For the Record

Allele Frequencies for Three STR Loci in a Population Sample from Catalonia (Spain) Using a Simple Manual Triplex PCR Method

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Population: Catalonia, West Mediterranean region from N.E. of Spain. $N = 230$ for D5S818, 228 for D13S317, 224 for D7S820

Keywords: forensic science, population genetics, short tandem repeats, triplex PCR amplification, DNA typing, D5S818, D13S317, D7S820, Caucasian, Catalonia, Spain

DNA was extracted from blood samples from healthy unrelated individuals from Catalonia using the phenol-chloroform-isoamyl alcohol method (1). PCR triplex amplification of D5S818, D13S317, and D7S820 was achieved using fluorescein labeled primers described in GenBank database accession numbers G08446, G09017, and G08616. Hot start PCR reactions were carried out in a 20 μL volume containing 5 ng of DNA template, 200 μM each dNTP, 12 pmol (D5S818 and D7S820) or 14 pmol (D13S317) of each primer, 50 mM KCl, 10 mM Tris-HCl (pH 8.3) at 2 mM $MgCl_2$ and 0.75 units of AmpliTaq Gold™ polymerase. Calculated sample temperature conditions were: 12 min at 95°C (hot start and denaturing step), and 35 cycles consisting of 1 min at 94°C, 45 s at 53°C and 1 min 20 s at 72°C. The last elongation step was 1 h at 60°C. Control DNA samples, previously typed using PE Biosystems genotyping kits and color sequencer devices, were used. Separation of the PCR products was carried out on polyacrylamide denaturing high-performance DNA sequencing gels (ReproGel™ High Resolution). The electrophoresis were carried out on the Automated Laser Fluorescent (ALF) DNA sequencer (Pharmacia) at 1500 V, 34 W, 60 mA and 40°C with laser power at 3 mW for 480 min. HWE was determined by calculating the exact test proposed by Guo and Thompson (2). The distribution of the genotypes at all three loci were in Hardy-Weinberg equilibrium.

The complete data are available to any interested researcher upon request or by accessing http://www.ub.es/spublica/database.html

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Allele	D5S818	D13S317	D7S820
7	0.002		0.018
8		0.156	0.141
9	0.039	0.072	0.123
10	0.070	0.055	0.308
11	0.398	0.296	0.201
12	0.330	0.250	0.170
13	0.161	0.132	0.033
14		0.039	0.007
P (exact test)	0.1280	0.1068	0.0877