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Population genetics of three STR markers (CYP19, D8S1132 and FGA) in North-East Italy

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Abstract

The genetic database of the population living in north-eastern Italy has been enlarged. Three new short tandem repeats (STR) loci (CYP19, D8S1132 and FGA) were analysed by PCR, polyacrylamide gel electrophoresis and silver staining. The data obtained from more than 100 unrelated subjects was analysed to establish allele frequency distributions and other forensic parameters.

Keywords: CYP19; D8S1132; FGA; Population genetics; PCR

1. Introduction

Short tandem repeats (STR) systems are highly polymorphic and sensitive markers for human identification and paternity testing [1–5].

The forensic application of polymorphic markers requires the availability of a database for the reference population. In the present work, we report the allele frequency distribution and other statistical parameters of three new STR loci (CYP19, D8S1132 and FGA) analysed in our laboratory.

2. Materials and methods

A population sample from healthy unrelated Caucasians living in north-eastern Italy was analysed at the three STR loci reported in Table 1.

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Table 1
Other statistical parameters

	CYP19	D8S1132	FGA
OH (observed heterozygosity)	0.657	0.902	0.890
EH (expected heterozigosity)	0.700	0.859	0.845
PM (probability of match)	0.126	0.047	0.050
DP (discrimination power)	0.974	0.953	0.950

DNA extraction was carried out using the standard phenol-chloroform method [6].

PCR amplification was performed in a final volume of 25 μl with 1.5 mM Mg^{2+} following standard procedures with minimal modifications. Published primer sequences

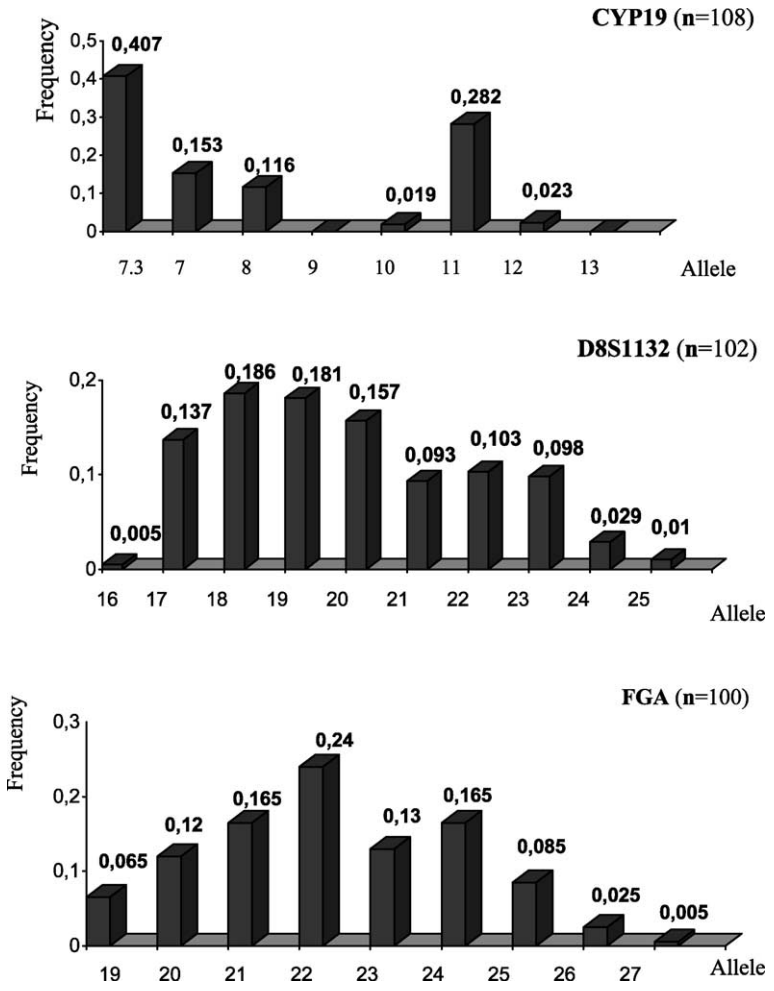

Fig. 1. Allele frequencies of the PCR-based systems analysed in our study; n = number of samples.

were employed [7–10]. A positive amplification control was represented by 1 ng of DNA from the K562 cell line.

After amplification, the PCR products of D8S1132 and CYP19 loci were analysed by non-denaturing 6% polyacrylamide/bis (19/1) gel electrophoresis in TBE buffer for 2–4 h at 400 V. To achieve an accurate separation of the inter-alleles, non-denaturing 8% polyacrylamide/bis (19/1) gel electrophoresis in TBE buffer for 15 h at 250 V was adopted for the locus FGA. Silver staining was performed for all three loci.

Genetic typing was achieved by comparison with sequenced allelic standards. The designation of alleles was given according to the number of repeats [11].

Statistical analysis included the estimation of allele frequencies, observed (OH) and expected (EH) heterozygosity, probability of match (PM) and discrimination power (DP). The Hardy–Weinberg equilibrium was verified using the χ^2-test.

3. Results

The observed allele frequencies of the three PCR-based systems are reported in Fig. 1.

No intermediate alleles were found for locus FGA. Other statistical parameters are summarised in Table 1. No departures from Hardy–Weinberg equilibrium were observed (data not shown) and our results do not differ from those of other open Caucasian populations as far as the degree of heterozygosity and the allele frequency distribution.

4. Discussion

In this study, we present genetic data concerning three STR systems usually tested in our laboratory. For all STR loci, the Hardy–Weinberg equilibrium was tested with the χ^2-test, and no significant deviation was found.

Moreover, the data reported here did not differ from those of other Caucasian populations concerning heterozygosity and the allele frequency distributions observed (data not shown).

With regard to the other forensic parameters (OH, EH, PM and DP), our data shows that the employment of a single STR system does not allow a high degree of discrimination between unrelated subjects. It is clear, however, that the combined analysis of many STR loci provides an irreplaceable tool both in personal identification and paternity testing.

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References

[1] A. Edwards, H.A. Hammond, L. Jin, C.T. Caskey, R. Chakraborty, Genetic variation at five trimeric and tetrameric tandem repeat loci in four human population groups, Genomics 12 (1992) 241–253.
[2] B. Brinkmann, The use of STRs in stain analysis, Proceedings from the third International Symposium on Human Identification, Promega, Madison, USA, 1992, pp. 357–374.
[3] C.P. Kimpton, A. Walton, P. Gill, A further tetranucleotide repeat polymorphism in the vWF gene, Hum. Mol. Genet. 1 (1992) 287.
[4] M.H. Polymeropoulos, H. Xiao, C.R. Merril, Tetranucleotide repeat polymorphism at the human myelin basic protein gene (MBP), Hum. Mol. Genet. 1 (1992) 658.
[5] P. Wiegand, B. Budowle, S. Rand, B. Brinkmann, Forensic validation of the STR systems SE33 and TC11, Int. J. Leg. Med. 105 (1993) 315–320.
[6] J. Sambrook, E.F. Fritsch, T. Maniatis, Molecular Cloning. A Laboratory Manual, Cold Spring Harbor Laboratory Press, New York, 1989.
[7] B. Caeiro, J.R. Luis, S. Dios, Genetic studies of the nucleotide repeat CYP19, in: B. Olaisen, B. Brinkmann, P.J. Lincoln (Eds.), Progress in Forensic Genetics, Singapore, Tokio, vol. 7, 1998, pp. 252–254.
[8] C. Weispfenning, C. Luckenbach, H. Ritter, CD4, CYP19 FABP2 and LPL: analysis, sequencing and frequency data, in: B. Olaisen, B. Brinkmann, P.J. Lincoln (Eds.), Progress in Forensic Genetics, Singapore, Tokio, vol. 7 (1998) pp. 378–380.
[9] P. Wiegand, H.R. Schneider, M. Schurenkamp, B. Brinkmann, Tetranucleotide STR system D8S1132: sequencing data and population genetic comparison, Int. J. Leg. Med. 111 (4) (1998) 180–182.
[10] N.J. Oldroyd, A.J. Urquhart, C.P. Kimpton, E.S. Millican, S.K. Watson, T. Downes, P. Gill, A higly discriminating octoplex short tandem repeat polymerase chain reaction system suitable for human individual identification, Electrophoresis 16 (1995) 334–337.
[11] Dna recommendations, Report concerning further recommendations of the DNA Commission of the ISFH regarding PCR-based polymorphisms in STR (short tandem repeat) systems, For. Sci. Int. 69 (1994) 103–104.