ANALYSIS OF THE LOCUS D1S80: FREQUENCY DISTRIBUTION IN NORTH-EAST ENGLAND AND APPLICATION TO PATERNITY TESTING

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Analysis of the VNTR locus D1S80 was first performed by Budowle et al. (1991) and the genetic and forensic applications were studied by Sajantile et al. (1992). Recently the reproducibility of the amplified fragment length polymorphism (AMP-FLP) technique to study the D1S80 locus has been tested by Thymann et al. (1993). However our knowledge about the variation of allele frequencies for the D1S80 locus is still very limited. The study describes the distribution of D1S80 alleles in the population of north-east England. A retrospective analysis of five paternity cases (trios), each excluded with three single locus VNTR systems detected by RFLP techniques, was also undertaken to assess the efficiency of the D1S80 locus in paternity testing.

Materials and Methods

High molecular weight DNA was extracted using phenol/chloroform procedure from 10ml EDTA blood samples, collected from 111 random individuals born and living in north-east England. Five paternity cases (trios) investigated previously, and each excluded with g3, MS1 and MS43 probes, were also investigated. DNA samples were air lifted to the Institute of Forensic Genetics, Copenhagen, where the characterisation of D1S80 phenotype was carried out.

Amplification of the D1S80 locus was performed using the following primers:

5'-AAA CTGG CCTC CAAA CACT GCCC GCCG-3'

5'-GTCT TGTT GGAG ATGC ACGT GCCC CTTGC-3'

The details of the PCR conditions, characterisation of the amplified product by polyacrylamide gel, visualisation by silver staining, and the allelic ladders used for assignment of the phenotype, are discussed in detail by Budowle et al. (1991) and Thymann et al. (1993).

Results and Discussion

Population Variation: The allele frequencies in the population of north-east England and other white Caucasian populations already studied are given in Table 1. In the population of north-east England a total of 18 alleles was observed for the D1S80 locus. The most frequent allele was with 24 repeats and its frequency was 32%, followed by the 28% frequency of the allele with 18 repeats. All other frequencies were less than 9% and alleles with 32 or more repeats all had very low frequencies

(0.05%). A single allele with more than 41 repeat units was also found in heterozygote combination with allele 23.

The allele frequencies of north-east England were very similar to the Caucasian populations of Finns, East Solvenians, Danes and US Caucasians (Table 1). However, there are significant differences compared to the Italian sample (Tagliabracci et al., 1992). In the Italians, the allele with 20 repeats was described as most frequent and both alleles 18 and 24 showed significantly low frequencies. Whether these differences are due to population structure or to the difference in technique needs further confirmation.

Heterozygosity and Power of Exclusion: The heterozygosity for the D1S80 locus in the population of north-east England is 75% and this value is similar to that in the Danes (77%) and other white Caucasian populations listed in Table 1. The theoretical chance of exclusion of non-father in the population of north-east England is 62%, a value once again very similar to that found for the Danish population (Thymann et al., 1993) and the match probability of the two randomly selected individuals is 6%.

Paternity Testing: The phenotypes of the five paternity cases where the exclusions were confirmed by three single locus probes are given in Table 2. Two cases (40%) were excluded by the D1S80 system.

It is evident from the results that the D1S80 locus is highly polymorphic and extremely useful in population and medical genetic studies. The discrimination power of D1S80 locus is 94%, suggesting its potential efficiency in forensic testing, however the proportion of cases independently excluded indicate that D1S80 can be used as an additional marker to confirm results.

References

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Table 1: Allele frequencies in 111 unrelated English from north-east England compared to other Caucasians

Allele (number of repeats)	Danes (a) n = 210	US-Caucasians (b) n = 99	Finns (c) n = 140	East Slovenian (d) n = 100	North-east England (e) n = 111
17	0.005	-	-	-	-
18	0.224	0.293	0.307	0.235	0.283
19	0.002	0.010	0.011	0.030	-
20	0.028	0.020	0.032	0.025	0.023
21	0.014	0.025	0.018	0.015	-
22	0.041	0.045	0.014	0.045	0.045
23	0.019	0.020	0.014	0.025	0.018
24	0.371	0.328	0.311	0.375	0.324
25	0.036	0.035	0.075	0.045	0.045
26	0.017	0.015	0.011	0.025	0.081
27	0.012	0.056	0.007	0.040	0.013
28	0.050	0.066	0.068	0.045	0.049
29	0.057	0.056	0.032	0.040	0.058
30	0.010	0.010	0.043	0.010	0.005
31	0.088	-	0.079	0.010	0.090
32	0.005	0.010	-	0.015	0.005
33	-	0.005	-	0.010	0.005
34	0.005	0.005	-	0.010	-
35	0.005	-	-	-	0.005
36	0.007	-	-	-	0.005
37	0.002	-	0.007	-	0.005
..	....	....	....	....	0.005
..	....	....	....	....	-
41	0.002	-	-	-	-
41	-	-	-	-	-
					0.005

(a) Thymann et al., 1993; (b) Budowle et al., 1981; (c) Sajantila et al., 1992; (d) Nasikov et al., 1992; (e) present study

Table 2: Phenotype of five paternity cases each excluded with three single locus VNTR probes (g3, MS1, MS43)

Relation	Case 1	Case 2	Case 3	Case 4	Case 5
Putative father	24, 31	24, 29	24	23, 36	18, 25
Child	22, 24	24, 29	24, 25	18, 22	18, 24
Mother	22, 24	18, 29	24	18, 25	18, 24
Conclusion	NE	NE	E	E	NE

NE not excluded; E excluded