PATERNITY TESTING WITH DNA SYSTEMS: APPLICATION OF D1S80 PHENOTYPING TO DANISH PATERNITY CASES ANALYSED WITH FIVE VNTR SINGLE LOCUS SYSTEMS.

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INTRODUCTION

This study presents the results of examination with D1S80 (MTC118) in 61 Danish cases of disputed paternity involving 74 men. The calculations were based on 372 unrelated Danes. The exclusion efficiency of the system D1S80 has been compared with the efficiency of the VNTR single locus systems D2S44 (YNH24), D5S43 (MS8), D7S21 (MS31), D7S22 (g3), and D12S11 (MS43a).

MATERIAL AND METHODS

The details of the technique used for D1S80 examinations has been described elsewhere (Thymann et al. 1993). Briefly the AMP-FLP technique used comprised DNA extraction from 3 ul blood by the chelex method followed by PCR amplification according to Budowle et al. (1990). Typing of the PCR products were performed by vertical electrophoresis in polyacrylamide gels and silverstaining. Paternity testing using the five VNTR-single locus systems has been published previously (Morling & Hansen 1993, Hansen & Morling 1993)

RESULTS AND DISCUSSION

Among 372 random, unrelated Danes 23 different D1S80 alleles were observed. The frequencies are given in table 1. Out of the 276 possible phenotypes 72 were observed. In 297 two D1S80 alleles, and in 75 persons only one allele were observed, giving a heterozygosity of 79.8%.
The investigations of 61 cases of disputed paternity with the system D1S80 and the five single locus systems used routinely in paternity investigations are summarized in table 2. Of the 74 men 18 were excluded. The comparatively high proportion of possible fathers (56/74) can be explained by the fact that the DNA-investigations were used as a second step for men not already excluded by the conventional genetic marker systems. All the 18 men excluded by D1S80 were also excluded by at least two of the other DNA systems used. Three men were excluded by the other DNA systems but not by the D1S80 system.
The positive evidence **for** paternity was examined by calculating the paternity index (PI) for 60 men not excluded by the D1S80 system. The PI-values were calculated as described by Gürtler (1956). A paternity index, PI <10, was found in 47 cases (78%), and in 10 cases 11 <PI< 50. In only 3 cases PI > 100.

For the D1S80 system RMNE-values (Random Man Not Excluded) were calculated in two ways: i) for the 62 mother-child pairs, RMNE = 0.380, ii) by means of random reallocation of the 74 men, RMNE = 0.297. The average value observed was RMNE = 0.339, or a rate of exclusion of 66,1%. Using the genefrequencies given in table 1, a theoretical rate of exclusion of 64.0% was calculated. For comparison the observed exclusion efficiency of the five VNTR systems used routinely in 70 cases were: D2S44 87.1%, D5S43 51.4%, D7S21 91.4%, D7S22 92.9%, and D12S11 78.6%. (Hansen & Morling 1993).
No mother-child exclusions were observed in the D1S80 system, and all paternal exclusions were confirmed by at least two other DNA systems. No evidence of the presence of a "blank" allele was found.

CONCLUSION

The system D1S80 (MCT118) has proven a reliable system for use in cases of disputed paternity. The amount of material required is small, the technique is not time-consuming, and together with the high exclusion efficiency this makes the system well fit for use in the first step of paternity investigations.

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Table 1. Observed distribution of 23 D1S80 alleles among 372 unrelated Danes.

Allele	Observed number	Phenotype frequency	Allele frequency
17	3	0.0081	0.0040
18	148	0.3978	0.2218
19	2	0.0054	0.0027
20	20	0.0538	0.0269
21	19	0.0511	0.0255
22	26	0.0699	0.0363
23	16	0.0430	0.0215
24	214	0.5753	0.3602
25	25	0.0672	0.0336
26	14	0.0376	0.0188
27	6	0.0161	0.0081
28	41	0.1102	0.0551
29	36	0.0968	0.0497
30	13	0.0349	0.0175
31	64	0.1720	0.0887
32	4	0.0108	0.0054
33	4	0.0108	0.0054
34	3	0.0081	0.0040
35	2	0.0054	0.0027
36	4	0.0108	0.0054
37	1	0.0027	0.0013
40	1	0.0027	0.0013
41	3	0.0081	0.0040

Table 2. Outcome of 61 cases of disputed paternity investigated with D1S80 and the five single locus systems D2S44, D5S43, D7S21, D7S22, and D12S11.

Number of men	Number of men/case				
	1	2	2 sibs	3	Total
Non-excluded	45	9	2	0	56
Excluded in D1S80 & other	2	9	1	3	15
Excluded in other systems	2	0	1	0	3
Total	49	18	4	3	74
Number of cases	49	9	2	1	61