Comparison of Minisatellite DNA Probes and Blood Group, Protein, and Enzyme Markers in Paternity Cases

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INTRODUCTION

Multilocus and single locus minitsatellite probes have been applied to the paternity expertise for some time (Jeffreys et al., 1985; Dykes et al., 1988; Henke et al., 1990; Jeffreys et al., 1991). They are now considered as very useful tools in complex cases of disputed paternity. There are, however, still controversal opinions concerning their application in the standard trio case. In the present study we have been interested in a comparision between the use of single locus minisatellite DNA probes (SLP) and conventional blood group, enzyme, and serological markers under routine conditions.

MATERIALS AND METHODS

In 50 paternity cases with three individuals and four cases with four individuals (two children or two men involved) the paternity expertise was carried out according to German federal regulations. In addition, the genetic markers C3, BF, PLG, ORM1, the subtypes of HP, GC 1, PGM1, TF C, BF F, and in some cases A2HS, PGP and C6 were investigated. Genomic DNA was digested with HinfI, submitted to eletrophoresis and Southern blotting, and hybridized with the DNA probes MS1, MS31, MS43, g3, and MS8 (ICI Diagnostics) as described elsewhere (Wong et al., 1987). For the detection of probe-specific fragments a non-radioactive digoxigenin-dUTP labeled flourescence AMPPD Kit (Boehringer Mannheim) was used, and initially compared with a radioactive ^{32}P assay.
The probability of paternity in the standard expertise as well as in the SLP expertise was calculated according to Essen-Möller (Essen-Möller, 1938). For the DNA expertises the likelihood values were calculated using upper 95 % confidence limits as conservative estimates for the allele frequencies derived from the non-related German adults of the total number of cases.

RESULTS AND DISCUSSION

In the technical evaluation the fluorescent assay was seen to be superior in time, sensitivity and reprobing as compared to the ^{32}P assay, but comparable in experimental effort to the conventional expertise without HLA.
The distribution of SLP fragments among the nonrelated individuals followed published frequencies for Caucasoids (Fig. 1). Remarkable, however, was the wider spectrum of fragments besides the major 6.6 kb fragement of MS8 and the lower frequency for the major 6.4 kb fragment of MS31, and also a lower frequency for the 1.7 kb fragment of g3. This may be related to the limited number of individuals tested here, and to the method of attributing fragment sizes. The rate of heterzygous individuals was in agreement with published frequencies: MS1 0.99, MS31 0.96, MS42 0.95, g3 0.97, and MS8 0.89.

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Concordance in exclusion of paternity between the SLP's and standard markers was seen in 17 cases (Fig. 2). In two cases with a single exclusion in the standard expertise all five SLP's were informative. In no case less than three SLP's showed exclusion of paternity. No exclusion was observed with minisatellites or with standard markers alone.
Thirty-six men not excluded by the two procedures had probabilities of paternity between 96.0 % and 99.99 % with conventional markers, between 98.77 and 99,99 % with SLP's (Table1).

Table 1. Comparison of the probability of paternity based on conventional markers and SLP's (no. of cases)

P	conventional markers	SLP's
> 0.9999	24	33
0.9973-0.9999	5	2
0.9900-0.9973	4	0
0.9600-0.9900	3	1

We conclude from our data that with the non-radioactive detection system minisattellites are reliabel for the paternity expertise. Although higher probability values were obtained with results from minisatellites there was no major improvement in the verbal statement of paternity for the non-excluded men. In view of the limited experience with DNA probes as compared conventional to markers and a similar technological effort, we consider that for the presence their application should be confined to complex cases such as involvement of related men, deficiency cases single exclusion in the standard expertise or unsatisfactory probability values.
For biostatistical evaluations more extensive data on SLP frequencies in different populations are needed. They were recently published for a larger West German population sample (Henke et al., 1991). But due to the absence of standardized procedures laboratory specific frequencies for single locus minisatellites may still some time be necessary.

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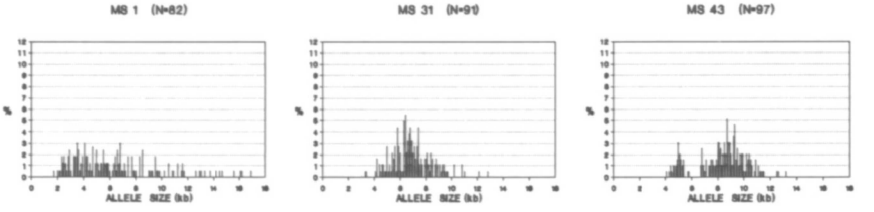
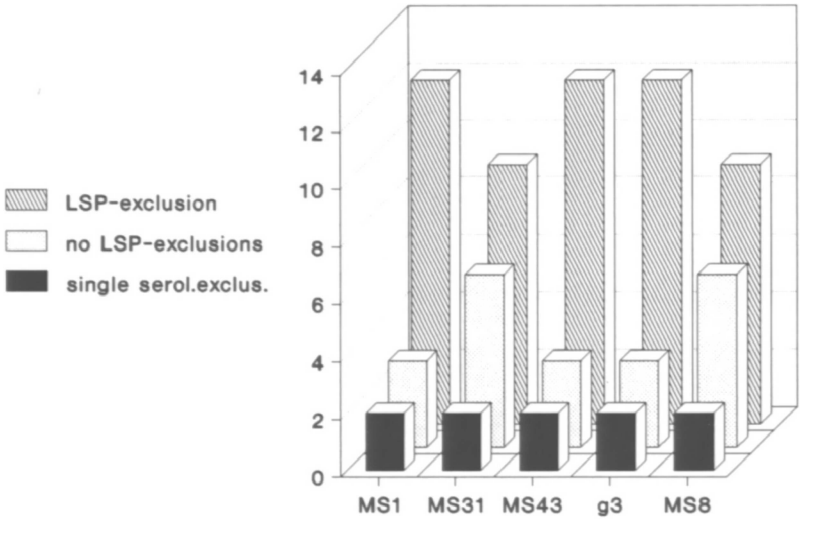


Figure 1: Allele Frequencies of SLP's
in Caucasoids (Cologne area)

Figure 2: Exclusion of Paternity:
5 LSP evaluated (N=17cases)



Exclusion with 1 to 7 serolog. markers