APPLICATION OF CONVENTIONAL AND DNA POLYMORPHISM TO PATERNITY CASES

Crestani C., Caenazzo L., Ponzano E., Bonan G., Cortivo P.

Institute of Legal Medicine, University of Padua, Via Falloppio 50, 35121 Padova, Italy

INTRODUCTION

Multilocus and single locus minisatellite probes have been applied to the paternity expertise for sometime.

DNA profiling has been routinely used in cases of disputed paternity at the Institute of Legal Medicine of Padua since 1991.

A series of 41 cases of paternity testing have been investigated using a single locus probe (pYNH24), up to 3 AMP-FLPs (ApoB, YNZ22, MCT118), HLA-DQalfa, and also 21 systems comprising red cell antigen, red cell enzyme and serum protein polymorphisms.

In the present study we have assesed and compared the use of both DNA testing and conventional markers in each of the above mentioned paternity cases.

MATERIALS AND METHODS

The material comprised 41 cases of disputed paternity.

Routine paternity testing consisted of the conventional analysis of the red cell antigen systems: ABO, MNSs, Rhesus, Kell-Cellano, Duffy, Kidd, Lutheran, the red cell enzymes AcP, PGM_1, GLO, EsD, ADA, AK, 6-PGD, the serum proteins Hp, Tf, Pi, Bf, ORM and PLG.

DNA was extracted from EDTA-blood using a non-organic solvent procedure. Genomic DNA was digested with HinfI, submitted to electrophoresis and Southern blotting, and hybridized with ^{32}P-labelled DNA probe, YNH24. The radioactive fragments were visualized by autoradiography.

PCR amplification of MCT118, ApoB and YNZ22 was performed using previously published conditions and primers (1-3). PCR products were separated by electrophoresis in 1x TBE Buffer using 6% polyacrylamide (29:1 Acrylamide/Bis) for ApoB, 8% for MCT118 and YNZ22. Electrophoresis was performed at 150 V in a C.B.S. apparatus for about 3-4 hours (depending on the genetic system). The marker used was Marker VI Boehringer. The amplification products were visualized directly after staining the gel in ethidium bromide.

HLA-DQ alfa were amplified by PCR using the Amplitype^{TM}DQalfa Kit (CETUS) according to the recommended protocol.

RESULTS

28 of the 41 cases gave no exclusions on conventional or DNA testing.

In 2 cases (see case 8 and 11) clear exclusions were found using only conventional systems; in 1 case (see case 4) DNA testing was useful for the exclusion of paternity.

In table 1 13 of 41 cases of disputed paternity are reported with exclusion.

Table 1 - Exclusion in 13 cases of disputed paternity

case 1 - exclusion: Duffy, PGM_1, DQalfa

case 2 - exclusion: Rh, Alfa-1-AT, Gc, ORM, DQalfa

case 3 - exclusion: Kidd, DQalfa, MCT118

case 4 - exclusion: DQalfa

case 5 - exclusion: AB0, Hp, MNSs, Kidd, Tf, Alfa-1-AT, C3, DQalfa, YNH24, MCT118

case 6 - exclusion: AB0, MNSs, Hp, Tf, DQalfa, YNH24, MCT118

case 7 - exclusion: MNSs, Kidd, PGM_1, Tf, Alfa-1-AT, DQalfa, YNZ22, ApoB

case 8 - exclusion: ADA, Alfa-1-AT, PGM_1

case 9 - exclusion: Rh, AcP, ORM, Tf, DQalfa

case 10 - exclusion: Rh, Alfa-1-AT, DQalfa, MCT118

case 11 - exclusion: MNSs, Kidd, ADA, PGM_1, Alfa-1-AT

case 12 - exclusion: Rh, Duffy, ApoB

case 13 - exclusion: AcP, YNZ22

The analysis by DNA polymorphisms appears to be quite valuable in the study of paternity cases.

REFERENCES

1) Budowle B., Chakraborty R., Giusti A.M., Eisenberg A.J., Allen R.C. (1991). Analysis by the PCR followed by high-resolution PAGE. Am. J. Hum. Genet. 48:137-144.

2) Boerwinke E., Xiong W., Fourest E., Chan L. (1989). Rapid typing of tandemly repeated hypervariable loci by the polymerase chain reactin: application to the Apolipoprotein B 3' hypervariable region. Proc. Natl. Acad. Sci. USA 86:212-216

3) Horn G.T., Richards B., Klinger K.W. (1989). Amplification of a highly polymorphic VNTR segment by the Polymerase Chain Reaction. Nucleic Acids Res. 17:2140.