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Investigation of chimerism in a healthy, adult female by means of minisatellite and microsatellite typing

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Abstract

DNA profiles were generated from blood and a tissue sample (buccal swab) of a healthy female showing irregular agglutinations in serological blood grouping and from whole blood samples of her parents in order to prove the presumption of chimerism and differentiate between its different forms. Genetic patterns of different cell types, including double maternal and paternal contribution of alleles at some loci, could be detected in whole blood and the buccal swab sample. Thus, the chimerism detected in the proposita was not restricted to the blood.

Keywords: Tetragametic chimerism; Microsatellite; Minisatellite

1. Introduction

Chimeric individuals are characterized by having cells with different genetic patterns originating from two or more zygotes. Within permanent chimeric individuals, ‘‘blood chimeras‘‘ (twin chimeras), which result from blood vessel junctions of dizygotic twins, can be distinguished from ‘‘whole body chimeras‘‘, where the coexistence of the different cell lines is not restricted to haemopoietic cells but spread over various tissues. Whole

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body chimerism (tetragametic or dispermic chimerism) is characterized by double parental or double paternal contribution of markers in all tissues [1]. In very rare cases, double maternal contribution has also been detected [2]. We investigated blood and a tissue sample of a healthy female, showing irregular agglutinations in serological blood grouping, in order to verify the chimeric status.

2. Materials and methods

EDTA whole blood samples were taken from the proposita and her parents. Additionally, a buccal swab was collected from the proposita. DNA extraction was either performed by Chelex® extraction [3], the "salting out method" [4] or Qiagen DNeasy Tissue kit®. STR typing was conducted employing the AmpF*l*STR® SGM Plus™ and Profiler Plus kits™ (Applied Biosystems) as well as the Powerplex™ 16 kit (Promega), the genRes HumACTBP2 (SE33)® system (SERAC) and an in-house method for D12S391 [5]. Subsequent minisatellite typing was conducted using non-commercially available methods for YNZ22, D1S80, Col2A1 and ApoB [6].

3. Results

STR typing with AmpF*l*STR® SGM Plus™ and Profiler Plus™ kits (13 different STR loci and Amelogenin) revealed the presence of two maternal and one paternal allele at seven loci (D3S1358, VWA, D21S11, D18S51, D19S433, TH01, FGA) in blood and buccal swab samples of the proposita. Amelogenin gave no indication for the presence of a Y-chromosome. In some loci, mixed patterns could not be observed due to shared alleles between the parents or homozygosity, whereas in one locus (D8S1179), no double maternal contribution was observed despite maternal heterozygosity. Further investigations including six more loci using Powerplex™ 16 (Penta E, CSF1PO, Penta D, TPOX), the genRES HumACTBP2 kit and D12S391 finally revealed the presence of two paternal alleles at the Penta E and the D12S391 locus (Table 1). Minisatellite typing also clearly confirmed that the chimerism of our proposita is a result of double parental contribution, showing both alleles of the father in typing of D1S80 and Col2A1 (Table 1).

Table 1
Chimerism in blood of a healthy, adult female—double paternal contributions at two microsatellite and two minisatellite loci

Individual	Microsatellite loci		Minisatellite loci	
	Penta E	D12S391	D1S80	Col2A1
Mother	16, 18	21, 21	24, 24	8, 8
Proposita	13, 14, 16, 18	18, 20, 21	18, 24, 37	5s, 8, 8f
Father	13, 14	18, 20	18, 37	5s, 8f

4. Discussion

Using the AmpF*l*STR® SGM Plus™ and Profiler Plus kits™, mixed genetic patterns could be obtained from blood samples and a buccal swab from the proposita. Initially, due to the lack of clear double paternal contribution, which most probably resulted from the high percentage of alleles shared between the parents as well as of homozygosities at several loci, the rare case of double maternal but single paternal contribution had to be assumed. Further investigations including six more microsatellite loci and four minisatellites finally revealed double paternal contributions at four loci, thus, the chimerism in the proposita was shown to be a result of double parental contribution.

References

[1] P. Tippett, Blood group chimeras, Vox Sang. 44 (1983) 333–359.
[2] C.R. Ford, Mosaics and chimeras, Br. Med. Bull. 25 (1) (1969) 104–109.
[3] P.S. Walsh, D.A. Metzger, R. Higuchi, Chelex® 100 as a medium for simple extraction of DNA for PCR-based typing from forensic material, BioTechniques 10 (4) (1991) 506–513.
[4] S.A. Miller, D.D. Dykes, H.F. Polesky, A simple salting out procedure for extracting DNA from human nucleated cells, Nucleic Acids Res. 16 (3) (1988) 121.
[5] B. Glock, E.M. Dauber, D.W.M. Schwartz, W.R. Mayr, Additional variability at the D12S391 STR locus in an Austrian population sample: sequencing data and allele distribution, Forensic Sci. Int. 90 (3) (1997) 197–203.
[6] D.W.M. Schwartz, E.M. Jungl, O.R. Krenek, W.R. Mayr, Simple and rapid typing for VNTR polymorphisms using high resolution electrophoresis on rehydratable polyacrylamide gels, Advances in Forensic Haemogenetics, vol. 5, Springer, Berlin, FRG, 1994, pp. 170–172.