

CASE REPORT

INHERITED ALLELES REVEALING AN INCESTUOUS PATERNITY

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Abstract: Some rape cases result in the pregnancy of the victim and if the case is not reported to the police after the act with a subsequent gynaecological examination of the girl and the taking of a vaginal swab, there is no way of connecting the rape case with the perpetrator, except by parentage determination using DNA (deoxyribonucleic acid) analysis after abortion or induced delivery. In order to solve the rape case of a minor girl of 14 years which resulted with pregnancy, where a 60-year-old man was accused of the rape, DNA was extracted from blood samples from the girl and the putative assailant and from the foetus after its induced delivery. The autosomal short tandem repeats (STR) typing for 15 different loci showed differences in 6 STR loci between the putative assailant as a father and the foetus, thus excluding the tested paternity. A large number of identical loci between the mother's and the child's genotype led us to consider the possibility of incestuous paternity. Analysis of DNA samples from the girl's father and brother clarified the case as brother-sister incest.

Key words: rape case, short tandem repeats (STRs) genotype, incest.

Introduction

The performance of forensic DNA analysis has considerably increased efficiency in solving paternity cases, sexual assaults or criminal cases. Regarding this improvement in forensic analysis, the short tandem repeat (STR) system, which is highly polymorphic, has an evidential value in clarifying these cases with a high degree of certainty [1, 2]. In cases of sexual assault, the victims are usually woman and children [3] or mentally retarded individuals [4], while the assailants are nearly always men.

Sometimes pointing to a particular sexual assailant can take away from the real case. Especially when the victim is a child, the situation can be more complicated. In comparison to adult victims there is need for special access when dealing with prepubertual victims because of their vulnerable nature and lower capacity to deal with trauma. Some of them refuse to cooperate and give true information about the problem. Sometimes work with specialists in children's psychology [5] can get us closer to the right solution. With the rapid advance in forensic DNA technologies, DNA analysis of biological samples offers a precise clue in solving sexual abuse cases.

Here we report a rape case which resulted in pregnancy, where the sexually abused girl accused a 60-year-old man as the assailant and as the father of the child she was carrying. In view of the medical and psychosocial implications, it was considered the pregnancy should be terminated. DNA extracted from blood samples of the girl and the assailant and from the foetus after its induced delivery was examined for 15 autosomal STR loci and Amelogenin locus. The results excluded the assailant as the father of the aborted foetus.

After the exclusion of the assailant as the father of the child (foetus), due to the large number of identical loci for both alleles between the mother's and the child's genotype, we considered that a close relative of the mother could be the father of the child. The other possibility accounting for such a high number of identical alleles between the mother's and the child's genotype could have been mating between individuals of a small, isolated population group. In this case the reason for sharing a large number of identical alleles between the foetus and the mother was due to the mating of individuals with a first degree of relationship.

The case was solved after making a DNA profile of girl's brother and father, clarifying that the brother of the girl was the father of her child (foetus).

Materials and Methods

DNA extraction

DNA was extracted from blood samples taken from the foetus, the girl, the assailant and the girl's brother and father, using the Chelex 100 method [6].

Polymerase chain reaction amplification

Polymerase chain reaction (PCR) amplification was performed according to the protocols and the manufacturers' instructions for AmpFISTR Identifier kit (Applied Biosystems). Amplification was carried out in a 9700 Thermal Cycler (Applied Biosystems).

Electrophoresis

After preparing a mix of 1 µl of the PCR product with 12 µl of formamide and 0,5 µl of GeneScan 500 LIZ size, standard samples were electrophoresed and analysed on an ABI Prism 310 Genetic Analyzer. Genotyping was done using Genotyper (Version 3.2, Applied Biosystems) Software.

Statistics

Statistical analysis was performed using DNA View Charles Brenner software for paternity testing.

Results and discussion

The results in this case were obtained in two parts. In the first approach, analysis of a set of 15 different autosomal loci from DNA extracted from the blood of the foetus, the girl (victim) and the assailant showed six exclusions for the assailant as the alleged father (Table 1). Thus the first conclusion was that the putative assailant of the girl was not the father of the child she was carrying.

Table 1 –Табела 1

STR profile of the foetus, victim (the girl) and the putative assailant
СТР профил на фетусот, жртивата (девојчето) и претпоставениот силувач

DNA locus	Foetus	Victim (the girl)	Putative assailant
D8S1179	13, 13	13, 13	12, 13
D21S11	29, 32.2	29, 32.2	27, 28
D7S820	10, 11	10, 11	8, 11
CSF1PO	10, 11	10, 11	10, 11
D3S1358	15, 16	15, 18	15, 16
TH01	9, 9	7, 9	9.3, 9.3
D13S317	8, 11	8, 10	11, 11
D16S539	10, 12	12, 12	12, 13
D2S1338	22, 25	22, 25	24, 25
D19S433	13, 15.2	13, 13	14, 14
vWA	17, 18	18, 18	16, 18
TPOX	8, 8	8, 8	8 , 11
D18S51	14, 21	15, 21	14 , 18
D5S818	13, 13	11, 13	12, 12
FGA	21, 22	21, 22	21 , 21
Amelogenin	X, X	X, X	X, Y

Exclusion of the putative assailant (at six loci) as the father of the child (foetus).

Претпоставениот силувач беше исклучен како татко на детето (фетус) во 6 локуси.

In the second approach, the observation of seven identical loci between the mother and the foetus with four homozygous loci in a foetal profile sug-

gested that pregnancy might have been a consequence of incest [7, 8]. The usual measure of inbreeding, often called the coefficient of inbreeding, gives the probability that alleles are identical by descent. From Wright's equation the coefficient of inbreeding of children of first degree relatives is $\frac{1}{4}$, which means that 25% of times a pair of alleles – for any gene being considered – is expected to be identical by descent [9]. Therefore the victim's first degree relatives, her father and her brother, were asked to provide blood samples for DNA testing.

Genetic analysis showed that the girl's father was not the father of the child, because of exclusion in three loci. The girl's brother's genotype, on the contrary, included at each locus an obligate paternal allele of the foetus (Table 2). The probability of paternity was calculated and gave a probability of paternity > 99.999984% (prior probability of paternity 50%).

Table 2 – Табела 2

STR profiles of the foetus, victim (the girl) and victim's brother and father
СТР профили на фетусот, жртвата (девојчето)
и браќот и таткото на жртвата

DNA locus	Foetus	Victim (the girl)	Victim's brother	Victim's father
D8S1179	13, 13	13, 13	13	13
D21S11	29, 32.2	29, 32.2	29, 32.2	32.2, 33.2
D7S820	10, 11	10, 11	10, 11	11
CSF1PO	10, 11	10, 11	10, 11	11
D3S1358	15, 16	15, 18	15, 16	16 , 18
TH01	9, 9	7, 9	7, 9	7, 9.3
D13S317	8, 11	8, 10	8, 11	8, 10
D16S539	10, 12	12, 12	10 , 11	10 , 12
D2S1338	22, 25	22, 25	25	22, 25
D19S433	13, 15.2	13, 13	13, 15.2	13, 15.2
vWA	17, 18	18, 18	17 , 18	18
TPOX	8, 8	8, 8	8	8 , 11
D18S51	14, 21	15, 21	14 , 15	14 , 21
D5S818	13, 13	11, 13	11, 13	13 , 14
FGA	21, 22	21, 22	22	22 , 24
Amelogenin	X, X	X, X	X, Y	X, Y

In the second approach, results showed exclusion of girl's father (at three loci) and confirmation of girl's brother as the father of the unborn child.

Во втората анализа, резултатите покажаа исклучување на таткото на девојчето (во три локуси) и потврда дека братот на девојчето е татко на нероденото дете.

Although the results of population studies show that most of the alleles from analysed loci such as al.8 (TOPX); al.22 (FGA); al.13 (D8S1179); al.29 (D21S11) and al.11 (D7S820) are among the most frequent in the Macedonian population [10], in this case their repetition was due to an incestuous mating.

Conclusion

The presented case shows the importance of complex assessment in solving forensic cases, particularly when it is a matter of cases of rape. It points to the importance of focused attention on obtained DNA results, with their correct interpretation, which can sometimes direct us towards further investigation in order to solve the case in its completeness.

REFERENCES

1. Brinkmann B., Pfeiffer H., Schurenkamp M., Hohoff C. (2001): The evidential value of STR. An analysis of exclusion cases. *Int J Legal Med*; 114: 173–177.
2. Delfin F., Madrid B., Tan M., De Ungria M.C. (2005): Y-STR analysis for detection and objective confirmation of child sexual abuse. *Int J Legal Med*; 119: 158–1632.
3. Collins KA. (1998): The laboratory's role in detecting sexual assault. *Lab Med*; 29: 361–365.
4. Robino C., Barilaro M., Gino S., Chiarle R., Palestro G., Torre C. (2006): Incestuous paternity detected by STR-typing of chorionic villi isolated from archival formalin-fixed paraffin-embedded abortion material using laser microdissection. *J Forensic Sci*; 51: 90–924.
5. Laraque D., DeMattia A., Low C. (2006): Forensic Child Abuse Evaluation: A Review. *The Mount Sinai Journal of Medicine*; 73: 1138–1147.
6. Walsh PS., Metzger DA., Higuchi R. (1991): Chelex 100 as a medium for simple extraction of DNA for PCR-based typing from forensic material. *Biotechniques*; 10: 506.
7. Schmidtke J., Kuhnau W., Wand D., Edelmann J., Szibor R., Krawczak M. (2004): Prenatal exclusion without involving the putative fathers of an incestuous father-daughter parenthood.
8. Macan M., Uvodic P., Botica V. (2003): Paternity testing in a case of brother-sister incest. *Croatian Medical Journal*; 44(3): 347–349.
9. Mange EJ., Mange A. (1999): Basic human genetics, Second edition published by Sunderland, Massachusetts.
10. Jakovski Z., Nikolova K., Furac I., Masic M., Janeska B., Kubat M. (2006): Allele frequencies for 15 STR loci in a population from the republic of Macedonia. *Int J Legal Med*; 120: 53–55.

Резиме

**ИНЦЕСТНО ТАТКОВСТВО ОТКРИЕНО ПРЕКУ НАСЛЕДЕНИ
АЛЕЛИ**

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Апстракт: Некои случаи на силување резултираат со забременување на жртвата и доколку случајот не биде пријавен во полиција по настанот со понатамошен гинеколошки преглед и земање на вагинален брис, силувањето не може да биде поврзано со извршителот, освен со одредување на татковство со ДНК (дезоксирибонуклеинска киселина) анализа по абортус или провоцирано пораѓање. За решавање на случај на силување на 14-годишно девојче кој резултирал со бременост, каде што обвинет за силувањето беше 60-годишен маж, беше направена ДНК изолација од примерок крв од девојчето, претпоставениот силувач и од фетусот по провоцираното пораѓање. При анализа на кратки повторувачки ДНК секвенци (СТР) за 15 различни локуси, во 6 локуси имаше несовпаѓање меѓу ДНК профилот на наводниот силувач и фетусот со што се исклучи како можен татко на фетусот. Но големиот број на идентични локуси меѓу мајката и фетусот не наведе да мислиме на можноста за инцестно татковство. Анализата на ДНК примероците од таткото и братот на девојчето го разјасни случајот за инцест брат–сестра.

Клучни зборови: силување, кратки повторувачки секвенци, генотип, инцест.

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