X-Chromosome Markers Used in Deficiency of Paternity Case

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Abstract

Cases of deficiency paternity, characterized by missing of the alleged father, are a task for forensic genetics. Recently these cases were determined using sets of extremely polymorphic autosomal short tandem repeats (STRs), which indicate to powerful likelihood ratios (LR). Some difficult cases emerge whenever the kinship highly remote or if the another hypotheses are not properly formulated because of the absence of information. In these situations, beyond the routinely used marker set, laboratories usually enlarge the number and/or the type of markers analysed. A set of 14X-STRs is further advantageous than 21 autosomal STRs (AS STRs) biallelic markers. Furthermore, the utility of X-STRs was also appear in cases shows that in father–daughter duos and merely a close family member of the alleged parent (father or mother) is existing for testing. Here we present a cases with biological daughter of the alleged father and her mothers and putative grandmother was the genotyped relative are available for reconstructing his haplotype . Aiming to increase the Paternity Index (PI), Probability of Paternity (PP), and obtain more reliable results. The result of X STR showed that the missing person (father) share 50% genotype with the donors (daughter)and (his mother) at each locus, there are no alleles in any one of these 13 loci conflict with the alleles inherited from her father and Posterior Probability of paternity was (99.999958%) that is mean the missing person was biological father for this donor daughter.

Keywords: *13 X-STRs with Amelogenin, Paternity case, Kinship analysis, Paternity index, Paternity deficiency.*

Introduction

The main benefit of X-chromosomal (ChrX) STRs rises in deficiency paternity cases, i.e. when a putative father is unavailable and DNA from paternal relatives has to be investigated instead [1,2]. In such cases, the power of exclusion of autosomal STRs is significantly decreased, whereas ChrX markers are (at least in some cases) more resourceful. Female individuals share their paternal ChrX when they are fathered by the same man [3,4]. Males receive their single ChrX from their mother. Consequently, in cases in which the supposed grandmother is available for genotyping, the possible ChrX alleles of the putative father can be verified. ChrX marker typing is very applicable in mother–son kinship and in father–daughter analyzing [5,6].

Case Report

We perform the use of this set of markers in paternity deficiency cases, containing a female child and her mother and the putative grandmother and a dead alleged father who was not analyzed.

Figure 1 displays the pedigrees of the families. All persons are shown in the pedigrees were examined for autosomal (AS) and X STRs.

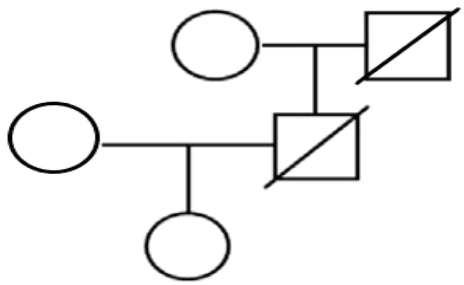

Figure 1: Pedigree of the deficiency paternity case. (paternal grandmother-granddaughter)

Materials and Methods

All blood samples were gathered utilizing finger lancets and kept on FTA® Classic Card (GE Healthcare, Pittsburgh, USA). For each sample a 1.2 mm punch was directly amplified according to the manufacturer's recommended instructions. PCR using two kits the PowerPlex 21® System)20 STR loci and Amelogenin), including D1S1656, D2S1338, D3S1358, D5S818, D6S1043, D7S820, D8S1179, D12S391,D13S317, D16S539, D18S51, D19S433, D21S11, Amelogenin, CSF1PO, FGA, Penta D, Penta E, TH01, TPOX and vWA. And 14ChrX STRs- GenePhile X-Plex(13STR loci and Amelogenin), including for (DXS8378, DXS9898, DSX8377, DSX8377, GATA172D05, DXS7423, DXS6809, DXS7132, DXS101, DXS6789, DXS9902, DXS9902, DXS7424). Genomic DNA (1 ng) was amplified in a 10 L total volume of PCR reaction mix was done using Applied Biosystems 9700 thermocycler according to standard manufacturer's recommended conditions. Capillary electrophoresis was achieved by using a 3500XL Genetic Analyzer (Applied Biosystems) in accordance with the manufacturer's instructions; ILS-500 (promega, 2017) and BTO (qiagen, 2015) were used as an internal size standard. Genotyping was completed using GeneMapper ID-X v1.4 software (Applied Biosystems).

Statistical Analysis

The statistical parameters that analyzed showed the Paternity Index (PI) and Probability of Paternity (PP), with the assumption of an a priori probability of 0.5 for the latter. Genotypes and allele frequencies of the X-liked STRs were transmitted by the deceased father to his daughter and the formulae applied when the putative paternal grandmother was the genotyped relative that can be seen in Table 1. Statistical analyses were done corresponding to the formulae proposed by Ayres and Powley in 2005 [7,8].

Table 1: Formulations used to determine PI considering the profile configurations depicted with ChrX STRs,

Mother	Daughter	Putative grandmother	Formulas
AB	AB	AB	1/(a+b)
AB	AA	AB	1/2a
AA	AB	AB	1/2b
AA	AA	AB	1/2a
AB	BC	AC or CD	1/2c
AC	CC	BC	1/2c
AC	AC	BC	1/[2(b+c)]
AA	AB	BB	1/b
Absent	AB	BC	1/4b
Absent	AB	BB	1/2b
Absent	BB	BC	1/2b

Results

The routine work in identification we started with autosomal using PowerPlex 21, Autosomal STR DNA profile of paternal grandmother, mother and female child trio is presented in Table 2. All the allele in the autosomal STR DNA profile of female child is matching with both mother and paternal grandmother on all the 20locus establishing the identity of the missing father.

Table 2: The autosomal STR DNA profile of both mother)wife) and Daughter, paternal grandmother. (powerplex 21 genotype)

STRs	Mother	Daughter	grandmother
Amelo	X,X	X,X	X,X
D3S1358	16,18	15,16	16,17
D1S1656	14,14	11,14	13,14
D6S1043	11.18	14,18	14,17
D13S317	11,11	9,11	8,14
Penta E	12,13	7,13	11,16
D16S539	10,10	10,13	12,12
D18S51	12,12	12,15	14,18
D2S1338	17,25	17,25	17,27
CSF 1PO	10,12	12,12	10,12
Penta D	9,15	9,9	11,11
TH01	9,10	9,9	7,9
vWA	16,17	16,17	15,17
D21S11	29,29	26,29	26,31.2
D7S820	10,10	10,12	8,12
D5S818	10,12	10,12	12,12
TPOX	8,10	8,8	11,11
D8S1179	9,15	9,14	9,14
D12S391	20,12	21,23	18,23
D19S433	13,15.2	13,15.2	14,15.2
FGA	24.25	24,24	24,24

Data of DNA profile of same three samples with - GenePhile X-Plex(13STR loci and Amelogenin), kit is presented in Table 3. In X STR also all the allele of female child is matching with both mother and paternal grandmother on all the 13 locus. As the female child is having two X, one from father and the other from mother, so this can be shown by generating X STR DNA profile of the trio.

Table (3) : The genotype of mother (wife), daughter and grandmother (genephile x-plex -13 STR kit)

STRs	mother	Daughter	grandmother
Amelo	X,X	X,X	X,X
DXS8378	11,12	10,11	10,11
DXS9898	8.3,8.3	8.3,14	8.3,14
DXS8377	46,54	47,54	43,47
HPRTB	11,13	11,13	11,12
GATA172D05	6,11	6,10	10,10
DXS7423	15,16	15,16	15,17
DXS6809	31,32	31,34	33,34
DXS7132	13,14	13,14	13,14

DXS101	21,21	21,24	14,24
DXS6789	20,20	19,20	15,19
DXS9902	10,12	10,12	10,10
DXS6807	11,14	13,14	11,13
DXS7424	14,16	12,14	12,13

So Paternity indices (PI) for full trio and deficient (father not available) cases are (23725241.4)

Prior odd=1/2

$$\text{Posterior Probability of paternity} = \frac{CPI * prior}{(CPI * priorodd) + (1 - priorodd)} = 0.99999958$$

Discussion

The autosomal matching was done to support these results and avoid the false positive due to huge number of close related people missing at the same time we did sex chromosomes genotyping. The result of X STR showed that the missing person (father) share 50% genotype with the donors (daughter)and (his mother) at each locus, there are no alleles in any one of these 13 loci conflict with the alleles inherited from her father and Posterior Probability of paternity was (99.999958%) that is mean the missing person was biological father for this donor daughter. This value is accepted according to lab. This value is accepted according to lab. of mass grave standard and the International Society for Forensic Genetics (ISFG) recommendation [9,10].

Conclusion

Recently the X-Chromosome STR loci transmission analysis are clearly available for use in forensics as in immigration, victim identification in mass disasters and it is possible that these loci will be used efficiently for paternity testing of female children, predominantly as supplementary tests to complement the information obtained with the autosomes and the differentiation between pedigrees in deficient or otherwise difficult cases. In this study the equations that used provide the means to evaluate the utility of such loci in the existence of substructure, and complement previously published equations used for autosomal loci.

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Ethical Clearance: The principles and the experimental protocol in this study was approved by the Medico-Legal Directorate, Ministry of Health, Baghdad, Iraq.

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Conflict of Interest: Nil.

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