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Allele frequency distribution of two X-chromosomal STR loci in the Han population in China

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Abstract. X-chromosome (ChrX) STR loci have been proven to be a useful tool for paternity testing, especially in some cases. In paternity testing, genetic markers on ChrX are only applicable when the disputed child is female. The author investigated allele frequency distribution of ChrX STR loci DXS6789 and HumSTRX1 in a sample of unrelated individuals (males and females) in Han population living in Chengdu, China, by using PCR and PAGE followed by silver staining. Ten and six different alleles of DXS6789 and HumSTRX1 loci were detected, respectively. The genotype frequencies of DXS6789 and HumSTRX1 loci were in good agreement with the Hardy–Weinberg equilibrium. The observed heterozygosity of DXS6789 and HumSTRX1 loci were 0.795 and 0.748 in females, respectively. It is suggested that these ChrX markers are appropriate for the purposes of forensic analyses.

Keywords: X-chromosome; DXS6789; HumSTRX1; Short tandem repeat; Polymorphism

1. Introduction

DNA typing of X-chromosome (ChrX) STR is underrepresented in paternity testing, and is only applicable in paternity cases when the disputed child is female. The mean exclusion (MEC) of genetic markers on X-chromosome as proposed by Kishida and Tamaki [1] were higher than those on autosomal chromosome as proposed by Krüger et al. [2]. Therefore, they are very useful in some special cases of deficiency paternity testing cases. Such as, in case of whether presumptive half-sisters have the same father when the mother is lacking. For the purpose of solving this problem, only a few ChrX markers are required. To date, the number of ChrX which have been studied in the Chinese population is still very small. The aim of this paper is to investigate the frequency distribution of both DXS6789 and HumSTRX1 in the Han population in Chengdu, China.

2. Survey methodology

Blood Specimens were obtained from 127 unrelated females and 118 unrelated males volunteer donors. DNA were extracted from blood specimens using Chelex-100 [3].

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Genotyping was carried out by PCR in a PE9600 cycler. The components of a 20 μl reaction mixture were as follows: template DNA 20 ng, primer 0.2 μmol/l each, dNTPs 200 μmol/l each, KCl 50 μmol/l, Tris–HCl (pH8.3) 10 mmol/l, $MgCl_2$ 1.5 mmol/l, Taq polymerase 1 U.

Primer sequences:

DXS6789:

P1:5′-TGT CCT ATT GTA TTA GTC AGG GAT C-3′;

P2:5′-ATG TAA GTT GGT ACT TAA TAA ACC CTC-3′.

HumSTRX1:

P1:5′-GTT TCC TCC TGC AAA ATA CAG C-3′;

P2:5′-TCC AGC ACC CAA GGA AGT C-3′.

PCR conditions: start at 94 °C for 3 min, followed by 30 cycles consisting of 35 s at 94 °C, 40 s at 57 °C, 50 s at 72 °C, followed by a 10 min extension at 72 °C. The amplified products were electrophoresed in 6% polyacrylamide gel by using 100 bp ladder and allelic markers as size markers, followed by silver staining. The amplified products were examined by using an ABI PRISM™ 310 Genetic Analyzer.

The Hardy–Weinberg equilibrium test (HWE) was performed by an exact test [4]. We calculated the polymorphism information content (PIC), average power of discrimination in females (PD^F) and in males (PD^M) [5].

3. Results

Both DXS6789 and HumSTRX1 loci are tetranucleotide [6–8]. DXS6789 exhibited 10 clearly distinguishable alleles ranging from 146 to 182 bp. HumSTRX1 exhibited six clearly distinguishable alleles ranging from 214 to 234 bp. The allele frequency distributions of both ChrX loci, their PD and PIC are shown in Table 1. The parameters of Het, and PIC, in normal family trio testing, as well as the average PD^F and PD^M confirm that both DXS6789 and HumSTRX1 loci are highly polymorphic.

4. Discussion

The genotype frequencies of both loci were shown in Table 2. Genotypes (25 and 12) were found in DXS6789 and HumSTRX1 loci, respectively, and they are in good

Table 1
Allele frequency distributions of DXS6789 and HumSTRX1

DXS6789						HumSTRX1					
Allele	Female number	Female (%)	Male number	Male (%)	Total frequency (%)	Allele	Female number	Female (%)	Male number	Male (%)	Total frequency (%)
14	3	1.2	0	0	0.6	11	0	0	2	0.8	0.4
15	53	20.9	40	16.9	19.0	12	7	2.8	8	3.4	3.1
16	77	30.3	74	31.4	30.8	13	41	16.1	40	16.9	16.5
17	8	3.1	14	5.9	4.5	14	117	46.1	98	41.5	43.9
18	1	0.4	0	0	0.2	15	75	29.5	76	32.2	30.8
19	14	505	2	0.8	3.3	16	14	5.5	12	5.1	5.3
20	51	20.1	46	19.5	19.8		–	–	–	–	–
21	37	14.6	44	18.6	16.5		–	–	–	–	–
22	8	3.1	16	6.8	4.9		–	–	–	–	–
23	2	0.8	0	0	0.4		–	–	–	–	–
Total	127	1	118	1	1		127	1	118	1	1
PD^F: 0.931; PD^M: 0.792; PIC: 0.77						PD^F: 0.837; PD^M: 0.691; PIC: 0.63					

Table 2
Genotypes of DXS6789 and HumSTRX1 found in females

DXS6789				HumSTRX1			
Genotypes	No.	Genotypes	No.	Genotypes	No.	Genotypes	No.
14–16	3	16–20	13	12–13	1	15–16	5
15–15	4	16–21	12	12–14	2	–	–
15–16	11	16–22	4	12–15	4	–	–
15–17	2	16–23	2	13–13	6	–	–
15–18	1	17–20	4	13–14	12	–	–
15–19	4	17–21	2	13–15	15	–	–
15–20	11	19–20	5	13–16	1	–	–
15–21	12	19–21	1	14–14	24	–	–
15–22	4	20–20	6	14–15	47	–	–
16–16	14	20–21	6	14–16	8	–	–
16–19	4	21–21	2	15–15	2	–	–
HWE exact test: $P=0.309>0.05$; Het: 0.795				HWE exact test: $P=0.174>0.05$; Het: 0.748			

agreement with the HWE, and *P*-values were 0.309 and 0.174, respectively. Investigation in 50 true family trios with female children suggested a codominant X-linked inheritance. No mutations and no mother–child exclusions were found. It is suggested that these ChrX markers are useful for forensic analyses, especially in solving complicated kinship testing and paternity testing of lacking mother.

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