International Congress Series 1239 (2003) 187–191

Polymorphisms of six STR loci on chromosome 22 in a Chinese population

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Abstract

To develop more STR markers for forensic DNA typing in Chinese populations, six STR loci on chromosome 22 were investigated. An Amp-FLP technique was employed for genotyping individuals from a Han population sample from North and South China. Hardy–Weinberg equilibrium was checked using the chi-square test and no significant deviation was found at any of the six loci. The results showed that the differences of genotype distributions at two loci were significant between two populations, but the distributions of genotypes at the other four loci were similar in both populations. Five of the six loci showed good levels of polymorphism in both populations, with heterozygosities of more than 0.59. The discrimination power and the exclusion probability were more than 0.82 and 0.40, respectively, making them suitable candidate markers for forensic applications. At another locus, only two alleles were found. The heterozygosity of this locus was less than 0.36 and the discrimination power and exclusion probability were less than 0.62 and 0.12, respectively. A two-step mutation was observed at one of six loci.

Keywords: Chromosome 22; STR; Polymorphisms; Chinese population

1. Introduction

STR loci have become routine markers for forensic applications. To develop more STR markers for forensic DNA typing in Chinese populations, we chose six STR loci on chromosome 22, named as D22S686, D22S533, D22S685, D22S683, D22S445 and D22S444. All six loci were tetranucleotide repeat STRs. We investigated the distributions

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0531-5131/03
PII: S0531-5131(02)00322-9

of genotypes in Han population samples from North and South China, and evaluated the forensic application of the six loci.

2. Materials and methods

2.1. Population samples

Forty-four and 63 EDTA–blood samples were collected from unrelated individuals in North and South China, respectively.

2.2. Experimental details

DNA was extracted using the Chelex method. PCR amplification was carried out using the primers according to GDB [1]. Each PCR reaction contained 2–10 ng human genomic DNA, 1 × Taq buffer, 1.5 mM $MgCl_2$, 200 μM each nucleotide, 1.5 U Taq polymerase (Promega, USA), 0.25 μM each primer in a total volume of 37 μl. A total of 30 cycles was carried out in a GeneAmp PCR System 9600 (Perkin-Elmer, USA) with denaturation for 50 s at 94 °C, annealing for 50 s at 59 °C and extension for 30 s at 72 °C. The PCR products were analyzed using non-denaturing polyacrylamide gel electrophoresis with a discontinuous buffer system [2]. The allelic ladders for STR typing were made in-house, constructed by mixing PCR products with different genotypes. Alleles of D22S683 were named in order of DNA fragments from short to long fragment: as 1, 2, 3, and so on, because the alleles of D22S683 were not sequenced. The alleles of the other five loci were named according to the recommendations of the International Society of Forensic Genetics [3].

2.3. Statistical calculations

The POWERSTATS software was used for the calculation of heterozygosity, allele frequencies, the discrimination power and the exclusion probability [4]. The chi-square test for Hardy–Weinberg equilibrium was carried out according to Hou et al [5]. The chi-square test was also used to compare the distributions of genotypes of the two populations.

3. Results and discussion

3.1. Population genetics

Using the chi-square test to compare the distributions of genotypes of North and South Chinese showed the significant differences between the two populations for the genotypes of the D22S683 and D22S444 loci. Also, polymorphism information content (PIC) at D22S444 was different between the two populations. The distributions of genotypes at D22S686, D22S533, D22S685 and D22S445 loci were similar in both populations, indicating that values of population genetics from South and North Chinese population samples could be combined in forensic applications (Tables 1 and 2).

Table 1
Allelic frequencies of D22S686, D22S533 and D22S685 in Chinese population

D22S686				D22S533				D22S685			
Allele	South	North	South and North	Allele	South	North	South and North	Allele	South	North	South and North
14	0.409	0.302	0.346	13	0.011	0.032	0.023	9	0	0.008	0.005
15	0.125	0.048	0.079	14	0	0.024	0.014	10	0.023	0.063	0.047
17	0	0.016	0.009	15	0.159	0.032	0.084	11	0.296	0.293	0.294
18	0.114	0.198	0.146	16	0.33	0.19	0.284	12	0.261	0.262	0.261
19	0.272	0.341	0.313	17	0.239	0.357	0.308	13	0.182	0.152	0.163
20	0.08	0.095	0.089	18	0.17	0.198	0.187	14	0.159	0.119	0.136
				19	0.091	0.167	0.136	15	0.057	0.087	0.075
								16	0.011	0.016	0.014
								17	0.011	0	0.005
HWE[a]	12.72	6.73	11.08		3.85	7.33	14.46		5.56	6.71	9.71
df	7	10	12		10	9	13		8	13	13
P	>0.05	>0.05	>0.05		>0.05	>0.05	>0.05		>0.05	>0.05	>0.05

[a] Test for Hardy–Weinberg equilibrium.

3.2. Forensic application

This study revealed that two alleles were found in the D22S444 locus, discrimination powers of D22S444 were 0.444 and 0.621 in South and North Chinese, respectively. The exclusion probabilities of D22S444 were 0.078 and 0.120 in South and North Chinese, respectively. It showed that the information content of D22S444 was limited, and D22S444 was not a good marker for forensic applications in these two populations. D22S686, D22S533, D22S685, D22S683 and D22S445 showed good polymorphisms in both populations. Their heterozygosities were more than 0.59. The discrimination power

Table 2
Allelic frequencies of D22S683, D22S445 and D22S44 in Chinese population

D22S683						D22S445				D22S444		
Allele	South	North	Allele	South	North	Allele	South	North	South and North	Allele	South	North
1	0	0.008	8	0.08	0.032	10	0.023	0	0.009	6	0.364	0.167
2	0.171	0.206	9	0.068	0.008	11	0.261	0.198	0.225	7	0.636	0.833
3	0.091	0.214	10	0.057	0.032	12	0.102	0.063	0.079			
4	0.045	0.087	11	0.011	0.04	13	0.182	0.183	0.182			
5	0.08	0.063	12	0.011	0.016	14	0.409	0.532	0.482			
6	0.216	0.135	13	0.034	0.016	15	0.23	0.024	0.023			
7	0.136	0.135	14	0	0.008							
HWE[a]				12.14	6.25		5.72	6.63	4.71		0.87	2.52
df				20	19		5	6	8		1	1
P				>0.05	>0.05		>0.05	>0.05	>0.05		>0.05	>0.05

[a] Test for Hardy–Weinberg equilibrium.

Table 3
Values of population genetics and forensic science of six STR loci

Locus	Population	PIC	DP	Pm	CE	OH	EH	SE
D22686	North	0.70	0.860	0.140	0.587	0.794	0.748	0.055
	South	0.68	0.860	0.140	0.435	0.705	0.731	0.067
	South and North	0.70	0.875	0.125	0.522	0.757	0.745	0.042
D22S533	North	0.73	0.889	0.111	0.647	0.825	0.733	0.053
	South	0.74	0.909	0.092	0.401	0.682	0.78	0.062
	South and North	0.75	0.913	0.087	0.538	0.766	0.786	0.040
D22S685	North	0.77	0.923	0.077	0.617	0.810	0.802	0.050
	South	0.75	0.909	0.091	0.633	0.818	0.818	0.061
	South and North	0.76	0.923	0.077	0.624	0.813	0.795	0.039
D22683	North	0.84	0.957	0.043	0.617	0.810	0.866	0.043
	South	0.86	0.960	0.040	0.472	0.727	0.884	0.048
D22445	North	0.59	0.822	0.178	0.402	0.683	0.645	0.060
	South	0.68	0.850	0.150	0.633	0.818	0.728	0.067
	South and North	0.63	0.845	0.155	0.490	0.738	0.682	0.045
D22S444	North	0.24	0.444	0.556	0.078	0.333	0.280	0.057
	South	0.36	0.621	0.379	0.120	0.409	0.468	0.075

PIC: polymorphism information content, DP: discrimination power, Pm: probability of match, CE: chance of exclusion, OH: observed heterozygosity, EH: expected heterozygosity, SE: standard error.

and the exclusion probability were more than 0.82 and 0.40, respectively. This showed that these loci would make suitable candidate markers for forensic applications. Forensic values of all six loci are shown in Table 3.

3.3. Mutation

A mutation event was observed at D22S685 transmitted from father to son in a pedigree. An allele 15 of the first offspring was inconsistent with an allele 13 in the father. Many researchers thought that mutations of repeats in STR loci were stepwise mutation, a mutation event increases or decreases a repeat [6], but this mutation event increased more than two repeats, a rare mutation event. The mechanism and biological significance of mutations in STR loci is unclear at this moment.

Acknowledgements

This study was supported by grants from the Alexander von Humboldt Foundation, Germany, and the Chinese Medical Board of New York, USA, and the National Nature Science Foundation as well as the Science Foundation of Sichuan Province, China.

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