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ORIGINAL ARTICLE

Seventeen Y-chromosomal short tandem repeat haplotypes in seven groups of population living in Taiwan

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Abstract The analysis of Y-chromosome short tandem repeat (Y-STR) loci is a powerful tool in forensic casework. The aim of this study was to present the 17 Y-STR loci haplotype distributions of groups of population living in Taiwan and to demonstrate genetic distances between the groups as well as multidimensional scaling plot based on Y-STR genotype data. Five hundred and fifteen DNA samples from unrelated males of seven groups of population, including Taiwanese Han, two groups of indigenous Taiwanese of Taiwan Island, Tao, mainland Chinese, Filipinos, and a group of people with European, Near Eastern, or South Asian ancestry, were analyzed using a commercial kit that co-amplifies 17 Y-STRs. A total of 471 different haplotypes with 440 unique haplotypes were identified. The overall haplotype diversity was 0.9995± 0.0002. High haplotype diversity was observed in six groups of population, except the Tao. These Y-STRs revealed a low discrimination capacity in the Tao population (36.84%), which should be considered in forensic practice. The multidimensional scaling plot of these seven groups of population constructed based on the genetic distances according to 17 Y-STR loci presented a clear patrilineal genetic substructure in the area. Partial Y-STRs profiling reduced the discrimination capacity in most groups of population and distorted the multidimensional scaling plot.

Keywords Y-chromosome · Short tandem repeats · Forensic genetics · Multidimensional scaling plot

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Introduction

The analysis of Y-chromosome short tandem repeat (Y-STR) loci has become a powerful tool in forensic casework. It is useful in patrilineal testing, human identification, and sexual assault cases with mixed male/female samples [1, 2]. Y haplotype distributions in different population groups and varied types of markers have been widely reported. Extensive large-scale population surveys have been carried out and are still ongoing to increase the forensic databases [3–7]. Population genetic diversity in relation to microsatellite heterogeneity has been investigated to trace human evolution and migration, and phylogenetic trees or multidimensional scaling (MDS) plots based on Y-STR have been described [6–13].

A commercial Y-STR kit (AmpF*l*STR® Yfiler) can be used to analyze 17 Y-STR loci, including the nine core Y-STR markers (minimal haplotype, DYS19, DYS389I/II, DYS390, DYS391, DYS392, DYS393, and DYS385a/b) and eight other loci (DYS437, DYS438, DYS439, DYS448, DYS456, DYS458, DYS635, and Y GATA H4) in a single multiplex polymerase chain reaction (PCR), and has been widely used for Y lineage identification in forensic laboratories recently. Because of environmental destruction and the scarcity of forensic samples, incomplete Y-STR profiles are often obtained from trace evidence material or mixed male/female samples in forensic casework. The DYS19, DYS389II, DYS448, and DYS635 loci have been reported to be among the most frequent noninformative loci, followed by DYS385a/b, DYS390, DYS392, DYS437, and DYS438 in the 17 Y-STRs system [6, 14, 15]. Loss of the signals will decrease the discrimination capacity.

The population in Taiwan is heterogeneous and is made up of two major groups, an indigenous population (about 480,000, 1.5%) and a Han population (about 22,470,000, 98.5%) [16]. The indigenous population is comprised of a specific tribe (Tao) living on Orchid Island and 13 tribes living mainly in the mountainous regions of Taiwan proper. Although a common origin is supposed, the origin of indigenous Taiwanese remains obscure. There were different clustering results for the tribes in Taiwan proper. However, Ami tribe located in the eastern portion of Taiwan separated from other tribes by Central Mountain Range is found to be relatively different from other tribes in Taiwan proper [17, 18]. The Taiwanese Han population includes the descendants of individuals who migrated from China in a number of waves of immigration beginning 400 years ago and up to 1949. There are also populations from other countries working or living in Taiwan presently (about 680,000) [19]. The available data on the Y-STR genetic diversity of population groups living in Taiwan is still limited.

The aim of this study is to present the 17 Y-STR loci haplotype distributions of populations living in Taiwan. Based on the Y-STR haplotype distributions, the genetic distances between these population groups can be estimated, and a patrilineal MDS plot can be demonstrated. We also calculated the genetic distances and constructed MDS plots based on partial Y-STR genotypic data in order to reveal the effect of missing signals of Y-STR loci.

Materials and methods

Sample sources and DNA extraction

This retrospective study was approved by the Institutional Review Board. A total of 515 DNA samples from apparently healthy and unrelated males were analyzed. There were 200 Taiwanese Han (TWH), 27 Ami (AMI), 63 indigenous Taiwanese of Taiwan proper other than Ami (TWI), 42 Tao (TAO), 101 mainland Chinese [CHI, 68.3% (69/101) from South China—Fujian and Guangdong], 55 Filipinos (FIL), and 27 people with European, Near Eastern, or South Asian ancestry (ENS, including seven from the USA, five from the UK, three from Australia, two from France, two from Germany, two from New Zealand, one from Denmark, one from Netherlands, one from Peru, one from Brazil, one from Jordan, and one from Syria). The blood samples and buccal swab samples were obtained from volunteer donors between 1993 and 2007. Standard methods of phenol–chloroform isoamyl alcohol extraction and QIAamp blood kit (Qiagen, Hilden, Germany) were used for DNA extraction from peripheral whole blood samples, whereas the Viogene Blood & Tissue Genomic DNA extraction Miniprep system (Viogene, Taipei, Taiwan) was used for DNA extraction from buccal cells.

Y-STR analysis

Seventeen Y-STR loci (DYS19, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS385a/b, DYS437, DYS438, DYS439, DYS448, DYS456, DYS458, DYS635, and Y GATA H4) were genotyped using the AmpF*l*STR® Yfiler PCR amplification kit (Applied Biosystems, Warrington, UK). All PCRs were performed according to the instructions provided by the manufacturer. Electrophoresis was performed using an ABI 3100 Genetic Analyzer (Applied Biosystems). Fragment sizes were automatically determined using GeneScan Analysis software v3.7 (Applied Biosystems). Genotyping was analyzed using either Genotyper v3.6 (Applied Biosystems) or GeneMapper ID V3.2 software (Applied Biosystems). The nomenclature of the Y-STR loci studied has followed the updated recommendations of the DNA Commission of the International Society of Forensic Genetics for analysis of Y-STR systems [20].

Statistical analyses

Characteristic parameters for each population, including discrimination capacity and the haplotype diversity, were calculated as described by Kwak et al. [4]. Briefly, gene or haplotype diversity was estimated according to the formula presented by Nei [21]. The discrimination capacity was assessed as the percentage proportion of individual-specific haplotypes [22]. The genetic relationship between different populations was calculated following the description by Excoffier et al. [23, 24]. Statistical analyses were performed using Arlequin 3.1 software [25] and tested for statistical significance using 10,000 randomizations. The non-

standard alleles (partial or duplicated) were treated as "genotypes," with the two alleles separated by a hyphen in genetic distance calculation [20]. The genetic distances among different populations were explored by analysis of molecular variance implemental in Arlequin 3.1 (http://cmpg.unibe.ch/software/arlequin3). A difference with a p value<0.05 was taken as statistically significant. Reynolds' genetic distances [26] between groups of population were estimated using PHYLIP v3.5 [27]. For analyses, DYS389 was considered as a haplotype of two independent loci: DYS389I and DYS389II–I. The repeat number of DYS389I was subtracted from that of DYS389II [28]. Y-STR data for 17 markers of 30 YRI (Yoruba in Ibadan, Nigeria, and Africa), 22 CHB (Han Chinese in Beijing), 22 JPT (Japanese in Tokyo), and 30 CEU (CEPH, Utah residents with ancestry from Northern and Western Europe), obtained from a previous report by He et al. [28], was incorporated in our study. Based on Reynolds' genetic distances, MDS plot was performed using the Statistical Package for the Social Sciences (SPSS) v16.0 (SPSS Inc., Chicago, IL, USA). We deleted the data of the four Y-SYR loci (DYS19, DYS389II, DYS448, and DYS635) with the highest profiling failure rate and further deleted the data of the six loci (DYS385a/b, DYS390, DYS392, DYS437, and DYS438) with a relatively high failure rate. Based on incomplete Y-STR genotype data, the consequent forensic parameters and Reynolds' genetic distances between groups were estimated, and MDS plots were constructed.

Results and discussion

Y-chromosomal STR profiling is important in forensic science and has been considered as the marker of choice for local population genetic studies [29]. A total of 515 subjects from seven groups of population were analyzed with 471 different haplotypes. Four hundred and forty haplotypes were unique, 23 occurred in two individuals, four occurred in three individuals, three occurred in four individuals, and one occurred in five individuals (Supplementary Table S1). Among 31 shared haplotypes, 28 were shared

Table 1 Combined Y-chromosome short tandem repeat (Y-STRs) forensic parameters of seven groups of population

Population (number of cases)	TWH (200)	TWI (63)	AMI (27)	TAO (42)	CHI (101)	FIL (55)	ENS (27)	Pooled (515)
Complete 17 loci profiling								
Ht No	194	55	26	19	101	52	27	471
Uni Ht	188	49	25	7	101	49	27	440
DC (%)	94.00	77.78	92.59	36.84	100.00	89.09	100.00	85.44
PD	0.999999989	0.999993844	0.999999596	0.999997323	0.999999989	0.999999982	0.999999955	0.999999992
HtD±SD	0.9997± 0.0005	0.9949± 0.0041	0.9972± 0.0111	0.9535± 0.0128	1±0.0014	0.998± 0.0039	1±0.0101	0.9995± 0.0002
Incomplete profiling A								
Ht No	187	49	24	15	99	52	27	440
Uni Ht	175	40	21	5	97	49	27	392
DC (%)	87.50	63.49	77.78	11.90	96.04	89.09	100.00	76.12
PD	0.999998175	0.999695346	0.999960212	0.999930237	0.999998190	0.999997212	0.999996361	0.999998508
HtD±SD	0.9993± 0.0006	0.9887± 0.0059	0.9915± 0.0125	0.9268± 0.0165	0.9996± 0.0015	0.998± 0.0039	1±0.0101	0.9992± 0.0002
Incomplete profiling B								
Ht No	153	48	23	14	85	49	26	350
Uni Ht	128	39	20	5	77	43	25	264
DC (%)	64.00	61.90	74.07	11.90	76.24	78.18	92.59	51.26
PD	0.998708735	0.996194198	0.997791096	0.998288917	0.998722123	0.999022839	0.998377649	0.999228023
HtD±SD	0.9955± 0.0014	0.9872± 0.0063	0.9858± 0.015	0.9152± 0.0183	0.9929± 0.0036	0.996± 0.0043	0.9972± 0.0111	0.9982± 0.0003

Incomplete profiling A: based on 13 Y-STR loci (DYS385a/b, DYS389I, DYS390, DYS391, DYS392, DYS393, DYS437, DYS438, DYS439, DYS456, DYS458, and Y GATA H4)

Incomplete profiling B: based on seven Y-STR loci (DYS389I, DYS391, DYS 393, DYS439, DYS456, DYS458, and Y GATA H4)

Ht No number of different haplotypes, *Uni Ht* unique haplotypes, *DC* discrimination capacity, *PD* power of discrimination, *HtD±SD* haplotype diversity±standard deviation

Table 2 Y-chromosome short tandem repeat haplotype pairwise Fst values between the seven groups of population

Below diagonal: pairwise Fst values. Non-significant value ($p>0.05$) is presented in italic. Above diagonal: p values. Non-significant value ($p>0.05$) is presented in bold

	TWH	TWI	AMI	TAO	CHI	FIL	ENS
TWH	–	<0.001	<0.001	<0.001	**0.14414**	<0.001	<0.001
TWI	0.12687	–	<0.001	<0.001	<0.001	<0.001	<0.001
AMI	0.05415	0.12480	–	<0.001	<0.001	0.01802	<0.001
TAO	0.11178	0.13055	0.09416	–	<0.001	<0.001	<0.001
CHI	*0.00185*	0.13197	0.04662	0.10360	–	<0.001	<0.001
FIL	0.03151	0.11394	0.02544	0.09082	0.02651	–	<0.001
ENS	0.09686	0.19489	0.12133	0.21681	0.09273	0.07924	–

within the same population, and three were found between two populations (two haplotypes shared between Taiwanese Han and Chinese; one haplotype shared between a Chinese and a Filipino; Supplementary Table S1).

The allelic frequencies of 17 Y-STR loci from these seven groups of population are shown in Supplementary Table S2. The allele frequency distribution of Taiwanese Han was similar to that of a previous report from Taiwan among 15 of 17 loci, except loci DYS448 and DYS635. The most frequent alleles at loci DYS448 and DYS635 in the previous report were 18 and 21 repeats, respectively, but 19 and 20 repeats, respectively, in our data [5]. The allele frequency distribution among ten (DYS19, DYS385a/b, DYS389I/II, DYS390, DYS391, DYS393, DYS438, and DYS439) of 11 Y-STR loci (except DYS392) of an American Caucasian population was similar to the data of our ENS [30]. In a previous report from Austria, the allele distribution of 13 Y-STRs loci (DYS19, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS439, DYS448, DYS456, DYS458, DYS635, and Y GATA H4) was similar to that of our ENS group. For loci DYS437 and DYS438, the most frequent alleles were 15 and 10, respectively, in the Austrian population, and 14 and 12, respectively, in our small ENS group [31]. The allele frequency distribution of all nine Y-STR loci (DYS19, DYS385a/b, DYS389I, DYS389II, DYS390, DYS391, DYS392, and DYS393) in a previous Filipino population report was similar to that of Filipinos in this study [32]. The allele frequency distribution among 14 of 16 Y-STR loci of a Chinese population reported previously was similar to that of mainland Chinese in this study [33]. However, the most frequent alleles of loci DYS392 and DYS635 were 14 and 20, respectively, in that Chinese group, and 13 and 21, respectively, in our Chinese group [33].

The duplicated marker DYS385a/b is the most informative marker for all population groups. For Taiwanese Han and mainland Chinese, the locus DYS391 is the least

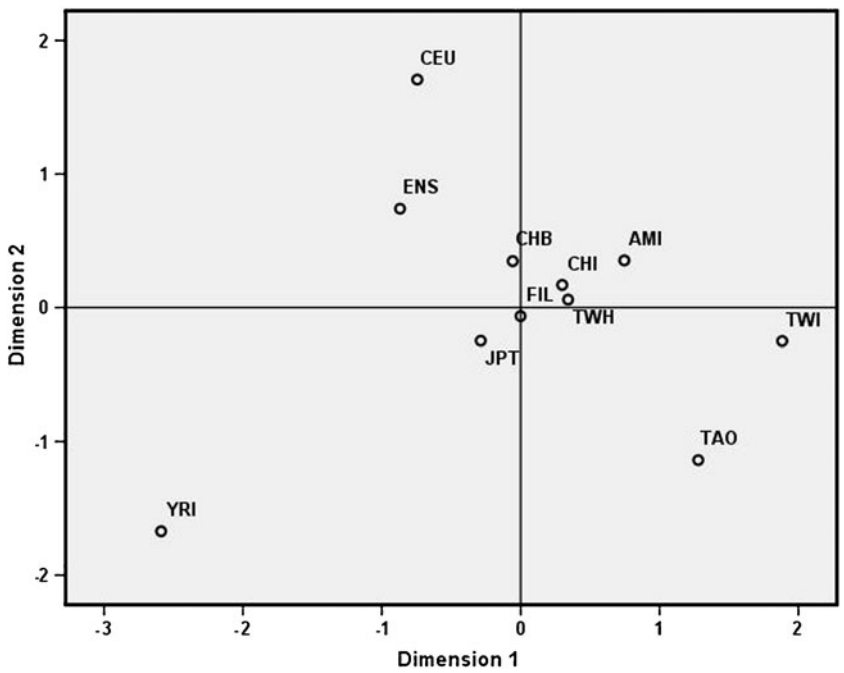

Fig. 1 The multidimensional scaling plot constructed based on the Reynolds' genetic distances between seven groups of population based on data of all 17 Y-chromosome short tandem repeat loci

discriminative marker. For the Tao, the locus DYS437 is the least informative locus, with only one kind of allele. For Ami, the locus DYS438 is the least informative locus, with one kind of allele. For all three groups of indigenous Taiwanese, the loci DYS437 and DYS438 are less discriminative. These findings may result from genetic drift and the isolated patrilineality of the indigenous Taiwanese groups [34].

Table 1 presents the forensic parameters of the seven groups of population in this study. The overall haplotype diversity was 0.9995±0.0002. High haplotype diversity was observed in six groups of population, except the Tao group (0.9535±0.0128). A reduced discrimination capacity (36.84%) was also noted in the small isolated Tao population, which was probably due to genetic drift. The low discrimination capacity of these Y-STRs markers in Tao population should be taken into account in forensic practice. After removing data of the four most labile loci and further removing the six relatively labile loci, the discrimination capacity reduced significantly in most of the groups.

The pairwise Fst between these seven groups of population are shown in Table 2. The genetic distance between Taiwanese Han and Chinese is non-significant. The other population groups revealed significant genetic distances between each other. This result is consistent with the migration of Taiwanese Han from mainland China in the past 400 years.

The MDS plot of our seven groups of population and four groups of population (YRI, CHB, JPT, and CEU) from He et al. [28], constructed on the basis of the Reynolds' genetic distances according to the data of 17 Y-STRs, is illustrated in Fig. 1. This plot showed a clear patrilineal genetic substructure in this area. The TWH, CHI, FIL, CHB, and JPT were clustered together. After removal of the data of the four most labile loci (DYS19, DYS389II, DYS 448, and DYS635), the JPT separated from this cluster (Supplementary Figure S1). After further deletion of the data of the six loci with relatively higher failure rates (DYS385a/b, DYS390, DYS392, DYS437, and DYS438), the TWH, CHI, and CHB still clustered together (Supplementary Figure S2). However, the JPT separated widely from this cluster seemed unreasonable. These results suggest that the particular loci being analyzed affect the structure of the MDS plot, and the MDS plots may be confusing without data of enough number of Y-STR markers.

In conclusion, this study presented the 17 Y-STR loci data of six groups of Asian population and a small group of subjects with European, Near Eastern, or South Asian ancestry. The low discrimination capacity of these Y-STRs in Tao should be considered in forensic practice of this population. The MDS plot constructed based on genetic distances between these population groups presented a clear genetic substructure for the area. Partial Y-STRs profiling reduced the discrimination capacity in most groups of population and distorted the MDS plot.

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