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Allele Frequencies of STR CODIS 13 of Madura Ethnic from Bangkalan and Probolinggo

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Abstract One of forensic identification that can be done is through DNA testing. FBI recommends to use 13 *short tandem repeat* loci known as CODIS 13 for forensic DNA testing. We tested such system to characterize and determine whether individuals with Madura ethnic background (Madurese) resides in different region differ in CODIS 13 pattern. Employing standard PCR-RFLP for STR CODIS 13 our results showed that indeed the CODIS 13 pattern between the two regions were distinct. Most frequent alleles appeared in Madura ethnic from Bangkalan were at locus D3S1358: allele 16; VWA: 16; FGA: 24 and 26; TH01: 8; TPOX: 9; CSF1PO: 10; D5S818: 11 and 13; D13S317: 14; D7S820: 9; D8S1179: 15; D21S11: 27, 28 and 30; D18S51: 16; and D16S539: 15. Most frequent alleles appeared in each locus of Madura ethnic from Probolinggo were locus D3S1358: allele 17; VWA: 17; FGA: 23 and 26; TH01: 7; TPOX: 10; CSF1PO: 10, 11 and 13; D5S818: 11 and 12; D13S317: 11; D7S820: 11; D8S1179: 11 and 12; D21S11: 30; D18S51: 14; and D16S539: 8. The heterozygosity levels of Madura ethnic from Bangkalan's CODIS 13 varied from 0.4180 (D5S818) to 0.9102 (FGA). The power of exclusion was between 0.1569 (D5S818) to 0.7667 (FGA). The power of discrimination was between 0.2816 (D5S818) to 0.9811 (FGA). The paternity index was between 0.8591 (D5S818) to 5.5653 (FGA). The heterozygosity index of Madura ethnic from Probolinggo varied between 0.63021 (VWA) to 0.94445 (FGA), power of exclusion was between 0.33418 (VWA) to 0.84779 (FGA), and power of discrimination ranged from 0.60501 (TPOX) to 0.99305 (FGA).

Introduction

Forensic identification plays role in victim identification, which can be done by comparing ante mortem and post mortem data. DNA testing is an alternative that can be done when the victim body suffered a severe damage. Nowadays, DNA testing in Indonesia uses FBI standard of CODIS 13 loci [1, 2]. Currently the database for Indonesia is limited [3]. As Indonesia consists of many ethnics who may reside in different part of this archipelagic country, it is interesting as well as mandatory to know whether the CODIS 13 pattern has a specific pattern for each existing ethnic. In the present study, as a model, Madura ethnic (Madurese) was chosen.

Madurese is one of minor ethnics in Indonesia which comprise 0.03% of the population, whereas the major ethnics are Javanese (40%) and Sundanese (15%) [4]. Madurese is one of the ethnics that spread in many regions in Indonesia. Most of them live in Bangkalan, Pamekasan, Sumenep, and Sampang in Madura Island. Some live in Java and other islands. In East Java province, most of Madurese live also in Probolinggo, Pasuruan, Situbondo,

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Lumajang, Malang and other cities. To represent the different regions of Madurese people, subjects for the present study were taken from Bangkalan of Madura Island and Probolinggo of eastern part of Java Island. It is suspected that the pattern of CODIS 13 of Madurese in the two regions differ due to socio-culture context, which rather closed and restrictive in Madura Island compared to other regions.

Materials and methods

The sample comprised of 60 volunteers (30 from Bangkalan and 30 from Probolinggo) aged 18–60 years, who were consent to donate blood for genetic studies. All participants were asked and confirmed that they were of Madurese ethnic for three generations. Blood were withdrawn from arm vein according to standard protocol for DNA isolation. All treatments were approved by the Ethical Committee for Health Research of Faculty of Medicine, University of Brawijaya.

DNA was isolated from blood using Nucleospin Blood Quick Pure kit essentially as described elsewhere [5].

Amplification of 13 STR loci were performed by using PCR application (Takara), with a total volume of 10 µl per-tube were consist of 6 µl ddH_2O, 10 µl PCR master mix kit (10x buffer Taq polymerase, dNTP, $MgCl_2$, primer, Taq DNA Polymerase, ddH_2O), 1 µl primer forward, primer reverse 1 and 2 µl of blood genomic DNA with PCR cycles 94° C 1 minute for pre-denaturation, followed by 35 cycles of 94°C 1 minute denaturation, 60° C 1 minute for annealing, and 72° C 1 minute for extension, and one cycle of 72° C for final incubation [6].

Electrophoresis were done using 8% polyacrylamide gel, visualized and documented using GelDoc BioRad as described [6].

Alleles for each CODIS 13 locus were analyzed for heterozygosity index, power of exclusion, power of discrimination and paternity index [7].

Results and Discussion

Our results showed that the most frequent alleles detected in each locus of Madura ethnic from Bangkalan were D3S1358, 16 ;VWA, 16; FGA, 24 and 26; TH01,8; TPOX, 9; CSF1PO, 10; D5S818, 11 and 13; D13S317, 14; D7S820, 9; D8S1179, 15; D21S11, 27, 28 and 30; D18S51, 16; and D16S539, 15. The calculated heterozygosity level of Madura ethnic from Bangkalan varied from 0.4180 (D5S818) to 0.9102 (FGA), the power of exclusion was from 0.1569 (D5S818) to 0.7667 (FGA), the power of discrimination was from 0.2816 (D5S818) to 0.9811 (FGA). The paternity index was from 0.8591 (D5S818) to 5.5653 (FGA) (Table 1).

Some of the alleles detected in Bangkalan were also found in Probolinggo, however most frequent alleles that appear in each locus of the Madurese in Probolinggo were different. At locus D3S1358, allele 17 was in high frequency. Other loci and most frequent alleles found in Probolinggo Madurese were VWA, 17; FGA, 23 and 26; TH01,7; TPOX, 10; CSF1PO, 10,11 and 13; D5S818, 11 and 12; D13S317, 11; D7S820, 11; D8S1179, 11 and 12; D21S11, 30; D18S51, 14; and D16S539, 8. The heterozygosity index of Madura ethnic from Probolinggo varied between 0.63021 (VWA) to 0.94445 (FGA). The power of exclusion was between 0.33418 (VWA) to 0.84779 (FGA). The power of discrimination was between 0.60501 (TPOX) to 0.99305 (FGA) (Table 2).

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Table 1. CODIS 13 allele distribution of Madurese in Bangkalan

Allele	TPOX	CSF1PO	TH01	vWA	D3S1358	FGA	D13S317	D16S539	D18S51	D5S818	D7S820	D8S1179	D21S11
5													
6			0.0.25										
7													
8	0.0625		0.375								0.0625		
9	0.5										0.3125		
10	0.1875	0.5	0.0625								0.125		
11		0.0625								0.375			
12							0.0625			0.1875		0.125	
13					0.0625		0.5	0.0625		0.375		0.125	
14				0.0625	0.0625		0.25	0.375				0.1875	
15				0.0625	0.25		0.125	0.5				0.5	
16				0.375	0.4375				0.375				
17				0.25					0.25				
18									0.125				
19				0.0833					0.0.25				
20													
21													
22													
23						0.125							
24						0.1875							0.0625
25						0.0625							
26						0.1875							0.125
27													0.25
28													0.25
29													
30													0.25
31													
32													
H*	0.7109	0.7461	0.6288	0.60069	0.7383	0.9102	0.6680	0.6055	0.7188	0.4180	0.8828	0.6836	0.7930
PE	0.4316	0.4780	0.5469	0.30885	0.4673	0.7667	0.3801	0.3136	0.4415	0.1569	0.7012	0.3982	0.5469
PD	0.7691	0.8086	0.8906	0.52853	0.8224	0.9811	0.7129	0.6064	0.8140	0.2816	0.9628	0.7356	0.9023
PI	1.7297	1.9692	2.4151	1.25219	1.9105	5.5653	1.5059	1.2673	1.7778	0.8591	4.2667	1.5803	2.4151

*Heterozygosity index (H), Power of Exclusion (PE), Power of Discrimination (PD), Paternity Index (PI)

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Table 2. CODIS 13 allele distribution of Madura ethnic in Probolinggo

Allele	TPOX	CSF1PO	TH01	vWA	D3S1358	FGA	D13S317	D16S539	D18S51	D5S818	D7S820	D8S1179	D21S11
5													
6								0.0833			0.1667		
7			0.5000								0.1667		
8			0.2500					0.4167			0.2500		
9	0.0833		0.0833					0.3333					
10	0.7500	0.8333	0.1667				0.1667	0.1667		0.1667	0.0833	0.1667	
11	0.1667	0.0833					0.5000			0.2500	0.3333	0.2500	
12							0.2500			0.2500		0.2500	
13		0.0833					0.0833		0.1667	0.1667			
14									0.4167	0.1667			
15									0.0833				
16				0.0833	0.2500							0.3333	
17				0.7500	0.4167				0.0833				
18				0.1667	0.2500								
19					0.0833				0.0833				
20													
21						0.0833							
22						0.2500							
23						0.3333			0.0833				
24									0.0833				
25						0.0833							
26						0.0833							0.0833
27													0.1667
28													0.1667
29						0.1667							0.0833
30													0.2500
31													0.1667
32													0.0833
H*	0.7326	0.78126	0.84897	0.63021	0.82119	0.94445	0.91945	0.77431	0.94098	0.90625	0.84029	0.82982	0.85244
PE	0.6275	0.52887	0.64228	0.33418	0.59303	0.84779	0.78781	0.51842	0.83917	0.75807	0.626465	0.60790	0.64874
PD	0.6050	0.86012	0.94497	0.60994	0.91531	0.99305	0.98282	0.87775	0.99109	0.96491	0.93572	0.92756	0.94849
PI	1.8701	2.28577	3.31051	1.35211	2.79619	9.00074	6.20694	2.2154	8.47170	5.33356	3.13069	2.93801	3.38845

*Heterozygosity index (H), Power of Exclusion (PE), Power of Discrimination (PD), Paternity Index (PI)

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The differences observed within same ethnicity but different region may due to the main contribution of genetic differentiation between populations linked to historical events and demographic processes leading to genetic drift [8, 9].

Conclusion

Our study concluded that individuals with Madura ethnic background (Madurese) resided in different region differed in their CODIS 13 pattern. Further studies using more samples from different region may result in distinct pattern which helpful in narrowing down the ethnic and residential origin of individuals.

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