

## FOR THE RECORD

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# Genetic Variation at Three DYS-STR Loci and Amelogenin Between the Australian Caucasians and Three Asian Ethnic Groups

**POPULATION:** Male population samples from Caucasians ( $n = 130$ ), Malays ( $n = 113$ ), Chinese ( $n = 113$ ) and Indians ( $n = 112$ ). The Caucasians are from Australia and the three Asian ethnic groups are from Malaysia. A mixed male DNA sample, derived from approximately 500 sperm donors is from an Australian fertility clinic in 1984.

**KEYWORDS:** forensic science, DNA typing, population genetics, DYS438, DYS390, DYS439, Australia, Malaysia

Blood samples were obtained from unrelated male individuals from the four population groups and placed on FTA<sup>®</sup> paper (Fitzco-Whatman). DNA was extracted using the aqueous processing method for FTA<sup>®</sup> paper. One extracted disc (1.2 mm diameter) was used in the PCR per sample. The mixed male DNA sample was loaded onto a pre-washed FTA<sup>®</sup> paper and subjected to an alkaline-isopropanol treatment. A combination of three Y-linked microsatellites loci, DYS438 (1), DYS390 (2), and DYS439 (1) and amelogenin were amplified simultaneously using a fluorescent multiplex system. DYS438 and amelogenin were labelled with 6-FAM, DYS390 with JOE and DYS439 with TAMRA.

Extracted discs were amplified with 15  $\mu$ L of master mix containing: DYS438 primer pair (each at 0.13  $\mu$ M), DYS390 primer pair (each at 0.10  $\mu$ M), DYS439 primer pair (each at 0.50  $\mu$ M), amelogenin primer pair (each at 0.01  $\mu$ M), 1X GeneAmp<sup>®</sup> PCR Buffer II (PE Applied Biosystems, containing 10 mM Tris-HCl pH 8.3, 50 mM KCl (PCR Buffer), 1.5 mM MgCl<sub>2</sub>, 200  $\mu$ M of each dNTPs (Perkin Elmer GeneAmp<sup>®</sup>), 1.2U AmpliTaq Gold<sup>™</sup> polymerase (PE Applied Biosystems) and 16 mg/mL Bovine Serum Albumin (BSA) from Sigma. Optimized cycling conditions were: 95°C for 11 min, 30 cycles at 94°C for 1 min, 57°C for 3 min and 72°C for 3 min, 72°C at 20 min. Amplified products were detected on an ABI PRISM<sup>™</sup> 377 DNA Sequencer. Allele designation (3) was achieved by using two Y-STR control samples from M.A. Jobling (University of Leicester, UK). Locus diversity values ( $D_L$ ), haplotype diversity values ( $D_H$ ) and standard error (SE) were calculated according to the equations by Nei (4).

Genetic variation of the three DYS-STRs loci between the four groups was greatest with respect to DYS438 and least with respect

to DYS439. Y-STR locus diversity values ranged from 0.365 to 0.774 (shown in Table 1). Locus diversity for DYS438 was highest in Indians (0.685), for DYS390 highest in Malays (0.774), and for DYS439 highest in Indians (0.734). A total of 74 different haplotypes were found in the four population groups (Table 2). Thirty seven haplotypes were unique. Two groups shared eight haplotypes and three groups shared 18 haplotypes. Only 11 haplotypes were shared by the four population groups. Haplotype diversity of the Y-STRs was highest in the Malays (0.964), followed by the Indians (0.955), the Caucasians (0.940), and the Chinese (0.928). An amelogenin null allele was encountered in four male Indian individuals. These four samples showed a total absence of the Y-allele (112 bp) in their Genescan<sup>™</sup> profiles.

DYS438 was found to have a strong ethnic affiliation in a comparison of the Caucasians and the Asians of this study. DYS438-allele 12 (232 bp) exists in high frequency (>50%) in the Caucasian group compared with the three Asian groups (0.9% in Malays, 0.9% in Chinese, and 1.8% in Indians). DYS438-allele13 (237 bp) was not observed in the three Asian ethnic groups and was only observed in the Caucasian group (3.9%). The mixed male DNA sample (predominantly from Australian Caucasians) produced the same number of alleles at DYS438 (5 alleles) and DYS439 (5 alleles) as the typed Caucasian population sample, except for DYS390, which had 6 alleles. The DYS438-allele12 (232 bp) in the mixed male DNA sample had a frequency of 52.6%, compared with 55.4% in the typed Caucasian population sample (Table 3), which further reinforces the presence of this characteristic allele in Australian Caucasians.

The complete data can be accessed at [http://daphne.bio.flinders.edu.au/jc-y\\_str/](http://daphne.bio.flinders.edu.au/jc-y_str/)

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TABLE 1— *Y-STR alleles for DYS438, DYS390 and DYS439 of the Caucasian, Malay, Chinese and Indian population groups.*

Locus	bp*	Allele Units†	Caucasian		Malay		Chinese		Indian	
			No.	Allele Freq. ( <i>p</i> )	No.	Allele Freq. ( <i>p</i> )	No.	Allele Freq. ( <i>p</i> )	No.	Allele Freq. ( <i>p</i> )
DYS438	212	8	0	0.0000	1	0.0088	0	0.0000	1	0.0089
	217	9	5	0.0385	12	0.1062	1	0.0088	39	0.3482
	222	10	34	0.2615	74	0.6549	87	0.7699	40	0.3571
	227	11	14	0.1077	25	0.2212	24	0.2124	30	0.2679
	232	12	72	0.5538	1	0.0088	1	0.0088	2	0.0179
	237	13	5	0.0385	0	0.0000	0	0.0000	0	0.0000
Total individuals ( <i>n</i> )			130	1	113	0.9999	113	0.9999	112	1
Locus diversity ( $D_L$ )			0.615		0.515		0.365		0.685	
Standard error (SE)			0.024		0.031		0.243		0.008	
DYS390	201	20	0	0.0000	1	0.0088	0	0.0000	0	0.0000
	205	21	0	0.0000	12	0.1062	0	0.0000	4	0.0357
	209	22	14	0.1077	10	0.0885	9	0.0796	45	0.4018
	213	23	37	0.2846	36	0.3186	49	0.4336	23	0.2054
	217	24	57	0.4385	30	0.2655	35	0.3097	17	0.1518
	221	25	21	0.1615	23	0.2035	19	0.1681	21	0.1875
	225	26	1	0.0077	1	0.0088	1	0.0088	2	0.0178
Total individuals ( <i>n</i> )			130	1	113	0.9999	113	0.9998	112	1
Locus diversity ( $D_L$ )			0.694		0.774		0.687		0.743	
Standard error (SD)			0.016		0.011		0.016		0.008	
DYS439	241	10	5	0.0385	10	0.0885	6	0.0531	23	0.2053
	245	11	51	0.3923	38	0.3363	36	0.3186	34	0.3036
	249	12	56	0.4308	52	0.4602	57	0.5044	38	0.3393
	253	13	16	0.1230	12	0.1062	11	0.0973	17	0.1518
	257	14	2	0.0154	1	0.0088	3	0.0265	0	0.0000
Total individuals ( <i>n</i> )			130	1	113	1	113	0.9999	112	1
Locus diversity ( $D_L$ )			0.649		0.662		0.637		0.734	
Standard error (SD)			0.015		0.018		0.021		0.009	

\* Amplicon sizes of Y-STRs in base pairs.

† Allele units determined by two Y-STR control samples (C1 and E1).

Locus diversity and standard error calculated according to formulae by Nei (4).

TABLE 2—Y-STR haplotypes and frequencies for DYS438, DYS390, and DYS439 of the Caucasian, Malay, Chinese and Indian population groups.

	Haplotypes	Caucasian	Malay	Chinese	Indian
1	213 205 245				1
2	213 213 249		1		
3	217 205 241				1
4	217 209 241				1
5	217 209 245		3		10
6	217 209 249		1		6
7	217 209 253				1
8	217 213 245	1	1		4
9	217 213 249		4	1	4
10	217 213 253				1
11	217 217 245		1		4
12	217 217 249	2			2
13	217 217 253	2			1
14	217 221 245		2		2
15	217 221 249				2
16	222 201 249		1		
17	222 205 245		6		1
18	222 205 249		5		
19	222 205 253		1		1
20	222 209 241			3	
21	222 209 245	10	2	1	2
22	222 209 249	3	2	4	15
23	222 209 253			1	8
24	222 213 241		1		
25	222 213 245	8	6	15	1
26	222 213 249	1	10	19	2
27	222 213 253	3	3	4	1
28	222 213 257			1	
29	222 217 241		1	2	
30	222 217 245	2	6	9	1
31	222 217 249	3	10	12	1
32	222 217 253	3	2	2	1
33	222 217 257	1			
34	222 221 241				2
35	222 221 245		5	5	
36	222 221 249		8	7	3
37	222 221 253		4		
38	222 221 257			1	
39	222 225 245		1		1
40	222 225 249			1	
41	227 209 245		1		
42	227 209 249		1		
43	227 209 253				1
44	227 213 241	2	1		5
45	227 213 245	1	2	2	2
46	227 213 249		4	6	2
47	227 213 253		1		
48	227 213 257		1	1	
49	227 217 241	1	3		4
50	227 217 245	1	2	3	3
51	227 217 249		4	4	
52	227 217 253		1	2	
53	227 221 241	2	4	1	9
54	227 221 245	5		1	2
55	227 221 249	2		2	
56	227 221 253			2	1
57	227 225 241				1
58	232 209 249	1			
59	232 209 253				1
60	232 213 245	6			
61	232 213 249	11	1		1
62	232 213 253	3			
63	232 217 245	12			
64	232 217 249	22		1	
65	232 217 253	4			
66	232 217 257	1			

(continues)

TABLE 2—(Continued)

	Haplotypes	Caucasian	Malay	Chinese	Indian
67	232 221 245	2			
68	232 221 249	8			
69	232 221 253	1			
70	232 225 249	1			
71	237 213 249	1			
72	237 217 245	2			
73	237 217 249	1			
74	237 221 245	1			
Total individuals analysed (n):		130	113	113	112
Number of different haplotypes:		35	38	28	40
Haplotype diversity value ( $D_H$ ):		0.9395	0.964	0.9283	0.9553
Standard error (SE):		0.0066	0.0035	0.0074	0.0055

TABLE 3—Comparison of *DYS438*, *DYS390* and *DYS439* allele frequencies of the mixed male DNA sample and the typed Caucasian population samples.

<i>DYS438</i> Alleles (in bp)	Frequency (in %)* of the Mixed Male DNA Sample	Frequency (in %) of the Typed Caucasian Population
217	8.37	3.85
222	26.17	26.15
227	10.49	10.77
232	52.59	55.38
237	2.38	3.85
<i>DYS390</i> Alleles (in bp)	Frequency (in %)* of the Mixed Male DNA Sample	Frequency (in %) of the Typed Caucasian Population
205	2.70	0
209	19.90	10.77
213	26.48	28.46
217	37.57	43.85
221	13.11	16.15
225	0.24	0.77
<i>DYS439</i> Alleles (in bp)	Frequency (in %)* of the Mixed Male DNA Sample	Frequency (in %) of the Typed Caucasian Population
241	5.47	3.85
245	34.41	39.23
249	47.66	43.08
253	10.43	12.30
257	2.02	1.54

\* Frequency estimated from peak area in GeneScan™ profile.

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