

Y-kromozomu 17 STR markırının Türkiye'deki polimorfizmi Polymorphism of 17 Y-chromosome STR markers in Türkiye

Gönül Filoğlu¹, Havva Altuncul¹, Özlem Bülbül¹, Elif Mertoğlu², Tolga Zorlu¹,
Gülten Rayimoğlu¹, Cengiz Salih¹

Corresponding author: Filiz Civil Arslan

Institute of Forensic Science, Istanbul University, Istanbul, Türkiye, email: gonulfiloglu@yahoo.com.tr

ÖZET

AMAÇ: Adli araştırmalarda, erkek ve kadın DNA örnekleri genellikle karışım halde bulunurlar. Bu tür örneklerin tiplendirilmesinde Y-STR kullanılmaktadır. Bu çalışmanın amacı Türkiye popülasyonuna ait homojen dağılımın gösteren bir Y-STR haplotip veri tabanını oluşturmak ve diğer dünya popülasyonlarında önceden yapılmış Y-STR sonuçlarıyla karşılaştırmaktır.

YÖNTEM: Kan veya ağız içi sürüntü örnekleri 218 sağlıklı, aralarında akrabalık ilişkisi bulunmayan ve çalışmaya gönüllü katılmayı kabul eden Türkiye'deki erkek bireylerden alınmıştır. 17 Y-STR lokuslarının haplotip ve alel sıklıkları AmpFISTR® Y-filer™ PCR Amplification Kiti kullanılarak çoğaltıldı ve ABI 3130 cihazında analiz edildi. Gen ve haplotip alel sıklıkları Arlequin Software 3.01 kullanılarak hesaplandı. Moleküler varyans analiz (AMOVA) sonuçları YHRD çevrimiçi araçları kullanılarak RST şeklinde değerlendirildi.

BULGULAR: Türkiye popülasyonunda 218 örnekte aynı haplotipe rastlanmadı. En yüksek gen çeşitliliği DYS385 a / b lokusunda görülürken ve en düşük DYS391 lokusunda gözlemlendi. Çalışılan popülasyona en yakın popülasyonlar sırasıyla Güneydoğu Anadolu, Kıbrıslı Türkler ve Çukurova popülasyonları oldu.

SONUÇ: Bu çalışma, adli uygulamalarda kullanılan yüksek ayırım gücüne sahip ve 17 Y-STR lokusunun Türk popülasyonuna ait son verilerini içermektedir.

Anahtar Kelimeler: Y-STR, Türkiye, adli bilimler, popülasyon genetiği.

INTRODUCTION

Türkiye, located on continents of Europe and Asia actually serves as a transcontinental bridge between East and West, has exposed a great number of changes since the Paleolithic era to the now present Republic of Turkey. Thus the connection feature of Turkey created a major variety of social and ethnic civilizations during many ages. Turkey is bordered by eight countries:

ABSTRACT

OBJECTIVE: Y- STR typing is useful forensic investigation tool as it often involves a mixture of male and female DNA. The aims of the present study were to type 17 Y-STR loci in order to establish a proper Y-STR haplotype database with homogenous distribution of the Turkish population and compare our results with previously published Y-STR researches about Turkish and other worldwide populations.

METHODS: The blood or buccal swab samples were collected from 218 healthy, unrelated male individuals in Turkey with their informed consent. DNA samples were amplified using the 17 Y-STR loci AmpFISTR Y-filer Amplification Kit and analyzed on ABI 3130 (both Applied Biosystems). Haplotype frequencies were estimated by gene counting. Gene / haplotype diversities and allele frequencies were calculated using the Arlequin Software 3.01. Analyses of Molecular Variance (AMOVA) results were assessed in the form of RST values using the YHRD online tool.

RESULTS: Among 218 samples of Turkey dataset, all of the haplotypes were unique. The highest gene diversity was determined at DYS385 a / b and the lowest was at DYS391 locus. The closest populations were Southeastern Anatolia, Turkish Cypriot and Cukurova populations respectively.

CONCLUSION: This study indicates recent population data of 17 YSTR markers in Turkish population with high discrimination power for forensic applications.

Keywords: Y-STR, Türkiye, forensic sciences, population genetics.

Bulgaria to the northwest; Greece to the west; Georgia to the northeast; Armenia, Iran and the Azerbaijani exclave of Nakhichevan to the east; and Iraq and Syria to the southeast. The Mediterranean Sea is to the south; the Aegean Sea is to the west; and the Black Sea is to the north (1,2). The characteristics that make Y-STRs unique in the human genome are the paternal inheritance remains unchanged among generations and another reason for its uniqueness is the lack of

a homologous chromosome. The polymorphism of Y chromosome has widely been used by forensic experts on identification and male population research (3). The determination of Y-STR haplotype frequencies in different populations is a major point for the proper interpretation of the genetic profile matches in paternity and forensic casework and information on mutation rates is also crucial in kinship analysis (4). Y Chromosome Haplotype Reference Database (YHRD) (www.yhrd.org), a global YSTR haplotype database provides haplotype frequency estimates of worldwide and multiregional population groups (5). Since different Y STR based population data from the various regions of Turkey are available, we analyzed a collective data from different geographic regions of Turkey in order to create a homogenous distribution of Turkish population haplotype frequencies using 17 Y-STR loci including DYS19, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS385a / b, DYS438, DYS439, DYS437, DYS448, DYS456, DYS458, DYS635, and Y-GATA H4 and compared our results with previously published Y-STR researches about Turkish and other relative / worldwide populations.

MATERIAL VE METHOD

The blood or buccal swab samples were collected from 218 healthy, unrelated male volunteers with their informed consent from Istanbul University Cerrahpaşa Faculty of Medicine, Blood Transfusion Center. The samples were chosen according to donors' place of birth in order to represent all regions of the Turkey based on population distribution data of Turkey Statistical Institute in 2010. DNA was extracted from blood using the QIAamp® DNA Blood Mini Kit (QIAGEN, Hilden, Germany) and from buccal swaps using Chelex method. PCR amplification was performed using the AmpFISTR Y-filer Amplification Kit, which includes 17 Y-STR loci (DYS19, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS385ab, DYS437, DYS438,

DYS439, DYS448, DYS456, DYS458, DYS635 and GATA H4). DNA samples were amplified following manufacturer's instructions (AmpFISTR® Y-filer™ PCR Amplification Kit, Applied Biosystems). The amplified products were separated and detected using an ABI Prism 3130 Genetic Analyzer (Applied Biosystems, Foster City, CA, USA) according to the manufacturer's recommended protocol. Results were analyzed using GeneMapper® IDX software (Applied Biosystems, Foster City, CA, USA). Alleles were named according to the updated International Society of Forensic Genetics (ISFG) guidelines for forensic Y-STR analysis (7).

Haplotype frequencies were estimated by gene counting. The discrimination capacity (DC) was determined by dividing the number of different haplotypes by the total number of samples in that population. Haplotype diversity was calculated according to Nei's formula (8). Allele frequencies and gene diversity were calculated using the Arlequin Software 3.01.

Population pairwise genetic distances (RST) were performed by using analysis of molecular variance (AMOVA) tests on YHRD online tool (<http://www.yhrd.org/Analyse/AMOVA>). RST values were ascertained at a significance level of 0.01 using 10,000 permutations. Also genetic distance was illustrated by using multi-dimensional scaling (MDS) analyses with SPSS 21 software. In genetic distance analyses, DYS385a / b was not considered and the number of repeats in DYS389I was subtracted from DYS389II.

Turkish population results (RST values were compared with Beijing (Han) (n = 446) (YA003470), Spain (Galicia) (n = 93) (YA000244), Cyprus (Turkish Cypriot) (n = 253) (YA000884), Israel and Palestinian Authority (n = 163) (YA003643), Macedonia (n = 351) (YA003354), Southeastern Anatolia (n = 86) (YA003727), Greece (Northern Greece) (n = 191) (YA003465), Cukurova region (Turkey) (n = 249) (YA003668), United States (Hispanic Ame-

1 Institute of Forensic Science, Istanbul University, Istanbul, Türkiye

2 Department of Molecular Biology and Genetics, Faculty of Science, Istanbul University, Istanbul, Türkiye

Table 1: Allele frequencies and gene diversity (GD) of 17 Y-STR in Turkish population (continued next page)

Allele	DYS19	DYS389I	DYS389II	DYS390	DYS391	DYS392	DYS393	DSY437	DSY438	DYS439	DSY448	DYS456	DYS458	DYS635	YGATA	DYS385 a/b Genotypes	DYS385
7																915	0.0046
8					0.0138	0.0184			0.0138						0.0275	1013	0.0046
9					0.0734	0.0229	0.0046		0.2752						0.0321	1015	0.0046
9.4																1016	0.0046
10					0.6972	0.0688	0.0183		0.3944	0.1284					0.0642	1021	0.0046
10.2																1022	0.0046
11	0.0138	0.0092			0.1881	0.6101	0.0459		0.1881	0.3991					0.4220	1112	0.0046
12	0.0046	0.1835			0.0275	0.0963	0.4771		0.1193	0.3211					0.3303	1113	0.0229
13	0.1514	0.6422				0.1055	0.3165		0.0092	0.1330		0.0138			0.1147	1114	0.1009
14	0.3991	0.1651				0.0550	0.1009	0.5321		0.0046		0.1193	0.0642		0.0092	1115	0.0321
14.3																1116	0.0229
15	0.2890					0.0138	0.0321	0.3119		0.0138		0.5046	0.2156			1118	0.0183
16	0.1055						0.0046	0.1560				0.2431	0.2615			1119	0.0046
16.2																1212	0.0092
17	0.0275					0.0046					0.0046	0.0963	0.2294			1213	0.0413
17.2																1214	0.0229
18	0.0091					0.0046					0.0458	0.0229	0.0917			1215	0.0413
18.2													0.0183			1216	0.0505
19				0.0046							0.2982		0.0413			1217	0.0183
19.2													0.0459			1218	0.0229
20				0.0046							0.3349		0.0321	0.0642		1219	0.0092
20.2																1220	0.0092
21				0.0550							0.2385			0.2982		1314	0.0459
22				0.1697							0.0505			0.2385		1315	0.0642

Allele	DYS19	DYS389I	DYS389II	DYS390	DYS391	DYS392	DYS393	DSY437	DSY438	DYS439	DSY448	DYS456	DYS458	DYS635	YGATA	DYS385 a/b Genotypes	DYS385
23				0.3532							0.0229			0.3028		1316	0.0550
24				0.2936							0.0046			0.0642		1317	0.0413
25				0.1055										0.0229		1318	0.0275
26			0.0045	0.0092										0.0092		1319	0.0321
27			0.0182	0.0046												1320	0.0092
28			0.0864													1321	0.0046
29			0.3909													1414	0.0046
30			0.2864													1415	0.0413
31			0.1500													1416	0.0367
32			0.0455													1417	0.0138
33			0.0182													1418	0.0229
																1421	0.0046
																1515	0.0138
																1516	0.0046
																1518	0.0138
																1519	0.0046
																1520	0.0046
																1616	0.0092
																1617	0.0275
																1618	0.0092
																1619	0.0138
																1717	0.0092
																1718	0.0092
																1719	0.0138
																1922	0.0046
GD	0.7254	0.5290	0.7358	0.7494	0.4743	0.6013	0.6616	0.5980	0.7221	0.7065	0.7402	0.6651	0.8186	0.7572	0.6969		0.9655

Table 2: Matrix of pairwise genetic distances based on RST (below the diagonal) and their corresponding P values (above diagonal) between the Turkish population and the rest of populations used in comparisons.

Population	Beijing, China [Han]	Cyprus [Turkish Cypriot]	Galicia, Spain [Spanish]	Israel & Palestinian Authority Area [Arab]	Macedonia [Macedonian]	Northern Greece, [Greek]	Southeastern Anatolia, Turkey [Turkish]	US [African American]	US [Hispanic American]	Çukurova, Turkey [Turk]	Turkey
Beijing, China [Han]	-	0.1626	0.1522	0.1572	0.2421	0.2108	0.1424	0.2385	0.1434	0.1372	0.1671
Cyprus [Turkish Cypriot]	0	-	0.0816	0.0645	0.036	0.0189	0.0064	0.1444	0.1369	0.015	0.005
Galicia, Spain [Spanish]	0	0	-	0.1174	0.0884	0.0884	0.0765	0.1696	0.0314	0.0428	0.0613
Israel & Palestinian Authority Area [Arab]	0	0	0	-	0.1216	0.1268	0.0562	0.1754	0.1788	0.0897	0.0686
Macedonia [Macedonian]	0	0	0	0	-	0.0101	0.0535	0.2042	0.1404	0.0469	0.0349
Northern Greece, [Greek]	0	0	0	0	0.0025	-	0.0324	0.1684	0.1407	0.0255	0.0176
Southeastern Anatolia, Turkey [Turkish]	0	0.0718	0	0	0	0	-	0.1406	0.1141	0.0056	0.0023
US [African American]	0	0	0.0001	0	0	0	0	-	0.21	0.1461	0.131
US [Hispanic American]	0	0	0	0	0	0	0	0	-	0.084	0.125
Çukurova, Turkey [Turk]	0	0	0	0	0	0	0.1126	0	0	-	0.0067
Turkey	0	0.0218	0	0	0	0	0.2435	0	0.0117	-	-

P values are shown above and Φ_{st} values below the diagonal.
P values in bold and italics represent statistically insignificant differences.
†The level of significance is $P < 0.01$.

ricans) (n = 1407) (YA003315) and United States (African Americans) (n = 1431) (YA003485 and YA003313) populations (9-18). A quality control check was performed using the proficiency testing of the Y STRs Haplotyping Quality Assurance Exercise 2013 for the markers included in this study. The data was submitted to Y-Chromosome haplotype reference database (YHRD; <http://www.yhrd.org/>) and is available with the accession number: YA003907.

RESULTS

Allele frequency, gene diversity, and identified haplotype results are shown in Table 1. Among 218 samples of Turkey dataset, all of the haplotypes were unique. Haplotype diversity where the diploid loci DYS385a / b excluded was calculated 0.9999 with discrimination capacity of 1.000. Gene diversity values of all loci differed from 0.4743 for DYS391 to 0.9655 for DYS385a / b. Although null alleles were not found in the Turkish dataset, two duplicate alleles were detected on locus DYS389II. Intermediate alleles of 18.2 (n = 4) and 19.2 (n = 10) were observed at DYS458 locus. In order to define the genetic relationship

between Turkey and previous regional Turkish population studies and also other populations, pairwise genetic distances (RST) comparison was calculated and results are shown in Table 3. Results showed that the minimum RST values were obtained from previous Turkish population studies such as; Southeastern Anatolia Turkey (RST = 0.0023, $P > 0.01$), Turkish Cypriot (RST = 0.0050, $P > 0.01$), Cukurova, Turkey (RST = 0.0067, $P > 0.01$) respectively. The observed P values were also confirmed that there is no significant genetic distance between our present and previous studies on Turkish populations (Table 2). Moreover significant genetic distances were observed in the comparison of Turkish population with other compared (whom are not Turkish) populations. Turkish population being more distant from the Balkan, European and Middle Eastern cluster, such as Northern Greece (RST = 0.0176, $P < 0.01$), Macedonia (RST = 0.0349, $P < 0.01$), Galicia, Spain (RST = 0.0613, $P < 0.01$), Israel & Palestinian (RST = 0.0686, $P \leftarrow 0.01$). Asian and American clusters such as Hispanic American (RST = 0.1250, $P < 0.01$), African American (RST = 0.1310, $P < 0.01$), China (RST = 0.1671, $P < 0.01$) were the farthest populations as expected. Illustrating the relationship between populations based on RST

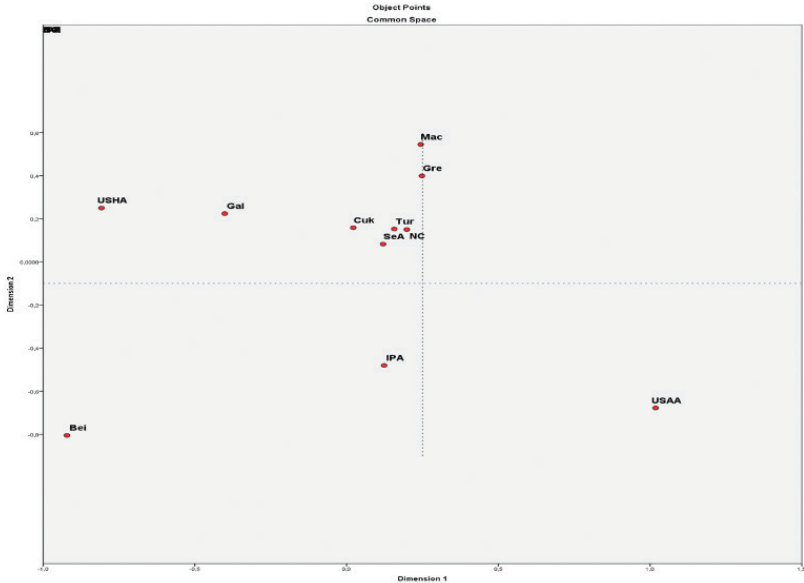


Figure 1: MDS plot based on the pairwise RST values between the Turkish population and neighboring / distant populations. Abbreviations: Tur: Turkish - Turkey; IPA: Palestinian - Israel and Palestinian Authority area; Gre: Greek - Northern Greece; SeA: Turkish - Southeastern Anatolia region; Cuk: Turkish - Cukurova region; USHA: Hispanic - United States Hispanic American; USAA: African - United States African American; Bei: Beijing, China; Gal: Spanish-Galicia, Spain; Mac: Macedonia; NC: Turkish - Cypriot (stress value = 0.020 and DAF = 0.980).

values at MDS plot is outlined in Figure 1 showed that Turkey clustered with Anatolian, Balkan and European populations.

DISCUSSION AND CONCLUSION

Y-STR markers, located on the non-recombining region of the Y chromosome (NRY), have become an essential tool in forensic investigation due to their unique heredity properties and male specificity. A statistical approach should be carried out when the evidence’s Y-STR profile matches the profile of the suspect. Y-STR haplotype databases are important for understanding how common / rare evidence profile is in the relevant population. So it is necessary to question population database (19). In this study we determined the allele frequency and forensic statistical calculation for of 17 Y-STR loci in the Turkish population and it is now available for forensic purposes.

Turkey is a large country with its rich historical background and unique geographical position from East to West continental mainland. Turkey has been a crossroad for many civilizations: Hattusa, Hittites, Hellenic, Phrygian, Urartu, Lydia, Lycia, Ionians, Persians, Macedonians, Armenians, Romans, Byzantines, Seljuks and Ottomans and thus is an abundant genetic pool with a high ethnic heterogeneity. There are few remarkable Y STR population studies on Turkish population that are mainly focused on regional part of Turkey or have limited number of Y STR loci (17, 18, 20, -, 25). Previous studies showed that the Turkey population is closer to the European populations than Asia (21, 26, 27). The results of

the present study indicated the existence of the greatest genetic likelihood between Turkish population studies and confirmed a close genetic relationship between Turkish and Balkan and European populations.

It was also important to emphasize the intermediate alleles that could increase the discrimination power of DNA evidence that were observed in DYS458 locus (18.2 and 19.2) on our data set. The high frequency of intermediate alleles occurs at locus DYS458 that is most frequently found in Northern / Eastern African, Caucasian (28) and also in Turkish populations (Turkish Cypriots, Cukurova and Southeastern Anatolian). Although there are data suggesting that locus DYS458 may be affected by a higher mutation rate compared to other tetrameric Y-STR loci (29). This study presents a recent population of data of 17 YSTR loci in the homogenous distribution of the Turkish population for the forensic community.

ETHICAL APPROVAL

This study was approved by the Ethics Committee of the Istanbul University, Faculty of Medicine.

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