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DNA profiling for forensic identification in Bulgarian Turks

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Abstract

Forensic statistical parameters based on allelic frequencies of commonly used STRs (Short Tandem Repeats) were estimated for the Bulgarian Turks. The 6-dye GlobalFiler™ PCR amplification kit incorporates 21 autosomal STRs, providing reliable DNA typing results with enhanced the power of discrimination. Here, we analyzed the GlobalFiler™ STR loci in 150 unrelated individuals from Bulgaria Turks who was born in Bulgaria, lived there and then have settled in Turkey. A total of 329 alleles were observed ranging between 5 and 37 repeat units, and SE33 showed the greatest power of discrimination (40 alleles) in Bulgaria Turks population.
Blood and intraoral swab samples were collected from 150 Bulgarian Turks whose consent was obtained beforehand. DNA isolation was performed by using QIamp DNA Mini Isolation Kit. After quantification of the obtained DNA, the next step, PCR, was carried out using the GlobalFiler ™ STR kit. The DNA amplified after PCR was analyzed for profiling by running on ABI 3130 Genetic Analyzer. Allele frequencies and forensic statistical calculations were performed with Arlequin v 3.5.2.2 program. Allele frequencies were compared with Spain, Europe, Middle East, Africa, America, Central Asia, Japan, and Turkish populations. When the loci were examined in general, it was observed that the Turkish population had the closest frequency value to the group we studied, and the African and Central Asian populations had the farthest frequency value. The present study provides precise reference database for forensic applications and population genetic studies.

Keywords: Population genetics, allele frequency, STR, DNA analysis, globalfiler, forensic genetics, statistics, Bulgarian Turks

Introduction

DNA analysis, which is used routinely in basic sciences for various purposes, is used in the forensic sciences to present objective evidence in courts. In the forensic sciences, DNA analyzes are used in paternity and / or maternity detection cases, criminal cases, identification of missing persons and disaster victims [1,2]. DNA analysis consists of two steps: determination of DNA profile from biological samples and interpretation of evidence obtained from DNA. In order to interpret the evidence obtained from the DNA molecule correctly, it is necessary to know how often genetic signs are seen in the relevant population [3,4].

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In the non-coding DNA regions (introns), especially the regions formed by repeating the number of nucleotide sequences of different lengths repeatedly, have a high polymorphic value [5]. All people have the same sequences, but the number of repeats varies from person to person. These repeated sequences are hereditary characters suitable for Mendelian inheritance and are polymorphic due to the variation in the number of repeats. In DNA profiling, these repeated sequences are used [4,5]. STR loci have been preferred in forensic sciences since 1993 because of their small allele length and short analysis time, yielding results even in old and poorly preserved biological samples, enabling multiple analyzes and their suitability for automation [6-8].

The microsatellites are preferred in forensic sciences because the fact that they are very small fragments, they can be obtained from

damaged or even small amounts of DNA, the obtained results can be repeated, that the mutation rate is low. In addition to these advantages, the fact that many STR regions are analyzed at the same time and the discrimination power has increased has made it one of the leading techniques used in forensic sciences [9].

The GlobalFiler ™ STR kit was created by upgrading the 13 CODIS (Combined DNA Index System) loci to 20 loci and was made available to the laboratories . In fact, the GlobalFiler ™ STR kit can be called a system created by combining and developing the Identifiler® Kit and NGM SElect ™ Kits. The NGM SElect ™ kit includes the STR regions defined by the ESS (European Standard Set of Loci), which is used mostly in Europe, while the Identifiler® Kit is a FBI and consequently a CODIS-based system. Since the last locus of CODIS is located within the ESS, the GlobalFiler ™ STR kit has become a global system combining CODIS and ESS regions [10]. Because of these distinctive features, GlobalFiler ™ STR kit was used in our study. One of the most important features of the GlobalFiler ™ STR kit is the use of the combination of Y-chromosomal DYS391 and Y-indel, which compensates for the deficiencies in highly degraded samples in which DYS391 may fail, and thus greatly assist in the determination of highly degraded samples. The use of this feature helps to identify the victim of the disaster, to identify generations / genealogies and to clarify cases of sexual assault. In addition, the inclusion of the SE33 locus within the ESS, which has a high discriminatory power, improves the quality of the studies [1,11,12].

The allele frequency of STR loci varies in all populations. Therefore, genetic frequencies of genetic markers used in forensic sciences should be determined for each population and a database should be established. In the statistical calculations made in the evaluation of the results of DNA analysis, it is important to use the exact community database[12-14]. Located on a major migration route and has hosted many civilizations territory of Turkey has many racial and ethnic groups. Bulgarian Turkish immigrants, who have a considerable population especially in the Marmara Region, are an effective resource for population studies as they are included in the ethnic and cultural diversity of our country. In this regard, the purpose of our study is to identify the gene frequency in the specified locus for Bulgaria Turks who was born in Bulgaria, lived there and then have settled in Turkey by using GlobalFiler ™ STR kit, and to provide a use in forensic laboratories.

Materials and Methods

Sample Collection

This study was conducted with the permission of Istanbul University, Cerrahpasa Faculty of Medicine Clinical Research Ethics Committee (No:381416, dated 03.12.2015). Blood samples and buccal cell swabs were collected from 150 unrelated healthy Bulgarian Turk donors following informed consent.

105 blood samples were taken into 2 mL EDTA tubes and brought to the laboratory in accordance with the cold transport chain. 45 intraoral swab samples were taken from the mouth with sterile swabs and dried at room temperature. The collected samples were stored at -20 ° C until the time of study. All the studies were carried out using devices and equipments in the Forensic Molecular Genetic Laboratory of Istanbul University-Cerrahpaşa Forensic Medicine Institute.

Application of the Method

DNA isolation and quantification

DNA isolation was performed with silica-based QIAamp® DNA Mini Kit (Qiagen, Stanford, CA, USA) from 2mL blood samples taken into EDTA tubes and intraoral swab samples taken from sterile swabs [16]. The DNA quantification of the isolates was determined using the Qubit® fluorometer (Applied Biosystems) using the Quant-iT dsDNA HS (High Sensetive) Assay kit (Invitrogen, Paisley, Renfrewshire, UK) [17].

Amplification of the Samples by Polymerase Chain Reaction

The GlobalFiler ™ STR kit, which was stored at -20ºC, was used at this stage of the study. PCR analysis was performed on the GeneAmp 9700 (Thermo Fisher Scientific Company) according to the parameters set forth in Table 1. A negative control was used for each amplification to control contamination during the PCR stage and a positive control was used to determine if the method was working correctly [18].

Table 1. Cycling Conditions

PCR Steps	Temprature	Time
Initial incubation	95°C	1 minute
Denate	94°C	20 seconds
Anneal	59°C	90 seconds
Extend		
Final Extension	60°C	10 minutes

Running Samples in Capillary Electrophoresis

Electrophoresis of PCR products was performed by using 4 capillary ABI PRISM® 3130 Genetic Analyzer (Applied Biosystems Foster City, California) in Istanbul University-Cerrahpaşa Forensic Molecular Genetics Laboratory.

Analysis of Capillary Electrophoresis Data

After electrophoresis, visualization, typing and evaluation of the raw data obtained from the samples were performed using GeneMapper® ID-X software v1.4 (Applied Biosystems) analysis program [19]. Statistical calculations of raw data (allele frequencies, Fst values obtained for comparison with other countries) were analysed with Excel based PowerStats and Arlequin v 3.5.2.2. analysis programs [20,21]. Allele frequencies were compared with Turkish, Spanish, European, Middle Eastern, African, American, Central Asian and Japanese populations.

Results

Blood samples and buccal cell swabs were collected from 150 unrelated healthy Bulgarian Turk donors following informed consent. The amount of DNA measured by fluorimetric method of all samples used in our study ranged between 0.88 and> 600 ng / µl. The silica based QIAamp® DNA Mini Kit (Qiagen, Stanford, CA, USA) was used for DNA isolation. PCR amplifications were performed using the GlobalFiler™ STR according to the manufacturers' recommendations. Amplified products were analyzed using ABI 3130 sequencers and allele designations

were made corresponding to GlobalFiler Allelic Ladder (Applied Biosystems).

Data from each study were analyzed using GeneMapper ID-X 1.4 software (Life Technologies / Thermo Fisher Scientific, USA) with a global cut-off value of 20%. This 20% cut-off filter removes small peaks less than or equal to 20% of the height of the highest peak in a given location. Electroprograms were checked using GeneMapper ID-X and, if necessary, a noise peak (s) such as plus and / or minus stutter peaks were manually removed. The low allele peaks, which are automatically removed by the 20% cut-off filter, are manually added. Electrophoregrams of 24 loci are shown in Figure 1 and Figure 2.

The allele frequencies (Table 2) and forensic parameters (Table 4) were determined for 21 STR autosomal loci (D8S1179, D21S11, D7S820, CSF1PO, D3S1358, TH01, D13S317, D16S539, D2S1338, D19S433, VWA, TPOX, D18S51, D5S818, FGA, D10S1248, D1S1656, D22S1045, D2S441, D12S391 and SE33). The allele frenquencies obtained from using Arlequin v 3.5.2.2 are shown in Table 2, the expected & observed heterozygosity rates, and Hardy-Weinberg equilibrium results from using Arlequin v 3.5.2.2 are shown in Table 3 [21].

Table 2. Allele frequencies of 21 autosomal STR loci obtained for a population of 150 Bulgarian Turk individuals genotyped using the GlobalFiler kit.

Alel	D3S1358	vWa	D16S539	CSF1PO	TPOX	D8S1179	D21S11	D18S51	D2S441	D19S433	TH01	FGA	D22S1045	D5S818	D13S317	D7S820	SE33	D10S1248	D1S1656	D12S391	D2S1338
5					0.007																
6				0.007	0.027					0.007	0.237	0.003				0.007					
6.3																	0.003				
7				0.027	0.010	0.003		0.003			0.160			0.007		0.037	0.010				
7.3					0.007						0.003										
8			0.047	0.010	0.073	0.020		0.003		0.007	0.100	0.003	0.003	0.007	0.180	0.193	0.003			0.003	
9			0.153	0.073	0.293	0.007	0.003	0.007	0.017	0.003	0.187	0.003		0.037	0.133	0.087	0.013				
9.3							0.007				0.223	0.003									
10		0.003	0.100	0.293	0.293	0.087		0.017	0.170	0.013	0.043		0.007	0.050	0.043	0.300	0.007				0.003
11			0.337	0.293	0.253	0.030		0.023	0.260	0.007	0.007		0.157	0.330	0.283	0.227	0.007	0.017	0.117		
11.2										0.013	0.003										
12	0.003	0.003	0.200	0.253	0.037	0.157		0.093	0.120	0.100	0.003		0.017	0.347	0.283	0.097	0.007	0.040	0.063		0.013
12.2										0.103											
12.3								0.003						0.003							
13	0.003	0.007	0.147	0.037		0.347	0.003	0.130	0.070	0.170	0.007		0.007	0.187	0.060	0.023	0.020	0.217	0.073	0.007	0.010
13.2										0.197											
13.3											0.003										
14	0.037	0.083	0.013	0.007	0.007	0.167		0.173	0.297	0.140		0.003	0.070	0.017	0.013		0.050	0.343	0.080		
14.2										0.063	0.003										
14.3										0.010	0.003						0.003		0.003		
15	0.253	0.097	0.003			0.123		0.133	0.060	0.090		0.010	0.373	0.007	0.003	0.003	0.053	0.160	0.183	0.060	0.003
15.2										0.023											
15.3																			0.023		
16	0.377	0.230				0.047		0.173	0.003	0.043		0.003	0.247	0.007			0.070	0.163	0.167	0.010	0.057
16.3																			0.027	0.003	
17	0.150	0.313				0.013		0.087	0.003	0.007		0.010	0.083	0.003		0.017	0.117	0.047	0.070	0.123	0.197
17.3								0.003											0.100	0.020	
18	0.150	0.147						0.070				0.003	0.020				0.070	0.007	0.050	0.180	0.093
18.3																			0.013	0.013	0.003
19	0.023	0.073						0.037				0.070	0.003				0.087		0.003	0.130	0.077
19.3												0.007								0.030	0.007
20	0.003	0.033						0.017			0.003	0.137	0.007			0.003	0.087		0.013	0.080	0.133
20.2												0.003									
20.3																				0.003	
21								0.010				0.163	0.003				0.033		0.003	0.137	0.037
21.2												0.003					0.007				
21.3												0.020									
22		0.003						0.007				0.193					0.017	0.003	0.003	0.073	0.060
22.2																	0.033				
22.3												0.003									
23		0.003						0.003			0.007	0.123	0.003							0.073	0.087
23.2																0.007	0.027				
23.3										0.003		0.010									
24		0.003									0.003	0.090					0.007			0.040	0.113
24.2							0.003										0.027		0.003		
24.3												0.003									
25							0.003					0.077					0.007		0.003	0.010	0.103
25.2																	0.020				
26							0.200				0.003	0.030					0.013			0.003	0.003
26.2																	0.023				
26.3												0.003									
27							0.017					0.007					0.003				
27.2																	0.017				
28							0.130					0.007					0.017				
28.2																	0.037	0.003			
28.3							0.003														
29							0.313	0.003				0.003									
29.2							0.007										0.040				
29.3							0.007														
30							0.173	0.003													
30.2							0.033										0.013				
31							0.043										0.007				
31.2							0.053										0.020				
31.3							0.003														
32							0.020					0.003									
32.2							0.110										0.013				
33.2							0.030														
34																	0.003				
34.2							0.007														
35																	0.003				
36							0.007														
36.2							0.003										0.003				
37																	0.003				

Using Arlequin v3.5.1.2, the Hardy-Weinberg Complete Test was performed to determine whether any tribal specimen exhibited detectable deviations from equilibrium expectations. The test phase used 1,000,000 Markov chains and 100,000 dememorization steps. The significance level was accepted as p = 0.05 (Table 3).

Table 3. Statistics obtained from using Arlequin v 3.5.2.2

	Na	Ne	I	Ho	He	uHe	F	P value	S.D.	Garza Williamson index
D3S1358	9	3.952916	1.550688	0.76	0.747022	0.749521	-0.01737	0.39091	0.00067	1,00000
VWA	13	5.115962	1.849563	0.806667	0.804533	0.807224	-0.00265	0.04300	0.00440	0,86667
D16S539	8	4.745334	1.707324	0.773333	0.789267	0.791906	0.020188	0.21300	0.00010	1,00000
CSF1PA	9	4.099854	1.589642	0.733333	0.756089	0.758618	0.030096	0.04000	0.00410	1,00000
TPOX	10	3.132832	1.420714	0.653333	0.6808	0.683077	0.040345	0.61818	0.00033	0,13158
D8S1179	12	5.052773	1.878937	0.84	0.802089	0.804771	-0.04727	0.03100	0.00520	1,00000
D21S11	24	6.058158	2.222919	0.853333	0.834933	0.837726	-0.02204	0.98000	0.00023	0,33333
D18S51	21	8.431703	2.35221	0.906667	0.8814	0.884348	-0.02867	0.05300	0.00302	0,45652
D2S441	10	4.824702	1.741015	0.773333	0.792733	0.795385	0.024472	0.00909	0.01401	1,00000
D19S433	18	8.127145	2.309121	0.793333	0.876956	0.879889	0.095355	0.04200	0.00355	0,32727
TH01	18	5.603985	1.919979	0.793333	0.821556	0.824303	0.034352	0.17020	0.00142	0,42857
FGA	29	8.490566	2.438231	0.853333	0.882222	0.885173	0.032746	0.87080	0.00039	0,38667
D22S1045	14	4.211117	1.719472	0.806667	0.762533	0.765084	-0.05788	0.11090	0.00262	0,87500
D5S818	12	3.727943	1.557305	0.786667	0.731756	0.734203	-0.07504	0.09701	0.00394	0,32432
D13S317	8	4.621072	1.673358	0.733333	0.7836	0.786221	0.064148	0.95000	0.00022	1,00000
D7S820	12	5.053341	1.835132	0.713333	0.802111	0.804794	0.11068	0.01500	0.00713	0,22222
SE33	40	18.51852	3.21684	0.926667	0.946	0.949164	0.020437	0.03400	0.00495	0,57143
D10S1248	10	4.520342	1.699022	0.74	0.778778	0.781382	0.049793	0.01421	0.00756	0,17857
D1S1656	18	9.127789	2.402548	0.873333	0.890444	0.893423	0.019216	0.03300	0.00508	0,37255
D12S391	19	9.3187	2.429389	0.8	0.892689	0.895674	0.103831	0.05132	0.00391	0,38776
D2S1338	17	9.041591	2.36577	0.786667	0.8894	0.892375	0.115509	0.39091	0.00067	0,37778
TOTAL MEAN	15.7619	6.46554	1.994247	0.795556	0.81652	0.81925	0.024298			0,58287
TOTAL SE	1.725032	0.74485	0.095406	0.014204	0.014414	0.014462	0.01186			

Na: No. of Different Alleles, Ne: No. of Effective Alleles = 1 / (Sum pi^2), I: Shannon's Information Index = -1* Sum (pi* Ln (pi)), Ho: Observed Heterozygosity= No. of Hets / N, He: Expected Heterozygosity= 1- Sum pi^2, uHe: Unbiased Expected Heterozygosity = (2N / (2N-1))*He, F: Fixation Index= (He-Ho) / He= 1-(Ho / He), S.d: Standart Deviation
Where pi is the frequency of the ith allele fort he population & Sum pi^2 is the sum of the squared population allele frequencies.

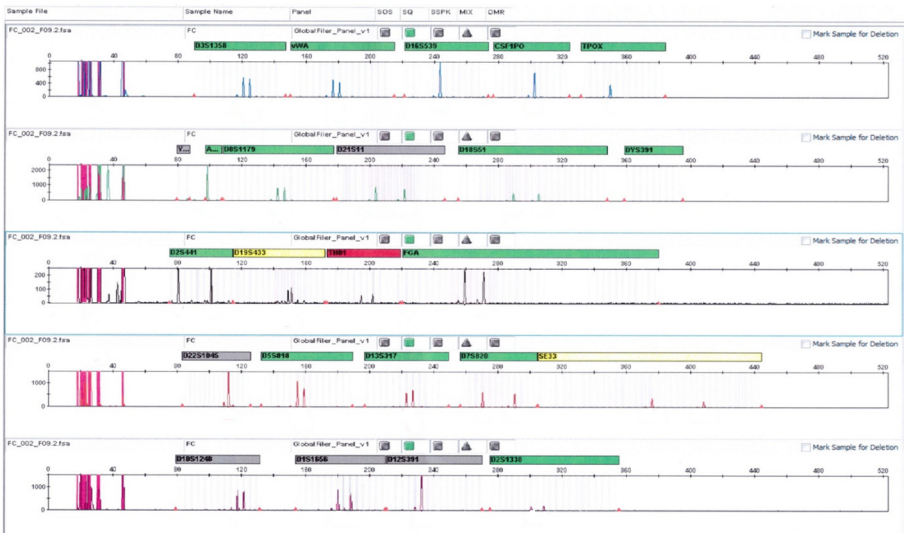

Figure 1. Female Electrophoregram

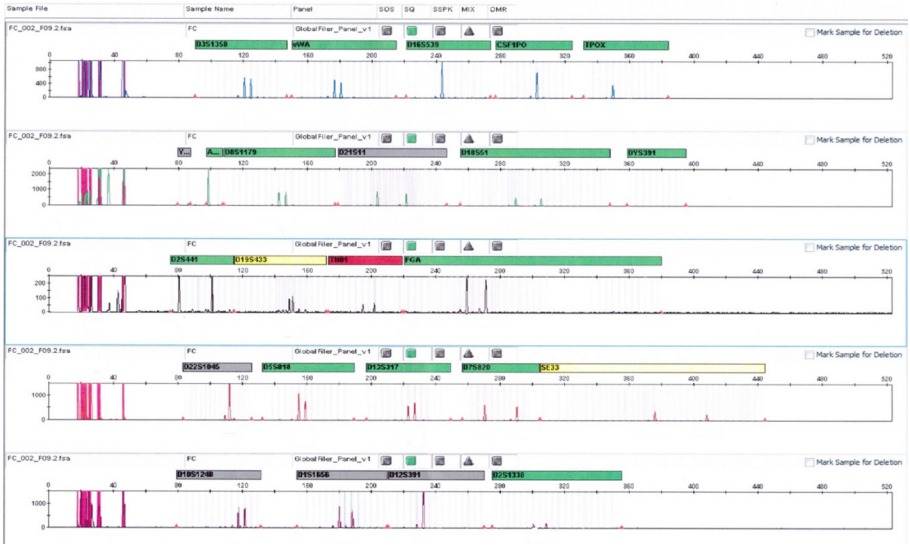

Figure 2. Male Electrophoregram

Probability of match (PM), power of discrimination (PD), polymorphism information content (PIC), power of exclusion (PE), typical paternity index (TPI) and Heterozygosity (He) were calculated roughly with the Excel based PowerStats (Table 4).

In the combined dataset, there were 9 loci that exhibited statistically significant deviations from HWE based on the exact test ($p<0.05$) which might be expected given population substructure. The marker SE33 exhibited the highest Hobs (0.92667), PD (0.9847), PIC (0.9418), PE (0.8471) and TPI values (6.6818) making it the most variable locus when compared to the other loci in this dataset. On the other hand, the less informative marker was TPOX (Table 4) .

Table 4. Forensic statistical parameters obtained from Power Stats

	PM	Expressed as 1 in ...	PD	PIC	PE	TPI	Homozygotes	Heterozygotes	Total Alleles
D3S1358	0.1188	8.4207	0.8812	0.7083	0.5270	2.0833	0.2400	0.7600	300
VWA	0.0716	13.9752	0.9284	0.7794	0.6115	2.5862	0.1933	0.8067	300
D16S539	0.0802	12.4723	0.9198	0.7604	0.5505	2.2059	0.2267	0.7733	300
CSF1PA	0.1089	9.1837	0.8911	0.7181	0.4817	1.8750	0.2667	0.7333	300
TPOX	0.1644	6.0811	0.8356	0.6302	0.3598	1.4423	0.3467	0.6533	300
D8S1179	0.0676	14.8026	0.9324	0.7790	0.6753	3.1250	0.1600	0.8400	300
D21S11	0.0720	13.8795	0.9280	0.8083	0.7031	3.4286	0.1458	0.8542	300
D18S51	0.0339	29.5276	0.9661	0.8699	0.8091	5.3571	0.0933	0.9067	300
D2S441	0.0868	11.5210	0.9132	0.7618	0.5478	2.1912	0.2282	0.7718	300
D19S433	0.0444	22.5350	0.9556	0.8646	0.5816	2.3871	0.2095	0.7905	300
THO1	0.0698	14.3312	0.9302	0.7973	0.5867	2.4194	0.2067	0.7933	300
FGA	0.0346	28.8700	0.9654	0.8693	0.6995	3.3864	0.1477	0.8523	300
D22S1045	0.1073	9.3206	0.8927	0.7299	0.6115	2.5862	0.1933	0.8067	300
D5S818	0.1297	7.7108	0.8703	0.6873	0.5745	2.3438	0.2133	0.7867	300
D13S317	0.0834	11.9936	0.9166	0.7510	0.4817	1.8750	0.2667	0.7333	300
D7S820	0.0747	13.3929	0.9253	0.7752	0.4492	1.7442	0.2867	0.7133	300
SE33	0.0153	65.2840	0.9847	0.9418	0.8471	6.6818	0.0748	0.9252	300
D10S1248	0.0856	11.6822	0.9144	0.7473	0.4928	1.9231	0.2600	0.7400	300
D1S1656	0.0291	34.4202	0.9709	0.8805	0.7396	3.9211	0.1275	0.8725	300
D12S391	0.0281	35.6356	0.9719	0.8825	0.5966	2.4833	0.2013	0.7987	300
D2S1338	0.0302	33.0882	0.9698	0.8795	0.5745	2.3438	0.2133	0.7867	300

Allelic frequencies were compared to previously published population data from Turkey (publication in preparation), as well as, others populations, namely, Spain, Middle East, Europe, Africa, America, Central Asia and Japan[22,23-26]. F-statics was performed Arlequin v3.5.1.2 and the results revealed that most of the molecular variation was due to variation between different populations. In the exact test of population differentiation, no major differences were observed with the populations of Turkey (FST=0,00014) and Middle East (FST=0,00027). Statistically significant differences were found with the populations of Japan (FST= -0,00001) and Spain (FST=0,00432) (Table 5).

Table 5. Computing conventional F-Statistics from haplotype frequencies

	BGTR	Turkey	Spain	Middleeast	Europe	Africa	America	Centralasia	Japan
BGTR	0.00000								
Turkey	0.00014	0.00000							
Spain	0.00432	0.00389	0.00000						
Middleeast	0.00027	-0.00003	0.00324	0.00000					
Europe	0.00051	0.00013	0.00375	0.00008	0.00000				
Africa	0.00090	0.00021	0.00383	0.00045	0.00049	0.00000			
America	0.00042	0.00007	0.00369	0.00010	0.00008	0.00027	0.00000		
Centralasia	0.00080	0.00078	0.00304	0.00051	0.00040	0.00136	0.00061	0.00000	
Japan	-0.00001	0.00010	0.00519	0.00017	0.00027	0.00058	0.00043	0.00064	0.00000

Discussion

The allele frequency of STR loci varies in all populations. For this reason, allele frequencies of genetic markers used in forensic sciences should be determined for each population and a database should be established. In the statistical calculations made in the evaluation of the results of DNA analysis, it is very important to use the accurate society database.

Combined with the need to get more information from the limited samples obtained and to finalize the procedures faster, studies have begun to analyze multiple STR systems together. To analyze multiple sites in the genome, multiple (Multiplex) PCR kits including the addition of multiple sets of PCR primers were able to implement this idea.

With this application depending on the principle that the primers used during PCR are labeled with fluorescent dyes and subsequently resolved spectrally, it is possible to amplify the labeled STR regions using different colored fluorescent dyes and to determine the size of the regions [14,27].

As a result of increasing 13 CODIS loci to 20 bymultiplying in the PCR following analyzing with electrophoresis and, a global system was established with the combination of CODIS and ESS regions and was made available to the use of the laboratories [14]. This system, called GlobalFilerTM STR kit, was first applied in the Forensic Molecular Genetic Laboratory of Istanbul University-Cerrahpaşa Forensic Medicine Institute, where the study was carried out, and optimized in our laboratory.

In our study, we preferred the GlobalFiler ™ STR kit to the Promega PowerPlex® 21 kit with similar features; Identifiler STR kit, which has been used in the forensic molecular genetics laboratory of Istanbul University Institute of Forensic Medicine for a while, has been used in routine analysis and project studies, since it is the same company, its primer indexes are similar and provide convenience in validation and optimization phase. Changes in primer sequences between commercial kits causes allele inconsistencies, allele losses and peak height ratio risk. This requires extra time to identify problems [13].

Conclusion

In conclusion, a Bulgarian Turks population database has been established for the 21 STR systems studied by using the GlobalFiler ™ PCR kit. The population data published in this paper can be freely used for any all state and privately funded forensic laboratory in the field of human identification and paternity testing. The obtained results support the idea that GlobalFiler ™ PCR kit is very informative for forensic purposes in Turkey Population and presents a very helpful tool for forensic reference samples analysis..

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Conflict of interests

The authors declare that they have no competing interests.

Financial Disclosure

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Ethical approval

This study was conducted with the permission of Istanbul University-Cerrahpaşa Medical Faculty Clinical Research Ethics Committee (No:381416, dated 03.12.2015). Blood samples and buccal cell swabs were collected from 150 unrelated healthy Bulgarian Turk donors following informed consent..

References

1. Hares, DR. Addendum to expanding the CODIS core loci in the United States. Forensic Science International. 2012;6: e135.

2. Vijg, J. Aging of the genome: the dual role of DNA in life and death. Oxford University Press. 2021;10(1)162-8 168.

3. Rudin N, Inman K. An introduction to forensic DNA analysis. CRC press, London, 2001.
4. Semikhodskii, A. Dealing with DNA evidence: a legal guide. Routledge-Cavendish, 2007.
5. Balding, D J. When can a DNA profile be regarded as unique?. Science & Justice. 1999;39:257-60.
6. Scientific Working Group on DNA Analysis Methods (SWGDAM). SWGDAM interpretation guidelines for autosomal STR typing by forensic DNA testing laboratories. 2010.
7. Aşıcıoğlu F. X-Kromozomal STR Polimorfizmi (DXS8377, DXS101, DXS6789, STRX-1, HUMHPRTB) Ve Türk Toplumundaki Alel Frekansları. Ph.D. thesis, Istanbul University, Istanbul, 2006.
8. Ezkurdia, I, Juan D, Rodriguez et al. Multiple evidence strands suggest that there may be as few as 19 000 human protein-coding genes. Human Molecular Genetics. 2014;23:5866-78.
9. Butler J M. Forensic DNA Typing. Biology, Technology, and Genetics of STR Markers. 2nd edition. Elsevier Academic Press, USA, 2005;87-117.
10. Martín, P, de Simón, L F, Luque G, et al. Improving DNA data exchange: validation studies on a single 6 dye STR kit with 24 loci. Forensic Science International: Genetics. 2014;13:68-78.
11. http://www.thermofisher.com/order/catalog/product/4476135 access date 28.07.2019
12. Hares D. Expanding the CODIS core loci in the United States. Forensic Science International: Genetics. 2012;6:52-4.
13. Yükseloğlu E H, Kara U. Hukukçular ve Genetikçiler için Temel Adli Genetik, 1. Baskı. Nobel Tıp Kitapevi, İstanbul, Türkiye. 2015.
14. Butler, J.M., Hill, C.R. and Coble, M.D. Variability of New STR Loci and Kits in US Population Groups. Promega Corporation Web site. http://www.promega.com/resources/articles/profiles-in-dna/2012/variability-of-new-str-loci-and-kits-in-uspopulation-groups access date 03.08.2019
15. Butler J M, Hill C R, Kline M C et al. The single most polymorphic STR Locus: SE33 performance in US populations. Forensic Sci. Int. Genet. Suppl. Ser. 2009;2:23-4.
16. https://www.qiagen.com/us/resources/download.aspx?id=62a200d6-faf4-469b-b50f-2b59cf738962&lang=en access date 20.07.2019
17. https://www.thermofisher.com/order/catalog/product/Q33120 access date 22.07.2019
18. http://www.thermofisher.com/order/catalog/product/4476135 access date 22.07.2019
19. http://tools.thermofisher.com/content/sfs/manuals/cms_041469.pdf. access date 22.07.2019
20. Crow J F. Hardy, Weinberg and language impediments. Genetics. 1999;152:821-5.
21. http://cmpg.unibe.ch/software/arlequin35/Arlequin35.html access date 12.06.2019
22. García O, Alonso J, Cano J A, et al. Population genetic data and concordance study for the kits Identifiler, NGM, PowerPlex ESX 17 System and Investigator ESSplex in Spain. Forensic Sci. Int. Genet. 2012;6:78-9.
23. http://spsmart.cesga.es access date 17.06.2019