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SHORT COMMUNICATION

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Population genetics of the 15 AmpF/STR Identifiler loci in Kosovo Albanians

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Abstract The 15 AmpF/STR Identifiler loci (D8S1179, D21S11, D7S820, CSF1PO, D3S1358, THO1, D13S317, D16S539, D2S1338, D19S433, VWA, TPOX, D18S51, D5S818 and FGA) were analyzed in a sample of 136 unrelated Albanian adults from Kosovo. The agreement with HWE was confirmed for all loci with the exception of TPOX (based on the exact test only). The combined power of discrimination (PD) and the combined power of exclusion (PE) for the 15 studied loci were 0.99999999999999997 and 0.9999995, respectively. According to the presented data, FGA proved to be the most informative marker. An interpopulation comparison between Kosovo Albanians and Croatians (as an example of a population from the Balkans) revealed significant differences in four out of nine loci.

Keywords STRs · AmpF/STR Identifiler · Population data · Kosovo Albanians

Introduction

Multilocus AmpFLP systems based on simultaneous detection of overlapping STR loci labeled with different fluorescent dyes allow more efficient forensic work since faster genotyping procedure is achieved [1, 2, 3, 4]. An example of a commercially available detection system is the AmpF/STR Identifiler PCR amplification kit which provides an easily reproducible and highly reliable method for typing 15 highly polymorphic STR loci: D8S1179, D21S11, D7S820, CSF1PO, D3S1358, TH01, D13S317, D16S539, D2S1338, D19S433, VWA, TPOX, D18S51, D5S818 and FGA.

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Materials and methods

Unrelated, autochthonous healthy adult Albanians from Kosovo (Prishtina, Vitia and Gjakova areas) participated in this study and gave their informed consent. Whole blood samples were obtained by venipuncture, collected into EDTA tubes and stored at –40°C. DNA was extracted from whole blood (10 ml) by the salting-out procedure [5] and quantified spectrophotometrically.

Multiplex PCR amplification was performed on approximately 1–3 ng of genomic DNA in a total reaction volume of 25 µl consisting of 9.5 µl AmpF/STR Identifiler PCR reaction mix, 0.5 µl of AmpliTaq Gold DNA polymerase, and 5.0 µl of AmpF/STR Identifiler primer set. Amplification was carried out in a 9600 Thermal Cycler (Applied Biosystems) performing 28 cycles under the following conditions (after an initial denaturation step of 11 min at 95°C): 94°C for 1 min, 59°C for 1 min, 72°C for 1 min.

Of the PCR product, 1 µl was combined with 12 µl formamide and 0.5 µl of size standard (GeneScan 500 LIZ). Electrophoresis, detection of PCR products and genotyping were carried out on the ABI PRISM 310 Genetic Analyzer (Applied Biosystems) using the ABI PRISM 310 data collection software and Genotyper 3.7 analysis software (Applied Biosystems).

Allele frequencies (since autosomal co-dominant) were computed using the gene counting method. The agreement with the Hardy-Weinberg expectations (HWE) of genotype frequencies was determined using the χ^2-test based on the number of observed and expected heterozygotes and the exact test based on the number of observed and expected genotypes [6], as implemented in a software developed at the Institute for Anthropological Research, Zagreb, Croatia. Forensic parameters were calculated using the software package PowerStats (Promega, Madison, WI).

Table 1 Observed allele frequencies and statistical parameters for forensic testing at the 15 STR loci in 136 unrelated Kosovo Albanians

Allele	D8S1179	D21S11	D7S820	CSF1PO	D3S1358	TH01	D13S317	D16S539	D2S1338	D19S433	VWA	TPOX	D18S51	D5S818	FGA
6						0.298						0.004			
7			0.026			0.129									
8	0.011		0.254			0.176	0.191	0.015				0.456			
9	0.022		0.081	0.022		0.202	0.107	0.129				0.081		0.018	
9.3						0.184									
10	0.055		0.232	0.173		0.011	0.077	0.074				0.074	0.007	0.085	
11	0.055		0.243	0.379			0.305	0.301			0.015	0.349	0.007	0.309	
12	0.103		0.140	0.327			0.239	0.283		0.096		0.037	0.037	0.375	
13	0.335		0.026	0.085			0.070	0.180		0.235			0.154	0.199	
13.2										0.022					
14	0.191			0.015	0.074		0.011	0.018		0.327	0.114		0.246	0.015	
14.2										0.077					
15	0.195				0.210				0.004	0.081	0.103		0.162		
15.2										0.033					
16	0.033				0.290				0.081	0.096	0.195		0.173		
16.2										0.029					
17					0.224				0.272		0.287		0.044		
17.2										0.004					
18					0.202				0.081		0.184		0.066		0.011
19									0.077		0.070		0.040		0.048
20									0.121		0.033		0.018		0.140
20.2															0.007
21													0.029		0.143
21.2															0.007
22									0.022				0.011		0.191
22.2															0.004
23									0.176						0.184
23.2															0.007
24									0.114				0.004		0.121
25									0.048						0.077
26		0.004							0.004						0.044
27		0.029													0.004
28		0.132													0.011
29		0.316													
29.2		0.026													
30		0.176													
30.2		0.033													
31		0.048													
31.2		0.125													
32		0.004													
32.2		0.063													
33		0.004													

Table 1 (continued)

Allele	D8S1179	D21S11	D7S820	CSF1PO	D3S1358	TH01	D13S317	D16S539	D2S1338	D19S433	VWA	TPOX	D18S51	D5S818	FGA
33.2		0.033													
34.2		0.004													
35.2		0.004													
H_{obs} [a]	0.787	0.816	0.838	0.743	0.765	0.853	0.772	0.809	0.868	0.779	0.860	0.794	0.882	0.728	0.831
H_{exp} [b]	0.795	0.826	0.796	0.712	0.775	0.789	0.791	0.774	0.845	0.804	0.816	0.777	0.849	0.717	0.864
χ^2-test	0.019	0.033	1.261	0.488	0.036	2.983	0.189	0.754	0.363	0.390	1.479	0.141	0.952	0.036	1.021
Exact test (*p*)	0.419	0.779	0.092	0.677	0.396	0.380	0.258	0.184	0.143	0.754	0.385	0.000	0.313	0.815	0.574
PM[c]	0.077	0.053	0.088	0.135	0.095	0.088	0.084	0.097	0.051	0.062	0.065	0.184	0.049	0.128	0.036
PD[d]	0.923	0.947	0.912	0.865	0.905	0.912	0.916	0.903	0.949	0.938	0.935	0.816	0.951	0.872	0.964
PE[e]	0.575	0.629	0.672	0.497	0.535	0.701	0.548	0.616	0.730	0.561	0.715	0.415	0.760	0.473	0.658
PIC[f]	0.77	0.81	0.76	0.66	0.74	0.76	0.76	0.74	0.83	0.78	0.79	0.60	0.83	0.67	0.85

[a] H_{obs} Observed heterozygosity.
[b] H_{exp} Expected heterozygosity.
[c] *PM* Probability of a match.
[d] *PD* Power of discrimination.
[e] *PE* Power of exclusion.
[f] *PIC* Polymorphism information content.

Table 2 Interpopulation comparison between Kosovo Albanian and Croatian populations

Locus	*P*-value
D3S1358	0.865
VWA	0.492
FGA	0.222
THO1	0.002
TPOX	0.011
CSF1PO	0.203
D5S818	0.570
D13S317	0.010
D7S820	0.047

Results and discussion

The observed allele frequencies and statistical parameters for forensic testing based on 15 AmpF*l*STR Identifiler loci in Kosovo Albanian population are summarized in Table 1. The agreement with Hardy-Weinberg expectations, tested by the exact test based on the number of observed and expected genotypes and the χ^2-test based on the number of observed and expected heterozygotes, was confirmed for all studied loci with the exception of TPOX (exact test). This departure is caused by the excess of genotypes 8-11 and 8-8. Considering the fact that only one test registered departure from HWE for one locus, this finding does not represent a basis for rejection of HWE. The combined power of discrimination (PD) and the combined power of exclusion (PE) for the 15 studied loci were 0.99999999999999997 and 0.9999995, respectively. Based on both measures of informativeness, heterozygosity and polymorphic information content, out of the 15 analyzed loci, FGA may be considered as the most informative.

An interpopulation comparison between Kosovo Albanians and Croatians (Table 2) (as an example of a population from the Balkans, see Table 3) revealed significant differences at the four loci TH01, TPOX, D13S317 and D7S820, whereas the remaining five loci examined showed uniform allelic frequencies.

In summary, based on presented allelic frequencies and statistical parameters for forensic testing for the AmpF*l*STR Identifiler detection system, the combination of these 15 STR loci presents a powerful strategy for individual identification and parentage analyses in the Albanian population from Kosovo.

Table 3 Allele frequencies of 9 STR loci in 102 unrelated adults from the Croatian mainland

Allele	D3S1358	VWA	FGA	TH01	TPOX	CSF1PO	D5S818	D13S317	D7S820
5									
6				23.04	0.49				
7				15.2					2.45
8				9.31	56.86			10.78	15.69
9				18.63	10.78	1.96	5.39	7.84	11.27
9.3				31.86					
10				1.96	4.9	21.57	7.35	4.41	29.9
11					25.98	31.37	34.31	41.67	22.55
12					0.98	37.75	36.27	21.57	14.22
13	0.49	0.49				5.88	16.18	10.29	3.92
14	9.31	7.84				1.47	0.49	3.43	
15	21.08	14.71							
16	26.96	17.65							
17	24.02	30.88							
18	17.16	21.57	0.98						
19	0.98	5.88	8.33						
20		0.98	17.65						
20.2			0.49						
21			17.65						
22			18.63						
22.2			1.47						
23			11.27						
23.2			1.47						
24			12.75						
24.2			0.49						
25			5.39						
26			2.94						
27			0.49						

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