

FOR THE RECORD

Damir Marjanovic,^{1,2} Ph.D.; Naris Pojskic,¹ Ph.D.; Jon Davoren,³ M.Sc.; Lejla Kovacevic,¹ B.Sc.; Adaleta Durmic,¹ M.Sc.; Narcisa Bakal,¹ B.Sc.; Katja Drobnic,⁴ Ph.D.; Dragan Primorac,⁵ Ph.D.; Vedrana Škaro,² B.Sc.; Kasim Bajrovic,¹ Ph.D.; and Rifat Hadziselimovic,¹ Ph.D.

Population Data at Two Short Tandem Repeat Loci D2S1338 and D19S433 in the Sample of Multinational Bosnia and Herzegovina Residents

POPULATION: We have analyzed the distribution of allele frequencies at two short tandem repeats loci (D2S1338 and D19S433) in a multinational sample of Bosnia and Herzegovina (B&H) residents. A total of 110 unrelated male and female individuals (Caucasians) from different regions of B&H were sampled for the analysis. We ensured that the sample reflected approximate proportional participation of the three main ethnic groups in the population of B&H (Bosniacs-Muslim [45%], Serbs [34%], Croats [21%]).

KEYWORDS: forensic science, DNA typing, short tandem repeats, D2S1338, D19S433, population data, Bosnia, Herzegovina

Some of the 110 tested subjects were involved in legal proceedings concerning paternity or other type of forensic testing, while the others were voluntary donors. Buccal swabs and blood samples (blood stains) were used as sources of DNA. The specimens were air-dried on the spot, placed in 1.5 mL tubes, and immediately transported to the Laboratory for Forensic Genetics at the Institute for Genetic Engineering and Biotechnology, Sarajevo, Bosnia and Herzegovina (B&H). Until DNA extraction, the samples were stored at -80°C . The Qiagen DnaeasyTM Tissue Kit was used for DNA extraction (1). An AmpFISTR[®] Identifiler[®] (ABI, Foster City, CA) was used to amplify simultaneously 15 short tandem repeat (STR) loci including D2S1338 and D19S433. Similar amounts of DNA were used in all PCR reactions. PCR amplification was carried out in a PE Gene Amp PCR System Thermal Cycler (ABI) according to the previously described reaction conditions (2). The total reaction volume was 12.5 μL . The amplified fragments were analyzed on an ABI PRISM 3100 instrument (ABI). GeneScan[®] 3.7.1 and Genotyper[®] 3.7 were applied in numerical allele designations. Deviation from Hardy–Weinberg equilibrium (3), observed, and expected heterozygosity (4) were calculated using Powermarker software (5), power of discrimination, and power of exclusion using Microsoft[®] Excel workbook template—POWERSTATS (6) and exact test of population differentiation (7) using Arlequin ver.

¹Institute for Genetic Engineering and Biotechnology, Kemalbegova 10, 71 000 Sarajevo, Bosnia and Herzegovina.

²Center for Integrative Genomics, Molecular Diagnostics, Cell and Gene Therapy, “Rudjer Boskovic” Institute, Zagreb, Croatia.

³International commission on missing persons, Alipasina 45a, 71.000 Sarajevo, Bosnia and Herzegovina.

⁴Forensic Laboratory and Research Center, Ministry of the Interior, Stefanova 2, 1501 Ljubljana, Slovenia.

⁵Laboratory for Clinical and Forensic Genetics, University Hospital Split, Spinciceva 1, 21 000 Split, Croatia.

3.01 (8). All results are shown in Table 1. The entire data are available at <http://www.ingeb.ba/edat/str/strbase.html>

TABLE 1—Bosnian and Herzegovinian allele frequencies for D2S1338 and D19S433 (N = 110).

Allele	D2S1338	D19S433
10	—	—
11	—	0.014
12	—	0.159
13	—	0.223
14	—	0.277
14.2	—	0.050
15	—	0.136
15.2	—	0.045
16	0.045	0.045
16.2	—	0.041
17	0.227	0.009
18	0.068	—
19	0.105	—
20	0.173	—
21	0.032	—
22	0.014	—
23	0.091	—
24	0.114	—
25	0.109	—
26	0.023	—
27	—	—
28	—	—
26	—	—
27	—	—
H(ob)	0.8545	0.8182
H(ex)	0.8621	0.8173
P	0.7943	0.1153
PD	0.964	0.935
PE	0.704	0.633

H(ob), observed heterozygosity; H(ex), expected heterozygosity; P, deviation from Hardy–Weinberg equilibrium; PD, power of discrimination; PE, power of exclusion.

In our previous population studies of B&H human population, we applied 15 STR loci included in the *PowerPlex 16[®] System* (9), 12 Y-chromosomal STRs loci incorporated in the *PowerPlex[®] Y System* (10), as well as 28 Y-chromosome NRY bi-allelic markers (11). Now, we have compared our data for these two loci with data obtained from geographically closer (neighboring) European populations. No statistically significant deviation ($p > 0.05$) from Hardy–Weinberg equilibrium was found for either of the observed loci. We compared B&H allele frequencies with the data from Slovenia (12), Serbia (Kosovo Albanians) (13), Serbia (Vojvodina) (14), and Macedonia (15). These exact tests of population differentiation show statistically significant differences (the significance level after Bonferroni's correction was $p = 0.01$) in allele frequencies at the D2S1338 locus between B&H and Serbia (Kosovo Albanians) samples. The same type of difference was noticed at the D19S433 locus between our's and the Serbia (Kosovo Albanians) and Macedonia samples.

References

1. Qiagen Companies. QIAGEN genomic DNA handbook. Wina: Qiagen, 2001.
2. Applied Biosystems. AmpFISTR[®] Identifiler[™] PCR amplification kit user's manual. Foster City, CA: Applied Biosystems, 2005.
3. Nei M. Molecular evolutionary genetics. New York: Columbia University Press, 1987.
4. Guo S, Thompson E. Performing the exact test of Hardy–Weinberg proportion for multiple alleles. *Biometrics* 1992;48:361–72.
5. Liu K, Muse S. PowerMarker: new genetic data analysis software—Version 3.0, 2004. Free program distributed by the authors over the internet from <http://www.powermarker.net>
6. Tereba A. Tools for analysis of population statistics. *Profiles in DNA* 1999;3:14–6.
7. Raymond M, Rousset F. An exact test for population differentiation. *Evolution* 1995;49:1280–3.
8. Excoffier L, Laval G, Schneider S. Arlequin ver. 3.0: an integrated software package for population genetics data analysis. *Evol Bioinform Online* 2005;1:47–50.
9. Marjanovic D, Bakal N, Pojskic N, Kapur L, Drobnic K, Primorac D, et al. Allele frequencies for 15 short tandem repeat loci in a representative sample of Bosnians and Herzegovinians. *Forensic Sci Int* 2006;156:79–81.
10. Marjanovic D, Bakal N, Pojskic N, Kapur L, Drobnic K, Primorac D, et al. Population data for the twelve Y-chromosome short tandem repeat loci from the sample of multinational population in Bosnia and Herzegovina. *J Forensic Sci* 2005;50(1):223–5.
11. Marjanovic D, Fomarina S, Montagna S, Primorac D, Hadziselimovic R, Vidovic S, et al. The peopling of modern Bosnia–Herzegovina: Y-chromosome haplogroups in the three main ethnic groups. *Ann Hum Genet* 2005;69(6):757–64.
12. Drobnic K, Regent A, Budowle B. STR data for the AmpFISTR SGM plus from Slovenia. *Forensic Sci Int* 2001;115:107–9.
13. Kubat M, Skavic J, Behluli I, Nuraj B, Bekteshi T, Behluli M, et al. Population genetics of the 15 AmpF/STR identifier loci in Kosovo Albanians. *Int J Legal Med* 2004;118:115–8.
14. Veselinovic I, Kubat M, Furac I, Skavic J, Martinovic-Klaric I, Tasic M. Allele frequencies of 15 AmpF/STR Identifier loci in the population of Vojvodina Province, Serbia and Montenegro. *Int J Legal Med* 2004;118:184–6.
15. Jakovski Z, Nikolova K, Furac I, Masic M, Janeska B, Kubat M. Allele frequencies for 15 STR loci in a population from the Republic of Macedonia. *Int J Legal Med* 2006;120:53–5.

Additional information and reprint requests:

Damir Marjanovic, Ph.D.

Institute for Genetic Engineering and Biotechnology (INGEB)

Kemalbegova 10

71000 Sarajevo

Bosnia and Herzegovina

E-mail: damir.marjanovic@ingeb.ba