

## Preliminary population study at fifteen autosomal and twelve Y-chromosome short tandem repeat loci in the representative sample of multinational Bosnia and Herzegovina residents

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**Abstract.** We have selected 100 unrelated healthy individuals born in the Bosnia and Herzegovina, from three main ethnical groups for our study. For the autosomal STR analysis we choose 100 male and female individuals (Bosniaks=44%, Serbs=31%, Croats=17% and others=8%). Additionally, 100 males, voluntary donors [ Bosniaks: Muslim (35), Serbs (31), Croats (34)], have been investigated for the Y-STR analysis. Buccal swabs and blood samples (blood spots) have been used as the DNA source and Qiagen Dnaeasy™ Tissue Kit was employed for DNA extraction. The *PowerPlex 16® System* and *PowerPlex® Y System* (Promega Corp., Madison, WI, USA) have been used to amplify 15 autosomal and 12 Y-STR loci. Amplification was carried out as described previously. Electrophoresis of the amplification products was performed on an ABI PRISM 377 genetic analyzer. We have compared three Bosniak ethnic group according to Y-STR data. Also, we have compared entire B&H autosomal STR data with data obtained from geographically closer (neighboring) European populations. These results will be used as guiding principle for our upcoming studies. © 2005 Elsevier B.V. All rights reserved.

**Keywords:** STR; Y-chromosome; Population data; Genetic distance; Bosniak ethnic group

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1. Introduction

According to archaeological and genetic data the Balkan territory was continuously settled since the Paleolithic and, during the last Glacial Maximum (LGM), has been an important refugee area [1]. Previous studies, based on NRY (non-recombining region of the Y-chromosome) bi-allelic markers, describe that three main groups of Bosnia–Herzegovina, in spite of some quantitative differences, share a large fraction of the same ancient gene pool [2]. In DNA analysis of forensic biological evidence, we are using 15 STR loci included in the *PowerPlex 16® System*, as well as twelve Y-chromosomal short tandem repeats loci incorporated in the *PowerPlex® Y System*. Autosomal and Y-STR allele frequencies from representative sample of B&H population for daily use in DNA forensic and paternity testing, as well as some other parameters, were calculated in our previous studies [3,4]. Now, we have performed some preliminary tests of inter- and intra-population diversity based on observed STR loci.

2. Material and methods

For the autosomal STR analysis we chose 100 male and female unrelated individuals (Bosniaks=44%, Serbs=31%, Croats=17% and others=8%), but for the Y-STR analysis 100 males, voluntary donors, have been tested. Buccal swabs and blood samples (blood spots) have been used as the DNA source. DNA extraction was performed by usage of Qiagen Dnaeasy™ Tissue [5]. 15 autosomal STR loci (D3S1358, TH01, D21S11, D18S51, Penta E, D5S818, D13S317, D7S820, D16S539, CSF1PO, Penta D, vWA, D8S1179, TPOX, and FGA) included in the *PowerPlex 16® System*, as well as twelve Y-chromosomal short tandem repeats loci (DYS19, DYS385a, DYS385b, DYS389I, DYS389II, DYS390, DYS391, DYS392, DYS393, DYS437, DYS438 and DYS439) incorporated in the *PowerPlex® Y System*, both manufactured by Promega Corp., Madison, WI, USA, were chosen to be amplified by PCR. Amplification was carried out as described previously [6]. The total volume of each reaction was 10 µl. The PCR amplifications have been carried out in PE Gene Amp PCR System Thermal Cycler (ABI, Foster City, CA, USA) according to the manufacturer’s recommendations. Electrophoresis of the amplification products was performed on an ABI PRISM 377 genetic analyzer (ABI), using 5% bis-acrilamide gel (Long Ranger® Single® Packs). Raw data have been compiled and analyzed using the accessory software: ABI PRISM® Data Collection Software and Gene Scan®. Numerical allele designations of the profiles were obtained by processing with Powertyper16 and PowertyperY Macro. Exact test of population differentiation and standard genetic distance [7] was calculated within Arlequin version 2000 and *DISPAN (Genetic Distance and Phylogenetic Analysis)*.

3. Results and discussion

81 different Y-STR haplotypes (from total number of 100 obtained) were detected: 69 of them were unique, 7 appeared twice, 4 appeared three and only 1 five times. Six of twelve not singleton

Table 1  
Genetic distances between B&H ethnic groups based on Y-STR results

Inconsistencies	Bosniaks	Croats	Serbs
Bosniaks	////////	////////	////////
Croats	0.0912	////////	////////
Serbs	0.0282	0.1114	////////

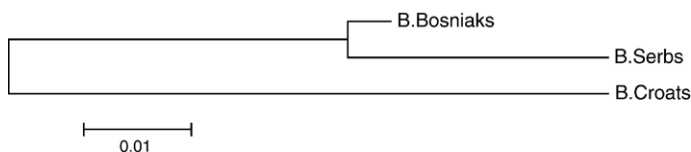


Fig. 1. Dendrogram constructed using Reynolds standard genetic distance calculated between observed population and the neighbor-joining clustering method.

haplotypes were shared by different population groups: two of them by Croats and Bosniaks, two by Bosniaks and Serbs and the last two by Serbs and Croats, thus testifying almost certainly recent gene flows between groups.

Genetic distances between these populations, based on Y-STR results, are given in Table 1. The dendrogram, constructed by using the neighbor-joining method (Fig. 1) and based on the results of the genetic distance analysis using pooled data, positioned Bosniaks in position between Croats and Serbs.

The calculated genetic distance and dendrogram indicate that, according these results, the Croatian population is relatively distanced from Bosniak and Serb populations. Similar results have been obtained in previous studies on bi-allelic [2]. As well as it was emphasized in previous studies, these results should be taken as preliminary results that have to be more investigated with larger and more geographically structured sample of all ethnic groups. Only in this manner could it be recognized if this differentiation is based on ethnical or geographical background.

Also, we have previously compared autosomal STR B&H data with data obtained from geographically closer (neighboring) European populations. No significant difference ( $P < 0.05$ ) is found at any individual locus except at FGA locus in comparative analysis of B&H and pooled Austrian data [4].

Joint results of this study are going to be used as guidelines in an additional investigation of genetic relationship between recent B&H and neighboring human populations, originated in this and our earlier studies on Y chromosome bi-allelic markers.

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