International Congress Series 1288 (2006) 307–309

www.ics-elsevier.com

Polymorphism of four X-chromosomal STRs in a religious minority of Old Believers residing in northeastern Poland

W. Pepinski [a,*], A. Niemcunowicz-Janica [a], M. Skawronska [a], J.R. Janica [b], E. Koc-Zorawska [a], J. Janica [a], I. Soltyszewski [c]

[a] *Department of Forensic Medicine, Medical University of Bialystok, Poland*
[b] *Department of Radiology, Medical University of Bialystok, Poland*
[c] *Central Forensic Laboratory of Police, Warsaw, Poland*

Abstract. Allele frequencies for four X-chromosomal STR were determined in a population sample of 210 unrelated volunteers from the north-eastern Poland by multiplex PCR and subsequent automated fluorescent detection (ABI 310) using a commercially available multiplex PCR kit (Mentype Argus X-UL). Kinship tests revealed a typical X-linked inheritance with no mutation. The analysed quadruplex is a potential extension to a battery of autosomal systems in forensic applications, especially in the investigation of kinship analysis and deficiency cases. © 2005 Published by Elsevier B.V.

Keywords: X-chromosome STRs; Old Believers; Northeastern Poland; Forensic genetics; Population genetics

1. Introduction

Old Believers are a fraction of the Russian Orthodox Church who came into existence as a result of schism introduced in 1653–1666 by Patriarch Nikon in opposition to the Russian Church Reform, adopting the liturgy and practices of the Greek Church. The Old Believers who resisted the Reform were condemned and declared dissidents; in 1667, they were separated from the Russian Orthodox Church and severely persecuted under the tsars, sought shelter in the most remote corners of Russian Siberia as well as abroad, including USA, Lithuania and Poland. In the 19th century, they moved to Suwalki Region (NE Poland), where they founded several villages and have struggled to maintain their religious

* Corresponding author. Tel.: +48 85 7485948; fax: +48 85 7485985.
E-mail address: pepinski@amb.edu.pl (W. Pepinski).

0531-5131/ © 2005 Published by Elsevier B.V.
doi:10.1016/j.ics.2005.08.031

identity and traditional ways of life in almost complete isolation for centuries. Buccal swabs were collected from 210 unrelated volunteers (140 males and 70 females). In order to investigate mutation rates, a population study was performed based on 52 casework family trios (38 male, 28 female offspring), with autosomally confirmed paternity at the probability level of 99.999% or higher.

2. Materials and methods

DNA was extracted using the Chelex 100 and proteinase K protocol [1]. 0.5–1 ng target DNA was amplified using a commercial kit Mentype Argus X-UL (Biotype AG, Germany) that allows a single-tube co-amplification and detection of four loci: DX8378, DX7132, HPRTB and DX7423. Electrophoresis and typing were performed in the ABI 310 Genetic Analyzer (Applera, USA). Reference sequenced ladder in combination with the Mentype Argus X-UL Template File included in the kit were used for genotype classification. For each locus, allele frequencies were calculated separately for males and females. Comparison of allele frequencies in the both groups was performed by the exact test of a RxC contingency table analysis [2]. Possible divergence from HWE was tested using the exact test [3] included in the GDA software v1.2 [4]. The following statistical parameters were calculated: observed and expected heterozygosity (Ho, He) [5], polymorphism information content (PIC) [6], mean exclusion chance (MEC) [7], expected probability of exclusion (PE) and discrimination power in males (DP_M) and in females (DP_F) [8]. Comparison of interpopulation allele frequency distributions was performed by means of RxC contingency test (G. Carmody, Ottawa, Canada).

Table 1
Allele distribution and biostatistical parameters for 4 X-chromosomal STR markers in a population sample of religious minority of old believers

Allele	DX8378			HPRTB			DX7423			DX7132		
	M	F	T	M	F	T	M	F	T	M	F	T
9	0.0214	0.0210	0.0214	0.0429	0.0500	0.0464						
10	0.3143	0.2720	0.2929	0.0071	0.0000	0.0036						
11	0.3357	0.3500	0.3429	0.0929	0.0857	0.0893				0.0071	0.0071	0.0071
12	0.2643	0.2930	0.2786	0.4357	0.4571	0.4464	0.0071	0.0000	0.0036	0.1214	0.1429	0.1321
13	0.0571	0.0640	0.0607	0.2000	0.1714	0.1857	0.1429	0.1640	0.1536	0.2929	0.2786	0.2857
14	0.0071	0.0000	0.0036	0.1429	0.1214	0.1321	0.2929	0.3210	0.3071	0.3071	0.3000	0.3036
15				0.0643	0.0929	0.0786	0.3286	0.2790	0.3036	0.1929	0.2000	0.1964
16				0.0143	0.0214	0.0179	0.2000	0.2000	0.2000	0.0643	0.0643	0.0643
17							0.0286	0.0360	0.0321	0.0143	0.0071	0.0107
P	0.1344			0.0440			0.0001			0.6466		
Ho	0.6429			0.7571			0.8286			0.8000		
He	0.7186			0.7332			0.7562			0.7733		
PIC	0.63			0.70			0.71			0.73		
MEC	0.6609			0.7017			0.7054			0.7280		
PE	0.5171			0.5632			0.5663			0.5932		
DP_M	0.7149			0.7349			0.7450			0.7636		
DP_F	0.8633			0.8969			0.8949			0.9083		

M: males, F: females, T: pooled, *P*: probability value (HWE analysis), Ho: observed heterozygosity, He: expected heterozygosity, PIC: polymorphism information content, MEC: mean exclusion chance, PE: expected probability of exclusion (father/daughter duos), DP_M: discrimination power in males, DP_F: discrimination power in females.

3. Results

Allele distribution and biostatistical parameters for X-chromosomal STR markers are shown in Table 1. The genotype distributions among the females conformed with HWE for all analysed loci ($0.05 < P$) except for DX7423 and HPRTB ($P = 0.0001$ and 0.0440, respectively). No significant differences were observed between allele distributions in males and females ($0.4230 < P < 0.9940$); therefore, the two groups were pooled into single-frequency distributions for respective loci. No mutation was detected at any of the four loci based on 38 maternal transfers and 28 paternal transfers.

4. Discussion

The markers are all tetranucleotide repeats and belong to the X chromosome coupling groups 1, 2, 3, and 4, respectively [9]. A pairwise comparison using the exact test disequilibrium analysis yielded no indication of allelic dependence ($0.2428 < P < 0.7059$); hence, the loci could be treated independently from each other. For the quadruplex evaluated, the combined MEC was 0.9919, and the combined DP was 0.9954 and 0.9999 (for males and females, respectively). A pairwise testing for heterogeneity using the RxC contingency table exact tests for population differentiation revealed statistical differences between the religious minority of Old Believers and the autochthonous Polish population at HPRTB and DX7132 ($P = 0.0010$ and 0.0110, respectively) [10]. Furthermore, at DX8378 the studied sample lacked allele 8 which was displayed in the Polish data set. On the other hand, at HPRTB and DX7423 alleles 10 and 12 were found, respectively, which were absent in autochthonous Poles.

Acknowledgements

This project was supported by the Medical University of Bialystok Grant No. 3-21589.

References

[1] P. Wiegand, T. Bajanowski, B. Brinkmann, PCR typing of debris from fingernails, Int. J. Leg. Med. 106 (1993) 81–84.
[2] D.A. Roff, P. Bentzen, The statistical analysis of mitochondrial DNA polymorphisms: chi 2 and the problem of small samples, Mol. Biol. Evol. 6 (1989) 539–545.
[3] S.W. Guo, E.A. Thompson, Performing the exact test of Hardy–Weinberg proportion for multiple alleles, Biometrics 48 (1992) 361–372.
[4] P.O. Lewis, D. Zaykin, Genetic Data Analysis: Computer program for the analysis of allelic data. Version 1.0 (d16c). Free program distributed by the authors over the internet from http://lewis.eeb.uconn.edu/lewishome/software.html.
[5] M. Nei, A.K. Roychoudhury, Sampling variances of heterozygosity and genetic distance, Genetics 76 (1974) 379–390.
[6] D. Botstein, et al., Construction of a genetic linkage map in man using restriction fragment length polymorphism, Am. J. Hum. Genet. 32 (1980) 314–331.
[7] T. Kishida, Y. Tamaki, Japanese population data on X-chromosomal STR locus AR, Nippon Hoigaku Zasshi 51 (1997) 376–379.
[8] D. Desmarais, et al., Development of a highly polymorphic STR marker for identity testing purposes at the human androgen receptor gene (HUMARA), J. Forensic Sci. 43 (1998) 1046–1049.
[9] R. Szibor, et al., Use of X-linked markers for forensic purposes, Int. J. Leg. Med. 117 (2003) 67–74.
[10] W. Pepinski, et al., Polymorphism of four X-chromosomal STRs in a Polish population sample, Forensic Sci. Int. 151 (2005) 93–95.