HLA-Antigens in the Romanian Population

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DURING the first millennium B.C., the present territory of Romania was invaded by Thracians (12th century B.C.), Illyrians (750–450 B.C.), and Getae and Celts (300 B.C.). Dacians, the northern Thracians, are known from Greek sources of the 4th and 3rd centuries B.C. In the period between 200 and 31 B.C., the Dacian state extended between the Tisza River and the lower Danube. In 85 A.D., the Roman emperor Domimitian's expedition against the Dacians consolidated his position north of the Danube. The complete conquest of Dacia followed two vigorous campaigns by the emperor Trajan (in 103 and 105–106); the process of Romanization was swift. In Romania, Latin completely supplanted the Dacian and Thracian languages. In 214, Dacia was conquered by the Goths. The devastation of the Gothic empire by the Huns, 200 years later, followed by that of the Bulgars at the end of the 5th century and that of the Avars in the late 6th century, prepared the ground for a wide Slavic dissemination from their homeland north of the Carpathians in the Ukraine. During the 10th–13th centuries, small Romanian principalities of Russian Orthodox religion and Latin language were formed. After a brief period of unification and independence, the Southern (Valachia) and Eastern (Moldavia) principalities fell under Turkish occupation (beginning of the 17th century), while the central state of Transylvania was conquered by Hungary and then by Austria (1696). It was only in 1856 that Valachia and Moldavia regained their independence, forming the United Romanian State, to which the Romanian provinces of Bucovina and Basarabia (Northeast), Transylvania (Central), and Banat (West) proclaimed their adherence in 1918. Basarabia and part of Bucovina were lost to the Russians after World War II.[1]

The work reported in this article represents an analysis of the genetic structure of the Romanian population with respect to the HLA system.

MATERIALS AND METHODS

Population Sample

Seventy-five unrelated Romanians were included in our study. Of these, 24 were HLA genotyped by studying their immediate relatives (not included in this survey). All individuals typed are native Christian Romanians, who have emigrated to the United States after 1948.

Typing Methods

HLA-A,B typing was performed by the microlymphocytotoxicity method using a set of 180 well defined antisera originating from the NIH, from our own laboratory, and from national and international exchanges.

Statistical Analysis

The gene frequencies (p) were calculated by the formula: $p = 1 - \sqrt{1 - f}$, where f is the frequency of the corresponding antigen. Gametic association between HLA-A and HLA-B antigens was measured by the formula of Mattiuz et al:[2]

$$D = \sqrt{\frac{d}{n}} - \sqrt{\frac{(b + d)(c + d)}{n^2}},$$

where n is the size of the sample, and a, b, c, d represent the numbers of $++$, $+-$, $-+$, $--$ phenotypes, respectively.

Haplotype frequencies h_{ij} were calculated as: $h_{ij} = P_iP_j + D_{ij}$, where P_i is the gene frequency of the i allele at the A locus and P_j is the frequency of the j allele at the B locus, while D_{ij} is the corresponding gametic association. The statistical significance of D value was calculated from the phenotype frequencies by the 2×2 χ^2, and the variance of the estimate of D was determined by the formula of Bodmer and Bodmer.[3]

Genetic distance (F) between Romanian and other

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Supported in part by National Institute of Arthritis and Infectious Diseases Contract NO 1-AI-82552.

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0041–1345/79/1104–0014$01.00/0

Table 1. Antigen and Gene Frequency in 75 Unrelated Romanians

HLA-A	Gene Frequency	Antigen Frequency	SD	HLA-B	Gene Frequency	Antigen Frequency	SD
A1	.113	.213	.027	B5	.113	−213	.027
A2	.270	.467	.042	B7	.034	−066	.015
A28	.048	.093	.017	B8	.076	−147	.022
A3	.136	.253	.030	B12	.120	−227	.028
Aw23	.010	.040	.012	B13	.027	−053	.013
Aw24	.113	.213	.027	B14	.013	−027	.009
A25	.041	.080	.016	B15	.041	−080	.016
A26	.076	.146	.023	B16	.062	−120	.020
A11	.069	.133	.021	B17	.027	−053	.013
A29	.020	.040	.011	B18	.083	−160	.024
Aw30	.007	.013	.007	Bw21	.027	−053	.013
Aw31	.020	.040	.011	Bw22	.041	−080	.016
Aw32	.034	.067	.015	B27	.048	−093	.018
Aw33	.013	.027	.009	Bw35	.120	−227	.028
				B40	.083	−160	.024
				Bw41	.041	−080	.016

populations was calculated by the formula:[3] $F = 4(1 - \text{Cos}\,\theta)/(k - 1)$, with

$$\text{Cos}\,\theta = \sum_{i=1}^{k} \sqrt{P_{i1} - P_{i2}},$$

where k is the number of alleles common to the two populations and P_{i1} and P_{i2} are the respective allele frequencies.

Additionally, the "angular" distance between two populations was calculated as:[4] $d = \sqrt{1 - \text{Cos}\,\theta}$.

RESULTS AND DISCUSSION

The frequencies of HLA-A and B antigens and genes are shown in Table 1. Compared to Northern Europeans,[5] Romanians have a significant increase in the frequency of A10, B5, B16, and B18 and a decrease in the frequencies of B7, B14, and B15. A number of significant gametic associations between antigens at the first and second locus were found. Table 2 lists these haplotype frequencies and the corresponding D values. Like most other European populations, highly significant gametic associations have been found for A1–B8, A2–B12, A26–B16. The haplotype A25–B18, which has high D in Germans and in North American whites[4] is also frequently found in Romanians. Linkage disequilibria between A3–B27 and A31–B35 seem to be peculiar to the Romanian population.

Table 3 shows the genetic distance between the Romanian and other European populations, such as British,[6] Danish,[7] Belgian,[8] French,[6] Spanish,[9] Italian,[6] Yugoslavian,[10]

Table 2. Significant Linkage Disequilibria (Δ) and Haplotype Frequencies (HF) of HLA-A and HLA-B Antigens in Romanians

	Δ	SE	HF	Fisher's p
A1,B8	.0370	.0078	.0456	1.15×10^{-3}
A2,B12	.0602	.0264	.0927	8.64×10^{-4}
A3,B27	.0257	.0053	.0322	9.81×10^{-3}
A25,B18	.0228	.0032	.0262	5.05×10^{-3}
A26,B16	.0353	.0041	.0400	1.59×10^{-4}
Aw31,B35	.0118	.0023	.0202	1.01×10^{-2}

Table 3. Genetic Distance Between Romanians and Other European Populations

	F			d	
	HLA-A	HLA-B	Average	HLA-A	HLA-B
Polish	.0035	.0067	.0053	.1120	.1644
French	.0047	.0061	.0055	.1285	.1569
Bulgarian	.0068	.0060	.0064	.1307	.1606
Italian	.0094	.0079	.0086	.1815	.1783
Yugoslavian	.0102	.0097	.0099	.1894	.1969
Russian	.0051	.0146	.0101	.1338	.2346
Czechoslovakian	.0078	.0142	.0113	.1659	.2390
British	.0059	.0166	.0115	.1437	.2501
Danish	.0074	.0152	.0119	.1492	.2448
Hungarian	.0087	.0143	.0119	.1690	.2395
Spanish	.0136	.0153	.0146	.2026	.2401
Greek	.0156	.0159	.0156	.2075	.2448
Romanian Jews	.0283	.0178	.0223	.2792	.2587
Belgian	.0457	.0139	.0250	.3024	.2286

Czechoslovakian,[11] Polish,[12] Hungarian,[13] Russian,[14] Bulgarian,[15] Greek,[16] and Romanian Jews.[17] While the distance to some Latin-speaking populations (French and Italian) is relatively small, the distance to others (Spanish and particularly Belgian) is significantly higher.

The common Roman ancestry cannot explain these differences, since Romania's colonization by the Romans occurred at the time when Roman expansion in Europe reached its culmination—comprising not only the Iberian (Spain, Portugal), Italic and Balcanic peninsulas (Yugoslavia, Bulgaria, Albania, Greece), but also the present territories of France, Germany, Belgium, Holland, Britain, Austria, Czechoslovakia, and Hungary (14–138 AD). The same holds true for the more recent Gothic invasions (4th–6th centuries), which involved about the same territories. It is more likely that the different rates of Slavic admixture have left their stigma on the genetic distance. Thus, the barbarian invasions of the 5th century (Huns, Bulgars, Avars) have left the Balkan Peninsula a sparsely populated wasteland destined to attract a wide Slavic dissemination from the upper Vistula in central Poland (where they have settled during the great prehistoric migration) toward southeastern Europe. This is consistent with the relatively small genetic distance between Romanians, Poles, and Bulgarians.

The Roman-German-Slavic admixture might thus explain why Romanians appear genetically equidistant from the extremes of Western Europeans (Britain) and Eastern Europeans (Russia).

REFERENCES

1. Stier HE, Kirsten E, Wuhr W: Westermann's Grosser Atlas Zur Weltgeschichte. Berlin, 1956
2. Mattiuz PL, Inde D, Piazza A, et al: In Terasaki PI (ed): Histocompatibility Testing 1970. Copenhagen, Munksgaard, 1970, pp 193–206
3. Bodmer J, Bodmer WF: Am J Hum Genet 22:396–411, 1970
4. Pickbourne P, Piazza A, Bodmer WF: In Bodmer WF, Batchelor JR, Bodmer JG, et al (eds): Histocompatibility Testing 1977. Copenhagen, Munksgaard, 1978, pp 259–278
5. Fog T, Schuller E, Jersild C, et al: In Dausset J, Svejgaard A (eds): HLA and Disease. 1977, pp 108–125
6. Bodmer J: In Kissmeyer-Nielsen F (ed): Histocompatibility Testing 1975. Copenhagen, Munksgaard, 1975, p 21
7. Hansen HE, OlsenLarsen S, Ryder LP, et al: Tissue Antigens 13:143, 1979
8. deBruyere M, Kulakowski S, Malchaire J, et al: Tissue Antigens 10:399, 1977
9. Moreno ME, Kreisler JM: Tissue Antigens 9:105, 1977
10. Kastelan A: In Bodmer WF, Batchelor JR, Bodmer JG, et al (eds): Histocompatibility Testing 1977. Copenhagen, Munksgaard, 1978, p 448
11. Gligorovic V, Susakovic N, Stolic C, et al: In Bodmer WF, Batchelor JR, Bodmer JG, et al (eds): Histocompatibility Testing 1977. Copenhagen, Munksgaard, 1978, p 423
12. Matej H, Roszak E, Kunicka J: Tissue Antigens 10:229, 1977
13. Onody C, Kristensen T, Lamm LU, et al: Tissue Antigens 12:323, 1978
14. Egorov IK, Vedernikov AA, Malenko AF, et al: In Kissmeyer-Nielsen F (ed): Histocompatibility Testing 1975. Copenhagen, Munksgaard, 1975, p 186
15. Mineu M, Mikhaylou T, Kastelan A, et al: Tissue Antigens 11:50, 1978
16. Renieri N, Stavropoulos C, Lepage V: Tissue Antigens 13:154, 1979
17. Zamir R, Brautbar C, Mizrachi Y, et al: Tissue Antigens 11:213, 1978