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Serogenetic Study of Romanies (Gypsies) from Region Spiš in Slovakia. Distribution of Blood and Serum Groups, Red Cell Isoenzymes, HLA Antigens of First Class and Apolipoprotein B Gene Polymorphism

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ABSTRACT The present paper informs about ABO, Rh, MN, P, Kell, Duffy, Kidd bood group frequencies, Gc serum group, ACP isoenzymes of red cells, frequencies of first class HLA antigen and allele frequency distributions for the ApoB3' VNTR locus. The results are compared with the data of the other in Slovakia living Romany groups, of European Romanies and non-Romany Slovak population. The Duffy*Fy allele frequency is much higher (0.129) as compared with the European standards. Both our and the European Romany data refer to a fact the Romany population genetic pool becomes different as compared with the host population. In our study of Spiš Romanies the inbreeding coefficient (F) 0.017 was observed. This paper is part of the scientific research program EUPECO-Biological History of European population n.ERBCIPDCT 940038 - Ethnogenesis of Romanies (Gypsies). Coordinator of the Contract : Prof. Alberto Piazza, Universita Torino, Italy.

In the Slovak Republic live 400,000 Romanies aproximately. The exact number of Romanies is not known, but it is estimated to be about 10 million in the world, about 5 million of whom live in Europe (McDowel, 1970). Romany history dates back to the first record of their stay in Europe. Earlier written, historical sources do not exist because the Romanies had no written records or hypotheses about their origin. As a result, various, often mysterious legends came into existence. One of the oldest legends maintains they came from Egypt. Despite misunderstandings and inaccuracies, some Romany groups misused this legend. Till the linguistic studies during the second half of century 18 suggested the Indian origin of Romanies (Rüdiger, 1782; Grellmann, 1787 ; Pott, 1845). Up to this time, however, their original India part, the question to what *jatis* (castes) they belonged and what was their social status in their ancient motherland are unknown. It may be seen, these questions are of mere academic interest, but the accurate data about the history of 5 million ethnicum living in Europe have important and wide-ranging social and political implications. Only few literature data are available to indicate that Romanies belonged to the low paria castes. It is true that at the present time *jatis* (caste) of Dom is "low", but it was not so in history, on contrary, Hindus caste vocabulary Jatikos confirms the political power of Doms in the past and Indian historicians reach the agreement they belonged to the original Indian population being of Dravid origin and shared the construction of one oldest civilization of protoindian civilization Mohendjodar, Harrape, Kalibanghán Lóthal etc. (Hübschmannová, 1996). Up to the present time, however, these facts were not published in any study about the Romany history. As no written direct documents about Romanies in India were found, other nonhistorical relevant scientific disciplines, *e.g.* population genetics help to solve this problem.

The goal of our program PECO, "Ethnogenesis of Romanies (Gypsies)" is to compare the genetic marker frequencies of the typed subgroups of Romanies living in Slovakia and of the members of typed some Indian castes: Gade Lohars, Dhamamis, Dholis, Doms, Domras, Banjaras, Kanjars, Sansis, Kal, Bahelias, Bhangis, Madaris, Sikligars, Rajputs, Bhils, Brahui living in Rajasthan, Uttar Pradesh and Punjab which typed Dr. Hübschmannová on the basis linguistic and ethnographic study. Based on the gene and haplotype frequencies (blood and serum groups, isoenzymes of red cells, HLA system and DNA polymorphisms), to determine the genetic distances, to determine some genetic parameter of population structure as the inbreeding coefficient and consanguinity frequency. These obtained gene frequencies will be used in calculating the genetic distances between the individual populations in reconstructing the

ethnogenesis of Romany populations concerned estimating the rate of European admixture in the present-day Romany pool etc. The present paper reports part of a large project PECO.

MATERIAL AND METHODS

Adult romanies of both sexy from Spiš region in East Slovakia were studied. Blood groups were determined by test-tube method according to instructions attached to diagnostic sera. Complete diagnostic sera were used to determine the ABO, Rh, MN,P and Kell blood group systems while with incomplete diagnostic sera (with the use of Coombs serum) the Duffy and Kidd systems were determined. Standard diagnostic sera were supplied by Immuno Diagnostica Vienna, Biotest Frankfurt, Immunocor Rodermark. The gene frequencies were calculated according to Mourant et al. (1976).

Serum factor Gc was typed according to Hirschfeld (1960). Horizontal starch gel electrophoresis was used to study acid phosphatase (ACP) according to the method of Hopkinson et al. (1963).

To standardize the HLA antigens the serologic method of the microlymphocytotoxic test (Staff, Research Branch NIAID, 1976) was applicated. The gene frequencies were estimated by the formula $g = 1 - \sqrt{1 - f}$ The associations between HLA-A,B,C, loci were calculated according to Mattiuz et al. (1970). The D value expressive of the gametic association was used. This value considering the alleles of 2 loci (Dausset et al., 1978).

Genomic DNA was purified from venous blood samples through saturated NaCl (Miller et al., 1988). The sequence of the 5' oligonucleotide used to prime the PCR was 5'-ATGGA AACGGAGAAATTATG-3'. The sequence of the 3' PCR primer was 5'-CCTTCTCACTT GGCAAATAC-3'. Genomic DNA was amplified in a total volume of 50 µl. One microgram of genomic DNA and 1 µg each of the oligonucleotides were used for each reaction. The four dNTPs were present each at a final concentration of 200 µM. The reaction buffer used was that recommended by the manufacturer. 1 unit of thermostable Taq polymerase (Boehringer Mannheim) was used to amplify the targeted sequence. The amplification was performed in a programmable thermal cycler Perkin Elmer Cetus Denaturing was carried out for 1 min at 94° C, annealing and extension was carried out for 6 min at 58°C. Twenty-six extension-denaturation cycles were carried out for each PCR (Boerwinkele, 1989). PCR products were visualized on 2% agarose gels containing ethidium bromide. For determination of fragment lengths we used markers VI and VIII (Boehringer Mannheim), and 100 bp Ladder (GibcoBRL).

The genetic distance was calculated using E index (Edwards and Cavalli-Sforza, 1972) and our results were compared with the European Romanies.

RESULTS AND DISCUSSION

In tables 1-12, the gene (haplotype) frequencies found in our Romany series are compared with relevant frequencies in other Romany samples known to us, as well as with the frequencies found in the non-Romany population of East Slovakia.

In all the polymorphic systems analyzed in this study genetic equilibrium was observed.

The data in ABO system show, that the *ABO*B* gene frequency, being especially the object of our interest, varies from the value of 0.049 in Swedish Romany population (Beckman et al., 1965) to the value of 0.316 in French Romanies (Cazal et al., 1951). The value of Spiš Romanies as well as of our investigated groups of probands are within the highest ones, excluding Olach Romanies, with very low (0.050) *ABO*B* gene frequency (Bernasovský et al., 1994 a). Small value of the gene frequency was likely determined in Swedish (Beckman et al., 1965). English (Clarke, 1973), Wales (Harper et al., 1977) and Yugoslavian Romanies (Hočevar, 1965).

The ratio of *ABO*A/B* genes frequency in Romanies in our study from Spiš, East Slovakia (Bernasovský et al., 1976c), Podskalka (Bernasovská et al., 1976) and Slovakia (Bernasovský et al., 1994b) varied between the values of 1.1 to 1.4. In Olach Romanies this ratio reaches the value of 2.2 (Bernasovský et al, 1994a). Great values of the ABO*A/B ratio were found in Swedish 7.9 (Beckman et al., 1965), English 4.7 (Clarke, 1973), Yugoslavian 3.9 (Hočevar, 1965) and in Romanies from Wales 3.3 (Harper et al.,

Table 1: A_1A_2BO gene frequencies in various European Romany and Non-Romany East Slovakian groups

Population	*Authors*	*N*	A_1	A_2	*B*	*O*
Romanies						
Spiš	Present Study	75	0.212	0.011	0.164	0.613
Slovakia	Bernasovský et al. (1994b)	140	0.223	0.015	0.227	0.535
East Slovakia	Bernasovský et al. (1976c)	2935	0.236	0.022	0.193	0.549
Podskalka	Bernasovská et al. (1976)	153	0.273	0.018	0.252	0.457
Olach	Bernasovský et al. (1994a)	119	0.101	0.024	0.050	0.825
Hungary	Rex-Kiss et al. (1973)	600	0.231	0.033	0.217	0.519
Yugoslavia	Hočevar (1965)	350	0.207	0.125	0.085	0.583
France	Cazal (1952)	113	0.185	0.042	0.316	0.457
Sweden	Beckman et al. (1965)	115	0.296	0.093	0.049	0.562
England	Clarke (1973)	109	0.200	0.109	0.064	0.627
Wales	Harper et al. (1977)	70	0.201	0.047	0.075	0.676
Non-Romanies						
East Slovakia	Juríčková et al. (1993 a)	1466	0.202	0.065	0.182	0.551

Table 2 : Rh haplotype frequencies in various European Romany and non-Romany East Slovakians

Population	*Authors*	*N*	*cde*	*Cde*	*cdE*	*cDe*	*CDe*	*cDE*	*CDE*
Romanies									
Spiš	Present Study	75	0.181	0.059	0.000	0.000	0.760	0.000	0.000
Slovakia	Bernasovský et al. (1994 b)	140	0.233	0.074	0.000	0.057	0.600	0.036	0.000
Podskalka	Bernasovská et al. (1976)	153	0.152	0.093	0.000	0.152	0.584	0.019	0.000
Olach	Bernasovský et al. (1994a)	119	0.430	0.003	0.000	0.000	0.543	0.018	0.000
Hungary	Rex-Kiss et al. (1973)	507	0.311	0.001	0.000	0.006	0.611	0.052	0.002
Yugoslavia	Hočevar (1965)	350	0.378	0.056	0.000	0.000	0.456	0.105	0.005
France	Cazal et al. (1952)	107	0.028	0.033	0.000	0.038	0.621	0.024	0.000
Sweden	Beckman et al. (1965)	115	0.191	0.026	0.000	0.000	0.684	0.103	0.000
Wales	Harper et al. (1977)	70	0.221	0.000	0.032	0.000	0.536	0.211	0.000
Non-Romanies									
East Slovakia	Juríčková et al. (1993 a)	1638	0.397	0.000	0.000	0.017	0.487	0.099	0.000

1977). In non-Romany East Slovakian population the ratio was evaluated to be 2.1 (Bernasovský et al., 1976 b). For Indian subcontinent population, where the world highest frequency of ABO*B blood group was found (Lehmann and Cutbush, 1952 ; Kirk et al., 1962; Glasgow et al., 1968; Race and Sanger, 1975; Mourant et. al., 1976 ; Papiha and Roberts, 1988; Walter et al., 1991, 1993), low values of ABO*A/B ratio are characteristic.

In the Rh system the frequency of *Rh*d* gene in the majority of European Romanies falls to the range of 0.300-0.450. In Spiš Romanies the value of *Rh*d* gene was found to be only 0.240, in Romanies from Podskalka (Bernasovská et al., 1976) 0.245. Similar low frequency of *Rh*d* gene were found in Swedish 0.217 (Beckman et al., 1965) and in Romanies from Wales 0.253 (Harper et al., 1977). In Indian populations the values vary from 0.100 to 0.346 (Mourant et al., 1976; Mastana and Papiha, 1992). A high frequency of the *Rh*CDe* haplotype was observed by Beckman et al., (1965). Cazal et al. (1951); Rex-Kiss et al. (1973). The *Rh*C* allele frequency, too, is elevated in almost all Romany populations. A very high *Rh* CDe* frequency seems to be characteristic for the Indian subcontinent, the data (Boyd and Boyd, 1954; Chaudhuri et al., 1967; Das et al., 1967; Walter et al., 1991,1993) are fairly consistent with a *Rh*CDe* frequency always above 0.400 and a maximum 0.760.

The distribution of MN blood system in Romanies from Spiš, East Slovakia (Bernasovský et al., 1976c), Podskalka (Bernasovská et

Table 3 : MN gene frequencies in various European Romany groups and non-Romany East Slovakians

Population	*Authors*	*N*	*M*	*N*
Romanies				
Spiš	Present Study	75	0.440	0.560
Slovakia	Bernasovský et al. (1994b)	140	0.604	0.396
East Slovakia	Bernasovský et al. (1976c)	405	0.530	0.470
Podskalka	Bernasovská et al. (1976)	153	0.627	0.373
Olach	Bernasovský et al. (1994a)	119	0.698	0.302
Hungary	Rex-Kiss et al. (1973)	536	0.559	0.441
Yugoslavia	Hočevar (1965)	350	0.426	0.574
France	Cazal (1951)	86	0.593	0.407
Sweden	Beckman et al. (1965)	115	0.479	0.521
England	Clarke (1973)	109	0.702	0.298
Wales	Harper et al. (1977)	70	0.464	0.536
Greece	Bartsocas et al. (1979)	200	0.678	0.323
Non-Romanies				
East Slovakia	Juríčková et al. (1993 a)	1455	0.668	0.332

Table 4 : P gene frequencies in various European Romany groups and non-Romany East Slovakians

Population	*Authors*	*N*	*P*1*	*P*QO*
Romanies				
Spiš	Present Study	75	0.774	0.226
Slovakia	Bernasovský et al. (1994b)	140	0.651	0.349
Podskalka	Bernasovský et al. (1976a)	125	0.387	0.613
Olach	Bernasovský et al. (1994a)	122	0.195	0.805
France	Cazal et al. (1951)	74	0.651	0.349
Wales	Harper et al. (1977)	48	0.487	0.513
Non-Romanies				
East Slovakia	Juríčková et al. (1993a)	1937	0.590	0.410

Table 5 : Kell gene frequencies in various European Romany groups and non-Romany East Slovakians

Population	*Authors*	*N*	*K*	*k*
Romanies				
Spiš	Present Study	75	0.013	0.987
Slovakia	Bernasovský et al. (1994b)	135	0.015	0.985
Podskalka	Bernasovský et al. (1976a)	134	0.015	0.985
Olach	Bernasovský et al. (1994 a)	119	0.063	0.937
England	Clarke et al. (1973)	108	0.034	0.976
Wales	Harper et al. (1977)	70	0.000	1.000
Greece	Bartsocas et al. (1979)	200	0.067	0.933
Non-Romanies				
East Slovakia	Juríčková et al. (1993a)	4463	0.032	0.968

al., 1976), Slovak Romanies (Bernasovský et al., 1994b) and the comparison of the European Romany data with those in East Slovakian non-Romanies (Juríčková et al., 1993a) is given in table 3.

Table 6 : Duffy gene frequencies in various European Romany groups and non-Romany East Slovakians

Population	*Authors*	*N*	*FY*A*	*FY*B*	*FY*QO*
Romanies					
Spiš	Present Study	75	0.481	0.390	0.129
Slovakia	Bernasovský et al. (1994 b)	140	0.483	0.348	0.169
Podskalka	Bernasovský et al. (1976a)	103	0.498	0.331	0.171
Olach	Bernasovský et al. (1994a)	119	0.525	0.475	0.000
England	Clarke (1973)	106	0.362	0.638	0.000
Wales	Harper et al. (1977)	69	0.398	0.602	0.000
Greece	Bartsocas et al. (1979)	118	0.606	0.394	0.000
Non-Romanies					
East Slovakia	Juríčková et al. (1993a)	1596	0.460	0.540	0.000

Table 7 : Kidd gene frequencies in various European Romany groups and non-Romany East Slovakians

Population	*Authors*	*N*	*JK*A*	*JK*B*
Romanies				
Spiš	Present Study	75	0.466	0.534
Slovakia	Bernasovský et al. (1994b)	134	0.480	0.520
Podskalka	Bernasovský et al. (1976a)	101	0.608	0.392
Olach	Bernasovský et al. (1994a)	111	0.487	0.513
Non-Romanies				
East Slovakia	Juríčková et al. (1993a)	2320	0.578	0.422

Table 8 : Gene frequencies of serum group GC in various European Romany groups and non-Romany East Slovakians

Population	*Authors*	*N*	*GC*1*	*GC*2*
Romanies				
Spiš	Present Study	75	0.798	0.202
Olach	Bernasovský et al. (1994a)	119	0.546	0.454
Yugoslavia	Fraser et al. (1969)	18	0.805	0.195
Non-Romanies				
East Slovakia	Juríčková et al. (1993b)	2458	0.677	0.323

Table 9 : Gene frequencies of red cell isoenzyme ACP in various European Romany groups and non-Romany East Slovakians

Population	*Authors*	*N*	*ACP*A*	*ACP*B*	*ACP*C*
Romanies					
Spiš	Present Study	75	0.328	0.672	0.000
Olach	Bernasovský et al. (1994a)	119	0.237	0.763	0.000
Slovakia	Siváková (1983)	235	0.289	0.711	0.000
Wales	Harper et al. (1977)	64	0.336	0.624	0.040
Non-Romanies					
East Slovakia	Juríčková et al. (1994)	2402	0.328	0.640	0.032

Gene *MN*M* frequency in our investigated Romany groups is within the range of 0.440 0.690 and does not differ from the frequency shown by Mourant et al. (1976) and Walter et al. (1991, 1993) in Indian subcontinent populations. The highest *MN*M* gene frequency was observed in Olach Romanies 0.698 (Bernasovský et al., 1994a). Likely the high occurrence of *MN*M* gene (0.702) was found by Clarke (1973) in English Romanies.

The other genetic systems studied are much less informative because few data from Indian and other Romany populations are available, and the difference in gene frequencies between Indians and Europeans are not much pronounced. An exception worth noticing is represented by the Duffy system. The gene frequency of the "amorph" allele *Duffy*Fy* was estimated to be 0.129 in Spiš Romanies and 0.169 in Slovak Romanies. A similar Duffy*Fy frequency has been reported from an East Slovak Romany isolate of Podskalka (Bernasovký et al., 1976a), while the other two investigations with Duffy typing included (Clarke et al., 1973 ; Harper et al., 1977) failed to find any *Duffy*Fy* case. In Europeans, the *Duffy*Fy* allele is rare, non-polymorphic, occasional reports of the *Duffy*Fy* individuals from Slovakia (Hrubiško et al., 1976) concern also Romanies. The *Duffy*Fy* allele is known to confer a complete resistance against the ineffective agent of the tertiary malaria, *Plasmodium vivax* (Miller et al., 1976), and is often in Negroes, as described by Lowe et al. (1971). From Indian region, data on *Duffy*Fy* gene distribution are scarce. Mourant et al. (1968) found 0.100 *Duffy*Fy* among Bhutanese, and Alciati (1968) among Pathans from West Pakistan, but both figures are based on small samples.

The analysis of the Gc factor shows the higher frequency of *Gc*1* gene which was higher in our sample as compared with Olach Romanies (Bernasovský et al., 1994a) and with the data in non-Romany East Slovakian population (Juričková et al., 1993b). As for ACP isoenzyme, our sample exibited higher occurrence of the *ACP*B* gene, whereas *ACP*C* gene was absent.

The HLA antigen frequencies of Spiš Romanies were compared with the Romany data of the Hungarian (Gyódi et al., 1981), Spanish (Imanishi et al., 1992), Indian populations (Imanishi et al., 1992) and finally with the East Slovakian (Table 10). The antigen *HLA*A1* showed the highest frequency, the antigen *HLA*A28* the lowest one. In locus B and C, the highest frequency was found for the antigen *HLA*B27 and HLA*Cw2.*

HLA A1-B27 and HLA*A1-B5* occur as the most frequent haplotypes in Spiš Romanies, each of them show low frequency among Slovaks. The *HLA*A1-B8, HLA*A2-B12 and HLA*A2-B7* haplotypes are very common among East Slovaks is absent in Romanies.

The considerable differences of HLA and blood markers in the Romany and the Slovakian population indicate their different ethnic origin.

In the present time we elaborate the results of the genealogical study of Spiš Romanies. In spite of the fact the genetic isolation degree is not yet determined in comparison with the Romany and non-Romany population, their isolation towards the other Romany and non-Romany population is clear. The above mentioned genetic variability could be caused by the mechanisms emerging in the isolated comities undoubtedly Spiš Romanies *i.e.* by the genetic drift and founder effect.

One hypervariable VNTR locus is located less than 100 bp 3' of the second transcriptional termination signal of the apolipoprotein B (ApoB) gene, which maps to human chromosome 2 (Knott et al., 1986). The length of the repeat unit at the apoB locus is 14-16 bp (Ludwig et al., 1989).

After electrophoretic separation of the amplified DNA products the size of the apoB alleles was determined by comparison with an allele marker. After this analyse we determined eight alleles with coresponded lengths : VNTR 51=900bp, VNTR 49=870bp, VNTR 47=840bp, VNTR 45=810bp, VNTR 37=690bp, VNTR 35=660bp, VNTR 33=630bvp. VNTR31=600bp (allelic designation by Boernwinkle et al., 1989). For comparison we present the data in non-Romanies from Slovakia (Kádasi et al., 1994) (Fig. 1). The most frequent allele was VNTR37 (60.61%). VNTR37/37 occur as the most frequent genotyp in Spiš Romanies (42.43%). The share of heterozygotes was 57.57%. The observed genotypes are given on table 12.

According to the data of population genetic investigation in Romanies result that the gene pool of the present-day Romany population in Europe is significantly different from the majority of European population.

Table 10: Gene frequencies of first class HLA antigen in Slovakian (Spiš), Hungarian and Spanish Romany samples compared with non-Romany East Slovakians and Indians

Antigen	*Romanies Spiš Present Study* N = 80	*Romanies Hungary Gyódi et al. (1981)* N=176	*Romanies Spain Imanishi et al. (1992)* N = 75	*India Imanishi et al. (1992)* N = 99	*Non-Romanies East Slovakia Juríčková et al. (1993c)* N = 672
HLA-A					
A1	0.500	0.254	0.281	0.111	0.155
A2	0.113	0.373	0.118	0.121	0.297
A3	0.025	0.017	0.057	0.079	0.098
A9	0.051	0.000	0.081	0.000	0.084
A10	0.065	0.084	0.070	0.000	0.064
A11	0.120	0.121	0.256	0.159	0.089
A19	0.113	0.003	0.100	0.000	0.000
A28	0.013	0.009	0.013	0.061	0.017
AX	0.163	0.380	0.000	0.000	0.177
HLA-B					
B5	0.149	0.154	0.140	0.000	0.079
B7	0.006	0.000	0.047	0.095	0.093
B8	0.013	0.006	0.013	0.038	0.076
B12	0.038	0.029	0.060	0.000	0.067
B13	0.000	0.006	0.000	0.010	0.023
B14	0.071	0.006	0.013	0.020	0.019
B15	0.013	0.026	0.013	0.000	0.030
B16	0.051	0.029	0.040	0.000	0.013
B17	0.045	0.031	0.067	0.000	0.025
B18	0.019	0.083	0.087	0.025	0.046
B21	0.099	0.012	0.040	0.000	0.007
B22	0.000	0.127	0.000	0.000	0.000
B27	0.275	0.074	0.073	0.028	0.055
B35	0.071	0.121	0.127	0.120	0.098
B37	0.000	0.003	0.040	0.038	0.000
B40	0.178	0.220	0.213	0.000	0.071
B41	0.006	0.049	0.000	0.000	0.000
BX	0.005	0.017	0.000	0.005	0.124
HLA-C					
Cw1	0.013	0.168	0.000	0.057	0.000
Cw2	0.608	0.065	0.115	0.025	0.054
Cw3	0.007	0.014	0.000	0.000	0.062
Cw4	0.066	0.096	0.097	0.134	0.127
Cw5	0.066	0.012	0.041	0.005	0.000
Cw6	0.000	0.038	0.000	0.112	0.000
Cw7	0.000	0.000	0.129	0.237	0.000
Cw8	0.000	0.003	0.000	0.000	0.000
Cw9	0.000	0.000	0.027	0.025	0.000
CX	0.642	0.606	0.000	0.005	0.539

Table 11: HLA haptotype associations in Romanies, Hungarian and East Slovak populations

Romanies present study N = 80		*Hungary Gyódi et al. (1981)* N=587		*East Slovakia Juríčková et al. (1993c)* N = 672	
Haplotype	*D*	*Haplotype*	*D*	*Haplotype*	*D*
Al, B5	0.0546	Al, B8	0.0443	A1B8	0.0640
Al, B27	0.0540	A2, B12	0.0265	A2B7	0.0215
All, B21	0.0214	B35, Cw4	0.0876	B35, Cw4	0.0746
B27, Cw2	0.0449	B27, Cw2	0.0249	B27, Cw2	0.0400

$D = \sqrt{\frac{d}{n}} - \sqrt{\frac{(b+d\ /\ /c+d)}{n^2}}$, where a, b, c, d represent the number of ++, +-, -+, -- phenotypes respectively, n is the sample size.

Table 12 : ApoBII-alleles and their frequencies in Romanies from Spiš and non-Romanies Slovakians

Slovak (n = 230) (Kádasi et al., 1994)		*Romanies (n = 33) (Present study)*		*3' VNTR (Present study)*	
Allele	*Frequency*	*Allele*	*Frequency*	*Genotype*	
29	0.0022	31	0.0909	VNTR 35/31	0.0303
31	0.0696	33	0.0606	35/33	0/0303
33	0.0696	35	0.1061	37/31	0.0909
35	0.2196	37	0.6061	37/33	0.0606
37	0.3457	45	0.0303	37/35	0.0909
39	0.0957	47	0.0454	37/37	0.4243
41	0.0087	49	0.0303	45/35	0.0303
43	0.0022	51	0.0303	45/37	0.0303
				47/31	0.0606
45	0.0153			47/37	0.0303
47	0.0739			49/33	0.0303
49	0.0870			49/37	0.0303
51	0.0109			51/35	0.0303
				51/37	0.0303

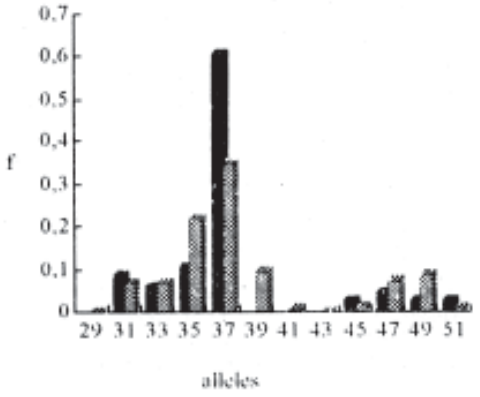

Fig. 1. Apo BII-Comparison of identified alleles in Romanies and non-Romanies and their frequencies

The investigation of Romany populations could be roughly divided into two categories : the first consisted in investigations of Romany isolates and the second consisted in some "ordinary" Romany population studies (Bernasovský, 1994).

Few publications of the first category are as follows : the study of Yugoslavian Romany population with approximate number of 700 persons (Hočevar, 1965), the study by Ely (1961, 1966) about two little isolated groups of French Romanies and our investigations of about 350 persons in Podskalka, as well as the study of 900 Olach Romanies from Vinodol and our study about 800 persons of Spiš Romanies.

With the regard of Indian origin of European Romanies to be expected a significant tendency to consanguinic marriage in them. Since this tendency is characteristic for a great part of populations living on the Indian subcontinent (Sanghvi, 1966), they have practically the extr- emly high values of inbreeding coefficient. The marriage customs are known as most stable and conservative features in the Indian society.

The results of population genetic structure have revealed high consanguinity and coefficient of inbreeding (F) in two other particular Romany subpopulations in Slovakia. According to the coefficient of inbreeding the Olach Romanies count F = 0.05 (Ferák et al., 1987), two series of the Slovakian Romanies from Gemer (a single region in Slovakia) attain F equal to F = 0.014 and F =0.021, respectively (Siváková et al., 1994).

As far as for the Slovak non-Romany population, tendency towards endogamy and high coefficient of inbreeding prevailed in the past among the Slovak rural populations, too (Ferák et al., 1980 ; Siváková et al., 1995 ; Siváková and Walter, 1996).

Sieglová and Siváková (1985) reported the results of genetic distances E (Edwards and Cavalli-Sforza, 1972) and D (Nei, 1971), calculated on the basis of 9 blood group and serum protein systems, among 4 population samples, 2 Romany populations from Slovakia, selected Indians populations and the Slovak non-Romany population. Both values have revealed the greatest distance value between the two Romany samples from Slovakia.

Our calculated index E (Edwards and Cavalli-Sforza, 1972) indicated the largest genetic distance and therefore the most different allele frequency between Olach and French Romanies (E = 0.3026). The differencies between distances were observed between Olach and Slovak Romanies (E = 0.2135), Olach and Spiš Romanies (E=0.2123), Romanies from Podskalka settlement and Walles Romanies (E = 0.2558), England Romanies (E=0.2243) and Yugoslavian Romanies (E = 0.2217).

The most evident similarity between Hungarian

and East Slovakian Romanies (E = 0.0283) was observed.

Both our and data from European Romanies point to the fact, that gene pool of Romany populations is different from that the host population.

In the frame of population genetic studies, we stated coefficient of inbreeding (F) in selected Slovak Romany population. The average value of inbreeding coefficient (F) was found to be 0.017. It suggested the estimated high F value may have expressive influence on wide production of homozygotes in this population. This fact is documented by high incident of homozygotes (42.43%) in the followed ApoB 3' locus, too.

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