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Biochemical comparisons in Yugoslavian rodents of the families Arvicolidae and Muridae

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Abstract

Twenty eight proteins from six species of Arvicolidae and three species of Muridae were examined electrophoretically. Animals of the species *Microtus arvalis*, *M. epiroticus*, *Clethrionomys glareolus*, *Pitymys subterraneus*, *P. felteni*, *Dinaromys bogdanovi*, *Apodemus flavicollis*, *A. sylvaticus* and *A. agrarius* were collected in various regions of Yugoslavia. The Nei genetic distances approximately correspond to previous conceptions of the relationship of the species. The distance between *Microtus* and *Pitymys* species is negligible, *Microtus* to *Clethrionomys* is 0.60, and the average distance from *Dinaromys* to the remaining arvicolids is 1.21. Thereby the special position of *Dinaromys* within the voles is again accentuated. *Apodemus sylvaticus* and *A. flavicollis* are not distinguishable in Yugoslavia with the loci examined. Their distance to *Apodemus agrarius*, on the other hand, is great.

Introduction

General agreement has not been reached on the classification of some of the species dealt with in this study, and differing classifications have been proposed (HINTON 1926; GROMOV and POLYAKOV 1977; CORBET 1978; CHALINE and MEIN 1979; HONACKI et al. 1982). CORBET (1978: 94) notes that: "Only a very comprehensive revision using all the

available characters is likely to result in a stable arrangement." The use of biochemical characters may aid substantially.

The purpose of this paper is to apply the results of electrophoretic analysis to the classification of certain species of the Arvicolidae and Muridae of the Yugoslavian fauna. The systematic value of electrophoretic data and the advantages of using it as a systematic tool have been stressed by AVISE (1974). Using electrophoretic analysis in the study of the Arvicolidae in parts of western Europe and North America, GRAF and coworkers (GRAF and SCHOLL 1975; GRAF and MEYLAN 1980; GRAF 1982) have found a general correspondence between classifications based on classical systematics and on genetic distances derived from electrophoretic data, with some differences.

This paper presents a preliminary survey of a number of Yugoslavian microtine species and species of the genus *Apodemus*. Joint analysis of samples collected at the same sites of the two families allows comparison of patterns of intra-family variation. The microtines, including voles and lemmings, are a distinct group assigned to the family Arvicolidae by HONACKI et al. (1982). About half the microtine species found in Yugoslavia are represented in this study, including *Microtus arvalis*, the common vole; *M. epiroticus* (*M. subarvalis*), which is distinguished from *M. arvalis* on the basis of its karyotype (MEYER et al. 1972) and of morphological and other characteristics (RUŽIĆ et al. 1975; PETROV and RUŽIĆ 1982); *Clethrionomys glareolus*, the bank vole; two species of the genus *Pitymys*, *P. subterraneus* and *P. felteni*; and *Dinaromys bogdanovi*, Martino's vole endemic to the mountains of southwestern Yugoslavia.

Apodemus is a fairly distinct genus within the Muridae. It consists of about 13 species (HONACKI et al. 1982) and is the dominant group of murid rodents in the Palaearctic (CORBET 1978). Three of the five *Apodemus* species found in Yugoslavia are represented in this study, *A. flavicollis*, the yellow-necked mouse; *A. sylvaticus*, the wood mouse; and *A. agrarius*, the striped field mouse.

Materials and methods

Individuals of the families Arvicolidae ($n = 55$) and Muridae ($n = 21$, all in the genus *Apodemus*) were live-trapped at 11 localities in Yugoslavia. The number of specimens trapped at each locality is given in parentheses.

Microtus arvalis (*M. a.*): Debrč (4); Brezovica, Šara Mts. (6); Knjaževac (2); Kanjiža (13).

Microtus epiroticus (*M. e.*): Skopje (2).

Clethrionomys glareolus (*C. g.*): Brezovica, Šara Mts. (1); Žljeb Mts. (3); Travna Gora, Ribnica (5); Ig, Ljubljana (1); Djerdap (2).

Pitymys subterraneus (*P. s.*): Ig, Ljubljana (2); Debrč (2); Crni Vrh, Žagubica (1); Kanjiža (1).

Pitymys felteni (*P. f.*): Brezovica, Šara Mts. (1).

Dinaromys bogdanovi grebenscikovi (*D. b. g.*): Žljeb Mts. (4 = ♀ + litter of 3).

Dinaromys bogdanovi bogdanovi (*D. b. b.*): Brezovica, Šara Mts. (5 = ♀ + litter of 4).

Apodemus flavicollis (*A. f.*): Beška, Srem (7); Ig, Ljubljana (3); Djerdap (1).

Apodemus sylvaticus (*A. s.*): Beška, Srem (1); Crni Vrh, Žagubica (2); Djerdap (1).

Apodemus agrarius (*A. a.*): Beška, Srem (5); Debrč (1).

Methods of tissue sample preparation and horizontal starch gel electrophoresis were essentially as described in SELANDER et al. (1971) with slight modifications. For Lithium hydroxide gels, 11.0 % Electrostarch Lot 307 (Electrostarch Co., Madison, WI, USA) was used and 12.4 % starch was used for all other gels. The buffer and stain systems for proteins screened in this study (Table 1) are described in SELANDER et al. (1971) except for stain a. 50 ml 0.2 M Tris HCl, pH 8.0, 250 mg D-sorbitol, 10 mg NAD, 15 mg NBT, 2 mg PMS; stain b. 37.5 ml H₂O, 12.5 ml 0.2 M Tris HCl, pH 8.0, 5 ml 0.1 M MgCl₂, 75 mg glucose, 1 ml G-6-PDH, 50 mg ATP, 10 mg NAD, 10 mg NADP, 10 mg MTT, 5 mg PMS; and stain c. 50 ml 0.2 M Tris HCl, pH 8.0, 2 ml 0.25 M MnCl₂, 40 mg leucylalanine, 10 mg snake venom, 20 mg peroxidase, 10 mg O-dianisidine-di-HCl. Alcohol dehydrogenase (ADH) does not have to be stained specifically and is seen on many dehydrogenase gels. It was read on gels stained for glycerol-3-phosphate dehydrogenase (GPD).

Table 1

Enzymes and nonenzymatic proteins screened

Enzyme or protein name	Abbreviation	Tissue	Buffer system ¹	Stain system ¹
Alcohol dehydrogenase	ADH	Liver	5. TC, pH 8.0	6
Glycerol-3-phosphate dehydrogenase	GPD	Liver	5. TC, pH 8.0	6
Sorbitol dehydrogenase	SORDH	Liver	5. TC, pH 8.0	a
Lactate dehydrogenase	LDH	Kidneys	5. TC, pH 8.0	10
Malate dehydrogenase	MDH	Liver	4. TC, pH 6.3	11
Malic enzyme	ME	Liver	4. TC, pH 6.3	11
Isocitrate dehydrogenase	ICD	Kidneys	5. TC, pH 8.0	8
Glucose-6-phosphate dehydrogenase	G-6PD	Kidneys	5. TC, pH 8.0	4
Superoxide dismutase	SOD	Kidneys	9. TME	7
Glutamate-oxaloacetate transaminase	GOT	Liver	or	7
			4. TC, pH 8.0	7
			2. LiOH	5
Hexokinase	HK	Kidneys	or	5
			5. TC, pH 8.0	5
Peptidase	PEP	Liver	5. TC, pH 8.0	b
Glucose phosphate isomerase	GPI	Liver	4. TC, pH 6.3	c
Hemoglobin	HB	Hemolysate	5. TC, pH 8.0	14
Albumin	ALB	Plasma	1. Tris HCl	3
Transferrin	TRF	Plasma	2. LiOH	3
General proteins	GP	Hemolysate	2. LiOH	3
			1. Tris HCl	3

¹ Given in SELANDER et al. (1971) except for stains a, b, and c, described in Materials and methods.

Mathematical methods: Gene frequencies, measures of genetic variation, NEI's (1978) unbiased genetic distance and a dendrogram based on the latter were derived from input on single individual genotypes (electromorphs), using the computer program BIOSYS-1 (SWOFFORD and SELANDER 1981). This program can handle missing data, basing pairwise genetic distances only on loci present in both species. All the allozymes were not detected over the range of species studied, and there were differences in enzyme activity among species. Allozymes not present or which could not be reliably discerned in particular species are left blank (Table 2). Loci not included are GPI-1 for *Microtus arvalis*, SOD-3 for *M. epiroticus*, PEP-1 for *Clethrionomys glareolus* and HK-1 for the two *Pitymys* species, leaving 27 loci analysed for each of these five species. *Dinaromys bogdanovi bogdanovi* was also analysed with 27 loci, missing GPI-2, and *D. b. grebenscikovi* with 24, missing GPI-2, ME, HK-1 and HK-4. G-6-PD, PEP and GP-2 are blank for all the *Apodemus* species. *A. flavicollis* also lacks ICD-2, ME, HK-4 and ADH, leaving 21 loci. There are 22 loci analysed for *A. sylvaticus*, which also lacks ME, HK-1 and HK-4, and 24 loci for *A. agrarius*, which lacks ICD-2. The unweighted pair group method with recomputation of coefficients by arithmetic averaging (UPGMA) was used in the BIOSYS-1 cluster analysis (SNEATH and SOKAL 1973).

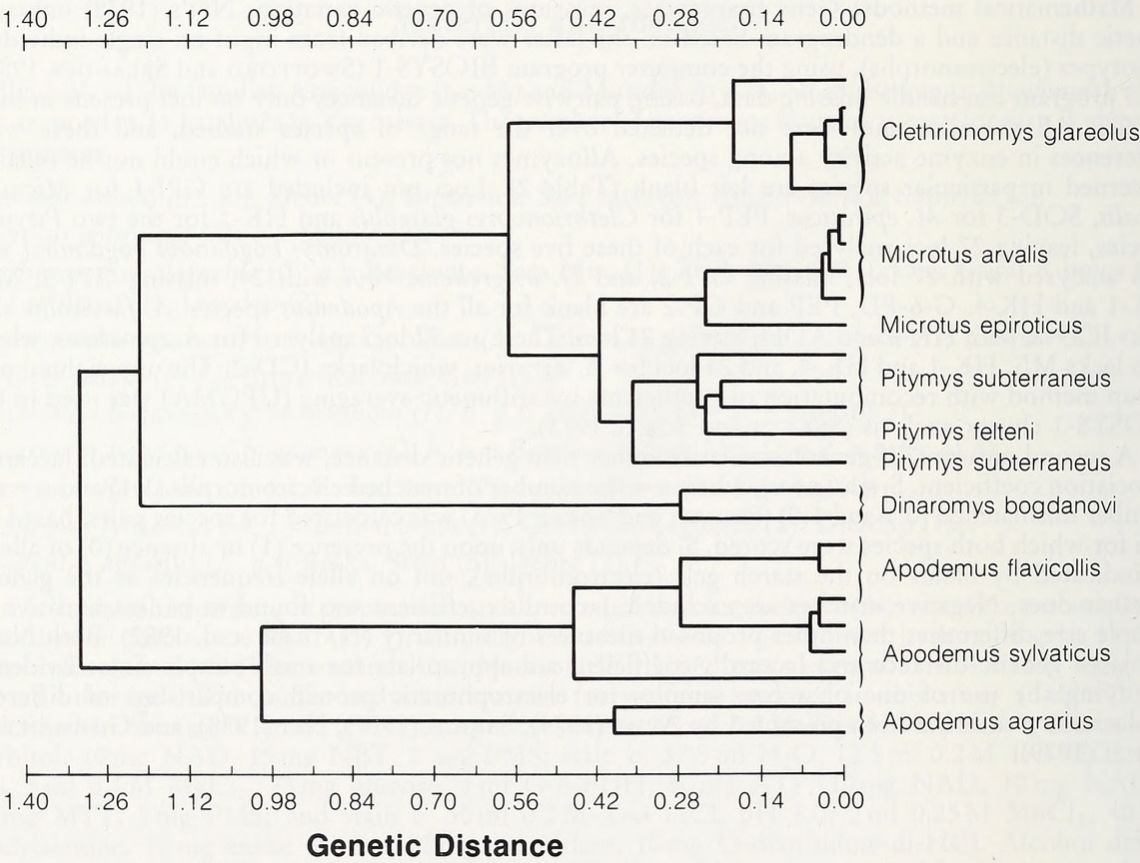
A second measure, of genetic similarity rather than genetic distance, was also calculated. Jaccard's association coefficient, $S_j = a/(a + u)$, where a = the number of matched electromorphs (1:1) and u = the number mismatched (0:1 and 1:0) (SNEATH and SOKAL 1973) was calculated for species pairs, based on loci for which both species were scored. S_j depends only upon the presence (1) or absence (0) of alleles as indicated by bands on the starch gels (electromorphs), not on allele frequencies as the genetic distance does. Negative matches are excluded. Jaccard's coefficient was found to be less sensitive to sample size differences than other proposed measures of similarity (NADLER et al. 1982). Both NEI's unbiased genetic distance and Jaccard's coefficient are appropriate for small sample sizes. Evidence justifying the use of one or a few samples for electrophoretic protein comparisons of different evolutionary units has been presented by AVISE (1974), SARICH (1977), NEI (1978), and GORMAN and RENZI (1979).

Results and discussion

Arvicolidae

The microtines have been recognized as a sharply defined group among the rodents, whether they have been given family status (CHALINE and MEIN 1979; HONACKI et al. 1982) or subfamily status (SIMPSON 1945; CORBET 1978). One of the most unstable areas of classification at the generic level is the *Microtus/Pitymys/Arvicola* group (CORBET 1978), and opinion differs as to whether the latter two should be retained as distinct genera or considered subgenera of *Microtus* (GROMOV and POLYAKOV 1977; HONACKI et al. 1982).

Biochemical similarities of the species in this study are based on electrophoretic data (Table 2) representing a broad range of glucose-metabolizing enzymes, other enzymes, and nonenzymatic proteins. Twenty eight presumptive loci were scored for the arvicolids and 25 for species of *Apodemus*, but all the allozymes were not detected over the range of species studied (see Materials and methods). Allozymes not present in a particular species are left blank in Table 2. The results given here are an initial survey of some of the protein variants present in these species and do not represent estimates of population frequencies. The interrelationship among the species based on their biochemical similarities (Table 2) is depicted in a dendrogram (Fig. 1). The *Microtus* and *Pitymys* samples form one major cluster at an average genetic distance $\bar{D}=0.58$ from the *Clethrionomys glareolus* cluster. The *Dinaromys bogdanovi* samples are distinct from the other microtines with $\bar{D}=1.21$. The distance of *Apodemus agrarius* from *A. sylvaticus* and *A. flavicollis* is almost 1.00, and $\bar{D}=1.30$ between the families of arvicolids and murids. The goodness of fit of the dendrogram to the cophenetic matrix derived from the NEI (1978) unbiased genetic distances is given by a cophenetic correlation of 0.913. NEI showed that the number of individuals used for estimating average heterozygosity and genetic distance can be very



Dendrogram of unbiased genetic distances (NEI 1978) derived from data of Table 2

Table 2

Allele frequencies of species of *Microtus*, *Clethrionomys*, *Pitymys*, *Dinaromys*, and *Apodemus* in Yugoslavia

Species* No of Individuals Loci and alleles**		<i>M.a.</i> 25	<i>M.e.</i> 2	<i>C.g.</i> 12	<i>P.s.</i> 6	<i>P.f.</i> 1	<i>D.b.g.</i> 4	<i>D.b.b.</i> 5	<i>A.f.</i> 11	<i>A.s.</i> 4	<i>A.a.</i> 6
I. Glucose-metabolizing enzymes											
LDH-A	a	1.00	1.00	0.96	1.00	1.00	1.00				
	b							1.00			
	c										1.00
	d			0.04					1.00	1.00	
LDH-B	a										1.00
	b								1.00	1.00	
	c	1.00	1.00	1.00	1.00	1.00	1.00	1.00			
ICD-1	a	0.95	1.00	1.00	1.00	1.00					
	b	0.05									
	c									0.25	
	d						1.00	1.00	1.00	0.75	
	e										1.00
ICD-2	a				0.60		1.00	1.00		1.00	
	b	0.80	1.00	0.87	0.40	1.00					
	c	0.20		0.13							
G-6PD	a		1.00								
	b	0.08		0.17							
	c	0.92		0.83	1.00	1.00	1.00	1.00			
GPD	a	0.98	1.00	1.00	1.00	1.00					
	b						1.00		1.00	1.00	
	c										1.00
	d	0.02						1.00			
GPI-1	a		1.00		1.00	1.00		1.00	1.00	1.00	1.00
	b			1.00							
	c						1.00				
GPI-2	a				1.00	1.00					
	b	0.92	1.00						1.00	1.00	1.00
	c			1.00							
	d	0.08									
MDH-1	a			1.00	0.17						
	b	1.00	1.00		0.83	1.00	1.00	1.00			1.00
	c								1.00	1.00	
MDH-2	a	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	
	b										1.00
ME	a			0.22							1.00
	b			0.78							
	c							1.00			
	d	1.00	1.00		1.00	1.00					

Table 2 (continued)

Species*		M.a. 25	M.e. 2	C.g. 12	P.s. 6	P.f. 1	D.b.g. 4	D.b.b. 5	A.f. 11	A.s. 4	A.a. 6
No of Individuals Loci and alleles**											
HK-1	a							0.25	0.50		0.50
	b	0.56	0.75	0.95							
	c							0.75			
	d								0.50		0.50
	e	0.11									
	f	0.33									
	g		0.25	0.05							
HK-2	a								1.00	0.75	
	b										0.83
	c	0.96	1.00	1.00	0.08	1.00	1.00	1.00		0.25	0.17
	d	0.04			0.92						
HK-3	a			0.14	0.33	0.50	1.00	1.00	1.00	1.00	1.00
	b	0.98	1.00		0.67						
	c	0.02		0.77		0.50					
	d			0.09							
HK-4	a	0.02									
	b	0.94									
	c		1.00					1.00			
	d	0.04		1.00	1.00						0.33
	e					1.00					0.67
II. Other enzymes											
GOT-1	a	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
GOT-2	a	0.14									0.17
	b	0.86	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00	0.83
SORDH	a	1.00	1.00	1.00	0.17				1.00	1.00	1.00
	b						1.00	1.00			
	c				0.83	1.00					
PEP-1	a	1.00	1.00		1.00	1.00					
	b						1.00	1.00			
SOD-1	a						1.00	1.00			
	b	1.00	1.00	1.00	1.00	1.00			1.00	1.00	1.00
SOD-2	a	1.00	1.00	1.00	1.00	1.00					
	b						1.00				
	c								1.00	0.75	
	d							1.00		0.25	
	e										1.00
SOD-3	a	1.00		1.00	1.00	1.00			1.00	1.00	
	b						1.00	1.00			
	c										1.00
ADH	a	1.00									
	b		1.00		1.00		1.00	1.00		0.25	
	c			1.00		1.00				0.75	1.00

Table 2 (continued)

Species*		M.a.	M.e.	C.g.	P.s.	P.f.	D.b.g.	D.b.b.	A.f.	A.s.	A.a.
No of Individuals		25	2	12	6	1	4	5	11	4	6
Loci and alleles**											
III. Nonenzymatic proteins											
HB-A	a			0.08	0.33				0.20	0.75	1.00
	b	1.00	1.00	0.92	0.67	1.00			0.80	0.25	
	c						1.00	1.00			
HB-B	a			0.08	0.33				0.20	0.75	1.00
	b	1.00	1.00	0.92	0.67	1.00			0.80	0.25	
	c						1.00	1.00			
ALB	a						0.13				
	b						0.87	1.00			1.00
	c			1.00					1.00	0.75	
	d	1.00	1.00		1.00	1.00				0.25	
GP-1	a						1.00	1.00			
	b	1.00	1.00	1.00	1.00	1.00			1.00	1.00	1.00
GP-2	a	0.96	0.50	1.00	0.33	0.50	1.00	1.00			
	b	0.04	0.50		0.67	0.50					

* Abbreviations for loci are given in Table 1, for species in Materials and methods. – ** Alleles are listed in order of increasing mobility; a is the slowest.

small if the genetic distance is large and the average heterozygosity low. For average heterozygosities of the order of 0.06 and genetic distances greater than 0.15, the expected magnitude of the bias is not important even if a single individual is sampled from each of the species compared (NEI 1978).

Several populations of *C. glareolus*, *M. arvalis*, and *P. subterraneus* were sampled, and the variability within these species is evident (Fig. 1). Due to the sample sizes, however, the estimates of intraspecific variability are tentative. There was no evidence of clines or other patterns in the variability within any of the species. Three major clusters within the Arvicolidae mirror the classical systematic classification: 1) *Clethrionomys*, 2) *Microtus/Pitymys* group, and 3) *Dinaromys* (Fig. 1). Within the second group there are further clear subdivisions. The four *M. arvalis* samples cluster tightly (D between 0.01 and 0.07). GRAF (1982) found very similar genetic distances between eight populations of western European *M. arvalis* (D between 0.00 and 0.08). The *M. epiroticus* sample is distinct from the *M. arvalis* cluster (D = 0.24). The karyotype of *M. epiroticus* is characterized by $2n = 54$, while for *M. arvalis* $2n = 46$ (MEYER et al. 1972; ŽIVKOVIĆ et al. 1976); hybrids between them are sterile (PETROV and RUŽIĆ 1982). The single individual of *P. felteni* is clustered among those of *P. subterraneus* but there is considerable biochemical variation within the *Pitymys* samples, D ranging from 0.19 to 0.37 (Fig. 1). The close biochemical affinity of *P. subterraneus* and *P. felteni* is also apparent in their association coefficient, $S_J = 0.68$, which is almost as high as that between the subspecies of *D. bogdanovi*, $S_J = 0.69$ (Table 3).

The *Pitymys* samples form a subcluster separate from the *Microtus* samples (Fig. 1), with an average distance of $\bar{D} = 0.42$ between the subclusters. While the genetic distances, based on allelic frequencies, indicate a difference between *Microtus* and *Pitymys*, this is not the case when one looks only at presence or absence of alleles as represented in Jaccard's coefficient of association (Table 3). *M. epiroticus* is more closely related to the *Pitymys* species than *M. arvalis* is; *M. arvalis* is just as similar to *P. felteni* as it is to *M. epiroticus*. The relationships among *M. arvalis*, *P. subterraneus*, and *C. glareolus* found in this study

Table 3

Jaccard's coefficient of association, showing the biochemical similarity between pairs of certain rodent species of Yugoslavia

	<i>M.e.</i>	<i>P.s.</i>	<i>P.f.</i>	<i>C.g.</i>	<i>D.b.g.</i>	<i>D.b.b.</i>	<i>A.f.</i>	<i>A.s.</i>	<i>A.a.</i>
<i>M.a.</i>	0.558	0.578	0.548	0.451	0.196	0.164	0.275	0.273	0.200
<i>M.e.</i>		0.649	0.618	0.400	0.231	0.222	0.294	0.361	0.214
<i>P.s.</i>			0.684	0.511	0.267	0.250	0.371	0.415	0.267
<i>P.f.</i>				0.476	0.244	0.227	0.303	0.333	0.250
<i>C.g.</i>					0.200	0.154	0.389	0.390	0.261
<i>D.b.g.</i>						0.690	0.171	0.220	0.162
<i>D.b.b.</i>							0.189	0.256	0.186
<i>A.f.</i>								0.846	0.333
<i>A.s.</i>									0.308

Abbreviations for species are given in Materials and methods.

are quite similar to those found by GRAF (1982), with *C. glareolus* being a little more closely associated with *P. subterraneus* than with *M. arvalis* (Table 3).

The monospecific genus *Dinaromys*, Martino's vole, was earlier referred to the genus *Dolomys*, but evidence indicates it is a separate, recent genus (CORBET 1978). Animals of this genus live in small isolated populations at altitudes from sea level to 2200 m (PETROV and TODORVIĆ 1982). KOENIGSWALD (1980, cited in HONACKI et al. 1982) believes, on the basis of the internal structure of the molar enamel, that *Dinaromys* probably has no close relationship with any other living arvicolid. The distinctiveness of the endemic relict species *D. bogdanovi* is strongly supported by the biochemical data (Fig. 1, Table 3). It is almost as distinct from members of its own family ($\bar{D}=1.21$) as it is from the *Apodemus* species in the family Muridae ($\bar{D}=1.30$). The species has been divided on morphological grounds into two subgroups, the *bogdanovi* group in the northern part of its distribution and the *grebensickovi* group in the southern part (TODORVIĆ 1956; PETROV and TODORVIĆ 1982). While the genetic variability of *D. bogdanovi* is low (Table 2), as is typical of isolates, most of the variation which does occur is between the groups, not within them, supporting their classification into separate subgroups (Fig. 1).

Muridae

Three species of the genus *Apodemus* are included in this study. There are taxonomic problems concerning the genus (CORBET 1966; Williams et al. 1980). *A. sylvaticus* and *A. flavicollis* are difficult to distinguish morphologically and often occupy the same deciduous woodland habitats (DULIĆ and TVRTKOVIĆ 1972). CORBET (1978) suggested that the two species may hybridize, and some investigators believe that introgression has occurred between the species in southern Europe, whereas others think they occur together without interbreeding (GEMMEKE 1980). *A. agrarius* is easily distinguished morphologically.

The genetic distance found between *A. sylvaticus* and *A. flavicollis* was small (\bar{D} between 0.28 and 0.46) compared to their great distance from *A. agrarius* ($\bar{D}=0.99$), in agreement with morphological (DULIĆ and TVRTKOVIĆ 1972; NIETHAMMER and KRAPP 1978), chromosomal (VUJOŠEVIĆ ET AL. 1984), and earlier electrophoretic data (GEMMEKE 1980). *A. sylvaticus* and *A. flavicollis* showed the strongest association of any of the taxa, with an association coefficient of 0.85 (Table 3). ENGEL et al. (1973) and GEMMEKE (1980) found greater biochemical differences between *A. sylvaticus* and *A. flavicollis* populations in western Europe. The dissimilarity of *A. agrarius* from the other two *Apodemus* species is much greater than that between other rodent congeners reported in the literature (AVISE 1974).

There was no heterozygosity found in the four *A. sylvaticus* individuals sampled. They were all homozygous for the same allele at 15 loci, but one anomalous individual from Beška was homozygous for different alleles than the other three at seven other loci. Because of the high degree of homozygosity the effect of this one unusual individual is magnified in the dendrogram and it is distinct from the others ($\bar{D}=0.46$). Its electrophoretic pattern for these variant loci differed from that of *A. flavicollis* except for the hemoglobins. Because of the unusual electrophoretic results for this individual, its species identity was double checked and verified. Moreover, the specimens of *A. sylvaticus* and *A. flavicollis* are from regions of Yugoslavia in which there is no problem in distinguishing them morphologically. GEMMEKE (1980) found geographical variation in western European *A. sylvaticus*, and both the present study and that of WILLIAMS et al. (1980) suggest geographical variation in Yugoslavian *A. sylvaticus*.

Electrophoretic analysis is not useful for investigating differences between families because the number of shared alleles become minimal. It is indicative of the distinctiveness of *D. bogdanovi* within the microtines and of *A. agrarius* within the genus *Apodemus* that the genetic distance between the two families is not much greater than the distances of these two species from other members of their respective groups (Fig. 1).

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Zusammenfassung

Biochemische Vergleiche von jugoslawischen Rodentia der Familien Arvicolidae und Muridae

28 Proteine wurden bei 6 Arten der Arvicolidae und 3 Arten der Muridae elektrophoretisch untersucht. Es handelt sich um die Arten *Microtus arvalis*, *M. epiroticus*, *Clethrionomys glareolus*, *Pitymys subterraneus*, *P. felteni*, *Dinaromys bogdanovi*, *Apodemus flavicollis*, *A. sylvaticus* und *A. agrarius* von denen Tiere in verschiedenen Gebieten Jugoslawiens gesammelt wurden. Die nach NEI berechneten genetischen Abstände entsprechen annähernd den bisherigen Vorstellungen über die abgestufte Verwandtschaft der Arten. Der Abstand zwischen den beiden *Microtus*- und *Pitymys*-Arten ist gering, der zu *Clethrionomys* beträgt 0,60, der mittlere Abstand von *Dinaromys* zu den übrigen Arvicoliden 1,21. Damit wird die Sonderstellung von *Dinaromys* innerhalb der Wühlmäuse erneut betont. *Apodemus sylvaticus* und *A. flavicollis* sind in Jugoslawien mit den untersuchten Loci nicht unterscheidbar. Ihr Abstand zu *Apodemus agrarius* ist hingegen groß.

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