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Skull features of the common vole (*Microtus arvalis* sensu lato) from Hungary: craniometrical evidence for its taxonomic detachment

Georgi MARKOV[1,*], Gabor CSORBA[2], Maria KOCHEVA[1], Milena GOSPODINOVA[1]

[1]Institute of Zoology, Bulgarian Academy of Sciences, 1000 Sofia - BULGARIA

[2]Hungarian Natural History Museum, Budapest, H-1088 - HUNGARY

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Abstract: A revision was carried out on 245 adult skull specimens of the common vole (*Microtus arvalis* sensu lato) kept in the mammal collection of the Hungarian Natural History Museum in Budapest. A highly effective craniometric classification function was used to reveal whether they belonged to the common vole (*M. arvalis* Pallas, 1778 sensu stricto) or to the East European vole (*Microtus levis* Miller, 1908). It was found that *M. levis* specimens were present in the Hungarian mammal fauna; the boundaries of that animal's European range were thus expanded in the west. The distribution of the East European vole and the common vole in Hungary showed that both of these vole species occur together in all of the studied regions, with different predominant presence. *M. levis* occurred in lower numbers than *M. arvalis* in the western part of the country while the 2 species were relatively equally present in the central part of the country, with an insignificant preponderance of *M. arvalis* in the northeastern part. Conversely, *M. levis* had a clearly expressed predominance over *M. arvalis* in several regions of the northern part of the country. The craniometrical specificity of the 2 vole species in Hungary was characterized on the basis of craniologically determined samples. Craniometrical characteristics of both species showed similar absolute variability of their corresponding parameters with poorly expressed cranial sexual dimorphism.

Key words: Skull features, *Microtus arvalis*, *Microtus levis*, taxonomic detachment, distribution, Hungary

Introduction

As a result of the systematic revision of the polytypic species *Microtus arvalis* Pallas, 1778, it was found that 2 species, *Microtus levis* Miller, 1908 = *M. rossiaemeridionalis* Ognev, 1924, and *M. arvalis* Pallas, 1778 (sensu stricto), are morphologically sibling species. The supervening karyotaxonomic and biochemical investigations of the species differentiation of these sibling voles outlined their ranges. At present, the ranges of the 2 species, generally accepted on the grounds of distribution evidence, are as follows. *M. arvalis* occurs in Eurasia from the European Atlantic coast in the west to the Mongolian Altai Mountains in the east, and from the Baltic Sea coast (Finland and Karelia) in the north to the Balkan Peninsula and Asia Minor in the south, also including the Middle Urals and West Siberia. *M. arvalis* (sensu stricto) has been found in the Transcaucasian region and Mongolia (Malygin, 1983; Baranovski et al., 1994; Malygin and Bashenina, 1994; Meyer et al., 1996).

* E-mail: georgimar@gmail.com

The range of the East European vole, *M. levis*, covers South Finland in the west, Latvia, Belarus, Ukraine, Moldova (Malygin, 1983; Zagorodnyuk et al., 1994), the Danube delta in Romania (Murariu, 1984), and the southern part of the Balkan Peninsula (Bulgaria, Greece) (Kral, 1975; Belcheva et al., 1977), as well as a large area of southern Serbia and Macedonia (Kral, 1975; Petrov et al., 1975; Ruźić et al., 1975; Zivković et al., 1974). Specimens of this species were found in northern Turkey (Doğramacı, 1989), and some isolated populations were found in the Ararat valley in Transcaucasia, as well (Malygin and Orlov, 1975). The species also occurs in the Don River valley (Malygin and Bashenina, 1994; Tikhonova et al., 1999), the Caspian lowland, around the Volga River outfall, and in the Urals (Malygin, 1983; Baranovski et al., 1994; Tikhonov et al., 1996).

The main part of the range of the East European vole thus covers the central part of the distribution area of the sibling species *M. arvalis* (sensu stricto) (Malygin and Bashenina, 1994), where the 2 species are distributed sympatrically. The data obtained in recent investigations of the karyotype and cranial morphology of the sibling species *M. arvalis* and *M. levis* added new locations to the distribution map of the East European vole in Europe (Suchentrunk et al., 1998; Mazeikyte et al., 1999; Kalcheva and Topashka-Ancheva, 2005; Markov and Kocheva, 2008) and Asia Minor (Yiğit et al., 2007), manifesting an expansion of *M. levis* presence in Eurasia.

The territory of Hungary is within the boundaries of the sympatric distribution of the 2 sibling species (from 40°N to 60°N). In the second half of the 20th century, when the greatest number of grey vole skulls were collected and lodged in the zoological collections of many countries in central and eastern Europe, the species determination of the sibling species through classical morphological features depended very much on the chosen methods of diagnostics and was very unreliable; for this reason, all of the grey vole skulls collected in this period were assigned to *M. arvalis* (sensu lato). Because of this approach, the huge amount of material collected in that period was not used properly to outline in detail the sympatric occurrence of the 2 species in Europe.

A new opportunity for species determination of massive craniological material was afforded by the morphological-craniometrical key for differentiation of the grey voles' sibling species from Europe (Markov and Kocheva, 2007), and this predetermined the aims of the present work as follows: 1) to carry out revision of skull specimens of the common vole (*Microtus arvalis* Pallas sensu lato) from the mammal collection of the Hungarian Natural History Museum in Budapest and to ascertain if *M. levis* is present among the Hungarian mammal fauna; 2) if it is in fact present, to define the regions with sympatric distribution of the studied sibling vole species in the country; and 3) to analyze the craniometrical characters of craniologically identified individuals of the common vole *M. arvalis* and the East European vole *M. levis* from the studied territory of their sympatric presence in Hungary.

Materials and methods

The study was based on 245 adult skull specimens of the common vole (*Microtus arvalis* sensu lato) kept in the mammal collection of the Hungarian Natural History Museum in Budapest. The localities examined covered the largest part of the presumable species habitats of the common vole (*Microtus arvalis* Pallas sensu lato) in Hungary and were grouped topographically into 14 regions: Bács-Kiskun (studied specimens n = 21), Borsod-Abaúj-Zemplén (n = 21), Fejér (n = 12), Györ-Moson-Sporon (n = 18), Hajdú-Bihar (n = 21), Heves (n = 17), Komárom-Esztergom (n = 10), Pest (n = 22), Somogy (n = 17), Szabolcs-Szatmár-Bereg (n = 13), Jasz-Nagykun-Szolnok (n = 22), Vas (n = 22), Veszprém (n = 22), and Tolna (n = 6). The studied localities differ significantly in terms of their physiographic conditions. The age of the specimens was determined according to the craniological criteria of Bashenina (1953).

The studied specimens were classified as either *M. arvalis* or *M. levis* using a discriminating craniological key (Markov and Kocheva, 2007). The discriminant function included a set of 4 variables that did not show sexual dimorphism in the studied species:

$$Y = 2.099 \times \text{length of incisive foramen} - 3.678 \times \text{alveolar length of upper molar series} + 7.433 \times (\text{zygomatic width} / \text{skull height between bullae osseae}) - 2.830.$$

An unclassified specimen would be assigned to one of the sibling species, *M. levis* or *M. arvalis*, after completion of the following steps:

1. Calculation of the canonical discriminant function, using the real values of the included craniometric characters of the analyzed specimen;
2. Comparison of the value of the calculated canonical discriminant function with the values of group centroids (0.803 for *M. arvalis* and –2.71 for *M. levis*); and
3. Assigning of the analyzed individual to that species with the centroid value closest to the calculated centroid value.

Further craniometrical analysis included craniologically differentiated specimens of both species of the sibling voles in Hungary: 16 specimens (7 males and 9 females) of *M. arvalis* and 15 specimens (8 males and 7 females) of *M. levis*. It was based on 25 cranial characters (Figure 1) taken from each specimen with the help of a digital caliper with 0.1-mm accuracy as follows: V1 – condylobasal

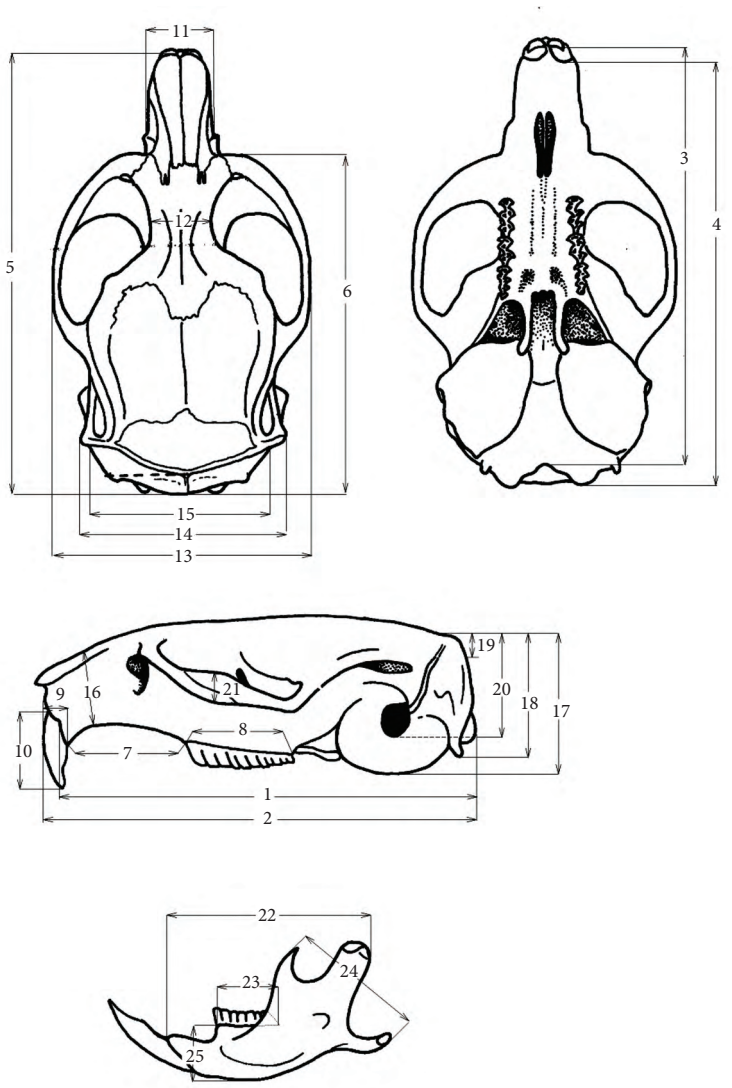

Figure 1. Studied cranial characters of males and females of the vole sibling species *M. arvalis* and *M. levis* in Hungary. A description of these characters is given in the "materials and methods" section.

length I; V2 – condylobasal length II; V3 – basal length; V4 – condylobasilar length; V5 – total skull length; V6 – occipitomaxillar length; V7 – length of upper diastema; V8 – alveolar length of upper molar series; V9 – thickness of upper incisives; V10 – length of upper incisives; V11 – rostrum width; V12 – interorbital width; V13 – zygomatic width; V14 – occipital width; V15 – mastoid width; V16 – rostrum height; V17 – cranial height at bullae osseae; V18 – cranial height at mastoids; V19 – interparietal-foramen magnum height; V20 – skull height between bullae osseae; V21 – height of zygomatic arch; V22 – mandible length; V23 – alveolar length of lower molars; 24 – articular height; V25 – mandible height (taken at M_2). Although previous investigations did not reveal any sexual dimorphism in these characters in the European range of sibling voles (Markov and Kocheva, 2007), the presence of cranial sexual dimorphism within each species and the differences between the means of the studied metric characters in the Hungarian samples were tested using Student's t-test at $P < 0.05$. All statistical analyses were carried out using the STATISTICA program, version 8.0 (StatSoft, Inc., 2008).

Results

The craniometrical revision of the taxonomic detachment of adult skull specimens of the common vole (*Microtus arvalis* sensu lato) from Hungary revealed the presence of both sibling vole species, *M. levis* and *M. arvalis* (sensu stricto).

In total, 92 of the studied specimens (37.55%) were classified as *M. levis*. The presence of the common vole *M. arvalis* (sensu stricto) in the studied sample was found to be significantly higher, at 153 (62.45%) of the studied skulls.

The established distribution of craniometrically differentiated sibling voles showed that the East European vole, *M. levis*, and the common vole, *M. arvalis*, occurred together in samples from all of the studied regions in the country with different relative predominant presences (Figure 2).

The relative numbers of the 2 species in the studied samples demonstrated that *M. arvalis* predominated in the samples from Vas (*M. levis* – 13.6%, *M. arvalis* – 86.4%), Hajdú-Bihar (*M. levis* – 23.8%, *M. arvalis* – 76.2%), Györ-Moson-Sporon (*M. levis* –

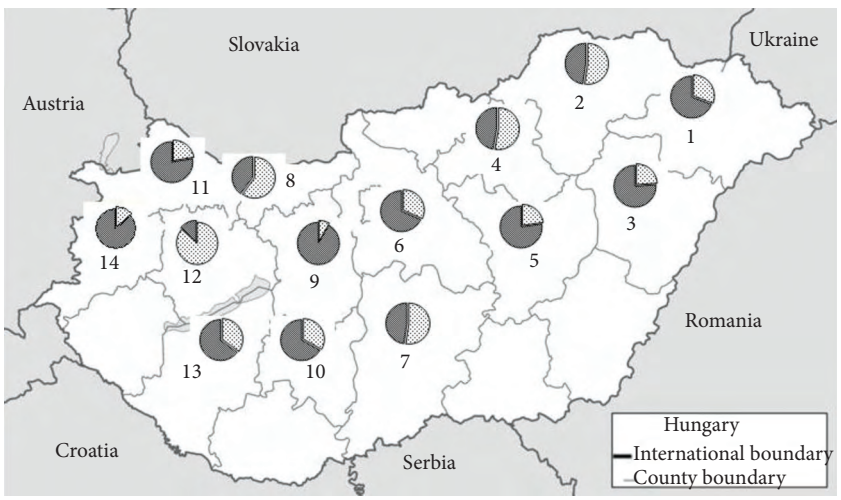

Figure 2. The relative presence (in percentages) of the craniometrically classified East European vole (*M. levis*) (dotted) and the common vole (*M. arvalis* sensu stricto) (hatched) in samples from the studied county regions in Hungary: 1) Szabolcs-Szatmár-Bereg (studied specimens n = 13); 2) Borsod-Abaúj-Zemplén (n = 21); 3) Hajdú-Bihar (n = 21); 4) Heves (n = 17); 5) Jasz-Nagykun-Szolnok (n = 22); 6) Pest (n = 22); 7) Bács-Kiskun (n=21); 8) Komárom-Esztergom (n = 10); 9) Fejér (n = 12); 10) Tolna (n = 6); 11) Györ-Moson-Sporon (n = 18); 12) Veszprém (n = 22); 13) Somogy (n = 17); 14) Vas (n = 22).

22.2%, *M. arvalis* – 77.8%), Pest (*M. levis* – 31.8%, *M. arvalis* 68.2%), Somogy (*M. levis* – 35.3%, *M. arvalis* – 64.7%), Szabolcs-Szatmár-Bereg (*M. levis* – 30.8%, *M. arvalis* 69.2%), Jasz-Nagykun-Szolnok (*M. levis* – 22.7%, *M. arvalis* 77.3%); and Tolna- (*M. levis* – 33.3%, *M. arvalis* – 66.7%), and, in the region of Fejér, the common vole accounted for 91.7% of the specimens presented. Although *M. levis* slightly predominated (at 4.68% higher) in the samples from the Bács-Kiskun region (*M. levis* – 52.34 %, *M. arvalis* – 47.66%), the presence of the 2 species was almost equal there. In the sample from the Borsod-Abaúj-Zemplén region (*M. levis* – 47.6%, *M. arvalis* – 52.4%), the 2 species were also represented comparatively equally; there were only 4.8% more specimens of *M. arvalis* than of *M. levis*. The regions with a higher relative presence of *M. levis* were Heves (*M. levis* – 52.9%, *M. arvalis* – 47.1%), Komárom-Esztergom (*M. levis* - 60%, *M. arvalis* – 40 %), and Veszprém (*M. levis* – 86.4%, *M. arvalis* – 13.6%); the highest predominance of *M. levis* over *M. arvalis* was found in the studied sample from Veszprém (86.4%).

The comparative detailed analysis (Student's t-test at $P < 0.05$) of mean values of the craniometrical characteristics and their absolute variation in the 2 morphologically sibling vole species (Table) confirmed the absence of statistically significant differences between the mean values of all studied craniometrical characters of the males and females in each species; that the specimens of both sexes could thus be pooled together to deduce the basic descriptive statistics of cranial measurements of the 2 species from Hungary; and that the mean values of respective craniometrical characters were very close and showed similar absolute variability.

Discussion

The craniometrical revision of the collection of skull specimens from the common vole *Microtus arvalis* Pallas (sensu lato) resulted in their differentiation as *M. arvalis* or *M. levis*; thus, the presence of *M. levis* in Hungarian mammal fauna was made manifest.

The high degree of craniometrical similarity found in the sibling voles showed that only multiple examinations of a relatively large number of craniometrical characters could enable the craniological differentiation of these 2 species in Hungary.

The fact that craniometrical examination revealed *M. levis* specimens in Hungary supports recent ideas about the distribution of *M. levis* in Europe (Zima 1999; Shenbrot and Krasnov, 2005) by extending the borders of its range into the west. At the same time, it is consistent with the generally accepted idea of *M. levis* as a widespread lowland species, tolerant to a wide range of habitat types (Zagorodnyuk et al., 2008).

The established craniometrical classification of the studied skulls of *Microtus arvalis* Pallas (sensu lato) from Hungary encourage the following conclusions: the 2 morphologically sibling voles are not equally present in most of the studied localities, differing by their physiographical characteristics; there were significantly fewer specimens of *M. levis* than of *M. arvalis* found in the western part of the country; the 2 species were both present in the central and eastern part of the country, with an overall preponderance of *M. arvalis*; and in several regions of the northern part of the country, *M. levis* dominated over *M. arvalis*.

Mosaic distribution of *M. levis* in Hungary suggests that further detailed investigation is necessary to determine the sympatric distribution of *M. levis* and *M. arvalis* with regard to their biotopic preferences. Namely, the East European vole prefers biotopes with tree stands and more humid or anthropogenized biotopes, such as vegetable gardens, undergrowths of large-stalk grass, or landmarks (Malygin, 1970, 1983; Dobrokhotov et al., 1985; Tikhonov et al., 1998); and the common vole prefers meadow-type coenoses and agricultural areas (Dobrokhotov et al., 1985; Teslenko and Zagorodnyuk, 1986; Tikhonov et al., 1992; Karaseva et al., 1994) and inhabits poorly timbered forest biotopes in river valleys, forest belts, and gorges in the zone of deciduous forests and forest steppes, avoiding strongly anthropogenized and transformed territories (Tikhonov et al., 1992; Tikhonov et al., 1998; Tikhonov and Tikhonova, 1997). The application of this approach together with the tentative data about the sympatric distribution of the 2 sibling species in Hungary obtained by craniological investigation, supported by the results of cytotaxonomic and biochemical-genetic analyses confirming unambiguously their species

Table. Basic descriptive statistics (mean and SD) of cranial measurements (mm) and estimations of their sexual dimorphism (t-test value; P < 0.05) of the sibling species *M. arvalis* and *M. levis* in Hungary. Variables without sexual dimorphism found are marked with an asterisk. A description of the variables is given in the "materials and methods" section and in Figure 1.

Variables	*Microtus arvalis*								*Microtus levis*							
	Males (n = 7)		Females (n = 9)		Student's test		All (n = 16)		Males (n = 7)		Females (n = 8)		Student's test		All (n = 15)	
	Mean	SD	Mean	SD	t	P	Mean	SD	Mean	SD	Mean	SD	t	P	Mean	SD
V1*	21.729	1.485	21.667	0.951	0.102	0.921	21.694	1.169	20.537	1.195	21.362	1.017	1.446	0.172	20.977	1.145
V2*	22.373	1.392	22.300	1.380	0.104	0.918	22.332	1.339	21.214	1.618	22.013	1.115	1.125	0.281	21.640	1.383
V3*	21.528	1.302	21.444	1.190	0.135	0.895	21.481	1.198	20.057	1.494	20.925	1.050	1.316	0.211	20.520	1.307
V4*	22.043	1.094	22.133	0.987	–0.174	0.865	22.094	1.000	20.714	1.578	21.600	1.488	1.118	0.284	21.187	1.544
V5*	22.743	1.074	23.056	0.932	–0.624	0.543	22.919	0.974	21.857	1.465	22.550	1.020	1,075	0,302	22.227	1.252
V6*	17.286	0.762	17.500	0.800	–0.542	0.596	17.406	0.765	16.729	0.991	17.425	0.860	1.458	0.169	17.100	0.959
V7*	7.171	0.588	7.144	0.424	0.107	0.916	7.156	0.484	6.714	0.478	7.075	0.440	1.523	0.152	6.907	0.479
V8*	5.114	0.248	5.367	0.316	–1.733	0.105	5.256	0.308	5.000	0.370	5.100	0.478	0.448	0.662	5.053	0.419
V9*	1.114	0.121	1.156	0.113	–0.702	0.494	1.138	0.115	1.086	0.195	1.125	0.089	0.514	0.616	1.107	0.144
V10*	3.857	0.439	3.722	0.496	0.566	0.580	3.781	0.462	3.971	0.723	4.125	0.301	0.551	0.591	4.053	0.525
V11*	3.614	0.227	3.656	0.394	–0.246	0.809	3.638	0.322	3.657	0.326	3.738	0.311	0.488	0.634	3.700	0.309
V12*	3.086	0.090	3.122	0.156	–0.549	0.592	3.106	0.129	3.143	0.223	3.313	0.247	1.387	0.189	3.233	0.244
V13*	12.886	0.803	13.002	0.530	–0.350	0.732	12.951	0.641	12.114	0.857	12.438	0.852	0.731	0.478	12.287	0.840
V14*	10.957	0.812	10.767	0.950	0.423	0.679	10.850	0.869	10.829	0.509	10.788	0.582	–0.144	0.887	10.807	0.530
V15*	9.800	0.876	9.589	0.410	0.643	0.531	9.681	0.639	9.100	0.688	9.538	0.437	1.491	0.160	9.333	0.591
V16*	3.928	0.377	4.000	0.394	–0.366	0.720	3.969	0.375	3.871	0.479	3.875	0.260	0.018	0.986	3.873	0.363
V17*	8.485	0.514	8.667	0.550	–0.671	0.513	8.588	0.525	8.300	0.346	8.400	0.262	0.636	0.536	8.353	0.297
V18*	6.143	0.700	6.390	0.639	–0.733	0.475	6.281	0.655	6.357	0.796	6.975	0.468	1.864	0.085	6.687	0.695
V19*	3.886	0.362	3.811	0.352	0.415	0.684	3.844	0.346	3.800	0.416	4.125	0.362	1.619	0.129	3.973	0.410
V20*	6.100	0.723	6.056	0.218	0.176	0.863	6.080	0.485	5.714	0.318	5.825	0.341	0.646	0.529	5.773	0.324
V21*	0.986	0.168	1.000	0.140	–0.185	0.856	0.994	0.148	0.944	0.140	0.950	0.093	0.094	0.926	0.947	0.113
V22*	13.000	0.557	13.267	0.646	–0.868	0.400	13.150	0.604	12.629	1.069	12.938	0.542	0.721	0.484	12.793	0.814
V23*	5.751	0.320	5.900	0.630	–1.252	0.231	5.756	0.530	5.400	0.545	5.775	0.578	1.288	0.220	5.600	0.576
V24*	6.900	0.839	7.144	0.773	–0.605	0.555	7.038	0.785	6.629	0.665	6.938	0.434	1.080	0.300	6.793	0.556
V25*	3.314	0.552	3.100	0.492	0.820	0.426	3.194	0.513	3.229	0.170	3.213	0.223	–0.155	0.879	3.220	0.193

differentiation, would allow the specification of the present distribution and biotopic adherence of *M. levis* in Hungary and the more accurate outlining of the zones of sympatric distribution of the 2 sibling vole species *M. levis* and *M. arvalis* in the country.

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