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DENTAL MORPHOLOGY OF *MICROTUS (TERRICOLA)* VOLES FROM CALABRIA (SOUTHERN ITALY) AND RELATIONSHIPS WITH *M. (T.) SAVII* (RODENTIA, ARVICOLIDAE)

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ABSTRACT - *Microtus (Terricola) savii brachycercus* (von Lehmann, 1961) was described as a Calabrian endemism. More recently, by chromosome evidence, it was considered as a valid species. Comparison of the first lower molar morphometry between Calabrian *Terricola* and *M. savii* populations show small differences. This may be explained by a recent separation of the two groups. Biogeographical, geographical and paleogeographical features of the Calabria, are also showed in order to understand a possible role in ground vole differentiation.

Key words: *Microtus brachycercus, M. savii,* dental morphology, differentiation, South Italy

RIASSUNTO – ***Morfologia dentaria delle arvicole* Microtus (Terricola) *della Calabria (Sud Italia) e relazioni con* M. (T.) savii *(Rodentia, Arvicolidae).*** *Microtus (Terricola) savii brachycercus* (von Lehmann, 1961) fu descritto come un endemismo calabro. Più recentemente, da evidenze cromosomiche, è stato considerato buona specie. Analisi comparative sulla morfometria del primo molare inferiore tra *Terricola* calabresi e popolazioni di *M. savii* mostrano piccole differenze. Ciò può essere spiegato da una segregazione recente dei due gruppi. Sono altresì illustrate le caratteristiche biogeografiche, geografiche e paleogeografiche della Calabria per comprendere un possibile ruolo nella differenziazione delle arvicole sotterranee.

Parole chiave: *Microtus brachycercus, M. savii,* morfologia dentaria, differenziazione, Sud Italia

INTRODUCTION

Microtus (Terricola) savii group, in Italy, can be an interesting model for evolutionary trend studies because of its morphological and genetic variations; the heterogeneous landscape characterising the Italian range also plays an important role. The following sub-species belong to this group: *M. s. savii* (de Sélys Longchamps, 1838) in the mainland, *M. s. nebrodensis* (Minà Palumbo, 1868) in Sicilia, *M. s. tolfetanus* Monti della Tolfa (Roma District) (Contoli, 2003), *M. s. niethammericus*

Gargano promontory (Foggia District) (Contoli, 2003) and the *taxon "brachycercus"* von Lehmann, 1961 endemic to Calabria (Amori *et al.*, 1999; Contoli, 2003), the southern most peninsular Italian region.

Pitymys savii brachycercus was described by von Lehmann (1961) upon seven specimens from La Sila mountains (Cosenza District); its main features are rounded skull, pale fur and very short tail. The holotype is a male trapped at Camigliatello Silano (27.7.1960) and kept in the Museum A. Koenig, Bonn, Germany (cat. N. 60.379).

More recently, karyological studies has been carried out, in order to define clearly the status of Calabrian *Terricola*. A male chromosomes analysis on Rosarno (Cosenza District) and Central Italy specimens (Galleni *et al.*, 1992), shows a giant sexual pair and some difference in bands in the Rosarno karyotypes. For this reason, a fertility reduction in crosses between *M. savii savii* is hypothesised and, in further experiments (Galleni *et al.*, 1994), the males F_1 crosses *savii* x *brachycercus* showed cell degeneration in seminiferous tubules and complete absence of spermatozoa, so that they are sterile and *M. savii brachycercus* should be regarded as a good species. This was confirmed by another study (Galleni *et al.*, 1998) dealing with new analyses on Fiumefreddo (Cosenza District) specimens, identified also as *M. brachycercus*. More recently, a genetic study on cytochrome *b* gene, shows a divergence with *M. savii* of 4-5%, but data set is limited (Jaarola *et al.*, 2004).

On the basis of these items of evidence, we studied, for the first time, the dental morphology of some Calabrian ground vole populations of the subgenus *Terricola* by a metrical method largerly used on these rodents (Brunet-Lecomte, 1988 and 1990; Brunet-Lecomte and Chaline, 1992; Brunet-Lecomte *et al.*, 2001).

As a first work's hypothesis, admitting that all the Calabrian *Terricola* populations belong to *M. brachycercus*, a discriminant analysis, based on the characters that separate Calabrian and *M. savii* populations (see later for details), has been run in order to verifying, on every population from Calabria, the possibility to belong to *M. savii* or *M. brachycercus* (Tab. 1). In this case, the more typical population is the one from Saporito.

Table 1 - Identification of the *Terricola* populations from Calabria after a discriminant analysis with *M. savii* (S). If *M. brachycercus* (B) is really distributed all over Calabria, the population from Saporito B6 turns out to be the best identified. Abreviations: B1 Bisignano, B2 Apollinara, Corigliano, B3 Aspromonte Spiti, B4 San Basile Morano, B5 Belvedere Superiore, B6 Saporito, B7 San Marco Argentano, Pollare.

	B1	B2	B3	B4	B5	B6	B7
B	73.7	79.3	82.3	65.5	65.7	93.0	69.2
S	26.3	20.7	17.7	34.5	34.3	7.0	30.8

MATERIALS AND METHODS

The Calabrian studied specimens belong to the "Microtheriological Calabria Collection", Calabria University, Department of Ecology, Arcavacata of Rende (Cosenza District) (Aloise *et al.*, 1990). They have been found in the localities (Fig. 1) listed in Tab. 2. The teeth have been compared with the those from 11 populations groups of *M. savii* (Tab. 3) and subsequently, a more detailed analysis, has been carried out by comparing the Calabrian populations with single populations of *M. savii* from South Italy (Tab. 4). On the occlusal surface of the first lower molar, the measurements – obtained by using a measuroscope at 1/1000mm precision but approximated at 1/100mm – and the indexes, follow Brunet-Lecomte (1988) and Laplana *et al.* (2000) with some modifications. The 27 measurements (V) are illustrated in Fig. 2 and the calculated indexes are listed in Tab. 5.

The general morphometry of the M_1 was studied by canonical discriminant analysis running on 27 measures taken on the occlusal surface of the tooth. The dental criteria were compared between groups of populations using variance analysis (ANOVA).

Figure 1 - Map of the Calabria region showing the locations of the examied specimens (*), the terra typica of *Pitymys savii brachycercus* (+) and the two stations where specimens with *brachycercus* karyotype (O) were trapped.

Table 2 - Calabrian populations of *Microtus* (*Terricola*) considered in this study.

Code	Locality	District	N. measured teeth
B1	Bisignano	Cosenza	38
B2	Apollinara, Corigliano	Cosenza	29
B3	Aspromonte Spiti	Reggio Calabria	34
B4	San Basile Morano	Cosenza	29
B5	Belvedere Superiore	Catanzaro	35
B6	Saporito	Cosenza	43
B7	San Marco Argentano Pollare	Cosenza	13

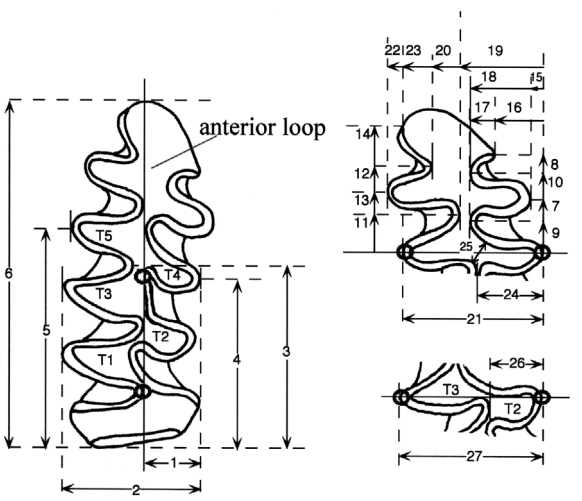

Figure 2 - Measurements (variables “V” in the text) of the first lower molar. T = triangle; T4 and T5 form the pitymyan rhombus.

Table 3 - Populations groups of *Microtus savii* used for a comparison with Calabrian *Terricola*

Subdivision	Region/s	Code	N. measured teeth
Northern Italy	Piemonte	PI	172
	Lombardia	LO	75
	Veneto + 3 Emilia Romagna populations	VE	229
	Emilia Romagna	ER	44
Central Italy	Abruzzo + Toscana + Umbria	AB	173
	Lazio	LA	127
	Campania	C1	122
Southern Italy	Campania	C2	156
	Puglia + Basilicata	P1	123
	Puglia	P2	50
	Sicilia	SI	21

Table 4 - Populations of *M. savii* from South Italy used for a comparison with Calabrian *Terricola.*

Regions	Code	Locality	District	N. measured teeth
Campania	C1	Maiano	Caserta	9
	C2	Frazione Sala, Caserta	Caserta	51
	C3	Capo d'Orso, Maiori	Salerno	6
	C4	Auletta	Salerno	19
	C5	San Giorgio del Sannio	Benevento	60
	C6	Torre del Greco	Napoli	52
	C7	Conza della Campania	Avellino	26
	C8	Ottaviano	Napoli	55
Puglia	P1	Palombara, Monteparano	Taranto	15
	P2	Masseria Colonia, Melissano	Lecce	23
	P3	Alberona	Foggia	42
	P4	North Tavoliere delle Puglie	Foggia	20
	P5	Ostuni, Upper Pleistocene	Brindisi	30
Basilicata	BA	Oasi WWF San Giuliano	Matera	43
Sicilia	S1	Tortorici	Messina	7
	S2	Fontasala	Trapani	8
	S3	Roccapalumba	Palermo	5
	S4	Piana degli Albanesi, Monreale	Palermo	8

Table 5 - List of the calculated indexes (for measurements see Fig. 2).

- relative length of the anterior part, LRPA = (V6-V3)/V6)*100;
- communication between the anterior loop and pitymyan rhombus, V1917 = (V19-V17)/V21*100;
- pitymyan rhombus slope, RP = V4-V3;
- communication between the anterior loop and pitymyan rhombus, V1917 = (V19-V17)/V21*100;
- V1817 = (V18-V17)/V21*100;
- labial-lingual asymmetry at T2-T3, V2627 = (V26/V27)*100;
- anterior loop breadth, BA = (V20-V18)/V21)*100;
- V621 = V6/V21;
- V109 = (V10-V9)/V6*100;
- V1211 = (V12-V11)/V6*100;
- V119 = (V11-V9)/V6*100;
- V1210 = (V12-V10)/V6*100;
- V2019 = (V20-V19)/V21*100.

RESULTS

A first canonical discriminant analysis was running between the 11 geographical groups of *M. savii* and the seven areas from Calabria. The plan 1-2 explains 50% of the variance, with respectively 32% and 18%, and the plan 1-3, that discriminates some the Calabrian populations, explains 44% of the variance, respectively 32% and 12% for the axes 1 and 3 (Fig. 3). This analysis shows that tree Calabrian populations (B1, B2 and B5) are more neighbours to Southern populations of *M. savii* and four Calabrian populations (B3, B4, B6 and B7) are separated from Northern and Southern populations of *M. savii*.

A second analysis is running between the 11 geographical groups of *M. savii* and the seven Calabrian populations pooled. The plan 1-2 explain 55% of the variance, respectively 38% and 17% for the axes 1 and 2 (Fig. 4). This analysis shows that the Calabrian group is closed to the Southern *M. savii* groups.

Finally, a third analysis is running between local Southern populations of *M. savii* and the seven Calabrian populations. The plan 1-3 that explain 33% of the variance, respectively 22% and 11% for the axes 1 and 3 (Fig. 5), and this plan shows that within the Calabrian populations, the Belvedere population is better separated from populations of *M. savii*.

Besides, the ANOVA test shows 11 parameters separating Calabrian *Terricola* and *M. savii* (Tab. 6).

DISCUSSION AND CONCLUSIONS

By considering the obtained data, a separation between Calabrian *Terricola* and *M. savii* from other regions is less clear and, if the specific status of *M. brachycercus* will be confirmed by further genetic studies, this can be explained by a recent speciation from

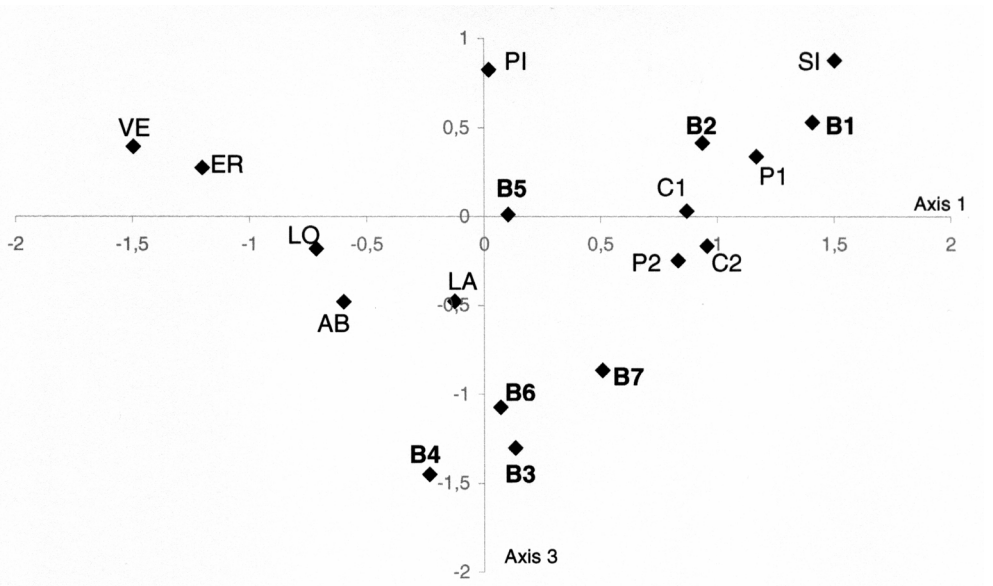

Figure 3 - Position of Calabrian *Terricola* (B) and *M. savii* populations groups centroids on the canonical axes. Abreviations: B1 Bisignano, B2 Apollinara, Corigliano, B3 Aspromonte Spiti, B4 San Basile Morano, B5 Belvedere Superiore, B6 Saporito, B7 San Marco Argentano, Pollare, PI Piemonte, LO Lombardia, VE Veneto + 3 Emilia Romagna populations, ER Emilia Romagna, AB Abruzzo + Toscana + Umbria, LA Lazio, C1 and C2 Campania, P1 Puglia + Basilicata, P2 Puglia, SI Sicilia.

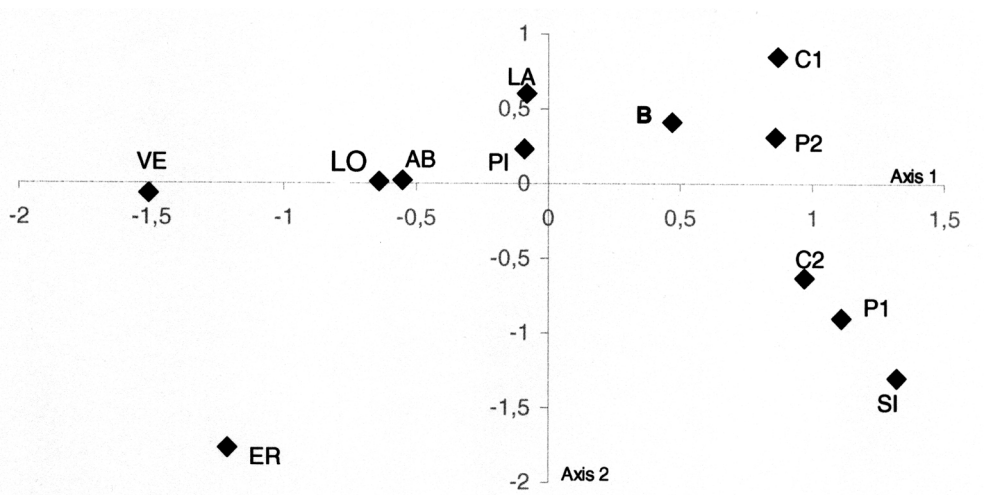

Figure 4 - Position of all Calabrian *Terricola* populations regrouped (B) and *M. savii* populations groups centroids on the canonical axes; for abreviations see Fig. 3 or text.

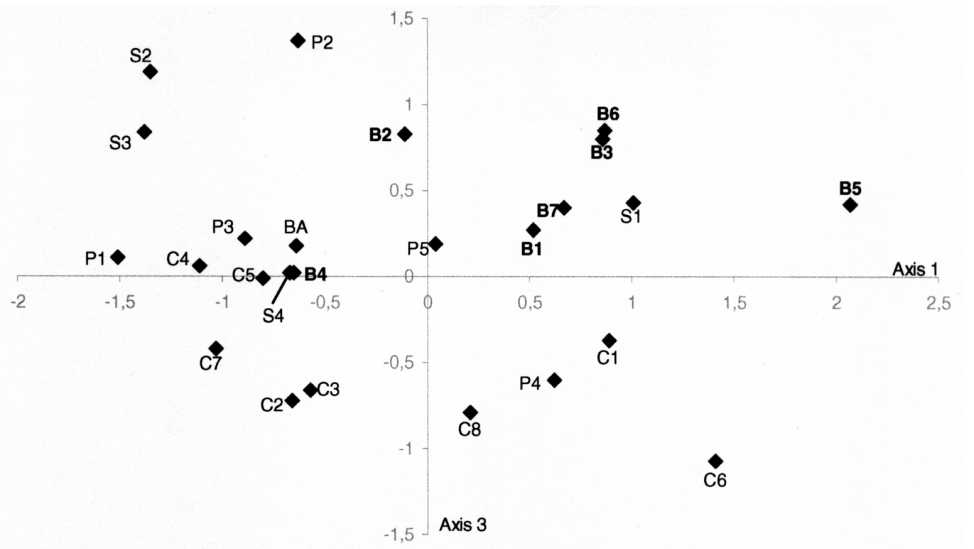

Figure 5 - Position of centroids on the canonical axes of Calabrian *Terricola* (B) and *M. savii* populations from South Italy; for abreviations see Fig. 3 or text.

M. savii, as suggested by Jaarola *et al.* (2004). This study, based on cytochrome *b* variations, shows a less genetic difference that can be included in the normal variability of *M. savii*, but other researches are necessary. Besides, in some Calabrian places: Aspromonte, Apennines and Catanzaro District, von Lehmann (1964; 1973; 1977) found specimens of ground voles ascribed to "*Pitymys savii savii* x *brachycercus*" because their intermediate features and, without a certain evidence of *M. savii* absence in region, a parapatric or sympatric situation between the two species is still possible (Aloise and Contoli, *in litteris*, 08-10-2004).

Nevertheless, considering the biogeography, the geography and the paleogeography of the Calabria, there are good conditions to be realized differentiation processes. At subspecific level, several endemic mammals (Amori *et al.*, 1999) occur: *Sorex antinorii silanus* (Insectivora, Soricidae), *Crocidura suaveolens bruecheri* (Insectivora, Soricidae), *Talpa romana adamoi* (Insectivora, Talpidae), *Clethrionomys glareolus hallucalis*, *C. g. curcio* (Rodentia, Arvicolidae) and another subspecies, *Dryomys nitedula aspromontis* (Rodentia, Gliridae), also occupied a small portion of a bordering region, the Basilicata.

According to Massa (1982), the peninsular effect determines a decrease in species richness along a North-South line but an increase in endemisms, specific or subspecific, along the same gradient. Regarding to rodent occurrences, more particularly, in Southern Italy, the Calabria seems to detach because a species richness (Contoli and Penko, 1996; Contoli, 2000).

During Würm, some glaciers reached Southern ltaly (Malatesta, 1985), so Calabria became a refuge region and, during Middle Pleistocene, its southern

Table 6 - Values of some parameters; NS = not significant, for abbreviation see text.

	Terricola from Calabria (221 teeth)			*Microtus savii* (1351 teeth)			p values
Parameter	Mean	SD	Min-Max	Mean	SD	Min-Max	(ANOVA test)
V6 (mm)	2.58	0.17	2.09 - 3.06	2.57	0.15	2.07 - 3.17	0.4209 NS
LRPA (%)	50.3	1.3	47.1 - 54.1	51.1	1.5	42.3 - 55.1	<0.0001
RP (mm)	-0.043	0.049	-0.137 - 0.403	-0.018	0.040	-0.219 - 0.187	<0.0001
BA (%)	29.7	7.9	13.8 - 58.1	25.7	8.1	3.6 - 57.7	<0.0001
V621 (ratio)	2.51	0.12	2.16 - 2.84	2.6	0.1	2.2 - 3.1	<0.0001
V109 (%)	13.0	1.9	7.9 - 18.1	13.9	1.7	6.1 - 23.6	<0.0001
V119 (%)	0.2	1.3	-3.5 - 4.7	0.3	1.4	-5.1 - 10.5	0.3558 NS
V1210 (%)	2.7	1.7	-4.4 - 9.3	2.2	1.5	-7.5 - 11.8	<0.0001
V1211 (%)	15.5	1.7	11.4 - 22.8	15.8	1.4	6.2 - 21.5	0.0080
V1817 (%)	-8.5	4.9	-24.1 - 4.2	-5.1	5.2	-27.0 -13.8	<0.0001
V1917 (%)	2.7	2.8	-3.7 - 12.4	2.2	2.3	-25.7 - 14.4	0.0043
V2019 (%)	18.5	4.2	9.4 - 31.5	18.4	5.1	3.8 - 45.4	0.6579 NS
V25 (mm)	0.17	0.04	0.07 - 0.28	0.16	0.03	0.06 - 0.27	<0.0001
V2627 (%)	38.3	2.2	30.3 - 48.7	37.8	2.2	29.4 - 55.2	0.0005

portion was separated from the rest of the peninsula, becoming an island with several endemism phenomena (Azzaroli, 1982; Malatesta, 1985). The complex morphology of the territory also played an important role and the ground voles - a group characterized by a very strong evolutionary radiation - were likely subjected to different pressures from the *M. savii* populations of other regions.

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