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A METHOD FOR THE MORPHOMETRIC IDENTIFICATION OF SOUTHERN ITALIAN POPULATIONS OF *APODEMUS* (*SYLVAEMUS*)

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ABSTRACT – The study of morphological and morphometric characteristics of *Apodemus* (*Sylvaemus*) *sylvaticus* and *A. (S.) flavicollis* is examined in this paper. The examined specimens were living in sympatry and allopatry in two Mediterranean habitat-types (a long-trunked forest and a cultivated treed field) in southern Italy, Through discriminant analysis, trends of **skull** measurements (which characterize the different situations of co-presence and/or absence of the two sibling species) are examined. Isometric dental measurements best discriminate the examined populations. Possible causal factors which could affect discriminant measures are discussed.

Key words: Rodents, *Apodemrrs*, morphometry, discriminant analysis, synecology, southern Italy.

RIASSUNTO – *Un metodo per la discriminazione morfometrica in popolazioni di* Apodemus (Sylvaemus) *dell'Italia meridionale* – In questo studio preliminare sono state prese in esame le caratteristiche morfologiche c morfometriche di *Apodemus (Sylvaemus) sylvaticus* e *A. (S.) flavicollis* in condizioni di simpatria e allopatria in due ambienti mediterranei (bosco maturo e campo prossimo ad un frutteto) del sud Italia. Mediante analisi discriminante sono state studiate le tendenze delle misure craniche a caratterizzare i diversi gruppi in situazione di allopatria e/o simpatria delle due specie sorelle. Le misure isometriche dentarie mostrano una maggiore capacità di discriminare i gruppi considerati. Vengono discussi i possibili fattori causali che potrebbero influire sulle misure discriminanti.

Parole chiave: Roditori, *Apodemzts*, morfometria, analisi discriminante, sinecologia, Italia meridionale.

INTRODUCTION

Many European studies (Judes, 1982) have dealt with discrimination criteria for the most common sibling species of wild mice (genus *Apodemus* Kaup, 1829) currently ascribed to the subgenus *Sylvaemus* Ognev, 1929. Present species classification criteria by electrophoresis raise the subgenus *Sylvaemus* to the rank of genus due to the numerous genetic similarities among the Mediterranean species ***Apodemus*** *(Sylvaemus) sylvaticus*, ***A.*** *(S.) flavicollis*, ***A.*** (S.) *hermonensis*, ***A.*** **(S.)** *alpicola*, ***A.*** *(S.) microps* (Filippucci, 1992).

Approximately 95% of *A. (S.) sylvaticus* and ***A.* (S.)** *flavicollis* from central southern Italy can be identified at the specific level through an index (see Amori et al., 1986) obtained by combaining morphologic and morphometric data with very reliable results from electrophoresis analysis (Filippucci et al., 1984). These populations present highly similar morphologic and morphometric characteristics, which makes difficult to distinguish between them, especially when they are sympatric (Niethammer, **1969;** Niethammer and Krapp, 1978; Recco et al., 1978; Cristaldi, 1984; Krapp, 1984; Amori et al., 1984, 1986).

The goal of the present paper is to use an approach allowing for more accurate morphometric discrimination by using univariate and multivariate systems (Cavedon et al., 1990). Furthermore, these methods should allow the identification of phenetic similarities between populations and, possibly, the genetic and ecological conditions that influence them (Amori and Contoli, 1986; Corti and Thorpe, 1989). Amori and Contoli (1986) analysed the patterns of morphometric characters of two sibling species of *Sylvaemus* and divided them into "isometric" and "allometric" with a reciprocal grade of correlation in order to evaluate the possible response to interspecific competition and to other macroenvironmental factors. The authors demonstrated the morphologic changes adaptation response of ***A.*** *sylvaticus* in sympatric situations with its sibling species and its potential increase in size in allopatric conditions. This size increase is typical in cases of isolation (Filippucci et al., 1984; Kotsakis, 1984; Amori and Contoli, 1986) where there are no specialised predators that select by minusvariance the size of small mammals.

Basing upon the method of Demeter and Lazar (1984), we chose the discriminating function analysis because it should enable to establish whether the morphometric characters lead to the identification of the following groups: i) *A. sylvaticus* and ii) *A. flavicollis* living in sympatry with its sibling species, iii) ***A.*** *sylvaticus* living in allopatry; furthermore, it should show how each single character contribute to discrimination. Additionally, this analysis should allow the identification of badly classified individuals, as well as the outliers constituting true exceptions to the assumed control group. After the identification of characters allowing discrimination, it should be possible identify logical combinations of them that can be illustrated in scatter plots (Dulic and Tvrtkovic, 1974) in order to readily identify craniometric characteristics of practical use.

MATERIALS AND METHODS

The study area is located in southrern Italy at the extreme western part of the province of Potenza as described in map nr. 187 **III SW** of the Italian Military Geographic Institute (scale 1:25,000) called "Ricigliano" (geogr.

coord.: 40°45'N, 15°29'E). The mice were captured in two sites separated by torrent "Fiumara di Muro", geologically stable, and are 1.7 km for apart. The first site is located on the eastern slope of Monte Paratiello, near the small village of S. Maria Indorata (469 m a.s.l.), with a mature coppice (CCR), exposed to the North and lying in a gorge grown with oak (*Quercus cerris*), hazelnut (*Corylus avellana*) and chestnut trees (*Castanea sativa)* and with dense underbrush (*Ilex aquifolium, Ruscus aculeatus, Urtica dioica, Pteridium aquilinum*, *Helleborus foetidus, Primula vulgaris*, *Cyclamen* sp., etc.), where **8** examples of *A. sylvaticus* **(WS)** and 12 *A. flavicollis* (WF) were captured in sympatria. The other site is located southeast of the village of Muro Lucano near the small village of Pascone (475 m a.s.l.) with arborated grain fields **(SEM),** a 20% incline, and southwest exposition, where **11** examples of *Apodemus sylvaticus (SS)* were caught.

The samples were classified according to the method of Filippucci et al. (1984) (Tab. 1) by which the length of the incisive foramina is subtracted from the sum of the lengths of the upper tooth row and the length of palatal bridge and the interorbital width to obtain an index >8.0 in *A. flavicollis* and <6.9 in *A. sylvaticus.* For values ranging between these extremes, the table published by Amori et al. (1986) can be considered. This table is based on the position of the tubercules t4 and t7 on M^1 and the possible presence of t9 on M^2 (Pasquier, 1974), as well as the position of the proximal margin of the incisor orifices as compared to the roots of M^1.

A taxonomic evaluation made by electrophoresis on 4 samples from S. Maria Indorata (Tab. 1) showed 5 loci that allowed us to distinguish between the two sibling species (Filippucci, personal communication).

The three age groups **(AGE)** of the samples were established according to the method of **Adamczewska-Andrezejewska** (1967) based on the erosion of the dental tubercules. They were divided into: **I** = juveniles and subadults under 2 months; **II** = adults between **2** and 9 months, III = elderly having more than 9 months.

Table 1 shows the craniometric values made with a Mauser caliber 1/20, as indicated by Cristaldi et al. (1985), done mainly on the left side: INW (interorbital width), PPL (length of palatal bridge), PAL (palatal length), DTL (diasthema length), FIL (incisive foramina length), RSW (rostrum width), ASL (alveolar superior length), AIL (alveolar inferior length), ISW (incisor superior width).

On the basis of these variables, the discriminating analysis was carried out in two successive phases using the statistic package SPSS/PC+ (1986) to compare the following groups first two by two, then all three groups together (Tab. 2): forest *A. sylvaticus* living in sympatry with its cosympatric sibling species *A. flavicollis*, *A. sylvaticus* living in open fields in allopatry in

Tab. 1 – List of the examples of *Apodemus* (*Sylvaemus*) coming from southern Italy (Muro Lucano: MLU). **WS** = ***A. sylvaticus*** sympatric in forest; WF = ***A. flavicollis*** sympatric in forest; SS = A. ***sylvaticus*** allopatric in arborated grain fields; SEX: F = female; M = male; AGE = I, II, III groups (cf. text). For the abbreviations referring to craniometric variables see text. For the morphometric index, the position of the incisor orifices, **the** position of the dental tubercules (t4, t7, **on** M^1; t9 **on** M^2) and the morphologic index cf. Filippucci et al. (1984), Amori et al. (1986). * = samples analyzed by electrophoresis.

SPECIES *Apodemus*	CAPTURE DATE	MLU CODE	MACRO MORPH. INDEX	SEX	AGE	INW	PPL	PAL	DTL	FIL	RSW	ASL	AIL	ISW	MORPH. METR. INDEX	FORINC. POS.	M^1 t4, t7	M^2 t9
sylvaticus	22.12.88	WS 9	s	**M**	II	3.94	4.61	11.37	7.24	5.52	4.31	3.91	4.01	1.79	6.49		t4/t7	t9
sylvaticus	24.12.88	WS 14	(f)	**M**	II	3.96	4.11	10.22	6.11	5.11	3.89	3.92	3.74	1.52	6.88		t4-t7	t9
sylvaticus	27.12.88	WS 35	s	**M**	II	3.82	4.14	10.74	6.84	5.18	4.36	3.99	3.66	1.78	6.77		t4/t7	t9
sylvaticus	31.12.88	WS 39	(f)	**M**	II	3.94	4.08	10.84	6.68	5.42	4.44	3.98	3.74	1.58	6.58		(t4-t7)	t9
sylvaticus	28. 3.89	WS 72	(f)	**M**	II	3.78	4.17	10.50	6.66	5.27	4.04	4.03	3.79	1.68	6.71		t4-t7	t9
sylvaticus	24.12.88	WS 15	(f)	**F**	II	3.74	4.58	11.07	6.82	5.19	4.66	4.12	3.72	1.89	7.25		t4-t7	t9
sylvaticus*	26. 3.89	WS 67	s	F	III	4.01	4.22	10.09	6.86	5.38	3.79	4.04	3.76	1.84	6.89		t4-t7	t9
sylvaticus*	28. 3.89	WS 70	s	F	III	3.94	4.36	11.23	7.56	5.71	4.24	4.02	3.51	1.99	6.61		t4-t7	t9
flavicollis	27.12.88	WF 32	f	M	II	4.12	5.29	12.49	7.91	5.89	5.31	4.46	4.00	2.00	7.98	+	t4-t7	(t9)
flavicollis	5. 2.89	WF 49	f	**M**	II	4.19	4.92	11.11.	6.74	5.19	4.46	4.38	4.06	1.82	8.30	+	t4-t7	(t9)
flavicollis	7 , 2.89	WF 53	f	M	II	4.01	4.69	11.29	7.06	5.53	4.76	4.02	3.90	1.84	7.19	+	t4/t7	(t9)
flavicollis	24.12.88	WF 13	f	F	II	4.09	4.99	11.66	7.14	5.28	5.01	4.32	4.04	1.98	8.12	+	t4-t7	(t9)
flavicollis	25.12.88	WF 31	f	F	II	4.21	4.42	10.72	6.52	5.21	4.72	4.32	3.91	1.72	7.74	0	t4/t7	(t9)
flavicollis	27.12.88	w F 3 4	f	F	II	4.06	4.66	10.82	6.38	5.09	4.78	4.24	3.94	1.75	7.87	+	t4/t7	(t9)
flavicollis	27. 3.89	WF 69	f	F	II	4.02	4.80	10.89	6.45	5.46	4.29	4.50	4.16	1.74	7.86	+	t4/t7	(t9)
flavicollis	28. 3.89	WF 71	f	F	II	4.34	4.72	10.72	6.50	**5.30**	5.05	4.20	3.94	1.92	7.96		t4/t7	(t9)
flavicollis	5. 3.89	WF 58	(f)	**M**	III	4.52	5.12	12.26	7.86	6.00	5.01	4.42	3.86	2.18	8.06	0	t4/t7	(t9)
flavicollis*	18.11.89	WF 218	(f)	**M**	III	4.22	5.20	12.29	7.66	**5.46**	4.92	4.42	3.74	1.89	8.38		eroded	
flavicollis*	18.11.89	WF 219	f	M	III	4.34	5.01	12.81	8.17	5.94	5.41	**4.44**	3.92	2.30	7.85		(t4-t7)	
flavicollis	6. 2.89	WF 50	f	F	III	4.32	4.36	11.64	7.52	6.04	4.34	4.32	3.90	1.98	6.96		eroded	
sylvaticus	1.12.88	SS 4	(f)	F	I	4.00	4.32	10.52	6.48	5.28	3.92	3.84	3.58	1.48	6.88	+	t4-t7	t9
sylvaticus	20.12.88	SS 5	(f)	F	I	3.76	4.14	9.82	5.48	4.48	3.78	3.82	3.62	1.48	7.24		t4-t7	t9
sylvaticus	23.12.88	SS 10	(f)	F	I	3.88	3.94	9.72	5.76	4.84	3.64	3.94	3.52	1.42	6.92		(t4-t7)	t9
sylvaticus	23.12.88	SS 11	(s)	M	II	3.88	4.12	9.99	6.28	4.74	3.99	3.72	3.54	1.68	6.98		t4-t7	t9
syivaticus	27.12.88	SS 36	(s)	**M**	II	4.04	4.02	10.44	6.88	5.44	4.42	3.78	3.68	1.67	6.40		t4/t7	t9
sylvaticus	28. 3.89	SS 74	(s)	**M**	II	3.88	3.82	10.31	6.42	5.48	4.03	3.62	3.66	1.48	5.84		t4-t7	t9
sylvaticus	29. 3.89	SS 77	s	**M**	II	3.76	4.12	10.61	6.84	5.49	4.34	3.74	3.54	1.64	6.13		t4-t7	t9
sylvaticus	29. 3.89	SS 78	s	**M**	II	3.98	4.14	10.96	7.06	5.52	3.98	3.98	3.46	1.58	6.58	+	t4-t7	t9
sylvaticus	22.11.88	SS 2	(f)	**F**	II	4.22	4.52	10.82	6.72	5.18	4.88	4.12	3.64	1.54	7.68		t4-t7	t9
sylvaticus	1.12.88	SS 3	(f)	**F**	II	3.92	4.84	11.32	6.98	5.26	4.06	3.94	3.54	1.66	7.44	+	t4-t7	t9
sylvaticus	23.12.88	SS 12	f	**M**	III	4.12	3.94	10.82	7.16	5.96	4.62	3.74	3.34	1.98	5.84		t4-t7	t9

Tab. 2 – Overview of results from discriminant analysis including two and three groups carried out on craniometric characters. The underlined measurements been a higher discriminating potential on the basis of the pooled within-groups correlation matrices obtained (Tab. **4**). Rel. = reliability (percent of "grouped cases correctly classified). For abbreviations cf. text and Tab. 1.

TWO GROUPS			ALL VARIABLES	ALL VARIABLES
				without AGE, SEX DTL, FIL, ISW:
1	***A. sylvaticus***	WS	**P≤0.0002**	P≤0.0019
	A. sylvaticus	**SS**	**AIL-ISW-ASL**	**AIL-ASL-PPL**
			PPL-INW-AGE	**PAL-INW**
			Rel. 100%	Rel. 100%
				without AGE, SEX:
2	***A. sylvaticus***	WS	**P≤0.00001**	**P≤0.00001**
	A. flavicollis	**WF**	ASL-INW-AGE	AIL-ISW-ASL
			Rel. 95%: exc. WF53	Rel. 100%
				without AGE, SEX DTL, FIL, **ISW:**
3	A. ***flavicollis***	**WF**	P≤0.00001	**P≤0.00001**
	A. *sylvaticus*	**SS**	a-ISW-ASL	**AIL-ASL-RSW**
			Rel. 100%	Rel. 100%
THREE GROUPS				without AGE, SEX DTL, FIL, ISW
4	***A. sylvaticus***	WS	P≤0.0023	P≤0.05
	A. flavicollis	**WF**	**ASL-AIL-AGE-INW**	**ASL-AIL-INW**
	A. sylvaticus	**SS**	Rel. 96,77%: exc. WF53	Rel. 90,32%: exc. WS70, WF53, SS2

comparison to the preceding groups. The sample age could cause an anomaly in the calculations because of the animals growth and thus in the measurements that are reflected in the allometric variations (Voss et al., 1990). A the pooled within-groups correlation matrix, was obtained by averaging the separate covariance matrices for all groups and then computing the correlation matrix. On the basis of this pooled within-groups correlation matrix, in the second phase of discriminating analysis, were excluded those variables whose correlation index in the age factor was close to a reference value of 0.70 (=70%) or above. A summary of the results of this procedure is shown in Table 2. Furthermore, in order to

Tab. 3 – Means and standard variation compared to all the variables of the three population groups (WS, WF, SS).

PARAMETERS	WS $\bar{X}$	WS SD	WF $\bar{X}$	WF SD	SS $\bar{X}$	SS SD
INW	3.89	.10	4.20	.15	3.95	.14
PPL	4.28	.21	4.85	.29	4.17	.29
PAL	10.76	.46	11.56	.75	10.48	.50
DTL	6.85	.43	7.16	.65	6.55	.54
FIL	5.35	.20	5.53	.35	5.24	.42
RSW	4.22	.29	4.84	.35	4.15	.37
ASL	4.00	.07	4.34	.13	3.84	.14
AIL	3.74	.14	3.95	.11	3.56	.10
ISW	1.76	.16	1.93	.18	1.60	.15

estimate the weight of the age factor, the same procedures were applied, excluding the samples from the age class I (Tab. 1: SS4, SS5, SS10), all included in the allopatric *A. sylvaticus* group.

RESULTS

Table 2 summarize the results of the following analyses:

1- Statistical analysis of two groups (*A. sylvaticus* sympatric with its sibling species, against allopatric *A. sylvaticus*).

The means (Tab, 3) show the plusvariance of the first population group (WS) and the widest standard deviation of the second (SS). From the discriminating analysis of all the variables a pooled within-groups correlation matrix was obtained (Tab. 4) which reflects the minor influence of the sex factor (SEX) on the variables, with the partial exclusion of ASL and secondly PPL, and the remarkable influence of age (AGE) on ISW, DTL, FIL. The results in Table 2 were obtained by excluding AGE, SEX, DTL, FIL and ISW from the discriminating analysis. It reflects the significance ($P < 0.0019$) of the following measurements that better characterise the canonic discriminating function: AIL, ASL, PPL, PAL, INW. All individuals were discriminated into their respective groups with 100% correct classification. (Fig, 1).

2 - Statistical analysis of two sympatric groups (*A. sylvaticus* and *A. flavicollis*).

From the mean values of Table 3 we can note the plusvariance of the second group (WF) as compared to the first one (WS). The pooled within-groups correlation matrix obtained from the discriminant analysis applied to two groups shows (Tab. 5) the minor influence of SEX on the variables and a relative influence of AGE (approx. 65%) on the variables

Tab. 4 – **Pooled within-groups correlation matrix of the three considered groups (WS, WF, SS).** The underlined **variables are carried over into the succeeding phase of the analysis (see text). * = correlation values** with age – 70%.

A. sylvaticus WS / *A. sylvaticus* SS

	AGE	SEX	INW	PPL	PAL	DTL	FIL	RSW	ASL	AIL	ISW
AGE	1.00										
SEX	-.17	1.00									
INW	.47	.05	1.0								
PPL	-.40	.52	.10	1.00							
PAL	.34	-.02	.25	.63	1.00						
DTL*	.71	-.14	.41	.34	.82	1.00					
FIL*	.69	-.31	.46	-.04	.62	.84	1.00				
RSW	.37	-.14	.36	.25	.61	.55	.45	1.00			
ASL	-.11	.61	.27	.53	.24	.09	-.16	.20	1.00		
ALL	-.44	-.11	-.04	.19	-.11	-.26	-.28	-.04	-.13	1.00	
ISW*	.76	.01	.18	.19	.46	.71	.55	.45	.06	-.47	1.00

A. sylvaticus WS / *A. flavicollis* WF

	AGE	SEX	INW	PPL	PAL	DTL	FIL	RSW	ASL	AIL	ISW
AGE	1.00										
SEX	.05	1.00									
	.64	-.13	1.0								
PPL	.13	-.34	.03	1.00							
PAL	.45	.45	.26	.70	1.00						
DTL	.61	-.33	.39	.58	.92	1.00					
FIL	.66	-.21	.51	26	.71	.84	1.00				
RSW	-.03	-.26	.07	.57	.66	.50	.22	1.00			
ASL	.32	.05	.11	.44	.39	.34	.26	.12	1.00		
AIL	-.56	-.02	-.27	.09	-.21	-.32	-.19	-.21	-.03	1.00	
ISW	.65	-.02	.45	.51	.73	.84	.69	.53	.34	-.29	1.00

A. flavicollis WF / *A. sylvaticus* SS

	AGE	SEX	INW	PPL	PAL	DTL	FIL	RSW	ASL	ATL	ISW
AGE	1.00										
SEX	-.51	1.00									
INW	.56	-.09	1.0								
PPL	.05	-.08	.15	1.00							
PAL	.61	-.44	.41	.65	1.00						
DTL*	.74	-.58	.50	.43	.94	1.00					
FIL*	.72	-.47	.49	.05	.69	.83	1.00				
RSW	.46	-.33	.49	.39	.58	.60	.42	1.00			
ALL	.05	.22	.35	.56	.40	.24	.06	.12	1.00		
AIL	-.54	.29	-.32	.01	-.33	-.40	-.33	-.19	.11	1.00	
ISW*	.75	-.49	.53	.25	.69	.77	.67	.61	.04	-.42	1.00

A. sylvaticus WS / *A. flavicollis* WF / *A. sylvaticus* SS

	AGE	SEX	INW	PPL	PAL	DTL	FIL	RSW	ASL	AIL	ISW
AGE	1.00										
SEX	-.24	1.00									
INW	.56	-.06	1.0								
PPL	.05	.01	.10	1.00							
PAL	.48	-.34	.32	.66	1.00						
DTL*	.71	-.38	.44	.45	.91	1.00					
FIL*	.69	-.35	.48	.08	.67	.83	1.00				
RSW	.29	-.25	.33	.40	.61	.55	.38	1.00			
ASL	.07	.28	.26	.52	.36	.23	.05	.38	1.00		
AIL	-.51	.05	-.22	.09	-.23	-.33	-.27	.15	.01	100	
ISW*	.72	-.19	.41	.31	.65	.78	.64	.54	.10	-.39	1.00

DTL, FIL, ISW, INW. The results are only 95% acceptable because one individual (WF **53**) classified as ***A. flavicollis*** tends to collocate in the other group. From the summary Table 2 obtained by excluding from the analysis only the variables **SEX** and **AGE.** The variables **AIL,** ISW and **ASL** better characterises the canonic discriminating function and the significance among the groups is significant ($P < 0.00001$). All the individuals were discriminated into their respective groups with 100% correct classification (Tab. 2).

3 - Statistic analysis of two groups (sympatric *A. flavicollis* with its allopatric sister species *A. sylvaticus*).

The mean values in Table **3** show the plusvariance of the first population group (WF) as compared to the second *(SS).* **A** pooled within-groups correlation matrix was obtained (Tab. 6) in which **SEX** affected the variables ISW, DTL, FIL much less than did **AGE.** In Table 2, which was obtained by eliminating **AGE, SEX, DTL, FIL** and **ISW** from the analysis, a significance of ($P < 0.00001$) of the following measurements better characterise the canonic discriminating function for: **AIL,** ASL, **RSW. All** the individuals were classified in the respective group with 100% correct classification (Tab. 2).

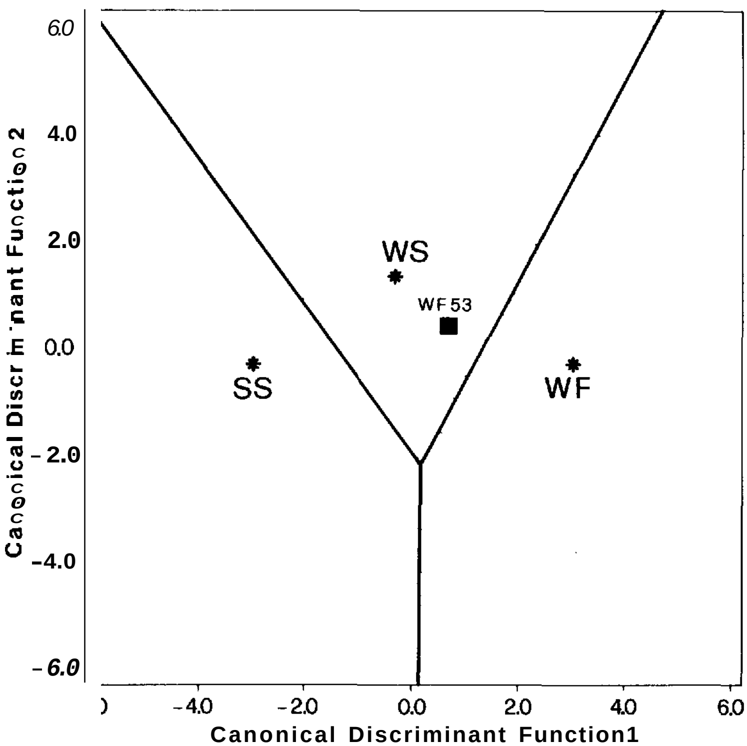


Fig. 1 — All variables scatterplot obtained for three groups by canonical discriminant functions. * = group centroid; exception: WF 53.

Tab. 5 – A posteriori classification obtained by discriminant analysis (see Fig. 1) for the three groups (WS, WF, SS).

ACTUAL GROUP				PREDICTED GROUP MEMBERSHIPS 1	2	3
group	1	*A. sylvaticus*	WS	100.0%	.0%	.0%
group	2	*A. flavicollis*	WF	8.3%	91.7%	.0%
group	3	*A. sylvaticus*	SS	.0%	.0%	100.0%
				percent of "grouped cases correctly classified 96.77%		

Tab. 6 – A posteriori classification obtained by discriminant analysis based on the most significant variables (see Fig. 2) for the three groups (WS, WF, SS).

ACTUAL GROUP				PREDICTED GROUP MEMBERSHIPS 1	2	3
group	1	*A. sylvaticus*	WS	87.5%	.0%	12.5%
group	2	*A. flavicollis*	WF	8.3%	91.7%	.0%
group	3	*A. sylvaticus*	SS	9.1%	.0%	90.9%
				percent of "grouped cases correctly classified 90.32%		

4 - Discriminating analysis of three groups (sympatric ***A.*** *sylvaticus*, sympatric ***A.*** *flavicollis*, allopatric ***A.*** *sylvaticus*).

A pooled within-groups correlation matrix was obtained (Tab. **4)** from the discriminating analysis of the variables in which there was a minor influence of the **SEX** factor on the variables and an influence of **AGE** on the **ISW,** DTL, and FIL measurements. The results are not completely acceptable as the classification of a single example (WF 53: attributed to syrnpatric group ***A.*** *sylvaticus* , which was classified as ***A.*** *flavicollis*) reduced the percentage of correct classification to 96.77% (Tabs. **2,** *5)*. From the summary Table **2** obtained by excluding from the analysis the variables **AGE, SEX,** DTL, FIL and ISW, there is a significance of ($P \leq 0.05$) of the following measurements which better characterises the canonic discriminating function: ASL, AIL, INW. Furthermore, applying the discriminating analysis to three groups with only the three above mentioned variables, an equal reliability of **90.32%** is obtained due to the misclassification of WS 70, WF 53 and SS 2 (Tab. **6;** Fig. **2).** Identical results were obtained by excluding the **SEX** and **AGE** factors and the variables correlated to these (ISW, DTL, FIL) while keeping PPL, PAL, **RSW.** The exclusion of three individuals from the discriminating analysis of the Age **I** group, included in allopatric group ***A.*** *sylvaticus* **(SS)** did not significantly change the results reported above. In the three-group analysis, the procedure tended to lower the reliability percentage. The coordinate diagrams (Figs. 3, 4, 5) summarising the results were effected by combining the measurements characterized by a major discriminating capacity: ASL, AIL, INW. The second (AIL) was furthermore compared with the morphometric index of Filippucci et al. **(1984)** (Fig. **6).** These diagrams show that some individuals were located in the range of the morphologically similar groups. It should be noted, however, that the extreme groups of allopatric *Sylvaemus,* plusvariant (WF) and minusvariant (SS) were never identical.

DISCUSSION

On the basis of our samples the results show that the **SEX** variable has a minor influence on the analysis, except for the higher correlation (61%) obtained with the ASL (Table 4) by comparing populations of *A. sylvaticus*. Generally, the **SEX** factor can be considered a very secondary factor in the craniometric variability of the taxon *Sylvaemus* (see Alcantara et al., 1991).

The age, as a variability factor, could cause an anomaly in the calculations because of the animals' growth and thus in the measurements that are reflected in the allometric variations (Voss et al., 1990).

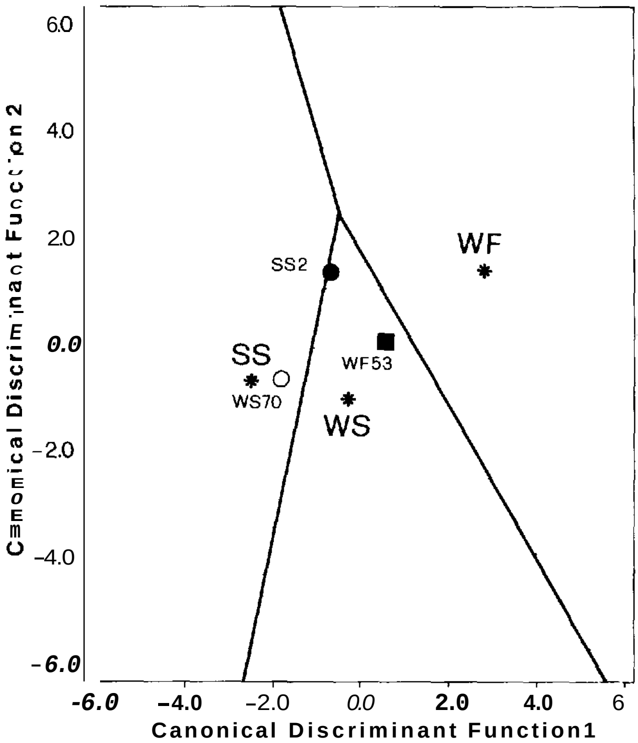


Fig. 2 – Three groups scatterplot of craniometric variables with **highest significance** (cf. **Tab. 2) obtained from Tab. 4 (AIL, ASL, INW), excluding AGE and SEX. * =** group **centroid; exceptions: WS 70, WF 53, SS 2.**

Moreover, **AGE** factor, linked to the variability of the seasonal presence of individuals, contributes to the discrimination of the groups. Eliminating the influence of high values for this factor of variability in the correlation matrix can be an useful strategy for identifying only those measurements that effectively weigh on the characterisation of the populations considered. Its purpose is to identify together with the morphologic factors that are more subject to natural selection, the predation for size, trophic

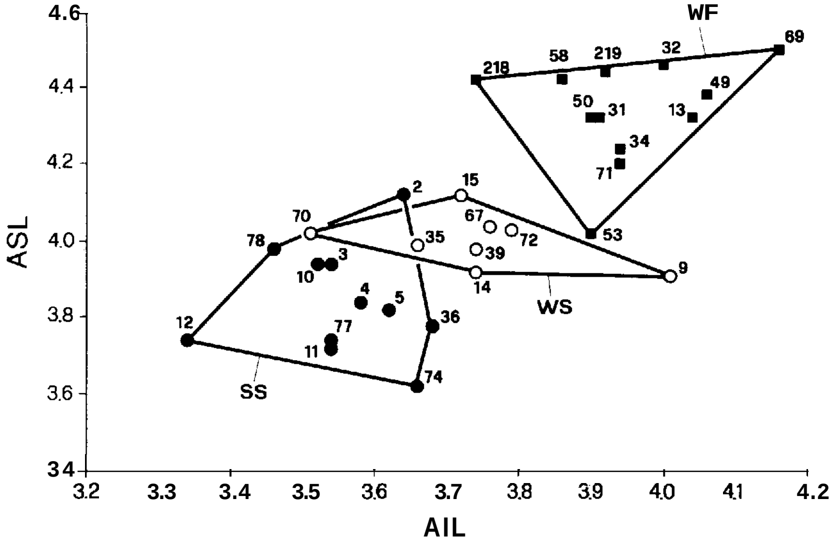


Fig. 3 – Scattergram of all the individuals (individual numbers in Tab. 1) for **the AIL and ASL variables.**

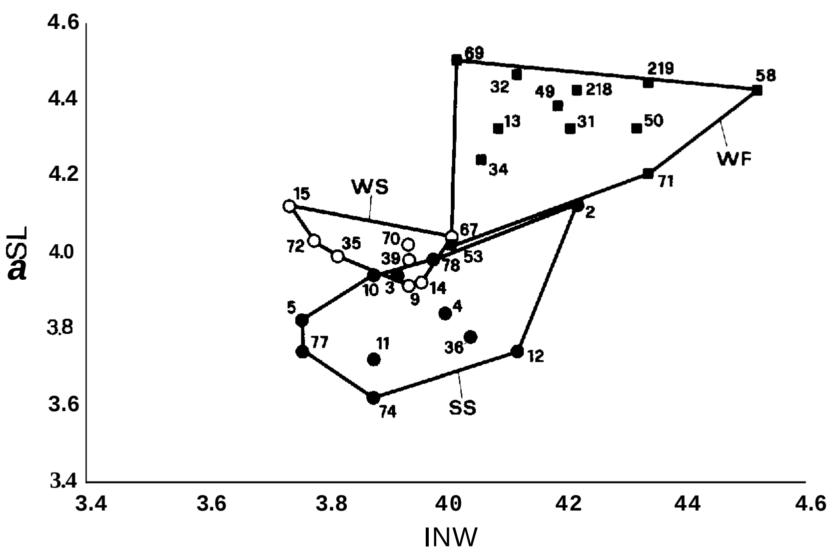


Fig. **4 – Scattergram for all the individuals (individual numbers in Tab. 1)** for **the INW and ASL variables.**

potentials, inter- and intraspecific competition, more or less directly responsible for the morphologic discrimination between conspecific populations. The complex of these factors involves the problem of the morphologic discriminability between populations and between demes within a single species. This phenomenon, as in this case, can be considered as being of even more biologic importance with respect to ability to discriminate between sibling species.

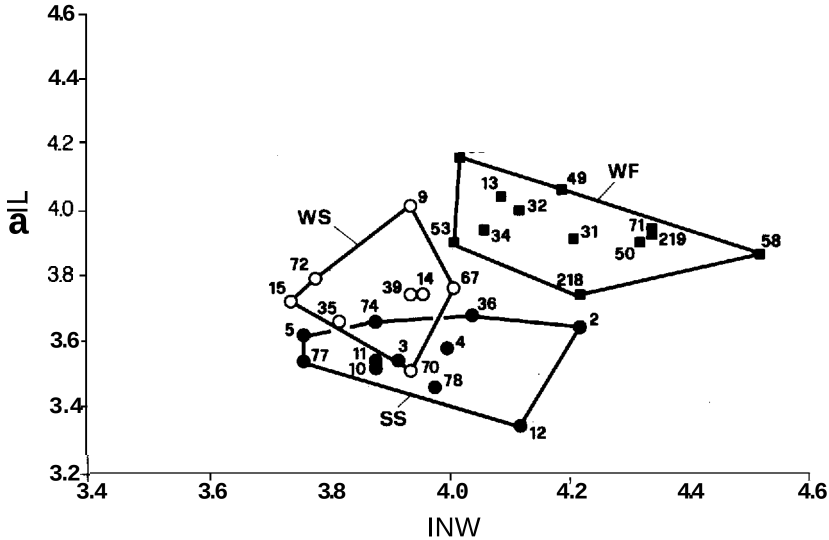
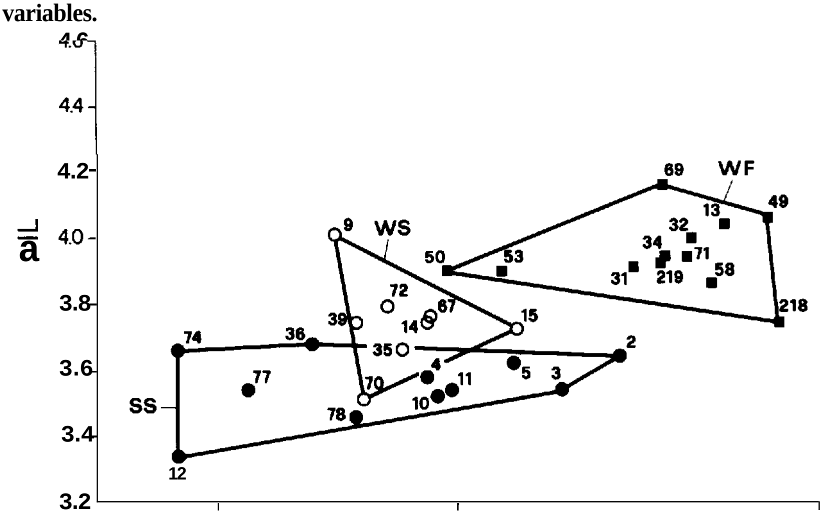


Fig. 5 – Scattergram of all the individuals (individual numbers in Tab. 1) for the INW and AIL variables.



The results of the present biometric analysis, which will be followed by other larger ones on more examples, populations and criteria (Cagnin et al., in prep.), indicate a tendency to minusvariance in allopatric *A. sylvaticus* and a tendency to plusvariant dimensional convergence in sympatric *A. sylvaticus* and *A. flavicollis*. It is not an easy task to explain this pattern in biologic-evolutionary terms. We may consider as an example the case Hagen described in 1954, where two ecotypes of presumed bioclimatic origin within the taxon *Apodemus sylvaticus dichrurus* of eastern Sicily were morphologically discriminable. The Etnean mountain type is larger with more distinct colours, while the one from the plain is smaller and greyer. Such a determination was then disputed first by Kahmann (1956), who attributed such variations to different distributions by age of the samples, then by von Lehmann and Schaefer (1973), who distinguished them first as a species and then correcting themselves, as subspecies (von Lehmann and Schaefer, 1976). Comparing the diagrams on these examples between AIL and ASL, (Fig. 6) with those done by Filippucci et al. (1984) as compared to the conspecific populations in Italy, it was demonstrated that there is a tendency to minusvariance in the range of values typical for *A. flavicollis*, in accordance with the biornetric distribution of the Garganic population, a disposition of the minusvariance of *A. sylvaticus* with a certain concordance with the Etnean population and an intermediate collocation of *A. sylvaticus* in concordance with the populations of Central Italy.

Comparing the means obtained by Amori and Contoli (1986) with those observed, referred to the values of ASL, PPL, FIL, INW, we note that in these, the ASL is generally distributed on the minusvariant values, in particular in the allopatric populations of *A. sylvaticus (SS)*. In general this population is minusvariant not only as compared to the allopatric insular populations (plusvariant for all the variables considered by the authors), but also as compared to the remaining sympatric populations of Italy observed by the authors with the single exception of INW.

In the light of studies done on morphometric characteristics of *Sylvaemus* it is necessary to ask:

- In allopatric *A. sylvaticus* is there a selection according to smaller sizes and age, with a wider standard deviation, which takes place inversely to the tendency assumed by the sympatric populations grouped in plusvariance?
- Are calculations of single craniornetric values sufficient to reveal the consequences of the different predatory pressure exerted on the size and coloration of the wild mice in the diverse environmental situations?
- What could be the influence of food availability and sampling season on the phenotype?
- What role do epidemiologic factors play?

- Can the size and habitus of the two sympatric species be reciprocally influenced by their copresence in forest environments and/or by the peninsular effect that could reduce the genetic flow?

- Can such factors cause a morphologic convergence in the two sympatric species to the extent that in contrast with the principle of the niche incompatibility, it would bring about a reciprocal masking in one single size and a common habitus more adapt to fleeing or dissuading the predator?

- Why do the discriminating dental variables tend to select in minus- or plusvariance even at short geographic distances (in the present study approx. 1.700 km)?

These questions remain for the moment insolved. The discrimination between sister species in the present study can be carried out using only a few key measurements such as ASL, AIL, INW (Tab. 2). From the results obtained keeping the PPL, PAL, RSW, sufficiently correlated to age, it can be shown that the later variables weigh only slightly on the results of the discriminating analysis. Confirming this analysis, the same individuals (WS 70, **WF 53, SS 2),** who lowered the reliability, behave anomalously in the scattergrams (Figs. **3, 4,** 5), including the one in which the index of Filippucci et al. **(1984)** (Fig. 6) was used. On the other hand, to discriminate the single populations of *Apodemus*, it is necessary to adopt discriminating analysis with a greater number of variables. Only through the automatic analysis of data can one deduce the variables that allow us to discriminate both the species and the intraspecific population variants, which oblige us to look for the discriminating variables one by one. We need to distinguish between the tendentially allometric measurements like dentals, and isometric ones subject to major entropy (see Demeter and Lazar, 1984), such as the lengths of the cranial bones. If one succeeds in keeping down the influence of growth (age factor) on the discriminating variables, the combinations that derive from it can represent the "mode" with which the skull with its adaptive characteristics for neurosensorial and feeding interacts with the environment.

The coordination diagrams taken from the significant measurements by the discriminating analysis **(ASL, AIL, TNW)** confirm the validity of the classical criteria used in craniometric analysis.

The identification of an example (WF 53) classifiable as ***A.*** *flavicollis*, having the craniometric characteristics of ***A.*** *sylvaticus*, can be considered an additional stimulus for focusing attention on those individuals considered as being "intermediates" between the two forms (Niethammer, 1969) and posing a problem that already complicated research before electrophoresis was used.

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REFERENCES

ADAMCZEWSKA-ANDRZEJEWSKA, K. A. **1967.** Age reference model for ***Apodemus flavicollis*** (Melchior, **1934).** Ekologia Polska, Ser. A, XV, **41:** 788-790.

ALCANTARA, M., M. DIAZ & F.J.P. PULIDO. **1991.** Variabilidad en las relaciones alométricas entre el peso y las medidas craneales en el raton de campo ***Apodenzus sylvaticus*** L.: efectos sobre su utilidad en estudios de ecologia trofica de aves rapaces. Doñana, Acta Vertebrata, **18 (2): 205-216.**

AMORI, G. & L. CONTOLI. **1986.** Variazioni allometriche ed isometriche in ***Apodemus sylvaticus*** e ***Apodemus flavicollis*** italiani, rispetto alle condizioni di allopatria e simpatria. Hystrix **1 (2): 161-188.**

AMORI, G., M. CRISTALDI & L. CONTOLI. **1986.** Sui Roditori (Gliridae, Arvicolidae, Muridae) dell'Italia peninsulare ed insulare in rapporto all'ambiente bioclimatico mediterraneo. Animalia **(1984), 11 (1-3): 217-269.**

AMORI, G., **R.** FEDERICI & D. PIRAS. **1984.** Utilizzazione di una banca di dati per la distribuzione del sottogenere ***Sylvaemus*** nell'Italia Centro Meridionale. In: "Recenti acquisizioni sul genere ***Apodemus*** in Italia". Suppl. Ricerche di Biologia della Selvaggina. Vol. IX, Ottobre **1984,** Nurnero unico: **65-73.**

CAVEDON, G., M. CRISTALDI & E PASQUALUCCI. **1990.** Tecniche statistiche di analisi multivariata applicate su parametri morfologici rilevati in Roditori selvatici. Hystrix, 2 (n.s.): **59-77.**

CORTI, M. & R.S. THORPE. **1989.** Morphological clines across a karyotypic zone of house mice in central Italy. J. Evol. Biol., 2 **253-264.**

CRISTALDI, M. **1984.** Problematiche di ricerca sui Roditori del genere ***Apodemus*** in Italia. In "Recenti acquisizioni sul genere *Apodemus* in Italia". Suppl. Ricerche Biologia della Selvaggina, Vol. IX, Nurnero unico: **13-23.**

CRISTALDI, M., G. SIMEONE & E. D'ARCANGELO. **1985.** Un approccio biometrico ai processi microevolutivi in *Rattus rattus* (L.). Atti Soc. ital. Sci. nat. Museo civ. Stor. nat. Milano, **126: 3-42.**

DEMETER **A.** & P. **LAZAR. 1984.** Morphometric analysis of field mice *Apodemus*: character selection for routine identification (Mammalia). Ann. Hist.-Nat. Musei Nationalis Hungarici, Budapest, 76: 297-322.

DULIC, B. & **N. TVRTKOVIC. 1974.** Some problems of taxonomy of ***Apodenzus*** *sylvaticus* Linn., **1758** and ***Apodemus flavicollis*** Melchior, **1834** in Yugoslavia. Sep. ex Symp. Theriologicum **II,** Brno **1971: 183-189.**

FILIPPUCCI, M.G. **1992.** Allozyme variation and divergence among European, Middle Eastern, and North African species of the genus *Apodemus* (Rodentia, Muridae). Isr. J. **Zool., 38(3-4): 193-218.**

FILIPPUCCI, M.G., M. CRISTALDI, L. TIZI & L. CONTOLI. **1984.** Dati morfologici **e** morfometrici in popolazioni di *Apodemus* (*Sylvaemus*) dell'Italia Centro-Meridionale determinati elettroforeticarnente. In: "Recenti acquisizioni sul genere *Apodemus* in Italia". Suppl. Ricerche Biologia della Selvaggina, Vol. IX, Numero unico: 85-126.

HAGEN, B. **1984.** Zur kleinsugerfauna Siziliens. Bonn. Zool. Beitr., **1-11** (15): 1-15.

JUDES, U. **1982.** Bibliography of the genus *Apodemus* (Rodentia, Muridae). Mammal Review, **12 (2/3): 59-142.**

KAHMANN, H. 1957. Gibt es zwei gestaltich unterscheidbare okotypen der Waldmaus auf der Insel Siziliens? **Zool. Anz., 159: 153-159.**

KOTSAKIS, T. 1984. Gli ***Apodemus*** (Muridae, Rodentia) fossili delle isole mediterranee. In: "Recenti acquisizioni sul genere ***Apodemus*** in Italia". Suppl. Ricerche Biologia della Selvaggina, Vol. IX, Numero unico: **25-49.**

KRAPP, F. 1984. Gli ***Apodemus*** d'Italia in rapporto alla situazione europea del genere. In: "Recenti acquisizioni sul genere ***Apodemus*** in Italia". Suppl. Ricerche Biologia della Selvaggina, Vol. IX, Numero unico: **51-63.**

von LEHMANN, E. & H.E. SCHAEFER. **1973.** Untersuchungen von Waldmausen ***(Apodemus sylsaticus)*** und Gelbhalsmausen ***(Apodemus flavicollis)*** in Kalabrien und Sizilien. Suppl. Ricerche Zool. Applicata alla Caccia, **5 (6): 175-184.**

von LEHMANN, E. & H.E. SCHAEFER. **1976.** Kreuzungsergebnisse mit Waldmausen (Apodemus) der Insel Sizilien. Saugetierkundl. Mitt., 24 **(3): 180-184.**

NIETHAMMER, J. **1969. Zur** Frage der Introgression bei den Waldmausen ***Apodemus sylsaticus*** und ***Apodemus flavicollis*** (Mamm., Rod.). *Z.* zool. System. Evolut.-forsch., 7: **77-127.**

NIETHAMMER, J. & E KRAPP (Eds.) **1978.** Handbuch der Säugetiere Europas. Band **1,** Rodentia I. Akad. Verlagsgesellschaft, Wiesbaden.

PASQUIER, L. **1974.** Dinamique évolutive d'un sous-genre de Muridae, ***Apodemiis*** (*Sylvaemus*). Etude biometrique des caractères dentaires des populations fossiles et actuelles d'Europe occidentale. Thèse de Doctorat en Paleontologie, Acad. Montpellier: **183** pp.

RECCO, M.A., R. FEDERICI & M. CRISTALDI. **1978.** Presenza simpatrica di ***Apodemus flavicollis*** e ***Apodemus sylsaticus*** nelle zone di Tolfa e Manziana: considerazioni critiche. Boll. Mus. Civ. St. Nat. Verona, **V: 313-353.**

SPSS/PC+. **1986.** Advanced Statistics TM, Copyright **1986** by SPSS Inc.

VOSS, R.S., L.F. MARCUS & P.P. ESCALANTE. 1990. Morphological evolution in muroid rodents. **1.** Conservative patterns of craniometric covariance and their ontogenetic basis in Neotropical genus *Zygodontomys*. Evolution, **44 (6): 1568-1587.**

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