ELSEVIER

Journal of Archaeological Science 35 (2008) 1867–1879

Journal of Archaeological SCIENCE

http://www.elsevier.com/locate/jas

Reappraisal of 'chronospecies' and the use of *Arvicola* (Rodentia, Mammalia) for biochronology

Elise Escudé [a,e,*], Sophie Montuire [b], Emmanuel Desclaux [c], Jean-Pierre Quéré [d], Elodie Renvoisé [e], Marcel Jeannet [f]

[a] *CNRS UMR 6569, Institut de Paléontologie Humaine, 1 rue René Panhard, F-75013 Paris, France*
[b] *CNRS UMR 5561, Biogéosciences & EPHE, Ecole Pratique des Hautes Etudes, Université de Bourgogne, 6 bd Gabriel, F-21000 Dijon, France*
[c] *Laboratoire départemental de Préhistoire du Lazaret, 33bis bd Frank Pilatte, F-06300 Nice, France*
[d] *INRA UMR 1062, CBGP (Centre Biologie et Gestion des Populations), Campus International de Baillarguet, CS 30 016, F-34988 Montferrier/Lez, France*
[e] *CNRS UMR 5561, Biogéosciences, Université de Bourgogne, 6 bd Gabriel, F-21000 Dijon, France*
[f] *ARPA (Association de Recherche Paléoécologique en Archéologie) 2 rue du 19 Mars 1962, F-71850 Charnay-lès-Macon, France*

Received 30 March 2007; received in revised form 13 November 2007; accepted 27 November 2007

Abstract

The water vole, genus *Arvicola*, is characterised by a broad geographic distribution throughout Europe and is widespread during the late Middle and Upper Pleistocene. This genus is used as a major biostratigraphic tool within the Quaternary. Specific determinations using the Schmelzband-Differenzierung-Quotient or SDQ have identified many chronospecies within the fossil species *Arvicola cantiana* (Hinton, 1910). As SDQ calculation remains limited, this study reappraises the *Arvicola* genus in terms of morphodiversity and morphospace using outline analysis which takes into account the tooth as a whole. Outline analysis suggests that one single species of *Arvicola*, *A. cantiana*, was present during the Pleistocene. This species shows great variability with no trends or patterns in morphospace. Thus, these results call into question the reliability of SDQ for specific determinations and throw doubt on the biochronological framework based on *Arvicola*.

Keywords: Rodentia; *Arvicola*; Enamel quotient; Morphology; Outline analysis; Biochronology

1. Introduction

The water vole, genus *Arvicola* (Lacépède, 1799), is characterised by a broad geographic distribution throughout Europe and a wide climatic tolerance, throughout the Quaternary (Shenbrot and Krasnov, 2005). As a result, this genus is present in both glacial and interglacial periods and was widespread during the late Middle and Upper Pleistocene. Its rapid evolution and successive migration waves have encouraged the use of the genus as a major biostratigraphic tool within the Quaternary. *Arvicola* was one of the most important components of the "*short chronology*" theory, based on Mammalian assemblage and expansion (Roebroeks and Kolsfschoten, 1994).

All palaeontological analyses within *Arvicolinae* are based on first lower molars (m1) because m1 have visible diagnostic features and are found in great quantities in the fossil record. In water voles, m1 are continuously growing rootless hypsodont molars, composed of a posterior loop, five closed enamel triangles (or salient angles) and an anterior loop (Chaline, 1972, 1987; Chaline and Laurin, 1986; Hinton, 1926).

In 1955, Heim de Balzac and Guislain (1955) showed the existence in Western Europe of three species in the extant

* Corresponding author. CNRS UMR 5561, Biogéosciences, Université de Bourgogne, 6 bd Gabriel, F-21000 Dijon, France.
E-mail addresses: elise.escude@netcourrier.com (E. Escudé), sophie.montuire@u-bourgogne.fr (S. Montuire), manu@lazaret-unice.fr (E. Desclaux), quere@ensam.inra.fr (J.-P. Quéré), elodie.renvoise@u-bourgogne.fr (M. Renvoisé), m.jeannet.arpa.mf@wanadoo.fr (M. Jeannet).

0305-4403/$ - see front matter © 2008 Elsevier Ltd. All rights reserved.
doi:10.1016/j.jas.2007.11.018

fauna: *Arvicola sapidus* (Miller, 1908) in the north and in the south, sympatric with *Arvicola scherman* (Shaw, 1801), the small Central European species and *Arvicola terrestris* (Linnaeus, 1758), the Eastern and Northern water vole.

Today, two main species are distinguished. *A. sapidus*, the largest in size, is restricted to Spain and France and is completely aquatic. The second extant species, *A. terrestris*, is characterised by a terrestrial lifestyle, and a geographic distribution covering the whole of Europe except Spain. Two ecotypes are defined according to habitat: the aquatic form *Arvicola terrestris terrestris*, and the terrestrial form *Arvicola terrestris scherman* (Le Louarn and Quéré, 2003). These forms are raised to species rank (Wilson and Reeder, 2005): *Arvicola amphibius* (Linnaeus, 1758) for the aquatic form and *Arvicola scherman* (Shaw, 1801) for the terrestrial form. Nevertheless, in Western Europe, morphological and genetic features are not sufficiently established to follow the new nomenclature (Wust-Saucy, 1998). Thus, for our work, *A. terrestris* will be used for the terrestrial form.

1.1. State of the art

Arvicola made its first appearance in Central Europe around 500,000 years ago, and in Eastern Europe around 400,000 years ago (Agadjhanian, 1977, 1983; Radulesco and Samson, 1977). Both the extant species and the fossil, *Arvicola cantiana* (Hinton, 1910), were probably derived from *Mimomys* lineage (*Mimomys occitanus–Mimomys polonicus–Mimomys pliocaenicus–Mimomys ostramosensis–Mimomys savini*) as suggested by the great similarity in molar occlusal surfaces (Chaline, 1972; Fejfar and Heinrich, 1983; Janossy, 1969; Koenigswald von, 1970, 1973, 1980; Kretzoi, 1969; Rabeder, 1981). Thus, the transition from *M. savini* to *A. cantiana*, which corresponds to the Biharian–Toringian boundary in the Cromerian complex, is an important stratigraphic marker (Koenigswald von and Kolfschoten van, 1996).

In *Arvicola*, the enamel band of salient angles underwent differentiation, and m1 length increased during the Quaternary. *A. cantiana* follows the *Mimomys* enamel pattern, the enamel is thicker on the posterior wall and thinner on the anterior wall, while in modern *Arvicola* the enamel is thinner on the posterior wall and thicker on the anterior wall. The thinning of the enamel band on the posterior wall reduces continually, and thus corresponds to an irreversible trend (Heinrich, 1978; Koenigswald von, 1970, 1973, 1980, 1994).

In previous works, enamel differentiation and m1 length have been used for specific determinations, relative dating and correlations of archaeological records (Heinrich, 1978, 1982).

Koenigswald von (1973, 1980, 1994) defined three species and proposed the following stratigraphic division:

- *A. cantiana* in the Middle Pleistocene.
- *A. cantiana/terrestris* in the Middle and Upper Pleistocene.
- *A. terrestris* in the Upper Pleistocene.

Heinrich (1978, 1982, 1987, 1990) traced the evolutionary trend in the *Arvicola* lineage and proposed a biostratigraphic framework based on *Arvicola* with the calculation of the "enamel quotient" (SDQ = Schmelzband-Differenzierung-Quotient). The species partition is *A. cantiana* (SDQ > 100), *A. terrestris* (SDQ < 100) and the transition between the two species coincides approximately with the Eemian–Weichselian boundary.

In the Middle Pleistocene, in the Netherlands and in the Rhine area, Kolfschoten van (1990) and Koenigswald von and Kolfschoten van (1996) distinguished three distinct taxa:

- *Arvicola terrestris cantiana* (SDQ > 120).
- *A. terrestris* ssp. A (120 > SDQ > 95).
- *A. terrestris* ssp. B (SDQ < 95).

In the south-east of France and Liguria, Abbassi and Desclaux (1996) and Abbassi et al. (1998) showed two taxa and proposed a biochronological scheme:

- *Arvicola* morphotype *cantiana/terrestris* (small, and T4–T5 confluent) and *Arvicola* morphotype *cantiana/sapidus* (large, and T4–T5 alternate) in the late Middle Pleistocene (120 < SDQ < 95).
- *A. terrestris* and *A. sapidus* in the Upper Pleistocene (with generally SDQ < 100, except in Italy and Spain, SDQ > 100).

Kolfschoten van (1992), Abbassi et al. (1998) and Desclaux et al. (2000) demonstrated a migration event of *A. terrestris* ssp. B to the Rhone Valley area during the Saalien and the migration of more primitive *A. terrestris* from the south into North-western Europe during the Eemian.

1.2. Aim of our study

However, as SDQ calculation remains limited (Kratochvil, 1980, 1981; Röttger, 1986, 1987), it appeared interesting to suggest a new approach to the genus using a new morphometric method (outline analysis) applied in systematic (Rohlf and Marcus, 1993; Schmittbuhl et al., 1997). At present, these methods are preferred as they provide a general quantitative description of shape and size which allows the study of the object as a whole (Rohlf and Marcus, 1993; Schmittbuhl et al., 1997).

The choice of morphological descriptor depends on the type of object described: geometric morphometry requires a precise determination of homologous landmarks, which are relatively difficult to define and sometimes uncertain on vole molars. This method would result in information loss through less accurate characterisation. For voles, the most appropriate descriptors for the analysis of the first lower molar (with enamel triangle) are linear measurements and outline methods, which allow discrimination between species and are robust for morphospace analysis (Navarro et al., 2004). For our study, we chose outline analysis, which takes into account shape as

a whole, rather than linear measurements which focus on a detailed and specific part of the tooth.

Our study consists of (1) a discussion of the validity and variability of the SDQ method and values and (2) remarks on the reliability of a biochronological framework based on *Arvicola*.

2. Material and methods

The fossil populations chosen for analysis come from France and Liguria. They are listed in Table 1. This material is compared with modern species for morphometric analysis. The present-day populations were obtained by traps in the north and east of France:

- *A. terrestris*, terrestrial form: Valleroy-le-Sec (Vosges, $N = 32$), Chapelle-d'Huin (Doubs, $N = 35$), La Grave (Hautes Alpes, $N = 31$).
- *A. sapidus*: samples ($N = 20$) were collected from all over the Paris Basin as this species is difficult to trap.

The localisation of present populations and fossil localities is given in Fig. 1.

2.1. Outline analysis

The closed outline of each tooth is defined by an empirical periodic function expressing the radius length between outline point and centre of gravity. Each function is resolved into Fourier series for mathematic description which is comparable function to function. Fourier series consist of the approximation of periodic function by a sum of trigonometric functions of decreasing wavelength (Lestrel, 1997; Renaud, 1997).

Vole molars have a complex outline with triangles and loops where the radius from the centre of gravity may cut through the outline several times (i.e. non-holomorph outline). Thus, the co-ordinates x and y of the outline are each expressed by a function $x(t)$ and $y(t)$.

Complex Discrete Fourier Transform, CDFT (Gonzalez and Woods, 1992), allows a complex outline to be treated under the same principle as Elliptic Fourier (Kuhl and Giardina, 1982). The two-dimensional signal is represented by complex number and processed in a single pass.

In order to limit measurement error linked to analysis point of origin (cf maximum of anterior loop), we chose the amplitude of each harmonic as the parameter of analysis.

The external enamel contour of each occlusal surface was hand-drawn then scanned at 300 dpi. For this analysis, 200 equidistant points along the curve were sampled and 20 harmonics were chosen permitting the reconstruction about 97% of the original outline (Fig. 2). The morphospaces are obtained for form (=size and shape) and shape. Shape requires a size-standardisation, using the square root of the area of the occlusal surface. Measurement error for hand drawing was not statistically significant (each tooth has been drawn and scanned several times).

Analyses were performed using MATLAB Toolbox CDFT 2.7 (Dommergues, 2000, 2001), modified by Navarro et al. (2004).

All variables stemming from CDFT were treated by multivariate analysis.

Factorial discriminant analysis (FDA) allows the validation or invalidation of the *a priori* hypothesis for individual classes. This technique aims to maximise inter-group differences and minimise intra-group differences. Mahalanobis distances and associated probabilities indicate the distance between each group and whether the groups are distinct. The statistical level of significance for each discriminant analysis was evaluated by Wilks' Lambda test. For each sample, all individuals were represented by a scatter plot.

Principal component analysis (PCA) is an exploratory technique used to visualise the morphospace to n-dimensions of each species and each population by reducing its dimensionality.

All analyses were performed with *Statistica StatSoft*® (Statsoft).

2.2. Schmelzband-Differenzierung-Quotient (SDQ)

The principle is to quantify the differences in enamel thickness by calculating a quotient (SDQ). In the first lower

Table 1
Fossil populations by archaeological sites, regions, individual number, dating, epoch and bibliographic references

Populations	Sites	Regions	*N*	OIS	Epoch	References
Gigny IX	Gigny cave	Jura	29	3	Upper Pleistocene	Campy (1982), Chaline and Brochet (1989)
Gigny XIX			40	3	Upper Pleistocene	
Gigny XXII			40	4	Upper Pleistocene	
Moula XIV–XV	Moula-Guercy cave	Ardèche	40	5e	Upper Pleistocene	Defleur et al. (1998)
Moula XIX–XVII			30	6	Middle Pleistocene	
Orgnac	Orgnac-3 sinkhole		30	9	Middle Pleistocene	Combier (1967, 1976)
Eglise	Eglise cave	Dordogne	32	3	Upper Pleistocene	Laville et al. (1973), Laville (1973)
Vaufrey	Vaufrey cave		67	6	Middle Pleistocene	Delpech (1988), Marquet (1989), Rigaud (1988)
Artenac	Artenac cave	Charente	37	5	Upper Pleistocene	Delagnes et al. (1999)
Suard	Suard cave		32	5	Upper Pleistocene	Debenath (1974), Marquet (1989)
Gaudry	Gaudry cave		63	6	Middle Pleistocene	Debenath (1974), Lumley de (1989)
Manie	Arma delle Manie	Côte d'azur	22	4	Upper Pleistocene	Arroda et al. (1976), Fornasiero (1989)
Lazaret	Lazaret cave	Liguria	40	6	Middle Pleistocene	Lumley de (1989)

N, individual number; OIS, Oxygen Isotopic Stage.

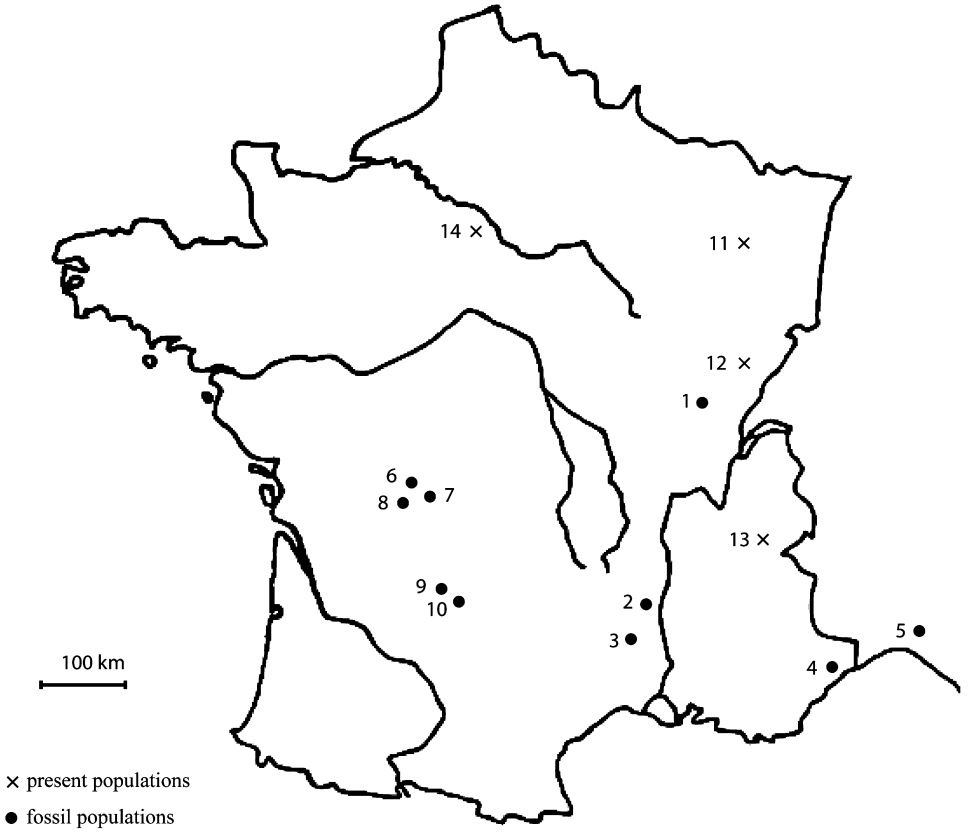

Fig. 1. Map of France and Liguria (Italy) with fossil and present populations of *Arvicola*: 1. Gigny cave, 2. Orgnac-3 sinkhole, 3. Moula-Guercy cave, 4. Lazaret cave, 5. Arma delle Manie, 6. Suard cave, 7. Gaudry cave, 8. Artenac cave, 9. Vaufrey, 10. Eglise, 11. Valleroy-le-Sec, 12. Chapelle-d'Huin, 13. La Grave, 14. The Paris Basin.

molar (m1), the enamel band of each triangle is measured and the thickness of the posterior wall is divided by the thickness of the anterior wall. The SDQ for each m1 is the mean of the SDQ values of each salient angle. The SDQ for a population is the mean of the SDQ of each m1. The landmarks of each measurement are the middle of the enamel band of the anterior and posterior walls (Heinrich, 1978) (Fig. 3).

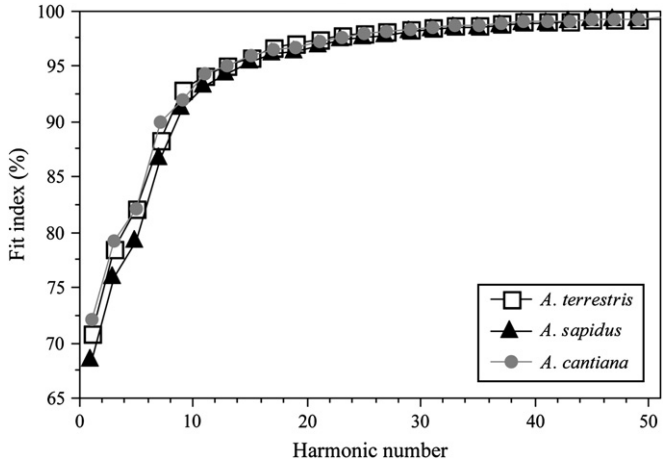

Fig. 2. Convergence index of occlusal surfaces for the 29 first harmonics.

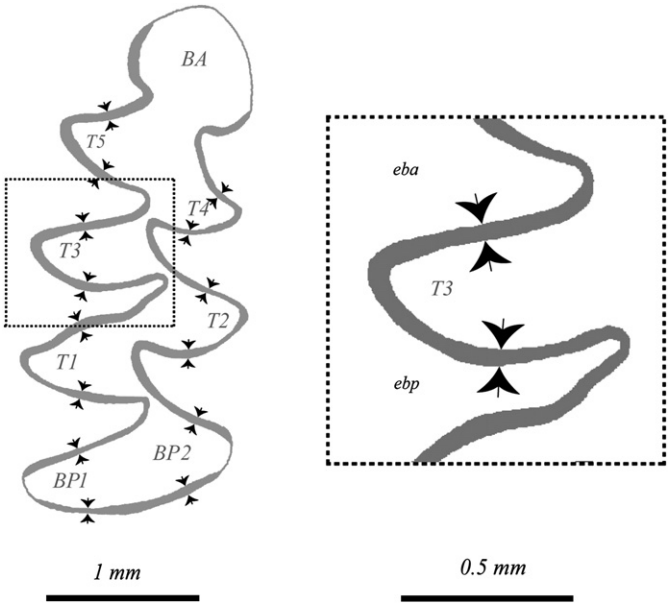

Fig. 3. SDQ calculation. With T: enamel triangle, BA: anterior loop, BP: posterior loop, eba: anterior wall, ebp: posterior wall.

$$\text{SDQ triangle} : \text{SDQ}_t = \frac{\text{ebp}}{\text{eba}} 100$$

$$\text{SDQ m1} : \text{SDQ}_{\text{m1}} = \frac{\sum \text{SDQ}_p}{7} 100$$

$$\text{SDQ population} : \text{SDQ}_{\text{pop}} = \frac{\sum \text{SDQ}_{\text{m1}}}{n} 100$$

SDQ values, morphology and dimensions of m1 are taken into account for specific determinations. As SDQ values decrease with time, the relative stratigraphic position of studied populations can be established in order to construct a biochronological framework.

Enamel thickness measurements are made with a measuring video (colour video camera).

Analysis of variance (ANOVA) is used to test the significance of differences between several means (F test, Snedecor) of SDQ calculation depending on the measurer, measurement series (inter- and intra-measurer variability), between right and left sides, and between individuals (inter- and intra-individual variability). Each ANOVA was performed after verification of distribution normality and variance homogeneity for all samples.

Regression techniques are probabilistic relationships allowing the study of the relationship between SDQ value decrease and time represented by Oxygen Isotopic Stage (OIS 9–1). The linear correlation degree was assessed by the correlation coefficient (R^2).

2.3. Artificial wear and thin-slides

First of all, to examine variation of SDQ values with wear, we reproduce wear artificially in successive steps. This technique was established by Viriot (1994) who showed that the great morphological variability of m1 between individuals of different ages has led to an over-estimation of the number of species. To reproduce the most natural way of chewing, we must consider skull characteristics. In rodents, elongate fosse with no postglenoid process only allows the lower jaw to move forwards and backwards with a pendulum movement, but no lateral movements (Lecointre and Le Guyader, 2003). This movement creates greater wear in the middle of the lower molar row and at the ends of upper molar row. In order to limit error measurement created by failure to reproduce the pendulum movement, only m1 was submitted to this procedure (Viriot, 1994). The following protocol was followed: the molar was fixed to the mobile plate at the same angle as in the alveolar bone of the mandible; the base of the tooth was fixed with ethyl methacrylate resin; the tooth was coated in black epoxy resin, to contrast with the light-coloured

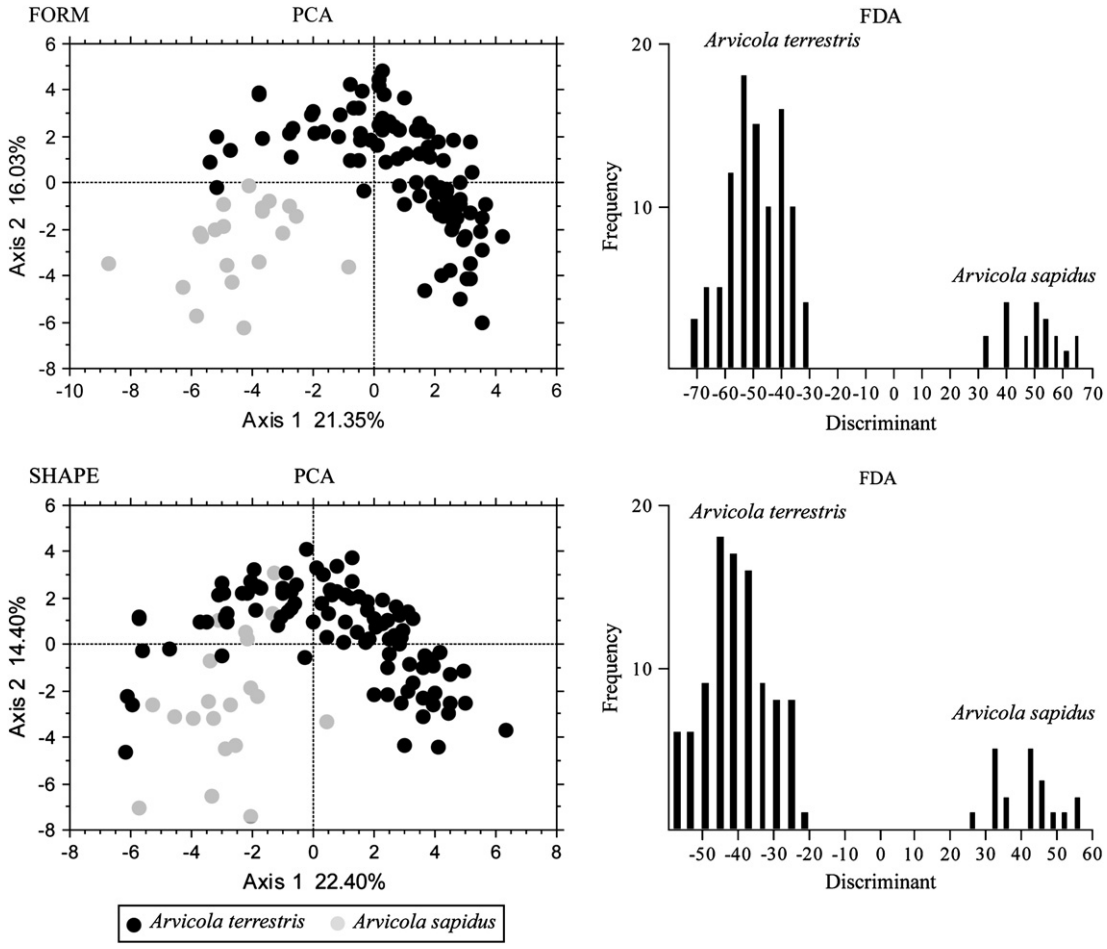

Fig. 4. Current model: *Arvicola terrestris* and *Arvicola sapidus* with principal component analysis (PCA) and factorial discriminant analysis (FDA).

Table 2
Specific determination according to SDQ values (Desclaux et al., 2000)

	Arvicola morphotype *cantiana/terrestris*	*Arvicola* morphotype *cantiana/sapidus*	*Arvicola terrestris*	*Arvicola sapidus*
Gigny IX			+	+
Eglise			+	
Gigny XIX			+	+
Suard			+	
Artenac			+	
Manie				+
Moula XIV–XV				+
Gigny XXII	+			
Moula XIX–XVIII	+	+		
Vaufrey	+	+		
Lazaret	+	+		
Orgnac	+			
Gaudry	+			

mineralised tissues; the tooth was worn down mechanically with a diamond wheel, with a wear step of 0.5 mm; SDQ measurement and calculation for each step (seven steps in all) on a sample of five molars from young individuals of various species.

Afterwards, to observe the variation of enamel band thickness along tooth crowns without wear, two thin-slides were made per tooth from a sample of two young individuals and three adult individuals, various species.

3. Results

3.1. Outline analysis

First of all, it was necessary to establish a current model from individuals as identified by criteria other than their first lower molar (e.g. fur colour, body mass, habitat) so as to verify the efficiency of the chosen specific determination method. Morphospaces of *A. terrestris* and *A. sapidus* are given in Fig. 4 (PCA) for form and shape. The morphospace of *A. sapidus* is separated from *A. terrestris* and the two first axes of the PCA expressed 37.4% (form) and 36.8% (shape) of total variance. To verify the statistical classification, discriminant analyses are performed and presented in Fig. 4 (FDA) for form and shape. All individuals are well classed (form: Wilks' Lambda: $0.0622F$ approx. $(40.77) = 27.592$, $p < 0.000$; shape: Wilks' Lambda: $0.07699F$ approx. $(40.77) = 23.088$, $p < 0.000$). Thus, outline analysis is robust and appropriate to separate *A. terrestris* from *A. sapidus*.

Afterwards, the fossil populations were inserted in the robust present model *A. terrestris*–*A. sapidus* in order to understand relationships between the different species. The groups were established *a priori* according to specific determination extracted from previous works (Desclaux et al., 2000), using SDQ (Table 2). Thus, six groups are distinguished: *Arvicola cantiana terrestris*, *Arvicola cantiana sapidus*, *A. terrestris* fossil, *A. sapidus* fossil, *A. terrestris* and *A. sapidus*. Sixty percent of total individuals are well classed for form and 60.6% for shape (form: Wilks' Lambda: $0.12244F$ approx. $(200.2917) = 7.6722$, $p < 0.000$; shape: Wilks' Lambda: $0.15469F$ approx. $(200.2912) = 6.6378$, $p < 0.000$). The Mahalanobis distances are significant between the centroids of each group (Tables 3 and 4). They are very small between fossil species, small between fossil species and *A. terrestris*. They are large between *A. sapidus* and *A. terrestris*, and *A. sapidus* and fossil species. The first axis permits the separation between *A. sapidus* and *A. terrestris* and fossil species. The second axis permits the separation between *A. sapidus*, *A. terrestris* and fossil species (Figs. 5 and 6). The *A. sapidus* group is spread out with few individuals whereas the *A. terrestris* and *A. cantiana* groups are more compact with many individuals. Taking into account the parameters of form then shape, the fossil group is distinct from the modern species and shows considerable variability within certain limits. There is no separation between species defined by SDQ in previous works: *A. cantiana terrestris*, *A. cantiana sapidus*, *A. terrestris* and *A. sapidus*.

Finally, an attempt was made to analyse changes in form and shape over time. Figs. 7A and 8A allow the morphospaces of each population to be visualised for form and shape over time. About 30–40% of total variance is explained by the first and second component of PCA. Populations are placed depending on their relative chronological position defined by radiometric data and SDQ (Desclaux et al., 2000). Figs. 7B and 8B represent boundary markers (minimum and maximum) of morphospace for the first and second principal component. As illustrated by these figures, we note that there are no significant changes in morphospace over time and that *Arvicola* shows great morphological variability.

3.2. Schmelzband-Differenzierung-Quotient (SDQ)

In a first part, we aim to quantify the error measurement of enamel band thickness and the variability of SDQ calculation between enamel salients of a same m1, along the crown of a same m1 with and without wear, a given individual (between

Table 3
Mahalanobis distance* between centroids of each species scatter plot and associated probability** for form

	A. sapidus fossil	*A. terrestris fossil*	*A. cantiana terrestris*	*A. cantiana sapidus*	*A. sapidus*	*A. terrestris*
A. sapidus fossil		1.80*	2.28	1.65	48.38	7.71
A. terrestris fossil	0.00**		3.12	1.98	51.50	5.94
A. cantiana terrestris	0.00	0.00		2.01	45.13	7.81
A. cantiana sapidus	0.01	0.00	0.00		51.64	8.11
A. sapidus	0.00	0.00	0.00	0.00		57.23
A. terrestris	0.00	0.00	0.00	0.00	0.00	

Table 4
Mahalanobis distance* between centroids of each species scatter plot and associated probability** for shape

	A. sapidus fossil	*A. terrestris fossil*	*A. cantiana terrestris*	*A. cantiana sapidus*	*A. sapidus*	*A. terrestris*
A. sapidus fossil		1.84*	2.40	1.61	37.27	7.91
A. terrestris fossil	0.00**		3.06	1.98	36.37	5.63
A. cantiana terrestris	0.00	0.00		1.97	29.21	7.76
A. cantiana sapidus	0.01	0.00	0.00		34.64	7.95
A. sapidus	0.00	0.00	0.00	0.00		38.81
A. terrestris	0.00	0.00	0.00	0.00	0.00	

right and left side) by measurement series and different measurers. Concerning the first remark, the enamel bands are rarely parallel along the same edge, thus thickness can vary greatly (e.g. in the T4 posterior wall, T4ebp, thickness varied from 56 μm in the middle to 85 and 90 μm at each end). The repetitive measurements of a same enamel band thickness vary greatly (e.g. in the BP1 posterior wall, BP1ebp, the thickness varied from 79 to 88 μm then 100 μm for three measurement series). This is significant for half of the measurements (BP1ebp: $F = 5.64$, $p = 0.004$; BP2eba: $F = 3.60$, $p = 0.030$; T1eba: $F = 8.32$, $p = 0.000$; T2eba: $F = 7.0$, $p = 0.001$; T2ebp: $F = 3.32$, $p = 0.040$; T3eba: $F = 3.39$, $p = 0.380$; T3ebp: $F = 9.22$, $p = 0.000$). Within a same m1 of *Arvicola* type *terrestris*, we note that some enamel salients have "*cantiana* values" and some *Arvicola* type *cantiana* have "*terrestris* values" (e.g. Eglise: BP1 114.3, BP2 98.8, T1 87.1, T2 88.2, T3 92.1, T4 120.3, T5 72.7; Gaudry: BP1 113.8, BP2 111.2, T1 110.4, T2 96.4, T3 112.1, T4 87.2, T5 113.9). Concerning the variability of SDQ calculation along the crown of a same m1 with and without wear (the artificial wear and thin-slides), in both cases (Tables 6 and 7), we observe a great variability in values without trends along tooth crowns in young individuals and in adult individuals. For a given individual, the significant difference of averages between right and left sides seems to indicate a directional asymmetry (right side: Mean 82.3 and left side: Mean 84.6; $F = 8.46$, $p = 0.005$). With three measurement series of the SDQ values for the same population ($N = 30$), the difference between each mean is not significant ($F = 0.738$, $p = 0.481$), but these values for one population (Mean: 83.3, 84.0, 82.5) replaced in a chronological context may change the relative stratigraphic position of this population compared to others. The comparison of previous works with present analyses (Table 5) shows that the relative position of populations changes considerably depending on sampling size and measurer. With differentiation of sampling, Moula XIV–XV (Table 1) value is 100.53 for 15 individuals and 93 for 40 individuals. Within the same sample, Manie average of SDQ values increases from 99.84 to 106.55, Vaufrey average decreases from 104 to 100, Lazaret average decreases from 107.9 to 98.03 and Gaudry decreases from 113.9 to 107.12. The unavailability of previous data does not allow the significance of the difference to be tested using ANOVA. Nevertheless, these differences involve problems for stratigraphic interpretations.

In a second part of our analyses, we attempt to verify if a statistical relationship clearly exits between the decrease in SDQ values and time, using regression techniques. Classically, the decrease in SDQ values in time is used by several authors

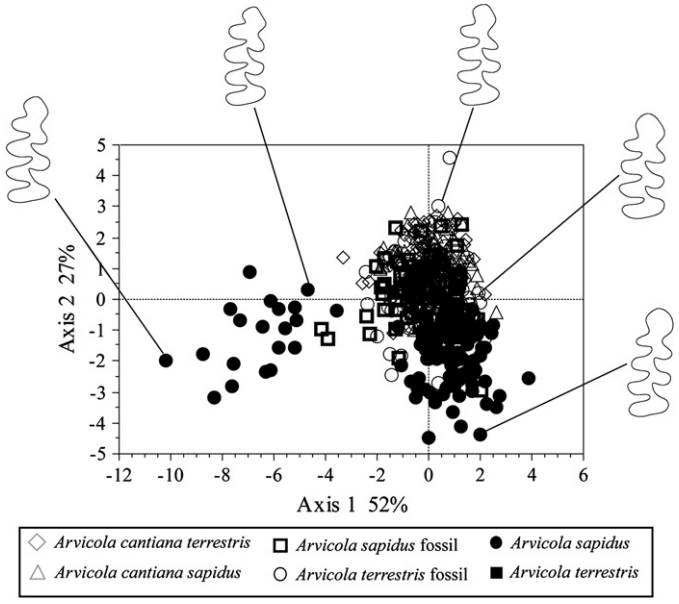

Fig. 5. Discriminant analysis between current and fossil species of *Arvicola* for form.

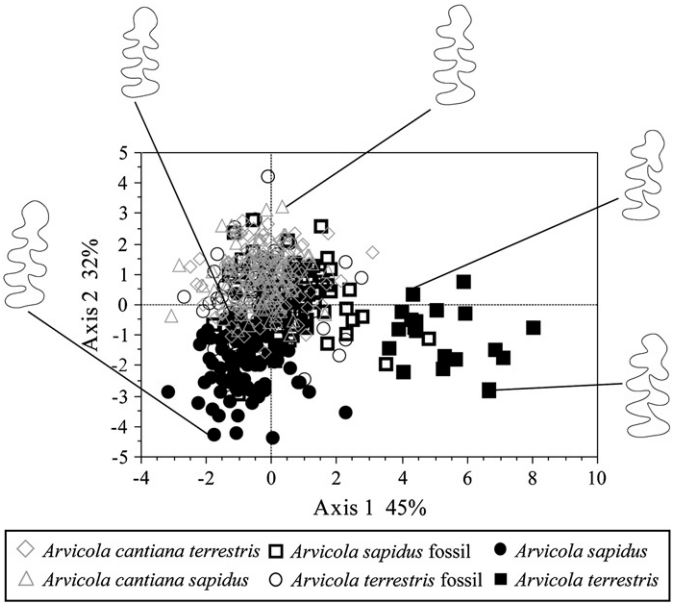

Fig. 6. Discriminant analysis between current and fossil species of *Arvicola* for shape.

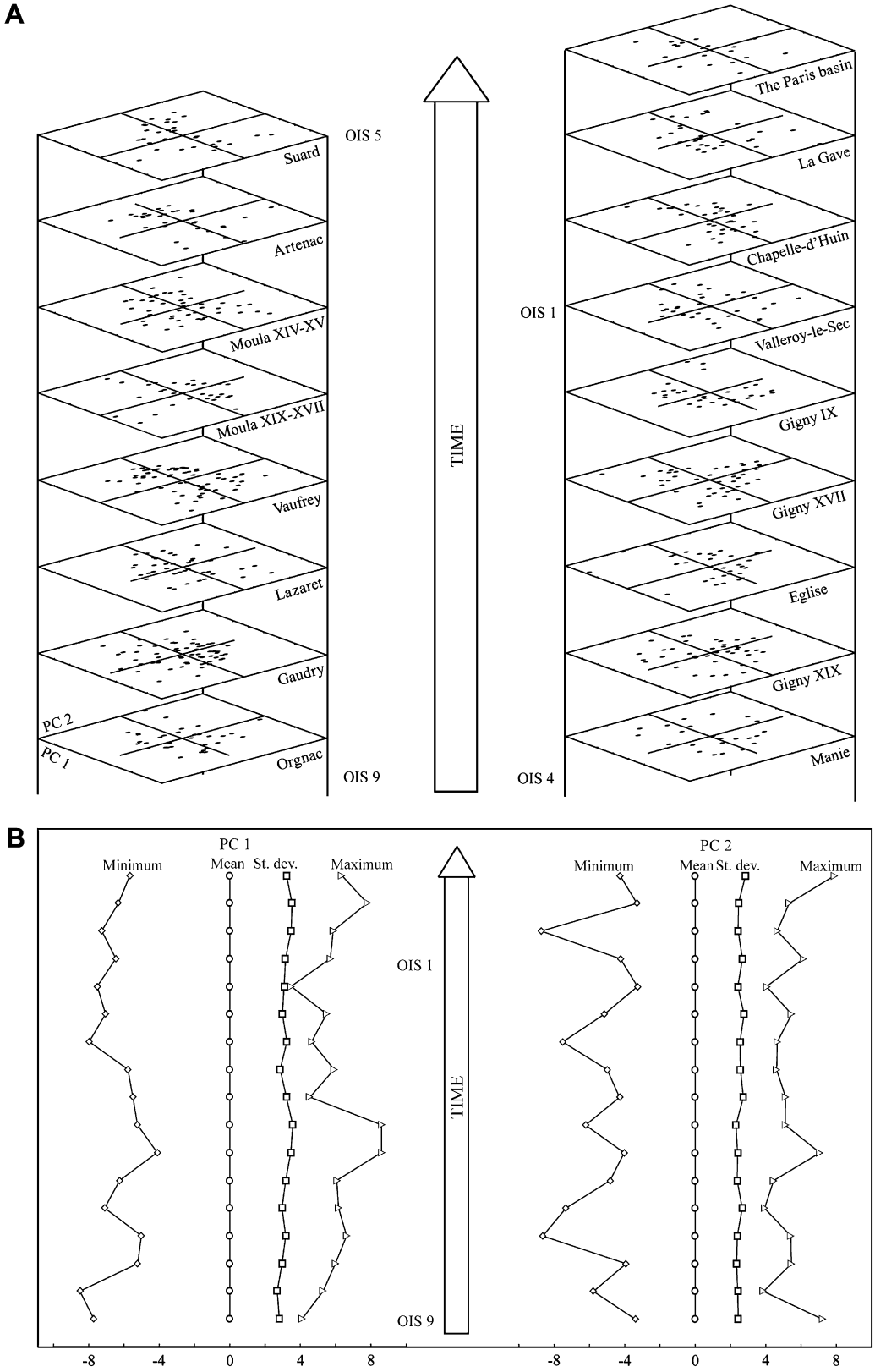

Fig. 7. Visualisation of population morphospaces of *Arvicola* during the time for form with principal component analysis (PCA), St. dev.: standard deviation.

(Abbassi and Desclaux, 1996; Abbassi et al., 1998; Desclaux et al., 2000; Heinrich, 1978, 1982, 1987, 1990; Kolfschoten van, 1990, 1992) to construct a biochronological framework, but this observation has never been statistically tested. Fig. 9 is a scatter diagram of the results of SDQ calculation for 547 individuals in extant and fossil fauna, and the Oxygen Isotopic Stage (OIS 9–1). The relationship between SDQ values and time is low. The correlation coefficient (R^2) reaches 0.165

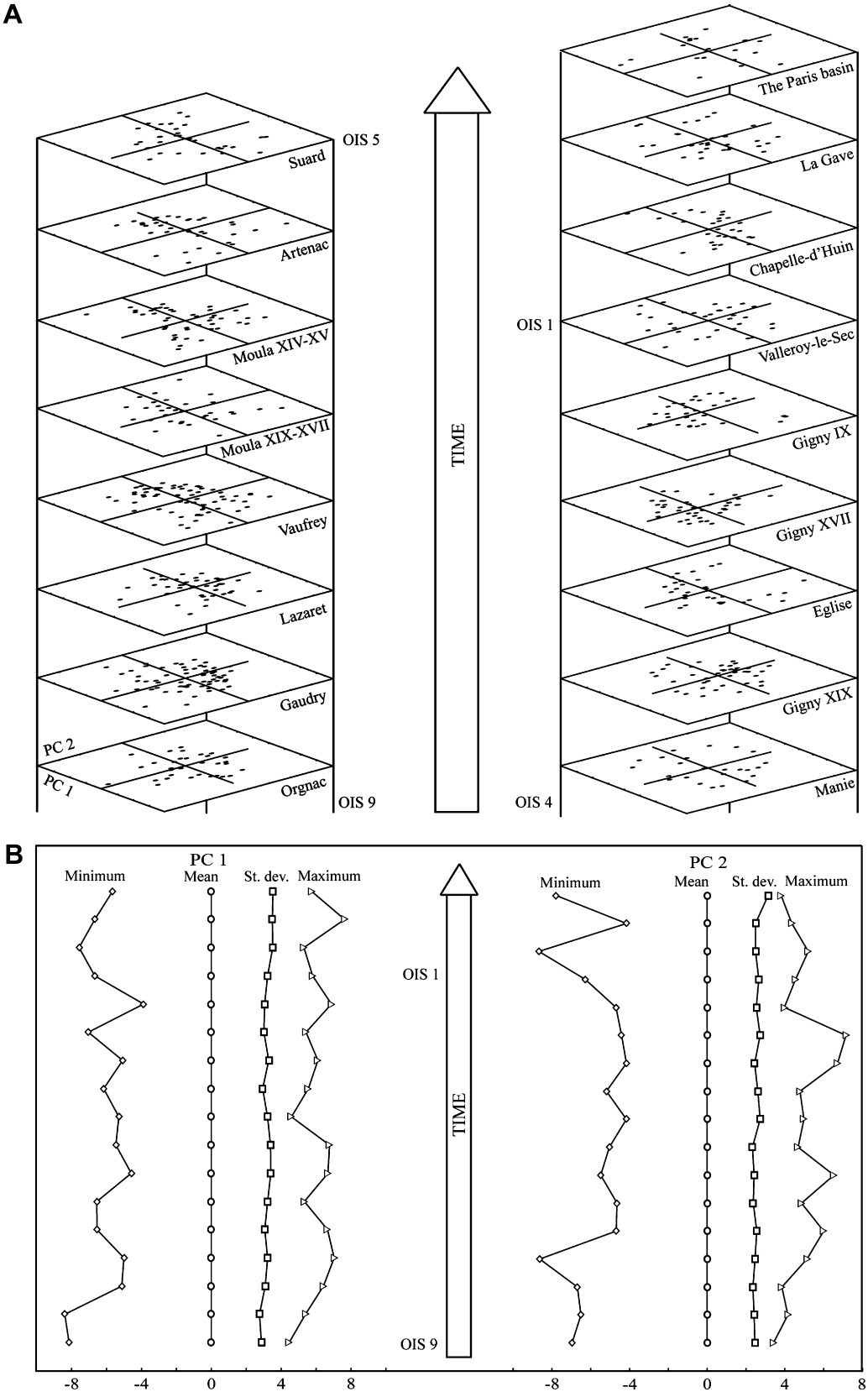

Fig. 8. Visualisation of population morphospaces of *Arvicola* during the time for shape with principal component analysis (PCA), St. dev.: standard deviation.

Table 5
Comparison of previous SDQ values with present study for each population according to different measurers and individual numbers

	Desclaux et al. (2000)			Escudé (present study)		
	N	Min–Max	Mean	*N*	Min–Max	Mean
Eglise	31	68.1–96.0	83.59	32	70.7–84.7	84.78
Gigny XIX	41	79.2–108.9	93.68	40	89.64–109.55	96.25
Gigny XXII	6	88.7–97.3	94.19	40	87.33–103.83	94.34
Manie	25	81.8–119.1	99.84	22	86.9–122.5	106.55
Suard	32	79.5–103.2	91.59	32	67–99	89.1
Artenac	14	80.5–110.8	93.85	37	81–111	93.73
Moula XIV–XV	15	81.8–114.3	100.53	40	73–117	93
Moula XIX–XVIII	31	74.5–106.1	87.41	30	76.24–122.56	87.9
Vaufrey	31	87.3–131	104	37	80.7–127.6	100.1
Lazaret	47	90.5–127.2	107.9	40	66.26–109.65	98.03
Gaudry	64	98–138.7	113.9	63	96–118	107.12

($p < 0.0001$). Only 40% of the variation of SDQ values can be explained by time. Thus, although the regression is significant, there is a high error risk in estimating the relative stratigraphic position of near populations.

4. Discussion

Our study suggests that perhaps only one species of *Arvicola*, *A. cantiana*, was present during the Pleistocene. This species shows great phenotypic variability in which it is difficult to show trends or patterns in morphospace. Therefore indications of glacial or interglacial periods and migration waves (Desclaux et al., 2000; Kolfschoten van, 1990, 1992) are not obvious.

It is necessary to remember that, in previous works, SDQ calculation has led to the description of subspecies, morphotypes and migration waves. SDQ also has been used in the construction of a biochronological framework based on *Arvicola* in the Quaternary. Nevertheless, some works show that the great variation in SDQ values can induce problems for specific determinations and biochronological interpretations. Kratochvil (1980, 1981) and Röttger (1986, 1987) showed that interpretations of SDQ values are more complex because of a chronomorphocline, and also geographical, climatological and altitudinal clines. Röttger (1986) established that there is a great variation in the enamel quotient related to the distribution of *Arvicola* species in Europe and the Middle East.

Table 6
SDQ values variability with different step of artificial wear

	A. terrestris	*A. terrestris*	*A. terrestris*	*A. sapidus*	*A. sapidus*
Step 1	85.42	78.33	87.07	89.27	75.84
Step 2	84.62	74.39	86.93	89.17	82.00
Step 3	76.14	82.70	88.39	81.07	76.81
Step 4	67.44	75.71	82.00	85.37	76.72
Step 5	67.08	63.10	80.88	83.14	84.60
Step 6	74.68	73.94	84.59	88.24	79.98
Step 7	77.89	80.97	81.57	78.82	74.79

Table 7
SDQ values variability along tooth crown (thin-slides)

	Young individual		Adult individual		
	A. terrestris	*A. sapidus*	*A. terrestris*	*A. sapidus*	*A. sapidus*
Thin-slide 1	100	100	98.17	112	123.6
Thin-slide 2	101.18	102.60	81.13	120.15	93.95
Thin-slide 3	92.04	100.66	85.37	110.93	112.72

Western European populations show *Arvicola* enamel differentiation ($75.5 < \text{SDQ m1} < 120.8$). Eastern populations have a *Mimomys* enamel quotient (e.g. Turkey, SDQ m1 = 124.6, and Iran, SDQ m1 = 134.4). Kratochvil (1981) demonstrated that the enamel quotient of living water vole populations generally increases with altitude, which is why *Arvicola* populations in Switzerland (Vaud Canton, for example, 1160 m above sea level) have an *A. cantiana* SDQ value. Kratochvil (1980) noted that enamel quotient generally decreases with the age of the animals concerned. However, if young specimens are discounted, no essential difference is found between SDQ values in adults and old specimens. At the first stage of tooth wear, the pattern is undifferentiated (SDQ is close to 100).

Thus, our results induce a discussion of the validity and variability of the "enamel quotient" method and values and remarks on the reliability of a biochronological framework based on *Arvicola*.

The reduction in the number of subspecies and morphotype defined is linked to methodological choice. The outline analysis method and multivariate analyses take into account the enamel contour of the occlusal surface; they position the object in a morphospace to *n*-dimensions and give an overall view of size and shape. In contrast, SDQ calculation, m1 length measurement and univariate analysis focus on one feature of m1. The multiplication of subspecies and morphotype is induced by the method itself and the great variability of SDQ values. The great measurement error of enamel band thickness and the great variability of SDQ calculation result from a problem of protocol. The enamel thickness values

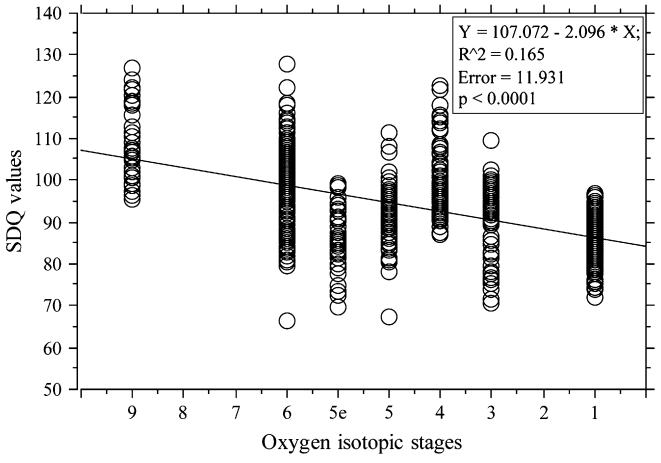

Fig. 9. Scatter diagram of SDQ values and the time (OIS).

are included between about 24 and 150 μm, consequently the acquisition of measurements requires great precision which may be affected by choice of landmarks. The enamel bands are rarely parallel along the same edge and the landmarks defined in the middle of the enamel wall reduce the precision of standardisation. Thus, the repeatability of measurements is less accurate and induces great variability in values between several measurers and between different measurement series of SDQ values. To work with the mean causes problems for biozonation definition, since the mean is a central point which requires consideration of dispersion (variance), and where values depend greatly on sample size and sample range. In this way, because of the great variability of SDQ values, the dispersion around the mean and the overlap of dispersion between each population, the arbitrary limitations of specific determination and biozonation are perhaps defined with too much precision. Another bias may be taphonomic effects such as abrasion by predator digestion (Andrews, 1991) leading to poor conservation and an ''artificial'' reduction in enamel thickness. These facts involve a loss of information and strongly distort SDQ calculation inducing the great variability of SDQ values. SDQ values show great variability within m1 and a given individual. The variability between salient angles and along tooth crowns with wear can be explained by chewing which is peculiar to each individual and is not a mechanical movement. Thus, irregular movements give different pressure points of wear. This fact explains the variability of SDQ calculation between the right and left side of a given individual. The variability along tooth crowns without wear has no trend in young individuals and in adult individuals whereas a decrease was supposed by Kratochvil (1980). This fact can be explained by irregularity of tooth growth.

In short, the variability in SDQ values involves considerable error in interpretation and is very important to assess the error risk incurred before specific determinations and stratigraphic interpretation.

The variation of SDQ values depends somewhat on time because of the low correlation between the decrease in SDQ values and the oxygen isotopic stages. The risk of a mistake in the relative stratigraphic positioning of a population is major (60%). In the morphological variability of *Arvicola*, there is no trend over time and no particular pattern with glacial and interglacial periods and geography. The chronomorphocline, and geographical, climatological and altitudinal clines and the migration wave of the northern form corresponding to SDQ values of *A. terrestris* ssp. B (Moula XIX–XVII) (Abbassi et al., 1998; Desclaux et al., 2000; Kolfschoten van, 1990, 1992) are not displayed. For archaeological records in a fluviatile context, the *Arvicola* lifestyle must be taken into account as the water vole digs tunnels in banks. Thus, *Arvicola* populations may be more recent than the archaeological levels where they have been found. This fact can explain cases where the SDQ values disagree with radiometric data. Consequently, it seems foolhardy to consider SDQ as a robust tool for dating and correlating archaeological records.

5. Conclusions

The present morphometric analyses suggest that a single species with great variability, *A. cantiana*, existed during the late Middle Pleistocene and the Upper Pleistocene.

The considerable difference between measurers, the complex interpretations of values, the high bias of measurement and calculation and also the error risk incurred throw doubt on the reliability of SDQ for specific determinations and stratigraphy.

To conclude, in terms of SDQ calculation and morphodiversity, *Arvicola* is not a useful tool for the construction of a biochronological framework. Taking into account the great phenotypic plasticity of m1, it seems difficult to apply biochronology methods within a rodent genus. It would be preferable to work with biozonation boundaries (FAD and LAD) between genera (e.g. *Mimomys*/*Arvicola* boundary).

Acknowledgements

This article is a contribution of the Institut de Paléontologie Humaine (CNRS UMR 6569) and Biogéosciences & EPHE, Ecole Pratique des Hautes Etudes (CNRS UMR 5561).

We wish to thank Alain Festeau and Dominique Champagnac for the establishment of the artificial wear protocol, Pascal Taubaty for thin-slides, Carmela Chateau for English corrections and François-Xavier Flotterer for discussions. We also thank two anonymous reviewers for their helpful remarks.

References

Abbassi, M., Desclaux, E., 1996. *Arvicola* Lacépède, 1799 (Rodentia, Mammalia) de quatre séquences du Sud-Est de la France et de Ligurie datant de la fin du Pléistocène moyen et du début du Pléistocène supérieur. Quaternaire 8 (1), 29–37.

Abbassi, M., Desclaux, E., Marquet, J.-C., Chaline, J., 1998. Répartition et évolution des *Arvicola* Lacépède, 1799 (Rodentia, Mammalia) au cours du Pléistocène moyen et supérieur en France et en Ligurie. Quaternaire 9 (2), 105–116.

Agadjhanian, A.-K., 1977. Quartäre Kleinsäuger aus der Russischen Ebene. Quartär 27/28, 111–145 (Bonn).

Agadjhanian, A.-K., 1983. Eine urtümliche *Arvicola* aus dem Mittlepleistozän der Russischen Ebene. Shcriftenr. Geol. Wiss. 19/20, 9–29 (Berlin).

Arroda, D., Giuggiola, O., Imperiale, G., Lamberti, A., Oxiola, M., Vicino, G., 1976. Le Manie. Il giacimiento musteriano dell'Arma (Finale, Savona). Archeologia in Liguria, Scavi e scoperte 1967–75. Doc. Soprin. Archeol. Della Liguria, Genova, pp. 113–144.

Andrews, P., 1991. Owls, Caves and Fossils Predation, Preservation and Accumulation of Mammals Bones in Caves, with an Analysis of the Pleistocene Caves Faunas from Westbury-sub-Mendip, Somerset. National History Museum Publications, London, U.K., pp. 231.

Campy, M., 1982. Le Quaternaire franc-comtois, Essai chronologique et paléoclimatique. Doctorat d'état, Fac. Sc. et Tech. de Franche-Comté, pp. 575.

Chaline, J., 1972. Les rongeurs du Pléistocène moyen et supérieur de France. Cahiers de Paléontologie. C.N.R.S, Paris, pp. 410.

Chaline, J., Laurin, B., 1986. Phyletic gradualism in a European Pliocene *Mimomys* lineage (Arvicolidae, Rodentia). Paleobiology 12 (2), 203–216.

Chaline, J., 1987. Arvicolid data (Arvicolinae, Rodentia) and evolutionary concepts. Evol. Biol. 21, 237–310.

Chaline, J., Brochet, G., 1989. Les Rongeurs et leurs significations paléoécologiques et paléoclimatiques. In: Campy, M., Chaline, J., Vuillemey, M. (Eds.), «La Baume de Gigny (Jura)» Gallia Préhistoire, 27ème suppl, pp. 97–110 (Paris).

Combier, J., 1967. Le Paléolithique d'Ardèche dans son cadre paléoclimatique. Publ. Inst. Préhist. Bord. Mem. 4, 31–127.

Combier, J., 1976. Gisement acheuléen d'Orgnac III. In: IXe congrès U.S.P.P. Nice. Livret guide de l'excursion A8, Paleolithique et Néolithique. pp. 212–226.

Defleur, A., Cregut-Bonnoure, E., Desclaux, E., 1998. Première mise en évidence d'une séquence éémienne à restes humains dans le remplissage de la Baume Moula-Guercy (Soyons, Ardèche). C. R. Acad. Sci. Paris 326 (6), 453–458.

Debenath, A., 1974. Recherches sur les terrains quaternaires charentais et les industries qui leur sont associées. Doctorat d'état, Univ Bordeaux I, pp. 678.

Delagnes, A., Tournepiche, J.-F., Armand, D., Desclaux, E., Diot, M.-F., Ferrier, C., Le Fillâtre, V., Vandermeersch, B., 1999. Le gisement Pléistocène moyen et supérieur d'Artenac (Saint-Mary, Charente): premier bilan interdisciplinaire. Bull. Soc. Préhist. Fr. 4, 469–496.

Delpech, F., 1988. Les grands mammifères, à l'exception des Ursidés. In: Rigaud, J.P. (Ed.), «La grotte Vaufrey, paléoenvironnements, chronologie, activités humaines». Mémoire de la Société préhistorique Française, pp. 213–289.

Desclaux, E., Abbassi, M., Marquet, J.-C., Chaline, J., Kolfschoten van, T., 2000. Distribution and evolution of *Arvicola* Lacepede, 1799 (Mammalia, Rodentia) in France and Liguria (Italy) during the Middle and the Upper Pleistocene. Acta Zool. Cracov. 43 (1–2), 107–125.

Dommergues, C.-H., 2000. Développement d'une application Matlab d'analyse de contour par transformées de Fourier, TX 5x Travaux de laboratoire Biogéosciences. Université de Bourgogne, Dijon. pp. 13.

Dommergues, C.-H., 2001. CDFT Complex Discret Fourier Transform (Matlab Package), Version 2.7. Biogeosciences-Dijon, UMR CNRS 5561, (Dijon).

Fejfar, O., Heinrich, W.-D., 1983. Arvicoliden-Sukzession und Biostratigraphie des Oberpliozän und Quartärs in Europa. Schriftenr. Geol. Wiss. 19/20, 61–109.

Fornasiero, O., 1989. Cronostratigrafia di due giacimienti Quaternari continentali a mammiferi della Liguria. Corso di Laurea in Scienze Geologiche. Univ. Degli Studi di Ferrara, pp. 119.

Gonzalez, R.-C., Woods, R.-E., 1992. Digital Image Processing. Addison Wesley Longman, Inc, pp. 716.

Heim de Balzac, H., Guislain, R., 1955. Evolution et spéciation des campagnols du genre *Arvicola* en territoire français. Mammalia 19 (3), 367–390.

Heinrich, W.-D., 1978. Zur biometrischen Erfassung eines Evolutiontrends bei *Arvicola* (Rodentia, Mammalia) aus dem Pleistozän Thüringens. Säugetierkd. Inform. 2/3, 3–21.

Heinrich, W.-D., 1982. Zur Evolution und Biostratigraphie von *Arvicola* (Rodentia, Mammalia) im Pleistozän Europas. Z. Geol. Wiss. 10, 683–735.

Heinrich, W.-D., 1987. Neue Ergebnissezue evolution und Biostratigraphie von *Arvicola* (Rodentia, Mammalia) in Quatärs Europas. Z. Geol. Wiss. 15, 389–406.

Heinrich, W.-D., 1990. Some aspects of evolution and biostratigraphy of *Arvicola* (Mammalia, Rodentia) in the central European Pleistocene. In: International Symposium on Evolution Phylogeny and Biostratigraphy of Arvicolids. pp. 165–182.

Hinton, M.A.C., 1926. Monograph of Voles and Lemmings (Microtinae) Living and Extinct, vol. 1. British Museum (Natural History), London.

Janossy, D., 1969. Stratigraphische Auswertung der europäischen mittelpleistozänen Wirbeltierfaunen. Ber. Dtsch. Ges. Geol. Wiss. A 14 (Teil 1: Heft 4), 367–438. Teil 2: Heft 5, 573–643.

Koenigswald von, W., 1970. Mittelpleistozäne Kleinsäugerfauna aus der Spaltenfüllung Petersbuch bei Eichstätt. Mitt. Bayer. Staatssamml. Paläontol. Hist. Geol. 10, 407–432.

Koenigswald von, W., 1973. Veränderungen in der Kleinsäugerfauna von Mitteleuropa zwischen Cromer und Eem (Pleistozän). Eiszeitalt. Ggw 23/24, 159–167.

Koenigswald von, W., 1980. Schmelzstruktur und Morphologie in dem molaren der Arvicoliden (Rodentia). Abh. Senckenb. Natforsch. Ges. 59 (1), 1–94.

Koenigswald von, W., 1994. Functional symmetries in the schmelzmuster and morphology of rootless rodent molars. Zool. J. Linn. Soc. 110, 141–179.

Koenigswald von, W., Kolfschoten van, T., 1996. The *Mimomys–Arvicola* boundary and the enamel thickness quotient (SDQ) of *Arvicola* as stratigraphic markers in the Middle Pleistocene. In: The Early Middle Pleistocene in Europe, Balkema, pp. 211–238.

Kolfschoten van, T., 1990. The evolution of mammal fauna in the Netherlands and the middle Rhine area (Western Germany) during the late middle Pleistocene. Meded. Rijks Geol. Dienst. 43 (3), 42–74.

Kolfschoten van, T., 1992. Aspect of the migration of mammals to northwestern Europe during the Pleistocene, in particular the reimmigration of *Arvicola terrestris*. Cour. Forschungsinst. Senckenb. 153, 213–220.

Kratochvil, J., 1980. Zur Phylogenie und Ontogenie bei *Arvicola terrestris* (Rodentia, Arvicola). Folia Zool. 29, 209–224.

Kratochvil, J., 1981. Arvicola cantiana vit-elle encore? Folia Zool. 30, 289–300.

Kretzoi, M., 1969. Skizze einer Arvicoliden-Phylogenie, Stand, 1969. Vertebr. Hung. 11, 155–193.

Kuhl, F.-P., Giardina, C.-R., 1982. Elliptic Fourier features of a closed contour. Comput. Graph. Image Process. 18, 236–258.

Laville, H., Prat, F., Thibault, C., 1973. Un gisement à faune du Pléistocène moyen: la grotte de l'Eglise à Cénac et Saint-Julien (Dordogne). Quaternaria XIV, 71–119.

Laville, H., 1973. Climatologie et chronologie du Paléolothique en Périgord. Etude sédimentologique des dépôts en grottes et en abris. Doctorat d'état, Univ Bordeaux I, pp. 716.

Lecointre, G., Le Guyader, H., 2003. Classification phylogénétique du vivant, 2e éd. Belin, pp. 543.

Lestrel, P.E., 1997. Fourier Descriptors and their Applications in Biology. Cambridge University Press, Cambridge.

Le Louarn, H., Quéré, J.-P., 2003. Les rongeurs de France. Faunistique et biologie. revue et augmentée, 2e éd. INRA édition, Paris, pp. 256.

Lumley de, H., 1989. Le Paléolithique inférieur et moyen du Midi Méditerranéen dans son cadre géologique. Vème suppl. à Galia Préhistoire. 2t, 896.

Marquet, J.-C., 1989. Paléoenvironnements et chronologie des sites du domaine atlantique français d'âge Pléistocène moyen et supérieur d'après l'étude des rongeurs. Doctorat d'état, Univ. Bourgogne, pp. 637.

Navarro, N., Zatarian, X., Montuire, S., 2004. Effects of morphometric descriptor changes on statistical classification and morphospaces. Biol. J. Linn. Soc. 83, 243–260.

Radulesco, C., Samson, P., 1977. *Arvicola* (Rodentia, Mammalia) dans le Pléistocène moyen de Roumanie. Trav. Inst. Spéol. «Emilie Racovita» 16, 151–162.

Rabeder, G., 1981. Die Arvicoliden (Rodentia, Mammalia) aus dem Pliozän und dem älterren Pleistozän von Niederösterreich. Beitr. Paläontol. Österreich 8 (1), 343.

Renaud, S., 1997. Modalité de l'évolution morpholgique: analyse morphométrique d'une spécialisation dentaire chez les Murinés, la stephanodontie. Thèse de doctorat, Université de Montpellier II.

Rigaud, J.P., 1988. La grotte Vaufrey à Cénac et Saint-Julien (Dordogne): Paléoenvironnement, chronologie et activités humaines. Mém. Soc. Préhist. Fr. 19, 616.

Roebroeks, W., Kolsfschoten, van T., 1994. The earliest occupation of Europe: a short chronology. Antiquity 68, 489–503.

Rohlf, F.J., Marcus, L.F., 1993. A revolution in morphometrics. Tree 8 (4), 129–132.

Röttger, U., 1986. Schmelzbandbreiten an Molaren der Gattung "*Arvicola*" Lacépède, 1799. Med. Dissertation 121.

Röttger, U., 1987. Schmelzbandbreiten an Molaren von Shermäusen (*Arvicola* Lacépède, 1799). Bonn. Zool. Beitr. 38 (2), 95–105.

Schmittbuhl, M., Allenbach, B., Schaaf, A., Viriot, L., 1997. Méthodes de Fourier elliptique appliqués à la quantification de la forme de contours complexes. Biom. Hum. Anthropol. 15, 233–241.

Shenbrot, G., Krasnov, B., 2005. An Atlas of the Geographic Distribution of the Arvicoline Rodents of the World (Rodentia, Muridae: Arvicolinae). Pensoft Publishers, Sofia, pp. 336.

Statsoft, Statistica® Version 7.1.

Viriot, L., 1994. Tendances évolutives des molaires chez les Arvicolidés (Rodentia, Mammalia). Université de Bourgogne, Centre des Sciences de la Terre, Dijon, pp. 160.

Wilson, D., Reeder, D., 2005. Mammal Species of the World a Taxonomic and Geographic Reference, third ed., Washington and London, pp. 1207.

Wust-Saucy, A.-G., 1998. Polymorphisme génétique et phylogéographie du Campagnol terrestre *Arvicola terrestris*. Diss. thesis, Université de Lausanne, pp. 162.