Available online at www.sciencedirect.com
ScienceDirect
Molecular Phylogenetics and Evolution 41 (2006) 266–278
MOLECULAR PHYLOGENETICS AND EVOLUTION
www.elsevier.com/locate/ympev

Phylogenetic systematics and tempo of evolution of the Viverrinae (Mammalia, Carnivora, Viverridae) within feliformians: Implications for faunal exchanges between Asia and Africa

Philippe Gaubert [a,*], Pedro Cordeiro-Estrela [b,1]

[a] *Estación Biológica de Doñana (CSIC), Departamento de Biología Aplicada, Avenida María Luisa s/n Pabellón del Perú, 41013 Sevilla, Spain*
[b] *Muséum National d'Histoire Naturelle, Département Systématique et Evolution, UMR 5202 CNRS/USM 601, "Origine, Structure et Evolution de la Biodiversité", CP 51, 55 rue Buffon, 75005 Paris, France*

Received 14 November 2005; revised 3 April 2006; accepted 19 May 2006
Available online 3 June 2006

Abstract

We reconstructed the phylogeny of the subfamily Viverrinae (Mammalia, Carnivora, Viverridae) using a ~3 kb data set in order to reassess timing and patterns of faunal exchanges between Asia and Africa. Maximum parsimony, maximum likelihood, and Bayesian analyses of separated and combined matrices (cytochrome *b*, transthyretin intron I and IRBP exon 1 [IRBP1]) recovered all the well-supported relationships within feliformian lineages. In addition, IRBP1 supported paraphyly of genus *Herpestes* and contributed to the resolution of equivocal hypotheses within Viverridae, including (1) the monophyly of Viverrinae, and (2) *Viverricula* sister-group of the other terrestrial civets (*Civettictis* and *Viverra*). The combined analysis yielded a robust phylogeny, recovering monophyly of Prionodontidae and yielding high posterior probabilities for nodes (1) (Prionodontidae, Felidae) and (2) ((Felidae, Prionodontidae), ((Hyaenidae, (Herpestidae, Eupleridae)), Viverridae)). Using a fossil cross-validation method, we estimated the emergence of Viverridae at 34.29 Myr, with a separation between the three traditional subfamilies Hemigalinae, Paradoxurinae, and Viverrinae during the Late Oligocene–Early Miocene. The terrestrial civets and the splits between (1) *Civettictis* and *Viverra* and (2) *Poiana* and *Genetta* were estimated to appear during the Middle Miocene. Parsimony- and maximum likelihood-based methods yielded unambiguous ancestral area reconstructions, including the Asian origin of the family Viverridae, the subfamily Viverrinae, the terrestrial civets and the clade (*Civettictis, Viverra*). On the grounds of genetic distances, morphological divergence, and divergence time estimates, we propose the erection of the subfamily Genettinae (including *Genetta* and *Poiana*). Our analyses suggested two independent migration events from Asia to Africa, during the Middle Miocene (*Civettictis*) and between the Late Oligocene and Middle Miocene (Genettinae). These results are in agreement with the hypothesis of Miocene routes from Asia to Africa—via the Arabian microplate—that would have involved several independent events of migrations. Couched in the context of the viverrid fossil record, our study calls for a revision of the paleontological data in order to fully appreciate the complexity of Afro-Asian faunal exchanges.

Keywords: Viverrinae; Genettinae; Phylogeny; Migration route; Miocene; Fossil cross-validation; Asia–Africa; Fossil record

1. Introduction

The Neogene collision between Asia and Africa via the Arabian microplate allowed intense faunal and floral exchanges, acting as a promoter of taxonomic evolutionary dynamics in the two continents (Kappelman et al., 2003). However, the time from which the Arabian microplate allowed effective taxa exchanges remains uncertain.

* Corresponding author. Present address: Institut de Recherche pour le Développement, UT 136 "Aires protégées", Centre IRD, 5 rue du Carbone, 45072 Orléans, France. Fax: +33 2 38 49 95 47.
E-mail address: gaubert@orleans.ird.fr (P. Gaubert).
[1] Present address: Universidade Federal do Rio Grande do Sul, Departamento de Genética, Laboratório de citogenética e evolução, Campus do Vale-Bloco III, Av. Bento Gonçalves, 9500 Agronomia, CP 15.053, Porto Alegre, RS, 91501-970, Brazil.

1055-7903/$ - see front matter © 2006 Elsevier Inc. All rights reserved.
doi:10.1016/j.ympev.2006.05.034

Tectonic data estimate microplate collision between ca. 16 and 20 Myr (Cox, 2000; Krijgsman, 2002), whereas paleontological evidence suggests intercontinental migrations antedating 27 Myr (Kappelman et al., 2003); the latter is in agreement with estimates from plant phylogenetic studies (Davis et al., 2002; Renner et al., 2004). The mammalian fossil record argues for an Asian origin of several extant African lineages (e.g., within Ruminantia, Rhinocerotidae, Hystricognathi, Carnivora—Antoine et al., 2003; Huchon and Douzery, 2001; Métais et al., 2001; Schmidt-Kittler, 1987), which implies at least one Miocene migration route to Africa throughout forested corridors within the Arabian Peninsula (Cox and Moore, 1993). The existence of such a migratory path has been supported by phylogenetic patterns in extant taxa (Juste et al., 1999; Kosuch et al., 2001).

However, recent advances in molecular phylogenetic studies have challenged the common point of view of a single "Asia to Africa" migration route via the Arabian Peninsula. Indeed, phylogenetic patterns suggest migrations between the two continents—including retro-migrations from Africa—at different Miocene periods (Chevret and Dobigny, 2005; Ropiquet and Hassanin, 2004) during variable climatic conditions in Arabia (Griffin, 2002).

We conducted phylogenetic analyses and estimated molecular divergence times in the subfamily Viverrinae (Mammalia, Carnivora, Viverridae) in order to re-assess timing and patterns of faunal exchanges between Africa and Asia. The Viverrinae are included in the sub-order Feliformia under the family Viverridae, the systematics of which has been recently clarified (Flynn and Nedbal, 1998; Gaubert et al., 2005; Yoder et al., 2003). They comprise mesopredators grouping into two distinct clades: (1) the large, digitigrade terrestrial civets (*Civettictis, Viverra, Viverricula*), and (2) the slender, semi-digitigrade genets (*Genetta*), and oyans or African linsangs (*Poiana*) (Gaubert and Veron, 2003). The Viverrinae are an appropriate group for questioning Afro-Asian faunal relationships because (1) they are distributed within the intertropical zone of both continents (Nowak, 1999), (2) the extant representatives are derived from a diversified fossil record present in Africa and Eurasia since the Early Miocene (Hunt, 1996), and (3) the clade of extant terrestrial civets combines both Asian (*Viverra, Viverricula*) and African (*Civettictis*) representatives—whereas the genets and oyans are endemic to Africa. Although the systematics of the Viverrinae has recently been re-assessed (exclusion of the Asian genus *Prionodon*—Gaubert and Veron, 2003; Gaubert et al., 2004b), several phylogenetic relationships within the group remain unresolved, notably among the terrestrial civets.

The aim of our study is first to provide a robust, comprehensive phylogeny of the extant Viverrinae, within a representative set of feliformian taxa. Second, we estimate lineage divergence times from multiple fossil calibrations to propose evolutionary scenarios with re-assessed timing of faunal exchanges between Africa and Asia. In return, we expect from these phylogenetic investigations a re-assessment of the systematics of the Viverrinae and of the accuracy of fossil calibration points used within feliformians.

2. Materials and methods

2.1. Taxonomic sampling

Nucleotide sequence variation was examined in 34 feliformians (ingroup) and 4 caniformians (outgroup). The three subfamilies of Viverridae were represented: seven genera/13 species of Viverrinae, two genera/two species of Paradoxurinae and one species of Hemigalinae (Table 1). The six other families of Feliformia (Gaubert et al., 2005; Wozencraft, 2005) were also represented in order to have all the major ingroup lineages included in our analysis: Nandiniidae (one species), Prionodontidae (two species/one genus), Felidae (two species/two genera), Hyaenidae (two species/two genera), Herpestidae (seven species/six genera), and Eupleridae (four species/four genera). Taxonomic sampling within Viverridae was achieved through field collection (PG) and thanks to the contribution of field collaborators (see Acknowledgments). Fresh tissues and hairs constituted original DNA sources.

2.2. DNA sequencing and nucleotide coverage

Our data set represents one mitochondrial gene (cytochrome *b*; cytb) and two nuclear gene fragments (transthyretin Intron I; TriI—and IRBP exon 1; IRBP1). The matrices for cytb (1140 bp) and TriI (1096 [indels included]—775 bp [indels removed]) were mostly compiled from previous works on the phylogeny of carnivorans (Arnason et al., 1995; Flynn and Nedbal, 1998; Gaubert and Veron, 2003; Gaubert et al., 2004b; Gaubert et al., 2005a; Hosoda et al., 2000; Ledje and Arnason, 1996; Sato et al., 2004; Yoder et al., 2003). We completed the cytb and TriI matrices with three and five additional species respectively, in order to increase the taxonomic data set and to allow additional calibration points (following Gaubert and Veron, 2003; Gaubert et al., 2004b). The taxa concerned are Prionodontidae, Hyaenidae, and Viverrinae (Table 1).

IRBP1 was sequenced for 15 species of feliformians, which were then aligned with sequences produced by Yoder et al. (2003) and Sato et al. (2004), totaling a data set of 1031 bp (1028 without indel). Total genomic DNA was prepared following standard CTAB method (Kocher et al., 1989). The amplification of IRBP1 was performed with a set of two pairs of primers (designed using Oligo v. 4.0—Rychlik, 1992) which produced two fragments with >100 bp of overlap. U and L were used for upper and lower strand, respectively: "IRBP U29" 5′-GCT CCT TGA ACG ACC CTC-3′— "IRBP L559" 5′-ACC CCC TAC AGT CCG CTC-3′; and "IRBP U424" 5′ ACC CTG CCC CAG GTC CT-3′—"IRBP L1117" 5′-GGC ATC GGC AAA GCT GTC-3′. Reaction parameters for PCR amplification were described in Veron and Heard (2000). We used annealing temperatures between 49 and 50 °C and the *Taq* polymerase RedTaq (Sigma–Aldrich, St. Louis, MO) for 35–45 PCR cycles. The reaction products were visualized in a 1.5% agarose gel and then purified from the PCR mixture

Table 1
Taxonomic sampling and GenBank accession numbers for nucleotide sequences used in this study

Taxon	Cytochrome *b*	Transthyretin intron 1	IRBP exon 1
Caniformia (outgroup)			
*Canis**	AB048590	AF039732	AY170074
Ursus arctos	X82308	AF039741	AB109333
*Mustela**	AB051240	AF039735	AB082969
Procyon lotor	X94930	AF039736	AB082981
Feliformia (ingroup)			
Nandiniidae			
Nandinia binotata	AF511057	AY232613	AY170083
Felidae			
*Felis**	X82296	AF039724	Z11811
Panthera leo	X82300	AF039725	AB109336
Prionodontidae			
Prionodon linsang	AF511065	DQ267551	DQ267572
Prionodon pardicolor	AF522349	AY23261	DQ267573
Hyaenidae			
Hyaena hyaena	AF511063	DQ267552	DQ267570
Crocuta crocuta	AF511064	AF039728	AY170087
Eupleridae			
Fossa fossana	AF511062	AY170019	AY170067
Cryptoprocta ferox	AF511070	AY232612	AY170066
Galidia elegans	AY170099	AY170020	AY170069
Mungotictis decemlineata	AF511061	AY170016	AY170064
Herpestidae			
Suricata suricatta	AF522346	AY170028	AY170084
Crossarchus obscurus	AF522327	AF039726	AY170071
Mungos mungo	AY170095	AY170017	AY170065
Herpestes javanicus	AY170108	AY170081	AY170081
Herpestes edwardsii	AY170107	AY170025	AY170080
Herpestes ichneumon	AF511059	AY232611	DQ267571
Cynictis penicillata	AF511060	AY170024	AY170079
Viverridae			
Hemigalinae			
Hemigalus derbyanus	AF511067	AY170027	AY170082
Paradoxurinae			
Paguma larvata	AF511069	AY232615	AB109339
Paradoxurus hermaphroditus	AF511056	AF039730	DQ267569
Viverrinae			
Viverricula indica	DQ267556	AY232618	DQ267568
Civettictis civetta	AF511043	AY232619	AY170078
Viverra tangalunga	AF511045	AF039731	AY170085
Viverra zibetha	AF511047	AY232616	DQ267566
Viverra megaspila	AF511046	AY232617	DQ267567
Poiana richardsonii	AY241891	AY232620	DQ267559
Genetta thierryi	AF511052	AY232625	DQ267560
Genetta servalina	AF511053	AY232626	AY170088
Genetta johnstoni	DQ267557	AY232624	DQ267561
Genetta genetta	DQ267558	DQ267555	DQ267565
Genetta pardina	AY397706	DQ267553	DQ267563
Genetta maculata	AY241901	AY232627	DQ267562
Genetta tigrina	AY241889	DQ267554	DQ267564

GenBank numbers starting with "DQ" constitute new sequences produced for this study.

* For the purpose of lineage representation in the concatenated analysis, sequences were combined between species representatives of the same genus in the following cases: *Canis familiaris* × *C. lupus* (*Canis*), *Mustela erminea* × *M. frenata* (*Mustela*), and *Felis catus* × *F. silvestris* (*Felis*).

using the MinElute PCR Kit (Qiagen S.A., Courtaboeuf, France). We directly sequenced the purified products in both forward and reverse directions with an automated DNA sequencer CEQ 2000 DNA Analysis System (Beckman–Coulter Inc., Fullerton, CA).

The combined matrix represents an alignment of 3267 (indels included) or 2943 (indels removed) bp. All the new sequences produced for this study are available in GenBank (see Table 1).

2.3. Phylogenetic analyses

Sequences were aligned by eye using BioEdit v. 5.0.6 (Hall, 1999). Maximum Parsimony (MP) analyses were

conducted with PAUP* v. 4.0b10 (Swofford, 2002) on an Apple Macintosh G4. We ran heuristic searches using tree bisection-reconnection branch-swapping and random stepwise addition (10 replicates). All sites were attributed equal weights, and indels were treated as fifth states. The support of nodes was measured through nonparametric bootstrapping (Felsenstein, 1985) with 1000 pseudoreplicates.

The models of molecular evolution for maximum likelihood (ML) analysis were selected using DT-ModSel (Minin et al., 2003). This program uses a Decision-Theoretic (DT) approach that weights the selection of a model by its performance in estimating the branch lengths of a given phylogenetic tree (in addition to its fit). This is different from the traditional likelihood ratio test-based method (Posada and Crandall, 1998), which has been shown to select complex models that may not contribute to the performance of the phylogenetic inference (Abdo et al., 2004; Pol, 2004). The result of the DT approach is the selection of the simplest model among a set of equivalent models, thus tending to discard high variations in the parameter estimates to which the more complex models can be subjected (Abdo et al., 2004; Burnham and Anderson, 2002). We performed model selection for the three separated data sets and for the combined matrix. All portions of the alignments containing indels were removed before statistical analysis (321 bp in TriI and 3 bp in IRBP1). The best-fitting model for cytb and TriI was K81uf (with unequal base frequencies—Kimura, 1981; Posada, 2004) with a gamma rate distribution (Γ), plus a proportion of invariable sites (I) in the case of cytb. DT-ModSel selected for IRBP1 a HKY+Γ+I model (Hasegawa et al., 1985). When the three genes were concatenated into a same matrix, the best-fitting model was GTR+Γ+I (Tavaré, 1986). Model parameters for IRBP1 and the combined matrix are detailed in Table 2. ML analyses with DT-ModSel parameters fixed *a priori* were run under PAUP*. Heuristic search conditions were the same than those considered in MP analysis. Node support was assessed using nonparametric bootstrapping with 100 pseudoreplicates.

Bayesian analyses were ran using MrBayes 3.1 (Ronquist et al., 2005) on a computer Intel Pentium 4 (2.8 GHz) under Mandrake Linux v. 10.1. Evolutionary models selected for single gene analysis by DT-ModSel were used or approximated (due to restriction in model availability under MrBayes, we used six substitution rates instead of 3 [K81] for cytb and TriI) with a γ-distribution divided into five categories. In what concerns the concatenated analysis, we attributed the respective models to each partition, and then used the MrBayes command lines for unlinking parameters and allowing partitions to evolve under different rates. According to Ronquist et al. (2005), we used the default priors for the following parameters: topology, branch lengths, nucleotide frequencies, nucleotide substitution rates, proportion of invariable sites (whenever relevant), and γ-distribution. Four Markov chains were run simultaneously for 2,000,000 Metropolis-coupled generations (sampled every 20 generations) in two independent runs. Convergence between runs was estimated visually by examining the plot "number of generations versus log likelihood values" and using the Potential Scale Reduction Factor (PSRF) provided in the "sump" output of MrBayes 3.1 (PRSF values nearing 1 were assumed to diagnose convergence). Burnin was fixed to 3000 and 2000 trees in separated and combined analyses, respectively. The total number of trees used to construct the majority-rule consensus phylograms under PAUP* was 17350 (cytb), 27174, (TriI) 18890 (IRBP1), and 283 (combined analysis).

Mean genetic distances among the four viverrid lineages (Paradoxurinae, Hemigalinae, terrestrial civets, oyans, and genets) was calculated under MEGA v. 3.1 (Kumar et al., 2004) using number of nucleotide differences (with "complete deletion" for handling gaps and missing data). The calculation of genetic distances from the number of nucleotide differences allows for a direct estimate of nucleotide divergence not depending on a specific model of molecular evolution (see Nei and Kumar, 2000).

2.4. Estimation of divergence time

Divergence time estimates were calculated from the concatenated molecular matrix using the software r8s v. 1.7 (Sanderson, 2003, 2004). The latter provides a method based on penalized likelihood (Green and Silverman, 1994) that allows different rates of evolution on every branch of a phylogenetic tree but applies a penalty function that smoothes rate variation between nearby branches (Sanderson, 2002). It also allows the simultaneous use of

Table 2
Models of evolution and parameters estimated using DT-ModSel for IRBP exon 1 and the combined nucleotide matrices (maximum likelihood analysis)

Data set	Model of evolution	Parameters				
		Base frequencies	Substitution rates	Ti/Tv ratio	Proportion of invariable sites	Gamma rate distribution
IRBP exon 1	HKY+Γ+I	$\pi_A = 0.179476$ $\pi_C = 0.315581$ $\pi_G = 0.317883$ $\pi_T = 0.187060$	—	3.553984	$I = 0.439824$	$\Gamma = 0.821494$
Combined matrix	GTR+Γ+I	$\pi_A = 0.264366$ $\pi_C = 0.305339$ $\pi_G = 0.211464$ $\pi_T = 0.218831$	r(A-T) = 1.406596 r(A-G) = 4.759453 r(A-C) = 1.211676 r(G-T) = 0.483862 r(C-T) = 12.709424	—	$I = 0.271408$	$\Gamma = 0.451798$

Table 3
Fossil calibration points used in this study

Lineage split/origin	Minimum age	Source
Panthera–Felis	4 Myr	Werdelin and Lewis (2005)
Crocuta–Hyaena	9.5 Myr	Werdelin (1996)
Herpestidae	18 Myr (*Leptoplesictis*)	Roth (1988), Hunt (1996)
Herpestes	9.5 Myr	Barry (1983)
Viverridae	23 Myr (*Herpestides*)	Hunt (1991, 1996)
Viverra	9 Myr	Petter and Howell (1977)
Genetta	7.5 Myr	Werdelin (2003)

Paleontological ages represent minimum time estimates.

several fossil calibration points for estimating divergence times. We first compiled a total of seven calibration points that we could extract from the paleontological literature for the feliformian taxa represented in our study (Table 3). Following the method of "fossil cross-validation" (Near and Sanderson, 2004; Near et al., 2005a; Near et al., 2005b) implemented in r8s, we ran a first round of analysis, using all fossil calibration points, in order to identify calibrations that produced inconsistent molecular divergence time estimates. This method is based on a two-step procedure described in Near and Sanderson (2004). We first calculated the differences between the molecular and fossil estimates (D) for all the fossil-dated nodes available in the phylogeny. We then summed the squared differences (SS) for each fossil calibration in order to better visualize calibrations that showed the most inconsistent values with respect to the other fossils used in the analysis. We then calculated the average squared deviation (s) to estimate the impact of the most inconsistent fossil calibrations, by removing one by one the fossils with the greatest SS values and recalculating (s) with the remaining calibration points. This procedure was applied until the removal of the fossils did not affect significantly the magnitude of (s) decrease. Following these results, we ran a final analysis using a pruned set restricted to the four most consistent fossil calibration points in order to estimate divergence times within feliformians for all the well-supported nodes of the combined analysis.

2.5. Estimation of ancestral areas

Estimation of ancestral areas among Viverridae was conducted using the tree topology yielded by the Bayesian combined analysis. We coded geographical areas as continents into one binary character (0: Asia–1: Africa). We used a first method of ancestral state reconstruction based on linear parsimony through the software MacClade v. 3.07 (Maddison and Maddison, 1992). Because MacClade uses a procedure from both "up and down" and reverse readings of the tree (Swofford and Maddison, 1987), and in order to avoid biased estimates caused by incomplete taxonomic sampling in lineages outside the group of interest, we only retained the subtree of Viverridae. This procedure is similar to fixing uncertainty state (?) on the branch leading to the hypothetical outgroup. We used this technique as a way of minimizing *a priori* hypotheses about the condition at root in Viverridae (but see Grandcolas et al., 2004).

In order to compare these estimations with a probabilistic method, we used the software Discrete v. 4.0 (Pagel, 2000), which takes into account branch lengths and evolutionary rates of characters to be reconstructed (see Mooers and Schluter, 1999; Pagel, 1994, 1999). The fit of the data was significantly improved using the gamma rate heterogeneity model of trait evolution (Likelihood Ratio statistics [LR] >2), which allows states to evolve at different rates along the tree (Yang, 1994). The final model we used implemented two transition rates for change between states ($0 \rightarrow 1 = 0.00298$ and $1 \rightarrow 0 = 0.0006$; significantly better than the one-rate model) with gamma rate = 0.33073. We compared ancestral state estimates obtained from both "local" and "global" calculations (Pagel, 1999). The first method re-calculates the parameters of the model for each state at nodes, and allows ancestral state reconstructions of neighbouring nodes to vary. The global option applies instead the parameter values estimated for the model and calculates the best simultaneous set of ancestral states on the tree, which may differ from the ancestral states obtained by calculating separately the most probable state at each node (Pagel, 2000).

3. Results

3.1. Phylogenetic analyses

A total of 1688 characters were variable, of which 1237 were parsimony informative (314 and 189, respectively, for IRBP1). Analyses of IRBP1 and combined matrix alignments yielded eight most parsimonious trees of 616 steps (CI = 0.636; RI = 0.757) and one most parsimonious tree of 5282 steps (CI = 0.458; RI = 0.594), respectively. MP, ML, and Bayesian analyses of both IRBP1 and combined data sets (cytb, TriI, and IRBP1) recovered similar topologies and nodal support measures (Figs. 1 and 2). All the well-supported relationships within the main feliformian lineages were recovered: (1) *Nandinia* was the sister-group to all other extant feliformians, (2) the clade (Hyaenidae, (Herpestidae, Eupleridae)), (3) the sister-group relationship between social and "solitary" mongooses, (4) the monophyly of Viverridae as newly defined, and (5) the clade (Hemigalinae, Paradoxurinae) (Flynn and Nedbal, 1998; Flynn et al., 2005; Gaubert and Veron, 2003; Gaubert et al., 2004b; Veron et al., 2004; Yoder et al., 2003).

Analysis of IRBP1 alone (Fig. 1) contributed to the resolution of equivocal or weakly supported phylogenetic hypotheses within Viverridae (Gaubert and Veron, 2003; Gaubert et al., 2004b), concerning (1) the monophyly of Viverrinae ($bp_{MP-ML} = 98–99\ \%$; $pp_{Bayes} = 1.00$), and (2) the sister-group relationship between *Viverricula* and the other genera of terrestrial civets (*Civettictis* and *Viverra*) ($bp_{MP-ML} = 98–95\%$; $pp_{Bayes} = 1.00$). The production of a new sequence for Herpestidae (*Herpestes ichneumon*) also allowed us to confirm the paraphyly of the genus *Herpestes* (Veron et al., 2004) from nuclear DNA sequences.

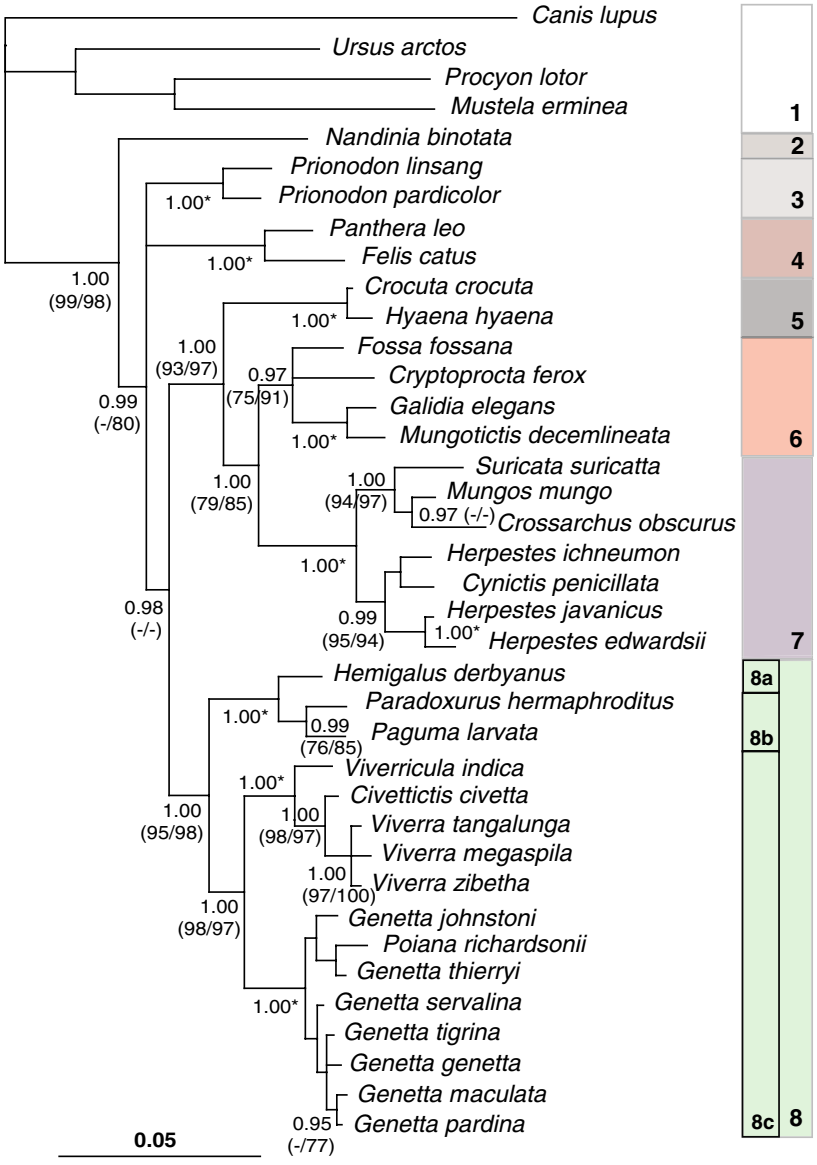

Fig. 1. Phylogram inferred from Bayesian analysis of IRBP1 (1028 bp). The model of sequence evolution is HKY+Γ+I. Values at nodes indicate Bayesian posterior probabilities >0.95 and bootstrap values >75% for MP and ML analyses, respectively (between parentheses). Bayesian posterior probabilities followed by asterisk represent nodes recovered with 100% of support in both MP and ML analyses. Scale bar corresponds to 5% sequence divergence. Numbers in boxes: 1, Caniformia (outgroup); 2, Nandiniidae; 3, Prionodontidae; 4, Felidae; 5, Hyaenidae; 6, Eupleridae; 7, Herpestidae; 8, Viverridae (8a, Hemigalinae; 8b, Paradoxurinae; 8c, Viverrinae).

Results from the combined analysis exemplified the complementarity between the phylogenetic signals of cytb, TriI, and IRBP1, which yielded generally high nodal supports along the feliformian tree (Fig. 2). The phylogenetic relationships supported by IRBP1 analysis within Viverridae and Herpestidae were recovered, and the monophyly of Prionodontidae was confirmed (Gaubert et al., 2004b). The combined analysis also yielded well-supported nodes for (1) the sister-group relationship between Prionodontidae and Felidae (bp_{MP-ML} = 86–82%; pp_{Bayes} = 1.00), and (2) the clade ((Felidae, Prionodontidae), ((Hyaenidae, (Herpestidae, Eupleridae)), Viverridae)) (pp_{Bayes} = 1.00; but bp_{MP-ML} < 50%).

Mean genetic distance values among the four viverrid lineages (Paradoxurinae, Hemigaline, terrestrial civets, and genets + oyans) are given in Table 4. Genetic distances had a low amplitude spectrum, ranging from 183.00 (SE ± 11.63) mean number of nucleotide differences between Paradoxurinae and Hemigalinae, to 213.38 (±12.00) between genets + oyans and Hemigalinae.

3.2. Divergence times and ancestral areas

The "fossil cross-validation" method estimated as inconsistent the calibration points used for Viverridae (23 Myr), *Panthera-Felis* (4 Myr) and *Viverra* (9 Myr). They were then removed in the final estimation of divergence times (Fig. 3). We obtained similar results to the few estimates available in the literature for recent and intermediate nodes within the feliformian tree (Fig. 4). The nodes (*Poiana, Genetta*) and (*G. pardina*, (*G. maculata*, *G. tigrina*)) were estimated at

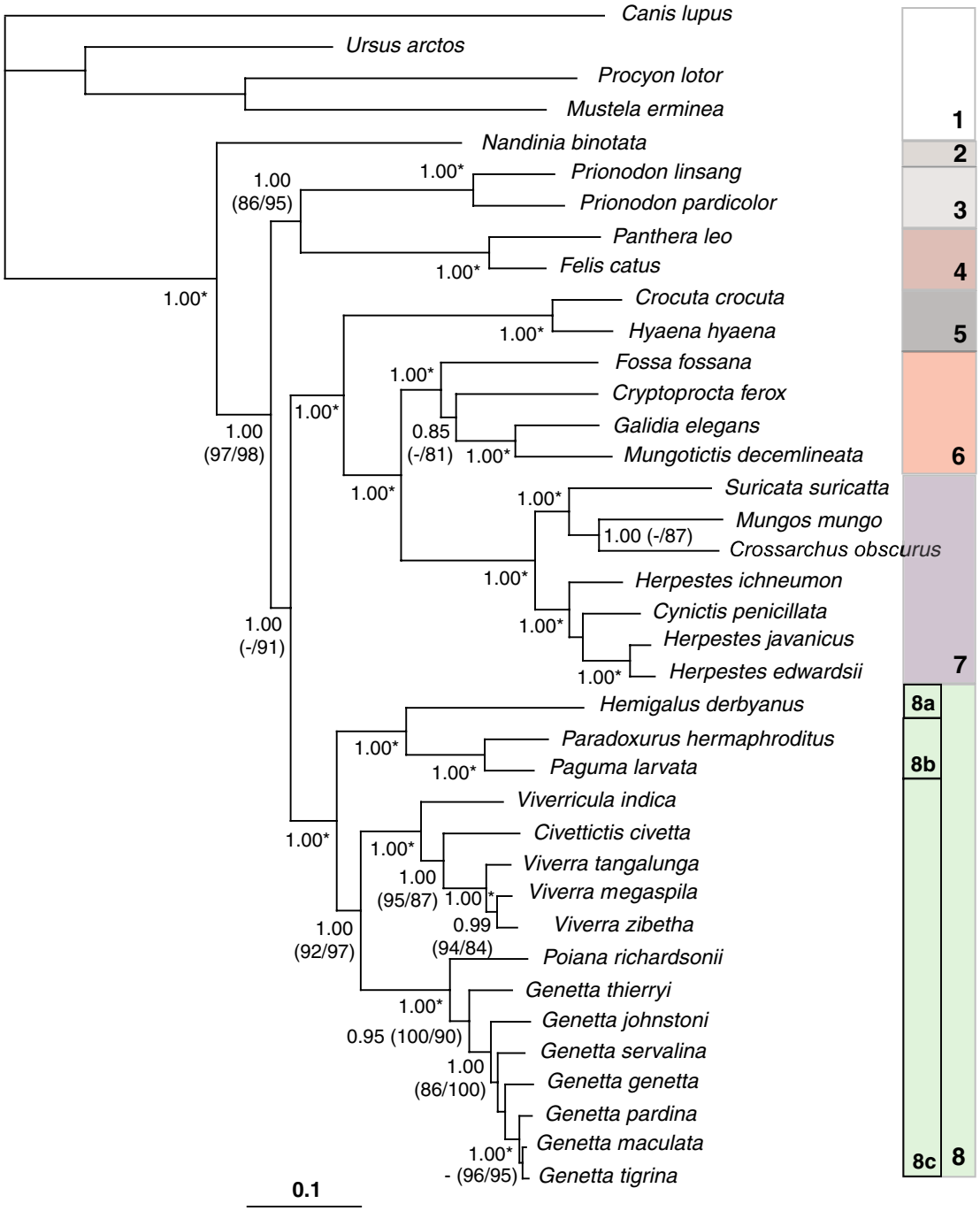

Fig. 2. Phylogram inferred from Bayesian analysis of the combined data set (cytb–TriI–IRBP1; 2943 bp). The model of sequence evolution is GTR+Γ+I. Values at nodes indicate Bayesian posterior probabilities >0.95 and bootstrap values >75% for MP and ML analyses, respectively (between parentheses). Bayesian posterior probabilities followed by asterisk represent nodes recovered with 100% of support in both MP and ML analyses. Scale bar corresponds to 10% sequence divergence. See Fig. 1 for numbers in boxes.

11.54 Myr (9.5–13.3 Myr—Gaubert and Veron, 2003) and 1.51 (1.42–2.51 Myr—Gaubert et al., 2004a), respectively. The origin of Eupleridae at 24.83 Myr approached the 18–24 Myr bracket of Yoder et al. (2003). On the other hand, estimates for deeper nodes yielded values older than previously suggested, as concerns for instance (*Felidae*, *Prionodon*): 42.32 Myr; Viverridae: 34.29 Myr; and Feliformia: 54.56 Myr (see Gaubert and Veron, 2003; Koepfli et al., 2006). The split ((Hyaenidae, (Herpestidae, Eupleridae)), Viverridae) was estimated to appear in the Middle Eocene (44.18 Myr). We calculated the minimum divergence time of the genus *Prionodon* (previously included in the Viverrinae) at the Middle Miocene (13.20 Myr). Within the Viverridae, the minimum divergence date for the separation between the Asian subfamilies Hemigalinae and Paradoxurinae was 23.83 Myr (Late Oligocene–Early Miocene), with the origin of the Paradoxurinae fixed to 9.57 Myr (Late Miocene). The origin of the subfamily Viverrinae was estimated as

Table 4
Estimates of genetic distances among viverrid lineages

	1	2	3	4
1. Genets and oyans	90.79 ± 5.19			
2. Terrestrial civets	183.93 ± 10.17	116.70 ± 7.60		
3. Paradoxurinae	203.88 ± 11.67	193.10 ± 10.86	120.00 ± 10.52	
4. Hemigalinae	213.38 ± 12.00	206.20 ± 11.81	183.00 ± 11.63	NA

Under diagonal, mean number of nucleotide differences ± SE between viverrid lineages; in diagonal (grey background), mean number of nucleotide differences ± SE within viverrid lineages.

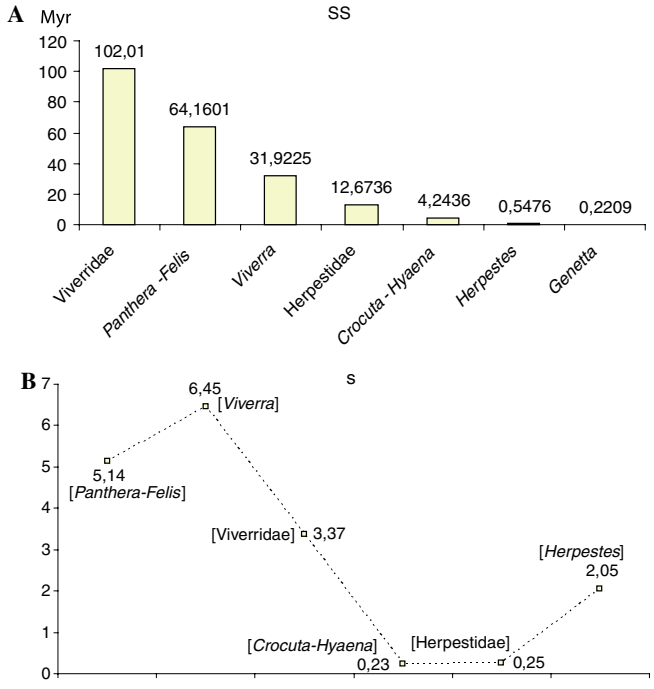

Fig. 3. Results of the fossil cross-validation method as implemented in r8s. (A) Squared differences (SS) between each fossil calibration and molecular estimate, ranked from greatest to smallest values. (B) Magnitude of average squared deviations calculated after a step-by-step removal of the most inconsistent fossil calibrations.

occurring in the Late Oligocene (28.49 Myr). The terrestrial civets appeared during the Middle Miocene (16.17 Myr), with a split between the African *Civettictis* and the Asian *Viverra* during the same geological period (12.26 Myr). The extant representatives of the genus *Viverra* were given a minimum date of origin in the Early Pliocene (4.60 Myr), as well as for the *Genetta* clade excluding *G. thierryi* (5.21 Myr).

The parsimony-based ancestral reconstruction using MacClade yielded unambiguous estimates (Fig. 5), including Asian origin for the family Viverridae, the subfamily Viverrinae, the terrestrial civets and the clade (*Civettictis*, *Viverra*). The lineage leading to (*Poiana*, *Genetta*) was attributed an African origin. The ML-based calculations obtained from Discrete gave the same results as parsimony, whether the "local" or "global" options were considered (Fig. 5). All the ancestral estimates obtained using the "local" method were significant (LR > 2).

4. Discussion

4.1. Phylogenetic re-assessment of the systematics of the Viverrinae

The combined analysis of cytb, TriI, and IRBP1 allowed us to propose an almost fully resolved phylogenetic tree of feliformians (Fig. 2). Our results gave strong support to the monophyly of (1) (Prionodontidae, Felidae), (2) (Viverridae, (Hyaenidae, (Herpestidae, Eupleridae))) and (3) Viverrinae, thus clarifying phylogenetic hypotheses within the group (Flynn et al., 2005; Gaubert and Veron, 2003; Gaubert et al., 2004b; Yoder et al., 2003). The phylogenetic affinities were also resolved among the terrestrial civets, the sister-group relationship between *Viverricula* and the clade (*Civettictis*, *Viverra*) being strongly supported.

Our analyses revealed substantial evolutionary distinctiveness, within Viverridae, between the terrestrial civets and the clade (*Poiana*, *Genetta*). First, the estimated genetic distances between the two lineages were similar to those relative to subfamilies Paradoxurinae and Hemigalinae (Table 4). Second, divergence time estimates fixed a deep splitting time between both Paradoxurinae and Hemigalinae, and the terrestrial civets and (*Poiana*, *Genetta*), around the Late Oligocene–Early Miocene (Fig. 4); the two latter lineages were estimated to appear in the Middle Miocene, a little earlier than what was calculated for the subfamily Paradoxurinae (Late Miocene; origin of Hemigalinae not calculable here). If we now consider the stringent morphological differences that distinguish the terrestrial civets from the oyans and genets (Gregory and Hellman, 1939; Nowak, 1999; Pocock, 1915; Veron, 1999), then genetic, divergence time, and morphological evidences support the splitting of the subfamily Viverrinae into two distinct subfamily level taxa. We propose the erection of the new subfamily Genettinae for designating the clade (*Poiana*, *Genetta*). We provide below diagnostic morphological characters and a list of extant genera for the two taxa newly defined following previous investigations (Gaubert et al., 2002; Gaubert, 2003; Gaubert and Veron, 2003; Gaubert et al., 2005b; Pocock, 1915; Wozencraft, 1993).

4.1.1. Subfamily Viverrinae Gray, 1821

Diagnosis: Feet digitigrade; thenar and hypothenar pads absent or residual; ratio between head + body and tail >1.5; black- and white-striped "collar" on the neck; perineal glands with a deep interglandular poach for musk storage (in females); caudal entotympanic bone ventrally inflated, showing a pyramidal shape; paraoccipital process strong, exhibiting a ventral prolongation; thick dentition, with a well-developed talonid on M_1.

- Genus *Viverra* Linné, 1758 [type genus]
- Genus *Viverricula* Hodgson, 1838
- Genus *Civettictis* Pocock, 1915

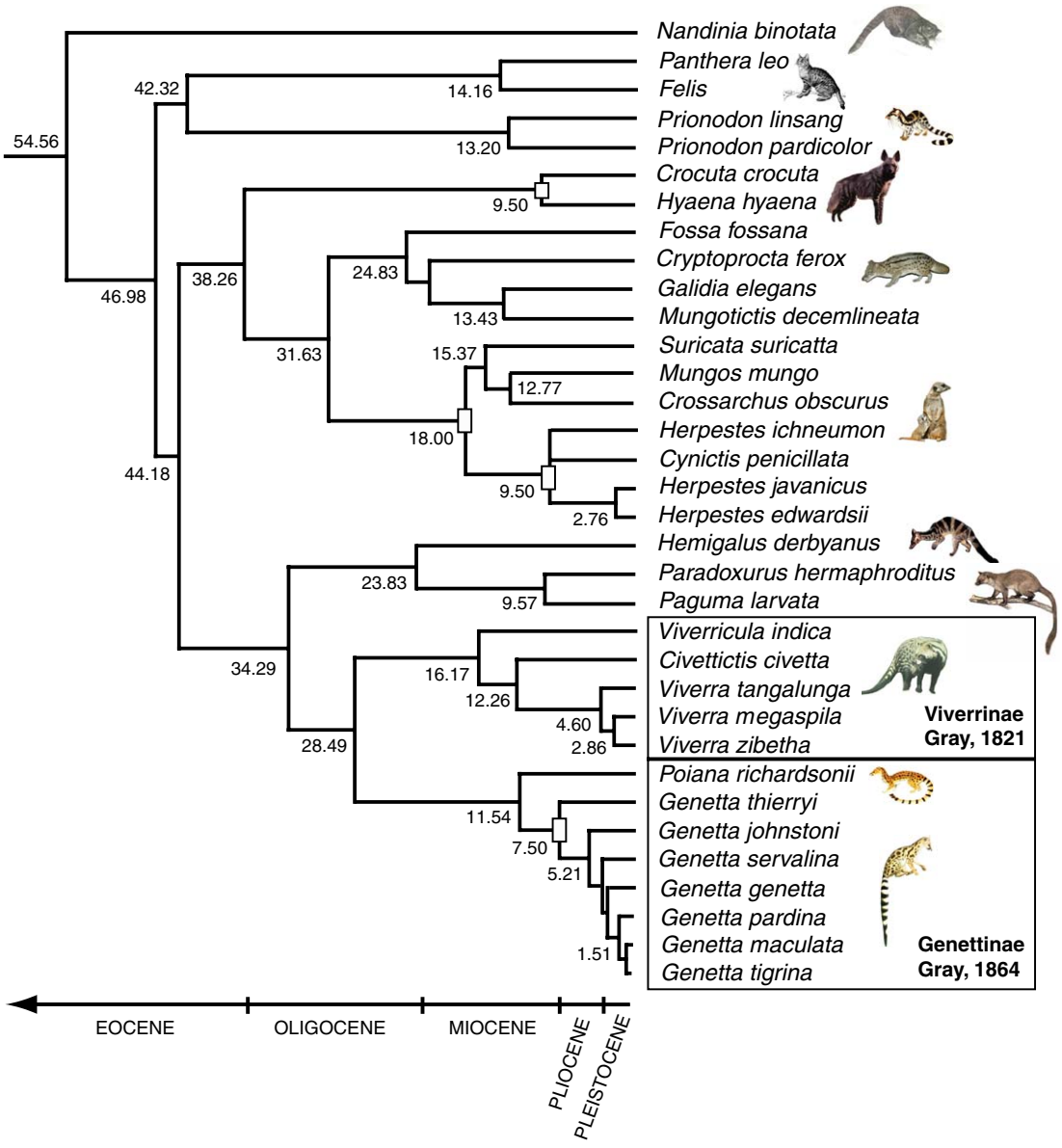

Fig. 4. Linearized tree showing minimum divergence time estimates (in Myr) within feliformians. White squares at nodes represent the four calibration points used in the final analysis. The taxonomic partition proposed by the authors within the traditional Viverrinae is indicated (see Section 4.1).

4.1.2. Subfamily Genettinae Gray, 1864 [following Principle of Coordination—Article 36.1; International Commission on Zoological Nomenclature (1999)]

Diagnosis: Posterior feet digitigrade and anterior feet plantigrade (semi-digitigrady); thenar and hypothenar pads well-developed; ratio between head + body and tail <1.5; facial mask with bright supra-orbital and supra-labial pairs of spots; perineal glands (absent in *Poiana*) with a shallow interglandular poach for musk storage (in females); caudal entotympanic bone weakly inflated, showing a globular shape; paraoccipital process rounded and short; dentition thin and sharp, with a short talonid on M_1.

- Genus *Genetta* G. Cuvier, 1816 [type genus]
- Genus *Poiana* Gray, 1865

4.2. Divergence time estimates, fossil records, and faunal exchanges between Asia and Africa

The fossil cross-validation method allowed us to assess the level of consistency among the different calibration points we had defined within feliformians. The application of this method showed that the calibration point used for *Felis-Panthera* (3.5 Myr) by Gaubert and Veron (2003) was underestimated, as recently confirmed by Johnson et al. (2006), whom estimated divergence time between the *Panthera* lineage and the rest of extant felids at ca. 10.8 Myr. Fossil cross-validation is thus of interest because it is likely to temper bias due to taxonomic misattribution of fossils and incompleteness of paleontological records; biases that are usually not taken into account in the routinely used

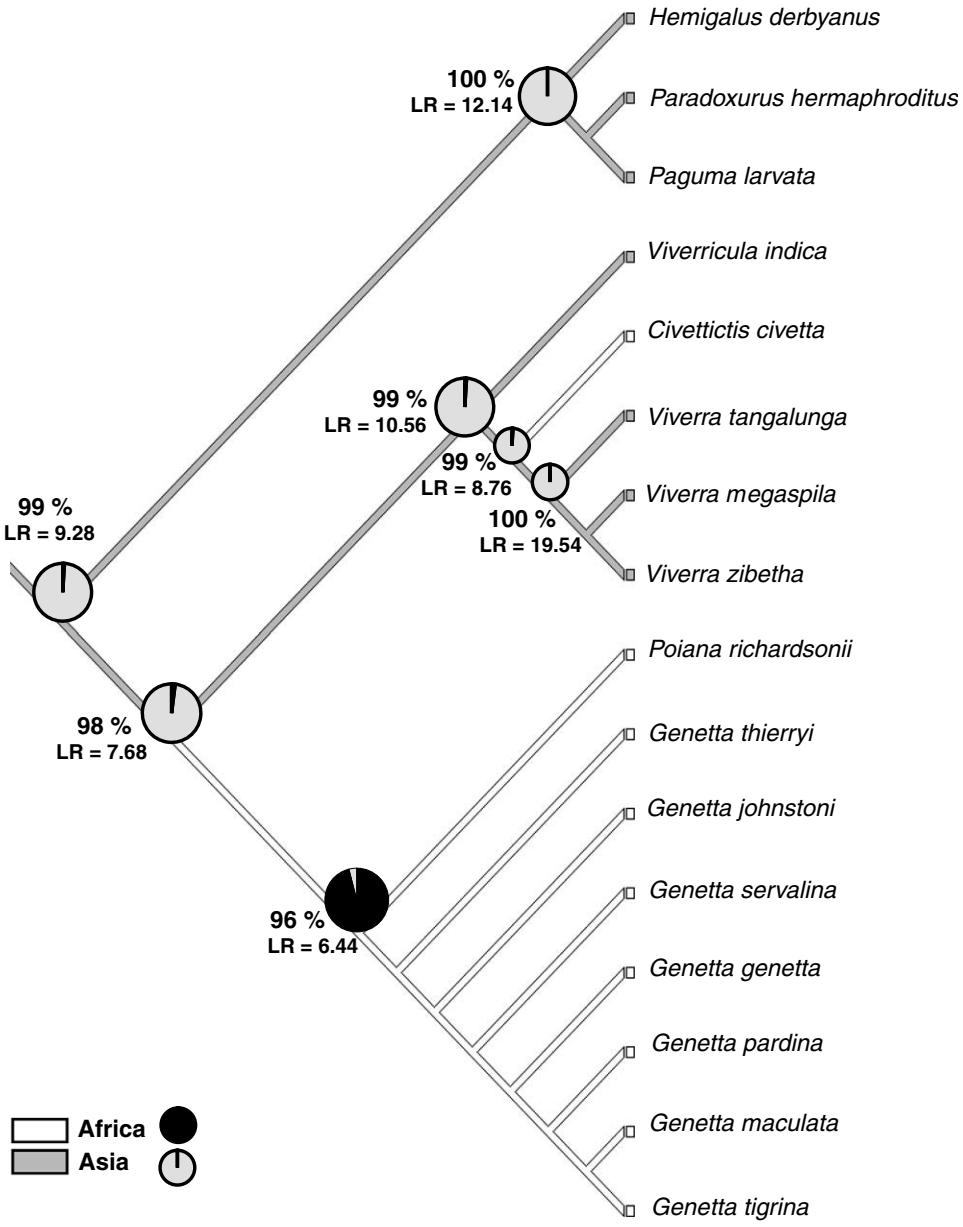

Fig. 5. Reconstruction of ancestral areas among the Viverridae. Parsimony-based estimates (MacClade) are superimposed on the tree. Maximum likelihood-based calculations (Discrete) are given as circles at nodes. Percentages with LR values correspond to the proportion of most probable ancestral state obtained using the "local" method.

method of simultaneous "independent" calibrations (see Thorne and Kishino, 2002). For instance, the simultaneous use of the seven calibration points first defined in our study yielded significantly younger divergence time estimates all along the tree (e.g., 28.41 Myr for (Prionodontidae, Felidae), 24.99 Myr for (Hyaenidae, (Herpestidae, Eupleridae)), and 20.54 Myr for (Viverrinae, Genettinae); other data not shown) because several calibrations with under-estimated ages were included. On the other hand, the utility of this method may be dependent on the series of calibration points selected for the analysis, a series of inconsistent calibration points including one "correct" datum potentially diagnosing as inconsistent the only valid fossil reference.

Our results suggested two independent migration events for the Viverridae from Asia to Africa. One was estimated to occur during the Middle Miocene (leading to *Civettictis*), and the other one occurring sometime between the Late Oligocene and the Middle Miocene (Genettinae). The absence of fossil taxa prior to 7.5 Myr attributable to the Genettinae hampers the definition of a more precise geological period to date this event. Nevertheless, these results confirm the hypothesis of Miocene routes from Asia to Africa across the Arabian microplate (Cox and Moore, 1993) that would have involved several independent events of migrations (Juste et al., 1999; Johnson et al., 2006).

Our findings, in terms of divergence times and biogeographic scenarios, can be compared with the available viverrid fossil record, even if its systematics and phylogenetic reconstruction are still poorly comprehended. Estimated times of appearance, geographical distribution and

morphological characters of fossils might then be viewed under a different light when framed into the inferred migration patterns within Viverridae.

We found inconsistency of *Herpestides* as a calibration point for the family Viverridae (fossil record: 23–25 Myr; divergence time estimates: 34.29 Myr), although Hunt (1991) recognized *Herpestides*, as the oldest fossil showing a "true" viverrid morphotype (Late Oligocene–Early Miocene of Eurasia). We observed that the dental and cranial morphology of *Herpestides* shared similarities with those characterizing terrestrial civets and genets, but clearly differed from the peculiar morphotypes -especially teeth- of Hemigalinae and Paradoxurinae (F. Solé and P. Gaubert, unpubl. data; see also Hunt, 2001). One plausible hypothesis would be to consider *Herpestides* as representing a fossil lineage of the extant Viverrinae and Genettinae, which would actually fit with our divergence time estimates for the clade (28.49 Myr).

The Eurasian origin of the clade Viverrinae–Genettinae is supported by the existence of another viverrine-like fossil genus from the Early Miocene of Eurasia, *Semigenetta* (Montoya et al., 2001; Qiu and Gu, 1986). Although our analyses yielded unambiguous estimations concerning the geographical origin of the family Viverridae, doubts remain over its precise location in Eurasia, especially given the lack of fossil records attributable to Paradoxurinae and Hemigalinae (Hunt, 1996; but see Morales et al., 2000).

Our estimations based on extant taxa are congruent with the fossil record in suggesting migration routes towards Africa during the Early and Middle Miocene, with the first viverrid fossil (civet-like) mentioned from Africa ca. 17.5 Myr ago (*Orangictis*—Morales et al., 2001). Posterior to this period, the fossil record however suggests that migration patterns within terrestrial civets and civet-like fossils were more complex. The peak of taxonomic diversity in Eurasia and Africa from Late Miocene to Late Pliocene (De Beaumont, 1973; Geraads, 1997; Hunt, 1996; Petter and Howell, 1977; Pilgrim, 1932; Rook and Martinez-Navarro, 2004) indeed suggests (1) additional migration events to Africa *via* the Arabian plate and possibly the Messinian junction between southwestern Europe and North Africa (Cox, 2000), and (2) subsequent retro-migrations to Eurasia. Migration routes may have persisted until the Early Pliocene, from which started the desertification of the Arabian Peninsula (Anton, 1984) and the opening of the strait of Gibraltar (Krijgsman, 2002). Phylogenetic relationships within the clade *Herpestes* (including *Cynictis*; family Herpestidae) also supported the hypothesis of a retro-migration from Africa to Asia, posterior to 9.50 Myr, as illustrated by the Asian clade (*H. javanica*, *H. edwardsii*).

The calibration point used for *Viverra* (9 Myr) proved highly inconsistent according to the cross-validation method. Indeed, we estimated the origin of the extant genus during the Middle Pliocene (4.60 Myr), a much later period than what was suggested by the fossil record (Petter and Howell, 1977). This result suggests taxonomic attributions misled by morphological convergences within *Viverra*-like fossils, and emphasizes the critical need for a taxonomic revision of the genus.

Finally, our study illustrates the necessity of carefully assessing the consistency of fossil calibrations when using multiple points for estimating divergence times. It also points out the necessity of integrating knowledge in fossil taxonomy in order to avoid "extantalism" (interpretation strictly based on patterns derived from extant taxa) when attempting to reconstruct evolutionary scenarios. Our results suggested that geological and molecular estimates were in agreement concerning the time of origin of Hyaenidae, Herpestidae, *Herpestes*, and *Genetta*. On the other hand, they pointed out the critical need for additional paleontological data and investigations concerning the origin of Felidae, Viverridae, and *Viverra*.

Acknowledgments

We are deeply grateful to the following people for having contributed to the taxonomic sampling effort: Raoul Benveniste (Laboratory of Genomic Diversity, Frederick, MD), François Catzeflis (ISEM, Université Montpellier 2, France), Christiane Denys and Vitaly Volobouev (Muséum National d'Histoire Naturelle, Paris, France), Amy Dunham (Harvard University, Cambridge, MA), Pascal Fournier (Mission Vison d'Europe, Villandraut, France), Laurent Granjon (Institut de Recherche pour le Développement, Bamako, Mali), Lon Grassman (Ceasar Kleberg Wildlife Research Institute, Kingsville, TX), Eric Harley (University of Cape Town, South Africa), Julian Kerbis (The Field Museum, Chicago, IL), Sébastien Lavoué (Ocean Research Institute, University of Tokyo, Japan), Francisco Palomares (Estacion Biologica de Doñana, Spain), Terri Roth (Cincinnati Zoo & Botanical Garden, OH), and Joe Walston (Wildlife Conservation Society, Cambodia). Chris W. Wozencraft and Peter J. Taylor greatly improved the early version of the manuscript. We are grateful to Lars Werdelin and Floréal Solé for fruitful discussions on feliformian fossils. PG thanks Louis Deharveng, Eric Pasquet and Géraldine Veron (Unité OSEB/CNRS UMR 5202 – Muséum National d'Histoire Naturelle, Paris, France) for having supported the molecular work at the Service Commun de Systématique Moléculaire (CNRS IFR 1541, Muséum National d'Histoire Naturelle, Paris, France). PG is thankful to Georges Gaubert for providing computer material. PCE was financed by CAPES Grant No. 0908201-6 (Ministry of Education of Brazil).

References

Abdo, Z., Minin, V.N., Joyce, P., Sullivan, J., 2004. Accounting for uncertainty in the tree topology has little effect on the decision–theoretic approach to model selection in phylogeny estimation. Mol. Biol. Evol. 22, 691–703.

Antoine, P.-O., Ducrocq, S., Marivaux, L., Chaimanee, Y., Crochet, J.-Y., Jaeger, J.-J., Welcomme, J.-L., 2003. Early rhinocerotids (Mammalia: Perissodactyla) from South Asia and a review of the Holarctic Paleogene rhinocerotid record. Can. J. Earth Sci. 40, 365–374.

Anton, D., 1984. Aspects of geomorphological evolution; paleosols and dunes in Saudi Arabia. In: Jado, A.R., Zotl, J.G. (Eds.), Quaternary Period in Saudi Arabia. Springer-Verlag, Vienna, pp. 275–296.

Arnason, U., Bodin, K., Gullberg, A., Ledje, C., Mouchaty, S., 1995. A molecular view of pinniped relationships with particular emphasis on the true seals. J. Mol. Evol. 40, 78–85.

Barry, J.C., 1983. *Herpestes* (Viverridae, Carnivora) from the Miocene of Pakistan. J. Paleontol. 57, 150–156.

Burnham, K.P., Anderson, D.R., 2002. Model Selection and Multimodel Inference: A Practical Information-Theoretic Approach. Springer Verlag, New York.

Chevret, P., Dobigny, G., 2005. Systematics and evolution of the subfamily Gerbillinae (Mammalia, Rodentia, Muridae). Mol. Phylogenet. Evol. 35, 674–688.

Cox, C.B., Moore, P.D., 1993. Biogeography: An Ecological and Evolutionary Approach. Blackwell Science, Oxford.

Cox, C.B., 2000. Plate tectonics, seaways and climate in the historical biogeography of mammals. Mem. Inst. Oswaldo Cruz, Rio de Janeiro 95, 509–516.

Davis, C.C., Bell, C.D., Fritsch, P.W., Mathews, S., 2002. Phylogeny of Acridocarpus–Brachylophon (Malpighiaceae): implications for tertiary tropical floras and Afroasian biogeography. Evolution 56, 2395–2405.

De Beaumont, G., 1973. Contribution à l'étude des Viverridés (Carnivora) du Miocène d'Europe. Arch. Sci. Genève 26, 285–296.

Felsenstein, J., 1985. Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39, 783–791.

Flynn, J.J., Nedbal, M.A., 1998. Phylogeny of the Carnivora (Mammalia): congruence vs incompatibility among multiple data sets. Mol. Phylogenet. Evol. 9, 414–426.

Flynn, J.J., Finarelli, J.A., Zehr, S., Hsu, J., Nedbal, M.A., 2005. Molecular phylogeny of the Carnivora (Mammalia): assessing the impact of increased sampling on resolving enigmatic relationships. Syst. Biol. 54, 317–337.

Gaubert, P., Veron, G., Tranier, M., 2002. Genets and "genet-like" taxa (Carnivora, Viverrinae): Phylogenetic analysis, systematics and biogeographic implications. Zool. J. Linn. Soc. 134, 317–334.

Gaubert, P., 2003. Systématique et phylogénie du genre *Genetta* et des énigmatiques "genet-like taxa" *Prionodon, Poia*na e*t Osbornict*is (Carnivora, Viverridae): caractérisation de la sous-famille des Viverrinae et étude des patrons de diversification au sein du continent africain. Unpublished PhD Thesis, Muséum National d'Histoire Naturelle, Paris, France.

Gaubert, P., Veron, G., 2003. Exhaustive sample set among Viverridae reveals the sister-group of felids: the linsangs as a case of extreme morphological convergence within Feliformia. Proc. R. Soc. Lond. Biol. Sci. 270, 2523–2530.

Gaubert, P., Fernandes, C.A., Bruford, M.W., Veron, G., 2004a. Genets (Carnivora, Viverridae) in Africa: an evolutionary synthesis based on cytochrome *b* sequences and morphological characters. Biol. J. Linn. Soc. 81, 589–610.

Gaubert, P., Tranier, M., Delmas, A.-S., Colyn, M., Veron, G., 2004b. First molecular evidence for reassessing phylogenetic affinities between genets (*Genetta*) and the enigmatic genet-like taxa *Osbornictis, Poiana* and *Prionodon* (Carnivora, Viverridae). Zool. Scr. 33, 117–129.

Gaubert, P., Taylor, P.J., Fernandes, C.A., Bruford, M.W., Veron, G., 2005a. Patterns of cryptic hybridization revealed using an integrative approach: a case study on genets (Carnivora, Viverridae, *Genetta* spp.) from the southern African subregion. Biol. J. Linn. Soc. 86, 11–33.

Gaubert, P., Taylor, P.J., Veron, G., 2005b. Integrative taxonomy and phylogenetic systematics of the genets (Carnivora, Viverridae, genus *Genetta*): a new classification of the most speciose carnivoran genus in Africa. In: Huber, B.A., Sinclair, B.J., Lampe, K.-H. (Eds.), African Biodiversity: Molecules, Organisms, Ecosystems. Springer, New York, pp. 371–383.

Gaubert, P., Wozencraft, W.C., Cordeiro-Estrela, P., Veron, G., 2005. Mosaic of convergences, noise and misleading morphological phylogenies: what's in a viverrid-like carnivoran? Syst. Biol. 54, 865–894.

Geraads, D., 1997. Carnivores du Pliocène terminal de Ahl al Oughlam (Casablanca, Maroc). Geobios 30, 127–164.

Grandcolas, P., Guilbert, E., Robillard, T., D'Haese, C.A., Murienne, J., Legendre, F., 2004. Mapping characters on a tree with or without the outgroups. Cladistics 20, 1–4.

Green, P.J., Silverman, B.W., 1994. Nonparametric Regression and Generalized Linear Models. Chapman and Hall, London.

Gregory, W.K., Hellman, M., 1939. On the evolution and major classification of the civets (Viverridae) and allied fossil and recent Carnivora: a phylogenetic study of the skull and dentition. Proc. Am. Philos. Soc. 81, 309–392.

Griffin, D.L., 2002. Aridity and humidity: two aspects of the late Miocene climate of North Africa and the Mediterranean. Paleogeogr. Paleoclimatol. Paleocol. 182, 65–91.

Hall, T.A., 1999. BioEdit: a user-friendly biological sequence alignment and analysis program for Windows 95/98/NT. Nucleic Acid Symp. 41, 95–98.

Hasegawa, M., Kishino, H., Yano, T.-A., 1985. Dating of the human-ape splitting by a molecular clock of mitochondrial DNA. J. Mol. Evol. 22, 160–174.

Hosoda, T., Suzuki, H., Harada, M., Tsuchiya, K., Han, S.H., Zhang, Y., Kryukov, A.P., Lin, L.K., 2000. Evolutionary trends of the mitochondrial lineage differentiation in species of genera *Martes* and *Mustela*. Genes Genet. Syst. 75, 259–267.

Huchon, D., Douzery, E.J.P., 2001. From the Old World to the New World: a molecular chronicle of the phylogeny and biogeography of hystricognath rodents. Mol. Phylogenet. Evol. 20, 238–251.

Hunt Jr., R.M., 1991. Evolution of the aeluroid Carnivora: viverrid affinities of the Miocene carnivoran *Herpestides*. Am. Mus. Novit. 3023, 1–34.

Hunt Jr., R.M., 1996. Biogeography of the order Carnivora. In: Gittleman, J.L. (Ed.), Carnivore Behavior, Ecology and Evolution. Cornell University Press, New York, pp. 451–485.

Hunt Jr., R.M., 2001. Basicranial anatomy of the living Linsangs *Prionodon* and *Poiana* (Mammalia, Carnivora, Viverridae), with comments on the early evolution of Aeluroid Carnivora. Am. Mus. Novit. 3330, 1–23.

International Commission on Zoological Nomenclature, 1999. International Code of Zoological Nomenclature. The International Trust for Zoological Nomenclature, London, United Kingdom.

Johnson, W.E., Eizirik, E., Pecon-Slattery, J., Murphy, W.J., Antunes, A., Teeling, E., O'Brien, S.J., 2006. The Late Miocene radiation of modern Felidae: a genetic assessment. Science 311, 73–77.

Juste, J.B., Alvarez, Y., Tabares, E., Garrido-Pertierra, A., Ibanez, C., Bautista, J.M., 1999. Phylogeography of African fruitbats (Megachiroptera). Mol. Phylogenet. Evol. 13, 596–604.

Kappelman, J., Rasmussen, D.T., Sanders, W.J., Feseha, M., Bown, T., Copeland, P., Crabaugh, J., Fleagle, J., Glantz, M., Gordon, A., Jacobs, B., Maga, M., Muldoon, K., Pan, A., Pyne, L., Richmond, B., Ryan, T., Seiffert, E.R., Sen, S., Todd, L., Wlemann, M.C., Winkler, A., 2003. Oligocene mammals from Ethiopia and faunal exchange between Afro-Arabia and Eurasia. Nature 426, 549–552.

Kimura, M., 1981. Estimation of evolutionary distances between homologous nucleotide sequences. Proc. Natl. Acad. Sci. USA 78, 454–458.

Kocher, T.D., Thomas, W.K., Meyer, A., Edwards, S.V., Pääbo, S., Villablanca, F.X., Wilson, A.C., 1989. Dynamics of mitochondrial DNA evolution in animals: amplification and sequencing with conserved primers. Proc. Natl. Acad. Sci. USA 86, 6196–6200.

Koepfli, K.-P., Jenks, S.M., Eizirik, E., Zahirpour, T., Valkenburgh, B.V., Wayne, R.K., 2006. Molecular systematics of the Hyaenidae: relationships of a relictual lineage resolved by a molecular supermatrix. Mol. Phylogenet. Evol. 38, 603–620.

Kosuch, J., Vences, M., Dubois, A., Ohler, A., Böhme, W., 2001. Out of Asia: mitochondrial DNA evidence for an oriental origin of tiger frogs, genus *Hoplobatrachus*. Mol. Phylogenet. Evol. 21, 398–407.

Krijgsman, W., 2002. The Mediterranean: *Mare Nostrum* of Earth sciences. Earth Planet. Sci. Lett. 205, 1–12.

Kumar, S., Tamura, K., Nei, M., 2004. MEGA3: integrated software for molecular evolutionary genetics analysis and sequence alignment. Brief. Bioinform. 5, 150–163.

Ledje, C., Arnason, U., 1996. Phylogenetic analyses of complete cytochrome *b* genes of the order Carnivora with particular emphasis on the Caniformia. J. Mol. Evol. 42, 135–144.

Maddison, W.P., Maddison, D.R., 1992. MacClade: Analysis of Phylogeny and Character Evolution. Sinauer Associates, Sunderland, MA.

Métais, G., Chaimanee, Y., Jaeger, J.-J., Ducrocq, S., 2001. New remains of primitive ruminants from Thailand: evidence of the early evolution of the Ruminantia in Asia. Zool. Scr. 30, 231–248.

Minin, V., Abdo, Z., Joyce, P., Sullivan, J., 2003. Performance-based selection of likelihood models for phylogeny estimation. Syst. Biol. 52, 1–10.

Montoya, P., Alcala, L., Morales, J., 2001. First find of a Viverridae (Carnivora, Mammalia) in the Upper Miocene of the Teruel Basin (Spain). Bol. Soc. Esp. Hist. Nat. Secc. Geol. 96, 101–109.

Mooers, A.O., Schluter, D., 1999. Reconstructing ancestor states with maximum likelihood: support for one- and two-rate models. Syst. Biol. 48, 623–633.

Morales, J., Pickford, M., Salesa, M., Soria, D., 2000. The systematic status of *Kelba*, Savage, 1965, *Kenyalutra*, Schmidt-Kittler, 1987 and *Ndamathaia*, Jcobs et al., 1987, (Viverridae, Mammalia) and a review of Early Miocene mongoose-like carnivores of Africa. Annu. Paleontol. 86, 243–251.

Morales, J., Pickford, M., Soria, D., Fraile, S., 2001. New Viverrinae (Carnivora, Mammalia) from the basal Middle Miocene of Arrisdrift (Namibia). Paleontol. Afr. 37, 99–102.

Near, T.J., Sanderson, M.J., 2004. Assessing the quality of molecular divergence time estimates by fossil calibrations and fossil-based model selection. Philos. Trans. R. Soc. Lond. B 359, 1477–1483.

Near, T.J., Bolnick, D.I., Wainwright, P.C., 2005a. Fossil calibrations and molecular divergence time estimates in centrarchid fishes (Teleostei: Centrarchidae). Evolution 59, 1768–1782.

Near, T.J., Meylan, P.A., Shaffer, H.B., 2005b. Assessing concordance of fossil calibration points in molecular clock studies: an example using turtles. Am. Nat. 165, 137–146.

Nei, M., Kumar, S., 2000. Molecular Evolution and Phylogenetics. Oxford University Press, New York.

Nowak, R.M., 1999. Walker's Mammals of the World. Johns Hopkins University Press, Baltimore, MD.

Pagel, M., 1994. Detecting correlated evolution on phylogenies: a general method for the comparative analysis of discrete characters. Proc. R. Soc. Lond. B 255, 37–45.

Pagel, M., 1999. The maximum likelihood approach to reconstructing ancestral character states of discrete characters on phylogenies. Syst. Biol. 48, 612–622.

Pagel, M., 2000. User's Manual for Discrete. University of Reading, Reading, UK.

Petter, G., Howell, F.C., 1977. Diversification des civettes (Carnivora, Viverridae) dans les gisements pléistocènes de l'Omo. C. R. Acad. Sci. Paris D 284, 283–286.

Pilgrim, G.E., 1932. The fossil Carnivora of India. Mem. Geol. Surv. India Pal. Indica, New Ser. 18, 1–232.

Pocock, R.I., 1915. On the feet and glands and other external characters of the Viverrinae, with the description of a new genus. Proc. Zool. Soc. Lond. 1915, 131–149.

Pol, D., 2004. Empirical problems of the Hierarchical likelihood ratio test for model selection. Syst. Biol. 53, 949–962.

Posada, D., Crandall, K.A., 1998. MODELTEST: testing the model of DNA substitution. Bioinform. Appl. Note 14, 817–818.

Posada, D., 2004. ModelTest 3.6. University of Vigo, Spain.

Qiu, Z., Gu, Y., 1986. The Middle Miocene vertebrate fauna from Xiacaowan, Sihong Co., Jiangsu Province. 3. Two species of fossil carnivores: *Semigenetta* and *Pseudaelurus*. Vertebr. PalAsiatic. 24, 1–31.

Renner, S.S., Zhang, L.-B., Murata, J., 2004. A chloroplast phylogeny of *Arisaema* (Araceae) illustrates Tertiary floristic links between Asia, North America, and East Africa. Am. J. Bot. 91, 881–888.

Ronquist, F., Huelsenbeck, J.P., van der Mark, P., 2005. MrBayes 3.1 Manual. Florida State University, Tallahassee, FL.

Rook, L., Martinez-Navarro, B., 2004. *Viverra howelli* n. sp., a new Viverrid (Carnivora, Mammalia) from the Baccinello-Cinigiano basin (Latest Miocene, Italy). Riv. Ital. Paleontol. Strategy 110, 719–723.

Ropiquet, A., Hassanin, A., 2004. Molecular phylogeny of caprines (Bovidae, Antilopinae): the question of their origin and diversification during the Miocene. J. Zool. Syst. Evol. Res. 43, 49–60.

Roth, C., 1988. *Leptoplesictis* Major 1903 (Mammalia, Carnivora, Viverridae) aus dem Orleanium und Astaracium/Miozän von Frankreich und Deutschland. Paleontol. Zeit. 62, 333–343.

Rychlik, W., 1992. OLIGO 4.0—OLIGO Primer Analysis Software. National Biosciences, Plymouth, MN.

Sanderson, M.J., 2002. Estimating absolute rates of molecular evolution and divergence times: a penalized likelihood approach. Mol. Biol. Evol. 19, 101–109.

Sanderson, M.J., 2003. r8s; inferring absolute rates of evolution and divergence times in the absence of a molecular clock. Bioinformatics 19, 301–302.

Sanderson, M.J., 2004. r8s, version 1.70. User's Manual. University of California, Davis, CA.

Sato, J.J., Hosoda, T., Wolsan, M., Suzuki, H., 2004. Molecular phylogeny of arctoids (Mammalia: Carnivora) with emphasis on phylogenetic and taxonomic positions of the ferret-badger and skunks. Zool. Sci. 21, 111–1118.

Schmidt-Kittler, N., 1987. The Carnivora (Fissipedia) from the Lower Miocene of East Africa. Paleontol. Abt. A 197, 85–126.

Swofford, D.L., Maddison, W.P., 1987. Reconstructing ancestral character states under Wagner parsimony. Math. Biosci. 87, 199–229.

Swofford, D.L., 2002. PAUP* 4.0: phylogenetic analysis using parsimony (* and other methods). Simmer Associates, Sunderland, MA.

Tavaré, S., 1986. Some probabilistic and statistical problems in the analysis of DNA sequences. In: Miura, R.M. (Ed.), Some Mathematical Questions in Biology—DNA Sequence Analysis. American Mathematic Society, Providence, pp. 57–86.

Thorne, J.L., Kishino, H., 2002. Divergence time and evolutionary rate estimation with multilocus data. Syst. Biol. 51, 689–702.

Veron, G., 1999. Pads morphology in the Viverridae (Carnivora). Act. Ther. 44, 363–376.

Veron, G., Heard, S., 2000. Molecular systematics of the Asiatic Viverridae (Carnivora) inferred from mitochondrial cytochrome *b* sequence analysis. J. Zool. Syst. Evol. Res. 38, 209–217.

Veron, G., Colyn, M., Dunham, A.E., Taylor, P., Gaubert, P., 2004. Molecular systematics and origin of sociality in mongooses (Herpestidae, Carnivora). Mol. Phylogenet. Evol. 30, 582–598.

Werdelin, L., 1996. Carnivores, exclusive of Hyaenidae from the later Miocene of Europe and western Asia. In: Bernor, R.L., Fahlbusch, V., Mittmann, H.-W. (Eds.), The Evolution of Western Eurasian Neogene Mammal Faunas. Columbia University Press, New York, pp. 271–289.

Werdelin, L., 2003. Mio-Pliocene Carnivora from Lothagam, Kenya. In: Leakey, M.G., Harris, J.M. (Eds.), Lothagam: The Dawn of Humanity in Africa. Columbia University Press, New York, pp. 261–330.

Werdelin, L., Lewis, M.E., 2005. Plio-Pleistocene Carnivora of eastern Africa: species richness and turnover patterns. Zool. J. Linn. Soc. 144, 121–144.

Wozencraft, W.C., 1993. Order Carnivora. In: Wilson, D.E., Reeder, D.M. (Eds.), Mammal Species of the World—A Taxonomic and Geographic Reference. Smithsonian Institution Press, Washington & London, pp. 279–348.

Wozencraft, W.C., 2005. Order Carnivora. In: Wilson, D.E., Reeder, D.M. (Eds.), Mammals species of the world. A taxonomic and geographic reference. The Johns Hopkins University Press, Baltimore, MD, pp. 532–628.

Yang, Z., 1994. Maximum likelihood phylogenetic estimation from DNA sequences with variable rates over sites: Approximate methods. J. Mol. Evol. 39, 306–314.

Yoder, A.D., Burns, M.M., Zehr, S., Delefosse, T., Veron, G., Goodman, S.M., Flynn, J.J., 2003. Single origin of Malagasy Carnivora from an African ancestor. Nature 421, 734–737.