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Original article

Calumma vohibola, a new chameleon species (Squamata: Chamaeleonidae) from the littoral forests of eastern Madagascar

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Abstract.—We describe *Calumma vohibola* sp. nov., a morphologically distinct chameleon species of the *Calumma nasutum* species group from littoral forest fragments of the north-central east coast of Madagascar. Males and females of this species differ from all other species of the *Calumma nasutum* group by an almost absent rostral appendage, by a characteristic stress colouration in the female consisting of a dark reddish ground colouration with many irregular light blue spots, and by significant sequence divergence in the mitochondrial *ND2* gene (10.5–19.4% pairwise distance) and no haplotype sharing in the nuclear *C-mos* gene compared with other members of the *C. nasutum* group. A molecular survey of *C. nasutum* populations occurring near the type locality of the new species reveals an extraordinarily high genetic diversity within this morphologically conservative chameleon group. We suspect that *C. vohibola* might be restricted to a rather small and fragmented distribution range within the last littoral forest fragments along the coast. Owing to the unsolved taxonomic situation of the *C. nasutum* group, no reliable distribution data of any member of this group are available at present, and hence we cannot ascertain whether the extent of occurrence of *C. vohibola* is indeed as restricted as suspected. Intensive surveys and field studies are necessary to clarify the distribution limits of this new species and to assess its conservation status reliably.

Key words.—Chamaeleonidae, *Calumma vohibola*, *Calumma nasutum*, new species, genetic diversity, littoral forest

Introduction

Madagascar harbours one of the world's most diverse herpetofaunas, characterised by an extreme degree of endemism. The knowledge of the diversity and distribution of this unique herpetofauna has rapidly increased in the last two decades, but still the taxonomic status and especially the distribution of many of its amphibian and reptile species remains poorly known. The intensive research activity during recent years and the comprehensive use of integrative taxonomical approaches, combining

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molecular and morphological data (Padial *et al.* 2010), has led to the identification of many new species and improved our understanding of Madagascar's magnificent biodiversity (Vieites *et al.* 2009). Recent comprehensive taxonomic rearrangements revealed that numerous widespread species in fact represent species complexes, leading to their splitting into several separate species. As a consequence, the knowledge on distribution ranges of species is changing rapidly as well. For example, Raxworthy and Nussbaum (2006) recognised that the chameleon species *Calumma brevicorne* as previously understood was a complex of various distinct species, and reliably assigning historical records of *C. brevicorne* to any of the newly delimited species would have required extensive re-analysis. Therefore, each of the newly recognised species of this complex was left with just a few localities in the summarising account of Glaw and Vences (2007). The same situation seems to hold true for the common species *Calumma nasutum*, which according to current taxonomy is widely distributed in eastern Madagascar's rainforests but is suspected to represent a complex of several species as well (Glaw & Vences 2007).

The genus *Calumma* is currently composed of 31 species which are distributed in the humid rainforests and montane regions of eastern and central Madagascar (Gehring, Pabijan *et al.* 2010). A further species from the Seychelles previously assigned to this genus has recently been transferred to a separate genus as *Archaius tigris*, taking into account its close relationships to the African leaf chameleons of the genus *Rieppeleon* rather than to Malagasy species of *Calumma* (Townsend *et al.* 2011). Moreover, recent molecular studies suggest that the genus *Calumma* might not be monophyletic (Raxworthy *et al.* 2002; Townsend & Larson 2002; Townsend *et al.* 2011), although the available molecular phylogenetic data neither reliably support nor reject monophyly of the genus. The relationships between the morphologically defined phenetic species groups within the genus are not yet sufficiently resolved and even the monophyly of these groups in their current definition is questionable. In contrast to the genus *Furcifer*, where only a few new species were discovered in recent times (Glaw *et al.* 2009), the species inventory of *Calumma* seems to be far from complete: 10 new *Calumma* species were described in the last 15 years, and a further 4 were elevated to species status (Böhme 1997; Andreone *et al.* 2001; Raxworthy & Nussbaum 2006; Glaw & Vences 2007; Gehring, Pabijan *et al.* 2010).

The five currently recognised species of small chameleons with soft dermal appendages on their snout tip from Madagascar are referred to the *C. nasutum* group (*C. boettgeri*, *C. guibei*, *C. gallus*, *C. nasutum* and *C. fallax*). All of them are easy to identify as members of this group by morphology, and most of them can also be diagnosed to species with some confidence. The presence of occipital lobes allows the identification of *C. boettgeri* and *C. guibei*, as well as *C. linotum*, which sometimes is seen as a synonym of *C. boettgeri* and sometimes as valid species. Males of *C. gallus* are uniquely characterised by their elongated, usually spear-shaped and pointed rostral appendage. Distinguishing *C. nasutum* and *C. fallax* can be challenging (especially of the females), because these two species are poorly defined and assumed to occur sympatrically along lowland areas and mid-elevations at Madagascar's central east coast, together with *C. gallus*. On a first glance the different *C. nasutum* populations are morphologically rather conserved but a closer look reveals important morphological variation (e.g. absence or presence of dorsal crests, shape, length and colouration of rostral appendages) between and within several populations ascribed to this species,

already pointing to a complex taxonomic situation (Hillenius 1959; Brygoo 1971; Glaw & Vences 2007).

As a first step to resolving the complex taxonomic situation within this chameleon group, we here provide information on the type material of *C. nasutum* and *C. fallax,* and molecular evidence for an extraordinarily high genetic diversity within this group of chameleons, indicating a very complex situation with numerous candidate species and/or deep conspecific lineages.

During recent fieldwork in littoral forests at Madagascar's north-central east coast, we collected specimens of short-nosed chameleons of the genus *Calumma* that differed obviously from hitherto known populations of *C. nasutum* by differences in morphology of the rostral appendage in both sexes and subtle differences in live colouration. Subsequent molecular analysis revealed a high mitochondrial divergence from other populations and taxa of short-nosed chameleons and consistent differences in one nuclear gene.

As a second step in the present study we therefore conclude that this population indeed constitutes a distinct species which we describe as *Calumma vohibola* sp. nov. In the following we will already refer to this species under the new scientific name coined later.

Materials and Methods

Specimens were collected in the field at night at the end of the rainy season using torches and headlamps to detect roosting chameleons in the vegetation. Voucher specimens were anesthetised and killed by injection with chlorobutanol, strengthened with 90% ethanol and stored in 70% ethanol. Muscle tissue samples for future molecular analyses were taken from all specimens from the right hind limb and preserved in pure ethanol. Geographic coordinates were recorded with global positioning satellite receivers.

Specimens from the following collections were studied: Naturhistorisches Museum Wien, Austria (NMW); Senckenberg Naturmuseum, Frankfurt, Germany (SMF); Université d'Antananarivo, Département de Biologie Animale, Antananarivo, Madagascar (UADBA); Zoologisches Forschungsmuseum Alexander Koenig, Bonn, Germany (ZFMK); Zoologisches Museum Berlin, Germany (ZMB); Zoologische Staatssammlung München, Germany (ZSM).

Tissue samples of additional individuals were taken by tail clipping from freshly collected specimens and stored in pure ethanol for further genetic analyses at the Technical University of Braunschweig, Zoological Institute, Braunschweig, Germany. ZCMV, FGZC, PSG and FAZC refer to field numbers of M. Vences, F. Glaw, P.-S. Gehring, and F. Andreone.

Measurements were taken with digital callipers to the nearest 0.1 mm by P.-S. Gehring. Snout–vent length is abbreviated SVL, total length is abbreviated TL. Rostral appendage length (RAL) was measured in lateral view along the anterior surface, from the appendage base to the tip of the appendage. Terminology in description of crests and ornamentation characters and other external morphology follows previously published chameleon descriptions (Brygoo 1971, 1978; Andreone *et al.* 2001), except that the canthus rostralis is here referred to as the rostral crest.

Mitochondrial and nuclear DNA variation was studied in 10 (for mtDNA) and 9 (for nDNA) specimens of the new species, including the holotype, 2 paratypes and

7 additional individuals sampled in the forest fragments at Vohibola and Ankanin'ny Nofy (approx. 2.5 km west of the type locality). Additionally, we incorporated available tissue samples from other species of the *C. nasutum* species group. Tissue samples of *C. fallax* were collected by PSG, FG, Jörn Köhler and Jason Lee Brown at Tsinjoarivo (19°39.56′S, 47°44.06′E, 1568 m asl) in April 2010, samples of *C.* aff. *nasutum* and *C. gallus* were collected by PSG, FR, Emile Rajeriarison and François Randrianasolo in a forest fragment called 'Sahafina' (18°48′38.3′′S, 48°58′49.2′′E, 56 m) close to Ampasimanolotra in April 2009. Tissue samples of *C.* aff. *nasutum* from Tampolo (17°17′19.2′′S, 49°24′41.6′′E, 4 m) were collected by PSG, FR, Emile Rajeriarison and François Randrianasolo in April 2009. Tissue samples of *C. nasutum* were collected by FG and MV on 18 February 2003 at Andasibe (no precise coordinates, ca. 900 m asl). Tissue samples of *C.* cf. *gallus* were collected by David R. Vieites and Ignacio de la Riva in Ambohitsara, south-east Madagascar (21°21.431′S, 47°48.941′E, 294 m a.s.l.). The samples of *Furcifer lateralis* (FGMV 2002.65), used as an outgroup, were collected by MV at Antoetra in January 2003.

Two molecular datasets were used for analyses: (1) a fragment of the mitochondrial *NADH dehydrogenase subunit 2* (*ND2*) gene; and (2) a gene encoding the oocyte maturation factor (*C-mos*). We used the *C-mos* gene to validate the relationships and to confirm the lack of inter-lineage gene flow between *Calumma vohibola* sp. nov. and closely related species.

Total genomic DNA was extracted from the tissue samples using proteinase K digestion (10mg/ml concentration) followed by a salt extraction protocol (Bruford *et al.* 1992). We Polymerase Chain Reaction-amplified the fragment of the *ND2* gene using the primers ND2F17 (5′-TGACAAAAAATTGCNCC-3′) (Macey *et al.* 2000) and ALAR2 (5′-AAAATRTCTGRGTTGCATTCAG-3′) (Macey *et al.* 1997), and a fragment of the nuclear oocyte maturation factor (*C-mos*) using the primers CO8 (5′-GCTTGGTGTTCAATAGACTGG-3′) and CO9 (5′-TTTGGGAGCATCCAAA GTCTC-3′) (Han *et al.* 2004), following standard protocols. After purification (EXOSAP), the fragments were resolved on an automated DNA sequencer (ABI 3130 XL Applied Biosystems). For the *ND2* dataset only the light strand was sequenced, while the *C-mos* gene fragment was sequenced in both directions in order to verify possible heterozygote sites. Sequences were validated and aligned manually with the software CodonCode Aligner (CodonCode Corporation).

After alignment and exclusion of ambiguous positions at the beginning and at the end of sequences, the fragment consisted of 593 nucleotides (*ND2*) and 471 nucleotides (*C-mos*). The amplified *ND2* fragment incorporates a short fragment (nine base pairs) of the flanking Trp-tRNA. A specimen of *Furcifer lateralis* (FGMV 2002.65) from Antoetra was used as an additional outgroup in the phylogenetic analyses of the *ND2* dataset. We deposited the newly resolved DNA sequences in Genbank, where the *ND2* fragment was submitted without the nine base pairs of the flanking tRNA (accession numbers JN030454–JN030489; Table 1, see Supplementary Material online).

Unpartitioned Bayesian inference searches were performed for the *ND2* dataset only. Model selection was carried out in MrModelTest version 2.3 (Posada & Crandall 1998; Nylander 2004). The Akaike information criterion as implemented in this software selected GTR + I + G as the best fitting model of nucleotide substitution in the *ND2* dataset. We implemented this model in MrBayes version 3.1.2, and programmed two runs with four chains run for a total of five million generations, sampled every thousandth generation. The effective sample size (ESS)

values ranged for all parameters in both runs between minimum 3 753.625 and maximum 8 490.028; standard deviation of split frequencies (SDSF) values dropped down to 0.0047, indicating that five million generations were sufficient and convergence of the two runs has occurred. The two runs had converged onto a stationary distribution after 30 000 generations; however, we conservatively discarded the first 25% of the five million generations as burn-in, leaving 3750000 generations from which parameter values were summarised and a majority rule consensus tree was produced with posterior probabilities calculated as the frequency of samples recovering each clade (Huelsenbeck & Ronquist 2001). As a further measure of node support, we used the sequence data in a maximum parsimony analysis with unordered and equally weighted characters in PAUP* version 4.0b10 software (Swofford 2002). We specified a heuristic search with 100 random addition sequences and tree bisection reconnection (TBR) branch swapping. Support for the resulting topology was obtained by bootstrapping with 1000 replicates.

Corrected (LogDet) and uncorrected pair-wise distances (*p*-distances transformed into percentages) for the *ND2* dataset were calculated group-wise in MEGA4 software (Tamura *et al.* 2007).

Several heterozygous nucleotide sites were detected in the *C-mos* (471 base pairs) dataset, therefore we used the software Phase version 2.1.1 (Stephens *et al.* 2001; Stephens & Scheet 2005) to infer haplotypes. The haplotypes were used in a network analysis using statistical parsimony (Templeton *et al.* 1992), as implemented in the program TCS version 1.21 (Clement *et al.* 2000) with a connection limit of 95%. We do not present here a combined analysis of both genes as our aim was not to study the phylogeny of the species involved (for which more markers would be necessary) but to assess concordance in patterns of separation of haplotypes among the nuclear and mitochondrial gene to support species delimitation.

For as yet unidentified species we used the names as in Glaw & Vences (2007) which usually prefix with 'sp. aff.' the name of the morphologically closest described species and a descriptor that is either geographic or refers to a characteristic trait of the candidate species.

RESULTS

Identity of *Calumma nasutum* and *C. fallax*

The exact delineation of all the different known populations assigned to *C. nasutum* and *C. fallax* is a very complex endeavour given that *C. nasutum* as currently understood (Glaw & Vences 2007) certainly represents a complex of several species.

The type material of *C. nasutum* was examined by FR in the Muséum National d'Histoire Naturelle in Paris in October 2009 and again by PSG in January 2011. According to the original description (Duméril & Bibron 1836) as well as Klaver and Böhme (1997), *C. nasutum* was described on the basis of four syntypes (MNHN 6643 ♀, MNHN 6643A ♀, MNHN 6643B ♂, MNHN 6643C ♂; for measurements see Table 2), although Mocquard (1900b) argues that the actual number of syntypes is nine. After more than 170 years in alcohol the four studied syntypes are still in good condition, although the colouration has almost faded completely to a brown-grey with some enlarged bluish-grey scales all over the body (Fig. 1). All four specimens show distinct rostral ridges fusing on the anterior snout in a soft, dermal laterally

Table 2. Morphological measurements of the type specimens of Malagasy short-nosed chameleons without occipital lobes: *Calumma vohibola*, *C. nasutum*, *C. fallax* and *C. gallus*.

Collection no.	Species	Locality	Sex	Status	SVL	TaL	TL	EN	NST	LRA	TSRA	RcA	ESC	DC	TC	PC	SFL	AP
ZSM 645/2009	*C. vohibola*	Vohibola forest	M	HT	45.8	40.1 (cut)	45.8	2	2.9	0.5	33	+	+	7 spines	+	–	+	–
ZSM 644/2009	*C. vohibola*	Vohibola forest	M	PT	45.6	33.7 (cut)	79.3	2	2	0.8	21	+	+	–	+	–	+	–
ZSM 643/2009	*C. vohibola*	Vohibola forest	F	PT	44.8	38.2	83	1.5	1.8	0.0	17	+	+	–	+	–	+	–
FAZC 14607	*C. vohibola*	Ivoloina	F	PT	43.1	40.1 (cut)	83.2	1.7	2	0.3	17	+	+	–	+	–	+	–
ZFMK 46112	*C. vohibola*	Ivoloina	M	PT	49.8	40.7	90.5	1.8	1.9	0.1	10	+	+	8 spines	+	–	+	–
ZFMK 50550	*C. vohibola*	40 km S Tamatave	F	PT	43.0	33.3	76.3	1.7	2.1	0.2	17	+	+	–	+	–	+	–
ZFMK 48176	*C. vohibola*	Ivoloina	F	PT	42.6	43.6	86.2	2	2.3	0.7	24	+	+	–	+	–	+	–
ZFMK 48177	*C. vohibola*	Ivoloina	F	PT	37.1	42.1	79.2	1.6	1.6	0.6	24	+	+	–	+	–	+	–
ZMB 18999	*C. vohibola*	Ille aux Prunes	F	PT	41.9	40.3	82.2	1.5	2.0	0.3	21	+	+	–	+	–	+	–
SMF 16465	*C. vohibola*	Ille aux Prunes	F	PT	42.1	40.7	82.8	–	–	0.4	–	+	+	–	+	–	+	–
MNHN 6643	*C. nasutum*	Madagascar	F	ST	49.1	43.9	92.9	1.7	2.0	0.9	24	–	–	–	+	–	–	+
MNHN 6643A	*C. nasutum*	Madagascar	F	ST	44.2	39.4	83.6	1.6	2.2	1.1	25	–	–	–	+	–	–	+
MNHN 6643B	*C. nasutum*	Madagascar	M	ST	45.5	42.8	88.3	2.1	2.1	3.4 cut nose	*>30*	–	–	–	+	–	–	+
MNHN 6643C	*C. nasutum*	Madagascar	M	ST	47.8	47.6	95.4	–	–	3.6	–	–	–	–	+	–	–	+
MNHN 6643D	*C. nasutum*	Madagascar	F	*	46.6	38.1	84.7	0.7	1.1	1.6	–	–	–	–	+	–	–	+
MNHN 6643E	*C. nasutum*	Madagascar	J	*	25.8	22.6	48.4	1.8	1.6	0.9	20	–	–	–	+	–	–	+
MNHN 6643F	*C. nasutum*	Madagascar	M	*	46.0	42.4	88.4	2	2.7	3.4	>50	–	–	–	+	–	–	+
MNHN 1899.317	*C. fallax*	Foret de Ikongo	M	ST	41.5	48.9	90.4	2.0	2.7	1.7	25	–	+	12 cones	+	+	+	–
MNHN 1890.430	*C. fallax*	Foret de Ikongo	M	*	43.4	48.9	92.3	2.3	2.4	2.5	32	–	+	7 cones	+	+	+	–
MNHN 1888.24	*C. fallax*	Foret de Ikongo	F	*	42.2	43	85.24	1.8	2.4	2.5	48	–	+	–	+	+	+	–

Notes: M, male; F, female; J, juvenile; HT, holotype; PT, paratype; ST, syntype; *type status of specimen unclear; SVL, snout–vent length in mm; TaL, tail length in mm; TL, total length in mm; EN, distance between anterior border of eye socket and nostril in mm; NST, distance between nostril and snout tip in mm; LRA, length of rostral appendage in mm; TSRA, total number of scales on rostral appendage in lateral view; RcA, rostral in direct contact with appendage; ESC, distinctly enlarged scales on cheeks; DC, dorsal crest absent (−) or number of dorsal spines/cones; TC, temporal crest present (+) or absent (−); PC, parietal crest present (+) or absent (−); SFL, scales on forelimbs homogeneous (−) or heterogeneous (+); AP, axillary pits present (+) or absent (−).

Figure 1. Selected individuals of the type series of *Calumma nasutum* in lateral and dorsal view. From top to bottom: female syntype MNHN 6643; male syntype MNHN 6643C; note the rostral appendage that clearly projects the snout. (A) Detailed dorsal view of the head of the female syntype (MNHN 6643); note the kinked rostral appendage that clearly projects beyond the snout. (B) Detailed lateral view of the head of the female syntype (MNHN 6643). Note: Not to scale.

compressed, ellipsoid rostral appendage that clearly projects beyond the upper snout tip. In *C. nasutum* a sexual dimorphism seems to exist in the length of this appendage that varies in the type material (females 0.9–1.1 mm, males 3.4–3.6 mm). The upper labials form dorsally a serrated line, a character mentioned by Angel (1942) as "dents de scie". All four type specimens of *C. nasutum* have no occipital lobes and no traces of gular, ventral or dorsal crest.

Calumma fallax was described twice in two different papers. One of the descriptions (Mocquard 1900a) is very short, comprising just 10 lines, whereas the second (Mocquard 1900b) is much more detailed, including an illustration. According to Mocquard (1900a,b), the description of *C. fallax* was based on two adult syntypes (a male and a gravid female) from the Ikongo forest, collected by Guillaume Grandidier in 1898–1899. However, Mocquard (1900b) mentioned that the museum has four additional specimens (three males, one female), catalogued as *C. nasutum*, which should be assigned to *C. fallax* as well. This statement and

perhaps entries in the MNHN catalogue might have prompted Klaver and Böhme (1997) to list six syntypes of *C. fallax* (MNHN 99.317–318; 88.24; 90.430–432), although the numbers suggest that only MNHN 1899.317–318 might have been collected by G. Grandidier in 1898–1899 and can be unambiguously considered as syntypes. Brygoo (1971), probably aware of this problem and not fully convinced on the validity of *C. fallax*, obviously considered only two specimens as syntypes ("1 M. et 1 F. gravide, G. Grandidier, 1898–1899"), although he did not mention – in contrast to other species accounts – any type status or catalogue number.

Mocquard (1900a,b) described a small median parietal crest in both sexes and a distinct dorsal crest in the male, consisting of small enlarged rounded tubercles, which are not spine-like: "... petites tubercules ... arrondies sensiblement et non spiniformes" (Mocquard 1900a, p. 345). The rostral appendage in males is longer than in females, both specimens of *C. fallax* are without axillary pits and enlarged separated scales on the casque and the cheeks (Mocquard 1900a,b). In a summarising re-examination of the *C. fallax* type material, Brygoo (1971) provided additional morphological information to distinguish it from *C. nasutum*: TL up to 110 mm in males and 96 mm in females, length of rostral appendage 2.5–3.0 mm; 4–6 enlarged temporal scales; occipital lobes absent; dorsal crest composed of 8–10 small, rounded tubercles in males, absent in females.

In our molecular analyses we refer to an adult male specimen assigned to *C. fallax* (ZSM 286/2010) collected at Tsinjoarivo on 23 April 2010 characterised by the presence of a parietal crest, presence of dorsal crest consisting of small rounded tubercles, six enlarged scales on the temple, absent axillary pits and a distinctly elevated casque (Fig. 6C).

Our comparisons with the type material of *C. nasutum* and *C. fallax* allow us to conclude that neither of these names constitutes a senior synonym of *C. vohibola.* The two male *C. nasutum* syntypes have distinctly longer rostral appendages than all available *C. vohibola* males (3.4–3.6 mm vs. 0.1–0.8 mm; Table 2) and the two female syntypes of *C. nasutum* differ from all available females of *C. vohibola* by a longer rostral appendage (0.9–1.1 mm vs. 0.0–0.7 mm; Table 2), and by the presence of a serrated line of the upper labials (vs. absence). The unambiguous male syntype of *C. fallax* (MNHN 1899.317) differs from the male holotype of *C. vohibola* by a longer rostral appendage (1.7 mm vs. 0.4 mm), the presence of a parietal crest (vs. absence), a dorsal crest composed of flat and rounded tubercles (vs. upright spines), the presence of distinct enlarged temporal scales (vs. indistinct) and an elevated casque (vs. not elevated).

Our examination of the type material of *C. gallus*, the only other nominal chameleon species from Madagascar with soft rostral appendage and without occipital lobes, indicated that this species and its identity, too, are in need of revision. However, the holotype differed in various characters from *C. vohibola* (very long and pointed appendage in males [vs. very short], the presence of distinct axillary pits [vs. absence], the more homogeneous scalation on the head [vs. more heterogeneous]) and, therefore, both taxa are certainly not conspecific. A detailed analysis of the identity of the type specimens of *C. nasutum*, *C. fallax* and *C. gallus* and their attribution to chameleon populations will be provided elsewhere. A preliminary identification key to the currently recognized species of Malagasy short-nosed chameleons is provided in Table 3. In the following, we will analyse the molecular and morphological differentiation of *C. vohibola* in comparison with the other

species of the *C. nastum* group, in order to provide evidence for its identity as independent evolutionary lineage, and thus as support for its species status.

Molecular Differentiation and Phylogenetic Relationships

The *ND2* alignment consisted of 19 sequences with 593 aligned nucleotide positions. A total of 263 nucleotides were variable, 170 of which were parsimony informative. The heuristic search in the maximum parsimony analysis produced three most parsimonious trees, all with a length of 445 steps (consistency index [CI] = 0.7326; retention index [RI] = 0.8065). The three obtained trees differed exclusively in the placement of *C. vohibola* individuals within the *C. vohibola* clade.

In all performed analyses, the basal phylogenetic relationships between *C. vohibola* and the other taxa of the *C. nasutum* group remained unresolved. Clearly, further genetic analyses including several more markers will be necessary to resolve the relationships within this group and the entire genus *Calumma*. However, the main purpose of this analysis was not reliably to establish relationships within this group but to give an indication of the molecular differentiation between taxa.

In the *ND2* gene dataset four clades were highly supported by the discrete character- and model-based phylogenetic analyses (Fig. 2). The haplotypes of *C. vohibola* showed a high divergence to all included taxa of the *C. nasutum* species group and formed a single distinct clade grouping all specimens assigned to *C. vohibola*. In the *ND2* dataset we detected marginal sequence variation within *C. vohibola* from no nucleotide substitutions up to 0.07% (uncorrected *p*-distance). The samples of the different *C. nasutum* populations formed well-supported clades for the most part, and the lowland populations from Sahafina and Tampolo formed a highly supported sister clade to *C. gallus* and *C. fallax*. The phylogenetic relationship of *C. fallax* from Tsinjoarivo remains unresolved in our tree and was placed in a basal position but without any statistical support. Assuming that the *C. nasutum* population from the mid-elevation locality Andasibe represents *C. nasutum sensu stricto* (see previous sub-section on *C. nasutum*), these two lowland *C.* sp. aff. *nasutum* populations represent two new unconfirmed candidate species (UCS), following the scheme suggested by Vieites *et al.* (2009).

The phylogenetic analysis of *C.* cf. *gallus* from Ambohitsara placed it with highest statistical support as sister taxon of *C. gallus,* indicating at least a new deep conspecific lineage in this species.

Groupwise uncorrected *p*-distances in the *ND2* fragment amount to 14.7% between *C. vohibola* and *C. nasutum* from Andasibe, 15.4% between *C. vohibola* and the *C.* sp. aff. *nasutum* population from Sahafina, 17.7% to the *C.* sp. aff. *nasutum* population from Tampolo, 14.8% to *C. fallax* from Tsinjoarivo, 17.7% to *C.* cf. *gallus* from Ambohitsara and finally 19.4% to *C. gallus* from Sahafina. The uncorrected *p*-distances between all *Calumma* species and the outgroup (*Furcifer lateralis*) reached from 22.3 to 26.1% (additional uncorrected and corrected *p*-distances are given in Table 4). As a comparison, *ND2* sequence divergences between species of south African dwarf chameleons (*Bradypodion*) ranged between around 2 and 16.5% (Tolley *et al.* 2004).

The LogDet-distance was designed to deal with unequal base frequencies in each pairwise sequence comparison – thus it allows base compositions to vary over the

Table 3. Key to males of the *Calumma nastum* group and the morphologically similar *C. vatosoa*.

Feature	Species/ instruction
1(a) Occipital lobes present	Go to 2
1(b) Occipital lobes absent	Go to 3
2(a) Occipital lobes in contact medially, but largely separated behind the occiput; tip of rostral appendage not blue coloured; only known from higher elevations of the Tsaratanana massif in central-northern Madagascar	*C. guibei*
2(b) Occipital lobes not notched medially; tip of rostral appendage sometimes blue coloured; low and mid-elevations of northern Madagascar	*C. boettgeri*
3(a) Axillary pits present; scalation on head and forelimbs rather homogeneous; dorsal crest always absent.	Go to 4
3(b) Axillary pits absent; scalation on head and forelimbs rather heterogeneous; dorsal crest present or absent	Go to 5
4(a) No trace of rostral appendage; a yellow to orange spot laterally at mid-body; north-eastern Madagascar (Anjanaharibe-Sud, Masoala)	*C. vatosoa*
4(b) Very long (5–11 mm), pointed, distinctively coloured rostral appendage; often two blue spots laterally at mid-body; central-eastern Madagascar	*C. gallus*
5(a) Rostral appendage indistinct, very short or nearly absent, if dorsal crest is present it consists of separated, upright, spiniform scales; direct contact of rostral and scales of rostral appendage; lowland and littoral forests of central-eastern Madagascar	*C. vohibola*
5(b) Rostral appendage distinct, moderately long (2–3 mm) and rounded; number and shape of cones or spines in dorsal crest variable; low and mid-elevation of eastern Madagascar	Go to 6
6(a) Parietal crest distinct; casque distinctly raised posteriorly; dorsal crest consisting of separated, rounded, cone-like scales; distinctively enlarged scales all over the body; mid-altitude and highland areas of south-central and central-eastern Madagascar	*C. fallax*
6(b) Parietal crest absent; casque not distinctly raised posteriorly; dorsal crest sometimes absent but usually present, consisting of separated cone- to spine-like scales; mid-altitude and lowland areas of eastern Madagascar	*C. nasutum* complex

tree and assumes that all sites can vary (Gu & Li 1996; Lockhart *et al.* 1994). The LogDet-distance in the present study is used to give a model-based estimate of nucleotide substitution within this group of chameleons. Groupwise corrected LogDet-distances in the *ND2* fragment amount to 19.2% between *C. vohibola* and *C. nasutum* from Andasibe, 20% between *C. vohibola* and the *C.* sp. aff. *nasutum* population from Sahafina, 24.1% to the *C.* sp. aff. *nasutum* population from Tampolo, 19.3% to *C. fallax* from Tsinjoarivo, 25.6% to *C.* cf. *gallus* from Ambohitsara and finally 22.3% to *C. gallus* from Sahafina.

Our analyses of variation in *C-mos* nuclear DNA sequences gave quite similar results to our estimate of relationships derived from mtDNA (Fig. 2). Out of the 471 base pairs analysed in the *C-mos* gene, 30 nucleotides were variable, of which 24 were

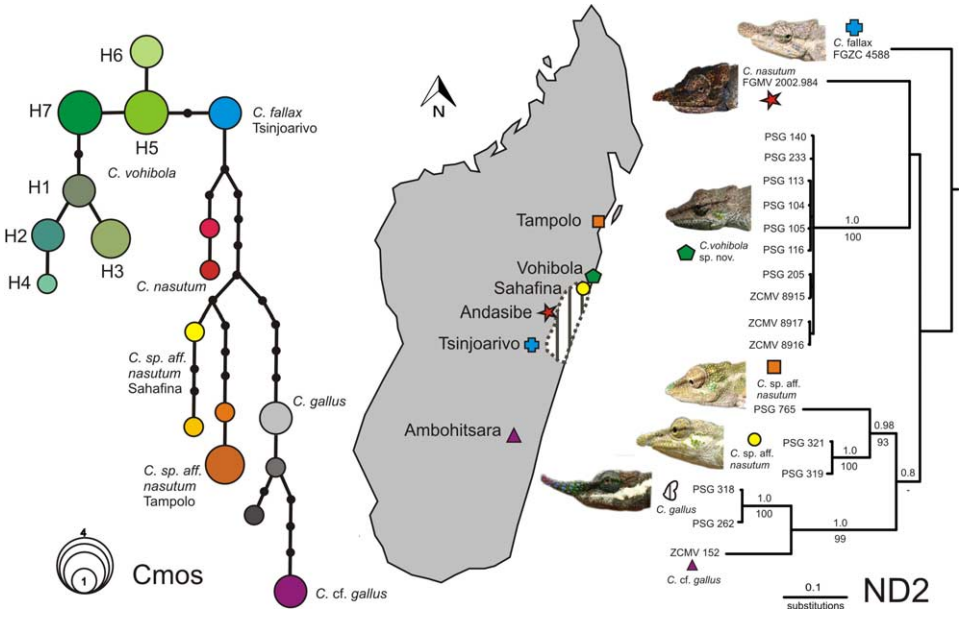

Figure 2. Results of a phylogenetic analysis of the mitochondrial *ND2* gene (to the right), and haplotype network of the nuclear *C-mos* gene (to the left), with a map showing collection localities of *Calumma* samples in Madagascar.
Notes: In the tree, values above branches are Bayesian posterior probabilities; maximum parsimony bootstrap proportions above 80% are given below corresponding branch; a sample of *Furcifer lateralis* was used as an outgroup (not shown); samples are given with their field numbers (see 'Materials and Methods' for abbreviations).

parsimony informative. Heterozygous sites in the *C-mos* dataset were detected in several individuals. In *C. vohibola*, we encountered three homozygous individuals, one individual with two heterozygous sites and four individuals with one heterozygous site. In *C. nasutum*, we detected only a single heterozygous site. The sequence of *C.* aff. *nasutum* from Sahafina showed three heterozygous sites, while one specimen of *C.* aff. *nasutum* from Tampolo was homozygous and the other was heterozygous at a single site. The individual of *C. fallax* from Tsinjoarivo was a homozygote, as well the *C.* cf. *gallus* from Ambohitsara. In *C. gallus* from Sahafina both individuals were heterozygous; one at three sites, the other at a single site. However, the haplotypes formed well-defined clusters separated from each other by a minimum of two mutational steps and without haplotype sharing between species or candidate species. *C. fallax* from Tsinjoarivo was separated by only 2 mutational steps from *C. vohibola*, whereas a maximum of 17 mutational steps separate *C. vohibola* and *C.* cf. *gallus* from Ambohitsara. Within the nine analysed individuals of *C. vohibola* seven haplotypes could be distinguished (Fig. 2); Haplotypes H1–H7 are separated by a maximum of six mutational steps (H4–H6).

Although the nuclear dataset refers to only a limited number of specimens, the fact that there is no haplotype-sharing between the several forms of short-nosed chameleons suggests that probably all of the studied taxa represent independent evolutionary lineages.

Table 4. Mean uncorrected *p*-distances (below the diagonal in plain font) and corrected *LogDet*-distances (above the diagonal, in italics) for the grouped *ND2* dataset.

	Cv	CnA	CaffnS	CaffnT	Cf	Cg	Ccfg
Cv		*19.2%*	*20%*	*24.1%*	*19.3%*	*25.6%*	*22.3%*
CnA	14.7%		*15.9%*	*19.8%*	*18.5%*	*24.4%*	*20.7%*
CaffnS	15.4%	13%		*13.7%*	*18.8%*	*21.8%*	*21.2%*
CaffnT	17.7%	14.8%	10.5%		*19.2%*	*25.7%*	*22.2%*
Cf	14.8%	14%	14.2%	14%		*23.2%*	*20.4%*
Cg	19.4%	18.8%	16.9%	19.3%	17.6%		*15.8%*
Ccfg	17.7%	17.2%	16.5%	17.7%	16.9%	11.4%	

Notes: Cv, *Calumma vohibola*; CnA, *Calumma nasutum* Andasibe; CaffnS, *Calumma* sp. aff. *nasutum* Sahafina; CaffnT, *Calumma* sp. aff. *nasutum* Tampolo; Cf, *Calumma fallax* Tsinjoarivo; Cg, *Calumma gallus* Sahafina; Ccfg, *Calumma* cf. *gallus* Ambohitsara.

Given the constant differences in morphology to the morphologically similar taxa *C. nasutum*, *C. fallax*, and *C. gallus* which fully correlate with high mitochondrial divergence, and with distinct haplotypes in one nuclear gene, we conclude that *C. vohibola* constitutes a separate and independent evolutionary lineage. Therefore, it should best be considered as a distinct species. In the following sub-sections we thus describe *C. vohibola* as a new species.

Calumma vohibola sp. nov. (Figs. 3 & 4)

Holotype.—ZSM 645/2009 (ZCMV 8915), adult male with incompletely everted hemipenes, collected in the littoral forest 'Vohibola' near the village Andranokoditra (18°35′22,9″S, 49°13′50,6″E, 9 m asl), Toamasina Province, Région Antsinanana, central eastern Madagascar, on 13 April 2009 by P.-S. Gehring, F.M. Ratsoavina, B. Mathevon, E. Rajeriarison & F. Randrianasolo.

Paratypes.—ZSM 644/2009 (ZCMV 8916), adult male, ZSM 643/2009 (ZCMV 8917), adult female, UADBA uncatalogued (ZCMV 8918) adult female, all three with same collection data as holotype; UADBA uncatalogued (ZCMV 8903) collected at Ankanin'ny Nofy (18°36′20,9″S, 49°12′49,8″E), Toamasina Province, Région Antsinanana, on 8 April 2009 by P.-S. Gehring, F.M. Ratsoavina, E. Rajeriarison & F. Randrianasolo; SMF 16465 and ZMB 18999, two adult females, collected at Ile aux Prunes, Toamasina Province, Région Antsinanana, by A. Voeltzkow on 21–23 October 1904; ZFMK 46112, adult male, with everted hemipenes, collected at Ivoloina, Toamasina Province, Région Antsinanana, by W. Schmidt in 1985; ZFMK 50550, adult female, collected 40 km south of Toamasina, 'Akininofi' (Ankanin'ny Nofy), Toamasina Province, Région Antsinanana, by F. Henkel, W. Schmidt & V. Müller in May 1989; ZSM 805/2010 (FAZC 14607), adult female, collected at Ivoloina, Toamasina Province, Région Antsinanana, on 16 October 2010 by A. Crottini.

Diagnosis.—A small-sized brown to bluish-grey chameleon (SVL 43.0–49.8 mm, TL 76.4–92.2 mm) that is characterised by a very short rostral appendage, a very low casque, the absence of axillary pits, presence or absence of a dorsal crest in males and a distinct stress colouration in the female (Fig. 3D), consisting of a dark reddish

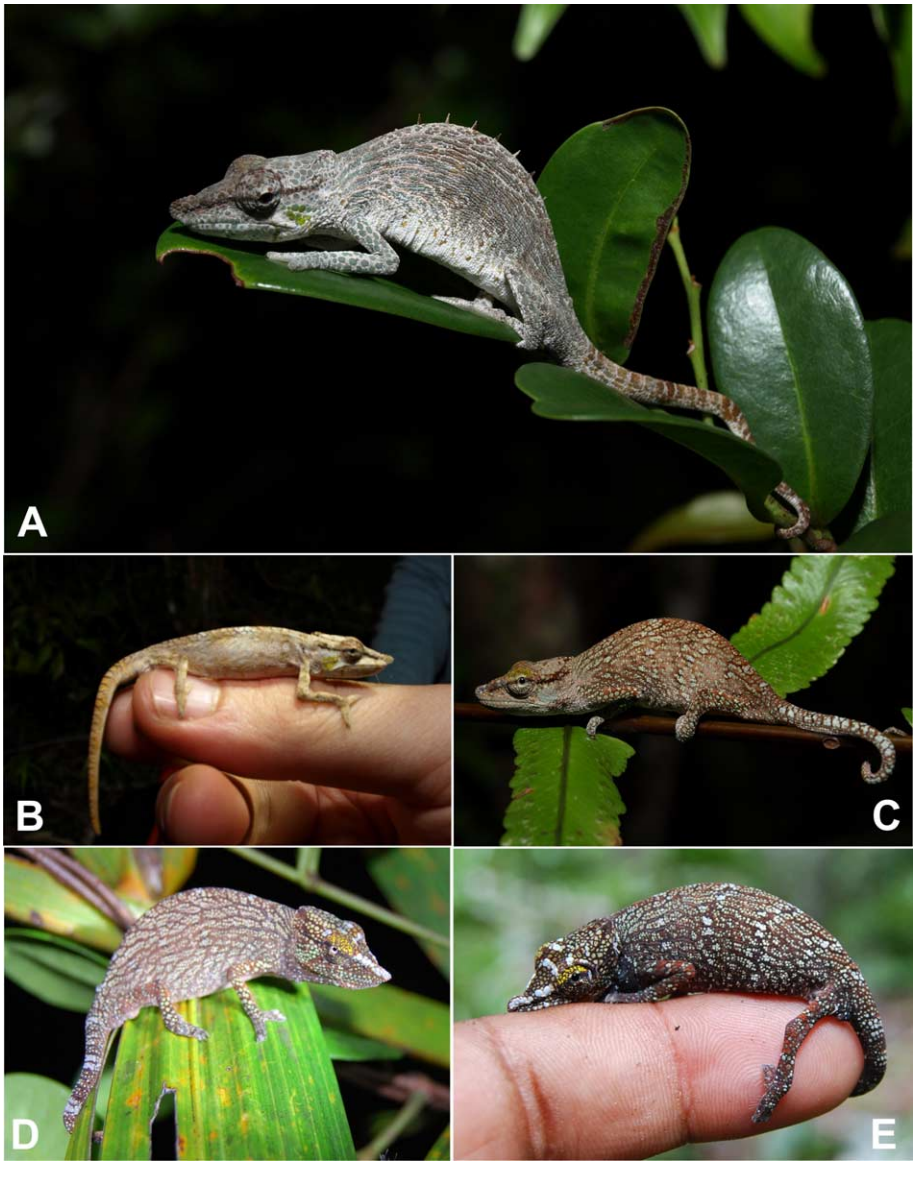

Figure 3. (A) Lateral view of male holotype of *Calumma vohibola* sp. nov. (ZSM 645/2009) in life when unstressed. (B) Lateral view of male paratype (ZSM 644/2009) in life when unstressed. (C) Lateral view of female paratype (ZSM 643/2009) in life when unstressed. (D–E) Female of *C. vohibola*, stress colouration fully developed.
Note: Photographs were taken in April 2009 at the type locality in the Vohibola forest.

ground colouration with many irregular light blue spots. *Calumma vohibola* is a member of the *C. nasutum* group based on its small size, low casque, absence of gular and ventral crest, and presence of a soft, dermal unpaired rostral appendage. This new species differs from all the other species in this group by the very short rostral appendage ($<$ 1 mm length in males, almost completely absent in females), which is $>$ 1 mm (usually 2–3 mm) in males of all other species.

Figure 4. Part of the type series of *Calumma vohibola* sp. nov. in lateral view. Left side from top to bottom: male holotype ZSM 645/2009; female paratype ZSM 643/2009; male paratype ZSM 644/2009; right side from top to bottom: female paratype ZFMK 50550; male paratype ZFMK 46112; female paratype SMF 16465; note the short or almost absent rostral appendages in all specimens and the dorsal crest in the male, consisting of seven single upright spines.
Note: Scale bar = 10 mm.

Besides this unique combination of characters, *C. vohibola* differs from *C. boettgeri* (including the holotype ZSM 21/1923 of *C. linotum* as a synonym) and *C. guibei* by the absence of occipital lobes (vs. distinctly recognisable); from *C. gallus* by the very short and rounded rostral appendage in males (vs. a very long and pointed appendage in males), the absence of distinct axillary pits (vs. presence), the more heterogeneous scalation on the head (vs. more homogeneous); from *C. fallax* by the absence of a distinctly posteriorly raised casque (vs. presence), the absence of a parietal crest (vs. presence), the presence of upright spine like dorsal tubercles (vs. rounded tubercles) and the absence of clearly enlarged scales on the temple and casque (vs. presence). *Calumma vohibola* is most similar to *C. nasutum* which, however, has a longer rostral appendage which is also evident in the four syntypes MNHN 6643 and 6643 a, b, c of *C. nasutum* and the holotype SMF 22132 of *C. radamanus*, a junior synonym of *C. nasutum*. In addition, *C. vohibola* differs from *C. nasutum*, *C. gallus* and *C. fallax,* and, as far as known, from all other species of *Calumma,* by a substantial genetic differentiation (see section on molecular differentiation below). *Calumma vohibola* also shows distinct similarities to *C. vatosoa* (which has been hitherto assigned to the *Calumma furcifer* species group) but differs from this species by presence of a short rostral appendage (vs. complete absence), absence of axillary pits (vs. presence) and absence of greenish ground colour (vs. presence).

Description of the holotype.—Adult male, in good state of preservation, but with a slit in the inner side of the right hind limb, where muscle tissue for genetic analyses was removed; left hemipenis completely, and right hemipenis incompletely, everted;

tongue outside of mouth cavity; SVL 45.8 mm; tail length 40.2 mm; further morphological measurements are provided in Table 5. Six slightly enlarged scales on the cheek with a darker pigmentation than the surrounding scales on the head. Distinct rostral ridges that fuse on the anterior snout in a very short soft, laterally compressed dermal rostral appendage that projects from the upper snout tip beyond 0.4 mm and has a round shape distally; parietal crest indistinct; supra-orbital crest rounded in lateral view and formed by single, rather smooth row of tubercles; lateral crest poorly developed and pointing straight posteriorly, fusing at the highest point of the casque. Temporal crest distinct, starting at the most posterior edge of the socket, curving upwards to the highest point of the casque. No traces of occipital lobes and no traces of gular and ventral crest. Dorsal crest present, starting 9.1 mm behind the casque and continuing until 11.2 mm above the cloaca; consisting of a row of seven separated upright spines, each with a length of 0.7 mm, and two distinct enlarged tubercles one at the beginning and one at the end of the crest. The single spines are separated by 1.8 mm continuously. Body laterally compressed with fine homogeneous scalation with the exception of slightly more heterogeneous larger scales on the head and on the legs, tail with heterogeneous scalation; axillary pits absent; tail without dorsal crest, feet without tarsal spines; tail base moderately

Table 5. Morphological measurements (in mm) of the holotype and two paratypes of *Calumma vohibola* sp. nov. Definition and selection of characters partly according to Andreone *et al.* (2001).

Character	ZSM 645/2009	ZSM 644/2009	ZSM 643/2009
Status	HT	PT	PT
Sex	Male	Male	Female
SVL	45.8	45.6	44.2
TaL	40.2	33.7 (cut)	38.2
TL	86	79.3	82.4
HL	13.1	11.6	11.9
HW	5.9	5.9	6.3
HD	7.4	7.6	8.2
App.–Eye	2.7	3	3
ESC	6	7	7
RA	0.4	0.5	0.1
DC (*n*)	7	–	–
DC–Head	9	–	–
DC–Tail	11.2	–	–
TC	+	+	+
HC	1.1	0.6	0.6
PC	5.5	5.1	4.2
SD	3.7	3.5	3.1
AGD	26.3	25.3	26.2

Notes: HT, holotype; PT, paratype; SVL, snout–vent length; TaL, tail length; TL, total length; HL, head length; HW, head width; HD, head depth; App.–Eye, distance between rostral appendage and eye socket; ESC, number of enlarged scales on the cheek; RA, length of rostral appendage (from snout tip to the tip of the appendage); DC, dorsal crest presence (−, absent) and number of spines; DC–Head, distance from head to first spine of the dorsal crest; DC–Tail, distance from last spine of the dorsal crest to the tail base; TC, temporal crest (+, present); HC, height of casque; PC, parietal crest; SD, socket diameter (measured horizontally); AGD, axilla–groin distance.

swollen. The hemipenis morphology will be described and analysed in a forthcoming comprehensive revision of genital morphology in the *C. nasutum* group.

Colouration in life. In a relaxed state the male holotype (Fig. 3A, photographed during the night) was characterised by a rather light bluish-grey colouration shading into a whitish colouration in ventral direction, with a network of darker horizontal stripes and irregular little brown spots all over the flanks, head and on the tail where they become more prominent posteriorly. Spines of dorsal crest uniformly brown. Turquoise blue enlarged scales on head, forelimbs, hind limbs, and tail, six yellow to green enlarged temporal scales. A dark horizontal line running from the tip of the snout, through the eyelid and extending horizontally until the posterior edge of the casque. Ventrally uniformly white. In preservative the colouration has almost disappeared, changing into brownish-blue (Fig. 4). Dorsum dark brownish with single blue spots that are arranged in horizontal bands, generally shading into lighter tones in ventral direction. Head almost completely dark-brownish with distinct single dark blue scales. Belly uniformly white. Tail almost completely brown with many single dark blue enlarged scales, ventrally with a white line.

Description of paratypes.—For measurements of available type specimens see Table 2. The female paratype ZSM 643/2009 is adult, in good state of preservation but with a slit in the inner side of the left hind limb, where muscle tissue for genetic analyses was removed. Head with almost absent rostral appendage that projects beyond the upper snout tip less than 0.2 mm. Parietal crest indistinct; supra-orbital crest rounded in lateral view and formed by single, rather smooth row of tubercles; lateral crest poorly developed pointing straight posteriorly and fusing at the highest point of the casque. On top of the casque, two slightly pronounced ridges form a V-like pattern, fusing posteriorly at the highest point of the casque. Temporal crest distinct, starting at the most posterior edge of the socket, curving upwards to the highest point of the casque; no traces of occipital lobes; no traces of gular and ventral crest. Rostral ridges fuse on the anterior snout in a very short rostral appendage. Upper labials form dorsally a straight line. Dorsal crest absent, body laterally compressed with fine homogeneous scalation, except on the head the scales are slightly larger; limbs and tail with heterogeneous scalation, tail without dorsal crest, feet without tarsal spines; tail base not swollen.

The adult male paratype ZSM 644/2009 is in good state of preservation but with a slit in the inner side of the right hind limb where muscle tissue for genetic analyses was removed; hemipenes fully everted. Distinct rostral ridges, that fuse on the anterior snout in a very short soft, dermal laterally compressed rostral appendage that projects the upper snout tip beyond 0.6 mm and has a round shape; parietal crest indistinct; supra-orbital crest rounded in lateral view and formed by single, rather smooth row of tubercles; lateral crest poorly developed and pointing straight posteriorly and fusing at the highest point of the casque, temporal crest distinct, starting at the most posterior edge of the socket, curving upwards to the highest point of the casque; no traces of occipital lobes; no traces of gular and ventral crest. Dorsal crest absent. Body laterally compressed with fine homogeneous scalation with the exception of slightly more heterogeneous larger scales on the head and on the legs, tail with heterogeneous scalation; axillary pits absent; tail without dorsal crest, feet without tarsal spines; tail base moderately swollen.

The adult male ZFMK 46112 is in rather good condition and has everted hemipenes (Fig. 4). Distinct rostral ridges that fuse on the anterior snout in a very short soft, dermal laterally compressed rostral appendage almost not projecting beyond the upper snout tip (less than 0.1 mm); parietal crest indistinct; supra-orbital crest rounded in lateral view and formed by single, rather smooth row of tubercles; lateral crest poorly developed and pointing straight posteriorly, fusing at the highest point of the casque; temporal crest distinct, starting at the most posterior edge of the eye socket, curving upwards to the highest point of the casque; no traces of occipital lobes; no traces of gular and ventral crests. Dorsal crest present, starting 4.6 mm behind the casque and reaching until 14.7 mm above the cloaca; consisting of a row of six separated upright spines, each with a length of 0.6 mm, and one distinct enlarged tubercle at the beginning and at the end of the crest. Axillary pits absent. Scalation on the extremities, the head, and cheeks rather heterogeneous with distinct enlarged bluish-grey scales. After 25 years in alcohol colouration has almost faded completely to brown grey with some enlarged bluish grey scales all over the body (Fig. 4).

The adult female ZFMK 50550 was collected at Ankanin'ny Nofy, only 2.5 km west of the type locality of *C. vohibola*, and is in rather good condition. It has no occipital lobes; no traces of gular, ventral or dorsal crest; parietal crest indistinct; supra-orbital crest rounded following the eye-socket in lateral view, formed by single, rather smooth row of tubercles; lateral crest poorly developed and pointing straight posteriorly and fusing at the highest point of the casque, temporal crest distinct, starting at the most posterior edge of the socket, curving upwards to the highest point of the casque. Distinct rostral ridges that fuse on the anterior snout in a very short soft, dermal laterally compressed rostral appendage, projecting beyond the upper snout tip for less than 0.1 mm. Axillary pits absent. After more than 20 years in preservative the colouration has almost faded to a rather uniformly dark brown with distinct enlarged dark blue scales all over the body, especially on the extremities and on the head and cheeks (Fig. 4).

Two adult females (ZMB 18999 and SMF 16465) of short-nosed chameleons from the Ile aux Prunes were identified by Boettger (1913) as females of *C. gallus* (Fig. 4). Based on the description of Boettger (1913) and after the re-examination of the two specimens, we assign them to *C. vohibola*, owing to the almost absent rostral appendages; the lack of gular, ventral or dorsal crest; absence of axillary pits, and a very low casque. The scalation on the head and especially on the cheeks is clearly heterogeneous in both specimens with single enlarged scales, which is in contrast to the rather homogeneous scalation in both sexes of *C. gallus*. After more than 100 years of preservation, the colouration faded almost completely into whitish-grey with single dark blue spots, although both specimens are in rather good condition. Interestingly, Boettger (1913) described the colouration of these two females as reddish-brown reticulated on whitish-yellow ground with vein-like stripes, based on the original field notes of Voeltzkow. This description fits astonishingly well with the observed stress colouration of a female *C. vohibola*, photographed at the type locality in April 2009 (Fig. 3D).

Colouration in life.—Males in relaxed state were rather uniform in life colouration (Fig. 3). Depending on temper, the colouration can brighten up and white, beige and brownish colour patterns become prominent, with several irregular light blue spots

all over the dorsum. The contrast to the dark horizontal line becomes more conspicuous and a vein-like pattern of dark lines occurs dorsally and extends on the dorsal parts of the tail. The enlarged temporal scales form a yellow triangle.

The colouration of the females can vary from beige to a reddish-brown ground colouration without any obvious colour patterns in a relaxed state to a conspicuous colouration consisting of an irregular pattern of light blue and red spots, when stressed (Fig. 3D; photographed during the day). In a stressed temper the upper half of the eyelids is brightly yellow coloured, a pattern that is not so obvious in relaxed animals, and six parallel arranged bright spots occur on the supra-orbital and rostral crests and another one on the tip of the snout. These spots can fuse to a broad white line forming a triangle between the eyes and the snout tip. A dark horizontal line through the eyelid is also present in females.

Etymology.—The species was discovered in the littoral forest of Vohibola, close to the village of Andranokoditra, a protected area approximately 60 km south of Toamasina, which is managed by the non-government organisation 'Man and the Environment' (MATE). Littoral forests are one of Madagascar's most endangered forest types and, therefore, we dedicate this new species to this protected area and in recognition for the work that has been done especially by Barbara Mathevon from MATE for the protection of this forest, and with the hope that it will help to protect this highly threatened habitat. The specific epithet is used as an invariable noun in apposition.

Natural history and ecology.—The protected littoral forest area 'Vohibola' is located on a narrow stretch of land between the Indian Ocean and the Canal des Pangalanes (Fig. 5). Within the protected area different habitat types exist: primary littoral forest; secondary forest; swamp; alluvial forest; cultivated; and strongly degraded area (Gehring, Ratsoavina *et al.* 2010). During our survey in Vohibola and in Ankanin'ny Nofy we detected *C. vohibola* within primary littoral forest and secondary forests. One of the predominant habitat types in Vohibola, large areas of secondary vegetation consisting of ericoid heathland (*Philippia* sp.) on sandy soils yielded no specimen of *C. vohibola*. All individuals were found in mid-April. At that time they were frequently found roosting in the vegetation at night or foraging at daytime, c. 0.5–4.0 m above the ground. Small and probably recently hatched juveniles were much more common than adults. No gravid females were found or identified as such during our survey. Adult females were much more abundant than fully grown males.

Distribution and conservation.—So far, *C. vohibola* is known only from localities within a 60 km-long stretch along the coastal area between Ivoloina and the small island Ille aux Prunes in the north and the Vohibola forest in the south (Fig. 5). Reports of *C. nasutum* in a survey of the Vohibola forest by Randrianirina (2005) almost certainly refer to *C. vohibola*.

Owing to intensive deforestation, the littoral forests of Madagascar today are persisting only as small fragments and they are no more than a few kilometres and never more than 10 km in width (de Gouvenain & Silander 2003; Ganzhorn *et al.* 2000). Forest remnants such as Vohibola still harbour a high species richness, especially of plants, with several genera endemic to this habitat (Bollen & Donati 2005; de Gouvenain & Silander 2003). However, it seems that some of the amphibian

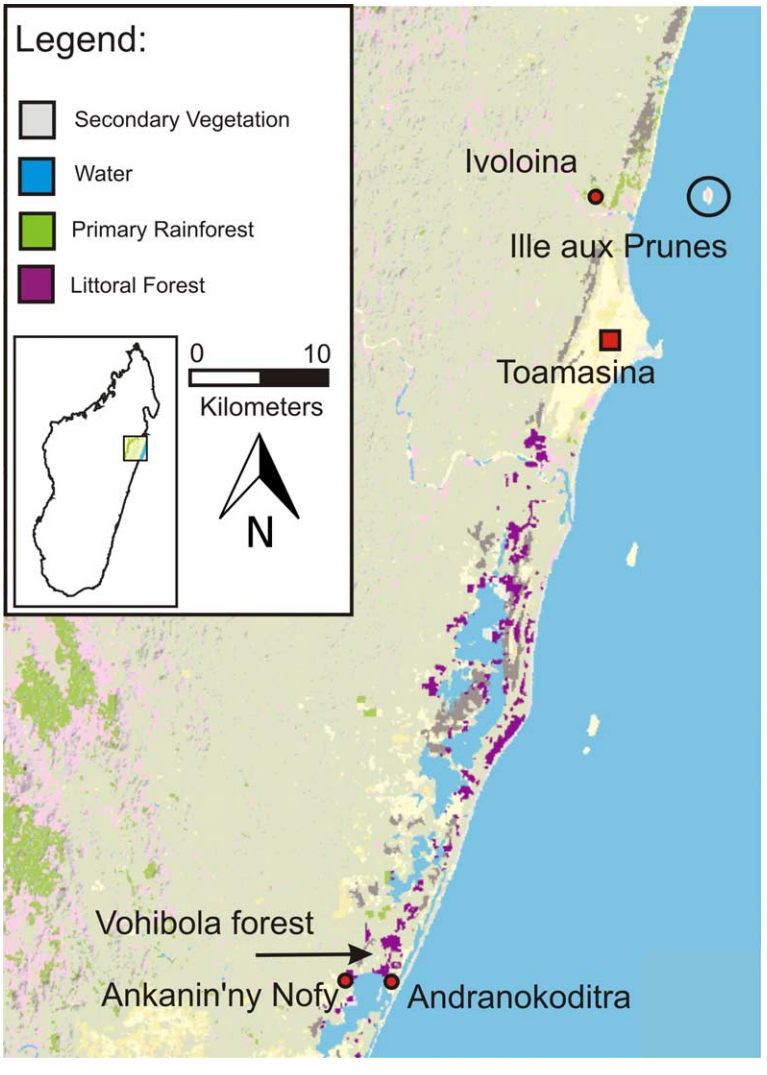

Figure 5. Map of the known localities and potential distribution areas of *Calumma vohibola* in central-eastern Madagascar.

and reptile species occurring in this area are somewhat tolerant to forest disturbance (e.g., species of *Blommersia*, *Mantidactylus*, *Phelsuma*, and probably also *Calumma vohibola*), and their conservation will, therefore, be feasible even in these relatively small reserves with partly degraded buffer zones (Gehring *et al.* 2010). Nevertheless, further studies are necessary to elaborate suitable management practices in more detail. Owing to its status as protected area, the Vohibola forest may be a stronghold for the species, especially in the case of continued habitat degradation in other littoral forests of this region. A full assessment of the conservation status of this species according to International Union for Conservation of Nature (IUCN) criteria (IUCN 2001) will be provided in the ongoing process of the 'global reptile assessment'.

Available names.—One junior synonym is currently recognised for *C. nasutum* and needs to be discussed as earlier available name for *C. vohibola*.

Chamaeleon radamanus Mertens, 1933 (synonymised with *C. nasutum* by Angel [1942]), was based on a male holotype (SMF 22132) and 16 adult plus 2 sub-adult paratypes, all from the Col Pierre Radama in north-eastern Madagascar, Maroantsetra Province, 1000 m asl. Several of the paratypes have been exchanged with other museums and we were able to trace the following five only: SMF 26394 (female), NMW 15999:1 (male with everted hemipenes), NMW 15999:2 (female) and NMW 15999:3 (female). According to the website Histoire de Maroantsetra (http://www.maroantsetra.com/pages/histoire.html), Col Pierre Radama is synonymous with the locality Ambatoledama which, according to the Gazetteer to Malagasy Botanical Collecting localities (http://www.mobot.org/mobot/gazetteer/results.asp), is situated at –15.28333333 (Lat_dec_deg), 50.00.00 (Long_min) and might be also identical with Ambaton'Radama or Col Radama, 547 m, c. 35–40 km north-east of Maroantsetra (Viette 1991).

After re-examination of the holotype by PSG and according to the original description the holotype has a SVL of 44 mm, tail length of 44 mm, without axillary mite pits, and enlarged scales on the extremities, the head and temples; the casque is indistinct concave and without any crests on the head; dorsal, gular and ventral crests are absent. The rostral appendage is in both sexes conspicuously developed, slightly denticulated and has a length between 2.2 and 3.0 mm in the males and 2.0 mm in the females. Mertens (1933) described that the rostral appendages are also fully developed in juveniles and not shorter than in the adults. This character and the absence of a dorsal crest in males differ substantially from *C. vohibola.*

The status of *C. radamanus* was also discussed by Brygoo (1971), who confirmed the synonymy with *C. nasutum*. After a comprehensive re-examination of the male holotype (SMF 22132) and the female paratype (SMF 26394) by PSG in September 2009, we agree with this interpretation, although this taxonomic status is tentative owing to the fact that *C. nasutum* most probably represents a complex of several distinct species.

Discussion

The present study highlights an extraordinarily high genetic diversity within a morphologically conservative chameleon group. The molecular data clearly separate species (*C. vohibola*, *C. nasutum*, *C. gallus* and *C. fallax*) and unconfirmed candidate species such as *C.* sp. aff. *nasutum* Tampolo, *C.* sp. aff. *nasutum* Sahafina (Fig. 6) and *C.* sp. aff. *gallus* Ambohitsara. The analysis of DNA sequences of the *ND2* gene confirmed that the *C. nasutum* species as currently defined consists of clearly distinct mitochondrial lineages and provides evidence that mitochondrial haplotype-sharing (and thus recent mitochondrial gene flow) between these species is very rare, and probably absent. The data further indicate the existence of two deep mitochondrial lineages that so far are taxonomically unnamed, only in the geographically restricted study area in the north-central east coast – the population from Sahafina and the population from Tampolo. As a result of this, the taxon *C. nasutum* is paraphyletic and represents a complex of several cryptic species hidden under a single name. The different genetically distinct lineages assessed in the mtDNA and the nDNA dataset

Figure 6. Males of two additional candidate species of short-nosed chameleons from lowland localities of Madagascar's north-east and central-east, and of *Calumma fallax* (in life). (A) *Calumma* sp. aff. *nasutum*, Tampolo. (B) *C.* sp. aff. *nasutum*, Sahafina. (C) Male assigned to *C. fallax*, from a mid-elevation rainforest Tsinjoarivo, central Madagascar.

suggest a rather long independent evolutionary history. The fact that the genealogies concord insofar as all species and candidate species are reciprocally monophyletic for the mitochondrial gene and do not share haplotypes in the nuclear gene suggests that all of these units are independent evolutionary lineages, possibly to be recognised on the species level.

The description of *C. vohibola* adds a distinctive new species to the genus *Calumma* but is only a first step to resolving the cryptic species diversity of short-nosed chameleons from Madagascar. Based on molecular similarities, *C. vohibola* appears to be most closely related to *C. nasutum* and *C. fallax*. Besides molecular differences, only limited diagnostic morphological characters are known. One of them seems to be the length and shape of the rostral appendage that varies within this group between species and at the intraspecific level between sexes. The generally bigger size of the appendage in males and its shorter size or near-absence in females suggests that the structure is possibly related to mate recognition and driven by sexual selection, although it could also be operating at an interspecific level. Since it is largely known that chameleons are mainly visually orientated animals (Bowmaker *et al.* 2005; Stuart-Fox & Moussalli 2008), the length, shape and colouration of rostral appendages may be an important optical signal (Parcher 1974). Clearly the soft, dermal appendages of the *C. nasutum* group are not useful in rival fights, unlike the robust appendages in other Malagasy chameleon species (e.g. in the Malagasy *Calumma parsonii* and *Furcifer bifidus* groups, or in African species of the genus *Trioceros*). In Malagasy chameleons soft and colourful appendages only occur within this group, and in some of these species (e.g.

C. boettgeri or *C. gallus*) they bear a different and more conspicuous colouration than the rest of the head. Mostly these colours consist of different shades of blue or red, colours that are known to be highly reflecting in the ultraviolet spectrum, visible to the chameleon's eye (Bowmaker *et al.* 2005; Gehring & Witte 2007; Stuart-Fox & Moussalli 2008). Especially in the diffuse light of rainforests these conspicuous 'nose-flags' can play an important role in finding mates. Experiments of Parcher (1974) using *C. nasutum* from Andasibe revealed that females with a rostral appendage are more readily recognised by males as conspecifics than females with an amputated rostral appendage.

The very short or near-absent rostral appendage in *C. vohibola* is enigmatic and might be related to a rather open canopy in its littoral forest habitat. No other species of the *C. nasutum* complex was found during surveys in littoral forests (Raselimanana *et al.* 1998; Raselimanana 2005; Gehring *et al.* 2010; Randrianirina 2005), whereas in other nearby localities at least two syntopic species of the *C. nasutum* group occur (e.g. Vohidrazana, Sahafina), maybe resulting in a stronger selection towards a conspicuous and easily recognisable contour.

Despite the very short rostral appendage as a congruent morphological character in *C. vohibola*, the relevance of the unconstant presence of a dorsal crest in males of this species remains unclarified. While in the male holotype (ZSM 645/2009) and in the male specimen from Ankanin'ny Nofy (ZFMK 46112) a dorsal ridge composed of seven single upright spines is obviously present, the male paratype (ZSM 644/2009) completely lacks any signs of a dorsal ridge. Hillenius (1959) already points to the problem of the intraspecific variation (dorsal crest absent or present and number of dorsal scales) within *C. nasutum* populations, but does not provide any hypotheses for it at all. Further studies and a larger sample size of mature males of *C. vohibola* may throw light on this intraspecific variation.

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REFERENCES

ANDREONE, F., F. MATTIOLI, R. JESU & J.E. RANDRIANIRINA. 2001. Two new chameleons of the genus *Calumma* from north-east Madagascar, with observations on hemipenial morphology in the *Calumma furcifer* group (Reptilia, Squamata, Chamaeleonidae). Herpetol. J. 11: 53–68.

ANGEL, F. 1942. Les lezards de Madagascar. Mém. Acad. Malgache 36: 1–139.

BÖHME, W. 1997. Eine neue Chamäleonart aus der *Calumma gastrotaenia*-Verwandtschaft Ost-Madagaskars. Herpetofauna 19: 5–10.

BOETTGER, O. 1913. Reptilien und Amphibien von Madagascar, den Inseln und dem Festland Ostafrikas. Pp. 269–375, pl. 23–30. *In* A. VOELTZKOW (Ed.), Reise in Ostafrika in den Jahren 1903–1905, Wissenschaftliche Ergebnisse. Vol. 3. Schweizerbartsche Verlagsbuchhandlung, Stuttgart, Germany.

BOLLEN, A. & G. DONATI. 2005. Phenology of the littoral forest of Sainte Luce, southeastern Madagascar. Biotropica 37: 32–43.

BOWMAKER, J.K., E.R. LOEW & M. OTT. 2005. The cone photoreceptors and visual pigments of chameleons. J. Comp. Physiol. A. 9: 925–932.

BRUFORD, M.W., O. HANOTTE, J.F.Y. BROOKFIELD & T. BURKE. 1992. Single-locus and multilocus DNA fingerprint. Pp. 225–270. *In* A.R. HOELZEL (Ed.), Molecular Genetic Analysis of Populations: A Practical Approach. IRL Press, Oxford, UK.

BRYGOO, E.R. 1971. Reptiles sauriens Chamaeleonidae – genre *Chamaeleo*. Faune Madag. 33: 1–318.

BRYGOO, E.R. 1978. Reptiles sauriens Chamaeleonidae. Genre *Brookesia* et complément pour le genre *Chamaeleo*. Faune Madag. 47: 1–173.

CLEMENT, M., D. POSADA & K.A. CRANDALL. 2000. TCS: a computer program to estimate gene genealogies. Mol. Ecol. 9: 1657–1659.

DE GOUVENAIN, R.C. & J.A. SILANDER. 2003. Littoral forest. Pp. 103–111. *In* S.M. Goodman & J.P. Benstead (Eds.), The Natural History of Madagascar. The University of Chicago Press, London.

DUMÉRIL, A.M.C. & G. BIBRON. 1836. Erpetologie Générale ou Histoire Naturelle Complete des Reptiles Vol. 3. Librairie Encyclopédique Roret, Paris.

GANZHORN, J.U., S.M. GOODMAN, J.B. RAMANAMANJATO, D. RAKOTONDAVONY, B. RAKOTOSAMIMANANA & D. VALLAN. 2000. Vertebrate species in fragmented littoral forest of Madagascar. Pp. 155–164. *In* W.R. LOURENÇO & S.M. GOODMAN (Eds.), Diversity and endemism in Madagascar. Mémoires de la Société de Biogéographie Edition. Museum Histoire Naturelle, Paris.

GEHRING, P.-S., M. PABIJAN, F.M. RATSOAVINA, J. KÖHLER, M. VENCES & F. GLAW. 2010. A Tarzan yell for conservation: a new chameleon, *Calumma tarzan* sp. n., proposed as a flagship species for the creation of new nature reserves in Madagascar. Salamandra 46: 167–179.

GEHRING, P.-S., F.M. RATSOAVINA & M. VENCES. 2010. Filling the gaps – amphibian and reptile records from lowland rainforests in eastern Madagascar. Salamandra 46: 214–234.

GEHRING, P.-S. & K. WITTE. 2007. Ultraviolet reflectance in Malagasy chameleons of the genus *Furcifer* (Squamanta: Chamaeleonidae). Salamandra 43: 43–48.

GLAW, F., J. KÖHLER & M. VENCES. 2009. A distinctive new species of chameleon of the genus *Furcifer* (Squamata: Chamaeleonidae) from the Montagne d'Ambre rainforest of northern Madagascar. Zootaxa 2269: 32–42.

GLAW, F. & M. VENCES. 2007. A field guide to the amphibians and reptiles of Madagascar, 3rd ed. Vences & Glaw Verlag, Cologne, Germany.

GU, X. & W.-H. LI. 1996. Bias-corrected paralinear and LogDet distances and tests of molecular clocks and phylogenies under nonstationary nucleotide frequencies. Mol. Biol. Evol. 13: 1375–1383.

HAN, D., K. ZHOU & A.M. BAUER. 2004. Phylogenetic relationships among gekkotan lizards inferred from *Cmos* nuclear DNA sequences and a new classification of the Gekkota. Biol. J. Linn. Soc. 83: 353–368.

HILLENIUS, D. 1959. The differentiation within the genus *Chamaeleo* Laurenti, 1768. Beaufortia 89: 1–92.

HUELSENBECK, J.P. & F. RONQUIST. 2001. MRBAYES: Bayesian inference of phylogeny. Bioinformatics 17: 754–755.

IUCN. 2001. IUCN red list categories. Version 3.1. Species Survival Commission, IUCN, Gland, Switzerland and Cambridge, UK.

KLAVER, C. & W. BÖHME. 1997. Chamaeleonidae. Das Tierreich. Part 112. Verlag Walter de Gruyter & Co, Berlin and New York.

LOCKHART, P.J., M.A. STEEL, M.D. HENDY & D. PENNY. 1994. Recovering evolutionary trees under a more realistic model of sequence evolution. Mol. Biol. Evol. 11: 605–612.

MACEY, J.R., A. LARSON, N.B. ANANJEVA, Z. FANG & T.J. PAPENFUSS. 1997. Two novel gene orders and the role of light-strand replication in rearrangement of the vertebrate mitochondrial genome. Mol. Biol. Evol. 14: 91–104.

MACEY, J.R., J.A. SCHULTE II, A. LARSON, N.B. ANANJEVA, Y. WANG, R. PETHIYAGODA, N. RASTEGAR-POUYANI & T.J. PAPENFUSS 2000. Evaluating trans-Tethys migration: an example using acrodont lizard phylogenetics. Syst. Biol. 49: 233–256.

MERTENS, R. 1933. Die Reptilien der Madagaskar-Expedition Prof. Dr. H. Bluntschli's. Senckenb. Biol. 15: 260–274.

MOCQUARD, M.F. 1900a. Diagnoses d'espèces nouvelles de reptiles de Madagascar. Bull. Mus. Natl. Hist. Nat. 6: 345–348.

MOCQUARD, M.F. 1900b. Nouvelle contribution à la faune herpétologique de Madagascar. Bull. Soc. Philomat. Paris 9: 93–111.

NYLANDER, J.A.A. 2004. MrModeltest v2. Program distributed by the author, Evolutionary Biology Centre, Uppsala University.

PADIAL, J.M., A. MIRALLES, I. DE LA RIVA & M. VENCES. 2010. The integrative future of taxonomy. Front. Zool. 7(May): 16.

PARCHER, S.R. 1974. Observations on the natural histories of six Malagasy Chamaeleontidae. Z. Tierpsychol. 34: 500–523.

POSADA, D. & K.A. CRANDALL. 1998. Modeltest: testing the model of DNA substitution. Bioinformatics 14: 817–818.

RANDRIANIRINA, J.E. 2005. Inventaire des reptiles et amphibians de Vohibola. Pp. 4–15. *In* J.E. RANDRIANIRINA, D. RANDRIANJAFINIASA, F. RAHOLIARISENDRA, C. RAFAMANTANANTSOA & J.B. RAMILANIRINA (Eds.), Inventaire biologique de la foret littorale de Vohibola, Madagascar (Amphibiens, Reptiles, Oiseaux, Lémuriens et Insectes). Unpubl. report by L' Homme et l' environnement.

RASELIMANANA, A.P. 2005. Suivi de l' herpétofaune. Pp. 105–120. *In* J. RATSIRARSON & S.M. GOODMAN (Eds.), Suivi de biodiversité de la foret littorale de Tampolo. Rech. Develop. Sér. Sci. Biol. 22: 1–134 (Ministère de la Recherche Scientifique, Antananarivo, Madagascar).

RASELIMANANA, A.P., D. RAKOTOMALALA & F. RAKOTONDRAPARANY. 1998. Les reptiles et amphibiens: diversité et conservations. Pp. 183–195. *In* J. RATSIRARSON & S.M. GOODMAN (Eds.), Inventaire biologique de la foret littorale de Tampolo (Fenoarivo Atsinanana). Rech. Develop. Sér. Sci. Biol. 14: 1–261 (Ministère de la Recherche Scientifique, Antananarivo, Madagascar).

RAXWORTHY, C.J., M.R.J. FORSTNER & R.A. NUSSBAUM. 2002. Chameleon radiation by oceanic dispersal. Nature 415: 784–487.

RAXWORTHY, C.J. & R.A. NUSSBAUM. 2006. Six new species of occipital-lobed *Calumma* chameleons (Squamata: Chamaeleonidae) from montane regions of Madagascar, with a new description and revision of *Calumma brevicorne*. Copeia 2006: 711–734.

STEPHENS, M. & P. SCHEET. 2005. Accounting for decay of linkage disequilibrium in haplotype inference and missing data imputation. Am. J. Human Genet. 76: 449–462.

STEPHENS, M., N. SMITH & P. DONNELLY. 2001. A new statistical method for haplotype reconstruction from population data. Am. J. Human Genet. 68: 978–989.

STUART-FOX, D. & A. MOUSSALLI. 2008. Selection for social signalling drives the evolution of chameleon colour change. PLoS Biol. 6: e25.

SWOFFORD, D.L. 2002. PAUP*. Phylogenetic Analysis Using Parsimony (*and Other Methods). Version 4. Sinauer Associates, Sunderland, MA.

TAMURA, K., J. DUDLEY, M. NEI & S. KUMAR. 2007. *MEGA4*: Molecular Evolutionary Genetics Analysis (MEGA) software version 4.0. Mol. Biol. Evol. 24: 1596–1599.

TEMPLETON, A.R., K.A. CRANDALL & C.F. SING. 1992. A cladistic analysis of phenotypic associations with haplotypes inferred from restriction endonuclease mapping and DNA sequence data. III. Cladogramm estimation. Genetics 132: 619–633.

TOLLEY, K., C.R. TILBURY, W.R. BRANCH & C.A. MATTHEE. 2004. Phylogenetics of the southern African dwarf chameleons, *Bradypodion* (Squamata: Chamaeleonidae). Mol. Phyl. Evol. 30: 354–365.

TOWNSEND, T. & A. LARSON. 2002. Molecular phylogeny and mitochondrial genomic evolution in the Chamaeleonidae (Reptilia, Squamata). Mol. Phylogenet. Evol. 23: 22–36.

TOWNSEND, T.M., K.A. TOLLEY, F. GLAW, W. BÖHME & M. VENCES. 2011. Eastward from Africa: palaeocurrent-mediated chameleon dispersal to the Seychelles islands. Biol. Letters 7: 225–228.

VIEITES, D.R., K.C. WOLLENBERG, F. ANDREONE, J. KÖHLER, F. GLAW & M. VENCES. 2009. Vast underestimation of Madagascar's biodiversity evidenced by an integrative amphibian inventory. Proc. Nat. Acad. Sci. USA. 106: 8267–8272.

VIETTE, P. 1991. Principales localités où des insectes ont été recueillis à Madagascar. Faune Madag. Suppl. 2: 1–88.

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Table 1. List of Genbank accession numbers and field numbers of specimens used in the present study.

Gene	Gen Bank accession number	Field number	Species
ND2	JN030471	PSG113	*C. vohibola*
Cmos	JN030454		
ND2	JN030473	PSG205	*C. vohibola*
Cmos	JN030459		
ND2	JN030474	PSG104	*C. vohibola*
Cmos	JN030456		
ND2	JN030475	PSG105	*C. vohibola*
Cmos	JN030457		
ND2	JN030476	PSG116	*C. vohibola*
Cmos	JN030458		
ND2	JN030478	PSG233	*C. vohibola*
Cmos	JN030461		
ND2	JN030477	PSG140	*C. vohibola*
Cmos	JN030460		
ND2	JN030479	ZCMV8915	*C. vohibola*
Cmos	JN030462		
ND2	JN030480	ZCMV8917	*C. vohibola*
Cmos	–		
ND2	JN030481	ZCMV8916	*C. vohibola*
Cmos	JN030455		
ND2	JN030482	FGMV_2002_984	*C. nasutum*
Cmos	JN030463		
ND2	JN030483	PSG321	*C.* aff. *nasutum*
Cmos	JN030466	PSG777	*C.* aff. *nasutum*
ND2	JN030484	PSG319	*C.* aff. *nasutum*
Cmos	JN030464		
ND2	JN030485	PSG765	*C.* aff. *nasutum*
Cmos	JN030465		
ND2	JN030486	FGZC4588	*C. fallax*
Cmos	JN030468		
ND2	JN030487	PSG318	*C. gallus*
Cmos	JN030470		
ND2	JN030488	PSG262	*C. gallus*
Cmos	JN030467		
ND2	JN030472	ZCMV152	*C.* cf. *gallus*
Cmos	JN030469		
ND2	JN030489	FGMV2002_65	*F. lateralis*