

The herpetofauna (Amphibia, Crocodylia, Squamata, Testudines) of the Isalo Massif, Southwest Madagascar: combining morphological, molecular and museum data

WALTER COCCA¹, GONÇALO M. ROSA^{2,3}, FRANCO ANDREONE⁴, GENNARO APREA⁵, PAOLO EUSEBIO BERGÒ⁴,
FABIO MATTIOLI⁶, VINCENZO MERCURIO⁷, JASMIN EMILE RANDRIANIRINA⁸, DANIELA ROSADO¹,
MIGUEL VENCES⁹ & ANGELICA CROTTINI¹

¹) CIBIO, Research Centre in Biodiversity and Genetic Resources, InBIO, Universidade do Porto, Campus Agrário de Vairão,
Rua Padre Armando Quintas, No 7, 4485-661 Vairão, Portugal

²) Institute of Zoology, Zoological Society of London, Regent's Park, NW1 4RY London, UK

³) Centre for Ecology, Evolution and Environmental Changes (CE3C), Faculdade de Ciências da Universidade de Lisboa,
Bloco C2, Campo Grande, 1749-016 Lisboa, Portugal

⁴) Museo Regionale di Scienze Naturali, Via G. Giolitti, 36, 10123 Torino, Italy

⁵) Dipartimento di Biologia, Università degli Studi di Napoli Federico II, Via Cinthia, 80126 Napoli, Italy

⁶) Acquario di Genova, Area Porto Antico, Ponte Spinola, 16128 Genova, Italy

⁷) Museum für Naturkunde, Leibniz-Institut für Evolutions- und Biodiversitätsforschung an der Humboldt-Universität zu Berlin,
Invalidenstr. 43, 10115 Berlin, Germany

⁸) Parc Botanique et Zoologique de Tsimbazaza, BP 4096, Antananarivo 101, Madagascar

⁹) Technische Universität Braunschweig, Zoological Institute, Mendelssohnstr. 4, 38106 Braunschweig, Germany

Corresponding author: WALTER COCCA, e-mail: walter.cocca85@gmail.com

Manuscript received: 28 November 2017

Accepted: 19 March 2018 by JÖRN KÖHLER

Abstract. This study summarizes the data of multiple surveys carried out at the Isalo Massif (southwestern Madagascar), both within and outside the National Park limits between 2004 and 2014, providing an updated list of the amphibians and non-avian reptiles present within the area. We used an integrative taxonomic approach, combining morphological and molecular data sets to identify collected samples as either described species, or as undescribed candidate species. We here report on one new amphibian candidate species (*Mantidactylus* sp. aff. *multiplicatus* Ca65 “Isalo”), and two new amphibian distribution records (*Mantella* sp. aff. *expectata* “South”, *Scaphiophryne* sp. aff. *calcarata* Cao2) compared to the former inventories of the area. Additionally, we provide molecular reference sequences for 23 of the 24 amphibian taxa and for 40 of the 47 reptile taxa known to occur in the area, combining all available sources. With this study, we validate Isalo as an important centre of biodiversity and micro-endemism within Madagascar, with four described endemic amphibian (*Gephyromantis azzurrae*, *Mantella expectata*, *Mantidactylus noralotae*, and *Scaphiophryne gottlebei*) and one reptile species (*Trachylepis nancycoutuae*). Of the taxa here listed for Isalo, seven amphibians and six reptiles are new candidate species, and among them at least one amphibian (*Mantidactylus* sp. aff. *multiplicatus* Ca65 “Isalo”) and three reptiles (*Lygodactylus* sp. aff. *tuberosus* Cao2 “Isalo”, *Paroedura* sp. aff. *bastardi* Cao2 “Isalo” and *P.* sp. aff. *bastardi* Cao3 “Ilakaka”) are currently known only from Isalo. The unique biodiversity of the Isalo Massif was likely favoured by the confluence of different biomes in the area and/or its role as a refugium during palaeoclimatic oscillations. Our study highlights the crucial importance of applying molecular taxonomic identification in updating the species lists even of previously well surveyed areas, and of maintaining and increasing museum voucher collections as a permanent and accessible testimony of the biodiversity inventory.

Key words. Dry forest, museum specimens, species survey, integrative taxonomy, candidate species, 16S rRNA, COI.

Introduction

Madagascar is one of the most important hotspots of biodiversity worldwide (MYERS et al. 2000, GOODMAN & BENSTEAD 2005, KREMEN et al. 2008, VIEITES et al. 2009, AY-

MOZ et al. 2013). The island is characterized by an incredibly high faunal endemism rate, ranging from 50% in birds to 100% in native amphibians (GLAW & VENCES 2007, PERL et al. 2014), and with about 400 non-marine squamate species only found in Madagascar (GLAW & VENCES 2007, JENKINS

et al. 2014). Madagascar is characterized by a strong climatic heterogeneity and it is roughly divided from north to south into a humid east and an arid west (VENCES et al. 2009, BROWN et al. 2016). Five broad bioclimatic zones are currently recognized: the eastern humid rainforest belt, the sub-humid highlands, the western dry deciduous forests, the southwestern sub-arid spiny forests, and the montane grasslands and shrublands (SCHATZ 2000, GANZHORN et al. 2001, GLAW & VENCES 2007, VENCES et al. 2009). The climate in the western part of the island is overall hot and dry, with a cooler-drier and a warmer-wetter season that each last 5–7 months (GOODMAN & BENSTEAD 2003). This climate supports a diversity of sclerophyllous deciduous forests, open woodland and savannas that change in composition from northern subhumid areas to southern spiny bushland (GOODMAN & BENSTEAD 2003). Diversity patterns of the herpetofauna largely reflect this subdivision, with bioclimatic boundaries coinciding with major areas of biotic turnover (BROWN et al. 2014, 2016). Dry deciduous forest in Madagascar has been experiencing a dramatic decrease in the last decades and, since the 1970s, has been reduced by almost 40% of its original extent (WAEBER et al. 2015). Knowledge is limited on the fauna and flora of this biome, and even less is known about their biology and ecological interactions.

While the richness of amphibians peaks in the eastern and northern rainforest areas (e.g., more than 100 amphibian species are known to occur in Ranomafana National Park, 80 in Betampona Strict Nature Reserve, and at least 70 in Marojejy National Park; VIEITES et al. 2009, ROSA et al. 2012, GOODMAN & WILMÉ 2003), the reptiles also exhibit high species richness in western and southwestern Madagascar (RASELIMANANA 2008, BROWN et al. 2016). In contrast, the patterns of local endemism appear to be distinctly higher, for both amphibians and reptiles, in the north (D'CRUZE et al. 2008, BROWN et al. 2016).

An analysis of the herpetofaunal diversity of the dry forests of western Madagascar (RASELIMANANA 2008) has confirmed this bioregion as an important centre for species diversity and local endemism. At least 30% of the island's named herpetofauna occurs in this region, with areas of higher habitat heterogeneity reaching distinctly higher species richness values (RASELIMANANA 2008, BORA et al. 2010, GARDNER et al. 2015, BROWN et al. 2016). For example, in the Tsingy de Bemaraha Massif, 19 species of amphibians and 60 of reptiles have been recorded, and of these, five amphibians and 18 reptile species are considered local endemics (BORA et al. 2010).

One other important area known to be highly heterogeneous in terms of available habitats and hosting many local endemic species is the Isalo Massif (MERCURIO et al. 2008). Part of this area is included in the Parc National de l'Isalo, established in its current borders in 1962 (GOODMAN & BENSTEAD 2003). In this study we update the previously published inventory of the amphibians of this area (MERCURIO et al. 2008), and report on the known reptiles (Squamata, Testudines and Crocodylia) inhabiting this area, complementing the former, unpublished inventory on the

herpetofauna of Isalo by HAWKINS (1994). We used an integrative taxonomic approach, combining the external morphology of specimens, photographs in life, and DNA sequences to assign collected/observed amphibians and reptiles to either described species or undescribed candidate species. To allow these data to be as informative as possible for a better management of the area's unique biodiversity, we also provide a list of the available voucher specimens hosted in the herpetological collection of the Museo Regionale di Scienze Naturali di Torino (Italy). The term 'reptiles' is used in this study as referring to non-avian reptiles only, i.e., in this case vertebrates in the orders Squamata, Testudines and Crocodylia.

Material and methods

Study site

The Isalo Massif is situated in the southwestern corner of the Province of Fianarantsoa (Fianarantsoa Faritany) and hosts the Parc National de l'Isalo, one of the largest protected areas of Madagascar (81,540 ha). Elevation ranges between 510 and 1268 m, with narrow canyons of up to 200 m deep, most of which occur in the eastern and northwestern sectors of the area. This area is characterized by the presence of sandstone outcrops and rocks with a mosaic of three main habitat types: savannas, open valleys, and narrow canyons (MERCURIO & ANDREONE 2006). Savannas are expansive prairies that are repeatedly exposed to fire during the dry seasons, with isolated trees and forested spots. Humidity is low and temperature fluctuations between night and day are high. In this kind of habitat, rivers and pools are usually temporary, fed by seasonal rains, and dry for most of the year. The open valleys are usually vegetated by gallery forests that vary in dimension. Here the aquatic habitats consist of permanent or semi-permanent torrents with both cascades and pools being present. The montane, rocky part of the massif is intersected by canyons of different dimensions and depths in which water is present to a variable extent. In these humid canyons, vegetation is absent or limited to few isolated trees due to the scarcity of light. The climate is dry tropical, with 90% of precipitation occurring between November and March and with mean temperatures varying between 17°C in June and 25°C in February (MERCURIO et al. 2008).

Survey methods

Multiple surveys were carried out in Isalo, both within and outside the National Park limits, between 2004 and 2014 (Fig. 1, Supplementary Table S1). Most of our sampling took place during the rainy season, when the activity of most species is at its highest. Sampling was conducted opportunistically during both day and night, using headlamps and hand-held torchlights, in a quest to explore all available habitats along transects or trails and by visiting seasonal streams and temporary ponds. Our search for amphibians

also included locating vocalising males. Locality data were recorded using a GPS receiver (Supplementary Table S1).

Most animals were photographed with analogical and/or digital cameras to document their coloration in life and subsequently released at the place of capture. For molecular analyses, toe clips of adult amphibians, fin tips of tadpoles, and tissue samples of reptiles were usually collected and stored in 99% ethanol. If individuals were suspected of possibly belonging to new and undescribed taxa, a limited number of specimens were collected as vouchers. They were anaesthetized by immersion in (amphibians), or injection with (reptiles) MS222, followed by euthanasia with an overdose of MS222. They were then fixed in 10% formalin or 90% ethanol and later transferred to 65–70% ethanol for long-term storage. The lists of tissue samples and voucher specimens analysed for this study are provided in Supplementary Tables S2–S5.

Species identification in the field was carried out mostly using the descriptions provided by GLAW & VENCES (2007), and additional photographic records from scientific publications. The final inventory of amphibian and reptile species of the Isalo Massif is given as a compilation of the work carried out by our team and complemented by records retrieved from other publications for species not found by us (GLAW & VENCES 1994, HAWKINS 1994, PUENTE et al. 2005, CROTTINI et al. 2008, MERCURIO et al. 2008).

Toponyms used in this study are those more consistently used in the literature (Supplementary Table S1). In most cases, anyhow, they correspond to informal local names.

Molecular species identification

Total genomic DNA was extracted from tissue samples using proteinase K digestion (10 mg/ml concentration) followed by a standard high-salt extraction method (BRUFORD et al. 1992). A fragment of ca. 550 bp of the 3' terminus of the mitochondrial 16S rRNA gene (hereafter referred to as 16S), proven to be suitable for amphibian identification (VENCES et al. 2005a), was amplified for 159 samples, while a fragment of around 650 bp of the standard barcoding region of the cytochrome oxidase I gene (COI; NAGY et al. 2012) was amplified for 73 reptile samples. For three amphibian samples, it was not possible to amplify the 16S fragment, and in these instances other mitochondrial gene fragments were used, i.e., 16S 5' terminus, or cytochrome b (Cytb). The molecular taxonomic identification using the COI fragment was not possible for some reptile species due to missing reference sequences. In these instances, other mitochondrial gene fragments (16S, ND1, ND2, ND4, Cytb) were used for identification (see Supplementary Table S2 and S4 for details and GenBank acces-

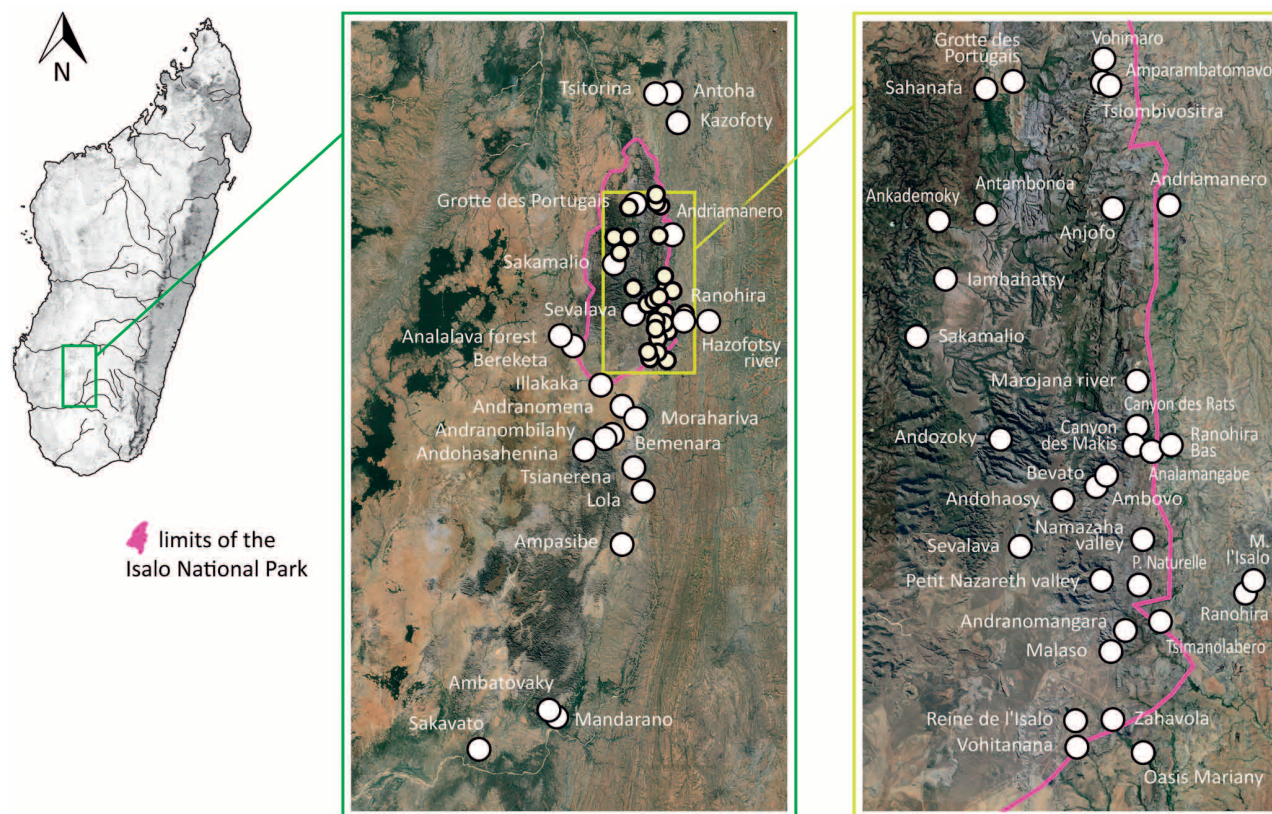


Figure 1. Maps of the study area with all sampling localities for both amphibians and reptiles. Andravaka not included because GPS data were not available and this site could therefore not be georeferenced unambiguously.

sion numbers of newly determined sequences [KX066545–KX066697, MH063278–MH063431], and Supplementary Table S6 for primers and cycling protocols). All fragments were sequenced using an ABI 3730XL automated sequencer at Macrogen Inc. Chromatograms were checked and sequences were manually edited where necessary, using the sequence alignment editor of BioEdit (V.7.2.0; HALL 1999).

To assess taxon identity, sequences of each morphologically identified taxon were compared among each other and each sequence was compared using the Basic Local Alignment Search Tool (BLAST, <http://blast.ncbi.nlm.nih.gov/Blast.cgi>, ALTSCHUL et al. 1997) with all homologous sequences available in GenBank (<http://www.ncbi.nlm.nih.gov/GenBank/>), using the nucleotide blast (nblast) option with default parameters. Average genetic distances within and between species were calculated by uncorrected pairwise genetic distance estimations (uncorrected p-distance) using MEGA 7 (KUMAR et al. 2016; Supplementary Table S7 and S8).

Some specimens could not be assigned to any described or identified candidate species (VIEITES et al. 2009, PERL et al. 2014, NAGY et al. 2012). For these taxa, we applied the terms and abbreviations, confirmed candidate species (CCS), unconfirmed candidate species (UCS), or deep conspecific lineage (DCL) as defined by VIEITES et al. (2009). Our denominations of already identified candidate species follow PERL et al. (2014) for amphibians and NAGY et al. (2012) for reptiles, although we decided to complement these working names with information on the morphologically closest described species by prefixing the species epithet with “sp. aff.” and a descriptor of the locality from where the taxon is known (this toponym was added in cases of species known only from Isalo) or of a characteristic trait of the candidate species.

Candidate species of amphibians were identified based on a threshold of 4% minimum divergence for the 16S gene fragment (see also VENCES et al. 2005b, FOUQUET et al. 2007, VIEITES et al. 2009). Candidate species of reptiles were identified following the different thresholds proposed for the different groups by NAGY et al. (2012). The sequences of the other gene fragments were used to refine species identification within a known species group and there was no need to apply a minimum threshold.

In this study, we also provide updated distributional range information within the Isalo Massif for amphibians based on locality records from museum specimens, molecular data, GenBank, and literature (mostly GLAW & VENCES 2007, MERCURIO et al. 2008; Supplementary Table S3). For reptiles, the distribution of each species includes locality records from museum specimens, molecular data, GenBank, and literature (mostly GLAW & VENCES 2007, HAWKINS 1994; Supplementary Table S5).

Museum specimens

Most of the specimens collected during our multiple surveys in Isalo have been deposited in the herpetological col-

lections of the Museo Regionale di Scienze Naturali (Turin, Italy) and the Parc Botanique et Zoologique de Tsimbazaza (Antananarivo, Madagascar). We refer to these specimens using their collection numbers preceded by acronyms MRSN and PBZT, respectively. In addition to these, we provide taxonomic information on some additional specimens hosted in various collections. For these records, we use their catalogue numbers preceded by their institution acronyms (SMF, Senckenberg Museum Frankfurt, Germany; UADBA, Université d'Antananarivo, Mention Zoologie et Biodiversité Animale, Antananarivo, Madagascar; ZMA, Zoologisch Museum Amsterdam, The Netherlands (collections currently in the process of being integrated in the Naturalis Biodiversity Center of Leiden); UMMZ, Museum of Zoology at the University of Michigan; ZSM, Zoologische Staatssammlung München, Germany). Field number acronyms are FAZC (Franco Andreone Zoological Collection) and ACZCV (Angelica Crottini Zoological Collection Voucher). In instances where specimens have already been deposited in institutional collections, but are not yet catalogued, they are given as FAZC/MRSN (for specimens in Turin) and FAZC/PBZT (for specimens in Antananarivo), respectively.

Specimens hosted in the MRSN collection (Supplementary Table S3 and S5) were visually inspected and their taxonomic identification was reassessed based on the results obtained in this study. A careful morphological inspection and verification of diagnostic traits was performed when molecular data were insufficient to confirm species identity.

Results

In total, 24 amphibian and 47 reptile species were recorded in Isalo by combining all available data sources, and we here provide DNA sequences for almost all of these species. Like MERCURIO et al. (2008), we failed to detect in the field *Scaphiophryne menabensis* that, at the moment, remains known from the area by a single voucher specimen (UMMZ 227489, University of Michigan, Museum of Zoology; VENCES et al. 2003, GLOS et al. 2005).

Among the amphibians, 16 taxa (65%) belong to the family Mantellidae while six (26%) are Microhylidae. Hyperolidae and Ptychadenidae are present with only one species each. We confirm the presence of 20 (out of the 21) amphibian species listed by MERCURIO et al. (2008) and provide evidence for the occurrence of one new candidate species (*Mantidactylus* sp. aff. *multiplicatus* Ca65 “Isalo”). We also report two new amphibian distribution records (*Mantella* sp. aff. *expectata* “South” and *Scaphiophryne* sp. aff. *calcarata* Cao2) compared to the previous inventories of the area. Four of the amphibian species and one candidate species seem to be Isalo endemics: *Gephyromantis azzurrae*, *Mantella expectata*, *Scaphiophryne gottlebei*, *Mantidactylus noralottae* and *M.* sp. aff. *multiplicatus* Ca65 “Isalo”. In this study, *Gephyromantis corvus* is tentatively not considered microendemic to Isalo, because there is a

record of this species from the Makay Massif (RAKOTONDRAVONY & GOODMAN 2011), even though this record is in need of confirmation and might turn out to represent a different species of the subgenus *Phylacomantis*. Finally, we report new locality records for ten amphibian taxa within the Isalo Massif: *Scaphiophryne* sp. aff. *calcarata* Ca01, *Dyscophus insularis*, *Boophis occidentalis*, *Laliostoma labrosum*, *Blommersia* sp. aff. *wittei* Ca05 “Isalo”, *Gephyromantis corvus*, *G. azzurrae*, *Mantidactylus* sp. aff. *ulcerosus* Ca14 “Isalo”, *M. noralottae*, and *M. sp. aff. cowani* Ca48 “small” (see Supplementary Table S3).

Our molecular and morphological analyses of specimen MRSN A5313 (part of the type series of *G. azzurrae* MERCURIO & ANDREONE 2007) suggest this specimen was wrongly assigned to *G. azzurrae* and we here propose to refer it to *G. corvus* (Fig. 4, Supplementary Table S2 and S3). This specimen was not analysed genetically by MERCURIO & ANDREONE (2007) when describing *G. azzurrae* and was erroneously assigned to this species.

The reptile species of Isalo belong to 10 of the 15 families known from the island (Supplementary Table S4 and S5). Most abundant are Gekkonidae and Lamprophiidae, which are represented by 13 and 10 species, respectively (or 27 and 21% of the total). Isalo represents an expansion of the known range for six taxa (*Crocodylus niloticus*, *Paroedura* sp. aff. *bastardi* Ca01 “Marofandilia/Miandrivazo”, *Trachylepis gravenhorstii* lineage 4b, *O. saxicola*, *Lycodryas guentheri*, and *L. sp.* Ca02 “Andohahela”). A total of six species are probably undescribed (*Lygodactylus* sp. aff. *pictus* Ca01 “Isalo”, *L. sp. aff. tuberosus* Ca02 “Isalo”, *Paroedura* sp. aff. *bastardi* Ca01 “Marofandilia/Miandrivazo”, *P. sp. aff. bastardi* Ca02 “Isalo”, *P. sp. aff. bastardi* Ca03 “Ilakaka”, and *L. sp. aff. pseudogranuliceps* Ca01 “Andohahela”); of these, three taxa are known only from here and might be micro-endemic to this massif (*Lygodactylus* sp. aff. *tuberosus* Ca02 “Isalo”, *Paroedura* sp. aff. *bastardi* Ca02 “Isalo”, and *P. sp. aff. bastardi* Ca03 “Ilakaka”). Note that *Lygodactylus* sp. aff. *pictus* Ca01 “Isalo” is excluded from this list, because in this

study we report this taxon also from the nearby Zombitse-Vohibasia National Park (details follow). Beside these, the only other reptile species endemic to Isalo is *Trachylepis nancycoutuae*.

To complement the work of MERCURIO et al. (2008), we here report on the distribution, habitat preferences, and intraspecific genetic variability of the new amphibian candidate species *Mantidactylus* sp. aff. *multiplicatus* Ca65 “Isalo” and the newly recorded *Scaphiophryne* sp. aff. *calcarata* Ca02 and *Mantella* sp. aff. *expectata* “South”, and subsequently provide an annotated species list of the Isalo reptiles.

Species accounts

Amphibians

Mantidactylus (Brygoomantis) sp. aff. multiplicatus Ca65 “Isalo” (UCS)

Fig. 2A

Locality records: Andohasahenina (MRSN A2964), Andriamane-ro (MRSN A6893, MRSN A6885, ACZCV 0281–0284), Namazaha Valley (also referred to as Namaza) (MRSN A2689, MRSN A2878, MRSN A6889, MRSN A6901), Piscine Naturelle.

This undescribed species represents a new record for Madagascar and could represent a new example of micro-endemism in Isalo. This lineage is phylogenetically close to *M. multiplicatus*, *M. sp. aff. betsileanus* Ca29 “Tolagnaro”, *M. sp. aff. betsileanus* Ca30 “Andohahela”, and the sympatric *M. noralottae*. Note that a formal resurrection of *M. multiplicatus* BOETTGER, 1913 is still needed, but this species name was already used by POTH et al. (2012) anticipating an upcoming taxonomic revision. The genetic distance between *M. sp. aff. multiplicatus* Ca65 “Isalo” and all the other species of the aforementioned group is ca. 4% (16S). This UCS represents the third *Brygoomantis* from Isalo, with the other two being *M. noralottae* and *M. sp.*

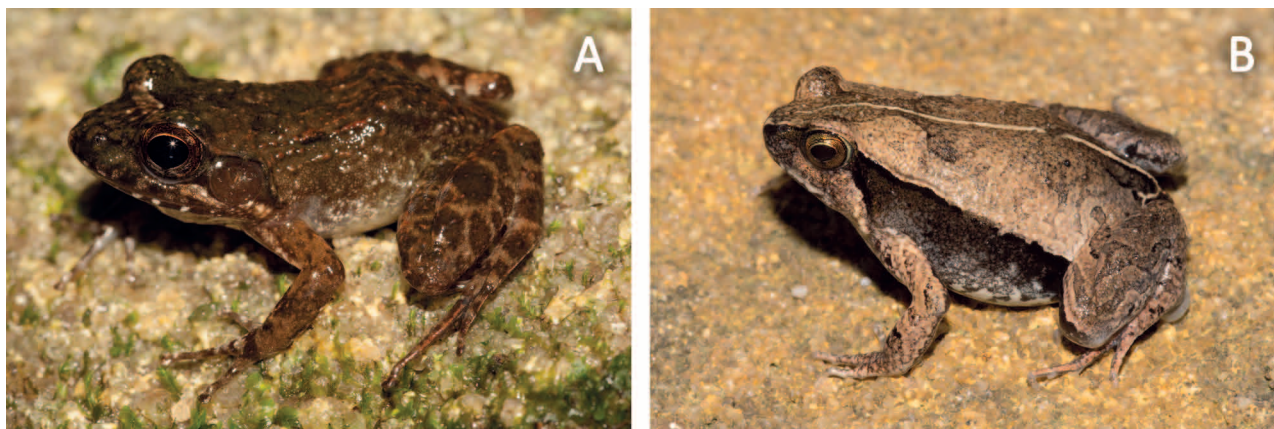


Figure 2. Two new amphibian records from the Isalo region: A) An adult male of *Mantidactylus* sp. aff. *multiplicatus* Ca65 “Isalo” (FAZC 14351) in dorsolateral view from Piscine Naturelle, Petit Nazareth (30 November 2009, not collected); B) adult *Scaphiophryne* sp. aff. *calcarata* Ca02 (MRSN A6895) in dorsolateral view from Ranohira (22 January 2011).

aff. *ulcerosus* Ca14 “Isalo”. New surveys followed up with molecular taxonomic identification of the samples will be needed to define its extent of occurrence in Isalo due to its superficial morphological similarity with *M. noralotiae*.

Scaphiophryne sp. aff. *calcarata* Cao2 (CCS)
Fig. 2B

Locality record: Ranohira (MRSN A6895).

The name *S. calcarata* (MOCQUARD, 1895) has not yet been unequivocally assigned to any population of *Scaphiophryne*, and three deeply divergent mitochondrial lineages exist in Madagascar: *S. sp. aff. calcarata* Cao1 (or *S. sp.*

aff. *calcarata* B; known from Tolagnaro and Berenty), *S. sp. aff. calcarata* Cao2 (or *S. sp. aff. calcarata* A; known from Kirindy and Menabe) (terminology follows VIEITES et al. [2009] and PERL et al. [2014], respectively), and *S. sp. aff. calcarata* Ankarafantsika (PERL et al. 2014). *S. sp. aff. calcarata* Cao1 was previously reported as *S. calcarata* by MERCURIO et al. (2008), and we here also report the occurrence of *S. sp. aff. calcarata* Cao2 (16S p-distance between populations from Kirindy and Isalo ca. 1%), revealing Isalo as a contact zone between these two taxa. The molecular divergence between *S. sp. Cao1* and *S. sp. Cao2* (ca. 4%; 16S) is supported by bioacoustic data (calls from Kirindy and Berenty are highly divergent, GLAW & VENCES [2007]), but no calls are known from Ankarafantsika or Isalo. The population from Ankarafantsika shows a 2% genetic dis-

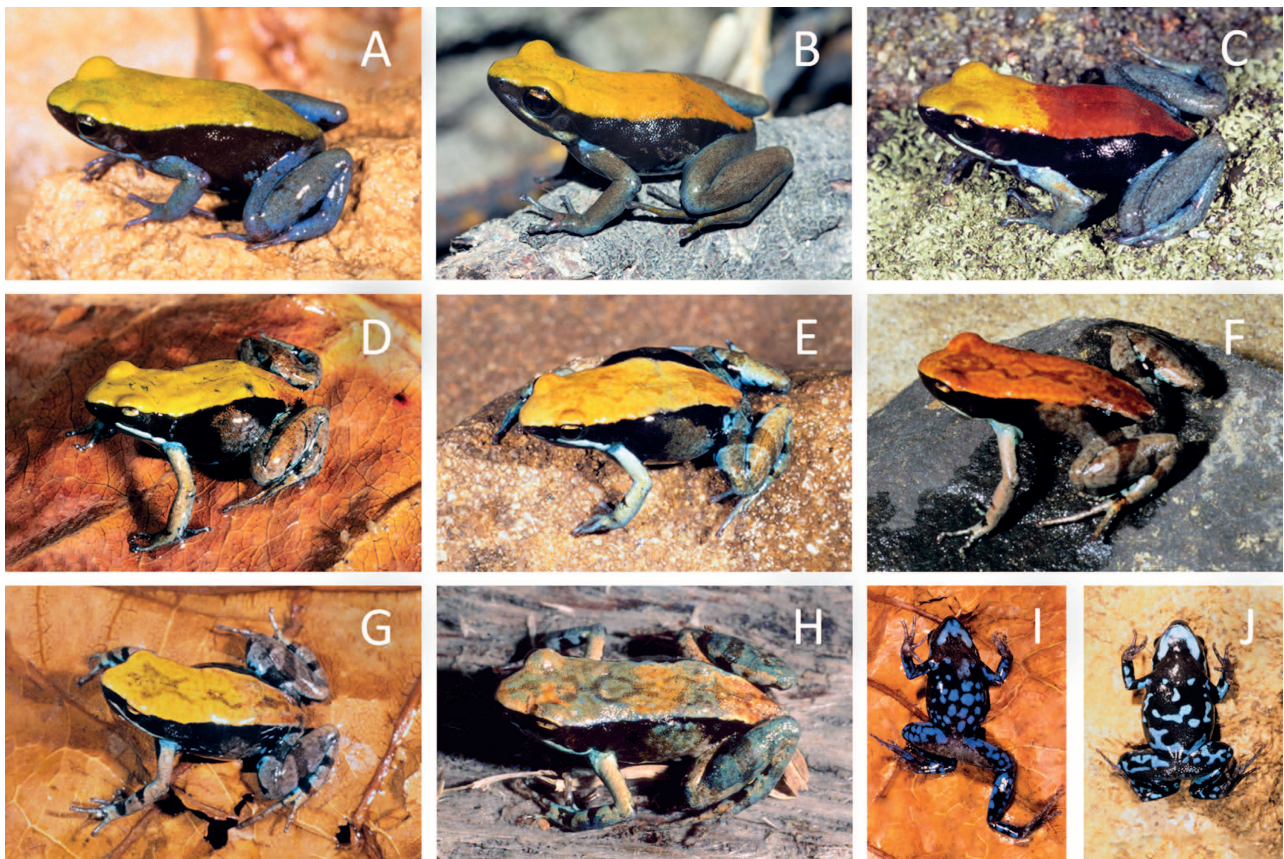


Figure 3. Diversity of *Mantella* species and corresponding morphotypes found in the Isalo Massif: A) Individual of *M. expectata* from Sakavato with typical coloration (MRSN A5204); B) individual of *M. expectata* from Malaso with darker limbs; C) individual of *M. expectata* from Antambonooa with a contrasting reddish lower back. This is a rather common coloration in *M. expectata* populations from the northern section of the Isalo National Park (CROTTINI et al. 2008) (B–C photos not reliably referable to certain vouchers, but confirmed by molecular means as *M. expectata* following CROTTINI et al. 2008); D–E) individuals from Sakavato representing the *M. sp. aff. expectata* “South” mitochondrial lineage (already known from Tranomaro and Tsingy de Bemaraha) (D: MRSN A5222; E: MRSN A5230), note the typical *M. expectata* coloration of MRSN A5222 and the intermediate (*M. expectata*/*M. betsileo*) chromatic traits of MRSN A5230; F) individual of *M. betsileo* from Andriamanero with a typical *M. betsileo* colour pattern (photo not reliably referable to a certain voucher but confirmed by molecular means as *M. betsileo*); G–H) individuals of *M. betsileo* from Sakavato (G: FAZC 12825) and Karofoty (H: photo not reliably referable to a certain voucher but confirmed by molecular means as *M. betsileo* following GONÇALVES et al. (in prep.)) exhibiting different levels of intermediate (*M. expectata* and *M. betsileo*) chromatic traits; I) ventral side of *M. betsileo* (FAZC 12825 as in G); and J) ventral side of *M. expectata* (MRSN A5204 as in A) exhibiting no distinctive differences in the ventral colour pattern.

tance (16S) to populations of both Kirindy and Isalo and ca. 4% from populations from Berenty and Tolagnaro.

Mantella sp. aff. *expectata* “South” (DCL)
Figs 3D–E

Locality record: Sakavato (MRSN A5222, MRSN A5230).

This lineage was already known from Tranomaro (in the southeast) and Bemaraha in the west (RABEMANAJARA et al. 2008). The 16S sequences of these two individuals (EF674846–EF674847) were already published (CROTTINI et al. 2008) and released in GenBank as *M. cf. expectata/betsileo*. Interestingly, individual MRSN A5222 was unequivocally identified as *M. expectata* (see Fig. 3D) due to its coloration, whereas MRSN A5230 exhibits chromatic traits that are intermediate between *M. expectata* and *M. betsileo* (see Fig. 3E). These samples have a genetic distance (16S) of ca 1% to both *M. betsileo* and *M. expectata*, whose interspecific genetic distance is only about 1.7% (see Supplementary Table S7 for more details). In general, *Mantella* species are genetically poorly differentiated from each other (SCHAEFER et al. 2002), and the taxonomy of *Mantella* sp. aff. *expectata* “South” remains difficult, although preliminary phylogeographic analyses suggest it might deserve to be treated as an independent taxon (unpublished data). It is likely that a more robust study of the validity of this lineage will be possible only when more samples are available.

Reptiles

PELOMEDUSIDAE

Pelomedusa subrufa (LACÉPÈDE, 1788)
Fig. 5A

Locality records: Ambatovaky (MRSN R3207), Zahavola (MRSN R3737), Isalo (no precise locality).

This species is the most common freshwater turtle in Madagascar and widely distributed from the Sahamaliza Peninsula in the northwest (PENNY et al. 2017) to all of the southwest. *Pelomedusa subrufa* is generally found in stagnant water bodies and temporary ponds. In Isalo, it was found in Ambatovaky and Zahavola in slow-flowing streams, and it was previously reported from here by HAWKINS (1994).

CROCODYLIDAE

Crocodylus niloticus LAURENTI, 1768
Fig. 5B

Locality records: Ranohira (FAZC 14784/PBZT-uncatalogued).

The species was found in the Ianakandrarezo River (visual record by PEB and GMR) and the Hazofotsy River in Ranohira. Subfossil remains of *C. niloticus* are known from Ampoza (GOODMAN & JUNGERS 2014). This site is close to the Analavelona Massif, which is located at a short distance (around 30 km) west of Isalo. However, with the recent recognition of the genus *Voay* BROCHU, 2007 the identification of these remains requires reassessment (BROCHU 2007, GOODMAN & JUNGERS 2014). The two records from the Ranohira area represent the first reliable report of the Nile crocodile from the Isalo Massif. Although occasionally hunted by local people, the species seems to be so secretive in the area that there is no information about the behaviour and ecology of the Isalo populations.

CHAMAELEONIDAE

Brookesia brygooi RAXWORTHY & NUSSBAUM, 1995
Fig. 6A

Locality records: Namazaha Valley (MRSN R2546), Isalo (no precise locality).

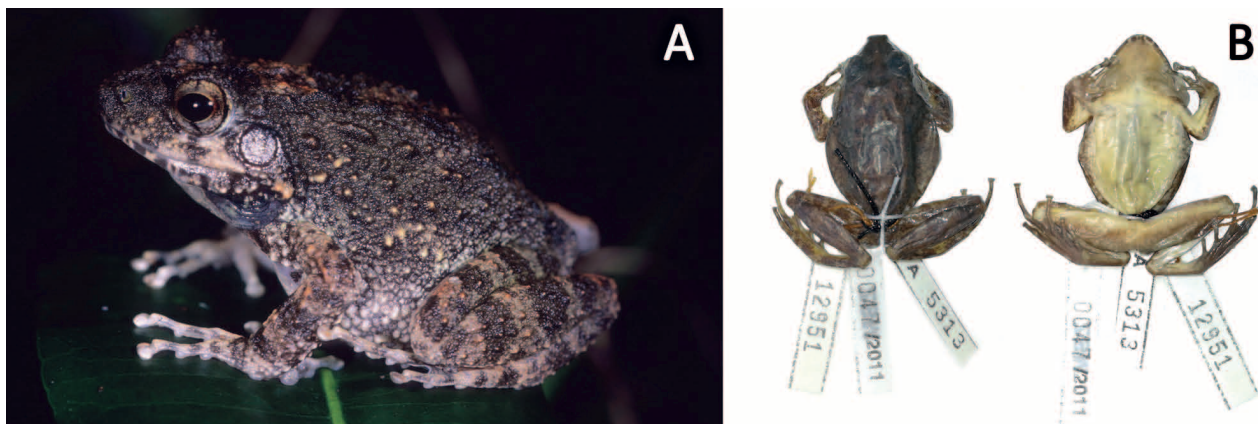


Figure 4. *Gephyromantis azzurrae* (MERCURIO & ANDREONE, 2007), paratype MRSN A5313 (A, lateral view in life; B, dorsal and ventral views in preservative), here genetically and morphologically reassigned to *G. corvus*.

Brookesia brygooi belongs to the *Brookesia decaryi* group, which includes aridity-adapted species that live within the dry-deciduous forests on the western slope of the central high plateau of the island (TOWNSEND et al. 2009). It inhabits the leaf litter stratum during the day and is found resting on low vegetation during the night. It can be quite difficult to observe in Isalo, although there is a record of one event in which several individuals were sampled in one single night (S. NDRIANTSOA, pers. comm.) This population is genetically similar to the populations of *B. brygooi* at Kirindy with which it shares a distinctive lineage when compared to *B. brygooi* from Ankarafantsika (CROTTINI et al. 2012).

Furcifer major (BRYGOO, 1971)
Fig. 6B

Locality records: Andranomena (MRSN R2891), Malaso, Ranohira (MRSN R1185), Zahavola (MRSN R2474–R2475), Isalo (no precise locality).

This taxon was formerly considered a large-sized subspecies of *Furcifer lateralis* (BRYGOO 1971, KLAVER & BÖHME 1997) inhabiting the arid southwestern region of Madagascar. A recent taxonomic revision of the carpet chameleons elevated this taxon to species rank (FLORIO et al. 2012), which therefore also applies to specimens from Isalo. HAWKINS (1994) reported this taxon as *F. lateralis*. The samples analysed in this study are genetically similar to each other (Supplementary Table S8) whereas they have a genetic distance at the COI fragment of ca. 2–3% to *F. viridis*, 3–4% to *F. lateralis*, and ca. 9–10% to sympatric *F. oustaleti* and *F. verrucosus*.

Furcifer oustaleti (MOCQUARD, 1894)
Fig. 6C

Locality records: Analalava Forest, Bereketa (MRSN R3161), Namazaha Valley.

A recent phylogeographic analysis revealed the presence of two distinctive lineages within this species, but refrained from formally describing a new species based on the still-limited evidence available (FLORIO & RAXWORTHY 2016). The populations from Isalo belong to *F. oustaleti* Clade D sensu FLORIO & RAXWORTHY (2016) and are genetically uniform for the marker analysed (Supplementary Table S8).

Furcifer verrucosus (CUVIER, 1829)
Fig. 6D

Locality records: Piscine Naturelle, Sakavato (MRSN R3165, MRSN R3169).

Slightly smaller, *F. verrucosus* can be easily mistaken for *F. oustaleti* that also widely overlaps its distribution range. *Furcifer verrucosus* is quite common in disturbed areas although it can be found in a wide array of habitats at sites below 120 m a.s.l. (RASELIMANANA & RAKOTOMALALA 2003). A recent phylogeographic analysis revealed the presence of a western and an eastern lineage (FLORIO & RAXWORTHY 2016). The Isalo population belongs to *F. verrucosus* Clade B sensu FLORIO & RAXWORTHY (2016), and the analysed samples from Isalo exhibit a genetic variability of 0.7% (COI; Supplementary Table S8) between each other.

GEKKONIDAE

Blaesodactylus sakalava (GRANDIDIER, 1867)

Locality record: Isalo (no precise locality).

In Isalo, this species is currently known from a record made by HAWKINS (1994) and a photographic record available online (flickrriver, Pete Read: <http://www.flickrriver.com/photos/128237756@No6/19835428834/>). No molecular data from Isalo are available for this species.

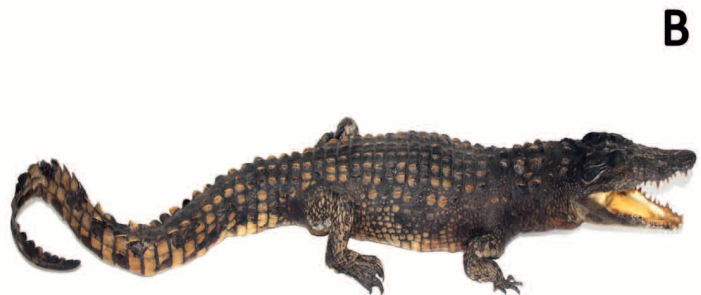


Figure 5. Chelonian and a crocodylian species found in the Isalo Massif: A) *Pelomedusa subrufa* (MRSN R3737) from Zahavola in life; B) *Crocodylus niloticus* preserved specimen (FAZC 14784, PBZT-uncatalogued) from Hazofotsy River (Ranohira).



Figure 6. Photos in life of lizard species recorded in the Isalo Massif: A) *Brookesia brygooi* (ACZC 2569) from the Namazaha Valley; B) *Furcifer major* (FAZC 14640) from Zahavola; C) *Furcifer oustaleti* from the Analalava Forest (photo not reliably referable to a certain sample or voucher); D) *F. verrucosus* from circuit Piscine Naturelle (photo not reliably referable to a certain sample or voucher); E) *Hemidactylus mercatorius* (ACZC 1945) from the Namazaha Valley; F) *Lygodactylus* sp. aff. *tuberosus* Ca02 (MRSN R3746) from Malaso; G) *Paroedura picta* from Zahavola (photo not reliably referable to a certain sample or voucher); H) *P. sp. aff. bastardi* Ca01 (FAZC 14631) from Zahavola; I) *P. sp. aff. bastardi* Ca03 from Ilakaka (photo not reliably referable to a certain sample or voucher); J) *P. vahiny* (MRSN R3748) from Malaso; K) *Phelsuma hielscheri* from Piscine Naturelle (tissue sample not collected); L) *P. mutabilis* from the Namazaha Valley (tissue sample not collected); M) *P. standingi* from Isalo (no precise locality, tissue sample not collected); N) *Zonosaurus laticaudatus* (MRSN R3213) from the Canyon des Makis; O) *Z. madagascariensis* from Andranonombilahy (tissue sample not collected); P) *Z. karsteni* from Analamangabe (tissue sample not collected); Q) *Chalarodon madagascariensis* from Ilakaka (tissue sample not collected); R) *Oplurus cyclurus* from Malaso (photo not reliably referable to a certain sample or voucher); S) *O. quadrimaculatus* from Zahavola (tissue sample not collected); T) *O. saxicola* (photo not reliably referable to a certain sample or voucher) from Sakavato; U) *Trachylepis elegans* from the Namazaha Valley (tissue sample not collected); V) *T. gravenhorstii* from the Namazaha Valley (tissue sample not collected); W) *T. nancykoutuae* from the Namazaha Valley (tissue sample not collected).

Geckolepis typica GRANDIDIER, 1867

Locality records: Analalava Forest (UMMZ 215902–215903), Beraketa (UMMZ 196575–196576), Sahanafa (UMMZ 215904), Isalo (no precise locality).

The only specimens of this taxon from Isalo are those in the collection of the Museum of Zoology at the University of Michigan, but another record stems from HAWKINS (1994). As for Isalo, neither molecular data nor information on its habitat are available.

Hemidactylus mercatorius GRAY, 1842

Fig. 6E

Locality records: Analalava Forest (MRSN R2406–R2407), Malaso (FAZC 14650/PBZT-uncatalogued), Namazaha Valley, Ranohira – Motel d'Isalo, Zahavola.

Hemidactylus mercatorius is one of the few non-endemic squamate species of Madagascar, being present and widely distributed in eastern Africa and on many islands of the Indian Ocean. Samples from Isalo belong to the most common haplotype, occurring both in the Sambirano and northeast, and in the southern central east, southeast, and south (VENCES et al. 2004). The analysed populations exhibit an average genetic variation of 2.6% (COI; Supplementary Table S8) between each other.

Lygodactylus sp. aff. *pictus* Cao1 “Isalo” (UCS)

Locality record: Analalava Forest (ZMA 19595).

This genus still contains several undescribed species (PUENTE et al. 2005, 2009, MEZZASALMA et al. 2017). Isalo lineage was previously reported by PUENTE et al. (2005) and refers to a specimen deposited in the collection of the Naturalis Biodiversity Center, Leiden (The Netherlands), collected in the Analalava Forest. It is morphologically similar to *L. pictus* although genetically it has at least 11% genetic distance to all other species of *Lygodactylus* for which 16S sequences are currently available. For comparison, we sequenced the 16S and COI fragment (16S: MHO63336–MHO63337, COI: MHO63413–MHO63414) of a recently collected specimen of *Lygodactylus* sp. aff. *pictus* Cao1 “Isalo” from the Forêt de Vohibasia (Zombitse-Vohibasia National Park) and the 16S fragment proved to be 99% identical to the 16S sequence of this taxon collected in the Analalava Forest (PUENTE et al. 2005; AY653238). This lineage is therefore currently known at least from Isalo and Zombitse-Vohibasia.

Lygodactylus sp. aff. *tuberosus* Cao2 “Isalo” (UCS)

Fig. 6F

Locality records: Malaso (MRSN R3746, MRSN R3749).

This candidate species is identified here for the first time. The analysed specimens were found in open grassland near canyon entrances and rocky microhabitat in Malaso. Morphologically, this taxon is quite similar to *L. tuberosus*. The genetic distance between *L. tuberosus* from Ifaty (MHO63412) and this UCS from Isalo is ca. 17% at the analysed COI fragment. The genetic distance in 16S between this UCS and the syntopic *Lygodactylus* sp. aff. *pictus* Cao1 “Isalo” is ca. 10%, and ca. 12% to *L. tuberosus* from Toliara and Belalanda (e.g., AY653283, LN998658).

Paroedura picta (PETERS, 1854)

Fig. 6G

Locality records: Andriamanero, Zahavola.

This ground-dwelling species is typically found in the dry forests or on sandy substrate in spiny scrub vegetation of southern Madagascar (GLAW & VENCES 2007). Within Isalo, it is commonly encountered in open savannah areas. The samples here analysed have a genetic distance of 6% (16S) to *P. picta* from Kirindy (AF215242) and only 1% to *P. picta* from the Toliara region (GU128988). In the COI fragment analysed, the samples of *P. picta* from Isalo have ca. 2–3% genetic distance to other *P. picta* from the pet trade or from Isalo (KR149293 and JQ909507, respectively), and ca. 13% distance to another *P. picta* from the pet trade (KMO47640), therefore suggesting the existence of a complex of cryptic species.

Paroedura sp. aff. *bastardi* Cao1 “Marofandilia/Miandrivazo” (UCS)

Fig. 6H; Fig. 8A

Locality records: Zahavola (MRSN R3736, MRSN R3745), Namazaha Valley (MRSN R2448), Isalo (no precise locality, MRSN R2553).

This UCS belongs to the *P. bastardi* species complex, which contains several undescribed species (JACKMAN et al. 2008, APREA et al. 2013, GLAW et al. 2014). It was found within the canyon in a shaded area close to the stream where humidity was high due to the presence of a small cave and a waterfall. It was already known from Marofandilia and Miandrivazo, and the 16S genetic distance of the Isalo population to these other populations is 3–4% (GU129005, Marofandilia and GU128989, Miandrivazo; APREA et al. 2013).

Paroedura sp. aff. *bastardi* Cao2 “Isalo” (UCS)

Locality records: Malaso, Zahavola (FAZC 14657/PBZT-uncatalogued).

This UCS also belongs to the *P. bastardi* species complex. In 2009, individuals belonging to this taxon were found on

rocky surfaces in open areas in close proximity to the Museum of the Isalo National Park, next to the Zahavola canyons (where also *Paroedura* sp. aff. *bastardi* Cao1 “Marofandilia/Miandrivazo” occurs). Other individuals were observed in open grassland near canyon entrances and in rocky microhabitats. These two sympatric lineages (*Paroedura* sp. aff. *bastardi* Cao1 “Marofandilia/Miandrivazo” and *Paroedura* sp. aff. *bastardi* Cao2 “Isalo”) are genetically highly differentiated, with values of uncorrected p-distance at the analysed COI and 16S gene fragment of ca. 26% (Supplementary Table S8) and 16%, respectively. The analysed samples from Isalo are 17% divergent (COI) from *P. bastardi* from Toliara (JQ909501, NAGY et al. 2012).

Paroedura sp. aff. *bastardi* Cao3 “Ilakaka” (UCS)
Fig. 6I; Fig. 8B

Locality records: Ilakaka, Kazofoty (MRSN R3205), Isalo (no precise locality, MRSN R2529, MRSN R2568–R2569).

This is another UCS of the *P. bastardi* species complex that is present in Isalo. This taxon was already known from previous work (APREA et al. 2013), and the genetic distance of this taxon to co-occurring species of the *P. bastardi* complex is 24% (COI, Supplementary Table S8), 18% (16S) to *P. sp. aff. bastardi* Cao1 “Marofandilia/Miandrivazo”, and 23% (COI, Supplementary Table S8) and 8% (16S) to *P. sp. aff. bastardi* Cao2 “Isalo”, respectively. The genetic distance between different samples of this species is ca. 1% at the analysed 16S fragment (GU129002, APREA et al. 2013).

Paroedura vahiny NUSSBAUM & RAXWORTHY, 2000
Fig. 6J

Locality records: Andohasahenina (MRSN R2400), Malaso (MRSN R3748), Isalo (no precise locality).

This poorly known species is characterized by having an overall smooth appearance, determined by the presence of small and scattered tubercles and numerous small scales on both body and head. In his report, HAWKINS (1994) listed *P. androyensis* for Isalo National Park, but this record almost certainly refers to *P. vahiny*, described few years after. In contrast to the holotype of *P. vahiny* that was found on a plant stem in the forest, this taxon was found on the ground in Isalo. It exhibits a genetic distance, at the COI fragment, of ca. 20–24% to all the other syntopic species of *Paroedura* (Supplementary Table S8), but the lack of a COI reference for *P. vahiny* from the type locality prevents us from assigning these samples on a molecular basis. Similarly, for the 16S gene fragment, samples of this taxon are 81–82% similar to almost all other *Paroedura* species. We here refer to this taxon as *P. vahiny* pending confirmation by means of a molecular comparison with samples from the type locality.

Phelsuma hielscheri RÖSLER, OBST & SEIPP, 2001
Fig. 6K

Locality records: Ranohira, close to Isalo Ranch (NAGY et al. 2012), Petit Nazareth Valley, Piscine Naturelle, Zahavola (MRSN R2486), Isalo (no precise locality, ZSM 192/2004).

Phelsuma hielscheri is a regionally endemic species of southwestern Madagascar that belongs to the *P. dubia* group (ROCHA et al. 2010). This species is so far known only from a few localities such as Kirindy, Morondava and Isalo. *Phelsuma hielscheri* is quite difficult to observe in Isalo. Individuals were spotted during the daytime, active on *Pandanus* sp. leaves. Only one specimen was collected in Zahavola from which no DNA sequences could be obtained.

Phelsuma mutabilis (GRANDIDIER, 1869)
Fig. 6L

Locality record: Namazaha Valley.

Phelsuma mutabilis is distributed along the southern and western Madagascar coasts up to Ankarafantsika in the northwest (CROTTINI et al. 2011). This species is known to inhabit both natural and anthropogenic areas and is adapted to live in different environments such as trees and bushes, but also on walls of buildings or fences. In Isalo, a representative of this species was observed in the Namazaha Valley where it was basking on a tree trunk (A. CROTTINI, pers. obs.). *P. mutabilis* exhibits an intraspecific genetic distance of 4.7% across Madagascar (16S, CROTTINI et al. 2011). The analysed sample from Isalo confirms this intraspecific variability by showing a genetic distance ranging from 4 to 6% (16S) to other populations (Toliara, Ejeda, Antsalova: ROCHA et al. 2009; Beroroha, Makay: SOUND et al. 2006).

Phelsuma standingi METHUEN & HEWITT, 1913
Fig. 6M

Locality records: Ranohira, Isalo (no precise locality).

Phelsuma standingi is a large species of day gecko with unresolved phylogenetic relationships (ROCHA et al. 2009, 2010), occurring only in the arid spiny forest of southwestern Madagascar. Like *P. hielscheri*, *P. standingi* is quite rarely seen in Isalo and our surveys did not yield any new observations. The only DNA sequence available for this species from Isalo refers to a published record (16S, DQ270564), of which the locality is given as “South of Isalo” (SOUND et al. 2006). Therefore, it might actually refer to the Zombitse-Vohibasia National Park, where we have observed the species as well, rather than to the Isalo Massif itself. On the other hand, BERGHOF & KRAUSE (1999) reported on a visual record of this species in Ranohira.

GERRHOSAURIDAE

Tracheloptychus madagascariensis PETERS, 1854

Locality record: Isalo (no precise locality).

This taxon was reported from Isalo by BRYGOO (1985). No molecular data from Isalo are available for this species.

Zonosaurus laticaudatus (GRANDIDIER, 1869)
Fig. 6N

Locality records: Canyon des Makis (MRSN R3213), Isalo (no precise locality, FAZC 12767/MRSN R-uncatalogued).

In Isalo, this species was observed in forested habitat inside large canyons. The analysed samples suggest a close relationship to *Z. laticaudatus* from Tsingy de Bemaraha (6% for COI, JQ909628; 2% for the 16S fragment, KC515139; NAGY et al. 2012, RECKNAGEL et al. 2013).

Zonosaurus madagascariensis (GRAY, 1831)
Fig. 6O

Locality records: Andranonombilahy, Canyon des Makis (MRSN R3232).

In contrast to *Z. laticaudatus*, this species is mostly distributed in eastern Madagascar, although there are areas in the northwest where these two species co-occur. In Isalo, the observed individuals were found in forested habitat inside or at the entrance of large canyons. The samples analysed show a genetic distance of ca. 1–2% to *Z. madagascariensis* from Tsingy de Bemaraha (JQ909629) and Mahasoia (JQ909630), and 7% to specimens from Marojejy (JQ909631) (COI). At the 16S fragment, *Z. madagascariensis* from Isalo is only 1% divergent from the closest sister species, *Z. haraldmeieri* from the Forêt d'Ambre (KC515136). More in-depth analyses will be needed to assess whether *Z. haraldmeieri* is a chromatic variant of *Z. madagascariensis* or represents a true sister species of recent divergence (RASELIMANANA et al. 2009, RECKNAGEL et al. 2013, BLAIR et al. 2015).

Zonosaurus ornatus (GRAY, 1831)

Locality record: Isalo (no precise locality).

Not encountered during our surveys, but reported from Isalo by GLAW & VENCES (1994), where it was observed in open grasslands. This visual record is in need of confirmation, as no photographic or physical voucher is available, and we cannot exclude this record was mistaken, e.g., by confusion with an unusually colored *Z. karsteni*. No molecular data from Isalo specimens are available.

Zonosaurus karsteni (GRANDIDIER, 1869)
Fig. 6P

Locality records: Analamangabe, Ranohira Bas (FAZC 14698/PBZT-uncatalogued), Isalo (no precise locality).

This species is currently known from fragmented and geographically distant populations in the dry forests of western Madagascar (GLAW & VENCES 2007). It has been reported before in Isalo by HAWKINS (1994). The populations of this species analysed so far (Andranomanintsy, Bemaraha, Kirindy, Lambokely, Mikea, and Petriky) show a low phylogeographic substructure (RASELIMANANA et al. 2009). *Zonosaurus karsteni* from Isalo confirms this pattern in that the samples had a genetic distance at both the 16S and ND1 fragment of only 1% to *Z. karsteni* from Toliara and Mikea-Ankindranoka.

OPLURIDAE

Chalarodon madagascariensis PETERS, 1854
Fig. 6Q

Locality records: Analalava Forest (UADBA 21059), Antambonoa (MRSN R3127–R3128), Canyon des Makis (MRSN R3219), Ilakaka, Vohimaro (FAZC 12792/PBZT-uncatalogued), Isalo (no precise locality, MRSN R2446–R2447).

This species was encountered in Vohimaro after it was already reported from this area (HAWKINS 1994) and Analalava (MÜNCHENBERG et al. 2008). Samples from Isalo are genetically more similar to the population of *C. madagascariensis* at Tsingy de Bemaraha (in the central west) than to the geographically closer populations at Toliara (MÜNCHENBERG et al. 2008).

Oplurus cyclurus (MERREM, 1820)
Fig. 6R

Locality records: Analalava Forest (ZSM 939/2003-940/2003), Andohasahenina (MRSN R2454), Andranomena (MRSN R2377), Antambonoa (MRSN R3238), Malaso (MRSN R3239).

Oplurus cyclurus is a largely arboreal and rupicolous iguana that can be easily identified by its tail bearing enlarged spines (GLAW & VENCES 2007). In Isalo, specifically in the Analalava Forest, two mitochondrial lineages with a genetic distance of up to 2% at the 16S occur (MÜNCHENBERG et al. 2008). Our samples of *O. cyclurus* from Malaso are genetically uniform for both 16S and COI (Supplementary Table S8) and equally divergent (1%; 16S) from the samples of both clades present in the Analalava forest.

Oplurus grandidieri MOCQUARD, 1900

Locality record: Antoha (MRSN R3215).

Only one specimen of *Oplurus grandidieri* was collected in Isalo. It exhibits a genetic distance of 2% to a sample of *O. grandidieri* from Zazafotsy (16S; EU099755; MÜNCHENBERG et al. 2008) and of 3% from two samples from the pet trade (COI, KM882905 – ALTMANOVA et al. 2015; AB218720, OKAJIMA & KUMAZAWA 2009). *Oplurus grandidieri* has a low genetic divergence compared to *O. fierinensis* (16S, c-mos uncorrected p-distances = 2.0 and 0%, respectively; MÜNCHENBERG et al. 2008), and according to our data, the COI distance is 6% compared to a specimen of *O. fierinensis* from Toliara; JQ909485, NAGY et al. 2012).

Oplurus quadrimaculatus DUMÉRIL, 1851
Fig. 6S

Locality records: Andohasahenina (MRSN R2380), Andranomena (MRSN R2378–R2379, MRSN R2381), Malaso (FAZC 14687/MRSN R-uncatalogued), Zahavola, Isalo (no precise locality, MRSN R3265–R3270).

This species lives throughout the arid areas of southwestern Madagascar, but can also be found in the drier parts of the central highlands up to 2050 m a.s.l. (Ibity Massif) and near the rainforest in the Tolagnaro area (GLAW & VENCES 2007). In Isalo, it was collected in Malaso, but, in contrast to *O. cyclurus*, which is more arboreal, this relatively common species was found on large rocks and boulders. This taxon was previously recorded by HAWKINS (1994). *Oplurus quadrimaculatus* is known to comprise two well-supported clades, one representing the highland populations (Antoetra, Ambositra, and Andringitra) and the other one the southern populations (Toliara, Tolagnaro, Andohahele, and Tranoroa) (MÜNCHENBERG et al. 2008). The population at Isalo belongs to the southern lineage, and one sample exhibits a 1% distance (16S) to this southern lineage. At the COI fragment, *O. quadrimaculatus* from Isalo shows a genetic distance of 3% to samples from Andohahele (JQ909486).

Oplurus saxicola GRANDIDIER, 1869
Fig. 6T

Locality record: Sakavato (MRSN R3243–R3246).

This rock-dwelling species inhabits the south of Madagascar (GLAW & VENCES 2007). In Isalo, it has been found only in Sakavato, and this represents the first observation in this area. Like *O. quadrimaculatus*, *O. saxicola* appears to comprise two divergent lineages: one in the southeast (Esomony, from where a new species of *Chalarodon* has recently been described; MIRALLES et al. 2015), and one in the southwest (MÜNCHENBERG et al. 2008). The sample from Isalo belongs to this last clade from the southwest, from which it differs by 2% (16S), while it exhibits 3% (16S) divergence from the sample from Esomony.

SCINCIDAE

Grandidierina rubrocaudata (GRANDIDIER, 1869)

Locality record: Isalo (no precise locality).

This taxon was reported for Isalo by BRYGOO (1981). No molecular data are available for this species from Isalo.

Trachylepis elegans (PETERS, 1854)
Fig. 6U

Locality records: Andohaosy (MRSN R3224), Andranombilahy (MRSN R2393), Andranomena (MRSN R2437), Malaso, Namazaha Valley, Ranohira – Motel d'Isalo, Vallee du Petit Nazareth (MRSN R2455), Zahavola (MRSN R3123), Isalo (no precise locality, MRSN R1173).

In Isalo, individuals were found in open grasslands and outside canyons, where they are often seen basking on rocky surfaces. A recent preliminary analysis of the genetic variability of this species across its distribution identified four genetic lineages (VENCES et al. 2014). The analysed samples from Isalo are genetically homogeneous (0.3%, Supplementary Table S8) and belong to the most widespread lineage of *T. elegans*, the lineage A sensu VENCES et al. (2014).

Trachylepis gravenhorstii (DUMÉRIL & BIBRON, 1839)
Fig. 6V

Locality records: Sambalahy (Andriamanero), Malaso (FAZC 14635/MRSN R-uncatalogued), Namazaha Valley (MRSN R2449), Ranohira, Zahavola (MRSN R3221).

Trachylepis gravenhorstii can be found both in the ground and on tree trunks (GLAW & VENCES 2007), but not inside canyons. In Isalo, we identified two different lineages, 4a and 4b (sensu VENCES et al. 2014), with a genetic distance at the analysed COI fragment of 7% between each other. As far as we know, lineage 4a was found only in the village of Ranohira, whereas lineage 4b occurred in at least two localities within the Massif (Malaso and Zahavola). Molecular taxonomic identification of the individuals collected in the Namazaha Valley was not possible. The presence of *T. gravenhorstii* lineage 4b in Isalo represents a new distributional record for this lineage.

Trachylepis nancycoutuae (NUSSBAUM & RAXWORTHY, 1998)
Fig. 6W

Locality records: Andranomena (MRSN R2401), Bememara (MRSN R2430), Bevato, Malaso (FAZC 14632/PBZT-uncatalogued), Namazaha Valley, Zahavola (MRSN R3212).

This skink species is endemic to Isalo, inhabiting rocky substrates including large boulders and cliff faces. *Trachy-*

lepis nancycoutuae was found in the southeastern sections of the Isalo NP and in the centre of the massif. This is a very secretive species, which seems to be quite abundant in this region, although it is difficult to collect. A phylogenetic analysis of the genus *Trachylepis* has recently been published and revealed that *T. nancycoutuae* probably is the sister species of an undescribed species inhabiting the area of Ambalavao and Andringitra (*T. sp. aff. vato*; LIMA et al. 2013).

LAMPROPHIIDAE

Dromicodryas bernieri (DUMÉRIL, BIBRON & DUMÉRIL, 1854)
Fig. 7A

Locality records: Andranomangara, Andranomena (MRSN R2367), Lola (MRSN R3227), Malaso, Ranohira (MRSN R2366), Zahavola (MRSN R3226).

In Malaso, this snake species was encountered during the day on rocks close to a shallow canyon, while in Ranohira, it was found along a secondary road. The analysed samples from Isalo are genetically similar to each other (0.5% Supplementary Table S8) and to the individuals from Ifaty (1%, JQ909361, NAGY et al. 2012).

Ithyocyphus oursi DOMERGUE, 1986
Fig. 7B

Locality records: Canyon des Makis (MRSN R3120), Anjofo, Namazaha Valley.

In Isalo, this species has been spotted in the Canyon des Makis, Anjofo, and in the Namazaha Valley where one adult individual was observed preying on an adult *Furcifer oustaleti* (CROTTINI et al. 2010). Irrespective of their morphological differences, *I. oursi* and *I. miniatus* are genetically poorly differentiated (ca. 3%, COI). The two analysed samples from Isalo are identical to each other in COI, but have a ca. 5% genetic divergence from a sample from Ranobe (JQ909394; NAGY et al. 2012).

Madagascarophis meridionalis (DOMERGUE, 1987)
Fig. 7C

Locality records: Andranomena (MRSN R2336), Canyon des Rats (MRSN R3741), Malaso (FAZC 14658/PBZT-uncatalogued, FAZC 14788/PBZT-uncatalogued), Namazaha Valley (MRSN R2334, MRSN R2339), Ranohira: Motel d'Isalo, Reine de l'Isalo (MRSN R3183), Tsitorina (FAZC 12807/MRSN R-uncatalogued), Vallée du Petit Nazareth (MRSN R2335), Zahavola (MRSN R3182), Isalo (no precise locality).

In Isalo, we found two distinct genetic lineages of this species with ca. 7% of genetic divergence at the analysed COI gene fragment. The sample from the Canyon des Rats be-

longs to a lineage also present in Ifaty, Ihosy and Ranomafana (NAGY et al. 2012, RUANE et al. 2016), whereas samples from Malaso, Ranohira and Zahavola belong to a lineage found also in Tsimanampetsotsa (NAGY et al. 2012). Both lineages are present in the Namazaha Valley. HAWKINS (1994) recorded this taxon as *M. colubrinus*.

Mimophis mahfalensis (GRANDIDIER, 1867)
Fig. 7D

Locality records: Andranomena (MRSN R2363), Bemenara (MRSN R2351, MRSN R2352), Bereketa (MRSN R3176), Malaso, Ranohira, Zahavola.

Mimophis mahfalensis and the recently described *M. occultus* RUANE, MYERS, LO, YUEN, WELT, JUMAN, FUTTERMAN, NUSSBAUM, SCHNEIDER, BURBRINK & RAXWORTHY, 2017 are the only Malagasy snakes that belong to the mostly African subfamily Psammophiinae (RUANE et al. 2017). In Isalo, *M. mahfalensis* can be found in dry forest, thornbush savannah, and anthropogenic habitats. The analysed samples from Isalo are identical to each other in the markers studied (Supplementary Table S8) and more similar to the individuals from Ibity (1%, COI) than to those from Toliara (5%, COI). Using specimens from Isalo, ROSA et al. (2014) provided data on the internal anatomy of *M. mahfalensis* to demonstrate the presence of toxin-secreting glands, supporting the notion to classify the Psammophiinae as a subfamily of the Lamprophiidae (PYRON et al. 2013).

Leioheterodon madagascariensis DUMÉRIL & BIBRON, 1854

Locality record: Bereketa, Isalo (no precise locality).

In Isalo, this species is currently known only from a report by HAWKINS (1994) and from Bereketa (GLAW & VENCES 2007), but no molecular data are available.

Leioheterodon modestus (GÜNTHER, 1863)
Fig. 7E

Locality records: Analalava Forest (FAZC 14695/MRSN R-uncatalogued), Andozoky (MRSN R3236), Canyon des rats (MRSN R3234), Lola (MRSN R3233), Namazaha Valley, Ranohira (FAZC 14704/PBZT-uncatalogued), Ranohira – Motel d'Isalo, Isalo (no precise locality).

This is a common species in Isalo and individuals were seen in open savannas, gallery forest, and in canyons. In Isalo, individuals of *L. modestus* have been recorded to predate upon the microendemic frog *Scaphiophryne gottlebei* (ROSA et al. 2010).

Specimen FAZC 14695, collected close to the Analalava Forest, was identified by molecular means (COI) as *Leioheterodon geayi* MOCQUARD, 1905. We compared a COI sequence of this specimen with sympatric and allopatric pop-

ulations of *L. modestus* (ACZC 1965, FAZC 14668, FAZC 14704 from Isalo; FAZC 14701 from Zazafotsy, MHO63415; FAZC 14769 from Pont Bevilany, MHO63416) and found that the COI sequence of specimen FAZC 14695 had an average genetic distance of 15% to the otherwise similar COI sequences of the other 5 analysed samples of *L. modestus*. We therefore amplified the 16S fragment of this specimen and of FAZC 14704 from Isalo and those sequences were identical (99% genetic distance), suggesting a possible amplification of a nuclear COI pseudogene in the case of specimen FAZC 14695. However, these 16S sequences had a 97–98% genetic similarity to *Leioheterodon madagascariensis* (AY188061 from NAGY et al. 2003). Morphologically, these six specimens (four from Isalo and two from elsewhere, details provided above) are identical to each other and they all correspond phenotypically to *L. modestus*. We therefore think that there might be a case of mislabelling the 16S sequence AY188061 (NAGY et al. 2003) and suggest that this sequence actually refers to *L. modestus*.

Liophidium vaillantii (MOCQUARD, 1901)

Fig. 7F

Locality record: Zahavola (MRSN R3210).

This species was rarely observed in Isalo. The known individual from Zahavola has a coloration that is quite different

from those from other localities (compare pictures of *L. sp. aff. vaillantii* “South” – which corresponds to this record – on page 435 and of *L. vaillantii* on page 433 in GLAW & VENCES 2007). However, analysed samples of this species exhibit limited genetic differentiation [1–2% to *L. vaillantii* from Kirindy (DQ979995, EU394720; Cytb) and Tsimanampetsotsa (GQ913677; Cytb); 1% to *L. vaillantii* from Tsimanampetsotsa (GQ913674; 16S)]. MERCURIO et al. (2006) observed that this species is at least partly ophiophagous.

Lycodryas guentheri (BOULENGER, 1896)

Fig. 7G

Locality records: Zahavola (MRSN R3739), Isalo (no precise locality).

We have only one record of this species from Zahavola, where the individual was found in a shrub outside the canyon. As for *L. carleti*, no sequences were previously available, but with 212 ventral scales (all other *Lycodryas* species have more), 17 scales at midbody, 78 subcaudals and 8 supralabial scales, our inspection of specimen MRSN R3739 suggests this record being referable to this taxon (VENCES et al. 2004, NAGY et al. 2010). A record of *L. gaimardi* exists from the Isalo area (HAWKINS 1994), but neither morphological nor precise locality information are available, so it could represent another record for *L. guentheri*. Geneti-



Figure 7. Photos of snake species in life recorded in the Isalo Massif: A) *Dromicodryas bernieri* from Malaso (tissue sample not collected); B) *Ithycyphus oursi* (ACZC 1932) from Namazaha Valley; C) *Madagascarophis meridionalis* (FAZC 14638) from Malaso (sample not sequenced); D) *Mimophis mahfalensis* (FAZC 14677) from Malaso; E) *Leioheterodon modestus* from the Namazaha Valley (tissue sample not collected); F) *Liophidium vaillantii* (MRSN R3210) from Zahavola; G) *Lycodryas guentheri* (MRSN R3739) from Zahavola; H) *Lycodryas sp. aff. pseudogranuliceps* Ca01 “Andohahela” (MRSN R3216) from Antoha; I) *Thamnosophis lateralis* (ACZC 1928) from the Namazaha Valley; J) *Acrantophis dumerili* from Ilakaka (tissue sample not collected); K) *Sanzinia voluntany* from the Namazaha Valley (tissue sample not collected); L) *Madatyphlops arenarius* (FAZC 14696) from near Ilakaka (sample not sequenced).

cally, this taxon seems to be closely related to *L. maculatus* from the Comoros (ZSM 38/2010) and Mayotte (ZSM 42/2010 and ZSM 665/2000) from which it has between 3 and 4% genetic distance at the analysed 16S gene, respectively, and 13% at the COI fragment.

Lycodryas sp. aff. *pseudogranuliceps* Cao1 “Andohahela” (CCS)
Fig. 7H

Locality record: Antoha (MRSN R3216).

This undescribed species of *Lycodryas* is already known from Andohahela in southeastern Madagascar (NAGY et al. 2010). Considering similar geographical patterns in *Lio-phidium vaillanti* and *Mantella* sp. aff. *expectata* “South”, this affinity of Isalo to Andohahela in southeastern Madagascar is interesting to note. In Isalo, this species was found on a small shrub at night. It seems to be closely related to *L. granuliceps* and *L. pseudogranuliceps*. The genetic distance between the specimen from Isalo and Andohahela is 1 and 5% at the analysed 16S and COI gene fragments, respectively. Our sample has 2% (16S) and 9% (COI) divergences from *L. pseudogranuliceps* from Befandriana-Avaratra in northern Madagascar (ZSM 193/2009, NAGY et al. 2010, 2012).

Pseudoxyrhopus quinquelineatus (GÜNTHER, 1881)

Locality record: Isalo (no precise locality).

This species is currently known from Isalo only by the record published by HAWKINS (1994). No molecular data from Isalo are available.

Thamnosophis lateralis (DUMÉRIL, BIBRON & DUMÉRIL, 1854)
Fig. 7I

Locality records: Andriamanero (FAZC 12632/MRSN R-uncatalogued), Namazaha Valley.

The individuals found in the Namazaha Valley and Andriamanero were active on trails during the day. The analysed samples from Isalo are quite similar to each other (0.8%, Supplementary Table S8) and genetically similar (3%; COI) to the individuals from Ranomafana as published by NAGY et al. (2012).

SANZINIIDAE

Acrantophis dumerili JAN, 1860
Fig. 7J

Locality records: Andohasahenina, Ilakaka, Namazaha Valley, Isalo (no precise locality).

This species was reported from Isalo by HAWKINS (1994). We observed it in Andohasahenina and Ilakaka, but no samples were collected. This species was recently (October 2017) found in Namazaha Valley (L. BUFFA, pers. comm.), inactive on the ground during the day. Two species are currently known in this genus, but molecular analyses revealed a third lineage in the south and southeast of Madagascar (OROZCO-TER WENGEL et al. 2008). The only data for this species from Isalo available to us are those of a published record (16S, EU419793; OROZCO-TER WENGEL et al. 2008).

Sanzinia voluntary VENCES & GLAW, 2004
Fig. 7K

Locality records: Canyon de Zahavola, Namazaha Valley, Isalo (no precise locality).

This taxon was formerly considered a subspecies of *S. madagascariensis* (OROZCO-TER WENGEL et al. 2008), but has recently been elevated to species level (REYNOLDS et al. 2014). As per its new definition, this species is restricted to the west of Madagascar where it has a patchy distribution mostly limited to habitats with trees and large shrubs. In Isalo, this species was found in both primary and secondary forests. In 2009, one young individual was observed resting for several days inside the Canyon of Zahavola, where it was probably hunting. None of the newly collected samples from Isalo could be successfully sequenced; we therefore here refer to a published record (16S, EU419803 – OROZCO-TER WENGEL et al. 2008).

TYPHLOPIDAE

Madatyphlops arenarius (GRANDIDIER, 1872)
Fig. 7L

Locality records: Ilakaka (FAZC 14696/MRSN R-uncatalogued), Ranohira (MRSN R2368–R2369).

Madatyphlops arenarius is a worm-like blind snake known from the west and south of Madagascar. It inhabits sandy substrates and can be encountered under stones and, rarely, active on the ground at night. In Isalo, this species is rarely found, and only a few records are currently available (three specimens have been collected). One specimen was found crossing a dirt road at night. Unfortunately, it was not possible to amplify DNA fragments from these recently collected specimens. The only genetic data available for this species from Isalo are those of a published record (COI, JQ909606; NAGY et al. 2012, 2015). This sample has 11% genetic distance to a sample of *Madatyphlops arenarius* from Ifaty (KT316451; NAGY et al. 2012). In fact, a recent publication indicated that *M. arenarius* might be a complex of at least four different taxa and a systematic revision of this group is therefore needed to assign this name to one or more populations (NAGY et al. 2015).

Discussion

In this study, we generated reference sequences for almost all amphibians and reptiles known from Isalo. These include 1) 16S sequences of 20 of the 21 amphibian taxa previously listed by MERCURIO et al. (2008), and 2) 16S sequences of one new taxon (*Mantidactylus* sp. aff. *multiplicatus* Ca65 “Isalo”) and two new species distribution records (*Mantella* sp. aff. *expectata* “South” and *Scaphiophryne* sp. aff. *calcarata* Cao2); finally, we 3) provide reference sequences for 40 of the 47 reptile taxa currently known from this area. For seven taxa (*Blaesodactylus sakalava*, *Geckolepis typica*, *Grandidierina rubrocaudata*, *Leioheterodon madagascariensis*, *Pseudoxyrhopus quinquelineatus*, *Tracheloptychus madagascariensis*, and *Zonosaurus ornatus*) we only have reports from literature (BRYGOO 1981, 1985, HAWKINS 1994, GLAW & VENCES 1994), as those taxa were not observed by our team during the multiple surveys we conducted in Isalo. In his species inventory from Isalo National Park, HAWKINS (1994) reported on *Lygodactylus tolampyae* (GRANDIDIER, 1872), a taxon already known from the Zombitse-Vohibasia National Park (GLAW & VENCES 2007), and on *L. gaimardi* (SCHLEGEL, 1837). Although we could not verify the taxonomic identities of these records we think they might refer to one of the two candidate species of *Lygodactylus* reported herein, and to *L. guentheri* or *Lycodryas* sp. aff. *pseudogranuliceps* Cao1 “Andohahela”, respectively.

Overall, when comparing our results with those available from other dry deciduous forests in Madagascar, it would appear that amphibian species richness (24 species-level taxa) in the Isalo Massif is relatively high and more

diverse than at other western sites such as the Tsingy de Bemaraha National Park [19 species; BORA et al. (2010)], the Kirindy dry forest [15; GLOS (2003)], Ankarafantsika National Park, and in the Montagne des Français [9; MORI et al. (2006) and D’CRUZE et al. (2007)]. The scenario is slightly different for reptiles: compared to the 47 species-level taxa of reptiles in Isalo, there are higher numbers in Tsingy de Bemaraha (60, BORA et al. 2010), 58 in Mikea (RASELIMANANA 2008), 52 in the Montagne des Français (D’CRUZE et al. 2007), whereas Kirindy and Ankarafantsika host similar numbers (43 and 47 taxa, respectively; BLOXAM et al. 1996, RAMANAMANJATO & RABIBISOA 2002). If our results are compared with values available from the eastern rainforests, amphibian richness is obviously much lower (see for comparison GEHRING et al. 2010, ROSA et al. 2012, HEINERMANN et al. 2015), while values for reptile richness are roughly similar (ANDREONE et al. 2003, GOODMAN et al. 2003, KREMEN 2003, D’CRUZE et al. 2008, GEHRING et al. 2010).

The high level of species richness and the apparently locally endemic species observed in Isalo can be interpreted as a result of palaeoclimatic oscillations (RAXWORTHY & NUSSBAUM 1997, MERCIER & WILMÉ 2013). If this is true, the rainforest species could have expanded their ranges and survived in the massif during warm and humid periods due to habitat availability and the presence of permanent water and sub-humid forests, while dryness-adapted species remained constrained to small refugia in suitable areas (CROWLEY 2010, MERCIER & WILMÉ 2013); and the situation would have been the opposite during colder and drier periods. On the other hand, the peculiar position of the Isalo Massif and the structure of the sandstone formation that provides a number of humid (canyons) and arid habitats might have favoured the coexistence of taxa typical of multiple biomes. This heterogeneity allows for the presence of species adapted to different habitats, from the eastern humid forest, via the western dry deciduous forest to the southern subarid savannas, as is demonstrated by the presence of typical rainforest (e.g., *Boophis luteus*) and dry habitat (e.g., *Brookesia brygooi*) species. For some taxa, Isalo can also be characterized as a contact zone of different lineages such as those found in *Trachylepis gravenhorstii* and *Madagascarophis meridionalis*, and of closely related taxa such as *Scaphiophryne* sp. aff. *calcarata* Cao1 and Cao2. In the case of *T. gravenhorstii*, lineage 4a was already known from Isalo (Analalava Forest) as well as Antoetra, Ranomafana and Vohiparara (VENCES et al. 2014). These latter three localities are all located on the humid eastern side of the island. Lineage 4b is instead widespread in the subarid-subhumid southern area (Tolagnaro, Andohahela, Manantany, Tranomaro, Ambolavao and Toliara; VENCES et al. 2014). Similarly, the samples from Zahavola, Ranohira and Malaso of *M. meridionalis* correspond to the lineage known from Tsimanampetsotsa (arid environment) whereas the one from the Canyon des Rats, together with samples known from Ifaty and Toliara, belongs to the lineage present at Ihosy and Ranomafana (humid environment).

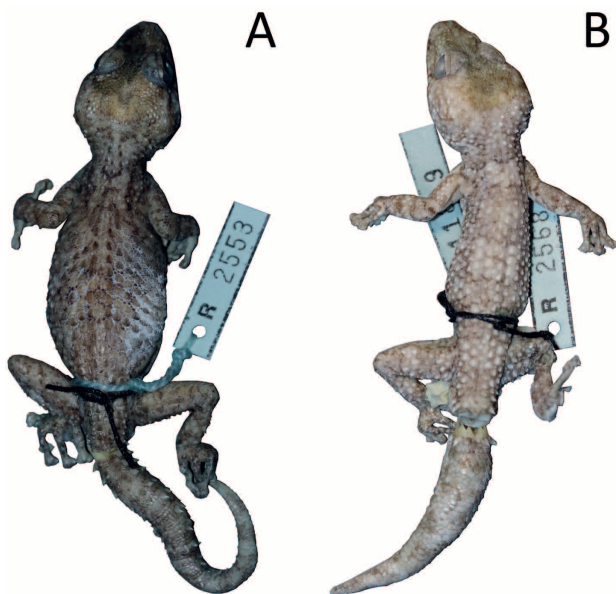


Figure 8. Photos of preserved specimens of *Paroedura* sp. aff. *bastardi* Ca01 “Marofandilia/Miandrivazo” (A; MRSN R2553) and *Paroedura* sp. aff. *bastardi* Ca03 “Ilakaka” (B; MRSN R2568).

The analysis of populations of *M. expectata* (Figs 3A–C, J), *M. sp. aff. expectata* “South” (Figs 3D–E) and *M. betsileo* (Figs 3F–I) revealed that these taxa vary greatly in their colour patterns (Fig. 3). Interestingly, in Sakavato, two morphologically similar individuals with intermediate (*M. expectata*/*M. betsileo*) chromatic traits (Figs 3E, G) were identified by their mitochondrial DNA and their analysis revealed that these individuals represent two different taxa (*M. sp. aff. expectata* “South” – Fig. 3E, and *M. betsileo* – Fig. 3G). This might be due to introgressive hybridization of the mtDNA (CHIARI et al. 2004, CROTTINI et al. 2008) or convergent evolution of the same coloration pattern promoted by living in the same environment and sharing the same selective pressures, for example predation (Müllerian mimicry; CHIARI et al. 2004).

In this study, we provide a thorough taxonomic identification of most of the Isalo specimens hosted in the MRSN collection (Supplementary Table S3 and S5) and found that only *Scaphiophryne menabensis* is not represented in its amphibian collection. As far as reptiles are concerned, *Crocodylus niloticus*, *Lygodactylus sp. aff. pictus* Cao1 “Isalo”, *Blaesodactylus sakalava*, *Geckolepis typica*, *Paroedura sp. aff. bastardi* Cao2 “Isalo”, *Phelsuma mutabilis*, *Phelsuma standingi*, *Zonosaurus karsteni*, *Zonosaurus ornatus*, *Tracheloptychus madagascariensis*, *Grandidierina rubrocaudata*, *Thamnosophis lateralis*, *Sanzinia volontany*, *Acrantophis dumerili*, *Leioheterodon madagascariensis*, and *Pseudoxyrhopus quinquelineatus* are currently not represented in the MRSN collection. The findings reported in this study highlight the crucial importance of combining extensive field research with molecular taxonomic identification and with the morphological inspection of voucher specimens hosted in museum collections (ROCHA et al. 2014). With this approach, it is possible to generate taxonomically accurate species lists and (when necessary) update previously published work to accurately assess the biodiversity value of a surveyed area. In some instances, the lack of reference sequences in public repositories did not allow a sample to be unequivocally assigned to a described or undescribed taxa. In these cases, their identification was facilitated only by the availability of corresponding museum specimens (e.g., *Paroedura vahiny* and *Lycodryas guentheri*). At the same time the availability of preserved specimens enabled us to ascertain the consistency of diagnostic characters. This was the case in our *Leioheterodon* samples that we assigned to *L. modestus* on the basis of diagnostic morphological traits when molecular data were misleading. It provided a possibility to extract new tissue samples from existing specimens that facilitated the sequencing and reassignment of one *G. azzurrae* paratype (MRSN A5313) to *G. corvus* and the investigation of other aspects of specimen morphology (e.g., coloration, as in some specimens of *Mantella* spp. analysed in our study). All this underscores the crucial role that historical and modern collections play in cataloguing world biodiversity and the importance of maintaining specimens in permanent and accessible museum collections (DE LA SANCHA et al. 2016, ANDREONE 2017).

Acknowledgements

Our thanks go to the Malagasy authorities for granting research and export permits, to the Madagascar Institut pour la Conservation des Ecosystèmes Tropicaux (MICET) for logistical help, and to the Madagascar National Parks Association (MNP) for granting us access to the Park. Particular thanks go to the colleagues, students and assistants who helped in the field: ANICET (MNP park guide), DEVIN EDMONDS, ISABELLA LAU, D. JAMES HARRIS, MARGARIDA GONÇALVES, IKER A. IRISARRI, ALEXANDRA LIMA, and SOLOHERY RASAMISON; and to HAZA. SUSANNE HAUSWALDT and MEIKE KONDERMANN who contributed to the laboratory work. ARONIAINA RAJAONARIVO helped us with logistics and assistance in Isalo. Our fieldwork was made possible with the support of the Mohamed bin Zayed Species Conservation Fund, EDGE, Zoological Society of London, Amphibian Specialist Group-Madagascar, Conservation International, Gondwana Conservation and Research, Regione Piemonte, Reptiland, and the Zurich Zoo. The Portuguese National Funds through FCT – Foundation for Science and Technology supported the PhD fellowship of WC (SFRH/BD/102495/2014) and the Investigador FCT (IF) grant to AC (IF/00209/2014). This work is funded by National Funds through FCT within the IF/00209/2014/CP1256/CT0011 Exploratory Research Project.

References

- ALTMANOVÁ, M., M. JOHNSON POKORNA, M. ROVATOS & L. KRATOCHVIL (2015): Focused on Old World iguanas: Karyotype evolution in the Madagascar family Opluridae (Squamata: Pleurodonta). – 20th International Chromosome Conference (ICC), **23**: 399–400.
- ALTSCHUL, S. F., T. L. MADDEN, A. A. SCHÄFFER, J. ZHANG, Z. ZHANG, W. MILLER & D. J. LIPMAN (1997): Gapped BLAST and PSI-BLAST: a new generation of protein database search programs. – *Nucleic Acids Research*, **25**: 3389–3402.
- ANDREONE, F. (2017): Collezioni sistematiche e ricerca: sfide e problemi per i musei naturalistici italiani. – *Museologia Scientifica*, **17**: 89–93.
- ANDREONE, F., F. GLAW, R. A. NUSSBAUM, C. J. RAXWORTHY, M. VENCES & J. E. RANDRIANIRINA (2003): The amphibians and reptiles of Nosy Be (NW Madagascar) and nearby islands: a case study of diversity and conservation of an insular fauna. – *Journal of Natural History*, **37**: 2119–2149.
- APREA, G., F. ANDREONE, D. FULGIONE, A. PETRACCIOLI & G. ODIERNA (2013): Chromosomal rearrangements occurred repeatedly and independently during species diversification in Malagasy geckos, genus *Paroedura*. – *African Zoology*, **48**: 96–108.
- ARÉVALO, E., S. K. DAVIS, & J. SITES (1994): Mitochondrial DNA sequence divergence and phylogenetic relationships among eight chromosome races of the *Sceloporus grammicus* complex (Phrynosomatidae) in Central Mexico. – *Systematic Biology*, **43**: 387–418.
- AYMOZ, B. G. P., V. R. RANDRIANJAFY, Z. J. N. RANDRIANJAFY & D. KHASA (2013): Community management of natural resources: a case study from Ankarafantsika National Park, Madagascar. – *Ambio*, **42**: 767–775.
- BERGHOF, H.–P. & P. KRAUSE (1999): Bemerkungen zum Verbreitungsgebiet von *Phelsuma standingi* Methuen & Hewitt, 1913. – *Herpetofauna*, **21** (123): 19–20.

- BLAIR, C., B. P. NOONAN, J. L. BROWN, A. P. RASELIMANANA, M. VENCES & A. D. YODER (2015): Multilocus phylogenetic and geospatial analyses illuminate diversification patterns and the biogeographic history of Malagasy endemic plated lizards (Gerrhosauridae: Zonosaurinae). – *Journal of Evolutionary Biology*, **28**: 481–492.
- BLOXAM, Q. M. C., J. L. BEHLER, E. R. RAKOTOVAO, H. J. A. R. RANDRIAMAHAZO, K. T. HAYES, S. J. TONGE & J. U. GANZHORN (1996): Effects of logging on the reptile fauna of the Kirindy forest with special emphasis on the Flat-tailed Tortoise (*Pyxis planicauda*). – pp. 189–203 in: GANZHORN, J. U. & J. P. SORG (eds): Ecology and economy of a tropical dry forest in Madagascar. – *Primate Report*, **46**: 189–201.
- BORA, P., C. J. RANDRIANANTOANDRO, R. RANDRIANAVELONA, E. F. HANTALALAINA, R. R. ANDRIANTSIMANARILAFY, D. RAKOTONDRAVONY, O. R. RAMILJAONA, M. VENCES, R. K. B. JENKINS, F. GLAW & J. KÖHLER (2010): Amphibians and reptiles of the Tsingy de Bemaraha plateau, western Madagascar: checklist, biogeography and conservation. – *Herpetological Conservation and Biology*, **5**: 111–125.
- BOSSUYT, F. & M. C. MILINKOVITCH (2000): Convergent adaptive radiations in Madagascan and Asian ranid frogs reveal covariation between larval and adult traits. – *Proceedings of the National Academy of Sciences of the USA*, **97**: 6585–6590.
- BROCHU, C. A. (2007): Morphology, relationships, and biogeographical significance of an extinct horned crocodile (Crocodylia, Crocodylidae) from the Quaternary of Madagascar. – *Zoological Journal of the Linnean Society*, **150**: 835.
- BROWN, J. L., A. CAMERON, A. D. YODER & M. VENCES (2014): A necessarily complex model to explain the biogeography of the amphibians and reptiles of Madagascar. – *Nature Communications*, **5**: e5046.
- BROWN, J. L., N. SILLERO, F. GLAW, P. BORA, D. R. VIEITES & M. VENCES (2016): Spatial biodiversity patterns of Madagascar's amphibians and reptiles. – *PLoS ONE*, **11**: e0144076.
- BRUFORD, M. W., O. HANOTTE, J. F. Y. BROOKFIELD & T. BURKE (1992): Single-locus and multilocus DNA fingerprinting. – Oxford, IRL Press.
- BRYGOO, E. R. (1971): Reptiles sauriens Chamaeleonidae: genre *Chamaeleo*. – Faune Madagascar, Orstom et CNRS, Paris, 318 pp.
- BRYGOO, E. R. (1981): Systématique des Lézards Scincides de la région malgache. VII Revision des genres *Voeltzkowia* Boettger, 1893, *Grandidierina* Mocquard, 1894, et *Cryptoscincus* Mocquard, 1894. – Mémoires du Muséum National d'Histoire Naturelle. Série 4, 3: 675–688.
- BRYGOO, E. R. (1985) : Les Gerrhosaurinae de Madagascar Sauria (Cordylidae). – Mémoires du Muséum National d'Histoire Naturelle. Série A, Zoologie, **134**: 1–65.
- BURBRINK, F. T., R. LAWSON & J. B. SLOWINSKI (2000): Mitochondrial DNA phylogeography of the polytypic North American rat snake (*Elaphe obsoleta*): a critique of the subspecies concept. – *Evolution*, **54**: 2107–2118.
- CHIARI, Y., M. VENCES, D. R. VIEITES, F. RABEMANANJARA, P. BORA, O. RAMILJAONA RAVOAHANGIMALALA & A. MEYER (2004): New evidence for parallel evolution of colour patterns in Malagasy poison frogs (*Mantella*). – *Molecular Ecology*, **13**: 3763–3774.
- CROTTINI, A., Y. CHIARI, V. MERCURIO, A. MEYER, M. VENCES & F. ANDREONE (2008): Into the canyons: the phylogeography of the Malagasy frogs *Mantella expectata* and *Scaphiophryne gottlebei* in the arid Isalo Massif, and its significance for conservation (Amphibia: Mantellidae and Microhylidae). – *Organisms Diversity & Evolution*, **8**: 368–377.
- CROTTINI, A., P. S. GEHRING, F. GLAW, D. J. HARRIS, A. LIMA & M. VENCES (2011): Deciphering the cryptic species diversity of dull-coloured day gekos *Phelsuma* (Squamata: Gekkonidae) from Madagascar, with description of a new species. – *Zootaxa*, **2982**: 40–48.
- CROTTINI, A., D. J. HARRIS, I. A. IRISARRI, A. LIMA, S. RASAMISO & G. M. ROSA (2010): Confirming Domergue: *Ithycyphus oursi* Domergue, 1986 predation upon *Furcifer oustaleti* (Mocquard, 1894). – *Herpetology Notes*, **3**: 127–131.
- CROTTINI, A., A. MIRALLES, F. GLAW, D. J. HARRIS, A. LIMA & M. VENCES (2012): Description of a new pygmy chameleon (Chamaeleonidae: *Brookesia*) from central Madagascar. – *Zootaxa*, **3490**: 63–74.
- CROWLEY, B. E. (2010): A refined chronology of prehistoric Madagascar and the demise of the megafauna. – *Quaternary Science Reviews*, **29**: 2591–2603.
- D'CRUZE, N., J. KÖHLER, M. FRANZEN & F. GLAW (2008): A conservation assessment of the amphibians and reptiles of the Forêt d'Ambre Special Reserve, North Madagascar. – *Madagascar Conservation & Development*, **3**: 44–54.
- D'CRUZE, N., J. SABEL, K. GREEN, J. DAWSON, C. GARDNER, J. ROBINSON, G. STARKIE, M. VENCES & F. GLAW (2007): The first comprehensive survey of amphibians and reptiles at Montagne des Français, Madagascar. – *Herpetological Conservation and Biology*, **2**: 87–99.
- DE LA SANCHA, N. U., S. A. BOYLE & B. D. PATTERSON (2016): Getting back to the basics: museum collections and satellite imagery are critical to analysing species diversity. – *BioScience*, **67**: 405–406.
- FLORIO, A. M., C. M. INGRAM, H. A. RAKOTONDRAVONY, E. E. LOUIS & C. J. RAXWORTHY (2012): Detecting cryptic speciation in the widespread and morphologically conservative carpet chameleon (*Furcifer lateralis*) of Madagascar. – *Journal of Evolutionary Biology*, **25**: 1399–1414.
- FLORIO, A. M. & C. J. RAXWORTHY (2016): A phylogeographic assessment of the Malagasy Giant Chameleons (*Furcifer verrucosus* and *Furcifer oustaleti*). – *PLoS ONE*, **11**: e0154144.
- FOUQUET, A., A. GILLES, M. VENCES, C. MARTY, M. BLANC & N. J. GEMMELL (2007): Underestimation of species richness in Neotropical frogs revealed by mtDNA analyses. – *PLoS ONE*, **2**: e1109.
- GANZHORN, J. U., P. P. II LOWRY, G. E. SCHATZ & S. SOMMER (2001): The biodiversity of Madagascar: one of the world's hottest hotspots on its way out. – *Oryx*, **35**: 2–5.
- GARDNER, C. J., C. J. RAXWORTHY, K. METCALFE, A. P. RASELIMANANA, R. J. SMITH & Z. G. DAVIES (2015): Comparing methods for prioritising protected areas for investment: a case study using Madagascar's dry forest reptiles. – *PLoS ONE*, **10**: e0132803.
- 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.

- GLAW, F., H. RÖSLER, I. INEICH, P.-S. GEHRING, J. KÖHLER & M. VENCES (2014): A new species of nocturnal gecko (*Paroedura*) from karstic limestone in northern Madagascar. – *Zoosystematics and Evolution*, **90**: 249–259.
- GLAW, F. & M. VENCES (1994): A field guide to the amphibians and reptiles of Madagascar. – M. Vences & F. Glaw Verlags GbR.
- GLAW, F. & M. VENCES (2007): A field guide to the amphibians and reptiles of Madagascar. – M. Vences & F. Glaw Verlags GbR.
- GLOS, J. (2003): The amphibian fauna of the Kirindy dry forest in western Madagascar. – *Salamandra*, **39**: 75–90.
- GLOS, J., F. GLAW & M. VENCES (2005): A new species of *Scaphiophryne* from western Madagascar. – *Copeia*, **2**: 252–261.
- GOODMAN, S. M. & J. P. BENSTEAD (2003): The Natural history of Madagascar. – The University of Chicago Press, 1728 pp.
- GOODMAN, S. M. & J. P. BENSTEAD (2005): Updated estimates of biotic diversity and endemism for Madagascar. – *Oryx*, **39**: 73–77.
- GOODMAN, S. M. & W. L. JUNGERS (2014): Extinct Madagascar: Picturing the Island's Past. – The University of Chicago Press, Chicago, Illinois, 296 pp.
- GOODMAN, S. M. & L. WILMÉ (2003): Nouveaux résultats d'inventaires biologiques faisant référence à l'altitude dans la région des massifs montagneux de Marojejy et d'Anjanaharibe-Sud. – Recherches pour le Développement, Série Sciences Biologiques, 302 pp.
- HAWKINS, F. (1994): Isalo faunal inventory. Final report to the Association Nationale pur la Gestion des Aires Protégées. Landell Mills Limited, City, UK. pp. 310–311 in: RAKOTONDRAVONY, H. A. & S. M. GOODMAN (eds): Rapid herpetofaunal surveys within five isolated forests on sedimentary rock in western Madagascar. – *Herpetological Conservation and Biology*, **6**: 297–311.
- HALL, T. A. (1999): BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. – *Nucleic Acids Symposium Series*, **41**: 95–98.
- HEINERMANN, J., A. RODRÍGUEZ, O. SEGEV, D. EDMONDS, R. DOLCH & M. VENCES (2015): Year-round activity patterns in a hyperdiverse community of rainforest amphibians in Madagascar. – *Journal of Natural History*, **49**: 35–36.
- JACKMAN, T. R., A. M. BAUER, E. GREENBAUM, F. GLAW & M. VENCES (2008): Molecular phylogenetic relationships among species of the Malagasy-Comoran gecko genus *Paroedura* (Squamata: Gekkonidae). – *Molecular Phylogenetics and Evolution*, **46**: 74–81.
- JENKINS, R. K. B., M. F. TOGNETTI, P. BOWLES, N. COX, J. L. BROWN, L. CHAN, F. ANDREONE, A. ANDRIAMAZAVA, R. R. ANDRIANTSIMANARILAFY, M. ANJERINIAINA, P. BORA, L. D. BRADY, E. F. HANTALALAINA, F. GLAW, R. A. GRIFFITHS, C. HILTON-TAYLOR, M. HOFFMANN, V. KATARIYA, N.H. RABISOA, J. RAFANOMEZANTSOA, D. RAKOTOMALALA, H. RAKOTONDRAVONY, N. A. RAKOTONDRAZAFY, J. RALAMBONIAIRAINY, J. B. RAMANNAMANJATO, H. RANDRIAMAHAZO, J. C. RANDRIANANTOANDRO, H. H. RANDRIANASOLO, J. E. RANDRIANIRINA, H. RANDRIANIZAHAN, A. P. RASELIMANANA, A. RASOLOHERY, F. M. RATSOAVINA, C. J. RAXWORTHY, E. RABOSMANITRANDRASANA, F. ROLLANDE, P. P. VAN DIJK, A. D. YODER & M. VENCES (2014): Extinction risks and the conservation of Madagascar's reptiles. – *PloS ONE*, **9**: e100173.
- KLAVER, C. J. J. & W. BÖHME (1997): Chamaeleonidae. Das Tierreich, 112. – Verlag Walter de Gruyter & Co, Berlin, New York, 85 pp.
- KREMEN, C. (2003): The Masoala peninsula. – pp 1459–1466 in: GOODMAN S. M. & J. P. BENSTEAD (eds): The Natural History of Madagascar. – The University of Chicago Press, Chicago and London.
- KREMEN, C., A. CAMERON, A. MOILANEN, S. J. PHILLIPS, C. D. THOMAS, H. BEENTJE, J. DRANSFIELD, B. L. FISHER, F. GLAW, T. C. GOOD, G. J. HARPER, R. J. HIJMANS, D. C. LEES, E. LOUIS, R. A. NUSSBAUM, C. J. RAXWORTHY, A. RAZAFIMPAHANANA, G. E. SCHATZ, M. VENCES, D. R. VIEITES, P. C. WRIGHT & M. L. ZJHRA (2008): Aligning conservation priorities across taxa in Madagascar with high-resolution planning tools. – *Science*, **320**: 222–226.
- KUMAR, S., G. STECHER & K. TAMURA (2016): MEGA7: Molecular Evolutionary Genetics Analysis Version 7.0 for bigger datasets. – *Molecular Biology and Evolution*, **33**: 1870–1874.
- LEACHÉ, A. D. & T. W. REEDER (2002): Molecular systematics of the eastern fence lizard (*Sceloporus undulatus*): a comparison of parsimony, likelihood, and Bayesian approaches. – *Systematic Biology*, **51**: 44–68.
- LIMA, A., D. J. HARRIS, S. ROCHA, A. MIRALLES, F. GLAW & M. VENCES (2013): Phylogenetic relationships of *Trachylepis* skink species from Madagascar and the Seychelles (Squamata: Scincidae). – *Molecular Phylogenetics and Evolution*, **67**: 615–620.
- MERCIER, J. & L. WILMÉ (2013): The Eco-Geo-Clim model: explaining Madagascar's endemism. – *Madagascar Conservation and Development*, **8**: 2–8.
- MERCURIO, V. & F. ANDREONE (2006): The tadpoles of *Scaphiophryne gottlebei* (Microhylidae: Scaphiophryninae) and *Mantella expectata* (Mantellidae: Mantellinae) from Isalo Massif, South-central Madagascar. – *Alytes*, **23**: 81–95.
- MERCURIO, V. & F. ANDREONE (2007): Two new canyon-dwelling frogs from the arid sandstone Isalo Massif, central-southern Madagascar (Mantellidae-Mantellinae). – *Zootaxa*, **1574**: 31–47.
- MERCURIO, V., G. APREA, A. CROTTINI, F. MATTIOLI, J. RANDRIANIRINA, T. RAZAFINDRABE & F. ANDREONE (2008): The amphibians of Isalo Massif, southern-central Madagascar: high frog diversity in an apparently hostile dry habitat. – *Monografie del Museo Regionale di Scienze Naturali di Torino*, 5–58.
- MERCURIO, V., F. MATTIOLI, T. J. RAZAFINDRABE & F. ANDREONE (2006): A possible attempt of predation of *Liophidium vaillanti* upon *Dromicodryas bernieri* observed in central-southern Madagascar (Serpentes: Colubridae). – *Salamandra*, **42**: 181–183.
- MEZZASALMA, M., F. ANDREONE, G. APREA, F. GLAW, G. ODIERNA & F. M. GUARINO (2017): Molecular phylogeny, biogeography and chromosome evolution of Malagasy dwarf geckos of the genus *Lygodactylus* (Squamata, Gekkonidae). – *Zoologica Scripta*, **46**: 42–54.
- MIRALLES, A., F. GLAW, F. M. RATSOAVINA & M. VENCES (2015): A likely microendemic new species of terrestrial iguana, genus *Chalarodon*, from Madagascar. – *Zootaxa*, **3946**: 201–220.
- MORI, A., I. IKEUCHI & M. HASEGAWA (2006): Herpetofauna of Ampijoroa, Ankarafantsika Strict Nature Reserve, a dry forest

- in northwestern Madagascar. – *Herpetological Natural History*, **10**: 31–60.
- MYERS, N., R. A. MITTERMEIER, C. G. MITTERMEIER, G. A. B. DA FONSECA & J. KENT (2000): Biodiversity hotspots for conservation priorities. – *Nature*, **403**: 853–858.
- MÜNCHENBERG, T., K. C. WOLLENBERG, F. GLAW & M. VENCES (2008): Molecular phylogeny and geographic variation of Malagasy iguanas (*Oplurus* and *Chalarodon*). – *Amphibia-Reptilia*, **29**: 319–327.
- NAGY, Z. T., U. JOGER, M. WINK, F. GLAW & M. VENCES (2003): Multiple colonization of Madagascar and Socotra by colubrid snakes: evidence from nuclear and mitochondrial gene phylogenies. – *Proceedings of the Royal Society, London, B*, **270**: 2613–2621.
- NAGY, Z. T., F. GLAW & M. VENCES (2010): Systematics of the snake genera *Stenophis* and *Lycodryas* from Madagascar and the Comoros. – *Zoologica Scripta*, **39**: 426–435.
- NAGY, Z. T., A. B. MARION, F. GLAW, A. MIRALLES, J. NOPPER, M. VENCES & S. B. HEDGES (2015): Molecular systematics and undescribed diversity of Madagascar scoleophidian snakes (Squamata: Serpentes). – *Zootaxa*, **4040**: 31–47.
- NAGY, Z. T., G. SONET, F. GLAW & M. VENCES (2012): First large-scale DNA barcoding assessment of reptiles in the biodiversity hotspot of Madagascar, based on newly designed COI primers. – *PLoS ONE*, **7**: e34506.
- OKAJIMA, Y. & Y. KUMAZAWA (2009): Mitogenomic perspectives into iguanid phylogeny and biogeography: Gondwanan vicariance for the origin of Madagascan oplurines. – *Gene*, **441**: 28–35.
- OROZCO-TER WENGEL, P., Z. T. NAGY, D. R. VIEITES, M. VENCES, JR. E. LOUIS (2008): Phylogeography and phylogenetic relationships of Malagasy tree and ground boas. – *Biological Journal of the Linnean Society*, **95**: 640–652.
- PALUMBI, S., A. MARTIN, S. ROMANO, W. O. McMILLAN, L. STICE & G. GRABOWSKI (1991): The simple fool's guide to PCR. – Department of Zoology, Honolulu.
- PENNY, S. G., A. CROTTINI, F. ANDREONE, A. BELLATI, L. M. S. RAKOTOZAFY, M. W. HOLDERIED, C. SCHWITZER & G. M. ROSA (2017): Combining old and new evidence to increase the known biodiversity value of the Sahamalaza Peninsula, Northwest Madagascar. – *Contribution to Zoology*, **86**: 273–296.
- PERL, R. G. B., Z. T. NAGY, G. SONET, F. GLAW, K. C. WOLLENBERG & M. VENCES (2014): DNA barcoding Madagascar's amphibian fauna. – *Amphibia-Reptilia*, **35**: 197–206.
- POTH, D., K. C. WOLLENBERG, M. VENCES & S. SCHULZ (2012): Volatile amphibian pheromones: macrolides of mantellid frogs from Madagascar. – *Angewandte Chemie International Edition*, **51**: 2187–2190.
- PUENTE, M., F. GLAW, D. R. VIEITES & M. VENCES (2009): Review of the systematics, morphology and distribution of Malagasy dwarf geckos, genera *Lygodactylus* and *Microscalabotes* (Squamata: Gekkonidae). – *Zootaxa*, **2103**: 1–76.
- PUENTE, M., M. THOMAS & M. VENCES (2005): Phylogeny and biogeography of Malagasy dwarf geckos, *Lygodactylus* Gray, 1864: preliminary data from mitochondrial DNA sequences (Squamata: Gekkonidae). – pp. 229–235 in: HUBER, B. A. & K. H. LAMPE (eds): *African Biodiversity – Molecules, Organisms, Ecosystems*. – Springer, Berlin.
- PYRON, R. A., F. T. BURBRINK & J. J. WIENS (2013): A phylogeny and revised classification of Squamata, including 4161 species of lizards and snakes. – *BMC Evolutionary Biology*, **13**: 93.
- RABEMANANJARA, F. C. E., N. RASOAMAMPIONONA RAMINOSOA, O. RAVOAHANGIMALALA RAMILJAONA, D. RAKOTONDRAVONY, F. ANDREONE, P. BORA, A. I. CARPENTER, F. GLAW, T. RAZAFINDRABE, D. VALLAN, D. R. VIEITES & M. VENCES (2008): Malagasy poison frogs in the pet trade: a survey of levels of exploitation of species in the genus *Mantella*. – pp. 277–300 in: ANDREONE, F. (ed.): *A Conservation Strategy for the Amphibians of Madagascar*. – Monografie del Museo Regionale di Scienze Naturali di Torino.
- RAKOTONDRAVONY, H. A. & S. M. GOODMAN (2011): Rapid herpetofaunal surveys within five isolated forests on sedimentary rock in western Madagascar. – *Herpetological Conservation and Biology*, **6**: 297–311.
- RAMANAMANJATO, J. B. & N. RABIBISOA (2002): Evaluation rapide de la diversité biologique des reptiles et amphibiens de la Réserve Naturelle Intégrale d'Ankarafantsika. – pp. 98–103 and 135–138 in: ALONSO, L. E., T. S. SCHULENBERG, S. RADILOFE & O. MISSA (eds): *Une evaluation biologique de la Réserve Naturelle Intégrale d'Ankarafantsika, Madagascar*. – Bulletin RAP d'évaluation rapide No. 23, Conservational International, Washington D.C., USA.
- RASELIMANANA, A. P. (2008): Herpétofaune des forets sèches malgaches. – *Malagasy Nature*, **1**: 46–75.
- RASELIMANANA, A. P., B. NOONAN, K. P. KARANTH, J. GAUTHIER & A. D. YODER (2009): Phylogeny and evolution of Malagasy plated lizards. – *Molecular Phylogenetics and Evolution*, **50**: 336–344.
- RASELIMANANA, A. P. & D. RAKOTOMALALA (2003): Chamaeleonidae, Chameleons. – pp. 961–969 in: GOODMAN S. M. & J. P. BENSTEAD (eds): *The Natural History of Madagascar*. – The University of Chicago Press, Chicago and London.
- RAXWORTHY, C. J. & R. A. NUSSBAUM (1997): Biogeographic Patterns of Reptiles in Eastern Madagascar. – pp. 124–141 in: GOODMAN S. M. & B. PATTERSON (eds): *Natural change and human impact in Madagascar*. – Washington D. C., Smithsonian Institution Press.
- RECKNAGEL, H., K. R. ELMER, P. N. BRICE, A. P. RASELIMANANA, A. MEYER & M. VENCES (2013): Multi-gene phylogeny of Madagascar's plated lizards, *Zonosaurus* and *Tracheloptychus* (Squamata: Gerrhosauridae). – *Molecular Phylogenetics and Evolution*, **69**: 1215–1221.
- REYNOLDS, R. G., L. M. L. NIEMILLER & L. J. REVELL (2014): Toward a Tree-of-Life for the boas and pythons: Multilocus species-level phylogeny with unprecedented taxon sampling. – *Molecular Phylogenetics and Evolution*, **71**: 201–213.
- ROCHA, L. A., A. ALEIXO, G. ALLEN, F. ALMEDA, C. C. BALDWIN, M. V. L. BARCLAY, J. M. BATES, A. M. BAUER, F. BENZONI, C. M. BERNS, M. L. BERUMEN, D. C. BLACKBURN, S. BLUM, F. BOLAÑOS, R. C. K. BOWIE, R. BRITZ, R. M. BROWN, C. D. CADENA, K. CARPENTER, L. M. CERÍACO, P. CHAKRABARTY, G. CHAVES, J. H. CHOAT, K. D. CLEMENTS, B. B. COLLETTE, A. COLLINS, J. COYNE, J. CRACRAFT, T. DANIEL, M. R. DE CARVALHO, K. DE QUEIROZ, F. DI DARIO, R. DREWES, J. P. DUMBACHER, A. ENGLISH JR, M. V. ERDMANN, W. ESCHMEYER, C. R. FELDMAN, B. L. FISHER, J. FJELDSÅ, P. W. FRITSCH, J. FUCHS, A. GETAHUN, A. GILL, M. GOMON, T. GOSLINER, G. R. GRAVES, C. E. GRISWOLD, R. GURALNICK, K. HARTEL, K. M. HELGEN,

- H. HO, D. T. ISKANDAR, T. IWAMOTO, Z. JAAFAR, H. F. JAMES, D. JOHNSON, D. KAVANAUGH, N. KNOWLTON, E. LACEY, H. K. LARSON, P. LAST, J. M. LEIS, H. LESSIOS, J. LIEBHERR, M. LOWMAN, D. L. MAHLER, V. MAMONEKENE, K. MATSUURA, G. C. MAYER, H. MAYS JR, J. MCCOSKER, R. W. MCDIARMID, J. MCGUIRE, M. J. MILLER, R. MOOI, R. D. MOOI, C. MORITZ, P. MYERS, M. W. NACHMAN, R. A. NUSSBAUM, D. Ó FOIGHIL, L. R. PARENTI, J. F. PARHAM, E. PAUL, G. PAULAY, J. PÉREZ-EMÁN, A. PÉREZ-MATUS, S. POE, J. POGONOSKI, D. L. RABOSKY, J. E. RANDALL, J. D. REIMER, D. R. ROBERTSON, M.-O. RÖDEL, M. T. RODRIGUES, P. ROOPNARINE, L. RÜBER, M. J. RYAN, F. SHELDON, G. SHINOHARA, A. SHORT, W. B. SIMISON, W. F. SMITH-VANIZ, V. G. SPRINGER, M. STIASSNY, J. G. TELLO, C. W. THOMPSON, T. TRNSKI, P. TUCKER, T. VALQUI, M. VECCHIONE, E. VERHEYEN, P. C. WAINWRIGHT, T. A. WHEELER, W. T. WHITE, K. WILL, J. T. WILLIAMS, G. WILLIAMS, E. O. WILSON, K. WINKER, R. WINTERBOTTOM & C. C. WITT (2014): Specimen collection: An essential tool. – *Science*, **344**: 814–815.
- ROCHA, S., H. RÖSLER, P.-S. GEHRING, F. GLAW, D. POSADA, D. J. HARRIS & M. VENCES (2010): Phylogenetic systematics of day geckos, genus *Phelsuma*, based on molecular and morphological data (Squamata: Gekkonidae). – *Zootaxa*, **2429**: 1–28.
- ROCHA, S., M. VENCES, F. GLAW, D. POSADA & D. J. HARRIS (2009): Multigene phylogeny of Malagasy day geckos of the genus *Phelsuma*. – *Molecular Phylogenetics and Evolution*, **52**: 530–537.
- ROSA, G. M., F. ANDREONE, A. CROTTINI, J. S. HAUSWALDT, J. NOËL, N. H. RABISOA, M. O. RANDRIAMBAHINIARIME, R. REBELO & C. J. RAXWORTHY (2012): The amphibians of the relict Betampona low-elevation rainforest, eastern Madagascar: an application of the integrative taxonomy approach to biodiversity assessments. – *Biodiversity and Conservation*, **21**: 1531–1559.
- ROSA, G. M., R. BOISTEL, E. CAMPANTICO, B. GILLET, P. EUSEBIO BERGÒ & F. ANDREONE (2014): Case solved: presence of toxin-secreting oral glands in the lamprophiid snake *Mimophis mahfalensis* (Grandidier, 1867) from Madagascar. – *Zoomorphology*, **133**: 417–423.
- ROSA, G. M., V. MERCURIO, A. CROTTINI & F. ANDREONE (2010): Predation of the snake *Leioheterodon modestus* (Günther, 1863) upon the microhylid frog *Scaphiophryne gottlebei* Busse & Böhme, 1992 at Isalo, southern Madagascar. – *Herpetology Notes*, **3**: 259–261.
- RUANE, S., F. T. BURBRINK, B. RANDRIAMAHATANTSOA & C. J. RAXWORTHY (2016): The cat-eyed snakes of Madagascar: phylogeny and description of a new species of *Madagascarophis* (Serpentes: Lamprophiidae) from the Tsingy of Ankarana. – *Copeia*, **104**: 712–721.
- RUANE, S., E. A. MYERS, K. LO, S. YUEN, R. S. WELT, M. JUMAN, I. FUTTERMAN, R. A. NUSSBAUM, G. SCHNEIDER, F. T. BURBRINK & C. J. RAXWORTHY (2017): Unrecognized species diversity and new insights into colour pattern polymorphism within the widespread Malagasy snake *Mimophis* (Serpentes: Lamprophiidae). – *Systematics and Biodiversity*, DOI: 10.1080/14772000.2017.1375046.
- SCHAEFER, H.-C., M. VENCES & M. VEITH (2002): Molecular phylogeny of Malagasy poison frogs, genus *Mantella* (Anura: Mantellidae): homoplastic evolution of colour pattern in aposematic amphibians. – *Organisms Diversity and Evolution*, **2**: 97–105.
- SCHATZ, G. E. (2000): Endemism in the Malagasy tree flora. – pp. 1–9 in: LOURENCO W. R. & S. M. GOODMAN (eds): Biogeography of Madagascar. – *Memoires de la Societe de Biogeographie*, Paris.
- SCHMITZ, A., M. C. BRANDLEY, P. MAUSFELD, M. VENCES, F. GLAW, R. A. NUSSBAUM & T. W. REEDER (2005): Opening the black box: phylogenetics and morphological evolution of the Malagasy fossorial lizards of the subfamily “Scincinae”. – *Molecular Phylogenetics and Evolution*, **34**: 118–133.
- SOUND, P., J. KOSUCH, M. VENCES, A. SEITZ & M. VEITH (2006): Preliminary molecular relationships of Comoroan day geckos (*Phelsuma*). – pp. 175–179 in: VENCES, M., J. KÖHLER, T. ZIEGLER & W. BÖHME (eds): *Herpetologia Bonnensis II*. – Proceedings of the 13th Congress of the Societas Europaea Herpetologica. – Zoologisches Forschungsmuseum A. Koenig and Societas Europaea Herpetologica, Bonn.
- TOWNSEND, T. M., D. R. VIEITES, F. GLAW & M. VENCES (2009): Testing species-level diversification hypotheses in Madagascar: the case of microendemic *Brookesia* leaf chameleons. – *Systematic Biology*, **58**: 641–656.
- VENCES, M., F. GLAW, V. MERCURIO & F. ANDREONE (2004): Review of the Malagasy tree snakes of the genus *Stenophis*. – *Salamandra* **40**: 161–179.
- VENCES, M., A. LIMA, A. MIRALLES & F. GLAW (2014): DNA barcoding assessment of genetic variation in two widespread skinks from Madagascar, *Trachylepis elegans* and *T. gravenhorstii* (Squamata: Scincidae). – *Zootaxa*, **3755**: 477–484.
- VENCES, M., C. J. RAXWORTHY, R. A. NUSSBAUM & F. GLAW (2003): A revision of the *Scaphiophryne marmorata* complex of marbled toads from Madagascar, including the description of a new species. – *Herpetological Journal*, **13**: 69–79.
- VENCES, M., M. THOMAS, R. M. BONETT, D. R. VIEITES (2005b): Deciphering amphibian diversity through DNA barcoding: chances and challenges. – *Philosophical Transactions of the Royal Society B: Biological Sciences*, **360**: 1859–1868.
- VENCES, M., M. THOMAS, A. VAN DER MEIJDEN, Y. CHIARI & D. R. VIEITES (2005a): Comparative performance of the 16S rRNA gene in DNA barcoding of amphibians. – *Frontiers in Zoology*, **2**: 5.
- VENCES, M., D. R. VIEITES, F. GLAW, H. BRINKMANN, J. KOSUCH, M. VEITH & A. MEYER (2003): Multiple overseas dispersal in amphibians. – *Proceedings of the Royal Society of London B*, **270**: 2435–2442.
- VENCES, M., S. WANKE, D. R. VIEITES, W. R. BRANCH, F. GLAW & A. MEYER (2004): Natural colonization or introduction? Phylogeographical relationships and morphological differentiation of house geckos (*Hemidactylus*) from Madagascar. – *Biological Journal of the Linnean Society*, **83**: 115–130.
- VENCES, M., K. C. WOLLENBERG, D. R. VIEITES & D. C. LEES (2009): Madagascar as a model region of species diversification. – *Trends in Ecology and Evolution*, **24**: 456–465.
- 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. – *Proceedings of the National Academy of Sciences of the United States of America*, **106**: 8267–8272.
- WAEBER, P. O., L. WILMÉ, B. RAMAMONJISOA, C. GARCIA, D. RAKOTOMALALA, Z. H. RABEMANANJARA, C. KULL, J. U. GANZHORN & J. P. SORG (2015): Dry forests in Madagascar: neglected and under pressure. *International Forestry Review* – Special Issue: Global Dry Forests, **17**: 126–147.

Supplementary material

8 Supplementary Tables:

Table S1. List of toponyms and corresponding GPS coordinates.

Table S2. List of amphibian samples analysed for this study.

Table S3. List of Isalo's voucher specimens of amphibians unequivocally identified based on morphology and/or molecular analyses.

Table S4. List of reptile samples analysed for this study.

Table S5. List of Isalo's voucher specimens of reptiles unequivocally identified based on morphology and/or molecular analyses.

Table S6. Primer sequences and PCR conditions used in the present study.

Table S7. Pairwise genetic distances (p-distances) between and within amphibian taxa according to 16S rRNA gene fragment sequence variation.

Table S8. Pairwise genetic distances (p-distance) between and within reptile taxa according to COI gene fragment sequence variation.