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HAL Id: hal-03176669
https://hal.inrae.fr/hal-03176669

Submitted on 22 Mar 2021

Article

A new endemic species of *Hemidactylus* (Squamata: Gekkonidae) from São Nicolau Island, Cabo Verde

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Abstract

A new species of gecko of the genus *Hemidactylus* (Squamata: Gekkonidae) is described from São Nicolau Island, Cabo Verde Archipelago, and the Sal and Boavista island populations of *Hemidactylus boavistensis* (i.e., *Hemidactylus boavistensis boavistensis* **comb. nov.** and *Hemidactylus boavistensis chevalieri* **comb. nov.**) are recognized as subspecies. *Hemidactylus nicolauensis* **sp. nov.** is genetically distinct from *H. bouvieri*, to which it has previously been referred, and from all other closely related endemic *Hemidactylus* from Cabo Verde Islands in mitochondrial (12S & cyt *b*) and nuclear (RAG2, MC1R) markers. It is characterized morphologically by its distinct colouration and a diagnostically different arrangement of digital lamellae. With the description of this new species, São Nicolau is now known to harbour three single-island endemic gecko species, and the documented reptile diversity in Cabo Verde is raised to 23 endemic species. As a result of our taxonomic changes, existing conservation regulations should be updated and the conservation status of these taxa should be re-evaluated.

Key words: Cryptic diversity, DNA, integrative taxonomic revision, Macaronesia, morphology, reptile

Resumo

Uma nova espécie de osga do género *Hemidactylus* (Squamata: Gekkonidae) da ilha de São Nicolau, arquipélago de Cabo Verde, é descrita, e as populações de *Hemidactylus boavistensis* das ilhas do Sal e da Boavista (*Hemidactylus boavistensis boavistensis* **comb. nov.** e *Hemidactylus boavistensis chevalieri* **comb. nov.**) são reconhecidas como subespécies. A espécie *Hemidactylus nicolauensis* **sp. nov.** é geneticamente distinta de *H. bouvieri*, à qual foi anteriormente associada, e de todos os outros *Hemidactylus* endémicos muito aparentados das ilhas de Cabo Verde nos marcadores mitocondriais (12S & cyt *b*) e nucleares (RAG2, MC1R). Caracteriza-se morfologicamente pela coloração distinta e por um arranjo diferente e diagnóstico das lamelas digitais. Com a descrição desta nova espécie, São Nicolau alberga agora três espécies de osga endémicas de uma única ilha e a diversidade de répteis documentada em Cabo Verde aumenta para 23 espécies endémicas. Como resultado das nossas mudanças taxonómicas, os regulamentos de conservação existentes deverão ser actualizados e os estatutos de conservação desses taxa deverão ser reavaliados.

Introduction

The genus *Hemidactylus* Oken, 1817 presently consists of 165 named species (Uetz *et al.* 2019) distributed across all tropical and subtropical continental landmasses, as well as hundreds of oceanic and continental islands. The ge-

nus originated from Asia and Africa and naturally colonized South America via transmarine dispersal (Carranza & Arnold 2006). Recent human-mediated colonization events have resulted in the spread of several species far from their natural ranges (Carranza & Arnold 2006). Recent works on the phylogeny of the genus resulted in its division into four main lineages: (i) the tropical Asian clade, (ii) the *H. angulatus* clade, (iii) the Arid clade, and (iv) the African–Atlantic clade (Bauer *et al.* 2010; Carranza & Arnold 2006). Endemic *Hemidactylus* species from Cabo Verde Islands belong to the last one.

The Cabo Verde Islands are an oceanic archipelago of volcanic origin located circa 450 km off the West African coast. They comprise 10 main islands plus several islets that are topologically arranged in three island groups, the northwestern, the eastern and the southern islands. São Nicolau is the closest island to mainland Africa in the north-western group (Fig. 1). Cabo Verde is currently home to three extant endemic reptile radiations: the Scincidae genus *Chioninia* (Miralles *et al.* 2011), the Phyllodactylidae genus *Tarentola* (Vasconcelos *et al.* 2010) and the Gekkoni-dae genus *Hemidactylus* (Arnold *et al.* 2008). The endemic *Hemidactylus* of Cabo Verde form a monophyletic group within the African–Atlantic clade. These geckos are small, the largest animals growing to about 50 mm snout–vent length (SVL), and characterized by a pointed snout, by the absence of enlarged tubercles on the dorsum (except some scales in *H. lopezjuradoi*), by having a reduced number of precloacal pores in males, and by a dorsal pattern of few dark transverse bands. These geckos differ from the species of *Tarentola* by having claws in all fingers and toes and divided subdigital lamellae.

Before Arnold *et al.* (2008) revised the taxonomy of the Cabo Verdean endemic *Hemidactylus*, all autochtho-nous populations were included in a single species, *Hemidactylus bouvieri* (Bocourt, 1870), with three recognized subspecies: *Hemidactylus bouvieri boavistensis* Boulenger, 1906 on Boavista and Sal, *Hemidactylus bouvieri razoensis* Gruber & Schleich, 1982 on Santa Luzia and Raso Islet (Desertas Islands group, Desertas herein) and *Hemidactylus bouvieri bouvieri* known from Santo Antão, São Vicente, Santiago, Fogo, and Brava (Schleich 1987; Mateo *et al.* 1997; see Fig. 1). Then, Arnold *et al.* (2008) re-elevated *H. boavistensis* to the species rank and de-scribed the Fogo population as a new species, *Hemidactylus lopezjuradoi* Arnold, Vasconcelos, Harris, Mateo and Carranza, 2008. In spite of its genetic divergence from typical *H. bouvieri*, a recently discovered population on the island of São Nicolau (see Köhler *et al.* 2007; Arnold *et al.* 2008) was retained within *H. bouvieri* because available material was too poorly-preserved to allow a detailed morphological description. However, Arnold *et al.* (2008) suggested that once better material became available, this form would warrant description as a distinct taxon. For the same reasons, the two island populations of *H. boavistensis* were retained as a single taxon despite the presented mitochondrial and nuclear divergences (Arnold *et al.* 2008), as well as supposed morphological differences between them (Angel 1935).

In addition to these endemic taxa, two non-native species, *Hemidactylus angulatus* Hallowell, 1852 and either *Hemidactylus mabouia* (Moreau de Jonnès, 1818) or *Hemidactylus mercatorius* Gray, 1842 (the taxonomy of these species and the allocation of the Cabo Verdean population needs further investigation) are now widespread on some of the islands of the archipelago (Vasconcelos *et al.* 2013).

Hemidactylus boavistensis is currently classified as Near Threatened based on the IUCN categories due to ongoing infrastructure development along coastal areas, spread of the introduced species *H. angulatus* and climate change (Vasconcelos 2013a). *Hemidactylus bouvieri* is listed as Critically Endangered on the basis that, while it is poorly-known due to its rarity, it has a very low extent of occurrence, its populations are thought to be severely fragmented and threatened by introduced predators and habitat loss (Vasconcelos 2013b). This species has rarely been recorded, and is known only from isolated records on the islands where it has been reported. Vasconcelos *et al.* (2013) recorded a single individual of *H. bouvieri* on Santo Antão in 2007, the first record in over a century, during 68 days of survey work. Similarly, two specimens collected in 1998 and 2016 on São Vicente represent the only reports from this island for 100 years (Andreone 2000, this study), from where only three specimens were previously known. On other islands the number of specimens and records is a little bit higher. On São Nicolau, ten specimens have been collected between 1997 and 2012 (Köhler *et al.* 2007; Arnold *et al.* 2008; this study); on the Desertas group, the subspecies *H. b. razoensis* is known from a total of 19 individuals, ten from Raso found in 1981 and reported in Gruber & Schleich (1982), three found on Raso by Mateo and Geniez in 1997 (this study), and three found on Raso in 2019 by Ricardo Rocha (pers. comm.), plus two from Santa Luzia found in 1995 or 1996 reported by Mateo *et al.* (1997) and one from Santa Luzia found in 2012 (this study). Only a handful of specimens are known from Santiago, all from the 19th century except for one collected in 1954 and reported by Mertens (1954). None has ever been genetically analysed. In the present work, we use molecular and morphological data to describe the

population of *Hemidactylus* from São Nicolau Island as a new endemic species and reassess the taxonomic status of the *Hemidactylus* populations from Sal and Boavista.

Material and methods

A list of all individuals included in the molecular and/or morphological analyses with their taxonomic identifications, sample codes, voucher references, corresponding geographical locations and GenBank accession numbers for all sequenced genes (when relevant) is presented in Table 1. The geographic distribution of samples used in this study is depicted in Fig. 1, and the haplotype networks in Fig. 2. Pictures of the voucher of the holotype is presented in Figs. 3 and 4, and a phylogenetic tree with integrative taxonomy results and the pairwise genetic distances in Figs. 5 and 6, respectively. Live pictures of the new taxa are depicted in Fig. 7.

Materials, DNA extraction, amplification and sequencing

A total of 72 sequences from two mitochondrial genes, a fragment of the 12S ribosomal gene (12S), and two portions of the cytochrome *b* (cyt *b* 1 and cyt *b* 2), and part of one nuclear marker encoding the recombination activating gene 2 (RAG2) of 22 of individuals of endemic Cabo Verde *Hemidactylus* were downloaded from GenBank (Table 1). Our field surveys were able to locate three more individuals, one from São Vicente and two from São Nicolau that we sampled (tail tips), analysed for the present study and released afterwards (except the São Nicolau individuals used for species description). Tissue samples housed in the collection of 'Biogéographie et Ecologie des Vertébrés', 'Centre d'Ecologie Fonctionnelle et Evolutive', Montpellier, are catalogued with BEV.T initials. Genomic DNA was extracted from ethanol-preserved tissue samples using the Qiagen DNeasy Blood & Tissue Kit. DNA samples from these new individuals were amplified and sequenced for the same available markers, using the same conditions as described in Arnold *et al.* (2008). We also sequenced a fragment of the melano-cortin 1 receptor (MC1R) nuclear marker for a selection of new and previously available tissue samples using the conditions described in Vasconcelos *et al.* (2012). The new sequences were deposited in GenBank (accession MT890017–MT890036 and MT905068; see Table 1). Sequences from two outgroup taxa (*Hemidactylus robustus* and *Hemidactylus turcicus*) were downloaded from GenBank. For *H. robustus*, all sequences should correspond to the same specimen (Šmíd *et al.* 2015) but for *H. turcicus* the concatenated alignment combines gene fragments from three different specimens from Spain. Given the low genetic structure of the species in Spain, this should not be a problem (Rato *et al.* 2011).

Molecular analyses

Sequencing chromatograms were checked manually, assembled and edited using Geneious v.6.1.6 (Biomatters Ltd.). DNA sequences were aligned using MAFFT (Katoh & Toh 2008). Coding mtDNA and nDNA gene fragments were translated into amino acids and no stop codons were observed. The two cyt *b* fragments were concatenated for distance calculations.

For nuclear loci, heterozygous individuals were identified based on the presence of two peaks of approximately equal height at a single nucleotide site. Phased nuclear sequences were used for the network analyses, and the software PHASE v. 2.1.1 implemented in DNAsp (Librado & Rozas 2009) was used to resolve phased haplotypes using all sequences.

Between and within groups mean genetic distances were calculated with MEGA version 6.0 (Tamura *et al.* 2013), using the 'pairwise deletion' option for handling ambiguous positions, missing data and indels for mtDNA markers with the K2P model (with uniform rates among sites). There were a total of 2 557 base pairs in the final dataset.

As in other studies of Cabo Verdean reptiles (Miralles *et al.* 2011; Vasconcelos *et al.* 2012), the genealogical relationships among taxa were assessed using cyt *b* 1 and MC1R markers with haplotype networks constructed using statistical parsimony (Templeton *et al.* 1992) implemented in the program TCS v.1.21 (Clement *et al.* 2005), using phased sequences with a connection limit of 95% and deletions treated as a fifth state. A maximum-likelihood tree was built with MEGA using a concatenated data set (12S, cyt *b*, MC1R and RAG2) and its support evaluated with 1 000 bootstrap pseudo-replicates. The best model of DNA evolution (GTR + G) was determined with MEGA. Default options were used for the tree building and the 'pairwise deletion' option was selected for missing data and indels. We also used MrBayes (Ronquist *et al.* 2012) to obtain a Bayesian analysis tree using the same model of

DNA evolution. The number of generations of the MCMC chain was adjusted until the potential scale reduction factor and effective sample size of each parameter met the criteria recommended in the MrBayes manual (available at http://mrbayes.net). The topology was exactly the same as with ML, except for Santa Luzia that was grouped with Raso in the Bayesian but not in the ML tree. Additionally, a species delimitation test incorporated in Geneious was performed.

Morphological analyses

For *Hemidactylus bouvieri* and the new species, a total of 30 specimens were examined using several sources: examination of colour photos of live and museum specimens, and examination of museum specimens (Table 1). This represent most known specimens of *Hemidactylus bouvieri* from Santo Antão, São Vicente, Santa Luzia, Raso, and São Nicolau. We also examined for comparative purposes specimens of *H. lopezjuradoi* and *H. boavistensis*. Specimens from Brava and Santiago were not included, but preliminary biogeographical evidence suggest they do not belong to *H. bouvieri* (Arnold *et al.* 2008). Examined voucher specimens originate from the following collections: 'Senckenberg Forschungsinstitute und Naturmuseen' (SMF), Frankfurt am Main; 'Zoologische Staatssammlung München' (ZSM), Munich; 'Muséum national d'Histoire naturelle' (MNHN), Paris; Natural History Museum of United Kingdom (NHMUK), London; 'Museo Civico 'Craveri' di Storia Naturale' (MCCSN), Bra; 'Biogéographie et Ecologie des Vertébrés' collection in 'Centre d'Ecologie Fonctionnelle et Evolutive' (BEV), Montpellier; and 'Universidade Técnica do Atlântico' (UCV), Mindelo, Cabo Verde. We also used photos of live specimens housed in Philippe Geniez's photo collection (PGe), Montpellier, now available on MorphoBank (project 2812).

The following characters were examined by the same person, Pierre-André Crochet (PAC), on voucher specimens and, for some of them, photos: snout–vent length (SVL), measured from tip of snout to vent; number of supralabials and infralabials (combined as total number of scales on both sides, IL+SL); number of enlarged ventral scale rows at midbody (VENL); number of scales in a line between the middle of the eyes on top of head (excluding superciliary granules, STH); and number and disposition of lamellae under the first and third finger and toe (especially number of divided lamellae under third finger, DTF, and third toe, DTT, respectively). Colour pattern, *habitus,* and additional potentially diagnostic characters of live and preserved specimen were also checked from photo or voucher by comparing specimens of each major genetic lineage. Small sample size for every population precluded meaningful statistical testing in most cases, but non-parametric tests (Mann-Whitney U test) were performed on some variables. Descriptive statistics are presented below.

For *H. boavistensis*, only photos of live specimens where studied (34 specimens from Boavista and 11 specimens from Sal).

Integrative taxonomy framework

For consistency, the same approach used in the taxonomic revision of the other Cabo Verde endemic lizard genus, *Chioninia* and *Tarentola* (Miralles *et al.* 2011; Vasconcelos *et al.* 2012) was followed here, retaining as valid taxa only candidate species that are supported by at least two independent lines of evidence. Three lines of evidence were defined on the basis of the independence of their data sets (mtDNA, nDNA, and morphology) to evaluate the divergence and decide on the taxonomic status of the São Nicolau lineage: (1) mtDNA—presence of independent cyt *b* parsimony networks with a connection limit of 95%; (2) nDNA—absence of shared haplotypes in the MC1R nuclear gene; (3) morphology—fixed diagnostic character state or a set of a unique combination of characters (Miralles *et al.* 2011; Vasconcelos *et al.* 2012).

Results

Molecular analyses

The results of the haplotype network analyses are shown in Fig. 2 and clearly show that, in cyt *b* 1*,* only one haplotype was shared between two populations, namely between Raso and Santa Luzia. The remaining haplotypes from the different island populations were singletons. Fourteen haplotypes were recovered from endemic *Hemidactylus* populations in the archipelago. São Nicolau unique haplotypes were disconnected from the other haplotypes recovered from other endemic *Hemidactylus* populations from the northwestern islands. *H. lopezjuradoi* haplotypes were also disconnected and formed an isolated network with two haplotypes. Haplotypes of *H. boavistensis* from

Boavista and Sal also formed isolated networks with three haplotypes each. In the MC1R network, the *Hemidactylus* populations from northwestern (*H. bouvieri* and São Nicolau population), eastern (*H. boavistensis*) and southern islands (*H. lopezjuradoi*) presented different haplogroups.

The phylogenetic analyses by Arnold *et al.* (2008) clearly indicated that the São Nicolau population is genetically distinct from *H. bouvieri,* and that the Sal and Boavista populations of *H. boavistensis* were also differentiated, although to a lower degree. The molecular analyses performed in this study support the hypothesis that the São Nicolau population and *H. bouvieri,* sensu stricto from the remaining northwestern islands are reciprocally monophyletic (Figs. 2, 5) and present a high level of genetic divergence between them (Table 2, Fig. 6), especially in the mitochondrial markers: K2P distance (12S, cyt *b*)= 3.9±1.0, 11.8±1.4, respectively. Moreover, the network analyses also depicted no connection in cyt *b* 1 and no haplotype sharing between those two species in the nuclear marker (Fig. 2). This study also confirms that Sal and Boavista populations are reciprocally monophyletic in mtDNA, with a moderate level of divergence in mitochondrial markers: K2P distance (12S; cyt *b*)= 0.9±0.4; 4.1±0.7, respectively.

The species delimitation test for São Nicolau's clade significantly supported it as a different taxonomical unit: P= 1.00 (range: 0.86–1.0), clade support of 100% (Rosenberg's P= 0.00023). Genetic distances from other clades in cyt *b* were always higher than 11% (Table 2).

Morphological analyses

Specimens from Santo Antão, São Vicente, Santa Luzia and Raso are all remarkably similar in appearance. As already noted by Arnold *et al.* (2008), the main difference between Raso and Santa Luzia (*H. b. razoensis*) and the two larger islands (*H. bouvieri bouvieri*) is body size (see Table 3, maximum SVL= 29.0 mm in *H. b. razoensis* versus 35.6 mm in *H. b. bouvieri*, Mann-Whitney U test, P= 0.004), while there was no significant difference in all other examined characters (results not shown, see Table 3 for descriptive statistics).

Specimens from São Nicolau are also very similar in appearance to those of the neighbouring islands, but they differ in a slightly different colouration and a diagnostically different arrangement of lamellae under the toes and finger (DTF and DTT are significantly different: Mann-Whitney U test, P= 0.004 and P= 0.002, respectively; see also diagnosis and variation below). Their general colouration makes them superficially more similar to *H. lopezjuradoi* than to *H. bouvieri* from Santo Antão, São Vicente, Santa Luzia and Raso, but they differ from *H. lopezjuradoi* in snout shape and pholidosis (see below; Figs. 3, 4). They are easily separated from *H. boavistensis* by the same characters that separate that species from *H. bouvieri* (see Arnold *et al.* 2008 and diagnosis below).

According to Angel (1935), specimens from Sal and Boavista are differentiated by the fact that the former usually have six lamellae under the first toe (versus four to five in Boavista specimens) and lack the transversal bands on the head and nape (versus present). We could not check ourselves if the differences on number of subdigital lamellae are real, but we found at least one specimen from Boavista with six lamellae under the first finger. The reported differences in colouration are in fact very weak and there is ample overlap between the populations from Boavista and Sal in this character (Fig. 7).

Taxonomic accounts

The genetic distinctiveness of the populations from São Nicolau from the remaining *Hemidactylus* populations of the Cabo Verde Archipelago in two mitochondrial and two nuclear gene fragments (Figs. 2, 5, 6), and their differentiation in several morphological traits (see morphological analyses of Results above and diagnosis below; Figs. 3, 4), demonstrate that the populations from São Nicolau constitute a distinct evolutionary unit, sister to *H. bouvieri*, that we recognize as a species level unit and that deserves to be named following all integrative approaches. Deciding which rank to allocate to this new taxon from São Nicolau is more problematical, as it is often the case for diverging allopatric lineages. In case of allopatric taxa, we follow a pragmatic approach, where lineages that differ as much as widely accepted species in the same group should be given species rank, as advocated for example by Helbig *et al.* (2002).

The genetic divergence between the São Nicolau lineage and *H. bouvieri* sensu stricto is more similar to the divergence among widely accepted species of *Hemidactylus* in Cabo Verde than among conspecific populations (Fig. 6, Table 2), both in mtDNA as well as for the combined mtDNA and nDNA dataset. The MC1R network (Fig. 2) revealed a complete lack of shared haplotypes with *H. bouvieri*, whereas populations of *H. bouvieri* or *H. boavistensis* on different islands share some polymorphisms. Finally, the distinctive colouration and diagnostic arrange-

ment of lamellae under toes and fingers of the specimens from São Nicolau sets it apart from *H. b. bouvieri* and *H. b. razoensis* as much as from *H. lopezjuradoi* and *H. boavistensis* (see Arnold *et al.* 2008). Thus, we think a specific rank is the most appropriate category for the new taxon from São Nicolau.

The Sal and Boavista populations present independent cyt *b* networks, but share haplotypes in the nuclear marker and lack fixed diagnostic morphological characters (Fig. 2, 5). Hence, following the integrative taxonomy framework, these island populations are best considered to be conspecific, but should be recognized as subspecies (Fig. 5). Even though *Hemidactylus* from Sal and Boavista show distinctiveness in the mitochondrial markers, their genetic divergence is smaller than the interspecific divergences reported above (Table 2), and some individuals do not exhibit the distinct colouration patterns (Fig. 7) or lamella counts typical of their population. As previously note by Loveridge (1947), colouration in such a variable species can scarcely be expected to form a stable basis for separation. Thus, we think that subspecific rank is the most appropriated category for the *Hemidactylus* populations from Sal and Boavista.

Nomenclature

None of the older nomina that are available for the *Hemidactylus* species from Cabo Verde can be used for the São Nicolau population. *Emydactylus bouvieri* Bocourt, 1870 is based on three syntypes from São Vicente and should remain the valid nomen for the species found in Santo Antão, São Vicente, Santa Luzia and Raso. *Hemidactylus Cessacii* Bocage, 1873 was described based on three syntypes from Santiago collected by M. de Cessac and housed in the former Lisbon museum. Several other specimens from Santiago and collected by Ferreira Borges were held in Lisbon as well (Bocage 1896). All specimens formerly housed in Lisbon are now lost due to a fire. The only remaining specimens from Santiago are the specimen reported by Mertens (1954) now held in Helsinki (pers. obs.), and the London specimen reported by Arnold *et al.* (2008). We are convinced that *cessacii* cannot be used for the São Nicolau population because: 1) we have examined photos of the Helsinki specimen (Finnish Museum of Natural History MZH MS 1953) and it does not show the features associated with the São Nicolau population, and 2) there is overwhelming evidence that the populations from São Nicolau and Santiago are not conspecific. No species is shared between these two islands in the other endemic genera *Chioninia* and *Tarentola* (Vasconcelos *et al.* 2013). The status of the nomen *cessacii* needs to be examined in due course, but we anticipate that it will prove to be either conspecific with *H. lopezjuradoi* from the neighbouring island of Fogo or to constitute a valid species endemic from Santiago. *Hemidactylus bouvieri razoensis* Gruber & Schleich, 1982 is clearly conspecific with the São Vicente population, so this name cannot be used for the new taxon. Lastly, *Hemidactylus boavistensis* Boulenger, 1906 and *Hemidactylus chevalieri* Angel, 1935 are based on specimens from Boavista and Sal, respectively, and cannot be used for the São Nicolau population. We thus need to introduce a new nomen to describe the São Nicolau *Hemidactylus* as a new species.

The *Hemidactylus* from Boavista was described as *Hemidactylus boavistensis* by Boulenger (1906) whereas the *Hemidactylus* from Sal was named as *Hemidactylus chevalieri* by Angel (1935). Both *nomina* are available, with *boavistensis* having precedence over *chevalieri*, so *Hemidactylus chevalieri* can be used as the valid nomen of the Sal population if it is treated as taxonomically distinct, otherwise it is a junior subjective synonym of *H. boavistensis*. Thus, considering these two taxa as conspecific but distinct at the subspecific level, the population from Boavista should be referred as *Hemidactylus boavistensis boavistensis* (**new comb.**) whereas the one from Sal is recognized as *Hemidactylus boavistensis chevalieri* (**new comb.**).

Hemidactylus nicolauensis sp. nov.
(Figs. 1–7, Tables 1–3)
urn:lsid:zoobank.org:act:9C484E9A-5A72-4186-9C6F-1C89ECC6721B

Holotype. SMF 86992 (Morphobank M585325–M585346), adult male, from Ribeira Funda around 250 m. a.s.l., São Nicolau Island, Cabo Verde (16.66°N, 24.34°W; WGS 84), collected 01 April 2007 by Gunther Köhler, GK.

Paratypes. UCV2018/004 (previously SMF 86991; Morphobank M585298–M585321) adult female from Ribeira Funda, collected by GK; SMF 90057 (Morphobank M585736–M585746) and SMF 90058 (Morphobank M585747–M585754), juveniles and same data as holotype.

Other specimens examined. In addition to the holotype and paratypes, we have examined four specimens housed in London: NHMUK 2005.1637–40 collected 1 km past Cachaço in direction to Tarrafal de São Nicolau (16.6189°N, 24.3335°W) on 4 August 1997 by J. A. Mateo and P. Geniez. As reported by Arnold *et al.* (2008), these

were in a too poor condition to be used in morphological analyses, but three of them have been included in the genetic analyses. Also, the animals corresponding to the tissue number BEV. T5652, DNA sample code H07, Morphobank M675961– M675962), adult female and tissue number BEV. T5651 (DNA sample code H06, Morphobank M673362–M673366), adult female, both from 200 m NE of Cachaço church, São Nicolau, Cabo Verde (16.6242°N, 24.3301°W), found 01 March 2012 by PAC were also examined. As far as we know these ten specimens constitute all the known material and records of the species. Five specimens were included in the genetic analyses, and eight voucher specimens were included in the morphological analyses (Table 1), including the holotype and paratypes.

Etymology. The species epithet refers to the island of São Nicolau, the only island of Cabo Verde where the new species is currently known to occur.

Diagnosis. A small *Hemidactylus* that shares all the morphological features common to the Cabo Verde endemic species (see Arnold *et al.* 2008), but characterized by the following combination of characters: no enlarged tubercles on dorsum or upper tail, two precloacal pores (one on each side) in males, two fully divided toe lamellae at the tip of the longest fingers and toes.

Hemidactylus nicolauensis **sp. nov.** is morphologically most similar to its sister taxon *H. bouvieri*, as both species have enlarged scales and toe lamellae (scansors) under toes and fingers, with minute hair-like structure under the toe lamellae. It is differentiated from *H. bouvieri* by the following characters: in *H. nicolauensis* **sp. nov.** the maximum size is larger (SVL 41 mm vs. 36 mm in *H. bouvieri*; Table 2), the toe lamellae tend to be more divided, with usually two fully divided toe lamellae (sometimes three) under the longest toes and fingers, whereas in *H. bouvieri*, the toe lamellae are entire (with a narrowing at the middle for the largest ones) or there is one or (rarely) two entirely divided toe lamellae near the tip of the fingers or toes (see Table 3). The difference is best appreciated under the third finger where it is diagnostic: there are at least two divided toe lamellae in *H. nicolauensis* **sp. nov.**, whereas in *H. bouvieri* there is at most one divided toe lamellae under third finger.
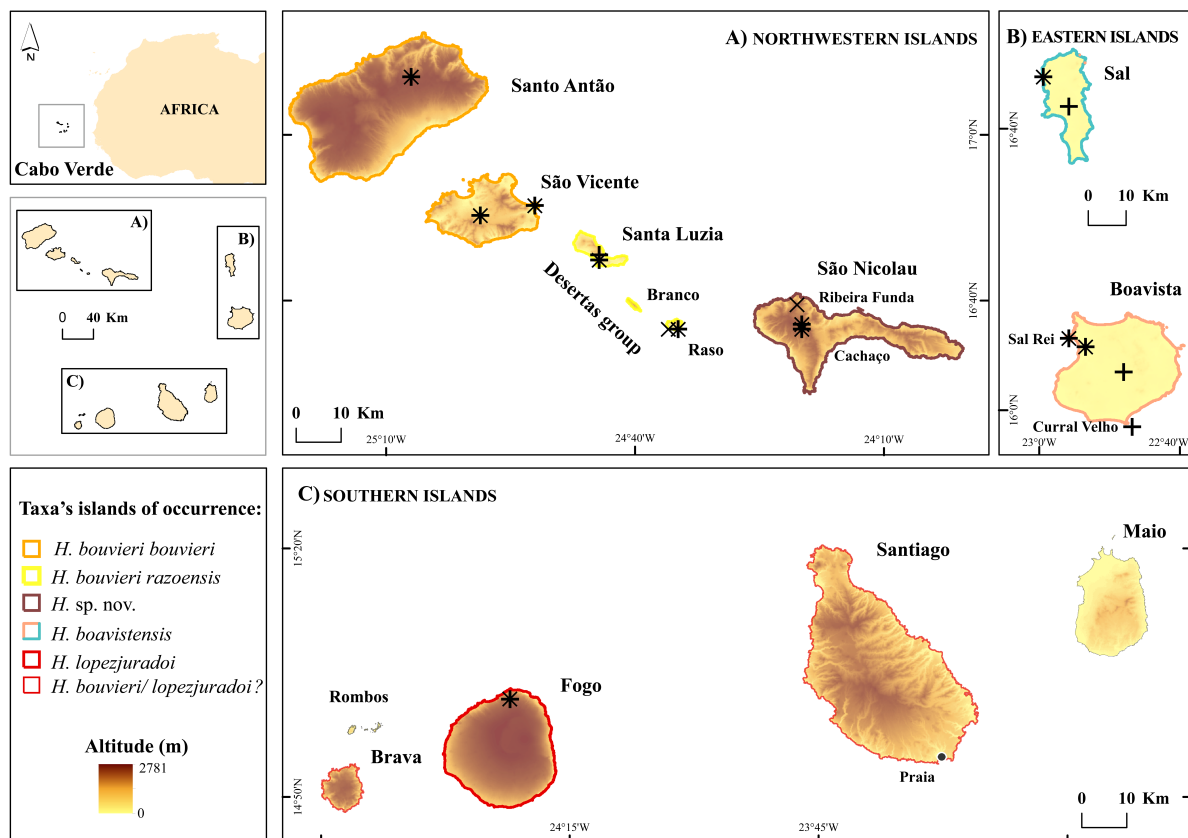
FIGURE 1. Location of the study area and samples. Map of the Cabo Verde Islands showing the geographical location (latitudes and longitudes) and altitudes of the islands and the origins of the *Hemidactylus* samples included in the morphological (X) and genetic (+) analyses (Geographic Coordinate System, Datum WGS 84). Island and taxa colours match the colours used in the network analyses. No specimens were found on Maio.

TABLE 1. Studied specimens for the molecular and morphological analyses. The location (island, locality, latitude, and longitude coordinates), year of record and name of the collector of each specimen is indicated. The DNA, voucher, GenBank and MorphoBank accession codes are also given. Taxa codes: *Hv, H. boavistensis*; *Hbb, H. bouvieri bouvieri*; *Hbr, H. bouvieri razoensis*; *Hl, H. lopezjuradoi*; *Hsp; H.* sp. nov., *Ht, H. turcicus*; *Hr, H. robustus.* Collector codes: FA, F. Andreone; GK, G. Köhler; HHS, H.H. Schleich; HJG, H.-J. Gruber; JAM, J.A. Mateo; JO, J. Oliveira; MAB, M. A. Bouvier; MAC, M. A. Carretero; PAC, P.-A. Crochet; PG, P. Geniez; PLS, P.L. Suárez; RTL, R.T. Lowe; RV, R. Vasconcelos; SM, S. Martins; SR, S. Rocha. For a few specimens (marked with *) we could not establish correspondence between tissue sample and specimen identity (although we know that the sample is one from of a series of specimens).

DNA code	Voucher code	Taxa code	Location				Year
			Island	**Locality**	**Longitude**	**Latitude**	
H01	-	*Hv*	Sal	Bunalema	16.72	-22.93	1997
H09	-	*Hv*	Sal	Buracona	16.79	-22.99	1997
H10	-	*Hv*	Sal	Buracona	16.79	-22.99	1997
HBV1	-	*Hv*	Sal	-	16.72	-22.93	-
H02	-	*Hv*	Boavista	Ribeira de água, S of Sal Rei	16.15	-22.89	1997
H11	-	*Hv*	Curral Velho	-	15.96	-22.78	-
H12	-	*Hv*	Sal Rei	-	16.17	-22.93	1997
CV38	-	*Hv*	Boavista	-	16.09	-22.80	-
CV125	-	*Hv*	Boavista	-	16.09	-22.80	-
Hrv1	-	*Hbb*	Santa Antão	Xôxô (= Lombo de Diogo)	17.12	-25.12	2006
Hrv3	-	*Hbb*	São Vicente	Praia Grande, Calhau	16.86	-24.87	2015
H54	MCCSN FA2	*Hbb*	São Vicente	Praia Grande, Calhau	16.84	-24.98	1998
-	NHMUK 1866.4.12.3	*Hbb*	São Vicente	-	16.84	-24.98	<1866
-	NHMUK 1866.4.12.4 (lost)	*Hbb*	São Vicente	-	16.84	-24.98	<1866
-	MNHN-RA-0.2444	*Hbb*	São Vicente	-	16.84	-24.98	<1870
-	MNHN-RA-1999.8221	*Hbb*	São Vicente	-	16.84	-24.98	<1870
-	MNHN-RA-1999.8222	*Hbb*	São Vicente	-	16.84	-24.98	<1870
H04	NHMUK 2005.1667	*Hbr*	Raso	Chã do Castelo, around the ruin	16.61	-24.58	1997
H14	NHMUK 2005.1666	*Hbr*	Raso	Chã do Castelo, around the ruin	16.61	-24.58	1997
H28	-	*Hbr*	Raso	Chã do Castelo, around the ruin	16.61	-24.58	1997
-	ZSM 130/1981/1	*Hbr*	Raso	near the coastline, W side	16.61	-24.60	1981
-	ZSM 130/1981/2	*Hbr*	Raso	near the coastline, W side	16.61	-24.60	1981
-	ZSM 130/1981/3	*Hbr*	Raso	near the coastline, W side	16.61	-24.60	1981

TABLE 1. (Continued)

DNA code	Voucher code	Taxa code	Locations		Longitude	Latitude	Year
			Island	Locality			
-	ZSM 130/1981/4	*Hbr*	Raso	near the coastline, W side	16.61	-24.60	1981
-	ZSM 1654/2009	*Hbr*	Raso	near the coastline, W side	16.61	-24.60	1981
-	ZSM 1655/2009	*Hbr*	Raso	near the coastline, W side	16.61	-24.60	1981
Hrv2	-	*Hbr*	Santa Luzia	above Ribeira. dos Penedos	16.75	-24.74	2012
H30	-	*Hbr*	Santa Luzia	above Ribeira dos Penedos	16.76	-24.74	1995
H29	-	*Hbr*	Santa Luzia	above Ribeira dos Penedos	16.76	-24.74	1995
H03	NHMUK 2005.1632	*Hl*	Fogo	Ribeira Culunjar, Rib. Ilhéu–Atalaia	15.03	-24.37	1997
H13	NHMUK 2005.1633	*Hl*	Fogo	Ribeira Culunjar, Rib. Ilhéu–Atalaia	15.03	-24.37	1997
-	NHMUK 2005.1634 - 36	*Hl*	Fogo	Ribeira Culunjar, Rib. Ilhéu–Atalaia	15.03	-24.37	1997
H06/BEV.T5651	-	*Hsp*	São Nicolau	200 m NE Cachaço church	16.62	-24.33	2012
H07/BEV.T5652	-	*Hsp*	São Nicolau	200 m NE Cachaço church	16.62	-24.33	2012
H05	NHMUK 2005.1639	*Hsp*	São Nicolau	1km past Cachaço	16.61	-24.33	1997
H16	NHMUK 2005.1640	*Hsp*	São Nicolau	1km past Cachaço	16.61	-24.33	1997
H17	NHMUK 2005.1638	*Hsp*	São Nicolau	1km past Cachaço	16.61	-24.33	1997
-	NHMUK 2005.1637	*Hsp*	São Nicolau	1km past Cachaço	16.61	-24.33	1997
-	UCV2018/0004/SMF 86991	*Hsp*	São Nicolau	Rib. Funda, 250 m. a.s.l.	16.66	-24.34	2007
-	SMF 86992	*Hsp*	São Nicolau	Rib. Funda, 250 m. a.s.l.	16.66	-24.34	2007
-	SMF 90057	*Hsp*	São Nicolau	Rib. Funda, 250 m. a.s.l.	16.66	-24.34	2007
-	SMF 90058	*Hsp*	São Nicolau	Rib. Funda, 250 m. a.s.l.	16.66	-24.34	2007
Hd03	-	*Ht*	-	Cabo de Gata, Spain	-	-	-
SPM002086	-	*Ht*	-	Barcelona, Spain	-	-	-
SPM003063	-	*Ht*	-	Granada, Spain	-	-	-
JS58	-	*Hr*	-	Yemen, Bir Ali	-	-	-

TABLE 1. (Continued)

DNA code	Collector code	GenBank accession code					Morphobank code
		12S	cyt *b* 1	cyt *b* 2	RAG2	MC1R	
H01	JAM/ PG	DQ120418	DQ120247	EU730670	EU730677	MT890020	M676398
H09	JAM/ PG	DQ120419	DQ120248	EU730671	-	-	M676396/7/M678219*
H10	JAM/ PG	DQ120420	DQ120249	EU730672	-	-	M676396/7/M678219*
HBV1	-	AF324812	AF324811	-	-	-	-
H02	JAM/ PG	DQ120422	DQ120251	EU730673	EU730678	MT890021	M678220–M678221*
H11	PLS	DQ120423	DQ120252	EU730674	EU730679	MT890022	-
H12	JAM/ PG	DQ120421	DQ120250	EU730675	EU730680	MT890023	M678222–M678224*
CV38	-	AF324810	AF324809	-	-	-	-
CV125	-	AF324808	AF324807	-	-	-	-
Hrv1	RV/ SM/ SR	EU730649	EU730659	EU730669	EF540744	MT890024	M676174–M676182
Hrv3	RV/ MAC	-	MT890017	MT890018	MT890019	MT890025	M676150–M676173
H54	FA	EU730648	DQ120253	EU730668	-	-	M676139–M676149
-	RTL	-	-	-	-	-	M676127–M676138
-	RTL	-	-	-	-	-	M676288–M676288
-	MAB	-	-	-	-	-	M676118–M676126
-	MAB	-	-	-	-	-	M676108–M676117
-	MAB	-	-	-	-	-	M676098–M676107
H04	JAM/ PG	EU730644	EU730655	EU730664	EF540738	MT890026	M676083–M676093
H14	JAM/ PG	-	-	-	-	-	M676079–M676082
H28	JAM/ PG	EU730645	EU730656	EU730665	EF540740	MT890027	M676097
-	HHS/ HJG	-	-	-	-	-	M676057–M676078
-	HHS/ HJG	-	-	-	-	-	M676044–M676056
-	HHS/ HJG	-	-	-	-	-	M676030–M676043
-	HHS/ HJG	-	-	-	-	-	M676012–M676029
-	HHS/ HJG	-	-	-	-	-	M675997–M676011
-	HHS/ HJG	-	-	-	-	-	M675993–M675996

TABLE 1. (Continued)

| DNA code	Collector code	GenBank accession code					Morphobank code
		12S	**cyt *b* 1**	**cyt *b* 2**	**RAG2**	**MC1R**	
Hrv2	RV/ PAC/ JO	-	MT905068	-	-	MT890028	M675971–M675978
H30	JAM	EU730647	EU730658	EU730667	EU730684	MT890029	-
H29	JAM	EU730646	EU730657	EU730666	EU730683	-	-
H03	JAM/ PG	EU730639	EU730650	EU730660	EU730681	MT890030	-
H13	JAM/ PG	EU730640	EU730651	EU730660	EU730682	MT890031	-
-	JAM/ PG	-	-	-	-	-	-
H06/BEV.T5651	PAC	-	-	-	-	MT890032	M673362–M673366
H07/BEV.T5652	PAC	-	-	-	-	MT890033	M675961–M675962
H05	JAM/ PG	EU730641	EU730652	EU730661	EF540737	MT890034	M670156–M670160
H16	JAM/ PG	EU730642	EU730653	EU730662	EF540742	MT890035	M670148–M670155
H17	JAM/ PG	EU730643	EU730654	EU730663	EF540743	MT890036	M670161–M670163
-	JAM/ PG	-	-	-	-	-	M669539–M669541
-	GK	-	-	-	-	-	M585298–M585321
-	GK	-	-	-	-	-	M585325–M585346
-	GK	-	-	-	-	-	M585736–M585746
-	GK	-	-	-	-	-	M585747–M585754
Hd03	-	-	HQ833728	HQ833728	-	-	-
SPM002086	-	DQ120313	-	-	-	JQ957301	-
SPM003063	-	-	-	-	HQ676103	-	-
JS58	-	KC818702	KC818854	KC818854	-	KC818929	-
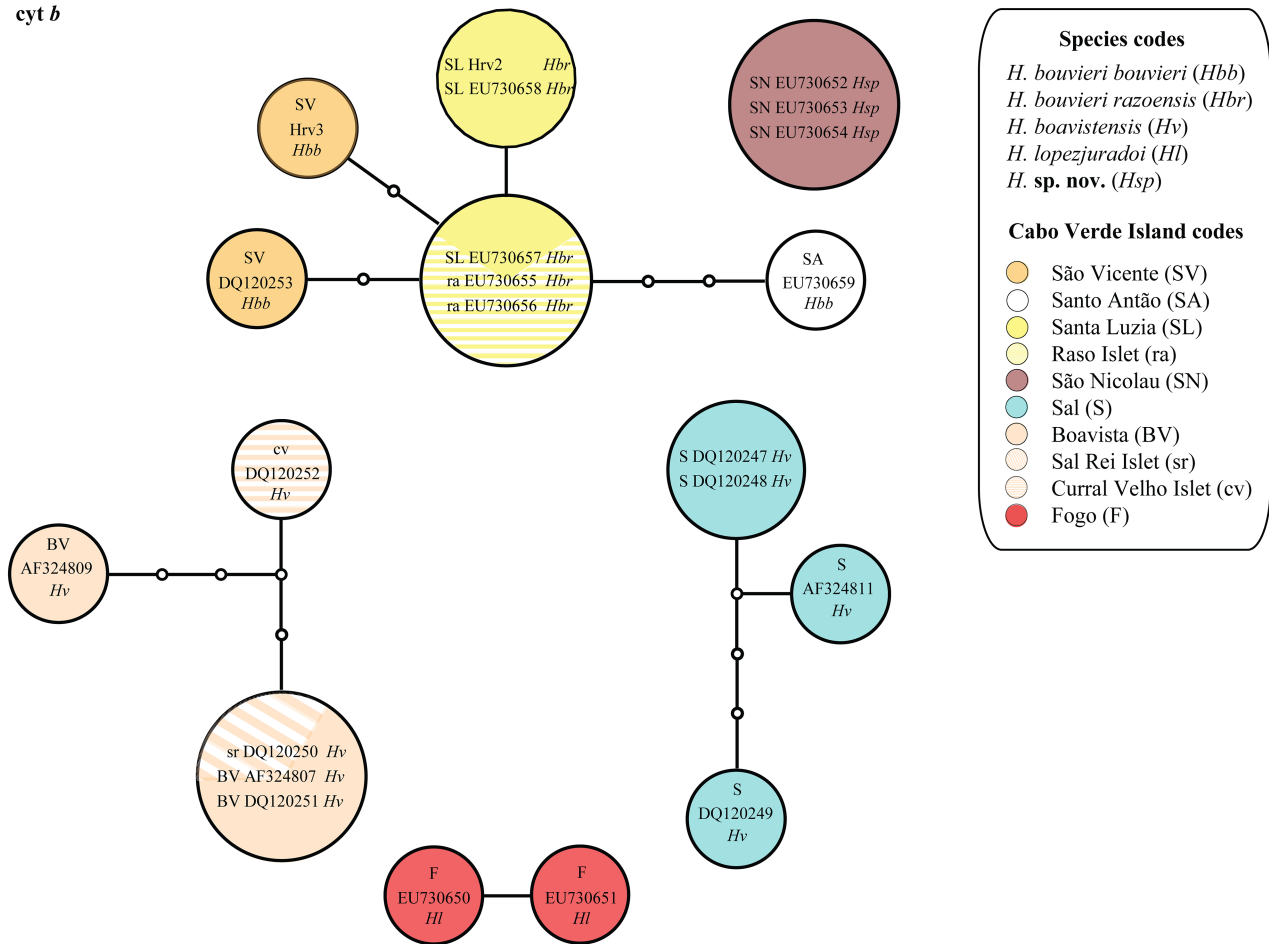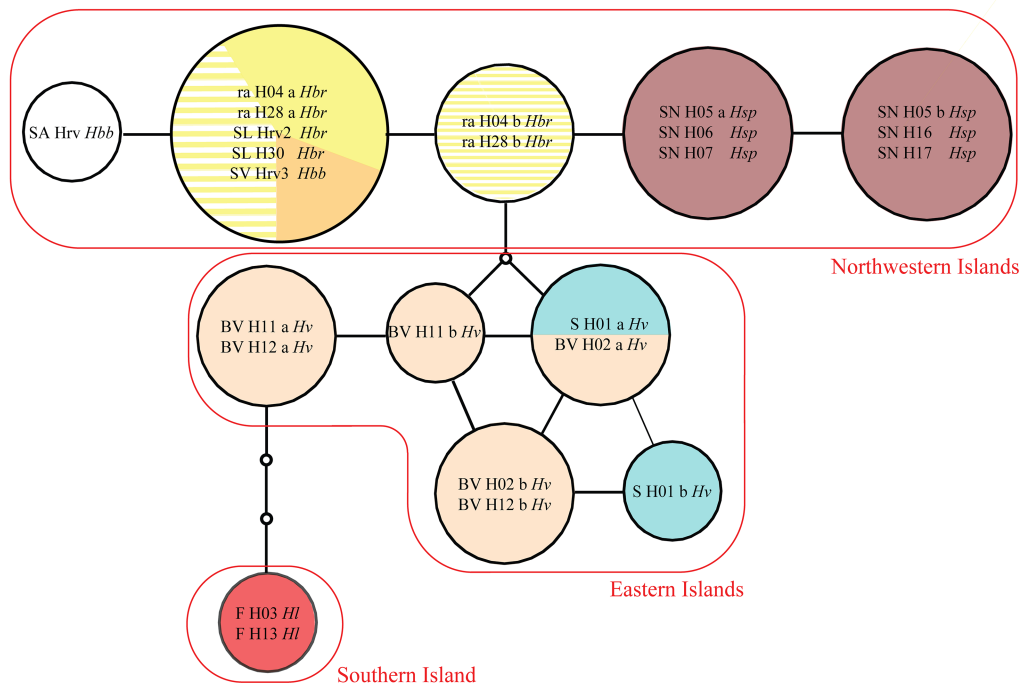
FIGURE 2. Parsimony networks corresponding to cyt *b* (mtDNA), and MC1R (nDNA) sequence variation. Lines represent mutational steps, circles haplotypes and dots unsampled haplotypes. The size of circles is proportional to the number of individuals. Samples from the same geographic location are similarly coloured. For correspondences of sample and location codes see Table 1.

Head colouration seems diagnostically different in the live specimens we have examined (colour photos of live specimens: six *H. nicolauensis* **sp. nov.** and five *H. bouvieri*; Fig. 7): the dark band that runs along the snout side in front of the eye is thinner and better defined in *H. bouvieri* (less well marked and/or thicker in *H. nicolauensis* **sp. nov.**) whereas the top of the head has many more dark elements in *H. nicolauensis* **sp. nov.**, resulting in a less clean and contrasted head pattern in all age classes (see Figs. 3, 7; Morphobank M585300 versus M676174). In addition, the dorsal colouration of *H. nicolauensis* **sp. nov.** is usually less contrasting and the dark bands across the dorsum do not reach as far down on the flanks as in *H. bouvieri* (Morphobank M585300 versus M676180; Fig. 7). In *H. bouvieri*, most individuals have a distinct pale yellowish background colouration with contrasting dark band on dorsum joining with a continuous dark band along the flanks. In *H. nicolauensis* **sp. nov.**, the colouration is never as contrasting as in most *H. bouvieri*, and most animals have shorter dark bands across dorsum, no continuous dark bands along flank, and a generally darker and less contrasting body and head colouration, although there is an overlap in body colouration between the two species (Morphobank M585300 versus M676180; Fig. 7).
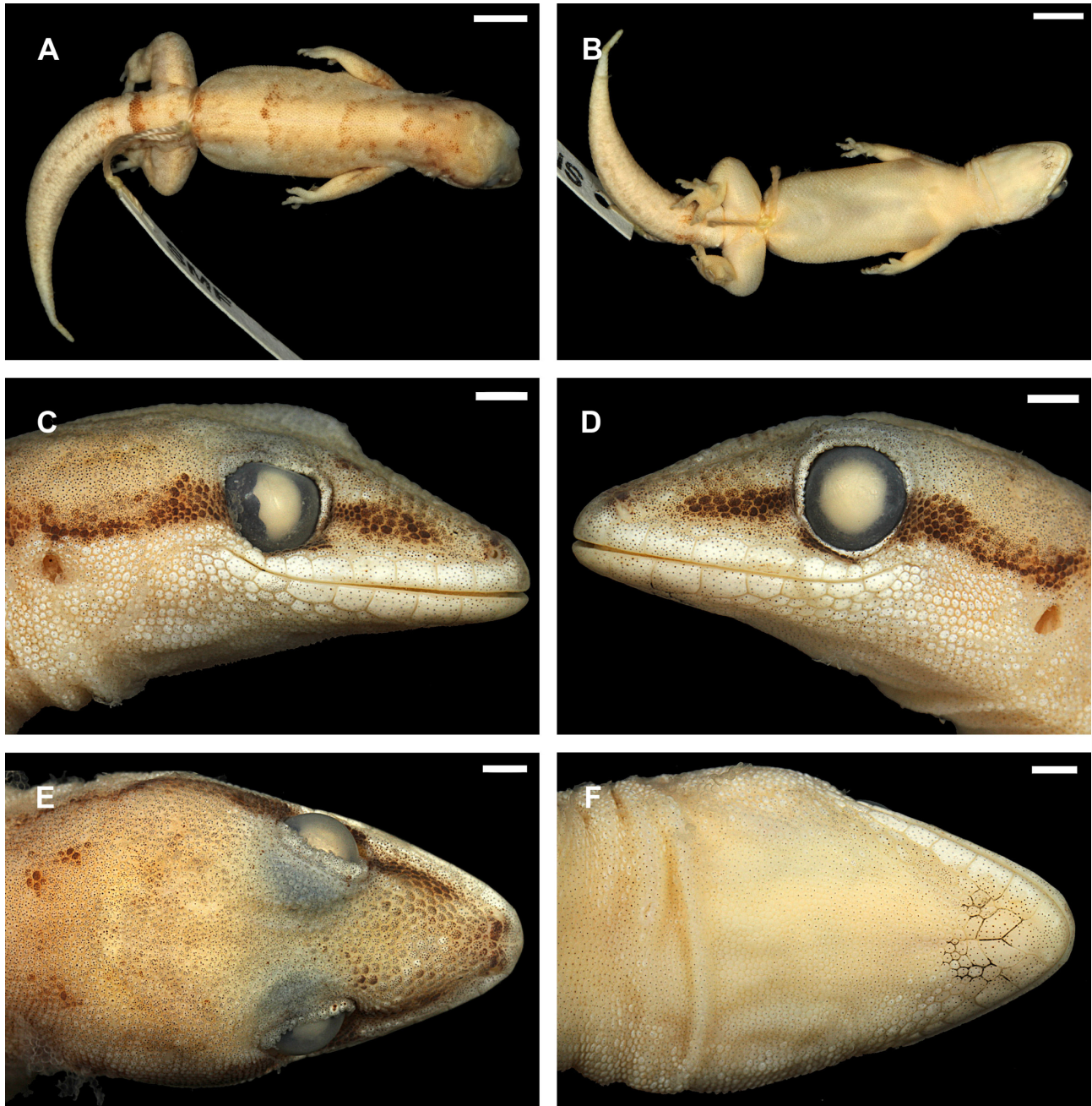

FIGURE 3. General and head views of the holotype of *Hemidactylus nicolauensis* (SMF 86992) from Cabo Verde. A) dorsal general view; B) ventral general view; C) right view of head; D) left view of head; E) dorsal view of head; F) ventral view of head. Scale bar = 5 mm in A and B, 1 mm in C to F.
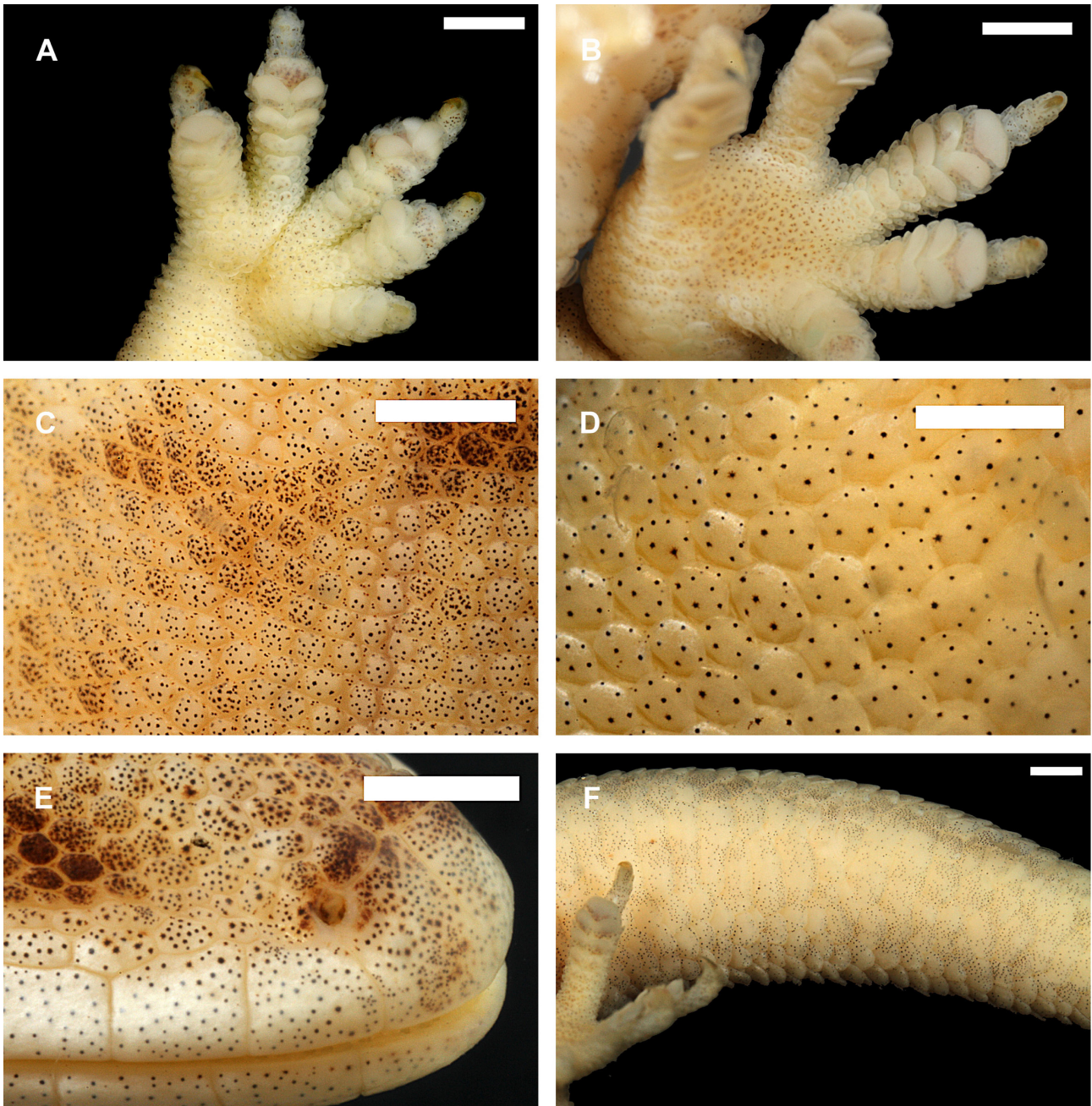
FIGURE 4. Scale details of the holotype of *Hemidactylus nicolauensis* (SMF 86992). A) Finger lamella; B) toe lamella; C) mid-dorsum scales; D) mid-venter scales; E) nasal scales; F) ventral tail scales. Scale bar = 1 mm.

Finally, head shape in most individuals differs between the two species, with *H. bouvieri* having a longer and more pointed snout than *H. nicolauensis* **sp. nov.**. Again, there is variation in this feature, as some *H. bouvieri* have a snout shape that is not clearly different from *H. nicolauensis* **sp. nov.** (see Figs. 3, 7 and Morphobank M676174 versus M585304).

Hemidactylus nicolauensis **sp. nov.** differs morphologically from *H. boavistensis* by its smaller size (the latter reaching up to 50 mm SVL), coarser and less numerous ventral scales (26 or 27 versus 35–38 across mid-belly in *H. boavistensis*), lower number of enlarged scales and digital lamellae under fourth hind toe (5–8 versus 7–9), and its narrower head. The two species have also a clearly different colouration (e.g.: Morphobank M585300 versus M678224; Fig. 7). Finally, *H. nicolauensis* **sp. nov.** differs morphologically from *H. lopezjuradoi* by its lower number of precloacal pores in males (one on each side versus three on each side in *H. lopezjuradoi*) and by the absence of the enlarged small tubercles on the back that are typical of *H. lopezjuradoi*.
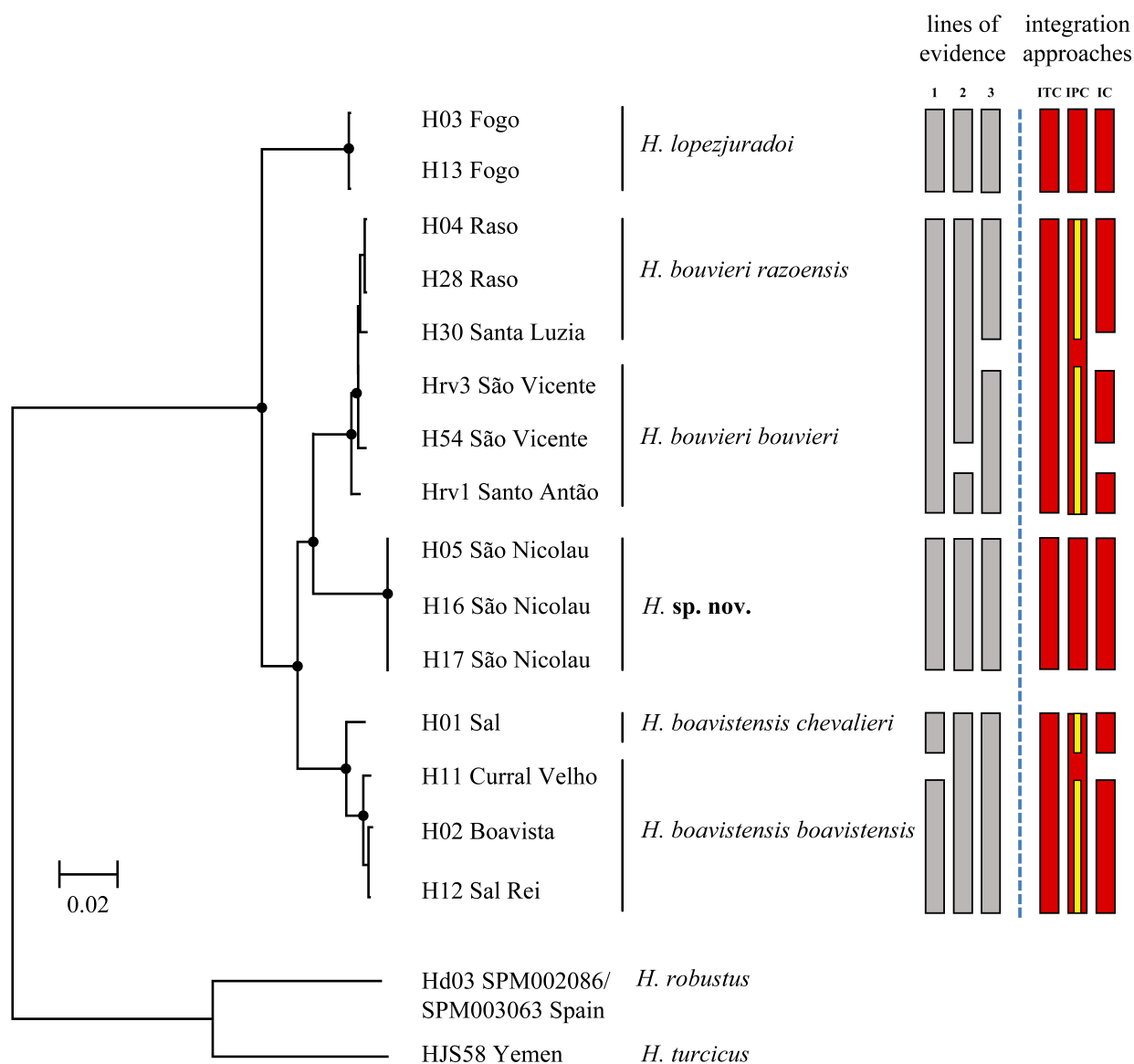
FIGURE 5. Phylogenetic tree with integrative taxonomy results. Relationships in endemic Cabo Verde *Hemidactylus* and their relatives based on a maximum-likelihood analysis of a 2524 base pairs (bp) concatenated alignment composed of two mitochondrial (669 bp of cyt *b* and approximately 380 bp of 12S rRNA) and two nuclear (668 bp of MC1R and 804 bp of RAG2) gene sequences. Black circles on top of nodes indicate bootstrap support for ML and Bayesian posterior probability values. Lines of evidence (in light grey): (1) Mitochondrial DNA (independent cyt *b* parsimony networks with a connection limit of 95%); (2) Nuclear DNA (absence of shared haplotypes in MC1R) and; (3) Morphology (detection of any diagnostic morphological character). Integration approaches (in red) from the most conservative to the most inflationist: ITC stands for an integration by total congruence (all lines of evidence should be congruent), IPC stands for integration by partial congruence which have been presently retained in this study (at least two lines of evidence are necessary); IC stands for an integration by cumulation (one line of evidence is sufficient). Species are represented in red bars, and for the IPC protocol, subspecies are represented within those bars in yellow. For fuller locality data and GenBank accession numbers see Table 1.

Description of the holotype and variation. The holotype is an adult male, 40.5 mm from tip of snout to cloaca, regenerated tail length 30 mm, eight supralabials on each side, seven (left side) and eight (right side) infralabials, two precloacal pores (one on each side) separated by a single scale, five enlarged scales and lamellae under second toe, six under third toe, seven under fourth hind toe and seven under fifth toe (Morphobank M585325–M585346).

Due to the poor state of conservation of the NHMUK specimens, the following description is mainly based on the four SMF and two BEV specimens. A small species, SVL varying between 33 and 41 mm in apparently adult

specimens (including a female carrying eggs and a male with well-developed precloacal pores). Tail long and thick, 70 to 80 % of the body length in the two specimens with complete tail, even thicker when regenerated. Long fore-limbs and hind limbs with five fingers and toes.

First finger and toe much smaller than other digits and without well-developed claw. Second and fifth fingers approximately same size, third finger longer, fourth finger the longest, all with a narrow tip carrying a claw projecting out of an enlarged middle part of the fingers. Hind foot and toes of same structure, but with fifth toe longer than second and toes with more developed tips and claws than fingers. Starting from the tip, the underside of each finger and toe has one large and elongated, but entire lamella, followed by 2–3 transversally elongated lamellae that are divided or centrally constricted, and 1–3 enlarged scales that are clearly larger than the other scales under hand and foot. The lamellae harbour tiny hair-like structures typical of many geckos. There are 3–5, 5, 5–6, 6 and 5–6 enlarged scales and lamellae under first to fifth finger and 3–4, 5, 5–6, 5–8 and 5–7 enlarged scales and lamellae under first to fifth toes.
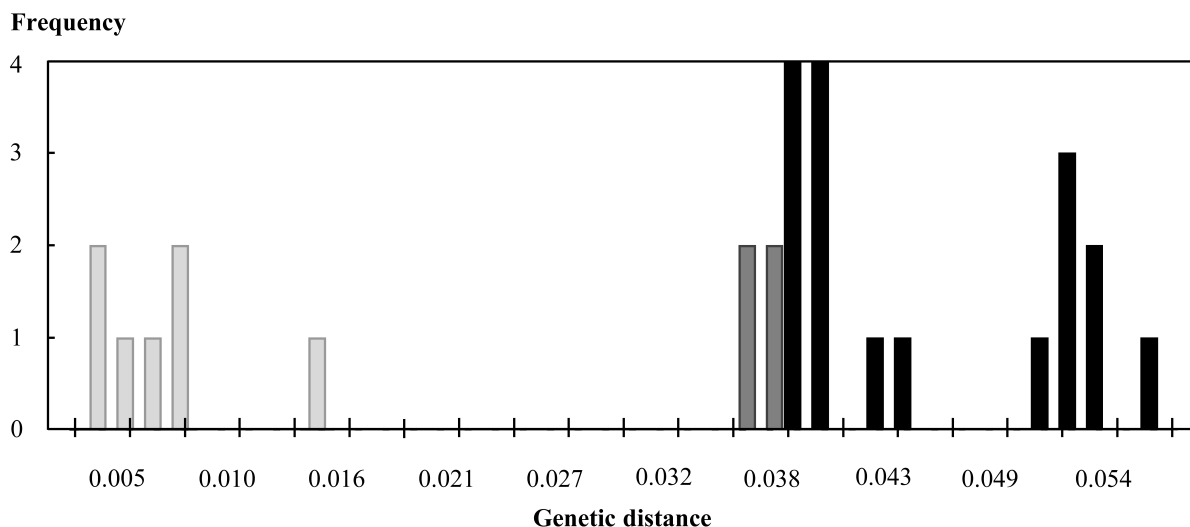
FIGURE 6. Pairwise genetic distances. Distribution of K2P distances of the concatenated dataset using the four markers between all pairs of samples for samples belonging to different species based on current taxonomy (black), to the same species based on current taxonomy (pale grey), and between samples of São Nicolau and other islands (dark grey).

Head elongated, snout long with straight or convex upper snout, hardly protruding small nostrils, large eyes with diameters larger than distance from eye to tip of snout. Distance from eye to snout equals distance from eye to ear opening. Ear opening very small (smaller than average supralabials), roundish. Supralabials and infralabials 7–8. Rostral large, reaching nostril and depressed in the middle as if it was semidivided. Three scales border the nostril in addition to the rostral. Mental scale triangular with rounded outer edge, approximately as broad as long, bordered by two postmental scales with a long suture. Three to four rows of enlarged scales border the postmental posteriorly. Rest of head, limbs and body scalation rather uniform, covered by small, round, flat scales, smaller on head and neck and even smaller in temporal area and on side of neck, 23–29 scales across top of head between the eyes (excluding superciliaries). No collar or gular fold. Ventrals gradually larger, triangular, broader than long, in 26 or 27 in longitudinal rows. Unregenerated tail (present in the two SMF juveniles) covered dorsally and laterally with larger flat rectangular scales, arranged in nearly regular tail rings; under the tail a single row of much larger scales, each tail covering most of the undertail width. Regenerated tail covered with larger scales arranged less regularly; undertail variable, covered of slightly larger scales with irregular disposition (hence without the single row of much larger scales of original tails). Both sexes have a few short blunt tubercles on each side of tail basis.

Colouration of live specimens available for one (adult female) SMF specimen, the two adult female BEV specimens and three of the NHMUK specimens. Body pale grey to brown, with five dark crossbands on back, the first one between forelimbs and the last one between hind limbs, larger than the intervening pale areas, often with irregular edges and/or invaded by pale elements; head same colour as body, with a dark patch on top of back of head, and various amount of dark elements on top of head, with a dark band running from the snout to the insertion of forelimbs, where it sometimes joins an irregular dark band running along flanks to tail basis, but this dark band

along flanks is often missing. Limbs and digits dark, about same colour as dark crossband on dorsum, often with scattered pale scales. Tail yellow to pale red, with dark irregular narrow crossbands when intact, and an even more irregular dark pattern when regenerated. Underparts pale, whitish to greyish, except undertail that is uniformly yellow to pale red. Lamellae and some enlarged scales under toes and fingers contrastingly paler.

TABLE 2. Estimates of genetic distances among endemic *Hemidactylus* populations and species (K2P distances). Values for the 12S mitochondrial marker are shown below the diagonal and for the two concatenated cyt *b* fragments above the diagonal. See Table 1 and material and methods for details.

		Hsp	*Hb*				*Hl*	*Hv*	
		SN	ra	SL	SV	SA	F	S	BV
H. sp. nov. (*Hsp*)	São Nicolau (SN)		0.117	0.118	0.119	0.117	0.187	0.144	0.139
H. bouvieri (*Hb*)	Raso (ra)	0.040		0.005	0.013	0.023	0.161	0.117	0.118
	Santa Luzia (SL)	0.037	0.003		0.011	0.021	0.165	0.118	0.117
	São Vicente (SV)	0.047	0.007	0.009		0.019	0.164	0.119	0.117
	Santo Antão (SA)	0.033	0.007	0.004	0.013		0.163	0.114	0.116
H. lopezjuradoi (*Hl*)	Fogo (F)	0.060	0.061	0.060	0.066	0.057		0.181	0.179
H. boavistensis (*Hv*)	Sal (S)	0.039	0.047	0.045	0.050	0.043	0.042		0.041
	Boavista (BV)	0.047	0.052	0.051	0.055	0.048	0.049	0.009	

TABLE 3. Morphological data. Mean, standard deviation and range (between squared brackets) of the morphological variables examined for *Hemidactylus* from São Nicolau and from other northwestern islands of Cabo Verde. The n stands for the maximum number of animals examined; SVL for snout-vent length; IL + SL, number of labials; VENL, number of enlarged ventral scale rows at midbody; SHT, number of scales between the eyes; and DTF and DTT, divided lamellae under third anterior and posterior digit, respectively.

	SVL	IL + SL	VENL	STH	DTF	DTT
H. b. bouvieri	33.9 ± 2.3	28.4 ± 2.6	23.0 ± 3.0	21.7 ± 3.0	0.75 ± 0.30	0.60 ± 0.23
(n=6)	[29.8–35.6]	[26–32]	[19–27]	[26–32]	[0–1]	[0.5–1.0]
H. b. razoensis	24.7 ± 4.7	27.3 ± 1.7	25.4 ± 3.6	26.6 ± 4.5	0.62 ± 0.44	0.78 ± 0.40
(n=9)	[16.5–29.0]	[25–30]	[22–32]	[21–32]	[0–1]	[0.5–1.0]
H. sp. nov.	31.5 ± 9.9	30.2 ± 1.2	26.5 ± 0.7	26.0 ± 2.6	2.28 ± 0.44	2.00 ± 0.55
(n=8)	[19.0–40.8]	[29–32]	[26–27]	[23–29]	[2–3]	[1.5–3.0]

Sexual dimorphism weakly developed, males have two well-developed precloacal pores (one on each side) situated in front of the cloaca and separated by a single scale.

Distribution. São Nicolau Island, Cabo Verde Archipelago. So far only known from three localities in two areas (central mountains around Cachaço and Ribeira Funda on the north coast, see Fig. 1 and Table 1), but doubtlessly far more widespread. Further research needed to establish its distribution range on São Nicolau.

Natural history. The rarity of these geckos makes their precise ecological requirements uncertain. Specimens have been found in the more humid areas of the island. Around Cachaço, the two localities where they have been found are situated at the foot of rocky outcrops, and, in one of them, the two animals were found in the most humid microhabitat, next to a spring with water dropping from the rock surface. In Ribeira Funda, the species has been collected in a narrow valley between cliffs and steep rocky slopes. These animals are supposed to be mainly nocturnal, and all specimens have been found inactive by day under stones or in rock crevices. In one spot, two females were found together (the two BEV specimens). One female from Ribeira Funda collected on 1 April 2007 (UCV2018/004 or former SMF 86991) carried a single, large, well visible egg.

Conservation status. Not Evaluated. All native reptiles in Cabo Verde are endemic to these islands, although this is not widely-recognized among the public and policy makers. In addition, geckos are generally feared due to traditional beliefs, and there may be occasional persecution (Vasconcelos *et al.* 2013). Education is therefore needed to raise awareness of these species and of their conservation requirements and stressing that these animals are inoffensive to humans. The existing conservation regulations should also be updated to reflect changes in both

taxonomy and conservation status (Vasconcelos 2013b). There is very little information on the population status and ecology of *H. nicolauensis* **sp. nov.**, so the precise impacts of identified and potential threats require further research. For *H. bouvieri, H. nicolauensis* **sp. nov.**, and *H. lopezjuradoi*, the paucity of records both historically and recently, in spite of active research on all the islands, indicates low population densities or severe fragmentation of the occupied range (rather than an extraordinary low detectability) and suggests a marked population decline since human colonization of the islands (Arnold *et al.* 2008).

Hemidactylus boavistensis Boulenger, 1906

Referred material. Morphobank M678219–M678224; M676396– M676398

Diagnosis. Relatively large animals, up to 50 mm from snout to vent with relatively broad head posteriorly and narrow snout with concave or straight upper profile (Arnold *et al.* 2008). No enlarged tubercles on the dorsum and small ventral scales and often longer than wide and about 35–40 across mid-belly (Arnold *et al.* 2008). Usually five enlarged scales under first hind toe and 7–9 under fourth, 5–7 under fifth (Arnold *et al.* 2008). Mental often narrowed posteriorly, and postmentals frequently longer than wide (Arnold *et al.* 2008). Two large precloacal pores in males and regular and expanded laterally subcaudal scales (Arnold *et al.* 2008). The dorsal pattern often consisting of broad transverse bands, but these may be divided on the midline, or the anterior ones broken in to several sections, or animals may be more uniform without bands (Arnold *et al.* 2008).

Distribution. Sal and Boavista islands, Cabo Verde Archipelago.

Conservation status. Classified as Near Threatened in the IUCN Red List on the basis that this species, although restricted to two islands and two islets in Cabo Verde, is still abundant despite ongoing pressures from infrastructure development and introduced species (Vasconcelos 2013a). The species almost meets the requirements for listing as threatened under the criterion of reduced geographic range (reduced number of locations, area of extent and estimated number of mature individuals), situation which needs monitoring (Vasconcelos 2013a). Measures are required to ensure the effective protection of this species.

Hemidactylus boavistensis boavistensis **comb. nov.** Boulenger, 1906

Name-bearing types. The description is based on several specimens from Boavista, from sea level to 600 m high (although the maximum elevation of Boavista is circa 360 m) collected by L. Fea and sent to the NHMUK. The following six specimens in the NHMUK collection were all collected by Fea on Boavista and are thus all syntypes: NHMUK 1946.8.25.68-73.

Type locality. Boavista Island, Cabo Verde Archipelago.

Diagnosis. Very similar to *H. b. chevalieri*, but usually with four to five lamellas under the first finger according to Angel (1935). Angel (1935) also claims that the transverse bands on the head and nape are usually more marked, but we found this to be highly variable and of little use for identification (see Fig. 7). Even though dorsal colouration is highly variable, Boavista individuals are usually lighter than Sal ones.

Distribution. Boavista Island and adjacent islets.

Conservation status. Not Evaluated. Threatened by ongoing touristic development on Boavista such as beach resorts, as the favourable habitat of this gecko is coastal areas with sand and dunes, and it seems avoiding urban and heavily-developed areas (Vasconcelos 2013a). Other authors (López-Jurado *et al.* 1999) reported disappearance of this gecko in areas where the introduced house gecko *Hemidactylus angulatus* occurs. This exotic gecko is widespreading and now it also occurs, apart from anthropogenic habitats, in natural areas on Boavista, posing a threat to the endemic *Hemidactylus* (Vasconcelos *et al.* 2013). In addition, climate change may worsen the ongoing drought conditions in this island, negatively affecting Boavista leaf-toed geckos (Vasconcelos 2013a).
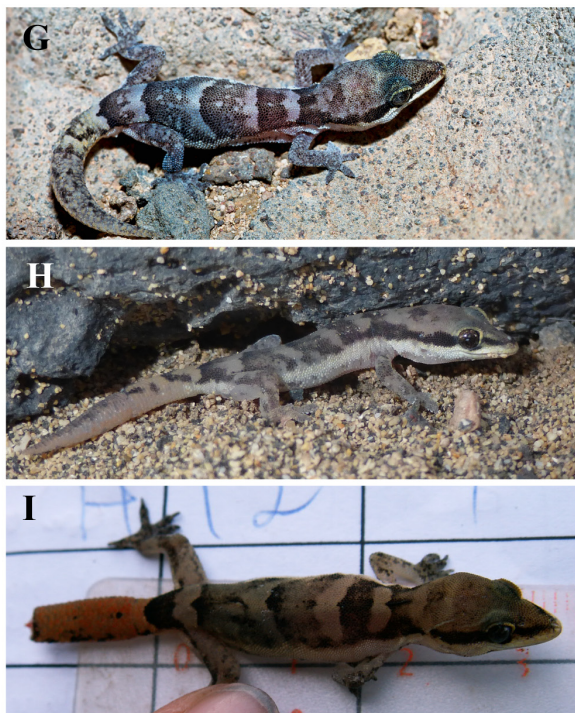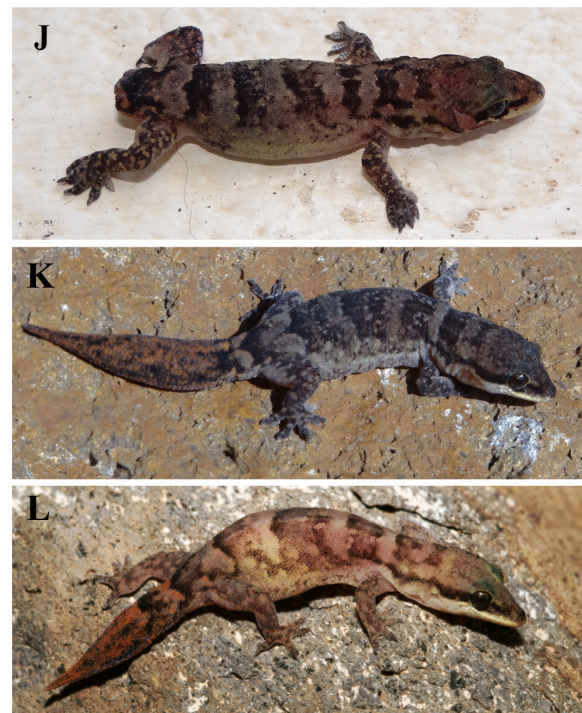
H. boavistensis boavistensis
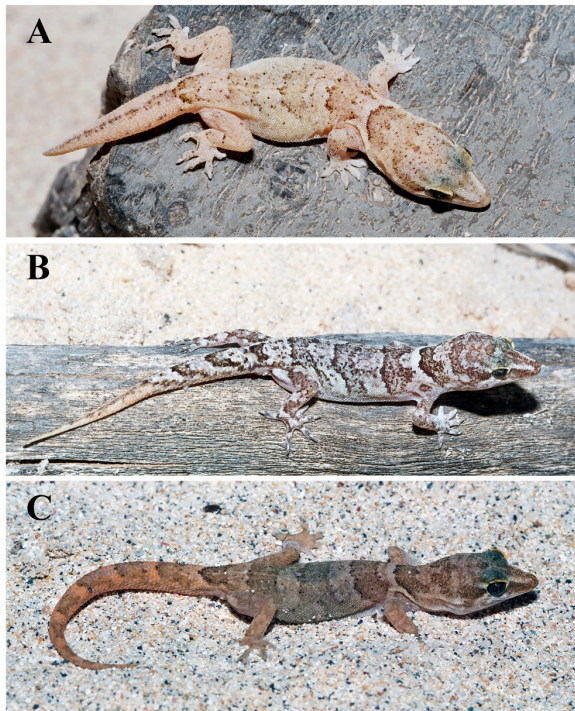
H. boavistensis chevalieri
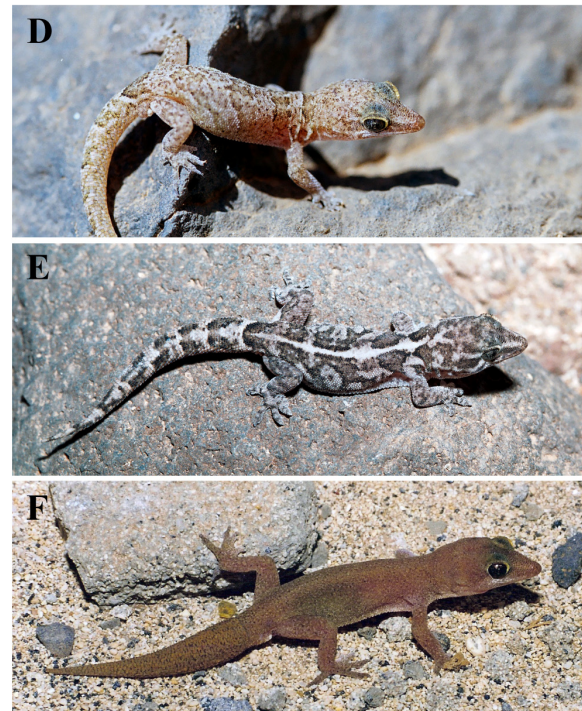
H. bouvieri

H. nicolauensis

FIGURE 7. Photos of live specimens of *Hemidactylus boavistensis boavistensis* (top left, A–C) versus *H. b. chevalieri* (top right, D–F), and *H. bouvieri* (bottom left, G–I) versus *H. nicolauensis* (bottom right, J–L). All photos by PG, GK, RV and PAC. A) Adult female from 1 km E of Sal Rei, Boavista (PGe.1479); B) adult male from 3 km E of Sal Rei, Boavista (PGe.1485); C) subadult female from 1 km E of Sal Rei, Boavista (PGe.1481); D) adult from 1 km SE Santa Maria, Sal (PGe.1499); E) adult from Buracona, Sal (PGe.1507); F) adult from 1 km ESE Santa Maria, Sal (PGe.1500); G) adult from Chã do Castelo, Raso (H28; PGe.1476); H) adult from Praia Grande, Calhau, São Vicente (Hrv3); I) adult from Lombo de Diogo, Santo Antão (Hrv1); J) adult female from Cachaço, São Nicolau (H06; BEV.T5651); K) adult female from Cachaço, São Nicolau (H07; BEV.T5652); L) adult female from Ribeira Funda, São Nicolau (UCV2018/004, paratype).

Hemidactylus boavistensis chevalieri comb. nov. Angel, 1935

Name-bearing types. Thirteen syntypes of which 12 are still present in the MNHN collections : MNHN-RA-1935.178, 179, 179A, 179B, 180, 180A, 181, 181A, 182, 183, 184 & 184A. Based on the information in the MNHN catalogue, the two beforelast come from 'Santa Maria' and the rest from an unkown locality on 'Sal'.

   Type locality. Santa Maria, Sal Island & unkown locality, Sal Island, Cabo Verde Archipelago.

   Diagnosis. Very similar to *H. b. boavistensis* but usually with six lamellas under the first finger according to Angel (1935). Angel (1935) also claims that head and nape colouration is generally also different as regards to the transverse bands, which are usually lacking or less clear than in the nominotypical subspecies (see Fig. 7), but we found this to be highly variable and of little use for identification. Even though dorsal colouration is highly variable, Sal individuals are usually darker than Boavista ones.

   Distribution. Sal Island, Cabo Verde Archipelago.

   Conservation status. Not Evaluated. Identical threats as referred above.

Discussion

We confirmed here previous results showing that *Hemidactylus nicolauensis* **sp. nov.** is genetically distinct from *H. bouvieri* (Figs. 2, 5). The lack of shared haplotypes in mitochondrial and MC1R nuclear genes (Fig. 2) indicates that gene flow was reduced between the two species. This was expected given the distance between São Nicolau and the Desertas (15 km; Fig. 1) and the current sea depths between them (more than 400 m along any possible path). São Nicolau started to form as a result of volcanic activity around 6 million years ago (Duprat *et al.* 2007) and sea level never varied enough to connect it with other islands after its formation (Fernández-Palacios 2016; Triantis *et al.* 2016). This fit nicely with the inferred timing of divergence between *H. bouvieri* and *H. nicolauensis* **sp. nov.** (4.7 ± 1.2 million years ago in Arnold *et al.* 2008) suggesting a colonisation of São Nicolau soon after its origin and subsequent genetic isolation.

   Differentiation between these species is also supported by morphology, although differences are subtle. Considering that *H. nicolauensis* **sp. nov.** and *H. bouvieri* occupy the same general niche, occurring in similar habitats and microhabitats on different islands where no other congeneric species naturally occurs, it is no surprise that phenotypic differences are not very marked between them.

   It was previously thought that *H. bouvieri bouvieri* might be restricted to humid, high montane areas between 600–700 m in elevation, and that *H. b. razoensis* may differ in their ecological requirements (Arnold *et al.* 2008). However, *H. bouvieri bouvieri* was recently found on São Vicente at sea-level, on a beach with some small bushes and scattered stones near the coast (R. Vasconcelos pers. obs.). *Hemidactylys b. razoensis* exhibits similar habitat preferences on Santa Luzia to the ones described for the nominal form, as it was found near the top of the island in the most vegetated and humid areas (J.A. Mateo pers. com., R. Vasconcelos & P.A. Crochet pers. obs.). But on Raso, a much arid, flatter, and less-vegetated islet, it occurs along dry streams in inland areas with dense vegetation, and uses cavities in rock or tree roots, or stonewall ruins as shelter sites (J.A. Mateo & P. Geniez pers. obs., Gruber & Schleich 1982). So, further research will be needed to evaluate the ecology, habitat requirements and conservation status of *H. nicolauensis* **sp. nov.**. The study of its diet, using DNA metabarcoding or isotope analyses, would probably give us some insight on this (Martín *et al.* 2017; Pinho *et al.* 2018).

   Regarding the two subspecies of *H. boavistensis*, a little more ecological information exists. The species is known to be abundant in very arid, open areas with little vegetation (Arnold *et al.* 2008), although it is absent from hyperarid areas in the south of Boavista (López-Jurado *et al.* 1999). Animals shelter under rock and rock piles, in sparse dune vegetation or fallen palm trees (Schleich 1987). They occur in large groups, and lay eggs in communal nests. However, more information and monitoring are needed to set conservation action plans on each island, such as evaluating the extent of threat by exotic species (Vasconcelos *et al.* 2013).

Acknowledgements

References

Andreone, F. (2000) Herpetological observations on Cape Verde: a tribute to the Italian naturalist LEONARD FEA, with complementary notes on *Macroscincus coctei* (Duméril & Bibron, 1839). *Herpetozoa*, 13, 15–26.

Angel, F. (1935) Lézards des Îles du Cap Vert, rapportés par M. le Professeur Chevalier. Description de espèces nouvelles. *Bulletin du Muséum national d'Histoire naturelle, Paris*, 2, 165–169.

Arnold, E.N., Vasconcelos, R., Harris, D.J., Mateo, J.A. & Carranza, S. (2008) Systematics, biogeography and evolution of the endemic *Hemidactylus* geckos (Reptilia, Squamata, Gekkonidae) of the Cape Verde Islands: based on morphology and mitochondrial and nuclear DNA sequences. *Zoologica Scripta*, 37, 619–636.
https://doi.org/10.1111/j.1463-6409.2008.00351.x

Bauer, A.M., Jackman, T.R., Greenbaum, E., Giri, V.B. & de Silva, A. (2010) South Asia supports a major endemic radiation of *Hemidactylus* geckos. *Molecular Phylogenetics and Evolution*, 57, 343–352.
https://doi.org/10.1016/j.ympev.2010.06.014

Bocage, J.V.B. (1896) Reptis de algumas possessões portuguezas d' África que existem no museu de Lisboa. *Jornal de Sciencias Mathematicas, Physicas e Naturaes* 2, 1–9, estampas I e II.

Boulenger, G.A. (1906) Report on the Reptiles collected by the late L. Fea in West Africa. *Annali del Museo Civico di Storia Naturale di Genova*, II, 196–216.

Carranza, S. & Arnold, E.N. (2006) Systematics, biogeography, and evolution of *Hemidactylus* geckos (Reptilia: Gekkonidae) elucidated using mitochondrial DNA sequences. *Molecular Phylogenetics and Evolution*, 38, 531–545.
https://doi.org/10.1016/j.ympev.2005.07.012

Clement, M., Derington, J., Woolley, S. & Posada, D. (2005) Tcs 1.21. *Young*, 21, 1–8.

Duprat, H.I., Friis, J., Holm, P.M., Grandvuinet, T. & Sørensen, R.V. (2007) The volcanic and geochemical development of São Nicolau, Cape Verde Islands: Constraints from field and 40Ar/39Ar evidence. *Journal of Volcanology and Geothermal Research*, 162, 1–19.
https://doi.org/10.1016/j.jvolgeores.2007.01.001

Fernández-Palacios, J.M. (2016) Island biogeography: Shaped by sea-level shifts. *Nature*, 532, 42–43.
https://doi.org/10.1038/nature17880

Gruber, H.-J. & Schleich, H.-H. (1982) *Hemidactylus bouvieri razoensis* nov. ssp. von den Kapverdischen Inseln. *Spixiana* 5, 303–310.

Helbig, A.J., Knox, A.G., Parkin, D.T., Sangster, G. & Collinson, M. (2002) Guidelines for assigning species rank. *Ibis*, 144, 518–525.
https://doi.org/10.1046/j.1474-919X.2002.00091.x

Katoh, K. & Toh, H. (2008) Recent developments in the MAFFT multiple sequence alignment program. *Briefings in Bioinformatics*, 9, 286–298.
https://doi.org/10.1093/bib/bbn013

Köhler, G., Hertz, A., Sunyer, J., Seipp, R. & Monteiro, A. (2007) Herpetologische Forschungen auf den Kapverden unter besonderer Berücksichtigung des Kapverdischen Riesenskinks, *Macroscincus coctei*. *Elaphe*, 15, 75–79.

Librado, P. & Rozas, J. (2009) DnaSP v5: A software for comprehensive analysis of DNA polymorphism data. *Bioinformatics*, 25, 1451–1452.
https://doi.org/10.1093/bioinformatics/btp187

López-Jurado, L.F., Mateo, J.A. & Geniez, P. (1999) Los reptiles de la isla de Boavista (archipiélagode Cabo Verde). *Boletín de la Asociación Herpetológica Española*, 10, 10–13.

Loveridge, A. (1947) Revision of the African lizards of the family Gekkonidae. *Bulletin of the Museum of Comparative Zoology*, 98, 1–469.

Martín, N., Martínez, S., Pujol-Buxó, E., Viñolas, A., Llorente, G.A., Sanpera, C., Vasconcelos, R., Carranza, S. & Santos, X. (2017) Stable isotopes and diet uncover trophic-niche divergence and ecological diversification processes of endemic reptiles on Socotra Island. *Zoologischer Anzeiger*, 267, 69–81.
https://doi.org/10.1016/j.jcz.2017.01.005

Mateo, J.A., Gárcia-Marques, M., López-Jurado, L.F. & Pether, J. (1997) Nuevas observaciones herpetologicas en las Islas Desertas (Archipiélago de Cabo Verde). *Boletín de la Asociación Herpetológica Española*, 8, 8–11.

Mertens, R. (1954) Die Eidechsen der Kapverden. *Societas Scientarum Fennica Commentationes Biologicae*, 15, 1–17.

Miralles, A., Vasconcelos, R., Perera, A., Harris, D.J. & Carranza, S. (2011) An integrative taxonomic revision of the Cape Verdean skinks (Squamata, Scincidae). *Zoologica Scripta* 40, 16–44.
https://doi.org/10.1111/j.1463-6409.2010.00453.x

Pinho, C., Santos, B., Mata, V., Seguro, M., Romeiras, M., Lopes, R. & Vasconcelos, R. (2018) What Is the Giant Wall Gecko Having for Dinner? Conservation Genetics for Guiding Reserve Management in Cabo Verde. *Genes*, 9, 599.
https://doi.org/10.3390/genes9120599

Rato, C., Carranza, S. & Harris, D.J. (2011) When selection deceives phylogeographic interpretation: The case of the Mediterranean house gecko, *Hemidactylus turcicus* (Linnaeus, 1758). *Molecular Phylogenetics and Evolution*, 58, 365–373.
https://doi.org/10.1016/j.ympev.2010.12.004

Ronquist, F., Teslenko, M., Van Der Mark, P., Ayres, D.L., Darling, A., Höhna, S., Larget, B., Liu, L., Suchard, M.A. & Huelsenbeck, J.P. (2012) Mrbayes 3.2: Efficient bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology*, 61 (3), 539–542.
https://doi.org/10.1093/sysbio/sys029

Schleich, H.-H. (1987) Herpetofauna Caboverdiana. *Spixian,a* 12, 1–75.

Šmíd, J., Moravec, J., Kratochvíl, L., Nasher, A.K., Mazuch, T., Gvoždík, V. & Carranza, S. (2015) Multilocus phylogeny and taxonomic revision of the *Hemidactylus robustus* species group (Reptilia, Gekkonidae) with descriptions of three new species from Yemen and Ethiopia. *Systematics and Biodiversity*, 13, 346–368.
https://doi.org/10.1080/14772000.2014.996264

Tamura, K., Stecher, G., Peterson, D., Filipski, A. & Kumar, S. (2013) MEGA6: Molecular evolutionary genetics analysis version 6.0. *Molecular Biology and Evolution*, 30, 2725–2729.
https://doi.org/10.1093/molbev/mst197

Templeton, A.R., Crandall, K.A. & Sing, C.F. (1992) A cladistic analysis of phenotypic associations with haplotypes inferred from restriction endonuclease mapping and DNA sequence data. III. Cladogram estimation. *Genetics* 132, 619–633.

Triantis, K.A., Whittaker, R.J., Fernández-Palacios, J.M. & Geist, D.J. (2016) Oceanic archipelagos: a perspective on the geodynamics and biogeography of the World ' s smallest biotic provinces. *Frontiers of Biogeography*, 8.2, e29605.
https://doi.org/10.21425/F5FBG29605

Uetz, P., Freed, P. & Hošek, J. (2019) The Reptile Database. Available from: http://www.reptile-database.org/ (accessed 17 October 2017).

Vasconcelos, R. (2013a) *Hemidactylus boavistensis*. *The IUCN Red List of Threatened Species*, 2013, e.T18486000A18486039.
https://doi.org/10.2305/IUCN.UK.2013-1.RLTS.T18486000A18486039.en

Vasconcelos, R. (2013b) *Hemidactylus bouvieri*. *The IUCN Red List of Threatened Species*, 2013, e.T203840A2771787.
https://doi.org/10.2305/IUCN.UK.2013-1.RLTS.T203840A2771787.en

Vasconcelos, R., Brito, J.C., Carranza, S. & Harris, D.J. (2013) Review of the distribution and conservation status of the terrestrial reptiles of the Cape Verde Islands. *Oryx*, 47, 77–87.
https://doi.org/10.1017/S0030605311001438

Vasconcelos, R., Carranza, S. & James Harris, D. (2010) Insight into an island radiation: the *Tarentola* geckos of the Cape Verde archipelago. *Journal of Biogeography*, 37, 1047–1060.
https://doi.org/10.1111/j.1365-2699.2009.02254.x

Vasconcelos, R., Perera, A., Geniez, P., Harris, D.J. & Carranza, S. (2012) An integrative taxonomic revision of the *Tarentola* geckos (Squamata, Phyllodactylidae) of the Cape Verde Islands. *Zoological Journal of the Linnean Society*, 164, 328–360.
https://doi.org/10.1111/j.1096-3642.2011.00768.x