

Arrival and diversification of mabuyine skinks (Squamata: Scincidae) in the Neotropics based on a fossil-calibrated timetree

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Background: The evolution of South American Mabuyinae skinks holds significant biogeographic interest because its sister lineage is distributed across the African continent and adjacent islands. Moreover, at least one insular species, *Trachylepis atlantica*, has independently reached the New World through transoceanic dispersal. To clarify the evolutionary history of both Neotropical lineages, this study aimed to infer an updated timescale using the largest species and gene sampling dataset ever assembled for this group. By extending the analysis to the Scincidae family, we could employ fossil information to estimate mabuyinae divergence times and carried out a formal statistical biogeography analysis. To unveil macroevolutionary patterns, we also inferred diversification rates for this lineage and evaluated whether the colonization of South American continent significantly altered the mode of Mabuyinae evolution.

Methods: A time-calibrated phylogeny was inferred under the Bayesian framework employing fossil information. This timetree was used to (i) evaluate the historical biogeography of mabuyines using the statistical approach implemented in BioGeoBEARS; (ii) estimate macroevolutionary diversification rates of the South American Mabuyinae lineages and the patterns of evolution of selected traits, namely, the mode of reproduction, body mass and snout-vent length; (iii) test the hypothesis of differential macroevolutionary patterns in South American lineages in BAMM and GeoSSE; and (iv) re-evaluate the ancestral state of the mode of reproduction of mabuyines.

Results: Our results corroborated the hypothesis that the occupation of the South American continent by Mabuyinae consisted of two independent dispersion events that occurred between the Oligocene and the Miocene. We found significant differences in speciation rates between the New World and the remaining Mabuyinae clades only in GeoSSE. The influence of phenotypic traits on diversification rates was not supported by any method. Ancestral state reconstruction suggested that the ancestor of South American mabuyine was likely viviparous.

Discussion: Our analyses further corroborated the existence of a transoceanic connection between Africa and South America in the Eocene/Oligocene period (Atlantogea). Following colonization of the isolated South America and subsequent dispersal through the continent by the ancestral mabuyine stock, we detected no difference in macroevolutionary regimes of New World clades. This finding argued against the ecological opportunity model as an explanation for the diversity of living mabuyines.

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Abstract

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51 World clades. This finding argued against the ecological opportunity model as an explanation for
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Introduction

The subfamily Mabuyinae comprises 22 genera and approximately 190 species of lizards and belongs to the highly diverse and worldwide spread Scincidae family (Lepidosauria; Squamata) (Uetz and Hosek, 2015). The Mabuyinae, as well as the Scincidae, are distributed on nearly all continents. Approximately one-third of the species in this subfamily are from the Neotropical region, and they are the only representatives of scincids in this area.

The entire diversity of mabuyine species was traditionally assigned to the single genus, *Mabuya*, but a previous analysis proposed four new monophyletic genera with well-defined geographical distributions (Mausfeld *et al.*, 2002): (1) *Trachylepis* (previously *Euprepis*), comprising African and Madagascan species with one South American representative (*T. atlantica*); (2) *Eutropis*, containing Asian species; (3) *Chioninia*, from the Cape Verde islands; and (4) *Mabuya*, containing New World species, recently rearranged into 16 new genera by Hedges and Conn (2012). Hedges and Conn (2012) treated South American Mabuyinae as a clade of family Mabuyidae, within the superfamily Lygosomoidea. The new Mabuyidae consisted of four new subfamilies: Mabuyinae, Chioniniinae, Dasiinae and Trachylepidinae. Hedges and Conn's arrangement disregarded many genera previously related to Mabuyinae, such as *Eutropis*, *Lankaskincus*, and *Ristella* (Pyrón *et al.*, 2013), and recent papers have questioned this classification (e.g. Pyron *et al.*, 2013, Pinto *et al.*, 2015, Karin *et al.*, 2016). Recently, alternative classifications were suggested. For instance, Karin *et al.* (2016) placed the Middle-Eastern *Trachylepis* (*T. aurata*, *T. vittatus* and *T. septemtaeniatus*) into the genus *Heremites* and *Eutropis novemcarinata* into *Toenayar novemcarinata*. Moreover, Pinto *et al.* (2015) assigned species from the genera *Maracaiba* and *Alinea* back to genus *Mabuya* and Metallinou *et al.*

(2016) reclassified *Trachylepis ivensii* as *Lubuya ivensii*. In this study, we followed the Reptile Database (Uetz and Hosek, 2015) taxonomy as of January 2017, which classified this clade as subfamily Mabuyinae of the family Scincidae.

Therefore, at least two phylogenetically distinct lineages of Mabuyinae are distributed in the New World, namely, *T. atlantica* and the Continental American Mabuyinae (CAM) clade. These lineages are distinguished by both morphological features – presacral vertebrae counts, keeled dorsal scales, coloration, and oviparity (Greer, 1970) – and molecular evidence (e.g. Mausfeld and Vrcibradic, 2002; Carranza and Arnold, 2003; Whiting *et al.*, 2006). It is customary to assume that the history of Mabuyinae in South American continent consisted of two independent transoceanic dispersal events from the Old World (Mausfeld and Vrcibradic, 2002; Carranza and Arnold, 2003; Whiting *et al.*, 2006). The spatial distribution of the single representative of the genus *Trachylepis* in an island closer to South America than to Africa, *T. atlantica*, is of particular interest. This issue is so intriguing that it has reached the pages of nontechnical literature (de Queiroz, 2014). *T. atlantica* is found in Fernando de Noronha, a small volcanic archipelago in the Atlantic Ocean that lies 375 km from the northeastern coast of Brazil and that was geologically formed from the Miocene (12.3 Ma) to the upper Pliocene, from 3.3 – 1.7 Ma (Almeida, 2002). Although other *Trachylepis* species have spread to several islands near the African continent, the presence of *T. atlantica* in Fernando de Noronha likely represents the farthest dispersal registered for the genus.

The CAM lineage, on the other hand, contains approximately 60 species of Mabuyinae. Previous studies have suggested that the split between this clade and the African Mabuyinae (genus *Trachylepis*) occurred from 28 – 34 Ma (Hedges and Conn, 2012; Karin *et al.*, 2016). Additionally, the age of the last common ancestor (LCA) of the CAM was dated at 7 – 9 Ma

(Carranza and Arnold, 2003; Pinto-Sanchez et al., 2015) and 11 – 14 Ma (Miralles and Carranza, 2010; Hedges and Conn, 2012; Karin et al., 2016). Considering the present-day Atlantic Ocean currents, Mausfeld *et al.* (2002) suggested that *T. atlantica* could have dispersed from the coast of Southwest Africa to South America, but no work to date has comprehensively evaluated this hypothesis. The older estimated ages could be consistent with the supposed faunal transoceanic connection between Africa and South America during the Eocene/Oligocene (e.g. hystricognath rodents, Loss-Oliveira *et al.*, 2012; Voloch *et al.*, 2013; anthropoid primates, Schrago *et al.*, 2012; 2013; amphisbaenians, Vidal *et al.*, 2008; emballonurid bats, Teeling, 2005; Leigh *et al.*, 2013; testudinid turtles, Le *et al.*, 2006), through a single or a series of islands constituting an Atlantic Ocean Ridge (the Atlantogea paleo province) (Simpson, 1950; Poux *et al.*, 2006; de Oliveira *et al.*, 2009; Ezcurra and Agnolin, 2012; de Queiroz, 2014). This transatlantic island corridor, associated with a drop in the sea level in the Oligocene, could explain faunal exchanges in this period (de Queiroz, 2014). Regarding *T. atlantica*, there is a lack of estimates of the age of the separation between this species and its African sister lineage.

Chronological information is indispensable for a full understanding of the scenario underlying the current geographic distribution and evolutionary history of extant lineages (Sanmartín *et al.* 2008; Loss-Oliveira *et al.* 2012). However, estimates of divergence times within the Mabuyinae have been hampered by the lack of fossils for Mabuyinae, which made previous researchers rely on evolutionary rates borrowed from the literature (e.g., Carranza and Arnold, 2003; Miralles and Carranza, 2010; Lima *et al.*, 2013; Barker et al., 2015; Karin *et al.*, 2016) or to employ biogeographic events as calibrations (Hedges and Conn, 2012; Pinto-Sanchez et al., 2015) to derive the timescale of this lineage. The fossil record, however, has been demonstrated to be much more informative than biogeographic events as a calibration tool

(Hedges, 2011). The use of biogeographic events to time-calibrate phylogenies requires the assumption of vicariant scenarios of diversification, which entails that the speciation is synchronous with the breaking apart of landmasses, or that island colonization occurs immediately following geological formation (Hedges, 2011; Mello and Schrago, 2012).

Revealing the origin and diversification of the CAM clade and *T. atlantica* also requires a robust phylogenetic hypothesis. Although it is generally accepted that *T. atlantica* is a member of the genus *Trachylepis* and therefore excluded from the main diversification of the CAM, its evolutionary affinity remains controversial (Mausfeld et al., 2002; Carranza and Arnold, 2003; Whiting et al., 2006). Early proposed phylogenies of Mabuyinae used a maximum of 35 species, a number that significantly underrepresents the diversity of this subfamily (Mausfeld et al., 2002; Carranza and Arnold, 2003; Whiting et al., 2006). Hedges and Conn (2012) studied the CAM clade exclusively and included 40 species, whereas the large-scale Squamata phylogeny of Pyron et al. (2013), which contains 4,161 species, sampled 71 mabuyine skink species. Recently, Pinto Sanchez et al. (2015) used 250 specimens to infer the phylogeny and species diversity of neotropical mabuyinae, focusing on Colombian populations. Karin et al. (2016) analyzed the higher-order relationships of Mabuyinae using 22 species (24 specimens). Considering that improvements in phylogenetic inference and divergence time estimation can be obtained by increasing taxon sampling (Linder et al., 2005; Albert et al., 2009; Soares and Schrago, 2012), this matter requires further investigation.

Moreover, the occupation of the South American mainland by the CAM motivates an analysis of differential rates of diversification rates phenotypic traits evolution in this clade. It has been broadly reported that the ecological opportunity of a new environment can induce acceleration of macroevolutionary rates (Yoder et al., 2010; Liedtke et al., 2016). This

phenomenon was documented for insular vertebrates (Losos JB, Mahler DL. 2010; Jonsson KA et al. 2012); but, as proposed by G.G. Simpson (1953), the occupation of a new continent also could theoretically trigger evolutionary radiations (Yoder JB et al. 2010; Pires MM, Silvestro D, Quental TB. 2015).

In this study, as a means of exploring the continental biogeographic and evolutionary patterns associated with the occupation of South America by mabuyine skinks, we estimated the molecular phylogeny and inferred the divergence times of its members employing fossil information. The inferred time-tree was used to performed, for the first time, a formal statistical historical biogeography analysis and macroevolutionary diversification rates inference of Mabuyinae. To this end, by data-mining previously published data, we assembled the largest dataset of species and gene sampling composed to date, with the aim of uncovering the evolution of the Mabuyinae.

Materials and Methods

Data collection, alignment and taxonomy

We assembled a dataset containing sequence information for eight genetic loci from 117 species of Mabuyinae. Moreover, a representative number of terminals were collected from other scincid subfamilies (102 genera), totaling 219 taxa. All sequences were downloaded from GenBank. Two genera of the family Xantusiidae were used as outgroups. All seven genes available from *Trachylepis atlantica* were used in our analysis: the mitochondrial ribosomal genes *12S rRNA*, *16S rRNA* and the coding gene *cytochrome b (cytb)*; as well as the nuclear genes alpha enolase (*enol*), oocyte maturation factor (*cmos*), glyceraldehyde-3-phosphate dehydrogenase (*gapdh*), myosin heavy chain (*myh*), and G protein-coupled receptor 149 (*gpr149*). Detailed information on each locus was listed in Table 1 and accession numbers are available in Supplementary File 2. When several sequences representative of each taxon were available, the longest sequence was selected. We assumed genera and species as monophyletic. Therefore, chimeric supermatrices were used. All protein-coding sequences were visually checked for stop codons and aligned individually in SeaView v. 4.4.3 (Gouy *et al.*, 2010) using the MUSCLE v3.8.31 (Edgar 2004) algorithm, whereas the ribosomal genes were aligned in MAFFT v. 7 (Katoh and Standley, 2013). Gblocks v. 0.91b (Castresana, 2000; Talavera and Castresana, 2007) was used to exclude poorly aligned bases and divergent regions in the *12S rRNA*, *16S rRNA* and *enol* genes. Individual genes were then concatenated into a single supermatrix using the R package Phyloch (Heibl, 2015). RogueNarok (Aberer *et al.*, 2013) was used to identify taxa without significant phylogenetic information using a ML tree and associated bootstrapped topologies. These datasets were inferred under a rapid bootstrapping algorithm

analysis with 200 replicates (Stamatakis et al., 2008), followed by a thorough search of the ML tree using the evolutionary model GTRGAMMA, performed in RAxML-HPC (8.1.24) (Stamatakis, 2014). The terminal nodes *Larutia* and *Otosaurus*, which each presented relative bipartition information criterion (RBIC) higher than 1.0, were removed from the analysis. Another dataset, assembled with a more stringent (RBIC > 0.5) criterion, was composed for comparison. Under this criterion, the genus *Lankascincus* and the mabuyine species *Eumecia anchietae* and *Trachylepis acutilabris* were excluded from the analyses. The results, however, were robust for both RBIC values, and we report the results under RBIC > 1.0 hereafter. The final supermatrix consisting of 4,131 base pairs are available in Supplementary File 2.

Evolutionary analyses

We investigated 18 candidate partitioning schemes using PartitionFinder heuristic search algorithm with the Bayesian information criterion (BIC) for model selection (Lanfear et al., 2012). The partitioning schemes also tested codon positions of protein-coding genes. The partitioning strategy with the best fit consisted of 7 partitions (Supplementary File 3), which were used throughout the analyses. Maximum likelihood (ML) phylogenetic inference was performed in RAxML-HPC (8.1.24) employing the evolutionary model GTRCAT. The GTR substitution model was applied to each partition, as this is the only model supported in RAxML. ML analyses used 200 initial searches for finding the optimal tree topology. Statistical support for clades was assessed using 2,000 standard nonparametric bootstrap replicates (PB).

Inference of node ages was performed with the mcmctree program of the PAML 4.8a package (Yang, 2007). For large alignments, the Bayesian inference of node ages via Markov chain Monte Carlo is computationally intensive. To make the analyses feasible, we used an

approximate likelihood calculation modified from Thorne et al. (1998) and implemented in mcmctree (Reis and Yang, 2011). Priors for the ρ gene and σ^2 parameters were set as $G(2, 200)$ and $G(1, 10)$, respectively. Markov chains were sampled every 1,000 generations until 50,000 trees were collected. The analysis was performed twice to check for convergence of the chains. Effective sample sizes (ESS) of parameters were calculated in Tracer v. 1.5, and all values were greater than 200.

Calibration priors

The age of the root, which corresponds to the split between the families Scincidae and Xantusiidae, was calibrated at a minimum value of 70.6 Ma and a maximum age of 209.5 Ma. The minimum was set according to the oldest scincid from the Late Cretaceous of North America (Campanian, 83.5 – 70.6 Ma), as previously adopted by Mulcahy *et al.* (2012) (Rowe *et al.*, 1992); while the maximum was set according to Benton *et al.* (2015), which proposed a maximum age of the ancestral of Squamata. Additional calibration information was gathered from the Paleobiology Database (paleobiology.org) and entered as minimum ages of the stem nodes of clades. The minimum age of the Scincidae stem node was calibrated at 20.4 Ma, based on the oldest crown scincid *Eumeces antiquus* classified as a member of the subfamily Scincinae (Holman, 1981; Estes, 1983). The stem node of the clade containing the extant genus *Eumeces* was calibrated at a minimum age of 13.6 Ma based on fossils from the Middle Miocene in North America (Holman, 1966; Voorhies *et al.*, 1987; Joeckel, 1988). The age of an extinct *Egernia* sp. from the Miocene of Hungary (>5 Ma) was used to calibrate the stem node of the clade containing this extant genus (Venczel and Hír, 2013). According to Böhme (2010), the fossil *Tropidophorus bavaricus* belongs to extant genus *Tropidophorus*, and it was used to calibrate the

stem node of this clade, setting its minimum age at 13.6 Ma (Böttcher *et al.*, 2009). Calibrate nodes are shown in the Supplementary Figure 4.

Ancestral area reconstruction

A historical biogeographical reconstruction was performed for the subfamily Mabuyinae and its sister clade (*Lankascincus* and *Ristella*). The R package BioGeoBEARS (Matzke 2013) was used to run likelihood methods: DIVALIKE (a likelihood interpretation of DIVA that allows for the same events as DIVA – Matzke, 2013) and DEC (Dispersal-Extinction-Cladogenesis, Ree and Smith, 2008). In BioGeoBEARS, we used the likelihood ratio to test whether the null models (DIVALIKE and DEC) fitted the data better than did the more sophisticated models (DIVALIKE+J and DEC+J). The “J” in models represents the addition of the founder-event speciation, thereby allowing dispersal without range expansion (Matzke 2014). The maximum range size, which limits the number of areas defined by tips and nodes, was set to two, based on the current geographic distribution of species. Constraints on dispersal or area availability were not included. To make the biogeographic analysis computationally feasible, islands were not considered independent regions. The rationale for choosing the seven biogeographic areas follows the zoogeographical regions found in the herpetological and biogeographical literature (Vitt and Caldwell, 2009; Lomolino, 2010; Morrone, 2014; Pyron, 2014): (1) Neotropical Brazilian Subregion (B), (2) Neotropical Chacoan Subregion (C), (3) West Indies – Caribbean Islands (W); (4) Oriental Region (O): Southeast Asia + Philippines + Indian Subcontinent (Pakistan to Bangladesh, including Sri Lanka, Nepal, and Bhutan); (5) Afrotropical (A): Sub-Saharan Africa; (6) Madagascar (M): Madagascar and adjacent islands (the Seychelles and the Comoros); and (7) Saharo-Arabian (S): Europe + North Africa + the northern portion of the

Arabian Peninsula + Southwest Asia. Using online distributional data from the Reptile Database (Uetz and Hosek, 2015), we classified the tips as belonging to one or more of these areas.

Rate of species diversification and diversification-phenotype rate correlation

Our dated phylogeny of Mabuyinae was used to infer the dynamics of species diversification using BAMM 2.5 (Bayesian Analysis of Macroevolutionary Mixtures – Rabosky, 2014), which simultaneously accounts for variation in evolutionary rates through time and among lineages using transdimensional (reversible-jump) Markov chain Monte Carlo (rjMCMC) (Rabosky 2014). Markov chains were sampled every 1,000th generation until 37,500 trees were collected after a burn-in of 25%. Prior distributions were set according to *setBAMMPriors* function from the BAMMtools R package (Rabosky *et al.* 2014). The frequencies of the species in each genus were considered.

We were also prompted to test for trait-dependent diversification. The following traits were collated from the literature: (i) data on the reproductive mode for 60 species of the Mabuyinae – 39 species as viviparous, 17 as oviparous, and 3 as ovoviviparous (Meiri *et al.* 2013; Pyron and Burbrink 2013); (ii) the SVL (snout–vent length) for 46 species (Meiri 2010; Miralles and Carranza, 2010; Das 2010; Hedges and Conn, 2012; Meiri *et al.* 2013; Pyron and Burbrink 2013); and (iii) the body mass data for 35 species (Meiri 2010; Hedges and Conn, 2012). Following previous works, we treated "ovoviviparity" as viviparity (Pyron and Burbrink 2013).

Differences in the rates of speciation (λ) and extinction (μ) between New World (*NW*) and Old World (\overline{NW}) areas and between viviparous (*V*) and non-viviparous (\overline{V}) lineages were tested using two approaches. Firstly, an ANOVA, implemented in the R package *diversitree* (FitzJohn 2012), was used to compare different macroevolutionary regimes. In the *NW*/ \overline{NW}

273 comparison, rates were calculated using the GeoSSE approach (Goldberg *et al.* 2011), whereas
 274 BiSSE was used to test V/\bar{V} rates, with both tests of binary characters as implemented in
 275 diversitree. In this sense, GeoSSE first optimised the parameters under an unconstrained full
 276 model, in which ML estimates were obtained for (i) the speciation rate of the New World lineage
 277 (λ_{NW}); (ii) the speciation rate of non-New World lineages ($\lambda_{\bar{NW}}$); (iii) the extinction rate of the
 278 New World clade (μ_{NW}); (iv) the extinction rate of non-New World lineages ($\mu_{\bar{NW}}$); (v) the
 279 intermediate speciation rate parameter ($\lambda_{NW,\bar{NW}}$); (vi) the dispersal rates from the New World
 280 clade ($d_{NW \rightarrow \bar{NW}}$); and (vii) the dispersal rates of the sister lineages ($d_{\bar{NW} \rightarrow NW}$). Similarly, BiSSE
 281 was used to infer (i) the speciation rate of the viviparous lineage (λ_V); (ii) the speciation rate of
 282 non-viviparous lineages ($\lambda_{\bar{V}}$); (iii) the extinction rate of the viviparous clade (μ_V); (iv) the
 283 extinction rate of non-viviparous lineages ($\mu_{\bar{V}}$); (v) transition rates from the viviparous clade ($q_{V \rightarrow \bar{V}}$); and (vi) transition rates of the sister clade ($q_{\bar{V} \rightarrow V}$). Initial parametric values were set
 284 according to the *starting.point* function from the diversitree package with an initial ratio of 0.5.
 285 This preliminary step was required to build the likelihood function (the *make* command). In
 286 BiSSE, species with unknown states were coded as ‘NA’ and assigned the sampling fraction of
 287 the species of Mabuyinae used in this work (~61%), independent of the character state.
 288 Subsequently, to perform the ANOVA test of the GeoSSE results, we chose to constrain the
 289 intermediate speciation and the dispersal rate parameters to zero ($\lambda_{NW,\bar{NW}} = d_{NW \rightarrow \bar{NW}} =$
 290 $d_{\bar{NW} \rightarrow NW} = 0$), in order to compare speciation and the extinction rates within regions exclusively.
 291 Finally, we tested macroevolutionary alternative models against the full models (Table 2).
 292
 293 The second comparison between NW/\bar{NW} and V/\bar{V} macroevolutionary regimes used the
 294 marginal posterior distributions of macroevolutionary parameters, inspected using the R package

diversitree. These distributions were obtained using the MCMC analyses, with samples taken in diversitree every 1,000th generation until 1,000 samples were collected. A broad exponential prior (mean of 0.5) for all parameters was used, as recommended, while the λ and μ rates were set as the values obtained in the ML full model (FitzJohn 2012). For the NW/\overline{NW} relationship, we also used the marginal posterior distributions obtained from BAMM, applying the *getCladeRates* function of the BAMMtools R package. For all approaches, we calculated the 95% highest posterior density (HPD) interval for the difference between the means of the NW/\overline{NW} and V/\overline{V} lineages (Bolstad 2007).

The diversification-phenotype rate correlation was performed in STRAPP (Rabosky and Huang 2015), as implemented in BAMMtools. To run STRAPP, phylogenies were pruned to match the available information for both tree terminals and traits. Diversification analysis results previously obtained using BAMM were pruned to match the available trait information using the function *subtreeBAMM* and were tested against the analyzed traits, using the Mann-Whitney method for binary characters and both Pearson and Spearman methods for continuous traits.

Rate of trait evolution and ancestral state reconstruction

We inferred ancestral states of reproduction mode in the BayesTraits software (Pagel *et al.* 2004) and in the R package BiSSE. Both approaches use the maximum likelihood method and allow the use of species with unknown character states. Results were visualized in the R package diversitree. The rates of trait evolution of the two continuous traits studied, namely, SVL and body mass, were inferred in BAMM, using logarithms of both measures. Phylogenies were also pruned to match the available information for both tree terminals and traits. We tested the correlation between SVL and body mass using phylogenetic independent contrasts (PIC;

318 Felsenstein, 1985) as implemented in the R package ape. Prior distributions to these subtrees
 319 were set according to *setBAMMPriors* function from the BAMMtools R package (Rabosky *et al.*
 320 2014). Chains were sampled every 1,000 generations until 50,000 trees were collected. The
 321 results were summarized and visualized in BAMMtools.

Results

Mabuyinae was recovered as a monophyletic group (BS = 76). The first split in this subfamily isolated the genus *Dasia* from the remaining Mabuyinae and was inferred to have occurred at 30 Ma, with an HPD interval ranging from 20 – 48 Ma (Supplementary File 4). The biogeographical model with the highest likelihood was the DEC+J (lnL: -94.34). Therefore, the addition of the J parameter for founder events significantly increased the likelihood of the DEC model ($p = 1.2e^{-9}$, Table 3). Our results supported a model in which the genera *Lankascincus* and *Ristella* split from Mabuyinae (BS = 20) in the Oriental region.

After dispersal from the Oriental region to Africa, the ancestor of the CAM clade split from Saharo-Arabian *Trachylepis* approximately 25 Ma ($\pm 16 - 41$ Ma) (BS=60). Subsequently, the stock that gave rise to New World Mabuyinae dispersed from the Saharo-Arabian region to the Neotropical Brazilian Subregion (Figure 1). The New World Mabuyinae were recovered as monophyletic (BS=97), and the age of their LCA was estimated as 19 Ma (12 – 31 Ma). Our results supported the hypothesis that this lineage reached the Neotropics via the Brazilian Subregion. From this region, the ancestor of *Spondylurus* and *Copeoglossum* dispersed to islands of the West Indies. The continental clade later dispersed along the Brazilian Subregion. Additional dispersal events to the West Indies were inferred to have occurred at least twice: in the ancestor of *Mabuya* and in that of *Alinea* and *Marisora*. *Marisora* was not recovered as monophyletic, with the Central American clade as sister to *Alinea* and the South American clade as sister to *Aspronema*.

The origin of *Trachylepis atlantica* consisted of a different history. Firstly, the monophyly of genus *Trachylepis* was not recovered. This genus was split into two lineages: one

sister lineage of the CAM clade and another major clade to which *T. atlantica* belongs. Our analysis supported the phylogenetic position of *T. atlantica* as a sister lineage of the clade including the Europa Island *T. infralineata*, the Madagascan *T. comorensis*, and the continental African *T. maculilabris* (BS = 29). We found that the *T. atlantica* ancestor diverged from the remaining *Trachylepis* in the Miocene, approximately 17 Ma (between 10 and 27 Ma) in tropical Africa. The younger diversification of *T. atlantica* indicates that the crossing of this species to Fernando de Noronha occurred more recently than did the occupation of South America by the CAM clade. Our analyses also suggested that once the ancestors of this major *Trachylepis* clade reached Africa, dispersal events from the African continent occurred to Madagascar and nearby islands at least three times, in addition to the dispersion to the Neotropics (*T. atlantica*). Our results showed that a monophyletic Madagascan *Trachylepis* (BS = 94) diverged from its sister group approximately 19 Ma (12 – 32 Ma) (BS = 23).

The ANOVA was used to test whether the full four-parameter GeoSSE model of the NW/\overline{NW} comparison (model 1) was statistically favored over simpler alternative models (models 2-4). When we constrained speciation rates to be equal (model 2) and to present equal speciation and extinction rates (model 4), the full model was supported over these alternative models ($p = 0.00782$ and $p = 0.02900$, respectively). The comparison with the model that constrained extinction rates (model=3), however, was not significantly different ($p = 0.15575$).

GeoSSE analysis of the difference between the posterior distributions of speciation rates in NW/\overline{NW} lineages indicated a positive credible interval (CI) (0.005 – 0.113), indicating that the speciation rates of South American clades (NW) were indeed higher than those of the remaining lineages (\overline{NW}). In BAMM, however, we failed to find a significant difference between the posterior distributions of speciation rates (CI: -0.030 – 0.031). We also found no evidence for

diversification shifts during mabuyine diversification. However, the macroevolutionary cohort matrix suggested heterogeneous macroevolutionary rate regimes in the South American Mabuyinae and the remaining clades (Figure 2a). In both GeoSSE and BAMM analyses, we failed to find differences between the extinction rates (CI_{GeoSSE} : -0.036 – 0.074, CI_{BAMM} : -0.027 – 0.027) and between the net diversification rates (CI_{GeoSSE} : -0.010 – 0.090, CI_{BAMM} : -0.027 – 0.027) (Figure 2b).

Concerning the influence of viviparity on diversification rates, no submodels were supported over the unconstrained model ($p > 0.05$). STRAPP analysis failed to find any correlation between diversification rates and mode of reproduction ($p > 0.05$). We also found no differences between the means obtained from the MCMC of the BiSSE (CI : -0.018 – 0.125, -0.038 – 0.055, -0.051 – 0.049, respectively for speciation, extinction, and transition rates).

The ancestor of CAM and their *Trachylepis* sister clade was recovered as viviparous by BayesTraits ($P_{\text{BTML}}=0.751$; $P_{\text{BTBA}}=0.787$) and as oviparous by BiSSE ($P_{\text{BiSSE}}=1$), while the ancestor of CAM alone was recovered as viviparous by all approaches ($P_{\text{BiSSE}}=0.953$; $P_{\text{BTML}}=0.992$; $P_{\text{BTBA}}=0.983$), suggesting that the ancestral species that colonised South America was likely viviparous (Figure 2a). The ancestor of the subfamily Mabuyinae was inferred as oviparous ($P_{\text{BiSSE}}=1$; $P_{\text{BTML}}=0.983$; $P_{\text{BTBA}}=0.980$). Viviparity seems to have emerged several times in the genus *Trachylepis*, as well as in the ancestor of *Chioninia* ($P_{\text{BiSSE}}=0.952$; $P_{\text{BTML}}=0.984$; $P_{\text{BTBA}}=0.971$).

According to PIC results, SVL measurement and body mass were correlated ($p_{\text{LRP}} < 0.01$), however, we had found differences in their evolutionary rates. The mean rates of trait evolution of SVL and body mass were smaller in the New World clade than in the \overline{NW} lineages. However, these differences were not significant. In BAMM, we found an increase in the rate of

391 SVL trait evolution in genus *Chioninia*. Only one clade presented an increase in the rate of body
 392 mass trait evolution: the Seychelles *Trachylepis* (*T. wrightii* and *T. brevicollis*). STRAPP
 393 analysis failed to find a significant correlation between the rate of species diversification and the
 394 rate of SVL or body mass trait evolution ($p > 0.05$).

Discussion

Our analyses based on a fossil-calibrated timetree support the hypothesis that the occupation of South America by the ancestors of the Continental America Mabuyinae (CAM) clade occurred between the Eocene and the Oligocene from the Saharo-Arabian region, corroborating recent estimates (e.g. Pinto-Sánchez *et al.* 2015; Karin *et al.*, 2016). This colonization occurred independently of that of *Trachylepis atlantica*, which split from its tropical African sister clade between the Oligocene and the Miocene. Therefore, our results corroborated the hypothesis that the ancestor of *T. atlantica* dispersed from tropical Africa as suggested by Mausfeld *et al.* (2002). The divergence between *T. atlantica* and its African ancestors was older than the estimated age of the formation of the Fernando de Noronha archipelago (12.3 – 1.7 Ma; Almeida, 2002). Thus, it is unlikely that the ancestors of *T. atlantica* reached South America in a single transoceanic dispersal event. It is more probable to hypothesize a complex dispersal scenario, with a series of dispersals through a transatlantic island corridor leading to Fernando de Noronha, consistent with the Atlantogea model (Oliveira *et al.* 2009; Ezcurra and Agnolin, 2012). This stepping-stone mode of dispersal could explain the discrepancy between the ages of genetic divergence and the formation of the archipelago because it would dissociate the geological formation of the Fernando de Noronha archipelago from the genetic isolation of the ancestor of *T. atlantica*. This result must be interpreted cautiously because choosing between alternative biogeographical scenarios is influenced by the inferred timescale. The lack of extant and extinct mabuyine species may have impacted divergence time estimates.

The age of the crown node of a living CAM species was inferred to be much older than the previously reported estimates by Carranza and Arnold (2003), 7 – 9 Ma. However, the ages

inferred by Miralles and Carranza (2010); Hedges and Conn (2012) and Karin *et al.* (2016), at approximately 14 Ma, lies within the range reported in our work (12 – 31 Ma). Nevertheless, all previous estimates were also incompatible with single-event scenarios of New World colonization. Note that a fossil of a putative species of Mabuyinae has been described in the mid-Atlantic Ascension Island, although no recent systematic re-evaluation of this specimen is available (Mausfeld and Vrcibradic, 2002). The direction of Atlantic Ocean currents from Africa to South America would explain the scarcity of dispersals in the opposite direction (Houle, 1999). The evolution of the two American lineages of Mabuyinae also corroborates this faunal connection and suggests that this dispersal may have occurred through island hopping, which is more likely than a single sweepstake event across the Atlantic Ocean.

The arrival of Mabuyinae to South America seems to have changed the diversification rate regime of this lineage. The evolution rates of both body mass and SVL traits were not significantly altered after the arrival of the ancestors of New World clades, but the increases in rates of both traits were found in island groups, such in Cape Verde's *Chioninia* and Seychelles' *Trachylepis*. The viviparous CAM clade presents peculiar reproductive traits, such as a specialized chorioallantoic placenta that provides the fetal nutrition, similarly to eutherian mammals (Vrcibradic and Rocha, 2011). This character was suggested to be synapomorphous by Mausfeld *et al.* (2002). According to our analysis, the CAM ancestor was already viviparous, in agreement with the large-scale analysis of Squamates (Pyron and Burbrink, 2013). However, Pyron and Burbrink (2013) analyses suggested an early origin of viviparity in Mabuyinae and multiple reversions to oviparity, including the clade consisting of *Chioninia* and CAM + *Heremites*. On the other hand, possibly because of topological differences, we estimated that the

ancestor of Mabuyinae was oviparous and multiple reversions to viviparity occurred later along the evolution of the lineage.

Regarding the higher systematics of Mabuyinae, Greer (1970), based on the primitive characteristics of Asian Mabuyinae, suggested that the ancestors of this subfamily first dispersed from Asia. Other works reasserted this hypothesis, although no formal statistical analyses were conducted (Honda *et al.*, 1999; 2003; Karin *et al.*, 2016). Our results suggested that the early members of Mabuyinae dispersed to Africa and then reached South America and several oceanic islands. The inferred time-dated phylogeny and biogeographic reconstruction similarly suggested an Oriental ancestral distribution, as corroborated by the distribution of the genera *Lankascincus* and *Ristella*, which were recovered as sister clades of Mabuyinae and are geographically distributed in the Oriental region (Sri Lanka and the Indian, respectively). Karin *et al.*, 2016 inferred that *Eutropis*, instead of *Dasia*, was the first Mabuyinae offshoot. Nevertheless, as both genera are distributed in the Oriental region, this topological rearrangement would not significantly alter the ancestral area reconstruction.

Finally, this study provided an updated timescale and estimates of macroevolutionary regimes of the diversification of the subfamily Mabuyinae. Our focus was the occupation of the South American continent by the subfamily Mabuyinae through an Oligocene/Eocene transoceanic connection (Atlantogea), which could be responsible for approximately 29% of the South American mammal diversity, suggesting the importance of this event to the extant vertebrate diversity in South America (Marshall *et al.*, 1982). CAM and *T. atlantica* are the only representatives of the family Scincidae in South America accounting for 4% of the approximately 1560 squamate species on this continent (Uetz, 2000). Although there are species of other subfamilies of Scincidae in Central America, these species seem to have never crossed

the Isthmus of Panama. The converse also seems true because the only mabuyine genus (*Marisora*) that partially occupied Central America based on our results had arrived via an intermediate dispersion through the West Indies. If this is the case, no representative of Scincidae would be found in present day South American fauna in the absence of this Atlantogea connection. Our results presented an overall picture of the timing, biogeography and macroevolutionary dynamics associated with the arrivals of the ancestors of the only vertebrate clade known to have undergone transoceanic dispersal twice.

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References

- Aberer AJ, Krompass D, Stamatakis A. 2013. Pruning Rogue Taxa Improves Phylogenetic Accuracy: An Efficient Algorithm and Webservice. *Systematic Biology* 62:162–166.
- Albert EM, San Mauro D, García-París M, Rüber L, Zardoya R. 2009. Effect of taxon sampling on recovering the phylogeny of squamate reptiles based on complete mitochondrial genome and nuclear gene sequence data. *Gene* 441:12–21.
- Almeida FFM. 2002. Arquipélago de Fernando de Noronha. *Sítios geológicos e paleontológicos do Brasil* 66:361–368.
- Barker FK, Burns KJ, Klicka J, Lanyon SM, Lovette IJ. 2015. New insights into New World biogeography, An integrated view from the phylogeny of blackbirds, cardinals, sparrows, tanagers, warblers, and allies. *The Auk* 132:333–348.
- Benton MJ, Donoghue P, Asher RJ. 2015. Constraints on the timescale of animal evolutionary history. *Palaeontologia Electronica* 18.1.1FC:1-106.
- Böhme M. 2010. Ectothermic vertebrates (Actinopterygii, Allocaudata, Urodela, Anura, Crocodylia, Squamata) from the Miocene of Sandelzhausen (Germany, Bavaria). and their implications for environment reconstruction and palaeoclimate. *Paläontologische Zeitschrift* 84:3–41.
- Böttcher R., Heizmann EPJ, Rasser MW, Ziegler R. 2009. Biostratigraphy and palaeoecology of a Middle Miocene (Karpadian, MN 5) fauna from the northern margin of the North Alpine Foreland Basin (Oggenhausen 2, SW' Germany). *Neues Jahrbuch für Geologie und Paläontologie - Abhandlungen* 254:237–260.
- Briggs JC. 2003. Fishes and Birds, Gondwana Life Rafts Reconsidered. *Systematic Biology*

498 52:548–553.

499 Carranza S, Arnold EN. 2003. Investigating the origin of transoceanic distributions, mtDNA
500 shows *Mabuia* lizards (Reptilia, Scincidae) crossed the Atlantic twice. *Systematics and*
501 *Biodiversity* 1:275–282.

502 Castresana J. 2000. Selection of Conserved Blocks from Multiple Alignments for Their Use in
503 Phylogenetic Analysis. *Molecular Biology and Evolution* 17:540–552.

504 Das I. 2010. *A Field Guide To The Reptiles Of South-East Asia*, London: New Holland
505 Publishers.

506 de Oliveira FB, Molina EC, Marroig G. 2009. Paleogeography of the South Atlantic, a Route for
507 Primates and Rodents into the New World? In Garber PA, Estrada A, Bicca-Marques JC,
508 Heymann EW, Strier KB, ed. *South American Primates, Developments in Primatology:*
509 *Progress and Prospects*. New York: Springer. p 55-68.

510 de Queiroz A. 2005. The resurrection of oceanic dispersal in historical biogeography. *Trends in*
511 *Ecology and Evolution* 20:68–73.

512 de Queiroz A. 2014. *The Monkey's Voyage*, How Improbable Journeys Shaped the History of
513 Life. New York: Basic Books.

514 Edgar RC. 2004. MUSCLE, multiple sequence alignment with high accuracy and high
515 throughput. *Nucleic Acids Research* 32:1792–1797.

516 Estes R. 1983. *Encyclopedia of Paleoherpetology*. Stuttgart: Gustav Fischer Verlag.

517 Ezcurra MD, Agnolin FL. 2012. A New Global Palaeobiogeographical Model for the Late
518 Mesozoic and Early Tertiary. *Systematic Biology* 61:553–566.

519 FitzJohn RG. 2012. Diversitree, comparative phylogenetic analyses of diversification in R.
520 *Methods in Ecology and Evolution* 3:1084–1092.

521 Goldberg EE, Lancaster LT, Ree RH. 2011. Phylogenetic Inference of Reciprocal Effects
522 between Geographic Range Evolution and Diversification. *Systematic Biology* 60:451–465.

523 Gouy M, Guindon S, Gascuel O. 2010. SeaView Version 4, A Multiplatform Graphical User
524 Interface for Sequence Alignment and Phylogenetic Tree Building. *Molecular Biology and*
525 *Evolution* 27:221–224.

526 Greer AE. 1970. A subfamilial classification of scincid lizards. *Bull. Mus. comp. Zool. Harvard*
527 139:151–183.

528 Heads M. 2011. Old Taxa on Young Islands, A Critique of the Use of Island Age to Date Island-
529 Endemic Clades and Calibrate Phylogenies. *Systematic Biology* 60:204–218.

530 Hedges SB, Conn CE. 2012. A new skink fauna from Caribbean islands (Squamata, Mabuyidae,
531 Mabuyinae). *Zootaxa* 3288:1–244.

532 Heibl C. 2015. Phyloch. Available at <http://www.christophheibl.de/Rpackages.html>.

533 Holman JA. 1966. A small Miocene herpetofauna from Texas. *Quarterly Journal of the Florida*
534 *Academy of Sciences* 29:267–275.

535 Holman JA. 1981. A herpetofauna from an early extension of the Harrison Formation (early
536 Miocene, Arikareean), Cherry County, Nebraska. *Journal of Vertebrate Paleontology* 1:49–
537 56.

538 Honda M, Ota H, Kobayashi M, Nabhitabhata J, Yong H, and Hikida T. 1999. Evolution of
539 Asian and African Lygosomine Skinks of the *Mabuya* Group (Reptilia, Scincidae), A
540 Molecular Perspective. *Zoological Science* 16:979–984.

541 Honda M, Ota H, Kohler G, Ineich I, Chirio L, Chen S-L, Hikida T. 2003. Phylogeny of the
542 Lizard Subfamily Lygosominae (Reptilia, Scincidae), with Special Reference to the Origin
543 of the New World Taxa. *Genes Genet. Syst.* 78:17–80.

544 Houle A. 1999. The Origin of Platyrrhines: An Evaluation of the Antarctic Scenario and the
545 Floating Island Model. *American Journal of Physical Anthropology* 109:1–19.

546 Joeckel RM. 1988. A new late Miocene herpetofauna from Franklin County, Nebraska. *Copeia*
547 3:787–789.

548 Karin BR, Metallinou M, Weinell JL, Jackman TR, Bauer AM. 2016. Resolving the higher-order
549 phylogenetic relationships of the circumtropical Mabuya group (Squamata: Scincidae): An
550 out-of-Asia diversification. *Molecular Phylogenetics and Evolution* 102:220–232.

551 Katoh K, Standley DM. 2013. MAFFT Multiple Sequence Alignment Software Version 7,
552 Improvements in Performance and Usability. *Molecular Biology and Evolution* 30:772–780.

553 Lanfear R, Calcott B, Ho SYW, Guindon S. 2012. PartitionFinder, Combined Selection of
554 Partitioning Schemes and Substitution Models for Phylogenetic Analyses. *Molecular*
555 *Biology and Evolution* 29:1695–1701.

556 Le M, Raxworthy CJ, Mccord WP, Mertz L. 2006. A molecular phylogeny of tortoises
557 (Testudines, Testudinidae). based on mitochondrial and nuclear genes. *Molecular*
558 *Phylogenetics and Molecular Phylogenetics and Evolution* 40:517–531.

559 Leigh EG, O'Dea A, Vermeij GJ. 2013. Historical biogeography of the Isthmus of Panama.
560 *Biological Reviews* 89:148–172.

561 Leite RN, Kolokotronis S-O, Almeida FC, Werneck FP, Rogers DS, Weksler M. 2014. In the
562 Wake of Invasion, Tracing the Historical Biogeography of the South American Cricetid
563 Radiation (Rodentia, Sigmodontinae). *PLoS ONE* 9:e100687.

564 Lima A, Harris DJ, Rocha S, Miralles A, Glaw F, Vences M. 2013. Phylogenetic relationships of
565 *Trachylepis* skink species from Madagascar and the Seychelles (Squamata, Scincidae).
566 *Molecular Phylogenetics and Evolution* 67:615–620.

567 Linder HP, Hardy CR, Rutschmann F. 2005. Taxon sampling effects in molecular clock dating,
568 An example from the African Restionaceae. *Molecular Phylogenetics and Evolution* 35:569–
569 582.

570 Lomolino MV. 2010. *Biogeography*. Sunderland: Sinauer Associates.

571 Loss-Oliveira L, Aguiar B, Schrago CG. 2012. Testing Synchrony in Historical Biogeography:
572 The Case of New World Primates and Hystricognathi Rodents. *Evolutionary Bioinformatics*
573 8:127–137.

574 Marshall LG, Webb SD, Sepkoski Jr JJ, Raup DM. 1982. Mammalian evolution and the Great
575 American Interchange. *Science* 215:1351–1357.

576 Matzke NJ. 2013. Probabilistic historical biogeography, new models for founder-event
577 speciation, imperfect, d. etection., and fossils allow improved accuracy and model-testing.
578 *Frontiers of Biogeography* 5:242–248.

579 Matzke NJ. 2014. Model Selection in Historical Biogeography Reveals that Founder-Event
580 Speciation Is a Crucial Process in Island Clades. *Systematic Biology* 63: 951–970.

581 Mausfeld P. 2000. First Data on the Molecular Phylogeography of Scincid Lizards of the Genus
582 *Mabuya*. *Molecular Phylogenetics and Evolution* 17:11–14.

583 Mausfeld P, Vrcibradic D. 2002. On the Nomenclature of the Skink (*Mabuya*). Endemic to the
584 Western Atlantic Archipelago of Fernando de Noronha, Brazil. *Journal of Herpetology*
585 36:292–295.

586 Mausfeld P, Schmitz A, Bohme W, Misof B, Vrcibradic D, Rocha CFD. 2002. Phylogenetic
587 Affinities of *Mabuya atlantica* Schmidt, 1945, Endemic to the Atlantic Ocean Archipelago
588 of Fernando de Noronha (Brazil), Necessity of Partitioning the Genus *Mabuya* Fitzinger,
589 1826 (Scincidae, Lygosominae). *Zoologischer Anzeiger* 241:281–293.

590 Meiri S. 2010. Length-weight allometries in lizards. *Journal of Zoology* 281:218–226.

591 Meiri S, Bauer AM, Chirio L, Colli GR, Das I, Doan TM, Feldman A, Herrera F-C, Novosolov
592 M, Pafilis P, Pincheira-Donoso D, Powney G, Torres-Carvajal O, Uetz P, Van Damme R.
593 2013. Are lizards feeling the heat? A tale of ecology and evolution under two temperatures.
594 *Global Ecology and Biogeography* 22: 834–845.

595 Mello B, Schrago CG. 2012. Incorrect handling of calibration information in divergence time
596 inference, an example from volcanic islands. *Ecology and Evolution* 2:493–500.

597 Miralles A, Carranza S. 2010. Molecular Phylogenetics and Evolution. *Molecular Phylogenetics*
598 *and Evolution* 54:857–869.

599 Morrone JJ. 2014. Biogeographical regionalisation of the Neotropical region. *Zootaxa* 3782:1–
600 110.

601 Mulcahy DG, Noonan BP, Moss M, Townsend TM, Reeder TW, Sites Jr JW, Wiens JJ. 2012.
602 Estimating divergence dates and evaluating dating methods using phylogenomic and
603 mitochondrial data in squamate reptiles. *Molecular Phylogenetics and Evolution* 65:974–
604 991.

605 Pagel M, Meade A, Barker D. 2004. Bayesian Estimation of Ancestral Character States on
606 Phylogenies. *Systematic Biology* 53:673–684.

607 Pinto-Sanchez NR, Calderón-Espinosa ML, Miralles A, Crawford AJ, Ramírez-Pinilla MP.
608 2015. Molecular phylogenetics and biogeography of the Neotropical skink genus *Mabuya*
609 *Fitzinger* (Squamata: Scincidae) with emphasis on Colombian populations. *Molecular*
610 *Phylogenetics and Evolution* 93:188–211.

611 Poux C, Chevret P, Huchon D, de Jong WW, Douzery EJP. 2006. Arrival and Diversification of
612 Caviomorph Rodents and Platyrrhine Primates in South America. *Systematic Biology*

55:228–244.

Pyron RA. 2014. Biogeographic Analysis Reveals Ancient Continental Vicariance and Recent Oceanic Dispersal in Amphibians. *Systematic Biology* 63:779-797.

Pyron RA, Burbrink FT. 2013. Early origin of viviparity and multiple reversions to oviparity in squamate reptiles. *Ecology Letters* 17:13–21.

Pyron R, Burbrink FT, Wiens JJ. 2013. A phylogeny and revised classification of Squamata, including 4161 species of lizards and snakes. *BMC Evolutionary Biology* 13:93.

Rabosky DL. 2014. Automatic detection of key innovations, rate, shifts, and diversity-dependence on phylogenetic trees. *PLoS ONE* 9:e89543.

Rabosky DL, Grudler M, Anderson C, Title CP, Shi JJ, Brown JW, Huang H, Larson JG. 2014. BAMMtools, an R package for the analysis of evolutionary dynamics on phylogenetic trees. *Methods in Ecology and Evolution* 5:701–707.

Renner SS. 2004. Plant dispersal across the tropical atlantic by wind and sea currents. *International Journal of Plant Science* 165:S23–S33.

Rowe TB, Cifelli RL, Lehman TM, Weil A. 1992. The Campanian Terlingua local fauna, with a summary of other vertebrates from the Aguja Formation. *Journal of Vertebrate Paleontology* 12:472–493.

Sanmartín I, van der Mark P, Ronquist F. 2008. Inferring dispersal, a Bayesian approach to phylogeny-based island biogeography, with special reference to the Canary Islands. *Journal of Biogeography* 35:428–449.

Schrago CG, Mello B, Soares AER. 2013. Combining fossil and molecular data to date the diversification of New World Primates. *Journal of Evolutionary Biology* 26: 2438–2446.

Schrago CG, Menezes AN, Moreira MAM, Pissinatti A, Seuánez HN. 2012. Chronology of

636 Deep Nodes in the Neotropical Primate Phylogeny, Insights from Mitochondrial Genomes.
637 *PLoS ONE* 7: e51699.

638 Simpson GG. 1950. History of the Fauna of Latin America. *American Scientist* 38: 361–389.

639 Simpson GG. 1953. The Major Features of Evolution. Columbia University Press, New York.

640 Sindaco R, Metallinou M, Pupin F, Fasola M, Carranza S. 2012. Forgotten in the ocean,
641 systematics, biogeography and evolution of the *Trachylepis* skinks of the Socotra
642 Archipelago. *Zoologica Scripta* 41:346–362.

643 Soares A, Schrago CG. 2012. The Influence of Taxon Sampling and Tree Shape on Molecular
644 Dating, An Empirical Example from Mammalian Mitochondrial Genomes. *Bioinformatics*
645 *and Biology Insights* 6:129-43.

646 Stamatakis A. 2014. RAxML Version 8, A tool for Phylogenetic Analysis and Post-Analysis of
647 Large Phylogenies. *Bioinformatics* 30:1312-1313.

648 Stamatakis A, Hoover P, Rougemont J. 2008. A Rapid Bootstrap Algorithm for the RAxML
649 Web Servers. *Systematic Biology* 57:758–771.

650 Talavera G, Castresana J. 2007. Improvement of Phylogenies after Removing Divergent and
651 Ambiguously Aligned Blocks from Protein Sequence Alignments. *Systematic Biology*
652 56:564–577.

653 Teeling EC. 2005. A Molecular Phylogeny for Bats Illuminates Biogeography and the Fossil
654 Record. *Science* 307:580–584.

655 Uetz P. 2000. How many Reptile Species? *Herpetological Review* 31: 13–15.

656 Uetz, P. 1996. The Reptile Database. Available at <http://www.reptile-database.org> (accessed 15
657 May 2015).

658 Venczel M, Hír J. 2013. Micro-computed tomography study of a three-dimensionally preserved

659 neurocranium of Albanerpeton (Lissamphibia, Albanerpetontidae) from the Pliocene of
660 Hungary. *Journal of Vertebrate Paleontology* 33:568–587.

661 Vidal N, Azvolinsky A, Cruaud C, Hedges SB. 2008. Origin of tropical American burrowing
662 reptiles by transatlantic rafting. *Biology Letters* 4:115–118.

663 Vitt LJ., Caldwell MW. 2009. *Herpetology: An Introductory Biology of Amphibians and*
664 *Reptiles*. Burlington: Elsevier.

665 Voloch CM, Vilela JF, Loss-Oliveira L, Schrago CG. 2013. Phylogeny and chronology of the
666 major lineages of New World hystricognath rodents, insights on the biogeography of the
667 Eocene/Oligocene arrival of mammals in South America. *BMC Research Notes* 6:1–9.

668 Voorhies MR, Holman JA, Xiang-xu X. 1987. The Hottell Ranch rhino quarries (basal Ogallala;
669 medial Barstovian), Banner County, Nebraska; Part I; Geologic setting, faunal lists, lower
670 vertebrates. *Contributions to Geology, University of Wyoming* 25:55–69.

671 Vrcibradic D, Rocha CFD. 2011. An overview of female reproductive traits in South American
672 *Mabuya* (Squamata, Scincidae), with emphasis on brood size and its correlates. *Journal of*
673 *Natural History* 45:813–825.

674 Whiting AS, Sites Jr JW, Pellegrino KCM, Rodrigues MT. 2006. Comparing alignment methods
675 for inferring the history of the new world lizard genus *Mabuya* (Squamata, Scincidae).
676 *Molecular Phylogenetics and Evolution* 38:719–730.

677 Yang Z. 2007. PAML 4: Phylogenetic Analysis by Maximum Likelihood. *Molecular Biology*
678 *and Evolution* 24:1586–1591.

Table 1(on next page)

General information on the loci used in this study.

1

	<i>12S</i>	<i>16S</i>	<i>enol</i>	<i>cytb</i>	<i>cmos</i>	<i>gapdh</i>	<i>myh</i>	<i>gpr149</i>	Total
Number of sequenced species	214	194	70	153	146	47	51	73	218
Alignment length	366	464	192	1,033	894	380	133	669	4,131
Frequency of sites with missing data	0.12	0.20	0.74	0.38	0.40	0.81	0.81	0.72	0.47
Frequency of indels	0.02	0.03	0.01	0.16	0.22	0.04	0.01	0.01	0.10

2

Table 2 (on next page)

Model comparisons performed in 'diversitree'.

Constraints in the macroevolutionary models subjected to ANOVA for differences in the rates of speciation (λ) and extinction (μ) between New World (NW) and Old World (NW) areas (GeoSSE) and between viviparous (V) and non-viviparous (V) lineages (BiSSE).

Model	Approach	Constraints	Parameters
Reduced full	GeoSSE	NA	$\lambda_{NW}, \lambda_{\overline{NW}}, \mu_{NW}, \mu_{\overline{NW}}$
	BiSSE	NA	$\lambda_V, \lambda_{\overline{V}}, \mu_V, \mu_{\overline{V}}$
Equal speciation	GeoSSE	$\lambda_{NW} = \lambda_{\overline{NW}}$	$\lambda, \mu_{NW}, \mu_{\overline{NW}}$
	BiSSE	$\lambda_V = \lambda_{\overline{V}}$	$\lambda, \mu_V, \mu_{\overline{V}}$
Equal extinction	GeoSSE	$\mu_{NW} = \mu_{\overline{NW}}$	$\lambda_{NW}, \lambda_{\overline{NW}}, \mu$
	BiSSE	$\mu_V = \mu_{\overline{V}}$	$\lambda_V, \lambda_{\overline{V}}, \mu$
Equal diversification	GeoSSE	$\lambda_{NW} = \lambda_{\overline{NW}}$	λ, μ
		$\mu_{NW} = \mu_{\overline{NW}}$	
	BiSSE	$\lambda_V = \lambda_{\overline{V}}$ $\mu_V = \mu_{\overline{V}}$	λ, μ

1
2
3

Table 3(on next page)

Model comparisons performed in BioGeoBEARS

Likelihood-ratio tests between null models (DIVALIKE and DEC) and more sophisticated models with the addition of the founder-event speciation (+J) (DIVALIKE+J and DEC+J). (LnL) Likelihood value. (DF) Degrees of freedom for the chi-square test.

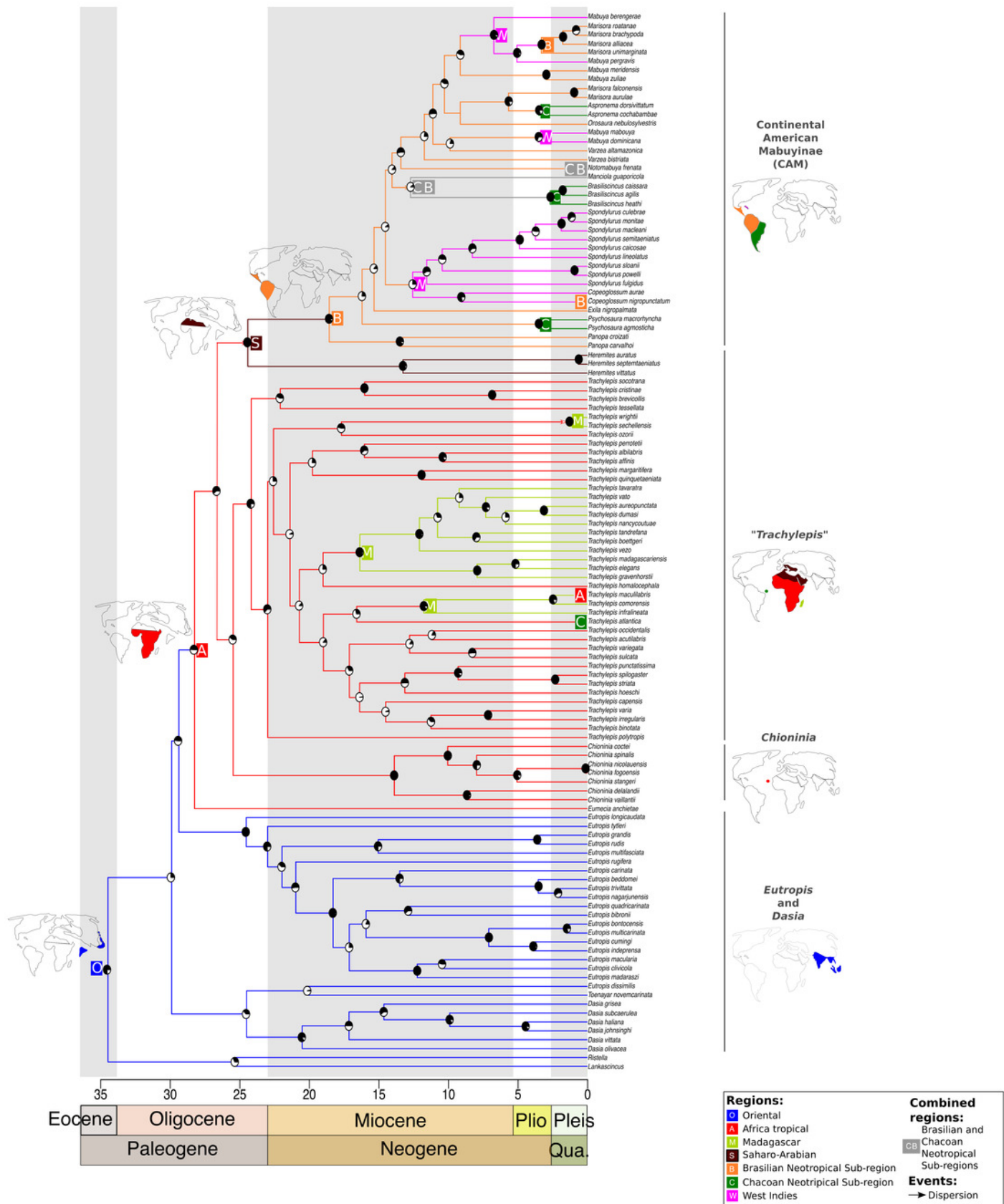
model	Null model	LnLalt	LnLnull	DF	pval
DEC+J	DEC	-94.34	-112.8	1	1.20E-09
DIVALIKE+J	DIVALIKE	-95.65	-119.2	1	6.70E-12

1

Figure 1

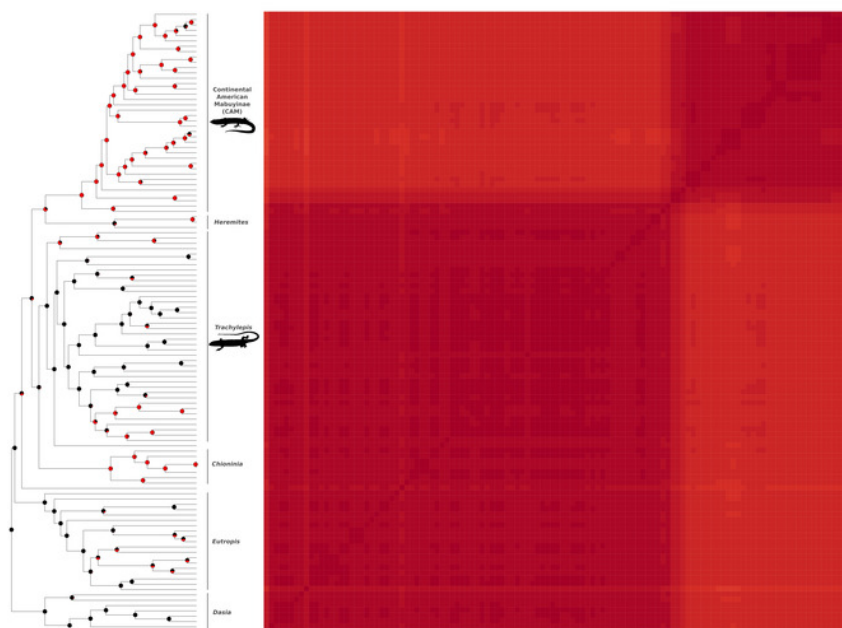
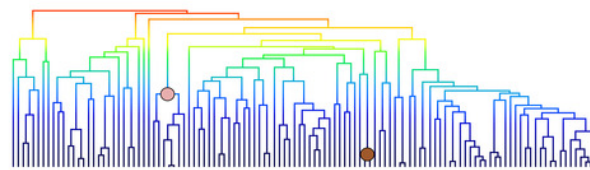
Time-dated phylogeny of the Mabuyinae with ancestral area reconstruction

Maps depict putative dispersals and vicariant events that culminated in the occupation of South America by this group. Ancestral area reconstruction was based on the results from DEC+J because this model produced a significantly higher log-likelihood score. The black regions in pie charts represent bootstrap supports.



Macroevolutionary cohort matrix for the Mabuyinae subfamily.

A



B

