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Chasing the Rainbow: Decoupled phenotypic and genotypic evolution in New Guinea's rainbow skinks (Squamata: Scincidae)

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Abstract

With the advent of molecular data, the discovery of cryptic species has become commonplace. New Guinea, a region of high vertebrate biodiversity and complex geological history, has been found to contain immense numbers of cryptic skink species. We present the first molecular phylogenetic analysis of *Carlia* Gray, 1845, and its sister genus *Lygisaurus* de Vis, 1884, across mainland New Guinea and the Solomon Islands. We find rainbow skinks exhibit significant genetic divergence with minimal morphological variation and our data suggest the existence of many undescribed species. Due to the morphologically cryptic nature of rainbow skinks, we demonstrate the efficacy of the COI gene as a "barcode" for difficult species determinations.

Divergence time and biogeographic analyses support four separate dispersal events from Australia to New Guinea for rainbow skinks from ~10–5 mya, with most groups arriving first in the East Papua Composite Terrane (EPCT) and dispersing from there to other terranes and islands. Exceptions to this pattern were observed in *Lygisaurus,* which dispersed first to the West Papuan portion of the Craton ~8.4mya, and the island clade of the *Carlia fusca* group, which dispersed to the Vogelkop peninsula ~4.7mya and from there to many islands.

Key words: Rainbow Skinks, Phylogenetics, Papua New Guinea, Vicariance, *Carlia, Lygisaurus,* Biogeography

Introduction

Historically, species delimitation has been based on distinct morphological characters. With the advent of DNA sequencing and molecular technologies, thousands of cryptic species have been identified (Li & Weins 2023, Funk *et al.* 2012, Oliver *et al*. 2009). Often, morphological characters are found *a posteriori* to support the molecular based species designations, but there are groups where phenotype does not appear to change at the same rate as genotype (Bruna *et al.* 1996; Cerca *et al.* 2020; Lavoué *et al.* 2011; Prates *et al.* 2023a; Prates *et al.* 2023b; Vacher *et al.* 2020). Mounting evidence suggests the presence of cryptic species to be far more commonplace in nature than previously predicted (Pérez-Ponce de León & Poulin 2016; Pfenninger & Schwenk 2007), at times accounting for more than half the species richness in a lineage (Adams *et al.* 2014; Eme *et al.* 2018; Lukić *et al.* 2019). Four causes have been identified for morphological similarity between genetically distinct species: parallelism, convergence, recent divergence, and molecular stasis (Fišer *et al.* 2018; Struck *et al.* 2018). Of these, recent divergence is the most common explanation for morphological similarity (Monro & Mayo 2022), resulting from a lack of time for the species to accumulate significant morphological differences (Knowlton 1993; Korshunova *et al.* 2017; Reidenbach *et al.* 2012). Yet deep genetic divergences with low morphological variation is a repeatedly observed phenomenon in many skink lineages (Austin 1995; Barley *et al*. 2013, Bruna *et al*. 1996, Linkem *et al.* 2010, Kornilios *et al*. 2018; Prates *et al.* 2023b).

New Guinea is the second largest island on the planet and is recognized as an area of high vertebrate biodiversity (Austin *et al.* 2008; Cámara-Leret *et al.* 2020; Mittermeier *et al.* 2003; Stattersfield 1998; Toussaint *et al.* 2014).

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Despite the immense biodiversity, New Guinea remains a taxonomic knowledge gap for many groups. The diversity of skinks in New Guinea is second only to Australia (Chapple *et al*. 2023), and recent molecular studies have found large numbers of new and cryptic species (Austin *et al.* 2011; Slavenko *et al.* 2023; Slavenko *et al.* 2020; Slavenko *et al.* 2022). New Guinea emerged from the collision of the Australian and Pacific plates (Hall & Spakman 2003; Johnson & Molnar 1972), with further interactions from the Sunda plate forming Northwestern New Guinea (Baldwin *et al.* 2012; Bird 2003). This tectonic activity resulted in the rapid formation of the central mountain range (Davies 2012) and high volcanic activity in the surrounding region, leading to the formation of Northern New Guinea and various island arcs (Baldwin *et al.* 2012; Lindley 1988). While the timing and sequence by which these events occurred is still debated (Dow 1972, 1977; Davies 2012; Hall 1998; 2002; Pigram and Davies 1987; Davies *et al*. 1996; Quarles van Ufford and Cloos 2005), it is well accepted that New Guinea is composed of 5 major geologic regions: the Australian Craton, the East Papua Composite Terrane (EPCT), the Fold Belt, the Accreted Terrane, and the Vogelkop Peninsula (Davies 2012). The complex geologic history of New Guinea has likely played a critical role in shaping species distributions and driving speciation.

Named for their iridescent scales and colorful male breeding coloration, the rainbow skinks are distributed across Australia, New Guinea, Wallacea, and the Solomon Islands. Most species are identified by the distinct breeding coloration of males, while morphological scalation characters used in species descriptions are often continuous and overlapping rather than discrete (Allison and Zug 2006; Stuart-Fox *et al.* 2002, Zug 2004). This is problematic when dealing with preserved museum specimens where color rapidly fades and makes species delimitation and recognition difficult (Kraus 2007; Zug 2004). Studies have found up to 10% sequence divergence among closely related species with only minor scalation differences (Couper *et al.* 2005; Donnellan *et al.* 2009). Currently rainbow skinks are composed of 64 species across three genera (*Lygisaurus, Liburnascincus* Wells and Wellington 1983, and *Carlia*) with the bulk of diversity found in north-eastern Australia and New Guinea. Relationships among the Australian rainbow skinks have historically been disputed due to the lack of morphological characters and the speed at which the group diversified. However, a multitude of recent molecular studies have established a well-supported phylogeny, including the resurrection of the genus *Lygisaurus*, creation of the genus *Liburnascincus*, and the description of many new species (Austin *et al.* 2011; Bragg *et al.* 2018; Dolman & Hugall 2008; Donnellan *et al.* 2009; Hoskin & Couper 2015; Ingram & Covacevich 1988, 1989; Stuart-Fox *et al.* 2002; Wells & Wellington 1983)

Within New Guinea, rainbow skinks are represented by three distinct species groups; *Carlia bicarinata* group, *Carlia fusca* group, and *Lygisaurus novaeguineae* group. Each group is thought to represent a separate colonization event from Australia followed by rapid diversification (Dolman & Hugall 2008; Stuart-Fox *et al.* 2002), consistent with the radiation of other scincid genera found in the region (Skinner *et al.* 2013). Kraus (2007) reviewed the *Lygisaurus novaeguineae* group, distinguished by small body size and smooth scales, from the southern portion of New Guinea and resurrected two species, *L. macfarlani* Günther 1877, and *L. curtus* Boulenger 1897*,* based on morphological data. The *C. bicarinata* group consists of three species (*C. Schmeltzii* Peters 1867*, C. bicarinata* Macleay 1877*,* and *C. storri* Ingram and Covacevich 1989) found in northern Australia and southern New Guinea, distinguished by small slender bodies, moderately sized limbs, and strongly bicarinate scales although *C. schmeltzii* has both bicarinate and tricarinate populations (Ingram & Covacevich 1989; Zug 2004, 2010). The *C. fusca* group, comprised of 18 species across Northern Australia, New Guinea, Wallacea, and the Solomon Islands (Donnellan *et al.* 2009; Zug 2004; Zug & Allison 2006), is united through weakly tricarinate to smooth scales, larger body size, and more robust limbs (Zug 2004, 2010). Zug's (2004) systematic morphological revision of the *C. fusca* group was the most complete work to date on the group and included the description of 6 new species but also left multiple populations in New Guinea unassigned. An additional two species were described from western New Guinea (Zug & Allison 2006), and *C. longipes* Macleay 1877 (distributed in northern Australia and the Torres strait) was split into three species (Donnellan *et al.* 2009). While investigating invasive island populations, Austin *et al.* (2011) found significant disagreement between molecular data and Zug's (2004) species delimitations for *C. ailanpalai* Zug, 2004*, C. mysi* Zug, 2004*, and C. tutela* Zug, 2004, again emphasizing the decoupling of phenotypic and genotypic evolution within rainbow skinks. Given the dearth of useful morphological characters, the apparent rapid diversification of rainbow skinks, and the complex geological history of New Guinea, we seek to test previous morphological species delimitations by generating a molecular phylogeny and inferring the biogeographic history of rainbow skinks in New Guinea.

Materials and Methods

Sampling

Tissues were obtained from 73 specimens and sequences for an additional 62 individuals were obtained from GenBank (see supplementary data, table S1). Sampling included all rainbow skink species from mainland New Guinea, with the exception of *C. fusca* Duméril and Bibron, 1839, and *C. diguliensis* Kopstein, 1926, (see Fig. 1 and 2 for sampling localities). Representatives of all three Australian rainbow skink genera were included, and two species of the closely related Eugongyline genus *Cryptoblepharus* Wiegmann 1834, were used as the outgroup (Brandley *et al.* 2015; Chapple *et al.* 2023).

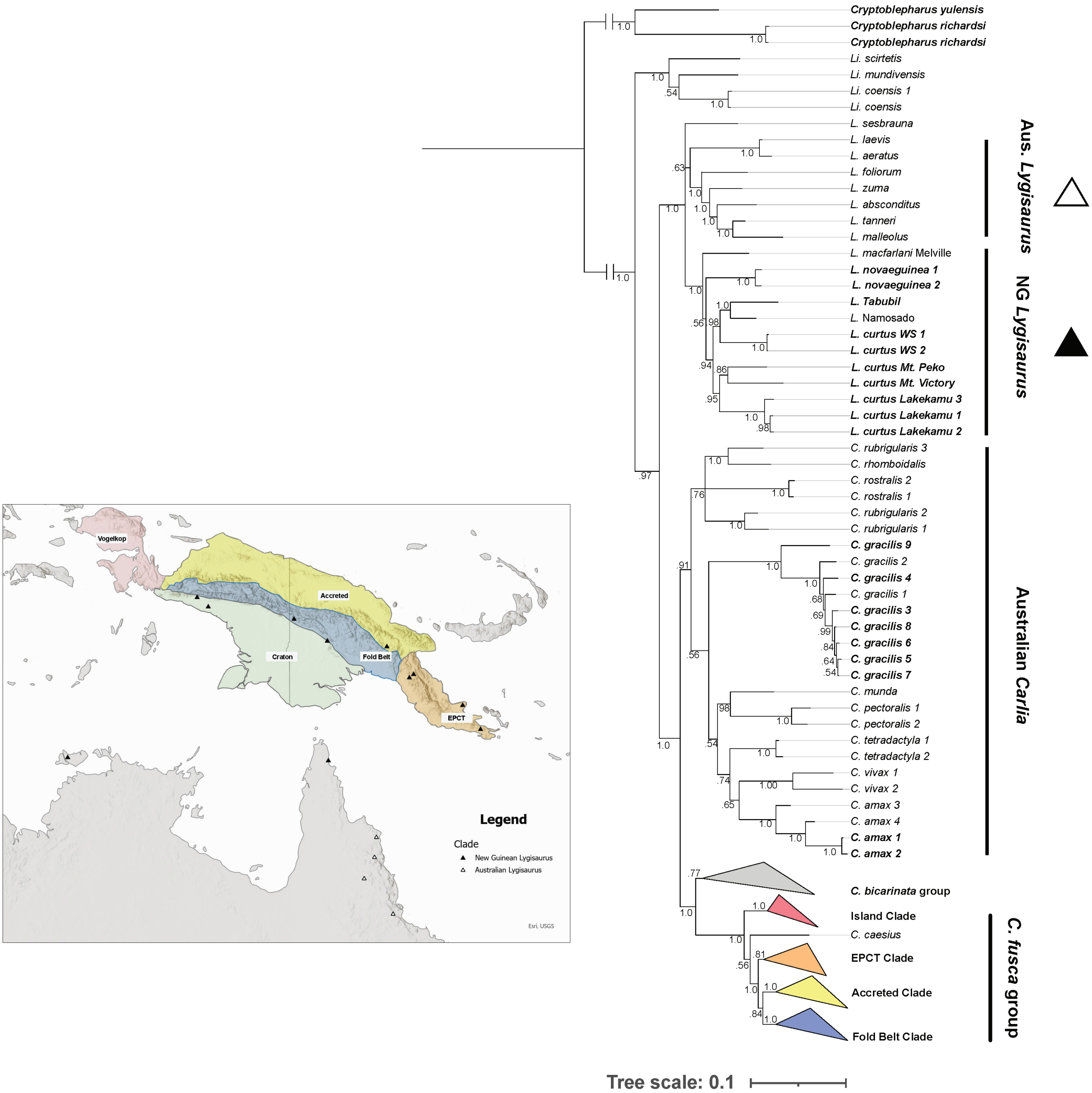

FIGURE 1. Phylogenetic relationships between three genera of rainbow skinks based on Bayesian analysis of the concatenated dataset. Posterior probabilities are listed below branches. *Liburnascincus* and *Lygisaurus* are represented by *Li.* and *L.* respectively. Clades within the *Carlia fusca* complex have been collapsed and are shown in detail in figure 2. A sampling map is provided for *Lygisaurus* with the 5 major geological terranes of New Guinea labeled according to Davies (2012) and drawn based on Hill *et al.* (2023). Clades and colors within the *Carlia fusca* group reflect relationships shown in figure 2.

Molecular methods

DNA was extracted from preserved tissues using the Zymo Research Quick-DNA Miniprep Plus Kit (Cat#D4068) following the solid tissues protocol with the following adaptations: tissue samples were incubated in Proteinase K overnight before DNA extraction, and DNA elution buffer was heated to 55°C before use. Portions of three mitochondrial genes; NADH dehydrogenase subunit 4 (ND4), 16S rRNA (16S), and cytochrome C oxidase subunit 1, as well as five nuclear genes; oocyte maturation factor (C-mos), aldolase exon 5(ALD), β-globin-intron 2 (β-Globin-2), recombination activating gene 1 (Rag-1), and myosin heavy chain 2 (MHC2), were amplified using the primers and cycling profiles listed in Supplementary data (Table S2). PCR was performed using Cytiva PuReTaq Ready-To-Go PCR Beads, and products were sent to Eton Bio (https://www.etonbio.com/) for purification and Sanger sequencing. Chromatographs were aligned in Pearl (https://www.gear-genomics.com) (Rausch *et al*. 2020) and checked manually for sequencing errors and single nucleotide polymorphisms.

Alignment and Phylogenetic inference

Sequences were aligned using MUSCLE (Edgar 2004) with UPGMA clustering as implemented in MEGA v11.0.1 (Tamura *et al.* 2021). Models of DNA sequence evolution were inferred via Akaike Information Criteria (AIC) in IQTREE v1.6.12 (Nguyen *et al.* 2015) for the following partitions; individual genes, mitochondrial genes, nuclear genes, and total concatenated dataset. Trees were constructed using Bayesian Inference (BI) with default priors in MrBayes v3.2.7a (Ronquist *et al.* 2012), run for 1E7 generations, sampling every 1000 generations, and discarding the first 25% as burn-in. Trees were visualized using Interactive Tree of Life (iTOL) (Letunic & Bork 2021).

Divergence Time Estimation and Biogeographical Reconstruction

We used the Chronos function through the ape package v5.8 (Paradis & Schliep 2019) in R to calibrate an ultrametric starter tree based on our concatenated Bayesian analysis with the following arguments (lambda = 0.1, model = relaxed). The minimum and maximum age for the MRCA of rainbow skinks was specified to be (12–22mya) according to the secondary calibration date of Chapple *et al*. (2023).

Divergence times were estimated with BEAST v2.7.7 (Bouckaert 2019; Drummond *et al*. 2012) using the concatenated data set under the substitution model GTR + F + R4. We used a birth-death speciation model with a lognormal relaxed clock. Due to a lack of fossil priors, we specified the prior divergence time as previously estimated in Chapple *et al*. (2023) to be 20.8 ± 2 Mya under log normal distribution (M = 3.035, S = 0.096). All unspecified priors were set as default. We conducted our run for 1E7 generations, with sampling at intervals of every 1000 generations. Tracer v1.7.2 was used to examine convergence, posterior trace plots, effective sample sizes, and burn-in. To generate a maximum clade credibility tree based on sample trees, we used TreeAnnotator with a 25% burn-in.

To examine the biogeographical history of New Guinean rainbow skinks, we performed biogeographic reconstruction analyses. Each sample in our analysis was assigned to a discrete region in New Guinea and surrounding islands based on the collection locality: Australia, EPCT, Accreted Terranes, Vogelkop Peninsula, Craton, Fold Belt, New Britain, Manus, New Ireland, Mariana Islands, Fergusson Island, Woodlark Islands, Sudest Island, Torres Strait, and Halmahera. The range of *Cryptoblepharus* was set to Australia given strong evidence for Australia being the point of origin for the common ancestor of *Cryptoblepharus*, *Saproscincus* Wells and Wellington 1983*, Lampropholis* Fitzinger 1843*, Lygisaurus, Liburnascincus*, and *Carlia* (Chapple *et a*l. 2023).

We performed the reconstruction on our time-calibrated phylogeny using the R package 'BioGeoBEARS 1.1.3' (Matzke 2013). We tested the dispersal-extinction-cladogenesis (DEC) against the DEC+J model, which includes an additional parameter allowing for founder-event speciation via jump dispersal (Matzke 2014). We ran this on the unconstrained data set to allow for all possible combinations, with the maximum range size limited to three regions.

Morphology

Morphological characters were recorded from 102 adult *Carlia* specimens representing four geographic regions that also formed monophyletic clades in the molecular analysis: Maimafu (m=7, f=1), East New Britain (m=19,f=8), Haia (m=11, f=1), and Lakekamu f=20, m=36. Localities with limited sampling (<8) were excluded, as was Alotau due to the discovery of two distinct species in this locality. The following measurements were taken: snout-vent length (SVL), head length (HL), head width (HW), head depth (HD), pectoral width (PeW), pectoral depth (PeD), pelvic width (PlW), pelvic depth (PlD), arm length (AL), forearm length (FAL), manus length (ML), femur length (FL), crus length (CL), and pes length (PL) according to Slavenko (2021). Limb measurements were taken from the left side of the body, unless damaged. Measurements were taken with digital calipers and rounded to the nearest .5 mm. The sum of arm length, forearm length, and manus length was represented as a single measurement: forelimb length (FrL). Similarly, femur length, crus length, and pes length were summed into hind limb length (HiL). Sex and reproductive maturity were determined through dissection and examination of gonads. All statistical analyses were conducted in R v4.4.0 (R Core Team 2013) through the stats package v4.4. To remove the effect of body size, residuals were taken from linear regressions against SVL for the individual measurements, which were then used in subsequent analyses. To ensure a normal distribution of variables, histograms were analyzed for each length measurement. Multivariate analysis of variance (MANOVA) was used to determine if there was a statistical difference within populations due to sexual dimorphism. If a significant difference exists between males and females, Welch's T-Test was applied to each measurement variable. Variables with significant differences due to sex were removed from interpopulation comparisons. To visualize the morphospace occupied by *Carlia* populations, we used principal component analysis (PCA) applying the broken stick method to determine which principal components were significant contributors to variance (Jackson 1993). The Analysis of Variance (ANOVA) test was used to compare the residual means of the morphological variables between the various geographic locations. If a statistical difference between localities was detected in a variable, we applied the Tukey Honesty Significant Difference test to assess the significance of differences.

DNA Barcoding

To test the efficacy of the COI-5P region in the identification of rainbow skinks, we calculated raw distances using sequences from this study and additional sequences obtained from BOLD (Ratnasingham & Hebert 2007) consisting of a total of 135 *Carlia* sequences and 37 *Lygisaurus* sequences. MEGAv11.0.1 (Tamura *et al.* 2021) was used to compute pairwise distances between individuals, and haplotype networks were constructed in SplitsTree v4.11 (Huson & Bryant 2006) under the NeighborNet algorithm (Bryant & Moulton 2004) with branch lengths used as an estimation of sequence divergence.

Results

Phylogenetic analyses

The total concatenated dataset consisted of 4,758 base pairs (best fit model = GTR + F + R4). Best fit models for each partition are as follows: ND4 (755bp, TIM2+F+R4), 16S (480bp, TIM2+F+I+G4), COI (715bp, TPM2u+F+R3), RAG-1 (810bp, TPM3u+F+I+G4), β-Globin-2 (853bp, TVM+F+G4), C-mos (400bp, TN+F+G4), ALD (235bp, SYM+R2), MHC-2 (510bp, TN+F+G4), mitochondrial (1950 bp, GTR+F+R4), and nuclear (2,808bp, TVMe+R3). We find no well supported conflict between the partitioned and concatenated results (see Supplementary Figure 1), therefore results from the concatenated analysis are presented here.

The concatenated Bayesian analysis (Fig. 1) strongly supports (1.0pp) the monophyly of all three genera of rainbow skinks, with *Liburnascincus* sister to *Lygisaurus* + *Carlia* (.97pp), as found in Dolman & Hugall (2008) but in contrast to Bragg *et al*. (2018). Within *Lygisaurus* our results support two distinct clades, one composed of Australian species, and the other with the Australian *L. macfarlani*, (referred to as *L. novaeguineae* Meyer, 1874, in Dolman & Hugall 2008; changed here based on locality) sister to all New Guinean samples. The placement of *L.*

sesbrauna Ingram & Covacevich, 1988, is problematic and unresolved in the concatenated tree. *Lygisaurus curtus,* is monophyletic (.94pp) with long branch lengths and deep genetic distances across localities.

Within *Carlia*, the *fusca* group is sister to the *bicarinata* group, with all remaining Australian taxa sister this clade (1.0pp). Support for the monophyly of the *C. bicarinata* clade is low (.77pp) and results in a basal tritomy, due to the placement of *C. schmeltzii*. Our results add to the support for the monophyly of the *C. fusca* group (1.0 pp) (Dolman & Hugall 2008; Donnellan *et al*. 2009; Stuart-Fox *et al*. 2002; Zug 2004) and divide it into four well supported clades, with the exception of *C. caesius* Zug and Allison, 2006 where placement is poorly supported (Fig. 2).

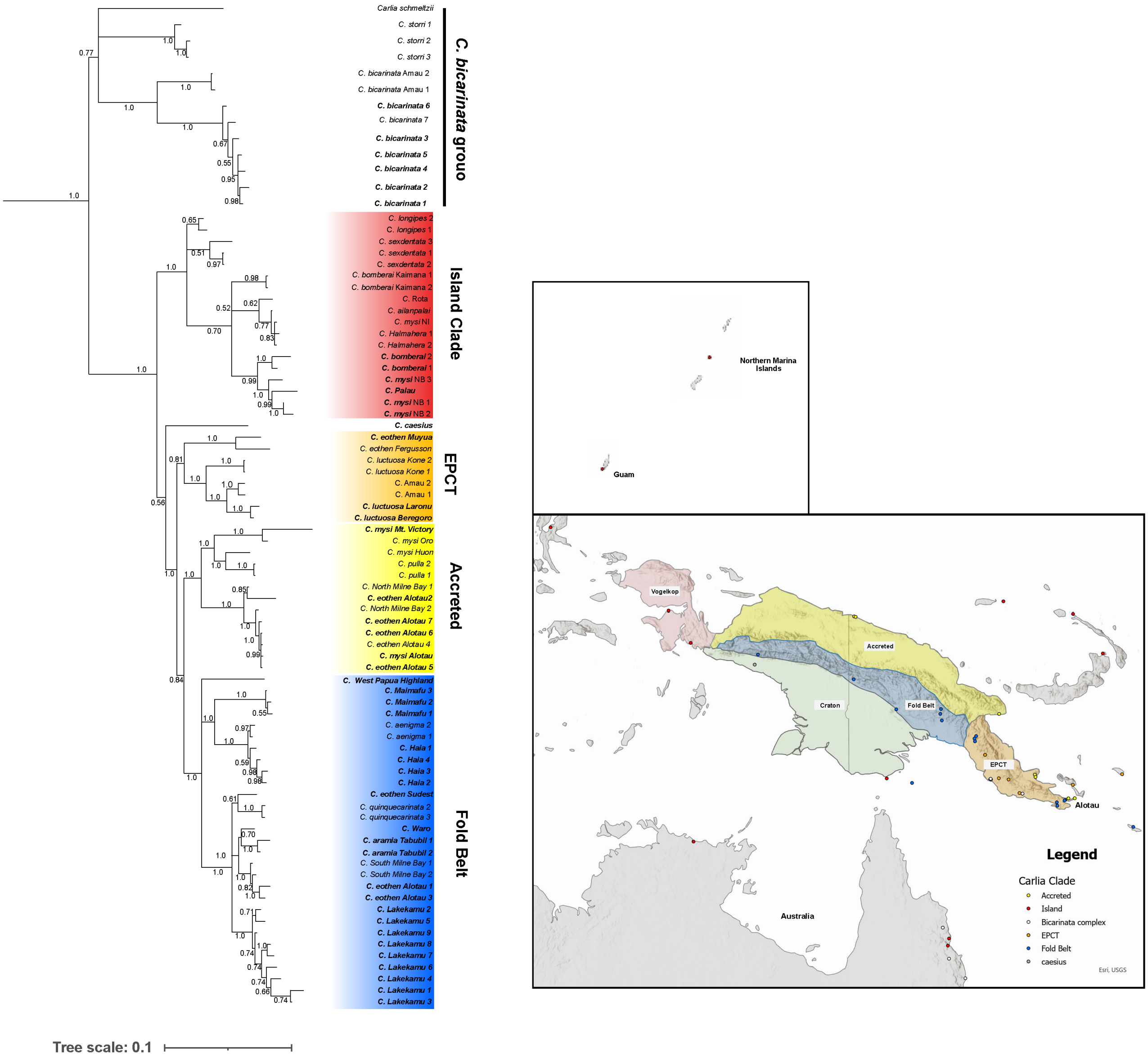

FIGURE 2. Pruned subtree based on Bayesian analysis of the con catenated dataset showing expanded relationships within the *Carlia fusca* and *C. bicarinata* groups. Posterior probabilities are displayed below branches. Clades are color coded according to their primary geologic terrane, and taxon names reflect the current species description and sampling locality. Taxa in bold represent sequences generated in this study, while those in plain text were taken from GenBank. The map shows sampling localities color coded by clade.

Estimation of Divergence Times and biogeographic history

We find the DEC+J model to be a significantly better fit to our phylogeny compared to the DEC model (AIC 268.4 v.s 333.7). There are problems with the use of the DEC+J model (Ree & Sanmartín 2018) with models returning zero dispersal and extinction probabilities. We find similar problems in our biogeographical analysis with d=0 (dispersal) and e=0 (extinction). Although this model seems appropriate for testing biogeographical hypotheses of *Carlia* considering their widespread distribution across the Islands of Oceania (Zug 2004; Zug & Allison 2010; Donnellan *et al*. 2009), we recommend caution in the interpretation of the results. We find Australian *Carlia* and the *fusca* + *bicarinata* group diverge ~16.9 mya (Fig. 3, for confidence intervals on dates see Supplementary Figure S2), followed by the rapid radiation of the *fusca* group beginning ~10.4 mya in Australia. Our data support four independent colonizations of New Guinea from Australia; single invasions by *Lygisaurus* and the *C. bicarinata* group, and two by the *C. fusca* group.

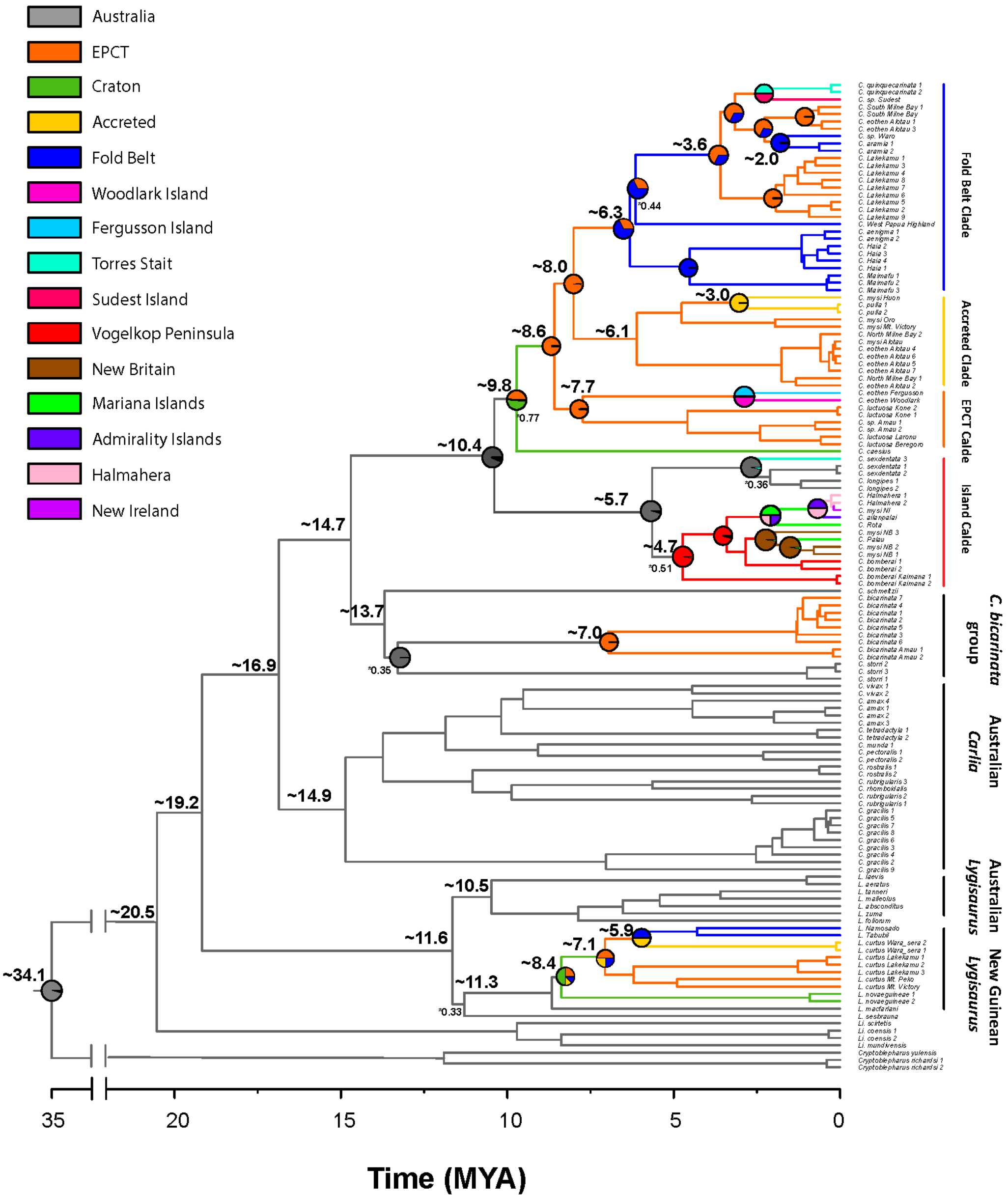

FIGURE 3. Time calibrated biogeographic reconstruction of rainbow skink dispersal under the best fit DEC+J model generated in BioGeoBears. Node probabilities are represented by pie charts, with the size of colored slices corresponding to the probability of each state at the given node. Nodes with 100% probability and unchanged from the previous node are not listed. For important colonization events, the mean node age is given to the left of the node. The color of the terminal branches represents the area from which the specimen originated. For relationships of New Guinean rainbow skinks that were unresolved in our phylogenetic analysis but resolved here, posterior probabilities are listed under the node indicated by an asterisk (*).

New Guinean *Lygisaurus* diverged ~8.7 mya, dispersing to the Craton and then the EPCT, while the *C. bicarinata* group dispersed directly to the EPCT (~7.0mya). The first invasion of the *C. fusca* group to New Guinea occurred ~9.8 mya into either the Craton or the EPCT (the relationships between *C. caesius* from the Craton is poorly resolved in our phylogenetic analysis (.56, see Fig. 2) and the probability of colonization through the Craton is only slightly higher than the EPCT). The EPCT appears to have served as a "launching point" for dispersal to the fold belt and Accreted terranes as well as island dispersal and speciation, with Darnley, Fergusson, Woodlark, and Sudest populations all originating from the EPCT. The second *fusca* group colonization event involves the dispersal of the Island clade to the VogelKop peninsula (~4.7mya), with additional long-distance dispersal events to Halmahera, New Ireland, Palau, and New Britain.

Morphometric analyses

All morphological measurements are reported in supplementary data (Table S3). MANOVA tests revealed significant sexual dimorphism in New Britain *Carlia* (p = 0.01), but not in Lakekamu (p=.174), as Welch's T-test showed significant differences in HL (p=0.00065, t= 4.20), HW (p=0.0076, t = 3.014), and PlD (p=0.0039, t= 3.50). To remove the effects of sexual dimorphism in interpopulation comparisons, these characters (HL, HW, and PlD) were removed from subsequent analyses.

The Broken stick method revealed only PC1 (pectoral width, pelvic width) and PC6 (pelvic depth, pelvic width) (Table 1) of our Principal Components Analysis to explain a substantial portion of variance, explaining 41.5% of the cumulative variance. The morphospace resulting from PCA reveals considerable overlap between regions (Fig. 4). Results of Tukey Honesty significant difference tests of ANOVA results are shown in Table 2, with significant differences marked with an asterisk. New Britain and Maimafu were found to contain no significant differences in characters analyzed. No more than 1–2 significant differences were found between other regions apart from New Britain and Lakekamu, with Lakekamu specimens possessing wider bodies and longer forelimbs.

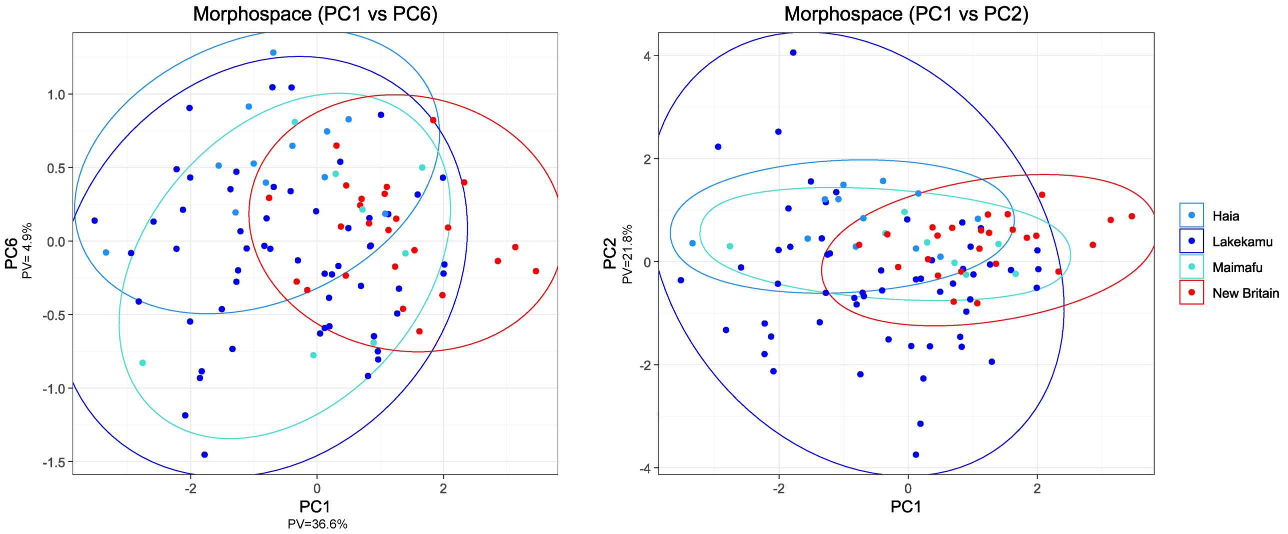

FIGURE 4. *Carlia* morphospace visualized by statistically significant principal components (PC1 and PC6) and PC2, depicting morphometric variables and their variance between regions. Samples are represented by a point, with an ellipse drawn around all points in a population to visualize the area occupied. The populations are color coded according to their designated clade from our phylogenetic analysis, with New Britain from the Island clade and Lakekamu, Maimafu, and Haia from the Fold Belt. Significant overlap is found between Carlia from all regions.

DNA Barcode

COI haplotype networks were able to differentiate between recognized species and identify multiple clades that may represent new species (see Fig. 5; for simplicity only the network for *Lygisaurus* is shown, see Supplementary Figure S2 for the *Carlia* haplotype network).

TABLE 1. Principal component analysis loadings from the Broken stick method. PC1 and PC6 were found to be significant.

Character	PC1	PC2	PC6
HD	-0.27401	0.681675	-0.47447
PeW	-0.50042	-0.26053	0.274893
PeD	-0.42909	0.508985	0.501551
PlW	-0.50493	-0.36697	-0.5988
FrL	-0.42801	-0.25657	0.26225
HiL	-0.22852	0.088809	-0.14289

TABLE 2. Results of Tukey Honesty Significant Differences test run on results of ANOVA. Statistically significant results are denoted by an asterisk (*).

Populations	HD	PeW	PeD	PlW	FrL	HiL
Lakekamu-Haia	-0.383	0.104	-0.88*	0.795*	0.0639	-0.884
New Britain-Haia	-0.467	-0.55	-1.18*	-0.216	-1.067*	-0.791
Maimafu-Haia	-0.537	-0.362	-0.57	0.371	-1.265	-2.165*
Maimafu-Lakekamu	-0.154	-0.466	0.309	-0.424	-1.329*	-1.281
New Britain-Maimafu	0.0696	-0.188	-0.61	-0.588	0.198	1.374
New Britain-Lakekam	-0.0847	-0.654*	-0.301	-1.011*	-1.131*	0.092

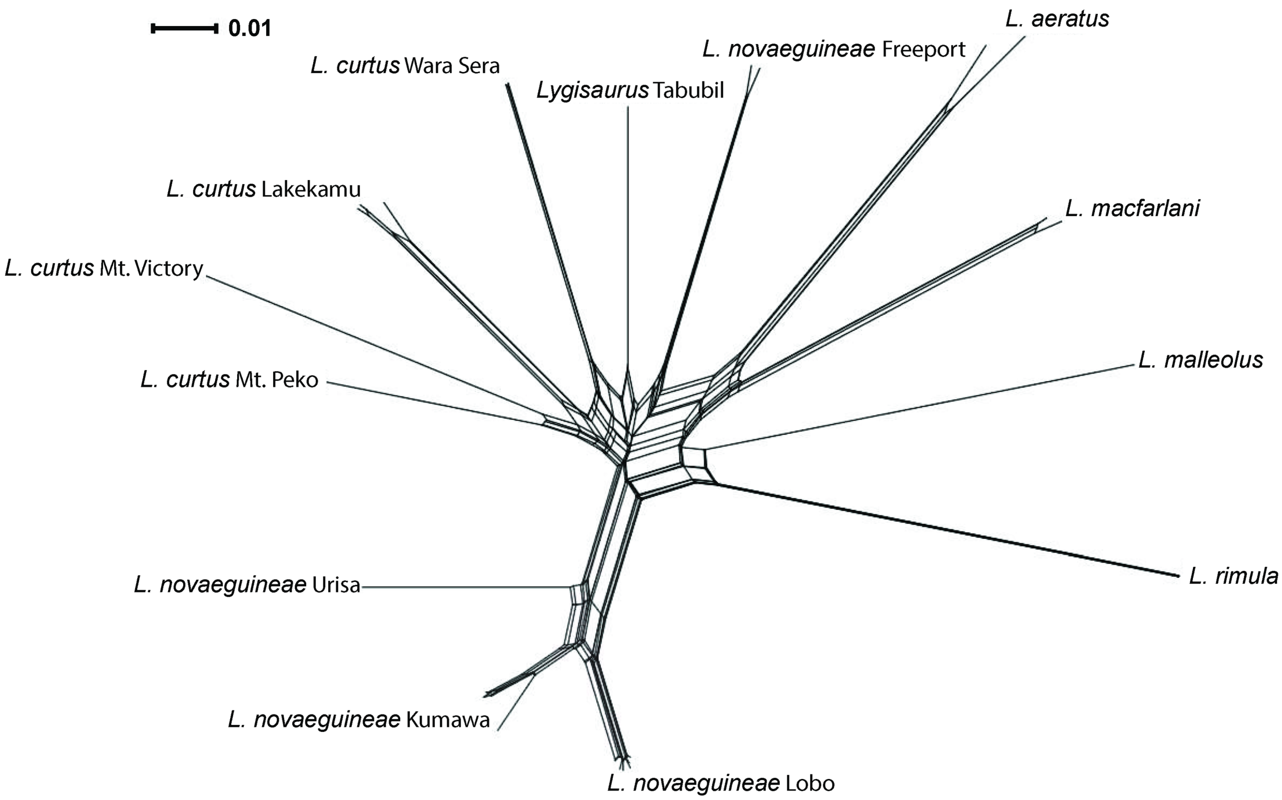

FIGURE 5. Haplotype Network for *Lygisaurus* generated from COI-5P sequences from this study and BOLD. Currently recognized species are found to represent distinct branches. Significant genetic divergence is found between geographic localities of *L. curtus*, and between *L. novaeguineae* from Bird Head Peninsula and Freeport.

Discussion

Phylogeny

Our molecular phylogeny highlights problems with previous morphological delimitations of rainbow skinks, similar to recent findings in other skink genera (Prates *et al*. 2023b; Slavenko *et al.,* 2020, 2021). As currently defined, *Carlia eothen* Zug 2004*, C. mysi*, *C. bomberai* Zug and Allison 2006*,* and *C. ailanpalai* are all paraphyletic and likely represent multiple species. Deep genetic divergence between localities of *C. mysi, C. luctuosa* Peters and Doria 1878*,* and *Lygisaurus curtus* suggests the presence of species complexes with multiple undefined species. Additional potential undescribed species are found in the *C. fusca* group in the following localities; the Lakekamu basin, central highlands (Maimafu), West Papua Highlands (Freeport), and the Milne Bay province (labeled in Fig. 2 by their localities). Additional species of rainbow skinks will undoubtedly be revealed with added sampling. The inclusion of in-life color patterns, morphological data, and more complete geographic sampling is necessary to describe and define the boundaries of these new species.

Despite evidence for the existence of several cryptic species, we find only a sole case of allopatric species within the *Carlia fusca* group; the coastal town of Alotau in the Milne Bay province. Austin (2011) lists two undescribed *Carlia* species from the Milne Bay province, with *C. sp.* "North Milne Bay ‘' containing samples from the northern coast to the Eastern Cape, and *C. sp*. "South Milne Bay" distributed from Alotau through the south of the province. In this study, samples from a single locality in Alotau group with both Austin's north and south Milne Bay species. Zug (2004) hinted at the possibility of two *Carlia* species within Milne Bay as he noted two distinct adult color patterns, yet no significant difference was detected in morphological characters (Zug 2004). Due to their distinct evolutionary history, we can assume that these two species came into sympatry secondarily due to range expansion.

While the molecular data has identified many cryptic species, we also find evidence of possible over-splitting within *Carlia.* Zug (2004) restricted *C. ailanpalai* to the Admiralty Islands, however Austin (2011) found that *Carlia* from the Bismarck archipelago, Halmahera, Palau, and the Mariana Islands (described as *C. mysi* and *C. tutela* by Zug, 2004) were all part of this same species group. Our results confirm Austin's findings and add additional populations found in West Papua (described as *C. bomberai* by Zug and Allison, 2006) to this group. Additional work is necessary to determine species boundaries within this group.

Biogeography

Results from divergence time and biogeographic analyses for rainbow skinks are similar to what has been found in other New Guinean clades including *Lobulia* Greer 1974*, Papuascincus* Allison and Greer 1986*, Prasinohaema* Greer 1974*,* and Microhylidae Günther 1858 (Hill *et al.* 2023); Hill *et al*. 2023), with original dispersal events to the EPCT followed by dispersal to the other terranes and islands. However, our results suggest later colonization dates of ~10–5 mya for the four rainbow skink dispersal events to New Guinea, as compared to 20–30 mya found in other groups. Clades resulting from the first invasion of the *C. fusca* (~9.8 mya) group appear to reflect the geological terranes of New Guinea (Davies 2012; Pigram 1987) (see Figure 2). The EPCT clade (~7.7 mya) is found on the southern side of Owen Stanley Mountain range and southeastern Islands. The Accreted clade (~6.1 mya) is distributed along the Northern Coast of the Accreted Terranes and EPCT, while the Fold Belt clade (~6.3 mya) is distributed primarily along the Fold Belt and the southern tip of the EPTC. The Craton is represented by a sole individual, *C. caesius,* whose placement in our phylogenetic analysis is recovered with low support.

Three primary hypotheses have been postulated to explain the geologic history of New Guinea (Hill *et al*. 2023), the "mobile belt" (Dow 1972, 1977; Davies 2012), "recent emergence", and "slow and steady" (Davies *et al*. 1997, Pigram and Davies 1987; Polhemus and Polhemus 1998; Quarles van Ufford and Cloos 2005; Davies 2012). Each hypothesis specifies different dates for the formation and docking of New Guinea's five terranes. Given that our results show *Carlia* and *Lygisaurus* colonizing the Craton or EPCT before 5 mya, the "recent emergence" hypothesis is not supported by our data. While our results cannot definitively reject either the "mobile belt" or "slow and steady" hypotheses, our Accreted *Carlia* taxa have a diversification date of ~3 mya, supporting a younger Accreted Terrane as found in the "slow and steady" hypothesis. However, because we used secondary calibrations to estimate divergence dates due to a lack of fossil priors, our dates may be erroneous (Schenk 2016) and not reflective of the true timing of divergence between clades. Although we find similar patterns of dispersal in *Lygisaurus,* we lack adequate sampling to demarcate discrete phylogenetic clades within the group. Furthermore, our *Lygisaurus* samples from the Accreted Terrane come from the Highlands of New Guinea and lie at the border of the Fold Belt and thus the Accreted Terrane may not be truly represented here. Both *Lygisaurus* and *Carlia* are widespread across Western Papua, the Torres Strait, and Wallacea; sampling of these areas is necessary to truly test biogeographic hypotheses.

Despite overall agreement with previous biogeographical studies, some patterns of dispersal remain perplexing, particularly within the Fold Belt clade. C*arlia* from Waro and Tabubil are recovered as sister group to *Carlia eothen* South Milne Bay despite these localities being geographically quite distant. Likewise, Sudest Island *Carlia* is found to be sister to *C. quinquecarinata* from the Torres Strait, despite a distance of nearly 1,000km. Similarly, the Island Clade defies traditional biogeographical hypotheses, dispersing from Australia to the Vogelkop Peninsula. The areas occupied by the Island Clade are some of the youngest formed in the region, supporting the much more recent date of dispersal for this clade of ~4.7 mya. Austin (2011) concluded human mediated dispersal was responsible for low genetic divergence between geographically disjunct populations of *C. ailanpalai*. However, between some localities within this clade the genetic divergence and divergence dates are greater than expected given human mediated dispersal.

Skinks have been shown to have impressive vagility across large bodies of water (Adler *et al.* 1995; Austin 1995; Austin & Zug 1999; Blom et al 2019; Kurita & Hikida 2014; Richmond *et al*. 2021; Whiting *et al*. 2006), and the small body size and oviparity of these skinks make them ideal for successful waif dispersal (Escoriza 2023). While life history strategies of *Carlia ailanpalai* remain unknown, reproductive studies in the Mariana Islands show year-round breeding with an average clutch size of n=20 (Goldberg & Kraus 2012), demonstrating their high reproductive capability, which would allow for rapid colonization of new islands. While the exact dispersal patterns of rainbow skinks cannot be determined in this study, we can conclude that overwater dispersal appears to be much more common than previously believed. Given the distribution of the *C. ailanpalai* group, sampling the islands of Wallacea, Micronesia (Buden 2009), and other West Papuan coastal populations (i.e. *C. fusca*) is likely to reveal additional populations of this widespread group, clarify species boundaries, and the pattern of dispersal.

Like island colonization and isolation, the formation of montane barriers has served as a key vicariance event for many squamate groups in New Guinea (Slavenko *et al.* 2020; Tallowin, *et al*. 2018). The central mountain range appears to play a key role in the speciation of *Lygisaurus*, as all samples from this region show high genetic divergence between localities. The uplift of the mountains creates highly fragmented montane habitats, resulting in novel habitat colonization and population isolation (Toussaint *et al*. 2014; Oliver *et al*. 2017; Tallowin *et al*. 2018). Montane vicariance is likely also responsible for the large genetic distance between *Carlia* populations from Maimafu and Haia, where a ~2,500m peak and a difference of 400 m elevation separates the two localities despite a distance of only 22km. Furthermore, the uplift of these mountain ranges appears to have promoted speciation of lowland taxa by forming barriers to gene flow between populations (Georges *et al*. 2014; Tallowin *et al*. 2018; Tallowin *et al*. 2019).

Cryptic Morphology

Our molecular analyses support the existence of multiple cryptic species of rainbow skinks. While sample size and sexual dimorphism may bias our morphological data, our results are similar to the largely unresolved morphological analysis of Zug (2004), and support the findings of others, that the meristic and mensural characters used to define species of *Carlia* often exhibit intraspecific variation equal to or greater than the interspecific variation (Allison and Zug, 2006; Stuart-Fox *et al*., 2002). While our morphological analysis detected a few significant morphological differences between clades of *Carlia*, there is considerable overlap in mensural characters as seen in the morphospace diagram (Fig. 4) and the variation within the Lakekamu population is greater than that between geographically and genetically distant localities such as New Britain and Maimafu. Slavenko *et al*. 2021 found that within New Guinean skinks, lowland species are larger, more robust, and possess longer limbs than highland species. The lowland populations of *Carlia* such as New Britain show no statistical morphological differences from the highland populations of Maimafu, despite an elevational difference of ~1,600 meters. Differences in limb length and body proportions do not appear to follow any observable patterns across localities.

The decoupling of genetic and morphological divergence has been previously reported within *Carlia* (Afonso Silva *et al.* 2017; Austin, 2011; Bragg *et al.* 2018; Potter *et al.* 2016; Storr 1974; Stuart-Fox *et al.* 2002) and is confirmed here. Given the divergence date of ~10.4 mya for the *fusca* group, and the diversity of environments inhabited, the lack of phenotypic differentiation is surprising. Yet, this decoupling of genotypic and phenotypic divergence is being reported more and more, especially within skinks (Austin 1995; Barley *et al*. 2013, Bruna *et al*. 1996, Linkem *et al.* 2010, Kornilios *et al*. 2018; Prates *et al.* 2023b; Zug and Allison 2006). Both morphological stasis and recent non-adaptive radiation could explain this phenomenon. The *C. fusca* group may have opted for a generalist strategy with a high tolerance for environmental fluctuation and increased dispersal capabilities, leading to conserved morphology across climatically variable habitats. This phenomenon may explain George Zug's observation that the *C. fusca* group is "geographically variable, yet geographically uniform" (Zug 2004), with high character similarity between distant localities.

Despite morphological similarity, male breeding coloration can vary drastically between species (Allison and Zug, 2006; Bragg *et al.* 2018; Cogger 2014; Ingram & Covacevich 1988; Zug 2004) and may serve as a prezygotic mating barrier in rainbow skinks. Similarly, UV reflectance has been identified as a method for signal communication in many squamate clades (Fleishman *et al.* 1993; Loew, E. *et al* 1996; Loew, E. R. 1994; Martin *et al.* 2015) and may be another mating barrier within rainbow skinks (Blomberg & Stuart-Fox 2001), allowing for morphological similarity in genetically distinct lineages without introgression.

DNA barcode

While breeding colors have been successful in distinguishing species, similar color patterns have emerged in geographically distant populations (Zug 2004), and color fades quickly upon preservation in ethanol (Donnellan *et al.* 2009 2004; Kraus 2007). A densely sampled, well supported molecular phylogeny will be the best solution to this conundrum, defining the geographic boundaries of each species. Even then, morphologically similar sympatric species, such as those identified in Alotau, will still be problematic to identify. Future morphological studies informed by molecular phylogeny may find new classes of characters that more accurately reflect the evolutionary history and genetic diversity of this group, but until these characters emerge, we suggest the use of a DNA barcode region to assist in identifications of difficult populations of rainbow skinks. Our data demonstrate the ability of COI-5P to successfully differentiate currently described and putative species of rainbow skinks, as visualized through haplotype networks (Figure 5, and Supplementary Figure S3).

Conclusion

Currently used meristic and mensural characters have likely led to both the over and under splitting of rainbow skinks in New Guinea, due to the apparent decoupling of genotypic and phenotypic evolution. New classes of morphological characters that more accurately represent the evolutionary history of this group should be investigated. The molecular phylogeny presented here provides the foundation for future investigation into the evolutionary history, biogeography, and taxonomy of the rainbow skinks of New Guinea, as well as principles of cryptic speciation and vicariance.

Data Availability

All sequences generated for this study are available through GenBank with the following accession numbers: Rag-1 (PQ761252–PQ761307), Aldolase (PQ761202–PQ761251), C-mos (PQ761308–PQ761363), ND4 (PQ761364–PQ761429), B-globin (PQ761430–PQ761470), Myosin (PQ761471–PQ761509), COI (PQ760909–PQ760976), and 16S (PQ759030–PQ759094).

Supplementary Material

All supplementary material is available here: doi.org/10.6084/m9.figshare.28035470.v1

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References

Adams, M., Raadik, T.A., Burridge, C.P. & Georges, A. (2014) Global biodiversity assessment and hyper-cryptic species complexes: more than one species of elephant in the room? *Systematic Biology*, 63 (4), 51–533.
https://doi.org/10.1093/sysbio/syu017

Adler, G.H., Austin, C.C. & Dudley, R. (1995) Dispersal and speciation of skinks among archipelagos in the tropical Pacific Ocean. *Evolutionary Ecology*, 9, 529–541.
https://doi.org/10.1007/BF01237834

Afonso Silva, A.C., Bragg, J.G., Potter, S., Fernandes, C., Coelho, M.M. & Moritz, C. (2017) Tropical specialist vs. climate generalist: diversification and demographic history of sister species of *Carlia* skinks from northwestern Australia. *Molecular Ecology*, 26 (15), 4045–4058.
https://doi.org/10.1111/mec.14185

Allison, A. & Greer, A.E. (1986) Egg shells with pustulate surface structures: basis for a new genus of New Guinea skinks (Lacertilia: Scincidae). *Journal of Herpetology*, 20 (1), 116–119.
https://doi.org/10.2307/1564142

Austin, C.C. (1995) Molecular and morphological evolution in South Pacific scincid lizards: morphological conservatism and phylogenetic relationships of Papuan *Lipinia* (Scincidae). *Herpetologica*, 51 (3), 291–300.

Austin, C.C., Hayden, C.J., Bigilale, I., Dahl, C. & Anaminiato, J. (2008) Checklist and comments on the terrestrial amphibian and reptile fauna from Utai, northwestern Papua New Guinea. *Herpetological Review*, 39 (1), 40.

Austin, C.C., Rittmeyer, E.N., Oliver, L.A., Andermann, J.O., Zug, G.R., Rodda, G.H. & Jackson, N.D. (2011) The bioinvasion of Guam: inferring geographic origin, pace, pattern and process of an invasive lizard (*Carlia*) in the Pacific using multi-locus genomic data. *Biological Invasions*, 13 (9), 1951–1967.
https://doi.org/10.1007/s10530-011-0014-y

Austin, C.C. & Zug, G.R. (1999) Molecular and morphological evolution in the south-central Pacific skink *Emoia tongana* (Reptilia: Squamata): uniformity and human-mediated dispersal. *Australian Journal of Zoology*, 47 (5), 425–437.
https://doi.org/10.1071/ZO99019

Baldwin, S.L., Fitzgerald, P.G. & Webb, L.E. (2012) Tectonics of the New Guinea region. *Annual Review of Earth and Planetary Sciences*, 40 (1), 495–520.
https://doi.org/10.1146/annurev-earth-040809-152540

Barley, A.J., White, J., Diesmos, A.C. & Brown, R.M. (2013) The challenge of species delimitation at the extremes: diversification without morphological change in Philippine sun skinks. *Evolution*, 67 (12), 3556–3572.
https://doi.org/10.1111/evo.12219

Bird, P. (2003) An updated digital model of plate boundaries. *Geochemistry, Geophysics, Geosystems*, 4 (3), 1–52.
https://doi.org/10.1029/2001GC000252

Blomberg, S.P. & Stuart-Fox, D. (2001) Ultraviolet reflectance in the small skink *Carlia pectoralis*. *Herpetological Review*, 32 (1), 16.

Boulenger, G.A. (1898) An account of the reptiles and batrachians collected by Dr. L. Loria in British New Guinea. *Annali del Museo Civico di Storia Naturale di Genova*, 38 (2), 694–710.

Bouckaert, R., Vaughan, T.G., Barido-Sottani, J., Duchêne, S., Fourment, M., Gavryushkina, A., Heled, J., Jones, G., Kühnert, D., De Maio, N., Matschiner, N., Mendes, F.K., Müller, N.F., Ogilvie, H.A., du Plessis, L., Popinga, A., Rambaut, A., Rasmussen, D., Siveroni, I., Suchard, M.A., Wu, C., Xie, D., Zhang, C., Stadler, T. & Drummond, A.J. (2019) BEAST 2.5: An advanced software platform for Bayesian evolutionary analysis. *PLoS computational biology*, 15 (4), e1006650.
https://doi.org/10.1371/journal.pcbi.1006650

Bragg, J.G., Potter, S., Afonso Silva, A.C., Hoskin, C.J., Bai, B.Y. & Moritz, C. (2018) Phylogenomics of a rapid radiation: the Australian rainbow skinks. *BMC evolutionary biology*, 18 (15), 1–12.
https://doi.org/10.1186/s12862-018-1130-4

Brandley, M.C., Bragg, J.G., Singhal, S., Chapple, D.G., Jennings, C.K., Lemmon, A.R., Lemmon, E.M., Thompson, M.B. & Moritz, C. (2015) Evaluating the performance of anchored hybrid enrichment at the tips of the tree of life: a phylogenetic analysis of Australian Eugongylus group scincid lizards. *BMC evolutionary biology*, 15 (62), 1–14. https://doi.org/10.1186/s12862-015-0318-0

Bruna, E.M., Fisher, R.N. & Case, T.J. (1996) Morphological and genetic evolution appear decoupled in Pacific skinks (Squamata: Scincidae: *Emoia*). *Proceedings of the Royal Society of London. Series B: Biological Sciences*, 263 (1371), 681–688. https://doi.org/10.1098/rspb.1996.0102

Bryant, D. & Moulton, V. (2004) Neighbor-net: an agglomerative method for the construction of phylogenetic networks. *Molecular biology and evolution*, 21 (2), 255–265. https://doi.org/10.1093/molbev/msh018

Buden, D.W. (2009) *Carlia ailanpalai* (Reptilia: Scincidae): An Invasive Species of Lizard in the Federated States of Micronesia. *Pacific Science*, 63 (2), 243–251. https://doi.org/10.2984/049.063.0206

Cámara-Leret, R., Frodin, D.G., Adema, F., Anderson, C., Appelhans, M.S., Argent, G., Arias Guerrero, S., Ashton, P., Baker, W.J., Barfod, A.S., Barrington, D., Borosova, R., Bramley, G.L., Briggs, M., Buerki, S., Cahen, D., Callmander, M.W., Cheek, M., Chen, C.W., Conn, B.J., Coode, M.J., Darbyshire, I., Dawson, S., Dransfield, J., Drinkell, C., Duyfjes, B., Ebihara, A., Ezedin, Z., Fu, L.F., Gideon, O., Girmansyah, D., Govaerts, R., Fortune-Hopkins, H., Hassemer, G., Hay, A., Heatubun, C.D., Hind, D.J., Hoch, P., Homot, P., Hovenkamp, P., Hughes, M., Jebb, M., Jennings, L., Jimbo, T., Kessler, M., Kiew, R., Knapp, S., Lamei, P., Lehnert, M., Lewis, G.P., Linder, H.P., Lindsay, S., Low, Y.W., Lucas, E., Mancera, J.P., Monro, A.K., Moore, A., Middleton, D.J., Nagamasu, H., Newman, M.F., Nic Lughadha, E., Melo, P.H., Ohlsen, D.J., Pannell, C.M., Parris, B., Pearce, L., Penneys, D.S., Perrie, L.R., Petoe, P., Poulsen, A.D., Prance, G.T., Quakenbush, J.P., Raes, N., Rodda, M., Rogers, Z.S., Schuiteman, A., Schwartsburd, P., Scotland, R.W., Simmons, M.P., Simpson, D.A., Stevens, P., Sundue, M., Testo, W., Trias-Blasi, A., Turner, I., Utteridge, T., Walsingham, L., Webber, B.L., Wei, R., Weiblen, G.D., Weigend, M., Weston, P., de Wilde, W., Wilkie, P., Wilmot-Dear, C.M., Wilson, H.P., Wood, J.R., Zhang, L. & van Welzen, P.C. (2020) New Guinea has the world's richest island flora. *Nature*, 584 (7822), 579–583. https://doi.org/10.1038/s41586-020-2549-5

Cerca, J., Meyer, C., Stateczny, D., Siemon, D., Wegbrod, J., Purschke, G., Dimitrov, D. & Struck, T.H. (2020) Deceleration of morphological evolution in a cryptic species complex and its link to paleontological stasis. *Evolution*, 74 (1), 116–131. https://doi.org/10.1111/evo.13884

Chapple, D.G., Slavenko, A., Tingley, R., Farquhar, J.E., Camaiti, M., Roll, U. & Meiri, S. (2023) Built for success: Distribution, morphology, ecology and life history of the world's skinks. *Ecology and Evolution*, 13 (12), e10791. https://doi.org/10.1002/ece3.10791

Chapple, D.G., Chapple, S.N., Smith, S.A., Shea, G.M., Brennan, I.G. & Sadlier, R.A. (2023) Phylogenetic relationships in the Eugongylini (Squamata: Scincidae): generic limits and biogeography. *Australian Journal of Zoology*, 70 (6), 165–203. https://doi.org/10.1071/ZO23007

Cogger, H.G. (2014) *Reptiles and Amphibians of Australia*. CSIRO Publishing, Collingwood, 1033 pp. https://doi.org/10.1071/9780643109773

Couper, P.J., Wilmer, J.W., Roberts, L., Amey, A.P. & Zug, G.R. (2005) Skinks currently assigned to *Carlia aerata* (Scincidae: Lygosominae) of north-eastern Queensland: a preliminary study of cryptic diversity and two new species. *Australian Journal of Zoology*, 53 (1), 35–49. https://doi.org/10.1071/ZO04010

Davies, H.L. (2012) The geology of New Guinea-the cordilleran margin of the Australian continent. *Episodes Journal of International Geoscience*, 35 (1), 87–102. https://doi.org/10.18814/epiiugs/2012/v35i1/008

Davies, H.L., Perembo, R.C., Winn, R.D. & KenGemar, P. (1997) Terranes of the New Guinea orogen. *In:* Hancock, G. (Ed.), *Proceedings of the Geology Exploration and Mining Conference, Madang*. Australasian Institute of Mining and Metallurgy, Melbourne, pp. 61-66.

Davies, H.L., Winn, R.D. & KenGemar, P. (1996) Evolution of the Papuan Basin-a view from the orogen. Papua New Guinea (PNG) Petroleum Convention Proceedings. *In:* Buchanan, P. (Ed.), *Petroleum exploration, development and production in Papua New Guinea*. PNG Chamber of Mines and Petroleum, Port Moresby, pp. 53–62.

De Vis, C. (1884) New Queensland lizards. *Proceedings of the Royal Society of Queensland*, 1 (2), 77–78. https://doi.org/10.5962/p.351005

Dolman, G. & Hugall, A.F. (2008) Combined mitochondrial and nuclear data enhance resolution of a rapid radiation of Australian rainbow skinks (Scincidae: *Carlia*). *Molecular Phylogenetics and Evolution*, 49 (3), 782–794. https://doi.org/10.1016/j.ympev.2008.09.021

Dolman, G. & Stuart-Fox, D. (2010) Processes driving male breeding colour and ecomorphological diversification in rainbow skinks: a phylogenetic comparative test. *Evolutionary Ecology*, 24, 97–113. https://doi.org/10.1007/s10682-009-9293-5

Donnellan, S., Couper, P., Saint, K. & Wheaton, L. (2009) Systematics of the *Carlia fusca* complex (Reptilia: Scincidae) from northern Australia. *Zootaxa*, 2227 (1), 1–31. https://doi.org/10.11646/zootaxa.2227.1.1

Dow, D.B. (1972) *Geology of the South Sepik Region, New Guinea. Bulletin/Dept. of National Development, Bureau of Mineral Resources, Geology and Geophysics*; *133*. Australian Government Publishing Service, Canberra, 88 pp.

Duméril, A.M.C. & Bibron, G. (1841) *Erpétologie Générale ou Histoire Naturelle Compléte des Reptiles. Vol. 8.* Roret, Paris, 792 pp.

Drummond, A.J., Suchard, M.A., Xie, D. & Rambaut, A. (2012) Bayesian phylogenetics with BEAUti and the BEAST 1.7. *Molecular biology and evolution*, 29 (8), 1969–1973.
https://doi.org/10.1093/molbev/mss075

Edgar, R.C. (2004) MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic acids research*, 32 (5), 1792–1797.
https://doi.org/10.1093/nar/gkh340

Eme, D., Zagmajster, M., Delić, T., Fišer, C., Flot, J.F., Konecny-Dupré, L., Pálsson, S., Stoch, F., Zakšek, V., Douady, C.J. & Malard, F. (2018) Do cryptic species matter in macroecology? Sequencing European groundwater crustaceans yields smaller ranges but does not challenge biodiversity determinants. *Ecography*, 41 (2), 424–436.
https://doi.org/10.1111/ecog.02683

Escoriza, D. (2023) Success factors of great oceanic dispersers: Case of Squamata in the Pacific Ocean. *Journal of Zoology*, 319 (3), 221–230.
https://doi.org/10.1111/jzo.13042

Fleishman, L.J., Loew, E.R. & Leal, M. (1993) Ultraviolet vision in lizards. *Nature*, 365 (6445), 397.
https://doi.org/10.1038/365397a0

Fišer, C., Robinson, C.T. & Malard, F. (2018) Cryptic species as a window into the paradigm shift of the species concept. *Molecular Ecology*, 27 (3), 613–635.
https://doi.org/10.1111/mec.14486

Fitzinger, L. (1843) *Systema Reptilium. Fasciculus primus, Amblyglossae*. Braumüller et Seidel, Vienna, 106 pp.
https://doi.org/10.5962/bhl.title.4694

Funk, W.C., Caminer, M. & Ron, S.R. (2012) High levels of cryptic species diversity uncovered in Amazonian frogs. *Proceedings of the Royal Society B: Biological Sciences*, 279 (1734), 1806–1814.
https://doi.org/10.1098/rspb.2011.1653

Georges, A., Zhang, X., Unmack, P., Reid, B.N., Le, M. & McCord, W.P. (2014) Contemporary genetic structure of an endemic freshwater turtle reflects Miocene orogenesis of New Guinea. B*iological Journal of the Linnean Society*, 111 (1), 192–208.
https://doi.org/10.1111/bij.12176

Goldberg, S.R. & Kraus, F. (2012) Reproduction in the invasive lizard, *Carlia ailanpalai* (Squamata: Scincidae) from Oceania. *Russian Journal of Herpetology*, 19 (3), 199–202.

Gray, J.E. (1845) *Cataloge of the specimens of lizards in the collection of the British Museum.* Trustees of the British Museum, London, 289 pp.
https://doi.org/10.5962/bhl.title.5499

Greer, A.E. (1974) The genetic relationships of the scincid lizard genus *Leiolopisma* and its relatives. *Australian Journal of Zoology Supplementary Series*, 22 (31), 1–67.
https://doi.org/10.1071/AJZS031

Günther, A.C.L.G. (1858) On the systematic arrangement of the tailless batrachians and the structure of Rhinophrynus dorsalis. *Proceedings of the Zoological Society of London*, 1858, 339–352.
https://doi.org/10.1111/j.1469-7998.1858.tb06387.x

Hall, R. (1998) The plate tectonics of Cenozoic SE Asia and the distribution of land and sea. *In:* Hall, R. & Holloway, J. (Eds.), *Biogeography and Geological Evolution of SE Asia.* Backhuys, Leiden, pp. 99–131.

Hall, R. (2002) Cenozoic geological and plate tectonic evolution of SE Asia and the SW Pacific: computer-based reconstructions, model and animations. *Journal of Asian earth sciences*, 20 (4), 353–431.
https://doi.org/10.1016/S1367-9120(01)00069-4

Hall, R. & Spakman, W. (2003) Mantle structure and tectonic evolution of the region north and east of Australia. *Special Papers-Geological Society of America*, 372, 361–382.
https://doi.org/10.1130/0-8137-2372-8.361

Hill, E.C., Gao, D.F., Polhemus, D.A., Fraser, C.J., Iova, B., Allison, A. & Butler, M.A. (2023) Testing geology with biology: plate tectonics and the diversification of microhylid frogs in the Papuan region. *Integrative Organismal Biology*, 5 (1), obad028
https://doi.org/10.1093/iob/obad028

Hoskin, C.J. & Couper, P.J. (2015) A new skink (Scincidae: *Liburnascincus*) from rocky habitat on Cape York, northeast Australia. *Zootaxa*, 3994 (2), 222–234.
https://doi.org/10.11646/zootaxa.3994.2.3

Huson, D.H. & Bryant, D. (2006) Application of phylogenetic networks in evolutionary studies. *Molecular biology and evolution*, 23 (2), 254–267.
https://doi.org/10.1093/molbev/msj030

Ingram, G. & Covacevich, J. (1988) Revision of the genus Lygisaurus de Vis (Scincidae: Reptilia) in Australia. *Memoirs of the Queensland Museum*, 25 (2), 335–354.

Ingram, G. & Covacevich, J. (1989) Revision of the genus Carlia (Reptilia, Scincidae) in Australia with comments on *Carlia bicarinata* of New Guinea. *Memoirs of the Queensland Museum*, 27 (2), 443–490.

Jackson, D.A. (1993) Stopping rules in principal components analysis: a comparison of heuristical and statistical approaches. *Ecology*, 74 (8), 2204–2214.
https://doi.org/10.2307/1939574

Johnson, T. & Molnar, P. (1972) Focal mechanisms and plate tectonics of the southwest Pacific. *Journal of Geophysical Research*, 77 (26), 5000–5032.
https://doi.org/10.1029/JB077i026p05000

Knowlton, N. (1993) Sibling species in the sea. *Annual review of ecology and systematics*, 24, 189-216.
https://doi.org/10.1146/annurev.es.24.110193.001201

Kopstein, P.F. (1926) Reptilien von den Molukken und den benachbarten Inseln. *Zoologische Mededelingen*, 9 (5), 71–112.

Kornilios, P., Kumlutaş, Y., Lymberakis, P. & Ilgaz, Ç. (2018) Cryptic diversity and molecular systematics of the Aegean *Ophiomorus* skinks (Reptilia: Squamata), with the description of a new species. *Journal of Zoological Systematics and Evolutionary Research*, 56 (3), 364–381.
https://doi.org/10.1111/jzs.12205

Korshunova, T., Martynov, A., Bakken, T. & Picton, B. (2017) External diversity is restrained by internal conservatism: new nudibranch mollusc contributes to the cryptic species problem. *Zoologica Scripta*, 46 (6), 683–692.
https://doi.org/10.1111/zsc.12253

Kraus, F. (2007) Taxonomic partitioning within Papuan members of the *Carlia novaeguineae* complex (Squamata: Scincidae). *Journal of Herpetology*, 41 (3), 410–423.
https://doi.org/10.1670/0022-1511(2007)41[410:TPWPMO]2.0.CO;2

Kurita, K. & Hikida, T. (2014) Divergence and long-distance overseas dispersals of island populations of the Ryukyu five-lined skink, *Plestiodon marginatus* (Scincidae: Squamata), in the Ryukyu Archipelago, Japan, as revealed by mitochondrial DNA phylogeography. *Zoological science*, 31 (4), 187–194.
https://doi.org/10.2108/zs130179

Lavoué, S., Miya, M., Arnegard, M.E., McIntyre, P.B., Mamonekene, V. & Nishida, M. (2011) Remarkable morphological stasis in an extant vertebrate despite tens of millions of years of divergence. *Proceedings of the Royal Society B: Biological Sciences*, 278 (1708), 1003–1008.
https://doi.org/10.1098/rspb.2010.1639

Letunic, I. & Bork, P. (2021) Interactive Tree Of Life (iTOL) v5: an online tool for phylogenetic tree display and annotation. *Nucleic acids research*, 49 (W1), W293–W296.
https://doi.org/10.1093/nar/gkab301

Li, X. & Wiens, J.J. (2023) Estimating global biodiversity: the role of cryptic insect species. *Systematic Biology*, 72 (2), 391–403.
https://doi.org/10.1093/sysbio/syac069

Lindley, D. (1988) Early Cainozoic stratigraphy and structure of the Gazelle Peninsula, east New Britain: an example of extensional tectonics in the New Britain arc-trench complex. *Journal of the Geological Society of Australia*, 35 (2), 231–244.
https://doi.org/10.1080/14400958808527943

Linkem, C.W., Hesed, K.M., Diesmos, A.C. & Brown, R.M. (2010) Species boundaries and cryptic lineage diversity in a Philippine forest skink complex (Reptilia; Squamata; Scincidae: Lygosominae). *Molecular Phylogenetics and Evolution*, 56 (2), 572–585.
https://doi.org/10.1016/j.ympev.2010.03.043

Loew, E., Govardovskii, V., Röhlich, P. & Szel, A. (1996) Microspectrophotometric and immunocytochemical identification of ultraviolet photoreceptors in geckos. *Visual Neuroscience*, 13 (2), 247–256.
https://doi.org/10.1017/S0952523800007483

Loew, E.R. (1994) A third, ultraviolet-sensitive, visual pigment in the Tokay gecko (*Gekko gekko*). *Vision research*, 34 (11), 1427–1431.
https://doi.org/10.1016/0042-6989(94)90143-0

Lukić, D., Waterkeyn, A., Rabet, N., Mioduchowska, M., Geudens, B., Vanschoenwinkel, B., Brendonck L. & Pinceel, T. (2019) High genetic variation and phylogeographic relations among Palearctic fairy shrimp populations reflect persistence in multiple southern refugia during Pleistocene ice ages and postglacial colonisation. *Freshwater Biology*, 64 (11), 1896–1907.
https://doi.org/10.1111/fwb.13380

Macleay, W. (1877) The lizards of the "Chevert" Expedition. *Proceedings of the Linnean Society of New South Wales*, 2 (1), 60–69.
https://doi.org/10.5962/bhl.part.12420

Martin, M., Le Galliard, J.F., Meylan, S. & Loew, E.R. (2015) The importance of ultraviolet and near-infrared sensitivity for visual discrimination in two species of lacertid lizards. *Journal of Experimental Biology*, 218 (3), 458–465.
https://doi.org/10.1242/jeb.115923

Matzke, N.J. (2013) Probabilistic historical biogeography: New models for founder-event speciation, imperfect detection, and fossils allow improved accuracy and model-testing. *Frontiers in Biogeography*, 5, 242–248.
https://doi.org/10.1093/sysbio/syu056

Matzke, N. J. (2014) Model selection in historical biogeography reveals that founder-event speciation is a crucial process in island clades. *Systematic biology*, 63 (6), 951–970.
https://doi.org/10.1093/sysbio/syu056

Meyer, A. (1874) *Platemys novaeguineae* sp. nov. Dr. WH Peters legte vor: Eine mittheilung von Hrn. Adolf Bernhard Meyer uber die von ihm auf Neu-Guinea under den Inseln Jobi, Mysore und Mafoor im Jahre 1873 gesammelten Amphibien. *Monatsberichte der Koniglich Preussischen Akademie der Wissenschaften zu Berlin*, 39, 128–140.

Mittermeier, R.A., Mittermeier, C.G., Brooks, T.M., Pilgrim, J.D., Konstant, W.R., Da Fonseca, G.A. & Kormos, C. (2003) Wilderness and biodiversity conservation. *Proceedings of the National Academy of Sciences*, 100 (18), 10309–10313.
https://doi.org/10.1073/pnas.1732458100

Monro, A.K. & Mayo, S.J. (2022) *Cryptic species: Morphological stasis, circumscription, and hidden diversity*. Cambridge University Press, Cambridge, 350 pp.
https://doi.org/10.1017/9781009070553

Nguyen, L.T., Schmidt, H.A., Von Haeseler, A. & Minh, B.Q. (2015) IQ-TREE: a fast and effective stochastic algorithm for estimating maximum-likelihood phylogenies. *Molecular biology and evolution*, 32 (1), 268–274.
https://doi.org/10.1093/molbev/msu300

Oliver, P.M., Adams, M., Lee, M.S., Hutchinson, M.N. & Doughty, P. (2009) Cryptic diversity in vertebrates: molecular data double estimates of species diversity in a radiation of Australian lizards (*Diplodactylus*, Gekkota). *Proceedings of the Royal Society B: Biological Sciences*, 276 (1664), 2001–2007.
https://doi.org/10.1098/rspb.2008.1881

Oliver, P.M., Kraus, F., Austin, C.C., Tedeschi, L.G., O'Brien, A.R. & Maddock, S.T. (2024) Lineage diversity in a Melanesian lizard radiation (Gekkonidae: *Nactus*) further highlights exceptional diversity and endemism in eastern Papua New Guinea. *Organisms Diversity & Evolution*, 2024, 1–16.
https://doi.org/10.1007/s13127-024-00655-w

Paradis, E. & Schliep, K. (2019) "ape 5.0: an environment for modern phylogenetics and evolutionary analyses in R." *Bioinformatics*, 35 (3), 526–528.
https://doi.org/10.1093/bioinformatics/bty633

Pérez-Ponce de León, G. & Poulin, R. (2016) Taxonomic distribution of cryptic diversity among metazoans: not so homogeneous after all. *Biology letters*, 12 (8), 20160371.
https://doi.org/10.1098/rsbl.2016.0371

Peters, W. (1864) Die Eidechsenfamilie der Scincoiden, insbesonder euber die Schneider'schen. *Wiegmann'schen und neue Arten des zoologishen Museums*, 1864, 44-58.

Peters, W., Beccari, O., Albertis, L., Bruijn, A. & Doria, G. (1878) Catalogo dei rettili e dei batraci raccolti da O. Beccari, LM D'Albertis e AA Bruijn nella Sotto-Regione Austro-Malese. *Annali del Museo Civico de Storia Naturale di Genova*, Series 1, 13, 323–450.

Pfenninger, M. & Schwenk, K. (2007) Cryptic animal species are homogeneously distributed among taxa and biogeographical regions. *BMC evolutionary biology*, 7 (1), 121.
https://doi.org/10.1186/1471-2148-7-121

Pigram, C. & Davies, H.L. (1987) Terranes and the accretion history of the Papua New Guinea Orogen. *Australian Bureau of Mineral Resources, Geology and Geophysics Bulletin*, 10, 193–211.

Polhemus, D.A. & Polhemus, J.T. (1998) Assembling New Guinea: 40 million years of island arc accretion as indicated by the distributions of aquatic Heteroptera (Insecta). *In:* Hall, R. & Holloway, J. (Eds.), *Biogeography and Geological Evolution of SE Asia.* Backhuys, Leiden, pp. 327–340.

Potter, S., Bragg, J.G., Peter, B.M., Bi, K. & Moritz, C. (2016) Phylogenomics at the tips: inferring lineages and their demographic history in a tropical lizard, *Carlia amax*. *Molecular Ecology*, 25 (6), 1367–1380.
https://doi.org/10.1111/mec.13546

Prates, I., Doughty, P. & Rabosky, D.L. (2023a) Subspecies at crossroads: the evolutionary significance of genomic and phenotypic variation in a wide-ranging Australian lizard (*Ctenotus pantherinus*). *Zoological Journal of the Linnean Society*, 197 (3), 768–786.
https://doi.org/10.1093/zoolinnean/zlac076

Prates, I., Hutchinson, M.N., Singhal, S., Moritz, C. & Rabosky, D.L. (2023b) Notes from the taxonomic disaster zone: Evolutionary drivers of intractable species boundaries in an Australian lizard clade (Scincidae: *Ctenotus*). *Molecular Ecology*, 33 (20), e17074.
https://doi.org/10.1111/mec.17074

R Core Team (2013) *R: A language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna. Available from: http://www.R-project.org/ (accessed 9 May 2024)

Rambaut, A., Drummond, A.J., Xie, D., Baele, G. & Suchard, M.A. (2018) Posterior summarization in Bayesian phylogenetics using Tracer 1.7. *Systematic biology*, 67 (5), 901–904.
https://doi.org/10.1093/sysbio/syy032

Ratnasingham, S. & Hebert, P.D. (2007) BOLD: The Barcode of Life Data System (http://www. barcodinglife. org). *Molecular ecology notes*, 7 (3), 355–364.
https://doi.org/10.1111/j.1471-8286.2007.01678.x

Rausch, T., Fritz, M.H., Untergasser, A. & Benes, V. (2020) Tracy: basecalling, alignment, assembly and deconvolution of sanger chromatogram trace files. *BMC Genomics*, 21, 230.
https://doi.org/10.1186/s12864-020-6635-8

Ree, R.H. & Sanmartín, I. (2018) Conceptual and statistical problems with the DEC+ J model of founder-event speciation and its comparison with DEC via model selection. *Journal of Biogeography*, 45 (4), 741–749.
https://doi.org/10.1111/jbi.13173

Reidenbach, K.R., Neafsey, D.E., Costantini, C., Sagnon, N.F., Simard, F., Ragland, G.J., Egan, S.P., Feder, J.L., Muskavitch, M.A. & Besansky, N.J. (2012) Patterns of genomic differentiation between ecologically differentiated M and S forms of *Anopheles gambiae* in West and Central Africa. *Genome biology and evolution*, 4 (12), 1202–1212.
https://doi.org/10.1093/gbe/evs095

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

Schenk, J.J. (2016) Consequences of secondary calibrations on divergence time estimates. *PLoS ONE*, 11 (1), e0148228.
https://doi.org/10.1371/journal.pone.0148228

Skinner, A., Hutchinson, M.N. & Lee, M.S. (2013) Phylogeny and divergence times of Australian *Sphenomorphus* group skinks (Scincidae, Squamata). *Molecular Phylogenetics and Evolution*, 69 (3), 906–918.
https://doi.org/10.1016/j.ympev.2013.06.014

Slavenko, A., Allison, A., Austin, C.C., Bauer, A.M., Brown, R.M., Fisher, R.N., Ineich, I., Iova, B., Karin, B.R., Kraus, F., Mecke, S., Meiri, S., Morrison, C., Oliver, P.M., O'Shea, M., Richmond, J.Q., Shea, G.M., Tallowin, O.J. & Chapple, D.G. (2023) Skinks of Oceania, New Guinea, and Eastern Wallacea: an underexplored biodiversity hotspot. *Pacific Conservation Biology*, 29 (6), 526–543.
https://doi.org/10.1071/PC22034

Slavenko, A., Allison, A. & Meiri, S. (2021) Elevation is a stronger predictor of morphological trait divergence than competition in a radiation of tropical lizards. *Journal of Animal Ecology*, 90 (4), 917–930.
https://doi.org/10.1111/1365-2656.13420

Slavenko, A., Tamar, K., Tallowin, O.J., Allison, A., Kraus, F., Carranza, S. & Meiri, S. (2020) Cryptic diversity and non-adaptive radiation of montane New Guinea skinks (*Papuascincus*; Scincidae). *Molecular Phylogenetics and Evolution*, 146, 106749.
https://doi.org/10.1016/j.ympev.2020.106749

Slavenko, A., Tamar, K., Tallowin, O.J., Kraus, F., Allison, A., Carranza, S. & Meiri, S. (2022) Revision of the montane New Guinean skink genus *Lobulia* (Squamata: Scincidae), with the description of four new genera and nine new species. *Zoological Journal of the Linnean Society*, 195 (1), 220–278.
https://doi.org/10.1093/zoolinnean/zlab052

Stattersfield, A. (1998) Identifying threatened species in the" south" using new criteria. *Pacific Conservation Biology*, 4 (1), 33–38.
https://doi.org/10.1071/PC980033

Storr, G. (1974) The genus *Carlia* (Lacertilia, Scincidae) in Western Australia and the Northern Territory. *Records of the Western Australian Museum*, 3 (2), 151–165.

Struck, T.H., Feder, J.L., Bendiksby, M., Birkeland, S., Cerca, J., Gusarov, V.I., Kistenich, S., Larsson, K.H., Liow, L.H., Nowak, M.D., Stedje, B., Bachmann, L. & Dimitrov, D. (2018) Finding evolutionary processes hidden in cryptic species. *Trends in Ecology & Evolution*, 33 (3), 153–163.
https://doi.org/10.1016/j.tree.2017.11.007

Stuart-Fox, D.M., Hugall, A.F. & Moritz, C. (2002) A molecular phylogeny of rainbow skinks (Scincidae: *Carlia*): taxonomic and biogeographic implications. *Australian Journal of Zoology*, 50 (1), 39–51.
https://doi.org/10.1071/ZO01051

Tallowin, O.J., Tamar, K., Meiri, S., Allison, A., Kraus, F., Richards, S.J. & Oliver, P.M. (2018) Early insularity and subsequent mountain uplift were complementary drivers of diversification in a Melanesian lizard radiation (Gekkonidae: *Cyrtodactylus*). *Molecular Phylogenetics and Evolution*, 125, 29–39.
https://doi.org/10.1016/j.ympev.2018.03.020

Tamura, K., Stecher, G. & Kumar, S. (2021) MEGA11: molecular evolutionary genetics analysis version 11. *Molecular biology and evolution*, 38 (7), 3022–3027.
https://doi.org/10.1093/molbev/msab120

Toussaint, E.F., Hall, R., Monaghan, M.T., Sagata, K., Ibalim, S., Shaverdo, H.V., Vogler, A.P., Pons, J. & Balke, M. (2014) The towering orogeny of New Guinea as a trigger for arthropod megadiversity. *Nature communications*, 5 (1), 4001.
https://doi.org/10.1038/ncomms5001

Vacher, J.P., Chave, J., Ficetola, F.G., Sommeria-Klein, G., Tao, S., Thébaud, C., Blanc, M., Camacho, A., Cassimiro, J., Colston, T.J., Dewynter, M., Ernst, R., Gaucher, P., Gomes, J.O., Jairam, R., Kok, P.J., Lima, J.D., Martinez, Q., Marty, C., Noonan, B.P., Sales Nunes, P.M., Ouboter, P., Recoder, R., Rodrigues, M.T., Snyder, A., Marques-Souza, S. & Fouquet, A. (2020) Large-scale DNA-based survey of frogs in Amazonia suggests a vast underestimation of species richness and endemism. *Journal of Biogeography*, 47 (8), 1781–1791.
https://doi.org/10.1111/jbi.13847

van Ufford, A.Q. & Cloos, M. (2005) Cenozoic tectonics of new Guinea. *AAPG Bulletin*, 89 (1), 119–140.
https://doi.org/10.1306/08300403073

Voje, K.L., Starrfelt, J. & Liow, L.H. (2018) Model adequacy and microevolutionary explanations for stasis in the fossil record. *The American Naturalist*, 191 (4), 509–523.
https://doi.org/10.1086/696265

Wells, R.W. & Wellington, C.R. (1984) A synopsis of the class Reptilia in Australia. *Australian Journal of Herpetology*, 1, 73–129.

Whiting, A.S., Sites Jr, J.W., Pellegrino, K.C. & Rodrigues, M.T. (2006) Comparing alignment methods for inferring the history of the new world lizard genus *Mabuya* (Squamata: Scincidae). *Molecular Phylogenetics and Evolution*, 38 (3), 719–730.
https://doi.org/10.1016/j.ympev.2005.11.011

Zug, G.R. (2004) Systematics of the *Carlia* “*fusca*” lizards (Squamata:Scincidae) of New Guinea and nearby islands. *Bishop Museum Bulletin in Zoology*, 5, 1–84

Zug, G.R. (2010) An outlying *Carlia* population from Java and comments on species groups within the genus *Carlia* (Reptilia: Squamata: Scincidae). *Proceedings of the California Academy of Sciences*, 61 (8), 389–408

Zug, G.R. & Allison, A. (2006) New *Carlia fusca* complex lizards (Reptilia: Squamata: Scincidae) from New Guinea, Papua-Indonesia. *Zootaxa*, 1237 (1), 27–44
https://doi.org/10.11646/zootaxa.1237.1.3