Fully-sampled phylogenies of squamates reveal evolutionary patterns in threat status

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ABSTRACT

Macroevolutionary rates of diversification and anthropogenic extinction risk differ vastly throughout the Tree of Life. This results in a highly heterogeneous distribution of Evolutionary distinctiveness (ED) and threat status among species. We examine the phylogenetic distribution of ED and threat status for squamates (amphisbaenians, lizards, and snakes) using fully-sampled phylogenies containing 9574 species and expert-based estimates of threat status for ~4000 species. We ask whether threatened species are more closely related than would be expected by chance and whether high-risk species represent a disproportionate amount of total evolutionary history. We found currently-assessed threat status to be phylogenetically clustered at broad level in Squamata, suggesting it is critical to assess extinction risks for close relatives of threatened lineages. Our findings show no association between threat status and ED, suggesting that future extinctions may not result in a disproportionate loss of evolutionary history. Lizards in degraded tropical regions (e.g., Madagascar, India, Australia, and the West Indies) seem to be at particular risk. A low number of threatened high-ED species in areas like the Amazon, Borneo, and Papua New Guinea may be due to a dearth of adequate risk assessments. It seems we have not yet reached a tipping point of extinction risk affecting a majority of species; 63% of the assessed species are not threatened and 56% are Least Concern. Nonetheless, our results show that immediate efforts should focus on geckos, iguanas, and chameleons, representing 67% of high-ED threatened species and 57% of Unassessed high-ED lineages.

1. Introduction

Human activities are the primary driver of the ongoing sixth mass-extinction event (Ceballos et al., 2015). The list of extinct taxa has increased significantly in recent decades and extinction risks are spread across the Tree of Life, in groups such as plants (Vamosi and Wilson, 2008), marine invertebrates (Solan et al., 2004), terrestrial arthropods (den Boer, 1990), and many vertebrate lineages (Hoffmann et al., 2010). Among vertebrates, the extinction risks of extant squamates (lizards, snakes, and amphisbaenians) are not well understood compared to birds, mammals, and amphibians understood (Isaac et al., 2007, 2012; Fritz and Purvis, 2010; Safi and Pettorelli, 2010; Jetz et al., 2014; Meiri and Chapple, 2016). This is despite the fact that squamates are one of the most diverse and widespread lineages of terrestrial vertebrate, with >9500 extant species.

A recent global analysis of extinction risk in squamates assessed only 15% (1450) of the >9500 squamate species (Böhm et al., 2013). As of 2015, the IUCN Red List has evaluated the threat status of about 4000 squamates, which includes approximately 70% (753 of 1073) of

squamate genera. These recent assessments of extinction risk and methodological advances in tree estimation allow us to explore the phylogenetic pattern of threat status in squamates, and their degree of Evolutionary Distinctiveness (ED). The ED metric measures the proportion of phylogenetic diversity (total evolutionary history, measured as the sum of branch lengths in millions of years weighted by the number of tips sharing that branch) represented by each individual species (Isaac et al., 2007).

First, we ask if species with high extinction-risk are more closely related than would be expected by chance. For example, certain lineages might be more prone to extinction due to specific traits (Purvis et al., 2000; Cardillo et al., 2008; Fritz et al., 2009; Lee and Jetz, 2011). This may be due to extinction risks associated with ecology and life-history factors such as body size, generation time, and population growth-rate, which are likely to be conserved among closely related species. In addition, extinction risk might correlate with environmental heterogeneity, geographical range, and human populations, which are expected to affect closely related species if they are endemic to regions highly impacted by climate change or habitat modification. In contrast, extinction risk might be phylogenetically overdispersed if threats are concentrated in specific geographic areas that contain a large number of distantly related lineages (such as Madagascar and Australia), but are relatively low

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Table 1
Summary of the IUCN data and reference taxonomy of the Reptile Database used in the present study. Dibamidae and *Sphenodon* were omitted.

Infraorder	Total genera	IUCN genera	Total species	IUCN species
Anguimorpha	20	15 (75%)	221	96 (43%)
Gekkota	122	91 (75%)	1602	473 (30%)
Iguania	115	84 (73%)	1759	586 (33%)
Lacertoidea	119	72 (61%)	892	270 (30%)
Scincoidea	172	117 (68%)	1721	510 (30%)
Serpentes	509	371 (73%)	3491	1358 (39%)

elsewhere. A lack of phylogenetic clustering or overdispersion might indicate that systemic threats are affecting a wide cross-section of species across the globe (Sinervo et al., 2010; Böhm et al., 2013).

Second, we ask if species at high risk of extinction represent a disproportionate amount of the total evolutionary history of squamates; if threatened lineages have higher ED. It has been shown that ED varies across lineages. For example, median ED is 6.2 Ma for all extant birds (Jetz et al., 2014), 7.9 Ma for mammals (Isaac et al., 2007), and 12.5 Ma for amphibians (Isaac et al., 2012). However, ED for squamates is unknown. Given limited resources for conservation, and often forcing triage in management decisions, threatened lineages with higher ED may be "worth more" than others (Bottrill et al., 2008; Isaac et al., 2012).

A species on a long branch with few relatives, such as the genus *Sphenodon* (tuataras), has very high ED, and represents a large proportion of the extant evolutionary diversity. From an evolutionary perspective, this would be a massive loss if they became extinct. Conversely, in a large radiation of relatively young species, such as the South American genus *Liolaemus* (tree iguanas), the loss of any single species may not have a huge impact on the amount of ED remaining in the group. Identifying whether extinction-prone species have particularly high ED, and which threatened species have the highest distinctiveness, allows more targeted allocation of funds and effort in the difficult field of conservation triage (Bottrill et al., 2008).

At least 929 of the 1073 squamate genera have species represented by DNA-sequence data (e.g. in GenBank), totalling ~5500 of the ~9500 species (~60%). Thus, we can create robust fully-sampled phylogenies containing all species, based on large-scale phylogenetic inference and
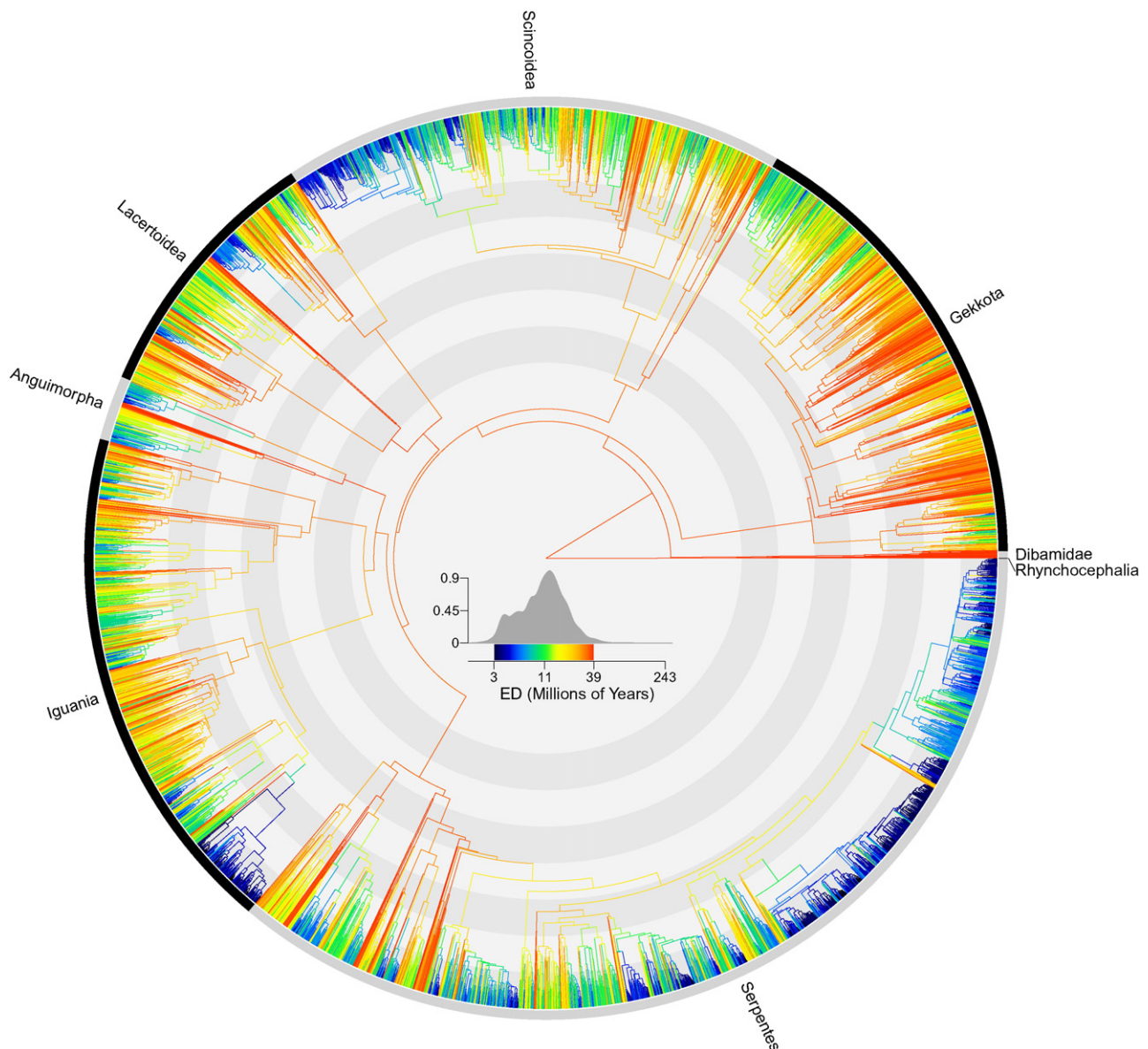

Fig. 1. Fully-sampled phylogeny of 9755 Lepidosauria species with branch color by ED values. Warmer and colder colors represent high and low ED, respectively. In the inset, the y-axis represents proportion of species and the x-axis ED in millions of years, on a log scale.

taxonomic imputation (Jetz et al., 2012). Our overall goal is to identify lineages and geographic areas of highest conservation priority. We restrict this study to Lepidosauria (Squamata [lizards, snakes, and amphisbaenians] and Rhynchocephalia [the tuatara]), to investigate a single well-known clade. Future studies might extend this research to crocodylians and turtles. We suggest future avenues for research, highlight unassessed taxa with the highest ED that merit immediate assessment, and identify the threatened taxa with the highest ED representing high priorities for current conservation efforts.

2. Material and methods

2.1. Threatened vs. non-threatened species

The most recent edition of the IUCN Red List of threatened species includes 4169 species of squamates, plus the one species of tuatara (total of 4170 species; IUCN, 2015). Based on the IUCN Red List categories and criteria, we defined extinction risk as a binary trait: threatened or non-threatened. As per IUCN definitions, species classified as Critically Endangered (CR: 133 species), Endangered (EN: 310 species), and Vulnerable (VU: 326 species) were considered 'threatened.' The 14 species classified as Extinct (EX) were also considered in our 'threatened' category because we were interested in species that have gone or are close to extinction. Species listed as Near Threatened (NT: 268 species), and Least Concern (LC: 2245 species) were considered 'non-threatened'. Data Deficient (DD: 775 species) and Unassessed (UA: 5684 species) species were not included in the analyses.

The species-level taxonomy and geographic distributions (limited to countries or other large, sub-national regions) follow the Reptile Database (Uetz and Hošek, 2015). Species-level distribution maps are not yet available for squamates. One hundred species in the IUCN dataset were removed because they did not match valid species listed in the Reptile Database (Appendix 1). Thus, the final data matrix comprises 3296 squamate species and *Sphenodon* (i.e., out of the total 4170 species, 100 were removed and 775 DD were not included), with 783 classified as threatened and 2513 species as non-threatened (Appendix 2).
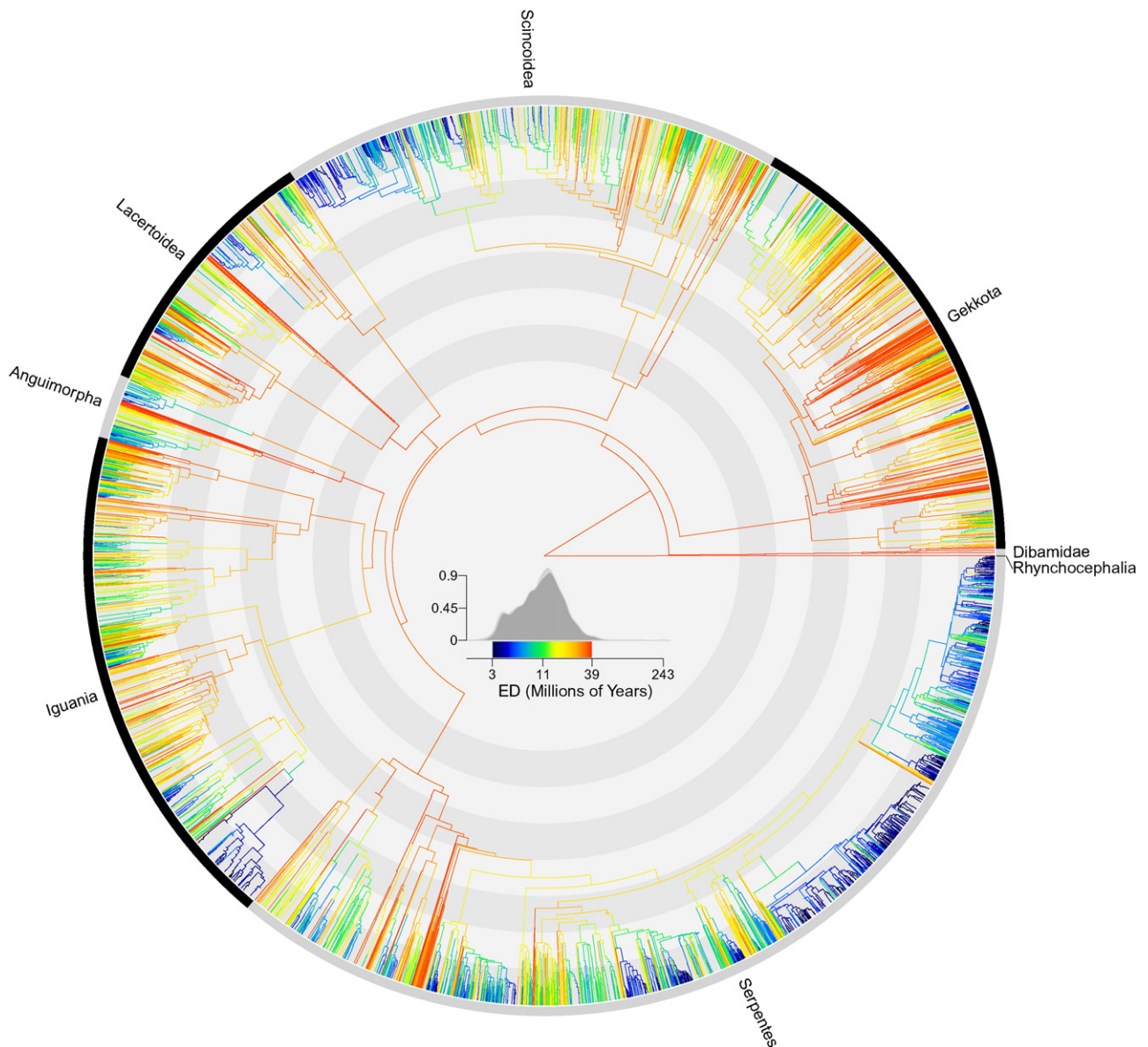

Fig. 2. Fully-sampled phylogeny trimmed to 3296 Lepidosauria species assessed by IUCN with branch color by ED values. Warmer and colder colors represent high and low ED, respectively. In the inset, the y-axis represents proportion of species and the x-axis ED in millions of years (Ma), on a log scale. In light gray is the distribution of ED across all lepidosaurs (Fig. 1), and dark gray is the distribution of ED of the assessed lineages in the tree.

Despite relative incompleteness at the species level, all major lineages and geographic areas are represented by assessments of at least a few species (IUCN, 2015), with 34% (3295) of the 9754 squamate species in the reference taxonomy from the Reptile Database, and 70% (751) of 1073 genera (see below).

We analyze squamates as a whole and the following clades separately (Table 1): Gekkota (geckos and relatives), Scincoidea (skinks and relatives), Lacertoidea (teiids, lacertids, amphisbaenians, and relatives), Anguimorpha (glass lizards, monitors, and relatives), Iguania (iguanas, chameleons, and relatives), and Serpentes (snakes). This infraorder classification follows previous authors (Jones et al., 2013; Pyron et al., 2013). We omitted dibamids from clade-specific analyses because only two have been assessed, both non-threatened.

2.2. Fully-sampled phylogeny of Squamata

We followed recent protocols, using a combination of phylogenetic inference and taxonomic assignment, to generate a posterior distribution of fully-sampled phylogenies for Squamata using the Phylogenetic Assembly with Soft Taxonomic InferenceS (PASTIS) approach (Jetz et al., 2012; Thomas et al., 2013). The data and full methods are presented in detail in the supporting information (data available in Dryad Digital Repository: http://dx.doi.org/10.5061/dryad.db005), and briefly outlined here. Using the March 2015 update of the Reptile Database, we created a baseline taxonomy consisting of 9754 currently recognized squamates, plus the tuatara. We revised an existing molecular supermatrix (Pyron et al., 2013) to include all available sequence data for 17 genes, 7 mitochondrial and 10 nuclear, for 5415 squamates plus the tuatara. Using ExaML/RAxML (Stamatakis, 2006), we estimated the Maximum-Likelihood (ML) topology for these species. This topology was enforced as a constraint for all subsequent analyses, for those species with data (Appendix Fig. 1). Topological variability is visualized as the coefficient of variation in estimated ED values across the 100 trees (Appendix Fig. 2).

We identified 175 subclades which accounted for all 9754 species. We then extracted a subset of the matrix containing 175 species
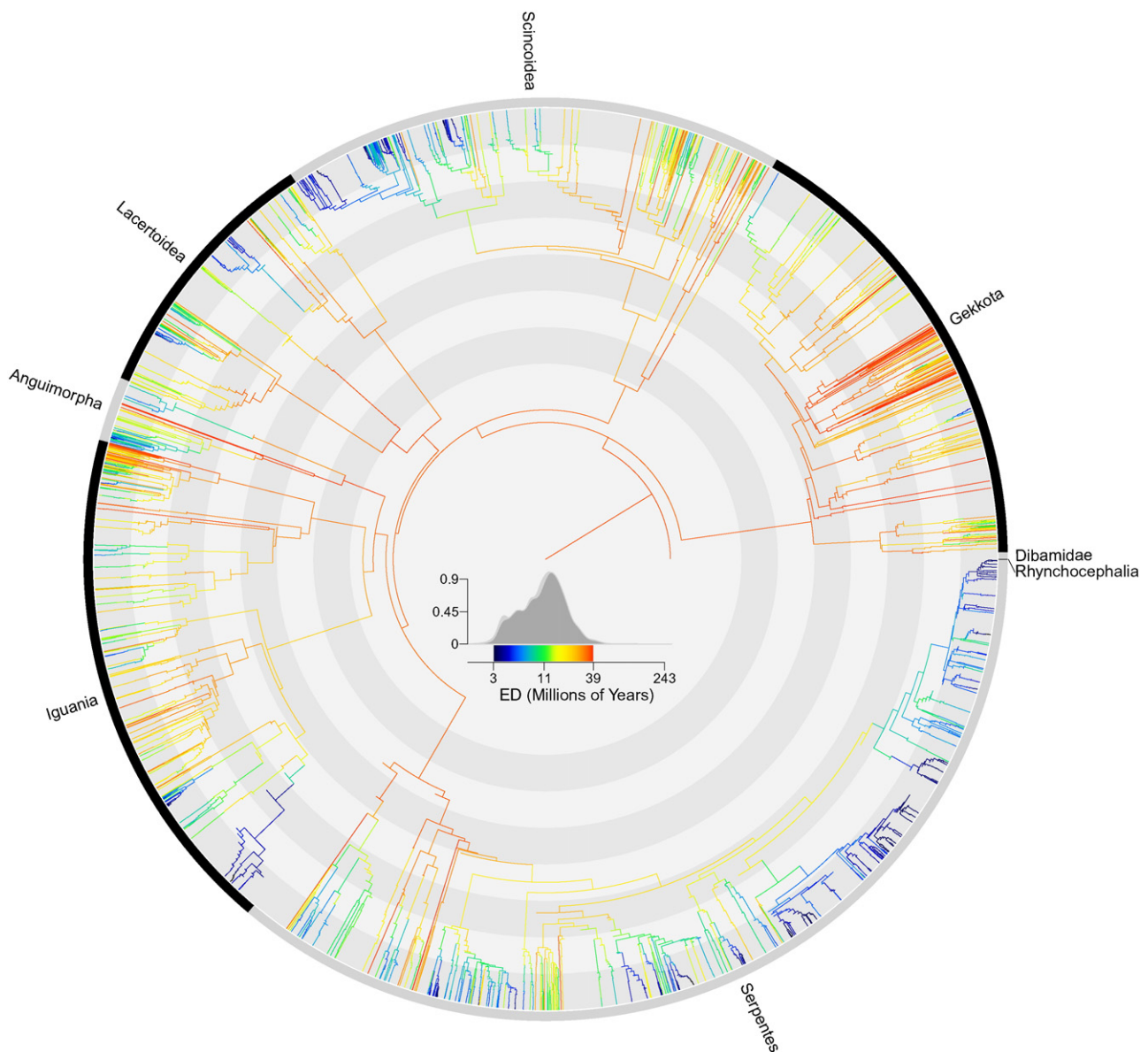

Fig. 3. Fully-sampled phylogeny trimmed to 783 squamate species classified as threatened with branch color by ED values. Warmer and colder colors represent high and low ED, respectively. In the inset, the y-axis represents proportion of species and the x-axis ED in millions of years (Ma). In light gray is the distribution of ED across all Lepidosaurs (Fig. 1), and dark gray is the distribution of ED of the assessed lineages in the tree.

representing each subclade, which we dated using MrBayes 3.2 (Ronquist et al., 2012) under a relaxed-clock model, with node-age calibrations taken from a recent integrative stratigraphic and molecular assessment of squamate divergence times (Jones et al., 2013). For each of the 175 subclades, we estimated trees scaled to relative time under the same relaxed-clock model. In these analyses, the topology of species with DNA-sequence data was fixed, and the remaining unsampled species were assigned randomly within their genus or higher-level clade. From these, one subclade tree from each subclade was grafted onto its parent lineage on the backbone tree, with the root age then re-scaled to absolute time.

Overall, this yields a distribution of 10,000 trees containing 9754 species (data available in Dryad Digital Repository: http://dx.doi.org/10.5061/dryad.db005). In each of these trees, the ML topology for 5415 species is constant, and the placement of the unsampled species is drawn from the posterior distribution of their possible locations within each genus or higher-level clade. Thus, the 10,000 trees are limited to a smaller, more probable region of tree space that accounts for the known phylogenetic relationships of sampled species, and the known taxonomic classification of unsampled species. For most analyses, we used the all-compatible consensus of these 10,000 trees, which integrate over the phylogenetic uncertainty of the missing species by collapsing poorly known clades into polytomies. For others (described below), we used a sample of 100 trees from the posterior to calculate a range of values.

This and similar approaches have shown good results for applications involving evolutionary rates and distinctiveness of threatened species, even for unsampled taxa (Isaac et al., 2012; Jetz et al., 2012, 2014). While these trees may not be suitable for estimating quantities such as rates of continuous-character evolution, simulations show them to be adequate for assessing branch-length related measures such as diversification rate, and by extension, ED (Rabosky, 2015). Thus, they should be useful for creating null models of the distribution of threat status that are conservative with respect to remaining phylogenetic uncertainty.

2.3. Phylogenetic measures of biodiversity

The measures we used to identify evolutionary patterns in threat status (see Helmus et al., 2007) are Phylogenetic Species Variability (PSV), which investigates average phylogenetic distance among all threatened species (hereafter 'average relatedness'), and Phylogenetic Species Clustering (PSC), which investigates average phylogenetic distance of threatened species to its non-threatened closest relative (hereafter 'nearest-NT-neighbor'). Values for both metrics range from 0 to 1. The PSV metric measures a hypothetical, neutral, continuously-valued trait shared by all species, which evolves randomly and independently along the phylogeny. When PSV is 1, the species phylogeny is a star (e.g. polytomy), indicating that the sampled species are maximally unrelated (overdispersed). The interaction between speciation and extinction through time drives diversification, increases relatedness, and consequently decreases PSV, indicating reduced variability and maximum relatedness (clustering) among species as PSV goes to 0. For PSC values approaching 1, species are maximally unrelated at the tips of the phylogeny (overdispersed), whereas values of 0 indicate strong clustering.

The PSV and PSC metrics are comparable to Net Relatedness Index (NRI) and Nearest Taxon Index (NTI), respectively (Helmus et al., 2007). However, NRI and NTI require an extra step to standardize their variance across communities and center their means at 0 using random selection from the species pool, while PSV and PSC measure phylogenetic signal that is not confounded with species richness. In addition, they are standardized against a hypothetical community of species that are maximally unrelated (i.e. a star phylogeny).

We tested for nonrandom phylogenetic patterns by randomly shuffling cells within rows (Kembel and Hubbell, 2006; Helmus et al., 2007;
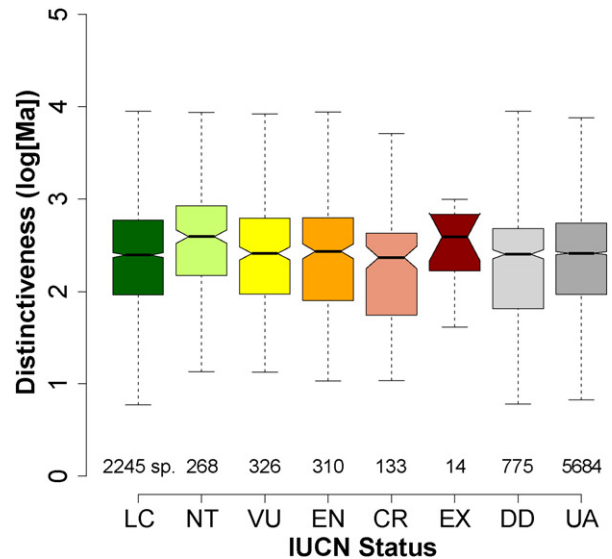

Fig. 4. Variation in ED values (log[Ma]) across 9755 Lepidosauria species. IUCN categories of threat status are Least Concern (LC), Near Threatened (NT), Vulnerable (VU), Endangered (EN), Critically Endangered (CR), Extinct (EX), Data Deficient (DD), and Unassessed (UA).

see also Webb et al., 2002). The null hypothesis assumes that if the expected phylogenetic variance of threatened species is equal to the variance of the entire squamate species pool, then the resulting phylogenetic pattern is random. In contrast, if the pool of threatened species does not represent a random sample from the total species pool, then the phylogenetic pattern of extinction risk is either 'clustered' or 'overdispersed.' If current extinction risk is more likely to affect a group of closely related species the pattern is 'clustered,' whereas if extinction risk is more likely to affect a group of more distantly related species, the pattern is 'overdispersed.'

For the null hypotheses, we generated 100 random sets of threatened species out of the total species pool and estimated PSV and PSC for each set, using the consensus tree (Null 1). In addition, we tested whether PSV of the whole species pool differs from the threatened species by sampling 100 trees from the posterior, and estimating PSV for each tree to create a distribution (Null 2). For PSV, both the random sets of threatened species with estimations using the consensus tree, and the comparison between the whole species pool and the threatened species across a sample of 100 posterior trees are valid nulls. Significance in Null 1 and randomness in Null 2 would imply that the variability among trees is greater than the signal contained in the consensus. Thus, Null 2 serves as a check on the strength of Null 1.

For PSC, the null hypothesis using the consensus tree and random sets of threatened species is ideal (e.g. Null 1), because polytomies make estimates more conservative in terms of tip clustering. Mean observed values within 2.5% and 97.5% quantiles of the null distribution were considered 'random,' values under the 2.5% quantile were

Table 2
Phylogenetic Species Variability (PSV) results. Values in the columns Null 1 and Null 2 correspond to the 5% quantile of PSV.

	Mean PSV	Null 1	Null 2	Phylostructure
Squamates	0.601	0.616–0.628	0.571–0.615	Clustered/Random
Anguimorpha	0.424	0.589–0.682	0.608–0.706	Clustered
Gekkota	0.852	0.803–0.844	0.815–0.865	Overdispersed/Random
Iguania	0.747	0.717–0.763	0.724–0.771	Random
Lacertoidea	0.707	0.762–0.791	0.763–0.793	Clustered
Scincoidea	0.553	0.498–0.577	0.502–0.588	Random
Serpentes	0.587	0.618–0.679	0.592–0.684	Clustered

Table 3
Phylogenetic Species Cluster (PSC) results. Values in the column Null 1 correspond to the 5% quantile of PSC.

	Mean PSC	Null 1	Phylostructure
Squamates	0.926	0.883–0.893	Overdispersed
Anguimorpha	0.873	0.807–0.886	Random
Gekkota	0.701	0.562–0.633	Overdispersed
Iguania	0.872	0.787–0.814	Overdispersed
Lacertoidea	0.884	0.764–0.812	Overdispersed
Scincoidea	0.886	0.813–0.834	Overdispersed
Serpentes	0.870	0.795–0.821	Overdispersed

considered 'clustered,' and values above the range of 97.5% quantile were considered 'overdispersed.' We ran analyses in R version 3.2.2 (R Core Team, 2015), using the package 'picante' (Kembel et al., 2010) and 'caper' (Orme et al., 2015).

The lack of assessment for the remaining 5684 squamate species prevents us from testing hypotheses correlating causes of threat with species-level phylogenetic patterns (as highlighted in Meiri and Chapple, 2016). For instance, if the result is clustered, we might identify which clusters of closely related species significantly drive the pattern, and test whether similar underlying threats affect distinct clusters of lineages. Therefore, we only highlight preliminary patterns. Below, we discuss potential future studies to address root causes at the species level.

2.4. ED and extinction risk

We sampled 100 trees from the posterior and calculated ED values for all 9755 species using the R package 'caper,' taking the median across each species (Figs. 1–3). For the 3296 species with a threat status (Fig. 4), we performed a simple t-test in R version 3.2.2 (R Core Team, 2015) to determine if median ED differed between threatened and non-threatened species. Given that ED is expected to co-vary strongly with phylogenetic relatedness, we then performed a phylogenetic ANOVA in the R package 'Geiger,' simulating 1000 null F-statistics to create a test distribution for comparison with the observed value.

Finally, we identified the upper 25% quartile of highest-ED Unassessed species. These are the taxa which should be the focus of

immediate assessment efforts, to plan future conservation strategies interested in maintaining maximum diversity of evolutionary history. We also identified the upper 25% quartile of highest-ED threatened lineages, which would represent the largest loss in evolutionary history if they were to go extinct, and thus should have conservation efforts increased. From these, we summarized the infraorders representing a majority of species, which indicate the broad-scale direction of priorities. We also present the top-25 species from each list to illustrate the geographic and taxonomic patterns.

3. Results

3.1. Phylogenetic patterns of threat status

There are strong phylogenetic patterns in threat for both squamates as a whole (Figs. 1-3) and the individual subclades (Tables 2 and 3). The permutation tests of PSV showed phylogenetic clustering, indicating that in terms of average relatedness, extinction-prone squamates are more closely related than expected by chance (Table 2). By analyzing each infraorder of squamates separately, threatened species of Anguimorpha, Lacertoidea, and Serpentes showed phylogenetic clustering as well (Table 2). In contrast, threats in Gekkota were phylogenetically overdispersed, whereas Iguania and Scincoidea did not differ from random (Table 2). For Null 2, Squamata and Gekkota were random; this implies greater variability among trees than the signal contained in the consensus tree (Table 2). More complete trees and assessments may help to confirm these results. We treat both groups as significant for the purposes of preliminary discussion.

For PSC, the phylogenetic pattern of threatened squamate species is overdispersion (Table 3). This shows that the nearest-NT-neighbors of extinction-prone species are more distantly related than expected at random. All major clades of squamate showed phylogenetic overdispersion, except Anguimorpha, which did not differ from random (Table 3).

3.2. ED and extinction risk

Squamates listed in IUCN threat categories do not have significant differences in median ED (Fig. 4). Furthermore, neither the standard

Table 4
Top-50 highest-ED Unassessed squamates.

Infraorder	Family	Species	ED	Range
Gekkota	Eublepharidae	*Aeluroscalabotes felinus*	92.13	Southeastern Asia
Scincoidea	Xantusiidae	*Cricosaura typica*	87.56	Cuba
Gekkota	Sphaerodactylidae	*Chatogekko amazonicus*	80.20	Amazonia
Gekkota	Gekkonidae	*Narudasia festiva*	72.41	Southwestern Africa
Serpentes	Gerrhopilidae	*Cathetorhinus melanocephalus*	71.91	Mauritius
Serpentes	Aniliidae	*Anilius scytale*	71.20	Amazonia
Anguimorpha	Lanthanotidae	*Lanthanotus borneensis*	61.04	Indonesia
Gekkota	Diplodactylidae	*Crenadactylus ocellatus*	60.58	Australia
Lacertoidea	Cadeidae	*Cadea blanoides*	57.67	Cuba
Lacertoidea	Cadeidae	*Cadea palirostrata*	57.67	Cuba
Lacertoidea	Gymnophthalmidae	*Rhachisaurus brachylepis*	56.68	Southeastern Brazil
Scincoidea	Scincidae	*Brachymeles apus*	54.73	Malaysia
Gekkota	Gekkonidae	*Kolekanos plumicaudus*	53.73	Southwestern Africa
Lacertoidea	Lacertidae	*Poromera fordii*	53.39	West-Central Africa
Serpentes	Boidae	*Calabaria reinhardtii*	53.16	West-Central Africa
Gekkota	Sphaerodactylidae	*Coleodactylus brachystoma*	50.43	Central South America
Scincoidea	Gerrhosauridae	*Broadleysaurus major*	50.24	Central and Eastern Africa
Lacertoidea	Teiidae	*Callopistes flavipunctatus*	49.81	Peru
Gekkota	Eublepharidae	*Hemitheconyx taylori*	49.73	East-Central Africa
Serpentes	Anomalepididae	*Typhlophis squamosus*	49.60	Northern South America
Dibamoidea	Dibamidae	*Dibamus tiomanensis*	48.30	West Malaysia
Scincoidea	Scincidae	*Brachymeles miriamae*	48.06	Thailand
Dibamoidea	Dibamidae	*Dibamus tebal*	47.01	Indonesia
Scincoidea	Scincidae	*Sphenomorphus praesignis*	46.81	Malay Peninsula

nor the phylogenetically corrected *t*-tests showed significant difference of ED between threatened and non-threatened taxa. Species in each group have a median ~12 Ma of ED. Geckos and iguanas represent 57% (595 and 288 species, respectively) of high-ED lineages with threat status unassessed (Appendix 3). Among top-25 highest-ED Unassessed taxa, median ED ranges from 40.63 Ma to 92.13 Ma (Table 4). Areas such as the Amazon, Borneo, New Guinea, Africa, and Southeast Asia have numerous species Unassessed by the IUCN.

Among high-ED squamates classified as threatened, 90 species are listed as Vulnerable, 84 as Endangered, 26 as Critically Endangered, and six are putatively Extinct (Appendix 3). The top-25 highest-ED threatened taxa reveal urgent needs to increase conservation priorities, again particularly in geckos and iguanas (Table 5). As might be expected, high-ED threatened taxa are also concentrated in areas of high anthropogenic pressures, such as Madagascar, Australia, the West Indies, and other tropical islands (e.g., New Caledonia). Gekkonids in particular represent a very large number of high-ED threatened species in these regions.

4. Discussion

Overall, our results suggest that current extinction risk is typically clustered at a broad level across the Squamate Tree of Life, concentrated particularly in geckos and iguanas. This result differs from birds, in which imperiled species are more likely to be overdispersed (Jetz et al., 2014). Within infraorders of squamates, extinction risk is either clustered or overdispersed (see also Böhm et al., 2013), which apparently results from the co-occurrence of multiple lineages in regions of high anthropogenic threat. Median ED for squamates is similar to amphibians, around 12 Ma (Isaac et al., 2012). As in birds and mammals, there is little variation in ED between IUCN Red List categories (Fig. 4; Arregoitia et al., 2013; Jetz et al., 2014).

Unsurprisingly, Madagascar tops the list of the top-25 highest ED threatened squamates (56% of species in Table 5) with a hugely diverse, high-ED, highly endemic fauna that is heavily endangered (Andreone et al., 2008; Hannah et al., 2008; Jetz et al., 2014). However, high-ED species also occur in areas such as the Amazon, Borneo, New Guinea, Africa, and southeast Asia, as well as lineages that are less affected by climate change (e.g., snakes), but these high-ED species have not been assessed (Table 4). Assessments of the remaining high-ED species (e.g., Table 4) will be crucial for complete conservation-management strategies worldwide, though many will likely be Data Deficient, with geographical range and population trends difficult to ascertain. Predicting threat status from trait data may assist in this effort (see below).

The low number of high-ED threatened species in areas like the Amazon, Borneo, and Papua New Guinea may thus represent a dearth of adequate risk assessments, rather than a true lack of human impact or extinction risk (Curran et al., 2004; Malhi et al., 2007; Shearman and Bryan, 2010; Natusch and Lyons, 2012). These regions are known to have many diverse and evolutionarily distinct lineages of squamates (da Silva Jr and Sites, 1995; Das, 2006; Allison, 2007), and in fact several of those are represented in Table 4. For instance, the Bornean earless monitor Lanthanotus is a monotypic lineage with high ED (61 Ma), and known from very few specimens. This lizard is likely to be highly threatened, but it has not been assessed by the IUCN. We suggest that threat assessments of squamates from regions of high biodiversity, such as Amazonia and the mainland and archipelagos of Southeast Asia, are a crucial next step toward effective conservation planning.

Species with both high and low levels of ED alike exhibit high extinction-risk, though a subset of high-ED, high-threat species present particularly urgent priorities for conservation effort (Table 5). Overall, lizards (e.g. geckos and iguanas) in degraded tropical regions (e.g. Madagascar, India, Australia, and the West Indies) seem to be at particular risk. This is likely due to temperature increases, caused by climate change or habitat modification, that affect heliotropic thermoconformers (see Huey et al., 2009; Sinervo et al., 2010). However, diurnal lizards are not the only species at risk. While we have not done an explicit analysis of fine-scale geographic or trait-based correlates of extinction risk, the threatened species with the highest ED span a qualitatively wide range of geographic regions, body sizes, and life histories other than basking lizards (see Meiri et al., 2012, 2013; Feldman et al., 2016).

For example, the monotypic genus *Shinisaurus* (Shinisauridae) is a large, semi-aquatic lizard from cool tropical forests in southeast Asia, and represents 103 Ma of ED, which is 29 Ma more than the next highest lineage (Tables 4, 5). In contrast, the monotypic genus *Xenotyphlops* (Xenotyphlopidae) is a small, fossorial blind snake from the dry regions

Table 5
Top-50 highest-ED threatened squamates.

Infraorder	Family	Species	Category	ED	Range
Anguimorpha	Shinisauridae	*Shinisaurus crocodilurus*	EN	103.00	Southeastern Asia
Gekkota	Sphaerodactylidae	*Saurodactylus fasciatus*	VU	74.00	Northern Africa
Serpentes	Xenotyphlopidae	*Xenotyphlops grandidieri*	CR	67.70	Madagascar
Serpentes	Bolyeriidae	*Bolyeria multocarinata*	EX	51.40	Mauritius
Serpentes	Bolyeriidae	*Casarea dussumieri*	EN	51.40	Mauritius
Gekkota	Gekkonidae	*Uroplatus malama*	VU	50.50	Madagascar
Gekkota	Gekkonidae	*Paragehyra gabriellae*	EN	45.80	Madagascar
Gekkota	Gekkonidae	*Paragehyra petiti*	VU	45.80	Madagascar
Gekkota	Gekkonidae	*Matoatoa brevipes*	VU	45.00	Madagascar
Iguania	Chamaeleonidae	*Palleon nasus*	VU	43.80	Madagascar
Gekkota	Eublepharidae	*Goniurosaurus kuroiwae*	EN	43.80	Japan
Gekkota	Gekkonidae	*Uroplatus guentheri*	EN	43.60	Madagascar
Gekkota	Gekkonidae	*Uroplatus malahelo*	EN	43.60	Madagascar
Gekkota	Gekkonidae	*Uroplatus ebenaui*	VU	42.90	Madagascar
Anguimorpha	Xenosauridae	*Xenosaurus newmanorum*	EN	42.10	Southern Mexico
Gekkota	Sphaerodactylidae	*Gonatodes daudini*	CR	40.80	Lesser Antilles
Gekkota	Gekkonidae	*Paroedura masobe*	EN	40.30	Madagascar
Gekkota	Gekkonidae	*Ebenavia maintimainty*	EN	39.80	Madagascar
Scincoidea	Cordylidae	*Smaug giganteus*	VU	39.70	Southern Africa
Gekkota	Gekkonidae	*Uroplatus pietschmanni*	EN	39.10	Madagascar
Gekkota	Gekkonidae	*Perochirus ateles*	EN	37.50	Micronesia
Iguania	Chamaeleonidae	*Archaius tigris*	EN	36.20	Seychelles
Gekkota	Gekkonidae	*Paroedura vazimba*	VU	36.10	Madagascar
Gekkota	Phyllodactylidae	*Asaccus montanus*	VU	35.70	Middle East
Iguania	Chamaeleonidae	*Furcifer balteatus*	EN	35.00	Madagascar

of northern Madagascar, but represents a proportionally large 68 Ma of ED. Despite their differences in body size, ecological traits, and life history, these species are both high ED, and highly threatened.

The situation is also not entirely bleak. To summarize, 63% of the assessed species are not threatened, and 56% are Least Concern. If these percentages hold across the Unassessed species, then a total of 6145 squamates are not threatened, and 5462 are Least Concern. The tuatara (*Sphenodon*) has a median ED value of 243 Ma, and is considered Least Concern. Additionally, the highest-ED squamate species is the amphisbaenian *Rhineura floridana*, the Florida Worm Lizard (Lacertoidea), with a median ED value of 130 Ma. This taxon is considered Least Concern, and is well protected in peninsular Florida. This is not to say that threats do not exist and that urgent conservation-management planning is not needed, but it seems we have not yet reached a tipping point of extinction risk affecting a majority of species or lineages.

The phylogenetic clustering of threat status suggests that it is crucial to assess extinction risks for close relatives of threatened lineages. Therefore, future research should be concentrated in three major areas. First would be in-depth assessments of the almost 6000 unsampled species, for robust evaluation of threat levels. Second would be to gather relevant spatial, geographic, ecological, and life-history data that could be used to evaluate predictors of extinction risk and identify clades and regions of exceptionally high conservation priority. Related to this, a third aim is to predict threat status using those data and models for species with little currently available data on population trends (Dickinson et al., 2014; Bland et al., 2015; Jetz and Freckleton, 2015; Schachat et al., 2015; Bland and Böhm, 2016). Preliminarily, our results suggest that conserving primary habitats containing diverse assemblages of species is likely to be the best overall strategy for preserving squamate diversity, difficult as this may be.

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Data accessibility

All PASTIS trees and related data are available in Dryad repository http://dx.doi.org/10.5061/dryad.db005.

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