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Detecting evolutionarily significant units above the species level using the Generalized Mixed Yule Coalescent method

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Summary

1. There is renewed interest in inferring evolutionary history by modelling diversification rates using phylogenies. Understanding the performance of the methods used under different scenarios is essential for assessing empirical results. Recently we introduced a new approach for analysing broadscale diversity patterns, using the Generalized Mixed Yule Coalescent (GMYC) method to test for the existence of evolutionarily significant units above the species (higher ESUs). This approach focuses on identifying clades as well as estimating rates and we refer to it as clade-dependent. However, the ability of the GMYC to detect the phylogenetic signature of higher ESUs has not been fully explored, nor has it been placed in the context of other, clade-independent approaches.

2. We simulated >32,000 trees under two clade-independent models: constant-rate birth-death (CRBD) and variable-rate birth-death (VRBD), using parameter estimates from nine empirical trees and more general parameter values. The simulated trees were used to evaluate scenarios under which GMYC might incorrectly detect the presence of higher ESUs.

3. The GMYC null model was rejected at a high rate on CRBD-simulated trees. This would lead to spurious inference of higher ESUs. However, the support for the GMYC model was significantly greater in most of the empirical clades than expected under a CRBD process. Simulations with empirically derived parameter values could therefore be used to exclude CRBD as an explanation for diversification patterns. In contrast, a VRBD process could not be ruled out as an alternative explanation for the apparent signature of hESUs in the empirical clades, based on the GMYC method alone. Other metrics of tree shape, however, differed notably between the empirical and VRBD-simulated trees. These metrics could be used in future to distinguish clade-dependent and clade-independent models.

4. In conclusion, detection of higher ESUs using the GMYC is robust against some clade-independent models, as long as simulations are used to evaluate these alternatives, but not against others. The differences between clade-dependent and clade-independent processes are biologically interesting, but most current models focus on the latter. We advocate more research into clade-dependent models for broad diversity patterns.

Keywords: birth-death, clade-dependent, clade-independent, diversification, phylogenetic clustering, rate shift, relative extinction rate, simulation

Introduction

There is currently widespread interest in understanding the evolutionary history of clades by inferring diversification dynamics from phylogenetic trees. Of particular interest has been identifying shifts in net diversification rates (Rabosky 2006; Alfaro *et al.* 2009) or rapidly diversifying clades (e.g. Hughes & Eastwood 2006; Valente, Savolainen & Vargas 2010) that might be associated with a particular trait or region (Maddison, Midford & Otto 2007; Goldberg, Lancaster & Ree 2011). Recent advances have focussed on making the widely used birth-death model (Nee, May & Harvey 1994) more flexible, allowing rates to vary more generally over time (Morlon, Parsons & Plotkin 2011), as a function of standing diversity (Etienne et al. 2012) or among lineages, to identify clades undergoing adaptive radiation (Etienne & Haegeman 2012) or with shared evolutionary dynamics (Rabosky 2014).

What processes could cause sharing and decoupling of rates among lineages? Recently, we proposed a model of evolutionarily significant units above the level of species (Barraclough 2010; Humphreys & Barraclough 2014). This model assumes that i) species within a wider clade occupy a range of geographical regions and/or ecological zones; ii) there are separate limits on the number of species within each geographical region or ecological zone; iii) species turnover occurs through ongoing speciation and extinction and iv) transitions between geographical regions or ecological zones are rare, meaning that closely related species tend to occupy the same region and/or zone. If these conditions are met, then species will fall into a set of clades, each of which occupies a separate geographical region or ecological zone, which we call higher evolutionarily significant units (hESUs; Fig. 1). Because of ongoing species turnover, species within a hESU share evolutionary fate as well as history (Barraclough & Humphreys 2015). This means that any event influencing the likelihood of lineages speciating or going extinct will be shared among species within but not among hESUs; hence, diversification rates are shared within and decoupled among hESUs.

The hESU model thus provides an explanation for diversity patterns that focuses on identifying units (clades) as well as estimating diversification rates. We therefore refer to it as a clade-dependent model (Fig. 1). The phylogenetic signature of hESUs is a significant increase in the rate of lineage accumulation toward the present. However, such a pattern may equally result from a clade-wide increase in diversification rates caused, for example, by a rebound from a mass extinction event (Crisp & Cook 2009) or a burst following environmental change (Stadler 2011). In other words, the pattern of an increase in branching rate, predicted to arise with hESUs, could also result from a uniform change in diversification rate, acting across an entire clade or independently of clade membership (clade-independent model, Fig. 1). Indeed, distinguishing alternative models for diversification is challenging because several processes can lead to indistinguishable patterns (Barraclough & Nee 2001; Rabosky 2009; Morlon, Potts & Plotkin 2010; Moen & Morlon 2014). Understanding the performance of the models used to study these patterns is therefore necessary if we are to have confidence in empirical inferences.

Here we use simulations to explore error rates of the generalised mixed Yule coalescent (GMYC; Pons *et al.* 2006) method, used to define hESUs, when trees actually derive from clade-independent processes. The GMYC method analyses waiting times between branching events in a time-calibrated phylogeny, where tips represent species, densely sampled (Humphreys & Barraclough 2014) for a broader clade, to identify significant shift(s) in the rate of branching. The approach uses a null model that no shift has occurred and that a single process is sufficient to describe phylogenetic branching across the entire clade. The alternative model finds one (single threshold version, ST) or more (multiple threshold version, MT) shifts in branching rate toward the present (Pons *et al.* 2006; Fontaneto *et al.* 2007; Monaghan *et al.* 2009; Fujisawa & Barraclough 2013), denoting the transition from among to within hESU branching. In its current formulation, the alternative model thus uses two branching parameters, λ, to explain the distribution of waiting times, one within and one among hESUs. In addition, the GMYC algorithm includes one (null) or two (ST, MT) scaling parameters, p, that allow the net branching rate to depart from a constant-rate process ($p = 1$), to either accelerate ($p > 1$) or decelerate ($p < 1$) toward the present.

Several studies have assessed the factors that influence the performance of the GMYC method applied at the species level (e.g. Papadopoulou *et al.* 2008; Reid & Carstens 2012; Fujisawa & Barraclough 2013; Tang *et al.* 2014), the main factor being the level of variation within species relative to divergence times among species (Fujisawa & Barraclough 2013). For analyses of higher clades, we previously recorded high error rates for trees simulated under a particular clade-independent model (with constant extinction rates), but found that empirical signatures of hESUs in were significantly stronger than expected under that model (Humphreys & Barraclough 2014). We build on these results here to assess error rates for a broader range of clade sizes, extinction rates and diversification processes. We examine performance of the GMYC approach on empirical trees and trees simulated under two different clade-independent processes that might generate similar phylogenetic patterns to the hESU model: constant-rate birth-death (CRBD) and variable-rate birth-death (VRBD) with a tree-wide shift in diversification rate. The CRBD model, commonly used for macroevolutionary analyses, generates an upturn in apparent branching rate towards the present when extinction rates are high (Nee *et al.* 1994). This might artefactually lead to the detection of hESUs using the standard GMYC method. Simulated trees were therefore used to estimate the rate of incorrect detection of hESUs for data generated using a CRBD model. More challenging still, the VRBD model generates a simultaneous increase in branching rate across an entire clade. This pattern should be indistinguishable from the predictions of hESUs as detected using the ST version of GMYC. There is no reason, however, to expect simultaneous transition times for all hESUs in a clade-dependent model and therefore the MT version of the GMYC might still reveal stronger evidence for hESUs than a VRBD model.

To focus our investigation on real datasets, we ran the simulations using parameter values estimated for nine empirical clades. We then compared the significance of hESUs in each clade relative to the standard GMYC null model, to trees simulated assuming a CRBD model, and to trees simulated assuming a VRBD model. Empirical trees yielded higher likelihoods under the alternative GMYC model than did trees simulated under the CRBD model, indicating that inferences of hESUs are robust to the effects of constant extinction rates on tree shapes. The likelihoods of empirical trees under the GMYC model were not, however, higher than expected under the VRBD model, even using the MT version. Additional measures of tree shape or of ecological trait variation are necessary to distinguish a signal of clade-dependent hESUs from a VRBD model.

Materials and methods

PHYLOGENETIC TREES FOR EMPIRICAL DATA

Empirical analyses were performed for nine clades, defined as representing at least one order and a manageable number of species (≤1000 species), for which densely sampled, time calibrated phylogenies were available or could be generated using published data. Such phylogenies were available for three clades of mammals (Carnivora, Euungulata and Lagomorpha; Humphreys & Barraclough 2014), birds (Afroaves (sensu Jarvis *et al.* 2014), nightbirds (except owls), swifts and hummingbirds (sensu Ericson *et al.* 2006, hereafter 'nightbirds') and core waterbirds plus pigeons and cuckoos (hereafter 'waterbirds'; Jetz *et al.* 2012; SI Text)) and conifers (Leslie et al. 2012). Phylogenies for cycads and Gnetales were generated using standard protocols from published matK, rbcL, 18S and, for cycads, PHYP sequences (Rydin & Korall 2009; Nagalingum *et al.* 2011; Hou *et al.* 2015; SI Text). Overall, all orders, families and genera in these clades were sampled and on average 80-90% of the species (Table S2).

GENERALISED MIXED YULE-COALESCENT ANALYSES FOR EMPIRICAL TREES

Null, ST and MT GMYC models were fitted to each bird and gymnosperm maximum clade credibility (MCC) tree using the R (R Development Core Team 2011) package splits (Ezard, Fujisawa & Barraclough 2014). The null model has two parameters (λ, p) and the alternative models four (λ_{among}, p_{among}, λ_{within}, p_{within}). The inferred threshold time does not constitute a model parameter but a constraint to model search space (Fujisawa & Barraclough 2013). Model inferences were summarised across the 95% confidence set of models. Mammal results were obtained from Humphreys & Barraclough (2014).

SIMULATING BIRTH-DEATH TREES WITH EMPIRICAL PARAMETER VALUES

To study the behaviour of GMYC models under alternative scenarios, CRBD trees (Nee, May & Harvey 1994) with the properties of each of the nine empirical clades (number of tips, speciation (λ) and extinction (μ) rates) were simulated. Parameter values were estimated using the birthdeath function in the R package ape (Paradis, Claude & Strimmer 2004), applied to 500 trees for all datasets except conifers, where a single tree was used. Five hundred CRBD trees were simulated using λ and relative extinction ($\mu/\lambda = \varepsilon$) rates sampled randomly from across the range of estimates for each clade using sim.bd.taxa in TreeSim (Stadler 2012) and drop.extinct in Geiger (Harmon *et al.* 2008). The estimate of ε for Gnetales was always 1.00 (Table S3) so trees were simulated with this parameter sampled between 0.98-0.99 to speed up the simulations. For the conifer CRBD trees, parameters were sampled from the same range as for cycads because the point estimate for conifers was identical to the median estimate for cycads. This resulted in 9 x 500 CRBD trees of varying size, λ and ε (Table S3). Finally, null, ST and MT GMYC models were fitted to each simulated tree and the difference in fit between null and alternative models determined using likelihood ratio (LR) tests. Error rates for CRBD trees were recorded as the proportion of trees in each set for which the null model was rejected. Note these are not type I error rates, but errors due to the null model being rejected in favour of the GMYC model when in fact a third model is true (i.e. CRBD).

SIMULATING BIRTH-DEATH TREES WITH GENERAL PARAMETER VALUES

To test GMYC performance above the species more generally, trees were simulated under a pure birth model (ε=0; Yule 1925) and CRBD models with ε = 0.1, 0.3, 0.5, 0.7 and 0.9. Pure birth trees were simulated using the tree.bd function in diversitree (FitzJohn 2012), “low” ε trees (ε = 0.1, 0.3) using sim.bdtree in Geiger and “high” ε trees (ε = 0.5, 0.7, 0.9) using sim.bd.taxa in TreeSim and drop.extinct in Geiger. The speciation rate was set to 1.0 for all simulations. For each rate of ε 500 trees were simulated, each with 100, 500 and 1000 tips. From these, sets of trees with 100%, 75% and 50% sampling were generated. Removing tips changes the shape of the lineages-through-time (LTT) plot, generating trees with an excess of early branching events ("slowdown") or, under high rates of ε, a less severe "pull of the present" effect. The sets of pruned trees therefore allowed assessing GMYC performance under departures from CRBD. In all, this resulted in 9 x 500 trees for each level of ε, i.e., a total of 54 x 500 CRBD trees of varying size, ε and branching process. Null, ST and MT GMYC models were fitted to each tree in turn and error rates recorded as above.

SIMULATING BIRTH-DEATH TREES WITH A CLADE-WIDE SHIFT IN RATES

To generate the phylogenetic signature of a clade-wide shift in rates and test the performance of the GMYC method under this scenario, VRBD trees were simulated using the empirically estimated shift position (*T*, threshold time in absolute time) and ratio of within:among hESU branching rate (λ_{within}:λ_{among}, interpreted as pre-shift and post-shift diversification rates, respectively, starting at the present and going back in time), scaled to match rates expected under a CRBD process (SI Text). Trees were simulated using sim.rateshift.taxa in TreeSim and nine combinations of parameters and tree characteristics obtained from the empirical clades (Table S4). Parameters were: λ_{among}, λ_{within} and ε, randomly sampled from the estimates across 500 trees. Tree characteristics (constraints to reconstructed tree space; Stadler 2011) were: clade size (number of extant tips) and location of the rate shift, expressed in absolute time and sampled randomly from the range of mean threshold times retained among the confidence set of GMYC models for each dataset. This resulted in 9 x 100 VRBD trees of varying size, overall λ, ε and position and severity of the rate shift. Null, ST and MT GMYC models were fitted to each tree in turn as above and the proportion of trees for which 1) the null was rejected in favour of the ST-GMYC, 2) the correct empirical position of the rate shift was recovered and 3) fit of the MT-GMYC was significantly better than ST-GMYC ($\Delta AIC \geq 5$; a somewhat conservative cutoff (Burnham & Anderson 2002), due to known sensitivity of MT-GMYC (Fujisawa & Barraclough 2013)) was recorded.

TREE CHARACTERISTICS

Clade size, root age, ε, clade imbalance and tree stemminess were recorded for each empirical and simulated tree to assess to what extent the simulated process captured other features of the empirical trees and what affects performance of the GMYC method. Tree imbalance was estimated using Colless' (Ic; Colless 1982; Heard 1992) and Sackin's (Is; Shao & Sokal 1990) indices because they have been found to perform well compared to other measures (Agapow & Purvis 2002). The former uses the difference in the number of nodes arising from the sister clades of each node and the latter the number of nodes that separates each tip from the root. In general, more imbalanced trees have a higher value under both indexes. To enable comparison among datasets they were normalized using a Yule model (Blum, François & Janson 2006). Tree stemminess was estimated using the non-cumulative stemminess index (St_N; Rohlf *et al.* 1990). Stemmier trees, i.e. those that have longer unbranched edges, have a higher value. However, values also tend to increase with increasing clade size so St_N was only compared among trees within each size set. Each index was calculated using apTreeshape (Ic and Is; Bortolussi *et al.* 2012) and customized R scripts (St_N; SI Text). Tree characteristics were correlated against error rates using linear regressions and Generalised Additive Models (Hastie & Tibshirani 1990) using the R package mgcv (Wood 2000; Wood 2011).

Results

GENERALISED MIXED YULE-COALESCENT RESULTS FOR EMPIRICAL CLADES

The GMYC null model was rejected in favour of the ST model for all clades except waterbirds and in favour of the MT model for all clades (Table 1). Only MT models were retained in the 95% confidence set of models for euungulates, conifers, Afroaves and nightbirds, both ST and MT models were retained for carnivores, lagomorphs, cycads and Gnetales and for waterbirds the null was included as well. Based on the confidence set of models, hESUs date to the Miocene (mean threshold: 5.65 Ma [Gnetales] – 16.4 Ma [conifers]; Table 1, Fig. S1) and correspond to traditionally named genera (gymnosperms, mammals), families (mammals) or clades of subfamilial, generic or subgeneric rank (birds; Fig. S2).

CONSTANT-RATE BIRTH-DEATH SIMULATED TREES

As suspected, the standard GMYC method often erroneously detected hESUs from CRBD trees. Error rates for CRBD trees based on empirical parameter estimates ranged from 6.6%–56.8% for ST and 21.1%–88.9% for MT GMYC, being lowest in trees simulated using carnivore parameter values and highest in those based on conifers (Table 2). There is no effect of clade size on error rates (linear regression ST: F = 0.28 on 7 d.f., P = 0.61; MT: F = 0.02 on 7 d.f., P = 0.89) but error rates increase with increasing ε (linear regression ST: F = 24.4 on 7 d.f., P = 0.0017, R^2 = 0.78; MT: F = 29.1 on 7 d.f., P = 0.0010, R^2 = 0.81). There is no interaction between clade size and ε and the relationship is stronger for MT than ST GMYC (slope = 49 and 26, respectively; Fig. 2).

Results for CRBD trees simulated with general parameters confirm these results (Fig. 2). Error rates increased non-linearly for all datasets except MT-GMYC with 100% sampling. For the other sets of trees, error rates remained around 10% (ST) and 30% (MT) until ε = 0.3, when they increased, non-linearly. The best regression model is a Gaussian process, which is indistinguishable from a quadratic polynomial model, based on AIC values (Table S5). Overall, error rates were higher for MT-GMYC and for higher levels of sampling. This is not because clades with more complete sampling are larger but because they have LTT plots with a more pronounced upturn (Fig. S3; the effect of sampling was marginally significant for the MT results when all clades were analysed together, P = 0.047).

Despite these effects, the empirical results are generally not explained by incorrect rejection of the null model due to constant-rate birth-death processes: the LR difference in fit between null and alternative GMYC models is much greater for empirical trees than CRBD-simulated trees for both ST and MT models and for all clades except Gnetales and waterbirds (Table 2). Thus, the evidence for hESUs is robust with respect to an alternative CRBD model.

VARIABLE-RATE BIRTH-DEATH SIMULATED TREES

The ST-GMYC detected a shift in ≥ 94% of trees for all datasets except those based on carnivore (47%) and lagomorph (75%) parameter values (Table 3). The simulated position of the shift was correctly inferred on average (estimated threshold time overlaps with range of threshold times under which trees were simulated) for all clades except those based on Afroaves values (Fig. S4). Fit of MT-GMYC was indistinguishable from ST-GMYC for the cycad-based trees and possibly those simulated using Gnetales parameter values (94% and 90% of simulated trees, respectively) but significantly better for all other datasets (in 14%–67% of simulated trees; Table S6).

The likelihood of the GMYC model for the empirical trees, however, was not greater than expected from VRBD-simulated trees, for either ST or MT versions. Indeed, for both versions, the empirical LR between null and alternative GMYC models was lower for the empirical trees than for the simulated VRBD trees (Table 4).

CHARACTERISTICS OF SIMULATED VERSUS EMPIRICAL TREES

The root height of the CRBD trees encompassed the root height of the empirical trees for all datasets except those based on euungulate and lagomorph parameter estimates, where simulated trees were too young (Fig. S1). The shape of the CRBD trees differed from the empirical trees by having too few deep lineages (carnivores, euungulates, conifers and all three bird clades), a less severe upturn in branching rate (lagomorphs, cycads and Gnetales) and by being more balanced and/or stemmy (Table S7). Exceptions are simulated trees based on carnivore, lagomorph and cycad parameter values, which were indistinguishable from empirical trees for both balance and stemminess.

The root height for VRBD trees was extremely old for all simulated trees, except those based on carnivore (overlapped empirical trees) and Afroaves (younger than empirical trees) parameter estimates (Fig. S4). The shape of the VRBD trees approximated that of the empirical tree for carnivore-based trees but differed in various ways for the other datasets. For example, VRBD conifer-based trees had too few surviving old lineages compared to the empirical tree, a completely different shape for all three simulated sets based on bird parameter values and were generally more balanced and/or stemmy than empirical trees (Table S7). Exceptions are trees based on carnivore and lagomorph parameter values, which were indistinguishable from empirical trees for both balance and stemminess.

Discussion

Our results show that the standard GMYC method is sensitive to high rates of extinction (above approximately 30% of the speciation rate) in CRBD models. Although the scaling parameters, *p*, were developed to allow for departures from a pure birth model, a constant extinction rate produces a recent upturn in branching rates rather than a gradual increase through the whole tree (Nee *et al.* 1994). This problem becomes more severe with increasing extinction rates but is relatively unaffected by clade size and ameliorated by incomplete sampling (c.f. Fujisawa & Barraclough 2013).

One solution, however, is to use a critical value for significance obtained from simulations (e.g. Maddison, Midford & Otto 2007; FitzJohn, Maddison & Otto 2009; Humphreys & Barraclough 2014). This entails comparing the LR difference in fit between null and alternative models for simulated data to that estimated from the empirical data. Using our CRBD simulations for this purpose reveals that the difference in fit between null and alternative models is significantly greater for empirical than simulated trees for all clades except Gnetales and waterbirds ($P < 0.01$; for lagomorphs, $P = 0.05$). For the other seven clades the CRBD model can be excluded. We therefore recommend use of simulations with empirical parameter values to judge significance of the GMYC model against alternative, clade-independent models. Based on the clades analysed here, a general rule of thumb seems to be that a LR ≥ 15 (ST) and ≥ 20 (MT) compared to the null model is indicative of empirical results that differ significantly from those expected under a BD process at $P = 0.05$ (Table 2). It is also possible to use specific estimates of the LR difference needed for significance for a given extinction rate, ε (Fig. 3).

In contrast, and as expected, the GMYC model could not discriminate clade-dependent hESUs from a clade-independent VRBD model, where the whole clade experienced a single shift in diversification rate (e.g. due to a change in environmental conditions). In some circumstances the GMYC model might still be able to distinguish these scenarios: for example, if origination of hESUs is staggered in time so that the most recent among-unit branching event postdates the most ancient within-unit branching event (Monaghan *et al.* 2009; Fujisawa & Barraclough 2013), but not in the clades analysed here.

How might we refine comparison of these alternatives, which are biologically interesting? The GMYC approach focuses on waiting intervals between branching events, but other features of tree shape might discriminate clade-dependent and clade-independent models. One possibility for improving model discrimination in future is to include additional metrics in model evaluation. We found that the clade-independent models analysed here do a poor job at capturing features of real (empirical) phylogenies. For example, the root height of VRBD trees is generally ridiculously old, e.g. on average 13.7 *billion* years for cycads, ~900 Ma for conifers and Gnetales and ~500 Ma for lagomorphs. In addition, there is a tendency for both CRBD and VRBD trees to be more balanced and stemmy than empirical trees but VRBD trees differ more from the empirical trees than do the CRBD trees, despite being simulated to more closely capture the LTT pattern of the empirical trees. The finding that CRBD models do not capture the shape of empirical trees is not new (Mooers & Heard 1997; Nee 2006) but less is known about VRBD models in this respect. Beyond this, and rather than comparing just one null and alternative model, a broad array of models could in principle be fitted to identify a confidence set of plausible models and parameter estimates consistent with the data.

Other considerations argue for biological relevance of the detected hESUs, whether those units result from a clade-dependent or clade-independent VRBD process. The hESUs correspond to various taxonomic ranks, revealing taxonomic inconsistencies among groups that are not surprising (e.g. Avise & Johns 1999; Holt & Jønsson 2014). However, the correspondence of hESUs with traditionally named taxa is striking for both mammals (families and genera) and gymnosperms (genera), suggesting that future efforts to understand diversification dynamics in these groups should focus on these ranks. There is less correspondence of bird hESUs with named higher taxa, although these results might be premature because the phylogenies analysed here are based on data for two thirds of the species only (Jetz *et al.* 2012). Intriguingly, the average age of hESUs in each bird, mammal and gymnosperm clade dates to the Miocene. In theory, this does not necessarily mean that anything special happened at that time (Fig. 1, and see Barraclough & Humphreys 2015) but might suggest similar, average turnover rates across clades. Previous analyses of birds and conifers have identified high rates of species turnover in regions characterised by climate fluctuations during the Neogene, including high latitude regions of the Northern Hemisphere and mountainous regions (Jetz *et al.* 2012; Leslie *et al.* 2012). Our results suggest not only general rules governing turnover rates among regions but that different types of organisms occupying these regions might be similarly affected by these rate-governing processes. Further research is needed to determine the causality and generality of these findings.

In conclusion, we have shown how inferences of hESU using the GMYC method are robust against some clade-independent models (CRBD), as long as simulations are used to evaluate these alternatives, but not against others (VRBD) that generate very similar patterns in waiting intervals between branching events. The differences between clade-dependent and clade-independent models are interesting biologically, however, and additional metrics either of tree-shape or evaluation of ecological trait distributions (Humphreys & Barraclough 2014) are needed to discriminate these alternatives. We suspect that clade-dependent models, focussing both on diversification rates and the units within which they operate, will prove important for explaining broadscale diversity patterns and encourage more research on this class of models.

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Data Accessibility

The R code used to calculate the non-cumulative stemminess index (St_N) is provided as online supporting information (SI Text S4). Trees simulated using empirical parameter values have been deposited in the Dryad Data Repository (doi: 10.5061/dryad.3rt26), as have the Gnetales and cycad trees. Mammal trees can be found in TreeBASE (study ID S15307).

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Supporting Information

The following supporting information is made available online as a single PDF file.

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Tables

Table 1. Fit of null, single (ST) and multiple (MT) threshold GMYC models for empirical clades and inferences across the confidence set of models.

Clade	**Lh (Null)**	**Lh (ST)**	**Lh (MT)**	**Models in 95% confidence set**	**hESUs**	**Mean threshold [Ma]**
Carnivores	319.46	326.05***	328.17***	6 *x* MT, 8 *x* ST	20 (17–24)	14.1 (13.1–15.4)
Euungulates	475.90	487.52***	491.87***	7 *x* MT	24 (18–29)	12.7 (11.2–15.5)
Lagomorphs	42.89	48.76**	49.35**	3 *x* MT, 8 *x* ST	6 (2–11)	8.36 (5.53–13.5)
Conifers	738.5	756.5***	761.7***	9 *x* MT	83 (75–90)	16.4 (14.9–17.4)
Cycads	318.8	367.5***	369.4***	2 *x* MT, 1 *x* ST	14 (12-16)	6.83 (6.34–7.63)
Gnetales	11.2	15.1*	16.0**	4 *x* MT, 25 *x* ST	17 (7–31)	5.65 (2.70–19.1)
Afroaves	3230.72	3238.98***	3262.43***	6 x MT	76 (71–81)	14.4 (14.1–14.8)
Nightbirds	1181.56	1186.08**	1201.13***	5 x MT	64 (60–66)	11.1 (10.8–11.5)
Waterbirds	2445.82	2447.45	2448.83*	5 x MT, 41 x ST, Null	42 (1–1027)	33.3 (79–0.00)

Asterices denote significance compared to the null at P = 0.05 (*), P = 0.01 (**) and P ≤ 0.001 (***).

Lh = Log likelihood

Ma = million years

Table 2. Performance of the GMYC applied to CRBD trees simulated using empirical parameter values for each study clade: the LR difference in fit between null and alternative models for empirical (LR_{obs}) and simulated trees (LR_{sim}) and error rate (rejection of the null).

		ST			MT		
Clade	**N**	**LR_{obs}**	**LR_{sim} (95%, 99%)[1]**	**Error rate**	**LR_{obs}**	**LR_{sim} (95%, 99%)[1]**	**Error rate**
Carnivores	235	13.2	6.95, 10.3	6.64%	17.50	10.2, 13.0	21.1%
Euungulates	302	23.4	9.58, 13.0	9.50%	31.94	12.5, 18.4	31.1%
Lagomorphs	71	11.7	9.22, 14.9	12.1%	12.92	13.2, 19.4	31.9%
Conifers	489	36.0	17.9, 22.5	56.8%	46.4	24.2, 29.1	88.9%
Cycads	204	97.4	11.6, 17.6	30.8%	101.2	19.9, 26.1	68.3%
Gnetales	72	7.80	12.5, 16.5	33.0%	9.60	15.6, 10.7	63.4%
Afroaves	1132	16.5	9.51, 13.0	12.2%	63.4	12.4, 14.5	44.9%
Nightbirds	556	9.04	10.2, 12.4	16.1%	39.1	16.4, 21.3	54.6%
Waterbirds	1028	3.26	8.45, 10.4	10.7%	6.02	10.4, 13.6	36.5%

[1] 95th and 99th percentiles

ST = single threshold GMYC method

MT = multiple threshold GMYC method

N = number of tips in phylogeny

LR = Likelihood ratio

Table 3. Median and 95% CI ST-GMYC inferences for VRBD-simulated trees where the null was rejected.

Clade	P ≤ 0.05[1]	hESUs	Threshold [Ma]	Relative depth of shift	$\lambda_{between}$	λ_{within}	λ_{within} / $\lambda_{between}$	$p_{between}$	p_{within}
Carnivores	47.4%	13 (2–28.9)	18.3 (13.4–45.6)	0.39 (0.20–1.00)	0.065 (2.0e-05–0.38)	0.31 (0.059–0.55)	4.77	0.87 (0.00–12.7)	0.41 (0.23–0.64)
Euungulates	100%	19 (11–34)	15.1 (12.0–21.9)	0.08 (0.047–0.16)	0.019 (0.0041–0.11)	0.33 (0.20–0.50)	17.37	0.86 (0.056–1.52)	0.43 (0.33–0.52)
Lagomorphs	74.5%	4 (2–12.4)	7.52 (4.92–11.3)	0.02 (0.33–0.0042)	5.6e-04 (6.1e-08–4.7e-02)	0.50 (0.25–1.54)	892.86	2.36 (-0.93–17.3)	0.45 (0.14–0.64)
Conifers	100%	29 (8.50–49.1)	28.0 (15.5–82.2)	0.031 (0.012–0.12)	0.0016 (0.00054–0.021)	0.071 (0.012–0.13)	44.38	1.34 (0.063–1.78)	0.84 (0.69–1.01)
Cycads	100%	28 (11–46)	6.79 (5.99–16.2)	0.00032 (0.00013–0.00099)	7.11e-05 (1.41e-05–9.81e-04)	0.32 (0.16–0.57)	4500.70	1.64 (0.94–2.35)	0.68 (0.41–0.88)
Gnetales	94%	6 (3–31.4)	6.59 (0.83–20.6)	0.0081 (0.0013–0.079)	7.35e-04 (4.48e-08–0.19)	0.24 (0.060–5.44)	326.53	1.76 (-0.31–12.9)	0.78 (0.048–1.16)
Afroaves	100%	49 (31–66)	31.4 (28.8–34.7)	0.60 (0.49–0.69)	0.088 (0.04–0.22)	0.85 (0.67–1.27)	9.66	0.98 (0.69–1.24)	2.22e-08 (1.03e-09–1.56e-07)
Nightbirds	100%	87 (63–112.1)	14.5 (11.7–15.5)	0.050 (0.031–0.076)	0.014 (0.0042–0.029)	0.32 (0.19–0.47)	22.86	1.00 (0.78–1.33)	0.41 (0.26–0.56)
Waterbirds	99%	15 (6.5–24)	40.2 (32.7–55.3)	0.20 (0.12–0.42)	0.015 (0.0050–0.092)	0.13 (0.096–0.19)	8.67	0.87 (0.0000082–1.56)	0.50 (0.45–0.55)

[1]Proportion of trees where single threshold (ST) GMYC detects a shift in diversification rate.

CI = confidence interval

VRBD = variable rate birth death

Ma = million years

Table 4. Performance of the GMYC applied to VRBD trees simulated using empirical parameter values for each study clade: the LR difference in fit between null and alternative models for empirical (LR_{obs}) and simulated trees (LR_{sim}).

Clade	ST		MT	
	LR_{obs}	LR_{sim} (95%, 99%)[1]	LR_{obs}	LR_{sim} (95%, 99%)[1]
Carnivores	13.2	15.5, 23.5	17.50	21.3, 29.7
Euungulates	23.4	84.6, 92.7	31.94	90.5, 100.2
Lagomorphs	11.7	80.0, 106.0	12.92	80.8, 106.3
Conifers	36.0	96.3, 107.6	46.4	100.9, 109.7
Cycads	97.4	332.1, 391.9	101.2	337.1, 395.7
Gnetales	7.80	62.9, 82.1	9.60	61.8, 83.9
Afroaves	16.5	319.3, 326.8	63.4	469.4, 498.6
Nightbirds	9.04	149.1, 153.3	39.1	156.2, 164.5
Waterbirds	3.26	85.4, 90.3	6.02	87.0, 93.8

[1]95th and 99th percentiles

ST = single threshold GMYC method

MT = multiple threshold GMYC method

LR = Likelihood ratio

Figure legends

Figure 1. Models of sharing and decoupling of speciation and extinction rates over time and among clades. In clade independent models these parameters apply across the entire clade and may be constant (CRBD) or variable (VRBD) over time. If variable over time, any rate shift that occurs at a given time, *T*, will affect all lineages equally, irrespective of clade membership. In contrast, in clade dependent models, speciation and extinction parameters will vary over time as well as being decoupled among clades, due to occupation of different geographical or ecological zones. Turnover through ongoing speciation and extinction will operate independently among such clades, referred to as higher evolutionarily significant units (hESUs). In this class of model, the threshold time, *T,* denotes the timing of the shift from among to within clade processes. However, *T* does not denote the timing of any particular event in the past, only the age of the most recent common ancestor of the oldest hESU, which depends on the rate of turnover in that hESU. CRBD = constant-rate birth-death; VRBD = variable-rate birth-death.

Figure 2. Error rates of the GMYC method applied above the species versus clade size (left) and relative extinction rate (ε, right). Results for CRBD trees simulated using parameter values estimated from the empirical clades (top row). Results for CRBD trees simulated using general parameter values, with 100%, 75% and 50% of the species retained (rows 2–4).

Figure 3. Rule-of-thumb likelihood ratio (LR) values needed for significance against a CRBD model based on relative extinction rate (ε) for the single-threshold (ST, top) and multiple-threshold (MT, bottom) version of the GMYC, at P = 0.05 (dashed line, open circles) and P = 0.01 (solid line, filled circles; LR values from Table 2). Fitted linear models: y=6.6x+7.4 (ST, P = 0.05); y=8.1x+10.6 (ST, P = 0.01); y=8.0x+10.5 (MT, P = 0.05); y=9.5x+14.3 (MT, P = 0.01). For example, a clade with average $\varepsilon = 0.2$ would need a LR $\geq$ 16.2 to reject the null in favour of the MT-GMYC at P = 0.01 and a clade with average $\varepsilon = 0.5$ would need a LR $\geq$ 19.1 (blue lines).

Detecting evolutionarily significant units above the species level using the Generalized Mixed Yule Coalescent method

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T.G. Barraclough

Supporting Information

Contents

SI Text

SI Text S1. SELECTION AND DEFINITION OF BIRD CLADES

Bird clades selected for analysis were generated by pruning the Hackett-backbone maximum clade credibility (MCC) tree of Jetz *et al*. (2012) to contain only the desired species. In addition, 500 posterior trees were obtained for each species set from birdtree.org (Jetz *et al.* 2012). The first clade is equivalent to Jarvis *et al.*'s (2014) Afroaves and includes groups such as the New World vultures, eagles, owls, mousebirds, cuckoo-roller, trogons, hornbills, woodpeckers, kingfishers, toucans, jacamars and bee-eaters. The second clade is Hackett *et al.*'s (2008) Caprimulgiformes (nightjars) and Apodiformes (hummingbirds and swifts) and Jarvis *et al.*'s (2014) Caprimulgimorphae, plus the kagu (*Rhynochetos*) and sunbittern (*Eurypyga*). Ericson *et al.* (2006) refer to this group as "nightbirds (except owls), swifts and hummingbirds". We adopt this name here ("nightbirds" for short), whilst being aware that as a whole, the clade is unsupported. The third clade is Jarvis *et al.*'s (2014) "core waterbirds" (including loons, penguins, fulmars, cormorants, ibises, herons and pelicans) plus turacos, rails, bustards, cuckoos, pigeons, mesites, sandgrouses, flamingos, grebes and the hoatzin. This forms a clade in the Hackett backbone tree of Jetz *et al.* (2012) although a relationship between core waterbirds and cuckoos and pigeons has not been found in other studies. We refer to this clade as "waterbirds".

SI Text S2. PHYLOGENETIC ANALYSES FOR CYCADS AND GNETALES

Phylogenies for cycads and Gnetales were generated using published sequences for chloroplast regions matK and rbcL, 18S of nuclear ribosomal DNA and, for cycads, the nuclear phytochrome-P gene (PHYP; Rydin & Korall 2009; Nagalingum *et al.* 2011; Hou *et al.* 2015). Sequences were aligned manually in Mesquite 2.74 (Maddison & Maddison 2010) because there were no alignment ambiguities. Using *Ginkgo biloba* as an outgroup, a RAxML (Stamatakis 2006; Stamatakis, Hoover & Rougemont 2008) tree was first inferred for each dataset. Then identical sequences were removed to improve convergence of downstream analyses. Based on the trimmed dataset, time-calibrated trees were generated in Beast (Drummond & Rambaut 2007; Drummond *et al.* 2012) using a number of topological constraints and age priors (Table S1), a GTR + G substitution model, unlinked among gene regions, a birth-death tree prior (Gernhard 2008) and either the RAxML (cycads) or a random (Gnetales) starting tree. A random starting tree together with more topological constraints improved convergence and sampling of parameters for Gnetales. For each dataset, four runs of $80 \, x \, 10^6$ generations, sampling every 1000, were performed. Convergence of runs and sampling and mixing of parameters were assessed in Tracer (Rambaut & Drummond 2007), measured as a combined effective sample size of ~200. A set of 20,000 trees was sampled from across all four (cycads) or three (Gnetales) runs post burnin and tree statistics were summarised on the MCC tree from that set, retaining the node heights of the MCC tree. Analyses were run on the Cipres Science Gateway (Miller *et al.* 2012).

SI Text S3. SCALING GMYC RATES TO MATCH THOSE OF THE BIRTH-DEATH PROCESS

Diversification rates inferred from the best-fitting MT-GMYC models are on average much higher than those inferred from CRBD models (except for waterbirds; Table S4). This is because the equations underlying the GMYC are quite different from those underlying the CRBD process. Preliminary analyses showed that simulating trees using the raw GMYC estimates produced trees that were extremely young, with an extremely deep relative shift position (i.e. the shift is pushed back in an attempt to accommodate the predefined absolute shift time but the process is so quick that the tree grows to the specified number of tips before it reaches the specified shift time). Therefore the GMYC rates were scaled to match those expected under a CRBD process by dividing the average GMYC rate with the CRBD diversification rate. This amendment allowed fixing the position of the shift in absolute time at a position that matched the empirically inferred threshold time.

SI Text S4. R CODE USED TO CALCULATE THE NON-CUMULATIVE STEMMINESS INDEX (St_N)

```
# Function for calculating Rohlf's Stemminess Index on a
 single tree
# Reference: Rohlf, F.J., Chang, W.S., Sokal, R.R. & Kim, J.
 (1990) Accuracy of estimated phylogenies: effects of tree
 topology and evolutionary model. Evolution, 44, 1671-1684.

stemminess= function(tr){t <- Ntip(tr)
h <- numeric()
for (i in (1:length(tr$edge[,1]))){
 times <- branching.times(tr)
 names(times) <- c((Ntip(tr)+1):(Ntip(tr)+Nnode(tr)))
 if (tr$edge[i, 2]>=(Ntip(tr)+1))
 h[i] <- times[names(times)==tr$edge[i,1]]
 else h[i] <- 0
 }
h <- h[which(h>0)]
w <- tr$edge.length[which(h>0)]
ST <- sum(w/h)
St <- (1/(t-2))*ST
return(St)
}
```

SI Tables

SI Table S1. Topological constraints and age priors used in the Beast analyses for cycads and Gnetales.

Taxon[1]	Age prior [Ma]	Reference
Cycadales		
Cycadales	208.0 ± 2	(Nagalingum *et al.* 2011)
Cycadales excl. *Cycas*	120.6 ± 2	(Nagalingum *et al.* 2011)
Cycas	10.5 ± 1	(Nagalingum *et al.* 2011)
Encephalartos	9.5 ± 0.5	(Nagalingum *et al.* 2011)
Zamia	8.0 ± 1	(Nagalingum *et al.* 2011)
Macrozamia	5.0 ± 1	(Nagalingum *et al.* 2011)
Ceratozamia	7.5 ± 1.5	(Nagalingum *et al.* 2011)
Encephalartos+*Lepidozamia*	40.0 ± 2	(Nagalingum *et al.* 2011)
Bowenia + its sister clade	102.0 ± 2	(Nagalingum *et al.* 2011)
Root height (gymnosperms)	387.0 ± 0.005	(Magallón 2010) [2]
Gnetales		
Gnetales	167 ± 2	(Rydin *et al.* 2006; Ickert-Bond, Rydin & Renner 2009)
Welwitschia + *Gnetum*	111.4 ± 2	(Ickert-Bond, Rydin & Renner 2009)
Gnetum	None	(Won & Renner 2006; Hou *et al.* 2015)
*Gnetum*_South America	None	(Won & Renner 2006; Hou *et al.* 2015)
*Gnetum*_trees	None	(Won & Renner 2006; Hou *et al.* 2015)
*Gnetum*_Asia I	None	(Won & Renner 2006; Hou *et al.* 2015)
*Gnetum*_Asia II	None	(Won & Renner 2006; Hou *et al.* 2015)
Ephedra	30.39 ± 5	(Ickert-Bond, Rydin & Renner 2009)
*Ephedra*_New World	None	(Rydin & Korall 2009)
*Ephedra*_China	None	(Rydin & Korall 2009)
*Ephedra*_Asia I+Horn of Africa	None	(Rydin & Korall 2009)
*Ephedra*_Asia I	None	(Rydin & Korall 2009)
*Ephedra*_Asia I+II+Horn of Africa	None	(Rydin & Korall 2009)
*Ephedra*_Asia II	None	(Rydin & Korall 2009)
Root height (gymnosperms)	387.0 ± 0.005	(Magallón 2010) [2]

[1]All listed clades were constrained to be monophyletic. Some were also given an age prior.

[2]The oldest estimate for seed plants (node 9; Magallón 2010). If gymnosperms are monophyletic (node 10), it is equivalent to their stem age.

Ma = million years

SI Table S2. Taxonomic diversity[1] and sampling of empirical clades.

Clade	Orders	Families	Genera	Species	Sampling
Carnivores	1	16	123	285	83%
Euungulates	2	24	138	346	87%
Lagomorphs	1	2	12	92	77%
Conifers	1	6	72	~550[2]	~89%
Cycads	1	2	10	~300[2]	~68%
Gnetales	1	3	3	~80	~90%
Afroaves	7	21	218	1132	100%[3]
Nightbirds	3	10	148	566	100%[3]
Waterbirds	17	31	223	1028	100%[3]

[1]Numbers were taken from Wilson & Reeder (2005) and IUCN (2011) species lists for mammals; Jetz *et al.* (2012) for birds, where the taxonomy is mainly based on BirdLife International (www.birdlife.org) and the International Ornithologists' Committee (www.worldbirdnames.org); Leslie *et al.* (2012) and Eckenwalder (2009) for conifers; Nagalingum *et al.* (2011) and the World List of Cycads (www.cycadlist.org) for cycads; and Kubitzki (1990) for Gnetales.

[2]These figures are low compared to some recent estimates. It is uncertain how much of recent increases in species numbers is due to true species discovery and how much to do with taxonomic splitting (e.g. Eckenwalder 2009).

[3]DNA data exist for two thirds of the species on average. The remainder have been modelled in by the authors based on prior taxonomic knowledge (Jetz et al. 2012).

SI Table S3. Parameter values for CRBD simulations based on estimates across each of the nine empirical clades.

Clade	**N**	**μ**	**λ**	**μ/ λ**
Carnivores	235	0.00 (0.00–0.00)	0.12 (0.11–0.12)	0.00 (0.00–0.00)
Euungulates	302	0.032 (0.01–0.05)	0.13 (0.11–0.14)	0.25 (0.08–0.35)
Lagomorphs	71	0.030 (0.00–0.10)	0.19 (0.16–0.23)	0.16 (0.00–0.44)
Conifers	489	0.30	0.31	0.98
Cycads	204	0.30 (0.25–0.36)	0.31 (0.26–0.36)	0.98 (0.96–0.99)
Gnetales	72	0.38 (0.23–0.71)	0.38 (0.23–0.71)	1.00 (1.00–1.00)
Afroaves	1132	0.038 (0.012–0.075)	0.12 (0.097–0.15)	0.32 (0.13–0.51)
Nightbirds	566	0.067 (0.041–0.091)	0.13 (0.11–0.16)	0.50 (0.35–0.59)
Waterbirds	1028	0.018 (0.0037–0.033)	0.083 (0.071–0.097)	0.22 (0.050–0.35)

Estimates are based on 500 trees apart from for conifers where 1 empirical tree was used only. Estimates for mammals are from Humphreys & Barraclough (2014) and are provided here for comparison only.

N = number of tips

SI Table S4. Parameter values for VRBD simulations based on estimates from the best-fitting GMYC models for each of the nine empirical clades.

Clade	N	λ_2	λ_1	λ_1/λ_2	p_2	p_1	T	Scale
Carnivores	235	0.19	0.42	2.21	0.40	0.25	13.1–15.4	2.58
Euungulates	302	0.062	0.82	13.23	0.79	0.17	11.2–15.5	4.54
Lagomorphs	71	0.0092	1.60	173.91	2.65	0.00	5.53–13.5	5.05
Conifers	489	0.016	0.15	9.38	0.93	0.49	14.9–17.4	8.3
Cycads	204	0.011	2.45	222.73	1.14	0.076	6.34–7.63	123.1
Gnetales	72	0.0082	0.41	50.16	1.47	0.34	2.70–19.1	20.9
Afroaves	1132	12.1	0.44	0.04	-1.63	0.25	14.1–14.8	76.8
Nightbirds	566	0.16	1.34	8.38	0.54	3.6×10^{-8}	10.8–11.5	11.5
Waterbirds	1028	0.0097	0.075	7.73	3.27	0.51	33.3	0.65

N = number of tips

λ_1 = pre-shift / within-hESU rate

λ_2 = post-shift / among-hESU rate

p_1 = pre-shift / within-hESU scaling parameter

p_2 = post-shift / among-hESU scaling parameter

T = shift position (for waterbirds mean threshold time is used; for all others the range estimated from confidence set of models is used)

Scale = average GMYC rate divided by the CRBD diversification rate (λ-μ)

SI Table S5. Models for explaining the relationship between relative extinction rate (ε) and error rates in CRBD trees simulated using general parameters values.

Dataset	Model	F	D.f.	GCV	P	R^2
ST 100%	Gaussian	19.13	15.35	140.47	< 0.001	0.75
MT 100%	Linear	105.4	16.00	-	<< 0.001	0.86
ST 75%	Gaussian	14.83	14.98	97.01	< 0.001	0.73
MT 75%	Gaussian	40.44	15.24	79.94	<< 0.001	0.87
ST 50%	Gaussian	7.32	14.77	59.21	0.0026	0.56
MT 50%	Gaussian	13.74	14.86	105.18	< 0.001	0.72

SI Table S6. Fit of the MT-GMYC model to trees simulated with the VRBD model.

Clade	Lh (Null) [1]	Lh (ST) [1]	Lh (MT) [1]	MT > ST[2]
Carnivores	375.7	379.0	381.6	46 (60)
Euungulates	573.1	600.5	602.2	19 (42)
Lagomorphs	67.7	83.7	84.8	14 (30)
Conifers	1394.0	1425.0	1427.0	23 (63)
Cycads	211.3	328.7	329.8	6 (27)
Gnetales	97.7	112.9	130.8	10 (24)
Afroaves	2666.0	2797.0	2837.0	67 (69)
Nightbirds	1197.0	1251.0	1256.0	51 (51)
Waterbirds	2930.0	2955.0	2956.0	11 (37)

[1]Average across 100 trees

[2]Number of trees where the MT-GMYC fits the data better than ST-GMYC at $\Delta AIC \geq 5$ (in brackets, at $\Delta AIC \geq 3$).

SI Table S7. Tree characteristics of empirical and simulated trees of each size set.

		I_C			I_S			St_N	
Dataset	**MCC**	**CRBD**[1]	**VRBD**[1]	**MCC**	**CRBD**[1]	**VRBD**[1]	**MCC**	**CRBD**[1]	**VRBD**[1]
Carnivores	0.81	0.16 (-1.15–2.35)	-0.0011 (-1.37–1.94)	0.75	0.15 (-0.82–1.88)	-0.040 (-0.97–1.63)	0.70	0.67 (0.51–0.94)	0.74 (0.54–1.25)
Euungulates	4.88	0.059 (-1.17–2.28)	-0.10 (-1.37–1.58)	4.15	0.056 (-0.89–1.96)	-0.032 (-1.03–1.33)	0.57	0.70 (0.55–1.04)	1.14 (0.75–2.04)
Lagomorphs	1.16	0.022 (-1.13–1.81)	-0.11 (-1.33–1.60)	0.90	0.024 (-0.79–1.52)	-0.10 (-0.91–1.37)	1.08	0.65 (0.47–1.43)	0.93 (0.44–23.4)
Conifers	2.76	-0.060 (-1.15–1.71)	-0.071 (-1.13–1.67)	2.44	-0.057 (-0.88–1.37)	-0.087 (-0.85–1.37)	1.39	1.69 (1.19–3.80)	5.45 (1.88–16.2)
Cycads	1.59	-0.026 (-1.23–2.11)	-0.11 (-1.22–1.28)	1.37	-0.0064 (-0.89–1.83)	-0.068 (-0.90–1.14)	1.75	1.43 (0.86–3.36)	123.4 (35.5–446.5)
Gnetales	0.16	-0.091 (-1.13–1.76)	-0.19 (-1.11–1.63)	0.13	-0.097 (-0.85–1.55)	-0.13 (-0.80–1.27)	0.52	1.33 (0.58–3.93)	8.27 (0.63–143.3)
Afroaves	4.04	-0.10 (-1.31–1.65)	-0.022 (-0.96–1.53)	3.28	-0.10 (-0.93–1.25)	-0.032 (-0.74–1.19)	0.63	0.76 (0.64–1.01)	0.45 (0.44–0.48)
Nightbirds	6.17	-0.092 (-1.23–1.85)	0.028 (-1.30–1.51)	5.54	-0.10 (0.91–1.46)	0.0041 (-0.89–1.28)	0.51	0.79 (0.64–1.09)	1.84 (1.24–3.36)
Waterbirds	2.45	-0.050 (-1.32–1.68)	-0.10 (-1.27–1.64)	2.29	-0.088 (-0.99–1.42)	-0.10 (-0.97–1.31)	0.53	0.72 (0.61–0.94)	0.78 (0.64–1.07)

[1]For simulated trees median and 95% confidence interval are shown.

I_c = Colless' index of tree imbalance

I_s = Sackin's index of tree imbalance

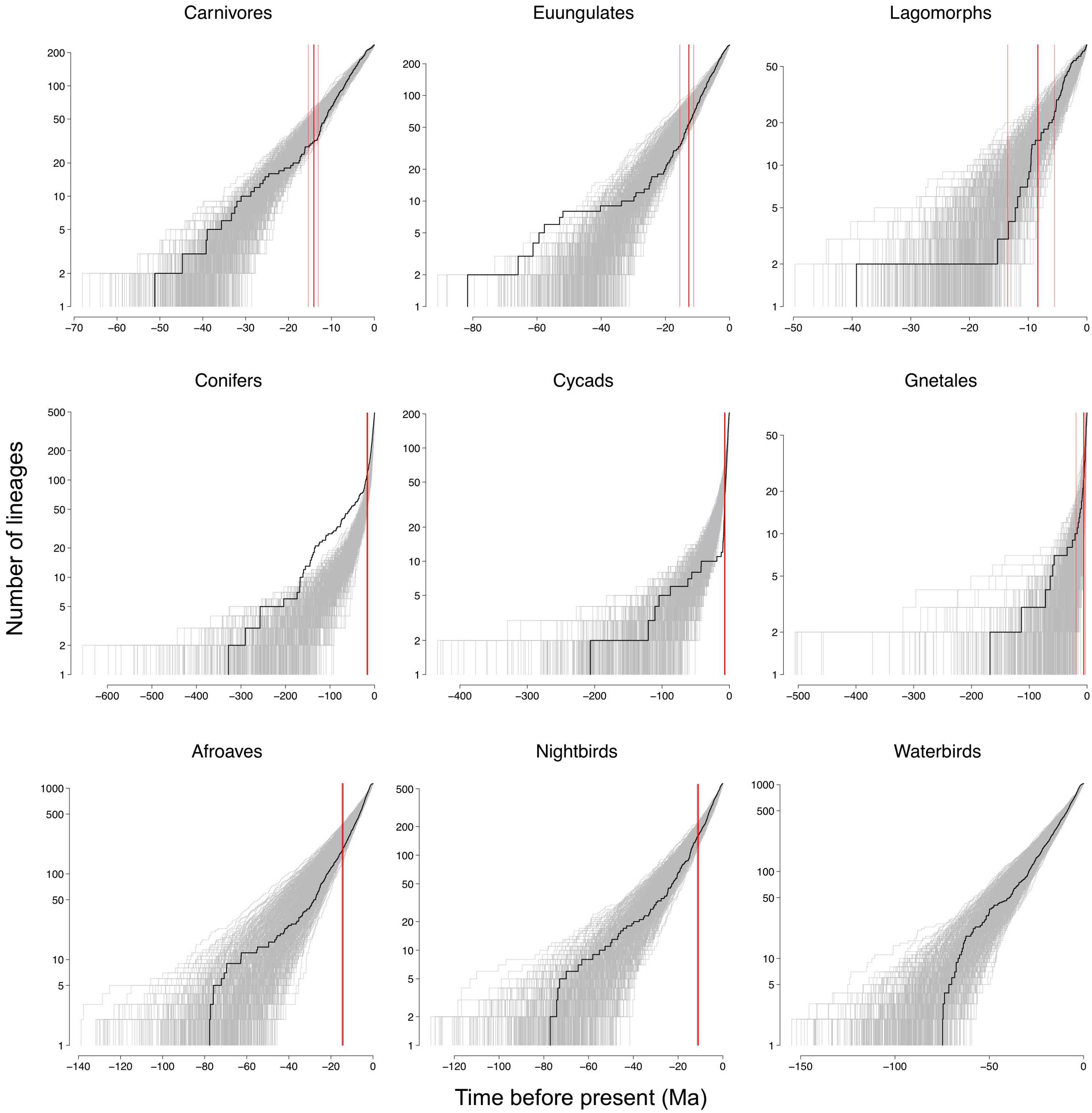
St_N = Rohlf's non-cumulative stemminess index

MCC = maximum clade credibility tree

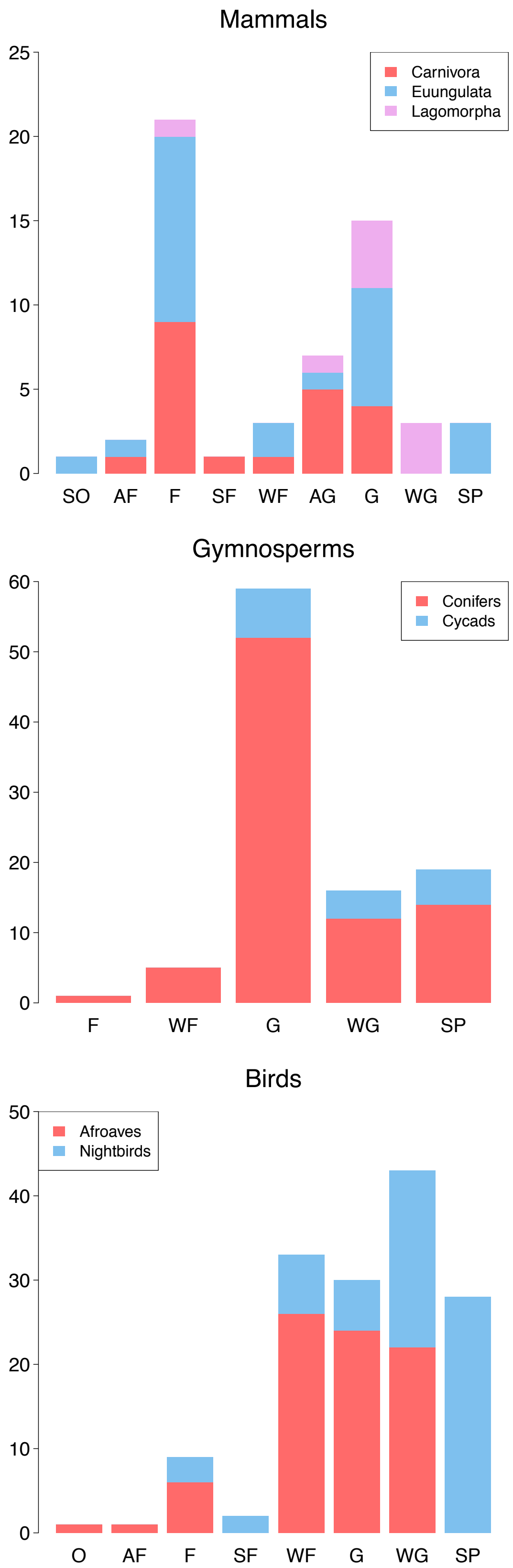
CRBD = trees simulated under a constant rate birth-death process

VRBD = trees simulated under a variable rates birth-death process

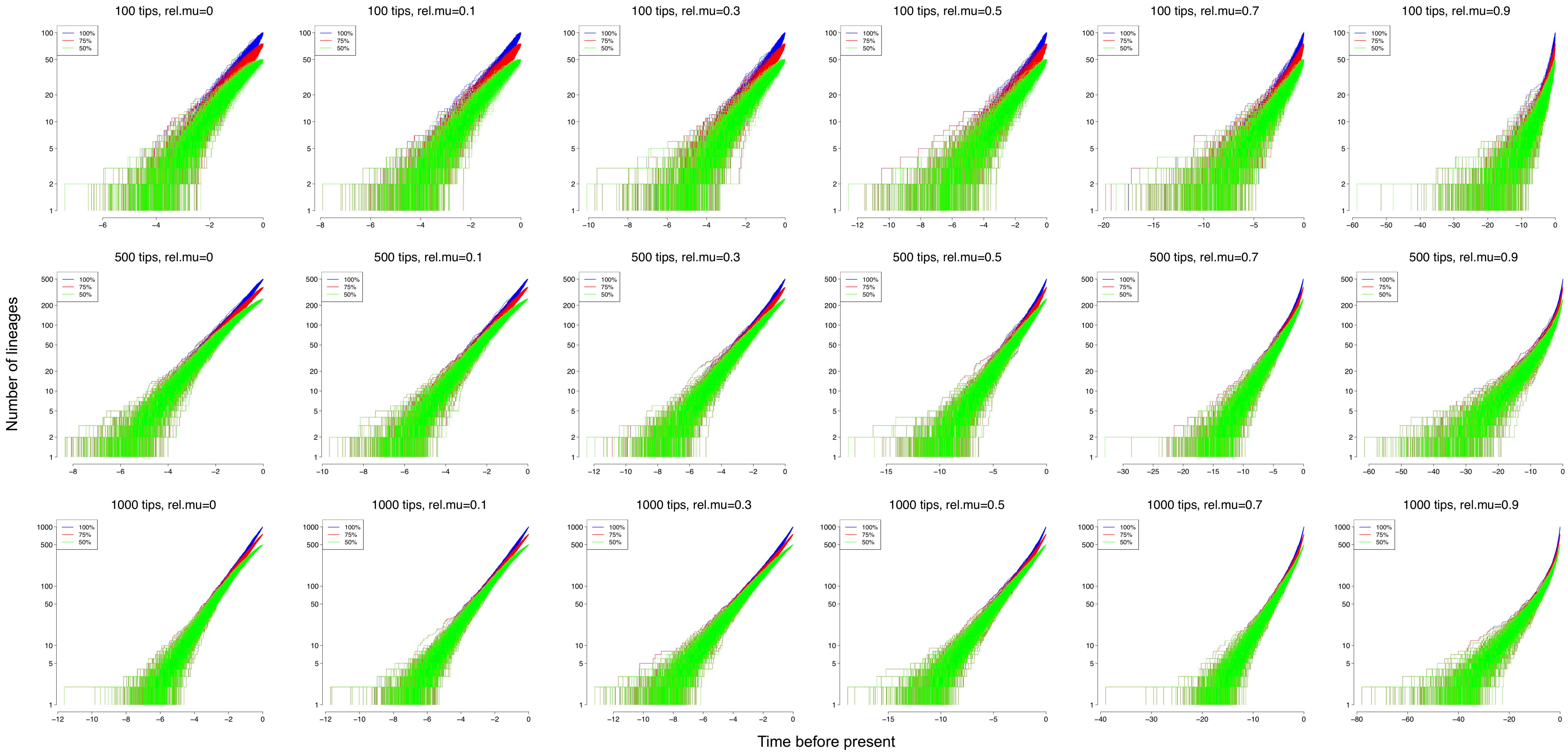
SI Figures



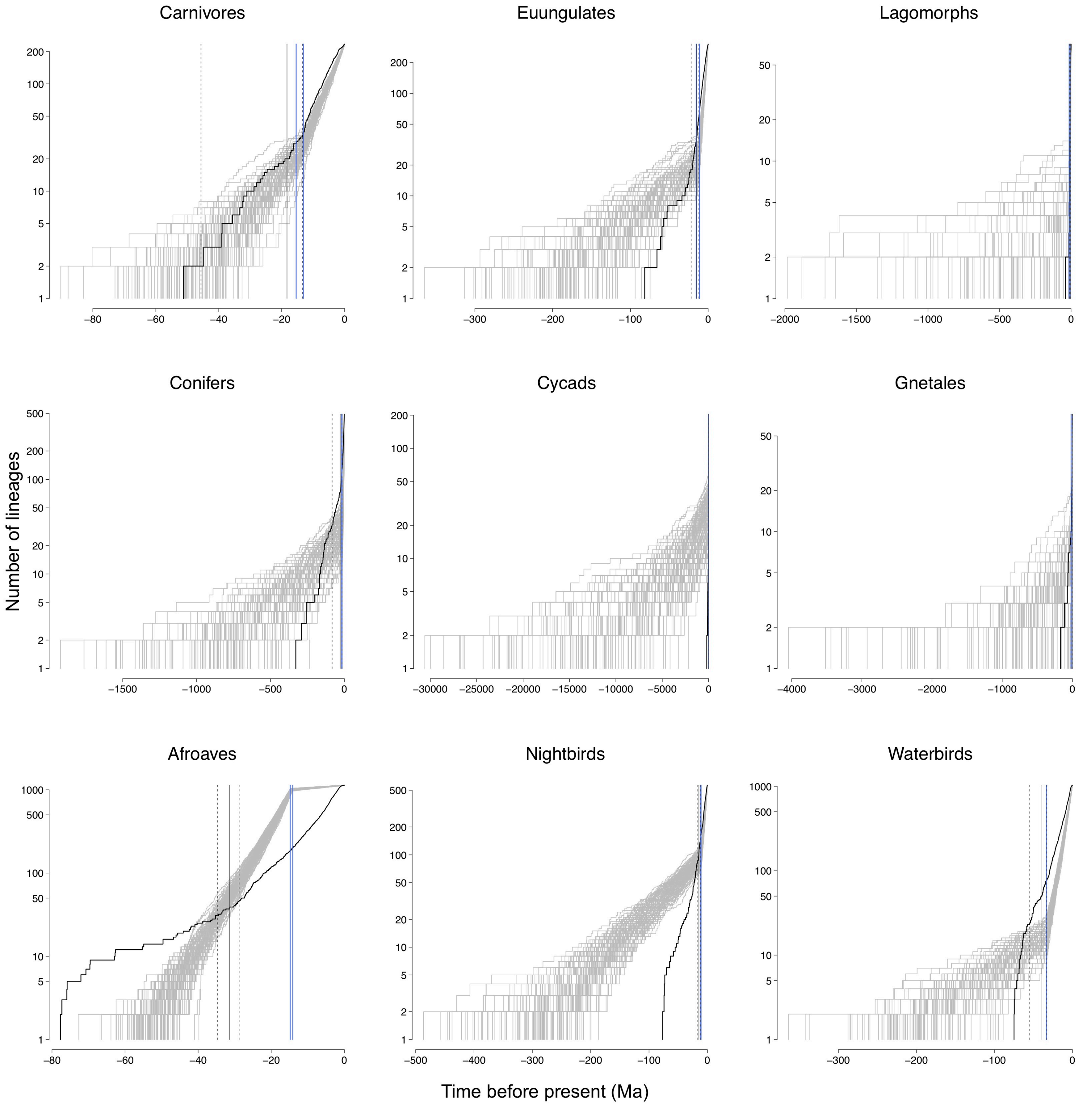
SI figure S1. Lineages-through-time plots for trees simulated under CRBD models (grey) and the MCC tree for each empirical clade (black), with the threshold range (red, vertical lines) inferred from the 95% confidence set of best GMYC models.



SI figure S2. Taxonomic rank of hESUs inferred for the a) mammal, b) gymnosperm and c) bird clades, where empirical results differ significantly from those expected under the constant-rate birth-death model (i.e. excluding results for Gnetales and waterbirds). hESUs = higher evolutionarily significant units. O = Order, SO = "Subor-der" (comprises one suborder, one infraorder and one superfamily), AF = Above-family, F = Family, SF = Subfamily, WF = Within-family, AG = Above-genus, G = genus, WG = Within-genus, SP = Species. (Bold text denotes formal taxonomic ranks.)



SI figure S3. Lineages-through-time plots for trees simulated under constant-rate birth-death models with general parameter values and 100% (blue), 75% (red) and 50% (green) sampling. Note that the plots are identical early on, with differences appearing only toward the present due to removal of tips.



SI figure S4. Lineages-through-time plots for trees simulated under VRBD models (grey) and the MCC tree for each empirical clade (black). Vertical lines are threshold times inferred with the GMYC method for simulated (grey) and empirical (blue) trees. The empirical threshold time overlaps with range of threshold times under which trees were simulated for all clades except those simulated using Afroaves parameter values. For trees simulated using lagomorph and Gnetales parameter values the overlap was almost perfect, for euungulate and cycad-based trees inferred shift times extended a little deeper and for the other five clades the range of inferred shift times extended much deeper than the range under which they were simulated.

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