Using Phylogenetic Comparative Methods To Understand Diversification and Geographic Range Evolution

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DEDICATION

I dedicate my work to the loved ones that I lost during my time as a graduate student – my father and my beautiful parrot, Izze. You both left this world far too soon.

Papi, gracias por siempre apoyarme cuando pequeña. Me diste los recursos para engendrar y motivar mis curiosidades científicas. Tu fé en mi me mantuvo enfocada.

Izze, you were my strength. There was nothing I cared for more in this world. You gave me hope when there was none. You taught me how to fight. This world is a lonelier place without you. Your bouts of flight inspired my third chapter.

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ABSTRACT

Two key processes that have been modeled in a phylogenetic comparative framework are diversification and historical biogeography. Many questions arise on what process have shaped the abundance (or lack) of species we see today and what influences their survival and interconnectedness with other species. Many methods have been developed to answer these questions. Over the past several decades there has been a rise in parametric modeling and development of more adequate frameworks to answer biological questions of interest. However, many models still lack the incorporation of ecological, mainly biotic factors, which influence the evolution and ecology of species, while accounting for phylogenetic relatedness.

In my dissertation, I studied a diverse set of questions in the realm of diversification and biogeography. I began my investigation by improving upon the widely used Dispersal-Extinction and Cladogenesis model and showing my method DEC* is a more adequate model that usually has better model fit and parameter estimation (Chapter 1). I then moved on to learn how a complex trait such a parasitism could influence the diversification of parasitic angiosperms with a wide array of diversification models in phylogenetics (Chapter 2). My results indicated that all parasitic flowering plants are not undergoing an evolutionary dead end, as many have postulated. In my final chapter, I merged trait and biogeographic evolution by assembling a model that would test the influence of body size in passerine birds. Additionally, I tested the influence of several traits upon the diversification of passerine birds (Chapter 3). The results showed that the larger body sizes are not associated with greater dispersal rates in passerines. According to the results, smaller birds have a greater rate of dispersal. I also show support for both body size and region of occurrence in the world (temperate versus tropical) as having an influence on

diversification. For the diversification analyses in Chapter 2 and 3, I find evidence of underlying biological trait and/process influencing the diversification of these groups.

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INTRODUCTION

There are many different processes by which organisms can evolve. As evolutionary biologists we try to quantify these processes by collecting data and inferring the likely scenarios describing the evolutionary history of species. Two main types of modeling frameworks to understand the evolution of species are diversification and geographic range evolution/historical biogeography. Under diversification models, we can understand the speciation and extinction processes underlying a group of species and with historical biogeography approaches, we can understand the evolutionary history of species ranges through time and what processes have shaped these.

Trait Evolution

Traits are commonly used to assess the diversification of species and many such methods have been developed to ascertain how a trait could influence the macroevolution of a clade. Trait types can vary from trophic level, morphology, behavior, pollinator specialism, host ranges and can be classified into discrete and continuous classes depending on the data type. Among these, morphological traits are commonly used and many studies have sought to understand how phenotypes have influenced evolution of a clade. In particular, body size has been shown to shape the evolutionary history of many species (Jablonski 1997; Maurer 1998; Hillebrand and Azovsky 2001; Ashton 2002; Thomas et al. 2009). A trait that is less commonly explored is parasitism. However, a focus of one of the chapters will be to understand how parasitism has influenced diversification in angiosperm lineages. The parasitic trait is more of a continuous trait, where a species will fall on a spectrum of degree of parasitism. At the most extreme end is an endoparasite – a species that has completely lost its ability to photosynthesize and lives within the host itself. On the other extreme are hemiparasites, which have chlorophyll and thus might

have the capacity to photosynthesize, but mildly depend on a host for nutrients, carbohydrates, or placements, for instance.

Evolutionary biologists have been interested in understanding the diversity of species. Different traits set species apart. These unique traits have been a focus of interest to understand whether they have been key innovations or have caused clades to have lower diversification rates (speciation – extinction). The field of phylogenetics has methods to explicitly test whether traits could be key innovations using phylogenies (an inferred evolutionary history of the relationship of species). The evolutionary history of a clade can be used in conjunction with traits and an inference method to learn about the underlying evolutionary process of interest. Different questions can then be asked with these methods, such as does trait *X* influence the diversification of clade A, has speciation increased through time due to trait *Y*, and how do birth and death rates compare between sister clades, among other questions. A review of methods used to test such questions and the connections between them can be found in O'Meara (2012).

There are two main types of diversification methods that my research will focus on using: lineage or non-trait, and trait-dependent diversification. MEDUSA (Modeling Evolutionary Diversification Using Stepwise Akaike Information Criterion) is one such method used to model lineage diversification, which detects multiple shifts in birth and death rates given a phylogeny (Alfaro et al. 2009). Other methods, such as Lineage Through Time Plots (Harvey et al. 1994; Stadler 2008), model the number of species through time and then fit the best model to understand the fluctuation in the number of species (Rabosky 2006; Rabosky and Lovette 2008; Rabosky 2014). Another method is BAMM (Bayesian Analysis of Evolutionary Mixtures), which models the speciation and extinction of a lineage given a phylogeny. It also tells the shifts based on the heterogeneity of the diversification (Rabosky et al. 2013). However, due to recent

contention regarding its accuracy of likelihood estimation (Moore et al. 2016), I decided to not pursue using it for my dissertation. A seminal model called BiSSE (Binary State Speciation and Extinction) models trait-dependent speciation and extinction (Maddison et al. 2007). Unfortunately, there are limitations for this model that need to be considered before usage, such as a minimum threshold of taxa and the ratio of the binary traits (Davis et al. 2013). My second chapter seeks to improve upon these shortcomings. Recently, there has been much debate on whether the trait of interest might be causing the pattern of evolution seen for a clade, or if another unknown trait that is hidden could also be influencing what we observe. The authors of BiSSE discussed this in Maddison et al. (2007) and others have commented regarding this issue. The method HiSSE (Beaulieu and O'Meara 2016) tests for this, so now the possibility of testing for the trait of interest and some other hidden trait that could be the causative agent as well, is possible. The field of diversification is ripe with methodology to test the diversification of lineages using a vast variety of modeling frameworks. I seek to use these methods to understand whether the results tell the same story among the study systems I use.

Historical Biogeography

Historical biogeography seeks to understand the distribution of biodiversity through space and time. Of broad interest is the curiosity over the past distribution of species: can we infer past ranges, and if so, can that give us more information to predict their future range? Is biogeography important to the evolution of biodiversity? How important was geological history in the evolution of taxa? As methods in geographic range evolution have evolved to allow the incorporation of more information required to answer questions of this caliber, we can expect in the near future to have more satisfying answers.

By knowing the current distribution of species, scientists have attempted to infer ancestral state distributions of organisms with phylogenetic methods. The history of this field has evolved from observing the general patterns of species, tracking their distribution (Croizat 1958), to incorporating events that explain biogeographic processes (such as vicariance and dispersal) (Ronquist 1997) to developing probabilistic approaches (Ree et al. 2005a; Ree and Smith 2008; Matzke 2014). During the time when non-parametric event-based methods in biogeography dominated, many scientists contended whether geological events (vicariance advocates) better explained species distribution patterns or recent migration events (dispersalists). However, with the advent of parametric methods based on Maximum Likelihood and Bayesian frameworks, researchers have been able to incorporate important data and ask a wider variety of research questions, therefore obviating the vicariance versus dispersal predicament, which has impeded progress in historical biogeography. Also, parametric methods allow for important information to be incorporated, such as branch lengths of a phylogeny. Please see Sanmartín (2012) for insights on the evolution of historical biogeography as a field and Ronquist and Sanmartín (2011) for a thorough review of methods.

However, there are a limited amount of model-based methods to test hypotheses regarding historical biogeography, and most of these only handle discrete data. Therefore, to further improve the field, it is important to develop new methods as well as improve current ones. Many scientists agree that more statistical methods in historical biogeography need to be developed (Donoghue and Moore 2003; Ree et al. 2005b; Ree and Sanmartín 2009; Goldberg et al. 2011; Landis et al. 2013; Matzke 2014). Parametric methods allow more information to be incorporated, such as fossils, to more accurately measure the historical biogeography of taxa (Wood et al. 2012). There are extensions to the Dispersal-Extinction-Cladogenesis model that

allow for the incorporation of distance between areas (Van Dam and Matzke 2016), jump dispersal (Matzke 2014), and stochastic mapping (Fabiola Soto-Trejo 2017). There are also methods that allow for historical biogeography inference when the number of ranges is large (Landis et al. 2013). There are methods that seek to use historical biogeography for dating species (Landis 2016). And lastly, there are methods that allow for the estimation of related geographic distribution tied to rates of diversification modeled in a probabilistic framework (Goldberg et al. 2011). Since I began my dissertation work in geographic range evolution, the use of probabilistic methods has increased (Figure 1.1). The number of citations increased from June 2015 to March 2017.

All the methods I previously mentioned are all based on discrete biogeography data. Methods in historical biogeography have focused on models which use pre-defined, discrete areas for species, indicating for each species its presence or absence in each area. An example of a pre-defined area dataset is one that uses the Hawaiian archipelago, such as Hawaii, Oahu, Maui, and Kauai as discrete areas. Therefore, areas are discrete units of geographic range that are generally arbitrarily defined, and are analogous to discrete character states (Ree et al. 2005b), such as parasitic or non-parasitic, or dioecious and monoecious. Despite the arbitrary designation of discrete states in biogeography studies, the use of discrete area state analyses in historical biogeography has been more popular than continuous biogeography, which has been proposed in the field of phylogeography (Lemmon and Lemmon 2008). This may be the case since most empirical biogeographic analyses are about clades found in islands systems, which are essentially discrete areas, and discrete models are easier to develop. Although, see Ronquist and Sanmartín (2011) for a more thorough explanation on this. However, if the clade of interest has a range within continents or islands, then discretizing oversimplifies the data. Lemmon and

Lemmon (2008) developed a maximum likelihood model that estimates the phylogeographic history of a gene and assesses the biogeography of the ancestors at nodes in the clade of interest in a continuous landscape, which could be adapted for historical biogeography.

Goals of the Dissertation Research

I am interested in addressing trait evolution and historical biogeography of species through phylogenetic methods. My work will seek to: 1) understand the diversification of parasitic plants and passerine birds, and 2) develop a new parametric method in historical biogeography. I also carry out a new approach using the BiSSE model to circumvent some of its limitations.

Historical biogeography has been hindered by much contention between which event-based processes are important. Probabilistic approaches have helped ameliorate this due to having parameters estimate these processes. It is important for the future of the field of biogeography to improve upon existing methods to make them more adequate to fit the biological data and to develop new methods to allow researchers to ask more questions of interest. Chapter 1 is an extension of the popular Dispersal-Extinction Cladogenesis model that improves parameter estimation for extinction and dispersal. This was tested via simulations. My model, named DEC*, also has better model fit and adequacy over the DEC model, tested on 15 independent datasets across different taxa, suggesting it should be considered in models of geographic range evolution.

The second chapter seeks to understand the diversification of parasitic angiosperms across an expansive range of parasitic clades. Parasitic plants have been understudied and work regarding their diversification and biogeography would be a substantial contribution. I use a wide range of phylogenetic comparative methods to assess diversification and observe robustness

across results. These results are compared with the non-parasitic sister clade counterparts to understand how the diversification process compares.

An integrated modeling approach that incorporates the influence of a trait upon dispersal and local extinction with regards to biogeography has not been studied much in the literature, except in Sukumaran et al. (2016). A trait and biogeography model would be useful for assessing how traits could affect biogeographic history. Merging traits and biogeography is an approach to assess how morphology (i.e. fruit type, growth habit), behavior (i.e. aggressive, non-aggressive), diet (i.e. carnivore, herbivore), and other factors may affect the range of species. In my third chapter, using popular models of geographic range evolution, I test whether body size influences dispersal in passerine birds. I also use methods of lineage and trait-dependent diversification to understand whether larger passerines have an increased diversification, and whether tropical species of passerines have increased diversification.

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Appendix A

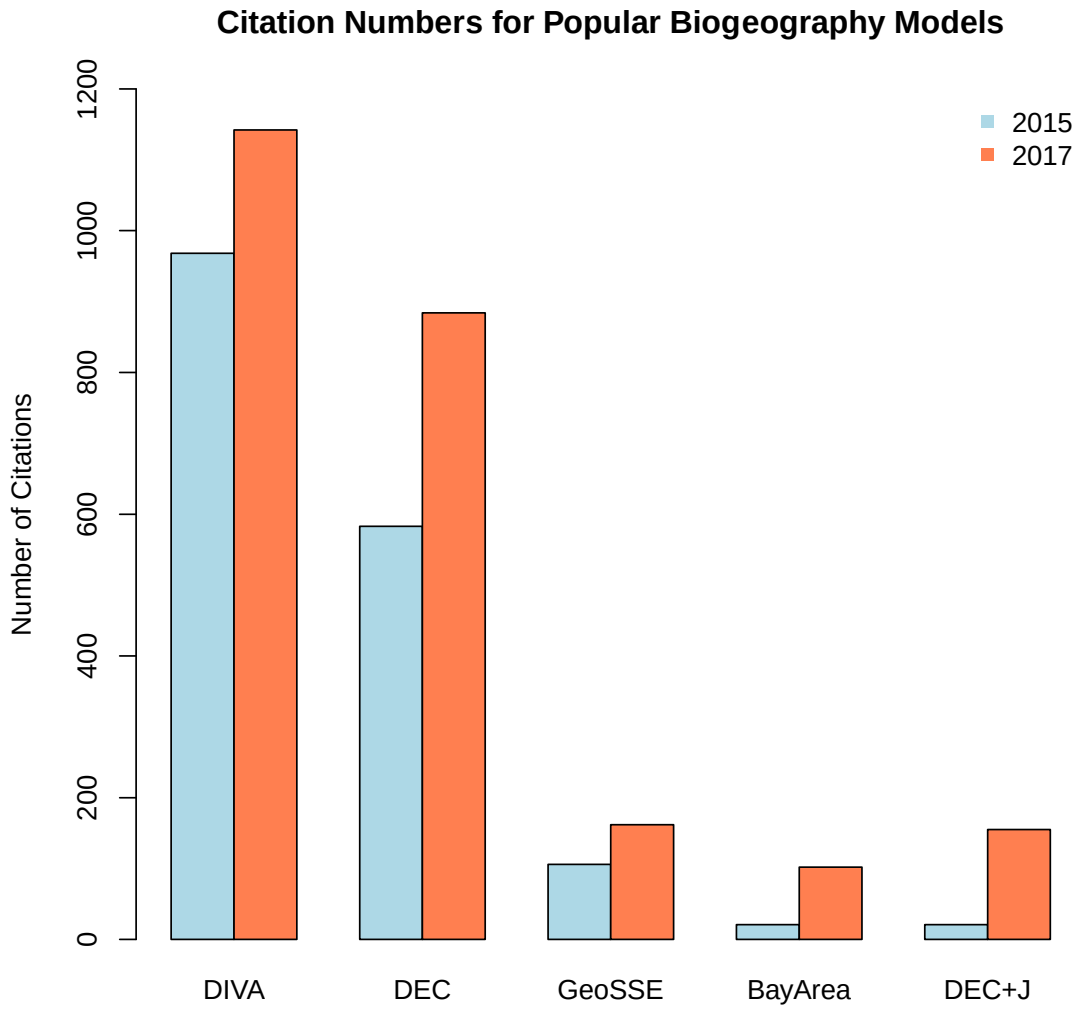

Figure 1.1. The number of citations for probabilistic models of historical biogeography in 2015 versus 2017. Data was acquired through a Google Scholar search where the number of citations were recorded for the publication describing each method. The results show that the use of parametric methods has been on the rise for these last two years. This is evident since 2015 when I started working on my project to now, March 2017.

CHAPTER I
NON-NULL EFFECTS OF THE NULL RANGE IN BIOGEOGRAPHIC MODELS: EXPLORING PARAMETER ESTIMATION IN THE DEC MODEL

A version of this chapter was originally submitted as an invited paper to *Systematic Biology*. It will be resubmitted to this journal pending revisions form reviewers. Currently it is available on the pre-print server, bioRxiv.

Massana, K. A., J. M. Beaulieu, N. J. Matzke, and B. C. O'Meara. 2015. Non-null Effects of the Null Range in Biogeographic Models: Exploring Parameter Estimation in the DEC Model. bioRxiv.

My primary contributions to this chapter are: (i) collection and analysis of the data and simulations, (ii) design of tables and figures, (iii) completion of the manuscript draft for submission, and (iv) response to reviewers for publication. The DEC* method is available through my github account found at https://github.com/kmassana/lagrange.

Abstract

Historical biogeography seeks to understand the distribution of biodiversity in space and time. The dispersal-extinction-cladogenesis (DEC) model, a likelihood-based model of geographic range evolution, is widely used in assessing the biogeography of clades. Robust inference of dispersal and local extinction parameters is crucial for biogeographic inference, and yet a major caveat to its use is that the DEC model severely underestimates local extinction. We suggest that this is mainly due to the way in which the model is constructed to allow observed species to transition into being present in no areas (i.e., null range). By prohibiting transitions into the null range in the transition rate matrix, we were able to better infer local extinction and support this with simulations. This modified model, DEC*, has higher model fit and model adequacy than DEC, suggesting this modification should be considered for DEC and other models of geographic range evolution.

Background

Historical biogeography has developed from simply observing the general patterns of species, to incorporating events that explain biogeographic processes (such as vicariance and dispersal), to developing explicit probabilistic approaches. With the advent of parametric methods based on maximum likelihood and Bayesian frameworks, researchers have been able to incorporate important information, such as branch lengths and fossils (e.g., allowing for better tree dating estimates) (Smith and Donoghue 2010; Wood et al. 2012; Beaulieu et al. 2013a; Pyron 2014).

A popular method (cited 884 times since its publication) to assess the historical biogeography of taxa is the dispersal-extinction-cladogenesis (DEC) model (Ree and Smith 2008), which estimates geographic range evolution for anagenetic (i.e., along branches) and cladogenetic (i.e., at nodes) change on a phylogeny. In the case of anagenetic change, range expansion and range contraction [modeled as parameters by way of the rate of dispersal from area *i* to area *j* (D_{ij}) and local extinction in area *i* (E_i)] are modeled as stochastic processes along branches. Published analyses with DEC assume the simplest model involving only a single dispersal rate and a single local extinction event, although it is possible to have more complex models (i.e., allowing different dispersal rates among each area pair by having n^2-n free parameters for the dispersal rates, and n free parameters for local extinction). A rate matrix can be assembled for a given number of geographic ranges and rate parameters (Figure 2.1).

Ree and Smith (2008) carried out simulations to test the accuracy of the DEC model on parameter estimation. They found that although the model worked reasonably well, dispersal was underestimated and local extinction was severely underestimated, often estimated as being effectively zero. Note that while there has been a robust discussion of whether extinction rates

can be estimated on molecular phylogenies (Nee et al. 1994; Rabosky 2010; Beaulieu and O'Meara 2015), the extinction rate in these cases relates to speciation and extinction of entire species and its signal on a phylogeny's topology and branch lengths. The extinction rate relevant for our purposes is the rate of having a population no longer occurring in a particular area, using a fixed tree. In terms of its role in a model, it is more like the rate of reduction in a meristic character than a rate at which a species goes extinct, even though biologically it appears more similar to the latter. Thus, its difficulty in being estimated is surprising.

One feature of the DEC model that has received little comment is that it includes a null range (a geographic range of 0 areas) in the anagenesis transition matrix (Figure 2. 2). In one sense, inclusion of the null range is a natural modeling decision, since the assumption that local extirpation is a process directly implies that the same process can reduce a single area geographic range to a range of size 0. However, the inclusion of a null range in the state space has some peculiar properties. For instance, no sampled species will ever occupy the null range state; even extinct species, if included in an analysis, are included because they occurred in some area. We suspect that the only way to fit any data pattern that does not observe null ranges is by driving down the rate of range contraction to the point where the probability of such an event is effectively zero. Unfortunately, given that all transitions to other extinction scenarios are linked through a global extinction parameter it seems unavoidable that when null ranges are allowed in the model extinction would generally be underestimated.

Of course, in some ways, the extinction rate is a nuisance parameter – that is, the hundreds of studies using the DEC model primarily focus on ancestral state estimates rather than on rates. However, given that this rate represents one of the two free parameters that are then used for inferring ancestral states on the tree, we expect that biased extinction estimates may

result in errors in the ancestral state estimation. In other words, given low extinction rate estimates, areas can only be lost at speciation events, so we predict a greater number of areas as we move rootward on the tree and few area losses along longer branches. This study attempts to modify the DEC model to improve estimation of extinction rate and then tests using simulated and empirical data to see if this results in a better model overall.

Methods

We modified the DEC model to omit transitions into the null state in the anagenesis transition rate matrix between ancestor and descendent pairs; we refer to this modified model as DEC* (Figure 2.1). It has the same number of parameters as DEC (dispersal and local extinction, *d* and *e* respectively), with the only change being fixing the transition rate to 0 for transitions from ranges of size 1 to the null range. DEC* is distinct from the three-parameter DEC+J model which allows for founder-event speciation associated with lineage-splitting with the addition of the free *j* parameter (Matzke 2014b). DEC+J retains the DEC assumption that a null geographic range is a valid state. To implement the DEC* model, we modified the original lagrange DEC C++ code (https://github.com/rhr/lagrange-cpp) to omit the transition into the null range in the anagenesis transition rate matrix. The DEC* model is implemented as a modification to the DEC C++ version, and is also allowed in the BioGeoBEARS R package (Matzke 2013) by changing the *include_null_range* setting from TRUE to FALSE in the BioGeoBEARS model setup.

We implemented our own DEC simulator in R that follows the procedures described by Ree and Smith (2008). The simulator produces birth-death phylogenetic trees with concurrent range evolution, combining the DEC model and stochastic cladogenesis. The simulator also does the same for the DEC* model. Trees were produced with the same known dispersal and local extinction parameter, constrained to vary between 0.01 and 0.2, while speciation was constrained

to be 0.4 events per million years. Geography consisted of three possible hard-coded geographic areas, meaning that there were 8 possible geographic ranges in the state space of the DEC simulation (A, B, C, AB, AC, BC, ABC, and null), and 7 possible ranges in the DEC* simulation (as the null range state is excluded). At cladogenesis, when the lineage had a widespread range, equal probabilities were assigned to each allowed range-inheritance scenario (vicariance or subset sympatry). For both DEC and DEC*, we performed 2,000 simulation-inference runs and compared dispersal and local extinction parameter estimates as well as the number of correctly inferred number of areas at internal nodes for all simulations. The simulations began by assigning the root node a range of a random single geographic area. The phylogeny was allowed to grow according to the DEC or the DEC* model until it reached 100 taxa (extant plus extinct). To match empirical datasets, the simulated phylogenies were pruned of branches that went extinct.

Our main objective was to understand DEC* versus DEC analyses on empirical datasets. Therefore, we searched the literature for published studies that used the DEC model. Then we compiled the phylogenies and geography presence/absence data available, which resulted in 15 empirical datasets. Most of these datasets were used in Matzke (2014b), and followed any modifications made therein (Kambysellis et al. 1995; Baldwin and Sanderson 1998; Hormiga et al. 2003; Jordan et al. 2003; Clark et al. 2008; Dunbar-Co 2008; Ree and Smith 2008; Benavides et al. 2009; Clark et al. 2009; Gillespie and Baldwin 2010; Smith and Donoghue 2010; Lerner et al. 2011; Nicholson et al. 2012; Bennett et al. 2013; Lapoint et al. 2013; Matzke 2014a). However, we also assessed the caecilian and salamander datasets from a recent published study comparing DEC and DEC+J (Pyron 2014), and a palpimanoid spider dataset (Wood et al. 2012) that only used DEC in the analysis. We performed unconstrained analyses with C++ DEC and

DEC* on each dataset. We compared analyses between DEC, DEC*, and DEC+J for all 15 datasets. If the values were not available, we used the package BioGeoBEARS (Matzke 2013) to run all DEC+J analyses. We also assessed model adequacy for each dataset by comparing the number of areas estimated per node between DEC and DEC* to the observed modern geographic range sizes.

Results

Simulations

Overall, the point estimate for local extinction was closer to the true value under DEC* than with DEC (Figure 2.4), although with higher variance. With simulations under the DEC model we found that the median for local extinction under a DEC* inference (*e*=0.0957) was closer to the true local extinction median estimate (*e*=0.0989), while the median for local extinction under DEC inference was close to zero (*e*=1.287e-06). Similarly, with simulations under the DEC* model we found that the median local extinction estimated under DEC* inference (*e*=0.1028) is almost identical to the true local extinction parameter (*e*=0.1030), whereas, again, local extinction is grossly underestimated under the DEC inference (*e*=1.200e-6).

Median estimates of dispersal under DEC simulations were closer to the median of the true dispersal parameter (*d*=0.0989) under DEC* than under DEC inference (*d*=0.0766) (Figure 2.4). When simulating under DEC* (Figure 2.5), the median dispersal under DEC* inference (*d=0.*1053) was again closer to the median dispersal rate used in the simulation (*d*=0.1030) than dispersal inferred under DEC (*d*=0.0789).

We calculated the root mean square error (RMSE) of the estimated parameter values for DEC and DEC*. The root mean square error gives the standard deviation associated with the differences between the true parameter and the inferred parameter estimates, and here a smaller

value indicates less error in the parameter inference. Results indicated that on the logarithmic scale the error for *e* was far better for DEC* than DEC and nearly the same for *d* (RMSE of *e* was 10.9120 for DEC and 3.8363 for DEC*; RMSE of *d* was 0.3784 for DEC and 0.3886 for DEC*). However, on a linear scale, error is far better for both parameters for DEC than DEC*, due to some tremendously high values of *e*. (RMSE of *e* was 0.1135 for DEC and 2.2345 for DEC*; RMSE of *d* was 0.0363 for DEC and 0.7816 for DEC*).

Finally, we assessed the accuracy of DEC against DEC* in estimating the geographic area range at the root. Under DEC simulation, the root state was correctly estimated 49.05% of the time, whereas under DEC*, 58.30% of root states were accurately estimated.

Empirical Datasets

In comparisons of DEC and DEC* on empirical datasets, likelihood was always better under DEC*. Thus, AIC always selected DEC* as the better model over DEC (as the two models have the same number of parameters). In 10 out of 15 empirical datasets, AIC selected DEC* over DEC+J (Figure 2.6). DEC+J has an extra parameter relative to DEC*, so if likelihoods were equal between DEC+J and DEC*, DEC* would be preferred by AIC. However, in all but one of the cases where AIC preferred DEC* over DEC+J, the likelihood was itself better with DEC* (which is possible, as the two models are not nested). The exception was *Psychotria*, where AIC gives DEC* 50.2% of the model weight despite slightly higher likelihood for DEC+J (model weight 20.4%).

Unlike the simulated data, for over half the empirical datasets the extinction rate inferred by DEC was substantially higher than zero, ranging from 16% to 546% of the estimated value of the dispersal rate. For DEC*, the extinction rates were even higher relative to dispersal: only for one empirical dataset was the extinction rate indistinguishable from zero, for the rest the

extinction rate was between 3.2 and 1389-fold higher than dispersal rate (median 104-fold higher). In some cases, the estimated extinction rate was at the maximum allowed by C++ DEC and DEC*; modifying it to increase the bound by tenfold improved the likelihood by a median of 0.061 log likelihood units and increased the extinction estimate up to the new maximum in most cases (Figure 2.6). The small magnitude of improvement, with the large magnitude of change in the estimate, suggests that the likelihood surface is very flat but that the unconstrained maximum likelihood estimate would be even higher. More simply put, for these datasets, the best estimate of extinction is extremely high, which would mean that after a species expands its range it nearly instantly contracts it (into either the new region or back to the old region). In only three of nine of these datasets was DEC+J chosen over DEC*, despite the apparent evidence for a jump-like dispersal model.

Model Adequacy

In addition to model choice, a key question to examine with new models is model adequacy: how well does the model fit overall? Even the best-fitting model may not do a good job predicting the data, which would point to the need for new models to better match reality. This has been increasingly emphasized in phylogenetics (Goldman 1993; Bollback 2002; O'Meara 2012; Beaulieu et al. 2013b; Pennell et al. 2015). To see if the DEC* model adequately describes the data, we counted the number of occupied areas estimated for each node and compared this between DEC and DEC* for each empirical dataset. We work under the assumption that the present should look like the past: a clade of island endemics is more likely to have been island endemics for much of their history, rather than being composed of very widespread species that only at the present suddenly became endemic to single islands. Of course, there are processes that could make the present not resemble the past (i.e., a sudden

change in climate causing suitable habitat to be divided into isolated patches), but this assumption should hold in most groups. For all but two empirical datasets, the DEC* model was the more adequate model, with estimated range sizes at ancestral nodes more closely matching the estimated mean range sizes observed at the tips of the phylogeny (Figure 2.5 and Figure 2.7). Inference under DEC usually yields ancestral distributions that are very widespread, which is not the case under DEC* (Figure 2.5 and Figure 2.8).

Discussion

Given the results, we argue that DEC* should be considered for use in biogeographic models. Testing models should be an intrinsic part of the research process, so most users should try DEC, DEC*, DEC+J, and future models, but if one were limited to just one model, in most cases DEC* would be preferred, based on the empirical results presented here. DEC* does a more adequate job at estimating ancestral ranges than does the canonical DEC model. However, while the median extinction and dispersal parameters were better estimated under DEC* than with DEC, the RMSE of the estimates on a linear scale was better under DEC, and DEC* often returns very high estimates for extinction rate. For estimating ancestral areas, DEC* is probably the better model, but we urge extreme caution when treating its rate parameters as parameters of interest rather than nuisance parameters. Of course, ancestral states are known to be difficult to estimate well (Cunningham 1999; Oakley and Cunningham 2000), so biologists should expect a great deal of uncertainty with estimates. We also note that the estimates of uncertainty in this model are always underestimates, due to other uncertainty (topology, branch lengths, states) that is typically not accounted for.

Another caveat to the use of DEC*, which also applies to DEC and DEC+J, is its treatment of the phylogeny: it assumes range evolves on a tree but that biogeography does not

directly influence speciation or extinction. Speciation often seems influenced by geographic context (Mayr 1963), such as through the divergence of two isolated populations. While DEC, DEC*, and DEC+J allow subdivision of ranges at speciation events, these models do not, for example, fit a higher speciation rate to species with larger ranges. There are models that jointly fit the diversification process and process of biogeographic evolution, such as GeoSSE (Goldberg et al. 2011) and ClaSSE (Goldberg and Igić 2012). However, even though these models are more realistic, they can require rather large data sets (Davis et al. 2013) and are feasible only for very few areas. The empirical datasets used in biogeographic models are typically small in comparison; the ones used in this study had an average of 75 taxa and 5.53 areas from all datasets, and a range of 4 to 10 areas and 9 to 469 taxa. Recent work (Matzke 2014b) showed that DEC vs. DEC+J model choice appears robust to some commonly postulated SSE processes (speciation and extinction depending on range size), and that ancestral range estimation is reasonably accurate if model choice is performed and if the dispersal rate is low, suggesting that for datasets that limit the power of SSE models, the DEC* model can still be used, with caution.

It is important to emphasize that the DEC* model is still relatively simple. Though some complexity can be incorporated with different dispersal rates between areas or at different time points, all species are treated as having the same rates of dispersal and extinction at a given time. We know, however, that species in a clade may vary in traits affecting successful dispersal (ability to inbreed, resting stages, wind versus animal dispersal, tolerance of saltwater, and so forth) or extinction (body size, trophic level, thermal tolerance, and so forth) and this variation is not yet incorporated in any of these models. There are additional sources of heterogeneity that also may result in misleading results if not incorporated.

The very high extinction estimates by DEC*, especially in empirical datasets, were unexpected. One partial explanation may come from the empirical distribution of range sizes in the empirical studies presented here, where the vast majority of species were found in just one area. Under DEC* (or DEC) the only way for a species to change its area is through expansion to a new area (i.e., dispersal), followed by other events. Given that all the studies have species in different areas (there is little point to inferring biogeographic history for a clade that only ever occupies one area), there must be a nonzero dispersal rate. Lineages can reduce range size in two ways: at cladogenesis, or through range contraction along branches. However, when *e* is high with respect to *d* and the speciation rate, lineages will spend almost no time in widespread ranges. Therefore, widespread ranges are essentially never available at cladogenesis, and all speciation will be sympatric. Moreover, on terminal branches, which represent over half the branches on the tree, any necessary dispersals cannot be "undone" by contraction at cladogenesis, and so the only way to have some dispersals along terminal branches, but have observed species in just one area each, is to have a substantial extinction rate. DEC* with high *e* is a model with all range change effectively occurring in anagenetic "jumps" along the branches: any expansion is followed almost immediately by a contraction. The fact that DEC* often outperforms DEC+J probably indicates that in these cases, the probability of sister taxa living in different areas correlates better with the branch length between the taxa than with the number of speciation events recorded in the observed tree. We expect a DEC*+J model may incorporate the best of both of these models, and that adding "*" to other models may also be beneficial.

The major use of parametric models in historical biogeography is for ancestral state estimation. For model adequacy, we expect that the present tends to resemble the past, so a model where past distributions are similar to present ones is probably a better fit to the data. For

empirical datasets used in biogeography, tip taxa are most often in one area. But, DEC often estimates ancestors as being in many areas, because the DEC model allows a transition into the null range, and since null ranges are not observed, the inference is pushed towards a low extinction rate. In contrast, the DEC* model returns estimates at internal nodes that usually resemble the number of areas present in the tips (see Figure 2.5). DEC* may return nearly equally likely single areas rather than a more confident estimation of the ancestral state being a union of areas. In many cases, especially given observed species that individually occupy few areas, this uncertainty about which single area an ancestor occupied represents reality. However, even though uncertainty exists in ancestral range estimates, statistical model choice is a fruitful way to assess models against the data.

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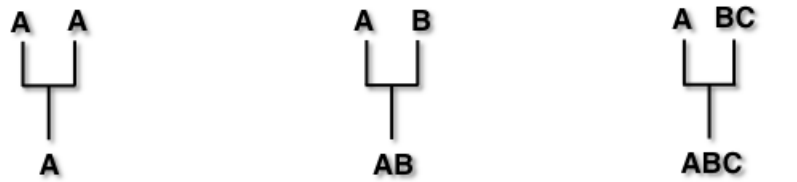
Appendix B

a) Fixed Cladogenesis



b) DEC Anagenesis

	Ø	A	B	C	AB	AC	BC	ABC
Ø	–	0	0	0	0	0	0	0
A	E_A	–	0	0	D_{AB}	D_{AC}	0	0
B	E_B	0	–	0	D_{BA}	0	D_{BC}	0
C	E_C	0	0	–	0	D_{CA}	D_{CB}	0
AB	0	E_B	E_A	0	–	0	0	$D_{AC} + D_{BC}$
AC	0	E_C	0	E_A	0	–	0	$D_{AB} + D_{CB}$
BC	0	0	E_C	E_B	0	0	–	$D_{BA} + D_{CA}$
ABC	0	0	0	0	E_C	E_B	E_A	–

c) DEC* Anagenesis

	Ø	A	B	C	AB	AC	BC	ABC
Ø	–	0	0	0	0	0	0	0
A	0	–	0	0	D_{AB}	D_{AC}	0	0
B	0	0	–	0	D_{BA}	0	D_{BC}	0
C	0	0	0	–	0	D_{CA}	D_{CB}	0
AB	0	E_B	E_A	0	–	0	0	$D_{AC} + D_{BC}$
AC	0	E_C	0	E_A	0	–	0	$D_{AB} + D_{CB}$
BC	0	0	E_C	E_B	0	0	–	$D_{BA} + D_{CA}$
ABC	0	0	0	0	E_C	E_B	E_A	–

Figure 2.1. Diagram showing (a) the allowed cladogenetic events for DEC and DEC*, (b) the anagenesis transition rate matrix for DEC, and (c) the DEC* anagenesis transition rate matrix, assuming three geographic states (A, B, C). At a cladogenetic event, if a species is in one area, the descendant species inherits that area. If the species is in multiple areas, one species inherits one area, while the other species inherits all areas (peripatric speciation; within area widespread) or it is allowed to inherit all areas but the area occupied by the first species (vicariant allopatric speciation; between area widespread). Note that extinction is not allowed in DEC* from one state to zero states. D = dispersal, E = local extinction.

	Ø	A / A	B / B	C / C	A / B	B / A	A / C	C / A	B / C	C / B	AB / A	A / AB	AB / B	B / AB	AC / A	A / AC	AC / C	C / AC	BC / B	B / BC	BC / C	C / BC	BC / A	A / BC	AC / B	B / AC	AB / C	C / AB	ABC / A	A / ABC	ABC / B	B / ABC	ABC / C	C / ABC
Ø	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
A	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
B	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
C	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
AB	0	0	0	0	1/6	1/6	0	0	0	0	1/6	1/6	1/6	1/6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
AC	0	0	0	0	0	0	1/6	1/6	0	0	0	0	0	0	1/6	1/6	1/6	1/6	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
BC	0	0	0	0	0	0	0	0	1/6	1/6	0	0	0	0	0	0	0	0	1/6	1/6	1/6	1/6	0	0	0	0	0	0	0	0	0	0	0	0
ABC	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1/12	1/12	1/12	1/12	1/12	1/12

Figure 2.2. DEC and DEC* cladogenesis transition rate matrix from ancestors (leftmost column) to descendants (first row) with the respective transition probabilities under the fixed events.

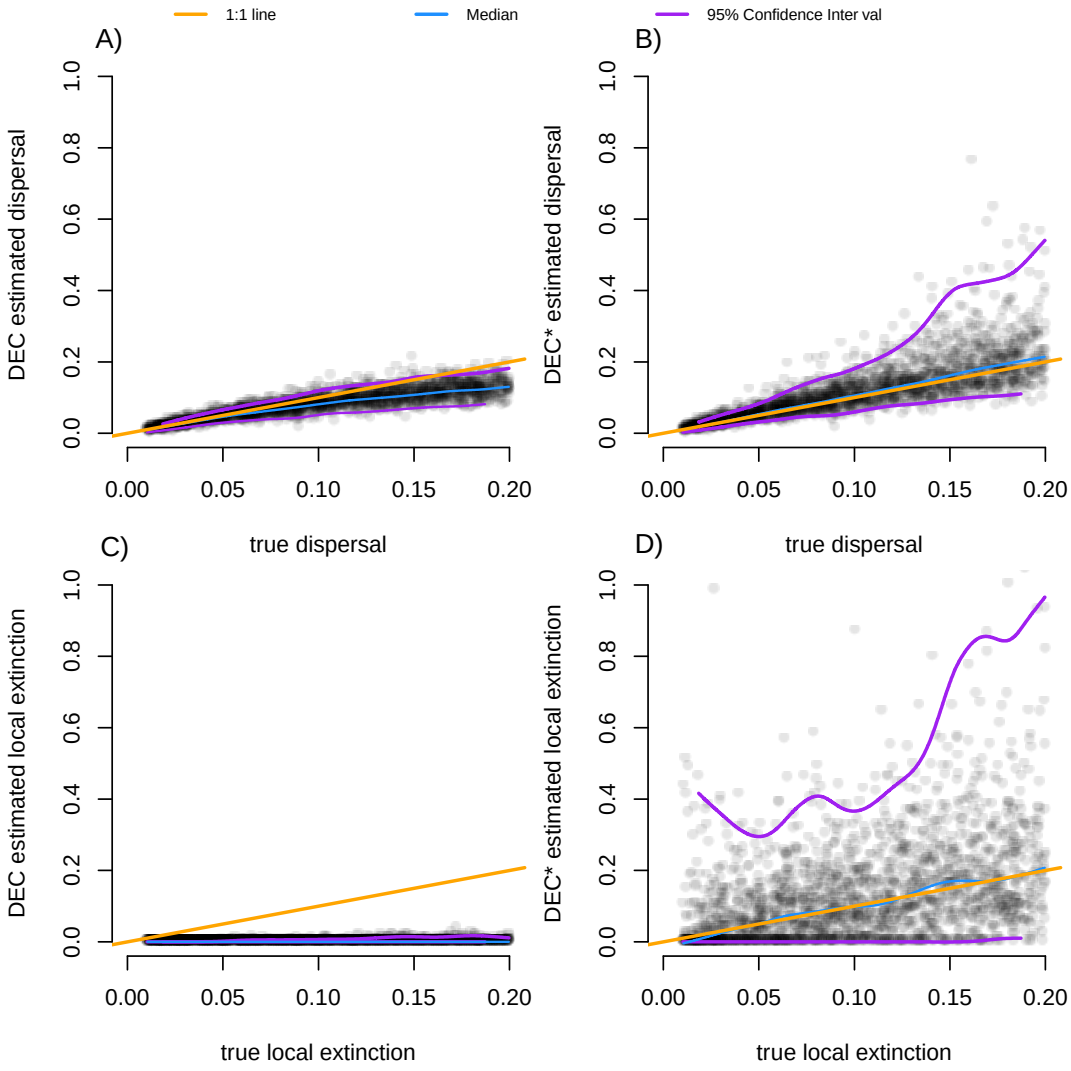


Figure 2.3. Plots showing parameter inference under the 2,000 DEC simulations. DEC inference of dispersal (A) was not as effective as dispersal inferred under DEC* (B). Local extinction under DEC inference (C) was highly underestimated. Local extinction under DEC* (D) was better estimated in comparison to DEC inference, although with more variance. Purple lines represent 95% confidence intervals; blue line shows the median; orange line shows the 1:1 line.

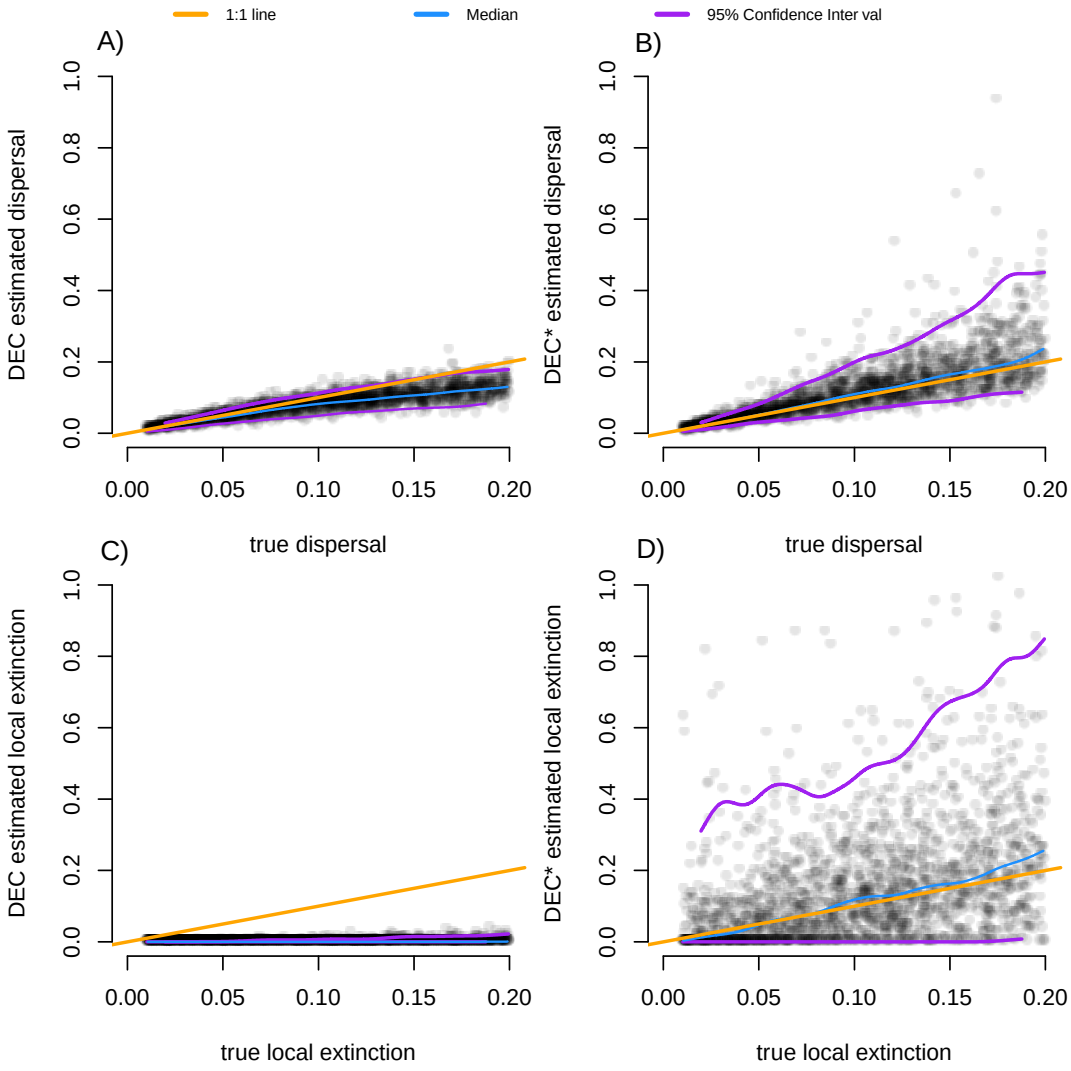


Figure 2.4. Plots showing parameter inference under the 2,000 DEC* simulations. DEC inference of dispersal (A) was not as effective as dispersal inferred under DEC* (B), but dispersal inferred under DEC* seems to have more variance (B). Local extinction under DEC inference (C) was highly underestimated. Local extinction under DEC* (D) was better estimated in comparison to DEC inference, although with more variance. Purple lines represent 95% confidence intervals; blue line shows the median; orange line shows the 1:1 line.

Figure 2.5. Model adequacy plots based on the number of areas occupied at nodes for the empirical datasets of the Galapagan *Microlophus* (A), Hawaiian *Plantago* (B), Pacific *Cyrtandra* (C) reconstructed with DEC or DEC* versus the tips which represent the current number of areas for each group. In each empirical case (A, B, C), DEC* was able to ancestrally infer the same number of areas as the tips in the current range. DEC inferred ranges that were more widespread. The last plot depicts the mean number of current areas at tips versus the mean number of areas estimated at nodes through DEC or DEC* for each group (D). In each case except for two, DEC* was able to estimate the ancestors to occupy the same number of areas as the tips.

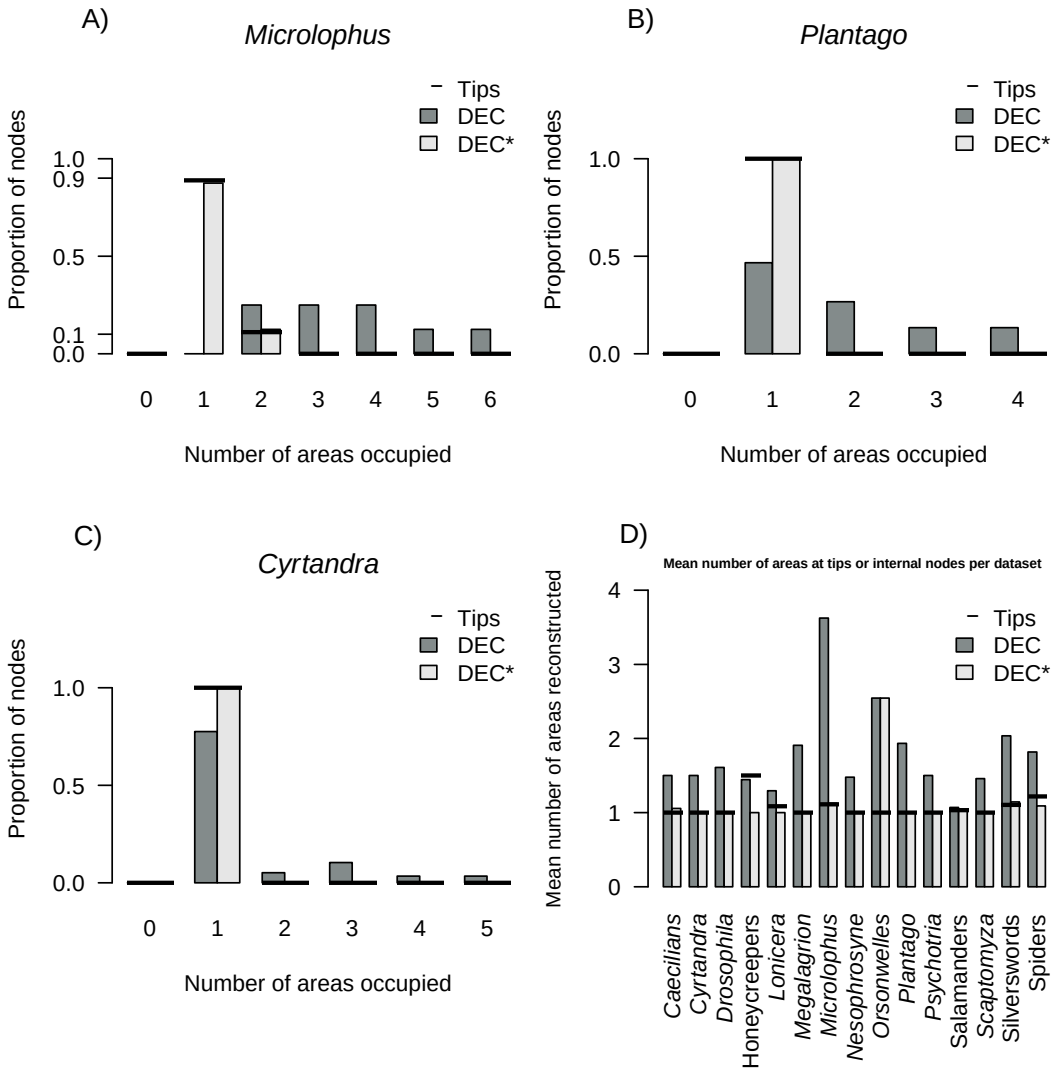


Figure 2.5. Continued.

Clade	DEC Δ AIC	DEC* Δ AIC	DEC+J Δ AIC	DEC* dispersal	DEC c++ dispersal	DEC* extinction	DEC c++ extinction
Caecilians	7.524	0	25.948	0.001	0	0.104	0
Cyrtandra	10.032	0	4.107	0.008	0.001	100	0
Drosophila	75.657	0	2.535	0.105	0.018	100	0.021
Honeycreepers	27.061	0	4.907	1.159	0.173	3.655	0.163
Lonicera	43.234	0	7.98	0.013	0.006	0.223	0.001
Megalogrion	59.018	0	8.88	0.424	0.067	100	0.055
Microlophus	27.494	0	3.898	2.145	0.078	100	0.426
Nesophrosyne	229.792	27.8	0	0.19	0.048	100	0.024
Orsonwelles	3.454	3.427	0	0	0	0	0
Plantago	15.28	0	2.44	0.105	0.013	100	0
Psychotria	25.3004	0	0.0164	0.178747	0.0350117	100	0.0282904
Salamanders	24.21	0	28.532	0.001	0	0.036	0
Scaptomyza	92.026	3.456	0	0.072	0.017	100	0.031
Silversword	8.777	0	6.148	0.236	0.041	2.669	0
Spiders	13.351	7.262	0	0.001	0.001	0.014	0

Figure 2.6. Results of comparisons of DEC and DEC* on 15 empirical datasets. AIC shows DEC* as a better model over DEC. In two-thirds of the datasets, AIC selected DEC* as a better model over DEC+J. Extinction rates inferred by both DEC and DEC* were high relative to dispersal, although DEC* inferred rates to be much higher, and in some cases the extinction rate was at the maximum allowed by the program. Modifying to increase the bound by tenfold improved the likelihood by a small fraction and increased the extinction estimate up to the new maximum in most cases.

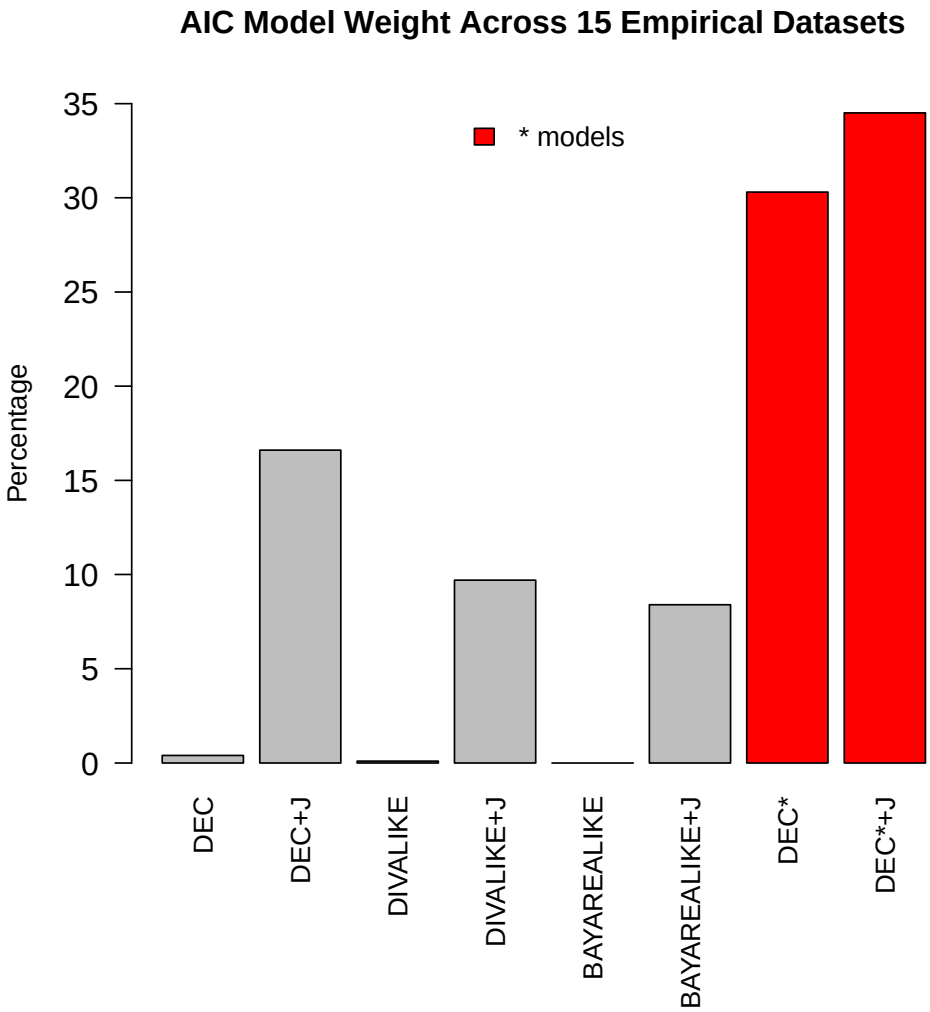


Figure 2.7. The AIC model weight across the 15 empirical datasets. The "*" models encompass most of the AIC weight (over 75%). Whereas, the other models, cumulatively, fall below.

Figure 2.8. Most probable number of biogeographic areas estimated in the Plantago clade under the DEC (A) versus the DEC* model (B). The number of areas at tips is also shown. The estimated ancestral range probabilities under DEC versus DEC* for node 8 (C) and node 10 (D) are shown. Ancestral state estimates under the DEC model are more widespread than under DEC*, therefore providing a different biogeographic history (C and D). Nodes closer to the root provide more variance in the probable ranges estimated (D).

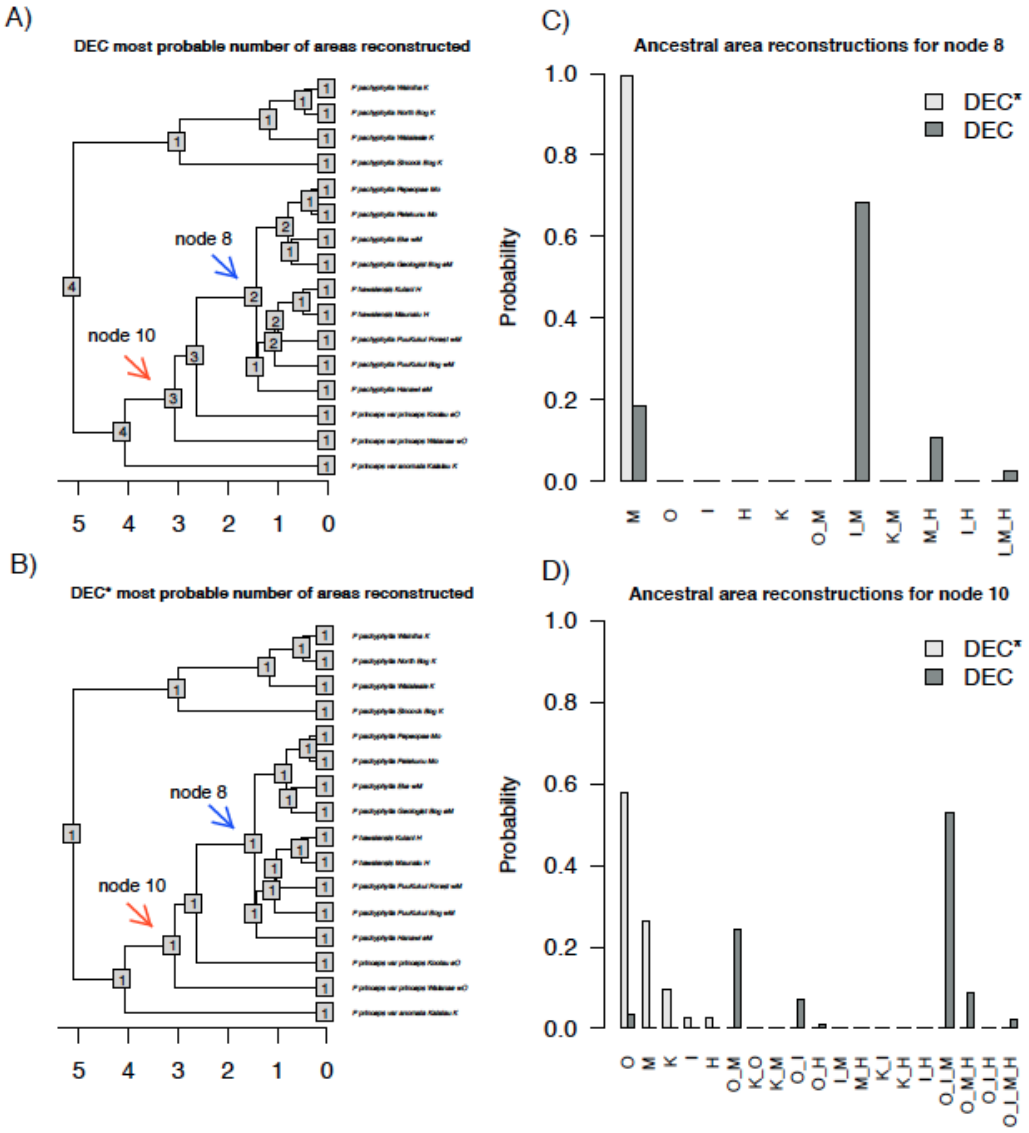


Figure 2.8. Continued.

CHAPTER II
THE DIVERSIFICATION OF PARASITIC ANGIOSPERMS

This chapter is intended to be submitted as a manuscript to *Science*.

I collaborated on this project with Anouk van't Padje while she was a Master's student at Harvard University and with Dr. Chuck Davis, a Professor at Harvard University. My primary contributions to this chapter include, (i) experimental design, (ii) collection and analysis of data, (iii) table and figure development, (iv) written completion of the manuscript draft for submission.

Abstract

Parasitic angiosperms have been hypothesized to undergo an evolutionary dead end. Accompanying an evolutionary dead end is a lower diversification rate (speciation – extinction) ascribed to lower speciation rates and higher extinction rates due to highly specialized behavior and structures. In addition, many species have small effective population sizes and high reliance on hosts. In this study, we used a wide array of diversification methods to test whether several lineages of parasitic angiosperms are indeed undergoing an evolutionary dead end. We find that the mode of parasitism (i.e. hemiparasitic, holoparasitic, endoparasitic) affects the number of species, that older clades tend to have more species, and that standard diversification models show each clade has different diversification processes. Trait-dependent methods show that parasitism is not an evolutionary dead end for flowering plant parasites and that there is another underling trait or process affecting the diversification of these species.

Background

Angiosperms that have lost or reduced photosynthetic ability either directly parasitize fungi (mycoheterotrophs) or plants via a specialized feeding structure called a haustorium (parasitic plants). There are 12-13 different origins of parasitism in flowering plants (Barkman et al. 2007). Mycoheterotrophy, on the other hand, has independently evolved many more times. In eudicots, it has evolved 7 times accounting for approximately 46 species (1 in Polygalaceae, 2 or 3 in Ericaceae and 4 in Gentiaceae). In monocots, 43 lineages have evolved mycoheterotrophy with at least 411 species, most of which are in Orchidaceae. At least one species in each of the following groups has evolved mycohetrotrophy: ferns, lycophytes and gymnosperms. Mycoheterotrophy is not known to have evolved in mosses and liverworts. Parasitic plants and mycoheterotrophs are widely distributed around the globe and exist in many different biomes, ranging from the tropical rainforests to the tundra (Merckx and Freudenstein 2010).

Plant parasites have a great diversity in morphology, anatomy and physiology (Kuijt 1969; Nickrent and Musselman 2004; Thorogood and Hiscock 2010). However, their morphology in many cases is reduced. Most studies in the literature support an evolutionary dead hypothesis of decreased diversification and high extinction (Cope 1904; Gould 1970; Futuyma and Moreno 1988), due to many plant parasites having highly specialized evolutionary structures and mechanisms. Unlike parasitism in animals, there are no known reversals from parasitism in angiosperms. Depending on host specificity, dispersal agents and other factors, parasitic plants and mycoheterotrophs have variable roles in community assemblies, which have fluctuating implications on food web interactions and community structure. Both parasitic plants and mycoheterotrophs have moved up the food chain, changing from producers to primary consumers. This escalation in the food chain could offer advantages and disadvantages, but how

does it affect their diversification? Small effective population sizes (characteristic of many parasitic plants) and reliance on hosts increases extinction in these plants, which lowers diversification and leads to parasitism in flowering plants as an evolutionary dead-end. However, other factors such as increased mutation rates and faster generation times compared to hosts (Bromham et al. 2013), as well as genetic drift, may confound this hypothesis, especially depending on the clade of angiosperm parasites.

Parasites increase connectedness and complexity in food webs (Hudson et al. 2006) and are involved in altering the physiology and anatomy of their hosts, which could in turn affect the influence hosts have on an ecological community. Host populations are affected by parasites: parasites affect host mortality, fitness, growth, nutritional uptake and energetic requirements. Such effects could also alter rates of predation and trophic cascades (Lafferty et al. 2006). The following is a broadly significant example: parasites in the genus *Striga* attack important agricultural crops in the Poaceae family, this in turn affects the plant crops involved and severely affects growth, increases mortality and decreases fitness. Understanding diversification of flowering plant parasites would help us understand more about parasitism as a trait.

Hardy and Cook (2012) suggest that the difference between the parasitic clades and their autotrophic sisters is due to differences in ecological limitations and/or exponential diversification. We seek to test the latter in this study, using sister group comparisons and diversification models.

Methods

Data Classification

In this study, we were interested in understanding how diversification rates compared between parasitic clades and their non-parasitic sister groups. We searched the literature to find the main parasitic plant clades and their closest non-parasitic relatives and included mycoheterotrophs (Musselman et al. 1995; Nickrent 1997; Janssen and Bremer 2004; Merckx et al. 2006; Goldblatt et al. 2008; Cameron 2009; Bell et al. 2010; Merckx and Freudenstein 2010; Chacón et al. 2012; Hardy and Cook 2012; Merckx 2012; Bellot and Renner 2013; List 2013; Mennes et al. 2013; Schwery et al. 2015). We classified these into sister clade comparisons.

Net Diversification Rates from Magallón and Sanderson (2001)

Magallón and Sanderson (2001) proposed a method to assess lineage diversification with species richness. We estimated the net diversification rates as a function of the accepted number of species per parasitic family using List (2013) and the crown group clade ages (Table 1.1). We calculated the expected number of species per age in the R package geiger (Harmon et al. 2008). We set the extinction rates to be low (extinction = 0) and high (extinction = 0.9) to estimate the rates in order to create a 95% confidence interval for species numbers with high and low extinction. Using the intervals estimated from the package, we could then test whether the number of species for each clade, given the age, was significant. We used the expected diversification rates from Magallón and Sanderson (2001) to calculate the expected rates based on all angiosperms. The rates we used for high and low extinction were $r_{e=0}$= 0.0893 and $r_{e=0.9}$= 0.0767.

We plotted the different modes of parasitism (hemiparasitism versus holoparasitism, and endoparasitism versus exoparasitism) and the site at which the parasite attached (stem versus root attachment) to assess diversification for the mode of parasitism.

Phylogenetic Generalized Least Squares

To check for a relationship between the trait state of interest and diversification, we performed a phylogenetic generalized least squares regression (PGLS) (Freckleton et al. 2002) for each parasitic clade. We used the List (2013) to record the number of species and the crown ages were extracted from the dated phylogeny of Zanne et al. (2014). Using the package caper in R (Orme 2013), we calculated the phylogenetic signal, the correlations between the stem and crown ages, the number of species and the diversification with the rate of the whole angiosperm family.

Sister Clade Comparisons

We used several sister group comparison tests: the binomial sign test, the classical sister group comparison by Slowinski and Guyer (1993) and the advocated sister groups by Käfer and Mousset (2014) to understand the diversification between each sister clade comparison. The binomial sign test tests whether the observed number of groups with lower diversification rates can be explained due to random chance. The advocated sister groups comparisons corrects the clade age for the stem branch length.

Phylogenetic Inference and Dating

We identified the overall clade that contained the parasitic plant clade and the sister group clade. We used the software PHLAWD (Smith et al. 2009) to download the sequences from GenBank (Buckner et al. 2014) for 17 genes (18S, 26S, atpB, matK, matR, ndhF, petG,

psbA, rpl16, rpl20, rps4, rps16, trnK, trnL, trnSG, ITS and rbcL). To align the sequences, we use the software MAFFT (Katoh et al. 2002). Phyutility and Mesquite were both used to clean sequences. To effectively clean the data for missing data, we removed taxa that had a sequence length shorter than half the length of the longest sequence in Mesquite (Maddison and Maddison 2001). The program Mesquite was also used to remove hybrid taxa and species only at the genus level. We then used the program phyutility to clean, align and concatenate the sequences (Smith and Dunn 2008). We created partition files so each gene in the concatenation could evolve under a different rate of molecular evolution under Maximum Likelihood for tree inference. We used RaxML-HPC BlackBox (Stamatakis 2006) on the CIPRES Science Gateway (Miller et al. 2010) on the concatenated alignment with a partition/mixed model to allow for differing rates among the genes under the recommended GTR + Γ model by Stamatakis (2006), which would also print the corresponding branch lengths. All tree dating was done with TreePL (Smith and O'Meara 2012) using the dating times from Bell et al. (2010). TreePL implements the penalized likelihood dating method described by Sanderson (2002). We identified the partial decisiveness scores with Decisivator (Zhbannikov et al. 2013) to assess how robust the trees were. We then pruned trees to parasitic clade and sister clade lineages. For the analysis with the phylogenies made with PHLAWD, there were a small proportion of states when the parasitic state was divided into more specific parasitic forms (i.e. hemiparasite, holoparasite, endoparasite, exoparasite) compared to the number of non-parasitic taxa, than recommended by Davis et al. (2013). Therefore, the trait-dependent analyses were restricted to using the parasitic versus non-parasitic binary state.

Lineage Through Time Plots and Diversification Analysis

Diversification analyses were carried out using the R package laser for pure birth, birth death, logistic dependent, exponential dependent, variable speciation, variable extinction and

variable speciation and extinction diversification models (Rabosky 2006). We also visually looked at the Lineage Through Time plots of the clade to understand the diversification process (Harvey et al. 1994; Stadler 2008). Incomplete lineage sampling can skew results to appear as having an early burst, followed by a decreased diversification. The Rafflesiaceae clade was the only clade with complete lineage sampling, therefore, we assessed the effect of incomplete lineage sampling on the diversification tests for each clade by calculating the gamma statistic and using the MCCR test in the package laser.

Joint BiSSE Inference

We implemented a joint BiSSE method to test trait-dependent diversification based on the method proposed in Maddison et al. (2007). In this approach, we linked the six different parameters estimated through the BiSSE model for all parasitic and non-parasitic sister clades jointly. We did this to overcome several challenges identified with the BiSSE model. Since some of the parasitic clades are small in species numbers, the trait ratios between the parasitic state and non-parasitic state are small for some clades, which has been identified as an issue by Davis et al. (2013). We also know that only studying diversification of one independent origin per clade biases inference because there are many independent origins of parasitism in angiosperms and this could potentially bias the results. For an in depth discussion, please see Maddison and FitzJohn (2014). We used the BiSSE model function from the diversitree R package (FitzJohn 2012) and developed our own code to jointly infer the BiSSE model across all the clades of interest. We tested an unconstrained model (all parameters free for all clades), a model where transitions were equal across each clade being compared ($q_{01} = q_{01}$), a model where extinction was the same across each clade being compared ($\mu_0 = \mu_0$ and $\mu_1 = \mu_1$) and a model where all parameters were equal across clade comparisons ($\lambda_0 = \lambda_0$; $\lambda_1 = \lambda_1$; $\mu_0 = \mu_0$; $\mu_1 = \mu_1$; $q_{01} = q_{01}$). We

did not allow for transition from parasitic to non-parasitic, since this is not something that has been biologically observed ($q_{10} \sim 0$).

HiSSE

We also used HiSSE (Beaulieu and O'Meara 2016) to test trait-dependent diversification through the hisse R package (Beaulieu et al. 2016). The advantage of the HiSSE method is that it allows for a hidden trait with a binary state to affect the diversification of a clade, so the rate heterogeneity inferred is not necessarily attributed to the specific trait being tested. We set up 24 different models including a fully unconstrained model and 22 different constrained HiSSE models where the parameters were fixed in different ways, as well as a BiSSE model. We corrected for incomplete sampling in all lineages since all of the lineages were missing species, except for Rafflesiaceae.

Results

The mode of parasitism affects the number of species

Parasitic plant taxa have differing results concerning tests of absolute diversification (Figure 3.1). Orobanchaceae, *Cuscuta* and Kramericeae fall above, Apodanthaceae, Cynomoriaceae, Cytanaceae, Hydnoraceae, Lennoaceae, Mitrastemonaceae and Rafflesiaceae fall below, and Santalales and *Cassytha* fall within the 95% confidence interval of the Magallón and Sanderson (2001) method. This shows that all endoparasitic plant families have a lower than average expected number of species (p-value: 0.0195), while hemiparasities generally have a higher number of species than the holoparasitic lineages. The type of parasitism, whether root or stem, and the place of attachment do not affect the number of species for each family. Using the PGLS approach, there was a significant association only with the crown age and the number of

species, showing that the older clades tend to have more species than the younger parasitic clades (Table 1.2).

Phylogenetic inference

We inferred 17 phylogenetic trees, most were of larger clades based on the Order that comprised the parasitic and non-parasitic sister lineages. This was carried out to help with dating of the trees and to ensure the most species mined from Genbank. The partial decisiveness scores from the inferred clades ranged from 0.521 (Lamiales) and 0.928 (Zygophyllales). Please refer to Table 1.3 for all partial decisiveness scores, the number of species in each clade's phylogeny and the number of characters in the sequence alignment. We determined that there were only five sister clade comparisons (Table 1.4) where enough species were represented in each clade for the trait-dependent diversification analyses: Caryophyllales + **Santalales**, Solanales + **Convolvulaceae**, Dioscoreaceae + **Burmanniaceae and Thismiaceae**, Euphorbiaceae + **Rafflesiaceae** and Hypoxidaceae + **Orchidaceae and Iridaceae**. The parasitic clades are bolded. All of these clades have partial decisiveness values that scored over 0.7, suggesting the sequence data is decisive for over 70% of the possible trees, except for Caryophyllales.

LTT plots and standard diversification models show varying diversification processes

The results from the LTT plots (Figures 3.2 - 3.6) and the diversification models show different results among the parasitic clades and their sister clades. Below, we sectioned the results based on the sister clade comparisons. All parameters are summarized in Table 1.5. The results from the MCCR test show non-significant findings, suggesting that even with incomplete lineage sampling, this diversification results are robust.

Dioscoreaceae versus Burmanniaceae + Thismiaceae

For the Burmanniaceae + Thismiaceae clade, the best model was the exponential density dependent model (Table 1.6) with an initial speciation parameter estimated as 0.472 and an *x* parameter of 0.878. For the Dioscoreaceae clade, the best model was the variable speciation model (Table 1.7) with an initial speciation parameter estimate of 0.371, a *k* parameter estimate of 0.001 and an extinction parameter of 0.337. The variable speciation is evident in the LTT plot (Figure 3.2) with an initial high speciation around ~85 million years ago, and then the speciation decreased, with a high speciation rate again around 15 million years ago. The initial speciation parameter is higher for the Burmanniaceae + Thismiaceae parasitic clade, however the Dioscoreaceae clade has a greater number of species. The diversification process between the clades is very different.

Solanaceae versus Convolvulaceae

For the Solanaceae clade, the best model was the variable extinction and speciation model (Table 1.8) with an initial speciation rate of 0.313, a *k* parameter of 0.001, an extinction estimate of 0.289 and a *z* parameter estimate of 0.110. For the Convolvulaceae clade, the best model was the exponential density dependent model (Table 1.9) with an initial speciation rate of 0.284 and a *x* parameter estimate of 0.468. The initial speciation rate is nearly the same for both clades, only slightly higher for the non-parasitic Solanaceae clade. The Solanaceae clade has a greater number of species, indicating a higher diversification rate.

Caryophyllales versus Santalales

The best model for Caryophyllales clade is the variable speciation model (Table 1.10) with an initial speciation rate of 0.273, a *k* parameter of 0.003 and an extinction parameter of 0.192. For the Santalales clade, the pure birth model (Table 1.11) is the best model with an initial

speciation rate of 0.043. The LTT plots look very similar for both these clades, with higher speciation around 100 million years ago, after which speciation is still increasing but at a lower rate. The Caryophyllales clade has a higher initial speciation rate than the Santalales clade. The Caryophyllales clade has more species, yet the diversification process for these clades is very similar.

Euphorbiaceae versus Rafflesiaceae

The best model for the Euphorbiaceae clade is the birth death model (Table 1.12) with an initial speciation rate of 0.091 and a death rate of 0.799. The best model for the Rafflesiaceae clade is also the birth death model (Table 1.13) with an initial speciation rate of 3.42E-08 and a death rate of 0.999. For Rafflesiaceae, the clade seems to have diversified around 20 million years ago and Euphorbiaceae around 40 million years ago. However, from the results, it is clear that the speciation rate for Rafflesiaceae is much lower than for Euphorbiaceae and the extinction rate is high for both clades, but much higher in Rafflesiaceae. It is clear from the LTT plots that the Euphorbiaceae clade has a much higher diversification of species.

Hypoxidaceae versus Orchidaceae + Iridaceae

The best model for the Hypoxidaceae clade is the pure birth model (Table 1.14) with a birth rate of 0.058. The best model for the Orchidaceae + Iridaceae clade is the birth death model (Table 1.15) with an initial speciation rate of 0.065 and a death rate of 0.736. The speciation is similar in both of these clades; however, the number of species in the Orchidaceae + Iridaceae clade is much higher.

Joint BiSSE analyses do not support an evolutionary dead end for all parasitic angiosperms

The best model from the BiSSE joint modeling framework is the full model with all state and transition rates unconstrained (Table 1.16). The parameter estimates under this model can be found in Tables 2.17 and 2.18. The net diversification rate for the non-parasitic trait is greater in the Caryophyllales + Santalales clade by ~3.3 times, the Solanales + Convolvulaceae clade by ~4.6 times, and Hypoxidaceae + Orchidaceae and Iridaceae by ~3.3 times. The net diversification is higher for the parasitic trait in the Dioscoreaceae + Burmanniaceae + Thismiaceae clade by ~14.5 times and for the Euphorbiaceae + Rafflesiaceae clade by 1.6 times. In the Caryophyllales + Santalales clade, for approximately every 340 speciation events in non-parasitic plants, there is a transition from non-parasitic to parasitic. In the Solanales and Convolvulaceae clade, for approximately every 1,431 speciation events in non-parasitic plants, there is a transition from non-parasitic to parasitic. In the Dioscoreaceae + Burmanniaceae and Thismiaceae clade for about 194 speciation events, there is a transition from non-parasitic to parasitic. Having the slowest rate, in the Euphorbiaceae + Rafflesiaceae there are about 1,756 speciation events, before there is a transition from non-parasitic to parasitic. With the fastest rate of transition to a parasitic state, the Hypoxidaceae and Orchidaceae + Iridaceae, there are about 132 speciation events, before there is a transition from non-parasitic to parasitic. These results show that for three out of the five clades (Caryophyllales + Santalales, Solanales + Convolvulaceae, Hypoxidaceae + Orchidaceae and Iridaceae), the net diversification for the non-parasitic trait is higher. This gives support that the all clades of parasitic plants do not have a lower diversification over the non-parasites. For the parameter estimations for the rest of the models tested, please see Tables 1.19 - 1.21.

HiSSE results show a hidden trait influences the diversification of parasitism for all sister clade comparisons

The model fit results for the given sister clade comparisons using the HiSSE models can be found in Table 1.22-1.26. The parameters estimated given the best model for each sister clade comparison can be found in Tables 1.27-1.32.

Dioscoreaceae versus Burmanniaceae + Thismiaceae

The best model for the Dioscoreaceae + Burmanniaceae and Thismiaceae sister clade comparison is the full HiSSE model with transition rates and states unconstrained (AIC model weight 99.92%). The net diversification rate for non-parasitic trait with hidden trait state B is 72.7 times greater than for the non-parasitic trait with state A. The parasitic trait with state B is 8.262e+19 times greater than for the parasitic trait with state A. The net diversification rate for the non-parasitic trait with state A is 4.060e+18 times greater than for the parasitic state with state A. For the non-parasitic trait with state B the net diversification rate is 3.572 times greater than for the parasitic trait with state B. The angiosperms with the parasitic state and state A have the lowest turnover and the highest extinction fraction. The results show support for the parasitic trait influencing diversification to be low for plants in these sister clades, but suggest another possible trait and/or biological process is contributing to the diversification outcomes we see here.

Solanaceae versus Convolvulaceae

The best model for this clade comparison was one where there was no parasitic trait with a hidden trait with state B, therefore not allowing for this as a possibility (AIC model weight of 96.17%). This suggests that only one state of a hidden trait influences the diversification of the parasitic trait, but both binary states of the hidden trait affect the non-parasites. The highest net diversification is for the non-parasitic plants with the hidden trait with state B. These also had the

highest turnover and lowest extinction fraction. Comparing the hidden trait with state A, the parasitic species have a 14.11 times greater diversification, 12% of the turnover and a 2.250e-06 lower extinction fraction. This suggests that parasitic species in this clade comparison have differing diversification in comparison to their autotrophic counterparts depending on the influence of the hidden trait. We also see no transition into the parasitic state with hidden state B, which could imply that the trait of interest or biological process could be tied to photosynthesis or photosynthetic potential. Since parasitic species in the Convolvulaceae clade are mostly holoparasites (lack the ability to photosynthesize), and there is very little evidence on species having the presence of chlorophyll, it seems like the ability to photosynthesize could be a good candidate for the hidden trait.

Caryophyllales versus Santalales

The best model for the Caryophyllales and Santalales sister clades is the full HiSSE model with transition rates and states unconstrained (AIC model weight 99.99%). The net diversification rate for non-parasitic trait with hidden trait state B is ~ 4.9 times greater than for the non-parasitic trait with state A. For the parasitic trait with state B, the net diversification is 277.975 times greater than for the non-parasitic trait with state B. The net diversification rate for non-parasitic trait with hidden trait state A is 6.758 times greater than for the non-parasitic trait with state B. The parasitic trait with state B is 201.214 times greater than for the parasitic trait with state A. The angiosperms with the non-parasitic state and state B have the lowest turnover and the highest extinction fraction. The results show mixed support for diversification of parasitic plants in these clades. A hidden trait also influences the diversification and depending on the state of the hidden trait, there is either lower diversification compared to non-parasites (state A) or higher diversification (state B). There is significant evidence to suggest another

possible trait and/or biological process is contributing to the diversification outcomes we see here, and the influence is more complex than a direct influence upon diversification.

Euphorbiaceae versus Rafflesiaceae

The best model for the Euphorbiaceae and Rafflesiaceae sister clade comparisons is a model where state B is fixed for both the non-parasitic and parasitic traits (0B = 1B, AIC model weight 35.71%). Although a model with the state A fixed has a delta AIC of 0.03611059 and 35.07% of the AIC model weight, which is a small difference in comparison to the best model. A model with both hidden states to be fixed has a delta AIC of 0.813196305 (which is also a small difference at 23.78% of the AIC model weight), supporting this conclusion. The net diversification for the hidden trait state B is higher than for the hidden trait state A regardless of whether the plant is a parasite or not (10.02061 times greater than parasitic plants and 649.2792 times greater than non-parasites). The turnover is much greater for the hidden state B. The extinction fraction is about the same regardless of the traits. The results suggest that either hidden state B or state A should be fixed. Therefore, there is evidence for the hidden trait with both states A and B and that the parasitic and non-parasitic traits influence one of those hidden traits. It is also possible that only the hidden trait matters. However, the relationship of the influence of parasitism is not clear on the diversification of these clades. Improving the sampling of the Euphorbiaceae clade may or may not help disentangle this story.

Hypoxidaceae versus Orchidaceae + Iridaceae

The final sister comparison, Orchidaceae + Iridaceae versus Hypoxidaceae had the best model where the hidden trait with state B was fixed (AIC model weight 100%). Whether a species is parasitic or not has no influence on the state B of the hidden trait in this model. Therefore, it seems that parasitism only influences one of the states of this hidden trait. The net

diversification and turnover was higher for the hidden state with state B regardless of whether the species was parasitic or not with the hidden strait with state A.

Discussion

Is parasitism an evolutionary dead end?

It is clear from the results that the process of diversification in parasitic angiosperms is different depending on the clade. We postulate that different diversification processes are likely due to the remarkable variation among parasites, which display different modes of parasitism, specialization, species dependence, and range, which could all be influencing this phenomenon. We showed evidence with the Magallón and Sanderson (2001) method that the mode of parasitism influenced the diversification rate of parasitic lineages: hemiparasities having a higher diversification over endoparasities. Our results, therefore, show support that parasitism alone is not causing an evolutionary dead end in angiosperms, but the extreme specialization to a host, for example, might be able to cause this. Therefore, parasitic lineages such as ones with endoparasites, may be under an evolutionary dead end, but the general umbrella trait of parasitism is not. We do not state with certainty that the endoparasitic trait causes lower diversification because we had mixed results from the analyses. The diversification models and the LTT plots showed Rafflesiaceae having a lower diversification and a higher extinction than Euphorbiaceae, yet the HiSSE and the BiSSE analyses showed that the parasitic state influenced a slightly higher diversification. Unfortunately, we currently do not have the data at this large-scale to explicitly test whether mode of parasitism or other species-dependent traits could be affecting the diversification of parasitic angiosperms. Further sequencing of individual parasitic angiosperms will need to be carried out to infer better phylogenies of these for future comparative work.

Hardy and Cook (2012) showed results similar to ours, in the case of Santalales, Orobanchaceae, *Cassytha* (Lauraceae) and *Cuscuta* (Convolvulaceae) having higher or equal diversification rates that their sister group counterparts. From our results, generally, there is a lower diversification rate for the parasitic lineages, but not in all comparisons. For the BiSSE joint analyses, we see three parasitic clades have lower diversification in comparison to their non-parasitic sister clade counterparts, except for Orchidaceae + Iridaceae which has an increased diversification in comparison to Hypoxidaceae, and Rafflesiaceae which has an increased diversification in comparison to Euphorbiaceae. The HiSSE analyses support a lower diversification for the parasitic trait in the Convolvulaceae, Santalales and Burmanniaceae and Thismiaceae. This is most likely due to the mode of parasitism or the photosynthetic potential. As we stated previously, there was not enough representation of traits for mode of parasitism to test the influence with trait-dependent analyses. Cumulatively, our results go against the widely held hypothesis that parasitism in angiosperms is an evolutionary dead end. Looking at parasitic plants for all angiosperms using the Zanne et al. (2014) phylogeny supported the evolutionary dead end hypothesis (data not shown here), but our individual clade comparisons show that the diversification has a unique story for each clade.

The degree of parasitism appears to very influential in the diversification of parasitic angiosperms. Hemiparasitic angiosperms that still have the capacity of photosynthesizing seem to have a higher diversification rates than the holoparasites and endoparasities, which do not have the ability of photosynthesizing anymore and live within the host itself. Endoparsitism is the most derived form of parasitism in angiosperms and is considered an adaptive peak (Barkman et al. 2007). This degree of parasitism has its benefits and costs, on the one hand the parasite is completely protected form predators and outside threats, at the cost of being

completely dependent on its host for survival (Combes 2001). If the host is abundant and the parasite has little threat of extinction, then this might be a good strategy, otherwise the endoparasitic would face the threat of extinction. Once an organism is so specialized, it is difficult, if not impossible, to revert back to a generalized state, not to mention gaining the ability to evolve the ability of photosynthesis. Therefore, this could lead to an evolutionary dead end for taxa with this mode of parasitism (Cruickshank and Paterson 2006).

Unfortunately, we were not able to compare all parasitic lineages with their non-parasitic sister counterparts. This was mostly due to many parasitic lineages having very few species in each origin (4 species in Lennoaceae, 6 in Ericaceae, 1 in Cassytha, 4 in Cytanaceae, 2 in Petrosaviaceae and Hydnoraceae and 1 in Kramericaceae), which makes it hard to use phylogenetic methods to yield informative results.

Justification for methodology

The methods we used were carefully selected to test our hypothesis. We specifically chose not to use the BAMM (Rabosky et al. 2014) software due to recent debate in the literature about its flawed likelihood calculation (Moore et al. 2016). Davis et al. (2013) showed that phylogenies with less than 200 have very little statistical power in BiSSE analyses. They also show that in reality it is necessary to have over 300 taxa to be able to interpret and trust the results well. Another requirement of BiSSE is to have a binary trait ratio of at least 1:10, per clade of interest. Parasitic lineages usually have a small number of parasites, so we find the number of parasites is too low to have a 1:10 ratio of binary states. Comparing each parasitic clade independently is also problematic, since there is only one origin of parasitism in each parasitic clade. This makes it hard for the BiSSE model to estimate transition rates based on only one transition. To ameliorate these issues, we developed the joint BiSSE modeling framework.

We also considered using HiSSE because we wanted to test whether the diversification rates we inferred were due to parasitism. Since HiSSE allows for a hidden trait with a binary state to affect the diversification of a clade, we were able to test whether the influence of diversification was attributed to parasitism or a hidden trait. This was informative for our study, since we found that in each clade comparison an underlying trait influences their diversification.

Lack of data for parasitic angiosperms

The partial decisiveness scores show that there is need for more sampling of angiosperms to gather sequence data. Hinchliff and Smith (2014) discuss the limitations of GenBank data for the use of phylogenetic inference with PHLAWD (Smith et al. 2009). They state that only about one third of recognized land plants have sequence data available. The rate of adding sequences to GenBank has stayed about the same since the mid-1990s, and assuming this same rate of increase, it would take until the year 2044 to reach the minimum number of estimated living land plants (~300,000).

There were many more questions we wanted to answer in this study. Does host range influence diversification? Are there more generalist or specialist parasites and how does this influence the diversity of the parasitic lineages? Data such as pollinator type, host range, biogeography was not available in a large-scale capacity to do comparative analyses. Based on our results that another trait influences the diversification of parasitic angiosperms, we think host range may also be a good predictor of diversification in addition to mode of parasitism in gathering more information of influencing factors of parasite diversification. We hope future studies will focus on collecting more data of this type for parasitic angiosperms. We are aware that this is not an easy task, since there is a high possibility of ascertainment bias here – stem parasites are usually easily identified, but endoparasites and root parasites are not, which would

show a higher rate of stem parasites over ones that live underground. Generating lists of host species is also difficult, especially for very generalist parasites that have a wide ranges of hosts. Incorporating uncertainty or studying a smaller, well-sampled clade would be something that should be further pursued if an analysis of this type is considered. There would be higher confidence in species that are sampled often as well.

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Appendix C

Table 1.1. The parasitic lineages and their sister groups. Crown ages are extracted from Zanne et al. (2014) mega-phylogeny. The number of species corresponds with the number of accepted species documented in the Plant List. The mycoheterotrophs were not included in this study because of the few representative species present in the phylogeny.

Order	parasitic lineage	Number of species	Stem age	Crown age	Sister group	Number of species
Piperales	Hydnoraceae	10[a]	58[b]	48[b]	Aristolochiaceae[c]	480[a]
Laurales	*Cassytha*	21[a]	24[d]	24[d]	Lauraceae	134[a]
Malvales	Cytinaceae	11[a]	72[b]	48[d]	Muntingiaceae[e]	3[a]
Zygophyllales	Krameriaceae	25[a]	61[b]	12[f]	Zygophyllaceae	211[a]
Malpighiales	Rafflesiaceae	22[a]	95[g]	82[g]	Euphorbiaceae[h]	5735[a]
Cucurbitales	Apodanthaceae	27[a]	73[i]	60[i]	Coriaceae Corynocarpaceae[j]	2295[a]
Saxifragales	Cynomoriaceae	1[a]	100[b]	100[b]	Crassulaceae[k]	1482[a]
	Santales	3655	110[b]	105[l]	Caryophyllales	11155[a]
Ericales	Mitrastemonaceae	2[a]	103[m]	103[m]	Balsaminaceae, Marcgraviaceae Tetrameristaceae[n]	626[a]
Boraginales	Lennoaceae	4[a]	65[b]	41[b]	Ehretiaceae[o]	150[a]
Solanales	*Cuscuta*	171[a]	34[b]	22[d]	Convolvulaceae (excl. *Humbertia, Jacquemontia, Dichondra)*	1032[a]
Lamiales	Orobanchaceae (Ex. *Lindenbergia*)	1613[a]	42[d]	35[d]	*Lindenbergia*[n]	4[a]

[a] theplantlist.com, [b]Naumann *et al.* (2013),[c] Nickrent *et al.* (2002), [d]Zanne *et al.* (2014), [e]Nickrent (2007), [20]Renner and Schaefer (2010), [g]Bendiksby *et al.* (2010), [h]Davis *et al.* (2007), [i]Bellot and Renner (2014), [j]Filipowicz and Renner (2010), [k]Nickrent *et al.* (2005), [l]Anderson *et al.* (2005), [m] Bremer *et al.* (2004), [n]Hardy and Cook (2012)

Table 1.2. The results from the Phylogenetic Generalized Least Squares analysis.

Correlation	Intercept	P-value	Slope	p-value
Stem age – state	177.6	<2e-16	-0.3018	0.9423
Stem age- number of species	1.776+02	<2e-16	2.7005e-05	0.9255
Crown age – state	129.0	2.146e-12	4.547	0.3915
Crown age – number of species	1.289e+02	1.782e-12	2.181	0.0289*
State – number of species	5.569e-03	0.9727	-1.280e-06	0.7025
Diversification rate (stem age; ε = 0) – state	0.02685	0.9497	-0.009608	0.9215
Diversification rate (stem age; ε = 0.9) – state	0.01045	0.9453	-0.007369	0.8708
Diversification rate (crown age;ε= 0) – state	0.03176	0.6495	-0.003536	0.8650
Diversification rate (crown age; ε= 0.9) – state	0.01954	0.6977	-0.002671	0.8584

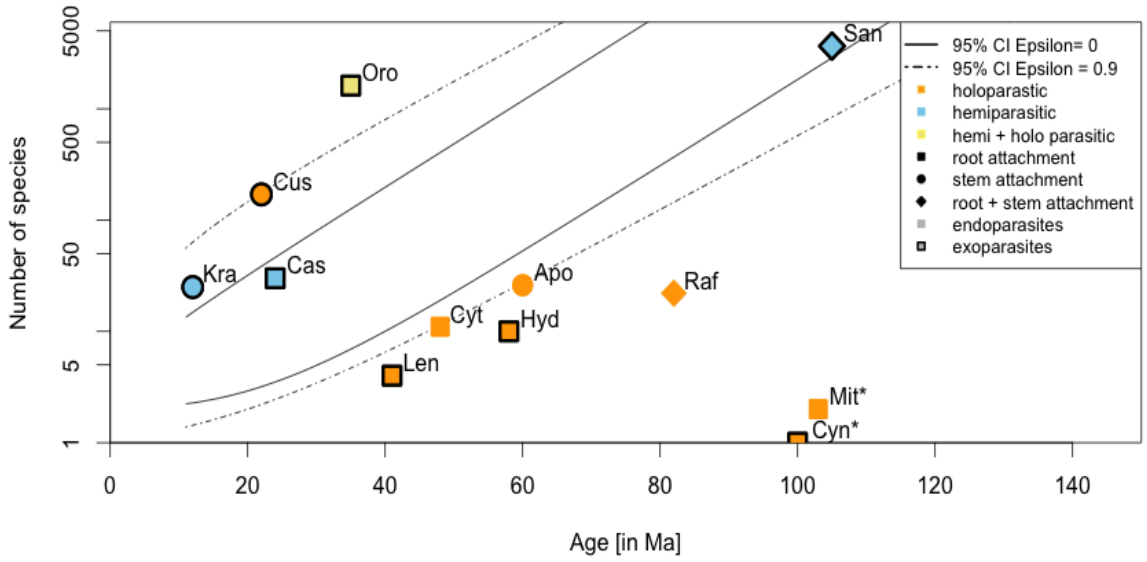

Figure 3.1. Net diversification of parasitic plant lineages; number of species plotted against the crown age. Apo: Apodanthaceae, Cas: *Cassytha*, Cus: *Cuscut*a, Cyn: Cynomoriaceae, Cyt: Cytinaceae, Hyd: Hydnoraceae, Kra: Krameriaceae, Len: Lennoaceae, Mit: Mitrastemonaceae, Oro: Orobanchaceae, San: Santalales.

Table 1.3. The number of species, number of characters in the sequence alignment and partial decisiveness scores for the overall angiosperm clades. The partial decisiveness scores range from Lamiales having the lowest score at 0.520734 and Zygophyllales having the highest score with 0.928333. There is a wide range of scores showing that some clades are better represented than others.

Clade	Number of species	Number of characters in alignment	Partial Decisiveness Score
Asparagales	7,178	14,369	0.714
Boraginaceae	779	15,122	0.610
Caryophyllales	3,246	12,068	0.544
Dioscoreales	323	11,874	0.741
Ericales	3,179	13,646	0.655
Fabales	4,649	12,690	0.613
Gentiales	4,320	14,411	0.560
Lamiales	4,882	14,528	0.521
Laurales	516	12,466	0.716
Liliales	889	10,688	0.618
Malpighiales	3,956	12,303	0.670
Malvales	1,375	13,178	0.572
Pandanales	209	10,818	0.675
Piperales	378	13,566	0.680
Santalales	294	10,140	0.738
Solanales	1,389	13,601	0.610
Zygophyllales	75	10,185	0.928

Table 1.4. The parasitic clades used for trait-dependent analyses. The rest of the parasitic and non-parasitic clades could not be used because they had too few species for phylogenetic comparison, either because this is a reality with Lennoaceae, or because there were too few sequences extracted from GenBank.

Order	**Parasitic Clade**	**Estimated Number of Species from the Plant List**	**Number of Species found from GenBank**	**Sister group**
Asparagales	Orchidaceae + Iridaceae	30116	3751	Hypoxidaceae
Dioscoreales	Burmanniaceae + Thismiaceae	226	62	Dioscoreaceae
Malpighiales	Rafflesiaceae	22	22	Euphorbiaceae
Solanales	*Cuscuta*	171	70	Solanaceae
Saxifragales	Santalales	3655	273	Caryophyllales

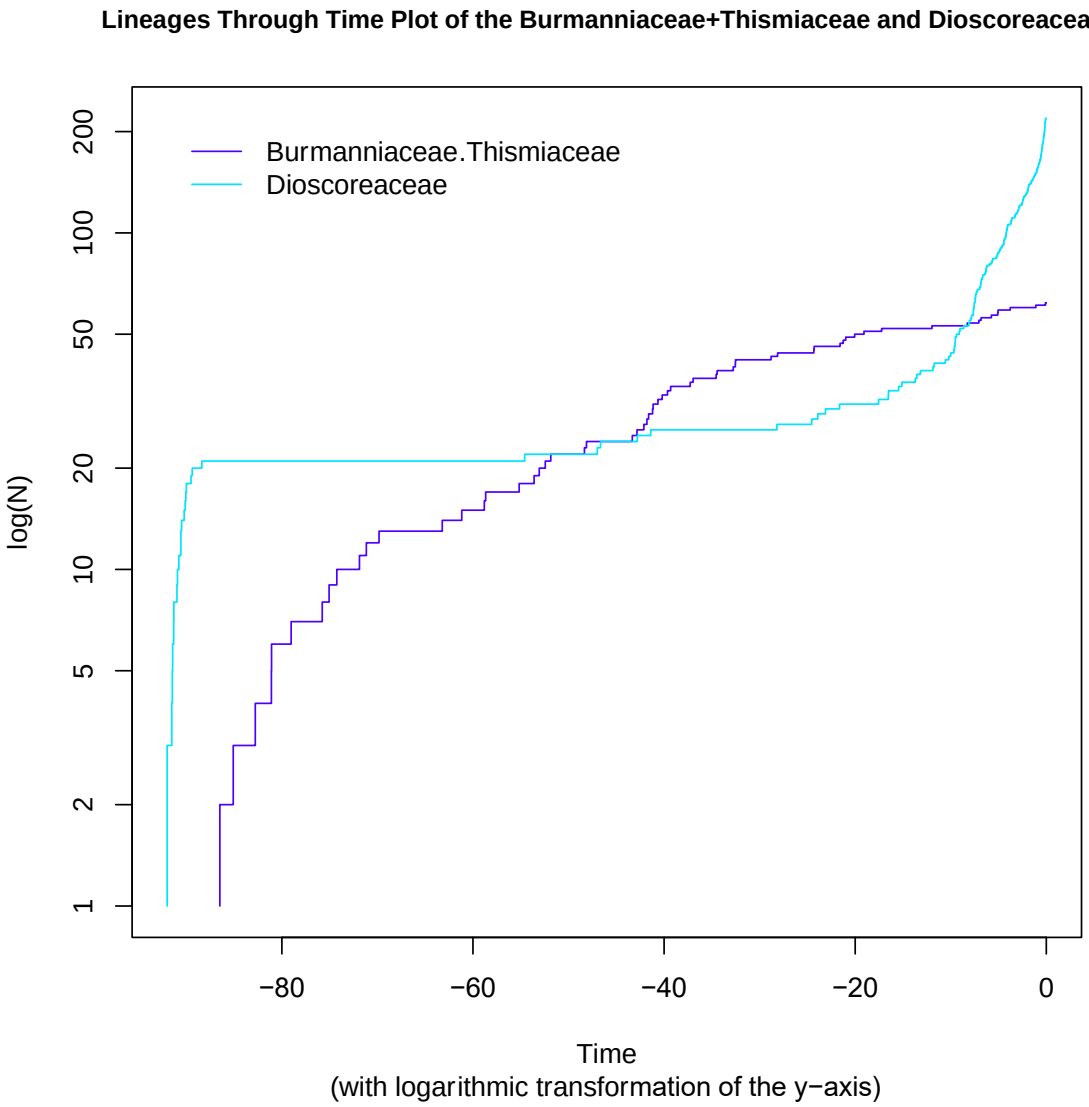

Figure 3.2. Lineage Through Time Plot for the parasitic clade Burmanniaceae + Thismiaceae (purple line) versus the non-parasitic sister clade Dioscoreaceae (blue line). The lines show the number of species in each lineage through their evolutionary history.

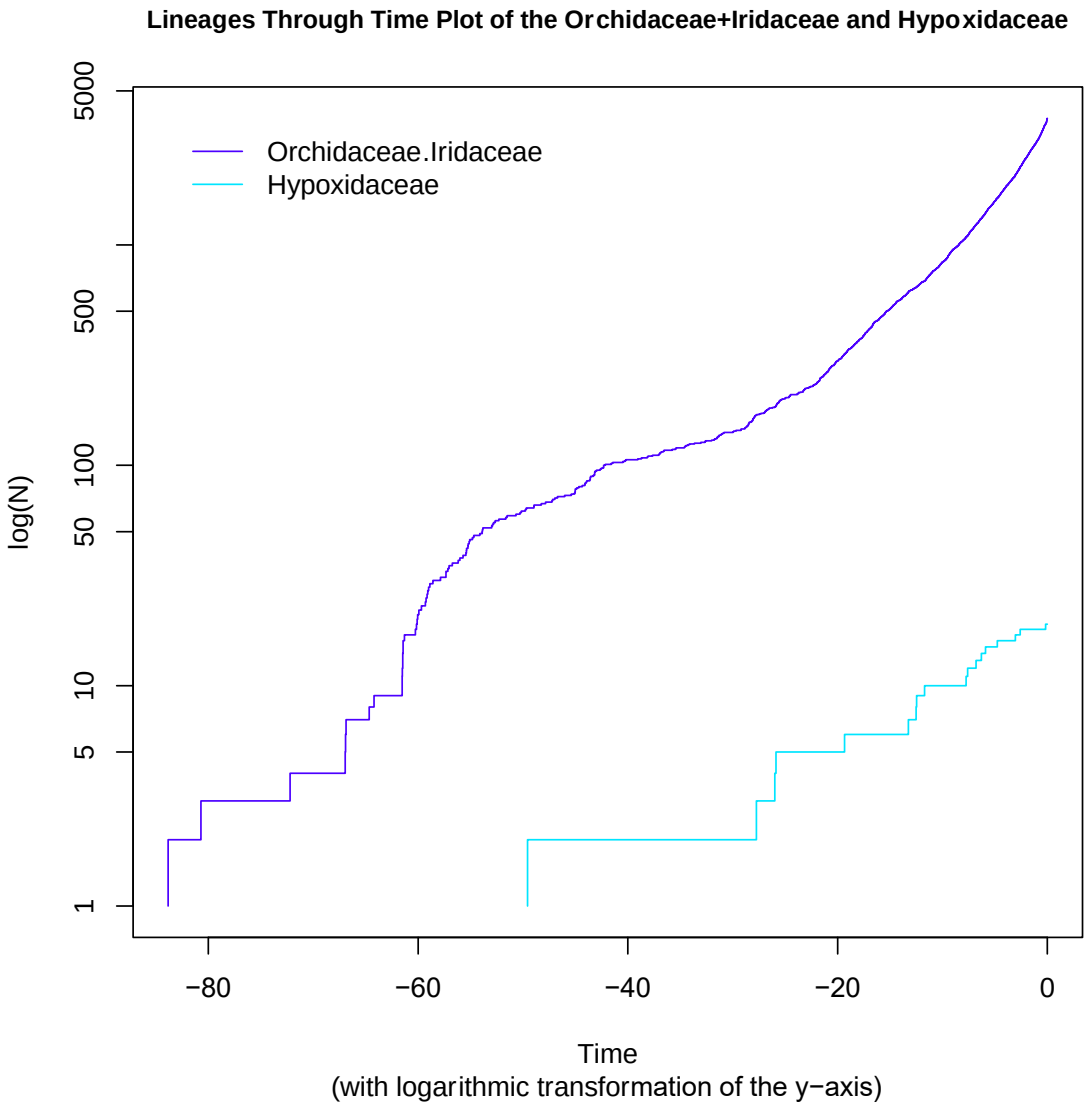

Figure 3.3. Lineage Through Time Plot for the parasitic clade Orchidaceae + Iridaceae (purple line) versus the non-parasitic sister clade Hypoxidaceae (blue line). The lines show the number of species in each lineage through their evolutionary history.

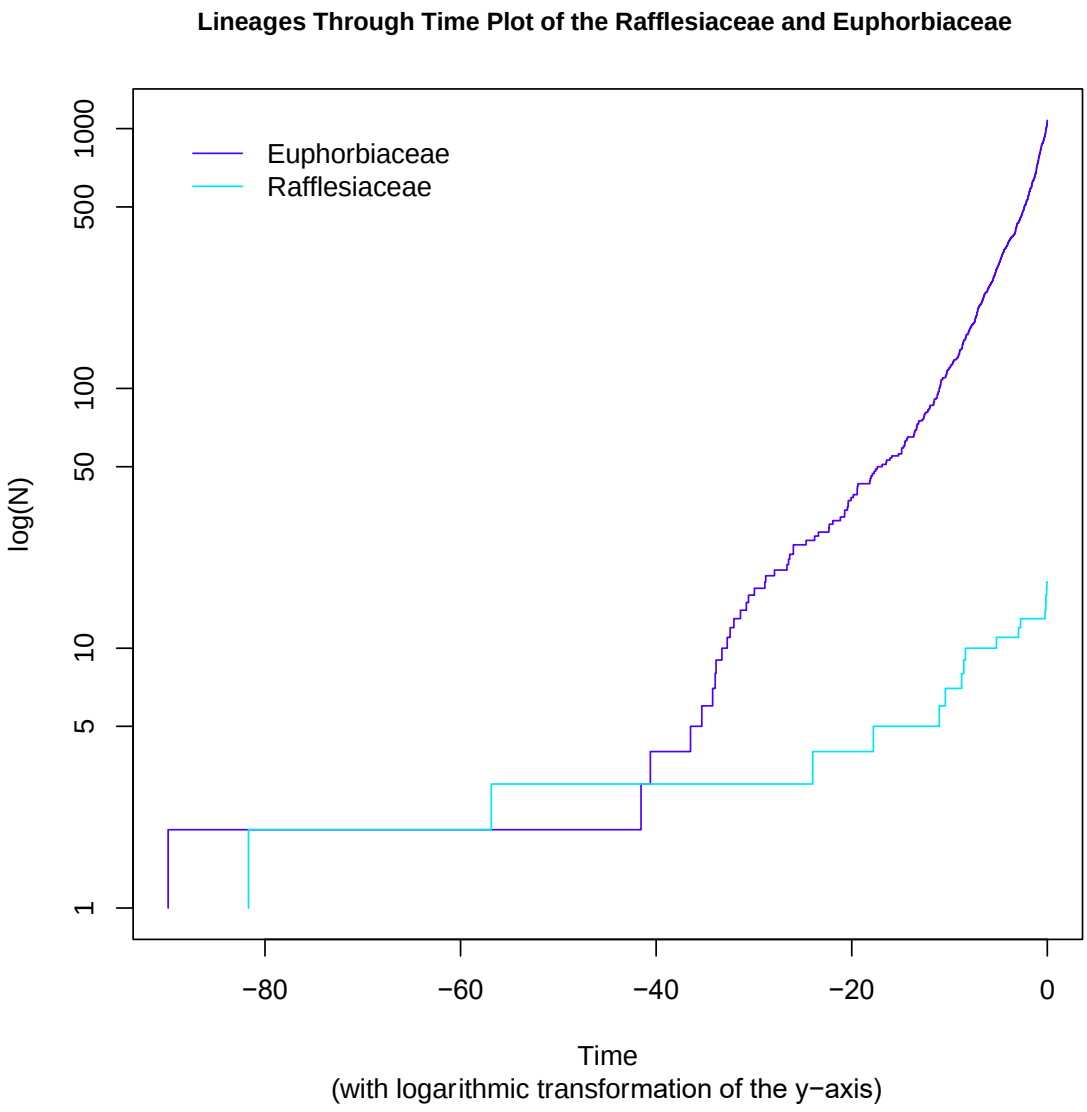

Figure 3.4. Lineage Through Time Plot for the parasitic clade Rafflesiaceae (blue line) versus the non-parasitic sister clade Euphorbiaceae (purple line). The lines show the number of species in each lineage through their evolutionary history.

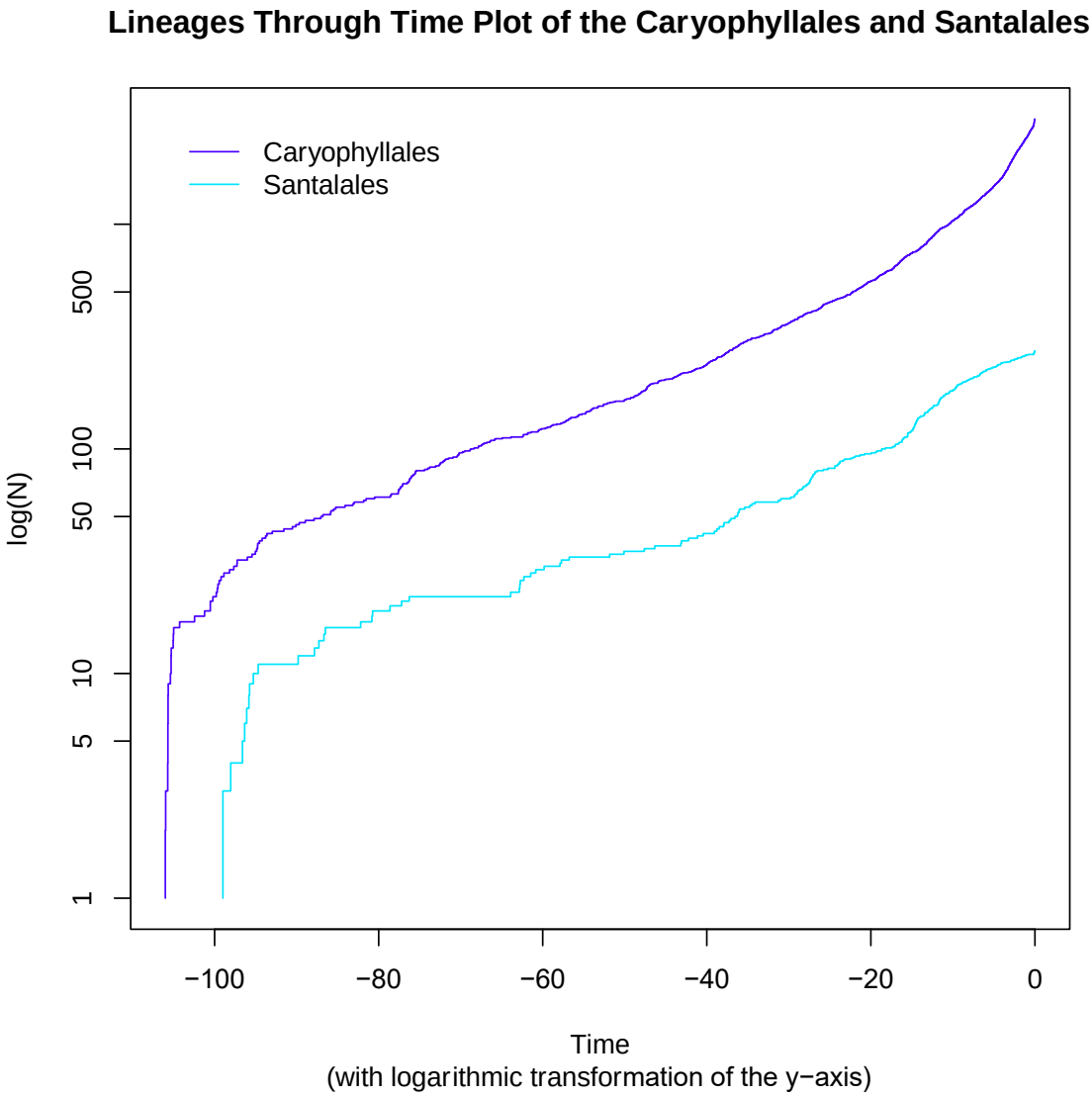

Figure 3.5. Lineage Through Time Plot for the parasitic clade Santalales (blue line) versus the non-parasitic sister clade Caryophyllales (purple line). The lines show the number of species in each lineage through their evolutionary history.

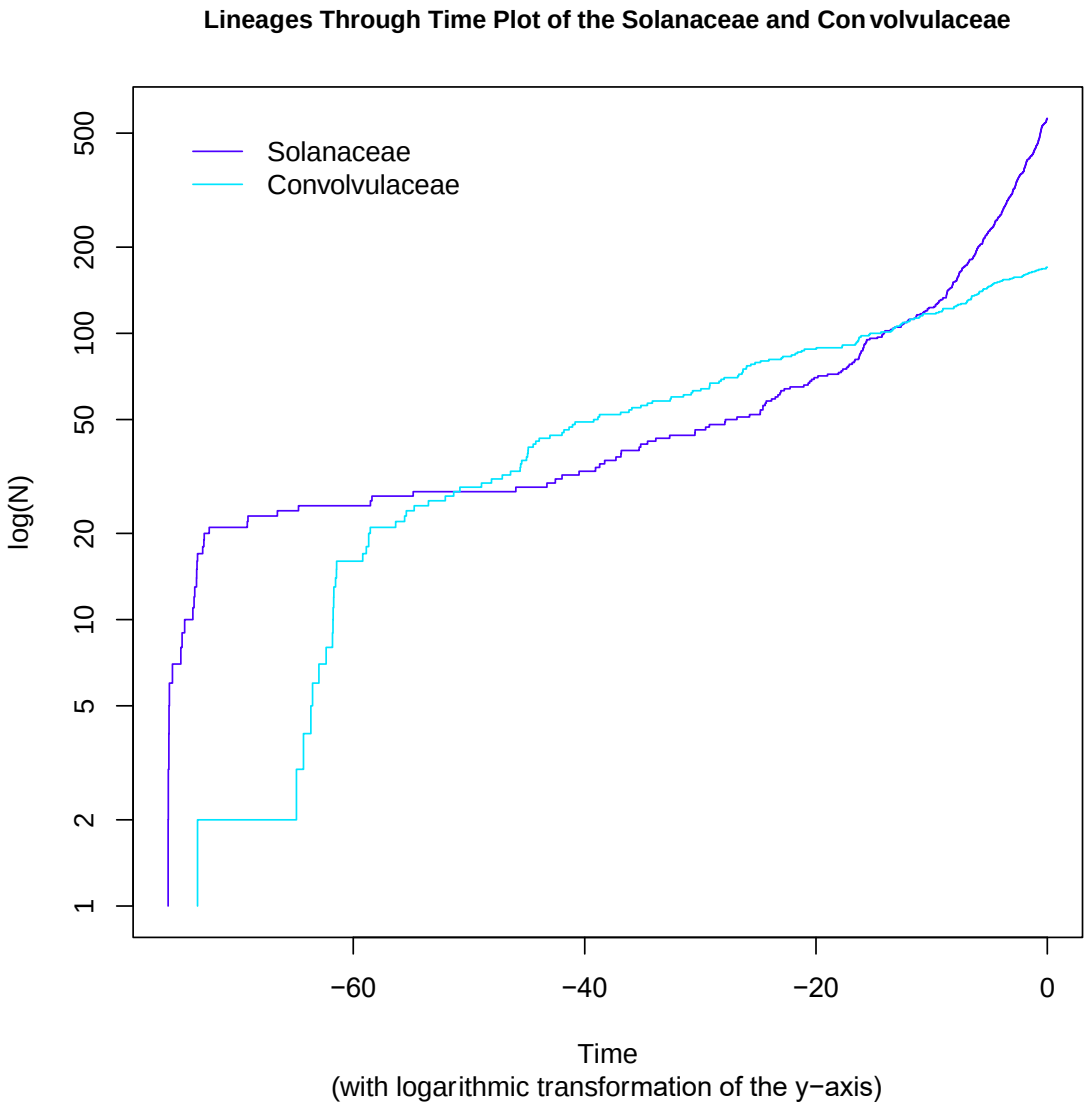

Figure 3.6. Lineage Through Time Plot for the parasitic clade Convolvulaceae (blue line) versus the non-parasitic sister clade Solanaceae (purple line). The lines show the number of species in each lineage through their evolutionary history.

Table 1.5. The estimated parameters under the best lineage diversification model for each clade. The parasitic clades are bolded. The non-parasitic clade and parasitic clade comparisons are grouped together.

Clade	Best Model	λ_0/r_1	k	μ_0	xparam	z	a
Dioscoreaceae	Variable Speciation	0.371	0.001	0.337			
Burmanniaceae + Thismiaceae	Exponential Density Dependent	0.472			0.878		
Solanaceae	Variable Extinction and Speciation	0.313	0.001	0.289		0.110	
Convolvulaceae	Exponential Density Dependent	0.284			0.468		
Caryophylalles	Variable Speciation	0.273	0.003	0.192			
Santalales	Pure Birth	0.043					
Euphorbiaceae	Birth Death	0.091					0.799
Rafflesiaceae	Birth Death	3.42E-08					0.999
Hypoxidaceae	Pure Birth	0.058					
Orchidaceae + Iridaceae	Birth Death	0.065					0.736

Table 1.6. The lineage diversification results for the parasitic clade Burmmaniaceae + Thismiaceae. The best model was the Exponential Density Dependent diversification model.

Models	AIC	Δ AIC
Pure Birth	188.820	21.517
Birth Death	190.820	23.517
Logistic Density Dependent	171.428	4.125
Exponential Density Dependent	167.303	0
Variable Speciation	173.112	5.810
Variable Extinction	193.460	26.157

Table 1.7. The lineage diversification results for Dioscoreaceae. The best model was the Variable Speciation model.

Models	AIC	Δ AIC
Pure Birth	-351.485	215.640
Birth Death	-559.589	7.536
Logistic Density Dependent	-345.630	221.496
Exponential Density Dependent	-530.298	36.827
Variable Speciation	-567.125	0
Variable Extinction	-557.578	9.547

Table 1.8. The lineage diversification results for Solanaceae. The best model was the Variable Extinction and Speciation Model.

Models	AIC	Δ AIC
Pure Birth	-2332.430	252.653
Birth Death	-2552.144	32.938
Logistic Density Dependent	-2185.680	399.402
Exponential Density Dependent	-2529.942	55.1409
Variable Speciation	-2553.735	31.348
Variable Extinction	-2550.132	34.951
Variable Extinction and Speciation	-2585.082	0

Table 1.9. The lineage diversification results for the parasitic clade Convolvulaceae. The best model was the Exponential Density Dependent model.

Models	AIC	Δ AIC
Pure Birth	35.009	18.543
Birth Death	37.009	20.543
Logistic Density Dependent	30.600	14.134
Exponential Density Dependent	16.466	0
Variable Speciation	17.994	1.528
Variable Extinction	39.421	22.955
Variable Extinction and Speciation	20.039	3.574

Table 1.10. The lineage diversification results for Caryophyllales. The best model was the Variable Speciation model.

Models	AIC	Δ AIC
Pure Birth	-20169.187	843.749
Birth Death	-20968.231	44.706
Logistic Density Dependent	-19503.377	1509.559
Exponential Density Dependent	-20804.279	208.658
Variable Speciation	-21012.937	0
Variable Extinction	-20963.864	49.073

Table 1.11. The lineage diversification results for Santalales. The best model was the Pure Birth model.

Models	AIC	Δ AIC
Pure Birth	-269.170	0
Birth Death	-263.662	5.508
Logistic Density Dependent	-246.910	22.260
Exponential Density Dependent	-263.023	6.147
Variable Speciation	-261.473	7.697
Variable Extinction	-261.671	7.499

Table 1.12. The lineage diversification results for Euphorbiaceae. The best model selected was the Birth Death model.

Models	AIC	Δ AIC
Pure Birth	-7521.940	201.390
Birth Death	-7723.330	0
Logistic Density Dependent	-7519.924	203.406
Exponential Density Dependent	-7710.260	13.069
Variable Speciation	-7717.479	5.851
Variable Extinction	-7721.329	2.000
Variable Extinction and Speciation	-7715.479	7.851

Table 1.13. The lineage diversification results for Rafflesiaceae. The best model was the Birth Death model.

Models	AIC	Δ AIC
Pure Birth	62.936	11.717
Birth Death	51.220	0
Logistic Density Dependent	64.937	13.717
Exponential Density Dependent	54.342	3.122
Variable Speciation	53.939	2.720
Variable Extinction	53.302	2.082
Variable Extinction and Speciation	55.939	4.720

Table 1.14. The lineage diversification results for Hypoxidaceae. The best model was the Pure Birth model.

Models	AIC	Δ AIC
Pure Birth	60.005	0
Birth Death	61.760	1.756
Logistic Density Dependent	61.999	1.995
Exponential Density Dependent	61.907	1.902
Variable Speciation	63.823	3.819
Variable Extinction	63.796	3.792
Variable Extinction and Speciation	65.823	5.819

Table 1.15. The lineage diversification results for Orchidaceae + Iridaceae. The best model was the Birth Death model.

Models	AIC	Δ AIC
Pure Birth	-31768.539	515.047
Birth Death	-32283.587	0
Logistic Density Dependent	-31111.451	1172.135
Exponential Density Dependent	-32221.986	61.600
Variable Speciation	-32280.794	2.792
Variable Extinction	-32281.044	2.543
Variable Extinction and Speciation	-32278.692	4.895

Table 1.16. The model fit results for the four models tested under the BiSSE joint model. The best model was the unconstrained model where all parameters were allowed to be estimated.

Models	lnL	AIC	Δ AIC
Unconstrained	-27631.598	55313.197	0
All Death Parameters Equal	-27686.311	55402.622	89.425
01 Transition Rate Equal	-27645.886	55331.772	18.575
All Parameters Equal	-28035.355	56080.710	767.513

Table 1.17. The BiSSE joint results for the unconstrained model, where all parameters are free. Speciation rate (lambda), extinction rate (mu), transition rate (q), 0 = non-parasitic, 1 = parasitic. Parasitic clades are bolded.

Sister Clades	lambda0	lambda1	mu0	mu1	q01
Caryophyllales + **Santalales**	0.425	0.733	0.409	0.729	0.001
Solanales + **Convolvulaceae**	0.607	0.707	0.596	0.709	0.000
Dioscoreaceae + **BurmanniaceaeThismiaceae**	0.584	0.773	0.581	0.720	0.003
Euphorbiaceae + **Rafflesiaceae**	0.704	0.893	0.556	0.663	0.000
Hypoxidaceae + **OrchidaceaeIridaceae**	0.425	0.679	0.381	0.691	0.003

Table 1.18. The BiSSE joint unconstrained model results, where all parameters are free. Net diversification rate (netdiv), speciation rate (lambda), transition rate (q), 0 = non-parasitic, 1 = parasitic. Parasitic clades are bolded.

Sister Clades	netdiv0	netdiv1	netdiv0/netdiv1	netdiv1/netdiv0	lambda0/q01
Caryophyllales + **Santalales**	0.016	0.005	3.323	0.301	340.488
Solanales + **Convolvulaceae**	0.011	-0.002	4.647	0.215	1431.087
Dioscoreaceae + **Burmanniaceae Thismiaceae**	0.004	0.052	0.069	14.475	194.032
Euphorbiaceae + **Rafflesiaceae**	0.148	0.230	0.644	1.552	1755.791
Hypoxidaceae + **Orchidaceae Iridaceae**	0.044	-0.013	3.484	0.287	132.0791

Table 1.19. The BiSSE joint results for the fully constrained model where all parameters are fixed. Speciation (lambda), extinction (mu), transition rate (q), 0 = non-parasitic, 1 = parasitic. Parasitic clades are bolded.

Sister Clades	lambda0	lambda1	mu0	mu1	q01
Caryophyllales + **Santalales**	0.285	0.046	0.250	0.005	0.000
Solanales + **Convolvulaceae**	0.285	0.046	0.250	0.005	0.000
Dioscoreaceae + **BurmanniaceaeThismiaceae**	0.285	0.046	0.250	0.005	0.000
Euphorbiaceae + **Rafflesiaceae**	0.285	0.046	0.250	0.005	0.000
Hypoxidaceae + **OrchidaceaeIridaceae**	0.285	0.046	0.250	0.005	0.000

Table 1.20. The BiSSE joint results for the equal death model, where extinction rates are fixed to be equal across clade comparisons. Speciation (lambda), extinction (mu), transition rate (q), 0 = non-parasitic, 1 = parasitic. Parasitic clades are bolded.

Sister Clades	lambda0	lambda1	mu0	mu1	q01
Caryophyllales + **Santalales**	0.228	0.509	0.197	0.510	0.002
Solanales + **Convolvulaceae**	0.221	0.515	0.197	0.510	0.000
Dioscoreaceae + **BurmanniaceaeThismiaceae**	0.210	0.507	0.197	0.510	0.007
Euphorbiaceae + **Rafflesiaceae**	0.474	0.619	0.197	0.510	0.001
Hypoxidaceae + **OrchidaceaeIridaceae**	0.262	0.511	0.197	0.510	0.004

Table 1.21. The BiSSE joint results for the equal transition rates model, where the transition rate from non-parasitic to parasitic was fixed to be the same across clade comparisons. Speciation (lambda), extinction (mu), transition rate (q), 0 = non-parasitic, 1 = parasitic. Parasitic clades are bolded.

Sister Clades	lambda0	lambda1	mu0	mu1	q01
Caryophyllales + **Santalales**	0.215	0.068	0.183	0.035	0.001
Solanales + **Convolvulaceae**	0.195	0.038	0.167	0.000	0.001
Dioscoreaceae + **BurmanniaceaeThismiaceae**	0.259	0.057	0.250	0.051	0.001
Euphorbiaceae + **Rafflesiaceae**	0.545	0.246	0.343	0.216	0.001
Hypoxidaceae + **OrchidaceaeIridaceae**	0.257	0.045	0.191	1.08E-08	0.001

Table 1.22. The fit of alternative models of parasitic angiosperm evolution across Dioscoreaceae + Burmanniaceae and Thismiaceae using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i), is denoted in bold.

SSE Models	AIC	ΔAIC	w_i (%)
parasite.bisse	2011.599	393.021	4.53E-84
parasite.hisse.full.model	**1618.578**	**0**	**99.919**
parasite.hisse.AandB.fixed	1632.803	14.225	0.081
parasite.hisse.0and1.fixed	2020.470	401.892	5.37E-86
parasite.hisse.A.fixed	1862.672	244.094	9.89E-52
parasite.hisse.B.fixed	1862.672	244.094	9.89E-52
parasite.hisse.1.fixed	1865.063	246.485	2.99E-52
parasite.hisse.0.fixed	2021.985	403.407	2.52E-86
parasite.hisse.1fixed.no0B	1728.390	109.812	1.43E-22
parasite.hisse.0fixed.no1B	1988.634	370.056	4.39E-79
parasite.hisse.no0B	1700.992	82.414	1.27E-16
parasite.hisse.no1B	1859.766	241.189	4.23E-51
parasite.bisse.ABand01Equal	2011.599	393.021	4.53E-84
parasite.hisse.full.model.ABand01Equal	1846.797	228.219	2.77E-48
parasite.hisse.AandB.fixed.ABand01Equal	1675.189	56.612	5.09E-11
parasite.hisse.0and1.fixed.ABand01Equal	2016.896	398.318	3.21E-85
parasite.hisse.A.fixed.ABand01Equal	1867.992	249.414	6.92E-53
parasite.hisse.B.fixed.ABand01Equal	1871.315	252.737	1.31E-53
parasite.hisse.1.fixed.ABand01Equal	1812.027	193.449	9.83E-41
parasite.hisse.0.fixed.ABand01Equal	1993.378	374.800	4.10E-80
parasite.hisse.1fixed.no0B.ABand01Equal	1905.990	287.412	3.88E-61
parasite.hisse.0fixed.no1B.ABand01Equal	1922.068	303.490	1.25E-64
parasite.hisse.no0B.ABand01Equal	1725.160	106.583	7.17E-22
parasite.hisse.no1B.ABand01Equal	1825.910	207.333	9.50E-44

Table 1.23. The fit of alternative models of parasitic angiosperm evolution across Caryophyllales and Santalales using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i), is denoted in bold.

SSE Models	AIC	ΔAIC	w_i
parasite.bisse	21432.240	2828.940	0
parasite.hisse.full.model	**18603.300**	**0**	**0.999**
parasite.hisse.AandB.fixed	18924.678	321.378	1.64E-70
parasite.hisse.0and1.fixed	21319.006	2715.706	0
parasite.hisse.A.fixed	18750.596	147.296	1.04E-32
parasite.hisse.B.fixed	18770.188	166.888	5.77E-37
parasite.hisse.1.fixed	18895.362	292.062	3.80E-64
parasite.hisse.0.fixed	21313.157	2709.857	0
parasite.hisse.1fixed.no0B	19145.751	542.451	1.62E-118
parasite.hisse.0fixed.no1B	21265.564	2662.263	0
parasite.hisse.no0B.raw	21242.362	2639.062	0
parasite.hisse.no1B	18745.562	142.262	1.28E-31
parasite.bisse.ABand01Equal	21432.240	2828.940	0
parasite.hisse.full.model.ABand01Equal	19054.950	451.649	8.43E-99
parasite.hisse.AandB.fixed.ABand01Equal	20434.079	1830.779	0
parasite.hisse.0and1.fixed.ABand01Equal	21410.903	2807.603	0
parasite.hisse.A.fixed.ABand01Equal	20522.187	1918.886	0
parasite.hisse.B.fixed.ABand01Equal	18720.683	117.382	3.24E-26
parasite.hisse.1.fixed.ABand01Equal	20527.504	1924.203	0
parasite.hisse.0.fixed.ABand01Equal	19128.922	525.622	7.29E-115
parasite.hisse.1fixed.no0B.ABand01Equal	20924.870	2321.569	0
parasite.hisse.0fixed.no1B.ABand01Equal	21336.075	2732.775	0
parasite.hisse.no0B.ABand01Equal	18634.753	31.453	1.48E-07
parasite.hisse.no1B.ABand01Equal	20548.039	1944.738	0

Table 1.24. The fit of alternative models of parasitic angiosperm evolution across Solanaceae and Convolvulaceae using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i), is denoted in bold.

SSE Models	AIC	ΔAIC	w_i (%)
parasite.bisse	5002.473	623.800	3.36E-134
parasite.hisse.full.model	4391.724	13.050	0.141
parasite.hisse.AandB.fixed	4421.055	42.381	6.03E-08
parasite.hisse.0and1.fixed	5014.471	635.800	8.35E-137
parasite.hisse.A.fixed	4387.385	8.712	1.235
parasite.hisse.B.fixed	4387.533	8.859	1.147
parasite.hisse.1.fixed	4387.302	8.628	1.287
parasite.hisse.0.fixed	4395.693	17.020	0.020
parasite.hisse.1fixed.no0B	4480.304	101.630	8.21E-21
parasite.hisse.0fixed.no1B	5014.221	635.547	9.46E-137
parasite.hisse.no0B	4484.303	105.630	1.11E-21
parasite.hisse.no1B	**4378.674**	**0**	**96.171**
parasite.bisse.ABand01Equal	5002.473	623.800	3.36E-134
parasite.hisse.full.model.ABand01Equal	4894.958	516.284	7.47E-111
parasite.hisse.AandB.fixed.ABand01Equal	4892.207	513.533	2.96E-110
parasite.hisse.0and1.fixed.ABand01Equal	5008.471	629.797	1.68E-135
parasite.hisse.A.fixed.ABand01Equal	4869.266	490.592	2.83E-105
parasite.hisse.B.fixed.ABand01Equal	4874.059	495.385	2.58E-106
parasite.hisse.1.fixed.ABand01Equal	4861.886	483.212	1.13E-103
parasite.hisse.0.fixed.ABand01Equal	4404.705	26.031	0.000
parasite.hisse.1fixed.no0B.ABand01Equal	4996.364	617.690	7.13E-133
parasite.hisse.0fixed.no1B.ABand01Equal	5008.222	629.548	1.90E-135
parasite.hisse.no0B.ABand01Equal	5000.368	621.694	9.64E-134
parasite.hisse.no1B.ABand01Equal	4899.038	520.364	9.71E-112

Table 1.25. The fit of alternative models of parasitic angiosperm evolution across Euphorbiaceae and Rafflesiaceae using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i), is denoted in bold.

SSE Models	AIC	ΔAIC	w_i (%)
parasite.bisse	4147.721	773.270	4.36E-167
parasite.hisse.full.model	3378.708	4.257	4.250
parasite.hisse.AandB.fixed	3375.264	0.813	23.778
parasite.hisse.0and1.fixed	4159.720	785.269	1.08E-169
parasite.hisse.A.fixed	3374.487	0.0361	35.069
parasite.hisse.B.fixed	**3374.451**	**0**	**35.708**
parasite.hisse.1.fixed	3390.260	15.809	0.013
parasite.hisse.0.fixed	4163.719	789.268	1.46E-170
parasite.hisse.1fixed.no0B	3798.557	424.106	2.88E-91
parasite.hisse.0fixed.no1B	4154.970	780.519	1.16E-168
parasite.hisse.no0B	3432.014	57.563	1.13E-11
parasite.hisse.no1B	3381.268	6.817	1.181
parasite.bisse.ABand01Equal	4147.721	773.270	4.36E-167
parasite.hisse.full.model.ABand01Equal	3504.409	129.958	2.15E-27
parasite.hisse.AandB.fixed.ABand01Equal	3902.984	528.533	6.07E-114
parasite.hisse.0and1.fixed.ABand01Equal	4153.719	779.268	2.17E-168
parasite.hisse.A.fixed.ABand01Equal	3850.023	475.572	1.92E-102
parasite.hisse.B.fixed.ABand01Equal	3724.923	350.471	2.81E-75
parasite.hisse.1.fixed.ABand01Equal	3498.861	124.410	3.45E-26
parasite.hisse.0.fixed.ABand01Equal	4148.644	774.193	2.75E-167
parasite.hisse.1fixed.no0B.ABand01Equal	3553.618	179.167	4.44E-38
parasite.hisse.0fixed.no1B.ABand01Equal	3576.870	202.419	3.96E-43
parasite.hisse.no0B.ABand01Equal	3613.735	239.284	3.92E-51
parasite.hisse.no1B.ABand01Equal	3869.327	494.876	1.24E-106

Table 1.26. The fit of alternative models of parasitic angiosperm evolution across Orchidaceae and Iridaceae + Hypoxidaceae using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i), is denoted in bold.

SSE Models	AIC	ΔAIC	w_i
parasite.bisse	22402.480	3029.955	0
parasite.hisse.full.model	19617.299	244.775	7.04E-54
parasite.hisse.AandB.fixed	19472.970	100.446	1.54E-22
parasite.hisse.0and1.fixed	22352.686	2980.162	0
parasite.hisse.A.fixed	19478.121	105.597	1.17E-23
parasite.hisse.B.fixed	**19372.524**	**0**	**1**
parasite.hisse.1.fixed	19598.676	226.152	7.79E-50
parasite.hisse.0.fixed	22247.306	2874.782	0
parasite.hisse.1fixed.no0B	19626.706	254.182	6.38E-56
parasite.no1B.raw.fixed	22281.957	2909.433	0
parasite.hisse.no0B	19535.996	163.472	3.18E-36
parasite.hisse.no1B.fixed	19526.030	153.506	4.64E-34
parasite.bisse.ABand01Equal	22402.480	3029.955	0
parasite.hisse.full.model.ABand01Equal	20326.333	953.809	7.64E-208
parasite.hisse.AandB.fixed.ABand01Equal	20262.574	890.049	5.35E-194
parasite.hisse.0and1.fixed.ABand01Equal	22343.621	2971.097	0
parasite.hisse.A.fixed.ABand01Equal	21178.378	1805.854	0
parasite.hisse.B.fixed.ABand01Equal	21523.331	2150.807	0
parasite.hisse.1.fixed.ABand01Equal	19902.999	530.475	6.44E-116
parasite.hisse.0.fixed.ABand01Equal	22348.579	2976.055	0

Table 1.27. The parameters inferred from the HiSSE best fitting model (unconstrained model) in the parasitic versus non-parasitic plant clade (Dioscoreaceae + Burmanniaceae and Thismiaceae). The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

Model	lambda	mu	netdiv	turnover	extinction fraction
Non-parasitic, A	0.114	0.122	-0.008	0.236	1.070
Parasitic, A	3.33E-23	1.99E-21	-1.96E-21	2.02E-21	59.691
Non-parasitic, B	6.283	5.704723	0.578	11.988	0.908
Parasitic, B	0.162	7.37E-11	0.162	0.162	4.55E-10

Table 1.28. The parameters inferred from the HiSSE best fitting model (unconstrained model) in the parasitic versus non-parasitic plant clade (Caryophyllales versus Santalales). The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

Model	lambda	mu	netdiv	turnover	extinction fraction
Non-parasitic,A	11.050	10.841	0.209	23.438	0.985
Parasitic, A	0.334	0.292	0.043	0.634	0.871
Non-parasitic, B	0.482	0.513	-0.0310	0.966	1.038
Parasitic, B	60.293	51.680	8.613	152.759	0.938

Table 1.29. The parameters inferred from the HiSSE best fitting model (unconstrained model) in the parasitic versus non-parasitic plant clade (Convolvulaceae versus Solanaceae). The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

Model	lambda	mu	netdiv	turnover	extinction fraction
Non-parasitic, A	0.493	0.521	-0.018	1.014	1.039
Parasitic, A	0.129	4.586E-08	0.250	0.129	2.338E-06
Non-parasitic, B	11.361	0.993	2.444	12.354	0.903
Parasitic, B	0	0	0	0	0

Table 1.30. The parameters inferred from the HiSSE best fitting model (unconstrained model) in the parasitic versus non-parasitic plant clade (Orchidaceae + Iridaceae versus Hypoxidaceae). The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

Model	lambda	mu	netdiv	turnover	extinction fraction
Non-parasitic, A	0.692	0.658	0.034	1.351	0.950
Parasitic, A	0.128	1.883E-08	0.128	0.128	1.474E-07
Non-parasitic, B	35.794	35.452	0.341	71.246	0.990
Parasitic, B	35.794	35.452	0.341	71.246	0.990

Table 1.31. The parameters inferred from the HiSSE best fitting model (unconstrained model) in the parasitic versus non-parasitic plant clade (Rafflesiaceae versus Euphorbiaceae). The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

Model	lambda	mu	netdiv	turnover	extinction fraction
Non-parasitic, A	2.387	2.393	-0.005	3.694	0.938
Parasitic, A	9.47E-03	3.65E-01	-3.55E-01	3.66E-01	1.069
Non-parasitic, B	108.345	104.786	3.557315	211.624	0.992
Parasitic, B	108.345	1.05E+02	3.557315	211.624	9.924E-01

CHAPTER III
DIVERSIFICATION AND HISTORICAL BIOGEOGRAPHY OF PASSERINES: A TRAIT-DEPENDENT APPROACH

This chapter is intended to be submitted as a manuscript to *Evolution*.

My primary contributions to this chapter include, (i) experimental design, (ii) collection and analysis of data, (iii) table and figure development, (iv) written completion of the manuscript draft for submission.

Abstract

Passerine birds are the most diverse group of Aves. Their diversification interests many because their morphology tends to be uniform across species. Passerines also are fairly cosmopolitan species, found nearly everywhere in the world, making their geographic range evolution of interest. We find that wing length explains most of the variation in these species. We model whether a larger body size (wing length and wing length normalized by body length) affects the dispersal of this group. We also test whether these two morphological traits and the region type they inhabit (tropical versus temperate) affect their diversification. Our results show that region type and wing length (small versus large) influence the diversification of passerine birds, yet the relationship of these is not clear, because we found evidence of another underlying trait or biological process, possibly wing area as influencing diversification. We show support that larger wing lengths do not influence increased dispersal rates in passerines. However, we recognize that complex historical biogeography models with more parameters have optimizations issues. Based on our results and other current studies, we recommend consideration of wing area, and larger sampling to more fully assess passerine diversification and biogeography.

Background

The Order Passeriformes includes the most species rich clades of birds with over 5,000 species (Gill and Donsker 2017), which comprise nearly 60% of extant birds (Sibley and Mondroe 1990). Not only are passerine birds the most diverse clade in Aves, they have a worldwide distribution and inhabit a remarkable diversity of ecological niches making them an interesting study for large-scale biogeographic studies (Sibley and Monroe 1990; Raikow and Bledsoe 2000; Claramunt and Cracraft 2015a, b; Moyle et al. 2016). Furthermore, although Passeriformes is the most diverse clade in Aves, their morphology is generally uniform (Ricklefs 2012). Ricklefs (2012) sought to understand whether the morphological space occupied by passerine birds increased directly with the number of species at a local regional scale, since he postulated the size of the niche could affect the occurrence of species and niche overlap. Through his analyses, Ricklefs concluded that passerine birds cluster toward the center of morphological space, with most species having a generalized morphology to be able to adapt to a wide variety of feeding substrates and prey items. However, there are certain species of passerines that do not follow this tendency and have varying morphology measurements, including varying wing lengths that differ from the uniform distribution of this trait. For example, the New Zealand wren is a poor flier and maintains a sedentary lifestyle. On the opposite spectrum, swallows have specialized morphology for aerial feeding, providing excellent larger wing to body area ratio for their necessary feeding behavior. Therefore, despite the general uniform trend of morphology amongst this group, there is still diversity. This begs the question: How does varying morphology linked to flight ability affect dispersal ability in passerine birds? Their fairly uniform morphology makes it interesting to look at how an outlier in morphology could

influence the dispersal rates in this group, making it a meaningful comparison between the structures of interest (Ricklefs 2012).

Biogeography

Recently, studies have sought to explain the biogeographic history of modern birds with probabilistic methods (Claramunt and Cracraft 2015; Moyle et al. 2016). One study sought to test multiple hypotheses of songbird evolution, but using phylogenomic data with fossil driven calibration. Their results supported an early songbird diversification from Australia into the Oligocene, followed by a geographic diversification and expansion into Southeast Asia in the early Miocene (Moyle et. Al. 2016). However, to date, no study has tried to link a trait, such as morphology tied to varying dispersal ability, as an impacting factor for this group's biogeographic history. A trait and biogeography model would be useful for assessing how traits could affect biogeographic history. Merging traits and biogeography is a novel approach to assess how morphology (i.e. fruit type, growth habit), behavior (i.e. aggressive, non-aggressive), diet (i.e. carnivore, herbivore), and other factors may affect the range of species. One objective of this study is to understand how a morphological trait can affect the dispersal and jump dispersal ability of passerine bird species. In this study, we test whether body size influences dispersal rates using two proxies: body size ratio (wing length/body length) and wing length, both treated as discretized traits, to understand their influence on dispersal ability anagenetic clade change of passerines. A meta-analysis by Jenkins et al. (2007) showed significant results supporting that dispersal distance depends on body size, with larger organisms achieving a greater dispersal distances. This study separated organisms into active (i.e. birds, mammals) versus passive dispersers (i.e. bacteria, certain seed types). Active dispersers had a positive dispersal-mass trend, meaning they dispersed significantly further and were significantly larger

in mass than passive dispersers. They also concluded that the size of the active dispersers mattered since their results supported that larger active dispersers reached greater dispersal distances than smaller active dispersers.

Diversification

Ricklefs (2006) discusses the diversification rates of passerines globally, with a focus of diversification of passerines for tropical versus temperate clades. The results suggest a higher speciation in tropical clades in South America than for North American temperate clades. The results also showed that there is a higher rate of diversification in the tropics than in the temperate regions, which support a latitudinal diversity gradient in passerines due to either higher speciation, lower extinction or both processes. The ages of the tropical versus temperate clades did not differ in the analyses, which suggests that tropical clades are not older than temperate ones. The authors stated that there were no methods to assess speciation and extinction parameters for species in these regions. However, at present we have such methods to assess diversification and estimation of extinction and speciation parameters through comparative phylogenetics (Maddison et al. 2007; Rabosky and Lovette 2008b; Beaulieu et al. 2013; Rabosky 2014; Beaulieu and O'Meara 2016). We seek to test whether tropical clades have a higher diversification rate over temperate clades. In this study, we also wanted to test whether accounting for phylogenetic relatedness affected the results of previous studies of morphological evolution in passerines.

The influence of body size on the diversification of different lineages has been studied by many (Brown and Maurer 1986; Damuth 1993; Jablonski 1997; Alroy 1998; Gilchrist et al. 2001; Laurin 2004; Thomas et al. 2009; Wollenberg et al. 2011), especially in relation to Cope's Rule, the generality that animal groups evolve toward larger body sizes (Stanley 1973; Hone and

Benton 2005). In passerines, there have been no studies explicitly testing whether body size correlates with increased diversification. Nudds (2007), using allometric scaling of wing-bone lengths, showed evidence that larger birds have longer wing lengths. Some studies have addressed how body size correlates, such as larger brain size, increase the diversification of birds due to a propensity for diverse behaviors to compete with other individuals, to allow for different feeding strategies and adaptability (Sol and Price 2008). The ability to have the capacity to fly further distances would allow for the separation of lineages and decreased genetic flow, thus increasing speciation, or better dispersal leading to higher gene flow.

Methods

Data Collection

The data used for the body size analyses is from Ricklefs (2012). This dataset of 1,642 species was reduced based on the overlap of the sequence data mined from GenBank (Buckner et al. 2014). The dataset was further reduced to not include invasive species, since these could skew the biogeography and diversification analyses. To do this, we used Long (1980) and Lever (1989) to further parse the list of species to not include invasive species. This left a total of 1,129 species for analyses. The biogeography presence/absence data was consolidated from 11 areas (Australia, Africa, New Guinea, Madagascar, Nearctic, Southern Asia, Neotropics, Palearctic, West Indies, Indonesia+Philippines, and New Zealand) to 6 areas (Nearctic, Palearctic, Afrotropic, Indomalaysia, Australasia and Neotropic) based on the biogeographic realms/ecozones (Udvardy and Udvardy 1975). This was necessary due to the large size of the data. The package BioGeoBEARS could not run analyses on 1,129 species with 11 possible area combinations and 2 discrete traits on 24 cores, as it crashed every time. After consolidating the areas, the analyses were able to run without an issue. The small and large body size was

classified in two ways, by wing length and wing length/body length. We used wing length, as it is the most common measure of body size (Ashton 2002). We also normalized wing length by body length to standardize species. To separate into small and large discrete body size categories, we looked at the average wing length and wing/length body ratios for passerine species known to be migratory species and aerial feeders and used their average values as a cutoff. We then looked at a histogram of the raw data values to determine the shape of the distribution and observed that this allowed an appropriate amount of small and large bodied species for our biogeographic analyses. The discrete data for the HiSSE analyses was determined from looking at the extant distribution of the passerine species. We used the tropical versus temporal classification of Ricklefs (2012) to assign species to either trait based on its range.

Sequence Data

In order to obtain sequence data for a phylogeny, we used the program PHLAWD (Smith et al. 2009) to mine the GenBank (Buckner et al. 2014) repository for all taxa available under the order Passeriformes for the following 11 genes: cmyc, cytb, musk, myoglobin, ornithine, RAG1, RAG2, rhodopsin, TGFB2, tropomyosin, ZENK. These are genes used in phylogenetic inference of passerines and relatives (Jetz et al. 2012). The sequences were aligned with MAFFT (Katoh et al. 2002). To effectively clean the data for missing data, we removed taxa that had a sequence length shorter than half the length of the longest sequence in Mesquite (Maddison and Maddison 2001). The program Mesquite was also used to remove hybrid taxa and species only at the genus level. We then used the program phyutility to clean, align and concatenate the sequences (Smith and Dunn 2008). The partial decisiveness (Sanderson et al. 2010) of the concatenated matrix was examined with the program Decisivator on a random simulation of 1,000 random trees (Zhbannikov et al. 2013).

Phylogenetic Inference and Dating

We created partition files so each gene in the concatenation could evolve under a different rate of molecular evolution under Maximum Likelihood for tree inference. We used RaxML-HPC BlackBox (Stamatakis 2006) on the CIPRES Science Gateway (Miller et al. 2010) on the concatenated alignment with a partition/mixed model to allow for differing rates among the genes under the recommended GTR + Γ model by Stamatakis (2006), which would also print the corresponding branch lengths. All tree dating was done with TreePL (Smith and O'Meara 2012) with fossil dates constraints (Boles 1993; Mayr and Manegold 2006; Worthy et al. 2007; Prum et al. 2015) and the Prum et al. (2015) estimated age for crown passerines of 53.5 million years. We also inferred dated phylogenies under only the fossil dates and only under the Prum et al. (2015) inferred crown age, but the dated phylogeny with all age constraints had better age estimates at the more shallow nodes.

PCA and Phylo PCA

From the morphological data from Ricklefs (2012) comprising wing length, body length, tail length, tarsus, middle toe, bill length, bill width and bill depth, we sought to carry out a principal components analysis (PCA) and compare these traits to see if there was a phylogenetic effect with phylogenetic PCA. We used the R package phytools (Revell 2012) for phylogenetic PCA analyses we used the stats package in R (Team 2000, 2014). We followed recommendations from Uyeda et al. (2014).

Diversification and Biogeographic Analyses

Diversification analyses were carried out using the R package laser for pure birth, birth death, logistic dependent, exponential dependent, variable speciation, variable extinction and

variable speciation and extinction diversification models (Rabosky 2006). We also visually looked at the Lineage Through Time Plot of the clade to understand the diversification process (Harvey et al. 1994; Stadler 2008). Our passerine tree does not have a complete sampling of all species. Incomplete lineage sampling can skew results to appear as having an early burst, followed by a decreased diversification. Therefore, we assessed the effect of incomplete lineage sampling on the diversification tests for each clade by calculating the gamma statistic and using the MCCR test in the package laser.

We used a HiSSE model to understand how speciation, extinction, net diversification, turnover and extinction fraction differed in tropical versus temperate clades through the hisse R package (Beaulieu et al. 2016; Beaulieu and O'Meara 2016). We set up 24 different models including a fully unconstrained model and 22 different constrained HiSSE models where the parameters were fixed in different ways, as well as a BiSSE model. We corrected for incomplete sampling. The advantage of the HiSSE method is that it allows for a hidden trait with a binary state to affect the diversification of a clade, so the rate heterogeneity inferred is not necessarily attributed to the specific trait being tested.

Biogeography analyses were carried out in BioGeoBEARS (Matzke 2013a) using the DEC, DEC*, DEC+J, DEC*+J, DIVAlike, DIVAlike+J, BayArealike and BayArealike+J (Ronquist 1997; Landis et al. 2013; Matzke 2013b; Matzke 2014). We ran unconstrained analyses and analyses that incorporated trait dependent dispersal for small and large sizes for two models: wing length and body size (wing length/body length). The trait dependent analyses do not allow for time stratification analyses, so we could not incorporate geological information. For the trait-dependent analyses we tried different starting point values to test parameter estimate

likelihood optimization using the GenSA optimization algorithm with slow and fast Maximum Likelihood searches (Matzke 2016).

Results

Phylogenetic Inference

We inferred a Maximum Likelihood phylogenetic tree of 1,129 species of passerine birds (Figure 4.1). The partial decisiveness score for the inferred phylogeny was 0.8453, suggesting the data is decisive for 84.53% of all possible trees.

Wing length explains most of the variation of passerines

The first principal component (PC) explained 76.7% of the total variation in the dataset for the standard PCA analysis. With the phylogenetic PCA analysis, the first PC explained 65.1% of the total variation. The first PC is a general size variable (wing length) and PCs 2-7 are variables that characterize different morphological traits associated with different foraging strategies, substrates and prey (Ricklefs 2012). The general size variable seems to explain most of the variation in in morphological space, which gives support to using it as a meaningful trait for diversification and biogeographic analyses. The first PC explains slightly less variation in the phylogenetic PCA.

Diversification Models and LTT plots support density-dependent diversification

The best diversification model was density dependent diversification (Table 2.1). The initial speciation parameter estimated with the dependent diversification was 0.097 and the *k* parameter, carrying capacity was 2,258 (Table 2.2). The LTT plot clearly shows an excess of the passerine lineages in the early radiation of passerines in relation to what would be expected under a constant rate model of lineage diversification (a line with a slope of 1) (Figure 4.2).

These findings are in accordance with the results of other studies of avian diversification (Nee et al. 1992; Rabosky and Lovette 2008a), where there is also a density dependent process showing an early excess of lineages in the early radiation of birds. The results from the MCCR test show non-significant findings, suggesting that even with incomplete lineage sampling, this diversification test shows that there are more cladogenetic events (speciation rate is higher) earlier in the tree than expected under constant rate model of lineage diversification.

HiSSE results suggest a hidden trait as influential in the diversification of passerines

Wing Length/Body Length Ratio

The best model had the hidden states constrained to be equal, meaning that the trait of interest small/large body size does not influence diversification (0A = 1A and 0B = 1B), rather a hidden trait does with states A and B (regardless of whether the bird has a small or large body size) (Table 2.2). For parameter estimates see Table 2.3. For hidden trait state A the net diversification and turnover that is ~ 5.2 times higher than for the hidden trait with state B, and an extinction fraction that is 7.656e-23 smaller. These results suggest that the wing length/body length ratio of a passerine bird does not influence the observed diversification at all. Instead, the differences seen in diversification are driven completely by hidden factors.

Wing Length

The best model had turnover and extinction fraction fixed for the hidden trait with state A (1A = 0A) with an 83.5% of the AIC model weight (Table 2.4). This supports the conclusion that the hidden trait with state A is not influenced by wing length, but the hidden trait with state B is influenced by the length of the wing. For parameter estimates please see Table 2.5. The net diversification rate for either trait (small versus large wing length) with state A of the hidden trait

is 0.074. The net diversification rate for the small wing length influenced by hidden trait state B is 0.389. The net diversification for the large wing length influenced by hidden trait with state B is 0.400. The hidden trait with state B has approximately a 1-time greater diversification rate for the larger wing length than for the smaller wing length. This model shows that the hidden trait with state B influences passerine birds to have a ~ 5.2 times greater diversification rate in birds with small wing lengths and ~ 5.4 times greater diversification rate for large wing lengths over birds influenced by the hidden trait with state A, regardless of the wing length size. The hidden trait with state A has 19% of the turnover for the hidden trait with state B, regardless of the wing length. The extinction fraction for the hidden trait with state A is also much lower that for the hidden trait with state B.

Tropical versus temperate

The best model was the one with the turnover and extinction fractions rates fixed for state B for the hidden trait (52.3% of the AIC model weight), followed by the HiSSE model weight with the turnover and extinction fraction for the hidden trait for A state fixed (47.7% of the AIC model weight) (Table 2.6). For the parameters estimates for both of these models, please see Tables 2.7 and 2.8. The model with both states A and B fixed was a poor model with 2.15E-15% of the AIC model weight (Table 2.6). For the model with the hidden trait state B fixed, the net diversification is about 227 times greater for tropical over temperate. However, when the hidden trait with state A is fixed, the temperate trait influences a 41 times greater diversification over the temperate trait with the influence of a hidden trait with state B. The results suggest that either hidden state B or state A should be fixed, but not both. Therefore, there is evidence for the hidden trait with both states A and B and that the tropical and temperate trait influences one of those hidden traits. However, the relationship of the influence of the tropical and temperate traits

is not clear on diversification of passerines. Improving the sampling of this group may or may not help disentangle this story.

Larger body sizes do not increase dispersal rates in passerines

For the unconstrained no trait-dependent scenario, the best model was the BayArealike +J. The ancestral state estimations were not consistent with previously estimated models of passerine biogeography. For instance, passerines were inferred to have originated from the West Indies in our analysis. This is problematic because these islands are younger than the estimated age of passerines. Previous studies using time stratified models account for the geological history of the Earth infer oscine passerines to have originated from Australia (Moyle et. al. 2016). For the trait-dependent biogeography model, parameter estimates and likelihood values showed issues with optimization for both models of wing length and wing length/body length (Tables 2.9 and 2.10). We used a slow optimization search and used the parameters estimated from the top 2 best models from each model type (DEC+J, DEC*+J, DIVAlike+J, BayArealike+J) as the starting parameters, in hopes of ameliorating the optimization issue. For the wing length/body length model, the likelihood score values were similar between the models (Table 3.9) indicating the model optimized parameters and likelihood optimized. However, for the wing length model, the likelihood scores were a difference of 745.47 –lnL, indicating the model did not optimize well (Table 3.10). This could potentially indicate that the parameters are hard to identify in the likelihood space and using a more robust optimization algorithm is needed. Therefore, based on these results, we feel slightly more confident in interpreting the parameter estimates from the wing length/body length ratio model. The dispersal, local extinction, jump dispersal and m_2 (large size multiplier on dispersal) parameter estimates were essentially the same, however, the t_{12} (transition rate from small to large body size) and t_{21} (transition rate from large to small body

size) parameter estimates were slightly different, please see Tables 2.9 and 2.10. The dispersal multiplier on the wing length/body length ratio indicated that the larger wing length/body length ratio does not have a higher influence over the small wing length/body length ratio. In fact, passerine birds with a smaller wing length/body length ratio have a ~ 2.3 greater dispersal over passerine birds with a larger wing length/body length ratio. Jump dispersal is ~ 10.5 times more likely than regular dispersal events.

Discussion

Our diversification results show support for diversity-dependent diversification of passerines, meaning they have an increase in the number of species quickly at first, but then species richness becomes limited as the diversification starts to slow down after they start to fill ecological space and have ecological constraints. There are many speciation events at the origin of the clade and these start to subside. This is in accordance with many studies (Ricklefs 2007; Rabosky 2009, 2013) and ones regarding bird diversification (Nee et al. 1992; Phillimore and Price 2008; Rabosky and Lovette 2008a). However, Ricklefs and Jønsson (2014) argue that passerines do not exhibit diversity dependent diversification, instead there is a random speciation-extinction process based on their F-ratio statistical test based on error sum of squares. According to Beaulieu and O'Meara (2016), a better approach would be to not assess significance, but rather use information theory, to assess the loss of information explained by the model, or through Bayesian approaches.

Many studies have supported a higher diversification for various groups of taxa in the tropics (Pianka 1966; Stevens 1989; Hillebrand 2004; Jablonski et al. 2006; Mittelbach et al. 2007), including passerines (Hawkins et al. 2006; Ricklefs 2006). There is also support that vertebrates and trees decrease in species richness the further away from the tropics, consistent

with a latitudinal diversity gradient in these species (Hillebrand and Azovsky 2001). However, in some groups, a reversed latitudinal diversity gradient has been shown (Kouki et al. 1994; Buckley et al. 2003; Tedersoo and Nara 2010), including in some avian species (Rabenold 1979). Our results do not show strong support for increased diversification of passerines in the tropics. Although the best model suggests a 227 times higher diversification rate for passerines in the tropics versus the temperate region, we question the power on the result from this model due to the AIC model weight not being much higher than for the next best model which supported a higher diversification in the temperate region (AIC model weights being 52.3% and 47.3%, respectively). It is evident from the results that there is another hidden trait that correlates with the region type in passerines, however region only influences one of the states of the hidden binary trait. Wing length might be a good candidate for the hidden trait. Ashton (2002) carried out a meta-analysis showing that the majority of birds are larger at higher latitudes, whereas tropical latitudes have birds that tend to be smaller in size, concluding that Bergmann's rule holds for birds throughout the world no matter whether they are migratory or sedentary species. The study only used data for 122 species, a small amount considering the estimated 10,672 extant bird species recognized (Gill and Donsker 2017).

We tested two different proxies for body size in passerines: wing length and wing length/body length. Wing length is used in most studies as a proxy for body size (Ashton 2002). We used the ratio proxy to normalize wing length by body length. Our results show evidence that wing length slightly influences increased diversification of passerines, but a hidden trait matters as well. This gives support that a body size influences the diversification of passerines. There was no evidence that the wing length/body length trait mattered at all, due the result of a hidden trait only mattering. These results suggest that the length, and possibly shape and area of the

wing matter more (discussed below). We have evidence that region type (temperate versus tropical) and wing length (small versus large) influence the diversification of passerine birds. Although, our results show evidence for region type and wing length (both with the influence of a hidden trait) affecting the diversification of passerines, our results do not prove that each of these traits is each other's hidden trait. Future studies would need to explicitly test both of these traits together through a Multiple State Speciation and Extinction Model (FitzJohn 2012) with a hidden state to explicitly test this conclusion.

A trait that would paint a better, and hopefully a more accurate picture, would be wing aspect ratio, where the square of the wingspan is taken as a ratio over the wing area. Another trait that is similar to the wing aspect ratio is the hand wing index described by Claramunt et al. (2012). The hand wing index accounts for the wing length and the secondary wing length to understand the shape and area of the wing. Another body size aspect to consider is the effect of body mass on the wing loading. Hayssen and Kunz (1996) discuss that carrying extra body mass has implications on wing loading, thus having a larger wing area relative to the body mass is more advantageous. Due to the nature of the dataset, this was not something that we could explicitly measure because we did not have wing area or body mass data. However, we would suggest future studies use aspect ratio to test diversification of passerines. Also, to date, the HISSE model does not allow for the use of continuous data. Even though we discretized the data based on a biological meaningful reason, having to discretize the body size data is still an arbitrary designation. It would be better to treat the data as continuous when assessing its influence on diversification.

The results of the trait-dependent biogeography model were not expected. The problems with optimization were not anticipated. The results show that the parameters linked to the trait

are hard to identify. Thus, studies that consider using this type of model need to carry out further measures until the parameters and likelihood are optimized. Our study was the first to explicitly test the hypothesis that passerine birds with larger body size (i.e. wing length) have greater dispersal ability than other passerines. Kennedy et al. (2016) used correlative methods to test this, but did not measure dispersal explicitly with historical biogeographic methods. The results show support for passerines inheriting an ancestor's biogeographic area, except for jump dispersal events in which the daughter lineage jumps into a new region. The results also show that jump dispersal events are more common than dispersal events. This makes intuitive sense: local dispersal facilitates continued gene flow among populations in each species and thus deters reproductive isolation, genetic differentiation and speciation, while through jump dispersal species colonize new areas and lineages become geographically isolated from one another.

A larger body size has been linked many times with increased dispersal, however, our results indicate otherwise. Kennedy et al. (2016) showed that when accounting for the evolutionary history of passerines, there was no relationship for migratory species versus sedentary species of passerines having a larger hand wind index. In this study, passerine species that were termed migratory (practiced a seasonal movement of more than 1,000 km between breeding and wintering grounds) did not have a larger wing shape and area. This suggests that low dispersal capabilities may not necessarily implicate low diversification, since low dispersal rate could aid reproductive isolation. In other words, if passerines have a low dispersal capability, then there might be a limitation on gene flow between populations of passerines that have become disconnected due to the low dispersal capability (Diamond et al. 1976; Kennedy et al. 2016), leading to allopatric speciation.

Future studies should seek to expand the dataset to include more species of passerines, since our study was limited to the species in our dataset and what was available in GenBank (approximately 20% of passerines species), while carefully considering the sampling of clades within passerines, so some lineages are not overrepresented over others. Another consideration for future studies is to replicate these analyses with other published phylogenies to see if the results are consistent among tress, since we know that not one single phylogeny is true. This would also see if the sampling and methodology used in this study is sufficient. This is especially relevant for the phylogenetic inference aspect of the study.

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Appendix D

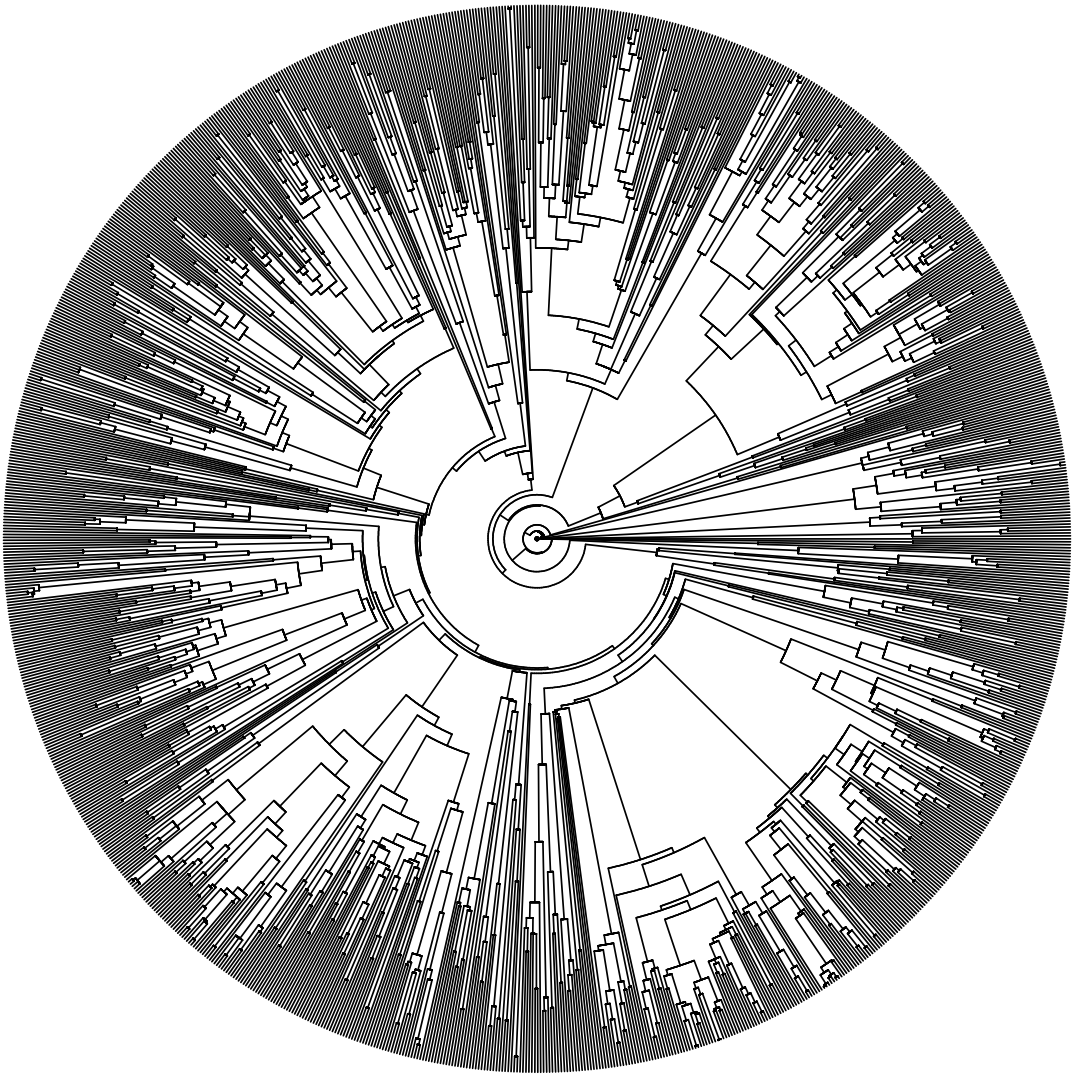

Figure 4.1. The inferred phylogeny under the program RaxML for 1,129 passerine birds from the concatenated alignment of 11 genes.

Table 2.1. The lineage diversification model results for passerine birds. The best model was the Logistic Density Dependent model.

Models	AIC	Δ AIC
Pure Birth	-5231.044	159.395
Birth Death	-5229.044	161.395
Logistic Density Dependent	-5390.439	0
Exponential Density Dependent	-5366.677	23.761
Variable Speciation	-5388.807	1.632
Variable Extinction	-5223.151	167.287
Variable Extinction and Speciation	-5388.061	2.378

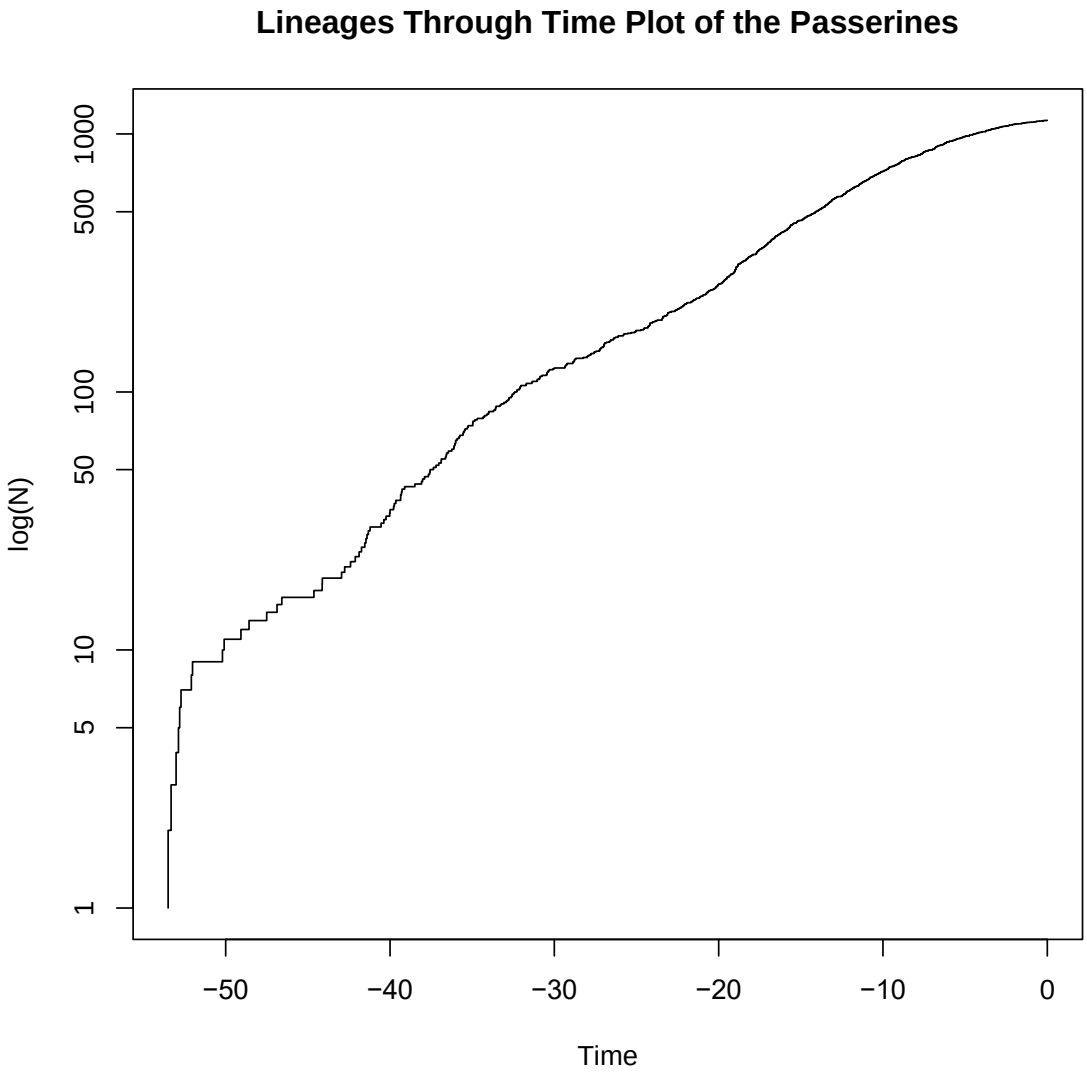

Figure 4.2. The Lineage Through Time Plot for passerines. The black line shows the number of species in the lineage through their evolutionary history.

Table 2.2. The fit of alternative models of normalized body size evolution across passerines using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i), is denoted in bold.

SSE Models	AIC	ΔAIC	w_i
area.bisse	8907.498	351.339	3.76036E-77
area.hisse.full.model	8658.022	101.863	5.60192E-23
area.hisse.AandB.fixed	**8556.159**	**0**	**0.737**
area.hisse.0and1.fixed	8883.850	327.691	5.13342E-72
area.hisse.A.fixed	8567.415	11.256	0.003
area.hisse.B.fixed	20000000028	19999991472	0
area.hisse.1.fixed	8808.205	252.046	1.36918E-55
area.hisse.0.fixed	8571.544	15.385	0.000
area.hisse.1fixed.no0B	8796.275	240.116	5.33316E-53
area.hisse.0fixed.no1B	8684.021	127.862	1.26684E-28
area.hisse.no0B	8561.750	5.591	0.045
area.hisse.no1B	20000000028	19999991472	0
area.bisse.ABand01Equal	8907.498	351.339	3.76036E-77
area.hisse.full.model.ABand01Equal	8563.723	7.564	0.017
area.hisse.AandB.fixed.ABand01Equal	8692.058	135.899	2.2776E-30
area.hisse.0and1.fixed.ABand01Equal	8817.678	261.520	1.2004E-57
area.hisse.A.fixed.ABand01Equal	8635.524	79.365	4.30278E-18
area.hisse.B.fixed.ABand01Equal	8560.345	4.186	0.091
area.hisse.1.fixed.ABand01Equal	8862.195	306.037	2.58568E-67
area.hisse.0.fixed.ABand01Equal	8564.220	8.062	0.013
area.hisse.1fixed.no0B.ABand01Equal	8814.242	258.083	6.69232E-57
area.hisse.0fixed.no1B.ABand01Equal	8622.208	66.049	3.35163E-15
area.hisse.no0B.ABand01Equal	8560.277	4.118	0.094
area.hisse.no1B.ABand01Equal	8625.787	69.628	5.59742E-16

Table 2.3. The parameters inferred from the HiSSE best fitting model (both hidden states A and B fixed) for the normalized body size in passerines model. The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

States	lambda	mu	netdiv	turnover	extinction fraction
Small Body Size, A	0.410	6.54E-37	4.10E-01	0.410	1.60E-36
Large Body Size, A	0.410	6.54E-37	4.10E-01	0.410	1.60E-36
Small Body Size, B	0.079	6.50E-16	7.90E-02	0.079	2.09E-14
Large Body Size, B	0.079	6.50E-16	7.90E-02	0.079	2.09E-14

Table 2.4. The fit of alternative models of wing length evolution across passerines using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i), is denoted in bold.

SSE Models	AIC	ΔAIC	w_i
wing.bisse	8931.091	356.798	2.78E-78
wing.hisse.full.model	8579.370	5.077	0.066
wing.hisse.AandB.fixed	8578.979	4.686	0.080
wing.hisse.0and1.fixed	8840.522	266.229	1.29E-58
wing.hisse.A.fixed	**8574.293**	**0**	**0.835**
wing.hisse.B.fixed	8582.133	7.840	0.017
wing.hisse.1.fixed	8593.410	19.117	0.000
wing.hisse.0.fixed	8779.907	205.615	1.87E-45
wing.hisse.1fixed.no0B	8651.384	77.092	1.52E-17
wing.hisse.0fixed.no1B	8775.907	201.614	1.38E-44
wing.hisse.no0B	8605.000	30.708	0.000
wing.hisse.no1B	8585.663	11.371	0.003
wing.bisse.ABand01Equal	8931.091	356.798	2.78E-78
wing.hisse.full.model.ABand01Equal	8783.762	209.469	2.73E-46
wing.hisse.AandB.fixed.ABand01Equal	8727.299	153.007	4.97E-34
wing.hisse.0and1.fixed.ABand01Equal	8910.876	336.583	6.81E-74
wing.hisse.A.fixed.ABand01Equal	8778.957	204.664	3.01E-45
wing.hisse.B.fixed.ABand01Equal	8666.871	92.578	6.58E-21
wing.hisse.1.fixed.ABand01Equal	8794.078	219.786	1.57E-48
wing.hisse.0.fixed.ABand01Equal	8753.603	179.311	9.65E-40
wing.hisse.1fixed.no0B.ABand01Equal	8825.714	251.421	2.12E-55
wing.hisse.0fixed.no1B.ABand01Equal	8769.942	195.649	2.73E-43
wing.hisse.no0B.ABand01Equal	8749.854	175.561	6.29E-39
wing.hisse.no1B.ABand01Equal	8692.186	117.893	2.1E-26

Table 2.5. The parameters inferred from the HiSSE best fitting model (both hidden states A and B fixed) for the wing length in passerines model. The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

States	lambda	mu	netdiv	turnover	extinction fraction
Small Wing Length, A	0.074	4.31E-26	7.36E-02	0.074	5.85E-25
Large Wing Length, A	0.074	4.31E-26	7.36E-02	0.074	5.85E-25
Small Wing Length, B	0.392	0.003	0.389	0.396	0.009
Large Wing Length, B	0.400	2.74E-12	4.00E-01	0.400	6.85E-12

Table 2.6. The fit of alternative models of tropical versus temperate habitat evolution across passerines using HiSSE. The best model, based on ΔAIC and Akaike weights (w_i) is denoted in bold.

SSE Models	AIC	ΔAIC	w_i
biome.bisse	9034.923	384.733	1.49532E-84
biome.hisse.full.model	8672.203	22.013	8.67865E-06
biome.hisse.AandB.fixed	8725.651	75.462	2.14882E-17
biome.hisse.0and1.fixed	8961.963	311.773	1.04173E-68
biome.hisse.A.fixed	**8650.374**	**0.184**	**0.477**
biome.hisse.B.fixed	**8650.190**	**0**	**0.523**
biome.hisse.1.fixed	8889.273	239.083	6.34105E-53
biome.hisse.0.fixed	8733.771	83.582	3.70619E-19
biome.hisse.1fixed.no0B	8888.225	238.032	1.07274E-52
biome.hisse.0fixed.no1B	8790.352	140.162	1.91692E-31
biome.hisse.no0B	8680.449	30.259	1.40552E-07
biome.hisse.no1B	8695.643	45.453	7.05466E-11
biome.bisse.ABand01Equal	9034.923	384.733	1.49532E-84
biome.hisse.full.model.ABand01Equal	8752.509	102.319	3.163E-23
biome.hisse.AandB.fixed.ABand01Equal	8782.288	132.098	1.08073E-29
biome.hisse.0and1.fixed.ABand01Equal	8955.237	305.047	3.00791E-67
biome.hisse.A.fixed.ABand01Equal	8766.828	116.638	2.45896E-26
biome.hisse.B.fixed.ABand01Equal	8739.045	88.855	2.654E-20
biome.hisse.1.fixed.ABand01Equal	8968.031	317.841	5.0135E-70
biome.hisse.0.fixed.ABand01Equal	8734.752	84.562	2.27004E-19
biome.hisse.1fixed.no0B.ABand01Equal	8966.478	316.288	1.08994E-69
biome.hisse.0fixed.no1B.ABand01Equal	8789.914	139.724	2.38663E-31
biome.hisse.no0B.ABand01Equal	8732.600	82.410	6.65911E-19
biome.hisse.no1B.ABand01Equal	8762.557	112.367	2.08065E-25

Table 2.7. The parameters inferred from the HiSSE best fitting model (state B fixed for the tropical versus temperate habitat in passerines model. The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

States	lambda	mu	netdiv	turnover	extinction fraction
Tropical, A	0.462	3.55E-15	4.62E-01	0.462	7.69E-15
Temperate, A	0.371	0.002	0.369	0.373	0.005
Tropical, B	0.075	3.84E-26	7.46E-02	0.075	5.15E-25
Temperate, B	0.075	3.84E-26	7.46E-02	0.075	5.15E-25

Table 2.8. The parameters inferred from the HiSSE best fitting model (state A fixed for the tropical versus temperate habitat in passerines model. The parameters are: speciation rate (lambda), extinction rate (mu), net diversification rate (netdiv), turnover rate (lambda + mu), and extinction fraction rate (lambda/mu).

States	lambda	mu	netdiv	turnover	extinction fraction
Tropical, A	0.075	8.99E-16	7.51E-02	0.075	1.20E-14
Temperate, A	0.075	8.99E-16	7.51E-02	0.075	1.20E-14
Tropical, B	0.464	1.47E-14	4.64E-01	0.464	3.16E-14
Temperate, B	0.382	0.020	19.135	0.402	0.052

Table 2.9. The BioGeoBEARS results under the normalized body size trait dispersal model. Dispersal rate (*d*), extinction rate (*e*), jump dispersal (*j*), transition rate from smaller body size to larger body size (t_{12}), transition rate from larger body size to smaller body size (t_{21}), multiplier on smaller-bodied dispersal (m_1), multiplier on larger-bodied dispersal (m_2).

Base Model	m_1 start	*d*	*e*	*j*	t_{12}	t_{21}	m_1	m_2	LnL
DECJ	1	0.002658318	1.00E-12	0.006	0.007142747	0.0233879	1	1.023019	-1921.307
DECJ	2	0.002234458	1.00E-12	0.006	0.006918588	0.04728451	1	1.984691	-1903.573
DECJ	3	0.001922548	1.00E-12	0.005790397	0.00681963	0.04333876	1	2.987029	-1895.949
DECJ	2.987029	0.001875416	1.00E-12	0.005407632	0.007097069	0.05092975	1	3.1834	-1894.413
DECJ	4	0.001708548	1.00E-12	0.004985932	0.006851397	0.0451244	1	3.977043	-1893.835
DECJ	3.977043	0.001695189	1.00E-12	0.005074587	0.007181003	0.05170289	1	3.978304	-1893.082
DECJ	5	0.001505583	1.00E-12	0.005038266	0.006276902	0.03530849	1	4.985469	-1899.508
DECJ	6	0.00139243	1.00E-12	0.0045	0.007164745	0.04930684	1	5.961001	-1896.531
DECJ	7	0.001255576	1.00E-12	0.0045	0.007460751	0.04947936	1	6.948485	-1899.729
DECJ	8	0.001131741	1.00E-12	0.0045	0.007803152	0.05715238	1	7.961701	-1902.362
DECJ	9	0.001078691	1.00E-12	0.0045	0.006920693	0.0381892	1	9.004935	-1913.132
DECJ	10	0.000966464	1.00E-12	0.0045	0.007619308	0.05646538	1	9.969506	-1910.139
DECstarJ	1	0.002700736	1.00E-03	0.006	0.007177714	0.02573757	1	1.025952	-1918.734
DECstarJ	2	0.002279623	0.001	0.005673607	0.006939566	0.04826315	1	1.987043	-1901.462
DECstarJ	3	0.001958448	0.001	0.005293286	0.007002586	0.05032383	1	2.999908	-1893.234
DECstarJ	2.999908	0.003007244	0.03601728	0.001794544	0.006978496	0.04935522	1	2.998	-1871.215
DECstarJ	4	0.001728894	0.001	0.004740519	0.006771313	0.04955539	1	3.987107	-1891.465
DECstarJ	3.987107	0.002651285	0.03575712	0.001720951	0.007103985	0.05122847	1	3.989899	-1870.281
DECstarJ	5	0.001596427	0.001	0.004523937	0.005953594	0.03657859	1	4.961073	-1897.07
DECstarJ	6	0.001392406	0.001	0.0045	0.007416726	0.05019147	1	5.993699	-1894.942

Table 2.9. Continued.

Base Model	m_1 start	d	e	j	t_{12}	t_{21}	m_1	m_2	LnL
DECstarJ	7	0.001301236	0.00086213	0.0045	0.006852313	0.03612404	1	6.988153	-1904.598
DECstarJ	8	0.001155482	0.001	0.0045	0.008008623	0.0571904	1	7.980752	-1901.218
DECstarJ	9	0.001096718	0.001	0.0045	0.006632422	0.04459309	1	8.980964	-1908.28
DECstarJ	10	0.001060772	0.001	0.0045	0.006542139	0.03230808	1	9.992066	-1921.587
DivalikeJ	1	0.002930668	0.001	0.004623425	0.1953642	0.5446911	1	0.8809421	-2092.435
DivalikeJ	2	0.001887809	1.00E-12	0.3050096	0.4908273	1.160942	1	2.338847	-3321.761
DivalikeJ	3	0.001819263	1.00E-12	0.3851827	0.126953	0.6248587	1	3.1565	-3372.622
BayarealikeJ	1	0.001325264	0.001615015	0.01293775	0.2156505	0.3414732	1	0.4989322	-1957.47
BayarealikeJ	**0.4989322**	**0.001215443**	**0.001895003**	**0.01280095**	**0.06573549**	**0.4038765**	**1**	**0.4729849**	**-1791.896**
BayarealikeJ	2	0.001226079	0.001864485	0.01258462	0.1970513	0.7089999	1	0.5588565	-1814.364
BayarealikeJ	**0.5588565**	**0.001226004**	**0.001876434**	**0.01292706**	**0.1058423**	**0.6532477**	**1**	**0.4267553**	**-1791.747**
BayarealikeJ	3	0.001110642	0.001525956	0.04527725	0.02711513	0.492139	1	2.79996	-1990.138
BayarealikeJ	4	0.000550596	0.001144798	0.1613485	0.2041158	0.2640377	1	4.063048	-2900.101
BayarealikeJ	5	0.000381167	0.04193535	0.06137402	0.01304488	0.2333874	1	4.727855	-2587.381
BayarealikeJ	6	0.00033868	0.006187981	0.1978429	0.2455164	0.2712708	1	6.040394	-3211.147
BayarealikeJ	7	0.000391158	0.000827546	0.1916671	0.2386926	0.2753311	1	7.042728	-3131.127
BayarealikeJ	8	0.000296663	0.004973375	0.1968338	0.2441902	0.2713527	1	8.046162	-3207.204
BayarealikeJ	9	0.000315578	0.001684211	0.1799639	0.2457681	0.2734568	1	9.053256	-3118.079
BayarealikeJ	10	0.000259519	0.005788514	0.1986835	0.2472963	0.2763127	1	10.03777	-3232.716

Table 2.10. The BioGeoBEARS results under the wing length trait dispersal model. Dispersal rate (*d*), extinction rate (*e*), jump dispersal (*j*), transition rate from smaller wing length to larger wing length (t_{12}), transition rate from larger wing length to smaller wing length (t_{21}), multiplier on smaller wing length dispersal (m_1), multiplier on larger wing length dispersal (m_2).

Base Model	m_1 start	*d*	*e*	*j*	t_{12}	t_{21}	m_1	m_2	LnL
DECJ	1	0.002706514	1.00E-12	0.006	0.02006587	0.006470938	1	0.9828187	-1925.605
DECJ		0.00313513	1.00E-12	0.007816755	0.02079061	0.006407644	1	0.8118855	-1924.192
DECJ	2	0.001428979	1.00E-12	0.0045	0.01824518	0.006726596	1	1.997105	-1938.103
DECJ		0.00146847	1.00E-12	0.003454689	0.01803153	0.00673065	1	1.975889	-1936.538
DECJ	3	0.000957375	1.00E-12	0.0045	0.01753549	0.00679581	1	2.999454	-1956.124
DECJ	4	0.000715752	1.00E-12	0.0045	0.01257596	0.006801564	1	4.000769	-1977.307
DECJ	5	0.000565187	1.00E-12	0.0045	0.01238567	0.0068395	1	4.997984	-1998.49
DECJ	6	0.000477359	1.00E-12	0.0045	0.01354567	0.006729886	1	5.999351	-2019.647
DECJ	7	0.000404377	1.00E-12	0.0045	0.01036253	0.006981412	1	6.996406	-2042.206
DECJ	8	0.000345174	1.00E-12	0.0045	0.009750745	0.007767589	1	8.001591	-2064.241
DECJ	9	0.000312551	1.00E-12	0.0045	0.01005704	0.008218787	1	8.999636	-2085.368
DECJ	10	0.000330041	1.00E-12	0.0045	0.04986988	0.006949338	1	9.990477	-2072.102
DECstarJ	1	0.002705214	0.001	0.006	0.02005302	0.006489938	1	1.00464	-1923.357
DECstarJ	1.00464	0.003900617	0.01	0.004988903	0.02095468	0.006391429	1	0.7654257	-1907.975
DECstarJ	2	0.001455846	0.001	0.0045	0.01778763	0.006724531	1	1.995731	-1936.334
DECstarJ	1.995731	0.001727821	0.01	0.002070115	0.01799805	0.006719518	1	1.992381	-1920.754
DECstarJ	3	0.000972785	0.001	0.0045	0.01814978	0.006842335	1	3.005709	-1955.108
DECstarJ	4	0.000725559	0.001	0.0045	0.01247128	0.007030535	1	3.997963	-1976.544
DECstarJ	5	0.000579778	0.000991008	0.0045	0.01408392	0.006933735	1	4.99667	-1997.398
DECstarJ	6	0.000480775	0.001	0.0045	0.01389507	0.007154099	1	5.998883	-2019.071
DECstarJ	7	0.000411789	0.000980396	0.0045	0.01087576	0.007615717	1	6.999738	-2041.937

Table 2.10. Continued.

Base Model	m_1 start	d	e	j	t_{12}	t_{21}	m_1	m_2	LnL
DECstarJ	8	0.000362225	0.001	0.0045	0.01085963	0.008285445	1	7.998319	-2063.919
DECstarJ	9	0.000313279	5.85E-05	0.0045	0.01070317	0.008935182	1	8.999882	-2086.212
DECstarJ	10	0.000321067	0.000112399	0.0045	0.03554753	0.005675631	1	10.00298	-2078.419
DivalikeJ	1	0.006084498	1.00E-12	0.02577953	0.5008378	0.4610135	1	0.000640179	-2343.773
DivalikeJ	2	0.001196258	3.93E-05	0.4289435	0.02165922	0.01821854	1	2.269721	-3809.989
DivalikeJ	3	0.01204187	0.07227757	0.9890984	1.864444	0.08368333	1	1.351692	-5032.436
BayarealikeJ	1	0.00104381	0.001208025	0.1971719	0.07366999	0.009383004	1	0.9025865	-2557.156
BayarealikeJ	2	0.000432932	0.002549307	0.03072917	1.317098	0.7271876	1	2.271084	-2146.89
BayarealikeJ	2.271084	0.000920131	0.001849116	0.0096963	1.802386	0.4778149	1	1.289882	-1912.845
BayarealikeJ	3	0.000479825	0.001209034	0.235097	0.1966351	0.0287777	1	3.012606	-3268.322
BayarealikeJ	4	0.00049722	0.000815683	0.1981273	0.2530342	0.2374875	1	4.045882	-3236.553
BayarealikeJ	5	0.000521083	0.003442918	0.04589805	0.2750875	0.3073891	1	4.922718	-2508.699
BayarealikeJ	**4.922718**	**0.000246158**	**0.002637172**	**0.002787992**	**0.01683097**	**0.007069097**	**1**	**4.802716**	**-1763.228**
BayarealikeJ	6	0.000355413	0.000957009	0.2344064	0.2522084	0.2092635	1	6.03907	-3403.255
BayarealikeJ	7	0.000302266	0.001019038	0.2258551	0.2538272	0.215739	1	7.036373	-3389.243
BayarealikeJ	8	0.00028054	0.001198302	0.2100122	0.2545795	0.2167487	1	8.033661	-3340.948
BayarealikeJ	9	0.000255419	0.000836188	0.2196454	0.2467989	0.2066056	1	9.025376	-3382.642
BayarealikeJ	10	0.000224045	0.001088419	0.2325281	0.2537055	0.211798	1	10.03695	-3439.188

CONCLUSION

Chapter 1

I improved upon the popular historical biogeography inference method Dispersal-Extinction-Cladogenesis. Through simulations for parameter estimation of dispersal and extinction, I found that my method, DEC*, performed better in comparison to DEC, although it yielded results with more variance. The DEC* model also was selected as the best model for most of the empirical models tested, and DEC* does a more adequate job at estimating ancestral ranges than does the canonical DEC model. Given the results, I suggest that DEC* should be considered for use in biogeographic models.

Chapter 2

Through a wide variety of diversification methods, I was able to show that parasitism does not always influence a lower diversification in angiosperms, thus rejecting the evolutionary dead end hypothesis. The results by the HiSSE model suggested that another underlying trait also influenced the diversification of parasitic angiosperms. The hidden trait of interest or biological process could be tied to photosynthesis or photosynthetic potential for parasites that almost or completely lack chlorophyll. For species of parasitic plants that can still photosynthesize, the specialization to host type, range or mode of attachment in terms of whether they have access to sunlight or pollinators could influence their diversification.

Chapter 3

In this chapter, I used a novel approach to test how a trait could influence the dispersal rate of a group of perching birds, the passerines. I also tested morphology and region type as traits that could influence diversification. I found that the both wing length and region type affect diversification in conjunction with a hidden trait or biological process. I also found evidence rejecting my hypothesis that larger passerines have greater dispersal rate, when the results suggest smaller birds have greater rates of dispersal. Results from another study suggest wing area as being important in the influence of dispersal and diversification.

Future Directions

The methods I focused on using to test diversification and biogeography were primarily for discrete data. Most data is hard to discretize into categories. The field of diversification has a variety of model to test trait-dependent diversification using discrete and continuous traits. Unfortunately, methods in historical biogeography have focused on models which use arbitrarily pre-defined, discrete areas, indicating for each species its presence or absence. An example of a pre-defined area dataset is one that uses the Hawaiian archipelago, such as Hawaii, Oahu, Maui, and Kauai as discrete areas. Therefore, areas are discrete units of geographic range that are generally arbitrarily defined, and are analogous to discrete character states (Ree et al. 2005), such as parasitic or non-parasitic, or dioecious and monoecious. Despite the arbitrary designation of discrete states in biogeography studies, the use of discrete area state analyses in historical biogeography has been far more popular than continuous biogeography. This may be the case since most empirical biogeographic analyses deal with clades found in island systems, which are essentially discrete areas. However, if the clade of interest has a range within continents or islands, then discretizing oversimplifies the data.

To date, as I mentioned before, only one likelihood-based model exists that infers the biogeographic history of a clade (an ancestor and all of its descendants) on a continuous landscape while accounting for phylogeny (evolutionary relationships). If the clade of interest has a range between island systems then discretizing seems reasonable, yet if the clade exists within continents or islands, then discretizing oversimplifies the data**.** Biogeography models assume range evolution across a homogenous landscape. However, we know geography influences species ranges. To simply incorporate a heterogeneous topography on continuous landscape, a grid-matrix can be superimposed with dispersal occupancy probabilities into grid-based areas dependent on probability of dispersal into areas (see Figure 5.1). More complex models would account for ecological data and reconstruct environmental parameters through time and determine the probability of grid occupancy based on those parameter estimates. I suggest that a new method assessing the historical biogeography of taxa, using a continuous data framework, is needed to get more accurate inference.

My work shows that many parasitic plants have been understudied and work regarding their ecology, evolution and biogeography would be a substantial contribution to understanding symbiosis. Parasitic plants are interesting organisms to study because they are reliant on hosts for survival. Currently, no study has tried to assess the biogeographic history of parasitic plants and we lack understanding of how hosts and dispersal agents have influenced the biogeographic history of parasitic plants**.** We currently cannot explicitly answer question such as: What biotic factors have structured the historical biogeography of parasitic species? Does host and dispersal agent biogeography correlate with parasite biogeography? These questions, at best, currently can be answered through correlative studies, not through joint inference of these traits. I, therefore,

suggest more phylogenetic comparative methods to incorporate more ecological networks and processes to make the models more biologically realistic.

There were many more questions that would be interesting to ask, but we cannot due to lack of data. Does host range influence diversification? Are there more generalist or specialist parasites and how does this influence the diversity of the parasitic lineages? Data such as pollinator type, host range, biogeography was not available in a large-scale capacity to do comparative analyses. We hope future studies will focus on collecting more data of this type for parasitic angiosperms. We are aware that this is not an easy task, since there is a high possibility of ascertainment bias here – stem parasites are usually easily identified, but endoparasites and root parasites are not, which would show a higher rate of stem parasites over ones that live underground. Generating lists of host species is also difficult, especially for very generalist parasites that have a wide range of hosts. Incorporating uncertainty or studying a smaller, well-sampled clade would be something that should be further pursued if an analysis of this type is considered. There would be higher confidence in species that are sampled often.

My results show that large-scale studies are limited in the types of questions that can be asked. This is largely due to lack of data from many sources. For instance, as I stated above, there are only about one third of recognized land plants that have sequence data available. The rate of adding sequences to GenBank has stayed about the same since the mid-1990s, and assuming this same rate of increase, it would take until the year 2044 to reach the minimum number of estimated living land plants (300,000). There is just so much diversity, and few scientists in comparison to study everything in depth. This could also be due to many scientists developing many great methods and undergoing more comparative work, but we need more naturalists. We need more scientists to go out and explore the world, work in the laboratory and

collect reliable and accurate data, so we can understand the world around us much better. And with that, I will end.

Appendix E

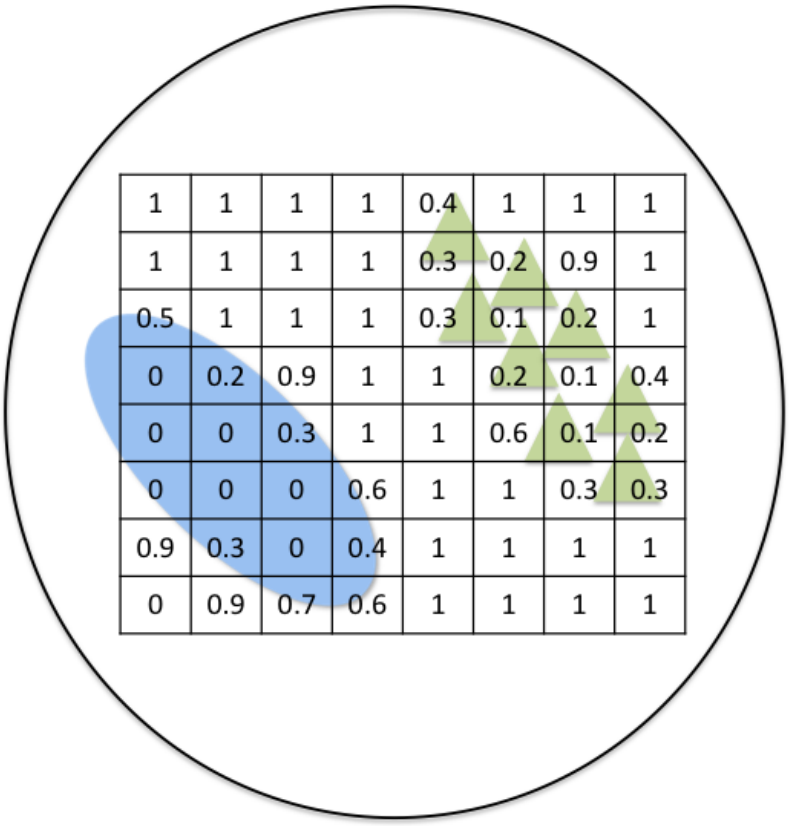

Figure 5.1. A simplified illustration of a superimposed grid matrix with dispersal/occupancy probabilities on a continuous landscape. The grid matrix allows the incorporation of heterogeneous topography. Dispersal/occupancy probability decreases as barriers are found in the landscape. A model like this has not been developed.

VITA

Kathryn Aurora Massana Baxley was born in San Juan, Puerto Rico in 1990. She is the first born, only child, oldest child, middle child and youngest child of Barbara Sue Baxley and the late, Jorge Carlos Massana Fajardo. She has six siblings. At a young age, she discovered her inherent interest to explore and discover the world. She went to La Academia San José in Guaynabo, Puerto Rico until she transferred to Madison High School in Asheville, North Carolina to finish her secondary education. She graduated from Berea College in May 2012 with a Bachelor of Arts in Biology. She studied abroad and engaged in several research projects, which led her to graduate school where she could pursue her own research interests. She completed her Master of Science in Statistics in December 2016 and Doctor of Philosophy in Ecology and Evolutionary Biology in May 2017 at the University of Tennessee. She served as a graduate teaching assistant for the Department of Ecology and Evolutionary Biology and a graduate research assistant for the Program for Excellence and Equity in Research and the National Institute for Mathematical and Biological Synthesis during her graduate career.