Diversification and Character Trait Analyses of the Core Goodeniaceae

A Senior Thesis Presented to

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By

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Introduction

The Goodeniaceae family has more than 420 formally described species and plays a major role in the makeup of the floristic landscapes of Australia. This family is spread throughout the entire Australian continent, across all biomes from the northern monsoonal forest to the arid Eremaean zone throughout the interior. The Southwest Australian Floristic Region (SWAFR) is one of the largest biodiversity hotspots for this family. While the majority of the family is endemic to Australia, *Scaevola* has consistently and effectively established populations on islands and coastlines throughout the Pacific, Atlantic, and Indian Oceans, with some dispersal events allowing for subsequent speciation events leading to island endemic species, particularly in Hawaii (Howarth & Baum, 2005).

The Goodeniaceae has 7 described genera and is split into two major clades, the Core Goodeniaceae and the LAD clade (Jabaily et al., 2012). The Core Goodeniaceae is made up of *Coopernookia, Goodenia,* and *Scaevola* and is sister to the genus *Brunonia*. These four genera are sister to the rest of the family, the LAD clade, which is comprised of the genera *Lechenaultia, Anthotium,* and *Dampiera*. Broader phylogenetic analyses of the Goodeniaceae shows the family sister to Asteraceae + Calyceraceae, with all three families further sister to the family Menyanthaceae (Tank & Donoghue, 2010). The taxonomic relationship of *Brunonia* to the rest of the family has been challenging, as it has been both included in Goodeniaceae and considered its own family multiple times in its history based on morphological traits. It is currently considered a monotypic genus in Goodeniaceae following a revision in 2002 (Cayzer et al., 2002). Jabaily et al. (2012), placed *Brunonia australis* with 100% support within the family sister to the Core Goodeniaceae clade.

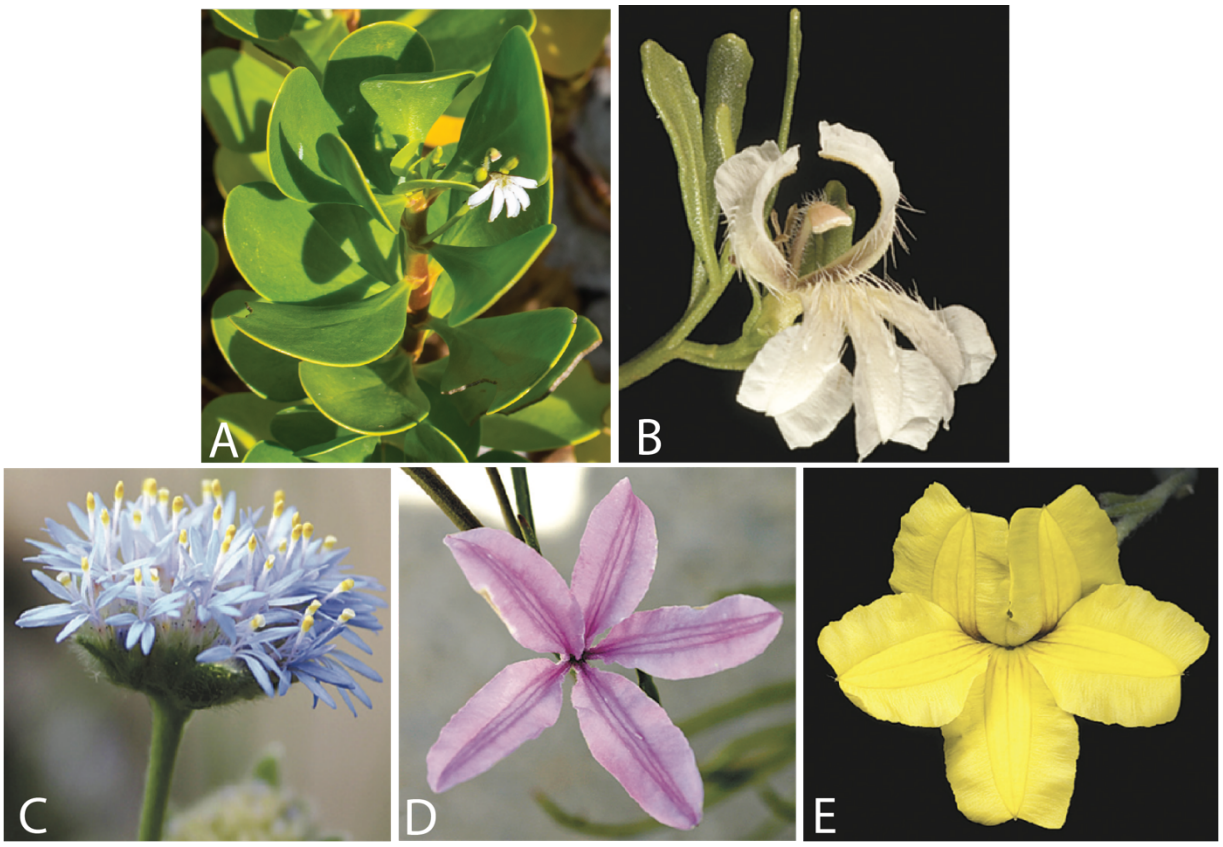


Figure 1. Images representing Goodeniaceae genera. **A**, *Scaevola plumieri*; **B**, *Coopernookia strophiolata*; **C,** *Brunonia australis*; **D**, *Scaevola filifolia*; **E,** *Goodenia convexa*. Photos – A by C. Brose; C, D by K.A. Shepherd, (Jabaily et al., 2012); B, E by K.R. Thiele, (Jabaily et al., 2012)

The significant defining feature for this family is the presence of the stylar indusium (Figure 1B), a cup-like structure on the style which packs pollen for secondary-pollen presentation (Jabaily et al., 2012). The wide range of diversity amongst other morphological traits, especially in floral structures (Figure 1), is worthy of deeper study as multiple independent evolutions and reversions back to ancestral states across the traits occur. Most species in this family have capsular dry fruits, but multiple evolutions of fleshiness and a reversion back to capsular fruits have occurred in *Scaevola*, as well as independent origins of fan-flowered symmetry in every major clade of the Core Goodeniaceae (Howarth et al., 2003; Gardner et al., 2016a) but no comparative methods have been used to analyze these traits.

The differing floral symmetry within the Goodeniaceae is an important character of interest and has been formally analyzed in a morphometrics study (Gardner et al., 2016a), which found that the fan-flower shape (Figure 1A) is significantly different from the other morphologies (Figure 1B-E). *Scaevola* is almost entirely fan-flowered, while *Goodenia* has only one clade, Clade C, plus a few species in Clades A and B, with this symmetry, as most of the genus is bilabiate or pseudoradial (Jabaily et al., 2012; Shepherd et al., 2020). Previous Goodeniaceae studies suggested that greater floral morphology diversity may drive additional speciation by employing a greater diversity of pollinators (Jabaily et al., 2014). The vegetative habit of the Goodeniaceae is also of interest as *Scaevola* is primarily woody while *Goodenia* has only one small clade of mostly shrubby taxa with the rest nearly all herbaceous. The habit and life history strategy of plants tends to play a role in the diversification rates, as herbaceous annuals have a much shorter generation time, allowing more accumulation of molecular change over the same period as woody perennials. However, this contradicts previous studies in the family which show a rapid burst of speciation in *Scaevola* with a much slower rate of molecular evolution in *Goodenia* (Jabaily et al., 2012).

The genus *Scaevola* is unique in the Goodeniaceae due to its ability to disperse across oceans and settle on islands throughout the Pacific, Atlantic, and Indian Oceans, mediated by its repeated evolution of fleshy fruits (Figure 2). *Scaevola* is still taxonomically circumscribed as it was by Carolin et al. (1992) with two exceptions: the exclusion of *S. collaris* (now part of the genus *Goodenia*), and the inclusion of the newly dissolved monotypic genus *Diaspasis* which adds *S. filifolia* to the genus, following the Shepherd et al. (2020) taxonomic revision. While *Scaevola* is primarily Australian, with 74 out of its 102 described species endemic, the dispersal of *Scaevola* species across islands, especially in the Pacific, was driven by the evolution of fleshy fruits. Nearly every extra-Australian *Scaevola* has this trait (Howarth et al., 2003), which has allowed for each species or its fleshy-fruited ancestor to travel across oceans both by floating through open ocean and through avian guts. *S. taccada* and *S. plumieri* are the two most effective species at long-distance dispersal, both with ranges

Figure 2. Fleshy fruits in *Scaevola plumieri.* Photo – C. Brose, Bill Baggs Cape Florida State Park, Miami, FL

across the tropics in the northern and southern hemispheres (Howarth et al., 2003). Previously, six independent events were identified where *Scaevola* dispersed outside Australia, two of which had subsequent radiations and the remaining four leading to single species (Howarth et al., 2003), however these conclusions may be limited by the exclusion of many Australian endemic *Scaevola*.

The infrageneric taxonomy of *Scaevola* was defined by Carolin (1990), based on morphological characters. The genus *Scaevola* has three sections – *Scaevola, Enantiophyllum,* and *Xerocarpa*. Section *Xerocarpa* is the only section with subsections, of which there are three: *Pogonanthera*, *Parvifoliae*, and *Biloculatae*. *Scaevola* sect. *Xerocarpa* subsect. *Biloculatae* has two series, *Globuliferae* and *Pogogynae* (Carolin, 1990). The molecular evidence from Howarth et al. (2003) only supports the monophyly of one out of the three sections defined by Carolin et al. (1990). *Scaevola* sect. *Enantiophyllum* is the only section with monophyly supported by both molecular data and morphological traits unique to the group. This section includes only two species, *S. enantophylla* and *S. oppositifolia*, both with the defining characters of opposite leaves and a vining habit. These species have fleshy fruits and distributions throughout Papua New Guinea and Indonesia, with *S. enantophylla* also extending through northern Australia and *S. oppositifolia* into the southern Philippines (Carolin et al., 1992; GBIF, 2022). All but five species of *Scaevola* (*S. gracilis*, *S. beckii*, *S. angulata*, S. *oppositifolia*, and *S. enantophylla*) with distributions ranging outside of Australia, as well as six endemic species, are included within *Scaevola* sect. *Scaevola*. This section is defined by fleshy fruits and shrubby to small tree habits with axillary inflorescences (Carolin et al., 1992). However, there are multiple species within this section with terminal inflorescences, which is a major defining character of the third section, *Xerocarpa*. *Scaevola* sect. *Xerocarpa* contains the remaining species of *Scaevola* and is additionally characterized by dry fruits and habits ranging from herbaceous to small shrubs. Although most species in this section have dry fruits, a few species, such as *S. gracilis* and *S. beckii,* have fleshy fruits. *S. gracilis* is included in sect. *Xerocarpa* due to its terminal inflorescences, but *S. beckii* has none of the defining characters for this section (Howarth et al., 2003). This mismatch between character traits among species, as well as the lack of monophyly for the two large sections of *Scaevola*, indicates the need for additional study of the sub-taxonomic grouping within the genus.

The topology of *Goodenia* is highly resolved, except for *Goodenia* Clade C, following the efforts in Shepherd et al., (2020), Jabaily et al., (2018), and Gardner et al., (2016b). *Goodenia* Clade C is difficult to untangle as the relationships between smaller clades within it are poorly supported. However, there is strong support for *Goodenia* Clade C and the infrageneric taxonomy discussed and formalized in Shepherd et al., (2020). The Jabaily et al. (2012) study identified major polytomies in the backbone of *Scaevola* but there have been no further efforts to increase sampling to mirror the work done in *Goodenia*, nor resolve the lack of support in relationships among these genera since. The phylogram produced in that study shows a very short and explosive speciation within *Scaevola sensu lato* (s.l.) and a much slower diversification in *Goodenia* s.l., and the disparities in rates of molecular evolution are evident (Jabaily et al., 2012). Molecular dating analyses were done by Jabaily et al. (2014) in the first study of the family in geologic time using a fossil from before the Asteroideae, which is a subfamily of the Asteraceae, dated to 47.5 million years ago (Mya) (Barreda et al., 2010). Following discovery of a new pollen fossil in the Asteraceae, a 2015 study published new revised molecular dates for the early evolution of the family and its close relatives, including members of the two major clades of the Goodeniaceae (Barreda et al., 2015).

There have been no formal analyses within the family to untangle the speciation rates and test hypotheses regarding the faster diversification in *Scaevola* s.l. compared to that of *Goodenia* s.l., nor have there been character trait analyses to compare correlations between specific trait evolution and subsequent shifts in speciation. Despite the lack of backbone support in *Scaevola* s.l., we consider these types of analyses to be important and necessary for understanding the evolutionary history of the Goodeniaceae as the topology can be resolved with additional genetic data after an initial run of diversification and character trait evolution analyses identify the gaps that would be filled with additional data and full resolution in the backbone phylogeny. In our study, we seek to 1) enhance genetic sampling of *Scaevola* by including new species and unpublished sequences in the phylogeny, 2) formally score traits and test their connection to increased diversification, 3) revisit the subgeneric grouping of *Scaevola*, and 4) update placement of clades in geologic time with newly calibrated molecular dating.

Methods

Taxon sampling and alignment

Taxon sampling includes 278 species in the Goodeniaceae, out of c. 420 described species, from *Brunonia*, *Coopernookia*, *Goodenia*, and *Scaevola*, representing all genera in the Core Goodeniaceae clade, as described in Jabaily et al., 2012 (Table 1). In addition, *Dampiera loranthifolia* was used as an outgroup from the LAD clade of the Goodeniaceae, which is sister to the Core Goodeniaceae + *Brunonia* (Jabaily et al., 2012). This study's sampling includes 117 taxa with unpublished sequences of either ITS or ITS and *trnL-F* loci, including 9 species, 1 *Goodenia* and 8 *Scaevola*, previously never included in a phylogeny. Accessions of *trnL-F* and ITS loci sequence data were acquired from Howarth et al. (2003) via GenBank, Jabaily et al. (2012), and unpublished data from Dr. Kelly Shepherd (Western Australian Herbarium). When multiple individuals were sequenced for a species, the new taxon with the longest sequence length with fewest ambiguities was chosen. For those with both loci from the same specimen, sequences were concatenated then aligned using the MAFFT aligner (Katoh & Standley, 2013) in Geneious Prime (Biomatters Ltd., 2005) and hand-checked for consistency.

Genera	**Number of described species**	**Number of species included**
Dampiera (outgroup)	66	1
Brunonia	1	1
Coopernookia	6	3
Goodenia	c. 251	196
Scaevola	102	77

Table 1. Number of species sampled in this study and total number of species in each genus across Goodeniaceae family.

Bayesian inference phylogeny

The two loci were tested separately to determine the best model for molecular evolution using jModelTest v. 2.1.10 (Darriba et al., 2012; Guindon & Gascuel, 2003). The test suggested

that the best model for *trnL-F* was general time reversible with invariable sites and gamma distribution (GTR + I + G) and the best model for ITS was the transitional model with equal base frequencies (TIM2ef + I + G). The TIM2ef + I + G model was replaced with the GTR + I + G model for ITS following recommendations from Lecocq et al. (2013). As both loci were found to have the same best fit model and independent analyses of each locus did not yield topological conflict, we then concatenated the alignment and analyzed each locus with the same model. A Bayesian inference tree with the concatenated alignment was made using MrBayes v.3.2.2 (Ronquist & Huelsenbeck, 2003) on the software portal Cyberinfrastructure for Phylogenetic Research (CIPRES; Miller et al., 2010). The analysis was run twice for 100,000,000 generations, with sampling every 10,000 generations using the GTR + I + G model. The backbone topology was constrained, following published topologies (Jabaily et al., 2012), to keep *Goodenia* monophyletic. The first 50% of each run was discarded as burn-in before being combined and used to produce a consensus tree in Geneious. All nodes with less than 50% consensus support were collapsed into a polytomy.

Molecular dating

Using the new dates acquired from the Barreda et al. (2015) study, a Bayesian Evolutionary Analysis by Sampling Trees (BEAST) v. 1.10.4 (Drummond & Rambaut, 2007) was performed on CIPRES for molecular dating. The input .xml file for BEAST was generated using the graphical user interface Bayesian Evolutionary Analysis Utility (BEAUti) v. 1.10.4 (Drummond & Rambaut, 2007), with dates for the stem of Core Goodeniaceae + *Brunonia* fixed at 68.1 Mya with 95% highest posterior density (HPD) between 30.8 and 91.1 Mya (Barreda et al., 2015). The crown of the Core Goodeniaceae was anchored at 25.7 Mya with 95% HPD between 5.2 and 52.4 Mya (Barreda et al., 2015). The dataset was partitioned into the two separate loci, with both sets sampled using a lognormal model with the Yule speciation process prior and uncorrelated relaxed clocks. Following the BEAST analysis, priors were checked in Tracer v. 1.7.2 (Rambaut et al., 2018) to ensure quality of the analyses, then the first 10% of trees were discarded as burn-in with TreeAnnotator v. 1.10.4 (Drummond & Rambaut, 2007) before being visualized in FigTree v. 1.4.4 (Rambaut 2018). A second BEAST analysis was run excluding the outgroup *Dampiera* and monotypic genus *Brunonia* and forced monophyly of *Goodenia* for subsequent character trait and diversification rate analyses. The ages for this analysis set the crown of *Scaevola* to 13.7 Mya (95% HPD 8.4 – 19.0 Mya), following estimates from the first BEAST analysis, with all other methods consistent with the previous run. Node ages from this analysis, containing only the Core Goodeniaceae, are used for downstream analyses and conclusions due to the enforced monophyly of *Goodenia*.

Character trait analyses

Character traits for floral symmetry, fruit form, and habit were scored binary for each species using species descriptions in *Flora of Australia* (Carolin et al., 1992), taxonomic grouping and descriptions (Shepherd et al., 2020), herbarium specimen photographs from the Global Biodiversity Information Facility (GBIF, 2022), and FloraBase (Western Australian Herbarium, 1998). Floral symmetry was scored as fan-flowered or not fan-flowered, following a morphometrics study which found a significant difference between fan-flower symmetry and all other symmetries found in the Goodeniaceae, primarily bilabiate or pseudoradial (Gardner et al.,

2016a). Each species' fruit form was considered either fleshy or dry, following descriptions found in *Flora of Australia* (Carolin et al., 1992) and Howarth et al. (2003). While some species are considered "partially fleshy" in the Howarth et al. (2003) study, we consider this to be fully fleshy for sake of model simplicity. Habit was scored as a very simple "herbaceous" or "not herbaceous", but more assumptions had to be made while scoring. For example, the two taxa with true vining habits, *Scaevola enantophylla* and *S. oppositifolia*, were scored as "not-herbaceous." Additionally, many herbaceous perennial species in the family are known to have woody growth underground (Shepherd et al., 2020), but we consider these "herbaceous."

Relative transition rates between the two states for each character trait were calculated using an MCMC statistics analysis in BayesTraits v. 3 (Pagel & Meade, 2006). The bifurcating ultrametric tree produced in BEAST and a matrix for character states for the trait of interest were used as the input file with the MCMC running for 1,000,000 generations, sampled every 1,000 generations, and the first 1% discarded as burn-in. Output rates in Q-matrices were averaged across all sampled generations and compared to the rate of transition in the opposite direction. Relative transition rates were visualized using arrows with widths corresponding to the rates from one state to another. Ancestral state reconstruction was done in Mesquite (Maddison & Maddison, 2021) using the cloud of several thousand post burn-in trees generated by MrBayes before consolidated into a consensus tree depicting scored character traits for each taxon and ancestral states.

Diversification rate analysis

Bayesian Analysis of Macroevolutionary Mixtures (BAMM; Rabosky et al., 2015) was used to analyze diversification rates in the Core Goodeniaceae. Initial program analyses were run in the command-line program, while the visualization and additional analyses were done with the R package BAMMtools v. 2.1.8 (Rabosky et al., 2014). The BEAST analysis of the Core Goodeniaceae, excluding *Brunonia* and *Dampiera*, was the input file used, with priors for the run estimated using BAMMtools and then run for 10,000,000 iterations. All other parameters in the BAMM control file are kept consistent with the suggested starting values in the manual. The output event file was analyzed in R to test for MCMC convergence and generate a phylorate plot to visualize net diversification rates. Net diversification was calculated as the difference between speciation and extinction rates. The significant rate shifts were added to the plot and 95% credible set of shift configurations with the probability for each was generated to show the shifts that makes up the 95% majority of the data.

Results

Topology

The topology within the genus *Goodenia* (Figure 3) closely follows the resolved topology in Shepherd et al. (2020). *Goodenia paniculata* is the only new species of the genus that has never been sequenced and placed in a phylogeny. This taxon is within a 100% supported polytomy with *G. bellidifolia*, *G. decurrens*, *G. racemosa var. racemosa*, and *G. glomerata* + *G. dimorpha* var. *angustifolia* and *G. stelligera*. Shepherd et al., (2020) has all these above taxa save *G. paniculata* within the clade that defines the subgenus *Monochila* section *Tetrathylax*. However, *G. paniculata* was placed within subg. *Porphyranthus* sect. *Porphyranthus*, due to

shared morphological characters, even though it was not sequenced. This suggests a potential deeper look into the sub-taxonomic grouping of *G. paniculata*.

Coopernookia is highly resolved and the relationships between species in the genus are highly supported. This is also reflected in Figure 3, with nearly 100% support following the topology represented in Jabaily et al. (2012). *Brunonia*, the monotypic genus, is similar, with its placement sister to the Core Goodeniaceae clade.

Scaevola, however, has not undergone any phylogenetic work since Jabaily et al. (2012), and the only taxonomic revisions were in Shepherd et al. (2020), where the monotypic genus *Diaspasis* was synonymized into *Scaevola* (Figure 3, Clade A), and *S. collaris* was renamed and included in *Goodenia*. The new unpublished sequences used in this study have begun to resolve into major clades. The placement of multiple species from *Scaevola* into subclades that were previously part of the large backbone polytomy prior to sequencing of the nrITS locus (Figure 3).

This tree places *Scaevola basedowii*, which has previously not been included in a phylogenetic study, sister to *S. depauperata*, while the clade these two taxa are in (Clade B, Figure 3), excluding *S. parvibarbata*, was considered sister to the rest of *Scaevola* in Jabaily et al. (2012). Here, this clade is instead included within the major backbone polytomy of *Scaevola* s.l., whereas *S. parvibarbata* was previously placed sister to *S. ovalifolia* with 100% posterior prior support (Jabaily et al., 2012). *Scaevola bursariifolia* and *S. spinescens* have been noted to have very similar morphologies (Carolin et al., 1992), but following the new addition of *S. acacioides* to the taxon sampling, we found *S. bursariifolia* is sister to *S. spinescens* + *S. acacioides* (Figure 3, Clade D3). *Scaevola basedowii* has been placed sister to *S. depauperata* with 100% bootstrap consensus support. Additionally, these taxa are sister to *S. angulata* + *S. restiacea* with 97% support. These are all part of a fully supported clade with *S. hamiltonii* + *S. parvibarbata* (Figure 3, Clade B). *Scaevola kalophylla* is decisively sister to *S. oldfieldii*. These taxa are part of a polytomy with *S. sericophylla* and *S. humifusa* + *S. paludosa* + *S. repens* (Figure 3, Clade D2), which is consistent with the topology of Jabaily et al. (2012). *Scaevola oxyclona* is highly supported as sister to *S. aemula* (97%) and more distantly related to *S. argentea* (Figure 3, Clade C). This provides greater resolution, with 100% support, than in the Jabaily et al. (2012) study, as *S. aemula* and *S. argentea* were previously part of the large backbone polytomy of *Scaevola* s.l. *Scaevola macrostachya* and *S. pulchella* are both newly placed in the large backbone polytomy of the genus (Figure 3, Clade D).

Diversification rates and ancestral character state reconstruction

The net diversification rate analysis in BAMM produced a phylorate plot (Figure 5) that is colored on a temperature scale to show relative rates of diversification across the family. The rates for *Scaevola* are significantly higher compared to those in *Goodenia*, especially in *Goodenia* Clade B (Figure 5). Two significant shifts in diversification rate were identified, one along the stem of *Goodenia* Clade A + Clade B and the other embedded within *Goodenia* Clade C. *Goodenia* Clade B shows a rapid increase in diversification at its crown followed by a dramatic decrease to the present. Ancestral state reconstruction for floral symmetry, fruit type, and vegetative habit in Mesquite sought to contribute explanations for the diversification rate changes in *Scaevola* and *Goodenia* Clade B (Figures 6A-C). Multiple independent origins of fleshy fruits are obvious within *Scaevola* (Figure 6A), with the ages of these nodes added from the BEAST analysis. Node I in Figure 6A is labeled with a range of ages to account for the uncertainty in relationships determined by Mesquite. The topology of the trees generated by

Mesquite and BEAST are slightly different, so we cannot predict the age of node I without it being younger than the nodes that follow it. There are also two tips that show a switch to fleshy fruits that we did not label with ages which is also due to the uncertainty of the topology between the two programs. Despite this, each of the labelled nodes suggest recent evolution of fleshy fruits, all of which occur within the same 8-million-year period.

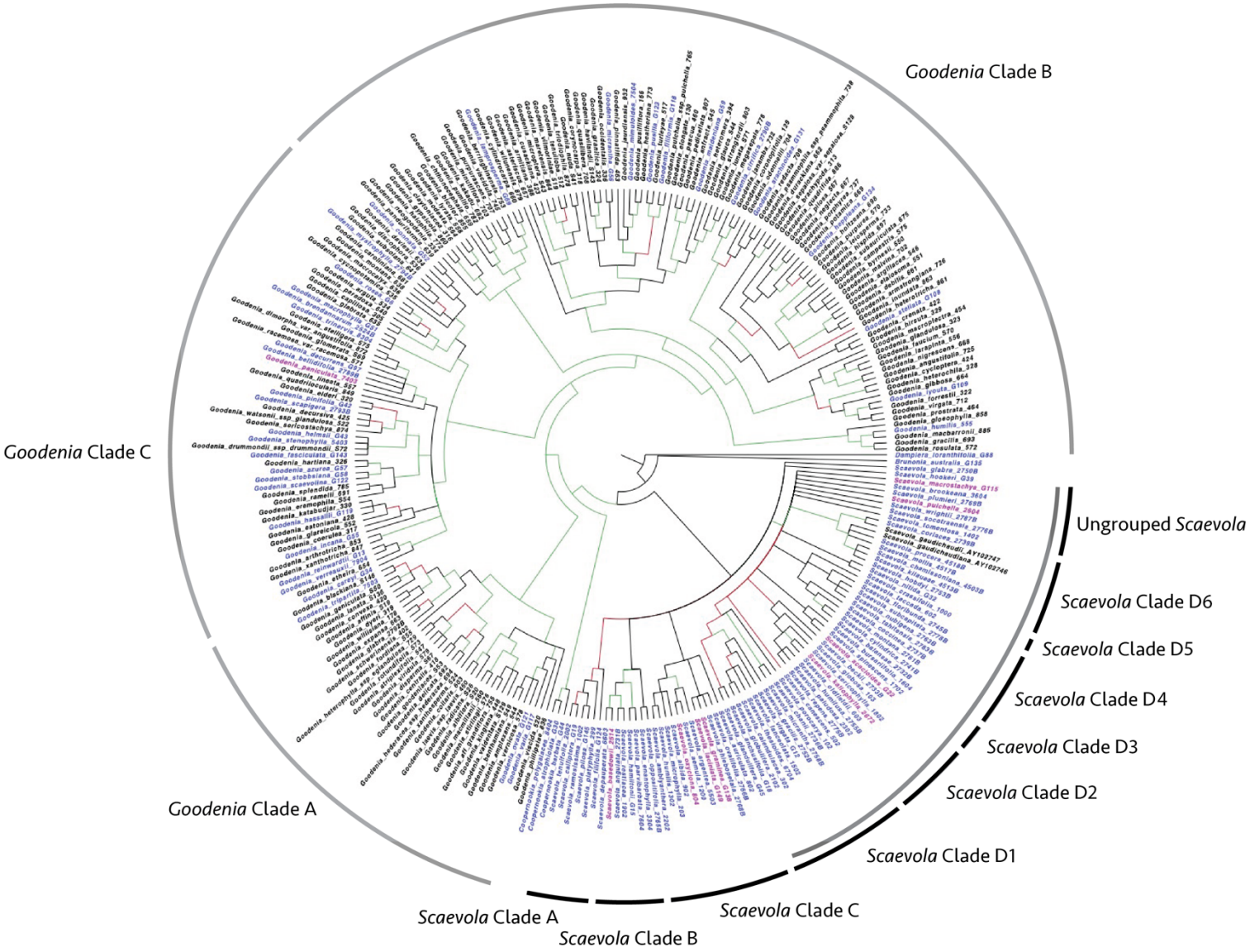
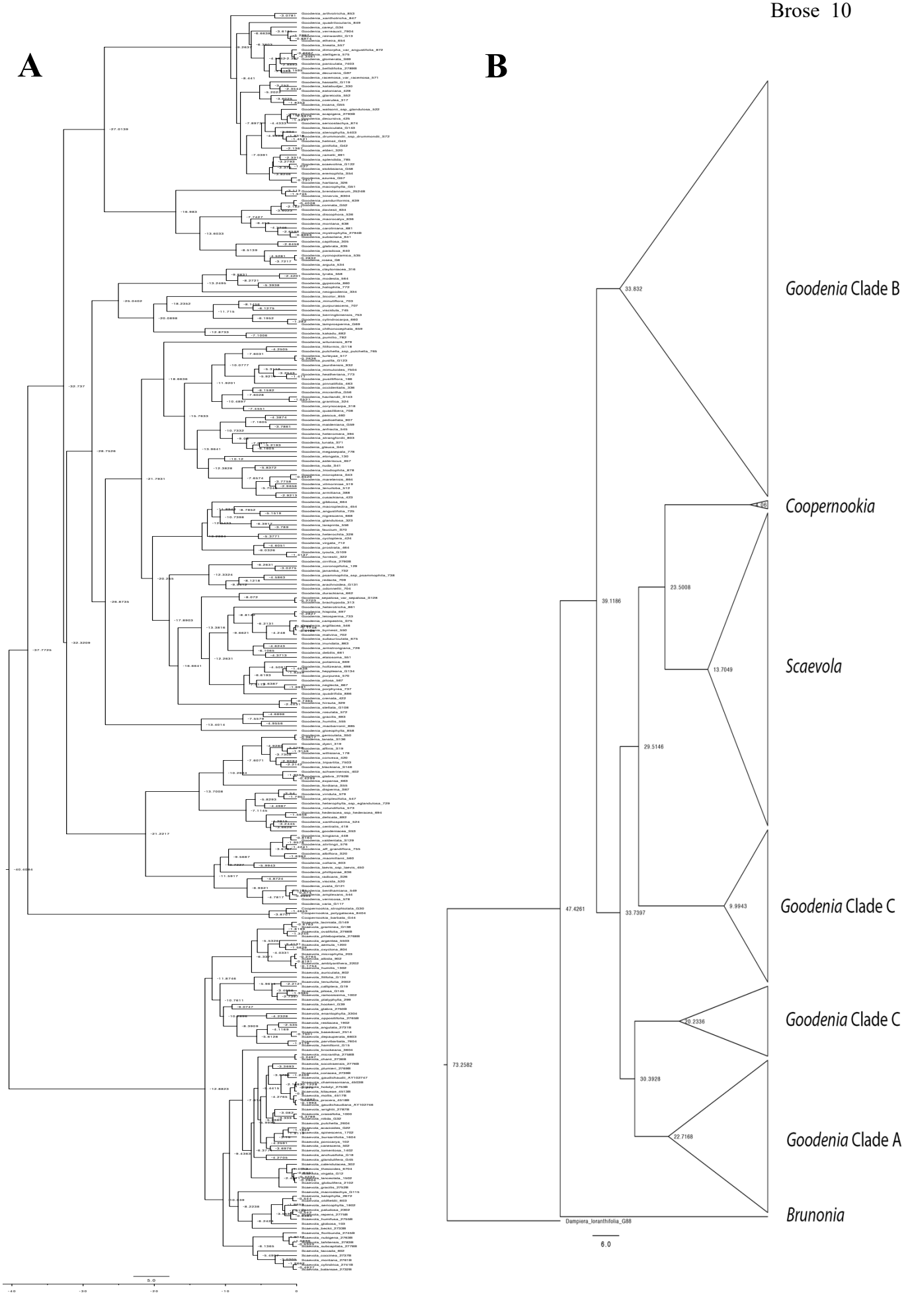


Figure 3. Topology and clade names for concatenated *nrITS* and *trnL-F* Bayesian inference tree of Core Goodeniaceae clade, with *Dampiera loranthifolia* as outgroup. Green branches represent ≥ 98% consensus support for topology, red branches show ≤ 65% support, and < 50% supported nodes collapsed into polytomies. Taxa in blue show unpublished sequences from K.A. Shepherd or R.S. Jabaily; taxa in purple show species never included in phylogenetic studies.

Figure 4. BEAST analysis of molecular dating, node ages in millions of years. **A**. Molecular dating of Core Goodeniaceae – excludes *Brunonia* and *Dampiera* outgroup. Monophyly of *Goodenia* forced with constraints, ages in millions of years before present for node calibration estimated from Fig. 4B. **B**. Whole taxon sampling BEAST tree, major clades collapsed and labelled with node ages. Calibration dates from Barreda et al. (2015) to root crown of *Scaevola* s.l. + *Goodenia* s.l.


A
B
Goodenia Clade B
Coopernookia
Scaevola
Goodenia Clade C
Goodenia Clade C
Goodenia Clade A
Brunonia
33.832
4.06
23.5008
13.7049
39.1186
29.5146
33.7397
9.9943
47.4261
20.2336
30.3928
22.7168
73.2582
Dampiera_loranthifolia_G88
6.0
5.0

Molecular dating

The two BEAST analyses show similar but slightly different values for the crown nodes of each of the four major clades in the Core Goodeniaceae: *Scaevola,* and *Goodenia* Clades A, B, and C. In general, the second molecular dating analysis (Figure 4A), which excludes *Dampiera* and *Brunonia*, shows crown ages younger than the original (Figure 4B) by 1-2 million years, apart from *Goodenia* Clade C. We estimate the origin of this clade to be approximately 27.0 Mya, consistent with the ages from Figure 4A, as *Goodenia* is not monophyletic in Figure 4B, where Clade C is paraphyletic with half of the clade sister to Clade A and the other half sister to *Scaevola* + *Coopernookia*. The node ages in our BEAST analyses were calibrated with new fossil dates (Barreda et al., 2015) that had not been published at the time of the last molecular dating in the Goodeniaceae (Jabaily et al., 2014). All ages are within 4-6 million years between the previous molecular dating and our new analysis, with the crown of Goodeniaceae, *Goodenia* s.l. crown, and the *Scaevola* s.l. + *Goodenia* s.l. crown all slightly older than in the Jabaily et al. (2014) study, and both the crown of the Core Goodeniaceae and the *Scaevola* s.l. crown slightly younger than the previous dates.

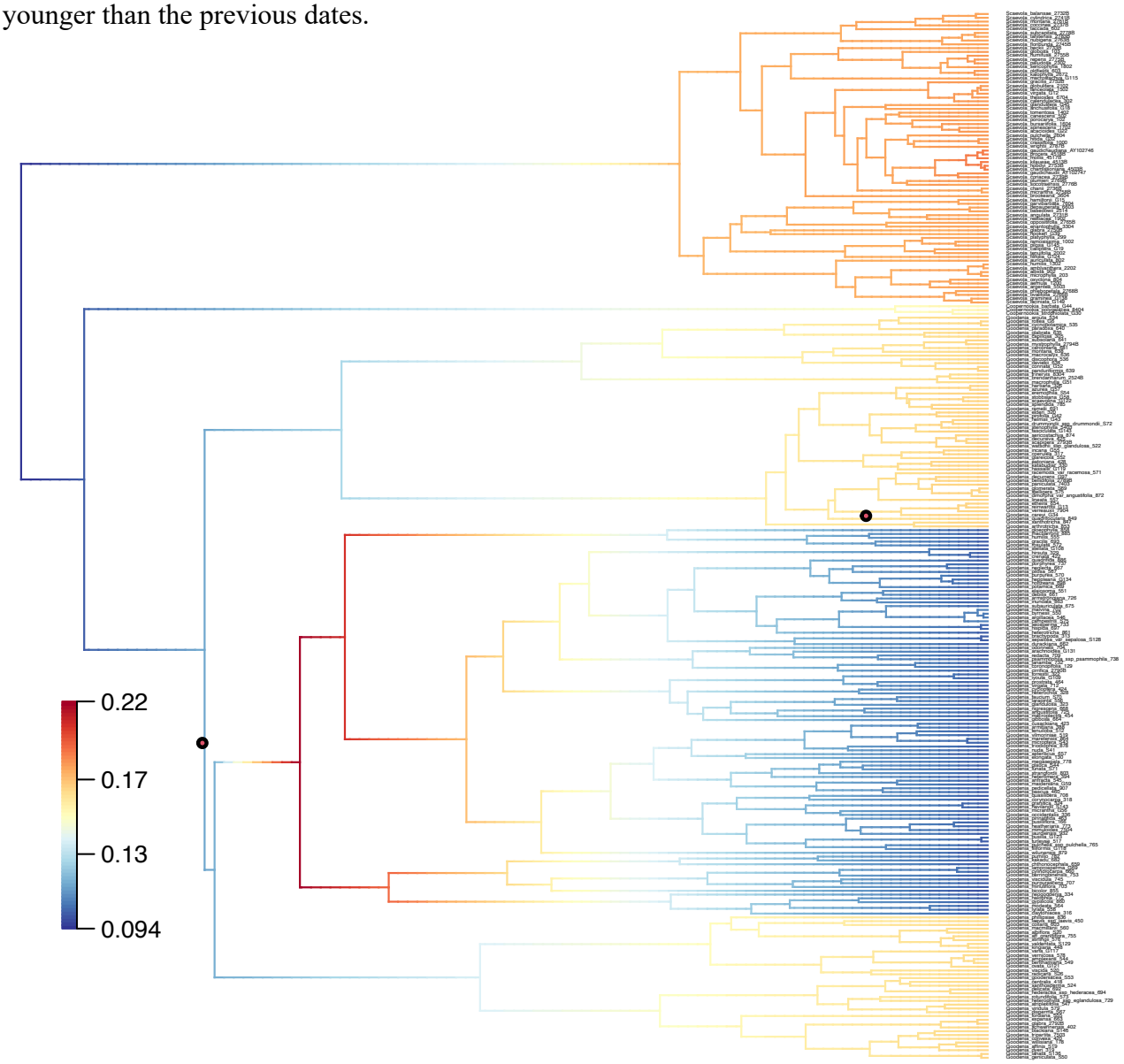


Figure 5. BAMM analysis of net diversification rates in Core Goodeniaceae visualized in R with BAMMtools, excludes *Dampiera* and *Brunonia*. Red dots on branches represent significant shifts in diversification. The input ultrametric tree comes from Figure 4A.

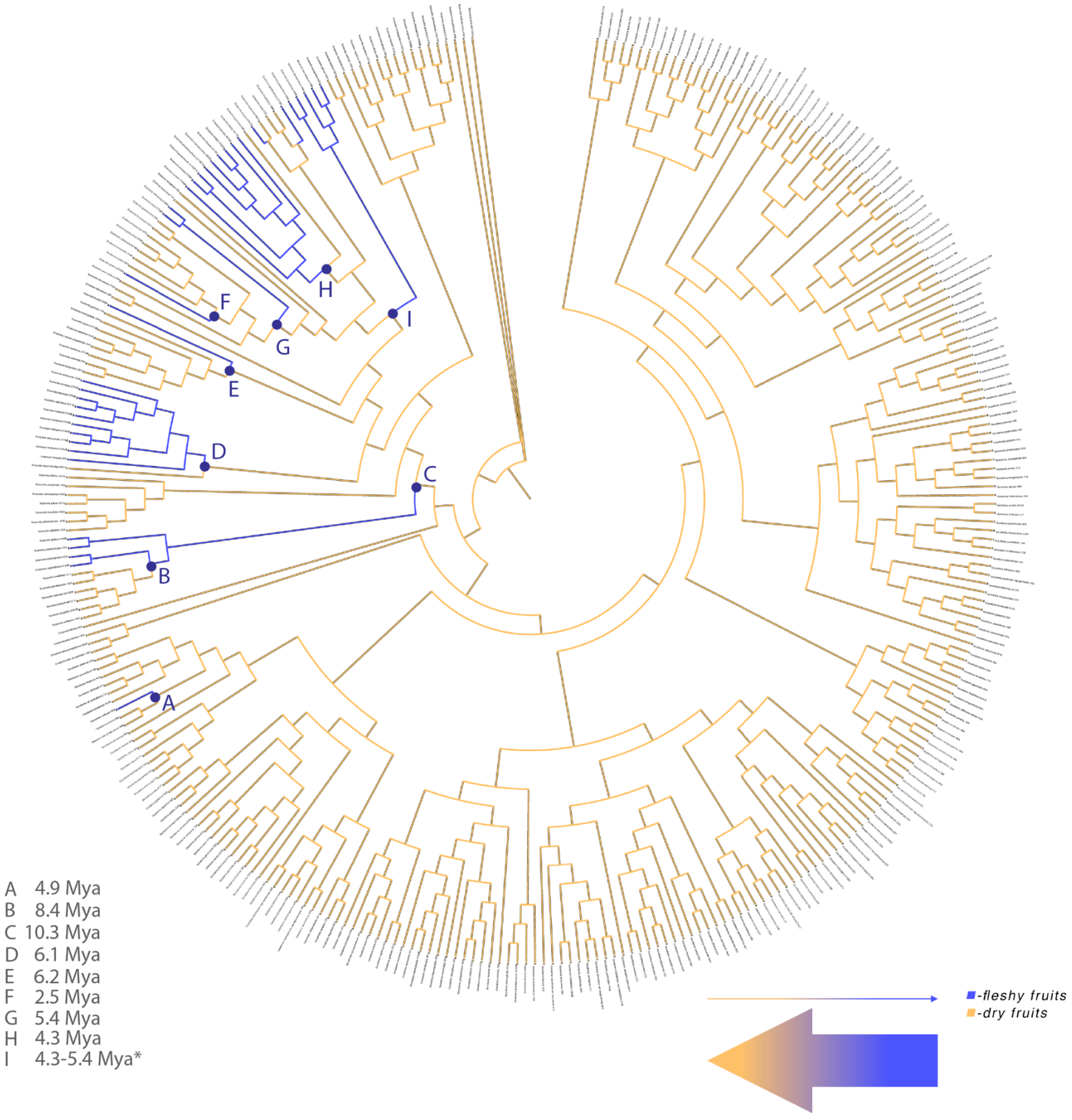


Figure 6A. Ancestral state reconstruction in Mesquite for fleshy fruits across the Core Goodeniaceae. Blue branches show fleshy fruits in ancestors or extant taxa, brown branches show dry fruits. Relative transition rates from one state to the other calculated in BayesTraits shown by arrows in bottom right. Node ages correspond to molecular dating from BEAST analysis (Fig. 4A). For explanation of age range at node I, see Results section "Diversification rates and ancestral character state reconstruction."

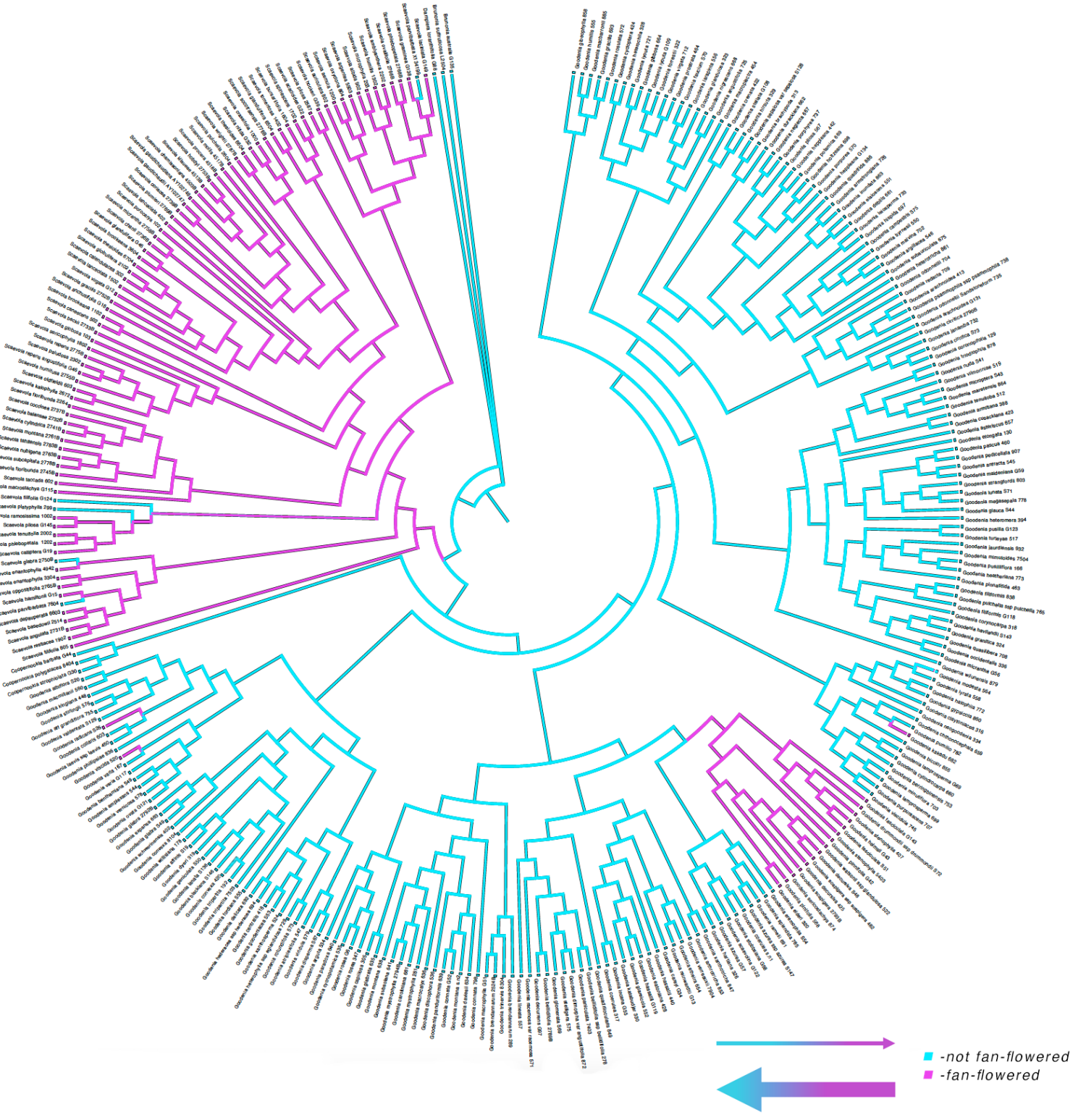


Figure 6B. Ancestral state reconstruction in Mesquite for floral symmetry across the Core Goodeniaceae. Blue branches show non-fan-flowered symmetry in ancestors or extant taxa, purple branches show fan-flowers. Relative transition rates from one state to the other calculated in BayesTraits shown by arrows in bottom right.

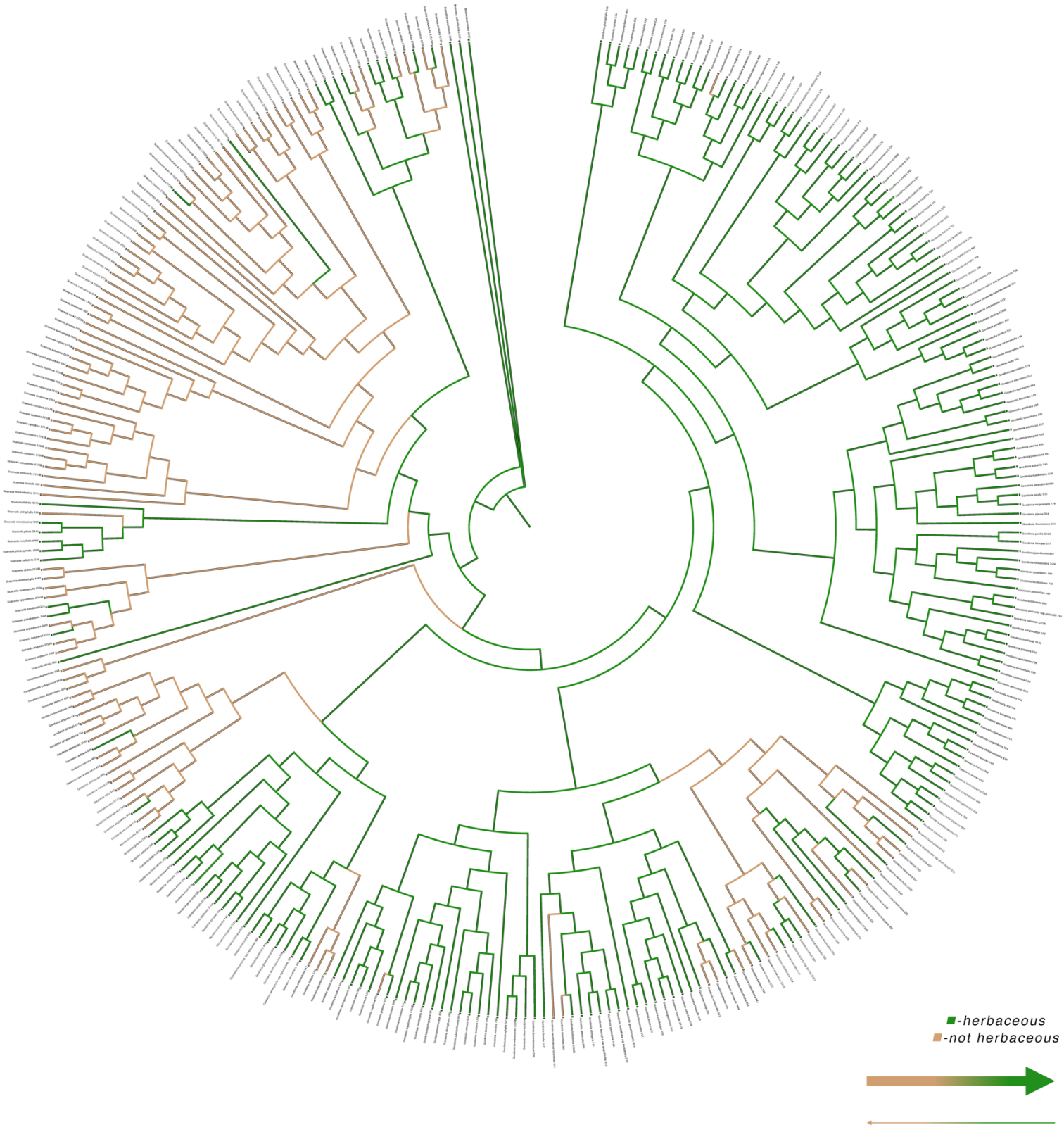


Figure 6C. Ancestral state reconstruction in Mesquite for vegetative habit across the Core Goodeniaceae. Green branches show herbaceous habit in ancestors or extant taxa, brown branches show shrubs. Relative transition rates from one state to the other calculated in BayesTraits shown by arrows in bottom right.

The floral symmetry ancestral states show a common ancestor to all *Scaevola* likely evolved fan-flowers, followed by few reversions back to bilabiate or pseudoradial more recently (Figure 6B). There are a few singular taxa with fan-flowers, *G. viscida* and *G. radicans* in Clade A and *G. kakadu* in Clade B, which we recognize as independent origins of fan-flowers from those in Clade C due to the amount of molecular evolution and the nodes between each of these taxa and *Goodenia* Clade C. The Core Goodeniaceae inferred ancestral state for habit, which is used as a proxy for life history strategy, is herbaceous (Figure 6C), which is contrary to previous observations that the Goodeniaceae is primarily perennial (Jabaily et al., 2012). This analysis shows multiple transitions toward secondary growth and woodiness, especially within *Scaevola*, and major clades within *Goodenia* Clades A and C made up primarily of shrubby habits. *Goodenia faucium* is the only species in Clade B with a non-herbaceous habit, supporting the statement in Shepherd et al. (2020) that most species in this clade are annuals.

Character trait relative transition rates

The BayesTraits relative transition rates shown in Figures 6A-C show the likelihood of transitioning from one state to another for each of the three-character traits analyzed in this study. The fruit form analysis of the Core Goodeniaceae shows there is a significantly higher probability, approximately 39.2 times, of reversion back to dry fruits than evolution of fleshy fruits. A potential explanation for this is the overall rarity of fleshy fruit evolution in the family. Since this state has few evolutions throughout the Goodeniaceae, the rate of transition to fleshiness would be low, and when considering the reversion back to dry fruits in Clade B, the relative rate of evolution of dry fruits would be much higher than initially expected. The floral symmetry analysis shows approximately 3.7 times greater likelihood of reversion back to bilabiate flowers than a shift to fan-flowered symmetry. When comparing this to the ancestral state reconstruction in Mesquite (Figure 6B), the potential explanation for this probability is like that in fruit form. While fan-flowers have evolved in every major clade of the Core Goodeniaceae, there are only two major clades with the evolution of this symmetry with subsequent speciation, and the multiple losses of fan-flowered symmetry could explain the large disparity in transition rates. The secondary growth analysis in BayesTraits shows an 11.5 times higher rate of reversion back to the ancestral state of an herbaceous habit compared to the evolution of woodiness.

Discussion

Topology and Taxonomy of Scaevola

There is still much uncertainty among the relationships within *Scaevola* s.l. which will require more genetic data to resolve. A previous study of *Scaevola* topology showed a small clade of *S. coccinea*, *S. montana*, *S. cylindrica*, and *S. balansae* (Howarth et al., 2003), and is reflected in this study where each node has higher support (Figure 3, Clade D4). *Scaevola acacioides, S. basedowii, S. graminea, S. kalophylla, S. lacinata, S. macrostachya, S. oxyclona,* and *S. pulchella* have previously never been included in a phylogenetic study, so the topology here for all eight taxa are novel. The resolution within *Scaevola* Clade B is similar and more resolved than the findings of Jabaily et al. (2012), as it resolves a major polytomy involving these taxa, excluding *S. parvibarbata*. *Scaevola graminea* and *S. lacinata* are both part of a

polytomy with *S. phlebopetala* and *S. ovalifolia* (Figure 3, Clade C). These latter two taxa were placed in separate, small inner clades of *Scaevola* s.l. in Jabaily et al., 2012, although still part of the same large polytomy in the genus' backbone. We are less inclined to accept these new sister relationships in contradiction to Jabaily et al., 2012 due to the lack of definitive bootstrap support.

The infrageneric taxonomy of *Scaevola*, especially among the two large sections, *Scaevola* and *Xerocarpa*, have lacked evidence for monophyly since the first molecular study (Howarth et al., 2003). The third section of *Scaevola*, *Enantiophyllum*, however, has been highly supported as monophyletic in every study. In Howarth et al. (2003), *Scaevola* section *Enantiophyllum* was placed sister to the rest of the genus, but with very poor bootstrap support. With the addition of more molecular data and a more comprehensive sampling of Australian *Scaevola*, our study shows the two taxa within sect. *Enantiophyllum* are within a clade in sect. *Xerocarpa* (Figure 3, *Scaevola* Clade B) with better support than its previous placement. While *Scaevola* sect. *Xerocarpa* and *S.* sect. *Scaevola* are well accepted as non-monophyletic, Howarth et al. (2003) argues for evidence of subsections and series within sect. *Xerocarpa* as potentially monophyletic. In that study, *S.* sect. *Xerocarpa* subsect. *Parvifoliae* appeared monophyletic with strong support, but with the addition of more Australian species in our study, the subsection no longer has evidence for such monophyly.

Within *Scaevola* Clade A, four of the six taxa included are all part of *S.* sect. *Xerocarpa* subsect. *Pogonanthera*. The two species not included in this grouping are *S. platyphylla*, sister to the four previous taxa, which is included in sect. *Xerocarpa* subsect. *Biloculatae* ser. *Pogogynae*, and *S. filifolia*, which is sister to all other species in Clade A and is part of sect. *Scaevola*. Similarly, *Scaevola* Clade B has the two sister taxa of sect. *Enantiophyllum* most closely related to the rest of the clade. Clade B includes six other taxa, one of which is part of sect. *Xerocarpa* subsect. *Biloculatae* ser. *Pogogynae* (*S. parvibarbata*), while the remaining five species are described as part of sect. *Xerocarpa* subsect. *Parvifoliae*. *Scaevola* Clade C is nearly entirely within subsect. *Biloculatae* ser. *Pogogynae*, with the exceptions of *S. oxyclona* and *S. phlebopetala*, which are part of sect. *Xerocarpa* subsect. *Parvifoliae* and sect. *Xerocarpa* subsect. *Pogonanthera*, respectively. The addition of these new Australian taxa disprove the Howarth et al. (2003) hypothesis that sect. *Xerocarpa* subsect. *Parvifoliae* forms a well-supported monophyletic clade. Additionally, due to the poor backbone support of *Scaevola* in our study, we cannot confirm the previous study's claim that all species within sect. *Xerocarpa* subsect. *Biloculatae* ser. *Globuliferae* forms a paraphyletic clade with multiple taxa from sect. *Scaevola*. Clade D is split into six smaller subclades, with every species included in either sect. *Scaevola* or sect. *Xerocarpa* subsect. *Biloculatae* ser. *Globuliferae*. Clades D2, D3, D4, and D6 are the only subclades that comprise of taxa entirely in one of these subgeneric groupings or the other, further supporting the lack of monophyly within sections.

The southern clade of extra-Australian *Scaevola*, as defined in Howarth et al. (2003), is reflected in this study (Figure 3, *Scaevola* Clade D4) as monophyletic. The northern hemisphere clade (Howarth et al., 2003) is represented in this study by Clade D6 plus three taxa embedded in the polytomy of Clade D, *S. plumieri, S. wrightii*, *S. socotraensis*. Excluding these three taxa, Clade D6 is highly resolved with more than 99% consensus support, compared to the poorly supported clade from the 2003 study which had these three taxa collapsed into a polytomy at the crown.

Fleshy fruit evolution and transoceanic dispersal ability in Scaevola

The multiple evolutions of fleshy fruits in *Scaevola* have been a long hypothesized to play a major part in the genus' dispersal outside of Australia (Howarth et al., 2003). This hypothesis is driven by the connection between the extra-Australian species and the occurrences of fleshy fruits, with only one case of fleshy fruit origin without dispersal, and one species with dry fruits occurring outside the continent. Still, there have been no formal studies into the number of independent evolutions of fleshy fruits in the family. An accepted number of six independent extra-Australian dispersals have occurred in the genus, as identified in Howarth et al. (2003), four of which were said to result in singular species. However, that study did not consider the species *S. enantophylla* to have dispersed outside of the country, as we do in our study (GBIF, 2022). Carolin et al. (1992) suggested in *Flora of Australia* that *Scaevola* sect. *Enantiophyllum* may have up to ten species, but could also be considered a single, highly variable species, but the species boundary between *S. enantophylla* and *S. oppositifolia*, while somewhat unclear, has been established by both Howarth et al. (2003) and Jabaily et al. (2012). The *Flora of Australia* considers *S. enantophylla* to be endemic to northwestern Australia, but multiple georeferenced records have identified the species in Papua New Guinea and Indonesia as well (GBIF, 2022). As such, we recognize only three single-species dispersal events within *Scaevola* in congruence with the dispersal event for the sister taxa *S. enantophylla* and *S. oppositifolia*. These three species resulting from their own dispersal events do not occur within Australia, suggesting that the *S. enantophylla* distribution in the southern Pacific could be the result of a range expansion.

Scaevola beckii is the only fleshy-fruited species within Clade D2. This species has distributions throughout New Caledonia and has been recognized as a single-species dispersal event due to its lack of morphological and molecular similarities to the other New Caledonian species, found in Clade D4, instead being more closely related to the Australian endemic species that comprise the rest of Clade D2. These species all have one-seeded fruits, small flowers, and awned corollas in common, which are not traits shared with the other New Caledonian species (Howarth et al., 2003). *Scaevola gracilis* is embedded within Clade D1, reflecting another significant evolution of fleshy fruits corresponding directly to the dispersal out of Australia in a single-species dispersal event. While the sister taxa *S. micrantha* and *S. chanii* are also within this clade, they represent a potential separate evolution of fleshy fruits as they do not have overlapping ranges. *Scaevola gracilis* occurs throughout New Zealand and Tonga, while the latter two taxa are both found in Borneo, with *S. micrantha* extending into the Philippines and *S. chanii* throughout Malaysia (GBIF, 2022). Carolin et al. (1992) noted that *S. gracilis* is very closely related to the Australian endemic *S. calendulacea*, which also can be found in Clade D1. However, with the addition of new taxa in both Howarth et al. (2003) and in this study, *S. calendulacea* is sister to *S. globulifera* and more distantly related to *S. gracilis* than previously thought.

Scaevola glabra is the last fleshy-fruited taxon that corresponds to a single-species dispersal event. This species is endemic to the Hawaiian Islands, and is the only known tetraploid taxa in the genus (Howarth et al., 2003). The MrBayes tree (Figure 3) shows *S. glabra* collapsed into the backbone polytomy for the entire genus, but when bifurcation is forced in during the ancestral state reconstruction in Mesquite this species is placed in a Clade B with the evolution of fruit fleshiness part of the same shift as the two taxa in section *Enantiophyllum*. Due to insufficient of support among the backbone of *Scaevola*, the lack of overlapping distribution

between *S. enantophylla, S. oppositifolia,* and *S. glabra*, and a mismatch in morphological and molecular characters with the other Hawaiian species of the genus, we support the conclusions of Howarth et al. (2003) that *S. glabra* falls under a single-species dispersal event.

The two most clear independent evolutions of fleshy fruits in *Scaevola* are represented in Clades D4 and D6 which correspond to the southern and northern dispersal clades identified in Howarth et al. (2003), respectively. *Scaevola* Clade D4 directly matches the nine taxa identified in the 2003 study, whose species are distributed throughout the Pacific Islands and New Caledonia. One of these species, *S. taccada*, is one of the two most successful species at transoceanic dispersal throughout the Pacific. The other successful disperser, found throughout the Pacific, Atlantic, and Indian Oceans, is *S. plumieri*, which is part of the northern transoceanic dispersal clade (Howarth et al., 2003). This clade is represented in this study as Clade D6, but also includes three additional taxa that had been collapsed into the greater backbone polytomy of Clade D, one of which was *S. plumieri*. Following the forced bifurcations for the ancestral state reconstruction, these three taxa, *S. plumieri, S. wrightii,* and *S. socotraensis,* are included in a larger clade made up of Clades D5 and D6 plus *S. pulchella*, which was also part of the backbone Clade D polytomy. *S. wrightii* and *S. socotraensis* are shown in our Mesquite analysis (Figure 6A) to be part of two separate individual shifts to fleshy fruits, despite being part of the northern dispersal clade in Howarth et al. (2003), likely due to the lack of backbone support in our analyses. Due to this, we cannot suggest that these two taxa are part of their own independent fleshy fruit evolutions. Additionally, *S. pulchella* was not previously included in the northern dispersal clade because it was not previously sampled in a phylogenetic study, and it is a dry-fruited Australian endemic species (Carolin et al., 1992). This would most likely be considered a reversion to dry fruits and endemicity, but due to the lack of support in placement of *S. pulchella* in the phylogeny, we cannot determine this with confidence until further sampling is done. The eleven species that make up the northern clade of extra-Australian species – eight species in Clade D6 and three included from the Clade D polytomy – are distributed throughout the Hawaiian Islands, Atlantic coastlines, and into Cuba and Socotra (Howarth et al., 2003). This clade's dispersal ability most likely stems from a single evolution of fleshy fruits near its crown.

Another species of note in Clade B is *Scaevola angulata*, which is the only species with dry fruits not endemic to Australia, with distributions across Indonesia and Papua New Guinea (GBIF, 2022; Howarth et al., 2003). Additionally, *S. angulata*, as seen in Figure 6A, has dry fruits following the only reversion back to this form following an evolution of fleshy fruits. Another exception to the correlation between fleshy fruit evolution and transoceanic dispersal, Clade D3 shows the three Australian endemic taxa with fleshy fruits: *S. bursariifolia, S. spinescens,* and *S. acacioides*. Similar to the placement of *S. glabra* in Clade B as visualized with required bifurcation in Mesquite, *S. tomentosa*, another endemic species, has been moved out of the polytomy of Clade D and placed in Clade D3 with the three aforementioned taxa, joining the only other Australian endemic species with fleshy fruits. When looking into the fruit form throughout the rest of the Core Goodeniaceae, there is a singular taxon in *Goodenia* with fleshy fruits: *G. radicans*. This species was formerly considered part of the monotypic herbaceous genus with fleshy fruits, *Selliera*, before it was synonymized into *Goodenia* to produce monophyly (Shepherd et al., 2020). In accordance with most fleshy fruited Goodeniaceae, *G. radicans* is distributed outside Australia, throughout coasts and highlands of New Zealand and southern Chilean salt marshes (Carolin et al., 1992).

When comparing the branches for each shift from capsular fruits to fleshy fruits in Mesquite to the ultrametric chronogram generated in BEAST, the first evidence of fleshy fruit

evolution is only as old as 10.3 Mya and the most recent evidence suggesting an evolution approximately 2.5 Mya. The oldest node at which a shift to fleshiness appears at the stem of Clade D4, which corresponds to the clade of southern Pacific distributed species (Howarth et al., 2003). Similar to previous chronograms (Jabaily et al., 2012), *Scaevola* speciates much faster than *Goodenia*. This is reflected also in the BAMM analysis of diversification rates where in each of the possible credible shifts in the 95% credibility set as well as in the highest probability phylorate plot *Scaevola* has a significantly faster rate of net diversification than *Goodenia* (Figure 5). Each of the nodes identified to show the evolution of fleshy fruits occurs within a relatively short period, suggesting that a biogeographic study of the genus and the evolution of this trait could help uncover a more complete story of these species.

Diversification rates and ancestral character state reconstruction

Two characters of interest that have been hypothesized to contribute to speciation in the Goodeniaceae are the floral symmetry and habit of the species. Floral symmetry in the family has been extensively studied due to the uniqueness of the fan-flower shape and its convergent evolution (Gardner et al., 2016a,b). We originally hypothesized that the differential flexibility in floral symmetry within *Goodenia* would lead to increased diversification rates compared to *Scaevola* with its putative increasing pollinator diversity and specificity; however, the higher diversification in *Scaevola* than *Goodenia* in general suggest that ubiquitous fan-flowered symmetry may not decrease diversification rates. *Goodenia* subgenus *Monochila* is fan-flowered, with the ancestor of this clade, within Clade C, also likely evolving this trait. This morphology shift within *Goodenia* does not coincide with a diversification rate shift hypothesized in previous studies of the family (Gardner et al., 2016a), instead there was no change in speciation among the taxa in this clade (Figure 5). The BAMM phylorate plot shows a much greater relative rate of speciation among *Scaevola* especially compared to *Goodenia* Clade B (Figure 5), with both approaching these extremes at around the same period (13-20 Mya). *Scaevola* is nearly entirely fan-flowered, with only a few taxa having reverted to the family ancestral state, while *Goodenia* Clade B has only 1 species with fan-flowers. If the hypothesis that the fan-flowered symmetry allows for greater speciation within the family holds true, these would be the expected diversification rate results.

Vegetative habit is often used as a simplifying assumption for life history strategies in plants. Although Goodeniaceae has previously been noted as primarily perennial (Jabaily et al., 2012), which often indicates woodiness in the species, the ancestral state for this trait shows the ancestor of the Core Goodeniaceae was herbaceous (Figure 6C). A theory regarding diversification rates and life history suggests the herbaceous or annual plants would have much greater diversification rates due to the decreased amount of time between generations (Smith & Donoghue, 2008). However, rapid diversifying *Scaevola* has a primarily shrubby habit and slower diversifying *Goodenia* Clade B has only a single shrub in the herbaceous clade, contradicting the hypothesis. One potential reason for this is that the ability to have secondary growth cannot always confidently be used to supplement data for life history strategies, especially in a family such as the Goodeniaceae, where many perennial plants are herbaceous above ground and in many specimens, and exist underground as long-term vegetative structures during less favorable conditions. This indicates the ancestral state of the clade could still be perennial, even if it cannot form secondary growth. In the case of an herbaceous perennial, the timing of reproduction has the potential to be as long as a shrubby perennial or tree, so the

diversification rate may not be as fast as previously thought due to the lack of wood, potentially explaining the decreased rate in the primarily herbaceous clade. The large proportion of shrubby perennial *Scaevola* species could support the opposite argument, though, suggesting the presence of secondary growth in perennial taxa may help increase speciation rates. Other studies have cited slower diversification rates in annual herbaceous taxa due to greater extinction rates among the lineage (Friedman, 2020), however the BAMM analysis showed no evidence of increased extinction in Clade B compared to *Scaevola*. The multiple shifts toward shrubby habit in *Scaevola*, especially within extra-Australian species, supports the island biogeography theory of increased diversification and adaptive radiation on islands (Carlquist, 1972). Many species that establish on islands often evolve perennial habits and become large shrubs or trees when populating in different niches on islands (Cox & Burns, 2017), whereas continental species are typically herbaceous. *Scaevola* has many species that support this case, as nearly every extra-Australian taxa can support secondary growth and the majority of herbaceous species are endemic to Australia.

Future directions and methods review

Our conclusions are strengthened by the enhanced sampling within *Scaevola* and are primarily limited by poor phylogenetic resolution from analyzing only two loci, sequenced more than a decade ago with many are previously unpublished. As some resolution has been achieved and sister relationships begin to become clearer, one of the most important resolved topological implications is the identification of the sister taxon to *S. aemula, S. oxyclona*. *Scaevola aemula* is a major horticultural species available worldwide and finding its closest relatives can help inform horticultural efforts in the future.

Including chloroplast genes in a study can help decipher the evolutionary history of the taxon set by unveiling chloroplast capture and other hybridization methods visible in maternally inherited genes. Chloroplast data is imperative for resolving the *Scaevola* phylogeny, especially because the genus is known to hybridize (Howarth & Baum, 2005). New developments in genetic sampling methods have made sequencing whole chloroplast genomes more accessible. As a result of this, high-throughput sequencing of both chloroplast and nuclear genomes is the best way to approach solving the topology of *Scaevola*.

In an attempt to resolve the topological issues determined in Jabaily et al. (2012), genome skimming next-generation sequencing was done on a select subset of taxa in the Core Goodeniaceae to determine backbone relationships, particularly within *Goodenia* s.l. (Gardner et al., 2016b). The results were promising, with fully resolved chloroplast phylogenies, supported by the previous efforts with nuclear genes (Jabaily et al., 2018). A continuation of this effort is needed to resolve the polytomies, especially within *Scaevola*, by using targeted and enriched next-generation sequencing approaches. Such efforts could start with the Genomics for Australian Plants (GAP) project (Genomics for Australian Plants, 2018), which seeks to provide genomic level sequences for every Australian plant species, starting with one representative for each genus. The project has currently utilized the probe set Angiosperms353 (Johnson et al., 2019) to target, amplify, and sequence in parallel 353 nuclear loci across 10+ species in the Goodeniaceae, including several *Scaevola*. A related project on the genus *Dampiera* in Goodeniaceae explored the existing Goodeniaceae GAP project data and developed *Dampiera* specific primers from several of the 353 loci for Sanger sequencing (Li & Fanestil, 2022). These

new loci were longer, more variable, and easier to amplify than the loci sequenced by Howarth et al. (2003).

BAMM diversification methods have been the topic of a heated debate on the reliability and credibility of the underlying statistical work in the analyses following a critical review article (Moore et al., 2016). The authors claimed there was extreme bias in the estimation of diversification rates due to the failure to account for hidden rate shifts in extinct lineages, as well the priors' unreliable sensitivity to posterior values (Moore et al., 2016). The following year, the developers of BAMM published a response, seeking to disprove the claims and conducting an entire review of the critique's claims (Rabosky et al., 2017). This review has since been published on the project website (bamm-project.org) as a tool for BAMM's users to test the reliability of the program on their own. This debate has divided the systematic community, but for our purposes, we acknowledge the potential flaws in the program and continue to use the results with caution. Due to the lack of backbone support in *Scaevola*, the diversification rate analysis will need to be revisited following more resolution in the genus, so a further review of the reliability of the analyses conducted can be done at that time.

The addition of new data and taxa in our study have begun to solve the lack of statistical support across the phylogeny and shed more light onto the taxonomic issues of *Scaevola*. A more fully resolved topology within *Scaevola* will be of use to the broader botanical community. We suggest the need for considerable additional sequence data within *Scaevola* to fully resolve the topology followed by a recircumscription of the subgeneric groups to reflect phylogeny. High throughput sequencing of *Scaevola* is the logical next step, as single-loci molecular data is no longer sufficient for the level of work required for resolution in this genus. Our analyses of diversification rates and character trait evolution are the first in the Goodeniaceae, providing the basis for further comparative method studies into the evolutionary history of this family.

References

Barreda, V. D., Palazzesi, L., Tellería, M. C., Katinas, L., Crisci, J. V., Bremer, K., Passalia, M. G., Corsolini, R., Brizuela, R. R., & Bechis, F. (2010). Eocene Patagonia Fossils of the Daisy Family. *Science*, *329*(5999), 1621–1621. https://doi.org/10.1126/science.1193108

Barreda, V. D., Palazzesi, L., Telleria, M. C., Olivero, E. B., Raine, J. I., & Forest, F. (2015). Early evolution of the angiosperm clade Asteraceae in the Cretaceous of Antarctica. *Proceedings of the National Academy of Sciences of the United States of America*, *112*(35), 10989–10994. https://doi.org/10.1073/pnas.1423653112

Biomatters Ltd. (2005). *Geneious Prime* (2022.0.2) [Computer software]. Geneious. www.geneious.com

Carlquist, S. (1972). Island Biology: We've Only Just Begun. *BioScience*, *22*(4), 221–225. https://doi.org/10.2307/1296389

Carolin, R. (1990). Nomenclatural notes, new taxa and the systematic arrangement in the genus Scaevola (Goodeniaceae) including synonyms. *Telopea Journal of Plant Systematics*, *3*(4), 477–515.

Carolin, R. C., Morrison, D. A., & Rajput, T. (1992). *Flora of Australia* (Vol. 35). Australian Government Publishing Service Canberra.

Cayzer, L., Adams, L., Cowley, K., Lepschi, B., & Whitbread, G. (2002). Australian Plant Name Index. *Conservation Science Western Australia*, *4*(3), 94–96.

Cox, B. T. M., & Burns, K. C. (2017). Convergent evolution of gigantism in the flora of an isolated archipelago. *Evolutionary Ecology*, *31*(5), 741–752. https://doi.org/10.1007/s10682-017-9909-0

Darriba, D., Taboada, G. L., Doallo, R., & Posada, D. (2012). jModelTest 2: More models, new heuristics and parallel computing. *Nature Methods*, *9*(8), 772–772. https://doi.org/10.1038/nmeth.2109

Drummond, A. J., & Rambaut, A. (2007). BEAST: Bayesian evolutionary analysis by sampling trees. *BMC Evolutionary Biology*, *7*(1), 1–8. https://doi.org/10.1186/1471-2148-7-214

Friedman, J. (2020). The Evolution of Annual and Perennial Plant Life Histories: Ecological Correlates and Genetic Mechanisms. *Annual Reviews of Ecology, Evolution, and Systematics*, *51*, 461–481.

Gardner, A. G., Fitz Gerald, J. N., Menz, J., Shepherd, K. A., Howarth, D. G., & Jabaily, R. S. (2016a). Characterizing Floral Symmetry in the Core Goodeniaceae with Geometric Morphometrics. *PloS One*, *11*(5), e0154736.

Gardner, A. G., Sessa, E. B., Michener, P., Johnson, E., Shepherd, K. A., Howarth, D. G., & Jabaily, R. S. (2016b). Utilizing next-generation sequencing to resolve the backbone of the Core Goodeniaceae and inform future taxonomic and floral form studies. *Molecular Phylogenetics and Evolution*, *94*, 605–617. https://doi.org/10.1016/j.ympev.2015.10.003

GBIF. (2022). *Global Biodiversity Information Facility*. GBIF. https://www.gbif.org/

Genomics for Australian Plants. (2018). *Genomics for Australian Plants*. https://www.genomicsforaustralianplants.com/

Guindon, S., & Gascuel, O. (2003). A Simple, Fast, and Accurate Algorithm to Estimate Large Phylogenies by Maximum Likelihood. *Systematic Biology*, *52*(5), 696–704. https://doi.org/10.1080/10635150390235520

Howarth, D. G., & Baum, D. A. (2005). Genealogical Evidence of Homoploid Hybrid Speciation in an Adaptive Radiation of Scaevola (goodeniaceae) in the Hawaiian Islands. *Evolution*, *59*(5), 948–961. https://doi.org/10.1111/j.0014-3820.2005.tb01034.x

Howarth, D. G., Gustafsson, M. H. G., Baum, D. A., & Motley, T. J. (2003). Phylogenetics of the genus Scaevola (Goodeniaceae): Implication for dispersal patterns across the Pacific Basin and colonization of the Hawaiian Islands. *American Journal of Botany*, *90*(6), 915–923. https://doi.org/10.3732/ajb.90.6.915

Jabaily, R. S., Shepherd, K. A., Gardner, A. G., Gustafsson, M. H. G., Howarth, D. G., & Motley, T. J. (2014). Historical biogeography of the predominantly Australian plant family Goodeniaceae. *Journal of Biogeography*, *41*(11), 2057–2067. https://doi.org/10.1111/jbi.12363

Jabaily, R. S., Shepherd, K. A., Gustafsson, M. H. G., Sage, L. W., Krauss, S. L., Howarth, D. G., & Motley, T. J. (2012). Systematics of the Austral–Pacific family Goodeniaceae: Establishing a taxonomic and evolutionary framework. *TAXON*, *61*(2), 419–436. https://doi.org/10.1002/tax.612012

Jabaily, R. S., Shepherd, K. A., Michener, P. S., Bush, C. J., Rivero, R., Gardner, A. G., & Sessa, E. B. (2018). Employing hypothesis testing and data from multiple genomic compartments to resolve recalcitrant backbone nodes in Goodenia s.l. (Goodeniaceae).

Molecular Phylogenetics and Evolution, *127*, 502–512. https://doi.org/10.1016/j.ympev.2018.05.005

Johnson, M. G., Pokorny, L., Dodsworth, S., Botigué, L. R., Cowan, R. S., Devault, A., Eiserhardt, W. L., Epitawalage, N., Forest, F., Kim, J. T., Leebens-Mack, J. H., Leitch, I. J., Maurin, O., Soltis, D. E., Soltis, P. S., Wong, G. K., Baker, W. J., & Wickett, N. J. (2019). A Universal Probe Set for Targeted Sequencing of 353 Nuclear Genes from Any Flowering Plant Designed Using k-Medoids Clustering. *Systematic Biology*, *68*(4), 594–606. https://doi.org/10.1093/sysbio/syy086

Katoh, K., & Standley, D. M. (2013). MAFFT Multiple Sequence Alignment Software Version 7: Improvements in Performance and Usability. *Molecular Biology and Evolution*, *30*(4), 772–780. https://doi.org/10.1093/molbev/mst010

Lecocq, T., Vereecken, N. J., Michez, D., Dellicour, S., Lhomme, P., Valterová, I., Rasplus, J.-Y., & Rasmont, P. (2013). Patterns of Genetic and Reproductive Traits Differentiation in Mainland vs. Corsican Populations of Bumblebees. *PLoS ONE*, *8*(6), e65642. https://doi.org/10.1371/journal.pone.0065642

Li, P., & Fanestil, N. (2022, April 1). *Designing Custom Genetic Loci to Improve the Phylogeny of Dampiera (Goodeniaceae)* [Poster].

Maddison, W. P., & Maddison, D. R. (2021). *Mesquite: A modular system for evolutionary analysis* (3.70) [Computer software]. http://www.mesquiteproject.org

Miller, M. A., Pfeiffer, W., & Schwartz, T. (2010, November 14). *Creating the CIPRES Science Gateway for inference of large phylogenetic trees*. Gateway Computing Environments Workshop.

Moore, B. R., Höhna, S., May, M. R., Rannala, B., & Huelsenbeck, J. P. (2016). Critically evaluating the theory and performance of Bayesian analysis of macroevolutionary mixtures. *Proceedings of the National Academy of Sciences*, *113*(34), 9569–9574. https://doi.org/10.1073/pnas.1518659113

Pagel, M., & Meade, A. (2006). Bayesian Analysis of Correlated Evolution of Discrete Characters by Reversible-Jump Markov Chain Monte Carlo. *The American Naturalist*, *167*(6), 808–825. https://doi.org/10.1086/503444

Rabosky, D. L., Grundler, M., Title, P., Anderson, C., Shi, J., Brown, J., Huang, H., & Mitchell, J. S. (2014). BAMMtools: Analysis and Visualization of Macroevolutionary Dynamics on Phylogenetic Trees. *Methods in Ecology and Evolution*, *5*, 701–707.

Rabosky, D. L., Mitchell, J. S., & Chang, J. (2015). *BAMM - Bayesian Analysis of Macroevolutionary Mixtures*. http://bamm-project.org/index.html

Rabosky, D. L., Mitchell, J. S., & Chang, J. (2017). Is BAMM Flawed? Theoretical and Practical Concerns in the Analysis of Multi-Rate Diversification Models. *Systematic Biology*, *66*(4), 477–498. https://doi.org/10.1093/sysbio/syx037

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

Rambaut, Andrew. (2018). *FigTree* (1.4.4) [Computer software]. http://tree.bio.ed.ac.uk/software/figtree/

Ronquist, F., & Huelsenbeck, J. P. (2003). MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics*, *19*(12), 1572–1574. https://doi.org/10.1093/bioinformatics/btg180

Shepherd, K. A., Lepschi, B. J., Johnson, E. A., Gardner, A. G., Sessa, E. B., & Jabaily, R. S. (2020). The concluding chapter: Recircumscription of Goodenia (Goodeniaceae) to include four allied genera with an updated infrageneric classification. *PhytoKeys*, *152*, 27–104. https://doi.org/10.3897/phytokeys.152.49604

Smith, S. A., & Donoghue, M. J. (2008). Rates of Molecular Evolution Are Linked to Life History in Flowering Plants. *Science*, *322*(5898), 86–89. https://doi.org/10.1126/science.1163197

Tank, D. C., & Donoghue, M. J. (2010). Phylogeny and Phylogenetic Nomenclature of the Campanulidae Based on an Expanded Sample of Genes and Taxa. *Systematic Botany*, *35*(2), 425–441. https://doi.org/10.1600/036364410791638306

Western Australian Herbarium, B. and C. S. (1998). *Florabase—The Western Australian Flora* (State of Western Australia). Western Australian Herbarium, Biodiversity and Conservation Science, Department of Biodiversity, Conservation and Attractions. https://florabase.dpaw.wa.gov.au/