Article title:

Recurrent genome duplication events likely contributed to both the ancient and recent rise of ferns

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The following Supporting Information is available for this article:

Supplementary Text

Comparison of changes in diversification rates in this study and those reported by Testo and Sundue (2016)

Testo and Sundue (2016) sampled nearly 4000 species of ferns (3973 ferns and 34 outgroups) and reconstructed phylogeny based on six chloroplast regions. The placements of several families are different from the phylogenetic backbone used here [Fig. 1; see Qi et al. (2018)], probably due to the use of sequences from different compartments (i.e. nucleus or chloroplasts). Nuclear sequences are inherited from both parents whereas chloroplast sequences are maternally inherited; thus they possibly represent somewhat different histories of evolutionary divergences. Also, plastid genes are more conserved (with lower molecular evolutionary rate) than nuclear sequences, potentially contributing to differences in species phylogeny. Nevertheless, we compared our results in diversification rates to those reported in Testo and Sundue (2016). In brief, placements of rate shifts in the previous study preferred to be at derived nodes; there was only one shift at deep branch (all ferns excluding Equisetales), while all other shifts were located within a family. Although the defined "rate shifts" seem quite different in the two studies, they become more similar when regarding the changes in rates rather than the selected rate shift nodes. In Testo and Sundue (2016), the identified rate shifts are in clades of Polypodiaceae, *Tectaria* (Tectariaceae), Dryopteridaceae, Athyriaceae, Thelypteridaceae, *Asplenium* (Aspleniaceae), Pteridaceae, Lindsaeaceae, Cyatheaceae, *Marsilea* (Salviniales), *Angiopteris* (Marattiales). In our study, we found signals for rate shifts in clades of Polypodiaceae, Dryopteridaceae, Lindsaeaceae, Cyatheaceae and Marattiales, with varied placements for the exact positions. Except these, we also observed changes in rates (as shown in varying colors of the branches in Fig. 2) of branches in Tectariaceae, Athyriaceae, Thelypteridaceae, Aspleniaceae and Pteridaceae, although these changes may be relative minor than the shifts identified here and thus are not defined as having a "core shift" in rates, which is probably due to the differences in the number of samplings and in

phylogenetic topologies.

Ancient WGDs and the variations in chromosome numbers along the phylogenetic backbone

The newly identified WGDs from transcriptomic datasets provide an opportunity to compare with earlier reconstruction of ancestral chromosome numbers (Clark et al., 2016). Among the WGDs shared by many fern families (the backbone ones), the one for most leptosporangiates, after the separation of Osmundales (event 1, Fig. 1), is not associated with an dramatic increase in reconstructed ancestral chromosome numbers (a2n) on nearby branches (48 to 46) (Clark et al., 2016), and even with decreasing chromosome sizes from 0.42 to 0.27 pg, It is possible that this early WGD was followed by chromosome rearrangements/fusions that maintained similar chromosome numbers with reduced sizes.

On the other hand, the other two backbone WGDs (events 2 and 3) were both associated with an increase in chromosome numbers. Event 2 shared by most core leptosporangiates except Salviniales (i.e. Cyatheales and Polypodiales) corresponds to an increase of a2n from 46 to 64 (Polypodiales) or 68 (Cyatheales), as well as the doubling of holoploid genome size of Polypodiales (9.67 to 16.07 pg) in cytogenetic study (Clark et al., 2016). A previous study of the *Ceratopteris richardii* (Pteridaceae, Polypodiales) genetic map suggests duplications for most loci, but with only a weak signal for synteny between duplicated segments (Nakazato et al., 2006). This weak syntenic signal might imply an old WGD event such as the one shared by Polypodiales and Cyatheales identified here.

Event 3 as position here would affect all eupolypods with 20 of the 26 families of Polypodiales and more than two thirds of extant fern diversity (PPG I, 2016), and is consistent with an increase of a2n from that of eupolypod (a2n=54) to that of eupolypods I (a2n=82) (Clark et al., 2016). Although there is no increase in a2n from eupolypods to eupolypods II in Clark et al. (2016), the same study showed that almost all families of eupolypods have a2n from 70 to 82,

with an exception of 50 for Thelypteridaceae. This low value of Thelypteridaceae might have contributed some uncertainties (such as variation in samplings) during the analysis, whereas in another study (Sundue and Rothfels, 2014), the estimated a2n for Thelypteridaceae showed a relatively large possible range of a2n from 64 to higher than 80. Taken together, it is possible that the overall trend of an increase in a2n values of eupolypods reflect the results of an ancestral WGD, representing a good agreement between the WGDs detected here and increases in ancestral chromosome numbers reported previously.

Genome duplications for orders and families

In addition to the three WGD events shared by multiple orders or families, we also found evidence for lineage-specific WGDs in eight orders, while the three orders not showing evidence of lineage-specific WGD diverged between events 1 and 2.

Equisetales, Psilotales, Ophioglossales, Marattiales and Osmundales each have only one family. The WGD events detected here using molecular evidence (events 4-6 in Fig. 1 and events 13-14 in Fig. S1) in four orders are further supported by the increases of a2n from the value of their corresponding ancestral stems to the crown groups (44 to 216 for Equisetales, 44 to 104 for Psilotales, 44 to 90 for Ophioglossales and 44 to 78 for Marattiales) (Clark et al., 2016). The same study also showed different patterns in the changes of genome and chromosome sizes for these orders, further suggesting that genomes of Ophioglossales (0.52 to 0.27 pg) and Marattiales (~0.45 to 0.23 pg) might have experienced rearrangement while Psilotales tend to maintain the doubled genome (with the ancestral chromosome size as 0.62 pg). In fact, genomes of Psilotales not only have a high ancestral chromosome number (a2n=104) (Clark et al., 2016) but also possessed the largest sizes (can be up to 1C=150.61 pg) in ferns (Hidalgo et al., 2017b) and even of eukaryotes in the current understanding (Hidalgo et al., 2017a). These observations in Psilotales fit the hypothesis of static genomes (Leitch and Leitch, 2013). Values of a2n at the branches near the base of Osmundales are similar (48 and 44), but the average chromosome sizes

increased evidently from 0.42 to 0.69 pg. As an additional support, observations on the pattern of chromosome rearrangements (Zhang et al., 2008) suggested interspecific hybridization in this order, whereas Tsutsumi et al. (2011) reported allopolyploidy in members of Osmundaceae via taxonomic, cytogenetic, and flow-cytometric analyses as well as molecular phylogenies, consistent with the WGD detected here in this order.

In Gleicheniales, the two families sampled here, Gleicheniaceae and Dipteridaceae, each have one detected WGD (events 7 and 8) for their stem groups. This is generally consistent with the a2n of the ancestors of the two families being 48 and 2n values of extant species ranging from 66 (*Dipteris* sp.) to 112 (*Diplopterygium* sp.) (Clark et al., 2016). The genome and chromosome sizes of some Gleicheniales members are among the smallest in ferns, and the a2n value is nearly half to that of Marattiales (Clark et al., 2016), suggesting that members of Gleicheniales might have experienced extensive genome rearrangements.

In Cyatheales, a WGD (event 9) was shared by the five families included here, consistent with an increase in the a2n from 46 for core leptosporangiates to 68 for the MRCA of Cyatheales. Two more WGD events (events 10 and 11) were supported here along the branches leading to *Alsophila* and to *Plagiogyria*. Such events are also supported by the observation that members of Cyatheales examined in Clark et al. (2016) have chromosome numbers greater than 100. Three other orders (Hymenophyllales, Schizaeales, Salviniales) showed no lineage-specific WGD signal in our analysis, although they shared with other orders the earliest WGD along the fern backbone (event 1). Among these orders, Salviniales have the smallest genomes in ferns (Obermayer et al., 2002; Clark et al., 2016); for example, the genome of *Slavinia* (0.26 Gb) being the smallest one ever reported in ferns (Li et al., 2018). This possibly implies limited number of lineage-specific WGDs for this order after the one shared by almost all leptosporangiate ferns. Consistently, Li et al. (2018) identified a potential WGD event for *Azolla* and the coincidently larger genome size as 0.75 Gb, but no WGD event across genera was found within this order.

Genomes of Hymenophyllales are large and the ancient genome/chromosome sizes and

numbers were reconstructed as intermediate among orders (Clark et al., 2016), but the samplings of this order was limited in the previous study, with four of the seven taxa lacking the information of chromosome numbers and genome sizes. Further analysis with more sampling in this order might yield more insights into the evolution of their chromosome number and genome size.

Maintaining many chromosomes or rearranging to fewer chromosomes

It has been suggested that chromosomes in ferns tended to be maintained but silenced (Haufler, 1987; Leitch and Leitch, 2013; Haufler, 2014) to a low (diploid) expression state (Haufler and Soltis, 1986). From the previous analyses of changes in chromosome numbers and our results for recurrent WGD events, we agree with the conclusion of (Clark et al., 2016) that the above hypothesis could explain some, but not all, cases across fern diversity. At the same time, substantial chromosome rearrangements might have occurred in some orders (such as Gleicheniales, Polypodiales and maybe Marattiales), after the earliest WGD detected here (event 1) shared by 97% of extant ferns during Paleozoic.

On the other hand, it is surprising that only two WGDs were detected within Polypodiales, the largest fern order accounting for more than 80% of extant fern diversity. Besides the WGD (event 3) shared by all eupolypods, only one WGD was identified near the base of Lindsaeaceae (sharing by at least three genera included here) among the lineages of Polypodiales, while signals of other nodes are very low. Although there are still chances to identify other WGDs for some families by increasing taxon sampling, we did not detect WGD signals across multiple families or shared by the members of the same family with multiple taxa here. In fact, some previous studies supported the idea that genome duplications in Polypodiales might be fewer than expected. Barker (2009) observed only one shared peak at Ks ~1.6 from the analysis with ESTs of two Pteridaceae species and suggested an ancient genome duplication (Barker, 2009; Barker and Wolf, 2010), but no peaks at smaller Ks was reported. In addition, study on the genetic linkage map of

C. richardii revealed a history of duplications for most loci, but there was only a weak signal of synteny among duplicated segments (Nakazato et al., 2006). The weak signal of synteny fits the suggestion of an old WGD. Nevertheless, the variations of genome and chromosome sizes are quite large in Polypodiales (Clark et al., 2016), leading to the suggestion that some lineages of this large order might have experienced independent genome rearrangements (Clark et al., 2016).

Genome rearrangement after WGDs is a common mechanism that has been reported by many studies in angiosperms (Song et al., 1995; Gaeta and Pires, 2010; Bomblies and Madlung, 2014), such that no substantial increase in chromosome numbers is observed even when there have been recurrent WGDs. It has been proposed that the vigorous genome rearrangements in angiosperms might be one of the main forces resulting in the high diversity and thus the prominent plant group on Earth (Pellicer et al., 2018; Simonin and Roddy, 2018). Similar case might also be true for some ferns; in contrast to the high diversity in Polypodiales, lineage-specific WGD is limited in this large order; on the other hand, genome rearrangements might be relatively vigorous, contributing to the large variations in genome and chromosome sizes among Polypodiales members (Clark et al., 2016). In other words, chromosome rearrangements following the ancient WGDs along the fern backbone might have facilitated speciation and divergence in this order, resulting in the highest diversity in extant fern species.

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Figure S1. WGD identification with additional five species of Equisetales and Osmundales. Analytical method here is the same as described in Materials and Method and in Fig. 1 except the addition of five datasets in Equisetales and Osmundales (red branches). The WGD IDs (left column) correspond to the numbers in Fig. 1A and 1B. The 12 candidate events in Fig. 1 are well supported in this extended analysis whereas additional two events were identified for Equisetales (13) and Osmundales (14). To the right of branch tips are species names, and names further to the right are the names of orders (in boldface, all except Polypodiales) or families (only for those in Polypodiales).

WGD ID	Pure-MCL			MCL-BLAST		
	%	count	> #8	%	count	> seed
13	39.97	2995	O	49.88	2119	O
6	21.96	1996	O	26.71	1212	O
10	16.73	1244	O	22.13	855	O
1	13.28	1920	O	9.2	378	O
3	12.6	2561	O	22.71	1045	O
4	12.28	667	O	10.08	331	O
11	12.19	1248	O	17.61	801	O
2	12.09	1875	O	7.57	268	O
9	11.98	1310	O	10.47	463	O
12	11.38	1037	O	13.2	594	O
14	9.94	854	O	6.77	304	O
7	9.38	884	O	8.78	405	O
5	8.83	554	O	9.14	367	O
8	8.66	769	O	11.54	523	O

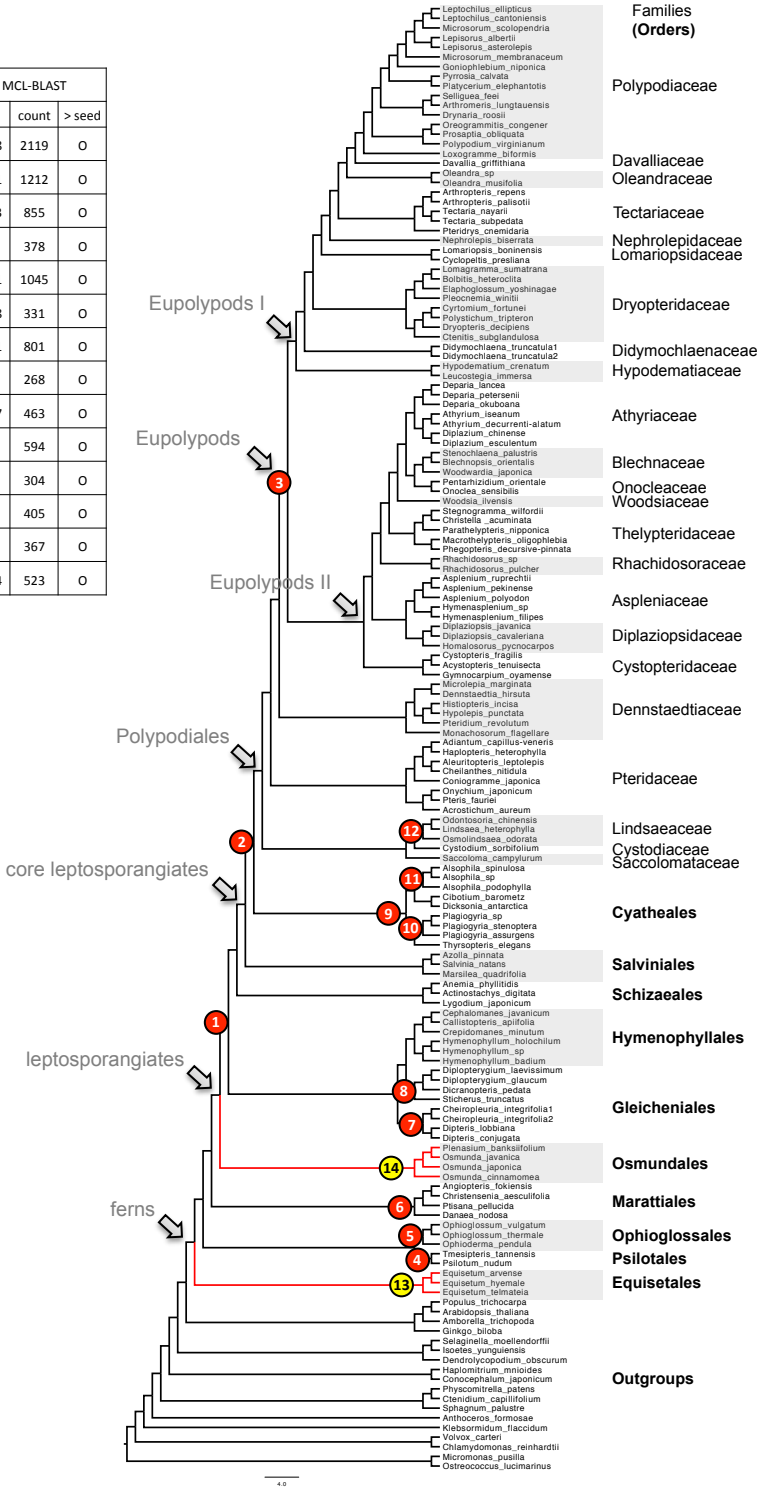

Figure S2. GO enrichment results for gene duplicates of the WGD events. This figure shows the over-representation of functional categories (defined by GO terms at level 2; adjusted $p < 0.05$) of gene duplicates for the WGD events in ferns. Values (colors) indicate the approximate fraction of genes in the indicated GO categories relative to total gene duplicates of the respective WGDs. We used the 4067 OGs as the background gene sets (see Materials and Methods, the section on identification of orthologous genes) in the enrichment analyses.

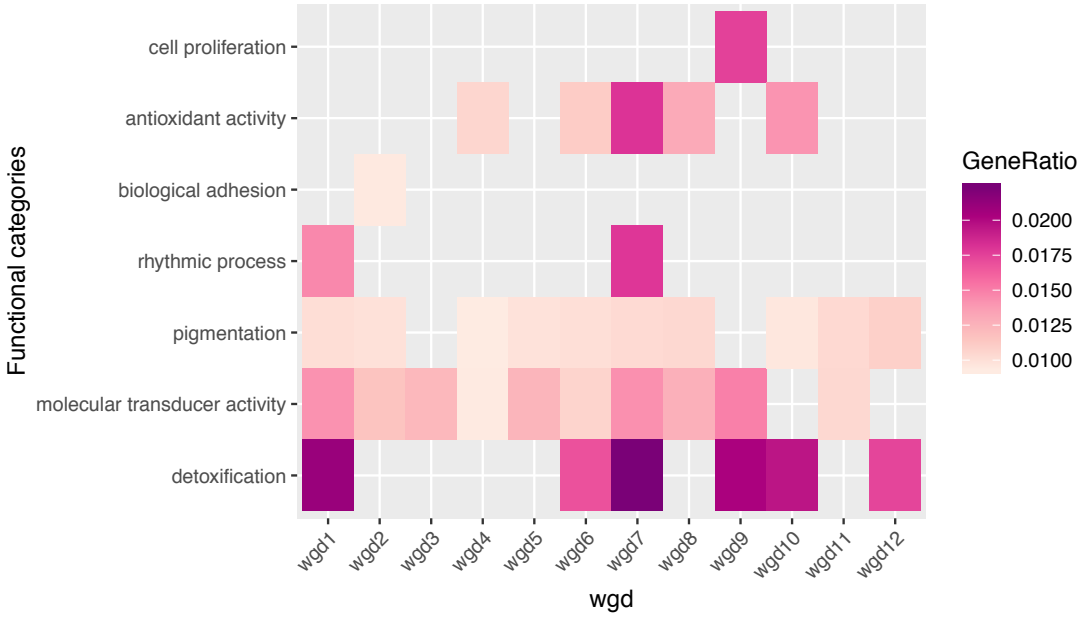

Figure S3. MEDUSA analysis under mixed model with AIC criterion (t17). This figure shows the direct result from MEDUSA analysis with turboMEDUSA as described in Materials and Methods, under the mixed model with AIC criterion. Blue circles indicate the positions of inferred shifts with sizes referring to the frequency of a shift. Because we used the ML tree rather than bootstrap replicates in this analysis, the frequency for a shift will only be 1. Branch colors indicate the median diversification rates. Ages of nodes and branches correspond to the geological time scale at bottom.

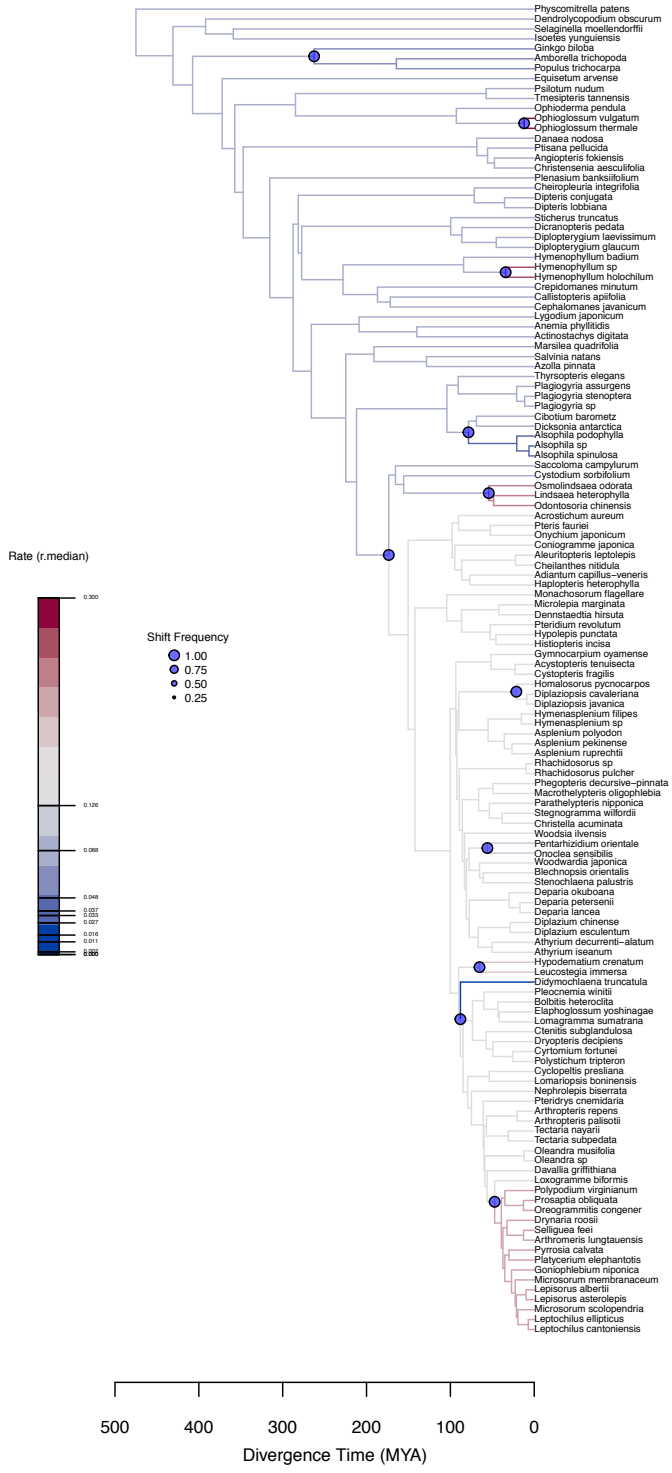

Figure S4. MEDUSA analysis under mixed model with AICC criterion (t18). This figure shows the direct result from MEDUSA analysis with turboMEDUSA as described in Materials and Methods, under the mixed model with corrected AIC criterion. Blue circles indicate the positions of inferred shifts with sizes referring to the frequency of a shift. Because we used the ML tree rather than bootstrap replicates in this analysis, the frequency for a shift will only be 1. Branch colors indicate the median diversification rates. Ages of nodes and branches correspond to the geological time scale at bottom.

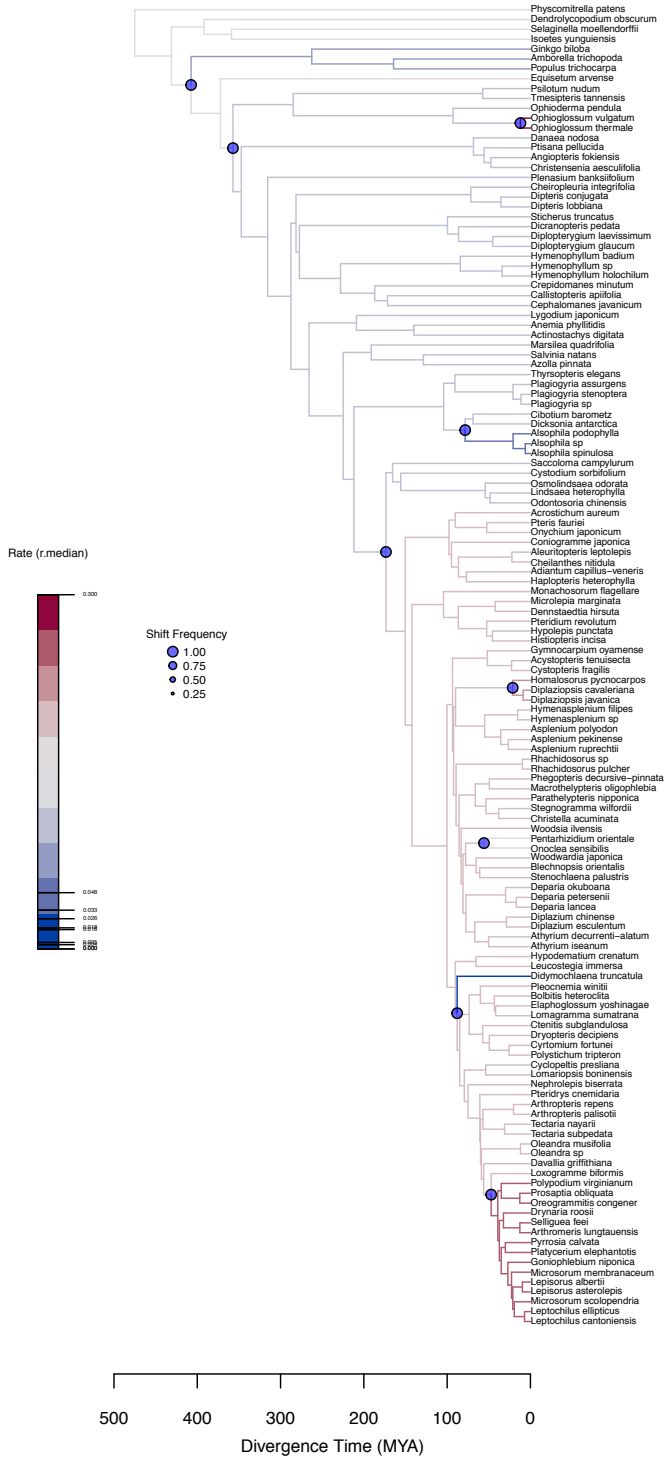

Figure S5. MEDUSA analysis under birth-death model with AIC criterion (t19). This figure shows the direct result from MEDUSA analysis with turboMEDUSA as described in Materials and Methods, under the birth-death model with AIC criterion. Blue circles indicate the positions of inferred shifts with sizes referring to the frequency of a shift. Because we used the ML tree rather than bootstrap replicates in this analysis, the frequency for a shift will only be 1. Branch colors indicate the median diversification rates. Ages of nodes and branches correspond to the geological time scale at bottom.

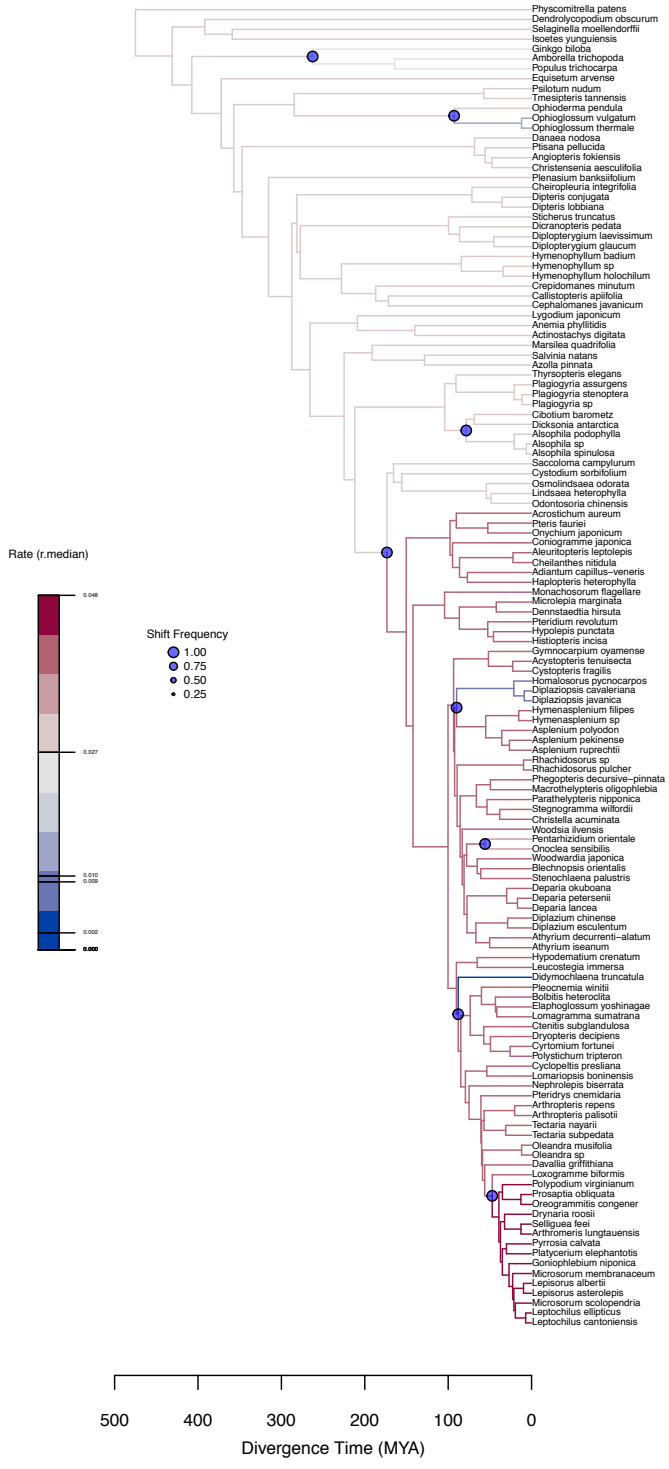

Figure S6. MEDUSA analysis under birth-death model with AICC criterion (t20). This figure shows the direct result from MEDUSA analysis with turboMEDUSA as described in Materials and Methods, under the birth-death model with corrected AIC criterion. Blue circles indicate the positions of inferred shifts with sizes referring to the frequency of a shift. Because we used the ML tree rather than bootstrap replicates in this analysis, the frequency for a shift will only be 1. Branch colors indicate the median diversification rates. Ages of nodes and branches correspond to the geological time scale at bottom.

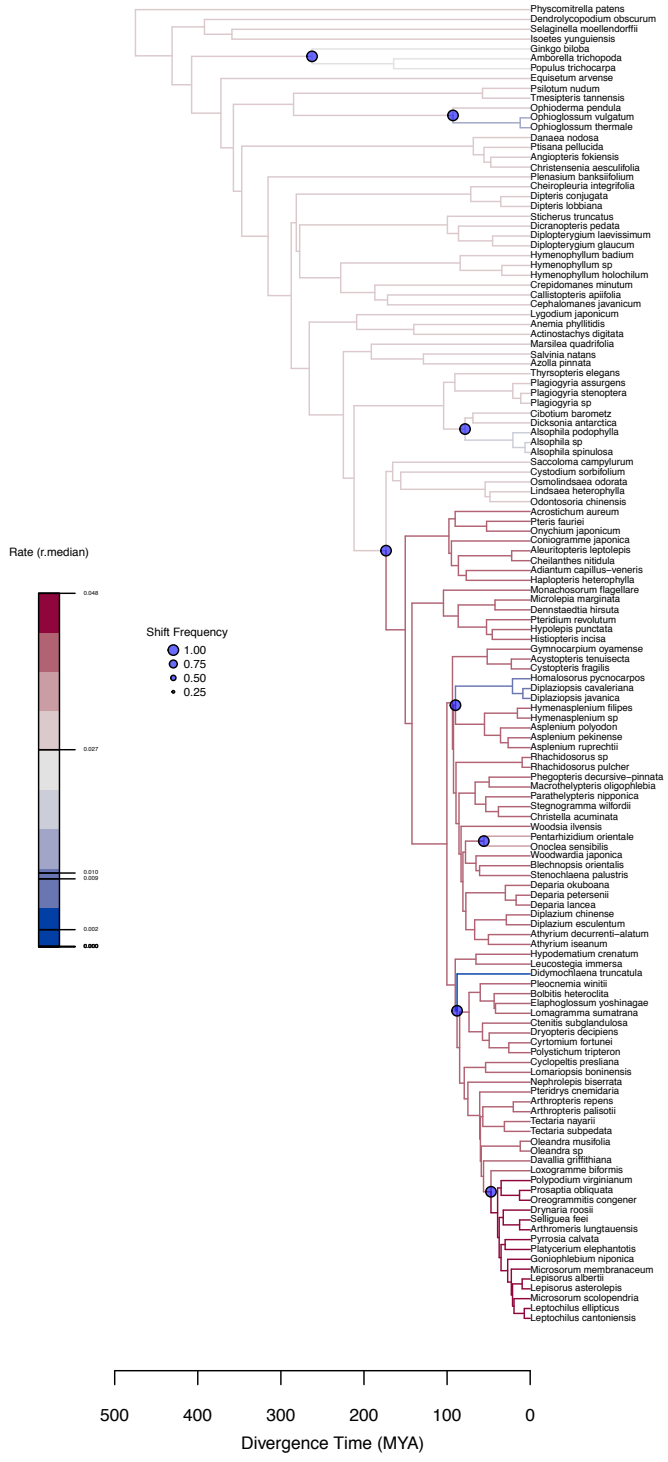

Figure S7. Posterior probability distribution from BAMM. The graphs show the prior (blue bars) and posterior (red bars) probabilities (*y*-axis) of the number (*x*-axis) of rate shifts from BAMM analyses. Figure titles (e.g., t1) indicate tests using different algorithms and parameters and correspond those in Table S1. All graphs show that 8 to 10 shifts have the highest posterior probabilities in most cases.

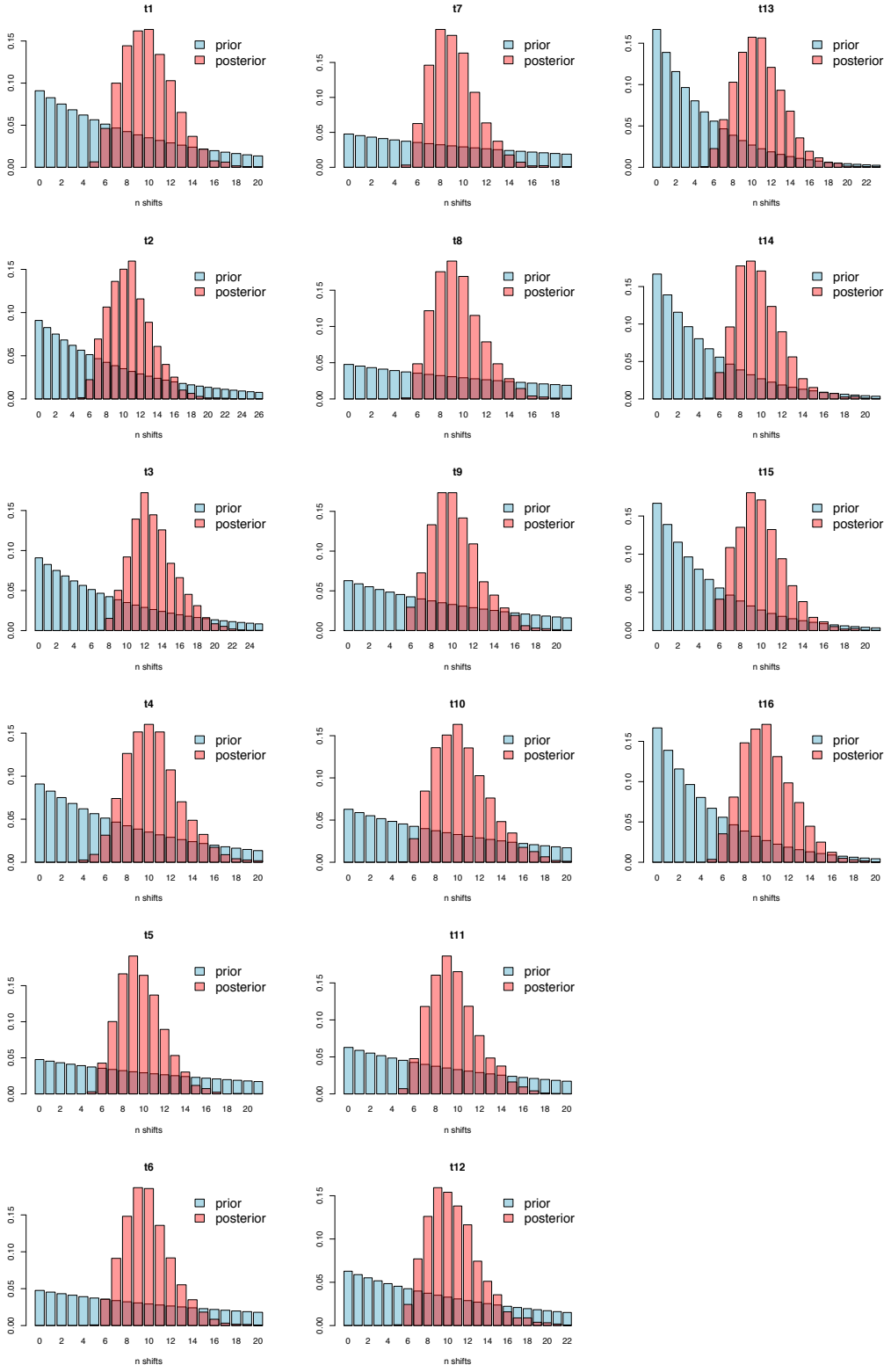

Figure S8. Identification numbers of nodes in Tables S2 and S5. All nodes are numbered in blue boxes and branch tips numbered in yellow boxes, and they correspond to the numbers in Tables S2 and S5.

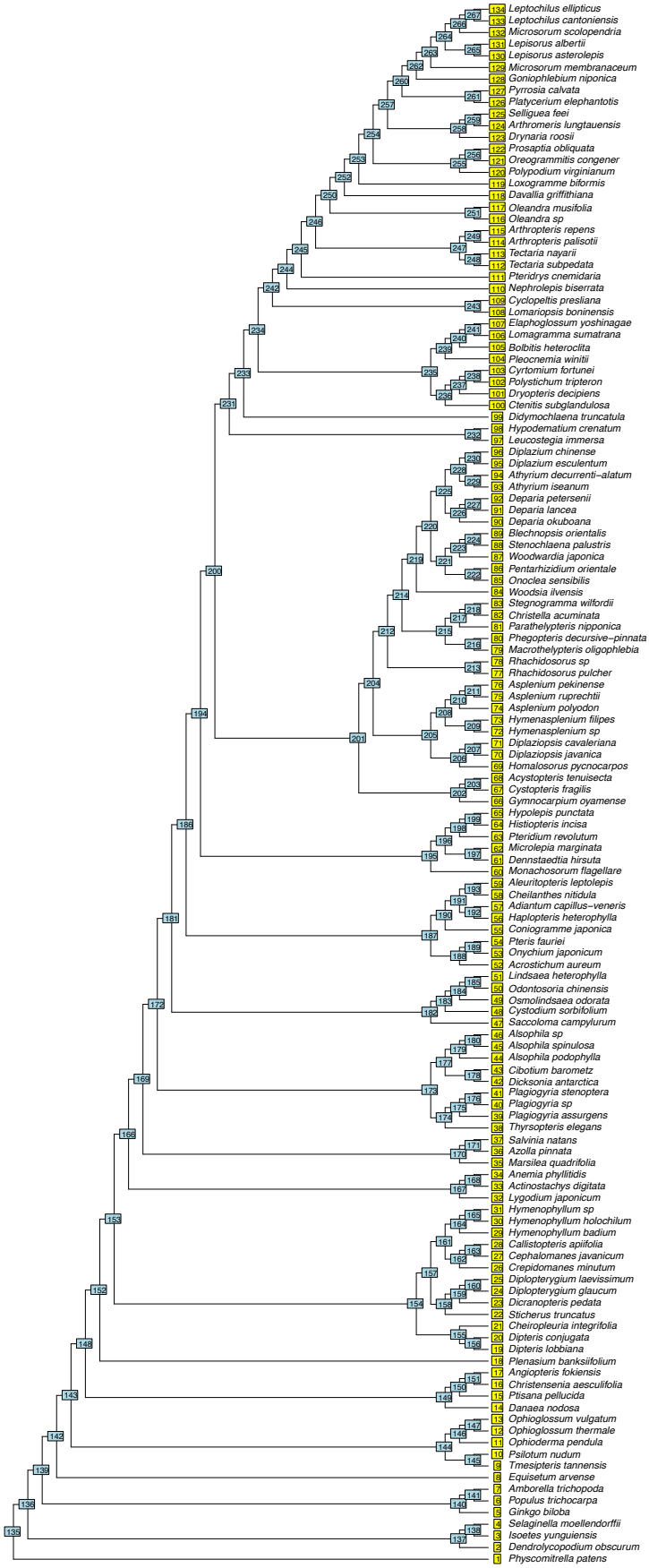

Figure S9. A statistic analysis for potential correlation of WGDs and all shifts. This figure shows the significances (*p* values; *y*-axis) of potential correlations of different shift patterns (summarized from the results using different combinations of parameters in BAMM and MEDUSA analyses; see Table S1 for details) and WGDs. Analyses in this figure are the same as Fig. 3b and are described in Materials and Methods. The only difference is that both up- and downshifts are included here while only up-shifts are considered for Fig. 3.

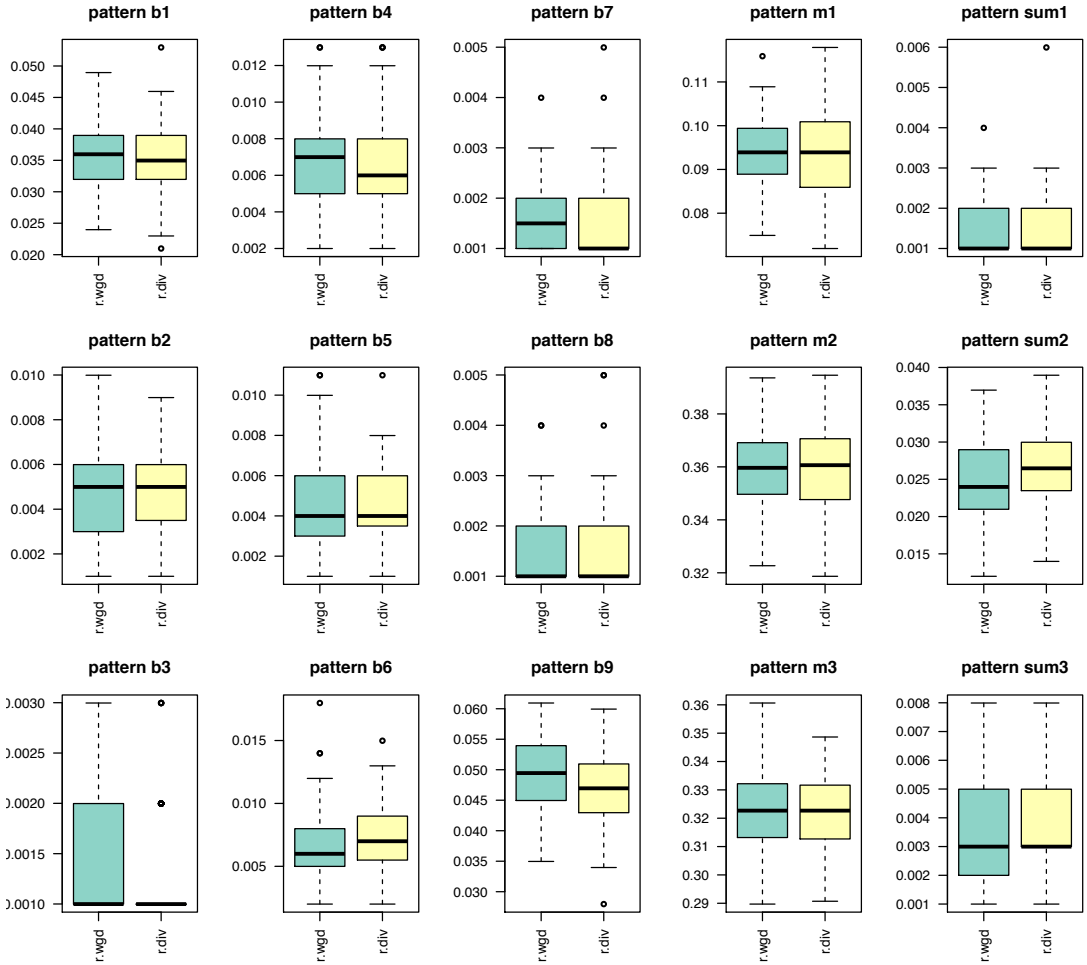

Figure S10. The procedure for WGD identification. (a) A simple illustration for the procedure of determining potential WGD events in this study. (b) Distribution of percent gene duplication from Pure-MCL method (see Materials and Methods for details).

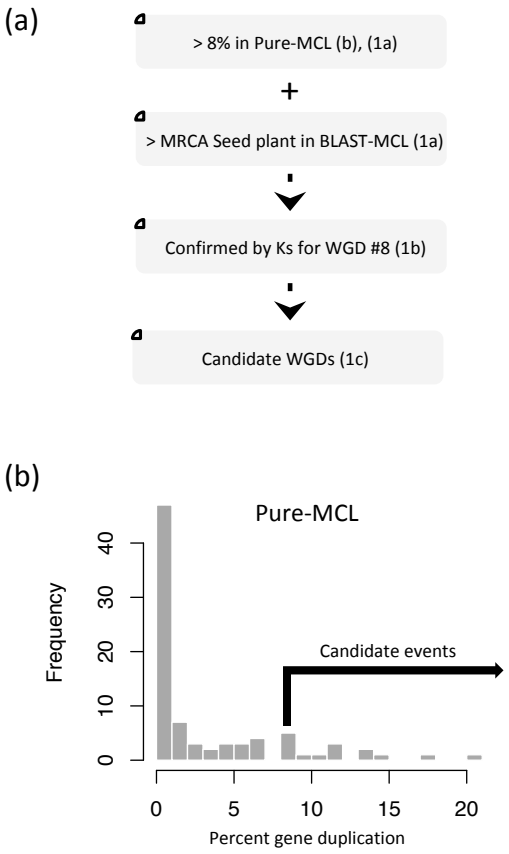

Table S1. Settings for various tests of diversification rate shifts and the summarized shift patterns in this study.

The values of parameters are poissonRatePrior for BAMM and criteria for MEDUSA analyses. Shift patterns correspond to those in Fig. 3B.

Test	method	model	parameter	shift pattern
t1	BAMM	time-constant	0.1	b3
t2	BAMM	time-constant	0.1	b6
t3	BAMM	time-variable	0.1	b4
t4	BAMM	time-variable	0.1	b5
t5	BAMM	time-constant	0.067	b6
t6	BAMM	time-constant	0.067	b7
t7	BAMM	time-variable	0.067	b5
t8	BAMM	time-variable	0.067	b5
t9	BAMM	time-constant	0.05	b3
t10	BAMM	time-constant	0.05	b7
t11	BAMM	time-variable	0.05	b8
t12	BAMM	time-variable	0.05	b6
t13	BAMM	time-constant	0.2	b9
t14	BAMM	time-constant	0.2	b2
t15	BAMM	time-variable	0.2	b1
t16	BAMM	time-variable	0.2	b5
t17	MEDUSA	mixed	AIC	m1
t18	MEDUSA	mixed	AICC	m2
t19	MEDUSA	birth-death	AIC	m3
t20	MEDUSA	birth-death	AICC	m3
	Summary-1	Supported by at least 2 models		sum1
	Summary-2	Supported by both BAMM and MEDUSA		sum2
	Summary-3	All shift nodes		sum3

Table S2. BAMM Results with different parameters.

Node identities correspond to Fig. S7. Test names correspond to those in Table S1. YES, a rate shift is inferred in the corresponding test.

	Time mode	Poisson rate prior	1	186	140	179	254	184	99	136	235	8	149	48	No. shifts
t1	constant	0.1	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES			10
t2	constant	0.1	YES	YES	YES	YES	YES	YES	YES		YES	YES	YES		10
t3	variable	0.1	YES	YES	YES	YES	YES	YES	YES	YES	YES		YES		10
t4	variable	0.1	YES	YES	YES	YES	YES	YES	YES		YES	YES			9
t5	constant	0.05	YES	YES	YES	YES	YES	YES	YES		YES	YES	YES		10
t6	constant	0.05	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES		11
t7	variable	0.05	YES	YES	YES	YES	YES	YES	YES		YES	YES			9
t8	variable	0.05	YES	YES	YES	YES	YES	YES	YES		YES	YES			9
t9	constant	0.067	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES			10
t10	constant	0.067	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES		11
t11	variable	0.067	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES	YES	12
t12	variable	0.067	YES	YES	YES	YES	YES	YES	YES		YES	YES	YES		10
t13	constant	0.2	YES	YES	YES	YES	YES	YES	YES		YES		YES		9
t14	constant	0.2	YES	YES	YES	YES	YES	YES	YES	YES	YES				9
t15	variable	0.2	YES	YES	YES	YES	YES	YES	YES		YES				8
t16	variable	0.2	YES	YES	YES	YES	YES	YES	YES		YES	YES			9
Number of supporting tests			16	16	16	16	16	16	16	7	16	12	8	1	

Table S3. Information of taxa and datasets included in this study.

Order	Family	Species	Number of unigenes	Source of data
Equisetales	Equisetaceae	*Equisetum arvense*	26419	Qi *et al*. (2018)
Equisetales	Equisetaceae	*Equisetum hyemale*	29830	1kp
Equisetales	Equisetaceae	*Equisetum telmateia*	53893	SRR4061752
Ophioglossales	Ophioglossaceae	*Ophioderma pendula*	23508	Qi *et al*. (2018)
Ophioglossales	Ophioglossaceae	*Ophioglossum thermale*	28355	Qi *et al*. (2018)
Ophioglossales	Ophioglossaceae	*Ophioglossum vulgatum*	16821	iplant
Psilotales	Psilotaceae	*Psilotum nudum*	25496	Qi *et al*. (2018)
Psilotales	Psilotaceae	*Tmesipteris tannensis*	27941	Qi *et al*. (2018)
Marattiales	Marattiaceae	*Angiopteris fokiensis*	32006	Qi *et al*. (2018)
Marattiales	Marattiaceae	*Christensenia aesculifolia*	22690	Qi *et al*. (2018)
Marattiales	Marattiaceae	*Danaea nodosa*	43415	Qi *et al*. (2018)
Marattiales	Marattiaceae	*Ptisana pellucida*	30708	Qi *et al*. (2018)
Osmundales	Osmundaceae	*Plenasium banksiifolium*	26583	Qi *et al*. (2018)
Osmundales	Osmundaceae	*Osmunda javanica*	27492	1kp
Osmundales	Osmundaceae	*Osmunda japonica*	28506	Shen *et al.* (2018)
Osmundales	Osmundaceae	*Osmunda cinnamomea*	23695	1kp
Gleicheniales	Dipteridaceae	*Cheiropleuria integrifolia*	40030	Qi *et al*. (2018)
Gleicheniales	Dipteridaceae	*Cheiropleuria integrifolia2*	32032	Qi *et al*. (2018)
Gleicheniales	Dipteridaceae	*Dipteris conjugata*	26737	Qi *et al*. (2018)
Gleicheniales	Dipteridaceae	*Dipteris lobbiana*	65635	Qi *et al*. (2018)
Gleicheniales	Gleicheniaceae	*Dicranopteris pedata*	28792	Qi *et al*. (2018)
Gleicheniales	Gleicheniaceae	*Diplopterygium glaucum*	34270	Qi *et al*. (2018)

Gleicheniales	Gleicheniaceae	*Diplopterygium laevissimum*	34681	Qi *et al*. (2018)
Gleicheniales	Gleicheniaceae	*Sticherus truncatus*	35205	Qi *et al*. (2018)
Hymenophyllales	Hymenophyllaceae	*Callistopteris apiifolia*	25563	Qi *et al*. (2018)
Hymenophyllales	Hymenophyllaceae	*Cephalomanes javanicum*	23603	Qi *et al*. (2018)
Hymenophyllales	Hymenophyllaceae	*Crepidomanes minutum*	19118	Qi *et al*. (2018)
Hymenophyllales	Hymenophyllaceae	*Hymenophyllum badium*	78996	Qi *et al*. (2018)
Hymenophyllales	Hymenophyllaceae	*Hymenophyllum holochilum*	51049	Qi *et al*. (2018)
Hymenophyllales	Hymenophyllaceae	*Hymenophyllum sp*	28789	Qi *et al*. (2018)
Schizaeales	Schizaeaceae	*Actinostachys digitata*	38503	Qi *et al*. (2018)
Schizaeales	Anemiaceae	*Anemia phyllitidis*	30786	Qi *et al*. (2018)
Schizaeales	Lygodiaceae	*Lygodium japonicum*	21975	Qi *et al*. (2018)
Salviniales	Marsileaceae	*Marsilea quadrifolia*	26751	Qi *et al*. (2018)
Salviniales	Salviniaceae	*Azolla pinnata*	31501	Qi *et al*. (2018)
Salviniales	Salviniaceae	*Salvinia natans*	45028	Qi *et al*. (2018)
Cyatheales	Cyatheaceae	*Alsophila podophylla*	35590	Qi *et al*. (2018)
Cyatheales	Cyatheaceae	*Alsophila sp*	33655	Qi *et al*. (2018)
Cyatheales	Cyatheaceae	*Alsophila spinulosa*	56985	Qi *et al*. (2018)
Cyatheales	Cibotiaceae	*Cibotium barometz*	52891	Qi *et al*. (2018)
Cyatheales	Dicksoniaceae	*Dicksonia antarctica*	37070	Qi *et al*. (2018)
Cyatheales	Plagiogyriaceae	*Plagiogyria assurgens*	21829	Qi *et al*. (2018)
Cyatheales	Plagiogyriaceae	*Plagiogyria sp*	67542	Qi *et al*. (2018)
Cyatheales	Plagiogyriaceae	*Plagiogyria stenoptera*	57227	Qi *et al*. (2018)
Cyatheales	Thyrsopteridaceae	*Thyrsopteris elegans*	37655	Qi *et al*. (2018)
Polypodiales	Saccolomataceae	*Saccoloma campylurum*	31735	Qi *et al*. (2018)
Polypodiales	Cystodiaceae	*Cystodium sorbifolium*	33008	Qi *et al*. (2018)

Polypodiales	Lindsaeaceae	*Lindsaea heterophylla*	34154	Qi *et al*. (2018)
Polypodiales	Lindsaeaceae	*Odontosoria chinensis*	47750	Qi *et al*. (2018)
Polypodiales	Lindsaeaceae	*Osmolindsaea odorata*	37837	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Acrostichum aureum*	26127	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Adiantum capillus-veneris*	29351	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Aleuritopteris leptolepis*	29695	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Cheilanthes nitidula*	24252	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Coniogramme japonica*	38958	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Haplopteris heterophylla*	27402	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Onychium japonicum*	41874	Qi *et al*. (2018)
Polypodiales	Pteridaceae	*Pteris fauriei*	67189	Qi *et al*. (2018)
Polypodiales	Dennstaedtiaceae	*Dennstaedtia hirsuta*	26536	Qi *et al*. (2018)
Polypodiales	Dennstaedtiaceae	*Histiopteris incisa*	30577	Qi *et al*. (2018)
Polypodiales	Dennstaedtiaceae	*Hypolepis punctata*	29620	Qi *et al*. (2018)
Polypodiales	Dennstaedtiaceae	*Microlepia marginata*	42447	Qi *et al*. (2018)
Polypodiales	Dennstaedtiaceae	*Monachosorum flagellare*	42392	Qi *et al*. (2018)
Polypodiales	Dennstaedtiaceae	*Pteridium revolutum*	36988	Qi *et al*. (2018)
Polypodiales	Cystopteridaceae	*Acystopteris tenuisecta*	37035	Qi *et al*. (2018)
Polypodiales	Cystopteridaceae	*Cystopteris fragilis*	27441	Qi *et al*. (2018)
Polypodiales	Cystopteridaceae	*Gymnocarpium oyamense*	35590	Qi *et al*. (2018)
Polypodiales	Diplaziopsidaceae	*Diplaziopsis cavaleriana*	37070	Qi *et al*. (2018)
Polypodiales	Diplaziopsidaceae	*Diplaziopsis javanica*	32632	Qi *et al*. (2018)
Polypodiales	Diplaziopsidaceae	*Homalosorus pycnocarpos*	31261	Rothfels lab
Polypodiales	Aspleniaceae	*Asplenium pekinense*	34933	Qi *et al*. (2018)
Polypodiales	Aspleniaceae	*Asplenium polyodon*	41804	Qi *et al*. (2018)

Polypodiales	Aspleniaceae	*Asplenium ruprechtii*	24490	Qi *et al*. (2018)
Polypodiales	Aspleniaceae	*Hymenasplenium filipes*	29705	Qi *et al*. (2018)
Polypodiales	Aspleniaceae	*Hymenasplenium sp*	18865	Qi *et al*. (2018)
Polypodiales	Rhachidosoraceae	*Rhachidosorus pulcher*	30928	Qi *et al*. (2018)
Polypodiales	Rhachidosoraceae	*Rhachidosorus sp*	35481	Qi *et al*. (2018)
Polypodiales	Thelypteridaceae	*Christella acuminata*	32060	Qi *et al*. (2018)
Polypodiales	Thelypteridaceae	*Macrothelypteris oligophlebia*	29493	Qi *et al*. (2018)
Polypodiales	Thelypteridaceae	*Parathelypteris nipponica*	37655	Qi *et al*. (2018)
Polypodiales	Thelypteridaceae	*Phegopteris decursive-pinnata*	34978	Qi *et al*. (2018)
Polypodiales	Thelypteridaceae	*Stegnogramma wilfordii*	55201	Qi *et al*. (2018)
Polypodiales	Woodsiaceae	*Woodsia ilvensis*	34189	Qi *et al*. (2018)
Polypodiales	Onocleaceae	*Onoclea sensibilis*	28302	Qi *et al*. (2018)
Polypodiales	Onocleaceae	*Pentarhizidium orientale*	39001	Qi *et al*. (2018)
Polypodiales	Blechnaceae	*Blechnopsis orientalis*	30101	Qi *et al*. (2018)
Polypodiales	Blechnaceae	*Stenochlaena palustris*	47750	Qi *et al*. (2018)
Polypodiales	Blechnaceae	*Woodwardia japonica*	43456	Qi *et al*. (2018)
Polypodiales	Athyriaceae	*Athyrium decurrenti-alatum*	24813	Qi *et al*. (2018)
Polypodiales	Athyriaceae	*Athyrium iseanum*	34957	Qi *et al*. (2018)
Polypodiales	Athyriaceae	*Deparia lancea*	33354	Qi *et al*. (2018)
Polypodiales	Athyriaceae	*Deparia okuboana*	33683	Qi *et al*. (2018)
Polypodiales	Athyriaceae	*Deparia petersenii*	45551	Qi *et al*. (2018)
Polypodiales	Athyriaceae	*Diplazium chinense*	25592	Qi *et al*. (2018)
Polypodiales	Athyriaceae	*Diplazium esculentum*	25563	Qi *et al*. (2018)
Polypodiales	Hypodematiaceae	*Hypodematium crenatum*	30987	Qi *et al*. (2018)
Polypodiales	Hypodematiaceae	*Leucostegia immersa*	25724	Qi *et al*. (2018)

Polypodiales	Didymochlaenaceae	*Didymochlaena truncatula*	32138	Qi *et al*. (2018)
Polypodiales	Didymochlaenaceae	*Didymochlaena truncatula2*	35364	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Bolbitis heteroclita*	47810	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Ctenitis subglandulosa*	31109	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Cyrtomium fortunei*	43135	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Dryopteris decipiens*	26112	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Elaphoglossum yoshinagae*	60399	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Lomagramma sumatrana*	27450	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Pleocnemia winitii*	31702	Qi *et al*. (2018)
Polypodiales	Dryopteridaceae	*Polystichum tripteron*	31797	Qi *et al*. (2018)
Polypodiales	Lomariopsidaceae	*Cyclopeltis presliana*	29872	Qi *et al*. (2018)
Polypodiales	Lomariopsidaceae	*Lomariopsis boninensis*	31685	Qi *et al*. (2018)
Polypodiales	Nephrolepidaceae	*Nephrolepis biserrata*	28252	Qi *et al*. (2018)
Polypodiales	Tectariaceae	*Arthropteris palisotii*	36707	Qi *et al*. (2018)
Polypodiales	Tectariaceae	*Arthropteris repens*	24579	Qi *et al*. (2018)
Polypodiales	Tectariaceae	*Pteridrys cnemidaria*	27689	Qi *et al*. (2018)
Polypodiales	Tectariaceae	*Tectaria nayarii*	33899	Qi *et al*. (2018)
Polypodiales	Tectariaceae	*Tectaria subpedata*	38125	Qi *et al*. (2018)
Polypodiales	Oleandraceae	*Oleandra musifolia*	27074	Qi *et al*. (2018)
Polypodiales	Oleandraceae	*Oleandra sp*	34642	Qi *et al*. (2018)
Polypodiales	Davalliaceae	*Davallia griffithiana*	30155	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Arthromeris lungtauensis*	36871	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Drynaria roosii*	39309	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Goniophlebium niponica*	49960	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Lepisorus albertii*	46204	Qi *et al*. (2018)

Polypodiales	Polypodiaceae	*Lepisorus asterolepis*	19368	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Leptochilus cantoniensis*	26106	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Leptochilus ellipticus*	30581	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Loxogramme biformis*	27046	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Microsorum membranaceum*	28136	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Microsorum scolopendria*	30967	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Oreogrammitis congener*	52099	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Platycerium elephantotis*	27105	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Polypodium virginianum*	28943	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Prosaptia obliquata*	33469	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Pyrrosia calvata*	68443	Qi *et al*. (2018)
Polypodiales	Polypodiaceae	*Selliguea feei*	51896	Qi *et al*. (2018)
Funariales	Funariaceae	*Physcomitrella patens*	28200	Rensing *et al*., (2008)
Lycopodiales	Lycopodiaceae	*Dendrolycopodium obscurum*	31936	Qi *et al*. (2018)
Isoëtales	Isoëtaceae	*Isoëtes yunguiensis*	33967	Qi *et al*. (2018)
Selaginellales	Selaginellaceae	*Selaginella moellendorffii*	21382	Banks *et al*., (2011)
Ginkgoales	Ginkgoaceae	*Ginkgo biloba*	25028	Zeng *et al*. (2014)
Amborellales	Amborellaceae	*Amborella trichopoda*	20974	Amborella Genome Project, 2013
Malpighiales	Salicaceae	*Populus trichocarpa*	41017	Tuskan *et al*., (2006)

Table S4. Species richness data used in MEDUSA and BAMM analyses.

Family	Richness	Sampled number
Anemiaceae	115	1
Aspleniaceae	730	5
Athyriaceae	650	7
Blechnaceae	265	3
Cibotiaceae	9	1
Cyatheaceae	643	3
Cystodiaceae	1	1
Cystopteridaceae	37	3
Davalliaceae	65	1
Dennstaedtiaceae	265	6
Dicksoniaceae	35	1
Didymochlaenaceae	1	1
Diplaziopsidaceae	4	3
Dipteridaceae	11	3
Dryopteridaceae	2115	8
Equisetaceae	15	1
Gleicheniaceae	157	4
Hymenophyllaceae	434	6
Hypodematiaceae	22	2
Lindsaeaceae	234	3
Lomariopsidaceae	69	2
Lygodiaceae	40	1
Marattiaceae	111	4
Marsileaceae	61	1
Nephrolepidaceae	19	1
Oleandraceae	15	2
Onocleaceae	5	2
Ophioglossaceae	112	3
Osmundaceae	18	1
Plagiogyriaceae	15	3
Polypodiaceae	1652	16
Psilotaceae	17	2
Pteridaceae	1211	8
Rhachidosoraceae	8	2
Saccolomataceae	18	1
Salviniaceae	21	2
Schizaeaceae	35	1
Tectariaceae	250	5
Thelypteridaceae	1034	5
Thyrsopteridaceae	1	1

Woodsiaceae	39	1
	Not included	
Culcitaceae	2	
Desmophlebiaceae	2	
Hemidictyaceae	1	
Lonchitidaceae	2	
Loxsomataceae	2	
Matoniaceae	4	
Metaxyaceae	6	
	Outgroups	
Seed plants	296433	3
Lycophytes	1290	3
Bryophytes	12000	1

Table S5. Data points for linear regression of peak Ks and ages in this study.
Nodes correspond to Fig. S7.

node	age	peak_ks	species1	species2
188	90.27438	0.585566329	Acrostichum_aureum	Pteris_fauriei
181	173.461055	0.931301922	Acrostichum_aureum	Saccoloma_campylurum
191	86.398128	0.516143341	Aleuritopteris_leptolepis	Adiantum_capillus-veneris
187	97.817885	0.60403552	Aleuritopteris_leptolepis	Pteris_fauriei
179	20.832014	0.067171157	Alsophila_podophylla	Alsophila_sp
179	20.832014	0.066197176	Alsophila_podophylla	Alsophila_spinulosa
177	78.393745	0.168859108	Alsophila_podophylla	Dicksonia_antarctica
177	78.393745	0.173407724	Alsophila_sp	Dicksonia_antarctica
173	104.150398	0.218421223	Alsophila_sp	Plagiogyria_stenoptera
247	57.011732	0.212894286	Arthropteris_repens	Tectaria_nayarii
205	89.904436	0.415892495	Asplenium_pekinense	Diplaziopsis_cavaleriana
208	54.992205	0.351190475	Asplenium_pekinense	Hymenasplenium_filipes
210	35.690573	0.252139648	Asplenium_polyodon	Asplenium_pekinense
240	43.346919	0.209212163	Bolbitis_heteroclita	Elaphoglossum_yoshinagae
155	71.509011	0.183422424	Cheiropleuria_integrifolia	Dipteris_conjugata
155	71.509011	0.177921232	Cheiropleuria_integrifolia	Dipteris_lobbiana
173	104.150398	0.216030455	Cibotium_barometz	Plagiogyria_stenoptera
173	104.150398	0.186270039	Cibotium_barometz	Thyrsopteris_elegans
190	94.781428	0.466427963	Coniogramme_japonica	Aleuritopteris_leptolepis
162	186.94119	0.444764741	Crepidomanes_minutum	Callistopteris_apiifolia
157	277.354898	0.916104252	Crepidomanes_minutum	Sticherus_truncatus
234	84.896841	0.325285495	Ctenitis_subglandulosa	Arthromeris_lungtauensis
236	57.385521	0.127370752	Ctenitis_subglandulosa	Cyrtomium_fortunei
235	73.611107	0.203444611	Ctenitis_subglandulosa	Pleocnemia_winitii
183	155.750106	0.833605726	Cystodium_sorbifolium	Lindsaea_heterophylla
183	155.750106	0.851169889	Cystodium_sorbifolium	Osmolindsaea_odorata
149	68.422962	0.226393125	Danaea_nodosa	Angiopteris_fokiensis
149	68.422962	0.21951073	Danaea_nodosa	Ptisana_pellucida
143	357.130382	1.383391202	Danaea_nodosa	Tmesipteris_tannensis
252	56.153711	0.347647924	Davallia_griffithiana	Leptochilus_cantoniensis
226	29.877476	0.066102669	Deparia_okuboana	Deparia_petersenii
172	211.961639	0.521489841	Dicksonia_antarctica	Cystodium_sorbifolium
173	104.150398	0.191634207	Dicksonia_antarctica	Plagiogyria_stenoptera
173	104.150398	0.160567104	Dicksonia_antarctica	Thyrsopteris_elegans
159	86.271743	0.22619494	Dicranopteris_pedata	Diplopterygium_laevissimum
233	88.01185	0.40628718	Didymochlaena_truncatula	Leptochilus_cantoniensis
231	90.173744	0.274054935	Didymochlaena_truncatula	Leucostegia_immersa
181	173.461055	1.277637899	Didymochlaena_truncatula	Lindsaea_heterophylla
228	66.822666	0.081885096	Diplazium_chinense	Athyrium_decurrenti-alatum

225	77.419527	0.110096403	Diplazium_chinense	Deparia_petersenii
220	80.96209	0.156229842	Diplazium_chinense	Woodwardia_japonica
154	281.438293	0.672977876	Diplopterygium_laevissimum	Cheiropleuria_integrifolia
153	287.533487	1.456626376	Dipteris_conjugata	Leptochilus_cantoniensis
258	32.419281	0.129840228	Drynaria_roosii	Arthromeris_lungtauensis
237	49.340499	0.113291413	Dryopteris_decipiens	Cyrtomium_fortunei
235	73.611107	0.285097768	Elaphoglossum_yoshinagae	Cyrtomium_fortunei
142	372.187585	1.827006285	Equisetum_arvense	Leptochilus_cantoniensis
142	372.187585	1.623301173	Equisetum_arvense	Tmesipteris_tannensis
262	27.067176	0.147846455	Goniophlebium_niponica	Leptochilus_cantoniensis
202	51.737272	0.095682648	Gymnocarpium_oyamense	Acystopteris_tenuisecta
194	142.268607	0.417236421	Gymnocarpium_oyamense	Monachosorum_flagellare
206	21.295635	0.063786249	Homalosorus_pycnocarpos	Diplaziopsis_cavaleriana
201	93.512914	0.13757314	Homalosorus_pycnocarpos	Gymnocarpium_oyamense
164	84.242912	0.10378816	Hymenophyllum_badium	Hymenophyllum_sp
161	228.000001	0.284886915	Hymenophyllum_sp	Callistopteris_apiifolia
157	277.354898	0.624454887	Hymenophyllum_sp	Diplopterygium_laevissimum
186	150.345423	0.988747243	Leptochilus_cantoniensis	Aleuritopteris_leptolepis
172	211.961639	0.899410611	Leptochilus_cantoniensis	Alsophila_sp
166	265.841667	1.581196975	Leptochilus_cantoniensis	Anemia_phyllitidis
148	347.097976	1.699968003	Leptochilus_cantoniensis	Angiopteris_fokiensis
257	37.332362	0.167995603	Leptochilus_cantoniensis	Arthromeris_lungtauensis
153	287.533487	1.44459615	Leptochilus_cantoniensis	Diplopterygium_laevissimum
231	90.173744	0.400136119	Leptochilus_cantoniensis	Hypodematium_crenatum
194	142.268607	0.621713065	Leptochilus_cantoniensis	Hypolepis_punctata
181	173.461055	1.347788442	Leptochilus_cantoniensis	Lindsaea_heterophylla
143	357.130382	1.776890278	Leptochilus_cantoniensis	Ophioglossum_thermale
169	224.798629	1.594297866	Leptochilus_cantoniensis	Salvinia_natans
200	100.228513	0.430474095	Leptochilus_cantoniensis	Woodwardia_japonica
200	100.228513	0.267441159	Leucostegia_immersa	Gymnocarpium_oyamense
166	265.841667	1.602465355	Lindsaea_heterophylla	Anemia_phyllitidis
148	347.097976	1.678356949	Lindsaea_heterophylla	Angiopteris_fokiensis
153	287.533487	1.507722529	Lindsaea_heterophylla	Diplopterygium_laevissimum
185	48.414494	0.282047422	Lindsaea_heterophylla	Odontosoria_chinensis
184	54.205417	0.355518952	Lindsaea_heterophylla	Osmolindsaea_odorata
169	224.798629	1.607414493	Lindsaea_heterophylla	Salvinia_natans
253	47.17812	0.320721657	Loxogramme_biformis	Leptochilus_cantoniensis
167	208.875789	1.3598391	Lygodium_japonicum	Anemia_phyllitidis
153	287.533487	1.362727678	Lygodium_japonicum	Dipteris_lobbiana
166	265.841667	1.79690129	Marsilea_quadrifolia	Lygodium_japonicum
170	191.153374	1.659169572	Marsilea_quadrifolia	Salvinia_natans
263	22.706326	0.112771177	Microsorum_membranaceum	Leptochilus_cantoniensis
266	19.457501	0.134710253	Microsorum_scolopendria	Leptochilus_cantoniensis

186	150.345423	0.76285428	Monachosorum_flagellare	Acrostichum_aureum
195	104.37687	0.269364911	Monachosorum_flagellare	Hypolepis_punctata
244	74.970709	0.444453794	Nephrolepis_biserrata	Leptochilus_cantoniensis
184	54.205417	0.353640579	Odontosoria_chinensis	Osmolindsaea_odorata
146	92.887675	0.453620537	Ophioderma_pendula	Ophioglossum_thermale
146	92.887675	0.454876965	Ophioderma_pendula	Ophioglossum_vulgatum
144	284.755896	1.370505523	Ophioderma_pendula	Tmesipteris_tannensis
143	357.130382	1.638420769	Ophioglossum_thermale	Dipteris_conjugata
144	284.755896	1.39822796	Ophioglossum_thermale	Psilotum_nudum
144	284.755896	1.363692005	Ophioglossum_thermale	Tmesipteris_tannensis
184	54.205417	0.349132104	Osmolindsaea_odorata	Lindsaea_heterophylla
184	54.205417	0.343280612	Osmolindsaea_odorata	Odontosoria_chinensis
217	53.463939	0.097580003	Parathelypteris_nipponica	Stegnogramma_wilfordii
175	20.933991	0.070403897	Plagiogyria_assurgens	Plagiogyria_sp
175	20.933991	0.073092823	Plagiogyria_assurgens	Plagiogyria_stenoptera
148	347.097976	1.225936952	Plenasium_banksiifolium	Danaea_nodosa
152	315.43628	0.798214862	Plenasium_banksiifolium	Dipteris_lobbiana
152	315.43628	1.53658909	Plenasium_banksiifolium	Leptochilus_cantoniensis
152	315.43628	1.536106232	Plenasium_banksiifolium	Lindsaea_heterophylla
239	60.114386	0.235301625	Pleocnemia_winitii	Elaphoglossum_yoshinagae
255	34.97788	0.185440043	Polypodium_virginianum	Prosaptia_obliquata
143	357.130382	1.761494367	Psilotum_nudum	Lindsaea_heterophylla
145	57.386509	0.217867897	Psilotum_nudum	Tmesipteris_tannensis
198	52.552321	0.156177107	Pteridium_revolutum	Hypolepis_punctata
245	60.725257	0.310703079	Pteridrys_cnemidaria	Leptochilus_cantoniensis
150	55.632545	0.117773474	Ptisana_pellucida	Angiopteris_fokiensis
182	165.495337	0.879007886	Saccoloma_campylurum	Lindsaea_heterophylla
172	211.961639	0.464450576	Saccoloma_campylurum	Thyrsopteris_elegans
215	66.248346	0.139434224	Stegnogramma_wilfordii	Phegopteris_decursive-pinnata
158	99.600001	0.288911777	Sticherus_truncatus	Dicranopteris_pedata
158	99.600001	0.230321507	Sticherus_truncatus	Diplopterygium_glaucum
158	99.600001	0.23443635	Sticherus_truncatus	Diplopterygium_laevissimum
154	281.438293	0.679088101	Sticherus_truncatus	Dipteris_lobbiana
169	224.798629	1.398910099	Thyrsopteris_elegans	Marsilea_quadrifolia
174	90.669238	0.155923326	Thyrsopteris_elegans	Plagiogyria_assurgens
174	90.669238	0.161665004	Thyrsopteris_elegans	Plagiogyria_stenoptera
219	83.186971	0.189946902	Woodsia_ilvensis	Woodwardia_japonica
201	93.512914	0.171065902	Woodwardia_japonica	Acystopteris_tenuisecta
200	100.228513	0.383542932	Woodwardia_japonica	Arthromeris_lungtauensis
204	92.271032	0.460204237	Woodwardia_japonica	Asplenium_pekinense
223	65.1983	0.172281628	Woodwardia_japonica	Blechnopsis_orientalis
194	142.268607	0.431987647	Woodwardia_japonica	Hypolepis_punctata
221	77.833641	0.15891412	Woodwardia_japonica	Pentarhizidium_orientale

200	100.228513	0.307704746	Woodwardia_japonica	Pleocnemia_winitii
200	100.228513	0.432094422	Woodwardia_japonica	Pyrrosia_calvata
214	85.673862	0.198081192	Woodwardia_japonica	Stegnogramma_wilfordii