the TIMETREE of LIFE

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Ferns

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Abstract

Ophioglossoids, whisk ferns, marattioids, horsetails, and leptosporangiates form a well-supported monophyletic group of seed-free vascular plants sometimes referred to as monilophytes (ferns). With approximately 10,000 species, ferns are the closest relatives of the seed plants. All five of the major extant fern lineages were present by the end of the Carboniferous (~299 million years ago, Ma). The Permian (299–251 Ma) and Triassic (251–200 Ma) witnessed the establishment of many leptosporangiate lineages. But despite these ancient origins, several successive radiations in the Cretaceous Period (146–66 Ma) and Cenozoic Era (66–0 Ma) generated most of modern fern diversity.

Extant ferns form a monophyletic group of vascular plants (*1–8*) that number about 10,000 species divided unequally among five major lineages—ophioglossoids, whisk ferns, marattioids, horsetails, and leptosporangiates—and recognized in 11 orders and 37 families (*9*) (Fig. 1). These lineages are all spore-bearing and "seed-free" (*10*). By tradition they were previously classified together with lycophytes (the only other seed-free lineage of vascular plants) under the umbrella-term "pteridophytes" or "ferns and fern allies." However, it is now clear that the five fern lineages together (without lycophytes) are the closest relatives of the seed plants (*1–8*), a position supported by the presence of euphylls—leaves with marginal or apical meristems and an associated leaf gap in the vascular stele (*11*)—and a 30-kb inversion in the plastid genome (*12*). An obvious morphological shared-derived character for ferns is lacking, but the monophyly of this clade is supported by sperm ultrastructure (*6*), sporophyte anatomy (*1*), and DNA sequence data (*2–5, 8*). Here, we provide an overview of fern relationships and divergence time estimates for the major groups.

Within ferns, the first dichotomy separates two robustly supported clades: one consisting of ophioglossoids plus whisk ferns and the other containing horsetails, marattioids, and leptosporangiates (*4, 13*). Whisk ferns and ophioglossoids are both relatively small lineages (~100 species total in two families and six genera; *9*) and both have a poor fossil record. Because of the extent of morphological simplification in both families, their close relationship was only recently recognized from molecular phylogenetic studies (*2, 3*). Horsetails, marattioids, and leptosporangiates are well supported as a clade, but relationships among these lineages remain equivocal (*2–5, 8, 13*). Horsetails are an ancient group of plants with fossil relatives dating back to the Late Devonian (385–359 Ma), but today consist of a mere 15 living species (all in *Equisetum*). The length of the branch (in terms of the number of DNA substitutions per site) leading to its few living species (with no other living taxa to sample) may be a complicating factor in determining the exact relationship of *Equisetum* to other fern lineages (*8, 14*). The marattioid ferns are also an ancient group, first appearing in the middle Carboniferous. In the Late Carboniferous and Permian, several large marattioid representatives originated, including *Psaronius*, which reached heights of about 8 m. Marattioids experienced a decline in diversity since the end of the Paleozoic (251 Ma), and today are represented by about 150 species in at least four genera (*9*).

The best-known and largest lineage of ferns is the leptosporangiates (Fig. 1), a monophyletic group of

Fig. 1 A leptosporangiate fern (*Matonia pectinata* R. Br.) from Malaysia. Credit: K. M. Pryer.
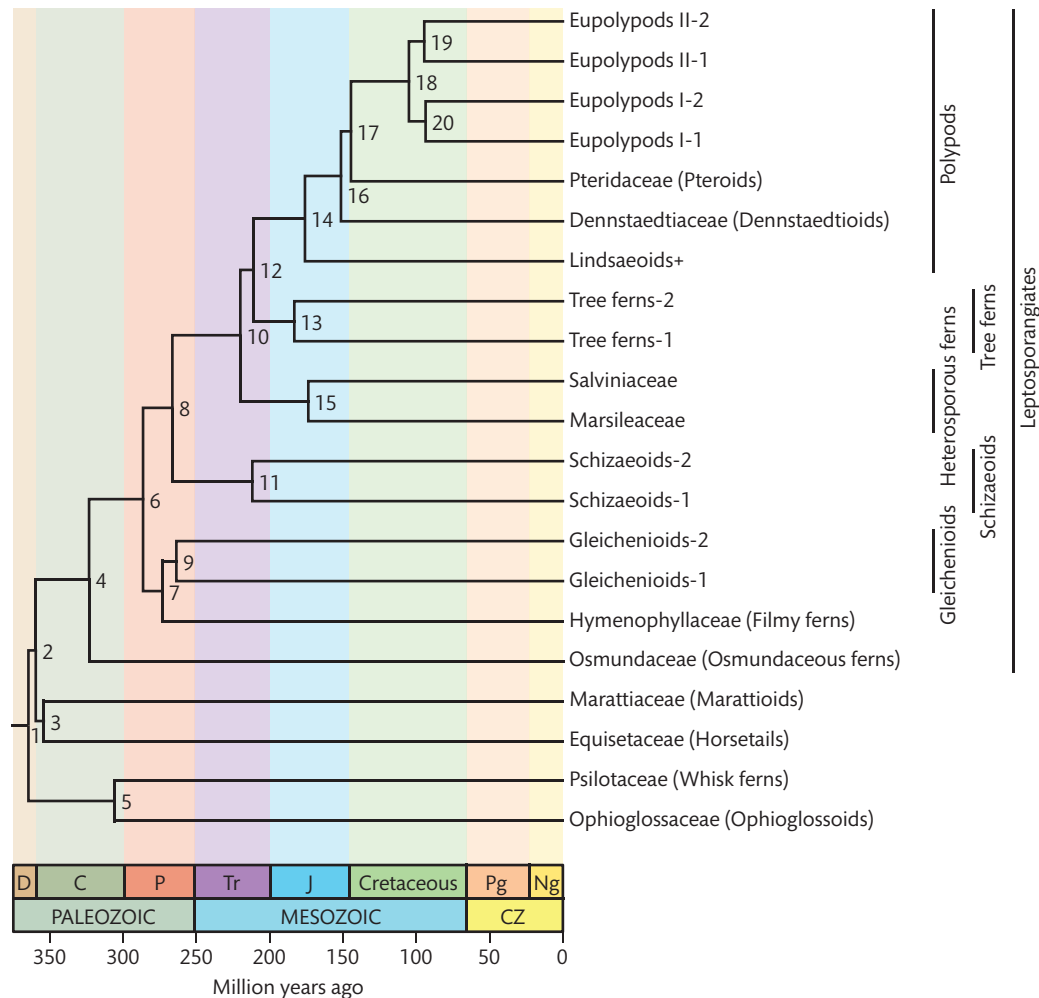
Fig. 2 A timetree of ferns. Divergence times are shown in Table 1. All numbered nodes (except 3, 7, and 17) are well supported. Gleichenioids-1 = Gleicheniaceae; Gleichenioids-2 = Dipteridaceae and Matoniaceae; Schizaeoids-1 = Lygodiaceae; Schizaeoids-2 = Anemiaceae and Schizaeaceae; Tree ferns-1 = Cibotiaceae, Cyatheaceae, Dicksoniaceae, and Metaxyaceae; Tree ferns-2 = Culcitaceae (not sampled in *4*), Loxomataceae, Plagiogyriaceae, and Thyrsopteridaceae (not sampled in *4*); Lindsaeoids+ = Lindsaeaceae and Saccolomataceae;

Eupolypods I-1 = Dryopteridaceae in part; Eupolypods I-2 = Davalliaceae, Dryopteridaceae (in part), Lomariopsidaceae, Oleandraceae, Polypodiaceae, and Tectariaceae (not sampled in *19*); Eupolypods II-1 = Aspleniaceae; and Eupolypods II-2 = Blechnaceae, Onocleaceae, Thelypteridaceae, and Woodsiaceae. *Abbreviations*: C (Carboniferous), CZ (Cenozoic), D (Devonian), J (Jurassic), Ng (Neogene), P (Permian), Pg (Paleogene), and Tr (Triassic).

more than 9000 species. These ferns have sporangia that develop from a single cell and have mature walls only one cell thick. Most leptosporangiate ferns also possess a distinctive annulus that serves to eject the (usually 64) spores. The osmundaceous ferns (Osmundaceae) are well supported as the closest relative of all other leptosporangiates (*4, 13, 15*). This position is consistent with the fossil record, as the oldest leptosporangiate fossils assignable to an extant lineage are members of this clade (*16, 17*). The filmy ferns, composing a single large family (Hymenophyllaceae) and the gleichenioid ferns,

with three smaller families (Dipteridaceae, Matoniaceae, and Gleicheniaceae), are both clearly monophyletic (*13, 15*). However, the precise relationships of filmy ferns and gleichenioids to one another and to the remaining leptosporangiate ferns remain unclear. The schizaeoid ferns are well supported as closest to the so-called "core leptosporangiates" (*4, 13, 15*), a large clade comprising three monophyletic groups: heterosporous ferns, tree ferns, and polypod ferns.

Although not always thought to form a natural group (*18*), the monophyly of polypod ferns receives

Table 1. Divergence time estimates (Ma) and their confidence/credibility intervals among ferns.

Timetree		Estimates			
Node	Time	Ref. (*4*)		Ref. (*19*)	
		Time	CI	Time	CI
1	364.4	364.4	364–356	–	–
2	359.6	359.6	–	–	–
3	354.0	354.0	–	–	–
4	323.1	323.1	330–310	–	–
5	305.6	305.6	318–267	–	–
6	286.2	286.2	297–272	–	–
7	272.9	272.9	283–259	–	–
8	266.3	266.3	281–250	–	–
9	263.3	263.3	–	–	–
10	220.0	220.0	232–206	–	–
11	211.6	211.6	237–188	–	–
12	210.8	210.8	–	–	–
13	182.9	182.9	195–169	–	–
14	175.8	–	–	175.8	200–163
15	173.3	173.3	186–156	–	–
16	151.4	–	–	151.4	174–137
17	144.5	–	–	144.5	167–130
18	104.7	–	–	104.7	124–91
19	94.5	–	–	94.5	112–79
20	93.6	–	–	93.6	112–81

Note: For each node, the confidence interval was calculated from ref. (*4*) or ref. (*19*), as the mean ± 1.96 times the standard deviation (when the standard deviation was equal to zero or not reported, no interval is given).

solid support in all recent analyses (*4, 13, 15, 19*). This clade, accounting for about 80% of all living fern species, is united by an unequivocal derived morphological character—sporangia each with a vertical annulus interrupted by the stalk. The smaller of the two clades arising from the first divergence within the polypod clade contains the lindsaeoid ferns and a few rather enigmatic fern genera (*Cystodium*, *Lonchitis*, and *Saccoloma*). The remaining polypods compose three well-supported clades: the small dennstaedtioid clade (Dennstaedtiaceae), the large pteroid clade (Pteridaceae), and the hyperdiverse eupolypod fern clade. The relationships among these three lineages are ambiguous. Within the eupolypods, two large clades of roughly equal size are resolved, which were recently dubbed "eupolypods I" and "eupolypods II" (*19*). This split is well supported by molecular data, but also by a frequently overlooked

morphological character, namely the vasculature of the petiole (*15*). Eupolypods I have three or more vascular bundles (with the exception of the grammitid ferns with one, and the genus *Hypodematium* with two), whereas eupolypods II have only two (with the exception of the well-nested blechnoid ferns with mostly three or more).

Integrating fossil time constraints together with molecular data from living taxa, two studies have estimated divergence times broadly across ferns (*4, 19*) (Fig. 2, Table 1). These divergence times are largely in accord with previous ideas about the times of origin and diversification of major fern clades (*16, 17, 20–23*). However, some clades (e.g., whisk ferns and ophioglossoids) with sparse fossil records are estimated to have originated much earlier than their meager fossil data would imply. The initial divergence among fern lineages occurred 364 Ma (Late Devonian). All four eusporangiate lineages, as well as the leptosporangiate ferns, were present by the end of the Carboniferous. Whisk ferns and ophioglossoids diverged from one another in the Late Carboniferous (306 Ma), and the earliest divergences among their living lineages occurred in the Late Cretaceous (88 Ma) and Middle Jurassic (162 Ma), respectively (*4*). As indicated by the fossil record, horsetails and marattioids had diverged from one another by the end of the Devonian; however, the divergences among living lineages within these groups appear to be more recent phenomena. Extant horsetails were estimated to have diversified in the Cenozoic (38 Ma; *4*, see also *14*). Extant marattioid lineages first diverged from one another in the Middle Triassic (237 Ma; *4*).

Within leptosporangiate ferns, the earliest divergences are estimated to have occurred in the Carboniferous and Permian. These divergences gave rise to the osmundaceous, filmy, gleichenioid, and schizaeoid ferns, as well as to the "core leptosporangiates" (Fig. 2). The initial divergence within the osmundaceous ferns is estimated to have occurred by the end of the Triassic (206 Ma; *4*) and the two major filmy fern lineages diverged from one another in the Jurassic (163 Ma; *4*, see also *24*). The earliest divergence within the gleichenioid ferns occurred in the Permian (263 Ma), but diversification within the extant gleichenioid families (Gleicheniaceae, Dipteridaceae, Matoniaceae) appears to be more recent (Cretaceous, see *4*). The initial divergence within schizaeoid ferns is estimated to have occurred in the Triassic, 212 Ma. A Late Triassic diversification gave rise to the three major lineages of "core leptosporangiates" (Fig. 2)—heterosporous ferns, tree ferns, and polypod ferns; the earliest divergences within each of these lineages occurred in the Jurassic.

All of the major polypod fern clades—lindsaeoids+, dennstaedtioids, pteroids, eupolypods I, and eupolypods II—had their origins in the Jurassic or Early Cretaceous (Table 1, Fig. 2). However, diversification in nearly all of these clades did not begin until the Late Cretaceous. This result alone suggests that at least 80% of extant fern diversity arose only in the last 100 million years.

Ferns attained remarkable levels of diversity from the Carboniferous to the Jurassic. But despite their ancient origins and early successes, it appears that several radiations in the Cretaceous and Cenozoic generated the bulk of modern fern diversity (Fig. 2; *4, 19*). This timing is suggestive of an ecological opportunistic response to the rise of angiosperms, as flowering plants came to dominate terrestrial ecosystems (*19*). Angiosperm-dominated communities likely promoted speciation in many lineages across the tree of life by creating new ecospaces into which these lineages could diversify (*25*). Polypod ferns may have been able to exploit new shady forest ecospaces specifically, through the evolutionary acquisition of a novel, physiologically more versatile, photoreceptor (*19, 26*). Better estimates of divergence times will allow us to more carefully evaluate potential links among profound biological phenomena and will help to elucidate those key events that have led to the many large, species-rich radiations in the long history of fern life on Earth.

Acknowledgment

References

1. P. Kenrick, P. R. Crane, *The Origin and Early Diversification of Land Plants: A Cladistic Study*. (Smithsonian Press, Washington, D.C., USA, 1997).
2. D. L. Nickrent, C. L. Parkinson, J. D. Palmer, R. J. Duff, *Mol. Biol. Evol.* **17**, 1885 (2000).
3. K. M. Pryer et al., *Nature* **409**, 618 (2001).
4. K. M. Pryer et al., *Am. J. Bot.* **91**, 1582 (2004).
5. Y.-L. Qiu et al., *Proc. Natl. Acad. Sci. U.S.A.* **103**, 15511 (2006).
6. K. S. Renzaglia, R. J. Duff, D. L. Nickrent, D. J. Garbary, *Phil. Trans. Roy. Soc. Lond. B* **355**, 769 (2000).
7. G. W. Rothwell, K. Nixon, *Int. J. Plant Sci.* **167**, 737 (2006).
8. N. Wikström, K. M. Pryer, *Mol. Phylogenet. Evol.* **36**, 484 (2005).
9. A. R. Smith et al., *Taxon* **55**, 705 (2006).
10. K. M. Pryer, H. Schneider, E. A. Zimmer, J. A. Banks, *Trends Plant Sci.* **7**, 550 (2002).
11. H. Schneider et al., in *Developmental Genetics and Plant Evolution*, Q. C. B. Cronk, R. M. Bateman, J. A. Harris, Eds. (Taylor & Francis, Philadelphia, 2002), pp. 330–364.
12. L. A. Raubeson, R. K. Jansen, *Science* **255**, 1697 (1992).
13. E. Schuettpelz, P. Korall, K. M. Pryer, *Taxon* **55**, 897 (2006).
14. D. L. Des Marais, A. R. Smith, D. M. Britton, K. M. Pryer, *Int. J. Plant Sci.* **164**, 737 (2003).
15. E. Schuettpelz, K. M. Pryer, *Taxon* **56**, 1037 (2007).
16. M. E. Collinson, in *Pteridology in Perspective*, J. M. Camus, M. Gibby, R. J. Johns, Eds. (Royal Botanic Gardens, Kew, UK, 1996), pp. 349–394.
17. W. D. Tidwell, S. R. Ash, *J. Plant Res.* **107**, 417 (1994).
18. A. R. Smith, *Amer. Fern J.* **85**, 104 (1995).
19. H. Schneider et al., *Nature* **428**, 553 (2004).
20. G. W. Rothwell, *Am. J. Bot.* **74**, 458 (1987).
21. G. W. Rothwell, *Rev. Palaeobot. Palynol.* **90**, 209 (1996).
22. J. E. Skog, *Brittonia* **53**, 236 (2001).
23. P. S. Soltis et al., *Proc. Natl. Acad. Sci. U.S.A.* **99**, 4430 (2002).
24. E. Schuettpelz, K. M. Pryer, *Syst. Biol.* **55**, 485 (2006).
25. C. S. Moreau et al., *Science* **312**, 101 (2006).
26. H. Kawai et al., *Nature* **421**, 287 (2003).