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Seed ferns and the origin of angiosperms

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DOYLE, J.A. (Section of Evolution and Ecology, University of California, Davis, California 95616, USA). Seed ferns and the origin of angiosperms. J. Torrey Bot. Soc. 133: 169–209. 2006.—If molecular analyses are correct in indicating that Gnetales are related to conifers and no other living gymnosperm group is directly related to angiosperms, studies on the origin of angiosperms must focus on fossil taxa, including ''seed ferns.'' Some authors have homologized the angiosperm carpel with the cupule of seed ferns, but because angiosperm ovules have two integuments rather than one, cupules are more likely to be homologous with the outer integument. Cupules of the earliest seed ferns may be derived from fertile appendages of ''progymnosperms,'' but those of later taxa appear to be modified leaves or leaflets, with ovules borne on the abaxial surface in some (peltasperms, corystosperms), the adaxial surface in others (glossopterids, *Caytonia*). Positional relationships and developmental genetic data suggest that the bitegmic ovule is comparable to a cupule with adaxial ovules. Analysis of a critically revised morphological data set for seed plants indicates that trees in which Gnetales are nested in conifers, as in molecular analyses, are almost as parsimonious as those in which Gnetales are linked with angiosperms, suggesting that the molecular arrangement should be accepted. When living taxa are constrained into the molecular topology, angiosperms are linked with glossopterids, *Pentoxylon*, Bennettitales, and *Caytonia*, supporting the homology of the cupule and the bitegmic ovule. Origin of the carpel poses more problems; it could correspond to the leaf portion of the glossopterid leaf-cupule complex, but its homologies in *Caytonia* are more obscure. New data on currently unknown characters of glossopterids, ''Mesozoic seed ferns,'' and Bennettitales are needed to test these hypotheses.

Key words: Seed plants, phylogeny, angiosperms, glossopterids, *Caytonia*

Many earlier discussions of the origin of angiosperms concluded that angiosperms were ''derived from'' seed ferns (e.g., Long 1966, Cronquist 1968, 1988, Takhtajan 1969). From a cladistic viewpoint, such statements are not very informative, since all phylogenetic analyses of living and fossil seed plants have indicated that ''seed ferns'' are a paraphyletic grade made up of lines that are basal to more derived groups (Crane 1985, Doyle and Donoghue 1986, Doyle et al. 1994, Nixon et al. 1994, Rothwell and Serbet 1994, Doyle 1996). By definition, the ancestor of a clade is not a paraphyletic group but a single species, which might be impossible to recognize as ancestral in the fossil record. However, taxa traditionally called seed ferns could still be the closest relatives of angiosperms, and these could say almost as much as a direct ancestor, by revealing the order of evolution of the various new features of the extant clade (the crown group) and more plesiomorphic homologs of its characteristic structures. This is especially true if we can recognize a series of successive outgroups, which may allow us to distinguish character states that existed on the stem lineage leading to the crown group from autapomorphies that arose in extinct side lines.

Whether or not seed ferns include the closest relatives of angiosperms, they may be important for understanding the origin of angiosperms and their distinctive features. Many authors have argued that the ovulate structures of other groups were too derived to be prototypes for the carpel, usually interpreted as a folded leaf bearing ovules on its adaxial side. For example, Bennettitales have been associated with angiosperms because they had flower-like structures (Arber and Parkin 1907) but rejected as ancestors because their ovules were borne directly on an ovuliferous receptacle, intermixed with interseminal scales, rather than on a leaflike structure. Similarly, other groups have been excluded as angiosperm ancestors because they are too advanced in their wood anatomy or other fea-

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tures (Bailey 1944, 1949, Cronquist 1968, 1988, Takhtajan 1969). Taxa with more derived sporophylls could still be the closest relatives of angiosperms, and they might have much to say about the origin of other features. However, it would be necessary to look lower in the phylogenetic tree for plants with leaflike sporophylls that might be transformed into a carpel, and these might be called seed ferns. This point was recognized by Arber and Parkin (1907), who argued that angiosperms were related to Bennettitales and Gnetales but all three groups came from a common ancestor with pinnate megasporophylls, which they hypothesized was derived from some group of seed ferns (cf. also Takhtajan 1969). The question becomes which group or groups of plants traditionally called seed ferns are related to angiosperms and how.

Understanding the homologies of the carpel also requires consideration of the angiosperm ovule, which usually differs from the ovules of other seed plants in having two integuments (bitegmic) and being bent back on itself (anatropous). The inner integument is presumably homologous with the single integument of other seed plants, but what is the outer integument? This problem is illustrated by the ''Mesozoic seed fern'' *Caytonia*, first described from the Yorkshire Jurassic by Thomas (1925). The ovulate structures of *Caytonia* consisted of an axis bearing two rows of fleshy cupules, each of which contained several ovules. Thomas (1925) compared these cupules with angiosperm carpels. However, this idea soon fell out of favor. First, Harris (1940) found that pollen got inside the cupule, to the micropyles of the ovules—the plant was functionally gymnospermous. This would not rule out the hypothesis that *Caytonia* was related to angiosperms but more primitive. However, other aspects of the morphology of *Caytonia* are inconsistent with a cupule-carpel homology. First, angiosperm carpels are thought to be modified leaves borne on a stem, whereas *Caytonia* cupules were borne in two rows on a dorsiventral axis, like leaflets on the rachis of a compound leaf. Second, as emphasized by Bailey and Swamy (1951), the cupules of *Caytonia* were enrolled circinately, from tip to base, whereas supposedly primitive carpels are folded lengthwise (conduplicate, or plicate). Finally, the ovules of *Caytonia* had only one integument, not two.

Similar problems affect Long's (1966) derivation of the carpel from the lobate, dichotomously organized cupule of Carboniferous seed ferns. On recognizing the problem of the bitegmic ovule, Long was forced to postulate that the second integument arose de novo as an outgrowth of the first. Meeuse and Bouman (1974) tried to circumvent the problem by homologizing the inner integument with the wall of the lagenostome (distal part of the nucellus) in early seed ferns. However, as recognized by Meeuse and Bouman, the inner integument of angiosperms develops from a ring of meristematic tissue (cf. Robinson-Beers et al. 1992, Umeda et al. 1994), whereas the lagenostome wall represents the epidermis of the nucellar apex, which separated from the central tissue of the apex to form a pollen chamber (Sporne 1965, Stewart and Rothwell 1993).

Such arguments led some to conclude that *Caytonia* was not related to angiosperms (e.g., Bailey 1949, Harris 1951, Cronquist 1968). However, there is another way to formulate homologies that might salvage the *Caytonia*-angiosperm relationship and solve the problem of the angiosperm ovule at the same time, first proposed by Gaussen (1946) and later supported by Stebbins (1974) and Doyle (1978; Fig. 1). Under this hypothesis the cupules of *Caytonia* correspond not to carpels, but rather to bitegmic ovules. The only change needed in the cupule would be reduction of the ovule number to one. The cupule wall would thus become the outer integument, and because of the circinate character of the cupule the bitegmic ovule would already be anatropous. However, this leaves a problem in explaining the carpel. In terms of positional relationships, the carpel should correspond to the *Caytonia* rachis, but this was narrow and not very leaflike. In Doyle (1978) I argued that the rachis was probably larger relative to the cupules early in ontogeny, so that it could be transformed into a carpel by modification of development at an early stage.

Stebbins (1974) preferred to compare angiosperms with another group, the Permian glossopterids of Gondwana, and this idea was adopted by Retallack and Dilcher (1981; Fig. 2). Glossopterids also had ovule-bearing structures that have been called cupules, but these were more leaflike—they were described as megasporophylls by Gould and Delevoryas (1977) and Taylor and Taylor (1992)—and did not enclose the ovules so completely. Either one or several of these cupules were borne on the adaxial side of a normal leaf. The resulting structure has been variously called a fertiliger (Schopf 1976), a bract-sporophyll complex (Doyle 1996), or a

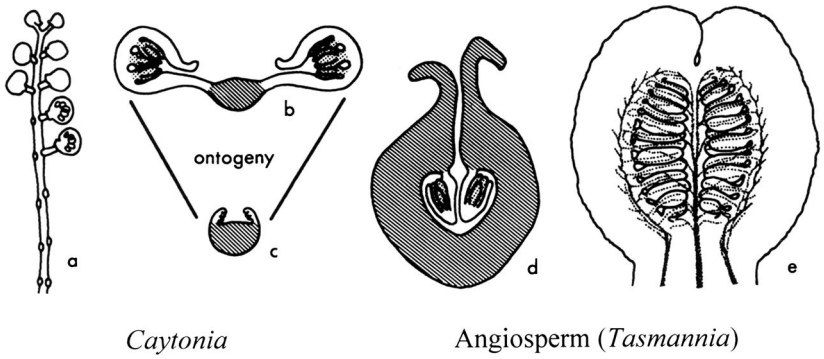

FIG. 1. Proposed homologies between ovulate structures of *Caytonia* (left) and angiosperms (right), from Doyle (1978), following Gaussen (1946) and Stebbins (1974).

bract-cupule complex (Doyle 1998a); in this article I will call it a leaf-cupule complex, because the subtending leaf was essentially unmodified. The cupule has been interpreted in many ways (Retallack and Dilcher 1981, Pigg and Trivett 1994): as a sporophyll fused to a leaf, a sporophyll on an axillary shoot fused to a leaf, or an adaxial fertile segment of a leaf (analogous to the fertile segment of Ophioglossales: Kato 1990). Whatever the cupule was, reduction to one ovule per cupule would yield an organ like a bitegmic ovule. Furthermore, the subtending leaf could be folded around the cupule to form the carpel wall—an advantage over the *Caytonia* hypothesis. Actually, the two hypotheses may not be mutually exclusive, since it is possible that *Caytonia* and glossopterids are related. This would be consistent with their simple reticulate leaf venation, with a midrib and one order of laminar venation. The main difference is that the *Glossopteris* leaf was simple but the *Caytonia* leaf was palmately compound, with four leaflets each resembling a *Glossopteris* leaf.

There are good reasons to believe that the many structures called cupules were not all homologous. Cupules of the first Late Devonian-Carboniferous seed ferns (*Archaeosperma*, *Elkinsia*, other hydraspermans, *Lyginopteris*) were

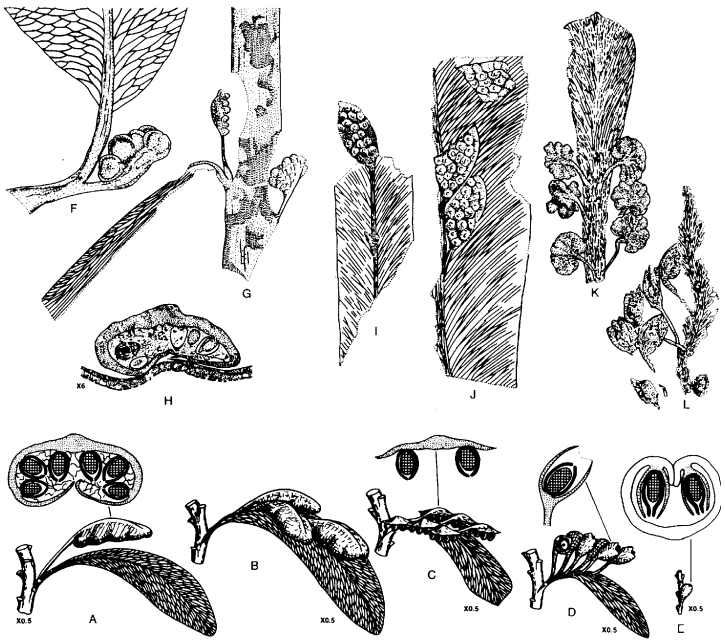

FIG. 2. Drawings of ovulate structures of glossopterids (above) and proposed steps in transformation of the leaf-cupule complex into an angiosperm carpel (below), from Retallack and Dilcher (1981).

dichotomously organized and borne apically on special fronds or segments of fronds (Kidston 1924, Long 1961, 1979, Galtier 1988, Retallack and Dilcher 1988, Serbet and Rothwell 1992). Kenrick and Crane (1997) argued that these cupules were homologous with the dichotomous fertile appendages of ''progymnosperms,'' while the ovules themselves were derived from groups of sporangia (or sporangium-bearing telomes), with the integument derived from the outer sporangia (telomes) by sterilization and fusion. In contrast, the cupules of *Caytonia* and glossopterids were dorsiventral and therefore more like modified leaves or leaflets (cf. Reymanówna 1974). This view is consistent with the stratigraphic distribution of the two types of cupules: dichotomously organized cupules appeared near the origin of typical compound fronds, whereas cupules of the dorsiventral type appeared much later.

The first cladistic analysis to address these questions was by Crane (1985). The best-known result of this study is the inference that angiosperms were related to Bennettitales, the Cretaceous genus *Pentoxylon*, and Gnetales, forming the ''anthophyte'' clade. However, the closest outgroups of anthophytes were Mesozoic seed ferns: corystosperms, *Caytonia*, and glossopterids. As Crane argued, these results were consistent with the cupule-ovule homology. He proposed a scheme starting with a megasporophyll bearing multiovulate cupules, as in *Caytonia*. Then the number of ovules per cupule was reduced to one, as in corystosperms. The carpel was derived from the whole sporophyll by expansion and folding of the rachis. Crane postulated that *Pentoxylon* and Bennettitales underwent further reduction to one cupule per sporophyll and a shift of the cupule to an orthotropous orientation. This relied in part on the view of Harris (1954) that some Bennettitales had a cupule, an interpretation recently rejected by Rothwell and Stockey (2002) and Stockey and Rothwell (2003) based on observations on *Cycadeoidea* and *Williamsonia*. Doyle and Donoghue (1986) obtained results similar to those of Crane, but with angiosperms at the base of the anthophytes and *Caytonia* as their sister group. They interpreted the flowers of Gnetales as still more reduced, with reduction to one ovule per flower, loss of the cupule, and formation of a new outer integument from two perianth parts (cf. Doyle 1994).

These results also supported the view that not all cupules were homologous. The basal Paleozoic seed ferns with dichotomous cupules were separated from Mesozoic groups by lines that lacked cupules and bore seeds directly on more or less leaflike sporophylls, such as medullosans, *Callistophyton*, and cycads. This implies that the original cupule was lost (or perhaps less plausibly that the original integument fused with the nucellus and the cupule was transformed into a new integument: Walton 1953, Meyen 1984), and that the cupules of more advanced seed ferns were modified ovule-bearing leaves or leaflets.

Other morphological analyses kept the anthophytes together but separated them from Mesozoic seed ferns (Nixon et al. 1994, Rothwell and Serbet 1994). Gnetales were monophyletic in Rothwell and Serbet (1994), but paraphyletic in Nixon et al. (1994), with angiosperms nested within them. In both analyses anthophytes were linked with conifers, while *Caytonia* and glossopterids formed a clade situated lower in the tree.

These results have been called into question by molecular phylogenetic analyses of living seed plants. Obviously such studies say nothing directly about relationships of angiosperms to fossil taxa, but they do address the view that angiosperms are related to Gnetales. Only a few molecular analyses have linked angiosperms and Gnetales, and this with low statistical support (Hamby and Zimmer 1992, Stefanovic et al. 1998, Rydin et al. 2002). Some analyses have placed Gnetales at the base of seed plants (Hamby and Zimmer 1992, Albert et al. 1994, Sanderson et al. 2000, Rydin et al. 2002), but tests using likelihood and other methods suggest that this arrangement is a result of long-branch attraction, particularly affecting third codon positions (Sanderson et al. 2000, Magallón and Sanderson 2002, Rydin et al. 2002, Soltis et al. 2002, Burleigh and Mathews 2004). Most analyses, especially those based on combining several genes, have associated Gnetales with conifers, either as their sister group or nested within them, as the sister group of Pinaceae (Goremykin et al. 1996, Chaw et al. 1997, 2000, Hansen et al. 1999, Qiu et al. 1999, Samigullin et al. 1999, Shindo et al. 1999, Winter et al. 1999, Bowe et al. 2000, Frohlich and Parker 2000, Sanderson et al. 2000, Rydin et al. 2002, Soltis et al. 2002, Burleigh and Mathews 2004, Nickerson and Drouin 2004, Kim et al. 2004). Trees of this sort offer a more plausible alternative to the anthophyte hypothesis, because many earlier authors pointed out similarities between Gneta-

les and conifers, such as linear leaves, elimination of scalariform pitting even in the primary xylem, and compound strobili constructed on a cordaite-like plan (Bailey 1944, 1949, Eames 1952, Bierhorst 1971, Doyle 1978, Carlquist 1996a).

Most such ''gnetifer'' and ''gnepine'' trees indicate that no other living gymnosperm group is any more closely related to the angiosperms: angiosperms and living gymnosperms are sister groups. This does not mean that angiosperms and gymnosperms were derived independently from non-seed plants, or that the molecular results conflict with the fossil record and should therefore be rejected (Axsmith et al. 1998). Any number of Paleozoic seed fern lines might branch off below the common ancestor of living angiosperms and gymnosperms, and other Paleozoic and Mesozoic taxa might be attached to the stem lineage leading to angiosperms. Consistent with this view, an analysis of the morphological data set of Doyle (1996) with living taxa constrained into the molecular arrangement indicated that both angiosperms and living gymnosperms are nested among Paleozoic seed ferns (Doyle 2001). However, the molecular results do mean that the search for relatives of angiosperms and steps in their origin must concentrate on fossil seed plants.

To address this question requires use of a morphological data set to determine how fossils fit into the tree of living taxa. This is a daunting task now that previous morphological analyses appear to have been so wrong about the relationship of angiosperms and Gnetales. However, the fact that morphology was wrong about Gnetales does not mean it is misleading everywhere—molecular phylogenetic analyses have confirmed many groups that were first recognized based on morphology—and in any case it is the only tool we have for the job. A general reassessment of previously used morphological characters is desirable, but a critical reevaluation of those characters that supported the anthophyte hypothesis is especially necessary, as emphasized by Donoghue and Doyle (2000). This article presents such an analysis, which incorporates a critique of supposed anthophyte synapomorphies, previously overlooked similarities between Gnetales and conifers, and new developments on the morphology of fossil seed plants and attempts to synthesize morphological data from living and fossil taxa with results of molecular analyses.

Materials and Methods. The starting point for this study was the data set of Doyle (1996), but many characters have been redefined, added, or eliminated after critical evaluation. All changes in characters and scoring of taxa are listed in Appendix 1. Some that are most relevant to angiosperm relationships or pose problems that require special argumentation are discussed here.

NEW DATA ON OVULE/CUPULE HOMOLOGIES. One kind of new data concerns homologies of the ''cupules'' of various taxa. As already noted, morphological evidence and previous phylogenetic analyses suggest that cupules of Permian and Mesozoic taxa were derived from leaves or leaflets with ovules on one surface. An important question is which surface, and how this compares with the condition in angiosperms. The same distinction can be extended to taxa with ovules borne on the surface of less modified leaves, thus avoiding semantic questions of whether a structure is a cupule, a sporophyll, or a leaflet of a sporophyll. This character was used by Doyle (1996), but it can now be scored in more taxa.

In some taxa the ovules were borne on the abaxial side of the cupule. This was previously known for Permian and Triassic peltasperms (*Peltaspermum*, *Autunia*, etc.), which had spoon-shaped or peltate cupules. Some peltasperm cupules are known attached to an axis in a spiral (helix) and can therefore be interpreted as simple sporophylls (Thomas 1933, Meyen 1987, Kerp 1988, Nixon et al. 1994, Doyle 1996), not leaflets of a pinnate sporophyll, as believed by Townrow (1960), Doyle and Donoghue (1986), and Retallack and Dilcher (1988). The fact that seeds in the peltate forms were attached to the underside of the peltate cap confirms their abaxial position, because leaves of living plants become peltate by formation of a cross-zone between the adaxially directed margins of the leaf primordium (Hagemann 1970).

An exciting recent discovery (Axsmith et al. 2000, Klavins et al. 2002) was that cupules of corystosperms from the Triassic of Antarctica had a similar orientation. Some authors had interpreted the branched structures bearing these cupules as compound sporophylls (Harris 1951, p. 38; Doyle and Donoghue 1986), but compression specimens with cupules arranged in terminal pseudowhorls (Axsmith et al. 2000) indicated that they were branches and the cupules were simple sporophylls, as in peltasperms, as argued by Thomas (1933) and Nixon et al.

(1994) and accepted as one of two possibilities by Doyle (1996). The cupules were curved downward relative to the axis, implying that the ovules were abaxial. In a study of petrified material, Klavins et al. (2002) confirmed this interpretation based on orientation of the xylem and phloem in the vascular bundles of the cupule: the ovules were on the phloem side of the bundles, which was presumably abaxial. Therefore I have rescored corystosperms as having paddle-like megasporophylls (1), rather than uncertain (0/1) in Doyle (1996), and abaxial ovules (1), rather than (1/2).

Cupules in other groups had adaxial ovules. It has been assumed that this was the case in *Caytonia*, with the cupules derived from adaxially enrolled leaflets (Harris 1940, 1951, Reymanówna 1974). Probably the best argument is the orientation of the cupules relative to the rachis, which is strongly dorsiventral: flatter on one side (presumably adaxial) and more convex on the other (presumably abaxial). The cupules are attached on either side of the flatter surface of the rachis and enrolled toward the middle (i.e., circinately), enclosing the ovules (Fig. 1). This orientation is more certain in Antarctic Triassic cupules described by Taylor et al. (1994) as *Petriellaea*. *Petriellaea* has not been associated with other organs, but vascular bundles are preserved in the cupule wall, and the ovules are attached to the xylem side and are therefore adaxial.

Glossopterids are another group with adaxial ovules, but they show complications due to the problematic double nature of the fertile structures. Early observations on compression fossils gave conflicting indications on whether the ovule-bearing surface of the cupule faced toward or away from the subtending leaf (e.g., Holmes 1974, Pant and Singh 1974, Fig. 2F, G). However, Schopf (1976) and Retallack and Dilcher (1981, 1988) concluded that the ovule-bearing surface faced the leaf, and this interpretation has been confirmed by analyses of numerous impression fossils of leaf-cupule complexes split along various planes (McLoughlin 1990, Adendorff 2005).

A major breakthrough in understanding glossopterid structures was the description by Taylor and Taylor (1992) of a silicified cupule (termed a megasporophyll) with the vascular bundles preserved, in which the ovules were borne on the xylem side and therefore adaxial. This cupule was not preserved in attachment to a leaf. Taylor and Taylor (1992) and Taylor (1996) claimed that their data contradicted the view that the ovules were borne on the side of the cupule facing the subtending leaf, but this does not necessarily follow. Schopf (1976) had confused the issue by describing the ovule-bearing surface as abaxial, apparently defined in terms of the relation of the whole leaf-cupule complex to the main stem, but he may have been correct about the orientation of the structure and incorrect about the morphological relations of its component parts. It is entirely possible that the adaxial side of the cupule faced the adaxial side of the leaf. The interpretation of the leaf-cupule complex that does the least violence to conventional morphological assumptions, one of three hypotheses discussed by Retallack and Dilcher (1981), is that the cupule was a sporophyll borne on an axillary branch that became adnate to the subtending leaf (Fig. 3). If the sporophyll was attached to the side of the axillary branch opposite the subtending leaf, like the adaxial prophyll of monocots and some magnoliids, ovules borne on its adaxial side would face the adaxial side of the subtending leaf. Taylor and Taylor (1992) and Taylor (1996) also questioned this orientation of the cupule because they thought it would mean that wind-borne pollen could not to get to the ovules. However, many living conifers

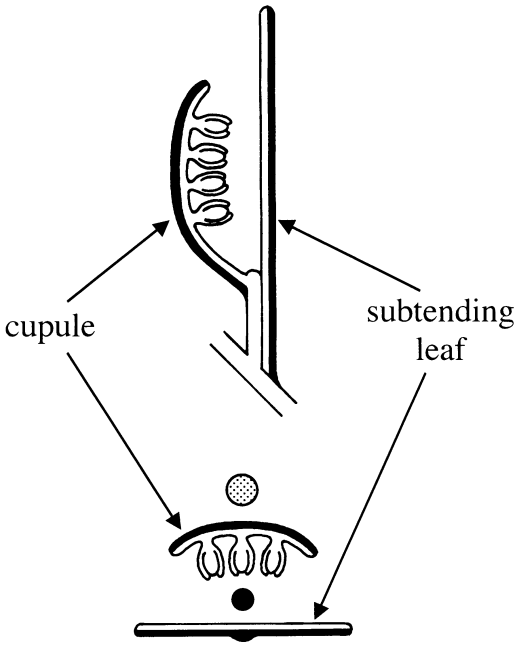

FIG. 3. Interpretation of positional relationships in the leaf-cupule complex of glossopterids, with abaxial surfaces indicated in black.

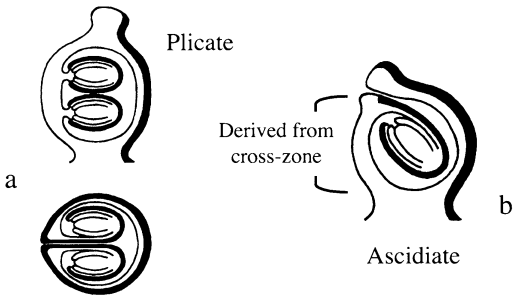

FIG. 4. Identification of the abaxial and adaxial surfaces of the carpel and the outer integument in (a) a plicate carpel and (b) an ascidiate carpel, with abaxial surfaces indicated in black.

manage to be pollinated during brief periods of separation of cone scales that are tightly appressed at other stages.

If the bitegmic ovule of angiosperms was derived from a cupule, this was presumably the type with adaxial ovules (Doyle and Donoghue 1986, Doyle 1996, Frohlich 2003), unless there was a reversal of cupule polarity in the origin of angiosperms (Frohlich 2003). Because there is much confusion about this point, it is important to realize this comparison is based on the presumed adaxial position of the nucellus and inner integument (together considered equivalent to the original seed plant ovule) relative to the outer integument, not on the adaxial position of the bitegmic ovule on the carpel. Klavins et al. (2002) took the contrast between the abaxial position of ovules in the corystosperm cupule and the adaxial position of ovules in the angiosperm carpel as evidence against a relationship between the two groups, but if the angiosperm bitegmic ovule corresponds not to a gymnosperm ovule but rather to a cupule containing an ovule, this is not the relevant comparison.

This hypothesis implies that the outer surface of the angiosperm outer integument is abaxial, the inner surface adaxial. In plicate carpels of the type shown in Fig. 1 (Doyle 1978), this polarity can be seen by tracing from the abaxial side of the carpel to the outside of the outer integument (Fig. 4a). The positional relationship is less obvious in ascidiate carpels, the ancestral type based on molecular phylogenies (Doyle and Endress 2000, Endress and Igersheim 2000), where the ovule is attached to a cross-zone on the adaxial side of a U-shaped or annular carpel primordium (Fig. 4b). However, the same identification of abaxial and adaxial surfaces is confirmed by the position of xylem and phloem in vascular bundles in the outer integument of the few angiosperms in which this feature has been described (Svoma 1997, Frohlich 2003).

Additional evidence is available from molecular developmental work on *Arabidopsis*. Based on studies of mutants and patterns of gene expression, genes of the YABBY family have been identified as specifying the abaxial side of leaves and other lateral organs (Bowman 2000). One of these is expressed in ovules: INO, for inner no outer, so-called because mutants have an inner integument but no outer integument. This gene is expressed in the outer integument (Villanueva et al. 1999, Bowman 2000), specifically in its outer epidermis (Balasubramanian and Schneitz 2000, 2002, Meister et al. 2002, Skinner et al. 2004), but not in its inner epidermis, nor in the inner integument. This implies that the outside of the outer integument is abaxial, as expected if it was derived from a leaf or leaflet with a unitegmic ovule on its adaxial surface. Meister et al. (2002), Yamada et al. (2003), and Skinner et al. (2004) recognized that these data suggest the outer integument is a leaflike organ, while Frohlich (2003) saw them as evidence that the bitegmic ovule was derived from a cupule with adaxial rather than abaxial ovules. These and other genetic data also suggest that the inner integument is fundamentally different from the outer integument, and not leaflike (Gross-Hardt et al. 2002, Yamada et al. 2003, Sieber et al. 2004, Skinner et al. 2004). As noted by Gross-Hardt et al. (2002), this is consistent with hypotheses that the inner integument was derived much earlier from the outer telomes of a fertile dichotomous branch or the outer sporangia of a synangium (Kenrick and Crane 1997).

There are reasons for caution in taking these data as confirmation of the positional and anatomical evidence on polarity of the bitegmic ovule. An alternative explanation of the gene expression data, proposed by Sieber et al. (2004), is that the two integuments of angiosperms were derived from a single integument by splitting. This hypothesis was suggested by the finding that PHB, a gene involved in specifying the adaxial side of leaves, is not expressed in the inner surface of the outer integument, but it is expressed on the inner surface of the inner integument. However, the data of Sieber et al. (2004) still suggest that the whole bitegmic ovule is a dorsiventral structure, because PHB is expressed on the presumed adaxial side of the ovule primordium well before the appearance of either integument.

Another complication is a report by Yamada et al. (2004) that YABBY genes in the basal angiosperm *Amborella* are expressed not on the abaxial side of leaves and carpels, but rather on the adaxial side. The relevance of this finding for ovule homologies is uncertain, because Yamada et al. (2004) did not find a homolog of INO, the YABBY gene involved in ovule development. Yamada et al. (2003) showed that INO is expressed in the outer epidermis of the outer integument in Nymphaeales, the next branch above *Amborella*. They also reported weak INO expression in the inner integument and the tip of the nucellus, but it is not clear that the signal was above background level (C. S. Gasser, pers. comm.). Yamada et al. (2004) surmised that YABBY genes are still involved in establishing dorsiventrality in *Amborella*, but that a reversal in their expression and function occurred within angiosperms. To judge whether the *Amborella* pattern is primitive or autapomorphic may require evidence on YABBY expression in other seed plants, which is not yet available. A potentially more fundamental marker of dorsiventral polarity is KANADI (Eshed et al. 2001, 2004), but involvement of this or related genes in ovule development has not been established.

Microsporangial position in glossopterids poses similar problems. Pollen-producing structures of glossopterids consisted of a leaf with a branched microsporangium-bearing unit attached to its adaxial side, recalling the female leaf-cupule complex (Surange and Maheshwari 1970, Schopf 1976, Gould and Delevoryas 1977, Retallack and Dilcher 1981). In Doyle (1996) I scored the microsporangia as either terminal or adaxial (0/2), consistent with definition of their position relative to either the branched unit or the subtending leaf. However, if the whole compound structure corresponds to a leaf-cupule complex, microsporangial position is better defined in terms of the branched unit. Pigg and Nishida (2005) have shown that microsporangia were at least sometimes borne on scale-like appendages. However, until the general morphological situation in the group is better understood, I have scored microsporangial position in glossopterids as unknown (?).

In Doyle (1996) I scored microsporangial position in *Pentoxylon* as terminal, but Sharma (2001) and Srivastava and Banerji (2001) described the microsporangia as borne in two lateral rows on the ultimate subdivisions of a branched structure. Because there is no laminar structure to serve as a reference for defining sporangial position, I have rescored *Pentoxylon* as unknown, like glossopterids.

NEW DATA ON GNETALES, ANGIOSPERMS, AND CONIFERS. Other changes in the data matrix are based on doubts concerning the analysis of characters that supported the anthophyte hypothesis, some mentioned briefly by Donoghue and Doyle (2000).

One such character is presence of a thick megaspore membrane in the seed, the basic state in seed plants, versus a reduced membrane in anthophytes and *Caytonia*, a difference recognized by Harris (1954) and emphasized by Crane (1985). However, although there is no megaspore membrane at all in angiosperms, there is a thin membrane in Gnetales, ranging in thickness from 1.0 to 2.3 μm (Martens 1971). Instead of lumping thin megaspore membrane with none, I have redefined this as a presence-absence character. Harris (1954) concluded that there was no megaspore membrane in seeds of Bennettitales and *Caytonia*. One could ask whether a megaspore membrane might have been present but was thin, as in Gnetales, and was therefore not preserved. An argument against this view is the fact that a megaspore membrane has been reported in at least one presumably gnetalian fossil with ephedroid pollen, the Early Cretaceous genus *Eoantha* (Krassilov 1986), and the more problematic but possibly gnetalian plants that produced *Eucommiidites* pollen (Pedersen et al. 1989a, Crane 1996). In Bennettitales, silicified specimens of *Williamsonia* (Stockey and Rothwell 2003) and *Cycadeoidea* (Wieland 1916) have been described as having a thin megaspore membrane. However, Harris (1954) remarked that there is no reason to assume the membrane in *Cycadeoidea* was cutinized. These cases illustrate the difficulty of comparing features of seeds preserved in different ways.

Another putative anthophyte synapomorphy is presence of a tunica in the apical meristem (an outer layer of cells that undergo only anticlinal divisions), found in angiosperms and Gnetales, specifically *Ephedra* and *Gnetum*. However, in *Ephedra* and *Gnetum* the tunica is only one cell thick (Gifford 1943, Johnson 1950), whereas in angiosperms it consists of two layers. This two-layered structure has been confirmed in several angiosperm taxa in the present data set (Nymphaeaceae, *Illicium*, Schisandraceae, Chloranthaceae, Winteraceae: Gifford 1950, 1954,

Ramji 1961, Wardlaw 1965). There are angiosperms with one tunica layer, but these belong to groups that are derived in current phylogenies, such as Cactaceae and asterids (Gifford 1954). Because the difference in number of layers raises doubts concerning homology of the tunica in Gnetales and angiosperms, I have redefined this as an unordered three-state character (tunica absent, single-layered, two-layered), which is equally consistent with the homology or non-homology of the two tunica types. *Welwitschia* is described as lacking a tunica (Johnson 1951, Martens 1971), but I have scored it as unknown, rather than (0) in Doyle and Donoghue (1986) and Doyle (1996), because its apex aborts at an early stage, calling into question comparisons with the meristems of mature shoots in other taxa. Among fossils, lack of a tunica was reported in cordaites by Rothwell and Warner (1984). The only other seed plant group known to have a tunica, Araucariaceae, is reported to have both one- and two-layered types (Griffith 1952, Jackman 1960) and is therefore scored as (1/2).

I have also added or rescored some characters of Gnetales that are potential synapomorphies with conifers. One, emphasized by Carlquist (1996a, 1996b), is the presence of a torus in the side-wall pits in vessels of *Ephedra* and *Gnetum*, a feature otherwise restricted to the tracheids of conifers and *Ginkgo*. Carlquist (1994) did not find a torus in *Gnetum gnemon*. However, the molecular analysis of Won and Renner (2003) implies that this absence is secondary, since a torus does occur in Won and Renner's ''Africa,'' ''South America,'' and ''SE Asia II'' clades (Carlquist and Robinson 1995, Carlquist 1996c, d), which are basal to the ''SE Asia I'' clade that contains *G. gnemon*. Hence I have scored *Gnetum* as having a torus. Bauch et al. (1972) recognized six types of pit membrane corresponding to differing degrees of development of a torus. However, types 1–4 overlap in their systematic distribution and seem best treated as one state, presence of a torus. Type 2 is found only in some Pinaceae that also have type 1 pits, while type 4 occurs only in some Cupressaceae and Podocarpaceae that also have type 3, and both types 1 and 3 occur in Cupressaceae-Taxodiaceae. In addition, *Agathis*, assigned to type 3, shows a conspicuous torus under SEM (Meylan and Butterfield 1978). Bauch et al. (1972) reported type 5 only in *Gnetum gnemon*, where Carlquist (1994) found no torus, and *G. scandens*, an Asian species of uncertain identity (S. Renner, pers. comm.), and a torus is clearly lacking in type 6 (*Welwitschia*, cycads), so I treat both types as absence of a torus. In fossil taxa, EM studies by Schmid (1967) demonstrated that a torus is absent in *Cordaites*. I have assumed a torus is absent in Bennettitales and other groups with scalariform pits (Carlquist 1996b). Pits of the Paleozoic conifer *Emporia* figured by Mapes and Rothwell (1984, pl. 11:2) show a ring that may be a torus, but because this is not mentioned by the authors and hard to interpret I have scored *Emporia* as unknown (?). In corystosperms, pits of *Kykloxylon* show a black central dot, but Meyer-Berthaud et al. (1993) considered this too small to be a functional torus and more likely an optical artifact caused by the conical pit aperture.

Another such character is a tiered proembryo, a conspicuous feature of conifers. After a free-nuclear phase of varying length, formation of cell walls and further divisions result in four tiers of cells in most Pinaceae (embryo, suspensor, rosette, upper), three tiers in most other conifers (embryo, suspensor, upper or open) (Doyle 1963, Sporne 1965, Singh 1978). Upper (proximal) cells derived from the embryo tier elongate to produce a secondary suspensor. The tiered condition contrasts with the more massive construction of the embryos of cycads and *Ginkgo*, in which discrete tiers are not visible. Early embryos of Araucariaceae are more massive than those of other conifers and have therefore been considered more primitive (Sporne 1965), but they are less massive and show more hints of tiers than embryos of cycads and *Ginkgo* (Singh 1978). Since the differences from other conifers may be due to the larger number of free nuclei that contribute to the embryo, I have scored Araucariaceae as tiered, as in Doyle (1996).

Previous analyses scored Gnetales as lacking tiers (Rothwell and Serbet 1994, Doyle 1996), and Donoghue and Doyle (2000) cited the tiered proembryo of conifers as evidence against nesting Gnetales within conifers. Embryogeny of Gnetales does differ from that of conifers in at least two respects. First, as discussed further below, in Gnetales each embryo is derived from a single cell without free-nuclear divisions. In *Ephedra* the zygote undergoes free-nuclear divisions, but each resulting diploid cell develops into a separate embryo by cellular divisions (Friedman 1992, 1994). Second, the primary suspensor cell remains as a single cell rather than giving rise to suspensor and open tiers (not

counting ramification of the suspensor in *Gnetum*: Martens 1971). However, after a few cell divisions the rest of embryo is organized into two more or less regular tiers (which were in fact designated as such in *Welwitschia*, as ''étages,'' by Martens 1971), with cells of the upper tier elongating into a secondary suspensor (Martens 1971, figs. 38D–G, 82, 120; Singh 1978, figs. 135H–N, 136). Since the differences between this and the conifer situation may be a consequence of elimination of a free-nuclear stage (treated as another character), I have rescored Gnetales as tiered after all. This agrees with the view of Martens (1971, p. 265) that the proembryo phase of Gnetales is generally close to that of conifers.

Angiosperms differ from other seed plants in lacking not only tiers but also a secondary suspensor, which Singh (1978) considered a universal feature of gymnosperms (although this seems obscure in cycads and *Ginkgo*). Whereas upper cells derived from the embryo tier or the embryonal mass contribute to the secondary suspensor in other seed plants, the primary suspensor cell of angiosperms usually contributes to the radicle of the embryo as well as the suspensor. An exception, the caryophyllad type, is restricted to some monocots and eudicots such as Caryophyllaceae and Saxifragales (Maheshwari 1950, Palser 1975, Sporne 1974) and can therefore be interpreted as derived. In Doyle (1996), I expressed these differences in terms of two characters, lack of tiers and lack of a secondary suspensor. However, it seems questionable to treat the lack of tiers in the presumably reduced angiosperm embryo as the same state as their absence in the very different, massive embryos of cycads and *Ginkgo*. Because it seems premature to dissect these overlapping distinctions into separate characters, I have combined tiers and secondary suspensor into an unordered three-state character, with the angiosperm state defined by lack of tiers and lack of a secondary suspensor. This avoids specific assumptions about the most probable transitions among these states.

A related distinction is between free-nuclear and cellular embryogeny. In gymnosperms other than Gnetales, the zygote undergoes a more or less prolonged phase of free-nuclear divisions, whereas in *Welwitschia*, *Gnetum*, and angiosperms even the first divisions are cellular. The most problematic case is *Ephedra*, in which each of the two zygotes formed by the *Ephedra* type of double fertilization undergoes two free-nuclear divisions, but each of the eight resulting cells then develops into a single embryo by cellular divisions (Friedman 1992, 1994). This contrasts with the situation in other gymnosperms, in which each embryo is derived from several free nuclei, and resembles that in *Welwitschia*, *Gnetum*, and angiosperms. In Doyle (1996) I treated these variations as one character: whether the embryo was derived from several free nuclei or from one uninucleate cell by cellular divisions. However, this ignored the similarity between the initial free-nuclear phase in *Ephedra* and the free-nuclear phase in conifers. I assumed that the presence of free-nuclear divisions was an autapomorphy of *Ephedra*, following Friedman (1992, 1994), but this may have obscured real evidence for a transition between the conditions in conifers and Gnetales. Hence I have now recognized both sets of similarities by splitting early embryogeny into two characters: whether the first division of the zygote is free-nuclear (*Ephedra*, conifers, etc.) or cellular (*Welwitschia*, *Gnetum*, angiosperms), and whether each embryo is derived from several nuclei or from one (all Gnetales, angiosperms).

Another relevant character, recognized by Friedman and Carmichael (1998) and cited as a similarity between Gnetales and conifers by Friedman and Floyd (2001), is timing of development of the nourishing tissue of the female gametophyte: before fertilization in cycads, *Ginkgo*, and medullosans; both before and after in conifers, *Ephedra*, and *Welwitschia*; and after in *Gnetum*. I have added this as a new character, scoring *Lyginopteris*, *Callistophyton*, and cordaites as well as medullosans as the cycad-*Ginkgo* state because their ovules too apparently reached full size while still unfertilized (Stewart and Rothwell 1993). Angiosperms might be assigned to the same state as *Gnetum* by defining the character in terms of provisioning of the nourishing tissue in the seed, whether female gametophyte or endosperm (Friedman and Carmichael 1998, Fig. 9). However, because the late development of the nourishing tissue in angiosperms is closely tied to its origin from double fertilization, I have instead treated the angiosperm condition as a fourth state. This avoids bias toward either the view that it is related to the *Gnetum* state or the view that it was derived independently from the basic seed plant state. However, the angiosperm state overlaps with presence of endosperm, previously treated as an independent character, which I have therefore eliminated. Typical endosperm formation may

be a consequence of this shift in developmental timing and two other derived features, double fertilization (angiosperms, *Ephedra*, *Welwitschia*) and the pattern of female gametophyte cellularization, where the presence of one or two polar nuclei may set the stage for endosperm formation by the second fertilization and determine whether the endosperm is diploid or triploid.

Several seed characters of Bennettitales need reappraisal in the light of well-preserved petrified specimens described by Rothwell and Stockey (2002) and Stockey and Rothwell (2003). Following Crane (1985), Doyle and Donoghue (1986) and Doyle (1996) scored Bennettiales as having a cupule, based primarily on the description by Harris (1932, 1954) of an extra layer of cuticle outside the integument in the Triassic fossils *Vardekloeftia* and *Bennetticarpus crossospermus*. It should be noted, though, that Harris (1954) reported a single integument in several other Bennettitales; because it is unclear which condition was ancestral, it might have been more appropriate to score Bennettitales as uncertain. Harris (1932, 1954) also described *Vardekloeftia* as differing from other Bennettitales in lacking a thickened nucellar cuticle; he could not determine whether the integument was free from the nucellus (as in other Bennettitales) or fused. His interpretation of *Vardekloeftia* was reaffirmed by studies of Pedersen et al. (1989b).

Although some earlier authors had also interpreted an outer layer in *Cycadeoidea* as a cupule, Rothwell and Stockey (2002) and Stockey and Rothwell (2003) showed that this layer is part of a single integument, histologically differentiated into a sarcotesta, as in *Williamsonia*, and that the nucellus in both genera ended in a nucellar plug with no pollen chamber. They also reinterpreted the observations on *Vardekloeftia* by Harris (1932, 1954) and Pedersen et al. (1989b) in the same terms, arguing that the two outer cuticles represented the inner and outer epidermis of a single integument and that *Vardekloeftia* too had a nucellar plug and no pollen chamber. However, this interpretation is difficult to reconcile with the fact that the inner cuticle appears to form a normal tubular micropyle protruding through the outer cuticle (Harris 1932, 1954, Pedersen et al. 1989b). Their claim that *Vardekloeftia* had no pollen chamber does not conflict with Harris (1932) and Pedersen et al. (1989b), who reconstructed the apex of the nucellus as lacking a pollen chamber. Friis (pers. comm.) questions their identification of a nucellar plug on the grounds that the darker appearance of the relevant area is an artifact. These conflicts underline the difficulty in correlating observations based on macerated compressions and petrifactions studied with peels. There is an understandable tendency in paleobotany to favor data based on petrifactions, but both modes of preservation have practical advantages and disadvantages, and some aspects of seed anatomy may be easier to visualize from macerated cuticles. Broad-scale comparative studies of seeds preserved in both ways are needed to interpret such cases more confidently.

Because the interpretation of *Vardekloeftia* and relationships among the three groups of Bennettitales are unresolved, I formulated two alternative scorings for Bennettitales: (A) assuming the states shown by Rothwell and Stockey (2002) and Stockey and Rothwell (2003) in *Cycadeoidea* and *Williamsonia* apply to Bennettitales as a whole, (B) scoring the group as uncertain for characters where Harris (1932, 1954) and Pedersen et al. (1989b) interpreted *Vardekloeftia* differently. Differences in scoring of all affected characters are as follows:

78. Ovule-enclosing structures: orthotropous cupule (3) in Doyle (1996); no cupule (1) in data set A; no cupule or orthotropous cupule (1/3) in data set B.

96. Integument fusion: free or fused (0/1) in Doyle (1996); free (0) in data set A; free or fused (0/1) in data set B.

102. Nucellar cuticle: thin or thick (0/1) in Doyle (1996); thick (1) in data set A; thin or thick (0/1) in data set B.

Following a suggestion by Owi Nandi (pers. comm), I have also added a previously overlooked character for presence or absence and distribution of fibers in the secondary phloem. Esau (1969) noted the presence of uniseriate tangential bands of fibers, usually in a highly regular alternation with parenchyma and sieve cells, as a feature of most conifers other than Pinaceae. A similar character (but without such regular alternation of cell types) has been used in angiosperms, where it appears to be a synapomorphy of Magnoliales (Doyle and Endress 2000, Sauquet et al. 2003). In contrast, fibers (not counting sclereids in the oldest phloem) are lacking in Pinaceae and Gnetales (Carlquist 1996b), which would be consistent with a position of Gnetales with or near Pinaceae rather than nested among other conifers. Cycads, *Ginkgo*, and Araucariaceae differ from both extremes in having varying numbers of fibers, usually in-

creasing in the older phloem, which sometimes form thicker, less regular tangential bands than those in most conifers. These three patterns of fiber distribution, recognized by den Outer (1967) and Smoot (1984b), are treated here as unordered states. Conditions in fossil taxa were reviewed by Smoot (1984b) and Taylor (1990). *Callistophyton* (Smoot 1984a), cordaites (Taylor 1988), and other Paleozoic seed ferns not included here had secondary phloem consisting entirely of alternating sieve cells and parenchyma (Pinaceae and Gnetales differ in that sieve cells predominate), and fibers are said to be lacking in *Lyginopteris* (Williamson and Scott 1896, cited by Smoot 1984b). However, *Medullosa* (Smoot 1984b) and Bennettitales (*Cycadeoidea*: Taylor 1990) had irregular tangential bands of fibers, increasing outward, a condition that Smoot (1984b) likened to that in cycads. Sharma and Bohra (1977) also reported irregular fiber bands in Bennettitales (*Bucklandia*), but not in *Pentoxylon*. The angiosperms in the present data set include none of the taxa with tangential fiber bands. Based on Metcalfe (1987) and Carlquist (1993, 1999, 2001), only Schisandraceae and possibly *Illicium* (scored ?) have phloem fibers.

The analysis of Doyle (1996) included *Piroconites*, a Jurassic fossil thought to be related to Gnetales because of its linear, multiveined, opposite leaves and striate ephedroid pollen (Kirchner 1992, van Konijnenburg-van Cittert 1992, Crane 1996). Because *Piroconites* had reproductive structures consisting of a scale-like ''sporophyll'' adnate to the top of a leaf, it seemed to offer evidence for a relationship between Gnetales and glossopterids (Doyle 1996). However, TEM work by Osborn (2000) showed that the description of the pollen as striate was due to misinterpretation of folded grains, so I have deleted *Piroconites* from the data set. The possibility that *Piroconites* is related to Gnetales cannot be excluded, but in the absence of striate pollen there are few characters to support its position. It illustrates the dangers of including taxa for which too few characters are preserved.

ANGIOSPERM TAXA AND CHARACTERS. I have modified the sampling of angiosperms from that of Doyle (1996), removing groups such as Magnoliales that now appear to be relatively nested and adding *Amborella* and other taxa that are basal in strongly supported molecular trees (e.g., Zanis et al. 2002). The taxon sampling is the same as in Eklund et al. (2004), including all the ANITA taxa, Chloranthaceae, and three relatively plesiomorphic representatives of divergent lines among the remaining groups, the eumagnoliid taxa Asaroideae (Aristolochiaceae), Saururaceae, and Winteraceae (see Eklund et al. 2004 for discussion), but with Nymphaeales and Chloranthaceae reduced to single taxa. When states varied within the last two taxa, they were scored as having ancestral states inferred from the internal topologies found by Les et al. (1999), Doyle and Endress (2000), and Eklund et al. (2004).

I added all characters from the angiosperm analysis of Doyle and Endress (2000) that are potentially informative with the present taxon sampling. Some of these were treated as additional states of characters used by Doyle (1996) at the seed plant level. Scoring of angiosperm taxa follows Doyle and Endress (2000), with modifications based on Sauquet et al. (2003) and Eklund et al. (2004). Several characters, for example involving floral morphology, were scored only in angiosperms. These are irrelevant in analyses in which angiosperm relationships were constrained to the molecular topology, but they are of interest in evaluating the congruence of morphological characters with molecular results.

I modified other characters to reflect the discovery that Nymphaeales and Austrobaileyales (but not *Amborella*) have diploid rather triploid endosperm, derived from a four- rather than eight-nucleate female gametophyte (Williams and Friedman 2002, 2004, Friedman et al. 2003). Because Friedman and Williams (2003, 2004) argued persuasively that the eight-nucleate embryo sac is a result of duplication of a four-nucleate module, with the three antipodals corresponding to the egg and two synergids, I have treated the two angiosperm conditions as elements of two separate characters: one a redefinition of the angiosperm state in the female gametophyte organization character (discussed above) based on the nature of the module (three grouped cells and one free nucleus), the other for presence of one or two modules. An alternative would be to treat the angiosperm conditions as two states of a single four-state character. However, this would have obscured the close similarity between the two angiosperm types and their marked differences from types seen in other seed plants.

ANALYSES. Data were analyzed with the parsimony program PAUP (Swofford 1990), using heuristic search methods. These involved 100

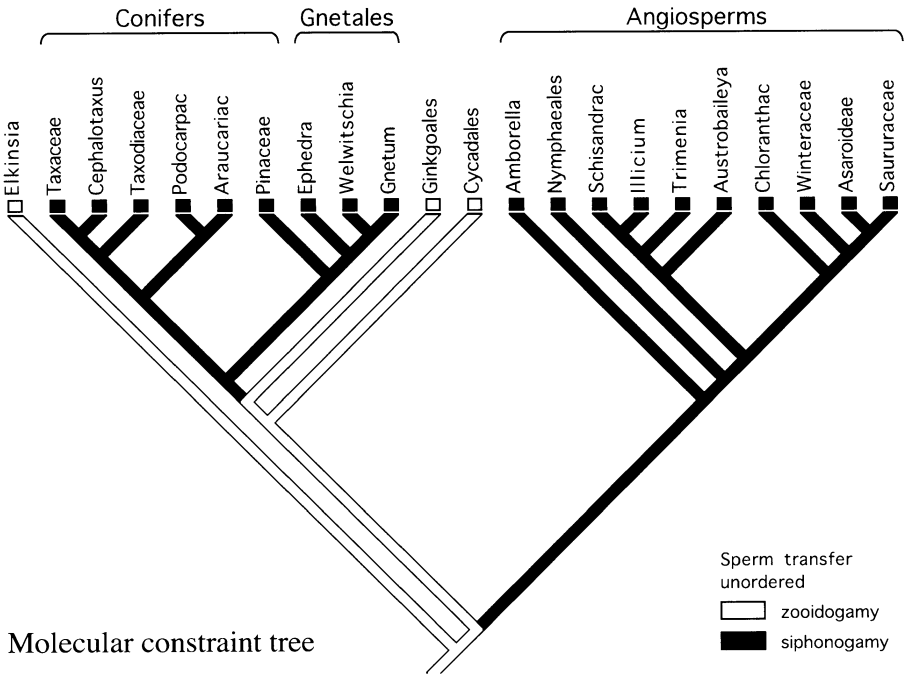

FIG. 5. Relationships among extant seed plants based on molecular data, used as a backbone constraint tree in subsequent analyses, with shading showing the inferred evolution of sperm transfer (character 73).

replicate analyses with stepwise random addition of taxa, holding multiple most parsimonious trees (MULPARS), and tree bisection-reconnection (TBR) branch swapping. In all analyses *Elkinsia* was specified as the outgroup to the remaining taxa. The relative parsimony of alternative hypotheses was determined by moving taxa with MacClade (Maddison and Maddison 2001) or by searching with PAUP for most parsimonious trees consistent or inconsistent with appropriate constraint trees, which were usually ''backbone'' constraints containing only a subset of taxa, such as living seed plants.

To evaluate the implications of molecular analyses, data were analyzed with the living groups forced into the currently best-supported molecular arrangement with a backbone constraint tree (Fig. 5). Fossil taxa attach to this framework wherever this is most parsimonious in terms of morphology.

Support for relationships was quantified with bootstrap analysis (Felsenstein 1985) and Bremer support, or decay analysis (Bremer 1988, Donoghue et al. 1992). Bootstrap analyses used 1000 replicates, each involving a single heuristic search with closest taxon addition sequence, in which five trees were held at each step during stepwise addition. Only 100 most parsimonious trees were saved per bootstrap replicate, but TBR branch swapping was allowed to continue after reaching this limit in order to increase the chance of finding shorter trees.

Decay analyses were conducted by searching for trees equal to or shorter than a given number of steps and then observing which clades were no longer present in the strict consensus. Decay indices of clades that remained when the search yielded more trees than could be retained in memory (30,000) were determined by searching for shortest trees not consistent with a constraint tree in which the relevant taxa formed a clade. This procedure was not possible in analyses in which modern groups were constrained into the molecular arrangement, but this was inconsequential because the only clades that remained when tree numbers exceeded memory were some of those whose relationships were constrained.

Character evolution was reconstructed by MacClade (Maddison and Maddison 2001), which optimizes character changes on a tree based on parsimony. MacClade was also used to identify characters that unequivocally change at each node and to study different optimizations of equivocal characters. When characters are cited as uniting clades, these are unequivocal syn-

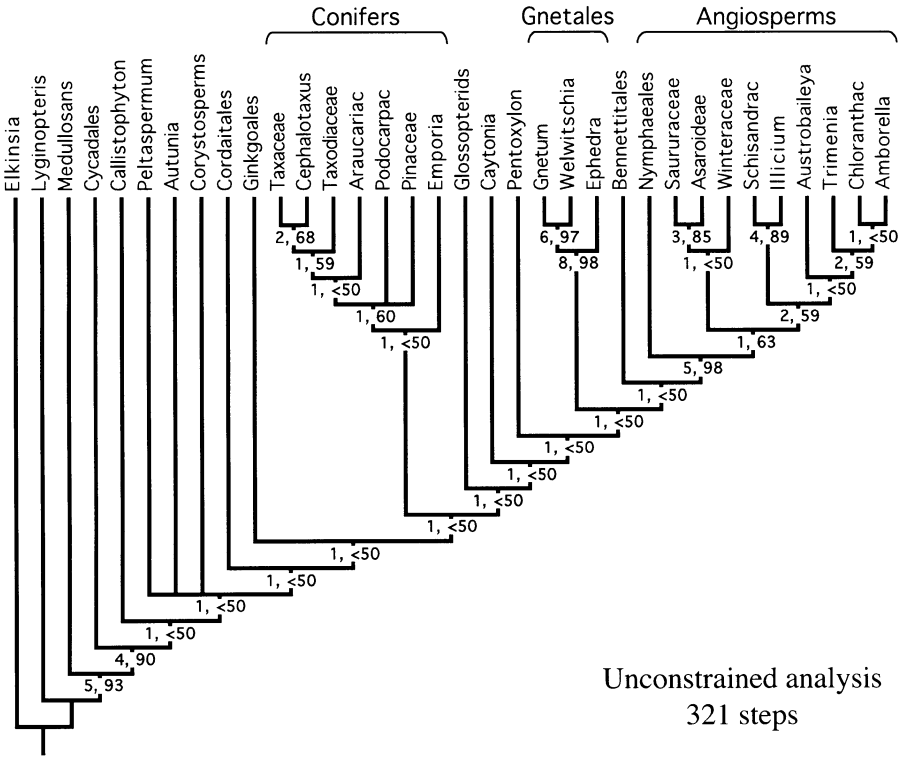

FIG. 6. Strict consensus of eight most parsimonious trees of 321 steps obtained from the unconstrained analysis, with decay and bootstrap support values for nodes.

apomorphies as determined by MacClade, unless otherwise indicated.

Results. Analyses of the two data sets (A and B) differing in interpretation of seed characters in Bennettitales gave identical trees of the same lengths, both with and without constraints. Subsequent remarks will refer to data set B, for reasons discussed further below.

The unconstrained analysis yielded eight most parsimonious trees of 321 steps (strict consensus in Fig. 6). These show the same arrangement of Devonian-Carboniferous seed ferns found in all previous analyses (allowing for variations in taxon sampling), with *Elkinsia*, *Lyginopteris*, and medullosans branching successively below a ''platysperm'' clade that contains cycads, the Late Carboniferous seed fern *Callistophyton*, and all remaining taxa, including coniferophytes (Crane 1985, Doyle and Donoghue 1986, Nixon et al. 1994, Rothwell and Serbet 1994, Doyle 1996). As in previous studies, Gnetales are the closest living relatives of angiosperms, but the two taxa belong to an anthophyte clade that also includes Bennettitales and *Pentoxylon*. These are linked with glossopterids and *Caytonia*, together forming a clade called glossophytes by Doyle (1996), but with a different internal arrangement of taxa. Glossophytes are nested within coniferophytes, as the sister group of conifers (including the Paleozoic genus *Emporia*).

Although this result reaffirms the anthophyte hypothesis, trees in which Gnetales are related to conifers rather than anthophytes are only one step less parsimonious. When I forced Gnetales and living conifers into a clade, using a backbone constraint tree of living taxa with all other relationships unresolved, I obtained 16 trees of 322 steps (representative tree in Fig. 7, with nodes not found in all trees indicated by arrows). Gnetales are nested within conifers, but not linked with Pinaceae; they may be sister to either Araucariaceae or a clade consisting of Araucariaceae, Taxodiaceae (including Cupressaceae), *Cephalotaxus*, and Taxaceae. Two pollen characters that support both positions are loss of air sacs and granular exine structure. The remaining glossophytes are much lower, linked with cycads by simple pinnate leaves and seed shed with mature embryo (scored as unknown

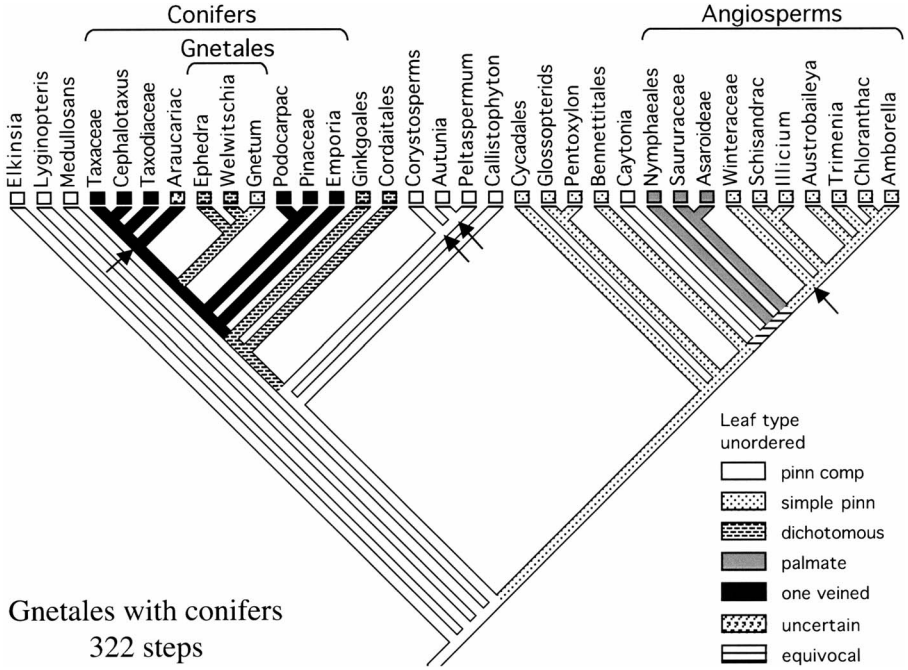

FIG. 7. Representative most parsimonious tree of 322 steps obtained from the analysis with Gnetales and conifers constrained to form a clade, with nodes not found in all most parsimonious trees indicated by arrows, and with shading showing the inferred evolution of leaf organization (character 26).

in glossopterids, *Pentoxylon*, and *Caytonia*). Their internal topology is more like that of Doyle (1996), with glossopterids and *Pentoxylon* forming a basal clade and *Caytonia* the sister group of angiosperms. Although trees in which cycads are related to glossophytes are most parsimonious, trees in which they are on the line leading to other living gymnosperms or basal to both lines are only two steps longer.

In the ''one-off'' trees in which Gnetales are nested in conifers (Fig. 7), the topology of living conifer families (setting aside Gnetales) is the same as that found by Doyle (1996), which differs from molecular trees (e.g., Magallón and Sanderson 2002, Quinn et al. 2002) only in that Podocarpaceae are linked with Pinaceae (based on two microsporangia per sporophyll) rather than Araucariaceae. However, in the unconstrained trees (Fig. 6), Podocarpaceae may be linked with either Pinaceae or the remaining conifers (based on tangential bands of phloem fibers).

Nymphaeales are sister to all other angiosperms in both sorts of trees (Figs. 6, 7), as in the analysis of Doyle (1996), which included only one other member of the basal ANITA grade (*Austrobaileya*). In contrast to molecular trees, the other ANITA taxa (*Amborella*, *Austrobaileya*, *Trimenia*, *Illicium*, Schisandraceae) form a clade that also includes Chloranthaceae, separated from Nymphaeales by Piperales (Saururaceae, Asaroideae) and Winteraceae. The only variation is that Winteraceae are linked with Asaroideae and Saururaceae in the unconstrained analysis (Fig. 6) but form an adjacent line in some trees with Gnetales in conifers (Fig. 7). These relationships are similar to those found in the morphological analysis of Doyle and Endress (2000), which included many more taxa, and identical to those in the unconstrained analysis of Eklund et al. (2004), which used the same sampling of angiosperms, allowing for the fact that those studies rooted the angiosperms on *Amborella*.

The analysis with living taxa forced into the molecular topology yielded 18 most parsimonious trees of 339 steps (Fig. 8), which form two islands of 6 and 12 trees. The same relationships outside angiosperms were found when *Amborella* and Nymphaeales were constrained to form a basal clade, as in Barkman et al. (2000). This represents an increase of 18 steps over the unconstrained analysis. Most of the extra steps are due to the different arrangement of taxa in angiosperms (10) and conifers (3) and the association of cycads with other living gymnosperms

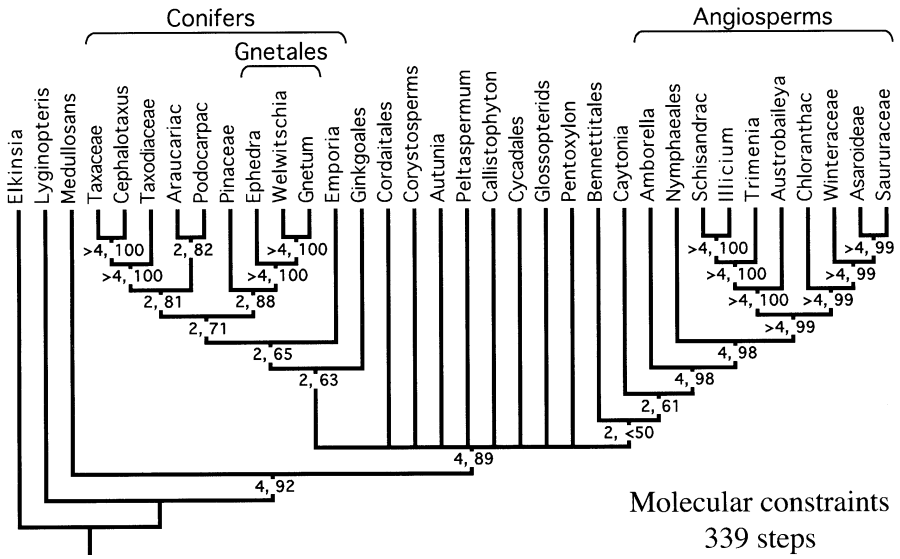

FIG. 8. Strict consensus of 18 most parsimonious trees of 339 steps obtained from the analysis with the molecular constraint tree (Fig. 5), with decay and bootstrap support values.

(2). Paleozoic seed ferns branch in the previously observed order at the base. Medullosans are united with the platysperms by bilateral pollen symmetry, loss of the original lobed cupule, loss of the central column in the lagenostome, sarcotesta, and vascularized nucellus. Synapomorphies of the platysperms include a sulcus, honeycomb-alveolar exine structure, sealed micropyle, platyspermic seeds (defined by presence of two vascular bundles or other anatomical signs of bilateral or bisymmetric organization, not necessarily flattened shape), and, in some trees, endarch primary xylem, simple pinnate leaves, abaxial microsporangia, and linear megaspore tetrad.

The poor resolution at the base of the platysperms (Fig. 8) reflects the existence of two islands of trees and the ambiguous position of *Callistophyton*. In island 1 (representative tree in Fig. 9, with nodes not found in all trees indicated by arrows), glossophytes diverge at the base of the platysperms, and their internal topology (not counting the molecular arrangement in angiosperms) is the same as in one-off trees with Gnetales in conifers (Fig. 7). Relationships are more poorly resolved in the consensus of island 2 (representative tree in Fig. 10), with a basal polytomy in the platysperms involving all groups except coniferophytes and the clade made up of Bennettitales, *Caytonia*, and angiosperms. However, inspection of individual trees shows that this lack of resolution is due to "jumping" of *Callistophyton* between two widely separated parts of the tree: nested within the clade including living gymnosperms in seven trees, between glossopterids and coniferophytes, along with corystosperms and peltasperms; and just below the common ancestor of living gymnosperms and angiosperms in five trees. In all 12 trees, cycads, *Pentoxylon*, and glossopterids are attached in that order at the base of the gymnosperm line. Thus the two islands represent different rootings of the platysperms and different unrooted relationships in the vicinity of cycads, glossopterids, and *Pentoxylon*.

Character state changes on the tree shown in Fig. 9, with nodes numbered in Fig. 11, are listed in Table 1. Unequivocal synapomorphies of each clade or terminal taxon are listed first, then equivocally optimized changes. Positions of the latter changes were sometimes chosen assuming accelerated transformation (acctran), with an early origin of the derived state followed by reversals, sometimes delayed transformation (deltran), with later multiple origins. Deltran was chosen when the derived state represented loss of a structure or some other such change that seemed more likely to have occurred twice than to have reversed. In angiosperms, results of the more extensive analysis of Doyle and Endress (2000) were sometimes used to choose between equally parsimonious optimizations. Synapomorphies of extant groups that involve characters not preserved in fossil outgroups were

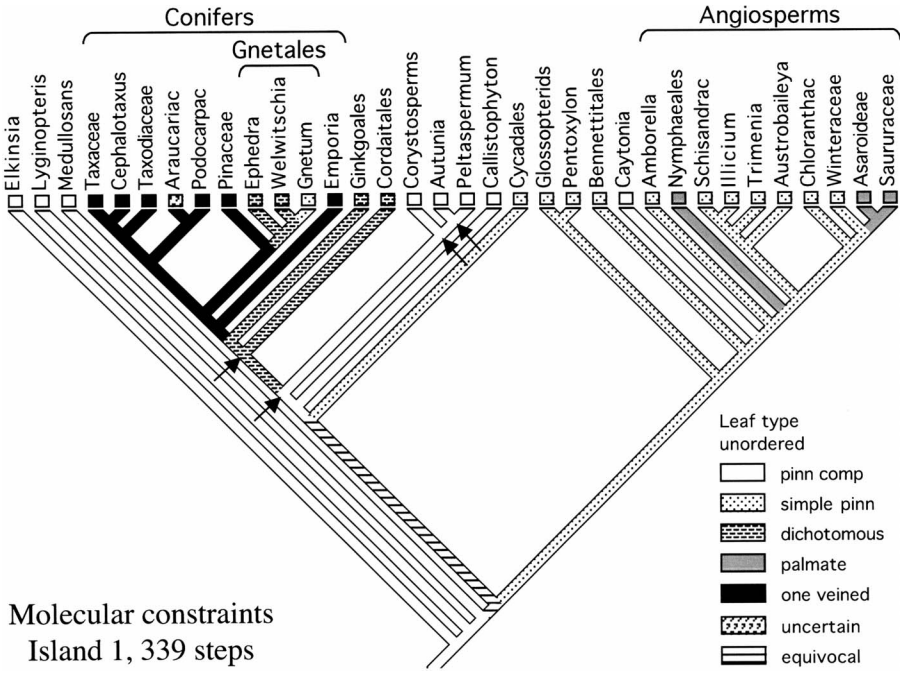

FIG. 9. Representative tree from island 1 from the analysis with molecular constraints, with nodes not found in all trees making up the island indicated by arrows, and with shading showing the inferred evolution of leaf organization (character 26).

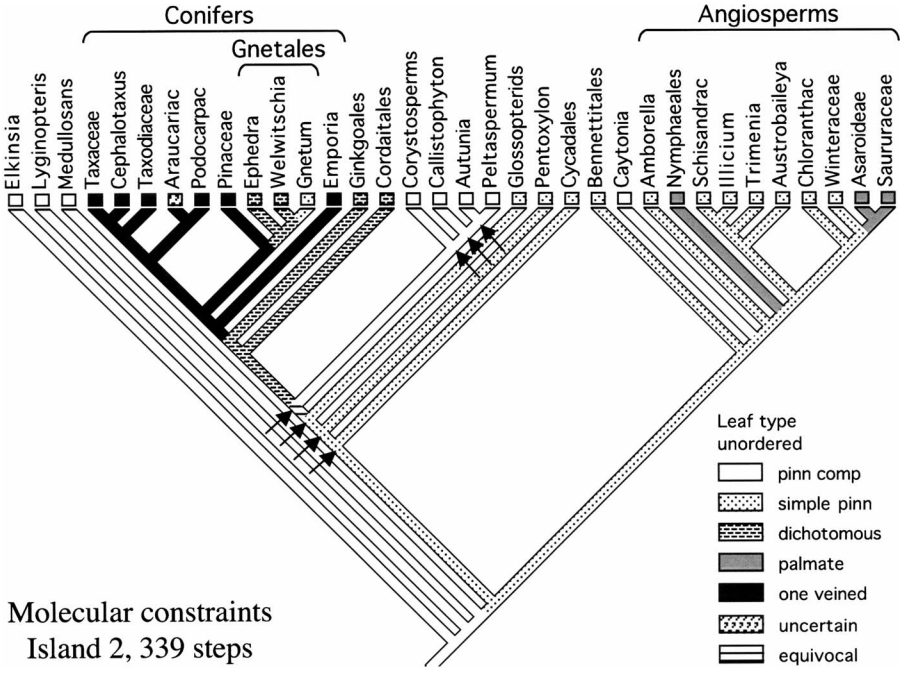

FIG. 10. Representative tree from island 2 from the analysis with molecular constraints, with nodes not found in all trees making up the island indicated by arrows, and with shading showing the inferred evolution of leaf organization (character 26).

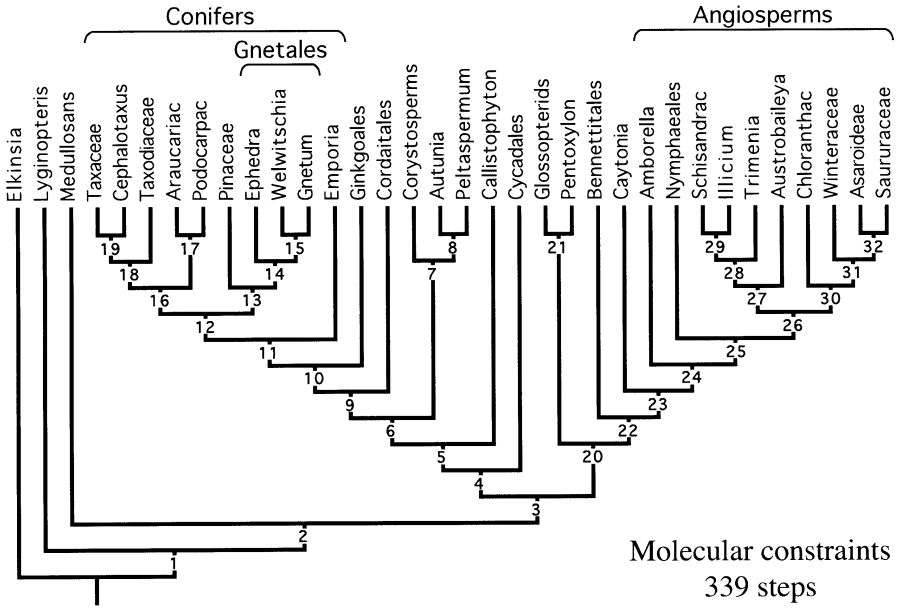

FIG. 11. Same tree from the analysis with molecular constraints as in Fig. 9, with nodes numbered for reference to the list of character state changes in Table 1.

placed at the crown-group node; such features may have arisen lower on the stem lineage.

Consistent with the ambiguous rooting of the platysperms, there is no unequivocal synapomorphy for the clade that includes extant gymnosperms; a possible candidate (equivocal because the basic state in glossophytes is unknown) is abaxial microsporangia. The only unequivocal synapomorphy of the glossophyte clade is loss of the lagenostome. Simple, pinnately veined leaves (i.e., with a midrib, unlike simple, dichotomously veined leaves in coniferophytes) are homologous throughout glossophytes (modified to palmately compound in *Caytonia*) but equivocal as a synapomorphy of the clade because they may or may not be homologous with the similar leaves of cycads (Fig. 9). Another derived feature that is restricted to glossophytes but equivocal as a synapomorphy is adaxial ovules, discussed further below. Glossophytes also share a thick nucellar cuticle, versus thin in all members of the living gymnosperm line where this character is known, but its status is equivocal because appropriate data are lacking for basal seed ferns. Loss of the megaspore membrane is a derived feature of all glossophytes except glossopterids, but because *Pentoxylon* is linked with glossopterids, it is equivocal whether this loss occurred independently in *Pentoxylon* and other glossophytes or (perhaps less plausibly, considering that glossopterids are the oldest members of the clade) occurred once and was reversed in glossopterids.

Within glossophytes, glossopterids and *Pentoxylon* are united by uniseriate rays, secondarily free microsporangia, and paddle-like megasporophylls. Bennettitales are linked with *Caytonia* and angiosperms by presence of scalariform pitting in the secondary xylem, endarch leaf traces, siphonogamy, and reduced pollen chamber (all but the last being characters that are unknown in *Caytonia*). *Caytonia* is linked with angiosperms by reticulate venation (initially of one vein order), unraised guard cell poles, anatropous cupules (including bitegmic ovules), and loss of nucellar vasculature. The present topology implies that reticulate venation is not homologous in glossopterids and *Caytonia*, because it would have to be lost independently in *Pentoxylon* and Bennettitales. Saccate pollen may be homologous throughout the living gymnosperm clade (*Callistophyton*, *Autunia*, corystosperms, cordaites, conifers), but not in glossopterids and *Caytonia*. Problems concerning these characters and other potential synapomorphies are discussed below.

Decay and bootstrap values from the unconstrained analyses (Fig. 6) reveal fairly strong support for relationships at the base of the seed plants, including the association of medullosans

Table 1. Character state changes on a representative tree with extant taxa constrained into the molecular arrangement (Fig. 11). Unequivocal changes listed first, then equivocally optimized changes in parentheses (see text for discussion).

Node 1: (5 0>1, 95 0>1).
Lyginopteris: 96 0>1 (6 0>1).
Node 2: 62 0>1, 78 0>1, 98 0>1, 100 0>1, 101 0>1 (20 0>2).
Medullosans: 18 0>1 (72 0>1).
Node 3 (platysperms): 7 0>1, 61 0>1, 64 0>1, 94 0>1, 99 0>1, 107 0>1 (6 0>1).
Node 4 (extant gymnosperms): (49 0>1).
Cycadales: 18 0>1, 23 0>1, 96 0>1 (26 0>1, 72 0>1, 76 0>3, 120 0>1).
Node 5: 63 0>1, 95 1>2 (20 2>1, 76 0>1).
Callistophyton: 7 1>0 (27 0>1).
Node 6: 15 0>1, 52 1>0.
Node 7: (74 0>1).
Corystosperms: 108 0>1 (23 0>1, 24 0>1).
Node 8 (peltasperms): 28 0>1 (27 0>1).
Autunia: none.
Peltaspermum: 63 1>0.
Node 9 (coniferophytes): 26 0>2, 48 0>1 (24 0>1, 74 0>2, 76 1>0).
Cordaitales: 39 0>2, 49 1>0, 61 1>0.
Node 10: 10 0>1, 101 1>0 (120 0>1).
Ginkgoales: 3 0>1, 18 0>1, 23 0>2, 53 0>1 (8 0>1, 63 1>0, 72 0>1, 96 0>1).
Node 11 (conifers): 26 2>4, 39 0>1, 40 0>1, 77 0>1, 100 1>0.
Emporia: 49 1>2, 61 1>0, 80 0>1, 99 1>0 (20 1>0).
Node 12: 73 0>1, 97 0>1, 98 1>2 (8 0>1, 20 1>2, 71 1>0, 96 0>1, 114 0>1, 118 0>1, 121 0>1).
Node 13: (113 0>1).
Pinaceae: 51 0>1 (41 0>1).
Node 14 (Gnetales): 2 0>1, 4 0>1, 12 0>1, 15 1>0, 21 0>1, 22 0>2, 23 0>2, 26 4>2, 39 1>2, 40 1>0, 49 1>0, 50 0>1, 52 0>1, 65 0>1, 75 0>1, 79 0>1, 95 2>3, 117 0>1 (63 1>0, 64 1>2, 101 0>1).
Ephedra: 20 2>0, 98 2>1 (61 1>2).
Node 15: 32 0>1, 34 0>1, 36 0>1, 70 0>1, 109 0>1, 110 0>1, 112 0>1, 116 0>1, 119 0>1 (31 0>1).
Welwitschia: 10 1>0 (14 0>1).
Gnetum: 23 2>1, 26 2>1, 62 1>2, 94 1>0, 114 1>2 (61 1>2, 101 1>0).
Node 16: 18 0>2 (41 0>1).
Node 17: 80 0>1.
Araucariaceae: 4 0>1/2, 18 2>1, 96 1>0 (42 0>1, 62 1>0, 63 1>0, 64 1>2).
Podocarpaceae: 51 0>1.
Node 18: 70 0>2 (62 1>0, 63 1>0, 64 1>2, 77 1>0).
Taxodiaceae: (42 0>1).
Node 19: 11 0>1, 60 0>1.
Cephalotaxus: 100 0>1.
Taxaceae: 39 1>0, 40 1>0, 61 1>2, 62 0>2, 75 0>1, 78 1>3.
Node 20 (glossophytes): 97 0>1 (23 0>2, 26 0>1, 76 0>2, 102 0>1).
Node 21: 15 0>1, 52 1>0, 74 0>1 (20 2>0).
Pentoxylon: 64 1>2 (3 0>1, 108 0>1).
Glossopterids: 31 0>1, 63 0>1, 65 0>1, 100 1>0.
Node 22: 9 0>1, 24 0>1, 73 0>1, 98 1>2 (34 0>1, 49 0>2, 108 0>1, 120 0>1).
Bennettitales: 18 0>1, 64 1>2, 95 1>3 (23 2>0).
Node 23: 31 0>1, 33 0>1, 78 1>2, 101 1>0 (100 1>0).
Caytonia: 26 1>0, 63 0>1 (34 1>0, 49 2>1).
Node 24 (angiosperms): 32 0>1, 48 0>2, 53 0>2, 64 1>3, 69 0>1, 74 0>3 (4 0>2, 17 0>1, 20 2>0, 21 0>1, 68 0>1, 70 0>3, 110 0>2, 113 0>1, 114 0>3, 116 0>1, 117 0>1, 118 0>2, 121 0>1).
Amborella: 22 0>1, 60 0>1, 67 0>1, 78 2>3, 111 0>1 (43 0>1, 87 0>2, 92 0>1).
Node 25: (104 1>0).
Nymphaeales: 1 0>1, 26 1>3, 35 0>1, 56 1>0/2, 57 0>1, 82 0>1, 105 0>1, 115 0>1 (30 1>0, 34 1>0, 44 0>1, 46 0>1, 54 0>1, 68 1>0, 87 0>1, 90 0>1, 121 1>0).
Node 26: 12 0>1, 37 0>1, 66 0>1.
Node 27 (Austrobaileyales): 103 1>2, 106 0>1 (22 0>2).
Austrobaileya: 86 0>1 (30 1>0, 85 0>1, 90 0>1, 91 0>1).
Node 28: 38 0>1, 105 0>1 (89 0>1, 104 0>1).
Trimenia: 49 2>3, 56 1>2, 67 0>1, 81 0>1, 87 0>2.
Node 29: 6 1>2, 23 2>0, 36 0>1, 57 0>1 (22 2>0, 29 1>0, 61 1>3, 62 1>0, 85 0>1).
Illicium: 19 1>0, 35 0>1, 86 0>1, 92 0>2, 93 0>1 (30 1>0, 91 0>2).
Schisandraceae: 43 0>1, 50 0>1 (18 0>1, 90 0>1, 91 0>1).
Node 30: 16 0>1, 19 1>0, 49 2>3, 111 0>1 (29 1>0, 44 0>1, 46 0>1).
Chloranthaceae: 43 0>1, 56 1>0, 59 0>1, 67 0>1, 78 2>3, 81 0>1 (22 0>2, 45 0>2, 58 0>1).
Node 31 (eumagnoliids): 23 2>3, 83 0>1, 84 0>1 (30 1>0, 47 0>1, 54 0>1, 55 0>1, 88 1>0, 90 0>1, 91 0>1).
Winteraceae: 12 1>0, 46 1>2, 62 1>0, 105 0>1 (55 1>2, 57 0>1, 58 0>1, 85 0>1).
Node 32 (Piperales): 1 0>1, 25 0>1, 26 1>3, 35 0>1, 56 1>2, 82 0>1, 86 0>1, 103 1>0 (13 0>1, 22 0>1, 29 0>1, 34 1>0, 45 0>1, 92 0>2, 93 0>1, 104 0>1).
Asaroideae: 14 0>1, 49 3>1, 87 0>1 (85 0>1).
Saururaceae: 23 3>1, 59 0>1, 78 2>3, 89 0>1, 115 0>1 (34 0>2, 45 1>2, 57 0>1, 66 1>0).

with platysperms (decay index 5 steps, bootstrap frequency 93%) and the monophyly of platysperms (4 steps, 90%), as well as for Gnetales (8 steps, 98%) and angiosperms (5 steps, 98%). However, support values for near-basal nodes in the platysperms are low, including those for nodes in the glossophyte clade (1 step, <50%). When living taxa are constrained into the molecular topology (Fig. 8), support values for relationships that involve fossil taxa are generally similar to those found without constraints. Decay support for the relationship of angiosperms with *Caytonia* and Bennettitales is slightly higher (2 steps) than other relationships in glossophytes, but bootstrap support remains low (61% for association of *Caytonia* with angiosperms, <50% for association of Bennettitales).

Discussion. The fact that both anthophyte and conifer relationships of Gnetales became almost equally parsimonious after the character revisions made here suggests that morphology does not conflict as strongly with molecular data on the position of Gnetales as it seemed. Apparently much of the conflict between previous morphological and molecular results was due to difficulties in assessing homology in certain morphological characters. A skeptic might argue that this change in parsimony only indicates that morphological characters can be reinterpreted at will to support any desired relationship. How-

ever, this would be unduly pessimistic. In the process of reciprocal illumination outlined above, closer examination showed that some characters thought to support the anthophyte hypothesis are instead equally consistent with either relationship, whereas others had been interpreted incorrectly. At the time of the first molecular analyses it seemed that morphology strongly favored the anthophyte hypothesis, whereas molecular data were equivocal, suggesting the morphological result should be accepted (Doyle 1998b). But now the situation is reversed: more voluminous and better-analyzed molecular data strongly contradict the anthophyte hypothesis, whereas morphological data are ambiguous. On a more positive note, the inference that morphology is less positively misleading than it seemed may be grounds for optimism about the prospects of using morphological data to fit fossil taxa into a molecular framework of living taxa.

As in Doyle (1996), the bootstrap and decay analyses (Figs. 6, 8) indicate that the strongest results concern relationships near the base of the seed plants, including the monophyly of platysperms, which correspond roughly to crown-group seed plants (depending on the position of *Callistophyton*), and the monophyly of Gnetales and angiosperms. In the constrained analysis (Fig. 8), bootstrap values below 100% in extant conifers and angiosperms, within which relationships were fixed, must be due to ''infiltration'' of fossil outgroups into the crown groups in some bootstrap replicates. A similar effect can be seen by examining trees found during the decay analyses. For example, in trees found when angiosperms were specified as not forming a clade (five steps longer than the shortest trees), *Caytonia* was nested within angiosperms. Thus the present numbers may underestimate the true support for angiosperm monophyly, because trees with *Caytonia* nested in angiosperms assume that *Caytonia* had angiosperm synapomorphies in embryological and other characters that are not preserved, which may be true in some cases, but probably not in all.

Unfortunately, the low support for relationships in basal platysperms and glossophytes means that the question of angiosperm relationships is still far from resolved. In the unconstrained analyses, the low values may reflect almost equal support for placement of Gnetales in glossophytes and in conifers, plus uncertain relationships among other fossil platysperms. Support in the vicinity of conifers is slightly higher in the constrained analysis (Fig. 8), where Gnetales were not allowed to ''jump'' out of conifers. The slightly higher decay support for relationships of angiosperms with *Caytonia* and Bennettitales in the constrained analyses may reflect the fact that Gnetales are no longer in the picture, but why bootstrap support for these relationships remains low is unclear. These results mean that the position of angiosperms among glossophytes is only a best guess, which may however serve as a focus for future investigations in paleobotany and in evolution and development. The strongest inference is that both angiosperms and other living seed plants are nested among Paleozoic seed ferns, in the platysperm clade, and that homologies for angiosperm organs are to be sought among fossil members of this clade, rather than in more basal seed ferns.

The unconstrained trees (Fig. 6, with leaf character in Fig. 12) are reminiscent of some trees that were one step longer than the shortest trees in the analysis of Doyle (1996, Fig. 9), which differed in relationships among glossophytes and the fact that their sister group was cordaites rather than conifers. This result recalls the view of Schopf (1976) that glossopterids were related to coniferophytes, with their simple, pinnately veined leaves derived from cordaite- or *Ginkgo*-like simple leaves with dichotomous venation by aggregation of veins into a midrib, and with their ''fertiliger'' (leaf-cupule complex) derived from a bract-axillary fertile short shoot unit of the type found across coniferophytes. It also recalls the suggestion of Crane et al. (2004) that among modern plants angiosperms are related to conifers and Gnetales, but cycads and *Ginkgo* are more basal, based on the restriction of siphonogamy to conifers, Gnetales, and angiosperms. Ironically, though, siphonogamy would not be a valid synapomorphy if fossil taxa are interpolated as found here, given the discovery of zooidogamy in glossopterids (Nishida et al. 2003, 2004) and its presumed occurrence in cordaites (e.g., Poort et al. 1996), in addition to more basal seed ferns (Benson 1908, Stewart 1951). However, trees found when Gnetales were forced together with conifers (Fig. 7) or living taxa were constrained into the molecular topology (Figs. 8, 9, 10) imply rather that the glossophyte line diverged earlier from platyspermic seed ferns. This emphasizes again that relationships near the base of the platysperms are poorly resolved, whether because of the smaller number of known characters in

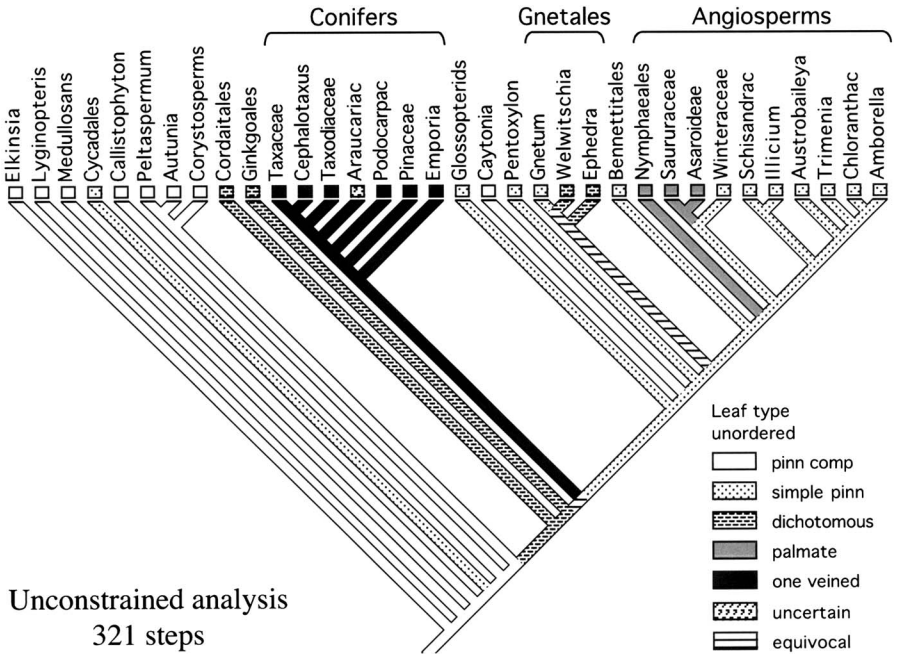

FIG. 12. Representative tree from the unconstrained analysis, with shading showing the inferred evolution of leaf organization (character 26).

Permian and Mesozoic fossils than in Carboniferous forms, very rapid radiation after origin of the clade, or both. Better evidence on anatomical and life cycle features in Permian and Mesozoic taxa could have a significant impact and should be a high priority for students of seed plant phylogeny.

Given that the analysis with Gnetales forced together with conifers placed cycads on the line leading to angiosperms (Fig. 7), but trees with cycads on the line leading to other living gymnosperms (as in most molecular analyses) or below both lines are only two steps longer, the conflict between morphological and molecular data on cycads is not severe. The position of cycads is one of the more weakly supported aspects of molecular phylogenies (cf. Magallón and Sanderson 2002, Soltis et al. 2002). Considering extant taxa alone, it might seem that different positions of cycads would have very different implications for character evolution in seed plants. However, trees with all three positions of cycads actually imply rather similar scenarios for the evolution of most characters, such as leaf morphology; differing positions of Permian and Mesozoic groups have a greater effect (compare Figs. 7, 9, 10, and 12). This is because Paleozoic seed ferns remain at the base of seed plants and relationships within the two main platysperm lines are often similar. This reaffirms the view that incorporation of fossil taxa into molecular trees can be necessary in order to gain a proper understanding of character evolution in ancient groups such as seed plants, even when relationships among living taxa are not affected (Doyle and Donoghue 1987, Donoghue et al. 1989).

All these trees are troubling in indicating that pollen germination through a distal sulcus originated at the base of platysperms, implying that the tetrad scar and proximal pollen germination of cordaites and Paleozoic conifers (such as *Emporia*) are not primitive features, as generally assumed (e.g., Poort et al. 1996), but rather reversals. This, together with the fact that cordaites are the oldest known platysperms, could be evidence for a more basal position of coniferophytes.

In discussing the implications of these results, there would be little justification for using a tree from the unconstrained analysis, since some of the relationships within angiosperms are strongly contradicted by molecular data. This is evident from the combined analysis of Doyle and Endress (2000), where molecular data overruled morphological data in most cases, for example in placing both Nymphaeales and other ANITA taxa together in a basal grade. Exceptions, where

morphology overcame weakly supported molecular relationships, concerned taxa not included in the present data set (e.g., association of Lauraceae with Hernandiaceae, Piperales with monocots). The ideal procedure would be to combine the present data with DNA sequences and analyze them together, but in the absence of this, and because the results of Doyle and Endress (2000) indicate that DNA would dominate in the taxa sampled here, I will instead concentrate on the analysis with living taxa constrained into the molecular arrangement (Fig. 8). I will consider data set B, with seed characters of Bennettitales scored as uncertain where they differ between *Vardekloeftia* and other taxa. Although I accept the interpretation of *Cycadeoidea* and *Williamsonia* by Rothwell and Stockey (2002) and Stockey and Rothwell (2003), I find the interpretation of *Vardekloeftia* by Pedersen et al. (1989b) more convincing than Rothwell and Stockey's (2002) critique of it, and it seems premature to assume which set of characters is ancestral. In any case, this is not a critical issue, because the two scorings of Bennettitales had no effect on inferred relationships.

Of the two types of trees found in the constrained analysis (Figs. 9, 10), trees from island 1 are more plausible in terms of the stratigraphic distribution of taxa and morphotypes. Island 2 (Fig. 10) implies that a line consisting of some of the youngest taxa of seed plants—Bennettitales and *Caytonia*, both unknown before the Late Triassic, and angiosperms—diverged at the same time as a line with members extending back to the Late Carboniferous (cordaites, conifers, probably cycads). This implies a long ghost lineage for the former line (where its existence is predicted by the tree but not attested in the fossil record: cf. Doyle 1998b). Furthermore, it implies that the first members of both lines had pinnately veined simple leaves, while the fernlike leaves of *Autunia*, *Peltaspermum*, corystosperms, and (in some trees) *Callistophyton* were secondarily compound (Fig. 10). In fact, compound leaves predominated in the Carboniferous, while simple pinnate leaves did not appear until the latest Carboniferous (*Taeniopteris*) and (with the notable exception of Permian glossopterids) remained subordinate until the Mesozoic. In contrast, trees in island 1 (Fig. 9) place older groups (including glossopterids) near the base of both lines and Mesozoic groups in more nested positions, and they allow pinnately compound leaves to be interpreted as primitive in all taxa where they occur (except *Caytonia*). Therefore I will use a tree from island 1 (Fig. 9) as a basis for discussion of evolutionary implications.

Implications for the evolution of ovulate structures can be introduced in terms of the ovule position character (Fig. 13), with apical, abaxial, adaxial, and marginal states. At the base are Paleozoic seed ferns—first *Elkinia* and *Lyginopteris* with cupules of the dichotomous type, then medullosans, with no cupule, in all of which the ovules appear to be apical. Seed ferns with abaxial ovules—*Callistophyton* (the most plesiomorphic example of this type), peltasperms, and corystosperms—form a grade on the line between cycads (in which ovules are basically marginal: Norstog and Nicholls 1997) and coniferophytes, except in one tree in which these taxa are linked with cordaites. Coniferophytes (including Gnetales), whose more plesiomorphic members (cordaites, Ginkgoales, and Paleozoic conifers such as *Emporia*) had ovules that were apparently apical on simple sporophylls, are nested in this clade. Their simple sporophyll morphology is presumably a consequence of a general shift from fernlike fronds to simple leaves, ascribed by Rothwell (1982) to heterochronic substitution of cataphylls for fronds. The glossophyte line includes all taxa with adaxial ovules, with glossopterids and *Pentoxylon* at the base, and with *Caytonia* linked with angiosperms. Because lines with adaxial, abaxial, and marginal ovules diverge from adjacent nodes, the most parsimonious ancestral state in platysperms is equivocal, and it is not clear which of these states are synapomorphies. One alternative is that all three states were separately derived from apical and are therefore synapomorphies of their respective clades.

Whereas most versions of the anthophyte hypothesis implied that the ''flowers'' of Gnetales were reduced from more complex structures (Crane 1985, Doyle and Donoghue 1986, Doyle 1994), the molecular results support the hypothesis that they are homologous with the axillary fertile short shoots of coniferophytes, best seen in cordaites and Paleozoic conifers. This view was proposed by Eames (1951) for *Ephedra* but later extended to the other genera (Bierhorst 1971, Doyle 1978), and it was discussed by Doyle (1994) in relation to trees in which anthophytes were nested in coniferophytes (e.g., Nixon et al. 1994). Shindo et al. (1999) argued that it was supported by developmental genetic data. The position of Gnetales within conifers, linked with Pinaceae, implies that the female

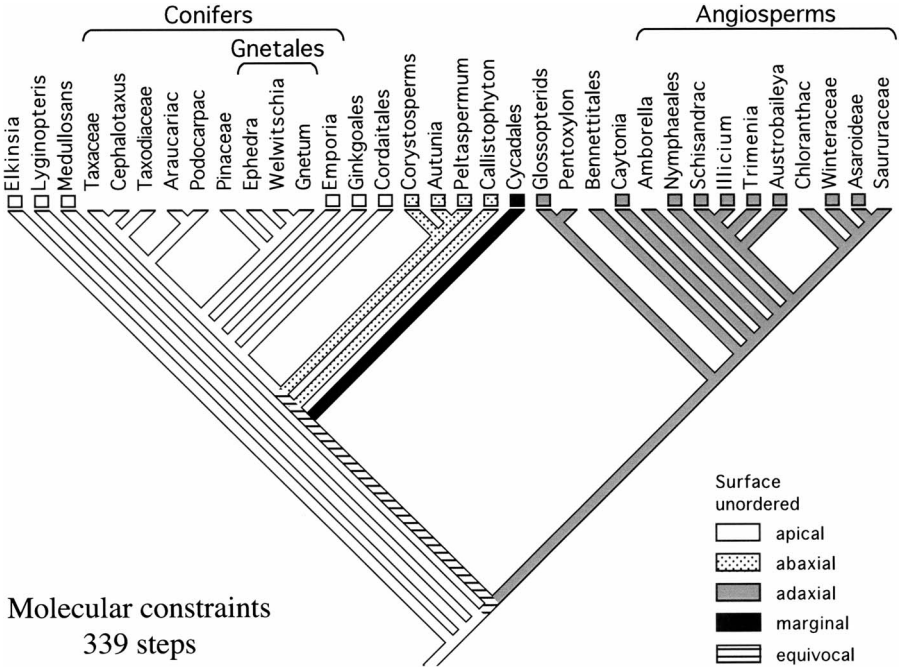

FIG. 13. Representative tree from the analysis with molecular constraints (Fig. 9), with shading showing the inferred evolution of ovule position (76).

fertile short shoot, which corresponds to the cone scale of living conifers (Florin 1951), was either transformed into a woody cone scale twice, in Pinaceae and in other conifers, or that the cone scale reverted to a shoot with scale-like appendages in Gnetales, a less plausible scenario. In either case, the constrained trees imply that the fertile short shoot, which had become dorsiventral in the common ancestor of *Emporia* and crown-group conifers, reverted to bisymmetric in Gnetales. In addition, there was a shift from simple to compound male strobili, perhaps by remodeling of the male structures on the female plan. Future studies may clarify whether the compound male strobili of the Paleozoic conifer *Thucydia* (Hernandez-Castillo et al. 2001) are relevant to this question. There is no character in the present data set that unequivocally links Pinaceae and Gnetales, although double fertilization of the *Ephedra* type, which has been reported in some Pinaceae but is not confirmed for the whole family (which was therefore scored as 0/1), could be such a synapomorphy (Friedman and Floyd 2001).

It may be significant that anthophyte trees implied that the inferred ancestral megasporophyll and cupule were completely lost by reduction in Gnetales (Doyle and Donoghue 1986, Doyle 1994, 1996). In hindsight the absence of these structures was a danger signal suggesting that Gnetales belonged elsewhere.

If a leaf-cupule complex of the glossopterid type existed on the line leading to angiosperms, their bitegmic ovule could be derived by enrolling of the cupule and reduction of the number of ovules on its adaxial surface to one, and the carpel wall could be derived from the subtending leaf (Stebbins 1974, Retallack and Dilcher 1981, Doyle 1996). These homologies are diagrammed in Fig. 14, with the abaxial surface of foliar structures indicated in black. In glossopterids, I have illustrated a unicupulate leaf-cupule complex (Fig. 14a) and two interpretations of the multicupulate type (e.g., *Lidgettonia*: Surange and Chandra 1975, Schopf 1976, Retallack & Dilcher 1981; Fig. 2J–L). In Fig. 14b the cupules are interpreted as leaflets of a single compound sporophyll, in Fig. 14c as several simple sporophylls. For angiosperms, I have shown an ascidiate carpel with one ovule (Fig. 14d), a common type in basal angiosperms (Endress and Igersheim 2000), and a classic plicate carpel with several ovules (Fig. 14e). The parallels between the unicupulate glossopterid type (Fig. 14a) and the ascidiate carpel (Fig. 14d) are especially close: the bitegmic ovule develops from

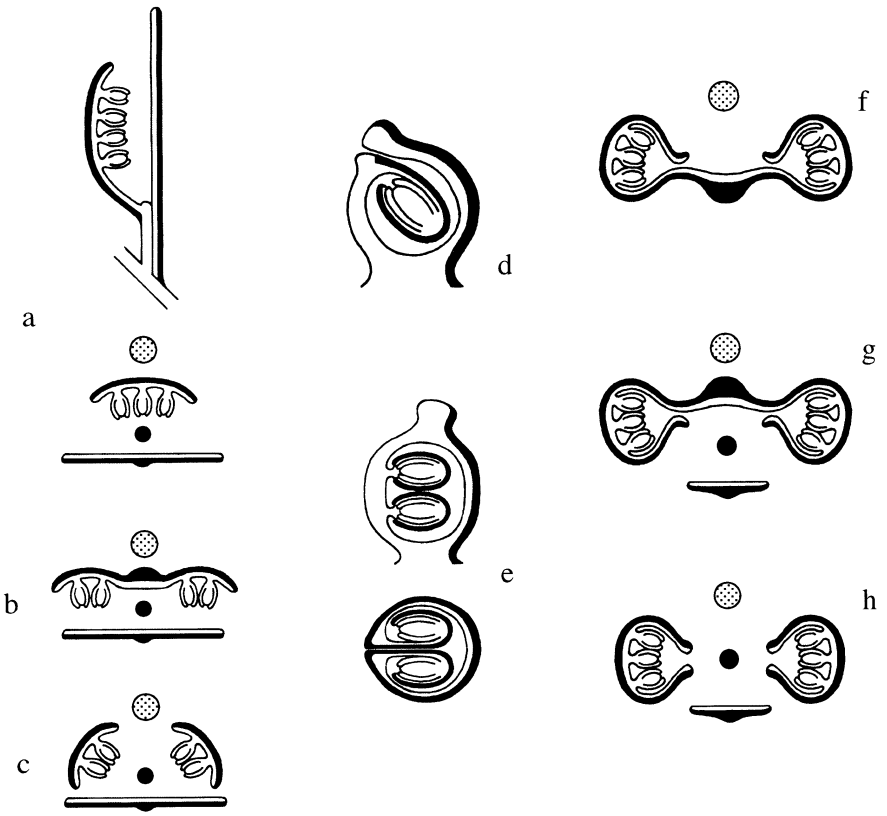

FIG. 14. Alternative interpretations and possible homologies of ovulate structures in glossopterids (a–c), angiosperms (d, e), and *Caytonia* (f–h), with abaxial surfaces indicated in black. See text for discussion.

the cross-zone on the ventral side of the carpel primordium, where one might expect an axillary branch, and the ovule has the proper orientation, allowing for tilting toward the carpel midrib. The positional relationships are less clearly comparable in the plicate carpel. However, the precise geometry may not be critical, considering the great flexibility in placentation within angiosperms.

The hypothesis that the carpel was derived from a glossopterid leaf-cupule complex is consistent with a widespread view among developmental geneticists (e.g., Skinner et al. 2004), based on mutants and gene expression patterns, that the placenta and carpel wall are distinct structures: the carpel wall corresponds to the leaf, the placenta to the axillary fertile branch. This also recalls the ''gonophyll theory'' of Melville (1963), but without his reliance on now-refuted reconstructions of glossopterids and his concept that angiosperm gynoecia were derived polyphyletically from glossopterid structures.

A weakness of this scheme is uncertainty in reconstructing characters of the first glossophytes. The present trees imply that several features of glossopterids are derived: uniseriate rays, unfused microsporangia, and paddle-like megasporophylls in both glossopterids and *Pentoxylon*, plus reticulate venation, air sacs and striations on the pollen, and loss of the sarcotesta in glossopterids alone. However, scenarios in which many of these features are ancestral in glossophytes are only one step less parsimonious (discussed below for reticulate venation). Uniseriate rays, an important aspect of the pycnoxylic wood syndrome, are suspiciously correlated with the cool temperate distribution of glossopterids and *Pentoxylon* (as well as conifers, ginkgos, and corystosperms). The inference that paddle-like megasporophylls were derived may be an artifact of the present character definition if they were homologous with the cupules of *Caytonia* and the bitegmic ovules of angiosperms. Alternatively, if multicupulate leaf-cupule complexes were ancestral in glossopterids and the ovule-bearing portion was a compound megasporo-

phyll (Fig. 14b), glossopterids would have the same state as *Caytonia*.

The concept that the leaf-cupule complex was ancestral in glossophytes is consistent with the age of glossopterids, and it would be enhanced if they were shown to be paraphyletic. Many authors have expressed suspicions that glossopterids were heterogeneous, noting for example the contrast between unicupulate types, in which the cupule was quite leaflike, and multicupulate types, in which the cupules were more modified and contained fewer ovules (cf. Pigg and Trivett 1994, Taylor 1996). There is little reason to believe that glossopterids were polyphyletic, since the different conditions could be a result of variation in the number of sporophylls per branch and the number of ovules per sporophyll, rather than fundamentally different starting points. It may be easier to imagine that some traditional glossopterids were more closely related to Mesozoic taxa than others, making the group paraphyletic. However, to demonstrate this would require showing that the apparent synapomorphies of glossopterids were outweighed by synapomorphies of some glossopterids and Mesozoic taxa.

The present scheme also requires that something like the leaf-cupule complex persisted on the line between glossopterids and angiosperms, but other taxa attached to this line show no sign of such a structure, with the possible exception of the cupule in the bennettitalian genus *Vardekloeftia* (Harris 1932, 1954, Pedersen et al. 1989b). Bennettitales had stalked ovules borne on a radial receptacle, while *Pentoxylon* had ovules borne on all sides of a structure that was originally assumed to be radial but was shown by Rothwell and Serbet (1994) to have a bilateral, leaflike anatomy. This may not be a problem if these conditions were autapomorphic specializations of structures of a glossopterid type, which I allowed by scoring Bennettitales as unknown for megasporophyll morphology and ovule position. The most common interpretation is that each stalked ovule of Bennettitales was a highly reduced sporophyll (Crane 1985, Rothwell and Stockey 2002). A more exotic alternative is that the receptacle was a sporophyll shifted to a terminal position and radialized (Doyle and Donoghue 1986, Doyle 1996), on analogy with *Pentoxylon*. However, both hypotheses imply that the ancestral leaf-axillary branch organization was lost, whether by reduction, fusion, or heterotopic transfer of the sporophyll to an axis of a lower order.

This problem would be less severe if Bennettitales and *Pentoxylon* formed a clade, as in Crane (1985), putting both deviant taxa on a sideline. This relationship is only one step less parsimonious with no other constraints, but three steps worse with molecular backbone constraints. The problem would disappear if it was shown that Bennettitales were not related to glossophytes but to some other group, such as cycads, a relationship that is two steps less parsimonious with no other constraints, three steps worse with the molecular backbone. Perhaps the situation is analogous to that of Gnetales, where lack of any vestige of the ancestral megasporophyll or cupule now appears to be evidence that Gnetales did not belong in the anthophytes. Discovery of more plesiomorphic relatives of Bennettitales or *Pentoxylon* could show either conditions more compatible with the glossopterid type or something different, strengthening or refuting the present scheme.

Caytonia fits better between glossopterids and angiosperms: it had cupules that correspond to the predicted intermediate, in containing several ovules but being anatropous, plus angiosperm-like advances in its seeds (no pollen chamber, thick nucellar cuticle, loss of the megaspore membrane). However, other aspects of its morphology are hard to explain in glossopterid terms. Several interpretations are possible, each of which raises new questions (Fig. 14f–h). Was the ovulate structure a compound leaf, with a rachis and leaflets converted into cupules, as believed by Harris (1940, 1951) and Reymanówna (1974) and assumed here in scoring *Caytonia* as having pinnate megasporophylls? If so, was it borne directly on a main stem (Fig. 14f), with the adaxially enrolled cupules facing upward? This would be difficult to reconcile with a glossopterid prototype, except by postulating that the sporophyll was transferred from the axillary branch to a stem of a lower order. Such a sporophyll might be transformed into an angiosperm carpel by expansion of the rachis (Gaussen 1946, Stebbins 1974, Doyle 1978; Fig. 1), an alternative to the proposed homologies with glossopterids. Or was the ovulate structure a compound leaf borne on an axillary shoot (Fig. 14g)? Such a system could be compared with a *Lidgettonia* leaf-cupule complex as interpreted in Fig. 14b, where the cupules correspond to leaflets. This would predict that *Caytonia* cupules faced downward, toward a subtending leaf or bract. Or, despite its dorsiventral appearance, was the *Caytonia* ovulate structure actually an

axillary branch bearing several simple sporophylls (Fig. 14h)? This would correspond to *Lidgettonia* as reconstructed in Fig. 14c, where each cupule is a simple sporophyll. In either of the latter schemes (Fig. 14g, h), was the subtending leaf distinct, or was it fused to the cupule-bearing axis, so that what appears to be a rachis was actually a composite structure?

Specimens that show the ovulate structures of *Caytonia* attached to a stem are needed to decide among these alternatives, although they might not be easy to interpret. Retallack and Dilcher (1988) reconstructed the cupules as facing downward, based on a sporophyll apparently attached to a stem in a specimen at Cambridge University, with no sign of a subtending leaf or bract. They interpreted this orientation as evidence that the ovules were on the abaxial surface of the cupules, although it might be consistent with Fig. 14g, in which the ovules are adaxial, if the bract was highly reduced or fused to the rachis. However, after examining this specimen I am not convinced that the relative orientation of the parts can be determined.

If nothing comparable to a glossopterid leaf-cupule complex can be found in the Mesozoic taxa associated here with glossopterids and angiosperms, it could mean that this structure was an autapomorphy of glossopterids that never existed on the line leading to angiosperms, thus refuting the proposed homologies. Or it could suggest an exclusive link between angiosperms and glossopterids. Answers to these questions, which might come from better information on the anatomy of glossopterids and Mesozoic fossils, could have a major impact on the angiosperm question.

Origin of the angiosperm stamen in terms of potential outgroups is less widely discussed, but it poses as many problems as origin of the carpel. Male structures of glossopterids consisted of a branched sporangium-bearing unit adnate to the adaxial side of a leaf (Surange and Maheshwari 1970, Schopf 1976, Gould and Delevoryas 1977, Retallack & Dilcher 1981), reminiscent of the leaf-cupule complex. Bennettitales had what appear to be sporophylls bearing microsynangia on their adaxial side, but it is worth considering that these were derived from compound structures of the glossopterid type. Whatever the origin of these structures, extreme reduction in the number of sporangia in either group might result in something like the stamens of basal angiosperms, which have two pairs of microsporangia borne on their adaxial side (Doyle and Endress 2000). Each pair of microsporangia would represent a separate synangium. As with the female structures, it is unclear how glossopterid and bennettitalian organs relate to the branched microsporophylls of *Pentoxylon* and *Caytonia*.

Another aspect of the angiosperm problem is origin of the angiosperm leaf. There is a wide morphological gap between the simple leaves of extant basal angiosperms, with pinnate secondary veins and reticulate fine venation, and Paleozoic seed ferns, which were pinnately compound and had dissected pinnules with open dichotomous venation. Leaves of the glossopterid type could fill part of this gap: not only were they already simple, but they also had reticulate laminar venation. The main difference is that the reticulum was simple, consisting of veins of only one order, as opposed to complex in angiosperms, consisting of several orders, the finest of which are freely ending veinlets. Elaborating on ideas of Stebbins (1974), Doyle and Hickey (1976) proposed that transformation of a seed fern frond into an angiosperm leaf involved radical reduction in a semiarid environment, but this is unnecessary under the present scheme, since glossopterid leaves were simple but not highly reduced. Origin of the angiosperm leaf could instead involve origin of a hierarchy of coarse to fine veins without any major change in size, which might reflect a shift in the type of meristematic activity responsible for production of the blade, from marginal to diffuse (Boyce 2005). This would be consistent with arguments of Feild et al. (2004) that the first angiosperms were adapted to disturbed habitats in the wet forest understory.

A problem for this scenario is the fact that taxa without reticulate venation, namely *Pentoxylon* and Bennettiales, are interpolated between glossopterids and angiosperms, although glossopterid-like venation does occur in the palmately compound leaves of *Caytonia*. As a result, although simple leaves with pinnate venation (like the *Taeniopteris*-type leaves of *Pentoxylon* and some Bennettitales) are reconstructed at the base of glossophytes, it is most parsimonious to assume that reticulate venation originated independently in glossopterids and the *Caytonia*-angiosperm clade. The alternative, that it originated once but was lost in *Pentoxylon* and Bennettiales, is one step less parsimonious. This picture would change if *Pentoxylon* and Bennettiales formed a clade, which would require only one reversal from reticulate to open venation. Also, some Bennettitales had

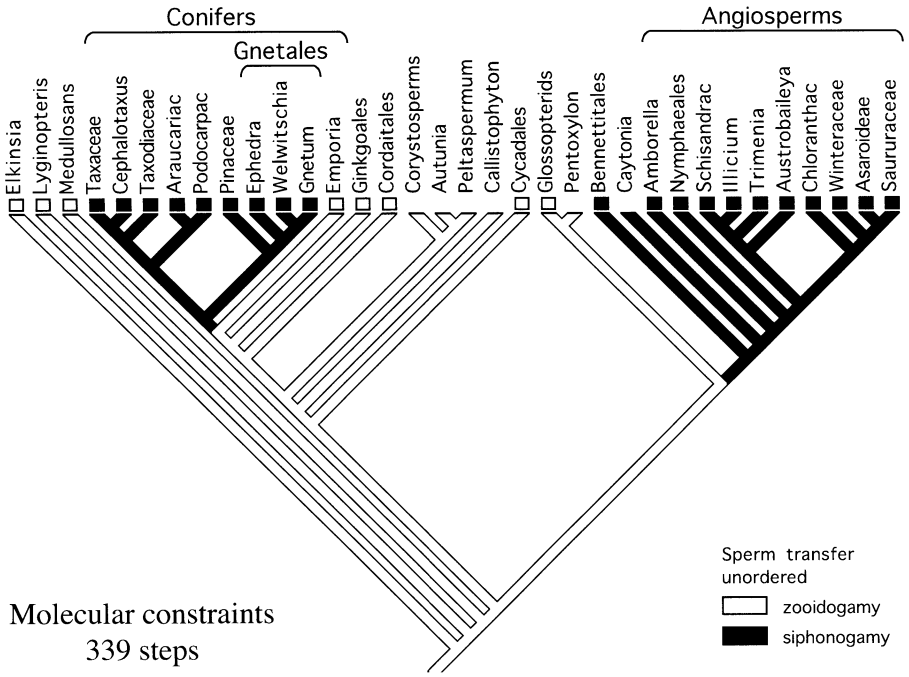

FIG. 15. Representative tree from the analysis with molecular constraints (Fig. 9), with shading showing the inferred evolution of sperm transfer (character 73).

reticulate venation (*Dictyozamites*); if these were shown to be plesiomorphic in the group, the hypothesis that reticulate venation was homologous in glossopterids, *Caytonia*, and angiosperms would be strengthened.

These observations bring out a general problem for the present scheme, the stratigraphic gap in the record of plants with glossopterid-like features after the mass extinction at the end of the Permian (Retallack 1995). The scheme predicts that some derivatives or relatives of glossopterids, with or without additional advances, survived into the Mesozoic. This picture could change with the discovery of new Mesozoic fossils or association of known but isolated organs. A possible example is *Mexiglossa*, a *Glossopteris*-like leaf from the Jurassic of Mexico (Delevoryas and Person 1975), which co-occurs with branched microsporophylls (*Perezlaria*) suggestive of the male structures of *Caytonia* (Delevoryas and Gould 1971). In general, if the plants associated as glossophytes do form a clade, they are probably not its only members. This is suggested by *Petriellaea* (Taylor et al. 1994), which also had cupules with adaxial ovules. Anderson and Anderson (2003) have described a remarkable array of plants from the Triassic Molteno flora of South Africa, some with anatropous cupules, that might also belong here.

The present results also provide a new perspective on the origin of siphonogamy, a feature of angiosperms and Gnetales that seemed to be an anthophyte synapomorphy and was more recently proposed as a synapomorphy of angiosperms, conifers, and Gnetales (Crane et al. 2004). Stockey and Rothwell (2003) showed that it probably existed in Bennettitales. A major new element is the report by Nishida et al. (2003, 2004) that glossopterids had motile sperm. This cannot be taken as evidence that glossopterids are not related to angiosperms. The molecular arrangement of living taxa (Fig. 5), where cycads and *Ginkgo*, with motile sperm, are attached between conifers and angiosperms, implies that siphonogamy arose independently in conifers (including Gnetales) and on the line leading to angiosperms. Considering living taxa alone, its origin on the angiosperm line could have occurred at any time between the Carboniferous and the Cretaceous. In terms of the tree of fossil and living taxa (Fig. 15), the discovery of motile sperm in glossopterids implies that siphonogamy arose between glossopterids and Bennettitales. This tree also predicts that *Pentoxylon* had motile sperm; discovery

that *Pentoxylon* too was siphonogamous might be evidence that it was closer to Bennettitales and angiosperms. Such examples illustrate not only the uncertainties caused by incomplete preservation, but also the potential impact of new data on previously unknown characters.

Of course, there are other fossils that might invalidate this scheme, such as Permian gigantopterids (cf. Taylor and Li 1997), which had even more angiosperm-like leaf venation but are too incompletely known to be included in an analysis. What is needed to test these hypotheses is better understanding of Permian and Mesozoic seed plant diversity and the morphology of fossil taxa that are already known. Examples include information on the nodal anatomy of glossopterids and *Caytonia* (two-trace unilacunar in *Pentoxylon* and basal angiosperms); wood anatomy, sporophyll attachment and associated structures in *Caytonia*; seed cuticle characters based on coordinated observations on both petrified and compressed material; details of the life cycle in Mesozoic fossil taxa; and recognition of more plesiomorphic relatives of Bennettitales, *Pentoxylon*, and *Caytonia*.

Literature Cited

ADENDORFF, R. 2005. A revision of the ovulate fructifications of *Glossopteris* from the Permian of South Africa. Ph.D. thesis. Univ. of the Witwatersrand, Johannesburg, South Africa.

ALBERT, V. A., A. BACKLUND, K. BREMER, M. W. CHASE, J. R. MANHART, B. D. MISHLER, AND K. C. NIXON. 1994. Functional constraints and *rbc*L evidence for land plant phylogeny. Ann. Mo. Bot. Gard. 81: 534–567.

ANDERSON, J. M. AND H. M. ANDERSON. 2003. Heyday of the gymnosperms: systematics and biodiversity of the Late Triassic Molteno fructifications. National Botanical Inst., Pretoria, South Africa.

ARBER, E. A. N. AND J. PARKIN. 1907. On the origin of angiosperms. J. Linn. Soc. Bot. 38: 29–80.

AXSMITH, B. J., E. L. TAYLOR AND T. N. TAYLOR. 1998. The limitations of molecular systematics: a palaeobotanical perspective. Taxon 47: 105–108.

AXSMITH, B. J., E. L. TAYLOR, T. N. TAYLOR, AND N. R. CUNEO. 2000. New perspectives on the Mesozoic seed fern order Corystospermales based on attached organs from the Triassic of Antarctica. Am. J. Bot. 87: 757–768.

BAILEY, I. W. 1944. The development of vessels in angiosperms and its significance in morphological research. Am. J. Bot. 31: 421–428.

BAILEY, I. W. 1949. Origin of the angiosperms: need for a broadened outlook. J. Arnold Arbor. Harv. Univ. 30: 64–70.

BAILEY, I. W. AND B. G. L. SWAMY. 1951. The conduplicate carpel of dicotyledons and its initial trends of specialization. Am. J. Bot. 38: 373–379.

BALASUBRAMANIAN, S. AND K. SCHNEITZ. 2000. *NOZZLE* regulates proximal-distal pattern formation, cell proliferation and early sporogenesis during ovule development in *Arabidopsis thaliana*. Development 127: 4227–4238.

BALASUBRAMANIAN, S. AND K. SCHNEITZ. 2002. *NOZZLE* links proximal-distal and adaxial-abaxial pattern formation during ovule development in *Arabidopsis thaliana*. Development 129: 4291–4300.

BARBACKA, M. AND K. BÓKA. 2000. The stomatal ontogeny and structure of the Liassic pteridosperm *Sagenopteris* (Caytoniales) from Hungary. Int. J. Plant Sci. 161: 149–157.

BARKMAN, T. J., G. CHENERY, J. R. MCNEAL, J. LYONS-WEILER, W. J. ELLISENS, G. MOORE, A. D. WOLFE, AND C. W. DEPAMPHILIS. 2000. Independent and combined analyses of sequences from all three genomic compartments converge on the root of flowering plant phylogeny. Proc. Natl. Acad. Sci. USA 97: 13166–13171.

BAUCH, J., W. LIESE, AND R. SCHULTZE. 1972. The morphological variability of the bordered pit membranes in gymnosperms. Wood Sci. Technol. 6: 165–184.

BENSON, M. 1908. On the contents of the pollen chamber of a specimen of *Lagenostoma ovoides*. Bot. Gaz. 45: 409–412.

BIERHORST, D. W. 1971. Morphology of vascular plants. Macmillan, New York, NY.

BOSE, M. N., P. K. PAL, AND T. M. HARRIS. 1985. The *Pentoxylon* plant. Philos. Trans. Roy. Soc. London Ser. B 310: 77–108.

BOWE, L. M., G. COAT, AND C. W. DEPAMPHILIS. 2000. Phylogeny of seed plants based on all three genomic compartments: extant gymnosperms are monophyletic and Gnetales' closest relatives are conifers. Proc. Natl. Acad. Sci. USA 97: 4092–4097.

BOWMAN, J. L. 2000. The *YABBY* gene family and abaxial cell fate. Curr. Opin. Plant Biol. 3: 17–22.

BOYCE, C. K. 2005. Patterns of segregation and convergence in the evolution of fern and seed plant leaf morphologies. Paleobiology 31: 117–140.

BREMER, K. 1988. The limits of amino acid sequence data in angiosperm phylogenetic reconstruction. Evolution 42: 795–803.

BURLEIGH, J. G. AND S. MATHEWS. 2004. Phylogenetic signal in nucleotide data from seed plants: implications for resolving the seed plant tree of life. Am. J. Bot. 91: 1599–1613.

CARLQUIST, S. 1993. Wood and bark anatomy of Aristolochiaceae; systematic and habital correlations. IAWA J. 14: 341–357.

CARLQUIST, S. 1994. Wood and bark anatomy of *Gnetum gnemon* L. Bot. J. Linn. Soc. 116: 203–221.

CARLQUIST, S. 1996a. Wood anatomy of primitive angiosperms: new perspectives and syntheses, pp. 68–90. *In* D. W. Taylor and L. J. Hickey [eds.], Flowering plant origin, evolution & phylogeny. Chapman & Hall, New York, NY.

CARLQUIST, S. 1996b. Wood, bark, and stem anatomy of Gnetales: a summary. Int. J. Plant Sci. 157(6 Suppl.): S58–S76.

CARLQUIST, S. 1996c. Wood, bark and stem anatomy of New World species of *Gnetum*. Bot. J. Linn. Soc. 120: 1–19.

CARLQUIST, S. 1996d. Wood and bark anatomy of lianoid Indomalesian and Asiatic species of *Gnetum*. Bot. J. Linn. Soc. 121: 1–24.

CARLQUIST, S. 1999. Wood and bark anatomy of Schisandraceae: implications for phylogeny, habit, and vessel evolution. Aliso 18: 45–55.

CARLQUIST, S. 2001. Observations on the vegetative anatomy of *Austrobaileya*: habital, organographic and phylogenetic conclusions. Bot. J. Linn. Soc. 135: 1–11.

CARLQUIST, S. AND A. A. ROBINSON. 1995. Wood and bark anatomy of the African species of *Gnetum*. Bot. J. Linn. Soc. 118: 123–137.

CHAMBERLAIN, C. J. 1935. Gymnosperms: structure and evolution. Univ. Chicago Press, Chicago, IL (reprinted by Dover, New York, NY).

CHAW, S.-M., A. ZHARKIKH, H.-M. SUNG, T.-C. LAU, AND W.-H. LI. 1997. Molecular phylogeny of extant gymnosperms and seed plant evolution: analysis of nuclear 18S rRNA sequences. Mol. Biol. Evol. 14: 56–68.

CHAW, S.-M., C. L. PARKINSON, Y. CHENG, T. M. VINCENT, AND J. D. PALMER. 2000. Seed plant phylogeny inferred from all three plant genomes: monophyly of extant gymnosperms and origin of Gnetales from conifers. Proc. Natl. Acad. Sci. USA 97: 4086–4091.

CHLONOVA, A. F. AND T. D. SUROVA. 1988. Pollen wall ultrastructure of *Clavatipollenites incisus* Chlonova and two modern species of *Ascarina* (Chloranthaceae). Pollen Spores 30: 29–44.

CONRAN, J. G., G. A. WOOD, P. G. MARTIN, J. M. DOWD, C. J. QUINN, P. A. GADEK, AND R. A. PRICE. 2001. Generic relationships within and between the gymnosperm families Podocarpaceae and Phyllocladaceae based on an analysis of the CP gene *rbc*L. Aust. J. Bot. 48: 715–724.

CORNER, E. J. H. 1976. The seeds of the dicotyledons. Cambridge Univ. Press, Cambridge, UK.

CRANE, P. R. 1985. Phylogenetic analysis of seed plants and the origin of angiosperms. Ann. Mo. Bot. Gard. 72: 716–793.

CRANE, P. R. 1996. The fossil history of the Gnetales. Int. J. Plant Sci. 157(6 Suppl.): S50–S57.

CRANE, P. R., P. HERENDEEN, AND E. M. FRIIS. 2004. Fossils and plant phylogeny. Am. J. Bot. 91: 1683–1699.

CRONQUIST, A. 1968. The evolution and classification of flowering plants. Houghton Mifflin, Boston, MA.

CRONQUIST, A. 1988. The evolution and classification of flowering plants, second edition. New York Botanical Garden, Bronx, NY.

D'ARCY, W. G. 1996. Anthers and stamens and what they do, pp. 1–24. *In* W. G. D'Arcy and R. C. Keating [eds.], The anther: form, function and phylogeny. Cambridge Univ. Press, Cambridge, UK.

DELEVORYAS, T. AND R. E. GOULD. 1971. An unusual fossil fructification from the Jurassic of Oaxaca, Mexico. Am. J. Bot. 58: 616–620.

DELEVORYAS, T. AND C. P. PERSON. 1975. *Mexiglossa varia* gen. et sp. nov., a new genus of glossopteroid leaves from the Middle Jurassic of Oaxaca, Mexico. Palaeontographica Abt. B 154: 114–120.

DEN OUTER, R. W. 1967. Histological investigations of the secondary phloem in gymnosperms. Meded. Landbouwhogesch. Wageningen 67(7): 1–119.

DICKISON, W. C. 1992. Morphology and anatomy of the flower and pollen of *Saruma henryi* Oliv., a phylogenetic relict of the Aristolochiaceae. Bull. Torrey Bot. Club 119: 392–400.

DONOGHUE, M. J. AND J. A. DOYLE. 2000. Seed plant phylogeny: demise of the anthophyte hypothesis? Curr. Biol. 10: R106–R109.

DONOGHUE, M. J. J. A. DOYLE, J. GAUTHIER, A. G. KLUGE, AND T. ROWE. 1989. The importance of fossils in phylogeny reconstruction. Annu. Rev. Ecol. Syst. 20: 431–460.

DONOGHUE, M. J., R. G. OLMSTEAD, J. F. SMITH, AND J. D. PALMER. 1992. Phylogenetic relationships of Dipsacales based on *rbc*L sequences. Ann. Mo. Bot. Gard. 79: 333–345.

DOYLE, J. 1963. Proembryogeny in *Pinus* in relation to that in other conifers—a survey. Proc. Roy. Irish Acad. Sect. B, 62: 181–216.

DOYLE, J. A. 1978. Origin of angiosperms. Annu. Rev. Ecol. Syst. 9: 365–392.

DOYLE, J. A. 1994. Origin of the angiosperm flower: a phylogenetic perspective. Plant Syst. Evol. Suppl. 8: 7–29.

DOYLE, J. A. 1996. Seed plant phylogeny and the relationships of Gnetales. Int. J. Plant Sci. 157(6, Suppl.): S3–S39.

DOYLE, J. A. 1998a. Phylogeny of vascular plants. Annu. Rev. Ecol. Syst. 29: 567–599.

DOYLE, J. A. 1998b. Molecules, morphology, fossils, and the relationship of angiosperms and Gnetales. Mol. Phylog. Evol. 9: 448–462.

DOYLE, J. A. 2001. Significance of molecular phylogenetic analyses for paleobotanical investigations on the origin of angiosperms. Palaeobotanist 50: 167–188.

DOYLE, J. A. AND M. J. DONOGHUE. 1986. Seed plant phylogeny and the origin of angiosperms: an experimental cladistic approach. Bot. Rev. 52: 321–431.

DOYLE, J. A. AND M. J. DONOGHUE. 1987. The importance of fossils in elucidating seed plant phylogeny and macroevolution. Rev. Palaeobot. Palynol. 50: 63–95.

DOYLE, J. A., M. J. DONOGHUE, AND E. A. ZIMMER. 1994. Integration of morphological and ribosomal RNA data on the origin of angiosperms. Ann. Mo. Bot. Gard. 81: 419–450.

DOYLE, J. A., H. EKLUND, AND P. S. HERENDEEN. 2003. Floral evolution in Chloranthaceae: implications of a morphological phylogenetic analysis. Int. J. Plant Sci. 164(5 Suppl.): S365–S382.

DOYLE, J. A. AND P. K. ENDRESS. 2000. Morphological phylogenetic analysis of basal angiosperms: comparison and combination with molecular data. Int. J. Plant Sci. 161(6 Suppl.): S121–S153.

DOYLE, J. A. AND L. J. HICKEY. 1976. Pollen and leaves from the mid-Cretaceous Potomac Group and their bearing on early angiosperm evolution, pp. 139–206. *In* C. B. Beck [ed.], Origin and early evolution of angiosperms. Columbia Univ. Press, NY.

EAMES, A. J. 1952. Relationships of the Ephedrales. Phytomorphology 2: 79–100.

EKLUND, H., J. A. DOYLE, AND P. S. HERENDEEN. 2004. Morphological phylogenetic analysis of living and fossil Chloranthaceae. Int. J. Plant Sci. 165: 107–151.

ENDRESS, P. K. 1980. The reproductive structures and systematic position of the Austrobaileyaceae. Bot. Jahrb. Syst. 101: 393–433.

ENDRESS, P. K. AND A. IGERSHEIM. 2000. Gynoecium structure and evolution in basal angiosperms. Int. J. Plant Sci. 161(6 Suppl.): S211–S223.

ESAU, K. 1969. The phloem. Encyclopedia of Plant Anatomy 5(2). Borntraeger, Berlin, Germany.

ESHED, Y., S. F. BAUM, J. V. PEREA, AND J. L. BOWMAN. 2001. Establishment of polarity in lateral organs of plants. Curr. Biol. 11: 1251–1260.

ESHED, Y., A. IZHAKI, S. F. BAUM, S. K. FLOYD, AND J. L. BOWMAN. 2004. Asymmetric leaf development and blade expansion in *Arabidopsis* are mediated by KANADI and YABBY activities. Development 131: 2997–3006.

FEILD, T. S., N. C. ARENS, J. A. DOYLE, T. E. DAWSON, AND M. J. DONOGHUE. 2004. Dark and disturbed: a new image of early angiosperm ecology. Paleobiology 30: 82–107.

FELSENSTEIN, J. 1985. Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39: 783–791.

FLORIN, R., 1951. Evolution in cordaites and conifers. Acta Horti Berg. 15: 285–388.

FRIEDMAN, W. E. 1992. Evidence of a pre-angiosperm origin of endosperm: implications for the evolution of flowering plants. Science 255: 336–339.

FRIEDMAN, W. E. 1994. The evolution of embryogeny in seed plants and the developmental origin and early history of endosperm. Am. J. Bot. 81: 1468–1486.

FRIEDMAN, W. E. AND J. S. CARMICHAEL. 1998. Heterochrony and developmental innovation: evolution of female gametophyte ontogeny in *Gnetum*, a highly apomorphic seed plant. Evolution 52: 1016–1030.

FRIEDMAN, W. E. AND S. K. FLOYD. 2001. The origin of flowering plants and their reproductive biology—a tale of two phylogenies. Evolution 55: 217–231.

FRIEDMAN, W. E., W. N. GALLUP, AND J. H. WILLIAMS. 2003. Female gametophyte development in *Kadsura*: implications for Schisandraceae, Austrobaileyales, and the early evolution of flowering plants. Int. J. Plant Sci. 164(5 Suppl.): S293–S305.

FRIEDMAN, W. E. AND J. H. WILLIAMS. 2003. Modularity in the angiosperm female gametophyte and its bearing on the early evolution of endosperm in flowering plants. Evolution 57: 216–230.

FRIEDMAN, W. E. AND J. H. WILLIAMS. 2004. Developmental evolution of the sexual process in ancient flowering plant lineages. Plant Cell (Suppl.) 16: S119–S132.

FROHLICH, M. W. 2003. An evolutionary scenario for the origin of flowers. Nature Rev. Genet. 4: 559–566.

FROHLICH, M. W. AND D. S. PARKER. 2000. The Mostly Male theory of flower evolutionary origins: from genes to fossils. Syst. Bot. 25: 155–170.

GABARAYEVA, N. I., V. V. GRIGORJEVA, AND J. R. ROWLEY. 2003. Sporoderm ontogeny in *Cabomba aquatica* (Cabombaceae). Rev. Palaeobot. Palynol. 127: 147–173.

GALTIER, J. 1988. Morphology and phylogenetic relationships of early pteridosperms, p. 135–176. *In* C. B. Beck [ed.], Origin and evolution of gymnosperms. Columbia Univ. Press, New York, NY.

GAUSSEN, H. 1946. Les Gymnospermes, actuelles et fossiles. Trav. Lab. For. Toulouse, Tome II Etud. Dendrol., sect. 1, vol. 1, Fasc. 3, ch. 5, 1–26.

GIBBS, R. D. 1957. The Mäule reaction, lignin, and the relationships between woody plants, pp. 269–312. *In* K. V. Thimann [ed.], The physiology of forest trees. Ronald Press, New York, NY.

GIFFORD, E. M. 1943. The structure and development of the shoot apex of *Ephedra altissima* Desf. Bull. Torrey Bot. Club 70: 15–25.

GIFFORD, E. M. 1950. The structure and development of the shoot apex in certain woody Ranales. Am. J. Bot. 37: 595–611.

GIFFORD, E. M. 1954. The shoot apex in angiosperms. Bot. Rev. 20: 477–529.

GOREMYKIN, V., V. BOBROVA, J. PAHNKE, A. TROITSKY, A. ANTONOV, AND W. MARTIN. 1996. Noncoding sequences from the slowly evolving chloroplast inverted repeat in addition to *rbc*L data do not support gnetalean affinities of angiosperms. Mol. Biol. Evol. 13: 383–396.

GOULD, R. E. AND T. DELEVORYAS. 1977. The biology of *Glossopteris*: evidence from petrified seed-bearing and pollen-bearing organs. Alcheringa 1: 387–399.

GRIFFITH, M. M. 1952. The structure and growth of the shoot apex in *Araucaria*. Am. J. Bot. 39: 253–263.

GROSS-HARDT, R., M. LENHARD, AND T. LAUX. 2002. *WUSCHEL* signaling functions in interregional communication during *Arabidopsis* ovule development. Genes Devel. 16: 1129–1138.

HAGEMANN, W. 1970. Studien zur Entwicklungsgeschichte der Angiospermenblätter. Bot. Jahrb. Syst. 90: 297–413.

HAMBY, R. K. AND E. A. ZIMMER. 1992. Ribosomal RNA as a phylogenetic tool in plant systematics, p. 50–91. *In* P. S. Soltis, D. E. Soltis, and J. J. Doyle [eds.], Molecular systematics of plants. Chapman & Hall, New York, NY.

HANSEN A., S. HANSMANN, T. SAMIGULLIN, A. ANTONOV, AND W. MARTIN. 1999. *Gnetum* and the angiosperms: molecular evidence that their shared morphological characters are convergent, rather than homologous. Mol. Biol. Evol. 16: 1006–1009.

HARRIS, T. M. 1932. The fossil flora of Scoresby Sound East Greenland. Part 3: Caytoniales and Bennettitales. Meddel. Grønland 85(5): 1–133.

HARRIS, T. M. 1940. *Caytonia*. Ann. Bot. N. S. 4: 713–734.

HARRIS, T. M. 1951. The relationships of the Caytoniales. Phytomorphology 1: 29–39.

HARRIS, T. M. 1954. Mesozoic seed cuticles. Svensk Bot. Tidskr. 48: 281–291.

HERNANDEZ-CASTILLO, G. R., G. W. ROTHWELL, AND G. MAPES. 2001. Thucydiaceae fam. nov., with a review and reevaluation of Paleozoic walchian conifers. Int. J. Plant Sci. 162: 1155–1185.

HESSE, M. 2001. Pollen characters of *Amborella trichopoda* (Amborellaceae): a reinvestigation. Int. J. Plant Sci. 162: 201–208.

HOLMES, W. B. K. 1974. On some fructifications of the Glossopteridales from the Upper Permian of New South Wales. Proc. Linn. Soc. N. S. Wales 98: 131–141.

JACKMAN, V. H. 1960. The shoot apex of some New Zealand gymnosperms. Phytomorphology 10: 145–157.

JEFFREY, E. C. AND R. E. TORREY. 1916. Ginkgo and

the microsporangial mechanisms of the seed plants. Bot. Gaz. 62: 281–292.

JOHNSON, M. A. 1950. Growth and development of the shoot of *Gnetum gnemon* L. I. The shoot apex and pith. Bull. Torrey Bot. Club 77: 354–367.

JOHNSON, M. A. 1951. The shoot apex in gymnosperms. Phytomorphology 1: 188–204.

KATO, M. 1990. *Ophioglossaceae*: a hypothetical archetype for the angiosperm carpel. Bot. J. Linn. Soc. 102: 303–311.

KEATING, R. C. 2000. Anatomy of the young vegetative shoot of *Takhtajania perrieri* (Winteraceae). Ann. Mo. Bot. Gard. 87: 335–346.

KENRICK, P. AND P. R. CRANE. 1997. The origin and early diversification of land plants: a cladistic study. Smithsonian Inst., Washington, DC.

KERP, J. H. F. 1988. Aspects of Permian palaeobotany and palynology. X. The West- and Central European species of the genus *Autunia* Krasser emend. Kerp (Peltaspermaceae) and the form-genus *Rhachiphyllum* Kerp (callipterid foliage). Rev. Palaeobot. Palynol. 54: 249–360.

KIDSTON, R. 1924. Fossil plants of the Carboniferous rocks of Great Britain. Mem. Geol. Surv. Gr. Brit. Palaeontol. 2: 379–522.

KIM, S., M.-J. YOO, V. A. ALBERT, J. S. FARRIS, P. S. SOLTIS, AND D. E. SOLTIS. 2004. Phylogeny and diversification of B-function MADS-box genes in angiosperms: evolutionary and functional implications of a 260-million-year-old duplication. Am. J. Bot. 91: 2102–2118.

KIRCHNER, M. 1992. Untersuchungen an einigen Gymnospermen der fränkischen Rhät-Lias-Grenzschichten. Palaeontographica Abt. B 224: 17–61.

KLAVINS, S. D., T. N. TAYLOR, AND E. L. TAYLOR. 2002. Anatomy of *Umkomasia* (Corystospermales) from the Triassic of Antarctica. Am. J. Bot. 89: 664–676.

KRASSILOV, V. A. 1986. New floral structure from the Lower Cretaceous of Lake Baikal area. Rev. Palaeobot. Palynol. 47: 9–16.

LES, D. H., E. L. SCHNEIDER, D. J. PADGETT, P. S. SOLTIS, D. E. SOLTIS, AND M. ZANIS. 1999. Phylogeny, classification and floral evolution of water lilies (Nymphaeaceae: Nymphaeales): a synthesis of non-molecular, *rbcL, matK,* and 18S rDNA data. Syst. Bot. 24: 28–46.

LONG, A. G. 1961. *Tristichia ovensi* gen. et sp. nov., a protostelic Lower Carboniferous pteridosperm from Berwickshire and East Lothian, with an account of some associated seeds and cupules. Trans. Roy. Soc. Edin. 64: 477–489.

LONG, A. G. 1966. Some Lower Carboniferous fructifications from Berwickshire, together with a theoretical account of the evolution of ovules, cupules and carpels. Trans. Roy. Soc. Edin. 66: 345–375.

LONG, A. G. 1979. Observations on the Lower Carboniferous genus *Pitus* Witham. Trans. Roy. Soc. Edin. 70: 111–127.

MADDISON, D. R. AND W. P. MADDISON. 2001. MacClade 4: analysis of phylogeny and character evolution, version 4.03. Sinauer, Sunderland, MA.

MADDISON, W. P. 1994. Missing data versus missing characters in phylogenetic analysis. Syst. Biol. 42: 576–581.

MAGALLÓN, S. AND M. J. SANDERSON. 2002. Relationships among seed plants inferred from highly conserved genes: sorting conflicting phylogenetic signals among ancient lineages. Am. J. Bot. 89: 1991–2006.

MAHESHWARI, P. 1950. An introduction to the embryology of angiosperms. McGraw-Hill, New York, NY.

MAPES, G. AND G. W. ROTHWELL. 1984. Permineralized ovulate cones of *Lebachia* from late Palaeozoic limestones of Kansas. Palaeontology 27: 69–94.

MARTENS, P. 1971. Les Gnétophytes. Encyclopedia of Plant Anatomy 12(2). Borntraeger, Stuttgart, Germany.

MCLOUGHLIN, S. 1990. Late Permian glossopterid fructifications from the Bowen and Sydney Basins, eastern Australia. Geobios 23: 283–297.

MEEUSE, A. D. J. AND F. BOUMAN. 1974. The inner integument—its probable origin and homology. Acta Bot. Neerl. 23: 237–249.

MEISTER, R. J., L. M. KOTOW, AND C. S. GASSER. 2002. SUPERMAN attenuates positive *INNER NO OUTER* autoregulation to maintain polar development of *Arabidopsis* ovule outer integuments. Development 129: 4281–4289.

MELVILLE, R. 1963. A new theory of the angiosperm flower: II. Kew Bull. 17: 1–63.

METCALFE, C. R. 1987. Anatomy of the dicotyledons, second edition, vol. III. Magnoliales, Illiciales, and Laurales (*sensu* Armen Takhtajan). Clarendon, Oxford, UK.

MEYEN, S. V. 1984. Basic features of gymnosperm systematics and phylogeny as evidenced by the fossil record. Bot. Rev. 50: 1–112.

MEYEN, S. V. 1987. Fundamentals of palaeobotany. Chapman & Hall, London, UK.

MEYER-BERTHAUD, B., T. N. TAYLOR AND E. L. TAYLOR. 1993. Petrified stems bearing *Dicroidium* leaves from the Triassic of Antarctica. Palaeontology 36: 337–356.

MEYLAN, B. A. AND B. G. BUTTERFIELD. 1978. The structure of New Zealand woods. New Zealand Department of Scientific and Industrial Research, Wellington.

MUHAMMAD, A. F. AND R. SATTLER. 1982. Vessel structure of *Gnetum* and the origin of angiosperms. Am. J. Bot. 69: 1004–1021.

NICKERSON, J. AND G. DROUIN. 2004. The sequence of the largest subunit of RNA polymerase II is a useful marker for inferring seed plant phylogeny. Mol. Phylog. Evol. 31: 403–415.

NISHIDA, H., K. B. PIGG, AND J. F. RIGBY. 2003. Swimming sperm in an extinct Gondwanan plant. Nature 422: 396–397.

NISHIDA, H., K. B. PIGG, K. KUDO, AND J. F. RIGBY. 2004. Zooidogamy in the Late Permian genus *Glossopteris*. J. Plant Res. 117: 323–328.

NIXON, K. C., W. L. CREPET, D. STEVENSON, AND E. M. FRIIS. 1994. A reevaluation of seed plant phylogeny. Ann. Mo. Bot. Gard. 81: 484–533.

NORSTOG, K. J. AND T. J. NICHOLLS. 1997. The biology of the cycads. Cornell Univ. Press, Ithaca, NY.

OSBORN, J. M. 2000. Pollen morphology and ultrastructure of gymnospermous anthophytes, p. 163–185. *In* M. M. Harley, C. M. Morton, and S. Blackmore [eds.], Pollen and spores: morphology and biology. Royal Botanic Gardens, Kew, UK.

OSBORN, J. M., T. N. TAYLOR, AND E. L. SCHNEIDER. 1991. Pollen morphology and ultrastructure of the

Cabombaceae: correlations with pollination biology. Am. J. Bot. 78: 1367–1378.

PALSER, B. F. 1975. The bases of angiosperm phylogeny: embryology. Ann. Mo. Bot. Gard. 62: 621–646.

PANT, D. D. AND R. S. SINGH. 1974. On the stem and attachment of *Glossopteris* and *Gangamopteris* leaves. Part II—Structural features. Palaeontographica Abt. B 147: 42–73.

PEDERSEN, K. R., P. R. CRANE, AND E. M. FRIIS. 1989a. Pollen organs and seeds with *Eucommiidites* pollen. Grana 28: 279–294.

PEDERSEN, K. R., P. R. CRANE, AND E. M. FRIIS. 1989b. The morphology and phylogenetic significance of *Vardekloeftia* Harris (Bennettitales). Rev. Palaeobot. Palynol. 60: 7–24.

PIGG, K. B. AND H. NISHIDA. 2006. The significance of silicified plant remains to the understanding of *Glossopteris*-bearing plants. J. Torrey Bot. Soc. 133: 46–61.

PIGG, K. B. AND M. L. TRIVETT. 1994. Evolution of the glossopterid gymnosperms from Permian Gondwana. J. Plant Res. 107: 461–477.

POORT, R. J., H. VISSCHER, AND D. L. DILCHER. 1996. Zoidogamy in fossil gymnosperms: the centenary of a concept, with special reference to prepollen of late Paleozoic conifers. Proc. Natl. Acad. Sci. USA 93: 11713–11717.

QIU, Y.-L., J. LEE, F. BERNASCONI-QUADRONI, D. E. SOLTIS, P. S. SOLTIS, M. ZANIS, E. A. ZIMMER, Z. CHEN, V. SAVOLAINEN, AND M. W. CHASE. 1999. The earliest angiosperms: evidence from mitochondrial, plastid and nuclear genomes. Nature 402: 404–407.

QUINN, C. J., R. A. PRICE, AND P. A. GADEK. 2002. Familial concepts and relationships of the conifers based on *rbcL* and *matK* sequence comparisons. Kew Bull. 57: 513–531.

RAMJI, M. V. 1961. Ontogenetic studies in *Sarcandra irvingbaileyi* Swamy. I. The structure of the shoot apex and ontogeny of leaf. Proc. Ind. Acad. Sci. Sect. B 53: 20–35.

RETALLACK, G. J. 1995. Permian-Triassic life crisis on land. Science 267: 77–80.

RETALLACK, G. J. AND D. L. DILCHER. 1981. Arguments for a glossopterid ancestry of angiosperms. Paleobiology 7: 54–67.

RETALLACK, G. J. AND D. L. DILCHER. 1988. Reconstruction of selected seed ferns. Ann. Mo. Bot. Gard. 75: 1010–1057.

REYMANÓWNA, M. 1974. On anatomy and morphology of *Caytonia*. Birbal Sahni Inst. Palaeobot. Spec. Publ. 2: 50–57.

ROBINSON-BEERS, K., R. E. PRUITT, AND C. S. GASSER. 1992. Ovule development in wild type *Arabidopsis* and two female-sterile mutants. Plant Cell 4: 1237–1249.

ROTHWELL, G. W. 1982. New interpretations of the earliest conifers. Rev. Palaeobot. Palynol. 37: 7–28.

ROTHWELL, G. W. AND R. SERBET. 1994. Lignophyte phylogeny and the evolution of spermatophytes: a numerical cladistic analysis. Syst. Bot. 19: 443–482.

ROTHWELL, G. W. AND R. A. STOCKEY. 2002. Anatomically preserved *Cycadeoidea* (Cycadeoidaceae), with a reevaluation of systematic characters for the seed cones of Bennettitales. Am. J. Bot. 89: 1447–1458.

ROTHWELL, G. W. AND S. WARNER. 1984. *Cordaixylon dumusum* n. sp. (Cordaitales). I. Vegetative structures. Bot. Gaz. 145: 275–291.

RYDIN, C., M. KÄLLERSJÖ, AND E. M. FRIIS. 2002. Seed plant relationships and the systematic position of Gnetales based on nuclear and chloroplast DNA: conflicting data, rooting problems, and the monophyly of conifers. Int. J. Plant Sci. 163: 197–214.

SAIKI, K. AND Y. YOSHIDA. 1999. A new Bennettitalean trunk with unilacunar five-trace nodal structure from the Upper Cretaceous of Hokkaido, Japan. Am. J. Bot. 86: 326–332.

SAMIGULLIN, T. K., W. F. MARTIN, A. V. TROITSKY, AND A. S. ANTONOV. 1999. Molecular data from the chloroplast *rpo*C1 gene suggest a deep and distinct dichotomy of contemporary spermatophytes into two monophyla: gymnosperms (including Gnetales) and angiosperms. J. Mol. Evol. 49: 310–315.

SAMPSON, F. B. 2000. Pollen diversity in some modern magnoliids. Int. J. Plant Sci. 161(6 Suppl.): S193–S210.

SANDERSON, M. J., M. F. WOJCIECHOWSKI, J.-M. HU, T. SHER KHAN, AND S. G. BRADY. 2000. Error, bias, and long-branch attraction in data for two chloroplast photosystem genes in seed plants. Mol. Biol. Evol. 17: 782–797.

SAUQUET, H., J. A. DOYLE, T. SCHARASCHKIN, T. BORSCH, K. W. HILU, L. W. CHATROU, AND A. LE THOMAS. 2003. Phylogenetic analysis of Magnoliales and Myristicaceae based on multiple data sets: implications for character evolution. Bot. J. Linn. Soc. 142: 125–186.

SCHMID, R. 1967. Electron microscopy of wood of *Callixylon* and *Cordaites*. Am. J. Bot. 54: 720–729.

SCHOPF, J. M. 1976. Morphologic interpretation of fertile structures in glossopterid gymnosperms. Rev. Palaeobot. Palynol. 21: 25–64.

SERBET, R. AND G. R. ROTHWELL. 1992. Characterizing the most primitive seed ferns. I. A reconstruction of *Elkinsia polymorpha*. Int. J. Plant Sci. 153: 602–621.

SHARMA, B. D. 2001. Misinterpretations about the 'Pentoxyleae'—a Mesozoic gymnospermous group of plants. Palaeobotanist 50: 255–265.

SHARMA, B. D. AND D. R. BOHRA. 1977. Structure of phloem in some plants of Bennettitales and Pentoxylales collected from the Rajmahal Hills, India. Geophytology 7: 214–216.

SHINDO, S., M. ITO, K. UEDA, M. KATO, AND M. HASEBE. 1999. Characterization of MADS genes in gymnosperm *Gnetum parvifolium* and its implication for the evolution of reproductive organs in seed plants. Evol. Devel. 3: 180–190.

SIEBER, P., J. GHEYSELINCK, R. GROSS-HARDT, T. LAUX, U. GROSSNIKLAUS, AND K. SCHNEITZ. 2004. Pattern formation during early ovule development in *Arabidopsis thaliana*. Devel. Biol. 273: 321–334.

SINGH, H. 1978. Embryology of gymnosperms. Encyclopedia of Plant Anatomy 10(2). Borntraeger, Berlin, Germany.

SKINNER, D. J., T. A. HILL, AND C. S. GASSER. 2004. Regulation of ovule development. Plant Cell Suppl. 16: S32–S45.

SMOOT, E. L. 1984a. Secondary phloem anatomy in *Callistophyton boyssetii* (Renault) Rothwell and

histological changes in the outer phloem. Bot. Gaz. 145: 395–406.

SMOOT, E. L. 1984b. Phloem anatomy of the Carboniferous pteridosperm *Medullosa* and evolutionary trends in gymnosperm phloem. Bot. Gaz. 145: 550–564.

SOLTIS, D. E., P. S. SOLTIS, AND M. J. ZANIS. 2002. Phylogeny of seed plants based on evidence from eight genes. Am. J. Bot. 89: 1670–1681.

SPORNE, K. R. 1965. The morphology of gymnosperms. Hutchinson Univ. Library, London, UK.

SPORNE, K. R. 1974. The morphology of angiosperms. Hutchinson Univ. Library, London, UK.

SRIVASTAVA, S. C. AND J. BANERJI. 2001. Pentoxylon plant: a reconstruction and interpretation. Plant Cell Biol. Devel. (Szeged) 13: 11–18.

STEBBINS, G. L. 1974. Flowering plants: evolution above the species level. Harvard Univ. Press, Cambridge, MA.

STEFANOVIC, S., M. JAGER, J. DEUTSCH, J. BROUTIN, AND M. MASSELOT. 1998. Phylogenetic relationships of conifers inferred from partial 28S rRNA gene sequences. Am. J. Bot. 85: 688–697.

STEWART, W. N. 1951. A new *Pachytesta* from the Berryville locality of southeastern Illinois. Am. Midl. Nat. 46: 717–742.

STEWART, W. N. AND G. W. ROTHWELL. 1993. Paleobotany and the evolution of plants, 2nd ed. Cambridge Univ. Press, Cambridge, UK.

STOCKEY, R. A. AND G. R. ROTHWELL. 2003. Anatomically preserved *Williamsonia* (Williamsoniaceae): evidence for bennettitalean reproduction in the Late Cretaceous of western North America. Int. J. Plant Sci. 164: 251–262.

SURANGE, K. R. AND S. CHANDRA. 1975. Morphology of the gymnospermous fructifications of the *Glossopteris* flora and their relationships. Palaeontographica Abt. B 149: 153–180.

SURANGE, K. R. AND H. K. MAHESHWARI. 1970. Some male and female fructifications of Glossopteridales from India. Palaeontographica Abt. B 129: 178–192.

SVOMA, E. 1997. Seed development and function in *Artabotrys hexapetalus* (*Annonaceae*). Plant Syst. Evol. 207: 205–223.

SWOFFORD, D. L. 1990. PAUP: phylogenetic analysis using parsimony, version 3.0. Illinois Natural History Survey, Champaign, IL.

TAKASO, T. 1985. A developmental study of the integument in gymnosperms 3. *Ephedra distachya* L. and *E. equisetina* Bge. Acta Bot. Neerl. 34: 33–48.

TAKHTAJAN, A. L. 1969. Flowering plants: origin and dispersal. Smithsonian Inst., Washington, DC.

TAYLOR, D. W. AND H. LI. 1997. Phylogenetic relationships of gigantopterids and their affinities to seed plants. Am. J. Bot. 84(6, Suppl.): 142–143.

TAYLOR, E. L. 1988. Secondary phloem anatomy in cordaitean stems. Am. J. Bot. 75: 1655–1666.

TAYLOR, E. L. 1990. Phloem evolution: an appraisal based on the fossil record, p. 285–298. *In* H.-D. Behnke and R. D. Sjoland [eds.], Sieve elements: comparative structure, induction and development. Springer, Berlin.

TAYLOR, E. L. 1996. Enigmatic gymnosperms? Structurally preserved Permian and Triassic seed ferns from Antarctica. Rev. Palaeobot. Palynol. 90: 303–318.

TAYLOR, E. L. AND T. N. TAYLOR. 1992. Reproductive biology of the Permian Glossopteridales and their suggested relationship to flowering plants. Proc. Natl. Acad. Sci. USA 89: 11495–11497.

TAYLOR, T. N., G. M. DEL FUEYO, AND E. L. TAYLOR. 1994. Permineralized seed fern cupules from the Triassic of Antarctica: implications for cupule and carpel evolution. Am. J. Bot. 81: 666–677.

THOMAS, H. H. 1925. The Caytoniales, a new group of angiospermous plants from the Jurassic rocks of Yorkshire. Philos. Trans. Roy. Soc. London Ser. B 213: 299–363.

THOMAS, H. H. 1933. On some pteridospermous plants from the Mesozoic rocks of South Africa. Philos. Trans. Roy. Soc. London Ser. B 222: 193–265.

TOBE, H., T. JAFFRÉ, AND P. H. RAVEN. 2000. Embryology of *Amborella* (Amborellaceae): descriptions and polarity of character states. J. Plant Res. 113: 271–280.

TOWNROW, J. A. 1960. The Peltaspermaceae, a pteridosperm family of Permian and Triassic age. Palaeontology 3: 333–361.

UMEDA, A., R. IMAICHI, AND M. KATO. 1994. Ovular development and morphology of the outer integument of *Magnolia grandiflora* (Magnoliaceae). Am. J. Bot. 81: 361–367.

VAN KONIJNENBURG-VAN CITTERT, J. H. A. 1992. An enigmatic Liassic microsporophyll, yielding *Ephedripites* pollen. Rev. Palaeobot. Palynol. 71: 239–254.

VILLANUEVA, J. M., J. BROADHVEST, B. A. HAUSER, R. J. MEISTER, K. SCHNEITZ, AND C. S. GASSER. 1999. *INNER NO OUTER* regulates abaxial-adaxial patterning in *Arabidopsis* ovules. Gen. Devel. 13: 3160–3169.

VISHNU-MITTRE. 1957. Studies on the fossil flora of Nipania (Rajmahal Series), India—Pentoxyleae. Palaeobotanist 6: 31–46.

WALTON, J. 1953. The evolution of the ovule in the pteridosperms. Advancem. Sci. 10: 223–230.

WARDLAW, C. W. 1965. The organization of the shoot apex. Encyclopedia of Plant Physiology 15(1): 966–1076.

WIELAND, G. R. 1916. American fossil cycads. Vol. 2. Taxonomy. Carnegie Inst. of Washington, Washington, DC.

WILLIAMS, J. H. AND W. E. FRIEDMAN. 2002. Identification of diploid endosperm in an early angiosperm lineage. Nature 415: 522–526.

WILLIAMS, J. H. AND W. E. FRIEDMAN. 2004. The four-celled female gametophyte of *Illicium* (Illiciaceae; Austrobaileyales): implications for understanding the origin and early evolution of monocots, eumagnoliids, and eudicots. Am. J. Bot. 91: 332–351.

WINTER, K. U., A. BECKER, T. MÜNSTER, J. T. KIM, H. SAEDLER, AND G. THEISSEN. 1999. MADS-box genes reveal that gnetophytes are more closely related to conifers than to flowering plants. Proc. Natl. Acad. Sci. USA 96: 7342–7347.

WON, H. AND S. S. RENNER. 2003. Horizontal gene transfer from flowering plants to Gnetum. Proc. Natl. Acad. Sci. USA 100: 10824–10829.

YAMADA, T., M. ITO, AND M. KATO. 2003. Expression pattern of *INNER NO OUTER* homologue in *Nymphaea* (water lily family, Nymphaeaceae). Devel. Genes Evol. 213: 510–513.

YAMADA, T., M. ITO, AND M. KATO. 2004. *YABBY2*-homologue expression in lateral organs of *Amborella trichopoda* (Amborellaceae). Int. J. Plant Sci. 165: 917–924.

ZANIS, M. J., D. E. SOLTIS, P. S. SOLTIS, S. MATHEWS, AND M. J. DONOGHUE. 2002. The root of the angiosperms revisited. Proc. Natl. Acad. Sci. USA 99: 6848–6853.

Appendix 1. Taxa and Characters

The data matrix for the present analysis is presented in Appendix 2.

TAXA

1. *Elkinsia* (Serbet and Rothwell 1992).
2. *Lyginopteris*.
3. Medullosans (*Quaestora* and *Medullosa*).
4. *Callistophyton*.
5. Cordaitales (consensus of *Mesoxylon* and *Cordaixylon* as reconstructed by Rothwell and Serbet 1994).
6. *Emporia* (Mapes and Rothwell 1984).
7. Pinaceae.
8. Podocarpaceae.
9. Araucariaceae.
10. Taxodiaceae (including Cupressaceae).
11. *Cephalotaxus*.
12. Taxaceae.
13. Ginkgoales.
14. Corystosperms.
15. *Autunia* (formerly *Callipteris*: Kerp 1988).
16. *Peltaspermum*.
17. Cycadales.
18. Glossopterids.
19. *Caytonia*.
20. Bennettitales.
21. *Pentoxylon*.
22. *Ephedra*.
23. *Welwitschia*.
24. *Gnetum*.
25. *Amborella*.
26. Nymphaeales (Cabombaceae and Nymphaeaceae).
27. *Austrobaileya*.
28. *Trimenia* (including *Piptocalyx*).
29. *Illicium*.
30. Schisandraceae.
31. Chloranthaceae (ancestral states based on Eklund et al. 2004, assuming *Hedyosmum* is basal to *Ascarina* and *Sarcandra* plus *Chloranthus*).
32. Saururaceae.
33. Asaroideae (Aristolochiaceae: *Saruma*, *Asarum*).
34. Winteraceae.

CHARACTERS

D96 designates characters of Doyle (1996); DE, characters of Doyle and Endress (2000). When changes from previous definitions or scorings are described, these are changes from the treatment of Doyle (1996) or Doyle and Endress (2000), as appropriate, unless otherwise indicated. Unmodified characters are documented and justified in those articles. All multistate characters except 103 are unordered.

General vegetative organization

1 (D96 1). Habit (0) woody, (1) (semi)herbaceous (secondary growth reduced or absent). Doyle and Endress (2000) recognized two related characters, DE 1 for tree or shrub vs. rhizomatous, scandent, or acaulescent, DE 5 for presence vs. absence of cambium. I have retained the original distinction because DE 1 is too difficult to extend to fossils and DE 5 is an autapomorphy of Nymphaeales. Chloranthaceae changed from (0/1) to (0) based on results of Eklund et al. (2004).

I deleted D96 2, radicle persistent vs. replaced by adventitious roots: with elimination of monocots, this is an autapomorphy of Nymphaeales.

2 (D96 3). Axillary buds (0) single, (1) multiple.

3 (D96 4). Vegetative short shoots (0) absent, (1) present.

Stem anatomy

4 (D96 14 modified). Apical meristem (0) without tunica, (1) one tunica layer, (2) two tunica layers. See text for discussion and references.

5 (D96 15). Cauline protoxylem (0) one central strand, (1) two or more sympodia.

6 (D96 16 modified). Stele (0) protostele or arcuate primary xylem segments, (1) eustele of more or less round bundles, (2) siphonostele or pseudosiphonostele. State (2) added from DE 2; Winteraceae scored as (1) based on Keating (2000).

7 (D96 18). Primary xylem (0) mesarch, (1) endarch.

8 (D96 19). Metaxylem (0) with scalariform pitting, (1) without scalariform pitting.

9 (D96 20 modified). Mature secondary xylem tracheids with (0) circular bordered pits or perforations only, (1) at least some scalariform pits or perforations. Redefined in terms of mature secondary xylem to address the condition in cordaites, where scalariform pitting extended into the early secondary xylem but gave way to circular bordered. Carlquist (1996a) stated that *Pentoxylon* had scalariform pitting, based on Vishnu-Mittre (1957), but this was refuted by Bose et al. (1985). Carlquist (1996a) questioned the previous scoring of cycads as (0/1) because scalariform pitting occurs in the early secondary xylem of *Cycas*, but because it is lacking in mature secondary xylem I have retained the previous scoring. I have changed *Gnetum* from (?) (based on Muhammad and Sattler 1982) to (0) because Carlquist (1996b) showed that the perforations are never truly scalariform. Doyle (1996) scored *Elkinsia* as (1), following Rothwell and Serbet (1994), but because Serbet and Rothwell (1992) described the meager secondary wood as having only circular bordered pits, I have rescored it accordingly.

10 (new). Torus in tracheid pits (0) absent, (1) present. See text for discussion and references.

11 (D96 21). Tertiary spiral thickenings in tracheids (0) absent, (1) present.

12 (D96 22, DE 7 modified). Tracheary elements (0) tracheids or cells with porose pit membranes, (1) vessel members with typical perforations. Redefined to include elements with porose membranes in (0), as in Eklund et al. (2004), and Nymphaeales are therefore still scored as (0).

13 (DE 8). Vessel grouping (0) predominantly solitary, (1) mostly pairs or multiples. Scored within an-

giosperms only; scoring in Gnetales would run risk of the Maddison effect (Maddison 1994), where the ancestral state in one taxon having a structure affects the inferred ancestral state in another taxon separated by lines in which the structure does not exist.

14 (D96 23, DE 9 in part). End-wall pits or vessel perforations (0) multiple, (1) simple. Like *Gnetum*, Schisandraceae, with mixed perforations, are scored as (?).

15 (D96 24 modified). Rays (0) at least some multiseriate, (1) uniseriate or uniseriate plus occasionally biseriate. State (1) redefined to express more clearly that it refers to woods with predominantly uniseriate rays.

16 (DE 11). Multiseriate rays (0) narrow (generally not more than four cells wide), (1) wide. Scored only in angiosperms because of uncertain applicability to gymnospermous taxa with uniseriate rays and lack of data in others.

17 (D96 25). Companion cells in phloem (0) absent, (1) present. Whether *Austrobaileya* has no companion cells in the normal sense (as sister cells of sieve-tube elements) or only very few is uncertain (Carlquist 2001), so I have changed its scoring from (1) to (?). I have not found data on *Amborella*.

18 (new). Fibers in secondary phloem (0) absent, (1) isolated or forming irregular groups or tangential bands, (2) forming regular, uniseriate tangential bands. See text for discussion and references. Not scored in groups with little or no secondary phloem.

19 (DE 17 modified). Pericycle (including modified protophloem) with (0) separate fiber bundles, (1) more or less continuous ring of fibers (or fibers and non-U-shaped sclereids). DE state (2), fibers alternating with U-shaped sclereids, is not present in this data set; state (3), no sclerenchyma, is found only in Nymphaeales (rescored ?) and questionably applicable because this character is associated with secondary growth, which is lacking in Nymphaeales. Saururaceae changed from (1) to (0/1) based on Eklund et al. (2004). I have not attempted to compile data on comparable features outside angiosperms, which would be especially difficult for fossils.

I deleted D96 26, sieve-tube plastid inclusions: with elimination of Magnoliales, Laurales, and monocots, the PI and PII types are autapomorphic for Pinaceae and Asaroideae, respectively.

20 (D97 27 modified). Secretory structures (0) absent, isolated cells, or groups of cells, (1) cavities, (2) canals. I have combined former states (0), isolated cells or groups of cells, and (3), absent, because of difficulty in distinguishing rare secretory cells from none, especially in fossils. Former state (4), oil cells, is treated as a separate character (37), since it is unlikely that these are transformations of the larger mucilage-containing structures in other groups. Because the canals of *Gnetum* (Carlquist 1996b) and Nymphaeales differ from those of other taxa in being laticifers, I have rescored them as (?).

21 (D96 28). Lignin with (0) no Mäule reaction, (1) Mäule reaction. Among added angiosperms, Gibbs (1957) recorded Mäule reaction in *Illicium*.

Leaf morphology and anatomy

22 (D96 5, DE 20). Phyllotaxy (0) spiral (helical), (1) distichous (at least on branches), (2) opposite or whorled.

23 (D96 17, DE 21 with different numbering of states). Nodes with (0) one trace from stele to each leaf, (1) more than three traces, (2) two traces from adjacent bundles, (3) three traces. Saiki and Yoshida (1999) compiled references for one-trace nodes in Bennettitales.

24 (D96 12). Leaf traces (0) mesarch, (1) endarch.

25 (DE 22). First appendage(s) on vegetative branch (0) paired lateral prophylls, (1) single distinct prophyll (adaxial, oblique, or lateral). Numbering of states was inadvertently reversed in Doyle and Endress (2000). *Trimenia* based on Eklund et al. (2004). Scored only in angiosperms because of lack of relevant data elsewhere, especially in fossils.

26 (D96 6, DE 27 in part). Leaf organization (0) pinnately compound, (1) simple and pinnately veined or compound but with parallel-veined leaflets, (2) linear or dichotomous with two or more veins, (3) palmately veined (actinodromous or acrodromous), (4) linear with one vein (rarely two; may fork apically).

27 (D96 7). Rachis (0) bifurcate, (1) simple. Scored only for pinnately compound leaves, except in *Caytonia*, in which the two pairs of leaflets are attached almost at one point.

28 (new). Rachial pinnules (0) absent, (1) present. A feature of peltasperms (*Autunia*, *Peltaspermum*) not used by Doyle and Donoghue (1986), where it was autapomorphic, or by Doyle (1996). Scored only for pinnately compound leaves, except *Caytonia* (cf. 27).

29 (DE 26 modified). Leaf shape (0) obovate to elliptical to oblong, (1) ovate. State (2), linear, deleted with elimination of monocots. *Trimenia* changed from (0) to (0/1) because it includes *Piptocalyx* (Eklund et al. 2004). Scored only in angiosperms because it is closely tied to their distinctive leaf architecture; although it could be scored in *Gnetum*, this would entail a risk of the Maddison effect (Maddison 1994).

30 (DE 30 modified). Chloranthoid teeth (0) absent, (1) present. DE states (2), monimioid, and (3), platanoid, are not represented. *Trimenia* scored as (1), *Illicium* as (0) following Eklund et al. (2004). Scored only in angiosperms (cf. 29).

31 (D96 8). Laminar venation (0) open, (1) reticulate.

32 (D96 9). Laminar vein orders (0) one, (1) two or more.

33 (D96 10). Guard cell poles (0) raised, (1) level with aperture. The similarity of *Caytonia* to angiosperms in this and other stomatal characters has been reaffirmed by Barbacka and Bóka (2000).

34 (D96 11, DE 31 modified). Stomata (predominant type on leaf when variable) (0) anomocytic (haplocheilic), (1) some or all paracytic (syndetocheilic), (2) stephanocytic (including tetracytic). Saururaceae are stephanocytic; in Chloranthaceae, *Hedyosmum* is stephanocytic, *Ascarina* encyclocytic, *Sarcandra* laterocytic, and *Chloranthus* laterocytic and paracytic (Eklund et al. 2004). Based on the topology of Chloranthaceae in Eklund et al. (2004), the most parsimonious ancestral state in the family could be any of these states. Treating laterocytic and encyclocytic as potentially related to either paracytic or stephanocytic, I have scored Chloranthaceae as (1/2), rather than paracytic in Doyle (1996) and laterocytic in Doyle and Endress (2000).

35 (DE 33). Palisade parenchyma (0) absent (mesophyll homogeneous), (1) present (mesophyll dorsi-

ventral). Chloranthaceae as a whole scored as (0), *Illicium* changed from (0) to (1), and Schisandraceae changed from (?) to (0) based on Eklund et al. (2004) and Feild et al. (2004). Data not compiled for nonangiospermous groups.

36 (D96 13, DE 34). Foliar astrosclereids (0) absent, (1) present. *Illicium* and Schisandraceae scored as (1), Winteraceae changed from (1) to (0) based on Doyle and Endress (2000).

37 (DE 35). Oil cells in mesophyll (0) absent, (1) present.

38 (DE 36). Mucilage cells in mesophyll (0) absent, (1) present. Scored only in angiosperms because of uncertainty in homology with mucilage cells and cavities in other groups (character 20).

General reproductive organization

39 (D96 46). Fertile appendages (0) not aggregated or in simple strobili, (1) simple male, compound female strobili, (2) compound male and female strobili. As in Doyle (1996), angiosperms with solitary flowers are scored as (0), as are those with botryoids and related inflorescences (*Amborella*, etc.), which differ from compound strobili of coniferophytes in having terminal flowers and intergrade with solitary flowers via types with occasional lateral flowers. Doyle (1996) scored spikes of Saururaceae and Chloranthaceae as (2), but because they differ from the gymnosperm states in being made up of flowers that are bisexual or probably derived from bisexual (Doyle et al. 2003), I have rescored them as (?).

Characters 40–42 are most informative for conifers and similar groups, but many other taxa can be reasonably assigned to the (0) state.

40 (D96 47). Symmetry of ovuliferous shoot (0) radial, (1) bilateral (dorsiventral). Radial includes bisymmetric, as in Gnetales.

41 (D96 48). Ovuliferous shoot (0) with distinct appendages, (1) cone scale without distinct appendages.

42 (D96 49). Bract and axillary female shoot (0) free, (1) fused.

Characters 43–47, which concern floral organization, are scored only in angiosperms because they cannot be applied to taxa with no flowers without making questionable assumptions on homology.

43 (DE 38 modified). Sex of flowers (0) bisexual, (1) unisexual. Because former state (1), bisexual and unisexual (usually male), occurs only in some *Trimenia* species, I have eliminated this state and rescored *Trimenia* as (0/1) (cf. Eklund et al. 2004).

44 (DE 40). Perianth phyllotaxy (0) spiral, (1) whorled.

45 (D96 50 modified, DE 41 in part). Perianth whorls (0) more than two, or spiral-irregular, (1) two whorls, (2) one or none. I have redefined state (2) to include one whorl in *Hedyosmum* as well as none in other Chloranthaceae.

46 (D96 51 modified, DE 42). Perianth merosity (0) irregular, (1) in threes, (2) in twos. DE state (2) also included fours and fives, not found in the present data set.

47 (DE 43). Outer perianth cycle (0) not clearly differentiated (or continuum of forms), (1) sepaloid.

I deleted D96 52, hypanthium, which occurs only in *Amborella*.

Microsporangiate structures

48 (D96 37). Microsporophylls (0) pinnate or paddle-like, (1) simple, one-veined, scale-like, (2) simple, one- (rarely three-) veined, with two pairs of longitudinal microsporangia. Doyle (1996) scored Chloranthaceae as (0/2) to allow for the possibility that the three-lobed androecium of *Chloranthus* is pinnate and ancestral, but because current phylogenies indicate that *Chloranthus* is derived (Eklund et al. 2004), I have rescored the family as (2). I have changed Gnetales from (0) to (0/1): the assumption that the androecium definitely consists of two branched rather than several simple sporophylls now seems premature, since the lateral grouping of sporangial units may be a consequence of the bisymmetric organization of the whole ''flower.''

49 (D96 39, DE 54 modified). Microsporangia (0) terminal, (1) abaxial, (2) adaxial, (3) lateral. See text for discussion of glossopterids and *Pentoxylon*. In addition to numbering the states differently, I have modified the limits used in Doyle and Endress (2000) character 54 (expressed as introrse, latrorse, extrorse) to restrict (1) and (2) to markedly extrorse and introrse, respectively, to avoid magnifying the variations among states in angiosperms, which are relatively minor compared to those differentiating other groups.

50 (D96 43, DE 48). Microsporophylls (0) free, (1) basally fused. Chloranthaceae changed from (?) to (0) because *Chloranthus*, interpreted as having fused stamens, is nested within the family (Eklund et al. 2004).

51 (D96 40). Microsporangia per sporophyll (0) more than two, (1) two.

52 (D96 41). Microsporangia (0) free, (1) fused at least basally.

53 (D96 42 modified). Microsporangial dehiscence (0) ectokinetic, (1) endokinetic, (2) endothecial. Following Nixon et al. (1994), Doyle (1996) scored several taxa as endokinetic, a condition recognized in *Ginkgo* by Jeffrey and Torrey (1916), with a fibrous layer below the epidermis of the microsporangia. However, Singh (1978), Martens (1971), and D'Arcy (1996) indicate that only *Ginkgo* is endokinetic; the scoring of other taxa was apparently an editing error (D. S. Stevenson, pers. comm.).

Characters 54–59, which concern androecial morphology, are scored only in angiosperms because they cannot be confidently applied to taxa with no flowers or depend on the special morphology of angiosperm stamens.

54 (DE 46 modified). Androecium phyllotaxy (0) spiral, (1) whorled. Former state (2), irregular, does not occur in this data set.

55 (D96 44 in part, DE 47 modified). Stamen merosity (0) irregular, (1) in threes, (2) in twos. DE state (2) also included fours and fives, not found in the present data set.

I deleted D96 45, inner staminodes; in the present data set these occur only in *Austrobaileya*.

56 (DE 49 modified). Stamen base (0) short (<2/3 length of anther), (1) long and wide (>1/2 width of anther), (2) long and narrow (typical filament). Replaces D96 38, stamens (0) laminar, (1) with well-differentiated filament. States (0) and (2) redefined as in Eklund et al. (2004).

57 (DE 51). Connective apex (0) extended, (1) truncated or smoothly rounded. Nymphaeales scored as (1), as in Cabombaceae and *Nuphar*, Chloranthaceae as (0), based on data and topology of Eklund et al. (2004).

58 (DE 56). Connective hypodermis (0) unspecialized, (1) endothecial or sclerenchymatous. Chloranthaceae scored as (1), based on *Hedyosmum* and *Ascarina*.

59 (DE 53). Pollen sacs (0) protruding, (1) embedded. *Trimenia* changed from (?) to (0), since its sacs are more protruding than those of *Ascarina*.

Pollen, microgametophyte

60 (D96 67, DE 58). Microspore cytokinesis (0) simultaneous, (1) successive. *Amborella* (1) based on Tobe et al. (2000).

61 (D96 68 modified, DE 61 in part). Pollen with (0) proximal tetrad scar, (1) distal sulcus or round germinal area, (2) no aperture, (3) tri- or hexacolpate. State (3) added with inclusion of *Illicium* and Schisandraceae. Several-armed aperture in *Hedyosmum* considered monosulcate for scoring of Chloranthaceae. Asaroideae (*Saruma* and *Asarum*) scored as (1/2) based on Doyle and Endress (2000), rather than (1) for Aristolochiaceae in Doyle (1996).

62 (D96 69). Pollen symmetry (0) radial, (1) bilateral, (2) global.

I deleted DE 60, pollen shape, because the only boat-shaped angiosperms in the present data set are Nymphaeales. I deleted DE 62, size, because only Nymphaeales (large) and Saururaceae (small) deviate from medium-sized.

63 (D96 70). Pollen (0) non-saccate or sub-saccate, (1) saccate.

64 (D96 71 modified, DE 63 in part). Infratectal structure (0) massive or spongy alveolar, (1) honeycomb alveolar, (2) granular, (3) columellar (including intermediate). I have combined the intermediate state of Doyle and Endress (2000), found in *Amborella* and some Nymphaeales (where it is inferred to be ancestral), with columellar; several authors have already considered it columellar (e.g., Osborn et al. 1991; Gabarayeva et al. 2003).

65 (D96 72). Exine striations (0) absent, (1) present. Not comparable to DE 65, which refers to a striate tendency of the muri.

66 (D96 73, DE 64 in part). Tectum (0) continuous or finely perforate, (1) foveolate-reticulate. DE state (2), reduced, is not represented in this data set. Asaroideae (0/1) based on Doyle and Endress (2000), rather than (0) for Aristolochiaceae in Doyle (1996).

67 (D96 74, DE 66). Supratectal spinules (0) absent, (1) present.

68 (D96 75, DE 68). Aperture membrane (0) smooth or weakly sculptured, (1) conspicuously sculptured. Asaroideae (1) based on Doyle and Endress (2000) and Dickison (1992) rather than (0) for Aristolochiaceae in Doyle (1996); Winteraceae (?) based on Doyle and Endress (2000) rather than (0) in Doyle (1996), to allow homology of the annulus with sculpture in other taxa.

69 (D96 76 modified). Endexine (0) uniformly thick (laminated), (1) thin (non-laminated), except under apertures, or absent. In angiosperms, Doyle (1996) distinguished (1) absent from (2), thin (non-laminated), except under apertures. However, with exclusion of Magnoliales and recognition of a thin endexine in *Amborella* (Hesse 2001), the only taxa in this data set that lack endexine are Chloranthaceae and some Nymphaeales (Cabombaceae), and even Chloranthaceae show what may be remnants of endexine (Chlonova and Surova 1988; Sampson 2000). Therefore I have combined the two states.

70 (D96 77). Microgametophyte with (0) five or more nuclei, (1) four nuclei, tube nucleus produced by the second division (no stalk cell), (2) four nuclei, tube nucleus produced by the first division (no prothallials), (3) three nuclei.

71 (D96 78). Sterile cell (0) colinear with other microgametophyte cells, (1) ring-shaped. In an oversight, Doyle (1996) scored Taxodiaceae as (0); because they resemble *Cephalotaxus* and Taxaceae in lacking prothallials, they are rescored as (?), like these taxa.

72 (new). Sperm size (0) small (<50 μm), (1) large (>50 μm). This character distinguishes medullosans, cycads, and *Ginkgo* from glossopterids and other modern taxa. Data compiled by Nishida et al. (2003, 2004). Benson (1908) reported sperm measuring about 41 μm in a pollen chamber of *Lagenostoma* (probably *Lyginopteris*), which was accepted by Rothwell and Serbet (1994) in scoring sperm of *Lyginopteris* as ''small, flagellate and zooidogamous.''

73 (D96 79). Sperm transfer (0) zooidogamous, (1) siphonogamous. Glossopterids scored as zooidogamous based on Nishida et al. (2003, 2004), Bennettitales as siphonogamous based on Stockey and Rothwell (2003). *Elkinsia* and *Lyginopteris* were inadvertently not scored in Doyle (1996) but can be assumed to be zooidogamous based on their spore-like pollen and Benson's (1908) report of sperm in *Lagenostoma* (cf. Nishida et al. 2004).

Ovulate structures

74 (D96 29 modified, D96 34 in part). Ovule-bearing structure (0) pinnate (ovules or ''cupules'' in two rows on a dorsiventral structure) or pinnate with a three-dimensional fertile portion, (1) simple, paddle-like (ovules not in two definite rows), (2) simple, stalk-like, with one ovule, or ovule sessile, (3) closed carpel with stigmatic pollen germination. In Doyle (1996) I scored multiovulate carpels as (0) and uniovulate carpels as unknown, but I have rescored all angiosperms as a new state (3), transferred from the former carpel character (D96 34). This avoids questionable assumptions that the carpel precursor was pinnate, allows for the possibility that paddle-like sporophylls in other taxa correspond to bitegmic ovules in angiosperms, and reduces the number of unknown scorings, which would otherwise increase relative to Doyle (1996) because of the greater number of uniovulate taxa. In Doyle (1996) I scored Cordaitales as (?) because their megasporophylls vary from simple to dichotomous, but I have changed this to (2) on the assumption that dichotomous is more likely related to stalk-like than to other types. See text for discussion of peltasperms, corystosperms, and *Caytonia*.

In angiosperms and Gnetales, ''ovule'' in characters 75–80 refers to the nucellus plus inner integument.

75 (D96 30). Ovule (0) on a lateral appendage or sessile but lateral on stem, (1) terminal on stem.

76 (D96 31 modified). Ovule position on supporting foliar structure (0) apical, (1) abaxial, (2) adaxial, (3) marginal. See text for discussion. Doyle (1996) lumped marginal, found only in cycads, with apical, but it makes fewer assumptions to separate the two states. I have changed the scoring of cycads from (0/1) to (3): although ovules are abaxial on a peltate sporophyll in some cycads (*Zamia*, *Encephalartos*, etc.), they are marginal in more basal taxa (*Cycas*, *Dioon*,

Stangeria: Norstog and Nicholls 1997). See text for discussion and references on other taxa.

77 (D96 32). Ovule orientation (0) erect, (1) inverted. In Podocarpaceae, Doyle (1996) assumed that taxa with erect ovules (e.g., *Phyllocladus*, *Microstrobus*) were derived based on then-available evidence, and this has been confirmed by more extensive analyses of Conran et al. (2000). Taxaceae and Gnetales were scored as erect, but because their orientation could be a consequence of their shift to a fully terminal position (75), it seems more prudent to score them as unknown.

78 (D96 33 modified, DE 85). Ovule (0) in radial, lobed ''cupule,'' (1) with no closely enclosing structure or in abaxially anatropous ''cupule,'' (2) in adaxially anatropous ''cupule'' or outer integument, (3) in orthotropous, unlobed ''cupule'' or outer integument. Because its opposite dorsiventral polarity implies that the anatropous cupule of corystosperms is not comparable to that of *Caytonia* and angiosperms (see text), I have redefined state (1) to include corystosperms, previously scored as (2). This distinction is not redundant with character 76, which also applies to groups with ovules on less modified leaves. Because the origin of the outer integument in Gnetales from two appendages seems well established (Martens 1971; Crane 1985; Takaso 1985), I have defined former state (4), bipartite outer integument derived from two primordia, as a separate character (79) and scored Gnetales as (0) for the present character. See text for discussion of Bennettitales.

79 (new). Bipartite outer integument around ovule (0) absent, (1) present.

D96 34, closed carpel, is included in character 74.

80 (new). Ovules per fertile short shoot or cone scale (0) more than one, (1) one. Scored only in conifers, cordaites, and ginkgos, in which homology of fertile short shoots is least contested. I have scored Taxaceae, with terminal ovules, as (?) because a cone scale is not recognizable. Although some Mesozoic fossils with two ovules per cone scale (*Rissikia*, *Mataia*: cf. Stewart and Rothwell 1993) have been assigned to Podocarpaceae, I have scored Podocarpaceae as (1), based on extant members, so that relationships of the fossils can eventually be tested rather than assumed.

Characters 81–93, which concern gynoecial morphology, are scored only in angiosperms because they cannot be confidently applied to taxa with no flowers or carpels. Here ''ovule'' refers to the whole bitegmic ovule.

81 (DE 71). Carpel number (0) more than one, (1) one.

82 (D96 35). Carpels (0) spiral or irregular, (1) whorled.

83 (DE 72 modified). Carpel form (0) ascidiate up to stigma, (1) completely plicate, or intermediate with some or all ovule(s) on the plicate zone. I have eliminated DE state (1), both plicate and ascidiate zones present below the stigma with ovule(s) on the ascidiate zone, because in this data set it occurs only in *Illicium*, which I have rescored as (?).

84 (DE 73 modified). Carpel sealing (0) by secretion, (1) complete postgenital fusion without canal. I have eliminated DE states (1), partial postgenital fusion with continuous unfused canal containing secretion, and (2), postgenital fusion to apex with partial canal containing secretion. Former state (1) occurs in *Illicium* only, which I have rescored as (?); former state (2) occurs only in some Nymphaeales (Nymphaeaceae).

85 (DE 74 modified). Pollen tube transmitting tissue (0) not prominently differentiated, (1) one prominently differentiated layer. State (2), more than one differentiated layer, does not occur in this data set.

86 (DE 75). Style (0) absent (stigma sessile or capitate), (1) present (elongated, distinctly constricted apical portion of carpel).

87 (DE 77). Stigma papillae (0) unicellular only (or stigma smooth), (1) some or all uniseriate pluricellular, (2) some or all pluriseriate pluricellular (including multicellular protuberances).

88 (DE 78). Extragynoecial compitum (0) absent, (1) present.

I deleted DE 79, carpel fusion: in this data set, only Saururaceae and Asaroideae are syncarpous, and they show different modes of carpel fusion (paracarpous and eusyncarpous, respectively).

89 (DE 80). Oil cells in carpels (0) absent or internal, (1) intrusive.

90 (D96 36, DE 82 modified). Ovules per carpel (0) one, (1) two or more. I have combined DE state (1), mostly two, with more than two: it occurs only in Schisandraceae, some Cabombaceae, and some Saururaceae. State (0) was previously defined as apical, but this is expressed in the next character.

91 (DE 84). Ovule direction (0) pendent, (1) horizontal, (2) ascendent.

92 (DE 93). Fruit wall (0) fleshy, (1) fleshy with hard endocarp (= drupe), (2) dry. In Doyle and Endress (2000), *Hedyosmum*, with a hard wall with aril-like outgrowths, was scored as (0/1), but because most of the fruit wall consists of the adnate perianth its correspondence to other types is unclear. Therefore I have considered *Hedyosmum* autapomorphic and scored Chloranthaceae as (0) based on the other genera.

93 (DE 94). Fruit dehiscence (0) indehiscent, (1) dehiscent. In Doyle and Endress (2000), numbering of the two states was inadvertently reversed in the state descriptions.

Ovule/seed morphology and anatomy

In angiosperms and Gnetales, ''ovule,'' ''integument,'' and ''seed'' in characters 94–100 refer to the nucellus plus inner integument.

94 (D96 53). Anatomical symmetry of ovule (0) radial (radiospermic), (1) bilateral or bisymmetric (platyspermic).

95 (D96 54). Apex of integument (0) free lobes, (1) simple, (2) bifid, (3) straight, tubular.

96 (D96 55). Integument (0) free from nucellus, (1) fused more than half way up from the base. See text for discussion of Bennettitales.

97 (D96 56). Lagenostome (0) present, (1) absent. Rothwell and Serbet (1994) scored corystosperms as having a lagenostome, but in Doyle (1996) I considered the character unknown. However, Klavins et al. (2002) showed a nucellar beak in petrified material, so I have rescored the group as (0).

98 (D96 57 modified). Pollen chamber (0) hydrasperman (with central column), (1) prominent but with no central column, (2) rudimentary to absent. Doyle (1996) recognized one state for ''nonhydrasperman or absent,'' which had been distinguished by Rothwell and Serbet (1994). At least a rudimentary pollen cham-

ber appears to be almost universally present in gymnospermous seed plants (Chamberlain 1935), but it is variably developed and inconsistently reported in the literature, and its complete absence in angiosperms may be a consequence of enclosure. However, it is possible to distinguish a prominent pollen chamber of the type seen in medullosans, *Callistophyton*, cordaites, *Ginkgo*, and cycads from the rudimentary and inconsistently developed chamber of conifers. Poort et al. (1996) proposed a correlation between prominent vs. vestigial pollen chamber and zooidogamy vs. siphonogamy, but this correlation is not perfect, since *Ephedra* has a prominent chamber (Martens 1971) but is siphonogamous, so it seems best to keep these as separate characters. Pollen chamber type is more often recognizable in fossil taxa. In light of these patterns, I have drawn a boundary between prominent and rudimentary pollen chamber, but not between the latter and no pollen chamber. This allows taxa for which the scoring of Rothwell and Serbet (1994) conflicted with previous reports to be scored as (2), such as *Welwitschia* (Martens 1971), Pinaceae, Podocarpaceae, and Taxaceae (Singh 1978).

A prominent pollen chamber is known in *Emporia* (Mapes and Rothwell 1984) and glossopterids (Nishida et al. 2003, 2004). Although the pollen chamber of *Gnetum* is somewhat intermediate, Martens (1971) considered it more like that of *Welwitschia* than *Ephedra*. In Doyle 1996 I scored corystosperms as unknown, but according to Klavins et al. (2002) they had a nucellar beak, so I have rescored them as (0), as in Rothwell and Serbet (1994). Reymanówna (1974) contrasted the small pollen chamber of *Caytonia* with the large chamber of *Callistophyton* (*Callospermarion*). I have not found clear evidence on the condition in *Pentoxylon* (Bose et al. 1985; Srivastava and Banerji 2001; Sharma 2001). See text for discussion of Bennettitales.

99 (D96 58). Micropyle (0) not sealed after pollination, (1) sealed.

100 (D96 59). Sarcotesta (0) absent or uniseriate, (1) multiseriate. As discussed in Doyle (1996), state (1) excludes what Rothwell and Serbet (1994) called a uniseriate sarcotesta, which they included in the same state as a classic thick, fleshy sarcotesta. By combining these conditions, they scored most gymnosperms as having a sarcotesta, including many not normally so described, such as *Lyginopteris*, conifers, and *Caytonia*. In the exceptions, corystosperms, Gnetales, and angiosperms, the absence of a sarcotesta was suspiciously correlated with ovule enclosure. Whether or not the uniseriate type is defined as a sarcotesta, it seems potentially more informative to distinguish it from the thick sarcotesta of cycads, *Ginkgo*, medullosans, and cordaites. Klavins et al. (2002) described the integument of corystosperms as consisting of outer thin-walled isodiametric cells and inner thicker-walled tabular cells, but because the whole integument is so thin and unsclerified I have continued to score the group as (?). Similarly, because the inner integument of angiosperms and Gnetales, the presumed homolog of the integument of other seed plants, is reduced and/or unsclerified (Martens 1971; Corner 1976), presumably as a result of enclosure in the outer integument, I have rescored these groups as (?) rather than (0). In Bennettitales, Rothwell and Stockey (2002) and Stockey and Rothwell (2003) described a sarcotesta in petrified material of *Cycadeoidea* and *Williamsonia*. Much of its thickness consists of one layer of radially elongated cells, but because it includes two cell layers in *Cycadeoidea* and multicellular pegs in *Williamsonia*, I have rescored Bennettitales as (1). The sarcotesta of *Austrobaileya* (see character 106) is in the outer integument and therefore not comparable.

101 (D96 60). Nucellus (0) not vascularized, (1) vascularized at least at base. Corystosperms were previously unknown, but Klavins et al. (2002) showed that they had a basal vascular disk.

102 (D96 61). Nucellar cuticle (0) thin, (1) thick. See text for discussion of Bennettitales.

Characters 103–106 are scored only in angiosperms because they depend on the bitegmic nature of the ovule.

103 (DE 89). Outer integument thickness (at middle of integument length) (0) two cells, (1) two and three to four, (2) four and five, or more. Ordered. Chloranthaceae scored as (0/1/2) based on data and topology of Eklund et al. (2004).

104 (DE 88). Outer integument lobation (0) unlobed, (1) lobed.

105 (D96 63, DE 96 in part). Exotesta (0) normal, (1) palisade. DE 96 state (2), tabular, is not represented in this data set.

106 (DE 97 modified). Mesotesta (0) unspecialized, (1) sclerotic. Two states in DE 97, (2) fibrous and (4) spongy, are not represented in this data set. State (3), sarcotesta, occurs in *Austrobaileya*, but this genus also has a sclerotic layer in the inner part of the mesotesta (Endress 1980) that may be homologous with that of other Austrobaileyales. This suggests that sarcotesta and sclerotic mesotesta should be treated under two independent characters, but in the present data set sarcotesta is autapomorphic and has therefore been eliminated.

I deleted D96 64, ruminations in the seed coat: with elimination of eumagnoliids, it occurs only in *Austrobaileya*.

107 (D96 65). Megaspore tetrad (0) tetrahedral, (1) linear.

108 (D96 66 modified). Cutinized megaspore membrane (0) present, (1) absent. See text for discussion. Nixon et al. (1994) scored Taxaceae as thick, Rothwell and Serbet (1994) as thin. Because I was unable to resolve this discrepancy I scored them as unknown (Doyle 1996), but with the present character definition they can be scored as (0).

Megagametophyte, fertilization, embryo

109 (D96 80). Megagametophyte (0) monosporic, (1) tetrasporic. In Doyle (1996), I scored Piperales (Piperaceae, Saururaceae) as (0/1), but Saururaceae alone are (0).

110 (D96 81 modified). Megagametophyte (0) large, cellular, with normal archegonia; (1) large, apical part and egg free-nuclear; (2) one or two four-nucleate modules, consisting of a group of three cells (including egg) and one free nucleus, no neck cells. See text for discussion.

111 (new). Megagametophyte modules (0) one, (1) two, one at each pole of gametophyte. See text for discussion.

112 (D96 82). Megagametophyte cellularization (0) enclosing single nuclei, resulting in uninucleate cells, (1) enclosing several nuclei, resulting in multinucleate-polyploid cells.

113 (D96 83). Fusion of (0) only one sperm with a female gametophyte nucleus, (1) regular fusion of both sperm. Friedman and Floyd (2001) reviewed scattered reports of double fertilization in conifers; except for *Thuja*, most are in Pinaceae (*Abies*, *Pinus*, *Pseudotsuga*), which I have therefore rescored as (0/1).

I deleted D96 84, fertilization producing embryo plus triploid endosperm tissue, as redundant with characters 111, 113, and 114 (see text for discussion).

114 (new). Provisioning of embryo (0) in female gametophyte before fertilization, (1) in female gametophyte before and after fertilization, (2) in female gametophyte after fertilization, (3) in endosperm derived from double fertilization. See text for discussion.

115 (DE 105, D96 85 in part). Perisperm (diploid nourishing tissue derived from the nucellus) (0) absent, (1) present. In D96 85, perisperm plus endosperm and endosperm only were treated as two states of one character. However, presence of endosperm is treated here in character 114, and as the previous definition acknowledged the two types of tissue are independent, since both occur in seeds of Nymphaeales and Piperales. For this reason they were split in Doyle and Endress (2000). Presence or absence of endosperm in the mature seed (DE 104) would be uninformative, because all angiosperms in the present data set have seeds with endosperm.

116 (new). First division of zygote (0) free-nuclear, (1) cellular. See text for discussion of this character and its relation to the next.

117 (D96 86). Embryo (0) derived from several free nuclei, (1) from a single uninucleate cell by cellular divisions.

118 (D96 87 and 88 modified). Proembryo (0) massive, no visible tiers, (1) tiered, cells of embryo tier elongating to form secondary suspensor, (2) not tiered, no secondary suspensor, derivatives of primary suspensor cell contributing to embryo. See text for discussion and references.

119 (D96 89). Feeder in embryo (0) absent, (1) present.

120 (D96 90). Seeds shed (0) without, (1) with well-developed embryo.

121 (D96 91, DE 108). Seed germination (0) hypogeal, (1) epigeal. Numbering of states reversed in Doyle and Endress (2000).

Appendix 2. Data Matrix

A = 0/1; B = 0/2; C = 1/2; D = 1/3; E = 2/3; F = 2/4; G = 0/1/2. See Appendix 1 for complete taxon names.

	1 2 3 4 5 6 7 8 9 0 1 2 12345678901234567890123456789012345678901234567890123456789012345678901234567890123456789012345678901
Elkinsia	0?0?00000?00?00????0?00??000??00??????0000????0000?00001?????????000?000?????0000000?????????????????00000000?????0000?0????????0?
Lyginopt	000?11000?00?00??0?0?000?000??0000?0??0000?????00001O??????O000000????00000000?????????????????01100000?????0000?0?0??????0?
Medullos	000?10000?00?00?01?2?B?0?000??0000?0??0000?????00001O??????O100000?0??1000001O?????????????????01001011?????0000?0?0??????0?
Callisto	000?11000?00?00?00?1?000?010??00???0??0000?????010010???????1111000000???001010?????????????????12001111??????0?0?0?0??????0?
Cordaita	0A00111000000?01?00?1?000?2????0000?0??2000?????100000???????0?11000?00??02000100?????????????????12001111??????000?0?0??????0?
Emporia	000?11100?00?01????0?00??4?????000????1100?????12000????????0111000?????02001101?????????????????12001000??????0????????????1?
Pinaceae	00A011110100?01?00?20001?4?????000?00?1110?????110100??????011110000000012O?1100?????????????????2112100?????100000A10001011
Podocarp	000011110100?01?02?2A001?4?????000?00?1110?????110100??????011110000000012O?1101?????????????????2112100?????100000010001011
Araucari	000C11110100?01?01?20B01?F????0000?00?1111?????110000??????01002000?000012O?1101?????????????????20121000????100000010001011
Taxodiac	000011110100?01?02?20001?4?????000?00?11A1?????110000??????0100200000 2?0120?A100?????????????????2A121000????100000010001011
Cephalot	000011110110?01?0??20001?4?????000?00?111??????110000??????11002000?02?0120?0100?????????????????12112110?????100000010001011
Taxaceae	000011110110?01?02?20001?4?????000?00?00???????110000??????122020 00?02?0121??30?????????????????12112100?????100000010001011
Ginkgoal	001011110100?01?01?10021?2????0000?00?0000?????1A0A01??????011010000001102000100?????????????????12101110O????100000000000010
Corystos	000?11100?00?01????1?011?000??0000?0??0000?????010000???????1111000000????101010?????????????????1200???10?????1??????????????
Autunia	????????????????????????????011??00?0????0000?????01000???????1111000000????101010?????????????????12????????????????????????????
Peltaspe	????????????????????????????011??00?0????0000?????010000???????110?0000?????101010?????????????????120?????0?????0??????????????
Cycadale	0?001110A000?00?01?20010?1????0000?00?0000?????0100A0??????011010000001100030 10?????????????????A11011110????100000000000010
Glossopt	00??11?00?00?01????0?0B0?1????1000?0??000???????0?000????????1111100000??001020 10?????????????????1101110?1?????0?0???????????
Caytonia	?0?????????????????????0???0?????1010????000???????010010???????1111000000????002020?????????????????11012?001?????1?????????????
Bennetti	000?11101000?00?01?2?001?1????0001????000??????02A010???????1102000000???1?0?0D0?????????????????3A12111A????110000????????1?
Pentoxyl	001?11?00?00?01?00?0?020?1????0000?0??000???????0?0000???????1102000000????10?00?????????????????110??1111?????1?????????????
Ephedra	0101111101O1?00?00?01221?2?????000?00?200??????A01010??????02102100?0000121??11?????????????????31111?10????100000110011011
Welwitsc	010?11110001?10?00?21221?2????1101?10?200??????A01010??????011021000 01?0121??11?????????????????131121?10?????01101?10111111
Gnetum	01?111101O1??0?00??1211?1????1101?10?200?????A01??0??????02202?00?01?0121??11?????????????????031121?00?????01101120111111
Amborell	000?11101000?000?010?12101??111111000000?10000220012001000111030011 13?0130?030?000000210001 0?10121???1100110210130112011
Nymphaea	1002111 0?000?0??1????0E103??1011101A00000?01A102200121AB10A011030000 13?0130202 0?01000A1A010B0?10121?01C0101102001311120 10
Austroba	000?111010010000?010122101??1011110010000?000002200120010000110301011 3?0130202 0?0000110101100?10121?0120011102001301120 11
Trimenia	000?11101001A00A1010?22101??A111110011000?A00002300120020000??03011113?0130202 0?1?00002?A0000?10121???2111110200130112 01?
Illicium	000212101001000 01?00100101??001111111110 00?000022001200110003003010113?0130202 0?00??1101102 21?10121???2111110200130112011
Schisand	000212101001 0?001110?00101??011111011100 0?10000 2?101200110003003010113?0130202 0?0000100111100?10121????2A11110200130112011
Chlorant	000211101001000110001221 01??01111C0010?00?1121?230012?000110110301111 3?0130?030?1?0000B?00000?10121?01GA0011021013011201 1
Saururac	100?11101001100 11?A0111113??101112101??00?0?2??2300121121010110300011 3?0130?030?01110100111BA?10121?01010?1102101311120 11
Asaroide	100?11101001110110A0113113??1011101010000?011112100121120000C1030A011 3?0130202 0?0111111001121?10121?010A0?1102101301120 11
Winterac	000211101000?001100010310 1??001111001 0000?0102123001212111001003010?13?0130202 0?00111000011 00?10121?0110101102101301120 11