

# Historical biogeography of two cosmopolitan families of flowering plants: Annonaceae and Rhamnaceae

J. E. Richardson<sup>1\*</sup>, L. W. Chatrou<sup>2</sup>, J. B. Mols<sup>3</sup>, R. H. J. Erkens<sup>2</sup> and M. D. Pirie<sup>2</sup>

<sup>1</sup>Nationaal Herbarium Nederland, Wageningen Universiteit Branch, Generaal Foulkesweg 37, 6703 BL Wageningen, The Netherlands

<sup>2</sup>Nationaal Herbarium Nederland, Universiteit Utrecht Branch, Heidelberglaan 2, 3584 CS Utrecht, The Netherlands

<sup>3</sup>Nationaal Herbarium Nederland, Universiteit Leiden Branch, PO Box 9514, 2300 RA Leiden, The Netherlands

Annonaceae are a pantropically distributed family found predominantly in rainforests, so they are megathermal taxa, whereas Rhamnaceae are a cosmopolitan family that tend to be found in xeric regions and may be classified as mesothermal. Phylogenetic analyses of these families are presented based on *rbcL* and *trnL-F* plastid DNA sequences. Likelihood ratio tests revealed rate heterogeneity in both phylogenetic trees and they were therefore made ultrametric using non-parametric rate smoothing and penalized likelihood. Divergence times were then estimated using fossil calibration points. The historical biogeography of these families that are species rich in different biomes is discussed and compared with other published reconstructions. Rhamnaceae and most lineages within Annonaceae are too young to have had their distribution patterns influenced by break-up of previously connected Gondwanan landmasses. Contrasts in the degree of geographical structure between these two families may be explained by differences in age and dispersal capability. In both groups, long-distance dispersal appears to have played a more significant role in establishing modern patterns than had previously been assumed. Both families also contain examples of recent diversification of species-rich lineages. An understanding of the processes responsible for shaping the distribution patterns of these families has contributed to our understanding of the historical assembly of the biomes that they occupy.

**Keywords:** molecular clocks; Gondwanaland; boreotropics; dispersal; diversification; biomes

## 1. INTRODUCTION

A biome may be defined as a 'major community, classified according to the predominant vegetation and characterized by adaptations of organisms to that particular environment' (Campbell 1996, p. 1106). Determination of the biogeographic histories of taxa that occupy a particular biome allow us to build up a picture of its historical assembly. Plate tectonic theory offered a seemingly plausible and potentially universal explanation for how organisms attained their modern distributions. Modern patterns could be explained by considering the changing positions of the continents since the Cretaceous. As well as tectonic changes, global distribution patterns may have been affected by climatic change. Zachos *et al.* (2001) reviewed trends in climate change over the past 65 Myr that have been measured using  $\delta^{18}\text{O}$  isotope records. A pronounced warming trend occurred from the Mid-Paleocene (59 Myr ago) to the Early Eocene (52 Myr ago) culminating in an EECO (52 to 50 Myr ago). This was followed by general cooling until 26–27 Myr ago when warming that peaked 17–15 Myr ago in the Late Middle Miocene climatic optimum was followed by gradual cooling and re-establishment of a

major ice sheet on Antarctica by 10 Myr ago. Climate on a regional scale has been affected by continental drift, for example isolation and poleward drift of Antarctica allowed the development of a cold circum-Antarctic (Benguela) current (Siesser 1980) that resulted in aridification of southwest Africa and southwest Australia. Closure of the Tethys Sea was also in part responsible for aridification throughout Africa (Axelrod & Raven 1978).

Fossil-based estimates by Magallón & Sanderson (2001) using the dataset used by the Angiosperm Phylogeny Group (1998) indicated that many cosmopolitan lineages of angiosperms radiated during comparatively recent times in the Tertiary, and do not extend back to the Cretaceous. Wikström *et al.* (2001) used a fossil representing the split between Cucurbitales and Fagales to evaluate divergence times within angiosperms using NPRS (Sanderson 1997) and penalized likelihood (Sanderson 2002) of a tree based on *rbcL*, *atp $\beta$*  and 18S rDNA genes and 567 taxa. Although Wikström *et al.* (2001) obtained slightly older estimates than Magallón & Sanderson (2001), many angiosperm lineages are still too young to have been influenced by break-up of larger landmasses, and it is therefore likely that continental history may have had less of an effect on the modern distributions of flowering plants than had previously been assumed.

Migration of organisms may depend on the availability of a suitable land connection and also on suitable climatic

\* Author for correspondence (james.richardson@wur.nl).

One contribution of 16 to a Discussion Meeting Issue 'Plant phylogeny and the origin of major biomes'.

conditions. The combination of tectonic and climatic changes presented new opportunities for migration and either *in situ* or post-dispersal diversification. Morley (2003) reviewed potential migration paths across the globe for megathermal angiosperms, suggesting, for example, that connections between South America and Africa could have existed up until the Oligocene via stepping stone dispersal across islands of the Rio-Grande Rise and the Walvis Ridge, which was above water southwest of the coast of Africa up until this time according to Parrish (1993), and also the Sierra Leone Rise. Dispersal through Laurasia at times during the EECO when climatic conditions supported tropical vegetation may provide the best explanation for many organisms that now have disjunct distributions between the tropics of South America, Africa and Southeast Asia. Tiffney (1985*a,b*) suggested that the common eastern Asian and eastern North American disjunct pattern may have had multiple origins throughout the Tertiary during which many taxa could have migrated between Asia and North America via the Bering land bridge or the North Atlantic land bridge. Manchester (1999) also supports the multiple patterns in the timings and routes for floristic exchanges between Asia and North America, with fossil floras documenting tropical forests at high latitudes in Laurasia during the EECO. These connections meant that migration could have been possible between the floras of Africa, Indochina and North America. Links between North and South America might have been possible via the proto Greater Antilles (*ca.* 50 Myr ago) or via the GAARlandia that existed 33–35 Myr ago (Iturralde-Vinent & MacPhee 1999; Graham 2003; see also figs 2 and 3 in Pennington & Dick 2004). Actinidiaceae, Annonaceae, Bignoniaceae, Lauraceae, Menispermaceae, Musaceae and groups of Myrtaceae, Rubiaceae, Palmae (Arecaceae) and Zingiberaceae are examples of boreotropical groups that were widespread in the Eocene and Oligocene Laurasia that, as a result of climatic changes, are now more or less restricted to Indomalaysia, tropical Africa, and/or the New World tropics (Mai 1995; Kubitzki & Krutzsch 1996; Manchester 1999).

Links for mesothermal taxa may also have been possible well after the break-up of Gondwanaland. There is fossil evidence for a Madrean–Tethyan xerophytic flora (Axelrod 1975) that existed from the Late Eocene along the Tethyan coast from North America to southeastern Eurasia and North Africa only disrupted by increased cooling and drying at the end of the Oligocene (see fig. 4, Pennington & Dick 2004). Dispersal of mesothermal taxa between North and South America could also have been possible via the proto Greater Antilles or GAARlandia in the Mid- to Late Eocene or along a Central American land bridge in the Late Miocene (Bermingham & Martin 1998). Raven (1982) suggested that continental connections between South America–Antarctica–Australia were possible from before the time of the origin of the angiosperms until the close of the Eocene (*ca.* 40 Myr ago). This route may have been possible for mesothermal taxa (e.g. Bombacaceae (Malvaceae ‘baobabs’), Polygonaceae, Restionaceae and Sapindaceae as suggested by Morley (2003)) but is less likely for frost-intolerant megathermals.

Molecular phylogenetic studies of the history of components of tropical biota (e.g. Lauraceae (Chanderbali *et al.* 2001), Melastomataceae (Renner *et al.* 2001) and

Malpighiaceae (Davis *et al.* 2002*a*)) have greatly improved our understanding of the assembly of pantropical floristic diversity. These studies suggest that a large proportion of the diversity of tropical regions may be a result of migration of boreotropical megathermal taxa into southern continents in the Oligocene and Miocene. Molecular studies also provide evidence for the break-up of Madrean–Tethyan links. For example, the 25 Myr ago estimate of the trans-Atlantic disjunction in the *Ocotea* (Lauraceae) complex as described by Chanderbali *et al.* (2001) is consistent with that expected for taxa with Madrean–Tethyan ranges. Vicariance among North American and Mediterranean lineages in Arbutioideae (Ericaceae) that split at approximately the same time, are also consistent with the break-up of this vegetation type (Hileman *et al.* 2001).

Molecular phylogenetic studies also demonstrate that the role of long-distance dispersal in explaining modern distributions appears to have been underestimated. Dates of clades that are disjunct between Africa and North America have been demonstrated to be too recent for migration via any of the routes discussed above (Lavin *et al.* 2004) and long-distance dispersal must therefore be invoked. Renner *et al.* (2001) demonstrated examples of intercontinental long-distance dispersal followed by significant radiation in Melastomataceae *s.str.*, and Malpighiaceae have recently (*ca.* 8 Myr ago) dispersed from Madagascar to New Caledonia (Davis *et al.* 2002*b*).

Annonaceae are a pantropical family of trees, shrubs and climbers with *ca.* 130 genera and 2500 species that are found predominantly in lowland tropical regions. This distribution and the fact that Annonaceae often represent a dominant component of tropical rainforest (e.g. Burnham & Johnson 2004) make them an excellent model group for the study of angiosperm diversification in this biome. Annonaceae may have dispersed via the migration routes for megathermal angiosperms that were suggested by Morley (2003). Rhamnaceae are another cosmopolitan family of trees, shrubs, climbers, and one herb consisting of *ca.* 50 genera and 900 species. They have a tendency towards xeromorphism and favour drier habitats. Raven & Axelrod (1974) stated that Rhamnaceae are so well represented both in tropical and temperate regions that it is difficult to trace the history of the family. Rhamnaceae may well have migrated by routes more suitable for mesothermal taxa as described above. Differences in dispersal capability between Annonaceae and Rhamnaceae may also have had a strong influence on modern distributions and patterns of diversity. Annonaceae propagules are dispersed primarily by primates (Van Tol & Meijdam 1991), whereas Rhamnaceae dispersal seems to be more effective (many more genera in Annonaceae are restricted to a single continent in comparison to many genera in Rhamnaceae that are widely distributed), and by a wider range of vectors (e.g. wind, *Gouania* (Mori & Brown 1994); or bird, *Frangula* (Hampe & Bairlein 2000) and *Maesopsis* (Clark *et al.* 2004)).

This study aims to determine the biogeographic histories of both Annonaceae and Rhamnaceae to ascertain which migration routes suggested by Morley (2003) and other routes described above may have been taken by these families and to determine the comparative contribution of vicariance and long-distance dispersal in shaping current distribution patterns in both groups. We also aim to

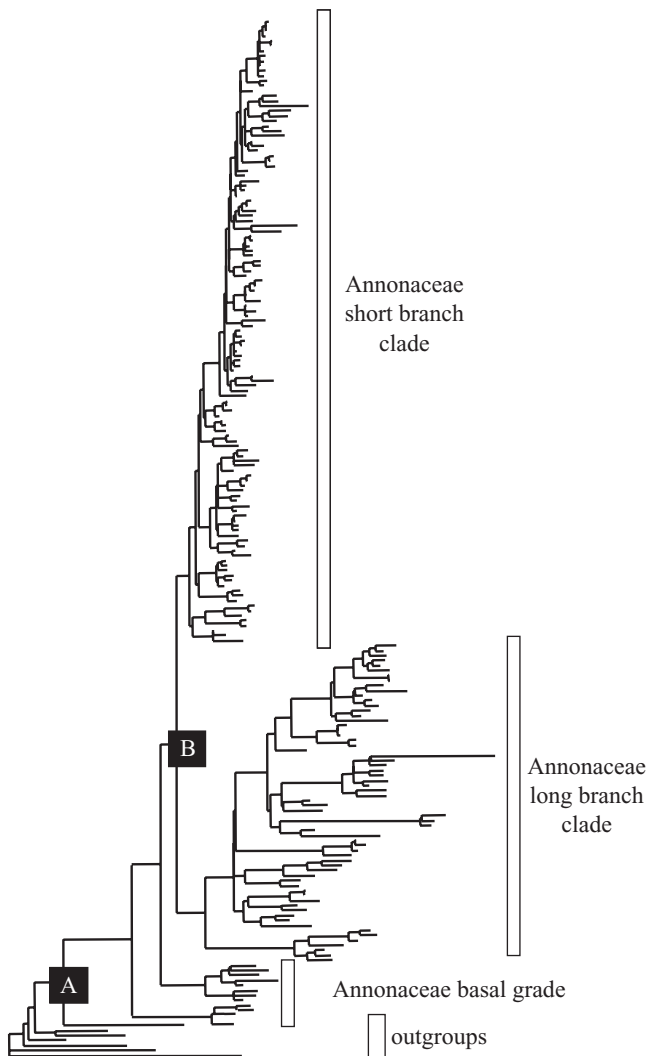


Figure 1. One of the most parsimonious trees from the analysis of Annonaceae *rbcL* and *trnL-F* DNA sequence data. Dates for nodes A and B are indicated in table 1.

demonstrate the timing of diversification of species-rich lineages within both groups.

## 2. MATERIAL AND METHODS

### (a) Taxon sampling and phylogenetic reconstruction

This study used previously published *rbcL* and *trnL-F* sequence data (Richardson *et al.* 2000; Fay *et al.* 2001; Mols *et al.* 2004; Pirie *et al.* 2004). Details of accessions are available in these publications. Alignment of *rbcL* sequences was performed manually and unambiguously because of the absence of insertions or deletions. An initial alignment was performed for five *trnL-F* sequences using CLUSTAL v. 1.61 (Higgins *et al.* 1992) and subsequent sequences were aligned manually. The Annonaceae dataset included sequences of 1421 *rbcL* and 1427 *trnL-F* characters representing species of 79 of the *ca.* 130 genera in Annonaceae, plus representatives of genera in Magnoliaceae, Myristicaceae, Degeneriaceae, Himantandraceae, Eupomatiaceae and Lauraceae, in a total of 208 terminal taxa. *Persea americana* (Lauraceae) was chosen as the ultimate outgroup because analyses of wider taxonomic range had shown Lauraceae to be a sister to the rest (Sauquet *et al.* 2003). For the Rhamnaceae data we used 1416 *rbcL* and 1375 *trnL-F* characters representing 43 out of the 50 genera in a total of 71 terminal taxa.

Outgroups were the same as those used in Richardson *et al.* (2000).

Data were analysed using the parsimony algorithm of the software package PAUP\* v. 4.0b10 (Swofford 1998). Searches were conducted on *rbcL* and *trnL-F* datasets combined. Tree searches were conducted under the equal and unordered weights criterion (Fitch parsimony (Fitch 1971)) with 1000 random sequence additions and TBR swapping, but permitting only five trees to be held at each step. All the shortest trees collected in the 1000 replicates were then used as starting trees for another round of heuristic search, and all these trees were swapped on to completion or swapped on until 5000 trees were produced, at which point we limited the number of trees and swapped on the 5000 trees collected. One thousand replicates of the bootstrap (Felsenstein 1985) were then carried out with simple addition of sequences, TBR branch swapping and holding five trees at each replicate. We applied the following scheme of support: bootstrap values of 50–74% represent weak support, 75–84% moderate support, and 85–100% strong support.

### (b) Likelihood ratio test

For both datasets we optimized the data on one of the trees from the combined *rbcL* and *trnL-F* parsimony analyses to estimate branch lengths. Branch lengths were calculated using maximum likelihood as implemented in PAUP\*, using the GTR +  $P_{inv}$  +  $I$  model that was indicated as optimal for both Annonaceae and Rhamnaceae datasets by MODELTEST v. 3.06 (Posada & Crandall 1998). Rate heterogeneity among lineages was evaluated using the likelihood ratio test, which compares log likelihoods of both constrained and unconstrained hypotheses on one of the trees from the parsimony analysis. If the molecular clock was rejected because the constrained and unconstrained analyses were significantly different, Sanderson's methods of NPRS (Sanderson 1997) and penalized likelihood (Sanderson 2002) were applied to estimate divergence times using the software package Rss (Sanderson 2003). We also estimated the standard deviation for the NPRS-derived dates by keeping the tree fixed and bootstrapping the dataset 100 times using PAUP\*. The resulting trees were then processed in Rss to calculate the standard deviation of divergence times for selected nodes.

### (c) Calibration

Both datasets used calibrations based on fossil material and it should be emphasized that all timings are therefore minimum age estimates. For the Annonaceae data we calibrated the tree at three nodes in absolute time: (i) using *Archaeanthus*; (ii) using the Wikström *et al.* (2001) age estimates for the stem of the family, i.e. 91 and 82 ± 4 Myr ago (using *Archaeanthus* gives an estimated age for stem Annonaceae of 90.6 ± 1.3 Myr ago, i.e. it matches that of one of the Wikström *et al.* (2001) age estimates so we report calibration (i) based on *Archaeanthus* and (ii) based on the younger (82 Myr ago) Wikström *et al.* (2001) estimate for Annonaceae); and (iii) using Maastrichtian seeds. *Archaeanthus* is an early Cenomanian fossil from North America (Dilcher & Crane 1984) that appears to be related to Magnoliaceae, based on the distinctive stipules, elongate receptacle and the fruits, which consist of numerous well-spaced follicles. It was therefore used here to assign a minimum age of 98 Myr for the stem of Magnoliaceae. The age estimates of Wikström *et al.* (2001) were calculated based on optimization of both parsimony (using ACCTRAN and DELTRAN) and maximum-likelihood branch lengths and transforming branch lengths using NPRS and penalized likelihood. These estimates had confidence intervals of ± 4 Myr but our calibrations did

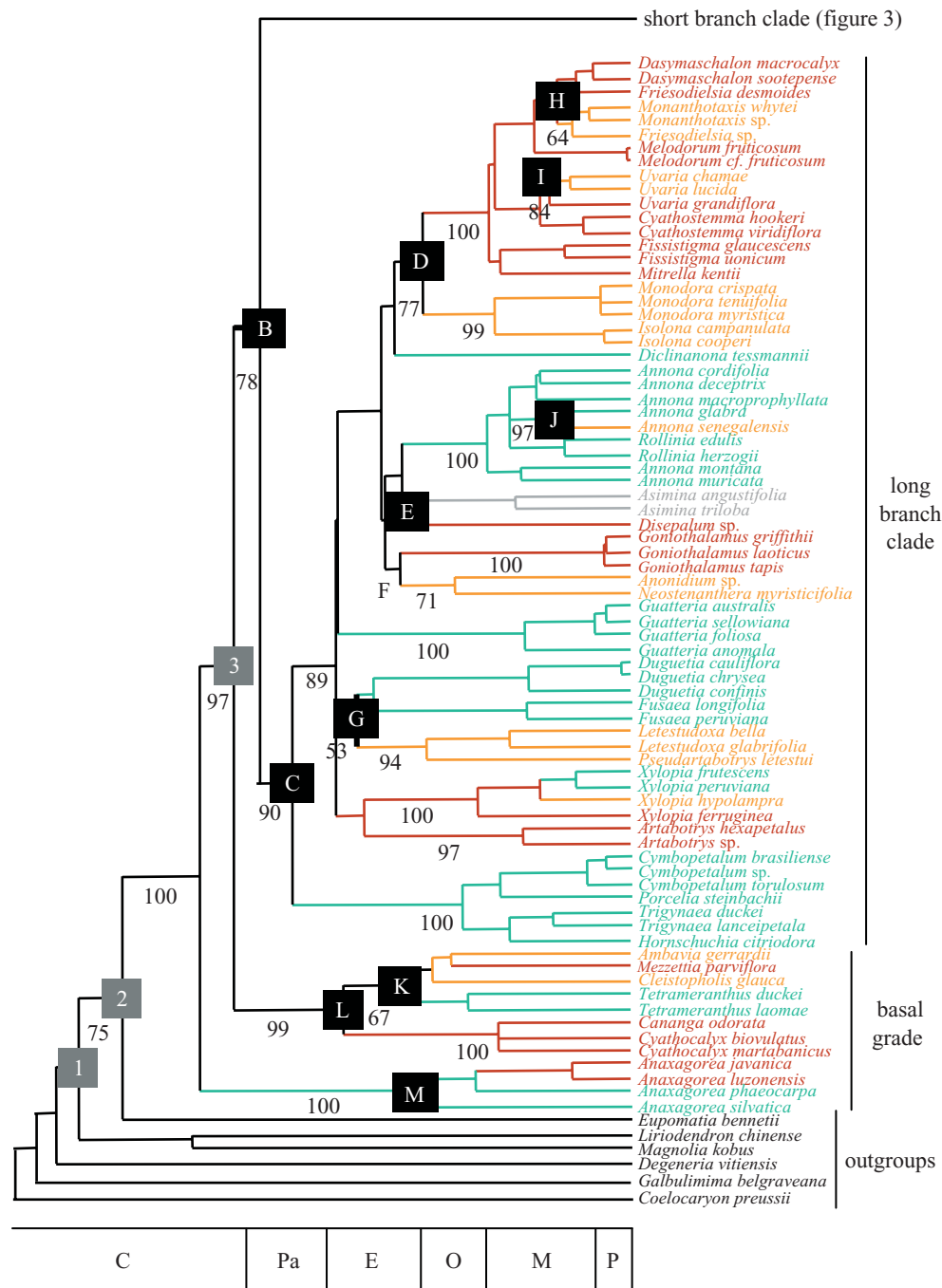


Figure 2. Detail of NPRS chronogram using calibration 1 (*Archaeanthus*) of the basal grade and long branch clade from one of the most parsimonious trees from the analysis of Annonaceae *rbcL* and *trnL-F* DNA sequence data. The three calibration points are indicated by grey boxes (*Archaeanthus* (1); Wikström stem Annonaceae dated at 82 Myr ago (2); and Maastrichtian seed (3)). Bootstrap values for key nodes with greater than 50% support are indicated below the branches. The dates of nodes are indicated in table 1. The line colour indicates geographical distribution: orange, Africa; red, Southeast Asia; green, South America; grey, North America. The time-scale represents the Cretaceous period (C) and Tertiary epochs (Pa, Palaeocene; E, Eocene; O, Oligocene; M, Miocene; P, Pliocene).

not account for these limits. The third calibration point used Maastrichtian seeds with a perichalazal ring and endosperm with lamelliform rumination (Chesters 1955) that may be contrasted with genera in the basal grade of Annonaceae that have seeds with irregular ruminations, this latter state is assumed to be ancestral. The lamelliform ruminations of the Nigerian Maastrichtian seeds (Chesters 1955) are similar to those found in most genera of the long branch clade and also some genera with similar ruminations are found in the short branch clade. Evolution of lamelliform ruminations could therefore have occurred at the stem of the long

and short branch clades as described in § 3, i.e. this node could be dated at between 65 and 70 Myr ago. We therefore applied a date of 68 Myr ago for this node. Maastrichtian reticulate monosulcate pollen from Colombia (Sole de Porta 1971) is reportedly similar to that of the malmeoid group of Walker (1971). However, precise placement of this fossil would require information on infratectal structure that is unavailable so it was not used as a calibration point.

For the Rhamnaceae data we calibrated the tree using dates of 62 and 64 Myr ago for the stem Rhamnaceae lineage based on age

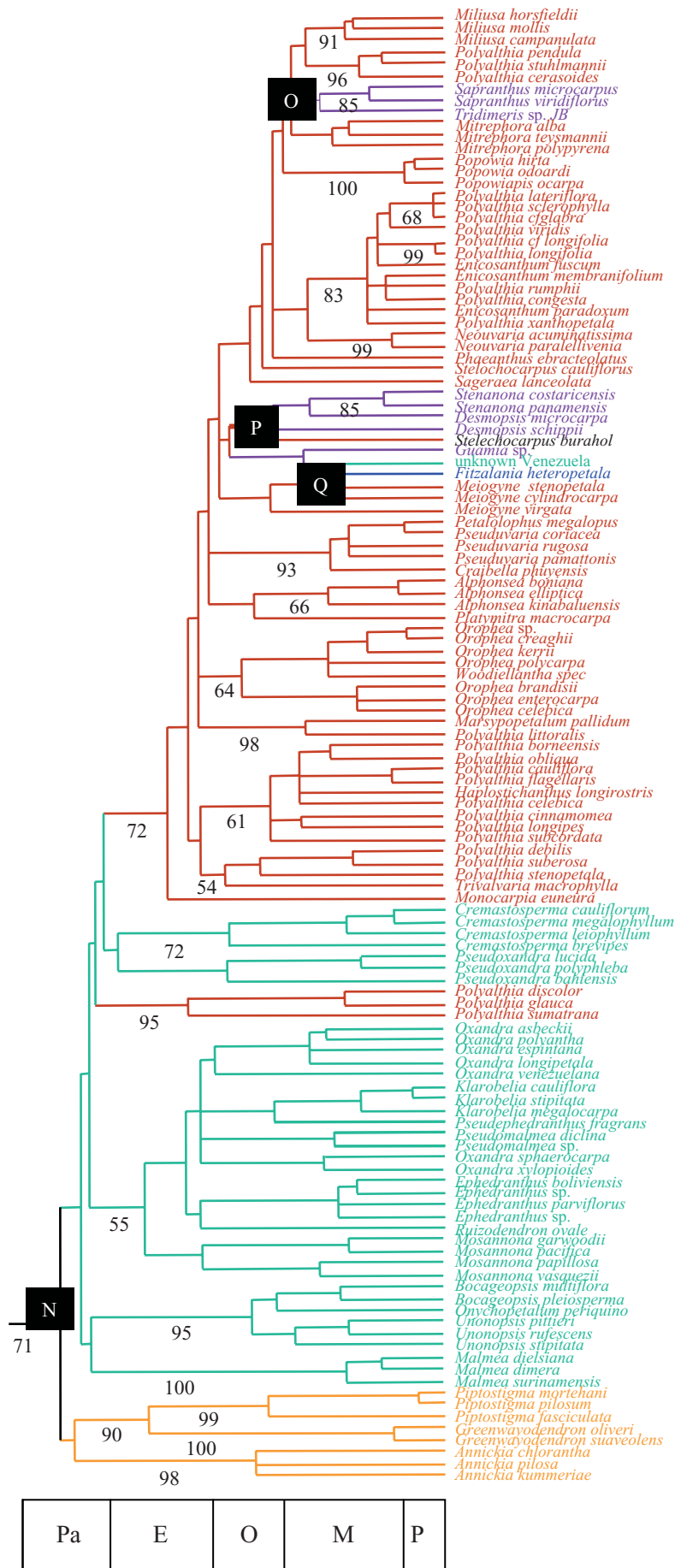


Figure 3. (Caption overleaf.)

Figure 3. Detail of NPRS chronogram using calibration 1 (*Archaeanthus*) of the short-branch clade from one of the most parsimonious trees from the analysis of Annonaceae *rbcL* and *trnL-F* DNA sequence data. The bootstrap values for key nodes with greater than 50% support are indicated below the branches. The dates of nodes are indicated in table 1. The line colour indicates geographical distribution: orange, Africa; red, Southeast Asia; green, South America; grey, North America; blue, Australia; purple, Central America. The time-scale represents Tertiary epochs (Pa, Palaeocene; E, Eocene; O, Oligocene; M, Miocene; P, Pliocene).

estimates for the family made by Wikström *et al.* (2001). These estimates had confidence intervals of  $\pm 3$  Myr but our calibrations did not account for these limits. The Rose Creek flower was originally ascribed to Rosaceae by Basinger & Dilcher (1984), but it could be a member of Rhamnaceae with obhapplostemonous flowers and seemingly rhamnaceous anther morphology. This fossil has been dated at 94 to 97 Myr ago, and if it is in fact Rhamnaceae, it establishes this as the minimum age for the family. However, this fossil was not used because we feel that its exact placement is still somewhat ambiguous and it could in fact represent an earlier diverging lineage within Rosales.

### 3. RESULTS

The Annonaceae parsimony search produced 5000 equally parsimonious trees of 2971 steps with CI = 0.494 and RI = 0.804. The molecular clock was rejected because the log likelihoods of the constrained and unconstrained analyses were significantly different (21 127.0 versus 21 681.6;  $p < 0.005$  respectively), so Sanderson's methods of NPRS (Sanderson 1997) and penalized likelihood (Sanderson 2002) were applied to produce ultrametric trees using the software package R8s. The time-scale bars shown on the figures are those obtained using NPRS and the *Archaeanthus* calibration point. Dates indicated in the text and the tables are those obtained from using NPRS and the younger Wikström *et al.* (2001) age estimate for Annonaceae (i.e. 82 Myr ago) and the *Archaeanthus* calibration points. Results from the penalized likelihood analysis are not shown because calibration using the older of these points with penalized likelihood usually yielded nodes with age estimates that fell within the confidence limits of the NPRS estimates. Those nodes for which penalized likelihood age estimates fell outside the confidence intervals of the NPRS estimates are indicated in the text. Using the third calibration point gave dates for nodes that were between those from the first and second calibrations and these results are also not shown.

Figure 1 shows one of the most parsimonious trees from the analysis of the Annonaceae data. The outgroups, basal grade and long branch clade are shown in more detail in figure 2. *Eupomatia* (Eupomatiaceae) from New Guinea and eastern Australia is a sister to Annonaceae with moderate bootstrap support. Annonaceae are a strongly supported monophyletic group. The genus *Anaxagorea* is a sister to the rest of Annonaceae and is part of a basal grade that also includes *Ambavia*, *Mezzetia*, *Cleistopholis*, *Tetrameranthus*, *Cananga* and *Cyathocalyx* (figure 2). Derived from within this grade is a strongly supported clade, the crown node of which is estimated as  $56.6\text{--}66.7 \pm 2.3$  Myr ago (figures 1 and 2, node B), consisting of a strongly supported long branch clade and a weakly supported short

branch clade. The crown of the long branch clade (shown in more detail in figure 2) is estimated at  $51.1\text{--}60.2 \pm 2.3$  Myr ago (node C) and that of the short branch clade (shown in more detail in figure 3) is  $53.1\text{--}62.5 \pm 3.6$  Myr ago (node N). Key nodes are indicated on the figures and their dates with standard deviations are indicated in table 1. Table 2 indicates the estimated dates for the diversification of species rich genera (i.e. those with more than 100 species).

The Rhamnaceae parsimony search produced 5000 trees with a length of 3023 steps, CI = 0.557 and RI = 0.690. For the Rhamnaceae dataset the molecular clock was rejected because the likelihoods of the constrained and unconstrained analyses were significantly different (4905.3 versus 4827.3;  $p < 0.005$ ), so Sanderson's (1997) methods of NPRS and penalized likelihood (Sanderson 2002) were applied to produce an ultrametric tree using R8s.

Figure 4 shows one of the most parsimonious trees from the analysis of the Rhamnaceae data. Rhamnaceae are strongly supported as monophyletic, with a clade consisting of Dirachmaceae and Elaeagnaceae forming their sister group. Within Rhamnaceae, strongly bootstrap-supported major groups are identified: a ziziphoid group; an ampeloziziphoid group that contains the genera *Ampeloziziphus*, *Doerpfeldia* and *Bathiorhamnus*; and a rhamnoid group. Other strongly supported groups within these larger groups include: (i) in the ziziphoid group: Pomaderreae, *Ceanothus*, Phyliceae, *Colubrina*, Colletieae, Paliureae and Gouanieae, and (ii) in the rhamnoid group: Rhamneae with the monotypic tribe Maesopsidae being a sister to Rhamneae and finally Ventilagineae. Estimated dates using both calibration points with NPRS and confidence intervals for key nodes are indicated in tables 3 and 4. The results from the penalized likelihood analysis are not shown because penalized likelihood usually gave age estimates that were within those estimated using NPRS. Any deviations in ages obtained using penalized likelihood that fell outside the confidence limits of the NPRS estimates are indicated in the text.

### 4. DISCUSSION

#### (a) *Biogeographic history of Annonaceae*

The oldest remains of Annonaceae are seeds with a perichalazal ring and endosperm with lamelliform rumination that were found in the Maastrichtian of Nigeria (Chesters 1955) and pollen from Colombia (Sole de Porta 1971). Other evidence for a west Gondwanan (i.e. formerly joined landmasses of South America and Africa) origin of Annonaceae includes the fact that the more primitive pollen types are found there, whereas most Asian taxa have more derived pollen morphology (Walker 1971; Raven & Axelrod 1974; Doyle & Le Thomas 1997). Other evidence against a Laurasian origin includes the fact that no fossils have been found from Cretaceous floras of Laurasia that can unambiguously be assigned to Annonaceae. A biogeographical analysis of Annonaceae by Doyle *et al.* (2004) indicate that when *Anaxagorea* is treated as being of South American origin (as indicated by our results) then the ancestral area of the family is either South American or African. The present level of sampling for the molecular tree does not permit determination of the precise origin of the family; however, most other lines of evidence point to an origin in west Gondwana.

The estimated age of stem Annonaceae according to Wikström *et al.* (2001) is between 82 and 91 Myr ago and this date is supported by that of the family ( $90.6 \pm 1.3$  Myr ago; figure 1, node A) that is obtained when calibrating the tree using *Archaeanthus* placed at the stem of Magnoliaceae. These age estimates for Annonaceae are older than those indicated by the fossil record. However, these estimates are still too young for most disjunctions within the family to be explained by continental drift. The comparatively young age of Annonaceae may be contrasted with dates obtained for Lauraceae by Chanderbali *et al.* (2001) of *ca.*  $174 \pm 32$  Myr ago, which is a family that includes Southern Hemispheric lineages showing strong evidence of Gondwanan vicariance dating from *ca.* 90 Myr ago.

According to Raven & Axelrod (1974), migration between Africa and South America could still have been possible after their break-up 90–105 Myr ago, by short-distance over-water dispersal and up until the Maastrichtian via 'filter dispersal' through island chains of the Rio Grande Rise–Walvis Ridge and the Sierra Leone Rise (Morley 2000, 2003). The timings of some splits, such as *Anaxagorea* from the rest of Annonaceae (64.9–76.8 Myr ago) and between the basal grade and the long or short branch clade (59.8–70.6 Myr ago), and the long branch–short branch split that occurred  $56.6\text{--}66.7 \pm 2.3$  Myr ago (figures 1 and 2, node B) according to our estimates could be consistent with either cessation of short-distance over-water dispersal that was possible for some time after the continents diverged, or they could be the result of break-up of the Rio Grande Rise–Walvis Ridge or the Sierra Leone Rise migration paths. These dates are consistent with Doyle & Le Thomas' (1997) suggestion that these lines radiated in the Late Cretaceous when migration might still have been possible between Africa and South America.

The young estimates for the dates of divergence that we obtain for most lineages within Annonaceae with sister taxa that are distributed on previously joined Gondwanan landmasses or in Southeast Asia require an alternative explanation. Migration between Gondwana and Laurasia in the Late Cretaceous–Early Tertiary could have been possible as the Tethys narrowed and India approached Asia. India may have acted as a raft that transported some taxa from Madagascar to Asia (Le Thomas & Doyle 1996). North and South American exchange could have occurred via a fragmented migrational pathway that may have existed through the Greater and Lesser Antilles in the Middle to Late Eocene (Pindell *et al.* 1988; Pindell & Barrett 1990; Iturralde-Vinent & MacPhee 1999). Doyle & Le Thomas (1997) suggested that a boreotropical distribution could have been attained when lines originating in northern Gondwana crossed into Laurasia and became established along the southern coast of Eurasia that had a tropical climate in the Eocene. Annonaceae fossils are also well represented in the Eocene of England (Chandler 1964) and Argentina (Menendez 1972), and Palaeocene Egypt (Chandler 1954) and Colombia (Van der Hammen & García de Mutis 1966). The diverse seeds of Annonaceae in the Early Eocene London clay flora show many affinities with present-day Southeast Asia. This flora has all the types of seed endosperm rumination observed in the family today (Chandler 1964). Little *et al.* (2002) have also reported the presence of Annonaceae in the Appian Way Locality

(Late Eocene) of Vancouver Island, British Columbia. Annonaceae had radiated extensively in tropical Asia by the Palaeogene according to Raven & Axelrod (1974) and our dates support radiation in Asia during this period (dates of the crown of some of the Asian lineages in the short and long branch clades are consistent with Palaeogene radiation; figures 2 and 3).

Post-Eocene cooling would result in disjunct distributions of taxa between tropical Africa, Asia and the New World (see fig. 6 of Pennington & Dick 2004). The *Tetrameranthus–Cleistopholis* ( $35.5\text{--}41.3 \pm 5.6$  Myr ago; figure 2, node K) and *Duguetia–Letestudoxa* New World–Old World splits ( $40.0\text{--}41.7 \pm 3.2$  Myr ago; figure 2, node G) and the *Dasymaschalon–Monodora* ( $31.6\text{--}37.2 \pm 3.3$  Myr ago; figure 2, node D) and *Goniothalamus–Neostenanthera* African–Southeast Asian ( $34.9\text{--}41.1 \pm 2.8$  Myr ago; figure 2, node F) estimated splits (figure 2) could be the result of disruption of boreotropical vegetation. *Asimina* from North America could also be part of this pattern (split from Asian *Disepalum*  $33.8\text{--}39.8 \pm 7.2$  Myr ago; figure 2, node E) and this estimate for the age of the split of *Asimina* from its MRCA correlates well with the age of Eocene–Oligocene fossil pollen attributed to *Asimina*. Davis *et al.* (2002a); Chanderbali *et al.* (2001) and Renner *et al.* (2001) also indicate groups in Malpighiaceae, Lauraceae and Melastomataceae *s.str.* respectively, with tropical amphiatlantic disjunctions that split up at a time consistent with the disruption of boreotropical ranges around the Eocene–Oligocene boundary. Fritsch (2001) provided more support for the boreotropics hypothesis in his study on the biogeography of the genus *Styrax*, although he did not calibrate phylogenies to determine the age of amphiatlantic splits.

The paucity of Annonaceae in Australia provides further support for migration after Gondwanan break-up. The nesting of *Fitzalania*, one of only two Australian endemic genera, within the southeast Asian clade and the estimated date of this group at *ca.*  $18.1\text{--}19.9 \pm 5.7$  Myr ago (figure 3, node Q) indicates that Annonaceae migrated from Southeast Asia into Australia comparatively recently. Migration from South America to Australia through the Antarctic was still possible up until *ca.* 40 Myr ago but this route was evidently not taken by Annonaceae and seems an unlikely migration path for megathermal taxa given the probable climatic conditions.

Figure 2 indicates examples of long-distance dispersal between South American, Southeast Asian and African tropical regions. The African *Monanthotaxis–Friesodielsia* lineage (*ca.* 70 species) is nested within a Southeast Asian clade and is dated at an estimated  $9.0\text{--}10.6 \pm 2.2$  Myr ago (figure 2, node H), indicating that it arrived by long-distance dispersal. Similarly, according to our estimates, *Uvaria* dispersed from Southeast Asia to Africa (where there are *ca.* 70 species)  $12.4\text{--}14.5 \pm 2.1$  Myr ago (figure 2, node I) and *Annona senegalensis* from South America to Africa  $13.9\text{--}16.3 \pm 3.2$  Myr ago (figure 2, node J). If these African lineages prove on further sampling to be monophyletic, then a substantial proportion of African species (*ca.* 20%) are comparatively recently derived as a result of radiation following long-distance dispersal. Long-distance dispersal may also account for short branch clade central American taxa nested within a southeast Asian clade that dispersed *ca.*  $24.9\text{--}30.8$  Myr ago (figure 3, nodes O and P),

Table 1. Age estimates for key events in the history of Annonaceae based on the branch lengths observed in the NPRS tree (figures 2 and 3), under the GTR +  $P_{inv}$  + G model using two calibration points: calibration 1 (based on 98 Myr ago for *Archaeanthus*), and calibration 2 (based on the younger Wikström age of the Annonaceae stem of 82 Myr ago). (s.d. was calculated as described in § 2.)

node	calibration 1	calibration 2	s.d.
A. Annonaceae	90.6	82	1.3
B. Long branch–short branch split	66.7	56.6	2.3
C. Long branch crown	60.2	51.1	2.3
D. <i>Dasydaschalon</i> – <i>Monodora</i>	37.2	31.6	3.3
E. <i>Asimina</i> – <i>Disepalum</i>	39.8	33.8	7.2
F. <i>Goniothalamus</i> – <i>Neostenanthera</i>	41.1	34.9	2.8
G. <i>Duguetia</i> – <i>Letestudoxa</i>	40.0	41.7	3.2
H. <i>Monanthotaxis</i> clade dispersal	10.6	9.0	2.2
I. <i>Uvaria</i> dispersal	14.5	12.4	2.1
J. <i>Annona</i> dispersal	16.3	13.9	3.2
K. <i>Tetrameranthus</i> – <i>Cleistopholis</i>	41.3	35.5	5.6
L. <i>Tetrameranthus</i> – <i>Cyathocalyx</i>	51.3	43.8	5.1
M. <i>Anaxagorea</i> crown	38.5	33.6	6.3
N. Short branch crown	62.5	53.1	3.6
O. <i>Tridimeris</i> clade dispersal	26.5	24.9	4.2
P. <i>Stelechocarpus</i> clade dispersal	30.8	27.7	6.3
Q. <i>Fitzalania</i> – <i>Meiogyne</i>	19.9	18.1	5.7

Table 2. Geographical distribution and age estimates for diversification (i.e. crown) of species-rich genera (more than 100 species) in Annonaceae.

(Two calibration points were used: calibration 1 (based on *Archaeanthus*) and calibration 2 (based on the younger Wikström age of the Annonaceae stem of 82 Myr ago). s.d. was calculated as described in § 2.)

	distribution	number of species	calibration 1	calibration 2	s.d.
<i>Xylopia</i>	pantropical	100	27.5	23.8	3.7
<i>Guatteria</i>	neotropical	250	19.1	16.3	3.3
<i>Annona</i>	tropical America and Africa (one species)	100	25.6	21.8	3.8
<i>Uvaria</i>	palaeotropics	100	14.8	12.4	2.4
<i>Goniothalamus</i>	Indomalesia	115	4.8	3.6	1.5
<i>Artabotrys</i>	palaeotropics	100	19.2	16.4	3.2

although this result must be interpreted with caution given the lack of support within the short branch clade. *Anaxagorea* from South America and southeast Asia could also have attained its current distribution more recently through long-distance dispersal (figure 2), as could *Xylopia*, a pantropical genus with a crown age estimated here as  $23.8\text{--}27.5 \pm 3.7$  Myr ago (figure 2). It is also possible that these genera may have attained their current distributions by migration through the boreotropics. Renner *et al.* (2001) demonstrated that palaeotropical Melastomeae form a clade that is deeply embedded in neotropical Melastomeae and is between 12 and 14 Myr old, indicating that West Africa was reached from the neotropics via long-distance dispersal. The only Asian genera of Melastomeae probably derived from long-distance dispersal from African or Madagascan ancestors, and two sub-lineages of Dissochaeteae–Sonerileae reached Madagascar by long-distance dispersal in the Late Miocene. Other examples of recent long-distance dispersal between tropical regions include *Acridocarpus austrocaledonicus*, which dispersed from Madagascar to New Caledonia *ca.* 8 Myr ago (Davis *et al.* 2002b), *Andira inermis* from South America to Africa (Pennington 2003) and *Symphonia globulifera* attained its current neotropical distribution by transoceanic dispersal from Africa *ca.* 17 Myr ago (Dick *et al.* 2003).

The molecular and fossil evidence indicate that the current distribution patterns of Annonaceae may be best explained by an origin in western Gondwana that was followed by interchange between Africa and South America by short-distance dispersal across the opening Atlantic, dispersal via the Walvis Ridge or, more likely, because of more suitable climate and shorter distances at lower latitudes, the Sierra Leone Rise. This could have been followed by migration into Laurasia during the EECO, resulting in a boreotropical distribution that was then disrupted by cooling. This scenario is largely congruent with that suggested by Doyle & Le Thomas (1997). Movement between continents after this period may have been restricted to rare long-distance dispersal events in the Neogene. The high level of continental generic endemism in Annonaceae with most genera either palaeotropical or neotropical suggests a high degree of geographical structure and also suggests that long-distance trans-oceanic dispersal has had a relatively minor role in determining present distribution patterns, which is not surprising given that the primary dispersal agents for Annonaceae are mammals. However, long-distance dispersal does occur, as evidenced by the molecular trees and the presence of Annonaceae on volcanic islands in the Lesser Antilles, indicating that propagules may be transported across water



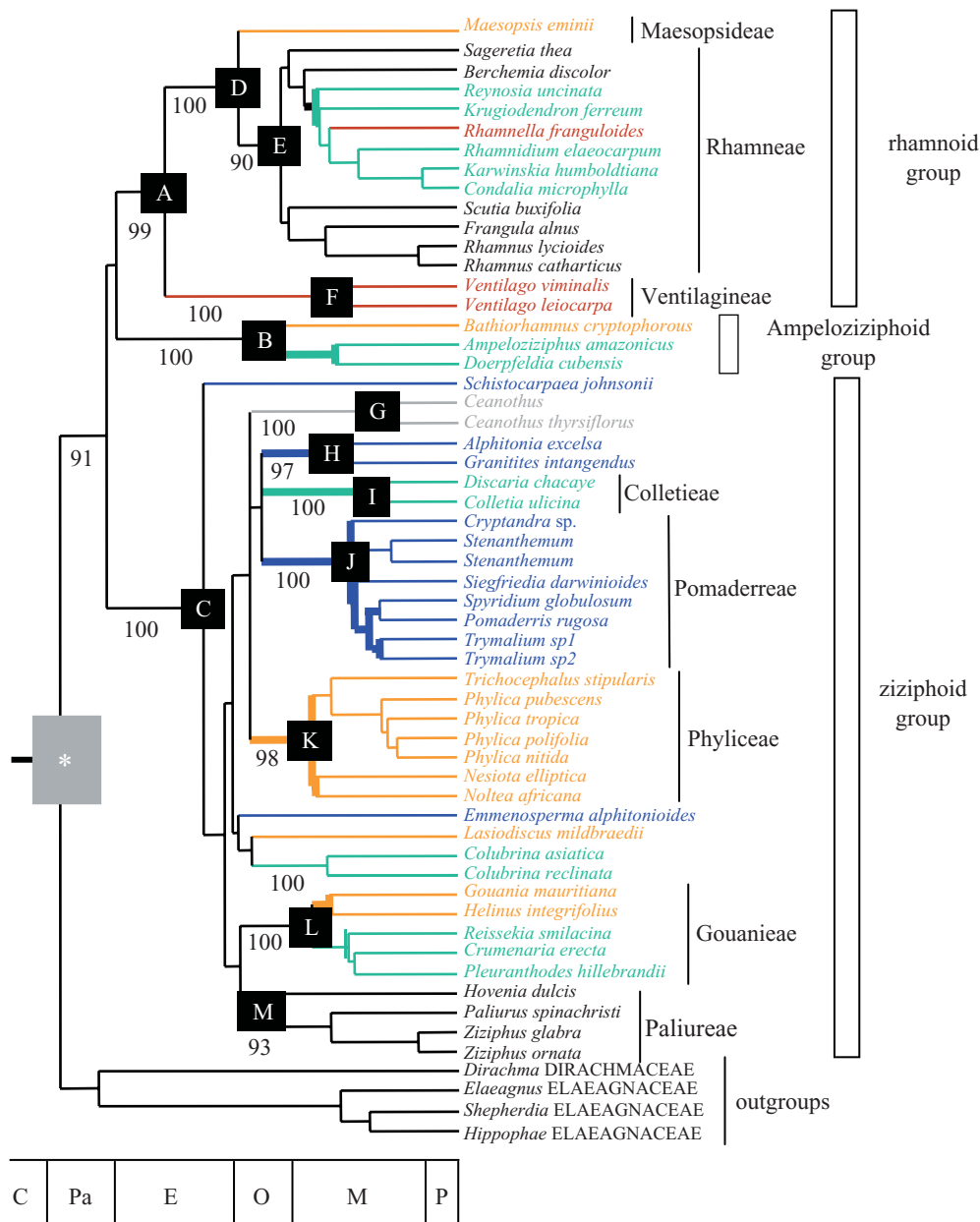


Figure 4. NPRS chronogram calibrated with the Rhamnaceae stem dated at 64 Myr ago from the analysis of *rbcL* and *trnL-F* DNA sequence data. The calibration point is indicated with an asterisk in a grey box. The bootstrap values for key nodes with greater than 50% support are indicated below the branches. The dates for nodes A–M are indicated in table 3. The line colour indicates geographical distribution: orange, Africa; red, Southeast Asia; green, South America; grey, North America; blue, Australia; black, broad distribution. The time-scale represents the Cretaceous period (C) and Tertiary epochs (Pa, Palaeocene; E, Eocene; O, Oligocene; M, Miocene; P, Pliocene).

barriers. Long-distance dispersal into Africa, though rare, has resulted in diversifications that could account for ca. 20% of Annonaceae species diversity on this continent.

Diversification of species-rich lineages in tropical rainforests had been attributed to gradual accumulation of species over a long geological period in stable equatorial climates (Stebbins 1974). However, Late Tertiary geological events and unstable Pleistocene climates may also have played a part in the generation of species diversity more recently (Gentry 1982; Haffer 1982; Whitmore & Prance 1987). DNA sequence data are consistent with recent diversification in *Inga*, a species-rich genus of neotropical trees in which speciation was concentrated in the past 10 Myr, with many species arising as recently as 2 Myr ago

(Richardson *et al.* 2001a). This coincides with major uplifts of the Andes, the bridging of the isthmus of Panama, and Quaternary glacial cycles. Table 2 shows our ages for the diversification of species-rich genera (i.e. those with more than 100 species) in Annonaceae. The most rapid radiation seems to have taken place in the Southeast Asian genus *Goniothalamus* (115 spp.), whose crown is estimated at  $3.6\text{--}4.8 \pm 1.5$  Myr ago. Other genera diversify from between 12.4 and 27.5 Myr ago, although further sampling could add more basal lineages and increase the crown ages of some of them. However, given this level of sampling it would seem that molecular data indicate comparatively recent diversification in species-rich lineages in Annonaceae.

Table 3. Age estimates for key events in the history of Rhamnaceae based on the branch lengths observed in the NPRS tree (figure 4) under the GTR +  $P_{inv}$  + G model using two calibration points: stem Rhamnaceae, 62 Myr ago; and stem Rhamnaceae, 64 Myr ago. (s.d. was calculated as explained in § 2.)

node	calibration	calibration	s.d.
	(62 Myr ago) NPRS	(64 Myr ago) NPRS	
A. Rhamnoid crown	45.8	47.3	2.2
B. Ampeloziziphoid crown	31.8	32.8	2.9
C. Ziziphoid crown	39.6	40.9	2.4
D. Rhamneae- <i>Maesopsis</i>	34.2	35.3	2.7
E. Rhamneae crown	27.6	28.5	2.7
F. <i>Ventilago</i> crown	19.4	20.0	3.0
G. <i>Ceanothus</i> crown	10.7	11.1	4.2
H. <i>Granitites</i> - <i>Alphitonia</i>	18.4	19.0	2.5
I. Colletieae crown	14.1	14.6	3.0
J. Pomaderreae crown	16.9	17.4	2.6
K. Phyliceae crown	22.9	23.6	3.1
L. Gouanieae crown	22.7	23.5	2.5
M. Paliureae crown	30.6	31.6	3.1

#### (b) *Biogeographic history of Rhamnaceae*

The distribution of Rhamnaceae suggests that patterns were strongly influenced by Gondwanan break-up. The ampeloziziphoid group illustrates a pattern of disjunction between northern South America and Madagascar also found in other groups (e.g. *Bixa* and *Diegodendron* (Fay *et al.* 1998) and Mimosoideae (Hughes *et al.* 2003)). The ziziphoid group is cosmopolitan with a predominantly Southern Hemisphere distribution (with the exception of *Ceanothus*, which has a North American distribution), suggesting a possible Gondwanan origin. This distribution suggests that this whole southern group was widespread throughout Gondwanaland and parts of Laurasia (in what is now North America) before these continents broke up and subsequently contracted, largely to the Southern Hemisphere. Genera and tribes within this group are usually confined to a particular continent (except *Colubrina*, Paliureae and Gouanieae). The rhamnoid group is also widespread, with Ventilagineae found in India and Southeast Asia, Maesopideae in Africa, and Rhamneae with a wide distribution throughout the tropics, sub-tropics and into temperate regions.

The age of stem Rhamnaceae determined by Wikström *et al.* (2001) is too young for continental drift to have affected patterns in its current distribution. This is supported by the fact that calibration of the Rhamnaceae tree using the Wikström *et al.* dates gives estimated dates of 22.9–23.6 ± 3.1 Myr ago for Phyliceae (figure 4, node K) in comparison with dates of 15–18 Myr ago estimated for this clade by Richardson *et al.* (2001b), who used an independent calibration based on dispersal of species of Phyliceae to volcanic oceanic islands of known age. The ziziphoid group includes a number of lineages whose relationships are poorly resolved and that, according to our estimates, diverged *ca.* 39.6–40.9 ± 2.4 Myr ago (figure 4, node C). Australian–Antarctic–South American links were

possible via a southern temperate route up until *ca.* 40 Myr ago, Madrean–Tethyan flora links were maintained from the Late Eocene until the end of the Oligocene and links through GAARLandia could have allowed migration between Africa and North and South America (33–35 Myr ago). Severing of these links would have resulted in splitting of the ziziphoid group between 30 and 40 Myr ago and the dates we obtain here are consistent with this. The Rhamneae–*Maesopsis* split (*ca.* 34.2–35.3 ± 2.7 Myr ago; figure 4, node D; penalized likelihood gave a date of 27.7 Myr ago) in the rhamnoid clade and the New World–African split in the Ampeloziziphoid group (*ca.* 31.8–32.8 ± 2.9 Myr ago; figure 4, node B) are also consistent with these dates. However, these distribution patterns may in fact be better explained by long-distance dispersal. A number of genera with widespread distributions are found in Rhamnaceae (*Ziziphus*, *Rhamnus*, *Berchemia*, *Gouania*, *Sageretia*, *Colubrina*, *Scutia*: table 4). More species in each of these genera need to be sequenced to make a fine-scale biogeographic assessment; however, given the estimated ages of the stems of each of these genera (between 19.6 and 33.4 Myr ago; table 4; penalized likelihood gave dates of between 16.9 and 34.3 Myr ago), it would seem that none of the migration pathways suggested above would have been available at these times, and therefore long-distance dispersal is a more feasible explanation for how they attained their current distributions. It is clear from the modern distribution of particular groups in Rhamnaceae that fruits or seeds are very effectively dispersed even though propagule morphology is not typical of widely dispersed taxa. For example, species of the tribe Phyliceae have dispersed to oceanic islands around Africa on several occasions (Richardson *et al.* 2001b). The dispersal capability of Gouanieae is indicated by the Hawaiian endemic genus *Pleuranthodes*. *Colubrina* is predominantly found in central South America and the Caribbean, although species are also found in Asia, Hawaii, Madagascar and South Africa. Our estimates indicate that *Colubrina* diverged from its MRCA 32.3–33.4 ± 2.7 Myr ago (this date matches the age of *Colubrina* fossil material in Colorado *ca.* 34 Myr ago), and although we have only two species, the age of crown *Colubrina* is *ca.* 20.3–21.0 Myr ago. This date also indicates that *Colubrina* attained its modern distribution by long-distance dispersal. The dispersal capabilities of species in this genus are clearly given the distribution of *C. asiatica* that is found in coastal regions all around the Pacific rim.

Our estimates for the time at which *Paliurus* split from *Hovenia* (30.6–31.6 ± 3.1 Myr ago; figure 4, node M) are a good match for the age of the oldest fossil of *Hovenia* (32.3–33.6 Myr ago; R. E. Dunn, personal communication). *Ziziphus*, as outlined by Liu & Cheng (1995), is widespread in tropical and sub-tropical regions (Asia, Africa, North America, South America, Oceania and Europe, with the centre of diversity in Asia) and the crown group age estimate is 6.2–6.4 Myr ago. The distribution of the sister taxa of *Ziziphus* in eastern Asia and the fact that the greatest species diversity is in Asia and that it and/or its sisters are well represented in the Late Eocene–Miocene of North America and Eurasia, suggests a Laurasian origin for this group followed by more recent migration into South America and Africa.

Table 4. Age estimates for the stems of widespread genera in Rhamnaceae.

	distribution	number of species	stem age based on calibrations of stem Rhamnaceae at 62 and 64 Myr ago (s.d.) using NPRS
<i>Ziziphus</i>	Asia, Africa, North America, South America, Oceania, Europe	150	19.6–20.2 (4.2)
<i>Rhamnus</i> (including <i>Frangula</i> )	Northern Hemisphere to Brazil and South Africa	125	26.5–27.4 (2.6)
<i>Berchemia</i>	East Africa, East Asia, western North America	12	24.0–24.8 (2.5)
<i>Gouania</i>	Americas (15), Africa (2), Madagascar and Indian Ocean (5), Asia (4)	26	20.1–20.7 (2.5)
<i>Sageretia</i>	Somalia and southwest Asia to Taiwan, tropical and warm Americas	35	26.5–27.3 (3.9)
<i>Colubrina</i>	tropical and warm especially Americas	31	32.3–33.4 (2.7)
<i>Scutia</i>	neotropical, palaeotropical and to South Africa	3	26.5–27.4 (2.6)

The distribution of Ventilagineae is Old World tropics with a centre of diversity in India. The Mid-Eocene split of Ventilagineae from its MRCA is not consistent with isolation on India as the sub-continent split from eastern Gondwana. However, these dates are consistent with dispersal from Africa to India as the latter migrated north, followed by diversification from *ca.* 19.4–20.0 ± 3.0 Myr ago (figure 4, node F) in India and Southeast Asia. The possibility of continued exchange between India and Africa as India moved north was suggested by (McKenna 1973). More sampling is necessary to clarify the biogeographic origins and patterns within this group.

The ziziphoid group contains a number of species-rich lineages that have diversified in Mediterranean biomes. That diversification of one particular lineage within Rhamnaceae coincides with the Miocene development of Mediterranean climate systems has been demonstrated by Richardson *et al.* (2001b). According to the *rbcL* and *trnL-F* data the lineage leading to *Noltea* diverged from its common ancestor with the rest of Phyliceae, 22.9–23.6 ± 3.1 Myr ago (figure 4, node K) according to our estimates. This is slightly older than the age estimate of 15–18 Myr ago based on an independent calibration using the dispersal of *Phylica* species to volcanic oceanic islands of known age, as estimated by Richardson *et al.* (2001b). However, this age is still consistent with divergence of *Noltea* from the rest of Phyliceae before extensive aridification of southwestern Africa began. *Noltea* does not have any of the adaptations to an arid environment that other species in the tribe have, and a *Noltea*-like ancestor could have been more widespread throughout the Cape region in the area now covered by fynbos when the vegetation consisted of wet, sub-tropical forests. Plant groups that made up this flora either no longer occur in Africa or exist only in refugia such as in the coastal forest vegetation of the southern Cape, where *Noltea* is presently found (see also the relict taxa found in mesic regions as indicated by Linder & Hardy (2004)). Richardson *et al.* (2001b) have shown that *Nesiota elliptica*, an endemic genus from St Helena, diverged at a time when morphological adaptations to an arid climate, such as those exhibited by this species (leaves with tomentose undersurfaces), might have occurred. *Phylica* species have an even more highly derived vegetative morphology (narrow leaves with tomentose undersurfaces) and this

genus began diversifying in the Late Miocene in the Cape at a time that coincides with the onset of extensive aridification in this region. *Alphitonia* together with *Granitites* and *Pomadereae* are Australasian taxa, representing isolated ziziphoid clades. Diversification in *Pomadereae* commenced from *ca.* 16.9–17.4 ± 2.6 Myr ago (figure 4, node J). *Granitites* diverged from *Alphitonia* at an estimated 18.4–19.0 ± 2.5 Myr ago (figure 4, node H) and is a component of the southwest Australian flora that may be an example of a relict of rainforest lineages that dominated the region in Cretaceous–Tertiary times (Hopper *et al.* 1996; Hill *et al.* 1999). The six species of the rainforest lineage *Alphitonia* are from Malesia, tropical Australia and the west Pacific. Diversification of *Pomadereae* and divergence of *Granitites* from *Alphitonia* coincides with aridification in southwest Australia. The Neogene climatic histories of the Cape Floristic Region and southwest Australia appear to have been similar, with Mid-Tertiary evergreen sub-tropical forests being replaced by heathy vegetation in the Late Miocene or Early Pliocene (Coetzee 1978; Truswell 1990; Hill *et al.* 1999) because of the onset of summer dry climates that resulted from the opening up of the Drake Passage (Siesser 1980). The timing of diversification of Phyliceae, *Pomadereae* and the *Alphitonia/Granitites* split coincides with these changes. Crisp *et al.* (2004) also document examples of radiations in similar time frames in a number of Australian angiosperm lineages that are found in sclerophyll communities. Calsbeek *et al.* (2003) used the rate of substitution of internal transcribed spacers of nuclear ribosomal DNA estimated by Richardson *et al.* (2001b) to estimate that diversification in *Ceanothus* commenced from *ca.* 16 Myr ago and coincided with the development of a summer dry climate in the Californian Floristic Province. This date correlates well with the estimates we obtain here (10.7–11.1 ± 4.2 Myr ago; figure 4, node G) that were based on a much smaller sample of the genus. Colletieae consists of *ca.* 40 species that are mostly found in regions of South America that have an arid climate. The timing of diversification of the crown of this predominantly xerophytic tribe (estimated at 14.1–14.6 ± 3.0 Myr ago; figure 4, node I) coincides with the diversification of other lineages found in Mediterranean climate zones.

Migration of Rhamnaceae between regions with Mediterranean climate might have been more possible up to ca. 30–40 Myr ago, after which routes overland or short-distance dispersal over water were severed. These Mediterranean biome 'islands' would then have become increasingly isolated and protected from a large influx of immigrants by long-distance oceanic barriers and the fact that these biomes occupy small areas. *In situ* endemic diversification with low rates of immigration could then have given rise to geographical structure as suggested by Lavin *et al.* (2004). The biogeographic patterns that result from continental vicariance or the break-up of migration pathways will be lost if frequent long-distance transoceanic dispersal occurs, as is the case in Fabaceae (Lavin *et al.* 2004). A better explanation for patterns in the phylogeny of legumes might be that the ecological setting and the amount of long-distance dispersal between habitats and the influence of these factors on neutral ecological processes determine the phylogenetic pattern (Hubbell 2001; Lavin *et al.* 2004; Schrire *et al.* 2004). The frequency of long-distance dispersal seen in Rhamnaceae seems to be similar to that found in legumes, and patterns in the Rhamnaceae phylogeny (particularly in the ziziphoid clade) could also be explained by the processes described by Lavin *et al.* (2004) and Schrire *et al.* (2004).

## 5. CONCLUSIONS

Current patterns in the diversity and distribution of Rhamnaceae and Annonaceae are explained by migration and dispersal that occurred predominantly after the break-up of Gondwanaland and also comparatively recent diversification of species-rich lineages. The combined evidence of molecular phylogenetics and the fossil record indicate that dispersal routes for Annonaceae, which are megathermal plants, may correspond closely to those predicted by Morley (2003). What is also evident from the molecular studies conducted so far is that long-distance and stepping-stone dispersal has played a greater role in the development of modern patterns than had previously been assumed. More molecular phylogenetic studies and continued refinements in the use of molecular data in combination with palaeobotanical, palaeoclimatological and geological data, such as those that have been outlined in this volume (e.g. Near & Sanderson 2004; Renner 2004), will further improve our understanding of how the Earth's major biomes evolved.

We thank P. C. van Welzen, R. T. Pennington and two anonymous reviewers for comments and helpful suggestions that improved this manuscript, and J. A. Doyle for providing access to an unpublished manuscript. The authors gratefully acknowledge Paul Bygrave at RBG Kew for generating some of the sequence data.

## REFERENCES

- Angiosperm Phylogeny Group 1998 An ordinal classification for the families of flowering plants. *Ann. Miss. Bot. Gard.* **85**, 531–553.
- Axelrod, D. I. 1975 Evolution and biogeography of Madrean–Tethyan sclerophyll vegetation. *Ann. Miss. Bot. Gard.* **62**, 280–334.
- Axelrod, D. I. & Raven, P. H. 1978 Late Cretaceous and Tertiary vegetation history of Africa. In *Biogeography and ecology of southern Africa* (ed. M. J. A. Werger), pp. 77–130. The Hague: Junk.
- Basinger, J. & Dilcher, D. L. 1984 Ancient bisexual flowers. *Science* **224**, 511–513.
- Bermingham, E. & Martin, A. P. 1998 Comparative mtDNA phylogeography of neotropical freshwater fishes: testing shared history to infer the evolutionary landscape of lower Central America. *Mol. Ecol.* **7**, 499–517.
- Burnham, R. J. & Johnson, K. R. 2004 South American palaeobotany and the origins of neotropical rainforests. *Phil. Trans. R. Soc. Lond. B* **359**, 1595–1610. (doi:10.1098/rstb.2004.1531)
- Calsbeek, R. G., Thompson, J. N. & Richardson, J. E. 2003 Patterns of molecular evolution and diversification in a biodiversity hotspot: the California Floristic Province. *Mol. Ecol.* **12**, 1021–1029.
- Campbell, N. A. 1996 *Biology*. Menlo Park, CA: The Benjamin/Cummings Publishing Company Inc.
- Chanderbali, A. S., Van der Werff, H. & Renner, S. S. 2001 Phylogeny and historical biogeography of Lauraceae: evidence from the chloroplast and nuclear genomes. *Ann. Miss. Bot. Gard.* **88**, 104–134.
- Chandler, M. E. J. 1954 Some Upper Cretaceous and Eocene fruits from Egypt. *Bull. Br. Mus. (Nat. Hist.) Geol.* **2**, 149–187.
- Chandler, M. E. J. 1964 *The Lower Tertiary floras of southern England. 4. A summary and survey of findings in the light of recent botanical observations*. London: British Museum of Natural History.
- Chesters, K. I. M. 1955 Some plant remains from the Upper Cretaceous and Tertiary of West Africa. *Ann. Mag. Nat. Hist.* (12) **8**, 498–504.
- Clark, C. J., Poulsen, J. R., Connor, E. F. & Parker, V. T. 2004 Fruiting trees as dispersal foci in a semi-deciduous tropical forest. *Oecologia* **139**, 66–75.
- Coetzee, J. A. 1978 Climate and biological changes in southwestern Africa during the Late Cainozoic. *Palaeoecol. Afr.* **10**, 13–29.
- Crisp, M., Cook, L. & Steane, D. 2004 Radiation of the Australian flora: what can comparisons of molecular phylogenies across multiple taxa tell us about the evolution of diversity in present-day communities? *Phil. Trans. R. Soc. Lond. B* **359**, 1551–1571. (doi:10.1098/rstb.2004.1528)
- Davis, C. C., Bell, C. D., Mathews, S. & Donoghue, M. J. 2002a Laurasian migration explains Gondwanan disjunctions: evidence from Malpighiaceae. *Proc. Natl Acad. Sci. USA* **99**, 6833–6837.
- Davis, C. C., Bell, C. D., Fritsch, P. W. & Mathews, S. 2002b Phylogeny of *Acridocarpus–Brachylophon* (Malpighiaceae): implications for tertiary tropical floras and Afroasian biogeography. *Evolution* **56**, 2395–2405.
- Dick, C. W., Abdul-Salim, K. & Bermingham, E. 2003 Neotropical differentiation of an ancient rainforest tree, *Symphonia globulifera* Linnaeus. *Am. Nat.* **162**, 691–703.
- Dilcher, D. L. & Crane, P. R. 1984 *Archaeanthus*: an early angiosperm from the Cenomanian of the Western Interior North America. *Ann. Miss. Bot. Gard.* **71**, 351–383.
- Doyle, J. A. & Le Thomas, H. 1997 Phylogeny and geographic history of Annonaceae. *Géogr. Phys. Quatern.* **51**, 353–361.
- Doyle, J. A., Sauquet, H., Scharaschkin, T. & Le Thomas, A. 2004 Phylogeny, molecular and fossil dating, and biogeographic history of Annonaceae and Myristicaceae (Magnoliales). *Int. J. Pl. Sci.* **165**(4), (Suppl.) S55–S67.
- Fay, M. F., Bayer, C., Alverson, W. S., de Bruijn, A. Y. & Chase, M. 1998 Plastid *rbcL* sequence data indicate a close affinity between *Diegodendron* and *Bixa*. *Taxon* **47**, 43–50.
- Fay, M. F., Lledó, M. D., Richardson, J. E., Rye, B. L. & Hopper, S. D. 2001 Molecular data confirm the affinities of *Granitites* with *Alphitonia* (Rhamnaceae). *Kew Bull.* **56**, 669–675.

- Felsenstein, J. 1985 Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* **39**, 783–791.
- Fitch, W. M. 1971 Toward defining the course of evolution: minimum change for a specified tree topology. *Syst. Zool.* **20**, 406–416.
- Fritsch, P. W. 2001 Phylogeny and biogeography of the flowering plant genus *Styrax* (Styracaceae) based on chloroplast DNA restriction sites and DNA sequences of the internal transcribed spacer region. *Mol. Phylogenet. Evol.* **19**, 387–408.
- Gentry, A. H. 1982 Neotropical floristic diversity: phytogeographical connections between Central and South America, Pleistocene climatic fluctuations, or an accident of the Andean orogeny? *Ann. Miss. Bot. Gard.* **69**, 557–593.
- Graham, A. 2003 Geohistory models and Cenozoic palaeoenvironments of the Caribbean region. *Syst. Bot.* **28**, 378–386.
- Haffer, J. 1982 General aspects of the refuge theory. In *Biological diversification in the tropics* (ed. G. T. Prance), pp. 6–24. New York: Columbia University Press.
- Hampe, A. & Bairlein, F. 2000 Modified dispersal-related traits in disjunct populations of bird-dispersed *Frangula alnus* (Rhamnaceae): a result of its Quaternary distribution shifts? *Ecography* **23**, 603–613.
- Higgins, D. G., Bleasby, A. J. & Fuchs, R. 1992 CLUSTAL: a new multiple sequence alignment program. *Computer Applic. Biosci.* **8**, 189–191.
- Hileman, L. C., Vasey, M. C. & Parker, V. T. 2001 Phylogeny and biogeography of the Arbutioideae (Ericaceae): implications for the Madrean–Tethyan hypothesis. *Syst. Bot.* **26**, 131–143.
- Hill, R. S., Truswell, E. M., McCloughlin, S. & Dettmann, M. E. 1999 Evolution of the Australian flora: fossil evidence. In *Flora of Australia*, 2nd edn, vol. 1 (ed. A. E. Orchard), pp. 251–320. Canberra: Australian Biological Resources Study/CSIRO.
- Hopper, S. D., Harvey, M. S., Chappill, J. A., Main, A. R. & Amin, B. Y. 1996 The Western Australian biota as Gondwanan heritage: a review. In *Gondwanan heritage: past, present and future of the Western Australian biota* (ed. S. D. Hopper, J. A. Chappill, M. S. Harvey & A. S. George), pp. 1–46. Chipping Norton, NSW: Surrey Beatty & Sons.
- Hubbell, S. P. 2001 *The unified neutral theory of biodiversity and biogeography*. Princeton University Press.
- Hughes, C. E., Bailey, C. D., Krosnick, S. & Luckow, M. A. 2003 Relationships among genera of the informal *Dichrostachys* and *Leucaena* groups (Mimosoideae) inferred from nuclear ribosomal ITS sequences. In *Advances in legume systematics. Part 10. Higher level systematics* (ed. B. B. Klitgaard & A. Bruneau), pp. 221–238. Kew, UK: Royal Botanic Gardens.
- Iturralde-Vinent, M. A. & MacPhee, R. D. E. 1999 Palaeogeography of the Caribbean region: implications for Cenozoic biogeography. *Bull. Am. Mus. Nat. Hist.* **238**, 1–95.
- Kubitzki, K. & Krutsch, W. 1996 Origins of east and south Asian plant diversity. In *Floristic characteristics and diversity of east Asian plants* (ed. Z. Auluo & W. Sugong), pp. 65–70. Berlin: Springer.
- Lavin, M., Schrire, B., Lewis, G., Pennington, R. T., Delgado-Salinas, A., Thulin, M., Hughes, C., Beyra Matos, A. & Wojciechowski, M. F. 2004 Metacommunity process rather than continental tectonic history better explains geographically structured phylogenies in legumes. *Phil. Trans. R. Soc. Lond. B* **359**, 1509–1522. (doi:10.1098/rstb.2004.1536)
- Le Thomas, A. & Doyle, J. A. 1996 Geographic relationships of Malagasy Annonaceae. In *Biogéographie de Madagascar* (ed. W. R. Lourenco), pp. 85–94. Paris: Institut Français de Recherche Scientifique pour le Développement en Coopération.
- Linder, H. P. & Hardy, C. R. 2004 Evolution of the species-rich Cape flora. *Phil. Trans. R. Soc. Lond. B* **359**, 1623–1632. (doi:10.1098/rstb.2004.1534)
- Little, S. A., Stockey, R. A. & Beard, G. 2002 Angiosperm fruits and seeds from the Eocene of Vancouver Island. Botany 2001 Abstracts. Botanical Society of America.
- Liu, M. J. & Cheng, C. Y. 1995 A taxonomic study of the genus *Ziziphus*. *Acta Hort.* **390**, 161–165.
- McKenna, M. C. C. 1973 Sweepstakes, filters, corridors, Noah's Arks, and beached Viking funeral ships in paleogeography. In *Implications of continental drift to the Earth Sciences* (ed. D. H. Tarling & S. K. Runcorn), pp. 291–304. London: Academic.
- Magallón, S. & Sanderson, M. J. 2001 Absolute diversification rates in angiosperm clades. *Evolution* **55**, 1762–1780.
- Mai, D. H. 1995 *Tertiäre vegetationsgeschichte Europas*. Jena, Germany: Fischer.
- Manchester, S. R. 1999 Biogeographical relationships of North American Tertiary floras. *Ann. Miss. Bot. Gard.* **86**, 472–522.
- Menéndez, C. A. 1972 Palaeophytologia kurtziana. III.9. La flora del Cretácico Superior de Cerro Guido, Chile. *Ameghiniana* **9**, 289–296.
- Mols, J. B., Gravendeel, B., Chatrou, L. W., Pirie, M. D., Bygrave, P. C., Chase, M. W. & Keßler, P. J. A. 2004 Identifying clades in Asian Annonaceae: monophyletic genera in the polyphyletic Miliuseae. *Am. J. Bot.* **91**, 590–600.
- Mori, S. A. & Brown, J. L. 1994 Report on wind dispersal in a lowland moist forest in central French Guiana. *Brittonia* **46**, 105–125.
- Morley, R. J. 2000 *Origin and evolution of tropical rainforests*. Chichester, UK: Wiley.
- Morley, R. J. 2003 Interplate dispersal routes for megathermal angiosperms. *Perspectives Pl. Ecol. Evol. Syst.* **6**, 5–20.
- Near, T. J. & Sanderson, M. J. 2004 Assessing the quality of molecular divergence time estimates by fossil calibrations and fossil-based model selection. *Phil. Trans. R. Soc. Lond. B* **359**, 1477–1483. (doi:10.1098/rstb.2004.1523)
- Parrish, J. T. 1993 The paleogeography of the opening South Atlantic. In *The Africa–South America connection* (ed. W. George & R. Lavocat), pp. 8–41. Oxford: Clarendon.
- Pennington, R. T. 2003 Monograph of *Andira* (Leguminosae: Papilionoideae). *Syst. Bot. Monogr.* **64**, 1–143.
- Pennington, R. T. & Dick, C. W. 2004 The role of immigrants in the assembly of the South American rainforest tree flora. *Phil. Trans. R. Soc. Lond. B* **359**, 1611–1622. (doi:10.1098/rstb.2004.1532)
- Pindell, J. L. & Barrett, S. F. 1990 Geological evolution of the Caribbean region: a plate tectonic perspective. In *The geology of North America, vol. H. The Caribbean region* (ed. G. Dengo & J. E. Case), pp. 405–432. Geological Society of America.
- Pindell, J. L., Cande, S. C., Pitman, W. C., Rowley, D. B., Dewey, J. F., LaBrecque, J. & Haxby, W. 1988 A plate-kinematic framework for models of Caribbean evolution. *Tectonophysics* **155**, 121–138.
- Pirie, M. D., Chatrou, L. W., Erkens, R. H. J., Maas, J. W., van der Niet, T., Mols, J. B. & Richardson, J. E. 2004 Phylogeny reconstruction and molecular dating in four Neotropical genera of Annonaceae: the effect of taxon sampling in age estimations. In *Plant species-level systematics: new perspectives on pattern and process* (ed. F. T. Bakker, L. W. Chatrou, B. Gravendeel & P. B. Pelser), *Regnum vegetabile* vol. 143. Koenigstein, Germany: Koeltz Scientific. (In the press.)
- Posada, D. & Crandall, K. A. 1998 MODELTEST: testing the model of DNA substitution. *Bioinformatics* **14**, 817–818.
- Raven, P. H. 1979 Plate tectonics and Southern Hemisphere biogeography. In *Tropical botany* (ed. K. Larsen & L. B. Holm-Nielsen), pp. 1–24. London: Academic.

- Raven, P. H. & Axelrod, D. I. 1974 Angiosperm biogeography and past continental movements. *Ann. Miss. Bot. Gard.* **61**, 539–673.
- Renner, S. S. 2004 Multiple Miocene Melastomataceae dispersal between Madagascar, Africa, and India. *Phil. Trans. R. Soc. Lond. B* **359**, 1485–1494. (doi:10.1098/rstb.2004.1530)
- Renner, S. S., Clausen, G. & Meyer, K. 2001 Historical biogeography of Melastomataceae: the roles of tertiary migration and long distance dispersal. *Am. J. Bot.* **88**, 1290–1300.
- Richardson, J. E., Fay, M. F., Cronk, Q. C. B., Bowman, D. & Chase, M. W. 2000 A phylogenetic analysis of Rhamnaceae using *rbcL* and *trnL-F* plastid DNA sequences. *Am. J. Bot.* **87**, 1309–1324.
- Richardson, J. E., Pennington, R. T., Pennington, T. D. & Hollingsworth, P. M. 2001a Rapid diversification of a species-rich genus of neotropical rainforest trees. *Science* **293**, 2242–2245.
- Richardson, J. E., Weitz, F. M., Fay, M. F., Cronk, Q. C. B., Linder, H. P., Reeves, G. & Chase, M. W. 2001b Rapid and recent origin of species richness in the Cape flora of South Africa. *Nature* **412**, 181–183.
- Sanderson, M. J. 1997 A non-parametric approach to estimating divergence times in the absence of rate constancy. *Mol. Biol. Evol.* **14**, 1218–1231.
- Sanderson, M. J. 2002 Estimating absolute rates of molecular evolution and divergence times: a penalized likelihood approach. *Mol. Biol. Evol.* **19**, 101–109.
- Sanderson, M. J. 2003 R8s, v. 1.6, user's manual (April 2003). Distributed by the author at <http://ginger.ucdavis.edu/r8s/>. Davis, CA: University of California.
- Sauquet, H., Doyle, J. A., Scharaschkin, T., Borsch, T., Hilu, K. W., Chatrou, L. W. & Le Thomas, A. 2003 Phylogenetic analysis of Magnoliales and Myristicaceae based on multiple datasets: implications for character evolution. *Bot. J. Linn. Soc.* **142**, 125–186.
- Schrire, B. D., Lavin, M. & Lewis, G. P. 2004 Global distribution patterns of the Leguminosae: insights from recent phylogenies. In *Plant diversity and complexity patterns: local, regional and global dimensions* (ed. I. Friis & H. Balslev). Biologiske Skrifter. (In the press.)
- Siesser, W. G. 1980 Late Miocene origin of the Benguela upwelling system off northern Namibia. *Science* **208**, 283–285.
- Sole de Porta, N. 1971 Algunos generos nuevos de polen procedentes de la formacion Guaduas (Maastrichtense-Paleocene) de Colombia. *Stud. Geol. Salamanca* **2**, 133–143.
- Stebbins, G. L. 1974 *Evolution above the species level*. Cambridge: Harvard University Press.
- Swofford, D. L. 1998 *PAUP\*, v. 4.0b2: phylogenetic analysis using parsimony. (\* and other methods)*. Sunderland, MA: Sinauer.
- Tiffney, B. 1985a Perspectives on the origin of the floristic similarity between eastern Asia and eastern North America. *J. Arnold Arboretum* **66**, 73–94.
- Tiffney, B. 1985b The Eocene North Atlantic land bridge: its importance in Tertiary and modern phytogeography of the northern hemisphere. *J. Arnold Arboretum* **66**, 243–273.
- Truswell, E. M. 1990 Australian rainforests: the 100 million year record. In *Australian tropical rainforests: science-values-meaning* (ed. L. J. Webb & J. Kikkawa), pp. 7–22. Melbourne, Australia: CSIRO.
- Van der Hammen, T. & García de Mutis, C. 1966 The Paleocene pollen flora of Colombia. *Leidse Geol. Med.* **35**, 105–114.
- Van Tol, I. A. V. & Meijdam, N. A. J. 1991 Field research into pollination and seed dispersal of Annonaceae. Internal report, Institute of Systematic Botany, Utrecht University, The Netherlands.
- Walker, J. M. 1971 Pollen morphology, phytogeography and phylogeny of the Annonaceae. *Cont. Gray Herb. Harvard Univ.* **202**, 1–133.
- Whitmore, T. C. & Prance, G. T. 1987 *Biogeography and Quaternary history in tropical America*. Oxford Science Publications.
- Wikström, N., Savolainen, V. & Chase, M. W. 2001 Evolution of the angiosperms: calibrating the family tree. *Proc. R. Soc. Lond. B* **268**, 2211–2220. (doi:10.1098/rspb.2001.1782)
- Zachos, J., Pagani, M., Sloan, L., Thomas, E. & Billups, K. 2001 Trends, rhythms and aberrations in global climate 65 Ma to present. *Science* **292**, 686–693.

## GLOSSARY

- CI: consistency index  
 EECO: Early Eocene climatic optimum  
 GAARlandia: Greater Antilles and Aves Ridge  
 MRCA: most recent common ancestor  
 NPRS: non-parametric rate smoothing  
 RI: retention index  
 TBR: tree bisection–reconnection