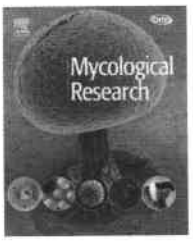
MYCOLOGICAL RESEARCH 112 (2008) 448–462

ELSEVIER

British Mycological Society promoting fungal science

journal homepage: www.elsevier.com/locate/mycres



Biogeography of *Hysterangiales* (*Phallomycetidae*, *Basidiomycota*)

Kentaro HOSAKA[a,*,†], *Michael A. CASTELLANO*[b], *Joseph W. SPATAFORA*[a]

[a]*Department of Botany and Plant Pathology, Cordley Hall 2082, Oregon State University, Corvallis, OR 97331-2902, USA*
[b]*USDA, Forest Service, PNW Research Station, Forestry Sciences Laboratory, 3200 Jefferson Way, Corvallis, OR 97331, USA*

ARTICLE INFO

Article history:
Received 29 November 2006
Received in revised form
22 May 2007
Accepted 10 June 2007
Corresponding Editor:
H. Thorsten Lumbsch

Keywords:
Ectomycorrhizae
False-truffles
Gomphoid-phalloid
Mycophagy
Phylogeography

ABSTRACT

To understand the biogeography of truffle-like fungi, DNA sequences were analysed from representative taxa of *Hysterangiales*. Multigene phylogenies and the results of ancestral area reconstructions are consistent with the hypothesis of an Australian, or eastern Gondwanan, origin of *Hysterangiales* with subsequent range expansions to the Northern Hemisphere. However, neither Northern Hemisphere nor Southern Hemisphere taxa formed a monophyletic group, which is in conflict with a strictly vicariant scenario. Therefore, the occurrence and importance of long-distance dispersal could not be rejected. Although a pre-Gondwanan origin of *Hysterangiales* remains as a possibility, this hypothesis requires that *Hysterangiales* exist prior to the origin of the currently recognized ectomycorrhizal plants, as well as the arrival of mycophagous animals in Australia. This also requires that a basal paraphyletic assemblage represents parallel evolution of the ectomycorrhizal symbiosis, or that *Hysterangiales* was mycorrhizal with members of the extinct flora of Gondwana. Regardless, models for both ancient and more recent origins of *Hysterangiales* are consistent with truffle-like fungi being capable of transoceanic dispersal.

Introduction

Although fungi are ubiquitous and play an important role in terrestrial ecosystems, biogeography of fungi (or mycogeography) has not been extensively studied within a phylogenetic framework. The cryptic nature of many fungi makes them difficult to sample, thus hindering global-scale biogeographical studies. Many mycogeographical studies have focused on Northern Hemisphere taxa. For example, *Grifola* (Shen *et al.* 2002) and *Amanita* (Oda *et al.* 2004) showed a Palearctic *versus* Nearctic pattern, which is a pattern that is common in biogeographical studies of plants, where a disjunct eastern North America *versus* eastern Asia distribution pattern is not supported by phylogenetic analyses (Xiang *et al.* 1998). Conversely, *Suillus* (Wu *et al.* 2000) and the *Tricholoma matsutake* group (Chapela & Garbelotto 2004) showed that eastern North American and eastern Asian taxa were more closely related to each other than to western North American taxa.

Several studies have dealt with fungi distributed both in the Northern and Southern Hemisphere. Interestingly, mycogeographical patterns frequently show a New World *versus* Old World pattern, not a Laurasia *versus* Gondwana pattern, which would be expected if the present distribution was caused by the break up of Pangaea. Examples of a New World *versus* Old World pattern include the *Pleurotus cystidiosus* group (Zervakis *et al.* 2004), *Schizophyllum* (James *et al.* 2001), and *Lentinula* (Hibbett 2001). Several exceptions to this pattern were observed in each study and they were usually explained

* Corresponding author.
E-mail address: khosaka@fieldmuseum.org
† Current address: Department of Botany, The Field Museum, 1400 S. Lake Shore Drive, Chicago, Illinois 60605-2496, USA.
0953-7562/$ – see front matter © 2007 The British Mycological Society. Published by Elsevier Ltd. All rights reserved.
doi:10.1016/j.mycres.2007.06.004

by rare, but recent long-distance dispersal. For example, long-distance dispersal between Australia and New Zealand was postulated for *Lentinula* (Hibbett 2001). Conversely, *Pleurotus* (Vilgalys & Sun 1994) and *Panellus* (Jin *et al.* 2001) were monophyletic in the Northern Hemisphere *versus* mono- or paraphyletic in the Southern Hemisphere. Although this pattern could be consistent with an ancient vicariant event in Pangaea, the authors proposed a more recent origin of the groups and dispersal scenario between the Northern and Southern Hemisphere continents. Similar patterns were observed for *Armillaria* (Coetzee *et al.* 2003) and *Pisolithus* (Martin *et al.* 2002), but ancient origins of these genera were suggested. Notably, Martin *et al.* (2002) suggested that the ancestor of the ectomycorrhizal genus *Pisolithus* was a generalist mycorrhizal symbiont, and originated before the break up of Pangaea in the Triassic period.

In this study, we assessed a global biogeographical pattern of *Hysterangiales*. *Hysterangiales* is an order of the subclass *Phallomycetidae* (*Basidiomycota*), which forms hypogeous fruiting bodies commonly referred to as false-truffles (Castellano *et al.* 1989; Hosaka *et al.* 2006). Because of its hypogeous habit, the spores of *Hysterangiales* are not thought to be disseminated by wind, as is the case of many epigeous mushroom-forming fungi that are capable of long distance spore dispersal (Thiers 1984). Instead, they produce a unique aroma that attracts small animals, which rely on hypogeous fungi as a large part of their diet (Castellano *et al.* 1989; Thiers 1984). Hypogeous fruiting bodies are eaten by small animals and the fungal spores are disseminated with the animal faeces (Castellano *et al.* 1989; Malajczuk *et al.* 1987b). Because spore dissemination of hypogeous fungi, including that of *Hysterangiales*, depends on such mycophagy, long-distance (e.g. transoceanic) dispersal of spores of hypogeous fungi is arguably less likely.

Despite its hypogeous habit and high dependency on animal mycophagy, *Hysterangiales* is distributed worldwide, both in the Northern and Southern Hemisphere (Castellano 1999). This is consistent with *Hysterangiales* being an old taxon, and the current distribution being the result of ancient vicariant events associated with the supercontinent Pangaea. Alternatively, it could be explained by *Hysterangiales* being a much more efficient disperser than predicted by morphology with a more recent origin. So far, Australia, North America, and Europe are documented centres of diversity for *Hysterangiales*, with more than 15 endemic species from each area (Castellano 1999), and recent studies also revealed relatively high diversity in New Zealand (Castellano & Beever 1994) and South America (Castellano & Muchovej 1996). The other known distributions of *Hysterangiales* include Africa, India, temperate and tropical Asia, New Caledonia, and Papua New Guinea (Castellano *et al.* 2000). Importantly, the distribution of each species appears to be restricted to a single continent or island (Castellano 1999). Therefore, areas of endemism can easily be defined as each continent (e.g. North America) or island (e.g. New Caledonia).

Most species of *Hysterangiales* are considered obligate ectomycorrhizal fungi. A wide range of trees, including both gymnosperms and angiosperms, are known as ectomycorrhizal hosts for *Hysterangiales* (Castellano 1999). Although host range varies, any one species of *Hysterangiales* only associates with hosts from one plant family and often only one genus or species (Castellano 1999). Major ectomycorrhizal host families for *Hysterangiales* include *Pinaceae*, *Myrtaceae*, *Fagaceae*, and *Nothofagaceae*. There are some examples of *Hysterangiales* associated with *Dipterocarpaceae*, *Ericaceae*, *Casuarinaceae*, and caesalpinioid legumes (*Caesalpinioideae*) (Castellano 1999; Castellano & Beever 1994; Castellano *et al.* 2000). This relative host specificity of the ectomycorrhizal systems enables us to assess the historical host–fungus associations, e.g. host-tracking *versus* host-shifting, and gives us clue to the historical biogeography of *Hysterangiales*.

Recent studies support the gomphoid-phalloid clade (*Phallomycetidae*), to which *Hysterangiales* belongs, as one of the basal clades of *Agaricomycetes* (mushroom-forming fungi) (Binder & Hibbett 2002; James *et al.* 2006; Lutzoni *et al.* 2004). Precambrian origins of major fungal lineages are postulated by some molecular clock studies (Heckman *et al.* 2001; Hedges *et al.* 2004). Halling (2001) and Martin *et al.* (2002) hypothesized, although not based on molecular clock or fossil records, that ectomycorrhizal fungi have diversified in the Jurassic or even older before the break up of Pangaea. These data are again consistent with the potentially ancient and vicariant origin of *Hysterangiales*.

All of the above features of global distribution with well-defined areas of endemism, ectomycorrhizal host range and specificity, hypogeous habit, and phylogenetic position make *Hysterangiales* an attractive system for testing numerous evolutionary hypotheses including dispersal *versus* vicariance, host-tracking *versus* host-shifting, and the ancient origin of extant fungal lineages. To address the overall goals of this study we sampled all available species of *Hysterangiales* with an emphasis on geographical distribution and ectomycorrhizal host association. As far as we know, this is the first phylogeographical study of globally distributed truffle-like fungi.

Materials and methods

Taxon sampling, PCR, and DNA sequencing

Taxa sampled, along with GenBank accession numbers, are listed in Supplementary Material Table 1. One hundred and fourteen taxa (two outgroup and 112 ingroup taxa) were sampled for this study. The selection of ingroup and outgroup taxa was based on the phylogeny of previous studies (Hosaka *et al.* 2006) to cover the diversity of *Hysterangiales*. Among the ingroup taxa, 107 taxa were ectomycorrhizal, and five taxa were considered as saprobic, based on the habitat information, or direct observation (morphological or molecular) of ectomycorrhizas (Agerer & Iosifidou 2004; Castellano 1990; Castellano & Beever 1994; Dell *et al.* 1990; Lu *et al.* 1999; Malajczuk *et al.* 1987a; Miller & Miller 1988; Molina & Trappe 1982; Müller & Agerer 1996; Raidl & Agerer 1998; Stewart & Trappe 1985).

DNA was extracted from glebal tissue of fresh or dried fruiting bodies using the protocol of Humpert *et al.* (2001). DNA sequence data were obtained from five independent loci: LR0R–LR3 region for nuLSU rDNA; MS1–MS2 region for mtSSU rDNA; ATPase subunit 6 (*atp6*); bRPB2-6F–bRPB2-7R region for the second largest subunit of RNA polymerase (*RPB2*);

EF1-983F–EF1-1567R region for translation elongation factor subunit 1α (EF-1α). The primers and PCR protocols were described previously (Kretzer & Bruns 1999; Liu et al. 1999; Matheny 2005; Rehner & Buckley 2005; Vilgalys & Hester 1990; White et al. 1990).

Phylogenetic analyses

DNA sequences were initially aligned using Clustal X (Thompson et al. 1997), followed by manual alignment in the data editor of BioEdit ver. 7.0.1 (Hall 1999). Ambiguously aligned regions and introns were excluded from the analyses.

To test for incongruence among the five individual datasets, 70 % BS trees from parsimony analyses of individual loci were compared. First, 70 % BS trees were calculated (100 BS replicates with five random addition sequences, TBR and Multrees options off) including only the taxa with sequences from all five loci. These trees were used as constraints in a different dataset (for example, parsimony analysis of the *atp6* dataset with the nuLSU rDNA tree as a constraint), using the 'Load Constraints' option in PAUP version 4.0b10 (Swofford 2002). Parsimony analyses (a two-step search approach described below) were conducted under these constraints, keeping only the trees that are compatible with these constraints. A total of ten constraint parsimony analyses were conducted for all pair-wise gene comparisons.

Comparisons of constraint and unconstraint trees were made using the 'Tree Scores' option in PAUP version 4.0b10 (Swofford 2002). Parsimony based comparisons were performed by the Templeton test (Templeton 1983), using non-parametric pairwise tests option. Likelihood-based comparisons were performed by the Shimodaira–Hasegawa test (Shimodaira & Hasegawa 1999), using RELL optimization with 1K BS replicates. Significance of results was determined by a P-value less than 0.05. After testing for incongruence, the individual gene datasets were combined and phylogenetic (both parsimony and Bayesian) analyses were conducted with a combined dataset of five loci described above.

Parsimony analyses were conducted under the equally-weighted parsimony criterion using PAUP version 4.0b10 (Swofford 2002). A two-step search approach was performed. In the first step, the heuristic search option [with tree bisection–reconnection (TBR), no Multrees] and 1K replicates of random addition sequence were performed, keeping only up to two shortest trees per replicate. In the second step, all of the shortest trees from the first step were used as starting trees for the heuristic search option (with TBR and Multrees on) with MAXTREES set to 10K. Support for the individual nodes was tested with BS analysis under the equally-weighted parsimony criterion. BS analysis was based on 500 BS replicates using the heuristic search option (TBR and Multrees off), with five random addition sequences.

Bayesian analyses were conducted using MrBayes version 3.0b4 (Huelsenbeck & Ronquist 2001). Eleven data partitions, including nuLSU rDNA and mtSSU rDNA, and one for each codon position for each of the three protein-coding loci, were delimited for Bayesian analyses. The $GTR + \Gamma + I$ model was employed separately for each of the 11 data partitions. Bayesian analyses were run with 5M MCMCMC generations with four chains, sampling trees every 100th generation. The log-likelihood scores of sample points against generation time were plotted using TRACER version 1.3 (http://evolve.zoo.ox.ac.uk/software.html) to determine if the run reached stationarity. We also observed the average standard deviation of split frequencies and verified that the values dropped below 0.01. The support of nodes was tested by PPs, obtained from a 50 % majority rule consensus after deleting the trees in the burn-in period.

Biogeographical analyses

Areas of endemism

The areas used in this study were delimited based on the geological history of Pangaean break up (Fig 1). Although some areas, especially the Holarctic (Northern Hemisphere) could be further divided into several smaller areas, these subdivisions were not used for the analyses. This is mainly because the area relationships for the Holarctic are not necessarily hierarchical. For example, the land connection between North America and Eurasia was connected and disconnected several times (Sanmartín et al. 2001). The following ten areas of endemism were used throughout this study for biogeographical analyses: (1) Australia (AUS): this is one of the better represented areas for *Hysterangiales*. Most AUS *Hysterangiales* are represented in Victoria and its vicinity, but some are from Western Australia, Queensland, or Tasmania. In this study, AUS including Tasmania was treated as one unit area. Forty-four species were represented for this study. (2) New Guinea (PNG): two species were represented, and both are from Papua New Guinea (Goroka and Lae). Although several areas of New Guinea Island could be treated separately due to its complex geological history, the whole island was treated as one area for this study. (3) New Zealand (NZ): both North and South Islands were treated as the same area. One species of *Hysterangium* (*H. youngii*) was collected in NZ, and it was demonstrated to be very closely related to the other Holarctic taxa. Although we do not have a detailed vegetation record of the collecting site, we strongly suspect that this species was collected under planted *Pinaceae* or *Fagaceae* originating in the Northern Hemisphere. Therefore for this study, *H. youngii* was treated as a Holarctic taxon. Fifteen species were represented for this study. (4) New Caledonia (NC): two species were represented for this study, and both are from the main island of NC (Grand Terre). (5) Southern South America (SSAM): defined here as the southern temperate region of South America, following the treatment of Sanmartín and Ronquist (2004). All known *Hysterangiales* distributed in SSAM are from Argentina or Chile. Four species were represented for this study. (6) Northern South America (NSAM): defined here as north-central South America east of the Andes, following the treatment of Sanmartín & Ronquist (2004). All known *Hysterangiales* distributed in NSAM are from Guyana. Four species were represented for this study. (7) Africa (AF): one species was represented for this study, and it is from Zimbabwe. (8) India (IND): one species was represented for this study, and it is from Karnataka Province (southwest IND). (9) Southeast Asia (SEA): defined here as Malaysian Peninsula. PNG was treated as an independent area, and not included in SEA. Three species were represented for this study. Two of them are from southern part of Thailand, and the other is from Singapore. (10) Holarctic

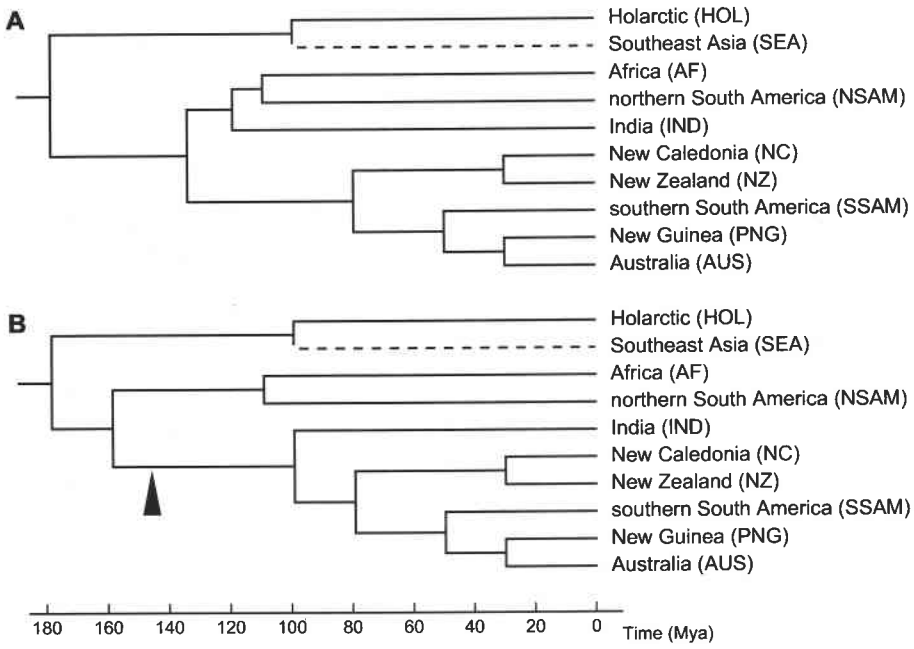


Fig 1 – Area cladograms based on geological data. (A) Based on Sanmartín & Ronquist (2004). (B) Based on McLoughlin (2001). The two cladograms differ only ien the relative position of India. The position of Southeast Asia was arbitrarily determined because of its complicated history (see Materials and methods). Time is in million years ago (Mya) from present. The black triangle in (B) indicates an alternative position of Southern South America based on Hallam (1994), showing the initial separation of Southern South America from the rest of Gondwana (see Materials and methods, *Geological scenario*).

(HOL): defined here as a combination of Palearctic and Nearctic. One species is from Central America (Costa Rica), and it was treated as a HOL taxon, following the treatment of Sanmartín & Ronquist (2004). Thirty-six species were represented for this study. HOL can be further divided into four regions; western North America (WNAM), eastern North America (ENAM), Asia, and Europe (EUR). However, these subdivisions were not used for biogeographical analyses.

Geological scenario

Two slightly different geological area cladograms are presented as the sequence of Pangaean breakup (Fig 1). One cladogram (Fig 1A) was based on Sanmartín & Ronquist (2004), where they used the opening of the South Atlantic Ocean (135 Mya) as a basis of the initial break up of Gondwana. However, several studies (McLoughlin 2001; Hallam 1994) suggest that the opening of the Weddell Sea (*ca* 160 Mya) separated eastern (including AUS, NZ, and IND) and western Gondwana (including AF and NSAM). This event was reflected in the alternative geological scenario (Fig 1B). Timing of the separation of IND from the rest of Gondwana and collision to the northern continents is also controversial. Sanmartín and Ronquist (2004) used 120 Mya as the timing of separation of IND (and Madagascar) from AF. However, some studies suggest that IND was connected to the rest of eastern Gondwana *via* Antarctica and the Kerguelen Plateau until 80 Mya (Sampson *et al.* 1998). Although it is currently submerged, the Kerguelen Plateau emerged above sea level during the Cretaceous and might have been an important corridor for terrestrial organisms (McLoughlin 2001).

The position of SEA was somewhat arbitrarily reflected in Fig 1 because of its hybrid nature. Geological evidence suggest that the present SEA was once located in the northern periphery of eastern Gondwana (Metcalfe 1998). The separation of SEA from Gondwana happened multiple times during the Paleozoic and Early Mesozoic. The geological history of Malaysian Peninsula is of particular interest herein, because all three SEA species of *Hysterangiales* sampled in this study are from this region. The present Malaysian Peninsula was largely composed of continental terrane known as Sibumasu, which was separated from Gondwana by the Late Permian and collided into Asia by the Late Triassic (Metcalfe 1998). Because these events occurred before the major break up of Pangaea, SEA was treated as a sister area of HOL.

SSAM retained a direct land connection to Antarctica until *ca* 30 Mya (McLoughlin 2001). However, Hallam (1994) suggested that there was a seaway separating the southern tip of South America from Antarctica in the Jurassic. Because a land connection between South America and Antarctica is well-documented throughout the Cretaceous (McLoughlin 2001), there was not a dispersal barrier between South America and Australia for terrestrial organisms with a Cretaceous origin. However, for the organisms with pre-Jurassic origin, this seaway could have served as an effective barrier.

Analytical tools

We used two computer programs for testing and refining the biogeography of *Hysterangiales*: DIVA 1.1 [Ronquist F, 1996. Computer program and manual available by anonymous FTP

from Uppsala University (ftp.uu.se or ftp.systbot.uu.se)] for dispersal–vicariance analysis (Ronquist 1997), and TreeFitter 1.0 (http://www.ebc.uu.se/systzoo/research/treefitter/treefitter.html) for parsimony-based tree fitting. Both methods are parsimony-based approaches that explicitly optimize the taxon-area cladogram by minimizing the total event cost. Four types of events are considered: vicariance, duplication (speciation within an area), dispersal, and extinction. Vicariance is a null hypothesis for speciation, and is, therefore, assigned zero cost, whereas dispersal and extinction receive positive costs (Ronquist 1997; Sanmartín & Ronquist 2004). By explicitly assigning the costs for each event, these methods differ from other computer programs for biogeographical analyses, such as COMPONENT (Page 1990) and TreeMap (Page 1995), which maximize the vicariance events at the expense of introducing more dispersal and extinction than necessary. In addition, DIVA can be applied without prior knowledge of the area relationships because it does not assume that area relationships are hierarchical (Ronquist 1997).

One obvious weakness of these methods is that optimizations depend on the specific value of the cost of individual events (Sanmartín *et al.* 2007). Because vicariance receives zero cost, the methods inevitably favour vicariance reconstructions over dispersal (Cook & Crisp 2005). A simulation study by Ronquist (2003) showed that optimizations under the current event cost settings (low cost to vicariance and duplication, and a higher cost to dispersal and extinction) performed well in a variety of host–parasite association patterns. However, how this result applies to real data is largely unknown. Parsimony-based methods are based only on a single topology, and information on branch lengths, uncertainty of phylogenies, and divergence time, is usually ignored. Efforts have been made to incorporate such information under likelihood (Ree *et al.* 2005) or Bayesian framework (Huelsenbeck *et al.* 2000). However, it is still premature to use these approaches for a large, real dataset like this study because a huge computational time is required and, in some cases, the model currently employed is too simplistic (Huelsenbeck *et al.* 2003). We suggest that readers who wish to use DIVA and TreeFitter consider the aforementioned advantages and, especially disadvantages, before drawing any conclusions about biogeographical hypotheses of organisms of interest.

Dispersal–vicariance analyses

Ancestral areas of each node in the phylogeny of *Hysterangiales* were reconstructed using DIVA. The reconstructions were made using the default event cost settings (zero for vicariance and duplication, 1 for dispersal and extinction). We used these event costs because the current version of DIVA does not allow users to change the cost settings. An initial attempt of ancestral area reconstructions resulted in highly unresolved reconstructions on many basal nodes. DIVA optimizations become less reliable at basal nodes, which tend to have large distributions that include most or all of the areas occupied by the terminals (Ronquist 1996). One solution for this problem is to constrain the maximum number of unit areas allowed in ancestral distributions. By doing this, we are asking 'if this group had a restricted distribution in the past, what is the most likely ancestral distribution of the group (Ronquist 1996)?' Therefore, reconstructions were made constraining the number of ancestral areas using the 'maxareas' command. The number of maximum ancestral areas was constrained from two to ten (=unspecified), and each result was compared.

Parsimony-based tree fitting

Searches for the best area cladograms. TreeFitter was used to find the best (most parsimonious) area cladograms, inferred from the taxon-area cladogram (Fig 3). The area cladogram with the lowest cost best explains the taxon distributions in the phylogeny (Sanmartín & Ronquist 2004). Searches were conducted using the 'exhaustive' option with the default settings for event costs (zero for vicariance and duplication, 1 for extinction, and 2 for dispersal).

Tree fitting and randomization tests. To assess the statistical significance of the fit between the geological area cladogram (Fig 1B) and the phylogeny of *Hysterangiales* (Fig 3), randomization tests were conducted. Randomization tests were based on 10K permutations of the terminals in the geological area cladogram to offer the maximum sensitivity for detecting vicariance patterns (Sanmartín & Ronquist 2004). The percentage of area cladograms obtained from permutations with a lower cost than the original area cladogram was used as the significance value ($P < 0.05$). The number of each event (vicariance, duplication, dispersal, and extinction) was also inferred by fitting the taxon-area cladogram (Fig 3) to a geological area cladogram (Fig 1B), using the 'calcevents' command of TreeFitter.

Sensitivity analyses. For both of the above analyses, a range of event cost settings were implemented to investigate the sensitivity of results. Analyses were conducted by changing the event costs for vicariance, dispersal, and extinction from zero to four while keeping the cost for duplication to zero. Searches for the best area cladograms, tree fitting, and randomization tests were performed under each of the event cost settings.

Ancestral ectomycorrhizal host reconstructions

Ancestral ectomycorrhizal hosts were reconstructed for most nodes of Fig 2 using MacClade version 4.06 (Maddison & Maddison 2003). Because the *Phallogastraceae* clade contains only non-mycorrhizal (saprobic) taxa, and the rest of *Hysterangiales* are all ectomycorrhizal, the *Phallogastraceae* clade was not included for reconstructions. Each taxon was coded for its known ectomycorrhizal host plant. Although more specific host information (genus or species of host plants) was available for some taxa, only family-level information was used for coding. Eight host families (*Myrtaceae*, *Nothofagaceae*, *Fagaceae*, *Pinaceae*, *Caesalpinioideae*, *Casuarinaceae*, *Dipterocarpaceae*, and *Ericaceae*) were used for coding. If presumable ectomycorrhizal hosts could not be identified to a single host family, taxa were coded as polymorphic. All reconstructions were based on unweighted parsimony criterion. Because some nodes were reconstructed only ambiguously, both maximum and minimum number of all possible changes from one host

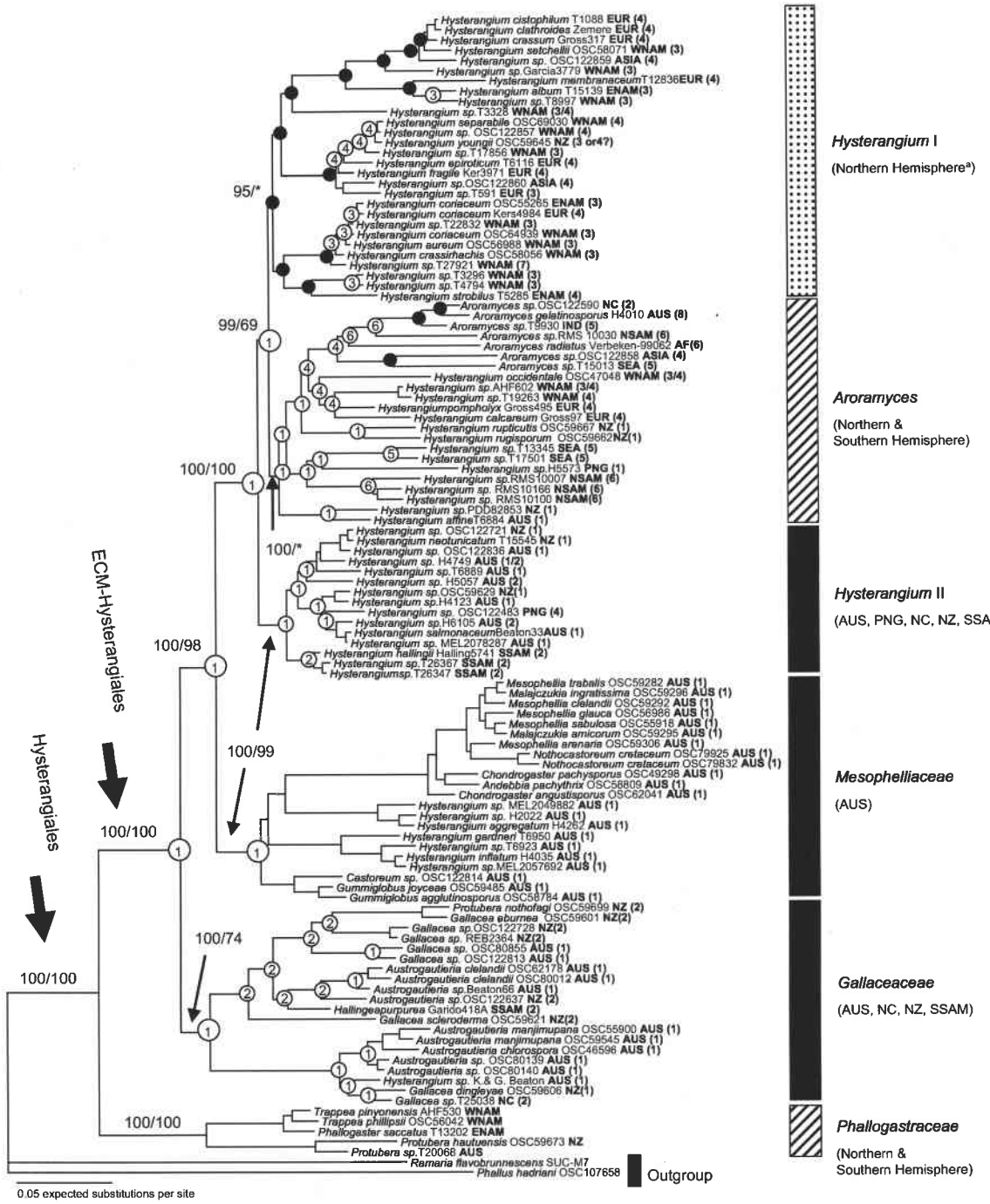


Fig 2 – A 50 % majority rule consensus of *Hysterangiales* phylogeny derived from Bayesian analysis. Taxon names followed by area of distribution (for abbreviation of areas, see Fig 1 and Materials and methods), and by presumable ectomycorrhizal host in parentheses (1 = *Myrtaceae*, 2 = *Nothofagaceae*, 3 = *Pinaceae*, 4 = *Fagaceae*, 5 = *Dipterocarpaceae*, 6 = *Caesalpinioideae*, 7 = *Ericaceae*, 8 = *Casuarinaceae*). Numbers in circles indicate the ancestral ectomycorrhizal host inferred from unweighted parsimony reconstructions. Black circles on nodes indicate ambiguous reconstructions. Numbers on branches are nodal supports shown as percentage (Bayesian PP/parsimony BS values; asterisk indicates no BS support. Only deep nodes are labelled). [a]One taxon distributed in New Zealand, but treated as a Holarctic taxon (see Materials and methods, *Areas of endemism*).

family to the others was recorded using the 'state changes and stasis' option.

Results and discussion

Phylogenetic analyses

Comparisons of 70 % BS trees from the individual gene analyses did not reveal any major conflicts among the datasets. The combined dataset after excluding the ambiguously aligned regions had an alignment length of 2842 bp, including 651 bp of *atp6*, 543 bp of nuLSU rDNA, 758 bp of *RPB2*, 483 bp of *EF-1α*, and 407 bp of mtSSU rDNA. The number of parsimony informative characters was 968 for the combined dataset, including 293 for *atp6* (77, 35, and 181 for 1st, 2nd, and 3rd codon position, respectively), 142 for nuLSU rDNA, 294 for *RPB2* (47, 18, and 229 for 1st, 2nd, and 3rd codon position, respectively), 155 for *EF-1α* (12, 8, and 135 for 1st, 2nd, and 3rd codon position, respectively), and 84 for mtSSU rDNA.

Parsimony analyses yielded 320 most parsimonious trees, of which 73 trees were found in the first step of the heuristic search. The most parsimonious trees had 6275 steps with a CI of 0.259, RI of 0.703, and RC of 0.183. Bayesian analyses reached the plateau of the log-likelihood at approximately 400K generations (based on the plotting using TRACER), and the average standard deviation of split frequencies went below 0.01 after approximately 800K generations. A 50 % majority rule consensus of the 40K Bayesian trees with the average log-likelihood of −36 882.42 (harmonic mean) is provided (Fig 2), after discarding the first 10K trees (1M generations) as the burn-in phase. The potential scale reduction factor was 1.000–1.002 for all parameters, indicating that the analyses were run for a sufficient number of generations.

The phylogenetic analyses with a combined five-gene dataset strongly supported the monophyly of *Hysterangiales* (100 % PP and BS value; Fig 2), consistent with the earlier study by Hosaka *et al.* (2006). In addition, all major groups found previously (Hosaka *et al.* 2006) were recovered with strong support. Six major clades within *Hysterangiales* were recognized, all of which, except for the *Hysterangium* I and *Aroramyces* clades, were well-supported by both the Bayesian and parsimony analyses (Fig 2). All saprobic taxa within *Hysterangiales* were confined to the *Phallogastraceae* clade (Castellano 1990; Miller & Miller 1988), and the rest of *Hysterangiales* were all ectomycorrhizal taxa (ECM-*Hysterangiales* clade in Fig 2). This suggests a single origin of the ectomycorrhizal habit for *Hysterangiales* or less parsimoniously, parallel gains of ectomycorrhizal habit. Although multiple losses of ectomycorrhizal habit have been hypothesized to have occurred during the evolution of *Agaricomycetes*, as well as multiple gains (Hibbett *et al.* 2000), no apparent loss of mycorrhizal habit was observed in *Hysterangiales*.

Higher-level phylogeny of *Hysterangiales* revealed that strong biogeographical patterns exist (Fig 2). While the *Phallogastraceae* clade was composed of both Northern and Southern Hemisphere taxa, the three basal clades within the ECM-*Hysterangiales* clade were composed strictly of Southern Hemisphere taxa (Fig 2). Northern Hemisphere taxa were restricted to the more terminal clades (*Hysterangium* I and *Aroramyces* clades; Fig 2). Neither Northern Hemisphere nor Southern Hemisphere taxa formed monophyletic groups, so the simple Northern *versus* Southern Hemisphere vicariant pattern could not be applied. Southern Hemisphere taxa comprised a basal paraphyletic assemblage, and Northern Hemisphere taxa were nested within Southern Hemisphere taxa. One major clade (*Hysterangium* I) was strictly composed of Northern Hemisphere taxa (Fig 2).

Several nodes showed congruent patterns with the geological history, including the sister relationships of AUS and PNG (*Hysterangium* II clade), and NC and NZ (*Gallaceaceae* clade). As far as we know, the area relationship observed in the phylogeny of *Hysterangiales* is one of a few biogeographical examples showing the sister relationship of NZ and NC. The *Phallogastraceae* clade showed a pattern consistent with Pangaean breakup (Fig 3). Although only five taxa are included in this clade, the pattern might correspond to the initial split of Pangaea into Gondwana and Laurasia (McLoughlin 2001; Hallam 1994; Fig 1). The area relationships observed in the *Aroramyces* clade (Fig 3) might also be a reflection of ancient geological event. This clade contains both Southern and Northern Hemisphere taxa, and it is the only clade containing AF and IND taxa. Furthermore, the area relationships within this clade seem to be congruent with the geological history of Pangaean break up (Fig 1). Taxa from AF, NSAM, IND, NC, and AUS form a monophyletic group, which is consistent with the fact that all those areas were once united as a single continent, Gondwana. The difference is that AF and NSAM did not form monophyly, but the general patterns closely resemble the sequence of Pangaean break up.

Several incongruent patterns with the geological history were also observed. The most frequent pattern was sister relationship of AUS and NZ, which was observed in six nodes (Fig 3). Generally, AUS and NZ are not considered the sister areas. Several geological evidences suggest that NZ and NC were separated from AUS by 80 Mya, while AUS was still connected to SSAM *via* Antarctica (McLoughlin 2001; Hallam 1994). If this is the case, we expect to see a sister relationship of AUS and SSAM, instead of AUS and NZ. Although SSAM was represented only in two clades in this study (*Gallaceaceae* and *Hysterangium* II clades; Fig 3), the patterns in both clades showed that AUS and NZ are more closely related to each other than either one of them is to SSAM. Many independent studies have shown that biogeographical patterns of AUS and NZ could only be explained by long-distance dispersal between these areas (Pole 1994; Knapp *et al.* 2005; Moyersoen *et al.* 2003). That significant areas of NZ have been submerged during Late Cretaceous to Mid-Tertiary (Pole 1994; McLoughlin 2001) also support the ideas that the presence of many, if not all, terrestrial organisms in NZ are due to long-distance dispersal, most likely from AUS.

Dispersal–vicariance analyses

DIVA suggested that the common ancestor of *Hysterangiales* had a wide distribution, but the reconstructions varied under different constraints on the maximum number of areas (Table 1). When 'maxareas' was not specified, the most basal node (node 1, Fig 3) was reconstructed as widespread in all ten unit areas (Table 1), whereas AUS, NZ and HOL were

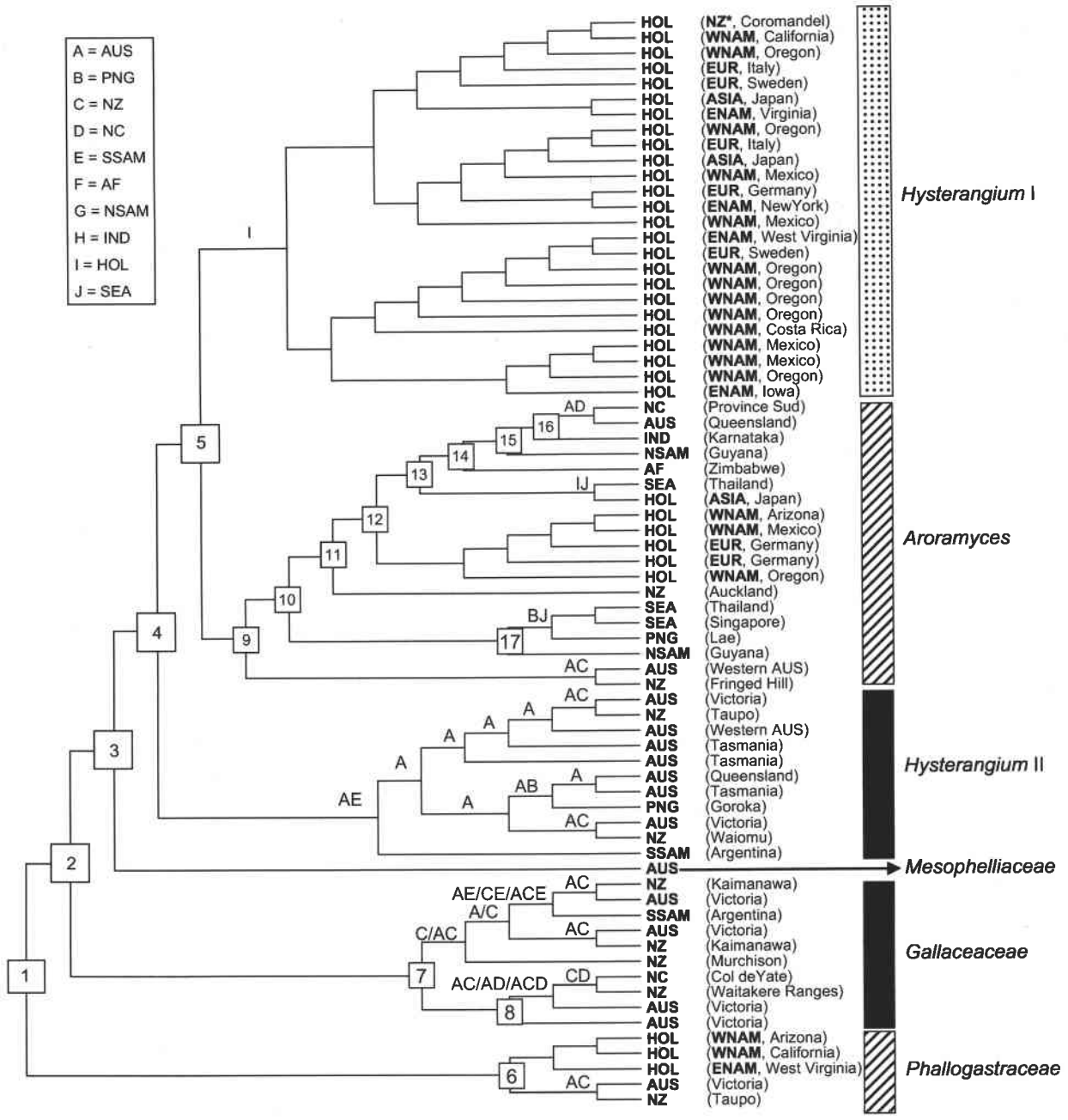


Fig 3 – Simplified taxon-area cladogram used for DIVA and TreeFitter analyses. Taxon names were replaced by areas of endemism (for abbreviation of areas, see Fig 1 and Materials and methods). Areas are followed by more specific locality information if available. Holarctic is subdivided into four unit areas, but these were not used for the analyses. Clade names follow Fig 2. Characters above branches indicate the results of the ancestral area reconstructions using DIVA. Two or more characters without a space indicate that the ancestors were widespread across those areas. Characters separated by a slash indicate the alternative equally parsimonious reconstructions. Ancestral areas reconstructed consistently throughout the 'maxareas' option of DIVA (ranging from two to ten) are shown. Numbers in squares (1–17) indicate the nodes that had different reconstructions throughout the 'maxareas' settings. See Table 1 for the reconstructions on nodes 1–17.

unambiguously reconstructed as the ancestral distribution under the 'maxareas' constraints from three to six. Regardless, a wide distribution of the common ancestor of *Hysterangiales*, both in the Northern and Southern continents, was suggested. Nodes 2–5 (Fig 3) were reconstructed more ambiguously (Table 1). When 'maxareas' was specified to nine or unspecified, at least four (to >100) equally parsimonious reconstructions were available for each node, and all nodes were reconstructed as a wide distribution that include all or most unit areas, including HOL (Table 1). However, it is

Table 1 – Ancestral area reconstructions inferred from dispersal–vicariance analyses

Node	Maxareas				
	10 (unspecified)	6[a]	7	8	9
1	ABCDEFGHIJ	ACI	ACI/ABCEFIJ	47 combinations (3–8)[d]	10 combinations (9)
2	6 combinations (7–10)	A	7 combinations (1–7)[b]	>100 combinations (1–8)[b]	47 combinations (7–9)
3	4 combinations (8–10)	A	5 combinations (1–7)[b]	>100 combinations (1–8)[b]	31 combinations (7–9)
4	8 combinations (7–10)	A	9 combinations (1–7)[b]	>100 combinations (1–8)[b]	59 combinations (6–9)
5	4 combinations (7–9)	AI	5 combinations (2–6)[c]	92 combinations (2–8)[c]	28 combinations (6–9)
6	AI/CI/ACI	CI/ACI	AI/CI/ACI	AI/CI/ACI	AI/CI/ACI
7	A/C/AC	A/AC	A/C/AC	A/C/AC/CD/ACD	A/C/AC
8	A/AC/ACD	A	A/AC/ACD	A/AC/AD/ACD	A/AC/ACD
9	6 combinations (7–9)	A	7 combinations (1–6)[b]	>100 combinations (1–8)[b]	42 combinations (6–9)
10	8 combinations (6–9)	28 combinations (2–6)	31 combinations (2–6)	>100 combinations (2–8)	56 combinations (5–9)
11	24 combinations (4–8)	A/AI/ACI	A/AI/CI/ACI	>100 combinations (1–8)[e]	>100 combinations (3–8)
12	8 combinations (4–7)	32 combinations (1–6)	33 combinations (1–7)	61 combinations (1–7)	32 combinations (3–7)
13	12 combinations (4–7)	77 combinations (2–6)	78 combinations (2–7)	92 combinations (2–7)	48 combinations (3–7)
14	4 combinations (3–5)	15 combinations (2–5)	15 combinations (2–5)	15 combinations (2–5)	12 combinations (2–5)
15	DFH/ADFH	7 combinations (2–4)	7 combinations (2–4)	7 combinations (2–4)	6 combinations (2–4)
16	DH/ADH	AH/DH/ADH	AH/DH/ADH	AH/DH/ADH	AH/DH/ADH
17	BF/BFJ	BF/FJ/BFJ	BF/FJ/BFJ	BF/FJ/BFJ	BF/FJ/BFJ

Reconstructions were conducted by changing the 'maxareas' option of DIVA from two to ten (unspecified). Two or more characters without a space indicate that the ancestors were widespread across those areas. Characters separated by a slash indicate the alternative equally parsimonious reconstruction. When more than three equally parsimonious reconstructions are available, the number of possible combinations is shown. Numbers in parentheses indicate the minimum and maximum number of unit areas for the equally parsimonious reconstructions. Node numbers and abbreviations of areas correspond to those of Fig 3.

a Reconstructions on nodes 1–9 were identical under the 'maxareas' settings from two to six except for node 1 and 6, which were reconstructed respectively as A/AC/AI and AI/CI under the 'maxareas' = 2.

b Only reconstruction with one unit area was A.

c Only reconstruction with two unit areas was AI.

d Only reconstruction with three unit areas was ACI.

e Equally parsimonious reconstructions included A, AI, CI, and ACI.

difficult to distinguish whether the results actually supported a wide ancestral distribution or they are the analytical artefacts of DIVA having unreliable reconstructions at basal nodes. However, the reconstructions were more consistent under the 'maxareas' constraints from two to eight (Table 1). For example, nodes 2–4 were either unambiguously reconstructed as AUS ('maxareas' = 2–6) or AUS was suggested as one of the most parsimonious reconstructions ('maxareas' = 7–8; Table 1). This suggests the origin and initial diversification of the ECM-*Hysterangiales* occurred in AUS, or eastern Gondwana, and range expansion of the ECM-*Hysterangiales* was the result of northward movement from the Southern Hemisphere.

DIVA was unable to resolve the ancestral areas for many nodes in the *Aroramyces* clade (Table 1). However, under the 'maxareas' constraints, node 9 was either unambiguously reconstructed as AUS ('maxareas' = 2–6) or AUS was suggested as one of the most parsimonious reconstructions ('maxareas' = 7–8; Table 1). If this scenario is correct, this suggests that range expansion of the ECM-*Hysterangiales* to the Northern Hemisphere happened more than once: once at node 5 and at least one more time at nodes 10–13. This is consistent with the polyphyly of Northern Hemisphere taxa (Figs 2 and 3), and that the phylogeny of *Hysterangiales* cannot be explained strictly by vicariance.

The ancestral areas of the *Hysterangium* II clade were unambiguously reconstructed for all nodes throughout the different 'maxareas' constraints (Fig 3). The reconstructions suggest that the common ancestor was distributed in AUS and SSAM, and the (potential) vicariant event caused a speciation into two lineages: one restricted to AUS and the other restricted to SSAM. Further range expansion to NZ (in two separate lineages) and to PNG were followed by another potential vicariant event. Because NZ was separated from Gondwana earlier than AUS and SSAM (Fig 1), two potential vicariant events depicted in Fig 3 (AE to A & E *versus* AC to A & C) are not compatible, unless the alternative geological hypothesis by Hallam (1994) is postulated (see Materials and methods, *Geological scenario*). Because of short branch lengths observed in the clade, phylogenetic patterns (e.g. NZ taxa deeply nested within AUS taxa), and the results of the ancestral area reconstructions, we hypothesize that NZ taxa in the *Hysterangium* II clade are the result of long-distance, transoceanic dispersal from AUS.

Parsimony-based tree fitting

Twenty-two equally parsimonious area cladograms were recovered from the phylogeny of *Hysterangiales* using TreeFitter (Fig 4), but none corresponded to a topology identical to the geological history of Pangaean break up (Fig 1). Among the 22 area cladograms, there were only two consistent patterns, i.e. sister relationships of AUS and NZ, and SEA and PNG (Fig 4A), both of which are incongruent with geological history (Fig 1). The results suggest that there have been long-distance dispersal events between AUS and NZ, and SEA and PNG. However, the searches for the best area cladograms were sensitive to the event cost settings. Different sets of area

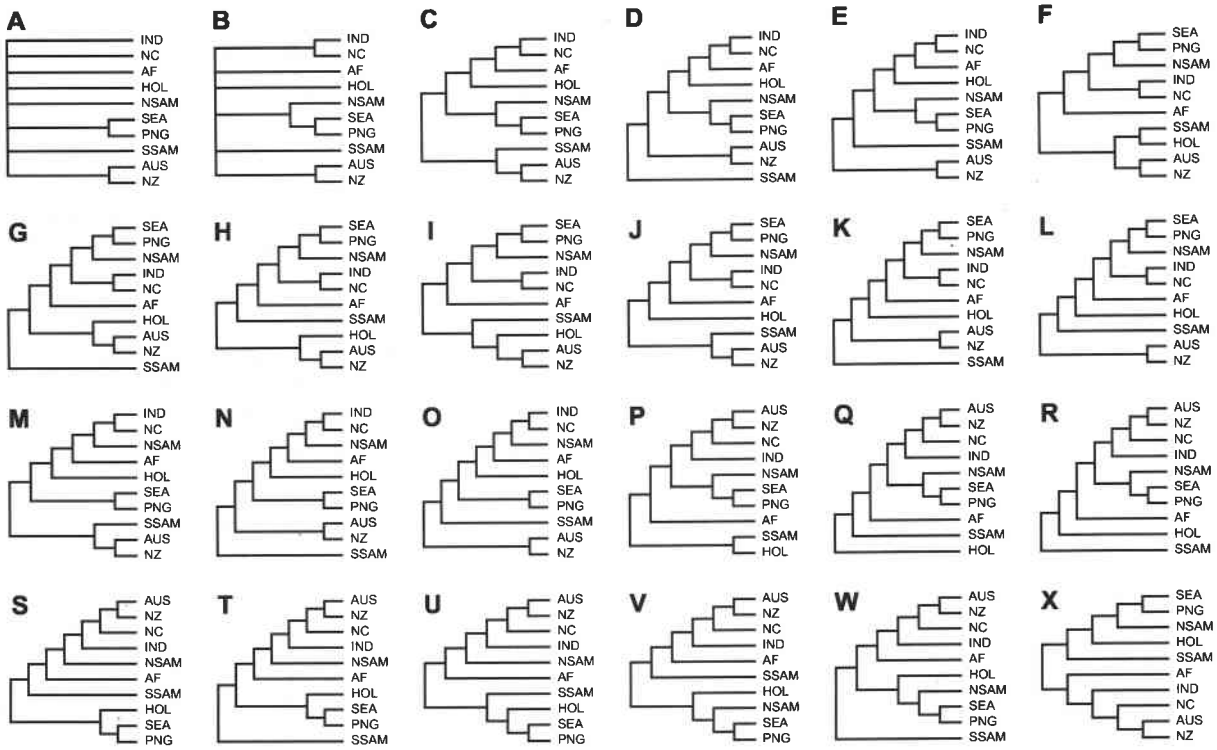


Fig 4 – The most parsimonious area cladograms inferred from the taxon-area cladogram in Fig 3. Strict consensus (A) and 50 % majority rule consensus (B) of 22 equally parsimonious area cladograms. (C)–(X) Equally parsimonious area cladograms. For abbreviations of areas, see Fig 1.

cladograms were obtained in each event cost settings, and the 22 trees obtained under the default cost settings (Analysis 1 of Table 2) were always suboptimal in terms of tree lengths (data not shown), except for analyses 9–13 and 15–16 (Table 2), which found more parsimonious trees than could be stored in memory. Despite these differences, most area cladograms obtained under a range of event cost settings consistently showed the sister area relationships of AUS and NZ, and SEA and PNG (data not shown).

Randomization tests showed that the fit between the phylogeny of *Hysterangiales* (Fig 3) and the geological history (Fig 1) was statistically not significant (Table 2). The results were consistent under the different event cost settings (Table 2). This suggests that the fit between the phylogeny of *Hysterangiales* and geological history could happen by chance, and that current distribution of *Hysterangiales* cannot be fully explained by Pangaean break up. The number of inferred biogeographical events also was relatively stable throughout the range of the event cost settings we have investigated (Table 2). In general, low number of vicariance (four or less) and extinction (11 or less) events and higher occurrence of dispersal (20 or more) events were inferred (Table 2). Only exceptional results were observed in the analysis 4, in which more vicariance (14) and extinction (59) events than dispersal (3) events were inferred (Table 2). In this analysis, dispersal events received a much higher cost than extinction (and vicariance) events (Table 2). This weighting scheme was identified as the most appropriate model to analyze the 'cospeciation–sorting pattern' (Ronquist 2003), which in a biogeographical context could be referred to as vicariance–extinction pattern. Although none of the randomization tests showed significant results, the lowest P-value (though well above 0.05) was obtained under this model (Table 2). We consider the results inconclusive, but it might be a weak indication that vicariance events are at least partially responsible for the current distribution of *Hysterangiales*.

Ancestral ectomycorrhizal host reconstructions

The phylogeny of *Hysterangiales* revealed many closely related species of *Hysterangiales* did not share the same host families (Fig 2). Most major clades within the ECM-*Hysterangiales* clade were represented by two or more ectomycorrhizal hosts, except the *Mesophelliaceae* clade, which is strictly associated with *Eucalyptus* (*Myrtaceae*). The results also indicate that frequent host shifts occurred during the evolution of *Hysterangiales* (Fig 5). However, host shifts are not necessarily between the two closely related groups of plants. For example, no host shifts between *Nothofagaceae* and *Fagaceae*, both of which belong to the order *Fagales*, were observed (Fig 5).Conversely, host shifts between distantly related plants, e.g. *Pinaceae* and *Fagaceae*, were frequently observed. This suggests that the phylogenies of *Hysterangiales* and its ectomycorrhizal host plants do not follow cospeciation patterns.

The host–fungus associations closely correlate with the current geographic distributions. That is, most Northern

Table 2 – Searches for the best area cladograms and frequency of events inferred using various event costs

Analysis no.	Event cost[a]				No. MPAC[b]	TL-MP[d]	TL-Geo[e]	Fit[f]	No. of inferred event[g]			
	Vic	Dup	Dis	Ext					Vic	Dup	Dis	Ext
1[h]	0	0	2	1	22	30	44	0.486	3–4	44–46	21–22	0–2
2	0	0	1	1	40	17	22	0.310	3–4	44–45	22	0
3	0	0	1	2	36	18	22	0.287	3–4	44–45	22	0
4	0	0	4	1	7	43	71	0.207	14	53	3	59
5	0	0	1	4	36	18	22	0.288	3–4	44–45	22	0
6	1	0	4	2	1	59	91	0.339	3	44–45	21–22	0–2
7	1	0	2	2	6	43	47	0.213	3	45	22	0
8	1	0	2	4	6	43	47	0.215	3	45	22	0
9	2	0	1	1	100K[c]	25	25	1	0	45	25	0
10	2	0	2	1	100K[c]	50	50	1	0–3	45–49	21–25	0–8
11	2	0	1	2	100K[c]	25	25	1	0	45	25	0
12	4	0	1	2	100K[c]	25	25	1	0	45	25	0
13	4	0	2	1	100K[c]	50	50	1	0	45–49	21–25	0–8
14	4	0	4	1	5	74	91	0.271	0	50	20	11
15	4	0	1	4	100K[c]	25	25	1	0	45	25	0
16	4	0	1	1	100K[c]	25	25	1	0	45	25	0

a Event costs for Vic, vicariance; Dup, duplication; Dis, dispersal; Ext, extinction.
b Number of equally parsimonious area cladograms obtained under the event cost settings implemented.
c More trees were found than could be stored in memory.
d Tree length (total cost) of the most parsimonious area cladograms obtained under the event cost settings implemented.
e Tree length (total cost) of the geological area cladograms (Fig 1B) obtained under the event cost settings implemented.
f P-value of fit between the taxon-area cladogram (Fig 3) and geological area cladogram (Fig 1B) tested by permutation of the areas in the geological area cladogram. Results are considered significant when $P < 0.05$.
g Maximum and minimum number of events (Vic, vicariance; Dup, duplication; Dis, dispersal; Ext, extinction) inferred by fitting the taxon-area cladogram (Fig 3) to geological area cladogram (Fig 1B).
h Default settings of TreeFitter.

Hemisphere species associate with *Fagaceae* or *Pinaceae* and Southern Hemisphere species associate with *Myrtaceae* or *Nothofagaceae*. Bidirectional host shifts were observed only between *Pinaceae* and *Fagaceae* (one to seven shifts in both directions), or between *Myrtaceae* and *Nothofagaceae* (two shifts from *Nothofagaceae*, five shifts from *Myrtaceae*) (Fig 5). All other patterns for host shifts, except for the shift from *Myrtaceae* to *Fagaceae* with three possible steps, occurred only once. This indicates that the biogeography of *Hysterangiales* has been shaped by host availability, i.e. co-occurrence with host trees in a common geographic area, and not by a pattern of speciation based on strict host-tracking.

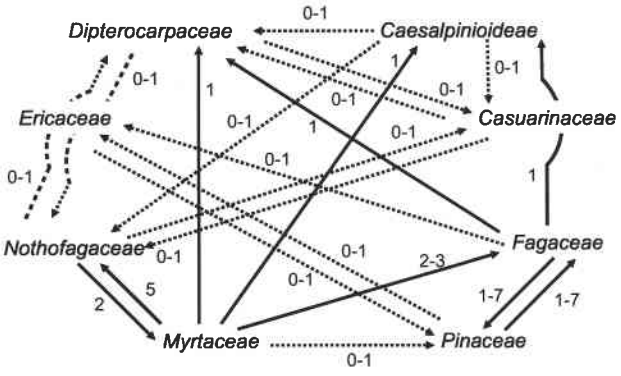


Fig 5 – Frequency of ectomycorrhizal host shifts inferred from unweighted parsimony reconstructions. Arrows indicate the direction of host shift. Numbers on arrows indicate the minimum and maximum possible steps. Host shifts with possible zero occurrences are indicated by dotted arrows. Ectomycorrhizal hosts are coded in eight states, each state corresponding to host family.

Myrtaceae was reconstructed as the most ancestral host of the ECM-*Hysterangiales* (Fig 2). Furthermore, the common ancestors of the *Hysterangium* II, *Mesophelliaceae* and *Gallaceaceae* clades were all reconstructed as *Myrtaceae* (Fig 2), and taxa associated with *Nothofagaceae* were confined to the more terminal clades (Fig 2). This contrasts with the traditional view that considered *Nothofagus* an ancestral host for many ectomycorrhizal fungi in the Southern Hemisphere, and host shifts to *Myrtaceae* happened more recently as *Myrtaceae* (especially *Eucalyptus*) expanded its distribution range (Malloch *et al.* 1980).

If *Myrtaceae* is indeed the most ancestral ectomycorrhizal host, the most parsimonious explanation is that *Hysterangiales* is as old as, or younger than, *Myrtaceae*. A molecular clock study suggested that the family originated less than 90 Mya (Sytsma *et al.* 2004). In this scenario, *Hysterangiales* could be even younger because not all members of *Myrtaceae* are ectomycorrhizal (Wang & Qiu 2006). This is inconsistent with the hypothesis that *Hysterangiales* originated before the break up of Gondwana. Furthermore, age estimates for any known ectomycorrhizal hosts of *Hysterangiales* are significantly younger than the initial break up of Gondwana (Schneider *et al.* 2004; Wang *et al.* 2000; Wikström *et al.* 2001).

Therefore, for *Hysterangiales* to be older than the Gondwanan break up, *ad hoc* hypotheses, such as novel ectomycorrhizal association with ancient plants or parallel gains of ectomycorrhizal habit, must be postulated.

Biogeography of animals with emphasis on mycophagy

The association between small animals and truffle-like fungi can be seen in many areas of the world. In the Northern Hemisphere, small mammals especially rodents (e.g. squirrels, mice, voles), and in the Southern Hemisphere several marsupials (e.g. potoroo, bettong, bandicoot) eat significant amount of fruiting bodies of truffle-like fungi as part of their diet (Castellano *et al.* 1989; Claridge 2002; Fogel & Trappe 1978; Malajczuk *et al.* 1987b). Because of the tight interaction between mycophagous animals and truffle-like fungi, some sort of co-evolutionary scenario is a possibility.

Two major animal groups containing mycophagous animals are *Eutheria* (including extant placental mammals) and *Metatheria* (including extant marsupials), and they are probably the sister groups (Phillips & Penny 2003). Although the age estimates for the origin of *Eutheria* and *Metatheria* vary from Permian to Mid-Cretaceous (Bromham *et al.* 1999), fossil evidence suggests that both groups originated in Asia (Luo *et al.* 2003). They expanded their distribution to North America and then to South America at around the K/T boundary (Nilsson *et al.* 2003). *Metatheria* did not reach AUS until Late Cretaceous or early Tertiary because the first appearance of marsupial fossils in AUS is from 55 Mya (Bromham *et al.* 1999). At that time, SSAM and AUS were still connected through Antarctica, so that marsupials could expand their range *via* Antarctica. This pattern is also consistent with the mammal phylogeny, which shows a more basal position of Asian and North American taxa with terminal AUS taxa (Luo *et al.* 2003). This means that one of the most important spore vectors for truffle-like fungi may not have been present in AUS until relatively recently, suggesting that mycophagous animals, e.g. marsupials, may not be the most important factor for the initial range expansion of *Hysterangiales*.

Although mycophagy by small animals, especially rodents and small marsupials, is usually emphasized for truffle–animal interactions, there may be other organisms that could serve as important spore vectors. For example, mycophagy by arthropods is well-documented for many groups of fungi (Martin 1979). The order *Phallales*, which is closely related to *Hysterangiales*, is one of the prime examples of insect mycophagy (Hosaka *et al.* 2006). Its phylogenetic affinity with *Hysterangiales* leaves a possibility that the ancestor of *Hysterangiales* was at least partially dependent on arthropods for spore dispersal. Exactly how arthropods are important for spore dispersal of *Hysterangiales* compared with mammals/marsupials is unclear. Some truffle-like fungi are known to emit chemical compounds to attract various insects (Pacioni *et al.* 1991), which implies that the interactions between *Hysterangiales* and arthropods should not be ignored.

There are some examples of mycophagy by birds (Simpson 2000; Claridge 2002) and deer (Ashkannejhad & Horton 2006). Lilleskov & Bruns (2005) suggested the potential importance of linkages between below ground and above-ground food webs, e.g. predatory animals as secondary spore vectors by feeding on mycophagous animals. In addition, abiotic factor, such as rainwater, may be equally important for physical movement of spores (Fogel 1976). These data imply that spores of truffle-like fungi could potentially be dispersed for long distances. Recent population genetics studies suggest that although the spores of truffle-like fungi can be dispersed for a few kilometers in continuous forest with a presence of mycophagous mammals (Kretzer *et al.* 2005), the existence of oceans (Wedén *et al.* 2004), mountain ranges (Murat *et al.* 2004), and valley systems (Grubisha *et al.* 2007) could be effective dispersal barriers.

Fossil records of fungi

Fossil records give us important clues for understanding the ancient fungal biota. Fungal fossils from the Proterozoic era are documented by Butterfield (2005), but the phylogenetic position of these fossils is difficult to evaluate. From the Paleozoic era, *Glomeromycota* (460 Mya; Redecker *et al.* 2000) and *Ascomycota* (400 Mya; Taylor *et al.* 2005) have been documented. One *Basidiomycota* fossil is also known as hyphae with clamp connections (290 Mya; Dennis 1970). However, it cannot be assigned to any specific group of *Basidiomycota*. The oldest fossil records of *Agaricomycetes* are from the Cretaceous period *ca* 100 Mya (Hibbett *et al.* 1997; Poinar & Brown 2003). A possibility of *Palaeoclavaria* (Poinar & Brown 2003) being a close relative of *Clavariadelphus* in *Phallomycetidae* (Hosaka *et al.* 2006) cannot be discarded, but is difficult to evaluate. There are much more recent fossils of ectomycorrhizas from the Eocene (LePage *et al.* 1997) and *Geastraceae*, which belongs to *Phallomycetidae*, and therefore is closely related to *Hysterangiales* (Hosaka *et al.* 2006), from the Miocene (Magallon-Puebla & Cevallos-Ferriz 1993). These fossil records cast no doubt about the existence of fungi in the Paleozoic era. However, to discuss the biogeography of *Hysterangiales*, scarce fossil records of mushroom-forming fungi give us little insight into the ancient mycobiota. Based on the fossil records, a Jurassic or older origin of *Agaricomycetes* is likely, but no direct evidence suggests the divergence time of *Hysterangiales*.

Conclusions

In conclusion, the phylogeny of *Hysterangiales*, as well as the results of ancestral area reconstructions, suggests that ectomycorrhizal lineages of *Hysterangiales* originated in AUS or eastern Gondwana with subsequent range expansions to the Northern Hemisphere. Because of the ambiguity associated with different constraints and event cost settings of biogeographical analyses, the origin of *Hysterangiales* and the timing of its range expansions are still inconclusive. However, consideration of the age of host trees and known mammalian dispersers are most consistent with a post-Pangean origin of *Hysterangiales*. Regardless, models for both ancient and more recent origins of *Hysterangiales* are consistent with hypogeous, truffle-like fungi being capable of transoceanic dispersal. Given its hypogeous and ectomycorrhizal habit, it is intriguing to know how and whether *Hysterangiales* could carry out such long-distance dispersal. If correct, then our initial hypothesis

that truffle-like fungi have more limited spore dispersal as compared with their epigeous counterparts is incorrect. Future research should focus on more sampling from the presently underrepresented areas, e.g. AF, IND, and Asia, to further clarify the biogeographical patterns of *Hysterangiales*. Additional paleontological studies of fossil ectomycorrhizas and mushroom-forming fungi from more ancient geologic periods would be of great benefit. Because no definitive fossils of *Hysterangiales* are currently known, more robust age estimates will have to be obtained with well-supported, higher-level phylogeny for *Agaricomycetes* using the external fossil records.

Acknowledgements

We thank all herbaria listed in Supplementary Material Table 1 and the following individuals for providing specimens: Cathie Aime, Youko Andoh, Bruce Caldwell, Efren Cazares, Lisa Grubisha, Steve Miller, Hiromi Sasaki, and James Trappe. We also acknowledge the following individuals for facilitating the field trips: Teresa Lebel and Nicole Middleton in Australia; Tanguy Jaffré, David Kurpisz, Joseph Manaute, and the park rangers of Parc Provincial de la Rivière Bleue in New Caledonia; Ross Beever, Jessica Beever, and Nick Singer in New Zealand; Roy Banka, Cepas Kayo, Charles Pepe, James Robins, Barbara Roy, Yoshika Sawairi, Roland Treu, Debra Wright, and the local people in the village of Maimafu in Papua New Guinea. This research project was financed in part by a grant to K.H. from the National Science Foundation, Doctoral Dissertation Improvement Grant (DEB-0309412) and by the NSF grant (DEB 9201421) to M.A.C.

Supplementary material

Supplementary data associated with this article can be found in the online version, at doi: 10.1016/j.mycres.2007.06.004.

REFERENCES

Agerer R, Iosifidou P, 2004. Rhizomorph structures of *Hymenomycetes*: a possibility to test DNA-based phylogenetic hypotheses? In: Agerer R, Piepenbring M, Blanz P (eds), *Frontiers in Basidiomycote Mycology* IHW-Verlag, Eching, pp. 249–302.

Ashkannejhad S, Horton TR, 2006. Ectomycorrhizal ecology under primary succession on coastal sand dunes: interactions involving *Pinus contorta*, suilloid fungi and deer. *New Phytologist* **169**: 345–354.

Binder M, Hibbett DS, 2002. Higher-level phylogenetic relationships of homobasidiomycetes (mushroom-forming fungi) inferred from four rDNA regions. *Molecular Phylogenetics and Evolution* **22**: 76–90.

Bromham L, Phillips MJ, Penny D, 1999. Growing up with dinosaurs: molecular dates and the mammalian radiation. *Trends in Ecology and Evolution* **14**: 113–118.

Butterfield NJ, 2005. Probable Proterozoic fungi. *Paleobiology* **31**: 165–182.

Castellano MA, 1990. The new genus *Trappea* (*Basidiomycotina*, *Hysterangiaceae*), a segregate from *Hysterangium*. *Mycotaxon* **38**: 1–9.

Castellano MA, 1999. *Hysterangium*. In: Cairney JWG, Chambers SM, Cairney SW (eds), *Ectomycorrhizal Fungi: key genera in profile*. Springer-Verlag, New York, pp. 311–323.

Castellano MA, Beever RE, 1994. Truffle-like *Basidiomycotina* of New Zealand: *Gallacea*, *Hysterangium*, *Phallobata*, and *Protubera*. *New Zealand Journal of Botany* **32**: 305–328.

Castellano MA, Muchovej JJ, 1996. Truffle-like fungi from South America: *Hysterangium sensu lato*. *Mycotaxon* **57**: 329–345.

Castellano MA, Trappe JM, Maser Z, Maser C, 1989. *Key to Spores of the Genera of Hypogeous Fungi of North Temperate Forests with Special Reference to Animal Mycophagy*. Mad River Press, Eureka, CA.

Castellano MA, Verbeken A, Walleyn R, Thoen D, 2000. Some new or interesting sequestrate *Basidiomycota* from African woodlands. *Karstenia* **40**: 11–21.

Chapela IH, Garbelotto M, 2004. Phylogeography and evolution in matsutake and close allies inferred by analyses of ITS sequences and AFLPs. *Mycologia* **96**: 730–741.

Claridge AW, 2002. Ecological role of hypogeous ectomycorrhizal fungi in Australian forests and woodlands. *Plant and Soil* **244**: 291–305.

Coetzee MPA, Wingfield BD, Bloomer P, Ridley GR, Wingfield MJ, 2003. Molecular identification and phylogeny of *Armillaria* isolates from South America and Indo-Malaysia. *Mycologia* **95**: 285–293.

Cook LG, Crisp MD, 2005. Directional asymmetry of long-distance dispersal and colonization could mislead reconstructions of biogeography. *Journal of Biogeography* **32**: 741–754.

Dell B, Malajczuk N, Grove TS, Thomson G, 1990. Ectomycorrhiza formation in *Eucalyptus*. IV. Ectomycorrhizas in the sporocarps of the hypogeous fungi *Mesophellia* and *Castorium* in Eucalypt forest of Western Australia. *New Phytologist* **114**: 449–456.

Dennis RL, 1970. A middle Pennsylvanian basidiomycete mycelium with clamp connections. *Mycologia* **62**: 579–584.

Fogel R, 1976. Ecological studies of hypogeous fungi. II. Sporocarp phenology in a western Oregon Douglas fir stand. *Canadian Journal of Botany* **54**: 1152–1162.

Fogel R, Trappe JM, 1978. Fungus consumption (mycophagy) by small animals. *Northwest Science* **52**: 1–31.

Grubisha LC, Bergemann SE, Bruns TD, 2007. Host islands within the California Northern Channel Islands create fine-scale genetic structure in two sympatric species of the symbiotic ectomycorrhizal fungus *Rhizopogon*. *Molecular Ecology* **16**: 1811–1822.

Hall TA, 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acids Symposium Series* **41**: 95–98.

Hallam A, 1994. *An Outline of Phanerozoic Biogeography*. Oxford University Press, Oxford.

Halling RE, 2001. Ectomycorrhizae: co-evolution, significance, and biogeography. *Annals of the Missouri Botanical Garden* **88**: 5–13.

Heckman DS, Geiser DM, Eidell BR, Stauffer RL, Kardos NL, Hedges SB, 2001. Molecular evidence for the early colonization of land by fungi and plants. *Science* **293**: 1129–1133.

Hedges SB, Blair JE, Venturi ML, Shoe JL, 2004. A molecular timescale of eukaryote evolution and the rise of complex multicellular life. *BMC Evolutionary Biology* **4**: 2–10.

Hibbett DS, 2001. Shiitake mushrooms and molecular clocks: historical biogeography of *Lentinula*. *Journal of Biogeography* **28**: 231–241.

Hibbett DS, Grimaldi D, Donoghue MJ, 1997. Fossil mushrooms from Miocene and Cretaceous ambers and the evolution of homobasidiomycetes. *American Journal of Botany* **84**: 981–991.

Hibbett DS, Gilbert LB, Donoghue MJ, 2000. Evolutionary instability of ectomycorrhizal symbioses in basidiomycetes. *Nature* **407**: 506–508.

Hosaka K, Bates ST, Beever RE, Castellano MA, Colgan W, Dominguez LS, Geml J, Giachini AJ, Kenney SR, Nouhra ER, Simpson NB, Spatafora JW, Trappe JM, 2006. Molecular

phylogenetics of the gomphoid-phalloid fungi with an establishment of the new subclass *Phallomycetidae* and two new orders. *Mycologia* **98**: 949–959.

Huelsenbeck JP, Ronquist F, 2001. MRBAYES: Bayesian inference of phylogenetic trees. *Bioinformatics* **17**: 754–755.

Huelsenbeck JP, Rannala B, Larget B, 2000. A Bayesian framework for the analysis of cospeciation. *Evolution* **54**: 352–364.

Huelsenbeck JP, Rannala B, Larget B, 2003. A statistical perspective for reconstructing the history of host–parasite associations. In: Page RDM (ed.), *Tangled Trees: phylogeny, cospeciation, and coevolution*. University of Chicago Press, Chicago, pp. 93–119.

Humpert AJ, Muench EL, Giachini AJ, Castellano MA, Spatafora JW, 2001. Molecular phylogenetics of *Ramaria* and related genera: evidence from nuclear large subunit and mitochondrial small subunit rDNA sequences. *Mycologia* **93**: 465–477.

James TY, Moncalvo J-M, Li S, Vilgalys R, 2001. Polymorphism at the ribosomal DNA spacers and its relation to breeding structure of the widespread mushroom *Schizophyllum commune*. *Genetics* **157**: 149–161.

James TY, Kauff F, Schoch C, Matheny PB, Hofstetter V, Cox CJ, Celio G, Gueidan C, Fraker E, Miadlikowska J, Lumbsch HT, Rauhut A, Reeb V, Arnold AE, Amtoft A, Stajich JE, Hosaka K, Sung GH, Johnson D, O'Rourke B, Binder M, Curtis JM, Slot JC, Wang Z, Wilson AW, Schüßler A, Longcore JE, O'Donnell K, Mozley-Standridge S, Porter D, Letcher PM, Powell MJ, Taylor JW, White MM, Griffith GW, Davies DR, Sugiyama J, Rossman AY, Rogers JD, Pfister DH, Hewitt D, Hansen K, Hambleton S, Shoemaker RA, Kohlmeyer J, Volkmann-Kohlmeyer B, Spotts R, Serdani M, Crous PW, Hughes KW, Matsuura K, Langer E, Langer G, Untereiner WA, Lücking R, Büdel B, Geiser DM, Aptroot A, Buck WR, Cole MS, Diederich P, Hillis DM, Printzen C, Schmitt I, Schultz M, Yahr R, Zavarzin A, Hibbett DS, Lutzoni F, McLaughlin DJ, Spatafora JW, Vilgalys R, 2006. Reconstructing the early evolution of the fungi using a six gene phylogeny. *Nature* **443**: 818–822.

Jin J, Hughes KW, Petersen RH, 2001. Biogeographical patterns in *Panellus stypticus*. *Mycologia* **93**: 309–316.

Knapp M, Ströckler K, Havell D, Delsuc F, Sebastiani F, Lockhart PJ, 2005. Relaxed molecular clock provides evidence for long-distance dispersal of *Nothofagus* (southern beech). *PLoS Biology* **3**: 38–43.

Kretzer AM, Bruns TD, 1999. Use of *atp6* in fungal phylogenetics: an example from the *Boletales*. *Molecular Phylogenetics and Evolution* **13**: 483–492.

Kretzer AM, Dunham S, Molina R, Spatafora JW, 2005. Patterns of vegetative growth and gene flow in *Rhizopogon vinicolor* and *R. vesiculosus* (*Boletales*, *Basidiomycota*). *Molecular Ecology* **14**: 2259–2268.

LePage BA, Currah RS, Stockey RA, Rothwell GW, 1997. Fossil ectomycorrhizae from the middle Eocene. *American Journal of Botany* **84**: 410–412.

Lilleskov EA, Bruns TD, 2005. Spore dispersal of a resupinate ectomycorrhizal fungus, *Tomentella sublilacina*, via soil food webs. *Mycologia* **97**: 762–769.

Liu YJ, Whelen S, Hall BD, 1999. Phylogenetic relationships among ascomycetes: evidence from and RNA polymerase II subunit. *Molecular Biology and Evolution* **16**: 1799–1808.

Lu X, Malajczuk N, Brundrett M, Dell B, 1999. Fruiting of putative ectomycorrhizal fungi under blue gum (*Eucalyptus globules*) plantations of different ages in Western Australia. *Mycorrhiza* **8**: 255–261.

Luo ZX, Ji Q, Wible JR, Yuan CX, 2003. An early Cretaceous tribosphenic mammal and metatherian evolution. *Science* **302**: 1934–1940.

Lutzoni F, Kauff F, Cox CJ, McLaughlin D, Celio G, Dentinger B, Padamsee M, Hibbett D, James TY, Baloch E, Grube M, Reeb V, Hofstetter V, Schoch C, Arnold AE, Miadlikowska J, Spatafora J, Johnson D, Hambleton S, Crockett M, Shoemaker R, Sung GH, Lücking R, Lumbsch T, O'Donnell K, Binder M, Diederich P, Ertz D, Gueidan C, Hansen K, Harris RC, Hosaka K, Lim YW, Matheny B, Nishida H, Pfister D, Rogers J, Rossman A, Schmitt I, Sipman H, Stone J, Sugiyama J, Yahr R, Vilgalys R, 2004. Assembling the Fungal Tree of Life: progress, classification, and evolution of subcellular traits. *American Journal of Botany* **91**: 1446–1480.

Maddison DR, Maddison WP, 2003. *Macclade*. Version 4.06. *Analysis of Phylogeny and Character Evolution*. Sinauer Associates, Sunderland, MA.

Magallon-Puebla S, Cevallos-Ferriz SRS, 1993. A fossil earthstar (*Geastraceae*; *Gasteromycetes*) from the Late Cenozoic of Puebla, Mexico. *American Journal of Botany* **80**: 1162–1167.

Malajczuk N, Dell B, Bougher NL, 1987a. Ectomycorrhiza formation in *Eucalyptus*. III. Superficial ectomycorrhizas initiated by *Hysterangium* and *Cortinarius* species. *New Phytologist* **105**: 421–428.

Malajczuk N, Trappe JM, Molina R, 1987b. Interrelationships among some ectomycorrhizal trees, hypogeous fungi and small mammals: Western Australian and northwestern American parallels. *Australian Journal of Ecology* **12**: 53–55.

Malloch DW, Pirozynski KA, Raven PH, 1980. Ecological and evolutionary significance of mycorrhizal symbioses in vascular plants (A Review). *Proceedings of the National Academy of Sciences, USA* **77**: 2113–2118.

Martin F, Díez J, Dell B, Delaruelle C, 2002. Phylogeography of the ectomycorrhizal *Pisolithus* species as inferred from nuclear ribosomal DNA ITS sequences. *New Phytologist* **153**: 345–357.

Martin MM, 1979. Biochemical implications of insect mycophagy. *Biological Reviews* **54**: 1–21.

Matheny PB, 2005. Improving phylogenetic inference of mushrooms with RPB1 and RPB2 nucleotide sequences (*Inocybe*; *Agaricales*). *Molecular Phylogenetics and Evolution* **35**: 1–20.

McLoughlin S, 2001. The breakup history of Gondwana and its impact of pre-Cenozoic floristic provincialism. *Australian Journal of Botany* **49**: 271–300.

Metcalfe I, 1998. Paleozoic and Mesozoic geological evolution of the SE Asian region: multidisciplinary constraints and implications for biogeography. In: Hall R, Holloway JD (eds), *Biogeography and Geological Evolution of SE Asia*. Backbuys Publishers, Leiden, pp. 25–41.

Miller OK, Miller HH, 1988. *Gasteromycetes: morphological and development features with keys to the orders, families, and genera*. Mad River Press, Eureka, CA.

Molina R, Trappe JM, 1982. Patterns of ectomycorrhizal host specificity and potential among Pacific Northwest conifers and fungi. *Forest Science* **28**: 423–458.

Moyersoen B, Beever RE, Martin F, 2003. Genetic diversity of *Pisolithus* in New Zealand indicates multiple long-distance dispersal from Australia. *New Phytologist* **160**: 569–579.

Müller WR, Agerer R, 1996. *Hysterangium crassirhachis* Zeller & Dodge + *Pseudotsuga menziesii* (Mirb.) Franco. *Descriptions of Ectomycorrhizae* **1**: 29–34.

Murat C, Díez J, Luis P, Delaruelle C, Dupré C, Chevalier G, Bonfante P, Martin F, 2004. Polymorphism at the ribosomal DNA ITS and its relation to postglacial re-colonization routes of the Perigord truffle *Tuber melanosporum*. *New Phytologist* **164**: 401–411.

Nilsson MA, Gullberg A, Spotorno AE, Arnason U, Janke A, 2003. Radiation of extant marsupials after the K/T boundary: evidence from complete mitochondrial genomes. *Journal of Molecular Evolution* **57**: S3–S12.

Oda T, Tanaka C, Tsuda M, 2004. Molecular phylogeny and biogeography of the widely distributed *Amanita* species, *A. muscaria* and *A. pantherina*. *Mycological Research* **108**: 885–896.

Pacioni G, Bologna MA, Laurenzi M, 1991. Insect attraction by *Tuber*: a chemical explanation. *Mycological Research* **95**: 1359–1363.

Page RDM, 1990. Temporal congruence and cladistic analysis of biogeography and cospeciation. *Systematic Zoology* **39**: 205–226.

Page RDM, 1995. Parallel phylogenies: reconstructing the history of host-parasite assemblages. *Cladistics* **10**: 155–173.

Phillips MJ, Penny D, 2003. The root of the mammalian tree inferred from whole mitochondrial genomes. *Molecular Phylogenetics and Evolution* **28**: 171–185.

Poinar GO, Brown AE, 2003. A non-gilled hymenomycete in Cretaceous amber. *Mycological Research* **107**: 763–768.

Pole M, 1994. The New Zealand flora—entirely long-distance dispersal? *Journal of Biogeography* **21**: 625–635.

Raidl S, Agerer R, 1998. *Hysterangium stoloniferum* Tul. & Tul. + *Picea abies* (L.) Karst. *Descriptions of Ectomycorrhizae* **3**: 31–35.

Redecker D, Kodner R, Graham LE, 2000. Glomalean fungi from the Ordovician. *Science* **289**: 1920–1921.

Ree RH, Moore BR, Webb CO, Donoghue MJ, 2005. A likelihood framework for inferring the evolution of geographic range on phylogenetic trees. *Evolution* **59**: 2299–2311.

Rehner SA, Buckley E, 2005. A *Beauveria* phylogeny inferred from nuclear ITS and EF1-α sequences: evidence for cryptic diversification and links to *Cordyceps* teleomorphs. *Mycologia* **97**: 84–98.

Ronquist F, 1997. Dispersal–vicariance analysis: a new approach to the quantification of historical biogeography. *Systematic Biology* **46**: 195–203.

Ronquist F, 2003. Parsimony analysis of coevolving species associations. In: Page RDM (ed), *Tangled Trees: phylogeny, cospeciation, and coevolution*. University of Chicago Press, Chicago, pp. 22–64.

Sampson SD, Witmer LM, Forster CA, Krause DW, O'Connor PM, Dodson P, Ravoavy F, 1998. Predatory dinosaur remains from Madagascar: implications for the Cretaceous biogeography of Gondwana. *Science* **280**: 1048–1051.

Sanmartín I, Ronquist F, 2004. Southern Hemisphere biogeography inferred by event-based models: plant versus animal patterns. *Systematic Biology* **53**: 216–243.

Sanmartín I, Enghoff H, Ronquist F, 2001. Patterns of animal dispersal, vicariance and diversification in the Holarctic. *Biological Journal of the Linnean Society* **73**: 345–390.

Sanmartín I, Wanntorp L, Winkworth C, 2007. West Wind Drift revisited: testing for directional dispersal in the Southern Hemisphere using event-based tree fitting. *Journal of Biogeography* **34**: 398–416.

Schneider H, Schuettpelz E, Pryer KM, Cranfill R, Magallón S, Lupia R, 2004. Ferns diversified in the shadow of angiosperms. *Nature* **428**: 553–557.

Shen Q, Geiser DM, Royse DJ, 2002. Molecular phylogenetic analysis of *Grifola frondosa* (maitake) reveals a species partition separating eastern North America and Asian isolates. *Mycologia* **94**: 472–482.

Shimodaira H, Hasegawa M, 1999. Multiple comparisons of log-likelihoods with applications to phylogenetic inference. *Molecular Biology and Evolution* **16**: 1114–1116.

Simpson JA, 2000. More on mycophagous birds. *Australian Mycologist* **19**: 49–51.

Stewart EL, Trappe JM, 1985. The new genus *Austrogautieria (Basidiomycotina)*, segregate from *Gautieria*. *Mycologia* **77**: 674–687.

Swofford DL, 2002. *PAUP*: phylogenetic analysis using parsimony (*and other methods)*. Version 4.0b10. Sinauer Associates, Sunderland, MA.

Sytsma KJ, Litt A, Zjhra ML, Pires JC, Nepokroeff M, Conti E, Walker J, Wilson PG, 2004. Clades, clocks, and continents: historical and biogeographical analysis of *Myrtaceae, Vochysiaceae*, and relatives in the Southern Hemisphere. *International Journal of Plant Sciences* **165**: S85–S105.

Taylor TN, Hass H, Kerp H, Krings M, Hanlin RT, 2005. Perithecial ascomycetes from the 400 million year old Rhynie chert: an example of ancestral polymorphism. *Mycologia* **97**: 269–285.

Templeton AR, 1983. Phylogenetic inference from restriction endonuclease cleavage site maps with particular reference to the evolution of humans and the apes. *Evolution* **37**: 221–244.

Thiers HD, 1984. The secotioid syndrome. *Mycologia* **76**: 1–8.

Thompson JD, Gibson TJ, Plewniak F, Jeanmougin F, Higgins DG, 1997. The Clustal X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucleic Acids Research* **25**: 4876–4882.

Vilgalys R, Hester M, 1990. Rapid genetic identification and mapping of enzymatically amplified DNA from several *Cryptococcus* species. *Journal of Bacteriology* **172**: 4238–4246.

Vilgalys R, Sun BL, 1994. Ancient and recent patterns of geographic speciation in the oyster mushroom *Pleurotus* revealed by phylogenetic analysis of ribosomal DNA sequences. *Proceedings of the National Academy of Sciences, USA* **91**: 4599–4603.

Wang B, Qiu Y-L, 2006. Phylogenetic distribution and evolution of mycorrhizas in land plants. *Mycorrhiza* **16**: 299–363.

Wang XQ, Tank DC, Sang T, 2000. Phylogeny and divergence times in *Pinaceae*: evidence from three genomes. *Molecular Biology and Evolution* **17**: 773–781.

Wedén C, Danell E, Camacho FJ, Backlund A, 2004. The population of the hypogeous fungus *Tuber aestivum* syn. *T. uncinatum* on the island of Gotland. *Mycorrhiza* **14**: 19–23.

White TJ, Bruns T, Lee S, Taylor JW, 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis MA, Gelfand DH, Sninsky JJ, White TJ (eds), *PCR Protocols: methods and applications*. Academic Press, San Diego, pp. 315–322.

Wikström N, Savolainen V, Chase MW, 2001. Evolution of the angiosperms: calibration the family tree. *Proceedings of the Royal Society of London, Series B* **268**: 2211–2220.

Wu Q-X, Mueller GM, Lutzoni FM, Huang Y-Q, Guo S-Y, 2000. Phylogenetic and biogeographic relationships of eastern Asian and eastern North American disjunct *Suillus* species (*Fungi*) as inferred from nuclear ribosomal RNA ITS sequences. *Molecular Phylogenetics and Evolution* **17**: 37–47.

Xiang Q-Y, Soltis DE, Soltis PS, 1998. The eastern Asian and eastern and western North American floristic disjunction: congruent phylogenetic patterns in seven diverse genera. *Moleculalr Phylogenetics and Evolution* **10**: 178–190.

Zervakis GI, Moncalvo J-M, Vilgalys R, 2004. Molecular phylogeny, biogeography and speciation of the mushroom species *Pleurotus cystidiosus* and allied taxa. *Microbiology* **150**: 715–726.