



Evolutionary origins of Gondwanan interactions: How old are *Araucaria* beetle herbivores?

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Studies of a variety of phenomena, ranging from rates of molecular substitution to rates of diversification, draw on estimates of geological age. Studies incorporating estimates of timing from fossils or other geological evidence are largely of relatively young, Tertiary divergences, to which older systems may provide useful comparisons. One apparently old assemblage comprises the beetle groups associated with the ancient genus *Araucaria* that share comparable, ostensibly Gondwanan distributions with their host. Our previous studies suggested a possibly Cretaceous age for *Araucaria* associations in bark beetles. However, the absence of confirmed bark beetle fossils earlier than the Tertiary has been taken as evidence of Cretaceous absence, and their confirmed phylogenetic position within the primitively angiosperm-feeding weevil family rules out pre-angiosperm, Jurassic origins. Nevertheless, an early shift from angiosperms to *Araucaria* seemed plausible in the light of *Araucaria* fossil history which spans the Mesozoic since the Jurassic. To resolve the phylogenetic affinities and to estimate divergence times of the Australian and South American bark beetle genera affiliated with *Araucaria* we analysed DNA sequences of nuclear and mitochondrial genes: protein coding elongation factor alpha, enolase and cytochrome oxidase I. The most parsimonious reconstruction of the host relationships of Tomicini from the combined dataset corroborates the ancestral association with the genus *Araucaria* of both South American and Australian Tomicini. Bayesian estimation of divergence times indicates that the divergence between the Australian and the South American *Araucaria*-feeding taxa occurred at the very latest in the Cretaceous/Paleocene border and that the age of the first Scolytinae–*Araucaria* association would then be during the later stages of the Late Cretaceous, while other known beetle/*Araucaria* associations are Jurassic.

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ADDITIONAL KEY WORDS: Tomicini – bark beetles – South America – Australia – phylogeny – cytochrome oxidase I – elongation factor 1 α – enolase – Bayesian inference – age estimation – Late Cretaceous divergence.

INTRODUCTION

The time over which species have been associated with particular hosts or geographic regions is relevant to study of a variety of phenomena, ranging from estimation of the absolute rate of nucleotide substitutions (Moran *et al.*, 1993; Peek, Vrijenhoek & Gaut, 1998), to assessing the apparent degree of adaptation (Farrell, 2001; Lutzoni & Pagel, 1997), evaluating the stability of mutualistic or parasitic interactions (Pellmyr & Leebens-Mack, 1999) and the diversity of radiations (Gillespie, Croom & Palumbi, 1994; Liebherr & Zimmerman, 1998; Sequeira *et al.*, 2000a; Sequeira, Normark & Farrell, 2000b). Studies of timing typically combine fossil or geological information with estimates of molecular divergence to calibrate systems ranging

up to the mid-Tertiary in age, over some 30 million years (Brower, 1994; Harrison & Crespi, 1999; Pellmyr & Leebens-Mack, 1999; Farrell, 2001; Sequeira *et al.*, 2000a; Gillespie *et al.*, 1994; Juan, Oromi & Hewitt, 1995, 1996; Juan *et al.*, 1998). Older associations (e.g. of Mesozoic age) seem much less frequently studied, perhaps because extinctions and shifts have obscured early patterns (but see Moran, 1996). However, it seems important to sample assemblages across as broad an array of time intervals as possible to provide the context for comparative studies.

A potential source of older comparisons may come from the disjunct biota associated with the now widely separated continents of South America, Africa and Australia of the south temperate zone. However, many groups occupy only two of these three formerly Gondwanan continents (precluding comparisons of their phylogenetic relationships to the temporal vicariance sequence predicted by the history of continental drift),

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Table 1. Some insect groups displaying disjunct distributions in the Southern Hemisphere. Initials in parentheses (in taxa 7, 14, 15 and 24 below) after the taxon name indicate the distribution is restricted to that area in the region: NZ: New Zealand, NC: New Caledonia; Tas: Tasmania

Rank Order: family	Disjunct group	Central/South America	South Africa	Australia Region	Reference
FAMILY					
(1) Lepidoptera: Palaephaltidae	Palaephaltidae	<i>Palaephaltus</i> <i>Apophaltus</i> <i>Plesiophaltus</i> <i>Metaphaltus</i> <i>Sesomata</i> <i>Caenominurus</i> <i>Chilecar</i>	—	<i>Azaleodes</i>	Davis, 1986; Nielsen, 1987
(2) Coleoptera: Caridae	Caridae		—	<i>Car</i> <i>Carodes</i> gen. sp.	Zimmerman, 1994 Kuschel, 1992 Riedel, 1995
SUBFAMILY					
(3) Diptera: Mydidae	Megascelinae	<i>Megascelus</i>	<i>Tongamyza</i>	<i>Neorhaphionides</i> <i>Alcides</i>	Yeates & Irwin, 1996* Coleman & Monteith, 1981
(4) Lepidoptera: Uramidae	Uraminae	<i>Urania</i>	—	<i>Nyctalemon</i> <i>Decogmus</i>	Darlington, 1965; Britton, 1970; Howden, 1981
(5) Coleoptera: Carabidae	Migadopinae	<i>Antractonomus</i> <i>Rhytidognathus</i> <i>Migadops</i> + four other genera	—	<i>Calypogonia</i> (Tas) <i>Stichonotus</i> (Tas) + four other genera	
(6) Coleoptera: Pyrochroidae	Pilipalpinae ²	<i>Pilipalpus</i> <i>Cycloderus</i>	—	<i>Paromarteon</i> <i>Temnopalpus</i> <i>Techmessa</i> <i>Techmessodes</i> <i>Exocalpus</i> <i>Binburrum</i> <i>Morpholycus</i> <i>Spilopyra</i> <i>Macrolema</i> <i>Richmondia</i> <i>Cheiloxena</i> <i>Bohumiliana</i> (NC) <i>Palophagus</i>	Pollock, 1995
(7) Coleoptera: Chrysomelidae	Spilopyrinae	<i>Hornius</i> <i>Stenomela</i>	—		Reid, 2000
(8) Coleoptera: Megalopodidae	Palophaginae ¹	<i>Palophagoides</i>	—		Kuschel & May, 1990, 1996
TRIBE					
(9) Lepidoptera: Papilionidae	Troidini	<i>Euryades</i>	—	<i>Cressida</i>	Parsons, 1996
(10) Coleoptera: Carabidae	Bembidiinae	<i>Xystosomus</i>	—	<i>Philis</i>	Baehr, 1995
(11) Coleoptera: Carabidae	Pelecini	<i>Pelecina</i>	Pelecina	Agoncina	Straneo & Ball, 1989
(12) Coleoptera: Carabidae	Homoloderini	several genera	—	several genera	Howden, 1981
(13) Coleoptera: Nemonychidae	Mecomacerini ¹	<i>Mecomacer</i> <i>Rhynchitomacerinus</i> <i>Rhynchitoplesius</i>	—	<i>Notomacer</i> <i>Aragomacer</i> <i>Eutactobius</i>	Kuschel, 1994; Kuschel & May, 1997
(14) Coleoptera: Nemonychidae	Rhinorhynchini ¹	<i>Brarus</i> <i>Nannomacer</i> <i>Atopomacer</i> (NA)	—	<i>Bunyaeus</i> <i>Rhinorhynchus</i> (NZ) <i>Basiliorhinus</i> <i>Basiliogeus</i> <i>Pagomacer</i>	Morrone, 1996

(15) Coleoptera: Belidae	Belini	<i>Homalocerus</i> <i>Trichophthalmus</i>	—	<i>Araibobelus</i> <i>Isacantha</i> <i>Isacanthodes</i> <i>Leptobelus</i> <i>Rhinotodes</i> <i>Rhinotia</i> (+ PNG and Pacific Islands)	Morrone, 1996
(16) Coleoptera: Curculionidae: Cossoninae	Araucariini ¹	<i>Araucarius</i>	<i>Amorphocerus</i> ³	<i>Coptocorynus</i> <i>Mastersinella</i> <i>Inosomus</i> <i>Xenocnema</i> <i>Hylurdectonus</i> <i>Pachycotes</i>	Kuschel, 1966; Morrone, 1997
(17) Coleoptera: Curculionidae: Scolytinae	Tomicini ¹	<i>Hylurgonotus</i> <i>Sinophloeus</i> <i>Xylechinossomus</i>	—		Wood, 1986; Wood & Bright, 1992
SUBTRIBE					
(18) Coleoptera: Belidae	Agnesioidina	<i>Dicordylus</i>	—	<i>Agathobelus</i> <i>Agnesioidis</i> <i>Apagobelus</i> <i>Basilobelus</i> <i>Macrobelus</i> <i>Cyrotypus</i> <i>Arhinobelus</i> <i>Brachybelus</i> <i>Habrobelus</i> <i>Pachyura</i> <i>Sphictobelus</i>	Morrone, 1996
(19) Coleoptera: Belidae	Pachyurina	<i>Atrachtuchus</i> <i>Callirhynchinus</i>	—		Morrone, 1996
GENUS					
(20) Neuroptera: Osmylidae	<i>Kempynus</i>	<i>Kampynus</i>	—	<i>Kempynus</i>	Wise, 1991
(21) Diptera: Apioceridae	<i>Apiocera</i>	<i>Anyperus</i> sub gen.	<i>Ripidosyrma</i> sub gen.	<i>Apiocera</i> sub gen.	Yeates & Irwin, 1996*
(22) Coleoptera: Lucanidae	<i>Sphaenognathus</i>	<i>Sphaenognathus</i>	—	<i>Sphaenognathus</i>	Moore, 1978
(23) Coleoptera: Buprestidae	<i>Curis</i>	<i>Curis</i>	—	<i>Curis</i>	Howden, 1981
(24) Coleoptera: Curculionidae	<i>Psepholax</i>	<i>Psepholax</i>	—	<i>Psepholax</i> (NZ)	Morrone, 1998

* Pangean distributions where the most plesiomorphic clades are found on a fragment of Laurasia and the next most plesiomorphic clades are found on fragments of Gondwanaland.

¹ Weevil and leaf beetle groups that feed on *Araucaria*.

² Group contains members in other land masses that formed part of West Gondwana (i.e. Madagascar).

³ New placement in Molytinae tribe Amorphocerini (Alonso-Zarazaga & Lyal, 1999).

and their distributions are thus assumed to reflect early to late Cretaceous divergences (depending on the pair), since postulation of more recent (i.e. Tertiary) origins would also posit very long distance dispersal that may be unrealistic for any but the most vagile groups of organisms. The many prominent examples of such disjunct distributions in vertebrates and plants have been the subject of debate for most of the century (e.g. Darlington, 1957, 1965; Nelson & Platnick, 1981) but an increasing number of insect groups have also been shown to have these disjunct distributions (Table 1), and thus provide a potentially useful comparative context within which to establish whether these share a common history, or arise in different ways, as would be suggested if they represent a wide range of ages. Indeed, some disparity in age is suggested by the range of taxonomic ranks of Gondwanan insect groups as these span levels from the family to the genus (Table 1), even within a given lineage. For example, of the very few groups of insects represented in all three areas, the mydid dipteran subfamily Mydinae and the genus *Apiocera* in the mydid subfamily Apiocerinae show phylogenetic concordance with the tectonic history of these three continents (Yeates & Irwin, 1996). However, most other groups of insects are restricted to only two of these Gondwanan continents, most often Australia and South America, a disjunction that suggests a minimum mid-Cretaceous age (Rosen, 1978; Smith, Hurley & Briden, 1981; Thayer, 1985; Pollock, 1995). While these distributions strongly suggest Gondwanan origins, more direct evidence on their ages (e.g. from fossils or molecular divergence) has not yet been assembled.

In comparison, several Gondwanan plant groups have substantial fossil records, potentially also illuminating the history of their associated insect faunas. Of the various Gondwanan plant groups (Craw, 1985; Humphries, Cox & Nielsen, 1986; Heads, 1990; Linder & Crisp, 1995; Renner, Foreman & Murray, 2000), the conifer genus *Araucaria* (Kuschel & May, 1990, 1996, 1997; Morrone, 1997; Farrell, 1998; Mecke, Engels & Galileo, 2000a; Mecke, Galileo & Engels, 2000b; Sequeira *et al.*, 2000b) and the angiosperm genus *Nothofagus* (McQuillan, 1993) have the best known herbivore faunas. The insect fauna associated with *Araucaria* is especially rich in beetles, of which five groups share comparable distributions with their Araucarian hosts in southern South America and Australia (Table 1). These present the possibility of joint study of both the assembly of a biogeographic realm and of a plant fauna (Farrell & Mitter, 1998; Sequeira *et al.*, 2000b; Farrell, 2001). Such systems should permit, in principle, distinguishing younger colonists from early inhabitants, perhaps illuminating patterns in timing and lending the historical context for evaluation of degree of adaptation and diversity (Farrell, 2001).

Araucaria has a rich fossil history extending from the Jurassic through the Early Tertiary (Fig. 1; Stockey, 1982; Stockey, Nishida & Nishida, 1994; Pole, 1995; Pole & Douglas, 1999), and *Araucaria*-containing beds also contain beetle body fossils in chrysomelid, nemomychid and belid groups today associated with these plants and which therefore represent some of the original Mesozoic herbivore associations (Arnol'di *et al.*, 1992; Kuschel & Poinar, 1993; Kuschel & May, 1990; Farrell, 1998). However, *Araucaria*-associated bark beetles in the curculionid subfamily Scolytinae are not known earlier than the Tertiary, and their phylogenetic position within an angiosperm-feeding group strongly implies a post-Jurassic origin (Kuschel, 1995; Marvaldi, 1997; Farrell, 1998; Kuschel, Leschen & Zimmerman, 2000; Marvaldi, Sequeira & Farrell, in prep.).

Our previous studies suggested a possibly Late Cretaceous age for *Araucaria* associations in bark beetles, because currently associated scolytine genera (*Hylurgonotus*, *Xylechinosomus* and *Sinophloeus*) in the tribe Tomicini in South America comprise the basal lineages of scolytid beetles (Sequeira *et al.*, 2000b). We have also recently confirmed the origin of the entire group within the primitively angiosperm-feeding weevil family Curculionidae (Marvaldi *et al.*, in prep.). Nevertheless, an early shift from angiosperms to *Araucaria* seemed plausible in light of *Araucaria* fossil history.

Although the 19 extant species of *Araucaria* today show a disjunct distribution in the Southern Hemisphere, each endemic either to Australia, New Caledonia, New Guinea or South America, *Araucaria* was much more widely distributed in the past, and was available for beetle colonization on all continents extending back at least into the Jurassic (Fig. 1). Thus the present distribution of *Araucaria*, though Gondwanan in character, is often said to be relictual, resulting from a different process of 'vicariance' via extinction in the northern part of their range over similar Cretaceous intervals as the tectonic separation of South America and Australia. Nevertheless, the absence of confirmed bark beetle fossils earlier than the Tertiary has been taken as evidence of Cretaceous absence, and their phylogenetic position rules out pre-angiosperm, Jurassic origins. Fortunately, potential clarification of the timing of origin of the bark beetles is presented by the existence of *Araucaria*-associated tomicine bark beetles in the Australian region. Two bark beetle genera, *Hylurdrectonus* and *Pachycotes*, are restricted to Australia, New Zealand and Papua New Guinea where they are associated with *Araucaria bidwillii*, *A. cunninghamii* and *A. heterophylla*. If these proved to be the closest relatives of the South American affiliates of *A. araucana* (Table 1) and their molecular divergence is commensurate with divergence in the

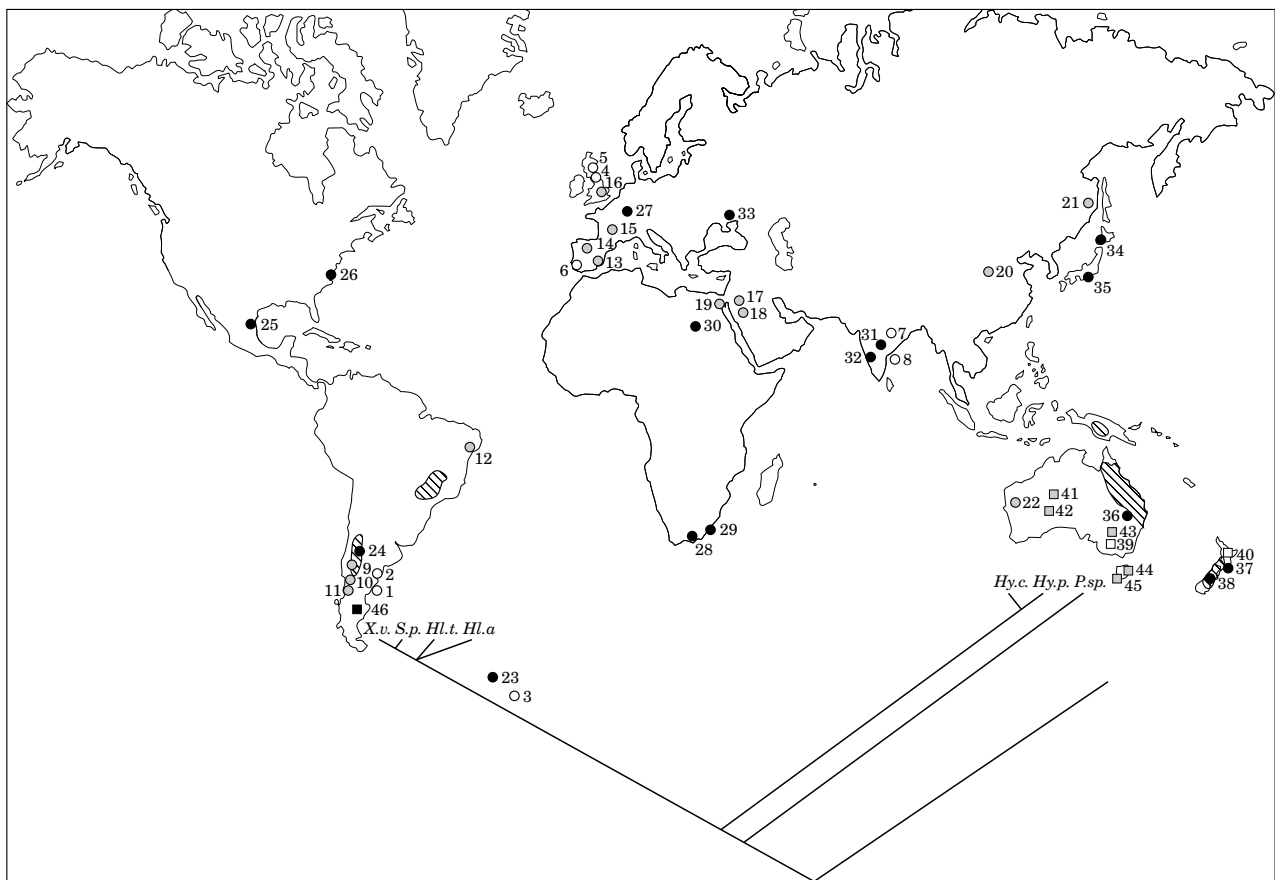


Figure 1. Cladogram depicting the relationships of the *Araucaria* feeding bark beetle lineages located according to the landmass distribution of each taxon (*X. v.*: *Xylechinosomus valdivianus*; *S. p.*: *Sinophloeus porteri*; *Hl. a.*: *Hylurionotus antipodus*; *Hl. t.*: *Hylurionotus tuberculatus*; *Hy. c.*: *Hylurdretonus corticinus*; *Hy. p.*: *Hylurdretonus pinarius*; *P. sp.*: *Pachycotes sp.*). Distributions of Araucariaceae fossils. Mesozoic fossils are circles and Cenozoic fossils are squares. Jurassic: white; Lower Cretaceous: gray; Upper Cretaceous: black; Palaeocene: white; Eocene: gray; Oligocene: black. Numbers beside each fossil location correspond to the publication of either the description of the fossil or the placement in a phylogenetic framework. 1: (Ibañez & Zamauer, 1996); 2: (Cuneo, 1991); 3: (Gee, 1989); 4: (Kendall, 1949); 5: (Stockey, 1980); 6: (Boureau, 1949); 7: (Bose & Jain, 1964); 8: (Sukh & Zeba, 1976); 9: (Archangelsky, 1994); 10: (del Fuego, 1991); 11: (Archangelsky, 1963); 12: (Duarte, 1993); 13: (Barale, 1992); 14: (Alvarez Ramis & Fernandez Marron, 1992); 15: (Wilde & Goth, 1987); 16: (Alvin, Watson & Spicer, 1994); 17: (Nissenbaum & Horowitz, 1992); 18: (Bandel & Vavra, 1981); 19: (Rottlaender & Mischer, 1970); 20: (Brattseva & Novodvorskaya, 1979); 21: (Krasilov, 1965); 22: (Cantrill, 1992); 23: (Zastawniak, 1994); 24: (Torres & Biro Bagoczky, 1986); 25: (Cevallos Ferriz, 1992); 26: (Raubeson & Gensel, 1991); 27: (Meijer, 1997); 28: (Erasmus, 1976); 29: (Schultze, 1966); 30: (Veillet Bartoszewska, 1956); 31: (Kar, Ambwani & Agarwal, 1998); 32: (Bose & Maheshwari, 1973); 33: (Voronova & Smykov, 1972); 34 and 35: (Stockey *et al.*, 1994); 36: (Pole, 2000); 37: (Pole, 1995); 38: (Pole & Douglas, 1999); 39: (Rowett, 1992); 40: (Pole, 1998); 41: (Carpenter & Pole, 1995); 42: (Martin, 1993); 43: (Christophel *et al.*, 1992); 44: (Hill & Bigwood, 1987); 45: (Pole, 1992); 46: (Barreda, 1997).

Cretaceous or earlier, this would disfavour the hypothesis that these are of Tertiary, post-Gondwanan origins.

Here we present results of the analysis of DNA sequences of nuclear and mitochondrial genes: protein coding elongation factor alpha (EF-1 α), enolase (eno) and cytochrome oxidase I (CO1) to both resolve the phylogenetic affinities and estimate divergence times

of the Australian and South American bark beetle genera affiliated with *Araucaria*.

MATERIAL AND METHODS

SAMPLES

Bark beetles from 9 of the 14 genera in the Tomicini and two of the three genera in the Hylastini were

collected from colonized hosts (see Table 2 for habits and distributions). Members of two different tribes from the weevil subfamily Cossoninae (Rhyncolini and Araucarini), the putative sister-group of Scolytinae (Kuschel *et al.*, 2000; Marvaldi *et al.*, in prep.), are included as outgroups.

DNA PREPARATION, PCR AMPLIFICATION AND SEQUENCING

DNA was extracted from individual beetles preserved in ethanol, DNA for six of the *Dendroctonus* species was kindly provided by Dr Scott Kelley. PCR and cycle sequencing were used to obtain partial sequences of three genes: EF-1 α , CO1 and enolase. Primers and conditions for amplification and sequencing are detailed in Normark, Jordal & Farrell (1999) for CO1, Sequeira *et al.* (2000b) for EF-1 α and Farrell *et al.* (2001) for enolase. Double stranded PCR products were purified using the Qiagen PCR purification kit to remove primers and unincorporated dNTPs prior to sequencing. Cycle sequencing reactions were performed with the ABI prism Dye Terminator Cycle Sequencing Kit (Perkin-Elmer). Both strands of the PCR product were sequenced in an ABI 370A automated sequencer. All sequences were compiled using Sequencher 3.1 (Genecodes Corporation, Ann Arbor, MI). For EF-1 α and enolase, evidence of two loci that differ in intron/exon structure was found in some taxa but only the copy with one middle intron was used for EF-1 α (Normark *et al.*, 1999) and the non-intron copy for enolase (eno1-ni) (Farrell *et al.*, 2001).

PHYLOGENETIC ANALYSIS

Saturation levels for each of the gene regions due to multiple substitutions were identified by plotting ts/tv ratios versus the number of transversions and by comparing with the ts/tv expected values based on the base composition as described in Holmquist (1983) (Fig. 2A,B,C). Each dataset was analysed separately and then combined in a total evidence matrix (1968 characters: for relative contribution of each gene region to the total of informative characters see Table 3, for accession numbers see Table 2). All substitutions were weighted equally. Phylogenetic analysis was performed by maximum parsimony using PAUP* (Swofford, 2000). The parsimony ratchet procedure (Nixon, 1999) was performed five times using 200 replicates each and repeated with varying percentages of weighted characters (12, 15 and 18). Batch files to implement this procedure were constructed using Pauprat (Sikes & Lewis, 2000) where an assigned percentage of characters selected at random in each replicate are given additional weight and branch swapping is performed on that tree using the reweighted matrix. This batch file is then executed in Paup*. The ratchet samples

many tree islands with fewer trees from each island providing faster accurate estimates of a consensus (Nixon, 1999). For bootstrapping and incongruence testing (Farris *et al.*, 1995) 100 replications and 20 random-addition starting trees were used. Autodecay 4.0 (Eriksson, 1998) was used to create the constraint trees for the nodes from the combined MP tree and TreeRoot (Sorenson, 1999) was used to calculate the partitioned Bremer support indices for each of the three gene regions (Baker & DeSalle, 1997; Baker, Yu & DeSalle, 1998). Decay indexes (Bremer, 1994) were calculated from the runs performed in Paup using heuristic searches with 100 random additions.

BAYESIAN INFERENCE OF DIVERGENCE TIMES

An approximation of the likelihood ratio test (LRT) (Felsenstein, 1981; Huelsenbeck & Rannala, 1997) was performed using maximum likelihood branch length optimizations of the three datasets independently, enforcing and not enforcing a molecular clock (thus testing for deviation from the molecular clock), over the strict consensus of the six MP trees obtained for the combined dataset using the 'describe tree' feature in Paup (HKY + Γ , estimating Γ , using the empirical base frequencies, estimating Ts/tv ratio).

Divergence times were estimated with Bayesian analysis and the nucleotide data with the clock tree (with optimized branch lengths) as a prior and running multiple Markov chain for 100 000 generations, with sampling every 100 generations using Mr Bayes 1.1 (Huelsenbeck, 2000). The first 10 000 generations of each chain were discarded (as recommended by the author). By calculating the posterior probabilities and using the provided dataset and tree with branch-lengths, Mr. Bayes will infer the branching time of the root of the tree and any additional parts of the tree that are provided as constraints. The median, mean and standard deviation of the age estimates resulting from the four 100 000 Markov chains are then used as estimates of the divergence times for the nodes of interest. The calibration specified for the origin of Scolytinae was as follows: a maximum of 135 million years ago (Mya), around the time of the origin of angiosperms, because Scolytinae appears to be nested deep within a primitively angiosperm-feeding clade of weevils (Kuschel, 1995; Marvaldi, 1997; Farrell, 1998; Kuschel *et al.*, 2000; Marvaldi *et al.*, in prep.), and a minimum of 40 Mya, the age of the oldest body fossil reported for any of the taxa in the tree: *Hylastes* and *Hylurgops*: Late Eocene amber; Larsson, 1978) (Fig. 3).

The utility of each gene partition was further investigated by plotting the partitioned Bremer support against the age estimated as above for that node. For nodes of the same age the mean and standard deviation

Table 2. Bark beetle taxa included in the study with the sequences analyzed, the feeding habit and distribution (subscripts beside accession numbers indicate studies by Kelley & Farrell 1998 (1), Sequeira *et al.*, 2000b (2) and Farrell *et al.*, 2001 (3))

	EF-1 α	COI	eno-ni	Host	Feeding habit	Distribution
Cossoninae						
<i>Araucarius major</i>	—	AY040285	AF375335 ₃	Araucariaceae	Phloeophagous	South American
<i>Araucarius minor</i>	AF308396 ₂	AF375307 ₃	AY040298	Araucariaceae	Phloeophagous	South American
<i>Stenanylus sp.</i>	AF375264 ₁	—	AF375334 ₃	polyphagous	Phloeophagous	South American
Scolytinae						
<i>Hylastes porculus</i>	AF308430 ₂	AF375321 ₃	AY040300	<i>Pinus</i> spp.	Phloeophagous	Holarctic
<i>Hylurgops rugipennis</i>	AF308408 ₂	AF375323 ₃	AY040299	<i>Picea</i> , <i>Pinus</i>	Phloeophagous	Holarctic
<i>Dendroctonus pseudotsugae</i>	AF308418 ₂	AF375318 ₃	AF375341 ₃	Pinaceae ¹	Phloeophagous	Holarctic
<i>Dendroctonus adjunctus</i>	AF308423 ₂	AY040286	AY040301	Pinaceae	Phloeophagous	Palaearctic
<i>Dendroctonus frontalis</i>	AF308424 ₂	AY040287	AY040302	Pinaceae	Phloeophagous	Palaearctic
<i>Dendroctonus jeffreyi</i>	AF308425 ₂	AY040288	AY040303	Pinaceae	Phloeophagous	Palaearctic
<i>Dendroctonus mexicanus</i>	AF308426 ₂	AF067988 ₁	AY040304	Pinaceae	Phloeophagous	Palaearctic
<i>Dendroctonus murrayanae</i>	AF308427 ₂	AF067989 ₁	AY040305	Pinaceae	Phloeophagous	Palaearctic
<i>Dendroctonus ponderosae</i>	AF308428 ₂	AF067987 ₁	AY040306	Pinaceae	Phloeophagous	Palaearctic
<i>Dendroctonus terebrans</i>	AF308429 ₂	AF375315 ₃	AF375338 ₃	Pinaceae	Phloeophagous	Palaearctic
<i>Hyludretonus corticianus</i>	AY040316	AT040289	AY040307	<i>A. cunninghamii</i>	Phloeophagous	Australian
<i>Hyludretonus pinarius</i>	AY040317	AY040290	AY040308	<i>A. cunninghamii</i>	Phloeophagous	Australian
<i>Hylurgonotus tuberculatus</i>	—	AF375313 ₃	AF375337 ₃	<i>A. araucana</i>	Xylophagous	South American
<i>Hylurgonotus antipodius</i>	AF308419 ₂	AY040291	AY040309	<i>A. araucana</i>	Phloeophagous	South American
<i>Hylurgonotus ligniperda</i>	—	AY040292	—	<i>Pinus</i>	Phloeophagous	Palaearctic
<i>Hylurgus micklitzi</i>	—	AY040293	AY040310	<i>Pinus</i>	Phloeophagous	Palaearctic
<i>Pachycotes sp.</i>	AY040318	AY040294	AY040311	<i>A. cunninghamii</i>	Xylophagous	Australian
<i>Pseudohylesinus granulatus</i>	AF308421 ₂	—	—	<i>Abies</i> , <i>Tsuga</i>	Phloeophagous	Nearctic
<i>Pseudohylesinus nebulosus</i>	AF308422 ₂	AF375316 ₃	AF375339 ₃	<i>Pseudotsuga</i>	Phloeophagous	Nearctic
<i>Sinophloeus porteri</i>	AF308420 ₂	AF375314 ₃	AY040312	<i>A. araucana</i>	Phloeophagous	South American
<i>Tomias destruens</i>	AY040313	AY040295	—	<i>Pinus</i> spp.	Phloeophagous	Palaearctic
<i>Tomias piniperda</i>	AY040314	AY040296	—	<i>Pinus</i> spp.	Phloeophagous	Palaearctic
<i>Tomias minor</i>	AY040315	AY040297	—	<i>Pinus</i> spp.	Phloeophagous	Palaearctic
<i>Xylechinomus valdivianus</i>	AF308410 ₂	AF375312 ₃	AF375336 ₃	<i>A. araucana</i>	Phloeophagous	South American

¹ For detailed host associations for *Dendroctonus* see Kelley & Farrell (1998) and references therein.

of the partitioned Bremer support values for those nodes were calculated (Fig. 2D).

RESULTS

Independent analysis of each dataset (EF-1 α , enolase-ni, CO1 amino acids and nucleotides), resolve relationships at different levels. CO1 nucleotides (almost entirely third codon positions) provide support close to the tips of the tree but not at higher levels (Fig 3). As in our earlier studies of bark beetles (Normark *et al.*, 1999; Farrell *et al.*, 2001), the higher ts/tv ratio in third codon positions (ts/tv ratio total = 1.16; 1st = 1.02; 2nds = 0.92; 3rds = 1.14) plus the asymptotic divergence (suggesting saturation) in third positions indicates the use of CO1 amino acids for the analysis. As in an earlier study in scolytine beetles (Jordal, Normark & Farrell, 2000), the level of saturation due to multiple substitutions was higher for mitochondrial genes, in this case CO1, than for the two other gene regions and mostly comprises changes in the third codon positions (Fig. 2C). However, the amino acid sequences for this rapidly evolving mitochondrial gene prove to be useful to resolve the deeper nodes of this phylogeny (Figs 2D, 3), exhibiting a higher proportion of informative characters (Table 3) and higher mean divergence values than the amino acid sequences of the other two nuclear regions (4% for EF-1 α , 12% for CO1 and 5% for enolase-ni).

Partition homogeneity tests indicate no significant incongruence among the three datasets ($P=0.0698$). Regardless of the percentage of characters weighted, all ratchet runs performed using the combined dataset resulted in 6 MP trees of 2469 steps, the strict consensus of which is shown in Figure 3. The phylogeny estimate from the combined dataset indicates ancestral association with *Araucaria* and Tomicini paraphyly with respect to Hylastini, though not highly supported, but concordant with previous studies (Sequeira *et al.*, 2000b). The five *Araucaria*-feeding genera (*Hylurgonotus*, *Xylechinosomus*, *Sinophloeus*, *Hylurdretonus* and *Pachycotes*) predominate in the basal branches of the tree (Fig. 3). The *Araucaria*-feeding genera from the Australian region are basal and the South American taxa form a well-supported monophyletic group.

The LRT performed as a maximum likelihood branch length optimization of the CO1 nucleotide data on the topology of the strict consensus of the 6 MP trees from the combined data indicates nonsignificant deviation from the molecular clock ($X=30$, $df=20$, $P=0.0698$), the other two gene regions rejected the molecular clock hypothesis ($P<0.005$). CO1 nucleotide data, though not resolving relationships at deep nodes (probably due to saturation at third codon positions: Fig. 2C), is the most suitable for dating this divergence due to its

clocklike variation rates and shows enough variation in first and second codon positions (reflected in the aminoacid sequences) that allow resolution of these deep nodes. The Bayesian estimation of divergence times is shown on a time scale on the phylogram in Figure 4. For several nodes the median (M), mean and standard deviation for the dates were calculated in the four chains of 100 000 generations, sampling every 100 generations (4000 estimations). These estimates indicate that the divergence between the Australian and the South American *Araucaria*-feeding taxa occurred at the very latest near the Cretaceous/Paleocene border (78 Mya [range 65–91 Mya] in Fig. 4). The age of the first Scolytinae-*Araucaria* association would then be during the latest stages of the Late Cretaceous (80 Mya [range 67–93 Mya] in Fig. 4).

As regards the relative utility of the three gene partitions (Fig. 2D), both nuclear genes, EF-1 α and enolase-ni, provide greater support for Oligocene–Miocene divergences and lose informativeness approaching the Paleocene, whereas CO1 amino acids contribute resolution closer to the Cretaceous/Paleocene border and even well within the late Cretaceous.

DISCUSSION

DIVERGENCE TIMES AND FOSSIL EVIDENCE

The divergence times and phylogenetic evidence that we present here supports the hypothesis advanced previously that the lineages of Tomicini that today breed in *Araucaria* are surviving remnants of a reversion to conifer-feeding in ancestral scolytids that appears to have occurred late in the Mesozoic, when *Araucaria* still formed a major component of the woody flora (Sequeira *et al.*, 2000b; Farrell *et al.*, 2001).

Fossil galleries in early Cretaceous conifer bark have been interpreted as evidence of Scolytinae (Brongniart, 1877; Wood cited in Boucot, 1990; Falder *et al.*, 1998), as well as reports of scolytines from the early/mid Cretaceous *Araucaria*-derived Lebanese amber (Whalley cited in Jarzembowsky, 1990). Our results lend support to an association with conifers that has probably persisted since the common origin of these tribes, some 78 Myr (± 13 Myr), somewhat later in the Cretaceous, closer to the Cretaceous/Paleocene border. This late Cretaceous age would be also consistent with the plant fossil record, because by the late Cretaceous the family Pinaceae was well established and the genus *Pinus*, host to the remaining Tomicini plus Hylastini, was already highly diversified and widespread (Miller, 1977, 1988; Millar, 1993; Savard *et al.*, 1994).

BIOGEOGRAPHIC SCENARIOS

The basal scolytine associations with *Araucaria* could result from contraction from a formerly more widespread (almost worldwide) distribution with their hosts

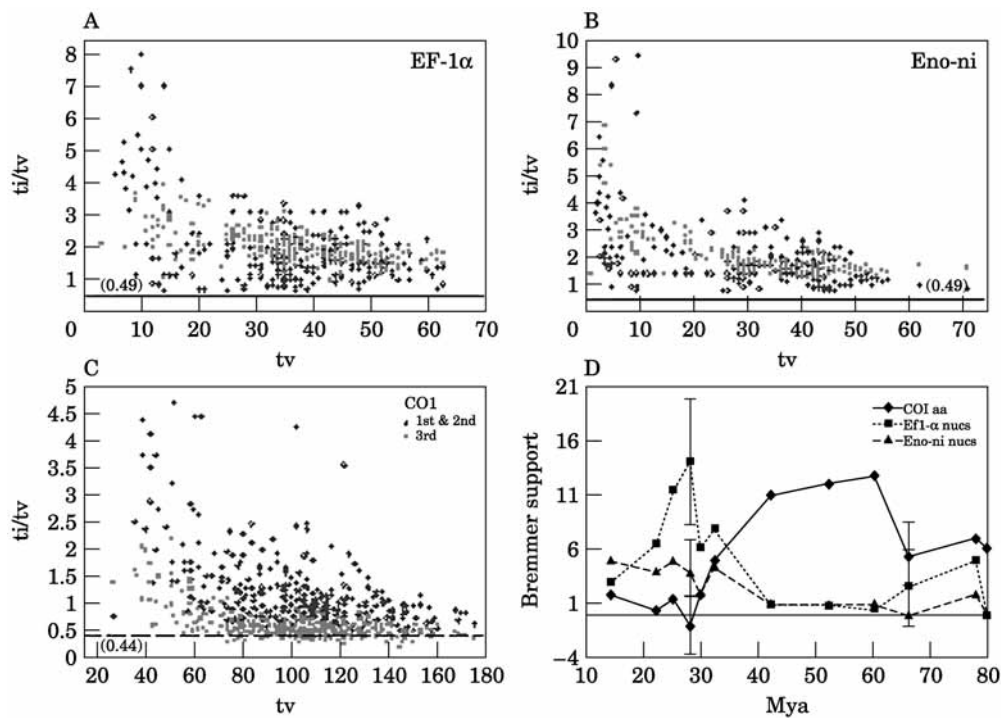


Figure 2. A–C, plots of transition/transversion ratios versus transversions for first and second codon positions combined and third positions in each gene region. Values of expected Ts/tv ratio ratio (Holmquist, 1983) are shown on the lines in each graph. D, partitioned Bremer support for each of the three data partitions against the divergence time estimated for that node with Bayesian analysis. The mean and SD are shown for nodes of the same age.

Table 3. Properties of gene partitions; Ti and tv signify transitions and transversions respectively

Gene region	Characters	% informative characters	Ti/tv ratios
EF-1α all pos	921	27	1.7
1	307	11	1.5
2	307	3	0.75
3	307	68	1.8
aa	307	7	—
COI all pos	1386	42	1.3
1	462	26	1.0
2	462	7	0.92
3	462	82	1.14
aa	462	23	—
Eno-ni all pos	585	11	1.5
1	195	11	1
2	195	4	0.8
3	195	70	1.1
aa	195	9	—
Combined	1968	EF1-α, 47 COI aa, 21 Eno-ni, 32	

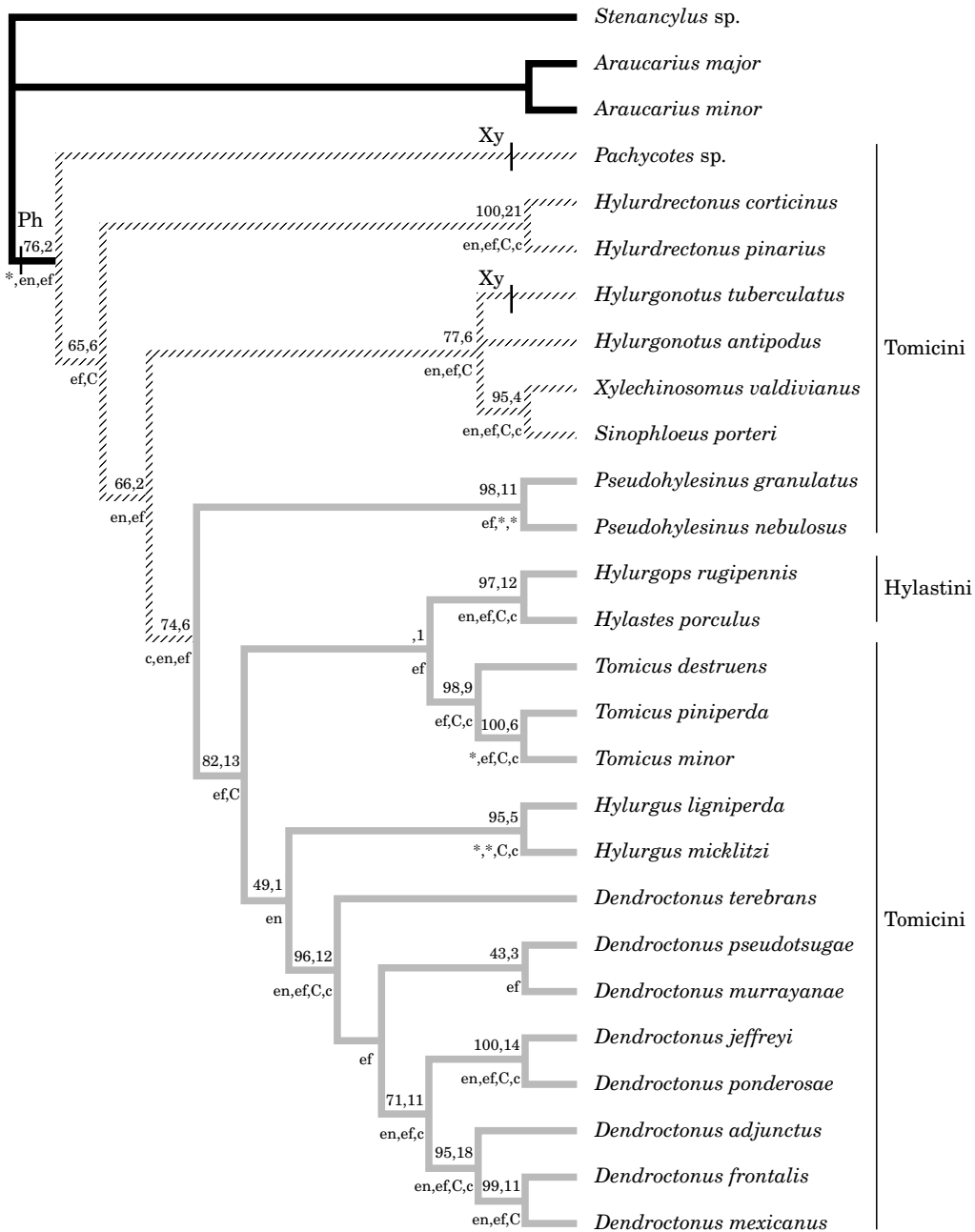


Figure 3. Strict consensus of six MP trees from the combined analysis of CO1 amino acids, EF-1 α and enolase-ni nucleotides (1968 characters: 462 CO1; 921 EF-1 α ; 585 enolase). Length = 2469; Ci = 0.587; Ri = 0.591. Numbers above the internal branches indicate bootstrap support and Bremer support for the node to the right in the combined analysis. Letters below the branches indicate which of the independent analysis supports that node with: more than 50% bootstrap support (letters in bold) or less than 50% bootstrap support (C: COI amino acids; c: COI nucleotides; ef, EF-1 α ; en: enolase non-intron). Patterns on the branches correspond to conifer family/beetle associations; stippled: Araucariaceae; solid grey: Pinaceae. Codes with bars on the branches mark changes in feeding modes (Ph: phloem-feeding, Xy: Feeding in xylem). Bars beside taxon names indicate tribe classification after Wood (1986; Wood & Bright, 1992) and Bright & Skidmore (1997). * marks the node where the calibration was specified.

(Fig. 1). A northern origin of the beetle *Araucaria* association plus worldwide dispersal before 80 Mya would seem to place the origin of the bark beetles

somewhat earlier in the Cretaceous, closer to the onset of angiosperm diversification, the main hosts of the apparent weevil ancestors of scolytines (Crowson,

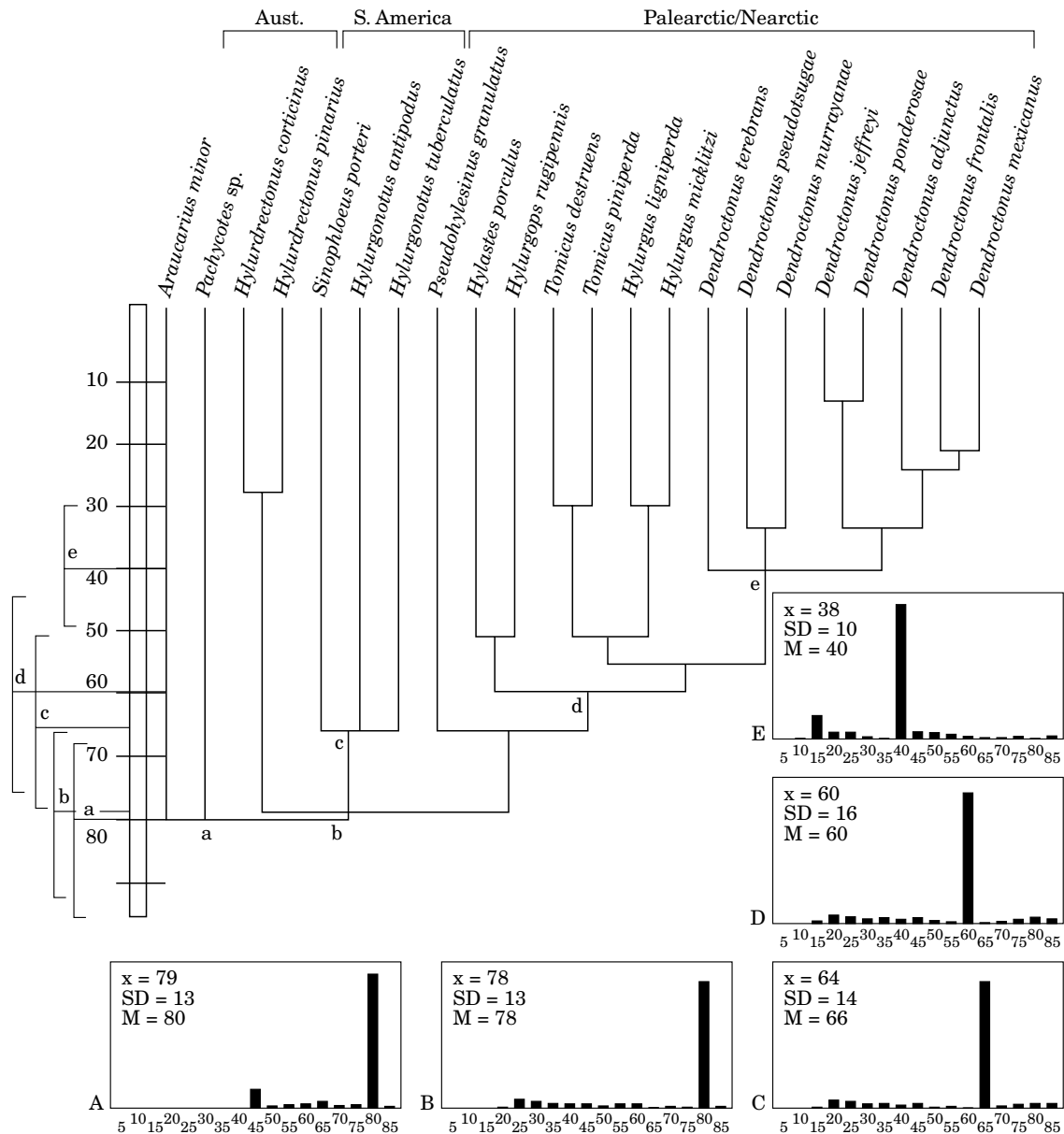


Figure 4. Phylogram showing the branch length optimization of the CO1 nucleotide data enforcing a molecular clock (HKY + Γ , estimating Γ , using the empirical base frequencies, estimating t_s/t_v ratio) over the strict consensus of the six MP trees obtained for the combined dataset. Histograms correspond to the distribution of age estimations for the constrained nodes obtained via the Markov chain in Mr. Bayes 1.0, with the SD of the mean for each estimation. Ranges on the time axis correspond to \pm SD for each estimation and the median of the distribution.

1967; Thompson, 1992; Kuschel, 1966, 1995; May, 1993; Lyal, 1995; Lyal & King, 1996; Marvaldi, 1997; Farrell, 1998; Farrell *et al.*, 2001; Marvaldi *et al.*, in prep.). However, a southern origin of the beetles and dispersal through Antarctica with later contraction to the present distribution could be on the more recent side of our estimated range. The surviving *Araucaria* associated bark beetle lineages, though now very isolated, could have originated either in Antarctica (where

Upper Cretaceous Araucariaceae fossils have been reported, Zastawniak (1994)) or in the Australian region and have dispersed to both Southern continents (South America and Australia) when these land masses were still close to each other (70 Mya: Pollock (1995); Rosen (1978) and Thayer (1985)).

The most compelling evidence on the question of origins is the restriction of Tomicini to the basal *Araucaria* sections *Bunya*, *Intermedia* and *Araucaria*,

plus the basal species in the most derived section Eutacta, which is thought to have radiated post-Eocene (Setoguchi *et al.*, 1998). The sections Bunya, Intermedia and Araucaria, in contrast, together form a monophyletic group whose fossil distribution is exclusively on New Zealand, Australia and Argentina in 70–80 Myr-old deposits (Fig. 1; Ohsawa, Nishida & Nishida, 1995; Stockey, Nishida & Nishida, 1992) and therefore favouring the model of southern vicariance during the mid to late Cretaceous for the distribution of Tomicini/Araucaria associations. More northern Araucaria fossils are not currently attributed to any of the sections used by Tomicini. If these fossils eventually prove to fall in derived sections of Araucaria, while sections Bunya, Intermedia and Araucaria remain basal, the origins of Araucaria would be in the Southern Hemisphere and their current distribution would be the result of Gondwanan vicariance, as with their beetle herbivores.

Further study of the bark beetle sister group, the Cossoninae, may permit resolution of the homology (and thus greater age) of the Araucaria associations of Araucariini with those of basal Scolytinae, possibly clarifying their geographic origins as well. Indeed, the distributions and associations of the remaining genera are similarly Gondwanan. The Australian genera *Coprocorynus*, *Mastersinella* and *Xenocnema* are all associated with Araucariaceae in Australia, while New Zealand harbours *Xenocnema* (also on New Caledonia) plus *Inosomus*, the sole genus associated with Podocarpaceae, the sister group to Araucariaceae (Setoguchi *et al.*, 1998). Moreover, the remaining genus previously placed in the Araucariini, *Amorphocereus* (new placement in the Molytinae, Alonso-Zarazaga & Lyal, 1999), is associated with Cycadaceae in South Africa, where Araucaria is now extinct. If these associations of Tomicini and Araucariini with Araucariaceae reflect descent from a shared ancestor, the origin of the association would be even deeper in the Cretaceous.

Also suggestive of age are the otherwise very rare shifts from phloem to xylem feeding in these Araucaria beetles, as well as a unique origin of leaf mining (Farrell *et al.*, 2001) (Table 2). Similarly, Kuschel (1966) suggested that the morphological disparity among the cossonine genera in the Araucariini indicates ancient divergences.

Even an early Cretaceous origin of association of Scolytinae, Araucariini or both with Araucariaceae could be reconciled with their placement in the primitively angiosperm-feeding weevils, but strongly pre-dates the previously assigned origin of the group in the Tertiary. While it is clear that these beetle tribes are thus Late Cretaceous in age, there remain a number of insect groups – their ranks ranging from families to genera – that have comparable distributions, whose

age remains to be investigated (Table 1). Because both the phylogeny and fossil record place other Araucaria herbivores well within the Jurassic, it seems that the evolutionary origins of the Gondwanan Araucaria fauna spans much of the Mesozoic. Demonstration that these plants were also colonized in the Tertiary by as yet unstudied herbivores would further expand the set of comparisons of older and younger herbivores, permitting assay of the relative closeness of their adaptation to these hosts.

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REFERENCES

- Alonso-Zarazaga M, Lyal CHC. 1999. A world catalogue of families and genera of Curculionoidea (Insecta: Coleoptera) (Excepting Scolytidae and Platypodidae). Barcelona, Spain, Entomopraxis.
- Alvarez Ramis C, Fernandez Marron T. 1992. An attempt to determine the genus of fossil remains considered as Araucariaceae. *Organisation Francaise de Paleobotanique Informations* 16-B: 4.
- Alvin KL, Watson J, Spicer RA. 1994. A new coniferous male cone from the English Wealden and a discussion of pollination in the Cheirolepidiaceae. *Palaeontology* 37: 173–180.
- Archangelsky S. 1963. A new Mesozoic flora from Tico, Santa Cruz Province, Argentina. *Bulletin of the British Museum Natural History, Geology Series* 8: 45–92.
- Archangelsky S. 1994. Comparative ultrastructure of three Early Cretaceous gymnosperm pollen grains: *Araucariacites*, *Balmeiopsis* and *Callialasporites*. *Review of Palaeobotany and Palynology* 83: 185–198.

- Arnol'di LV, Zherikhin VV, Nikritin LM, Ponomarenko AG. 1992.** *Mesozoic Coleptera*. New Delhi: Amerind Publishing Co.
- Baehr M. 1995.** Revision of *Philipis* (Coleoptera: Carabidae: Bembidiinae). A genus of arboreal tachyine beetles from rainforests of eastern Australia: Taxonomy, phylogeny and biogeography. *Memoirs of the Queensland Museum* **38**: 315–381.
- Baker RH, DeSalle R. 1997.** Multiple sources of character information and the phylogeny of Hawaiian drosophilids. *Systematic Biology* **46**: 645–673.
- Baker RH, Yu X, DeSalle R. 1998.** Assessing the relative contribution of molecular and morphological characters in simultaneous analysis trees. *Molecular Phylogenetics and Evolution* **9**: 427–436.
- Bandel K, Vavra N. 1981.** A Lower Cretaceous fossil resin from Jordan. *Neues Jahrbuch für Geologie und Paläontologie Monatshefte* **1**: 19–33.
- Barale G. 1992.** New remains of Araucariaceae in the lithographic limestones from the Lower Cretaceous of the Montsec, Spain. *Organisation Francaise de Paleobotanique Informations* **16-B**: 12.
- Barreda VD. 1997.** Palinoestratigrafía de la Formación San Julian en el area de Playa la Mina (provincia de Santa Cruz), Oligoceno de la cuenca Austral. *Ameghiniana* **34**: 283–294.
- Bose MN, Jain KP. 1964.** A megastrobilus belonging to the Araucariaceae from the Rajmahal hills, Bihar, India. *The Palaeobotanist* **12**: 229–230.
- Bose MN, Maheshwari HK. 1973.** Some detached seed-scales belonging to Araucariaceae from the Mesozoic rocks of India. *Geophytology* **3**: 205–214.
- Boucot AJ. 1990.** *Evolutionary Paleobiology of Behaviour and Coevolution*. New York, NY: Elsevier.
- Boureau E. 1949.** *Dadoxylon* (Araucarioxylon) *teixeirae* n. sp., bois fossile du jurassique superieur portugais. *Comunicacoes dos Servicos Geologicos de Portugal* **29**: 187–194.
- Brattseva GM, Novodvorskaya IM. 1979.** Palynological characteristics of the Lower Cretaceous deposits of the find-localities of Buylyasutuin-Khuduk and Anda-Khuduk, Bayan-Hongor Province, Mongolia. *Paleontologicheskaya Ekspeditsiya* **8**: 98–99.
- Bremer K. 1994.** Branch support and tree stability. *Cladistics* **10**: 295–304.
- Bright DE, Skidmore RE. 1997.** *A catalog of Scolytidae and Platypodidae (Coleoptera) Supplement 1 (1990–1994)*. Ontario: NRC Research Press.
- Britton EB. 1970.** Coleoptera (Beetles). In: *CSIRO Insects of Australia*. Melbourne: Melbourne University Press.
- Brongniart C. 1877.** Perforations observées dans deux morceaux de bois fossile. *Annales Société Entomologique France* **7**: 215–220.
- Brower AVZ. 1994.** Rapid morphological radiation and convergence among races of the butterfly *Heliconius erato* inferred from patterns of mitochondrial DNA evolution. *Proceedings of the National Academy of Sciences, USA* **91**: 6491–6495.
- Cantrill DJ. 1992.** Araucarian foliage from the lower Cretaceous of southern Victoria, Australia. *International Journal of Plant Sciences* **153**: 622–645.
- Carpenter RJ, Pole M. 1995.** Eocene plant fossils from the Lefroy and Cowan paleodrainages, Western Australia. *Australian Systematic Botany* **8**: 1107–1154.
- Cevallos Ferriz SRS. 1992.** Three Cretaceous gymnospermous woods from northern Mexico. *Anales del Instituto de Biología Universidad Nacional Autónoma de México Serie Botánica* **63**: 111–137.
- Christophel DC, Scriven LJ, Greenwood DR, Rowett AI. 1992.** Dispersed cuticular floras of South Australian Tertiary coalfields; Part 2, Lochiel. *Transactions Royal Society of South Australia* **116**: 65–76.
- Coleman NC, Monteith GB. 1981.** Life history of the North Queensland day-flying moth, *Alcides zodiaca* Butler (Lepidoptera: Uraniidae). *The North Queensland Naturalist* **45**: 2–6.
- Craw RC. 1985.** Classic problems of Southern Hemisphere biogeography re-examined. *Zeitschrift für Zoologische Systematik und Evolutionsforschung* **23**: 1–10.
- Crowson RA. 1967.** *The natural classification of the families of Coleoptera*. Middlesex, UK: E. W. Classey.
- Cuneo R. 1991.** Structural and paleoenvironmental analysis of the Jurassic *Araucaria* petrified forest from Patagonia Argentina. *American Journal of Botany* **78**: 111.
- Darlington PJ. 1957.** *Zoogeography: the geographical distribution of animals*. New York: Wiley.
- Darlington PJ. 1965.** *Biogeography of the southern end of the world; distribution and history of far-southern life and land, with an assessment of continental drift*. Cambridge, Mass.: Harvard University Press.
- Davis DR. 1986.** A new family of monotrystian moths from austral South America (Lepidoptera: Palaephatidae) with a phylogenetic review of the Monotrystia. *Smithsonian Contributions to Zoology* **434**: 1–202.
- del Fuego GM. 1991.** A fertile Araucariaceae from the Cretaceous of Santa Cruz; Argentina. *Ameghiniana* **28**: 203.
- Duarte L. 1993.** Araucariaceae remains in the Santana Formation, Crato Member (Aptian); northeastern Brazil. *Anais da Academia Brasileira de Ciências* **65**: 357–362.
- Erasmus T. 1976.** A new species of *Dammroxylon* Schultze-Motel, D. Natalense sp. nov. from the Cretaceous of Natal, South Africa. *Palaeontologia Africana* **19**: 135–139.
- Eriksson T. 1998.** *AutoDecay ver. 4.0*. Department of Botany, Stockholm University, Stockholm (program distributed by the author).
- Falder AB, Rothwell GW, Mapes G, Mapes RH, Doguzhaeva LA. 1998.** *Pityostrobus milleri* sp. nov., a pinnaceous cone from the Lower Cretaceous (Aptian) of southwestern Russia. *Review of Palaeobotany and Palynology* **103**: 253–261.
- Farrell BD. 1998.** Inordinate fondness explained: Why are there so many beetles? *Science (Washington)* **281**: 555–559.
- Farrell BD. 2001.** Evolutionary assembly of the milkweed fauna: Cytochrome oxidase I and the age of *Tetraopes* beetles. *Molecular Phylogenetics and Evolution* **18**: 467–478.

- Farrell BD, Mitter C. 1998. The timing of insect/plant diversification: Might Tetraopes (Coleoptera: Cerambycidae) and Asclepias (Asclepiadaceae) have co-evolved? *Biological Journal of the Linnean Society* **63**: 553–577.
- Farrell BD, Sequeira AS, O'Meara B, Normark BB, Chung J, Jordal BH. 2001. The evolution of agriculture in beetles (Curculionidae: Scolytinae and Platypodinae). *Evolution* (in press).
- Farris JS, Källersjö M, Kluge AC, Bult C. 1995. Testing significance of incongruence. *Cladistics* **10**: 315–319.
- Felsenstein J. 1981. Evolutionary trees from DNA sequences: A maximum likelihood approach. *Journal of Molecular Evolution* **17**: 368–376.
- Gee CT. 1989. Revision of the Late Jurassic–Early Cretaceous flora from Hope Bay Antarctica. *Palaeontographica Abteilung B Palaeophytologie* **213**: 149–214.
- Gillespie RG, Croom HB, Palumbi SR. 1994. Multiple origins of a spider radiation in Hawaii. *Proceedings of the National Academy of Sciences, USA* **91**: 2290–2294.
- Harrison MK, Crespi BJ. 1999. Phylogenetics of *Cancer* crabs (Crustacea: Decapoda: Brachyura). *Molecular Phylogenetics and Evolution* **12**: 186–199.
- Heads MJ. 1990. Integrating earth and life sciences in New Zealand's natural history: the parallel arcs model. *New Zealand Journal of Zoology* **16**: 549–585.
- Hill RS, Bigwood AJ. 1987. Tertiary gymnosperms from Tasmania; Araucariaceae. *Alcheringa* **11**: 325–335.
- Holmquist R. 1983. Transitions and transversions in evolutionary descent: an approach to understanding. *Journal of Molecular Evolution* **19**: 134–144.
- Howden HF. 1981. Zoogeography of some Australian Coleoptera as exemplified by the Scarabeoidea. In: A. Keast, ed. *Ecological biogeography of Australia*. The Hague: W. Junk.
- Huelsenbeck JP, Rannala B. 1997. Phylogenetic methods come of age: Testing hypotheses in an evolutionary context. *Science (Washington)* **276**: 227–232.
- Huelsenbeck JP. 2000. MrBayes: Bayesian inference of phylogeny. Distributed by the author. Department of Biology, University of Rochester.
- Humphries CJ, Cox JM, Nielsen ES. 1986. *Nothofagus* and its parasites: a cladistic approach to coevolution. In: Stone AR, Hawksworth DL, eds. *Coevolution and systematics*. Oxford: Clarendon Press, 55–74.
- Ibanez CG, Zamauer AB. 1996. Hyphomycetes (Deuteromycetes) in cones of *Araucaria mirabilis* (Spegazzini) Windhausen, Middle Jurassic of Patagonia, Argentina. *Mycotaxon* **59**: 137–143.
- Jarzewowski EA. 1990. A boring beetle from the Wealden of the Weald. In: Boucot AJ, ed. *Evolutionary Paleobiology of Behaviour and Coevolution*. New York: Elsevier, 373–376.
- Jordal BH, Normark BB, Farrell BD. 2000. Evolutionary radiation of an inbreeding haplodiploid beetle lineage (Curculionidae, Scolytinae). *Biological Journal of the Linnean Society* **71**: 483–499.
- Juan C, Oromi P, Hewitt GM. 1995. Mitochondrial DNA phylogeny and sequential colonization of Canary Islands by darkling beetles of the genus *Pimelia* (Tenebrionidae). *Proceedings of the Royal Society of London Series B* **261**: 173–180.
- Juan C, Oromi P, Hewitt GM. 1996. Phylogeny of the genus *Hegeter* (Tenebrionidae, Coleoptera) and its colonization of the Canary Islands deduced from Cytochrome Oxidase I mitochondrial DNA sequences. *Heredity* **76**: 392–403.
- Juan C, Ibrahim K, Oromi P, Hewitt GM. 1998. The phylogeography of the darkling beetle *Hegeter politus*, in the eastern Canary Islands. *Proceedings of the Royal Society of London Series B* **265**: 135–140.
- Kar RK, Ambwani K, Agarwal A. 1998. First occurrence of araucaroid wood from Pondicherry Formation (Late Cretaceous), Pondicherry, South India. *Current Science (Bangalore)* **75**: 1119–1121.
- Kelley S, Farrell BD. 1998. Is specialization a dead end? Phylogeny of host use in *Dendroctonus* (Scolytidae: Coleoptera). *Evolution* **52**: 1731–1743.
- Kendall MW. 1949. Jurassic member of the Araucariaceae. *Annals of Botany* **13**: 151–161.
- Krasilov VA. 1965. Araukariyevye iz nizhnego mela Primor'ya. *Paleontologicheskii Zhurnal* **2**: 109–117.
- Kuschel G. 1966. A cossonine genus with bark-beetle habits, with remarks on relationships and biogeography (Coleoptera: Curculionidae). *New Zealand Journal of Science* **9**: 3–29.
- Kuschel G. 1992. Reappraisal of the Baltic Amber Curculionoidea described by E. Voss. *Mitteilungen aus dem Geologisch-Palaeontologischen Institut der Universität Hamburg* **73**: 191–215.
- Kuschel G. 1994. Nemonychidae of Australia, New Guinea and New Caledonia. In: Zimmerman's *Australian Weevils*, Volume I. Melbourne, CSIRO.
- Kuschel G. 1995. A phylogenetic classification of Curculionoidea to families and subfamilies. *Memoirs of the Entomological Society of Washington* **14**: 5–33.
- Kuschel G, Leschen RAB, Zimmerman EC. 2000. Platypodidae under scrutiny. *Invertebrate Taxonomy* **14**: 771–805.
- Kuschel G, May BM. 1990. Palophaginae, a new subfamily for leaf-beetles, feeding as adult and larva on *Araucaria* pollen in Australia (Coleoptera: Megalopodidae). *Invertebrate Taxonomy* **3**: 697–719.
- Kuschel G, May BM. 1996. Discovery of Palophaginae (Coleoptera: Megalopodidae) on *Araucaria araucana* in Chile and Argentina. *New Zealand Entomologist* **19**: 1–13.
- Kuschel G, May BM. 1997. A new genus and species of Nemonychidae (Coleoptera) associated with *Araucaria angustifolia* in Brazil. *New Zealand Entomologist* **20**: 15–22.
- Kuschel G, Poinar GO. 1993. *Libanorhinus succinus* gen. & sp.n (Coleoptera; Nemonychidae) from Lebanese amber. *Entomologica Scandinavica* **24**: 143–146.
- Larsson SG. 1978. *Baltic Amber – a Palaeobiological Study*. Klampenborg, Denmark: Scandinavian Science Press Ltd.
- Liebherr JK, Zimmerman EC. 1998. Cladistic analysis, phylogeny and biogeography of the Hawaiian Platynini (Coleoptera: Carabidae). *Systematic Entomology* **23**: 137–172.

- Linder HP, Crisp MD. 1995. *Nothofagus* and Pacific biogeography. *Cladistics* **11**: 5–32.
- Lutzoni F, Pagel M. 1997. Accelerated evolution as a consequence of transitions to mutualism. *Proceedings of the National Academy of Sciences, USA* **94**: 11422–11427.
- Lyal CHC. 1995. The ventral structures of the weevil head (Coleoptera: Curculionoidea). *Memoirs of the Entomological Society of Washington* **14**: 35–51.
- Lyal CHC, King T. 1996. Elytro-tergal stridulation in weevils (Insecta: Coleoptera: Curculionoidea). *Journal of Natural History* **30**: 703–773.
- Martin HA. 1993. The palaeovegetation of the Murray Basin, Late Eocene to mid-Miocene. *Australian Systematic Botany* **6**: 491–531.
- Marvaldi A. 1997. Higher level phylogeny of Curculionidae (Coleoptera: Curculionoidea) based mainly on larval characters, with special reference to broad-nosed weevils. *Cladistics* **13**: 285–312.
- May BM. 1993. Larvae of Curculionoidea (Insecta: Coleoptera): A systematic overview. *Fauna of New Zealand* **28**: 1–223.
- McQuillan PB. 1993. *Nothofagus* (Fagaceae) and its invertebrate fauna – an overview and preliminary synthesis. *Biological Journal of the Linnean Society* **49**: 317–354.
- Mecke R, Engels W, Galileo MHM. 2000a. New records of Coleoptera and Hymenoptera from *Araucaria angustifolia* trees in the South of Brazil. XXI International Congress of Entomology, Foz de Iguazu, Brasil.
- Mecke R, Galileo MHM, Engels W. 2000b. Insetos e ácaros associados a *Araucaria angustifolia* (Araucariaceae: Coniferae) no Sul do Brasil. *Iheringia, Série Zoológica* **88**: 165–172.
- Meijer JJJ. 1997. Wood remains from the Late Cretaceous Aachen Formation. *Proceedings of the 4th European palaeobotanical and palynological conference* **58**: 131–137.
- Millar CI. 1993. Impact of the Eocene on the evolution of *Pinus* L. *Annals of the Missouri Botanical Gardens* **80**: 471–498.
- Miller CN. 1977. Mesozoic conifers. *Botanical reviews* **43**: 217–280.
- Miller CN. 1988. The origin of modern conifer families. In: Beck CB, ed. *Origin and evolution of Gymnosperms*. New York: Columbia University Press, 448–486.
- Moore BP. 1978. A new Australian stag beetle (Coleoptera: Lucanidae) with neotropical affinities. *Journal of the Australian Entomological Society* **17**: 99–103.
- Moran NA, Munson MA, Baumann P, Ishikawa H. 1993. A molecular clock in endosymbiotic bacteria is calibrated using the insect hosts. *Proceedings of the Royal Society of London Series B* **253**: 157–171.
- Moran NA. 1996. Accelerated evolution and Muller's ratchet in endosymbiotic bacteria. *Proceedings of the National Academy of Sciences USA* **93**: 2873–2878.
- Morrone JJ. 1996. Austral biogeography and relict weevil taxa (Coleoptera: Nemonychidae, Belidae, Brentidae and Caridae). *Journal of Comparative Biology* **1**: 123–127.
- Morrone JJ. 1997. Weevils (Coleoptera: Curculionidae) that feed on *Araucaria araucana* (Araucariaceae) in Southern Chile and Argentina, with an annotated checklist. *Folia Entomologica Mexicana* **100**: 1–14.
- Morrone JJ. 1998. On Udvardy's Insularctica province: a test from the weevils (Coleoptera: Curculionoidea). *Journal of Biogeography* **25**: 947–955.
- Nelson G, Platnick N. 1981. *Systematics and biogeography: cladistics and vicariance*. New York: Columbia University Press.
- Nielsen ES. 1987. The recently discovered primitive (non-Ditrysian) family Palaephatidae (Lepidoptera) in Australia. *Invertebrate Taxonomy* **1**: 201–229.
- Nissenbaum A, Horowitz A. 1992. The Levantine amber belt. *Journal of African Earth Sciences* **14**: 295–300.
- Nixon KC. 1999. The parsimony ratchet, a new method for rapid parsimony analysis. *Cladistics* **15**: 407–414.
- Normark BB, Jordal BH, Farrell BD. 1999. Origin of a haplodiploid beetle lineage. *Proceedings of the Royal Society of London Series B* **266**: 2553–2559.
- Ohsawa T, Nushida H, Nishida M. 1995. Yesonia, a new section of *Araucaria* (Araucariaceae) based on permineralized vegetative and reproductive organs of *A. vulgaris* comb. nov. from the upper Cretaceous of Hokkaido, Japan. *Journal of Plant Research* **108**: 25–39.
- Parsons MJ. 1996. A phylogenetic reappraisal of the birdwing genus *Ornithoptera* (Lepidoptera: Papilionidae: Troidini) a new theory of its evolution in relation to Gondwanan vicariance biogeography. *Journal of Natural History* **30**: 1707–1736.
- Peek AS, Vrijenhoek RC, Gaut BS. 1998. Accelerated evolutionary rate in sulfur-oxidizing endosymbiotic bacteria associated with the mode of symbiont transmission. *Molecular Biology and Evolution* **15**: 1514–1523.
- Pellmyr O, Leebens-Mack J. 1999. Forty million years of mutualism: Evidence for Eocene origin of the yucca-yucca moth association. *Proceedings of the National Academy of Sciences USA* **96**: 9178–9183.
- Pole M. 1992. Eocene vegetation from Hasties Northeastern Tasmania. *Australian Systematic Botany* **5**: 431–475.
- Pole M. 1995. Late Cretaceous macrofloras of Eastern Otago, New Zealand: Gymnosperms. *Australian Systematic Botany* **8**: 1067–1106.
- Pole M. 1998. Paleocene gymnosperms from Mount Somers, New Zealand. *Journal of the Royal Society of New Zealand* **28**: 375–403.
- Pole MS. 2000. Mid-Cretaceous conifers from the Eromanga Basin, Australia. *Australian Systematic Botany* **13**: 153–197.
- Pole M, Douglas B. 1999. Plant macrofossils of the upper Cretaceous Kaitangata coalfield, New Zealand. *Australian Systematic Botany* **12**: 331–364.
- Pollock DA. 1995. Classification, reconstructed phylogeny and geographical history of genera of Pilipalpinae (Coleoptera: Tenebrionoidea: Pyrochroidae). *Invertebrate Taxonomy* **9**: 563–708.
- Raubeson LA, Gensel PG. 1991. Upper Cretaceous conifer leaf fossils from the Black Creek formation with an assessment of affinities using principal components analysis. *Botanical Gazette* **152**: 380–391.

- Reid CAM. 2000.** Spilopyrinae Chapuis: a new subfamily in the Chrysomelidae and its systematic placement (Coleoptera). *Invertebrate Taxonomy* **14**: 837–862.
- Renner SS, Foreman DB, Murray D. 2000.** Timing transantarctic disjunctions in the Atherospermataceae (Laurales): Evidence from coding and noncoding chloroplast sequences. *Systematic Biology* **49**: 579–591.
- Riedel A. 1995.** [Research archives and Interests]. *Curculio* **39**: 3.
- Rosen D. 1978.** Vicariant patterns and historical explanation in biogeography. *Systematic Zoology* **27**: 159–188.
- Rottlaender R, Mischer G. 1970.** Chemical studies of lower Cretaceous amber from Lebanon. *Neues Jahrbuch für Geologie und Paläontologie Monatshefte* **11**: 668–673.
- Rowett AI. 1992.** Dispersed cuticular floras of South Australian Tertiary coalfields; Part 2, Lochiel. *Transactions of the Royal Society of South Australia* **116**: 95–107.
- Savard L, Li P, Strauss SH, Chase MW, Michaud M, Bousquet J. 1994.** Chloroplast and nuclear gene sequences indicate Late Pennsylvanian time for the last common ancestor of extant seed plants. *Proceedings of the National Academy of Sciences USA* **91**: 5163–5167.
- Schultze MJ. 1966.** Ergebnisse der Forschungsreise Richard Krausel's 1954 nach Sued- und Suedwest-Afrika, I, 9; gymnospermen-Hoelzer aus den oberkretazischen Umzamba-Schichten von Ost-Pondoland (S-Afrika). *Senckenbergiana Lethaea* **47**: 279–323.
- Sequeira AS, Lanteri AA, Scataglini MA, Confalonieri VA, Farrell BD. 2000a.** Are flightless *Galapaganus* weevils older than the Galapagos Islands they inhabit? *Heredity* **85**: 20–29.
- Sequeira AS, Normark BB, Farrell BD. 2000b.** Evolutionary assembly of the conifer fauna: Distinguishing ancient from recent associations in bark beetles. *Proceedings of the Royal Society of London Series B* **267**: 2359–2366.
- Setoguchi H, Osawa TA, Pintaud JC, Jaffre T, Veillon JM. 1998.** Phylogenetic relationships within Araucariaceae based on RBCL gene sequences. *American Journal of Botany* **85**: 1507–1516.
- Sikes DS, Lewis PO. 2000.** *PAUP Ratchet*. University of Connecticut.
- Smith AG, Hurley AM, Briden JC. 1981.** *Phanerozoic paleocontinental world maps*. Cambridge: Cambridge University Press.
- Sorenson MD. 1999.** *TreeRot.v2*. Boston University, Boston.
- Stockey RA. 1980.** Jurassic araucarian cone from southern England. *Palaeontology* **23**: 657–666.
- Stockey RA. 1982.** The Araucariaceae: an evolutionary perspective. *Review of Palaeobotany and Palynology* **37**: 133–154.
- Stockey RA, Nishida H, Nishida M. 1992.** Upper Cretaceous araucarian cones from Hokkaido: *Araucaria nihongii* sp. nov. *Review of Paleobotany and Palynology* **72**: 27–40.
- Stockey RA, Nishida H, Nishida M. 1994.** Upper Cretaceous araucarian cones from Hokkaido and Saghalien: *Araucaria nipponensis* sp. nov. *International Journal of Plant Sciences* **155**: 806–815.
- Straneo SL, Ball GE. 1989.** Synopsis of the genera and subgenera of the tribe Peleciini, and revision of the Neotropical and Oriental Species (Coleoptera: Carabidae). *Insecta Mundi* **3**: 73–142.
- Sukh D, Zeba B. 1976.** *Araucaria indica* and two other conifers from the Jurassic-Cretaceous rocks of Madhya Pradesh, India. *The Palaeobotanist* **25**: 496–508.
- Swofford DL. 2000.** *PAUP**. *Phylogenetic Analysis Using Parsimony (*and Other Methods)*. Version 4. Sunderland, Massachusetts: Sinauer Associates.
- Thayer MK. 1985.** Revision, phylogeny and biogeography of the austral genus *Metacoraeolabium* Steel (Coleoptera: Staphylinidae: Omaliinae). In: Ball GE, ed. *Taxonomy and zoogeography of beetles and ants*. Dordrecht: Junk, 113–179.
- Thompson RT. 1992.** Observations on the morphology and classification of weevils (Coleoptera, Curculionoidea) with a key to major groups. *Journal of Natural History* **26**: 835–891.
- Torres T, Biro Bagoczky L. 1986.** Xylotomy of fossil conifers from Quiriquina Island, Chile. *Serie Comunicaciones – Departamento de Geología, Facultad de Ciencias Físicas y Matemáticas, Universidad de Chile* **37**: 65–80.
- Veillet Bartoszewska M. 1956.** Sur l'anatomie et les affinités du Dadoxylon (*Araucarioxylon*) sahariense nov. sp., bois fossile du Continental intercalaire de l'Emi-Fezzan. *Bulletin de la Société Géologique de France* **6**: 227–236.
- Voronova MA, Smykov SY. 1972.** Stratigraphic subdivision of the Cretaceous deposits in the middle Ingul River region. *Geologicheskii Zhurnal (Kiev, 1968)* **32**: 69–77.
- Wilde V, Goth K. 1987.** *Araucaria* seeds from the Lower Cretaceous of Brilon-Nehden. *Geologie und Paläontologie in Westfalen* **10**: 45–50.
- Wise KAJ. 1991.** Distribution and zoogeography of the New Zealand Megaloptera and Neuroptera. *Rec Auckland Inst Mus* **28**: 211–227.
- Wood SL. 1986.** A reclassification of the genera of Scolytidae (Coleoptera). *Great Basin Naturalist Memoirs* **10**: 1–126.
- Wood SL, Bright DE. 1992.** A catalog of Scolytidae and Platypodidae (Coleoptera), part 2: taxonomic index. *Great Basin Naturalist Memoirs* **13**: 1–1533.
- Yates DK, Irwin ME. 1996.** Apioceridae (Insecta: Diptera): cladistic reappraisal and biogeography. *Zoological Journal of the Linnean Society* **116**: 247–301.
- Zastawniak E. 1994.** Upper Cretaceous leaf flora from the Blaszyk Moraine (Zamek Formation), King George Island, South Shetland Islands, West Antarctica. *Acta Palaeobotanica* **34**: 119–163.
- Zimmerman EC. 1994.** *Australian Weevils*. Melbourne: CSIRO.