ORIGINAL ARTICLE

Placing the origin of two species-rich genera in the late cretaceous with later species divergence in the tertiary: a phylogenetic, biogeographic and molecular dating analysis of *Piper* and *Peperomia* (Piperaceae)

**James F. Smith · Angela C. Stevens ·
Eric J. Tepe · Chris Davidson**

Abstract Nearly all of the species diversity in Piperaceae is encompassed within *Piper* and *Peperomia*. Both genera are pan-tropical with areas of diversification in the Neotropics and Southeast Asia. Piperaceae are less diverse in Africa with only two native species of *Piper*. This study examines the distribution of both *Piper* and *Peperomia* with representative samples from the Neotropics, Asia, Pacific Islands, and Africa. Molecular dating is used to place an age for the crown clades of *Piper* and *Peperomia* as well as ages for diversification within the clades. Both genera have origins in the late Cretaceous, but species level diversification occurred much later in the Tertiary. Biogeography of both genera are correlated with paleoclimate evidence to better explain the distribution and diversification of these large genera.

Keywords Piperaceae · *Piper* · *Peperomia* · *g3pd* · *trn*L-F · *trn*L intron · *ndh*F · Biogeography

Introduction

The distribution of the world's biota, and how it got to where it presently is, has been a question of systematics and evolution since Darwin (1859). Modern tools are able to improve our ability to assess biogeography. Strongly supported phylogenetic hypotheses, and molecular sequence age estimations have greatly enhanced our understanding of plant and animal distribution and movement (Thorne et al. 1998; Rambaut and Bromham 1998; Kishino et al. 2001; Thorne and Kishino 2002; Aris-Brosou and Yang 2002; Sanderson 2002, 2003; Drummond and Rambaut 2005; Ree et al. 2005; Smith 2006). For example, hypotheses on the putative number of plant introductions into the Hawaiian islands were initially based on morphological differences and similarities to presumed ancestral groups which often resulted in suggestions that many plant taxa had arrived via multiple independent introductions (Wagner et al. 1990). Phylogenetic analyses, however, have indicated that a single introduction was more of the norm for most groups where several separate introductions had been proposed (Baldwin et al. 1990, 1991; Baldwin and Robichaux 1995; Givnish et al. 1996; Cronk et al. 2005) and in at least one case, two introductions were proposed where only one had been hypothesized (Howarth et al. 1997).

Recent efforts have been especially informative regarding biogeography of islands (DeJoode and Wendel 1992; Knox et al. 1993; Wagner and Funk 1995; Givnish et al. 1995, 1996; Howarth et al. 1997, 2003; Stuessy et al. 1998a, b; Alice and Campbell 1999; Ganders et al. 2000; Helfgott et al. 2000; Wright et al. 2000, 2001; Lindqvist and Albert 2002; Nepokroeff et al. 2003; Carine et al. 2004; Cronk et al. 2005) and northern hemisphere or temperate groups (Wen and Stuessy 1993; Qiu et al. 1995; Wen and Jansen 1995; Lee et al. 1996; Wen et al. 1996; Wen and Zimmer 1996; Hasebe et al. 1998; Kim and Jansen 1998; Schnabel and Wendel 1998; Xiang et al. 1998; Wen et al. 1998; Kim and Kim 1999; Swenson et al.

J. F. Smith (✉) · A. C. Stevens
Department of Biological Sciences, Boise State University,
1910 University Drive, Boise, ID 83725-1515, USA
e-mail: jfsmith@boisestate.edu

E. J. Tepe
Department of Biology, University of Utah, Salt Lake City,
UT 84112, USA

C. Davidson
Idaho Botanical Research Foundation, 637 Warm Springs,
Boise, ID 83712, USA

2001; Bell and Donoghue 2005; Alsos et al. 2007; Bell 2007), but attention has generally been lacking for tropical organisms. This is especially true for plants with species or clades in South America, Southeast Asia and Africa (Givnish et al. 2000, 2004; Conti et al. 2002; Davis et al. 2002). Vicariance has often been used to explain distributions that cross major barriers to dispersal (Nelson and Platnick 1981; Humphries and Parenti 1999; Conti et al. 2002; Crisp and Cook 2007), especially for clades with relatively older origins. However, much recent data have begun to show that many plant and animal groups that seemed to fit a vicariant distribution pattern are actually the result of more recent, long distance dispersal events (Renner 2004; Sanmartín and Ronquist 2004; de Queiroz 2005; McGlone 2005; Poux et al. 2005; Winkworth et al. 2005; Smith et al. 2006; Morris et al. 2007).

Piperaceae provide a means of examining the biogeography of a pan-tropical group. Piperaceae consist of only five genera, and the vast majority of the estimated 3,600 species occur nearly equally in two genera, *Piper* and *Peperomia*. Both genera are found in tropical regions throughout the world (Miquel 1843–1844; Grieg 2004; Wanke et al. 2007a; Quijano-Abril et al. 2006). Previous phylogenetic analyses that have examined *Piper* on the global level identified three major clades, one each in the Neotropics, Southeast Asia, and Pacific Islands (Jaramillo and Manos 2001; Jaramillo et al. in press).

The origin of the African species of *Piper* pose an interesting question. There are two species indigenous to Africa. *Piper guineense* is a dioecious vine, reminiscent of the majority of the species of Southeast Asia. *Piper capense* is a shrub with bisexual flowers, more similar in habit and floral form to species of the Neotropics. The African species imply either a single origin for the genus on the continent and a shift in breeding system, or multiple origins. Likewise unusual is the paucity of species. Africa was previously dominated by lowland tropical rainforest and Neogene aridification is thought to have resulted in the extinction of many African tropical lineages with the result that the African flora is less diverse than that of Asia or South America (Axelrod 1972; Axelrod and Raven 1978; Raven and Axelrod 1974; Whitmore 1998). Alternatively, the low diversity of *Piper* may reflect a more recent introduction event into Africa with insufficient time for species diversification as has been proposed for Rapateaceae (Givnish et al. 2004).

Most species of *Piper* tend to occupy relatively small geographic ranges (Marquis 2004). Some species are widespread (e.g. *Pi. umbellatum*), spread on purpose for their utility (*Pi. nigrum, Pi. methysticum*) or have been distributed by accident or have escaped from cultivation (*Pi. auritum*). However, most species of *Piper* are known only from a single geographic region, island, or continent (Marquis 2004) with a few exceptions that are often thought to be recent human-mediated introductions (e.g. *Pi. aduncum*). The distributional pattern is similar in *Peperomia*, and again some species are geographically widespread. Some of this distribution of *Peperomia* may have occurred as a combined result of their weedy nature and thriving in disturbed areas, human-mediated introductions and producing an abundance of small seed-like fruits (e.g. *Pe. pellucida*). However, other species are found in relatively natural and undisturbed areas, suggesting that their distributions are not the result of human intervention. *Peperomia tetraphylla* is such an example with populations known from China, the Neotropics, Pacific Islands, Africa, Australia, New Zealand, Nepal, India and Indonesia (S. Wanke, personal communication).

It is the goal of this study to undertake a phylogenetic analysis of *Piper* and *Peperomia* species from the Neotropics, Asia, Pacific Islands, Australia, and Africa to better resolve the biogeographic relations within this large pan-tropical group of plants. We have sampled from all geographic regions for *Piper* and nearly all for *Peperomia*, including individuals of three *Peperomia* species from different geographic regions. Our phylogenetic analysis is based on DNA sequences of three chloroplast regions: *ndh*F, *trn*L intron, and *trn*L-*trn*F spacer, and the low copy nuclear gene glyceraldehyde 3-phosphate dehydrogenase (*g3pd*; Strand et al. 1997).

Materials and methods

Species used in the analysis, voucher information and Genbank accession numbers for all sequences are listed in Table 1. Data are available from the first author on request. DNA was extracted using Qiagen DNeasy extraction kits following the manufacturer's instructions. Amplification of *ndh*F, *trn*L intron, and the *trn*L-*trn*F spacer followed protocols used for other taxa (Smith et al. 2004). The amplification, cloning and sequencing of the low copy number nuclear gene *g3pd* used the primers of Strand et al. (1997) with the addition of BSA (final concentration of 1 μg/μL) and 2% DMSO to the reaction mix. Thermal cycling parameters entailed an initial cycle of 3 min at 94°C, followed by 2 cycles of 1 min at 94°C, 1 min at 52°C and 1.5 min at 72°C followed by cycles of step-down annealing temperatures, each repeated twice ranging from 52 to 40°C with a final 24 cycles at 39°C. The pGEM-T vector system kit (Promega) was used to clone *g3pd*. Up to 20 clones for some species were sequenced in an attempt to detect potential paralogs. Sequences were obtained from both strands of all gene regions with a Li-Cor LongreadIR 4200 automated sequencer.

Table 1 Species used in this analysis, their collection locality, voucher information, and Genbank accession numbers

Species	Voucher (herbarium acronym)	Collection locality	Genbank numbers, *g3pd*, *trn*L-*trn*F, *trn*L intron, *ndh*F
Pi. aduncum L.	A. Bornstein 765 (SEMO)	Nicaragua	EU519540, EU519630, EU519810, EU519720
Pi. aduncum L.	L. A. Hahn 438 (SRP)	Dominican Republic	EU519539, EU519629, EU519809, EU519719
Pi. aequale Vahl.	A. Bornstein 716 (SEMO)	Honduras	EU519550, EU519640, EU519820, EU519730
Pi. amalago L.	A. Bornstein 712 (SEMO)	Honduras	EU519543, EU519633, EU519813, EU519723
Pi. arboreum Aubl.	Aburri Botanical Garden collection number 042 (Aburri Botanical Garden)	Cultivated, Ghana	EU519525, EU519610, EU519795, EU519705
Pi. arboreum Aubl.	A. Bornstein 699 (SEMO)	Honduras	EU519524, EU519614, EU519794, EU519704
Pi. augustum Rudge	E. Tepe 525 (MU)	French Guiana	EU519644, EU519634, EU519814, EU519724
Pi. auritum H. B. & K.	C. Davidson 10892 (SRP)	La Selva, Costa Rica	EU519535, EU519625, EU519805, EU519715
Pi. auritum H. B. & K.	A. Rincon 2409 (XAL)	Veracruz, Mexico	EU519534, EU519624, EU519804, EU519714
Pi. austro-caledonicum C. DC.	G. McPherson 19190 (MO)	New Caledonia	EU519508, EU519598, EU519778, EU519688
Pi. avellanum C. DC.	E. Tepe 616 (MU)	French Guiana	EU519553, EU519643, EU519823, EU519733
Pi. betle L.	C. Davidson 11001 (SRP)	Tanzania	EU519505, EU519595, EU519775, EU519685
Pi. betle L.	J. F. Smith 5808 (SRP)	Cultivated	EU519506, EU519596, EU519776, EU519686
Pi. borbonense C. DC.	Accession number 87.3.616 for Conservatoire et Jardins Botaniques de Nancy, not vouchered	Cultivated, originally from Reunion	EU519495, EU519585, EU519765, EU519675
Pi. caninum Blume	T. Flynn 6750 (PTBG)	Cultivated, Australia	EU519501, EU519591, EU519771, EU519681
Pi. capense L. f.	J. F. Smith 4925 (SCA)	Cameroon	EU519488, EU519578, EU519758, EU519668
Pi. capense L. f.	C. Davidson 11009 (SRP)	Kenya	EU519489, EU519579, EU519759, EU519669
Pi. chandocanum C. DC.	Li J.-M. 06182 (PE)	Guangxi, China	EU519515, EU519605, EU519785, EU519695
Pi. colonense C. DC.	A. Bornstein 753 (SEMO)	Nicaragua	EU519538, EU519628, EU519808, EU519718
Pi. concepcionis Trel.	C. Davidson 10880 (SRP)	Costa Rica	EU519545, EU519635, EU519815, EU519725
Pi. costatum C. DC.	T. Flynn 4261 (PTBG)	Cultivated	EU519499, EU519589, EU519769, EU519679
Pi. flaviflorum C. DC.	Li J.-M. 06171 (PE)	Yunnan, China	EU519518, EU519608, EU519788, EU519698
Pi. guahamense C. DC.	T. Flynn 6748 (PTBG)	Cultivated	EU519498, EU519588, EU519768, EU519678
Pi. guazacapanense Trel. & Standl.	M. A. Perez Farrera 2923 (HEM)	Chiapas, Mexico	EU519547, EU519637, EU519817, EU519727
Pi. guineense Schumach. & Thonn.	C. Davidson 11005 (SRP)	Uganda	EU519492, EU519582, EU519762, EU519672

Table 1 continued

Species	Voucher (herbarium acronym)	Collection locality	Genbank numbers, *g3pd*, *trn*L-*trn*F, *trn*L intron, *ndh*F
Pi. guineense Schumach. & Thonn.	C. Davidson 11006 (SRP)	Uganda	EU519493, EU519583, EU519763, EU519673
Pi. guineense Schumach. & Thonn.	C. Davidson 11008 (SRP)	Kenya	EU519494, EU519584, EU519764, EU519674
*Pi. guineens*e Schumach. & Thonn.	J. F. Smith 4923 (SCA)	Cameroon	EU519490, EU519580, EU519760, EU519670
Pi. guineense Schumach. & Thonn.	J. F. Smith 4924 (SCA)	Cameroon	EU519491, EU519581, EU519761, EU519671
Pi. hancei Maximowicz	Li J.-M. 06211 (PE)	Guizhou, China	EU519512, EU519602, EU519782, EU519692
Pi. hispidum Sw.	A. Rincon 2327 (XAL)	Veracruz, Mexico	EU519541, EU519631, EU519811, EU519721
Pi. hostmannianum (Miq.) C. DC.	E. Tepe 599 (MU)	French Guiana	EU519545, EU519636, EU519816, EU519726
Pi. humistratum Görts & K. U. Kramer	E. Tepe 542 (MU)	French Guiana	EU519549, EU519639, EU519819, EU519729
Pi. imperiale (Miq.) C. DC.	C. Davidson 10882 (SRP)	Costa Rica	EU519552, EU519642, EU519822, EU519732
Pi. methysticum G. Forster	C. W. Morden 2957 (HAW)	Hawaii	EU519496, EU519586, EU519766, EU519676
Pi. methysticum G. Forster	C. W. Morden 2975 (HAW)	Hawaii	EU519497, EU519587, EU519767, EU519677
Pi. muricatum Blume	W. S. Wong 2 (SRP)	Malaysia	EU519526, EU519616, EU519796, EU519706
Pi. nigrum L.	J. F. Smith 5807 (SRP)	Cultivated	EU519509, EU519599, EU519779, EU519689
Pi. nudifolium C. DC.	C. Davidson 10897 (SRP)	Costa Rica	EU519536, EU519626, EU519806, EU519716
Pi. obliquum Ruiz & Pav.	A. Bornstein 738 (SEMO)	Nicaragua	EU519537, EU519628, EU519807, EU519717
Pi. peltatum L.	C. Davidson 10898 (SRP)	Costa Rica	EU519532, EU519622, EU519802, EU519712
Pi. peltatum L.	E. Tepe 589 (MU)	French Guiana	EU519533, EU519623, EU519803, EU519713
Pi. pingbienense Y. C. Tseng	Li J.-M. 06151 (PE)	Yunnan, China	EU519517, EU519607, EU519787, EU519697
Pi. porphyrophyllum N. E. Br.	W. S. Wong 3 (SRP)	Malaysia	EU519519, EU519609, EU519789, EU519699
Pi. pseudofuligineum C. DC.	A. Bornstein 710 (SEMO)	Honduras	EU519542, EU519632, EU519812, EU519722
Pi. puberulum (Benth.) Maxim.	T. Flynn 6749 (PTBG)	Cultivated	EU519500, EU519590, EU519770, EU519680
Pi. rothianum F. M. Bailey	J. F. Smith 6545 (SRP)	Queensland, Australia	EU519502, EU519592, EU519772, EU519682
Pi. sanctum Schltdl. ex. Miq.	A. Bornstein 744 (SEMO)	Nicaragua	EU519521, EU519611, EU519791, EU519701
Pi. sanctum Schltdl. ex. Miq.	H. G. Domingues 251 (HEM)	Chiapas, Mexico	EU519520, EU519610, EU519790, EU519700
Pi. sanctum Schltdl. ex. Miq.	A. Rincon 2352 (XAL)	Veracruz, Mexico	EU519522, EU519612, EU519792, EU519702
Pi. sanctum Schltdl. ex. Miq.	M. A. Perez Farrera 2820 (HEM)	Chiapas, Mexico	EU519523, EU519613, EU519793, EU519703
Pi. sarmentosum Roxburgh	J. F. Smith 5806 (SRP)	Cultivated	EU519510, EU519600, EU519780, EU519690

Table 1 continued

Species	Voucher (herbarium acronym)	Collection locality	Genbank numbers, *g3pd*, *trn*L-*trn*F, *trn*L intron, *ndh*F
Pi. semiimmersum C. DC.	Li J.-M. 06161 (PE)	Yunnan, China	EU519514, EU519604, EU519784, EU519694
Pi. submultinerve C. DC.	Li J.-M. 061410 (PE)	Yunnan, China	EU519516, EU519606, EU519786, EU519696
Pi. subpenninerve Ridl.	W. S. Wong 1 (SRP)	Malaysia	EU519503, EU519593, EU519773, EU519683
Pi. thomsonii (C. DC.) Hook. f.	Li J.-M. 061511 (PE)	Yunnan, China	EU519513, EU519603, EU519783, EU519693
Pi. umbellatum L.	A. Rincon 2325 (XAL)	Mexico	EU519528, EU519618, EU519798, EU519708
Pi. umbellatum L.	J. F. Smith 4926 (SCA)	Cameroon	EU519529, EU519619, EU519799, EU519709
Pi. umbellatum L.	C. Davidson 11010 (SRP)	Kenya	EU519530, EU519620, EU519800, EU519710
Pi. umbellatum L.	C. Davidson s.n. (SRP)	Tanzania	EU519531, EU519621, EU519801, EU519711
Pi. umbellatum L.	A. Bornstein 709 (SEMO)	Honduras	EU519527, EU519617, EU519797, EU519707
Pi. urophyllum C. DC. ex Pittier	C. Davidson 10902 (SRP)	Costa Rica	EU519551, EU519641, EU519821, EU519731
Pi. wallichii (Miq.) Handel-Mazzetti	Li J.-M. (PE)	Yunnan, China	EU519511, EU519601, EU519781, EU519691
Pi. yucatanense C. DC.	M. A. Perez Farrera 2898 (SRP)	Tabasco, Mexico	EU519548, EU519638, EU519818, EU519728
Pi. sp.	Harder 5565 (MO)	Vietnam	EU519507, EU519597, EU519777, EU519687
Pi. sp.	C. Davidson 11000 (SRP)	Tanzania	EU519504, EU519594, EU519774, EU519684
Peperomia blanda (Jacquin) Kunth	Li J.-M. 06262 (PE)	Guangxi, China	EU519471, EU519561, EU519741, EU519651
Pe. blanda (Jacquin) Kunth	J. E. Burrows 8521 (herbarium at Buffelskloof Private Nature Reserve, Mpumalanga,South Africa)	South Africa	EU519470, EU519560, EU519740, EU519650
Pe. caperata Yunck.	J. F. Smith 4029 (SRP)	Cultivated	NA, EU519556, EU519736, EU519646
Pe. cavalierii C. DC.	Li J.-M. 06201 (PE)	Guangxi, China	EU519472, EU519562, EU519742, EU519652
Pe. emarginella C. DC.	C. Davidson 10867 (CR)	Costa Rica	EU519486, EU519576, EU519756, EU519666
Pe. fernandopoiana C. DC.	J. F. Smith 4927 (SCA)	Cameroon	EU519475, EU519565, EU519745, EU519655
Pe. glabella (Sw.) A. Dietr.	C. Davidson 10895 (SRP)	Costa Rica	EU519484, EU519574, EU519754, EU519664
Pe. hernandiifolia Trel.	C. Davidson 10905 (CR)	Costa Rica	EU519479, EU519569, EU519749, EU519659
Pe. hesperomannii Wawra	Hawaiian Plant DNA Laboratory 1783 (HAW)	Hawaii	EU519481, EU519571, EU519751, EU519661
Pe. kamerunana C. DC.	J. F. Smith 4928 (SCA)	Cameroon	EU519480, EU519570, EU519750, EU519660
Pe. leptostachya Hook. & Arnott	Hawaiian Plant DNA Laboratory 37 (HAW)	Hawaii	EU519483, EU519573, EU519753, EU519663

Table 1 continued

Species	Voucher (herbarium acronym)	Collection locality	Genbank numbers, *g3pd*, *trn*L-*trn*F, *trn*L intron, *ndh*F
Pe. pellucida (L.) Kunth	Aburri Botanical Garden 040 (Aburri Botanical Garden Herbrium)	Ghana	EU519477, EU519567, EU519747, EU519657
Pe. pellucida (L.) Kunth	J. F. Smith 4929 (SCA)	Cameroon	EU519476, EU519566, EU519746, EU519656
Pe. pellucida (L.) Kunth	E. Tepe 578 (MU)	French Guiana	EU519478, EU519568, EU519748, EU519658
Pe. retusa A. Dietr.	J. E. Burrows 8923 (Buffelskloof herbarium at Buffelskloof Private Nature Reserve, Mpumalanga,South Africa)	South Africa	EU519473, EU519563, EU519743, EU519653
Pe. retusa A. Dietr.	C. Davidson 11003 (SRP)	South Africa	EU519474, EU519564, EU519744, EU519654
Pe. sandwicensis Miq.	Hawaiian Plant DNA Laboratory 1784 (HAW)	Hawaii	EU519482, EU519572, EU519752, EU519662
Pe. serpens Sw.	E. Tepe 548 (MU)	French Guiana	EU519487, EU519577, EU519757, EU519667
Pe. tetraphylla (G. Forster) Hook. & Arnott	C. Davidson 10907 (SRP)	South Africa	NA, EU519557, EU519737, EU519647
Pe. tetraphylla (G. Forster) Hook. & Arnott	J. E. Burrows 8924 (Buffelskloof herbarium at Buffelskloof Private Nature Reserve, Mpumalanga,South Africa)	South Africa	NA, EU519558, EU519738, EU519648
Pe. tetraphylla (G. Forster) Hook. & Arnott	Li J.-M. 061417 (PE)	Yunnan, China	NA, EU519559, EU519739, EU519649
Pe. urocarpa Fisch. & C. A. Mey.	C. Davidson 10889 (SRP)	Costa Rica	EU519485, EU519575, EU519755, EU519665
Houttuynia cordata Thunb.	J. F. Smith 4922 (SRP)	Cultivated	EU519469, EU519555, EU519735, EU519645
Saururus cernuus L.	A. Bornstein 863 (SEMO)	Cultivated	NA, EU519554, EU519734, EU519644

NA not amplified

Alignment of all sequences was done manually. Prior to combining *g3pd* and cpDNA sequences an ILD test (Farris et al. 1994) was run, implemented as the partition homogeneity test (PHT) of PAUP*. As this test is often sensitive to low signal in some of the partitions (Reeves et al. 2001; Yoder et al. 2001), separate maximum parsimony (MP) bootstrap (BS; Felsenstein 1985) analyses were run for each of the partitions (cpDNA and *g3pd*) to search for areas of hard incongruence (Seelanen et al. 1997). The data were analyzed using PAUP* 4.0b10 (Swofford 2002) for MP and BS. Indels were either treated as missing data, or rescored following the methods of Simmons and Ochoterena (2000). For the MP analyses, the data were analyzed using the search option of Olmstead and Palmer (1994) where five searches are performed using 1,000 replicates with nearest neighbor interchange and MulTrees Off saving all shortest trees. The results of each of these searches are then used as the starting trees

for a search using tree-bisection reconnection (TBR) and MulTrees on. The shortest trees from all searches were combined and a strict consensus was created. Bootstrap replicates were performed with 100 searches with 10 random replicates using full heuristic search with TBR and MulTrees on.

Modeltest Version 3.27 (Posada and Crandall 1998) was used to determine the best model that fit the data for Bayesian inference (BI) analyses. Results of these tests were used as priors in BI analyses with a complex (different model for each partition) or simple model for the combined data using either the likelihood ratio test (LRT), or Aikake information content (AIC) criteria (Table 2).

Bayesian inference analyses employed MRBAYES 3.0B4 (Huelsenbeck and Ronquist 2001). Analyses were run using two million generations saving trees every 100 generations with a burn-in of 25,000 generations. Analyses

Table 2 Details of the different DNA regions used in this analysis

Size average (range)	*ndh*F 2,521 (2,320–2,583)	*trn*L intron 499 (413–533)	*trn*L-*trn*F spacer 423 (357–510)	*g3pd* 838 (715–1,007)
Aligned size	2,705	617	704	1,209
Number of potentially informative characters (MP)	908	211	169	742
Modeltest result (LRT)	TVM + G			HKY + G + I
Modeltest result (AIC)	TVM + G			TVM + G + I

Note that the three cpDNA markers were combined into a single partition in all analyses

were repeated for each run with the order of species scrambled between runs.

The Kishino–Hasegawa and Shimodaira–Hasegawa tests (KH, Kishino and Hasegawa 1989; SH, Shimodaira and Hasegawa 1999) were both used to test trees where constraints were imposed on the search to make geographic regions (Neotropics and Africa for *Piper*; Asia, Neotropics, Africa, and Pacific Islands for *Peperomia*) that were not monophyletic into monophyletic groups, as well as constrain *Pe. retusa* as monophyletic. We include results from both tests but acknowledge that there are problems with KH tests (Goldman et al. 2000). Parameters from the AIC selection criteria of Modeltest for the combined data set were used to compute the likelihoods since AIC has been shown to provide better results (Posada and Buckley 2004). Search parameters utilized optimization with 1,000 replicate bootstrap samples.

Ancestral distribution areas were constructed with dispersal-vicariance analysis (DIVA Version 1.1; Ronquist 1996, 1997). DIVA minimizes the number of dispersal and extinction events needed to explain the pattern of terminal taxa. We scored terminal taxa based on their current distribution. For the most part this is relatively easy as species of *Piper* tend to be fairly local endemics (Marquis 2004). However, for some species we included accessions in our phylogenetic analysis that were not from their presumed native distribution (e.g., *Pi. umbellatum*). In cases where we assumed a species had been introduced (cultivated species) we scored the terminals for their native range. *Piper caninum* is known from both Australia and Malaysia (Chew 1972). The accession we used was from cultivation and presumably Australian in origin, however, since the species has a broader distribution we scored it as present in both Australia and Asia. For *Peperomia* species, this poses a greater challenge since some species are more widespread and their ancestral native range is not currently known. For *Peperomia* we scored current distribution based on the locality for which the individual in the analysis was from. Multiple analyses using different biogeographic regions as alternate tests also were run. Previous phylogenetic analyses had indicated that *Piper* fit three major clades that corresponded to three distinct

biogeographic regions; Asia, Pacific Islands, and Neotropics (Jaramillo and Manos 2001). We used these as our basis, but since we included additional species from China, Malaysia, Australia, Africa, and Reunion, we did additional analyses with these areas separated from the main three. To distinguish among the effects of *Peperomia* and *Piper* on the results, we also analyzed the two clades independently of each other. DIVA also requires a fully bifurcated tree, therefore we arbitrarily selected a single MP tree with arbitrary relationships among the polytomy of Asian species that was recovered. Previous analyses (Jaramillo and Manos 2001; Jaramillo et al. in press) have indicated that the Neotropical species of *Piper* are likely to be a monophyletic group. To examine the impact of altering the position of *Pi. sanctum*, *Pi. aequale*, and *Pi. urophyllum*, we also examined biogeographic patterns in DIVA constraining these three species at the base of the Neotropical clade with *Pi. sanctum* sister to all remaining Neotropical species.

DIVA does not allow the user to separate the histories of the areas that taxa are found in and the lineages of the taxa themselves (Ree et al. 2005). Since information on the geographic regions of Piperaceae is known, it would be valuable to include the information as part of the model. Therefore, we also employed AReA v. 2.1 (Smith 2006) as a means of re-assessing the biogeographic history of Piperaceae. AReA requires an ultrametric tree, therefore, we utilized the program r8s (Sanderson 2003) to put dates on our tree.

In addition to an ultrametric tree, AReA allows the user to implement priors regarding potential connections between geographic regions, which can vary over geological time. Other priors include the likely longevity of taxa and their vagility. We designated our global distribution to comprise five regions: Australia (including New Caledonia), Asia, Africa, the Neotropics, and the Pacific Islands (excluding New Caledonia). We tested two different models of connections. The first of these is designated the dispersal model since it reflects potential connections that may have been present due to dispersal, generally longer distances and over water barriers. The following connections were set to an arbitrary number of 0.25 prior to

60 mya to reflect the lower possibility of dispersal between these regions for the dispersal model: Pacific Islands to the Neotropics (Smith et al. 2006), Pacific Islands to Australia (Wright et al. 2000, 2001), Pacific Islands to Asia (Cronk et al. 2005), and Australia to Asia (Cronk et al. 2005). The only other connections allowed were Africa to the Neotropics (Sanmartín and Ronquist 2004; Givnish et al. 2004; Särkinen et al. 2007) and Africa to Australia (Swenson and Bremer 1997) prior to 60 mya, which were arbitrarily set at 1.0. From 60 mya to the present, the same six connections were allowed, but all with an arbitrary value of 0.5. The other model is termed the vicariant model and makes connections that would have occurred as a result of either vicariance, or dispersals that could have occurred over short distances (all land based). Connections in this model were more structured over time and were based on models of continent distributions during these time periods (Cox and Moore 2000). For the time between 140 and 110 mya the following arbitrary values were used: Neotropics to Africa 1.0, Neotropics to Australia (via Antarctica) 0.75. For 110–90 mya the connection between the Neotropics to Australia was reduced to 0.6 and from 90 to 40 mya the connection between the Neotropics and Africa was set at 0.75, between the Neotropics and Australia at 0.5 and a new connection between Africa and Asia was added at 0.6. From 40 mya to the present, it was assumed that all connections would be the result of dispersal and were not designated. Two different runs were completed using different arbitrary values for vagility (0.01 and 0.1) both with a single arbitrary value for longevity (0.01) for each model. As with DIVA, we also examined the effects while constraining all Neotropical *Piper* as monophyletic.

Piperaceae have a poor fossil record with a few fossils dating to the Pleistocene (Horn et al. 2003). However, since nearly all recent molecular phylogenetic analyses have indicated that Piperales are a relatively early divergent lineage (Qiu et al. 1999; Soltis et al. 2000; Doyle and Endress 2000; Zanis et al. 2002; Jaramillo and Kramer 2007), it is extremely unlikely that the family has only a recent history and it is more likely that the small flowers and their fleshy fruits have not been well preserved. The recent discovery of a fossil Saururaceae is encouraging in that Piperaceae fossils may yet be discovered (Smith and Stockey 2007). However, the recent age (middle Eocene) and placement phylogenetically as sister to *Saururus* (Smith and Stockey 2007), preclude the use of this fossil to calibrate our tree. *Appomatoxia* has been suggested to resemble the extant species *Zippelia* (Piperaceae) on the basis of shared spiny fruits (Friis et al. 1995). However, in the original description of this fossil Friis et al. (1995) did not think the similarity sufficient to place it within Piperales and recent analyses that have included *Appomatoxia* in phylogenetic analyses indicate that a more parsimonious

explanation maintains its status as a relative of Chloranthaceae, although placement within Piperales is only one step longer (Doyle and Endress 2007). Therefore, we resolved to look outside of Piperaceae and examine closely related families to determine if any of these had a better fossil record. *Lactoris* has a good fossil record placing the genus in the late Turonian at 91.2 mya (Zavada and Benson 1987; MacPhail et al. 1999; Magallón and Sanderson 2001). Since recent molecular data have placed *Lactoris* within or close to Aristolochiaceae (Nickrent et al. 2002; Borsch et al. 2005; Neinhuis et al. 2005; Wanke et al. 2007a) and Aristolochiaceae are sister to Saururaceae/Piperaceae (Nickrent et al. 2002; Borsch et al. 2005; Neinhuis et al. 2005; Wanke et al. 2007a) Piperaceae/Saururaceae would have diverged from Aristolochiaceae by that time if not earlier. This gives us a minimum stem age for Piperaceae/Saururaceae, but we are uncertain about the crown age. However, to avoid adding arbitrary bias, we used the age of 91.2 mya to calibrate our tree. We recognize that this date is potentially erroneous, but provides a starting point to begin the investigation of biogeography and evolution within Piperaceae and to establish approximate dates for the evolution within this family.

Penalized likelihood (PL; Sanderson 2002) was used to estimate the ages of nodes for Piperaceae. Cross validation was conducted to obtain the optimal smoothing value. Increments were set at 0.5 with 14 steps. To examine the impact of smoothing values on our data, we ran several separate analyses with smoothing values ranging from 2 to 100. As with DIVA, we opted to use a fully resolved topology and we used the same tree used with DIVA.

Results

Successful amplifications were obtained for all DNA regions for nearly all individuals sampled. Glyceraldehyde 3-phosphate dehydrogenase was not successfully amplified from any of the accessions of *Peperomia tetraphylla* included in the analysis, *Pe. caperata*, or *Saururus cernuus*. Phylogenetic analyses were conducted with missing data for these individuals, as well as excluding them from the analysis. Exclusion did not change the topology of the trees, therefore the results presented here are based on analyses that included all accessions. Table 2 summarizes information on each of these regions as well as the results of Modeltest 3.27 (Posada and Crandall 1998). The model selected for the combined, four gene data set was TVM + G with G = 0.8658, using LRT and AIC.

The PHT indicated significant differences between the two paritions ($p = 0.01$). However, this is likely due to suboptimal trees being found in some of the data partitions where low signal did not result in strongly supported
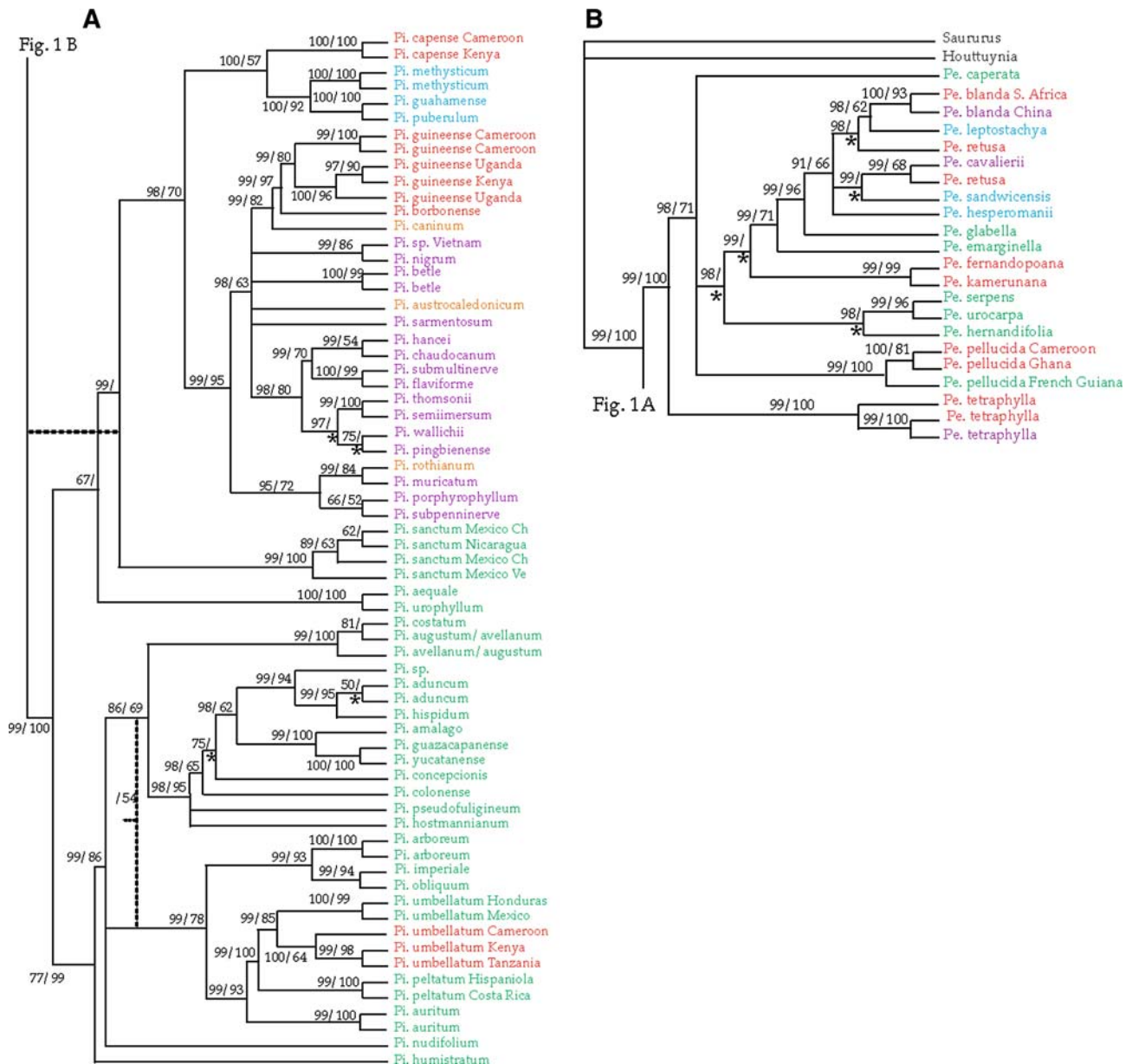
Fig. 1 a Majority rule tree based on BI using a different model for each of the two partitions (cpDNA and *g3pd*) with AIC showing the part of the tree that includes only *Piper*. *Dashed lines* represent relationships in the strict consensus of two MP trees of 8,381 steps each (CI = 0.53, RI = 0.80) with indels scored as missing data. *Numbers* above branches are PP values before the slash and BS values >50% after. *Asterisks* indicate clades that have PP < 50%, but are in agreement with the MP trees. Constraining all Neotropical species of *Piper* to a monophyletic clade cannot be rejected with SH tests. The slashes after *Pi. augustum* and *Pi. avellanum* reflect the switch of the position of these two species in the MP analysis. Species names are colored according to their native distribution: *red* Africa, *Blue* Pacific Island, *Purple* Asia, *Orange* Australia/New Caledonia, *Green* Neotropics. Pi., *Piper*; Ch, Chiapas; Ve, Veracruz. **b** Majority rule tree based on BI using a different model for each of the two partitions (cpDNA and *g3pd*) with AIC showing the part of the tree that includes only *Peperomia*. *Numbers* above branches are PP values before the slash and BS values >50% after. *Asterisks* indicate clades that have PP < 50%, but are in agreement with the MP trees. Species names are colored according to their native distribution: *red* Africa, *Blue* Pacific Island, *Green* Neotropics. Pe., *Peperomia*

topologies. When examined separately, each partition did not exhibit any area of incongruence that had BS > 75. Since hard incongruence was not detected (Seelanen et al. 1997), we combined data.

Maximum parsimony analysis with indels treated as missing data resulted in two equally parsimonious trees each

with 8,381 steps, CI = 0.53, RI = 0.80. The strict consensus of these trees is presented in Fig. 1. Rescoring indels resulted in eight most-parsimonious trees, each of 8,682 steps, CI = 0.53, RI = 0.49. The topologies of the trees were nearly identical, but differed among the relationships within some of the Asian clades and some Neotropical species.

Regardless of the scoring, MP resulted in both *Peperomia* and *Piper* as monophyletic. *Piper capense* is sister to a clade of otherwise exclusively Pacific Island species. *Piper guineense* is sister to *Pi. borbonense*, a species from the island of Reunion and sister to these two species is *Pi. caninum*, an Australian/Indonesian species. This clade lies within what would otherwise be a clade of exclusively Asian species with the exception of a second Australian species, *Pi. rothianum* which is in a clade of Malaysian species, and *Pi. austrocaledonicum* from New Caledonia. The Neotropical species are nearly monophyletic except that *Pi. sanctum*, a species from Mexico, is sister to all remaining species of *Piper*.

The BI analyses resulted in essentially the same topology regardless of the model used to infer the tree. The majority rule consensus of the trees from the analysis using a complex model based on AIC is presented in Fig. 1. Minor differences among analyses using LRT or a single model for all data produced a similar overall topology but differed slightly in posterior probability (PP) values, none of which was greater than 10 points different between analyses.

The BI and MP trees are largely in agreement with each other. Both *Peperomia* and *Piper* are monophyletic. Relationships among *Peperomia* species differ slightly in that the resolution among *Pe. retusa*, *Pe. cavalieri* and *Pe. sandwicensis* in BI is unresolved, as are the relationships among *Pe. fernandopoiana/Pe. kamerunana*, *Pe. pellucida*, *Pe. urocarpa/Pe. serpens*, and *Pe. hernandiifolia* and *Pe. retusa* to *Pe. blanda/Pe. leptostachya* (Fig. 1).

Within *Piper* it is again clear that the native African species are not monophyletic and their sister group relationships are identical between the MP and BI analyses (Fig. 1). The greatest number of differences between MP and BI topologies are the result of greater resolution within the Old World clade (Fig. 1). Relationships among species in the Neotropical clade are nearly identical between MP and BI trees, differing slightly in the placement of *Pi. concepcionis*, *Pi. colonense*, *Pi. pseudofuligineum*, *Pi. hostmannianum*, *Pi. augustum*, *Pi. avellanum*, and *Pi. nudifolium*. However, the greatest difference between the two topologies is in the placement of *Pi. sanctum*. This species is sister to all remaining species of *Piper* in the MP analysis, but is sister to all Old World species of *Piper* with BI. Likewise, the *Pi. aequale/Pi. urophyllum* clade which is sister to all remaining Neotropical *Piper* species in MP (excluding *Pi. sanctum*) is sister to the *Pi. sanctum*/Old World clade with BI (Fig. 1).

Both KH and SH tests indicated that constraining African *Piper* to a monophyletic clade (with placement corresponding either to the position of *Pi. capense* or *Pi. guineense* of Fig. 1) was significantly different ($p < 0.001$). Likewise constrained monophyly for Asian, Neotropical, Pacific, and African *Peperomia* each generated significantly longer trees ($p < 0.001$). Only two

Fig. 2 **a** Ultrametric tree using the topology of the MP analysis of ▶ Fig. 1a derived from PL placing the age of Piperaceae at 91.2 mya. Species names are colored according to their distribution and branches are colored according to consensus of models recreating distribution patterns: *red* Africa, *Blue* Pacific Island, *Purple* Asia, *Orange* Australia/New Caledonia, *Green* Neotropics. Where models disagree, *colored squares* above the lines and before the slash follow distribution patterns determined by AReA using the dispersal model with a vagility of 0.01, *colored squares* after the slash are based on the dispersal model with a vagility of 0.1 or the vicariance model (with two exceptions) with vagility of 0.1 or 0.01 (see text for a more detailed description of the models used), *triangles* reflect the two exceptions determined by the vicariance model that differed from the dispersal model, *circles* below the branches correspond to DIVA. Constraining all Neotropical species of *Piper* to a monophyletic clade cannot be rejected with SH tests and does not alter the results of AReA as they are presented here. Pi., *Piper*; Ch, Chiapas; Ve, Veracruz. **b** Ultrametric tree using the topology of the MP analysis of Fig. 1b derived from PL placing the age of Piperaceae at 91.2 mya. Species names are colored according to their distribution and branches are colored according to consensus of models recreating distribution patterns: *red* Africa, *Blue* Pacific Island, *Purple* Asia, *Green* Neotropics. Where models disagree, *colored squares* above the lines and before the slash follow distribution patterns determined by AReA using the dispersal model with a vagility of 0.01, *colored squares* after the slash are based on the dispersal model with a vagility of 0.1 or the vicariance model (with two exceptions) with vagility of 0.1 or 0.01 (see text for a more detailed description of the models used), *triangles* reflect the two exceptions determined by the vicariance model that differed from the dispersal model, *circles* below the branches correspond to DIVA. Pe., *Peperomia*

comparisons produced results that were not significant. These were the constraint of all Neotropical species of *Piper* ($p = 0.168$ and 0.161 for KH and SH, respectively) and a monophyletic *Pe. retusa* ($p = 0.153$ and 0.098 for KH and SH, respectively).

Irrespective of the number of regions or the tree that was used, DIVA ultimately indicated that the ancestor to either *Piper* or *Peperomia* was widespread (present in all designated regions) and required 18–19 (all included Piperaceae), 9–10 (*Piper* only), or 11–12 (*Peperomia* only) dispersal events to explain the extant sampled distribution. Constraining all Neotropical species to a single clade did not affect the results.

Regardless of whether *Piper*, *Peperomia*, or all of Piperaceae were set at a date of 91.2 mya, the results of r8s were similar. Cross validation gave an optimal smoothing value of 16 which was used for the dates presented here. Varying the smoothing value did not substantially change the dates obtained for nodes on the tree and ranged within five to ten mya from the values obtained with the value obtained from cross validation. Fixing the age of Piperaceae at 91.2 mya, the crown age of *Piper* was 71.75 mya and *Peperomia* was 88.89 mya. We reiterate that these dates are likely to be minimums since the crown age of Piperaceae is likely to be older than 91.2 mya, but by an unknown number of years.

A

B
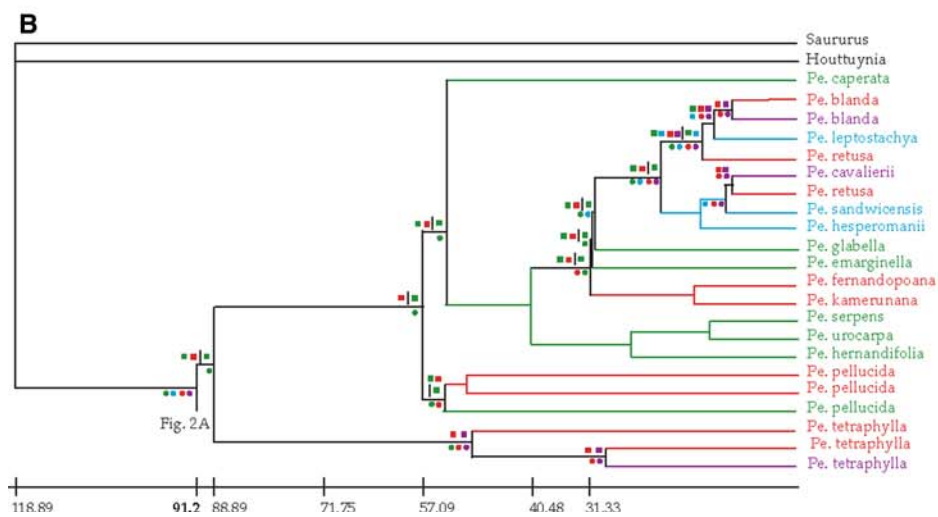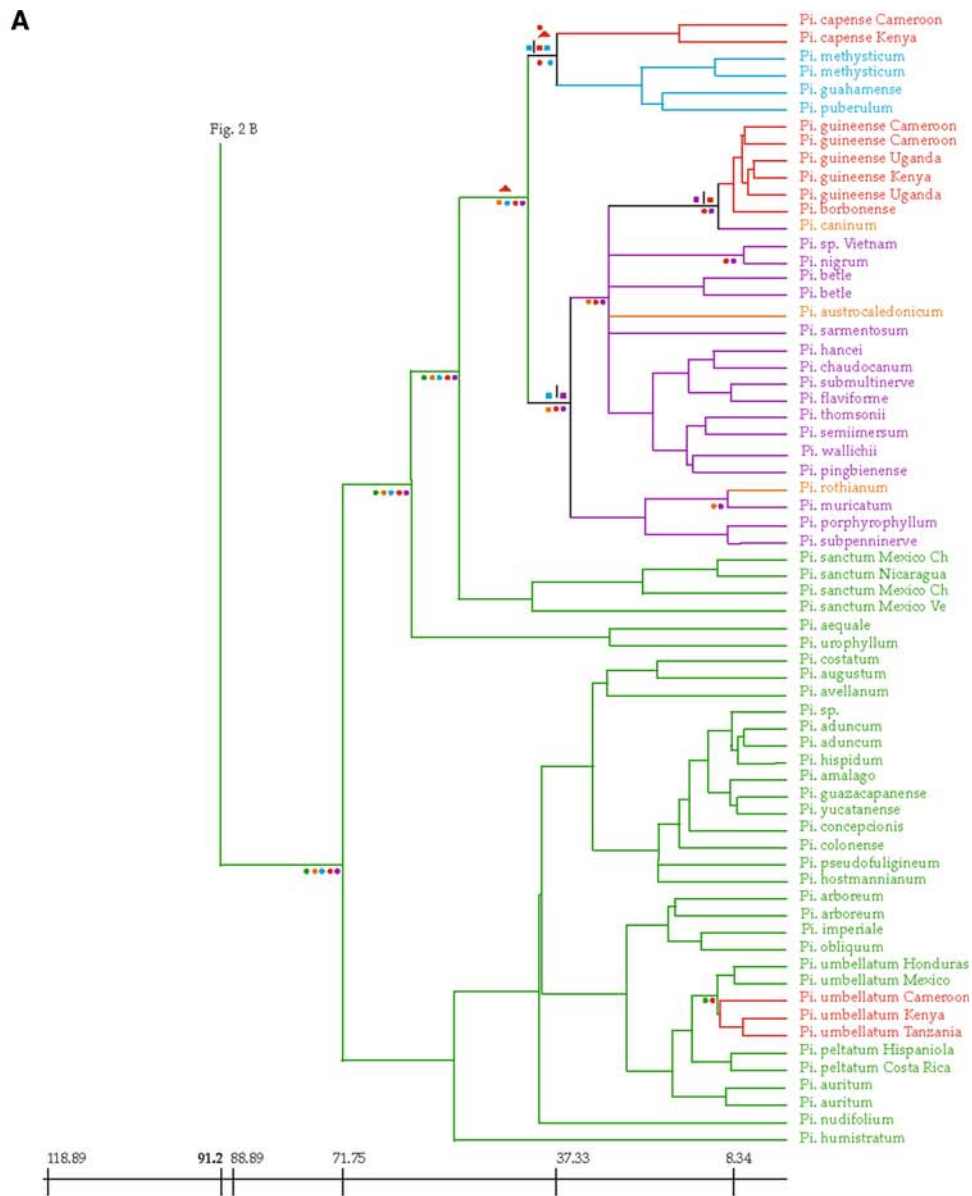
A comparison of the results of AReA using two different values of vagility indicated that the higher likelihood was obtained with the vicariant model and vagility at 0.01 (−ln = 595.8) although this did not differ greatly from the vicariant model with vagility at 0.1 (−ln = 628.2) or either of the dispersal models (−ln = 638.7, vagility = 0.01, −ln = 651.9, vagility = 0.1). The majority of distribution patterns within the tree were the same, however, some minor differences were apparent and these are also presented on Fig. 2. The majority of the differences were within *Peperomia* with the higher vagility rate (0.1) indicating a Neotropical origin for the genus with subsequent, multiple dispersal events to Africa, Asia, and the Pacific Islands, whereas the lower vagility value (0.01) implied a more widespread ancestor with a Neotropical and African origin, eventual extinction in Africa with subsequent dispersal back to Africa and multiple dispersal events to Asia and the Pacific Islands (Fig. 2). The vicariant models resulted in the same distribution regardless of the vagility value selected and were nearly in full agreement with the dispersal model with vagility at 0.1. In contrast to *Peperomia*, the biogeography for *Piper* was relatively consistent regardless of model selected with a Neotropical origin for the genus followed by either (1) dispersal to the Pacific Islands and further dispersals to Asia, Australia and Africa (dispersal model vagility = 0.01), (2) a dispersal to Asia followed by dispersals to Africa and Australia and a separate dispersal to Africa/Pacific Islands (dispersal model vagility at 0.1), or (3) dispersal to Africa with subsequent dispersal to Asia, Pacific Islands, Australia, and back migration to Africa (vicariant model) (Fig. 2). Constraining all Neotropical species of *Piper* to a single clade did not affect the results.

Discussion

Although we did not include representatives of *Zippelia*, *Verhuellia*, or *Manekia* in our analyses, we were able to demonstrate that the two largest genera of Piperaceae were each monophyletic and sister to each other in agreement with the results of all previous phylogenetic analyses of the family (Jaramillo and Manos 2001; Jaramillo et al. 2004; Wanke et al. 2007a, b; Jaramillo et al. in press.). Scoring of indels and different methods of data analysis provided some different results within Piperaceae, but in general these were only in areas of weak support. Regardless of how the data were analyzed it is clear that *Peperomia* and *Piper* have different biogeographic patterns (Fig. 2). This was not fully a surprise since previous phylogenetic analyses of *Piper* had indicated clear monophyletic groups that corresponded to major biogeographic regions (Jaramillo and Manos 2001; Jaramillo et al., in press), each of which

were also found here (Figs. 1, 2). *Peperomia* in contrast, has several species that are known to be widespread, therefore such a pattern was not expected. However, the monophyly of some of these widespread species had not yet been tested in a phylogenetic analysis.

An unusual result of this analysis is the placement of three Neotropical species: *Piper sanctum, Pi. aequale* and *Pi. urophyllum*. Bayesian analyses placed these three species as sister to the Old World clades of *Piper* and MP analyses rooted the tree with *Pi. sanctum* sister to all remaining species of *Piper* (Fig. 1). Although these are interesting results, it should be noted that KH and SH tests could not reject a monophyletic clade of all Neotropical species of *Piper* that included *Pi. sanctum, Pi. aequale*, and *Pi. urophyllum*. The placement of these three species may be the result of limited sampling. Inclusion of additional, and critical species may result in the placement of these species within, or at least sister to an exclusively Neotropical clade of *Piper*. A study that includes over 300 species and over 600 accessions of *Piper* provides evidence that *Pi. sanctum* is sister to *Pi. cinereum* C. DC. and this clade is sister to the remainder of Neotropical species excluding species in section *Schilleria*, few of which were included in the present analysis (Jaramillo et al. in press). The exclusion of *Pi. cinereum* and species of section *Schilleria* included in Jaramillo et al. (in press) is likely the best explanation for the unusual position of *Pi. sanctum* in the present study. However, since the impact of constraining all Neotropical species of *Piper* into a single clade did not affect the overall results of the biogeographic analyses, we will discuss our results using the trees obtained directly from the analyses of our data.

The biogeographic origin of *Piper*

Speciation within *Piper* has clearly followed a distinct biogeographic pattern with radiations occurring in three main biographic regions as previously outlined by Jaramillo and Manos (2001) and Jaramillo et al. (in press). These regions are Asia, the Pacific, and Neotropics with support for the sister group relationship of the Pacific and Asian clades. The results from AReA imply that *Piper* has an origin in the Neotropics with a dispersal to either the Pacific Islands, or the Pacific Islands/Africa and Asia using our dispersal based model (Fig. 2). An alternative perspective is presented with connections based on vicariance and land connections between the Neotropics and Africa. Under this model, *Piper* dispersed first from the Neotropics to Africa and then to the Pacific and Asia with subsequent dispersals from Asia to Australia and back to Africa (Fig. 2). DIVA analysis does not support these models and implies a widespread ancestor of *Piper* followed by later extinctions and dispersals (Fig. 2). However, DIVA does not allow for

any prior knowledge of histories of areas to be considered independently of the history of lineages (Ronquist 1996, 1997; Ree et al. 2005). In contrast, AReA makes use of prior assumptions assigned by the user related to biogeographic connections and the ages of clades to determine a distribution pattern. The dispersal model used in AReA allowed for a connection between the Neotropics and the Pacific Islands that was greater than that between the Neotropics and Asia but only prior to 60 mya. However, this time period pre-dates the greater diversification that occurred within *Piper* and *Peperomia* (Fig. 2). During the time period when lineage diversification was occurring, the dispersal model used in AReA allowed for all connections to be equal. The dispersal model optimized an initial dispersal event to the Pacific Islands when vagility is low (0.01) with subsequent dispersals from the Pacific Islands to Africa and Asia and further dispersal events from Asia to Africa, New Caledonia, and Australia (Fig. 2). With an increase in vagility (0.1), the dispersal model has a slightly different optimization and allows for a direct connection between the Neotropics and Asia, as well as a dispersal to both Africa and the Pacific Islands. Subsequent dispersals from Asia are nearly equivalent for either values of vagility under the dispersal model, the exception being an Asian ancestor for the *Pi. guineense/Pi. borbonense/Pi. caninum* clade with low vagility followed by dispersal to Africa for the ancestor to *Pi. borbonense* and *Pi. guineense*. With a higher vagility value, this same clade is proposed to be African in origin (albeit from within an Asian clade) and dispersal of *Pi. caninum* back to Australia/Asia since this species occurs in both Australia and Indonesia (Fig. 2). The vicariant model produced a nearly identical distribution pattern to the dispersal model with higher levels of vagility with two main exceptions: (1) the first clade outside of the Neotropics in *Piper* was African in origin, and (2) the dispersal to the Pacific was from Africa, not vice versa as implied with the dispersal model (Fig. 2). The stem and crown ages for the non-Neotropical clade are 53.0 and 41.86 mya, respectively. These age ranges just barely fit into the model where the connection between Africa and the Neotropics was still higher (0.75) than between all areas being equal and is the most likely explanation for these results. This connection is intriguing and the similarity between habit and breeding system of species of *Piper* in the Pacific, Neotropical species and *Pi. capense* argue for this closer connection. However, the low diversity of native *Piper* species in Africa argues against this. If the African origin of all non-Neotropical clades of *Piper* is correct then either widespread extinction or a reduction in the rate of speciation in Africa would be necessary to explain the depauperate *Piper* flora there. In light of this, it seems more likely that dispersal to Africa occurred independently from Asia and the Pacific, not from the Neotropics.

Piper biogeography: Asia/Pacific

It is yet to be fully resolved if the sister group relationship between the Asian and Pacific clades will be maintained, or if further sampling will reveal that one is derived from within the other. Studies with greater species sampling have maintained the sister group relationship between these clades (Jaramillo et al. in press). Pacific Islands are generally volcanic in origin and have a more recent origin relative to the Asian mainland. As a result, most studies that have examined plant groups with significant Pacific and mainland distributions have shown that the Pacific species are derived from mainland clades (Baldwin et al. 1990, 1991; Wright et al. 2000, 2001; Cronk et al. 2005).

From the data we have here, it appears that the Australian and New Caledonian species are also part of the Asian radiation. Australia has only 13 species of *Piper* (Australia Plant Names Index 2007) and we were only able to obtain material from two which appear as independent introductions (Fig. 2). We have only one of 16 species of Piperaceae from New Caledonia (Morat 1993), which also appears to be a separate introduction event from the Australian introductions and is clearly not a part of the Pacific clade. However, these conclusions should be regarded as preliminary until we have better sampling from these regions.

Piper biogeography: Africa

It is clear that the native African species are not the result of a single introduction. *Piper capense* is a part of the Pacific clade and represents an introduction into Africa (but see discussion above regarding the results of the vicariant model) independent of *Pi. guineense* (Figs.1–2). The latter species is sister to *Pi. borbonense*, a species from Reunion. The proximity of Reunion to the East African coast suggests that since the sister to these two species is *Pi. caninum*, an Indonesian/Australian species (our sample presumably is from Australia), that dispersal went from Asia to Australia to Reunion and Africa. However, the limited sampling of Asian species in our analysis precludes this conclusion. Further sampling of Asian species will be essential to resolve whether this pattern is retained in the future.

The phylogenetic analysis does resolve an interesting question regarding the habit, breeding system, and diversity of *Piper* in Africa. *Piper capense* is an erect shrub with hermaphroditic flowers, whereas *Pi. guineense* is a dioecious vine. Since the majority of Neotropical species are shrubs with hermaphroditic flowers and Asian species are dioecious vines, it had been considered a possibility that the two African species were, respectively, from the Neotropics and Asia (Jaramillo and Marquis 2004). If both species were from only one region, then a shift of habit

and/or breeding system occurred within Africa. These considerations, however, ignored the Pacific group which also are generally shrubs (Smith 1975). The data presented here clearly indicate separate introductions and it is likely that both species have retained characteristics of their parental clade with regard to habit and breeding system.

It has also been surprising that Africa is home to only two endemic species of *Piper* whereas the Neotropics are home to approximately two-thirds of the 1,000 species and Asia (including Australia and East Indian Islands) has about 300 species (Grieg 2004; Jaramillo and Marquis 2004). The much smaller land mass of the Pacific Islands is home to only about ten species all of which correspond to section *Macropiper* (Smith 1975; Jaramillo et al. in press). Explanations for the paucity of African species have invoked extinction that coincided with aridification of much of the African continent and loss of suitable *Piper* habitat (Axelrod 1972; Raven and Axelrod 1974; Axelrod and Raven 1978), or recent introductions. In light of the phylogenetic results, and the crown ages of *Pi. capense* and *Pi. guineense* at approximately 8 mya, it seems much more likely that the low species diversity of *Piper* in Africa is the result of a relatively recent arrival and the large species-level radiation that is seen in other areas where *Piper* is found has not had sufficient time to occur. This scenario has been invoked for African Rapateaceae and Bromeliaceae (Givnish et al. 2000, 2004). Givnish et al. (2004) conclude that, despite the similarity in habitats between *Maschalocephalus* and its closest relatives in South America and the fact that the two regions were likely adjacent prior to the opening of the Atlantic, the presence of Rapateaceae in Africa is the result of a recent (7.3 mya; Givnish et al. 2000, 2004) long distance dispersal event. Likewise, the presence of a single species of *Pitcairnia* (Bromeliaceae) is also proposed to be the result of a relatively recent (12 mya) long distance dispersal event (Givnish et al. 2004).

Multiple introductions into Africa are rare with other tropical groups that have a global distribution. Davis et al. (2002) found that the African acridocarpoid Malpighiaceae had a single origin from South America with a subsequent long distance dispersal into New Caledonia. Likewise Möller and Cronk (2001) found a single origin for all sampled species of African Gesneriaceae and have speculated that a single origin from Asia is likely the best explanation for the family in Africa. Conti et al. (2002) utilized phylogenetic relationships to show that three small African families previously placed within Crypteroniaceae (Oliniaceae, Penaeaceae, and Rhynchocalycaceae) formed a monophyletic group. In contrast, de Groot et al. (2006) have demonstrated up to five independent introductions into Africa for *Aristolochia*.

Piper umbellatum: Species of *Piper* that are not in cultivation are generally only found in a single biogeographic region (Asia, Africa, Pacific Islands, Neotropics). The one exception to this in our analyses is *Pi. umbellatum*. This study includes individuals from the Neotropics and Africa, but other studies have sampled individuals from Asia (Jaramillo and Manos 2001). In all studies, all individuals have formed a monophyletic group (Jaramillo and Manos 2001; Figs. 1, 2). The most common explanation for this widespread distribution has been recent human introductions. Transfer of plant material by humans is either intentional because the plant has utility, or accidental as a contaminant in other materials. The former likely explains the distribution of *Pi. nigrum*, *Pi. betle*, *Pi. methysticum*, and *Pi. auritum* which are economically important and widely cultivated in tropical regions throughout the world. *Piper umbellatum* is a weedy species typical of disturbed habitats, and its current distribution outside of the Neotropics has been considered to be the result of accidental introductions. However, this species also has medicinal uses in Cameroon (J. F. Smith, personal observation) and local botanists consider the species to be native (Chris Fouminyam, personal communication). The data presented here clearly show that the African individuals of *Pi. umbellatum* are monophyletic and nested within an otherwise exclusively Neotropical clade. However, the age of the African *Pi. umbellatum* clade is estimated at 10.88 mya which would preclude human transfer (Fig. 2). Thus *Pi. umbellatum* reflects a shift in dispersal ability within *Piper* that is more similar to that of *Peperomia* or an unusual dispersal event for this species. Placing dates on clades relies on smoothing the substitution rate from one branch to the next without allowing for major shifts in rate change (Sanderson 1997, 2002). A range of smoothing values never brought the date for the *Pi. umbellatum* clade within a range that could be attributed to human actions. However, the weedy habit of *Pi. umbellatum* may allow for a greater substitution rate as a result of small founding populations and a shorter generation time. As such, the greater divergence among the sequences of *Pi. umbellatum* included in this study may estimate the age of this clade to be older than it actually is.

Peperomia biogeography

Some species of *Peperomia* are known to have species distributed throughout the tropics. However, the monophyly of these widely distributed species had not yet been tested with rigorous molecular phylogenetic methods (Wanke et al. 2006). As it turns out, at least one species examined here is not monophyletic, *Pe. retusa*. An individual of this species from South Africa was sister to *Pe. cavalierii* from China. *Peperomia retusa* and *Pe. cavalierii* are considered close, and possibly may represent

subspecies rather than distinct species (Cheng et al. 1999). Another individual of *Pe. retusa* from South Africa was sister to the Asian accession of *Pe. blanda* and Pacific accession of *Pe. leptostachya* (Figs. 1, 2). However, KH and SH tests could not reject a monophyletic *Pe. retusa* and the results found here may simply reflect divergence of the sequences sampled among different populations. Alternatively, since a global revision of *Peperomia* has not been conducted recently, it may simply be that a more careful and diligent examination of morphological characters will reveal distinct differences among these populations that may merit their separation as distinct species. Similar studies of *Piper* have found that some of the widespread species may actually represent a number of distinct and different lineages as multiple populations from different regions have not resulted in monophyletic species in several instances (Jaramillo et al. in press).

In contrast to *Pe. retusa*, three additional species were included with samples from different biogeographic regions, (*Pe. blanda*, *Pe. pellucida*, and *Pe. tetraphylla*), each of which proved to be monophyletic with obvious dispersals to other regions following speciation (Fig. 2). As with *Pi. umbellatum* it had been speculated that some of these species may represent human-mediated introductions as they are generally found in disturbed habitats (*Pe. pellucida*). However, two of these species, with samples from either Africa and the Neotropics (*Pe. pellucida*) or from Africa and Asia (*Pe. tetraphylla*) have intraspecific divergences that are nearly as old as the majority of clades leading to different species within the genus (Fig. 2). The occurrence of several species with populations on different continents implies that *Peperomia* species are capable of long distance dispersal. The fact that the populations on the different continents are distinct at the molecular level and that the distinction can be dated to 49.41–53.75 mya for at least two of the species indicates that strong and similar selection pressures are maintaining morphological similarities of populations that are widely disjunct and have been disjunct for long periods. Alternatively, it is possible that the ages for *Peperomia* are distorted by the shorter generation time and presumably smaller founder populations of *Peperomia* in comparison to *Piper* which may affect rate heterogeneity. Likewise, it may be possible that these species have dispersed multiple times to different biogeographic regions and our sampling here by chance reflects some of the older dispersal events. Greater sampling may demonstrate more recent dispersal events and it is possible that gene flow among the newer dispersal events and established populations is maintaining morphological unity across continents. Coalescence of all genes, including the ones sampled here for phylogenetic analyses, simply has either yet to occur, or has coalesced to the copies from the earlier immigrants.

The sampling of *Peperomia* in this study was low, but sufficient to show that major biogeographic patterns within the genus are not as clear-cut as they are in *Piper*. Multiple introductions into Africa, Asia, and the Pacific Islands are necessary for either model used in AReA for *Peperomia* with an ancestor either in the Neotropics, or in the Neotropics and Africa (Fig. 2). DIVA indicates a widespread ancestor for *Peperomia* with subsequent extinctions and re-introductions to explain the current distribution of species (Fig. 2). As with AReA, DIVA requires multiple introductions into all geographic regions except the Neotropics. Further sampling of *Peperomia* species may yet reveal some patterns of biogeography.

It is likely that the smaller fruits of *Peperomia* allow for a greater ability to disperse than the larger, heavier fruits of *Piper*. All of the known fruit dispersal in *Piper* is carried out by bats primarily (Fleming 1988, 2004), and birds secondarily (Gorchov et al. 1993, 1995). In both cases, the fruits are ingested and, thus, are only carried over a relatively short distance before they are expelled. Furthermore, the fruits of most species of *Peperomia* are sticky and readily stick to the feet and feathers of birds (Ridley 1930). In fact, one of the longest documented instances of seed dispersal by birds is in *Peperomia* (Valdebenito et al. 1990). Not only does it appear that birds carried *Peperomia* seeds from southern South America to the Pacific Juan Fernandez Islands, 600 km from the coast of Chile, but then from the Juan Fernandez Islands to Tristan de Cunha Island in the South Atlantic; a voyage of over 5,000 km (Valdebenito et al. 1990). Numerous birds are known to fly among these island groups (Valdebenito et al. 1992).

An additional explanation is that the epiphytic habit of *Peperomia* allows for greater likelihood of establishment on different continents than would occur if the species were directly rooted in the soil. It may be that dispersal of *Piper* fruits is as likely as those of *Peperomia* and that competition restricts most species from becoming established. If competition among epiphytes were lower than that of terrestrial species, it may be possible that *Peperomia* could disperse and establish at a greater rate than *Piper* and thus the same species can be found on several continents. Lastly, it may be that the widespread species of *Peperomia* are more tolerant, or even require, more disturbed habitats than species of *Piper*. This is definitely true of *Pe. pellucida* which generally is found in weedy, disturbed habitats in many tropical parts of the world (S. Wanke, personal communication).

Divergence times

Ideally we would have studied divergence times within Piperaceae using the phylogenetic history of the family in

conjunction with multiple fossil calibration points. Unfortunately, Piperaceae have a poor fossil record with only a handful of recent fossils attributed to the family (Horn et al. 2003) and its sister family, Saururaceae (Smith and Stockey 2007). Rather than avoid estimating divergence times we opted to use the fossil record of *Lactoris* (Zavada and Benson 1987; MacPhail et al. 1999) as a starting point. Recent phylogenetic analyses have placed *Lactoris* within Piperales, and more specifically, part of Aristolochiaeae, the sister to Piperaceae/Saururaceae (Nickrent et al. 2002; Borsch et al. 2003; Jaramillo et al. 2004; Jaramillo and Kramer 2007; Wanke et al. 2007a, b). Thus the presence of *Lactoris* in the Turonian implies that at the minimum, its sister clade, Piperaceae/Saururaceae, must also have diverged at this point. It is possible that our dates underestimate the divergence points within Piperaceae since it may be that this clade diverged well before the origin of *Lactoris*: however, since we do not know when Saururaceae and Piperaceae diverged, it may be that our estimates are greater than what actually occurred. Until other fossils are discovered that can confirm the divergence points that are estimated here, these dates provide a middle ground on which we can start to form hypotheses for future testing.

It may be assumed that since Piperaceae are among some of the oldest lineages of flowering plants (Qiu et al. 1999; Soltis et al. 2000; Doyle and Endress 2000; Zanis et al. 2002; Jaramillo and Kramer 2007) that dispersals and radiations within the family would also be ancient. Our data indicate that with the stem ages of *Piper* and *Peperomia* defined at 91.2 mya, the crown ages for the two genera are still both in the Cretaceous at 71.75 and 88.89 mya, respectively (Fig. 2).

Despite the older crown ages for both *Piper* and *Peperomia*, most extant clades within these genera do not diverge until much later with major diversification of *Piper* into several Neotropical clades, a Pacific/*Pi. capense*, and a predominantly Asian clade at 37.33 mya (Fig. 2). Likewise, although the crown age of *Peperomia* is placed at 88.89 mya, the majority of diversification within the genus occurred between 57.09 and 31.33 mya (Fig. 2). Both of these results may be an artefact of limited species sampling, but broader phylogenetic analyses indicate that *Pi. sanctum* is one of the earliest divergent lineages within *Piper* and that other early divergent clades with the exception of section *Schilleria* are represented here as well (Jaramillo et al. in press). Likewise, sampling within *Peperomia* represents some of the earliest divergent clades (Wanke et al. 2006). Therefore, additional sampling may reveal branches of earlier dates, but it would be unlikely to discover many branches diverging prior to the ones discovered here.

Recent radiations and dispersals are not in conflict with biogeographic and phylogenetic studies of other groups

that have been able to estimate dates on radiations. Won and Renner (2006) found that although the fossil record indicates a diverse and widespread Cretaceous *Gnetum,* the current species' distributions are the result of divergence that occurred in the Upper Oligocene, Miocene, and Pliocene. Similar cases of relatively recent radiations in ancient lineages have been found for *Ephedra* (Huang and Price 2003: Ickert-Bond and Wojciechowski 2004) and *Equisetum* (Des Marais et al. 2003), as well as several angiosperm taxa with ancient and widely distributed fossil records such as Chloranthaceae (Zhang and Renner 2003; Eklund et al. 2004), Calycanthaceae (Zhou et al. 2006), *Illicium* (Morris et al. 2007), Anisophylleaceae (Zhang et al. 2007), and Nymphaeaceae (Yoo et al. 2005).

A pattern of an ancient stem lineage with a more recent crown age is not restricted to taxa with origins in the Cretaceous or earlier. Renner et al. (2001) proposed a stem age for Melastomataceae and Memecylaceae at 53 mya based on fossil data. This age marked a trichotomy in the tree among Memecylaceae, tribe Kibessieae of Melastomataceae and the remainder of Melastomataceae. Using fossils to establish two calibration points across the tree and assuming a clock, the crown ages for the three groups are 13.3–14.5, 9.2–10.0, and 45–49 mya, respectively. Although the crown age for the majority of the Melastomataceae seems relatively recent (4–8 mya after the stem origin), it should be emphasized that this is the point where major lineages within the family diverge and that species radiations within these lineages occur more recently (10–15 mya). Pennington et al. (2004) also found a pre-Pleistocene crown age (11–20) for a diversity of taxonomically unrelated groups that occur in seasonally dry tropical forests (*Coursetia*, *Poissonia*, *Ruprechtia*, *Loxopterygium* and *Chaetocalyx/Nissolia*).

The occurrence of old stem ages and recent crown ages implies long periods of stasis, high levels of extinction among earlier divergent lineages (creating "naked" stems), or a combination of both. Lack of fossils for Piperaceae prevents us from determining which of these hypotheses is more likely for this family. Either hypothesis is possible and data from different taxonomic groups support either hypothesis. Some groups have extensive fossil records and demonstrate what appears to be early extinct species arguing for extinctions that have produced long stem lineages (Melastomataceae: Renner et al. 2001, Juglandaceae: Manchester 1989, 1991; Manchester and Wheeler 1993).

In contrast, the amphi-Atlantic species *Symphonia globulifera* (Clusiaceae) can be dated using ITS sequence divergence to 28.52 mya, and the split between African and Neotropical clades is placed at 17.36 mya (Dick et al. 2003). Fossils of this species from Mesoamerica and South America can be dated to the Miocene. The dating of the African/American split, the retention of similar morphology between

populations on the two continents and the fossils found in the Americas imply that there has been at least a 15–17 my period of morphological stasis (Dick et al. 2003).

Long periods of stasis, followed by diversification may also be seen in more recent examples of migration such as invasive species (Mack et al. 2000), albeit over a much shorter time span. One current model for invasive species proposes that a species may become established in a new area, but remain in a quiescent stage for long periods before becoming invasive (Mack et al. 2000). The invasiveness may either be the result of recombination among genotypes introduced into the new area that were not sympatric in their native range (Ellstrand and Schierenbeck 2000; Petit et al. 2004; Williams et al. 2005), or evolution of new alleles that allow the species to move beyond its original introduction. Likewise we may envision the establishment of a single species in a new area that takes a long period before a mutation or recombination produces a genotype that allows for range expansion and subsequent speciation. This may explain the older stem and more recent crown ages seen for Piperaceae and other plant families.

A possible explanation for a long period of stasis in at least neotropical *Piper* is a limited amount of suitable habitat for the species to diversify into. In the Andes of South America numerous groups such as *Anthurium*, Gesneriaceae, *Miconia*, Campanulaceae, and *Piper* have high levels of diversity between 1,500 and 2,500 m in elevation. Prior to 10–15 mya, it is unlikely that the Andes had an extensive belt of elevations at this height (Lamb 2004) and thus, the cloud forest that contains high diversity in *Piper* was limited in its extent, and thus limited the diversity within the genus. More recent uplifts created a larger range of suitable habitats for *Piper* and correspond to the period in time where our phylogenetic and molecular dating indicates the greatest level of diversification occurred in the Neotropics (Fig. 2).

Quijano-Abril et al. (2006) have reasoned that *Piper* is not an Andean genus and that areas of diversity and endemism are centered in lower elevations with significant areas of diversity in the Caribbean and Amazonian regions. However, these areas of diversity and endemism do not preclude the Andean orogeny as a mechanism for increased diversification within *Piper*. The uplift of the Andes Mountains in the Miocene-Pliocene also had an enormous effect on speciation in many plant groups (Gentry 1982, 1989; Richardson et al. 2001; Hughes and Eastwood 2006; Ritz et al. 2007). The initial rise of the Andes as early as the Eocene produced localized habitat change, and isolated the lowland forests west of the orogeny from the much larger Amazonian forest (Gregory-Wodzicki 2000; Young et al. 2002). The orogeny also produced a highly dissected and constantly changing landscape, and generated new microclimatic niches through valleys of varying slope and

orientation, newly exposed substrates, rain shadows, and regular openings of new areas due to frequent landslides (Gentry 1982). Thus, even if *Piper* does not fit the model of an Andean genus, it is still likely that the emergence of the Andes had a direct influence on the speciation in this group.

Paleoclimate

The co-occurrence of species radiations among many taxonomically unrelated groups and lineages with different times of origin implies that there is something characteristic of the Tertiary that allowed for extensive species radiations. Episodic glaciations which result in both a cooler and drier climate in tropical regions have been proposed as an explanation for high levels of tropical diversity as areas of tropical wet and dry forests would contract and expand with each glaciation, respectively. Species surviving in refugia during each glaciation/interglaciation would result in a new wave of species radiation during each cycle (Prance 1982; Whitmore and Prance 1987; Behrensmeyer et al. 1992; Richardson et al. 2001; Dutech et al. 2003). However, the refuge hypothesis was based on Quaternary changes in the Pleistocene and dates for most plant groups place crown ages before the Pleistocene, at least in South America (some Central American plant groups have Pleistocene age crown groups; Pennington et al. 2004).

Early Eocene global temperatures reached a maximum allowing tropical forests to extend into high latitudes. Many plant families that are known to be restricted to tropical regions today were found in Eocene and Oligocene Laurasia (Manchester 1999; Mai 1995, Kubitzki and Krutzsch 1996). Diversity within the Neotropics reached a maximum at this point in time corresponding to either the shift in climate, or the amount of land encompassed by a suitable climate explaining the increase in diversity (Jaramillo et al. 2006). Paleoclimatic analyses indicate that dispersal to different regions was generally not limiting with regard to many factors such as light, temperature, moisture (Tiffney and Manchester 2001), or physical continuity such as the closing of the Tethys seaway (Hall 1998) and connections across the North Atlantic (Tiffney 1985a, b; Davis et al. 2002) at this time. Therefore, it is possible both *Piper* and *Peperomia* diverged in the Cretaceous, but did not radiate and disperse until the Eocene temperature maximum. Provided that the current physiological requirement of a tropical climate has been uniform in Piperaceae, subsequent cooling and episodic drying in the late Eocene to Miocene may have disrupted the further dispersal of these plants. Thus, barriers to dispersal and gene flow for the larger plants and fruits of *Piper* may have isolated these to their major biogeographic regions and only radiations within the tropical zones of the Americas,

Pacific Islands and Asia have occurred since, with dispersal to Africa and Australia occurring more recently.

Although some species of *Piper* and *Peperomia* are found in open areas, often at the edges of forest, the majority of species in Piperaceae occur in rain-forest understories (Grieg 2004). Applying physiological uniformitarionism to the species representing the ancestor of these genera, it would have been essential for tropical rain forests to also have existed at their time of origin in the late Cretaceous. Although much fossil evidence indicates that forests in the late Cretaceous were drier and more open than current rain forests (Tiffney 1984; Upchurch and Wolfe 1987, 1993, Wolfe and Upchurch 1987; Wing and Boucher 1998; Morley 2000; Johnson and Ellis 2002; Ziegler et al. 2003), molecular divergence time estimates for lineages of Malpighiales (major components of modern tropical rain forests) indicated that 28 major lineages were derived in tropical rain forests primarily between 112 and 94 mya (Davis et al. 2005). These data imply that tropical rain forests must also have been present at this time for the diversification of these lineages. The data for the origins of *Piper* and *Peperomia* presented here provide additional support that tropical forests existed during the Cretaceous.

References

Alice LA, Campbell CS (1999) Phylogeny of *Rubus* (Rosaceae) based on nuclear ribosomal DNA internal transcribed spacer region sequences. Amer J Bot 86:81–97

Alsos IG, Eidesen PB, Ehrich D, Skrede I, Westergaard K, Jacobsen FH, Landvik JY, Taberlet P, Brochmann C (2007) Frequent long-distance plant colonization in the changing arctic. Science 316:1606–1609

Aris-Brosou S, Yang Z (2002) Effects of models of rate evolution on estimation of divergence dates with special reference to the metazoan 18S rRNA phylogeny. Syst Biol 51:703–714

Australia Plant Names Index. http://www.anbg.gov.au/cpbr/databases/apni.html. Last visited October 2007

Axelrod DI (1972) Edaphic aridity as a factor in angiosperm evolution. Amer Naturalist 106:311–320

Axelrod DI, Raven PH (1978) Late Cretaceous and Tertiary vegetation history of Africa. In: Werger MJA (ed) Biogeography and ecology of southern Africa. Dr W. Junk bv Publishers, The Hague, pp 77–130

Baldwin BG, Robichaux RH (1995) Historical biogeography and ecology of the Hawaiian silversword alliance (Asteraceae): new molecular phylogenetic perspectives. In: Wagner WL, Funk VA (eds) Hawaiian biogeography: evolution on a hot spot archipelago. Smithsonian Institution Press, Washington, DC, pp 259–189

Baldwin BG, Kyhos DW, Dvorak J (1990) Chloroplast DNA variation and adaptive radiation in the Hawaiian silversword alliance (Asteraceae- Madiinae). Ann Missouri Bot Gard 77:96–109

Baldwin BG, Kyhos DW, Dvorak J, Carr GD (1991) Chloroplast DNA evidence for a North American origin of the Hawaiian silversword alliance (Asteraceae). Proc Natl Acad Sci USA 88:1840–1843

Behrensmeyer AK, Damuth J, DiMichele W, Potts R, Sues HD, Wing S (1992) Terrestrial ecosystems through time. University of Chicago Press, Chicago

Bell CD (2007) Phylogenetic placement and biogeography of the North American species of *Valerianella* (Valerianaceae: Dipsacales) based on chloroplast and nuclear DNA. Molec Phylogenet Evol 44:929–941

Bell CD, Donoghue MJ (2005) Phylogeny and biogeography of Valerianaceae (Dipsacales) with special reference to the South American valerians. Organisms. Divers Evol 5:147–159

Borsch T, Hilu KW, Quandt D, Wilde V, Neinhuis C, Barthlott W (2003) Noncoding plastid *trn*T-*trn*F sequences reveal a well resolved phylogeny of basal angiosperms. J Evol Biol 16:558–576

Borsch T, Löhne C, Müller K, Wanke S, Worberg A, Barthlott W, Neinhus C, Hilu KW, Quandt D (2005) Towards understanding basal angiosperm diversification: recent insights using fast evolving genomic regions. Nova Acta Leopoldina 342:85–110

Carine MA, Russell SJ, Santos-Guerra A, Francisco-Ortega J (2004) Relationships of the Macaronesian and Mediterranean floras: molecular evidence for multiple colonizations into Macaronesia and back-colonization of the continent in *Convolvulus* (Convolvulaceae). Amer J Bot 91:1070–1085

Cheng Y-Q, Xia N-H, Gilbert MG (1999) Piperaceae. In: Wu Z-Y, Raven PH (eds) Flora of China, Cycadaceae through Fagaceae. Missouri Botanical Garden Press. St. Louis, Missouri, USA, pp 110–129

Chew W-L (1972) The genus *Piper* (Piperaceae) in New Guinea, Solomon Islands, and Australia, 1. J Arnold Arbor 53:1–25

Conti E, Eriksson T, Schönenberger J, Sytsma KJ, Baum DA (2002) Early Tertiary out-of-India dispersal of Crypteroniaceae: evidence from phylogeny and molecular dating. Evolution 56:1931–1942

Cox CB, Moore PD (2000) Biogeography. An ecological and evolutionary approach, 6th edn. Blackwell, London

Crisp MD, Cook LG (2007) A congruent molecular signature of vicariance across multiple plant lineages. Molec Phylogenet Evol 43:1106–1117

Cronk QCB, Kiehn M, Wagner WL, Smith JF (2005) Evolution of *Cyrtandra* (Gesneriaceae) in the Pacific Ocean: the origin of a supertramp clade. Amer J Bot 92:1017–1024

Darwin C (1859) On the origin of species by means of natural selection, or the preservation of favoured races in the struggle for life. John Murray, London

Davis CC, Bell CD, Fritsch PW, Mathews S (2002) Phylogeny of *Acridocarpus-Brachylon* (Malpighiaceae): implications for

Tertiary tropical floras and Afroasian biogeography. Evolution 56:2395–2405

Davis CC, Webb CO, Wurdack KJ, Jaramillo CA, Donoghue MJ (2005) Explosive radiation of Malpighiales supports a mid-Cretaceous origin of modern tropical rain forests. Amer Naturalist 165:E36–E65

de Groot H, Wanke S, Neinhuis C (2006) Revision of the genus *Aristolochia* (Aristolochiaceae) in Africa, Madagascar and adjacent islands. Bot J Linn Soc 151:219–238

DeJoode DR, Wendel JF (1992) Genetic diversity and origin of the Hawaiian islands cotton, *Gossypium tomentosum*. Amer J Bot 79:1311–1319

de Queiroz A (2005) The resurrection of oceanic dispersal in historical biogeography. Trends Ecol Evol 20:68–73

Des Marais DL, Smith AR, Britton DM, Pryer KM (2003) Phylogenetic relationships and evolution of extant horsetails, *Equisetum*, based on chloroplast DNA sequence data (*rbc*L and *trn*L-F). Int J Pl Sci 164:737–751

Dick CW, Abdul-Salim K, Bermingham E (2003) Molecular systematic analysis reveals cryptic Tertiary diversification of a widespread tropical rain forest tree. Amer Naturalist 162:691–703

Doyle JA, Endress PK (2000) Morphological phylogenetic analysis of basal angiosperms: comparison and combination with molecular data. Int J Pl Sci 161:S121–S151

Doyle JA, Endress PK (2007) Integrating early Cretaceous fossils into the phylogeny of recent angiosperms. Plant Biology and Botany 2007, p 150 (abstract)

Dutech C, Maggia L, Tardy C, Joly HI, Jarne P (2003) Tracking a genetic signal of extinction-recolonization events in a neotropical tree species: *Vouacapoua americana* Aublet in French Guiana. Evolution 57:2753–2764

Drummond A. J., Rambaut A. (2005) BEAST v 1.3. http://evolve.zoo.ox.ac.uk/beast/

Eklund H, Doyle JA, Herendeen PS (2004) Morphological phylogenetic analysis of living and fossil Chloranthaceae. Int J Pl Sci 165:107–151

Ellstrand NC, Schierenbeck KA (2000) Hybridization as a stimulus for the evolution of invasiveness in plants? Proc Natl Acad Sci USA 97:7043–7050

Farris SJ, Källersjö M, Kluge AG, Bult C (1994) Testing significance of incongruence. Cladistics 10:315–319

Felsenstein J (1985) Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39:783–791

Fleming T (1988) The short-tailed fruit bat: a study in plant-animal interactions. University of Chicago Press, Chicago

Fleming T (2004) Dispersal ecology of Neotropical *Piper* shrubs and treelets. In: Dyer LA, Palmer ADN (eds) *Piper*: A model genus for studies of Phytochemistry, Ecology, and Evolution. Kluwer, New York, pp 58–77

Friis EM, Pedersen KR, Crane PR (1995) *Appomattoxia ancistrophora* gen. et sp. nov., a new early Cretaceous plant with similarities to *Circaeaster* and extant Magnoliidae. Amer J Bot 82:933–943

Ganders FR, Berbee M, Pirseyedi M (2000) ITS based sequence phylogeny in *Bidens* (Asteraceae): evidence for the continental relatives of Hawaiian and Marquesan *Bidens*. Syst Bot 25:122–133

Gentry AH (1982) Neotropical floristic diversity: phytogeographical connections between Central and South America, Pleistocene climatic fluctuations, or an accident of the Andean orogeny? Ann Missouri Bot Gard 69:557–593

Gentry AH (1989) Speciation in tropical forests. In: Holm-Nielsen LB, Nielsen IC, Balslev H (eds) Tropical forests: botanical dynamics, speciation and diversity. Academic Press, London, pp 113–134

Givnish TJ, Sytsma KJ, Hahn WJ, Smith JF (1995) Molecular evolution, adaptive radiation, and geographic speciation in *Cyanea* (Campanulaceae, Lobelioideae). In: Wagner WL, Funk VA (eds) Hawaiian biogeography: evolution on a hot spot archipelago. Smithsonian Institution Press, Washington, DC, pp 288–337

Givnish TJ, Knox E, Patterson TB, Hapeman JR, Palmer JD, Sytsma KJ (1996) The Hawaiian lobelioids are monophyletic and underwent a rapid initial radiation roughly 15 million years ago. Amer J Bot 83:159 (abstract)

Givnish TJ, Evans TM, Zjhra ML, Patterson TB, Berry PE, Sytsma KJ (2000) Molecular evolution, adaptive radiation, and geographic diversification in the amphiatlantic family Rapateaceae: evidence from *ndh*F sequences and morphology. Evolution 54:1915–1937

Givnish TJ, Millam KC, Evans TM, Hall JC, Pires JC, Berry PE, Sytsma KJ (2004) Ancient vicariance or recent long-distance dispersal? Inferences about phylogeny and South American-African disjunctions in Rapateaceae and Bromeliaceae based on *ndh*F sequence data. Int J Pl Sci 165:S35–S54

Goldman N, Anderson JP, Rodrigo AG (2000) Likelihood-based tests in phylogenetics. Syst Biol 49:652–670

Gorchov DL, Cornejo F, Ascorra CF, Jaramillo M (1993) The role of seed dispersal in the natural regeneration of rain forest after strip-cutting in the Peruvian Amazon. Pl Ecol 107–108:339–349

Gorchov DL, Cornejo F, Ascorra CF, Jaramillo M (1995) Dietary overlap between frugivorous birds and bats in the Peruvian Amazon. Oikos 74:235–250

Gregory-Wodzicki KM (2000) Uplift history of the central and northern Andes: a review. GSA Bull 112:1091–1105

Grieg N (2004) Introduction. In: Dyer LA, Palmer ADN (eds) *Piper*: A model genus for studies of Phytochemistry, Ecology, and Evolution. Kluwer, New York, pp 1–4

Hall R (1998) The plate tectonics of Cenozoic Southeast Asia and the distribution of land and sea. In: Hall R, Holloway JD (eds) Biogeography and geological evolution of Southeast Asia. Backhuys Publishers, Leiden, The Netherlands, pp 99–131

Hasebe M, Ando T, Iwatsuki K (1998) Intrageneric relationships of maple trees based on the chloroplast DNA restriction fragment length polymorphisms. J Pl Res 111:441–451

Helfgott DM, Francisco-Ortega J, Santos-Guerra A, Jansen RK, Simpson BB (2000) Biogeography and breeding system evolution of the woody *Bencomia* alliance (Rosaceae) in Macaronesia based on ITS sequence data. Syst Bot 25:82–97

Howarth DG, Gardner DE, Morden CW (1997) Phylogeny of *Rubus* subgenus *Ideobatus* (Rosaceae) and its implications toward colonization of the Hawaiian islands. Syst Bot 22:433–441

Howarth DG, Gustafsson MHG, Baum DA, Motley TJ (2003) Phylogenetics of the genus *Scaevola* (Goodeniaceae): implication for dispersal patterns across the Pacific Basin and colonization of the Hawaiian islands. Amer J Bot 90:915–923

Horn SP, Sanford RL, Dilcher D, Lott TA, Renne PR, Wiemann MC, Cozadd D, Vargas O (2003) Pleistocene plant fossils in and near La Selva Biological Station, Costa Rica. Biotropica 35:434–441

Huang J, Price RA (2003) Estimation of the age of extant *Ephedra* using chloroplast *rbc*L sequence data. Molec Biol Evol 20:435–440

Huelsenbeck JP, Ronquist F (2001) MRBAYES: Bayesian inference of phylogeny. Bioinformatics 17:754–755

Hughes C, Eastwood R (2006) Island radiations on a continental scale: exceptional rates of plant diversification after uplift of the Andes. Proc Natl Acad Sci USA 103:10334–10339

Humphries CJ, Parenti LR (1999) Cladistic biogeography. Oxford University Press, Oxford, UK

Ickert-Bond SM, Wojciechowski MF (2004) Phylogenetic relationships in *Ephedra* (Gnetales): evidence from nuclear and chloroplast DNA sequence data. Syst Bot 29:834–849

Jaramillo C, Rueda MJ, Mora G (2006) Cenozoic plant diversity in the Neotropics. Science 311:1893–1896

Jaramillo MA, Manos PS (2001) Phylogeny and patterns of floral diversity in the genus *Piper* (Piperaceae). Amer J Bot 88:706–716

Jaramillo MA, Marquis R (2004) Future research in *Piper* biology. In: Dyer LA, Palmer ADN (eds) *Piper*: A model genus for studies of Phytochemistry, Ecology, and Evolution. Kluwer, New York, pp 199–203

Jaramillo MA, Kramer EM (2007) Molecular evolution of the petal and stamen identity genes, *APETALA3* and *PISTILLATA*, after petal loss in the Piperales. Molec Phylogenet Evol 44:598–609

Jaramillo MA, Manos PS, Zimmer EA (2004) Phylogenetic relationships of the perianthless Piperales: reconstructing the evolution of floral development. Int J Pl Sci 165:403–416

Jaramillo MA, Callejas R, Davidson C, Smith JF, Stevens AC, Tepe EJ (in press) A phylogeny of the tropical genus *Piper* (Piperaceae) using ITS and the chloroplast intron *psbJ-petA*. Syst Bot

Johnson KR, Ellis B (2002) A tropical rainforest in Colorado 1.4 million years after the Cretaceous-Tertiary boundary. Science 296:2379–2383

Kim Y-D, Jansen RK (1998) Chloroplast DNA restriction site variation and phylogeny of the Berberidaceae. Amer J Bot 85:1766–1778

Kim Y-D, Kim S-H (1999) Phylogeny of *Weigela* and *Diervilla* (Caprifoliaceae) based on nuclear rDNA ITS sequences: biogeographic and taxonomic implications. J Pl Res 112:331–341

Kishino H, Hasegawa M (1989) Evaluation of the maximum likelihood estimate of the evolutionary tree topologies from DNA sequence data, and the branching order in Hominoidea. J Molec Evol 29:170–179

Kishino H, Thorne JL, Bruno WJ (2001) Performance of a divergence time estimation method under a probabilistic model of rate evolution. Molec Biol Evol 18:352–361

Knox EB, Downie SR, Palmer JD (1993) Chloroplast genome rearrangements and the evolution of giant lobelias from herbaceous ancestors. Molec Biol Evol 10:414–430

Kubitzki K, Krutzsch W (1996) Origins of East and South Asian plant diversity. In: Auluo Z, Sugong W (eds) Floristic characteristics and diversity of East Asian plants. Springer, Berlin, pp 65–70

Lamb S (2004) Devil in the mountain: a search for the origin of the Andes. Princeton University Press, Princeton, NJ

Lindqvist C, Albert VA (2002) Origin of the Hawaiian endemic mints within North American *Stachys* (Lamiaceae). Amer J Bot 89:1709–1724

Lee NS, Sang T, Crawford DJ, Yeau SH, Kim S-C (1996) Molecular divergence between disjunct taxa in eastern Asia and eastern North America. Amer J Bot 83:1373–1378

Mack RN, Simberloff D, Lonsdale WM (2000) Biotic invasions: causes, epidemiology, global consequences, and control. Ecol Appl 10:689–710

MacPhail MK, Partridge AD, Truswell EM (1999) Fossil pollen records of the problematical primitive angiosperm family Lactoridaceae in Australia. Pl Syst Evol 214:199–210

Magallón S, Sanderson MJ (2001) Absolute diversification rates in angiosperm clades. Evolution 55:1762–1780

Mai DH (1995) Tertiäre Vegetationsgeschichte Europas. Fischer, Jena

Manchester SR (1989) Early history of the Juglandaceae. Pl Syst Evol 162:231–250

Manchester SR (1991) *Cruciptera*, a new Juglandaceous winged fruit from the Eocene and Oligocene of western North America. Syst Bot 16:715–725

Manchester SR (1999) Biogeographical relationships of North American Tertiary floras. Ann Missouri Bot Garden 86:472–522

Manchester SR, Wheeler EA (1993) Extinct juglandaceous wood from the Eocene of Oregon and its implications for xylem evolution in the Juglandaceae. IAWA J 14:103–111

Marquis RJ (2004) Biogeography of Neotropical *Piper*. In: Dyer LA, Palmer ADN (eds) *Piper*: A model genus for studies of Phytochemistry, Ecology, and Evolution. Kluwer, New York, pp 78–96

McGlone MS (2005) Goodbye Gondwana. J Biogeography 32:739–740

Miquel F. A. (1843–1844) *Systema Piperacearum*. H. A. Kramer, Rotterdam.

Möller M, Cronk QCB (2001) Phylogenetic studies in *Streptocarpus* (Gesneriaceae): reconstruction of biogeographic history and distribution patterns. Syst Geogr Plants 71:545–555

Morat P (1993) Our knowledge of the flora of New Caledonia: endemism and diversity in relation to vegetation types and substrates. Biodivers Lett 1:72–81

Morley RJ (2000) Origin and evolution of tropical rain forests. Wiley, New York

Morris AB, Bell CD, Clayton JW, Judd WS, Soltis DE, Soltis PS (2007) Phylogeny and divergence time estimation in *Illicium* with implications for New World biogeography. Syst Bot 32:236–249

Nelson G, Platnick NI (1981) Systematics and biogeography: cladistics and vicariance. Columbia University Press, New York

Neinhuis C, Wanke S, Hilu KW, Müller K, Borsch T (2005) Phylogeny of Aristolochiaceae based on parsimony, likelihood, and Bayesian analyses of *trnL-trn*F sequences. Pl Syst Evol 250:7–26

Nepokroeff M, Sytsma KJ, Wagner WL, Zimmer EA (2003) Reconstructing ancestral patterns of colonization and dispersal in the Hawaiian understory tree genus *Psychotria* (Rubiaceae): a comparison of parsimony and likelihood approaches. Syst Biol 52:820–838

Nickrent D, L, Blarer A, Qiu Y-L, Soltis DE, Soltis PS, Zanis M (2002) Molecular data place Hydnoraceae with Aristolochiaceae. Amer J Bot 89:1809–1817

Olmstead RG, Palmer JD (1994) Chloroplast DNA systematics: a review of methods and data analysis. Amer J Bot 81:1205–1224

Pennington RT, Lavin M, Prado DE, Pendry CA, Pell SK, Butterworth CA (2004) Historical climate change and speciation: neotropical seasonally dry forest plants show patterns of both Tertiary and Quaternary diversification. Phil Trans Royal Soc London. Series B 359:515–537

Petit RJ, Bialozyt R, Garnier-Géré P, Hampe A (2004) Ecology and genetics of tree invasions: from recent introductions to Quaternary migrations. Forest Ecol Management 197:117–137

Posada D, Buckley TR (2004) Model selection and model averaging in phylogenetics: Advantages of Akaike information criterion and Bayesian approaches over likelihood ratio tests. Syst Biol 53:793–808

Posada D, Crandall KA (1998) Modeltest: testing the model of DNA substitution. Bioinformatics 14:817–818

Poux C, Madsen O, Marquard E, Vieites DR, Jong WWD, Vences M (2005) Asynchronous colonization of Madagascar by the four endemic clades of primates, tenrecs, carnivores, and rodents as inferred from nuclear genes. Syst Biol 54:719–730

Prance GT (1982) Biological diversification in the tropics. Columbia University Press, New York

Qiu Y-L, Parks CR, Chase MW (1995) Molecular divergence in eastern Asian-eastern North American disjunct section *Rytidospermum* of *Magnolia* (Magnoliaceae). Amer J Bot 82:1589–1598

Qiu Y-L, Lee J, Bernasconi-Quadroni F, Soltis DE, Soltis PS, Zanis M, Zimmer EA, Chen Z, Savolainen V, Chase MW (1999) The earliest angiosperms: evidence from mitochondrial, plastid, and nuclear genomes. Nature 402:404–407

Quijano-Abril MA, Callejas-Posada R, Miranda-Esquivel DR (2006) Areas of endemism and distribution patterns for Neotropical *Piper* species (Piperaceae). J Biogeogr 33:1266–1278

Rambaut A, Bromham L (1998) Estimating divergence dates from molecular sequences. Molec Biol Evol 15:442–448

Raven PH, Axelrod DI (1974) Angiosperm biogeography and past continental movements. Ann Missouri Bot Gard 61:539–673

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

Reeves G, Chase MW, Goldblatt P, Fay MF, Cox AV, LeJeune B, Souza-Chies T (2001) Molecular systematics of Iridaceae: evidence from four plastid DNA regions. Amer J Bot 88:2074–2087

Renner SS (2004) Plant dispersal across the tropical Atlantic by wind and sea currents. Int J Pl Sci 165:S23–S33

Renner SS, Clausing G, Meyer K (2001) Historical biogeography of Melastomataceae: the roles of Tertiary migration and long-distance dispersal. Amer J Bot 88:1290–1300

Richardson JE, Pennington RT, Pennington TD, Hollingsworth PM (2001) Rapid diversification of a species-rich genus of Neotropical rain forest trees. Science 293:2242–2245

Ridley H (1930) The dispersal of plants throughout the world. Ashford, Reeve

Ritz CM, Martins L, Mecklenberg R, Goremykin V, Hellwig FH (2007) The molecular phylogeny of *Rebutia* (Cactaceae) and its allies demonstrates the influence of paleogeography on the evolution of South American mountain cacti. Amer J Bot 94:1321–1332

Ronquist F. (1996) DIVA. Ver. 1.1 Computer program and manual. Available by anonymous FTP (ftp.sysbot.uu.se) from Uppsala University, Uppsala, Sweden

Ronquist F (1997) Dispersal-vicariance analysis: a new approach to the quantification of historical biogeography. Syst Biol 45:195–203

Sanderson MJ (1997) A nonparametric approach to estimating divergence times in the absence of rate constancy. Molec Biol Evol 14:1218–1231

Sanderson MJ (2002) Estimating absolute rates of molecular evolution and divergence times: a penalized likelihood approach. Molec Biol Evol 19:101–109

Sanderson MJ (2003) r8s: inferring absolute rates of molecular evolution and divergence times in the absence of a molecular clock. Bioinformatics 19:301–302

Sanmartín I, Ronquist F (2004) Southern hemisphere biogeography inferred by event-based models: plant versus animal patterns. Syst Biol 53:216–243

Särkinen TE, Newman MF, Maas PJM, Maas H, Poulsen AD, Harris DJ, Richardson JE, Clark A, Hollingsworth M, Pennington RT (2007) Recent oceanic long-distance dispersal and divergence in the amphi-Atlantic rain forest genus *Renealmia* L. f. (Zingiberaceae). Molec Phylogenet Evol 44:968–980

Schnabel A, Wendel JF (1998) Cladistic biogeography of *Gleditsia* (Leguminosae) based on *ndh*F and *rpl*16 chloroplast gene sequences. Amer J Bot 85:1753–1765

Seelanen T, Schnabel A, Wendel JF (1997) Congruence and consensus in the cotton tribe (Malvaceae). Syst Bot 22:259–290

Shimodaira H, Hasegawa M (1999) Multiple comparisons of log-likelihoods with applications to phylogenetic inference. Molec Biol Evol 16:1114–1116

Simmons MP, Ochoterena H (2000) Gaps as characters in sequence-based phylogenetic analyses. Syst Biol 49:369–381

Smith SA (1975) The genus *Macropiper* (Piperaceae). Biol J Linn Soc 71:1–38

Smith S. A. (2006) AReA: Ancestral range analysis v. 2.1. http://blackrim.org/programs/area.html

Smith SY, Stockey RA (2007) Establishing a fossil record for the perianthless Piperales: *Saururus tuckerae* sp. nov. (Saururaceae) from the Middle Eocene Princeton Chert. Amer J Bot 94:1642–1657

Smith JF, Funke MM, Woo VL (2006) A duplication of g*cyc* predates divergence within tribe Coronanthereae (Gesneriaceae): phylogenetic analysis and evolution. Pl Syst Evol 261:245–256

Smith JF, Hileman LC, Powell M, Baum DA (2004) Evolution of g*cyc*, a Gesneriaceae homolog of *CYCLOIDEA*, within subfamily Gesnerioideae (Gesneriaceae). Molec Phylogenet Evol 31:765–779

Soltis DE, Soltis PS, Chase MW, Mort ME, Albach DC, Zanis M, Savolainen V, Hahn WH, Hoot SB, Fay MF, Axtell M, Swensen SM, Prince LM, Kress WJ, Nixon KC, Farris JS (2000) Angiosperm phylogeny inferred from 18S rDNA, *rbc*L, and *atp*B sequences. Bot J Linn Soc 133:381–461

Strand AE, Leebens-Mack J, Milligan BG (1997) Nuclear DNA-based markers for plant evolutionary biology. Molec Ecol 6:113–118

Stuessy TF, Crawford DJ, Anderson GJ, Jensen RJ (1998a) Systematics, biogeography and conservation of Lactoridaceae Pers. Pl Ecol Evol Syst 1:267–290

Stuessy TF, Crawford DJ, Marticorena C, Rodriguez R (1998b) Island biogeography of angiosperms of the Juan Fernandez archipelago. In: Stuessy TF, Ono M (eds) Evolution and speciation of island plants. Cambridge University Press, Cambridge UK, pp 121–138

Swenson U, Bremer K (1997) Pacific biogeography of the Asteraceae genus *Abrotanella* (Senecioneae, Blennospermatinae). Syst Bot 22:493–508

Swenson U, Hill RS, McLoughlin S (2001) Biogeography of *Nothofagus* supports the sequence of Gondwana break-up. Taxon 50:1025–1041

Swofford D. L. (2002) PAUP* Phylogenetic analysis using parsimony (* and Other Methods). Version 10. Sinauer Associates, Sunderland, Massachusetts

Thorne JL, Kishino H (2002) Divergence time estimation and rate evolution with multilocus data sets. Syst Biol 51:689–702

Thorne JL, Kishino H, Painter IS (1998) Estimating the rate of evolution of the rate of molecular evolution. Molec Biol Evol 15:1647–1657

Tiffney BH (1984) Seed size, dispersal syndromes, and the rise of the angiosperms: evidence and hypothesis. Ann Missouri Bot Gard 71:551–576

Tiffney BH (1985a) Perspectives on the origin of the floristic similarity between eastern Asia and eastern North America. J Arnold Arbor 66:73–94

Tiffney BH (1985b) The Eocene North Atlantic land bridge: its importance in Tertiary and modern phytogeography of the Northern hemisphere. J Arnold Arbor 66:243–273

Tiffney BH, Manchester SR (2001) The use of geological and paleontological evidence in evaluating plant phylogenetic hypotheses in the Northern hemisphere Tertiary. Int J Pl Sci 162:S3–S17

Upchurch GR, Wolfe JA (1987) Mid-Cretaceous to early Tertiary vegetation and climate: evidence from fossil leaves and wood. In: Friis EM, Chaloner WG, Crane PR (eds) The origins of angiosperms and their biological consequences. Cambridge University Press, Cambridge, UK, pp 75–105

Upchurch GR, Wolfe JA (1993) Cretaceous vegetation of the western interior and adjacent regions of North America. In: Kauffman EG, Caldwell WGE (eds) Cretaceous evolution of the western interior basin. Geological Association of Canada, Special Paper, vol 39, pp 243–281

Valdebenito HA, Stuessy TF, Crawford DJ (1990) A new biogeographic connection between islands in the Atlantic and Pacific oceans. Nature 347:549–550

Valdebenito HA, Stuessy TF, Crawford DJ, Silva OM (1992) Evolution of *Peperomia* (Piperaceae) in the Juan Fernandez Islands, Chile. Pl Syst Evol 182:107–119

Wagner WL, Funk VA (1995) Hawaiian biogeography: evolution on a hot spot archipelago. Smithsonian Institution Press, Washington, DC

Wagner W. L., Herbst D. R., Sohmer S. H. (1990) Manual of the flowering plants of Hawai'i. University of Hawai'i Press, Honolulu, Hawai'i, USA

Wanke S, Samain M-S, Vanderschaeve L, Mathieu G, Goetghebeur P, Neinhuis C (2006) Phylogeny of the genus *Peperomia* (Piperaceae) inferred from the *trn*K/*mat*K region (cpDNA). Pl Biol 8:93–102

Wanke S, Jaramillo MA, Borsch T, Samain M-S, Quandt D, Neinhuis C (2007a) Evolution of the Piperales - *mat*K and *trn*K intron sequence data reveal lineage specific resolution contrast. Molec Phylogenet Evol 42:477–497

Wanke S, Vanderschaeve L, Mathieu G, Neinhuis C, Goetghebeur P, Samain M-S (2007b) From forgotten taxon to a missing link? The position of the genus *Verhuellia* (Piperaceae) revealed by molecules. Ann Bot 99:1231–1238

Wen J, Stuessy TF (1993) The phylogeny and biogeography of *Nyssa* (Cornaceae). Syst Bot 18:68–79

Wen J, Jansen RK (1995) Morphological and molecular comparisons of *Campsis grandiflora* and *C. radicans* (Bignoniaceae), an eastern Asian and eastern North American vicariad species pair. Pl Syst Evol 196:173–183

Wen J, Zimmer EA (1996) Phylogeny and biogeography of *Panax* L. (the ginseng genus, Araliaceae): inferences from ITS sequences of nuclear ribosomal DNA. Molec Phylogenet Evol 6:167–177

Wen J, Janzen RK, Zimmer EA (1996) Phylogenetic relationships and DNA sequence divergence of eastern Asian and eastern North American disjunct plants. In: Nei M, Takahata N (eds) Current topics of molecular evolution. Jointly published by the Pennsylvania State University and the Graduate University for Advanced Studies, Hayama, Japan, pp 37–44

Wen J, Shi S, Jansen RK, Zimmer EA (1998) Phylogeny and biogeography of *Aralia* sect. *Aralia* (Araliaceae). Amer J Bot 85:866–875

Whitmore TC (1998) An introduction to tropical rain forests, 2nd edn. Oxford University Press, Oxford

Whitmore TC, Prance GT (1987) Biogeography and Quaternary history in tropical America. Oxford University Press, New York

Williams DA, Overholt WA, Cuda JP, Hughes CR (2005) Chloroplast and microsatellite DNA diversities reveal the introduction history of Brazilian peppertree (*Schinus terebinthifolius*) in Florida. Molec Ecol 14:3643–3656

Wing SL, Boucher LD (1998) Ecological aspects of the Cretaceous flowering plant radiation. Annual Rev Earth Planet Sci 26:379–421

Winkworth RC, Wagstaff SJ, Glenny D, Lockhart PJ (2005) Evolution of the New Zealand mountain flora: origins, diversification and dispersal. Organisms Div Evol 5:237–247

Wolfe JA, Upchurch GR (1987) North American nonmarine climates and vegetation during the Late Cretaceous. Palaeog Palaeoclim Palaeoecol 61:33–77

Won H, Renner SS (2006) Dating dispersal and radiation in the Gymnosperm *Gnetum* (Gnetales) - Clock calibration when outgroup relationships are uncertain. Syst Biol 55:610–622

Wright SD, Yong CG, Dawson JW, Whittaker DJ, Gardner RC (2000) Riding the ice age El Niño? Pacific biogeography and evolution of *Metrosideros* (Myrtaceae) inferred from nuclear ribosomal DNA. Proc Natl Acad Sci USA 97:4118–4123

Wright SD, Yong CG, Dawson JW, Whittaker DJ, Gardner RC (2001) Stepping stones to Hawaii: a trans-equatorial dispersal pathway for *Metrosideros* (Myrtaceae) inferred from nrDNA (ITS plus ETS). J Biogeogr 28:769–774

Xiang Q-Y, Crawford DJ, Wolfe AD, Tang Y-C, DePamphilis CE (1998) Origin and biogeography of *Aesculus* (Hippocastanaceae): a molecular phylogenetic perspective. Evolution 52:988–997

Yoder AD, Irwin JA, Payseur BA (2001) Failure of the ILD to determine data combinability for slow loris phylogeny. Syst Biol 50:408–424

Yoo MJ, Bell CD, Soltis PS, Soltis DE (2005) Divergence times and historical biogeography of Nymphaeales. Syst Bot 30:693–704

Young KR, Ulloa UC, Luteyn JL, Knapp S (2002) Plant evolution and endemism in Andean South America: an introduction. Bot Rev 68:4–21

Zanis MJ, Soltis DE, Soltis PS, Mathews S, Donoghue MJ (2002) The root of the angiosperms revisited. Proc Natl Acad Sci USA 99:6848–6853

Zavada MS, Benson JM (1987) First fossil evidence for the primitive angiosperm family Lactoridaceae. Amer J Bot 74:1590–1594

Zhang L-B, Renner SS (2003) The deepest splits in Chloranthaceae as resolved by chloroplast sequences. Int J Pl Sci 164:S383–S392

Zhang L-B, Simmons MP, Renner SS (2007) A phylogeny of Anisophylleaceae based on six nuclear and plastic loci: ancient disjunctions and recent dispersal between South America, Africa, and Asia. Molec Phylogenet Evol 44:1057–1067

Zhou S, Renner SS, Wen J (2006) Molecular phylogeny and inter- and intracontinental biogeography of Calycanthaceae. Molec Phylogenet Evol 39:1–15

Ziegler AM, Eshel G, Rees PM, Rothfus TA, Rowley DB (2003) Tracing the tropics across land and sea: Permian to present. Lethaia 36:227–254