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Phylogeny of the Clusioid Clade (Malpighiales): Evidence from the Plastid and Mitochonrial Genomes

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Phylogeny of the clusioid clade (Malpighiales): Evidence from the plastid and mitochondrial genomes[1]

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- *Premise of the study*: The clusioid clade includes five families (i.e., Bonnetiaceae, Calophyllaceae, Clusiaceae s.s., Hypericaceae, and Podostemaceae) represented by 94 genera and ~1900 species. Species in this clade form a conspicuous element of tropical forests worldwide and are important in horticulture, timber production, and pharmacology. We conducted a taxon-rich multigene phylogenetic analysis of the clusioids to clarify phylogenetic relationships in this clade.
- *Methods*: We analyzed plastid (*matK*, *ndhF*, and *rbcL*) and mitochondrial (*matR*) nucleotide sequence data using parsimony, maximum likelihood, and Bayesian inference. Our combined data set included 194 species representing all major clusioid subclades, plus numerous species spanning the taxonomic, morphological, and biogeographic breadth of the clusioid clade.
- *Key results*: Our results indicate that *Tovomita* (Clusiaceae s.s.), *Harungana* and *Hypericum* (Hypericaceae), and *Ledermanniella* s.s. and *Zeylanidium* (Podostemaceae) are not monophyletic. In addition, we place four genera that have not been included in any previous molecular study: *Ceratolacis*, *Diamantina*, and *Griffithella* (Podostemaceae), and *Santomasia* (Hypericaceae). Finally, our results indicate that *Lianthus*, *Santomasia*, *Thornea*, and *Triadenum* can be safely merged into *Hypericum* (Hypericaceae).
- *Conclusions*: We present the first well-resolved, taxon-rich phylogeny of the clusioid clade. Taxon sampling and resolution within the clade are greatly improved compared to previous studies and provide a strong basis for improving the classification of the group. In addition, our phylogeny will form the foundation for our future work investigating the biogeography of tropical angiosperms that exhibit Gondwanan distributions.

Key words: *Garcinia mangostana*; Guttiferae; *Hypericum perforatum*; mangosteen; *matK*; *matR*; morphology; *ndhF*; *rbcL*; St. John's wort.

The clusioids are a clade of flowering plants in the large rosid order Malpighiales (Savolainen et al., 2000; Soltis et al., 2000; Wurdack and Davis, 2009). Species in this clade are morphologically heterogeneous and ecologically diverse. Growth forms include large tropical rainforest trees, temperate and high altitude tropical herbs and shrubs, and aquatic plants of swift-flowing rivers and streams. Although their distribution is nearly cosmopolitan, their greatest species diversity is in the tropics. This well-supported clade contains five families (APG III, 2009; Wurdack and Davis, 2009) representing 94 genera and ~1900 species (Kato, 2006; Cook and Rutishauser, 2007; Stevens, 2007a, b; Weitzman et al., 2007; Thiv et al., 2009; Koi and Kato, 2010; Tippery et al., in press): Bonnetiaceae, Calophyllaceae, Clusiaceae s.s., Hypericaceae, and Podostemaceae. The clusioids, excluding Podostemaceae, are an important component of tropical forests and comprise ~3% of the total species diversity in the Center for Tropical Forest Science's global network of tropical forest research plots (CTFS, 2009). Podostemaceae, the largest strictly aquatic flowering plant family, play a key role in river systems—especially through their impact on the ecology and nutrition of fish and invertebrates (Allan, 1995; Machado-Allison et al., 2003). This family occupies a unique ecological niche for angiosperms: growing firmly attached to solid substrates in swift-flowing, nutrient-poor rivers and waterfalls (Philbrick and Novelo, 2004). Their ability to attach to substrates in these harsh environments is facilitated by biofilms partially composed of cyanobacteria, which may function as an important source of nitrogen for the plants

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(Jäger-Zürn and Grubert, 2000). The clusioid clade also contains problematic invasive species, such as *Hypericum perforatum* L., which has been shown to outcompete native species and is toxic to livestock (Huffaker, 1951; Giese, 1980; Mitich, 1994; Vandenbogaerde et al., 1998; Buckley et al., 2003).

Clusioids are also economically important. Many species are cultivated in the horticultural trade (e.g., *Hypericum* spp.) or harvested for timber (e.g., *Calophyllum brasiliense* Cambess., *Mesua ferrea* L.). Several species have pharmacological activity and are potentially useful for the treatment of tumors, depression, and AIDS (Bennett and Lee, 1989; Burkhardt et al., 1994; McKee et al., 1998; Ernst, 2003). St. John's wort (*H. perforatum*), for example, is one of the best-selling herbal medicines worldwide, with annual sales in the United States of around $200 million (Ernst, 2003). Furthermore, members of this clade produce the important tropical fruits the mangosteen (*Garcinia mangostana* L.) and the mammey apple (*Mammea americana* L.).

The current circumscription of the clusioid clade differs from previous morphology-based classifications, and molecular data were required to detect its component families and their interrelationships (Savolainen et al., 2000; Soltis et al., 2000; Gustafsson et al., 2002; Wurdack and Davis, 2009). Cronquist (1981), for example, placed the clusioids in two distantly related orders, Theales and Podostemales, in his subclasses Dilleniidae and Rosidae, respectively. Terrestrial members of this clade (i.e., Bonnetiaceae, Calophyllaceae, Clusiaceae s.s., and Hypericaceae) have long been considered closely related, and the name Clusiaceae (alternately called Guttiferae) has historically been applied to various combinations of taxa now found in these four families (e.g., Cronquist, 1981; Takhtajan, 1997; Mabberley, 2008). The alternate-leaved clusioids, Bonnetiaceae, and some Calophyllaceae were considered closely related to Theaceae s.l. (e.g., Baretta-Kuipers, 1976; Cronquist, 1981; Takhtajan, 1997; Weitzman and Stevens, 1997), but subsequent phylogentic evidence placed Theaceae s.l. in the asterid order Ericales (Stevens, 2001 onward; APG III, 2009). The wholly aquatic Podostemaceae have been very difficult to place owing to their highly atypical morphology, but were never thought to be closely related to other clusioids (Stevens, 2007b). They have long been considered morphological misfits and are so unlike most angiosperms that some systematists suggested they be recognized as their own class, equal in rank to monocots and dicots (Cusset and Cusset, 1988).

These newly discovered relationships have led to a reexamination of morphological characteristics that revealed several putative synapomorphies for the clusioid clade and its major subclades. All clusioid families share distinctive xanthones, and many members of the clade possess exotegmic seeds (Bonnetiaceae, some Calophyllaceae, some Clusiaceae, Hypericaceae, and Podostemaceae). Bonnetiaceae, Clusiaceae s.s., and Hypericaceae share staminal fascicles opposite the petals, and Hypericaceae and Podostemaceae share tenuinucellate ovules. Additionally, Bonnetiaceae, some members of Hypericaceae, and Podostemaceae have papillate stigmas, and Hypericaceae, Calophyllaceae, Clusiaceae s.s., and some Podostemaceae share resin-containing glands or canals that are especially visible in the leaves (Cook and Rutishauser, 2007; Stevens, 2007a, b; Weitzman et al., 2007).

Several molecular phylogenetic studies have focused on individual clusioid families, subfamilies, or genera (Kita and Kato, 2001, 2004a; Abdul-Salim, 2002; Gustafsson and Bittrich, 2002; Gustafsson et al., 2002, 2007; Notis, 2004; Moline et al., 2006, 2007; Sweeney, 2008; Koi et al., 2009; Thiv et al., 2009; Wurdack and Davis, 2009; Tippery et al., in press), but only two of these studies have addressed relationships broadly within the clade. Gustafsson et al. (2002) provided evidence for several major clusioid subclades, most notably Podostemaceae + Hypericaceae. Relationships within and between most subclades, however, were not well resolved. This lack of resolution is likely due to their limited taxon sampling and the use of a single plastid gene, *rbcL*. Wurdack and Davis (2009) analyzed 13 genes from three genomes and provided strong resolution among the major clusioid subclades. In particular, their results included the unexpected finding that Clusiaceae s.l., as traditionally circumscribed, were not monophyletic. However, their taxon sampling was also narrow, including only 17 genera (of 94), each represented by a single placeholder taxon. Despite these insights, many questions remain unanswered. In particular, molecular results surprisingly suggest that the pantropical Symphonieae (Clusiaceae s.s.), with their unique stigmas, are not monophyletic (Gustafsson et al., 2002; Sweeney, 2008). Additionally, intergeneric relationships in most clusioid subclades are unknown, and it is thought that some genera are likely not monophyletic (e.g., *Hypericum*, *Garcinia*, *Ledermanniella* s.s.; Stevens, 2007a, b; Sweeney, 2008; Thiv et al., 2009; Nürk and Blattner, 2010). The major goal of our study is to assemble the first well-supported multigene phylogeny of the clusioid clade with dense taxonomic sampling. This will allow us to better assess the classification of the group, elucidate patterns of character evolution, establish synapomorphies for the major clusioid subclades, and pave the way for larger biogeographic analyses. To achieve our goal, we sampled three plastid genes (*matK*, *ndhF*, and *rbcL*) and the mitochondrial gene *matR* from the broadest clusioid taxon sampling to date.

MATERIALS AND METHODS

Taxon sampling—Our taxon sampling comprises 222 terminals including outgroups. Of these, 194 are clusioid species representing 71 of the 94 currently recognized genera and ~10% of the species diversity in this clade (Cook and Rutishauser, 2007; Stevens, 2007a, b; Weitzman et al., 2007; Thiv et al., 2009; Koi and Kato, 2010; Tippery et al., in press). Voucher information and GenBank numbers for all sequences are provided in Appendix 1. Most missing genera were from Podostemaceae (19 of 23; see Table 1). Tippery et al. (in press) have shown that several genera of Podostemaceae are not monophyletic. The species of *Oserya* that were transferred to *Noveloa* by Tippery et al. are represented here by *N. coulteriana* (Tul.) C.T. Philbrick. In addition, Tippery et al. found that the monotypic *Vanroyenella* was embedded within a Central American clade of *Marathrum.* Accordingly, we have included this species as *Marathrum plumosum* (Novelo & C.T. Philbrick) C.T. Philbrick & C.P. Bove.

Only four small genera outside Podostemaceae are missing from our analyses: *Lebrunia* (monotypic, Africa; Calophyllaceae), *Lianthus* (monotypic, China; Hypericaceae), *Neotatea* (four species, South America; Calophyllaceae), and *Thysanostemon* (two species, South America; Clusiaceae s.s.). Despite several attempts, we were unable to obtain polymerase chain reaction (PCR) amplicons from these taxa, perhaps due to the difficulty of obtaining high quality clusioid DNA from herbarium vouchers (Gustafsson and Bittrich, 2002). Our sampling included four genera that have not been included in previous molecular studies: *Ceratolacis*, *Diamantina*, and *Griffithella* (Podostemaceae), and *Santomasia* (Hypericaceae). We have also increased the taxon sampling across the biogeographical range of the clusioid clade and within numerous genera to begin assessing generic circumscriptions and infrageneric relationships. In some instances, gene sequences from different vouchers of a single species were combined (see Appendix 1). The sister group of the clusioid clade is unclear; therefore, we included 26 taxa representing all major lineages of Malpighiales sensu Wurdack and Davis (2009) as outgroups. Two taxa from the more distant outgroups Celastrales (Celastraceae) and Oxalidales (Oxalidaceae) were also included. Celastraceae were used to root our trees based on the findings by Wang et al. (2009).

TABLE 1. Updated classification of the clusioid clade reflecting the findings of this and other recent studies (see text). Taxa are listed in alphabetical order. Genera marked with "*" are represented in this study. Genera marked with "⊗" have been suggested to be nonmonophyletic with molecular data but taxonomic changes have yet to be made. Recent taxonomic changes sensu Tippery et al. (in press) are marked with "$".

I. Family Bonnetiaceae L. Beauvis. ex Nakai
- *Archytaea* Mart. *
- *Bonnetia* Mart. *
- *Ploiarium* Korth. *

II. Family Calophyllaceae J. Agardh
- A. Tribe Calophylleae Choisy
  - *Calophyllum* L. *
  - *Caraipa* Aubl. *
  - *Clusiella* Planch. & Triana *
  - *Haploclathra* Benth. *
  - *Kayea* Wall. *
  - *Kielmeyera* Mart. & Zucc. *
  - *Mahurea* Aubl. *
  - *Mammea* L. *
  - *Marila* Sw. *
  - *Mesua* L. *
  - *Neotatea* Maguire
  - *Poeciloneuron* Bedd. *
- B. Tribe Endodesmieae Engl.
  - *Endodesmia* Benth. *
  - *Lebrunia* Staner

III. Family Clusiaceae Lindl.
- A. Tribe Clusieae Choisy
  - *Chrysochlamys* Poepp. *
  - *Clusia* L. *
  - *Dystovomita* (Engl.) D'Arcy *, ⊗
  - *Tovomita* Aubl. *, ⊗
  - *Tovomitopsis* Planch. & Triana *
- B. Tribe Garcinieae Choisy
  - *Garcinia* L. * (including *Allanblackia* Oliv. *)
- C. Tribe Symphonieae Choisy
  - *Lorostemon* Ducke *
  - *Montrouziera* Planch. & Triana *
  - *Moronobea* Aubl. *
  - *Pentadesma* Sabine *
  - *Platonia* Mart. *
  - *Symphonia* L.f. *
  - *Thysanostemon* Maguire

IV. Family Hypericaceae Juss.
- A. Tribe Cratoxyleae Benth. & Hook.f.
  - *Cratoxylum* Blume *
  - *Eliea* Cambess. *
- B. Tribe Hypericeae Choisy
  - *Hypericum* L. * (including *Lianthus* N. Robson, *Santomasia* N. Robson *, *Thornea* Breedlove & E.M. McClint. *, and *Triadenum* Raf. *)
- C. Tribe Vismieae Choisy
  - *Harungana* Lam. *, ⊗
  - *Vismia* Vand. *, ⊗

V. Family Podostemaceae Rich. ex Kunth
- A. Subfamily Podostemoideae Wedd.
  - *Angolaea* Wedd.
  - *Autania* C.T. Philbrick $
  - *Apinagia* Tul. *, ⊗
  - *Butumia* G. Taylor
  - *Castelnavia* Tul. & Wedd. *
  - *Ceratolacis* (Tul.) Wedd. *
  - *Cipoia* C.T. Philbrick, Novelo & Irgang
  - *Cladopus* H.A. Möller *
  - *Diamantina* Novelo, C.T. Philbrick & Irgang *
  - *Dicraeanthus* Engl. *
  - *Diplobryum* C. Cusset
  - *Djinga* C. Cusset *
  - *Endocaulos* C. Cusset *
  - *Farmeria* Willis
  - *Griffithella* (Tul.) Warm. *
  - *Hanseniella* C. Cusset *
  - *Hydrobryum* Endl. *
  - *Hydrodiscus* Koi & M. Kato
  - *Inversodicraea* Engl. ex R.E. Fr. *
  - *Jenmaniella* Engl. ⊗
  - *Ledermanniella* Engl. *, ⊗
  - *Leiothylax* Warm. *
  - *Letestuella* G. Taylor *
  - *Lophogyne* Tul.
  - *Macarenia* P. Royen
  - *Macropodiella* Engl. *
  - *Marathrum* Humb. & Bonpl. *, ⊗, $ (including *Vanroyenella* Novelo & C.T. Philbrick *)
  - *Monandriella* Engl. *
  - *Monostylis* Tul. *
  - *Mourera* Aubl. *, $ (including *Lonchostephus* Tul. and *Tulasneantha* P. Royen)
  - *Noveloa* C.T. Philbrick *, $ (*Oserya* Tul. & Wedd. pro parte)
  - *Oserya* Tul. & Wedd.
  - *Paleodicraeia* C. Cusset
  - *Paracladopus* M. Kato *
  - *Podostemum* Michx. * (including *Crenias* Spreng. * and *Devillea* Tul. & Wedd.)
  - *Polypleurum* Warm. *
  - *Rhyncholacis* Tul. *
  - *Saxicolella* Engl.
  - *Sphaerothylax* Bisch. ex Krauss
  - *Stonesia* G. Taylor *
  - *Thawatchaia* M. Kato, Koi & Y. Kita *
  - *Thelethylax* C. Cusset *
  - *Wettsteiniola* Suess.
  - *Willisia* Warm.
  - *Winklerella* Engl.
  - *Zehnderia* C. Cusset
  - *Zeylanidium* (Tul.) Engl. *, ⊗
- B. Subfamily Tristichoideae Engler
  - *Cussetia* M. Kato
  - *Dalzellia* Wight *
  - *Indodalzellia* Koi & M. Kato *
  - *Indotristicha* P. Royen *
  - *Terniopsis* H.C. Chao *
  - *Tristicha* Thouars *
- C. Subfamily Weddellinoideae Engler
  - *Weddellina* Tul. *

Molecular methods—PCR amplification and automated sequencing mostly followed Wurdack and Davis (2009). When these protocols were unsuccessful, we used additional primers from the literature (*matK*: trnk-710F, 1168R [Johnson and Soltis, 1995], pod2R, pod3F, pod7F [Kita and Kato, 2001]; *ndhF*: 536F, 1318F, 1318R, 1603R [Olmstead and Sweere, 1994] and 2153R [Wang et al., 2009]; and *rbcL*: 1204R [Zurawski et al., 1981]) plus several designed here (see Table 2). Primers were frequently optimized independently for each major clusioid subclade. Primer mismatch was also addressed using a step-down PCR procedure (Korbie and Mattick, 2008). Depending on the quality of the DNA template and the presence of homopolymer regions (which were particularly common in Hypericaceae and Podostemaceae), gene regions were sometimes amplified and sequenced in smaller fragments and assembled into a larger contig. PCR products were sequenced using the facilities and protocols at Functional Biosciences (Madison, Wisconsin, USA).

In addition, we included plastid data (*matK*, *ndhF*, and *rbcL*) from seven clusioid plastid genomes: *Clusia rosea* Jacq. and *Garcinia mangostana* L. (Clusiaceae s.s.); *Hypericum kalmianum* L., *H. perforatum* L., *Triadenum fraseri* (Spach) Gleason, and *Vismia guianensis* (Aubl.) Choisy (Hypericaceae); and *Podostemum ceratophyllum* Michx. (Podostemaceae). These data were

TABLE 2. Primer table.

Gene	Primer	Sequence	Original publication	Clade/Use
matK	Afm	5′-ATCCACTTATCTTTCAGGAG-3′	(Ooi et al., 1995)	P
	400fm	5′-TCAGAATTTACGATCCATTCTTTCAAT-3′	(Cameron et al., 2001)	H
	1053Fm1	5′-CAATRTCATTTTWMTGTRTG-3′	(Wurdack and Davis, 2009)	B, C
	1053Fm2	5′-TCAATRKCATTTTTHTGTRTGG-3′	(Wurdack and Davis, 2009)	H, K
	1159Rm1	5′-TSTARYATTTGACTYCGKACCACBG-3′	(Wurdack and Davis, 2009)	B, C
	1159Rm2	5′-AGCATTTGACTTCGTAYCRCTG-3′	(Wurdack and Davis, 2009)	H, K
	EHypR	5′-AACTCTCGAKCAAGATGTGTAGG-3′	New to this study	H
ndhF	1098F	5′-AATGGAAGCTATTGTTGGTTATTCTC-3′	New to this study	All clades
	1676R	5′-GAATTGATTGAAAGGAATTCCKA-3′	K. Wurdack, unpublished	Degraded templates
rbcL	cRm	5′-GCAGCAGCTARTTCMGGACTCCA-3′	(Hasebe et al., 1994)	All clades
	636Fm	5′-ATGCGWTGGAGRGAYCGNTT-3′	(Lledo et al., 1998)	All clades
	724Rm	5′-TCRCATGTACCNGCRGTWG-3′	(Lledo et al., 1998)	All clades
	1204Rm	5′-CAAGGATGNCCTAARGTTCC-3′	(Zurawski et al., 1981)	All clades
matR	879Fm	5′-AGTTATTMTCAKGTCAGAGA-3′	(Meng et al., 2002)	All clades
	1002Rm	5′-CACCKWHGATTCCYAGTAGT-3′	(Meng et al., 2002)	All clades

Notes: Primers have the same name as in the publication listed followed by an "m" to indicate that they have been modified for use in the clusioid clade. Bonnetiaceae (B), Calophyllaceae (K), Clusiaceae s.s. (C), Hypericaceae (H), and Podostemaceae (P).

collected as part of a larger study to use complete plastid genomes to resolve relationships of the major subclades of Malpighiales (Xi et al., 2010).

Sequence assembly and phylogenetic analyses—Chromatograms were assembled into contiguous sequences and checked for accuracy using the program Sequencher ver. 4.9 (Gene Codes Corp., Ann Arbor, Michigan, USA). Primer regions were removed and sequences were aligned by eye as translated amino acids using the program MacClade ver. 4.08 (Maddison and Maddison, 2005). The ragged ends of the alignments and ambiguous internal regions were trimmed prior to analysis. Data matrices and trees are available in the database TreeBASE (http://www.treebase.org; accession S10995) and from the first author.

Maximum parsimony (MP), maximum likelihood (ML), and Bayesian inference (BI) were conducted to infer the phylogeny of the clusioid clade. We analyzed potential conflict between the individual and combined data sets using alternative topology testing (see below). Analyses of the combined data were conducted on reduced and expanded data sets. The reduced data set contained fewer taxa, but greater character density (ntax = 169, missing data = 8.4%). The expanded data set contained more taxa, but some taxa were missing data from one or more gene regions (ntax = 222, missing data = 19.4%). The expanded data set was important for including the most morphological, taxonomic, and biogeographic diversity in the group. Taxa with missing characters or characters lacking data from some taxa are often excluded from phylogenetic studies due to concerns surrounding the adverse effects of missing data on phylogenetic inference. However, recent work suggests that including taxa with missing data can provide increased phylogenetic resolution (McMahon and Sanderson, 2006; Wiens, 2006; Wiens and Moen, 2008).

The MP analyses were conducted with the program PAUP* ver. 4.0b10 (Swofford, 2003) using the parsimony ratchet (Nixon, 1999) as implemented in the program PAUPRat (Sikes and Lewis, 2001; distributed by D. Sikes at http://users.iab.uaf.edu/~derek_sikes/software2.htm). We conducted 10 replicates of 200 iterations each with 15% of characters reweighted per iteration. Gaps were treated as missing data and included in the analyses. Bootstrap percentage (BP) support (Felsenstein, 1985) for each clade was estimated from 1000 heuristic search replicates using PAUP* (10 random taxon addition replicates, tree-bisection-reconnection [TBR] swapping, option MULTREES = yes, and holding no more than 10 trees per replicate).

The ML analyses were implemented with the parallel versions of the program RAxML ver. 7.2.5 or 7.2.6 (Stamatakis, 2006; distributed by A. Stamatakis at http://wwwkramer.in.tum.de/exelixis/software.html). Two partitioning schemes for each data set were used: unpartitioned and partitioned by gene region. Each analysis was conducted five times with different starting trees to check for convergence in likelihood values. We determined the optimal model of evolution for the unpartitioned and partitioned data sets by using the Akaike information criterion (AIC) as implemented in the program ModelTest ver. 3.7 (Posada and Crandall, 1998; Posada and Buckley, 2004). However, because RAxML does not allow for the specification of the TVM+I+Γ model (Table 3), the GTR+Γ model of evolution was applied to each partition in the partitioned data sets with all parameters estimated from the data. The TVM and GTR models differ only by a single parameter; TVM constrains transition rates to be equal while transition rates are allowed to vary in the GTR model (Posada and Buckley, 2004). We chose not to estimate the proportion of invariant sites in the ML and BI analyses as suggested in the RAxML manual. The invariant sites model, in particular, can fail to find important patterns of variation in the data as discussed by Pagel and Meade (2005). For each analysis, the optimal ML tree and BP values were estimated in the same run using the default settings. The ML BP values were obtained from 1000 bootstrap replicates using the rapid bootstrap algorithm implemented in RAxML (Stamatakis et al., 2008).

The BI analyses were conducted using the parallel version of the program BayesPhylogenies ver. 1.1 (Pagel and Meade, 2004; distributed by M. Pagel at http://www.evolution.rdg.ac.uk/BayesPhy.html) using a reversible-jump implementation of the mixture model as described in Venditti et al. (2008). This approach allows the fitting of multiple models of sequence evolution to the data without a priori partitioning. Default settings were applied, and a GTR model was used with among-site rate variation estimated by a gamma distribution with four rate categories. We performed three independent analyses on each data set (six total runs) to determine consistency of stationary-phase likelihood values and estimated parameter values between runs. Each Markov chain Monte Carlo run consisted of 10 million generations, with sampling of trees and parameters every 1000 generations. Convergence was assessed using the program Tracer ver. 1.5 (distributed by A. Rambaut at http://tree.bio.ed.ac.uk/software/tracer/). Posterior probabilities (PP) were determined by building a 50% majority rule consensus tree after discarding the burn-in generations (the first 20% of the topologies were excluded in the first five runs; 40% of the topologies were excluded in the sixth).

Alternative topology tests—Alternative topology tests were conducted in a ML framework using the approximately unbiased (AU) test (Shimodaira, 2002) as implemented in the R software package, scaleboot ver. 0.3-2 (Shimodaira, 2008; distributed by CRAN at http://www.r-project.org). All constrained searches were conducted, as described above, using the reduced and expanded data sets. We initially determined whether the combined data could reject any of the topologies produced by individual genes, thereby indicating potential problems for analyzing these genes simultaneously. To achieve this goal, we conducted separate tree searches on single gene data sets (*matK*, *ndhF*, *rbcL*, and *matR*). We considered two topologies to be at odds if both contained conflicting clades supported by ≥80 BP. As such, clades supported by ≥80 BP in these individual gene analyses were then used to constrain searches on the combined data. In addition, we also tested the monophyly of several traditionally recognized taxa that were found to be nonmonophyletic in our analyses. We separately enforced monophyly for Clusiaceae s.l. (Calophyllaceae + Clusiaceae s.s), *Dystovomita*, *Garcinia*, *Harungana*, *Hypericum*, *Ledermanniella*, *Tovomita*, and *Zeylanidium*. Testing the monophyly of *Dystovomita* and *Zeylanidium* using the reduced data set was not possible due to insufficient taxon sampling. Finally, we assessed the alternative placement of *Mourera* as found in the MP analyses. In the MP analyses of both combined data sets, *Mourera* was placed sister to the Podostemoideae excluding *Diamantina,* while in the ML and BI analyses it was placed sister to a clade containing *Apinagia*, *Castelnavia*, *Marathrum*, *Monostylis*, *Noveloa*, and *Rhyncholacis*. The MP placement was enforced and tested against the unconstrained ML trees.

TABLE 3. Data set characteristics. Values listed for individual genes are for the alignments derived from the reduced / expanded data sets, respectively. Percentage of missing data is calculated as the total number of ?'s in the analyzed matrix divided by the total number of characters including gaps. Models of sequence evolution were chosen by the Akaike information criterion using ModelTest 3.7. pt, plastid; mt, mitochondrial.

Characteristic	pt *matK*	pt *ndhF*	pt *rbcL*	mt *matR*	Reduced total	Expanded total
Terminals	169 / 209	169 / 204	169 / 201	169 /190	169	222
Characters analyzed	1455	1086	1296	2400	6237	6237
% missing data	8.4 / 10.9	15.4 / 17.6	5.1 / 5.1	6.9 / 9.6	8.4	19.4
% gaps plus missing data	31.4 / 32.7	28.0 / 29.8	5.4 / 8.1	35.4 / 36.0	26.9	35.6
Constant characters	555 / 528	403 / 400	781 / 770	1467 / 1450	3206	3148
Variable characters	900 / 927	683 / 686	515 / 526	933 / 950	3031	3089
Parsimony informative characters	732 / 766	550 / 560	371 / 382	586 / 606	2239	2314
% Parsimony informative characters	50 / 53	51 / 52	29 / 29	24 / 25	36	37
Model of sequence evolution	TVM+I+Γ / TVM+I+Γ	TVM+I+Γ / TVM+I+Γ	GTR+I+Γ / TVM+I+Γ	GTR+Γ / GTR+Γ	GTR+I+Γ	GTR+I+Γ

RESULTS

Sequences/matrices—Our combined alignment included 6237 nucleotide bases. One hundred fifty-seven, 161, 125, and 144 sequences for *matK*, *ndhF*, *rbcL*, and *matR* were newly obtained for this study, respectively (Appendix 1; GenBank numbers HQ331542-HQ332128). These additions include the first published *ndhF* sequences for Podostemaceae. Genes *matK* and *ndhF* were the most variable markers and had a nearly equal percentage of parsimony informative characters; *rbcL* was slightly more informative than *matR*. Relevant characteristics for each gene region and data set are listed in Table 3.

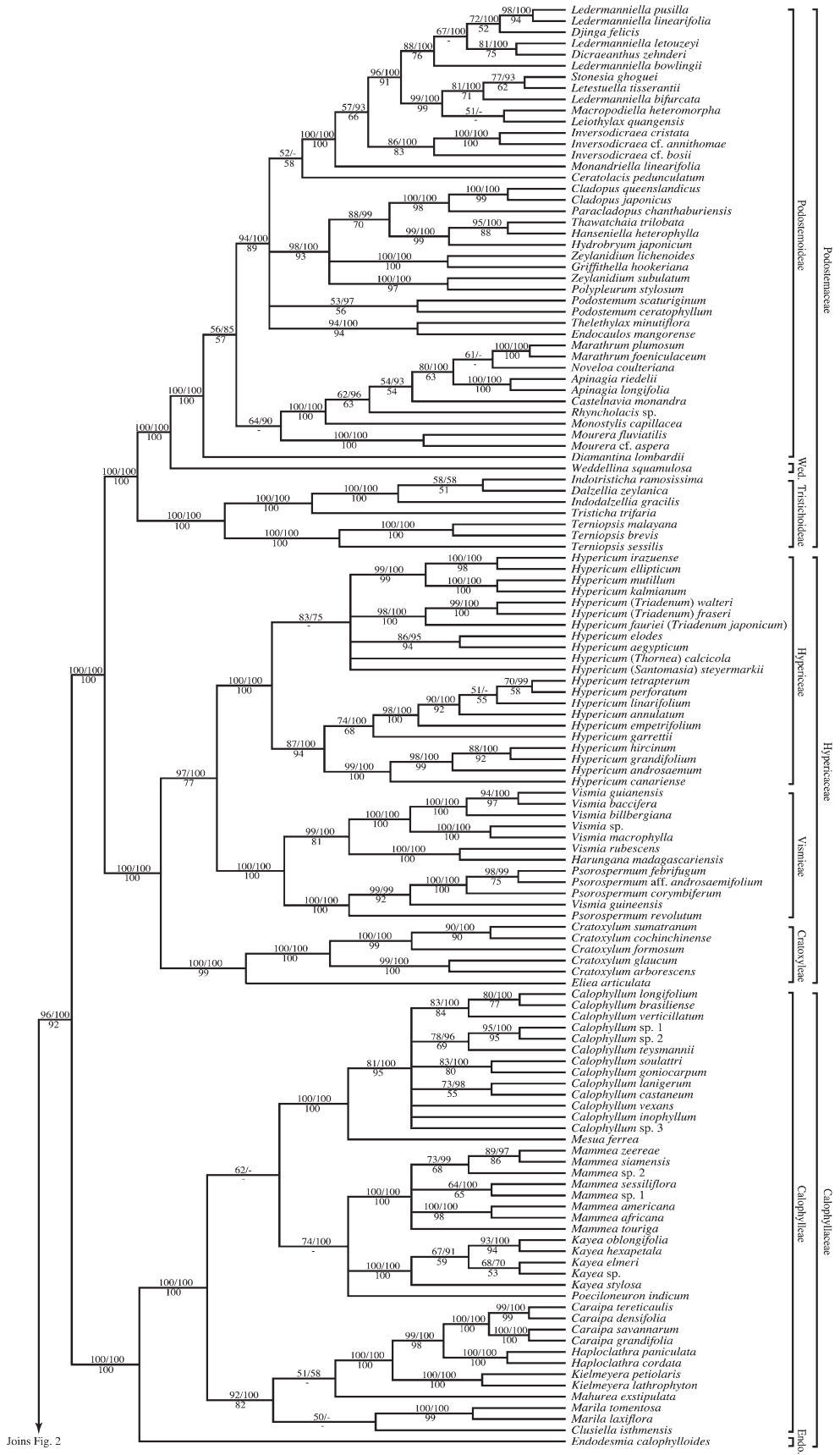
Phylogenetic analyses—Topologies derived from the combined data sets using MP, ML, and BI methods were largely congruent and contained no well-supported differences. Additionally, ML topologies resulting from unpartitioned and partitioned data sets were also congruent within and between partitioning schemes. The MP BP values were often lower than ML BP values, while BI support values were sometimes much higher (see Figs. 1–4). Furthermore, artificially inflated support values in BI analyses have been previously noted (Suzuki et al., 2002; Douady et al., 2003; Simmons et al., 2004). For these reasons, we will focus our discussion below on the 50% ML majority-rule consensus tree of the partitioned expanded data set (Figs. 1, 2). In addition, results from the partitioned reduced data set (Figs. 3, 4) and BI support values from each figure will be mentioned where relevant.

Outgroup relationships are generally in agreement with those reported in Wurdack and Davis (2009): Malpighiales are strongly supported (100 BP; data not shown) as monophyletic, but relationships between its major subclades are largely unresolved. One difference in the Bayesian analyses relates to the placement of *Bruguiera* (Rhizophoraceae) as sister to *Cyrillopsis* (Ixonanthaceae) with 95 PP and 98 PP with the reduced and expanded data sets, respectively (data not shown). *Irvingia* (Irvingiaceae) is in turn sister to this clade with 98 PP and 90 PP with the reduced and expanded data sets, respectively (data not shown). The placements of Irvingiaceae and Ixonanthaceae were unresolved by Wurdack and Davis (2009), and Rhizophoraceae + Erythroxylaceae were instead placed as sister to Ctenolophonaceae. The latter was unplaced in our results. We advise caution when interpreting these results, however, because our sampling includes a relatively small representation of non-clusioid taxa and far fewer genes than in the study by Wurdack and Davis (2009).

The clusioid clade and each of its five families are strongly supported (100 BP) as monophyletic in all analyses. Moreover, the interfamilial relationships reported here are the same as those in Wurdack and Davis (2009). Within the clusioid clade, Bonnetiaceae and Clusiaceae s.s. form a clade (88 BP; Fig. 2). This clade is sister to a strongly supported (96 BP; Fig. 1) clade containing the remaining three families Calophyllaceae, Hypericaceae, and Podostemaceae. Calophyllaceae are sister to a strongly supported (100 BP) clade containing Hypericaceae and Podostemaceae.

Alternative topology tests—No individual gene topologies from the expanded data set were rejected by the combined expanded data set. The individual gene topologies of *ndhF* and *matR* derived from the reduced data set, however, were rejected by the reduced combined data (Table 4). In the *ndhF* topology, well-supported conflict was identified in Hypericaceae and Podostemaceae. In Hypericaceae, conflict involved the placement of *Hypericum grandifolium* Choisy. This taxon was sister to *Hypericum androsaemum* L. in the *ndhF* topology (85 BP; data not shown), but sister to *Hypericum hircinum* L. in the combined data topology (88 BP; Fig. 3). Conflict in Podostemaceae involved the placement of *Dicraeanthus zehnderi* H.E. Hess, which was placed sister to *Ledermanniella bowlingii* (J.B. Hall) C. Cusset in the *ndhF* topology (91 BP; data not shown) but sister to *Ledermanniella letouzeyi* C. Cusset in the combined data topology (83 BP; Fig. 3). In the *matR* topology, well-supported conflict was identified in the *Caraipa* (Calophyllaceae) and *Cratoxylum* (Hypericaceae) clades. *Caraipa densifolia* Mart. was placed sister to a well-supported (81 BP; data not shown) clade containing the remaining *Caraipa* species. In the combined reduced topology, *C. densifolia* was instead strongly placed

Fig. 1. Fifty percent maximum likelihood (ML) majority-rule consensus tree of the clusioid clade based on the combined four-gene expanded data set (ntax = 222, missing data = 19.4%). Support values ≥ 50% are indicated. Values above branches are ML bootstrap values (left) and Bayesian inference posterior probabilities converted to percentages (right). Maximum parsimony bootstrap values are given below each branch. A hyphen indicates that the node was not present in a particular analysis. Endo., Endodesmieae; Wed., Weddellinoideae. Revised names for Hypericeae genera are given; former names are included in parentheses. Tree continued in Fig. 2.


Ledermanniella pusilla
Ledermanniella linearifolia
Djinga felicis
Ledermanniella letouzeyi
Dicraeanthus zehnderi
Ledermanniella bowlingii
Stonesia ghoguei
Letestuella tisserantii
Ledermanniella bifurcata
Macropodiella heteromorpha
Leiothylax quangensis
Inversodicraea cristata
Inversodicraea cf. annithomae
Inversodicraea cf. bosii
Monandriella linearifolia
Ceratolacis pedunculatum
Cladopus queenslandicus
Cladopus japonicus
Paracladopus chanthaburiensis
Thawatchaia trilobata
Hanseniella heterophylla
Hydrobryum japonicum
Zeylanidium lichenoides
Griffithella hookeriana
Zeylanidium subulatum
Polypleurum stylosum
Podostemum scaturiginum
Podostemum ceratophyllum
Thelethylax minutiflora
Endocaulos mangorense
Marathrum plumosum
Marathrum foeniculaceum
Novelova coulteriana
Apinagia riedelii
Apinagia longifolia
Castelnavia monandra
Rhyncholacis sp.
Monostylis capillacea
Mourera fluviatilis
Mourera cf. aspera
Diamantina lombardii
Weddellina squamulosa
Indotristicha ramosissima
Dalzellia zeylanica
Indodalzellia gracilis
Tristicha trifaria
Terniopsis malayana
Terniopsis brevis
Terniopsis sessilis
Hypericum irazuense
Hypericum ellipticum
Hypericum mutillum
Hypericum kalmianum
Hypericum (Triadenum) walteri
Hypericum (Triadenum) fraseri
Hypericum fauriei (Triadenum japonicum)
Hypericum elodes
Hypericum aegypticum
Hypericum (Thornea) calcicola
Hypericum (Santomasia) steyermarkii
Hypericum tetrapterum
Hypericum perforatum
Hypericum linarifolium
Hypericum annulatum
Hypericum empetrifolium
Hypericum garrettii
Hypericum hircinum
Hypericum grandifolium
Hypericum androsaemum
Hypericum canariense
Vismia guianensis
Vismia baccifera
Vismia billbergiana
Vismia sp.
Vismia macrophylla
Vismia rubescens
Harungana madagascariensis
Psorospermum febrifugum
Psorospermum aff. androsaemifolium
Psorospermum corymbiferum
Vismia guineensis
Psorospermum revolutum
Cratoxylum sumatranum
Cratoxylum cochinchinense
Cratoxylum formosum
Cratoxylum glaucum
Cratoxylum arborescens
Eliea articulata
Calophyllum longifolium
Calophyllum brasiliense
Calophyllum verticillatum
Calophyllum sp. 1
Calophyllum sp. 2
Calophyllum teysmannii
Calophyllum soulattri
Calophyllum goniocarpum
Calophyllum lanigerum
Calophyllum castaneum
Calophyllum vexans
Calophyllum inophyllum
Calophyllum sp. 3
Mesua ferrea
Mammea zeereae
Mammea siamensis
Mammea sp. 2
Mammea sessiliflora
Mammea sp. 1
Mammea americana
Mammea africana
Mammea touriga
Kayea oblongifolia
Kayea hexapetala
Kayea elmeri
Kayea sp.
Kayea stylosa
Poeciloneuron indicum
Caraipa tereticaulis
Caraipa densifolia
Caraipa savannarum
Caraipa grandifolia
Haploclathra paniculata
Haploclathra cordata
Kielmeyera petiolaris
Kielmeyera lathrophyton
Mahurea exstipulata
Marila tomentosa
Marila laxiflora
Clusiella isthmensis
Endodesmia calophylloides
Podostemaceae
Podostemoideae
Weddellinoideae
Tristichoideae
Hypericaceae
Hypericeae
Vismieae
Cratoxyleae
Calophyllaceae
Calophylleae
Endo.
Joins Fig. 2

(98 BP; Fig. 3) as sister to *Caraipa tereticaulis* Tul. Conflict within *Cratoxylum* involved the placement of *Cratoxylum formosum* (Jack) Dyer. This taxon was sister to *Cratoxylum sumatranum* (Jack) Blume in the *matR* topology (88 BP; data not shown) but sister to the well-supported (89 BP; Fig. 3) clade containing *Cratoxylum cochinchinense* (Lour.) Blume and *C. sumatranum* in the combined topology. Upon further inspection, it appears that partially missing *matR* data for *Caraipa densifolia* and *Cratoxylum cochinchinense* may explain these incongruencies. The *matR* sequences of the taxa within each of these clades are identical, and as a result, taxa with partial *matR* data may be spuriously placed.

These topological conflicts suggest possible concerns with combining our data in the reduced data set for phylogenetic analyses. However, topologies derived from the individual genes and the topology produced by the combined reduced data set are largely congruent, and where topological differences occur, very few of these are moderately to strongly supported. Importantly, these differences are only near the tips and between closely related taxa, indicating that conflict in the backbone of the topology was not evident. We advise readers to proceed cautiously when interpreting areas where conflict was discovered in the *ndhF* gene topology when compared to the combined reduced topology. Nevertheless, none of these areas are the focus of our study, and as such their implications will not be discussed further.

Finally, the monophyly of Clusiaceae s.l. (Calophyllaceae + Clusiaceae s.s.) could not be rejected (Table 4). Specific results concerning the topology and topological tests within each family are addressed in the Discussion.

DISCUSSION

Our results have provided several new insights into the clusioid phylogeny. We increased ingroup taxon sampling by at least a factor of 4.5 compared to previous studies (Gustafsson et al., 2002; Wurdack and Davis, 2009), and resolution within the clade is much greater than in previous studies: over 60% of the clades in the ML tree were resolved with ≥80 BP (Figs. 1, 2). We resolved the position of four genera that have not been included in previous molecular studies (i.e., *Diamantina*, *Ceratolacis*, *Griffithella*, and *Santomasia*), and identified several genera that are not monophyletic as currently circumscribed (i.e., *Harungana*, *Hypericum*, *Ledermanniella* s.s., *Tovomita*, and *Zeylanidium*). This phylogeny provides a firm foundation for reassessing the current classification of the clusioid clade (see Table 1 for a summary of our proposed changes). We discuss important results for each family below.

Bonnetiaceae—Bonnetiaceae are a small family of 35 species with a disjunct distribution between South America and Southeast Asia. *Archytaea* and *Bonnetia* are distributed exclusively in the New World, while *Ploiarium* are found only in Southeast Asia. Bonnetiaceae are split into two strongly supported (100 BP) subclades: the first containing the genera *Archytaea* and *Ploiarium,* and the second containing *Bonnetia*. These two subclades are well defined by anatomical, vegetative, and floral features (Baretta-Kuipers, 1976; Dickison and Weitzman, 1996, 1998; Weitzman and Stevens, 1997; Weitzman, 2005; Weitzman et al., 2007). *Archytaea* and *Ploiarium* share unilacunar nodes, vascularized disciform structures on leaves and/or bracts, and marginal setae of the leaves associated with vascular tissue. Shared floral features between these two genera include a five-locular ovary that develops into a capsule that dehisces from the proximal end. Additionally, their androecium is fasciculate with five staminodes. In *Bonnetia*, nodes are trilacunar, no disciform structures are present on the leaves and/or bracts, and marginal setae are not associated with vascular tissue. The ovary in *Bonnetia* is three- to four-locular and develops into a capsule that dehisces normally from the distal end. The androecium is apparently not fasciculate (but see Steyermark, 1984), and staminodes are absent. *Bonnetia* additionally have a mucilaginous epidermis, a foliar endodermis, and foliar sclereids, which are not present in the *Archytaea* + *Ploiarium* clade.

All previous molecular studies that included *Bonnetia* sampled only a single species. We include eight species representing the entire biogeographic range of the genus. Within *Bonnetia*, *B. roraimae* Oliv. is placed sister to the remaining *Bonnetia* species. This relationship is weakly supported by ML (53 BP), but strongly supported by BI (97 PP). *Bonnetia ahogadoi* (Steyerm.) A.L. Weitzman & P.F. Stevens was placed by Steyermark (1984) in a separate genus, *Acopanea.* Weitzman and Stevens (1997) transferred *Acopanea* into *Bonnetia* on the basis of anatomy and morphology, a conclusion which is supported by our analyses. Only three *Bonnetia* species [i.e., *B. cubensis* (Britton) R.A. Howard, *B. stricta* (Nees) Nees & Mart., and *B. paniculata* Spruce] occur outside of the Guiana Shield region in adjacent areas in South America and Cuba. These species are embedded within the *Bonnetia* clade (Fig. 2). The phylogenetic distribution of *Bonnetia* species occurring in the Guiana Shield suggests that this region is not only the center of diversity for the genus, but may also be its center of origin.

Calophyllaceae—All genera of Calophyllaceae are monophyletic in our analyses. The monotypic genus *Endodesmia* is well supported (100 BP) as sister to the remaining Calophyllaceae. This latter clade represents tribe Calophylleae, which contains three moderately to well-supported subclades, whose interrelationships are unclear. The first is strongly supported (92 BP) and contains the strictly New World genera *Caraipa*, *Clusiella*, *Haploclathra*, *Kielmeyera*, *Mahurea*, and *Marila*. The alternate-leaved genera *Caraipa*, *Kielmeyera*, and *Mahurea* occur together in a weakly supported clade (51 BP) with the opposite-leaved *Haploclathra*, which is sister to *Caraipa* (99 BP). In contrast to other Calophyllaceae, these four genera, as well as the unsampled *Neotatea*, possess winged seeds (Notis, 2004). Taxa with cordate cotyledons (*Caraipa*, *Haploclathra*, and *Kielmeyera*) form a strongly supported (100 BP) clade. *Clusiella* and *Marila* are weakly supported (50 BP) as a clade in the expanded data set, but support for this relationship increases greatly in the reduced data set analysis (71 BP; Fig. 3). This relationship has been suggested by Hammel (1999b) based on the shared features of small foveolate seeds and an embryo with well-developed cotyledons. In addition, investigations of the cotyledon-to-hypocotyl ratio in Calophyllaceae indicate that *Clusiella*, *Marila*, *Neotatea*, and *Mahurea* possess ratios between 0.2 to 2, while all other Calophyllaceae have a ratio greater than 2 (P.F. Stevens, Missouri Botanical Garden and University of Missouri, St. Louis, unpublished data).

The second and third subclades together form a poorly supported clade (62 BP). The second subclade is moderately supported (74 BP) and includes *Kayea*, *Mammea*, and *Poecilloneuron*; the third subclade is strongly supported (100 BP) and includes *Calophyllum* and *Mesua*. Although molecular support for the sister-group relationship of these subclades is weak, a

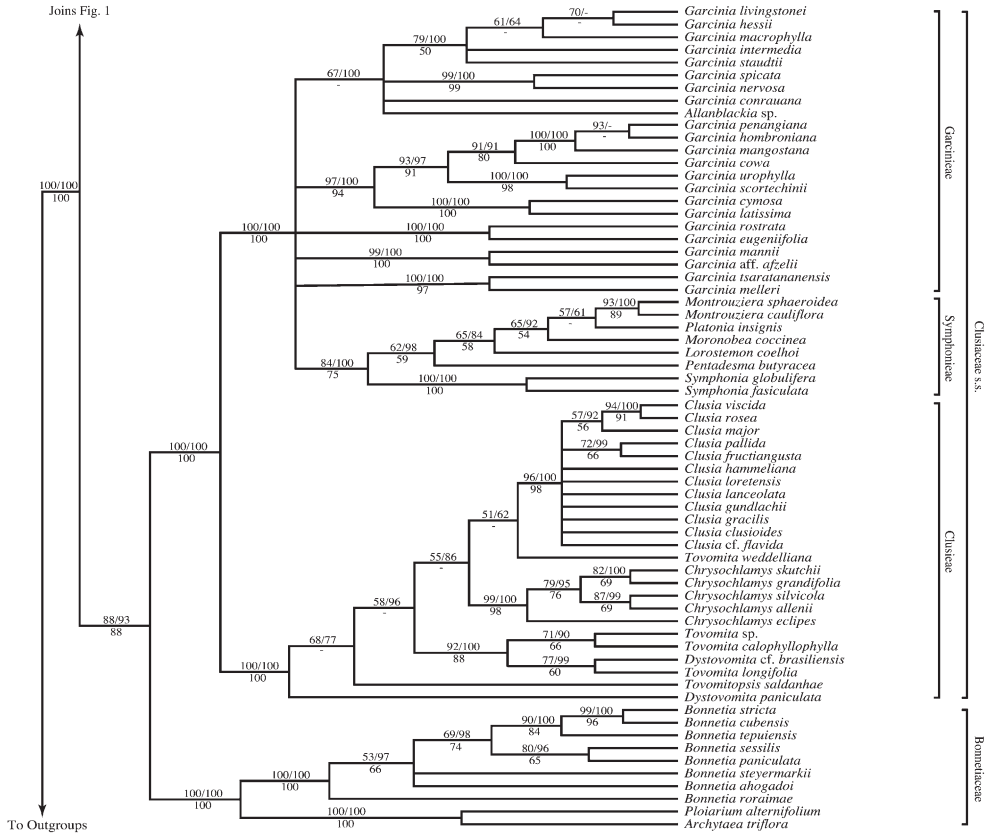


Fig. 2. Continuation of Fig. 1. Fifty percent maximum likelihood majority-rule consensus tree of the clusioid clade based on the combined four-gene expanded data set (ntax = 222, missing data = 19.4%). Outgroups removed to show only the clusioid clade.

close relationship among these taxa has been suggested based on morphology (Engler, 1925; Stevens, 1980). Taxa in these clades possess ovules with basal placentation, and commonly two to four sepals, petals, and carpels. Genera in these two subclades also share primarily Old World distributions. The larger genera, *Calophyllum* and *Mammea,* additionally include a small number of New World species. The New World species of these genera that we sampled are embedded within these principally Old World clades (*Calophyllum brasiliense* Cambess. and *C. longifolium* Willd.; *Mammea americana*; Fig. 1), suggesting a possible Old World origin for *Calophyllum* and *Mammea*. In contrast to members of these principally Old World subclades, members of the strictly New World subclade described above tend to have axile or intruded parietal placentation, five sepals and petals, and three carpels (Notis, 2004; Stevens, 2007a). Our placement of *Mammea* differs strongly from Notis (2004), who found it to be sister to all other Calophylleae. Relationships between *Kayea*, *Mammea*, and *Poeciloneuron* are unresolved in our trees. Although *Kayea* and *Poeciloneuron* are poorly supported as sister taxa (<50 BP), this relationship is corroborated by Notis (2004) and by morphology. These genera share a punctate stigma that differs from the expanded stigma of *Mammea* (Notis, 2004).

Our sampling within *Mammea* allowed us to partially examine the phylogenetic hypothesis of Dunthorn (2009) who proposed species groups based on variation in leaf and petiole anatomy. Our results indicate that species of his "Americana group" (represented by *Mammea americana* L. and *Mammea africana* G. Don in Fig. 1) are strongly monophyletic (100 BP), but members of his "Eugenioides group" (represented by *Mammea siamensis* (Miq.) T. Anderson, *M.* sp. 1, and *M.* sp. 2 in Fig. 1) are not. The position of the distinctive *Mammea touriga* (C.T. White & Francis) L.S. Sm., a species that lacks lamina fibers (Dunthorn, 2009), is not well supported in our ML analyses. Nevertheless, in both ML trees (data not shown), this taxon is placed sister to a clade containing *M. americana* and *M. africana*, which also lack lamina fibers. Finally, results within *Mammea* are interesting biogeographically because the Malagasy species (represented by *M.* sp. 1, *M. sessiliflora* Planch. & Triana, and *Mammea zeereae* P.F. Stevens in Fig. 1) do not form a clade.

We were unable to sample the genera *Lebrunia* (Endodesmieae) and *Neotatea* (Calophylleae). *Lebrunia* is considered to be a close relative of *Endodesmia* (Stevens 2007a), and these genera together constitute tribe Endodesmieae. *Endodesmia* and *Lebrunia* are each monotypic and found in western tropical Africa. They possess a single, apical ovule, which in Calophyllaceae, is found only in tribe Endodesmieae (Stevens, 2007a). *Neotatea* was originally described as a genus in Bonnetiaceae (Maguire, 1972) and was once considered a species of *Bonnetia* (Steyermark, 1984). However, the placement of this species was problematic due to its possession of unilacunar nodes, latex, an indumentum, smooth stigmatic surfaces, and anther glands (Weitzman and Stevens, 1997). More recently, it was transferred to Clusiaceae s.l. (including Hypericaceae; Weitzman and Stevens, 1997) and then placed in tribe Calophylleae (Stevens 2007a). *Neotatea* possesses alternate leaves and winged seeds, which as noted previously, appear in only one Calophyllaceae clade. Thus, *Neotatea* is likely to be placed

somewhere among these taxa. This hypothesis is supported by Notis (2004) who found *Neotatea* to be sister to *Mahurea*, based on the shared presence of intruded axile placentae bordered by in-curled carpel walls and seeds with a vascularized wing that does not completely surround the seed.

Clusiaceae s.s.—Clusiaceae s.s. include two strongly supported (100 BP) subclades. The first contains all genera of the strictly New World tribe Clusieae. Clusieae are characterized by a lack of bud scales, prevalent dioecy, nonfasciculate androecia, and fleshy capsules with arillate seeds (Stevens, 2007a). Support for intergeneric relationships within Clusieae is generally weak. Morphological characters indicating phylogenetic relationships are mostly lacking, but characters of the aril, leaf bases, and sepals seem promising for future study. *Chrysochlamys* and *Clusia* are strongly supported as monophyletic (96 and 99 BP, respectively); *Dystovomita* and *Tovomita* are nonmonophyletic, but their monophyly could not be rejected (Fig. 2; Table 4). *Dystovomita paniculata* (Donn. Sm.) Hammel is weakly placed as sister to all other Clusieae and *Dystovomita* cf. *brasiliensis* D'Arcy is strongly (92 BP) embedded within a clade of *Tovomita* spp. The nonmonophyly of *Dystovomita* should be interpreted cautiously, however, because the name *D. brasiliensis* was applied to this taxon in the Flora Reserva Ducke (Ribeiro, 1999) with the hope of eventually comparing it to the type specimen. Unfortunately, the type appears to have been lost. Thus, we cannot validate the identification of our specimen and cannot know with certainty if *Dystovomita* sensu D'Arcy (1978) is nonmonophyletic. However, we can say that the taxon labeled as *D.* cf. *brasiliensis* in our analyses and the taxon listed as *D. brasiliensis* in the Flora Reserva Ducke are better attributed to *Tovomita*, a genus that may also be nonmonophyletic. *Tovomita weddelliana* Planch. & Triana is weakly placed (51 BP) as sister to *Clusia* rather than with the remaining *Tovomita* species (Fig. 2). Interestingly, *T. weddelliana* and species of *Clusia* are both found at relatively high altitudes in the Neotropics. All other members of the tribe are generally found in lowland tropical forests (Gustafsson et al., 2007). It is surprising that *Tovomitopsis* is not placed near *Chrysochlamys* because the two are morphologically similar and have often been considered synonymous (Hammel, 1999a). It may be that biogeography is more helpful than morphology for separating these two genera: *Chrysochlamys* occurs in Central America, the Caribbean, and northwestern South America; *Tovomitopsis* occurs in southeastern Brazil (Bittrich, 2010).

The second subclade in Clusiaceae s.s. includes all Garcinieae and Symphonieae. In contrast to Clusieae, this group is characterized by a fasiculate androecium (Stevens, 2007a; Sweeney, 2008). We provide the first strongly supported evidence that Symphonieae are monophyletic (84 BP; Fig. 2). Previous results have suggested that they may not be monophyletic (Gustafsson et al., 2002; Sweeney, 2008), which was surprising based on morphology. Members of this clade possess a branched style with each branch having no exposed stigmatic surface. Instead, there is a small apical pore in the stigma through which pollen enters the stigmatic cavity, which is unique in Malpighiales (Bittrich and Amaral, 1996). Within Symphonieae, *Pentadesma* and *Symphonia* are genera with Old World origins (Dick et al., 2003; Stevens, 2007a; Dick and Heuertz, 2008) and are successive sister groups to a clade containing the New World taxa *Lorostemon*, *Moronobea*, and *Platonia* plus the New Caledonian genus *Montrouziera*. The only genus in Symphonieae we were not able to include was the poorly known *Thysanostemon* from Guyana. *Thysanostemon* is certainly a member of the tribe Symphonieae, based on both vegetative and floral characteristics, and may be closely related to *Lorostemon.* These two genera have very elongated flower buds and pollen with supratectal elements, features not present in other Symphonieae (Maguire, 1964; Seetharam, 1985).

We found no support for a monophyletic Garcinieae. In contrast, Sweeney (2008) found Garcinieae to be strongly monophyletic using nuclear data (ITS and GBSSI). Additionally, members of Garcinieae possess several characters that unite the group: colleters, dioecy, capitate stigmas, eperulate buds (common), and introrse anthers (often). These features contrast with Symphonieae, which lack colleters, are hermaphroditic, and possess porose stigmas, perulate buds, and extrorse anthers (Stevens, 2007a; Sweeney, 2008). Relationships within *Garcinia* presented here are in agreement with Sweeney (2008). Importantly, we also find *Allanblackia* embedded within *Garcinia* (67 BP; Fig. 2). Support for this placement increases in the analysis of the reduced data set (82 BP; Fig. 4) and is strong in both BI analyses (100 PP). This corroborates the recommendation by Sweeney (2008) that *Allanblackia* be transferred to *Garcinia*. Furthermore, floral characters also support this placement: *Allanblackia* and all *Garcinia* species in this subclade have nectariferous appendages in the flower, unlike other members of *Garcinia* (Sweeney, 2008). However, a monophyletic *Garcinia* (excluding *Allanblackia*) could not be rejected by the combined data sets (Table 4).

Hypericaceae—Three strongly supported subclades (100 BP) are recovered in Hypericaceae corresponding to tribes Cratoxyleae, Hypericeae, and Vismieae (Stevens, 2007b; see also Wurdack and Davis, 2009). Cratoxyleae are sister to a strongly supported (97 BP) clade containing Hypericeae + Vismieae. Within Cratoxyleae, *Cratoxylum* and the monotypic *Eliea* are sister taxa. We sampled five of the six *Cratoxylum* species representing the three sections recognized by Gogelein (1967). This sampling allowed us to test his hypothesis of relationships in the group, which agreed with our results. Species in section *Isopterygium* [*Cratoxylum arborescens* (Vahl) Blume and *Cratoxylum glaucum* Korth.] are evergreen trees with straight secondary leaf venation and a wing that surrounds the seed. This section is sister to a clade containing sections *Cratoxylum* [*Cratoxylum sumatranum* (Jack) Blume and *Cratoxylum cochinchinense* Blume] and *Tridesmos* [*Cratoxylum formosum* (Jack) Benth. & Hook.f. ex Dyer and *Cratoxylum maingayi* Dyer (not sampled)], which are more or less deciduous trees with curved secondary leaf venation and a unilateral seed wing.

Vismieae have been previously treated by Bamps (1966) and most recently by Stevens (2007b). Bamps recognized three genera: *Harungana*, *Psorospermum*, and *Vismia*. Bamps' *Harungana* and *Psorospermum* are found in Africa and Madagascar, while his *Vismia* is divided into two subgenera, *Vismia* and *Afrovismia,* found in the Americas and Africa, respectively. More recently, Stevens (2007b) considered the tribe to have only two genera, *Harungana* and *Vismia*, distributed in the Old World (Africa and Madagascar) and New World (Central and South America), respectively. Formal taxonomic changes however, were not made to reflect this viewpoint. Morphological characteristics that Stevens used to separate these two genera included the fusion of bracts to the pedicels (unfused in *Vismia* vs. fused in *Harungana*) and staminode pubescence (pubescent in *Vismia* vs. glabrous in *Harungana*; Bamps, 1966; Stevens, 2007b).

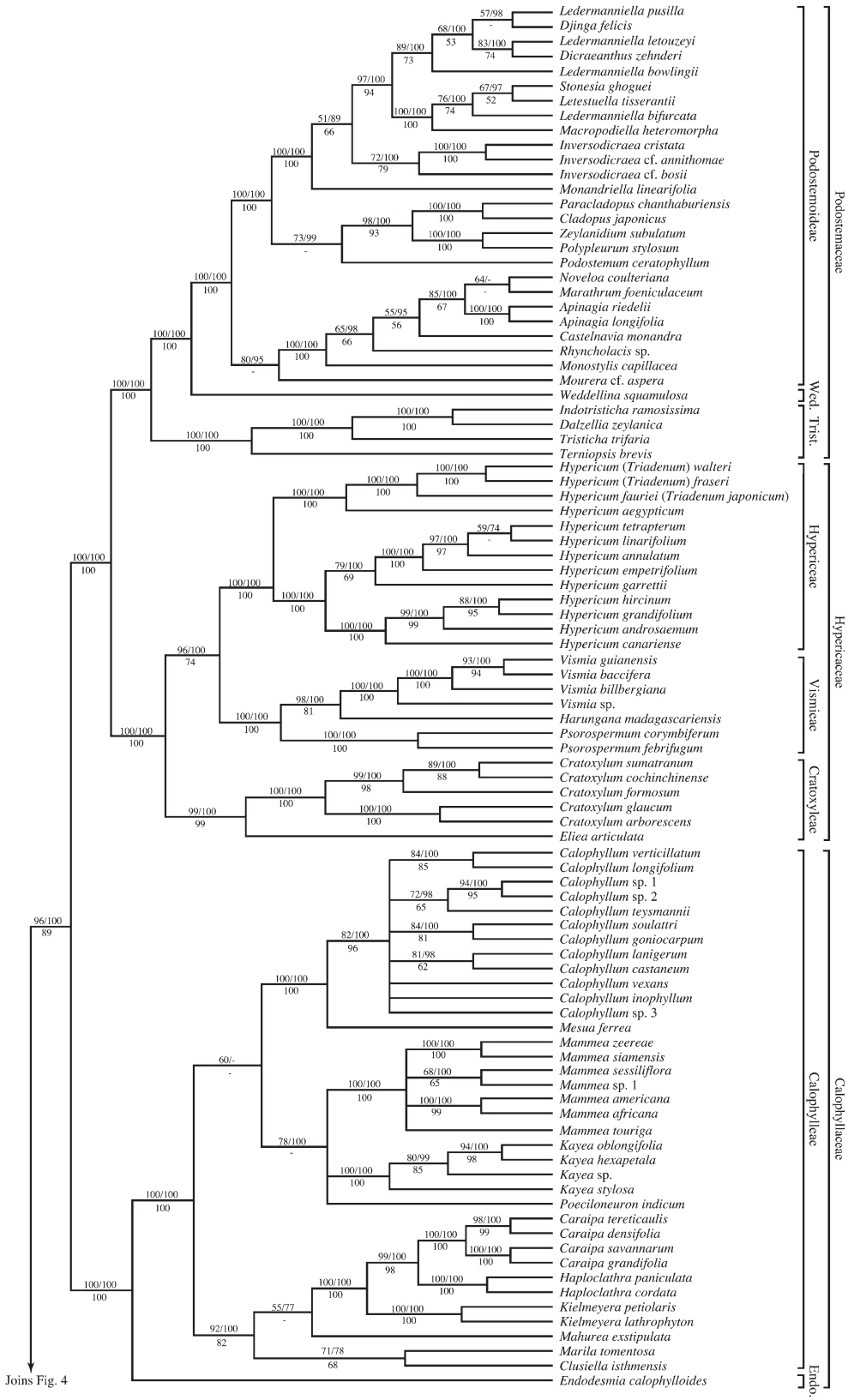


Fig. 3. Fifty percent maximum likelihood (ML) majority-rule consensus tree of the clusioid clade based on the combined four-gene reduced data set (ntax = 169, missing data = 8.4%). Support values ≥ 50% are indicated. Values above branches are ML bootstrap values (left) and Bayesian inference posterior probabilities converted to percentages (right). Maximum parsimony bootstrap values are given below each branch. A hyphen indicates that the node was not present in a particular analysis. Endo., Endodesmieae; Trist., Tristichoideae; Wed., Weddellinoideae. Revised names for Hypericeae genera are given; former names are included in parentheses. Tree continued in Fig. 4.

Our results indicate that neither of these classifications reflect phylogenetic relationships (Fig. 1). *Harungana* sensu Stevens (i.e., Old World Vismieae) is paraphyletic and includes American Vismieae. *Vismia* subgenus *Afrovismia* sensu Bamps is also not monophyletic: *Vismia guineensis* (L.) Choisy is embedded in *Psorospermum*, and *Vismia rubescens* Oliv. is sister to *Harungana madagascariensis* Poir. We believe that the sampling here is too preliminary to propose taxonomic revisions. However, restricting *Harungana* to include only *H. madagascariensis* (the type species of the genus) and *Vismia rubescens*, and including all other African and Malagasy species in an extended *Psorospermum* is a reasonable solution if these relationships are further corroborated by additional data. Morphological distinctions between these groups are lacking, but characters of the cotyledons and the position of the bracteoles on the inflorescence may be useful.

Within the third subclade, Hypericeae, *Hypericum* sensu Robson (1977 onward) and Stevens (2007b) is not monophyletic (Fig. 1; Table 4). These authors recognize four small genera (*Lianthus*, *Santomasia, Thornea*, and *Triadenum*) as separate from *Hypericum*, primarily based on the possession of staminodes, which are mostly absent in *Hypericum* (Robson, 1972, 1977; Stevens 2007b). White, pink, or reddish petals further separate *Lianthus*, *Thornea*, and *Triadenum* from *Hypericum*, which has yellow petals (Breedlove and McClintock, 1976; Robson, 1981, 2001; Stevens 2007b). However, in our analyses, *Santomasia, Thornea*, and *Triadenum* are well supported as members of a subclade of *Hypericum* (83 BP). This result does not agree with a recent morphological analysis of Hypericaceae where only *Santomasia* was found to be embedded within *Hypericum* (Nürk and Blattner 2010). The distribution of staminodes in the androecium of Hypericeae species offers additional support for our result. As stated previously, staminodes are present in *Lianthus*, *Santomasia, Thornea*, and *Triadenum*, as well as in all members of Cratoxyleae and Vismieae. However, staminodes are largely absent in *Hypericum*, except in sections *Adenotrias* and *Elodes* (represented in our study by *H. aegypticum* L. and *H. elodes* L., respectively [Robson, 1996; Fig. 1]). All Hypericeae taxa with staminodes occur in the same *Hypericum* subclade. We were unable to sample *Lianthus*, but it is very likely that this monotypic genus is also a member of this subclade because it possesses staminodes and shows strong affinities with *Thornea* and *Triadenum* (Robson, 2001). Given the embedded position of these smaller genera in *Hypericum*, we propose that *Lianthus*, *Santomasia*, *Thornea*, and *Triadenum* be reinstated as members of *Hypericum* (Table 1). These taxa have all previously been described as members of *Hypericum*, and as such, appropriate names are available (Table 5).

Podostemaceae—Our results generally agree with previous studies but include much denser character and taxon sampling (Kato et al., 2003; Kita and Kato, 2004a, b; Moline et al., 2006, 2007; Koi et al., 2008, 2009; Pfeifer et al., 2009; Thiv et al., 2009; Koi and Kato, 2010; Tippery et al., in press). We recognize the three subfamilies proposed by Engler (1930), which are each strongly supported (100 BP) as monophyletic here and elsewhere (Kita and Kato, 2001; Moline et al., 2007).

Tristichoideae are strongly supported as monophyletic (100 BP) and are sister to a clade containing subfamilies Podostemoideae + Weddellinoideae. Tristichoideae have tricarpellate ovaries and pantoporate pollen, in contrast to Podostemoideae and Weddellinoideae, which have bicarpellate ovaries and mostly tricolporate or tricolpate pollen (Kita and Kato, 2001; Cook and Rutishauser, 2007). Within the Tristichoideae clade, bootstrap support for *Dalzellia* + *Indotristicha* is weak (58 BP; Fig. 1), which is surprising because this clade has received strong support elsewhere (Koi et al., 2009). The *Dalzellia* + *Indotristicha* clade is also supported by morphology: a leafy cupule surrounding the flower bud is a putative synapomorphy for this clade (Koi et al., 2009). The only genus in this subfamily we were unable to include was the recently described *Cussetia,* which shows affinities to *Terniopsis* and *Tristicha* (Kato, 2006, 2009; Koi et al., 2009).

Podostemoideae are strongly supported as monophyletic (100 BP) and are characterized by the presence of a spathella that encloses the flower bud prior to anthesis. Its sister clade, Weddellinoideae, differs from Podostemoideae by the absence of a spathella and the presence of a distinct perianth, which are likely plesiomorphic characters shared with Tristichoideae (Kita and Kato, 2001). For the first time, we present evidence that the monotypic New World genus *Diamantina* is sister to the remaining Podostemoideae (Fig. 1). Its position is poorly supported (56 BP), likely because we were only able to obtain a portion of *matK* for this taxon. However, previous authors have hypothesized a similar phylogenetic placement of *Diamantina* (Philbrick et al., 2004b; Rutishauser et al., 2005; Koi et al., 2006). Among the remaining Podostemoideae, there are two subclades, an exclusively New World clade represented by *Apinagia*, *Castelnavia*, *Marathrum*, *Monostylis*, *Mourera*, *Noveloa*, and *Rhyncholacis* (Fig. 1) and a primarily Old World clade containing all other genera sampled here. The two New World genera, *Ceratolacis* and *Podostemum*, are an exception and are embedded within this primarily Old World clade. Kita and Kato (2001) showed that *Podostemum* was more closely related to the Old World members of Podostemoideae, but our results are the first strong evidence that *Ceratolacis* belongs to the Old World clade (94 BP; Fig. 1). This mostly Old World clade is loosely characterized by the possession of an andropodium, one or two stamens per flower, and pollen dyads (which are sometimes secondarily lost). The strictly New World clade is characterized by often having several free stamens per flower and pollen in monads (Cook and Rutishauser, 2007).

Much greater taxon sampling is needed in the New World Podostemoideae clade before evolutionary, taxonomic, and biogeographical patterns can be inferred (see also Tippery et al., in press). In particular, sampling in the genera *Apinagia*, *Marathrum,* and *Rhyncholacis* will need to be improved to further determine their limits. Furthermore, the New World genera *Cipoia*, *Macarenia*, and *Wettsteiniola* have never been included in a molecular phylogenetic study. *Macarenia* and *Wettsteiniola* are likely members of this clade based on morphological analysis (C.T. Philbrick, unpublished data). *Cipoia,* however, shares traits with members of the primarily Old World clade, such as pollen in dyads (Philbrick et al., 2004b; Bove et al., 2006). All New World taxa with dyad pollen sampled to date have been placed in the primarily Old Word clade (i.e., *Ceratolacis* and *Podostemum*).

The mostly Old World Podostemoideae clade is composed of four subclades whose interrelationships are unresolved: (1) the New World genus *Podostemum*; (2) the Malagasy genera *Endocaulos* and *Thelethylax*; (3) the Asian and Australian genera *Cladopus*, *Griffithella*, *Hanseniella*, *Hydrobryum*, *Paracladopus*, *Polypleurum*, *Thawatchaia*, and *Zeylanidium*; and (4) the Brazilian genus *Ceratolacis* plus the African genera *Dicraeanthus*, *Djinga*, *Inversodicraea*, *Ledermanniella*, *Leiothylax*, *Letestuella*, *Macropodiella*, *Monandriella*, and *Stonesia*. *Podostemum* is a

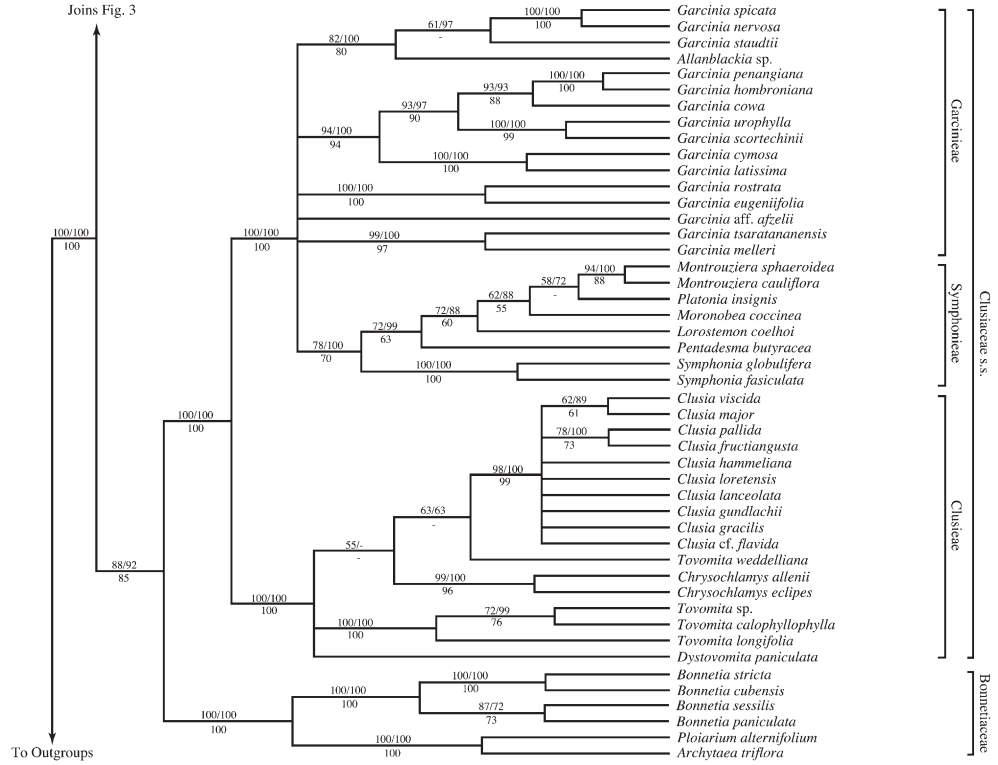


Fig. 4. Continuation of Fig. 3. Fifty percent maximum likelihood (ML) majority-rule consensus tree of the clusioid clade based on the combined four-gene reduced data set (ntax = 169, missing data = 8.4%). Outgroups removed to show only the clusioid clade.

weakly supported (53 BP) clade in our ML expanded data set analysis, but is strongly supported by BI (97 PP). The latter results are corroborated by previous studies, which provide strong morphological and molecular evidence that *Podostemum* is monophyletic (Philbrick and Novelo, 2004; Moline et al., 2006). Although *Podostemum* forms a polytomy with the three other subclades in our expanded analysis (Fig. 1), the reduced analysis provides moderate support (73 BP; Fig. 3) for it being the sister group to the Asian and Australian taxa *Cladopus*, *Paracladopus*, *Polypleurum*, and *Zeylanidium*. In contrast, Moline et al. (2007) placed *Podostemum* sister to a clade of the African/Malagasy taxa, although with weak bootstrap support (Moline et al., 2007).

We were unable to obtain material of the Malagasy taxa *Endocaulos* and *Thelethylax* and were limited to available *matK* sequence data from GenBank (Appendix 1). Recent studies (Moline et al., 2007; Pfeifer et al., 2009) used these same sequences in their analyses and found a sister group relationship between these Malagasy taxa and the African Podostemoideae

TABLE 4. Log likelihoods of optimal tree, constraint trees, and results from AU topology tests.

	Reduced data set		Expanded data set	
Topology	Likelihood	*P*	Likelihood	*P*
Optimal	−72673.656770	83.13	−78244.206942	83.76
80 BP ML constraints				
matK	−72690.359865	6.37	−78261.127486	13.43
ndhF	−72705.056806	2.98 *	−78270.805141	7.26
rbcL	−72673.656776	81.23	−78277.842951	7.52
matR	−72727.284331	0.04 *	−78259.285849	14.04
Monophyly constraints				
Clusiaceae s.l.	−72693.412489	5.91	−78265.061339	5.63
Dystovomita	—	—	−78257.353943	9.47
Garcinia	−72693.567356	12.26	−78263.982016	14.12
Harungana	−72686.106027	4.93 *	−78260.341692	3.25 *
Hypericum	−72710.968640	0.11 *	−78285.931353	0.66 *
Ledermanniella	−72754.773013	0.02 *	−78321.085180	0.19 *
Tovomita	−72677.789966	42.70	−78260.488872	5.11
Zeylanidium	—	—	−78336.063417	0 *
Alternate MP placement				
Mourera	−72676.969678	32.69	−78247.380007	55.47

Notes: P values less than 5% (marked with a "*") indicate topologies that differ significantly from the best tree.

TABLE 5. Proposed taxonomic changes for Hypericaceae.

Synonym in use prior to this study	Proposed name
Lianthus ellipticifolius (H.L. Li) N. Robson	*Hypericum ellipticifolium* H.L. Li
Santomasia steyermarkii (Standl.) N. Robson	*Hypericum steyermarkii* Standl.
Thornea calcicola (Standl. & Steyerm.) Breedlove & E.M. McClint.	*Hypericum calcicola* Standl. & Steyerm.
Thornea matudae (Lundell) Breedlove & E.M. McClint.	*Hypericum matudae* Lundell
Triadenum breviflorum (Wall. ex Dyer) Y. Kimura	*Hypericum breviflorum* Wall. ex Dyer
Triadenum fraseri (Spach) Gleason	*Hypericum fraseri* (Spach) Steudel
Triadenum japonicum (Blume) Makino	*Hypericum fauriei* R. Keller
Triadenum tubulosum (Walter) Gleason	*Hypericum tubulosum* Walter
Triadenum virginicum (L.) Raf.	*Hypericum virginicum* L.
Triadenum walteri (J.F. Gmel.) Gleason	*Hypericum walteri* J.F. Gmel.

clade. They proposed that completely or partially inverted flower orientation in bud might be a synapomorphy for the African/Malagasy clade (Grob et al., 2007; Moline et al., 2007). However, we find that the New World *Ceratolacis*, rather than the Malagasy taxa, are sister to the African clade, albeit with poor support (52 BP). Although *Ceratolacis* shares two stamens, an andropodium, and dyad pollen with many members of the primarily Old World clade, it also shares an asymmetrically placed stipule and an andropodial tepal with some members of *Podostemum* (Philbrick et al., 2004a, b) and forms a clade with *Podostemum* in a morphological analysis of the family (C.T. Philbrick, unpublished data).

We present new relationships and increased support within the clade of African taxa recently studied by Thiv et al. (2009). The monotypic *Monandriella* is weakly supported (57 BP) as sister to the remaining taxa from mainland Africa rather than embedded within the clade as in Thiv et al. (2009). Thiv et al. proposed that this genus might form a clade with other African taxa that shed their pollen in monads [their "*Ledermanniella*-monad" group; here represented by *Ledermanniella bifurcata* (Engler) C. Cusset, *Leiothylax, Letestuella, Macropodiella and Stonesia*; Fig. 1]. Our data do not support this suggestion, although our placement of *Monandriella* does support maintaining it as a separate genus. We also find strong support (86 BP) for a monophyletic *Inversodicraea* (*Ledermanniella* subgenus *Phyllosoma* sensu C. Cussett), for which there was no previous molecular support, confirming the separation of *Inversodicraea* from *Ledermanniella* s.l. sensu Thiv et al. The *Inversodicraea* clade is also supported by morphology: these taxa possess stem scales (Cusset, 1983; Thiv et al., 2009). Two clades containing taxa whose pollen is shed primarily in monads (mentioned above, excluding *Monandriella*) or dyads [here represented by *Dicraeanthus*, *Djinga*, *Ledermanniella bowlingii* (J.B. Hall) C. Cusset, *Ledermanniella letouzeyi* C. Cusset, *Ledermanniella linearifolia* Engl., and *Ledermanniella pusilla* (Warm.) C. Cusset in Fig. 1] are also moderately to strongly supported here but not in Thiv et al. (2009). Pollen shed in monads appears only in a few subclades in the mostly Old World clade, particularly among the mainland African taxa, suggesting that other African members that possess monads not sampled here (e.g., *Winklerella* and *Zehnderia*) belong among these taxa. Furthermore, we find strong support that the genus *Ledermanniella* s.s. as proposed by Thiv et al. (2009; former *Ledermanniella* subgenus *Ledermanniella* minus *Monandriella* sensu C. Cusset) is not monophyletic (Fig. 1; Table 4).

Within the Asian Podostemoideae clade, we show that *Zeylanidium* is not monophyletic (Fig. 1; Table 4): *Zeylanidium subulatum* (Gardner) C. Cusset is sister to *Polypleurum* (100 BP) and *Zeylanidium lichenoides* Engl. is sister to *Griffithella* (100 BP). Koi and Kato (2010) also demonstrated the nonmonophyly of *Zeylanidium*, but *Griffithella* was not included in their study. We believe that the sampling here is too preliminary to consider taxonomic changes.

Conclusions and future directions—The phylogeny of the clusioid clade presented here provides a greatly improved understanding of the evolutionary history of this morphologically and ecologically diverse clade. Taxon sampling and resolution within the clade is greatly improved compared to previous studies, which has allowed us to propose a more refined classification of the group. In the future, we will concentrate on two main areas of research using the clusioid clade as a study system.

Increased taxon and character sampling—Many important clusioid taxa have not been sampled with molecular data, and key areas in our phylogeny remain unresolved or poorly supported. To address these issues further, future taxon sampling should focus on unsampled genera, as well as on expanding sampling of distinct morphological or biogeographical groups within several larger genera (e.g., *Apinagia*, *Calophyllum*, *Chrysochlamys*, *Clusia*, *Garcinia*, *Hypericum*, *Ledermanniella*, *Mammea*, and *Marathrum*). In several genera, such as *Chrysochlamys* and *Clusia*, particularly in Andean countries, the alpha taxonomy is poorly known, and many species are undescribed. In these groups, revisionary taxonomic studies should be well integrated with phylogenetic investigations. Additionally, obtaining well-sampled phylogenies of *Calophyllum, Hypericum*, and *Mammea* will be important for future biogeographic studies of the clusioid clade because the early biogeographic histories of these widely distributed genera are unknown and are critical to assessing ancestral areas within the clusioids (see below). Character sampling, in addition to taxon sampling, should also be increased to help provide better resolution and support in various areas of the tree. Increased sampling of the plastid and mitochondrial genomes will be valuable, but nuclear markers should also be used in future studies to represent the evolutionary history of all three genomes. A particularly useful marker may be the low-copy nuclear gene *PHYC*, which has been shown to be very informative at both the familial and ordinal levels in Malpighiales (Davis et al., 2002; Davis and Chase, 2004; Kathriarachchi et al., 2005; Samuel et al., 2005; Wurdack and Davis, 2009; B.R. Ruhfel, unpublished data).

Biogeography—The clusioids offer a unique opportunity to study the biogeography of tropical angiosperms with Gondwanan distributions because they are of ancient origin and possess

a pantropical distribution. Fossil representatives of the clade are known from the Cretaceous (~90 Ma; Crepet and Nixon, 1998) and the Eocene (~45 Ma; Jan-du-Chene et al., 1978) and their stem group age dates to the mid-Cretaceous (99–109 Ma; Davis et al., 2005). The clusioids are prominently featured in the classic work by Raven and Axelrod (1974), which integrated plate tectonics with angiosperm evolution and biogeography. Raven and Axelrod hypothesized that various clusioid clades date back to Gondwanan times when Africa and South America were in close proximity to one another. More recent analyses, however, have indicated that at least some intercontinental disjunctions within this group are far more recent and are more consistent with long-distance dispersal rather than ancient Gondwanan vicariance (Dick et al., 2003; Kita and Kato, 2004b). Biogeographical studies of pantropical groups are few (see Clayton et al., 2009 and references therein) and are needed to increase our understanding of the relative roles of ancient vicariance and more recent dispersal in the assembly of the modern tropical biota (Pennington and Dick, 2004). Determining which of these two factors is most plausible for the many intercontinental disjunctions implied in our trees is testable and is a major focus of our future efforts.

While many disjunctions involving former Gondwanan landmasses can now be localized in our topology, an assessment of the influence of ancient vicariance vs. more recent dispersal cannot be determined until we know where and when these events occurred. This information can be gleaned from ancestral area reconstructions and divergence time estimation. It is of utmost importance that these analyses include appropriately placed fossils. *Paleoclusia chevalieri* Crepet & Nixon dates back to the Turonian, (~90 Ma), is among the oldest rosid macrofossils, and has been attributed to Clusiaceae s.s. (Crepet and Nixon, 1998). Its exact phylogenetic placement within the clusioid clade, however, remains to be determined. Analysis of a data set containing both molecular and morphological data may allow us to place this and other critical clusioid taxa that lack molecular data (Wiens, 2009; B.R. Ruhfel, P.F. Stevens, and C.C. Davis, unpublished manuscript). The placement of this fossil will be important for estimating divergence times in the clusioid clade as well as in the broader rosid clade. A further benefit of estimating divergences times within this clade concerns the response of tropical angiosperms to the Cretaceous–Tertiary (K-T) mass extinction event. The ancient age of the clusioids makes this group amenable to examine what effect, if any, the K-T mass extinction had on tropical rain forest diversity. A biogeographical study of the clusioids (B.R. Ruhfel, C.P. Bove, C.T. Philbrick, and C.C. Davis, unpublished manuscript) will enable the exploration of these important topics and will help to clarify the origin and maintenance of diversity in modern tropical rain forests.

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APPENDIX 1. Voucher information and GenBank accessions for sequences used in this study. New data have GenBank numbers beginning with HQ (HQ331542–HQ332128), and accessions in brackets are from a different voucher source. A dash (—) indicates that the sequence was unavailable. Herbaria acronyms follow Holmgren and Holmgren (1998 [continuously updated]).

FAMILY. Species, *voucher* (herbarium), GenBank accessions: *matK*, *ndhF*, *rbcL*, *matR*.

ACHARIACEAE. ***Acharia tragodes*** Thunb., *Cloete s.n.* (BOL), EF135500, AY425028, AF206728, AY674472.

BALANOPACEAE. ***Balanops vieillardi*** Baill., *Chase 1816* (K), EF135505, AY425032, AF089760, AY674479.

BONNETIACEAE. ***Archytaea triflora*** Mart., *Kubitzki & Feuerer 97-26* (HBG), HQ331545, AY425029, AY380342, AY674475; ***Bonnetia ahogadoi*** (Steyer.) A.L. Weitzman & P.F. Stevens, *Weitzman et al. 409* (K), HQ331546, AY425035, HQ332007, —; ***Bonnetia cubensis*** (Britton) R.A. Howard, *J. Gutierrez et al. HAJB 81795* (WIS), HQ331547, HQ331846, HQ332008, HQ331702; ***Bonnetia paniculata*** Spruce ex Benth., *P. Berry 7789* (MICH), HQ331548, HQ331847, HQ332009, HQ331703; ***Bonnetia roraimae*** Oliv., *Weitzman et al. 402* (K), —, HQ331848, AJ402930, —; ***Bonnetia sessilis*** Benth., *Berry s.n. 25.7.98* (MO), EF135509, HQ331849, HQ332010, EF135292; ***Bonnetia steyermarkii*** Kobuski, *Weitzman et al. 403* (K), —, HQ331850, HQ332011, HQ331704; ***Bonnetia stricta*** (Nees) Nees & Mart., *Amorim 3958* (CEPEC), HQ331549, HQ331851, HQ332012, HQ331705; ***Bonnetia tepuiensis*** Kobuski & Steyerm., *P. Berry 7788* (MICH), —, HQ331852, HQ332013, —; ***Ploiarium alternifolium*** Melchior, *Sugumaran 165* (US), FJ669999, FJ670063, FJ670161, FJ670352.

CALOPHYLLACEAE. ***Calophyllum brasiliense*** Cambess., *C. Notis 387* (FLAS), HQ331550, HQ331853, —, HQ331706; ***Calophyllum castaneum*** P.F. Stevens, *Ruhfel 111* (A), HQ331551, HQ331854, HQ332014, HQ331707; ***Calophyllum goniocarpum*** P.F. Stevens, *F. Damon 318* (MO), HQ331552, HQ331855, HQ332015, HQ331708; ***Calophyllum inophyllum*** L., *Ruhfel 115* (A), HQ331553, HQ331856, HQ332016, HQ331709; ***Calophyllum lanigerum*** Miq., *Ruhfel 104* (A), HQ331554, HQ331857, HQ332017, HQ331710; ***Calophyllum longifolium*** Willd., *Aguilar 11657* (NY), HQ331555, HQ331858, HQ332018, HQ331711; ***Calophyllum soulattri*** Burm. f., *Chase 1217* (K), HQ331556, AY425037, [*F. Damon 320* (MO), AY625021], AY674484; ***Calophyllum sp. 1***, *Ruhfel 108* (A), HQ331557, HQ331859, HQ332019, HQ331712; ***Calophyllum sp. 2***, *Ruhfel 113* (A), HQ331558, HQ331860, HQ332020, HQ331713; ***Calophyllum sp. 3***, *Ruhfel 114* (A), HQ331559, HQ331861, HQ332021, HQ331714; ***Calophyllum teysmannii*** Miq., *Ruhfel 112* (A), HQ331560, HQ331862, HQ332022, HQ331715; ***Calophyllum verticillatum*** P.F. Stevens, J. *Rabenantoandro et al. 733* (MO), HQ331561, HQ331863, HQ332023, HQ331716; ***Calophyllum vexans*** P.F. Stevens, *F. Damon 321* (MO), HQ331562, HQ331864, HQ332024, HQ331717; ***Caraipa densifolia*** Mart., *C. Grandez 16239* (FLAS), HQ331563, HQ331865, AY625012, HQ331718; ***Caraipa grandifolia*** Mart., *C. Grandez 16244* (FLAS), HQ331564, HQ331866, HQ332025, HQ331719; ***Caraipa savannarum*** Kubitzki, *G. Aymard s.n.* (PORT), HQ331565, HQ331867, HQ332026, HQ331720; ***Caraipa tereticaulis*** Tul., *Vormisto 578* (AAU), HQ331566, HQ331868, HQ332027, HQ331721; ***Clusiella isthmensis*** Hammel, *M. Whitten 2657* (FLAS), HQ331585, HQ331889, AY625019, HQ331738; ***Endodesmia calophylloides*** Benth., *Burgt 762* (WAG), FJ670005, FJ670069, FJ670163, FJ670356; ***Haploclathra cordata*** R.Vásquez, *C. Grandez 16237* (FLAS), HQ331613, HQ331918, AY625017, HQ331764; ***Haploclathra paniculata*** Benth., *C. Grandez 16246* (FLAS), HQ331614, HQ331919, HQ332068, HQ331765; ***Kayea elmeri*** Merr., *Ruhfel 110* (A), HQ331636, —, HQ332086, HQ331784; ***Kayea hexapetala*** Pierre, *Ruhfel 119* (A), HQ331637, HQ331939, HQ332087, HQ331785; ***Kayea oblongifolia*** Ridl., *Ruhfel 116* (A), HQ331638, HQ331940, HQ332088, HQ331786; ***Kayea sp.***, *E. Wood and G. A. Teck 5500* (A), HQ331639, HQ331941, HQ332089, HQ331787; ***Kayea stylosa*** Thw., *Kostermans 11106* (HUH), HQ331640, HQ331942, AY625025, HQ331788; ***Kielmeyera lathrophyton*** Saddi, *F. Feres s.n.* (UEC), HQ331641, HQ331943, AY625015, HQ331789; ***Kielmeyera petiolaris*** Mart., *F. Feres 75* (UEC), HQ331642, HQ331944, AY625016, HQ331790; ***Mahurea exstipulata*** Benth., *Kubitzki et al. 97-*

27 (HBG), HQ331650, HQ331954, AY625018, HQ331799; ***Mammea africana*** Sabine, *D. Kenfack 2055* (MO), HQ331651, HQ331955, HQ332098, HQ331800; ***Mammea americana*** L., *C. Notis 392* (FLAS), HQ331652, HQ331956, AY625029, HQ331801; ***Mammea sessiliflora*** Planch. & Triana, *McPherson 18377* (MO), HQ331653, HQ331957, AY625027, HQ331802; ***Mammea siamensis*** T. Anderson, *Chase 1216* (K), FJ670006, FJ670070, AY625028, FJ670357; ***Mammea sp. 1***, *P. Sweeney 1305* (MO), HQ331654, HQ331958, HQ332099, HQ331803; ***Mammea sp. 2***, *T.G. Laman et al. TL 727* (A), HQ331655, HQ331959, HQ332100, —; ***Mammea touriga*** (C.T. White & W.D. Francis) L.S. Sm., *H. van der Werff and B. Gray 17055* (MO), HQ331656, HQ331960, HQ332101, HQ331804; ***Mammea zeereae*** P.F. Stevens, *P. Sweeney 1273* (MO), HQ331657, HQ331961, HQ332102, HQ331805; ***Marila laxiflora*** Rusby, *van der Werff et al. 16246* (MO), HQ331659, HQ331963, —, HQ331807; ***Marila tomentosa*** Poepp. & Endl., *van der Werff et al. 16215* (MO), HQ331660, HQ331964, AY625010, HQ331808; ***Mesua ferrea*** L., *M. Sugumaran et al. SM 120* (KLU), HQ331661, HQ331965, [*C. Notis 390* (FLAS), AY625024], HQ331809; ***Poeciloneuron indicum*** Bedd., *U. Ghate s.n.* (FLAS), HQ331673, HQ331977, AY625023, HQ331819.

CARYOCARACEAE. ***Caryocar glabrum*** Pers., *Mori 22997* (NY), EF135515, AY425039, Z75671, AY674486.

CELASTRACEAE. ***Celastrus orbiculatus*** Thunb., *Simmons 1773* (BH), EF135517, FJ670145, AY788194, EF135295.

CENTROPLACACEAE. ***Centroplacus glaucinus*** Pierre, *White 128, ser. 1* (MO), FJ670002, FJ670066, AY663646, FJ670355.

CHRYSOBALANACEAE. ***Chrysobalanus icaco*** L., *Wurdack D711* (US), EF135519, FJ670067, L11178, AY674491.

CLUSIACEAE S.S. ***Allanblackia sp.***, *E. Ndive s.n.* (YU), HQ331542, HQ331843, HQ332004, HQ331699; ***Chrysochlamys allenii*** (Maguire) Hammel, *R. Kriebel 2289* (INB), HQ331569, HQ331871, HQ332030, HQ331723; ***Chrysochlamys eclipes*** L.O. Williams, *BCI 158121* (STRI), HQ331570, HQ331872, HQ332031, HQ331724; ***Chrysochlamys grandifolia*** (L.O. Williams) Hammel, *R. Aguilar ra12291* (NY), —, HQ331873, HQ332032, HQ331725; ***Chrysochlamys silvicola*** (Hammel) Hammel, *B. Hammel 25293* (MO), HQ331571, HQ331874, —, HQ331726; ***Chrysochlamys skutchii*** Hammel, *R. Aguilar ra12292* (NY), HQ331572, HQ331875, —, —; ***Clusia* cf. *flavida*** (Benth.) Pipoly, *M. H. G. Gustafsson 454* (AAU), HQ331575, HQ331878, HQ332035, HQ331728; ***Clusia clusioides*** (Griseb.) D'Arcy, *M. H. G. Gustafsson 272* (NY), —, HQ331879, AF518388, HQ331729; ***Clusia fructiangusta*** Cuatrec., *M. H. G. Gustafsson 485* (AAU), HQ331576, HQ331880, HQ332036, HQ331730; ***Clusia gracilis*** Standl., *Ruhfel 23* (A), HQ331577, HQ331881, HQ332037, HQ331731; ***Clusia gundlachii*** Stahl, *Chase 341* (NCU), EF135520, AY425041, Z75673, AY674493; ***Clusia hammeliana*** Pipoly, *M. H. G. Gustafsson 451* (AAU), HQ331578, HQ331882, HQ332038, HQ331732; ***Clusia lanceolata*** Cambess., *C. Notis 389* (FLAS), HQ331579, HQ331883, HQ332039, HQ331733; ***Clusia loretensis*** Engl., *M. H. G. Gustafsson 500* (AAU), HQ331580, HQ331884, HQ332040, HQ331734; ***Clusia major*** L., *M. H. G. Gustafsson 396* (AAU), HQ331581, HQ331885, HQ332041, HQ331735; ***Clusia pallida*** Engl., *M. H. G. Gustafsson 464* (AAU), HQ331582, HQ331886, HQ332042, HQ331736; ***Clusia rosea*** Jacq., *Kent s.n.* (A), HQ331583, HQ331887, HQ332043, —; ***Clusia viscida*** Engl., *M. H. G. Gustafsson 444* (AAU), HQ331584, HQ331888, HQ332044, HQ331737; ***Dystovomita* cf. *brasiliensis*** D'Arcy, *Sothers 452* (UEC), —, —, AF518387, —; ***Dystovomita paniculata*** (Donn. Sm.) Hammel, *B. Hammel 25295* (MO), HQ331594, HQ331897, [*B. Hammel 22728* (INB), HQ332051], HQ331746; ***Garcinia* aff. *afzelii*** Engl., *P. W. Sweeney 1411* (MO), HQ331595, HQ331898, HQ332052, HQ331747; ***Garcinia conrauana*** Engl., *S. Moses 961* (MO), —, HQ331899, HQ332053, —; ***Garcinia cowa*** Roxb., *M. Sugumaran et al. SM 146* (KLU), HQ331596, HQ331900, HQ332054, HQ331748; ***Garcinia cymosa*** (K. Schum.) I.M.Turner & P.F.Stevens, *P. Sweeney 1000* (MO), HQ331597, HQ331901, [*T. Motley s.n.* (AAU) AF518379], HQ331749; ***Garcinia eugeniifolia*** Wall. ex T. Anderson, *P. W. Sweeney 985* (MO), HQ331598, HQ331902, HQ332055, HQ331750; ***Garcinia hessii*** (Britton) Alain, *Axelrod 4537* (UPR), EF135543, —, AJ402952, DQ110341; ***Garcinia hombroniana*** Pierre, *M. Sugumaran et al. SM 124* (KLU), HQ331599, HQ331903, HQ332056, HQ331751; ***Garcinia intermedia*** (Pittier) Hammel, *M.J Balick 3570* (GH), HQ331600, HQ331904, —, HQ331752; ***Garcinia latissima*** Miq., *Chase 2100* (K), FJ670008, FJ670072, AF518386, FJ670359; ***Garcinia livingstonei*** T. Anderson, *P. Sweeney 1007* (MO), —, HQ331905, —, HQ331753; ***Garcinia macrophylla*** Mart., *Chase 1219* (K), —, FJ670073, FJ670165, FJ670360; ***Garcinia mangostana*** L., *Kent s.n.* (A), HQ331601, HQ331906, HQ332057, —; ***Garcinia mannii*** Oliver, *G. Walters et al. 604* (MO), HQ331602, HQ331907, —, HQ331754; ***Garcinia melleri*** Baker, *J. Rabenantoandro and G. McPherson 689* (MO), HQ331603, HQ331908, HQ332058, HQ331755; ***Garcinia nervosa*** Miq., *Ruhfel 106* (A), HQ331604, HQ331909, HQ332059, HQ331756; ***Garcinia penangiana*** Pierre, *Ruhfel 118* (A), HQ331605, HQ331910, HQ332060, HQ331757; ***Garcinia rostrata*** Hassk. ex Hook. f., *P. W. Sweeney 1071* (MO), HQ331606, HQ331911, HQ332061, HQ331758; ***Garcinia scortechinii*** King, *P. W. Sweeney 994* (MO), HQ331607, HQ331912, HQ332062, HQ331759; ***Garcinia spicata*** Hook. f., *C. Notis 388* (FLAS), HQ331608, HQ331913, HQ332063, HQ331760; ***Garcinia staudtii*** Engl., *P. Sweeney et al. 1445* (MO), HQ331609, HQ331914, HQ332064, HQ331761; ***Garcinia tsaratananensis*** (H. Perrier) P. Sweeney & Z.S. Rogers, *P. Sweeney 1232* (MO), HQ331610, HQ331915, HQ332065, HQ331762; ***Garcinia urophylla*** Scort. ex King, *P. W. Sweeney 1081* (MO), HQ331611, HQ331916, HQ332066, HQ331763; ***Lorostemon coelhoi*** Paula, *V. Bittrich 95-170* (UEC), HQ331648, HQ331952, [*Assuncão 492* (UEC), AF518401], HQ331797; ***Montrouziera cauliflora*** Planch. & Triana, *Lowry 5601* (MO), FJ670007, FJ670071, FJ670164, FJ670358; ***Montrouziera sphaeroidea*** Planch. ex Planch. & Triana, *K. Cameron 981* (NY), HQ331664, HQ331968, [*Cameron 981* (NY), AF518390], HQ331812; ***Moronobea coccinea*** Aubl., *SM 24698* (NY), HQ331665, HQ331969, AF518378, HQ331813; ***Pentadesma butyracea*** Sabine, *Kitjima s.n.* (A), HQ331669, HQ331973, [*Nagata 951*, (HLA), AF518383], HQ331817; ***Platonia insignis*** Mart., *V. Bittrich s.n. 3.01.05* (INB), HQ331670, HQ331974, [*Mori 23699* (NY), AF518394], HQ331818; ***Symphonia fasciculata*** (Noronha ex Thouars) Vesque, *J.S. Miller et al 8836* (MO), HQ331679, HQ331984, HQ332117, HQ331825; ***Symphonia globulifera*** L. f., *Ruhfel 21* (A), HQ331680, HQ331985, [*Mori 24792* (NY), AF518381], HQ331826; ***Tovomita calophyllophylla*** García-Villacorta & Hammel, *J.Vormisto 579* (AAU), HQ331683, HQ331988, HQ332119, HQ331828; ***Tovomita longifolia*** (Rich.) Hochr., *R. Aguilar ra12290* (NY), HQ331684, HQ331989, HQ332120, HQ331829; ***Tovomita sp.***, *J. Vormisto 562* (AAU), HQ331685, HQ331990, HQ332121, HQ331830; ***Tovomita weddelliana*** Planch. & Triana, *M. H. G. Gustafsson 478* (AAU), HQ331686, HQ331991, HQ332122, HQ331831; ***Tovomitopsis saldanhae*** Engl., *V. Bittrich s.n.* (UEC), HQ331687, HQ331992, HQ332123, —.

CTENOLOPHONACEAE. ***Ctenolophon englerianus*** Mildbr., *McPherson 16911* (MO), EF135524, FJ670074, AJ402940, AY674499.

ELATINACEAE. ***Elatine triandra*** Schkuhr, *Burton et al. 13384* (MICH), [EF135532], AY425049, [AY380349], AY674507.

EUPHORBIACEAE. ***Ricinus communis*** L., *Wurdack D9* (US), EF135590, FJ670089, AY788188, AY674560.

GOUPIACEAE. ***Goupia glabra*** Aubl., *Prevost 3031* (CAY), EF135544, AY425054, AJ235780, AY674516.

HUMIRIACEAE. ***Humiria balsamifera*** Aubl., *Anderson 13654* (MICH), EF135549, AF351007, L01926, AY674523.

HYPERICACEAE. ***Cratoxylum arborescens*** (Vahl) Blume, *Ruhfel 121* (A), HQ331586, HQ331890, HQ332045, HQ331739; ***Cratoxylum cochinchinense*** (Lour.) Blume, *Church et al. 2699* (A), HQ331587, HQ331891, HQ332046, HQ331740; ***Cratoxylum formosum*** (Jack) Dyer, *Ruhfel 107* (A), HQ331588, HQ331892, HQ332047, HQ331741; ***Cratoxylum glaucum*** Korth., *Ruhfel 102* (A), HQ331589, HQ331893, HQ332048, HQ331742; ***Cratoxylum sumatranum*** (Jack) Blume, *Chase 1218* (K), FJ670022, FJ670095, AF518395, FJ670373; ***Eliea articulata*** Cambess., *Razakamalala 295* (MO), FJ670023, FJ670096, FJ670167, FJ670374; ***Harungana madagascariensis*** Poir., *B. Pettersson and L. A. Nilson 37* (UPS), HQ331615, HQ331920, [*Naugona 139* (NY), AF518396], HQ331766; ***Hypericum aegypticum*** L., *M. Gustafsson MG 1148* (AAU), HQ331617, HQ331922, HQ332069, HQ331767; ***Hypericum androsaemum*** L., *J. Christiansen s.n.* (AAU), HQ331618, HQ331923, HQ332070, HQ331768; ***Hypericum annulatum*** Moris, *J. Christiansen s.n.* (AAU), HQ331619, HQ331924, HQ332071, HQ331769; ***Hypericum canariense*** L., *J. Christiansen s.n.* (AAU), HQ331620, HQ331925, HQ332072, HQ331770; ***Hypericum ellipticum***

Hook., *C.C. Davis s.n.* (A), HQ331621, HQ331926, —, HQ331771; ***Hypericum elodes*** L., *Halliday s.n., 6/7 1964* (AAU), HQ331622, —, HQ332073, HQ331772; ***Hypericum empetrifolium*** Willd., *Chase 837* (K), HQ331623, AY425060, HQ332074, AY674525; ***Hypericum garrettii*** Craib, *J. Christiansen s.n.* (AAU), HQ331624, HQ331927, HQ332075, HQ331773; ***Hypericum grandifolium*** Choisy, *M. Gustafsson MG1147* (AAU), HQ331625, HQ331928, HQ332076, HQ331774; ***Hypericum hircinum*** L., *J. Christiansen s.n.* (AAU), HQ331626, HQ331929, HQ332077, HQ331775; ***Hypericum irazuense*** Kuntze ex N. Robson, *Ruhfel 8* (A), —, —, HQ332078, HQ331776; ***Hypericum kalmianum*** L., *C.C. Davis s.n.* (A), HQ331627, HQ331930, HQ332079, —; ***Hypericum linarifolium*** Vahl, *J. Christiansen s.n.* (AAU), HQ331628, HQ331931, HQ332080, HQ331777; ***Hypericum mutilum*** L., *C.C. Davis s.n.* (A), HQ331629, HQ331932, —, HQ331778; ***Hypericum perforatum*** L., *Ruhfel s.n.* (A), HQ331630, HQ331933, HQ332081, —; ***Hypericum tetrapterum*** Fr., *J. Christiansen s.n.* (AAU), HQ331631, HQ331934, HQ332082, HQ331779; ***Psorospermum*** **aff.** ***androsaemifolium*** Baker, *R. Randrianaivo et al. 145* (UPS), HQ331675, —, HQ332111, —; ***Psorospermum corymbiferum*** Hochr., *J.E. Lawesson and Goudiaby 7578* (AAU), HQ331676, HQ331979, HQ332112, HQ331821; ***Psorospermum febrifugum*** Spach, *M. Hedren et al. 394* (UPS), HQ331677, HQ331980, HQ332113, HQ331822; ***Psorospermum revolutum*** (Choisy) Hochr., *M. Thulin, P. Kornhall, and M. Popp 10312* (UPS), HQ331678, —, HQ332114, HQ331823; ***Santomasia steyermarkii*** (Standl.) N. Robson, *E. Matuda S-228* (A), —, HQ331982, —, —; ***Thornea calcicola*** (Standl. & Steyerm.) Breedlove & E.M. McClint., *D.E. Breedlove 37070* (MO), HQ331682, [*J.A. Steyermark 48946* (A), HQ331987], —, —; ***Triadenum fraseri*** (Spach) Gleason, *C.C. Davis s.n.* (A), HQ331688, HQ331993, HQ332124, [*C.C. Davis s.n.* (A), HQ331832]; ***Triadenum japonicum*** (Blume) Makino, *S. Kobayashi 2713* (A), HQ331689, HQ331994, HQ332125, HQ331833; ***Triadenum walteri*** (J.F. Gmel.) Gleason, *Brant 4792* (MO), HQ331690, FJ670097, FJ670168, FJ670375; ***Vismia baccifera*** (L.) Triana & Planch., *Ruhfel 20* (A), HQ331692, HQ331996, [*Gustafsson 302* (NY), AF518382], HQ331835; ***Vismia bilbergiana*** Beurl., *B. Hammel 25285* (MO), HQ331693, HQ331997, [*STRI:BCI 734543* (STRI), GQ981917], HQ331836; ***Vismia guianensis*** (Aubl.) Choisy, *Amorim 7659* (CEPC), HQ331694, HQ331998, HQ332126, [*Amorim 3978* (CEPC), HQ331837]; ***Vismia guineensis*** (L.) Choisy, *M. Merello et al. 1149* (UPS), HQ331695, HQ331999, —, HQ331838; ***Vismia macrophylla*** Kunth, *Amorim 3972* (CEPC), HQ331696, HQ332000, —, HQ331839; ***Vismia rubescens*** Oliv., *R. Niangadouma et al. 374* (MO), —, HQ332001, HQ332127, HQ331840; ***Vismia sp.,*** *Miller et al. 9313* (MO), EF135601, FJ670098, FJ670169, AY674571.

IRVINGIACEAE. ***Irvingia malayana*** Oliv., *Simpson 2638* (K), EF135553, AY425061, AF123278, EF135300.

IXONANTHACEAE. ***Cyrillopsis paraensis*** Kuhl., *Hentrich 68* (NY), FJ670024, FJ670100, FJ670170, FJ670376.

LACISTEMATACEAE. ***Lacistema aggregatum*** Rusby, *Pennington et al. 583* (K), FJ670025, AY425064, AF206787, AY674529.

LINACEAE. ***Reinwardtia indica*** Dumort., *Chase 230* (NCU), AB048380, FJ670104, L13188, AY674559.

LOPHOPYXIDACEAE. ***Lophopyxis maingayi*** Hook. f., *Adelbai P-10203* (US), EF135560, FJ670105, AY663643, AY674534.

MALPIGHIACEAE. ***Acridocarpus natalitius*** Adr. Juss., *Goldblatt s.n.* (PRE), AF344525, AF351016, AF344455, EF135290.

OCHNACEAE. ***Ochna multiflora*** DC., Chase 229 (NCU), EF135572, AY425072, Z75273, EF135302.

OXALIDACEAE. ***Averrhoa carambola*** L., *Chase 214* (NCU), FJ670048, FJ670141, FJ670180, AY674478.

PANDACEAE. ***Panda oleosa*** Pierre, *Schmidt et al. 2048* (MO), FJ670032, FJ670111, AY663644, FJ670383.

PASSIFLORACEAE. ***Paropsia madagascariensis*** (Baill.) H. Perrier, *Zyhra 949* (WIS), EF135576, AY757164, AF206802, AY674547.

PERACEAE. ***Pera bicolor*** (Klotzsch) Müll. Arg., *Gillespie 4300* (US), EF135578, AY425075, AY794968, AY674549.

PHYLLANTHACEAE. ***Phyllanthus epiphyllanthus*** L., *Wurdack D56* (US), EF135581, AY425078, AY663604, AY674552.

PICRODENDRACEAE. ***Podocalyx loranthoides*** Klotzsch, *Berry & Aymard 7226* (MO), EF135583, FJ670117, AY663647, AY674553.

PODOSTEMACEAE. ***Apinagia longifolia*** (Tul.) P. Royen, *C.T. Philbrick 6023* (WCSU), HQ331543, HQ331844, HQ332005, HQ331700; ***Apinagia riedelii*** Tul., *C.T. Philbrick 5960* (WCSU), HQ331544, HQ331845, HQ332006, HQ331701; ***Castelnavia monandra*** Tul. & Wedd., *C.T. Philbrick 5982* (WCSU), HQ331567, HQ331869, HQ332028, HQ331722; ***Ceratolacis pedunculatum*** C. Philbrick, *Novelo & Irgang, C.T. Philbrick 5761* (MO), HQ331568, HQ331870, HQ332029, —; ***Cladopus japonicus*** Imamura, *S. Koi and N. Katayama JP-404* (TNS), HQ331573, HQ331876, HQ332033, HQ331727; ***Cladopus queenslandicus*** (Domin) C.D.K. Cook & Rutish., *J.J. Bruhl and I.R. Telford 2542* (MO), HQ331574, HQ331877, HQ332034, —; ***Dalzellia zeylanica*** Wight, *M. Kato and N. Katayama SL-101* (TNS), HQ331590, HQ331894, [SL-04 (TNS), AB113760], HQ331743; ***Diamantina lombardii*** Novelo, *C. Philbrick & Irgang, C.T. Philbrick 5783* (WCSU), HQ331591, —, —, —; ***Dicraeanthus zehnderi*** H.E. Hess, *Ghogue GHO-1650* (Z/ZT), HQ331592, HQ331895, HQ332049, HQ331744; ***Djinga felicis*** C. Cusset, *Ghogue et al. GAR-09* (Z/ZT), HQ331593, HQ331896, HQ332050, HQ331745; ***Endocaulos mangorense*** (H. Perrier) C. Cusset, *Kato et al. MD-02* (TI), AB038191, —, —, —; ***Griffithela hookeriana*** (Tul.) Warm., *C.T. Philbrick 4683* (WCSU), HQ331612, HQ331917, HQ332067, —; ***Hanseniella heterophylla*** C. Cusset, *Kato et al. TL-311* (TI), AB104562, —, —, —; ***Hydrobryum japonicum*** Imamura, *S. Koi and N. Katayama JP-401* (TNS), HQ331616, HQ331921, —, —; ***Indodalzellia gracilis*** (C.J. Mathew, Jäger-Zürn, & Nileena) Koi & M. Kato, *KI-115* (TNS), AB450015, —, —, —; ***Indotristicha ramosissima*** (Wight) Royen, *M. Kato et al. KI-210* (TNS), HQ331632, HQ331935, [*KI-26* (TNS), AB124844], HQ331780; ***Inversodicraea*** **cf.** ***annithomae*** (C. Cusset) R.Rutish. and Thiv, *Ghogue et al. GAHR-23* (Z/ZT), HQ331633, HQ331936, HQ332083, HQ331781; ***Inversodicraea*** **cf.** ***bosii*** (C. Cusset) R.Rutish. & Thiv, *Ghogue et al GAR-01* (Z/ZT), HQ331634, HQ331937, HQ332084, HQ331782; ***Inversodicraea cristata*** Engler, *Ghogue GHO-1664* (Z/ZT), HQ331635, HQ331938, HQ332085, HQ331783; ***Ledermanniella bifurcata*** (Engler) C. Cusset, *Ghogue GHO-1597* (Z/ZT), HQ331643, HQ331945, HQ332090, HQ331791; ***Ledermanniella bowlingii*** (J.B. Hall) C. Cusset, *Ameka and Rutishauser AR-021010* (Z/ZT), HQ331644, HQ331946, HQ332091, HQ331792; ***Ledermanniella letouzeyi*** C. Cusset, *Ghogue et al. GAR-12* (Z/ZT), HQ331645, HQ331947, HQ332092, HQ331793; ***Ledermanniella linearifolia*** Engler, *Ghogue et al. GAHR-41* (Z/ZT), —, HQ331948, HQ332093, HQ331794; ***Ledermanniella pusilla*** (Warming) C. Cusset, *Ghogue et al. GAHR-17* (Z/ZT), HQ331646, HQ331949, HQ332094, HQ331795; ***Leiothylax quangensis*** (Engler) Warming, *Ghogue GHO-1667* (Z/ZT), FM877842, HQ331950, HQ332095, —; ***Letestuella tisserantii*** G. Taylor, *Ghogue GHO-1660* (Z/ZT), HQ331647, HQ331951, HQ332096, HQ331796; ***Macropodiella heteromorpha*** (Baillon) C. Cusset, *Ghogue et al. GAHR-24* (Z/ZT), HQ331649, HQ331953, HQ332097, HQ331798; ***Marathrum foeniculaceum*** Bonpl., *C.T. Philbrick 5958* (WCSU), HQ331658, HQ331962, HQ332103, HQ331806; ***Marathrum plumosum*** (Novelo & C.T.Philbrick) *C.T.Philbrick & C.P.Bove, MX-05* (TI), AB048378, —, [Les et al., U68090], —; ***Monandriella linearifolia*** Engler, *Ghogue GHO-1663* (Z/ZT), HQ331662, HQ331966, HQ332104, HQ331810; ***Monostylis capillacea*** Tul., *C.T. Philbrick 6076* (WCSU), HQ331663, HQ331967, HQ332105, HQ331811; ***Mourera*** **cf.** ***aspera*** (Bong.) Tul., *C.T. Philbrick 6093* (WCSU), HQ331666, HQ331970, [Les et al, U68086], HQ331814; ***Mourera fluviatilis*** Aubl., *GU-24* (TI), AB038200, —, [not listed, AB113759], —; ***Noveloa coulteriana*** (Tul.) C.T.Philbrick, *C.T. Philbrick 6270* (WCSU), HQ331667, HQ331971, HQ332106, HQ331815; ***Paracladopus chanthaburiensis*** Koi & M. Kato, *S. Koi et al. TKF-24* (TNS), HQ331668, HQ331972, HQ332107, HQ331816***;*** ***Podostemum ceratophyllum*** Michx., *Ruhfel s.n.* (A), HQ331671, HQ331975, HQ332108, [*Horn s.n.* (DUKE), EF135304]; ***Podostemum scaturiginum*** (Mart.) C. Philbrick & Novelo, *C.T. Philbrick et al. 5602* (MO), HQ331672, HQ331976, HQ332109, —; ***Polypleurum stylosum*** (Wight) J.B. Hall, *M. Kato and N. Katayama SL-103* (TNS), HQ331674, HQ331978, HQ332110, HQ331820; ***Rhyncholacis sp.***, *Amaral s.n.* (INPA), EF135564, HQ331981, HQ332115, AY674537; ***Stonesia ghoguei*** E. Pfeifer and Rutishauser, *Ghogue GHO-1665* (Z/ZT), FM877841, HQ331983, HQ332116, HQ331824; ***Terniopsis brevis*** M. Kato, *S. Koi et al. TKF-25* (TNS), HQ331681, HQ331986, HQ332118, HQ331827; ***Terniopsis malayana*** (J.Dransf. & Whitmore) M.Kato, *TL-106, 107* (TNS), AB048827, —, AB083098, —; ***Terniopsis sessilis***

Hsiu C. Chao, *CH-03* (TI), AB048377, —, AB083100, —; ***Thawatchaia trilobata*** M.Kato, Koi & Y.Kita, *Kato et al. TL-419* (TI), AB104563, —, —, —; ***Thelethylax minutiflora*** (Tul.) C. Susset, *Kato et al. MD-01* (TI), AB038196, —, —, —; ***Tristicha trifaria*** (Bory ex Willd.) Spreng., *C.T. Philbrick 6090* (WCSU), HQ331691, HQ331995, [*BR-01*, AB113746], HQ331834; ***Weddellina squamulosa*** Tul., *C.T. Philbrick 5827* (WCSU), HQ331697, HQ332002, [not listed, AB113758], HQ331841; ***Zeylanidium lichenoides*** Engl., *Kato et al. KI-35* (TI), AB048828, —, —, —; ***Zeylanidium subulatum*** (Gardner) C. Susset, *M. Kato and N. Katayama SL-102* (TNS), HQ331698, HQ332003, HQ332128, HQ331842.

PUTRANJIVACEAE. ***Putranjiva roxburghii*** Wall. (as *Drypetes roxburghii* [Wall.] Hurus.), *Wurdack D57* (US), EF135530, [AY425048], [M95757], [AY674505].

RHIZOPHORACEAE. ***Bruguiera gymnorhiza*** Lam., *Chase 12838* (K), EF135511, AY425036, [AF127693], AY674483.

SALICACEAE. ***Populus maximowiczii*** Henry, *Chase 996* (K), EF135587, AY425080, AJ418836, AY674556.

VIOLACEAE. ***Hybanthus concolor*** Spreng., *Alford 3056* (BH), EF135550, AY757141, AY788178, AY674524.