

The Shenzhen Declaration on Plant Sciences: Too late or just in time?

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On 29 July 2017 in the closing session of the XIX International Botanical Congress held in Shenzhen, China, nearly 7,000 plant scientists from 77 countries unanimously endorsed a statement to focus their research and educational efforts on finding solutions to the growing problems of “environmental degradation, unsustainable resource use, and biodiversity loss.” This moment was the culmination of a seven-day conference that brought together botanists from around the world to discuss and share their latest exciting research findings on a wide range of topics across the plant sciences. Yet, in addition to the scientific advances being communicated, an awareness and recognition was pervasive and much discussed throughout the week of the IBC that our planet is changing in substantial ways that will affect the social, political, and economic frameworks of our lives into the foreseeable future. And all agreed that these immense changes are the result of unbridled human activities.

The Shenzhen Declaration, conceived and composed by a broadly representative group of scientists, aims to raise the awareness of botanists to the accelerating rate of environmental change around the globe. More importantly it calls on them to make a commitment to take action now in both their lifestyles and their research programs to find solutions before an environmental threshold is crossed that will inevitably lead to

irreversible degradation of our societies, natural habitats, and biodiversity. Although many scientists believe that humanity and the planet may have already crossed that threshold, the authors of the Declaration and the botanists who have endorsed it believe that time still exists for answers to be found and implemented. However, that time is short.

The seven priorities outlined in the Declaration range broadly from increased action by scientific communities, more cooperation and integration across disciplines, the implementation of new technologies, valuing local and traditional knowledge, to greater engagement with the public. Realizing and achieving these priorities will be a major challenge that will require re-orienting research agendas and new resources. However, the enthusiastic response to the Declaration by the thousands of plant scientists in Shenzhen suggests that the botanical community now has a solid and inspiring roadmap for the future. If we are to successfully build a green and sustainable Earth, all scientists and citizens, not just botanists, should carefully read, study, and take steps to participate in collective action to make the seven priorities of the Shenzhen Declaration a reality for our common future.

The Shenzhen Declaration on Plant Sciences – Uniting plant sciences and society to build a green, sustainable Earth

Shenzhen Declaration Drafting Committee

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VITAL CONNECTIONS

Actions and priorities to connect the global community of plant scientists with the world's changing societies are today more imperative than ever. Environmental degradation, unsustainable resource use, and biodiversity loss all require integrated, collaborative solutions.

A CHANGING WORLD

As plant scientists we are increasingly aware and concerned with the accelerating rate of change of our planet and our societies. In our lifetimes we have witnessed major alterations in the structure and make-up of land, water, and the atmosphere, in use of natural resources and agricultural practices, in migration of plants, animals, and people, in rates of urbanization, and in the rise and spread of infectious diseases. The rate of species extinction is greater now than at any time in the last 65 million years. It is clear that this tremendous transformation, with its profound effect on nature, is primarily the result of human activities. The degree of pressure on the environment has never been greater — far beyond the level at which natural systems will be able to maintain sustainable productivity. The need to act is urgent.

Equally in transition are our own disciplines in the plant sciences: taxonomy and systematics, morphology and development, evolution and ecology, physiology and genetics. New technologies that generate immense quantities of data are often limited by current infrastructure and information management capabilities; a growing emphasis on laboratory investigations is overshadowing the need for priority field work in rapidly disappearing environments; and balance in training for pure and applied research careers is shifting. In many nations, funding support for basic science is declining along with public trust in science. Parallel to these changes within the plant sciences are those affecting social, political, and economic contexts within which scientific research is conducted. Factors such as growing income inequality among peoples, the uneven redistribution of resources across the globe, and rising levels of conflict within and among nations all impact our ability to conduct meaningful science.

At this time of extraordinary challenges, the International Botanical Congress is being held for the first time in China. The increasing wealth of China and the prosperity of its people, coupled with the country's need for and interest in tackling serious national environmental problems, have given the country a key role in combatting climate change. China also has the potential to address biodiversity loss through the development and implementation of a strong national plan in this area. The Chinese linking of "risks" with "opportunities" has never carried more meaning than it does now, at a time when all countries need for their own sake, and for the world, to help achieve global sustainability. The hosting of IBC 2017 in Shenzhen, this Declaration, and the establishment of the Shenzhen International Award in Plant Sciences are measures of China's clear commitment to action.

THE SHENZHEN CALL FOR ACTION: SEVEN PRIORITIES

We endorse the following seven priorities for strategic action in the plant sciences. Vigorous development of these areas will allow society, with the help of science, to mitigate impacts of human activities on plant species, habitats, and distributions, and to approach formation of a sustainable world for ourselves and those who follow us.

To become responsible scientists and research communities who pursue plant sciences in the context of a changing world.

Plant scientists must contribute to regional and global sustainability as directly and efficiently as possible. Key efforts, such as the urgent preservation of plant diversity and the adaptation of agriculture to increasingly warm climates, must be strengthened greatly if we are to meet the challenges ahead. Our research is not conducted in a vacuum, and we cannot continue to act as if we have a great deal of time available, when we simply and clearly do not. We must confront challenges swiftly and directly to mitigate rapidly deteriorating environmental conditions.

To enhance support for the plant sciences to achieve global sustainability.

Plants play a central role in functioning ecosystems. They also are our sole source of food (directly or indirectly), and provide many of our medicines, building materials, clothing materials, and other essential products. Plants deserve a far greater level of scientific attention through enhanced public and private funding than they are receiving at present. Integrated studies are necessary to develop robust solutions to environmental problems. Support across plant sciences, from description to use, should be provided at adequate levels and sustained.

To cooperate and integrate across nations and regions and to work together across disciplines and cultures to address common goals.

Science is by its very nature international, with the plant sciences no exception. Although progress has been made in moving forward with together, stronger international cooperation will be needed to halt biodiversity loss, improve agriculture, and maintain a stable climate. Working together has never been more important. Stable global partnerships are badly needed to overcome barriers and provide integrated, effective solutions to urgent environmental challenges as rapidly as possible.

To build and use new technologies and big data platforms to increase exploration and understanding of nature.

New technical approaches to information and information sharing will only accelerate in the years to come, making sustainability of data platforms imperative. Increasingly large, linked databases reveal new connections and relationships about life on Earth. Our rapidly advancing ability to sequence genomes leads to new ways of understanding the diversity, evolution, and functioning of life on our planet. As these and other new technologies expand, we must apply them in timely, integrated, and practical ways to organize information and address environmental problems.

To accelerate the inventory of life on Earth for the wise use of nature and the benefit of humankind.

More than half of the land plant species could be extinct in nature by the end of the present century. Although we have given names to many, we know very little about most of them, and there are more that await discovery. Those we know now can be protected or preserved, but the urgency of finding and learning about the unknowns before they become extinct is clear. Doing so will require integration and collaboration on a scale we have not yet achieved. We need to know plants in order to save them, but time is short.

To value, document, and protect indigenous, traditional, and local knowledge about plants and nature.

Indigenous, traditional, and local knowledge about nature is disappearing even more rapidly than is biodiversity itself. Once lost, such knowledge, with its unique insights into nature, can never be regained. Plant scientists must work together with holders of this knowledge to understand and achieve sustainable environmental stewardship. Cultural diversity, coupled with crop genetic diversity, will be of central importance for future food security. We will need informed collaboration coupled with urgent, rigorous planning and implementation across cultures and knowledge systems.

To engage the power of the public with the power of plants through greater participation and outreach, innovative education, and citizen science.

We need to engage the power of the public with the power of nature. People who care about the environment are motivated to do a great deal to protect it and ensure its future. The creation of an ecological civilization, where societies work together in the creation of knowledge and implementation of solutions, cannot remain only an abstract concept. We all need plants, and plants need our care now more than ever—we depend absolutely upon them for our very existence. Embedding that need into the very fabric of our societies will require global engagement, across nations and cultures — this will require all of us.

We believe that, by working together, we can unite innovative plant sciences with the needs and strengths of human societies, helping to create new paths to a green, sustainable future for Earth, with plants and people in harmony.

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On merging *Acer* sections *Rubra* and *Hyptiocarpa*: Molecular and morphological evidence

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Abstract

In this study, we expanded *Acer* sect. *Rubra* Pax to include *A.* sect. *Hyptiocarpa* Fang. Traditionally, section *Rubra* comprises two iconic species, *Acer rubrum* Linnaeus (red maple) and *A. saccharinum* Linnaeus (silver maple), of eastern North American forests as well as the rare Japanese montane species, *A. pycnanthum* K. Koch. Section *Hyptiocarpa* consists of *A. laurinum* Hasskarl and *A. pinnatinervium* Merrill, which occur in subtropical and tropical regions of southwestern China to southeast Asia. Here, we confirm prior phylogenetic results showing the close relationship between sects. *Rubra* and *Hyptiocarpa*, and we use scanning electron microscopy to demonstrate that leaves of species within these sections have similar arrangements of cuticular waxes, which account for the silvery color of their abaxial surfaces. We describe that the sections also share labile sex expression; inflorescences that range from compound racemose thyrses, to racemes or umbels and that may have undergone evolutionary reduction; and several features of their fruits, such as seed locules without keels, basal portion of wings straight, acute attachment angle between mericarps, and production of some mericarps that are seedless and partially developed at maturity. Our expansion of sect. *Rubra* to include sect. *Hyptiocarpa* better elucidates the biogeographic and evolutionary history of these species. Additionally, we show that *A. laurinum* and *A. pinnatinervium* have intergrading morphology and are probably synonymous, but we note that further studies are required to conclude their taxonomic status.

Keywords

Acer laurinum, *Acer pycnanthum*, *Acer rubrum*, *Acer saccharinum*, cuticle, ITS, scanning electron microscopy

Introduction

Acer Linnaeus, the maple genus, is remarkable for comprising 125+ species and for representing one of the largest woody plant genera in the Northern Hemisphere next to oaks and willows (Ogata 1967; de Jong 1976; Murray 1970; Delendick 1981; Xu et al. 2008; van Gelderen et al. 1994; Weakley 2011). *Acer* exhibits a classical pattern of biogeographic disjunction across Europe, northern Africa, Asia, and North America with the greatest species richness in eastern Asia (Raven 1972; Thorne 1972; de Jong 1976; van Gelderen et al. 1994; Wen 1999; 2001; Qian and Ricklefs 2000; Xu et al. 2008; Wen et al. 2016; Harris et al. 2017). The genus is highly valued horticulturally and for timber and sugar products (Larsson and Jaciw 1967; Delendick 1981; van Gelderen et al. 1994; de Beaulieu and Mechelynck 2003; Barrett 2004).

Acer and the closely-related genus, *Dipteronia* Oliver (2 spp.), formerly comprised Aceraceae but are now treated in tribe Acereae of Sapindaceae (Acevedo-Rodriguez et al. 2011). Acereae belongs to subfamily Hippocastanoideae with tribe Hippocastaneae, which includes *Aesculus* Linnaeus, or the horsechestnuts and buckeyes, and two other small genera (Judd et al. 1994; Harrington et al. 2005; Buerki et al. 2010). Acereae has been fairly taxonomically stable, and consists of a well-supported clade based on morphological and molecular data (Willis 1980; Harrington et al. 2005; Pan et al. 2008; Buerki et al. 2010).

Within *Acer*, the circumscription of infra-generic groups has been controversial. Some groups are reasonably well agreed upon, such as section *Macrantha*, which includes species that have conspicuously white- or green-striped bark, and the recognition of *Acer carpinifolium* Siebold & Zuccarini as the sole member of sect. *Indivisia* (e.g. Momotani 1962; Ogata 1967; Murray 1970; de Jong 1976; Delendick 1981; Wolfe and Tanai 1987; van Gelderen et al. 1994). On the other hand, some sections have been wildly unstable, such as sect. *Negundo*, which possesses one to several species and is sometimes raised to generic or subgeneric status (Fang 1966; de Jong 1976). The controversy regarding infra-generic groups in *Acer* ultimately reflects uncertainty about the evolutionary relationships among species.

Another maple that has not enjoyed taxonomic stability is *A. laurinum* Hasskarl. *Acer laurinum* was described as *A. javanum* (Junghuhn, 1841), an impressive tree with leaves and fruits that stood out from a distance. Hasskarl (1843) referred to Junghuhn's description and renamed the taxon to *A. laurinum* (*nomen novum*) two years later, because the prior name was already in use (i.e., *A. javanicus*, now recognized as a species in either *Actinomorpha* or *Colona*, see Hasskarl 1857, Murray 1970). *Acer laurinum* has undergone considerable taxonomic splitting and lumping, and disagreements on its delimitation and the number of subspecific entities suggest that it is highly variable, but its variants may lack notable limits (e.g., Fang 1966; Murray 1970; van Gelderen et al. 1994). *Acer laurinum* is most often treated within sect. *Hyptiocarpa* (Fang 1966; = sect. *Laurinum* of Ogata 1967) or within sect. *Integrifolia* (Merrill 1941; Pax 1885). In the most recent global treatment of *Acer* van Gelderen et al. (1994) asserted that sect. *Hyptiocarpa* comprised *A. laurinum* and *A. garrettii* Craib, the latter of which the

authors speculated may not merit species status. In contrast, Flora of China (Xu et al. 2008) represents a more recent and regional treatment of sect. *Hyptiocarpa* and recognizes two species: *A. laurinum* (including *A. garrettii*) and *A. pinnatinervium* Merrill, which the global treatment had synonymized with *A. garrettii*.

The affinities of sect. *Hyptiocarpa* remain highly speculative, and its closest relatives may be within sects. *Integrifolia*, *Trifoliata*, *Rubra*, or *Lithocarpa* (Pax 1885; Tanai 1978; Fang 1966; van Gelderen et al. 1994). *Acer laurinum* is an anomaly among maples and is of biogeographic importance, because it has a geographic range from southern China, Vietnam, and Thailand to the Philippines and Java. Therefore, it is the only maple to have a distribution that crosses the equator into the Southern Hemisphere.

Recently, phylogenetic studies using chloroplast and nuclear DNA with several methods of analysis have repeatedly shown strong support for the somewhat unexpected sister relationship between *Acer* sects. *Hyptiocarpa* and *Rubra* (Suh et al. 2000; Tian et al. 2002; Grimm et al. 2006; Renner et al. 2008). Section *Rubra* is a highly supported clade (Grimm et al. 2006; Renner et al. 2008) that possesses three species: *A. rubrum* Linnaeus (red maple) and *A. saccharinum* Linnaeus (silver maple), which are iconic species in eastern North America, and *A. pycnanthum* K. Koch, which occurs in montane areas of Honshu in Japan (Ohwi 1965; van Gelderen et al. 1994). The molecular phylogenetic studies that support the relationship between sects. *Hyptiocarpa* and *Rubra* have included sequences from all species of sect. *Rubra* and from *A. laurinum* or, in one case, a sequence from a specimen of *A. garrettii* collected by its authority, Craib (Suh et al. 2000; Tian et al. 2002; Grimm et al. 2006; Renner et al. 2008). Infrequently, taxonomic and morphological studies have also speculated on a possible relationship between sects. *Rubra* and *Hyptiocarpa* (Delendick 1981; van Gelderen et al. 1994), and we observed that the possibility is reinforced by the conspicuous silvery abaxial leaf surfaces that are common to all four species (Fig. 1) and noted as a distinctive feature of sect. *Hyptiocarpa* (Merrill 1941).

In this study, we present evidence for the relationship between sects. *Rubra* and *Hyptiocarpa* from nuclear and chloroplast phylogenies and from an analysis of leaf cuticular wax ultrastructures. We also compare other morphological features of the sections according the available literature and specimens and discuss these in the context of biogeography and evolutionary radiation. Based on the results of our study, we propose combining sects. *Rubra* and *Hyptiocarpa* within sect. *Rubra* s.l. Throughout the study, we apply the taxonomy of van Gelderen et al. (1994) except as otherwise noted and for sect. *Hyptiocarpa*, for which we apply the more recent treatment from the Flora of China (Xu et al. 2008).

Methods

Phylogenetic reconstruction

We reconstructed phylogenies of Acereae at the section-level using sequences of nuclear Internal Transcribed Spacer (ITS) and the chloroplast spacer *trnD-psbM* (hereafter,

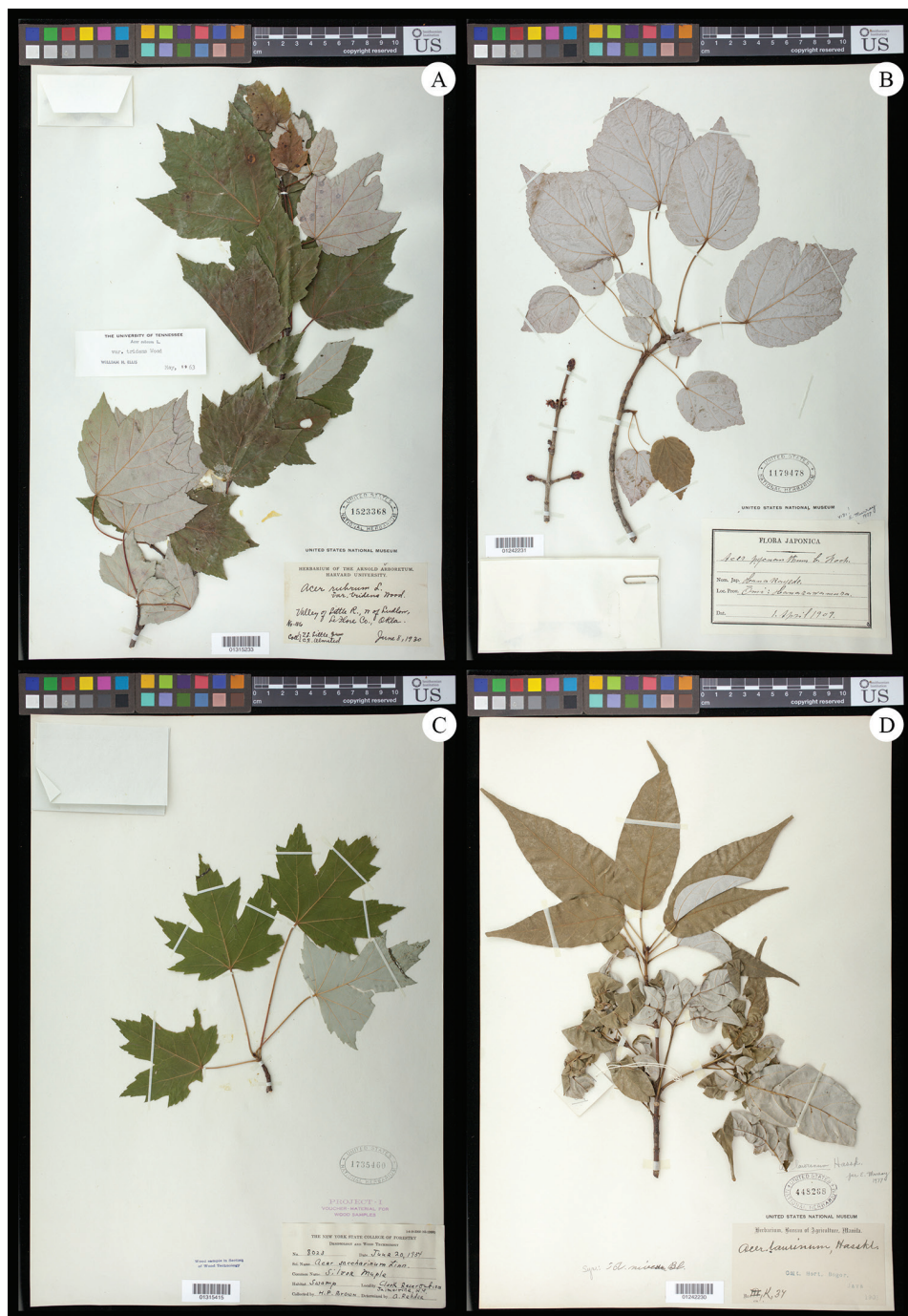


Figure 1. Typical specimens of *Acer* sects. *Rubra* and *Hyptiocarpa*, especially exhibiting leaf macromorphology. **A** *A. rubrum* **B** *A. pycnanthum* **C** *A. saccharinum* **D** *A. laurinum*. Specimens deposited at US national herbarium, and accession information visible in images. Detailed specimen records are available via the US online catalog (<http://collections.nmnh.si.edu/search/botany/>).

psbM). We selected these markers because of their utility as DNA barcodes in plants (Dong et al. 2012; Li et al. 2011; Shaw et al. 2005; Zuo et al. 2011, 2017), their demonstrated utility in *Acer* for the sections in question (Grimm et al. 2006; Renner et al. 2008), and our preliminary observations about the information content of *psbM* for *Acereae*. We reconstructed phylogenies for this study even though prior studies have generated phylogenies of *Acereae* using chloroplast DNA, ITS, and nuclear genes (Suh et al. 2000; Tian et al. 2002; Grimm et al. 2006; Buerki et al. 2010; Renner et al. 2008; Harris et al. 2017), because doing so gave us more control over sampling of representative species, the ability to curate data and make decisions about data quality, and first-hand knowledge of all analysis parameters.

We obtained sequences of *psbM* and ITS from GenBank for representative samples of sections of *Acer sensu* Wolfe and Tanai (1987), *A. laurinum*, and all species of sect. *Rubra sensu* van Gelderen et al. (1994). We used Wolfe and Tanai (1987) to guide our taxonomic sampling, because their treatment splits *Acer* into smaller sections, which are more consistent with large, published molecular phylogenies (e.g., Grimm et al. 2006, Renner et al. 2008) than the most recent treatment by van Gelderen (1994). Wolfe and Tanai recognized 21 sections of extant *Acer*, and maintained *A. saccharinum* in a separate, monotypic section from *A. rubrum* and *A. pycnanthum*. Our section-level sampling according to Wolfe and Tanai (1987) may underrepresent diversity in *Acer*, especially within sect. *Acer*, which has been the least taxonomically stable section and probably includes species that are phylogenetically distant (Ogata 1967; Wolfe and Tanai 1987; van Gelderen et al. 1994; Grimm et al. 2006, Renner et al. 2008). Nevertheless, resolving relationships in sect. *Acer* is beyond the scope of our study and, based on outcomes from prior molecular phylogenetic studies (Grimm et al. 2006, Renner et al. 2008), species variously treated in sect. *Acer* are distant from sects. *Rubra* and *Hyptiocarpa*. Of the 21 sections recognized by Wolfe and Tanai, we sampled 20, but the missing section, sect. *Integrifolia*, may be represented by *Acer pentaphyllum* Diels. Wolfe and Tanai (1987) treated *A. pentaphyllum* in sect. *Acer*, but the species is included in sect. *Pentaphylla* in van Gelderen et. al (1994) with other species of Wolfe and Tanai's (1987) sect. *Integrifolia* and is resolved with species of sect. *Integrifolia* in molecular phylogenies (Suh et al. 2000; Grimm et al. 2006; Renner et al. 2008). For all sections of *Acer*, we sampled the type species when possible. In addition to species of *Acer*, we included both species of *Dipteronia* in our analyses, and we utilized one sequence each of *Sapindus* Linnaeus, *Koelreuteria* Laxmann, and *Xanthoceras sorbifolium* Bunge as outgroups. *Xanthoceras* may be sister to all Sapindaceae and *Sapindus* and *Koelreuteria* represent the core Sapindaceae (*sensu* Buerki et al. 2010), which is sister to Hippocastanoideae (Buerki et al. 2010). We did not include Hippocastaneae among the outgroup or ingroup, because it has ITS sequences that are very difficult to align with *Acer* according to a prior report (Grimm et al. 2006) and our personal experience. Nevertheless, prior molecular phylogenetic studies of *Acer* have used cpDNA and have included *Aesculus* of Hippocastaneae (Renner et al. 2008; Tian et al. 2002). Therefore, we compare results of those studies with our own. The ITS and *psbM* datasets comprised 27 sequences each. The details of our sampling, including additional explana-

tion of taxonomic representativeness and GenBank accession numbers, are presented in Table 1.

Two sequences were new to this study: *psbM* of *Acer sterculiaceum* K. Koch subsp. *franchettii* (Pax) A.E. Murray and *A. cissifolium* (Siebold & Zucc.) K. Koch. We obtained the new sequences using fresh material, which we collected from the United States National Arboretum. Our collections consisted of leaves for DNA extractions, which we preserved in silica at the time of sampling, and voucher specimens, which we deposited at the United States National Herbarium (US; <http://n2t.net/ark:/65665/396759747-a431-4859-b4a7-8c57db1cc2a2> and <http://n2t.net/ark:/65665/36583930c-3354-4039-9e29-f9e0f9699ecb>). We performed DNA extractions using a Qiagen Plant Mini Kit (Venlo, Netherlands) according to manufacturer recommendations, and we amplified *psbM* using forward and reverse PCR primers from Lee and Wen (2004). We performed PCR, sequencing, and purification steps using the reactions, thermocycling scheme, and protocols reported in Harris et al. (2017), except that the thermocycling included 35, rather than 40, cycles. Our primers for sequencing were the same as those that we used for PCR amplification. We reported the new sequences to GenBank (Table 1).

We performed sequence alignment using the MAFFT algorithm (Katoh et al. 2002; Katoh et al. 2005) on the GUIDANCE 2 (Sela et al. 2015) webserver (<http://guidance.tau.ac.il/ver2/>; Penn et al. 2010). GUIDANCE 2 helps to identify uncertain regions of an alignment by comparing alignments derived from bootstrap guide trees. The GUIDANCE 2 webserver also facilitates removing uncertain portions of an alignment and realigning through an iterative, interactive process. We performed initial alignments on our ITS and *psbM* data matrices with up to five MAFFT iterations for refinement and 100 bootstrap replicates. We used a conservative confidence score of 0.853 (GUIDANCE 2 Overview, <http://guidance.tau.ac.il/ver2/overview.php>), and we removed all sites with lower confidence scores. Following this step, we performed a new alignment in GUIDANCE 2 with the uncertain sites excluded, and we checked that the new alignment had a confidence score of at least 0.95 (out of 1.0 possible) averaged across all sites. We also checked the final alignment visually with sites color-coded according to their GUIDANCE 2 score using JALVIEW (Waterhouse et al. 2009) on the GUIDANCE 2 webserver. We concatenated aligned matrices using SEQUENCEMATRIX (Vaidya et al. 2011), and our concatenated matrix comprised composite taxonomic entities of the same section in *Acer* and usually of the same species, except in the case of sects. *Rubra* and *Hyptiocarpa*, for which composite entities were always of the same species (see Table 1 for *Dipteronia* and outgroups). We provide all final alignments in Dryad: <http://dx.doi.org/10.5061/dryad.n26nd>

Prior to phylogenetic analyses, we assessed the data matrices for base compositional heterogeneity and to determine the best nucleotide substitution model. We sought to detect base compositional heterogeneity, because it can lead to errors in phylogenetic inferences especially in the placement of outgroups and other long branches (Tarrío et al. 2000; Jermini et al. 2004; Sheffield 2013). We performed the analysis for base compositional heterogeneity using a chi square test in PAUP*

Table 1. Representative sampling of species used in this study for molecular phylogenetic analysis.

	Species	GenBank-ITS	GenBank- <i>psbM</i>	Section affiliation <i>sensu</i> van Geldren (1994)	Section affiliation <i>sensu</i> Wolfe and Tanai (1987)
Acereae	<i>Acer argutum</i> Maximowicz	AF401153	DQ659842	<i>Glabra</i>	<i>Arguta</i>
	<i>Acer campestre</i> Linnaeus	LK022558	DQ659844	<i>Platanoides</i>	<i>Campestria</i>
	<i>Acer carpinifolium</i> Siebold & Zuccarini	AF401148	DQ659845	<i>Indivisia</i>	<i>Indivisa</i>
	<i>Acer cisifolium</i> (Siebold & Zuccarini) K. Koch	AY605402	KY682748	<i>Negundo</i>	<i>Cisifolia</i>
	<i>Acer distylum</i> Siebold & Zuccarini	DQ238354	DQ659850	<i>Parviflora</i>	<i>Distyla</i>
	<i>Acer glabrum</i> Torrey	DQ23834	DQ659892	<i>Glabra</i>	<i>Glabra</i>
	<i>Acer laurinum</i> Hasskarl	AF241490	DQ659854	<i>Hyptiocarpa</i>	<i>Laurina</i>
	<i>Acer macrophyllum</i> Pursh	DQ238352	DQ659860	<i>Lithocarpa</i>	<i>Macrophylla</i>
	<i>Acer negundo</i> Linnaeus	AY605407	DQ659864	<i>Negundo</i>	<i>Negundo</i>
	<i>Acer nipponicum</i> H. Hara	DQ366143	DQ659865	<i>Parviflora</i>	<i>Parviflora</i>
	<i>Acer palmatum</i> Thunberg	KT160159	DQ659867	<i>Palmata</i>	<i>Palmata</i>
	<i>Acer pensylvanicum</i> Linnaeus	AY605398	DQ659869	<i>Macrantha</i>	<i>Macrantha</i>
	<i>Acer pentaphyllum</i> Diels	DQ238478	DQ659870	<i>Pentaphylla</i>	<i>Acer</i>
	<i>Acer pentapomicum</i> Stewart ex Brandis	-	DQ659888	<i>Pubescens</i>	<i>Pubescens</i>
	<i>Acer pilosum</i> Maximowicz	DQ238345	-	<i>Pubescens</i>	<i>Pubescens</i>
	<i>Acer platanoides</i> Linnaeus	LK022676	DQ659871	<i>Platanoides</i>	<i>Platanoides</i>
	<i>Acer pseudoplatanus</i> Linnaeus	AM238269	DQ659872	<i>Acer</i>	<i>Acer</i>
	<i>Acer pycnanthum</i> K. Koch	AM113529	DQ659873	<i>Rubra</i>	<i>Rubra</i>
	<i>Acer rubrum</i> Linnaeus	AJ634580	DQ659874	<i>Rubra</i>	<i>Rubra</i>
	<i>Acer saccharinum</i> Linnaeus	AM113537	DQ659875	<i>Rubra</i>	<i>Eriocarpa</i>
	<i>Acer spicatum</i> Lamarek	AJ634578	DQ659879	<i>Parviflora</i>	<i>Spicata</i>
	<i>Acer sterculiaceum</i> K. Koch subsp. <i>franchetii</i> (Pax) A.E. Murray	DQ366145	KY682749	<i>Lithocarpa</i>	<i>Lithocarpa</i>
	<i>Acer tataricum</i> L. subsp. <i>ginnala</i> (Maximowicz) Maximowicz	AY605364	DQ659855	<i>Ginnala</i>	<i>Trilobata</i>
	<i>Dipteronia dyeriana</i> Henry	AM182900	DQ659838	-	-
	<i>Dipteronia sinensis</i> Oliver	AY605292	DQ659839	-	-

	Species	GenBank-ITS	GenBank- <i>psbM</i>	Section affiliation <i>sensu</i> van Geldren (1994)	Section affiliation <i>sensu</i> Wolfe and Tanai (1987)
Outgroups	<i>Koeberuteria</i> Laxmann	EU72057	DQ659835	-	-
	<i>Sapindus</i> Linnaeus	AY207570	DQ659836	-	-
	<i>Xanthoceras</i> Bunge	FJ375202	DQ659837	-	-

Notes: We indicate the section affiliation of the species according to van Gelderen et al. (1994) and Wolfe and Tanai (1987), who largely followed Ogata (1967). We include the GenBank accession number for the sequences of ITS that we used for phylogenetic analyses. All species typify the sections recognized by Wolfe and Tanai (1987) except as follows: (A) We included *Acer pycnanthum* of sect. *Rubra* even though it does not typify the section, because of the objectives of the study; (B) We included *A. pentaphyllum* of sect. *Pentaphyllum sensu* van Gelderen et al. to stand in as a representative of sect. *Integrifolia* of Wolfe and Tanai. *Acer pentaphyllum* typifies sect. *Pentaphyllum* in van Gelderen et al. and Ogata, and van Gelderen's sect. *Pentaphyllum* also includes species of Ogata's sect. *Integrifolia*. While Wolfe and Tanai recognized sect. *Integrifolia*, they did not recognize sect. *Pentaphyllum*, and they treated *A. pentaphyllum* in sect. *Acer*; (C) In the *psbM* dataset, we utilized *A. garrettii* to stand in for *A. laurinum*, because there were no available sequences of *psbM. Acer garrettii* was not mentioned (and probably not recognized) by Ogata and was hesitantly given species status by van Gelderen.

(Swofford 2002). We estimated the best model of nucleotide substitution from among 1-, 2-, and 6- parameter models with and without gamma rate variation (see Yang 1996 regarding invariance) in JMODELTEST (Posada 2008) under the Bayesian information criterion (BIC), and determined that the 6-parameter SMY+G (BIC=6544.4) and 2-parameter K80+G (BIC=5387.7) models were the best fit for ITS and *psbM*, respectively.

We performed phylogenetic analyses using neighbor-joining (NJ), maximum likelihood (ML), and Bayesian inference (BI) methods independently for ITS and *psbM* as well as for the concatenated data matrix. We performed the NJ analyses in GENEIOUS TREE BUILDER using Jukes Cantor distance and 1000 NJ bootstrap (BS) replicates to assess support. We reconstructed the ML trees in MEGA 6.06 (Tamura et al. 2013). In MEGA, we set models according to the results from JMODELTEST except that we used GTR+G for ITS, because it is the only 6-parameter model available in MEGA. We performed the analyses with five gamma rate categories and the subtree pruning and recrafting method of branch swapping. We also performed 500 BS replicates under the same parameters to determine support for clades. For BI, we utilized the GTR+G model of nucleotide substitution *a priori* (see Huelsenbeck and Rannala 2004; Ronquist et al. 2011) and unlinked models for the two markers in the analysis of concatenated data. The BI analysis comprised two simultaneous runs of 20 million generations with 12 incrementally heated MCMC chains each in MRBAYES 3.2.6 (Ronquist and Huelsenbeck 2003; Ronquist et al. 2011; Ronquist et al. 2012). We sampled the MCMC every 5000 generations and used Tracer 1.6 (Rambaut and Drummond 2007) to confirm stationarity and that a 10% burnin per independent analysis was appropriate. We combined results for simultaneous analyses using LOG COMBINER of the BEAST 1.8.0 software package (Drummond and Rambaut 2007; Drummond et al. 2012). We summarized the combined trees for each gene by selecting a maximum clade credibility tree with TREE ANNOTATOR, also of the BEAST 1.8.0 software package, and we obtained branch lengths for the selected tree using the median lengths from among the posterior distribution of trees. We also generated alternative summaries of the combined BI trees in GENEIOUS using 50% majority rule consensus with compatible groups with less than 50% support allowed. We visualized and rooted the final NJ, ML, and summarized BI trees in FIGTREE 1.4.0 (Rambaut and Drummond 2009). All final trees with clade support values are available in Dryad: <http://dx.doi.org/10.5061/dryad.n26nd>.

Examination of cuticular wax ultrastructure of leaves

For the morphological study of leaves, we examined individuals representing all four species comprising *Acer* sects. *Rubra* and *Hyptiocarpa*. We sampled leaves from all available specimens of *A. laurinum* and *A. pycnanthum* and four specimens each of *Acer rubrum* and *A. saccharinum* (Table 2). Our sampling of *A. rubrum* and *A. saccharinum*

Table 2. Specimens of *Acer* sections *Rubra* and *Hyptiocarpa* from which we obtained leaf material for study. All specimens are deposited at the United States National Herbarium (US; <http://collections.nmnh.si.edu/search/botany/>). Locations are given as state/province, county or with as much information as is available. Refer to Table 1 for taxon authorities.

Species	Collector name and number	Location	Stable URL to online specimen record
<i>Acer laurinum</i>	<i>Cult., in Hort. Bog. III, K 37</i>	Island of Java, Indonesia	http://n2t.net/ark:/65665/36a9749fe-e79a-456c-86bd-cb7d9ff2f6d2
<i>Acer laurinum</i>	<i>Sandkuhl 21296</i>	Benguet, Philippines	http://n2t.net/ark:/65665/3537d77b8-a9eb-460d-bd60-e16ce673f833
<i>Acer laurinum</i>	<i>Wen 13386</i>	Guangzhou (Cult.), China	http://n2t.net/ark:/65665/3b7b44164-fff4-4d7f-99a4-5d3d7adc2cbc
<i>Acer pycnanthum</i>	<i>Wilson 7729</i>	Island of Honshu, Japan	http://n2t.net/ark:/65665/369db8fd3-3e39-4f54-91d9-d8b631a4455f
<i>Acer pycnanthum</i>	<i>Wilson 6882</i>	Island of Honshu, Japan	http://n2t.net/ark:/65665/3744ee9ab-cddd-4013-9b52-cbda00eb02e3
<i>Acer pycnanthum</i>	unknown	Omi, Japan	http://n2t.net/ark:/65665/3255f723b-0e91-495a-94cc-e4d058c90352
<i>Acer rubrum</i>	<i>Arsene 11583</i>	Louisiana, USA	http://n2t.net/ark:/65665/37fa4f77a-b5eb-4c61-95ec-d967147cc603
<i>Acer rubrum</i>	<i>Thieret 22942</i>	Louisiana, USA	http://n2t.net/ark:/65665/3b286464c-77be-4e11-bf53-aa52e3f8299
<i>Acer rubrum</i>	<i>Stevens 2617</i>	Oklahoma, USA	http://n2t.net/ark:/65665/3e46d528c-6d03-4ab4-abfc-34c8b6821ceb
<i>Acer rubrum</i>	<i>Little & Ulmsted 186</i>	Oklahoma, USA	http://n2t.net/ark:/65665/39b4b7e0b-6ebc-46f9-b95a-d46b60d635ec
<i>Acer rubrum</i>	<i>Harris 2016-63</i>	West Virginia, USA	http://n2t.net/ark:/65665/329687fb5-8d3b-4d04-906e-c85a93d58334
<i>Acer saccharinum</i>	<i>Broum 8023</i>	New York, USA	http://n2t.net/ark:/65665/37328cb10-f106-4a89-a265-b7d8ade2aff7
<i>Acer saccharinum</i>	<i>Coville s.n.</i>	New York, USA	http://n2t.net/ark:/65665/3e52409ce-5840-4236-ad62-51434f8259a7
<i>Acer saccharinum</i>	<i>Richardson & Robertson 915</i>	Kansas, USA	http://n2t.net/ark:/65665/302ee09b6-3742-4616-b6fe-1af6c08c1e12
<i>Acer saccharinum</i>	<i>Norton 69</i>	Kansas, USA	http://n2t.net/ark:/65665/3f46187ca-30f8-442b-a449-c90d7a2f6807

included mid- and late-season specimens from two or more geographically distant parts of the ranges of the species and was designed to facilitate detection of population-level and seasonal variation in cuticular wax features (Sargent 1922; de Jong 1976; Delendick 1981). We obtained leaf samples near the center of leaves from sites adjacent to the midvein. The samples were dry when we obtained them from herbarium sheets. Air-dried samples, such as from herbarium sheets, are suitable for examination of cuticles without additional preparations and do not typically develop structural artifacts from drying or during examination with SEM (Pathan et al. 2010). We used specimens deposited at the United States National Herbarium (US) to obtain all leaf materials (Table 2).

We used a Hitachi TM300 scanning electron microscope (SEM) to examine the ultrastructure of the abaxial and adaxial surfaces of the leaves following standard protocols. We used a standard working depth of 10mm and took SEM micrographs under 15kv after determining that this intensity of the electron beam would not melt the cuticular wax. All of our scanning electron micrographs of the leaf surfaces are available from in Dryad: <http://dx.doi.org/10.5061/dryad.n26nd>.

Throughout, we apply the term ‘cuticle’ to all parts of the wax layer(s) above the cellulose wall of the epidermal cells. We acknowledge that the cuticle is a complex structure comprised of many well-delimited and/or intergrading components (reviewed in Fernández et al. 2016). However, our imaging is from a birdseye view, such that we are not able to distinguish among cuticular layers. We use terminology for cuticular wax forms following Barthlott et al. (1998). For discussion of leaf characters, especially veins, we follow The Manual of Leaf Architecture of the Leaf Architecture Working Group (1999).

Examination of specimens

We examined numerous herbarium specimens to complete this study. In particular, we examined specimens in person at US, South China Botanical Garden (IBSC), and the United States National Arboretum (NA). We also examined high resolution images of specimens online using JSTOR Global Plants (<http://plants.jstor.org/>) and SEINet (<http://swbiodiversity.org/seinet>).

Results

The aligned sequence matrices of ITS and *psbM* (<http://dx.doi.org/10.5061/dryad.n26nd>) each had alignment scores of 0.96. The ITS matrix comprised 564 characters, and *psbM* had 856 characters. Neither *psbM* nor ITS had significant differences in base composition (χ^2 crit = 10.2, $p=1.0$ and χ^2 crit = 23.8, $p=1.0$, respectively).

Phylogenetic analyses of ITS showed weak support for the monophyly of the clade comprising sects. *Rubra* and *Hyptiocarpa*: NJ BS 45%, BI posterior probability (PP) 0.78, ML BS 48%. The *psbM* data matrix had few informative characters to distin-

guish a clade of sects. *Rubra* and *Hyptiocarpa* from *Acer distylum* Siebold & Zucc. of the monotypic sect. *Distyla*. Sections *Rubra* and *Hyptiocarpa* formed a trivially supported clade in the NJ phylogeny. However, a clade of sects. *Rubra* and *Hyptiocarpa* included *A. distylum* in the ML phylogeny. The BI results from *psbM* highlight the low support for the relationships among sects. *Rubra*, *Hyptiocarpa*, and *Distyla* in that the maximum clade credibility summary showed a clade of sects. *Rubra* and *Hyptiocarpa*, while the majority rule summary showed *A. distylum* included in a clade with sects. *Rubra* and *Hyptiocarpa*. The concatenated data matrix of ITS and *psbM* yielded moderate support for a clade of sects. *Rubra* and *Hyptiocarpa* in NJ, ML, and BI analyses. The support for the *Rubra*-*Hyptiocarpa* clade was NJ BS of 45%, BI PP of 0.74 in the maximum clade credibility tree and of 0.71 in the majority rule topology, and ML BS of 74% (Fig. 2). All trees in which sect. *Rubra* and *Hyptiocarpa* form a clade suggest that *A. pycnanthum* is sister to a clade of *A. rubrum* and *A. saccharinum* and that *A. laurinum* is sister to the other three species.

Our examination of leaf surface features in *Acer rubrum* shows that the adaxial surface bears pavement cells that are generally ovoid in shape and have wavy, jig-saw puzzle-piece-like margins (Fig. 3A–B). Across the surfaces of the pavement cells, the cuticle comprises a smooth layer and forms a striate pattern (Fig. 3B). The abaxial surfaces of leaves in *Acer rubrum* bear a cuticular wax layer comprised of membranous platelets, for which the membranes sometimes coalesce into structures appearing as terraced or non-terraced wax splatters and finger-like extensions of the membranes are thin, polygonal (Fig. 3C–D).

Wax features of leaves of *Acer saccharinum* are similar to those of *A. rubrum*. Specifically, the adaxial surface bears wavy pavement cells (Fig. 4A–B) and the abaxial surface is covered by a cuticular wax layer comprised of membranous platelets with thin extensions (Fig. 4C–D). However, the wax splatter features formed by the coalescing of the membranous platelets appear larger and more frequent on the leaf surface (Fig. 4D).

Leaves of *Acer pycnanthum* bears wax similar to those of *A. rubrum* and *A. saccharinum* and show wavy pavement cells with striate cuticular wax (Fig. 5A–B). On its abaxial surface, *Acer pycnanthum* bears the coalescing membranous platelets (Fig. 5C–D). The platelets sometimes have rounded extensions instead of or alongside of polygonal ones (compare raw images provided in <http://dx.doi.org/10.5061/dryad.n26nd>).

In *Acer laurinum*, the cuticular smooth layer on the adaxial leaf surface has wrinkles that make it appear thicker than in species of sect. *Rubra* s.s. The smooth layer may be slightly or extensively wrinkled across the adaxial surface (Fig. 6A–B, also <http://dx.doi.org/10.5061/dryad.n26nd>), and it obscures the shapes of the pavement cells. The membranous platelets on the abaxial surface (Fig. 6C) are wide and do not taper into finger-like projections at their ends (Fig. 6D). The wax splatter feature is sometimes granular on its surface (Fig. 6D).

We did not detect differences in the leaf wax features based on geographic range or, in most cases, seasonality. However, we observed one late-season *Acer pycnanthum* specimen with some leaves partially lacking the silvery color on the abaxial surface

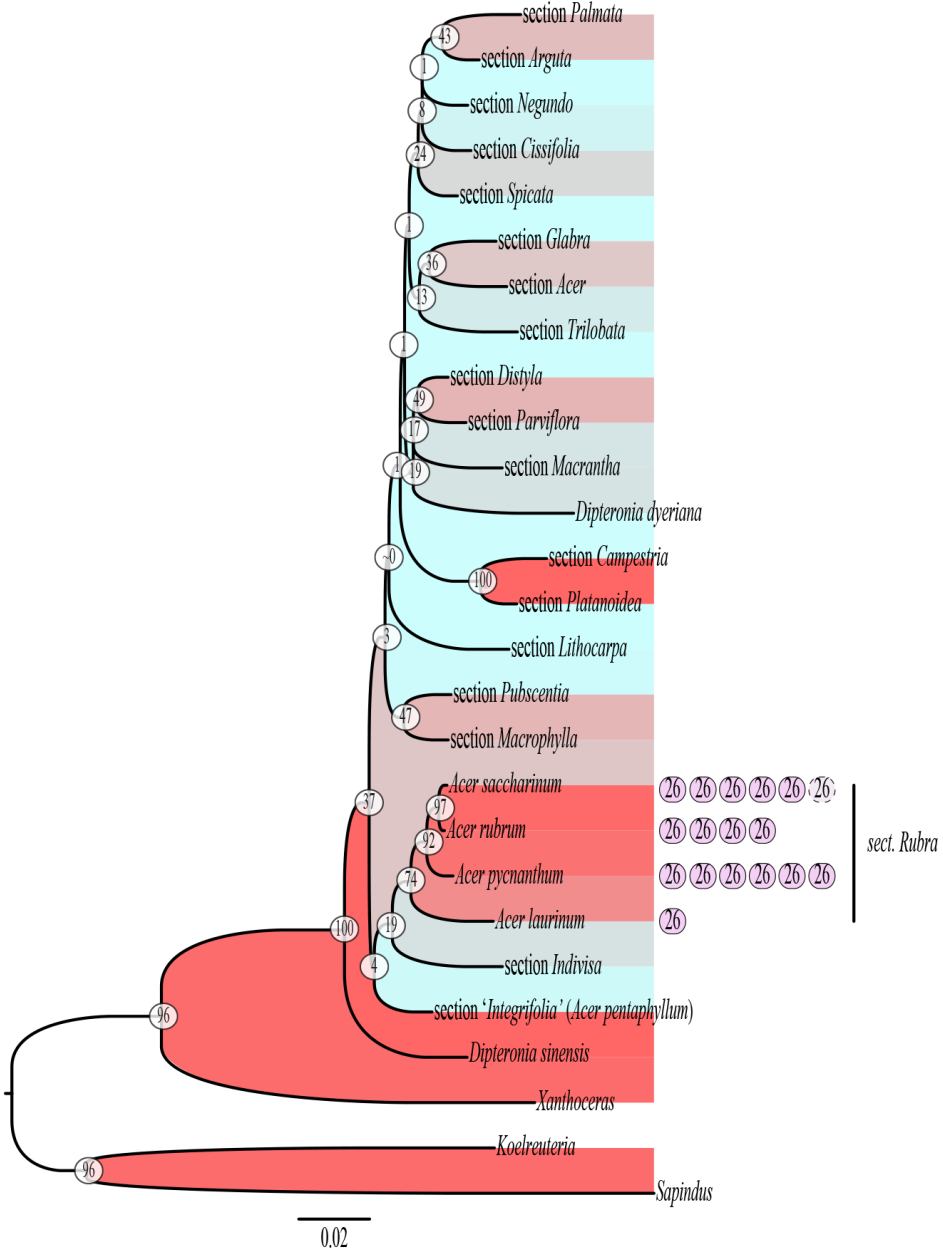


Figure 2. Maximum likelihood tree. Composite individuals represent sections except in the case of sects. *Rubra* and *Hyptiocarpa*, where composite individuals represent species. See Table 1 for GenBank accession numbers. Bootstrap values for nodes are shown in white circles. Purple circles to the right of species in sects. *Rubra* and *Hyptiocarpa* represent one set each of 26 chromosomes (i.e., $2n=26$) and show ploidy levels in sects. *Rubra* and *Hyptiocarpa* (e.g., *A. saccharinum* is tetraploid). Color coding of red and blue among branches shows relative support, respectively, from high ($=100\%$ BS) to low ($\sim 0\%$ BS). Branches are scaled according to the bar below the tree.

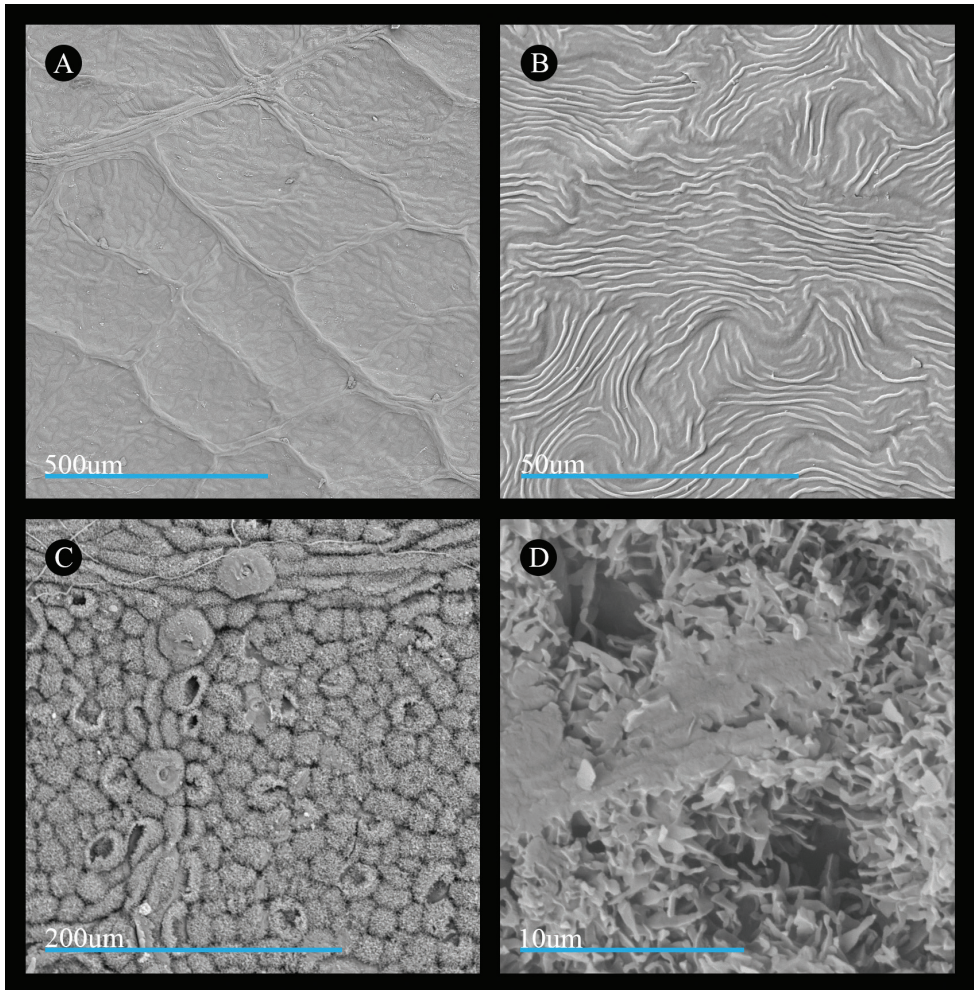


Figure 3. Micrographs of the leaf surfaces of *Acer rubrum*. **A** Adaxial surface showing cell shape and organization (Thieret 22942) **B** Adaxial surface showing cuticle (Harris 2016-63) **C**, Abaxial surface showing cell shape and organization of cells and stomata (Stevens 2617) **D** Abaxial surface showing cuticular wax (Thieret 22942). All leaf materials are from specimens deposited at US, and parenthetical information in this legend refers to the collector name and number for the source specimen.

(Fig. 7A). The silvery portion showed cuticular waxes similar to those on the mid-season leaves (Fig. 5B, see also <http://dx.doi.org/10.5061/dryad.n26nd>) while the non-silvery portion clearly lacked the plate-like cuticular features altogether (Fig. 7A, inset). However, on an equivalently late-season specimen of *A. rubrum* (Fig. 7B), we did not observe any parts of the leaves lacking the silvery component and membranous plates appeared the same as on the mid-season leaves (Fig. 7B, and insert).

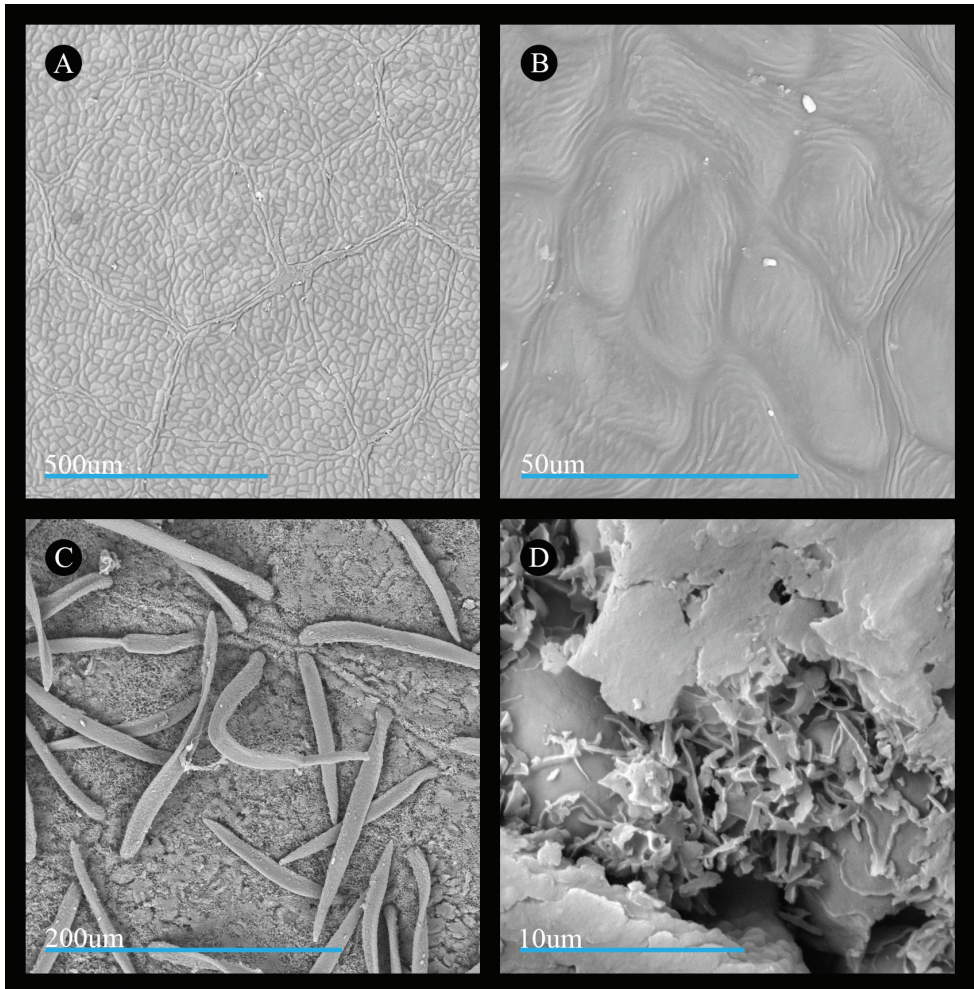


Figure 4. Micrographs of the leaf surfaces of *Acer saccharinum*. **A** Adaxial surface showing cell shape and organization (Norton 69) **B** Adaxial surface showing cuticle (Richardson & Robertson 915) **C** Abaxial surface showing cell shape and organization of cells and stomata (Brown 8023) **D** Abaxial surface showing cuticular wax (Coville s.n.). All leaf materials are from specimens deposited at US, and parenthetical information in this legend refers to the collector name and number.

Discussion

Phylogenetic relationship between *Acer* sections *Rubra* and *Hyptiocarpa*

Our phylogenetic results are congruent with previous molecular studies, which have found well-supported close relationships between *Acer* sects. *Rubra* and *Hyptiocarpa*. For example, Renner et al. (2008) reconstructed a phylogeny of Acereae from six chloroplast genes, including *psbM*, and using all four species comprising sects. *Rubra*

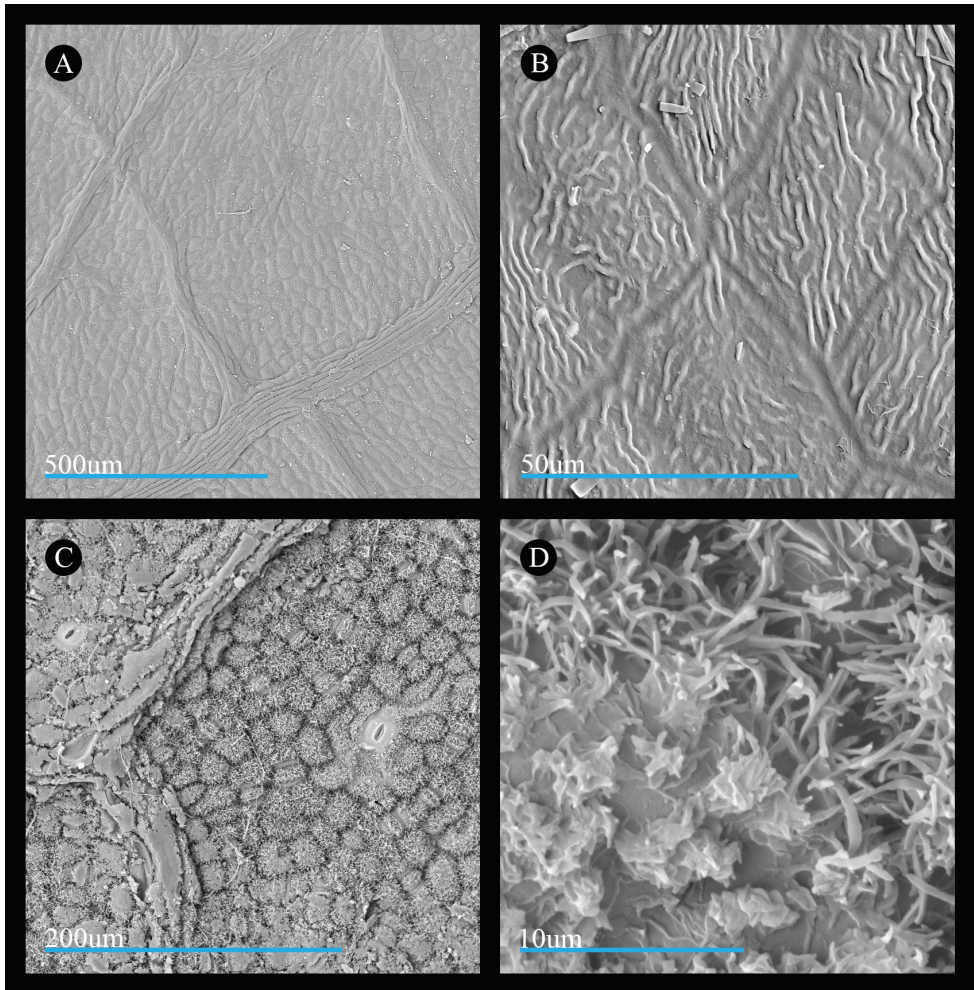


Figure 5. Micrographs of the leaf surfaces of *Acer pycnanthum*. **A** Adaxial surface showing cell shape and organization (Wilson 6882) **B** Adaxial surface showing cuticle (Collector unknown, s.n.) **C** Abaxial surface showing cell shape and organization of cells and stomata (Wilson 7729) **D** Abaxial surface showing cuticular wax (Wilson 6882). All leaf materials are from specimens deposited at US, and parenthetical information in this legend refers to the collector name and number..

and *Hyptiocarpa*. They found 99%BS support for a *Rubra-Hyptiocarpa* clade based on an ML analysis and showed the same relationships within the clade as in our analyses (Fig. 2; data in Dryad) (Renner et al. 2008). Similarly, Li et al. (2006) performed an NJ analysis of *Acereae* and found 100%BS support for a clade of sects. *Rubra* and *Hyptiocarpa* according to two chloroplast genes, including *psbM*, and Grimm et al. (2006) obtained the same result using MP and BI analyses of ITS. Tian et al. (2002) also recovered the *Rubra-Hyptiocarpa* clade from concatenated ITS and one chloroplast

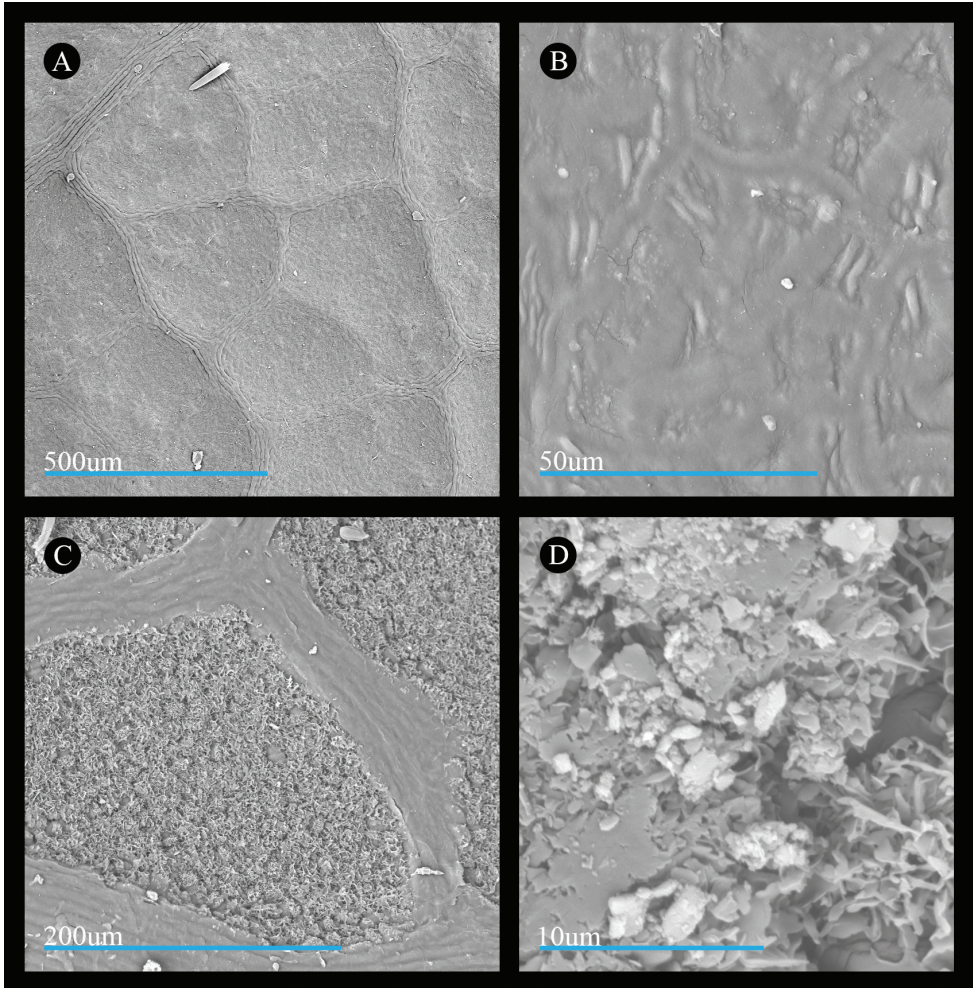


Figure 6. Micrographs of the leaf surfaces of *Acer laurinum*. **A** Adaxial surface showing cell shape and organization (*Cult., in Hort. Bog. III, K, 37*) **B** Adaxial surface showing cuticle (*Sandkuhl 21296*) **C** Abaxial surface showing cell shape and organization of cells and stomata (*Sandkuhl 21296*) **D** Abaxial surface showing cuticular wax (*Wen 13386*). All leaf materials are from specimens deposited at US, parenetical information in this legend refers to the collector name and number.

gene, *trnL-F*, except that they did not include *Acer pycnanthum* in their study. In addition to phylogenetic reconstruction, network analyses have also shown strong support for the grouping of the *Rubra-Hyptiocarpa* (Grimm et al. 2006; Renner et al. 2008). By comparison to other studies, we found relatively low molecular phylogenetic support for the *Rubra-Hyptiocarpa* clade, and this is probably due to stringent removal of uncertain portions of our alignments and because our chloroplast dataset is small, comprising only one gene.



Figure 7. Late-season specimens of *Acer* sect. *Rubrum* approaching leaf senescence. **A** *A. pycnanthum* with black arrow indicating silvery abaxial surface and green arrow indicating non-silvery surface. The inset in the upper left shows an SEM micrograph of a portion of an abaxial leaf surface from this specimen that lacks the silvery color such as the area referred to by the green arrow. Blue scale bar = 50µm. For an SEM micrograph showing a silvery portion of leaf surface from this specimen, see Figure 5C **B** *A. rubrum* exhibiting exclusively silvery abaxial leaf surface (upper, right leaf). The inset in the upper left shows an SEM micrograph of a portion of an abaxial leaf surface from this specimen bearing the characteristic silvery surface, and we did not observe late season leaves of *A. rubrum* lacking the silvery surface. Herbarium specimens deposited at US, and accession information visible in images. Detailed specimen records are available via the US online catalog (<http://collections.nmnh.si.edu/search/botany/>).

Similarities in leaf wax features between *Acer* sections *Rubra* and *Hyptiocarpa*

We observed very similar cuticular wax configurations on the abaxial leaf surfaces of species of sect. *Rubra* and in *A. laurinum*. In general, these configurations comprised membranous crystals that coalesce in formations appearing as wax splatters on the surface. We unexpectedly showed evidence that cuticles comprised of membranous plates are the source of the classic silvery appearance in sects. *Rubra* and *Hyptiocarpa* by showing that when cuticular wax formation is absent in *A. pycnanthum* (Fig. 7A), so is the silvery color. We expect that cuticular waxes are probably responsible for the silvery color in all species of sects. *Rubra* and *Hyptiocarpa*, and the relationship between the silver color and cuticular waxes has been previously noted and explored (e.g., Baker 1974; Caddah et al. 2012).

Some authors have speculated that cuticular wax configurations may be of limited taxonomic value, because they could vary with environment (Baker 1974; Mayeux et al. 1981). However, cuticular waxes have been informative in other groups (e.g., *Jatropha* Linnaeus, Dehgan 1980; *Rosa* Linnaeus sect. *Caninae*, Wissemann 2000; and *Aralia* Linnaeus, Wen 2011) and often have clear evolutionary significance (Eglinton and Hamilton 1967). Moreover, we did not find notable differences in the waxes among specimens collected in different parts of their geographic ranges or during different seasons (compare images at <http://dx.doi.org/10.5061/dryad.n26nd> from specimens of *Acer rubrum* and *A. saccharinum*). The striking cuticular waxes on the abaxial surfaces of all four *Rubra-Hyptiocarpa* species probably reflects descent from a common ancestor and could function in insect interactions (e.g., limiting insect walking on the abaxial surfaces; Baker 1974; Eigenbrode and Espelie 1995; Federle et al. 1997; Gorb et al. 2008; Müller 2008) or reducing water loss (Sutter and Langhans 1982; Clarke and Richards 1988).

The cuticle layer on the adaxial surface of *Acer laurinum* appears less similar to the species of section *Rubra*. While both sects. *Rubra* and *A. laurinum* have striations, these differ in the size of the striae, or ridges, which are wider and taller in *A. laurinum* (compare Fig. 6B with Figs 3B, 4B, 5B). Additionally, the size of the striae in *A. laurinum* makes the cuticle appear thicker than in the other species. A thick cuticle in *A. laurinum* would be consistent with its distribution in subtropical and tropical regions (Bloembergen 1948; van Gelderen et al. 1994; Xu et al. 2008), as tropical species often exhibit thick cuticles to reduce leaching via regular rainfall (Martin and Juniper 1970; Boeger et al. 2004). Striations of different widths and heights between *A. laurinum* and sect. *Rubra* may represent specialized adaptations to local conditions but the presence of striations may arise from a common genetic architecture. Recent studies on the genetic basis for cuticular wax phenotypes in model organisms such as *Sorghum* L. (Punnuri et al. 2017) and *Arabidopsis* Heynh. (Lee and Suh 2015) provide a foundation for future investigations of the evolutionary origins of cuticular wax forms in sects. *Rubra* and *Hyptiocarpa* and other maples.

Acer sects. *Rubra* and *Hyptiocarpa* cannot be united strictly based on the appearance of the abaxial surfaces of their leaves. Although this feature may have taxonomic value (Merrill 1941; Krause 1978; Delendick 1981) and it appears monomorphic in sects. *Rubra* and *Hyptiocarpa*, it also occurs elsewhere in the genus (van Gelderen et al. 1994). In particular, silvery or glaucous surfaces occur in most species of sect. *Pentaphyllum* and in some species of sect. *Acer*. Nevertheless, the taxonomic informativeness of cuticular waxes in *Acer* may warrant further investigation to compare both the fine features of ultrastructure and wax chemical composition especially within and among glaucous and non-glaucous sections and species.

Acer laurinum and other species of section *Hyptiocarpa*

Different taxonomic treatments of *Hyptiocarpa* do not all agree on species delimitation within the section. The large number of synonyms in *Hyptiocarpa* and confusion over the boundaries of species may reflect high variability and the need for additional field

work to elucidate species limits or intergradation (Bloembergen 1948; van Gelderen et al. 1994). The most recent treatment of sect. *Hyptiocarpa* in Flora of China (Xu et al. 2008) recognizes two species: *Acer laurinum* and *A. pinnatinervium*. *Acer pinnatinervium* is considered a synonym of *A. laurinum* by van Gelderen (1994) and in the Plant List (<http://www.theplantlist.org>), but its status within *Hyptiocarpa* merits discussion here.

According to Xu et al. (2008), *Acer laurinum* and *A. pinnatinervium* differ in fruit size, the number of primary veins per leaf, and their geographic distributions. *Acer laurinum* has fruits 4–7 cm and leaves with three primary veins, while *Acer pinnatinervium* has fruits 2–4 cm and only one primary vein, i.e., it is truly pinnately veined. The pinnate venation in *Acer pinnatinervium* may be particularly noteworthy, because most species of *Acer* have leaves with three main veins (Merrill 1941). Therefore, pinnate venation in *A. pinnatinervium* is considered the primary character for distinguishing it from *A. laurinum* (Merrill 1941). With respect to geographic distributions, Xu et al. (2008) report that *Acer laurinum* has a broader range, being found from southwestern China to India, Vietnam, Indonesia, and the Philippines, while *A. pinnatinervium* occurs in southwest China, Thailand, and India.

Closer examination of *Acer laurinum* and *A. pinnatinervium* shows that they intergrade on the number of primary veins. Some collections of *A. laurinum* (e.g., *Blume* 466, L; *Blume s.n.*, L) show strong basal acrodromous veins, while isotypes of *Acer pinnatinervium* (*F. Kingdon-Ward* 9102, A, BM) show pinnate venation with brochidodromous secondary veins near the leaf base. However, the holotype and isotype of *A. laurinum* (*F.W. Junghuhn s.n.*, L, U, respectively) each show variability in venation such that some leaves have acrodromous veins and others are pinnately veined with weak brochidodromous secondaries. We also observed this variability within a specimen of *A. laurinum* utilized in the SEM component of this study, *Cult., in Hort. Bog. III, K, 37* (see Table 2), and in many specimens that are ascribed to *A. pinnatinervium* and digitized in the Chinese Virtual Herbarium (<http://www.cvh.ac.cn/>). In the latter case, intra-individual variability of leaf veins may account for recent disagreements in the identities of specimens as either *A. laurinum* or *A. pinnatinervium* evidenced by the annotation labels. Based on these observations, we suspect that the number of primary veins is not sufficient to distinguish *Acer pinnatinervium* from *A. laurinum*, and combining the two species may be needed pending an additional study of more strategically sampled individuals.

Morphology of *Acer* sections *Rubra* and *Hyptiocarpa*

Leaves in sects. *Rubra* and *Hyptiocarpa*, hereafter sect. *Rubra sensus latu*, exhibit shapes that vary within and among species from elongate to orbicular (Fig. 1). *Acer laurinum* and *A. pinnatinervium* have highly elongate leaves, while, in *A. rubrum*, leaves vary from being orbicular (Fig. 1A) to having slight elongation (Fig. 8A). Similarly, leaves in mature *A. pycnanthum* may also possess roughly orbicular leaves (Fig. 1B) to leaves that



Figure 8. Elongate leaf shape in *Acer rubrum* and *A. pycnanthum*. **A–B** *A. rubrum* **C–D** *A. pycnanthum*. Unfortunately, there is no scale for the images of *A. pycnanthum*, but the leaf size is similar to that illustrated in Figure 1B. Herbarium specimens in **A** and **B** deposited at US, and accession information visible in images. Detailed specimen records are available via the US online catalog (<http://collections.nmnhs.si.edu/search/botany/>).

are highly elongated and nearly lacking lobes (Fig. 8C–D). In *A. saccharinum*, most individuals have leaves that are more-or-less orbicular (Fig. 1C), but some have elongated leaves (e.g., *Chaney 290*, LSU). Many species of *Acer* exhibit elongation of juvenile leaves, including in *A. saccharinum* (e.g., *Longbottom 8925*, DOV), *A. pycnanthum* (e.g., *Meyer 12513*, NA), and *A. rubrum* (Fig. 8B). Additionally, leaves and leaflets in other Sapindaceae are also often elongate (Acevedo-Rodríguez et al. 2011; Harris et al. 2017). Leaf elongation in seedlings of *Acer* may indicate an underlying genetic architecture in the genus and, consequently, ontogenic recapitulation (Haeckel 1866; e.g., Mishler 1998). Thus, while variable leaf shape in sect. *Rubra* does not unite its species, the tendency towards elongation is likely a noteworthy plesiomorphy in *Acer*.

Section *Rubra* s.l. has variable inflorescence architecture (Fig. 9). *Acer rubrum* (Fig. 9A–B), *A. pycnanthum* (Fig. 9C), and *A. saccharinum* (Fig. 9D) have inflorescences that are umbels (de Jong 1976; van Gelderen et al. 1994), while *A. laurinum* and allied taxa have inflorescences that may be racemes (*F.W. Junghuhn s.n.*, L) or paniculate thyrses (*Lindley, 418*, K) (de Jong 1976; van Gelderen et al. 1994). The umbels, which are unique within *Acer*, probably represent evolutionarily reduced racemes, while the racemes, which are more common in *Acer*, may represent reduced paniculate thyrses (de Jong 1976, Singer 2008). Inflorescences throughout sect. *Rubra* s.l. are almost exclusively lateral (Ohwi 1965, de Jong 1976, van Gelderen et al. 1994), although some authors report occasional terminal inflorescences in *A. pycnanthum* (van Gelderen et al. 1994; but contrast with *A. pycnanthum* in Ohwi 1965, de Jong 1976). While lateral inflorescences are common to other sections of *Acer*, exclusively lateral ones (or nearly so) occur only in sects. *Rubra*, *Lithocarpa*, and *Glabra*.

Species of sect. *Rubra* s.l. except *A. pycnanthum* may be monoecious or dioecious and exhibit labile sex expression among individuals (de Jong 1976; Primack and McCall 1986; Santamour 1993), and within-individual and within-clade labile sex expression occurs in some other groups of *Acer* and other Sapindaceae (Acevedo-Rodríguez et al. 2011; Renner et al. 2007). *Acer pycnanthum* is thought to be exclusively dioecious (de Jong 1976, Saeki 2008). Among monocious individuals of *Acer rubrum*, *A. saccharinum*, and *A. laurinum*, individual inflorescences are usually exclusively comprised of staminate or pistillate flowers. One prior study inferred that dioecy was ancestral in sect. *Rubra* s.l., but that inference was based on scoring *A. laurinum* as dioecious (Renner et al. 2007), which is not accurate (Bloembergen 1948; de Jong 1976; Xu et al. 2008). All flowers in sect. *Rubra* s.l. emerge from leafless buds, and this is a taxonomically informative trait that delimits some sections of *Acer* from others (de Jong 1976; van Gelderen et al. 1994).

Fruits in sect. *Rubra* s.l. also share many features (Fig. 10), especially from among those identified as taxonomically informative in a comprehensive study by Wolfe and Tanai (1987). We have observed that the fruits of all species in sect. *Rubra* have slightly inflated seed locules without keels, wings that are straight at the base, and mericarps forming an acute angle with respect to one another. Each of these traits tends to be monomorphic within sections. Each trait occurs in about half of all sections, but this suite of traits may be unique to sect. *Rubra* s.l. Additionally, species in sect. *Rubra* s.l.



Figure 9. Inflorescences of *Acer* sects. *Rubra* and *Hyptiocarpa*. **A** *Acer rubrum* with umbels of pistillate flowers **B** *Acer rubrum* with umbels of staminate flowers **C** *A. pycnanthum* with umbels of pistillate flowers **C** *A. saccharinum* with umbels of pistillate flowers. Note flowers with two, divided persistent styles **D** *A. pinnatinervium* with racemose thyrses. Specimens in A–D deposited at US, and specimen in D deposited at the British National Museum (BM). Accession information visible in images, and detailed specimen records are available via the US online catalog (<http://collections.nmnh.si.edu/search/botany/>) and at the data portal of BM (<http://data.nhm.ac.uk/>).

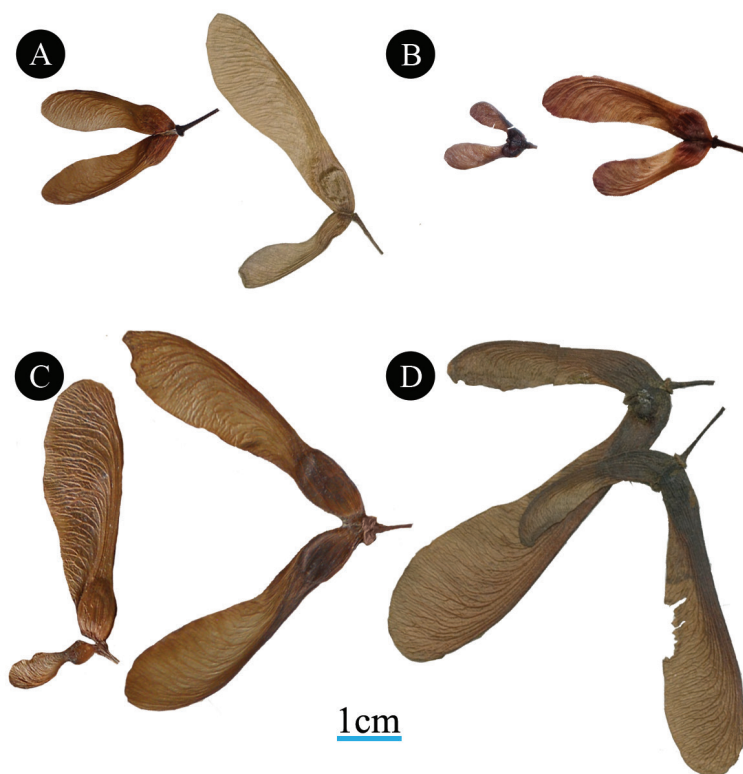


Figure 10. Fruits of species of *Acer* section *Rubra*. **A** *A. rubrum*. Specimen on left deposited at US National herbarium (US) with collection name and number: *Lilian 62*. Specimen on right deposited at Kew (K) as *Acer drummondii* Nutt. (= *A. rubrum*) with collection name and number: *Drummond 53*. Image of fruits obtained from image of specimen deposited in JSTOR Plants (<http://plants.jstor.org/>) **B** *A. pycnanthum*, used with attribution to Chinese Virtual Herbarium (<http://www.cvh.ac.cn/>); *Miyoshi Furuse 54050*, PE **C** *A. saccharinum* showing fruit with two fertilized ovules (upper) compared with one fertilized ovules and one partially developed, unfertilized ovule (lower). Specimens deposited at US with collection name and number: *Wolf s.n.* and *Pringle s.n.*, respectively **D** *A. laurinum* Specimen deposited at K with collection name and number: *Lindley, 418*. Image of fruits obtained from image of specimen deposited in JSTOR Plants. Scale bar of 1cm applies to all images.

are capable of producing partially developed seedless mericarps (Fig. 10), compared to complete or extremely minimal (e.g., roughly pinhead-sized) development in other species and sections (de Jong, 1976). The degree of development of seedless mericarps in *Acer* is well-characterized by de Jong (1976) and is taxonomically informative. Partially developed, seedless mericarps occur in about half of sections of *Acer*, and most sections are monomorphic for this trait. Fruits of sect. *Rubra* s.l. are highly variable in size within species with the largest fruits occurring in *A. saccharinum* and *A. laurinum* (Townsend 1972; van Gelderen et al. 1994; Xu et al. 2008).

Prior studies have proposed other plausible relationships for sect. *Hyptiocarpa* based on morphology. In particular, leaf morphology has often been used to link

sect. *Hyptiocarpa* with *Acer oblongum* Wallich ex de Candolle and its close relatives in sect. *Pentaphylla* or *Integrifolia* (Pax 1885; Momotani 1962; Fang 1966). *Acer oblongum* has entire, unlobed elongate leaves and silvery abaxial surfaces (van Gelderen et al. 1994) that are similar to leaves in *A. laurinum*. Nevertheless, any association between *Acer oblongum* and sect. *Hyptiocarpa* has not been supported by molecular phylogenies, which show that *Acer oblongum* is associated with sect. *Pentaphyllum* and distant from sect. *Rubra* (Suh et al. 2000; Renner et al. 2008). Morphologically, *A. oblongum* differs from *A. laurinum* by having mostly terminal inflorescences and by flowers and leaves arising from the same buds (van Gelderen et al. 1994). Additionally, the waxes of *A. oblongum* may differ from those in sect. *Rubra* by extending partially onto the midrib. While we made this observation on many specimens at IBSC, we used a low magnification hand lens, and a more detailed study using higher magnification may be warranted. Another possible association for sect. *Hyptiocarpa* was with sect. *Lithocarpa*, which has a relatively large number of bud scales, axillary inflorescences from leafless buds, and insertion of stamens on a staminal disk (Ogata 1967); features that are also shared with sect. *Rubra* s.s., except for stamen insertion (Pax 1885; Ogata 1967; de Jong 1976; van Gelderen et al. 1994). In sect. *Rubra* stamens are inserted outside of the disk or the disk is absent in some individuals of each species (van Gelderen et al. 1994). Thus, the disk may be relatively labile within sect. *Rubra* s.s. and in sect. *Rubra* s.l. Section *Hyptiocarpa* differs from sect. *Lithocarpa* (except *A. macrophyllum* Pursh.) by having wood rays 3-4 cells wide rather than cells wide. Overall, in prior taxonomic work, recognition of a distinct sect. *Hyptiocarpa*, seems more motivated by uncertainties about its affinities (Ogata 1967; de Jong 1976; Delendick 1981; Wolfe and Tanai 1987; van Gelderen et al. 1994) than affirmation of its significant uniqueness within *Acer* (e.g., contrasted with *A. carpinifolium* and *A. negundo* Linnaeus).

Section *Rubra* s.l. and evolutionary radiation

Section *Rubra* s.l. may have radiated out of the tropics and into temperate areas of Japan and North America based on our phylogenetic results (Fig. 2) and results presented in other molecular phylogenetic studies (Grimm et al. 2006; Renner et al. 2008). In particular, results suggest that *A. laurinum* is the earliest diverging species within sect. *Rubra* s.l. An out- of-the-tropics radiation in sect. *Rubra* s.l. may have been accompanied by, or even spurred by, polyploidization. According to the literature, *Acer laurinum* is diploid ($2n=26$), *A. saccharinum* is tetraploid, *A. pycnanthum* is hexaploid, and *A. rubrum* includes hexaploid, heptaploid (rarely), and octaploid individuals (Fig. 2), and these counts reflect attempts to avoid hybrid individuals and use materials originating from wild populations (Löve 1971; Santamour 1965; van Gelderen et al. 1994; Chromosome Count Database, <http://ccdb.tau.ac.il/home/>). Polyploidization is assumed to have played a role in adaptation to less equitable environments in some plant groups, and polyploidy sometimes shows clear positive correlation with latitude (Beaton and

Hebert 1988). Future studies may examine the timing of evolutionary radiation, such as by using fossils and divergence time dating, to better understand possible correlations between ploidy level and past environments.

Synopsis of *Acer* section *Rubra* s.l.

Acer sect. *Rubra* Pax, Bot. Jahrb. Syst. 6: 326. 1885.

Acer sect. *Hyptiocarpa* W.P. Fang, Acta Phytotax. Sin. 11: 172. 1966.

Acer sect. *Laurina* Ogata, Bull. Tokyo Univ. Forests 63: 151. 1967.#

Type species. *Acer rubrum* Linnaeus.

Description. Trees, deciduous or evergreen, with labile sex expression ranging from monoecy to dioecy (possibly exclusively dioecious in *A. pycnanthum*). Wood distinctly ring-porous, rays 1–4(10) cells wide. Bud scales imbricate, decussate, in pairs of 4–11. Leaves entire, unlobed, or 3- or 5-lobed, elliptic to ovate, toothed or entire, glaucous to blue-colored beneath; cuticular waxes of leaves comprising a smooth layer on the adaxial surface and bearing membranous platelets and wax splatter features abaxially; primary veins 1 or 3, 5 in 5-lobed individuals of *A. saccharinum*; petioles sometimes turning red (e.g., new growth, late season). Inflorescences axillary (rarely terminal) from leafless buds, usually emerging before leaves, panicle thyrses, racemes, or umbels. Sepals 5. Petals 0 or 5, red, red-green, or green when present. Stamens 5–12, inserted on (*A. laurinum* and *A. pinnatinervium*) or outside of staminal disk, disk sometimes reduced or absent (*A. rubrum*, *A. saccharinum*, *A. pycnanthum*). Carpels 2. Fruits schizocarps with partially inflated seed locules, sometimes turning red during maturation, partitioning wall generally narrower than the seed locules; mericarps diverging from each other at less than 90°, wings straight to slightly convex on the proximal (vein-dense) side, curved on the distal side. Some fruits seedless and partially developed at maturity.

Five species showing a disjunct distribution between eastern and southeastern Asia (3 spp.) and eastern North America (2 spp.), a common biogeographic pattern among Northern Hemisphere plant groups (Donoghue and Smith 2004; Harris et al. 2013, 2017; Li 1952; Wen 1999, 2001; Xiang et al. 2015).

Acer laurinum Hasskarl, Tijdschr. Natuurl. Gesch. Physiol. 10: 138. 1843.

Acer javanicum Junghuhn, 1841

Acer niveum Blume, 1847

Acer cassiifolium Blume, 1847 (as *cassiaefolium*)

Acer philippinum Merrill, 1906

Acer garrettii Craib, 1920

Acer decandrum Merrill, 1932

Acer chionophyllum Merrill, 1941

- Acer longicarpum* Hu & W. C. Cheng, 1948
Acer macropterum T. Z. Hsu & H. Sun, 1997
Acer pinnatinervium Merrill, Brittonia 4: 109. 1941.
Acer machilifolium Hu & Cheng, 1948
Acer jingdongense T. Z. Hsu, 1983
Acer pycnanthum K. Koch, Ann. Mus. Bot. Lugduno-Batavi 1: 250. 1864.
Acer rubrum Linnaeus, Sp. Pl. 1055. 1753.
Acer carolinianum Walter, 1788
Acer barbatum Michaux, 1803, *pro parte*
Acer sanguineum Spach, 1834
Saccharodendron barbatum (Michaux) Nieuwland, 1914, *pro parte*
Rufacer carolinianum (Walter) Small, 1933
Rufacer rubrum (Linnaeus) Small, 1933
Acer saccharinum Linnaeus, Sp. Pl. 1055. 1753.
Acer sylvestre Young, 1783
Acer glaucum Marshall, 1785
Acer rubrum Linnaeus var. *pallidum* Aiton
Acer dasycarpum Ehrhart, 1789
Acer eriocarpum Michaux, 1803
Acer tomentosum Steudel, 1821
Acer coccineum F. Michaux
Saccharosphenclamnus saccharina (Linnaeus) Nieuwland, 1914
Argentacer saccharinum (Linnaeus) Small, 1933

Conclusions

Based on evidence from molecular phylogeny, morphology, and leaf ultrastructure, we propose uniting sects. *Rubra* and *Hyptiocarpa* within *Acer* sect. *Rubra*. Our molecular phylogenetic results are in agreement with prior studies, which suggest that *Acer* sects. *Rubra* and *Hyptiocarpa* are sisters. Within these sections, species share taxonomically important characteristics including leaves with silvery abaxial surfaces resulting from similar cuticular wax structures, typically lateral inflorescences, labile sex expression, partial development of seedless fruits, and many aspects of fruit morphology. The unity of these sections yields better and more complete understanding their evolutionary and biogeographic history. We speculate that sect. *Rubra* s.l. radiated out of the tropics and that the radiation coincided with polyploidization.

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Integrative research identifies 71 new plant species records in the state of Rio Grande do Norte (Brazil) and enhances a small herbarium collection during a funding shortage

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Abstract

A National Forest Inventory (NFI) encompassing the entire territory of Brazil is in progress. It is coordinated and promoted by the Brazilian Forest Service of the Ministry of Environment. In each state, the NFI collaborates with local herbaria by receiving collected plant material and performing species identification. Consultants are hired by the NFI and work at the local herbaria under the supervision of a curator. In exchange for curatorial assistance, the NFI provides equipment and consumables for the herbarium. Other public projects collaborating with NFI are Refflora and the Brazilian Biodiversity Infor-

mation System (SiBBR). Both projects have online platforms that seek to connect herbaria and make all their data freely available, including high quality digital images of specimens. Through inter-institutional collaboration, the joint interests of NFI, Refflora, SiBBR and local herbaria have improved collections, expanded the online Refflora database, and provided the NFI with verified species lists. These strategic uses of public funding are positively affecting Botany, particularly during a period of economic crisis and cuts in research. Here, we illustrate the increase in floristic knowledge through the improvement of a herbarium collection in Rio Grande do Norte (RN) – the Brazilian state with the lowest levels of plant richness. We report 71 new occurrences of vascular plants for RN, belonging mainly to the Poaceae, Fabaceae and Malvaceae. Most of the species with new occurrences have a Neotropical distribution (21 spp.) and only seven are restricted to the Brazilian Northeast. Our findings highlight previous gaps in RN's floristic knowledge. The partnership NFI, Refflora, SiBBR and the UFRN herbarium improved herbarium curation, digital collection, and quality of data. Finally, a fellowship provided by Refflora and SiBBR allowed improving curation by distributing duplicates and incorporating the Herbarium of Câmara Cascudo Museum.

Keywords

Brazil Flora Group, Flora, floristics, Herbarium collection, Inventory, Refflora, SiBBR, IFN

Introduction

A National Forest Inventory (NFI) covering all of Brazil is currently in progress. The NFI is a major undertaking by the Brazilian government, specifically, the Brazilian Forest Service, a public section of the Ministry of Environment, to periodically gather information about the forests and land cover of Brazil, through a systematic sampling of the territory using a 20 km × 20 km grid. In each state, the NFI invites local herbaria to receive and identify the collected specimens. Consultants are hired by the NFI and stay at the local herbarium under the supervision of a curator to identify plants to species. In exchange for this curatorial assistance, the NFI helps the herbarium by providing equipment and consumables. In Rio Grande do Norte (RN), the NFI started in 2014. Two additional public projects that are working with the NFI are Refflora and the Brazilian Biodiversity Information System (SiBBR). Refflora and the SiBBR are online platforms that connect herbaria, making data and high quality images of specimens in their collections freely available. The main goal of Refflora is to complete the Flora do Brasil 2020 online project, which relies on specimen data and images from herbaria in Brazil, the USA, and Europe. Inter-institutional collaboration serves the interests of NFI, Refflora, SiBBR and local herbaria, improving collections, expanding the Refflora database, and providing the NFI with accurate lists of plants. In this paper, we discuss the details and results of a four-part collaboration that makes strategic use of public funding to positively impact the study of Botany during tough economic times.

Rio Grande do Norte (RN) is a Brazilian state that consists of two phytogeographical domains: Dry Woodlands (Caatinga) and Atlantic Forest (Floresta Atlântica). The savanna (Cerrado) vegetation is scattered in small patches throughout the state. Different vegetation types occur within these phytogeographical domains: deciduous, semi-deciduous, subperennial and seasonal mixed palm forest (dominated by *Copernicia*

prunifera (Mill.) H.E. Moore), dunes and coastal sand plain vegetation (restinga), xeric rocky outcrops, natural and anthropic fields, mangroves, saline desert and aquatic vegetation (SUDENE 1971). Both of the main phytogeographical domains in RN have been profoundly altered by human activities. The Atlantic Forest, where most of the sugar cane cultivation has been done for centuries, is fragmented and degraded and urgently needs ecological restoration. Its remaining coverage varies from 8–17%, depending on whether mangroves and restinga coastal plains are included or excluded in the estimate (Maciel et al. 2011). The Caatinga has lost 45% of its original coverage in RN (C. Fonseca, Dept. Ecology, UFRN, pers. com.); what remains is within a few protected areas.

The *Lista de Espécies da Flora do Brasil* (Forzza et al. 2010, 2012) and the *Checklist das plantas do Nordeste Brasileiro* (Barbosa et al. 2006) gathered preliminary knowledge of the flora of RN. Recent taxonomic work has complemented this knowledge by focusing on specific taxonomic groups or on floristic studies. Groups that have been studied recently include *Chamaecrista* (Queiroz and Loiola 2009), Turneraceae (Rocha et al. 2012), *Paspalum* (Oliveira et al. 2013a), Leguminosae-Papilionoideae (São-Mateus et al. 2013), Erythroxylaceae (Costa-Lima et al. 2014), Capparaceae (Soares-Neto and Jardim 2015), *Cyperus* (Ribeiro et al. 2015a), Fabaceae (Amorim et al. 2016), and Bignoniaceae (Colombo et al. 2016). Recent floristic studies focused on specific areas or vegetation types, such as the deciduous and semi-deciduous forests (Cestaro and Soares 2004; 2008), savanna (Oliveira et al. 2012), riparian vegetation (Oliveira et al. 2013b, Ribeiro et al. 2014) and the herbaceous vegetation in Seridó (Santana and Souto 2006, Amorim et al. 2006, Ferreira et al. 2009, Queiroz et al. 2015). Furthermore, field work in RN has produced new records (e.g. Versieux et al. 2013a, 2013b). It is likely that the historically limited number of herbaria (only two in Index Herbariorum), graduate programs focused on biodiversity, and taxonomists may have led to insufficient sampling and underestimation of the true taxonomic diversity of the state.

The most recent account listed 1,222 species of angiosperms in RN, only five of which are endemic to the state (BFG 2015, *Flora do Brasil* 2020): *Aspilia procumbens* Baker (Asteraceae), *Arachis seridoensis* Valls et al. (Fabaceae), *Sida macaibae* Mont. (Malvaceae), *Eugenia pipensis* A.R.Lourenço & B.S.Amorim (Myrtaceae), and *Gouinia virgata* (J. Presl) Scribn. (Poaceae). The growing knowledge of flora in RN is striking and is illustrated by estimates of species richness. In 2010 the RN list included 707 species of angiosperms (Forzza et al. 2010), while in 2015 this number nearly doubled to 1,222 species – a 73% increase in five years (BFG 2015). Research investments that supported these results include the participation of researchers in inter-institutional projects, the creation of two new graduate programs in biodiversity in the largest university of the state (UFRN), and an increase in the number of botanical monographs. We also expect an increase in the publication of floristic studies in the next few years, since many recently-collected data are still in preparation.

Although the UFRN herbarium is a small collection (~25,000 specimens), it is the most representative of RN's flora. The objective of this paper is to describe the progress in the RN floristic knowledge after joint efforts dedicated to the NFI in RN. Together,

IFN, SiBBR and Refflora projects have been addressing poor species coverage and lack of investments in botanical collections. Having completed this field inventory, we can show the new species records for the RN flora, and whether these species are restricted to the northeast of Brazil or else are widely-distributed species that have been previously overlooked in RN. Finally, we report how participation in this joint initiative has influenced and affected the infrastructural legacy of the UFRN herbarium.

Material and methods

The NFI fieldwork in RN was carried out from March to October 2014 by private environmental consultants, and specimens began to be deposited in the UFRN herbarium in December 2014. The sampling units of the NFI are distributed according to the National Sampling Points Grid (*Grade Nacional de Pontos Amostrais* – GNPA), established by the Brazilian Forest Service. The grid density is 20×20 km, covering all of Brazil (IFN 2012). A total of 133 sampling units, called conglomerates, were placed systematically throughout RN. A conglomerate is composed of four subunits (20×50 m), which are established in the field following magnetic cardinal directions, and radiating 50 m from a central point. Inside each subunit, representative specimens of each species of herbaceous and woody plants were collected following specific protocols for Caatinga and Atlantic Forest (IFN 2012). Biophysical data including necromass, litter and soil characteristics, as well as socio-environmental data were also collected in each conglomerate (Freitas et al. 2016). Our summaries presented here are based only on the species richness data from conglomerates, as well as on the new occurrences indicated by specialists that visited our collections or received duplicates of material previously collected and deposited in UFRN herbarium.

A total of 556 voucher specimens were collected and analyzed to estimate the number of new occurrences for RN. All specimens collected were identified at the UFRN Herbarium using appropriate taxonomic literature and floras, comparisons with specimens identified by specialists, or direct determination by taxonomic specialists. We also incorporated the collection of the Câmara Cascudo Museum (MCC), which used to be an independent collection within UFRN. The MCC collection was partially revised by a technician provided by Refflora and SiBBR projects. All vouchers from the NFI were deposited at UFRN (including non-fertile material) and duplicates were sent to other herbaria (RB, HUEFS, UFP, SP, MG; acronyms follow Thiers continuously updated). Furthermore, during the project, the entire UFRN herbarium collection was digitalized into high quality images that are now available in Jabot platform <http://ufrn.jbrj.gov.br>.

We used *Lista de Espécies da Flora do Brasil* (<http://floradobrasil.jbrj.gov.br>), now updated to *Flora do Brasil 2020*, to determine whether species identified were new records for the state. Though *Flora do Brasil 2020* should be continuously updated, we highlight new records in our list, in case that new occurrences reported in scientific literature have been missed.

To get a better picture of the flora of RN, we checked whether the new occurrences are taxa with broad or restricted ranges, as this information may indicate the degree to which they are absent from collections. We defined five categories of distribution according to geographical and political boundaries to infer whether new species records had a distribution restricted to the northeast of Brazil or else they are more widely distributed: 1. Pantropical (“Cosmopolite”): occurring in many places even outside of the tropics, 2. American: occurring all over the Americas, 3. Neotropical: occurring in the Neotropical region, 4. Brazilian: occurring in many states of Brazil – not exclusive to the Northeast region, 5. Northeast: occurring in the Northeast region of Brazil. In addition, we provide comments about each new recorded taxon, including the phyto-geographical domains and municipalities where it occurs in RN. Maps were created in QGIS 2.14 (QGIS Development Team 2016) using TEOW (Terrestrial Ecoregions of the World) as a cartographic base (Olsen et al. 2001).

The Brazilian states are abbreviated as follows: AC: Acre, AL: Alagoas, AP: Amapá, AM: Amazonas, BA: Bahia, CE: Ceará, DF: Distrito Federal, ES: Espírito Santo, GO: Goiás, MA: Maranhão, MT: Mato Grosso, MS: Mato Grosso do Sul, MG: Minas Gerais, PA: Pará, PB: Paraíba, PR: Paraná, PE: Pernambuco, PI: Piauí, RJ: Rio de Janeiro, RN: Rio Grande do Norte, RS: Rio Grande do Sul, RO: Rondônia, RR: Roraima, SC: Santa Catarina, SP: São Paulo, SE: Sergipe, TO: Tocantins.

Results

The NFI sampled 133 conglomerates in RN, including 127 in Caatinga and six in Atlantic Rainforest (Figure 1). The sampling covered 86 out of a total of 167 municipalities in the state. We recorded a total of 556 specimens, 285 species and 57 families. The Cerrado was not sampled.

We found 71 newly-occurring species (Table 1) in RN, 43 of which were a result of the NFI inventory (see Fig. 1, overlapping new records and conglomerates points) and 28 of them resulted from additional research developed at the UFRN herbarium and the MCC collection. These new occurrences include species from 21 families, most of them belonging to Poaceae (14 spp.), Fabaceae (13), Malvaceae (13) and Cyperaceae (7). New occurrences have been reported in 55 different municipalities in RN and the municipalities with the highest number of new occurrences were Canguaretama (7 spp.), Ceará-Mirim (6), and Macaíba (4). Most newly-occurring species have a Neotropical distribution (21) and only seven species are restricted to the Northeast of Brazil (Table 2).

Considering the curatorial improvement of the UFRN collection we highlight the merging of the Museu Câmara Cascudo into UFRN herbarium. The specimens from the Museu Câmara Cascudo included 405 angiosperms (54 families), and 1,224 macroalgae (31 families). Algae were not studied in this work, but we indicate the new occurrences among angiosperms (Table 2).

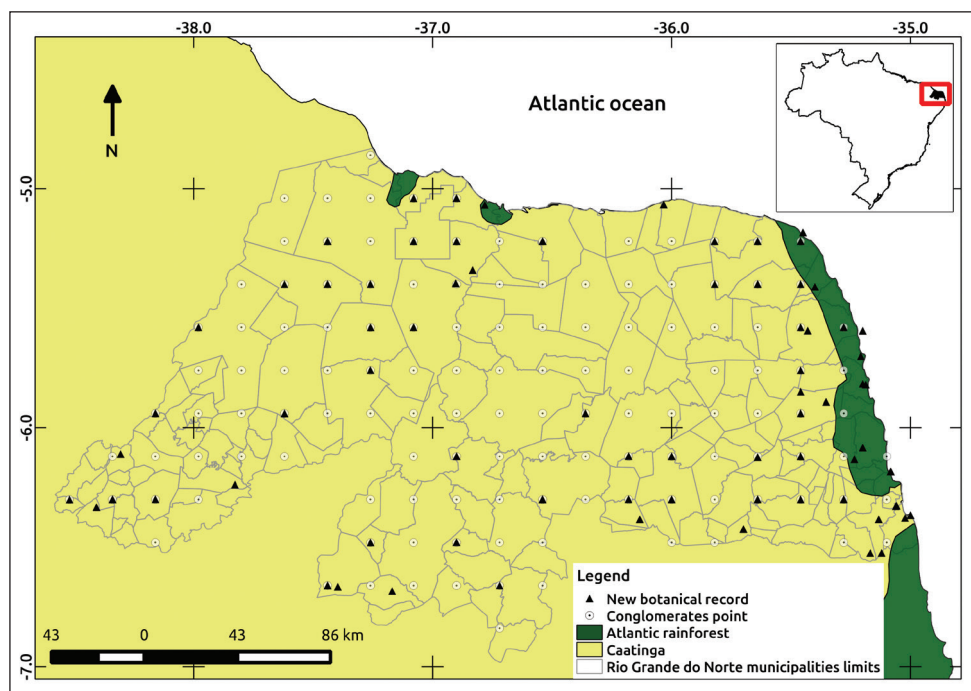


Figure 1. Map of Rio Grande do Norte state and municipalities, phytogeographical domains (Atlantic Rainforest and Caatinga), conglomerates points and new botanical records.

Distribution, phytogeographical domain, and habitats for each new species recorded in RN

AMARANTHACEAE

Froelichia humboldtiana (Roem. & Schult.) Seub.

This species occurs in Brazil and Venezuela (Funk et al. 2007, Marchioretto et al. 2005). Inside Brazil it occurs in AL, BA, CE, GO, MG, PB, PE, and PI states in the Caatinga phytogeographical domain (Marchioretto et al. 2005, Marchioretto 2015). In RN, it inhabits anthropic areas with sandy and stony soils.

ANACARDIACEAE

Spondias purpurea L.

This species is widely distributed from North and Central America to Brazil, occurring in dry or semi-deciduous forests (Mitchell and Daly 2015). In Brazil, it occurs in AC,

Table 1. List of new floristic records of the National Forest Inventory from Rio Grande do Norte state, Brazil. *Sterile specimen **Species previously cited in the literature but not in *Flora do Brasil* 2020.

Family	Species	Voucher	Municipality	Distribution
Amaranthaceae	<i>Froelichia humboldtiana</i> (Roem. & Schult.) Seub.	Silva, A.F. 43; Moura, E.O. 125	Serra do Mel; Timbaúba dos Batistas	Neotropical
Anacardiaceae	<i>Spondias purpurea</i> L.	Gonçalves, F.B. 447*	Ceará-Mirim	Neotropical
Asteraceae	<i>Stilpnopappus laiseae</i> R.Barros & R.Estevés	Silva, A.F. 54	Serra Negra do Norte	Northeast
Burseraceae	<i>Protium heptaphyllum</i> (Aubl.) Marchand	Gonçalves, F.B. 405; Dantas, A. 191	Canguaretama; Touros	Neotropical
Cactaceae	<i>Melocactus ernestii</i> Vaupel	Souza, A.C.D. 13	Serra de São Bento	Brazilian
Celastraceae	<i>Maytenus acanthophylla</i> Reissek	Jardim, J.G. 6393	Serra de São Bento	Brazilian
Chrysobalanaceae	** <i>Hirtella ciliata</i> Mart. & Zucc.	Gonçalves, F.B. 417; 418; 441; MCC	Touros; Pureza	Neotropical
Chrysobalanaceae	** <i>Hirtella racemosa</i> Lam.	Santos, L.A.S. 1268	Ceará-Mirim	Neotropical
Cyperaceae	<i>Bacquerelia cymosa</i> Brongn.	Roque, A.A. 1375	Canguaretama	Neotropical
Cyperaceae	<i>Eleocharis flavescens</i> (Poir.) Urb.	Jardim, J.G. 5823	Extremoz	American
Cyperaceae	<i>Eleocharis maculosa</i> (Vahl) Roem. & Schult.	Jardim, J.G. 5624	Extremoz	American
Cyperaceae	<i>Eleocharis montana</i> (Kunth) Roem. & Schult.	Oliveira, A.C.P. 1357; Roque, A.A. 929	Ceará-Mirim; São João do Sabugi	American
Cyperaceae	<i>Rhynchospora caracasana</i> (Kunth) Boeckeler	Moura, E.O. 149; Ribeiro, A.R.O. 48	Caraúbas; Canguaretama	Neotropical
Cyperaceae	<i>Rhynchospora gigantea</i> Link	Jardim, J.G. 6756	Canguaretama	American
Cyperaceae	<i>Scleria macrophylla</i> J.Presl & C.Presl	Jardim, J.G. 6757	Canguaretama	American
Euphorbiaceae	<i>Bernardia sidosides</i> (Klotzsch) Müll. Arg.	Dantas, R. 63; Silva, A.F. 20	Currais Novos; Açu	Neotropical
Euphorbiaceae	<i>Croton campestris</i> A.St.-Hil.	Gonçalves, F.B. 423	Parazinho	Neotropical
Euphorbiaceae	<i>Ditaxis malpighiacea</i> (Ule) Pax & K.Hoffm.	Gonçalves, F.B. 371	Macaíba	Brazilian
Euphorbiaceae	<i>Manihot esculenta</i> Crantz	Gonçalves, F.B. 349	Serra Caiada	Pantropical
Fabaceae	** <i>Amburana cearensis</i> (Allemão) A.C.Sm.	Gonçalves, F.B. 465*; Silva, A.F. 30*; Dantas, R. 78*	Campo Redondo; Upanema; Venha Ver	Neotropical
Fabaceae	** <i>Ancistrotropis peduncularis</i> (Kunth) A.Delgado	Silva, A.F. 60	Florânia	Neotropical

Family	Species	Voucher	Municipality	Distribution
Fabaceae	<i>Bauhinia dubia</i> G.Don	Moura, E.O. 218	Areia branca	Brazilian
Fabaceae	** <i>Dioclea violacea</i> Mart. ex Benth	Borges, S. 257	Natal	American
Fabaceae	<i>Calliandra depauperata</i> Benth.	Dantas, R. 3; 103*	Macau; Mossoró	Brazilian
Fabaceae	<i>Calliandra sessilis</i> Benth.	Dantas, R. 102*	Macau	Brazilian
Fabaceae	<i>Inga</i> cf. <i>vena</i> Willd.	Dantas, R. 47A*	Jardim de Piranhas	Neotropical
Fabaceae	** <i>Mimosa invisa</i> Mart. ex Colla	Silva, A.F. 100	Riacho de Santana	Neotropical
Fabaceae	** <i>Parkinsonia aculeata</i> L.	Silva, A.F. 91; Dantas, R. 61; Santos, L.A.S. 1246	Pilões; São José do Seridó; Santo Antônio	Pantropical
Fabaceae	** <i>Poincianaella bracteosa</i> (Tul.) L.P. Queiroz	Silva, A.F. 5; Dantas, A. 295	Mossoró; Touros	Neotropical
Fabaceae	** <i>Poincianaella pyramidalis</i> (Tul.) L.P. Queiroz	Dantas, R. 27; Silva, R.E. 7	Governador Dix-Sept Rosado; Caraubas	Brazilian
Fabaceae	** <i>Sylosanthus humilis</i> Kunth	Dantas, R. 66; Silva, A.F. 93	Currais Novos; Marcelino Vieira	Neotropical
Fabaceae	** <i>Trischidium molle</i> (Benth.) H.E. Ireland	Dantas, R. 33*; Gonçalves, F.B. 431; Santos, L.A.S. 1267	Carnaubais; Ceará-Mirim; Pureza	Brazilian
Lecythidaceae	<i>Eschweilera ovata</i> (Cambess.) Mart. ex Miers	Dantas, A. 241	Canguaretama	Brazilian
Malvaceae	<i>Ayenia erecta</i> Mart. ex K.Schum.	Moura, E.O. 133	Severiano Melo	northeast
Malvaceae	<i>Ceiba glaziovii</i> (Kuntze) K.Schum.	Santos, L.A.S. 1229	Santa Cruz	northeast
Malvaceae	<i>Helicteres</i> cf. <i>guazumifolia</i> Kunth	Gonçalves, F.B. 382*	Macaíba	Neotropical
Malvaceae	<i>Herissantia crispa</i> (L.) Brizicky	Dantas, R. 39; Silva, A.F. 1002; Silva R.E. 10	Carnaubais; Santa Cruz; Porto do Mangue	American
Malvaceae	<i>Malachra fasciata</i> Jacq.	Dantas, A. 215	Tibau do Sul	American
Malvaceae	<i>Melochia tomentosa</i> L.	Santos, L.A.S. 1226	Santa Cruz	American
Malvaceae	<i>Pavonia cancellata</i> (L.) Cav.	Silva, A.F. 52; Gonçalves 394	Serra Negra do Norte; Lagoa Salgada	American
Malvaceae	** <i>Pseudobombax marginatum</i> (A.St.-Hil., Juss. & Cambess.) A. Robyns	Silva, A.F. 99*	Riacho de Santana	Neotropical
Malvaceae	<i>Sida acuta</i> Burm.f.	Gonçalves, F.B. 360	Goianinha	Pantropical
Malvaceae	<i>Sida ciliaris</i> L.	Gonçalves, F.B. 397; 490	Lagoa Salgada; Santa Maria	Pantropical
Malvaceae	<i>Sidastrum paniculatum</i> (L.) Fryxell	Santos, L.A.S. 1249; Dantas, A. 162	Santo Antônio; Tibau do Sul	Neotropical
Malvaceae	<i>Waltheria brachypetala</i> Turcz.	Dantas, A. 298	São Bento do Norte	northeast
Malvaceae	<i>Wissadula hernandioides</i> (U'Hér.) Garcke	Santos, L.A.S. 1240	São José do Campestre	American

Family	Species	Voucher	Municipality	Distribution
Melastomataceae	<i>Clidemia hirta</i> (L.) D. Don	Moura, E.O. 21	Macabá	Neotropical
Melastomataceae	<i>Tibouchina gardneri</i> (Naudin) Cogn.	Jardim, J.G. 6295	Serra de São Bento	northeast
Myrtaceae	<i>Eugenia astringens</i> Cambess.	Dantas, A. 110	Nísia Floresta	Brazilian
Nyctaginaceae	<i>Guaipira campestris</i> (Netto) Lundell	Roque, A.A. 1103	São José de Mipibu	Brazilian
Nyctaginaceae	<i>Guaipira</i> cf. <i>noxia</i> (Netto) Lundell	Silva, R.E. 05*; Gonçalves, F.B. 497*	João Câmara; Porto do Mangue	Brazilian
Phyllanthaceae	<i>Savia sessiliflora</i> (Sw.) Willd.	Gonçalves, F.B. 362*	Macabá	American
Poaceae	<i>Andropogon fastigiatus</i> Sw.	Oliveira, R.C. 1743	Canguaretama	Pantropical
Poaceae	<i>Aristida ekmanniana</i> Henrad	Flor. R. 268	Ceará-Mirim	Brazilian
Poaceae	<i>Aristida recurvata</i> Kunth	Oliveira, A.C.P. sn	Rio do fogo	American
Poaceae	<i>Cenchrus echinatus</i> L.	Dantas, A. 189	Baía Formosa	Pantropical
Poaceae	<i>Chrysopogon zizanioides</i> (L.) Robery	Cestaro, L.A. 97-0014	Natal	American
Poaceae	<i>Digitaria ciliaris</i> (Retz.) Koeler	Dantas, R.A. 70	Encanto	Pantropical
Poaceae	<i>Digitaria horizontalis</i> Willd.	Dantas, A. 190	Baía Formosa	American
Poaceae	<i>Gymnopogon fastigiatus</i> Nees	Souza, E.B. 3520; Silva, A.F. 57	Apodi; Serra Negra do Norte	American
Poaceae	<i>Hyparrhenia diplandra</i> (Hack.) Stapf	Jardim, J.G. 5788	Jucurutu	Pantropical
Poaceae	<i>Lasiacis divaricata</i> var. <i>austramericana</i> Davidse	Dantas, A. 116	Nísia Floresta	American
Poaceae	<i>Piretia leptophylla</i> Soderstr.	Jardim, J.G. 6251	Baía Formosa	American
Poaceae	<i>Schizachyrium condensatum</i> (Kunth) Nees	Dantas, A. 231; Flor, R. 266; 267	Ceará-Mirim; Tibau do Sul	American
Poaceae	<i>Setaria viridis</i> (L.) P.Beauv.	Santos, L.A.S. 1271	João Câmara	Pantropical
Poaceae	<i>Sorghum bicolor</i> var. <i>arundinaceum</i> (Desv.) de Wet & J.R.Harlan ex Davidse	Oliveira, R.C. 2236; 2164	Riacho de Santana; Luís Gomes	Pantropical
Peridaceae	** <i>Adiantum deflectens</i> Mart.	Moura, E.O. 146	São Francisco do Oeste	Neotropical
Rubiaceae	<i>Mitracarpus baturitensis</i> Sucre	Moura, E.O. 138; 212; 213	Carnaubais; Parelhas	Brazilian
Rubiaceae	<i>Mitracarpus longicalyx</i> E.B.Souza & M.F.Sales	Silva, A.F. 119A	Rio do fogo	northeast
Sapindaceae	<i>Allophylus quercifolius</i> (Mart.) Radlk.	Santos, L.A.S. 1232	São Tomé	northeast
Selaginellaceae	** <i>Selaginella convoluta</i> (Arn.) Spring	Silva, A.F. 68	Cruzeta	Neotropical

Table 2. Distribution patterns of species treated here as new botanical records for Rio Grande do Norte state, Brazil.

Distribution pattern	Number of species
Neotropical	21
American	19
Brazilian	14
Pantropical	10
Northeast Brazil	7

AM, BA, and MS states (Mitchell and Daly 2015). In RN, it is recorded from coastal regions with sandy soils.

ASTERACEAE

Stilpnopappus laiseae R.Barros & R.Esteves

This species is only known from PI state in Brazil occurring in Caatinga areas (Barros and Esteves 2004) and BA (Loeuille 2015). In RN, it inhabits anthropic areas with shallow grounds or stony soils.

BURSERACEAE

Protium heptaphyllum (Aubl.) Marchand

This species has a Neotropical distribution (Pirani 2003). In Brazil, it is widely distributed except in the south region (PR, RS, SC) and in a few states of Northeast (PI, PB, RN). It occurs in the Amazon Rainforest, Caatinga, Central Brazilian Savanna and Atlantic Rainforest (Daly 2015). In RN, it inhabits coastal areas with sandy soils.

CACTACEAE

Melocactus ernestii Vaupel

Endemic to Brazil, it is distributed in AL, BA, MG, PB, PE, and SE states and in Caatinga and Atlantic Rainforest (Zappi et al. 2015). In RN, it inhabits rock outcrops.

CELASTRACEAE

Maytenus acanthophylla Reissek

This species is endemic to Brazil, where it occurs in BA and MG states where it grows in Caatinga (Lombardi et al. 2016). In RN, it was collected in coastal seasonal forested areas.

CYPERACEAE

Becquerelia cymosa Brongn.

This species occurs from Nicaragua and Costa Rica in Central America, Trinidad and Tobago and the Guianas to Brazil in South America (Gómez-Laurito 2003). Previously unknown from DF, GO, MS, PI and RN states where it grows in Amazon Rainforest, Central Brazilian Savanna and Atlantic Rainforest (Alves et al. 2015). In RN, it was collected in seasonal forest in Atlantic Rainforest and Caatinga areas.

Eleocharis flavescens (Poir.) Urb.

This species is distributed in the United States, Mexico, Central America, Antilles, and South America (Trevisan and Boldrini 2008). In Brazil, it is widely distributed in the northeast (BA, CE, PB, PE), southeast (MG, RJ, SP) and south (PR, RS, SC) except in AL, SE, and ES states. It occurs in Caatinga and Atlantic Rainforest domains (Alves et al. 2015). In RN, it is found in wetland Atlantic Rainforest areas within restinga.

Eleocharis maculosa (Vahl) Roem. & Schult.

This species is widely distributed in the Americas from Central America to South America (Trevisan and Boldrini 2008). In Brazil, it is known for BA, CE, ES, MG, PA, PE, PR, RJ, RR, RS, SC, and SE states. It is widely distributed in all phytogeographical domains (Amazon Rainforest, Caatinga, Central Brazilian Savanna, Atlantic Rainforest, Pampa, Pantanal) (Alves et al. 2015). In RN, it occurs in restinga.

Eleocharis montana (Kunth) Roem. & Schult.

This species is distributed from United States to South America including Antilles (Trevisan and Boldrini 2008, Silveira and Longhi-Wagner 2008). It occurs in BA, DF,

ES, GO, MG, MS, MT, PE, PR, RS, SC, and SP states, where it grows in Caatinga, Central Brazilian Savanna, Atlantic Rainforest and Pampa (Alves et al. 2015). In RN, it occurs in seasonal wetland areas in Caatinga.

Rhynchospora caracasana (Kunth) Boeckeler

This species is distributed in Brazil, Bolivia, Suriname, Guyana and Venezuela (Strong 2006). In Brazil, occurs in BA, CE, DF, MG, and PE states and it is found in Caatinga and Central Brazilian Savanna (Alves et al. 2015). In RN, it was collected in Caatinga areas.

Rhynchospora gigantea Link

This species is distributed from Mexico, Central America to Brazil in South America (Guaglianone 2001). In Brazil, it occurs in AL, BA, ES, PB, PE, PR, RJ, RS, SC, SE, and SP states, where it grows in Amazon Rainforest, Caatinga, Central Brazilian Savanna and Atlantic Rainforest (Alves et al. 2015). In RN, it was collected in riparian forest.

Scleria macrophylla J.Presl & C.Presl

This species is distributed from Mexico to Brazil including Antilles (Gómez-Laurito 2003). In Brazil, it occurs in BA, DF, GO, MA, MG, MS, MT, PE, PI, RO, and TO states. It is found in Amazon Rainforest, Central Brazilian Savanna and Atlantic Rainforest (Alves et al. 2015). In RN, it was recorded in riparian forest.

EUPHORBIACEAE

Bernardia sidoides (Klotzsch) Müll. Arg.

This species is widely distributed from North America to Brazil (Govaerts et al. 2000). In Brazil, it occurs in BA, PE, and MT states, growing in Caatinga and Central Brazilian Savanna (Cordeiro and Secco 2015). In RN, it occurs in shrubby Caatinga with sandy soils and rocky outcrops.

Croton campestris A. St.-Hil.

This species occurs in Bolivia and Brazil (Forzza et al. 2010, Jørgensen et al. 2014). It has been previously recorded in AL, BA, CE, DF, ES, GO, MG, MS, PB, PE, PI,

PR, RJ, RS, and TO states, where it grows in Amazon Rainforest, Caatinga, Central Brazilian Savanna and Atlantic Rainforest (Cordeiro et al. 2015a). In RN, it occurs in Caatinga and secondary forest with clay soils.

Ditaxis malpighiacea (Ule) Pax. & K. Hoffm.

This species is endemic to Brazil and it is only recorded for AL, BA, PI, PE, and PB states, in Caatinga domain (Lucena and Alves 2009, Steinmann 2015). In RN, it was recorded in Caatinga areas.

Manihot esculenta Crantz

Native of South America and originated in the Amazon but widely distributed as a cultivated plant (Olsen and Schaal 1999). It has been previously recorded in Brazil for AC, AL, AM, AP, BA, CE, DF, GO, MA, MT, MG, PA, PE, PI, RO, and SP state, in Amazon Rainforest and Caatinga (Cordeiro et al. 2015b). In RN, it was collected in Caatinga and anthropic agricultural areas.

FABACEAE

Bauhinia dubia G. Don

Species endemic to Brazil where it is found in AM, CE, MA, PA, PI, and TO states, in Amazon Rainforest and Central Brazilian Savanna (Vaz and Tozzi 2003, Vaz 2015). In this study it was recorded in shrubby Caatinga.

Calliandra depauperata Benth.

Endemic to Brazil and previously recorded in BA, CE, PE, and PI states, in Caatinga (Souza 2015). In RN, it occurs in coastal areas with stony soils and shrubby caatinga.

Calliandra sessilis Benth.

This species occurs only in Brazil. It has been reviously recorded in BA, CE, MA, MT, MG, PA, PE, and PI states (Souza 2015). It occurs in Amazon Rainforest, Caatinga and Central Brazilian Savanna (Souza 2015). In RN, it was collected in transitional areas between Atlantic Rainforest and Caatinga, and between Atlantic Rainforest and restinga.

Inga vera Willd.

Inga vera is widely distributed from Mexico to Argentina (Zamora 2010). In Brazil, it is widely distributed so far not recorded only in Al and SE. It occurs in Amazon Rainforest, Central Brazilian Savanna, Atlantic Rainforest and Pantanal phytogeographical domains (Garcia and Fernandes 2015). In RN, it was collected sterile in Caatinga areas.

LECYTHIDACEAE

Eschweilera ovata (Cambress.) Mart. ex Miers

This species is known from Brazil in AL, AP, BA, ES, MA, MG, MT, PA, PR, PE, and SE state and Amazon and Atlantic Rainforest domains (Smith et al. 2015). In RN, it was recorded in coastal Atlantic forest areas.

MALVACEAE

Ayenia erecta Mart. ex K.Schum.

This species is endemic to Brazil and recorded only in PI state in Caatinga domain (Esteves 2015a). In RN, it was collected in Caatinga with sandy soils and also in anthropic areas.

Ceiba glaziovii (Kuntze) K.Schum.

This species is endemic to Brazil (Gibbs and Semir 2003). It is distributed in BA, CE, PB, and PE states in Caatinga, Central Brazilian Savanna and Atlantic Rainforest (Duarte 2015a). In RN, it was recorded in shrubby or forested Caatinga.

Helicteres guazumifolia Kunth

Helicteres guazumifolia occurs from Mexico to Brazil, in the states of BA, MT, PE, PI, RO, and SE (Cristóbal 2001, Esteves 2015b). In RN, it was collected in transitional areas of Caatinga and Atlantic Rainforest with stony soils.

Herissantia crispa (L.) Brizicky

This species is recorded from United State to Argentina (Alves et al. 2011). In Brazil, it occurs in AL, BA, PE, and SE states, in Caatinga and Central Brazilian Savanna (Bovini 2015a). In RN, it was collected in Caatinga with sandy soils.

Malachra fasciata Jacq.

This species occurs from Mexico to Bolivia in South America (Fryxell 2007). In Brazil, it has been recorded in BA, MA, MG, PE, and RJ states, in Amazon Rainforest, Central Brazilian Savanna and Atlantic Rainforest (Esteves 2015c). In RN, it was recorded in coastal areas.

Melochia tomentosa L.

This species is distributed from United States to Paraguay (Goldberg 1967, Rondón 2007). In Brazil, it occurs in AL, BA, CE, MT, MS, PB, PE, and PI states, in Caatinga, Central Brazilian Savanna and Atlantic Rainforest (Esteves 2015d). In RN, it was collected in shrubby and forested Caatingas.

Pavonia cancellata (L.) Cav.

It is distributed from Mexico to Brazil (Esteves and Krapovickas 2009). In Brazil, it occurs in AL, AM, BA, DF, CE, ES, GO, MA, MG, MT, MS, PA, PE, PI, PB, RJ, SE, and SP states, in Amazon Rainforest, Caatinga, Central Brazilian Savanna and Atlantic Rainforest (Esteves 2015e). In RN, it occurs in anthropic areas and stony soils.

Pseudobombax marginatum (A.St.-Hill.) A. Robyns

It occurs in South America (Amorim et al. 2009). In Brazil, it has been recorded in BA, CE, DF, ES, GO, MA, MG, MS, MT, PB, PR, RJ, RO, and SP states (Duarte 2015b). During the NFI we recorded it in Caatinga areas.

Sida acuta Burm. f

It has a Pantropical distribution (Krapovickas 2007). In Brazil, it occurs in BA, CE, GO, MA, MG, PE, PI, PA, and TO states in Amazon Rainforest, Caatinga, Central

Brazilian Savanna and Atlantic Rainforest (Bovini 2015b). In RN, it was recorded in Caatinga areas with shallow and stony soils.

Sida ciliaris L.

This species has a Pantropical distribution (Krapovickas 2007). It is a very polymorphic taxon with questionable delimitation (Fryxell 1985, Krapovickas 2007). In Brazil, it occurs in PE state (Amorim et al. 2009). In RN, it was collected in anthropic areas with clay soils and Caatinga with shallow soils.

Sidastrum paniculatum (L.) Fryxell

Widely distributed in the Neotropics (Alves et al. 2011), this species occurs in BA, MG, MT, MS, PB, PE, RJ, and SP states, in Amazon Rainforest and Caatinga (Bovini 2015c). In RN, it was collected in shrubby Caatinga and coastal areas.

Waltheria brachypetala Turcz.

This species is endemic to Brazil where it occurs in BA, CE, PE, and PI states in Caatinga (Esteves 2015f). In RN, it was recorded in coastal anthropic areas.

Wissadula hernandioides (L. Hér.) Garcke

Widely distributed from United States, Mexico, West Indies, Venezuela, Colombia, Bolivia, Paraguay, Argentina, and Brazil (Bovini and Baumgratz 2016). In Brazil, it occurs in BA, MT, MG, PA, PR, RJ, RR, RS, and SP states and in Amazon Rainforest, Central Brazilian Savanna, Atlantic Rainforest and Pantanal (Bovini 2015d). In RN, it was collected in Caatinga areas.

MELASTOMATACEAE

Clidemia hirta (L.) D. Don

This species is widely distributed from Mexico to Brazil (Goldenberg et al. 2005). Previously, it was recorded for all Brazilian states except to RN state. It grows in Amazon Rainforest, Caatinga, Cerrado, Atlantic Rainforest (Michelangeli and Reginato 2015). In RN, it grows in Caatinga areas.

Tibouchina gardneri (Naudin) Cogn.

This species is endemic to Brazil, where it occurs in CE and PE states and in Caatinga and Central Brazilian Savanna (Guimarães 2015). In RN, it was recorded in decidual seasonal forest.

MYRTACEAE

Eugenia astringens Cambess.

This species is endemic to Brazil, it occurs in BA, ES, PR, RJ, SC, and SP states in Atlantic forest (Sobral et al. 2015). In RN, it was recorded in coastal Atlantic forest areas.

NYCTAGINACEAE

Guapira campestris (Netto) Lundell

This species is known only from Brazil in BA, DF, GO, MG, and PI states, growing in Central Brazilian Savanna (Sá 2015). In RN, it occurs in semidecidual seasonal forest.

Guapira noxia (Netto) Lundell

Guapira noxia is endemic to Brazil, where it occurs in DF, GO, MG, MS, MT and SP state in Campo Rupestre and Central Brazilian Savanna (Sá 2015). In RN, it was collected sterile in shrubby Caatinga with stony soils.

PHYLLANTACEAE

Savia sessiliflora (Sw.) Willd.

This species occurs from Mexico, Cuba, Puerto Rico, Hispaniola, Caribbean, Venezuela and Brazil (Webster 1998). In Brazil, is recorded only for northeast (BA, CE, PE, SE) and reported only in Caatinga (Secco et al. 2015). In RN, it was collected in ecotonal areas between Atlantic forest and Caatinga.

POACEAE

Andropogon fastigiatus Sw.

This species occurs from Mexico, Central America, Antilles to South America and the Old World (Morales 2003). In Brazil, it is recorded in North, Northeast, Central West and Southeast regions in Amazon Rainforest, Caatinga and Central Brazilian Savanna (Zanin 2015a). In RN, it was recorded in anthropic areas with sandy soils.

Aristida ekmaniana Henrard

Species endemic to Brazil where it occurs in BA, DF, GO, MG, PR, and SP states in Central Brazilian Savanna (Longhi-Wagner 1990, 2015). In RN, it was recorded in coastal savanna areas.

Aristida recurvata Kunth

American species distributed from Belize, Venezuela, Guayanas, Bolivia to Brazil (Morales 2003). In this latter country, it is recorded for BA, DF, GO, MG, MS, MT, PR, RJ, RR, and SP states. It grows in Amazon Rainforest, Central Brazilian Savanna and Atlantic Rainforest (Longhi-Wagner 2015). In RN, it was recorded in Savanna areas the central portion the state.

Cenchrus echinatus L.

Pantropical species (Morales 2003). In Brazil, it occurs in BA, CE, DF, GO, MS, MT, PA, PB, PR, RO, RR, and SC state and in Amazon Rainforest, Caatinga, Central Brazilian Savanna, Atlantic Rainforest, Pantanal (Filgueiras 2015a). In RN, it was collected in coastal Atlantic forest areas.

Chrysopogon zizanioides (L.) Roberty

This species occurs from United States to Argentina (Filgueiras 2003a). In Brazil, it occurs in BA, RJ, and SP states and in Amazon Rainforest, Central Brazilian Savanna and Atlantic Rainforest (Filgueiras 2015b). In RN, it occurs in white-sand coastal areas.

Digitaria ciliaris (Retz.) Koeler

This species occurs from United States to Argentina and in the Old World (Morales 2003). In Brazil, it occurs in AM, BA, DF, ES, GO, MA, MG, MS, MT, PA, PE, PB, PR, RJ, RS, SC, SE, and SP states in Amazon Rainforest, Caatinga, Central Brazilian Savanna, Atlantic Rainforest, Pampa and Pantanal (Canto-Dorow 2015). In RN, it occurs in Caatinga areas.

Digitaria horizontalis Willd.

Distributed from United States, Central America to Argentina (Vega and Rúgolo de Agrasar 2003). In Brazil, it occurs in AC, AL, AM, AP, BA, CE, GO, MA, MS, PA, PE, PB, PR, RJ, SC, SP, and TO states in Amazon Rainforest, Caatinga, Central Brazilian Savanna, Atlantic Rainforest and Pantanal (Canto-Dorow 2015). In RN, it was recorded in coastal areas.

Gymnopogon fastigiatus Nees

This species occurs from Central America to Bolivia (Morales 2003). In Brazil, it occurs in AM, DF, GO, MG, MT, MS, RO, and SP states in Amazon Rainforest and Central Brazilian Savanna (Valls 2015). In RN, it occurs in anthropic areas with stony soils.

Hyparrhenia diplandra (Hack.) Stapf

This species is worldwide distributed. In Brazil, it is only known from PE state Atlantic Rainforest (Filgueiras et al. 2015a). In RN, it was recorded in Caatinga areas.

Lasiacis divaricata var. *austroamericana* Davidse

This variety occurs in South America from Ecuador to Argentina (Davidse 2003). In Brazil, it was recorded in BA, CE, ES, MA, MG, PR, SP, and SC states in Caatinga, Central Brazilian Savanna and Atlantic Rainforest domains (Filgueiras et al. 2015b). In RN, it was recorded in coastal areas.

Piresia leptophylla Soderstr.

This species is distributed from Colombia, Ecuador to Brazil (Judziewicz et al. 2000, Giraldo-Cañas 2011). In Brazil, it occurs in AM, BA, and PE states in the Amazon Rainforest and Atlantic Rainforest domains (Carvalho and Oliveira 2015). In RN, it occurs in white-sand restinga coastal areas.

Setaria viridis (L.) P.Beauv.

This species is widely distributed in the new and old world (Pensiero 2003, Morrone et al. 2014). In Brazil, it occurs in AP, DF, GO, MG, RS, and SP states in Amazon Rainforest, Central Brazilian Savanna and Atlantic Rainforest (Shirasuna and Rodrigues 2015). In RN, it was recorded in Caatinga areas.

Schizachyrium condensatum (Kunth) Nees

This species occurs from Mexico, Central American, Caribbean to Argentina in South America (Filgueiras 2003b). In Brazil, it was recorded in BA, DF, GO, MG, MS, MT, PR, RS, SC, and SP states in Central Brazilian Savanna, Atlantic Rainforest and Pampa domains (Zanin 2015b). In RN, it was found in coastal areas.

Sorghum bicolor var. *arundinaceum* (Desv.) de Wet & J.R. Harlan ex Davidse

This is a cultivated and naturalized species originally from Africa that now is worldwide distributed (Longhi-Wagner 2001, Giraldo-Cañas 2011). In Brazil, previously recorded only from Acre state (Filgueiras and Valls 2015). In RN, it was recorded in river banks with clay soil.

RUBIACEAE

Mitracarpus baturitensis Sucre

This species is endemic to Brazil (Souza et al. 2010). Recorded in BA, CE, DF, GO, MG, MT, PI, PB, and PE states in Central Brazilian Savanna and Caatinga phyto-geographic domains (Souza 2015). In RN, it was collected in shrubby Caatingas with stony soils or secondary forest.

Mitracarpus longicalyx E.B.Souza & M.F.Sales

This species is endemic to Brazil, where it occurs in BA, CE, PE, and PI states, restricted to Caatinga domain (Souza et al. 2010, Souza 2015). In RN, it was recorded in anthropic areas with banana plantation.

SAPINDACEAE

Allophylus quercifolius (Mart.) Radlk.

This species is restricted to northeast of Brazil. It occurs in AL, BA, CE, PE, and SE states in Caatinga and Central Brazilian Savanna phytogeographical domains (Somner et al. 2015). In RN, it was recorded in Caatinga areas.

Discussion

Systematic sampling in the entire state of RN during the NFI covered many municipalities that have seen little or no floristic attention in the past. Before the recent surveys, three municipalities – Natal, Mossoró and Serra Negra do Norte – had the highest botanical collecting effort for RN and 129 municipalities had less than 189 records (including 21 without any collection effort whatsoever) (Silva 2015). The low amount of previous effort is reflected in the high number of new occurrences to RN reported in the present study. The municipalities that were previously sampled were mostly concentrated along the Central Potiguar and Agreste Potiguar mesoregions of RN. This study reports new occurrences from the under-sampled Agreste mesoregion (e.g., Pureza), although it also reports new occurrences from some intensively-sampled regions, such as Mossoró, Serra Negra do Norte and Natal. This finding suggests that the botanical sampling in RN remains insufficient even in areas with the highest floristic efforts, such as the capital of the state and the second largest city, which also hosts an herbarium (Mossoró).

Most new occurrences belong to Poaceae, Fabaceae and Malvaceae. Fabaceae and Poaceae are the most species-rich families reported for the Caatinga (BFG 2015) and most new records are from this domain. Although the Atlantic Forest is the most species-rich ecoregion in Brazil (BFG 2015) the NFI conducted more sampling in the Caatinga than in the Atlantic Forest in RN. Sampling effort was allocated in this manner because Caatinga is geographically predominant in this state (Figure 1), while the Atlantic Forest covers only a narrow strip along the east coast, and its area has been severely reduced. Colombo et al. (2016) observed that increased efforts to sample the

flora of RN have resulted in improved knowledge of the flora of the Caatinga. Despite our focus on the Caatinga, we found that two of the three municipalities with the highest number of new occurrences, Ceará-Mirim and Canguaretama, were within the Atlantic Forest. This likely reflects the intrinsic diversity of the Atlantic Forest. We believe the Caatinga is the most under-studied area where future sampling efforts should be focused.

Our sampling efforts improved occurrence data of widely-distributed species in Brazil. Some species were originally absent only in RN state, such as *Clidemia hirta* (Melastomataceae), while others were unknown in several other northeastern Brazilian states, such as *Inga vera* (Fabaceae) (also unknown in Alagoas and Sergipe) and *Amburana cearensis* (Fabaceae), which is categorized as endangered (Americas Regional Workshop 1998). *Amburana cearensis* was recently recorded for RN (Amorim et al. 2016) and currently is only absent in Sergipe. We also improved occurrence data of species with restricted distributions, such as *Stilpnopappus laiseae* (Asteraceae) and *Ayenia erecta* (Malvaceae), which were previously reported only from Piauí, and *Sorghum bicolor* var. *arundinaceum* (Poaceae), which was previously known only from Acre. We added occurrence data for a few other species already registered for the RN flora but with few known localities. This is the case of the Cactaceae *Tacinga subcylindrica* and *Brasiliopuntia brasiliensis*, previously known from only one collection in the Açu municipality in 2013. New collections from 2015 onwards considerably expanded the documented occurrence of *T. subcylindrica* in RN (to the municipalities of Equador, João Câmara, Areia Branca and Macau). Only one collection of *B. brasiliensis* was reported in 2001 in the municipality of Macaíba, however, new collections were made from 2011 onwards in the municipality of Ceará-Mirim.

Only seven out of 71 newly-occurring species (~10%) have a distribution that is restricted to northeastern Brazil. These low numbers of endemic species are a general pattern for northeast Brazil flora compared to other regions. According to BFG (2015), only 22.7% of 10,661 species of the northeast Brazil are endemic. Also, most of the area in the northeast is Caatinga, which shows a lower percentage of endemic taxa (19.7% of 4,657, BFG 2015), despite being a unique biome in Brazil.

The results of this NFI indicate that the number of species for RN is still underestimated. However, the knowledge of RN's flora is growing rapidly. New species have recently been discovered (Sobral 2010, Araújo and Alves 2013, Lourenço et al. 2013, Terra-Araújo et al. 2013, Ribeiro et al. 2015b, Souza et al. 2016) and new occurrences for genera or species have been reported for RN (Versieux et al. 2013a, 2013b, São-Matheus et al. 2013, Magalhães et al. 2014, Amorim et al. 2016, Colombo et al. 2016, Gomes-Costa and Alves 2016). In five years, the number of species records for RN increased by 78% (BFG 2015). To continue this trend, we recommend intensive effort focusing on areas that have not been explored by modern taxonomists (Silva 2015), continuous collection across seasons, investment in training of local taxonomists, and improvements to the infrastructure of herbaria.

We emphasize the importance of collaboration among institutions to improve herbarium collections. Most Brazilian herbaria can be considered small, having less than

20,000 records (Vieira 2015). Currently Refflora hosts 54 collections, out of which 40 can be considered small. In our view, small collections are more prone to difficulties due to limited number of staff and funding, lack of visits from specialists (influencing the quality of the data), fewer type specimens, and curators with an overload of tasks (most of them professors). Lack of recognition within institutions, when an herbarium is regarded as belonging to a lab or to a particular professor, leave many collections vulnerable to loss or damage. By facilitating visits by specialists to study specimens, and funding technical fellowships, we improved the quality of our data and incorporated a valuable collection that otherwise would have been abandoned. The impact of visits from specialists on our floristic list is demonstrated by the number of new records reported for Poaceae and Cyperaceae and also by the merging of Museu Câmara Cascudo collection to UFRN herbarium, revealing new occurrences from specimens collected decades ago. The Museu Câmara Cascudo collection for algae is still awaiting examination by specialists, which will likely lead to more new botanical occurrences to RN.

Conclusion

The partnership between NFI, Refflora, SiBBR and UFRN Herbarium has advanced knowledge of biodiversity by exploring areas with few botanical records and adding new species records for the Flora of Rio Grande do Norte. The geographical distribution of newly-added species is mostly Neotropical (21 spp.), while fewer (seven spp.) are endemic to northeastern Brazil. From these results, we conclude that species that have not been recorded to date may occur in different habitats, and that the entire state requires additional floristic inventories. Furthermore, we revealed areas that were poorly covered by existing botanical collections. We also recorded new species from areas with relatively high previous effort of collection, indicating that the species richness in RN remains underestimated. Even our collections may contain specimens that should be further analyzed and studied by specialists, who rarely have opportunities to visit small herbaria. Future botanical projects should fill these remaining gaps in collections, particularly focusing on seasonality. Finally, the NFI/Refflora/SiBBR projects in RN improved the UFRN herbarium collection by digitizing all specimens and improved the curation of the collection through exchange of material among institutions, increased visibility of our specimens online, and attention from specialists.

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Revisions of *Ruizodendron* and *Pseudephedranthus* (Annonaceae) including a new species and an overview of most up-to-date revisions of Neotropical Annonaceae genera

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Abstract

We present revisions of the Neotropical genera *Ruizodendron* and *Pseudephedranthus* (Annonaceae). *Ruizodendron* includes a single species *R. ovale*. *Pseudephedranthus* now comprises two species, with the description of the new species *P. enigmaticus* **sp. nov.** extending the range of the genus beyond the Upper Rio Negro region of Brazil (Amazonas) and adjacent Venezuela (*P. fragrans*), to include Guyana, Suriname, and the Brazilian state of Pará. An overview is provided of current revisions of Neotropical Annonaceae genera that will aid in accessing proper species information for this frequently encountered tropical rain forest family.

Keywords

Biodiversity, Neotropics, new species, systematics, taxonomy

Introduction

Annonaceae are frequent components of tropical rain forests worldwide (Gentry 1993; Tchouto et al. 2006; Punyasena et al. 2008; Sonké and Couvreur 2014), for which it is vital to have a good overview of the most recent knowledge of the ca. 2450 spe-

cies (Rainer and Chatrou 2006). Revisions and monographs are an important tool to document this knowledge, be it online or in a more traditional printed format. This documentation is not a trivial exercise. For instance, Ter Steege et al. (2016) wanted to estimate how many tree species occur in the Amazon basin. They used, amongst many other sources, the most recent revision of the Annonaceae genus *Guatteria* (Maas et al. 2015) to check their preliminary taxa list. Since Maas et al. (2015) reduced 62 Amazonian tree species into synonymy and described 10 new Amazonian species the check of Ter Steege et al. reduced the taxonomic error for this group substantially.

With respect to Neotropical Annonaceae, renewed revisionary efforts have been underway since the early eighties (Maas 1984; Chatrou 1999; Oliveira and Sales 1999). To date, 29 out of 34 Neotropical genera (Maas et al. 2011) have been revised, of which 11 published after 2000 (Table 1). Revisionary work on three further genera (*Desmopsis* Saff., *Ephedranthus* S.Moore, and *Sapranthus* Seem.) is at an advanced stage, and the final two, *Annona* L. (including *Rollinia* A.St.-Hil.) and *Xylopia* L., are subject of ongoing research. The *Duckeanthus* R.E.Fr. revision (Fries 1934) is the oldest still in use and together with a more recent addition by Maas et al. (1993) provides an adequate summary of all knowledge available on this monospecific genus. However, not all recently revised genera might be monophyletic (e.g. *Desmopsis*, *Klarobelia* Chatrou, *Oxandra* A.Rich. and *Stenanona* Standl.) and some of the taxonomic treatments will need to be modified in the light of future molecular phylogenetic evidence.

Two genera so far still awaiting revision were the monospecific genus *Ruizodendron* R.E.Fr. and the non-related genus *Pseudephedranthus* Aristeg.

Taxonomic history of *Ruizodendron*

The monospecific genus *Ruizodendron* occurs from Amazonian Brazil to Bolivia, Peru, Ecuador, and Colombia (Fig. 1). It is readily recognized by the asymmetrical transversely ellipsoid monocarps (Fig. 2b, Fig. 3d, e, f). The genus name consists of the Greek word for tree (*dendron*) and part of the name of the Spanish botanist Hipolito Ruiz López (1754–1815). Ruiz undertook a field trip throughout South America from 1779 to 1788 together with J. Pavón after which they published *Florae Peruvianae et Chilensis Prodrum* (1794, reprinted 1797) and *Flora Peruviana et Chilensis* (1798).

Ruizodendron was not originally described as a separate genus. During the field expedition of Ruiz and Pavón in South America in 1778–1788 they collected a specimen near Pozuzo in Peru that they described as *Guatteria ovalis* Ruiz & Pav. (Fries 1936; Maas et al. 2011; Maas et al. 2015). The specimen bore solely fruits and the description was therefore incomplete: “G. foliis oblongis ovalibusque. Arbor quadriorgyalis.” The epithet “ovalis” was appropriate for their material since the type has oval, on both sides rounded, leaves (Fig. 3a) of ca. 10 cm long (Fries 1939, p. 544; although we now know that leaves can be found that are less typically oval). Fries studied this type specimen (B) and found that vegetative parts and structure of the monocarps differed from that typical of *Guatteria*, concluding that this specimen should not be attributed to that

Table 1. 34 Neotropical genera of Annonaceae (based on Maas et al. 2011) and their most up-to-date revision. A more elaborate overview of taxonomic literature of these taxa can be found in Erkens et al. (2012).

Genus:	Most up-to-date revision:
<i>Anaxagorea</i> A.St.-Hil.	Maas and Westra 1984, 1985 with a new species in Maas et al. 1986
<i>Annona</i> L. (including <i>Rollinia</i> A.St.-Hil.)	Ongoing work (for inclusion of <i>Rollinia</i> see Rainer 2007)
<i>Asimina</i> Adanson (including <i>Deeringothamnus</i> Small)	Kral 1960 (for inclusion of <i>Deeringothamnus</i> see Maas et al. 2011)
<i>Bocagea</i> A.St.-Hil.	Johnson and Murray 1995
<i>Bocageopsis</i> R.E.Fr.	Maas et al. 2007
<i>Cardiopetalum</i> Schtdl.	Johnson and Murray 1995
<i>Crematosperma</i> R.E.Fr.	Pirie et al. 2005
<i>Cymbopetalum</i> Benth.	Murray 1993
<i>Desmopsis</i> Saff.	Schatz et al. in prep. (pers. comm.)
<i>Diclinanona</i> Diels	Erkens et al. 2014
<i>Duckeanthus</i> R.E.Fr.	Fries 1934; Maas et al. 1993
<i>Duguetia</i> A.St.-Hil.	Maas et al. 2003, including all African species with a new species in Maas and Westra 2010, and a new species in Bazante and Alves 2017
<i>Ephedranthus</i> S.Moore	Carvalho Lopes in prep. (pers. comm.)
<i>Froesiodendron</i> R.E.Fr.	Johnson and Murray 1995
<i>Fusaea</i> (Baill.) Saff.	Chatrou and He 1999
<i>Guatteria</i> Ruiz & Pav.	Maas et al. 2015
<i>Hornschuchia</i> Nees	Johnson and Murray 1995
<i>Klarobelia</i> Chatrou	Chatrou 1998
<i>Malmea</i> R.E.Fr.	Chatrou 1998
<i>Mosannona</i> Chatrou	Chatrou 1998
<i>Onychopetalum</i> R.E.Fr.	Maas et al. 2007
<i>Oxandra</i> A.Rich.	Junikka et al. 2016
<i>Porcelia</i> Ruiz & Pav.	Murray 1993
<i>Pseudephedranthus</i> Aristeg.	Oliveira and Sales 1999 and this work
<i>Pseudomalmea</i> Chatrou	Chatrou 1998
<i>Pseudoxandra</i> R.E.Fr.	Maas and Westra 2003 with a new species in Maas and Westra 2005
<i>Ruizodendron</i> R.E.Fr.	This work
<i>Sapranthus</i> Seem.	Schatz et al. in prep. (pers. comm.)
<i>Stenanona</i> Standl.	Schatz and Maas 2010 with a new species in Ortiz-Rodriguez et al. 2014
<i>Tetrameranthus</i> R.E.Fr.	Westra and Maas 2012
<i>Tridimeris</i> Baill.	Schatz 1987 with a new species in Ortiz-Rodriguez et al. 2016
<i>Trigynaea</i> Schtdl.	Johnson and Murray 1995 with a new species in Lobão 2017
<i>Unonopsis</i> R.E.Fr.	Maas et al. 2007
<i>Xylopia</i> L.	Ongoing work

genus. On the basis of the axillary flower buds with imbricate petals, and stamen and carpel morphology he concluded that this species should be placed close to *Crematosperma* R.E.Fr. (Fries 1936, 1939).

In 1925 the same species was collected by Steinbach in Santa Cruz in Bolivia, but also this specimen solely bore fruits. Only with the finding by Klug of a flowering specimen in the Upper (“oberen”) Río Huallaga region (San Martín; not far from the type locality of *Guatteria ovalis*) the placement of the genus within Annonaceae could

be studied for the first time. From the flower it was clear that this indeed was not *Guatteria*. Fries (1936) even thought that the flowers were quite aberrant for Neotropical Annonaceae: they had very thin, long and narrow petals (“linear lanzettlich”; Fig. 2a, Fig. 3b). Based on the position of the articulation on the pedicel and the position of the bracts Fries (1939) stated that *Ruizodendron* was related to *Crematosperma*. However, the fruiting pedicel was placed in the middle of the long side of the monocarp (“...indem der Same bei *Guatteria ovalis* quer gestellt und in horizontaler Fläche ausgestreckt zu sitzen kommt mit dem Stiel des Monokarpiums mitten an der Längseite befestigt”; Fig. 2b, 3d, f). Based on this fruit type Fries concluded that *Ruizodendron* was unique among American Annonaceae (“Wir erhalten dadurch ein Fruchttyp, der allein dasteht unter den amerikanischen Annonaceen”). Furthermore, based on dried material he judged that the specimens had fleshy fruit *in vivo*. This was not similar to his dried material of *Crematosperma* species, which had a dry, fragile pericarp. All this led Fries to describe *Ruizodendron ovale* (Ruiz & Pav.) R.E.Fr.

More recently, Van Setten and Koek-Noorman (1992) used fruit and seed features to place *Ruizodendron* as part of a group with six other Neotropical genera (be it together with other non-Neotropical genera): *Crematosperma*, *Ephedranthus*, *Malmea* R.E.Fr., *Oxandra*, *Pseudephedranthus* and *Pseudoxandra* R.E.Fr. This is in line with an earlier grouping of Walker (1971) based on pollen characters and a concurrent conclusion by Van Heusden (1992) based on flower morphology. One year later, Keßler (1993) included *Ruizodendron* with *Oxandra*, *Pseudoxandra*, *Crematosperma*, and *Ephedranthus* in his “*Oxandra* group”, characterised by axillary flowers, imbricate sepals and petals, sulcate pollen grains, a single basal or lateral ovule, and free monocarps.

At this moment, based on molecular phylogenetic work (Chatrou et al. 2012), *Ruizodendron* is placed in a well-supported clade within tribe Malmeeae Chatrou & R.M.K.Saunders (subfamily Malmeeae Chatrou, Pirie, Erkens & Couvreur) with *Ephedranthus*, *Klarobelia*, *Mosannonna*, *Oxandra*, *Pseudephedranthus*, and *Pseudomalmea* Chatrou, although its exact phylogenetic placement within this clade is not yet known.

Taxonomic history of *Pseudephedranthus*

Until now, *Pseudephedranthus* consisted of a single species: *P. fragrans* (R.E.Fr.) Aristeg. known only from Brazil in the state of Amazonas (Upper Rio Negro Region, Rio Cauaburi), and adjacent Venezuela (Piedra de Cucuy; Fig. 1). In 1993, Maas collected a specimen (Maas et al. 6878, Fig. 6, 7) of *P. fragrans* during a botanical expedition in the Upper Rio Negro Region of Brazil and adjacent Venezuela and reported that the species was fairly common in the forested hills at the base of Piedra de Cucuy. Newly described here, *P. enigmaticus* Maas & Westra is distributed across Guyana, Suriname, and the Brazilian state of Pará (Fig. 1).

Pseudephedranthus (Aristeguieta 1969) can be easily recognised vegetatively. Its leaves exhibit some quite distinctive features (fig 4a): 1) the lamina is shiny on the upper side; 2) the primary vein is both raised and ribbed at the upper side of the lamina; 3) relatively few, widely spaced secondary veins (5–9) are present, and 4) the upper side

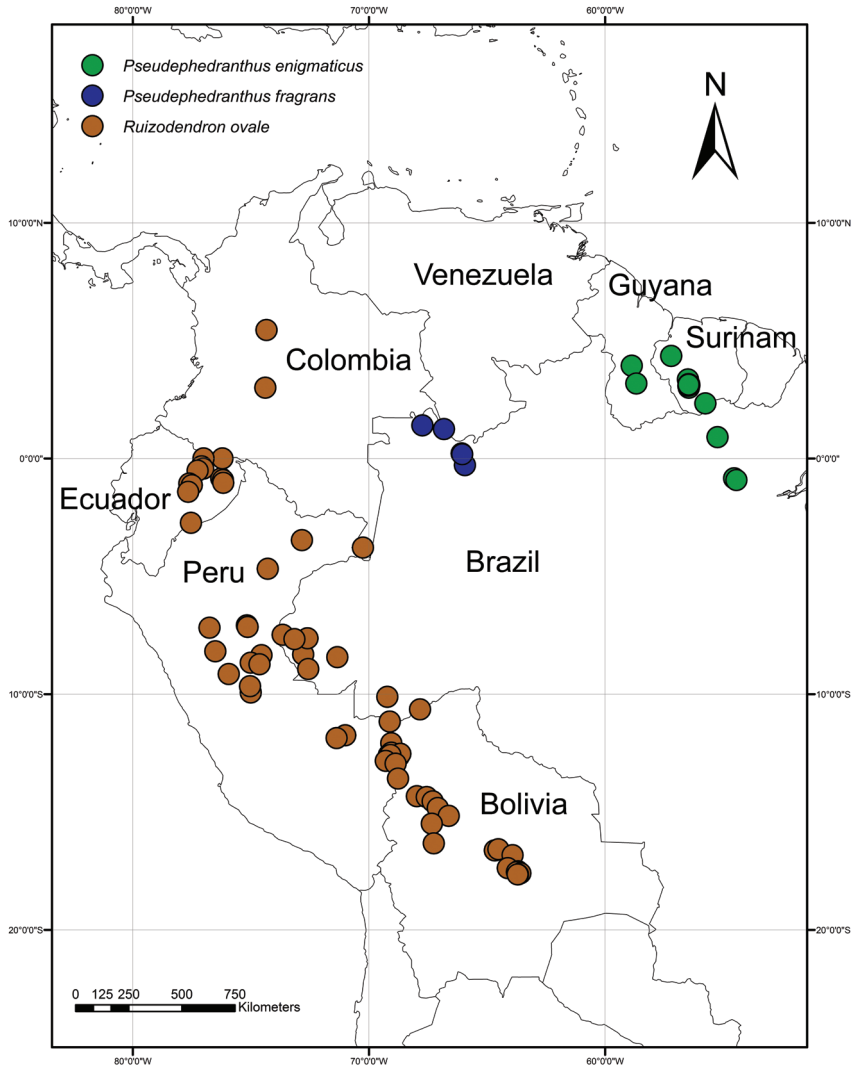


Figure 1. Distribution map of *Ruizodendron ovale*, *Pseudephedranthus fragrans* and *P. enigmaticus*.

shows a reticulate venation. The latest revision dated from 1999 (Oliveira and Sales 1999), written in Portuguese.

Maas et al. (1993) hypothesised that *Pseudephedranthus* is closest to *Oxandra* by its elongate connective appendages, and to *Ephedranthus* by its androdioecious flowers and its extremely large seeds (25–30 × 13 mm, Fig. 5b).

Pirie et al. (2006) already showed that *Pseudephedranthus* is nested within *Klarobelia*. More recent molecular phylogenetic work (Chatrou et al. 2012) confirmed this and demonstrated as well that *Pseudephedranthus* is part of the same clade as *Ruizodendron* together with *Ephedranthus*, *Klarobelia*, *Mosannonna*, *Oxandra*, and *Pseudomalmea* (in tribe Malmeeae Chatrou & R.M.K.Saunders).

Materials and methods

Herbarium material was investigated from the following herbaria: A, AAU, CGE, E, F, G, GB, GH, INPA, K, L, LPB, MICH, MO, P, NY, RB, S, U, UC, US, WAG, and WIS. Dried herbarium material was used for measurements, colour indications and descriptions of surface structures. If measurements were done on material kept in spirit this is indicated by curly brackets { }. We have indicated the density of hair cover according to the following gradations: densely, rather densely and sparsely.

Taxonomic treatment

Ruizodendron R.E.Fr.

Ruizodendron R.E.Fr., *Arkiv för Botanik Ser.* 28B (4): 3. 1936.

Type. *Ruizodendron ovale* (Ruiz & Pav.) R.E.Fr. (= *Guatteria ovalis* Ruiz & Pav.)

Description. *Trees*, often with buttresses; young twigs lenticellate. *Leaves*: distichous, simple, entire, shortly petiolate, exstipulate; lamina medium-sized, mostly elliptic, venation brochidodromous to eucamptodromous, articulated at the base, primary vein slightly raised to flat above, glaucous below. *Flowers* actinomorphic, bisexual, perianth consisting of one whorl of sepals and two whorls of petals; pedicels with basal articulation and 2 soon falling bracts; sepals 3, imbricate, free, much shorter than the petals, soon falling; petals 6, imbricate, free, subequal; stamens ca. 40, extrorse, filament very short, connective apex discoid, glabrous; carpels ca. 25, spirally arranged, free, ovary 1-locular with 1 ovule attached below the middle, style absent, stigma ovoid, hairy. *Fruit* apocarpous, composed of few indehiscent monocarps, these transversely ellipsoid, the stipe laterally attached. *Seed* 1, filling the whole monocarp, orange-brown, rumination lamellate in 2 parts, raphe a fine, shallow groove.

Distribution. Monotypic, occurring in the western part of South America (Fig. 1).

Ruizodendron ovale (Ruiz & Pav.) R.E.Fr.

Figs 2, 3

Ruizodendron ovale (Ruiz & Pav.) R.E.Fr., *Arkiv för Botanik Ser.* 28B (4): 3. 1936. – *Guatteria ovalis* Ruiz & Pav., *Systema vegetabilium florum peruvianae et chilensis* 1: 146. 1798.

Type. PERU. Huánuco: Pozuzo, anno 1782, *Ruiz & Pavón s.n.* (holotype MA [barcode MA811639], only photograph seen; isotype B).

Description. *Tree*, 5–45 m tall, 10–70 cm diam., buttresses to ca. 1.5 m high, young twigs longitudinally fissured, sparsely covered with appressed, silvery hairs

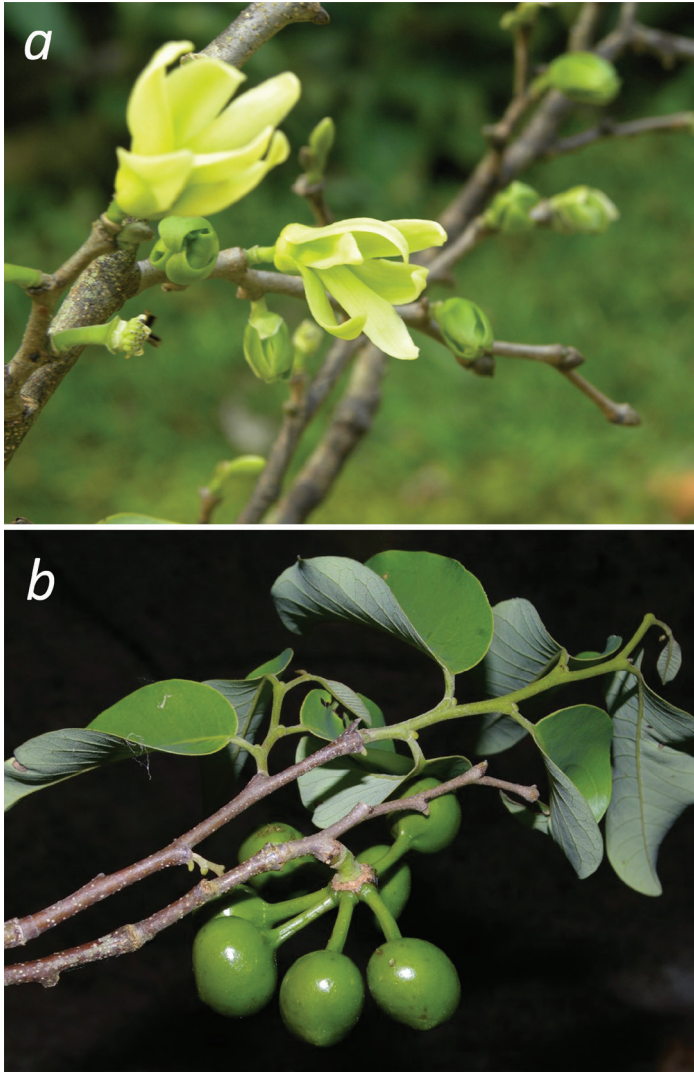


Figure 2. a Flowering branch of *Ruizodendron ovale* **b** The asymmetrical, transversely-ellipsoid mono-carps of *Ruizodendron ovale* (van der Werff 22749, MO). Missouri Botanical Garden, used with permission. Photograph 1b downloaded from <http://www.tropicos.org>. Photograph 1a taken by Abel Monteagudo, 1b taken by Rodolfo Vásquez.

0.1–0.4 mm long, soon glabrous. *Leaves*: petioles 5–10(–15) by 1–2 mm; lamina elliptic to ovate, rarely narrowly so, 6–25 by 4–12 cm (index 1.5–2(–3)), chartaceous, olive green to greenish brown and shiny above *in sicco*, glaucous to pale brown below, glabrous above, sparsely covered with appressed, silvery hairs to ca. 0.4 mm long mainly on larger veins below, base rounded with the extreme base slightly attenuate; apex obtuse, rarely acute; secondary veins 7–14 on either side of primary vein, slightly raised to flat above, shortest distance between secondary veins

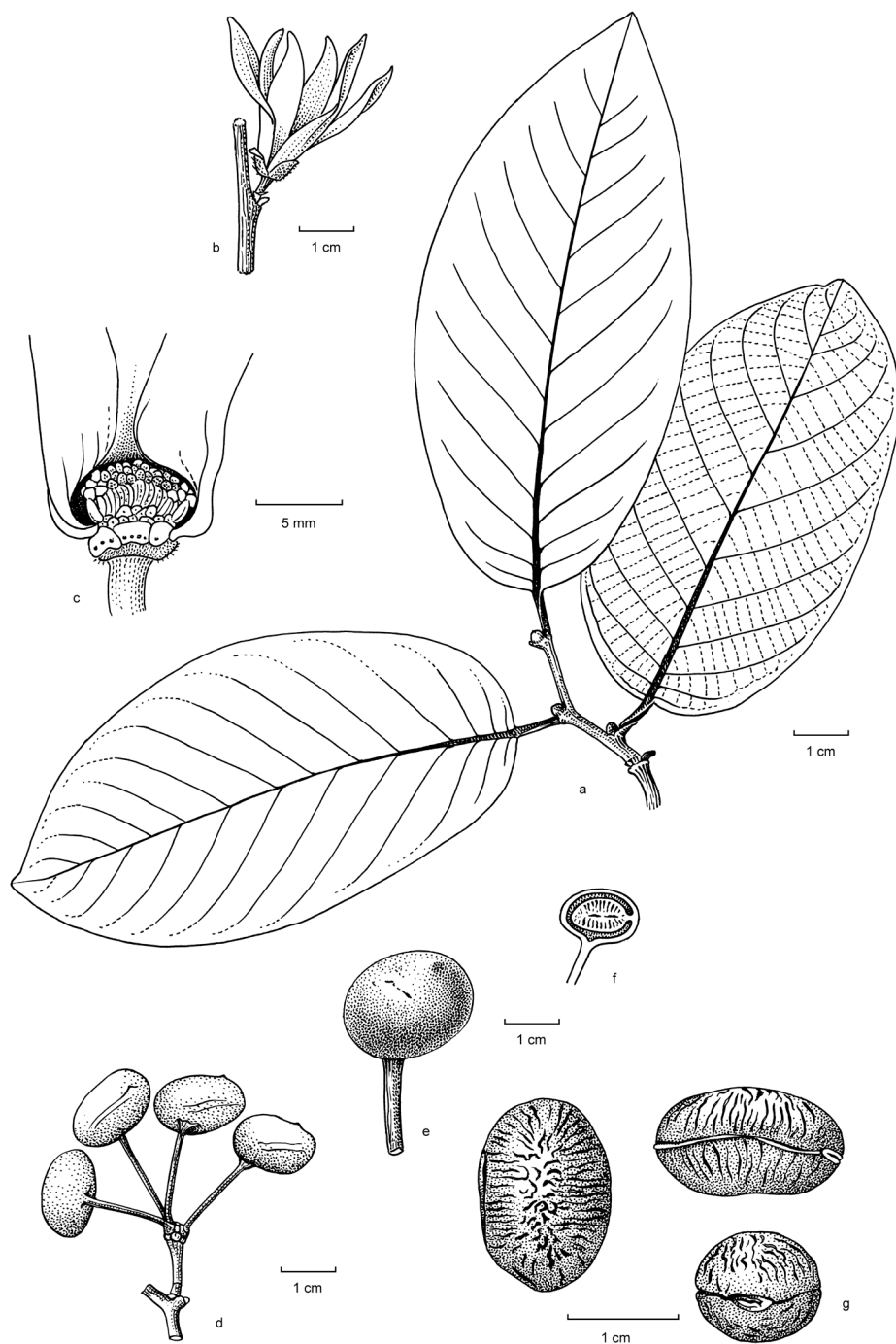


Figure 3. Drawing of *Ruizodendron ovale*. **a** Top of a branch **b** Flower **c** Base of flower with part of petals removed to show the interior **d** Fruit **e** Monocarp **f** Monocarp in longitudinal section showing laterally attached seed **g** Seed viewed clockwise starting from right: longitudinal view, hilum side, and in longitudinal section. Drawings by H. Rypkema.

and margin 1–3 mm, tertiary veins strongly raised above, percurrent to reticulate. *Inflorescences* axillary, 1-flowered, rarely 2 together through auxiliary bud formation, often appearing on small, leafless lateral branches. Indument: pedicels, outer side of bracts and sepals densely covered with appressed, silvery hairs to ca. 0.4 mm long, petals sparsely so; pedicels 4–8 by 1 mm, fruiting pedicels to ca. 10 by 1–3 mm; bracts 2, soon falling, one bract placed just below the articulation, ovate, ca. 4 mm long, other bract placed at ca. 0.2 from the base of the pedicel, ovate-triangular, ca. 4 mm long; sepals ovate-triangular, 5–6 by 3–4 mm, soon falling; petals white to cream *in vivo*, black *in sicco*, narrowly elliptic, 20–50 by 3–5 mm, membranous, base widened enclosing the pollination chamber; torus slightly raised; stamens ca. 1.5 mm long; carpels ca. 1 mm long. *Monocarps* 1–10, green, maturing black *in vivo*, brownish black *in sicco*, transversely ellipsoid, 15–20{–25} by 10–16 mm, glabrous, apex excentrically apiculate (apiculum to ca. 1 mm long), wall 0.2–0.5 mm thick; stipes 8–25 by 1–1.5 mm, excentrically placed. *Seed* transversely ellipsoid, 14–22 by 9–13 mm, orange-brown.

Distribution. Colombia (Amazonas, Cundinamarca, Meta), Ecuador (Morona-Santiago, Napo, Orellana, Pastaza, Sucumbios), Peru (Huánuco, Loreto, Madre de Dios, Pasco, San Martín, Ucayali), Bolivia (Beni, Cochabamba (?), La Paz, Pando, Santa Cruz), Brazil (Acre). Fig. 1.

Habitat and ecology. In tropical rain forest, often on alluvial soil. At elevations of up to 800 m. Flowering: July to September; fruiting: August to May.

Vernacular names. Bolivia: Blanquillo (*Vargas C. et al.* 1133), Fariña seca de bajo (Dominguez & Gonzales 42), Ojojo blanco (*DeWalt et al.* 283, Neill & Quevedo 9386, Seidel *et al.* 5640), Ojoso (*Vargas C. et al.* 5366), Ojoso blanco (*Del Aguila et al.* 95), Ojoso negro (*Cruz & Terceros* 208, *Meneces & Terceros* 473, *D.N. Smith et al.* 14160, 14194A, 14215, 14346), Palo fino (*Cruz & Medina* 153), Periquina hoja menuda (*Meneces & Hartshorn* 777); Tumuqui (Tacana name) (*DeWalt et al.* 283). Brazil: Envira orelha de onça (*Silveira et al.* 1094), Piaçaba (*Campbell et al.* 9473a). Ecuador: Aparina cara caspi (Quichua name) (*Zuleta* 364), Shampipian (*Ortega U.* 233). Peru: Ochabaja (*Begazo* 61), Panjil ruro (*Albán C.* 5878, *Schunke V.* 1203), Paujil ruro (*Tello* 257), Paujil ruro amarillo (*Kröll* 217).

Specimens examined. **Bolivia.** Beni: Prov. Moxos, 130 km S of San Ignacio, *Del Aguila et al.* 95 (U), 27 km from San Borja, along road to Trinidad, 260 m, *D.N. Smith et al.* 14160 (U), 14194A (U), 14215 (MO); Prov. Ballivian, 35 km on the road from Yucumo to Rurrenabaque, 230 m, *D.N. Smith et al.* 14346 (MO). La Paz: Prov. Iturralde, Comunidad de Santa Fé, 250 m, *DeWalt et al.* 510 (U), Upper Madidi ridge top, 7 km NE of camp, 370–380 m, *Gentry et al.* 70691 (MO); Prov. Iturralde, Ixiamas, 90 m, *Couvreux & Vargas* 213 (MO, RB); Prov. Sur Yungas, Basin of Río Bopi, Asunta, near Evenay, 690–750 m, *Krukoff* 10669 (A, F, G, K, MICH, MO, NY, S, U, UC, US); Prov. Sur Yungas, Upper Río Beni, concession of cooperativa Sapecho, 600 m, *Seidel et al.* 5640 (U). Pando: Prov. Manuripi, Batraja, 35 km E of Puerto Rico, 150 m, *Chatrou et al.* 453, 457 (U). Santa Cruz: San Rafael de Amboró, Centro de Estudios Ambientales, 400 m, *Maas et al.* 8762 (U); Prov. Ichilo,

Parque Nacional Amboró, Campamento Molle, San Germán, road from Yapacaní to Río Ichilo, 400 m, *Maas et al.* 8773 (U); Prov. Ichilo, Reserve Forestal 'El Choré', 240 m, *Meneces et al.* 473, 476 (U); Prov. Ichilo, Parque Nacional Amboró, 23 km S of Buena Vista, along Río Chonta, 420 m, *Nee* 36863 (LPB, MO, NY, U); Prov. Ichilo, Parque Nacional Amboró, 4 km SW of Buena Vista, S side of Río Surutu, 315 m, *Nee* 39391 (MO, NY, U, WIS); Prov. Ichilo, Reserva Forestal Choré, Río Ibabo, bosque experimental "Elias Meneces", 180 m, *Neill et al.* 9386 (G, MO); Prov. Sara, 400 m, *Steinbach* 7248 (A, E, F, G, K, LPB, MO, NY, U); Prov. Ichilo, Parque Nacional Amboró, Río Yapoje and Saguayo, 8 km SW of El Carmén, 360 m, *Vargas C. et al.* 1129 (MO), 1133 (NY, MO); Prov. Ichilo, El Carmén, 8 km SSW of Buena Vista, on the way to Huayti, 400 m, *Vargas C. & Méndez F.* 5366 (U). **Brazil.** Acre: Upper Rio Moa near Fazenda Arizona, *Campbell et al.* 6196 (U); Fazenda Bom Sossego, between Igarapé Cujubim and Igarapé Jacamin, *Campbell et al.* 8633, 9473a (U); Mun. Cruzeiro do Sul, Igarapé Humaitá, aflente do Rio Juruá, Colocação Várzea Grande, *Cid et al.* 10453 (U); Mun. Cruzeiro do Sul, Rio Juruá, entrada do Igarapé Viseu, *Cid et al.* 10535 (U); Mun. Cruzeiro do Sul, Upper Rio Juruá, Fazenda Calila Sara, *Cid et al.* 10839 (MO, U), Mun. Sena Madureira, Basin of Rio Purus, Rio Iaco, right bank, Nova Olinda, between Igarapé Santo Antonio and Igarapé Boa Esperança, *Daly et al.* 7909 (U); Mun. Marechal Thaumaturgo, Basin of Rio Juruá, Rio Bagé, *Daly et al.* 10245 (U); Mun. Manoel Urbano, Rio Chandles (tributary of Rio Purus), downstream from Igarapé Canamarí, *Daly et al.* 11234 (U); Colonia Cinco Mil ("Seita do Cipó"), *Nelson* 818 (MICH, MO, NY, RB, U, US); Mun. Xapuri, Reserva Extrativista Chico Mendes, Serigal Floresta, Colocação Rio Branco, *Figueiredo* 270 (U); Tarauacá, Bacia do Alto Juruá, Rio Tarauacá, Reserva Indígena Praia do Carapanã, Colocação Nova Morada. *Silveira et al.* 1094 (U). **Colombia.** Linden s.n. (CGE); Cundinamarca: Mun. Yacopí, vereda El Morro, 405 m, *Rangel et al.* 13563 (U). Meta: Parque Nacional Natural Tinigua, Serranía Chamusa, Centro de Investigaciones Primatólogicas La Macarena, *Stevenson* 299 (COL). **Ecuador.** Napo: Parque Nacional Yasuní, Pozo petrolero Daimi, 200 m, *Cerón et al.* 4227 (MO); Cantón Tena, Estación Biológica Jatun Sacha, 8 km E of Misahuallí, 400 m, *Cerón et al.* 5976 (G, GB, MO, U), Cantón Aguarico, Reserva Etnica Huaorani, 260 m, *Dik* 1220 (U), Parque Nacional Yasuní, Pozo petrolero Daimi I, Conoco, 230 m, *Hurtado et al.* 21 (MO, U); Cantón Tena, Estación Biológica Jatun Sacha, 8 km E of Misahuallí, 450 m, *Maas et al.* 8600 (U), Río Napo, 8 km down the river from Puerto Misahuallí, 400 m, *Neill & Palacios* 7130 (U); Reserva Biológica Jatun Sacha, Río Napo, 8 km Río E of Misahuallí, 450 m, *Neill & Manning* 8014 (U); Estación Experimental IN-IAP Payamino, 5 km NE of Coca, 250 m, *Palacios et al.* 1036 (AAU, F, U); Reserva Biológica Jatun Sacha, Río Napo, 8 km down the river from Misahuallí, on right side, 450 m, *Palacios* 1421 (U, MO); Cantón Loreto, Payamino-Loreto road, Comunidad Jumandi, 350 m, *Palacios* 10950 (F, U); 30 km NNW of Coca, Río Huashito, Site of Proyecto Huashito, 250 m, *T.D. Pennington* 10609 (U); Cantón Tena, Estación

Biológica Jatun Sacha, 400 m, *Revelo* 65 (U); Cantón Orellana, Sector Huashito, 20 km N of Coca, Propriedad de Palmoriente, 250 m, *Rubio* 249 (G, GB, K, MO); Estación Biológica Jatun Sacha, confluence of Río Arajuno and Río Puní, 450 m, *Zuleta* 39 (U), Estación Biológica Jatun Sacha, 400 m, *Zuleta* 364 (U). Pastaza: Cantón Arajuno, Campamentos temporales 11–12, 785 m, *Freire et al.* 3468 (U). Sucumbios: Reserva de Producción Faunística Cuyabeno, 1 km N of Laguna Grande, 265 m, *Valencia et al.* 68558 (AAU). **Peru.** Huánuco: Tingo Maria, *Asplund* 13193 (G, NY, S); Prov. Puerto Inca, Distr. Yuyapichis, Dantas, 270 m, *Kröll* 217 (G, NY), 569 (G), 623 (G), 781 (G); Prov. Pachitea, Distr. Honoria, *Schunke V.* 1203 (F, G, GH, K, MO, NY); Distr. Yuyapichis, Prov. Puerto Inca, Unidad Modelo de Manejo y Producción Forestal Dantas, 270 m, *Tello* 257 (G, MO, ibidem, *Timaná* 143 (G); Shapajilla, 630 m, *Woytkowski* 32 (F). Loreto: Prov. Maynas, Yanamono, Explorama Tourist Camp, Río Amazonas halfway between Indiana and mouth of Río Napo, 130 m, *Gentry et al.* 42403 (MO). Madre de Dios: Prov. Tambopata, Río Heath, Pto. San Antonio, 210 m, *Aguilar & Castro* 916 (U), 1076 (U), Comunidad Nativa, Sonene, ca 8 km above confluence with Río Madre de Dios, 180 m, *Alexiades et al.* 1210 (U); Prov. Manú, Parque Nacional de Manu, Estación Biológico Cocha Cashu, *Chatrou et al.* 157 (U), Parque Nacional “Bahuaja-Sonene”, *Díaz et al.* 9037 (U), *Díaz & Ramírez* 9548 (U); Cashucocha Camp, Río Manu, Parque Nacional de Manu, 380 m, *Gentry et al.* 26821 (U), 26952 (F); Cocha Cashu Biológico Station, Parque Nacional de Manu, 400 m, *Gentry* 43334 (U), *Gentry* 43537 (MO); Tambopata Nature Reserve, junction of Río La Torre and Río Tambopata, 250 m, *Gentry et al.* 57751 (MO); Prov. Tambopata, 15 km ENE of Puerto Maldonado, Cusco Amazónico, 200 m, *Gentry et al.* 68713 (MO); Prov. Manu, Puerto Maldonado, Los Amigos Biological Station, Madre de Dios River, *Janovec et al.* 2014 (U), ibidem, 270 m, *Maceda* 7 (U); Prov. Tambopata, Cusco Amazónico, 200 m, *Núñez et al.* 10607 (MO); Prov. Manu, Río Manu, Pakitsa - behind station, 350 m, *Sobrevila et al.* 1782 (F); Prov. Tambopata, Las Piedras, Cusco Amazónico, 200 m, *Timaná et al.* 1722, 2795, 3060 (MO). San Martín: Prov. Huallaga, Barranca, 380 m, *Albán C.* 5878 (F); Juanjuí, Alto Río Huallaga, 400 m, *Klug* 3798 (F, GH, K, MO, NY, S); Moyobamba - Tarapoto, km 20, 1000 m, *T.D. Pennington & Daza* 16708 (U); Prov. Mariscal Cáceres, Distr. Tocache Nuevo, Quebrada de Saule Grande (margen derecho del Río Huallaga), *Schunke V.* 4387 (G, GH, K, MO, NY, US). Ucayali: Prov. Pucallpa, Estación Experimental Alexander von Humboldt, *Begazo* 61, 99 (U); Prov. Coronel Portillo, Humboldt station, near Campo Verde, ca. 34 km SW of Pucallpa, 200 m, *Morawetz & Wallnöfer* 11-19985 (U); Prov. Coronel Portillo, Bosque Nacional Alexander von Humboldt, km 68 of Pucallpa-Tingo Maria road, 270 m, *Oliveira* 15 (MO); Prov. Ucayali, Distr. Pampa Hermosa, Pampa Hermosa, 240 m, *Reynel R.* 832 (U); Prov. Coronel Portillo, Bosque Nacional Alexander von Humboldt, km 68 of Pucallpa-Tingo Maria road, 270 m, *Salazar* 32 (MO); Canchahuayo, Río Ucayali, 500 m, *Vásquez et al.* 6958 (AAU, F, NY, MO, U).

Notes. This species is easily recognizable by a combination of oval and on both sides rounded leaves with an olive green colour, long and narrow petals (often drying blackish), and stipes and apicules that are excentrically placed.

Maas et al. 8773 from Bolivia: Inner layer of monocarps ca. 5 mm thick, orange-yellow, sweet and edible.

***Pseudephedranthus* Aristeg.**

Pseudephedranthus Aristeg., *Memoirs of the New York Botanical Garden* 18(2): 43. f. 10. 1969.

Type. *Pseudephedranthus fragrans* (R.E.Fr.) Aristeg. (= *Ephedranthus fragrans* R.E.Fr.).

Description. *Trees*; young twigs glabrous. *Leaves*: distichous, simple, entire, shortly petiolate, exstipulate; lamina medium-sized, elliptic, venation brochidodromous, primary vein raised above. *Inflorescences* axillary, 1–4-flowered with 2nd order flowers originating from axils of lower bracts (or possibly also through accessory buds), often persisting on older leafless branchlets, pedicels with articulation in lower part and with 3–5 bracts, the uppermost bract above the articulation. *Flowers* actinomorphic, bisexual or staminate (androdioecious), 3-merous, perianth consisting of one whorl of sepals and two whorls of petals; sepals 3, valvate, basally connate, much shorter than the petals; petals 6, imbricate, elliptic, free, subequal; staminate flowers: torus conical, stamens numerous, extrorse, filament very short, apical prolongation of connective broadly ovoid in basal stamens to discoid in distal stamens; bisexual flowers: torus slightly raised, stamens numerous, but less so than in staminate flowers, apical prolongation of connective broadly ovoid; carpels numerous, spirally arranged, free, ovary 1-locular, with 1 basal ovule, style absent, stigma ovoid, papillate. *Fruit* apocarpous, composed of few, indehiscent monocarps, these ellipsoid, distinctly stipitate. *Seed* 1, pale brown, rumination lamellate in 2–4 parts, raphe a distinct groove.

Distribution. Two species in the Amazon regions of Venezuela and Brazil, and in Guyana and Suriname.

Key to the species of *Pseudephedranthus*

- 1a Petioles 3–5 mm long; leaf base acute to attenuate; outer side of outer petals (rather) densely covered with appressed hairs, inner side of outer petals and both sides of inner petals densely covered with white or greyish white curly hairs (Suriname, and the Brazilian state of Pará)..... ***P. enigmaticus***
- 1b Petioles 8–12 mm long; leaf base obtuse to less often acute, the extreme base shortly attenuate or not; outer side of outer and inner petals densely covered with appressed, not curly hairs to ca. 0.1 mm, inner side of outer and inner petals sparsely covered with appressed hairs or glabrous (Upper Río Negro region of Brazil and Venezuela) ***P. fragrans***

***Pseudephedranthus enigmaticus* Maas & Westra, sp. nov.**

urn:lsid:ipni.org:names:77165709-1

Figs 4, 5

Diagnosis. Differing from *P. fragrans* by shorter petioles. Moreover, petals in *P. enigmaticus* are for a large part covered by a very dense indument of curly hairs, those of *P. fragrans* covered by a less dense indument of appressed hairs. Also, seeds in *P. enigmaticus* are ellipsoid instead of ovoid and smaller than in *P. fragrans*.

Type. SURINAME, Sipaliwini, Central Suriname Nature Reserve, ca. 4 km ENE of Kayserberg Airstrip, alt. 235 m, 4 June 2003, *Evans et al.* 3437 (holotype WAG! [barcode WAG.1584983]; isotype L! [barcode L.3724851]).

Description. *Tree*, 3–15 m tall, 12–20 cm diam.; young twigs glabrous. *Leaves*: petioles 3–5 by 1–2 mm; lamina narrowly elliptic, 12–22(–26) by 4–6(–9) cm (index 2.8–4), chartaceous, pale gray to greenish gray above *in sicco*, somewhat bullate above *in vivo*, greenish brown to pale brown below *in sicco*, base acute, apex acuminate (acumen 5–10 mm long), primary vein raised above, secondary veins 6–10 on either side of primary vein, raised above, smallest distance between secondary veins and margin 4–7 mm, tertiary veins raised, rarely flat above, reticulate. Only staminate flowers seen, *Inflorescence* axillary, 1–2(–several)-flowered, pedicels 3–12 mm by 0.5–2 mm, rather densely to sparsely covered with erect to appressed, brown hairs to ca. 1 mm long, soon glabrous; bracts 4–5, depressed ovate, 1–2 mm long, outer side rather densely to sparsely covered with erect to appressed, brown hairs; flower buds ellipsoid; sepals shallowly ovate-triangular, ca. 2 by 2–3 mm, outer side rather densely to sparsely covered with erect to appressed, brown hairs; petals white, tinged with pale green *in vivo*, oblong-elliptic to narrowly so, 7–12 by 3–6 mm, outer side of outer petals densely to rather densely covered with appressed, brown hairs, inner side densely covered with whitish or greyish-white, curly hairs except for the glabrous base, outer side and apical part of inner petals densely covered with curly, white hairs; staminate torus conical, 2–2.5 mm long, ca. 1 mm diam. at base; stamens ca. 50, 2–2.5 mm long, apical prolongation of connective discoid, broadly elliptic. *Monocarps* 3–15, green *in vivo*, black *in sicco*, ellipsoid, 12–32 by 7–15 mm, glabrous or sparsely covered with appressed hairs, apex rounded, wall 0.2–0.5 mm thick, stipes 1–4 mm long, 1–1.5 mm diam. *Seed* ellipsoid, 12–19 by 7–10 mm, pale brown, transversely striate.

Distribution. Guyana, Suriname, and the Brazilian state of Pará. Fig. 1.

Habitat and ecology. In periodically inundated or non-inundated forest, on sandy or loamy soil, alt. 100–600 m. Flowering: May, June; fruiting: June, July, September.

Specimens examined. **Brazil.** Pará: Parque Indígena do Tumucumaque, Rio Pará de Oeste, Missão Tiriyo, *Cavalcante* 2579 (U); Rio Maicuru, Igarapé do Mutum, 31/2 hrs. por canoa de motor de poupa acima da pista de pouso do Lageiro, *Jangoux & Ribeiro* 1555 (L, RB); W bank of Rio Maicuru, ca. 23 km upstream from Lageira airstrip, N side of Mutum stream, *Strudwick et al.* 3808 (U). **Guyana.** Takutu-U Region, Rupununi River, between Kwattamang Landing and Rewa Village, 100 m, *Clarke et al.* 6750 (NY, US). **Suriname.** Suriname, Sipaliwini District, Sipaliwini River, Werehpai, 5 September 2010, *Bánki et al.* 1674 (L); Sipaliwini District, Si-

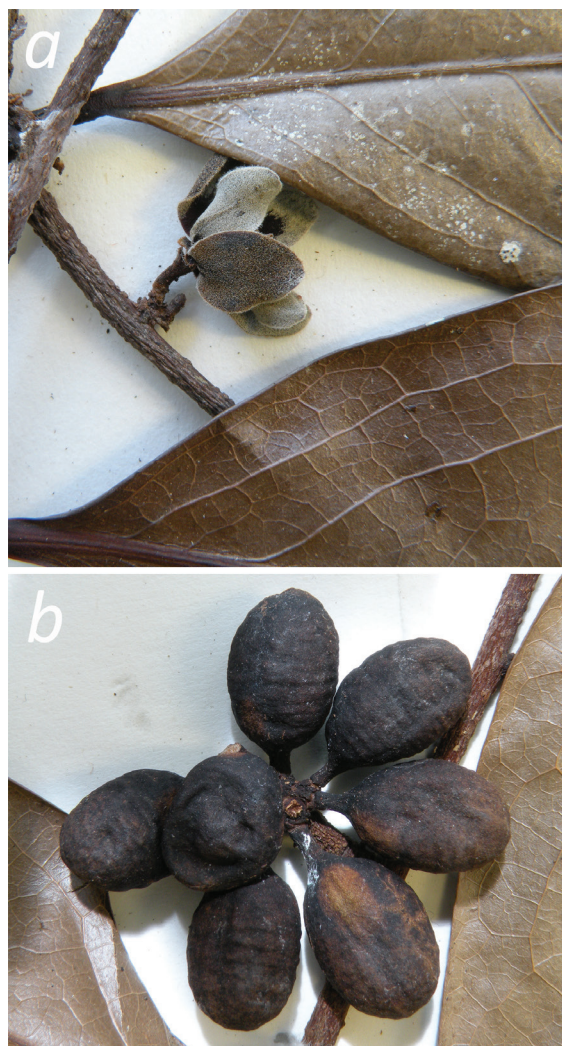


Figure 4. **a** Close up of flowering branch of *Pseudephedranthus enigmaticus* (Evans et al. 3437) **b** Close up of fruiting branch of *Pseudephedranthus enigmaticus* (Herrera C. et al. 9959).

paliwini River, *Bánki et al.* 1579 (L); Sipaliwini, vicinity of camp on W bank of Zuid River, across river (i.e. W and outside of) Central Suriname Nature Reserve, ca. 10 km straight-line distance SSE of Kayserberg Airstrip, 240 m, *Evans et al.* 3485 (L); Sipaliwini, Central Suriname Nature Reserve, on S slope of the first peak in Eilerts de Haan mountain range, ca. 7 km ENE of Kayserberg Airstrip, 400–600 m, *Herrera C. et al.* 9959 (L, WAG); Distr. Nickerie, area of Kabalebo Dam project, 30–130 m, *Lindeman & de Rooy* 752 (U); Sipaliwini, Morro Grande camp forest island, 6 km W of Morro Grande dome, 360 m, *Oldenburger et al.* 416 (U); Sipaliwini, Central Suriname Nature Reserve, 2–5 km SE of E end of Kayserberg Airstrip, 235 m, *Rosário* 1796 (L); Sipaliwini, Central Suriname Nature Reserve, 2–5 km ENE of Kayserberg

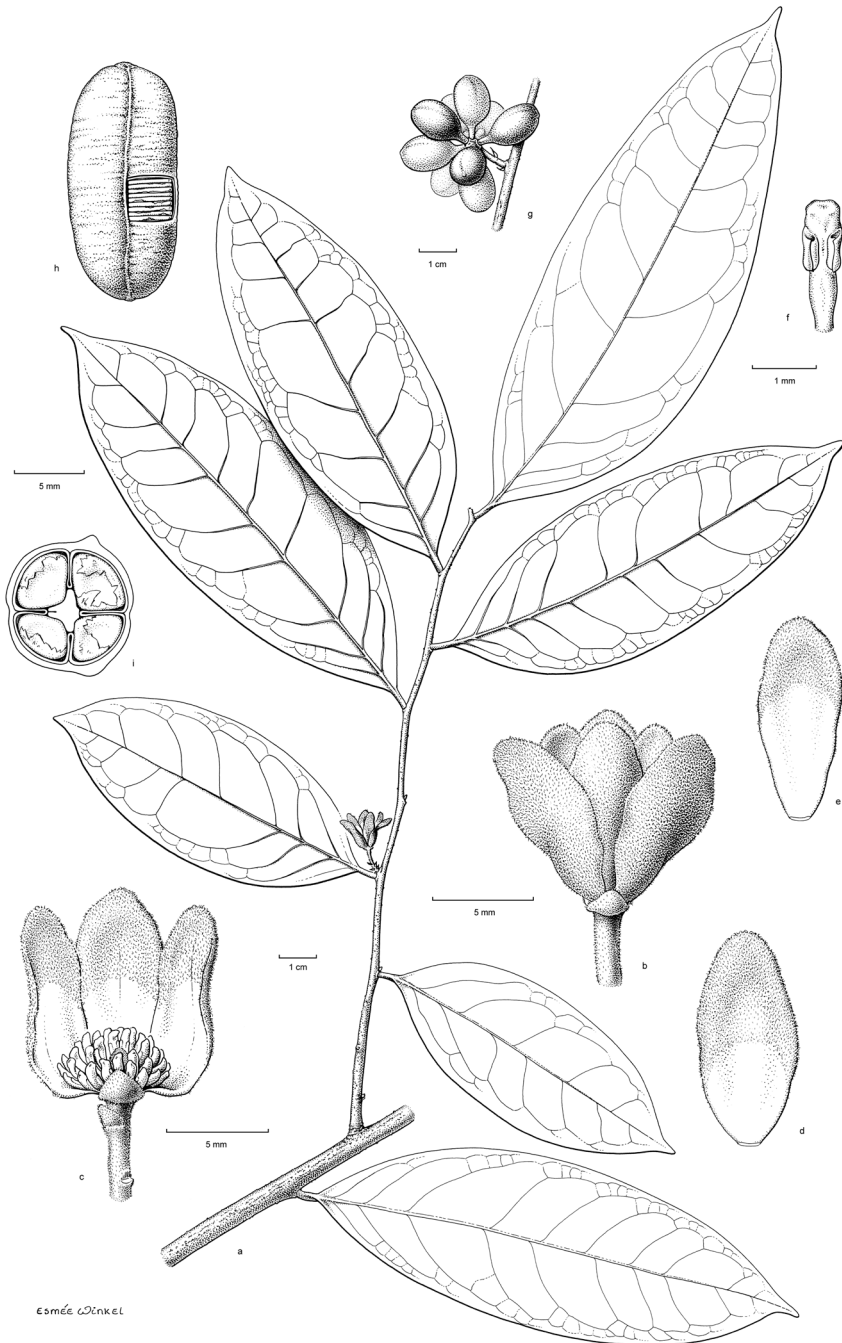


Figure 5. Drawing of *Pseudephedranthus enigmaticus*. **a** Flowering branch (Clarke 3420, U) **b** Flower in lateral view **c** Staminate flower in longitudinal section **d** Outer petal **e** Inner petal **f** Stamen (**b–f** Evans et al. 3437, WAG) **g** Fruit (Herrera C. 9959, WAG) **h** Seed, small part of seed coat removed to show lamellate ruminations **i** Cross section of monocarp and enclosed seed showing 4-parted rumination (**h–i** Strudwick et al. 3808, U). Drawings by Esmée Winkel.

Airstrip, 235 m, *Rosário et al.* 1829 (MO); Sipaliwini, Central Suriname Nature Reserve, vicinity of camp at southern base of the first peak in Eilerts de Haan mountain range, 250–350 m, *C.S. & D.O. Rosario* 2176 (L).

Notes. Material of this species had previously been filed in herbaria under different generic names such as *Crematosperma*, *Guatteria*, *Klarobelia*, *Malmea*, *Oxandra*, and *Rollinia* (which is quite aberrant!). The confusion is aptly expressed in the epithet “enigmaticus”. This new species fits quite well, however, within the genus *Pseudephedranthus* (segregated from *Ephedranthus* by Aristeguieta in 1969), among others by the leaf venation, fruit and seed structure, and the strong similarity of the flowers. We acknowledge the fact that Pirie et al. (2006) demonstrated that *Pseudephedranthus fragrans* is nested in *Klarobelia*. From a morphological point of view (leaves and venation; flower morphology) this is quite surprising, given that overall morphology of *Klarobelia* is homogenous, and *Pseudephedranthus* is deviant from the general *Klarobelia* morphology. Therefore, we prefer to describe this new species in *Pseudephedranthus* to reflect the morphological similarity to *P. fragrans*. *P. enigmaticus* is distinct from *P. fragrans* by shorter petioles and, particularly, by the much denser indument of small curly hairs on most of the inner side of the petals (versus mostly small straight hairs). Also, seeds in *P. enigmaticus* are ellipsoid and 12–19 by 7–10 mm in contrast to *P. fragrans* where they are ovoid and larger (25–30 by 13–15 mm).

The specimens investigated here were either fruiting or flowering, the flowers all being staminate. Carpel bearing flowers are still needed to complete the description.

***Pseudephedranthus fragrans* (R.E.Fr.) Aristeg.**

Figs 6, 7

Pseudephedranthus fragrans (R.E.Fr.) Aristeg., *Memoirs of the New York Botanical Garden* 18(2): 43. f. 10. 1969.

Ephedranthus fragrans R.E.Fr., *Memoirs of the New York Botanical Garden* 9(3): 327. 1957.

Type. VENEZUELA. Amazonas: Río Negro, occasional on lower slopes of Piedra Nuncia, just N of Piedra de Cucuy, 100–150 m, 10 April 1953, *Maguire & Wurdack* 34954 (holotype S [barcode S.85211]; isotypes F, GH, NY, P, S).

Description. *Tree*, 10–20 m tall, 15–30 cm diam.; young twigs glabrous, fissured, covered with a white, waxy surface. *Leaves*: petioles 8–12 by 1–2 mm; lamina elliptic to narrowly elliptic, (5–)10–25 by (2.5–)4–7(–9) cm (index 2–3), chartaceous to coriaceous, dark greenish-brown above, pale green below, shiny and glabrous on both sides, base obtuse to less often acute, the extreme base shortly attenuate or not, apex shortly acuminate (acumen to ca. 15 mm long), secondary veins 5–9 on either side of primary vein, raised above, smallest distance between secondary veins and margin 2–6 mm, tertiary veins raised above, reticulate. *Inflorescence* axillary, 1–2(–more)-flowered, usually on older parts of branchlets; pedicels 4–8 by ca. 1 mm, fruiting

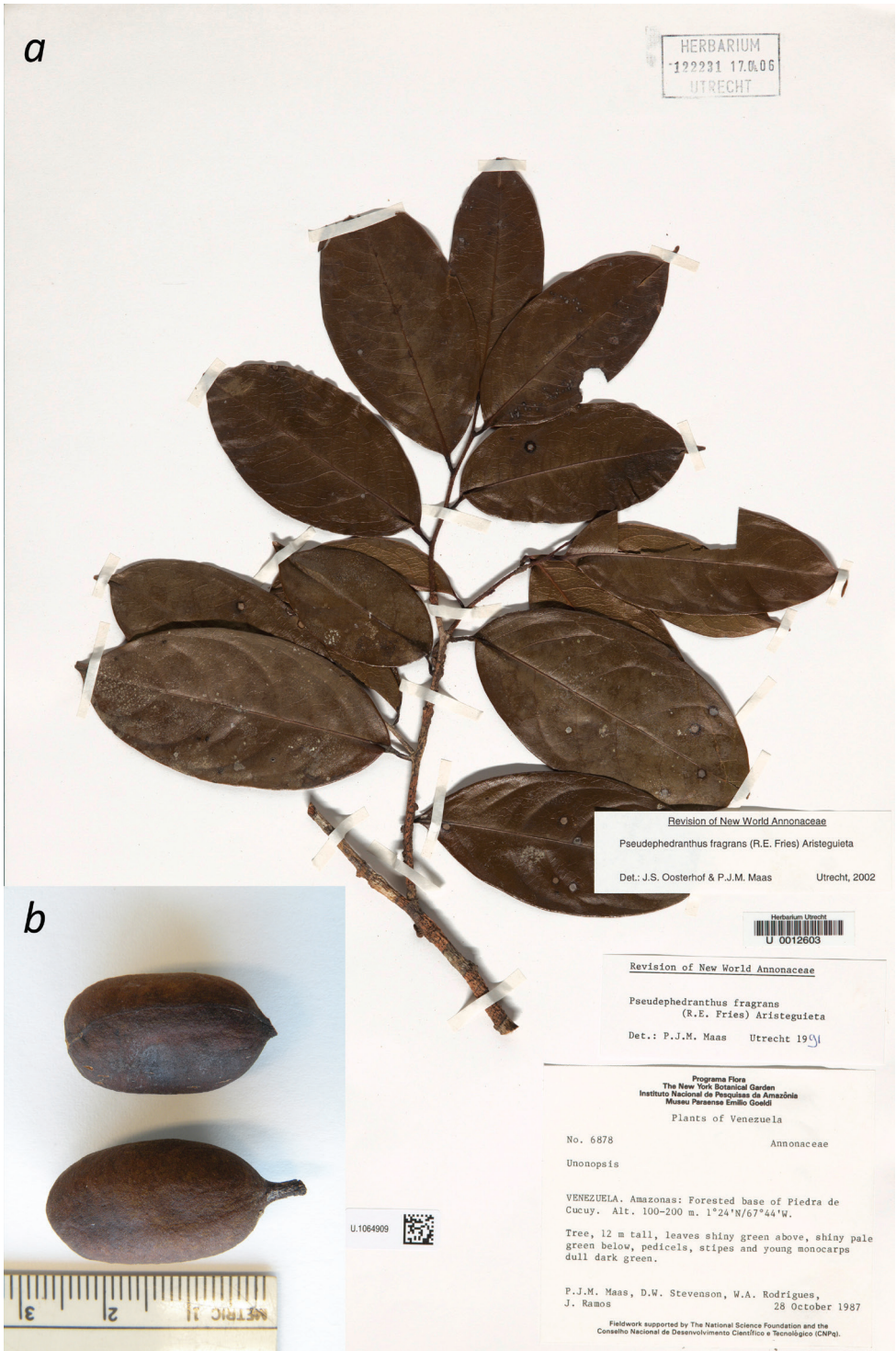


Figure 6. a Branch of *Pseudephedranthus fragrans* b Monocarps (Maas et al. 6878, U).



Figure 7. Fruit of *Pseudephedranthus fragrans*. Venezuela, Amazonas: Piedra de Cucuy (Maas et al. 6878).

pedicels to ca. 10 by 2–4 mm, densely covered with appressed hairs to ca. 0.1 mm long; bracts 3–5, triangular, 0.7–1 mm long, outer side densely covered with appressed hairs to ca. 0.1 mm long; flower buds broadly ovoid; sepals shallowly ovate-triangular, 1.5–2 by 3 mm, outer side densely covered with appressed hairs to ca. 0.1 mm long; petals white to cream *in vivo*, narrowly elliptic, 9–15 by 3–5 mm, outer side densely covered with appressed hairs to ca. 0.1 mm long; staminate flowers: torus concave, ca. 2.5 mm long, stamens ca. 50, 1.5–2 mm long, basal stamens with a broadly ovoid prolongation of connective, the distal ones with a discoid prolongation of connective, carpels absent; bisexual flowers: torus slightly raised, ca. 1 mm long, stamens ca. 15, 1.5–2.5 mm long, apical prolongation of connective broadly ovoid, carpels ca. 25, ca. 3 mm long, densely covered with appressed hairs to ca. 0.1 mm long, stigma ca. 1 mm long. *Monocarps* 3–10, green, maturing orange *in vivo*, pale brown *in sicco*, ellipsoid, 25–32 by 12–15 mm, apex rounded, sparsely covered with appressed hairs to ca. 0.4 mm long to glabrous, wall 0.2{–1} mm thick; stipes 2–4 by 1.5 mm. *Seed* ovoid, 25–30 by 13–15 mm, pale brown, transversely striate.

Distribution. Restricted to the Upper Rio Negro region of Brazil and adjacent Venezuela. Fig. 1.

Habitat and ecology. In lowland rain forest (in forested hills at the base of Piedra de Cucuy, Maas, pers. comm.). At elevations of 50–600 m. Flowering: April, December; fruiting: October, December.

Specimens examined. Venezuela. Amazonas: Río Negro, forested base of Piedra de Cucuy, 100–200 m, *Maas et al.* 6878 (INPA, MO, NY, RB, S, U, WU). **Brazil.** Amazonas: Rio Cauaburí, Caño Tucano, 125 m, *Maguire et al.* 60189 (F, GH, MO, NY, S), 60191 (MO, NY, S, US); Rio Negro, Rio Cauaburí, Rio Tucano, Camp Tucano (Camp No. 2), 850 ft., *Maguire et al.* 60332 (MO); between Palmito Camp and Camp Tatu, 400–600 m, *N.T. Silva & Brazão* 60683 (NY, US); Rio Negro, Rio Cauaburí, Rio Maturacá, between Camp III and Maloca, *N.T. Silva & Brazão* 60738 (NY).

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Two new endemic tree species from Puerto Rico: *Pisonia horneae* and *Pisonia roqueae* (Nyctaginaceae)

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Abstract

In this paper, we describe two endemic tree species of *Pisonia* (Caryophyllales: Nyctaginaceae) from Puerto Rico that were erroneously catalogued under the single name *Pisonia subcordata* var. *typica* f. *gigantophylla*, misidentified as *P. albida* or *P. subcordata*, and informally named as “*P. borinquena*” and “*P. woodburyana*”. The species here named as *P. horneae* is a rare to locally occasional tree from low elevations in the Northern Karst and the Sierra de Cayey. The other species, here named as *P. roqueae*, is a rare to locally common tree from mid to high elevations in the Central Mountain Range and the Luquillo Mountains. We provide an account of the taxonomical and nomenclatural history of both species, images, conservation notes, a distribution map, and a key to distinguish the species of *Pisonia* present in Puerto Rico.

Resumen

En esta publicación se describen dos especies de árboles endémicos de *Pisonia* (Caryophyllales: Nyctaginaceae) de Puerto Rico, las cuales fueron erróneamente catalogadas bajo un solo nombre como *Pisonia subcordata* var. *typica* f. *gigantophylla*, mal identificadas como *P. albida* o *P. subcordata*, e informalmente nombradas como “*P. borinquena*” y “*P. woodburyana*”. La especie aquí nombrada como *P. horneae* es un árbol raro a localmente ocasional de bajas elevaciones en el Karso Norteño y la Sierra de Cayey. La segunda especie aquí nombrada como *P. roqueae* es un árbol raro o localmente común de elevaciones medias a altas en la Cordillera Central y las Montañas de Luquillo. Se provee un recuento de la historia taxonómica y nomenclatural de ambas especies, imágenes, notas de conservación, un mapa de distribución, y una clave para distinguir las especies de *Pisonia* presentes en Puerto Rico.

* Both authors contributed equally to this work.

Keywords

Ana Roqué de Duprey, Caribbean, Caryophyllales, Frances W. Horne, *Pisonia subcordata* var. *typica* f. *gigantophylla*, “*Pisonia borinquena*”, “*Pisonia woodburyana*”, West Indies

Introduction

The Caribbean Islands are considered one of the hottest hotspots of world biodiversity (Myers et al. 2000) with a richness that has not been fully documented yet, as evidenced by the dozens of new species described from the region every year (Hedges and Conn 2012, Corgosinho and Schizas 2013, Mercado-Díaz et al. 2014, Cardoso and Braga 2015, Aguirre-Santoro et al. 2016, Carmenate-Reyes et al. 2017). For example, Puerto Rico – the smallest of the Greater Antilles – is probably the island with the best documented flora of the region with about 2331 species of native plants (Gann et al. 2015–2017). In spite of the intensive botanical work conducted in the island for more than a century, new species and rediscoveries of plants are still being documented (e.g., in the last decade: Trejo-Torres 2008, 2009, Vega et al. 2010, Caraballo-Ortiz 2013, Bornstein et al. 2014, Trejo-Torres et al. 2014).

An uncertain name used for a *Pisonia*

Pisonia subcordata Swartz var. *typica* Heimerl f. *gigantophylla* Heimerl (Caryophyllales: Nyctaginaceae) was described by Heimerl (1896) using a set of sterile specimens gathered by Paul E. E. Sintenis from at least two localities in central Puerto Rico. Nine years later, Heimerl and Urban (1905) synonymized this name under *Pisonia subcordata* var. *typica* Heimerl, as they considered the type collections to be juvenile parts of this species. In 1918, Standley reclassified *P. subcordata* var. *typica* f. *gigantophylla* as a synonym of *P. albida* (Heimerl) Britton ex Standley (Standley 1918). Coincidentally, in 1980 Westra examined and annotated the duplicates of specimen Sintenis 2705 (B, K) and considered them as a mixed collection containing parts of *P. albida* and *P. subcordata* Swartz.

After examining the type material of the abovementioned taxa, additional specimens, and living plants, we have concluded that the three sets of specimens used to describe *P. subcordata* var. *typica* f. *gigantophylla* (Sintenis 2141, 2705, and 4355 [sic. 4335]) comprise two taxa, which however, do not match any of the currently recognized *Pisonia* species from Puerto Rico or the Caribbean. While the two duplicates of Sintenis 2141 (GH, K) represent the same taxon, at least two of the three duplicates of Sintenis 2705 (B, K, and possibly GH) are mixed and include the same taxon as Sintenis 2141 as well as a second taxon. Although these specimens clearly represent two taxa, their sterile and fragmented nature, as well as the scant protologue, brings uncertainties to assign identities to each of the pieces. Moreover, Sintenis 4355 has not been located yet, and we lack information on its identity or locality.

Two informal names for *Pisonia*

In 1985 and 1993, George R. Proctor informally recognized two species of *Pisonia* from Puerto Rico and annotated a series of specimens as “*Pisonia borinquena* Proctor” (Britton 5628 (NY); Caminero 282, 443 (MAPR); Little 13380, 13404 (NY); Proctor 45465 (FTG, UPR); Proctor 43322, 44432, 48064 (US)) and “*Pisonia woodburyana* Proctor” (Britton 1515 (NY); Gleason A134 (NY); Liogier 10670 (NY, US); Proctor 41173 (IJ, UPR); Proctor 43284, 45450 (US); Woodbury s.n. (NY)). These unpublished names have been used in other herbarium specimens and databases, as well as in floristic assessments, scientific presentations and publications, and technical reports (e.g., Francis et al. 1998, Acevedo-Rodríguez and Axelrod 1999, Antoun et al. 1999, 2001, Alemañy-Merly 2000, Caraballo-Ortiz and Santiago-Valentín 2005, Caraballo-Ortiz 2007, Axelrod 2011, Trejo-Torres et al. 2011, Gann et al. 2015–2017).

Our analysis of living and dried specimens shows that the two species of *Pisonia* recognized by Proctor represent the same two taxa involved in the description of *P. subcordata* var. *typica* f. *gigantophylla*. Therefore, our objective in this paper is to clarify the identities and assign new names for these two species of *Pisonia*. For each species, we provide descriptions, illustrations, and comments on their abundance, natural history and conservation. In addition, we provide a distribution map and a dichotomous artificial key for the six species of *Pisonia* reported for Puerto Rico.

Materials and methods

Information on living plants and natural history was obtained from field observations and cultivated plants growing at the nursery of the Conservation Trust of Puerto Rico at San Juan. Species descriptions and measurements were obtained from field notes and dried herbarium specimens.

Taxonomic treatment

Pisonia borneae Trejo & Caraballo, sp. nov.

urn:lsid:ipni.org:names:77165902-1

Figs 1, 2, 3, 4

Pisonia subcordata sensu Heimerl and Urban (1905), not Swartz (1788).

Pisonia subcordata Swartz var. *typica* Heimerl f. *gigantophylla* Heimerl, Bot. Jahrb. Syst. 21: 630. 1896. Pro parte (see Additional specimens examined section).

“*Pisonia woodburyana* Proctor”, *nomen nudum*.

Type. PUERTO RICO. Municipality of Quebradillas, Bo. Terranova, old train road to El Tunel Negro, from Highway #2, border of dirt road near GPS [18.4840°, -66.9536°],

5 m, 12 Jul 2003, J.C. Trejo, P. Vives, A.M. Camacho, and M.A. Caraballo 2310 (holotype: UPR!; isotypes: FLAS! [#230768, 230769]).

Diagnosis. *Pisonia horneae* is distinguished from congeners from Puerto Rico by a combination of the following characters: leaves membranaceous and puberulous, twigs puberulous, fruits elliptic-ob lanceolate and grayish with 10 rows of viscid glands along their whole length.

Description. *Trees* dioecious to 10 m high with trunks to 30 cm in diameter. *Bark* finely and vertically striated, hazel, grayish, or silvery, sometimes lustrous, with lenticels about 3 mm in diameter. *Twigs* slightly ancipitous, greenish, and puberulous when young; terete, puberulent, and grayish when old. *Leaves* deciduous, clustered towards the ends of branches; opposite or subopposite, decussate; petioles to 7 cm long, grayish; blades ovate, elliptic, obovate, or roundish, sometimes asymmetric, 9–26 (–30) × 11–16 (–20) cm, apex acute to rounded, sometimes cuspidate, base acute to rounded, sometimes cuneate, cordate, or oblique, margin entire or wavy; adaxial side glabrous or puberulent; abaxial side puberulous, 0.1–0.4 mm long; chartaceous, drying membranaceous, brittle, light green above, paler below; veins pinnate, reticulate, secondary veins arcuate, up to twelve pairs, opposite or alternate, pale brown; abaxially pubescent and raised up to the secondary ramifications, main vein purplish on young plants. *Inflorescences* terminal or axillary, dendroid, to 7 cm long, pale green, puberulous; crown compact, flabellate; *Flowers* slightly fragrant; staminate flowers campanulate at anthesis, greenish white, 4 mm long; pistillate flowers cylindrical-campanulate at anthesis, 2 mm long. *Infructescences* dendroid, 10–16 cm long, drying grayish; peduncle terete, angled at base; crown lax, branches forked or pseudo-dichotomous; terminal branches with minute subulate bracteoles close to the anthocarps. *Fruits* anthocarps (achenes), elliptic to oblanceolate, 10–13 × 3 mm, longitudinally striate, tip cuspidate; husk softly ligneous, about 0.3 mm thick, with ten lines of viscid glands over ribs along its whole length, puberulous; glands capitate, about 0.5 mm long.

Habitat and ecology. *Pisonia horneae* is found in moist limestone forests at low elevations in the Northern Karst of Puerto Rico (10–150 m; Fig. 3), frequently on slopes and forest edges with relatively undisturbed vegetation. In eastern Puerto Rico, the species have been recorded from volcanic moist substrates from approximately 10 to 300 m (Fig. 3).

Although *P. horneae* is a species from low elevations and can grow near the coast, it has not been observed as part of the coastal vegetation. In contrast, the congener *P. subcordata* is distributed on coastal zones directly exposed to sea spray on elevations typically less than 10 m.

Phenology. *Pisonia horneae* sheds leaves during March and April. After shedding, the plant usually produces flowers by April, though flowers have been also recorded in January. Fruits have been recorded in May and June.

Vernacular names. As with other tree species of *Pisonia* from Puerto Rico, *P. horneae* is locally known as *corcho*.

Etymology. We are honored to dedicate this species to Frances Elvira Worth Horne (1873–1967), an American illustrator who lived in Puerto Rico and, for 45

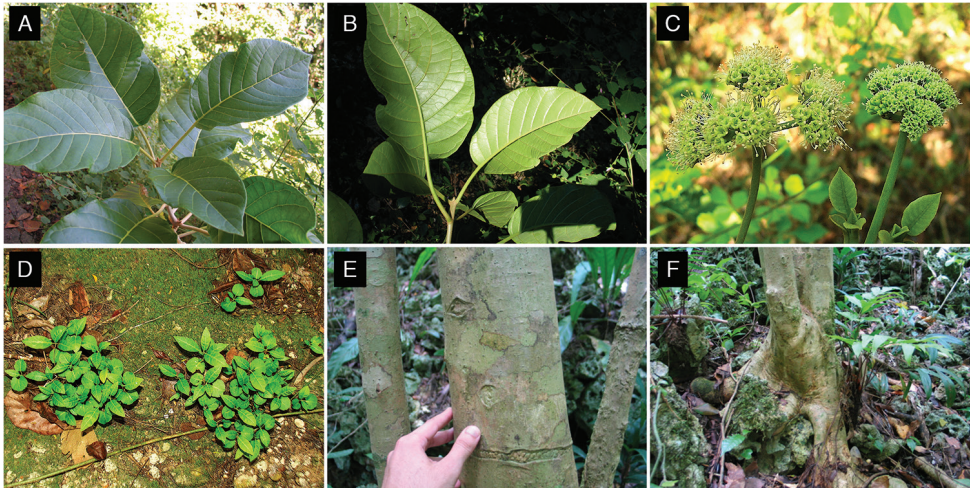


Figure 1. *Pisonia horneae*. **A** Branch showing adaxial side of leaves **B** Abaxial side of leaves **C** Staminate (left) and pistillate (right) flowers at anthesis **D** Seedlings **E** Bark **F** Lower trunk of an adult tree. Note the characteristic swollen base observed in all *Pisonia* from Puerto Rico, where the trunk base wraps over the rocks as it grows. Photo credits: **A–B, E–F**: JCTT; **C–D**: MACO.

years, painted its plants and birds. Ms. Horne composed 750 watercolors of Puerto Rican plants to complement Nathaniel Lord Britton's *Flora Borinqueña* (Jackson 1997), a popular book on common plants of Puerto Rico which was never published. Only 44 of her plant watercolors were published in the botanical journal *Addisonia* from 1922 to 1932 (Jackson, 1997), while 48 were included in the book *Common Trees of Puerto Rico and the Virgin Islands* (Little et al. 1977). She donated her collection of plant watercolors to The New York Botanical Garden in 1963 (Jackson 1997), which can be consulted at <http://plants.jstor.org> [accessed 02.08.2017]).

Abundance and conservation. *Pisonia horneae* can be considered a rare to locally occasional species. Despite its broad distributional range along the Northern Karst of Puerto Rico, trees of *P. horneae* are usually found isolated or forming small groups. The only localities where we have observed the species as locally common include a few karstic ravines in the municipality of Quebradillas and some limestone hills with relatively undisturbed forest in Ciales (Bo. Hato Viejo) and Dorado (Bo. Rio Lajas). *Pisonia horneae* seems to be extremely rare outside the Northern Karst, as the species is only known from three scattered localities in eastern Puerto Rico in the municipalities of Rio Grande, San Lorenzo, and Yabucoa. In Rio Grande, *P. horneae* seems to be uncommon and is only known from a single specimen (described below) and an unvouchered observation of a solitary sapling made in 2006 by biologist J. A. Sustache from the SJ Herbarium (pers. comm.). The authors visited the historical locality of Cerro Gregorio in San Lorenzo in December 2016, but no *Pisonia* were located. The locality in Yabucoa contained a single individual; however, it was fruiting and multiple seedlings were observed around the tree, suggesting the presence of additional individuals at the site (see Discussion section).

Additional unvouchered localities where *P. horneae* has been reported include Arecibo (Bo. Cambalache, Cambalache State Forest, 30 m, 25 Jul 2007, MACO), Bayamón (Bo. Volcán, 45 m, 26 Dec 2007, MACO; Bo. Juan Sánchez, Parque Julio Enrique Monagas, 35 m, 13 Jan 2008, MACO) and Carolina (Bo. Hoyo Mulas, 10 m, 07 Feb 2008, MACO and N.I. Cacho).

Pisonia horneae is currently designated as a critical element of the flora of Puerto Rico by the Department of Natural and Environmental Resources of The Commonwealth of Puerto Rico (listed as *P. woodburyana*) (DNER 2008). It is also preliminary considered as an “Imperiled” (*sensu* NatureServe) or “Vulnerable” (*sensu* IUCN Red List) species by Gann et al. (2015–2017).

Additional specimens examined. PUERTO RICO. Arecibo: Cambalache, 50 m, 19 Jan 1989, Liogier 36633 (NY, UPR); Bo. Dominguito, Mata de Plátano Biological Station, 93 m, 25 Jul 2003, Trejo 2349 (UPR); 142 m, 25 Jul 2003, Acevedo-Rdgz. 13404 (FTG, US); 140 m, 12 Jan 2016, Caraballo and Rivera 3380 (PAC, UPR); Finca El Tallonal, 101 m, 15 Aug 2003, Trejo 2364 (UPRRP); 100 m, 16 Mar 2004, Trejo 2585 (UPR); Bo. Miraflores, sector Biáfara, Finca Dentón, 175 m, 14 Sept 2003, Trejo 2397 (UPR); 169 m, 28 Nov 2003, Trejo 2482 (UPR); **Bayamón:** 21 Feb 1959, Woodbury s.n. (NY [#00680786], UPR [#000772]); Bo. Hato Tejas, 45 m, 21 May 2009, Caraballo et al. 2871 (UPR); **Canóvanas:** Bo. San Isidro, 10 m, 2 Jan 2017, Caraballo 3382 (PAC); **Ciales:** Bo. Hato Viejo, 160 m, 9 Jun 2007, Caraballo et al. 1818 (UPR); 130 m, 9 Jun 2007, Caraballo et al. 1822 (UPR); **Dorado:** Bo. Higuillar, 8 Feb 1964, Liogier 10670 (F, MAPR, NY); Bo. Sabana, northeast of Regadera, off Road 691, 5–10 m, 12 May 1985, Proctor 41173 (UPR); Bo. Río Lajas, 15 m, 20 Jun 2003, Trejo and Caraballo 2279 (UPR); **Hatillo:** Bo. Capáez, ca. 5 m, 12 Jan 2006, Axelrod 13357 (UPRRP); **Isabela:** Quebrada La Sequia, 75 m, 2 Oct 2005, Caraballo et al. 639 (UPR); Guajataca gorge, 75 m, 26 Jan 2006, Caraballo et al. 1425 (UPR); Cordillera Jaicoa, 200 m, 23 Aug 2006, Caraballo 1196 (UPR); **Loíza:** Bo. Piñones, Piñones Forest Reverse, 13 Mar 1980, Del Llano s.n. (UPRRP [#96]); **Manatí:** Bo. Tierras Nuevas Saliente, 03 May 2001, Axelrod and Zachariades 11754 (UPRRP); Bo. Tierras Nuevas Poniente, Hacienda La Esperanza, 10 m, 14 Mar 2006, Caraballo et al. 919 (UPR); 12 m, 28 Mar 2006, Caraballo and Rivera 976 (UPR); **Moca:** Bo. Aceitunas, 175 m, 1 Mar 2008, Caraballo 2455 (UPR); **Quebradillas:** Bo. San José, Quebrada Bellaca, 52 m, 14 Oct 2003, Trejo and Caraballo 2444 (UPR); Bo. Cacao-Terranova, Quebrada Las Talas [El Gallo], 85 m, 21 Aug 2004, Trejo and Caraballo 2751 (UPR); Bo. Cocos, Quebrada Bellaca, 101 m, 9 Mar 2005, Trejo et al. 2873 (NY, UPR); limestone hills, ca. 150 m, 22 May 2005, Axelrod et al. 13062 (UPRRP); **Río Grande:** Rio Mar, 100 m, 5 Aug 1988, Liogier and Martorell 36637 (NY, UPR); **San Lorenzo:** Prope Hato Grande in sylva primaera montis Gregorio, 31 Aug 1885, Sintenis 2705 [mixed specimens, see Introduction and additional specimens examined for *P. roqueae*] (B [10-0217019]; GH [00037445]; K [K000036122]); **Toa Baja:** Bo. Candelaria, E sector, Mogotes de Nevárez, 20–90 m, 27 Mar 1989, Proctor 45450 (US); 5 Mar 1914, Britton et al. 1515 (NY); Campanillas, 8 Jan 1959, Woodbury s.n. (UPR [#027151]); Nevárez, 75 m, 16 Mar 2006, Caraballo et al. 931 (UPR); **Vega Alta:** Bo.



Figure 2. Isotype specimen of *Pisonia horneae* from the University of Florida Herbarium (Trejo et al. 2310, FLAS [230768]). Photo courtesy of The University of Florida Herbarium (FLAS) – Florida Museum of Natural History.

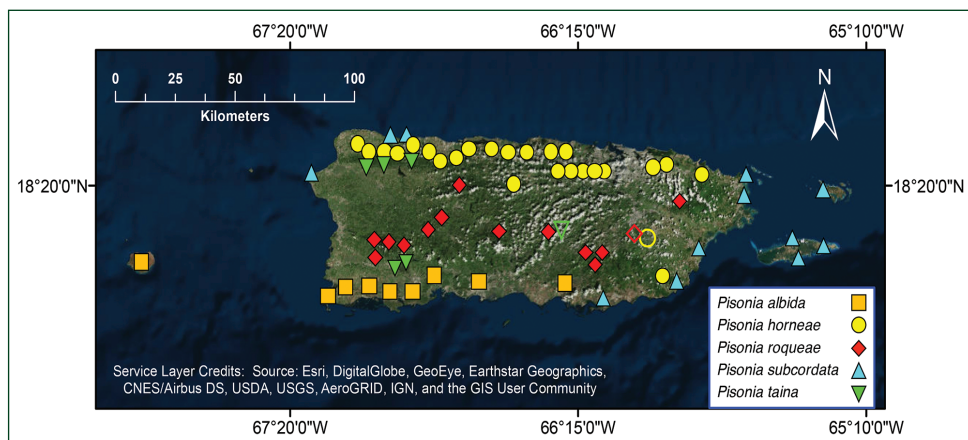


Figure 3. Distribution of *Pisonia* (excluding *P. aculeata*) in Puerto Rico and adjacent islands based on herbarium specimens. Empty symbols represent historical localities where populations have been presumably extirpated (see Trejo-Torres 2005 and this study for details).

Espinosa, 11 Feb 1926, Gleason and Cook A134 (NY); Bo. Sabana, 5–10 m, 12 May 1985, Proctor 41173 (IJ); hill NE of Regadera, 5 m, 1 Apr 1987, Proctor 43284 (US); Bo. Regadera, Axelrod 84 (UPRRP); **Yabucoa**: Cuchilla de Pandura, 175 m, 13 June 2006, Caraballo and Flecha 1088 (UPR); **Without locality**: without locality, 8 Nov 1886, Sintenis 4355 [see Introduction and additional specimens examined for *P. roqueae*] (B, n.v., probably lost).

***Pisonia roqueae* Trejo & Caraballo, sp. nov.**

urn:lsid:ipni.org:names:77165903-1

Figs 3, 4, 5, 6

Pisonia subcordata sensu Heimerl and Urban (1905), not Swartz (1788).

Pisonia subcordata Swartz var. *typica* Heimerl f. *gigantophylla* Heimerl. Bot. Jahrb. Syst. 21: 630. 1896. Pro parte (see Additional specimens examined section). Lectotype (designated here): Barranquitas: Prope Barranquitas in sylva primaera montis Torrecilla, 30 Oct 1885, Sintenis 2141 (GH [00037444]; K [H2005/00995]).

“*Pisonia borinquena* Proctor”, *nomen nudum*.

Type. PUERTO RICO. Municipio de San Germán, Bo. Caín Alto, Maricao State Forest, in a moist ravine, south of Road 120 km 15.6, 850 m, 23 June 1992, G.R. Proctor, R. Padrón, and R. Rivera 48064 (holotype: US! [#00707324]; isotype: MO! [#04580139]).

Diagnosis. *Pisonia roqueae* is distinguished from congeners from Puerto Rico by a combination of the following characters: staminate inflorescences with globose crowns, twigs glabrescent, leaves coriaceous and glabrescent, and fruits clavate and reddish-black with five rows of viscid glands on their distal half.

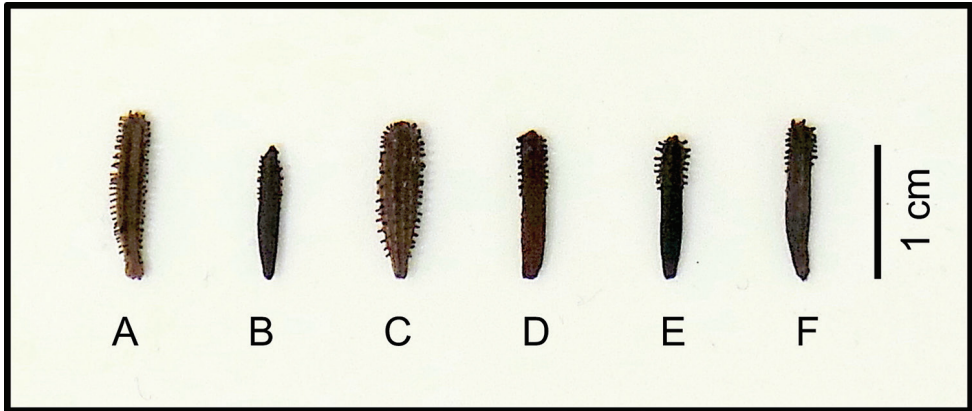


Figure 4. Fruits of the six species of *Pisonia* from Puerto Rico. **A** *P. aculeata* **B** *P. albida* **C** *P. horneae* **D** *P. roqueae* **E** *P. subcordata* **F** *P. taina*. Reference specimens: **A** Rose 3548, US **B** Little 13219, US **C** Acevedo-Rdgz. 13404, US **D** Proctor 44432, US **E** Breckon 7766, US; and **F** Trejo 2371, US.

Description. *Trees* dioecious up to 25 m high with trunks up to 1 m in diameter. *Bark* finely and vertically striated, grayish with lenticels about 3 mm in diameter. *Twigs* ancipitous, ferruginous-brownish, and pubescent when young; terete, glabrescent, and grayish when old. *Leaves* clustered towards the ends of branches, opposite, subopposite, or sub-verticillate, decussate; petioles up to 3.5 cm long, yellowish green; blades elliptic, obovate, or roundish, 10–17 (–25) cm × 7–12 (–16) cm, apex acute to rounded, sometimes cuspidate, base acute to cuneate, margin entire or slightly wavy, adaxial and abaxial sides glabrous, coriaceous, drying coriaceous, dark green above, slightly paler below; veins pinnate, reticulate, up to eleven pairs, opposite or alternate, blackish. *Inflorescences* terminal or axillary, dendroid, to 7 cm long; crowns compact; staminate inflorescences with a globose crown; pistillate inflorescences with a flabellate crown. *Flowers* fragrant; pistillate flowers with perianth cylindrical-campanulate-oblong at anthesis, 2 mm long, puberulent; staminate flowers perianth campanulate at anthesis, green, about 3 mm long, puberulent. *Infructescences* dendroid, 4–7 cm long, drying reddish or brownish; peduncle angled, 2–3 cm long; branches irregularly forked, with minute bracteoles around the base of anthocarps. *Fruits* anthocarps (achenes), clavate, 10–15 mm × 2 mm, longitudinally striate, tip cuspidate; husk glabrous, with five lines of glands over discrete ribs along the distal third or half; glands capitate, about 0.5 mm long, viscid.

Habitat and ecology. *Pisonia roqueae* is mainly distributed from middle to high elevations. In the Central Mountain Range and Sierra de Luquillo, the species have been found on wet and moist serpentine or volcanic forests from 480 to 950 m. The only known locality from the Northern Karst is on a wet limestone forest at ca. 330 m.

Unlike other species of *Pisonia* from Puerto Rico, adult trees of *P. roqueae* can reach considerable heights (> 15 m). Some large trees can be found at the Maricao State Forest and at the Luquillo Mountains in EL Verde Field Station, including the Luquillo Forest Dynamics Plot (<http://luq.lternet.edu>), where the species (treated as either “*P. borinquena*” or *P. subcordata*) has been included in long-term studies (e.g., Kress et al. 2010).

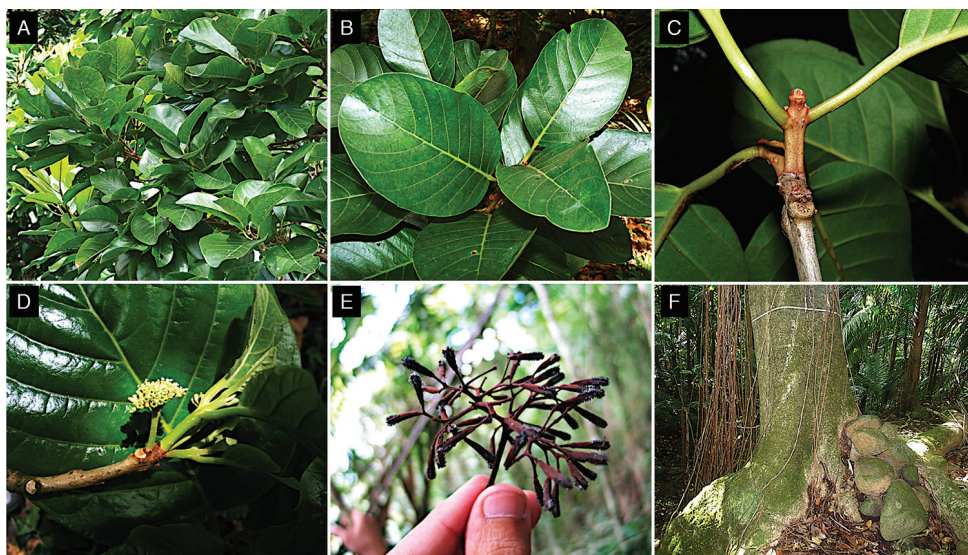


Figure 5. *Pisonia roqueae*. **A** Foliage **B** Adaxial side of mature leaves **C** Twig with ferrugineous-brownish pubescence, turning grayish as it ages **D** Pistillate inflorescence and young leaves **E** Ripe infructescence **F** Trunk of an adult tree. Photo credits: **A–C, F:** Fabiola Areces (Areces Berazain et al. 2013–2017); **D–E:** JCTT.

Some species of *Pisonia* from Puerto Rico, including *P. roqueae*, are common hosts for native species of mistletoes. For example, *P. albida* and *P. roqueae* are important hosts for *Phoradendron anceps* (Spreng.) M. Gómez (Santalaceae) at the Guánica State Forest and the Maricao State Forest, respectively (Pinto 2005). Similarly, *P. subcordata* has been reported as host for the mistletoes *Dendropemon caribaeus* Krug & Urb. (Loranthaceae) and *P. anceps* in northern Puerto Rico (M.A. Vives-Heyliger, pers. comm.).

Phenology. *Pisonia roqueae* has been recorded flowering in April, June, and July, and fruiting from January to April.

Vernacular names. As with other tree species of *Pisonia* from Puerto Rico, *P. roqueae* is locally known as *corcho* or *corcho blanco*.

Etymology. It is our honor to name *Pisonia roqueae* after Dr. Ana Cristina Roqué Geigel de Duprey (1853–1933), an amateur ethnobotanist from Puerto Rico who dedicated over three decades of her life to prepare the bilingual manuscript “*Botánica Antillana: Introduction to the study of the picturesque flora of Porto Rico and West Indies*”, aimed to make botany accessible to the general public. Her manuscript (Roqué de Duprey 1925) was never published and remained in oblivion to the botanical community until recently when JCTT and collaborators divulged its existence (Martínez 2015). Roqué de Duprey is mostly known for being an educator, writer, suffragist, and one of the founders of the University of Puerto Rico-Mayagüez, among other educational institutions.

Abundance and conservation. *Pisonia roqueae* has been observed as a locally common tree in two localities in the eastern and western Central Mountain Range (Monte La Torrecilla in Barranquitas and Maricao State Forest, respectively), occasional at the El Yunque



Figure 6. Holotype specimen of *Pisonia roqueae* from the National Herbarium of the Smithsonian Institution (Proctor et al. 48064, US [00707324]). Note the dendroid staminate inflorescences with globose crowns and the leafless branches during the flowering period. Photo courtesy of The United States National Herbarium, Smithsonian Institution.

National Forest (Luquillo Mountains), and rare elsewhere. It has been also recorded from Cayey (including an unvouchered locality at Bo. Jájome, 300–350 m, 18 Feb 2015, by O. Monzón), Guayama at Sierra de Cayey, and a single record from the Northern Karst

at Sabana Hoyos, Arecibo. The species is preliminary considered as “Imperiled” (*sensu* NatureServe) or “Vulnerable” (*sensu* IUCN Red List) by Gann et al. (2015–2017).

Additional specimens examined. PUERTO RICO. Arecibo: Bo. Sabana Hoyos, Finca Las Abras, 330 m, 9 Sept 2002, Trejo 1773 (UPRRP); 300 m, 10 Jul 2005, Trejo 2996 (UPR); **Barranquitas:** Bo. Barrancas, Monte La Torrecilla, 900–1100 m, 19–20 Mar 1915, Britton et al. 5628 (NY); 950 m, 1 Apr 1989, Proctor 45465 (FTG, UPR); 930 m, 24 Jul 2003, Trejo 2340 (FLAS, GH, UPR); Trejo 2342 (NY, UPR); Trejo 2343 (UPR); 5 Mar 2004, Trejo 2552 (UPR); Trejo 2553 (UPR); **Cayey:** Cercadillo, 600 m, 10 Feb 1983, Liogier 33875 (UPR); **Ciales:** Bo. Toro Negro, Tres Picachos, 700 m, 29 Jul 1993, Axelrod 6694 (JBSD, UPRRP); Bo. Cialitos, Los Tres Picachos, 675 m, 4 Apr 2008, Caraballo et al. 2561 (UPR); **Guayama:** Bo. Carmen, 290 m, 4 Jun 2016, Areces et al. 1140 (UPRRP); **Maricao:** Maricao Forest, 800 m, 26 Jun 1938, Sargent 506 (MO); 500 m, 2 Oct 1938, Sargent 633 (MO); Vivero de Peces, 850 m, 13 Jul 1950, Little 13380 (F, NY, UPR); 880 m, 15 Jul 1950, Little 13404 (F, NY, UPR); 26 Nov–4 Dec 1963, Duke 7134 (MO); Monte Alegrillo, 890–900 m, 5 Apr 1987, Proctor 43322 (US); Monte del Estado, 700 m, 6 May 1990, Caminero 282 (MAPR); Bo. Maricao Afuera, Monte del Estado, 850 m, 15 May 1991, Caminero 443 (MAPR); 480 m, 3 Apr 1996, Cedeño 881 (MAPR); 550 m, 4 Apr 1996, Cedeño 904 (MAPR); 840 m, 29 Feb 2004, Carlo 21 (UPR); 875 m, 27 May 2009, Caraballo et al. 2878 (UPR); s.c. s.n. (MAPR [016698]); **Sabana Grande:** Bo. Sabana-Tabonuco, 750 m, 5 Mar 2004, Trejo 2648 (UPR); **San Germán:** Bo. Caín Alto, Maricao Forest, 810–830 m, 23 Jan 1988, Proctor 44432 (US); **San Lorenzo:** Prope Hato Grande in sylva primaera montis Gregorio, 31 Aug 1885, Sintenis 2705 [mixed specimens, see Introduction and Additional specimens examined for *P. horneae*] (B [10-0217019]; GH [00037445]; K [K000036122]); **Río Grande:** El Verde Experimental Station, 29 Jun 1963, Smith 345 (EVFS [El Verde Field Station herbarium]); August 1963, Smith 470 (EVFS); 15 May 1964, Smith 1122 (EVFS); LTER plot, 350 m, 18 Jun 1990, Moestl 9 (EVFS); 419 m, 2 Mar 2013, Areces 748 (UPRRP); Río Sonadora, 350 m, 18 Jun 1991, Taylor 10453 (MO); 19 May 1994, Taylor 11689 (MO); **Utua:** without locality, 1916, Strube s.n. “A” (MO [#653326]); Bo. Roncador, Hacienda Verde, 514 m, 8 Aug 2004, Trejo 2731 (UPR); **Without locality:** without date, 8 Nov 1886, Sintenis 4355 [see Introduction and Additional specimens examined for *P. horneae*] (B, n.v., probably lost).

Discussion

With the formal addition of *P. horneae* and *P. roqueae*, the genus *Pisonia* in Puerto Rico is represented by six species, including three Puerto Rican endemics, two West Indian-restricted, and one pantropical. The distribution of the six species of *Pisonia* from Puerto Rico seems to follow a geographic and environmental pattern (Fig. 4): the vine *P. aculeata* L., the most widespread species in the genus with a worldwide distribution, is found at lower and middle elevations across the island; *P. albida*, a tree from Hispaniola and Puerto Rico, is distributed along dry coastal forests in southern lowlands; *P. horneae* is

distributed in the moist limestone forests of the Northern Karst and the eastern Central Mountain Range; *P. roqueae* is a species restricted to wet forests with elevations above 300 m along the Central Mountain Range and the Luquillo Mountains; *P. subcordata*, a tree distributed throughout the West Indies, is mainly found on coastal forests and thickets along the northern and eastern coast; and *P. taina* Trejo, an endemic and rare tree from Puerto Rico, has a scattered distribution across the central and western parts of the island (Trejo-Torres 2005). The distributional ranges of *P. albida*, *P. horneae*, *P. roqueae*, and *P. subcordata* seems to be consistent with specific substrate types and/or environmental conditions. However, we still lack scientific studies exploring possible biotic or abiotic factors shaping these distributions. A possible explanation is the existence of species-specific interactions with ectomycorrhizal fungi, which can help species survive in particular abiotic conditions (Van der Putten et al. 2010, Pickles et al. 2015) and have been found to interact with *Pisonia* and other members of the Pisonieae clade for at least 14 Myr (Hayward and Horton 2014). We also lack molecular studies to infer the phylogenetic relationships and species divergence times among Caribbean *Pisonia*.

The most reliable characteristics to set apart the six species of *Pisonia* from Puerto Rico are the morphology of inflorescences, flowers, and fruits along with leaf shape and size. However, *Pisonia* trees have a fugacious reproductive season and herbarium specimens are often found either sterile or fertile but lacking well-developed leaves (Fig. 6). As a result, many specimens are incomplete representations of the species, which has partially contributed to their misidentification (Trejo-Torres 2005). Therefore, when collecting *Pisonia* in the field we recommend to include, whenever possible, multiple vouchers from a single plant or population throughout seasons to properly document the range of phenological stages of plants, including fertile and sterile material. *Pisonia* fruits usually persist on branches or on the ground below trees during several months. Hence, we included a comparison of fruit morphology for the six species of *Pisonia* from Puerto Rico to assist with their identification (Fig. 4). Observational information may also assist with identification of the two-new species here described and we have noted that, unlike other *Pisonia* from Puerto Rico, fresh leaves of *P. horneae* are usually light green and young plants often have a purplish midvein. Likewise, leaves of *P. roqueae* are typically dark green and shiny.

Notes on reproduction and conservation

Most of the observed plants of *P. horneae* and *P. roqueae* were either isolated or forming small groups. Although both species have broad distributions within their respective habitats, most trees are restricted to ravine banks, cliffs, or rocky areas, especially for *P. horneae*. These observations might suggest that either trees tend to colonize these particular habitats due to physiological requirements, or that they represent relicts of a former, more continuous population that was severely fragmented during the intense deforestation period experienced in the island for the past centuries.

As with other *Pisonia* species, both *P. horneae* and *P. roqueae* are dioecious and their flowers might require pollen to set fruits (Douglas and Manos 2007, Nores et al. 2015).

However, at least two species of *Pisonia* from Puerto Rico (*P. taina* (Trejo-Torres 2005) and *P. subcordata* (M.A. Vives-Heyliger, pers. comm.)) have been reported as rarely sub-androecious, a condition that can help mitigate the impact of reproductive isolation by inbreeding. We currently lack information on reproductive biology for the vast majority of *Pisonia* species (Douglas and Manos 2007), but field observations made by one of the authors suggest that *P. horneae* and *P. subcordata* can achieve high fruit sets in sites where trees are locally abundant (MACO, pers. obs.). This suggests that *Pisonia*, as other dioecious tropical trees, can be particularly susceptible to reproductive failure from isolation and low population sizes (House 1992). Reproductive success of dioecious plants will depend on multiple factors such as the spatial distribution of trees, the male-female ratio within populations, and the foraging range of their effective pollinators (Lin et al. 2015). Thus, conservation efforts, besides protecting extant trees and natural areas where the species are present, should take into consideration increasing neighborhood densities by planting additional individuals near isolated trees, even though these actions would not warrant maintenance of genetic diversity and the long-term survival of the species.

To our knowledge, there are no reports of seed dispersal for any Neotropical *Pisonia*, which contrast with the Indo-Pacific region where seed dispersal has been well-documented, especially for *P. grandis* R. Br. and *P. umbellifera* (J.R. Forst. & G. Forst.) Seem. (e.g., Ridley 1930, Cleland 1952, Walker et al. 1991, Murphy and Legge 2003, Burger 2005). In 2005 after an intense fruiting season at one of the densest known populations of *P. horneae* at the municipality of Quebradillas in northwestern Puerto Rico, infructescences detached from branches when fruits were ripe, but they remained under their maternal trees. Subsequently, seedlings grew densely clustered and the vast majority of them died within the next few months (Fig. 1D; MACO, pers. obs.). These observations, along with the low recruitment detected beyond parental trees, suggest that at least some populations of *P. horneae* might be experiencing low seed dispersal rates.

Key to the species of *Pisonia* from Puerto Rico

- 1 Woody liana; twigs armed with spines ***P. aculeata***
- Shrubs or trees; twigs unarmed **2**
- 2 Leaf veins at abaxial side raised up to the finer ramifications ***P. taina***
- Leaf veins at abaxial side not raised up to the finer ramifications **3**
- 3 Twigs glabrescent (except at leaf axils); dried leaves coriaceous; fruits drying black or reddish black **4**
- Twigs puberulous; dried leaves chartaceous or membranaceous; fruits drying brown or gray **5**
- 4 Leaves puberulent, especially on the abaxial side and along veins; fruits ca. 1.3 cm long × 2.5 mm wide in average; staminate inflorescences with a globose crown ***P. roqueae***
- Leaves glabrous, sometimes slightly puberulent on the abaxial side at base of main vein; fruits ca. 1 cm long × 1.5 mm wide in average; staminate inflorescences with a flabellate crown ***P. subcordata***

- 5 Leaves up to 14 cm long, usually with a constant elliptic shape, chartaceous when dried, and whitish on the abaxial side; fruits brown, clavate with five rows of viscid glands along the distal end.....***P. albida***
- Leaves up to 40 cm long, variable in shape, membranaceous when dried, and greenish on the abaxial side; fruits gray, ellipsoid with ten rows of viscid glands along the whole fruit..... ***P. borneae***

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Exsiccatae

- Acevedo-Rdgz., P.A. 13404 (horneae)
Areces, F. 748, 1140 (roqueae)
Axelrod, F.S. 84 (horneae); 6694 (roqueae); 11754, 13062, 13357 (horneae)
Britton, N.L. 1515 (horneae); 5628 (roqueae)
Caminero, G. 282, 443 (roqueae)
Caraballo, M.A. 639, 919, 931, 976, 1088, 1196, 1425, 1818, 1822, 2455 (horneae);
2561 (roqueae); 2871 (horneae); 2878 (roqueae); 3380, 3382 (horneae)
Carlo, T.A. 21 (roqueae)
Cedeño, J.A. 881, 904 (roqueae)
Duke, J.A. 7134 (roqueae)
Gleason, H.A. A134 (horneae)
Liogier, H.A. 10670 (horneae); 33875 (roqueae); 36633, 36637 (horneae)
Little, E.L. 13380, 13404 (roqueae)
Moestl, S. 9 (roqueae)
Proctor, G.R. 41173, 43284 (horneae); 43322, 44432 (roqueae); 45450 (horneae);
45465, 48064 (roqueae)
Taylor, C.M. 10453, 11689 (roqueae)
Trejo, J.C. 1773 (roqueae); 2279, 2310 (hornena); 2340, 2342, 2343 (roqueae); 2349,
2364, 2397, 2444, 2482 (horneae); 2552, 2553 (roqueae); 2585 (horneae); 2648,
2731 (roqueae); 2751, 2873 (horneae); 2996 (roqueae)
Sargent, F.H. 506, 633 (roqueae)
Sintenis, P.E.E. 2141 (roqueae); 2705 (horneae and roqueae)
Smith, R.F. 345, 470, 1122 (roqueae)
Strube, L.B. s.n. "A" (roqueae)

A new disjunct *Dendrothrix* (Euphorbiaceae, tribe Hippomaneae): a Guiana Shield element in sub-Andean cordilleras of Ecuador and Peru

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Abstract

Dendrothrix condorensis K.Wurdack, **sp. nov.** from the sub-Andean cordilleras of Ecuador and Peru is described and illustrated. The new species is geographically widely separated from its likely closest relative, *D. yutajensis*, which is endemic to the Guiana Shield region of southern Venezuela and adjacent Brazil, and notably differs in leaf morphology. Vegetative (i.e., epidermal micropapillae, trichomes) and reproductive (i.e., cymule glands, flowers, pollen) micromorphological features were examined with SEM. Rare tristaminate flowers were documented among the typical bistaminate ones. Seeds and diagnostic features among the four species of *Dendrothrix* are compared.

Keywords

Cordillera del Cóndor, *Dendrothrix*, Hippomaneae, leaf morphology, seeds

Introduction

One of the most challenging groups of Euphorbiaceae with regard to identification and classification is the hippomanoid clade of subfamily Euphorbioideae. The clade presently includes 32–40 genera and 300 species grouped in three tribes (Hippomaneae, Hureae, and Pachystromateae; Esser 2001, 2012) or a single large one (Hippomaneae s.l., Webster 2014). Their taxonomy still needs considerable revision, especially at the

generic level, in light of molecular phylogenetic results that reveal multiple non-monophyletic groups (Wurdack et al. 2005; Wurdack, unpublished). *Dendrothrix* Esser is one of the many small genera of hippomanoids (19 of the 40 genera contain three or fewer species) that have been relatively recently recognized, and one whose systematics had been confused with other taxa in the clade including *Sapium* Jacq. and *Senefelderopsis* Steyerl. (Esser 1993, 1995). The shared similarities with *Sapium* include their bistaminate flowers with a partly fused calyx, and with *Senefelderopsis* their shared inflorescence structure and Guiana Shield distribution.

Dendrothrix contains four species of trees and shrubs with white latex, tiny apetalous flowers, and an unusual distribution in northern South America that includes tepuis in the Guiana Shield region and two disjunct outliers (Fig. 1). The genus is distinguished within the hippomanoids by a combination of relatively rare character states including compound thyrses inflorescences, dendritic trichomes, large disc- or cup-shaped bract glands, staminate flowers with two connate stamens, and small, dry fruits. Since Esser's original studies (1993, 1995) little new information has been published about *Dendrothrix* except for wood anatomy (Mennega 2005). The relationships of *Dendrothrix* were thought to lie with *Senefelderopsis*, and although the former is a distinctive group, Webster (2014) suggested that it might be reduced to a section of *Senefelderopsis*. Recent work on its molecular phylogenetic placement (Wurdack, unpublished) has shown that they are not sister groups and should not be merged. Moreover, a close relationship with *Mabea* Aubl., which had been suggested based on the shared multicellular dendritic trichomes from which *Dendrothrix* gets its generic name (Esser 1993), was also not supported.

Dendrothrix yutajensis (Jabl.) Esser was reported for the flora of Ecuador (Neill and Ulloa Ulloa 2011), which represented a considerable range extension. After further study of *Dendrothrix* collections from sub-Andean cordilleras of southern Ecuador and adjacent Peru, I have concluded that they are morphologically distinctive enough to deserve recognition as the new species described herein. These mountains along the wet northeastern flank of the Andes, also referred to as "Andean tepuis," are rich in endemic taxa, especially in the Cordillera del Cóndor (Neill 2005). Floristic inventory work begun there in the 1990s has yielded numerous new taxa in diverse families. For the Cordillera del Cóndor in particular, its location, climate, and unusual geology of mid-Cretaceous Hollin Formation sandstone in tepui-like mesas support biogeographic affinities with the Guiana Shield biota to the east on pre-Cambrian sandstones, e.g., *Phainantha* Gleason, Melastomataceae (Ulloa Ulloa and Neill 2006); *Stenopadus* S. F. Blake, Asteraceae (Pruski 1998). The new *Dendrothrix* adds the first member of Euphorbiaceae to the growing list of floristic disjunctions between the Andean tepuis and their counterparts in the Guiana Shield. The Euphorbiaceae diversity for the Cordillera del Cóndor region otherwise mostly appears typical of that seen in other montane Andean floras and is not especially rich, with less than 20 taxa. However, among those taxa are the unusual endemic *Croton condorensis* Riina & Cerón (Riina et al. 2014), and the more widely distributed new genus *Incadendron* (Wurdack and Farfan-Rios 2017).

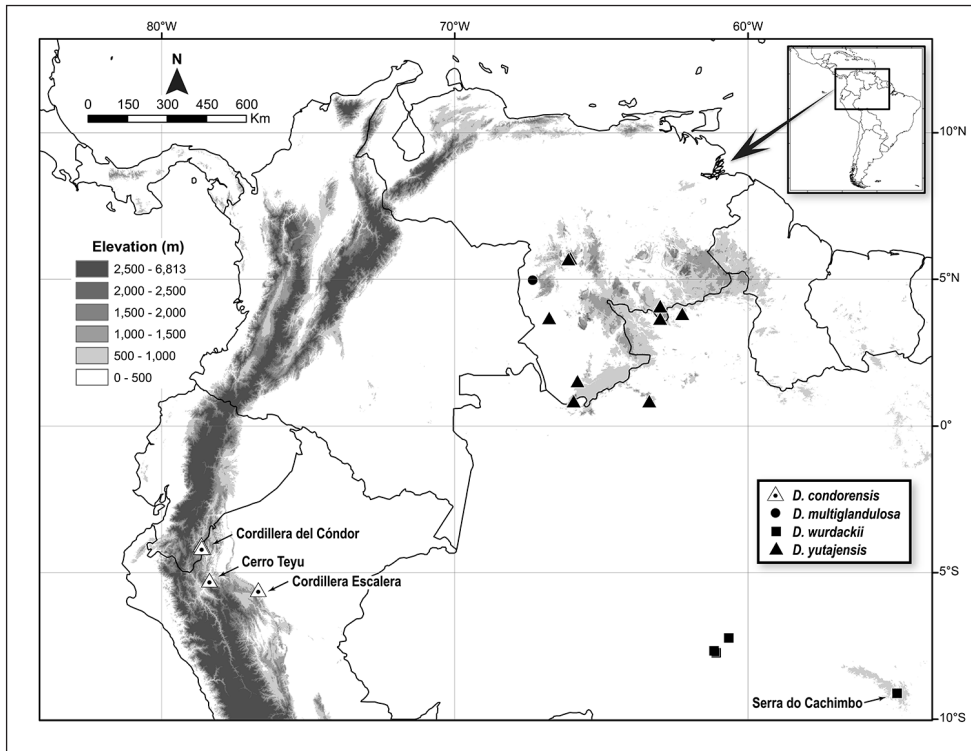


Figure 1. Distribution map of *Dendrothrix* in northern South America.

Taxonomic treatment

Dendrothrix condorensis K.Wurdack, sp. nov.

urn:lsid:ipni.org:names:77165931-1

Figure 2

Diagnosis. Differs from *Dendrothrix yutajensis* in larger, thinner leaves with more secondary veins, percurrent tertiary venation, acuminate leaf apex, and seeds lacking a caruncle.

Type. ECUADOR. Zamora-Chinchi: Nangaritza. Cordillera del Cóndor region, upper Río Nangaritza valley, along road 14.5 km south of Guayzimi, above west bank of Río Nangaritza, sandstone slope with relatively low forest, canopy 20 m, forest being cleared for mining of silica sand for glass manufacture, 04°02'27"S, 78°38'44"W, 930 m, 25 Nov 2005 (fl, fr), *D. Neill & W. Quizhpe* 14939 (holotype QCNE; isotypes: AAU, GB, HUT, K, LOJA, MO-6106711, NY, US).

Description. *Shrub* or small tree to 8 m tall, monoecious; stems of leafy branchlets 2–3 mm dia., bark smooth, pith soft. *Exudate* present, white latex. *Indumentum* of multicellular trichomes to 0.1 mm long, ramified, dark reddish-brown. *Leaves* alternate, petiolate, stipulate, simple. *Stipules* free, paired, scale-like, 0.5 × 0.3 mm (width at base), triangular, persistent, eglandular. *Petioles* 40–65 × 1–2.5 mm (dia. mid-length), terete, adaxially slightly canaliculate towards distal end, petiolar glands absent. *Leaf*

blades: laminar size class mesophyll, blade 14.5–19.8 × 6.5–8 cm, length:width ratio 2.00–2.98 (mean = 2.63, $n = 11$), symmetrical, shape elliptic, apex angle acute, apex shape short acuminate to attenuate, base angle acute, base shape cuneate to obtuse; margin entire, slightly revolute; blades sparsely pubescent when mature (young leaf not seen), primary and secondary veins more densely so; lamina thin and brittle when dry, adaxial surface smooth except for prominulous venation, abaxial surface pale, dull due to minute papillae; basilaminar glands present, abaxial, one on each side of primary the vein at the attachment of petiole and hidden under minutely auricled extension of lamina, narrowly elliptic, 1.4–1.8 × 0.4 mm, shallowly sunken into laminar surface and without raised edge, gland surface smooth; embedded laminar gland-like structures (perhaps necrotic, see Discussion), 0–6 per leaf, abaxial, scattered but usually adjacent to secondary or tertiary veins in distal portion of lamina, circular to widely elliptic, 0.5 × 0.3–0.4 mm. *Venation* pinnate, brochidodromous, 18–22 secondaries per side, spacing mostly regular, secondary vein angle gradually increasing along the series from ca 0° for proximal secondaries to 30° for distally diverging veins, course usually straight until curving for distal <1/4 of length, insertion shortly decurrent; intersecondaries parallel to secondaries, infrequent (1–5 per side); intercostal tertiaries alternate percurrent (occasionally opposite); primary to tertiary veins slightly prominulous on both surfaces. *Inflorescence* terminal, erect, compound thyrses, 21–31 cm long, with 2–3 orders of branching, axes moderately pubescent; leaves at base of inflorescence often of reduced size; lateral branches usually bisexual (rarely staminate) with pistillate flowers proximal and staminate distal, branch nodes subtended by usually eglandular bract (rarely at more distal nodes with pair of glands similar to those on cymules); bract acuminate, 2–2.5 × 0.5–0.7 mm (width at base). *Staminate cymules* spirally arranged, 20–22 per lateral inflorescence branch, subtended by bract and pair (1 on each side of bract base) of elongate disc- or cup-shaped glands; bract to 1 (deep) × 2 mm (wide), elliptic, margin sparsely ciliate; glands of proximal cymules 1.2–2 × 0.8–1.5 mm, becoming smaller at distal cymules (unclear if the size decrease is due to serial reduction or decreasing maturity), fleshy, 0.1 mm thick when dried, surface smooth and without pores; bracteoles absent. *Staminate flower* buds 10–16 per cymule, tightly clustered, erect when emergent from subtending bract, globose, to 0.7 mm diameter just before opening; anthetic flowers articulate at base, shortly pedicellate on persistent pedicel to 0.5 mm long; sepals 2 (rarely irregularly 3), connate at base to 0.3 mm, distal lobes 0.4–0.6 × ca 0.5 mm, margin sparsely ciliate; stamens 2(–3), 1–1.5 mm long, barely protruding beyond calyx at anthesis; filaments connate, 0.8–1 × 0.1 mm; anthers 0.4–0.5 mm long, bithecate, apicifixed to subapicifixed with very short connective and pendulous thecae, longitudinally dehiscent via slit 1/2–2/3 length of thecae; pistillode absent; flowers yellow in life. *Pistillate flowers* solitary at 1–2 proximal nodes of lateral thyrses branches; subtending bract 2–2.5 × ca 1.5 mm (width at base), acuminate; bract glands present (distal flower) or reduced to absent (proximal flower), similar to those subtending staminate cymules; short pedicellate, pedicel 0.5–0.8 × 0.5–0.6 mm; flower 3.5–4 mm long; sepals 3, 2–2.5 × 1 mm, free to minutely connate at base (to 0.2 mm), cymbiform, narrowly acute tip, sparsely pubescent, margin sparsely ciliate;

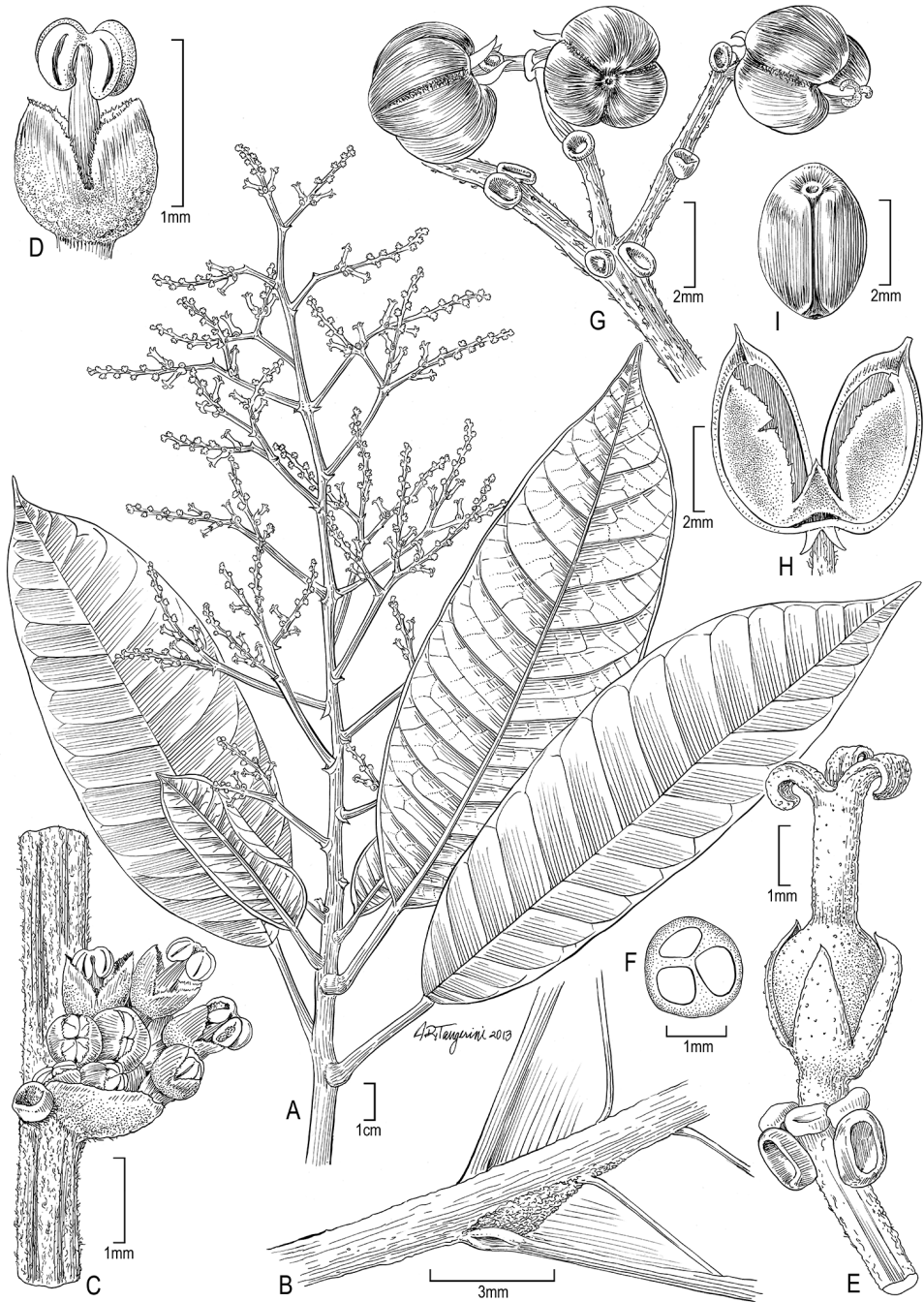


Figure 2. Illustration of *Dendrothrix condorensis*. **A** Habit **B** Leaf base (abaxial) showing basilar glands **C** Staminate cymule **D** Staminate flower **E** Pistillate flower **F** Cross section of ovary **G** Portion of infructescence **H** Mericarp valve **I** Seed. (Source: **A** Neill *et al.* 15747, MO and using life photos of this collection; **B, E–F** van der Werff *et al.* 16331, MO; **C–D, G** Neill & Quizhpe 14939, MO; **H–I** Neill *et al.* 15750, MO).

ovary 3-locular, ovoid, 1×1 mm, top tapering, densely pubescent, distinguished from styles by change in pubescence density; styles connate into trigonous column $1.5\text{--}2 \times 0.5\text{--}0.7$ mm, sparsely pubescent; stigmas 3, undivided, slightly flattened, $0.7\text{--}1.1$ (long) $\times 0.3$ (thick) $\times 0.4$ mm (wide at base), recurved to coiled at anthesis, surface coarsely papillose; placentation apical pendulous with a single ovule per locule; staminodes absent. *Infructescence* consisting of primary axis with lateral fruiting nodes, distal staminate portions of lateral branches deciduous; fruit pedicels $1\text{--}2(3) \times 0.5\text{--}0.7$ mm; bracts persistent. *Fruit* subglobose, 5×7 mm, sparsely pubescent, apex trilobed due to sunken styler region, ventral (septal) sutures sulcate, dorsal (loculicidal) sutures with slight ridge; mericarps equal, 2-valved, splitting septically then loculicidally to release seeds; sepals, styles, and stigmas persistent. *Pericarp* dry, woody, mericarp wall 0.3 mm thick (equatorial at dorsal suture); exocarp extremely thin (ca 0.05 mm) but locally thickened to 0.2 mm along ventral suture, adherent to mesocarp on dehiscence; mesocarp woody, in equatorial section varying from 0.3 mm thick at dorsal suture to 0.7 mm toward ventral suture; septa woody, nearly continuous except for distal semicircular gap where traversed by funicle, $0.7\text{--}1$ (wide) $\times 0.3\text{--}0.4$ mm (deep); mericarp valves (cocci) remaining attached together after dehiscence via basal triangle $1 \times 1\text{--}1.3$ mm (width at base), slightly twisted when dehiscent; septa of mericarps with one bifurcate vascular strand; funicle short, stout, 0.3×0.3 mm; columella (carpopore) persistent, $5 \times 0.45\text{--}0.5$ mm (width at narrowest point in middle), dilating to $1\text{--}1.5$ mm at both tip and base, trigonous, narrowly alate. *Seeds* 3 per fruit, dry, ellipsoid, 4 (long) $\times 2.7$ (wide; lateral-lateral) $\times 2$ mm (deep; raphe-antiraphe); apex with short beak, flattened or depressed around hilar zone, ventral face with shallow groove along which raphe runs as ca. 0.1 mm wide prominulous line; testa dry, smooth, uniformly dark brown, thin (ca 0.05 mm thick); caruncle absent; embryo not seen.

Etymology. The specific epithet refers to the Cordillera del Cóndor, where the type was collected. The mountain range name in turn comes from “condor” based on “kuntur” (Quechua) and refers to the Andean condor (*Vultur gryphus* L.), an important part of the ecology and culture of the Andes.

Distribution and ecology. The new species mostly occurs at 800–1000 m in dense, low, wet forest and sclerophyllous scrub over nutrient poor, acidic, sandstone-derived soils. Such habitats resemble those in the Guiana Highlands occupied by *D. yutajensis*. The three well-separated localities (Cordillera del Cóndor, Cordillera Escalera, Cerro Teyu; Fig. 1) are discoveries due to recent exploration in remote sub-Andean cordilleras, and continued floristic work is likely to extend its range to similar habitats further south in Peru. Floristic affinities between the Cordillera del Cóndor and Cordillera Escalera are notable (Neill et al. 2014). Flowers and fruits were collected during September–November and flowers in March (also fruits in July fide Croat 91402, not seen). *Dendrothrix yutajensis* with more collections to finely document phenology, is reproductive from November to May and appears to flower and fruit continuously during this period.

Conservation status. Following the criteria and categories of IUCN (2012), *D. condorensis* is given a preliminary status of Vulnerable (VU) under geographic range criteria B2 area of occupancy <2000 km² (B2a, known to exist at no more than 10

locations; B2b, continuing decline projected). Threats to this taxon in the Cordillera del Cóndor include mining for the underlying silica sand. The Cordillera Escalera is protected as a conservation area.

Additional collections. ECUADOR. Zamora-Chinchipec: Nangaritza, Cordillera del Cóndor region, west side of upper Río Nangaritza, along road about 13 km south of Guayzimi, silica mine “La Daniela”, dense wet forest on sloping Hollín sandstone plateau, being mined for silica sand for glass manufacture, 04°08'35"S, 78°35'45"W, 970 m, 15 Sep 2007 (fl.), *D. Neill, C. Davidson, S. Christoph & W. Quizhpe 15747* (AAU, LOJA, MO, NY, QCNE). [Same locality], 15 Sep 2007 (fr.), *D. Neill, C. Davidson, S. Christoph & W. Quizhpe 15750* (AAU, GB, LOJA, MO, NY, QCNE, US). Along road from near Paquisha south to Las Orchídeas, and end of river at Río Nangaritza, via Guayzimi, beginning at 15.9 km E of Zumbi and Río Zamora, then 37.3 km S of junction, 12.3 km N of Las Orchídeas, 04°08'25"S, 78°38'31"W (-4.1402700, -78.6419400), 886 m, 17 July 2004 (fr.), *T. Croat, L. Hannon, G. Walbert & T. Katan 91402* (MO; not seen but tentatively included here based on TROPICOS record). **PERU. Amazonas:** Bagua District, upper slopes and summit of Cerro Teyu, summit with sclerophyll scrub, 05°15'56"S, 78°22'07"W, 1030 m, 22 Mar 2001 (fl.), *H. van der Werff, R. Vasquez & B. Gray 16331* (MO, US). **Loreto:** Alto Cahuapanas, Campsite #3 (“Alto Cahuapanas”) on Rapid Biological and Social Inventory #26, -5.66438889, -76.839, 1000–1350 m, 28 Sep 2013, *M. Ríos Paredes 3480* (F). [Same locality], 29 Sep 2013, *M. Ríos Paredes 3517* (F).

Discussion. The four species of *Dendrothrix* are morphologically similar, and major differences are presented in Table 1. *Dendrothrix condorensis* and *D. yutajensis* are likely closely related as they have nearly identical floral features. However, they are easily distinguished by foliar characters (i.e., size, shape, thickness, and details of leaf architecture), seed caruncle differences, and biogeography. Although leaf architecture has not been rigorously compared through clearings and anatomy, several orders of the varyingly prominulous venation are evident in unprepared specimens that allow gross comparisons such as were shown by Esser (1993) to be informative for the group. In *D. condorensis*, secondaries are 18–22 pairs, mostly straight with few course deflections, and distally curving upward for $<1/4$ of course; intersecondaries are rare, and tertiaries are percurrent (Fig. 3G). In *D. yutajensis*, secondaries that are fewer (usually 7–9 pairs), not straight due to a slight zigzag (fractiflexuous) appearance where tertiaries diverge, and distally curving upward for half of their course; intersecondaries are frequent, and tertiaries are reticulate (Fig. 3J). Even on unusually robust specimens of *D. yutajensis* (e.g., *Maguire & Maguire 35103*, US), the largest leaves (16.5–18 × 6.5–7.8 cm) have only up to 11 pairs of secondaries. The leaves of *D. condorensis* more closely resemble those of *D. multiglandulosa* Esser with which it shares the generally larger blades and percurrent venation. *Dendrothrix multiglandulosa* is only known from two collections from the Cuao-Sipapo massif in Venezuela and potential variation is poorly understood. *Dendrothrix yutajensis* is the most frequently collected and wide-ranging species of *Dendrothrix*, known from many well-separated tepuis, but intraspecific variation appears low. *Dendrothrix wurdackii* Esser has previously unrecognized intraspecific variation with an odd collection (i.e., BRAZIL, Pará: km 872 Cuiabá-Santarém [Highway],

Table 1. Diagnostic features to distinguish among the species of *Dendrothrix*.

Character	<i>D. condorensis</i>	<i>D. yutaensis</i>	<i>D. multi glandulosa</i>	<i>D. wurdackii</i>
Leaf features	Blade elliptic, 14.5–19.8 × 6.5–8 cm, apex acuminate to attenuate, base cuneate to obtuse; secondary veins 18–22 pairs, straight; tertiary veins mostly alternate (occasionally opposite) percurrent; texture thin and brittle	Blade elliptic, 7–12.5(18) × 3.5–6.5(7.8), apex acute to subobtusely base cuneate to obtuse; secondary veins 7–9 (11) pairs, flexuous; tertiary veins reticulate; texture coriaceous	Blade elliptic to obovate, 13.5–16.5 × 5.5–9 cm, apex acute to obtuse, base cuneate to obtuse; secondary veins 9–14 pairs, straight; tertiary veins mostly alternate (rarely opposite) percurrent; texture coriaceous	Blade ovate, 8–10 × 6–7 cm, apex acute to acuminate, base subcordate to rounded; secondary veins 5–9 pairs, including prominent basal pair of major secondaries such that the leaf appears triplinerved, straight; tertiary veins reticulate; texture coriaceous
Basilaminar glands	Abaxial, hidden under leaf extension	Abaxial, hidden under leaf extension	Abaxial, hidden under leaf extension	At leaf margin and sometimes nearly adaxial (never hidden by laminar extension)
Pubescence	Reddish brown	Reddish brown	Pale, whitish	Reddish brown
Staminate cymule bract glands	1 per side	1(2) per side	(1)2–3(4) per side	1 per side
Pistillate sepals	Free to minutely connate at base (to 0.2 mm), lobes narrowly acute; margin entire, sparsely ciliate	Free to minutely connate at base (to 0.1 mm), lobes narrowly acute; margin entire, pubescent/ciliate	Distinctly connate at base (to 0.8 mm), lobes rounded to broadly acute; margin entire, ciliate	Distinctly connate at base (to 0.5 mm), lobes rounded to broadly acute; margin erose or irregularly minutely toothed, very sparsely ciliate
Pistil	Style 1.5–2 × 0.5–0.7 mm; stigma branches long (to 1.1 mm), thin, recurved/coiled	Style 1.5(–2) × 0.5–0.7; stigma branches long (to ca 1 mm), thin, recurved/coiled	Style 1–1.3 × 0.5–0.7 mm; stigma branches short (to 0.5 mm), spreading but not recurved	Style 1–1.5 × 0.5–0.6 mm; stigma branches long (to ca 1 mm), thin, recurved/coiled
Caruncle	Absent (Neill 15750, MO; Neill & Quizhpe 14939, MO, although more immature)	Present, small (Maguire 30694, NY; Neill 31120, NY, US; Amaral 1523, MO)	Absent (Maguire & Poltri 27683, NY)	Present, large (Calderón <i>et al.</i> 2682, US; Esser 1993)
Distribution	Ecuador (Zamora-Chinipe), Peru (Amazonas, Loreto)	Brazil (Amazonas), Venezuela (Bolívar, Amazonas)	Venezuela (Amazonas)	Brazil (Amazonas, Pará)

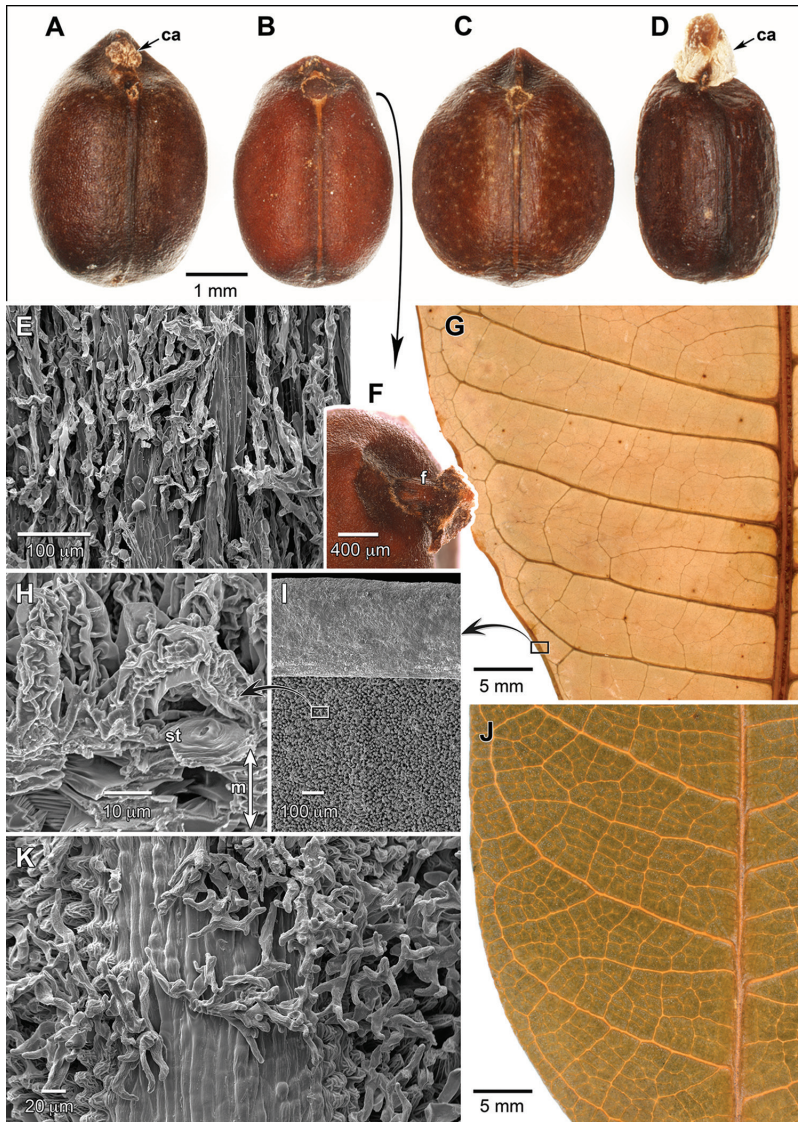


Figure 3. Seeds and surfaces of *Dendrothrix*. **A–D** Ventral views of seeds (ca = caruncle) **A** *D. yutajensis* **B** *D. condorensis* **C** *D. multiglandulosa* **D** *D. wurdackii* **E** *D. condorensis*, inflorescence axis showing dendritic trichomes **F** *D. condorensis*, top of seed showing short funicle (f) and caruncle absence **G** Abaxial view of *D. condorensis* leaf **H** *D. condorensis* transverse fractured leaf showing abaxial papillae and stomata (st), above intercellular space in the spongy parenchyma (m = mesophyll) **I** *D. condorensis* abaxial leaf micropapillae contrasting with smooth revolute adaxial leaf margin at image top **J** Adaxial view of *D. yutajensis* leaf **K** *D. wurdackii*, abaxial leaf surface along secondary vein showing gradient of micropapillae development at vein edges, and overlying dendritic trichomes. (**A–D**, **F–G**, **J** imaged with a Olympus DSX100 **E**, **H–I**, **K** imaged with a Zeiss EVO MA15 SEM at 10–12 kV after sputter coating with 25 nm of Au/Pd; SEM samples untreated and directly mounted from dried herbarium specimens. Source: **A** *Nee 31120*, NY; **B**, **F** *Neill et al. 15750*, MO; **C** *Maguire & Politi 27683*, NY; **D** *Calderón et al. 2682*, US; **E**, **G–I** *Neill & Quizhpe 14939*, MO; **J** *Amaral 1523*, MO; **K** *Cid Ferreira 5797*, NY).

Serra do Cachimbo, forest beside small stream, 6 Nov 1977, *G. Prance* 24947, MO, NY, US), having much larger (to 18×8.5 cm), long acuminate, glabrous leaves with poorly developed abaxial micropapillae as compared with the four other collections of the species that closely resemble the type. This collection, which Grady Webster annotated as a new species of *Senefeldera* Mart., clearly has closest affinities with *D. wurdackii* based on similarities in leaf shape and venation, basilaminar glands, pistillate sepal shape, and biogeography. While the morphological differentiation and slight disjunction (Fig. 1) may be significant, it is not to the degree used here to justify the recognition of *D. condorensis* and I have refrained from further taxonomic adjustments until *D. wurdackii* is better understood.

Leaf micromorphological features are similar among the taxa of *Dendrothrix*. The distinctive, loosely attached, branched trichomes (Fig. 3E, K) are structurally very similar but can differ in pigmentation (rusty brown versus whitish; see Table 1). Epidermal microrelief such as micropapillae or striations that give an often light-colored, matte sheen to abaxial leaf surfaces has evolved in many genera of Euphorbiaceae, including *Dendrothrix*, and other hippomanoids (e.g., *Gymnanthes hypoleuca* Benth., *Sebastiania larensis* Croizat & Tamayo, *Senefelderopsis croizatii* Steyererm.). The microrelief likely has functional significance related to optimized stomatal conductance and reduced leaf wetting (Neinhuis and Barthlott 1997). The morphology of the leaf micropapillae in *Dendrothrix* is unusual and may be synapomorphic. They have positional size variation in being short to absent on major veins and grading to longer in intercostal regions (Fig. 3K). In *D. condorensis*, the micropapillae are finger-like, 25–30 μm tall, ornamented with ridges, and form a protective canopy that obscures the stomata (Fig. 3H–I). Epicuticular waxes are not well developed, in keeping with the non-glaucous appearance. The stomata are only abaxial, with the adaxial surface being relatively featureless (Fig. 3I). Glands are often present on leaves and/or inflorescences of Euphorbiaceae, and *Dendrothrix* is no exception where they occur in pairs at the base of the leaf and subtending the bracts. The nature of scattered laminar glands in *Dendrothrix* is partly unclear. In the case of *D. wurdackii* they resemble typical Euphorbiaceae laminar glands with a well defined raised border and an abaxial distribution adjacent to secondary veins towards the leaf margin. The glands on *Prance* 24947 contain drops of clear exudate that are still sticky after 40 years. I have not found clearly homologous structures on the other taxa of *Dendrothrix*. Circular to widely elliptic, small (<0.5 mm diameter), gland-like structures with smooth surfaces and without raised edges are sparsely present in *D. condorensis* and more abundantly so in *D. multiglandulosa*, where in the latter they were considered of systematic significance (Esser 1993). Although not examined anatomically, these appear to be wound response to microbial damage rather than typical glands as evidenced by their absence in the few young leaves available, erratic distribution, the presence of apparently intermediate epidermal blistering stages that are clearly necrotic, and margins that are not clean under SEM (not shown) but rather contain fragments that suggest micropapillae residue from surficial degradation.

Among reproductive characters that are variable among the species are details of the pistillate flowers including sepal and stigma morphology (see Table 1). Staminate

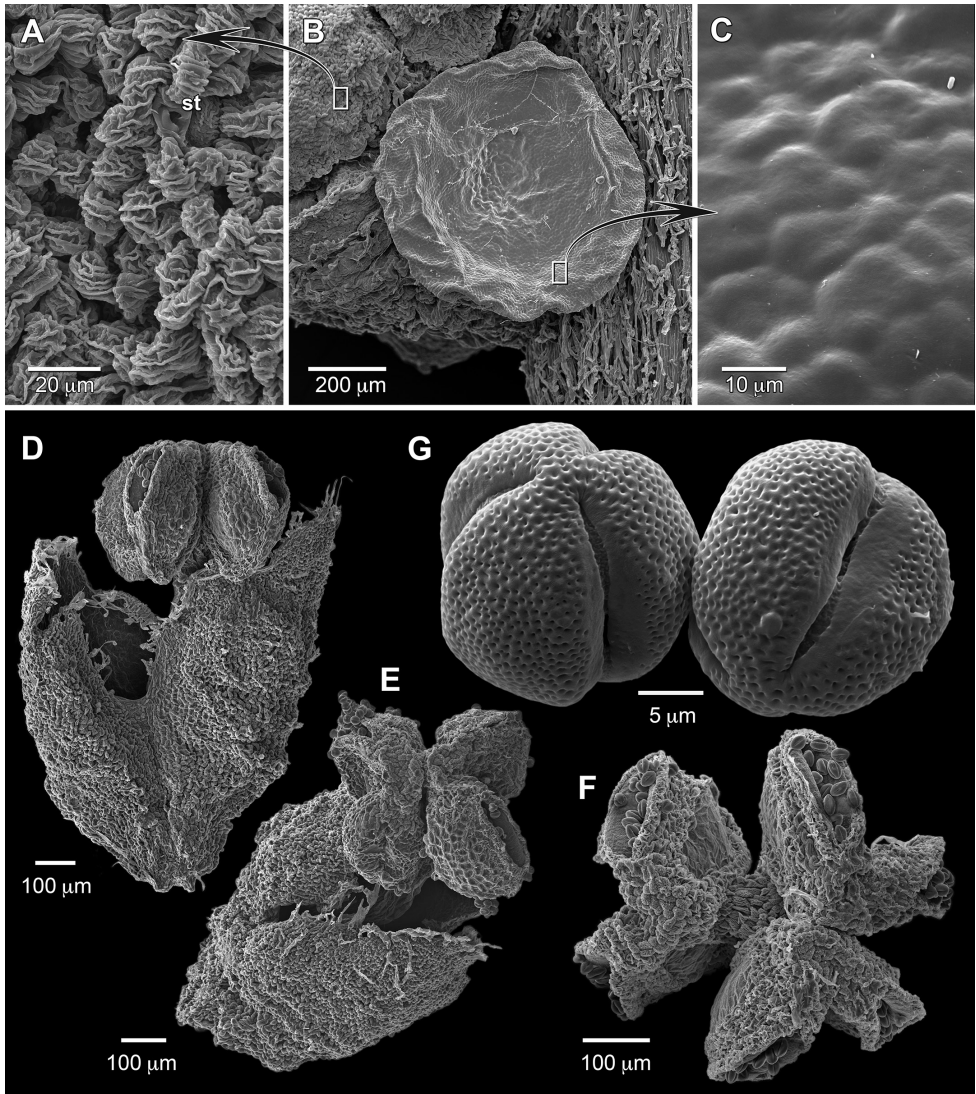


Figure 4. Floral morphology of *Dendrothrix condorensis*. **A** Staminate sepal outer surface showing papillae and stomata (st) **B** Staminate cymule base showing subtending discoid gland **C** Surface of staminate cymule gland **D** Staminate flower **E** Staminate flower **F** Triandrous flower, top view of slightly asymmetrically fused androecium **G** Pollen. (Source: SEM of Neill & Quizhpe 14939, MO).

bract glands vary in number with *Dendrothrix multiglandulosa* usually having 2–3 pairs (variable from 1–4, and paired or unequal in number between the bract sides), while the other species have one pair (*D. yutajensis* rarely has a second gland on one side, e.g., Maguire & Maguire 35103, NY). Inflorescence branching is potentially variable, with 2–3 orders for *D. condorensis* and *D. multiglandulosa*, and the other species have only two orders. It is unclear if this represents noteworthy interspecific variation or possible

collector bias for smaller inflorescences that better fit herbarium sheets. *Dendrothrix* is characterized by staminate flowers with two connate anthers (Fig. 4D–E). Rare flowers with three anthers are present on collections of *D. condorensis* (i.e., Neill & Quizhpe 14939, MO) and *D. yutajensis* (i.e., Maguire & Maguire 35103, NY). Such flowers have more or less symmetrical androecia, connate filament columns, partial third sepals, and developed pollen, which indicates they may be functional despite being teratological in gross structure (Fig. 4F). The triandrous flowers may be of significance in reflecting the likely plesiomorphic condition for the tribe and the ease of merosity change even in groups that appear fixed. The bract glands are smooth and without pores (Fig. 4B–C). The staminate sepals bear stomata and are externally minutely papillose (Fig. 4A). The pollen of *Dendrothrix* (Fig. 4G; Esser 1994) is tricolporate with a perforate exine, which closely resembles that of other Hippomaneae (see Park and Lee 2013). Seed variation occurs among the taxa of *Dendrothrix* (Fig. 3A–D) with a wider, more globose seed in *D. multiglandulosa*, and a well-developed caruncle in *D. wurdackii*. In *D. yutajensis* the caruncle is small, and there is no caruncle in *D. condorensis* or *D. multiglandulosa*. The 13 available seeds of *D. condorensis* from dehiscent fruits (some slightly immature; note lighter brown coat in Fig. 3B relative to other samples) do not show evidence of caruncle growth (Fig. 3F), while all seeds that I examined of *D. yutajensis* have a small caruncle.

Dendrothrix has a noteworthy Guiana Shield disjunct distribution (Fig. 1), which now combines two patterns with its over 1000 km distant outliers including elements in both the Andean (i.e., *D. condorensis*) and Amazon (i.e., *D. wurdackii*) phytogeographic regions. Such a tripartite distribution does not appear to be shared with other Guiana Shield near-endemic plant genera, although many have disjunct species in just one of those regions (Berry and Riina 2005). *Raveniopsis* Gleason (Rutaceae) also has a distribution of Guiana Shield endemics and two Brazilian Amazon lowland outliers, and interestingly, one of the few localities of *D. wurdackii* (Calderón *et al.* 2682, NY) is also the type locality for one of those outliers, *Raveniopsis campinicola* Kallunki (Calderón *et al.* 2722, US).

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