

The fern genus *Adenoderris* (family incertae sedis) is artificial

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Abstract For over a century the relationships of the rare fern genus *Adenoderris* J. Sm. have been confused. Here, we (1) present a molecular analysis of the genus based on multiple chloroplast markers with the goal of placing its species phylogenetically, (2) provide insights into its morphology and complex taxonomy, and (3) make relevant nomenclatural changes. In seeking a resolution of the problems with *Adenoderris*, we investigated the morphological, historical, ecological, and biogeographical factors that have made *Adenoderris* so difficult to place with certainty. The key findings are that (1) *Adenoderris* comprises two species that were included in the genus on the basis of convergent and symplesiomorphic morphological features, and (2) though the two species both lie in the Dryopteridaceae, they belong to different genera. The correct names for these two species are *Polystichum glandulosum* C. Presl and *Dryopteris sororia* (Maxon) M. McHenry, Sundue & Barrington, comb. nov.

Keywords Dryopteridaceae; morphology; taxonomy

Supplementary Material The alignment is available in the Supplementary Data section of the online version of this article (<http://ingentaconnect.com/content/iapt.tax>).

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■ INTRODUCTION

Daunting problems remain in the circumscription of genera near *Dryopteris* Adans. and *Polystichum* Roth in the diverse fern family Dryopteridaceae. For instance, among the genera tentatively recognized by Smith & al. (2006), several Old World genera including *Leptorumohra* (H. Itô) H. Itô in Nakai & Honda and *Phanerophlebiopsis* Ching appear to be artificial based on recent molecular analyses (e.g., Liu & al., 2007). In the New World, the genus *Adenoderris* J. Sm., also recognized by Smith & al. (2006) as a member of the Dryopteridaceae, remains an enigma, because of its controversial classification history and extraordinary rarity.

In 1875 John Smith established *Adenoderris* J. Sm., based on a fern species from Jamaica and Cuba originally described in 1831 as *Aspidium glandulosum* Hook. & Grev. Smith reported the species to be close to *Polystichum*, but found its habit to be different enough to merit generic recognition. Maxon (1905) broadened the generic concept to accommodate a newly discovered Guatemalan species, *Adenoderris sororia* Maxon. Maxon characterized the genus as comprising “small plants of lax habit, distinct from *Polystichum* by their herbaceous texture, aspinulose margins, and dense glandular-pilose covering.” Christ, in a letter to Maxon from 1905 (attached to US 831633), wrote that describing the specimens in question as a new genus was premature because the plants looked like a diminutive of a normally larger plant. In closing the note he exhorted Maxon to make further investigations. Still, it was Maxon’s short but authoritative 1905 publication that has led to the continued recognition of *Adenoderris* as a distinct genus.

The generic placement of *Adenoderris* has been a problem for almost 200 years. Presl (1836) in establishing *Polystichum* as a genus in the modern sense, included *Polystichum glandulosum* C. Presl, the future type species of *Adenoderris*, but with a query after the genus name, thus doubting its placement. The middle of the 19th century saw the species settled in alliance with *Polystichum*. Mettenius (1858) coined a nomen novum, *Aspidium viscidulum* Mett., for *Aspidium glandulosum* and placed the species between two diminutive West Indian species that pertain to *Polystichum* s.str. in the modern sense (e.g., sensu Li & al., 2008). Similarly, Hooker (1862) placed the species at the beginning of his broadly defined *Aspidium*, followed by species placed in *Polystichum* now. Smith (1875) renewed this controversy as he established the genus *Adenoderris*, mentioning both *Polystichum* and *Lastrea* Bory (i.e., *Thelypteris* Schmidel s.l.) as similar genera. Maxon (1905), in providing a synopsis of the genus *Adenoderris*, emphasized its distance from *Polystichum*, but did not provide a hypothesis as to its relationships.

Recent authors have continued to struggle with the relationships of *Adenoderris* within the higher leptosporangiate ferns (Polypodiales). The problem in the 20th century was that the genus combined characteristics of *Athyrium* Roth and allies in the Eupolypods II (sensu Rothfels & al., 2012) and *Dryopteris* and allies in the Eupolypods I (sensu Smith & al., 2006). Tryon & Tryon (1982) placed the genus in their Phymatidiaceae (i.e., close to *Athyrium*) but pointed out that the plants presented characters of both Dryopteridaceae and *Athyrium*. In contrast, Kato (1984) argued that the structure of the leaf traces, a peltate indusium, and spore morphology suggested that *Adenoderris* is close to *Polystichum*. He viewed the two vascular bundles

in the petiole (a common feature of the Eupolypods II) as an independent reduction, not an indication of affinity to the athyrioids. Tryon & Lugardon (1991) provided further insight into the two *Adenoderris* species by a close examination of their spore morphology. They found that the general spore architecture favored an alliance with the Dryopteridaceae, not the athyrioids. Moran (1995) placed the genus in his Woodsiaceae, reporting the two leaf traces as support for this placement while mentioning the similarity of the spores to those of *Polystichum* and *Dryopteris*. Mickel & Smith (2004) presented a review of the recent literature and, using Cranfill's unpublished molecular dataset as a basis, argued for maintaining its placement among the dryopteridaceous ferns such as *Polystichum*.

The two described species, as Maxon (1905) represented them, have much in common, but whether these similarities are a consequence of common ancestry has not been made clear. They are both diminutive plants with lax, thin-textured, aspinulose leaves covered with minute, gland-tipped hairs (Maxon, 1905). Maxon (1905) reported the indusia of both species to be peltate, though Smith (1875) had reported both orbicular and reniform indusia in the type species. Maxon (1905) found differences in leaf dissection, density of the minute indument, and sorus position between the two species. He reported the sori to be abaxial on veins that reach the margin in Antillean *Adenoderris glandulosa*, but terminal on veins that end before the margin in the Mesoamerican *A. sororia*. More recent work has revealed that though the spores of *Adenoderris* have a perispore pattern common in the Dryopteridaceae, the two species have strikingly different perispore ornamentation from each other (Tryon & Tryon, 1982; Tryon & Lugardon, 1991).

The geography of the two species in *Adenoderris* is consistent with floristic patterns in the literature evidencing a strong connection between the Greater Antilles and the southern Mexico-Guatemala region, treated by Takhtajan (1986) as part of a single geographic entity, the Caribbean region.

What are the affinities of the genus *Adenoderris*, and are its two species each other's closest allies? Resolution of the controversies is hindered by the rarity of the species: *A. glandulosa* has not been collected in 70 years in Cuba, and not for 80 years in Jamaica, where it is presumably extinct, given Proctor's intensive study of that island's fern flora (Proctor, 1985). *Adenoderris sororia* is known from only two collections, one the type from Guatemala (1887, *H. von Türckheim* 868, NY) and the other from Chiapas, Mexico (1981, *D.E. Breedlove* 56215, CAS-673414).

The recent considerable progress in resolving relationships among higher leptosporangiate ferns using molecular datasets (Hasebe & al., 1995; Smith & al., 2006; Schuettpelz & Pryer 2007; Rothfels & al., 2012) facilitates a more realistic assessment of *Adenoderris*'s placement. Here, we (1) present a molecular analysis of the genus with the goal of placing its species phylogenetically, (2) provide insights into its morphology and complex taxonomy, and (3) make relevant nomenclatural changes. In seeking a resolution of the problems with *Adenoderris*, we were especially interested in the morphological, historical, ecological, and biogeographical factors that have made *Adenoderris* so difficult to place with certainty.

■ MATERIALS AND METHODS

Taxon sampling. — The taxon sample (Appendix 1) consists of 110 taxa, including representatives from each family in Eupolypods I and II (sensu Smith & al., 2006 and all families but Diplaziopsidaceae sensu Rothfels & al., 2012). A preliminary analysis using the plastid marker *rbcL* run with representatives from each fern family in the Eupolypods placed *Adenoderris* in the Dryopteridaceae; the two species resolved in different genera, *Dryopteris* and *Polystichum*. Consequently, we built a denser sample of representatives from each family in Eupolypods I and II, with robust sampling of Dryopteridaceae, including at least two taxa from each major clade of *Dryopteris*, as outlined by Sessa & al. (2012), and *Polystichum*, as outlined by Driscoll & Barrington (2007). Sequences for *Polystichum* were obtained from material that was collected in the field, donated by numerous collaborators, or extracted from herbarium specimens. *Adenoderris* sequences were obtained from extractions of herbarium material for which we obtained permission to sample destructively. All other sequences were downloaded from GenBank.

DNA extraction, amplification, and sequencing. — Total genomic DNA was extracted from pinnules following a modified CTAB protocol (Doyle & Doyle, 1987). Three plastid DNA sequences were amplified using the polymerase chain reaction (PCR); the intergenic spacer between *trnL* and *trnF* (*trnLF*), and two plastid genes, *rbcL* and *rps4*. The plastid marker utilized, *matK*, consisted only of sequences downloaded from GenBank. Primers for amplification and sequencing were taken from the literature as follows: *rbcL* (Little & Barrington, 2003), *trnLF* (Taberlet & al., 1991), and *rps4* (Souza-Chies & al., 1997). In addition, four new internal primers were developed to amplify and sequence *Adenoderris* accessions for *rbcL* (reported in Table 1). The amplification of *rps4* for *Adenoderris* accessions was unsuccessful. Amplification by the polymerase chain reaction was performed in a TC-312 or TC-3000 thermal cycler (Techne, Burlington, New Jersey, U.S.A.) in 25 μ L aliquots with the following components: 150 ng of genomic template; 0.1 μ M of each primer; 1 \times ExTaq Buffer (TaKaRa, Gene Clone, Madison, Wisconsin, U.S.A.); 200 μ M/L of each dNTP; and 0.625 U Ex Taq polymerase (TaKaRa). All sequences were amplified as follows: initial denaturation at 94°C for 7 min; followed by 40 cycles (94°C for 30 s, 58°C for 1 min, 72°C for 1 min); and a final extension at 72°C for 7 min. PCR products were cleaned using ExoSAP-IT (USB Corporation, Cleveland, Ohio, U.S.A.). Sequencing of the cleaned PCR

Table 1. Primers developed for use in this study to amplify *Adenoderris* accessions for the gene *rbcL*.

Primer	5'-primer sequence-3'
MAMrbcL34F	GCTGGTGTCAAAGATTATCG
MAMrbcL635R	GATCTCTCCAACGCATGAATGG
MAMrbcL546F	AGAGCCGTCTACGAATG
MAMrbcL1200R	TTTCCCAAGGATGTCCCAAGGTTT

products employed a cycle sequence reaction using the BigDye Terminator Cycle Sequence Ready Reaction Kit v.3.1 (Perkin-Elmer/Applied Biosystems, Foster City, California, U.S.A.). Sequences were resolved on an ABI Prism 3100-Avant Genetic Analyzer (Vermont Cancer Center DNA Analysis Facility, Burlington, Vermont, U.S.A.). The raw chromatographs from each amplified PCR product were aligned using both the forward and reverse sequences; consensus sequences were assembled for each gene using Sequencher v.4.5 (Genes Code Corporation, Ann Arbor, Michigan, U.S.A.) or Geneious Pro v.5.0.3 (Drummond & al., 2007).

Sequence alignment and phylogenetic analysis. — Consensus sequences were aligned with MUSCLE (Edgar, 2004) as implemented in Geneious Pro v.5.0.3 (Drummond & al., 2007). All phylogenetically informative indels were coded following “the simple gap coding” of Simmons & Ochoterena (2000) and added as additional binary characters at the end of the NEXUS file.

The data were partitioned by plastid region, and optimal evolutionary models (Table 2) were selected for each partition using jModeltest v.2 under the Akaike information criterion (AIC; Durriba & al., 2012). The concatenated sequences were analyzed by Bayesian inference (BI) using MrBayes v.3.2 (Ronquist & al., 2012). This program ran a mixed-model Bayesian analysis allowing each region to evolve under its own best-fit model. BI was analyzed in MrBayes by running two independent analyses for 4 million generations with trees sampled every 1000 generations. Posterior probabilities (PP) were calculated at the conclusion of the Markov Chain Monte Carlo analysis in MrBayes. All trees prior to stationarity (10%) were discarded as the burn-in phase for PP calculation, then a 50% majority-rule consensus tree was calculated for the remaining trees. Stationarity was determined using the log-likelihood scores for each run plotted against generation in the program Tracer v.1.5 (Rambaut & Drummond, 2007). The program FigTree v.1.3 (Rambaut, 2008) was used to view the 50% majority-rule consensus tree with posterior probabilities.

The concatenated sequences were also analyzed by maximum parsimony (MP) analyses, using a heuristic search in TNT v.1.1 (Goloboff & al., 2008). The analysis employed 1000 independent iterations of the parsimony ratchet (Nixon, 1999), with 20 trees per iteration (up and down-weighting set to 5%), followed by 100 rounds of tree fusion with TBR-max, swapping among all most parsimonious trees to completion. Relative support for each node (bootstrap support; BS) was calculated by conducting bootstrap replicates (Felsenstein, 1985) for the

combined dataset, doing 10 ratchets per replicate, holding 20 trees per ratchet, and keeping only the strict consensus tree. MP trees were viewed in WinClada v.1.5 (Nixon, 2004).

Morphology. — Herbarium materials from GH, NY, and YU of either *Adenoderris glandulosa* from the Greater Antilles (Cuba, Jamaica) or *A. sororia* from northern Central America (Guatemala) were used to examine the variation within the genus and between the genus and its candidate allies. We applied characters that have traditionally been used along with new characters that have proven useful in recent analyses of the Eupolypods (Sundue & Rothfels, unpub. data). These characters were interpreted in the light of the molecular phylogeny.

■ RESULTS

DNA sequence and alignments. — The aligned, concatenated data matrix totaled 3504 characters of which 1996 (57%) were variable and 753 (21%) were parsimony informative.

Phylogeny. — The MP analysis recovered 1440 equally parsimonious trees 3063 steps long. The shortest trees had a consistency index (CI) of 0.32 and a retention index (RI) of 0.63. The topology of the Bayesian inference 50% majority-rule tree (Fig. 1) was congruent with the topology of the strict consensus MP tree with the following exceptions; the MP analysis failed to resolve many clades in *Dryopteris*, and the MP analysis was incongruent with the BI topology for the Eupolypod II taxa. Both analyses resolved a topology similar to but less resolved than those in previous work (e.g., Sessa & al., 2012; Zhang & al., 2012). The exception was the placement of *D. fragrans*, most likely an artifact of the smaller sample number of *Dryopteris* accessions included in this study.

The results from the BI and MP analyses placed *Adenoderris* in the Dryopteridaceae. The two *Adenoderris* species resolved within two different well-supported genera (Fig. 1); *Dryopteris* (PP = 1.0, BS = 99) and *Polystichum* (PP = 1.0, BS = 95). The sampled *A. sororia* accession was resolved within a clade of Neotropical *Dryopteris* species (Clade 1; PP = 1, BS = 99), and the sampled *A. glandulosa* accession was nested within a clade of Neotropical *Polystichum* species (Clade 2; PP = 1, BS = 100). *Adenoderris glandulosa* resolved with strong support (PP = 1, BS = 100) among polystichums reported as endemics to the Greater Antilles by Mickel (1997).

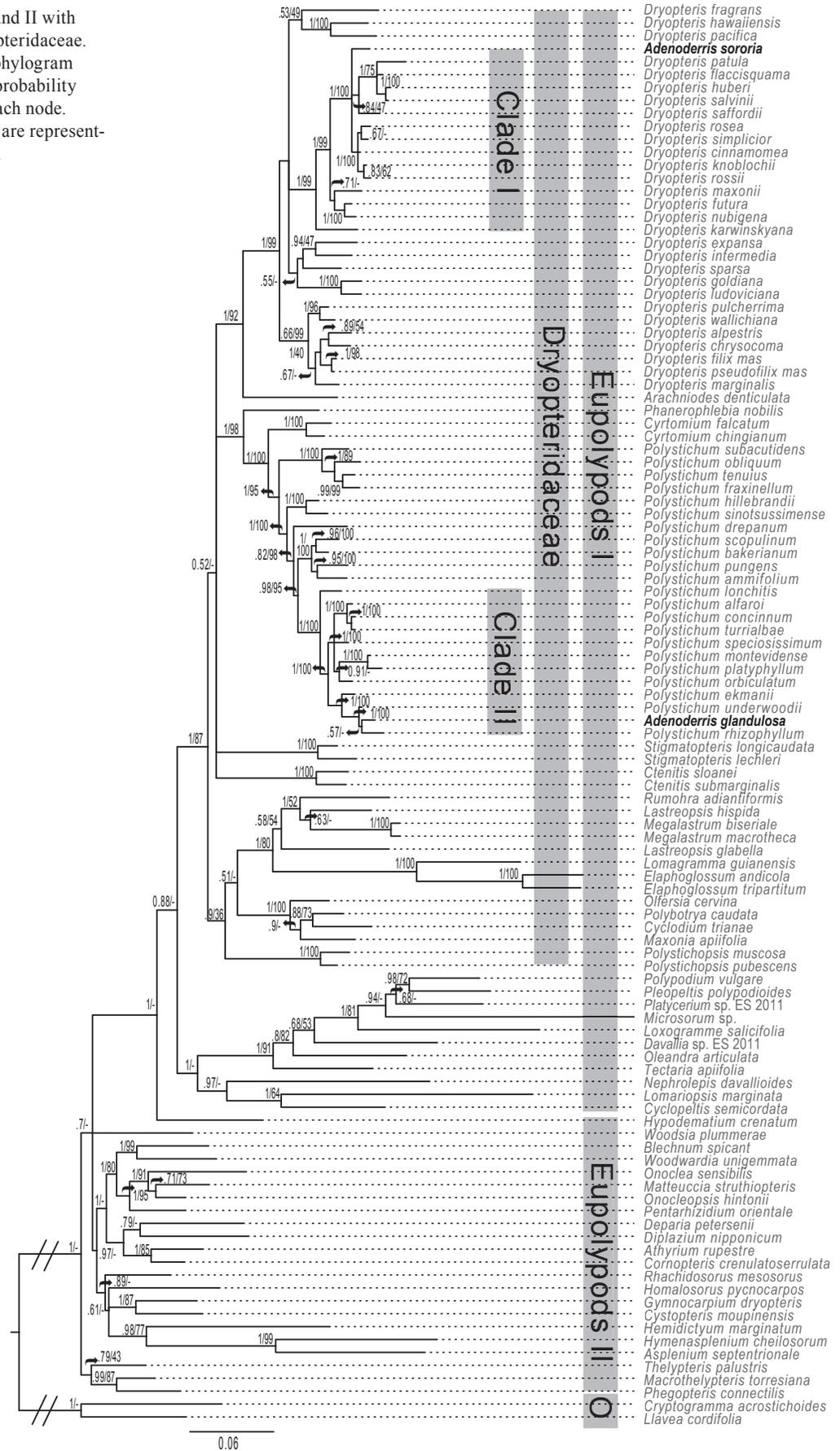
Morphology. — *Adenoderris* species are small, lax plants with short-creeping rhizomes and two leaf traces per petiole (Table 3). They have thin-textured leaves with a well-developed minute glandular-pilose indument (less developed in *A. sororia*). Spinules on the lamina edges are either absent or barely developed. The round sori are protected by a superior indusium. Spores are monolete with a prominent folded perispore. Where label data are specific, the habitat is reported to be along streams in ravines on banks or rocks at low elevations (below 1000 m).

The two *Adenoderris* species differ in a number of characters (Table 3). Whereas *A. glandulosa* has forking ultimate veins that end at the margin, *A. sororia* has simple ultimate veins that end shy of the margin. The lamina is more dissected

Table 2. Characteristics of the cpDNA markers used in the phylogenetic analysis.

Marker	Model (AIC)	Marker length	Sampled taxa
<i>rbcL</i>	SYM+G	1353	110
<i>trnLF</i>	TVM+G	366	56
<i>rps4</i>	TPM1uf+G	446	22
<i>matK</i>	TrN+G	1332	10

Fig. 1. Phylogeny of Eupolypods I and II with placement of *Adenoderris* in Dryopteridaceae. The tree is the 50% majority-rule phylogram from the BI analysis. BI posterior probability (PP)/MP bootstrap (BS) given at each node. Nodes not resolved in MP analysis are represented by -. Outgroups are labeled "O".



in *A. sororia*. Though neither has the strong spinule development of many other taxa in the Dryopteridaceae, *A. glandulosa* is weakly and intermittently spinulose whereas *A. sororia* is completely without spinules. The sori are abaxial on the veins in *A. glandulosa* but terminal on the veins in *A. sororia*; the indusia are peltate in the former and reniform in the latter. *Adenoderris* spores, based on SEM images in Tryon & Lugardon (1991), differ in that *A. glandulosa* spores have thin crests (“winglike folds”, Tryon & Lugardon, 1991) whereas those of *A. sororia* have broad folds (“inflated-saccate”, Tryon & Lugardon, 1991).

■ DISCUSSION

Relationships of the species. — The key finding of the molecular analysis is that the two *Adenoderris* species, though both allied to members of the Dryopteridaceae, are not closely related to one another. The phylogenetic distance between the species leads us to reject the idea that the biogeographic distribution of the two taxa is due to divergence from a common ancestor in Takhtajan’s Caribbean region. *Adenoderris glandulosa*’s allies are diminutive epilithic once-pinnate species of *Polystichum* from the Greater Antilles, whereas *A. sororia* belongs to a lineage of New World *Dryopteris*, a clade with taxa

rich in species limited to the northern Neotropics including the *D. patula* complex of Mickel & Smith (2004). The morphology of the plants (Table 3) is consistent with this molecular finding. *Adenoderris glandulosa*, like *Polystichum* and its immediate allies, has veins ending at the margin and a peltate indusium. Like many Greater Antillean polystichums, it has a perispore with thin crests. In contrast, *A. sororia* has veins ending shy of the margin and a reniform indusium, characters typical of *Dryopteris*. Similar to many Neotropical *Dryopteris* taxa it has a perispore with broad folds.

The temptation to place *Adenoderris* with the Athyriaceae (sensu Rothfels & al., 2012) has presumably derived from emphasis on two characters; (1) the heavy glandularity, until recently thought to be rare or absent in *Polystichum* and infrequent in *Dryopteris*, and (2) the two leaf traces, rare in Dryopteridaceae. However, juvenile polystichums are often glandular (McHenry, 2012), and given the small size of the *Adenoderris* species, simpler petiole vasculature is to be expected (Bower, 1930). Hence the resolution of the century-old debate is that *Adenoderris* species are dryopteridaceae; those features suggesting the Athyriaceae are independently derived within the family.

The possibility exists that the suite of features that brought these two unrelated fern species together in a single

Table 3. Characters relevant to the generic placement of the two *Adenoderris* species.

Character	<i>A. glandulosa</i>	<i>A. sororia</i>
Rhizome habit	short-creeping	short-creeping
Rhizome and petiole scales	narrow-lanceolate, sparsely glandular on edge and face, ochraceous	lanceolate, densely glandular on edge and face, ochraceous
Lamina dissection	once-pinnatisect with decurrent, shallowly lobed segments	once-pinnate with deeply lobed segments
Lamina texture	herbaceous	herbaceous
Indument	dense 2- or 3-celled uniseriate hairs	moderately dense 2-celled uniseriate hairs
Petiole vascular bundles	2	2
Spinules	weakly and intermittently spinulose	aspinulose
Vein forking	2 or 3 times forked	simple or forked 2 or 3 times acroscopically in the larger segments
Vein termination	at the margin	submarginal
Sorus position	abaxial on the vein	terminal on the vein
Indusium	peltate; erose to denticulate; glandular	reniform; erose denticulate, or fimbriate; glandular
Perispore	thin crests	broad folds
Distribution	Greater Antilles	northern Central America

genus may be explained by their convergent evolution in exploring low-light, stream-bank habitats. An array of miniature polystichums is known from low-light habitats on rocks, especially along streams (Mickel, 1997) and at cave entrances (e.g., Zhang & He, 2012). As juvenile polystichums are documented to be glandular-hairy, it seems possible that the glandularity in both *adenoderrises* is a convergent, pedomorphic trait. It is also possible, especially for *A. sororia* given its diminutive size and rarity, that they are simply precociously fertile individuals of a species that is ordinarily a much larger plant. Our results, however, argue that this is not the case. The autapomorphic single-nucleotide polymorphisms found in *A. sororia*, and our complete sample of the Central American clade, argue that it has a unique history among Central American *Dryopteris*.

Taxonomic implications. — Taxonomically, the genus *Adenoderris* is a synonym of *Polystichum*, and the two *Adenoderris* species belong to different genera. The nomenclature is as follows:

Polystichum glandulosum C. Presl, Tent. Pterid.: 82. 1836, non Dulac 1867 ≡ *Aspidium glandulosum* Hook. & Grev., Icon. Filic. 2: t. 140. 1829, non Blume 1828, nom. illeg. ≡ *Aspidium viscidulum* Mett. in Abh. Senckenberg. Naturf. Ges. 2: 322 1858 ≡ *Adenoderris glandulosa* (C. Presl) J. Sm., Hist. Fil.: 223. 1875 ≡ *Dryopteris viscidula* (Mett.) Kuntze, Revis. Gen. Pl. 2: 811. 1891 ≡ *Adenoderris viscidula* (Mett.) Maxon in Bot. Gaz. 39: 367, fig. 1. 1905, nom. superfl. (Art. 52.3) ≡ *Polystichum viscidulum* (Mett.) C. Chr., Index Filic. 75. 1905, nom. superfl. (Art. 52.3) – Lectotype (designated by Proctor, Ferns Jam.: 412. 1985): Hooker & Greville, Icon. Filic. 2: t. 140, based on material from Jamaica provided by several collectors.

Dryopteris sororia (Maxon) M. McHenry, Sundue & Barrington, **comb. nov.** ≡ *Adenoderris sororia* Maxon in Bot. Gaz. 39(5): 368, f. 2. 1905 – Holotype: GUATEMALA. Alta Verapaz. Sesisp., Apr. 1887, *H. von Türckheim* 868 (YU No. 000681!; isotypes: BM No. 000937852!, FI No. 005045!, K No. 000632727! [fragm. ex UC], MICH No. 1190003!, NY No. 00179342!, UC No. 678562!, US No. 831633!).

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Appendix 1. Taxonomic sampling for this study.

Taxa are arranged alphabetically by genus. *Taxon*; voucher specimen (for newly reported sequences) locality, collector and collection number (herbarium); and GenBank accession numbers for sequences utilized (*rbcL*, *trnL-F*, *rps4*, *matK*, listed respectively). “–” indicates missing data.

Adenoderris [= *Polystichum*] *glandulosum* Hook. & Grev.; Cuba, *Maxon 4078* (NY); KC878850, KC878869, –, –, *Adenoderris* [= *Dryopteris*] *sororia* Maxon; Mexico, *D.E. Breedlove 56215* (NY); KC878849, KC878866, –, –, *Asplenium septentrionale* (L.) Hoffm.; JF832054, –, –, JF832254. *Athyrium rupestre* Kodama; EU329045, –, –, *Blechnum spicatum* (L.) J. Sm.; JF832262, –, –, *Cornopteris christenseniana* (Koidz.) Tagawa; EU329061, –, –, *Cryptogramma acrostichoides* R.Br.; JN189571, –, –, *Ctenitis sloanei* (Poepp. ex Spreng.) C.V. Morton; EF463172, –, –, *Ctenitis submarginalis* (Langsd. & Fisch.) Ching; EF463173, –, –, *Cyclodium trianae* (Mett.) A.R. Sm.; EF463175, –, –, *Cyclopetis semicordata* (Sw.) J. Sm.; EF463234, –, –, *Cyrtogonellum caducum* Ching; AY694811, –, –, *Cyrtogonellum fraxinellum* (Christ) Ching; AY694810, –, –, *Cyrtomium falcatum* (L.f.) C. Presl; AF537226, EF177268, KF020442, –, *Cyrtomium chingianum* P.S. Wang; AY694803, –, –, *Cystopteris moupinensis* Franch; JF832268. *Davallia* sp.; ES-2011; JN189562, –, –, *Deparia petersenii* (Kunze) M. Kato; JF832065, –, –, *Diplazium nipponicum* Tagawa; AB574979, –, –, *Dryopteris alpestris* Tagawa ex Ching & S.K. Wu; JN189536, JN189105, –, *Dryopteris chrysocoma* (Christ) C. Chr.; JN189542, JN189111, –, *Dryopteris cinnamomea* (Cav.) C. Chr.; JN189528, JN189097, –, *Dryopteris expansa* (C. Presl) Fraser-Jenk. & Jermy; AY268844, AY268775, –, *Dryopteris filix-mas* (L.) Schott; JN189507, JN189075, –, *Dryopteris flaccisquama* A. Rojas; JN189519, JN189088, –, *Dryopteris fragrans* (L.) Schott; JN189512, JN189080, –, *Dryopteris futura* A.R. Sm.; JN189534, JN189103, –, *Dryopteris goldiana* (Hook.) A. Gray; JN189505, JN189073, –, *Dryopteris hawaiiensis* (Hillebr.) W.J. Rob.; JN189580, JN189144, –, *Dryopteris huberi* (Christ) C. Chr.; JN189520, JN189089, –, *Dryopteris intermedia* Kuntze; WIS; JN189503, –, –, *Dryopteris karwinskyana* (Mett.) Kuntze; JN189521, JN189090, –, *Dryopteris knoblochii* A.R. Sm.; JN189522, JN189091, –, *Dryopteris ludoviciana* (Kunze) Small; JN189513, JN189081, –, *Dryopteris marginalis* (L.) A. Gray; JN189502, JN189071, –, *Dryopteris maxonii* Underw. & C. Chr.; JN189529, JN189098, –, *Dryopteris nubigena* Maxon & C.V. Morton; JN189501, JN189070, –, *Dryopteris pacifica* (Nakai) Tagawa; JN189594, JN189161, –, *Dryopteris patula* (Sw.) Underw.; JN189391, JN189610, –, *Dryopteris pseudofilix-mas* (Fée) Rothm.; JN189415, JN189633, –, *Dryopteris pulcherrima* Ching; JN189406, JN189083, –, *Dryopteris rosea* Mickel & Beitel; JN189524, JN189093, –, *Dryopteris rossii* C. Chr.; JN189531, JN189100, –, *Dryopteris saffordii* C. Chr.; JN189525, JN189094, –, *Dryopteris salvinii* (Baker) Kuntze; JN189526, JN189095, –, *Dryopteris simplicior* Mickel & Beitel; JN189527, JN189096, –, *Dryopteris sparsa* (D. Don) Kuntze; JN189601, –, –, *Dryopteris wallichiana* (Spreng.) Hyl; JN189497, JN189067, –, *Elaphoglossum andicola* (Fée) T. Moore; EF463185, –, –, *Elaphoglossum tripartitum* (Hook. ex Grev.) Mickel; EF463204, –, –, *Gymnocarpium dryopteris* (L.) Newman; JF832068, –, –, JF832277. *Hemidictyum marginatum* (L.) C. Presl; EF463318, –, –, *Homalosorus pycnocarpus* (Spreng.) Pic. Serm.; JF832070, –, –, *Hymenasplenium cheilosorum* (Kunze ex Mett.) Tagawa; JF832071, –, –, *Hypodematium crenatum* (Forssk.) Kuhn; JF832072, –, –, *Lastreopsis glabella* (A. Cunn. in Hook.) Tindale; EF463206, –, –, *Lastreopsis hispida* (Sw.) Tindale; EF463207, –, –, *Llavea cordifolia* Lag.; U27726, –, –, *Lomagramma guianensis* (Aubl.) Ching; EF463208, –, –, *Lomariopsis marginata* (Schrad.) Kuhn; AY818677, –, –, *Loxogramme salicifolia* Makino; JN189567, –, –, *Matteuccia struthiopteris* (L.) Tod.; JF832074, –, –, *Macrotelypteris torresiana* (Gaudich.) Ching; JF832073, –, –, *Maxonia apiifolia* (Sw.) C. Chr.; EF463209, –, –, *Megalastrum biseriale* (Baker) A.R. Sm. & R.C. Moran; EF463210, –, –, *Megalastrum macrotheca* (Fée) A.R. Smith & R.C. Moran; EF463211, –, –, *Microsorium* sp. (misidentified on GenBank as *Robititis heteroclita* by Skog & al., 2004); AY818679, –, –, *Nephrolepis davallioides* (Sw.) Kunze; JF832075, –, –, *Olfersia cervina* Kunze; JN189565,

Appendix 1. Continued.

–, –, –. *Onoclea sensibilis* L.; JF832076, –, –, –. *Onocleopsis hintonii* Ballard; JF832077, –, –, JF832281. *Oleandra articulata* (Sw.) C. Presl; EF463242, –, –, –. *Pentarhizidium orientale* (Hook.) Hayata; JF832079, –, –, JF832283. *Phegopteris connectilis* (Michx.) Watt; JF832080, –, –, –. *Phanerophlebia nobilis* (Schltdl. & Cham.) C. Presl var. *nobilis*; AF537231, EF177269, –, –, –. *Platyserium* sp.; ES-2011; JN189563, –, –, –. *Pleopeltis polypodioides* (L.) E.G. Andrews & Windham; JN189568, –, –, –. *Polypodium vulgare* L.; JF832081, –, –, –. *Polybotrya caudata* Kunze; JN189572, –, –, –. *Polystichopsis muscosa* (M. Vahl) Proctor; JN189559, JN189128, –, –, –. *Polystichopsis pubescens* (L.) C.V. Morton; JN189560, –, –, –. *Polystichum alfaroi* (H. Christ) Barrington; Costa Rica; *D. Barrington 1213* (VT); –, –, KC890808, –; Costa Rica; *D. Barrington 1978* (VT); AF537236, EF177271, –, –, –. *Polystichum ammifolium* (Poir.) C. Chr.; La Réunion; *T.A. Ranker 1537* (VT); AF537237, EF177287, KC890807, –. *Polystichum bakerianum* (Atkins. ex Clarke) Diels; China, *D. Barrington 2246* (VT); KC878851, KC878859, KC890809, –. *Polystichum concinnum* Lellinger ex Barrington; Costa Rica; *J. Kluge 1441* (VT); EF177320, EF177276, KC890810, –. *Polystichum drepanum* (Sw.) C. Presl; Madeira, *C.P.G. 8864* (VT); KC878852, KC878861, KC890811, –. *Polystichum ekmanii* Maxon; Dominican Republic; *P. Wieczorek 215* (VT); AF537242, EF177272, KC907700, –. *Polystichum hillebrandii* Carruth.; United States; *H. Driscoll 315* (VT); EF177323, EF177279, KC890812, –. *Polystichum lonchitis* (L.) Roth; United States; *Little 344* (VT); AF537247, KC878862, KC890813, –. *Polystichum montevidense* (Spreng.) Rosenst.; Brazil, *J. Condack 687* (RIO); KC819966, KC819981, KC819996, –. *Polystichum obliquum* (D. Don) T. Moore; China; *D. Barrington 2090* (VT); EF177328, EF177284, KC890823, –. *Polystichum orbiculatum* (Desv.) J. Rémy & Fée; Costa Rica, *D. Barrington 2143* (VT); KC878853, KC878863, KC890814, –. *Polystichum platyphyllum* (Willd.) C. Presl.; Ecuador, *M.A. McHenry 10-55* (VT); KC878854, KC878864, KC890815, –. *Polystichum pungens* (Kaulf.) Presl; South Africa; *J.P. Roux* (VT); AF537253, EF177295, KC890816, –. *Polystichum rhizophyllum* (Swartz) C. Pr.; Puerto Rico, *M. Sundue 2069* (VT); KC878855, KC878867, KC890821, –. *Polystichum scopulinum* (D.C. Eaton) Maxon; Canada, *P. Zika 18579* (VT); KC878856, KC878860, KC890817, –. *Polystichum sinotsussimense* Ching & Z.Y. Liu, China, *S.G. Lu Z16* (PYU); KC878857, KC878858, KC890820, –. *Polystichum speciosissimum* (A. Braun ex Kunze) Copel.; Costa Rica; *D. Barrington 2150* (VT); AF537255, EF177317, KC907701, –. *Polystichum subacutidens* Ching ex L.L. Xiang; China; *S.G. Lu D8* (PYU); AY545488, DQ150418, KC890822, –. *Polystichum turrialbae* H. Christ; Costa Rica; *Little 295* (VT); AF537259, KC878865, KC890818, –. *Polystichum underwoodii* Maxon; Dominican Republic; *Wieczorek 258* (VT); AF537260, KC878868, KC890819, –. *Rhachidosorus mesosorus* (Makino) Ching; JF832084, –, –, JF832287. *Rumohra adiantiformis* (G. Forst.) Ching; U05942, –, –, –. *Stigmatopteris lechleri* (Mett.) C. Chr.; EF463221, –, –, –. *Stigmatopteris longicaudata* (Liebm.) C. Chr.; EF463222, –, –, –. *Tectaria apiifolia* (Schkuhr) Copel.; EF463270, –, –, –. *Thelypteris palustris* Schott; JF832085, –, –, JF832292. *Woodsia plummerae* Lemmon; JF832088, –, –, JF832295. *Woodwardia unigemmata* (Makino) Nakai; JF832297, –, –, –.