

Polystichum lilianae sp. nov. (Dryopteridaceae) and its relationships to *P. fournieri* and *P. turrialbae*

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Barrington, D. S. (Pringle Herbarium, Department of Botany, University of Vermont, Burlington, VT 05405-0086, U.S.A.; email: David.Barrington@uvm.edu). *Polystichum lilianae* sp. nov. (Dryopteridaceae) and its relationships to *P. fournieri* and *P. turrialbae*. *Brittonia* 55: 317–325. 2003.—Morphological, isozyme, and cytological analyses of the small, pale-scaled polystichums from oak-dominated montane rain forests in Costa Rica and Mexico reveal the presence of a separable undescribed species endemic to the Cordillera de Talamanca of Costa Rica. The new taxon, **Polystichum lilianae**, is an allotetraploid hypothesized to have the sympatric *P. turrialbae* as one diploid progenitor based on isozyme characters. The isozyme and morphometric data also support the inclusion of *P. smithii*, described from southern Mexico, in *P. turrialbae*, described from Costa Rica. The name *Polystichum fournieri*, formerly used for all of these plants, applies to species endemic to Oaxaca and Chiapas, Mexico. It is not a progenitor of *P. lilianae*.

Key words: Costa Rica, Dryopteridaceae, Mexico, *Polystichum*.

Barrington, D. S. (Pringle Herbarium, Department of Botany, University of Vermont, Burlington, VT 05405-0086, U.S.A.; email: David.Barrington@uvm.edu). *Polystichum lilianae* sp. nov. (Dryopteridaceae) and its relationships to *P. fournieri* and *P. turrialbae*. *Brittonia* 55: 317–325. 2003.—Análisis de morfología, isozimas y cromosomas de poblaciones de *Polystichum* de las selvas montañosas lluviosas de Costa Rica y México indican que existe una nueva especie endémica en la Cordillera de Talamanca de Costa Rica. La especie nueva, **Polystichum lilianae**, es allotetraploide y parece tener la especie simpátrica *P. turrialbae* como una de sus progenitores diploides. Además, datos morfológicos y de isozimas sugieren que *P. turrialbae* incluye como sinonimo *P. smithii* del sur de México. *Polystichum fournieri*, nombre anteriormente usado para todas estas plantas, es endémica a Oaxaca y Chiapas, México, y no es un progenitor de *P. lilianae*.

One of the significant problems remaining in Central American *Polystichum* relates to plants traditionally determined as *P. fournieri* A. R. Sm. These plants are small relative to most other Central American *Polystichum* species (fertile fronds 30–60 cm long), but they often have large indusia (>1 mm diam.) They typically inhabit wet oak forests between 2300 and 3200 m, where they are especially common along watercourses in deep shade. They are unusual among polystichums in the region in having pale basal petiole scales.

There are three specific epithets that have recently been applied to these small polystichums: 1) *Polystichum fournieri* A. R. Sm. is based on a lectotype chosen by Smith (1980) from pine forest in Chiapas, Mexico. The type matches modern collections such as *Barrington 1747* (VT) and *Pringle 5603* (VT). All have large indusia, small leaves, and normal circinate vernation. I found these plants growing on wet, seeping rockfaces and along stream courses in the region of Sierra de San Felipe and Cerro Pelón, Oaxaca, Mexico. 2) Plants named *P. smithii*

Mickel & Beitel come from Oaxaca, Mexico. They have basal pinnae shorter than the next distal pinnae and droop-tip circinate veneration (sensu Conant and Cooper-Driver, 1980: partly uncoiled croziers initially bent downward and later straightened up). They tend to grow at higher elevations in the open forest instead of on seepy rockfaces. 3) *Polystichum turrialbae* H. Christ, the type of which was collected by Wercklé on Volcán Turrialba, Costa Rica, resembles the Mexican *P. smithii*. Plants similar to the type are widespread in the upper oak forests of Costa Rica.

At the time I completed the treatment of *Polystichum* for the *Flora Mesoamericana* (Barrington, 1995), the relationships of these names were unclear. Seeking an understanding of species relationships in these polystichums, I launched a morphometric, cytological, and isozyme study centered on my own collections from Cerro de la Muerte, Costa Rica and Cerro Pelón, Oaxaca, Mexico. This work has led to a systematic review of *P. fournieri*, *P. smithii*, and *P. turrialbae*.

Methods

FIELD METHODS

Specimens were collected on Volcán Irazú and Cerro de la Muerte, Costa Rica, from 1978 to 2001 and on Cerro Pelón, Oaxaca, Mexico, in 1990 and 1992. Living plants were imported under the appropriate permits and maintained under shade cloth or in terraria in the University of Vermont greenhouses. Material for both isozyme analysis and chromosome work was obtained from these plants. Herbarium specimens serving as vouchers for these plants are listed in Appendix I.

ISOZYMES

The isozyme sample comprised 46 plants from Cerro de la Muerte, Costa Rica, and 52 (29 of *Polystichum fournieri* and 23 of *P. smithii*) from Cerro Pelón, Oaxaca, Mexico. Isozyme electrophoresis and staining methods were primarily as reported in Barrington (1990). I resolved phosphoglucose isomerase (PGI), phosphoglucose mutase (PGM), and

triosephosphate isomerase (TPI) on a modified system 6 of Soltis et al. (1983). Amino aspartate transferase (AAT), diaphorase (DIA), hexokinase (HK), and leucine aminopeptidase (LAP) were resolved on system 8 of Soltis et al. (1983). Their system 11 was modified to resolve 6-phosphogluconate dehydrogenase (6PGD), aconitase (ACN), NADP-dependent glyceraldehyde-3-phosphate dehydrogenase [NADP](G3PDH), isocitrate dehydrogenase (IDH), malate dehydrogenase (MDH), shikimate dehydrogenase (SkDH), and uridine diphosphoglucose pyrophosphorylase (UGPP). Staining of isozymes followed Soltis et al. (1983) and Werth (1985) except that staining of UGPP was modified from Jech and Wheeler (1984). All liquid stains except those for LAP and AAT were converted to agarose overlays by reducing the quantity of stain buffer to 3 ml, reducing the quantity of solid ingredients proportionately, and adding 6 ml of 1% agarose.

Fourteen isozymes were resolved, yielding 20 putative loci in the Costa Rican plants. Only sixteen of these were resolved for the Mexican plants. Apparently *g3pdh*, *pgil*, *tpil*, and *6pgd* were not expressed in these plants because they were grown in glass-covered terraria.

CYTOLOGY

Only plants from Cerro de la Muerte, Costa Rica, were used for cytological analysis. Developing leaves were fixed in Farmer's solution (3:1 glacial acetic Acid: 100% ethanol) for 18–24 hr, then transferred to 70% ethanol for storage at 0°C. Material in prophase of meiosis I was prepared by isolating sporangia on a slide, staining in 2% ferric acetocarmine for 2–3 minutes, and clearing and squashing in Hoyer's solution.

MORPHOMETRIC ANALYSIS

Herbarium specimens of the *Polystichum fournieri* complex from Mexico, Costa Rica, and Panama (Appendix I) were included in a principal-components analysis (PCA) to provide insight into the nature of variation in the complex. Leaves of 31 collections, each from a different sporophyte, were

scored for 22 characters (Appendix II). The characters were chosen based on a preliminary study of living plants from Costa Rica after two entities had been distinguished by isozyme analysis. Color character states are standard descriptive terms for Munsell color chips. The PCA was performed using the statistical analysis package JMP version 3.0.1 (SAS Institute, 1994).

Results

ISOZYME RESULTS

Polystichum sampled in this study, as in previous studies of Costa Rican *Polystichum* (Barrington, 1990), revealed little infraspecific variation. Six loci were invariant in all plants sampled: these were *aat*, *acn*, *dia1*, *mdh1*, *mdh2*, and *pgm1*. The four loci not expressed in Mexican plants (*6pgd*, *g3pdh*, *pgi1*, and *tpi1*) were present and invariant in all Costa Rican plants.

The polystichums from Cerro de la Muerte yielded two phenotypes. The first was monomorphic and presumably homozygous at all 20 sampled loci. The second, comprising 37 of the 46 plants sampled, showed constant paired bands suggesting fixed heterozygosity at six of the 20 loci (*hk*, *idh*, *mdh-3*, *pgi-2*, *pgm-2*, and *ugpp-1*). One of the two allozymes at five of these loci consistently comigrated with the single band evident in the nine homozygous plants. In the sixth locus (*hk*), the same individuals were heterozygous as in the other five, but neither of the constituent bands was shared with the homozygotes. At *lap*, the individuals heterozygous at the other loci had a different single band than the rest. This pattern of isozyme variation is consistent with there being two distinct entities in the forests of the Talamanca range, a tetraploid and one of its diploid progenitors.

Based on the 29 plants from Oaxaca, *Polystichum fournieri* has the same genetic profile as the first of the Costa Rican phenotypes (monomorphic at all loci). However, it is fixed for different alleles at five out of the 16 loci sampled (*dia2*, *hk*, *lap*, *pgi2*, and *skdh*), a difference typical of congeneric species in the ferns (e.g., Hauffler, 1985; Werth et al., 1985). In contrast, the

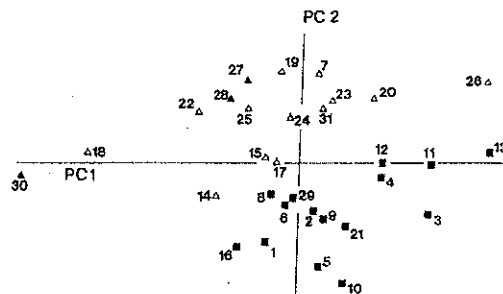


FIG. 1. Plot of principal-components axes 1 and 2 from a principal-components analysis of *Polystichum lillianae* (squares), *P. turrialbae* (open triangles), and *P. smithii* (solid triangles). Numbers are collections cited in Appendix II: Cytologically determined tetraploid is #2; cytologically and isozymically determined tetraploid is #16; types of *P. lillianae* are #s 11 & 12; isozymically determined diploid is #24; holotype of *P. smithii* is #27; holotype of *P. turrialbae* is #31.

23 *P. smithii* plants from the single Oaxaca population have a single phenotype that is identical at 15 of 16 loci to the first Costa Rican phenotype; they are homozygous and fixed for a different allele at *tpi2*. This close similarity is typical of different populations of the same species. Thus, from the Mexican isozyme data, it appears that neither of the Costa Rican entities is conspecific with *P. fournieri*, but Mexican *P. smithii* appears to be conspecific with the diploid cytotype from Costa Rica.

CYTOLOGY RESULTS

Two collections from Costa Rica (Barrington 677 and Barrington 2051) gave tetraploid counts of $n = ca. 82$ bivalents at meiosis I. The latter was among the plants that constantly yielded heterozygotes at six loci in the isozyme sample.

MORPHOMETRIC ANALYSIS

The first three vectors in the principal-components analysis accounted for 67% of the variance. These vectors were used to assess variation patterns in the Costa Rican plants. Plotting the distribution of plants using their eigenvalues for principal components 1 and 2 revealed two separate clusters (Fig. 1). Groups were best separated along the second principal component. Barrington 677 (#2, Fig. 1), cytologically determined to

be tetraploid (this paper), lies in one cluster. *Barrington 2051* (#16, Fig. 1), a cytologically determined tetraploid (this paper) that also showed the fixed-heterozygote isozyme pattern, is in the same cluster. The holotype of *Polystichum turrialbae* (#31, Fig. 1) lies near the center of the other cluster of plants, as does a plant inferred to be a diploid based on its isozyme phenotype (#24, Fig. 1). The holotype of *P. smithii* (#27, Fig. 1) and the other *P. smithii* sheets scored lie with the diploid plants from Costa Rica.

The most important characters in distinguishing the clusters discerned in the PCA, as judged from the loadings for principal component 2, were (in order of importance) lamina texture, pinna-rachis scale hue value, pinna-rachis scale shape, second pinna pinnule number, and pinna shape. Less important were the characters important for principal component 1, all of which were measures of segment length. In order of importance these were length of the first acroscopic pinnule of the medial pinna, length of the second basicopic pinnule of the medial pinna, length of the first pinna, length of the first basicopic pinnule of the medial pinna, and length of the second acroscopic pinnule of the basal pinna.

CONCLUSIONS

Combining the inferences from the three data sets, it appears that there are two genetically and morphologically distinct entities in Costa Rica, one of which is a diploid progenitor of the other. *Polystichum turrialbae* is the correct name for the diploid cytotype in Costa Rica. Isozyme and morphometric data indicate that Mexican *P. smithii* is conspecific with *P. turrialbae*; the latter is the older name.

The morphometric differences between the two Costa Rican entities indicate that they are different species. Thus, the upper oak forests of the Talamanca Mountains harbor two similar *Polystichum* species, one of which, *P. turrialbae*, is one of the diploid progenitors of the other, a tetraploid that I describe here. *Polystichum fournieri*, the name widely used for all of these plants, appears to be a species narrowly endemic

to southern Mexico; it is not the second progenitor of the Costa Rican tetraploid.

Taxonomic Section

POLYSTICHUM FOURNIERI A. R. Sm., Amer. Fern J. 70: 27. 1980, nom. nov. for *Polystichum mülleri* Mett. ex E. Fourn. 1872 not *Polystichum mülleri* Schumacher. 1803. *Polystichum mülleri* Mett. ex E. Fourn., Mexic. Pl. 1: 91. 1872.

TYPE: MEXICO. Chiapas: forests des pins. *J. J. Linden s.n.* (LECTOTYPE: designated by Smith, 1980, P).

Rhizome 1.5–2 cm diam., branching not seen. *Fronde* with normal circinate vernation; basal petiole scales narrow to long-lanceolate, coriaceous, light orangish yellow to light yellowish brown, conform, the cilia short to minute, flaccid; distal petiole scales similar, but broader, chartaceous to papyraceous, light orangish yellow; small petiole scales ovate-caudate, broadly to narrowly lanceolate, orangish yellow. *Lamina* 15–30 cm long, 2-pinnate, attenuate, chartaceous, dull green adaxially when fresh; rachis without a proliferous bud, the rachis scales orangish yellow. *Pinnae* 2–8 × 1–2 cm, acute to acuminate, attached at right angles to rachis, the basal pinnae usually as long as or longer than rest; abaxial pinna-rachis scales mostly narrow-lanceolate, deep orangish yellow to orangish yellow, with cilia occasional. *Pinnules* crenate, often attached at oblique angles, acute to obtuse, flat, the basal acroscopic pinnules longer than next distal, the basal basicopic pinnules near lamina base shorter than next distal pair; veins all free; short spinules usually developed on most vein tips. *Sorus* terminal on vein, the true *indusium* 0.6–1.7 mm diam., peltate, orangish yellow or light yellowish brown.

Distribution and ecology.—*Polystichum fournieri* is a rare species known only from Oaxaca and Chiapas, Mexico, where it is found between 2300 and 2700 m. It grows in the deep shade of streambeds and on dripping rock faces in forests of mixed oak, pine, and fir.

Additional specimens examined: MEXICO. Oaxaca: Dist. Ixtlán, Km 104.8 on Rte. 175, 2500 m, 3 Jan

1990, *Barrington* 1747, 2023 (VT); Dist Ixtlán, 40 km from Rancho Tejas on rd. from Ixtlán to San Miguel de Talea, 2300 m, 4 Jan 1990, *Barrington* 1753 (VT); Sierra de San Felipe, 9000 ft [2700 m], 21 Nov 1894, *Pringle* 5603 (VT).

***Polystichum lilianae* Barrington, sp. nov.**
(Fig. 2)

TYPE: COSTA RICA. Cartago: Villa Mills, in dense thicket in oak forest below and just E of Hotel La Georgina at Km 95 on the Pan-American Hwy., 3100 m, 15 Jan 1993. *D.S. Barrington* 2042 (HOLOTYPE: VT; ISOTYPES: NY, UC).

Species haec ab *Polysticho turrialbae* differt pinnis attenuatis non acuminatis, squamis petiolarum linea centrali fuscata manifeste signatis, squamulis rhachidis pinnarum angustis interdum uncinatis, pinnis basalibus longioribus quam ceteris, pinnulis ad rhachidin pinnarum oblique inclinatis.

Rhizome 1.5–2.3 cm diam., sometimes forking. *Fronde* with droop-tip veneration; basal petiole scales narrow to long-lanceolate, coriaceous (the edge rarely chartaceous), light orangish yellow to light yellowish brown with a well-developed and sharply delimited central deep brown stripe, conform, the cilia short to minute, flaccid; distal petiole scales similar, but broader, chartaceous to papyraceous, light orangish yellow or with a brownish orange to orangish yellow central stripe; small petiole scales ovate-caudate, broad-lanceolate, or long-lanceolate, orangish yellow with or without a brown central stripe. *Lamina* 30–45 cm long, 2-pinnate, acuminate, coriaceous, lustrous green adaxially when fresh; rachis without a proliferous bud, the rachis scales deep orange to brown. *Pinnae* 8–14 × 1.5–3.2 cm, attenuate, attached at right angles to rachis, the basal pinnae usually as long as or longer than rest; abaxial pinna-rachis scales abruptly contracted to an elongate apex (rarely long-lanceolate to narrow-lanceolate), deep orangish yellow to yellowish brown, with cilia more frequent at scale base. *Pinnules* crenate, often attached at oblique angles, acute to obtuse, flat, the basal acroscopic pinnules longer than next distal, the basal basiscopic pinnules near lamina base shorter than next distal; veins all free; short spinules usually developed on most vein tips. *Sorus* terminal on vein, the

true *indusium* 1.0–1.5 mm diam., peltate, deep orangish yellow to yellowish brown.

Distribution and ecology.—*Polystichum lilianae* is known only from Costa Rica, where it is found between 2500 and 3000 m on the Talamanca range and on the volcanoes of the Cordillera Central. It is likely to be found in western Panama. This species occurs in wet oak forests in dark sites near watercourses and on moist, shaded talus with *P. turrialbae*.

The species epithet honors my mother, Edith Lilian Barrington (1906–1992), my first botanical mentor.

Additional specimens examined: COSTA RICA. Cartago: Villa Mills, S of Hotel La Georgina, NNE of hwy. across from first borrow pit, 3100 m, 2 Jan 1980, *Barrington* 675, 676, 678, 679, 683 (VT), 677 (CR, VT); a few km N along dirt rd. N from Pan-American Hwy., 0.5 km N of La Georgina, *Barrington* 720 (VT); Villa Mills, below and just E of Hotel La Georgina at Km 95 on Pan-American Hwy., 3100 m, 15 Jan 1993, *Barrington* 2042 (NY, VT, UC), 2042a (UC [spores abortive, possible hybrid]); Cerro de la Muerte, Km 94 on Pan-American Hwy., 3100 m, 15 Jan 1995, *Barrington* 2044, 2045, 2046, 2047, 2048, 2049, 2051 (VT); vic. of Millsville, Pan-American Hwy., ca. 3 km above Nivel, 3000–3300 m, *Holm & Iltis* 548 (US); S slope of Volcán de Turrialba, nr. the Finca de Volcán de Turrialba, 2000–2400 m, *Standley* 35112 (US). San José: Villa Mills, S of Pan-American Hwy., ca. 1 km W of Hotel La Georgina, ca. 3100 m, 3 Jan 1980, *Barrington* 693, 699 (VT); NW along Pan-American Hwy. from Hotel La Georgina, 3100 m, 3 Jan 1980, *Barrington* 702, 703 (VT).

POLYSTICHUM TURRIALBAE H. Christ

Polystichum turrialbae H. Christ, Bull. Herb. Boissier, sér. 2, 6: 163. 1906.

TYPE: COSTA RICA. Heredia: Volcán Turrialba, 1905, *Wercklé* s.n.(P).

Polystichum smithii Mickel & Beitel, Mem. New York Bot. Gard. 46: 315, figs. 105I–K. 1988. TYPE: MEXICO. Oaxaca: Dist. Centro, top of Cerro San Felipe, 18 km N of Rte. 175 on lumber rd., 9500 ft [900 m], 15 Sep 1973, *J.T. Mickel* 7056. (HOLOTYPE: NY).

Rhizome ca. 1.5–2 cm diam., occasionally forking. *Fronde* with droop-tip veneration; basal petiole scales narrow to long-lanceolate, chartaceous, light orangish yellow to light yellowish brown throughout or sometimes with a weakly developed and poorly delimited yellowish brown to deep

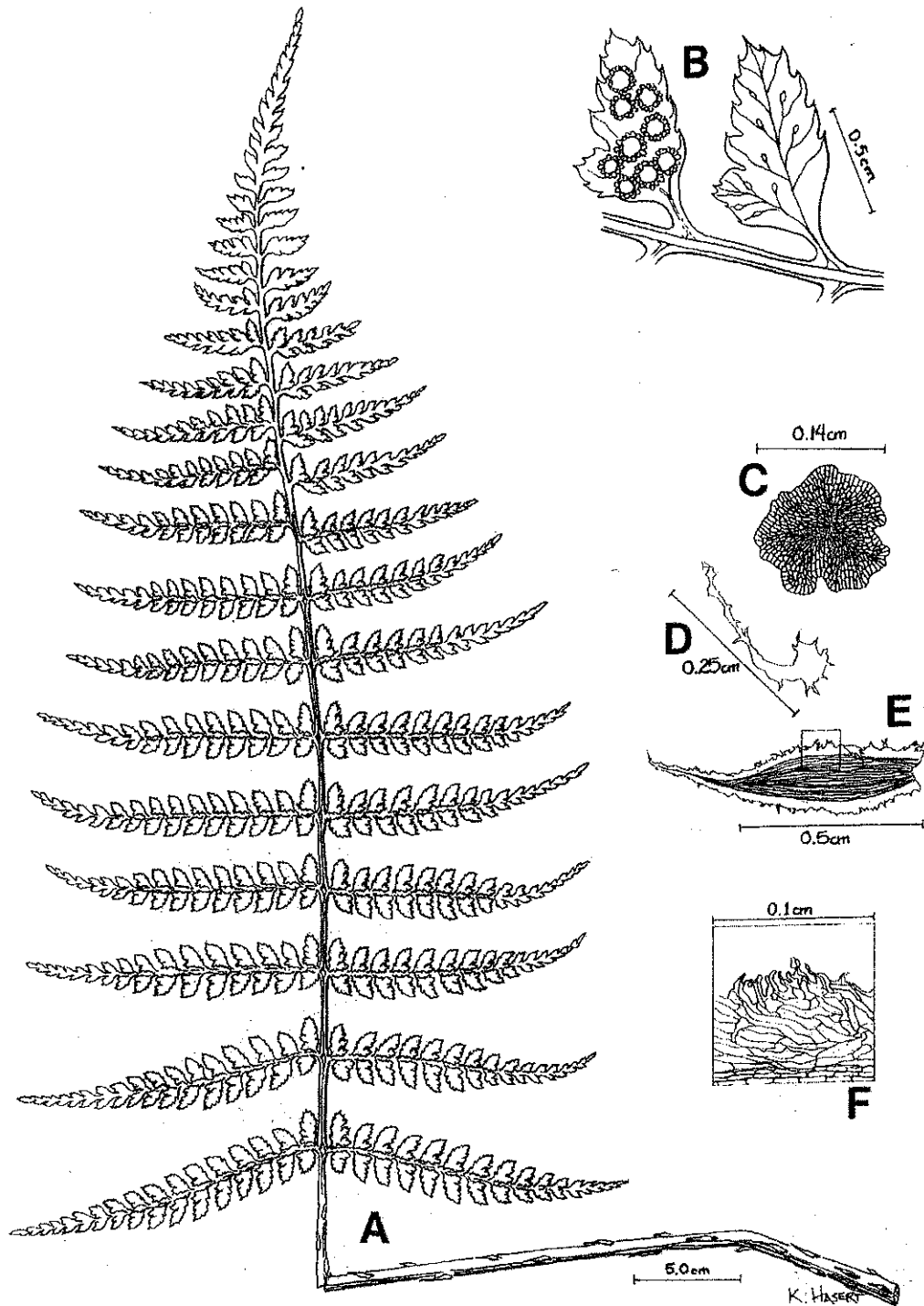


FIG. 2. *Polystichum lilianae* Barrington. A. Frond. B. Pinnules. C. Indusium. D. Abaxial pinna-rachis scale. E. Scale from base of petiole. F. Margin of basal petiole scale from location indicated in E. (From the holotype, Barrington 2042, VT).

yellowish brown center, conform, the cilia short to minute, flaccid; distal petiole scales similar; small petiole scales ovate-caudate, broad-lanceolate, or long lanceolate, orangish yellow. *Lamina* 15–45 cm long, 2-pinnate, attenuate, coriaceous, dull green adaxially when fresh; rachis without a proliferous bud, the rachis scales orangish yellow to light orangish yellow. *Pinnae* 6–12 × 1.1–2.7 cm, acute to acuminate, attached at right angles to rachis, the lowest pinnae often shorter than next distal, never longer; abaxial pinna-rachis scales mostly filiform, orangish yellow, rarely ciliate. *Pinnules* crenate, often attached at right angles, acute to obtuse, flat, the basal acroscopic pinnules longer than next distal, the basal basicopic pinnules near lamina base shorter than next distal; veins all free; spinules usually developed on most vein tips. *Sorus* terminal on vein, the true *indusium* 0.9–1.3 mm diam., peltate, yellowish brown to pale orangish yellow.

Distribution and ecology.—*Polystichum turrialbae* is widespread in tropical America at high altitudes, 2700–3200 m. It is disjunct between the mountains of S Mexico and Guatemala and the mountains of Costa Rica and Panama. It also extends into the Andes of Venezuela and Colombia, and there is a reliable report of the species from Bolivia (Smith et al., 1999). In Mexico it prefers streambanks in the oak-pine-fir forests of Cerro Pelón. In Costa Rica it is common in the high oak forests of the Talamanca range and the volcanoes, especially on talus slopes and roadbanks in shade, but also on wet forest slopes. The scant label data on Andean collections suggest similar habitats.

Additional specimens examined: MEXICO. **Distrito Federal:** Cañada de Contrera, 3100 m, *Rzedowski 19492* (ENCB). **Oaxaca:** Dist. Ixtlán, Km 104.8 on Rte. 175, 2500 m, 3 Jan 1990, *Barrington 2025* (VT); Dist. Central: top of Cerro San Felipe, 18 km N of Rte. 175 on lumber rd., 2900 m, 15 Sep 1973, *Mickel 7056* (NY); Dist. Ixtlán, above Ixtlán, Rancho Tejas, 3000 m, 22 Feb 2000, *Sundue 185* (VT). **Chiapas:** 1864–1870, *Ghiesbreght 371* (BM).

COSTA RICA. **Cartago:** Cerro de la Muerte, Km 94 on Pan-American Hwy., 3100 m, 15 Jan 1995, *Barrington 2050* (VT); above sanatorium nr. Cot, 2750 m, *Brown CR-51* (US); Irazú, près de nouveau volcán aux source du Río Sueci, 3000 m, 12 Dec 1888, *Pittier 948* (CR). **Heredia:** N of Heredia, ca. 1 km beyond

Porrosatí, 2100 m, 1970, *Lellinger & White 1680* (F. US). **Limón:** Cordillera de Talamanca, headwaters of unnamed W branch of the Río Teribe, between the Río Sini and the continental divide at Cerro Bekom, 9°10'45"N, 83°03'30"W, 2500–2600 m, 1984, *Davidse et al. 25740, 25761* (CR). Chirripó National Park, by path between Agua Potable and highest point of cloud forest on way to Chirripó, ca. 2800 m, 17 Feb 1983, *Garwood et al. 1330* (BM). **San José:** Villa Mills, S of Pan-American Hwy. ca. 1 km E of Hotel La Georgina, 3100 m, 3 Jan 1980, *Barrington 696, 697* (VT); 0.5 km E of Hotel La Georgina, 3100 m, 7 Jan 1981, *Barrington 801* (VT); Cerro de la Muerte, nr. soccer field ca. 1.5 km W of Hotel La Georgina, 3000 m, 24 Jan 1985, *Barrington 1270* (VT); Fila División, Cuenca, 2800 m, 15 Sep 1969, *Gómez P. 2378* (US).

PANAMA. **Bocas del Toro:** Upper Río Colubre, 2500–3000 m, Aug 1983, *Gómez P. et al. 21948* (CR, UC). **Chiriquí:** Deep wooded draw going W from "La Nevera" head of Río Chiriquí Viejo on Baru, 3200 m, 14 Mar 1979, *Hammel & D'Arcy 6397* (MO); Camp Cañassa, Chiriquí Volcano, 2500 m, 26 Feb 1918, *Killip 5445* (US, BM); above El Potrero Camp, Chiriquí Volcano, 2800–3100 m, 1918, *Killip 5455* (US).

VENEZUELA. **Merida:** above Chachopo, 3000 m, 26 Jan 1939, *Alston 6635* (BM).

COLOMBIA. **Norte de Santander:** La Mesita, Pamplona, 2725 m, 23 Feb 1939, *Alston 7227* (BM).

Key to the species of the *Polystichum* *fournieri* group

1. Basal pinnae usually shorter than the next; pinnules usually at right angles to pinna-rachis; abaxial pinna-rachis scales paler (orangish yellow) and filiform; indusia yellowish brown to pale orangish yellow, fresh leaves dull adaxially (diploid, Mexico to Bolivia) *P. turrialbae*
1. Basal pinnae as long as or longer than the next; pinnules usually oblique to pinna-rachis; abaxial pinna-rachis scales darker (usually deep orangish yellow to yellow brown) and broader at least at base; indusia pale to deep orangish yellow or yellow brown; fresh leaves lustrous or dull adaxially.
 2. Basal petiole scales light orangish yellow to light yellowish brown throughout; fronds with normal circinate veneration; abaxial pinna-rachis scales commonly narrow-lanceolate; indusia orangish yellow or light yellowish brown; fresh leaves dull adaxially (diploid, Mexico) *P. fournieri*
 2. Basal petiole scales light orangish yellow to light yellowish brown with a sharply delimited central deep brown stripe; fronds with droop-tip circinate veneration; abaxial pinna-rachis scales commonly widest at base and abruptly contracted to an elongate apex claw; indusia deep orang-

ish yellow to yellowish brown; fresh leaves lustrous adaxially (tetraploid, Costa Rica) *P. liliana*

Acknowledgments

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Appendix I

Herbarium specimens used in principal-components analysis (PCA). Species determinations match those in additional specimens examined, which see for full specimen data.

1. *Polystichum liliana*, Barrington 675 (VT); 2. *P. liliana*, tetraploid, Barrington 677 (VT); 3. *P. liliana*, Barrington 678 (VT); 4. *P. liliana*, Barrington 679 (VT); 5. *P. liliana*, Barrington 683 (VT); 6. *P. liliana*, Barrington 693 (VT); 7. *P. turrialbae*, Barrington 696 (VT); 8. *P. liliana*, Barrington 699 (VT); 9. *P. liliana*, Barrington 702 (VT); 10. *P. liliana*, Barrington 703 (VT); 11. *P. liliana*, Barrington 2042.1. (Isotype, NY); 12. *P. liliana*, Barrington 2042.2 (Holotype, VT); 13. *P. liliana*, Barrington 2042a (UC); 14. *P. lilia-*

nae, Barrington 2047 (VT); 15. *P. turrialbae*, Barrington 2050 (VT); 16. *P. liliana*, Barrington 2051, tetraploid (VT); 17. *P. turrialbae*, Brown CR-51 (US); 18. *P. turrialbae*, Davidse et al. 25761 (UC); 19. *P. turrialbae*, Gómez P. 2378 (US); 20. *P. turrialbae*, Gómez P. et al. 21948 (UC); 21. *P. liliana*, Holm & Iltis 548 (US); 22. *P. turrialbae*, Killip 5445 (UC); 23. *P. turrialbae* Killip 5455 (US); 24. *P. turrialbae*, diploid, Kintzer s.n. (VT); 25. *P. turrialbae*, Lellinger & White 1680 (F); 26. *P. turrialbae*, Maxon 5320 (US); 27. *P. turrialbae* (Holotype of *P. smithii*), Mickel 7056 (NY); 28. *P. turrialbae*, Rzedowski 19492 (ENCB); 29. *P. liliana*, Standley 35112 (US); 30. *P. turrialbae*, Sundue 185 (VT); 31. *P. turrialbae*, Werckle s.n. in 1905 (Holotype, P).

Appendix II

Characters used in the morphological analysis. The first pinna or pinnule is the most basal (lowest); the medial pinna is the one nearest the midpoint of the rachis. Pinnule lengths are taken from the base of the costa to the tip of the terminal spinule.

1. Pinna shape: attenuate (1), long-acuminate (2), acuminate (3).
2. Second pinna length.
3. First pinna length.
4. Number of pinnules on second pinna. A pinnule must be basally separated at least half its width from the pinna-rachis to be counted.
5. Number of pinnules on first pinna. See 4, above.
6. Indusium diameter. Measured from a dry indusium on the herbarium sheet; thus the size is underestimated.
7. Abaxial pinna-rachis scale shape: filiform (10), widest at base and abruptly contracted to an elongate apex (20), narrow-lanceolate (30), long-lanceolate (40). Scales between pinnule bases were used to assess character because the scales at the pinnule base vary.
8. Abaxial pinna-rachis scale cilia: absent (0), present (1), abundant (2).
9. Abaxial pinna-rachis scale hue value (amount of black): continuous, taken from Munsell color chips.
10. Abaxial pinna-rachis scale hue chroma (amount of pigment): continuous, taken from Munsell color chips.
11. Basal petiole scale hue: concolorous or weakly bicolorous (1), abruptly bicolorous (2). Measured from basal petiole scales.
12. Number of pinnae. A pinna must be basally separated at least half its width from the rachis to be counted.
13. Lamina length. Measured from lowest pinna-rachis to tip.
14. Lamina texture: coriaceous, high luster (1), intermediate (2), chartaceous, low luster (3).
15. First acroscopic pinnule of the first pinna, length.
16. Second acroscopic pinnule of the first pinna, length.
17. First basispic pinnule of the first pinna, length.
18. Second basispic pinnule of the first pinna, length.
19. First acroscopic pinnule of the medial pinna, length.
20. Second acroscopic pinnule of the medial pinna, length.
21. First basispic pinnule of the medial pinna, length.
22. Second basispic pinnule of the medial pinna, length.