

Pteris × *caridadiae* (Pteridaceae), a new hybrid fern from Costa RicaWESTON L. TESTO¹, JAMES E. WATKINS², JARMILA PITTERMANN³, AND REHMAN MOMIN²¹ Pringle Herbarium, Plant Biology Department, University of Vermont, 27 Colchester Avenue, Burlington, VT 05405, USA; e-mail: wtesto@uvm.edu² Biology Department, Colgate University, 13 Oak Drive, Hamilton, NY 13346, USA; e-mail: jwatkins@colgate.edu³ Department of Ecology and Evolutionary Biology, University of California Santa Cruz, 1156 High Street, Santa Cruz, CA 95064, USA; e-mail: jpitterm@ucsc.edu

Abstract. *Pteris* × *caridadiae*, a new hybrid fern from Costa Rica, is described and its relationships to its parents and other *Pteris* species are discussed. This is the first hybrid reported among a taxonomically complicated group of large, tripartite-leaved neotropical *Pteris* species.

Key Words: Fern, hybrid, Pteridaceae, *Pteris*, systematics, taxonomy.

The cosmopolitan fern genus *Pteris* L. comprises approximately 250 species and is most diverse in low- to mid-elevation forests in the tropics (Chao et al., 2014; Zhang et al., 2014). The group has received limited attention from taxonomists, and despite the contributions of recent phylogenetic studies (e.g., Bouma et al., 2010; Chao et al., 2012; Jaruwattanaphan et al., 2013; Chao et al., 2014; Zhang et al., 2014), the delineation of many species complexes remains problematic. Evidence from cytological (Manton, 1958; Walker 1958, 1960, 1962; Verma & Khullar 1965; Ammal & Bhavanandan, 1991) and phylogenetic (Denda et al., 2001; Chao et al., 2012; Jaruwattanaphan et al., 2013) studies highlights the importance of reticulate evolution in the group's evolutionary history. Despite this and the genus' large size, few *Pteris* hybrids have been described (Walker, 1958; Wagner & Nauman, 1982; Denda et al., 2001). Here we describe a new hybrid in the genus *Pteris* from Costa Rica, discuss its relationships to its parental taxa and distinguish it from other similar *Pteris* species.

Materials and methods

Field work was carried out at the Savegre Lodge and Reserve, San Gerardo de Dota, Provincia de San José, Costa Rica (9°54'94"N, 83°80'44"W) on 7–10 July, 2014. The study site was at an elevation of 2250 m, and comprised moist

upper montane forest adjacent to a small stream. The forest understory at the site was dominated by large terrestrial fern taxa, including *Diplazium diplazoides* (Klotzsch & H. Karst.) Alston, *Dicksonia sellowiana* Hook., *Thelypteris thomsonii* (Jenman) Proctor, *Pteris livida* Mett. (Testo 633, VT), and *Pteris podophylla* Sw. (Testo 634, MO, VT). The two *Pteris* species were particularly abundant at the site, with numerous large (to 2 m tall) sporophytes and sizeable populations of gametophytes and young sporophytes found growing in wet soil and on rocky stream banks (Fig. 1). Identification of small plants was made possible by observing a transitional growth series of both species. During field work at this site, we encountered one plant growing on a disturbed stream bank which resembled both *P. livida* and *P. podophylla* by its large size, tripartite leaves, and 2-pinnate-pinnatifid laminae. Closer inspection showed the plant to be intermediate to those species in a number of characters, suggesting a hybrid origin and prompting more detailed examination, which is presented here.

To characterize the morphology of this plant and its affinities to both *P. livida* and *P. podophylla*, we took quantitative measurements of 15 morphological characters (Table I) and qualitative measurements of scale color and size. Measurements were obtained from all three mature leaves on the single potential hybrid plant and from two or three leaves per plant from five plants each of *P. livida* and *P. podophylla* from the

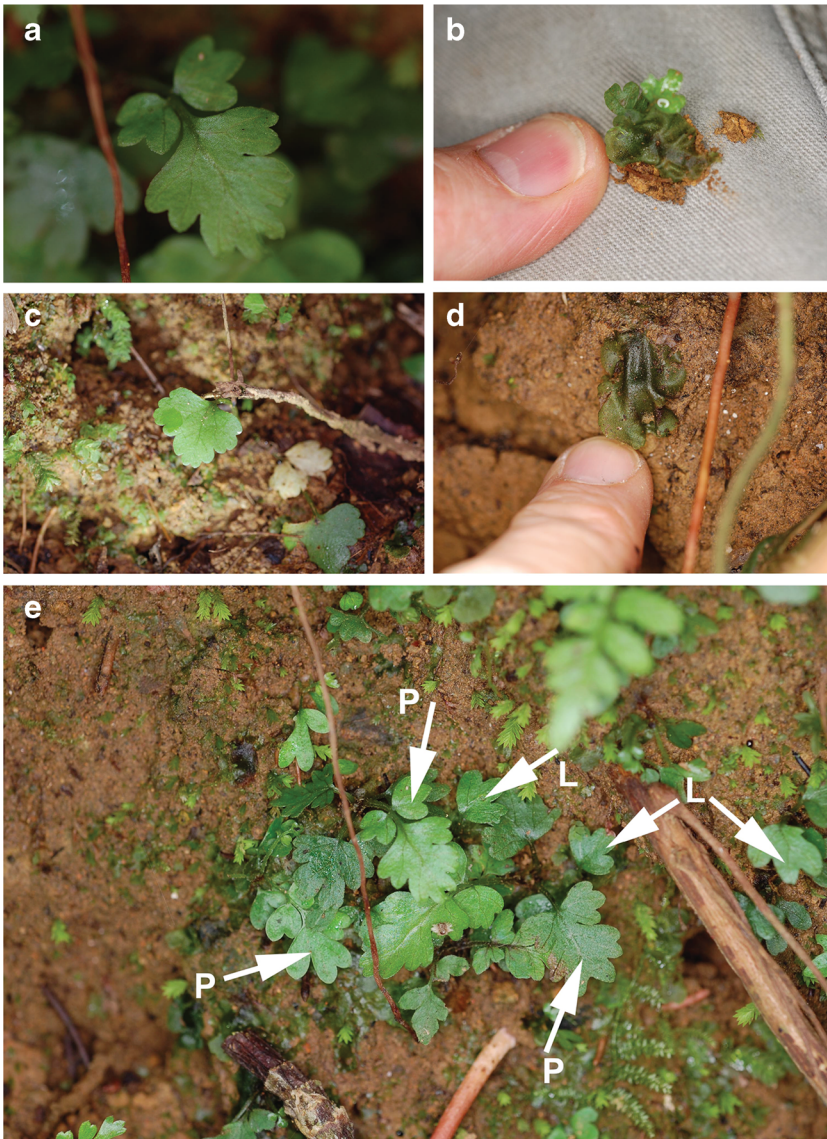


FIG. 1. Gametophytes and young sporophytes of *Pteris livida* and *P. podophylla* at type locality of *P. × caridadiae*. **A.** Young sporophyte of *P. podophylla*. **B.** Gametophyte of *P. livida*. **C.** Young sporophyte of *P. livida*. **D.** Gametophyte of *P. podophylla*. **E.** Gametophytes and young sporophytes of *P. livida* and *P. podophylla* growing on disturbed mud bank. “L” denotes *P. livida*, “P” denotes *P. podophylla*

same site. Stomata guard cell measurements were obtained by viewing epidermal peels at 20× magnification with an Olympus BX60 microscope; 27 stomata (length and width) were measured from each a single pinnule from each of the parents and the hybrid. Stomatal area was compared across taxa using one-way ANOVA. Rhizome scale measurements (15 per species) were obtained by hand with a Leica MX6 stereoscope at 125×

magnification. Unfortunately, no fertile leaves were present on the plant to assess whether the spores were normal or misshapen.

Results

The hybrid was intermediate between *Pteris livida* and *P. podophylla* in the majority of traits examined (Table I). Like that of both parental

TABLE I
MORPHOMETRIC COMPARISON OF *PTERIS* × *CARIDADIAE* AND ITS PROGENITOR SPECIES. ASTERISKS (*) DENOTE CHARACTERS FOR WHICH VALUES OF THE HYBRID ARE INTERMEDIATE BETWEEN ITS PARENTS.

Character	<i>P. livida</i>		<i>P. × caridadiae</i>		<i>P. podophylla</i>	
	Mean	SE	Mean	SE	Mean	SE
Stipe length (cm)	150.4	1.08	136.7	1.69	209.0	2.07
Stipe diameter (cm)	2.6	0.11	1.9*	0.14	1.9	0.24
Lateral pinna length (cm)	89.6	0.69	95.3	0.54	78.3	0.53
Lateral pinna width (cm)	58.8	0.52	45.3*	0.36	27.3	0.22
No. pinnule pairs on lateral pinna	8.8	0.18	11.3*	0.24	17.6	0.24
Length of medial pinnule of lateral pinna (cm)	34.3	0.39	25.0*	0.31	14.4	0.12
Width of medial pinnule of lateral pinna (cm)	12.7	0.24	5.7*	0.09	3.0	0.06
Apical pinna length (cm)	120.2	0.98	116.3*	1.41	101.3	0.57
Apical pinna width (cm)	71.0	0.37	58.0*	-	30.5	0.46
No. pinnule pairs on medial pinna	9.5	0.20	13.7*	0.48	24.8	0.36
Length of medial pinnule of apical pinna (cm)	31.4	0.27	34.3	0.48	16.1	0.13
Width of medial pinnule of apical pinna (cm)	9.1	0.22	11.5	0.29	3.2	0.06
Length of innermost basioscopic pinnule (cm)	31.0	0.34	51.7	0.80	35.6	0.38
Width of innermost basioscopic pinnule (cm)	11.1	0.23	37.0	0.38	21.6	0.29
Stomate size (μm^2)	1549.6	56.0	1422.1*	31.8	1126.9	22.6

taxa, the rhizome of the putative hybrid is massive, erect and scaly. The density and color of the rhizome indument was intermediate between those of *P. livida* and *P. podophylla*. The rhizome of *P. livida* is sparsely covered with concolorous light brown scales, whereas that of *P. podophylla* is densely covered with bicolorous dark brown/black scales. The rhizome of the hybrid is somewhat densely covered with concolorous dark brown scales.

The hybrid's petiole is green to stramineous and similar in size to that of both parents (1–1.5 m long, 1.8–2.2 cm wide). Transverse sections of the petiole revealed that the vascular bundles were arranged in a discontinuous omega-shape and weakly and incompletely surrounded by sclerotized tissue. This pattern resembles that of *P. livida* but differs from that of *P. podophylla*, which had an uninterrupted omega-shaped vascular bundle surrounded by a layer of sclerotized tissue (Fig. 2A–C). Petiole armament differed conspicuously between both parental taxa and was intermediate in the hybrid. The petioles of *P. livida* are armed with dark-brown, relatively large (to 1.5 mm) spines, whereas those of *P. podophylla* are unarmed. The hybrid had extremely small (to 0.2 mm long) spines that render the petiole scabrous.

The color, shape, size and distribution of the petiole scales were intermediate to the parental taxa (Fig. 2). The scales of the hybrid plant were uniformly dark brown, ovate-lanceolate, 7–11 × 3–4 mm and distributed sparsely throughout the

length of the petiole. In contrast, those of *P. livida* were uniformly light brown, ovate, 8–10 × 4–6 mm, densely distributed throughout the length of petiole and those of *P. podophylla* were light brown with a dark brown center, linear-lanceolate, 10–12 × 1.5–2 mm, and densely distributed only at the base of the petiole. The margin of the petiole scales of both *P. livida* and the hybrid were entire, whereas those of *P. podophylla* are conspicuously fimbriate (Fig. 2D–F).

Overall leaf morphology and lamina size of the hybrid resembled *P. livida* and *P. podophylla* (Fig. 3A–C). The fronds were tripartite and approximately 1.8–2 m long and 1.5–2 m wide. The hybrid was intermediate to its parents with respect to most aspects of leaf gross morphology, including apical pinna length and width, lateral pinnae width. The shape of the medial portion of the laminae (i.e., excluding the basal pinnae) was also intermediate: broadly lanceolate in *P. livida*, narrowly elliptic in *P. podophylla* and ovate in the hybrid (Fig. 3D–F). A conspicuous feature of all three taxa is the presence of enlarged innermost basal basioscopic pinnules on the lateral pinnae. These pinnules were smaller (28–36 × 10–13 cm) and less-divided (1-pinnate) on *P. livida* than either the new hybrid (45–55 × 36–38 cm; 1-pinnate-pinnatifid) or *P. podophylla* (31–43 × 21–27 cm, 1-pinnate-pinnatisect).

The hybrid plant was also intermediate to its parents with respect to the number, size and shape of the pinnules on both the lateral and apical

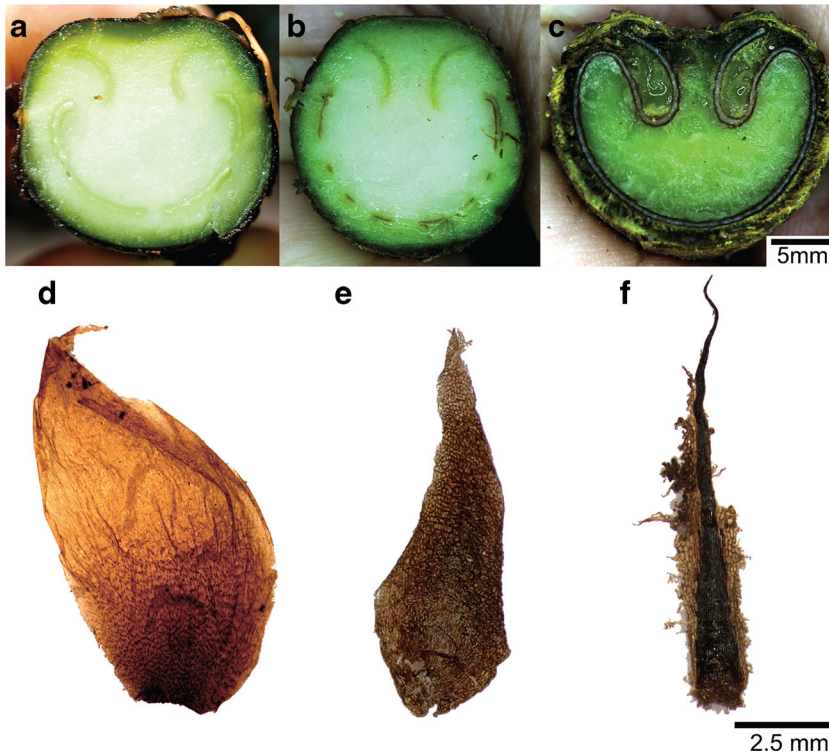


FIG. 2. Petiole cross-sections (A–C) and petiole scale morphology (D–F) of three taxa of *Pteris*. A, D. *P. livida*. B, E. *P. ×caridadae*. C, F. *P. podophylla*.

pinnae (Table I, Figs. 3, 4). The orientation of the pinnule ultimate segments ranged from ascending (*Pteris livida*) to weakly falcate (the hybrid) to strongly falcate (*P. podophylla*) (Fig. 4A–C). The lamina was chartaceous in both *P. livida* and the hybrid and subcoriaceous in *P. podophylla*. The leaf margin of both *P. livida* and the hybrid was crenate, whereas that of *P. podophylla* was spinulose. A single areole between the costule and the leaf margin was present in all three taxa (Fig. 4D–F). Whereas stomatal density was similar across the three taxa, stomatal size (calculated as length \times width) was variable: those of the hybrid were smaller than those of *P. livida* and larger than those of *P. podophylla* (Table I, $F_{81,2}=2.31$, $P<0.001$).

Several searches of the area failed to turn up additional individuals of the new hybrid; however, given the commonness of the parental species and their frequent co-occurrence in mid- to upper-elevation forests throughout the Neotropics, it is probable that this hybrid exists elsewhere. In addition to its progenitors, it is superficially similar to several other large *Pteris* species with tripartite laminae, most notably *P. navarrensis* Christ,

which differs by having bicolorous petiole scales and larger spines. With the similarities of these species in mind, careful observation in the field and examination of herbarium specimens may result in the discovery of additional discoveries of this hybrid. Because binomials are more stable nomenclaturally than formula names (Wagner 1969, Moran & Watkins, 2004), we propose the following binomial for this hybrid:

***Pteris ×caridadae* Testo & J. E. Watkins, hybrid nov.** Type: COSTA RICA. San José: San Gerardo de Dota, Savegre Natural Reserve, 9°54'94"N, 83°80'44"W, 2200 m, edge of stream at trailhead of El Canto de las Aves trail, montane oak forest, 8 Jul 2014, *W. Testo 585* (holotype: VT; isotype: CR). (Figs. 2, 3, and 4)

Hybrida inculta e *Pteride livida* et *P. podophylla* genita. Folia 1.8–2 m longa et 1.5–2 m lata. Folia pinnati-pinnatifida, pinnulis basalibus basisopicis, dilatatis 45–55 cm longis et 36–38 cm latis et stricte pinnatis-pinnatifidis. Folia fertilia ignota.

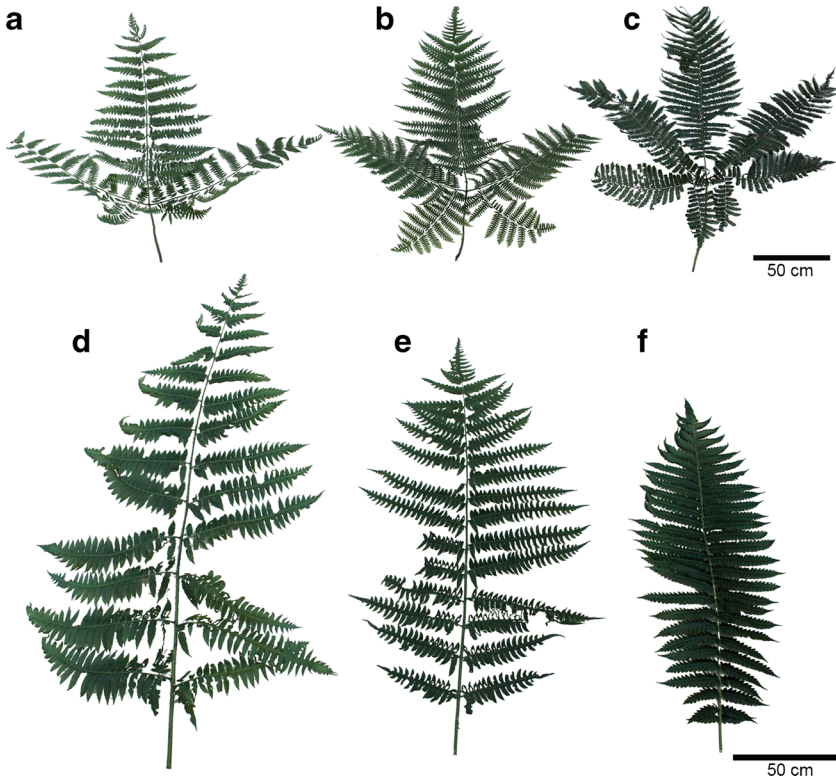


FIG. 3. Morphology of the fronds (A–C) and of central portion of laminae (D–F) in three taxa of *Pteris*. A, D. *P. livida*. B, E. *P. ×caridadiae*. C, F. *P. podophylla*

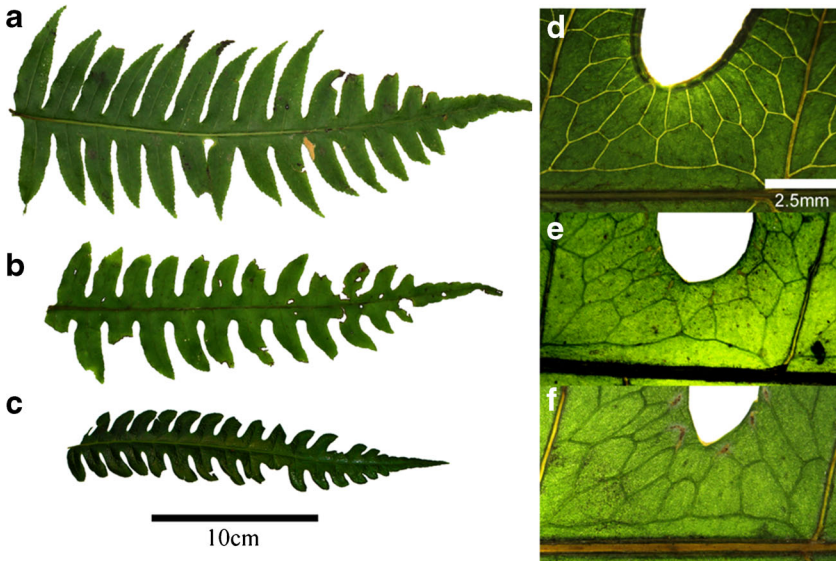


FIG. 4. Pinnule morphology (A–C) and leaf venation (D–F) in three taxa of *Pteris*. A, D. *P. livida*. B, E. *P. ×caridadiae*. C, F. *P. podophylla*.

Plant terrestrial with massive erect rhizome. Petiole armed with small spines, petiole scales sparse with entire margins, dark brown and ovate-lanceolate. Fronds 1.8–2 m long and 1.5–2 m wide, apical pinnae ovate, enlarged basal basioscopic pinnule 45–55 cm long and 36–38 cm wide. Fertile leaves unknown.

Etymology.—The hybrid epithet is named in honor of Caridad Zúñiga Calvo, who along with her husband Efraín Chacón Ureña and their eleven children, leads the preservation of the 400 ha Savegre Reserve where the hybrid was discovered. The reserve is within the Parque Nacional Los Quetzales and represents a pristine example of the rapidly disappearing upper montane forest in Central America.

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Literature Cited

- Ammal, S.L. & K. Bhavanandan.** 1991. Studies on the cytology of *Pteris silentvalliensis* N.C. Nair. *Indian Fern Journal* 8: 31–33.
- Bouma, W. L. M., P. Ritchie & L. R. Perrie.** 2010. Phylogeny and generic taxonomy of the New Zealand Pteridaceae ferns from chloroplast *rbcl* DNA sequences. *Australian Systematic Botany* 23: 143–151.
- Chao, Y.-S., H.-Y. Liu, Y.-C. Chiang & W.-L. Chiou.** 2012. Polyploidy and speciation (Pteridaceae). *Journal of Botany* doi:10.1155/2012/817920
- , **G. Rouhan, V. B. Amoroso & W.-L. Chiou.** 2014. Molecular phylogeny and biogeography of the fern genus *Pteris* (Pteridaceae). *Annals of Botany* 114: 109–124.
- Denda, T., H. Tomiyama & M. Yokoto.** 2001. An intersectoral hybrid in the genus *Pteris* (Pteridaceae) on Okinawa Island: evidence from nuclear and chloroplast DNA. *Acta Phytotaxonomica et Geobotanica* 52: 159–165.
- Jaruwattanaphan, T., S. Matsumoto & Y. Watano.** 2013. Reconstructing hybrid speciation events in the *Pteris cretica* group (Pteridaceae) in Japan and adjacent regions. *Systematic Botany* 38: 15–27.
- Manton, I.** 1958. Chromosomes and fern phylogeny with special reference to “Pteridaceae.” *Journal of the Linnean Society of London, Botany* 56: 73–92.
- Moran, R. C. & J. E. Watkins.** 2004. *Lomariopsis × farrarii*: a new hybrid fern between *L. japurensis* and *L. vestita* (Lomariopsidaceae) from Costa Rica. *Brittonia* 56: 205–209.
- Verma, S. C. & S. P. Khullar.** 1965. Cytogenetics of the Western Himalayan *Pteris cretica* complex. *Annals of Botany* 29: 673–681.
- Wagner, W. H.** 1969. The role and taxonomic treatment of hybrids. *BioScience* 19: 785–795.
- & **C. E. Nauman.** 1982. *Pteris × delchampsii*, a spontaneous fern hybrid from southern Florida. *American Fern Journal* 72: 97–102.
- Walker, T. G.** 1958. Hybridization in some species of *Pteris* L. *Evolution* 12: 82–92.
- . 1960. The *Pteris quadriaurita* complex in Ceylon. *Kew Bulletin* 14: 321–332.
- . 1962. Cytology and evolution in the fern genus *Pteris* L. *Evolution* 16: 27–43.
- Zhang, L., C. J. Rothfels, A. Ebihara, E. Schuettpelz, T. Le Péchon, P. Kamau, H. He, X. M. Zhou, J. Prado, A. Field, G. Yatskievych, X. F. Gao & L. B. Zhang.** 2014. A global plastid phylogeny of the brake fern genus *Pteris* (Pteridaceae) and related genera in the Pteridoideae. *Cladistics* doi: 10.1111/cla.12094.