Phytotaxa 550 (3): 215–223
https://www.mapress.com/pt/

Article

ISSN 1179-3155 (print edition)
PHYTOTAXA
ISSN 1179-3163 (online edition)

https://doi.org/10.11646/phytotaxa.550.3.1

Pteris pseudoamoena (Pteridaceae), a new species from Guangxi, China and Vietnam

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Abstract

A new species of fern, *Pteris pseudoamoena* (Pteridaceae), was identified and characterized. It is currently found in Guangxi Province, China, and Vietnam. *Pteris pseudoamoena* shares similar morphological characters with *P. amoena* and *P. mcclurei*, but can be distinguished by spores with echinate-tuberculate and auriculate ornamentation, straw-colored stipes, rachises and costae, and mostly free, occasionally interlinked veins. In addition, molecular evidence indicated that the new species is sister to a clade consisting of *P. amoena*, *P. nakasimae* and *P. mcclurei*.

Keywords: morphology characters, phylogeny, *Pteris*, taxonomy

Introduction

Pteris L. (1753: 1073) (Pteridaceae) is a large fern genus and contains ca. 200 (Tryon & Tryon 1982) to 250 species (Tryon *et al.* 1990) distributed throughout the tropical, subtropical, and temperate areas of all continents except Antarctic. Seventy species, 17 varieties and one form have been reported in China (Yang 2011, Zhang *et al.* 2013). Due to hybridization and apogamy, *Pteris* has complicated genetic relationships and some species in *Pteris* are highly similar in morphology (Li *et al.* 2004, Schuettpelz *et al.* 2007, Chao *et al.* 2012, Zhang *et al.* 2015, Wang *et al.* 2019). Therefore, the identification of *Pteris* is difficult.

In recent years, we conducted a study for assessment of *Pteris* in China. During the study, we concluded that samples collected in Guangxi Province, China and Vietnam represented a new species of *Pteris* because of its spore ornamentation and the color of stipes and rachises. We also found it has a unique phylogenic position among species of *Pteris*. Therefore, we described this species as a new one here.

Material and methods

Spores of the new species were collected from two specimens (*J. M. Wang 20150130-2, J. M. Wang 20150131-4 A7*) after the specimens were dried naturally. Spores were placed on double-sided tape on bronze stubs and sputter-coated with gold-palladium. Spore measurements and photomicrographs were made using a S-3000N Scanning Electron

Accepted by Joel Nitta: 31 May 2022; published: 16 Jun. 2022

Microscope (HITACHI, Japan). In our study, a total of 30 spores from the two samples were measured. Terms for describing the ornamentation of spores follow Wang & Yu (2003).

Thirty-five samples representing 30 *Pteris* species were used for phylogenetic study, including four individuals of the new species (Table 1). Ten *Pteris* samples were collected from the field by us and four chloroplast DNA (cpDNA) regions *(atpA, rbcL, rps4-trnS, trnL-trnF*) were obtained using the primers and protocol in Zhang *et al.* (2015). The cpDNA sequences of other *Pteris* samples were downloaded from Genbank mentioned in previous studies (Ebihara *et al.* 2010, Zhang *et al.* 2015, Zhang & Zhang 2018). DNA sequences were aligned using BioEdit v7.2.5 and manually edited. The combined cpDNA data set was used to constructed phylogenetic trees using maximum likelihood (ML), maximum parsimony (MP) and Bayesian methods. For ML analysis, the "GTRCAT" model was selected manually and 1,000 bootstrap replicates were conducted in RAxML software (Stamatakis 2014). For MP analysis, 1,000 tree-bisection-reconnection (TBR) searches were used for equally weighted maximum-parsimony analyses conducted for each locus in PAUP* v4.0. One thousand replicates were performed with 10 TBR searches per replicate and a maximum of 100 trees held per TBR search (Zhang & Zhang 2018). For Bayesian analysis, a phylogenetic tree was constructed using MrBayes v3.1.2 with the best-fit model (TIM+I+G) selected by AIC in Modeltest 3.7 1 (Ronquist & Huelsenbeck 2003). The parameter values were: base frequencies = (0.3030 0.1998 0.2225 0.2747), rate matrix = (1.0000 2.4808 0.4531 0.4531 2.8497 1.0000), Nst = 6, rates = gamma, shape = 0.9303, Pinvar = 0.4669. Four chains were run for 1,000,000 generations, with sampling every 1,000 generations. It was confirmed that the runs had converged by verifying that the standard deviation of the split frequencies was below 0.01. The majority-rule consensus tree was finally constructed with the first 25% of samples discarded as burnin.

TABLE 1. Taxa, locations and Genbank accession numbers of *Pteris* used for phylogenetic analysis in this study. Backslash (/) indicates missing data.

Taxon	**Location**	**Genbank accession no.**				**Voucher**	**Reference**
		rbcL	*atpA*	*trnL-trnF*	*rps4-trnS*		
P. pseudoamoena	Banbi, Guangxi, China	OM397928	OM397918	OM397938	OM397948	J. M. Wang 20150130-2 (IBSC)	this study
P. pseudoamoena	Banbi, Guangxi, China	OM397929	OM397919	OM397939	OM397949	J. M. Wang 20150130-4 (IBSC)	this study
P. pseudoamoena	Xialei, Guangxi, China	OM397930	OM397920	OM397940	OM397950	J. M. Wang 20150131-4 A7 (IBSC)	this study
P. pseudoamoena	Xialei, Guangxi, China	OM397931	OM397921	OM397941	OM397951	J. M. Wang 20150131-4 A8 (IBSC)	this study
P. angustipinnula	Wengʻang, Guizhou, China	OM397926	OM397916	OM397936	OM397946	J. M. Wang 20150517008 (IBSC)	this study
P. fauriei	Wengʻang, Guizhou, China	OM397925	OM397915	OM397935	OM397945	J. M. Wang 20150517007 (IBSC)	this study
P. fauriei var. *chinensis*	Longgang, Guangxi, China	OM397922	OM397912	OM397932	OM397942	J. M. Wang 20150129 (IBSC)	this study
P. majestica	Wuliangshan, Yunnan, China	OM397927	OM397917	OM397937	OM397947	J. M. Wang 0925012 (IBSC)	this study
P. splendida	Maolan, Guizhou, China	OM397924	OM397914	OM397934	OM397944	J. M. Wang 20150514015 (IBSC)	this study
P. splendida var. *longlinensis*	Luose, Guizhou, China	OM397923	OM397913	OM397933	OM397943	J. M. Wang 20150510011 (IBSC)	this study

.....continued on the next page

TABLE 1. (Continued)

Taxon	Location	Genbank accession no.				Voucher	Reference
		rbcL	*atpA*	*trnL-trnF*	*rps4-trnS*		
P. amoena	Medog, Tibet, China	KM008148	KM007583	KM008039 & KM007927	KM007814	Qingzang 74-5078 (KUN)	Zhang *et al.* 2015
P. biaurita	Nan, Thailand	KM008153	KM007588	KM008043 & KM007931	KM007818	A. Larsen 44455 (MO)	Zhang *et al.* 2015
P. bella	Guangdong, China	KM008151	KM007586	KM008041 & KM007929	KM007816	S. G. Wu s.n. (KUN)	Zhang *et al.* 2015
P. bella	Hainan, China	KM008152	KM007587	KM008042 & KM007930	KM007817	L. Zhang 1300 (CDBI)	Zhang *et al.* 2015
P. cadieri	Shixing, Guangdong, China	KM008158	KM007593	KM008048 & KM007936	KM007823	L. Zhang 1240 (CDBI)	Zhang *et al.* 2015
P. cretica	Bingchuan, Yunnan, China	KM008163	KM007598	KM008053 & KM007940	KM007828	X. H. Jin 11016 (CDBI)	Zhang *et al.* 2015
P. decrescens	Guizhou, China	KM008166	KM007601	KM008056 & KM007943	KM007831	L.T. Pan 053 (GZTM)	Zhang *et al.* 2015
P. dispar	Pujiang, Sichuan, China	KM008217	KM007651	KM008105 & KM007993	KM007882	L. Zhang 1463 (CDBI)	Zhang *et al.* 2015
P. ensiformis	Atherton Tablelands, Australia	KM008173	KM007608	KM008063 & KM007950	KM007838	ARF3539 (cult.)	Zhang *et al.* 2015
P. finotii	Wuzhishan, Hainan, China	KM008176	KM007611	KM008066 & KM007953	KM007841	L. Zhang 1323 (CDBI)	Zhang *et al.* 2015
P. insignis	Ruyuan, Guangdong, China	KM008186	KM007621	KM008076 & KM007963	KM007851	L. Zhang 1274 (CDBI)	Zhang *et al.* 2015
P. longipes	Baoting, Hainan, China	KM008195	KM007629	KM008085 & KM007972	KM007860	L. Zhang 1321 (CDBI)	Zhang *et al.* 2015
P. longipes	Dulongjiang, Gongshan, Yunnan, China	KM008194	KM007628	KM008084 & KM007971	KM007859	X. H. Jin 11507 (CDBI)	Zhang *et al.* 2015
P. mcclurei	Lechang, Guangdong, China	KM008196	KM007630	KM008086 & KM007973	KM007861	L. Zhang 1289 (CDBI)	Zhang *et al.* 2015
P. morii	Baoting, Hainan, China	KM008197	KM007631	KM008087 & KM007974	KM007862	L. Zhang 1314 (CDBI)	Zhang *et al.* 2015
P. multifida	Chongyi, Jiangxi, China	KM008200	KM007634	KM008090 & KM007977	KM007865	L. Zhang 1167 (CDBI)	Zhang *et al.* 2015
P. semipinnata	Pahang, Malaysia	KM008171	KM007606	KM008061 & KM007948	KM007836	E. Schuettpelz 893 (DUKE)	Zhang *et al.* 2015
P. setuloso-costulata	Sichuan, China	KM008218	KM007652	KM008106 & KM007994	KM007883	L. Zhang 1379 (CDBI)	Zhang *et al.* 2015
P. tripartita	Atherton, Australia	KM008224	KM007658	KM008112 & KM008000	KM007889	ARF3538 (cult.)	Zhang *et al.* 2015
P. vittata	Chengdu, Sichuan, China	KM008233	KM007667	KM008121 & KM008009	KM007898	L. Zhang 1466 (CDBI)	Zhang *et al.* 2015
P. wallichiana	Guangdong, China	KM008235	KM007669	KM008123 & KM008011	KM007900	L. Zhang 1284 (CDBI)	Zhang *et al.* 2015
P. aspericaulis	Yuanyang, Yunnan, China	MF972805	MF972644	MF972846	MF972843	L. B. Zhang 4925 (CDBI)	Zhang & Zhang 2018
P. grevilleana	Jerantut, Pahang, Malaysia	MF972811	MF972650	MF972852	MF972831	E. Schuettpelz 891 (DUKE, KEP)	Zhang & Zhang 2018
P. terminalis	West Maui, Hawaii, USA	MF972822	MF972657	/	MF972841	H. Oppenheimer s.n.	Zhang & Zhang 2018
P. nakasimae	Kagoshima Pref., Japan	AB574838	/	/	/	771437 (TNS)	Ebihara *et al.* 2010

Taxonomic treatment

Pteris pseudoamoena D. M. Yang & R. Guo, *sp. nov.* Figs. 1–3.

Type:—CHINA. Guangxi Province, Chongzuo City, Longzhou County, Banbi Village, under jungles in calcareous hills, 22.31°N, 106.99°E, elev. 270 m, 30 January 2015, *J. M. Wang 20150130-2* (holotype, IBSC!; isotypes, IBSC!), *J. M. Wang 20150130-3* (paratype, IBSC!), *J. M. Wang 20150130-4* (paratype, IBSC!); CHINA, Guangxi Province, Chongzuo City, Daxin County, Xialei Nature Reserve, on dry calcareous rocks, 22.91°N, 106.75°E, elev. 545 m, 31 January 2015, *J. M. Wang 20150131-4 A7* (paratype, IBSC!), *J. M. Wang 20150131-4 A8* (paratype, IBSC!).

Diagnosis:—Morphological features of the new species are similar to those of *P. amoena* Blume (1828: 210) and *P. mcclurei* Ching (1933: 28) with tripinnatifid laminae, but differ by having spores with echinate-tuberculate and auriculate ornamentation, straw-colored stipes, rachises and costae, free veins but occasionally interlinked veins, while *P. amoena* having spores with tuberculate and verrucate ornamentation, castaneous-brown to sorrel-red stipes, rachises and costae, and free veins, and *P. mcclurei* having spores with rugulate ornamentation, castaneous-brown to lightly brown stipes, rachises and costae, and interlinked veins (Table 2, Figs. 2 & 3).

Description:—Terrestrial, 65–130 cm tall. Rhizomes erect or ascending, short, 0.8–1.5 cm in diam., clothed with brown scales. Stipes, rachises and costae straw-colored, or stipes light brown at base, the upper parts, rachises and costae straw-colored. Stipes 32–65 cm long, 3–6 mm in diam., scaly at base, upper parts glabrous; laminae ovate, tripinnatifid, 26.6–58 × 36.6–55.5 cm, papery when dried; lateral pinnae 3–8 pairs, opposite or subopposite, obliquely extended; basal pinnae with 1–2 pairs of basiscopic pinnules, 13.3–29 × 3.3–6.5 cm, with 0.5–2.5 cm long petiolules; upper pinnae lanceolate, pectinate, sessile or subsessile, sometimes with one pair of basiscopic pinnules; middle pinnae 12–20 × 2.2–5 cm, base cuneate and slightly oblique, apex caudate, 1–6 cm long; terminal pinnae similar but slightly wider and with 2–5 cm long petiolules; segments 15–30 pairs, alternate, 2–7 mm apart, subfalcate, 1–3.5 × 0.4–0.6 cm, base slightly expanded and decurrent, sterile margins lightly serrulate, apex obtuse or mucronate, basal segments of pinnae slightly shorter; costae prominent and glabrous on the abaxial side, grooved on the adaxial side, with short spines along grooves and the base of midribs; veins decumbent, forked, free, or 2 opposite veins at the base of segments interlinked into a triangular mesh, and other veins outward from the mesh separate. Sori linear, along pinna margins. Spores tetrahedral, 37.2 × 31.6 μm, with echinate-tuberculate and auriculate ornamentation on the surface.

Distribution and Habitat:—*Pteris pseudoamoena* is currently found in Longzhou County and Daxin County of Guangxi Province, China, and in Kim Hy Nature Reserve of Bac Kan, Vietnam. It grows in calcareous hills with elevation 216–600 m.

Etymology:—The specific epithet refers to the most similar species *P. amoena* of the new species.

Common name (assigned here):—Ni Hong Gan Feng Wei Jue (拟红杆凤尾蕨; Chinese name).

TABLE 2. Morphological comparisons between *Pteris pseudoamoena*, *P. amoena* and *P. mcclurei*.

Taxon	Stipe	Rachis	Costa	Lamina	Vein	Spore
P. pseudoamoena	straw-colored or light brown at base, and straw-colored at upper part	straw-colored	straw-colored	tripinnatifid	mostly free but interlinked occasionally	echinate-tuberculate and auriculate on the surface
P. amoena	castaneous-brown at base and sorrel-red at upper part	sorrel-red	sorrel-red	tripinnatifid	free	tuberculate and verrucate on the surface
P. mcclurei	castaneous-brown at base and lightly brown at upper part	lightly brown	lightly brown to straw-colored	tripinnatifid	forming regularly arranged costal areoles	rugulate on the surface

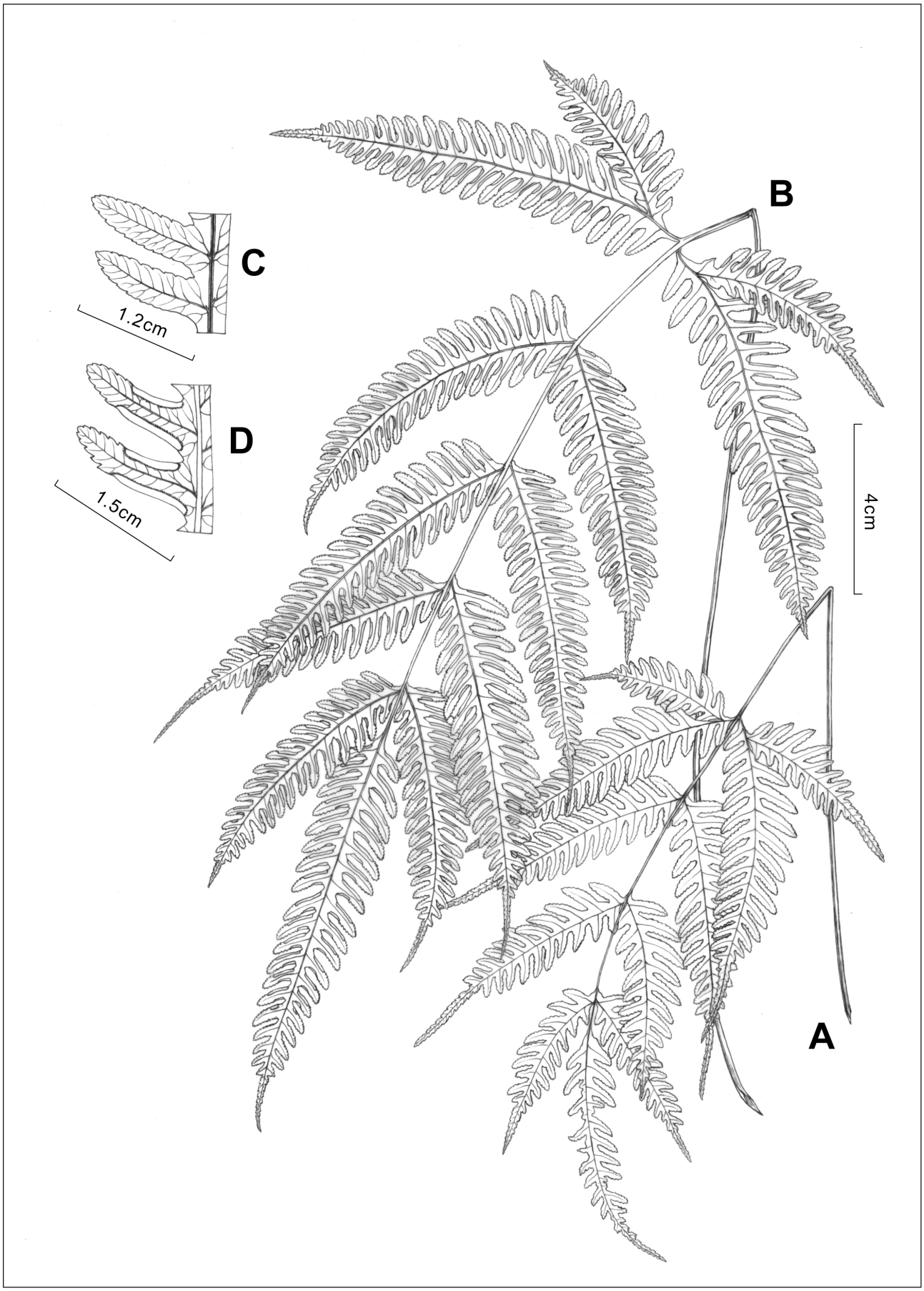

FIGURE 1. Line drawing of *Pteris pseudoamoena* D. M. Yang & R. Guo. Drawn by Yun-Xiao Liu based on the holotype J. M. Wang 20150130-2 (IBSC). A. Sterile frond; B. Fertile frond; C. Sterile segment; D. Fertile segment showing occasional interlinking of veins.

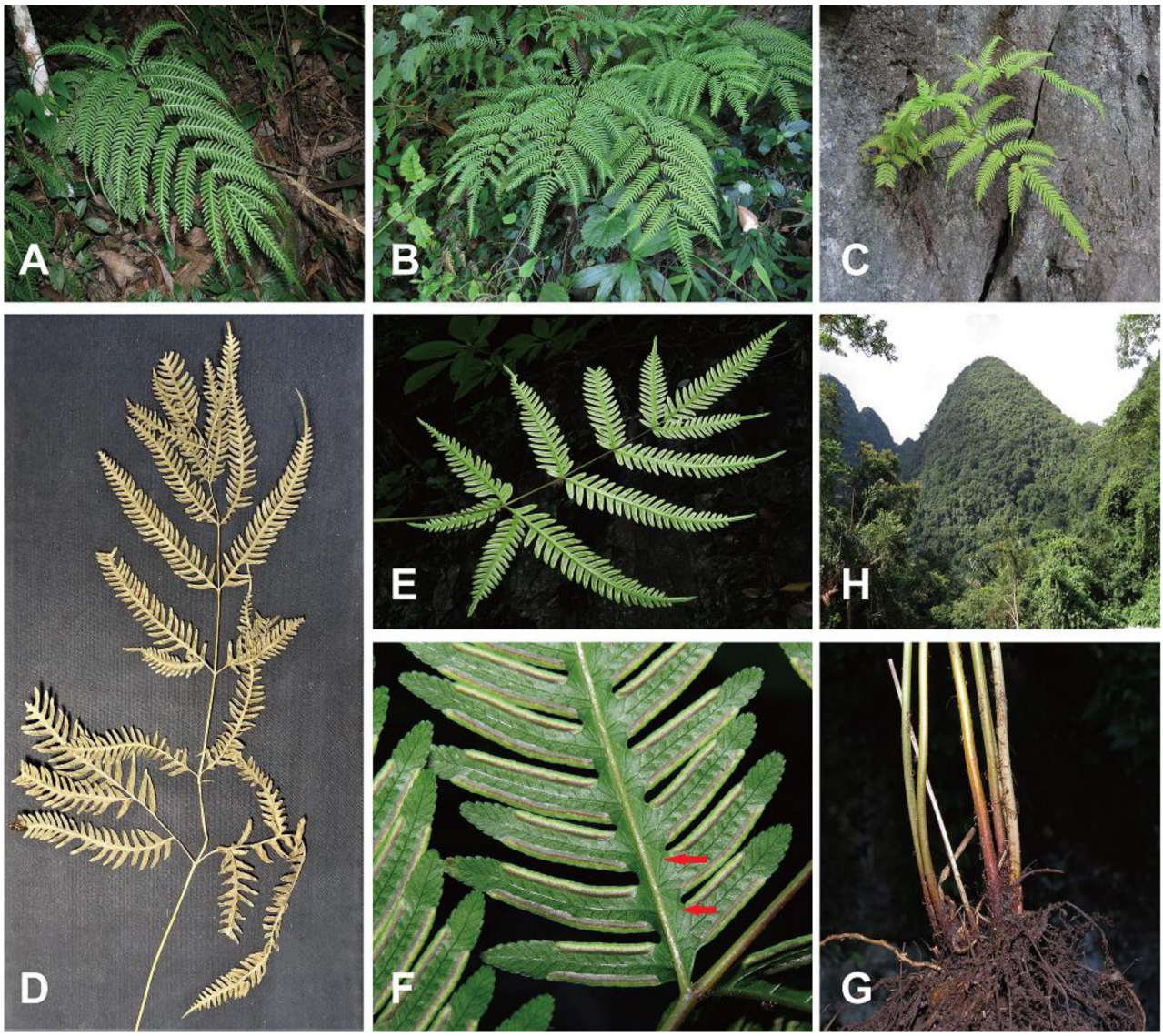

FIGURE 2. The habitat and morphology of *Pteris pseudoamoena* (photographed by Hong-Jin Wei except D). A. An individual with long apices of pinnae growing in a jungle on a calcareous hill; B. An individual with short apices of pinnae growing in a jungle on a calcareous hill; C. An individual growing in a calcareous rock crevice; D. An individual with two pairs of basiscopic pinnules at basal pinnae (*J. M. Wang 20150131-4 A7*); E. The abaxial side of a fertile frond; F. Segments of a fertile frond with interlinked veins indicated by arrows; G. Stipes with scales at base; H. Calcareous hills in Xialei Town, Daxin County, Guangxi Province, China.

Conservation status:—*Pteris pseudoamoena* is currently found in two counties of Guangxi, China and one location in Vietnam, and short of a profound study. According to the IUCN Red List criteria (IUCN 2012), it is considered data deficient (DD). Due to its current narrow geographical distribution, further investigations are still needed.

Phylogenetic evidence:—A maximum likelihood tree, Bayesian phylogenetic tree and maximum parsimony tree were constructed based on the sequences of *P. pseudoamoena* and other *Pteris* samples (Fig. 4). It is found that *P. pseudoamoena* is sister to a clade consisting of *P. amoena*, *P. nakasimae* and *P. mcclurei*. The new species forms a lineage independent from the other *Pteris* samples.

Other specimens examined:—CHINA, Guangxi Province, Chongzuo City, Longzhou County, Xiangshui Town, Siqing Village, in calcareous hills, 22.45°N, 107.11°E, elev. 216 m, 23 October 2019, *S. L. Jin JSL7327* (CSH). CHINA, Guangxi Province, Chongzuo City, Longzhou County, in calcareous hills, 2019, *S. L. Jin JSL7342* (CSH, KUN). CHINA, Guangxi Province, Chongzuo City, Daxin County, Xialei Town, Renyi Village, in calcareous rock crevices under jungles, 22.88°N, 106.76°E, elev. 541 m, 28 April 2014, *S. L. Jin YYH13555* (CSH); ibid., elev. 520–600 m, 2016, *S. L. Jin JSL4188* (CSH), *S. L. Jin JSL4198* (CSH), *S. L. Jin JSL4503* (CSH), *S.*

L. Jin JSL4504 (CSH), *S. L. Jin JSL4508* CSH), *S. L. Jin JSL4509* (CSH), *S. L. Jin JSL4509A* (CSH), *S. L. Jin JSL4512* (CSH), *S. L. Jin JSL4517* (CSH), *S. L. Jin JSL4522* (CSH); ibid., elev. 550 m, 20 May 2008, *HCAS GX Exp ed. 0792* (IBK); ibid., elev. 600 m, 11 August 2008, *HCAS GX Exped. 2676* (IBK). VIETNAM, Bac Kan, Na Ri, An Tinh Com., Kim Hy NR., Na Duong Village, inside a cave, elev. 300–530 m, 3 December 2013, *L. B. Zhang, L. Zhang & T. N. Lu 6661* (CDBI).

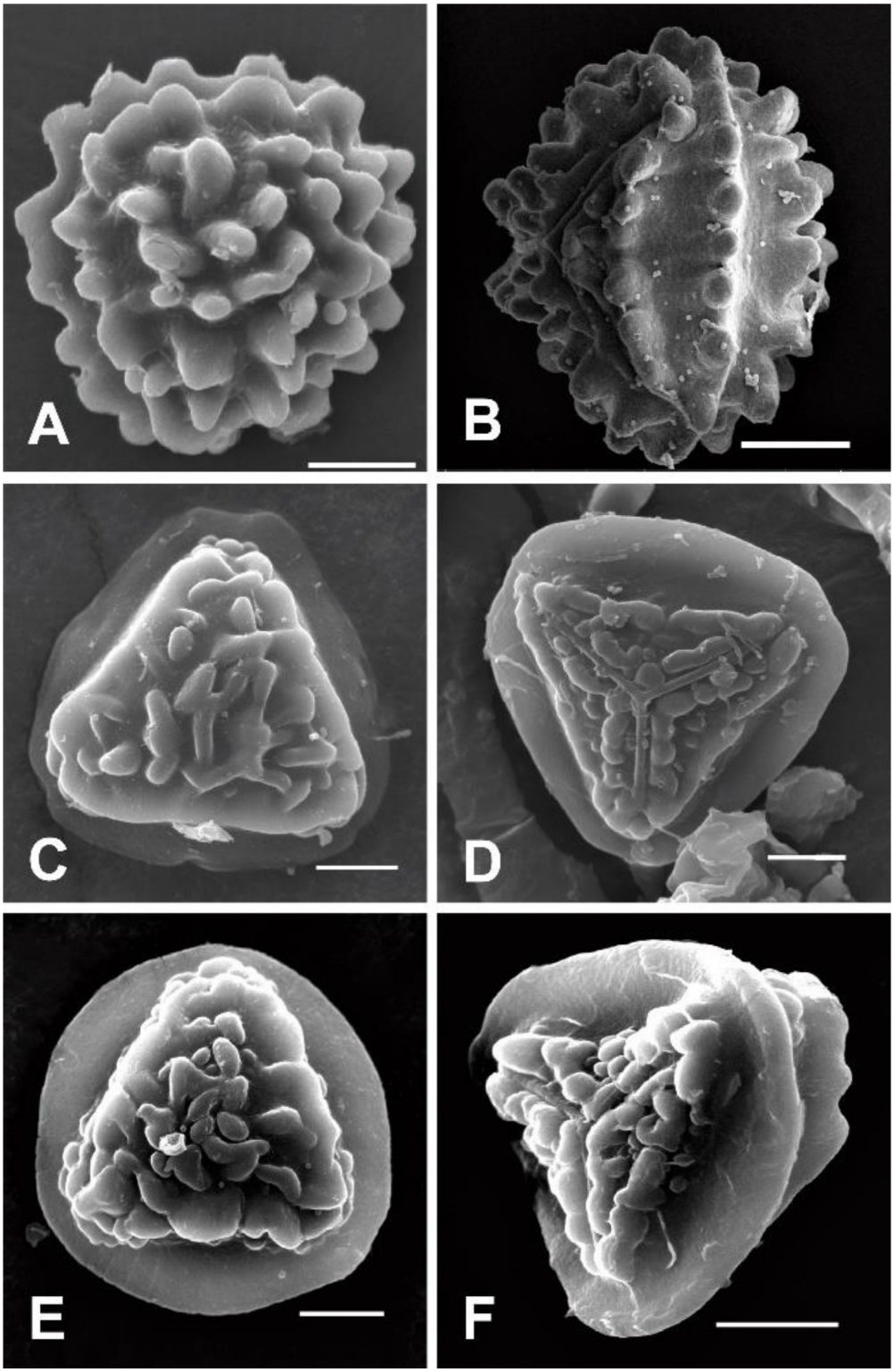

FIGURE 3. Spores of *Pteris pseudoamoena*, *P. amoena* and *P. mcclurei.* A–B. Spores of *P. pseudoamoena* with echinate-tuberculate and auriculate ornamentation (J. M. Wang 20150130-2 IBSC); C–D. Spores of *P. amoena* with tuberculate and verrucate ornamentation cited from Yang 2011 (B. G. Li 45755 HITBC); E–F. Spores of *P. mcclurei* with rugulate ornamentation cited from Yang 2011 (H. F. Qin 700276 IBSC). Scale bars: 10 μm (A–F).

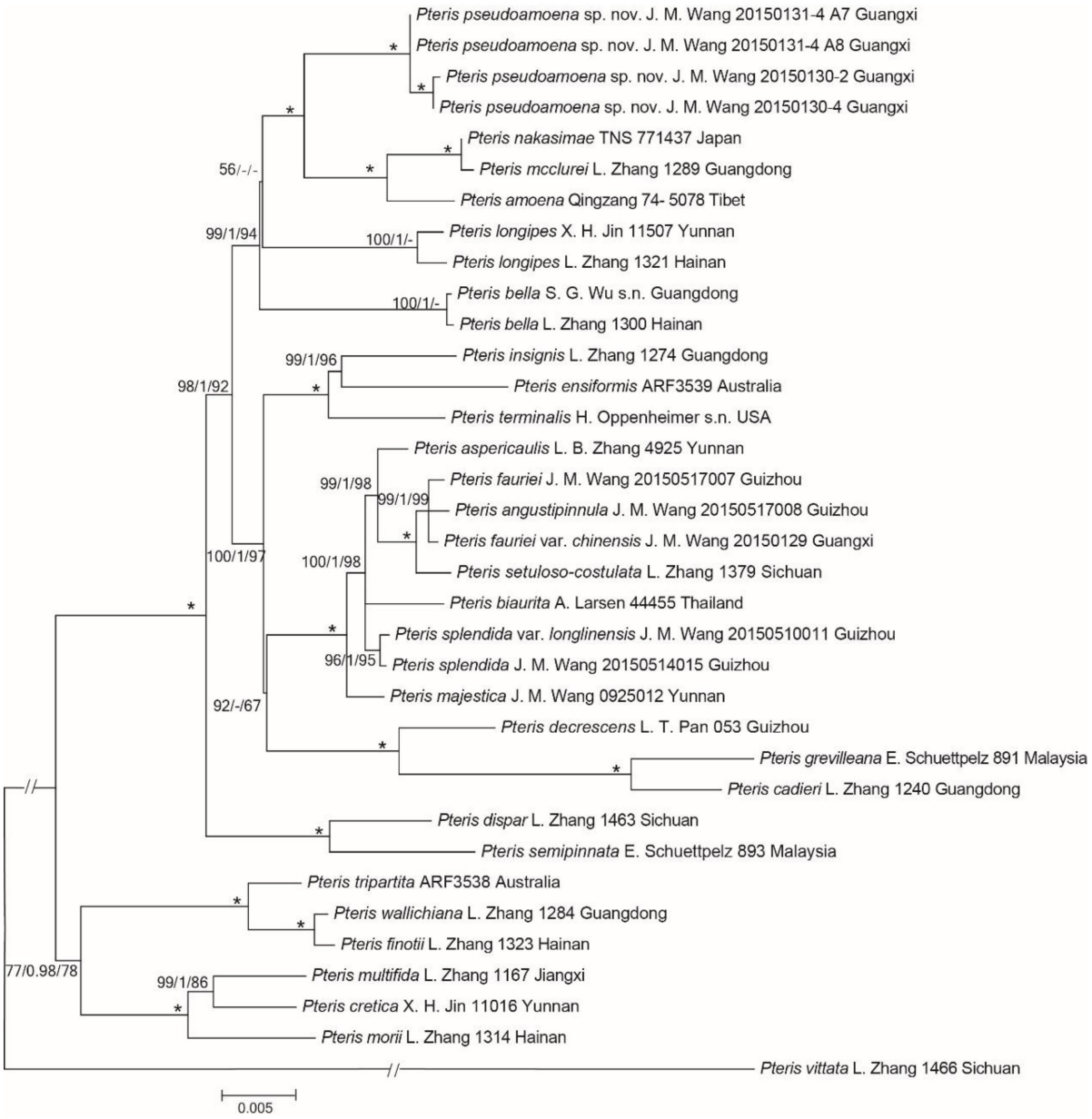

FIGURE 4. Maximum likelihood phylogeny of *Pteris*. Support values (maximum likelihood bootstrap support, Bayesian inference posterior probability and maximum parsimony jackknife support) are shown along the branches. Values of MLBS = 100%, BIPP = 1.0, and MPJK = 100%, are indicated by an asterisk. Dash (-) indicates nodes with BIPP < 0.9 or MPJK < 50%. "//" indicates that the branch length of the outgroup is shortened.

Acknowledgments

This research was funded by Hainan Provincial Natural Science Foundation of China (grant number 322RC571). The authors thank Hong-Jin Wei for sharing specimen information and habitat photos of the new species. We appreciate the editors and anonymous reviewers for carefully correcting the manuscript.

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