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Morphological and phylogenetic studies on *Whalleya*, with W. *hainanensis* sp. nov.

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Abstract: Two species of *Whalleya* from Guizhou and Hainan Provinces were collected during the investigation of *Xylariales* from 2019 to 2021. According to morphological and molecular characteristics, *Whalleya hainanensis* sp. nov. and *W. microplaca* are introduced. *Whalleya hainanensis* differs from *W. microplaca* by its obovoid or tubular perithecia and the slightly raised stromata; and differs from *W. maculata* by its smaller ascospores. Their detailed morphological descriptions and illustrations are provided. The taxonomic position of *Whalleya* is discussed.

Key words: *Lopadostomataceae*, 1 new species, morphology, phylogeny, taxonomy

1. Introduction

Whalleya J.D. Rogers, Y.M. Ju & F. San Martín. was proposed by Rogers et al. (1997), and affiliated with *Lopadostomataceae*, *Xylariales* by Hyde et al. (2017). *Whalleya* anamorph (wet scolecosporous conidia) is similar to that of *Lopadostoma turgidum* (Pers.: Fr.) Traverso, which had been classified in *Diatrypaceae* (Wehmeyer, 1975; Vasilyeva, 1998; Vasilyeva et al., 2007). Maharachchikumbura et al. (2016) classified *Whalleya* into *Xylariales incertae sedis* based on both xylariaceous and lopadostomaceous morphological affinities. Based on morphology of its conidial status and molecular data, *Whalleya* was accepted in *Lopadostomataceae* (Daranagama et al., 2018; Hyde et al., 2020).

Common morphological features of *Whalleya* are dark brown stromata, lacking KOH-extractable pigments, carbonaceous; white to brownish fungal tissue existing between the perithecia; ostioles below the surface of stromatal and appear as dots on the stromatal surface; ascospores usually have straight germ slits, light brown, perispore indehiscent in 10% KOH (Rogers et al., 1997). Currently, only two species of *Whalleya*, *W. microplaca* (Berk. & M. A. Curtis) J. D. Rogers, Y. M. Ju & F. San Martín and *W. maculata* (Theiss.) J. D. Rogers, Y. M. Ju & F. San Martín have been reported. *Whalleya microplaca*, the type species, is widely distributed in China (Rogers et al., 1997).

During the investigation of *Xylariales* in China, two taxa of Whalleya-like were encountered. Phylogenetic and morphological evidence shows that our new collection includes a novel species of *Whalleya* from Hainan province, and a known species *W. microplaca* from Guizhou province.

2. Materials and methods

2.1. Sample, isolation, and morphology

The specimens referred to in this paper were collected from Hainan and Guizhou provinces in China. The collected samples were placed in envelopes with basic information, such as latitude, longitude, and altitude, and brought back to the laboratory for microscopic observation in distilled water. The Melzer iodine reagent was used to check for

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the ascal apical apparatus. KOH (10%) was used to check for stromatal pigments and dehiscence of the perispore. The macroscopic traits were observed under Olympus SZ61 stereoscopic microscope. Micrographs were taken by a Canon digital camera (700D) under the composite microscope (Nikon Ni). At least 30 asci, 30 ascospores, and 20 perithecia were measured by the Tarosoft® image framework (v.0.9.0.7). Photomicrographs were improved and combined with Adobe Photoshop CS6 (USA).

The single spore isolation method was used to obtain pure cultures and the cultures were grown on oatmeal agar (OA) and potato dextrose agar (PDA) (Long et al., 2019; Pi et al., 2021). Pure colonies were transferred into 3 spiral capped centrifuges with sterile water and 2 filled with 10% glycerin water, which were sealed and then stored in refrigerators at 4 °C and –20 °C, respectively. The living cultures are stored in Guizhou Medical University Culture Collection (GMBC). Materials are stored in the Herbarium of Guizhou Medical University (GMB) and Herbarium of Cryptogams, Kunming Institute of Botany Academia Sinica (KUN-HKAS).

2.2. DNA extraction and sequencing

Fresh hyphae were scraped off on the medium with a clean scalpel blade for DNA extraction. According to the manufacturer protocol, the Fungal gDNA Isolation Kit (BW-GD2416, Biomiga, USA) was used to extract genomic DNA and kept under –20 °C.

The composition of the mixture and the thermal cycling procedure were followed by Pi et al. (2020) and Su et al. (2016). Primers for ITS5/ITS4, LR0R/LR5, T1/T22 or Bt2a/Bt2b, fRPB2-5F/fRPB2-7cR were used for amplifications of internal transcribed spacers (ITS) RNA gene, large subunit ribosomal (LSU) RNA gene, β-tubulin (*tub*2) gene and RNA polymerase II second largest subunit (*rpb*2) gene, respectively (Vilgalys and Hester, 1990; White et al., 1990; Glass and Donaldson, 1995; Liu et al., 1999; Hsieh et al., 2010). The amplified polymerase chain reaction (PCR) produces were detected with 1.5% agarose gel electrophoresis stained by GoldenView (G8140, Solarbio), and then sent to Sangon Biotech Co. (Shanghai), China for purification and sequencing. All new sequences generated were uploaded to GenBank.

2.3. Sequence alignment and construction of a phylogenetic tree

The information on sequences for the phylogenetic tree is listed in Table. All sequences were combined for phylogenetic analysis. All sequences were obtained from GenBank according to BLAST search and published literature (Ju et al., 2007; Daranagama et al., 2018; Hyde et al., 2020). Sequence alignment was carried out with the MAFFT v.7.110 online programs (Katoh and Standley, 2013) with the default settings. Sequences were trimmed with TrimAl (v.1.3) (Gappyout option) (Capella-Gutierrez et al., 2009; Samarakoon et al., 2022). Using BioEdit v.7.0.5.3 (Hall, 1999) assembled into ITS–LSU–*rpb2*–*tub2* datasets. The online tool ALTER (Alignment Transformation EnviRonment) was conducted to convert the file formats for RAxML and MrBayes (Ronquist et al., 2012; Xie et al., 2019).

Maximum likelihood analysis (ML) was generated with the GTR+G+I model of site substitution by using the RAxML 7.4.2 black box (Stamatakis et al., 2008). The branch support was calculated with a bootstrapping (BPP) method of 1000 replicates (Hillis and Bull, 1993). The Bayesian Inference analysis (BI) was performed using a Markov Chain Monte Carlo (MCMC) algorithm with Bayesian posterior probabilities (Rannala and Yang, 1996). Six Markov Chain Monte Carlo (MCMC) chains were run for 1,000,000 generations and sampled every 100 generations with a burning value of 25%. The posterior probabilities (PP) were counted to assess the remaining trees (Rannala and Yang, 1996). The final phylogenetic tree was viewed and checked with Figtree v1.4.0 (Rambaut, 2006) and polished using Photoshop CS6 software. Final alignments were registered in TreeBASE (ID 29057).

3. Results

3.1. Phylogenetic analyses

The combined ITS–LSU–*rpb*2–*tub*2 dataset included 44 taxa, 3121 positions including gaps (ITS: 1–579, LSU: 580–1,429, *rpb*2: 1,430–2,512, *tub*2: 2,513–3,121). Most genera of *Xylariales* were selected to construct the phylogenetic tree. *Achaetomium macrosporum* J.N. Rai, Wadhwani & J.P. Tewari (CBS 532.94), *Chaetomium elatum* Kunze (CBS 374.66) and *Sordaria fimicola* (Roberge ex Desm.) Ces. & De Not. (CBS 723.96) were selected as the outgroup taxa. The phylogenetic tree obtained from combining the alignment of ITS, LSU, *rpb*2, and *tub*2 sequences is represented in Figure 1. In the phylogenetic tree, new strains of *W. microplaca* and *W. hainanensis* show a close relationship with *W. microplaca* (YMJ 91111215). *W. hainanensis* and *W. microplaca* (YMJ 91111215) are separated into two distinct clades with high support values (99/1, Figure 1). *Xylariaceae* sp. FL0662, *Xylariaceae* sp. FL0638 and *Xylariacea*e sp. AK1116 as endophytic fungi clustered in the genus of *Whalleya*. Species of *Whalleya* cluster as a monophyletic clade with high support values (100/1, Figure 1), while the statistical support with other families of *Xylariales* is low (60% ML, 0.97 BYPP).

3.2. Taxonomy

Whalleya hainanensis Y.H. Pi & Q.R. Li, sp. nov. Figure 2

Mycobank No: 842380

Etymology—In reference to collection location, Hainan province.

Table. All taxa used in phylogenetic research are listed.

Species	Specimen number	Status	GenBank accession numbers				Reference
			ITS	LSU	*rpb2*	*tub2*	
Achaetomium macrosporum J.N. Rai, Wadhwani & J.P. Tewari	CBS 532.94	-	KX976574	KX976699	KX976797	KX976915	Wang et al., 2016
Amphibambusa bambusicola D.Q. Dai & K.D. Hyde	MFLUCC 11-0617	-	KP744433	KP744474	N/A	N/A	Senanayake et al., 2015
Anthostoma turgidum (Pers.) Nitschke	LT2	HT	KC774618	KC774618	KC774563	MF489024	Jaklitsch and Voglmayr, 2014
Arecophila bambusae Umali & K.D. Hyde	HKUCC 4794	-	N/A	AF452038	N/A	N/A	Jeewon et al., 2003
Astrosphaeriella erumpens (Berk. & M.A. Curtis) Theiss.	SMH 1291	-	N/A	AF279410	AY641073	N/A	Debashish et al., 2000
Barrmaelia rappazii Jaklitsch, Friebes & Voglmayr	CBS 142771	HT	MF488989	MF488989	MF488998	MF489017	Voglmayr et al., 2017
B. rhamnicola Rappaz	CBS 142772	ET	MF488990	MF488990	MF488999	MF489018	Voglmayr et al., 2017
Biscogniauxia nummularia (Bull.) Kuntze	MUCL 51395	ET	KY610382	KY610427	KY624236	KX271241	Wendt et al., 2018
Cainia anthoxanthis Senan., Camporesi & K.D. Hyde	MFLUCC 15-0539	HT	KR092787	KR092777	N/A	N/A	Senanayake et al., 2015
C. graminis (Niessl) Arx & E. Müll.	MFLUCC15-0540	-	KR092793	KR092781	N/A	N/A	Senanayake et al., 2015
Camillea obularia (Fr.) Læssøe, J.D. Rogers & Lodge	ATCC 28093	-	KY610384	KY610429	KY624238	KX271243	Voglmayr et al., 2017
Chaetomium elatum Kunze	CBS 374.66	-	KC109758	KC109758	KF001820	KC109776	Wang et al., 2016
Clypeosphaeria perfidiosa (De Not.) M.E. Barr	CBS 142773	ET	MF488993	MF488993	MF489003	MF489021	Voglmayr et al., 2017
Coniocessia maxima Asgari & Zare	CBS 593.74	HT	NR137751	MH878275	N/A	N/A	Vu et al., 2019
C. nodulisporioides (D. Hawksw.) Dania García, Stchigel, D. Hawksw. & Guarro	CBS 281.77	IT	MH861061	AJ875224	N/A	N/A	Vu et al., 2019
Creosphaeria sassafras (Schwein.) Y.M. Ju, F. San Martín & J.D. Rogers	STMA14087	-	KY610411	KY610468	KY624265	KX271258	Wendt et al., 2018
Daldinia loculatoides Wollw. & M. Stadler	CBS 113279	ET	MH862918	KY610438	KY624247	KX271246	Wendt et al., 2018
Diatrype palmicola Jian K. Liu & K.D. Hyde	MFLUCC 11-0018	-	KP744439	KP744481	N/A	N/A	Liu et al., 2015
D. whitmanensis J.D. Rogers & Glawe	DCHES100	-	GQ293951	N/A	N/A	GQ294008	Trouillas and Gubler, 2010
Entosordaria quercina Voglmayr & Jaklitsch	CBS 142774	HT	MF488994	MF488994	MF489004	MF489022	Voglmayr et al., 2017
Eutypa flavovirens (Pers.) Tul. & C. Tul.	MFLUCC 13-0625	-	KR092798	KR092774	N/A	N/A	Senanayake et al., 2015
E. lata (Pers.) Tul. & C. Tul.	CBS 208.87	-	MH862066	MH873755	KF453595	DQ006969	Vu et al., 2019

Table. (Continued).

Graphostroma platystomum (Schwein.) Piroz.	CBS 270.87	HT	JX658535	DQ836906	KY624296	HG934108	Voglmayr et al., 2017
Hypoxylon fragiforme (Pers.) J. Kickx f.	MUCL51264	ET	KC477229	KM186295	KM186296	KX271282	Wendt et al., 2018
Jackrogersella multiformis (Fr.) L. Wendt, Kuhnert & M. Stadler	CBS 119016	ET	KC477234	KY610473	KY624290	KX271262	Wendt et al., 2018
Kretzschmaria deusta (Hoffm.) P.M.D. Martin	CBS 163.93	-	KC477237	KY610458	KY624227	KX271251	Stadler et al., 2013
Longiappendispora chromolaenae Mapook & K.D. Hyde	MFLUCC 17-1485	HT	NR_169723	NG_068714	N/A	N/A	Mapook et al., 2020
Lopadostoma americanum Jaklitsch, J. Fourn., J.D. Rogers & Voglmayr	LG8	HT	KC774568	KC774568	KC774525	N/A	Jaklitsch and Voglmayr, 2014
L. dryophilum (Nitschke ex G.H. Otth) Jaklitsch, J. Fourn. & Voglmayr	LG21	HT	KC774570	KC774570	KC774526	MF489023	Jaklitsch and Voglmayr, 2014
L. fagi Jaklitsch, J. Fourn. & Voglmayr	LF1	HT	KC774575	KC774575	KC774531	N/A	Jaklitsch and Voglmayr, 2014
L. quercicola Jaklitsch, J. Fourn. & Voglmayr	LG27	HT	KC774610	KC774610	KC774558	N/A	Jaklitsch and Voglmayr, 2014
Podosordaria muli J.D. Rogers, Y.M. Ju & F. San Martín	WSP 167	HT	GU324761	N/A	GQ853038	GQ844839	Hsieh et al., 2010
Poronia punctata (L.) Fr.	CBS 656.78	HT	KT281904	KY610496	KY624278	KX271281	Senanayake et al., 2015
Sordaria fimicola (Roberge ex Desm.) Ces. & De Not.	CBS 723.96	-	MH862606	MH874231	DQ368647	DQ368618	Vu et al., 2019
Whalleya hainanensis Y.H. Pi & Q.R. Li	GMB0078	HT	OL689196	OL741533	N/A	OL742654	This study
W. hainanensis Y.H. Pi & Q.R. Li	GMB0079	-	OL689197	OL741534	N/A	OL742655	This study
W. microplaca (Berk. & M.A. Curtis) J.D. Rogers, Y.M. Ju & F. San Martín	YMJ 91111215	-	EF026129	N/A	N/A	EF025614	Ju et al., 2007; Hsieh et al., 2010
W. microplaca Y.H. Pi & Q.R. Li	GMB0080	-	OL689194	OL741535	N/A	OL742656	This study
W. microplaca Y.H. Pi & Q.R. Li	GMB0081	-	OL689195	OL741536	N/A	OL742657	This study
Xylaria hypoxylon (L.) Grev.	CBS 122620	ET	KY610407	KY610495	KY624231	KX271279	Wendt et al., 2018
X. polymorpha (Pers.) Grev.	MUCL 49884	-	KY610408	KY610464	KY624288	KX271280	Wendt et al., 2018
Xylariaceae sp.	FL0662	-	JQ760360	N/A	KU684234	KU684151	U'Ren et al., 2016
Xylariaceae sp.	FL0638	-	JQ760345	N/A	KU684231	N/A	U'Ren et al., 2016
Xylariaceae sp.	AK1116	-	JQ759464	N/A	KU684211	KU684113	U'Ren et al., 2016

Notes: Type samples are labeled with HT (holotype), ET (epitype). N/A: not available.

Description: Saprobic on unknown deadwood. **Sexual morph**: **Stromata** 10–40 mm long × 5–18 mm wide × 0.25–0.35 mm high, irregular, flat or slightly undulate, surface dark blackish-grey, thin, carbonaceous crust, the tissue between perithecia is white, coriaceous; lacking KOH-extractable pigments. **Perithecia** 140–240 × 160–260 µm ($\bar{x}$ = 190 × 230 µm, n = 20), obovoid or tubular. **Ostioles** lower than the level of stromatal, with umbilicate and appear as dots on the stromatal surface, black. **Asci** 50–75 × 4–6 µm ($\bar{x}$ = 61.5 × 5 µm, n = 30), 8-spored, unitunicate, cylindrical,

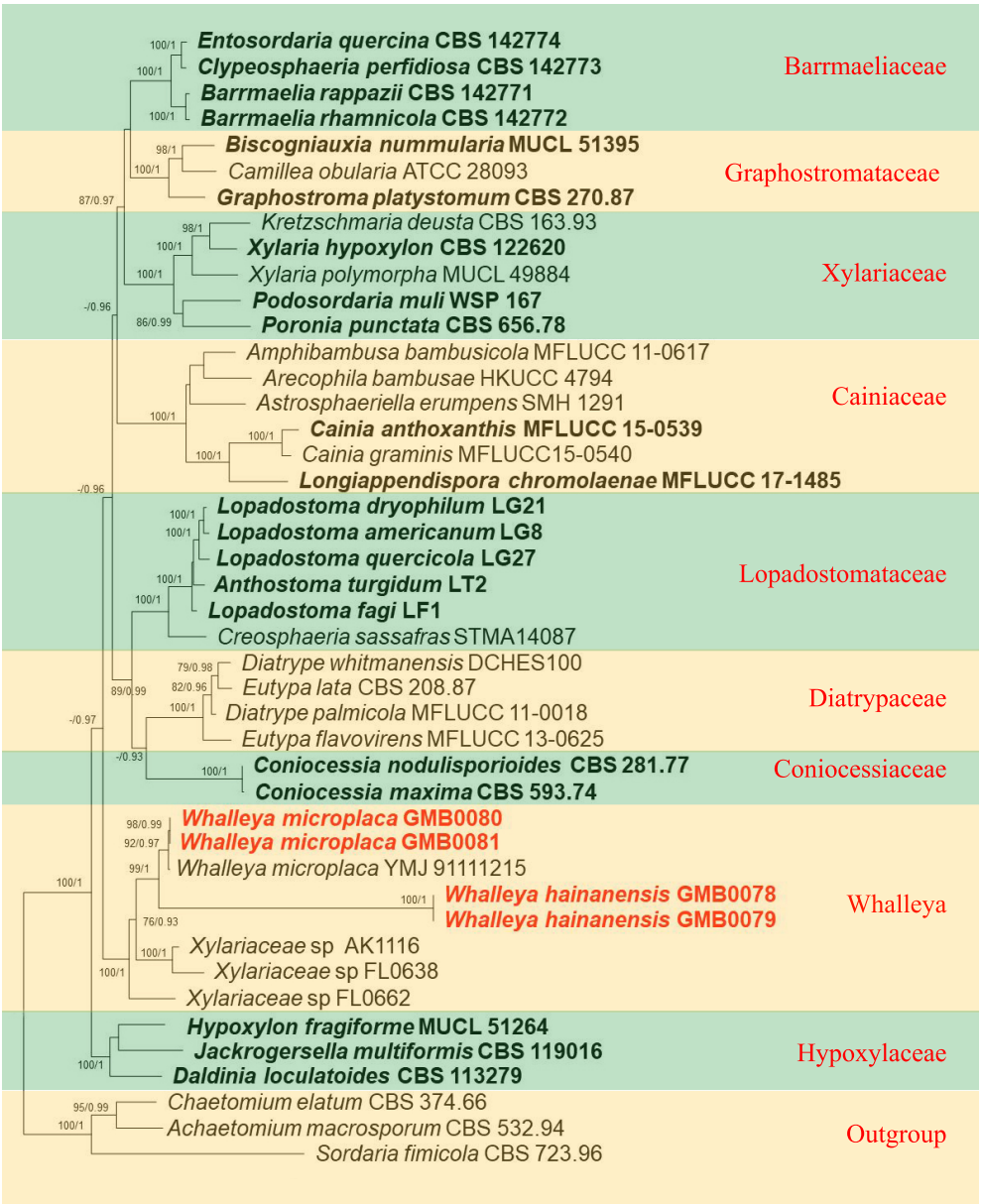

Figure 1. RAxML tree about *Whalleya* and related genera was constructed based on the combination of ITS, LSU, *rpb2* and *tub2* gene sequences. Bootstrap support values for maximum likelihood (ML) > 75% and Bayesian posterior probability (BYPP) > 0.90 are expressed as ML/BYPP and in node, otherwise indicated by dashes (" – "). Type strains are shown in bold and newly generated taxa are shown in red.

short-stipitate, with apparatus, discoid, blue in Melzer's reagent. **Ascospores** 4.5–5.5 × 2–3.5 µm ($\bar{x}$ = 5 × 2.7 µm, n = 30), light to dark brown, ellipsoid-inequilateral, with rounded ends, with an inconspicuous straight germ slit along the entire spore length or half; perispore indehiscent in 10% KOH; epispore smooth.

Culture characteristics: Ascospores germinated and grew on OA for two weeks in an incubator at 25 °C, and the colony diameter reached 5 cm. Colony surface white to the pale yellow, compact, round, flat, rough edge. Colonies were yellow on the reverse. Conidia were not observed on the medium.

Material examined: CHINA, Hainan Province, Wenchang City, Tongguling National Nature Reserve (19°39'38.95"N, 111°0'50.88"E, altitude: 58 m), on unknown dead wood, 11 November 2020, Y.H. Pi and L.L. Liu, 2020TGL8 (GMB0078, holotype; GMBC0078, ex-type living culture; KUN-HKAS 122635, isotype).

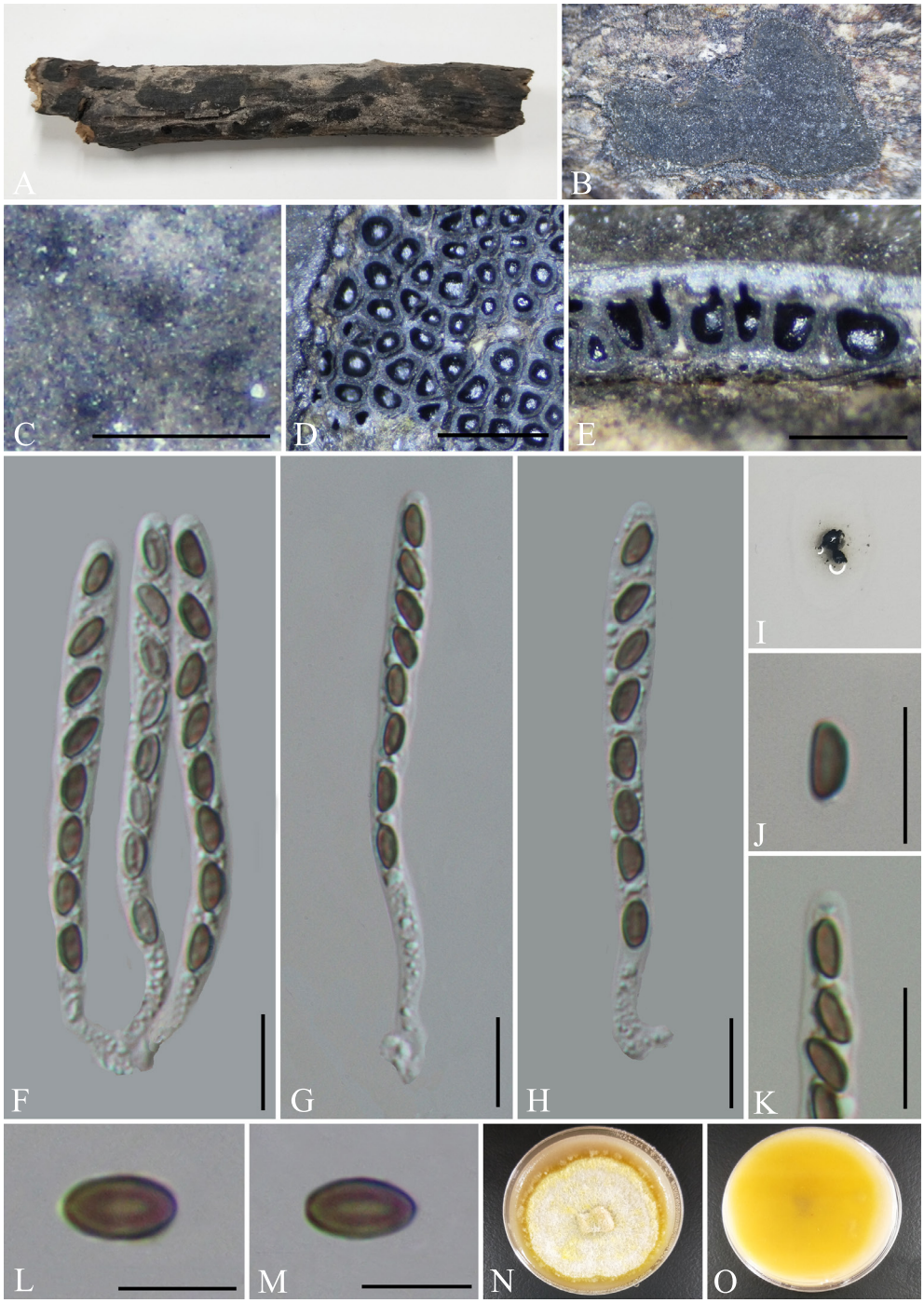

Figure 2. ***Whalleya hainanensis*** (GMB0078, holotype). A. Type material. B. Stroma on the surface of host. C. Ostioles. D. Transverse sections of stroma. E. Longitudinal section of stroma. F–H. Asci. I. No pigments in 10% KOH. J. Ascospores in 10% KOH. K. Asci apical ring (stained in Melzer's reagent). L, M. Ascospores. N, O. Colonies on OA (N-upper, O-lower). Scale bars: 0.5 mm (C–E); 10 μm (F–H, J, K); 5 μm (L, M).

Other examined material: CHINA, Hainan Province, Wenchang City, Tongguling National Nature Reserve (19°39'45.12"N, 111°0'55.41"E, altitude: 88 m), on unknown dead wood, 12 November 2020, Y.H. Pi, 2020TGL81 (GMB0079, holotype; GMBC0079, living culture; KUN-HKAS 122638, isotype).

Notes: In the phylogenetic analysis, *W. hainanensis* formed a distinct branch, which is a sister to *W. microplaca* (Figure 1). *W. hainanensis* resembles *W. microplaca* in having a similar size of ascospores (5 × 2.5 µm), but differs by having obovoid or tubular perithecia rather than globose (500–700 µm diam.) and the perithecia of *W. microplaca* immersed in stromal tissue, arranged in two layers, peridium walls composed of outermost carbonaceous cell layers, thick-walled (Daranagama et al., 2018). Moreover, *W. hainanensis* has slightly raised, widely effused stromata that are significantly higher than the bark surface which distinguish it from *W. microplaca* (Rogers et al., 1997). *W. hainanensis* differs from *W. maculata* by its smaller ascospores (4.5–5.5 × 2–3.5 µm vs. 5–7 × 2–4 µm) (Rogers et al., 1997).

Whalleya microplaca (Berk. & M.A. Curtis) J.D. Rogers, Y.M. Ju & F. San Martín, Mycotaxon 64: 48 (1997)

Synonymy:

Diatrype microplaca Berk. & M.A. Curtis, in Berkeley, J. Linn. Soc., Bot. 10 (no. 46): 386 (1868) [1869].

Anthostoma microplacum (Berk. & M.A. Curtis) Sacc., Syll. fung. (Abellini) 1: 298 (1882).

Nummularia microplaca (Berk. & M.A. Curtis) Cooke, in Winter, Grevillea 12 (no. 61): 8 (1883).

Nummularia scutata Berk. & Cooke, in Winter, Grevillea 12 (no. 61): 7 (1883).

Biscogniauxia scutata (Berk. & Cooke) Kuntze, Revis. gen. pl. (Leipzig) 2: 398 (1891).

Nummularia gracilenta Syd. & P. Syd., Annls mycol. 8 (1): 37 (1910).

Nummularia lamprostoma Syd. & P. Syd., Annls mycol. 18 (1/3): 99 (1920).

Hypoxylon microplacum (Berk. & M.A. Curtis) J.H. Mill., Mycologia 33 (1): 77 (1941).

Hypoxylon lamprostoma (Syd. & P. Syd.) P.M.D. Martin [as 'lamprostomum'], Jl S. Afr. Bot. 33: 319 (1967).

Numulariola microplaca (Berk. & M.A. Curtis) P.M.D. Martin, Jl S. Afr. Bot. 35: 229 (1969).

Hypoxylon lamprostoma (Syd. & P. Syd.) P.M.D. Martin [as 'lamprostomum'], Jl S. Afr. Bot. 42 (1): 72 (1976).

MycoBank No: 437284

Description: Saprobic on unknown deadwood. **Sexual morph**: **Stromata** 5–15 mm long × 4–15 mm wide × 0.3–0.5 mm high, applanate, orbicular, immersed in the surface of the bark, outer dehiscing, solitary, gray-brown, thin, carbonaceous; white or brownish soft tissue between the perithecia, coriaceous; lacking KOH-extractable pigments. **Perithecia** 160–200 × 170–210 µm ($\bar{x}$ = 180 × 195 µm, n = 20), globose, immersed in stromal tissue, arranged in two layers, tissue between ascomata black, peridium walls composed of outermost carbonaceous cell layers, thick-walled. **Ostioles** slightly below the surface of stromatal and appear as dots on the stromatal surface. **Asci** 55–75 × 4–6 µm ($\bar{x}$ = 61.5 × 4.7 µm, n = 30), 8-spored, cylindrical, short-stipitate, with apparatus, discoid, blue in Melzer's reagent. **Ascospores** 4.5–6 × 2–3 µm ($\bar{x}$ = 5 × 2.5 µm, n = 30), light to dark brown, ellipsoid-inequilateral, with narrowly rounded ends, with a straight germ slit 1/2 spore length; perispore indehiscent in 10% KOH; epispore smooth.

Culture characteristics: Ascospores germinated and grew on OA for two weeks in an incubator at 25 °C, and the colony diameter reached 5 cm. Colonies were white, cotton, velvety, round, and thin. Colony reverse center was yellow, irregular, and edge white. Conidia were not observed on the medium.

Material examined: CHINA, Guizhou Province, Kaili City, Leigongshan Nature Reserve (26°22'54.22"N, 108°21'50.86"E, altitude: 886 m), on unknown dead wood, 29 August 2020, Y.H. Pi, 2020LGS84 (GMB0080, KUN-HKAS 122636), living culture, GMBC0080; CHINA, Guizhou Province, Qiandongnan Miao and Dong Autonomous Prefecture, Kaili City, Leigongshan Nature Reserve (26°20'28.35"N, 108°17'19.12"E, altitude: 840 m), on unknown dead wood, 30 August 2020, Y.H. Pi, 2020LGS193 (GMB0081, KUN-HKAS 122637), living culture, GMBC0081.

Notes: New collections (GMB0080 and GMB0081) are morphologically consistent with *W. microplaca* having applanate carbonaceous stromata immersed in the surface of the host, light brown ellipsoid-inequilateral ascospores (4–6 × 2–3 µm) with rounded ends and a straight germ slit (Rogers et al., 1997). In phylogenetic analyses, new collections clearly show a close affiliation with *W. microplaca* (YMJ 91111215) (100% ML, 1 BYPP, Figure 1).

4. Discussion

Whalleya microplaca as the type species has been widely distributed in China, USA, Mauritius, and the Philippines (Glawe and Rogers, 1986; Rogers et al., 1997; Daranagama et al., 2018). While *W. maculata* has only been reported in Brazil, not domestically (Rogers et al., 1997). *Whalleya* only accepts two species currently, few morphological differences were found between these two species, and there may be many cryptic species. In this paper, we identified a novel species of *Whalleya* and a known species. It was found that the difference between our new species and those two species of *Whalleya* reported so far is very small and difficult to distinguish by morphological characteristics alone. It is difficult to use the germ slit on the ascospore as a trait for the species identification as the ascospore of *Whalleya* are small. The molecular data, however, can distinguish them.

Currently, there are only 2 morphologic species in the genus of *Whalleya* (Species Fungorum 2022) and 1 species

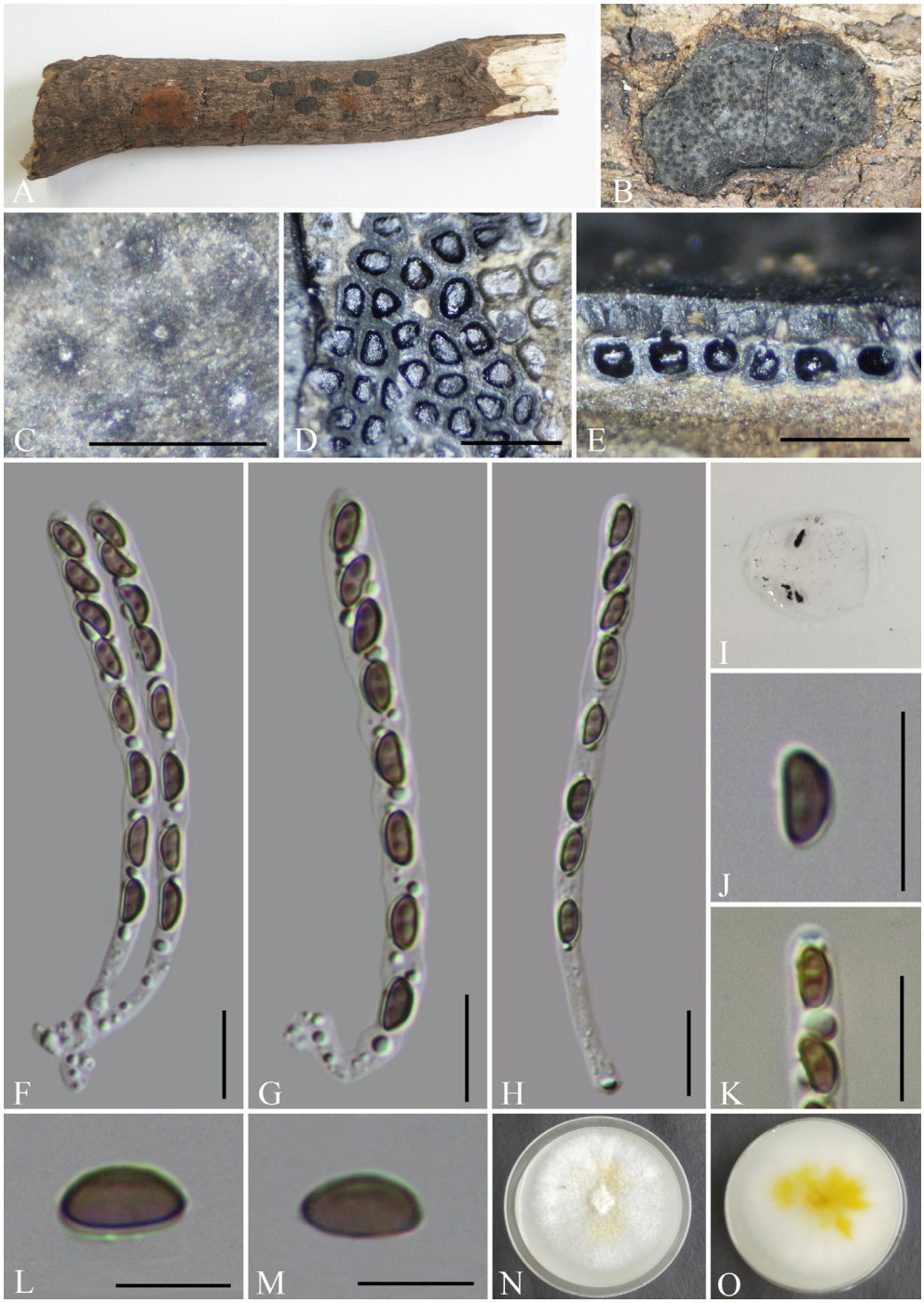

Figure 3. ***Whalleya microplaca*** (GMB0080). A. Material examined. B. Stroma on the surface of host. C. Ostioles. D. Transverse sections of stroma. E. Longitudinal section of stromata. F–H. Asci. I. No pigments in 10% KOH. J. Ascospore in 10% KOH. K. Asci apical ring (stained in Melzer's reagent). L, M. Ascospores. N, O. Colonies on OA (N-upper, O-lower). Scale bars: 0.5 mm (C–E); 10 µm (F–H, J, K); 5 µm (L, M).

with sequence data. *W. Microplaca*, published in 1997, there was no type strain sequence. However, YMJ91111215 is designed as the authority strain of *W. microplaca*. YMJ91111215 was identified and introduced by Ju Y.M. and Rogers J.D who originally established the genus of *Whalleya* and the specie of *W. microplaca* (Rogers et al.,

1997; Ju et al., 2007; Maharachchikumbura et al., 2016; Daranagama et al., 2018; Hyde et al., 2020). Moreover, the sequence of the type strain did not appear in their later studies. We have reason to think that YMJ91111215 can represent *W. microplaca*. In molecular analysis, *W. hainanensis* forms a separate evolutionary branch which is different from *W. microplaca*.

Whalleya was classified into *Lopadostomataceae* by Daranagama et al. (2018). However, Figure 1 shows that *Whalleya* does not fall in *Lopadostomataceae* with *Lopadostoma* (Nitschke) Traverso and *Creosphaeria* Theiss. *Whalleya* formed a single branch with a support rate (100/1), which contained endophytic fungi. The affiliation of *Whalleya* with other families of *Xylariales* is uncertain as to the low support values of the node connecting *Whalleya* branch and others. The position of *Whalleya* clade in the phylogenetic tree is unstable, and the taxonomic status of this genus should be further studied in the future.

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