

Three newly recorded wood-decay fungi in subtropical montane cloud forests of Taiwan and a field observation of fungal feeding by the endemic Swinhoe's pheasant

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ABSTRACT

Three wood-decay polypores, *Buglossoporus eucalypticola*, *Foraminispora concentrica*, and *Steccherinum subcollabens*, are newly recorded in subtropical montane cloud forests of Taiwan. Detailed descriptions and illustrations are provided. Phylogenetic analyses using internal transcribed spacer (ITS rDNA) sequences were conducted to confirm our morphology-based identifications. Additionally, we document the endemic male Swinhoe's pheasant (*Lophura swinhoii*) feeding on *B. eucalypticola*, highlighting the ecological significance of this fungal species in its native habitat.

Key words: fungal diversity, Basidiomycota, phylogeny, Polyporales, mycophagy, taxonomy

Introduction

Subtropical montane cloud forests (SMCFs) are unique and fragile ecosystems frequently immersed in mist and low clouds, resulting in high air humidity and lower temperatures (Bruijnzeel et al. 2010). In Taiwan, SMCFs are situated between elevations of 1,400 and 2,600 meters above sea level (m a.s.l.) (Su 1984; Li et al. 2013), which are home to diverse fungal species. Wood-decay fungi play an important role in nutrient recycling within SMCFs by decomposing cellulose,

hemicelluloses, and/or lignin from woody biomass (Härkönen et al. 2015; Tura et al. 2016).

In this study, we report three wood-decay polypore species within the order Polyporales (Basidiomycota): *Buglossoporus eucalypticola*, *Foraminispora concentrica*, and *Steccherinum subcollabens*. These species are newly recorded for Taiwan's SMCFs based on morphological and molecular characterizations. Additionally, we document an interaction between *B. eucalypticola* and the endemic male Swinhoe's pheasant (*Lophura swinhoii*), highlighting the ecological

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#Co-first authorship

Table 1. Species and sequences used in phylogenetic analyses. Newly generated sequences are shown in boldface.

Name	Samples	ITS	Locality
<i>Antrodia serialis</i>	KHL 12010 (GB)	JX109844	Norway
<i>Buglossoporus eucalypticola</i>	Dai 13660 (holotype)	KR605808	China
<i>Buglossoporus eucalypticola</i>	GC 1805-1	PQ328929	Taiwan
<i>Buglossoporus eucalypticola</i>	GC 1805-16	PQ328930	Taiwan
<i>Buglossoporus eucalypticola</i>	GC 1805-18	PQ328931	Taiwan
<i>Buglossoporus eucalypticola</i>	GC 2103-1	PQ328935	Taiwan
<i>Datronia mollis</i>	RLG6304sp	JN165002	USA: Montana
<i>Fomitopsis pinicola</i>	AFTOL-770	AY854083	NA
<i>Foraminispora concentrica</i>	Cui 12644 (holotype)	MK119812	China
<i>Foraminispora concentrica</i>	Cui 12646	MK119813	China
<i>Foraminispora concentrica</i>	Wu 0307-12	PQ328932	Taiwan
<i>Gelatoporia subvermispora</i>	FD-354	KP135312	NA
<i>Heterobasidion annosum</i>	AFTOL-ID 470	DQ206988	NA
<i>Hyphoderma setigerum</i>	FD-312	KP135297	USA: Massachusetts
<i>Irpex lacteus</i>	DO 421/951208	JX109852	Sweden
<i>Steccherinum subcollabens</i>	Dai 19344	MN871758	China
<i>Steccherinum subcollabens</i>	Dai 19345 (holotype)	MN871759	China
<i>Steccherinum subcollabens</i>	GC 2307-10	PQ328933	Taiwan
<i>Steccherinum subcollabens</i>	GC 2307-12	PQ328934	Taiwan
<i>Lopharia cinerascens</i>	FP-105043-sp	JN165019	USA: Mississippi
<i>Obba rivulosa</i>	FP-135416-Sp	KP135309	USA: Idaho
<i>Phanerochaete chrysosporium</i>	PC139	MZ422797	Taiwan
<i>Skeletocutis nivea</i>	ES2008-1 (GB)	JX109858	Sweden
<i>Steccherinum ochraceum</i>	KHL 11902 (GB)	JQ031130	Sweden
<i>Stereum hirsutum</i>	AFTOL-ID 492	AY854063	NA
<i>Trametes versicolor</i>	FP-135156-sp	JN164919	USA: New York
<i>Tyromyces chioneus</i>	FD-4	KP135311	USA: Massachusetts

significance of this fungal species in its native habitat.

Materials and Methods

Morphological studies

All studied specimens were deposited at the herbarium of the National Museum of Natural Sci-

ence (TNM), Taichung, Taiwan. Free-hand thin sections of fruiting bodies were prepared under an Olympus SZ51 stereomicroscope (Olympus, Tokyo, Japan) and mounted in 5% potassium hydroxide (KOH) with 1% phloxine for observation and measurements, and in Melzer's reagent (IKI) to test amyloidity and dextrinoidity. IKI- indicates both inamyloid and indextrinoid reac-

tions. The prepared slides were examined using a Leica DM2500 microscope (Leica, Wetzlar, Germany) at magnifications up to 1000 \times .

DNA extraction, amplification and sequencing

DNA was extracted from fungal cultures grown on malt extract agar (MEA) or from fruiting bodies using the NautiaZ Plant DNA Extraction Mini Kit (Nautia Gene, Taipei, Taiwan) according to the manufacturer's instructions. The internal transcribed spacer (ITS rDNA) region was amplified using primers ITS1F and ITS4 (White et al. 1990). Polymerase chain reaction (PCR) conditions were as follows: initial denaturation at

94 °C for 5 min; followed by 35 cycles of denaturation at 94 °C for 30 s, annealing at 55 °C for 30 s, and extension at 72 °C for 1 min; and a final extension at 72 °C for 7 min. PCR products were purified and directly sequenced by Genomics BioSci & Tech (New Taipei, Taiwan). Newly generated sequences were manually assembled using BioEdit v. 7.2.5 (Hall 1999).

Phylogenetic analysis

An ITS-based phylogenetic analysis using the Maximum Likelihood (ML) method was conducted to confirm the morphological identifications of *Buglossoporus eucalypticola*, *Foraminispora concentrica*, and *Steccherinum subcollabens*

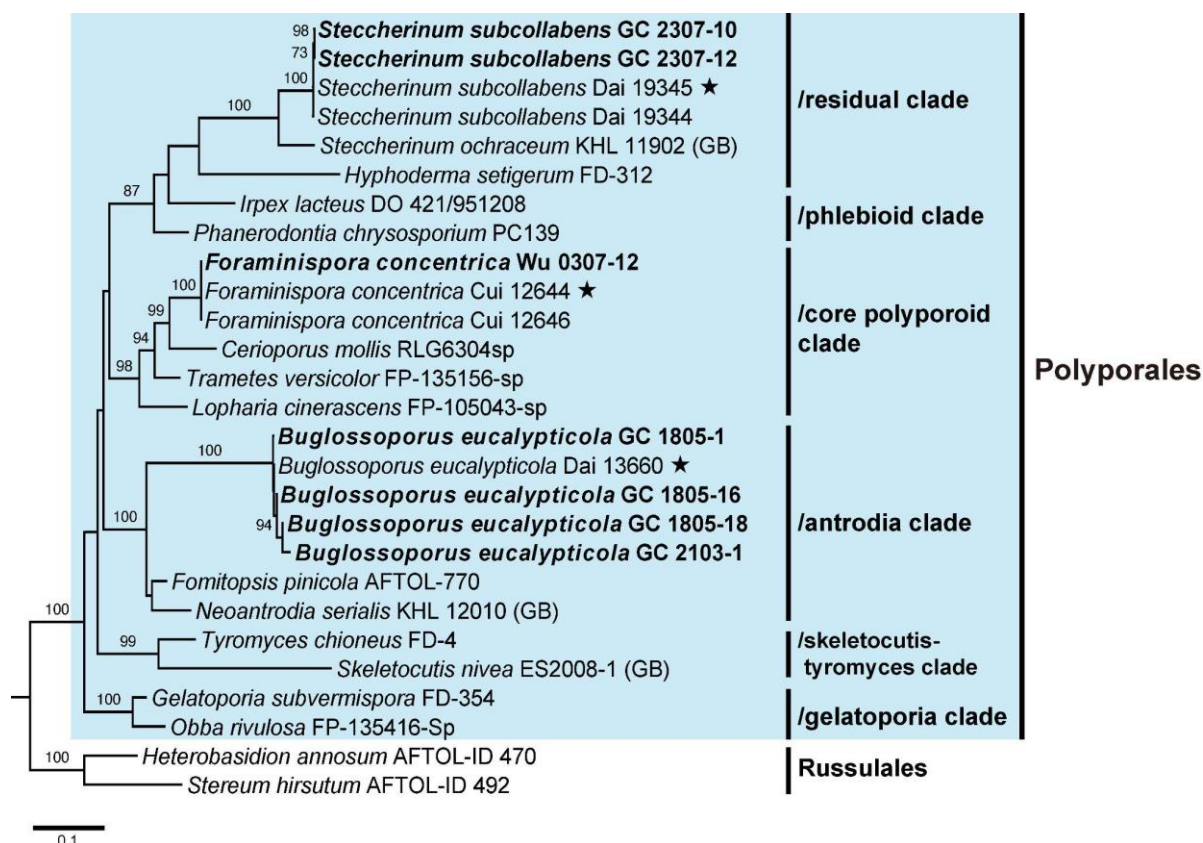


Fig. 1. The maximum likelihood (ML) phylogenetic tree, reconstructed using ITS rDNA gene sequences, reveals the placement of *Buglossoporus eucalypticola*, *Foraminispora concentrica*, and *Steccherinum subcollabens* within the order Polyporales. Bootstrap values ($\geq 70\%$), based on 1,000 replications, are labeled on branch nodes. Black stars (★) represent holotypes. Newly generated sequences in this study are shown in bold. Scale bar = substitutions per site.

bens. Newly generated sequences, along with representative sequences retrieved from GenBank (NCBI) (Table 1), were aligned using MAFFT v. 7 and manually adjusted with BioEdit v. 7.2.5. The ML analyses were performed using RAxML BlackBox (Stamatakis et al. 2014) on the CIPRES Science Gateway (<http://www.phylo.org/>), following the parameter settings of Chen et al. (2021).

RESULTS

Phylogenetic analysis

We generated seven new ITS sequences (GenBank accession numbers PQ328929–PQ328935) representing three species and seven specimens from Taiwan. The ITS dataset comprised 27 sequences from 18 species (Table 1). The final alignment included 727 positions with gaps. A best-scoring maximum likelihood (ML) tree with proportional bootstrap value (BS) was reconstructed (Fig. 1), revealing six main clades within the Polyporales ingroup: the antrodia clade, the core polyporoid clade, the gelatoporia clade, the phlebioid clade, the residual clade, and the skeletoncutis-tyromyces clade. Within the Polyporales, *Buglossoporus eucalypticola*, *Foraminispora concentrica*, and *Steccherinum subcollabens* belong to the antrodia clade, the core polyporoid clade, and the residual clade, respectively. The Taiwanese sequences of these three species all clustered with the holotype sequences from China with high support (BS = 100, Fig. 1).

Taxonomy

Buglossoporus eucalypticola M. L. Han, B. K. Cui & Y. C. Dai, in Han, Chen, Shen, Song,

Vlasák, Dai & Cui, Fungal Diversity 80 351. 2016. Fig. 2

Basidiomata up to 7 mm thick at the base, up to 17.5 cm wide, projecting up to 11.5 cm, annual, solitary, pileate, usually with a central to lateral stipe. Pileus appanate to slightly convex, flabelliform or semicircular with a series of inward-pointing notches; pileal surface peach to brownish orange when fresh, becoming cinnamon when dry, usually with a pellicle, glabrous, azonate, rugose; margin flesh-pink when fresh, becoming cinnamon to dark brown when dry. Pore surface white when fresh, turning dark brown when dry; pores 2–3 per mm, round to angular; tubes up to 0.5 mm deep, concolorous with pore surface; dissepiments up to 5.5 μm thick, entire. Context thicker than tubes, up to 7.5 mm, tissues becoming orange in KOH. Stipe glabrous, often with a pellicle, cream to dark brown, up to 9 cm long and 2.5 cm thick. Hyphal system dimitic in context, monomitc in trama; generative hyphae with clamp connections, IKI-. In context, generative hyphae 2.5–9 μm diam, dominant, colorless, occasionally branched, interwoven; skeletal hyphae infrequent, 3.2–5.6 μm diam, thick-walled with a wide lumen. In trama, generative hyphae 1.6–3.8 μm diam, colorless, thin-walled, moderately branched. Cystidia absent; cystidioles present, fusoid, colorless, thin-walled, 11–34 \times 2.8–4 μm . Basidia 18–37 \times 4–6.5 μm , clavate, 4-sterigmate, bearing a basal clamp connection; basidioles similar in shape to basidia but slightly smaller. Basidiospores mostly 5.4–6.4 \times 2.1–2.8 μm , cylindrical to fusiform, colorless, tapering at the apex, smooth, thin-walled, mostly bearing one guttule, IKI-.

Specimens examined. TAIWAN. Taichung

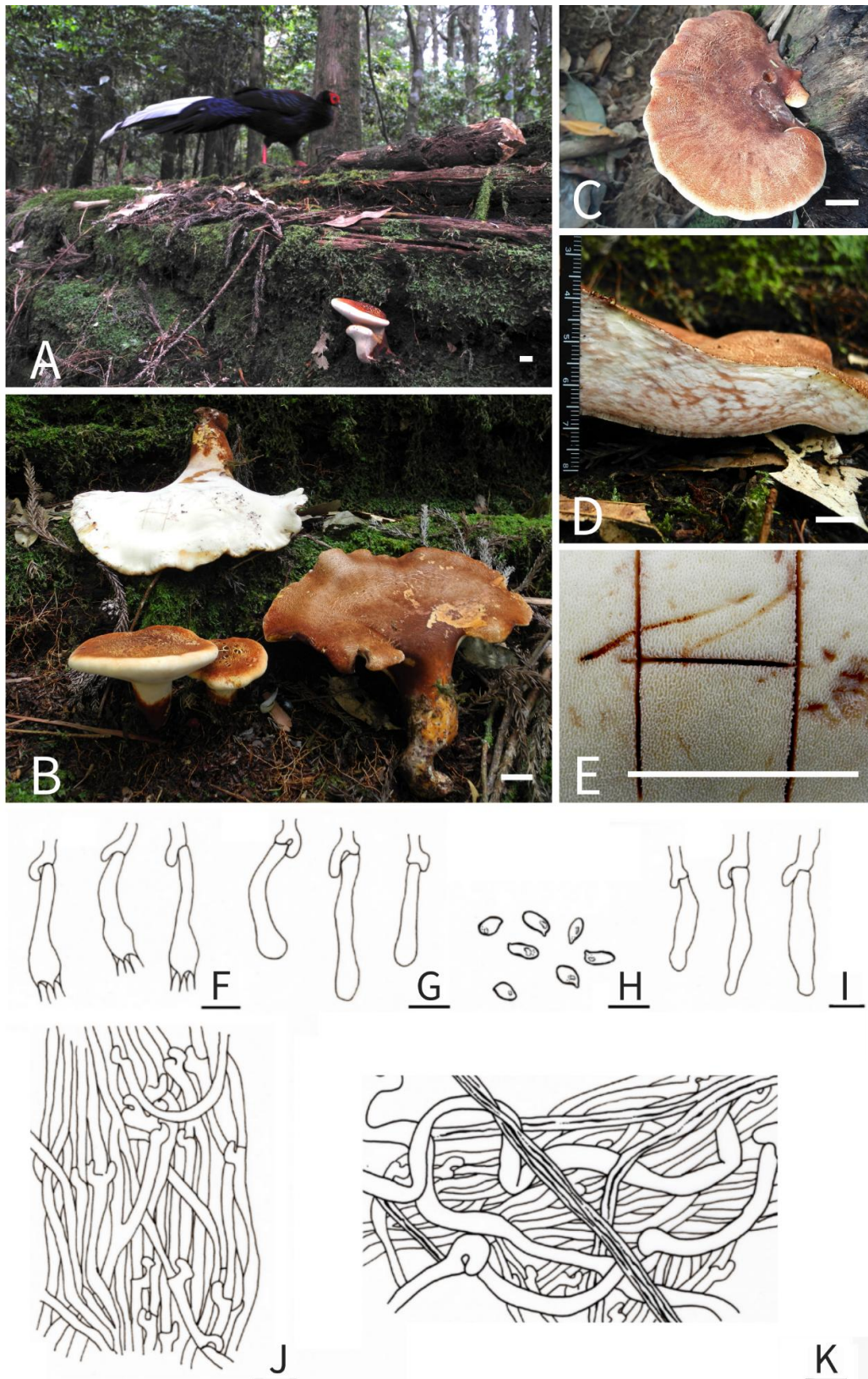


Fig. 2. *Buglossoporus eucalypticola* (from GC 1805-1 except for C, which is from GC 2103-1). A–C. Basidiomata in native habitat. D. Lateral view of basidiomata. E. Pore surface showing red bruises when injured. F. Basidia. G. Basidioles. H. Basidiospores. I. Cystidioles. J. Hyphae from trama. K. Hyphae from context. Scale bars: A–E = 1 cm; F–K = 10 μ m.

City, Hoping District, between 19–20 km of Dasyueshan Forestry Road, on fallen trunk of wood, 6 May 2018, leg. C.C. Chen & C.M. Hu, *GC 1805-1* (TNM F0033499); *ibid.*, 19 km of Dasyueshan Forestry Road, on fallen trunk of wood, 12 May 2018, leg. C.C. Chen, *GC 1805-16* (TNM F0033511); *ibid.*, between 32–33 km of Dasyueshan Forestry Road, on living trunk of Fagaceae sp., 20 May 2018, leg. C.M. Hu, *GC 1805-18* (TNM F0033513); Hsinchu County, Jianshi Township, Ptlaman, on wood, 28 Mar 2021, leg. Y.Y. Tang, *GC 2103-1* (TNM F0036739).

Ecology and distribution. On dead or living angiosperm wood (e.g., *Eucalyptus* and Fagaceae). Southern China (Hainan) (Han et al. 2016) and Taiwan (this study). Occurring in March, May, and June.

Notes. The morphology of our specimens aligns well with the protologue of *Buglossoporus eucalypticola* (Han et al. 2016). While Han et al. (2016) reported that *B. eucalypticola* grows exclusively on dead *Eucalyptus* trees, it also grows on the living trunk of Fagaceae species in Taiwan (e.g., *GC 1805-18*). *B. eucalypticola* resembles *B. malesianus* Corner and *B. quercinus* (Schrad.) Kotl. & Pouzar in having annual, pileate basidiomata with a dimitic hyphal system in context, monomitic in trama, and cylindrical to fusiform basidiospores. However, *B. eucalypticola* differs from *B. malesianus* by its pileal surface usually with a pellicle and slightly smaller basidiospores ($5.5\text{--}7 \times 2.5\text{--}3.2 \mu\text{m}$ in *B. malesianus*; Hattori 2000). It also differs from *B. quercinus* by having a pileal surface with a pellicle and smaller basidiospores ($6\text{--}8 \times 2.5\text{--}3.5 \mu\text{m}$ in *B. quercinus*;

Ryvarden and Melo 2017, as *Piptoporus quercinus* (Schrad.) P. Karst.).

Between 2 and 3 pm on 6 May 2018, in a coniferous and broadleaved mixed forest within the Dasyueshan Forest Recreation Area, Taichung, Taiwan, we observed a male Swinhoe's pheasant (*Lophura swinhoii*), an endemic bird in Taiwan, foraging on the forest floor. The bird was attracted by the fragrance of the basidioma of *B. eucalypticola* growing on a fallen trunk and subsequently fed on it (Fig. 2A). Swinhoe's pheasants have diverse omnivorous diets, including acorns, berries, leaves, earthworms, millipedes and insects (McGowan et al., 2020). To our knowledge, this is the first report of a Swinhoe's pheasant (*Lophura swinhoii*) feeding on fungi. Fungi are rich in amino acids, protein, and water, making them valuable seasonal food sources (Miles and Chang 2004). *B. eucalypticola* may provide essential nutrition to the pheasant during specific seasons, highlighting its conservation value.

Foraminispora concentrica (J. Song, X. L. He & B. K. Cui) Y. F. Sun & B. K. Cui in Sun, Costa-Rezende, Xing, Zhou, Zhang, Gibertoni, Gates, Glen, Dai & Cui, *Persoonia* 44: 218.2020.

Fig. 3
≡ *Amauroderma concentricum* J. Song, X. L. He & B. K. Cui in Song, Xing, Decock, He & Cui, *Phytotaxa* 260: 47. 2016.

Basidiomata centrally to laterally stipitate. Pileal surface yellowish brown, tomentose, with prominent concentric zones and radial wrinkles. Pore surface pale yellow to straw color when dry; pores circular to angular, 4–5 per mm. Tubes 60–

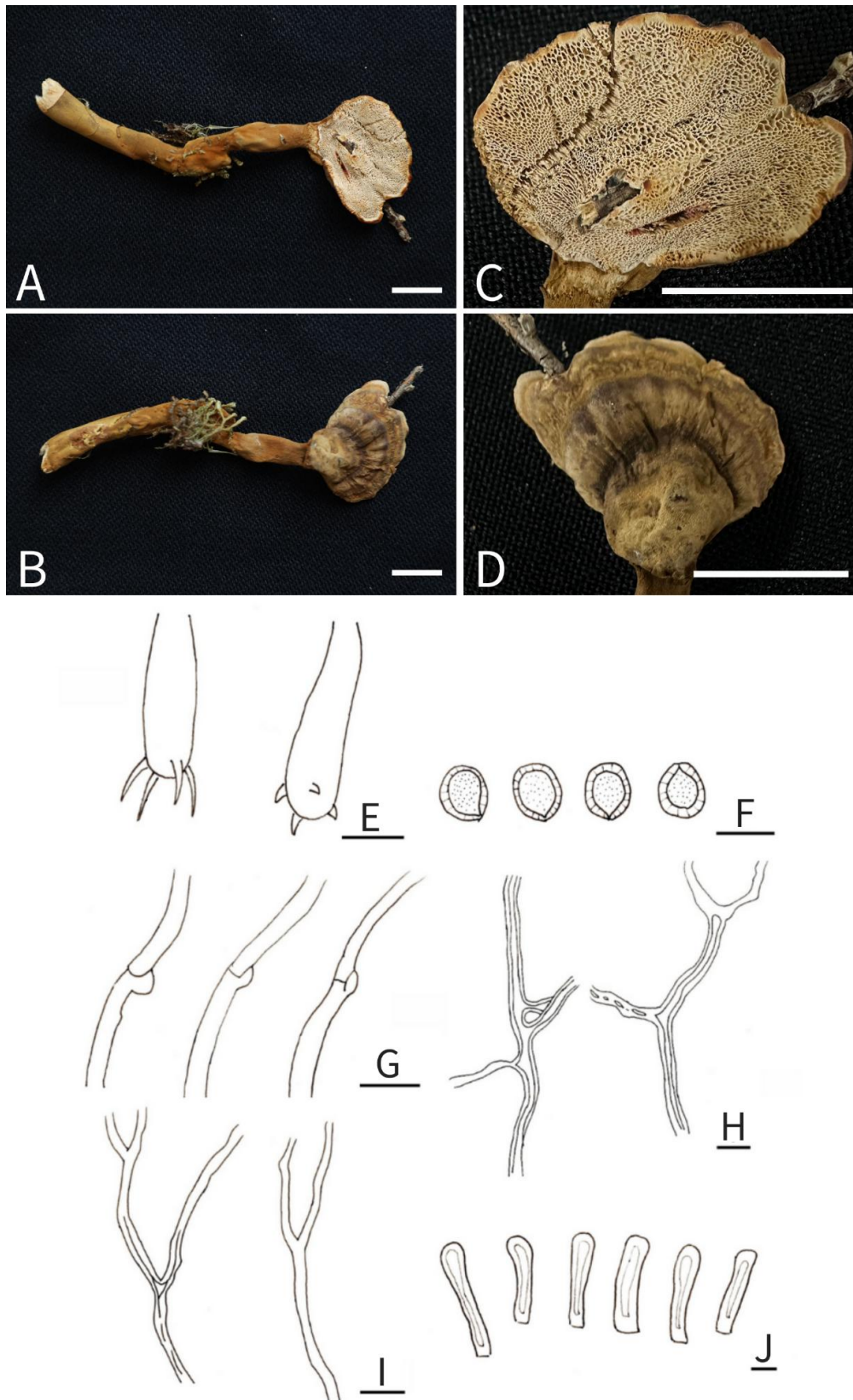


Fig. 3. *Foraminispora concentrica* (Wu 0307-12). A–B. Basidiomata in dried condition. C. Pore surface. D. Pileal surface. E. Basidia. F. Basidiospores. G. Generative hyphae. H. Skeletal hyphae. I. Binding hyphae. J. Terminal cells of pileipellis. Scale bars: A–D = 1 cm; E–J = 10 μ m.

75 μm thick. Context white to cream. Stipe concolorous with pileal surface. Tubes up to 50 μm deep, concolorous with pore surface. Hyphal system trimitic; generative hyphae with clamp connections, all hyphae IKI-. In context, generative hyphae 3 μm diam, colorless, thin-walled; skeletal hyphae 2–4 μm diam, colorless, thick-walled with a wide to narrow lumen or subsolid, branched; binding hyphae 1–2 μm diam, colorless, with narrow lumen or subsolid, branched. Pileal cover composed of thick-walled generative hyphae; apical cells clavate, inflated or constricted, yellowish brown, forming a regular palisade. Cystidia and cystidioles absent. Basidia clavate, colorless, thin-walled; basidioles similar in shape to basidia, colorless, thin-walled. Basidiospores 5.5–8 \times 5–7 μm , subglobose to broadly ellipsoid, pale yellow, slightly dextrinoid, with double and distinctly thick walls; exospore wall smooth; endospore wall with conspicuous echinules.

Specimens examined. TAIWAN. Taichung City, Hoping District, Anmashan, 24°16'N, 121°00'E, on the ground associated with mosses, 3 Jul 2003, leg. S.H. Wu, *Wu 0307-11* (TNM F0015194); *ibid.*, *Wu 0307-12* (TNM F0015195).

Ecology and distribution. On ground of broad-leaved forests, or on angiosperm stump. Subtropical to temperate regions of southwestern China (Sichuan, Yunnan, Guizhou) (Song et al. 2016; Sun et al. 2020) and Taiwan (this study). Occurring in July, September, and October.

Notes. *Foraminispora concentrica* was originally placed in *Amauroderma* (i.e., *A. concentricum*) due to its yellowish-brown to reddish-

brown pileal surface with concentric zones, single stipe, and subglobose, double-walled basidiospores (Song et al. 2016). Sun et al. (2020) later transferred this species to *Foraminispora* based on phylogenetic analyses. Morphologically, our specimens of *F. concentrica* align with the protologue (Song et al. 2016) in most respects. However, they have smaller basidiospores (5.5–8 \times 5–7 μm vs. 8–9.2 \times 7.2–8 μm in Song et al. 2016), likely due to immature basidiomata. This species resembles *F. austrosinensis* (J.D. Zhao & L.W. Hsu) Y.F. Sun & B.K. Cui in having yellowish-brown, tomentose pilei with distinct concentric zones. However, *F. austrosinensis* can be distinguished by its smaller pores (6–7 per mm) and indextrinoid basidiospores (Sun et al. 2020).

Steccherinum subcollabens (F. Wu, P. Du & X. M. Tian) Z. B. Liu & Y. C. Dai, *Phytotaxa* 483: 9. 2021.

Fig. 4

\equiv *Junghuhnia subcollabens* F. Wu, P. Du & X.M. Tian, in Du, Wu & Tian, *MycoKeys* 72: 10. 2020.

Basidiomata annual, resupinate. Pore surface pinkish-buff; pores round to angular, 11–12 per mm; dissepiments 50–80 μm thick. Subiculum brown, darker than pores. Tubes up to 2.5 mm deep, brown, distinctly darker than pore surface. Hyphal system dimitic; generative hyphae with clamp connections; skeletal hyphae IKI-. In context, dominated by skeletal hyphae: generative hyphae 2.5–3 μm diam, colorless, thin- to fairly thick-walled, frequently branched; skeletal hyphae 2.5–3 μm diam, thick-walled with a wide to narrow lumen, flexuous, occasionally branched, more or less gelatinized, interwoven. In trama: generative hyphae 2–2.5 μm diam, colorless, frequently branched, with both simple septa and

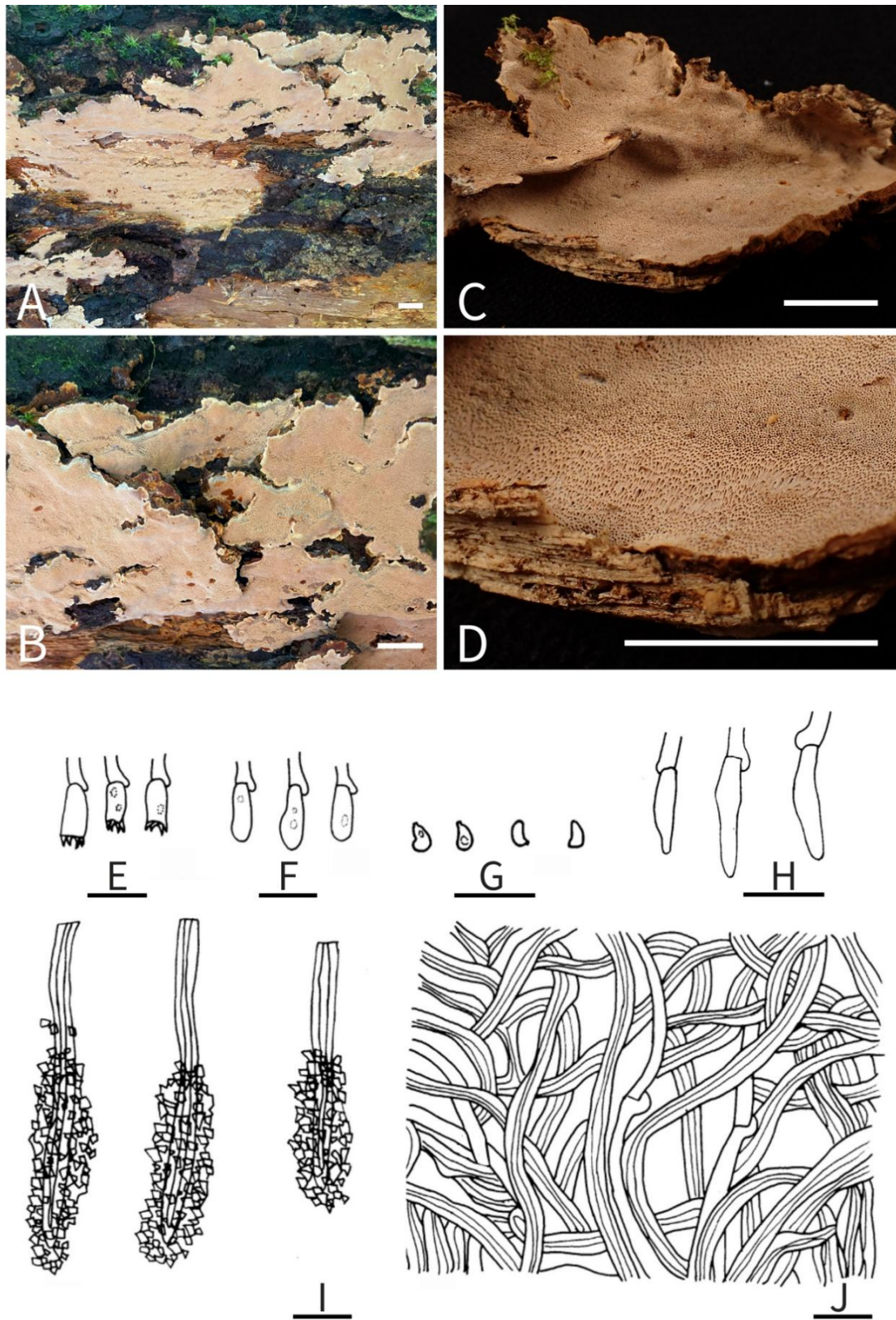


Fig. 4. *Steccherinum subcollabens* (GC 2307-10). A–B. Basidiomata in natural habit. C. Basidiomata in dried condition. D. Pore surface. E. Basidia. F. Basidioles. G. Basidiospores. H. Cystidioles. I. Skeletocystidia. J. Hyphae from trama. Scale bars: A–D = 1 cm; E–J = 10 μ m.

clamp connections (simple septa especially common at dissepiment edges); skeletal hyphae 2.5–3 μm diam, thick-walled with a wide to narrow lumen, rarely branched, flexuous, interwoven. Skeletocystidia 32–52 \times 9–11 μm , clavate, thick-walled, originating from trama, apex covered with crystals. Cystidioles 15–18 \times 3–4.2 μm , fusoid. Basidia clavate, bearing four sterigmata and a basal clamp connection; basidioles 10.5–11 \times 4–4.3 μm , similar in shape to basidia but smaller. Basidiospores 2–3.1 \times 1.2–1.5 μm , mostly ellipsoidal to lunate, colorless, thin-walled, sometimes with one or two small guttules, IKI-

Specimens examined. TAIWAN. Taichung City, Hoping District, Dasyueshan National Forest Recreation Area, Forest Trail, 24°15'26"N, 121°00'28"E, on rotten gymnosperm trunk, 13 Jul 2023, leg. J.J. Tung, P.Y. Chen, S.Y. Wu, W.Y. Chen, W.Y. Liao, *GC 2307-10* (TNM F0038152); *ibid.*, *GC 2307-12* (TNM F0038153).

Ecology and distribution. On rotten angiosperm or gymnosperm wood. In warm temperate forests of southwestern China (Yunnan) (Du et al. 2020) and Taiwan (this study). Occurring in Jul, Nov.

Notes. *Steccherinum subcollabens* was originally described under the genus *Junghuhnia* (i.e., *J. subcollabens*) (Du et al. 2020) and was recently transferred to *Steccherinum* based on phylogenetic analyses (Liu and Dai 2021). Morphologically, our specimens of *S. subcollabens* align well with the protologue (Du et al. 2020). It resembles *Steccherinum collabens* (Fr.) Vesterh. in having pinkish-buff pore surfaces and the

presence of skeletocystidia. However, *S. collabens* has larger pores (6–8 per mm) and longer basidiospores (3.2–3.6 \times 1.4–1.7 μm) (Niemelä 2016).

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臺灣亞熱帶山地雲霧森林中新記錄的三種木材腐朽真菌及 臺灣特有種鳥類藍腹鵲取食真菌的野外觀察

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摘 要

臺灣亞熱帶山地雲霧森林中首次記錄到三種木腐多孔菌：椴牛舌孔菌 (*Buglossoporus eucalypticola*)、環心空洞孢芝 (*Foraminispora concentrica*) 及亞皺齒耳菌 (*Steccherinum subcollabens*)。本文提供此三種菌詳細的特徵描述及繪圖，並使用核糖體核酸 (rDNA) 內轉錄間隔區 (internal transcribed spacers, ITS) 序列進行親緣關係分析以確定形態鑑定結果。此外，記錄臺灣特有種藍腹鵲 (*Lophura swinhoii*) 雄鳥取食椴牛舌孔菌的行為，突顯此菌在其原生棲地中的生態重要性。

關鍵詞：真菌多樣性、擔子菌門、親緣關係、多孔菌目、食菌行為、分類學