



Full paper

A new species of *Fusichalara* (Sclerococcaceae, Eurotiomycetes) from Taiwan

Chang-Hsin Kuo^a, Sung-Yuan Hsieh^b, Teik-Khiang Goh^{a*}^a Department of Plant Medicine, National Chiayi University, 300 Syuefu Road, Chiayi City 60004, Taiwan.^b Bioresource Collection and Research Centre, Food Industry Research and Development Institute, No. 331, Shihpin Road, Hsinchu City 300, Taiwan.

ABSTRACT

Fusichalara pallida sp. nov. is described from decaying wood submerged in a freshwater stream in Taiwan. The phylogenetic relationship of *Fusichalara* species was sought among representative taxa from related fungal lineages, namely the *Chaetosphaeriales* and *Glomerellales* in the *Sordariomycetes*, and various other ordinal groups in the *Eurotiomycetes*, by comparing the concatenated ITS and LSU sequences of their nuc rDNA. The novel *Fusichalara* species from Taiwan clustered with *F. minuta* within the *Sclerococcales* besides other ordinal groups in the *Eurotiomycetes*. Morphologically, *F. pallida* is comparable with *F. dimorphospora* and *F. novae-zelandiae* in having long-cylindrical first-formed conidia and fusiform subsequent conidia with paler end cells, however, they differ in conidial dimensions. With the addition of this novel taxon, *Fusichalara* now comprises seven species. A synopsis of these species and a composite illustration of their conidial morphology are given to ease identification.

Keywords: Asexual freshwater fungi, dimorphic conidia, lignicolous fungi, phialoconidia, phylogeny

Article history: Received 22 January 2024, Revised 20 May 2024, Accepted 22 May 2024, Available online 29 June 2024.

1. Introduction

This paper is a taxonomic treatment of a *Fusichalara* strain collected from Taiwan that could not be identified and provides its phylogenetic placement.

Fusichalara is a small genus currently comprising six species (Index Fungorum, 2024). It was established by Hughes and Nag Raj (1973) for three *Chalara*-like anamorphic fungi: *F. dimorphospora* S. Hughes & Nag Raj (the type species), *F. dingleyae* S. Hughes & Nag Raj, and *F. novae-zelandiae* S. Hughes & Nag Raj. They are lignicolous, dematiaceous hyphomycetes with cylindrical phialides but differ from *Chalara* in the presence of a pronounced wall-thickening inside the phialide at the point of transition from venter to collarette, and in producing two morphologically different kinds of phialoconidia; the first-formed conidium is cylindrical whereas those produced subsequently are fusiform or sigmoid (Hughes & Nag Raj, 1973). Three other species, all with hyaline septate conidia with truncate bases, vaguely similar to the three core species of *Fusichalara*, have been subsequently added to the genus: *F. minuta* Hol.-Jech. (Gams & Holubova-Jechova, 1976), *F. clavatispora* P.M. Kirk (Kirk & Spooner, 1984), and *F. goanensis* Bhat & W.B. Kendrick (Bhat & Kendrick, 1993).

Recent phylogenetic studies showed that *Fusichalara* is a polyphyletic genus. Except for *F. dingleyae* (Réblová, 2004) and *F. minuta* (Réblová et al., 2017), molecular data of the other four *Fusichala-*

lara species are not available and their phylogenetic relationships are unknown. *Fusichalara dingleyae* was confirmed experimentally by ascospore isolation and molecular data to be the asexual morph of *Chaetosphaeria fusichalaroides* Réblová (*Chaetosphaeriales*, *Sordariomycetes*) (Réblová, 2004), whereas *F. minuta* was phylogenetically placed in the *Sclerococcales* (*Eurotiomycetes*) (Réblová et al., 2017). Since the sequence data of the generic type, *Fusichalara dimorphospora*, is currently unavailable, therefore, the genus is placed in *Ascomycota* genera *incertae sedis* (Wijayawardene et al., 2018).

During our continuing survey of microfungi occurring on plant litter submerged in freshwater streams of Taiwan (Goh & Kuo, 2018, 2020, 2021; Hsieh et al., 2021a, 2021b; Kuo & Goh 2018a, 2018b, 2019, 2021; Kuo et al., 2022), we collected an undescribed species of *Fusichalara*. We identify this fungus morphologically as a new species by comparing it with all other previously described *Fusichalara* species. In the present paper, we investigated its phylogenetic lineage, whether it belong to the *Chaetosphaeriales* (as *F. dingleyae*) or the *Sclerococcales* (as *F. minuta*). Since the genus is polyphyletic and phylogenetic positions of the type and three other species are yet to be resolved, we follow Réblová et al. (2017) to describe the present novel taxon as a species of *Fusichalara* based on morphology. A synopsis of the form-species and a composite diagram showing their conidial morphology are provided for comparison and ease of identification.

* Corresponding author, Department of Plant Medicine, National Chiayi University, 300 Syuefu Road, Chiayi City 60004, Taiwan.
E-mail address: tkgohmagic2005@yahoo.com.hk (T.K. Goh)



2. Materials and Methods

2.1. Sample collection and morphological studies

Plant litter including wood was collected in plastic bags and returned to the laboratory where the samples were incubated at room temperature on moist filter paper in sterile plastic boxes. Materials were examined periodically for the presence of sporulating fungal structures and species were identified primarily based on morphology. Squashed mounts of fungal structures were prepared using fresh materials from their natural substrata (wood) in lactophenol. Conidia were measured using Axiocam 506 Color with operating software ZEN 2 BLUE LITE linked to a Zeiss Axioskop 2 Plus microscope. The average conidial size was calculated based on 20 measurements. Single-spore isolations were made by using a hand-made glass needle. Isolated conidia were cleaned by dragging and rolling them on the surface of 3% water agar (Goh, 1999). The process was examined by using a stereo-microscope. Several small agar blocks, each containing a single cleaned conidium were eventually transferred to potato dextrose agar (PDA) slants or plates. The agar slants and plates containing the single conidia were incubated at 20 °C to obtain pure cultures. The holotype specimen was deposited in the Herbarium at the National Museum of Natural Science (NMNS), Taichung, Taiwan (herbarium code, TNM). An ex-type culture was deposited at the Bioresource Collection and Research Centre (BCRC), Food Industry Research and Development Institute, Hsinchu, Taiwan. A duplicate of dried specimen (isotype) was deposited at the Department of Plant Medicine, National Chiayi University (NCYU), Chiayi, Taiwan. All the fungal DNA sequences generated from the present study were deposited at GenBank.

2.2. DNA extraction, PCR amplification, and sequencing for fungal isolates

DNA extraction was carried out following Sambrook and Russell (2001) by using biomass from 60-d-old PDA cultures. DNA amplification was performed by polymerase chain reaction (PCR) using an experimental sample cocktail consisting of 2–8 ng DNA template, 0.4 ng forward primer and reverse primer, PCR Master Mix II (5×) (Biomax Scientific Co., Ltd., Taiwan). The large subunit ribosomal RNA gene (LSU) and the internal transcribed spacer regions (ITS) were amplified using the primers LR0R (5'-ACCCGCT-GAAGTAAAGC-3') and LR5 (Vilgalys & Hester, 1990), and ITS5/ITS4 (White et al., 1990), respectively. Sequencing was performed on a Cycle Sequencing Applied Biosystems 3730 DNA Analyzer, with BigDyeR Terminator v3.1 Cycle Sequencing Kit (Applied Biosystems, USA), by using the Sanger dideoxy sequencing method (Zimmermann et al., 1988) and the final sequence was assembled from two or more overlapping sequence reads.

2.3. Phylogenetic analysis

The LSU and ITS sequences generated in this study were supplemented with additional sequences retrieved from GenBank representing taxa from several related ordinal lineages of fungi (Table 1) from the *Eurotiomycetes* (i.e., namely *Chaetothyriales*, *Coryneliales*, *Mycocaliciales*, *Onygenales*, and *Sclerococcales*), and the *Sordariomycetes* (i.e., *Chaetosphaeriales* and *Glomerellales*). *Trichoglossum hirsutum* AFTOL-ID 64 (*Leotiomycetes*) was selected as the outgroup. Each dataset containing 41 sequences was analyzed. MUSCLE was used for DNA alignment (Edgar, 2004). Poorly aligned positions of DNA alignment were manually modified where necessary. After alignment, uneven ends were trimmed off.

The alignment block of concatenated sequences was 1903 bp long (including gaps), and the dataset was partitioned at the 1106–1107 bp positions separating the LSU-ITS segments. The alignment file is attached to this paper as supplemental information. The evolutionary history of the combined dataset was inferred using the Maximum Likelihood (ML) and Bayesian Inference (BI) methods in RAxML version 8.2.10 and MrBayes v3.2.6 under UBUNTU 19.10 (64-bit) operating system (Ronquist & Huelsenbeck, 2003; Stamatakis, 2014). The substitution model was GTR + Gamma. The best substitutional model was tested separately for each of the gene segments and the concatenated dataset. For RAxML, all random seeds for rapid bootstrapping and tree inferences were explicitly set to 5566 during the analysis to ensure reproducibility. Analyses were repeated based on 1000 bootstrapped data sets to obtain non-parametric bootstrap support (BS). MrBayes was run for 1,000,000 generations, with trees sampled every 100 generations. The first 25% of sampled trees were discarded (relburnin). Additional phylogenetic trees inferred from the concatenated sequence and the individual gene segments (ITS and LSU) using the Neighbour-Joining method were run in MEGA7 (Kumar et al., 2016) and the results are attached with this paper as supplemental information.

3. Results

3.1. Phylogeny

As MegaBlast search results show, the ITS barcode similarity of the new species to the most closely related *F. minuta* is 93.79%, indicating that they are distinct species. The phylogenetic tree inferred from the aligned sequences of 41 taxa from related fungal lineages is shown in Fig. 1. Numbers on nodes represent ML bootstrap values (greater than 70%) and Bayesian posterior probabilities (greater than 0.90) are shown in the tree. Within the alignment block, there were a total of 1065 distinct alignment patterns for analysis (i.e., LSU = 501, ITS = 564), with 991 variable sites including 764 parsimony-informative sites and 193 singleton sites. Molecular data revealed several small clusters of taxa belonging to various fungal groups representing various ordinal lineages within the *Eurotiomycetes* and *Sordariomycetes*. The new *Fusichalara* species collected from Taiwan (BCRC-FU31906) clustered together with *F. minuta* (CBS 709.88) with a ML bootstrap support of 74% but without support by Bayesian inference. In the phylogenetic tree inferred from the concatenated dataset generated by the neighbour-joining method (not shown), however, the two *Fusichalara* species clustered together with a higher bootstrap support of 91%. In both trees, the two species of *Fusichalara* were adjacent to a clade comprising *Cylindroconidius aquaticus* (MFLUCC 11-0294) and *Rhopalophora clavispota* (CBS 281.75), with a 100% bootstrap support. These four taxa were adjacent to a clade consisting of *Dactylospora* and *Sclerococcum* species, and all formed a cluster representing members of the *Sclerococcales* with a 100% bootstrap support. *Chaetosphaeria fusichalaroides* (LAMIC0149/13), the sexual morph of *Fusichalara dingleyae*, clustered with the other two *Chaetosphaeria* species with an ML bootstrap value of 90% (Bayesian posterior probabilities = 1.00), and they were among other representative taxa in the *Chaetosphaeriales* with a 100% bootstrap support.

3.2. Taxonomy

Fusichalara pallida C.H. Kuo, S.Y. Hsieh & Goh, sp. nov.

Figs. 2, 3

MycoBank No.: MB 851385.

Table 1. Taxa and sources of sequences used in the present phylogenetic analysis.

Fungal taxon	Fungal strain/isolate	ITS	LSU	Fungal lineage
<i>Aphanoascus verrucosus</i>	NBRC 32381	JN943440	JN941553	Onygenales
<i>Ascosphaera apis</i>	CBS 402.96	MH862580	FJ358275	Onygenales
<i>Brunneodindenasporium brasiliense</i>	CBS 112007	JQ889272	JQ889288	Chaetosphaeriales
<i>Caliciopsis orientalis</i>	CBS 138.64	KP881690	DQ470987	Coryneliales
<i>Camptophora hylomeconis</i>	CBS 113311	KC455241	EU035415	Chaetothyriales
<i>Ceramothyrium carniolicum</i>	CBS 175.95	KC978733	KC455251	Chaetothyriales
<i>Chaenothecopsis savonica</i>	Tibell 15876	AY795868	AY796000	Mycocaliciales
<i>Chaetosphaeria decastyla</i>	NN055410	OL627834	OL655134	Chaetosphaeriales
<i>Chaetosphaeria fusichalaroides</i>	LAMIC0149/13	KX499466	KR363058	Chaetosphaeriales
<i>Chaetosphaeria obovoidea</i>	GZCC 22-0085	ON502901	ON502894	Chaetosphaeriales
<i>Chloridium chloroconium</i>	CBS 149055	OP455398	OP455505	Chaetosphaeriales
<i>Chloridium peruense</i>	CBS 126074	OP455424	OP455531	Chaetosphaeriales
<i>Cladophialophora humicola</i>	CBS 117536	EU035408	KC809987	Chaetothyriales
<i>Cladophialophora minutissima</i>	CBS 121758	MH863155	KJ636047	Chaetothyriales
<i>Codinaeella lutea</i>	CBS 624.77	OL654086	OL654143	Chaetosphaeriales
<i>Corynelia africana</i>	ARW 247	KP881693	KP881714	Coryneliales
<i>Corynelia fruitigena</i>	ARW 250	KP881704	KP881716	Coryneliales
<i>Cylindroconidium aquaticum</i>	MFLUCC 11-0294	MH236576	MH236579	Sclerococcales
<i>Cylindrotrichum clavatum</i>	CBS 125239	GU291799	GU180649	Glomerellales
<i>Cyphellophora laciniata</i>	CBS 190.61	KF928483	FJ358239	Chaetothyriales
<i>Cyphellophora sessilis</i>	CBS 243.85	MH861875	EU514700	Chaetothyriales
<i>Dactylospora stygia</i>	HN2022090370	OQ534124	OQ534408	Sclerococcales
<i>Dactylospora vrijmoediae</i>	NTOU 4002	KJ958534	KC692153	Sclerococcales
<i>Fusichalara minuta</i>	CBS 709.88	KX537754	KX537758	Sclerococcales
<i>Fusichalara pallida</i>	BCRC FU31906	OR944928	OR944931	Sclerococcales
<i>Kylindria chinensis</i>	MFLUCC 16-0965	MH120190	MH120186	Glomerellales
<i>Kylindria peruamazonensis</i>	CBS 838.91	GU180628	GU180638	Glomerellales
<i>Lagenulopsis bispora</i>	ARW 249	KP881709	KP881717	Coryneliales
<i>Menispora tortuosa</i>	AFTOL-ID 278	KT225527	AY544682	Chaetosphaeriales
<i>Menisporopsis theobromae</i>	MFLUCC 15-0055	KX609957	KX609954	Chaetosphaeriales
<i>Mycocalicium polyporaenum</i>	ZWGeo60Clark	AY789363	AY789362	Mycocaliciales
<i>Onygena corvina</i>	CBS 225.60	MH857958	MH869510	Onygenales
<i>Reticulascus tulasneorum</i>	CBS 570.76	MH861002	MH872775	Glomerellales
<i>Rhopalophora clavispora</i>	CBS 281.75	KX537752	KX537756	Sclerococcales
<i>Sclerococcum martyinii</i>	F-1570b	MZ221616	MZ221623	Sclerococcales
<i>Sclerococcum simplex</i>	MFLU 21-0117	MZ664325	MZ655912	Sclerococcales
<i>Spiromastix warcupii</i>	CBS 576.63	MH858362	DQ782909	Onygenales
<i>Sporoschisma hemisporium</i>	DLUCC 0700	KX455869	KX455862	Chaetosphaeriales
<i>Sporoschismopsis angustata</i>	CBS 136360	KF730739	KF730740	Glomerellales
<i>Stenocybe pullatula</i>	Tibell 17117	AY795878	AY796008	Mycocaliciales
<i>Trichoglossum hirsutum</i>	AFTOL-ID 64	DQ491494	AY544653	Leotiomyces

Diagnosis: This species differs from other *Fusichalara* species in conidial morphology (first-formed conidia 8-septate, subsequent conidia 7-septate, with pale coloration)

Type: TAIWAN, Taoyuan County, Fuxing District (24.647–121.444, 1099.11 m a.s.l.), on decaying wood submerged in a freshwater stream, leg. Chang-Hsin Kuo, 14 May 2022, 111FX4-2A3 (**holotype**, TNM F0037300; **isotype**, NCYU-111FX4-2A3); ex-holotype living culture BCRC:FU31906).

DNA sequences from ex-holotype strain: OR944928 (ITS), OR944931 (LSU).

Etymology: *pallida*, referring to the paler coloration of the conidia when compared with that of the type species (*F. dimorphospora*).

Colonies on natural substratum scattered, black, hairy. Mycelium immersed, composed of subhyaline to pale brown, smooth hyphae. Conidiophores scattered, solitary or in loose fascicle of 2–3, erect, unbranched, straight or slightly curved, cylindrical, brown to dark brown, smooth-walled, up to 5–7-septate, 280–320 × 9.5–10.5 μm, not distinctly inflated to demarcate the venter of the phialide, bearing a distinctly darker tubular collarette at the distal end. Conidiogenous cells phialidic, integrated, terminal, determinate, subcylindrical, 110–120 × 9.5–11 μm, composed of a poorly differentiated, very slightly inflated venter and a tubular collarette 95–117 ×

9.5–11 μm, the point of transition from venter to collarette slightly constricted (6.5–7.5 μm wide), with pronounced wall-thickening of the inner wall; distal end of tubular collarette bearing a conspicuous rim of ragged marginal frills at the opening, producing endogenous conidia successively which accumulate in a slimy mass after emergence from the tubular collarette. Phialoconidia of two kinds: first-formed conidia long-cylindrical, rounded at the apex, obconic-truncate at the base, straight, very pale orange-brown, 8-septate, 87–92 × 5.5–6 μm; subsequent conidia elongate-fusiform to slightly sigmoid, narrowly rounded at the apex (ca 2 μm wide), obconic-truncate at the base (1.0–1.5 μm wide), predominantly 7-septate, versicolorous, median cells very pale orange-brown, end cells subhyaline, (47.5–)52–67 × 5–6(–6.5) μm (\bar{x} = 58.5 × 5.5 μm, n = 20). Sexual morph undetermined.

Single conidia in PDA slants germinated readily (100%), but growth was extremely slow, with a growth rate of about 0.5 mm/d. Colonies on PDA slants (Fig. 3A) were pale creamy brown, with a broad fuzzy margin composed of thin aerial mycelium. On PDA plates (Fig. 3B), colonies attained 4.7 cm diam in 100 d at 20 °C, more or less effuse, funiculate, with sparse mycelium on the surface, pale creamy brown, with a slightly wavy margin, evenly colored, reverse side similar in appearance to upper side (Fig. 3C). Vegetative hyphae (Fig. 3H) smooth, branched, septate, hyaline,

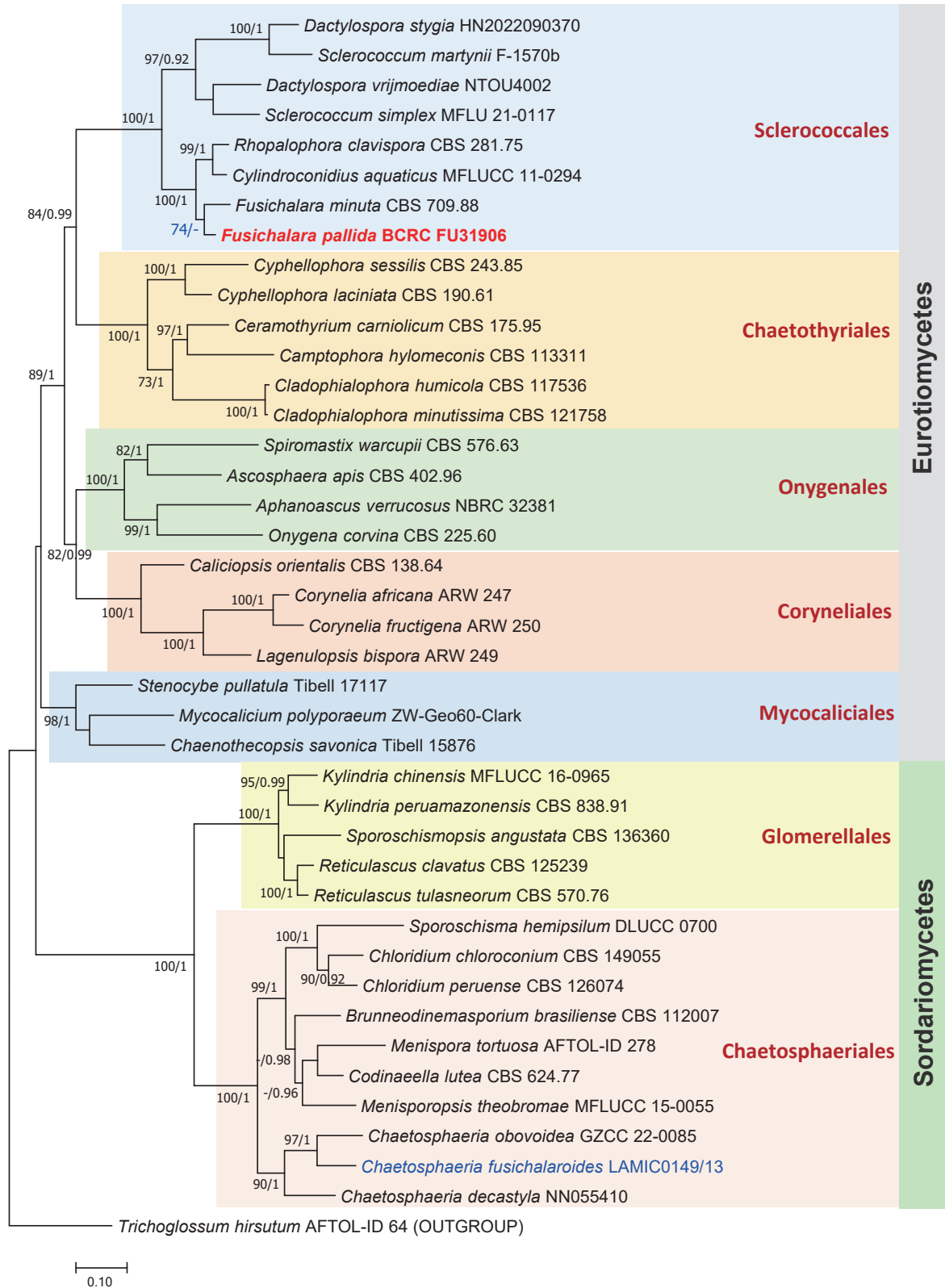


Fig. 1 – RAxML phylogenetic tree with BI inferred from concatenated ITS and LSU partial sequences of the rDNA showing relationships of *Fusichalara* species with other taxa in the Sclerococcales and other fungal lineages. The tree is rooted with *Trichoglossum hirsutum* AFTOL-ID 64 (*Leotiomycetes*). Numbers on nodes represent ML bootstrap values (greater than 70%) and Bayesian posterior probabilities (greater than 0.90). The new *Fusichalara* species (in bold red font) from Taiwan is among members of Sclerococcales. *Chaetosphaeria fusichalaroides* (in blue font), the teleomorph of *Fusichalara dingleyae*, is within the clade comprising several representative chaetosphaeriaceous taxa.

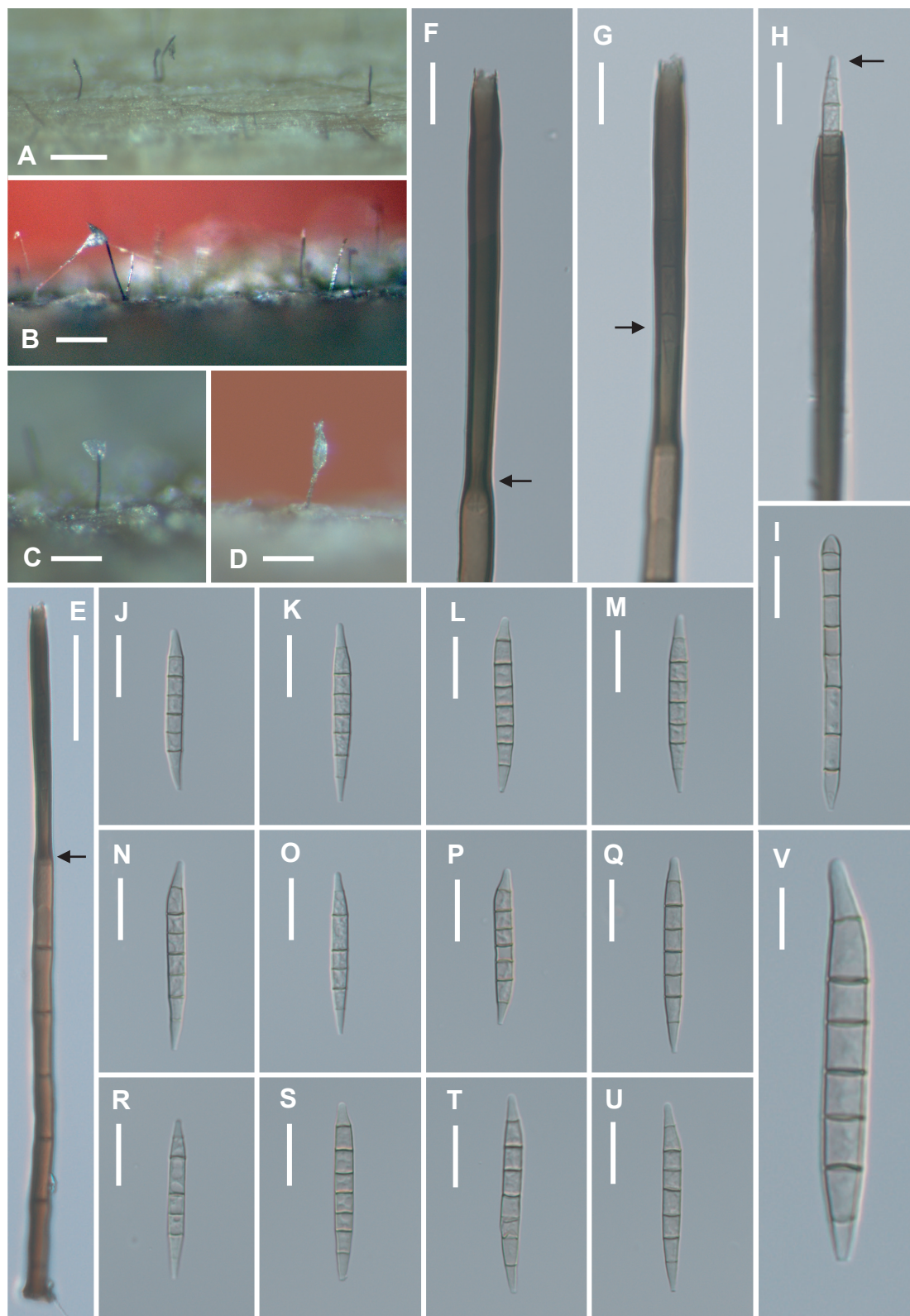


Fig. 2 – *Fusichalara pallida* (TNM F0037300, holotype). A, B: Colonies on natural substratum. C, D: Conidiophores on natural substratum, bearing a tuft of conidial mass at the apex. E: A conidiophore. Arrow indicates the point of transition from venter to the darker tubular collarette. F: Closer view of phialide. Note the pronounced wall-thickening (arrow) inside the phialide at the base of the tubular collarette. G: Closer view of phialide. Arrow indicates a conidium in the tubular collarette. H: Closer view of phialide showing a conidium emerging at the opening of the tubular collarette. Arrow points to the conically rounded conidial apex. I: A first-formed conidium which is cylindrical, with a rounded apex and an obconically truncate base. J–U: Subsequent conidia which are typically fusiform, 7-septate, and have paler end cells. Note that each conidium is conically rounded at the apex and obconically truncate at the base. V: Higher magnification of a subsequently formed conidium. Bars: A 500 μm ; B–D 200 μm ; E 50 μm ; F–U 20 μm ; V 10 μm .

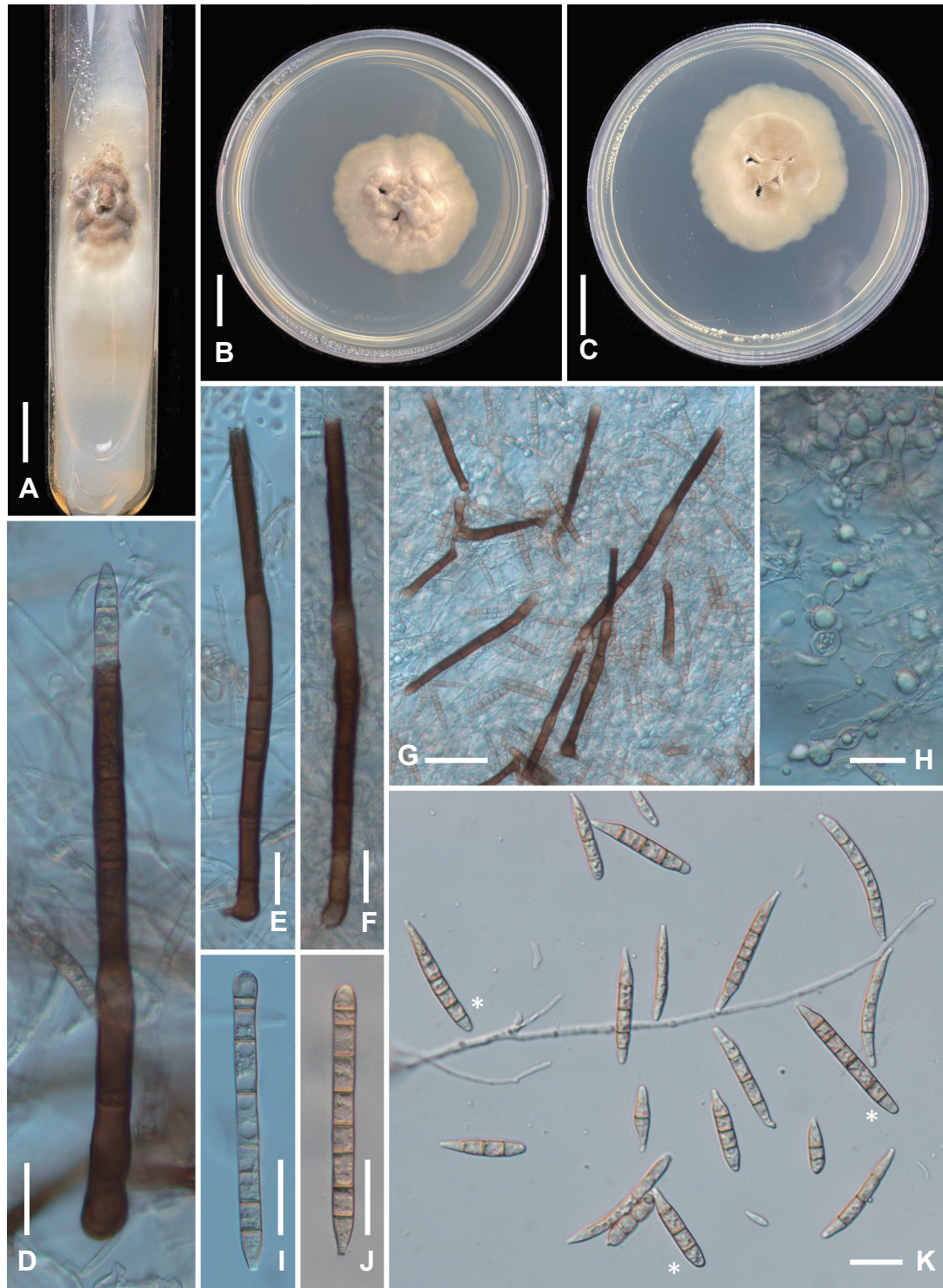


Fig. 3 – Ex-type cultures of *Fusichalara pallida* (BCRC-FU31906). A: Colony on a PDA slant developed from single-spore isolate. B: Colony on PDA plate (surface view). C: Colony on PDA plate (reverse side). D: A conidiophore showing a subsequent conidium emerging at the tip of the phialide. E, F: Conidiophores. G: Squashed mount showing conidiophores, conidia, and chlamydozooids. H: Mycelium showing chlamydozooid formation (monilioid hyphae). I, J: First-formed conidia. K: Conidia, first-formed conidia (indicated by asterisks) are cylindrical whereas subsequent conidia are fusiform. Bars: A–C 1 cm; D–F, H–K 20 μ m; G 50 μ m.

1.5–2.5 μm wide, frequently appearing moniloid and becoming intercalarily swollen, forming chlamydo-spores of 3–6 μm diam (Fig. 3H). Conidiophores in culture arising from submerged mycelium, often in a terminal position (Fig. 3D–G), morphologically similar to those produced on natural substratum. First-formed conidia in culture (Fig. 3I, 3J) cylindrical, pale brown, (5–)6–8(–9)-septate, 87–92 \times 5.5–6 μm ; subsequent conidia (Fig. 3K) fusiform, narrowly rounded at the apex (ca 2 μm wide), obconic-truncate at the base (1.5–1.5 μm wide), (1–)2–4(–5)-septate, versicolorous, median cells pale orange-brown, end cells subhyaline, (47.5–)52–67 \times 5–6(–6.5) μm (\bar{x} = 58.5 \times 5.5 μm , n = 20).

Habitat and distribution: Saprobic on decaying wood submerged in freshwater streams; known only from the type locality

(Taiwan).

4. Discussion

Morphologically, *Fusichalara pallida* is comparable with *F. dimorphospora* and *F. novae-zealandiae* in having long-cylindrical first-formed conidia and fusiform subsequent conidia with paler end cells (Fig. 4), however, they differ in conidial dimensions (Table 2). *Fusichalara dimorphospora* differs in having first-formed conidia which are wider (7–9 μm vs. 5.5–6 μm), occasionally with one or a few oblique septa, and subsequent conidia which are wider (7–10 μm vs. 5–6.5 μm). *Fusichalara pallida* is most similar to *F. novae-zealandiae* but the latter differs in having first-formed conid-

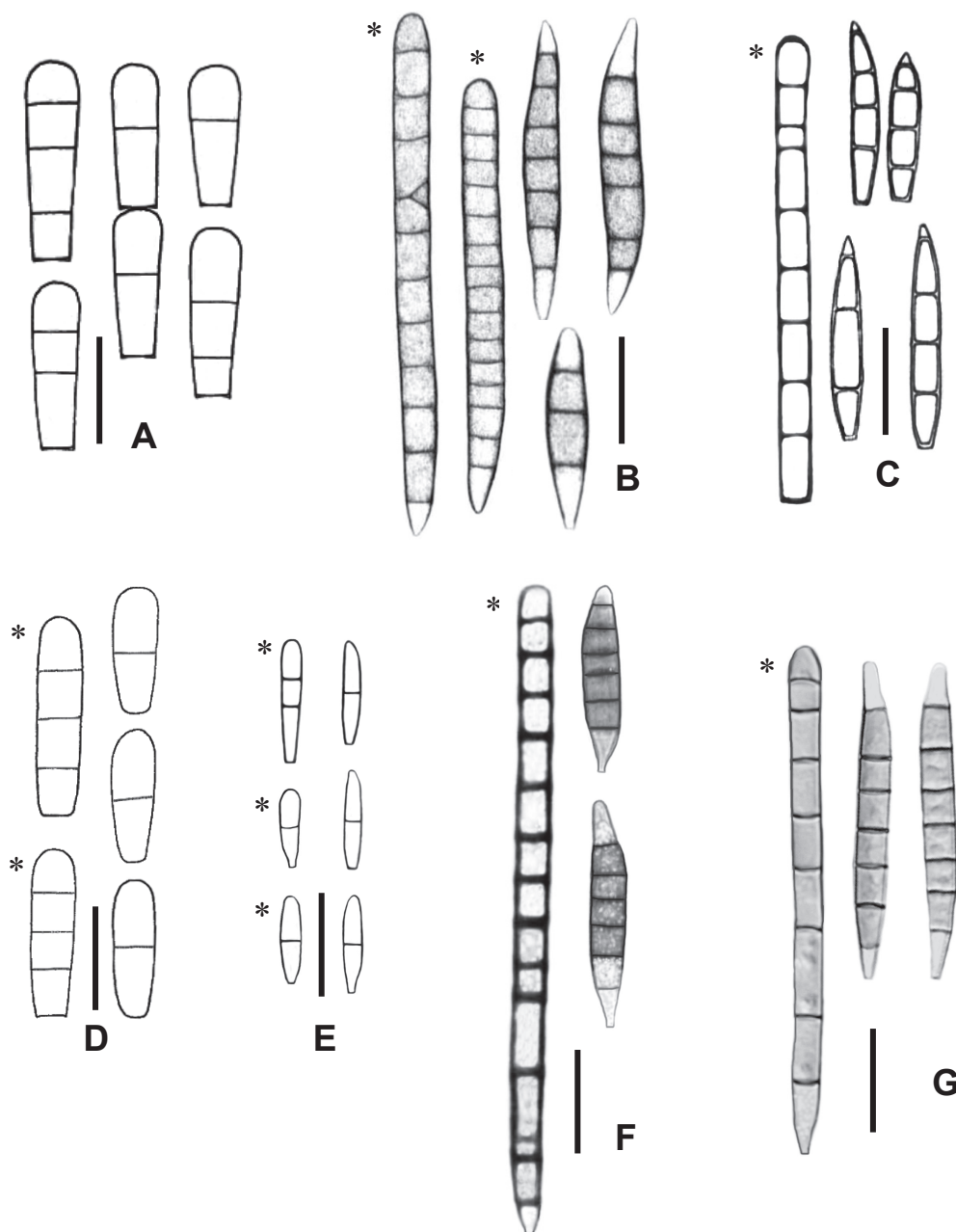


Fig. 4 – Conidia of *Fusichalara* species, re-illustrated with reference to the literature (Hughes & Nag Raj, 1973; Kirk & Spooner, 1984; Bhat & Kendrick, 1993; Réblová et al., 2017). A: *F. clavatispora*. B: *F. dimorphospora*. C: *F. dingleyae*. D: *F. goanensis*. E: *F. minuta*. F: *F. novae-zealandiae*. G: *F. pallida*. First-formed conidia are indicated by an asterisk (*). Bars: A, D 10 μm ; B, C, E–G 20 μm .

Table 2. Comparison of *Fusichalara* species.

Species	Current ordinal position				First formed conidia				Subsequent conidia				Holotype and type locality		Natural substratum	References
	Current ordinal position	Size of the tubular collarettes (μm)	Shape	Septation	Coloration	Size (μm)	Shape	Septation	Coloration	Size (μm)	Locality	Substrate				
<i>F. clavatispora</i>	ascomycetous incertae sedis	12–20 × 4–5	elongate cuneiform to clavate (apex broadly rounded, base truncate)	1–3	hyaline	12–16 × 3.5–4	elongate cuneiform to clavate (apex broadly rounded, base truncate)	1–3	hyaline	12–16 × 3.5–4	IMI 25873a; The Isle of Aran (Scotland)	dead stem of <i>Rubus fruticosus</i>	Kirk & Spooner, 1984			
<i>F. dimorphospora</i>	ascomycetous incertae sedis	70–130 × 13.5–15.3	Long-cylindrical (apex bluntly rounded, base obconic)	11–17; sometimes with one or a few oblique septa	pale brown to brown, base subhyaline	85–126 × (6–)7–9	fusiform or slightly sigmoid (base more tapered than the apex)	(3–5)7(–8)	median cells brown, end cells subhyaline	7-septate conidia (50–80–72 × 9–10; 3-septate conidia 36–38 × 7.2–8.3)	PDD 30402; Westland District (New Zealand)	dead bark of <i>Weinmannia racemosa</i>	Hughes & Nag Raj, 1973			
<i>F. dingleyae</i>	Chaetosphaeriales	65–110 × 8–9.5	Long-cylindrical (apex bluntly round, base truncate)	7–16	hyaline	62.5–83.5(–95) × 5.5–6.5	fusiform (apex narrowly conical, base truncate)	3–5	hyaline to subhyaline	36–58 × 5–5.6	PDD 21599; Auckland Province (New Zealand)	rotten wood	Hughes & Nag Raj, 1973			
<i>F. goanensis</i>	ascomycetous incertae sedis	18–24 × 6.5–8	nearly cylindrical (apex rounded, base truncate)	3	hyaline	17–22 × 5–6	nearly cylindrical (apex rounded, base truncate)	1	hyaline	12–15 × 4.5–5.5	DAOM 214604; Goa State (India)	decaying twig	Bhat & Kendrick, 1993			
<i>F. minuta</i>	Sclerococcales	11–14 × 2.5–3	clavate (apex rounded, base truncate)	1–2	hyaline	(9.5–)10.5–15 × 2–2.5	fusiform or fusiform-clavate (apex narrowly rounded, base obconic-truncate)	1	hyaline	(10–)11–12.5 × 2–2.5	PRM 795927; Central Bohemia Region (Czech Republic)	decaying trunk of <i>Quercus parva</i>	Gams & Holubová-Jechová, 1976; Řeblová et al., 2017			
<i>F. novae-zelandiae</i>	ascomycetous incertae sedis	(60–)80–100 (–120) × 8–11	Long-cylindrical (apex rounded, base narrowly truncate)	9–12(–18)	subhyaline to pale brown	83–105 × 5.5–6.5	fusiform to slightly sigmoid (apex narrowly rounded, base obconic-truncate)	7	median cells pale brown, end cells subhyaline	(27–)30–40(–47) × 5–6.5(–7)	PDD 30404; Auckland Province (New Zealand)	rotten wood of <i>Leptospermum scoparium</i>	Hughes & Nag Raj, 1973			
<i>F. pallida</i>	Sclerococcales	95–117 × 9.5–11	Long-cylindrical (apex rounded, base narrowly truncate)	8	very pale orange-brown	87–92 × 5.5–6	elongate-fusiform to slightly sigmoid (apex narrowly rounded, base obconic-truncate)	7	median cells very pale orange-brown, end cells subhyaline	(47.5–)52–67 × 5–6(–6.5)	TNM F0037300; Fuxing District (Taiwan)	Submerged wood	This paper			

ia with more septa (9–18 vs. 8) and subsequent conidia which are distinctly shorter (27–47 μm vs. 47.5–67 μm). Phylogenetically, *F. pallida* clustered with *F. minuta* within the *Sclerococcales* (Fig.1) and was closely related to the genera *Cylindroconidius* X.D. Yu & H. Zhang (as '*Cylindroconidiis*', Yu et al. 2018) and *Rhopalophora*. However, *Cylindroconidius* differs in having polyblastic conidiogenous cells that produce *Diplococcium*-like conidia (Yu et al., 2018). On the other hand, both *Fusichalara* and *Rhopalophora* have phialidic conidiogenous cells, but the conspicuous wall thickening at the base of the tubular collarette and the longer, septate, dimorphic conidia in *Fusichalara* can readily distinguish it from *Rhopalophora* (Řeblová et al., 2017).

Fusichalara species have distinct dematiaceous conidiophores bearing an integrated phialide with a tubular collarette. This feature is reminiscent of the erect, lageniform conidiophores with a tubular collarette in species of *Sporoschisma* Berk. & Broome and *Sporoschismopsis* Hol.-Jech. & Hennebert (Goh et al., 1997). With sufficient molecular studies in the past decade, *Sporoschisma* species have been proven to be members of the *Chaetosphaeriales* (Yang et al., 2016) whereas the genus *Sporoschismopsis* was positioned in the *Glomerellales* (Řeblová, 2014). The present phylogenetic analysis concurred with these findings, and confirmed that *Fusichalara pallida* and *F. minuta*, although both are comparable in having similar conidiogenous features, belong to the *Sclerococcales* (*Eurotiomycetes*), which is phylogenetically distant from species of *Sporoschisma* and *Sporoschismopsis* that are members of the *Sordariomycetes*.

Fusichalara dingleyae is one of the core species described by Hughes and Nag Raj (1973) for the establishment of the genus. It has dark, robust, dark conidiophores with a terminal, long-cylindrical phialide, producing septate conidia occasionally in readily seceding chains. These conidiogenous features are typically found in the chaetosphaeriacean genus *Sporoschisma*. Moreover, *Fusichalara dingleyae* has been experimentally confirmed to be the asexual morph of *Chaetosphaeria fusichalaroides* (Řeblová, 2004) and phylogenetic studies have also proven its position in the *Chaetosphaeriales*. These results clearly showed that the genus *Fusichalara* is polyphyletic, species of which could be members of *Chaetosphaeriales*, *Sclerococcales*, or further orders.

Fusichalara dingleyae is recognized by its reddish-brown, coarsely roughened conidiophores growing in fascicles on a thin stroma and hyaline, predominantly 3–7-septate fusiform conidia (Hughes & Nag Raj, 1973). The other six known species of *Fusichalara* have brown to black conidiophores that are solitary rather than fasciculate on the natural substrata. The present collection of *F. pallida* from Taiwan has morphological features comparable with *F. dimorphospora* and *F. novae-zelandiae* in having predominantly 7-septate, versicolorous conidia with median cells that are uniformly pigmented and end cells that are hyaline to subhyaline. In contrast, *Fusichalara clavatispora*, *F. goanensis* and *F. minuta* have hyaline conidia that differ from those of *F. dingleyae*, *F. dimorphospora*, *F. novae-zelandiae*, and *F. pallida* in shape, size, septation, and coloration. *Fusichalara clavatispora* is also somewhat atypical of the genus because its phialides possess a swollen venter and there is no distinction between the first-formed and subsequent conidia (Kirk & Spooner, 1984). These differences indicate that the genus *Fusichalara* is somewhat morphologically heterogeneous. Due to the phylogenetic uncertainty of the genus, we refrain from constructing a taxonomic key to the species for the time being, but merely provide a synopsis and a composite illustration of the form-species to ease their identification.

Disclosure

The authors declare no conflicts of interest. All the experiments undertaken in this study comply with the current laws of Taiwan where they were performed.

Acknowledgments

This research was financially supported, in part, by the National Science and Technology Council of Taiwan (Grant Number 111-2621-B-415-001) and the Ministry of Agriculture of Taiwan (Grant Number 113AS-1.7.1-AS-03). We would like to thank Dr Jie-Hao Ou for his constructive opinion regarding the phylogenetic analysis of fungi. Thanks are extended to Ms. Shing-Yu Lin and Ms. Hsin-Yi Peng for general technical support.

References

- Bhat, D. J., & Kendrick, B. (1993). Twenty-five new conidial fungi from the Western Ghats and the Andaman Islands (India). *Mycotaxon*, *49*, 19–90.
- Edgar, R. C. (2004). MUSCLE: multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Research*, *32*, 1792–1797. <https://doi.org/10.1093/nar/gkh340>
- Gams, W., & Holubová-Jechová, V. (1976). *Chloridium* and some other dematiaceous hyphomycetes growing on decaying wood. *Studies in Mycology*, *13*, 1–99.
- Goh, T. K. (1999). Single-spore isolation using a hand-made glass needle. *Fungal Diversity*, *2*, 47–63.
- Goh, T. K., Ho, W. H., Hyde, K. D., & Umali, T. E. (1997). New records and species of *Sporoschisma* and *Sporoschismopsis* from submerged wood in the tropics. *Mycological Research*, *101*, 1295–1307. <https://doi.org/10.1017/S0953756297003973>
- Goh, T. K., & Kuo, C. H. (2018). A new species of *Helicoön* from Taiwan. *Phytotaxa*, *346*, 141–156. <https://doi.org/10.11646/phytotaxa.346.2.2>
- Goh, T. K., & Kuo, C. H. (2020). *Jennwenomyces*, a new hyphomycete genus segregated from *Belemnospora*, producing versicolored phragmospores from percurrently extending conidiophores. *Mycological Progress*, *19*, 869–883.
- Goh, T. K., & Kuo, C. H. (2021). Reflections on *Canalisporium*, with descriptions of new species and records from Taiwan. *Mycological Progress*, *20*, 647–680. <https://doi.org/10.1007/s11557-021-01689-6>
- Hsieh, S. Y., Goh, T. K., & Kuo, C. H. (2021a). A taxonomic revision of *Stanjehughesia* (*Chaetosphaeriaceae*, *Sordariomycetes*), with a novel species *S. kaohsiungensis* from Taiwan. *Phytotaxa*, *484*, 261–280. <https://doi.org/10.11646/phytotaxa.484.3.2>
- Hsieh, S. Y., Goh, T. K., & Kuo, C. H. (2021b). New species and records of *Helicosporium* sensu lato from Taiwan, with a reflection on current generic circumscription. *Mycological Progress*, *20*, 169–190. <https://doi.org/10.1007/s11557-020-01663-8>
- Hughes, S. J., & Nag Raj, T. R. (1973). New Zealand fungi. 20. *Fusichalara* gen. nov. *New Zealand Journal of Botany*, *11*, 661–671. <https://doi.org/10.1080/0028825X.1973.10430307>
- Index Fungorum (2024). Retrieved March 31, 2024, from <http://www.indexfungorum.org/names/Names.asp>
- Kirk, P. M., & Spooner, B. M. (1984). An account of the fungi of Arran, Gigha and Kintyre. *Kew Bulletin*, *38*, 503–597.
- Kumar, S., Stecher, G., & Tamura, K. (2016). MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Molecular Biology and Evolution*, *33*, 1870–1874. <https://doi.org/10.1093/molbev/msw054>
- Kuo, C. H., & Goh, T. K. (2018a). Two new species of helicosporous hyphomycetes from Taiwan. *Mycological Progress*, *17*, 557–569. <https://doi.org/10.1007/s11557-018-1384-7>
- Kuo, C. H., & Goh, T. K. (2018b). A new species and a new record of *Helicomycetes* from Taiwan. *Mycoscience*, *59*, 433–440. <https://doi.org/10.1016/j.myc.2018.04.002>
- Kuo, C. H., & Goh, T. K. (2019). *Ellisembia acicularia* sp. nov. and four new records of freshwater hyphomycetes from streams in the Alishan area, Chiayi County, Taiwan. *Nova Hedwigia*, *108*, 185–196. https://doi.org/10.1127/nova_hedwigia/2018/0493
- Kuo, C. H., & Goh, T. K. (2021). Two new species and one new record of *Xenosporium* with ellipsoidal or ovoid conidia from Taiwan. *Mycologia*, *113*, 434–449. <https://doi.org/10.1080/00275514.2020.1837566>
- Kuo, C. H., Hsieh, S. Y., & Goh, T. K. (2022). *Wenhsuisporus taiwanensis* gen. et sp. nov., a peculiar setose hyphomycete from submerged wood in Taiwan. *Mycological Progress*, *21*, 409–426. <https://doi.org/10.1007/s11557-021-01748-y>
- Réblová, M. (2004). Four new species of *Chaetosphaeria* from New Zealand and redescription of *Dictyochaeta fuegiana*. *Studies in Mycology*, *50*, 171–186.
- Réblová, M. (2014). *Sporoschismopsis angustata* sp. nov., a new holomorph species in the *Reticulascaceae* (*Glomerellales*), and a reappraisal of *Sporoschismopsis*. *Mycological Progress*, *13*, 671–681.
- Réblová, M., Untereiner, W. A., Štěpánek, V., & Gams, W. (2017). Disentangling *Phialophora* section *Catenulatae*: disposition of taxa with pigmented conidiophores and recognition of a new subclass, *Sclerococcomycetidae* (*Eurotiomycetes*). *Mycological Progress*, *16*, 27–46. <https://doi.org/10.1007/s11557-016-1248-y>
- Ronquist, F., & Huelsenbeck, J. P. (2003). MrBayes 3: Bayesian phylogenetic inference under mixed models. *Bioinformatics*, *19*, 1572–1574.
- Sambrook, J., & Russell, D. W. (2001). *Molecular cloning: a laboratory manual*. Cold Spring Harbor Laboratory Press.
- Stamatakis, A. (2014). RAXML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics*, *30*, 1312–1313.
- Vilgalys, R., & Hester, M. (1990). Rapid genetic identification and mapping of enzymatically amplified ribosomal DNA from several *Cryptococcus* species. *Journal of Bacteriology*, *172*, 4238–4246
- White, T. J., Bruns, T., Lee, S., & Taylor, J. (1990). Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: M. A. Innis, D. H. Gelfand, J. J. Sninsky, & T. J. White (Eds.), *PCR protocols: A guide to methods and applications* (pp. 315–322). Academic Press.
- Wijayawardene, N. N., Hyde, K. D., Lumbsch, H. T., Liu, J. K., Maharachchikumbura, S. S. N., Ekanayaka, A. H., Tian, Q., & Phookamsak, R. (2018). Outline of Ascomycota: 2017. *Fungal Diversity*, *88*, 167–263. <https://doi.org/10.1007/s13225-018-0394-8>
- Yang, J., Liu, J. K., Hyde, K. D., Bhat, D. J., JONES, E. G., & Liu, Z. Y. (2016). New species of *Sporoschisma* (*Chaetosphaeriaceae*) from aquatic habitats in Thailand. *Phytotaxa*, *289*, 147–157. <https://doi.org/10.11646/phytotaxa.289.2.4>
- Yu, X. D., Dong, W., Bhat, D. J., Boonmee, S., Zhang, D., & Zhang, H. (2018). *Cylindroconidiis aquaticus* gen. et sp. nov., a new lineage of aquatic hyphomycetes in *Sclerococaceae* (*Eurotiomycetes*). *Phytotaxa*, *372*, 79–87. <https://doi.org/10.11646/phytotaxa.372.1.6>
- Zimmermann, J., Voss, H., Schwager, C., Stegemann, J., & Ansoerge, W. (1988). Automated Sanger dideoxy sequencing reaction protocol. *FEBS Letters*, *233*, 432–436. [https://doi.org/10.1016/0014-5793\(88\)80477-0](https://doi.org/10.1016/0014-5793(88)80477-0)