Fungal Diversity (2011) 51:63–102
DOI 10.1007/s13225-011-0147-4

Revision of lignicolous *Tubeufiaceae* based on morphological reexamination and phylogenetic analysis

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Received: 18 October 2011 / Accepted: 23 October 2011 / Published online: 17 November 2011

Abstract In this paper we revisit the family *Tubeufiaceae* with notes on genera that we have re-examined where possible. Generic type specimens of *Acanthophiobolus, Kamalomyces, Podonectria, Thaxteriella* and *Thaxteriellopsis* were re-examined, described and illustrated and shown to belong to *Tubeufiaceae*. Notes are provided on *Acanthostigma, Chaetosphaerulina, Thaxterina* and *Tubeufia*, which are retained in *Tubeufiaceae*; however, we were unable to locate the types of these genera during the time frame of this study. *Allonecte* is excluded from the *Tubeufiaceae*, as the ascospores are fusiform-ellipsoidal, grey-brown and 1-septate and the asci are cylindrical, all of which are features more typical of *Pleosporaceae*, where it is transferred. *Byssocallis* has yellow to orange ascomata and clavate ascospores which is atypical of *Tubeufiaceae*. Thus its taxonomic status needs to be reevaluated. *Lentendraeopsis* has an endophytic habit, cylindro-clavate asci and two-celled ascospores more typical of *Pleosporales*, where it is transferred. *Taphrophila* has small ascomata, a thin peridium, branching setae around the apex of the ascomata, clavate to saccate asci and lacks pseudoparaphyses. These are features atypical of the *Tubeufiaceae*, and *Taphrophila* should be placed in the Dothideomycetes *incertae cedis.* Twelve new collections of *Tubeufiaceae* from Thailand were isolated, and their DNA was extracted. The sequence data of LSU, SSU and ITS rDNA were amplified and analyzed using parsimony and likelihood methods. The results of phylogenetic analysis was used to establish the inter-generic relationships in *Tubeufiaceae. Thaxteriellopsis lignicola,* epitypified in this investigation, is a sister taxon in the family *Tubeufiaceae* based on phylogenetic analysis of rRNA sequence data. *Chlamydotubeufia* is introduced as a new genus based on the production of dictyochlamydosporous anamorphs, including two new species. Three new species, one each in *Acanthostigma*, *Tubeufia* and *Thaxteriella* are also described and illustrated. The phylogenetic placement of these genera is also discussed.

Keywords Anamorph · *Aquaphila* · Dothideomycetes · Helicosporous · Molecular phylogeny · Woody litter fungi

Electronic supplementary material The online version of this article (doi:10.1007/s13225-011-0147-4) contains supplementary material, which is available to authorized users.

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Introduction

The family *Tubeufiaceae sensu* Barr (1979) included the type genus *Tubeufia* and four representative genera (*Letendraea, Melioliphila, Podonectria, Thaxteriella*) and was considered to be pleosporaceous. The representatives were saprobic, hyperparasitic or hypersaprobic on ascomycetes and scale insects, and characterized by brightly pigmented, fleshy, superficial ascomata, bitunicate asci, and mostly hyaline to pale brownish, narrowly-elongate, obovoid or oblong septate ascospores. Barr (1980) included ten genera while Rossman (1987) studied the family in detail and included 12 genera (Table 1). Several other genera have since been added and the family now encompasses 23 genera (Lumbsch and Huhndorf 2010) or 22 genera (Kirk et al. 2001). Most taxa of *Tubeufiaceae* are commonly found on woody litter, although some species can also be found on leaf litter or even decaying cloth and some are associated with other fungi or scale insects (Barr 1980; Rossman 1987; Kodsueb et al. 2006; Promputtha and Miller 2010; Sánchez and Bianchinotti 2010). The anamorphs of *Tubeufiaceae* have been well-studied and are mostly related to helicosporous taxa such as *Helicoma, Helicomyces* and *Helicosporium* (Tsui et al. 2006, 2007; Hyde et al. 2011).

In this study, we have examined the generic types or authentic specimens of several tubuefiaceous genera that we have been able to locate. We have also surveyed woody litter in forests throughout northern Thailand and obtained several new collections of these fungi, which have been described, isolated and sequenced. We compare the sequence data of these new collections to those from the generic types and to provide insights on the inter-generic and species relationships in *Tubeufiaceae*. One new genus and five new species encountered during the course of our field work are also described.

Material and methods

Sample collection and specimen examination

Decaying wood was randomly collected from forests at various sampling sites in northern Thailand (i.e. Chiang Mai, Chiang Rai, Lam Pang and Pha Yao). Material was examined under a Motic SMZ 168 Series microscope. Micromorphological structures were photographed using a Nikon ECLIPSE 80i compound microscope with a Canon 450D digital camera, and measurements were made using

Table 1 Various treatments of *Tubeufiaceae*

Barr, 1979	Barr 1980	Rossman 1987	Kirk et al. 2001	Lumbsch and Huhndorf 2010	This paper
Letendraea	*Allonectria*	*Allonectria*	*Acanthophiobolus*	*Acanthostigma*	*Acanthostigma*
Melioliphila	*Boerlagiomyces*	*Boerlagiomyces*	*Acanthostigmella*	*Acanthophiobolus*	*Acanthophiobolus*
Podonectria	*Byssocallis*	*Byssocallis*	*Allonecte*	*Acanthostigmella*	*?Acanthostigmella*
Rebentischia	*Letendraea*	*Letendraea*	*Amphinectria*	*Allonecte*	*?Amphinectria*
Thaxteriella	*Melioliphila*	*Malacaria*	*Boerlagiomyces*	*?Amphinectria*	*Aquaphila*
Tubeufia	*Paranectriella*	*Melioliphila*	*Borinquenia*	*Boerlagiomyces*	*?Boerlagiomyces*
	Podonectria	*Paranectriella*	*Byssocallis*	*Byssocallis*	*?Chaetocrea*
	Puttemansia	*Podonectria*	*Chaetocrea*	*Chaetocrea*	*Chaetosphaerulina*
	Rebentischia	*Puttemansia*	*Glaxoa*	*Chaetosphaerulina*	*?Glaxoa*
	Tubeufia	*Rebentischia*	*Letendraea*	*Glaxoa*	*Kamalomyces*
		Tubeufia	*Letendraeopsis*	*Letendraeopsis*	*?Malacaria*
		Uredinophila	*Malacaria*	*Malacaria*	*?Melioliphila*
			Melioliphila	*Melioliphila*	*?Paranectriella*
			Paranectriella	*Paranectriella*	*Podonectria*
			Podonectria	*Podonectria*	*?Puttemansia*
			Puttemansia	*Puttemansia*	*Rebentischia*
			Rebentischia	*Rebentischia*	*Thaxteriella*
			Taphrophila	*Taphrophila*	*Thaxteriellopsis*
			Thaxterina	*Thaxteriella*	*Tubeufia*
			Tubeufia	*?Thaxteriellopsis*	*?Uredinophila*
			Uredinophila	*Thaxterina*	
				Tubeufia	
				Uredinophila	

the Tarosoft (R) Image Frame Work program. Images used for figures were processed with Adobe Photoshop CS3 Extended version 10.0 software (Adobe Systems Inc., The United States). The type herbarium material is deposited in the herbarium of Mae Fah Luang University (MFLU), Chiang Rai, Thailand, and cultures are deposited in the Mae Fah Luang University Culture Collection (MFLUCC), BIOTEC Culture Collection (BCC) and IFRD culture collections, International Fungal Research & Development Centre, Kunming, China, the latter under material transfer agreement No. 4/2010 (MTA).

Isolation of fungi

Single spore isolates were cultured on water agar (WA) or malt extract agar (MEA, Difco Laboratories, Detroit, Michigan, USA). Germinating spores were transferred to MEA media and incubated at room temperature for 7 days. Isolates were grown for 1–2 months when morphological characters in culture, such as the extent of the mycelium, colour, shape and texture were recorded. These cultures were also used for DNA extraction and sequencing.

DNA extraction, PCR amplification and sequencing

Fungal mycelium was scraped from colonies growing on MEA. The DNA was extracted using the Biospin Fungus Genomic DNA Extraction Kit (BioFlux®, China), according to the manufacturer's instructions. Sequences of ribosomal RNA were amplified using the primer pairs NS1 and NS4, ITS5 and,ITS4, as well as LROR/LR5, which target small subunit, intergeneric spacer and large subunit regions, respectively (White et al. 1990; Vilgalys and Hester 1990). Amplification reaction mixtures contained 50 ng of template DNA, PCR Master Mix 1X, 0.5 μM of each primer in a 25 μL volume, 50 U of Taq DNA Polymerase 400 μM of each dNTP, 3 mM of $MgCl_2$. The PCR reaction used consisted of initial denaturation at 95°C for 2 min, 94°C for 1 min, 50°C for 1 min and 72°C for 1 min, and 35 cycles, followed by a final extension step of 72°C for 10 min. The PCR amplicons were visualized by staining with gel red or ethidium bromide on 1% agarose gel electrophoresis. The PCR products were purified and sequenced at Shanghai Sangon Biological Engineering Technology & Services Co., Ltd (China).

Phylogenetic analyses

A BLAST search was performed to verify identities of the cultures and to look for closely related sequences in the GenBank database. Analysis of sequence data (Online resource 1) were aligned using BioEdit (Hall 1999) and Clustal X (Larkin et al. 2007). The aligned datasets were analyzed using PAUP* v. 4.0b10 (Swofford 2002). Ambiguous regions in the alignments were excluded from the phylogenetic analyses. Maximum Parsimony (MP) was performed with stepwise additions of sequences using 1,000 random replicates and tree-bisection-reconnection (TBR) branch-swapping algorithm, with MAXTREES setting at 1000. The parsimony tree scores including tree length (TL), consistency index (CI), retention index (RI), rescaled consistency index (RC) and homoplasy index (HI) were also calculated. Bootstrap support for the branches was estimated based on 500 MP replicates with a single sequence addition replicates in each bootstrap replicate.

Models of nucleotide substitution for each gene were determined using MrModeltest 2.3 (Nylander 2004). GTR + I + G evolutional model were selected for likelihood analysis using MrBayes v. 3.1.2 (Ronquist and Huelsenbeck 2003). The Markov Chain Monte Carlo (MCMC; Rannala and Yang 1996) algorithm was used to estimate posterior probabilities (PP). Six MCMC chains were run from a random starting tree for 1,000,000 generations and trees sampled every 100 generations. The first 2,000 trees were discarded as burn-in prior to convergence of the chains. The remaining 10,001 trees based on 1,000 replicates were used to construct 50% majority rule consensus tree and to calculate posterior probabilities were determined more than 95% PP given each on branches.

Results

Ten new sequences were generated in this study. The new sequences of partial SSU and LSU were aligned with sequences from 42 taxa retrieved from GenBank (Online resource 1), using *Schismatomma decolorans* as an outgroup. The alignment of combined partial SSU and LSU rRNA contained 52 taxa and 1046 characters. Parsimony analysis of the dataset resulted in 90 trees (759 steps). Out of 1046 characters, 765 characters were constant and 222 were parsimony informative. A likelihood tree was generated by Bayesian analysis with both parsimony bootstrap values and posterior probabilities shown on each branch (Fig. 1a). The overall tree topology did not differ from previous analyses of Dothideomycetes (Schoch et al. 2009), with the fact that the placement of *Tubeufiaceae* was not clearly resolved. *Tubeufiaceae* appeared to form a distinct sister relationship to *Botryosphaeriales* and *Tubeufiaceae* might represent a new order (Fig. 1a). Most of the newly collected fungi nested in the *Tubeufiaceae*, forming a strong monophyletic cluster (Fig. 1a). Surprisingly, *Thaxteriellopsis lignicola* did not cluster with other representatives in *Tubeufiaceae* but became a sister taxon in the family *Tubeufiaceae* with 100% bootstrap support and 100% PP. Our material of *Thaxteriellopsis lignicola* highly resembled

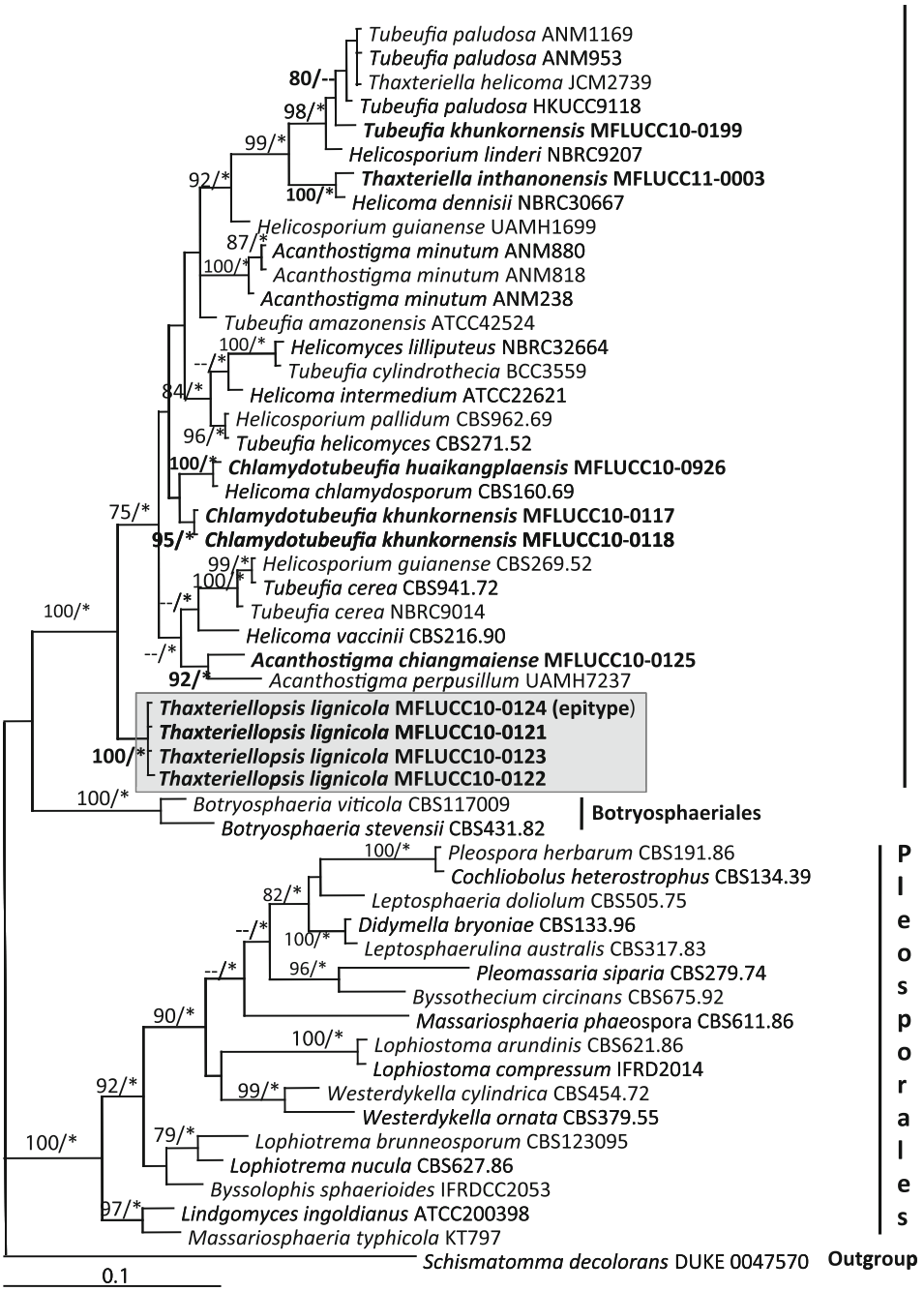


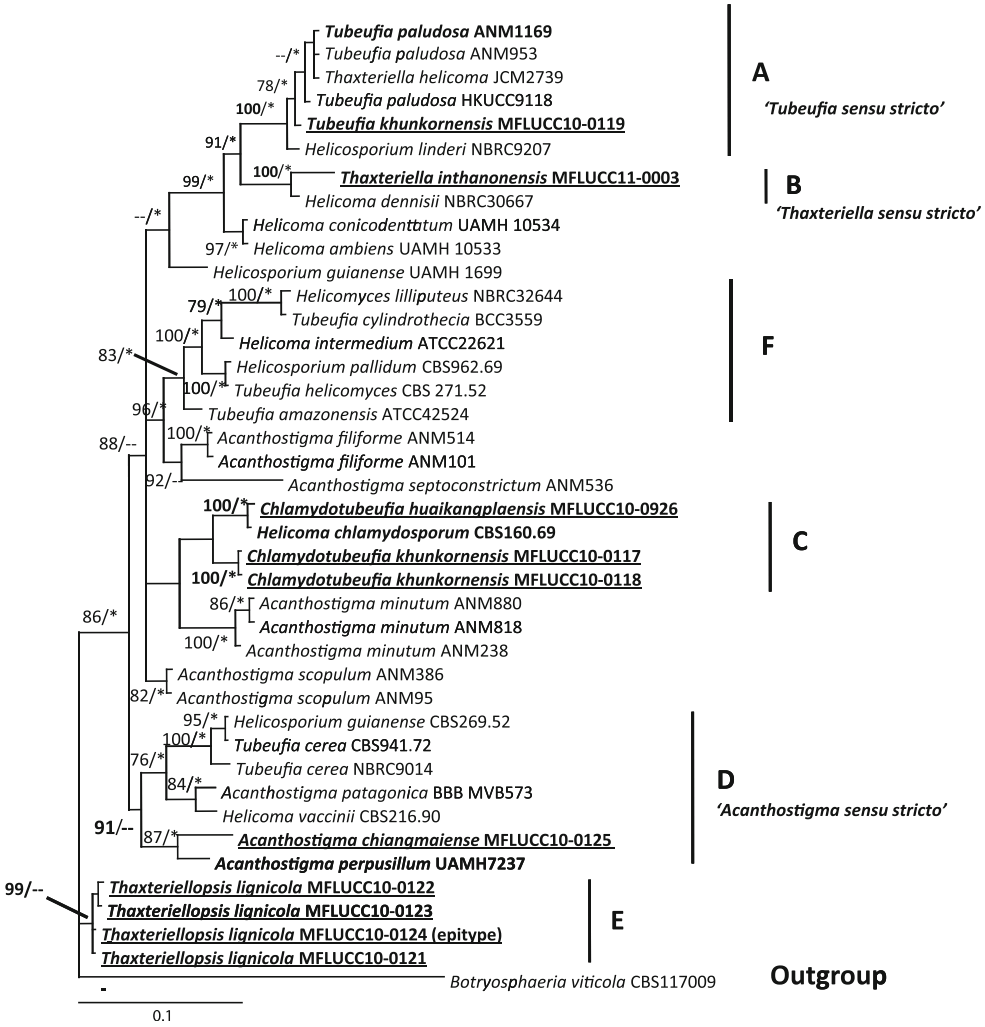
Fig. 1 **a** A likelihood tree showing the relationships of taxa in *Tubeufiaceae* to other ascomycetes, focusing on *Pleosporales*, inferred from partial SSU and LSU. The designated outgroup is *Schismatomma decolorans*. Parsimony bootstrap percentages over 75% and Bayesian posterior probabilities of more than 95% (marked by *) are shown above the branches. New collections of taxa in *Tubeufiaceae* are in bold. **b** One of the likelihood trees from MrBayes showing the phylogenetic relationships among representatives in *Tubeufiaceae* inferred from ITS and partial LSU. Bootstrap percentage values (>75%) generated from 500 replicates from maximum parsimony and posterior probabilities (.95%) from Bayesian analysis (marked by *) are shown above the branches. The designated outgroup is *Botryosphaeria viticola*. New collections of taxa in *Tubeufiaceae* are in bold and italic

the generic type of *Thaxteriellopsis*, which also belong in *Tubeufiaceae*, in terms of morphological features (Figs. 19 and 20). Therefore we designated one of our strains as the epitype of *Thaxteriellopsis lignicola* (see below).

The combined ITS and LSU sequence dataset consisted of 41 taxa and 1141 characters. MP analysis of the dataset resulted in 5 trees (1325 steps), and out of 1141 characters, 703 characters were constant and 334 were parsimony informative. Based on the results of parsimony and Bayesian analysis (Fig. 1b), representatives of *Tubeufiaceae* can be divided into multiple well-supported clades (>95% PP). Herein, we concentrate on clades A-F since we have performed thorough morphological examination of specimens within these clades, including five new taxa described in the current investigation (Fig. 1b).

Clade A comprised of six taxa and received strong bootstrap support (100%). This clade includes three strains of *Tubeufia paludosa,* the generic type of *Tubeufia,* the new species *T. khunkornensis* and *Helicosporium linderi,* plus a putative strain of *Thaxteriella helicoma*. This clade may represent *Tubeufia sensu stricto*. *Thaxteriella inthanonensis* clustered with *Helicoma dennisii* with high confidence

Fig. 1 (continued)



support in clade B (100% PP). We consider this clade as *Thaxteriella sensu stricto* because *T. inthanonensis* is morphologically very similar to the *T. corticola,* the generic type of *Thaxteriella*. Helicosporous anamorphs such as *Helicoma ambiens, Helicoma conicodentatum*, and *Helicosporium guianense* are closely related to members in clades A and B (Fig. 1b).

Members in clade C produced dictyochlamydosporous asexual stages, albeit the fact the clade received moderate bootstrap support (66%, data not shown on Fig. 1b). Two species are described under a new genus *Chlamydotubeufia* as *C. huaikangplaensis* and *C. khunkornensis* (Fig. 10, 12).

Clade D was considered as *Acanthostigma sensu stricto* because it contained *Acanthostigma perpusillum,* the type species. This clade also included a new species of *Acanthostigma* described in this paper as well as five other taxa. Clade E contained multiple strains of *Thaxteriellopsis lignicola*.

Tubeufia amazonensis, T. helicomyces, T. cylindrothecia, Helicosporium pallidum, Helicomyces lilliputeus and *Helicoma intermedium* clustered in a clade F with strong support (85% bootstrap support and 99% PP). Since the clade contained three *Tubeufia* species, this casts doubt on the monophyly of the genus *Tubeufia* because the type species nested in clade A.

Taxonomy

Tubeufiaceae M.E. Barr

Saprobic on dead plant material, especially wood, fungi or scale insects. *Ascomata* mostly forming on a subiculum, superficial, solitary, clustered to gregarious, globose to subglobose or turbinate, light yellow, orange, brown, dark brown to black, shiny, occasionally collapsing when dry, central ostiolate, papillate, with or without setae, or with mycelial covering. *Peridium* relatively wide, comprised of relatively thick-walked angular to globose cells. *Pseudoparaphyses* cellular, filiform, branched, anastomosing, attached to the hymenium and to the ascomata wall above. *Asci* 8-

spored, bitunicate, fissitunicate, usually cylindrical to broadly clavate, rounded at the apex, with or without an ocular chamber, with or without a pedicel. *Ascospores* filiform, cylindrical to narrowly fusiform, tapering towards the rounded to sub-acute ends, trans-septate, hyaline, pale yellow or brown, smooth-walled.

Anamorphs helicosporous, staurosporous or dictyosporous, belonging to *Annellospermosporella*, *Aquaphila*, *Araneomyces*, *Guelichia*, *Helicoma*, *Helicoon*, *Helicomyces*, *Helicosporium*, *Kamalomyces*, *Monodictys*-like, *Pendulispora*, *Peziotrichum*, *Tetracrium*, *Titaea* and *Xenosporium* (Hyde et al. 2011; Kirk et al. 2008).

Members of the *Tubeufiaceae* are easily recognized and usually occur on decaying woody plant material, including cloth, while there is a group that is fungicolous or occur in association with scale insects. The ascomata mostly form in a gregarious mass on a dark subiculum or are often covered in mycelium. Ascomata are usually dark and globose, but may also be light coloured. Asci are bitunicate, cylindrical and ascospores are fusiform to filamentous, trans-septate and hyaline or pale yellow or brown. The distinction between genera, is however, not clear and previous molecular studies have not resolved genera well.

Generic type: *Tubeufia.*

Accepted genera

Tubeufia Penz. & Sacc., Malpighia 11: 517 (1898) [1897].

= *Linobolus* Syd. & P. Syd., Annls mycol. 15(3/4): 204 (1917)

Saprobic on dead wood. *Ascomata* without a subiculum, superficial, solitary to gregarious, globose to subglobose, or clavate to obclavate, yellow to orange, becoming dark, occasionally hairy. *Peridium* composed of pseudoparenchymatous cells. *Asci* 8-spored, clavate to broadly cylindrical, apex rounded, short pedicellate (Fig. 2b). *Ascospores* cylindrical to fusiform, elongate, ends rounded, trans-septate, not constricted at septa, hyaline to pale yellow or brown, smooth-walled.

Anamorphs reported for genus: *Aquaphila* (Goh, K.D. Hyde & W.H. Ho 1998), *Helicoma* (Corda 1837), *Helicosporium* (Nees 1816), *Monodictys*-like, *Pendulispora* (M. B. Ellis 1961).

Type species

Tubeufia javanica Penz. & Sacc. Malpighia 11: 517 (1898) [1897] (Fig. 2)

Saprobic on culms of bamboo. *Ascomata* 350×150 μm, without a subiculum, superficial, solitary, gregarious, globose to subglobose or clavate to obclavate, white, orange, black, ostiole central occasionally hairy. *Peridium* pseudoparenchymatous (Fig. 2a). *Asci* 150–200×18–25 μm, (4–)8-spored, clavate to broadly cylindrical, apically rounded, short pedicellate (Fig. 2b). *Ascospores* 5–6 μm wide (spores length not reported), cylindrical to fusiform, elongated, ends rounded, trans-septate, hyaline to pale yellow or brown, smooth walled (Fig. 2c).

We have been unable to locate the type material, and our description and drawing are based on those provided by Penzig and Saccardo (1904). The generic type *Tubeufia javanica* was considered as a synonym of *T. paludosa* (Crouan & H. Crouan) Rossman, which is an earlier name (Rossman 1977). However, Barr (1980) considered Rossman's (1977) concept of *T. paludosa* to be rather broad and suggested that the collections grouped under this species eventually might be separated. Certainly, the illustrations of Penzig and Saccardo (1904) of *T. javanica* and those by Rossman (1977) of *T. paludosa* and Sivanesan (1984) of *T. paludosa* are quite different (perhaps stylized) and may not be the same species. Sequence data for a few strains of *Tubeufia*, including *T. paludosa*, are available in GenBank; however, it is difficult to establish if these are correctly named. The three strains of *T. paludosa* included in this study cluster together with *Thaxteriella helicoma*, with 65% bootstrap support. Fresh collections of *T. javanica* from Java are needed to establish if this is the same species as *T. paludosa* from a dead stem of *Rubus* in a stream bed in France, and both species need epitypifying with living cultures.

In this study we accept *Tubeufia* in a narrow sense, based on *T. javanica* and *T. paludosa*. Ascomata of *Tubeufia* are often globose to subglobose or clavate to obclavate, lack setae and may form on a subiculum, while asci have a wide thickened region at the apex and the ascospores are cylindrical to fusiform and trans-septate, and there is a helicosporous asexual state. Our new collections of *Tubeufia* also resemble *T. paludosa*; however, the ascomatal shape is slightly different. It does, however, cluster with putative strains of *Tubeufia paludosa* and *Thaxteriella helicoma* (Fig. 1b). Therefore, we consider *Tubeufia khunkornensis* sp. nov. to belong in *Tubeufia*.The anamorphs of *Tubeufia* are recorded as mostly helicosporous. *Aquaphila* is tubeufiaceous but did not cluster with the *Tubeufia paludosa* clade in the phylogenetic analyses (Tsui et al. 2006, 2007; Promputtha and Miller 2010) and appears to be a distinct genus. *Helicoma* and *Helicosporium* are shown to be polyphyletic in this and other studies (Tsui et al. 2006, 2007; Promputtha and Miller 2010; Fig. 1). The anamorphs of the genera *Tubeufia* will need to be reconsidered following this study.

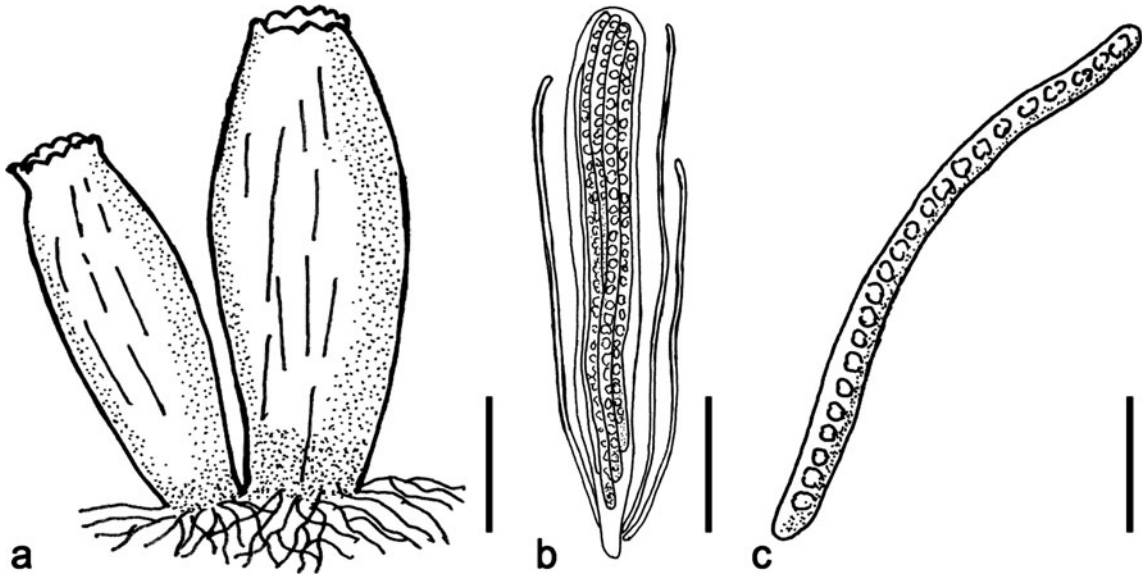


Fig. 2 *Tubeufia javanica* (redrawn from Penzig & Saccardo 1904). **a** Ascomata. **b** Ascus with pseudoparaphyses. **c** Ascospore. Scale bars: **a**= 100 μm, **b**, **c**=50 μm

Tubeufia khunkornensis Boonmee & K.D. Hyde, **sp. nov.** (Figs. 3 and 4)

MycoBank 563498

Tubeufia paludosa (Cr. & Cr.) Rossman. similis, sed ascospores (99–)111–118(−123)×(4–)5–7(−8) μm et 20–23-septate.

Etymology: from the Latin *–ensis* referring to an association with the *Khun Korn* Waterfall, the place of collection.

Saprobic on dead wood. *Ascomata* (133–)203–240 μm high×(193–)207–230 μm diam ($\bar{x}$= 192 × 210μm, n=5), borne on a dark brown subiculum, superficial, solitary or scattered, globose to subglobose, oval, to widely obclavate, brown to dark brown, ostiole unknown (Fig. 3a-c). *Peridium* 39–42 μm thick, comprised of several-layers of cells of *textura angularis*; inner cells hyaline to pale brown and angular, outer layer cells brown to dark brown and subglobose (Fig. 3d). *Hamathecium* cellular, filiform, *ca*. 2 μm wide, hyaline pseudoparaphyses, embedded in a gelatinous matrix (Fig. 3e). *Asci* (128.5–)130–153(−161)× (−13)16–19 μm ($\bar{x}$ = 141 × 17μm, n=20), 8-spored, bitunicate, cylindrical, apex thickened and rounded, large apical area lacking contents, ocular chamber not observed, with a 20–25 μm long pedicel (Fig. 3f, g). *Ascospores* (99–)111–118(−123)×(4–)5–7(−8) μm ($\bar{x}$ = 114 × 6μm, n=20), 3–4-seriate, cylindrical to fusiform, narrowly elongate, straight or slightly curved, 20 –23-septate, tapering towards the sub acute to rounded ends, not constricted at the septa, hyaline to pale grayish, smooth-walled (Fig. 3h-k).

Colonies in culture: Ascospores germinating on MEA within 24–36 h and germ tubes produced from both ends (Fig. 4a). Colonies growing slowly on MEA, reaching 9 mm in 1 week at 28°C, effuse, velvety to hairy, edge fimbriate, olive to olive brown, pigmented in media (Fig. 4b, c). Mycelium superficial and immersed, branched, septate, smooth, pale yellowish brown to reddish brown. Club-shaped, brown, muriform conidia-like structures formed on hyphae (Fig. 4d-g).

Material examined: THAILAND, Chiang Rai, Muang, Khun Korn Waterfall, elev. as 671 msl., N19°51–54' E 99° 35.39', on dead wood of unidentified trees, 13 November 2009, S. Boonmee KK-08 (MFLU10–0052, holotype), ex-type culture MFLUCC10–0119 = IFRD2180 = BCC.

Habitat: terrestrial, saprobic, on dead wood of unidentified trees.

Known distribution: Chiang Rai, Thailand.

Tubeufia khunkornensis is different from *T. javanica* and *T. paludosa* in ascomatal shape and colour, being clavate to obclavate and pale yellow to orange for the latter and dark for the former (Fig. 2; Penzig and Saccardo 1904; Samuels et al. 1979). Based on DNA sequences comparisons, *T. khunkornensis* is related to *T. paludosa* and *Thaxteriella helicoma* and receives 92% Bayesian support (Fig. 1).

Acanthostigma De Not., Sfer. Ital., 85 (1863).

Saprobic on dead plant debris. *Ascomata* superficial, scattered, globose to subglobose, reddish-brown to dark brown, collapsing when dry, sparsely setose on the upper part, setae dark brown to opaque. *Peridium* consisting of several-layers of polyhedral cells. *Hamathecium* numerous, cellular, branching and anastomosing pseudoparaphyses developing from the basal hymenium. *Asci* 8-spored, bitunicate, fissitunicate, clavate, broadly rounded and thickened at the apex, short pedicellate. *Ascospores* fusiform, narrowly rounded at both ends, one of middle cells often broader than the others, trans-septate, straight or slightly curved, not-constricted or slightly constricted at the septa, hyaline.

Anamorphs reported for genus: *Helicomyces*, *Helicosporium* (Hyde et al. 2011)

Type species

Acanthostigma perpusillum De Not., Sfer. Ital.: 207 (1863) (Fig. 5)

Acanthostigma clintonii (Peck) Sacc., Syll. fung. (Abellini) 2: 210 (1883)
Sphaeria clintonii Peck, Ann. Rep. N.Y. St. Mus. nat. Hist. 30: 65, tab. 2, Figs. 19 and 20 (1878) [1877]
Tubeufia clintonii (Peck) M.E. Barr, Mycotaxon 12 (1): 163 (1980)

Saprobic on dead bark. *Ascomata* 100–110 μm high × 150–155 μm diam, superficial, scattered, globose to subglobose, reddish-brown to dark brown, collabent towards when dry, sparsely setose on the upper part. *Setae* 1-celled, dark brown, opaque, acute, (10–)28–97 μm long × 5–6 μm wide at the base. *Peridium* 15–22 μm thick, comprising 3–4 layers of polyhedral, pale brown to brown, thick-walled cells (Fig. 5a). *Hamathecium* 2–3.5 μm wide, numerous, cellular, branched, anastomosing pseudoparaphyses developing from the basal hymenium (Fig. 5b). *Asci* 77–79 × 14–16 μm, 8-spored, bitunicate, fissitunicate, cylindro-clavate, broadly rounded and thickened at the apex, ocular chamber not observed, short-pedicellate (Fig. 5c). *Ascospores* 30.5–35.5(−42) × 5–6 μm, 2–4-seriate in the ascus, fusiform to clavate, narrowly rounded at both ends, one of middle cells often broader than the others, (5–)6–7(−8)-septate, straight or slightly curved, not-constricted or slightly constricted at the septa, hyaline (Fig. 5d).

Description and drawing based on Réblová and Barr (2000).

Acanthostigma was monographed by Réblová and Barr (2000) and six species were accepted. Berlese (1894), and Réblová and Barr (2000) redescribed the type and provided illustrations and a detailed description. We were unable to locate the type species during the time frame of this study and therefore refer to the excellent description of the type provided by Réblová and Barr (2000).

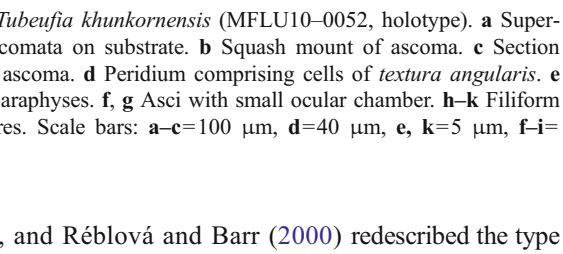


Fig. 3 *Tubeufia khunkornensis* (MFLU10–0052, holotype). **a** Superficial ascomata on substrate. **b** Squash mount of ascoma. **c** Section through ascoma. **d** Peridium comprising cells of *textura angularis*. **e** Pseudoparaphyses. **f**, **g** Asci with small ocular chamber. **h–k** Filiform ascospores. Scale bars: **a–c**=100 μm, **d**=40 μm, **e, k**=5 μm, **f–i**=50 μm

Acanthostigma was confirmed as belonging to *Tubeufiaceae* based on LSU and SSU, ITS and LSU rDNA sequence analysis (Tsui et al. 2007; Schoch et al. 2009, Promputtha and Miller 2010). In our study *Acanthostigma* is polyphyletic within *Tubeufiaceae* with representatives scattered all over the tree (Fig. 1b). *Acanthostigma chiangmaiense* sp. nov. clustered with *A. perpusillum* (strain no. UAMH 7237, Tsui and Berbee 2006) with 87% bootstrap support, and can be considered to belong in *Acanthostigma sensu stricto*. *Acanthostigma minutum, A. scopulum, A. filiforme* and *A. septoconstrictum* however, were distant from *A. perpusillum* and clustered in other clades (Fig. 1b). Promputtha and Miller (2010) also assigned *A. perpusillum* in *Tubeufiaceae*, although Kodsueb et al. (2006) placed one collection of *A. perpusillum (*AY856892) in the *Herpotrichiellaceae* clade with high bootstrap support.

Both *Helicomyces* and *Helicosporium* have been reported as anamorphs of *Acanthostigma* and are polyphyletic (Tsui et al. 2007; Promputtha and Miller 2010) and *A. chiangmaiense* only formed chlamydospores in culture. Thus further work is required to establish the anamorphs of *Acanthostigma sensu stricto*.

Acanthostigma chiangmaiense Boonmee & K.D. Hyde, **sp. nov.** (Figs. 6 and 7)

MycoBank 563499

Acanthostigma perpusillum De Not. similis sed ascosporis (35–)37–44(−48)×5–8 μm differt.

Etymology: From the Latin *–ensis,* meaning associated with and Chiang Mai in reference to the type locality of Chiang Mai Province.

Saprobic on dead wood. *Ascomata* (75–)108–118.5 μm high × (71–)113–123(−146.5) μm diam ($\bar{x} = 99 \times 108.5\mu m, n = 5$, n=5), superficial, solitary, scattered, globose to subglobose, dark brown to black, ostiole central, covered with black 26.5–74 μm long, 1-celled, tapering setae (Fig. 6a-c). *Peridium* 11.5–18 μm wide, dark

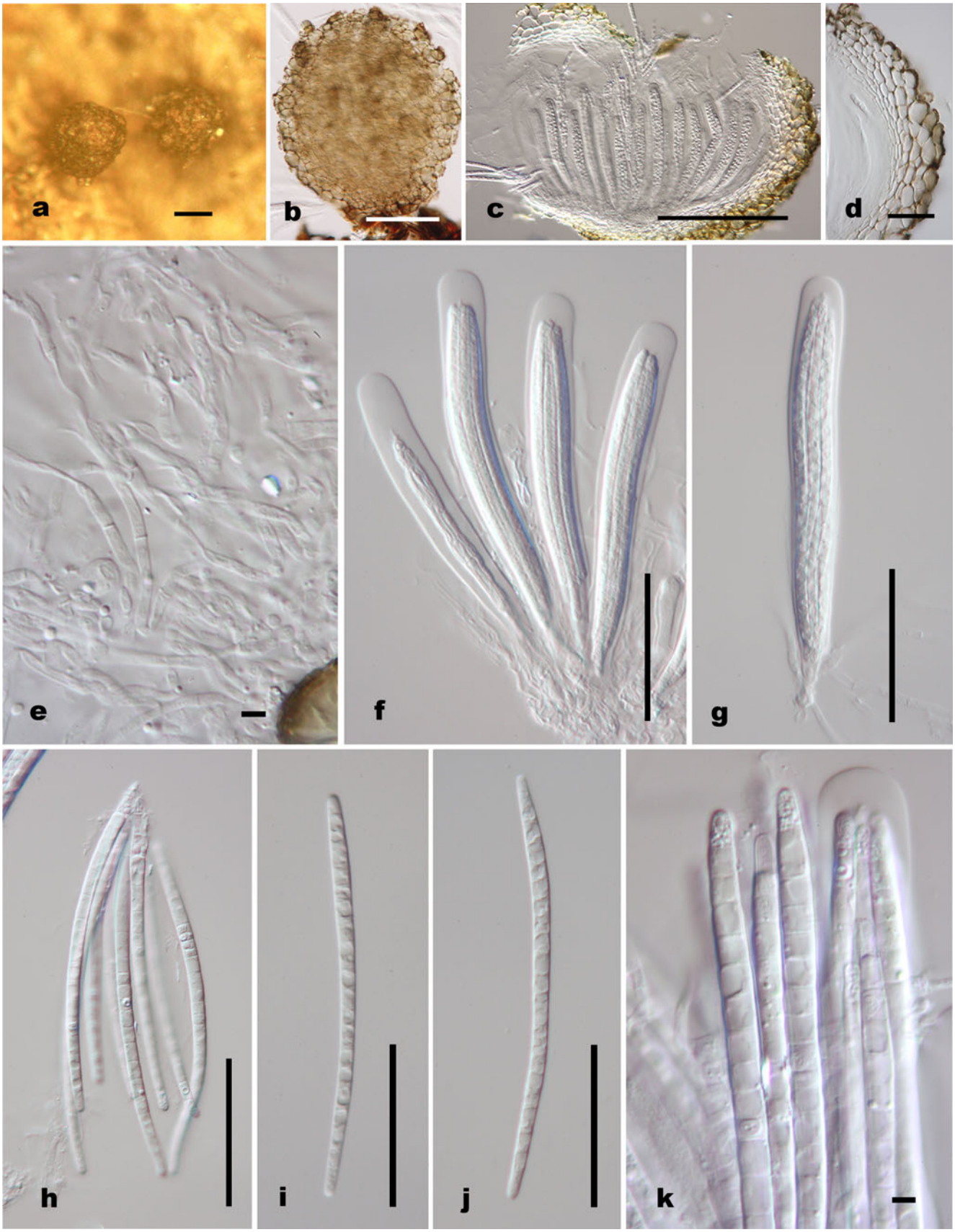

a
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Fig. 4 *Tubeufia khunkornensis* (MFLUCC10–0119, holotype). Colonies on MEA. **a** Germination of ascospore. **b, c** Colonies on MEA from above and below. Note colonies are olive to olive brown with pigment diffusing in the media. **d–i** Mycelium development on culture. Note the brown, club-shaped, muriform conidia-like structures formed at the ends of hyphae. Scale bars: **a**=50 μm, **b, c**=10 mm, **d–i**=10 μm

brown, a single stratum comprised of 3–4 cell layers of *textura angularis* (Fig. 6d). *Hamathecium ca*. 2 μm wide, filiform, cellular, anastomosing pseudoparaphyses surrounded by a gelatinous matrix, forming from the hymeni-

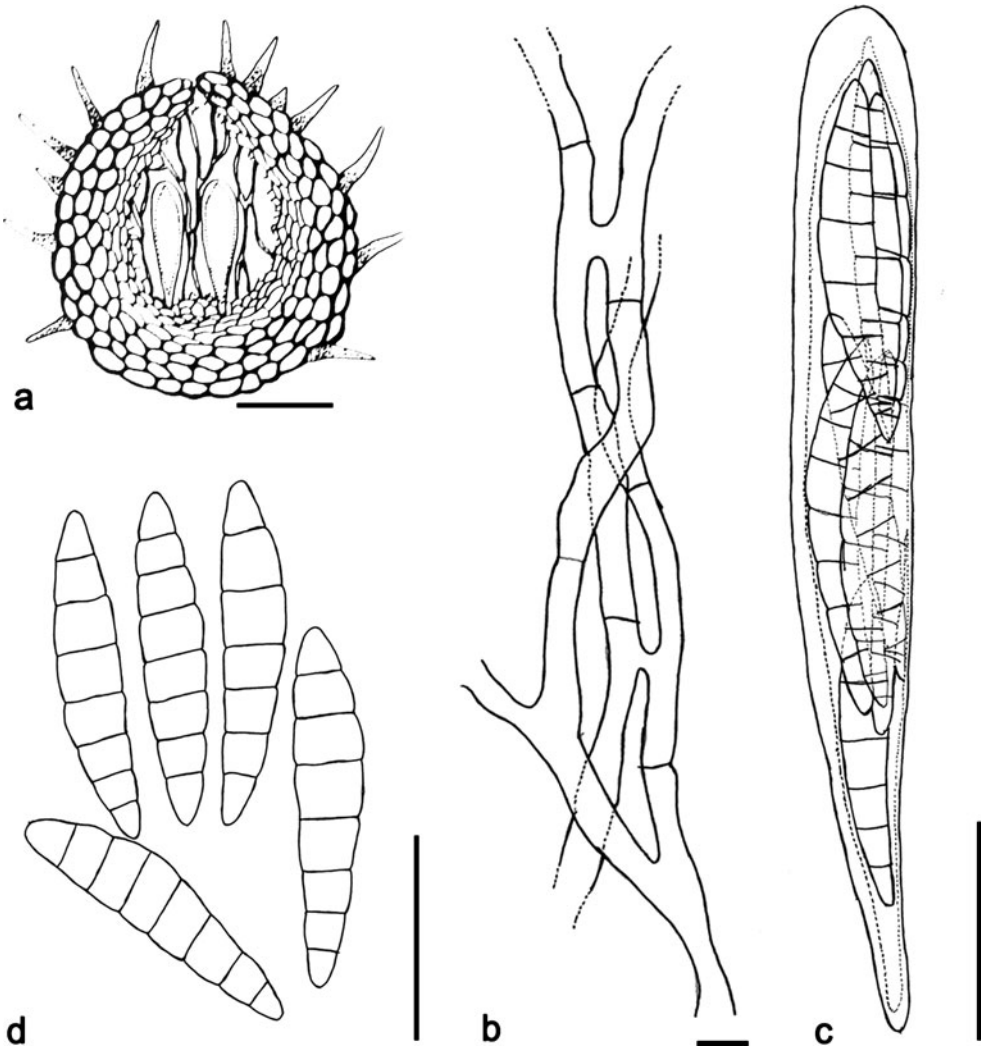

Fig. 5 *Acanthostigma perpusillum* (redrawn from Réblová and Barr 2000). **a** Ascomata cross section on substrate. **b** Pseudoparaphyses. **c** Ascus. **d** Ascospores. Scale bars: **a**=50 μm, **b**=5 μm, **c, d**=20 μm

um at the base/and sides of the ascoma (Fig. 6e). *Asci* (62.5–)70–92(−94)×(16–)17.5–20(−24.5) μm ($\bar{x}$= 79 × 18μm, *n*=20), 8-spored, bitunicate, fissitunicate, cylindro-clavate, broadly rounded at the apex (some with broadly rounded upper part), with a small ocular chamber, short pedicellate (Fig. 6f-h). *Ascospores* (35–)37–44(−48)× 5–8 μm ($\bar{x}$= 40 × 6μm, *n*=20), 3–4-seriate, fusiform-clavate, slightly tapering toward and sub acute ends, straight or slightly curved, 8–9-septate, widest at the third to fourth cell from the apex, not noticeably constricted or slightly constricted at the septa, hyaline, smooth walled (Fig. 6i-l).

Cultural characteristics: Ascospores germinating on WA within 12 h and germ tubes produced from both ends (Fig. 7a). Colonies growing slowly on MEA, reaching a diam of 2 mm. after 7 days at 28°C, effuse, velvety, entire to slightly undulate edge, dark to blackish (Fig. 7b, c). Mycelium superficial, branched or anastomosing, septate, smooth, subhyaline to pale brownish and olivaceous brown. Chlamydospores forming between hyphae, olive brown, becoming dark brown with age, globose, at first single celled, becoming multicellular, muriform, (16–)23–37 μm diam (Fig. 7d-m).

Material examined: THAILAND, Chiang Mai, Mae Rim, Mae Sa, Pong Yaeng, elev. *ca* 1000 msl., on dead wood of unidentified trees, 7 September 2009, S. Boonmee, PYW-02 (MFLU10–0058, holotype), ex-type culture MFLUCC10–0125 = IFRD2198 = BCC.

This new species belongs in *Acanthostigma* because it has similar morphological features such as a darkened ascomata surrounded by setae, cylindric-clavate asci and clavate-fusiform, trans-septate hyaline ascospores; it is also very similar to *A. perpusillum* (Réblová and Barr 2000). The taxon grouped with *A. perpusillum*, the generic type in the phylogenetic analysis with high support (100% PP). *A. chiangmaiense* can be distinguished from *A. perpusillum* by dimensions and shape of the ascospores, being (35–)37–44(−48)×5–8 μm and 8–9-septate in the

Fig. 6 *Acanthostigma chiangmaiense* (MFLU10–0058, holotype). **a** Superficial ascomata on substrate. **b** Section through of ascoma. Note ascomata are covered with dark brown setae. **c** Seta. **d** Section of peridium comprising brown cells of *textura angularis*. **e** Pseudoparaphyses, which are cellular and embedded in a mucilaginous matrix. **f–h** Young and mature asci with eight 3–4-seriate ascospores. Note asci are short-stalked. **i–l** Ascospores with 8–9-transverse septa. Scale bars: **a–c**=50 μm, **d**, **i–l**=10 μm, **e**=5 μm, **f–h**=30 μm

former and 30.5–35.5(−42)×5–6 μm and (5–)6–7(−8) septate in *A. perpusillum*. The species are however closely related. It is not similar to any other species of *Acanthostigma* (Réblová and Barr, 2000; Promputtha and Miller, 2010).

Acanthophiobolus Berl., Atti Congl. Bot. Intern. di Genova, 1892: 571 (1893) [1892].

Saprobic on cloth. *Ascomata* superficial, globose to subglobose, reddish brown to dark brown, ostiole central,

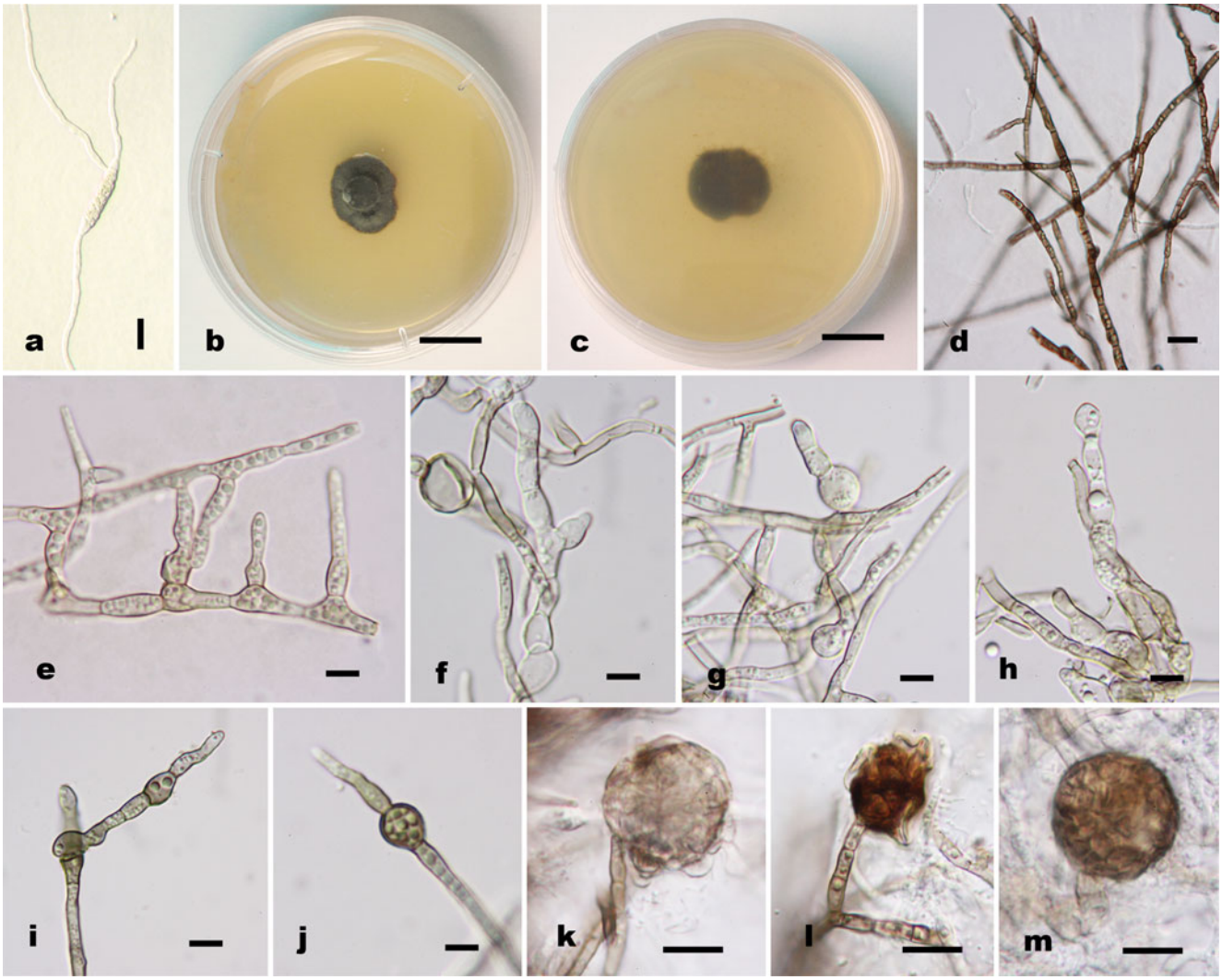

Fig. 7 *Acanthostigma chiangmaiense* (MFLUCC10–0125, holotype). Colonies on MEA. **a** Germinating ascospore. **b–c** Colonies on MEA from surface and reverse. Note colonies are dark brown. **d–m** Mycelium development on culture. Note the formation of chlamydospores. Scale bars: **a, k–m**=10 μm, **b–c**=5 mm, **d–j**=5 μm

with red brown to dark brown setae, which are septate, and taper towards the apex. *Peridium* comprising several-layers; inner layer composed of *textura prismatica-porrecta* and hyaline to pale, outer layer composed of 2–3 layers of dark brown cells of *textura angularis*. *Hamathecium* comprising filiform pseudoparaphyses. *Asci* 8-spored, bitunicate, elongate, cylindro-clavate, short pedicellate. *Ascospores* filiform, septate, spiral in the ascus, hyaline.

Anamorphs reported for genus: none.

Type species

Acanthophiobolus helminthosporus (Rehm) Berl., Die Pilze des Weinstockes, Vienna: 571 (1893) [1892] (Fig. 8)

≡ *Leptospora helminthospora* Rehm

Saprobic on rotten cloth. *Ascomata* 82–122×68–111 μm ($\bar{x}$ = 101 × 96μm, n=5), superficial, globose to subglobose, brown to dark brown or red brown, ostiole central, covered in setae which are 82–163.5 μm long, brown to red brown, darkened at the base, septate, and taper towards the apex (Fig. 8a-e). *Peridium* comprising two substrata, an inner layer composed of hyaline to pale cells of *textura prismatica-porrecta* and an outer layer composed of 2–3 layers of dark brown cells of *textura angularis* (Fig. 8f). *Hamathecium* filiform, 1–1.5 μm wide, pseudoparaphyses developing from the basal hymenium (Fig. 8g). *Asci* 126–168×8–10 μm ($\bar{x}$= 148 × 9μm, n=20), 8-spored, bitunicate, elongate cylindric-clavate, ocular chamber not observed, short pedicellate (Fig. 8h-i). *Ascospores* filiform, spiral in the ascus, hyaline (ascospores length could not be recorded but they were longer than asci) (Fig. 8j).

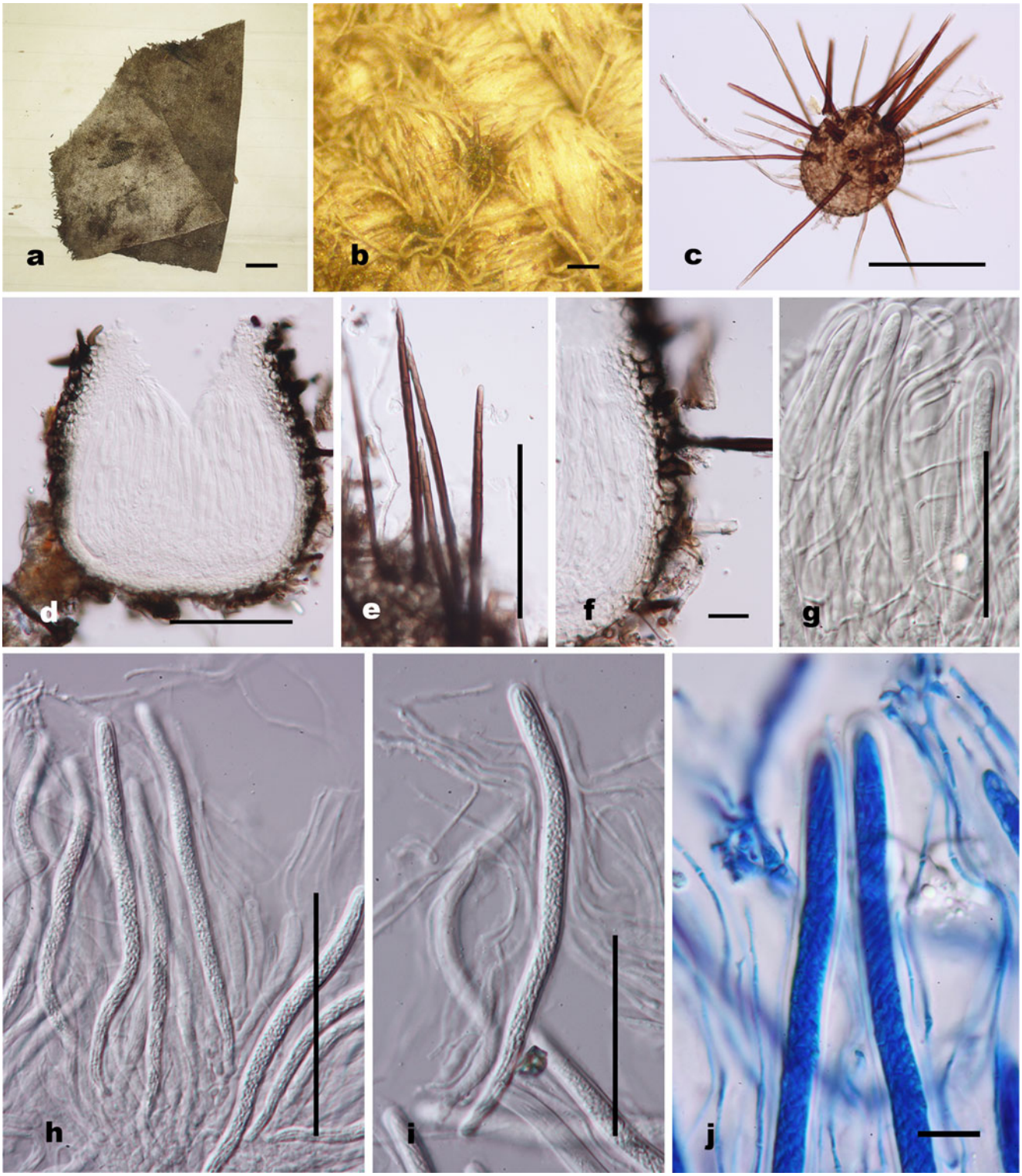

Fig. 8 *Acanthophiobolus helminthosporus* (S, holotype). **a** Habit of fungus on rotten cloth. **b** Ascomata on substrate. **c** Squash mount of ascoma. **d** Section of ascoma. **e** Setae. **f** Peridium. **g** Pseudoparaphyses. **h**, **i** Asci. **j** Ascospores spiraled in asci. Scale bars: **a–e**, **g–i**=100 μm, **f**, **j**=10 μm

***Material examined*:** GERMANY, Augsburg, on rotten cloth, 14 February 1880, M. Britzelmayr No. F12985 (S, holotype).

Acanthophiobolus was introduced by Berlese (1893) and six species are listed in *Index* Fungorum. An earlier name was found in *Acanthophiobolus helicosporus* (Berk. &

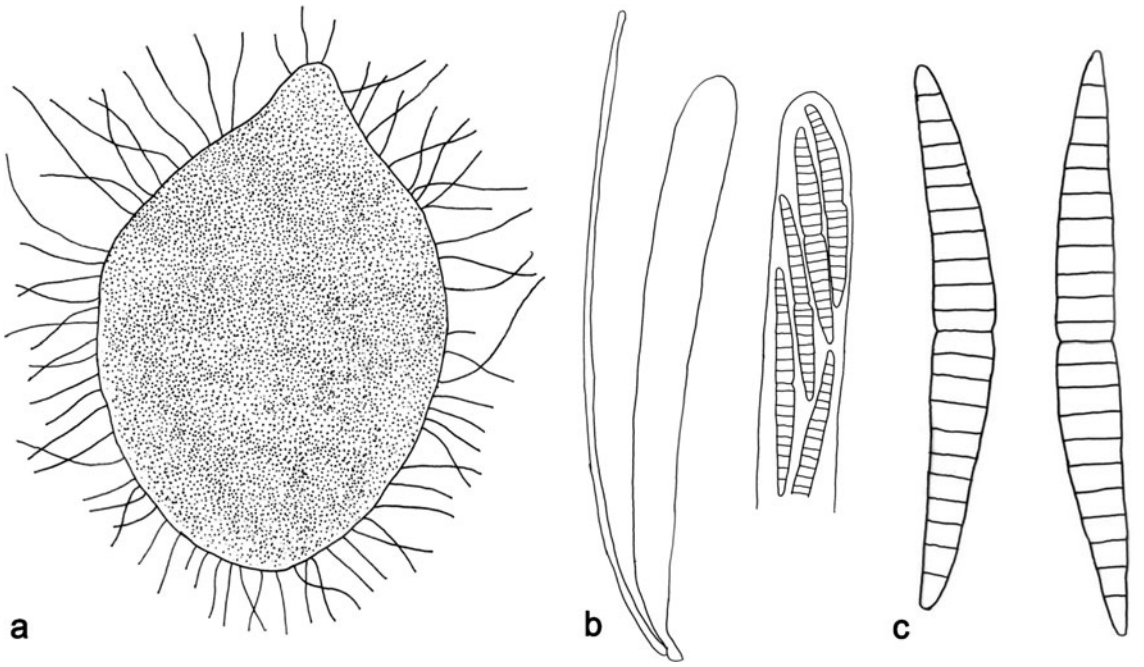

Fig. 9 *Chaetosphaerulina yasudai* (redrawn from Hino 1938) redrawn from Hino. **a** Ascoma. **b** Asci with paraphyses. **c** Ascospore

Broome) J. Walker based on *Sphaeria helicospora* Berk. & Broome and *Acanthophiobolus helminthosporus* was considered to be a synonym (Walker 1980). Ascomata are superficial on the substrate and globose to subglobose, covered by red brown to dark brown and long setae. Asci are cylindrical with filiform and spiral ascospores. It was not possible to obtain single ascospores from the asci for measurement as they were spiraled in the asci (Walker, 1980; Fig. 8h-j).

No molecular phylogenetic study has been conducted to *Acanthophiobolus*. Although the ascoma characters are somewhat similar to *Acanthostigma*, *Acanthophiobolus* clearly differs in having bright colour ascomata, septate setae which are much longer, and cylindrical, elongate asci with long filiform, trans-septate ascospores (Figs. 9, 12). We therefore maintain *Acanthophiobolus* is a distinct genus in *Tubeufiaceae*. The type needs recollecting and epitypifying.

Chaetosphaerulina I. Hino, Bulletin Miyazaki Coll. Agric. Forest. 10: 62 (1938)

Chaetosphaerulina (generic type = *C. yasudai* I. Hino 1938). *Saprobic* on dead wood. *Subiculum* superficial, attenuate apex, velutinous, dark to black, irregular, effuse brown hyphae, septate, attenuate apex. *Ascomata* on subiculum, ovoid to ellipsoid, ostiolate, hairy surrounded. *Hamathecium* filiform, cellular pseudoparaphyses. *Asci* 8-spored, bitunicate, cylindrical, slightly truncate apex. *Ascospores* hyaline, multiseptate, constricted at the median. *Chlamydospores* at the apex of hyphae, with verticillate production, 2–8-septate, oblong, fusoid to vermiform, frequently curved.

Anamorph reported for genus: *Xenosporium* (Hyde et al. 2011)

Type species

Chaetosphaerulina yasudai I. Hino, Canad. J. Plant Sci. 10: 62 (1938) (Fig. 9)

≡ *Herpotrichia yasudai* (I. Hino) Piroz. [as '*yasudae*'], Mycol. Pap. 129: 16 (1972)

≡ *Tubeufia yasudai* (I. Hino) Sivan. [as '*yasudae*'], Bitunicate Ascomycetes and their Anamorphs (Vaduz): 589 (1984)

Saprobic on culms of bamboo. *Subiculum* 311–555×3–6 μm, superficial, dark to black, comprising effuse brown septate hyphae, attenuate at the apex. *Ascomata* 289–366×244–333 μm (Fig. 9a), forming on a subiculum, ovoid to ellipsoid, ostiolate, hairy. *Hamathecium* filiform, cellular pseudoparaphyses. *Asci* 208–221×10–16 μm (Fig. 9b), 8-spored, bitunicate, cylindrical, with slightly truncate apex. *Ascospores* 61–67×6 μm (Fig. 9c), hyaline, 21-septate, constricted at the median. *Chlamydospores* 11–61×5–16 μm, forming at the ends of hyphae, oblong, fusoid to vermiform, frequently curved, verticillate, 2–8-septate.

Thaxteriellopsis bambusicola Sivan. & N.D. Sharma is very similar to *Chaetosphaerulina yasudai* and was

transferred to *Chaetosphaerulina bambusicola* (Sivan. & N. D. Sharma) by Crane et al. (1998). The ascospores overlap in size but *C. bambusicola* differs in have a few longitudinal septa. These may well turn out to be the same species but needs recollecting.

Chlamydotubeufia Boonmee & K.D. Hyde, **gen. nov**.

MycoBank 563500

Acanthostigma De Not. similis sed status anamorphosis dictyochlamydosporae differt.

Etymology: A combination of *Chlamydo-* and *–tubeufia* in reference to the similarity to the genus *Tubeufia* but dictyochlamydosporous anamorphs.

Saprobic on dead wood. *Ascomata* superficial, solitary, scattered or in small groups, globose or slightly subglobose, with or without an ostiole, covered by darkened setae. *Peridium* dark brown to black. *Hamathecium* cellular pseudoparaphyses, embedded in a mucilaginous matrix. *Asci* 8-spored, bitunicate, fissitunicate, cylindro-clavate, broadly clavate, rounded at the apex short-pedicellate. *Ascospores* 2–3-seriate, hyaline, narrowly fusiform, broad at supra-median, slightly curved, multiseptate, slightly constricted septum, ends asymmetrical. *Anamorph* helicosporous and also producing a dictyochlamydosporous state in culture and often on wood. Dictyochlamydospores broadly-oblong, elongate, multiseptate, at first red brown becoming darkening to black.

The new genus is introduced to accommodate *Tubeufia*-like species with globose ascomata, a helicosporous anamorph form as well as producing dictyochlamydospores. *Helicoma chlamydosporum* Shearer also have dictyochlamydospores and clustered with the three *Tubeufia*-like species producing dictyochlamydospores in culture. This cluster clearly separates from *Tubeufia* species in Clade A which include the generic type of *Tubeufia paludosa* and therefore we introduce the new genus *Chlamydotubeufia* based on morphological and molecular data. The helicospores produced in *H. chlamydosporum* are produced on blunt-tipped denticles on swellings on the hyphae, directly on hyphae, or on short lateral 0–3-septate conidiophores whereas in most typical *Helicoma* species conidiophores are rather long (Shearer 1987; Zhao et al. 2007).

We introduce two new species and transfer *H. chlamydosporum* and *H. depressispora* to *Chlamydotubeufia*, the latter also producing dictyochlamydospores. Another genus with similar chlamydospores and an aquatic habitat is *Intercalispora* and this may be an earlier name for *Chlamydotubeufia*. The type *I. nigra* is illustrated in Seifert et al. (2011) and although quite similar to *Chlamydotubeufia*, the septation in *Intercalarispora nigra* J.L. Crane & Schokn. is irregular, while in *Chlamydotubeufia* species the septa is distinctly transverse with up to two vertical septa. It is not apparent if this different has any taxonomic value in separating genera but until *Intercalarispora* is recollected, isolated and subjected to sequence analysis we prefer to use the new name *Chlamydotubeufia.* Another species with *Intercalarispora*-like chlamydospores is *Tubeufia amazonensis* Samuels, Rossman & E. Müll.; the ascomata differ from *Chlamydotubeufia* as they lack setae in this species (Samuels and Müller 1978). Seifert et al. (2011) also illustrate a *Monodictys* species as being anamorphic *Tubeufiaceae,* this however, may also be a species of *Intercalarispora,* but with the apical hyphae missing giving it the appearance of *Monodictys*.

Type species:

Chlamydotubeufia huaikangplaensis Boonmee & K.D. Hyde, **sp. nov**. (Figs. 10 and 11)

MycoBank 563501

Acanthostigma perpusillum De Not. similis sed ascospores 32–41.5×5–8 μm et 5–6-septate et status anamorphosis dictyochlamydosporae (45–)50–77×(25–)39–42 μm differt.

Etymology: from *–ensis* meaning pertaining to and *Huai Kang Pla,* the place of collection in reference to the collecting site.

Saprobic on dead wood of *Pinus*. *Ascomata* 161–192 μm high×142–184 μm diam ($\bar{x}$= 173 × 160μm, n=5), superficial, solitary or scattered, globose to subglobose, black, with a central ostiolate, covered with darkened and one-celled setae (Fig. 10a-c). *Peridium* 17–23 μm thick, a single substratum, composed of 3–4 layers of cells of *textura angularis* (Fig. 10d). *Hamathecium* 2–2.5 μm wide, branched, hyaline pseudoparaphyses embedded in a gelatinous matrix (Fig. 10e). *Asci* 50–83×16–21 μm ($\bar{x}$= 64 × 18μm, n=20), 8-spored, bitunicate, fissitunicate, cylindrical to clavate or saccate, apically rounded, ocular chamber not apparent, short pedicellate (Fig. 10f-i). *Ascospores* 32–41.5×5–8 μm ($\bar{x}$= 37 × 7μm, n=20), fusiform, slightly curved, guttulate, 5–6-septate, slightly constricted at the septa, hyaline to pale gray (Fig. 10j-l).

Colonies in culture: Ascospores germinating on MEA within 24–36 h and germ tubes produced from both ends (Fig. 11a). Colonies growing on MEA slowly, reaching 9 mm in 1 month at 28°C, flat to slightly effuse, edge entire-erose or dentate, darkened to blackish (Fig. 11b). Mycelium mostly superficial, branched, septate, smooth, subhyaline to pale brown, hyphae becoming dark brown

Fig. 10 *Chlamydotubeufia huaikangplaensis* (MFLU10–0972, holotype). **a** Ascomata on substrate. **b** Ascoma cross section. **c** Setae. **d** Peridium comprising *textura angularis* with 3–4 layers. **e** Pseudoparaphyses. **f–i** Asci with biseriate ascospores, short-stalked. **j–l** Ascospores with 6–7 cells. Scale bars: **a–b**=100 μm, **c–l**=20 μm

due to the development of dictyochlamydospores (Fig. 11c-e). Conidia (45–)50–77 μm long, (25–)39–42 μm wide ($\bar{x}$= 56 × 35μm, n=20), blastic, broadly oval to ellipsoid, dictyoseptate, light coloured when immature, becoming darkened to black when mature (Fig. 11f-k).

Material examined: THAILAND, Chiang Rai, Mae Chan, Huai Kang Pla Waterfall, elev. 512 msl., N19°51–54' E 99° 35.39', on dead wood of *Pinus*, 25 October 2010, S. Boonmee, HKP-01 (MFLU10–0972, holotype), ex-type culture MFLUCC10–0926 = BCC.

Chlamydotubeufia huaikangplaensis has superficial, globose to subglobose, black ascomata covered with 1-celled darkened setae, clavate asci, and fusiform-oblong, hyaline, trans-septate ascospores. In addition, dictyochlamydospores

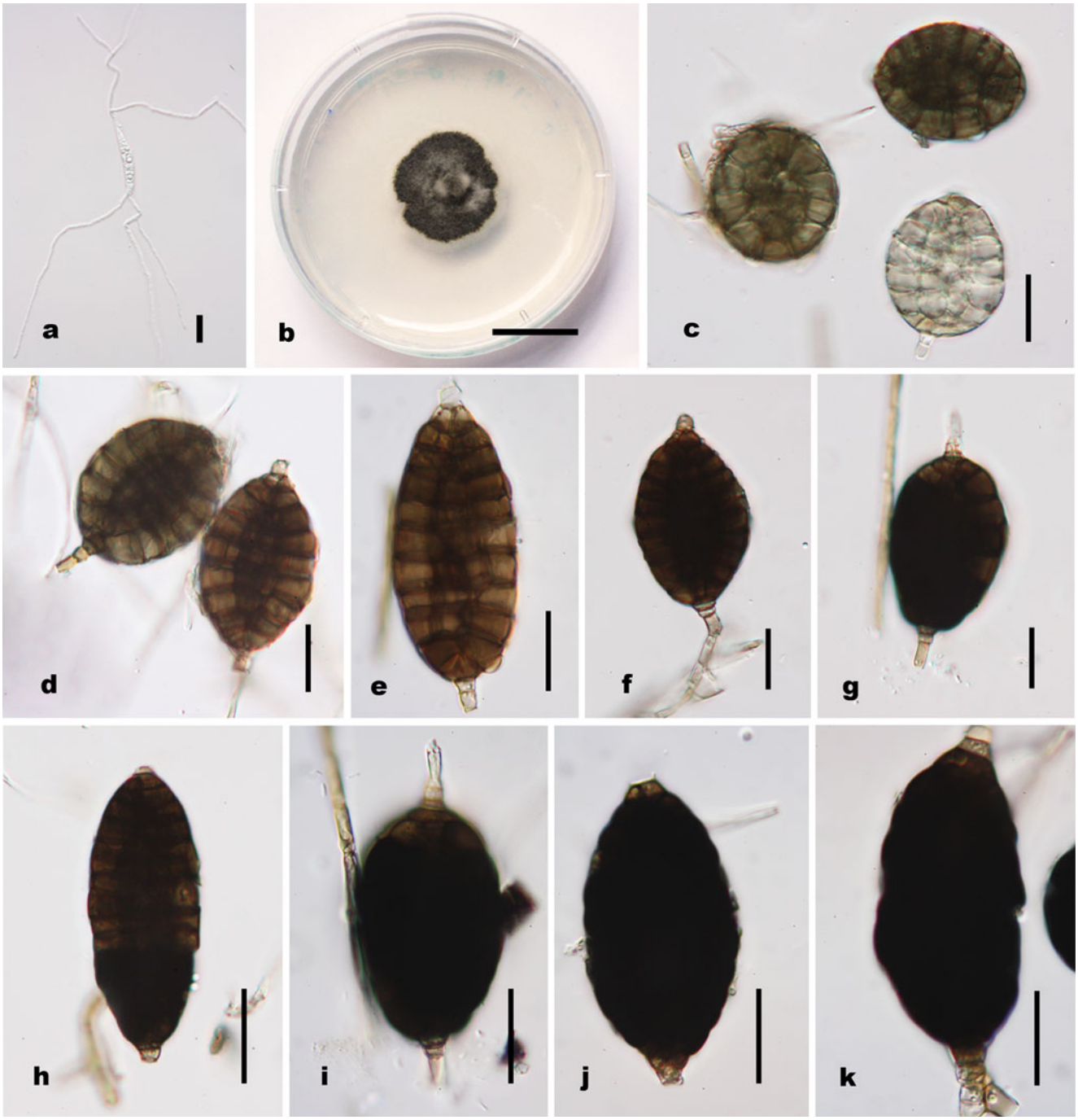

Fig. 11 *Chlamydotubeufia huaikangplaensis* (MFLUCC10–0926, holotype). Colonies on MEA. **a** Germination of ascospore. **b** Colonies on MEA in surface view. Note colonies are dark brown. **c–k** Dictyochlamydospores. Scale bars: **a–b**=10 mm, **c–k**=20 μm

form in culture (Fig. 11c-k). In molecular analysis (Fig. 1b) *C. huaikangplaensis* clusters with *Helicoma chlamydosporum* with strong support (100% PP).

Chlamydotubeufia khunkornensis Boonmee & K.D. Hyde, **sp. nov**. (Figs. 12 and 13)

MycoBank 563502

Acanthostigma perpusillum De Not. similis sed ascospores 34–42×5–7 et 6–7-septate et status anamorphosis dictyochlamydosporae (60–)73.5–130.5(−150)×(39–)48–65.5 (−70) μm differt.

Etymology: from *–ensis* meaning pertaining to and *Khun Korn,* the place of collection in reference to the collecting site.

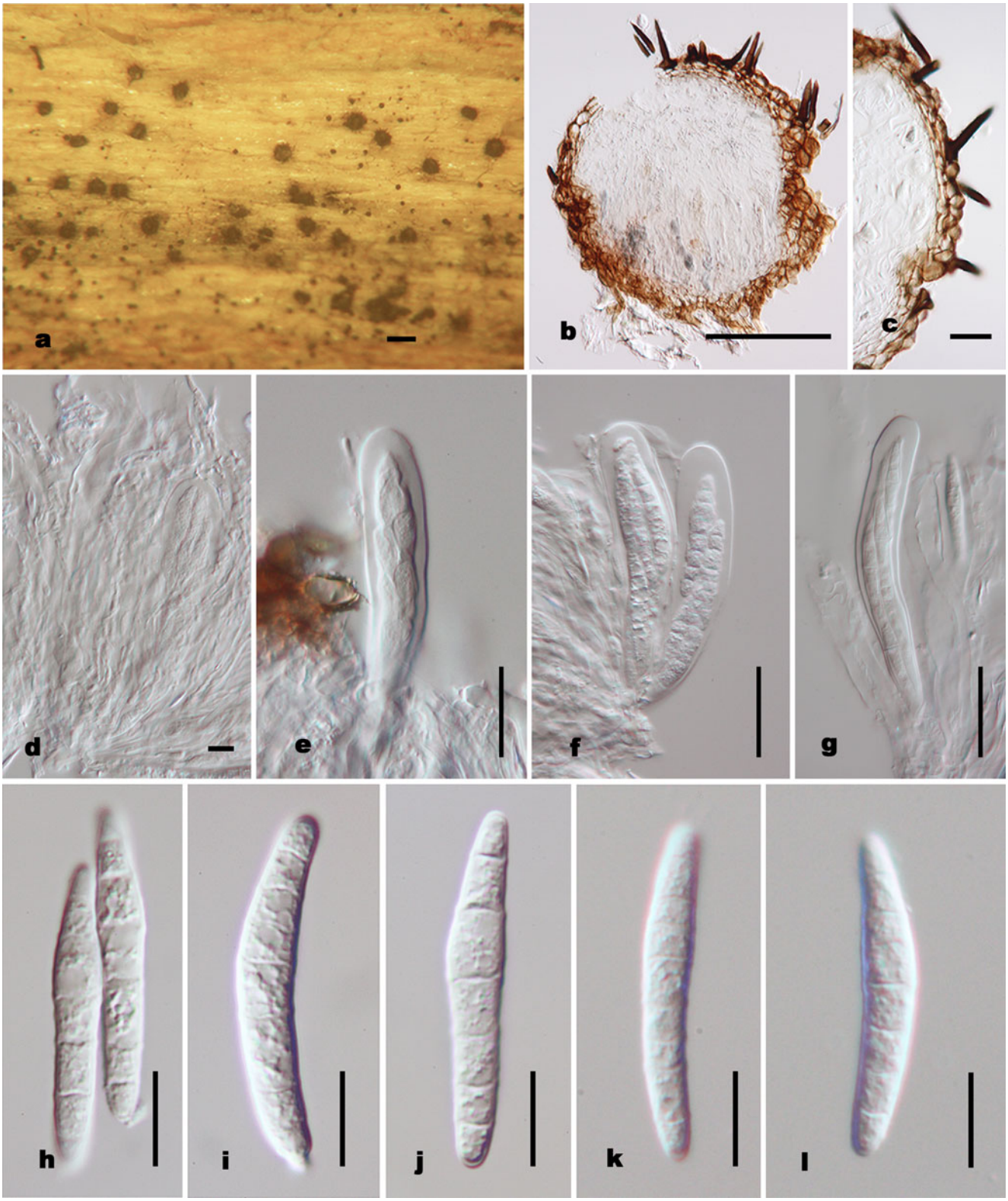

Fig. 12 *Chlamydotubeufia khunkornensis* (MFLU10–0051, holotype). **a** Superficial ascomata on substrate. Note ascomata covered with dark brown setae. **b, c** Section through ascoma and peridium that comprises brown-walled cells of *textura angularis*. **d** Pseudoparaphyses, cellular, embedded in a mucilaginous matrix. **e-g** Asci at young and mature stages. **h–l** Ascospores. Scale bars: **a**, **b**=100 μm, **c**, **e–g**=20 μm, **d**=5 μm, **h–l**=10 μm

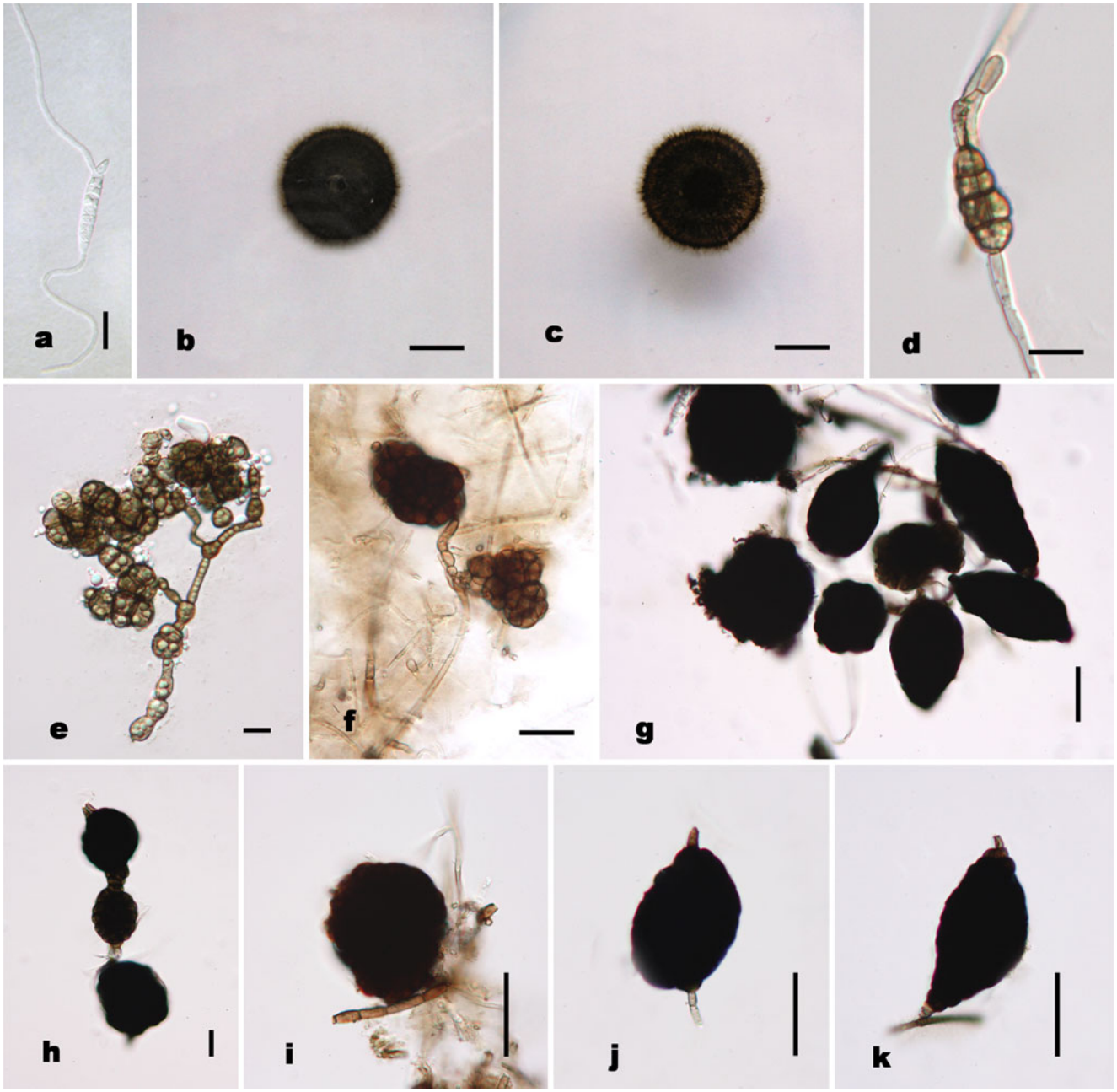

Fig. 13 *Chlamydotubeufia khunkornensis* (MFLUCC10–0118, holotype). Colonies on MEA. **a** Germinating ascospore. **b, c** Colonies on MEA from surface and in reverse. Note darkened colonies. **d–f** Mycelium development on culture. **g–k** Dictyochlamydospores. Scale bars: **a–c**=10 mm, **d–f**=10 μm, **g–k**=20 μm

Ascomata 180–250 μm high×199–249 μm diam ($\bar{x}$= 210.5×220μm, n=5), superficial, solitary or scattered, globose to subglobose, with a central ostiolate 20–30 μm wide, covered with dark brown setae, which taper to an acute apex (Fig. 12a, b). *Peridium* 18–28 μm thick, composed of 3–5 layers of light brown cells of compressed *textura angularis* (Fig. 12c). *Hamathecium* numerous, cellular, cylindrical, filiform pseudoparaphyses, hyaline, embedded in a gelatinous matrix (Fig. 12d). *Asci* 79.5–106×13–21 μm ($\bar{x}$= 96.5 × 15.5μm, n=20), 8-spored, bitunicate, cylindrical to clavate, apically rounded, ocular chamber not observed, short pedicellate (Fig. 12e-g). *Ascospores* 34–42×5–7 μm ($\bar{x}$ = 38 × 6μm, n=20), 2–3-seriate, fusiform, broad at the supra-median cell, 6–7-septate, ends asymmetrical, not constricted at any septum, hyaline, smooth walled (Fig. 12h-l).

Colonies in culture: Ascospores germinating on MEA within 24–36 h with germ tubes produced from both ends (Fig. 13a). Colonies growing on MEA slowly, reaching 5 mm in 1 month at 28°C, flat to slightly effuse, sparsely

hairy, with entire to fimbriate edge, darkened to blackish (Fig. 13b, c). Mycelium partly superficial, partly immersed, composed of branched, septate, smooth, subhyaline to olivaceous brown, dark brown hyphae, due to the development of dictyochlamydospores (Fig. 13d-f). Conidia (60–)73.5–130.5(−150) µm length, (39–)48–65.5(−70) µm wide ($\bar{x}$= 97.5 × 56.5μm, n=20), dictyochlamydospores, multicellular, darkening to black (Fig. 13g-k).

Material examined: THAILAND, Chiang Rai, Muang, Khun Korn Waterfall, elev. 671 msl., N19°51–54' E 99° 35.39', on dead wood of *Aleurites moluccana* (Candlenut tree), 13 November 2009, S. Boonmee KK-06 (MFLU10–0051, holotype), ex-type culture MFLUCC10–0118 = IFRDCC2186 = BCC.

The teleomorph state of *Chlamydotubeufia khunkornensis* is morphologically similar to *C. huaikangplaensis*, but differs by bright coloured ascomata, short-setae, and ascospores without guttulates (Figs. 10, 12). The dictyochlamydospores are also larger being 97.5×56.5 µm in *C. khunkornensis* and 56×35 µm in *C. huaikangplaensis.* The two species cluster together with low support (Fig. 1a).

Chlamydotubeufia chlamydospora (Shearer) Boonmee & K.D. Hyde, **comb nov.**

≡ *Helicoma chlamydosporum* Shearer, Mycologia 79: 468, 1987.

MycoBank 563503

Chlamydotubeufia depressispora (Matsush.) Boonmee & K.D. Hyde, **comb nov.**

≡ *Helicoma depressispora* Matsush., Matsush. Mycol. Mem. 7: 52 1993.

MycoBank 563504

Kamalomyces R.K. Verma, N. Sharma & Soni, Forest Fungi of Central India: 196 (2008).

Saprobic on dead bamboo. *Ascomata* forming on a subiculum of crowded black mycelium, superficial, clustered to solitary, globose to subglobose, stalked. *Peridium* comprising 3–4 layers of darkened cells of *textura angularis*, dull at margin. *Hamathecium*-cellular, filiform, branched pseudoparaphyses, embedded in a gelatinous matrix. *Asci* 8-spored, bitunicate, thick-walled, saccate-clavate, with an ocular chamber, pedicellate. *Ascospores* fusiform to clavate, slightly tapering toward the rounded ends, trans-septate with crowded septa, straight or slightly curved, upper part broad, hyaline.

Anamorphs reported for genus: none.

Type species:

Kamalomyces indicus R.K. Verma, N. Sharma & Soni, Forest Fungi of Central India: 196 (2008) (Fig. 14)

Saprobic on dead culms of bamboo (*Poaceae*). *Ascomata* (169–)216.5–253 µm high, (184–)225.5–295.5(−331) µm ($\bar{x}$= 218 × 263μm, n=5), forming on a subiculum of crowded black mycelium, superficial, clustered to solitary, subglobose-globose, stalked (Fig. 14a-d). *Peridium* (14.5–)19–28 µm, comprising of 3–4 layers of darkened cells of *textura angularis*, dull at margin (Fig. 14e). *Hamathecium* 1–1.5 µm wide, cellular, filiform, hyaline, branched pseudoparaphyses, embedded in a gelatinous matrix (Fig. 14f). *Asci* (140–)157–214(−223) µm×(27.5–)29–36(−38) µm ($\bar{x}$= 185 × 34μm, n=20), 8-spored, bitunicate, clavate, cylindric-clavate, subclavate or broadly obovoid, with an ocular chamber, pedicel (21–)25–49.5(−68.5) µm long (Fig. 14g-h). *Ascospores* (92–)95.5–104.5(−107)×8–10.5 µm ($\bar{x}$= 99 × 9μm, n=20), 3–4-seriate in the ascus, fusiform-clavate, slightly tapering towards the rounded ends, straight or slightly curved, upper part broad, (34–)38–42-trans-septate with crowded septa, constricted at the septum, hyaline (Fig. 14i-l).

Material examined: INDIA, Madhya Pradesh, Bori, on culms of *Dendrocalamus strictus* Nees (*Poaceae*), 5 April, 2004, R.K. Verma and K.K. Soni, No. TF0095 (TFRI, holotype).

This taxon appears to be typical of *Tubeufiaceae* as it has superficial, globose to subglobose, dark brown to black ascomata which form on a subiculum of crowded black mycelium, clavate asci and cylindrical-fusiform to elongate, trans-septate hyaline ascospores. No other genus in *Tubeufiaceae* have such a suite of characters and the crowded septa are particular distinct.

Podonectria Petch, Trans. Br. mycol. Soc. 7: 146 (1921)

Habit occurring in associations with scale insects. *Ascomata* superficial, scattered, solitary, subglobose-globose, light yellow-reddish, covered with light brown mycelium. *Peridium* composed of several-layers of pale yellowish brown cells of *textura angularis*. *Hamathecium* filiform, branched, septate, hyaline pseudoparaphyses embedded in a gelatinous matrix. *Asci* 8-spored, bitunicate, cylindro-clavate, short pedicellate. *Ascospores* 2–3-seriate, clavate to fusiform, hyaline to pale brown, trans-septate, wall minutely verruculose.

Anamorphs reported for genus: *Peziotrichum*, *Tetracrium* (Hyde et al. 2011)

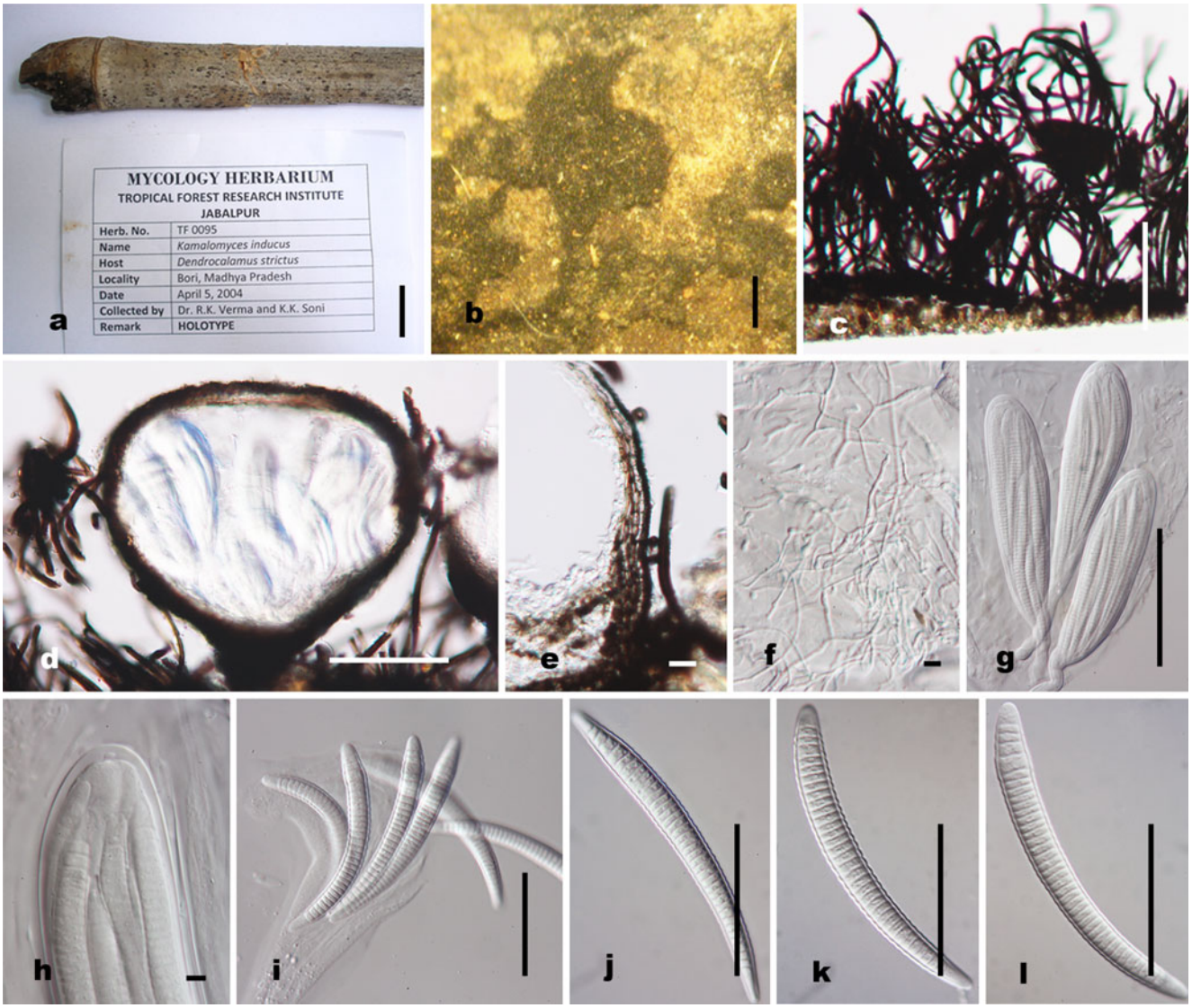

Fig. 14 *Kamalomyces indicus* (TFRI, holotype). **a** Appearance of fungus on bamboo. **b, c** Ascomata in subiculum on substrate. **d** Section of ascoma with stalk. **e** Peridium. **f** Pseudoparaphyses. **g–h** Asci showing pedicel (in g) and ocular chamber (in h). **i–l** Ascospores with crowded septa. Scale bars: **a–c**=150 μm, **d**, **g**=100 μm, **e**=10 μm, **f**, **h**=5 μm, **i–l**=50 μm

Lectotype species

Podonectria coccicola (Ellis & Everh.) Petch, Trans. Br. mycol. Soc. 7: 146 (1921) (Fig. 15)

≡ *Nectria coccicola* Ellis & Everh., J. Mycol. 2(4): 39 (1886)
≡ *Ophionectria coccicola* (Ellis & Everh.) Berl. & Voglino, in Saccardo, Syll. fung., Addit. I-IV (Abellini): 218 (1886)
≡ *Puttemansia coccicola* (Ellis & Everh.) Höhn., Sber. Akad. Wiss. Wien, Math.-naturw. Kl., Abt. 1 127: 625 (1918)
≡ *Scoleconectria coccicola* (Ellis & Everhart) Seaver, Mycologia 1(5): 198 (1909)
≡ *Tubeufia coccicola* (Ellis & Everh.) Lar.N. Vassiljeva, Nizshie Rasteniya, Griby i Mokhoobraznye Dalnego Vostoka Rossii, Griby. Tom 4. Pirenomitsety i Lokuloaskomitsety (Sankt-Peterburg): 317 (1998).

Habit occurring in association with scale insects. *Ascomata* (440.5–)476–496.5(−524.5) μm×357.5–458 (−599) μm ($\bar{x}$= 487 × 433μm, n=5), superficial, scattered, solitary, subglobose-globose, light yellow to reddish brown, brighter or reddish at the centre, covered with light brown mycelium (Fig. 15a-c). *Peridium* 51–69(−74) μm thick, composed several layers of pale yellowish brown cells of *textura angularis* (Fig. 15d). *Hamathecium ca.* 1 μm wide, cellular, filiform, branched, septate pseudoparaphyses embedded in a gelatinous matrix (Fig. 15e). *Asci* (133.5–)142–

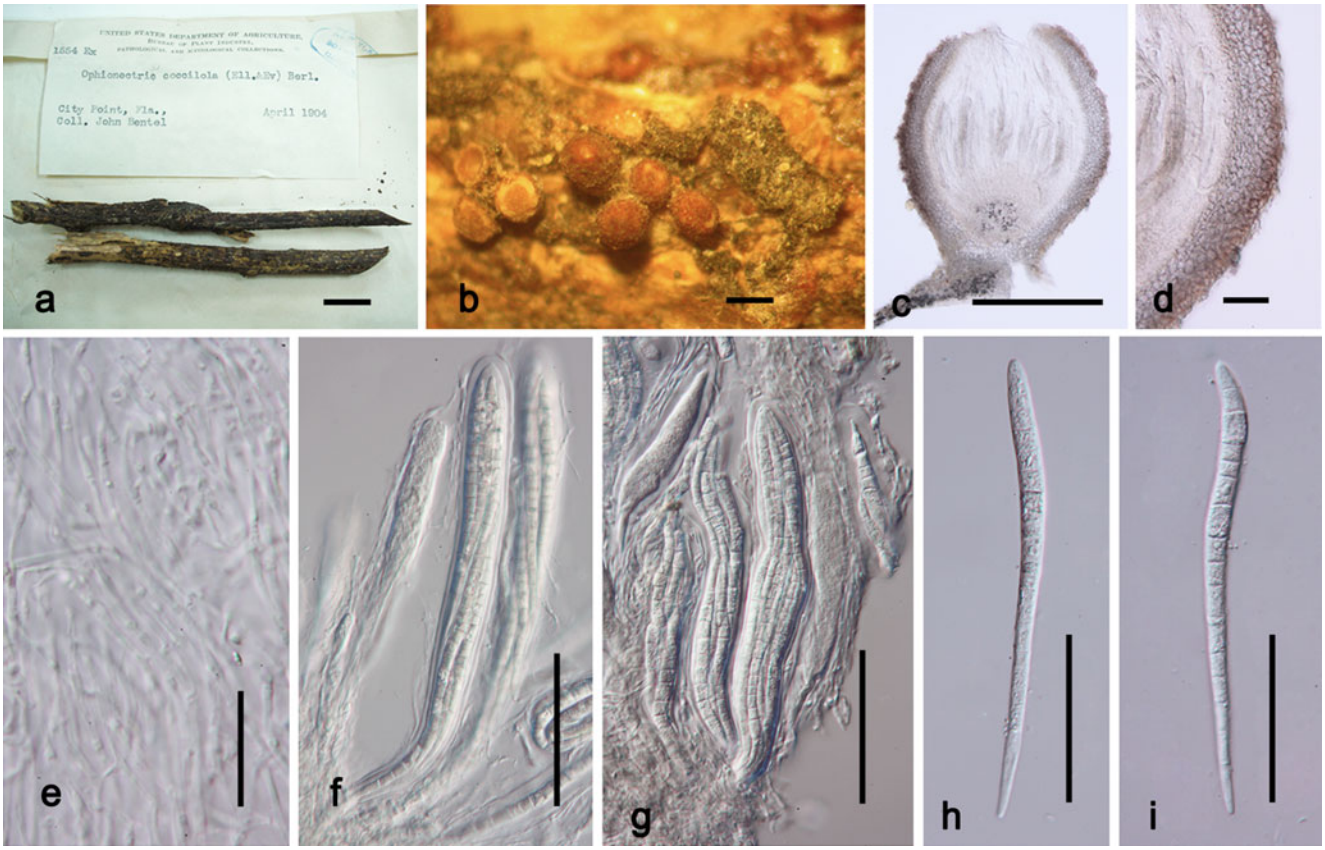

Fig. 15 *Podonectria coccicola* (NY, ex BPU 1554). **a** Material label. **b** Orange ascomata on substrate. **c** Section of ascoma. **d** Peridium. **e** Pseudoparaphyses. **f**, **f** Asci. **h**, **i** Ascospores with minutely verruculose walls. Scale bars: **a–c**=200 μm, **d**=40 μm, **e**=20 μm, **f–i**=50 μm

183(−202)×14–16(−21.5) μm ($\bar{x}$= 165 × 16.5μm, n=10), 8-spored, bitunicate, cylindric-clavate, ocular chamber not obvious, short pedicellate (Fig. 15f, g). *Ascospores* (101–)114–134.5(−139)×5.5–8.5(−10) μm ($\bar{x}$= 124 × 8μm, n= 10), 2–3-seriate, narrowly clavate to fusiform, (12–)14–18-trans-septate, ends rounded to sub acute, hyaline to pale grayish, wall minutely verruculose (Fig. 15h, i).

Material examined: USA, Florida, City Point, on scale insects on bark of orange trees, April 1904, John Bentel, ex BPU 1554 (NY); Gainesville, Florida, on scale insects on orange leaves, J. Matz, Herb. U. Mass. 632 (NY); Florida, Melbourne, on scale insects, 28 June 1937 J. Young (NY); Florida, Southworth, on dead scale insects on the bark of living orange trees, 24 January to 5 April 1923, Fred J. Seaver and Carlos E. Chardon No. 1064 (NY).

Podonectria is a genus that occurs on scale insects with long trans-septate ascospores and has been monographed by Rossman (1978) with eight accepted species. A collection determined as *P. coccicola* by Rossman (1978) was examined and is illustrated here. *Podonectria* was introduced by Petch (1921) for a taxon with light-reddish brown, hairy ascomata with clavate-fusiform, pale hyaline to grayish ascospores There are eleven species recorded in Index fungorum (www.indexfungorum.org/Names/Names.asp). *Podonectria coccicola*, the type species of the genus, was described from scale insects on orange trees from Florida (Rossman 1978). The habitat of the fungus and colour of the ascomata are unusual for *Tubeufiaceae* and this species needs recollecting to confirm its placement in the family.

Thaxteriella Petr., Annls mycol. 22(1/2): 63 (1924)

Saprobic on dead bark. *Mycelium* forming a dense velvety subiculum over the surface of the bark, comprising black hyphae. *Ascomata* superficially develop on the basal subiculum or slightly embedded, solitary or densely gregarious, globose to turbinate, black, shiny, often with small apical papillae at the central ostiolate apex. *Peridium* comprising two strata, outer layer of thick-walled, dark brown cells of *textura angularis* and inner layer of brown cells of *textura globosa*. The apical region is composed of less thick-walled cells which disintegrate to form an ostiole. *Hamathecium* cellular, filamentous, branched, anastomosing pseudoparaphyses attached to the hymenium and to the ascomata wall above. *Asci* 8-spored, bitunicate, cylindrical to broadly clavate or subclavate, thickened at the apex with

an ocular chamber, usually with a short pedicel. *Ascospores* cylindrical to long fusiform, tapering towards the rounded to sub-acute ends, trans-septate, hyaline.

Anamorphs reported for genus: *Helicoma* (Hyde et al. 2011)

Type species

Thaxteriella corticola Petr., Annls mycol. **22**(1/2): 63 (1924) (Fig. 16)

Saprobic on dead bark. *Mycelium* forming a dense velvety subiculum over the surface of the bark, comprising black hyphae. *Ascomata* 230–320×230–310 μm ($\bar{x}$ = 284× 296μm, *n*=10) diam, superficially developing on a basal subiculum or slightly embedded, solitary or densely aggregated, globose to turbinate, black, shiny, often with small apical papillae at the apex (Fig. 16a, b). *Ostiole* (10–)12.5–15 μm diam ($\bar{x}$= 13.5μm, *n*=5), central. *Peridium* 30–60 μm ($\bar{x}$= 39μm, *n*=10) diam, comprising two layers, outer layer of thick-walled, dark brown cells of *textura angularis* and inner layer of brown cells of *textura globosa* (Fig. 16c). The apical region is composed of less thick-walled cells which disintegrate to form an ostiole. *Hamathecium* 1–2 μm wide, cellular, filamentous, filiform, branched, anastomosing pseudoparaphyses attached to the hymenium and to the ascomata wall above (Fig. 16d). *Asci* (67.5–)77.5–101.5 (−112.5)×25–35 μm ($\bar{x}$= 92 × 28μm, *n*=10), 8-spored, bitunicate, cylindrical to broadly clavate or subclavate, thickened at the apex with an ocular chamber, usually with a short 5–10×5 μm ($\bar{x}$= 7.5 × 5μm, *n*=5) pedicel (Fig. 16e-g). *Ascospores* 45–55×(7.5–)10–12.5 μm ($\bar{x}$= 49.5 × 11μm, *n*=10), 3–6-seriate or fasciculate, cylindrical to long fusiform, tapering towards the rounded to sub acute ends, 7-septate, straight or slightly curved, sometimes constricted in some of the septa, hyaline to pale-yellowish (Fig. 16h-k).

Material examined: PUERTO RICO, Vega Baja., on dead bark, 15 January 1916, B. Fink No. 2145, F. Petrak, Pilzherbarium, Acqu. 1974 No. 24118 (W, holotype).

Thaxteriella was introduced by Petrak (1924) characterized by globose, black ascomata forming on a subiculum comprising smooth, thick-walled hyphae and bitunicate asci resembling *Herpotrichia* Fuckel (Bose 1961). Petrak (1953) proposed a new combination with *Thaxteriella pezizula* (Berk. & M.A. Curtis) Petr. and considered *Thaxteriella corticola* a synonym. Morphologically, *Thaxteriella* is similar to *Tubeufia* and has a similar anamorph (Samuels et al. 1979). *Thaxteriella* is typified by *T. corticola* Petr. (Petrak 1924) which Crane et al. (1998) considered to be a lost species, but is identical to *T. pezizula* (Berk. & M.A. Curtis) Petr. We have been able to locate type material and therefore redescribe *T. corticola*. Crane et al. (1998) mentioned that *Thaxteriella* differs from *Tubeufia* in pigmentation and structure of the peridium and he transferred *T. helicoma* and *T. amazonensis* to *Thaxteriella* based on morphological characters.

Thaxteriella is treated as a distinct genus in *Tubeufiaceae* because ascomata are subglobose, shiny and become cupulate when dry, in having a distinct subiculum, and cylindrical to long fusiform ascospores which tapering towards the rounded to sub-acute ends, and a *Helicoma* anamorph. In the phylogenetic tree (Fig. 1b) *T. inthanonensis* forms a distinct cluster with *Helicoma dennisii*. *T. inthanonensis* is characteristic of *Thaxteriella* and anamorphs are typical of *Helicoma*. *Thaxteriella helicoma* however clusters with *Tubeufia paludosa*, but we cannot confirm the correct identity of this taxon from GenBank. The anamorph of *Thaxteriella* is reported as *Helicoma* (Sivanesan 1984, Hyde et al. 2011), which is also polyphyletic.

Thaxteriella inthanonensis Boonmee & K.D. Hyde, **sp. nov**. (Figs. 17 and 18)

MycoBank 563505

Thaxteriella corticola Petr. similis sed ascospores (29.5–) 31.5–36.5(−37.5)×(5.5–)7–8 μm, 7-septate, anamorphosis helicosporae (10–)13–20×4–7 μm.

Etymology: from the Latin *–ensis* referring to an association and *Inthanon* the place of collection.

Saprobic on dead bark. *Ascomata* 258.5–307(−322) μm diam×(216–)227–247 μm high ($\bar{x}$= 232 × 293.5μm, *n*=5), superficial, solitary or scattered, globose to subglobose, subiculum or turbinate, collapsing when mature or dry, dark brown, shiny, without setae, central ostiolate (Fig. 17a, b). *Peridium* (42–)61–70 μm thick, comprising several layers of thickened cells of *textura angularis* to *globularis*; inner wall having subhyaline cell layers, become brown to dark brown inwardly (Fig. 17c). *Hamathecium ca*. 2 μm wide, hyaline, filiform, septate, anastomosing pseudoparaphyses embedded in a gelatinous matrix (Fig. 17d). *Asci* (73–)80–91×(21–)24–30(−32.5) μm ($\bar{x}$= 83.5 × 28μm, *n*=10), 8-spored, bitunicate, clavate-broadly, rounded and thickened at the apex, without a pedicel, 2–3-seriate in the ascus (Fig. 17e-g). *Ascospores* (29.5–)31.5–36.5(−37.5)×(5.5–)7–8 μm ($\bar{x}$= 33 × 7μm, *n*=20) allantoid, cylindrical to long fusiform, tapering towards the ends, slightly curved, 7-septate, slightly constricted at septa, symmetrical, granular when immature, hyaline to pale, smooth-walled at maturity (Fig. 17h-k).

Colony in culture: *Ascospores* germinating on MEA within 12–24 h and germ tubes produced from both ends

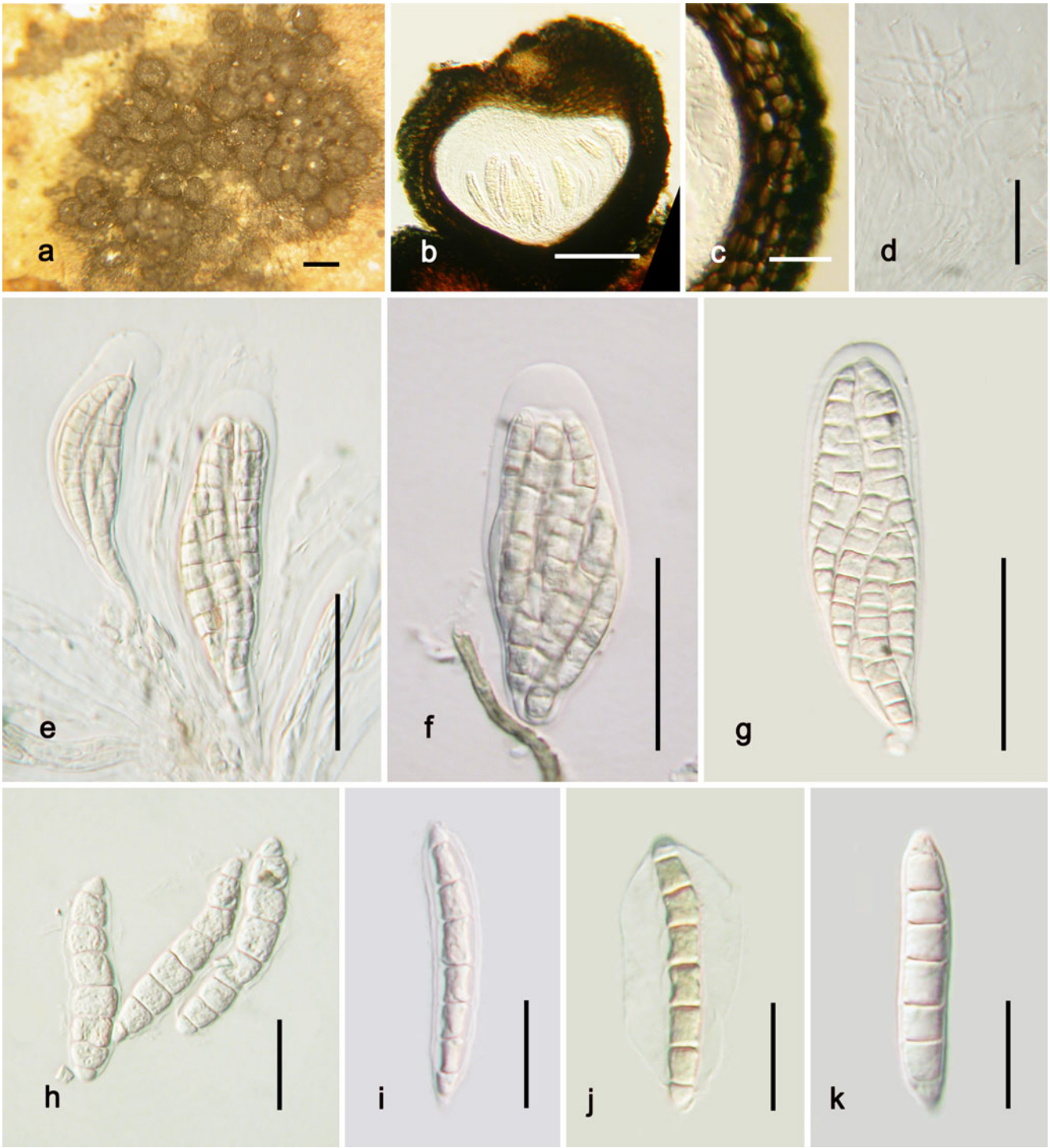

Fig. 16 *Thaxteriella corticola* (W, holotype). **a** Superficial gregarious ascomata on bark. **b** Section through ascomata. **c** Peridium. **d** Pseudoparaphyses. **e–g** Asci. Note the ocular chamber and short pedicels. **h–k** Ascospores. Notes the sheath in j. Scale bars: **a**, **b**, **d**= 100 μm, **c**, **e–g**=40 μm, **d**=100 μm, **h–k**=20 μm

(Fig. 18a). *Colonies* growing on MEA slowly, reaching 8 mm in 1 week at 28°C, slightly effuse, edge entire to erose or dentate, darkened to blackish (Fig. 18b). Mycelium mostly superficial, branched, septate, smooth, subhyaline to pale brown (Fig. 18c, d). *Conidiophores* (14.5–)26.5–34 (−42) μm in diam, 3 μm wide, septate, smooth, pale brown (Fig. 18e, f). *Conidia* (10–)13–20 μm in diam, 4–7 μm wide ($\bar{x}$= 14 × 6μm), acropleurogenous, helicosporous, at first hyaline to pale yellowish brown, becoming brown, 7-septate, smooth or rough (Fig. 18g-k).

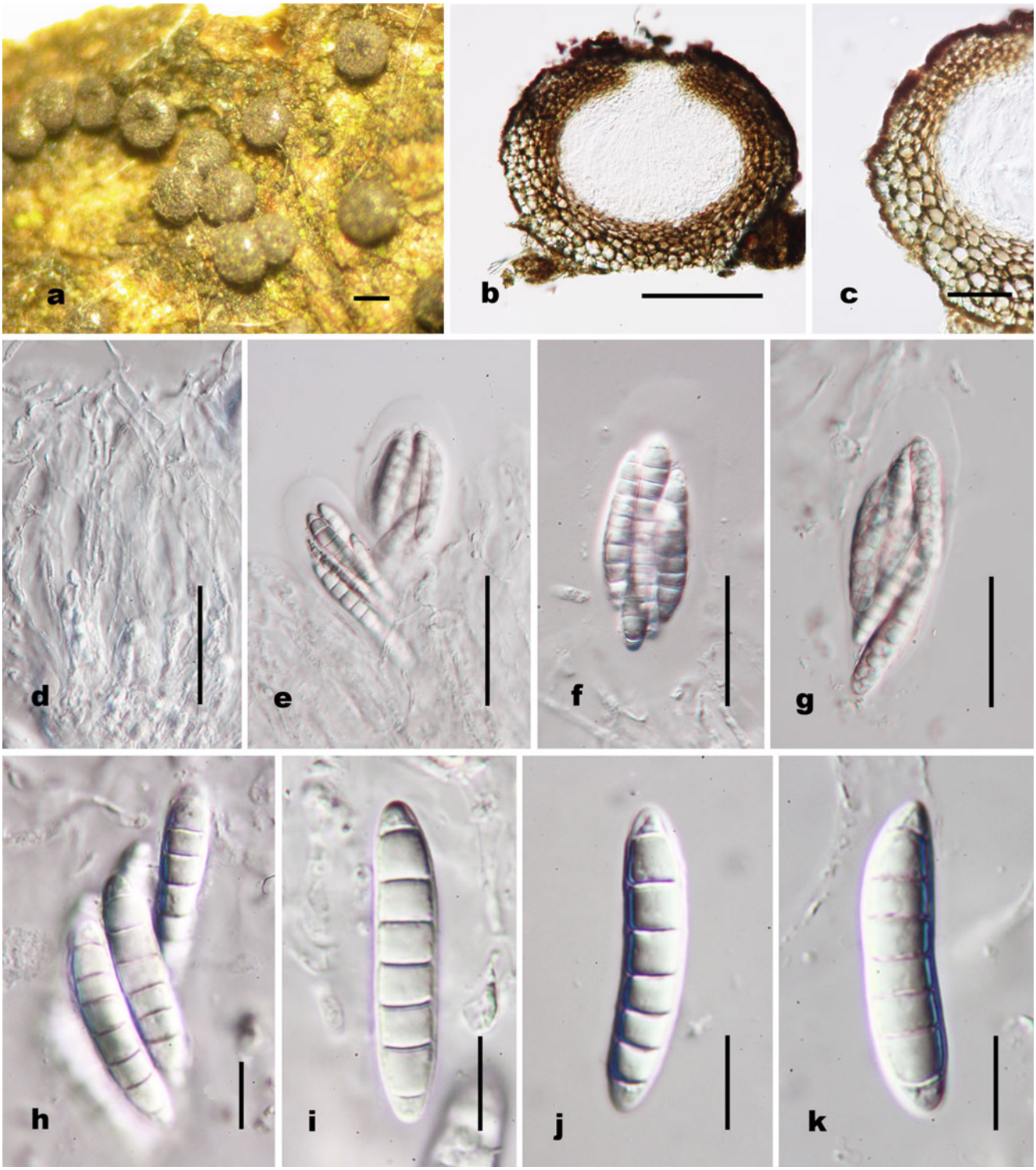

Fig. 17 *Thaxteriella inthanonensis* (MFLU11–0003, holotype). **a** Ascomata on substrate. **b** Ascoma cross section. **c** Peridium comprising cells of *textura angularis*. **d** Pseudoparaphyses. **e–g** Asci. **h–k** Ascospores. Scale bars: μm, **a**, **b**=100 μm, **c–g**=40 μm, **h–k**=10 μm

Material examined: THAILAND, Chinag Mai, Doi Inthanon, Jom Thong, elev. 800–1000 msl., N18°31.576' E 98°29.790', on dead bark of unidentified trees, 16 November 2010, Rungtiwa Phookamsak (MFLU11–0003, holotype), ex-type culture MFLUCC11–0003 = BCC.

Thaxteriella inthanonensis is very similar to *T. corticola* and obviously represents the same genus. *Thaxteriella inthanonensis* however, differs in the colour of the ascomata (Fig. 16a, 17a, Sivanesan 1984) being dark-brown in the new species and black in *T. corticola*. In *T.*

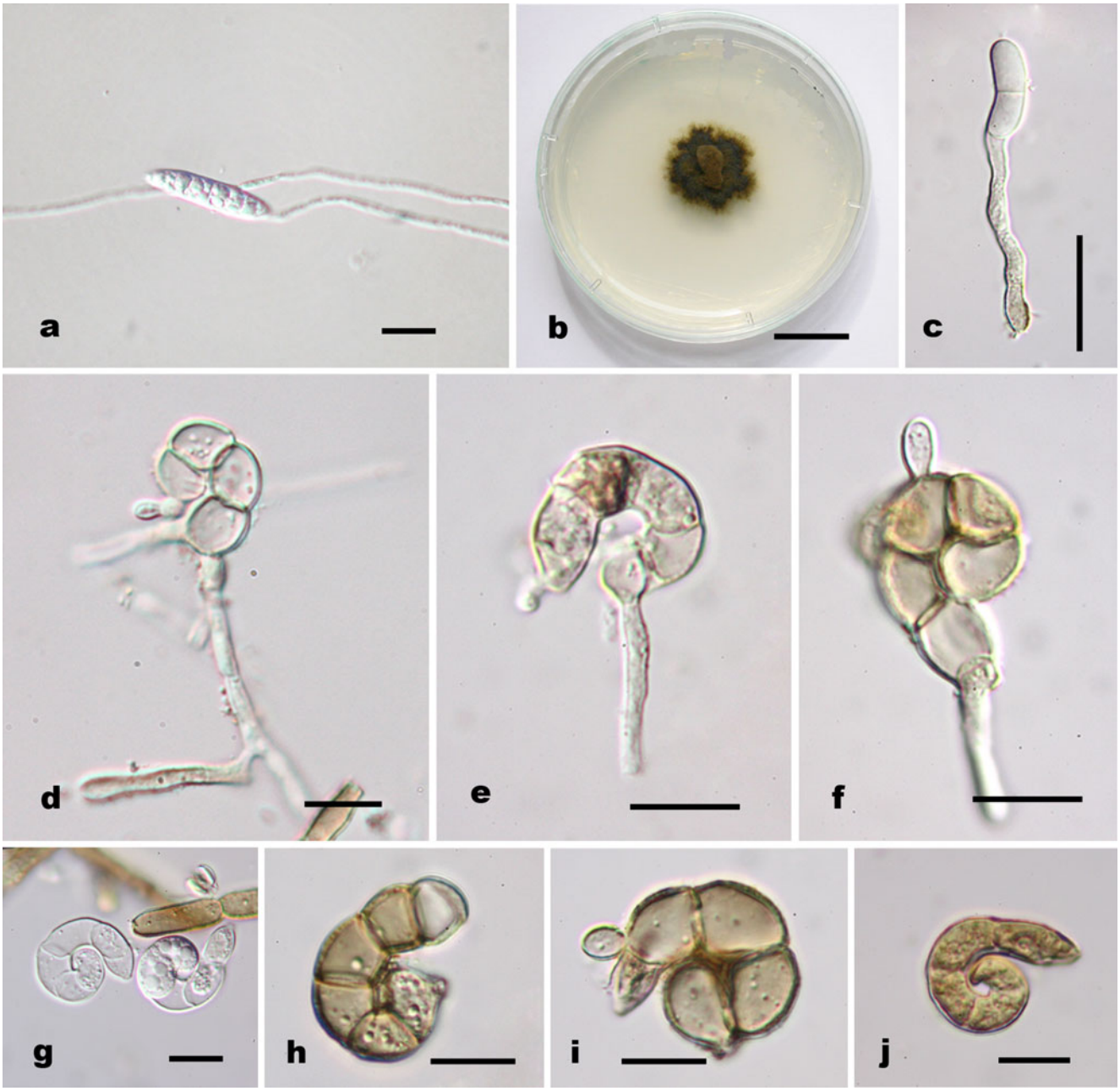

Fig. 18 *Thaxteriella inthanonensis* (MFLUCC11–0003, holotype). Colonies on MEA. **a** Germination of ascospore. **b** Colonies on MEA in surface view. Notes colonies are dark brown. **c–f** Conidiophores with conidia. **g–j** Conidia. Scale bars: **a, d–j**=10 μm, **b**=10 mm, **c**=20 μm

inthanonensis ascospores are also smaller than *T. corticola* (33×7 μm vs 49.5×11 μm). In addition *T. inthanonensis* produces pale brownish, septate *Helicoma*-like conidia (Fig. 18c-j), while in *T. corticola* no anamorphs are reported. *T. inthanonensis* clusters in an individual clade with 100% PP (Fig. 1b) and is representative of *Thaxteriella* which is a distinct genus from *Tubeufia*.

Thaxteriellopsis Sivan., Panwar & S.J. Kaur, Kavaka 4: 39 (1977) [1976]

Saprobic on dead wood. *Ascomata* born on a thin dark brown subiculum, superficial, solitary or scattered, globose to subglobose, reddish brown to dark brown, with brown to dark brown, septate, setae mostly on the top. *Peridium* comprising 3–4 layers of red brown to dark brown cells of *textura angularis*. *Hamathecium ca*. 2 μm wide, cellular, filiform pseudoparaphyses embedded in a gelatinous matrix. *Asci* 8-spored, bitunicate, cylindrical to clavate, apically rounded, short pedicellate, with long apical region with amorphous contents. *Ascospores* 2–3-seriate in an ascus,

fusiform to clavate, broader above, straight to slightly curved, 5-septate, constricted at the septum, hyaline, smooth-walled.

Anamorphs reported for the genus: *Helicosporium*, *Moorella*, *Xenosporium* (Hyde et al. 2011)

Type species

Thaxteriellopsis lignicola Sivan., Panwar & S.J. Kaur, Kavaka 4: 39 (1977) [1976] (Figs. 19, 21)

≡ *Chaetosphaerulina lignicola* (K.S. Panwar & S.J. Kaur), J.L. Crane, Shearer & M.E. Barr, *Can. J. Bot.* 76(4): 608 (1998).

Description from holotype (Fig. 19)

Saprobic on dead wood. *Ascomata* (142.5–)150–165(−179) μm high×(182–)204.5–248.5(−258.5) μm diam ($\bar{x}$= 157 × 217.5μm, *n*=5), borne on a thin dark brown subiculum, superficial, solitary or scattered, globose to subglobose, reddish brown to dark brown, with brown to dark brown, septate setae, mostly on the top (Fig. 19a-c). *Peridium* (35–)38–45 μm thick, comprising 3–4 layers of red brown to dark brown cells of *textura angularis* (Fig. 19d). *Hamathecium ca*. 2 μm wide, cellular, filiform pseudoparaphyses embedded in a gelatinous matrix (Fig. 19e). *Asci* (73.6–)76–92(−99)×(13–)15–18(−19) μm ($\bar{x}$= 84 × 16μm, *n*=20), 8-spored, bitunicate, cylindrical to clavate, apically rounded with long apical region with amorphous contents, short pedicellate (Fig. 19f). *Ascospores* (23–)24.5–27(−29)×6–8(−9) μm ($\bar{x}$= 25.5 × 7μm, *n*=20), 2–3-seriate, fusiform to clavate, broader above, straight to slightly curved, 5-septate, constricted at then septum, hyaline, smooth-walled (Fig. 19g, h).

Description from epitype (Fig. 20 and 21)

Ascomata 116–118(−148) μm high×130–132(−179.5) μm diam ($\bar{x}$= 127.5 × 147μm, *n*=3), superficial, solitary and scattered, globose to subglobose, shiny, reddish brown to dark brown, collapsed when dry, with central ostiole 17–19 μm wide, with dark brown setae up to 83.5 μm long with an acute apex (Fig. 20a-c). *Peridium* 17.5–28 μm thick, comprising up to 4-layers of dark brown cells of *textura angularis* (Fig. 20d). *Hamathecium ca*. 1.5 μm

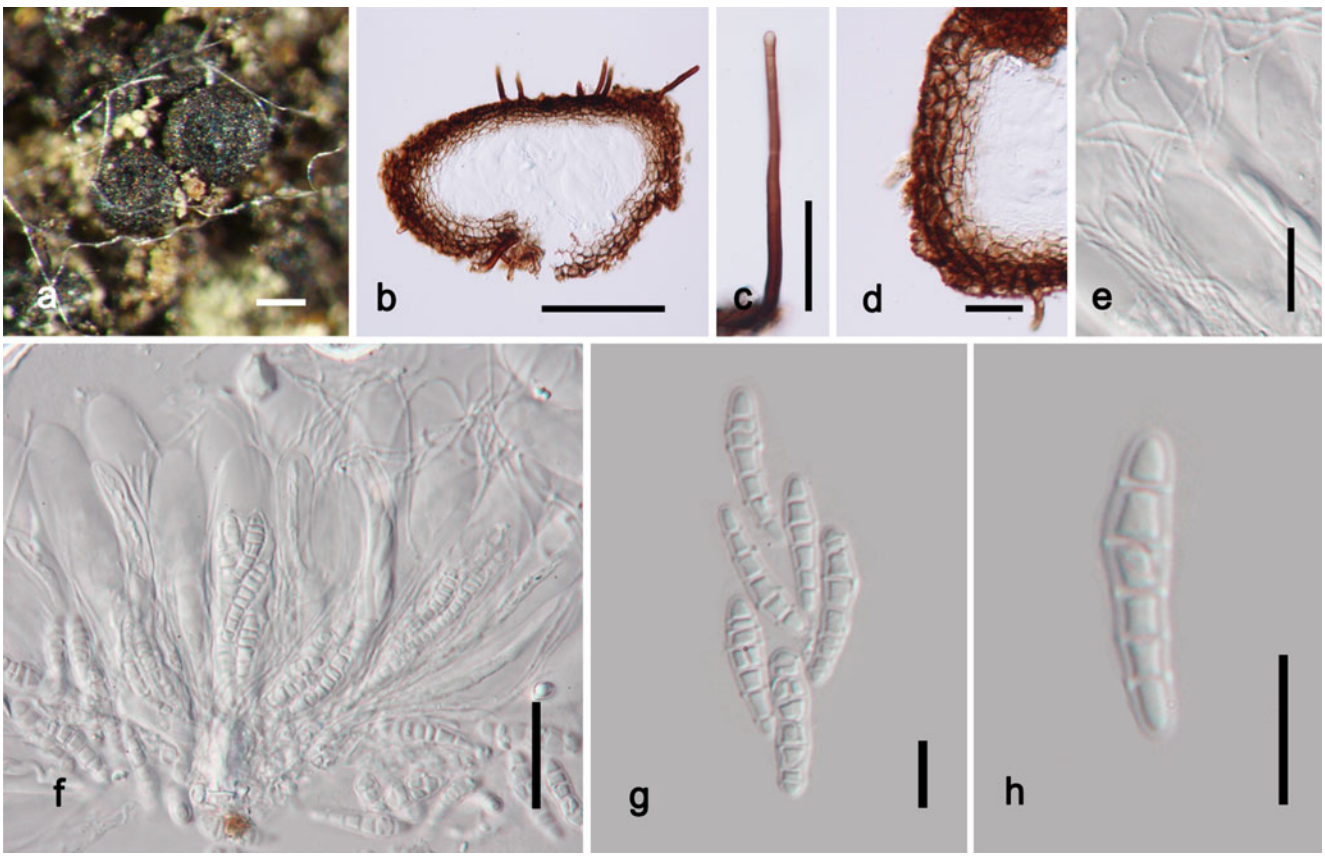

Fig. 19 *Thaxteriellopsis lignicola* (IMI, 197065: holotype). **a** Ascomata on substrate. **b** Section of ascoma. **c** Single seta. **d** Section of peridium. **e** Pseudoparaphyses. **f** Squash mount showing asci. Note the long apical region with amorphous contents. **g**, **h** Ascospores. Scale bars: **a**, **b**=100 μm, **c**=50 μm, **d**, **e**=20 μm, **f**, **g**=10 μm

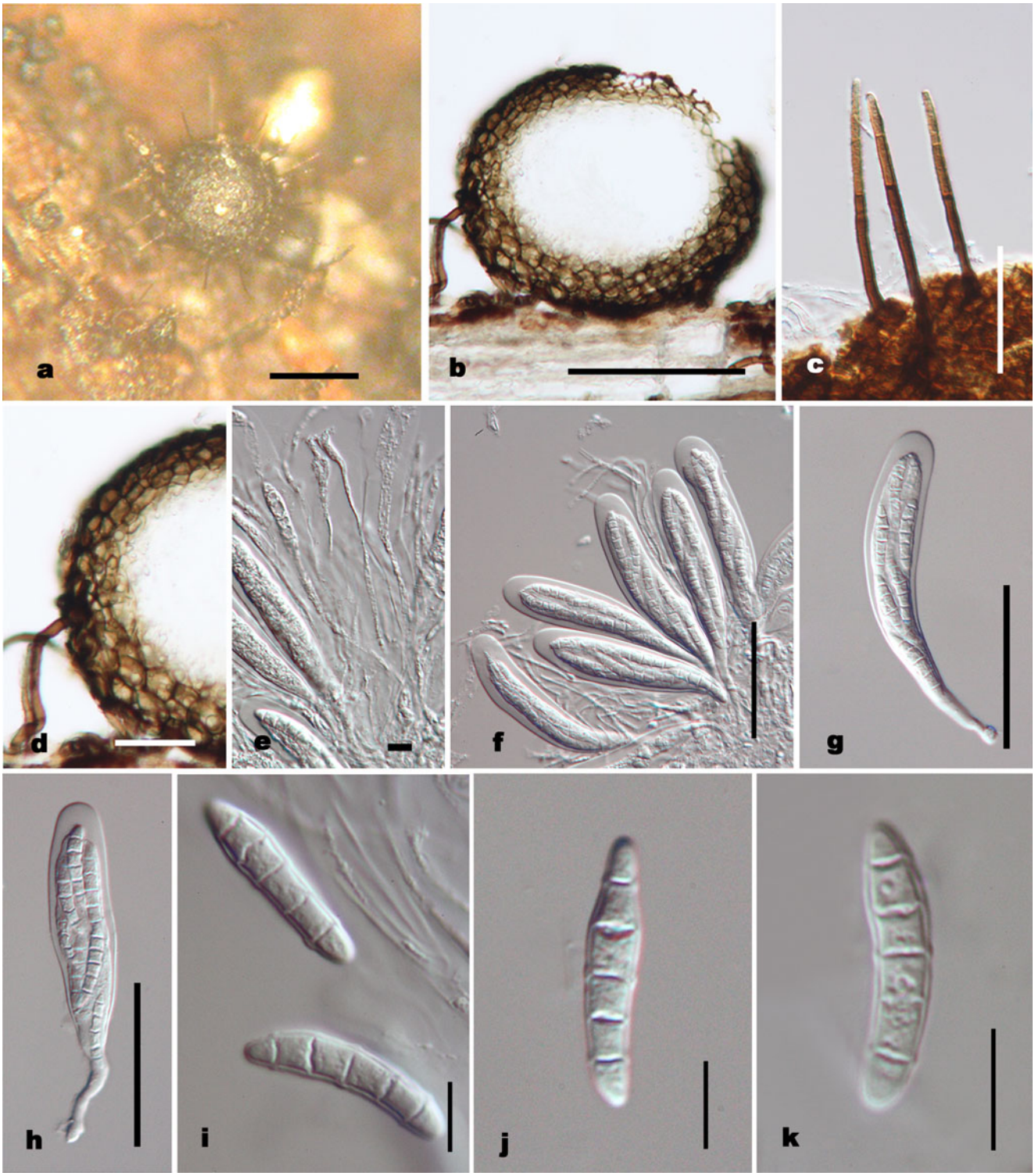

Fig. 20 *Thaxteriellopsis lignicola* (MFLU10–0057, epitype). **a** Superficial ascomata on substrate. Note ascomata covered with dark brown setae. **b** Section of ascoma. **c** Section of peridium. **d** Close up of brown setae, tapering to an acute apex. **e** Cellular pseudoparaphyses embedded in a gelatinous matrix. Pseudoparaphyses are longer than asci. **f–h** Asci at young and mature stages. Note the amorphous thickened apical region, low ocular chamber and long pedicel. **i–k** Ascospores. Scale bars: **a**, **b**=100 μm, **c**, **f–h**=50 μm, **d**=20 μm, **e**= 5 μm, **i–k**=10 μm

wide, cellular, filiform, hyaline pseudoparaphyses (Fig. 20e). *Asci* (78.5–)82–98(−106)×(14.5–)15–17(−18.5) μm ($\bar{x}$= 90 × 16μm, n=20), 8-spored, bitunicate, cylindrical to clavate, apically rounded, and amorphous thickened apical

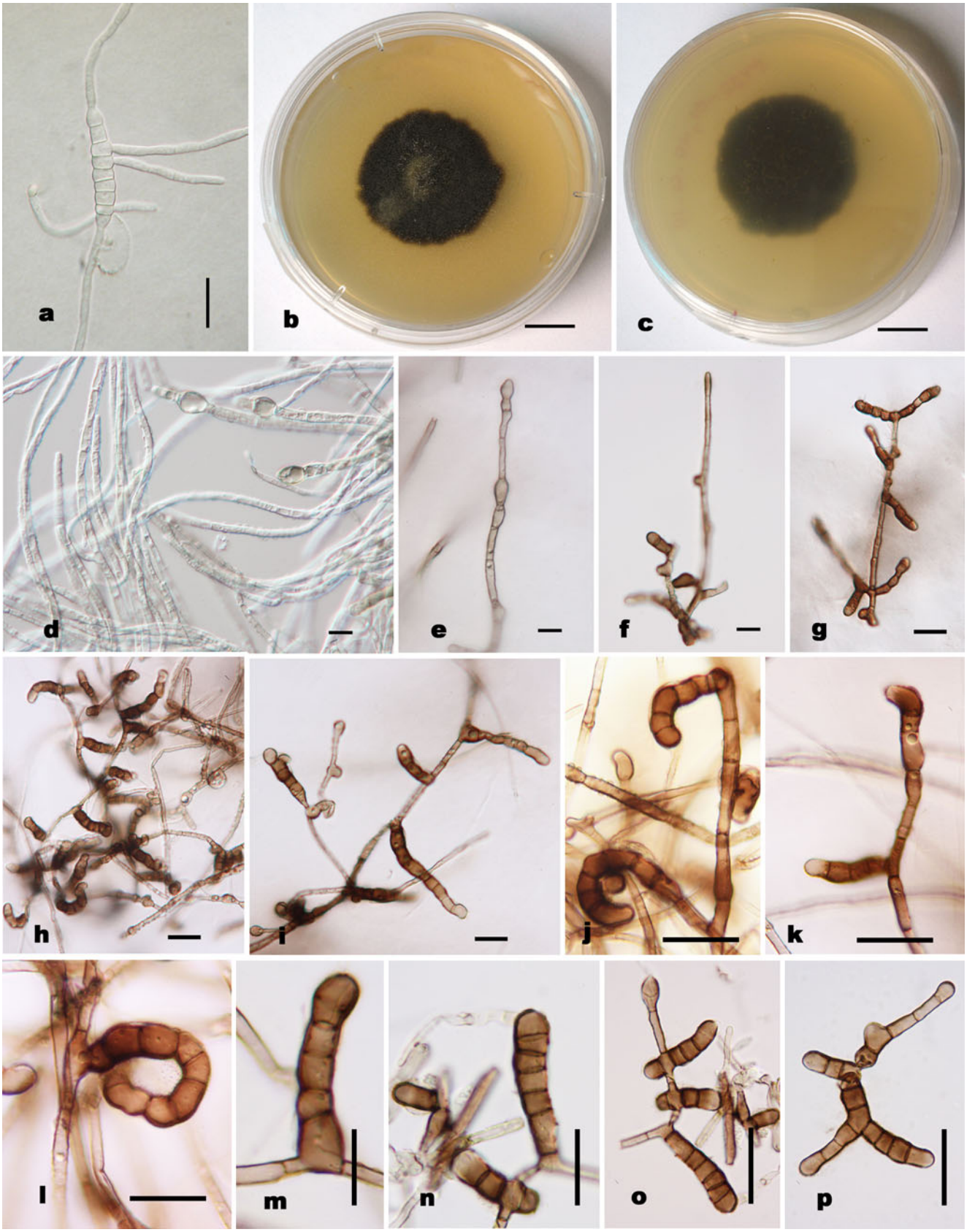

Fig. 21 *Thaxteriellopsis lignicola* (MFLUCC10–0124, epitype). Colonies on MEA. **a** Germinating ascospore. **b**, **c** Colonies on MEA from surface and reverse. Note colonies are dark brown. **d–g** Mycelium development in culture. **h–p** Conidia-like structures. Scale bars: **a**=20 μm, **b**, **c**=10 mm, **d–g**=5 μm, **h–p**=20 μm

region, ocular chamber low, with a 13–24 μm long pedicel (Fig. 20f-h). *Ascospores* (21.5–)23.4–28.5(−31)×5–6(−7) μm ($\bar{x}$= 26 × 5.5μm, *n*=20), 2–3-seriate in the ascus, clavate-fusiform, upper part broad, straight or slightly curved, ends rounded, 5-septate, slightly constricted at the septa, hyaline, smooth-walled (Fig. 20i-k).

Colonies in culture: Ascospores germinating on WA within 12 h and germ tubes produced from both ends (Fig. 21a). Colonies growing slowly on MEA, reaching 6 mm in 1 week at 28°C, effuse, edge entire to fimbriate, dark to blackish, aerial mycelium raised (Fig. 21b, c). Mycelium superficial, composed of branched, septate, smooth, subhyaline to pale yellow brown hyphae (Fig. 21d-f). Micronematous conidia-like structures (29–)40–80(−96) μm long, 5–7 μm wide, develop directly from hyphae (Fig. 21g-p).

Material examined: INDIA, Rajasthan, Mount Abu, on dead wood of *Lingo emortuo*, 10 September 1975, leg. K.S. Panwar 555 (IMI 197065, holotype); THAILAND, Chiang Mai Province, Mae Rim, Pong Yaeng, elev. *ca* 1000 msl., on dead wood of *Zizyphus mauritiana* Lamk (Jujube), 7 September 2009, S. Boonmee, PYJJ-01 (MFLU10–0057, epitype designated here), ex-epitype culture MFLUCC10–0124 = IFRD2197 = BCC; Lam Pang Province, Wang Neua, elev. *ca* 800–900 msl., on dead wood of unidentified trees, 7 January 2011, S. Boonmee, LP-01 (MFLU10–0055), living culture MFLUCC10–0122 = IFRD2175 = BCC; Chiang Rai Province, Muang, Khun Korn Waterfall, elev. 671 msl., N19°51–54' E 99°35.39', on dried bark of unidentified trees, 18 December 2009, S. Boonmee, KK-10 (MFLU10–0054), living culture MFLUCC10–0121 = IFRD2167 = BCC.

Thaxteriellopsis was introduced by Sivanesan et al. (1976) with *Thaxteriellopsis lignicola* as the type species and *T. eucalypti* A. Pande & V.G. Rao and *T. bambusicola* Sivan. & N.D. Sharma have since been added Crane et al. 1998). General characters as well as the ability to produce helicosporous anamorphs is are similarity to *Thaxteriella* (Subramanian and Sekar 1982, Sivanesan 1984, Fig. 18), but *Thaxteriellopsis* differs in having ascomata with long setae up to 180 μm and ascospores occasionally with longitudinal septate and becoming pale brown at maturity (Sivanesan et al. 1976). *Thaxteriellopsis bambusicola* has been transferred to *Chaetosphaerulina* by Crane et al. (1998) and this is probably correct, however we doubt that *Thaxteriellopsis lignicola* is a species of *Chaetosphaerulina*. The short ascospores, globose reddish-brown ascomata which collapse when dry and large thickened amorphous region at the ascus apex lead us to believe *Thaxteriellopsis* and *Chaetosphaerulina* are distinct genera. We did not find ascospores of *Thaxteriellopsis lignicola* with as many septa as reported by Sivanesan et al. (1976), but have clearly examined the same taxon. We did however find three new collections of *T. lignicola* which we consider to be the same taxon as the type and therefore epitypify the species here. *Thaxteriellopsis lignicola* forms a distinct clade in the phylogenetic tree with 99% PP (Fig. 1a) and thus is justified as a genus distinct from *Thaxteriella*. *Thaxteriellopsis lignicola* was associated with *Moorella*-like anamorphs (Subramanian and Sekar 1982).

Genera excluded from *Tubeufiaceae*

Allonecte Syd., Annls mycol. 37(4/5): 378 (1939)

Parasitic on living leaves. *Ascomata* superficial, subglobose, arranged in small clusters forming on a black, basal subiculum, ostiole central. *Peridium* composed of cells of *textura angularis*, outer layers dark brown to black, inner layers brown. *Hamathecium* filiform, branched, septate, hyaline pseudoparaphyses, mostly longer than asci. *Asci* 8-spored, bitunicate, cylindrical to fusiform, with a knob-like pedicel. *Ascospores* ellipsoid to broadly-fusiform, slightly obovoid, rounded ends, 1-septate, constricted at the septum, hyaline, thick-walled.

Anamorphs reported for genus: none.

Type species

Allonecte lagerheimii (Pat.) Syd., Annls mycol. 37(4/5): 379 (1939) (Fig. 22)

≡ *Broomella lagerheimii* Pat., Bull. Soc. mycol. Fr. 11(4): 229 (1895)

Parasitic on living leaves of *Poaceae* (bamboo). *Ascomata* 240–300 μm high×210–310 μm wide ($\bar{x}$= 266 × 254μm, *n*= 5), superficial, subglobose, arranged in small clusters on a black, basal subiculum ostiole central (Fig. 22a-c). *Peridium* 40–60 μm. wide, comprising cells of *textura angularis,* outer cells dark brown to black and inner cells brown (Fig. 22d). *Hamathecium* 1 μm wide, hyaline pseudoparaphyses, mostly longer than asci (Fig. 22e). *Asci* 125–177.5×7.5–12.5 μm. ($\bar{x}$= 150 × 12μm, *n*=10), 8-spored, bitunicate, fissitunicate, cylindrical, pedicellate, with an ocular chamber (Fig. 22f-h). *Ascospores* 17.5–20×7.5 μm ($\bar{x}$= 19.4 × 7.5μm, *n*=25), uniseriate, ellipsoid to broad fusiform, slightly obovoid, rounded ends, two-celled, constricted at the septum, hyaline, thick-walled (Fig. 22i-k).

Material examined: ECUADOR, on living leaves of *Poaceae* (bamboo), July 1892, Lagerheim San Jorge (FH, holotype).

This genus was erected by Sydow (1939) to accommodate *Allonecte lagerheimii*. It differs from *Tubeufiaceae*

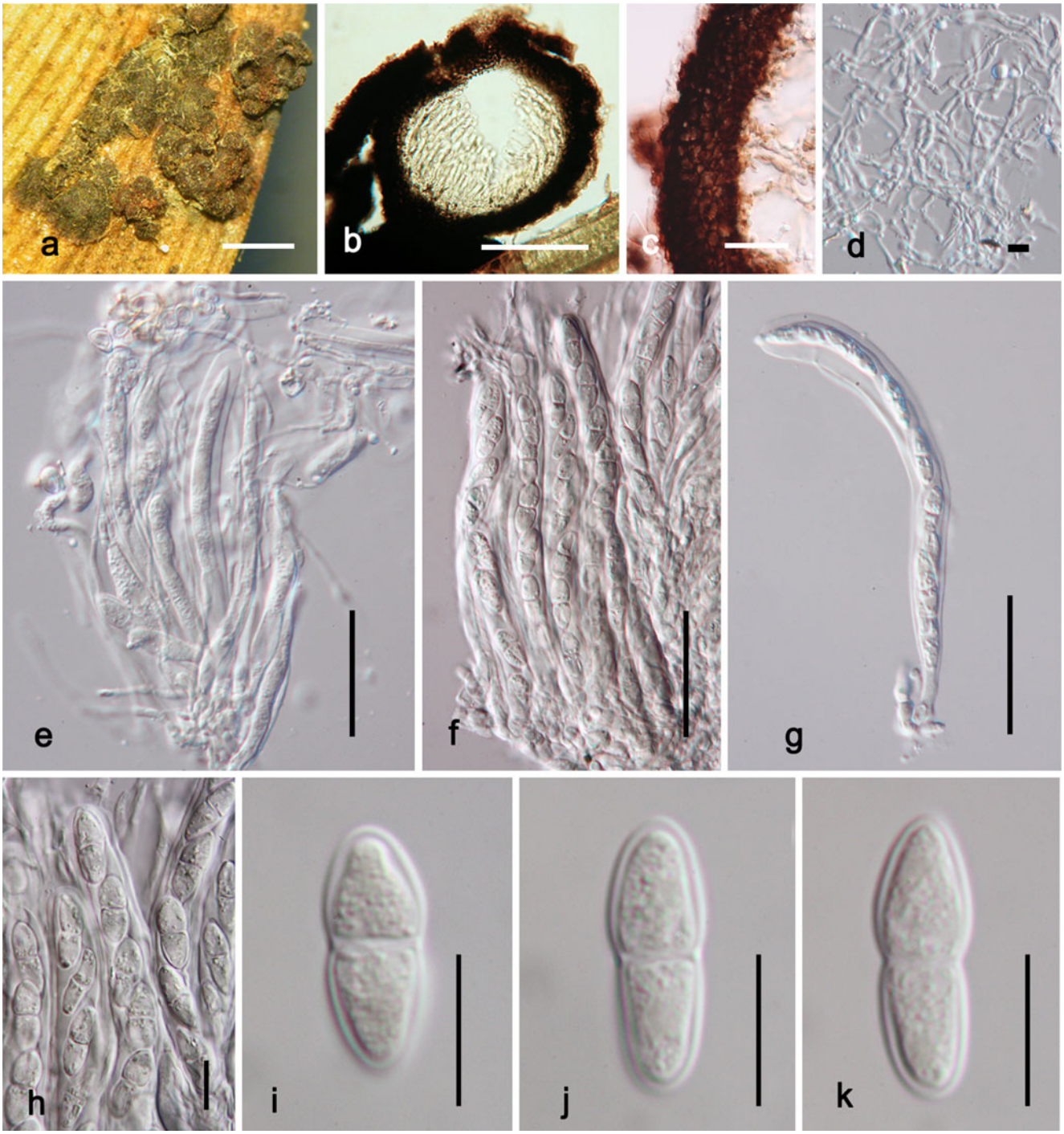

Fig. 22 *Allonecte lagerheimii* (FH, holotype of *Broomella lagerheimii*). **a** Ascomata on substrate. **b** Section of ascoma. **c** Peridium. **d** Pseudoparaphyses. **e–h** Asci with ocular chamber. **i–k** Ascospores. Scale bars: **a**, **b**=100 μm, **c**, **e–g**=40 μm, **d**=5 μm, **h–k**=10 μm

because it has ellipsoid, 2-celled ascospores with thick walls and asci are cylindrical and more typical of *Pleosporaceae* (Müller 1962, Rossman 1987, Crane et al. 1998, Fig. 22).

Byssocallis Syd., Annls mycol. 25(1/2): 14 (1927)

Parasitic on leaves. *Ascomata* superficial, scattered, solitary, subglobose-globose, light yellow-reddish, with a opening at the centre, covered with light brown mycelia. *Peridium* composed of pale yellow cells of *textura angularis*. *Hamathecium* filiform, branched, septate, hyaline pseudoparaphyses embedded in gelatinous matrix. *Asci* 8-spored, bitunicate,

cylindrical to clavate, thick-walled, slightly curved, short-stalked. *Ascospores* biseriate in the ascus, obovoid, broadly-narrowly, usually 3-trans-septate, occasionally with vertical septate, symmetrical ends, hyaline to grayish, smooth walled.

Anamorphs reported for genus: none.

Type species

Byssocallis phoebes Syd., Annls mycol. 25(1/2): 14 (1927) (Fig. 23)

≡ *Puttemansia phoebes* (Syd.) Petr., Annls mycol. 29(5/6): 343 (1931)

Parasitic on mycelium of *Meliolaceae* on leaves of *Phoebes jonduzii*. *Ascomata* (110–)166–200(−208.5) μm×163–281 (−306.5) μm ($\bar{x}$= 180 × 249μm, *n*=5), superficial, scattered, solitary, subglobose-globose, light yellow-reddish, with a opening at the centre, covered with light brown mycelia (Fig. 23a-c). *Peridium* 27.5–36 μm thick, composed 3–4 layers of pale yellow cells of *textura angularis* (Fig. 23d). *Hamathecium* 1–1.5 μm wide, hyaline, filiform, branched, septate pseudoparaphyses embedded in a gelatinous matrix (Fig. 23e). *Asci* (83–)96–121(−126) μm×18–22 (−24.5) μm ($\bar{x}$= 106 × 20μm, *n*=20), 8-spored, bitunicate, cylindrical to clavate, slightly curved, short pedicellate (Fig. 23f, g). *Ascospores* (35–)36.5–45(−48) μm×9–12.5 μm ($\bar{x}$= 42 × 10.5μm, *n*=20), fusiform-clavate, upper part widest, 3-trans-septate, occasionally vertically septate, slightly constricted, hyaline to grayish, smooth-walled (Fig. 23h-j).

Material examined: COSTA RICA, Grecia, on leaves of *Phoebes jonduzii* on *Meliolaceae*, 19 January 1925, H. Sydow 160a, ILL 8149 (S, BPI, isolectotype).

Byssocallis has yellow to orange ascomata and clavate ascospores, which is atypical of *Tubeufiaceae* (Barr 1980). Thus its taxonomic status needs to be reevaluated. We treat it as a member of Dothideomycetes *incertae cedis*.

Letendraeopsis K.F. Rodrigues & Samuels, Mycologia 86 (2): 255 (1994)

Endophytic in living leaves. *Ascomata* superficial, scattered to gregarious, globose to subglobose, without papilla or ostiole, colorless, becoming yellowish, smooth, glabrous. *Peridium* comprising a few layers of small, compressed cells. *Hamathecium* cellular, branched and anastomosing pseudoparaphyses, present at earliest stages of ascal development, attached apically and basally, persisting among mature asci arranged in a basal layer. *Asci* bitunicate, subglobose to obclavate at early stages, becoming clavate to broadly cylindrical, few in each ascoma, apex with an angular ocular chamber. *Ascospores* biseriate in ascus, fusiform, 1-septate, septum submedian, each cell with one to three globules, hyaline becoming yellow brown at maturity, with a broad hyaline sheath, smooth-walled.

Anamorphs reported for genus: none.

Type species

Letendraeopsis palmarum K.F. Rodrigues & Samuels, Mycologia 86(2): 255 (1994) (Fig. 24)

Ascomata (88–)113–227(−293) μm high, (100–)138–227 (−293) μm diam, superficial, scattered to gregarious, globose to subglobose, without papilla or ostiole, colourless, becoming yellowish, smooth, glabrous. *Peridium* less than 20 μm wide, comprised a few layers of small, compressed cells. *Hamathecium* of 2.5–3.5 μm wide, cellular, branched, anastomosing pseudoparaphyses, present at earliest stages of ascal development, persisting among mature asci, arranged in a basal layer (Fig. 24a). *Asci* 60–80×(19–)22–38(−40) μm, subglobose to obclavate at the early stages, becoming clavate to broadly cylindrical, few in each ascoma, with an angular ocular chamber (Fig. 24b, c). *Ascospores* (14–)19.2–26.4 (−27.5)×(7.2–)8.2–13.3(−14.7) μm, biseriate in ascus fusiform, 1-septate, septum submedian, each cell with one to three globules; at maturity, hyaline, becoming yellow brown, with a broad, hyaline sheath, smooth-walled (Fig. 24d-e).

Description revised from Rodrigues and Samuels (1994).

Material examined: BRAZIL, Para, Belém, Ilha Combu, on leaflets of *Euterpe oleracea*, September 1990, K.F. Rodrigues 427i3d, dry culture BPI (holotype).

The description follows Rodrigues and Samuels (1994). *Letendraeopsis* comprises globose to subglobose, colourless to yellowish ascomata, containing clavate to broadly cylindrical asci, with fusiform, hyaline to yellow brown, 1-septate ascospores (Fig. 24). These characters are atypical of *Tubeufiaceae* and *Letendraeopsis* are more typical of *Pleosporales* (*sensu* Zhang et al. 2012) where it is transferred.

Taphrophila Scheuer, Biblthca Mycol. 123: 171 (1988)

Saprobic on rotten plants. *Ascomata* superficial, solitary, globose to subglobose, with apically branched setae on the top, ostiole central. *Hamathecium* lacking. *Peridium* comprising 1–2 layers of brown cells of *textura angularis*. *Asci* 8-spored, bitunicate, broadly-clavate, slightly curved, short pedicellate. *Ascospores* multiseriate in ascus, elongate-fusiform, trans-septate, ends rounded, hyaline to grayish, wall roughened.

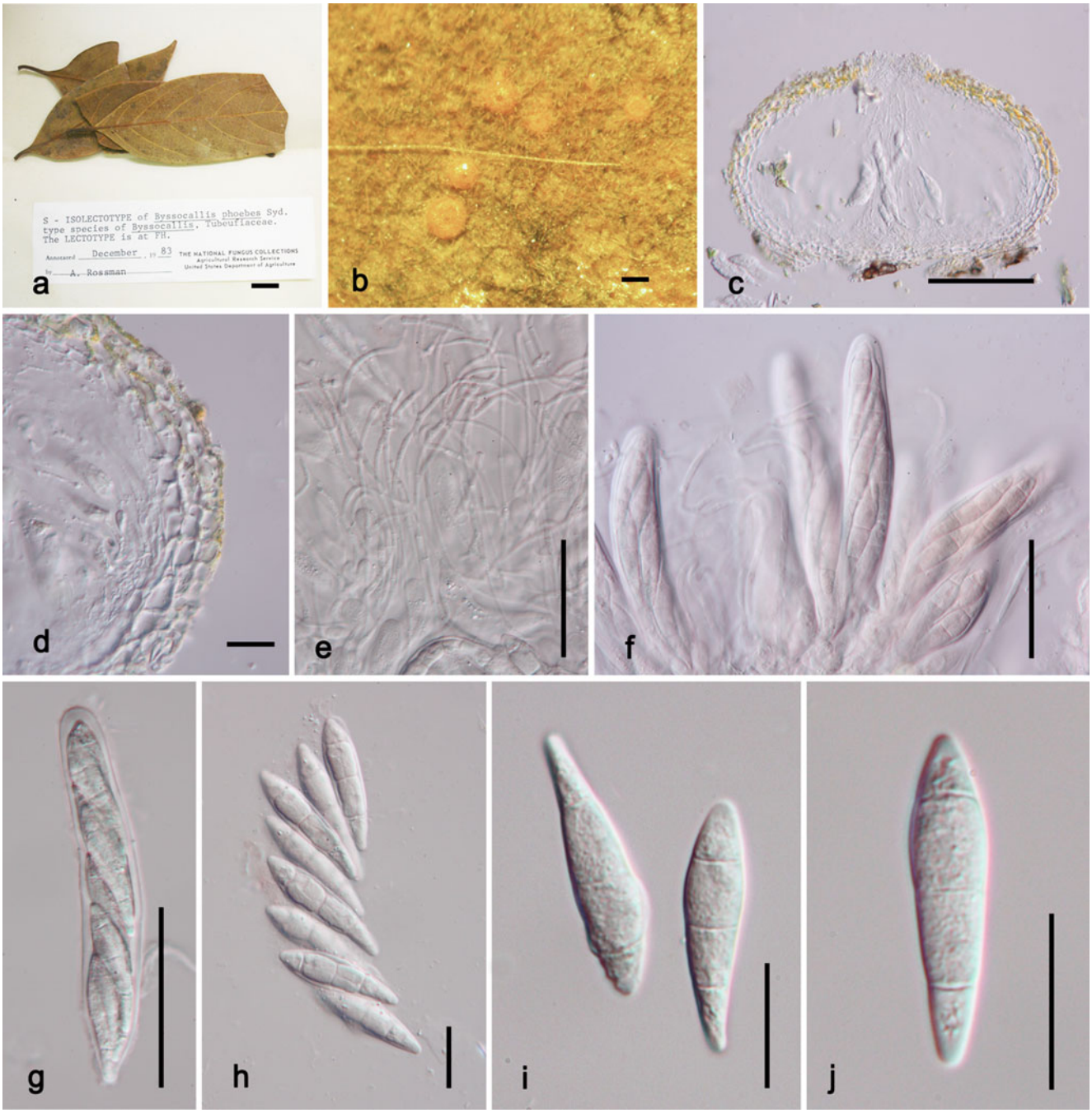

Fig. 23 *Byssocallis phoebes*. (S, isolectotype). **a** Type material. **b** Ascomata on substrate. **c** Section of ascoma. **d** Peridium. **e** Pseudoparaphyses. **f**, **g** Asci. **h–j** Ascospores. Scale bars: **a–c**=100 μm, **d**, **h–j**=20 μm, **e–g**=50 μm

Anamorph reported for genus: *Mirandina* G. Arnaud ex Matsush. (1975).

Type species

Taphrophila cornu-capreoli Scheuer, Biblthca Mycol. 123: 172 (1988) (Fig. 25)

Saprobic on rotten leaves or stems of *Carex paniculata*. *Ascomata* 27–33(−44) μm high×27–34(−47) μm diam, ($\bar{x}$= 33 × 34μm, n=5), globose to subglobose, superficial, solitary, branched setae on the top, ostiole central. *Hamathecium* lacking (Fig. 25a, b). *Peridium* 2–3 μm thick, composed of 1–2 layers of brown cells of *textura angularis* (Fig. 25c). *Asci* 19.5–24×7–9 μm ($\bar{x}$= 21.5 × 8μm, n=20),

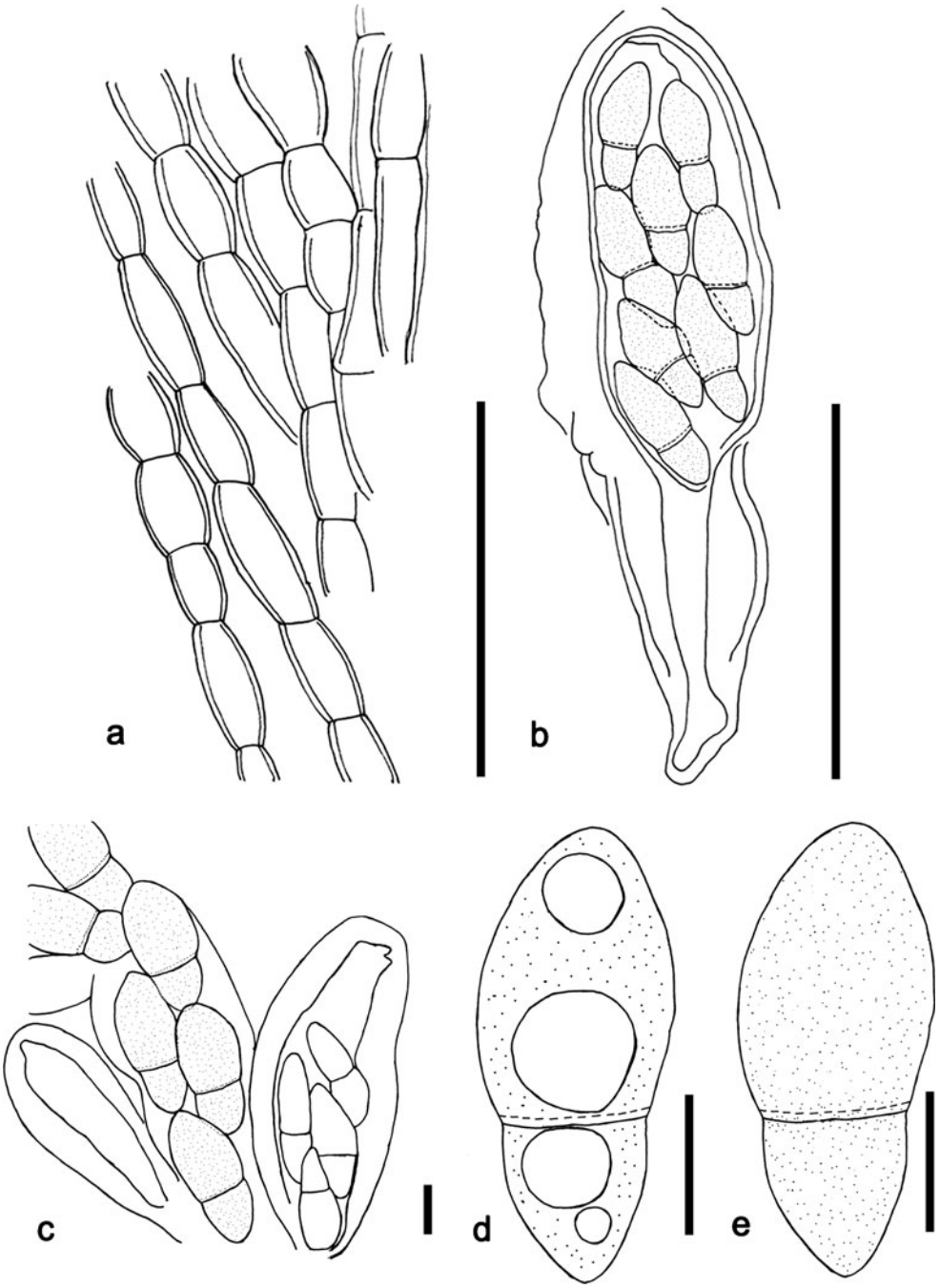

Fig. 24 *Letendraeopsis palmarum* (redrawn from Rodrigues and Samuels 1994). **a** Ascus with 8-spores. **b** Ascus with 8-spores. **c** Ascospores released. **d**, **e** Ascospores. Scale bars: **a**, **b**=40 μm, **c, e**=10 μm

8-spored, bitunicate, broadly-clavate, slightly curved, short pedicellate (Fig. 25d, e). *Ascospores* 13–18×1–2 ($\bar{x}$= 15 × 2μm, n=10), multi-seriate in ascus, narrowly fusiform, 5–6-septate, ends rounded, hyaline to grayish, wall roughened (Fig. 25f).

Material examined: AUSTRIA, Styria, Rottenmanner Tauern, Pöls valley, along the brook Pöls S of St. Johann am Tauern, elev. 1020 msl., 47°20'32" N, 14°28'17, grid ref. 8652/4; soggy brookside with Alnus incana, Filipendula ulmaria, Cirsium oleraceum, Scirpus sylvaticus, on rotten leaves and stems of *Carex paniculata*, 12 August 1984, Ch. Scheuer Nr. 1151 (GZU, holotype).

Taphrophila was introduced by Scheuer (1988) as a genus of *Tubeufiaceae* because of its ascomata having dark brown setae (Scheuer 1991, Fig. 24b). The very small ascomata, thin peridium, branching setae around the ascomata apex, clavate to saccate asci and lack of pseudoparaphyses are atypical of *Tubeufiaceae* and *Taphrophila* should be placed

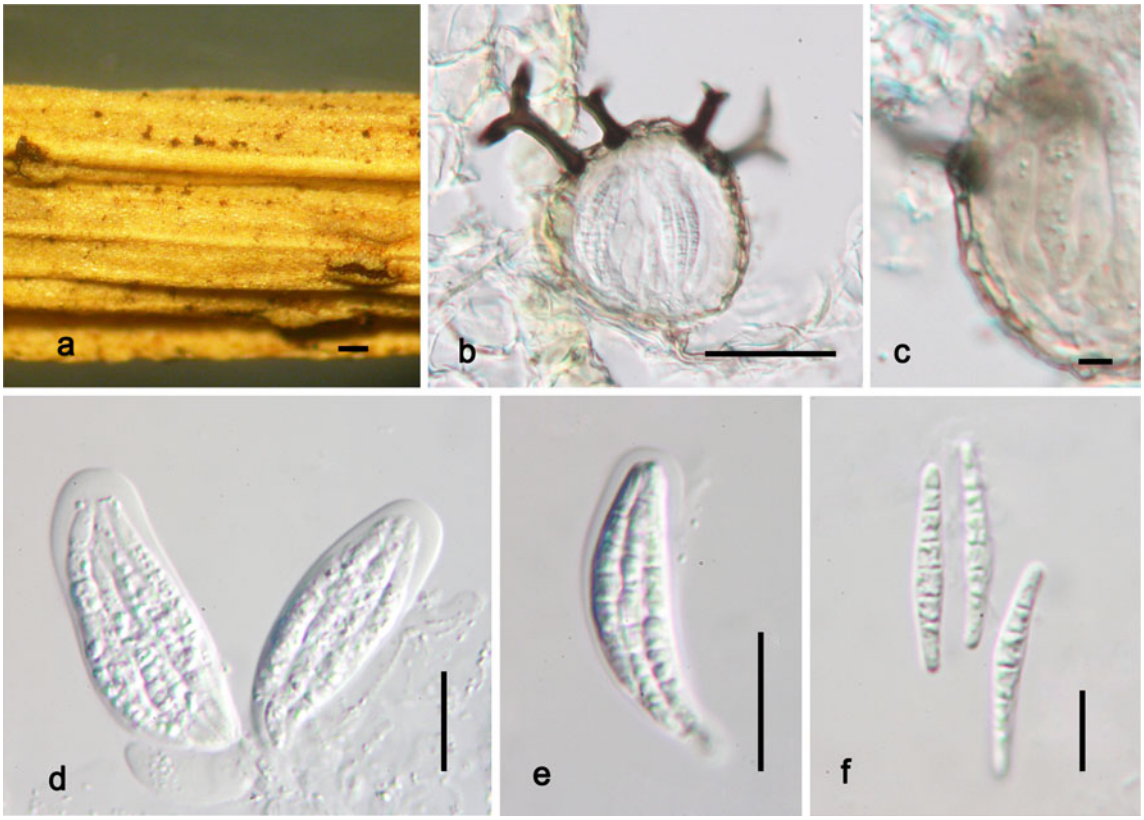

Fig. 25 *Taphrophila cornu-capreoli* (GZU, holotype). **a** Ascomata on substrate. **b** Section of ascoma illustrating the branched setae. **c** Thin-walled peridium. **d**, **e** Asci. **f** Ascospores. Scale bars: **a**, **b**, **d**, **e**=20 μm, **c**, **f**=5 μm

in Dothideomycetes *incertae cedis* until it can be sequenced (Scheuer 1991). The anamorph was reported as *Mirandina* by Matsushima (1975).

Moristroma A.I. Romero & Samuels, Sydowia 43: 246 (1991)

Thaxterina Sivan., R.C. Rajak & R.C. Gupta, Trans. Br. mycol. Soc. 90(4): 662 (1988)

Saprobic on dead wood. *Ascomata* forming on a subiculum, superficial, clustered, gregarious, globose to subglobose, red brown to dark brown, ostiole not observed, with setae, collapsed when dry. *Peridium* comprising several-layers of red brown to dark brown cells of *textura angularis*. *Hamathecium* hyaline, filiform, cellular pseudoparaphyses embedded in a gelatinous matrix. *Asci* multisporous, bitunicate, wide fusiform to saccate, rounded and thickened at the apex, with an ocular chamber. *Ascospores* numerous, oblong to elliptic-fusiform, ends rounded,1–3-septate, constricted at septum, thick-walled, hyaline, smooth-walled.

Anamorph reported for genus: none.

Type species: *Thaxterina multispora* Sivan., R.C. Rajak & R.C. Gupta

Moristroma multisporum (Sivan., R.C. Rajak & R.C. Gupta), Boonmee and K.D. Hyde, comb. nov. (Fig. 26)

≡ ***Thaxterina multispora*** Sivan., R.C. Rajak & R.C. Gupta, Trans. Br. mycol. Soc. 90(4): 662 (1988)

MycoBank 563506

Saprobic on dead wood of *Terminalia arjuna*. *Ascomata* (96–)128–196(−236) μm high×129–147(−292) μm diam ($\bar{x}$= 160 × 194μm, n=5), superficial, clustered, gregarious, globose to subglobose, forming on a subiculum, red brown to dark brown, collapsed when dry, ostiole not observed, with 39–145 μm long setae (Fig. 26a-c). *Peridium* (19.5–)27–38.5 μm thick, comprising 3–4 layers of red brown to dark brown cells of *textura angularis* (Fig. 26d). *Hamathecium ca*. 2 μm wide, cellular, filiform, hyaline pseudoparaphyses, embedded in a gelatinous matrix (Fig. 26e). *Asci* (102–)104–138.5(−144)×(28–)37–47(−48) μm ($\bar{x}$= 121 × 42μm, n=10), multisporous, bitunicate, wide fusiform to saccate, rounded and thickened at the apex, with an ocular chamber and short pedicel (Fig. 26f, g). *Ascospores* numerous, 7–10(−12)×2–3.5 μm ($\bar{x}$= 9 × 3μm, n=20), oblong to elliptic-fusiform, 1–3-septate, constricted at septum, thick-walled, hyaline, smooth-walled (Fig. 26h-k).

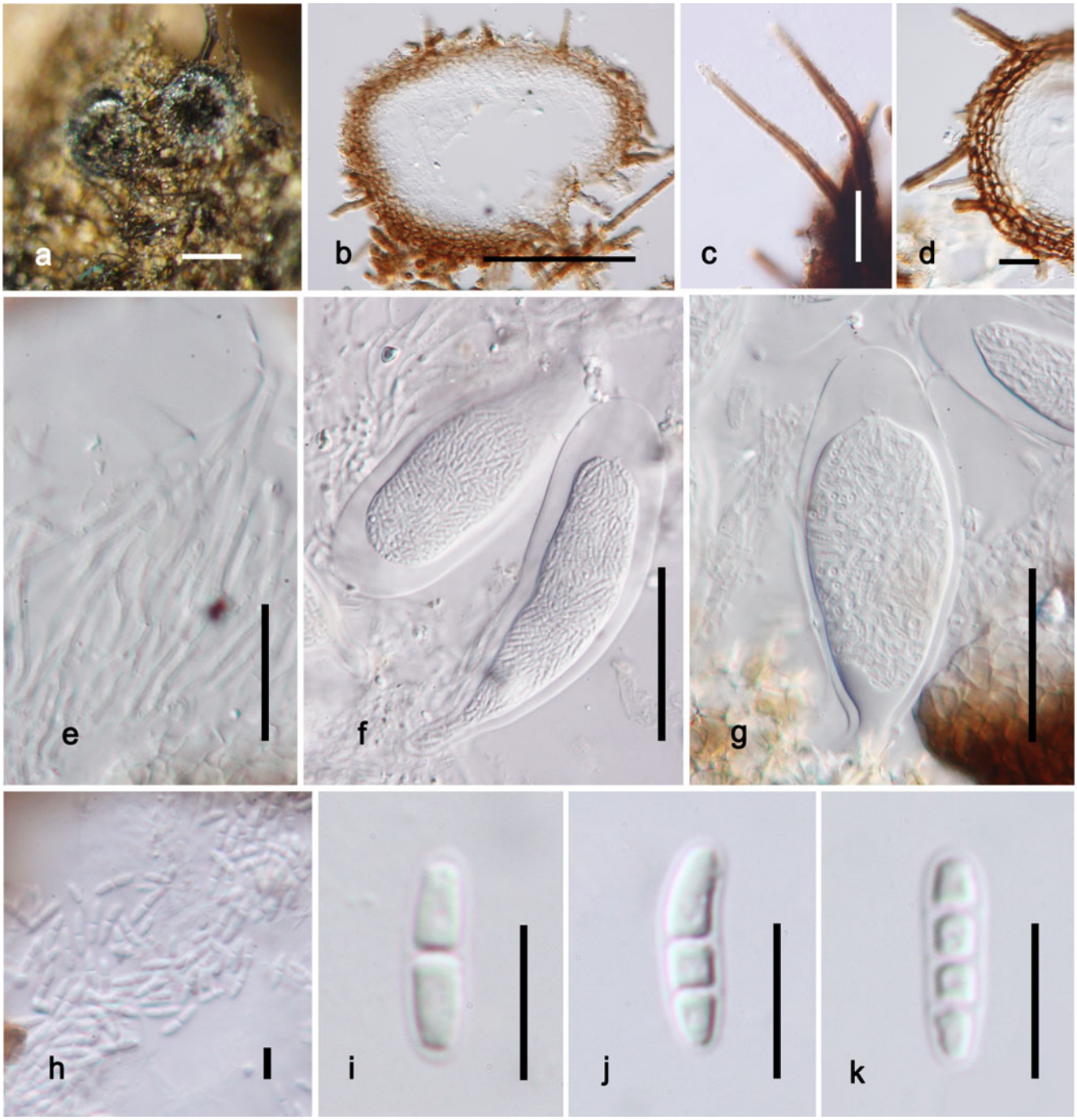

Fig. 26 *Thaxterina multispora* (IMI, holotype). **a** Ascomata on substrate. **b** Section of ascoma. **c** Setae. **d** Peridium. **e** Pseudoparaphyses. **f**, **g** Asci. **h–k** Ascospores. Scale bars: **a**, **b**=100 μm, **c**, **e**, **f**=50 μm, **d**=20 μm, **g–j**=5 μm

Material examined: INDIA, Madhya Pradesh, on dead wood of *Terminalia arjuna*, March 1987, RC Gupta 5 (IMI, holotype)

Sivanesan et al. (1988) introduced *Thaxterina* as the genus in *Tubeufiaceae* having ascomata characters similar to *Thaxteriella* and *Thaxteriellopsis*, but differing in being multisporous (Sivanesan et al. 1988). This taxon is very similar to *Moristroma polysporum* (Zhang et al. 2012) and thus we transfer *T. multispora* to this genus. Phylogenetic analysis of *Moristroma* by Nordén et al. (2005) showed it to be closely related to Chaetothyriomycetes instead of Dothideomycetes. The ordinal status of *Moristroma* has not been resolved. *Thaxterina* was also considered to be similar to *Moristroma polysporum* because it produced multispores in the ascus (Romero and Samuels 1991).

Discussion

There have been several molecular studies of *Tubeufiaceae* and its anamorphs (Kodsueb et al. 2004; Tsui et al. 2006, 2007; Promputtha and Miller 2010; Sánchez et al. 2011) but these studies have only partially resolved the generic concepts used within the family. This is mainly because these studies were unable to use strains of generic types or other species because they did not exist. They also used sequences downloaded from GenBank and it has not always been established if these taxa were correctly named. In other genera such as *Colletotrichum* the names in GenBank have been shown to be highly erroneous (Cai et al. 2009; Ko Ko et al. 2011). Our study represents an effort to resolve this problem, at least in part, by epitypifying genera and species and providing sequences of species very similar to the generic types.

The study makes some progress towards establishing the generic concepts in the *Tubeufiaceae* and define five distinct genera based on morphology and molecular data and accept a further 14 genera based on morphology only. We also suspect that *Aquaphila* should be retained as a distinctly aquatic genus and consider *Tubeufia asiana* as a synonym of *Aquaphila* (*Aquaphila albicans* Goh, K.D. Hyde & W.H. Ho, Mycol. Res. 102(5): 588 (1998) ≡ *Tubeufia asiana* Sivichai & K.M. Tsui, Mycologia 99(6): 885 (2008) [2007]). We have also linked anamorphs to their teleomorph genera, although the anamorphic genera are still mostly unresolved. It is important that generic types are recollected and sequenced and designated as epitypes so that the generic boundaries can be further defined based on molecular data analysis as has been suggested by Hyde et al. (2010) and Vasilyeva and Stephenson (2010).

An alternative approach would be to clump all *Tubeufia*-like species illustrated above and anamorphs in a single large speciose genus (see Promputtha and Miller 2010; Sánchez et al. 2011). We however feel that there is enough morphological differences and molecular evidence to support the differentiation of genera and follow this option.

The anamorph genera of *Tubeufiaceae* are mostly helicosporous and most of these genera have been shown to be polyphyletic (see Tsui et al. 2006, 2007; Fig. 1a, b in this paper). In the present study we have shown that species with dictyochlamydospores form a distinct clade and, as a consequence, are placed in the new genus *Chlamydotubeufia*. This genus however, also produces helicosporous anamorphs (e.g. *Helicoma chlamydosporum*). Tsui et al. (2007) showed *Aquaphila albicans* to be an anamorph of a species of *Tubeufia* that they described as *T. asiana*, however our studies would suggest this might represent a unique genus. On the other hand, *Helicoma*, *Helicosporium* and *Helicomyces* were shown to be highly polyphyletic by Tsui et al. (2006) and (Tsui et al. 2007), clustering throughout seven and nine clades, respectively, based on an analysis of ITS and partial LSU ribosomal sequences.

Several genera included in *Tubeufiaceae* by Lumbsch and Huhndorf (2010) were not studied because we have been unable to obtain a loan of type material during the time frame of this study and have not collected fresh material. Several of these genera were discussed by Barr (1980), Rossman (1987), Rossman et al. (1999) and Kodsueb et al. (2006), and only brief mention is made here. These taxa can be split into two groups, the first made up of those growing on dead plant material and the second consisting of those growing on other fungi.

Genera of *Tubeufiaceae* growing on dead plant material

Acanthostigmella (generic type = *A. genuflexa* Höhn.) was monographed by Barr (1977), and the ascomata are small, with setae around the ostioles, and develop on a delicate hyphal subiculum. Pseudoparaphyses are lacking and ascospores are 1-several septate. The genus has seven species (www.indexfungorum.org/Names/Names.asp). The lack of pseudoparaphyses would be unusual for *Tubeufiaceae* and Crane et al. (1998) suggested that this genus should be retained in *Herpotrichiellaceae* which is the concept followed herein. *Acanthostigmella brevispina* M.E. Barr & Rogerson clustered with members of *Pleosporales* in Untereiner et al. (1995); however, this species was transferred to *Tubeufia* by Crane et al. (1998), based on the fact it has pseudoparaphyses.

Amphinectria (generic type = *A. portoricensis* Speg.) consists of two species. The genus has been studied by various authors (Petrak 1951; Rossman 1987; Rossman et al. 1999) and is considered an ambiguous member of *Tubeufiaceae* as no good type material had been located. *Boerlagiomyces* is a well defined genus typified by *B. velutinus* (Penz. & Sacc.) Butzin and has been reviewed by Crane et al. (1998). There are several freshwater species (Cai et al. 2006). The genus is characterized by superficial, setose, membraneous ascomata with a peridium of large pseudoparenchymatous cells and hyaline to pale brown ascospores with numerous transverse septa and with vertical septa in most cells (Crane et al. 1998). One sequence in GenBank of *Boerlagiomyces websteri* Shearer & J.L. Crane does not cluster with *Tubeufiaceae* (Kodsueb et al. 2006) and this genus needs recollecting and sequencing to confirm if this result is correct.

Glaxoa (generic *G. pellucida* P.F. Cannon [Cannon 1997]) is also a monotypic genus, having been described from scales of dead cones of *Cupressus sempervirens* in the United Kingdom. We have examined the type material (IMI 362099) and it is in poor condition. The taxon has hyaline, rounded ascomata, and two-celled

hyaline ascospores. Cannon (1997) stated that this genus is most similar to *Letendraea* which Kodsueb et al. (2006) showed it to cluster in *Pleosporales* where *Glaxoa* should also be placed. However, fresh collections are needed so that the genus can be sequenced and analyzed. *Rebentischia* (generic type = *R. pomiformis* P. Karst.) has two species reviewed by Barr (1980) that appear to belong in *Tubeufiaceae* but are unusual in having a setiform basal appendage and a septate, pale brown upper part (Barr 1980).

Fungicolous genera

We have examined only the generic type of *Podonectria* associated with scale insects in this study, and there is a question as to whether or not this and other fungicolous genera belong in *Tubeufiaceae. Chaetocrea* (generic type *C. parasitica* Syd.) is a monotypic genus and is discussed and illustrated by Rossman et al. (1999) who placed it in *Tubeufiaceae*. The taxon grows on the stroma of other fungi. *Malacaria* (generic type = *M. meliolicola* Syd. was described by Sydow (1930) and is parasitic on *Meliolaceae.* There are eight species recorded in Index fungorum (www.indexfungorum.org/Names/Names.asp). *Malacaria* is unusual in *Tubeufiaceae* in having pale smoke-grey ascospores and unbranched septate pseudoparaphyses (Rossman, 1987). *Melioliphila* (generic type = *M. graminicola* (F. Stevens) Speg. was introduced by Spegazzini (1924) and is also parasitic on *Meliolaceae.* It differs from *Malacaria* in having hyaline, fusiform to clavate ascospores (Rossman, 1987). *Paranectriella* (generic type = *P .juruana* [Henn.] Höhn.) is also parasitic on other fungi and differs from other members of *Tubeufiaceae* by having a distinct papilla or cellular appendages at each end of the ascospore (Rossman 1987). *Puttemansia* (generic type = *P. lanosa* Henn.) also occurs on fungi and its ascospores are similar to those of *Paranectriella*. *Uredinophila* (generic type = *U. tropicalis* (Speg.) Rossman was introduced by Rossman (1987) to accommodate two species parasitic on rusts and has narrowly cylindrical to fusiform ascospores. Representative species from each of these fungicolous genera need recollecting, epitypifying and sequencing to establish if the genera are distinct and whether or not they belong in the *Tubeufiaceae*.

Acknowledgements The study reported herein was supported by Thailand Research Fund BRG528002. Appreciation is extended to the Global Research Net work for Fungal Biology and King Saud University for their support. The International Fungal Research and Development Centre, The Research Institute of Resource Insects, the Chinese Academy of Forestry (Bailongsi, Kunming 650224, China) and the Key Laboratory of Systematic Mycology and Lichenology, Institute of Microbiology, Chinese Academy of Sciences (Beijing, 100101, China) are also acknowledged for providing laboratory facilities. Thanks go to Cai Lei, Itthayakorn Promputtha, Rampai Kodsueb, Rungtiwa Phookamsak, Jian-Kui Liu and Kritsana Jatuwong for advice and discussions.

References

Barr ME (1977) Notes on some 'dimeriaceous' fungi. Mycotaxon 65:149–171

Barr ME (1979) A classification of Loculoascomycetes. Mycologia 71:935–957

Barr ME (1980) On the family Tubeufiaceae (Pleosporales). Mycotaxon 12:137–167

Berlese AN (1893) Descrizione di alcuni nuovi generi di pirenomiceti. Reprint from Atti del Congresso Botanico Internazionale: 1–10.

Berlese AN (1894) Icones fungorum omnium hucusque cognitorum. Biblioth Mycol 16A:1–243

Bose SK (1961) Studies on *Massarina* Sacc. and related genera. Phytopathologische Zeitschrift 41:151–213

Cai L, Hyde KD, Tsui CKM (2006) Genera of freshwater fungi. Fungal Diver Res Ser 18:1–261

Cai L, Hyde KD, Taylor PWJ, Weir BS, Waller J, Abang MM, Zhang JZ, Yang YL, Phoulivong S, Liu ZY, Prihastuti H, Shivas RG, McKenzie EHC, Johnston PR (2009) A polyphasic approach for studying *Colletotrichum*. Fungal Divers 39:183–204

Cannon PF (1997) Two new genera of Ascomycota, and other new or interesting fungi from Slapton Ley National Nature Reserve and its environs. Systema Ascomycetum 15:121–138

Crane JL, Shearer CA, Barr ME (1998) A revision of *Boerlagiomyces* with notes and a key to the saprobic genera of Tubeufiaceae. Canad J Bot 76:602–612

Hall TA (1999) BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. Nucleic Acids Symposium Series 41:95–98

Hyde KD, Chomnunti P, Crous P, Groenewald J, Damm U, Ko Ko TW, Shivas R, Summerell B, Tan Y (2010) A case for re-inventory of Australia's plant pathogens. Persoonia 25:50–60

Hyde KD, McKenzie EHC, KoKo TW (2011) Towards incorporating anamorphic fungi in a natural classification–checklist and notes for 2010. Mycosphere 2:1–88

Kirk PM, Cannon PF, David JC, Stalpers JA (2001) Ainsworth & Bisby`s dictionary of the fungi, 9th edn. CABI, Wallingford

Kirk PM, Cannon PF, Minter DW, Stalpers JA (2008) Ainsworth & Bisby`s dictionary of the fungi, 10th edn. CABI, Wallingford

Kodsueb R, Lumyong S, Lumyong P, McKenzie EHC, Ho WH, Hyde KD (2004) *Acanthostigma* and *Tubeufia* species, including *T. claspisphaeria* sp. nov., from submerged wood in Hong Kong. Mycologia 96:667–674

Kodsueb R, Jeewon R, Vijaykrishna D, Mckenzie EHC, Lumyong P, Lumyong S, Hyde KD (2006) Systematic revision of Tubeufiaceae based on morphological and molecular data. Fungal Divers 21:105–130

Ko Ko TW, Stephenson SL, Bahkali AH, Hyde KD (2011) From morphology to molecular biology: can we use sequence data to identify fungal endophytes? Fungal Divers 50:113–120

Larkin MA, Blackshields G, Brown NP, Chenna R, McGettigan PA, McWilliam H, Valentin F, Wallace IM, Wilm A, Lopez R, Thompson JD, Gibson TJ, Higgins DG (2007) Clustal W and Clustal X version 2.0. Bioinformatics 23:2947–2948

Lumbsch HT, Huhndorf SM (eds.) (2010) Outline of *Ascomycota* – 2009. Myconet 14:1–64

Matsushima T (1975) Icones microfungorum a Matsushima lectorum. Matsushima, Kobe, Japan 209:1–100

Nordén B, Sunhede S, Larsson E (2005) New species of *Moristroma* (Ascomycetes) and phylogenetic position of the genus. Mycol Prog 4:325–332

Nylander JAA (2004) MrModeltest 2.3. Program distributed by the author. Evolutionary Biology Centre, Uppsala University

Penzig O, Saccardo PA (1904) Icones fungorum javanicorum. E.J. Brill, Leiden

Petch T (1921) Studies in entomogenous fungi. 1. The nectriae parasitic on scale insects. Trans Brit Mycol Soc 7:89–167
Petrak F (1924) Mykologische Notizen VII. Ann Mycol 22:1–182
Petrak F (1951) Ergebnisse einer Revision der Grundtypen verschiedener Gattungen der Askomyzeten und Fungi imperfecti. Sydowia 26:169–198
Petrak F (1953) Ein Beitrag zur Pilzflora Floridas. Sydowia 7:103–132
Promputtha I, Miller AN (2010) Three new species of *Acanthostigma* (Tubeufiaceae, Dothideomycetes) from Great Smoky Mountains National Park. Mycologia 102:574–587
Rannala B, Yang Z (1996) Probability distribution of molecular evolutionary trees: a new method of phylogenetic inference. J Mole Evol 43:304–311
Réblová M, Barr M (2000) The genus *Acanthostigma* (Tubeufiaceae, Pleosporales). Sydowia 52:258–285
Rodrigues KF, Samuels GJ (1994) *Letendraeopsis palmarum*, a new genus and species of loculoascomycetes. Mycologia 86:254–258
Ronquist F, Huelsenbeck JP (2003) MrBayes 3: Bayesian phylogenetic inference under mixed models. Bioinformatics 19:1572
Romero AI, Samuels GJ (1991) Studies on xylophilous fungi from Argentina: VI. Ascomycotina on *Eucalyptus viminalis* (Myrtaceae). Sydowia 43:228–248
Rossman AY (1977) The genus *Ophionectria* (Euascomycetes, Hypocreales). Mycologia 69:355–391
Rossman AY (1978) *Podonectria*, a genus in the Pleosporales on scale insects. Mycotaxon 7:163–182
Rossman AY (1987) The Tubeufiaceae and similar Loculoascomycetes. Mycol Pap 157:1–71
Rossman AY, Samuels GJ, Rogerson CT, Lowen R (1999) Genera of Bionectriaceae, Hypocreaceae and Nectriaceae (Hypocreales, Ascomycetes). Stud Mycol 42:1–248
Samuels GJ, Müller E (1978) Life history studies of Brazilian Ascomycetes 2. Sydowia 31:137–141
Samuels GJ, Rossman AY, Müller E (1979) Life history studies of Brazilian Ascomycetes 6. Three species of *Tubeufia* with, respectively, dictyosporouspycnidial and helicosporous anamorphs. Sydowia 31:180–192
Sánchez RM, Bianchinotti MV (2010) New records in the Tubeufiaceae from Andean Patagonian forests of Argentina. Mycotaxon 111:131–141
Sánchez RM, Miller AN, Bianchinotti MV (2011) A new species of *Acanthostigma* (Tubeufiaceae, Dothideomycetes) from the southern hemisphere. Mycologia, doi:10.3852/11-105 (in press)
Scheuer C (1988) Ascomyceten auf Cyperaceen und Juncaceen im Ostalpen-raum. Biblioth Mycol 123:1–124
Scheuer C (1991) *Taphrophila* (Dothideales: Tubeufiaceae) and two species of *Tubeufia* with dark setae. Mycol Res 95:811–816
Schoch CL, Crous PW, Groenewald JZ et al (2009) A class-wide phylogenetic assessment of Dothideomycetes. Stud Mycol 64:1–15
Seifert KA, Morgan-Jones G, Gams W, Kendrick B (2011) The Genera of Hyphomycetes. CBS Biodiversity Series 9:1–997
Shearer C (1987) *Helicoma chlamydosporum*, a new hyphomycete from submerged wood in Panama. Mycologia 79:468–472
Sivanesan A (1984) The Bitunicate Ascomycetes and their Anamorphs Strauss and Cramer Vaduz, Germany
Sivanesan A, Panwar K, Kaur SJ (1976) *Thaxteriellopsis lignicola*, gen et sp nov, a new Loculoascomycete from India. Kavaka 4:39–42
Sivanesan A, Rajak RC, Gupta RC (1988) *Thaxterina*, a new tubeufiaceous genus with multispored asci from India. Trans Brit Mycol Soc 90:662–665
Spegazzini CL (1924) Algunos honguitos portoriqueños. Bol Acad Nac Ci 26:335–368
Subramanian C, Sekar G (1982) *Thaxteriellopsis lignicola* and its *Moorella* anamorph. Proc Plant Sci 91:1–7
Swofford DL (2002) PAUP*. Phylogenetic Analysis Using Parsimony (* and Others Methods), version 4.0b10 edition. Sinauer Associates, Sunderland, Massachusetts
Sydow H (1939) Fungi Venezuelani. Annales Mycol 28:29–224
Tsui CKM, Berbee ML (2006) Phylogenetic relationships and convergence of helicosporous fungi inferred from ribosomal DNA sequences. Mole Phyl Evol 39:587–597
Tsui CKM, Sivichai S, Berbee ML (2006) Molecular systematics of *Helicoma*, *Helicomyces* and *Helicosporium* and their teleomorphs inferred from rDNA sequences. Mycologia 98:94–104
Tsui CKM, Sivichai S, Rossman AY, Berbee ML (2007) Tubeufia asiana, the teleomorph of Aquaphila albicans in the Tubeufiaceae, Pleosporales, based on cultural and molecular data. Mycologia 99:884–894
Untereiner WA, Straus NA, Malloch D (1995) A molecular-morphotaxonomic approach to the systematics of the Herpotrichiellaceae and allied black yeasts. Mycol Res 99:897–913
Vasilyeva LN, Stephenson SL (2010) The problems of traditional and phylogenetic taxonomy of fungi. Mycosphere 1:45–51
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
Walker J (1980) *Gaeumannomyces*, *Linocarpon*, *Ophiobolus* and several other genera of scolecospored ascomycetes and *phialophora* conidial states, with a note on hyphopodia. Mycotaxon 11:1–129
White T, Bruns T, Lee S, Taylor J (1990) Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. PCR protocols: a guide to methods and applications 18:315–322
Zhang Y, Crous PW, Schoch CL, Hyde KD (2012) *Pleosporales*. Fungal Divers 52:1–221; doi: 10.1007/s13225-011-0117-x (in press)
Zhao G, Liu X, Wu W (2007) Helicosporous hyphomycetes from China. Fungal Divers 26:313–524