A MONOGRAPH OF THE FRESHWATER ASCOMYCETE FAMILY ANNULATASCACEAE: A MORPHOLOGICAL AND MOLECULAR STUDY

BY

STEVEN EDWARD ZELSKI

DISSERTATION

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Doctoral Committee:

Professor Emerita Carol A. Shearer, Chair, Director of Research
Research Professor Andrew N. Miller, Co-Chair, Co-Director of Research
Professor Emeritus David S. Seigler
Professor Stephen R. Downie

ABSTRACT

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Steven Edward Zelski, Ph.D.
Department of Plant Biology
University of Illinois at Urbana-Champaign, 2015
Carol A. Shearer and Andrew N. Miller, Co-advisors

Freshwater fungi are important agents decomposing submerged dead plant material. Roughly ten percent of the known teleomorphic (sexually reproducing) freshwater ascomycetes have been referred to or included in the family Annulatascaceae. Placement in this family is based on characters that include perithecial ascomata, unitunicate cylindrical asci with relatively large J- (Melzer's reagent negative) apical rings, and the presence of long tapering septate paraphyses. However, the large refractive apical apparati are the distinctive feature of the family. As sparse molecular data were available prior to the beginning of this study, a broad survey of freshwater temperate and tropical areas was conducted to collect these taxa for morphological examination, digital imagery, and extraction of DNA for phylogenetic inference. Thirty-five of roughly 70 described species in 21 genera of Annulatascaceae were assessed molecularly, and forty-five illustrated from holotypes and/or fresh collections. Twelve new taxa with relatively large non-amyloid apical apparati were discovered, only one of which, *Longicollum biappendiculatum*, belongs in Annulatascaceae. Material examined in this study was preserved as permanent slides, dried specimens, and/or living cultures. Phylogenetic analyses of 4 genes alone and in combination revealed a polyphyletic Annulatascaceae, with a *sensu stricto* clade consisting of the type, *Annulatascus velatisporus*, and nine other species. The other twenty-five taxa assessed molecularly were located in clades along the Sordariomycetes tree, basal to the Ophiostomatales, the nearest outgroup to Annulatascaceae in these analyses. Annulatascaceae *sensu stricto* lacks likelihood support at its root node in single and combined gene analyses, but is strongly supported by Bayesian posterior probability in

analyses of combined genes. This strongly suggests a familial if not an ordinal relationship of these ten taxa. One limitation of this study was a constraint on the number of genes sequenced. Some taxa only had GenBank sequence data and newly sequenced species did not all provide Mcm7 sequences as the primers are highly degenerate and annealing temperatures were variable. Addition of other genes (e.g. RPB1, TEF1α) to future analyses may increase confidence at both basal and distal nodes for this clade. A second limitation of this study was taxonomic coverage using sequence data. Though great effort was given to collecting, not all Annulatascaceae taxa were encountered. Collecting freshwater ascomycetes entails randomly selecting submerged woody debris and incubating it in moist chambers with the hopes of obtaining these microscopic organism. This is in contrast to collecting macroscopic organisms, which are readily recognizable in the field. Much work remains to be done on taxa thus far known only from the Eastern Hemisphere. Molecular data from these taxa would provide additional resolving power for lineages with large apical rings.

DEDICATION

This is dedicated to my ma n pa, without whose love I might not have been encouraged to explore.

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Without Dr. Steve Stephenson I would not have been able to visit Southeast Asia. Molecular data from collections made there have corroborated tropical distributions of many taxa in the course of the work presented here. The invitation to collect at Pea Ridge National Military Park increased the breadth of distribution patterns of some cosmopolitan species of freshwater fungi. What I learned about myxomycete distribution patterns has tempered my ideas about freshwater ascomycete distribution patterns.

To my brother Dr. John Janovec, collecting was almost a secondary goal to learning the cultures of your adopted home. We knocked out the collections like true professionals (thankfully the science needed incubation) and you introduced me to the social, economic, and political factors affecting a true biodiversity hotspot. Thanks to you, I can still feel optimistic about the future of these places we've explored. Rock n Roll.

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CHAPTER 1

INTRODUCTION

The Kingdom Fungi is estimated to consist of approximately 3.5-5.1 million species (O'Brien et al. 2005). Kirk et al. (2008) reported 97,861 described species. Of the described species, approximately 64,000 belong to the phylum Ascomycota, fungi that reproduce asexually by producing conidia or sexually by producing ascospores in sac-like structures called asci. Ascomycetes occupy nearly every habitat on Earth. They are reported from such disparate locations as Ernest Shackleton's camp in the Antarctic (Blanchette et al. 2010) to the high temperature geothermal formations in Yellowstone National Park (Redman et al. 1999). They can be found in frigid trenches and in high altitude streams (Le Calvez et al. 2009, Shearer et al. 2015). Ascomycetes perform heterotrophic roles, though when lichenized, they become part of a producer cooperative. They are plant and animal pathogens, saprobes, and symbionts.

Freshwater ascomycetes represent a relatively unstudied, taxonomically broad group of species. The group consists of species that spend the majority of their life cycle in water (indwellers) and species that may only spend part of their life cycle in freshwater (transients) (Park 1972). Freshwater ascomycetes have been studied in depth only over the last 60 years (Ingold 1942, Shearer 1972, Dudka 1985, Shearer 1993, Jones 2014, Shearer and Raja 2015, Shearer et al. 2015). C.T. Ingold focused on freshwater hyphomycetes whose spores accumulate in stream foam created by running water (Ingold 1942). His studies opened up the realm of an ecological group of filamentous fungi which reproduce asexually, and are typically associated with submerged substrates such as leaves and herbaceous material, as well as woody debris (Ingold 1942, Shearer 1972, Bärlocher 1985, Gulis et al. 2009). This work preceded DNA sequencing, and thus it was not known that the majority of these new taxa were members of the Ascomycota, though it was assumed due to the lack of hyphal clamp connections.

Freshwater ascomycetes are thought to play an important role in the decomposition of allochthanous and autochthanous woody and herbaceous debris in lentic and lotic aquatic habitats (Shearer 1992, Gessner and Chauvet 1994, Wong et al. 1998, Gessner and Van Ryckegem 2003). As saprobic organisms, they break down dead plant material such as autumn shed leaves, aquatic macrophytes, and woody debris (logs, branches, sticks, and roots) that are an important source of energy and nutrients in stream and lake food webs (Harmon et al. 1986). Their functional importance lies in their ability to break down complex plant compounds that invertebrates at the base of the food web cannot digest. Recalcitrant substrates such as cellulose and lignin are thereby reduced to smaller molecules that are then used as a food source for stream invertebrates (Zare-Maiven and Shearer 1988a, 1988b, Simonis et al. 2008). Fungi themselves are also consumed as food by a host of organisms (Bärlocher 1985).

Prior to Shearer undertaking the study of freshwater ascomycetes, which occur on long lasting lignocellulosic substrates, these organisms were poorly studied (Shearer 1972, 1993). Sexually reproducing ascomycetes in aquatic habitats had been sparsely reported prior to that. For example, the oldest species examined in this study, *Submersisphaeria vasicola* (Ellis and Everhart) Z. Wang, Aptroot & K.D. Hyde was reported in 1884 (Ellis and Everhart 1884), followed by *Pseudoproboscispora caudae-suis* (Ingold) J. Campb., Shearer, J.L. Crane & Fallah (Ingold 1951). All other taxa in this study were described beginning in 1992 and later, starting with *Annulatascus velatisporus* K.D. Hyde (Hyde 1992) the type species of the genus *Annulatascus* and family Annulatascaceae.

As an ecological assemblage of organisms, the freshwater ascomycetes do not themselves form a monophyletic group. It has been hypothesized that the adaptation to the freshwater environment was accomplished multiple times from various lineages within the classes Dothideomycetes, Orbiliomycetes, and Sordariomycetes (Shearer 1989, Belliveau and Bärlocher 2005, Vijaykrishna et al. 2006). Within the Sordariomycetes, the family Annulatascaceae, the focus of this study, accounts for nearly 10% of all described freshwater ascomycetes. Its position was considered *incertae sedis* at the subclass and ordinal levels (Huhndorf et al. 2004) until Maharachchikumbura et al. (2015) performed multi-gene analyses with the purpose of assessing the Sordariomycetes backbone tree and delimiting higher level classifications. At present, Annulatascaceae is placed in the order Annulatascales in the subclass Diaporthomycetidae (Maharachchikumbura et al. 2015).

The Annulatascaceae (roughly translated as "the family with rings in the asci") was created for freshwater ascomycetes which possessed the prominent feature of a relatively massive, Meltzer's reagent negative (J-) ring at the apex of the ascus (Wong et al. 1998) (**Fig. 1**). This morphological feature has proven to be symplesiomorphic at the generic to family level (Raja et al. 2003, Boonyuen et al. 2011, Ferrer et al. 2012, Raja et al. 2012, Jones et al. 2014). Though early molecular analyses warned of the symplesiomorphic nature of the annulus as a unifying character for the family, more genera and species were added to Annulatascaceae despite mounting evidence against the monophyly of the group (Ranghoo et al. 1999, Raja et al. 2003). These additions to the family included many taxa with much smaller apical rings, e.g. *Aquaticola*, than taxa originally included in the family. According to Kevin Hyde, one of the authorities of the family, basing the family on a large J- apical ring may have been misguided (Hyde pers. com. 2012). As a structural feature, ascus apical rings, whether staining with IKI or not, are common in Orbiliomycetes, Sordariomycetes, and Xylariomycetes (Beckett and Crawford 1973, Zhang et al. 2006, Kumar et al. 2012). As relatively inexpensive high throughput DNA sequencing was in its infancy at the time the family was established, morphology was paramount in grouping taxa. The family Annulatascaceae was initially placed in the Sordariales by Erikkson et al. (2001), but was moved to Sordariomycetes *incertae sedis* by Huhndorf et al. (2004) based on 28S rRNA analysis, in keeping with the opinions of Kirk et al. (2001). Thongkantha et al. (2009) showed that Ophiostomatales was closely related to Annulatascaceae *sensu stricto* in combined 18S and 28S analyses. The majority of publications formally describing members of the family have been based on morphology alone, with the relatively massive ascus apical ring (not so massive in some taxa) as the unifying character. An early study using molecular data by Ranghoo et al. (1999) showed that members of the genus *Ascotaiwania* Sivan. & H.S.

Fig. 1. Representative ascus apical rings of Annulatascaceae species. A. Ascus illustrating apical ring of *Annulatascus velatisporus*. B. Ascus apical ring of *Annulatascus biatriisporus* stained with aqueous nigrosin. C. Ascus apical ring of *Torrentispora crassiparietis*. D. Ascus apical ring of *Annulatascus velatisporus*. E. Ascus apical ring of *Torrentispora crassiparietis* end on. F. Ascus apical ring of *Cataractispora viscosa*. G. Ascus apical ring of *Submersisphaeria aquatica*. Bars: A = 40 µm; B-F = 20 µm; G = 10 µm.

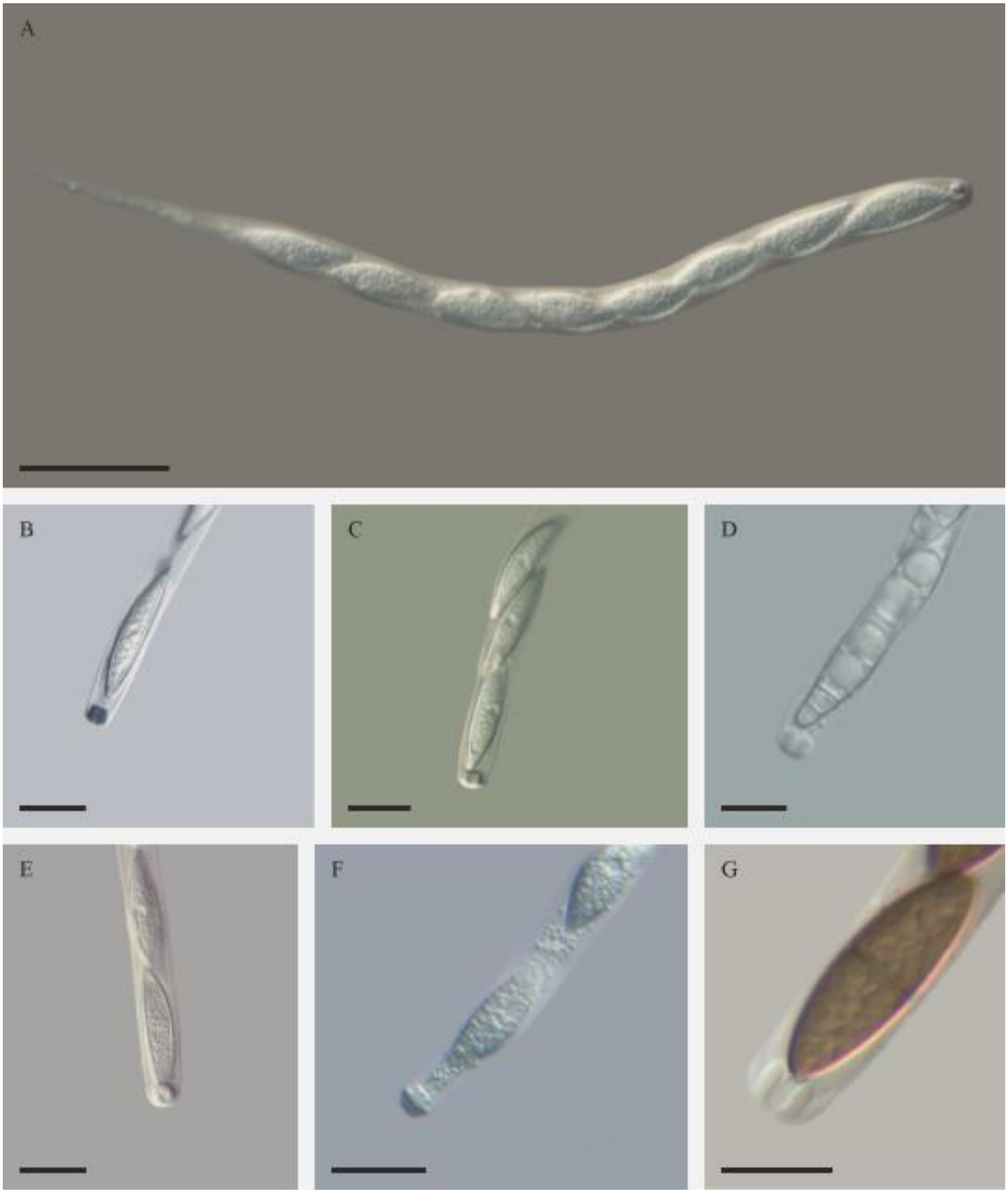


Fig.1

Chang, which were originally thought to belong in the Annulatascaceae due to their large bipartite apical rings, were distantly related to the type of the family, *A. velatisporus*. *Ascotaiwania* is currently placed in the Savoryellales based on a five gene phylogeny, though it has not yet been placed in a family (Boonyuen et al. 2011). Annulatascaceae, as currently circumscribed, is composed of 18 genera with approximately 71 species. Initially, the family consisted of six genera: *Annulatascus, Ascotaiwania, Clohiesia* K.D. Hyde*, Frondicola* K.D. Hyde*, Proboscispora* S.-W. Wong and K.D. Hyde*,* and *Submersisphaeria* K.D. Hyde (Wong et al. 1998). As mentioned above, *Ascotaiwania* has been removed and placed in the order Savoryellales based on molecular data (Boonyuen et al. 2011); *Proboscispora* was renamed *Pseudoproboscispora* Punith. as the name had been previously given to an unrelated species (Punithalingham 1999); and 21 new genera have been added while 3 were removed. Currently the following genera are recognized or referred to Annulatascaceae: *Annulatascus, Annulusmagnus* J. Campbell & Shearer*, Aqualignicola* Ranghoo, K.M. Tsui & K.D. Hyde, *Aquaticola* W.H. Ho, K.M. Tsui, Hodgkiss & K.D. Hyde*, Ascitendus* J. Campbell & Shearer*, Ascolacicola* Ranghoo & K.D. Hyde*, Ayria* Fryar & K.D. Hyde*, Brunneosporella* Ranghoo & K.D. Hyde*, Cataractispora* K.D. Hyde, S.W. Wong & E.B.G. Jones, *Chaetorostrum* Zelski, Raja, A.N. Mill & Shearer, *Clohiesia*, *Conlarium* F. Liu & L. Cai, *Cyanoannulus* Raja, J. Campbell & Shearer*, Diluviocola* K.D. Hyde, S.W. Wong & E.B.G. Jones, *Fluminicola* S.W. Wong, K.D. Hyde & E.B.G. Jones*, Frondicola*, *Fusoidispora* Vijaykr., Jeewon & K.D. Hyde, *Longicollum Zelski*, Raja, A.N. Mill & Shearer, *Pseudoproboscispora, Rhamphoria* Niessl, *Rivulicola* K.D. Hyde*, Submersisphaeria*, *Teracosphaeria* M. Réblová & K.A. Seifert, *Torrentispora* K.D. Hyde, W.H. Ho, E.B.G. Jones, K.M. Tsui & S.W. Wong, and *Vertexicola* K.D. Hyde, Ranghoo & S.W. Wong.

Four genera in Annulatascaceae, *Annulatascus*, *Aquaticola, Cataractispora, and Submersisphaeria*, account for 34 species, or approximately 50% of the family. These genera are relatively easy to distinguish based on morphology. Preliminary molecular studies with a limited number of taxa have shown the genera segregate with strong Maximum likelihood (ML) and Bayesian posterior probability (PP) branch support from one another (Raja et al. 2003, Campbell and Shearer 2004, Abdel-Wahab et al. 2011), but not as a monophyletic group. Furthermore, other molecular studies have shown that terrestrial genera such as *Ambrosiella* Brader, *Fragosphaeria* Shear, and *Ophiostoma* Syd & P. Syd, which differ morphologically from Annulatascaceae taxa, show phylogenetic affinities with this predominantly freshwater ecological group (Vijaykrishna et al. 2005, Boonyuen et al. 2012, Ferrer et al. 2012). In fact, when representatives of the latter genera are included in phylogenetic analyses, they divide the Annulatascaceae into *sensu stricto* and *lato* clades, and effectively insert whole non-related lineages between them (Jones et al. 2014).

The family Annulatascaceae is essentially an assortment of species primarily based on the presence of a J- apical ring that varies in size among genera. Additional characteristics of Annulatascaceae include: dark or light colored perithecia with a cylindrical neck; long, tapering, septate paraphyses; long cylindrical asci with relatively large apical rings which do not stain blue in the iodine based Meltzer’s reagent, but do stain in aqueous nigrosin; and ascospores that are aseptate or septate, mostly hyaline, occasionally brown, and with or without gelatinous sheaths or appendages (Wong et al. 1998). Studies of the ascus apices of taxa currently included in the Annulatascaceae have revealed variation in the size, shape, structure, and staining in various reagents (Wong et al. 1999, Ho et al. 1999, Ranghoo et al. 2001, Raja et al. 2003). An apical ring may be an important structural element of the ascus which leads to back door dehiscence of asci to release ascospores rather than forceful ejection from the ascus tip. The backdoor release mechanism of ascospore discharge is a common feature of Annulatascaceae taxa. Backdoor dehiscence occurs when the ascus wall splits open 1/3 to 2/3 of the length of the ascus from the tip, typically breaking in half, and thus releasing the ascospores. Other variable features used to delimit genera within Annulatascaceae include the size and morphology of ascospores and the presence/absence of gelatinous sheaths and/or appendages. Gelatinous sheaths and appendages are thought to be adaptations for ascospore adherence to substrates in an aquatic environment (Shearer 1993).

In the case of Annulatascaceae, a natural classification based on morphology and backed by molecular data is difficult as there are few morphological characters or suites of characters with which to group taxa when molecular evidence is included. Suites of characters are problematic as they often overlap in one or more categories. Molecular analyses are the gold standard in determining the monophyly of particular groups and then morphology is evaluated in relation to those those hypotheses.

Morphology-based taxonomy of Annulatascaceae has resulted in a large and variable assemblage which is only slowly being teased apart using molecular techniques (Ranghoo et al. 1999, Réblová and Winka 2001, Campbell et al. 2003, Raja et al. 2003, Campbell and Shearer 2004). This recent molecular work has suggested polyphyly of Annulatascaceae, with authors recently referring to a *sensu stricto* clade clustering with *A. velatisporus*, and a series of *sensu lato* clades that are not phylogenetically related (Abdel-Wahab et al. 2011, Jones et al. 2014). Predominantly due to studies using only genes that resolve tip taxa (e.g. 28S nrRNA) and which are less effective in resolving deeper branches in phylogenetic analyses, the family itself is currently considered *incertae sedis* at the subclass and ordinal levels. Thus genes more reflective of deeper branches such as SSU and single-copy protein coding genes such as Mcm7 require investigation (Aguileta et al. 2008, Schmitt et al. 2009, Raja et al. 2011). Addition of such gene sequences would also increase the characters available for comparisons. In addition, taxon sampling for molecular studies of Annulatascaceae has been somewhat limited. Species of Annulatascaceae have been reported from every continent except Antarctica and adding morphologically similar taxa from additional biogeographic regions to molecular analyses would increase resolving power.

Taxa in the Annulatascaceae *sensu stricto* clade agree with one another in that they possess relatively large J- ascus apical rings (Wong et al 1998, Ho and Hyde 2000) but have marked variation in traits such as ascomatal structure and pigmentation and ascospore shape, pigmentation, septation, guttulation, appendages, and sheaths. Therefore, to further understand phylogenetic relationships among Annulatascaceae *sensu lato*, this study was comprised of three main goals. The first was to perform a broad geographic survey of Annulatascaceae in order to isolate and extract DNA for molecular analyses for as many taxa as possible. The second was to illustrate these taxa and compare them using a database constructed of morphological characters. The third goal was to conduct molecular analyses using single and multi-gene alignments with as many taxa as possible to assess the genetic relatedness of Annulatascaceae-like taxa and determine which, if any morphological characters are phylogenetically informative based on molecular results.

These efforts were made to address the following questions: 1) Which of the currently accepted taxa in the Annulatascaceae form the monophyletic group with the type species of the family and consequently are the Annulatascaceae *sensu stricto*? 2) Which morphological character or set of characters are phylogenetically informative for the circumscription of the family and the genera therein? 3) Which species should be retained in Annulatascaceae, and which need taxonomic reassignment in light of combined morphological and molecular evidence? and 4) What is the placement of the Annulatascaceae *sensu stricto* at the subclass and ordinal levels, and what are the placements of those taxa removed from that clade?

In addition to addressing these questions, the following hypotheses and predictions were tested: 1) If bipartite J- ascus apical rings are widespread throughout Sordariomycetes, then this character alone will not be phylogenetically informative for the Annulatascaceae *sensu stricto*. 2) Since ascospore morphology is under selective pressure in aquatic habitats, then ascospore characters such as shape, pigmentation, and presence/absence of gelatinous appendages and/or sheaths will not be phylogenetically informative for the Annulatascaceae *sensu stricto*. 3) If ascomal architecture given its complexity is a more conserved trait it will therefore be more phylogenetically informative for Annulatascaceae than ascospore characters.

This study has merit in that it tackles the longstanding problem of the phylogenetic systematics of Annulatascaceae. It also provides distributional information, new illustrations, a compilation of morphological data, and phylogenetic inference. Vouchered herbarium specimens are deposited at the University of Illinois Herbarium, allowing U.S. and foreign institutions to access the material via herbarium-to-herbarium loans. Gene sequences are deposited through NCBI into GenBank and contribute to the information available to the scientific community. This dissertation will be made widely available for future research on this topic and the electronic morphological database constructed for the study will be disseminated upon request.

CHAPTER 2

MATERIALS AND METHODS

Collecting sites:

Sample collecting for this research was conducted in primarily tropical freshwater habitats in Asia and in North, Central, and South America, with permission to collect and export from the countries in which the collecting was performed. Collections were also made in the temperate U.S. From these collections, many Annulatascaceae taxa described from the Eastern Hemisphere and for which prior molecular data did not exist were recovered. See Table 1 for a complete list of collection sites and environmental factors. Of special note are the Los Amigos Biological Field Station/Centro de Investigación y Capacitación Rio Los Amigos (CICRA) and the Great Smoky Mountains National Park (GSMNP), that are abbreviated for the remainder of the text.

Habitat characterization:

Sites were characterized by geographic location, and with GPS coordinates (including altitude (m) above sea level) (Table 1). Environmental conditions were also recorded, e.g., pH, temperature (°C), dissolved oxygen content (DOC), and electroconductivity (µS). When possible, surrounding vegetation was included in site descriptions. These observations were used to characterize the abiotic and biotic factors influencing species distributions. Original descriptions served as historical site characterization.

Field Collection and Herbarium Requests:

One of the main goals of this study was to collect fresh specimens of Annulatascaceae for morphological and molecular characterization. These organisms are microscopic, in the order of 1 mm or smaller, and cannot be readily identified in the field. Therefore, random sampling of submerged woody debris followed by microscopic examination in the laboratory was the chosen method for recovering the highest diversity of freshwater ascomycetes, and in turn the most taxa of Annulatascaceae. Submerged woody debris was collected randomly from various lentic and lotic freshwater habitats according to procedures outlined by Shearer et al. (2004). Particular care was given to selecting submerged plant material that had been submerged for an extended period of time, i.e., long enough to permit colonization of the substrates by freshwater ascomycetes and the exclusion of terrestrial species. To ensure substrate suitability, debris was subjected to three tests. The first was a visual inspection of the substrate to determine if the material was of recent deposition; in which case it was discarded. The second test was to perform a “fingernail test” to determine if the material was easily penetrated and thus decomposing. Finally, suitable substrate was subjected to a quick snap to determine if it was breakable or still green, as recently senesced woody debris has not had time to be colonized. Satisfactory substrates were put in sealable plastic bags along with sheets of moist paper towels and shipped to the University of Illinois at Urbana-Champaign.

It was anticipated from the outset that not all taxa would be encountered, and thus type descriptions and herbarium specimens were used when fresh material was not available. Herbarium requests were made for all type specimens. Table 2 documents the herbarium location, collection details, and collector(s) for each holotype, as well as whether the holotype was available. Many of the holotypes requested could not be found by the curators of HKU or IFRD and one from IMI, and this is noted in the rest of the text. Some may have been misplaced in the move of the Hyde Collection from HKU to IFRD.

Examination of Substrates:

Fresh samples of woody debris were incubated in moist chambers and examined for reproductive structures within one week of arrival and periodically thereafter for 12 months. All available holotypes were examined and additional dried herbarium specimens were examined as time allowed. Reproductive structures were located with an AO Spencer Zoom Stereomicroscope (American Optical Co., Southbridge, MA). Ascomata or conidiophores were removed from the substrate with a sterilized dissecting needle, gently placed (conidiophores) or teased apart (ascomata) in a drop of distilled water on a 25 × 25 mm cover slip and covered with an 18 × 18 mm cover slip (Corning Inc., Corning, NY; VWR International, LLC, Radnor, PA) on a 25 × 75 × 1mm, plain, pre-cleaned microscope slides (Corning Inc., Corning, NY; Fisher Scientific, Pittsburg, PA). Freshly made slides were preserved according to the methods of Volkmann-Kohlmeyer and Kohlmeyer (1996). In cases where material was unavailable, protologues were used to study the fungi.

Isolation:

Isolation in pure culture was performed according to procedures modified from Fallah and Shearer (2001) and Shearer et al. (2004). For single spore isolation, sterile dissecting needles were used to gently tease apart the contents of ascomata on antibiotic water agar [AWA, 20 g agar (Difco), 0.5 g streptomycin sulfate (Sigma), 0.5 g penicillin G (Sigma) and 1000 mL deionized H_2O]. Single germinated ascospores were transferred to sterile Petri dishes containing peptone yeast glucose agar with antibiotics [PYG + Ab, 1.25 g peptone, 1.25 g yeast extract, 18 g agar, 5 g D-glucose (Acros), 0.5 g streptomycin sulfate, 0.5 g penicillin G, and 1000 mL deionized H_2O] and grown at ambient temperature with ~ 12/12 hr light/dark conditions. Antibiotics were added to media after autoclaving with a brief cool down period (5 min). For taxa that did not germinate, either ~30 ascomata or a mass of ascospores were harvested and put in AP1 buffer from the DNeasy Plant Mini Kit (Qiagen Sciences Inc., Valencia CA).

Imaging:

Macroscopic digital images of herbarium packets and their contents were taken with an iPhone 5s (Apple Inc., CA). Macroscopic digital images of fruiting structures were taken using an Olympus SZX7 stereomicroscope (Olympus Optical Co. Ltd., Tokyo, Japan) fitted with a SPOT RT color camera using SPOT Advanced software (Diagnostics instruments Inc., Sterling Hts., MI). Light microscopic examination of ascomal architecture was performed on an Olympus BHS microscope equipped with

Nomarski interference and phase optics (Olympus Optical Co. Ltd., Tokyo, Japan). Digital micrographs were obtained with a SPOT Insight 12 Mp color camera and Spot Advanced software. Images were processed with Adobe Photoshop v. 9.0.2 and assembled with Adobe InDesign v. 4.0.5.

Specimen preservation:

Permanent slides were prepared using the protocol of Volkmann-Kohlmeyer and Kohlmeyer (1996). Field material, when available, was also dried and deposited with permanent slides in ILL.

Molecular Methods

DNA extraction:

DNA was extracted using different methods depending on whether pure cultures were obtainable. In cases where cultures were obtained, DNA was extracted from mycelium scraped with a sterile spatula from agar plates. Mycelium was ground into a fine powder in liquid nitrogen with a mortar and pestle and DNA was extracted using the DNeasy Plant Mini Kit (Qiagen Sciences Inc., Valencia CA) according to the manufacturer's instructions. In cases where spores did not germinate, fruit bodies or spores were pooled in AP1 buffer (Qiagen Sciences Inc., Valencia CA) and subjected to three freeze/thaw cycles in liquid nitrogen (-196 °C) with micropestle pulverization before extraction with the DNeasy Plant Mini Kit.

PCR:

PCR amplification of extracted DNA was performed using Illustra Ready-To-Go™ PCR Beads (GE Healthcare) on an MJ Research PTC-200 thermocycler. PCR was performed using appropriate primer pairs (Table 3) for the 28S nrDNA, 18S nrDNA, ITS nrDNA, and the nuclear single copy protein Mcm7 genes. The following parameters were used for ribosomal genes: initial denaturation at 95 °C for 5 min, followed by 40 cycles at 95 °C for 30 s, 50 °C for 15 s, 72 °C for 10 s with a final extension step of

72 °C for 10 min. The following parameters were used for Mcm7: initial denaturing at 94 °C for 5 min; 30 cycles of denaturing at 94 °C for 45 s, annealing at 50-60 °C (empirically determined) for 50 s; extension at 72 °C for 1 min; and a final extension step of 72 °C for 5 min. PCR products were purified using a QIAquick PCR Purification Kit (Qiagen Sciences Inc., Valencia CA) according to the manufacturer's instructions.

Sequencing:

Sequencing reactions were carried out using the BigDye® Sequence Terminator Kit ver. 3.1 (Applied Biosystems, Foster City, CA) using appropriate primers (Table 3). Sanger DNA sequencing was performed on an AB 3730xl DNA Analyzer at the W. M. Keck Center for Comparative and Functional Genomics at the University of Illinois at Urbana-Champaign.

Taxon sampling:

As many Annulatascaceae or Annulatascaceae-like species as possible were examined and sequenced. Additional representatives of the major orders of Sordariomycetes were added to alignments of Annulatascaceae taxa. The majority of these taxa were drawn from the Sordariomycetes study of Zhang et al. (2006) and the recent molecular phylogenetic study of freshwater fungi, specifically Chapter 3, by Jones et al. (2014). In addition, Sordariomycetes taxa with Mcm7sequences from Raja et al. (2011) were included to provide taxonomic coverage for that gene. A complete list of taxa and sequence information can be found in Table 6.

Sequence assembly:

Sequences were assembled and initially aligned using Sequencher 4.9 (Gene Codes Corporation, Ann Arbor, MI). Further alignment was performed using MUSCLE 3.6 (Edgar 2004) in SeaView 4 (Gouy et al. 2010) followed by refinement by eye.

Phylogenetic analyses:

Maximum likelihood analyses were performed using RAxML-HCP2 v.7.0.4 on XSEDE (Stamatakis 2006, Stamatakis et al. 2008) on the CIPRES Portal v.2.0 (Miller et al. 2010) under default settings and GTRCAT model to increase inference time with 1000 rapid bootstrap searches. Most likely trees from likelihood analyses were initially viewed using Dendroscope v.3 (Huson and Scornavacca 2012) and then illustrated in Adobe Illustrator CS2. Bayesian analyses were conducted using MrBayes v.3.2.3 on XSEDE (Huelsenbeck et al. 2001, Ronquist and Huelsenbeck 2003) on the CIPRES Portal v.2.0 (Miller et al. 2010) using parameters obtained with jModelTest (Posada 2008, Darriba et al. 2012) with two runs and four chains. A total of 10,000,000 generations were run with trees sampled every 1000th generation, resulting in a total of 10,000 trees. The first 1,000 trees were discarded as burn in. All analyses had reached stationarity by that point, and the remaining 9,000 trees were used to establish posterior probabilities (PP). The consensus tree was viewed in Dendroscope 3 (Huson and Scornavacca 2012).

CHAPTER 3

RESULTS

PHYLOGENETIC ANALYSES

Thirty-five species, or approximately half of the taxa referred to Annulatascaceae, were evaluated molecularly in this study. Twenty of these species were evaluated using molecular data for the first time or with greater gene coverage. After ends were trimmed due to missing data and one 19 bp ambiguous region was removed, the final 28S alignment consisted of 203 sequences from 148 taxa, and had a length of 632 bp. Only the D1-D2 (LROR-LR3) region was used due to its variability and to reduce sequencing costs. After ends were trimmed due to missing data the final 18S alignment (NS1-NS4) consisted of 113 sequences from 90 taxa, and had a length of 1132 bp. After ends were trimmed due to missing data and ambiguous regions excluded, the final ITS alignment (ITS1F-ITS4) consisted of 73 sequences from 49 taxa, and had a length of 763 bp. After ends were trimmed due to missing data the final Mcm7 alignment (709for-1348rev) consisted of 43 sequences from 35 taxa, and had a length of 645 bp. The combined alignment of 18S and 28S consisted of 201 sequences from 148 taxa, and had a length of 1764 bp. The combined alignment of 18S, 28S, and Mcm7 consisted of 203 sequences from 148 taxa and had a length of 2409 bp (see Table 4). ITS sequences were not included in combined gene analyses as it is difficult to align beyond the familial level.

For Maximum likelihood and Bayesian analyses, jModelTest2 (Darriba et al. 2012) was used to determine the best fit models of nucleotide evolution for each gene. Parameters for each gene and combined datasets are listed in Table 5.

Tree results constructed from maximum likelihood analyses performed with RAxML are presented below first on a gene by gene basis (**Figs. 2-5**), followed by combinatorial variations (**Figs. 6-**

8). Bayesian PP values ≥ 95 and Maximum Likelihood bootstrap values ≥ 70 are used to indicate supported branches on all trees.

Maximum-likelihood RAxML analysis of the 28S dataset produced a single most likely tree (**Fig. 2**). Annulatascaceae *sensu stricto* is a monophyletic group with Ophiostomatales as the nearest basal order. Included in the *sensu stricto* clade along with the type of the family, *Annulatascus velatisporus,* are *A. aquatorba, A. hongkongensis, A. nilensis, Annulusmagnus triseptatus, Ascitendus austriacus, Longicollum biappendiculatum, Paoayensis lignicola, Pseudoproboscispora caudae-suis,* and *Submersisphaeria aquatica*. Additionally, and in contrast to combined gene analyses, *Annulatascus citriosporus* is placed as a sister taxon to *A. aquatorba,* although without support and on a long branch. Other Annulatascaceae taxa with 28S data are scattered throughout the Sordariomycetes in the three gene analysis results below.

Maximum-likelihood RAxML analysis of the 18S dataset produced a single most likely tree (**Fig. 3**). Annulatascaceae *sensu stricto* as inferred from 28S analysis is paraphyletic in 18S analyses. Rather, what would constitute the *sensu stricto* clade inferred from 18S analyses includes two members of the Diaporthales, *Ophioceras tenuisporum* and *Ceratosphaeria lampadophora*, though the branch leading to them is unsupported. Aside from these two species, the Annulatascaceae *sensu stricto* clade contains the same taxa as that produced in the 28S analyses except for *A. citriosporus*, for which 18S data was not obtained (see below and **Fig. 2**). Other Annulatascaceae taxa with 18S data included in this study are scattered throughout the Sordariomycetes in the three gene analysis results below.

Maximum-likelihood RAxML analysis of the ITS dataset produced a single most likely tree (**Fig. 4**). Annulatascaceae *sensu stricto* is paraphyletic group compared to the group in 28S analyses (see below and **Fig. 2**) with the exception that *Annulatascus hongkongensis, A. nilensis,* and *Paoayensis lignicola* are not included as ITS data are currently unavailable for these taxa as these taxa were not collected in this study. *Annulatascus citriosporus,* which was on a long branch in 28S analyses, is placed sister to

Submersisphaeria aquatica on a short branch in ITS analyses. Other Annulatascaceae taxa with ITS data included in this study are scattered throughout the Sordariomycetes in the three gene analysis results below. Exceptions to this are *Aquaticola tropicalis* and the informally described *Aq. hongkongensis* (*nomen invalidum* with sequence) which are only represented in GenBank with ITS sequences. *Aquaticola tropicalis* belongs in a clade with the new *Aquaticola*-like species PE0316 and *Aq. miniguttulata*, while *Aq. hongkongensis* is placed in a polytomy sister to *Conlarium duplumascospora* on a relatively long branch with no statistical support.

Maximum-likelihood RAxML analysis of the Mcm7 dataset produced a single most likely tree (**Fig. 5**). Annulatascaceae *sensu stricto*, compared to 28S analysis, is not a monophyletic group in this analysis as it consists of two clades. Inclusion of *Aquaticola miniguttulata*, two new *Aquaticola*-like species (AK1838, PE0316), *Chaetorostrum quincemilense, Fluminicola bipolaris*, and *Vertexicola triseptata* would be required for monophyly..

Maximum-likelihood RAxML analysis of the combined 18S and 28S datasets produced a single most likely tree (**Fig. 6**). Annulatascaceae *sensu stricto* is a monophyletic group with Ophiostomatales as its sister group. This clade is taxonomically congruent to that produced in 28S analysis, with the exception that *Annulatascus citriosporus* is not a member, but rather is placed sister to members of Melanosporales and Coronophorales. Other Annulatascaceae taxa with 28S and/or 18S data included in this study are scattered throughout the Sordariomycetes and are broken into clades in the three gene analyses.

Maximum-likelihood RAxML analysis of the combined 18S, 28S and Mcm7 datasets (the most robust analysis with the most data in this study, and that which is used for morphological/molecular synthesis) produced a single most likely tree (**Fig. 7**). Annulatascaceae *sensu stricto* forms a ten species clade within **Clade 1** in three gene analyses and is taxonomically congruent to the same genera and species as in the 28S analysis above, with the exception of *A. citriosporus*, which is rather distantly related. The clade is divided into 4 subclades. The nearest sister group consists of members of the Ophiostomatales and *Xylomelasma sordida*. Currently accepted or referred Annulatascaceae species with molecular data not in the Annulatascaceae *sensu stricto* clade are scattered throughout the Sordariomycetes in eight Annulatascaceae *sensu lato* clades as follows (referred to in **Fig. 7** as **Clades** for simplicity).

Clade 1 (treated as a clade here to facilitate the results and discussion) consisting of numerous Annulatascaceae *sensu lato* members is a large group of predominantly freshwater taxa (exceptions are *Lentomitella* spp. (although *Lentomitella pallibrunnea* has been recovered from freshwater (Shearer et al. 2015)) and *Rhamphoria delicatula*) and is placed sister to Ophiostomatales and Annulatascaceae *sensu stricto* (shaded taxa in **Fig. 7, Clade 1**). Inclusion of Ophiostomatales and Annulatascaceae *sensu stricto* causes **Clade 1** to become monophyletic. **Clade 1** is further subdivided into **Clades 1a-1k**. This large lineage does not have statistical support (< 70 ML, < 95 PP) at its basal node. Other Annulatasceae *sensu lato* species examined in this study are dispersed among **Clades 2-8** (**Fig. 7**).

Annulatascaceae *sensu lato* **Clade 1a** is composed of a group of freshwater fungi statistically supported by high Bayesian posterior probability values but not by Maximum-likelihood bootstrap support values. The clade is composed of *Brunneosporella aquatica, Chaetorostrum quincemilense, Fluminicola bipolaris*, and *Papulosa amerospora* (**Fig. 7, Clade 1a**).

Annulatascaceae *sensu lato* **Clade 1b** is composed of the *Cataractispora* species *C. appendiculata, C. receptaculorum*, and *C. viscosa,* as well as *Aquaticola hyalomura*, although without Bayesian posterior probability or Maximum-likelihood bootstrap support (**Fig. 7, Clade 1b**). Annulatascaceae *sensu lato* **Clade 1c** is composed of the *Aquaticola*-like species PE0157 and PE0264 as well as *Ayria nubispora.* The *Aquaticola*-like species are well supported sister taxa, but *Ayria nubispora* does not have Bayesian posterior probability or Maximum-likelihood bootstrap support (**Fig. 7, Clade 1c**).

Annulatascaceae *sensu lato* **Clade 1d** is composed of the freshwater fungi *Conlarium duplumascospora*, *Hydromelitis pulchella*, and *Riomyces rotundus*. *Conlarium duplumascospora* and *R. rotundus* are statistically well supported sister taxa, while *H. pulchella* is sister to the two species without Bayesian posterior probability or Maximum-likelihood bootstrap support (**Fig. 7, Clade 1d**).

Annulatascaceae *sensu lato* **Clade 1e** is composed of *Vertexicola caudatus* and the new species *V. triseptata* from Perú. This clade is strongly supported by Bayesian posterior probability and Maximum-likelihood bootstrap support values (**Fig. 7, Clade 1e**). These species agree well morphologically but differ in measurements of characters, the lack of a spike-like ascus tail in *V. triseptata*. The ascospores of *V. triseptata* are consistently 3-septate rather than 5-septate as in *V. caudatus*.

Annulatascaceae *sensu lato* **Clade 1f** is well supported as a sister to **Clade 1g** by Bayesian posterior probability but not by Maximum-likelihood bootstrap values. **Clade 1f** is composed of *Annulatascus biatriisporus*, a new *Annulatascus*-like species (PE0035), *Aquaticola*-like species (AK-1838, PE0316), *Aq. miniguttulata*, and *Fusoidispora aquatica*. Within **Clade 1f**, PE0316, *Aq. miniguttulata*, and *F. aquatica* are strongly united by both statistical metrics (**Fig. 7, Clade 1f**).

Annulatascaceae *sensu lato* **Clade 1g** is a monotypic clade with *Torrentispora fibrosa* as its sole member. It is separated from **Clade 1f** by a clade comprised of *Lentomitella* species, which possess large J- apical rings, but have thick ascomal walls and ill-defined necks. The latter two clades are sister to one another, supported by Bayesian posterior probability. The relationship of *T. fibrosa* to those clades lacks statistical support with either metric used in this study (**Fig. 7, Clade 1g**). *Torrentispora fibrosa* has black ascomata with well-defined necks, but differs from taxa in **Clade 1f** in that its ascospores are aseptate, ellipsoidal and have thick walls. The only taxon in **Clade 1g** with thick-walled ascospores is *Annulatascus biatriisporus*, but they are fusiform and have a thin sheath.

Annulatascaceae *sensu lato* **Clade 1h** is a monotypic clade with *Cyanoannulus petersenii* as its sole member (**Fig. 7, Clade 1h**). A feature that sets *C. petersenii* apart from other taxa is that the ascomata are reddish-brown as opposed to brown, black, or hyaline.

Annulatascaceae *sensu lato* **Clade 1i** is a monotypic clade, with *Hyalorostratum brunneisporum* as its sole member, sister to **Clade 1h** without Bayesian posterior probability or Maximum-likelihood bootstrap support values (**Fig. 7, Clade 1i**).

Annulatascaceae *sensu lato* **Clade 1j** consists of three species of freshwater ascomycetes not referred to Annulatascaceae per se and placed in the genus *Bullimyces* (**Fig. 7, Clade 1j**).

Annulatascaceae *sensu lato* **Clade 1k** is a monotypic clade with *Teracosphaeria petroica* as its sole member (**Fig. 7, 1K**).

Annulatascaceae *sensu lato* **Clade 2** is a monotypic clade with *Annulatascus citriosporus* (represented by two sequences generated in this study) as its sole member. It is on a relatively long branch sister to Chaetosphaeriales, although without any statistical support (**Fig. 7, Clade 2**).

Annulatascaceae *sensu lato* **Clade 3** is a polytomy that is divided into 3 subclades. **Clade 3a** is comprised of *Aquaticola triseptata* and the new *Aquaticola*-like species PE0159 (**Fig. 7, Clade 3a**). **Clade 3b** is monotypic with *Submersisphaeria rattanicola* as its sole member (**Fig. 7, Clade 3b**). **Clade 3c** is monotypic with *Clohiesia corticola* as its sole member (**Fig. 7, Clade 3c**

Annulatascaceae *sensu lato* **Clade 4** is a strongly statistically supported clade composed of two *Aquaticola* species, *Aq. ellipsoidea* and the informally describe *Aq. hongkongensis* (**Fig. 7, Clade 4**).

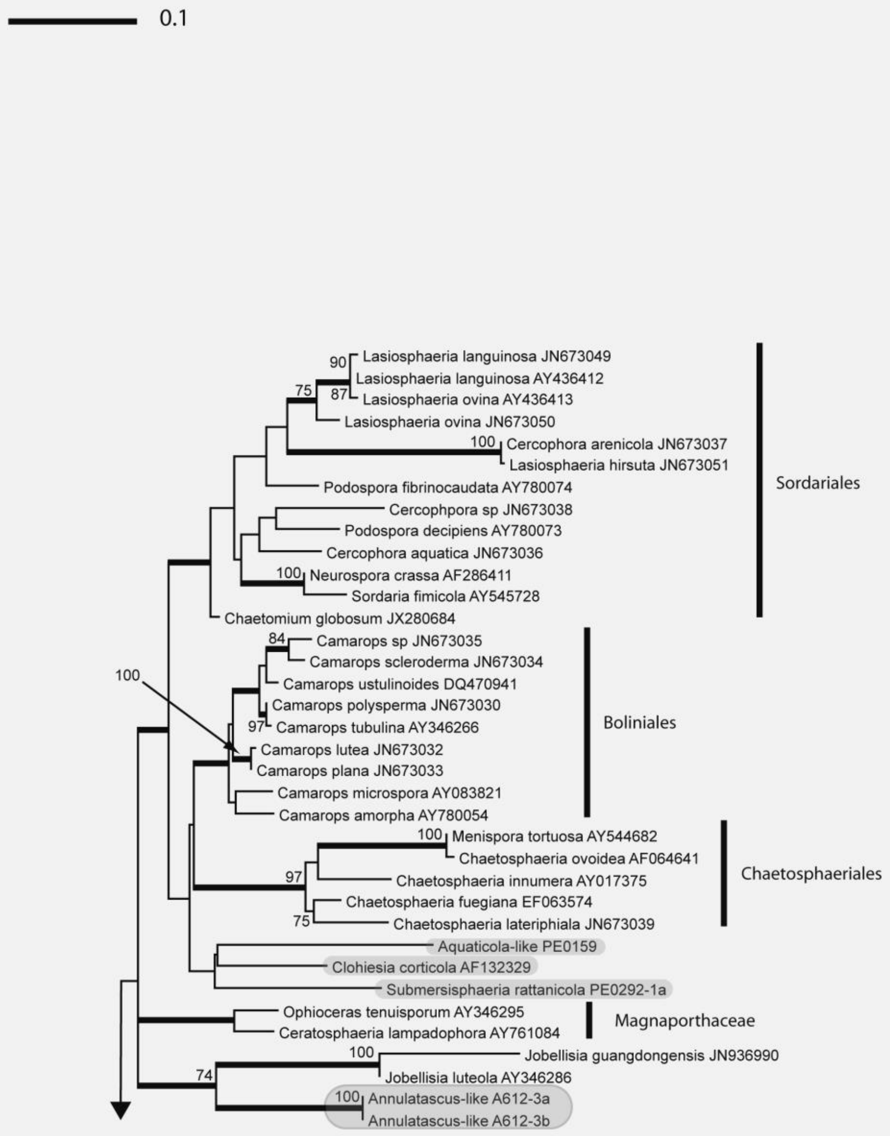
Annulatascaceae *sensu lato* **Clade 5** is a monotypic clade with *Torrentispora crassiparietis* as its sole member (**Fig. 7, Clade 5**).

Annulatascaceae *sensu lato* **Clade 6** is a monotypic clade with *Rivulicola incrustata* as its sole member (**Fig. 7, Clade 6**).

Annulatascaceae *sensu lato* **Clade 7** is composed of a new *Annulatascus*-like species (PE0324) and a new *Ayria*-like species (PE0323) which are strongly supported sister taxa Although PE0323 is on a longer branch. (**Fig. 7, Clade 7**).

Annulatascaceae *sensu lato* **Clade 8** monotypic with the new *Annulatascus*-like species A612 as its sole member (**Fig. 7, Clade 8**).

Fig. 2. Phylogram of the most likely tree from 28S nrRNA gene analysis obtained with RAxML (-ln = -17440.773436). Numbers ascribed to branches indicate ML bootstrap support values ≥70%, posterior probability values ≥95% are indicated by thickened branches. Taxa referred to but not members of Annulatascaceae *sensu stricto* are indicated by shaded rectangles.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Podospora decipiens AY780073
Cercophora aquatica JN673036
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Sordariales
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Camarops ustulinoides DQ470941
Camarops polysperma JN673030
100
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Camarops lutea JN673032
Camarops plana JN673033
Camarops microspora AY083821
Camarops amorpha AY780054
Boliniales
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Menispora tortuosa AY544682
Chaetosphaeria ovoidea AF064641
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Chaetosphaeria innumera AY017375
Chaetosphaeria fuegiana EF063574
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Chaetosphaeria lateriphiala JN673039
Chaetosphaeriales
Aquaticola-like PE0159
Clohiesia corticola AF132329
Submersisphaeria rattanicola PE0292-1a
Ophioceras tenuisporum AY346295
Ceratosphaeria lampadophora AY761084
Magnaporthaceae
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Jobellisia guangdongensis JN936990
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Annulatascus-like A612-3b

Fig. 2

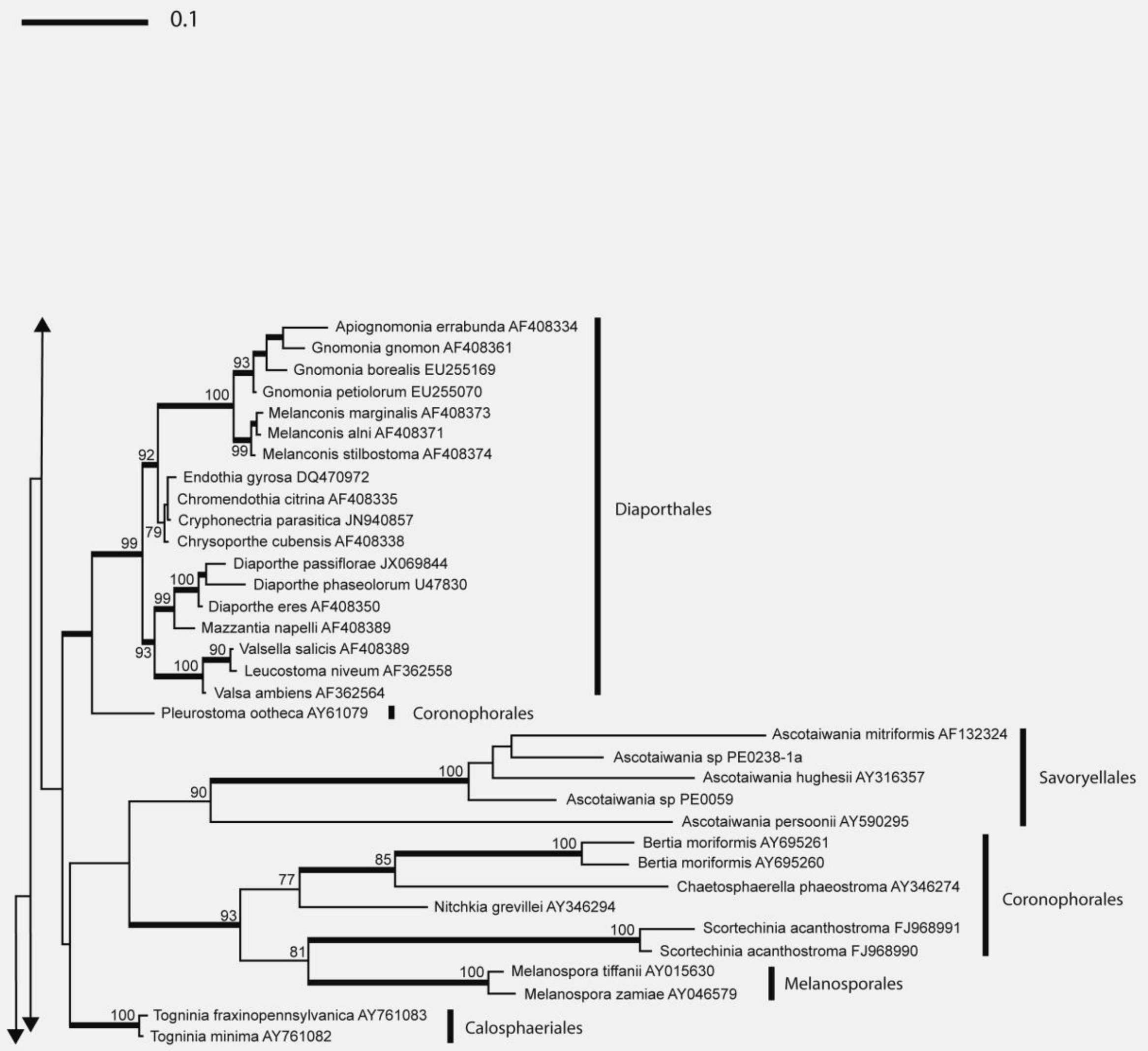
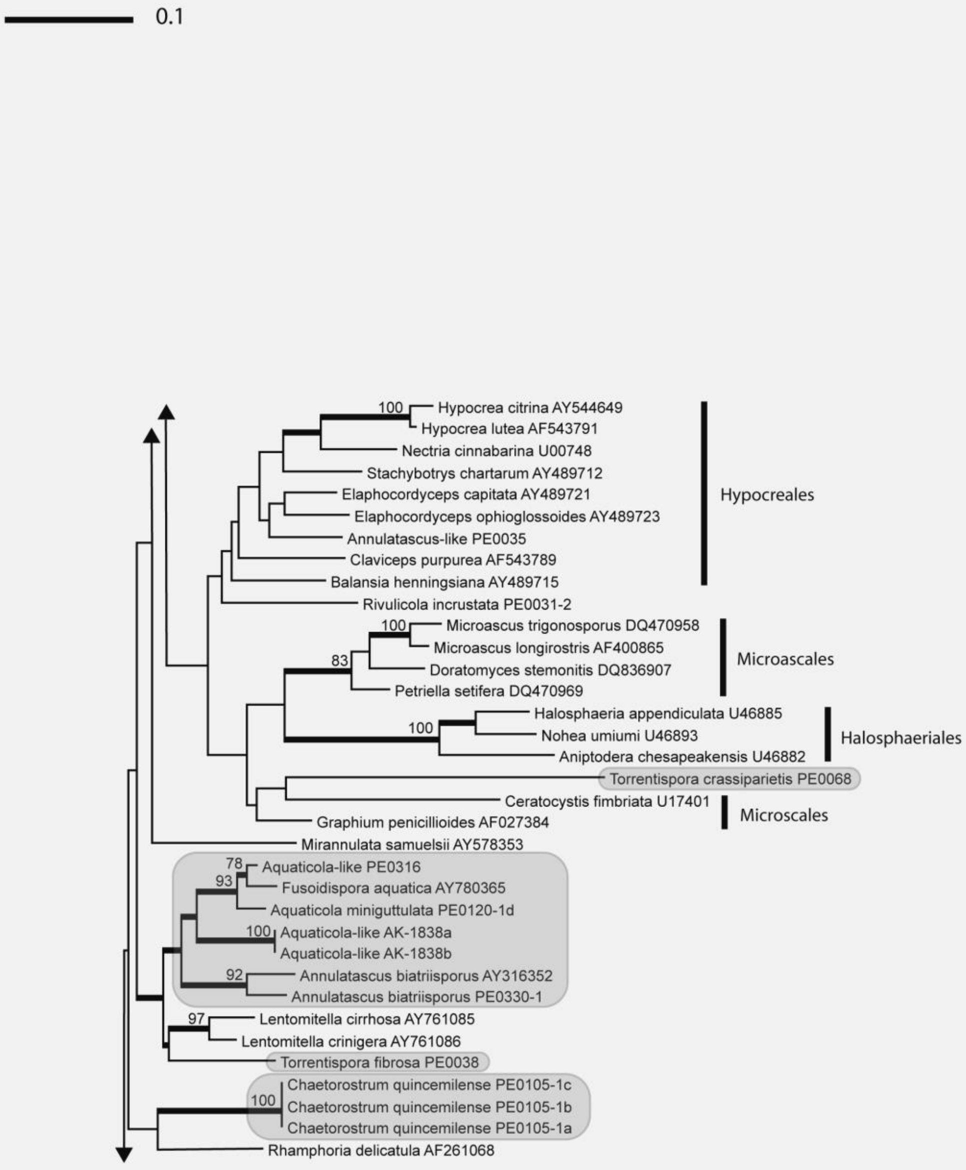


Fig. 2 continued.



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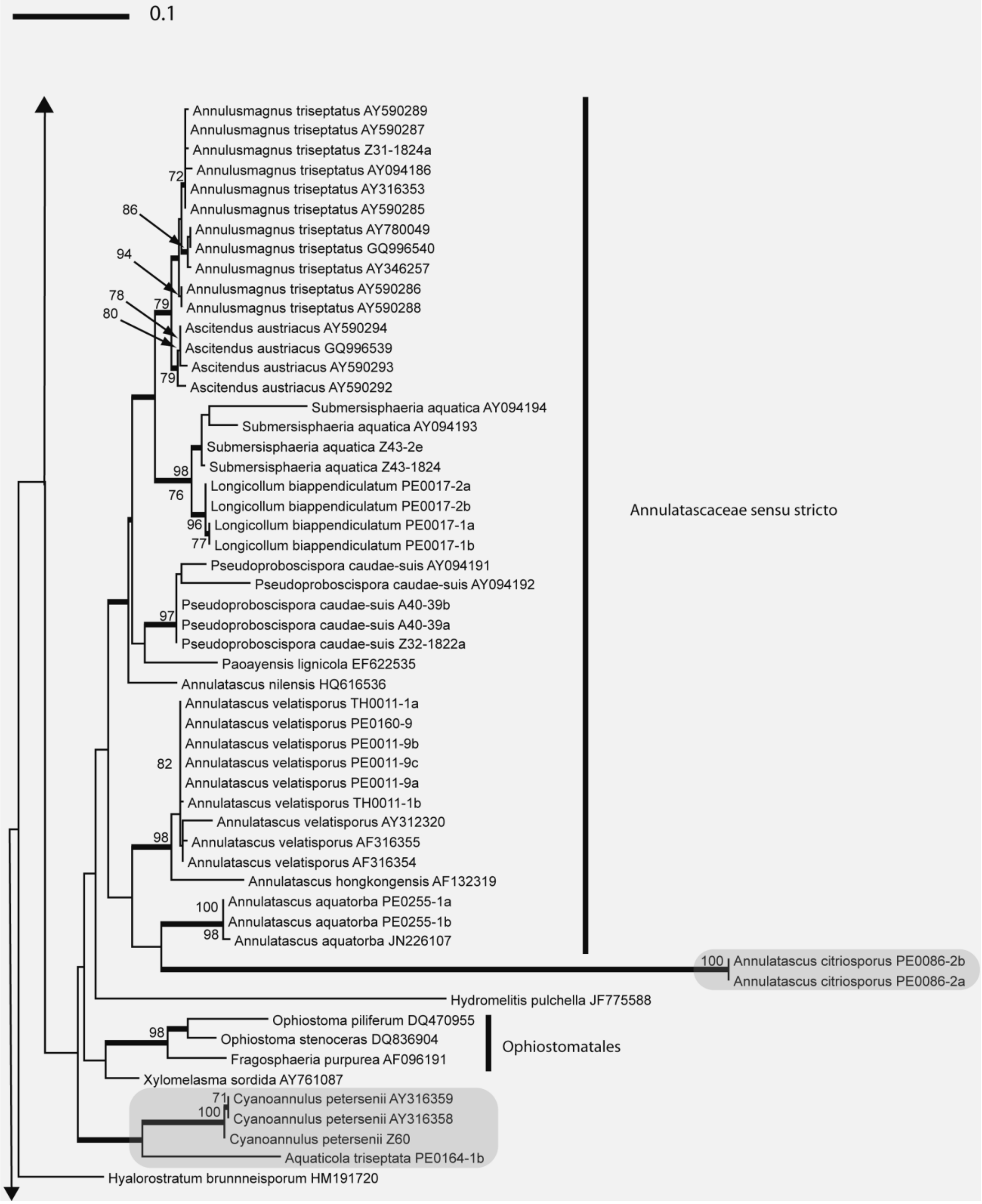

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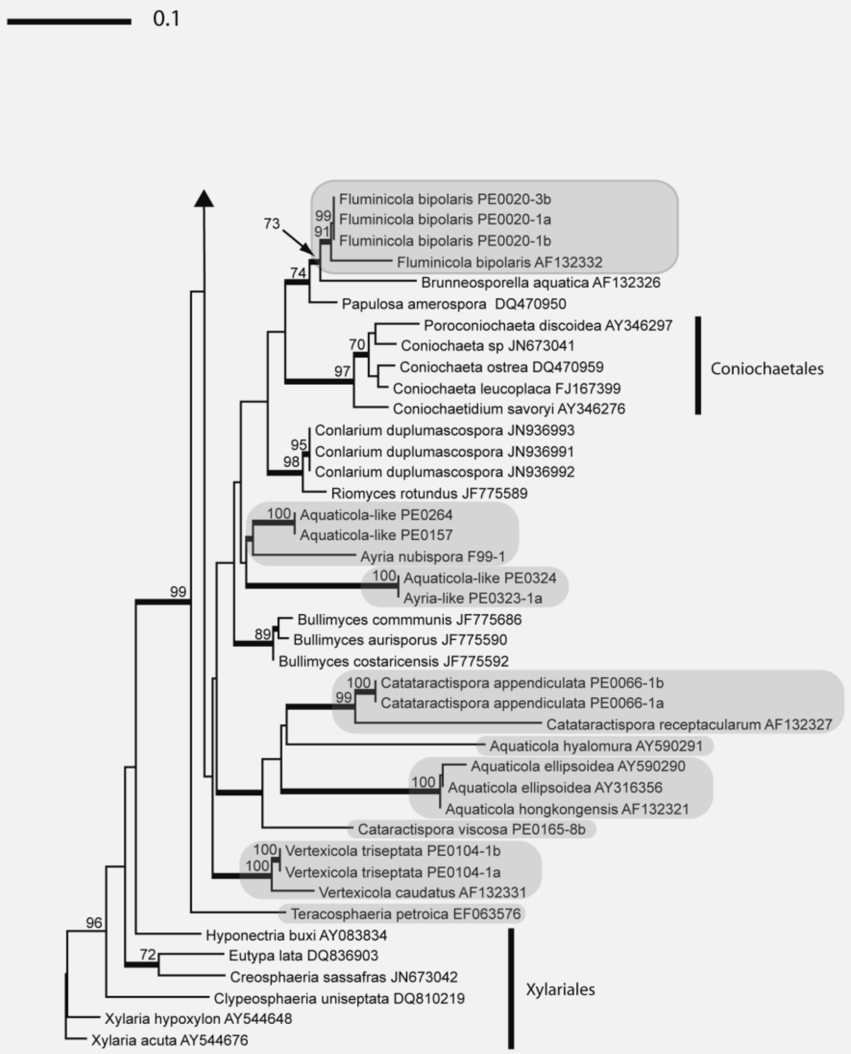
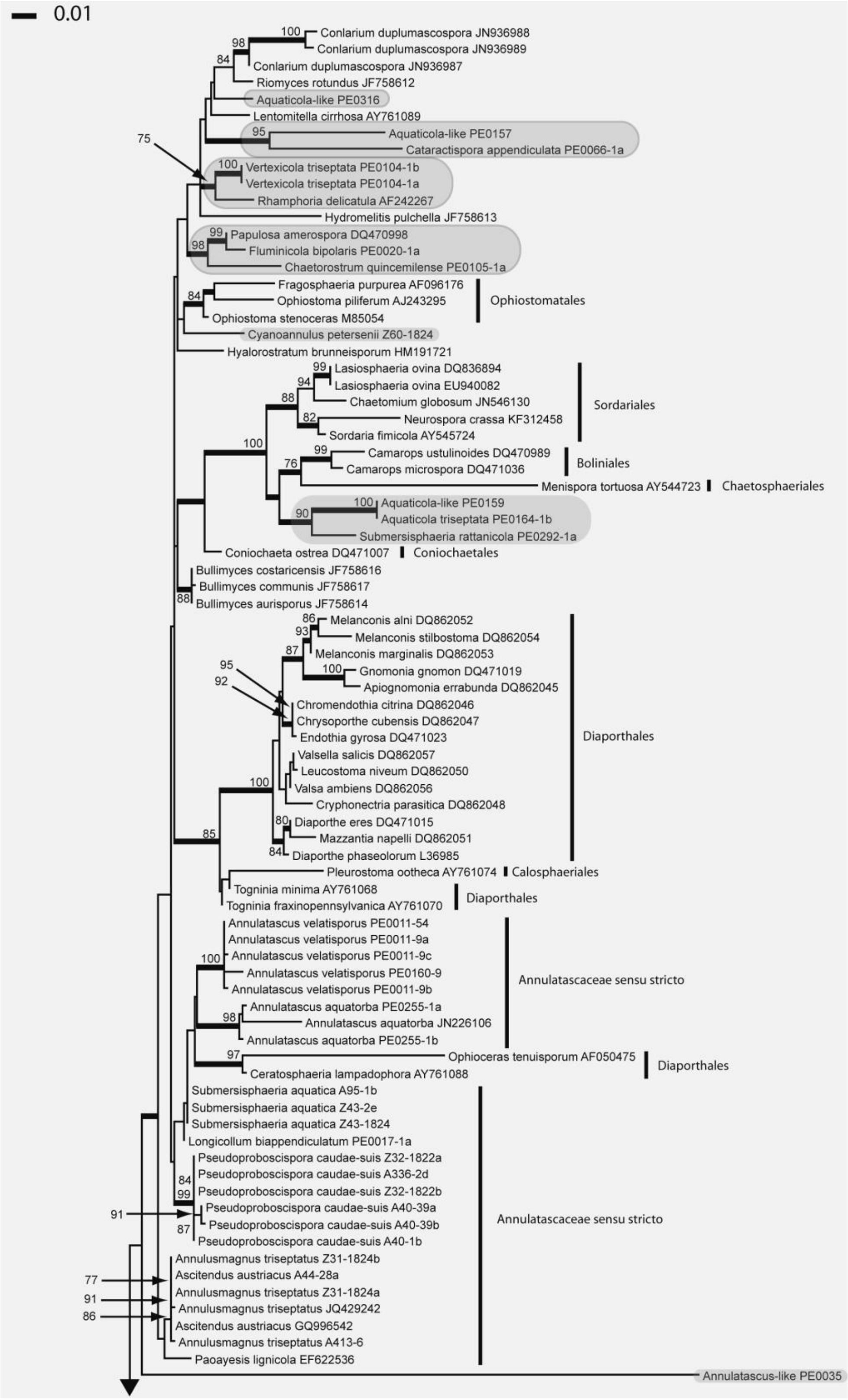


Fig. 2 continued.

Fig. 3. Phylogram of the most likely tree from 18S nrRNA gene analysis obtained with RAxML (-lnL = 10160.534721). Numbers ascribed to branches indicate ML bootstrap support values ≥70%, posterior probability support values ≥95% are indicated by thickened branches. Taxa referred to but not members of Annulatascaceae *sensu stricto* are indicated by shaded rounded rectangles.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Aquaticola-like PE0316
Lentomitella cirrhosa AY761089
Aquaticola-like PE0157
Cataractispora appendiculata PE0066-1a
Vertexicola triseptata PE0104-1b
Vertexicola triseptata PE0104-1a
Rhamphoria delicatula AF242267
Hydromelitis pulchella JF758613
Papulosa amerospora DQ470998
Fluminicola bipolaris PE0020-1a
Chaetorostrum quincemilense PE0105-1a
Fragosphaeria purpurea AF096176
Ophiostoma piliferum AJ243295
Ophiostoma stenoceras M85054
Ophiostomatales
Cyanoannulus petersenii Z60-1824
Hyalorostratum brunneisporum HM191721
Lasiosphaeria ovina DQ836894
Lasiosphaeria ovina EU940082
Chaetomium globosum JN546130
Neurospora crassa KF312458
Sordaria fimicola AY545724
Sordariales
Camarops ustulinoides DQ470989
Camarops microspora DQ471036
Boliniales
Menispora tortuosa AY544723
Chaetosphaeriales
Aquaticola-like PE0159
Aquaticola triseptata PE0164-1b
Submersisphaeria rattanicola PE0292-1a
Coniochaeta ostrea DQ471007
Coniochaetales
Bullimyces costaricensis JF758616
Bullimyces communis JF758617
Bullimyces aurisporus JF758614
Melanconis alni DQ862052
Melanconis stilbostoma DQ862054
Melanconis marginalis DQ862053
Gnomonia gnomon DQ471019
Apiognomonia errabunda DQ862045
Chromendothia citrina DQ862046
Chrysoporthe cubensis DQ862047
Endothia gyrosa DQ471023
Valsella salicis DQ862057
Leucostoma niveum DQ862050
Valsa ambiens DQ862056
Cryphonectria parasitica DQ862048
Diaporthe eres DQ471015
Mazzantia napelli DQ862051
Diaporthe phaseolorum L36985
Diaporthales
Pleurostoma ootheca AY761074
Calosphaeriales
Togninia minima AY761068
Togninia fraxinopennsylvanica AY761070
Diaporthales
Annulatascus velatisporus PE0011-54
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Annulatascus velatisporus PE0011-9c
Annulatascus velatisporus PE0160-9
Annulatascus velatisporus PE0011-9b
Annulatascus aquatorba PE0255-1a
Annulatascus aquatorba JN226106
Annulatascus aquatorba PE0255-1b
Annulatascaceae sensu stricto
Ophioceras tenuisporum AF050475
Ceratosphaeria lampadophora AY761088
Diaporthales
Submersisphaeria aquatica A95-1b
Submersisphaeria aquatica Z43-2e
Submersisphaeria aquatica Z43-1824
Longicollum biappendiculatum PE0017-1a
Pseudoproboscispora caudae-suis Z32-1822a
Pseudoproboscispora caudae-suis A336-2d
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Pseudoproboscispora caudae-suis A40-39a
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Pseudoproboscispora caudae-suis A40-1b
Annulatascaceae sensu stricto
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Ascitendus austriacus A44-28a
Annulusmagnus triseptatus Z31-1824a
Annulusmagnus triseptatus JQ429242
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Paoayesis lignicola EF622536
Annulatascus-like PE0035

Fig. 3.

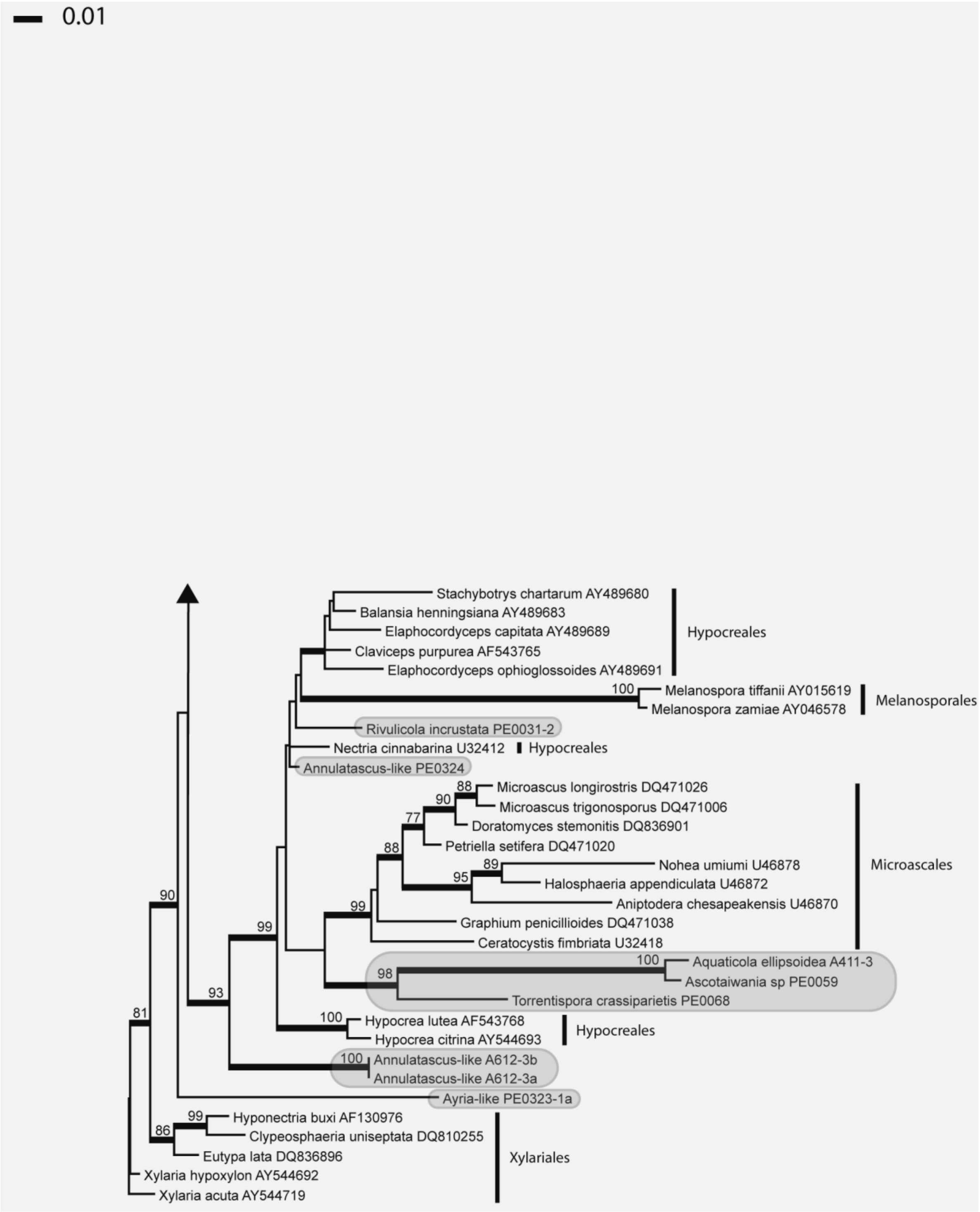
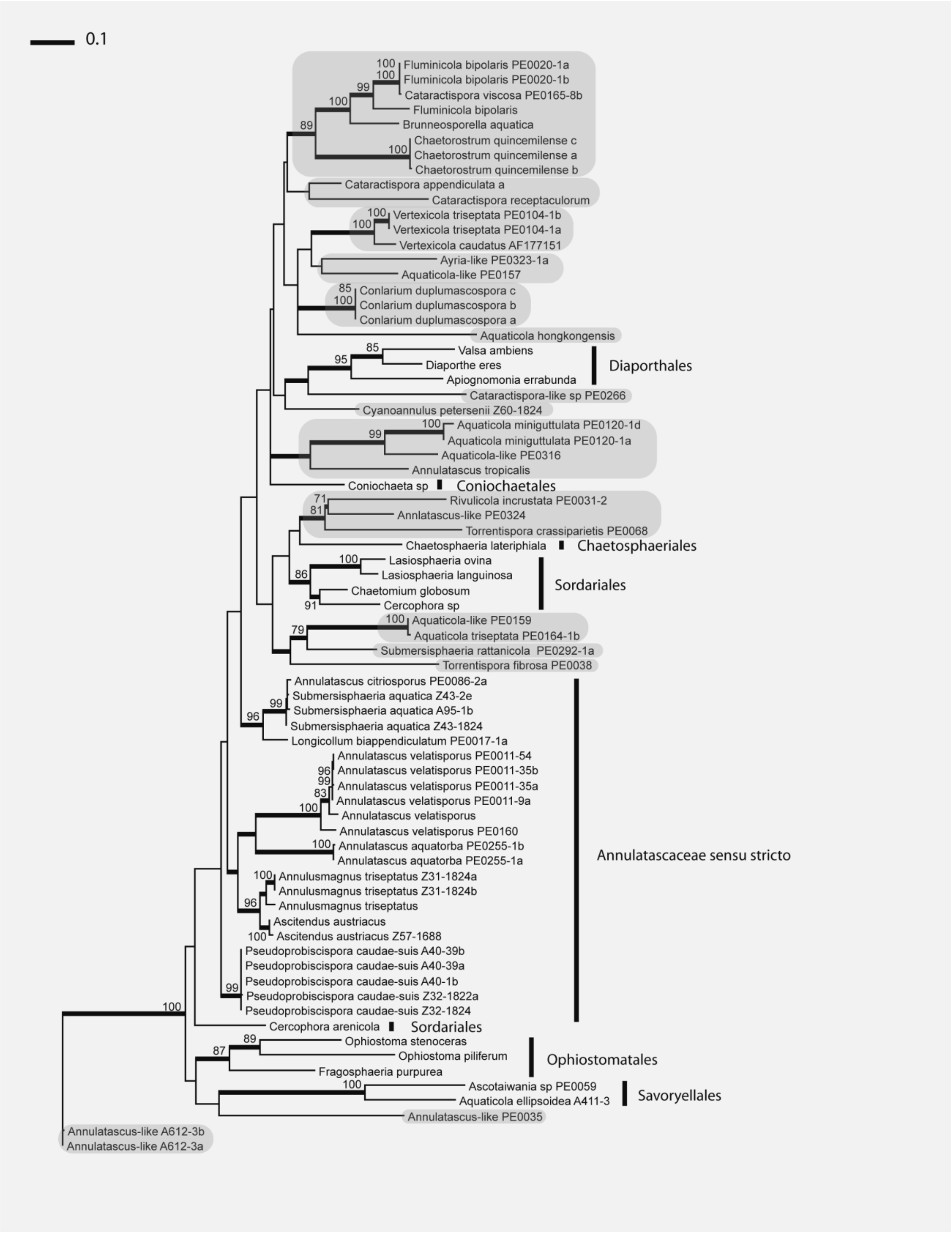


Fig. 3 continued.

Fig. 4. Phylogram of the most likely tree from ITS nrRNA gene analysis obtained with RAxML (-lnL = -13882.173909). Numbers ascribed to branches indicate ML bootstrap support values ≥70%, posterior probability support values ≥95% are indicated by thickened branches. Taxa referred to but not members of Annulatascaceae *sensu stricto* are indicated by shaded rounded rectangles.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99
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100
Fluminicola bipolaris
89
Brunneosporella aquatica
Chaetorostrum quincemilense c
100
Chaetorostrum quincemilense a
Chaetorostrum quincemilense b
Cataractispora appendiculata a
Cataractispora receptaculorum
100
Vertexicola triseptata PE0104-1b
100
Vertexicola triseptata PE0104-1a
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Ayria-like PE0323-1a
Aquaticola-like PE0157
85
Conlarium duplumascospora c
100
Conlarium duplumascospora b
Conlarium duplumascospora a
Aquaticola hongkongensis
85
Valsa ambiens
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Diaporthe eres
Apiognomonia errabunda
Diaporthales
Cataractispora-like sp PE0266
Cyanoannulus petersenii Z60-1824
100
Aquaticola miniguttulata PE0120-1d
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Aquaticola-like PE0316
Annulatascus tropicalis
Coniochaeta sp
Coniochaetales
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Rivulicola incrustata PE0031-2
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Annlatascus-like PE0324
Torrentispora crassiparietis PE0068
Chaetosphaeria lateriphiala
Chaetosphaeriales
100
Lasiosphaeria ovina
86
Lasiosphaeria languinosa
Chaetomium globosum
91
Cercophora sp
Sordariales
100
Aquaticola-like PE0159
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Aquaticola triseptata PE0164-1b
Submersisphaeria rattanicola PE0292-1a
Torrentispora fibrosa PE0038
Annulatascus citriosporus PE0086-2a
Submersisphaeria aquatica Z43-2e
99
Submersisphaeria aquatica A95-1b
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Submersisphaeria aquatica Z43-1824
Longicollum biappendiculatum PE0017-1a
Annulatascus velatisporus PE0011-54
96
Annulatascus velatisporus PE0011-35b
99
Annulatascus velatisporus PE0011-35a
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Annulatascus velatisporus PE0011-9a
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Annulatascus velatisporus
Annulatascus velatisporus PE0160
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Annulatascus aquatorba PE0255-1b
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Annulatascaceae sensu stricto
100
Annulusmagnus triseptatus Z31-1824a
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Annulusmagnus triseptatus
Ascitendus austriacus
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Pseudoprobiscispora caudae-suis A40-39b
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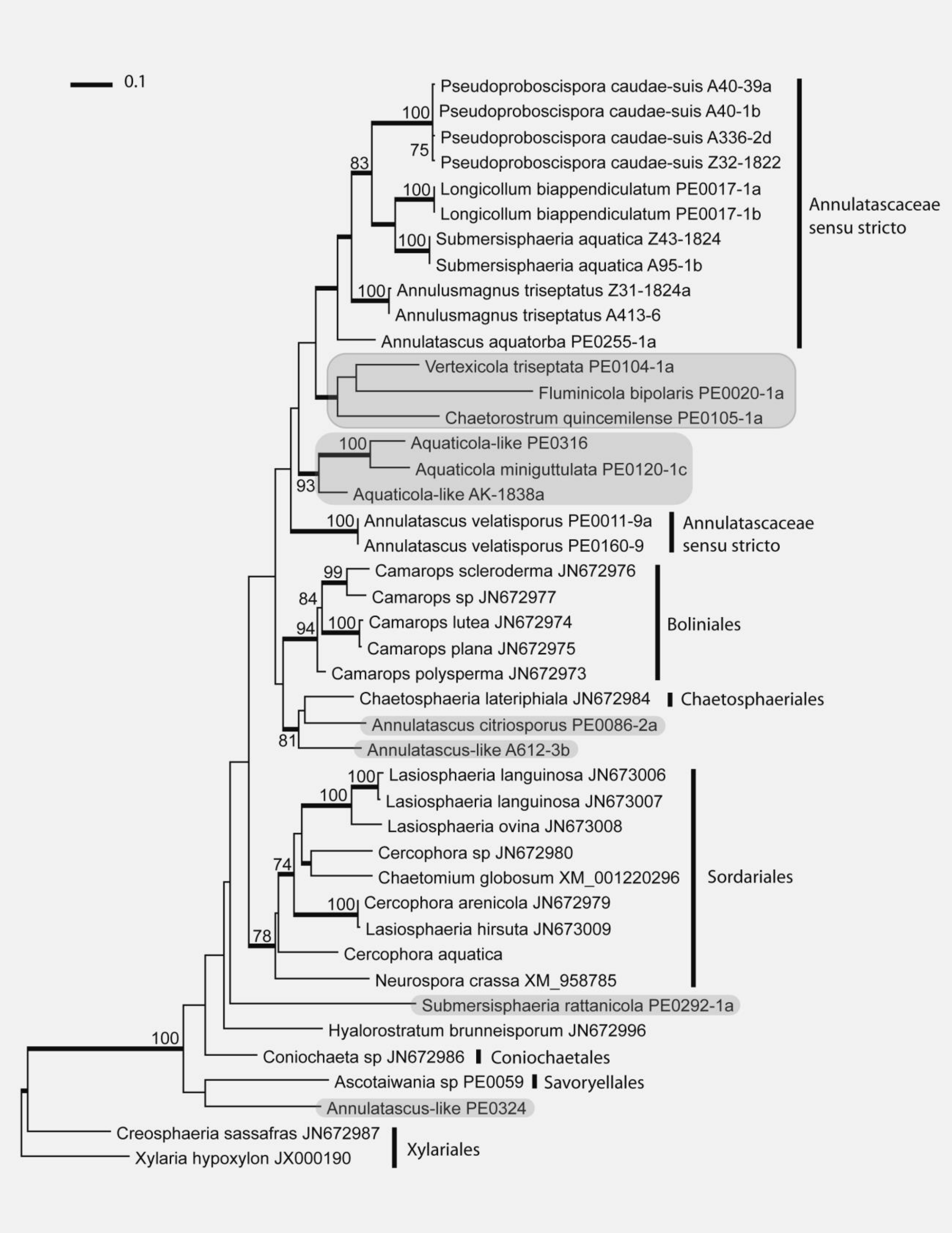
Pseudoprobiscispora caudae-suis Z32-1824
100
Cercophora arenicola
Sordariales
89
Ophiostoma stenoceras
87
Ophiostoma piliferum
Ophiostomatales
Fragosphaeria purpurea
100
Ascotaiwania sp PE0059
Aquaticola ellipsoidea A411-3
Savoryellales
Annulatascus-like PE0035
Annulatascus-like A612-3b
Annulatascus-like A612-3a

Fig. 4.

Fig. 5. Phylogram of the most likely tree from Mcm7 gene analysis obtained with RAxML (-lnL = -10190.729590). Numbers ascribed to branches indicate ML bootstrap support values ≥70%, posterior probability support values ≥95% are indicated by thickened branches. Taxa referred to but not members of Annulatascaceae *sensu stricto* are indicated by shaded rounded rectangles.



0.1
Pseudoproboscispora caudae-suis A40-39a
100
Pseudoproboscispora caudae-suis A40-1b
Pseudoproboscispora caudae-suis A336-2d
75
Pseudoproboscispora caudae-suis Z32-1822
83
100
Longicollum biappendiculatum PE0017-1a
Longicollum biappendiculatum PE0017-1b
100
Submersisphaeria aquatica Z43-1824
Submersisphaeria aquatica A95-1b
100
Annulusmagnus triseptatus Z31-1824a
Annulusmagnus triseptatus A413-6
Annulatascus aquatorba PE0255-1a
Annulatascaceae sensu stricto
Vertexicola triseptata PE0104-1a
Fluminicola bipolaris PE0020-1a
Chaetorostrum quincemilense PE0105-1a
100
Aquaticola-like PE0316
Aquaticola miniguttulata PE0120-1c
93
Aquaticola-like AK-1838a
100
Annulatascus velatisporus PE0011-9a
Annulatascus velatisporus PE0160-9
Annulatascaceae sensu stricto
99
Camarops scleroderma JN672976
84
Camarops sp JN672977
94
100
Camarops lutea JN672974
Camarops plana JN672975
Camarops polysperma JN672973
Boliniales
Chaetosphaeria lateriphiala JN672984
Chaetosphaeriales
Annulatascus citriosporus PE0086-2a
81
Annulatascus-like A612-3b
100
Lasiosphaeria languinosa JN673006
100
Lasiosphaeria languinosa JN673007
Lasiosphaeria ovina JN673008
Cercophora sp JN672980
74
Chaetomium globosum XM_001220296
100
Cercophora arenicola JN672979
Lasiosphaeria hirsuta JN673009
78
Cercophora aquatica
Neurospora crassa XM_958785
Sordariales
Submersisphaeria rattanicola PE0292-1a
Hyalorostratum brunneisporum JN672996
100
Coniochaeta sp JN672986
Coniochaetales
Ascotaiwania sp PE0059
Savoryellales
Annulatascus-like PE0324
Creosphaeria sassafras JN672987
Xylaria hypoxylon JX000190
Xylariales

Fig. 5.

Fig. 6. Phylogram of the most likely tree from combined 28S and 18S nrRNA gene analysis obtained with RAxML (-lnL = -40317.731427). Numbers ascribed to branches indicate ML bootstrap support values ≥70%, posterior probability support values ≥95% are indicated by thickened branches. Taxa referred to but not members of Annulatascaceae *sensu stricto* are indicated by shaded rounded rectangles.

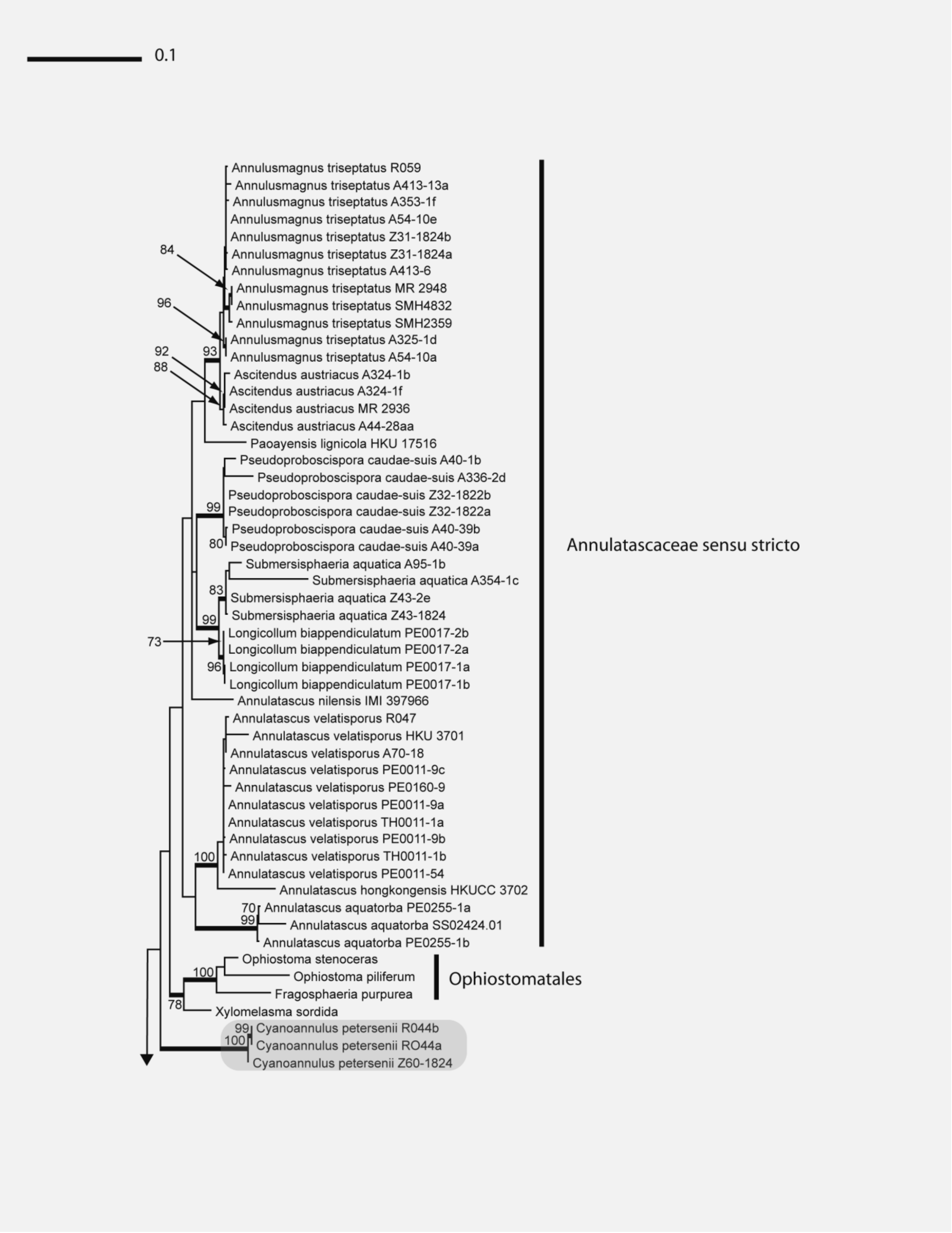

0.1
Annulusmagnus triseptatus R059
Annulusmagnus triseptatus A413-13a
Annulusmagnus triseptatus A353-1f
Annulusmagnus triseptatus A54-10e
Annulusmagnus triseptatus Z31-1824b
Annulusmagnus triseptatus Z31-1824a
Annulusmagnus triseptatus A413-6
Annulusmagnus triseptatus MR 2948
Annulusmagnus triseptatus SMH4832
Annulusmagnus triseptatus SMH2359
Annulusmagnus triseptatus A325-1d
Annulusmagnus triseptatus A54-10a
Ascitendus austriacus A324-1b
Ascitendus austriacus A324-1f
Ascitendus austriacus MR 2936
Ascitendus austriacus A44-28aa
Paoayensis lignicola HKU 17516
Pseudoproboscispora caudae-suis A40-1b
Pseudoproboscispora caudae-suis A336-2d
Pseudoproboscispora caudae-suis Z32-1822b
Pseudoproboscispora caudae-suis Z32-1822a
Pseudoproboscispora caudae-suis A40-39b
Pseudoproboscispora caudae-suis A40-39a
Submersisphaeria aquatica A95-1b
Submersisphaeria aquatica A354-1c
Submersisphaeria aquatica Z43-2e
Submersisphaeria aquatica Z43-1824
Longicollum biappendiculatum PE0017-2b
Longicollum biappendiculatum PE0017-2a
Longicollum biappendiculatum PE0017-1a
Longicollum biappendiculatum PE0017-1b
Annulatascus nilensis IMI 397966
Annulatascus velatisporus R047
Annulatascus velatisporus HKU 3701
Annulatascus velatisporus A70-18
Annulatascus velatisporus PE0011-9c
Annulatascus velatisporus PE0160-9
Annulatascus velatisporus PE0011-9a
Annulatascus velatisporus TH0011-1a
Annulatascus velatisporus PE0011-9b
Annulatascus velatisporus TH0011-1b
Annulatascus velatisporus PE0011-54
Annulatascus hongkongensis HKUCC 3702
Annulatascus aquatorba PE0255-1a
Annulatascus aquatorba SS02424.01
Annulatascus aquatorba PE0255-1b
Ophiostoma stenoceras
Ophiostoma piliferum
Fragosphaeria purpurea
Xylomelasma sordida
Cyanoannulus petersenii R044b
Cyanoannulus petersenii RO44a
Cyanoannulus petersenii Z60-1824
Annulatascaceae sensu stricto
Ophiostomatales
84
96
92
88
93
99
80
83
99
73
96
100
70
99
100
78
99
100

Fig. 6.

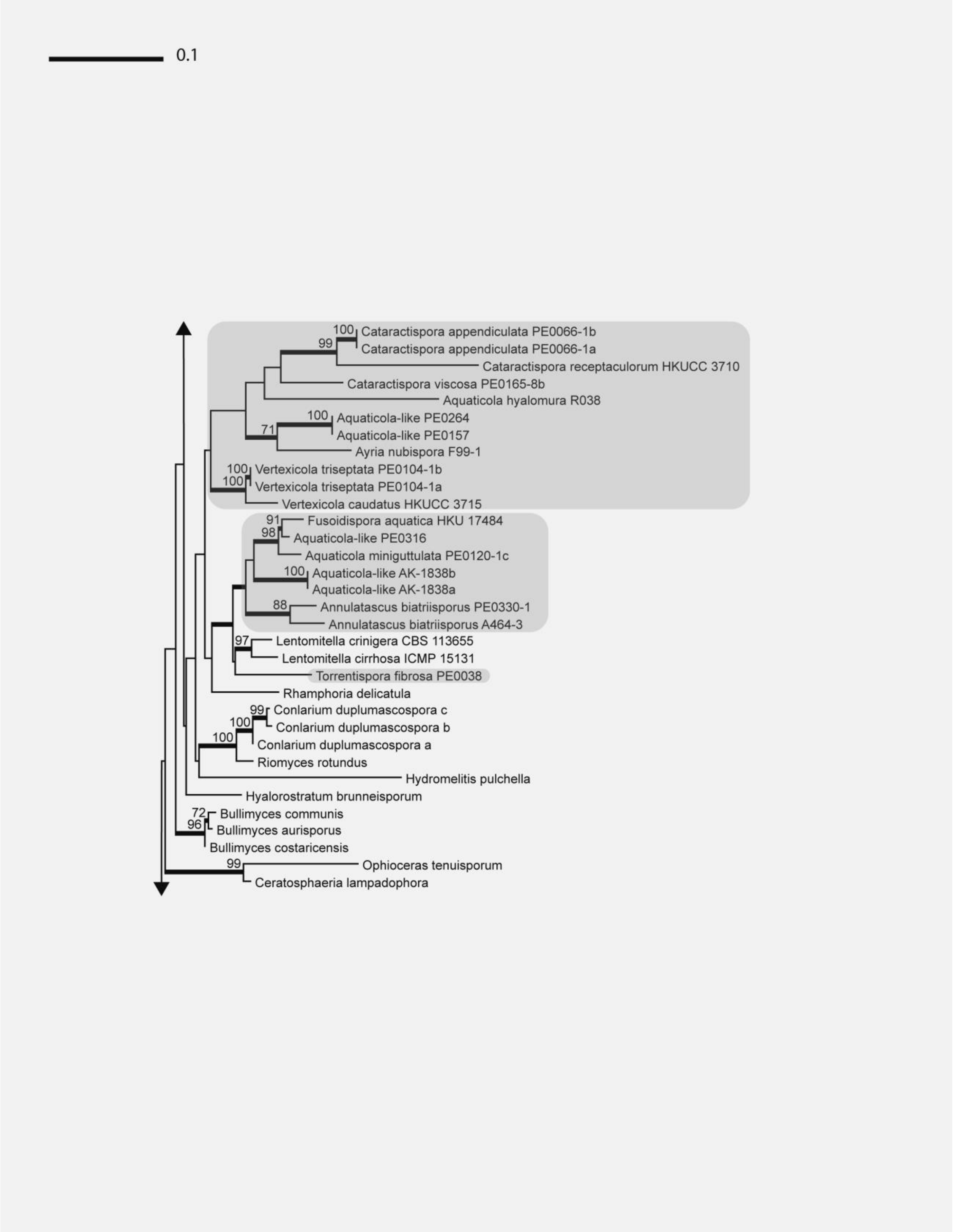


Fig. 6 continued.

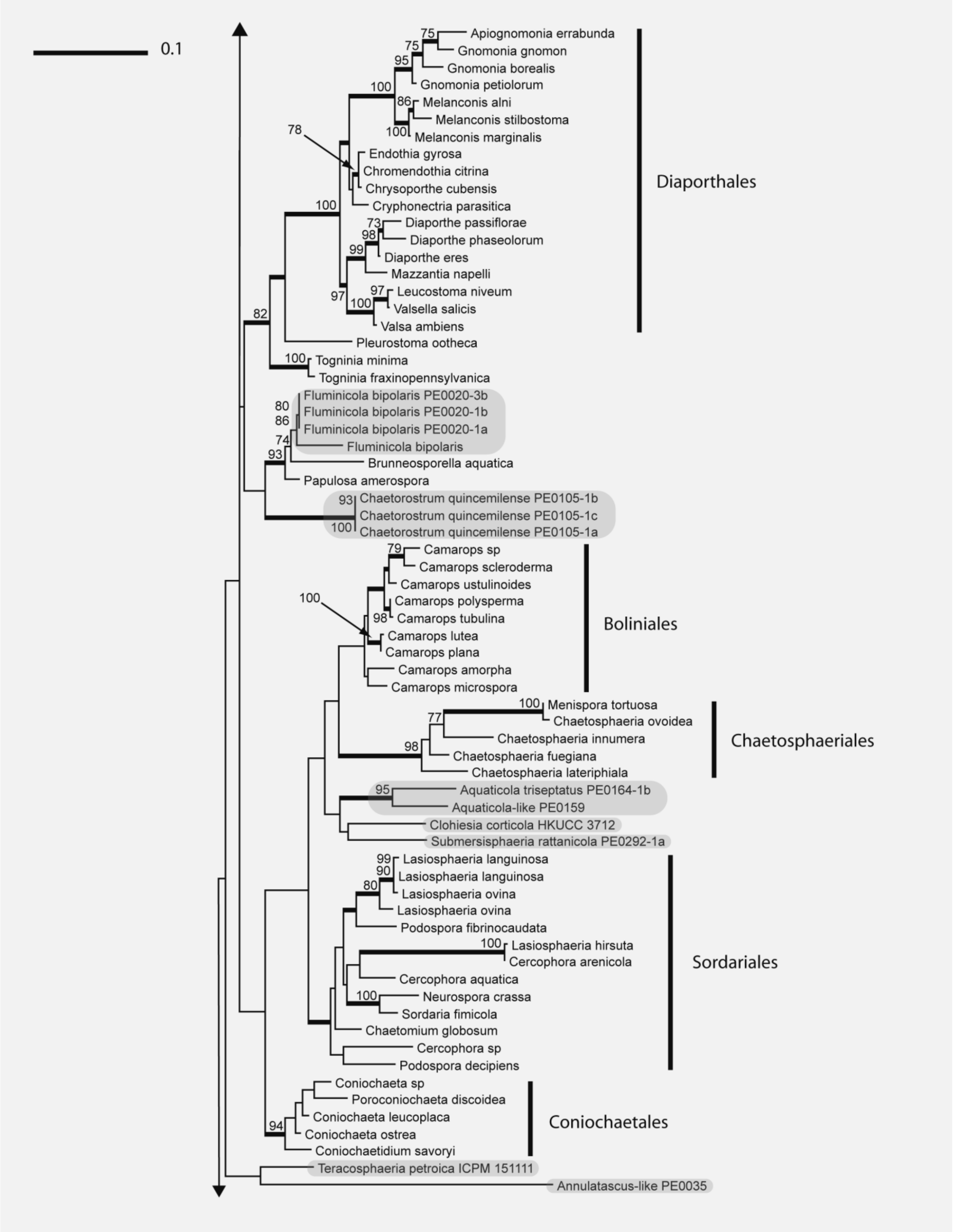


Fig. 6 continued.

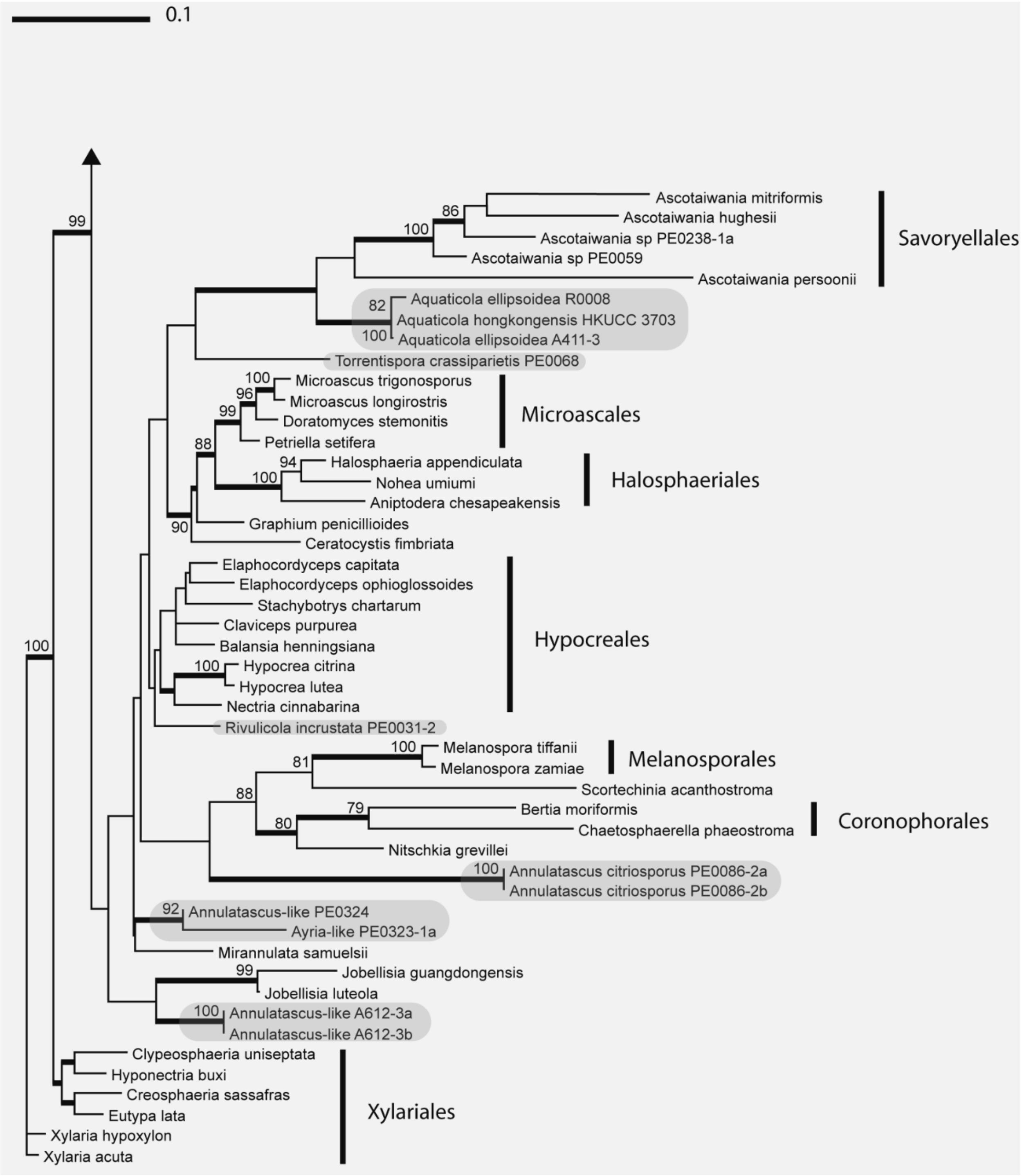


Fig. 6 continued.

Fig. 7. Phylogram of the most likely tree from combined 28S nrRNA, 18S nrRNA, and Mcm7 gene analysis obtained with RAxML (-lnL = -39137.212976). Numbers ascribed to branches indicate ML bootstrap support values ≥70%, posterior probability support values ≥95% are indicated by thickened branches. Taxa referred to but not members of Annulatascaceae *sensu stricto* are indicated by shaded rounded rectangles.

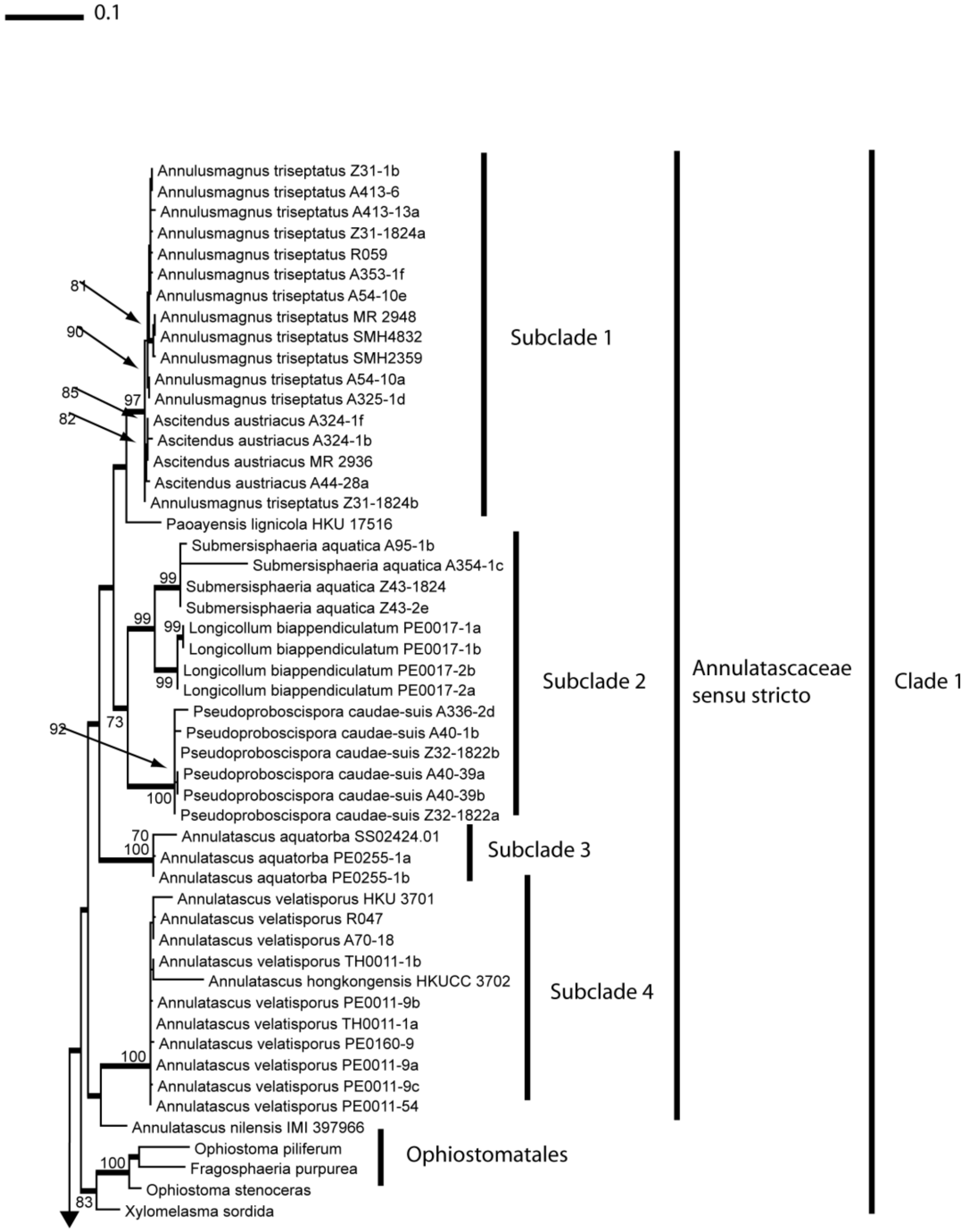

0.1
Annulusmagnus triseptatus Z31-1b
Annulusmagnus triseptatus A413-6
Annulusmagnus triseptatus A413-13a
Annulusmagnus triseptatus Z31-1824a
Annulusmagnus triseptatus R059
Annulusmagnus triseptatus A353-1f
81
Annulusmagnus triseptatus A54-10e
Annulusmagnus triseptatus MR 2948
90
Annulusmagnus triseptatus SMH4832
Annulusmagnus triseptatus SMH2359
Annulusmagnus triseptatus A54-10a
85
97
Annulusmagnus triseptatus A325-1d
82
Ascitendus austriacus A324-1f
Ascitendus austriacus A324-1b
Ascitendus austriacus MR 2936
Ascitendus austriacus A44-28a
Annulusmagnus triseptatus Z31-1824b
Paoayensis lignicola HKU 17516
Subclade 1
Submersisphaeria aquatica A95-1b
Submersisphaeria aquatica A354-1c
99
Submersisphaeria aquatica Z43-1824
Submersisphaeria aquatica Z43-2e
99
99
Longicollum biappendiculatum PE0017-1a
Longicollum biappendiculatum PE0017-1b
Longicollum biappendiculatum PE0017-2b
99
Longicollum biappendiculatum PE0017-2a
73
Pseudoproboscispora caudae-suis A336-2d
Pseudoproboscispora caudae-suis A40-1b
92
Pseudoproboscispora caudae-suis Z32-1822b
Pseudoproboscispora caudae-suis A40-39a
100
Pseudoproboscispora caudae-suis A40-39b
Pseudoproboscispora caudae-suis Z32-1822a
Subclade 2
70
Annulatascus aquatorba SS02424.01
100
Annulatascus aquatorba PE0255-1a
Annulatascus aquatorba PE0255-1b
Subclade 3
Annulatascus velatisporus HKU 3701
Annulatascus velatisporus R047
Annulatascus velatisporus A70-18
Annulatascus velatisporus TH0011-1b
Annulatascus hongkongensis HKUCC 3702
Annulatascus velatisporus PE0011-9b
Annulatascus velatisporus TH0011-1a
Annulatascus velatisporus PE0160-9
100
Annulatascus velatisporus PE0011-9a
Annulatascus velatisporus PE0011-9c
Annulatascus velatisporus PE0011-54
Subclade 4
Annulatascus nilensis IMI 397966
Annulatascaceae sensu stricto
Ophiostoma piliferum
100
Fragosphaeria purpurea
Ophiostoma stenoceras
83
Xylomelasma sordida
Ophiostomatales
Clade 1

Fig. 7.

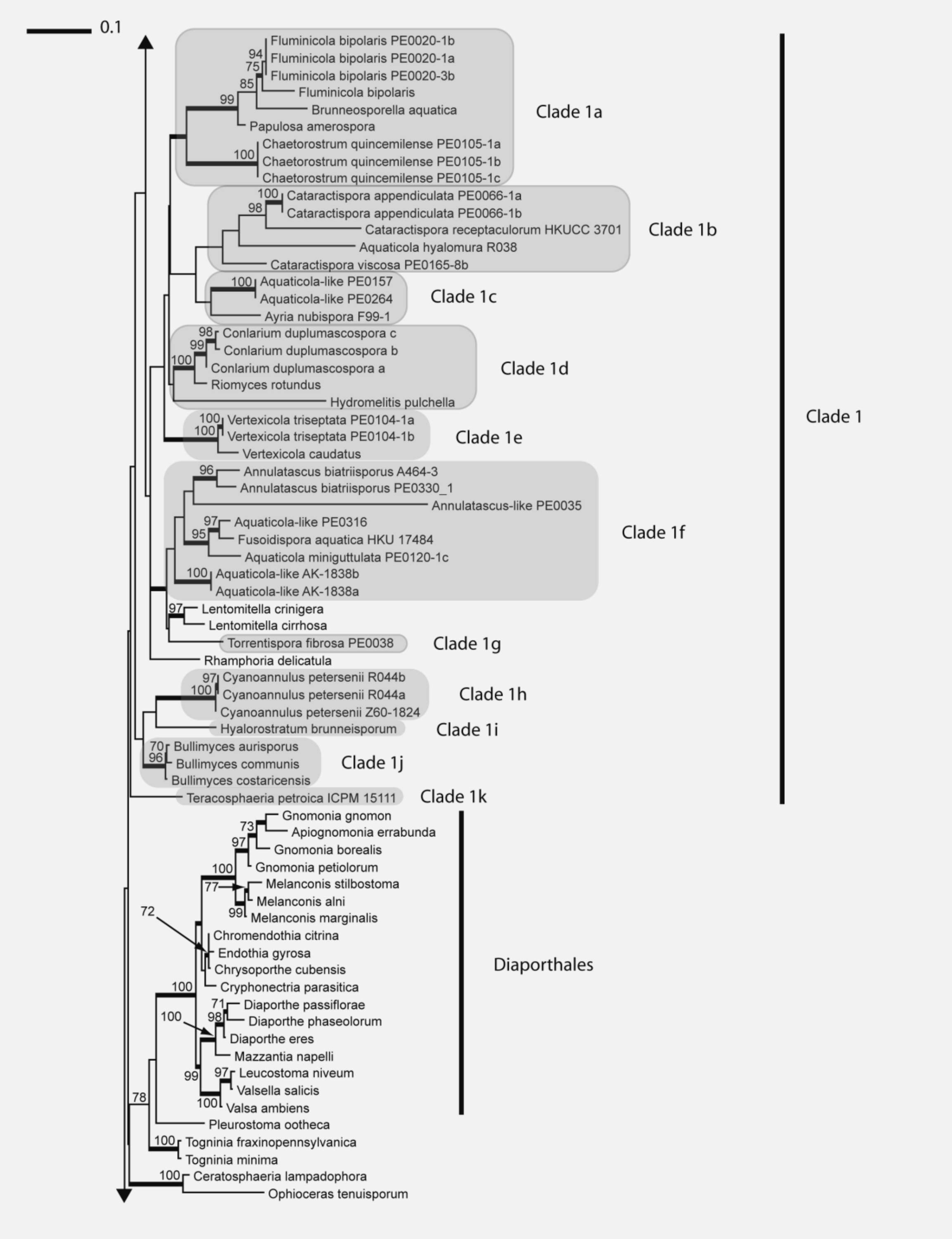


Fig. 7 continued.

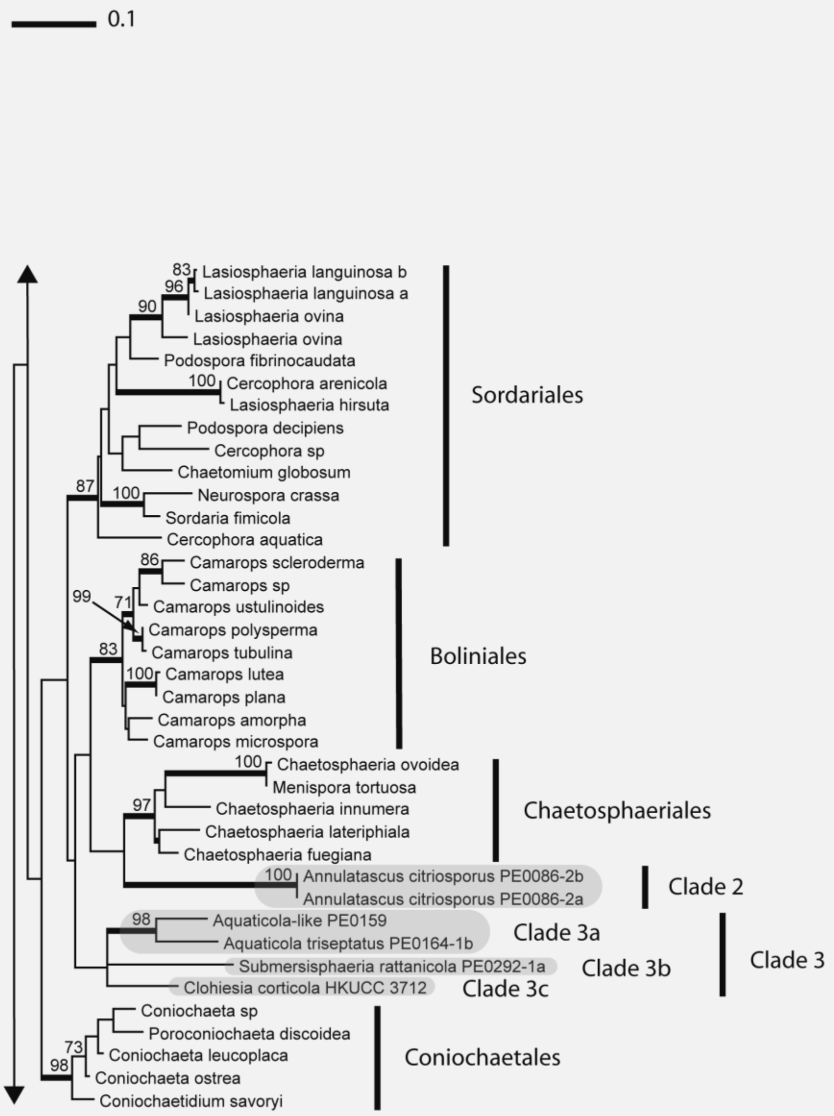


Fig. 7 continued.

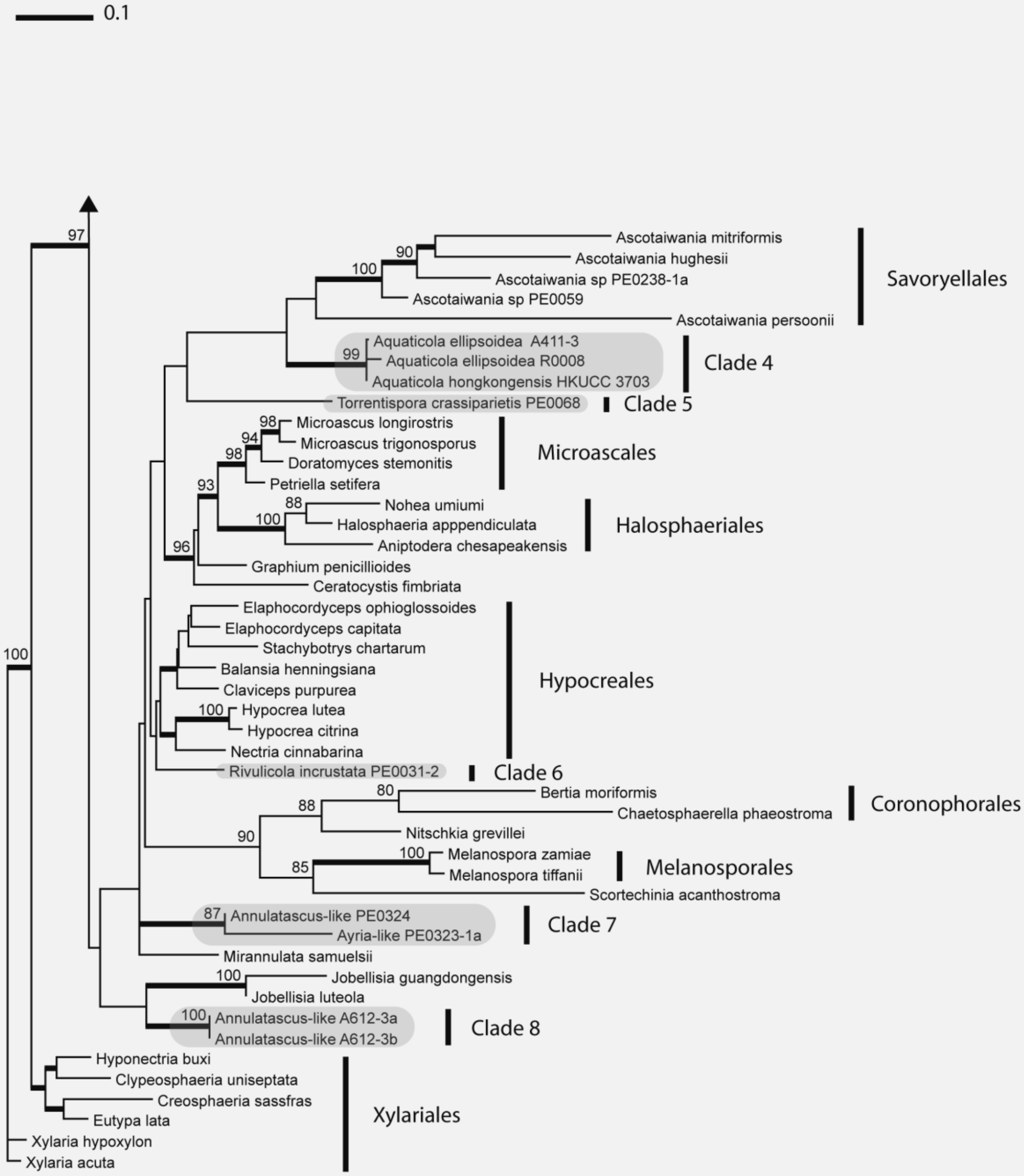


Fig. 7 continued.

MORPHOLOGY AND DATABASE:

Fifty-eight taxa were illustrated and measured either from the holotypes, fresh material, or both. In addition to this number are 14 newly discovered taxa with relatively large J- apical rings. A comprehensive database of Annulatascaceae species and their morphological characters was also compiled in Excel containing 71 taxa and 58 categories of morphological data. This database was used to write species descriptions, as a key to identify species, and to compare morphological characteristics among taxa. A synthesis of molecular and morphological data is presented in the discussion. This database is presented (in part) in Table 7 and is available in electronic form in full from the author by request, as the electronic version is far more utile.

CHAPTER 4

DISCUSSION

Annulatascaceae, as previously circumscribed, is a highly polyphyletic assemblage of taxa as evidenced in this study. An Annulatascaceae *sensu stricto* clade consisting of 10 taxa in seven genera was revealed in the three gene analyses with good Bayesian support (≥ 95 PP) though without maximum likelihood support (**Fig. 7**). The ML tree generated from three gene analyses is preferred as it is the most robust, incorporating the greatest number of molecular data. Four monotypic genera (*Annulusmagnus, Ascitendus, Paoayensis,* and *Longicollum)* and three genera with more than one species (*Annulatascus, Pseudoproboscispora* (only one species with sequence data), and *Submersisphaeria)* are included in the family. The nearest outgroup to the family is composed of members of Ophiostomatales. Maharachchikumbura et al. (2015) introduced the new order Annulatascales in the tentative subclass Diaporthomycetidae to accommodate Annulatascaceae in a higher level classification of the Sordariomycetes. Higher level taxonomic placement was one of the main questions to be addressed by this study, and while the ordinal name is appropriate Annulatascales needs revised circumscription and the subclass placement needs further refinement since Diaporthomycetidae as currently circumscribed is not widely recognized (Miller, Raja, and Shearer pers. comms.). The Annulatascaceae *sensu stricto* clade is thus the Annulatascales in the strict interpretation of the study of Maharachchikumbura et al. (2015), but also includes *L. biappendulatum* based on the work herein.

The most recently published detailed phylogenetic analysis of Annulatascaceae (Jones et al. 2004) will serve for comparison to the results of the combined three gene analyses of this study. Jones et al. (2014) mined GenBank for all available Annulatascaceae species with available 28S sequences. No new sequences were generated. That analysis divided the Annulatascaceae into an Annulatasceae *sensu stricto* clade and four Annulatascaceae *sensu lato* clades, although one clade (Clade II) could be construed as being composed of two separate lineages (Jones et al. 2014). Of note, the authors included

Annulatascus [sic] *apiculatus* in the Annulatascaceae *sensu stricto* clade, though the sequence is actually attributed to *A. aquatorba* (GenBank JN226107). No sequence data are available for *A. apiculatus*, as Barbosa et al. (2009) did not obtain a culture, and this species was not collected in this study. Thus the phylogenetic placement of *A. apiculatus* remains uncertain at this time.

Annulatascaceae *sensu stricto* is a well supported clade in multi-gene analyses in terms of Bayesian posterior probabilities (≥95 PP), but not Maximum Likelihood bootstrap values (**Fig. 7**). This clade corresponds to the Annulatascaceae *sensu* [sic] *lato* clade (= *sensu stricto*) (Clade I) of Jones et al. (2014) which contains the family type *A. velatisporus*, and with the exception that *Longicollum biappendiculatum* is included in this study. It has been argued that Bayesian support values are more robust, especially when dealing with short internode lengths compared to likelihood approaches which are subject to limitations in search strategies and the specific model of nucleotide evolution chosen (Alfaro et al. 2003). Within the Annulatascaceae *sensu stricto* clade of this study are four well supported subclades (**Fig. 7**).

Morphologically, the taxa in the *sensu stricto* clade share the following characteristics: brown to black predominantly coriaceous ascomata (with the exception of *Longicollum biappendiculatum* which has membranous ascomata); periphysate, cylindrical, mostly dark necks of variable lengths (with the exception of *Paoayensis lignicola* which has clustered ascomata with a common ostiole); hyaline tapering septate paraphyses, unitunicate cylindrical asci with mostly bipartite ascus apical rings (exceptions being *Paoayensis lignicola* with an evanescent ring, and *Pseudoproboscispora caudae-suis* with a flat apical ring with an apical pore); and uniseriate to overlapping uniseriate ascospores. The taxa differ in their ascospore morphologies with some being brown and some hyaline, their septation, and the presence or absence of gelatinous sheaths and/or appendages. Ascospore shapes are somewhat conserved as they range from fusoid to ellipsoidal.

Subclade 1 in Annulatascaceae *sensu stricto* consists of *Annulusmagnus triseptatus*, *Ascitendus austriacus*, and the dubious (from a morphological perspective) *Paoayensis lignicola* (**Fig. 7**). These taxa share the feature of straw colored to brown pigmented ascospores, although *Pa. lignicola* is distinctive in having extremely large ascospores with a germ slit and stromatic fruiting structures (Cabanela et al. 2007), features not found in other Annulatascaceae taxa. A fresh collection to confirm the molecular identity of *Pa. lignicola* should be obtained to corroborate its placement in Annulatascaceae *sensu stricto*. Removal of *Pa. lignicola* from three gene analysis leaves the Annulatascaceae *sensu stricto* clade intact and in the same order (results not shown). ITS and combined three gene analyses place *Asc. austriacus* and *An. triseptatus* as very closely related (**Fig. 4, Fig. 7**) and these taxa may need to be assimilated as congeneric. Morphologically, the species are distinguishable based on ascospore morphology. Both species have three septate ascospores but *Asc. austriacus* possesses ascospores that are smaller on average than those of *An. triseptatus* (14-27 × 4-9 µm ($\bar{x}$ = 20.5 × 6.5) vs. 16-37 × 5-10 µm ($\bar{x}$ = 26.5 × 7.5)). In addition, ascospores of *Asc. austriacus* are pale brown with central cells darker than end cells and have darkened septa, while those of *An. triseptatus* are hyaline to pale brown in their entirety without darkened septa.

Subclade 2 in Annulatascaceae *sensu stricto* consists of *Submersisphaeria aquatica*, *Longicollum biappendiculatum* and *Pseudoproboscispora caudae-suis* (**Fig. 7**). These taxa share the features of dark brown to black ascomata, well-defined periphysate necks, long tapering septate hyaline paraphyses, and a relatively large J- ascus apical ring, but they differ in ascospore morphology. *Longicollum biappendiculatum* possesses aseptate hyaline ascospores with bipolar, pad like appendages (Zelski et al. 2011a). *Pseudoproboscispora caudae-suis* possesses hyaline, 1-septate ascospores with coiled, thread-like appendages which unfurl to great length in water (**Fig. 52 F-H**). *Submersisphaeria aquatica* has brown 1–septate ascospores and lacks appendages or a sheath (**Fig. 57 H,I; Fig. 58 K,L**). This is a morphologically diverse clade with respect to ascospore morphology, which is subject to selective pressure (Shearer 2007), but the three taxa are linked by possessing dark brown to black, coriaceous ascomata with well-developed necks, asci with J- apical rings, and tapering septate paraphyses.

Subclade 3 of Annulatascaceae *sensu stricto* in combined three gene analyses consists of *Annulatascus aquatorba*, with both Asian and South American specimens included (**Fig. 7**). Though considered to be an *Annulatascus* species, it does not cluster with the type of the family, *A. velatisporus,* in three gene analyses. ITS analyses, however, do place the two species as sister taxa (**Fig. 4**), which may stem from the difficulty of aligning ITS sequences, which evolve more rapidly than the other genes evaluated in this study. *Annulatascus aquatorba*, which has brown pigmented ascospores and is quite similar to *Asc. austriacus*, does not resemble the taxa from the fourth clade in Annulatascaceae *sensu stricto* housing *A. velatisporus*, and all species in that clade having hyaline ascospores. Morphologically, this species most closely resembles *Asc. austriacus* and would not have been easily diagnosed as a separate taxon without molecular data. *Annulatascus aquatorba* has ascospores that are not constricted at the septa, which distinguishes it from *Asc. austriacus* whose ascospores are constricted at the septa.

Subclade 4 in Annulatascaceae *sensu stricto* in combined three gene analyses is composed of the type of the family, *A. velatisporus*, *A. hongkongensis* and *A. nilensis* (**Fig. 7**). All three taxa possess immersed to semi-immersed dark brown to black coriaceous perithecia of *textura angularis*. The peridium thickness of *A. velatisporus* (38–60 µm) is more than double that of the other two species. *Annulatascus hongkongensis* is morphologically quite similar to *A. velatisporus*, differing only in that its ascospores are consistently 3–septate and constricted at the septa. Only one sequence of *A. hongkongensis* (partial 28S) is available in GenBank and in single gene analyses, this species clusters within *A. velatisporus* specimens, although on a longer branch. Further molecular data from fresh collections, including a complete 28S sequence, would aid in determining if *A. hongkongensis* is conspecific with *A. velatisporus* or a separate taxon. *Annulatascus nilensis* also clusters in this clade in 28S and combined two and three gene analyses (**Fig. 2,6,7**). This taxon is the only *Annulatascus* species that possesses ascospores with more than five septa. It is anticipated that discovery of new species and sequencing of known species without molecular data will aid in refining the Annulatascaceae *sensu stricto* clade further. At present, Annulatascaceae *sensu stricto* (with the exception of *Paoayensis lignicola*) can be characterized by possessing dark brown to black ascomata with cylindrical periphysate necks, long tapering septate hyaline paraphyses, pedicellate 8-spored asci with J- ascus apical rings, and uniseriate to overlapping uniseriate ascospores.

Annulatascaceae *sensu lato* Clade 1 from molecular analyses is a large group of taxa that are somewhat related but not statistically supported as monophyletic by Bayesian or Maximum-likelihood values (**Fig. 7, Clade 1**). This large clade is on a branch nested between Diaporthales and Ophiostomatales and is composed of predominantly freshwater species. **Clade 1** forms a monophyletic group when Ophiostomatales and Annulatascaceae *sensu stricto* included, but is treated here based on its subclades (Clades 1a-1k) to facilitate the discussion. While the root of this lineage has no statistical support there are subclades within it that do have good Bayesian and likelihood support. Included in this group (**Fig. 7, Clade 1**) are the members of the Annulatascaceae *sensu lato* Clades II, IV, and V of Jones et al. (2014).

In the analyses presented herein, the species comprising Annulatascaceae *sensu lato* Clade II of Jones et al. (2014), *A. biatriisporus*, *Cyanoannulus petersenii*, and *Fusoidispora aquatica* are further separated from one another. These species are placed as follows: *A. biatriisporus* (**Fig. 7, Clade 1f**), *Cy. petersenii* (**Fig. 7, Clade 1h**), and *F. aquatica* (**Fig. 7, Clade 1f**). Morphologically, *A. biatriisporus* and *F. aquatica* share dark brown to black ascomata compared to *Cy. petersenii* which has reddish-brown ascomata. *Annulatascus biatriisporus* and *F. aquatica*, while in the same subclade, are distinguishable by ascospore morphology. *Annulatascus biatriisporus* has ascospores that are aseptate, fusiform, with bipolar apical chambers and a thin irregular sheath, while *F. aquatica* has 0–5 septate, fusoid to sickle-shaped ascospores and no sheath. Since *A. biatriisporus* is not related to *Annulatascus*, a new genus and combination, *Ascuslongus biatriisporus*, is proposed (see Nomenclatural Changes).

Taxa in Clade IV of Jones et al. (2014), *Aquaticola ellipsoidea*, *Aq. hongkongensis*, *Aq. hyalomura*, and *Cataractispora* [sic] *recepticuli*, are separated in the analyses presented here. *Cataractispora receptaculorum* and *Aq. hyalomura* belong in a clade with *C. viscosa* and *C. appendiculata* (**Fig. 7, Clade 1b**). Species in **Clade 1b** all possess ascospores bipolar appendages with the exception of *Aq. hyalomura*. The provenance of the *Aq. hyalomura* sequence (R038) used in this study has been a concern. The sequence does not come from the type, and *Aq. hyalomura* is not morphologically similar to *Cataractispora* species as it has a hyaline membranous ascomata and ascospores without bipolar appendages. *Cataractispora aquatica*, however, has been reported from the GSMNP (Raja pers. comm., Zelski unpub.) where the isolate from which putative *Aq. hyalomura* sequence was derived. Thus it is a possibility that this sequence was derived from *C. aquatica* and not *Aq. hyalomura*, which would strengthen the case for a monophyletic *Cataractispora*. *Aquaticola ellipsoidea* and *Aq. hongkongensis*, the two other *Aquaticola* species assessed molecularly, are placed together with strong Bayesian and Maximum-likelihood support in **Clade 4** (**Fig. 7, Clade 4**), removed from the type of the genus, *Aq. hyalomura*. Molecular data from fresh material collected at the type location is desirable to address this issue of polyphyly.

Annulatascaceae *sensu lato* Clade V of Jones et al. (2014), consisting of *Conlarium duplumascospora* and *Riomyces rotundus* is reproduced in this study with *Hydromelitis pulchella* as a statistically unsupported sister taxon (**Fig. 7, Clade 1d**). Ferrer et al. (2012) noted that *Riomyces* and *Hydromelitis* could be related to Annulatascaceae *sensu lato* but were unable to fully evaluate this due to lack of SSU data. Liu et al. (2012) placed *Conlarium* in Annulatascaceae *sensu lato* based on molecular analysis. The sterile cells in the hamathecium of *Riomyces* consist of large irregular cells, while those of *Conlarium* and *Hydromelitis* contain filamentous paraphyses. Asci of *Riomyces* and *Hydromelitis* have uniseriate ascospores and lack apical pores or other structures while *Conlarium* has biseriate ascospores and a prominent bipartite ascus apical ring. *Riomyces* and *Conlarium* are highly supported as sister taxa even though they differ morphologically and this may be an artifact of similarity of sequences and/or incomplete taxon sampling (or yet to be discovered taxa) in this region of the Sordariomycetes tree. *Hydromelitis* is not supported by Bayesian or Likelihood values and thus is a suspect member of **Clade 1d**.

Aside from the Annulatascaceae *sensu lato* clades of Jones et al. (2014) mentioned above, **Clade 1** from combined three gene analyses in this study includes numerous taxa not evaluated in that study (one exception being *Fluminicola* [sic] *coronata* (= *F. bipolaris*) which was included but not assigned to an Annulatasceae *sensu lato* clade).

Annulatascaceae **Clade 1a** contains *F. bipolaris* in a Bayesian and Maximum-likelihood supported clade together with *Brunneosporella aquatica* and *Papulosa amerospora* sister to *Chaetorostrum quincemilense* with Bayesian but not Maximum-likelihood support (**Fig. 7, Clade 1a**). *Chaetorostrum quincemilense*, originally described as an Annulatascaceae species (Zelski et al. 2011b), is placed here and is thus removed from the Annulatascaceae *sensu stricto* clade. Morphologically, the combination of the presence of setae on the necks, versicolored ascospores, and the production of a *Taeniolella*-like anamorph in culture and on natural substrates distinguishes this taxon from other Annulatascaceae members and thus from other members of **Clade 1a**. The presence of a large J- bipartite apical ring combined with the presence of prominent dark setae on the necks of perithecia is a feature shared with *Aqualignicola* spp., which have no available molecular data. The seta of *Aqualignicola* spp., however, extend to the ascomata and are not restricted to the distal portion of the neck as in *Ch. quincemilense*. The phylogenetic relationship of *Ch. quincemilense* to *Aqualignicola* spp. and species with setose necks still needs to be evaluated molecularly. It should be noted that *Ch. quincemilense* is unrelated to other species in this study that possess setae, such as *Chaetomium globosum*, *Coniochaeta*, and *Lasiosphaeria* spp. which suggests that this character alone is not phylogenetically informative.

Fluminicola bipolaris is sister to *B. Aquatica*, though with weak likelihood support and no Bayesian support. Ascomata of these species are similar in their coloration (dark brown to black) and shape (globose to subglobose) but differ in that those of *F. bipolaris* are smaller (250-275 µm wide × 175-188 µm high vs. 75-177 µm wide × 39-80 high) and coriaceous vs. membranous. In addition the ascospores of *F. bipolaris* are hyaline while those of *B. aquatica* are brown. *F. bipolaris* also differs from *Pap. amerospora* which has distinctively shield-like ascomata (Winka and Eriksson 2000). At present *F. bipolaris* as well as *Br. aquatica* should be considered Sordariomycetes *incertae sedis*, while *Pap. amerospora* is the type species of the monotypic family Papulosaceae. Morphologically, *B. aquatica*, *C. quincemilense*, and *F. bipolaris* share globose brown to black ascomata, periphysate cylindrical necks, cylindrical, pedicellate asci with J- discoid (*B. aquatica*) or bipartite apical rings, and uniseriate to overlapping uniseriate ascospores with one or more septa that are slightly constricted. *Papulosa amerospora* shares dark brown ascomata, periphysate cylindrical neck, cylindrical asci with an apical ring with the other member of the clade. However the ascus apical ring of *P. amerospora* is J+, ascospores are one-celled, and the species is found in salt marshes rather than in fresh water. Though not addressed in this study as it was focused on taxa with J- ascus apical rings, the difference in staining (i.e. chemical makeup of the ring) may be informative in relation to the terrestrial origins of aquatic fungal taxa and their adaptation to freshwater or saltwater (see Vijaykrishna et al. 2006).

Annulatascaceae *sensu lato* **Clade 1b** is composed of the *Cataractispora* species *C. appendiculata*, *C. receptaculorum*, and *C. viscosa*, as well as *Aquaticola hyalomura* (**Fig. 7, Clade 1b**). Morphologically, *Aq. hyalomura* does not agree with the *Cataractispora* species in this clade, and it may be that the GenBank sequence represents *C. aquatica* (found in the same location in GSMNP) rather than *Aq. hyalomura*. The *Cataractispora* species in this clade share the common features of coriaceous immersed to semi-immersed dark brown to black ascomata, periphysate necks, unitunicate cylindrical asci with long tapering pedicels and bipartite ascus apical rings, and hyaline fusiform or ellipsoidal ascospores of varying septation and having bipolar appendages. This suite of features unify the genus and thus is utile in delimiting the genus. An exception to this rule may be the unrelated *Conlarium duplumascospora*, which shares all of these features except that the ascus base is truncate and not a long tapering pedicel as in *Cataractispora* spp. Therefore, the morphology of the pedicel and its attachment to the hamathecium is an important character to consider when evaluating ascomycetes with a limited number of morphological characters. If the sequence of *A. hyalomura* represents *C. aquatica*, then that taxon would agree with the above morphological characteristics reported in the protologue and the *Cataractispora* clade would be monophyletic. If the sequence is indeed that of *Aq. hyalomura,* then this taxon differs morphologically from *Cataractispora* species in that the ascomata are hyaline, asci and ascus apical rings are considerably smaller, and the ascospores lack appendages. This would lead to a nomenclatural problem for this clade as both genera were established at the same time (Ho et al. 1999, Hyde et al. 1999), and neither of the types of the genera have sequences from type locations. Either *Aq. hyalomura* would be required to be renamed as *Cataractispora hyalomura* comb. nov., or the *Cataractispora* species would have to change to *Aquaticola* spp. Vouchered sequences from the type locations are critical to resolve this problem.

Sister to the aforementioned Annulatascaceae *sensu lato* **Clade 1b** is a statistically unsupported subclade including two new *Aquaticola*-like taxa (PE0157, PE0264) and *Ayria nubispora* (**Fig. 7, Clade 1c**). Morphologically the *Aquaticola*-like taxa are similar, but differ from *Ay. nubispora*. PE0157 and PE0264 have hyaline ascomata (although those of PE0157 are superficial rather than immersed), hyaline periphysate necks, small cigar shaped asci with small J- ascus apical rings, and hyaline ellipsoidal one-celled ascospores without sheaths or appendages. The two new *Aquaticola*-like species lack ascospore appendages and have considerably smaller ascospores compared to *Ay. nubispora*. *Ayria nubispora* differs in all these features save for the presence of hyaline ascospores. *Ayria nubispora* possesses ascospores with long uncoiling bipolar ascospore appendages similar to those of *Pseudoproboscispora* species, although it differs in ascomal morphology in having light colored membranous ascomata rather than the brown to black colored ascomata in *Pseudoproboscispora* species. **Clade 1c** requires further analysis as PE0157 and PE0264 are morphologically similar to *Aq. hyalomura* and comparison to molecular evidence from a specimen of *Aq. hyalomura* from the type location is desirable as the sequence used in this study is suspect (see above).

Annulatascaceae *sensu lato* **Clade 1d** is composed of the freshwater fungi *Conlarium duplumascospora*, *Hydromelitis pulchella*, and *Riomyces rotundus* (**Fig. 7, Clade 1d**). Morphologically, *C. duplumascospora* and *R. rotundus* share some morphological characters, but differ markedly from *H. pulchella* which is on a long branch. Of these three species, only *C. duplumascospora* has asci with apical rings. Other morphological features differ among the taxa. **Clade 1d** of this study was discussed fully in terms of morphology and molecular relationships above in relation to Annulatascaceae *sensu lato* Clade V of Jones et al. (2014).

Annulatascaceae *sensu lato* **Clade 1e** is comprised of *Vertexicola caudatus* and the new species from Perú, *V. triseptata* (PE0104) in combined three gene analyses in this study (**Fig. 7, Clade 1e**). These species agree well morphologically but differ in measurements of characters, ascospore septation, and the lack of a spike-like ascus tail in *V. triseptata*. The ascospores of *V. triseptata* are consistently 3-septate rather than 5-septate as in *V. caudatus*. The spike-like tail of *V. caudatus* is a feature is shared with *Ch. quincemilense*, although most other morphological features differ between these species, (e.g. *Ch. quincemilense* has setose necks and pigmented ascospores). The spike-like tail may be an artifact of separating the ascus from the base of the hamathecium during dissection and thus may not be a phylogenetically informative trait. *Vertexicola caudatus* (= *V. confusa* in GenBank) was not included in the study by Jones et al. (2004) and thus cannot be compared to that study. However, Raja et al. (2003) included the sequence in their evaluation of *Cyanoannulus petersenii*, and although it clustered in their

Annulatascaceae clade, the branch had no support. This is the first study to show *Vertexicola* is not a member of Annulatascaceae *sensu stricto*.

Annulatascaceae *sensu lato* **Clade 1f** was discussed above in relation to the Annulatascaceae *sensu lato* clades II and IV of Jones et al. (2014). This clade has Bayesian, though not Maximum-likelihood support. *Annulatascus biatriisporus*, a previously undescribed species (PE0035), *Aquaticola miniguttulata*, two new *Aquaticola*-like species (AK1838, PE0316), *Fusoidispora aquatica*, *Lentomitella* species, and *Torrentispora fibrosa* are included. Morphologically these taxa all share dark brown to black ascomata, and pedicellate asci with a J- refractive ascus apical ring. However, ascomal architecture, neck morphology, ascus apical ring morphology, and ascospore morphology vary among the taxa. These species all possess hyaline ascospores, although the ascospore shapes, ascospore wall thickness, and presence or absence of gelatinous sheaths serve to distinguish them. As *Aq. miniguttulata* is not related to the type of the genus, *Aq. hyalomura*, a new genus, *Breviascus*, is proposed to house *B. miniguttulata* (see Nomenclatural Changes).

Annulatascaceae *sensu lato* **Clade 1i** is monotypic and contains the species *Hyalorostratum brunneisporum* (**Fig. 7, Clade 1i**). Combined three gene analyses place this taxon as sister to *Cyanoannulus petersenii* without significant Bayesian or Maximum-likelihood support. The two species differ morphologically in that *H. brunneisporum* has pale to dark brown ascomata, necks with a tomentum of hyaline hyphae, and brown, 1–septate ascospores while *Cy. petersenii* has reddish-brown ascomata, reddish-brown necks, and hyaline, 3–septate ascospores. *Hyalorostratum brunneisporum* differs morphologically from other members of **Clade 1** in that it has pale to brown ascomata and a long hyaline neck. At present, *H. brunneisporum* remains *incertae sedis* at the level of family and above.

Annulatascaceae *sensu lato* **Clade 1j** consists of three species in the genus *Bullimyces, Bu. aurisporus, Bu. communis, Bu. costaricensis.* (**Fig. 7, Clade 1j**). One distinguishing feature of these taxa is that the centrum of the ascomata is filled with globose cells as opposed to the hyaline, tapering, septate paraphyses found in all other members of Annulatascaceae *sensu lato*. In the case of *Bullimyces* species, morphology of the sterile centrum cells appears to be a good phylogenetic character in agreement with molecular data.

Teracosphaeria petroica is the final species in Annulatascaceae *sensu lato* **Clade 1** and, although previously compared to Annulatascaceae (Réblová and Seifert 2007), is not related to the Annulatascaceae *sensu stricto* clade based on the phylogenetic analyses performed in this study (**Fig. 7, Clade 1k**). *Teracosphaeria petroica* differs from other members of **Clade 1** in that it has a combination of glabrous ascomata and fusiform-clavate asci with long stipitate ascus pedicels ending in a club-like structure. In addition, *Te. petroica* differs morphologically from Annulatascaceae *sensu stricto* taxa in having club shaped asci with small apical rings and the production of a phialidic anamorph.

Annulatascaceae *sensu lato* **Clade 1,** while lacking both Bayesian and Maximum-likelihood support at the basal node connecting it to the main tree, is a strong contender for higher level classification. It is composed primarily of freshwater ascomycetes with ascus apical rings that do not belong in the clade containing the type of the family, *Annulatascus velatisporus*, but have previously been referred to Annulatascaceae. Both Ophiostomatales and Annulatascaceae *sensu stricto* arise from this clade. Further refinement of the clade would be afforded with the addition of additional taxa and more genes for those taxa. There is a reasonable case for Annulatascaceae *sensu lato* **Clade 1** being ascribed a subclass, or more conservatively, an order between Diaporthales and Ophiostomatales. As this study was primarily focused on the family Annulatascaceae, resolving this higher level classification is a logical future direction.

Annulatascaceae *sensu lato* clade 2 consists of *Annulatascus citriosporus* which was determined to be sister to members of the Chaetosphaeriales clade in combined three gene analyses in this study (**Fig. 7, Clade 2**). Mcm7 and combined 18S and 28S analyses placed it in a similar position (**Fig. 5, Fig. 6**). ITS analyses placed *A. citriosporus* as a sister taxon to *Submersisphaeria aquatica* without statistical support (**Fig. 4**). 28S analyses placed it basal to the Annulatascaceae *sensu stricto* clade on a long branch without statistical support (**Fig. 2**). Based on the combined three gene analyses, this species is far removed from Annulatascaceae *sensu stricto* and *Annulatascus*. This species has all the morphological hallmarks of an Annulatascus species (see *sensu stricto* clade) save for having the only citriform ascospores in this study, and its type location was terrestrial. Along with *A. licualae,* this species was first described from a wet terrestrial habitat (palm glade), although it was recovered from freshwater in this study. Thus *A. citriosporus* may not represent a true freshwater indweller but rather a transient aquatic species. As *A. citriosporus* is unrelated to *Annulatascus* the new genus and combination, *Amphibioannulatascus citriosporus,* is proposed (see Nomenclatural Changes).

Annulatascaceae *sensu lato* Clade 3 is basal to the Chaetosphaeriales in combined three gene analyses and is a polytomous clade without Bayesian or Maximum-likelihood support composed of *Aq. triseptata*, a new *Aquaticola*-like species (PE0159), *Clohiesia corticola*, and *Submersisphaeria rattanicola* (**Fig. 7, Clade 3a,b** respectively). Due to the nature of this polytomy, it is difficult to determine the phylogenetic relationships of all these taxa at this time. As a polytomy, it is no surprise that there are few morphological characters uniting the clade. *Clohiesia corticola*, for instance forms its ascomata under a clypeus (Hyde 1995). It was placed in the Annulatascaceae *sensu lato* Clade III in the study of Jones et al. (2004) as its only member and with insignificant support sister to two *Chaetosphaeria* species. A review of previous phylogenies including this species shows that it is variably placed with very weak support (Raja et al. 2003, Duong et al. 2004, Abdel-Wahab et al. 2011). There is only one sequence (LSU) available for the entire genus and thus obtaining sequences from *Cl. curvispora* and *Cl. lignicola* and including other genes would aid in delimiting the genus and placing it in the Sordariomycetes phylogeny (Heath et al. 2008). *Submersisphaeria rattanicola* differs from the Annulatascaceae *sensu stricto* member, *S. aquatica,* in having brown uniseptate ascospore with well-defined septal constriction. *Submersisphaeria rattanicola* may thus be distinguished from *S. aquatica* by this feature of the ascospore morphology and is thus more similar to *S. vasicola* with its constricted ascospore septa. *S. vasicola* was not evaluated molecularly in this study (see taxonomic part) but may be congeneric with *S. rattanicola* as it agrees well morphologically. *Aquaticola triseptata* and PE0159, however, are supported as sister taxa (**Fig. 7, Clade 3a**) but differ in that PE0159 has uniseptate ascospores with striations on the ascospore surface rather than 3-septate ascospores with smooth walls. Other morphological features of these two species agree with the exception of measurements. *Aquaticola triseptata* and PE0159 are not related to *Aq. hyalomura* in this study, thus these two taxa represent a potentially new genus. *Insolensaquaticola triseptata* is proposed as a new combination for *Aq. triseptata* and *Pseudosubmersisphaeria rattanicola* for *S. rattanicola.* (see Nomenclatural Changes).

Annulatascaceae *sensu lato* Clade 4 is sister to the Savoryellales and is a clade consisting of the *Aquaticola* species, *Aq. ellipsoidea* and the informally described *Aq. hongkongensis*, partially corresponding taxonomically to the Annulatascaceae *sensu lato* Clade IV of Jones et al. (2014) (**Fig. 7, Clade 4**). Morphologically they both possess immersed to semi-immersed, gregarious, globose to subglobose black ascomata, cylindrical asci with small J- apical rings, and aseptate, hyaline ascospores. They differ, however, in the sizes of their asci and ascospores. These two taxa are in a clade with Bayesian and Maximum-likelihood support, though they are not related to the type of the genus, *Aq. hyalomura*. *Aquadulcemyces ellipsoidea* is proposed as a new combination for *Aquaticola ellipsoidea* (see Nomenclatural Changes). *Aquaticola hongkongensis* requires formal publication but should be added to *Aquadulcemyces*.

Annulatascaceae *sensu lato* Clade 5 consists of *Torrentispora crassiparietis* but the branch has no statistical support. Since this species does not cluster with the type of the genus, *T. fibrosa* (**Fig. 7, Clade 1g**), it should be considered Sordariomycetes *incertae sedis* at present and a new combination, *Pseudotorrentispora crassiparietis,* is proposed (see Nomenclatural Changes). From a morphological standpoint, *T. crassiparietis* and *T. fibrosa* (**Clade 1g**) agree well and would be expected cluster together, although *T. crassiparietis* has an ascus apical apparatus nearly three times as large and ascospores nearly double in size as those of *T. fibrosa*. In addition, an SSU sequence was not obtained for *T. fibrosa*, perhaps due to a long intron, and thus these taxa do not have a congruent molecular dataset. However, the LSU tree generated in this study still does not place these two species together, thus in spite of their similarities, the aforementioned morphological differences must be used to separate the taxa.

Annulatascaceae *sensu lato* Clade 6 is a monotypic clade with *Rivulicola incrustata* as its only member. It is on a branch sister to Hypocreales but with no support (**Fig. 7, Clade 6**). This species was isolated by plating germinated single ascospores and can be confidently considered as uncontaminated. The placement near Hypocreales has occasionally been associated with *Trichoderma* species contamination (Yeung at al. 2006, Zelski et al. 2014), but care was taken in this study to ensure a pure culture (all six single spore isolates had the same color and growth characteristics and did not produce a *Trichoderma* anamorph). The placement in the basal portion of the Sordariomycetes tree was replicated in all analyses with the exception of Mcm7 for which a sequence was not obtained (**Figs. 2-4, 6,7**) *Rivulicola incrustata* is the only species evaluated in this study which secretes a green pigment on woody substrates. In addition to this trait, the ascomata are hyaline and firmly immersed in the substrate, traits that separate it from Annulatascaceae *sensu stricto*. Teleomorphs in the Hypocreales tend to produce brightly colored ascomata, and thus *R. incrustata* fits in the order based on this characteristic.

Annulatascaceae *sensu lato* Clade 7 consists of two new taxa, an *Annulatascus*-like species (PE0324) and an *Ayria*-like species (PE0323) that form a group sister with Melanosporales (**Fig. 7, Clade 7**). This clade has good Bayesian and Maximum-likelihood support, but is polytomous at its root. These taxa both have hyaline ascospores but differ from one another in that PE0324 has ascospores with a thin sheath, while PE0323 has ascospores with coiled unfurling bipolar appendages and no sheath, as is found in the genera *Ayria* and *Pseudoproboscispora*. These taxa are different enough to warrant new genera (see New Taxa) and should be considered Sordariomycetes *incertae sedis*.

Annulatascaceae *sensu lato* Clade 8 is monotypic with an *Annulatascus*-like species recovered from Tennessee and Wisconsin (A612) which is a sister taxon to *Jobellisia* species **(Fig. 7, Clade 8**), although without statistical support. This species is the most basal of the Sordariomycetes species evaluated in this study (**Fig. 4**). It agrees with the majority of Annulatascaceae *sensu stricto* members in that it possesses long tapering septate paraphyses, and cylindrical unitunicate asci with a relatively massive bipartite J- ascus apical ring. It differs, however, in that its ascomata and necks are golden brown rather than dark brown to black. While superficially resembling Annulatascaceae by possessing a bipartite J- apical ring, the ascus apical ring is relatively small. Based on molecular evidence it is placed in Sordariomycetes *incertae sedis* **(Fig. 7, Clade 8**).

One of the main questions this study aimed to address was which taxa were included in the Annulatascaceae *sensu stricto* clade (**Fig. 7**). Ten of the ~35 taxa evaluated molecularly were found to belong, and it might be conjectured that a similar proportion of unsampled taxa will be found to belong in future studies. The majority of unsampled taxa are currently ascribed to the genus *Annulatascus*, however, and are thus the most likely candidates for inclusion based on the authors' determination of morphological similarity to that genus and not to other Annulatascaceae genera.

Another main question addressed in this study was which morphological characters tie the Annulatascaceae *sensu stricto* clade together. It was hypothesized that if large, J- bipartite ascus apical rings are widespread in Sordariomycetes then this trait was predicted to be phylogenetically uninformative. This was supported as evidenced by the dispersal of taxa with large, bipartite J- ascus apical rings in multiple Annulatascaceae *sensu lato* clades (**Fig. 7**). These taxa include *Cataractispora* spp., *Fluminicola* spp., *Mirannulata samuelsii*, and *Vertexicola* spp. It was also hypothesized that if ascospore morphology is under selective pressure in aquatic habitats, then this trait would also be phylogenetically uninformative. This hypothesis was supported by the presence of ascospores of widely varied morphology (size, shape, presence/absence of sheaths and/or appendages) even within the

Annulatascaceae *sensu stricto* clade. Finally, ascomal morphology was predicted to be phylogenetically informative as it is a more conserved trait, at least at the generic level (Miller and Huhndorf 2005). This was found to be true for the Annulatascaceae *sensu stricto* though in combination with other characters. Annulatascaceae *sensu stricto* species share the following combination of features: freshwater habitat, dark brown to black coriaceous ascomata; well-developed dark brown to black periphysate necks; filamentous, hyaline, tapering, septate paraphyses; 8–spored, and unitunicate asci with a J- ascus apical apparatus. The exception to this rule is *Paoayensis lignicola*, for reasons previously mentioned.

One further consideration is the route by which species become aquatic. Shearer (1993) proposed a number of scenarios for the adaptation of terrestrial taxa to the aquatic niche. Pathogens, endophytes and saprobes could all be subjected to the submerged environment through various routes. Senescence of aquatic macrophytes may have led to a facultative shift of endophytes to saprobic habit. In the same vein, terrestrial endophytes or pathogens can be carried into water via leaf drop or woody debris directly, or washed in later indirectly. Saprobes of terrestrial plant debris would also be transported into water along with their substrates. A final possibility is that spores themselves may be blown/washed into freshwater. As such, a polyphyletic freshwater mycota with traits adapted to the aquatic habitat forms the ecological group we know as freshwater ascomycetes. With respect to Annulatascaceae (in the broad sense) ascus apical rings could be advantageous (or at least not being deleterious) in preventing asci from swelling and rupturing and by favoring back-door dehiscence and ascospore dispersal. The trait may also be useful for periodic drying due to fluctuating water levels to maintain the integrity of the ascus.

Belliveau and Bärlocher (2005), using SSU molecular evidence for 30 taxa, put forth the idea that multiple origins of entry to the aquatic environment explains the presence of several species of aquatic hyphomycetes in the classes Dothideomycetes, Leotiomycetes, Orbiliomycetes, and Sordariomycetes. Vijaykrishna et al. (2006), again using SSU data, but with 94 sequences, demonstrated that freshwater fungi evaluated from asexual and sexual morphs have multiple origins from terrestrial species in widely separated lineages. These taxa reside in the subclasses Dothideomycetidae, Leotiomycetidae, and Sordariomycetidae. Shearer et al. (2009), using SSU and LSU data, showed that many orders of Dothideomycetes have representatives adapted to the aquatic habit, both in sexual and asexual state with spore dispersal adaptations in their reproductive structures as well as their spores.

Taxa with ascus apical rings of different chemical composition (J-, J+) and architecture that do not explosively discharge ascospores are common throughout the Hypocreomycetidae, Orbiliomycetidae, Sordariomycetidae, and Xylariomycetidae (Shearer 1989, Belliveau and Bärlocher 2005, Vijaykrishna et al. 2006). For instance, *Chaetomium* species, and taxa in Diaporthales and Microascales do not always forcibly discharge ascospores. Rather these asci typically deliquesce to release ascospores, a potential adaptation to a wet (fluid) vs. dry (air) environment. As an example in a terrestrial setting, ascospores of *Sordaria* spp. commonly have mucilaginous sheaths or appendages that afford the ability to cling to substrates when dispersed.

One interpretation of apical rings is that they provide for the structural integrity of the ascus as ascospores mature. The apical apparatus physically reinforces the weakest part of any cylinder, the apex. This feature may prevent apical discharge in favor of back door dehiscence, which is common in Annulatascaceae taxa. In the Annulatascaceae *sensu stricto*, ascospores are considerably wider than the apical rings, and should only be forcibly ejected if the annulus is stretchable or the ascospores are highly compressible in order to pass through. A sturdy ascus apical ring may be one solution to retaining ascospores until release via back-door dehiscence. It may prevent premature ascus rupture due to swelling of the ascus by imbibing water. In the majority of species examined in this study, asci were observed to dehisce 1/3 to 2/3 the distance of the asci from the ascus apex. This mode of ascospore release, coupled with asci being discharged from the ascoma in many taxa rather than ascospores, may be advantageous in ascospore dispersal in an aquatic environment, and may have come from a pre-adaptation developed in a moist terrestrial environment.

CHAPTER 5

TAXONOMY

Annulatascales D'souza, Maharachch. & K.D. Hyde, Fungal Diversity 72: 212. 2015

Index Fungorum IF551133

Type family: Annulatascaceae

Comments: The order Annulatascales was introduced by Maharachchikumbura et al. (2015) essentially as a higher order classification of Annulatascaceae. The order was based on a taxon sampling of four Annulatascaceae taxa in a larger multi-gene analysis used to investigate the phylogenetic structure of the Sordariomycete backbone. Sequences used in the study were mined from GenBank and did not include newly generated sequences. The description of the order was essentially the description of the family which follows. The 18 genera described in the introduction to this manuscript were included in Annulatascales, which is placed in the newly erected subclass Diaporthomycetidae Senan., Maharachch. & K.D. Hyde. These names are not widely being accepted by the mycological community and while Annulatascales is a good name for the order containing Annulatascaceae, its circumscription requires revision based on the results herein..

Annulatascaceae S.W. Wong, K.D. Hyde & E.B.G. Jones, Systema Ascomycetum 16: 18. 1998.

MycoBank 81939.

Type genus: *Annulatascus.*

Comments: This family was introduced by Wong et al. (1998) and included six genera. The family was reexamined by Ho and Hyde (2000) with a focus on ultrastructural characteristics and an additional five genera were included. In addition to the ascus apical apparatus, the ultrastructure and ontogeny of the asci and ascospores were considered important for the inclusion in the family (Ho and Hyde 2000). Annulatascaceae is characterized by having dark brown to black coriaceous ascomata with well developed periphysate necks, long, tapering, septate paraphyses, 8-spored unitunicate asci with relatively massive J- apical rings, and ascospores of varying septation and pigmentation that possess sheaths, appendages, both or neither.

Annulatascus K.D. Hyde, Australian Systematic Botany 5: 118. 1992. MycoBank 25398.

Type species: *Annulatascus velatisporus.*

Annulatascus apiculatus F.R. Barbosa & Gusmão, Mycotaxon 106: 405. 2008. MycoBank 512121.

Ascomata black, coriaceous, semi-immersed, perpendicular to surface, clustered, 400–550 µm wide × 240–410 µm high (x̅ = 475 × 325 µm, w:h = 1.5), globose. **Ascomal wall** composed of dark brown cells, 15–60 µm thick. **Neck** 100–250 µm long × 100–180 µm wide, periphysate. **Paraphyses** hyaline, 2.5–6 µm wide × 75–100 µm long, tapering, septate. **Asci** unitunicate, cylindrical, 8–spored, 175–250 µm long × 10–13 µm wide (x̅ = 212.5 × 11.5 µm, l:w = 18.5). **Ascus base** pedicellate, long, tail-like, 50 µm from *pars sporifera*. **Annulus** J-, bipartite, 6–7.2 µm long × 1.8–2.4 µm wide, (x̅ = 6.6 × 2.1 µm, l:w = 3.1). **Ascospores** hyaline, overlapping uniseriate, fusiform, 0–3 septate, not constricted at septa, 23–36.5 µm long × 8.8–10 µm wide (x̅ = 29.8 × 9.5 µm, l:w = 3.2), with bipolar apiculi, with a narrow sheath (Barbosa et al. 2008).

HOLOTYPE: BRAZIL. Bahia, Santa Terezinha, Serra da Jibóia, stream, on submerged wood, 19 Feb 2008, *F.R. Barbosa,* **HUEFS 134723.**

Known distribution: Brazil.

Comments: The holotype of *Annulatascus apiculatus* was not examined. Based on the protologue, this species best fits morphologically within the genus *Annulatascus*. *A. apiculatus* is most similar to *A. velatisporus*. Both taxa have black ascomata with well developed necks, asci of similar length with relatively large apical rings, and 0–3 septate ascospores of comparable size. The taxa differ in that *A. apiculatus* possesses ascospores with bipolar apiculi and a narrow sheath, while *A. velatisporus* lacks apiculi and has a spreading sheath. A culture was not obtained from the type collection (Barbosa pers. comm.) and therefore molecular data should be obtained from fresh collections made at or near the type location in order to determine its phylogenetic placement, which should currently be considered Sordariomycetes *incertae sedis*.

Annulatascus aquaticus W.H. Ho, K.D. Hyde & I.J. Hodgkiss, Fungal Diversity 2: 121. 1999. MycoBank 450113.

Ascomata dark brown, coriaceous, mostly immersed to superficial, perpendicular to surface, solitary, 240–600 µm wide × 220–600 µm high ($\bar{x}$ = 420 × 410 µm, w:h = 1), globose to subglobose. **Ascomal wall** brown, composed of layers of dark brown compressed cells of *textura angularis* in surface view. **Neck** dark brown, periphysate, 400–800 µm long × 60–80 µm wide. **Paraphyses** hyaline, 3–4.5 µm wide at base, tapering, septate. **Asci** unitunicate, cylindrical, 8–spored, 150–175 µm long × 10–12 µm wide ($\bar{x}$ = 160 × 11 µm, l:w = 14.6). **Ascus base** pedicellate, slightly tapering to small knob, 40 µm from *pars sporifera*. **Annulus** J-, bipartite, 4.5–5 µm long × 2.5–4 µm wide ($\bar{x}$ = 4.75 × 3.25 µm, l:w = 1.5). **Ascospores** hyaline, ellipsoidal, aseptate, overlapping uniseriate, 19–24 µm long × 6–7 µm wide ($\bar{x}$ = 21.5 × 6.5 µm, l:w = 3.3), with 2–4 lipid guttules, with gelatinous sheath evident at SEM level (Ho et al. 1999).

HOLOTYPE: HONG KONG. Tai Po Kau Forest Stream, on submerged decaying wood, 27 Jun 1996, *K.D. Hyde, WH238,* **HKU(M) 4526.**

Known distribution: Hong Kong.

Comments: The holotype of *Annulatascus aquaticus* was unavailable for examination and fresh specimens were not obtained in this study, thus the original description was used herein for morphological study and for inclusion in the Annulatascaceae morphological database. Ultrastructural studies performed on *A. aquaticus* (Ho et al. 1999) showed the ascus walls to be bilamellate and the apical rings to be bipartite which agrees with the type of the family, *A. velatisporus*, and the species *A. triseptatus*. In addition, a sheath was found to be present at the SEM level, which did not appear at the light microscope level. Morphologically *A. aquaticus* is most similar to *A. joannae*, although *A. joannae* has larger ascospores, blunter ascospore ends and thicker ascospore walls. In both species, ascospores are aseptate and show a distinct pattern of guttulation with two large guttules in the center of the ascospore flanked by smaller guttules at either end. A culture was not indicated in the protologue and if the holotype is lost, neotypification followed by molecular analyses should be performed to resolve the phylogenetic placement of this species, which should be considered Sordariomycetes *incertae sedis* at present.

Annulatascus aquatorba Boonyuen & Sri-indrasutdhi, Mycologia 104(3): 752. 2012. **Fig. 8 A-G.** MycoBank 519473.

Ascomata brown to dark brown, semi-immersed to superficial, perpendicular to surface, solitary, 390–500 µm wide × 310–350 µm high (x̅ = 445 × 312 µm, w:h = 1.4, n = 4), globose. **Ascomal wall** brown, of thick-walled brown pseudoparenchymatous cells in the outer layers, inner layers of thinner walled compressed cells. **Neck** 400–480 µm long × 100–120 µm wide (x̅ = 450 × 110 µm). **Paraphyses** hyaline, 2.5 µm wide at base, septate, filiform, simple, rarely branched. **Asci** unitunicate, cylindrical, 8–spored, 103–153 µm long × 6.3–8.8 µm wide (x̅ = 114 × 7.5 µm, l:w = 15.2) (**Fig. 8 A,B**). **Ascus base** pedicellate, 20 µm from *pars sporifera* (**Fig. 8 A**). **Annulus** J-, wedge-shaped, 5.0–7.5 µm wide (**Fig. 8 C,D**). **Ascospores** with brown central cells, end cells subhyaline, with rounded apices, fusoid to lunate, 3–septate, slightly constricted at septa, 15–22.5 µm long × 5–7.5 µm wide (x̅ = 19.2 × 6.5 µm, l:w = 3), with no appendages or sheath (**Fig. 8 E-G**) (Boonyuen et al. 2012).

HOLOTYPE: THAILAND. Narathiwat Province, Sirindhorn Peat Swamp Forest, on submerged wood test block of *Erythrophleum teysmannii*, 22 Feb 2003, *N. Boonyuen and V. Sri-indrasutdhi,* **BBH 29936.**

Specimens examined:

PERU. Cusco, Camanti, Quincemil, 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 21.3 °C, pH 6, on submerged woody debris, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0255-2;* Madre de Dios, palm swamp (agaujal) off the Interoceanic Highway outside of Puerto Maldonado, 12°42’48.0954”S, 69°28’11.28”W, 239m, on submerged woody debris, water 23.3 °C, pH 5.9, 20 May 2010, *Zelski S.E. and H.A. Raja, PE0255-4;* CICRA, Stream along at Trail 23, 12°33’31.03”S, 70°05’56.96”W, 280m, on submerged woody debris, water 23.3 °C, pH 7.8, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0255-1.*

Known Distribution: Perú, Thailand.

Comments: The holotype of *Annulatascus aquatorba* was not examined as the species was described toward the end of this study. However, fresh specimens with high sequence identity to those of the holotype were obtained from collections in Perú. Peruvian specimens were a good match to the holotype description, and in sequence analyses clustered with good support with sequences from the holotype deposited in GenBank. The type specimen was found on Fabaceae test blocks (Boonyuen et al. 2012), while the Peruvian specimens were found on unknown natural substrates. This species bears a striking similarity to *Ascitendus austriacus*, and Peruvian specimens were first diagnosed as such. The only

Fig. 8 A-G. *Annulatascus aquatorba* from specimen PE0255. A-B. Asci. C-D. Ascus apices. E-F. Ascospores. G. Ascospore. Bars: A-B = 20 µm; C-G = 10 µm.

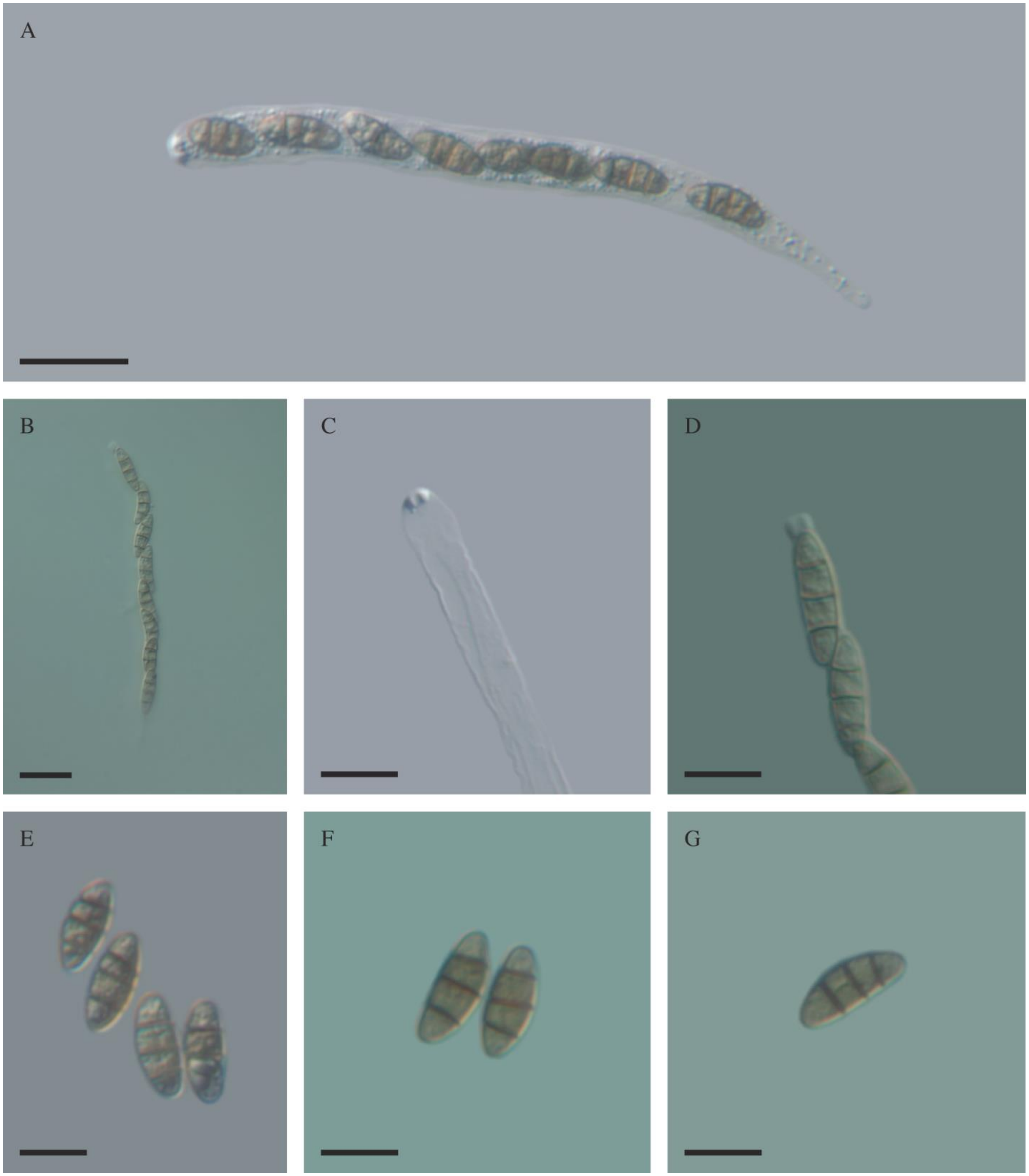


Fig. 8.

differences noted between these two taxa were that *A. aquatorba* has shorter asci (**Fig. 8 A,B**), a wider annulus (**Fig. 8 C,D**), and smooth walled ascospores that are slightly constricted at the septa (**Fig. 8 E-G**) compared to *As. austriacus*.

Annulatascus aquatorba has good statistical support for affinity with the type of the genus, *A. velatisporus* in this study, and is a member of the Annulatascaceae *sensu stricto* clade (**Fig. 7**). Boonyuen et al. (2012) placed this species in a subclade of Annulatascaceae taxa that contained *A. velatisporus, A. hongkongensis,* and *A. nilensis*. Whether *A. aquatorba* is congeneric with *A. velatisporus* remains to be determined, as gene selection and taxon sampling influence its phylogenetic placement in this study. In Mcm7 and the three gene analyses (**Fig. 5, Fig. 7**), *A. aquatorba* is separated from *A. velatisporus,* while in all other analyses it is a close neighbor (**Figs. 2-4, Fig. 6**).

Annulatascus biatriisporus K.D. Hyde, Nova Hedwigia 61: 120. 1995. **Fig. 9 A-P, Fig. 10 A-J.** MycoBank 412469.

Ascomata black, coriaceous, immersed, perpendicular to surface, solitary or gregarious, 195–325 µm wide × 390–520 µm high (x̅ = 260 × 455 µm, w:h = .6), ellipsoidal (**Fig. 9 G**). **Ascomal wall** to 50 µm thick, outer layers dark, of *textura angularis*, inner layers hyaline, thin walled, elongate. **Neck** black, ~ 390 µm long × 110 µm wide, periphysate. **Paraphyses** hyaline, as long as asci, 4 µm wide at base, tapering toward apex, septate (**Fig. 10 B)**. **Asci** unitunicate, long-cylindrical, 8–spored, 210–260 µm long × 12–17 µm wide (x̅ = 235 × 14.5 µm, l:w = 16.2) **(Fig. 9 G, Fig. 10 A)**. **Ascus base** short, pedicellate, 20 µm from *pars sporifera*, tapering to rounded end (**Fig. 9 H; Fig. 10 A,E**). **Annulus** J-, large, 7–8 µm long × 3–4 µm wide (x̅ = 7.5 × 3.5 µm, l:w = 2.1) (**Fig. 9 I-K; Fig. 10 C,D**). **Ascospores** hyaline, fusiform, aseptate, overlapping uniseriate, 48–65 µm long × 7.5–10 µm wide (x̅ = 52.5 × 8.8 µm, l:w = 6), with weakly swollen ends, with a thin, irregular gelatinous sheath (**Fig. 9 L-F, Fig.10 G-J**) (Hyde 1995).

Specimens examined:

HOLOTYPE: AUSTRALIA. Northern Queensland, Babinda, The Boulders, on submerged wood, Dec 1991, *K.D. Hyde 880,* **BRIP 21481.**

PERU. Cusco, Camanti, Stream at Quincemil Trail 1, 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0330-1.*

Known distribution: Australia, Brazil, China, Costa Rica, Perú.

Comments: The holotype of *Annulatascus biatriisporus* was in good condition (**Fig. 9 A-F)**. The initial name, recorded in documentation included in the herbarium packet, was *A. biatriospora* (**Fig. 9 A,D,E)**. Holotype substrate material consisted of a 16 × 2.5 cm piece of dried wood (**Fig. 9 F**). Two permanent slides with one ascoma per slide were prepared, examined, and returned to BRIP. Asci were scant and in poor condition (**Fig. 9 F**). The originally described bipolar apical chambers of the ascospores on closer scrutiny appear to be massive apical thickenings of the ascospore walls (**Fig. 9 L-P**). A significant feature of this fungus suggesting placement in *Annulatascus* is its relatively massive ascus apical ring (**Fig. 9 I-K; Fig. 10 E,D**). Peruvian specimens of *A. biatriisporus* (PE0330) agree well with the holotype, differing only in having shorter ascospores on average (44 µm vs. 52 µm long). These differences may be attributable to changes in the techniques in measurement in the last 20 years. Peruvian specimens had different ascus apical ring measurements on average (5 × 6.3 µm) than those reported in the protologue (7.5 × 3.5 µm). Examination of the holotype in this study resulted in annulus measurements of 6 × 7 µm, which is more in keeping with Peruvian specimen annulus measurements. Again, these differences may be attributable to changes in the techniques in measurement in the last 20 years. Raja et al. (2003) first showed *A. biatriisporus* to be in a weakly supported Annulatascaceae clade, sister to *Cyanoannulus petersenii*. Abdel-Wahab et al. (2011) in a 28S study showed that *A. biatriisporus* is in a separate lineage from Annulatascaceae *sensu stricto*. Boonyuen et al. (2012), in a combined 18S and 28S molecular study,

Fig. 9 A-P. *Annulatascus biatriisporus* from the holotype **BRIP 21481**. A. Herbarium packet (box). B. Side of herbarium packet. C. Contents of herbarium packet. D. Original correspondence for deposit at BRIP. E. Herbarium label (note original spelling) F. Dried holotype material. G. Cavities containing partial venters (arrows). H. Ascus. I-K. Ascus apices. L-P. Ascospores. Bars: G = 500 µm, H-P = 20 µm.

UNIVERSITY OF HONG KONG
DEPARTMENT OF BOTANY
Pokfulam Road, Hong Kong
香港大學
植物學系
香港薄扶林道

Tel:
Fax: (852) 8583477
Telex: 71919 CEREB HX

14/12/93
Dr. J. Alcorn,
Division of Plant Protection,
DPI.,
Meiers Road, Indooroopilly
Australia

23 November 1993

Dear John,

Please find enclosed 15 specimens (ascomycetes) to be deposited at BRIP.

1 *Annulatascus biatriospora* K.D. Hyde sp. nov. Australia, north Queensland, Babinda, The Boulders, on submerged wood, Dec 1991, K.D. Hyde 880, BRIP 21481 (holotype).

HERBARIUM BRIP
NATIONAL COLLECTION OF FUNGI

BRIP 21481 (Holotype) Ascomycotina

Annulatascus biatriospora K.D. Hyde

Host:

Dis.:
Loc.: The Boulders Babinda Qld Australia
Coll.: K.D. Hyde Date: Dec 1991
Det.: K.D. Hyde
Dup.:
Remarks: Boxed specimen. Nova Hedwigia 61: 120. 1995.

Fig. 9.

Fig. 10 A-J. *Annulatascus biatriisporus* from specimen PE0330-1. A. Ascus. B. Paraphyses. C-D. Ascus apices. E. Ascus base. F. Overlapping uniseriate ascospores. G-J. Ascospores. Bars: A = 40 µm, B-J = 20 µm.

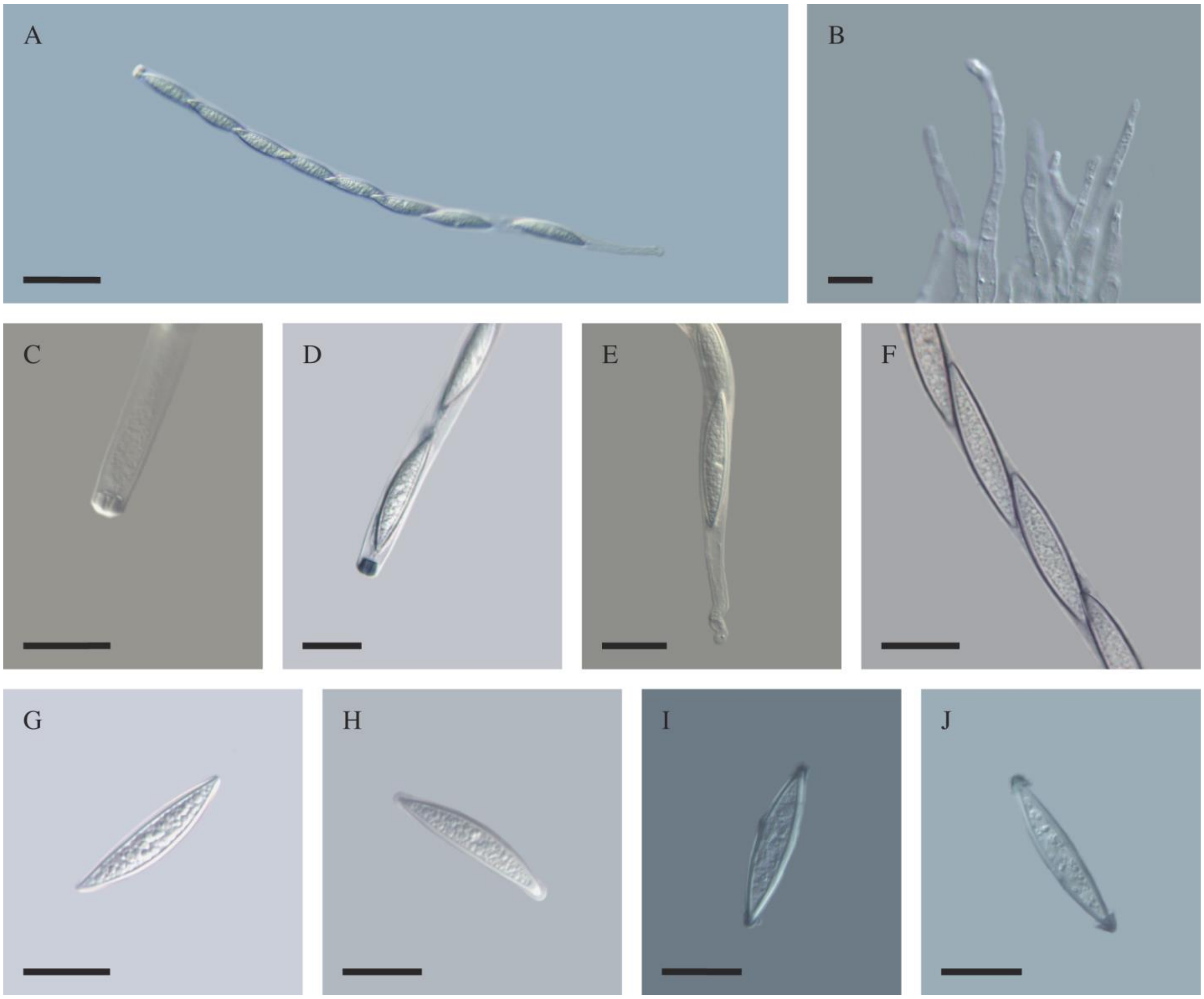


Fig. 10.

also excluded this species. The three gene analyses in this study also support the exclusion of *A. biatriisporus* from *Annulatascus* and Annulatascaceae. Additional specimens of *A. biatriisporus* have been reported from China (Tsui et al. 2002) and most recently from Costa Rica (Barbosa et al. 2013). Molecular data from specimens collected in these locations would aid in assessing this geographically broad distributional pattern. *A. biatriisporus* clusters with a new *Annulatascus*-like species (PE0035), *Fusoidispora aquatica*, *Aquaticola*-like species (PE0316, AK-1838), and *Lentomitella* species in combined three gene molecular analyses in this study (**Fig. 7, Clade 1f**). As *A. biatriisporus* is not part of the Annulatascaceae *sensu stricto* clade and is distantly related to *A. velatisporus*, it is removed from the genus as well as the family and a new genus and combination *Ascuslongus biatriisporus* is proposed (see Nomenclatural Changes). In the multi-gene analyses of this study, *A. biatriisporus* is in a clade separating Ophiostomatales and Diaporthales which places it in Sordariomycetes *incertae sedis* at present (**Fig. 7, Clade 1f**).

Annulatascus citriosporus J. Fröhl. & K.D. Hyde, Palm Microfungi. Fungal Diversity Series 3: 79. 2000. **Fig. 11 A-L, Fig. 12 A-F.** MycoBank 482750 (syn. *Annulatascus citrisporus* MycoBank 580506, legitimate).

Ascomata black, immersed, perpendicular to surface, 350–485 µm wide × 224–310 µm high, ($\bar{x}$ = 417.5 × 267 µm, w:h = 1.6), subglobose (**Fig. 11 D**). **Ascomal wall** brown, 5–6 layers of thick walled brown cells 4–8.8 × 2.8–3.8 µm, 3.8–17.5 µm thick (**Fig. 11 E**). **Neck** black, erumpent ostiole visible at surface of substrate, periphysate. **Paraphyses** hyaline, 6–8 µm wide × 72.5–155 µm long, tapering, septate. **Asci** unitunicate, cylindrical, 8–spored, overlapping uniseriate, 137.5–178.8 µm long × 10–12 µm wide ($\bar{x}$ = 158.2 × 11 µm, l:w = 14.4). **Ascus base** short pedicellate (**Fig. 11 J, Fig. 12 A**). **Annulus** J-, lens shaped, 2.75–3.25 µm long × 5–5.5 µm wide ($\bar{x}$ = 3 × 5.25 µm, l:w = .6) (**Fig. 11 F-I, Fig. 12 A-C**). **Ascospores** hyaline, citriform (lemon shaped), 0–3 septate, not constricted at septa, uniseriate, 22.5–30.4 µm long ×

6.4–8.5 µm wide (x̅ = 26.5 × 7.5 µm, l:w = 3.6), lipids arranged as 1–2–1 large guttules, without appendages, with a thin gelatinous sheath (**Fig. 11 K,L; Fig. 12 D-F**) (Fröhlich and Hyde 2000).

Specimens examined:

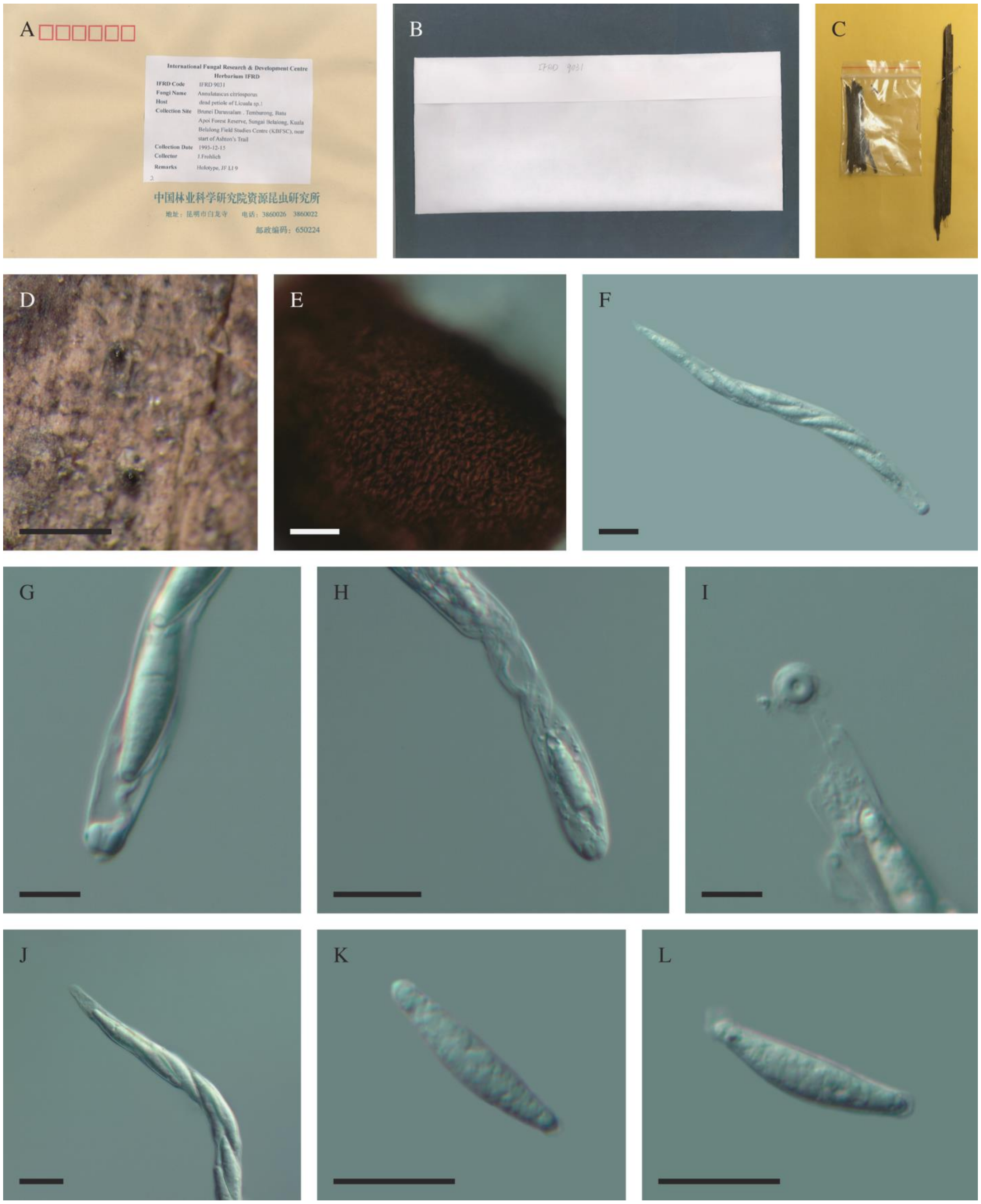
HOLOTYPE: BRUNEI DARUSSALAM. Temburong, Batu Apoi Forest Reserve, Kuala Belalong Field Studies Centre (KBFSC), Sungai Belalong, near start of Ashton's Trail, on dead petiole of *Licuala sp.*, Dec 1993, *J. Fröhlich, (HKU(M)JF LI 9),* **IFRD 9031.**

PERU. Cusco, Camanti, stream along Quincemil trail 1, 13°14'22.5594"S, 70°46'12.6114"W, 688m, on submerged woody debris, water 19.2 °C, pH 7.7, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0086-1;* Stagnant ditch along Quincemil trail 2, 13°13'40.404"S, 70°45'14.184"W, 659m, on decorticated wood, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0086-3;* Stream at Quincemil Trail 3, on submerged woody debris, 13°17'7.008"S, 70°47'13.632"W, 737m, water 21.7 °C, pH 7.6, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0086-7;* Stream at Quincemil trail 3, on submerged woody debris, 13°18'27.756"S, 70°48'44.9274"W, 757m, water 21.4 °C, pH 7.5, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0086-2;* Madre de Dios, CICRA, Pozo Don Pedro, palm swamp (aguajal), Trail 17, 12°33'34.00"S, 70°6'37.75"W, 243m, on submerged decorticated wood, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0086-4.*

Known distribution: Brunei, Perú.

Comments: The holotype of *Annulatascus citriosporus* is in good condition (**Fig. 11 A-C)**. *A. citriosporus* was first described from a dead palm petiole in the genus *Licuala* collected in Brunei (Fröhlich and Hyde 2000). *A. citriosporus*, as well as *A. licualae* were described in that study and represent rare members of the Annulatascaceae reported from a terrestrial habitat rather than an aquatic one (Fröhlich and Hyde 2000, Hyde and Fröhlich 2003). Peruvian specimens of *A. citriosporus* were collected in freshwater and are a good match morphologically to the holotype. Phylogenetically, *A.*

Fig. 11 A-L. *Annulatascus citriosporus* from the holotype **IFRD 9031**. A. Herbarium packet. B. Envelope containing specimen. C. Dried petiole of *Licuala sp*. D. Necks protruding from the surface of the substrate. E. Peridium. F. Ascus. G-H. Ascus apices. I. Annulus end on. J. Ascus base. K-L. Ascospores. Bars: D = 500 µm; F, H, J-L = 20 µm; G, I = 10 µm.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 9031
Fungi Name	Annulatascus citriosporus
Host	dead petiole of Licuala sp.1
Collection Site	Brunei Darussalam . Temburong, Batu Apoi Forest Reserve, Sungai Belalong, Kuala Belalong Field Studies Centre (KBFSC), near start of Ashton's Trail
Collection Date	1993-12-15
Collector	J.Frohlich
Remarks	Holotype, JF LI 9

2

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

Fig. 11.

Fig. 12. *Annulatascus citriosporus* from specimen PE0086. A. Ascus. B-C. Ascus apices. D. Ascospore. E. Ascospore with mucilaginous sheath. F. Ascospore. Bars: A = 20 µm; B-F = 10 µm.

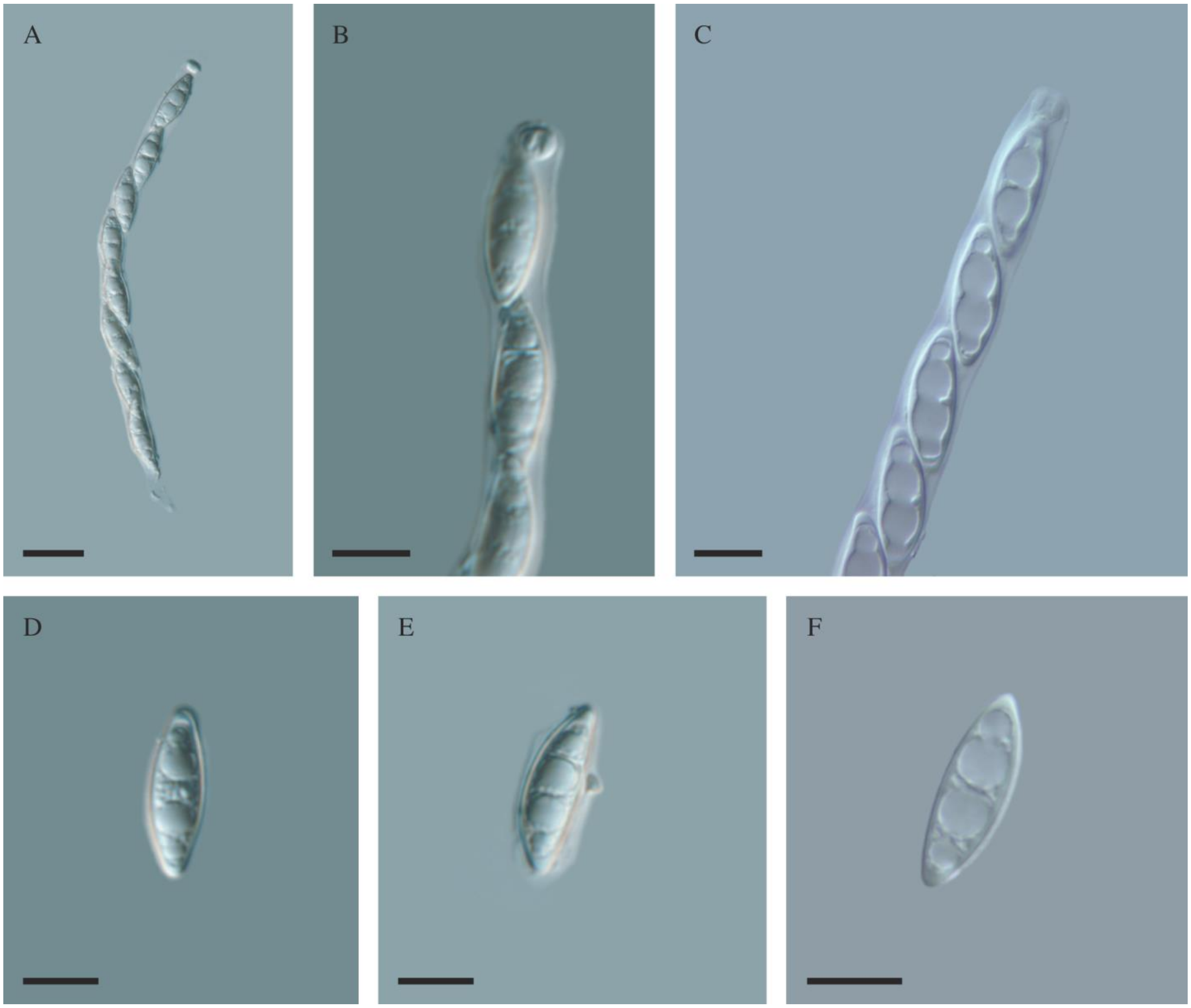


Fig. 12.

citriosporus is removed from *Annulatascus* and Annulatascaceae *sensu stricto* based on the analyses of this study. *A. citriosporus* is a separate lineage with a long branch in all analyses conducted in this study. 28S analysis places it on a long branch close to Annulatascaceae *sensu stricto* (**Fig. 2**), while combined three gene analyses place it on a long branch sister to Chaetosphaeriales (**Fig. 7, Clade 2**). Morphologically, *A. citriosporus* differs from the type of *Annulatascus, A. velatisporus*, in that it has immersed ascomata with only the tip of the neck appearing at the surface of the substrate, a lens-shaped annulus, and lemon-shaped ascospores with a distinctive guttulation. A new genus is thus proposed (see Nomenclatural Changes) which is placed in a clade with Sordariales, Boliniales and Chaetosphaeriales in the combined gene analyses (**Fig. 7, Clade 2**). Thus *A. citriosporus* is considered Sordariomycetes *incertae sedis.*

Annulatascus crassitunicatus Ho Ph.D. thesis, 53. 1998.

Ascomata dark brown, immersed to superficial, scattered, 280–400 µm wide × 200–380 µm high, globose to subglobose. **Ascomal wall** brown, coriaceous, of *textura angularis* in surface view. **Neck** dark brown, cylindrical, 550–950 µm long × 75–90 µm wide, periphysate. **Paraphyses** hyaline, 8–14 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 95–120 µm long × 14–16 µm wide ($\bar{x}$ = 116 × 15.2 µm, l:w = 7.6, n = 10). **Ascus base** short, not differentiated from ascus body. **Annulus** J-, 4.5–5 µm long × 4–5.5 µm wide ($\bar{x}$ = 4.75 × 4.75 µm, l:w = 1). **Ascospores** hyaline, ellipsoidal with acute ends, 3–septate, thick walled, overlapping uniseriate, 24–29 µm long × 6–8.5 µm wide ($\bar{x}$ = 26.4× 7.2 µm, l:w = 3.7, n = 25), guttulate, without appendages or sheath (Ho 1998).

HOLOTYPE: HKU(M) 2971

Comments: *Annulatascus crassitunicatus* has only been published in Ho's Ph.D. thesis (1998) and is not a recognized name in Index Fungorum or MycoBank. The holotype was not examined in this study as

Ho's thesis was only obtained near the completion of this manuscript. Thus only the protologue was used to assess this species morphologically. Ho noted that the thesis would only have 5 copies made and that due to this limited distribution, the ICBN would not allow new species to be considered valid (*nomen invalidum*) until published independently and broadly distributed. As this thesis is freely available online at this time, this species is being included in this study for completeness, with all credit given to Ho (1998). Though described as cylindrical, the asci appear clavate in the micrographs, and ascospores are overlapping uniseriate to nearly biseriate. In addition, the shape of the ascus apical ring appears to be wedge-shaped. Morphologically, these features support placement in *Annulatascus* and *Annulatascaceae*. However, *Annulatascus* and Annulatascaceae *sensu stricto*, as assessed molecularly, do not contain members with thick walled ascospores. The thick ascospore walls are reminiscent of some *Aquaticola* species, *Cataractispora* species, and *Torrentispora* species. *Torrentispora* and *Cataractispora* species possess long cylindrical asci as opposed to the clavate, short asci in *A. crassitunicatus*. Furthermore, *Torrentispora* species have aseptate, thick walled ascospores, while *Cataractispora* species have ascospores with appendages. Among *Aquaticola* species, only *Aq. triseptata* possesses 3–septate ascospores, though the walls are not nearly as thick, and the ascospore ends are not acute. Further work is necessary to formally publish and place *A. crassitunicatus* at the generic level and above.

Annulatascus fusiformis K.D. Hyde & S.W. Wong, Mycologia 92(3): 553. 2000. MycoBank 467429.

= *Gurgispora fusiforma* K.D. Hyde, S.W. Wong & E.B.G. Jones, Wong Ph.D. thesis, 153. 1996.

Ascomata black, immersed to semi-immersed, perpendicular to surface, solitary, 170–220 µm wide, globose to subglobose. **Ascomal wall** hyaline and brown, coriaceous, outer part several layers of thick-walled, brown, compressed cells, inner cell layers hyaline and elongate. **Neck** brown to black, long, with hyaline apex, periphysate. **Paraphyses** hyaline, 7.5 µm wide, tapering, septate, constricted at septa. **Asci**

unitunicate, long-cylindrical, 8–spored, 141–235 µm long × 7.5–10.5 µm wide (x̅ = 188 × 9 µm, l:w = 21). **Ascus base** short pedicel, 10 µm long from *pars sporifera*. **Annulus** J-, bipartite, 4–5 µm long × 3–4 µm wide (x̅ = 4.5 × 3.5 µm, l:w = 1.3). **Ascospores** hyaline, fusiform, 0–5 septate, not constricted at septa, uniseriate to overlapping uniseriate, 16.5–25.5 µm long × 6–9 µm wide (x̅ = 21× 7.5 µm, l:w = 2.8), multiguttulate, verruculose at SEM level, with bipolar mucilaginous pad-like appendages, without sheath (Hyde and Wong 2000).

HOLOTYPE: PHILIPPINES. Mindanao, Bukidnon, Impalutao, Natigbasan Creek, on submerged wood, Jan 1994, *K.D. Hyde*, **HKU(M) 3102.**

Known distribution: Philippines.

Comments: The holotype of *Annulatascus fusiformis* was unavailable for examination and fresh specimens were not obtained in this study. Thus, the protologue was used for morphological comparisons and included in the Annulatascaceae database. Of nomenclatural note is the original tentative name, *Gurgispora fusiforma*, presented in Wong's Ph.D. thesis (1998). *A. fusiformis* was placed in *Annulatascus* due to its J- bipartite refractive apical ring, as well as its bipolar, pad-like, ascospore appendages, which were seen as indicative of a primitive sheath (Wong and Hyde 2000). *A. fusiformis* differs from *A. velatisporus*, the type of the genus, in having smaller ascomata, shorter and thinner asci, a smaller annulus, and smaller ascospores which are up to 5–septate. With a massive apical ring and other typical features of *Annulatascus* species, the current genus is the most appropriate placement for *A. fusiformis*. Neotypification is recommended from fresh material collected at the type location in the Philippines. In addition, molecular evidence derived from pure culture or fungal tissue should be obtained to determine the placement of this species in Sordariomycetes, which should be considered Sordariomycetes *incertae sedis*.

Annulatascus hongkongensis W.H. Ho, Ranghoo, K.D. Hyde & I.J. Hodgkiss, Mycologia 91(5): 886. 1999. MycoBank 460558.

Ascomata dark brown, coriaceous, mostly immersed, perpendicular to surface, solitary, 250–280 µm wide, subglobose to ellipsoidal. **Ascomal wall** brown, 18–23 µm thick, of *textura angularis* in surface view. **Neck** brown, 140–150 µm long × 35–40 µm wide, 5–7 layers thick composed of globular brown cells, periphysate. **Paraphyses** hyaline, 6.5–8.5 µm wide × 200–250 µm long, tapering, septate. **Asci** unitunicate, cylindrical, 8–spored, 250–275 µm long × 25–30 µm wide ($\bar{x}$ = 262.5 × 27.5 µm, l:w = 9.6). **Ascus base** pedicellate. **Annulus** J-, large, 3.5–4.5 µm long × 4.7–6 µm wide ($\bar{x}$ = 4 × 5.35 µm, l:w = .75). **Ascospores** hyaline, ellipsoidal, 3–septate, not constricted at septa, uniseriate to overlapping uniseriate, 35–37.5 µm long × 12.5–15 µm wide ($\bar{x}$ = 36.3 × 13.8 µm, l:w = 2.7), guttulate, without appendages, with a thick sheath (Ho et al. 1999).

HOLOTYPE: HONG KONG. Plover Cove Reservoir, on submerged decaying wood, 15 Nov 1996, *K.D. Hyde and M. Wong, PC 21*, **HKU(M) 4702.**

Known distribution: Hong Kong.

Comments: The holotype of *Annulatascus hongkongensis* was unavailable for examination and fresh collections were not obtained in this study. This species may require neotypification. If the type culture HKUCC 3702 is available, procuring additional gene sequences would be beneficial for phylogenetic analyses. Molecular data in GenBank from the type culture was used in this and previous studies to assess the phylogenetic placement of *A. hongkongensis*. Previous studies have shown that *A. hongkongensis* resides in the Annulatascaceae *sensu stricto* and has been consistently shown to be closely related to the type of the genus, *A. velatisporus* (Raja et al., 2003, Campbell et al. 2003, Abdel-Wahab et al. 2011, Jones et al. 2014). Combined three gene molecular analyses place this species extremely close to *A. velatisporus* (i.e. on a long branch within the clade of *A. velatisporus* isolates used in this study) in the

Annulatascaceae *sensu stricto* clade (**Fig. 7**), and it may represent a recently diverged species, or a morphological variation of *A. velatisporus*. Morphologically, its distinctive septation and blunt ascospore ends suggest it is a separate species. Further molecular data from fresh collections would help resolve the relationship of *A. hongkongensis* with respect to *A. velatisporus.*

Annulatascus incrustatus Ho Ph.D. thesis, 54. 1998.

Ascomata dark brown, coriaceous, immersed to superficial, scattered, 200–340 µm wide × 100–160 µm high (x̅ =270 × 130 µm, w:h = 2.1), subglobose to ellipsoidal. **Ascomal wall** brown, 15-30 µm thick, composed of 4–6 layers of brown compressed cells, of *textura angularis* in surface view. **Neck** dark brown, cylindrical 200–350 µm long × 50–90 µm wide, composed of 2 layers of brown globular cells, periphysate. **Paraphyses** hyaline, 2.5–4.5 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 100–140 µm long × 7–8 µm wide (x̅ = 132 × 7.5 µm, l:w = 17.6, n = 10). **Ascus base** pedicellate. **Annulus** J-, refractive, 4.5–5 µm long × 3.5–4 µm wide (x̅ = 4.75 × 3.75 µm, l:w = 1.3). **Ascospores** hyaline, ellipsoidal, thick walled, 1–septate, uniseriate to overlapping uniseriate, 16–21 µm long × 5–6 µm wide (x̅ = 19 × 5.5 µm, l:w = 3.5, n = 25), with 2 large lipid guttules, without appendages, with a thin mucilaginous sheath (Ho 1998).

HOLOTYPE: HKU(M) 4841.

Comments: *Annulatascus incrustatus* is informally published in Ho's Ph.D. thesis (1998) and is not a recognized name in Index Fungorum or MycoBank. Ho noted that the thesis would only have 5 copies made and that due to this limited distribution, the ICBN would not allow new species to be considered valid (*nomen invalidum*) until published independently and broadly distributed. As this thesis is freely available online at this time, this species is being included in this study for completeness, with all credit given to Ho (1998). The holotype was not examined in this study as Ho's thesis was only obtained near the completion of this manuscript. Thus only the protologue was used to assess this species morphologically. Though described as ellipsoidal, the ascospores appear to be fusiform in the micrographs. Morphologically, this species has features typical of *Annulatascus* and Annulatascaceae *sensu stricto* species, with the exception of having thick walled ascospores. This feature is commonly found in *Aquaticola*, *Cataractispora*, and *Torrentispora* species. *Aquaticola* does not include species with 1–septate, fusiform ascospores. The genus *Cataractispora* has species with ascospores with appendages and no sheaths, while *Torrentispora* has species with much longer cylindrical asci and thick walled ascospores that are definitively ellipsoidal. These differences make the generic placement of *A. incrustatus* a difficult task and formal publication and further research, especially molecular, is warranted for this species.

Annulatascus joannae K.M. Tsui, I.J. Hodgkiss & K.D. Hyde, Mycoscience 43: 484. 2002. **Fig. 13 A-H.** MycoBank 489213.

Ascomata black, coriaceous, immersed, perpendicular to surface, gregarious, 180–200 µm wide × 150–250 µm high ($\bar{x}$ =190 × 200 µm, w:h = .95), globose to subglobose. **Ascomal wall** hyaline and brown, several outer layers of melanized cells of *textura angularis* in surface view, with inner pale brown cells, 20 µm thick. **Neck** 150–200 µm long × 40–60 µm wide, periphysate. **Paraphyses** hyaline, 4 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, long cylindrical to broadly cylindrical, 8–spored, 150–200 µm long × 10–13 µm wide ($\bar{x}$ = 175 × 11.5 µm, l:w = 15.2) (**Fig. 13 D,E**). **Ascus base** pedicellate. **Annulus** J-, refractive, 2.5–4 µm long × 5–6.5 µm wide ($\bar{x}$ = 3.25 × 5.75 µm, l:w = .57). **Ascospores** hyaline, thick walled, ellipsoidal to fusiform, aseptate, overlapping uniseriate, 20–28 µm long × 9–12 µm wide ($\bar{x}$ = 24 × 10.5 µm, l:w = 2.3), 2–3 large lipid guttules, without appendages, with a thin gelatinous sheath (**Fig. 13 F-H**) (Tsui et al. 2002).

Fig. 13 A-H. *Annulatascus joannae* from the holotype **IFRD 8648**. A. Herbarium packet. B. Photographic plate included with dried specimen (Dianming Hu). C. Dried specimen. D. Fragmented asci. E. Young asci. F. Ascus apex. G. Ascospores. H. Ascospore illustrating thick walls. Bars: D-H = 20 µm.

International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code IFRD 8648
Fungi Name Annulatascus joannae (light blue pin)
Host on submerged wood
Collection Site Lam Tsuen River,Tai Po, N.T.Hong Kong
Collection Date December 1998
Collector Clement Tsui
Remarks Holotype, KM 164

2

Massarina thalassioided (km 41) orange
Massorina purpurascens (km 15t) dark blue
Helicomyces torquatus (km 298) purple

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224

by Dianming Hu

Fig. 13.

Specimens examined:

HOLOTYPE: CHINA. Hong Kong. Tai Po, Lam Tsuen River, on submerged wood, Dec 1998, *K.M. Tsui, KM 164,* **HKU(M) 12177.**

Known distribution: Brazil, China.

Comments: The holotype of *Annulatascus joannae* consisted of dried material and was in reasonable condition for examination and one permanent slide was prepared, examined, and returned to IFRD (**Fig. 13 A-C**). A photographic plate included with the holotype was composed of images that differ from those given in the protologue (**Fig. 13 B**) (Dianming Hu). *A. joannae* remains to be evaluated molecularly, perhaps from ex type culture HKUCC 4370. Morphologically, *A. joannae* fits well in Annulatascaceae and differs from *A. velatisporus* in having smaller ascomata of *textura epidermoidea* in surface view, a smaller annulus, and ellipsoidal-fusiform, aseptate ascospores with 2–3 large lipid guttules. A significant morphological feature which may preclude inclusion in the genus and family is that *A. joannae* possesses thick walled ascospores, a feature not found in any of the taxa included in the Annulatascaceae *sensu stricto* clade determined by the molecular analyses of this study. Thus molecular data is needed to determine the phylogenetic placement of *A. joannae*.

Annulatascus lacicola V.M. Ranghoo & K.D. Hyde, Ranghoo Ph.D. thesis, 69. 1998.

Ascomata dark brown to black, coriaceous, semi-immersed, gregarious, 220–250 µm wide × 225–250 µm high, (x̅ = 170 × 115 µm, w:h = 1.5), globose to subglobose. **Ascomal wall** dark brown, 20–25 µm thick, composed of several layers of light brown elongated cells, of *textura angularis* in surface view. **Neck** dark brown, 175–200 µm long × 30–40 µm wide, periphysate. **Paraphyses** hyaline, 5–7.5 µm wide at base × 75–88 µm long, tapering toward apex, septate, branched. **Asci** unitunicate, cylindrical, 8–spored, 75–100 µm long × 19–25 µm wide, (x̅ = 87.5 × 22 µm, l:w = 4). **Ascus base** pedicellate, 10 µm from *pars* *sporifera*. **Annulus** J-, refractive, 2.5–4 µm long × 3–4 µm wide ($\bar{x}$ = 3.25 × 3.5 µm, l:w = .93). **Ascospores** hyaline, fusiform, 2–3–septate, not constricted at septa, uniseriate to overlapping uniseriate, 35–38 µm long × 13–16 µm wide ($\bar{x}$ = 36.5 × 14.5 µm, l:w = 2.5), without appendages or sheath (Ranghoo 1998).

HOLOTYPE: MAURITIUS. Flacq, deep river, on submerged wood, Sep1997, *Y. Sanmukhiya and S. Sanmukhiya*, **HKU(M) 5204.**

Known distribution: Mauritius.

Comments: This species is not recognized in MycoBank or Index Fungorum. The holotype was not examined in this study as Ranghoo's thesis (1998) was only obtained near the completion of this manuscript. Thus only the protologue was used to assess this species morphologically. Ranghoo noted that the thesis would only have 5 copies made, and that due to this limited distribution the ICBN would not permit new species to be considered valid (*nomen invalidum*) until published independently and broadly distributed. As this thesis is freely available from Hong Kong University at this time, this species is being included in this study for completeness, with all credit given to Ranghoo (1998). Though described as cylindrical, the asci of *Annulatascus lacicola* appear to be clavate in the micrographs. The asci also do not match the width reported above and appear to be 9–11 µm wide using the bar size reported in the figure legend. In addition, the ascospores appear to have thin irregular granular sheaths. Without molecular data it is difficult to determine the phylogenetic placement of this species, though it does have the morphological features which agree well with the genus *Annulatascus*.

Annulatascus lacteus K.M. Tsui, I.J. Hodgkiss & K.D. Hyde, Mycoscience 43: 383. 2002. MycoBank 489214.

Ascomata milky to pale brown, membranous, superficial, parallel to perpendicular to substrate, gregarious, 140–200 µm wide × 100–130 µm high, ($\bar{x}$ = 170 × 115 µm, w:h = 1.5), subglobose to pyriform. **Ascomal wall** several layer of hyaline, compressed pseudoparenchymatous cells of *textura angularis*, 20 µm thick. **Neck** milky to pale brown, 50 µm long × 40–50 µm wide, periphysate. **Paraphyses** hyaline, 2–3 µm wide, tapering, septate. **Asci** unitunicate, cylindrical, 8–spored, 130–170 µm long × 9–10.5 µm wide, ($\bar{x}$ = 150 × 9.6 µm, l:w = 15.4). **Ascus base** pedicellate, tapering to small knob, 20 µm from *pars sporifera*. **Annulus** J-, refractive, 3–4 µm long × 4–5 µm wide ($\bar{x}$ = 3.4 × 4.5 µm, l:w = .78). **Ascospores** hyaline, fusiform, aseptate, uniseriate, 24–28 µm long × 6–8 µm wide ($\bar{x}$ = 26 × 7 µm, l:w = 3.7), multiguttulate, without appendages or sheath (Tsui et al. 2002).

HOLOTYPE: CHINA. Hong Kong, Tai Po, Lam Tsuen River, on submerged wood, Sep1996, *K.M. Tsui, KM313,* **HKU(M) 4623.**

Known distribution: China.

Comments: The holotype of *Annulatascus lacteus* was unavailable for examination and fresh specimens were not obtained in this study. *A. lacteus* is morphologically similar, especially in terms of ascus and ascospore size, to taxa such as *A. hongkongensis*, *A. velatisporus*, and *Annulusmagnus triseptatus*. With aseptate ascospores, *A. lacteus* is most similar to *A. velatisporus*, though it does not have verruculose ascospore ornamentation and lacks a sheath. With only the protologue as evidence, *A. lacteus* should remain in *Annulatascus* and Annulatascaceae until molecular data are obtained.

Annulatascus licualae J. Fröhl. & K.D. Hyde, Palm Microfungi. Fungal Diversity Research Series 3: 80. 2000. **Fig. 14 A-L.** MycoBank 482751.

Ascomata black, immersed, perpendicular to substrate, gregarious, 77.5–95 µm wide × 202–460 µm high, ($\bar{x}$ = 86.3 × 331 µm, w:h = 1.5), lenticular. **Ascomal wall** medium to dark brown, 2–4 layers of irregular, roughly globose cells, 3.3–7.5 µm diam. **Neck** black, 56–82 µm wide, periphysate. **Paraphyses** hyaline, 2.6–3.6 µm wide × 74–120 µm long, septate, very slightly tapering to rounded apices. **Asci** unitunicate, cylindrical, 8–spored, 89–125 µm long × 5.8–7.8 µm wide ($\bar{x}$ = 107 × 6.8 µm, l:w = 15.7) (**Fig. 14 F,G**). **Ascus base** pedicellate (**Fig. 14 K**). **Annulus** J-, lens-shaped, 1.2–2 µm long × 2.5–2.75 µm wide ($\bar{x}$ = 1.6 × 2.6 µm, l:w = .61) (**Fig. 14 H-J**). **Ascospores** hyaline, fusoid-rhomboid, aseptate, overlapping uniseriate, 15–17.8 µm long × 3.8–5 µm wide ($\bar{x}$ = 16.4 × 4.4 µm, l:w = 3.7), without appendages, with a thin, inconspicuous mucilaginous sheath (**Fig. 14 L**) (Fröhlich and Hyde 2000).

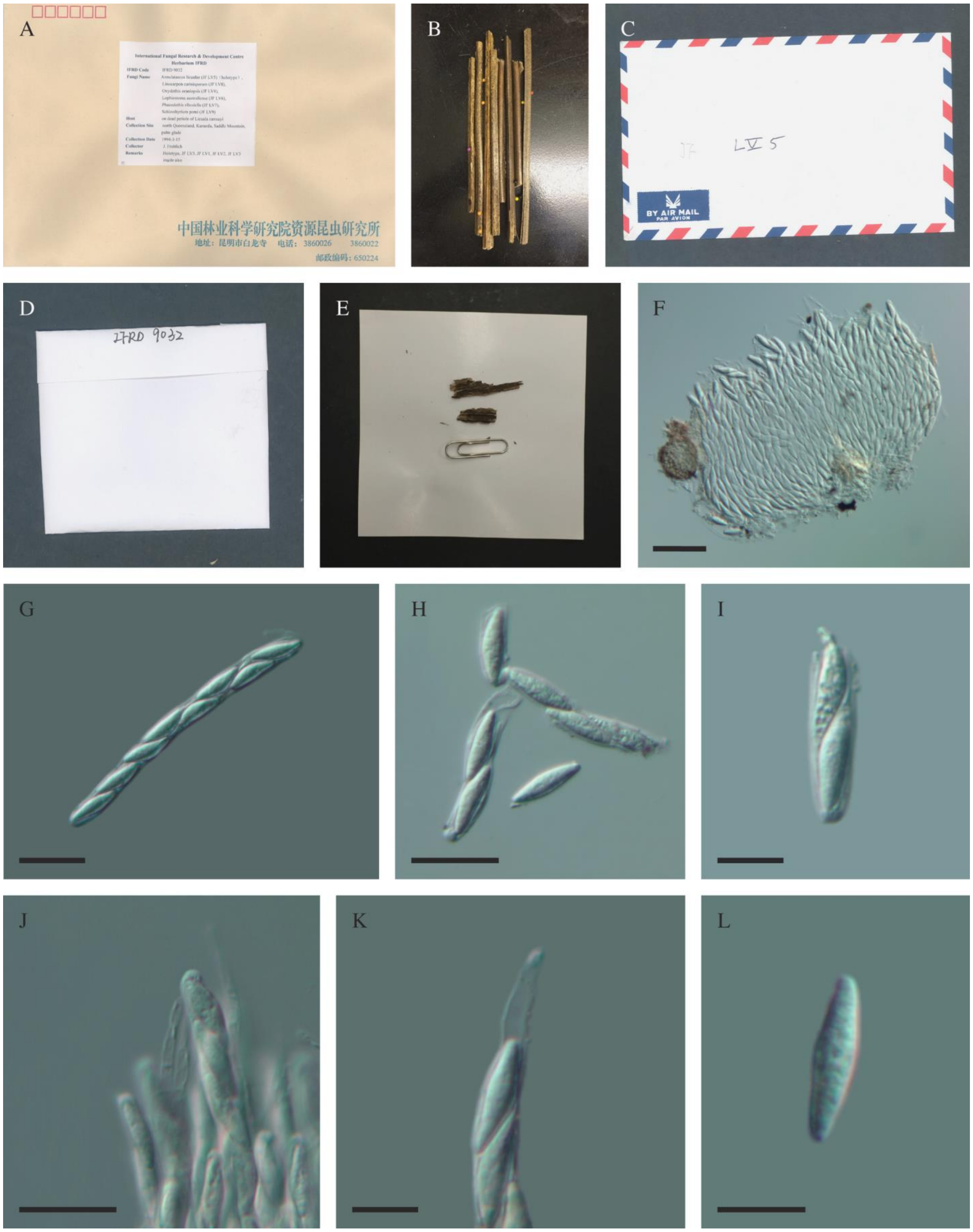
Specimens examined:

HOLOTYPE: AUSTRALIA. North Queensland, Kuranda, Saddle Mountain, palm glade, on dead petiole of *Licuala ramsayi*, Mar 1994, *J. Fröhlich, (HKU(M)JF LV 5),* **IFRD 9032.**

Known distribution: Australia.

Comments: The holotype of *Annulatascus licualae* is in good condition (**Fig. 14 A-E**). Two permanent slides were made in this study and returned with the holotype. Fresh specimens were not obtained in this study. One of the petioles in the first envelope had a mark where holotype material was to be found, but no ascomata were present. The second packet contained material from which multiple ascomata were removed. *A. licualae* as well as *A. citriosporus* were described from plant material found in a palm glade, but it was noted that the material was from a terrestrial habitat (Fröhlich and Hyde 2000, Hyde and Fröhlich 2003). Morphologically this species should remain in *Annulatascus* and Annulatascaceae until molecular evidence is evaluated. A feature which may be of discriminatory taxonomic note is the

Fig. 14 A-L. *Annulatascus licualae* from the holotype **IFRD 9032**. A. Herbarium packet. B. Dried petioles of the palm *Licuala ramsayi*. C. Mailing envelope of J. Fröhlich containing specimen JL LV5. D. IFRD herbarium packet containing dried material. E. Dried material. F. Asci and paraphyses. G. Ascus. H. Ascus apex and ascospores. I. Ascus apex. J. Ascus apex and paraphyses. K. Ascus base. L. Ascospore. Bars: F = 40 µm; G-H, J = 20 µm; I, K-L = 10 µm.


A
International Fungal Research & Development Centre
Herbarium IFRD
IFRD Code IFRD 9032
Fungi Name Annulatascus licualae (JF LV5) (holotype),
Linocarpon cariniisporum (JF LV8),
Oxydothis oraniopsis (JF LV4),
Lophiostoma australiense (JF LV6),
Phaeodothis ribesiella (JF LV7),
Schizothyrium pomi (JF LV9)
Host on dead petiole of Licuala ramsayi
Collection Site north Queensland, Kuranda, Saddle Mountain, palm glade
Collection Date 1994-3-15
Collector J. Fröhlich
Remarks Holotype, JF LV3, JF LV1, JF LV2, JF LV5 inside also
中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224
B
C
JF LV5
BY AIR MAIL
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IFRD 9032
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Fig. 14.

possession of a lens shaped ring (**Fig. 14 H-J**). This is inconsistent with the annulus description of type of the genus, *A. velatisporus*, which has a relatively massive bipartite ring. The terrestrial habit is also not consistent with the circumscription of Annulatascaceae. *A. citriosporus*, the other terrestrial taxon in this family (though recovered from freshwater in this study) is shown to be removed from the family in multi-gene analyses. *A. licualae* should be reevaluated with fresh material from the type location from which DNA can be obtained.

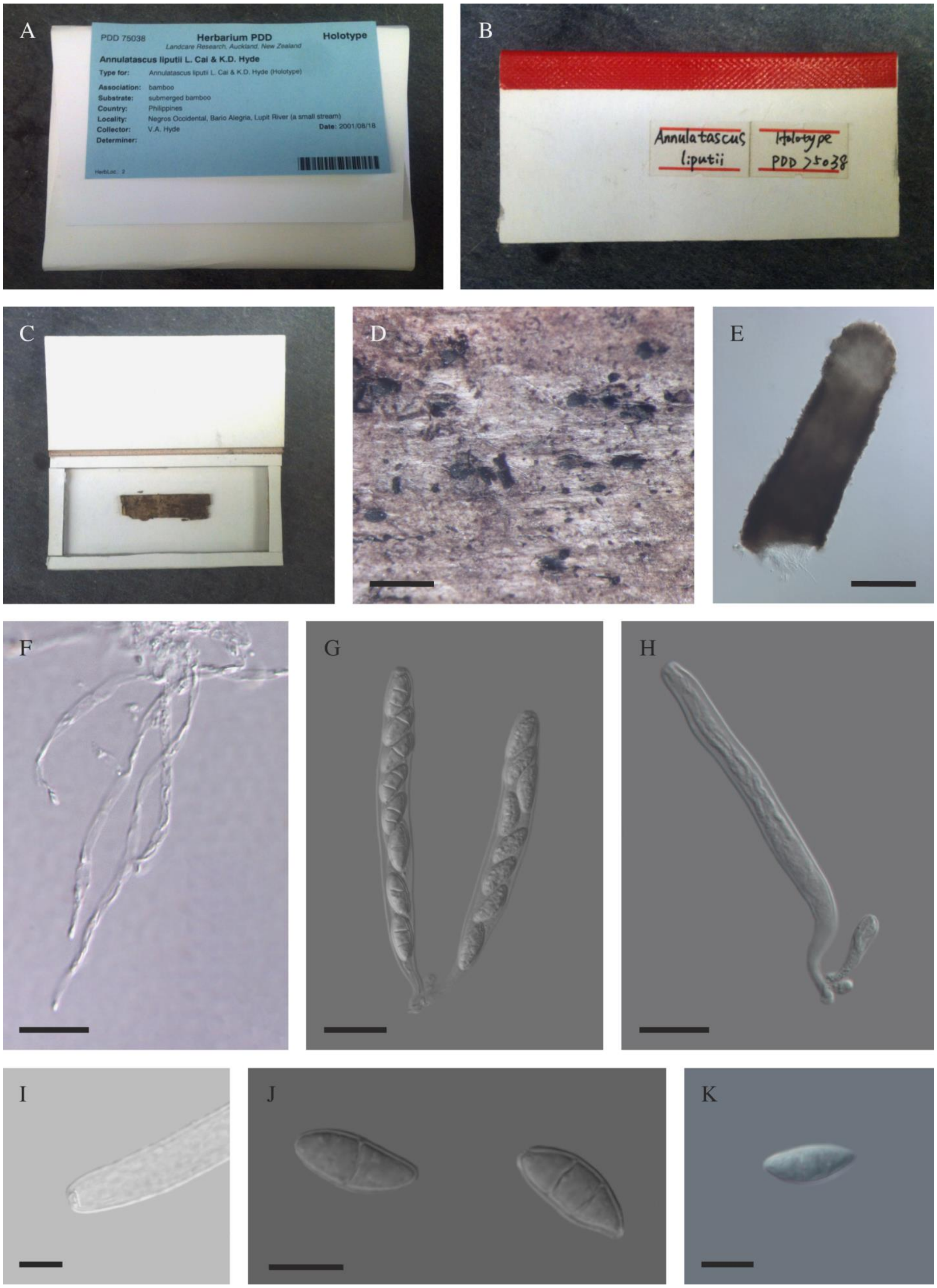
Annulatascus liputii L. Cai & K.D. Hyde, Mycotaxon 84: 256. 2002. **Fig. 15 A-K.** MycoBank 465316.

Ascomata dark brown to black, coriaceous, immersed to superficial, perpendicular to parallel to substrate surface, solitary or gregarious, 180–260 µm wide × 200–300 µm high (x̅ = 220 × 250 µm, w:h = .9), globose to subglobose (**Fig. 15 D**). **Ascomal wall** hyaline and brown, 15–30 µm thick, composed of dark brown-walled compressed cells laterally, with 3–4 inner layers of hyaline elongate cells, of *textura angularis* in surface view. **Neck** dark brown to black, 250–400 µm long × 45–70 µm wide, periphysate (**Fig. 15 E**). **Paraphyses** hyaline, 4–5 µm wide at base, tapering toward apex, septate (**Fig. 15 F**). **Asci** unitunicate, cylindrical, 8–spored, 130–187.5 µm long × 8.5–10 µm wide (x̅ = 159 × 9.3 µm, l:w = 17.2) (**Fig. 15 G,H**). **Ascus base** pedicellate, broadly tapering, 40 µm from *pars sporifera* (**Fig. 15 H**). **Annulus** J-, refractive, 2 µm long × 3.5 µm wide (x̅ = 2 × 3.5 µm, l:w = .57) (**Fig. 15 G-I**). **Ascospores** hyaline, fusiform, 0–2 septate, not constricted at septa, overlapping uniseriate, 15–22.5 µm long × 6.5–7.5 µm wide (x̅ = 18.75 × 7 µm, l:w = 2.7), without appendages, with a thin mucilaginous sheath (**Fig. 15 J,K**) (Cai et al. 2002).

Specimens examined:

HOLOTYPE: PHILIPPINES. Negros Occidental, Ario Alegria, Lupit River (a small stream), on submerged bamboo, 18 Aug 2001, *V.A. Hyde,* **PDD 75038.**

Fig. 15 A-K. *Annulatascus liputii* from the holotype **PDD 75038**. A. Herbarium packet. B. Slide holder. C. Dried bamboo. D. Ascomata on substrate. E. Neck illustrating periphyses. F. Long septate paraphyses. G. Asci. H. Young asci. I. Ascus apex. J. Ascospores. K. Ascospore. Bars: D = 500 µm; E = 50 µm; F-H = 20 µm; I-K = 10 µm.



PDD 75038 Herbarium PDD Holotype
Landcare Research, Auckland, New Zealand
Annulatascus liputii L. Cai & K.D. Hyde
Type for: Annulatascus liputii L. Cai & K.D. Hyde (Holotype)
Association: bamboo
Substrate: submerged bamboo
Country: Philippines
Locality: Negros Occidental, Bario Alegria, Lupit River (a small stream)
Collector: V.A. Hyde Date: 2001/08/18
Determiner:
HerbLoc.: 2

Annulatascus liputii
Holotype PDD 75038

Fig. 15.

Known Distribution: Costa Rica, Philippines.

Comments: The holotype of *Annulatascus liputii* consists of a small piece of dried bamboo, 3.5 × 1 cm, in a slide holder (**Fig. 15 A-C**). Ascomata are present and in good condition (**Fig. 15 D**). Two permanent slides were made, examined, and returned to PDD with the holotype. Fresh specimens were not obtained in this study. *A. liputii* has smaller ascomata and much smaller, 2–septate ascospores (**Fig. 15 J,K**) compared to the type of the genus, *A. velatisporus*, though both have thin mucilaginous ascospore sheaths (Cai et al. 2002). The ascus apical rings of *A. liputii* (**Fig. 15 G-I**) are also significantly smaller than those of *A. velatisporus*. These morphological differences and the polyphyletic nature of the genus as evidenced in this study, warrant the molecular evaluation of *A. liputii*. If viable, ex-type cultures HKUCC 2168 or 2169 should be used for obtaining molecular data. If not, a fresh specimen from the Lupit River in the Philippines should be obtained.

Annulatascus menglensis D.M. Hu, L. Cai & K.D. Hyde, Mycotaxon 120: 82. 2012. MycoBank 563810.

Ascomata black, coriaceous, superficial, perpendicular or parallel to surface of substrate, solitary, 120–150 µm wide × 180–200 µm high (x̅ = 190 × 135 µm, w:h = .71)**. Ascomal wall** of laterally compressed cells, outer layers dark brown, inner layers hyaline. **Neck** hyaline, 150–340 µm long × 40–60 µm wide, cylindrical, membranous, periphysate. **Paraphyses** hyaline, 5–6 µm wide at base, tapering toward apex, septate, smooth-walled. **Asci** unitunicate, cylindrical, 8–spored, 115–163 µm long × 10–13 µm wide (x̅ = 139 × 11.5 µm, l:w = 12). **Ascus base** pedicellate. **Annulus** J-, refractive, bipartite, 3 µm long × 4 µm wide (l:w = .75). **Ascospores** hyaline, fusiform, aseptate, uniseriate to overlapping uniseriate, 21–25 µm long × 8–10 µm wide (x̅ = 23 × 9 µm, l:w = 2.6), guttulate, without appendages or sheath (Hu et al. 2012).

HOLOTYPE: CHINA. Yunnan Province, Mengla, Wudaoban Stream, 21°24'N 101°36'E, 660m, 3 Apr 2009, *D.M. Hu,* **IFRDC 023-002.**

Comments: This species was published in the late stages of this study, and the holotype was not examined. Morphologically *Annulatascus menglensis* is currently best placed in *Annulatascus* and Annulatascaceae. However, it differs from the type of the genus, *A. velatisporus,* in having smaller ascomata, a hyaline neck, smaller asci, and a smaller annulus (Hu et al. 2012). Molecular data are needed to confirm generic and familial placement of this species, especially as no members of Annulatascaceae *sensu stricto* possess hyaline necks.

Annulatascus nilensis Abdel-Wahab and Abdel-Aziz, IMA Fungus 2(1): 3. 2011. MycoBank 517837.

Ascomata immersed, obpyriform, black, immersed, coriaceous, parallel to surface, solitary or gregarious, 400–600 µm wide × 220–280 µm high, (x̅ = 500 × 250 µm, w:h = 2.7). **Ascomal wall** hyaline and brown, 20–60 µm thick, of *textura angularis* in surface view, outer layer comprised of thick-walled flattened dark brown cells, encrusted with pigment outward, inner cells hyaline. **Neck** black, 240–360 µm long × 96–112 µm wide, periphysate. **Paraphyses** hyaline, 3–12 µm wide, tapering, rarely septate. **Asci** unitunicate, cylindrical, 8–spored, 260–400 µm long × 12–14 µm wide (x̅ = 330 × 13 µm, l:w = 25.4). **Ascus base** tapered, elongate base ending in small knob 140 µm from *pars sporifera*. **Annulus** J-, bipartite, 3–4 µm long × 5–6 µm wide (x̅ = 3.5 × 5.5 µm, l:w = .64). **Ascospores** hyaline, fusoid, 5–9–11 septate, constricted at septa, 32–52 µm long × 7–10 µm wide (x̅ = 42 × 8.5 µm, l:w = 5), without appendages, with a large, irregular, granular sheath (Abdel-Wahab and Abdel-Aziz 2011).

HOLOTYPE: EGYPT. Sohag, in the River Nile, on decayed submerged stems of *Phragmites australis*, Feb 2006, *A.E. Abdel-Aziz,* **IMI 397966.**

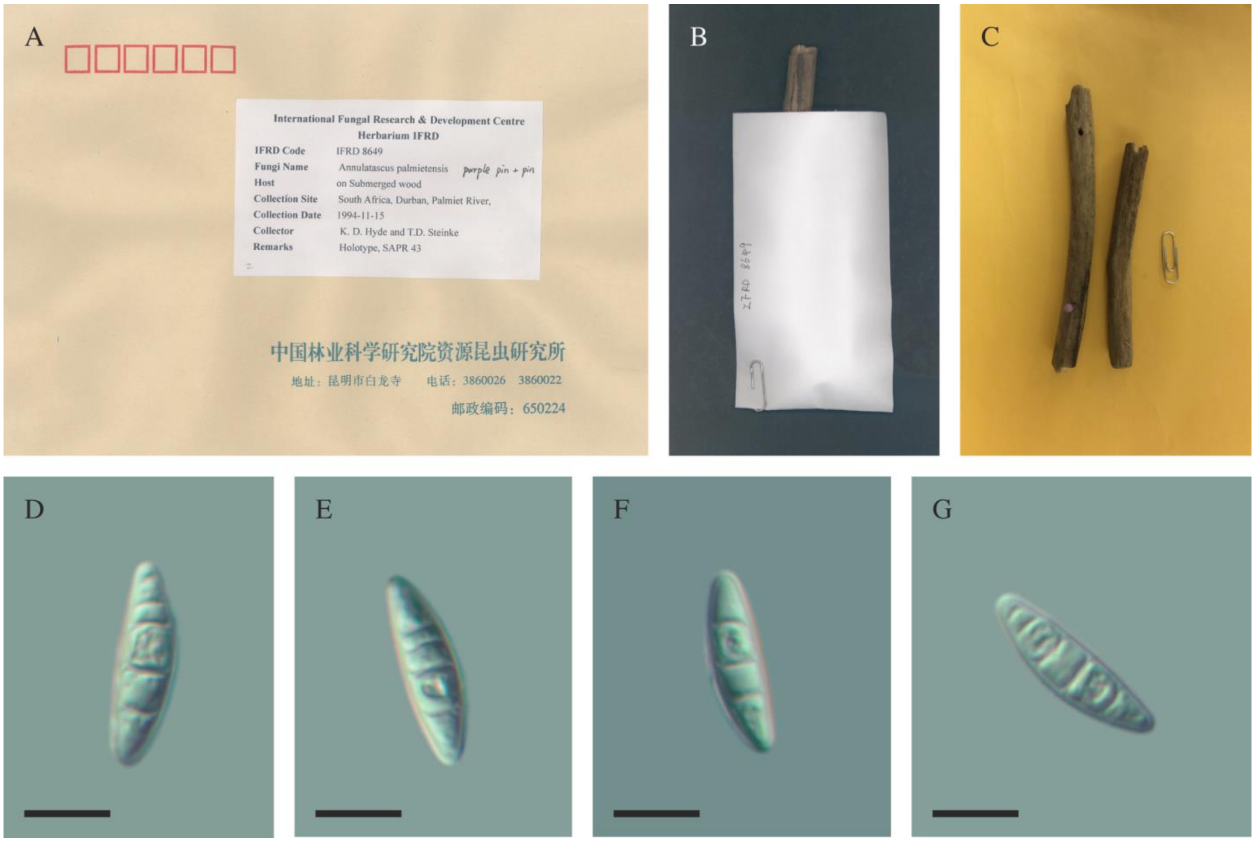
Known distribution: Egypt.

Comments: This species of Annulatascaceae is the first that possesses ascospores with septation greater than 5. The holotype of *Annulatascus nilensis* was not examined, as it was being processed at IMI. An ex type culture (MF 808) is also of uncertain location. Using the 28S sequence deposited in GenBank, this fungus changes position in trees generated during analyses and may depend on taxon sampling. *A. nilensis* caused a polytomy in combined 18S and 28S analyses (**Fig. 6**) as well as in preliminary molecular analyses performed in the course of this work (results not shown). *A. nilensis* clusters in the Annulatascaceae *sensu stricto* clade, though basally with only Bayesian support in combined three gene analyses (**Fig.7**), or as a long branch with only Bayesian support in 28S analyses (**Fig. 4**). Jones et al. (2014) noted that this species is distantly related to *A. velatisporus*, the type of the genus, though in the Annulatascaceae *sensu stricto* clade. Indeed, *A. nilensis* differs morphologically from *A. velatisporus* in having larger ascomata, longer asci with extremely long pedicels, shorter annuli, and longer up to 11–septate ascospores. These differences suggest that a new genus for this species may be necessary.

Annulatascus palmietensis K.D. Hyde, Goh, & T.D. Steinke, South African Journal of Botany 64: 151. 1998. **Fig. 16 A-G.** MycoBank 446190.

Ascomata dark brown to black, coriaceous, immersed, semi-immersed to superficial, perpendicular to surface, solitary to gregarious, 150–440 µm wide, ellipsoidal or subglobose. **Ascomal wall** dark brown to black, consisting of several rows of brown irregular angular cells, 20 µm thick. **Neck** dark brown to black, 200 µm long × 70 µm wide, periphysate. **Paraphyses** hyaline, 3 µm wide at base, tapering toward apex, septate, sparse. **Asci** unitunicate, cylindrical, 8–spored, 98–142 µm long × 7–10.5 µm wide ($\bar{x}$ = 120 × 8.8 µm, l:w = 13.7). **Ascus base** pedicellate, 20 µm from *pars sporifera*. **Annulus** J-, relatively massive, 3–4 µm long × 4–4.5 µm wide ($\bar{x}$ = 3.5 × 4.25 µm, l:w = .82). **Ascospores** hyaline, short fusiform, 3–septate, not constricted at septa, uniseriate to biseriate, 20–26 µm long × 6–7 µm wide ($\bar{x}$ = 23 × 6.5 µm, l:w = 3.5), guttulate, no appendages or sheath (**Fig. 16 D-G**) (Hyde et al. 1998).

Fig. 16 A-G. *Annulatascus palmietensis* from the holotype **IFRD 8649**. A. Herbarium packet. B. Envelope containing dried substrate. C. Dried substrate. D-G. Ascospores. Bars: D-G = 10 µm.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8649
Fungi Name	Annulatascus palmietensis purple pin + pin
Host	on Submerged wood
Collection Site	South Africa, Durban, Palmiet River,
Collection Date	1994-11-15
Collector	K. D. Hyde and T.D. Steinke
Remarks	Holotype, SAPR 43

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

Fig. 16.

Specimens examined:

HOLOTYPE: SOUTH AFRICA. Durban, Palmiet River, on submerged wood, Nov 1994, *K.D. Hyde and T.D. Steinke, SAPR 43, (HKU(M) 2206),* **IFRD 8649.**

Known distribution: South Africa.

Comments: The holotype of *Annulatascus palmietensis* is in fair condition with only ascospores preserved (**Fig. 16 A-C**). Two permanent slides were prepared, examined and returned with the holotype material to IFRD. Fresh material was not obtained in this study and therefore DNA was not obtained. On holotype material asci were not observed. Only fragments of ascomata and ascospores remain in the dried material. The protologue notes a resemblance to *A. velatisporus* but that *A. palmietensis* differs in having smaller, differently shaped (blunt ascospore ends), consistently 3–septate ascospores with no gelatinous sheath (**Fig. 16 D-G**) (Hyde et al. 1998). Fresh material should be collected from the Palmiet River, South Africa to obtain DNA and assess the relationship of *A. palmietensis* to *Annulatascus* and Annulatascaceae *sensu stricto*.

Annulatascus triseptatus S.W. Wong, K.D. Hyde, & E.B.G. Jones, Mycological Research 103(5): 563. 1999. **Fig. 17 A-G.** MycoBank 459600.

Ascomata black, coriaceous, immersed to semi-immersed, perpendicular to surface, gregarious, 222–353 µm wide, globose to subglobose. **Ascomal wall** hyaline and brown, outer layers brown of *textura epidermoidea* in surface view, inner layer of compressed hyaline cells. **Neck** black with hyaline apex, 110 µm long. **Paraphyses** hyaline, 3 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 140–218 µm long × 9–14 µm wide ($\bar{x}$ = 179 × 11.5 µm, l:w = 15.6) (**Fig. 17 D,E**). **Ascus base** pedicellate, broadly tapering, 20 µm from *pars sporifera* (**Fig. 17 G**). **Annulus** J-, bipartite, 3 µm long × 5.5 µm wide ($\bar{x}$ = 3 × 5.5 µm, l:w = .55) (**Fig. 17 F**). **Ascospores** hyaline, fusiform, 3–septate,

Fig. 17 A-G. *Annulatascus triseptatus* from the holotype **IFRD 8650**. A. Herbarium packet. B. Envelope containing dried material. C. Dried substrate. D-E. Asci. F. Ascus apex. G. Ascus base. Bars: D-G = 20 µm.

International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8650
Fungi Name	Annulatascus triseptatus
Host	on Submerged wood
Collection Site	Brunei
Collection Date	1994-4-11
Collector	K. D. Hyde and S. W. Wong
Remarks	Holotype

A small quantity. Please be careful

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

HKU(M) 3129
IFRD: 8650

Fig. 17.

slightly constricted at septa, overlapping uniseriate, 18–33 µm long × 6–12 µm wide ($\overline{x}$ = 25.5 × 8 µm, l:w = 2.8), multiguttulate, without appendages, with a thin sheath (Wong et al. 1999).

Specimens examined:

HOLOTYPE: BRUNEI. Temburong, Kuala Belalong Field Studies Centre, Sungai Anak, on submerged wood, Feb 1994, *K.D. Hyde and S.W. Wong, (HKU(M) 3129),* **IFRD 8650.**

Known distribution: Australia, Brunei, Hong Kong, Perú.

Comments: The holotype material of *Annulatascus triseptatus* was scant (**Fig. 17 A-C**). One permanent slide made in this study was examined and returned with the holotype to IFRD. Ascospores could not be freed from the asci and therefore only images of asci with ascospores inside were obtained (**Fig. 17 D-G**). *A. triseptatus* differs from *A. velatisporus* in that the ascospores are consistently three septate. In this study, a 3–septate species, PE0160, was initially diagnosed as *A. triseptatus* due to constriction at the ascospore septa, but molecular analyses place 3–septate (PE0160) and aseptate (PE0011) specimens as conspecific, i.e. as *A. velatisporus*. Campbell and Shearer (2004) re-evaluated *A. triseptatus* and created the new combination *Annulusmagnus triseptatus*. However, *Annulusmagnus* ascospores have blunter ends and ascospores with light brown pigmented septa, while the original publication of *A. triseptatus* describes completely hyaline ascospores. These differences make the synonymy questionable and thus *A. triseptatus* is treated as a legitimate and distinct taxon due to morphology in addition to the geographical separation of *A. triseptatus* from *An. triseptatus* (tropics vs. temperate locales). As no type culture has been reported, fresh material of *A. triseptatus* should be collected from Brunei and DNA extracted to resolve the relationship to *An. triseptatus*. Molecular data would also resolve the relationship of *A. triseptatus* with respect to Annulatascaceae *sensu stricto*.

Annulatascus tropicalis Ranghoo & K.D. Hyde, Mycoscience 43: 387. 2002. **Fig. 18 A-L.** MycoBank 489356.

= *Annulatascus longispora* V.M. Ranghoo and K.D. Hyde, Ranghoo Ph.D. thesis, 67. 1998.

Ascomata black, coriaceous, semi-immersed, gregarious, 263–275 µm wide × 250–263 µm high ($\bar{x}$ = 269 × 257 µm, w:h = 1.1), globose to subglobose (**Fig. 18 D**). **Ascomal wall** hyaline and brown, 20–30 µm thick, comprised of 5–7 outer layers of dark brown cells and 2–3 inner layers of hyaline cells, of *textura angularis* in surface view. **Neck** black, 100-150 µm long × 30–50 µm wide, periphysate. **Paraphyses** hyaline, 6–7.5 µm wide at base × 163–200 µm long, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 190–255 µm long × 12–18 µm wide ($\bar{x}$ = 222.5 × 15 µm, l:w = 14.8) (**Fig. 18 E**). **Ascus base** short, pedicellate, with a spike-like tail, 10 µm from *pars sporifera* (**Fig. 18 E,I,J**). **Annulus** J-, large, 3–5 µm long × 3–5 µm wide ($\bar{x}$ = 4 × 4 µm, l:w = 1) (**Fig. 18 F-H**). **Ascospores** hyaline, fusiform, 1–3 septate, not constricted at septa, uniseriate to overlapping uniseriate, 42.5–52.5 µm long × 7.5–10 µm wide ($\bar{x}$ = 47.5 × 8.8 µm, l:w = 5.4), multiguttulate, without appendages or sheath (**Fig. 18 K,L**) (Tsui et al. 2002).

Specimens examined:

HOLOTYPE: CHINA. Hong Kong, Tai Po, Plover Creek Reservoir, on submerged wood, Jan 1997, *V.M. Ranghoo, (HKU(M) 5253),* **IFRD 9036.**

Known distribution: China.

Comments: The holotype of *Annulatascus tropicalis* is in good condition, with many ascomata on the substrate (**Fig. 18 A-D**). Two permanent slides were made, examined and returned with the holotype to IFRD. Holotype material exhibited asci with spike-like tails (**Fig. 18 E,I,J**) which was not noted in the protologue, but are found in some Annulatascaceae-like taxa, such as *Vertexicola caudatus* and *Chaetorostrum quincemilense*. Fresh specimens were not obtained in this study and thus no new

Fig. 18 A-L. *Annulatascus tropicalis* from the holotype **IFRD 9036**. A. Herbarium packet. B. Dried substrate. C. Photographic plate included with dried specimen (Dianming Hu). D. Ascomata on surface of substrate. E. Ascus. F-H. Ascus apices. I-J. Ascus bases illustrating spike-like tails. K. Ascospores. L. Ascospore. Bars: D = 500 µm; E-L = 20 µm.

International Fungal Research & Development Centre
Herbarium IFRD
IFRD Code IFRD 9036
Fungi Name Annulatascus tropicalis dark blue
Host on Submerged wood
Collection Site China. Hong Kong, Tai Po, Plover Cove Reservoir
Collection Date January 1997
Collector V.M. Ranghoo
Remarks Holotype

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224

Fig. 18.

molecular data were obtained. This species was originally described under the name *A. longispora* in Ranghoo's Ph.D. thesis (1998) and an ITS sequence is deposited in GenBank under that name. In ITS analyses, *A. tropicalis* clusters with *Aquaticola miniguttulata* and a new *Aquaticola*-like species (PE0316) with strong Bayesian support, but not in a clade with *A. velatisporus*, the type of the genus and family (**Fig. 4**). Its placement in *Annulatascus* was largely based on possession of a large J- refractive ascus apical ring, but it ascospores are much longer, thinner, have more acute apices, and lack a sheath (**Fig. 18**) compared to *A. velatisporus*. No other species in Annulatascaceae *sensu stricto* possess these attributes, and thus the generic and familial placement of *A. tropicalis* remains uncertain. Further molecular evidence (28S) is needed to assess the phylogenetic placement of *A. tropicalis*.

Annulatascus velatisporus K.D. Hyde, Australian Systematic Biology 5: 118. 1992. **Fig. 19 A-H, Fig. 20 A-K.** MycoBank 804603. (*Annulatascus velatispora* MycoBank 355478, orthographic variant).

Ascomata black, coriaceous, immersed, semi-immersed, to superficial, perpendicular to surface, solitary or gregarious, 260–410 µm wide, globose to subglobose (**Fig. 20 A**). **Ascomal wall** dark, brown, hyaline, outer layer of dark indistinguishable cells, middle layer of brown cells of *textura angularis*, inner layer of compressed hyaline cells of *textura epidermoidea*. **Neck** black, 384 µm long × 140 µm wide, periphysate. **Paraphyses** hyaline, wide, tapering toward apex, septate (**Fig. 20 E**). **Asci** unitunicate, cylindrical, 8–spored, 220–290 µm long × 12–18 µm wide (x̅ = 220 × 15 µm, l:w = 14.7) (**Fig. 20 B**). **Ascus base** pedicellate, broadly tapering, 20 µm from *pars sporifera*. **Annulus** J-, bipartite, 7–8 µm long × 4–5 µm wide (x̅ = 47.5 × 8.75 µm, l:w = 5.4) (**Fig. 19 D,F,G; Fig. 20 C,D**). **Ascospores** hyaline, fusiform, 0–3 septate, not constricted at septa, with verruculose wall ornamentation, uniseriate, 21–30 µm long × 8–11 µm wide (x̅ = 25.5 × 9.5 µm, l:w = 2.7), multiguttulate, without appendages, with a thin, irregular expanding to wide sheath (**Fig. 19 E,H; Fig. 20 F-K**) (Hyde 1992).

Specimens examined:

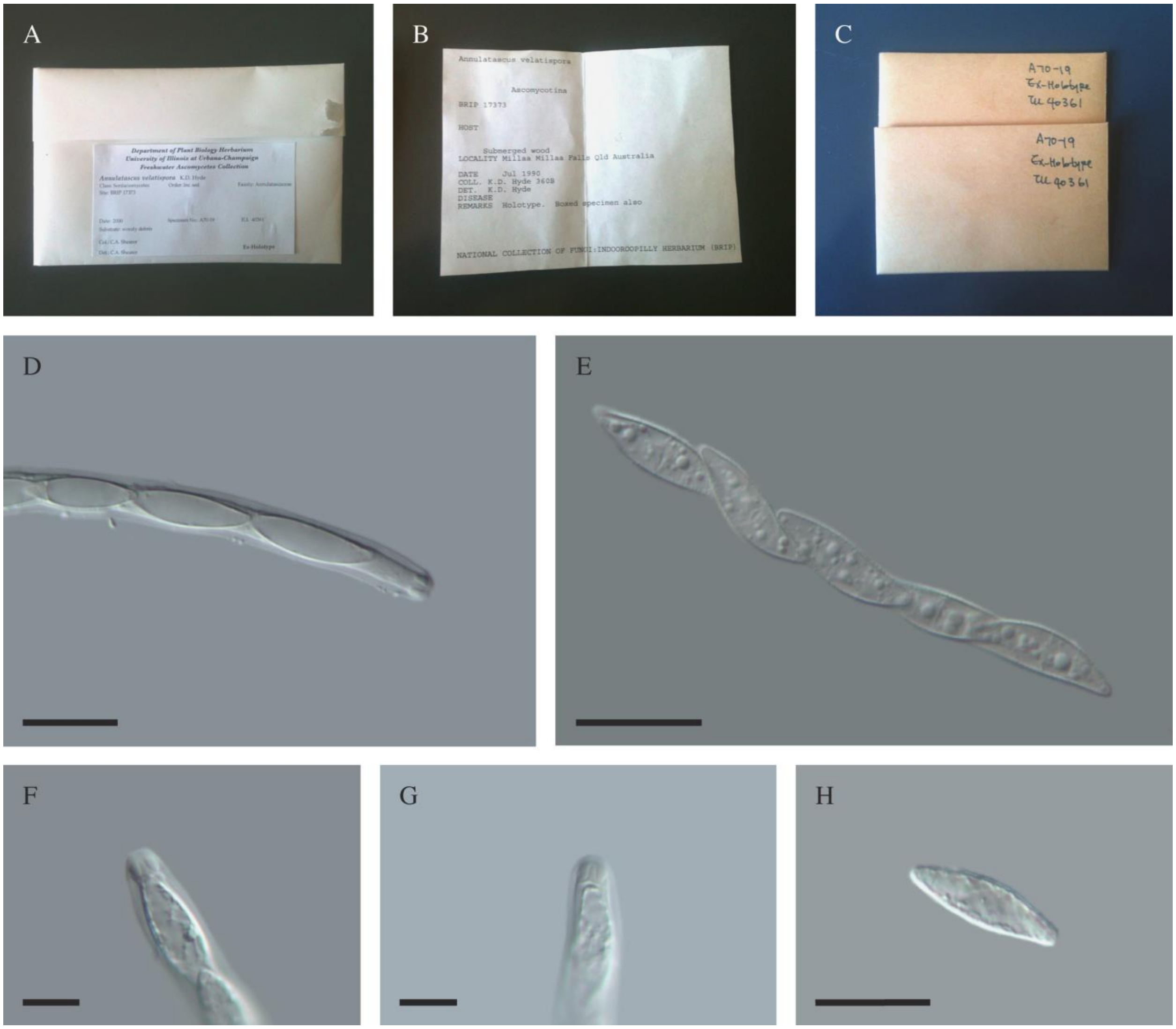
HOLOTYPE. AUSTRALIA. North Queensland, Millaa Millaa Falls, on submerged wood, Jul 1990, *K.D. Hyde 360B, ILL 40361 (ex-holotype),* **BRIP 1737**3.

COSTA RICA. Heredia, La Selva Biological Station, Arboleda Bridge, 10°25’48”N, 84°0’34”W, on partially decorticated woody debris, water 25 °C, pH 6, 20 May 2000, *Anderson J. and Wulffen R., A70-18, ILL 40360;* Rio Puerto Viejo, 10°25’48”N, 84°0’17”W, on submerged decorticated woody debris, water 23 °C, pH 5.5, 20 May 2000, *Anderson J. and Wulffen R., A70-20, ILL 40362;* Rio Saltito, 10°25’30”N, 84°0’36”W, on submerged decorticated woody debris, water 25 °C, pH 5.5, 17 May 2000, *Anderson J. and Wulffen R., A70-22, ILL 40367.* Limon, Las Cruces Biological Station, La Toma II, 8°47’5”N, 82°57’40”W, on submerged woody debris, water 19 °C, pH 5, 3 Jan 2006, *Salazar M., AF219-4, ILL 40756.* ECUADOR. Mirador I, on submerged decorticated woody debris, water 24.5 °C, pH 5.5-6, 2 Apr 2004, *J. Dalling, A70-24, ILL 40365;* Perú 1 stream, on submerged decorticated woody debris, water 24.5 °C, pH 5-5.5, 4 Apr 2004, *J. Dalling, A70-27, ILL 40368;* Napo, Yasuni National Park, Laguna I, 0°40’09”S, 76°24’04”W, on submerged woody debris, water 29 °C, pH 5, 29 Sep 2005, *A. Ferrer, AF219-1, AF219-2, ILL 40754, ILL 40755.* FRENCH GUIANA. Saül, Les Eaux Claires Resort, Crique Eaux Claires, 30°39’41”N, 53°13’17’W, on submerged plant debris, 28 Sep 1995, *J. L. Crane, A70-12, ILL 40355.*PERU. Cusco, Camanti, Quincemil, stream at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 22.3 °C, pH 7.5, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0011-3*; Stream at Quincemil Trail 2, 13°13’31.0434”S, 70°45’10.6194”W, 653m, on submerged woody debris, water 24.0 °C, pH 7.4, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0011-8*; Stream across road to Cusco from Quincemil, 13°17’7.008”S, 70°47’13.632”W, 737m, on submerged woody debris, water 22.0 °C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0011-13;* Madre de Dios, CICRA, stream at Trail 28, 12°34’02.81”S, 70°05’42.96”W, 272m, on submerged decorticated wood, water 22.7 °C, pH 5.9, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0011-1;* Rio Amigos, 12°34’3.2514”S, 70°4’55.92”W, 218m, on submerged woody debris, water 25.3 °C, pH 7.9, 23 May 2010, *Zelski S.E. and H.A. Raja, PE0011-2;* CICRA, Pozo Don Pedro, palm swamp (aguajal) at end of Trail 17, 12°33’34.27”S, 70°06’38”W, 243m, on submerged palm debris, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0011-4, PE0011-16;* CICRA, seasonally flooded oxbow lake (cocha seca) at Trail 14, 12°34’14.7”S, 70°05’23.69”W, 241m, on submerged woody debris, water 23.0 °C, pH 6.7, 23 May 2010, *Zelski S.E. and H.A. Raja, PE0011-54;* CICRA, stream at Trail 28, 12°34’02.81”S, 70°05’42.96”W, 272m, on submerged decorticated woody debris, water 23.3 °C, pH 6.8, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0011-9;* CICRA, Stream at Trail 20, 12°33’25.22”S, 70°05’59.89”W, 288m, on submerged woody debris, water 23.5 °C, pH 6.6, 9 Sep 2010, *Zelski S.E. and H.A. Raja, PE0011-10;* Rio Amigos, 12°34’13.008”S, 70°41’14.7714”W, 218m, on submerged bamboo, 25.3 °C, pH 7.9, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0011-34;* Palm swamp (agaujal) off the Interoceanic Highway outside of Puerto Maldonado, 12°42’48.0954”S, 69°28’11.28”W, 239m, on submerged woody debris, water 23.3 °C, pH 5.9, 20 May 2010, *Zelski S.E. and H.A. Raja, PE0011-36;* Semi-aquatic habitat along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.2 °C, pH 6.8, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0160-3;* River at end of Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.6 °C, pH 8.3, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0160-10;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 20.5 °C, pH 7.2, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0160-8;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 21.4° C, pH 7.5, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0160-1;* 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0160-17;* Stream flowing into backwater of river at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.2 °C, pH 7.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0160-16;* Stream with red algae along Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 21.8 °C, pH 7.2, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0160-14;* Madre de Dios, CICRA, semi-aquatic habitat on Trail 1, 12°34'06.52"S, 70°06'04.57"W, 263m, on submerged palm debris, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0160-13;* CICRA, Stream at Trail 10, 12°37'48.95"S, 70°05'23.69"W, 287m, on submerged woody debris, water 22.3 °C, pH 5.6, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0160-2;* CICRA, Pozo Don Pedro, palm swamp (aguajal) at end of Trail 17, 12°33'34.27"S, 70°06'38"W, 243m, on submerged woody debris, water 31.7 °C, pH 6.8, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0160-6;* CICRA, Rio Amigos, 12°33'25.22"S, 70°05'59.89"W, 288m, on submerged woody debris, water 31.4 °C, pH 8, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0160-7;* CICRA, Rio Amigos, 12°34'13.008"S, 70°41'14.7714"W, 218m, on submerged woody debris, water 31.4 °C, pH 8, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0160-18;* CICRA, Rio Amigos, 12°34'0.336"S, 70°4'59.052"W, 218m, on submerged woody debris, water 31.4 °C, pH 8, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0160-11;* CICRA, Pozo Don Pedro, palm swamp (aguajal) at end of Trail 17, 12°33'34.27"S, 70°06'38"W, 243m, on submerged woody debris, water 25.4 °C, pH 7.9, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0160-9.* THAILAND. Chiang Mai, Tham Rusee Nature Trail, 18°48'24.4794"N, 98°54'38.3754"E, 1149m, on submerged woody debris, water 22.4° C, pH 7.2, 6 Jun 2012, *Zelski S.E., TH0011-1*; USA. Florida, Blackwater River State Forest, Horns Creek Swamp, 30°46'31"N, 86°54'43"W, on submerged decorticated woody debris, water 30 °C, pH 6, 10 Jul 2004, *C. Brown. and H.A. Raja, F0002-1;* Big Coldwater Creek East Fork, 30°50'47"N, 86°59'02"W, on submerged woody debris, water 25 °C, pH 5-5.5, 7 Jul 2006, *H.A. Raja and J.L. Crane, F0002-14;* Hawaii, Maui, 13 Jan 1994, on submerged woody debris, *Charles, A70-6, ILL 40335;* Illinois, Johnson County, Cache River at foot Bridge to Heron Pond, 37°21'07"N, 88°55'27"W, on submerged decorticated wood, 3 Oct 1992, *J.L. Crane and C.A. Shearer, A70-1, ILL 40344;* Union County, spring at La Rue, Pine Hills Ecological Area, 37°32'40.4"N, 89°26'29.1"W, on submerged partially decorticated wood, water 3.5 °C, pH 6, 25 Jun 1994, *J.L Crane. and C.A. Shearer, A70-9, ILL 40352;* North Carolina, Coheeta Lab Stream, 35°04'N, 83°26'W, 685m, on submerged partially decorticated wood, 2000, *C.A.*

Shearer, A70-25, ILL 40366; GSMNP, Tapoco, Cheoah River, 35°26’18”N, 83°55’07”W, 390m, on submerged decorticated wood, water 26 °C, pH 5, 18 Jul 2000, *J. Campbell, A70-26, ILL 40367;* GSMNP, still water leading to Fontana Lake, 35°26’58”N, 83°31’38”W, 518m, on submerged woody debris, water 29 °C, pH 5.5, 16 Jul 2000, *J. Campbell, R47-1*; Santeetlah Lake, 35°22’26”N, 83°52’09”W, 590m, on submerged woody debris, water 28 °C, pH 5, 18 Jul 2000, *J. Campbell, R47-2;* Chilhowee Lake, 35°32’45”N, 83°59’32”W, 267m, on submerged decorticated wood, water 34 °C, pH 5, 19 Jul 2000, *J. Campbell, A70-21, ILL 40363;* Pigeon River, 35°37’27”N, 83°25’01”W, 1380m, on submerged woody debris, water 28 °C, pH 4.5, 20 Jul 2000, *J. Campbell, R47-3*; GSMNP, Cades Cove, Gourley Pond, 35°35’36”N, 83°47’21”W, 575m, on submerged woody debris, 12 Jan 2005, *A.N. Miller and H.A. Raja, R47-4;* Tennessee, Blount County, Laurel Creek, 35°38’32”N, 83°42’55”W, 415m, on submerged woody debris, 25 Sep 1999, *C.A. Shearer, A70-15, ILL 40358;* GSMNP, Cades Cove, 35°35’00”N, 83°47’00”W, water 9.5 °C, pH 6.4, on submerged corticated woody debris, 10 Dec 1999, *C.A. Shearer and Hurley, A70-16, ILL 40349;* GSMNP, Mill Creek, Cades Cove Loop Road, 35°00’00”N, 83°51’00”W, water 11 °C, pH 4.5-5, on submerged partially decorticated wood, 10 Dec 1999, *C.A. Shearer and Hurley, A70-17, ILL 40359;* Wisconsin, Iron County, Manitowish River at junction with Rt. 51, Manitowish waters, 46°8’14”N, 89°54’42.4”W, 485m, on submerged woody debris, 2 May 1997, *P.M. Fallah, A70-14, ILL 40357.* VENEZUELA. Barinas, Caño Yaure at junction with Barinas-PTA De Piedra Road, Santa Barbara, 7°42’9.6”N, 71°17’39.6”W, on submerged woody debris, *J.D. Schoknecht, J.L. Crane and D.C. Taphorn, A70*-3, *ILL 40346;* Tachira, Branch of Quebrada, La Pulida, Road to La Fundación, José De Bolivar, 7°43’22.8”N, 71°57’52.2”W, on submerged woody debris, 26 Jan 1993, *J.D. Schoknecht, J.L. Crane and D.C. Taphorn, A70-2, ILL 40345;* Trujillo, Distrito Muñoz, Laguna de Aguas Negros along road to Bocano, 9°18’N, 70°09’W, 2100m, on submerged woody debris, water 20.0 °C, pH 4.5, 19 Jan 1993, *Shearer C.A., A70-4, ILL 40347.*

Known distribution: Australia, Costa Rica, Ecuador, French Guiana, Perú, Thailand, USA, Venezuela.

Fig. 19 A-H. *Annulatascus velatisporus* from the holotype **BRIP 17373**. A. Ex-type herbarium packet. B. BRIP documentation. C. Contents of herbarium packet (2 slides). D. Ascus apex. E. Ascospores illustrating verruculose ornamentation. F-G. Ascus apices. H. Ascospore. Bars: D-E, H = 20 µm; F-G = 10 µm.



Department of Plant Biology Herbarium
University of Illinois at Urbana-Champaign
Freshwater Ascomycetes Collection

Annulatascus velatispora K.D. Hyde

Site: BRIP 17373

Date: 2000

Specimen No.: A70-19

Col.: C.A. Shearer

Det.: C.A. Shearer

Ex-Holotype

```
Annulatascus velatispora

              Ascomycotina

BRIP 17373

HOST

      Submerged wood
LOCALITY Millaa Millaa Falls Qld Australia

DATE      Jul 1990
COLL. K.D. Hyde 360B
DET.  K.D. Hyde
DISEASE
REMARKS  Holotype.  Boxed specimen also

NATIONAL COLLECTION OF FUNGI:INDOOROOPILLY HERBARIUM (BRIP)
```

A70-19
Ex-Holotype
ILL 40361

A70-19
Ex-Holotype
ILL 40361

Fig. 19.

Fig. 20 A-K. *Annulatascus velatisporus* from specimen PE0011. 1. Ascomata on substrate. B. Ascus. C-D. Ascus apices. E. Long septate paraphyses. F-G. Ascospores. H. Ascospore illustrating gelatinous sheath. I-J. Gelatinous sheath stained with nigrosin. K. Ascospore with three septa. Bars: A = 100 µm; B = 40 µm; C-K = 20 µm.

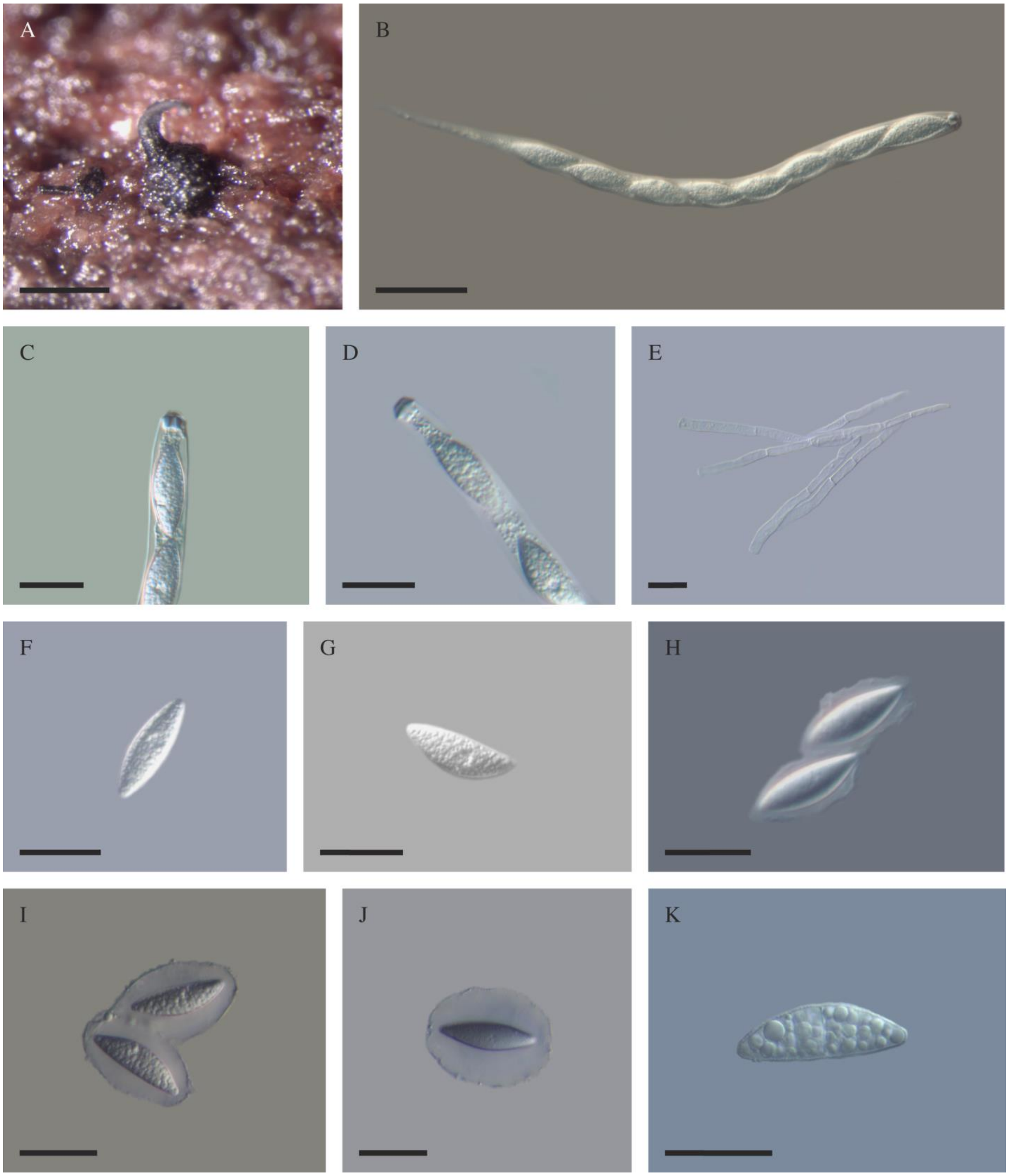


Fig. 20.

Comments: Examination of ex-holotype material of *Annulatascus velatisporus* was performed as a proxy for the holotype (**Fig. 19 A-C**). Fresh specimens were obtained and sequenced from Perú, Thailand and the USA in this study, and all grouped together with good support in molecular analyses with previously deposited sequences from other locations. This species is the most geographically widespread taxon in this study and also the type species of Annulatascaceae. *Annulatascus velatisporus* was first described by Hyde under the name *A. velatispora* (Hyde 1992). Based on this species, the genus *Annulatascus* was erected to accommodate saprophytic fungi with a combination of morphological characters which include dark, globose to subglobose, immersed to superficial ascomata with periphysate necks; long, tapering, septate paraphyses; unitunicate, cylindrical, pedicellate, 8–spored asci, each with a relatively massive, bipartite, J- apical ring; and hyaline, aseptate to multi-septate ascospores with mucilaginous sheaths or appendages (Hyde 1992). *Annulatascus*, as currently circumscribed, is composed of 16 species. This common and easily recognizable species is the standard to which all potential family members must be compared morphologically (**Figs. 19, 20**) and molecularly (**Figs. 2-7**). At the ordinal level, *A. velatisporus* is the type species of Annulatascales (Maharachchikumbura et al. 2015).

Annulusmagnus J. Campbell & Shearer, Mycologia 96 (4): 826. 2004. MycoBank 28870.

Type species: *Annulusmagnus triseptatus.*

Annulusmagnus triseptatus (S.W. Wong, K.D. Hyde & E.B.G. Jones) J. Campb. & Shearer, Mycologia 90(4): 826. 2004. **Fig. 21 A-G.** MycoBank 488707. (*Annulatascus triseptatus* MycoBank 459600, legitimate).

≡ *Annulatascus triseptatus* S.W. Wong, K.D. Hyde & E.B.G. Jones, Mycological Research 103(5): 563. 1999.

Ascomata black, coriaceous, immersed to semi-immersed, erumpent or superficial, perpendicular to surface, separate to clustered, 375–660 µm wide × 400–700 high µm, (x̅ = 517 × 550 µm, w:h = .94), oblate to suboblate to globose. **Ascomal wall** black, 42–50 µm thick , comprised of 3 layers, outer layer of thick walled cells occluded with dark brown material, of *textura angularis* in surface view, middle layer comprised of brown thin walled isodiametric to laterally compressed elongated cells, inner layer comprised of hyaline laterally compressed elongated cells, of *textura prismatica* in surface view. **Neck** black to dark brown, yellow at apex, 88–340 µm long × 80–150 µm wide, 2 layered, outer layer dark brown with laterally compressed cells, inner laterally of compressed hyaline cells, periphysate. **Paraphyses** hyaline, 4–10 µm wide at base × 165–210 µm long, tapering toward apex, septate (**Fig. 21 E**). **Asci** unitunicate, cylindrical, 8–spored, 138–283 µm long × 7–14 µm wide (x̅ = 210.5 × 10.5 µm, l:w = 20) (**Fig. 21 A**). **Ascus base** pedicellate, 10 µm from *pars sporifera*. **Annulus** J-, doughnut shaped, bipartite, subapical flange, 2–4 µm long × 3–5 µm wide (x̅ = 3 × 4 µm, l:w = .75) (**Fig. 21 B-D**). **Ascospores** hyaline to pale brown, fusoid, 3–septate, not constricted at septa, overlapping uniseriate to uniseriate, 16–37 µm long × 5–10 µm wide (x̅ = 26.5 × 7.5 µm, l:w = 3.5), multiguttulate, without appendages, with a thin adpressed sheath (**Fig. 21 F,G**) (Campbell and Shearer 2004).

Specimens examined:

HOLOTYPE: AUSTRALIA. Koah, Clohesy River, on submerged wood, 31 Dec 1991, *K.D. Hyde,* **HKU 889 (Neotype).**

USA. Pennsylvania, Ricketts Glen State Forest, River, 41°20’1.356”N, 76°16’15.4194”W, 632m, on submerged woody debris, water 20.3 °C, pH 9, 632 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z31-1824*.

Additional specimens were examined from Canada (ON, MB), USA (AR, IL, ME, MI, MN, NC, NH, NY, OR, TN, VA, WI), and Venezuela.

Fig. 21 A-G. *Annulusmagnus triseptatus* from specimen Z31-1824. A. Ascus. B-D. Ascus apices. E. Paraphysis. F-G. Ascospores. Bars: A-C, E-G= 20 µm; D = 10 µm.

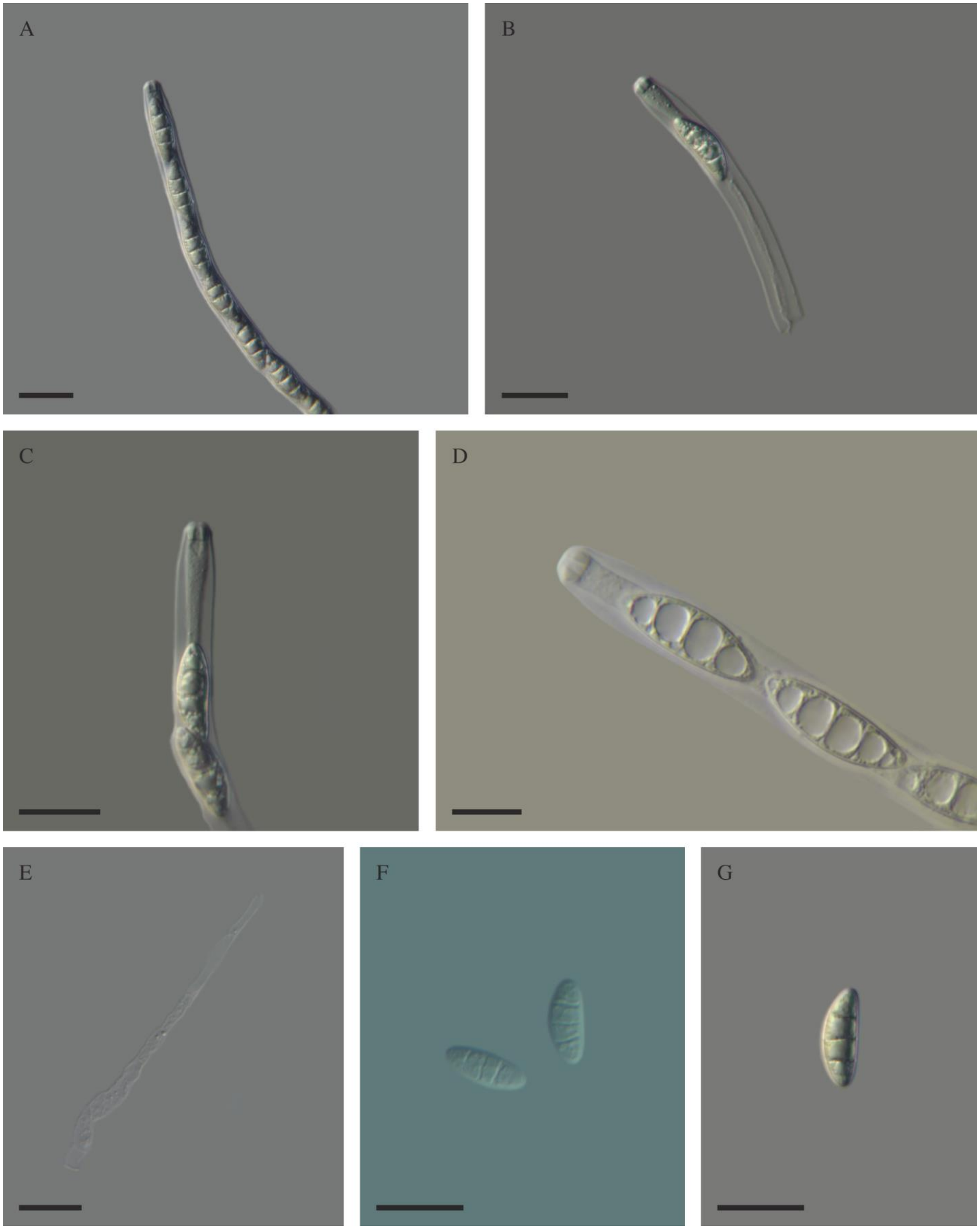


Fig. 21.

Known distribution: Australia, Canada, USA, Venezuela.

Comments: The holotype of *Annulusmagnus triseptatus* is in good condition. Fresh specimens of *An. triseptatus* obtained in this study were used in phylogenetic analyses. This species thus far appears to be restricted to North America. As noted above, the transfer of *Annulatascus triseptatus* to *Annulusmagnus* (Campbell and Shearer 2004) may be questionable due to pigmentation of ascospores and the widely separated geographic locations of collections. Molecular analyses place *An. triseptatus* in Annulatascaceae *sensu stricto*, but not in *Annulatascus*. *An. triseptatus*, represented by North American specimens, is considered by this author as a distinct genus, while *A. triseptatus* remains to be evaluated from fresh specimens from Brunei.

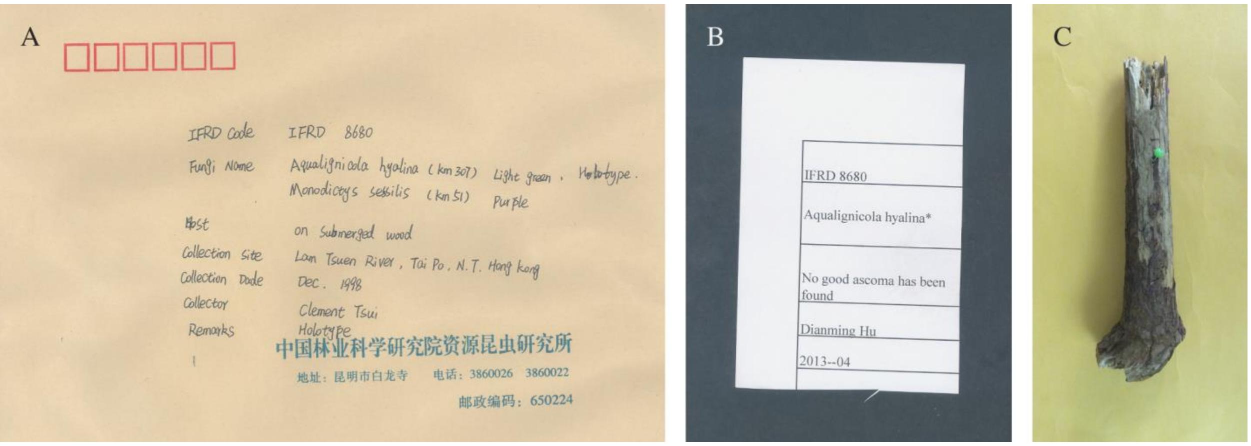
Aqualignicola Ranghoo, K.M. Tsui & K.D. Hyde, Mycological Research 105 (5): 628. 2001. MycoBank 28493.

Type species: *Aqualignicola hyalina.*

Aqualignicola hyalina V. M. Ranghoo, K. M. Tsui & K. D. Hyde, Mycological Research 105(5): 628, 2001. **Fig. 22 A-C.** MycoBank 467769.

Ascomata hyaline to pale brown, membranous, semi-immersed, perpendicular to surface, gregarious, 140–150 µm wide × 120–125 µm high (x̅ = 145 × 122.5 µm, w:h = 1.2). **Ascomal wall** light brown, 22.5–27.5 µm thick, several layers of light brown pseudoparenchymatous cells, of *textura angularis* in surface view. **Neck** dark brown, 150–180 µm long × 35–40 µm wide (x̅ = 165 × 37.5 µm, l:w = 4.4), dark brown with lanceolate, stiff, dark brown setae. **Paraphyses** hyaline, 3.75–5 µm wide at base × 95–100 µm long, tapering toward apex, septate, flexuous. **Asci** unitunicate, cylindrical, 8–spored, 137.5–150 µm

Fig. 22 A-C. *Aqualignicola hyalina* from the holotype **IFRD 8680**. A. Herbarium packet. B. Note from Dianming Hu stating that no good ascomata were found on the substrate. C. Dried substrate.



IFRD Code IFRD 8680
Fungi Name Aqualignicola hyalina (km 307) Light green, Holotype.
Monodictys sessilis (km 51) Purple
Host on Submerged wood
Collection Site Lam Tsuen River, Tai Po, N.T. Hong kong
Collection Dade Dec. 1998
Collector Clement Tsui
Remarks Holotype

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224

IFRD 8680
Aqualignicola hyalina*
No good ascoma has been found
Dianming Hu
2013--04

Fig. 22.

long × 9–12 µm wide (x̅ = 144 × 10.5 µm, l:w = 13.7). **Ascus base** pedicellate. **Annulus** J-, wedge-shaped, 3–3.5 µm long × 3.5–4 µm wide (x̅ = 3.25 × 3.75 µm, l:w = .87). **Ascospores** hyaline, ellipsoidal to fusiform, aseptate, uniseriate to biseriate 14–15 µm long × 6.25–7.5 µm wide (x̅ = 14.5 × 6.9 µm, l:w = 2.1), with verruculose ornamentation at SEM level, with two large lipid guttules, without appendages or sheath (Ranghoo et al. 2001).**Specimens examined:**

HOLOTYPE: CHINA. Hong Kong, New Territories, Tai Po, Lam Tsuen River, on submerged wood, Dec 9 1998, *K.M. Tsui*, *KM 307, HKU(M)12178,* **IFRD 8680.**

Reports: CHINA. Hong Kong, New Territories, Plover Creek Reservoir, on submerged wood, Jan 1997, *V.M. Ranghoo, PC25, HKU(M)5226.*

Known distribution: China.

Comments: The holotype of *Aqualignicola hyalina* is in poor condition (**Fig. 22 A-C**). A written note included with the holotype (**Fig. 22 B**) (Dianming Hu) suggested that no ascomata of *Aqual. hyalina* were found on the dried wood substrate (**Fig. 22 C**) from which the holotype was first described. Examination in this study corroborates this finding. Morphologically, *Aqual. hyalina* differs from all members of Annulatascaceae *sensu stricto* in having dark brown setae arising from the ascomata and necks. Ranghoo et al. (2001) compared *Aqual. hyalina* to *Aquaticola hyalomura* due to its membranous hyaline ascoma and to *Fluminicola bipolaris* due to its hyaline verruculose ascospores. *Aqual. hyalina* differs from *Aq. hyalomura* in having setae on the neck and in its verruculose ascospore ornamentation. *Aqual. hyalina* differs from *F. bipolaris* in having hyaline ascomata and lacking ascospore appendages. A culture is reported in the protologue (HKUCC 3798 culture ex holotype). If the culture is not viable, neotypification from the type location is recommended with subsequent molecular work in order to determine the phylogenetic placement of *Aqual. hyalina*. At present, *Aqual. hyalina* should be considered Sordariomycetes *incertae sedis.*

Aqualignicola vaginata D.M. Hu, L. Cai & K.D. Hyde, Mycotaxon 120: 84. 2012. MycoBank 563811.

Ascomata brown to black, membranous, immersed, perpendicular to surface, solitary, 100–140 µm wide × 100–140 µm high ($\bar{x}$ = 120 × 120 µm, w:h = 1), globose. **Ascomal wall** brown to black, 7–14 µm thick, composed of 5 layers of light-brown pseudoparenchymatous cells, of *textura angularis* in surface view. **Neck** hyaline, 200–350 µm long × 40–60 µm wide ($\bar{x}$ = 275 × 50 µm, l:w = 5.5), with brown, lanceolate setae, periphysate. **Paraphyses** hyaline, 4–6 µm wide at base × 85–100 µm long, tapering toward apex, septate, flexuose. **Asci** unitunicate, cylindrical, 8–spored, 145–156 µm long × 5.5–6.5 µm wide ($\bar{x}$ = 151 × 6 µm, l:w = 25). **Ascus base** pedicellate, 15 µm from *pars sporifera*. **Annulus** J-, large. **Ascospores** hyaline, ellipsoidal-fusiform, aseptate, uniseriate, 11–15 µm long × 5–6 µm wide ($\bar{x}$ = 13 × 5.5 µm, l:w = 2.4), multiguttulate, with a unipolar mucilaginous sheath (Hu et al. 2012).

HOLOTYPE: CHINA. Yunnan Province, Mengla, Wudaoban Stream, 21°32’N, 101°29’E, 620m, 2 Apr 2009, *D.M. Hu,* **IFRDC 021-043.**

Comments: The holotype of *Aqualignicola vaginata* was not examined as it was described late in this project, and fresh specimens were not obtained. However, the protologue was scrutinized and morphological details were added to the Annulatascaceae database. Morphologically, this species fits well within the type of the genus, *Aqual. hyalina*. The dark setae on the neck, the hyaline ascospores, and the freshwater habit indicate relatedness (Hu et al. 2012). *Chaetorostrum quincemilense,* which also has dark setae on its neck, differs from *Aqual. hyalina* in that it has 1–septate, versicolored ascospores. Molecular data is not available for either species of *Aqualignicola*, and as these species were not collected during this study, future collections and phylogenetic analyses are warranted. The generic delimitation has merit, but the familial placement in Annulatascaceae is doubtful due to the lack of setose necks in the Annulatascaceae *sensu stricto* clade. At present, *Aqual. vaginata* should be considered Sordariomycetes *incertae sedis.*

Aquaticola W.H. Ho, K.M. Tsui, Hodgkiss, & K.D. Hyde, Fungal Diversity 3: 88. 1999. MycoBank 28389.

Type species: *Aquaticola hyalomura.*

Aquaticola ellipsoidea W.H. Ho, K.M. Tsui, Hodgkiss & K.D. Hyde, Fungal Diversity 3: 90. 1999. **Fig. 23 A-I.** MycoBank 460000.

Ascomata dark brown to black, coriaceous, immersed to semi-immersed, perpendicular to parallel to surface, scattered to gregarious, 150–200 µm wide × 150–250 µm high ($\bar{x}$ = 175 × 200 µm, w:h = .9), globose to subglobose (**Fig. 23 A,B**). **Ascomal wall** hyaline and brown, 15–30 µm thick, composed of 2 layers, outer layer 2–3 rows of brown, angular cells, inner layer 4–8 rows of hyaline angular cells, of *textura angularis* in surface view (**Fig. 23 C**). **Neck** dark brown to black, setae at tip, periphysate. **Paraphyses** hyaline, 5 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 90–125 µm long × 10–12.5 µm wide ($\bar{x}$ = 107.5 × 11.3 µm, l:w = 9.6) (**Fig. 23 C-E**). **Ascus base** pedicellate, short truncate, club-like, 10 µm from *pars sporifera*. **Annulus** J-, small (**Fig. 23 F-H**). **Ascospores** hyaline, ellipsoidal, aseptate, overlapping uniseriate, 12–14 µm long × 5–7 µm wide ($\bar{x}$ = 13 × 6 µm, l:w = 2.2), guttulate with no appendages or sheath (**Fig. 23 I**) (Ho et al. 1999).

HOLOTYPE: HONG KONG. Tai Po, Tai Po Kau Forest Stream, on *Machilus velutina* wood bait, 29 Mar 1997, *W.H. Ho,* **HKU(M) 6033.**

Specimens examined:

USA. North Carolina, GSMNP, Noland Creek, fast flowing river, 37°27’00”N, 83°31’40”W, 600m, on submerged woody debris, water 19 °C, pH 5, 16 Jul 2000, *J. Campbell, R0008-1;* Fontana Village, Cheoah Lake, 35°26’51”N, 83°48’46”W, 583m, on submerged woody debris, water pH 5.5, 19 Jul 2000, *J. Campbell, R0008-2;* Noland Creek, fast flowing river, 35°27’14”N, 83°31’42”W, 564m, on submerged woody debris, 16 Jul 2000, *J. Campbell, R0008-3*; Lake Santeetleh, 35°22’26”N, 83°52’09”W, 612m, on submerged woody debris, water 28 °C, pH 5, 18 Jul 2000, *J. Campbell, R0008-4.*

Comments: The holotype of *Aquaticola ellipsoidea* was unavailable for examination. Alhough described at the same time as *Aq. hyalomura*, the type of the genus, *Aq. ellipsoidea*, which has black ascomata, was not designated as the type species of the genus. The ascomatal coloration difference between the two taxa, one black and one hyaline, marks them as separate species and this study finds them to belong to separate genera. With relatively few, simple morphological characters, *Aquaticola* species *sensu* Ho (Ho et al. 1999), and *Aquaticola*-like species require molecular data to ascertain phylogenetic placement at the generic level and above. *Aq. ellipsoidea* does not belong in Annulatascaceae *sensu stricto*, but is a sister taxon to the informally described *Aq. hongkongensis* with good support in combined three gene analyses (**Fig. 7, Clade 4**). *Aq. ellipsoidea*, along with the informally described *Aq. hongkongensis*, are placed in a clade between Savoryellales and Microascales, and thus *Aq. ellipsoidea* is considered Sordariomycetes *incertae sedis*. As *Aq. ellipsoidea* is not related to the type of the genus, *Aq. hyalomura*, a new genus and combination, *Aquadulcemyces ellipsoidea*, is proposed (see Nomenclatural changes).

Aquaticola hongkongensis Ranghoo, K.D. Hyde & E.C.Y. Liew, Fungal Diversity 2:161. 1999.
MycoBank 580431.

= *Aquaticola hongkongensis* V.M. Ranghoo & K.D. Hyde, Ranghoo Ph.D. thesis, 76. 1998.

Ascomata black, membranous, semi-immersed, mostly gregarious, 250–275 µm wide × 250–263 µm high ($\overline{x}$ = 262.5 × 256.5 µm, w:h = 1), globose or subglobose. **Ascomal wall** 25–30 µm thick, comprised of 5–7 layers of this walled dark brown compressed cells, of *textura angularis* in surface view. **Neck** dark

Fig. 23 A-I. *Aquaticola ellipsoidea* from specimen R0008. A. Ascomata on surface of substrate. B. Squash mount illustrating lighter colored necks. C Ascus and view of peridial surface. D-E. Asci. F-H. Ascus apices. I. Ascospores. Bars: A = 100 µm; B = 50 µm; E-G, I = 20 µm; H = 10 µm.

Fig. 23.

brown, 100–120 µm long × 30–40 µm wide, periphysate. **Paraphyses** hyaline, 6–8µm wide at base × 143–150 µm long, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 150–175 µm long × 13–15 µm wide ($\bar{x}$ = 162.5 × 14 µm, l:w = 11.6). **Ascus base** pedicellate, short, acute, 20 µm from *pars sporifera*. **Annulus** J-, 3–4 µm long × 3–4 µm wide (l:w = 1). **Ascospores** hyaline, ellipsoidal, aseptate, uniseriate to overlapping uniseriate, 23–26 µm long × 7.5–11 µm wide ($\bar{x}$ = 24.5 × 9.25 µm, l:w = 2.7), without appendages, with a thin mucilaginous sheath (Ranghoo 1998).

HOLOTYPE: CHINA. Hong Kong, New Territories, Plover Creek Reservoir, on submerged wood, Jan 1997, *V.M. Ranghoo,* **HKU(M) 4254.**

Comments: *Aquaticola hongkongensis* is listed in MycoBank and was referred to that database from Index Fungorum. Index Fungorum considers this an invalid name (Art. 36.1(c) (Melbourne) which would mean it may be "merely cited as a synonym" (McNeill et al. 2012). The holotype of *Aq. hongkongensis* was not examined in this study as Ranghoo's thesis (1998) was only obtained near the completion of this manuscript. Thus only the protologue was used to assess this species morphologically. As this thesis is freely available from Hong Kong University at this time, this species is being included in this study for completeness, with all credit given to Ranghoo (1998). ITS and 28S sequences of *Aq. hongkongensis* were available in GenBank and used for molecular analyses. ITS analyses place *Aq. hongkongensis* on a separate long branch from other *Aquaticola* and *Annulatascus* species (**Fig. 4**). Combined three gene molecular analyses place *Aq. hongkongensis* as a sister taxon to *Aq. ellipsoidea* with good support in this study (**Fig. 7, Clade 4**). Morphologically, *Aq. ellipsoidea* can be distinguished from *Aq. hongkongensis* by its shorter asci (90–125 µm vs. 150–175 µm) and smaller ascospores (12–14 × 5–7 µm vs. 23–26 × 7.5–11 µm). *Aq. hongkongensis*, along with the *Aq. ellipsoidea*, are placed in a clade between Savoryellales and Microascales, and thus *Aq. hongkongensis* should be considered Sordariomycetes *incertae sedis* at present. A formal publication of the species by Ranghoo is warranted.

Aquaticola hyalomura W.H. Ho, K.M. Tsui, Hodgkiss, & K.D. Hyde, Fungal Diversity 3: 88. 1999. **Fig. 24 A-L.** MycoBank 459999.

Ascomata white to pale brown, coriaceous, immersed to superficial, mostly solitary, 160–290 µm wide × 160–300 µm high (x̅ = 225 × 230 µm, w:h = .98), globose to subglobose (**Fig. 24 A**). **Ascomal wall** of *textura angularis* in surface view. **Neck** white to pale brown, 100–150 µm long × 45–65 µm wide (x̅ = 125 × 55 µm, l:w = 2.3), periphysate (**Fig. 24 B,C)**. **Paraphyses** hyaline, 2–3.5 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, broadly oblong (clavate), 8–spored, 52–64 µm long × 7–8 µm wide (x̅ = 58 × 7.5 µm, l:w = 7.7) (**Fig. 24 E,F**). **Ascus base** pedicellate, short, acute, 10 µm from *pars sporifera* (**Fig. 24 E,F)**. **Annulus** J-, faintly bipartite, 2 µm long × 4 µm wide (l:w = .5) (**Figs. 24 G,H**). **Ascospores** hyaline, ellipsoidal, aseptate, overlapping uniseriate to biseriate, 10–14 µm long × 5–7 µm wide (x̅ = 12 × 6 µm, l:w = 2), with two large lipid droplets, without appendages, with a thin sheath (**Fig. 24 I-L**) (Ho et al. 1999).

HOLOTYPE: HONG KONG. Tai Po Kau Forest Stream, on submerged decaying wood, 27 Jun 1996, *K.D. Hyde, WH189,* **HKU(M) 2969.**

Specimens examined:

USA. Pennsylvania, Orangeville Fishing Creek, 41°7'42.744"N, 76°21'26.6034"W, 191m, on submerged woody debris, water pH 9.5, 80 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z39-3;* Rickett’s Glen State Forest, river, 41°20'1.356"N, 76°16'15.4194"W, 632m, on submerged woody debris, water pH 9, 632 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z39-4.* North Carolina, GSMNP, Twenty Mile Creek, 35°28’02”N, 83°52’37”W, 306m, on submerged woody debris, water pH 5, 19 Jul 2000, *J. Campbell, R0038-1;* Stream in Okanaluftee watershed, near Cherokee, Newfound Gap, 35°35'6"N, 83°21'28.7274"W, 868m, on submerged woody debris, water 17.4 °C, pH 7.4, *Zelski S.E and H.A. Raja,* *Z39-1*. Tennessee, Pigeon River watershed, near Sugarlands, 35°40'32.8434"W, 83°43'37.848"N, 536m, on submerged woody debris, water 20 °C, pH 7.4, *Zelski S.E. and H.A. Raja*, *Z39-2*.

Known distribution: Hong Kong, USA.

Comments: The holotype of *Aquaticola hyalomura* was unavailable for examination. Many different collections made in this study were similar to the holotype description and were nearly identical to the GSMNP collection (R038) from which molecular data was first obtained. Molecular evidence from these highly similar fungi from the tropics, however, places these taxa in separate clades (**Fig. 7 Clade 1b, Clade 1c**). Two of these new species that are morphologically most similar to *Aq. hyalomura* (PE0157, PE0264) are described in the section on new taxa. Due to few morphological differences in these taxa (all possess hyaline ascomata, short clavate asci, small ascus apical rings, and small aseptate ascospores), visual observations must be supplemented with molecular evidence to determine species identity. As there is no reported culture from the holotype, fresh collections at the type locality in Hong Kong should be made to obtain DNA which could then be compared to North American and South American taxa that are extremely similar. Based on analyses in this study, *Aquaticola* is polyphyletic and not affiliated with Annulatascaceae *sensu stricto*. *Aq. hyalomura* is placed in a statistically unsupported clade with *Cataractispora* species in this study and should be considered Sordariomycetes *incertae sedis* (**Fig. 7, Clade 1b**).

Aquaticola longicolla K.M. Tsui, Hodgkiss & K.D. Hyde, Nova Hedwigia 77 (1-2): 162. 2003. **Fig. 25 A-J.** MycoBank 489211.

Ascomata black, coriaceous, immersed, perpendicular to surface, solitary, 212–248 µm wide, globose (**Fig. 25 C**). **Ascomal wall** dark brown, 15–25 µm thick, 10–15 layers of dark brown cells, small and rounded externally, long and narrow internally. **Neck** black, to 1200 µm long, periphysate. **Paraphyses**

Fig. 24 A-L. *Aquaticola hyalomura* from specimen PA C-1823. A. Ascomata on surface of substrate. B. Neck illustrating periphyses. C. Surface texture of neck. D. Paraphyses. E-F. Asci. G-H. Ascus apices. I-L. Ascospores. Bars: A = 500 µm; B-F= 20 µm; G-L = 10 µm.

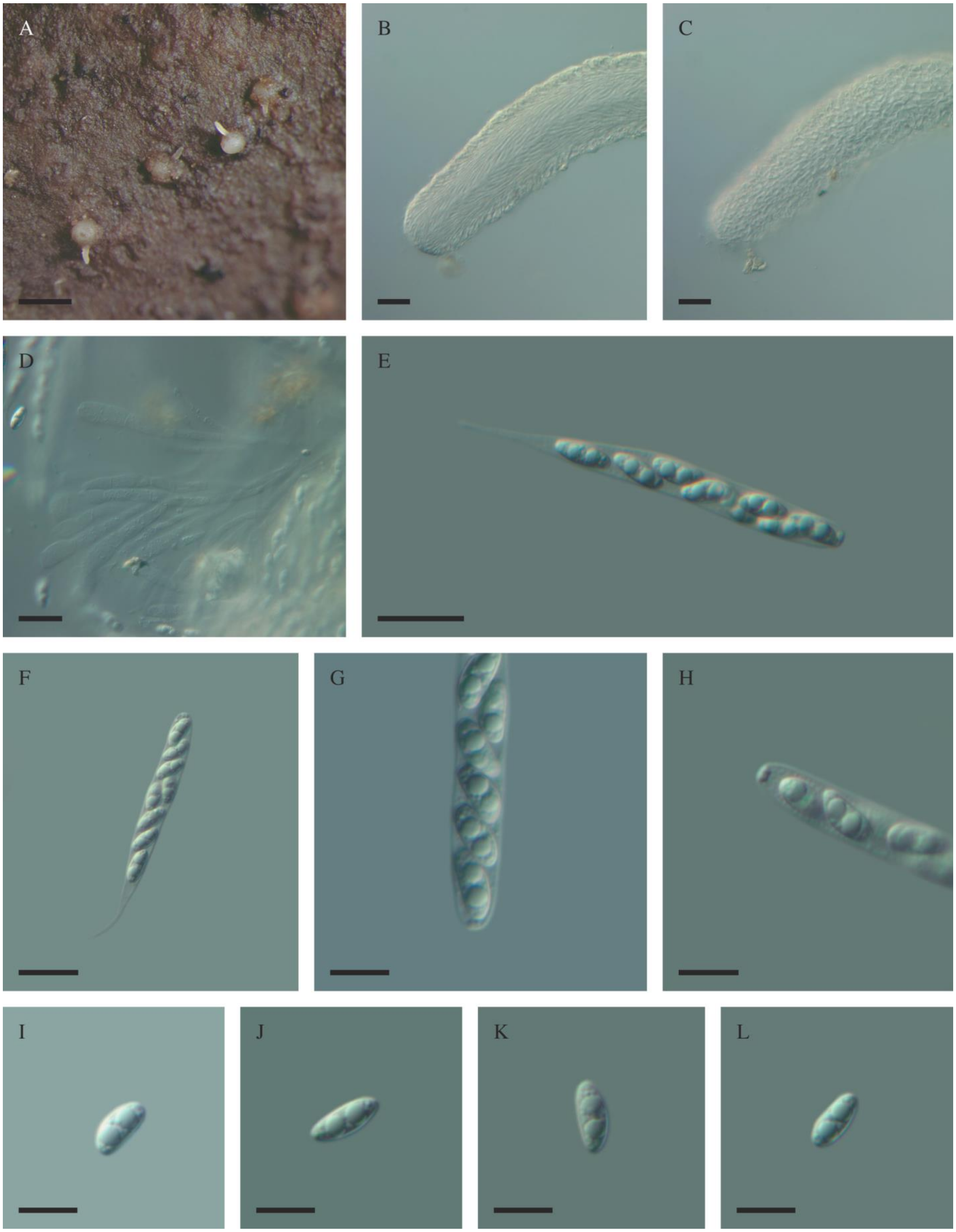


Fig. 24.

hyaline, 7.5 µm wide at base, tapering toward apex, septate, filamentous, flexuose (**Fig. 25 H)**. **Asci** unitunicate, short cylindrical to cylindrical-clavate, 8–spored, 75–83 µm long × 9–12 µm wide ($\bar{x}$ = 79 × 10.5 µm, l:w = 7.5). **Ascus base** pedicellate, broadly tapering, 5 µm from *pars sporifera* (**Fig. 25 E**).**Annulus** J-, discoid, 1.5 µm long × 3 µm wide (l:w = .5) (**Fig. 25 F,G**). **Ascospores** hyaline, ellipsoidal to fusiform, aseptate, 2–3 seriate, 12–16.5 µm long × 4.5–6 µm wide ($\bar{x}$ = 14.25 × 5.25 µm, l:w = 2.7), with two large lipid guttules, without appendages or sheath (**Fig. 25 I,J**) (Tsui et al. 2003).

Specimens examined:

HOLOTPYE: AUSTRALIA. North Queensland, near Ravenshoe, ca. 20 km along the Tully Palls Road to Koomoolooba Dam, 17°45'S, 145°35'E, submerged in creek, Mar 1997, *K.D. Hyde, RH 49, (HKU(M) 5159),* **IFRD 8682.**

PERU. Cusco, Camanti, Quincemil, 2nd stream along Quincemil Trail 1, trailhead 13°14'22.5594"S, 70°46'12.6114"W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0334-1;* 1st stream along Quincemil Trail 1, trailhead 13°14'22.5594"S, 70°46'12.6114"W, 688m, on submerged woody debris, water 21.7 °C, pH 6.8, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0334-2.*

Known distribution: Australia, Perú.

Comments: The holotype of *Aquaticola longicolla* was in good condition (**Fig. 25 A,B**). Fresh collections were discovered from Peruvian material very late in this study and thus not included in this manuscript. Thus phylogenetic placement remains uncertain, though an effort to isolate this species in pure culture is underway. This species differs from other described *Aquaticola* species in its ascus morphology. Rather than short cylindrical asci with short club-like pedicels, *Aq. longicolla* has asci which

Fig. 25 A-J. *Aquaticola longicolla* from the holotype **IFRD 8682**. A. Herbarium packet. B. Dried specimen. C. Immersed ascomata with protruding necks. D. Ascus. E. Ascus base. F-G. Ascus apices. H. Paraphyses. I. Ascospores. J. Ascospore. Bars: C = 500 µm; D-E, H = 20 µm; F-G, I-J = 10 µm.


A
中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224
B
C
D
E
F
G
H
I
J

Fig. 25.

become inflated and possess thin ascus tails (**Fig. 25 D,E)**. DNA is required to assess the phylogenetic placement of this species in relation to *Aquaticola* and Annulatascaceae *sensu stricto*, as all other *Aquaticola* and *Aquaticola*-like taxa examined in this study do not form a monophyletic group and are also removed from the family. *Aq. longicolla* should be considered Sordariomycetes *incertae sedis* at present.

Aquaticola miniguttulata K.M. Tsui, Hodgkiss & K.D. Hyde, Nova Hedwigia 77(1-2): 162. 2003. **Fig. 26 A-M, Fig. 27 A-L.** MycoBank 489210.

Ascomata black, coriaceous, immersed to semi-immersed, perpendicular to surface, scattered to gregarious, 150–220 µm wide × 200–300 µm high ($\bar{x}$ = 185 × 200 µm, w:h = 2.7), globose to subglobose (**Fig. 26 D, Fig. 27 A)**. **Ascomal wall** hyaline and brown, a few outer layers of dark brown angular cells, becoming hyaline inward, 25–40 µm thick, of *textura angularis* in surface view (**Fig. 26 E**). **Neck** black, 150–300 µm long × 25–30 µm wide ($\bar{x}$ = 225 × 27.5 µm, l:w = 8.2), periphysate (**Fig. 27 E**). **Paraphyses** hyaline, 10 µm wide × 150 µm long, tapering, septate (**Fig. 27 F**). **Asci** unitunicate, cylindrical, 8–spored, 96–130 µm long × 10–12 µm wide ($\bar{x}$ = 113 × 11 µm, l:w = 10.3) (**Fig. 26 F-H, Fig. 27 B-E**). **Ascus base** pedicellate, short, club-like, 10 µm from *pars sporifera*. **Annulus** J-, small, 2–2.5 µm long × 3–4 µm wide ($\bar{x}$ = 2.25 × 3.5 µm, l:w = .64) (**Fig. 26 I,J; Fig. 27 G-I**). **Ascospores** hyaline, ellipsoidal, aseptate, uniseriate to overlapping uniseriate, 9–12 µm long × 6–8 µm wide ($\bar{x}$ = 10.5 × 7 µm, l:w = 1.5), multiguttulate, no appendages, with a thin sheath that spreads out (**Fig. 26 K-M, Fig. 27 J-L**) (Tsui et al. 2003).

Specimens examined:

HOLOTYPE: CHINA. Hong Kong, Sai Kung, Hang Cho Shui, on submerged wood, Sep 1998, *Clement Tsui and Edmund Shek, KM52,* **IFRD 8683.**

PERU. Cusco, Camanti, Quincemil, river backwater at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 22 °C, pH 7.1, 14 Apr 2011, *Zelski S.E. and H.A. Raja, PE0120-10;* Stream crossing the Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 737m, on submerged woody debris, water 22°C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0120-11;* River at Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21 °C, pH 6.3, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0120-*9*;* Stream at Quincemil Trail 2, trailhead 13°13’31.0434”S, 70°45’10.6194”W, 653m, on submerged woody debris, water 21.9 °C, pH 6.4, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0120-1;* River at Quincemil Trail 3, 13°18’53.128”S, 70°48’44.8194”W, 817m, on submerged woody debris, water 20.3 °C, pH 7.6, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0120-8;* 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.1 °C, pH 6.7, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0120-4;* River backwater at Quincemil Trail 3, on submerged decorticated wood, 13°18’27.756”S, 70°48’44.9279”W, 757m, water 22 °C, pH 7.1, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0120,-10;* Madre de Dios, CICRA, cocha seca (dry lake) at Trail 29, 12°34’16.98”S, 70°05’06.70”W, 244m, on submerged decorticated wood, water 23.2 °C, pH 6.4, 23 May 2010, *Zelski S.E. and H.A. Raja, PE0120-2;* CICRA, Rio Amigos, 12°34’02.86”S, 70°04’56.26”W, 218m, on submerged woody debris, water 25.3 °C, pH 7.9, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0120-6;* CICRA, oxbow lake at Trail 14, 12°34’14.74”S, 70°05’23.69”W, 241m, on submerged decorticated wood, water 23.0 °C, pH 6.7, 23 May 2010, *Zelski S.E. and H.A. Raja, PE0120-4;* CICRA, Rio Amigos, 12°34’3.2514”S, 70°4’55.92”W, 218m, on submerged woody debris, water 25.3 °C, pH 7.9, 23 May 2010, *Zelski S.E. and H.A. Raja, PE0120-5;* CICRA, Stream at Trail 19, 12°34’01.04”S, 70°05’43.24”W, 275m, water 23.7 °C, pH 5.1, on submerged decorticated wood, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0120-12.* THAILAND. Chiang Mai, Tham Rusee Nature Trail, 18°40’24.4794”N, 90°54’38.3754”E, 1149m, on submerged woody debris, water 22.4 °C, pH 7.2, 18 Jun 2012, *Zelski S.E., TH0120-1.*

Reports:

CHINA. Hong Kong, Tai Po, Lam Tsuen River, on submerged wood, Sep 1996, *K.M. Tsui, KM52 (HKU(M) 4632, 4653)*; *ibid.*, Sep 1997, *K.M. Tsui, (HKU(M) 8087);* Lantau Island, Tung Chung River, on submerged wood, Aug 1997, *K.M. Tsui and Ken Wong, (HKU(M) 5532);* Tsuen Wan, Shing Mun Reservoir, on submerged wood, Jan 1998, *K.M. Tsui, (HKU(M) 8184); ibid.*, Oct 1998, *K.M. Tsui and T. Umali, (HKU(M) 12333)*.

Known distribution: China, Perú, Thailand.

Comments: The holotype of *Aquaticola miniguttulata* was in fair condition, with scant fungal material (**Fig. 26 A,B**). Included with the holotype is a photographic plate that differs from the protologue (**Fig. 26 C**) (Dianming Hu). Only two ascomata were located on the dried substrate near the pin demarcating the location of the fungus. One ascoma was removed, examined, preserved as a permanent slide, and returned with the holotype. Paraphyses were not preserved in the dried specimen but were described in the protologue as being numerous, wide at the base and tapering toward the apex, septate, filamentous, and embedded in a gelatinous matrix. However, the ascoma was replete with asci and ascospores. Fresh specimens were collected in Perú and Thailand. These specimens were good matches to the holotype. Peruvian specimens (PE0120) were grown in culture and sequenced. Three taxa were added to *Aquaticola* at the time *Aq. miniguttulata* was described (*Aq. longicolla, Aq. miniguttulata,* and *Aq. triseptata)* and were included based on morphological similarities (Tsui et al. 2003). The authors acknowledged that resolution of the phylogenetic relationships among these taxa required further molecular investigation. Characters which place *Aq. miniguttulata* in *Aquaticola* include membranous, thin walled peridia, well developed necks, paraphyses with cells tapering towards their apices, extending slightly beyond the asci, and cylindrical asci. Peruvian specimens (PE0120) agreed well with *Aq. miniguttulata* collected from Hong Kong, though ascospore measurements (9.9–12.22 × 4.6–6.1 µm, $\bar{x}$ = 11.1 × 5.4 µm, n = 30) were slightly smaller than those of the holotype (12–16.5 × 4.5–6 µm). The apical ring of the Peruvian specimen agrees well with the type, (1.5–2.1 × 2.6–3, x̅ = 1.9 × 2.8, n = 10), vs. (1.5 × 3 µm). Other than a slight difference in ascospore measurements, Peruvian specimens agree well with the type description of *Aq. miniguttulata*. The ascospores are telling as they are relatively thick walled (1 µm), and range from ellipsoid to fusiform. Peruvian specimens were nearly identical to Thai specimens. In combined three gene analyses, *Aq. miniguttulata* does not cluster with *Aq. hyalomura,* the type of the genus, nor does it belong in Annulatascaceae *sensu stricto* (**Fig. 7, Clade 1f**). As *Aq. hyalomura* has hyaline membranous ascomata, this is not surprising. *Aq. miniguttulata* clusters with *Fusoidispora aquatica* and a new *Aquaticola*-like species (PE0316), in a well supported clade in the three gene analyses. This clade is nested within in a larger clade with only Bayesian support that includes *Annulatascus biatriisporus*, *Lentomitella* species, *Aquaticola*-like species (AK-1838, PE0316), and *Torrentispora fibrosa*. *Aq. miniguttulata* should be considered Sordariomycetes *incertae sedis* at present. As *Aq. miniguttulata* is not related to the type of the genus, *Aq. hyalomura*, a new genus, *Breviascus,* is proposed to house *B. miniguttulata* (see Nomenclatural Changes).

Aquaticola rhomboida Ho Ph.D. thesis, 58. 1998.

Ascomata dark brown, immersed to superficial, coriaceous, 80–140 µm wide × 160–260 µm high (x̅ = 110 × 210 µm, w:h = .5), globose to subglobose. **Ascomal wall** brown, of *textura angularis* in surface view. **Neck** 60–140 µm long × 10–14 µm wide, periphysate. **Paraphyses** hyaline, 3–4.5 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, overlapping uniseriate, 80–125 µm long × 8.7–10 µm wide (x̅ = 108 × 9.4 µm, l:w = 10.9, n = 10). **Ascus base** pedicellate. **Annulus** 3–3.5 µm long × 2–2.5 µm wide (x̅ = 3.25 × 2.25, l × w = 1.44). **Ascospores** hyaline, rhomboid with one plane flattened, aseptate, overlapping uniseriate to biseriate, 16–20 µm long × 7–8.5 µm wide (x̅ = 18.3 × 7.6 µm, l:w = 1.5, n = 25), with two large guttules, without appendages or sheath (Ho 1998).

Fig. 26 A-M. *Aquaticola miniguttulata* from the holotype **IFRD 8683**. A. Herbarium packet. B. Dried substrate. C. Photographic plate enclosed with holotype (Dianming Hu). D. Neck protruding from surface of substrate. E. Peridium. F-H. Asci. I. Ascus apex. J. Ascus base. K-M. Ascospores. Bars: D = 500 µm; E-H = 20 µm; I-M = 10 µm.

International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8683
Fungi Name	Aquaticola miniguttulata orange
Host	on Submerged wood
Collection Site	CHINA, Hong Kong, Sai Kung, Hang Cho Shui
Collection Date	1998-9-8
Collector	Clement Tsui and Edmund Shek
Remarks	Holotype, KM52

Monotosporella tunicata (km 267) dark blue
Gonytrichum chlamydosporium var. chlaymelosporium (km 127) purple
Canalisporium caribense (km 153) red.

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

by Dianming Hu
2013. 9

Fig. 26.

Fig. 27 A-L. *Aquaticola miniguttulata* from specimen PE0120. A. Habit view with neck protruding from substrate. B-D. Asci. E. Immature ascus. F. Paraphyses. G-H. Asci illustrating back door dehiscence. I. Annulus. J-L. Ascospores. Bars: A= 500 µm, B-H = 20 µm, J-L = 10 µm.

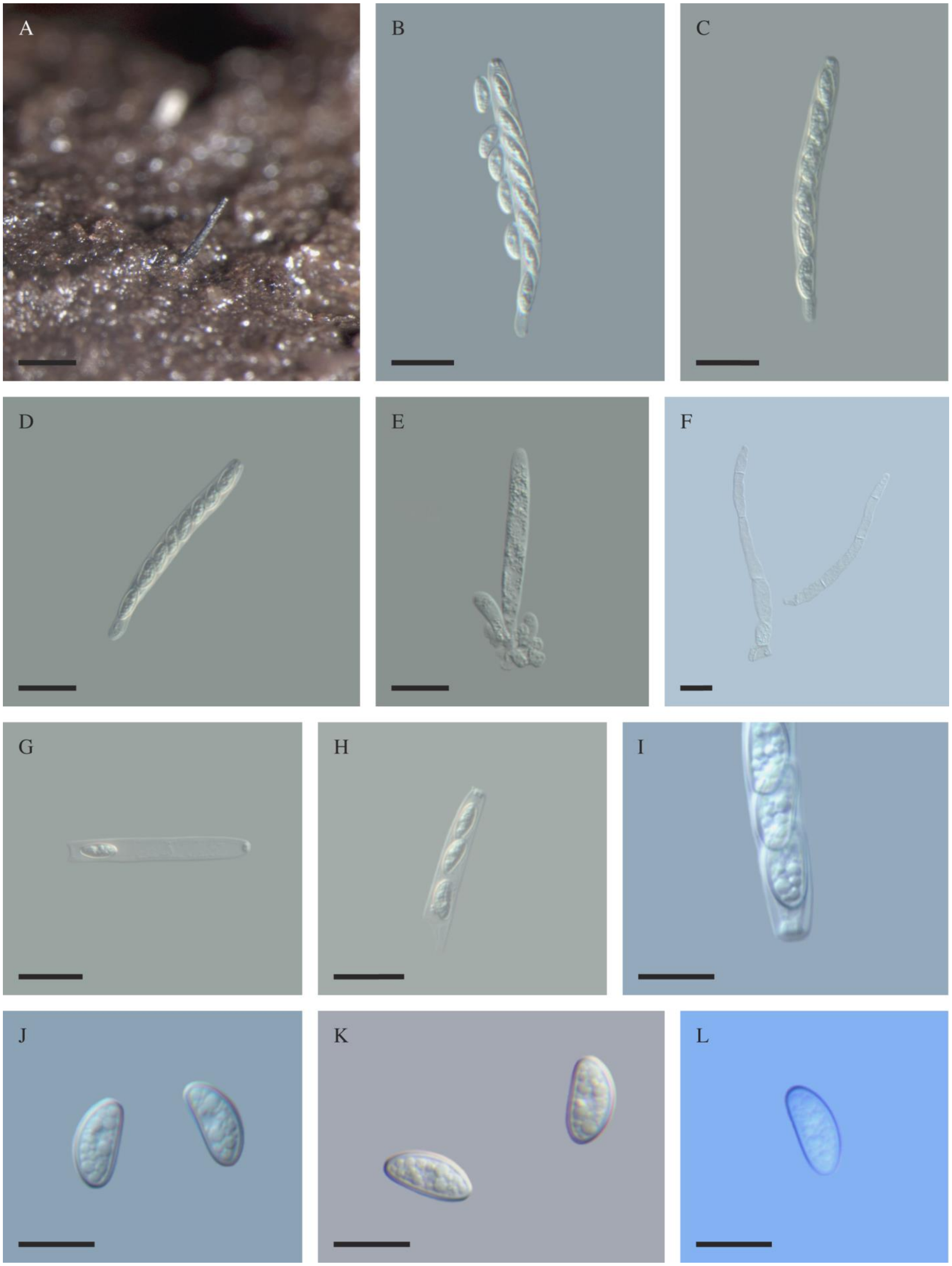


Fig. 27.

HOLOTYPE: HKU(M) 2924

Comments: Though informally published in Ho's Ph.D. thesis (1998) *Aquaticola rhomboida* is not recognized in MycoBank or Index Fungorum (*nomen invalidum*). This thesis was not obtained until this manuscript neared completion. Thus the protologue was used to assess this species morphologically. As this thesis is freely available online at this time, this species is being included in this study for completeness, with all credit given to Ho (1998). *Aq. rhomboida,* with its dark brown ascomata, is in the same category as the majority of the members of this genus in that it differs from the type of the genus, *Aq. hyalomura*, which has hyaline ascomata. Aside from this difference, the asci, ascus apical apparatus, and ascospores are typical of members of *Aquaticola*. This species requires formal publication and further investigation, especially from a molecular perspective. As *Aquaticola* is shown to be polyphyletic in this study, *Aq. rhomboida* should be considered Sordariomycetes *incertae sedis* at present.

Aquaticola triseptata K.M. Tsui, Hodgkiss & K.D. Hyde, Nova Hedwigia 77(1-2): 165. 2003. **Fig. 28 A-C, Fig. 29 A-K.** MycoBank 489212.

Ascomata reddish-brown to brown, immersed to semi-immersed, perpendicular to surface, solitary to gregarious, 200–240 µm wide × 240–320 µm high ($\bar{x}$ = 220 × 280 µm, w:h = .8), globose to subglobose (**Fig. 29 A**). **Ascomal wall** reddish-brown to brown, 15–30 µm thick, 5–6 layers of angular cells compressed at ascomal base, of *textura angularis* in surface view (**Fig. 29 B,C**). **Neck** reddish-brown to brown, 240 µm long × 60 µm wide, periphysate. **Paraphyses** hyaline, 5 µm wide at base, tapering toward apex, septate, readily deliquescent (**Fig. 29 F**). **Asci** unitunicate, broadly cylindrical to cylindrical-clavate, 8–spored, 50–80 µm long × 8–12 µm wide ($\bar{x}$ = 65 × 10 µm, l:w = 6.5) (**Fig. 29 D,E**). **Ascus base** short pedicellate (**Fig. 29 E**). **Annulus** J-, small, 3 µm long × 4 µm wide (l:w = .75) (**Fig. 29 G,H**). **Ascospores** hyaline, ellipsoidal, 3–septate, not constricted at septa, overlapping uniseriate to biseriate, 15–19 µm long

× 5–7 µm wide ($\bar{x} = 17 \times 6$ µm, l:w = 2.8), with a large lipid guttule in each cell, without appendages, with a thin sheath (**Fig. 29 I-K**) (Tsui et al. 2003).

Specimens examined:

HOLOTYPE: CHINA. Hong Kong, Tai Po, Lam Tsuen River, on submerged wood, Dec 1998, *K.M. Tsui and Ginimi Chan, KM111, (HKU(M) 12222),* **IFRD 8684.**

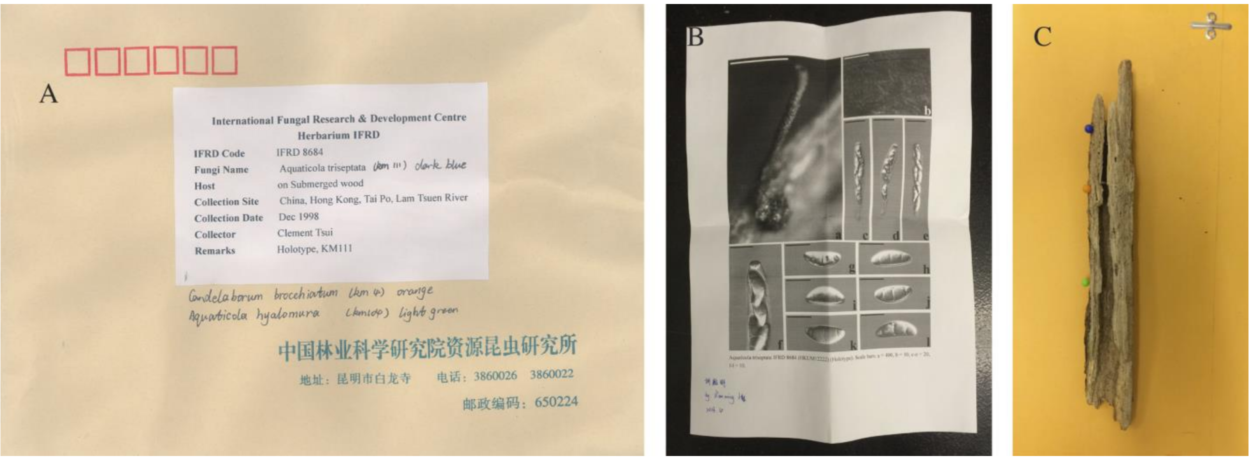
PERU. Cusco, Camanti, Quincemil, stream at Quincemil Trail 2, trailhead 13°13’31.0434”S, 70°45’10.6194”W, 653m, on submerged woody debris, water 21.9 °C, pH 6.4, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0164-2;* Stream crossing Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 653m, on submerged woody debris, water 21.7 °C, pH 7.6, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0164-1;* 5th stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.0 °C, pH 8.3, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0164-12;* 4th stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.2 °C, pH 7.7, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0164-15;* 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.1 °C, pH 6.7, Oct 2010, *Zelski S.E. and H.A. Raja, PE 0164-5;* 1st stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.7 °C, pH 5.8, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0164-4;* Rio Caliente, 1km south of Quincemil, 13°13’20.87”S, 70°44’30.07”W, 626m, on submerged woody debris, water 25.0 °C, pH 7.2, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0164-7;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, water 20.5 °C, pH 7.2, on submerged woody debris, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0164-13;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 21.4 °C, pH 7.5, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0164-11;* Stream crossing the Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 737m, on submerged woody debris, water 22.0 °C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0164-9;* Rio

Frio (Yana Mayo), 13°13’20.87”S, 70°44’30.07”W, 626m, on submerged woody debris, water 25.3 °C, pH 8, *Zelski S.E. and H.A. Raja, PE0164-23;* 4th stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.4 °C, pH 7.8, 12 Apr 2011 *Zelski S.E. and H.A. Raja, PE0164-22;* 2nd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0164-21;* 1st stream at Quincemil Trail 3, trailhead 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.7 °C, pH 6.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0164-17;* Stream flowing into river backwater at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.2 °C, pH 7.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0164-20;* Madre de Dios, CICRA, Creek at Trail 23, 12°33’31.03”S, 70°05’56.96”W, 280m, on submerged decorticated wood, water 22.2 °C, pH 6.4, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0164-3;* CICRA, Stream at Trail 28, 12°34’02.81”S, 70°05’42.96”W, 272m, on submerged decorticated woody debris, water 23.3 °C, pH 6.8, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0164-27.*

Known distribution: China, Perú.

Comments: The holotype of *Aquaticola triseptata* is in poor condition and should be neotypified with fresh specimens from Hong Kong (**Fig. 28 A-C**). Fresh collections from Perú yielded a fungus that agreed well with *Aq. triseptata.* This species was originally placed in *Aquaticola* based on morphology and the only species in the genus with 3–septate ascospores (Tsui et al. 2003). Molecular analyses from the Peruvian specimen, however, does not support inclusion in *Aquaticola*, as it is in a separate lineage from the type of the genus, *Aq. hyalomura*. *Aq. triseptata* itself does not have affinity to Annulatascaceae *sensu stricto* in this study. A morphological examination suggests *Aq. triseptata* could be related to *Cyanoannulus petersenii*, which was described at nearly the same time but the two species differ in that *Aq. triseptata* has apical rings that do not stain in cotton blue (Raja et al. 2003). 28S analyses place the

Fig. 28 A-C. *Aquaticola triseptata* from the holotype **IFRD 8684**. A. Herbarium packet. B. Photographic plate included with dried specimen (Dianming Hu). C. Dried wood lacking fungal material.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code IFRD 8684
Fungi Name Aquaticola triseptata (km 111) dark blue
Host on Submerged wood
Collection Site China, Hong Kong, Tai Po, Lam Tsuen River
Collection Date Dec 1998
Collector Clement Tsui
Remarks Holotype, KM111

Candelaborum brocchiatum (km 4) orange
Aquaticola hyalomura (km100) light green

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

Fig. 28.

Fig. 29 A-K. *Aquaticola triseptata* from specimen PE0164-1. A. Ascomata. B. Midsaggital section of ascomata. C. Peridium of *textura angularis*. D. Immature ascus. E. Ascus. F. Paraphysis. G-H. Ascus apices. I-K. Ascospores. Bars: A = 100 µm; B = 40 µm; C-E = 20 µm; F-K = 10 µm.

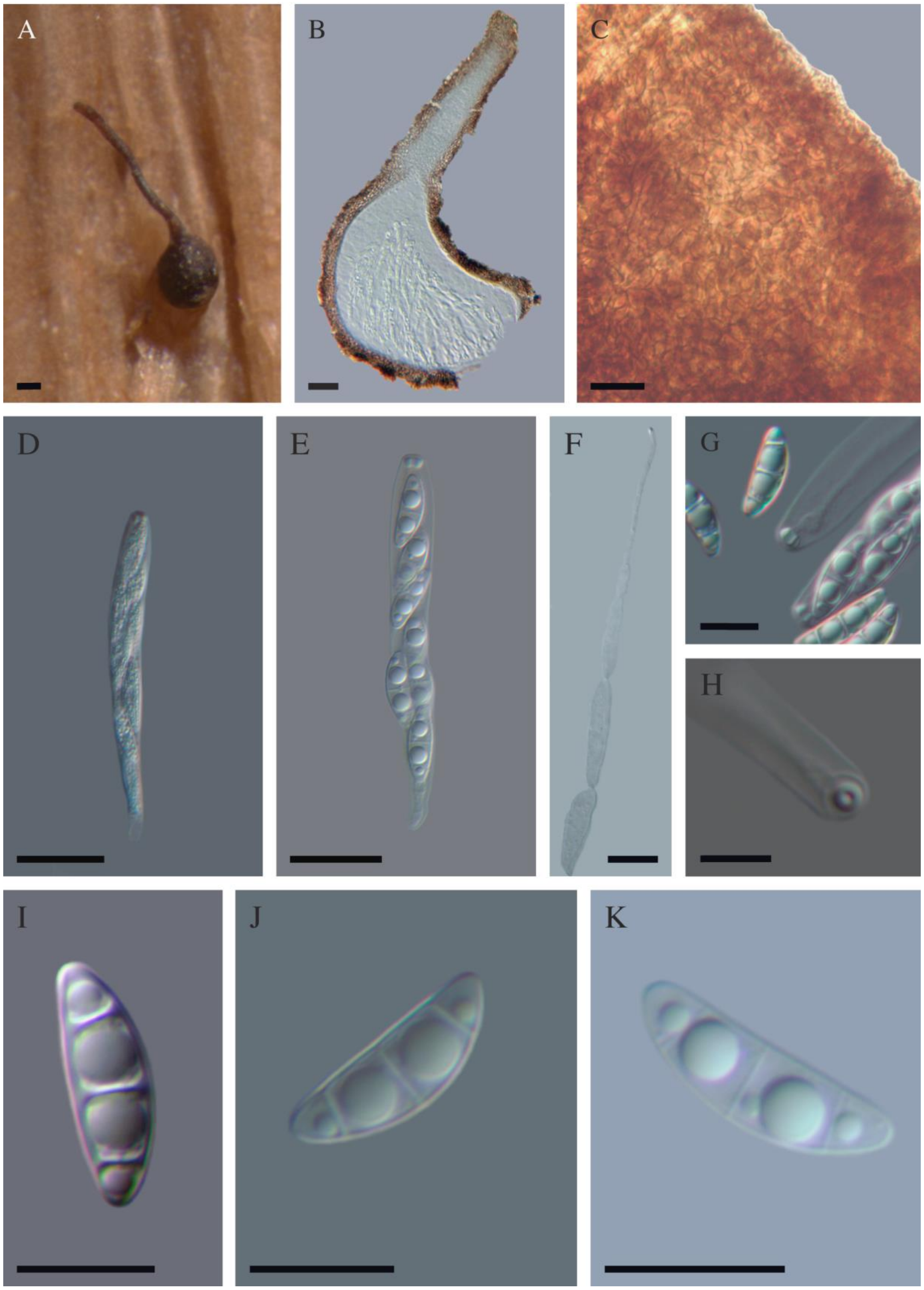


Fig. 29.

two as sister taxa (**Fig. 2**), but multi-gene analyses separate these taxa (**Fig. 6; Fig. 7 Clades 1b,3a**). *Aq. triseptata* is placed in a clade that includes Sordariales, Boliniales, and Chaetosphaeriales and should be considered Sordariomycetes *incertae sedis*. As *Aq. triseptata* is not related to the type of the genus, *Aq. hyalomura*, a new genus and combination, *Insolensaquaticola aquatica*, is proposed (see Nomenclatural Changes).

Ascitendus J. Campb. & Shearer, Mycologia 96(4): 829. 2004. MycoBank 28871.

Type species: *Ascitendus austriacus.*

Ascitendus austriacus (Réblová, Winka & Jaklitsch) J. Campb. & Shearer. Mycologia 96(4): 829. 2004. **Fig. 30 A-E, Fig. 31 A-I.** MycoBank 488706. (*Ascolacicola austriaca* MycoBank 467795, legitimate).

≡ *Ascolacicola austriaca* Réblová, Winka & Jaklitsch. Mycologia 93(3): 487. 2001.

Ascomata black, coriaceous, immersed to superficial, perpendicular to surface, separate to clustered to densely aggregated, 350–585 µm wide × 295–505 µm high ($\bar{x}$ = 467.5 × 480 µm, w:h = 1.2), globose to subglobose (**Fig. 31 A**). **Ascomal wall** brown and hyaline, 28–42 µm thick, 3 layers, outer pseudoparenchymatous with brown amorphous material, middle layer of brown laterally compressed elongated cells, inner layer of hyaline, laterally compressed elongate cells, pseudoparenchymatous in surface view. **Neck** black at base, pale yellow at apex, 85–300 µm long × 50–140 µm wide ($\bar{x}$ = 193 × 95 µm, l:w = 2), 2–layered, outer layer of dark brown laterally compressed cells occluded with brown material, inner layer laterally compressed hyaline cells, periphysate (**Fig. 31 A**). **Paraphyses** hyaline, 4–10 µm wide at base × 120–190 µm long, tapering toward apex, 1–9 septate (**Fig. 31 D**). **Asci** unitunicate, cylindrical, 8–spored, 100–260 µm long × 4–8 µm wide ($\bar{x}$ = 180 × 6 µm, l:w = 30) (**Fig. 31 B,C**). **Ascus**

base pedicellate, with a slight taper, 40 µm from *pars sporifera*. **Annulus** J-, cylindrical to flaring, 2.5–3.5 µm long × 3–3.8 µm wide (x̅ = 3 × 3.4 µm, l:w = .88) (**Fig. 31 E,F**). **Ascospores** pale brown, middle cells darker than end cells, fusoid, 3–septate, not constricted at septa, 14–27 µm long × 4–9 µm wide (x̅ = 20.5 × 6.5 µm, l:w = 3.2), large lipid guttule in each cell, without appendages or sheath (**Fig. 30 D,E; Fig. 31 G-I**) (Campbell and Shearer 2004).

Specimens examined:

HOLOTYPE: AUSTRIA. Wien 18, Hermanuskopel, on submerged wood of *Fagus sylvatica*, 29 Aug 1998, *W. Jaklitsch, WJ 1131-98*, **PRM 842991.**

USA. Illinois, Vermilion County, Jordan Creek, 40°4’24.492”N, 87°49’13.1514”W, on submerged decorticated woody debris, water 6.6 °C, pH 10.1, 3 Dec 2011, *Zelski S.E. and V.P. Hustad, JC0006-1*. Ohio, Cleveland Metroparks, Euclid Creek, 41°32’12.8034”N, 91°31’11.748”W, on submerged woody debris, water 21.1 °C, pH 9.4, 1263 µS, 18 Jul 2012, *Zelski S.E. and V.P. Hustad, Z57-1825;* Pennsylvania, Ricketts Glen State Forest, stream along Falls Trail, 41°20’1.356”N, 76°16’15.4194”W, 632m, on submerged woody debris, water 20.3 °C, pH 9, 632 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z57-1824;* Tennessee, GSMNP, riparian stream in Pigeon River watershed, 35°40'32.8434"N, 83°31'37.848"N, 536m, water 20 °C, pH 7.4, on submerged woody debris, 17 Jul 2010, *Zelski S.E. and H.A. Raja, Z57-1688.*

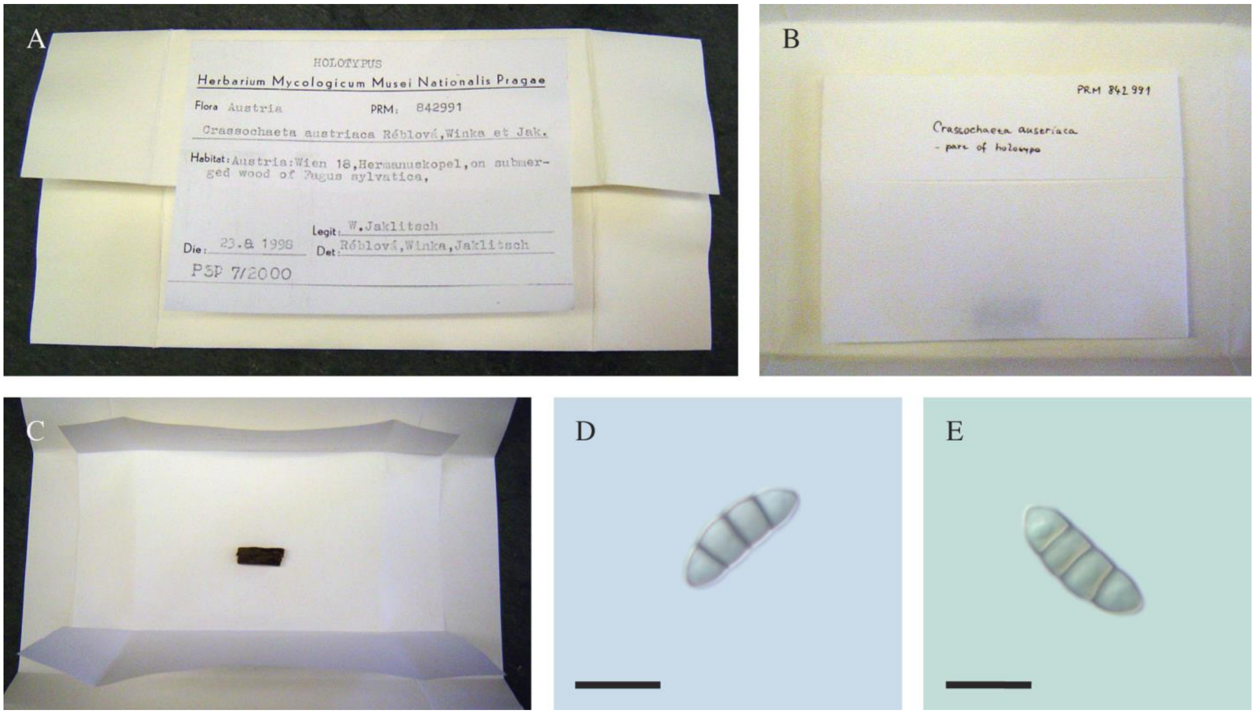
Additional specimens were examined from Canada (ON and MB), USA (AR, IL, ME, MN, MS, NC, NH, NY, OR, TN, VA, WI), and Venezuela.

Known distribution: Austria, Canada, USA, Venezuela.

Comments: The holotype of *Ascitendus austriacus* was in poor condition, only ascospores were located on an extremely small piece of dried substrate (**Fig. 30 A-C)**. The herbarium documentation suggests an initial tentative name of *Crassochaeta austriaca*, which is listed in MycoBank (MycoBank 491287, legitimate), though it was never formally published as such (**Fig. 30 B**). Members of *Crassochaet*a possess large, wedge-shaped apical rings, as opposed to those of those of *Asc. austriacus*, which are cylindrical to flaring. This fungus, however, was first placed in *Ascolacicola* as *Ascol. austriaca* (Réblová and Winka 2001), and later transferred to *Ascitendus* (Campbell and Shearer 2004). Early molecular work suggested *Asc. austriacus* was affiliated with Sordariales, Annulatascaceae, and/or Trichosphaeriaceae (Réblová and Winka 2001, Campbell and Shearer 2004). In these early analyses, *Asc. austriacus* clustered with *Annulatascus hongkongensis*, *Rhamphoria delicatula*, and the informally described *Aquaticola hongkongensis* (represented by GenBank sequences and the informal description in Ranghoo's thesis (1998)).

Fresh specimens of *Asc. austriacus* were collected and DNA was extracted for this study. Specimens collected in this study are a good match to those used in the first molecular study of the species (Campbell and Shearer 2004). Fresh specimens have brown ascospores with septa with central canals, the middle cells of the ascospores appear darker than the end cells, and there are longitudinal striations apparent on the surface of the ascospores (**Fig. 31 G-I**). This species is morphologically most similar to *A. aquatorba*, and Peruvian specimens (PE0255) were initially diagnosed as such. Molecular analyses place *A. aquatorba* as the taxon most closely related to *A. velatisporus*, the type of the genus, while *Asc. austriacus* is placed in Annulatascaceae *sensu stricto* as a separate genus (**Fig. 7**). An ex-type culture of *Asc. austriacus* is reported (CBS 102665), and it would be worthwhile to obtain sequences to compare European and North American species. Phylogenetically, the taxon most closely related at present to *Asc. austriacus* is *Annulusmagnus triseptatus* (**Figs. 7**). These species share the morphological features of three septate, brown pigmented ascospores, but *Asc. austriacus* has a smaller ascus apical apparatus and ascospores with darker septa compared to *An. triseptatus*.

Fig. 30 A-E. *Ascitendus austriacus* from the holotype **PRM 842991**. A. Herbarium packet. B. Envelope containing specimen (first tentative name *Crassochaeta austriaca*). C. Dried fragment of *Fagus sylvatica* with scant ascomata. D-E. Ascospores. Bars: D-E = 10 µm.



HOLOTYPUS

Herbarium Mycologicum Musei Nationalis Pragae

Flora Austria PRM: 842991

Crassochaeta austriaca Réblová,Winka et Jak.

Habitat: Austria:Wien 18,Hermanuskopel,on submerged wood of Fagus sylvatica,

Legit: W.Jaklitsch

Die: 23.8 1998 Det: Réblová,Winka,Jaklitsch

PSP 7/2000

PRM 842991

Crassochaeta austriaca
- part of holotype

Fig. 30.

Fig. 31 A-I. *Ascitendus austriacus* from specimen Z57-1824. A. Ascoma. B. Ascus. C. Ascus in cotton blue. D. Septate paraphyses. E. Wedge shaped annulus. F. Annulus end on. G-I. Ascospores. Bars: A = 100 µm; B-D = 20 µm; G-I = 10 µm; E-F = 5 µm.

Fig. 31.

Ascocollumdensa V.M. Ranghoo & K.D. Hyde, Ranghoo Ph.D. thesis, 64. 1998.

Type species: *Ascocollumdensa aquatica.*

Ascocollumdensa aquatica V.M. Ranghoo & K.D. Hyde, Ranghoo Ph.D. thesis, 66. 1998.

Ascomata hyaline, membranous, superficial, gregarious, 210–225 µm wide × 470–510 µm high ($\bar{x}$ = 217.5 × 490 µm, w:h = 1.3), globose to subglobose. **Ascomal wall** hyaline, 27.5–37 µm thick, composed of 5–7 layers of hyaline, thin walled, compressed pseudoparenchymatous cells. **Neck** hyaline, 230–250 µm long × 220–250 µm wide, covered with mass of hyaline setae, periphysate. **Paraphyses** hyaline, 5.6–6.6 µm wide at base × 120–150 µm long, septate, rarely branched. **Asci** unitunicate, cylindrical, 8–spored, 210–215 µm long × 12.5–15 µm wide ($\bar{x}$ = 212.5 × 13.8 µm, l:w = 12.8). **Ascus base** pedicellate, with a knob-like end, 15 µm from *pars sporifera*. **Annulus** J-, 2.5 µm long × 5 µm wide (l:w = .5). **Ascospores** hyaline, fusiform-ellipsoidal, 5–6–septate, not constricted at septa, overlapping uniseriate, 25–30 µm long × 6–10 µm wide ($\bar{x}$ = 27.5 × 8 µm, l:w = 3.4), without appendages or sheath (Ranghoo 1998).

HOLOTYPE: HONG KONG. New Territories, Plover Creek Reservoir, on submerged wood, Jan 1997, *V.M. Ranghoo,* **HKU(M) 5224.**

Known distribution: Hong Kong.

Comments: *Ascocollumdensa aquatica* is listed in MycoBank and was referred to that database from Index Fungorum. Index Fungorum considers this an invalid name (Art. 36.1(c) (Melbourne) which would mean it may be “merely cited as a synonym” (McNeill et al. 2012). No synonymy was uncovered in this study. The holotype was not examined in this study as Ranghoo’s thesis (1998) was only obtained near the completion of this manuscript. Thus, only the protologue was used to assess this species morphologically, and the 28S sequence was used for molecular analyses. Ranghoo noted that the thesis would only have 5 copies made and that due to this limited distribution, the ICBN would not allow new species to be considered valid (*nomen invalidum*) until published independently and broadly distributed. As this thesis is freely available from Hong Kong University at this time, this species is being included in this study for completeness, with all credit given to Ranghoo (1998). Early molecular analyses placed *Ascoc. aquatica* in a polytomous clade with putative Annulatascaceae taxa (Ranghoo et al. 1999) or basal to the Lulworthiales (Jones et al. 2014). Preliminary molecular analyses during the course of this study confirmed that *Ascoc. aquatica* does not belong in Annulatascaceae *sensu stricto* (results not shown). Morphologically, this species possesses hyaline membranous ascomata with densely setose necks, features which are not found in any of the taxa in the family. *Ascoc. aquatica* should be considered Sordariomycetes incertae sedis.

Ascolacicola Ranghoo & K.D. Hyde, Mycologia 90: 1055. 1998. MycoBank 27878.

Type species: *Ascolacicola aquatica.*

Ascolacicola aquatica Ranghoo & K.D. Hyde, Mycologia 90(6): 1056. 1998. **Fig. 32 A-B.** MycoBank 4461951.

Ascomata black, coriaceous, superficial, perpendicular to parallel to surface, solitary to gregarious, 250–375 µm wide × 225–275 µm high (x̅ = 312.5 × 250 µm, w:h = 1.25), subglobose. **Ascomal wall** brown, wall 5–7 layers of brown flattened angular cells, 18–27.5 µm thick. **Neck** black, 175–200 µm long × 62.5–75 µm wide (x̅ = 187.5 × 68.8 µm, l:w = 2.7), periphysate. **Paraphyses** hyaline, 2.5–3.8 µm wide at base × 175–200 µm long, apically rounded, septate, filamentous. **Asci** unitunicate, cylindrical, 8–spored, 125–163 µm long × 10–12.5 µm wide (x̅ = 144 × 11.3 µm, l:w = 12.8). **Ascus base** pedicellate, short, club-like, 30 µm from *pars sporifera*. **Annulus** J-, discoid, 2.5–3.7 µm long × 6.2 µm wide (x̅ = 3.1 × 6.2

µm, l:w = .5). **Ascospores** brown with pale end cells, ellipsoidal, 3–septate, not constricted at septa, uniseriate, 12.5–16.5 µm long × 4–7.5 µm wide ($\bar{x}$ = 14.5 × 5.8 µm, l:w = 2.5), guttulate, without appendages or sheath (Ranghoo 1998).

Specimens examined:

HOLOTYPE: HONG KONG. The New Territories, Plover Cove Reservoir, on submerged wood, Mar 1997, *V.M. Ranghoo, (HKU(M)5542),* **IFRD 8693.**

Known distribution: Hong Kong.

Comments: The holotype material was devoid of fungal material of *Ascolacicola aquatica* (**Fig. 32 A,B**). Fresh specimens were not obtained in this study. It is recommended that fresh material for neotypification be collected from the type location. *Ascol. aquatica* possesses ascospores that are 4-celled with brown pigmentation centrally and hyaline end cells. These features might suggest affiliation with *Ascitendus austriacus* with versicolored ascospores, or *Annulusmagnus triseptatus* which has ascospores that gradually become light brown, or even *Ascotaiwania* species. However, previous molecular analyses have shown *Ascol. aquatica t*o be a sister taxon to *Sordaria fimicola* (Ranghoo 1999) or even basal to the Helvellalaceae (Raja et al. 2003) and early analyses in this study (results not shown) have shown *Ascol. aquatica* is distant from Annulatascaceae *sensu stricto* and is considered Sordariomycetes *incertae sedis*.

Fig. 32 A-B. *Ascolacicola aquatica* from the holotype **IFRD 8693**. A. Herbarium packet. B. Dried substrate.

International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8693
Fungi Name	Ascolacicola aquatica
Host	on Submerged wood
Collection Site	Hong Kong.The New Territories. Plover Creek reservoire
Collection Date	Mar 1997
Collector	V.M. Ranghoo
Remarks	Holotype

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

Fig. 32.

Ayria Fryar & K.D. Hyde, Cryptogamie Mycologie 25(3): 248. 2004. MycoBank28835.

Type species: *Ayria appendiculata.*

Ayria appendiculata Fryar and K.D. Hyde, Cryptogamie Mycologie 25(3): 248. 2004. **Fig. 33 A-E.**

MycoBank 369503.

Ascomata light to dark brown, membranous, immersed to erumpent to superficial, solitary, 150 µm wide, globose to subglobose (**Fig. 33 C**). **Ascomal wall** light brown to dark brown, outer layer of brown cells, inner layer of hyaline flattened cells, 10 µm thick, of *textura intricata* in surface view. **Neck** 150 µm long × 44 µm wide. **Paraphyses** hyaline, 5–5.5 µm wide at base, septate, constricted at septa, sparse. **Asci** unitunicate, clavate, 8–spored, 115–150 µm long × 16–29 µm wide ($\bar{x}$ = 132.5 × 22.5 µm, l:w = 5.9) (**Fig. 33 D**). **Ascus base** pedicellate, 50 µm from *pars sporifera*. **Annulus** lacking. **Ascospores** hyaline, ellipsoid, aseptate, biseriate, 21–26 µm long × 7.5–10 µm wide ($\bar{x}$ = 23.5 × 8.75 µm, l:w = 2.7), multiguttulate, with bipolar unraveling appendages, without sheath (**Fig. 33 E**) (Fryar and Hyde 2004).

Specimens examined:

HOLOTYPE: BRUNEI. Sungai Kelakas, Tutong River, Site 2, 4°48’59.06”N, 114°40’47.78”E, on submerged rotting wood, 1998, *S.C. Fryar, (HKU(M) 15553),* **IFRD 8706.**

ECUADOR. Napo, Yasuni National Park, 0°39’30”S, 76°22’32”W, water 27 °C, pH 6, 30 Sep 2005, *A. Ferrer*, *AF227-1, ILL 40777*.

Known distribution: Brunei, Ecuador.

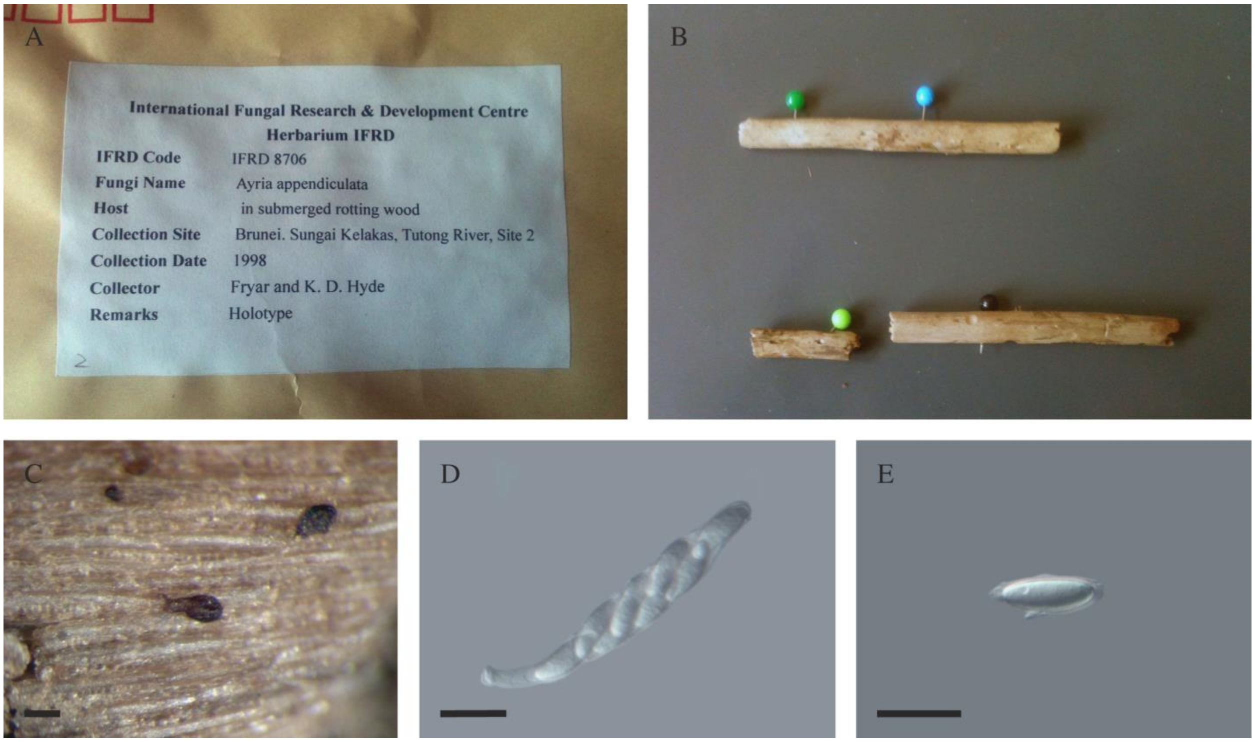
Comments: The holotype specimen of *Ayria appendiculata* is in fair condition, with only a few asci and ascospores recovered (**Fig. 33 A,B**). Three permanent slides were prepared, examined, and returned to

IFRD. Fryar and Hyde (2004) tentatively placed *Ay. appendiculata* in Annulatascaceae noting that molecular analysis should be performed. The fungus was found in two brackish and one marine site in their ecological study. Of note is that this species lacks an ascus apical ring even though affinity to Annulatascaceae was suggested (Fryar and Hyde 2004, Raja pers. comm.). Fresh specimens resembling *Ayria* (PE0323) were collected in this study, and sequence data for a second species, *Ay. nubispora* (F99), was available. *Ay. nubispora* is distinguished by having smaller ascospore dimensions. Peruvian specimens (PE0323) were more similar to *Ay. appendiculata*, though they were collected in freshwater rather than brackish habitats and had smaller ascomata. Neither *Ay. nubispora* nor PE0323 were related to Annulatascaceae *sensu stricto* nor to one another (**Fig. 7; Clades 1c, 7**). As no type culture was noted, fresh specimens from the Tutong River in Brunei are needed to assess the phylogenetic relationships of these taxa with similar ascospores. One species in Annulatascaceae *sensu stricto* has ascospores with these distinctive bipolar unraveling appendages, *Pseudoproboscispora caudae-suis*, though its spores are 1-septate. Other species with this feature also occur in the Halosphaeriales, though neither taxon in this study was placed in the Halosphaeriales clade. *Ay. appendiculata* should be considered Sordariomycetes *incertae sedis*.

Ayria nubispora Raja, A. Ferrer & Shearer, Fungal Diversity 34: 81. 2009. **Fig. 34 A-I.** MycoBank 512705.

Ascomata dark brown, membranous, semi-immersed to superficial, scattered, 250–270 µm wide × 158–168 µm long (x̅ = 260 × 163 µm, w:h = 1.6), globose to subglobose. **Ascomal wall** hyaline and brown, 18–20 µm thick, outer layer of dark brown isodiametric cells occluded by brown amorphous material, inner layer of hyaline elongated cells, of *textura angularis* in surface view (**Fig. 34 E**). **Neck** black at base to hyaline at apex, 40–50 µm long × 30–60 µm wide (x̅ = 45 × 45 µm, l:w = 1). **Paraphyses** hyaline, 7–9 µm wide × 90–120 µm long, tapering, septate, constricted at septa. **Asci** unitunicate, cylindrical to

Fig. 33 A-E. *Ayria appendiculata* from the holotype **IFRD 8706**. A. Herbarium packet. B. Dried substrate. C. Ascomata on substrate. D. Ascus. E. Ascospore. Bars: C = 100 μm; D-E = 20 μm.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8706
Fungi Name	Ayria appendiculata
Host	in submerged rotting wood
Collection Site	Brunei. Sungai Kelakas, Tutong River, Site 2
Collection Date	1998
Collector	Fryar and K. D. Hyde
Remarks	Holotype

2

Fig. 33.

clavate, 8–spored, 212–250 µm long × 34–40 µm wide (x̅ = 231 × 37 µm, l:w = 6.2). **Ascus base** pedicellate, short, 5 µm from *pars sporifera*. **Annulus** lacking. **Ascospores** hyaline, ellipsoidal to fusiform, aseptate, overlapping uniseriate to biseriate, 48–56 µm long × 16–20 µm wide (x̅ = 52 × 18 µm, l:w = 2.9), multiguttulate, with bipolar unraveling appendages, with a 10–18 µm wide sheath (**Fig. 34 F-I**) (Raja et al. 2009).

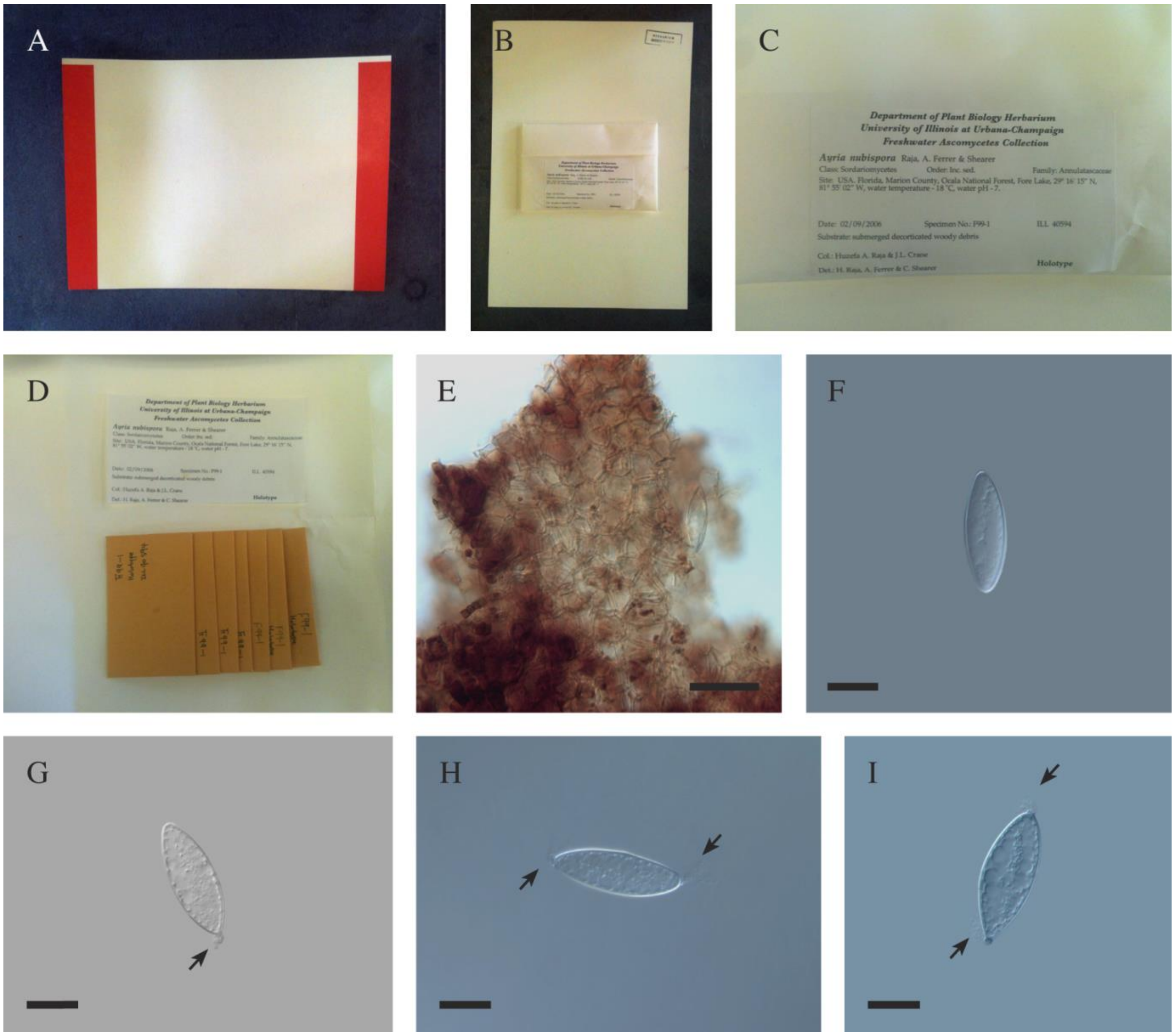
Specimens examined:

HOLOTYPE: USA. Florida, Marion County, Ocala National Forest, Fore Lake, 29°16'15"N, 81°55'02"W, on submerged woody debris, water 18 °C, pH 7, 9 Feb 2006, *H.A. Raja and J.L. Crane, F99-1*, ILL 40594. COSTA RICA. Heredia, Stream at La Selva, 10°25'7.26"N, 84°1'27.3"W, on submerged wood, water 25 °C, pH 5, 10 Jan 2006, *M. Salazar, AF268-1, ILL 40785;* Stream at La Selva, 10°25'43.86"N, 84°0'17.34"W, water 25 °C, pH 5, 9 Jan 2006, *M. Salazar, AF268-3, ILL 40787;* Limon, Las Palmas stream, 10°35'56.52"N, 83°31'52.62"W, water 25 °C, pH 5, 18 Dec 2005, *A. Ferrer and M. Salazar, AF268-2, ILL 40786*. ECUADOR. Napo, Yasuni National Park, 0°39'30"S, 76°22'32"W, water 27 °C, pH 6, 30 Sep 2005, *A. Ferrer, AF227-1, ILL 40777*.

Known distribution: Costa Rica, Ecuador, USA.

Comments: The holotype of *Ayria nubispora* contained only ascomatal remnants and few ascospores (**Fig. 34 A-D**). The protologue, however, has good illustrations (Raja et al. 2009). Fresh specimens of *Ay. nubispora* were not obtained in this study, though an *Ayria*-like species (PE0323) with aseptate ascospores and unfurling bipolar appendages was found in Perú. Preserved specimens of *Ay. nubispora* from Ecuador (AF268) only yielded ascospores. The type culture (F99-1) was dead and thus other genes could not be sequenced. The molecular analyses of this study place PE0323 sister to an *Annulatascus*-like Peruvian species (PE0324) with good support, removed from Annulatascaceae *sensu stricto* and in a clade containing orders such as Coronophorales, Hypocreales and Savoryellales (**Fig. 7, Clade 7**). In this study

Fig. 34 A-I. *Ayria nubispora* from the holotype **ILL 40594**. A. Herbarium folder. B. Herbarium packet. C. Holotype information. D. Holotype material (7 slides). E. Peridium illustrating large polygonal cells. F-I. Ascospores (remnants of appendages arrowed). Bars: E = 100 µm; F-I = 20 µm.



Department of Plant Biology Herbarium
University of Illinois at Urbana-Champaign
Freshwater Ascomycetes Collection

Ayria nubispora Raja, A. Ferrer & Shearer
Class: Sordariomycetes Order: Inc. sed. Family: Annulatascaceae
Site: USA. Florida, Marion County, Ocala National Forest, Fore Lake, 29° 16' 15" N, 81° 55' 02" W, water temperature - 18 °C, water pH - 7.

Date: 02/09/2006 Specimen No.: F99-1 ILL 40594
Substrate: submerged decorticated woody debris

Col.: Huzefa A. Raja & J.L. Crane
Det.: H. Raja, A. Ferrer & C. Shearer **Holotype**

Fig. 34.

Ay. nubispora clusters with *Aquaticola*-like species (PE0157, PE0264) in a clade between Ophiostomatales and Diaporthales (**Fig. 7, Fig. 1c**), but this clade is distant from PE0323. Additional collections from Florida are needed to further assess the species phylogenetically. At this time *Ay. nubispora* should be considered Sordariomycetes *incertae sedis*.

Brunneosporella Ranghoo & K.D. Hyde, Mycological Research 105 (5): 625. 2001. MycoBank 28492.

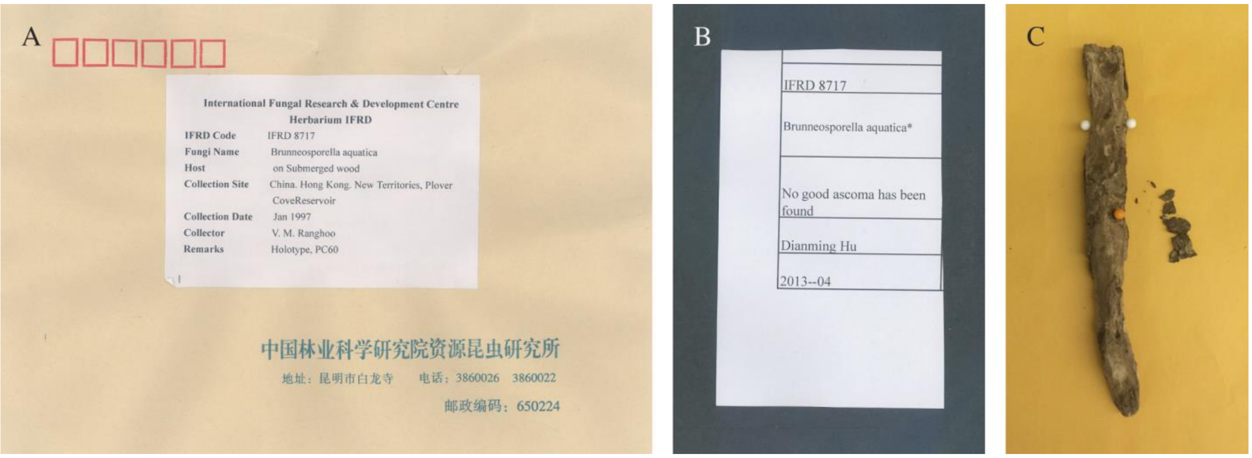
Type species: *Brunneosporella aquatica.*

Brunneosporella aquatica V.M. Ranghoo & K.D. Hyde, Mycological Research 105(5): 625. 2001. **Fig. 35 A-C.** MycoBank 467768.

= *Ascobrunneispora aquatica* V.M. Ranghoo & K.D. Hyde, Ranghoo Ph.D. thesis, 58. 1998.

Ascomata dark brown, membranous, immersed, perpendicular to surface, solitary to gregarious, 250–275 µm wide × 175–188 µm high (x̅ = 2662.5 × 181.5 µm, w:h = 1.5), globose to subglobose. **Ascomal wall** light brown, 20–25 µm thick, composed of several layers of light brown, angular elongate pseudoparenchymatous cells, of *textura angularis* in surface view. **Neck** dark brown, 130–150 µm long × 35–40 µm wide, periphysate. **Paraphyses** hyaline, 4–5 µm wide at base × 150–163 µm long, tapering toward apex, septate, flexuose. **Asci** unitunicate, cylindrical, 8–spored, 150–175 µm long × 10–13 µm wide (x̅ = 162.5 × 11.5 µm, l:w = 14.1). **Ascus base** pedicellate, cylindrical, rounded at tip, 15 µm from *pars sporifera*. **Annulus** J-, discoid, 2–3 µm long × 2–3 µm wide (x̅ = 2.5 × 2.5 µm, l:w = 1). **Ascospores** brown, ellipsoidal to fusiform, 1–septate, slightly constricted at septum, uniseriate to overlapping uniseriate 17.5–20 µm long × 9–10 µm wide (x̅ = 18.75 × 9.5 µm, l:w = 2), multiguttulate, without appendages or sheath (Ranghoo et al. 2001).

Fig. 35 A-C. *Brunneosporella aquatica* from the holotype **IFRD 8717**. A. Herbarium packet. B. Note from Dianming Hu stating that no good ascomata were found on the substrate. C. Dried material.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8717
Fungi Name	Brunneosporella aquatica
Host	on Submerged wood
Collection Site	China. Hong Kong. New Territories, Plover CoveReservoir
Collection Date	Jan 1997
Collector	V. M. Ranghoo
Remarks	Holotype, PC60

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

IFRD 8717
Brunneosporella aquatica*
No good ascoma has been found
Dianming Hu
2013--04

Fig. 35.

Specimens examined:

HOLOTYPE: CHINA. Hong Kong, New Territories, Plover Cove Reservoir, on submerged wood, Jan 1997, *V.M. Ranghoo, PC 60, (HKU(M) 5251),* **IFRD 8717.**

Known distribution: Australia, China.

Comments: The holotype of *Brunneosporella aquatica* is in poor condition **(Fig. 35 A-C).** The holotype came with a note that no good ascomata were found by IFRD (Dianming Hu) (**Fig. 25 B**). Examination of the holotype in this study corroborates this. It is recommended that neotypification be done from fresh material collected in China. This species was originally informally published as a new member of Annulatascaceae under the name *Ascobrunneispora aquatica* as in the Ph.D. thesis of Ranghoo (1998). Of morphological note is that *B. aquatica* has small discoid apical rings rather than large distinct rings as are found in the majority of species in Annulatascaceae. This species had previously been shown (Raja 2003, Abdel-Wahab et al. 2011) to be removed from Annulatascaceae *sensu stricto* and the same results were arrived at in this study. *B. aquatica* is placed in a well supported clade with *Fluminicola bipolaris*, which has hyaline ascospores, and *Papulosa amerospora*, which has J+ ascus apical rings (Winka and Eriksson 2000) in combined three gene analyses (**Fig. 7, Clade 1a**). *B. aquatica* should be considered Sordariomycetes *incertae sedis*.

Cataractispora K.D. Hyde, S.W. Wong & E.B.G. Jones, Mycological Research 103(8): 1019. 1999. MycoBank 28328.

Type species: *Cataractispora aquatica.*

Cataractispora appendiculata K.D. Hyde, S.W. Wong, & E.B.G. Jones, Mycological Research 103(8): 1025. 1999. **Fig. 36 A-K, Fig. 37 A-G.** MycoBank 460660.

Ascomata brown to black, coriaceous, immersed, semi-immersed, to superficial, perpendicular to parallel to surface, solitary, 224–390 µm wide × 128–167 µm high (x̅ = 307 × 147.5 µm, w:h = 2.1), globose, subglobose, or ellipsoidal. **Ascomal wall** hyaline and brown, outer layer of brown compressed cells and inner layer of hyaline compressed cells (**Fig. 36 E**). **Neck** brown to black, short, lateral. **Paraphyses** hyaline, 5 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 129–195 µm long × 11–20 µm wide (x̅ = 162 × 15.5 µm, l:w = 10.5) (**Fig. 37 A**). **Ascus base** pedicellate, tail elongate, 20 µm from *pars sporifera* (**Fig. 36 F, Fig. 37 B**). **Annulus** J-, bipartite, 4.5 µm long × 6 µm wide (x̅ = 4.5 × 6 µm, l:w = .75) (**Fig. 36 G,H; Fig. 37 C,D**). **Ascospores** hyaline, fusiform, aseptate, overlapping uniseriate to biseriate, 23–30 µm long × 7–10 µm wide (x̅ = 26.5 × 8.5 µm, l:w = 3.1), multiguttulate, with bipolar unraveling appendages, without a sheath (**Fig. 36 I-K, Fig. 37 E-G**) (Hyde et al. 1999).

Specimens examined:

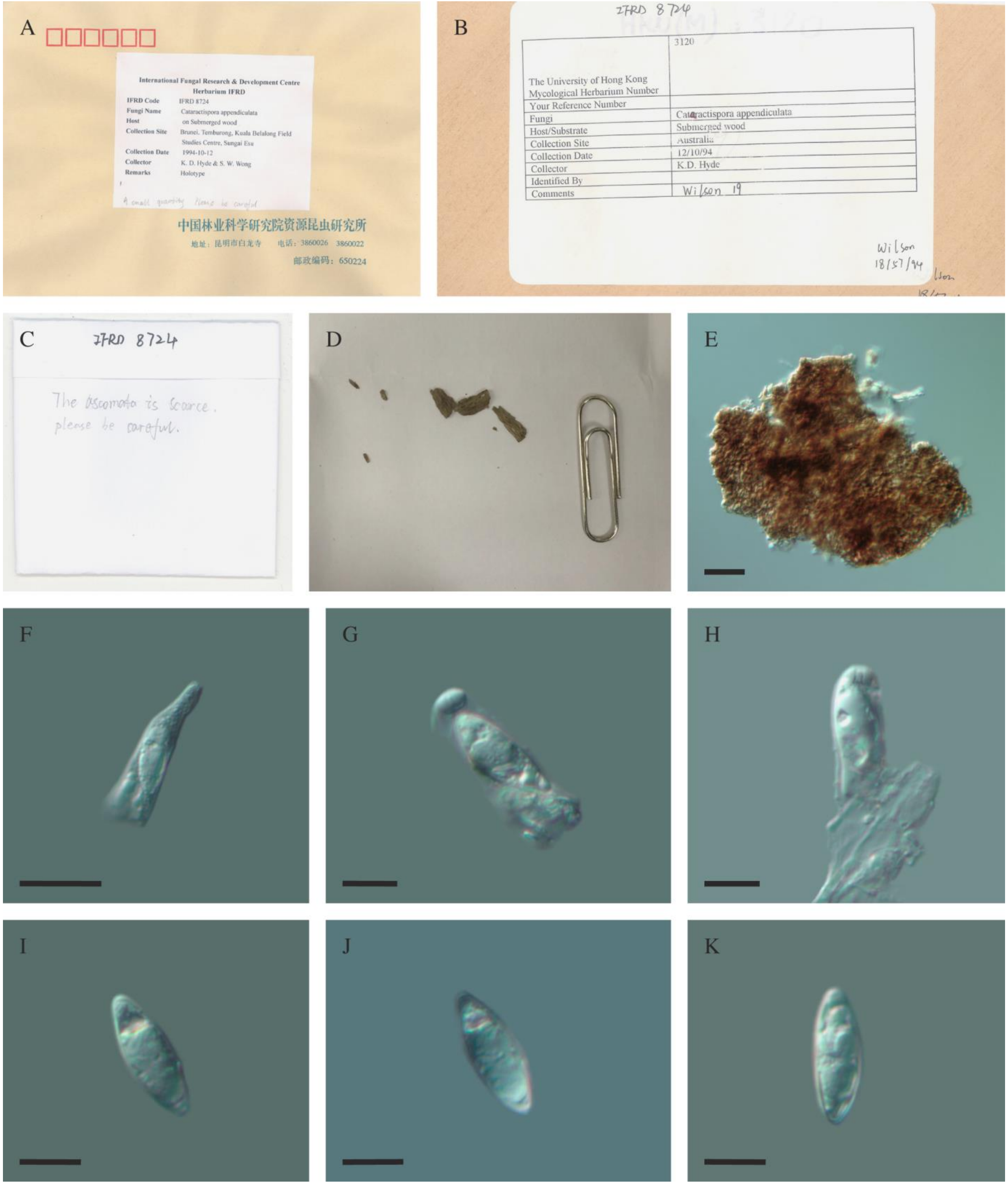
HOLOTYPE: BRUNEI. Temburong, Kuala Belalong Field Studies Centre, Sungai Esu, on submerged wood, Feb 1994, *K.D. Hyde and S.W. Wong, (HKU(M) 3120)*, **IFRD 8724.**

PERU. Madre de Dios, palm swamp (aguajal) near the Interoceanic Highway near Puerto Maldonado, trail head 12°42’48.0954”S, 69°28’11.28”W, 239m, water 23.3 °C, pH 5.9, 20 May 2010, *Zelski S.E. and H.A. Raja*, *PE0066-1*.

Known distribution: Brunei, Perú.

Comments: The holotype material of *Cataractispora appendiculata* was extremely scant with fewer than 5 ascomata observed (**Fig. 36 A-D**). In this study, one permanent slide was made of ascomal remnants, ascus fragments, and ascospores, examined, and returned with the holotype. Ascospores did not exhibit

Fig. 36 A-K. *Cataractispora appendiculata* from the holotype **IFRD 8724**. A. Herbarium packet. B. Original herbarium packet from HKU. C. Folder containing dried material. D. Scant amount of dried substrate. E. Peridium. F. Ascus base. G-H. Ascus apices illustrating large apical rings. I-K. Ascospores. Bars: E-F = 20 µm; G-K = 10 µm.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8724
Fungi Name	Cataractispora appendiculata
Host	on Submerged wood
Collection Site	Brunei, Temburong, Kuala Belalong Field Studies Centre, Sungai Esu
Collection Date	1994-10-12
Collector	K. D. Hyde & S. W. Wong
Remarks	Holotype

A small quantity Please be careful

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224

IFRD 8724

The University of Hong Kong Mycological Herbarium Number	3120
Your Reference Number	
Fungi	Cataractispora appendiculata
Host/Substrate	Submerged wood
Collection Site	Australia
Collection Date	12/10/94
Collector	K.D. Hyde
Identified By	
Comments	Wilson 19

Wilson
18/57/94

IFRD 8724

The ascomata is scarce.
please be careful.

Fig. 36.

Fig. 37 A-G. *Cataractispora appendiculata* from specimen PE0066-1. A. Ascus. B. Ascus base. C. Ascus apices illustrating large apical rings. D. Apical ring seen end on. E-F. Ascospores. G. Ascospore illustrating coiled appendage. Bars: A-B, E-G = 20 µm; C-D = 10 µm.

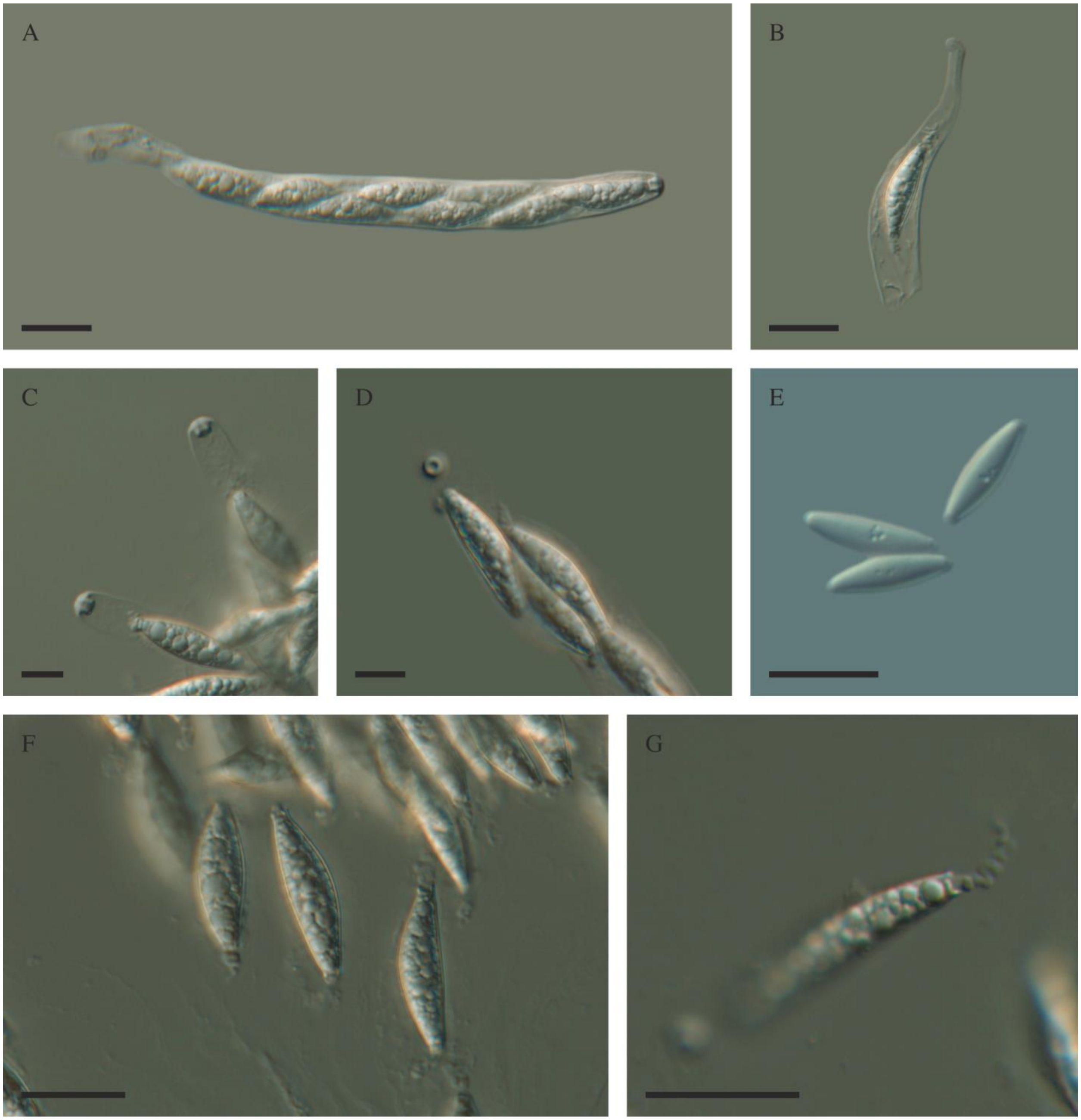


Fig. 37.

bipolar appendages when revived in KOH, but apical rings were well preserved. Due to the poor condition of the holotype material neotypification of this species should be performed. Peruvian specimens of *C. appendiculata* matched the type specimen well. *C. appendiculata* clusters in a clade with *Aquaticola hyalomura*, *C. receptaculorum*, and *C. viscosa* (**Fig. 7, Clade 1b**). This clade is not affiliated with Annulatascaceae *sensu stricto* thus *C. appendiculata* and the other species are removed from the family herein. Though the type of the genus, *C. aquatica*, has not been evaluated molecularly, the genus and these species should be considered Sordariomycetes *incertae sedis*. Fresh material of *C. aquatica* from Brunei should be collected to obtain molecular information for the type of the genus.

Cataractispora aquatica K. D. Hyde, S. W. Wong & E. B. G. Jones, Mycological Research 103 (8): 1020. 1999. **Fig. 38 A-J.** MycoBank 460659.

Ascomata dark brown to black, coriaceous, immersed to superficial, perpendicular to parallel to surface, solitary, 240–370 µm wide, globose to subglobose. **Ascomal wall** hyaline and brown, outer layer of brown compressed cells and inner layer of hyaline compressed cells. **Neck** black, short. **Paraphyses** hyaline, 3.3 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, long cylindrical, 8–spored, 344–366 µm long × 11–12 µm wide ($\bar{x}$ = 355 × 11.5 µm, l:w = 31) (**Fig. 38 E**). **Ascus base** pedicellate, short, club-like, 10 µm from *pars sporifera*. **Annulus** J-, refractive, bipartite, 5 µm long **×** 6 µm wide (l:w = .83) (**Fig. 38 F-H**). **Ascospores** hyaline, fusiform, 0–5 septate, uniseriate to overlapping uniseriate, 36–65 µm long × 8–14 µm wide ($\bar{x}$ = 50.5 × 11 µm, l:w = 4.6), with a large guttule in each cell, with bipolar unraveling appendages, without sheath (**Fig. 38 I,J**) (Hyde et al. 1999).

Specimens examined:

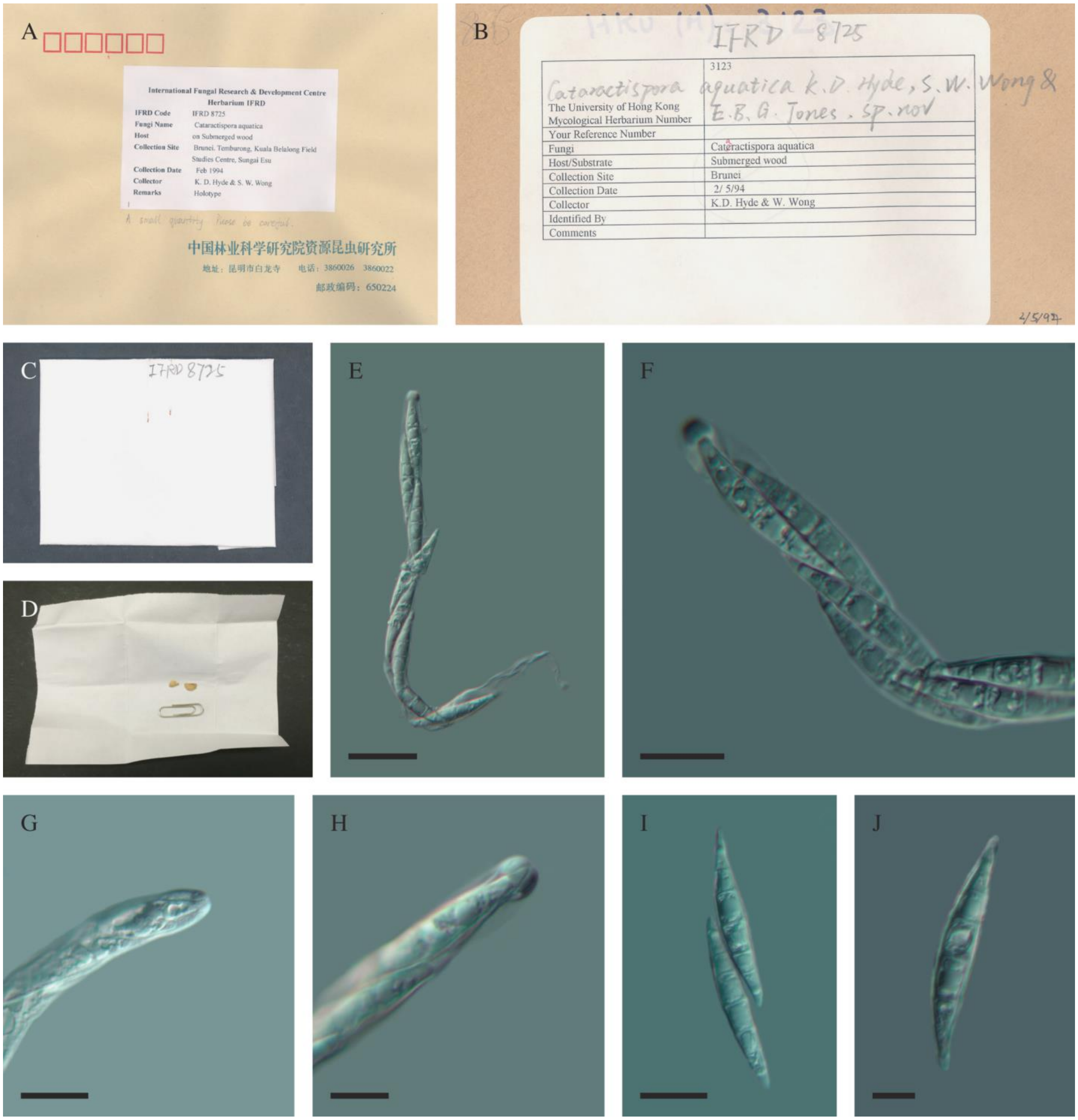
HOLOTYPE: BRUNEI. Temburong, Kuala Belalong Field Studies Centre, Sungai Esu, on submerged wood, Feb 1994, *K D. Hyde & S.W. Wong, (HKU(M) 3123)*, **IFRD 8725.**

PERU. Cusco, Camanti, Quincemil, stream at Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.2 °C, pH 6.8, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0189-1;* Stream crossing Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 653m, on submerged woody debris, water 21.7 °C, pH 7.6, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0189-2;* River at end of Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.6 °C, pH 8.3, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0189-4;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 20.5 °C, pH 7.2, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0189-3;* Stream at Quincemil Trail 3, on submerged decorticated wood, trailhead 13°18’27.756”S, 70°48’44.9279”W, 757m, water 21.5 °C, pH 7, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0189-5.* Madre de Dios, CICRA, Semi-aquatic habitat on Trail 1, 12°34’06.52”S, 70°06’04.57”W, 263m, on submerged woody debris, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0189-6.*

Known distribution: Australia, Brunei, Seychelles.

Comments: The holotype material of *Cataractispora aquatica* is extremely scant (**Fig. 38A-D**). One permanent slide made in this study, examined, and returned with the herbarium packet. Only one ascoma was found on the substrate. Paraphyses were not well preserved. Ascospores were agglomerated in the positions they were in when the asci dried, perhaps due to the bipolar appendages binding them together. The apex of the neck appeared white. Ascospore appendages were not well preserved, appearing as short extensions from the bipolar apical chambers. Fresh collections from Brunei should be made for neotypification and obtaining molecular data. Ascospores from fresh material in the collections failed to germinate. *C. aquatica* is most similar to *C. viscosa* morphologically, though it does not have a hyaline neck. *C. viscosa*, *C. appendiculata* and *C. receptaculorum*, form a clade that includes *Aquaticola hyalomura* in the combined three gene analyses (**Fig. 7, Clade 1b**). This clade is not affiliated with

Fig. 38 A-J. C*ataractispora aquatica* from the holotype **IFRD 8725**. A. Herbarium packet. B. Original HKU herbarium packet. C. Envelope with dried substrate inside. D. Scant amount of dried material. E. Ascus. F. Ascospores in ascus. G-H. Ascus apices illustrating large apical rings. I-J. Ascospores. Bars: E = 40 µm; F-G, I = 20 µm; H,J = 10 µm.



International Fungal Research & Development Centre
Herbarium IFRD
IFRD Code IFRD 8725
Fungi Name Cataractispora aquatica
Host on Submerged wood
Collection Site Brunei. Temburong. Kuala Belalong Field Studies Centre, Sungai Esu
Collection Date Feb 1994
Collector K. D. Hyde & S. W. Wong
Remarks Holotype

A small quantity. Please be careful.

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224

HKU (M) 3123
IFRD 8725

Cataractispora The University of Hong Kong Mycological Herbarium Number	3123 aquatica K.D. Hyde, S.W. Wong & E.B.G. Jones. sp. nov
Your Reference Number	
Fungi	Cateractispora aquatica
Host/Substrate	Submerged wood
Collection Site	Brunei
Collection Date	2/ 5/94
Collector	K.D. Hyde & W. Wong
Identified By	
Comments	

2/5/94

IFRD 8725

Fig. 38.

Annulatascaceae *sensu stricto*. Until *C. aquatica* can be evaluated molecularly, it should be considered Sordariomycetes *incertae sedis*.

Cataractispora bipolaris (K.D. Hyde) K.D. Hyde, S.W. Wong & E.B.G. Jones, Mycological Research 103(8): 1031. 1999. **Fig. 39 A-L.** MycoBank 460662 (*Annulatascus bipolaris* MycoBank 355477, legitimate).

≡ *Annulatascus bipolaris* K.D. Hyde, Australian Systematic Botany 5: 120. 1992.

Ascomata black, coriaceous, immersed, semi-immersed to superficial, solitary, 155–235 µm wide, globose to subglobose. **Ascomal wall** outer layer of elongate, angular, melanized cells, lighter toward interior (**Fig. 39 F**). **Neck** long, black, lateral, periphysate. **Paraphyses** hyaline, 4 µm wide at base, tapering toward apex, septate, longer than asci. **Asci** unitunicate, cylindrical, 8–spored, 174–205 µm long × 9–10.5 µm wide ($\bar{x}$ = 190 × 9.75 µm, l:w = 18.5) (**Fig. 39 G,H**). **Ascus base** pedunculate, 30 µm from *pars sporifera* (F**ig. 39 H**). **Annulus** J-, 4 µm long × 6 µm wide, (l:w = .66) (**Fig. 39 I**). **Ascospores** hyaline, fusiform, aseptate, uniseriate, 21–30 µm long × 6.5–8.5 µm wide ($\bar{x}$ = 25.5 × 7.5 µm, l:w = 3.4), with polar pad-like appendages extending to strands, without a sheath (**Fig. 39 J-L**) (Hyde 1992).

Specimens examined:

HOLOTYPE: AUSTRALIA. North Queensland, Clohesy River, on submerged wood, in the Koah River tributary, Nov 1990, *K.D. Hyde 361*, **BRIP 27374.**

Known distribution: Australia.

Fig. 39 A-J. *Cataractispora bipolaris* from the holotype **BRIP 17374**. A. Herbarium packet. B. BRIP documentation. C. Shearer correspondence. D. Holotype collection information. E. Dried substrate showing circled area (arrow) where the fungus was located. F. Peridium of *textura angularis*. G-H. Asci. I. Ascus apices illustrating large apical rings. J. Ascospore with bipolar appendages. K-L. Ascospores. Bars: F-H = 20 µm; I-L = 10 µm.

QUEENSLAND PRIMARY INDUSTRIES & FISHERIES
PLANT PATHOLOGY HERBARIUM (BRIP)

BRIP 17374 (Holotype)

Ascomycota
Sordariomycetes

Cataractispora bipolaris (K.D. Hyde) K.D. Hyde, S.W. Wong & E.B.G. Jones

Substrate: Wood
Locality: Tablelands, Koah River QLD Australia
Collector(s): K.D. Hyde
Coll. No: 361 **Coll. Date:** Nov 1990
Determiner: K.D. Hyde

17374

University of Illinois
at Urbana-Champaign

Department of Plant Biology

Dr. J.L. Alcorn
Plant Pathology Bldg
Department of Primary Industries
Meiers Road
Indooroopilly Q 4068
Australia

13 September, 2000

Dear Dr. Alcorn:

I am sorry about holding on to these specimens so long. I was not notified of their arrival and then I had to pack and unpack my laboratory twice. Recently, I moved my laboratory and have been going through everything as I unpack. I found the specimens about two weeks ago and immediately examined them.

The specimen of *Annulatascus velatisporus* (note spelling change – the species name had an incorrect ending) had numerous fruiting bodies. I could find no ascomata on the collection of *Annulatascus bipolaris* so you might want to notify Kevin so he can provide some slides or a neotype.

Thank you very much for sending the specimens and again I apologize for the delay in returning them.

With best regards,
Sincerely,

Carol Shearer

Carol Shearer
Professor

17373
17374

BRIP 17374

HOST

Submerged wood
LOCALITY Tablelands Nth Qld Koah River Qld Australia

DATE Nov 1990
COLL. K.D. Hyde 361
DET. K.D. Hyde
Holotype
DISEASE
REMARKS Aust. Syst. Bot. 5: 117, 1992.

Fig. 39.

Comments: The holotype of *Cataractispora bipolaris* contained one ascoma in good condition which was made into a permanent slide, examined, and returned to BRIP (**Fig. 39 A-E**). In 2000, Shearer obtained the holotype of *C. bipolaris*. According to the correspondence within the herbarium packet, efforts to locate ascomata of the fungus were unsuccessful. In this study, a small amount of water was added to the substrate using a sterile syringe, which revitalized the substrate so that one immersed ascoma was located within the zone demarcated on the dried wood. After rehydration in water, images were taken. Fresh collection from the Clohesy River in Australia for the purpose of obtaining DNA would assist in evaluating the phylogenetic placement of this species in relation to the type of the genus, *C. aquatica*, for which molecular data are also lacking. The three *Cataractispora* species evaluated in this study form a monophyletic group without Bayesian or Maximum-likelihood support in combined three gene analyses that also includes *Aquaticola hyalomura* (**Fig. 7, Clade 1b**). These species are not affiliated with Annulatascaceae *sensu stricto*. This leaves the question open as to the true placement of *Cataractispora* based on the type species. *C. bipolaris* should be considered Sordariomycetes *incertae sedis*.

Cataractispora receptaculorum W.H. Ho, K.D. Hyde & I.J. Hodgkiss, Mycologia 96(2): 411. 2004. MycoBank 488579.

Ascomata dark brown, coriaceous, immersed, perpendicular to surface, solitary, 275–300 µm wide × 250–275 µm high (x̅ = 288 × 263 µm, w:h = 1.1), globose to subglobose. **Ascomal wall** light brown to brown, 17–23 µm thick, 2–3 outer layers of angular brown cells and inner layer of light brown compressed cells, of *textura angularis* in surface view. **Neck** dark brown, 250–275 µm long × 113–125 µm wide. **Paraphyses** hyaline, 4–5 µm wide at base × 175–200 µm long, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 213–233 µm long × 15–22.5 µm wide (x̅ = 223 × 18.8 µm, l:w = 11.9). **Ascus base** pedicellate, broadly tapering, 50 µm from *pars sporifera*. **Annulus** J-, refractive, 5–7.5

µm long × 3.8–4 µm wide (x̅ = 6.25 × 3.9 µm, l:w = 30.9). **Ascospores** hyaline, ellipsoidal, 3–septate, not constricted at septa, uniseriate to overlapping uniseriate, 27.5–31 µm long × 7.5–10 µm wide (x̅ = 29.25 × 8.75 µm, l:w = 3.3), guttulate, with appendages developing from pads to strands, without sheath (Ho et al. 2004).

HOLOTYPE: HONG KONG. Plover Cove Reservoir, on bamboo submerged in freshwater, 15 Feb 1997, *V.M. Ranghoo, PC38,* **HKU(M) 5239.**

Comments: The holotype of *Cataractispora receptaculorum* was unavailable for examination. The protologue must serve for comparisons until neotypification can be performed. Fortunately, GenBank sequence data is available. Combined three gene analyses revealed that *C. receptaculorum* does not belong in the Annulatascaceae *sensu stricto*, but rather clusters with *C. appendiculata, C. viscosa, Aquaticola hyalomura* (**Fig. 7, Clade 1b**), sister to a clade containing *Aquaticola*-like species (PE0157, PE0264) and *Ayria nubispora* (**Fig. 7, Clade 1c**). *C. appendiculata* is a strongly supported sister taxon to *C. receptaculorum*. These clades are belong to a lineage dividing the Ophiostomatales and the Diaporthales and thus *C. receptaculorum* should be considered Sordariomycetes *incertae sedis*.

Cataractispora viscosa K.D. Hyde, S.W. Wong & E.B.G. Jones, Mycological Research 103(8): 1029. 1999. **Fig. 40 A-D, Fig. 41 A-J.** MycoBank 460661.

Ascomata black, coriaceous, immersed, perpendicular to surface, gregarious, 160–260 µm wide, globose to subglobose (**Fig. 41 A,B**). **Ascomal wall** hyaline and brown, to 36.5 µm thick outer layer of brown angular cells and inner layer of hyaline angular cells (**Fig. 41 B**). **Neck** white, 50 µm long. **Paraphyses** hyaline, 2 µm wide at base, tapering toward apex, septate (**Fig. 41 B**). **Asci** unitunicate, cylindrical, 8–spored, 126–230 µm long × 8–12 µm wide (x̅ = 178 × 10 µm, l:w = 18) (**Fig. 41 C**). **Ascus base** pedicellate, broadly tapering to flaring club-like tip, 25 µm from *pars sporifera* (**Fig. 41 D**). **Annulus** J-, refractive, 5 µm long × 6.5 µm wide (l:w = .77) (**Fig. 41 E-G**). **Ascospores** hyaline, fusiform, 5–septate, not constricted at septa, uniseriate to overlapping uniseriate, 25–34 µm long × 6–6.5 µm wide ($\overline{x}$ = 29.5 × 6.25 µm, l:w = 4.7), large lipid guttule in each cell, with thin unfurling appendages, without sheath (**Fig. 41 H-J**) (Hyde et al. 1999).

Specimens examined:

HOLOTYPE: HONG KONG. New Territories, Tai Po Kau Country Park, on twigs submerged in a stream, Sep 1993, *S.W. Wong & K.D. Hyde, (HKU(M) 3130),* **IFRD 8726.**

PERU. Cusco, Camanti, stream at Quincemil trail 1, 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged herbaceous debris, water 21.2 °C, pH 7.1, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0165-14;* Stream at Quincemil trail 1, 13°13’31.0434”S, 70°45’10.6194”W, 688m, on submerged woody debris, water 19 °C, pH 8, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0165-8;* Stream at Quincemil trail 2, 13°13’31.0434”S, 70°45’10.6194”W, 653m, on submerged woody debris, water 25 °C, pH 7.3, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0165-13;* Stream at Quincemil Trail 2, trailhead 13°13’31.0434”S, 70°45’10.6194”W, 653m, on submerged woody debris, water 21.9 °C, pH 6.4, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0165-2;* Stream at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 21.1 °C, pH 7.4, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0165-3;* Rio Caliente, 1km south of Quincemil, 13°13’20.87”S, 70°44’30.07”W, 626m, on submerged woody debris, water 25 °C, pH 7.2, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0165-12;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 20.5 °C, pH 7.2, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0165-6;* Stream crossing the Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 737m, on submerged woody debris, water 22 °C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0165-16;* River at end of Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.5 °C, pH 7.7, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0165-10;* 1st stream at Quincemil Trail 3, trailhead 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.7 °C, pH 6.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0165-15;* Stream at Quincemil Trail 3, trailhead 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.5 °C, pH 7, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0165-21;* River backwater at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 22 °C, pH 7.1, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0165-23;* Stream flowing into river backwater at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.2 °C, pH 7.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0165-19;* Stream with red algae along Quincemil trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, water 21.8 °C, pH 7.2, on submerged woody debris, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0165-20.* Madre de Dios, CICRA, Rio Amigos, 12°34’0.336”S, 70°4’59.052”W, 218m, on submerged woody debris, water 31.4 °C, pH 8, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0165-9;* CICRA, Stream at Trail 14, 12°34’14.7”S, 70°05’23.69”W, 241m, on submerged woody debris, water 25.1 °C, pH 7.3, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0165-5;* CICRA, Rio Amigos, 12°33’25.22”S, 70°05’59.89”W, 288m, on submerged woody debris, water 31.4 °C, pH 8, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0165-4.*

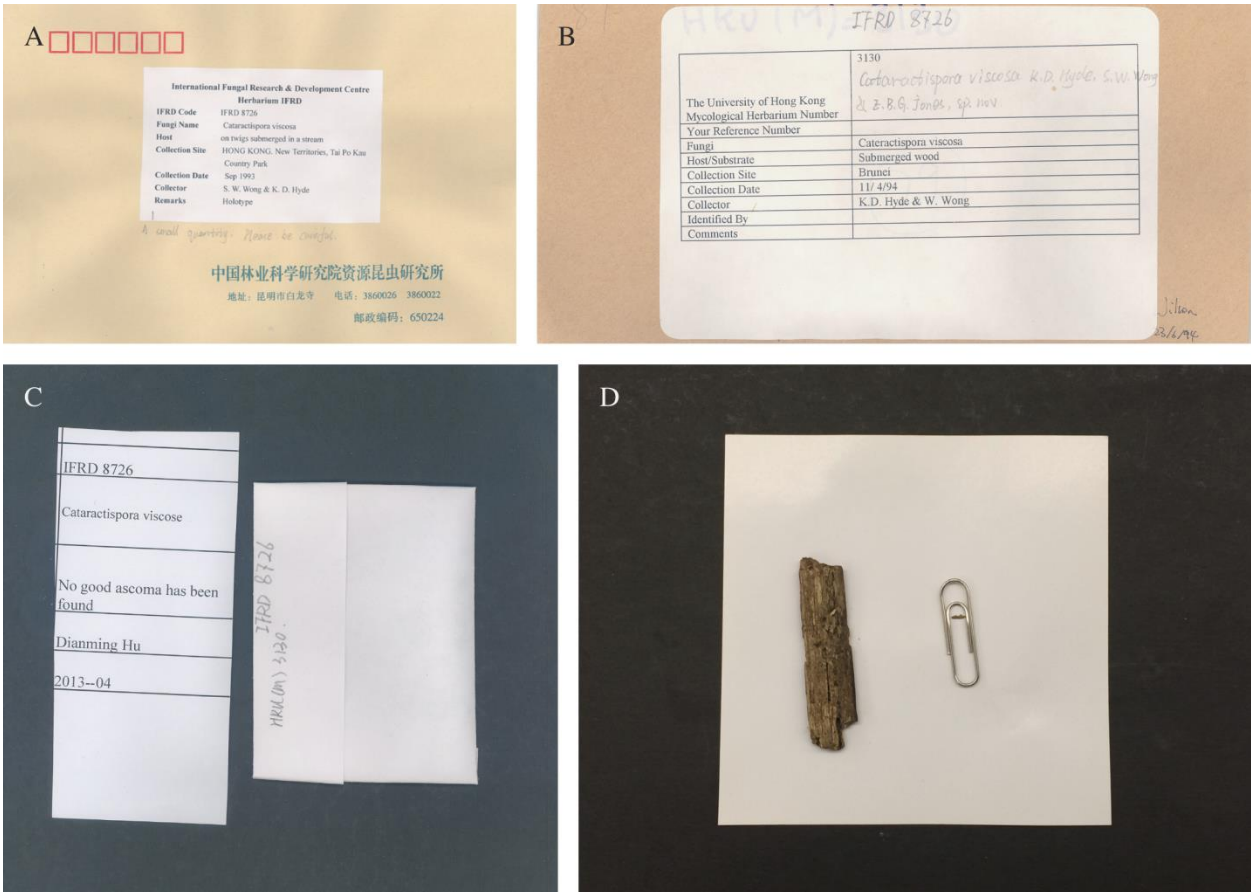
Reports:

AUSTRALIA. North Queensland, Cow Bay, freshwater stream, on submerged wood, Jun 1995, *K.D. Hyde & T.M. Hyde (HKU(M) CB24)*. MAURITIUS. Black River National Park, Black River, Aug 1995, *K D. Hyde & A. Poonyth. (HKU(M) Maur 53)*.

Known distribution: Australia, Hong Kong, Mauritius, Perú.

Comments: The holotype of *Cataractispora viscosa* was in poor condition (**Fig. 40 A-D**) and lacked fungal material as was noted in the accompanying documentation (Dianming Hu) (**Fig. 40 C**). Neotypification from Hong Kong should be done. Fresh specimens of *C. viscosa* were obtained from Perú and sequenced. Peruvian specimens were a good match to the protologue (Hyde et al. 1999). The type of

Fig. 40 A-D. *Cataractispora viscosa* from the holotype **IFRD 8726**. A. IFRD herbarium packet. B. Original HKU herbarium packet (HKU(M) 3130). C. Contents of packet. D. Dried substrate.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8726
Fungi Name	Cataractispora viscosa
Host	on twigs submerged in a stream
Collection Site	HONG KONG. New Territories, Tai Po Kau Country Park
Collection Date	Sep 1993
Collector	S. W. Wong & K. D. Hyde
Remarks	Holotype

A small quantity. Please be careful.

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

HKU (M)
IFRD 8726

The University of Hong Kong Mycological Herbarium Number	3130 Cataractispora viscosa K.D. Hyde, S.W. Wong & E.B.G. Jones, sp. nov
Your Reference Number	
Fungi	Cateractispora viscosa
Host/Substrate	Submerged wood
Collection Site	Brunei
Collection Date	11/ 4/94
Collector	K.D. Hyde & W. Wong
Identified By	
Comments	

Jilson
23/6/94

IFRD 8726

Cataractispora viscose

No good ascoma has been found

Dianming Hu

2013--04

IFRD 8726
HKU(M) 3130.

Fig. 40.

Fig. 41 A-J. *Cataractispora viscosa* from specimen PE0165-8. A. Ascomata on substrate. B. Ascoma in water illustrating hyaline neck. C. Ascus. D. Ascus base. E-F. Ascus apices illustrating large apical rings. H-J. Ascospores. Bars: A = 500 µm; B = 100 µm; C = 40 µm; D-E, H = 20 µm; F-G, I-J = 10 µm.

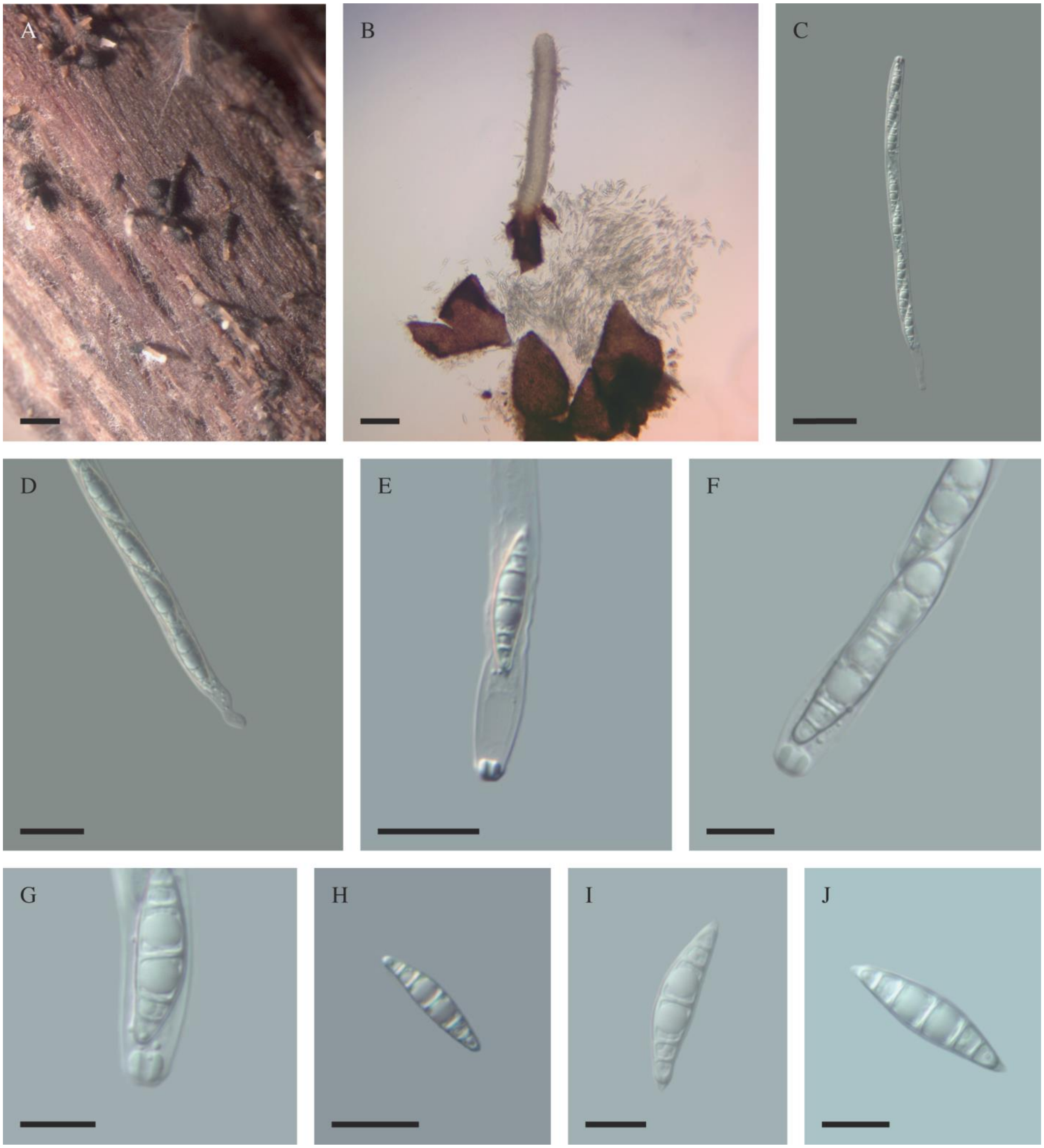


Fig. 41.

the genus, *C. aquatica*, was not evaluated molecularly in this study. *C. aquatica* and *C. viscosa* share ascospores with similar morphology though the ascospores of *C. aquatica* are longer. The neck of *C. viscosa* is also consistently white, as opposed to the black necks of *C. aquatica*. The high degree of morphological similarity places these species as congeneric, but the lack of molecular data for *C. aquatica* makes the placement of *C. viscosa* in the genus a question that still needs to be addressed. *C. viscosa* clusters in a clade with and *C. receptaculorum, C. appendiculata, Aquaticola hyalomura* (**Fig. 7, 1b**), sister to a clade containing *Aquaticola*-like species (PE0157, PE0264) and *Ayria nubispora* (**Fig. 7, 1c**). However, these taxa are not affiliated with Annulatascaceae *sensu stricto*. *Cataractispora viscosa* should be considered Sordariomycetes *incertae sedis*.

Chaetorostrum Zelski, Raja, A.N. Mill. & Shearer, Mycosphere 2(5): 594. 2011. MycoBank 563571.

Type species: *Chaetorostrum quincemilense.*

Chaetorostrum quincemilense Zelski, Raja, A.N. Mill. & Shearer, Mycosphere 2(5): 595. 2011. MycoBank 587628 (syn. *Chaetorostrum quincemilensis* MycoBank 563571, Legitimate)

Ascomata brown to light brown, membranous, immersed to semi-immersed, parallel to surface, scattered, 200–270 µm wide × 800–900 µm high (x̅ = 235 × 850 µm, w:h = .28), elongated globose. **Ascomal wall** of *textura angularis* in surface view. **Neck** brown to hyaline at apex, cylindrical, 600–700 µm long × 64–70 µm wide, with rigid, light brown, 90–150 µm long × 2–4 µm wide, pointed, 8–10 septate setae, periphysate. **Paraphyses** hyaline, 5–7 µm wide at base × 137–162 µm long, tapering toward apex, septate, filamentous. **Asci** unitunicate, cylindrical, 8–spored, 180–240 µm long × 12–15 µm wide (x̅ = 235 × 850 µm, l:w = 15.6). **Ascus base** long, narrow, elongate tail with a spike-like extension, 40 µm from *pars sporifera*. **Annulus** J-, bipartite, cylindrical, 5–6 µm long × 7–8 µm wide (x̅ = 5.5 × 7.5 µm, l:w = .74). **Ascospores** versicolored with brown central cells and hyaline end cells, broadly ellipsoidal, 3–septate, slightly constricted at middle septum, overlapping uniseriate, 30–38 µm long × 10–12 wide µm ($\bar{x}$ = 33 × 11 µm, l:w = 3), with two large lipid guttules, with gelatinous apiculate appendages, adpressed ephemeral sheath. **Colonies** on PYG + Ab grey-brown, dark brown in reverse view, irregular, raised; on CMA + alfalfa light brown to dark brown, floccose, in reverse view, producing phragmospores. **Conidiophores** hyaline, micronematous, mononematous. **Conidia** brown, paler at apex, dry, schizolytic, monoblastic from ends of hyphae, 20–280 µm long × 7-13 µm wide, 2–40 septate, with rough walls that slough off.

Specimens examined:

HOLOTYPE: PERU. Cusco, Camanti, Quincemil, Stream at Quincemil Trail 1, 13°14’23”S, 70°46’13”W, on submerged woody debris, 688m, water 21.2 °C, pH 6.8, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0105-1,* **ILL 40822.**

PERU. Cusco, Camanti, Quincemil, 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S,

70°46’12.6114”W, 688m, water 21.3 °C, pH 6, on submerged woody debris, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0105-2.*

Known distribution: Perú.

Comments: Illustrations can be found in Zelski et al. (2011b). Originally referred to the Annulatascaceae due to its large non-amyloid ascus apical ring, this species is not affiliated with Annulatascaceae *sensu stricto*. The presence of setae on the neck is a feature not found in any Annulatascaceae species. Its placement changes with the genes and taxa sampled in separate and combined analyses, and is on a branch sister to *Fluminicola bipolaris*, *Brunneosporella aquatica,* and *Papulosa amerospora,* all three aquatic Sordariomycetes, in combined three gene analyses (**Fig. 7, Clade 1a**). This species should be considered Sordariomycetes *incertae sedis* at present as it is in a lineage separating the Diaporthales and Ophiostomatales.

Clohiesia K.D. Hyde, Nova Hedwigia 61 (1-2): 125. 1995. MycoBank 27574.

Type species: *Clohiesia corticola.*

Clohiesia corticola K.D. Hyde, Nova Hedwigia 61: 126. 1995. **Fig. 42 A-K.** MycoBank 412816.

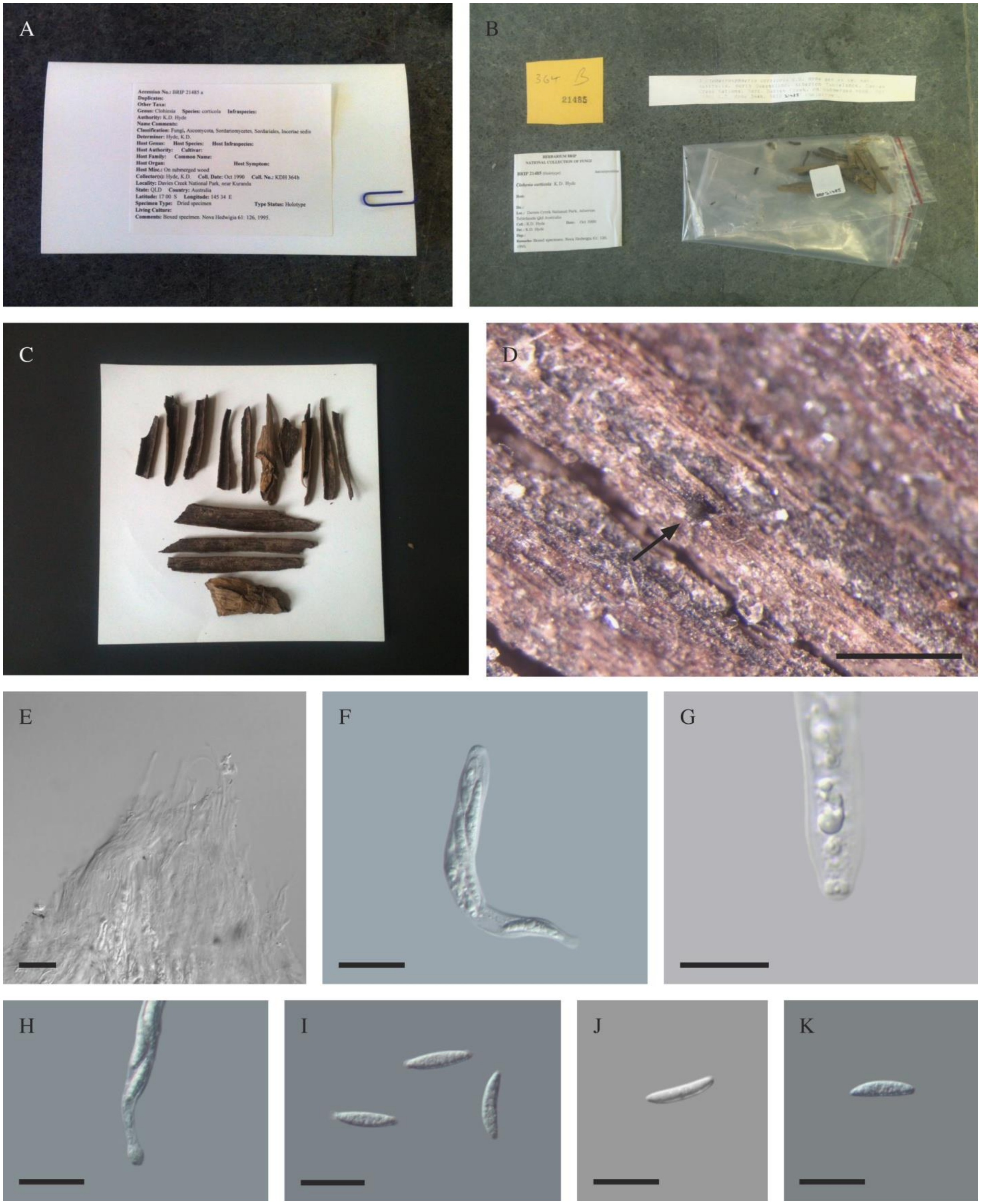
Ascomata light brown, coriaceous, immersed, 200 µm wide, clypeate (clypeus to 300 µm diam.), perpendicular to surface, solitary, depressed-globose, conical (**Fig. 42 D**). **Ascomal wall** light brown, 12–20 µm thick, 3–4 layers of light brown oblong cells. **Neck** dark brown 200 µm long × 120 µm wide, pseudoparenchymatous, of *textura angularis.* **Paraphyses** hyaline, tapering, septate, filamentous, branched (**Fig. 42 E**). **Asci** unitunicate, cylindro-clavate, 8–spored, 125 µm long × 75 µm wide (l:w= 16.7) (**Fig. 42 F**). **Ascus base** pedicellate (**Fig. 42 H**). **Annulus** J-, refractive (**Fig. 42 G**). **Ascospores** hyaline, curved fusiform, aseptate, overlapping uniseriate to biseriate, 18.5–26.5 µm long × 3.7–5 µm wide ($\bar{x}$ = 22.5 × 4.35 µm, l:w = 5.2) guttulate, without appendages, with a thin spreading sheath (**Fig. 42 I-K**) (Hyde 1995).

Specimens examined:

HOLOTYPE: AUSTRALIA. North Queensland, Atherton Tablelands, Davies Creek National Park, near Kuranda, Davies Creek, 17°0’0”S, 145°20’24”E, on submerged wood, Oct 1990, *K.D. Hyde, KDH 364b,* **BRIP 21485.**

Known distribution: Australia.

Fig. 42 A-K. *Clohiesia corticola* from the holotype **BRIP 21485**. A. Herbarium packet. B. Contents of packet. C. Dried wood. D. Ostiole protruding through surface of substrate (arrow). E. Paraphyses. F. Ascus. G. Annulus. H. Ascus base. I-K. Ascospores. Bars: D = 500 µm; E-F, H-K = 20 µm; G = 10 µm.



Accession No.: BRIP 21485 a
Duplicates:
Other Taxa:
Genus: Clohiesia **Species:** corticola **Infraspecies:**
Authority: K.D. Hyde
Name Comments:
Classification: Fungi, Ascomycota, Sordariomycetes, Sordariales, Incertae sedis
Determiner: Hyde, K.D.
Host Genus: **Host Species:** **Host Infraspecies:**
Host Authority: **Cultivar:**
Host Family: **Common Name:**
Host Organ: **Host Symptom:**
Host Misc.: On submerged wood
Collector(s): Hyde, K.D. **Coll. Date:** Oct 1990 **Coll. No.:** KDH 364b
Locality: Davies Creek National Park, near Kuranda
State: QLD **Country:** Australia
Latitude: 17 00 S **Longitude:** 145 34 E
Specimen Type: Dried specimen **Type Status:** Holotype
Living Culture:
Comments: Boxed specimen. Nova Hedwigia 61: 126, 1995.

Fig. 42.

Comments: The holotype of *Clohiesia corticola* was in good condition (**Fig. 42 A-C**). According to the documentation accompanying the herbarium packet, Hyde originally intended to name this fungus *Clohesyosphaeria corticola* (**Fig. 42 B**). The herbarium specimen consists of dried pieces of wood with embedded preserved ascomata (**Fig. 42 C**). No fresh collections of this species were made in this study. A morphological feature that separates this species, and the genus, from Annulatascaceae *sensu stricto* is the formation of ascomata under a clypeus. In addition, the asci are cylindro-clavate compared to other members of Annulatascaceae *sensu stricto*, save for *Pseudoproboscispora caudae-suis* and the dubious *Paoayensis lignicola*. The 28S sequence used in this study and previous studies (Ranghoo et al. 1999, Raja et al. 2003, Abdel-Wahab et al. 2011) show that *Cl. corticola* is removed from Annulatascaceae *sensu stricto* based on molecular analyses. In combined three gene analyses in this study, *Cl. corticola* is placed in a clade with Boliniales, Chaetosphaeriales, and Sordariales and should be considered Sordariomycetes *incertae sedis* (**Fig. 7, Clade 3c**).

Clohiesia curvispora L. Cai & K.D. Hyde, Mycoscience 48: 182. 2007. MycoBank 529577.

Ascomata dark brown to black, carbonaceous, semi-immersed to superficial, clypeate, perpendicular, gregarious, 400–600 µm wide × 400–500 µm high ($\bar{x}$ = 500 × 450 µm, w:h = 1.1), globose to subglobose. **Ascomal wall** light brown, brown-black, 30–50 µm thick, composed of light brown oblong cells, of *textura angularis* in surface view. **Neck** dark brown, short, pseudoparenchymatous, of *textura angularis*, periphysate. **Paraphyses** hyaline, 3–4 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 125–175 µm long × 8.5–10.5 µm wide ($\bar{x}$ = 150 × 9.5 µm, l:w = 15.8). **Ascus base** pedicellate, broadly tapering, 30 µm from *pars sporifera*. **Annulus** J-, refractive, 1.5 µm long × 2.5–3 µm wide ($\bar{x}$ = 1.5 × 2.75 µm, l:w = .55). **Ascospores** hyaline, curved elongate fusiform, aseptate, overlapping uniseriate to biseriate, 32.5–39 µm long × 3.2–5 µm wide ($\bar{x}$ = 35.8 × 4.1 µm, l:w = 8.7), guttulate, without appendages, with a thin sheath (Cai and Hyde 2007).

HOLOTYPE: CHINA. Yunnan, Jinghong, on submerged wood in a small forest stream, 15 Sep 2002, *L. Cai, CAI-9BNA38,* **HKU(M) 10854.**

Known distribution: China.

Comments: The holotype of *Clohiesia curvispora* was unavailable for examination and fresh specimens were not obtained in this study. The genus *Clohiesia* was initially referred to Annulatascaceae primarily due to its large ascus apical rings, but is shown to be a separate lineage based on the phylogenetic placement of *Cl. corticola* in this and previous studies (Ranghoo et al. 1999, Raja et al. 2003, Abdel-Wahab et al. 2011). The presence of a clypeus in *Clohiesia* appears to be a strong morphological feature precluding inclusion in Annulatascaceae despite the presence of large apical rings. Fresh material from China would shed light on the phylogenetic placement of this species with respect to the genus. *Cl. curvispora* should be considered Sordariomycetes *incertae sedis* as the type of the genus, *Cl. corticola* is placed there in this study (**Fig. 7, Clade 3c**).

Clohiesia lignicola K.M. Tsui, K.D. Hyde, & I. J. Hodgkiss, Mycoscience 39: 257. 1998. **Fig. 43 A-L.** MycoBank 446571.

Ascomata black, coriaceous, semi-immersed to superficial, clypeate, perpendicular, solitary to gregarious, 440–500 µm long × 400–440 µm high (x̅ = 470 × 420 µm, w:h = 1.2), globose to subglobose (**Fig. 43 D**). **Ascomal wall** hyaline and brown, 20–30 µm thick, outer layer of dark brown cells, inner layer of hyaline elongate cells, cells flattened at base, of *textura angularis* in surface view (**Fig. 43 E**). **Neck** black, 20–30 µm wide, periphysate. **Paraphyses** hyaline, 5–8 µm wide at base × 200 µm long, tapering toward apex, septate, numerous (**Fig. 43 I,J)**. **Asci** unitunicate, cylindrical to cylindro-clavate, 8–spored, 120–140 µm long × 8–12 µm wide (x̅ = 130 × 10 µm, l:w = 13) (**Fig. 43 F**). **Ascus base** pedicellate, cylindrical, rounded, 30 µm from *pars sporifera*. **Annulus** J-, relatively massive, 2 µm long ×

3–4 µm wide (x̅ = 2 × 3.5 µm, l:w = .57) (**Fig. 43 G,H**). **Ascospores** hyaline, fusoid ellipsoidal, aseptate, overlapping uniseriate, 14–28 µm long × 4.5–6 µm wide (x̅ = 21 × 4.1 µm, l:w = 8.7), guttulate, without appendages, with a thin sheath (**Fig. 43 K,L**) (Tsui et al. 1998).

Specimens examined:

HOLOTYPE: HONG KONG. Lantau Island, Tung Chung River, on submerged wood, 28 Jul 1997, *K.M. Tsui & Ken Wong, KM202, (HKU(M) 5539),* **IFRD 8732.**

Known distribution: Hong Kong.

Comments: The holotype of *Clohiesia lignicola* was in good condition with many ascomata present on the dried substrate (**Fig. 43 A-C**). Two permanent slides were made and returned with the holotype material. A photographic plate is included in the herbarium packet, which differs from the plate included in the protologue (Dianming Hu) (**Fig. 43 C**). Asci were difficult to tease apart. They were fused together along with copious long septate tapering paraphyses. It has been previously shown that *Cl. corticola*, the type of the genus, does not belong to Annulatascaceae based on 28S sequence data (Ranghoo et al. 1999, Raja et al. 2003, Abdel-Wahab et al. 2011) and thus the genus itself has been excluded. These results are supported in this study as well. Fresh material of *Cl. lignicola* was not collected during the course of this study, and thus this species and the genus remain to be assessed with further molecular data. Morphologically, *Clohiesia* differs from other taxa with J- ascus apical rings in that it is clypeate and possesses fusoid-ellipsoidal ascospores, characters which are not present in other members of Annulatascaceae *sensu stricto*. *Cl. lignicola* should be considered Sordariomycetes *incertae sedis* as the type of the genus, *Cl. corticola* is placed there in this study (**Fig. 7, Clade 3c**).

Fig. 43 A-L. *Clohiesia lignicola* from the holotype **IFRD 8732**. A. Herbarium packet. B. Dried substrate. C. Photographic plate included with dried specimen (Dianming Hu). D. Semi-immersed ascomata on substrate. E. Peridium. F. Ascus. E-H. Asus apices. I. Long tapering septate paraphyses. J. Base of paraphyses. K. Ascospores. L. Ascospore. Bars: D = 500 µm; E-F, I-L = 20 µm; G-H = 10 µm.

International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code IFRD 8732
Fungi Name Clohesia lignicola dark green
Host on Submerged wood
Collection Site HONG KONG, Lantau Island, Tung Chung Riv
Collection Date Aug 1997
Collector Clement & Ken Wong
Remarks Holotype, KM202

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺 电话：3860026 3860022
邮政编码：650224

Fig. 43.

Cyanoannulus Raja, J. Campbell & Shearer, Mycotaxon 88: 8. 2003. MycoBank 28772.

Type species: *Cyanoannulus petersenii.*

Cyanoannulus petersenii Raja, J. Campb., and Shearer, Mycotaxon 88: 11. 2003. **Fig. 44 A-O, Fig. 45 A-I.** MycoBank 469353.

Ascomata pale reddish brown, immersed to semi-immersed, slightly horizontal to surface, scattered, 405–800 µm wide × 214–297 µm high ($\bar{x}$ = 602.5 × 255.5 µm, w:h = 2.4), globose to subglobose (**Fig. 44 F, Fig. 45 A**). **Ascomal** wall hyaline and dark, outer 5–6 layers of dark cells, inner 2–3 layers of hyaline elongated pseudoparenchymatous cells, of *texture angularis* in surface view (**Fig. 44 G, Fig. 45 B**). **Neck** pale reddish brown, lighter at apex, 340–760 µm long × 25–65 µm wide, periphysate (**Fig. 44 H**). **Paraphyses** hyaline, 9–12 µm wide at base × 55–110 µm long, tapering toward apex, septate, constricted at septa (**Fig. 44 I**). **Asci** unitunicate, fusoid, 8–spored, 73–142 µm long × 8–12 µm wide ($\bar{x}$ = 107.5 × 10 µm, l:w = 10.75) (**Fig. 44 J,K; Fig. 45 C,D**). **Ascus base** pedicellate, short, club-like, 5 µm from *pars sporifera*. **Annulus** J-, bipartite, 2–3 µm long × 4–5 µm wide ($\bar{x}$ = 2.5 × 4.5 µm, l:w = .56) (**Fig. 44 L, Fig. 45 E**). **Ascospores** hyaline, fusiform, 3–septate, constricted at septa, overlapping uniseriate, 20–26.5 µm long × 5–7 µm wide ($\bar{x}$ = 23.25 × 6 µm, l:w = 3.9), guttulate, without appendages, with a 3–5 µm wide sheath (**Fig. 44 M-O, Fig. 45 F-I**) (Raja et al. 2003).

Specimens examined:

HOLOTYPE: USA. North Carolina, GSMNP, Ocanaluftee River, on submerged decorticated wood, 35°34’12.06”N, 83°20’7.38”W, on submerged decorticated wood, water 20 °C, pH 5, 3 Jul 2005, *H.A. Raja and Nate Hamburger, R44-1,* **ILL 40101.**

Pennsylvania, Ricketts Glen State Forest, river, 41°20’1.356”N, 76°16’15.4194”W, 632m, on submerged woody debris, water 20.3 °C, pH 9, 632 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z60-1824.*

Known distribution: Costa Rica, USA.

Comments: The holotype of *Cyanoannulus petersenii* consists of 5 slides: 3 ascomata fixed for sectioning as well as three pieces of dried wood (**Fig. 44 A-E)**. Raja et al. (2003) described *Cy. petersenii* at nearly the same time as Tsui et al. (2003) described *Aquaticola triseptata*. These species would be nearly indistinguishable save for the staining of the annulus of *Cy. petersenii* in cotton blue. Examination of the holotype of *Aq. triseptata* and Peruvian specimens did not reveal staining of the annuli in cotton blue, nor did preserved slides show this feature. Raja et al. (2003) used 28S molecular evidence to show affinity of *Cy. petersenii* with Annulatascaceae, though taxon sampling was limited. Raja et al. (2003) also noted that additional specimens of *Cy. petersenii* were examined from Costa Rica, though molecular data for these specimens was not obtained. Tsui et al. (2003) based their placement of *Aq. triseptata* solely on morphology. In this study, Peruvian specimens of *Aq. triseptata* were initially diagnosed as *Cy. petersenii.* 28S analysis placed the two taxa as sister taxa with low support (**Fig. 2**). In combined three gene analyses, these taxa are not related (**Fig. 7; Clades 1h, 3c**). Fresh specimens of both taxa were used in this study, *Aq. triseptata* from the tropics and *Cy. petersenii* from temperate zones. It would be worthwhile to obtain fresh material from Costa Rica to determine whether there is a temperate vs. tropical distribution of these highly similar species. *Cy. petersenii* is in a lineage separating Diaporthales and Ophiostomatales in this study and should be considered Sordariomycetes *incertae sedis*.

Fig. 44 A-O. *Cyanoannulus petersenii* from the holotype **ILL 40101**. A. Herbarium folder. B. Herbarium sheet. C. Contents of herbarium packets. D. Dried wood. E. Herbarium label. F. Neck protruding from substrate. G. Midsaggital section. H. Neck. I. Paraphysis. J-K. Asci. L. Ascus apex. M-O. Ascospores. Bars: F = 500 µm; G-H = 40 µm; I-N = 20 µm; O = 10 µm.

Department of Plant Biology Herbarium
University of Illinois at Urbana-Champaign
Freshwater Ascomycetes Collection

Cyanoannulus petersenii Raja, J. Campb. & Shearer
Class: Sordariomycetes Order: Inc. sed. Family:Annulatascaceae
Site USA: North Carolina. Great Smoky Mountains National Park, Ocanaluftee River, 35°34.201'N, 83°20.123'W, UTM Zone mE, 3936678 mN, water temperature 20° C, pH 5

Date: 3 July 2005 Specimen No.: R44-1 ILL 40101
Substrate: on submerged, decorticated wood

Col.: Huzefa Raja & Nate Hamburger
Det.: Huzefa Raja **Holotype**

Fig. 44.

Fig. 45 A-I. *Cyanoannulus petersenii* from specimen Z60-1824. A. Necks arising from submerged ascomata. B. Peridium. C-D. Asci. E. Annulus. F-H. Ascospores. Bars: A = 500 µm; B-D, F = 20 µm; E, G-I = 10 µm.

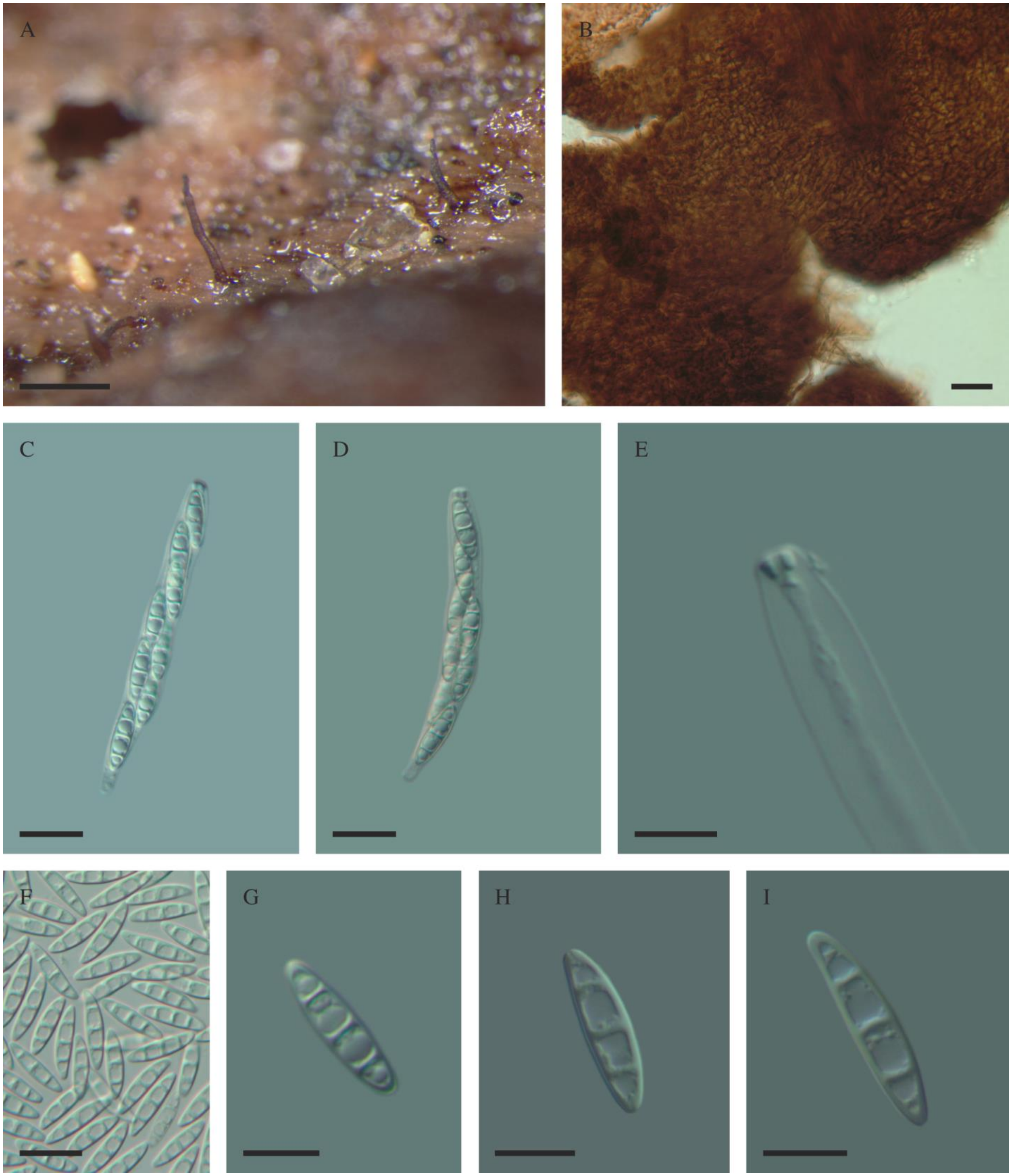


Fig. 45.

Diluviicola K.D. Hyde, S.W. Wong & E.B.G. Jones, Fungal Diversity 1: 134. 1998. MycoBank 24138.

Type species: *Diluviicola capensis.*

Diluviicola capensis K.D. Hyde, S.W. Wong & E.B.G. Jones, Fungal Diversity 1:134. 1998. **Fig. 46 A-B.** MycoBank 444849.

Ascomata black, coriaceous, immersed to semi-immersed, parallel to surface, solitary, 93–128 µm wide × 32–80 µm high ($\bar{x}$ = 110.5 × 56 µm, w:h = 2), obpyriform. **Ascomal wall** brown, several layers of brown elongate cells. **Neck** hyaline, short, periphysate. **Paraphyses** hyaline, 4 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, long cylindrical, 8–spored, 266–326 µm × 11–14 µm ($\bar{x}$ = 296 × 12.5 µm, l:w = 23.7). **Ascus base** pedicellate, tapering, 50 µm from *pars sporifera*. **Annulus** J-, bipartite, 3–4 µm long × 2–3 µm wide ($\bar{x}$ = 3.5 × 2.5 µm, l:w = 1.4). **Ascospores** hyaline, fusiform, aseptate, uniseriate to overlapping uniseriate, 26–34 µm long × 6–11 µm wide ($\bar{x}$ = 30 × 8.5 µm, l:w = 3.9), multiguttulate, with bipolar unraveling appendages, without sheath (Hyde et al. 1998).

Specimens examined:

HOLOTYPE: BRUNEI DARUSSALAM. Temburong, Kuala Belalong Field Studies Centre, Sungai Esu, on submerged wood, Feb 1994, *K.D. Hyde & S.W. Wong*, **IFRD 9033.**

Known distribution: Brunei**.**

Comments: The dried holotype substrate of *Diluviicola capensis* was extremely scant and lacked fungal tissue of *D. capensis* (**Fig 46 A,B**). Fresh collections of this fungus were not obtained in this study. The protologue should serve for comparison to freshly collected specimens until a neotype is designated. If the paratype (HKU(M) BRUNEI 29) cited in the protologue, is available, it could be designated as the neotype. Of note is a name change since the protologue (Wong et al. 1999) was published. *Diluviocola*

Fig. 46 A-B. *Diluviicola capensis* from the holotype **IFRD 9033**. A. Herbarium packet. B. Dried material.

International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 9033
Fungi Name	Diluviocola capensis
Host	on Submerged wood
Collection Site	Brunei
Collection Date	1994-4-11
Collector	K.D. Hyde and S.W. Wong
Remarks	Holotype

A small quantity. Please be careful.

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

Fig. 46.

was at some point changed to the proper Latin name recognized in Index Fungorum and MycoBank, *Diluviicola*. Fresh collections would also allow sequencing of DNA for phylogenetic placement of *D. capensis* with relation to Annulatascaceae *sensu stricto*, though morphologically its ascospore morphology and ascospore appendages mark it as a distinct genus compared to the rest of the genera in the family.

Fluminicola S.W. Wong, K.D. Hyde & E.B.G. Jones, Fungal Diversity 2: 190. 1999. MycoBank 28031.

Type species: *Fluminicola bipolaris.*

Fluminicola bipolaris S.W. Wong, K.D. Hyde & E.B.G. Jones, Fungal Diversity 2: 190. 1999. **Fig. 47 A-D, Fig. 48 A-E.** MycoBank 450146.

= *Fluminicola coronata* S.W. Wong, K.D. Hyde & E.B.G. Jones, Wong Ph.D. thesis, 141. 1996.

Ascomata black, coriaceous, immersed to semi-immersed, perpendicular to surface, solitary, 75–177 µm wide × 39–80 µm high (x̅ = 126 × 60 µm, w:h = 2.1), globose, subglobose, or ellipsoidal. **Ascomal wall** brown, composed of several layers of brown compressed cells. **Neck** black, short, periphysate. **Paraphyses** hyaline, 11 µm wide at base to 5 µm wide at apex, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 107–192 µm long × 9–12 µm wide (x̅ = 150 × 10.5 µm, l:w = 14.3) (**Fig. 47 C, Fig. 48 A**). **Ascus base** pedicellate, long, tapered to a point, 30 µm from *pars sporifera*. **Annulus** J-, bipartite, 3–6 µm long × 1.5–3 µm wide (x̅ = 4.5 × 2.25 µm, l:w = 2) (**Fig. 47 D, Fig. 48 B**). **Ascospores** hyaline, fusiform, 1–3 septate, slightly constricted at septa, uniseriate to overlapping uniseriate, 15–21 µm long × 6–9 µm wide (x̅ = 18 × 7.5 µm, l:w = 2.4), with a large guttule in each cell, with bipolar cup-like appendages, without sheath (**Fig. 48 C-E**) (Wong et al. 1999).

Specimens examined:

HOLOTYPE: PHILIPPINES. Mindanao, Bukidnon, Impalutao, Natigbasan Creek, on submerged wood, Jan 1994, *K.D. Hyde, (HKU(M)3103),* **IFRD 9034.**

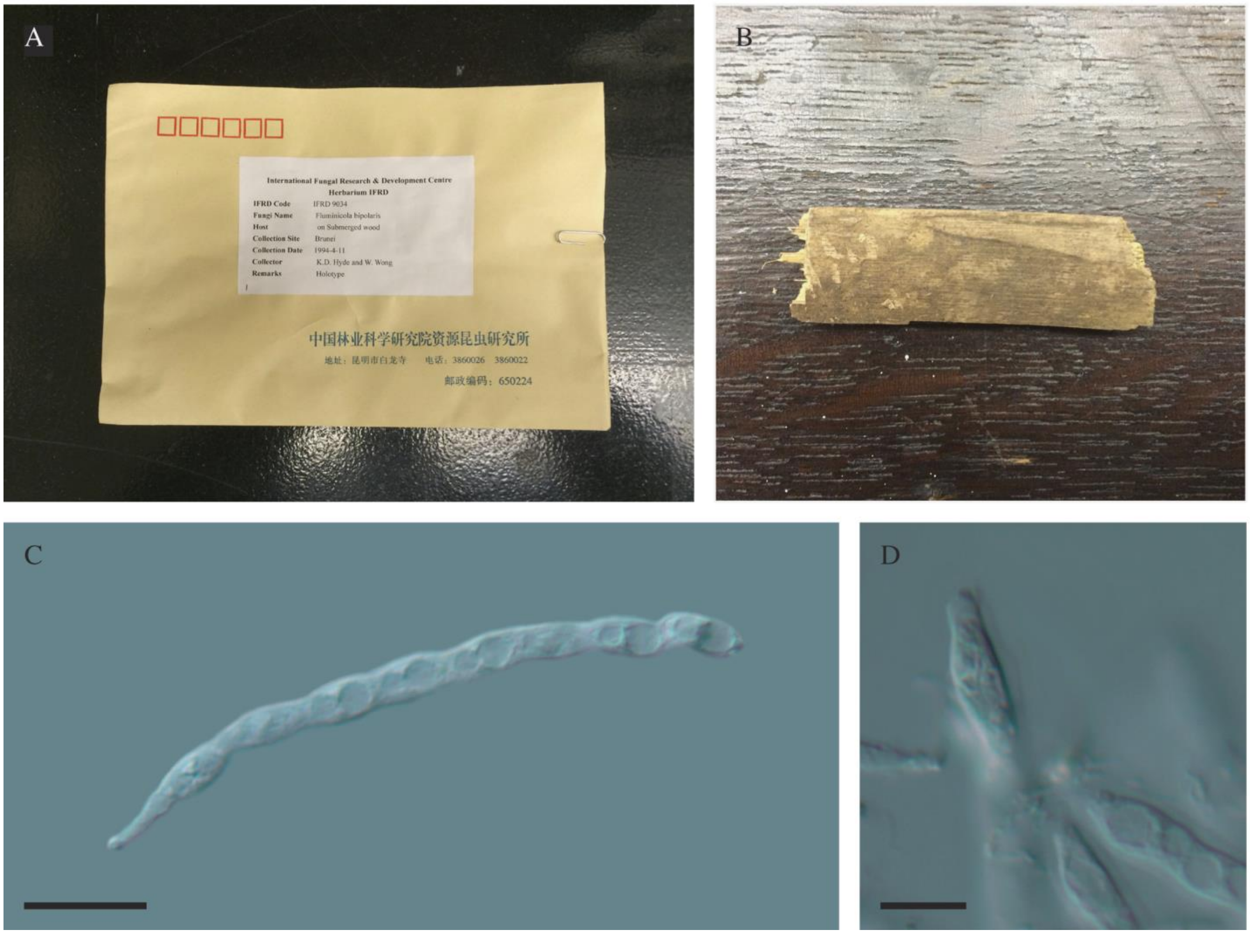
PERU. Cusco, Camanti, stream at Quincemil Trail 3, 13°18'27.756"S, 70°48'44.9274"W, 757m, on submerged woody debris, water 20.7 °C, pH 6, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0020-2;* Madre de Dios, Rio Amigos, 12°34'3.2514"S, 70°4'55.92W, 218m, water 25.3 °C, pH 7.9, 23 May 2010, *Zelski S.E and H.A. Raja, PE0020-1;* CICRA, stream at Trail 14, 12°34'14.7"S, 70°05'23.69"W, 241m, water 25.1 °C, pH 7.3, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0020-3.*

Known distribution: China, Perú, Philippines, Thailand.

Comments: The holotype of *Fluminicola bipolaris* is in fair condition and two permanent slides were made, examined and returned to IFRD (**Fig. 47 A,B)**. The genus *Fluminicola* was introduced to accommodate *F. bipolaris* (Wong et al. 1999). This species is synonymously listed in Index Fungorum and MycoBank as *F. coronata*, and sequences deposited in GenBank are listed under *Flumicola coronata* (note spelling difference). The holotype herbarium number for *F. coronata* (Wong et al. 1996) is identical to that of *F. bipolaris* (Wong et al. 1999). Before the formal description of *F. bipolaris* was published, *Fluminicola* was placed in Annulatascaceae largely based on its large apical ring, (Ho and Hyde 2000), though not in *Annulatascus* due to the unique ontogeny of its bipolar ascospore appendages.

Fresh material of *F. bipolaris* was collected in Perú in this study and DNA was successfully extracted. Peruvian specimens agree well with the holotype, collected in the Philippines (Wong et al. 1999). This species has also been reported from a heavily polluted lake in Yunnan, China (Luo et al. 2004) and from freshwater habitats in Narathiwat and Chiang Rai, Thailand (Pinruan et al. 2007, Kurniawati et al. 2010). Morphologically, the neotropical specimens examined varied little from the type,

Fig. 47 A-D. *Fluminicola bipolaris* from the holotype **IFRD 9034**. A. Herbarium packet. B. Dried material. C. Ascus. D. Ascus apices. Bars: A = 20 µm; B = 10 µm.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 9034
Fungi Name	Fluminicola bipolaris
Host	on Submerged wood
Collection Site	Brunei
Collection Date	1994-4-11
Collector	K.D. Hyde and W. Wong
Remarks	Holotype

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

Fig. 47.

Fig. 48 A-E. *Fluminicola bipolaris* from specimen PE0020-2. A. Ascus. B. Ascus apex illustrating annulus. C-E. Ascospores. Bars: A, C-E = 20 µm; B = 10 µm.

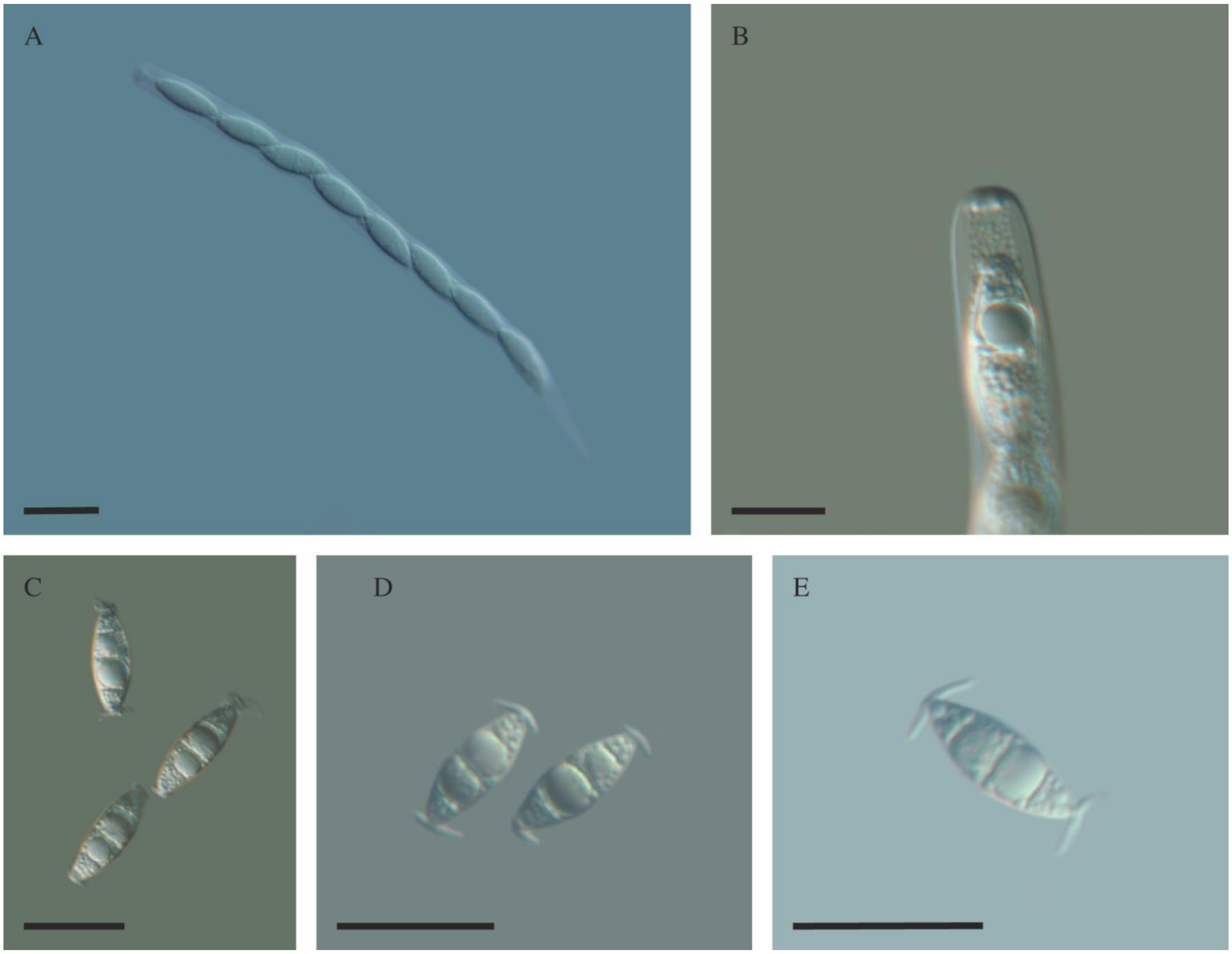


Fig. 48.

and expanded records of ascus length and width from 107–192 × 9–12 µm to 107–205.6 × 8.3–12 µm. Ascospore measurements fell within the type ranges and ascospore appendages were distinctly bifurcate-cupulate. In three gene molecular analyses, Peruvian specimens of *F. bipolaris* belong to a well supported clade with sequences taken from GenBank (**Fig. 7, Clade 1a**). The genus *Fluminicola* is not related to Annulatascaceae *sensu stricto*, and as it resides in a clade between Diaporthales and Ophiostomatales, it should be considered Sordariomycetes *incertae sedis*.

Frondicola K.D. Hyde, Botanical Journal of the Linnean Society 110: 100. 1992. MycoBank 26302.

Type species: *Frondicola tunitricuspis.*

Frondicola tunitricuspis K.D. Hyde, Botanical Journal of the Linnean Society 110: 102. 1992. **Fig. 49 A-P.** MycoBank 357594.

Ascomata light brown, immersed, perpendicular to surface, solitary, 340–620 µm wide × 155–280 µm high (x̅ = 480 × 218 µm, w:h = 2.2), lenticular (**Fig. 49 F,G**). **Ascomal wall** brown, 8.5–25.5 µm thick, elongate cells at sides, thick walls of *textura epidermoidea* at base, surrounded by brown hyphae of *textura porrecta*, of *textura epidermoidea* in surface view. **Neck** short, periphysate. **Paraphyses** hyaline, 6 µm wide at base, tapering toward apex, septate (**Fig. 49 H**). **Asci** unitunicate, cylindrical, 8–spored, 144–181 µm long × 144–181 µm wide (x̅ = 162.5 × 9.2 µm, l:w = 17.7) (**Fig. 49 H,I**). **Ascus base** short, pedunculate, with club-like foot, 15 µm from *pars sporifera*. **Annulus** J-, small (**Fig. 49 J**). **Ascospores** hyaline, ellipsoidal, aseptate (to 3 pseudoseptate), not constricted at septa, 17–26 µm long × 4.8–7.1 µm wide (x̅ = 21.5 × 6 µm, l:w = 3.6), verruculose, with large guttules, without appendages, with a sheath 12–13 × 9–12 µm, with 3 extensions (**Fig. 49 K-P**) (Hyde 1992).

Fig. 49 A-P. *Frondicola tunitricuspis* from the holotype **BRIP 17110**. A. Herbarium packet B. Original documentation. C. Packet contents. D. Slides. E. Dried specimen. F. Surface view of substrate immersed ascomata as raised areas. G. Single immersed ascoma illustrating the ostiole and circular area of light tan crust. H. Asci and paraphyses. I. Ascus. J. Ascus apex illustrating annulus. K. Surface of ascospores illustrating verruculose texture. L-O. Ascospores in aqueous nigrosin illustrating sheaths. P. Ascospore in water. Bars: F-G = 500 µm; H-I,L-P = 20 µm; J-K = 10 µm.

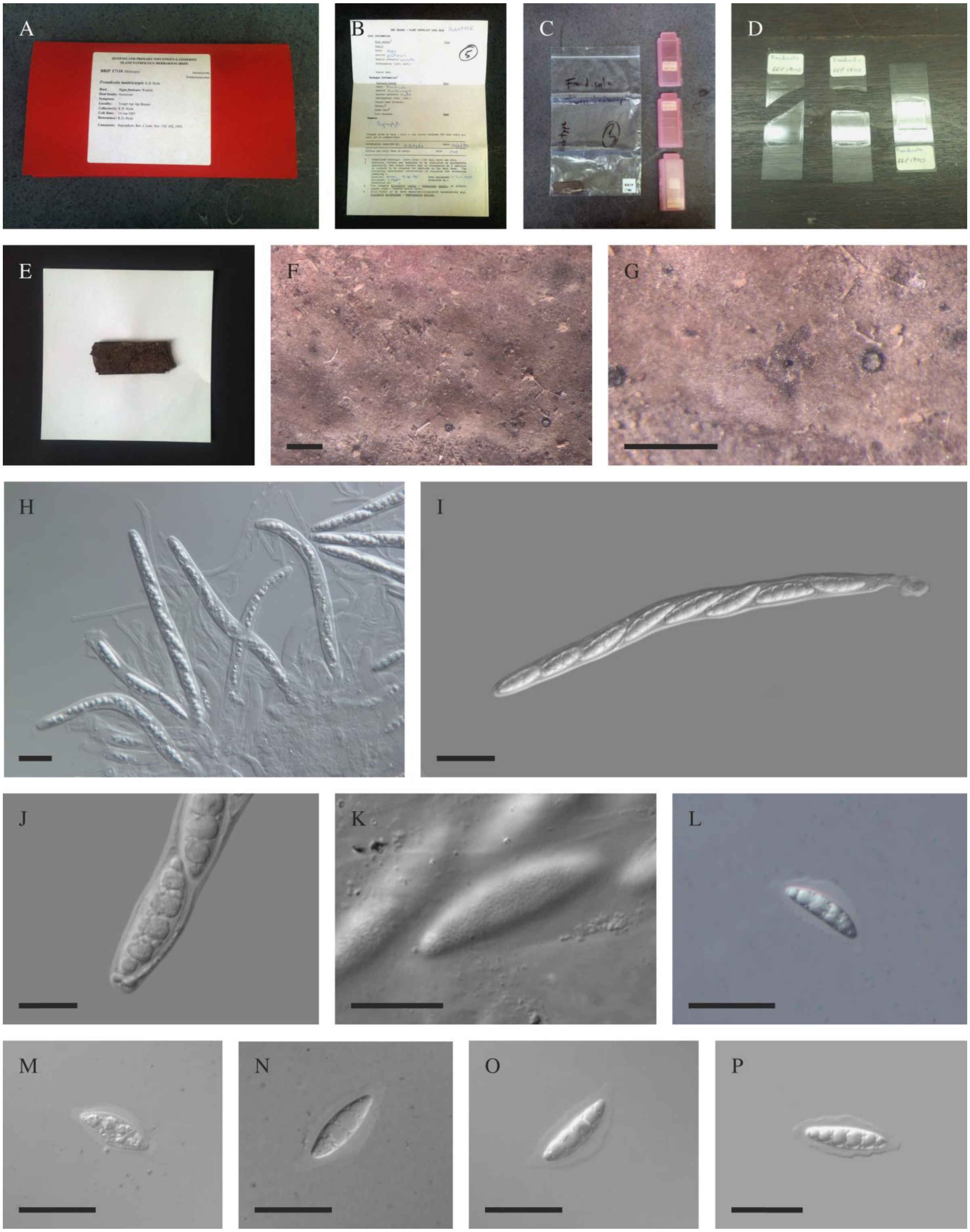


Fig. 49.

Specimens examined:

HOLOTYPE: BRUNEI. South China Sea, Tungit Api Api mangrove, on decaying fronds of *Nypa fruticans* collected from the intertidal region, 14 Jun 1987, *K.D. Hyde,* **BRIP 17110.**

Known distribution: Brunei.

Comments: The holotype of *Frondicola tunitricuspis* consists of three slides, two broken, with one broken one composed of two mismatched pieces. In addition, a dried piece of *Nypa fruticans* was included, 5 × 2 cm, and was replete with ascomata. Small ostioles surrounded by light tan circles betray the location of the ascomata immersed beneath clypeus-like structures. The use of 5% KOH to rehydrate fungal tissue renders the apical rings and ascospore sheaths unobservable. Water, however, preserves these features. Ascospore walls are verruculose as in *Annulatascus velatisporus* when viewed at 100 × magnification, a feature not noted in the type description. Although this feature is found in *A. velatisporus*, the type species of Annulatascaceae, the saltwater habitat and ascus morphology are in contrast to the known habitat and ascus morphology of other members of Annulatascaceae *sensu stricto*. A brackish to salt water species, *Fr. tunitricuspis* was initially included in Annulatascaceae due to its J- ascus apical ring, though it is relatively small (Wong et al. 1998). *Frondicola* should be evaluated molecularly from fresh material to determine its phylogenetic placement, and should be considered Sordariomycetes *incertae sedis*.

Fusoidispora Vijaykr., Jeewon & K.D. Hyde, Sydowia 57(2): 272. 2005. MycoBank 28975.

Type species: *Fusoidispora aquatica.*

Fusoidispora aquatica D. Vijaykrishna, R. Jeewon & K.D. Hyde, Sydowia 57(2): 274. 2004. MycoBank 357159.

Ascomata dark brown, coriaceous, immersed to erumpent, parallel to surface, solitary to gregarious, 115–215 µm wide × 80–150 µm high (x̅ = 165 × 115 µm, w:h = 1.4), ampulliform. **Ascomal wall** hyaline and brown, outer layer of three rows of compressed brown cells, inner layer of hyaline cells, of *textura angularis* in surface view. **Neck** dark brown, 75 µm long × 45 µm wide, periphysate. **Paraphyses** hyaline, 4.5 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 150–178 µm long × 8.5–11 µm wide (x̅ = 164 × 9.8 µm, l:w = 16.8). **Ascus base** pedicellate with club-like end, 15 µm from *pars sporifera*. **Annulus** J-, discoid, 2 µm wide. **Ascospores** hyaline, fusoid to sickle shaped, 0–5 septate, not constricted at septa, overlapping uniseriate to biseriate, 42–50 µm long × 4–6 µm wide (x̅ = 46 × 5 µm, l:w = 9.2), guttulate, with apical pad-like appendages, without sheath (Vijaykrishna et al. 2005).

HOLOTYPE: CHINA. Hong Kong SAR, New Territories, Tai Po Kau Country Park, Tai Po Kau Forest Stream, on submerged wood, 26 Nov 2003, *D. Vijaykrishna, PDD 78746 (isotype)*, **HKU(M) 17484.**

Known Distribution: China.

Comments: The holotype of *Fusoidispora aquatica* was unavailable for examination and fresh specimens were not obtained in this study. Although a key word of the protologue, early analysis of 28S sequence data showed that *Fu. aquatica* was not affiliated with Annulatascaceae, but rather was placed basal to the Ophiostomatales (Vijaykrishna et al. 2005). These findings were reproduced in this study, and *Fu.*

aquatica is strongly supported by Bayesian and Maximum-likelihood values as a sister taxon to *Aquaticola miniguttulata* and a new *Aquaticola*-like species (PE0316) (**Fig. 7, Clade 1f**). **Clade 1f** is a lineage with Bayesian but not Maximum-likelihood support containing *Annulatascus biatriisporus*, a new *Annulatascus*-like species (PE0035), a new *Aquaticola*-like species (AK-1838), *Lentomitella* species, and *Torrentispora fibrosa*. Morphologically, the similarity of *Fu. aquatica* to Annulatascaceae rests on the presence of a J- ascus apical ring. Ascospores, however are long fusoid, a feature not seen in other members of Annulatascaceae *sensu stricto*. A type culture is not indicated in the protologue, and further molecular analysis is warranted using multiple genes to further hone the phylogenetic placement of this species. As *Fu. aquatica* is placed in a clade separating Diaporthales and Ophiostomatales it should be considered Sordariomycetes *incertae sedis*.

Longicollum Zelski, F.R. Barbosa, Raja, A.N. Mill. & Shearer, Mycosphere 2(5): 540. 2011. MycoBank 563241.

Type species: *Longicollum biappendiculatum.*

Longicollum biappendiculatum Zelski, H.A. Raja, A.N. Mill. & Shearer, Mycosphere 2(5): 540. 2011. MycoBank 653241.

Ascomata brown, immersed, perpendicular to surface, scattered, 155–160 µm wide × 205–220 µm high (x̅ = 157.5 × 212.5 µm, w:h = 1.4), subglobose. **Ascomal wall** brown, membranous, composed of 4–6 layers of pseudoparenchymatous cell, brown externally, subhyaline and laterally compressed internally, 15–20 µm, thick. **Neck** hyaline, cylindrical, 315–370 µm long × 74–82 µm wide, periphysate. **Paraphyses** hyaline, 5–7 µm wide at base × 140–170 µm long, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 180–225 µm long × 15–17 µm wide (x̅ = 205 × 16 µm, l:w = 12.8).

Ascus base thin tapering pedicel, 5 µm from *pars sporifera*. **Annulus** J-, bipartite, staining blue in aqueous nigrosin, 2–4 µm long × 3–5 µm wide ($\overline{x}$ = 3 × 4 µm, l:w = .75). **Ascospores** hyaline, broadly ellipsoidal, aseptate, uniseriate, 26–35 µm long × 12–14 µm wide ($\overline{x}$ = 28 × 13 µm, l:w = 2.3), multiguttulate, with short, ephemeral, bipolar mucilaginous appendages, 3–6 × 6–9 µm, without sheath.

Specimens examined:

HOLOTYPE: PERU. Cusco, Camanti, stream at Quincemil Trail 1, 13°14'22.5594"S, 70°46'12.6114"W, on submerged woody debris, water 20 °C, pH 6, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0017-2,* **ILL 40794.**

BRAZIL. Bahia, stream at Serra da Jibóia, 12°51'00"S, 39°28'46"W, on submerged bark debris, water 21.8°C, pH 4.3, 21 Jan 2009, *F.R. Barbosa and L.F.P. Gusmão, HUEFS 158104*. COSTA RICA. Heredia, La Selva Biological Station, La Selva stream, 10°25'7"N, 84°01'27"W, on submerged woody debris, pH 5, water temp 25°C, 10 Jan 2006, *M. Salazar and A. Ferrer, AF267-1*. PERU. Cusco, Camanti, stream at Quincemil Trail 1, 13°14'23"S, 70°46'13"W, on submerged woody debris, water 21°C, pH 6, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0017-1*. USA. Florida, Wildcat Lake, Ocala National Forest, 29°10'14''N, 84°37'40''W, on submerged woody debris, water 14 °C, pH 5, 2 February 2006, *J.L. Crane and H.A. Raja, F107-1;* Dorr Lake, Ocala National Forest, 29°00'24''N, 81°38'13''W, on submerged woody debris, water 33 °C, pH 5, July 16 2006, *J.L. Crane and H.A. Raja, F107-2*.

Known distribution: Brazil, Costa Rica, Perú, USA.

Comments: Illustrations of *Longicollum biappendiculatum* can be found in Zelski et al. 2011a. This species is a close sister to *Submersisphaeria aquatica* within the Annulatascaceae *sensu stricto* clade. This relationship holds for four genes evaluated individually as well as in combined dataset analyses of two and three genes, all with good support (**Figs. 2-7**). This pairing is somewhat surprising as the ascospores of *L. biappendiculatum* are hyaline and those of *Submersisphaeria aquatica* and other

S*ubmersisphaeria* species are dematiaceous. In addition, the ascus apical ring in *L. biappendiculatum* is much smaller than that of *S. aquatica*.

Mirannulata Huhndorf, F.A. Fern., A.N. Mill. & Lodge, Sydowia 55(2): 173. 2003. MycoBank 28775.

Type species: *Mirannulata samuelsii.*

Mirannulata samuelsii Huhndorf, F.A. Fern., A.N. Mill. & Lodge, Sydowia 55(2): 173. 2003. MycoBank 488322.

Ascomata dark brown to black, superficial, perpendicular to surface, numerous, 480–525 µm wide × 550–630 µm high (x̅ = 502.5 × 509 µm, w:h = .9), obpyriform to ovoid. **Ascomal wall** dark brown, glabrous, composed of 2 layers, polygonal to elongate parenchymatous cells, melanized crust darker externally, lighter to hyaline and compressed internally. **Neck** dark brown, bluntly conical to broadly rounded, 100–150 µm high × 120–130 µm wide at apex, 250–300 µm wide at base, periphysate. **Paraphyses** hyaline, 3–4.5 µm wide at base, tapering toward apex, septate, abundant, persistent. **Asci** unitunicate, cylindrical, 8–spored, clavate, 140–185 µm long × 13–18 µm wide (x̅ = 165.5 × 15.5 µm, l:w = 10.6). **Ascus base** short, 15 µm from *pars sporifera*. **Annulus** J-, large, bipartite, 4.8–6 µm long × 5–6.5 µm wide (x̅ = 5.4 × 5.75 µm, l:w = .94). **Ascospores** hyaline, fusiform, 3–septate, not constricted at septa, biseriate, 33.6–43.3 µm long × 5.4–7.3 µm wide (x̅ = 38.5 × 6.3 µm, l:w = 6.1), with prominent guttules in each cell, without appendages or sheath (Huhndorf et al. 2003).

HOLOTYPE: UNITED STATES. Puerto Rico, El Verde Research Area, Luquillo Mountains, 18°19’26”N, 65°49’1”W, 372m, on 25 cm diam. stump of *Tetragastris balsamifera*, ***SMH1880, Holotype F***.

Known distribution: Costa Rica, Ecuador, French Guiana, Puerto Rico.

Comments: The holotype of *Mirannulata samuelsii* was not examined and no fresh collection was made in this study. Huhndorf et al. (2003) compared the species to Annulatascaceae, suggesting affinity to the genera *Annulatascus* and *Cataractispora*. The authors however suggested that this species, with a large bipartite apical ring, did not belong in Annulatascaceae *sensu stricto* and instead placed it as Sordariomycetes *incertae sedis*. The ascomata are superficial with bluntly conical necks while members of the Annulatascaceae are immersed to erumpent, but with well-developed cylindrical necks. The large apical rings, shape of the ascospores, and the thickness of ascospore walls resemble *C. viscosa* or *C. aquatica*, but *Cataractispora* ascomata are quite different, having thinner walls and well developed necks. Miller (unpub.) in an analysis of Sordariomycetes found *M. samuelsii* to be unrelated to Annulatascaceae and other large-ringed taxa sampled (Huhndorf et al. 2003). A second species, *M. costaricensis* Huhndorf, F.A. Fern., A.N. Mill. & Lodge, has also been described but does not have sequences in GenBank. It would be worthwhile to sequence *M. costaricensis* and compare it with *M. samuelsii* and Annulatascaceae, as the species does possess relatively large J- apical rings (Huhndorf et al. 2003). This study corroborates that *Mirannulata* is not a member of Annulatascaceae *sensu stricto* and should still be considered Sordariomycetes *incertae sedis* (**Fig. 7, sister to Clade 7**).

Paoayensis Cabanela, Jeewon & K.D. Hyde, Cryptogamie Mycologie 28(4): 303. 2007. MycoBank 510824.

Type species: *Paoayensis lignicola.*

Paoayensis lignicola Cabanela, Jeewon & K.D. Hyde, Cryptogamie Mycologie 28 (4): 304. 2007. MycoBank 510825.

Ascomata immersed to slightly erumpent, small groups developing under a pseudostroma, 546–626 µm wide × 520–586 µm high ($\bar{x}$ = 586 × 553 µm, w:h = 1.1), globose to obpyriform. **Ascomal wall** light brown to black, darker externally, lighter to hyaline and compressed internally. **Neck** small common ostiole. **Paraphyses** hyaline, 3–10 µm wide at base, tapering toward apex, septate, rarely branching, early deliquescent. **Asci** unitunicate, clavate, 4-spored, clavate, 45–130 µm long × 13–35 µm wide ($\bar{x}$ = 90 × 23 µm, l:w = 2.6). **Ascus base** short, pedicellate. **Annulus** J-, discoid, early deliquescent, rarely seen. **Ascospores** brown to dark brown, lemoniform, with a 10–12 µm long germ slit, 0–3 septate, not constricted at septa, uniseriate to slightly overlapping uniseriate, 53–90 µm long × 30–58 µm wide ($\bar{x}$ = 77 × 4 µm, l:w = 1.8), without appendages or sheath (Cabanela et al. 2007).

HOLOTYPE: Philippines, Ilocos Norte Province, Paoay, Paoay Lake, 18°12’N, 123°53’E, on submerged wood, 6 Jan 2006, *M.V. Cabanela*, **HKU(M) 17516.**

Known distribution: The Republic of the Philippines.

Comments: The holotype of *Paoayensis lignicola* was not examined nor were fresh specimens collected in this study. While molecular analyses demonstrate an affinity to Annulatascaceae *sensu stricto* with poor support, this species is a morphological anomaly compared with the other members. Ascospores are massive and have germ slits, features not present in any other Annulatascaceae *sensu stricto* taxa. In addition, ascomata are clustered in a pseudostroma and the asci possess discoid apical rings. Fresh collections should be made at Paoay Lake in the Philippines to obtain new sequence data to assure the veracity of the original assessment. DNA from a fresh culture could be compared to the ex type culture HKUCC 10927. Due to morphological differences, low support values, and being a separate branch within the Annulatascaceae *sensu stricto* clade, this species is best regarded at present as Sordariomycetes *incertae sedis*.

Pseudoproboscispora Punith., Kew Bulletin 54 (1): 234. 1999. MycoBank 28391.

Type species: *Pseudoproboscispora aquatica.*

Pseudoproboscispora aquatica (S.W. Wong & K.D. Hyde) Punith., Kew Bulletin 54(1): 234. 1999. **Fig. 50 A-B.** MycoBank 460710 (*Proboscispora aquatica* MycoBank 450112, legitimate).

≡ *Proboscispora aquatica* S.W. Wong & K.D. Hyde, Mycological Research 103(1): 82, 1999.

Ascomata light or dark brown, coriaceous, immersed, solitary to gregarious, 196–280 µm wide, ellipsoidal. **Ascomal wall** brown, several layers of compressed brown cells. **Neck** hyaline, short. **Paraphyses** hyaline, tapering, septate. **Asci** unitunicate, cylindrical, 8–spored, 135–140 µm long × 14–15 µm wide ($\overline{x}$ = 137.5 × 14.5 µm, l:w = 9.5). **Ascus base** pedicellate, broadly tapering to rounded base, 50 µm from *pars sporifera*. **Annulus** J-, refractive, angled, cylindrical, 3 µm long × 5 µm wide (l:w = .6). **Ascospores** hyaline, fusiform, 3–septate, uniseriate to overlapping uniseriate, 22.5–24 µm long × 7.5–8 µm wide ($\overline{x}$ = 23.25 × 7.75 µm, l:w = 3), guttulate, with bipolar filamentous appendages, without sheath (Wong and Hyde 1999).

Specimens examined:

HOLOTYPE: AUSTRALIA. North Queensland, near Cairns, Crystal Cascades, on submerged wood, Jun 1996, *K.D. Hyde and T.M. Hyde, CC63,* **HKU(M) 2792.**

Fig. 50 A-B. *Pseudoproboscispora aquatica* from the holotype **HKU(M) 2792**. A. Herbarium packet. B. Dried wood substrate.

Fig. 50.

Reports:

HONG KONG. New Territories, Tsuen Wan, Shing Mun Reservoir, on submerged wood, 14 Jan 1988, *Clement Tsui, HKU(M) 8145;* Tai Po, Lam Tsuen River, on submerged wood, Dec 1996, *Clement Tsui, HKU(M) 5364, HKU(M) 4667, HKU(M) 5363;* Apr 1997, *Clement Tsui HKU(M) 5424, HKU(M) 5425, HKU(M) 5426*; Jan 1998, *Clement Tsui, HKU(M) 5375*. PHILIPPINES. Negros Occidental, Bacolod, Kaliban River, on submerged wood, 18 Nov 1994, *K.D. Hyde and E. Arimas, HKU(M) 3119*.

Known distribution: Australia, Hong Kong, Philippines.

Comments: The holotype of *Pseudoproboscispora aquatica* was in poor condition and consisted of one piece of wood twig 20 cm long × 2 cm in diameter (**Fig. 50 A,B**). The dried material was devoid of fungal material matching the description of *P. aquatica,* though multiple ascomata were inspected (**Fig. 50 B)**. For now the protologue (Wong et al. 1999) must serve for comparison until neotypification from Australia can be performed. Fresh specimens are required for molecular analyses. *P. caudae-suis*, the second species in the genus, belongs in the Annulatascaceae *sensu stricto,* and thus *P. aquatica* cannot be ruled out as a member at this time. The main feature separating these two taxa with coiled unfurling bipolar ascospore appendages is that *P. aquatica* has shorter, thinner, 3–septate ascospores vs. the uniseptate (sometimes 3–septate) ascospores of *P. caudae-suis*. These distinctive appendages are also found in *Ayria* species and in members of the Halosphaeriales which are found to be unrelated to *P. caudae-suis* in this study.

Pseudoproboscispora caudae-suis (Ingold) J. Campb., Shearer, J.L. Crane & Fallah, Mycologia 95(1): 45. 2003. **Fig. 51 A-J, Fig. 52 A-H.** MycoBank 373568 (*Ceriospora caudae-suis* MycoBank 294658, legitimate).

≡ *Ceriospora caudae-suis* Ingold, Transactions of the British Mycological Society 34(2): 210. 1951.

Ascomata black, immersed to semi-immersed, scattered to gregarious, 400–700 µm wide × 300–500 µm high (x̅ = 550 × 400 µm, w:h = 1.4), oblong naviculoid, obpyriform (**Fig. 52 A**). **Ascomal wall** brown and subhyaline, 19.5–27-5 µm thick, 5–7 layers of brown pseudoparenchymatous cells, thicker at base of beak, inner cells subhyaline19.5–27-5 µm thick, of *textura angularis* in surface view (**Fig. 51 D**). **Neck** black, subhyaline at apex, short, 136.5–156 µm long × 109–117 µm wide, periphysate. **Paraphyses** hyaline, 5–6 µm wide at base × 150–200 µm long, tapering toward apex, septate (**Fig. 52 B**). **Asci** unitunicate, cylindrical, 8–spored, 190–343 µm long × 15–21 µm wide (x̅ = 266.5 × 16.5 µm, l:w = 16.1) (**Fig. 51 E,F; Fig. 52 C**). **Ascus base** pedicellate, narrow, 25 µm from *pars sporifera* (**Fig. 52 D**). **Annulus** J-, apical pore, or extremely shallow disc (**Fig. 51G-I, Fig. 52 E**). **Ascospores** hyaline, oval, 1–septate (occasionally 3–septate), uniseriate, 18–36 µm long × 9–18 µm wide (x̅ = 27 × 13.5 µm, l:w = 2), multiguttulate, with bipolar unraveling appendages, 2–3 µm wide × 50–120 µm long, without sheath (**Fig. 51 J, Fig. 52 F-H**) (Campbell et al. 2003).

Specimens examined:

HOLOTYPE: UNITED KINGDOM. Westmoreland, Windemere, on submerged decorticated *Fraxinus* twigs, 14 Nov 1949, *Ingold C.T., K(M) 180916*, **IMI 38506.**

USA. Sleeping Giant State Park, Quinipiac River, 41°25'54.624"N, 72°54'18.0714"W, 93m, on submerged woody debris, water 19.1 °C, pH 9.7, 237 µS, 17 Jul 2012, *Zelski S.E. and V.P. Hustad, Z32-1822.* North Carolina, GSMNP, riparian stream in Okanaluftee watershed, 35°30’7.2354”N, 83°17’49.56”W, 622m, on submerged woody debris, water 21.8 °C, pH 8.5, 16 Jul 2010, *Zelski S.E. and H.A. Raja, Z32-1;* Pennsylvania. Ricketts Glen State Park, stream along Falls Trail, 41°20’1.356”N,

Fig. 51 A-J. *Pseudoproboscispora caudae-suis* from the holotype **IMI 38506**. A. Herbarium packet. B. Herbarium packet contents. C. Dried culture. D. Peridium. E. Ascus. F-G. Asci. H. Ascus illustrating annulus and annulus plug. I. Annulus plug. J. Annulus. Bars: D-J = 20 µm.

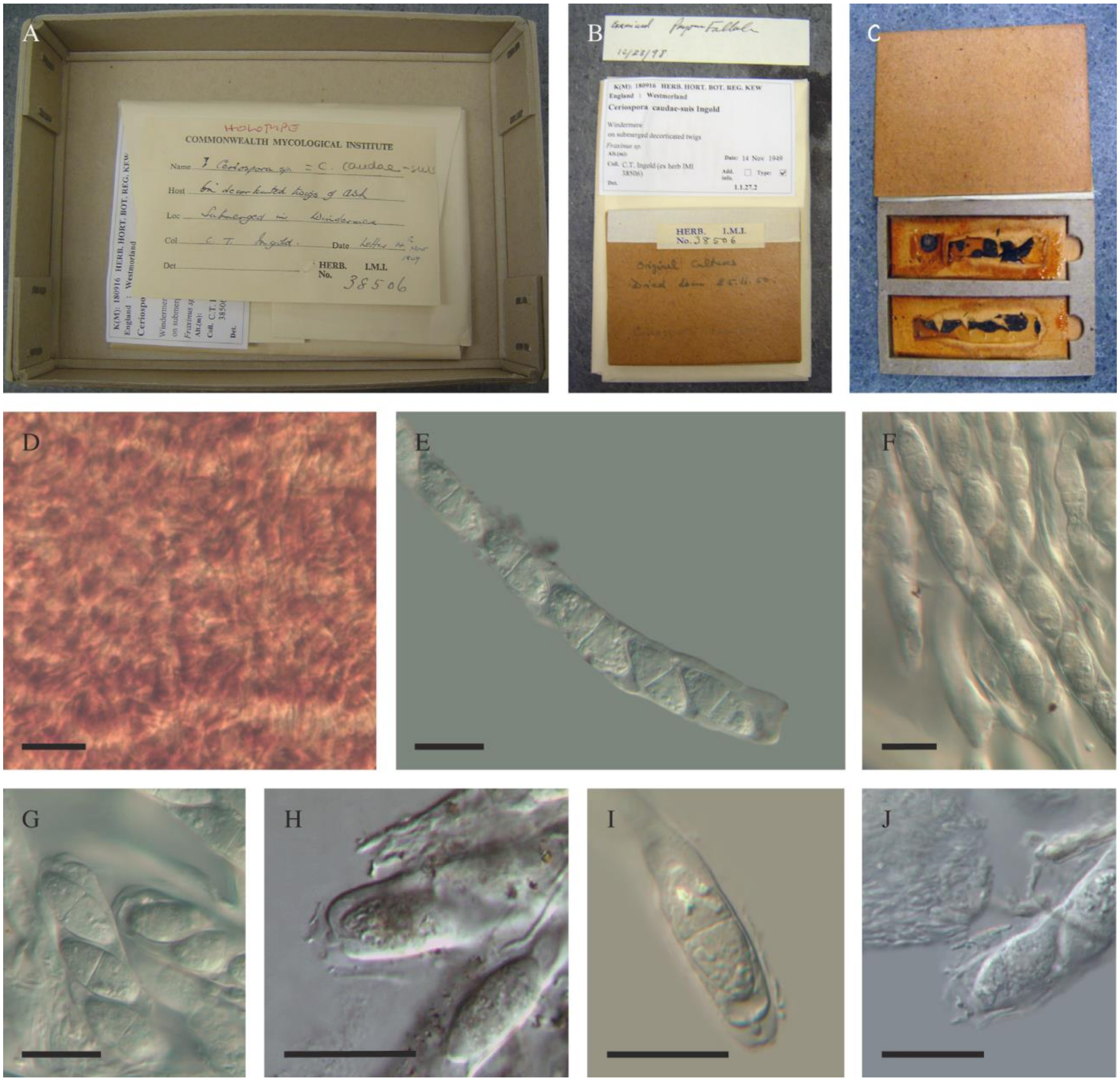
A
HOLOTYPE
COMMONWEALTH MYCOLOGICAL INSTITUTE
Name ? Ceriospora sp. = C. caudae-suis
Host on decorticated twigs of ash
Loc Submerged in Windermere
Col C. T. Ingold. Date Letter 14th Nov 1949
Det
HERB. I.M.I. No. 38506
K(M): 180916 HERB. HORT. BOT. REG. KEW
England : Westmorland
B
examined
12/28/98
K(M): 180916 HERB. HORT. BOT. REG. KEW
England : Westmorland
Ceriospora caudae-suis Ingold
Windermere
on submerged decorticated twigs
Fraxinus sp.
Alt.(m):
Date: 14 Nov 1949
Coll. C.T. Ingold (ex herb IMI 38506)
Add. info. ☐ Type: ☑
Det.
1.1.27.2
HERB. I.M.I. No. 38506
Original cultures
Dried down 25.11.50.
C
D
E
F
G
H
I
J

Fig. 51.

Fig. 52 A-H. *Pseudoproboscispora caudae-suis* from Z32-1824. A. Ascomata on substrate. B. Paraphyses. C. Ascus. D. Pedicellate ascus base. E. Ascus apex. F. Ascospore with two large lipid drops. G. Ascospore in aqueous nigrosin illustrating coiled appendages. H. Ascospore with appendages extended. Bars: A = 500 µm; B-D, F-H = 20 µm; E = 10 µm.

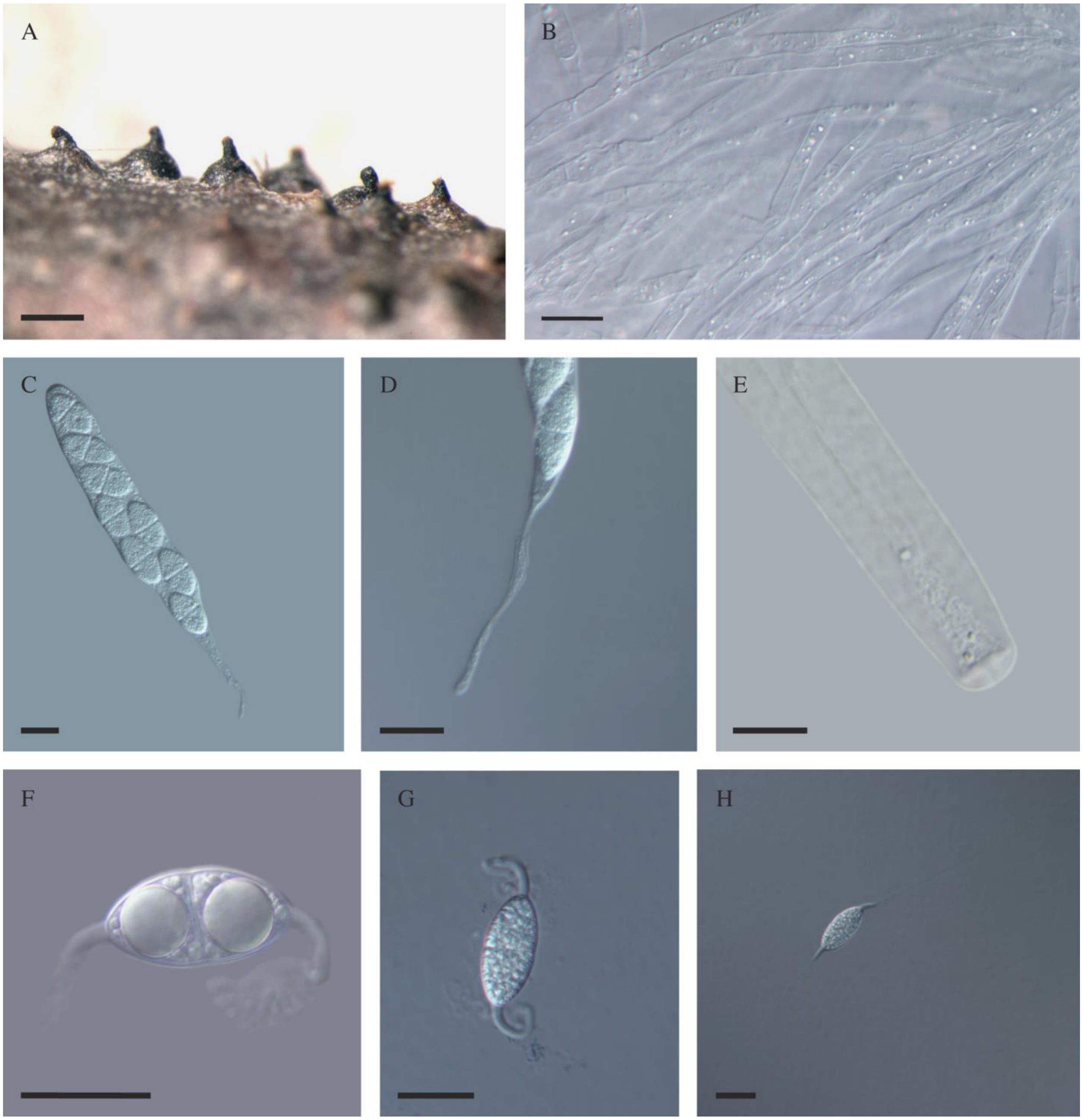


Fig. 52.

76°16’15.4194”W, on submerged woody debris, water 19.3 °C, pH 9, 38 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z32-1824;* Tennessee, GSMNP, riparian stream in Pigeon River watershed, 35°40’32.8434”N, 83°31’37.848”W, 536m, on submerged woody debris, water 20 °C, pH 7.4, 17 Jul 2010, *Zelski S.E. and H.A. Raja, Z32-2;*Wisconsin, Lemonweir River, on submerged decorticated wood, 5 Oct 2009*, H.A. Raja and A.N. Miller, A40-40.*

Known distribution: United Kingdom, United States.

Comments: The holotype of *Pseudoproboscispora caudae-suis* consists of two dried cultures plated in a 2 slide mailer and original documentation (**Fig. 51 A-C**). Ascomata formed in culture are present in the dried culture. Asci were difficult to tease apart after 5 min in 5% KOH and appeared cemented together, perhaps due to the ascospore appendages. The ascus apical apparatus of the holotype appears to consist of a small ring and the remnants of an apical plug. In fresh collections, appendages strongly adhered to dissecting needles when manipulated.

Campbell et al. (2003) reassessed *Ceriospora caudae-suis* Ingold and noted two morphological features not noted by Ingold in his original description. The first difference was that the distal ends of the bipolar appendages were found to separate into many long filaments. The second was that the asci readily separate from the hymenium when mounted in water. Variation in septation (1–3) has been noted in this fungus, and was observed in examination of the holotype. Campbell et al. (2003) placed this species in *Pseudoproboscispora* (S. W. Wong and K. D. Hyde) Punith. based on its high degree of similarity to *P. aquatica*, the type of the genus. Molecular analysis of *P. caudae-suis* also suggested affinity to *Annulatascus velatisporus*, the type species of Annulatascaceae, and it was thus was included in the family (Campbell et al. 2003). This study places it in Annulatascaceae *sensu stricto* as well (**Figs. 2-7**). Of note is the lack of a massive ascus apical apparatus (rather a shallow ring or ascus apical pore), typical of other members of the family.

Rivulicola K.D. Hyde, Nova Hedwigia 64: 186. 1997. MycoBank 27726.

Type species: *Rivulicola incrustata.*

Rivulicola aquatica V. M. Ranghoo & K. D. Hyde, Mycologia 92(5): 1023. 2000. **Fig. 53 A-B.** MycoBank 467603.

= *Vertexicola lignicola* V.M. Ranghoo & K.D. Hyde, Ranghoo Ph.D. thesis, 75. 1998.

Ascomata hyaline, membranous, semi-immersed, perpendicular to surface, solitary to gregarious, 140–150 µm wide × 175–180 µm high ($\bar{x}$ = 145 × 177.5 µm, w:h = .8), subglobose. **Ascomal wall** hyaline, 5–7 layers of hyaline compressed cells, 20–30 µm thick. **Neck** hyaline to brown, 50–88 µm long × 50–60 µm wide, periphysate. **Paraphyses** hyaline, 5.6–7 µm wide at base × 120–150 µm long, tapering toward apex cylindrical, septate, branched. **Asci** unitunicate, cylindrical, 8–spored, 125–162 µm long × 22–25 µm wide ($\bar{x}$ = 143.5 × 23.5 µm, l:w = 6). **Ascus base** pedicellate, tapering to club-like base, 15 µm from *pars sporifera*. **Annulus** J-, discoid, 2.5–4 µm long × 4–5 µm wide ($\bar{x}$ = 3.25 × 4.5 µm, l:w = .72). **Ascospores** hyaline, ellipsoidal, 1–3 septate, not constricted at septa, overlapping uniseriate to biseriate, 15–20 µm long × 7.5–12.5 µm wide ($\bar{x}$ = 17.5 × 10 µm, l:w = 1.75), multiguttulate, with thin appendages with fibrillar ornamentation, without sheath (Ranghoo et al. 2000).

Specimens examined:

HOLOTYPE: CHINA. Hong Kong, Tai Po Cove Reservoir, on submerged wood, Jan 1997, *V.M. Ranghoo, PC 61, (HKU(M) 5214)*, **IFRD 8843.**

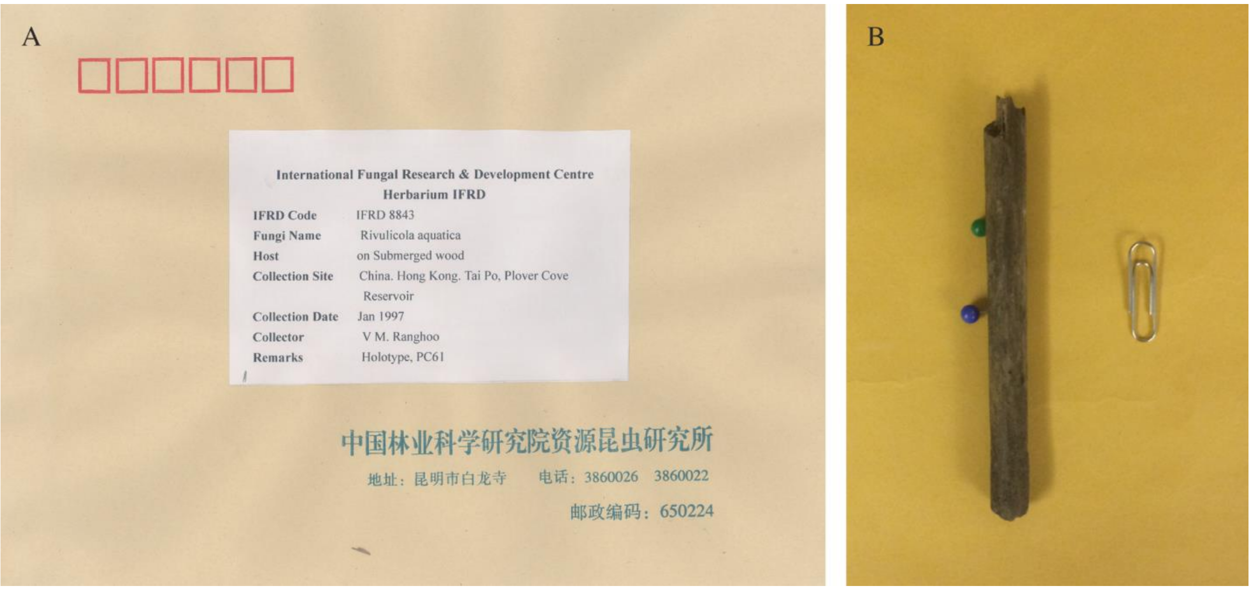
Known distribution: China.

Comments: The holotype lacked fungal material of *Rivulicola aquatica* and fresh specimens were not obtained in this study (**Fig. 53 A,B**). It is recommended that the species be neotypified. Hyde introduced the genus *Rivulicola* for a freshwater ascomycete, *R. incrustata*, found on submerged wood in Australia (Hyde et al. 1997). A key feature of the type of the genus is that it stains its substrate and its hyaline ascomata green, leaving protruding predominantly hyaline necks. *R. aquatica* was originally informally published as *Vertexicola lignicola* (Ranghoo 1998) though it was the only species in *Vertexicola* with 3-septate ascospores, with *V. aquatica* and *V. caudatus* having 5–septate ascospores. Ranghoo et al. (2000) included *R. aquatica,* collected in Hong Kong, as the second species in *Rivulicola*. It was included based on the hyaline necks of the ascomata, and the morphologically similar ascospores which differ only in septation from those of *R. incrustata* (1–3 septate in *R. aquatica* vs. 3–5 septate in *R. incrustata*). That study contributed curious observations of *R. incrustata* collected in Hong Kong that did not turn the substrate green. These may represent morphological variation in *R. aquatica*. Peruvian specimens were all found on green stained substrates. Molecular data are needed to determine the placement of *R. aquatica* in the genus *Rivulicola*, which is shown not to be affiliated with Annulatascaceae *sensu stricto* in this study based on sequence data from *R. incrustata*, but rather has affinity to Hypocreales (**Fig. 7, Clade 6**). The genus and *R. incrustata* should thus be considered Sordariomycetes *incertae sedis* at present.

Rivulicola cygnea Raja & Shearer, Fungal Diversity 34: 83. 2009. **Fig. 54 A-I.** MycoBank 512706.

Ascomata hyaline to pale brown, membranous, immersed, parallel to surface, scattered to gregarious, 470–680 µm wide × 140–170 µm high ($\bar{x}$ = 575 × 155 µm, w:h = 3.7), globose to subglobose. **Ascomal wall** hyaline to light brown, 4–5 layers of hyaline to light brown, elongated, flattened cells, 10–12 µm thick. **Neck** hyaline, 240–500 µm long × 30–40 µm wide, with short hyaline protruding hyphae, 10–15

Fig. 53 A-B. *Rivulicola aquatica* from the holotype **IFRD 8843**. A. Herbarium packet. B. Dried material.



International Fungal Research & Development Centre
Herbarium IFRD

IFRD Code	IFRD 8843
Fungi Name	Rivulicola aquatica
Host	on Submerged wood
Collection Site	China. Hong Kong. Tai Po, Plover Cove Reservoir
Collection Date	Jan 1997
Collector	V M. Ranghoo
Remarks	Holotype, PC61

中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224

Fig. 53.

µm long, periphysate. **Paraphyses** hyaline, 3 µm wide at base, tapering toward apex, septate, fasciculate. **Asci** unitunicate, cylindrical, 8–spored, 122–155 µm long × 7–8 µm wide (x̅ = 138.5 × 7.5 µm, l:w = 18.5) (**Fig. 54 E,F**). **Ascus base** pedicellate, short, broadly tapering to a rounded base, 20 µm from *pars sporifera*. **Annulus** J-, refractive 4 µm long × 2 µm wide (l:w = 2) (**Fig. 54 G**). **Ascospores** hyaline, ellipsoidal, 3–septate, not constricted at septa, overlapping uniseriate, 20–24 µm long × 6–7 µm wide (x̅ = 22 × 6.5 µm, l:w = 3.4), multiguttulate, without appendages, with a sheath 2–3 µm wide, sheath constricted at first septum (**Fig. 54 H,I**) (Raja et al. 2009).

Specimens examined:

HOLOTYPE: USA. Florida, Apalachicola National Forest, unnamed lake east of Lost Lake, 30°21’48”N, 84°22’56”W, on submerged decorticated wood, water 35 °C, pH 7.3, 13 Jul 2004, *H.A. Raja and Chris Brown, F41-1*, **ILL40111.**

Comments: The holotype of *Rivulicola cygnea* consists of 4 slides (**Fig. 54 A-D**). Ascospores still appear fresh, though not in as good condition as those used in the protologue micrographs. Evidence of an apical ring was found in only one ascus in the holotype material, which may be due to preservation in lactic acid. The annulus was neither illustrated nor were annulus dimensions reported on the protologue. *R. cygnea* possesses ascospores with sheaths that are well developed, while ascospores of *R. incrustata* possess irregular granular sheaths. Raja et al. (2008) included this species in *Rivulicola* and differentiated it from *R. incrustata* based on the superficial position of the ascomata, though *R. incrustata*, as its name implies, has deeply immersed, or incrusted ascomata. Green staining of the surrounding substrate by *R. incrustata*, but not in *R. cygnea*, is another feature that distinguishes the two species. In addition, the ascospores of *R. cygnea* are consistently 3–septate with a gelatinous sheath constricted at the first septum, while those of *R. incrustata* are 3–4–5 septate and have an irregular sheath. This genus has been referred to Annulatascaceae, though it appears to have a relatively small ascus apical apparatus. Based on molecular analyses, *R. incrustata* is not affiliated with Annulatascaceae *sensu stricto* in this study (**Fig. 7,**

Clade 6) thus *R. cygnea* should thus be excluded as well. The genus should thus be considered Sordariomycetes *incertae sedis*. Fresh collections from Florida to obtain molecular data are warranted to determine generic and higher level phylogenetic placement of *R. cygnea*.

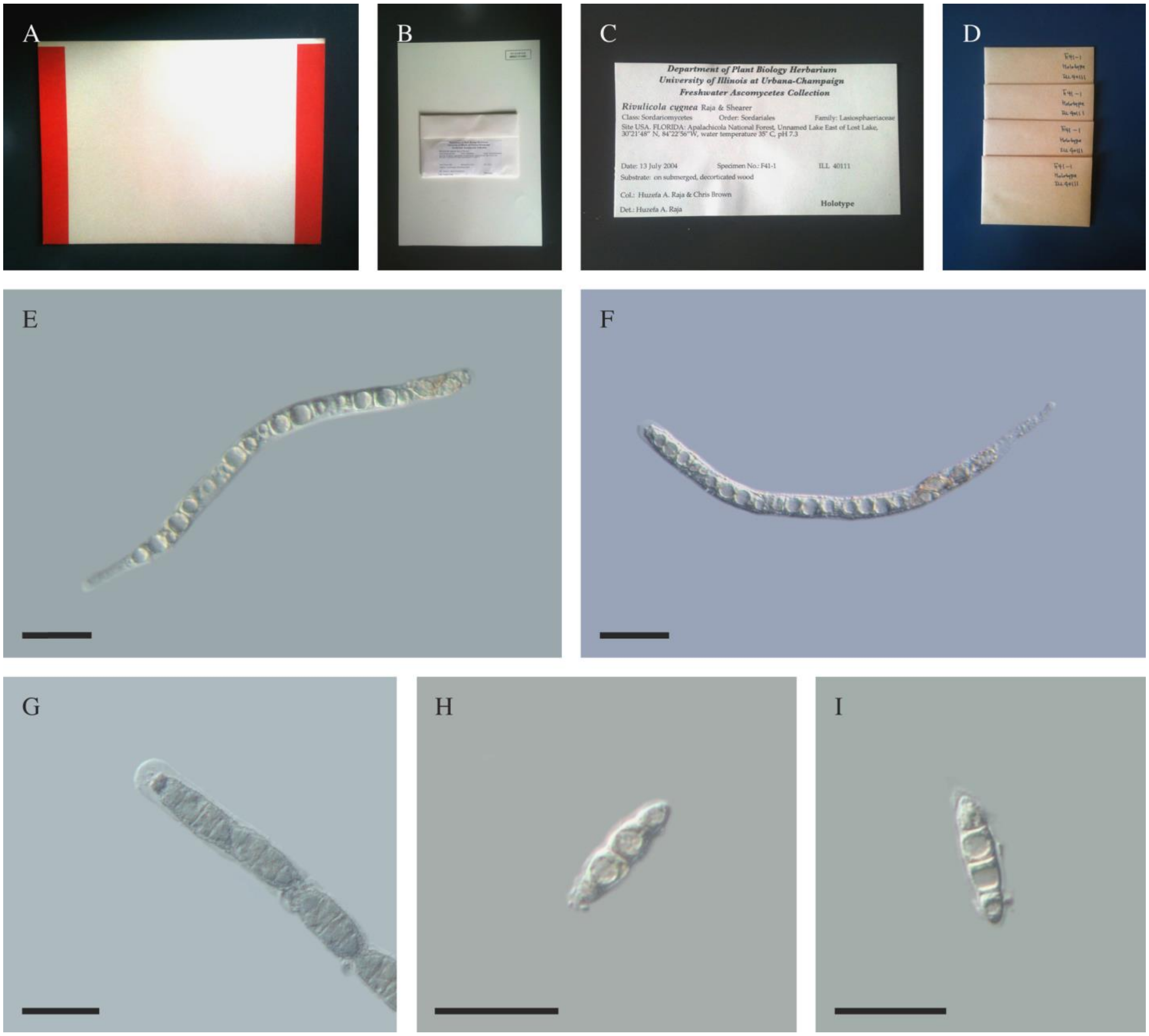
Rivulicola incrustata K.D. Hyde, Nova Hedwigia 64: 186. 1997. **Fig. 55 A-P, Fig. 56 A-H.** MycoBank 436671.

Ascomata hyaline, immersed, parallel to surface, scattered to gregarious, 160 µm wide × 280 µm high (w:h = .6), compressed-globose (**Fig. 55 H-J**). **Ascomal wall** hyaline, elongate near base, of *textura angularis* near neck, 20 µm thick, of *textura angularis* in surface view (**Fig. 55 K,L**). **Neck** hyaline and green, periphysate. **Paraphyses** hyaline, 4 µm wide at base, septate, filamentous. **Asci** unitunicate, cylindrical, 8–spored, 130–205 µm long × 11–12 µm wide ($\overline{x}$ = 167.5 × 11.5 µm, l:w = 14.6) (**Fig. 55 M, Fig. 56 A**). **Ascus base** pedicellate, short, 5–25 µm from *pars sporifera* (**Fig. 55 M, Fig. 56 A,D**). **Annulus** J-, refractive, 4.1 µm wide (**Fig. 56 B,C**). **Ascospores** hyaline, ellipsoidal, 3–4–5 septate, not constricted at septa, uniseriate, 21–27 µm long × 8–12 µm wide ($\overline{x}$ = 24 × 10 µm, l:w 2.4), multiguttulate, without appendages, with an irregular, granular sheath (**Fig. 55 N-P, Fig. 56 E-H**) (Hyde et al. 1997).

Specimens examined:

HOLOTYPE: AUSTRALIA. Queensland, Clohesy River near Kurunda, on submerged wood, Mar 1991, *K.D. Hyde, KDH 601*, **BRIP 23340.** PERU. Cusco, Camanti, stream at Quincemil Trail 3, 13°17’7.008”S, 70°47’13.632”W, 737m, on submerged woody debris, water 21.7 °C, pH 7.6, 27 May 2010, *Zelski S.E. and H.A. Raja., PE0031-1;* Stream at Quincemil Trail 3, on submerged woody debris, 13°17’7.008”S, 70°47’13.632”W, 737m, water 21.7 °C, pH 7.6, 4 Oct 2010, *Zelski S.E. and H.A Raja, PE0031-2;* Stream crossing the Interoceanic Highway, on submerged woody debris, 13°17’7.008”S, 70°47’13.632”W,

Fig. 54 A-I. *Rivulicola cygnea* from the holotype **ILL 40111**. A. Herbarium folder. B. Herbarium sheet. C. Herbarium label. D. Herbarium packet contents (4 slides). E-F. Asci. G. Ascus apex illustrating apical ring. H-I. Ascospores. Bars: E-I = 20 µm.



Department of Plant Biology Herbarium
University of Illinois at Urbana-Champaign
Freshwater Ascomycetes Collection

Rivulicola cygnea Raja & Shearer
Class: Sordariomycetes Order: Sordariales Family: Lasiosphaeriaceae
Site USA. FLORIDA: Apalachicola National Forest, Unnamed Lake East of Lost Lake, 30°21'48" N, 84°22'56"W, water temperature 35° C, pH 7.3

Date: 13 July 2004 Specimen No.: F41-1 ILL 40111
Substrate: on submerged, decorticated wood

Col.: Huzefa A. Raja & Chris Brown
Holotype
Det.: Huzefa A. Raja

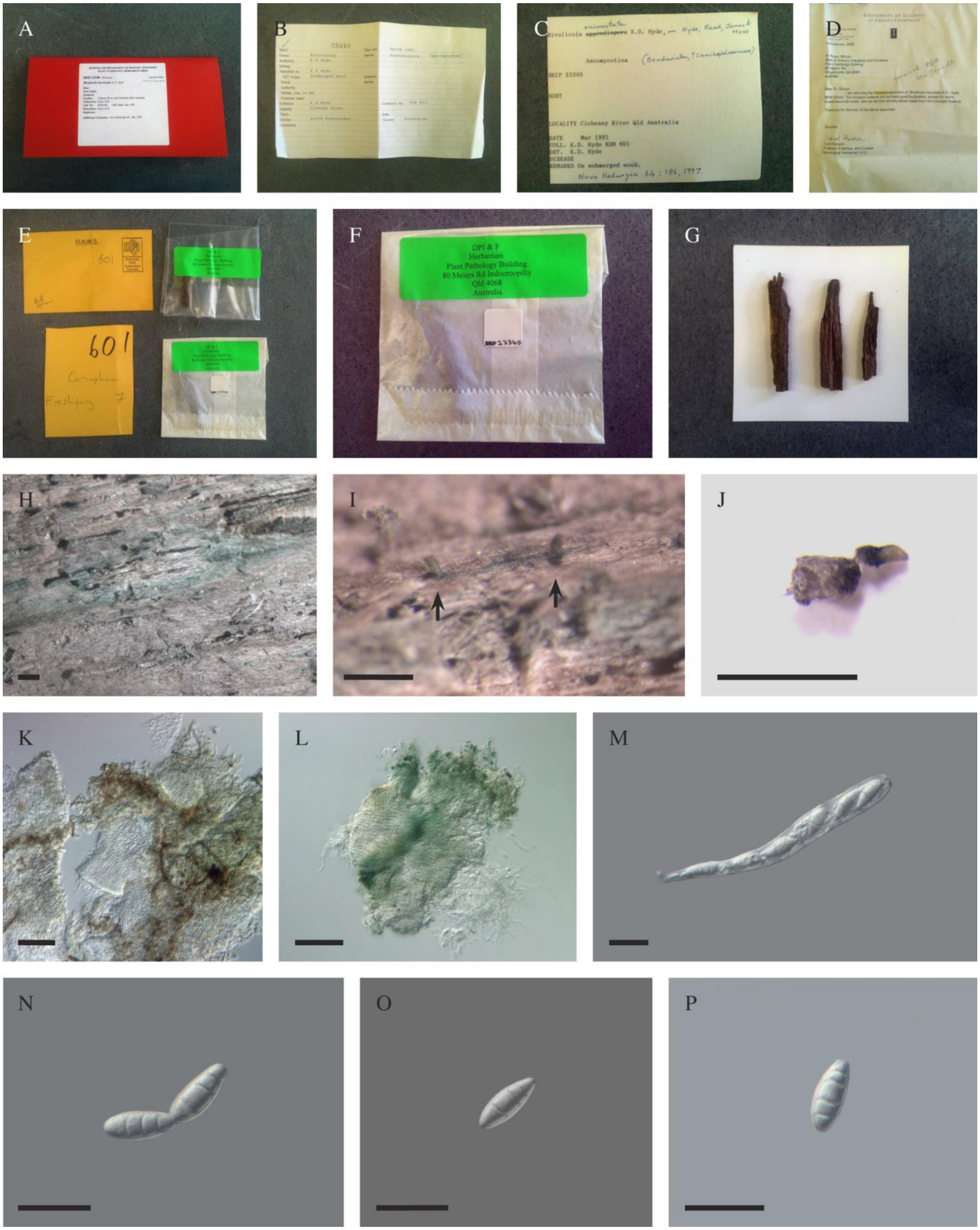
Fig. 54.

737m, water 22 °C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0031-7;* Stream at Quincemil Trail 3, on submerged woody debris, 13°18’27.756”S, 70°48’44.9274”W, 737m, water 21.2 °C, pH 7.8, 13 May 2011, *Zelski S.E. and H.A. Raja., PE0031-8;* Madre de Dios, Rio Amigos, on submerged woody debris, 12°34’13.008”S, 70°4’14.7714”W, 218m, water 31.4 °C, pH 8, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0031-5.*

Known distribution: Australia, Perú.

Comments: The holotype of *Rivulicola incrustata* was in good condition (**Fig. 55 A-G**) and fresh specimens were obtained from Perú from which DNA was extracted. The genus *Rivulicola* was erected by Hyde et al. (1997) to accommodate a *Ceratosphaeria*-like ascomycete, *R. incrustata*, found on submerged wood in the Clohesy River in north Queensland, Australia. The authors used the presence of a relatively massive ascus apical ring was used to suggest inclusion in Annulatascaceae. One striking feature of this fungus is that it stains the substrate, its hyaline venter, and part of its hyaline neck green. This fungus forms ascomata within dense woody substrates and is difficult to dissect out of the substrate. Though originally described as 3–4–5 septate, only ascospores with two septa were present in the holotype material. *R. incrustata* is not affiliated with Annulatascaceae *sensu stricto* in the molecular analyses performed in this study, and it, as well as the genus should thus be considered Sordariomycetes *incertae sedis* at present (**Fig. 7, Clade 6**).

Fig. 55 A-P. *Rivulicola incrustata* from the holotype **BRIP 23340**. A. Herbarium packet. B-D. Specimen documentation. E. Specimen documentation and dried material. F. Packet containing dried material. G. Dried wood. H. Surface of substrate showing green staining. I. Necks protruding above surface of substrate (arrows). J. Ascoma dissected from substrate. K. Hyaline peridium. L. Peridium with green staining. M. Ascus. N-O. Ascospores. Bars: H-J = 500 µm; K-L = 40 µm; M-P = 20 µm.



Rivulicola ~~appendiospora~~ inornatata K.D. Hyde, in Hyde, Read, Jones & Moss

Ascomycotina (Sordariales, ? Lasiosphaeriaceae)

BRIP 23340

HOST

LOCALITY Clohesey River Qld Australia

DATE Mar 1991
COLL. K.D. Hyde KDH 601
DET. K.D. Hyde
DISEASE
REMARKS On submerged wood.
Nova Hedwigia 64: 186, 1997.

DPI & F
Herbarium
Plant Pathology Building
80 Meiers Rd Indooroopilly
Qld 4068
Australia

BRIP 23340

Fig. 55.

Fig. 56 A-H. *Rivulicola incrustata* from PE0031. A. Ascus with immature ascospores. B. Immature ascus apex. C. Ascus apex illustrating apical ring. D. Ascus base. E. Immature ascospore. F-H. Ascospores. Bars: A-D = 20 µm; E-H = 10 µm.

Fig. 56.

Submersisphaeria K.D. Hyde, Nova Hedwigia 62: 172. 1996. MycoBank 27640.

Type species: *Submersisphaeria aquatica.*

Submersisphaeria aquatica K.D. Hyde, Nova Hedwigia 62: 172, 1996. **Fig. 57 A-I, Fig. 58 A-L.**

MycoBank 415480.

Ascomata brown, coriaceous, immersed, perpendicular to surface, scattered, 180–250 µm wide, globose to subglobose (**Fig. 57 D,E; Fig. 58 A,B**). **Ascomal wall** light brown and brown, outer layer of brown angular cells and inner layer of light brown flattened cells, 30 µm thick (**Fig. 57 G, Fig. 58 D**). **Neck** black, 560 µm long × 70 µm wide, composed of host tissue, angular fungal cells and black particles, periphysate (**Fig. 58 C**). **Paraphyses** hyaline, 3–4 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 175–210 µm long × 10–12.5 µm wide ($\bar{x}$ = 192.5 × 11.3 µm, l:w = 17.1) (**Fig. 58 E,F**). **Ascus base** short-pedunculate, with a club-like end, 20 µm from *pars sporifera*. **Annulus** J-, refractive, 4–5 µm long × 6–7 µm wide ($\bar{x}$ = 4.5 × 6.5 µm, l:w = .69) (**Fig. 58 E-J**). **Ascospores** brown, ellipsoidal to fusiform, 1–septate, slightly constricted at each septum, 23–27 µm long × 7.5–10 µm wide ($\bar{x}$ = 25 × 8.75 µm, l:w = 2.9), with granular contents, hyaline germ pores, without appendages or sheath (**Fig. 57 H,I; Fig. 58 K,L**) (Hyde 1996).

Specimens examined:

HOLOTYPE: AUSTRALIA. North Queensland, Atherton Tablelands, Mt. Lewis, rainforest stream, on submerged wood, Aug 1992, *K.D. Hyde 1543*, **BRIP 22743.**

USA. Pennsylvania, Orangeville Fishing Creek, river, 41°7'42.744"N, 76°21'26.6034"W, 191m, on submerged woody debris, water 21.4 °C, pH 9.8, 80 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z43-1823;* Ricketts Glen State Forest, River, 41°20'1.356"N, 76°16'15.4194"W, 632m, on submerged woody debris, water 20.3 °C, pH 9, 632 µS, 19 Jul 2012, *Zelski S.E. and V.P. Hustad, Z43-1824.*Tennessee, GSMNP, riparian stream in Pigeon River watershed, 35°37’15.672”N, 83°25’25.3554”W, 1330m, on submerged woody debris, water 14.6 °C, pH 7.2, 17 Jul 2010, *Zelski S.E. and H.A. Raja, Z43-1686;* GSMNP, riparian stream in Pigeon River watershed, 35°37’57.108”N, 83°25’25.3554” W, 1144m, on submerged woody debris, water 15.1 °C, pH 7.5, 17 Jul 2010, *Zelski S.E. and H.A. Raja, Z43-1687.*

Known distribution: Australia, United States.

Comments: The holotype of *Submersisphaeria aquatica* is in poor condition, only yielding ascospores, and should be neotypified with a fresh specimen from Australia (**Fig 57 A-E**). The protologue (Hyde 1996) is the best material for comparison until that time. Fresh specimens obtained in this study are in good agreement with the holotype and molecular analyses place it in Annulatascaceae *sensu stricto*, and sister to *Longicollum biappendiculatum* (**Fig. 7**). *S. aquatica* has dematiaceous ascospores, a feature shared only with *Paoayensis lignicola* in Annulatascaceae *sensu stricto*, though the latter species has poor support. *S. rattanicola* is shown to be in a separate linage in this study, and thus the combination of a J- ascus apical ring and brown uniseptate ascospores needs to be supplemented with molecular data in order to determine generic and familial affiliation for the remainder of *Submersisphaeria* species.

Fig. 57 A-I. *Submersisphaeria aquatica* from the holotype **BRIP 22743**. A. Herbarium packet (plastic box). B. Contents of herbarium packet. C. Herbarium packet edge on. D. Herbarium packet label. E. Empty ascomata on dried wood. F. Intact ascomata on dried wood. G. Peridium. H-I. Ascospores. Bars: E-F = 500 µm; G-I = 20 µm.

Accession No.: BRIP 22743 a
Duplicates:
Other Taxa:
Genus: Submersisphaeria **Species:** aquatica **Infraspecies:**
Authority: K.D. Hyde
Name Comments:
Classification: Fungi, Ascomycota, Sordariomycetes, Incertae sedis, Annulatascaceae
Determiner: Hyde, K.D.
Host Genus: **Host Species:** **Host Infraspecies:**
Host Authority: **Cultivar:**
Host Family: **Common Name:**
Host Organ: **Host Symptom:**
Host Misc.:
Collector(s): Hyde, K.D. **Coll. Date:** Aug 1992 **Coll. No.:** KDH 1543
Locality: Mount Lewis, near
Place: Julatten **State:** QLD **Country:** Australia
Latitude: 16 35 S **Longitude:** 145 17 E
Specimen Type: Dried specimen
Living Culture:
Comments: On submerged wood in rainforest stream. Boxed specimen. Nova Hedwigia 62: 172, 1996.

Fig. 57.

Fig. 58 A-L. *Submersisphaeria aquatica* from Z43-1823. A. Gregarious ascomatal necks protruding from substrate. B. Neck protruding from substrate with mass of ascospores adhering to the outer surface. C. Neck illustrating periphyses. D. Surface texture of neck. E. Ascus. F. Young and mature asci. G. Annulus in young ascus. H-J. Apices of mature asci illustrating relatively massive apical rings. K. Ascospore with multiple guttules. L. Ascospore exhibiting two large guttules in each ascospore chamber. Bars: A = 1 mm; B = 500 µm; C-G = 20 µm; H-L = 10 µm.

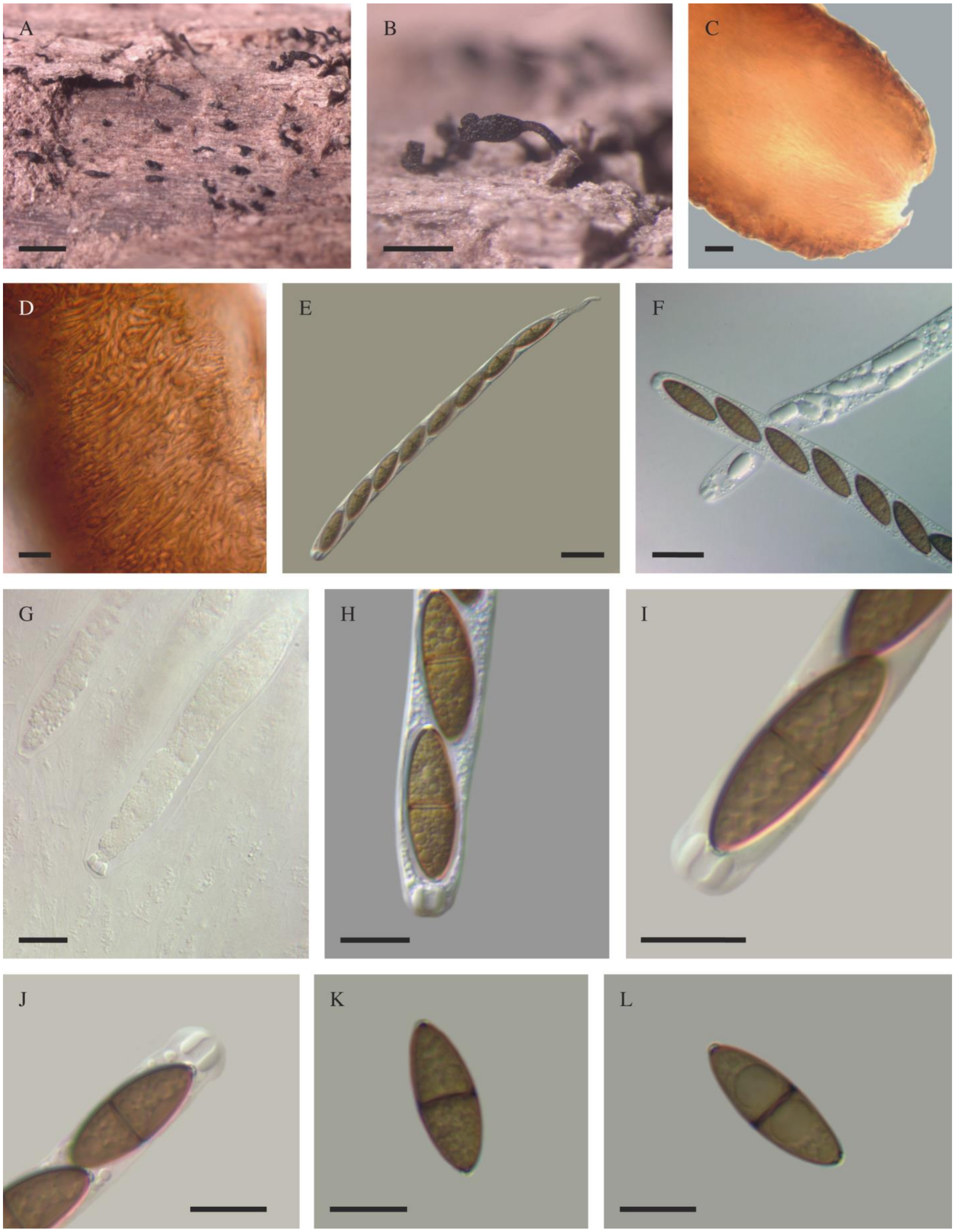


Fig. 58.

Submersisphaeria bambusicola D.Q. Zhou & K.D. Hyde, Fungal Diversity 4: 182. 2000. **Fig. 59 A-J.** MycoBank 464759.

Ascomata brown, immersed, perpendicular to surface, solitary, 420–580 µm wide × 350–540 µm high (x̅ = 500 × 445 µm, w:h = 21.1), globose to subglobose (**Fig. 59 C**). **Ascomal wall** brown outer layer and inner hyaline layer, 50–60 µm thick. **Neck** hyaline, 210 µm long × 60 µm wide, periphysate. **Paraphyses** hyaline, 4–8 µm wide at base, tapering toward apex, septate, filamentous. **Asci** unitunicate, cylindrical, 8–spored, 235–290 µm long × 10–12 µm wide (x̅ = 262.5 × 11 µm, l:w = 24) (**Fig. 59 D**). **Ascus base** pedicellate, long, tapering, 50 µm from *pars sporifera* (**Fig. 59 G**). **Annulus** J-, refractive, bipartite, 5 µm long × 5 µm wide, (l:w = 1) (**Fig. 59 E,F**). **Ascospores** dark brown, ellipsoid to fusiform, aseptate, uniseriate to overlapping uniseriate, 28–36 µm long × 6–8 µm wide (x̅ = 32 × 7 µm, l:w = 4.6), with one large lipid guttule, with hyaline germ pores, without appendages, with bipolar thin sheath (**Fig. 59 H-J**) (Zhou and Hyde 2000).

Specimens examined:

HOLOTYPE: HONG KONG. Hong Kong Island, Lung Fu Shan, on dead culm of *Arundinaria hindsii*, 19 Jul 1998, *Dequn Zhou, HKU(M) 9045*, **IFRD 8860.**

Known distribution: Hong Kong.

Comments: The holotype of *Submersisphaeria bambusicola* was in good condition and one permanent slide was prepared, examined, and returned to IFRD. (**Fig. 59 A,B**). Fresh collections of *S. bambusicola* were not obtained in this study, and thus the holotype and protologue were used for morphological study. *S. bambusicola* differs from the type of the genus, *S. aquatica* in having long-pedicellate asci and unicellular ascospores. *S. aquatica* belongs to Annulatascaceae *sensu stricto* based on molecular analyses, though *S. rattanicola* does not in this study. This polyphyly combined with these morphological

Fig. 59 A-J. *Submersisphaeria bambusicola* from the holotype **IFRD 8860**. A. Herbarium packet. B. Dried bamboo (*Arundinaria hindsii*). C. Ostiole on surface of bamboo surrounded by ascospores. D. Ascus. E-F. Ascus apices. G. Ascus base. H-J. Ascospores. Bars: C = 200 µm; D-J = 20 µm.


A
International Fungal Research & Development Centre
Herbarium IFRD
IFRD Code
Fungi Name
Host
Collection Site
HONG KONG, Hong Kong Island, Lung Fu Shan
Collection Date
1998-7-19
Collector
Remarks
Holotype
中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺
电话：3860026 3860022
邮政编码：650224
B
C
D
E
F
G
H
I
J

Fig. 59.

differences, indicate that a molecular evaluation of the phylogenetic placement of *S. bambusicola* is warranted. Zhou and Hyde (2000) noted that cultures were not obtained as ascospores failed to germinate. Collection of fresh specimens from Hong Kong should be made for the purpose of obtaining DNA.

Submersisphaeria palmae Pinnoi, Sydowia 56(1): 73. 2004. MycoBank 488198.

Ascomata dark brown, coriaceous, immersed, perpendicular to surface, clustered, 200–300 µm wide, globose to subglobose. **Ascomal wall** dark brown. **Neck** black. **Paraphyses** hyaline, 2.5–3.75 µm wide at base, tapering toward apex, septate, filamentous. **Asci** unitunicate, cylindrical, 8–spored, 100–127.5 µm long × 6.25–8.75 µm wide ($\bar{x}$ = 113.75 × 7.5 µm, l:w = 15.2). **Ascus base** pedicellate, tapering to club-like base, 25 µm from *pars sporifera*. **Annulus** J-, refractive, 1–2 µm long × 4–5 µm wide ($\bar{x}$ = 1.5 × 4.5 µm, l:w = .33). **Ascospores** olivaceous-brown, ellipsoidal, aseptate, uniseriate, 17.5–22.5 µm long × 5–7.5 µm wide ($\bar{x}$ = 20 × 6.25 µm, l:w = 3.2), guttulate, with bipolar pads as appendages, without a sheath (Pinnoi et al. 2004).

HOLOTYPE: THAILAND. Narathiwat, Sirindhorn Peat Swamp Forest, on submerged rachis of *Eleiodoxa conferta*, 13 Feb 2002*, A. Pinnoi*, **BBH Aom 152.**

Reports:

THAILAND. Narathiwat, Sirindhorn Peat Swamp Forest, on submerged rachis of *Eleiodoxa conferta*, 12 May 2001, *A. Pinnoi*, *BBH Aom 42; ibid.* on submerged rachis of *Nenga pumila,* 12 Feb 2002, *A. Pinnoi, BBH Nen 27; ibid.* on submerged trunk of *Licuala longecalycata,* 22 Jun 2001, *U. Pinraun, BBH Wah 72; ibid.* on submerged petiole of *Licuala longecalycata,* 22 Jun 2001, *U. Pinraun, BBH Wah 101; ibid.* on submerged trunk of *Licuala longecalycata,* 26 Sep 2001, U. Pinraun, *BBH Wah 125.*

Known distribution: Thailand.

Comments: The holotype of *Submersisphaeria palmae* was unavailable for examination and fresh specimens were not obtained in this study, thus the protologue was used for morphological study. *S. palmae* possesses bipolar pad-like appendages, a feature not found in the type of the genus, *S. aquatica*. *S. palmae* also has smaller ascospores which are aseptate vs. uniseptate in *S. aquatica*. *S. aquatica* is placed in molecular analyses in Annulatascaceae *sensu stricto*, though *S. rattanicola* does not in this study. This polyphyly of *Submersisphaeria* species combined with the morphological differences calls into question the relationship of *S. palmae* with the genus, and the species should be recollected, neotypified, and evaluated molecularly.

Submersisphaeria rattanicola J. Fröhl. & K.D. Hyde, Palm Microfungi: 82. 2000. **Fig. 60 A-J, Fig. 61 A-E.** MycoBank 482752.

Ascomata brown, immersed, clustered, 104–164 µm wide × 94–112 µm high (x̄ = 134 × 103 µm, w:h = 1.3), spherical (**Fig. 60 C**). **Ascomal wall** dark brown, 8–21 µm thick, 4–9 layers of dark brown flattened cells, of *textura angularis* in surface view. **Neck** periphysate. **Paraphyses** hyaline, to 8 µm wide at base, tapering to 2–4 µm at apex, septate. **Asci** unitunicate, long cylindrical, 8–spored, 123.8–154.5 µm long × 7–8.5 µm wide (x̄ = 139.2 × 7.8 µm, l:w = 18) (**Fig. 60 D, Fig. 61 A**). **Ascus base** pedicellate, 15 µm from *pars sporifera* (**Fig. 60 E, Fig. 61 B**). **Annulus** J-, refractive, 2–2.75 µm long × 3.25–4.75 µm wide (x̄ = 2.4 × 4 µm, l:w = .6) (**Fig 60 F,G**). **Ascospores** brown, fusiform to ellipsoid, 1–septate, slightly constricted at each septum, uniseriate, 14.3–20.8 µm long × 5–6.8 µm wide (x̄ = 17.6 × 5.9 µm, l:w = 3), multiguttulate, with hyaline apical germ pores, without appendages or sheath (**Fig. 60 H-I, Fig. 61 C-E**) (Fröhlich and Hyde 2000).

Specimens examined:

HOLOTYPE: AUSTRALIA. North Queensland, Palmerston, Palmerston National Park, on dead rattan of *Calamus radicalis*, Mar 1994, *J. Fröhlich, JF 306,* **IFRD 9035.**

PERU. Cusco, Camanti, Quincemil, stream at Quincemil Trail 1, trailhead 13°14'22.5594"S, 70°46'12.6114"W, 688m, on submerged woody debris, water 21.2 °C, pH 6.8, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0292-3;* Quincemil, 1st stream along Quincemil Trail 1, trailhead 13°14'22.5594"S, 70°46'12.6114"W, 688m, on submerged woody debris, water 19.7 °C, pH 5.8, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0292-1;* Quincemil, 3rd stream along Quincemil Trail 1, trailhead 13°14'22.5594"S, 70°46'12.6114"W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0292-2.*

Known distribution: Australia, Perú.

Comments: The holotype of *Submersisphaeria rattanicola* is in good condition and the dried herbarium material consisted of a rattan of *Calamus sp*., 30 cm long × 1 cm in diameter (**Fig. 60 A,B**). Three permanent slides were made, examined, and returned with the dried specimen. Paraphyses were not preserved in the holotype. The Peruvian samples are a good match to the protologue and the holotype material. Morphologically, *S. rattanicola* is most similar to *S. aquatica,* as noted in the protologue. However, ascospores are more fusoid in *S. rattanicola*. Combined three gene analyses place *S. rattanicola* distant to both *Submersisphaeria* and Annulatascaceae *sensu stricto* (**Fig. 7, Clade 3b**), and the species should be considered Sordariomycetes *incertae sedis* at present. As *S. rattanicola* is not related to the type of the genus, *S. aquatica*, a new combination, *Pseudosubmersisphaeria rattanicola*, is proposed (see Nomenclatural Changes).

Fig. 60 A-J. *Submersisphaeria rattanicola* from the holotype **IFRD 9035**. A. Herbarium packet. B. Dried rattan (*Calamus carotoides*). C. Dried substrate illustrating upraised areas of host tissue and areas where host tissue was removed to access ascomata. D. Ascus. E. Ascus base. F-G. Ascus apices. H-J. Ascospores. Bars: C = 1 mm; D-E, H = 20 µm; F-G, I-J = 10 µm.

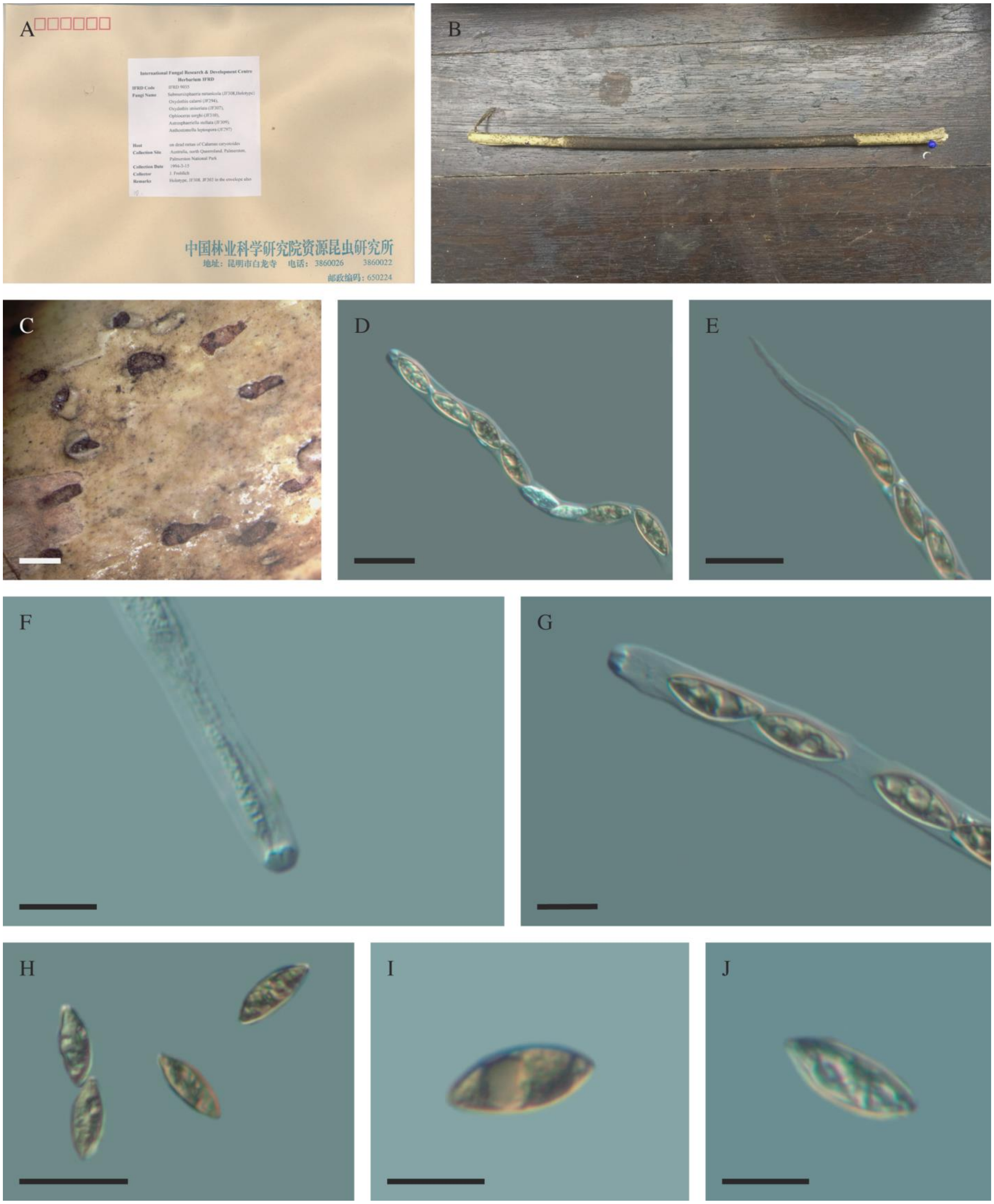

A
International Fungal Research & Development Centre
Herbarium IFRD
IFRD Code
Fungi Name
Host
Collection Site
Collection Date
Collector
Remarks
中国林业科学研究院资源昆虫研究所
地址：昆明市白龙寺　电话：3860026　3860022
邮政编码：650224
B
C
D
E
F
G
H
I
J

Fig. 60.

Fig. 61 A-E. *Submersisphaeria rattanicola* from specimen PE0292-2. A. Ascus apex. B. Ascus base. C-D. Ascospores. E. Ascospore under 100× magnification. Bars: A-D = 20 µm; E = 10 µm.

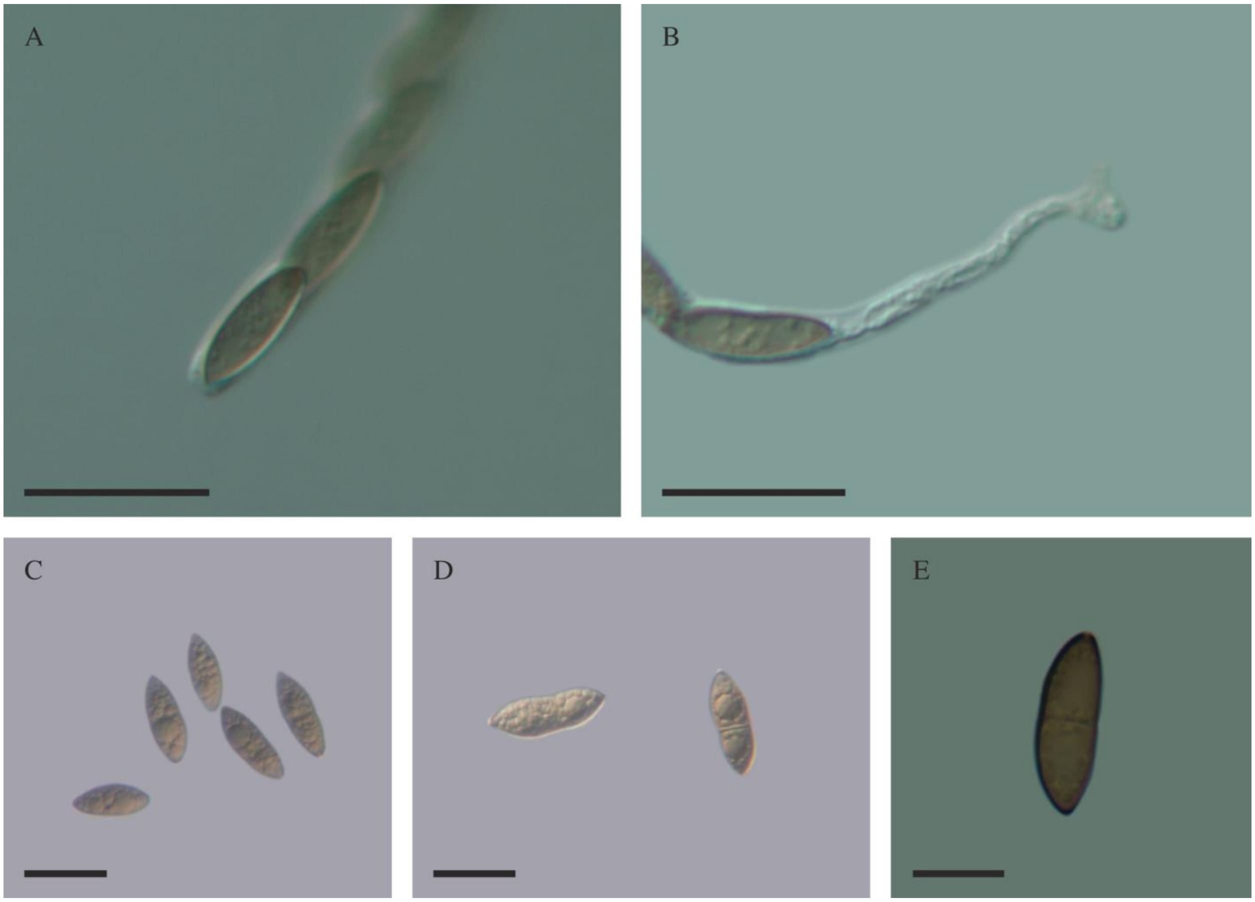


Fig. 61.

Submersisphaeria vasicola (Ellis & Everh.) Y.Z. Wang, Aptroot & K.D. Hyde, Revision of the genus *Amphisphaeria*: 62. 2004. **Fig. 62 A-G, Fig. 63 A-M.** MycoBank 370395.

≡ *Sphaeria aquatica* Ellis & Everh., Bulletin of the Torrey Botanical Club 11(4): 42. 1884. MycoBank 144608.

≡ *Amphisphaeria aquatica* (Ellis & Everh.) Berl. & Vogl., Sylloge Fungorum, Additamenta ad volumina 1-4: 125. 1886. MycoBank 189396.

≡ *Didymosphaeria aquatica* (Ellis & Everh.) Ellis & Everh., Catalog of Plants Found in New Jersey: 523. 1889. MycoBank 370395.

Ascomata brown, semi-immersed to erumpent, perpendicular to surface, solitary to gregarious, 250 µm wide, subglobose (**Fig. 62 E-G**)**. Ascomal wall** brown, several layers of brown compressed cells. **Neck** present. **Paraphyses** 3.5 µm wide at base, tapering toward apex, septate, flexuose (**Fig. 63 C,D**). **Asci** unitunicate, cylindrical, 8–spored, 140 µm long × 8 µm wide ($\bar{x}$ = 140 × 8 µm, l:w = 17.5) (**Fig. 63 A,B,E**). **Ascus base** pedicellate. **Annulus** J-, refractive (**Fig. 63 F-H**). **Ascospores** brown, cylindro-ellipsoidal, 1–septate, slightly constricted at each septum, uniseriate, 16–22 µm long × 6–7 µm wide ($\bar{x}$ = 19 × 6.5 µm, l:w = 3), each cell with lipid guttule, without appendages, with sheath (**Fig. 63 I-M**) (Wang et al. 2004).

Specimens examined:

HOLOTYPE: USA. New Jersey, Newfield, in cedar pail in constant use, Dec 1882, Ellis **NY, holotype of *Sphaeria aquatica*.**

Known distribution: USA.

Comments: The holotype of *Submersisphaeria vasicola* is in good condition and 2 permanent slides were made, examined, and returned to NY (**Fig. 63 A-D**). Originally described as *Sphaeria aquatica* (Ellis & Everhardt 1884), and ultimately transferred to *S. vasicola* (Wang et al. 2004), this species represents the

Fig. 62 A-G. *Submersisphaeria vasicola* from **NY holotype of *Sphaeria aquatica*** (barcode 00875084). A. Herbarium folder. B. Documentation. C. Documentation and dried specimen. D. Dried specimen. E-F. Ascomal remnants. G. Submersed ascomata. Bars: E-F = 500 µm; G = 1 mm.

Sphaeria aquatica E. & E.
Amphisphaeria
on bottom (inside) of a cedar
water pail in constant
use. Newfield N. J.
Dec. 1883 Spor. 15–20x6–7μ
asci 150x8–9μ

SPHÆRIA AQUATICA.—Perithecia scattered, membranaceous, .25^{mm} in diameter, partly buried in the wood, subglobose or a little elongated; ostiolum papilliform; asci linear, 150x8–6μ with abundant paraphyses; sporidia uniseriate (end to end), oblong, uniseptate and slightly constricted at the septum, straight or slightly curved, pale at first, with a single large nucleus in each cell, at length clear dark brown, 15–20x6–7μ.

Inside a cedar water-pail in constant use; on the bottom and around the lower part of the sides where it must have been almost constantly under water. Newfield N. J., Dec., 1883.

Ellis + Everhart: Bull. Torrey Club. 11: 42. 1884.

Sphaeria aquatica E & E
On bottom of cedar water pail in
the house - Water had been standing
in the pail almost continuously for
a year or more - pail in constant use
Newfield Dec.

Fig. 62.

Fig. 63 A-M. *Submersisphaeria vasicola* from **NY holotype of *Sphaeria aquatica*** (barcode 00875084). A-B. Asci. C-D. Paraphyses. E. Ascus bases. F-H. Ascus apices illustrating apical rings. I-J. Ascospores. K-M. Germinating ascospores. Bars: A-E, G, I-M = 20 µm; F,H = 10 µm.

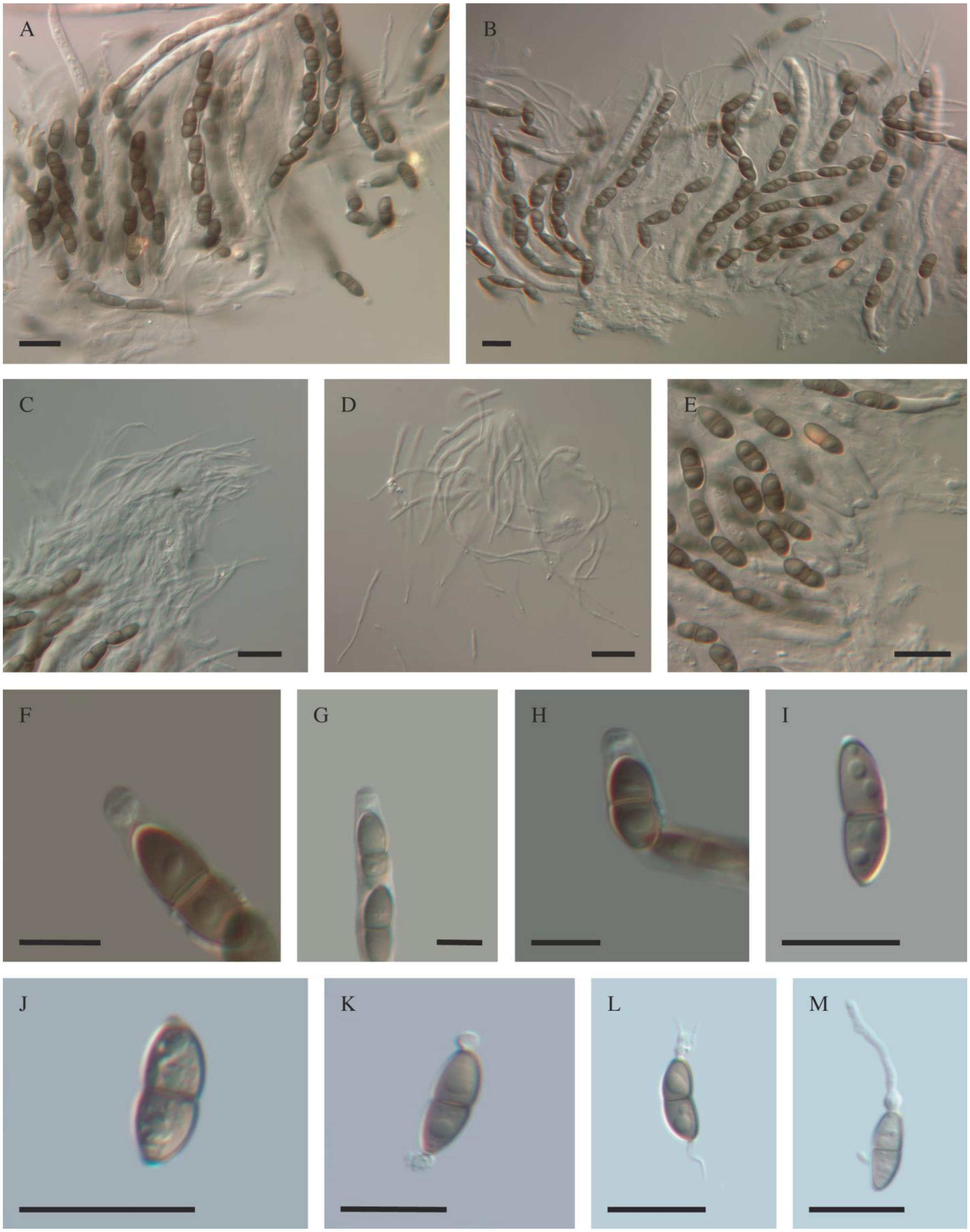


Fig. 63.

oldest described member of Annulatascaceae. The dried specimen yielded scattered, numerous, immersed, semi-immersed to erumpent, black, ostiolate, ascomata; numerous, cylindrical, unitunicate asci; and ascospores that were 17.8–23.2 µm long × 6.2–8.6 µm wide (x̅ = 20 × 7.5 µm, l:w = 2.7, n = 30). DNA extraction was not attempted due to the age of the material, thus the phylogenetic placement of this fungus in relation to *Submersisphaeria* and Annulatascaceae was not assessed with molecular data. Obtaining fresh material for this fungus may be tricky due to the rather unusual type location – inside a cedar pail in Newfield, New Jersey. Morphologically, the species fits well in *Submersisphaeria* and consequently in Annulatascaceae.

Teracosphaeria M. Réblová & K.A. Seifert, Mycological Research 111(3): 291. 2007. MycoBank 510202.

Type species: *Teracosphaeria petroica.*

Teracosphaeria petroica Réblová & Seifert, Mycological Research 111: 292. 2007. **Fig. 64 A-L.** MycoBank 510203.

Ascomata dark brown to black, glabrous, leathery to fragile, immersed, perpendicular to surface 450–600 µm wide × 400–550 µm high (x̅ = 525 × 475 µm, w:h = 1.1), globose to subglobose (**Fig. 64 C,D**). **Ascomal wall** outer wall of thin polyhedral cells, inner layer of hyaline elongated and compressed cells, 60–75 µm thick, of *textura angularis* to *textura prismatica* in surface view. **Neck black,** 600–900 µm long × 190–210 µm wide, of *textura prismatica,* periphysate. **Paraphyses** hyaline, 5–6 µm wide, tapering, septate (**Fig. 64 I**). **Asci** unitunicate, fusiform-clavate, 8–spored, 98–132 µm long × 12–17 µm wide (x̅ = 115 × 15.5 µm, l:w = 3) (**Fig. 64 E-G**). **Ascus base** pedicellate, ending in club-like base, 15 µm from *pars sporifera*. **Annulus** J-, refractive, 1.5 µm long × 4–5 µm wide (x̅ = 1.5 × 4.5 µm, l:w = .33)

(**Fig. 64 H**). **Ascospores** hyaline, ellipsoidal, 3–septate, not constricted at septa, uniseriate to biseriate, 15–20 µm long × 5–6 µm wide ($\bar{x}$ = 17.5 × 5.5 µm, l:w = 3.2), without appendages or sheath (**Fig. 64 J-L**) (Réblová and Seifert 2007).

Specimens examined:

HOLOTYPE: NEW ZEALAND. Westland, Lewis Pass, Victoria Forest Park, Riordan's Creek Valley, ca. 30km from Springs Junction, on decayed wood of *Nothofagus sp.,* 20 Feb 2003, *M. Réblová and K.A. Seifert, MR 2570/03,* **PDD 81436.**

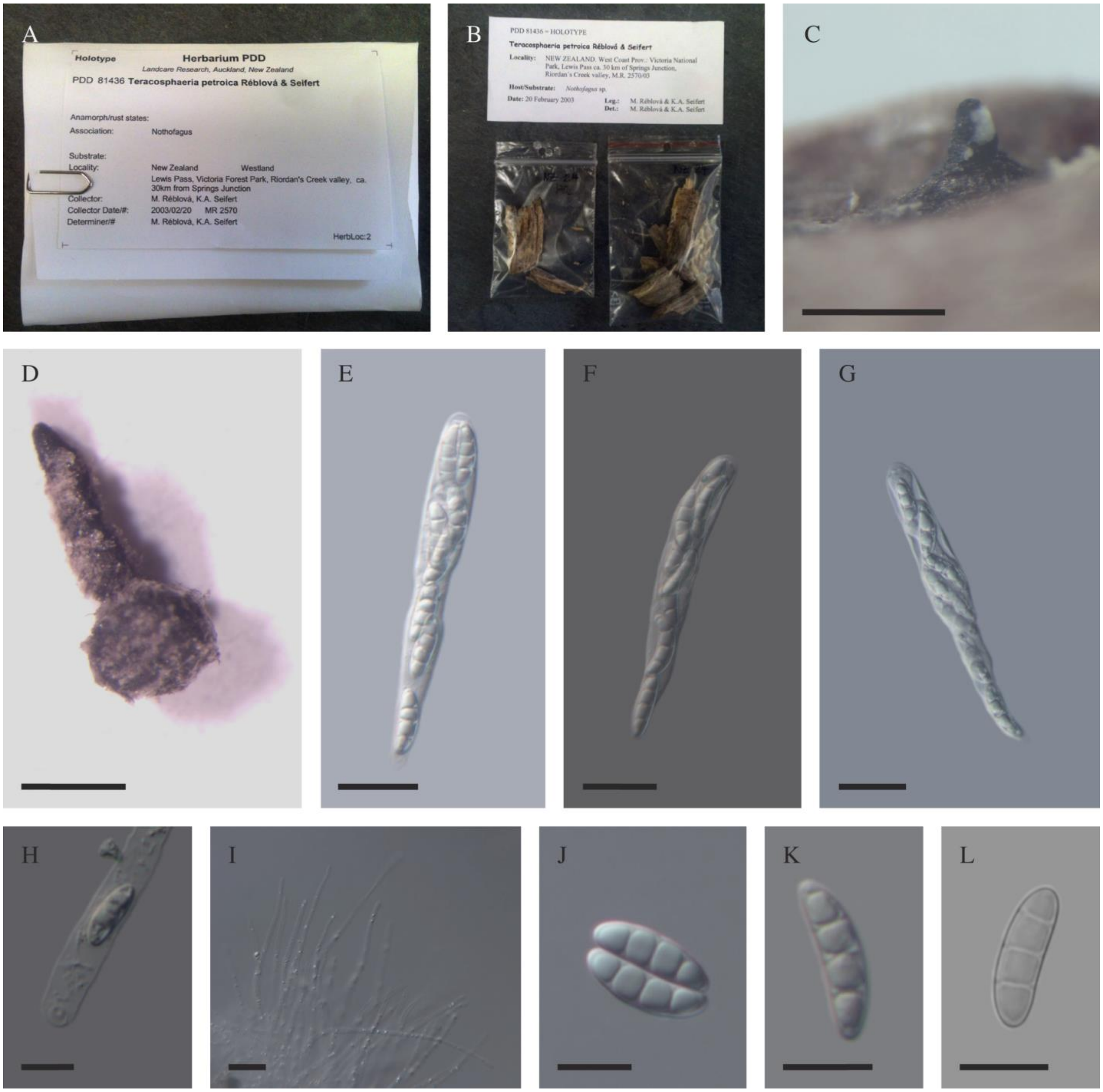
Comments: The holotype of *Teracosphaeria petroica* is in good condition and two slides made, examined, and returned with the holotype material (**Fig. 64 A,B**). Fresh collections of this fungus were not obtained in this study. 28S sequence analysis initially placed this species as a sister taxon to *Mirannulata samuelsii* (Réblová and Seifert 2007), but not related to Annulatascaceae. In the combined three gene analyses, *Te. petroica* is placed in a clade separating Diaporthales and Ophiostomatales, though not with *M. samuelsii* (**Fig. 7, Clade 1k**). Both taxa had been compared to Annulatascaceae due to the presence of relatively large refractive ascus apical rings. *Te. petroica* is best considered Sordariomycetes *incertae sedis*.

Torrentispora K.D. Hyde, W.H. Ho, E.B.G. Jones, K.M. Tsui & S.W. Wong, Mycological Research 104 (11): 1399. 2000. MycoBank 28453.

Type species: *Torrentispora fibrosa.*

Torrentispora crassiparietis S.C. Fryar & K.D. Hyde, Cryptogamie Mycologie 25 (3): 255. 2004. **Fig. 65 A-F.** MycoBank 368913.

Fig. 64 A-L. *Teracosphaeria petroica* from the holotype **PDD 81436**. A. Herbarium packet. B. Packet contents (2 small bags containing dried substrate). C. Neck protruding from substrate. D. Ascoma dissected from substrate. E-G. Asci. H. Annulus seen end on. I. Long tapering septate paraphyses. J-L. Ascospores. Bars: C-D = 500 µm; E-G, I = 20 µm; H, J-L = 10 µm.



Holotype Herbarium PDD
Landcare Research, Auckland, New Zealand
PDD 81436 Teracosphaeria petroica Réblová & Seifert

Anamorph/rust states:
Association: Nothofagus

Substrate:
Locality: New Zealand Westland
Lewis Pass, Victoria Forest Park, Riordan's Creek valley, ca. 30km from Springs Junction
Collector: M. Réblová, K.A. Seifert
Collector Date/#: 2003/02/20 MR 2570
Determiner/# M. Réblová, K.A. Seifert
HerbLoc:2

PDD 81436 = HOLOTYPE
Teracosphaeria petroica Réblová & Seifert
Locality: NEW ZEALAND. West Coast Prov.: Victoria National Park, Lewis Pass ca. 30 km of Springs Junction, Riordan's Creek valley, M.R. 2570/03
Host/Substrate: *Nothofagus* sp.
Date: 20 February 2003
Leg.: M. Réblová & K.A. Seifert
Det.: M. Réblová & K.A. Seifert

Fig. 64.

Ascomata black, coriaceous, immersed, perpendicular to surface, solitary, 220–315 µm wide, globose to subglobose. **Ascomal wall** black. **Neck** black, 200 µm long × 50 µm wide, periphysate. **Paraphyses** hyaline, 3 µm wide at base, tapering toward apex, septate (**Fig. 65 D**). **Asci** unitunicate, long cylindrical, 8–spored, 212–300 µm long × 10.5–12 µm wide (x̅ = 256 × 11.3 µm, l:w = 22.8) (**Fig. 65 A,B**). **Ascus base** pedicellate broadly tapering to club-like base, 25 µm from *pars sporifera*. **Annulus** J-, refractive, 7–8 µm long × 7–9 µm wide (x̅ = 7.5 × 8 µm, l:w = .94) (**Fig. 65 A-C**). **Ascospores** hyaline, thick walled, aseptate, ellipsoidal to fusiform, uniseriate, 31–42.5 µm long × 7.5–10 µm wide (x̅ = 36.75 × 8.75 µm, l:w = 4.2), multiguttulate, without appendages, with a thin fibrillar sheath (**Fig. 65 E,F**) (Fryar and Hyde 2004).

HOLOTYPE: BRUNEI. Tutong District, Sungai Kelakas, Site 3, 4°49’40.58”N, 114°41’25.16”E, 11m, on submerged rotting wood, water 29 °C, pH 6.3, 27 Oct 1998, *Fryar S.C.*, **HKU(M) 15667.**

Specimens examined:

PERU. Cusco, Camanti, Quincemil, stream at Quincemil trail 1, on submerged herbaceous debris, 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 21.2 °C, pH 7.1, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0068-4;* Rio Caliente, 1km south of Quincemil, 13°13’20.87”S, 70°44’30.07”W, 626m, water 25 °C, pH 7.2, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0068-5;* Stream with red algae along Quincemil trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, water 21.8 °C, pH 7.2, on submerged woody debris, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0068-6*; Madre de Dios, palm swamp (aguajal) off the Interoceanic Highway near Puerto Maldonado, trail head 12°42’48.0954”S, 69°28’11.28”W, 239m, water 23.3 °C, pH 5.9, 20 May 2010, *Zelski S.E. and H.A. Raja, PE0068-1;* CICRA, Rio Amigos, 12°34’13.008”S, 70°41’14.7714”W, 218m, on submerged woody debris, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0068-3.*

Known distribution: Brazil, Costa Rica, Brunei, Perú.

Comments: The holotype of *Torrentispora crassiparietis* was unavailable for examination. Hyde erected the genus *Torrentispora* to accommodate an *Annulatascus*-like species of freshwater ascomycetes, *T. fibrosa*, with thick walled ascospores and a large apical apparatus (Hyde et al. 2000). Morphologically, *T. crassiparietis* is similar to *T. fibrosa* in having thick walled ascospores but it lacks a fibrillar sheath. Peruvian specimens (PE0068) examined in this study agree with the protologue of *T. crassiparietis* in gross morphology as well as ascospore morphology. Ascospores are uniseriate, thick-walled, ellipsoid-fusiform, flattened on one side, unicellular, and hyaline as in Fryar and Hyde (2004). Dimensions of PE0068 ascospores differ slightly compared to the type (32.5–48.4 µm long × 7.6–12.1 µm wide (x̅ = 40.7 × 10.1 µm, l:w = 4, n = 90) vs. 31–42.5 µm long × 7.5–10 µm wide)). Asci and annuli were also comparable in measurements. *T. crassiparietis* is not related to *T. fibrosa*, the type of the genus, nor to the Annulatascaceae *sensu stricto* (**Fig. 7, Clade 5**). Therefor a new combination, *Pseudotorrentispora crassiparietis,* is proposed (see Nomenclatural Changes) and the species should be considered Sordariomycetes *incertae sedis*.

Torrentispora fibrosa K.D. Hyde, W.H. Ho, E.B.G. Jones, K.M. Tsui & S.W. Wong, Mycological Research 104(11): 1139. 2000. **Fig. 66 A-H.** MycoBank 466188.

Ascomata black, coriaceous, immersed to superficial, perpendicular to surface, solitary, 135–255 µm wide, globose to subglobose. **Ascomal** hyaline and brown, 21.5 µm thick, wall outer layer of dark brown angular cells and inner layer of hyaline compressed cells (**Fig. 66 A**). **Neck** black, long, periphysate. **Paraphyses** hyaline, 5.5 µm wide at base, tapering toward apex, septate, constricted at septa. **Asci** unitunicate, long cylindrical, 8–spored, 154–254 µm long × 6–9 µm wide (x̅ = 204 × 7.5 µm, l:w = 27.2) (**Fig. 66 B**). **Ascus base** pedicellate, cylindrical, rounded at end, 30 µm from *pars sporifera* (**Fig. 66 E**). **Annulus** J-, refractive, 1.75–2.25 µm long × 2.8–3.75 µm wide (x̅ = 1.9 × 3.1 µm, l:w = .56) (**Fig. 66 C,D**). **Ascospores** hyaline, thick walled, ovoid to fusiform, aseptate, uniseriate, 13.5–19.5 µm long × 5–7

Fig. 65 A-F. *Torrentispora crassiparietis* from specimen PE0068. A-B. Asci. C. Annulus ring end on. D. Paraphyses. E-F. Ascospores. Bars: A-F = 20 µm.

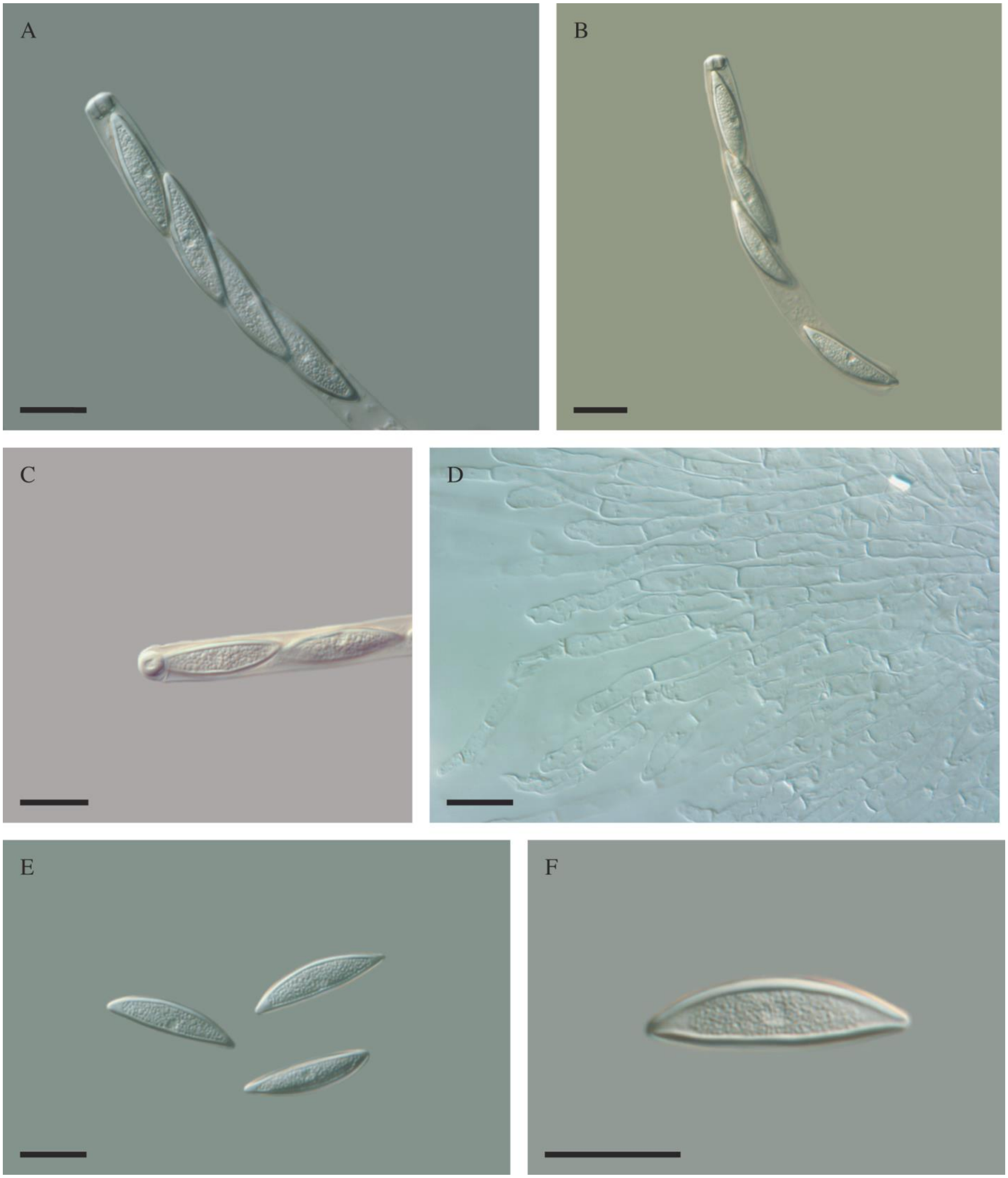


Fig. 65.

µm wide (x̅ = 16.5 × 6 µm, l:w = 2.75), multiguttulate, without appendages, with a narrow fibrillar sheath (**Fig. 66 F-H**) (Hyde et al. 2000).

HOLOTYPE: HONG KONG. New Territories, Tai Po Kau Forest Reserve, Tai Po Kau Forest Stream, on submersed decomposing wood, 10 Dec 1995, *W.H. Ho*, **HKU(M) 4519.**

Specimens examined:

PERU. Cusco, Camanti, Quincemil, stream crossing the Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 737m, on submerged woody debris, water 22 °C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0038-3;* Stream at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9274”W, 688m, on submerged woody debris, water 22.2 °C, pH 7.1, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-5;* Quincemil Trail 1, semi-aquatic habitat, 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0038-6;* Stream at Quincemil Trail 1, 13°13’58.25945”S, 70°46’37.7754”W, 675m, on submerged woody debris, water 21.2 °C, pH 7.1, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0038-4;* Rio Caliente, 1km south of Quincemil, 13°13’20.87”S, 70°44’30.07”W, 626m, on submerged woody debris, water 25 °C, pH 7.2, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0038-2;* Rio Frio (Yana Mayo), 13°13’20.87”S, 70°44’30.07”W, 626m, on submerged woody debris, water 25.3 °C, pH 8, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0038-14;* 4th stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.4 °C, pH 7.8, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-8;* 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-13;* 1st stream at Quincemil Trail 3, trailhead 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.7 °C, pH 6.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-10;* Stream at Quincemil Trail 3, trailhead 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.5 °C, pH 7, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-12;* Stream flowing into river backwater at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 21.2 °C, pH 7.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-7;* Madre de Dios, palm swamp (aguajal) near the Interoceanic Highway near Puerto Maldonado, trail head 12°42’48.0954”S, 69°28’11.28”W, 239m, water 23.3 °C, pH 5.9, 20 May 2010, *Zelski S.E. and H.A. Raja, PE0038-2;* CICRA, Stream at intersection of Trail 14/15, 12°34’17.08”S, 70°05’47.60”W, 244m, on submerged woody debris, water 23.5 °C, pH 6.7, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0038-11;* 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 21.3 °C, pH 6, on submerged woody debris, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-13;* River backwater at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 22 °C, pH 7.1, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0038-5.* THAILAND. Chiang Mai, Pa Pha Deng Temple stream, 19°06’37.61”N, 98°44’17.20”E, 1020m, on submerged woody debris, water 22.8 °C, pH 6.8, 27 Jun 2012, *Zelski S.E., TH0038-1.*

Reports:

HONG KONG. New Territories, Tai Po, Lam Tsuen River, on submerged decaying wood, 1 Oct 1997, *K.M. Tsui (HKU(M) 8093)*; *ibid. (HKU(M) 8107);* Tai Po Kau Forest Reserve, Tai Po Kau Forest Stream, on submerged decaying wood, 27 Jun 1996, *K.D. Hyde (HKU(M) 4534); ibid. (HKU(M) 4538); ibid.* 21 Sep 1996, *W.H. Ho* & *K.M. Tsui (HKU(M) 4818); ibid.*, 20 Dec 1996, *W.H. Ho* & *S.Y. Ho (HKU(M) 5919); ibid. (HKU(M) 5932); ibid.* 28 Jun 1997, *W.H. Ho (HKU(M) 6076); ibid. (HKU(M) 6084); ibid. (HKU(M) 6086); ibid. (HKU(M) 6089); ibid. (HKU(M) 6099); ibid. (HKU(M) 6101); ibid. (HKU(M) 6115); ibid. (HKU(M) 6118); ibid.* 28 Sep 1997, *W.H. Ho (HKU(M) 6167); ibid. (HKU(M) 6178);* Shing Mun Reservoir, on submerged wood, 14 Jan 1998, *K.M. Tsui (HKU(M) 8129); ibid. (HKU(M) 8197).*

Comments: The holotype of *Torrentispora fibrosa* was unavailable for examination. Hyde et al. (2000) erected the genus based on this species, which was found on submerged wood in streams in Hong Kong.

Fig. 66 A-H. *Torrentispora fibrosa* from specimen PE0038-2. A. Peridium. B. Ascus. C-D. Ascus apices. E. Ascus base. F-H. Ascospores. Bars: A = 40 μm; B-C,E-G = 20 μm; D,H = 10 μm.

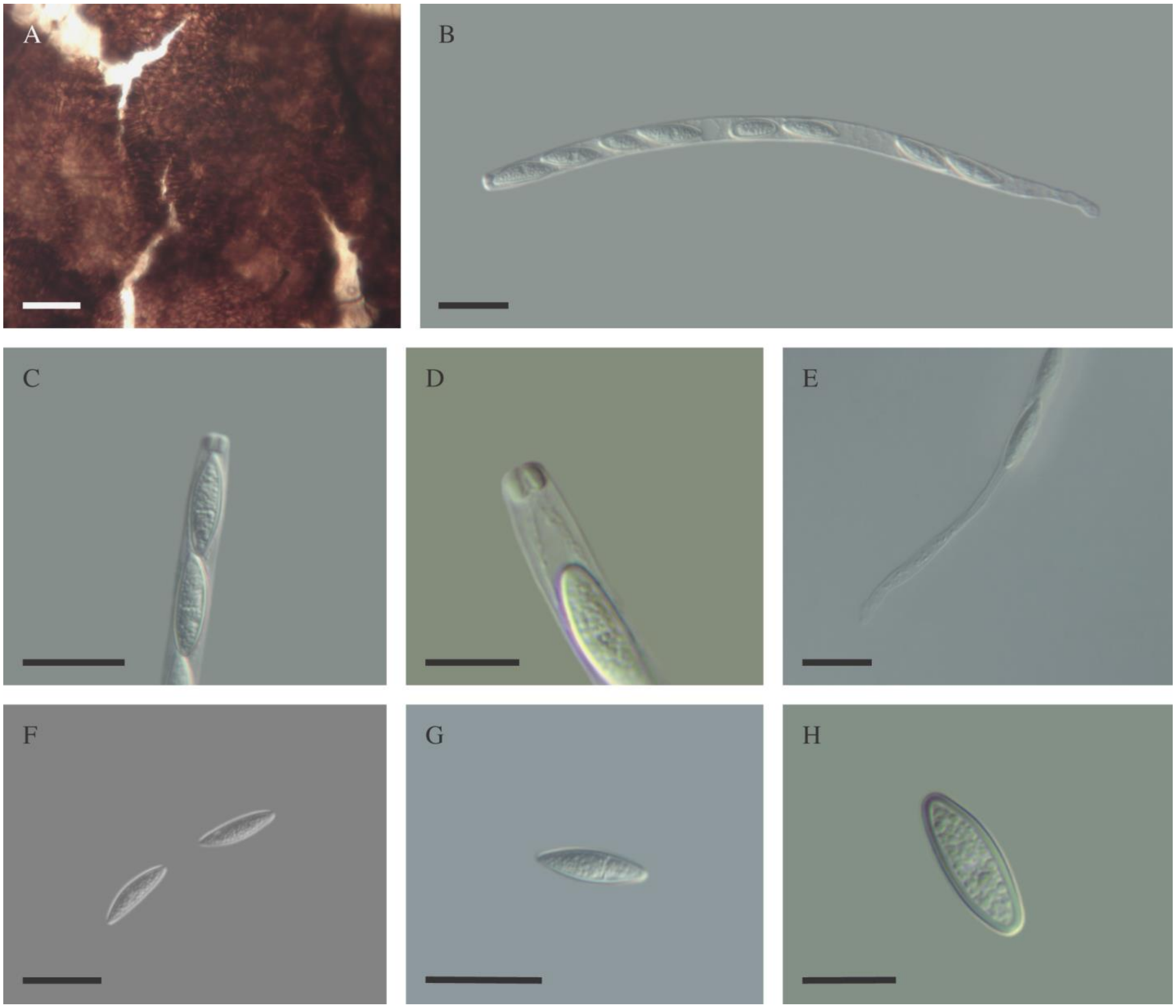


Fig. 66.

It was included in Annulatascaceae due to the presence of a relatively massive refractive non-amyloid ascus apical ring. In this study, ascomata were used for extraction as ascospores did not germinate. *T.fibrosa* nests in a clade well supported by Bayesian but not likelihood analyses (**Fig. 7, Clade 1g**). It is not related to Annulatascaceae *sensu stricto*. *T. fibrosa* grouped with *Aquaticola miniguttulata, Annulatascus biatriisporus,* a new *Annulatascus*-like species (PE0035), a new *Aquaticola*-like species (AK-1838), *Fusoidispora aquatica, Lentomitella* species, and a new *Aquaticola-like* species (PE0316) (**Fig. 7, Clade 1f**). *Torrentispora fibrosa* and these taxa should be considered Sordariomycetes *incertae sedis*.

Torrentispora fusiformis Fryar & K.D. Hyde, Cryptogamie Mycologie 25: 256. 2004. **Fig. 67 A-I.** MycoBank 368912.

Ascomata black, coriaceous, immersed, perpendicular to surface, solitary, 220–315 µm wide, globose to subglobose. **Ascomal wall** black, 16 µm thick, of *textura prismatica* in surface view (**Fig. 67 A**). **Neck** black, long, periphysate. **Paraphyses** hyaline, 4 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, long cylindrical, 8–spored, 200–230 µm long × 9–10 µm wide ($\bar{x}$ = 215 × 9.5 µm, l:w = 22.6) (**Fig. 67 B**). **Ascus base** pedicellate, 20 µm from *pars sporifera* (**Fig. 67 C). Annulus** J-, refractive, 4 µm long × 6 µm wide (l:w = .67) (**Fig. 67 D-G**). **Ascospores** hyaline, thick walled, fusiform, aseptate, uniseriate, 24–32.5 µm long × 6–9 µm wide ($\bar{x}$ = 28.25 × 7.5 µm, l:w = 4.8), multiguttulate, without appendages or sheath (**Fig. 67 H,I**) (Fryar and Hyde 2004).

HOLOTYPE: BRUNEI. Tutong District, Sungai Kelakas, a tributary of the Tutong River, Site 1, 4°48’01.38”N, 114°30’38.87”E, 8m, on submerged rotting wood, water 26.1-30 °C, pH 4.8-5.6, 27 Oct 1998, *S.C. Fryar*, **HKU(M) 16048.**

Specimens examined:

PERU. Cusco, Camanti, Quincemil, stream crossing the Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 737m, on submerged woody debris, water 22 °C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0262-9;* River at end of Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 19.6 °C, pH 8.3, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0262-5;* 2nd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 20 °C, pH 5.8, 3 Oct 2010, *Zelski S.E. and H.A. Raja, PE0262-8;* River at Quincemil Trail 2, 13°13’31.0434”S, 70°45’10.6194”W, 653m, on submerged woody debris, water 24.9 °C, pH 7.2, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0262-4;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 20.5 °C, pH 7.2, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0262-6;* 2nd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0262-1;* 1st stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.7 °C, pH 6.8, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0262-12;* River backwater at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9279”W, 757m, on submerged decorticated wood, water 22 °C, pH 7.1, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0262-11;* Madre de Dios, CICRA, Stream at Trail 10, 12°37’48.95”S, 70°05’23.69”W, 287m, on submerged woody debris, water 22.3 °C, pH 5.6, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0262-7;* CICRA, Creek at Trail 23, 12°33’31.03”S, 70°05’56.96”W, 280m, on submerged decorticated wood, water 22.2 °C, pH 6.4, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0262-3;* CICRA, Pozo Don Pedro, palm swamp (aguajal) at end of Trail 17, 12°33’34.27”S, 70°06’38”W, 243m, on submerged woody debris, 22 May 2010, *Zelski S.E. and H. A. Raja, PE0262-2.*

Reports:

BRUNEI. Sungai Kelakas, a tributary of the Tutong River, Site 4, 4°48’19.73”N, 114°40’37.76”E, 13m, on submerged rotting wood, water 26.6-29 °C, pH 5.4, *S.C. Fryar, HKU(M) 15575; ibid. HKU(M) 15609; ibid. HKU(M) 15615*.

Known distribution: Brunei, Perú.

Comments: The holotype of *Torrentispora fusiformis* was unavailable for examination. The protologue illustrates a thick walled ascospore which is a feature not present in Annulatascaceae *sensu stricto*. Though collected in this study, DNA was not obtained as ascospores did not germinate and extractions from fruit bodies were contaminated with other fungal material. As two other *Torrentispora* species, *T. crassiparietis* and *T. fibrosa*, were shown to be in separate lineages in this study, *T. fusiformis* should be considered Sordariomycetes *incertae sedis* and needs to be evaluated molecularly for its phylogenetic placement.

Torrentispora pilosa Shearer & F.R. Barbosa, Mycologia 105(2): 339. 2013. MycoBank 564458.

Ascomata black, hairy, coriaceous, semi-immersed, perpendicular to surface, scattered, 247–450 µm wide × 320–400 µm long (x̅ = 348.5 × 360 µm, w:h = .97). **Ascomal wall** brown, of *textura prismatica* in surface view. **Neck** black, hairy, 44–770 µm long × 55–180 µm wide, hairy. **Paraphyses** hyaline, 4–6 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 164–204 µm long × 7–8 µm wide (x̅ = 184 × 7.5 µm, l:w = 24.5). **Ascus base** pedicellate, tapering at the base, 25 µm from *pars sporifera*. **Annulus** J-, refractive, 3–4 µm long × 4–5 µm wide (x̅ = 3.5 × 4.5 µm, l:w = .78). **Ascospores** hyaline, ellipsoidal to fusiform, 0–2 septate, uniseriate, 21–30 µm long × 7–8 µm wide (x̅ = 25.5 × 7.5 µm, l:w = 3.4), without appendages or sheath (Barbosa et al. 2013).

Fig. 67 A-I. *Torrentispora fusiformis* from specimen PE0262-9. A. Peridium. B. Ascus. C. Ascus base. D-G. Ascus apices. H-I. Ascospores. Bars: A-B = 40 µm; C-F = 20 µm; G-H = 10 µm.

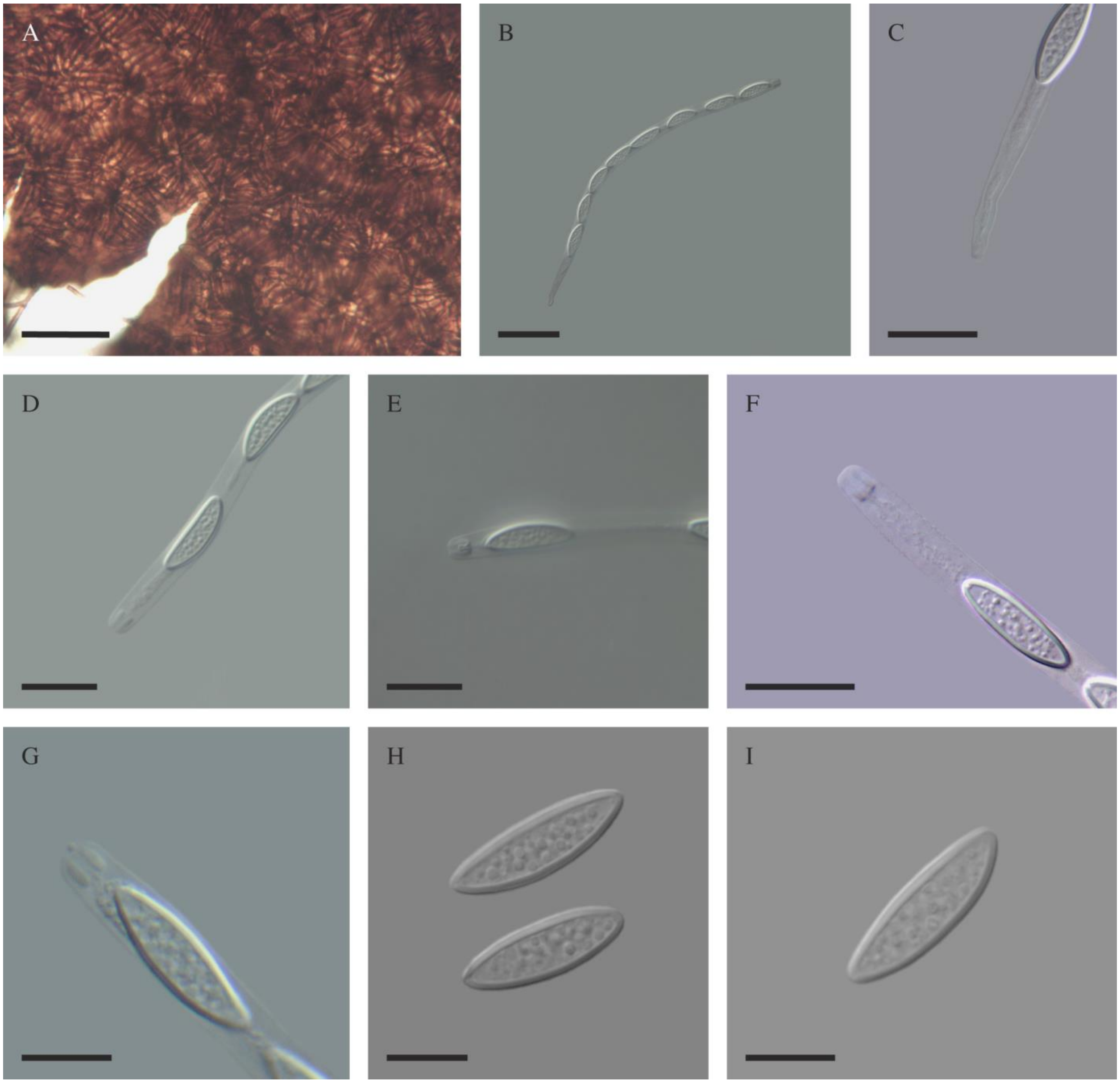


Fig. 67.

HOLOTYPE: COSTA RICA. Heredia, La Selva Biological Station, 10°25’48”N, 84°1’32”W, on submerged wood, water 25 °C, pH 5, 18 May 2000, *J. Anderson & R. Wulffen, A652–1*, **ILL 40814.**

Comments: The holotype of *Torrentispora pilosa* was not examined as it was described late in this study. The protologue however, is compelling in its placement in *Torrentispora* based on morphology alone. A culture is not noted in the original description. The two species evaluated molecularly in this study, *T. crassiparietis* and *T. fibrosa*, were shown to be unrelated, and not related to Annulatascaceae *sensu stricto*. To date, there are no species with relatively thick-walled ascospores in Annulatascaceae *sensu stricto*. At this time, *T. pilosa* should be considered Sordariomycetes *incertae sedis* at the level of family and order, and needs to be molecularly evaluated in terms of its generic placement.

Vertexicola K.D. Hyde, Ranghoo & S.W. Wong, Mycologia 92: 1019. 2000. MycoBank 28465.

Type species: *Vertexicola caudatus.*

Vertexicola aquatica V.M. Ranghoo & K.D. Hyde, Ranghoo Ph.D. thesis, 71. 1998.

Ascomata orange-brown, membranous, semi-immersed, gregarious, 250–275 µm wide × 250–263 µm high ($\bar{x}$ = 263 × 257 µm, w:h = 1), globose or subglobose. **Ascomal wall** 20–25 µm thick, composed of several layers of light brown elongated angular cells, of *textura angularis* in surface view. **Neck** orange-brown, 100–120 µm long × 30–40 µm wide, periphysate. **Paraphyses** hyaline, 4–5 µm wide at base × 120–250 µm long, tapering toward apex, septate. **Asci** unitunicate, cylindrical, 8–spored, 125–150 µm long × 33–38 µm wide ($\bar{x}$ = 235 × 11 µm, l:w = 21.4). **Ascus base** pedicellate, 25 µm from *pars sporifera*. **Annulus** J-, refractive, 4–5 µm long × 3–4 µm wide ($\bar{x}$ = 4.5 × 3.5 µm, l:w = 1.3). **Ascospores**

hyaline, fusiform, 3–5–septate, slightly constricted at septa, uniseriate to biseriate, 15–20 µm long × 7.5–10 µm wide (x̅ = 17.5 × 8.75 µm, l:w = 2), without appendages or sheath (Ranghoo 1998).

HOLOTYPE: CHINA. Hong Kong, New Territories, Plover Creek Reservoir, on submerged wood, Jan 1997, *V.M. Ranghoo,* **HKU(M) 5252.**

Comments: *Vertexicola aquatica* is not recognized in MycoBank or Index Fungorum. The holotype was not examined in this study as Ranghoo's thesis (1998) was only obtained near the completion of this manuscript. Thus only the protologue was used to assess this species morphologically. Ranghoo noted that the thesis would only have 5 copies made, and that due to this limited distribution the ICBN would not permit new species to be considered valid until published independently and broadly distributed. As this thesis is freely available from Hong Kong University at this time, this species is being included for the sake of completeness, with all credit given to Ranghoo (1998). The ascus measurements do not match the width reported above and appear to be 8–9 µm wide in the micrographs using the bar size reported in the figure legend. The ascomal color of *V. aquatica* is orange-brown, while the type of the genus, *V. caudatus*, has black ascomata. In addition *V. caudatus*, has asci with spike-like extensions of the pedicel, which are not present in *V. aquatica*. Without molecular data it is difficult to determine the phylogenetic placement of *V. aquatica* but the different ascomatal coloration and the lack of the spike-like tail makes placement in *Vertexicola* questionable. *V. aquatica* requires formal publication and molecular data to determine its phylogenetic placement.

Vertexicola ascoliberatus Shearer & F.R. Barbosa, Mycologia 105(2): 339. 2103. MycoBank 564459.

Ascomata brown to black, coriaceous, semi-immersed, perpendicular to parallel, scattered, 737–869 µm wide × 214–285 µm high (x̅ = 803 × 287 µm, w:h = 2.8), globose to subglobose. **Ascomal wall** black to brown. **Neck** black, to brown towards apex, 500–550 µm long × 100–120 µm wide, periphysate.

Paraphyses hyaline, 4–9 µm wide at base, septate, occasionally branching, filamentous. **Asci** unitunicate, cylindrical, 8–spored, 194–273 µm long × 10–12 µm wide (x̅ = 235 × 11 µm, l:w = 21.4). **Ascus base** tapering pedicel, with a spike-like tail, 15 µm from *pars sporifera*. **Annulus** J-, refractive, 4–5 µm long × 6–8 µm wide (x̅ = 4.5 × 7 µm, l:w = .64). **Ascospores** hyaline, ellipsoidal to fusiform, 0(3)–septate, 30–34 µm long × 10–12 µm wide (x̅ = 32 × 11 µm, l:w = 2.9), without appendages, with gelatinous sheath expanding in water (Barbosa et al. 2013).

HOLOTYPE: COSTA RICA. Heredia, La Selva Biological Station, Salto 30, 10°25'39"N, 84°0'9"W, on submerged wood, water 26 °C, pH 7, 18 May 2000, *J. Anderson & R. Wulffen, A653–1,* **ILL 40815.**

Known distribution: Costa Rica.

Comments: The holotype of *Vertexicola ascoliberatus* was not examined as it was described late in this study, and thus the protologue was used for morphological comparisons (Barbosa et al. 2013). A key feature used to place *V. ascoliberatus* in *Vertexicola* is the spike-like tail of the ascus. Whether this morphological feature is taxonomically informative is debatable. *Chaetorostrum quincemilensis*, which is distantly related to *V. caudatus*, and *Annulatascus tropicalis*, also distantly related, both exhibit spike-like ascus tails. *V. ascoliberatus* differs significantly from the type, *V. caudatus*, in which the ascus apical ring is shorter, the ascospores are consistently 5–septate, and a sheath is lacking. Molecular data are needed to determine whether *V. ascoliberatus* belongs in *Vertexicola*. Molecular analyses in this study with data from *V. caudatus* and the new species *V. triseptata* (PE0104) support the removal of *Vertexicola* from Annulatascaceae *sensu stricto* (**Fig. 7, Clade 1e**). *V. ascoliberatus* should be considered Sordariomycetes *incertae sedis*.

Vertexicola caudatus K.D. Hyde, V.M. Ranghoo & S.W. Wong, Mycologia 92(5): 1020. 2000. MycoBank 467602.

= *Vertexicola confusa* K.D. Hyde, S.W. Wong & E.B.G. Jones, Wong Ph.D. thesis, 165. 1996.

Ascomata black, coriaceous, semi-immersed to superficial, perpendicular to surface, solitary, 240–360 µm wide, globose to subglobose. **Ascomal wall** black, of *textura prismatica* in surface view. **Neck** black, 80–120 µm long × 40–60 µm wide, periphysate. **Paraphyses** hyaline, 6 µm wide at base, tapering toward apex, septate, constricted at septa. **Asci** unitunicate, cylindrical, 8–spored, 170–290 µm long × 7.5–12 µm wide ($\bar{x}$ = 230 × 9.8 µm, l:w = 23.6). **Ascus base** tapering pedicel, with a spike-like tail, 20 µm from *pars sporifera*. **Annulus** bipartite, 4 µm long × 5 µm wide (l:w = .8). **Ascospores** hyaline, ellipsoidal to fusiform, 5–septate, not constricted at septa, uniseriate to overlapping uniseriate, 18–24 µm long × 6–9 µm wide ($\bar{x}$ = 21 × 7.5 µm, l:w = .64), guttulate, without appendages or sheath (Hyde et al. 2000).

HOLOTYPE: PHILIPPINES. Bukidnon, Natigbasan Creek, Impalutao, Mindanao, on submerged wood, Jan 1994, *K.D. Hyde,* **HKU(M) 3108.**

Reports:

CHINA. Hong Kong, Tai Po, Lam Tsuen River, on submerged wood, Sep 1997, *K.M. Tsui, HKU(M) 8111;* Sai Kung, Hang Cho Shui stream, on submerged wood, Sep 1998, *K.M. Tsui, HKU(M) 12237*.

Known distribution: China, Philippines.

Comments: The holotype of *Vertexicola caudatus* was unavailable for examination. *V. caudatus* with a sequence deposited in GenBank under the name *V. confusa* forms a well supported monophyletic group with the new species *V. triseptata* (PE0104) (**Fig. 7, Clade 1e**). The genus *Vertexicola* was first introduced in Wong's doctoral thesis (1996). Two other putative *Vertexicola* species, *V. aquatica* and *V. lignicola* (= *Rivulicola aquatica*), were described in the doctoral thesis of Ranghoo (1998). *V. aquatica* differs from *V. caudatus* in having orange-brown ascomata and the lack of a spike-like ascus tail. *V. lignicola* was later formally published as *Rivulicola aquatica* (Ranghoo and Hyde 2000). The *Vertexicola* clade does not have affinity with Annulatascaceae *sensu stricto* and is a separate lineage. The third species in the genus, *V. ascoliberatus,* as mentioned above, lacks available molecular data, and differs morphologically from *V. caudatus*. In *V. caudatus* the neck is shorter, has narrower, unbranched paraphyses, and has a shallower apical ring compared to *V. ascoliberatus*. *V. caudatus* lacks an ascospore sheath and its ascospores are narrower and 0–6 septate as compared to 0–3 septate in *V. ascoliberatus*. *Vertexicola* should be ascribed to Sordariomycetes *incertae sedis* in terms of higher level taxonomic placement.

NEW TAXA

***Submersomyces triseptatus* gen. et sp. nov. [A612]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 68 A-I.**

Ascomata pale golden brown, immersed. **Ascomal wall** golden brown. **Neck** hyaline, erumpent, periphysate. **Paraphyses** hyaline, long, tapering, septate, filamentous (**Fig. 68 F**). **Asci** unitunicate, 8–spored, 100–140 µm long × 7.5–10 µm wide ($\bar{x}$ = 120 × 8.75 µm, l:w = 13.7, n = 20) (**Fig. 68 A-D**). **Annulus** bipartite, 2.5 µm long × 4 µm wide (l:w = .63, n = 10) (**Fig. 68 E**). **Ascospores** hyaline, ellipsoidal to fusiform, 3-septate upon maturity, not constricted at septa, uniseriate to overlapping uniseriate, 12–16 µm long × 6.5–8 µm wide ($\bar{x}$ = 14 × 7.25 µm, l:w = 1.9, n = 30), multiguttulate, without appendages or sheath (**Fig. 68 G-I**).

Specimens examined:

USA. Tennessee, GSMNP, The Sinks, lentic swamp, water 11 °C, pH 5, on submerged woody debris, 20 Mar 2007, *H.A. Raja, A.N. Miller, and P. Chaudhary, A612-1;* Wisconsin, Lemonweir River, on submerged woody debris, 5 Oct 2009, *H.A. Raja and A.N. Miller, A612-3*.

Comments: Taxon A612 was assessed molecularly due to its similarity to *Annulatascus* species, i.e. it possesses long tapering septate paraphyses, and unitunicate 8–spored asci with relatively massive J- ascus apical rings. A few permanent slides of decent quality were available to construct a photographic plate, but ascomata were not measurable or photographed. Taxon A612 is not genetically related to Annulatascaceae *sensu stricto* or any other putative Annulatascaceae taxa. In combined three gene analyses it is placed on a long branch basal to nearly all other Sordariomycetes taxa (**Fig. 7, Clade 8**). At present the species should be considered Sordariomycetes *incertae sedis*.

Fig. 68 A-I. ***Submersomyces triseptatus*** A612. A-C. Asci and paraphyses. D. Ascus. E. Ascus apex illustrating apical refractive annulus. F. Paraphyses. G-H. Ascospores in water. I. Ascospore in glycerin illustrating septa. Bars: A = 200 µm; B = 20 µm; D-G = 10 µm.

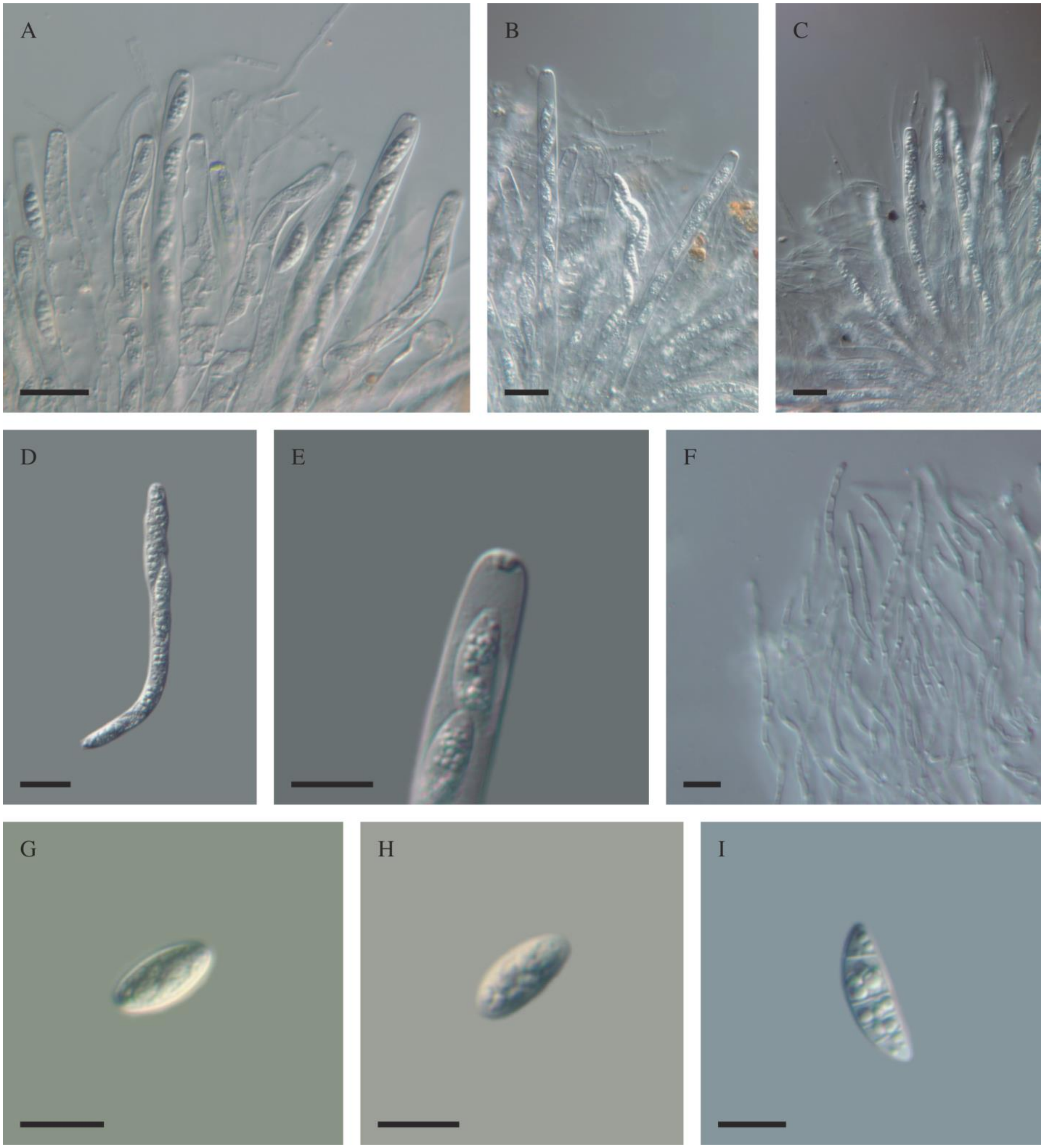


Fig. 68.

***Aquanigriparvum pearidgensis* gen. et sp. nov. [AK-1838]** Zelski, A.N. Mill. & Shearer. **Fig. 69 A-G.**

Ascomata black, coriaceous, semi-immersed to superficial, perpendicular to surface, solitary, 240–300 µm wide × 250–280 µm high, globose to subglobose (**Fig. 69 A**). **Ascomal wall** dark brown to black (**Fig. 69 B**). **Neck** black, 80–120 µm long × 40–60 µm wide, periphysate (**Fig. 69 A**). **Paraphyses** hyaline, tapering, septate. **Asci** unitunicate, 8–spored, 60–80 µm long × 6.5–8 µm wide ($\bar{x}$ = 72 × 7.3 µm, l:w = 9.9, n = 10) (**Fig. 69 C**). **Annulus** bipartite, 2 µm long × 2.5 µm wide (l:w = .8, n = 10) (**Fig. 69 D**). **Ascospores** hyaline, ellipsoidal to fusiform, uniseptate, not constricted at septa, thick walled, uniseriate to overlapping uniseriate, 14–16 µm long × 6–8 µm wide ($\bar{x}$ = 14.4 × 7.5 µm, l:w = 1.9, n = 30), guttulate, without appendages or sheath (**Fig. 69 E-G**).

Specimens examined:

USA. Arkansas, Pea Ridge National Military Park, Park proper, on submerged woody debris, Sep 2013, *Zelski S.E. and V.P. Hustad, AK-1837;* Pea Ridge National Military Park, Park annex, on submerged woody debris, Sep 2013, *Zelski S.E. and V.P. Hustad, AK-1838.*

Comments: Taxon AK-1838 was initially diagnosed as *Aquaticola ellipsoidea* due to the shape of the ascus, the morphology of the ascospores, and a general lack of morphological characteristics. Molecular analyses, however, place AK-1838 removed from the type of the genus, *Aq. hyalomura*, as well as *Aq. ellipsoidea*, and instead in a large clade containing *Annulatascus*-like species, *Aquaticola*-like species, *Fusoidispora aquatica*, *Lentomitella* species, and *Torrentispora fibrosa* (**Fig. 7, Clade 1f**). This clade is supported by Bayesian but not likelihood values. These taxa all share a J- apical apparatus, but beyond that there is a wide range of ascospore morphologies and annulus sizes. *Aquanigriparvum pearidgensis* should be considered Sordariomycetes *incertae sedis*.

Fig. 69 A-G. ***Aquanigriparvum pearidgensis*** AK-1838. A. Ascomata on surface of substrate. B. Peridium. C. Ascus. D. Ascus apex illustrating small refractive annulus. E-G. Ascospores. Bars: A = 200 µm; B = 20 µm; D-G = 10 µm.

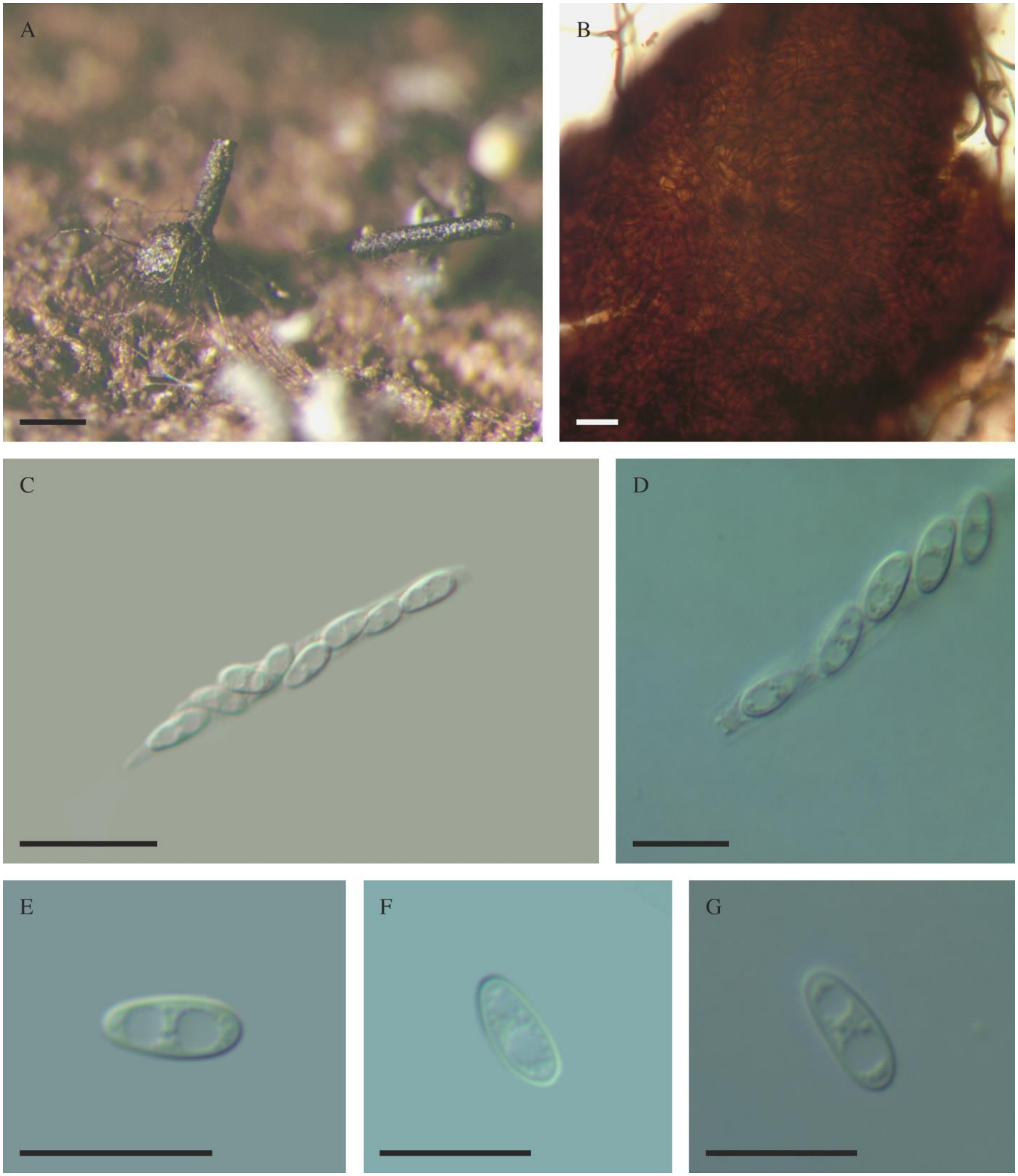


Fig. 69.

***Andesclivomyces submersus* gen. et sp. nov. [PE0035]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 70 A-I.**

Ascomata dark brown to black, coriaceous, semi-immersed to superficial, perpendicular to surface, gregarious, 240–360 µm wide, globose to subglobose (**Fig. 70 A**). **Ascomal wall** dark brown to black (**Fig. 70 B**). **Neck** black, 80–120 µm long × 40–60 µm wide, periphysate (**Fig. 70 A**). **Paraphyses** hyaline, 6 µm wide at base, tapering toward apex, septate. **Asci** unitunicate, 8–spored, 170–290 µm long × 7.5–12 µm wide ($\bar{x}$ = 230 × 9.8 µm, l:w = 23.6, n = 20) (**Fig. 70 C**). **Ascus base** short rounded knob (**Fig. 70 G**). **Annulus** bipartite, 4 µm long × 5 µm wide (l:w = .8, n = 10) (**Fig. 70 D-F**). **Ascospores** hyaline, ellipsoidal to fusiform, 5-septate, not constricted at septa, uniseriate to overlapping uniseriate, 18–24 µm long × 6–9 µm wide ($\bar{x}$ = 21 × 7.5 µm, l:w = .64, n = 30), guttulate, without appendages or sheath (**Fig. 70 H,I**).

Specimens examined:

PERU. Cusco, Camanti, Quincemil, stream at Quincemil Trail 2, on submerged woody debris, 13°13’34.0674”S, 70°45’12.6714”W, 669m, water 21.8 °C, pH 7.6, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0035-1;* Rio Frio (Yana Mayo), on submerged woody debris, 13°13’20.69”S, 70°44’25.80”W, 627m, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0035-4;* Madre de Dios, Palm swamp (aguajal) near the Interoceanic Highway near Puerto Maldonado, trail head 12°42’48.0954”S, 69°28’11.28”W, 239m, water 23.3 °C, pH 5.9, 20 May 2010, *Zelski S.E. and H.A. Raja, PE0035-3.*

Known distribution: Perú.

Comments: Taxon PE0035 was first diagnosed as *Annulatascus joannae*. However, upon maturation the ascospores changed from unicellular and blunt ended to 5-septate and fusiform-lunate. Combined three gene analyses place this species removed from Annulatascaceae *sensu stricto* and on a long branch as a separate lineage (**Fig. 7, Clade 1f**). Thus, a new genus and species are warranted for PE0035. It has been

Fig. 70 A-I. ***Andesclivomyces submersus*** PE0035. A. Ascomata on surface of substrate. B. Peridium. C. Ascus. D-F. Ascus apices illustrating refractive annulus. G. Ascus base. H-I. Ascospores. Bars: A = 100 µm; B-E, G-I = 20 µm; F = 10 µm.

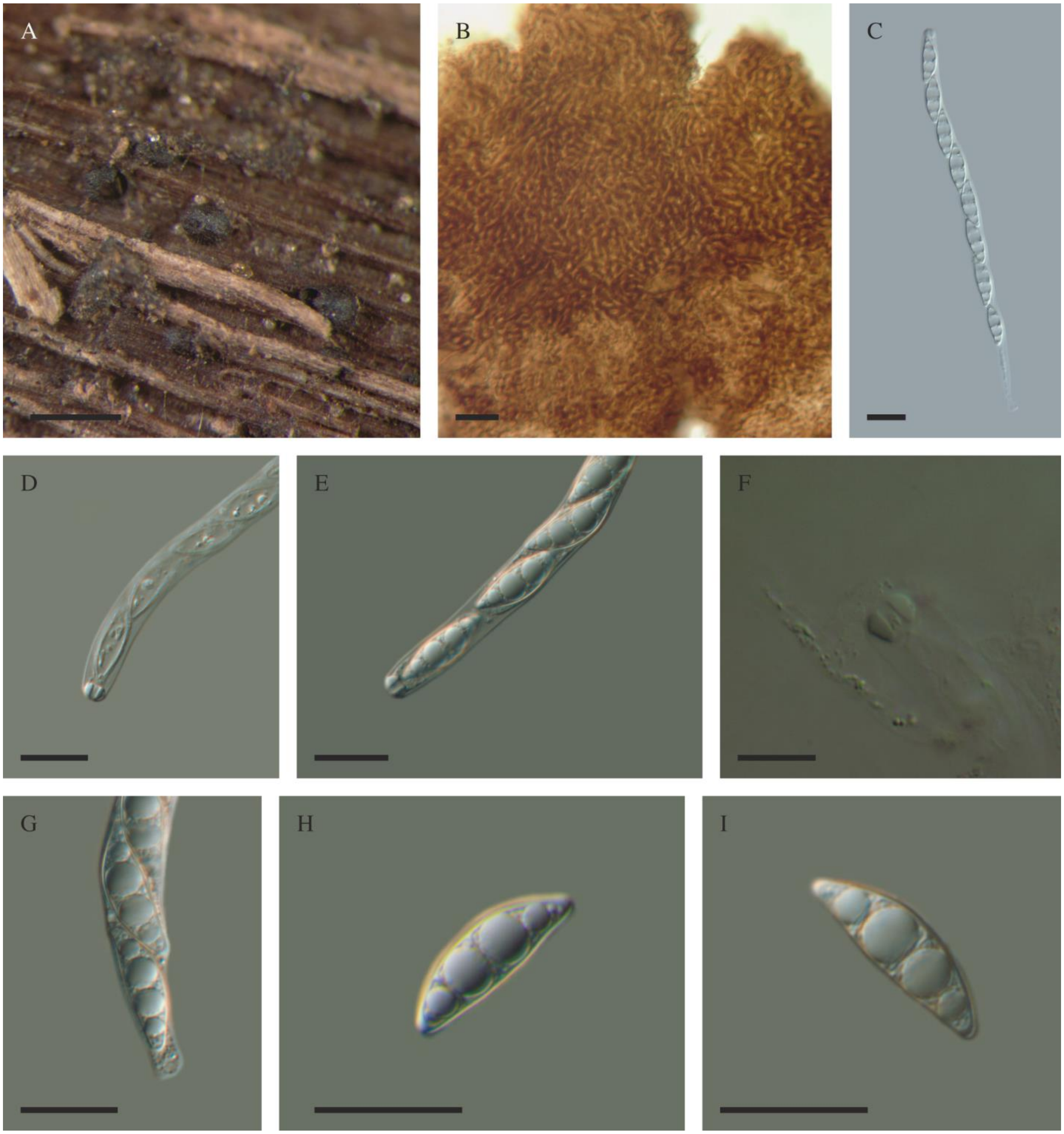

A
B
C
D
E
F
G
H
I

Fig. 70.

noted throughout this study that the presence of a large J- ascus apical apparatus, while being a defining feature of Annulatascaceae, is not a sufficient trait for inclusion in the family. This species adds further credence to this idea, even though morphological examination alone would lead to an Annulatascaceae diagnosis. In a clade separating Diaporthales and Ophiostomatales, PE0035 should be considered Sordariomycetes *incertae sedis*.

***Ascotaiwania* sp. [PE0059]** Zelski, Raja, A.N. Mill. & Shearer.

Specimens examined:

PERU. Cusco, Camanti, Quincemil, Stream crossing Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 653m, water 21.7 °C, pH 7.6, on submerged woody debris, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0059-1*; River at end of Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.5 °C, pH 7.7, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0058-2;* Stream crossing the Interoceanic Highway, 13°17’7.008”S, 70°47’13.632”W, 653m, on submerged woody debris, water 22°C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A Raja., PE0059-3*.

Known distribution: Perú.

Comments: A full description is not included here as this species is not a member of Annulatascaceae, rather it is a member of Savoryellales. In addition, preserved material is scant and in poor condition. This species was sequenced as it was initially diagnosed as *Rivulicola aquatica*, and it may be that ascospores matured to a pigmented state after initial observations. It is hoped that fresh material from the fourth collection from Perú will allow good photomicrographs to be taken. PE0059 is included here as its sequences were included in the phylogenetic analyses.

Albumglobomyces peruviensis **sp. nov. [PE0157]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 71 A-H.**

Ascomata hyaline, membranous, superficial, perpendicular to surface, highly gregarious, 150 µm wide × 150 µm, (w:h = 1) globose (**Fig. 71 A**). **Ascomal wall** hyaline. **Neck** hyaline, protruding from surface of substrate, 100 µm long × 60 µm wide, periphysate (**Fig. 71 B**). **Paraphyses** hyaline, tapering, septate (**Fig. 71 C**). **Asci** unitunicate, 8–spored, 80–85 µm long × 6–8 µm wide ($\bar{x}$ = 82.5 × 7 µm, l:w = 15, n = 20) (**Fig. 71 D**). **Ascus base** short, ending in club-like pedicel, to 20 µm from *pars sporifera*. **Annulus** bipartite, 2 µm long × 2.5 µm wide (l:w = .8, n = 10) (**Fig. 71 E,F**). **Ascospores** hyaline, ellipsoidal to fusiform, aseptate, uniseriate to overlapping uniseriate, 10.1–14.6 µm long × 6.2–7.4 µm wide ($\bar{x}$ = 11.8 × 6.8 µm, l:w = 1.7, n = 30), multiguttulate, without appendages or sheath (**Fig. 71 G,H**).

Specimens examined:

PERU. Cusco, Camanti, Quincemil, river at Quincemil trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21 °C, pH 6.3, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0157-1;* Stream at Quincemil trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114’W, 688m, on submerged woody debris, water 21.2 °C, pH 6.8, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0157-2;* Rio Frio (Yana Mayo), 13°13’20.87”S. 70°44’30.07”W, 626m, water 25.3 °C, pH 8, on submerged woody debris, *Zelski S.E. and H.A. Raja, PE0157-6;* Stream with red algae along Quincemil trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, water 21.8 °C, pH 7.2, on submerged woody debris, 13 Apr 2011, *Zelski S.E. and H.A. Raja,* PE0157-5. Madre de Dios, CICRA, Stream on Trail 14, 12°34’14.7”S, 70°05’23.69’W, 241m, on submerged woody debris, water 25.1 °C, pH 7.3, 30 Sep 2010, *Zelski S.E. and H.A. Raja, PE0157-3.*

Known distribution: Perú.

Fig. 71 A-H. *Albumglobomyces peruviensis* PE0157. A. Superficial hyaline ascomata with ascospores amassed at neck apices. B. Neck. C. Paraphyses. D. Ascus. E. Ascus apices. F. Ascus apex. G. Ascospores. H. Ascospore. Bars: A = 200 µm; G-H = 10 µm; B-E = 20 µm; F = 5 µm.

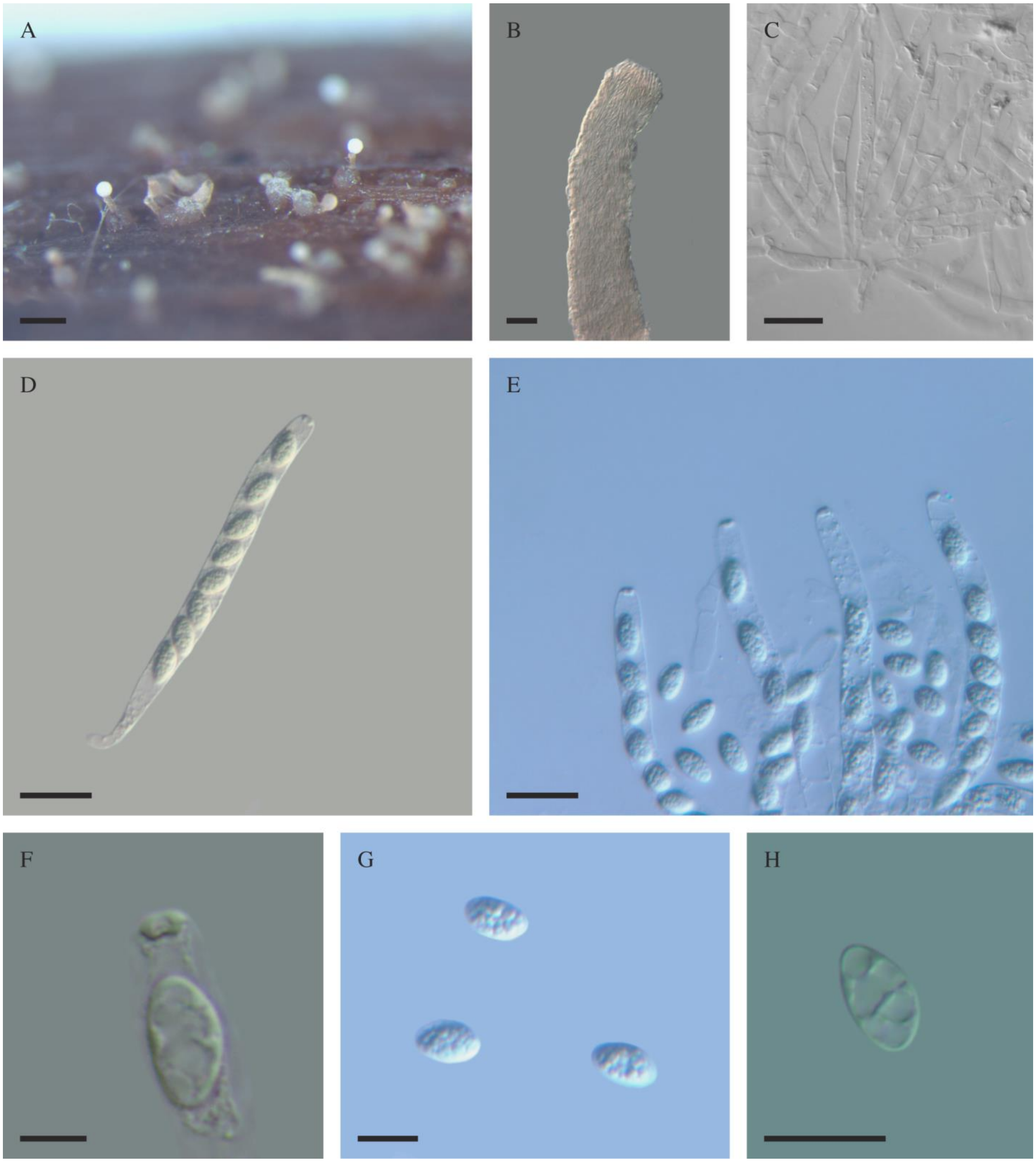


Fig. 71.

Comments: Taxon PE0157 is morphologically similar to *Aquaticola hyalomura*, and has ascospores of similar dimensions (10.1–14.6 × 5.8–7.4 µm vs. 10-14 × 5-7 µm). One collection, PE0157-2, had slightly brown tinted ascomata while the other specimens of PE0157 had distinctive hyaline ascomata. This pigmentation may have come from the substrate. Ascospores differ from *Aq. hyalomura* in that they are multiguttulate rather than possessing two large lipid guttules. Phylogenetic analyses place PE0157 as a sister taxon to PE0264 with high support although not closely related to *Aq*. hyalomura (**Fig. 7; Clade 1b, Clade 1c**). PE0157 differs from PE0264 in having superficial ascomata and shorter asci. Neither PE0157 nor PE0264, though originally thought to be specimens of *Aq. hyalomura*, are placed sister to that species (**Fig. 7, Clade 1b**). They do however cluster in a clade with no statistical support sister to the clade containing *Aq. hyalomura* as well as *Cataractispora* species. The closest neighbor to these new Peruvian species is *Ayria nubispora* in these analyses although there is no statistical support for the node (**Fig 7, Clade 1c**). *Ay. nubispora* shares few morphological similarities (one exception being hyaline ascospores) with PE0157. Taxon PE0157 is in a clade separating Diaporthales and Ophiostomatales and should be considered Sordariomycetes *incertae sedis*.

***Insolensaquaticola striatispora* sp. nov. [PE0159]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 72 A-H.**

Ascomata dark brown, coriaceous, immersed to superficial, perpendicular to surface, scattered, 240–360 µm wide × 250–270 µm high (w:h = 1.2), globose to subglobose. **Ascomal wall** black. **Neck** black, 80-120 µm long × 40-60 µm wide, periphysate. **Paraphyses** hyaline, 2–3 µm wide at base, long, tapering, septate (**Fig. 72 E**). **Asci** unitunicate, 8–spored, overlapping uniseriate, 190–200 µm long × 10–12 µm wide (x̅ = 195 × 11 µm, l:w = 18.2, n = 8) (**Fig. 72 A,B**). **Ascus base** pedicellate, 15 µm from *pars sporifera*. **Annulus** bipartite, 2 µm long × 4 µm wide (l:w = .5, n = 10) (**Fig. 72 C,D**). **Ascospores** hyaline, ellipsoidal to fusiform, aseptate, 20–22 µm long × 9–10 µm wide (x̅ = 21.2 × 9.8 µm, l:w = 2.2, n

Fig. 72 A-H. ***Insolensaquaticola striatispora*** PE0159. A. Asci and paraphyses. B. Ascus. C. Ruptured ascus. D. Ascus apex. E. Paraphyses. F. Ascospore illustrating longitudinal striations. G. Ascospores illustrating sheaths. H. Ascospore with sheath extended. Bars: A-H = 20 µm.

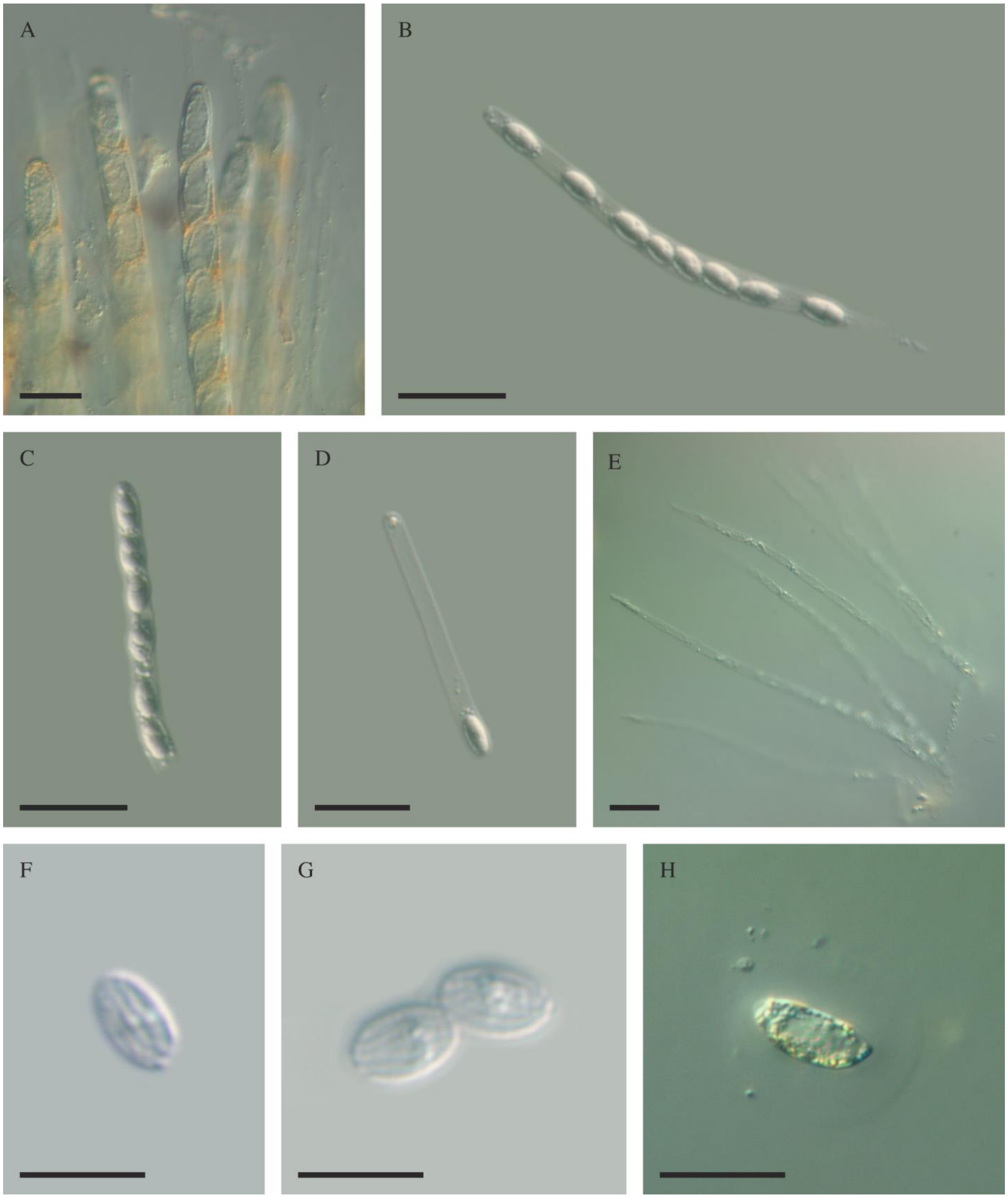


Fig. 72.

= 10), multiguttulate, with longitudinal striations, without appendages, with a spreading mucilaginous sheath (**Fig. 72 F-H**).

Specimens examined:

PERU. Cusco, Camanti, Quincemil, river at Quincemil Trail 1, trailhead 13°14'22.5594"S, 70°46'12.6114"W, 688m, water 21 °C, pH 6.3, on submerged woody debris, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0159-1*.

Known distribution: Perú.

Comments: PE0159 is a sister taxon with strong Bayesian and Maximum-likelihood support to *Aquaticola triseptata* (**Fig. 7, Clade 3a**). Although it is also in a clade with *Submersisphaeria rattanicola* and *Clohiesia corticola*, this larger grouping is a polytomy. PE0159, as well as *Aq. triseptata* are not related to Annulatascaceae *sensu stricto* nor the type of the genus *Aquaticola*, *Aq. hyalomura*. Images of PE0159 were taken from preserved specimens and the mucilaginous sheaths were not preserved in glycerin. PE0159 represents another *Aquaticola*-like species which does not belong in the genus. It does, however have morphological features, such as the ascospore striations and sheath, which serve to distinguish the species. PE0159 is considered Sordariomycetes *incertae sedis* at present and placed in the proposed new genus *Insolensaquaticola* with *I. triseptata*.

***Ascotaiwania* sp. [PE0238]** Zelski, Raja, A.N. Mill. & Shearer.

Specimens examined:

PERU. Cusco, Camanti, Quincemil, river at Quincemil Trail 3, trailhead 13°18'22.756"S, 70°48'44.9274"W, 757m, on submerged woody debris, water 21 °C, pH 7.7, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0238-1;* Junin, Central jungle, stream, 10°52'20.352"N, 74°59'35.2674"W, 529m, on submerged woody debris, water 23.4 °C, pH 9.4, 846 µS, DO 8.5, 27 May 2012, *Zelski S.E. and J. Balto, PE0238-2.*

Comments: A full description is not included here as this species is not a member of Annulatascaceae, rather it is a member of Savoryellales. In addition, preserved material is scant and in poor condition. This species was sequenced as it was initially diagnosed as a good match to *Ascolacicola aquatica* due to its 3-septate light brown ascospores with hyaline end cells. It is hoped that fresh material from the fourth collection from Perú will allow quality photomicrographs to be taken. This species is included here as its sequences were included in the phylogenetic analyses.

Albumglobomyces immersus **gen. et sp. nov. [PE0264] (**Zelski, Raja, A.N. Mill. & Shearer. **Fig. 73 A-J.**

Ascomata hyaline, membranous, immersed, perpendicular to surface, gregarious, 200–250 µm wide × 200–250 µm high, (w:h = 1) globose (**Fig. 73 A,B**). **Ascomal wall** hyaline, 25 µm thick, composed of an outer layer of hyaline globular cells and inner layer of 3–5 hyaline compressed cells, of *textura epidermoidea* in surface view (**Fig. 73 B**). **Neck** hyaline, protruding from surface of substrate, 100 µm long × 60 µm wide, periphysate (**Fig. 73 C,D**). **Paraphyses** hyaline, tapering, septate. **Asci** unitunicate, 8–spored, 100–110 µm long × 6–8 µm wide ($\bar{x}$ = 105 × 7 µm, l:w = 15, n = 20) (**Fig. 73 E**). **Ascus base** short, ending in club-like pedicel, 20 µm from *pars sporifera* (**Fig. 73 F**). **Annulus** bipartite, 2 µm long × 2.5 µm wide (l:w = .8, n = 10) (**Fig. 73 G,H**). **Ascospores** hyaline, ellipsoidal to fusiform, aseptate, uniseriate to overlapping uniseriate, 9–10 µm long × 6–8 µm wide ($\bar{x}$ = 9.5 × 7.5 µm, l:w = 1.3, n = 30), multiguttulate, without appendages or sheath (**Fig. 73 I,J**).

Specimens examined:

PERU. Cusco, Camanti, Quincemil, 5th stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 19 °C, pH 8.3, on submerged woody debris, 3 Oct 2010, *Zelski S.E. and*

H.A. Raja,PE0264-6; 3rd stream along Quincemil Trail 1, on submerged woody debris, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 19.1 °C, pH 6.7, Oct 2010, *Zelski S.E. and H.A. Raja, PE0264-7;* 2nd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 20 °C, pH 5.8, 3 Oct 2010, on submerged woody debris, *Zelski S.E. and H.A. Raja, PE0264-8;* Stream at Quincemil Trail 3, trailhead 13°18’22.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 21.5 °C, pH 7.5, 4 Oct 2010, *Zelski S.E. and H.A. Raja, PE0264-5;* Rio Frio (Yana Mayo), 13°13’20.87”S. 70°44’30.07”W, 626m, on submerged woody debris, water 25.3 °C, pH 8, *Zelski S.E. and H.A. Raja PE0264-35;* River at end of Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.5 °C, pH 7.7, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-23;* 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-12;* 2nd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-27;* 1st stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 21.7 °C, pH 6.8, on submerged woody debris, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-14;* Stream flowing into river backwater at Quincemil Trail 3, on submerged decorticated wood, 13°18’27.756”S, 70°48’44.9279”W, 757m, water 21.2 °C, pH 7.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-24;* River backwater at Quincemil Trail 3, on submerged decorticated wood, 13°18’27.756”S, 70°48’44.9279”W, 757m, water 22 °C, pH 7.1, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-28;* Stream with red algae along Quincemil trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, water 21.8 °C, pH 7.2, on submerged woody debris, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-31;* Madre de Dios, CICRA, Rio Amigos, 12°33’25.22”S, 70°05’59.89”W, 288m, water 31.4 °C, pH 8, on submerged woody debris, 1 Oct 2010, *Zelski S.E. and H.A. Raja, PE0264-4;* Stream along at Trail 20, 12°33’25.22”S, 70°05’59.89”W, 288m, water 23.1 °C, pH 8.3, on submerged woody debris, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-11;* Semi-aquatic habitat on Trail 1, 12°34’06.52”S, 70°06’04.57”W, 263m, on submerged woody debris, water 23.7 °C, pH 5.1, on submerged woody debris, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-10;* Stream at Trail 19, 12°34’01.04”S, 70°05’43.24”W, 275m, water 23.6 °C, pH 6.7, on submerged decorticated wood, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0264-32.*

Known distribution: Perú.

Comments: Taxon PE0264 was originally diagnosed as *Aquaticola hyalomura* due to its hyaline ascomata and the morphological similarity of its asci and ascospores. Combined three gene sequence analyses, however, did not confirm this diagnosis. Taxon PE0264 does cluster in a clades sister to a clade in which *Aq. hyalomura* is a member, although the clade and its constituents are a weakly statistically supported group (**Fig. 7, Clade 1c**). There is some doubt whether the GenBank sequence of *Aq. hyalomura* is genuine or represents *Cataractispora aquatica*, as mentioned in the results section. PE0264 is supported as a sister taxon to another Peruvian species initially diagnosed as *Aq. hyalomura*, PE0157. These two taxa differ in that PE0157 has superficial ascomata and PE0264 has immersed ascomata. They also differ in that PE0157 has shorter asci compared to those of PE0264 (80–85 µm long × 6–8 µm wide vs. 100–110 µm long × 6–8 µm) and longer, wider ascospores (10.1–14.6 µm long × 6.2–7.4 µm wide vs. 9–10 µm long × 6–8 µm wide). These species potentially are conspecific as evidenced by the support values on the branch terminating in these taxa (**Fig. 7, Clade 1c**). The holotype of *Aq. hyalomura* was unavailable, thus comparison was made to the protologue description. PE0264 differs in having a longer asci (100–110 µm vs. 52– 64 µm) and shorter ascospores compared to *Aq. hyalomura* (9–10 µm long vs. 10-14). In addition to PE03264 has multiguttulate ascospores as opposed to *Aq. hyalomura* which has ascospores containing two large lipid droplets. *Aquaticola* species are difficult to differentiate because morphological features are few thus molecular data is required to establish phylogenetic placement. PE0264 is in a clade separating Diaporthales and Ophiostomatales and is thus considered Sordariomycetes *incertae sedis* (**Fig. 7, Clade 1c**).

Fig. 73 A-J. ***Albumglobomyces immersus*** PE0264. A. Hyaline necks of gregarious ascomata protruding from substrate. B. Ascoma illustrating hyaline wall. C. Neck with ascospores in central canal. D. Neck illustrating periphyses. E. Ascus. F. Ascus base. G. Ascus apex. H. Ascus apex stained with nigrosin. I-J. Ascospores. Bars: A = 500 µm; B-C = 100 µm; D-F = 20 µm; G, I-J = 10 µm; H = 5 µm.

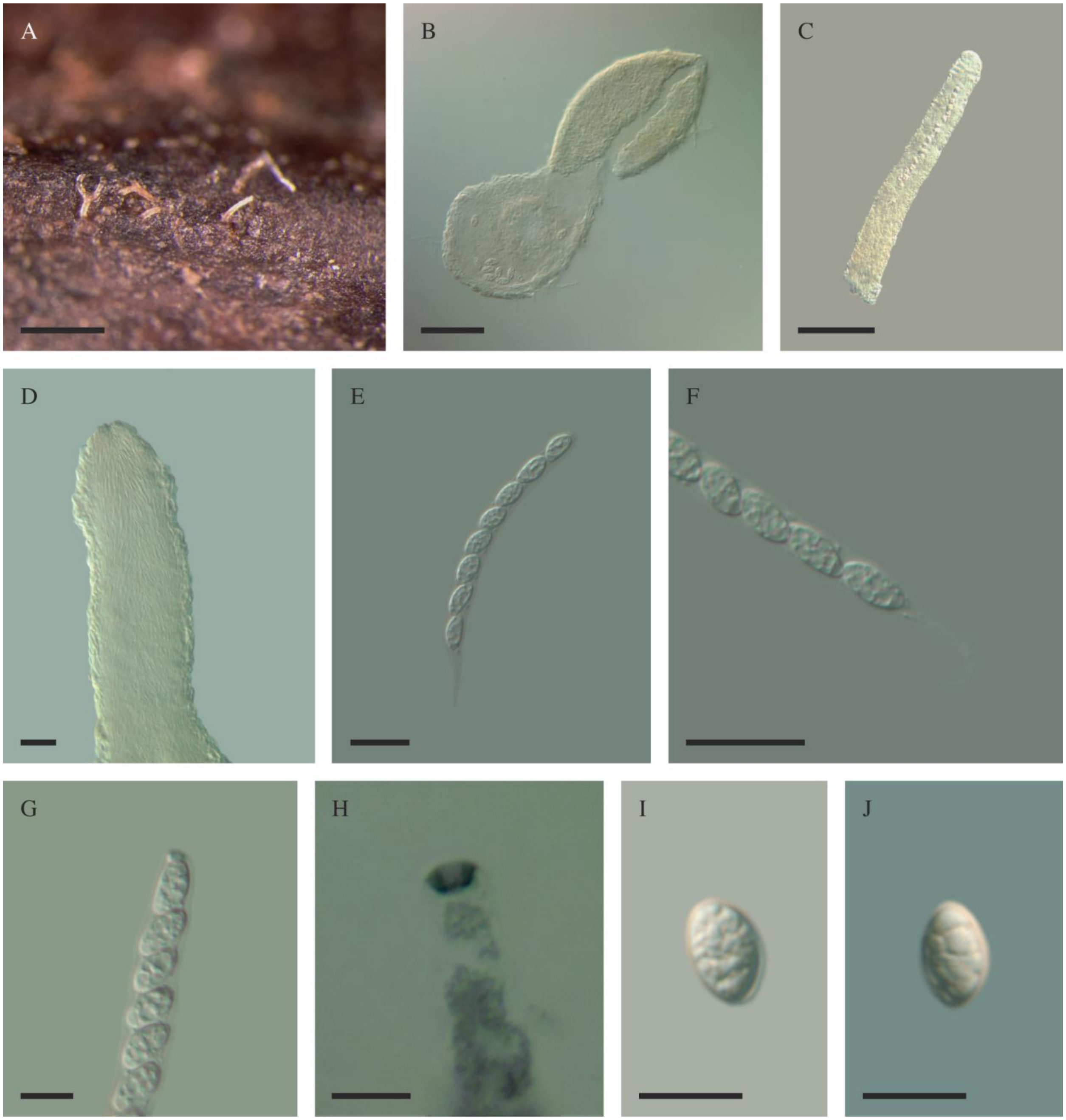


Fig. 73.

***Rubrobrunneorostrum aggregatofructum* gen. et sp. nov. [PE0266]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 74 A-J.**

Ascomata reddish-brown, membranous, superficial, perpendicular to surface, highly gregarious, 150 µm wide × 150 µm high, globose (**Fig. 74 A**). **Ascomal wall** reddish-brown, pseudoparenchymatous in surface view (**Fig. 74 C**). **Neck** reddish-brown, 200-210 µm long × 40-50 µm wide, periphysate (**Fig. 74 B**). **Paraphyses** hyaline, 5 µm wide at base, tapering, septate, constricted at septa (**Fig. 74 F**). **Asci** unitunicate, 8–spored, 80-100 µm long × 7.5-12 µm wide ($\overline{x}$ = 90 × 9.8 µm, l × w = 9.2) (**Fig. 74 D,E**). **Annulus** bipartite, 2.5 µm long × 2.5 µm wide (l × w = 1) (**Fig. 74 G**). **Ascospores** hyaline, fusiform, 3-septate, not constricted at septa, septa thick-walled, uniseriate to overlapping uniseriate, 15-17 µm long × 7-8 µm wide ($\overline{x}$ = 16 × 7.5 µm, l × w = 2.1), guttulate, without appendages or sheath (**Fig. 74 H-J**).

Specimens examined:

PERU. Cusco, Camanti, Quincemil, 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0266-1.*

Comments: This species was sequenced for ITS as part of a barcoding effort and the extracted DNA has gone missing. The species morphologically resembles *Cyanoannulus petersenii* due to its reddish-brown ascomata and 3-septate ascospores. Its ascomata, however, were densely clustered on the surface of the substrate compared to those of *Cy. petersenii*, which tend to be scattered. In addition, the ascus apical apparatus does not stain in cotton blue. ITS analyses place the two species as sister taxa but without statistical support, and PE0266 belongs in a clade with Diaporthales species with Bayesian, but not Maximum-likelihood support (**Fig. 4**). This species is not related to Annulatascaceae *sensu stricto* and should be considered Sordariomycetes *incertae sedis* at present.

Fig. 74 A-J. ***Rubrobrunneorostrum aggregatofructum*** PE0266. A. Superficial gregarious ascomata. B. Neck. C. Peridium. D-E. Asci. F. Paraphyses. G. Ascus apices. H-J. Ascospores. Bars: A = 200 µm; B = 40 µm; C-F = 20 µm; G-J = 10 µm.

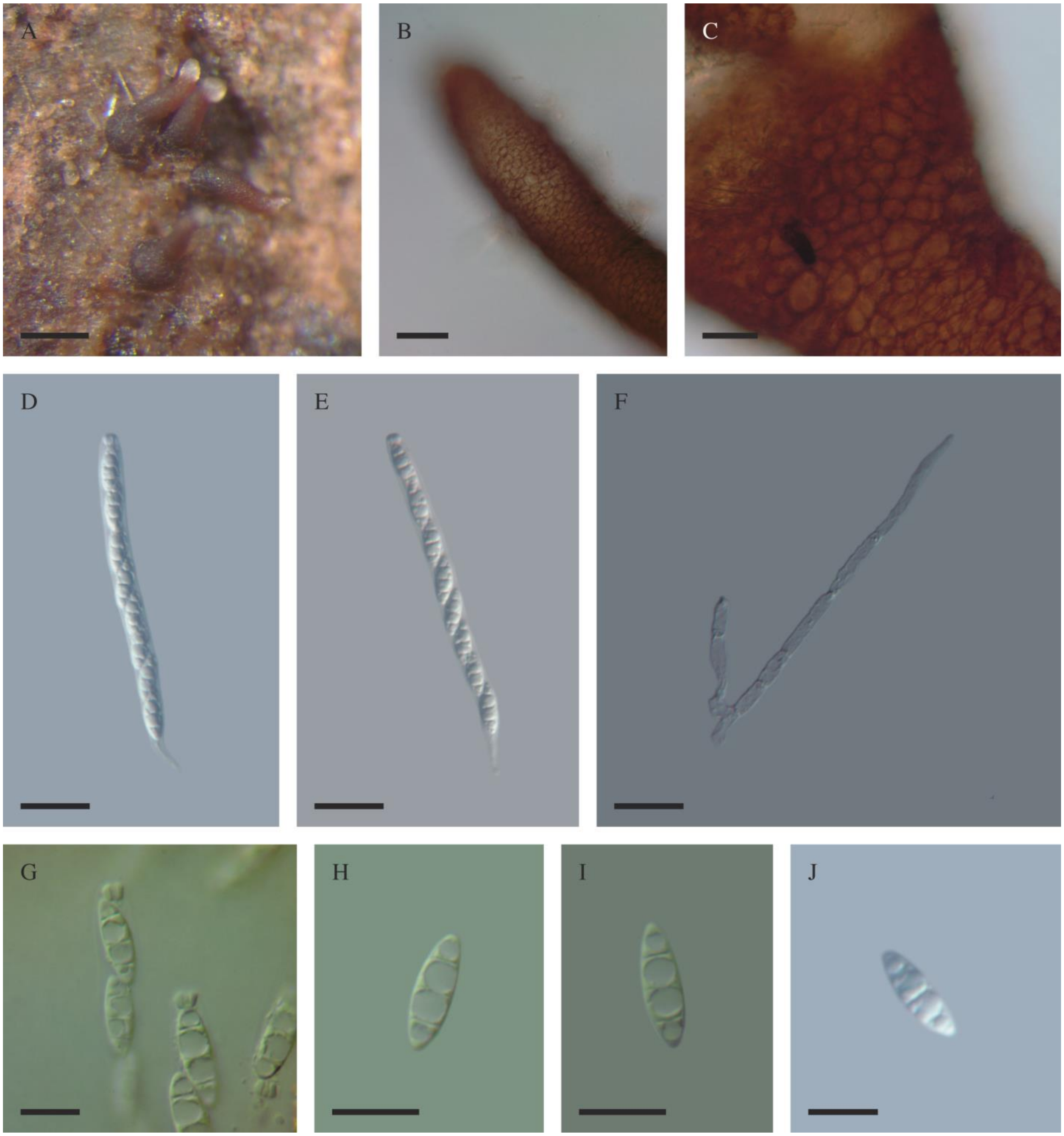


Fig. 74.

***Sororofusoidispora quincemilensis* gen. et. sp. nov. [PE0316]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 75 A-G.**

Ascomata dark brown, coriaceous, semi-immersed to superficial, perpendicular to surface, scattered, globose to subglobose. **Ascomal wall** brown, of *textura prismatica* in surface view (**Fig. 75 A**). **Neck** brown, short, periphysate. **Paraphyses** hyaline, tapering, septate (**Fig. 75 E**). **Asci** unitunicate, 8–spored, 110–140 µm long × 10–12 µm wide ($\bar{x}$ = 122 × 10.2 µm, l:w = 12, n = 10) (**Fig. 75 B**). **Ascus base** short, blunt, 10 µm from *pars sporifera* (**Fig. 75 D**). **Annulus** bipartite, 3–3.5 µm long × 5–5.5 µm wide ($\bar{x}$ = 3.25 × 5.2 µm l:w = .63, n = 10) (**Fig. 75 C**). **Ascospores** hyaline, ellipsoidal to fusiform, aseptate, uniseriate to overlapping uniseriate, 30–35 µm long × 10–12 µm wide ($\bar{x}$ = 32.5 × 11 µm, l:w = 3, n = 30), thick walled, multiguttulate, without appendages or sheath (**Fig. 75 F,G**).

Specimens examined:

PERU. Cusco, Camanti, Quincemil, stream flowing into river backwater at Quincemil Trail 3, on submerged decorticated wood, 13°18’27.756”S, 70°48’44.9279”W, 757m, water 21.2 °C, pH 7.8, 13 Apr 2011, *Zelski S.E. and H.A. Raja, PE0316-1.*

Comments: PE0316 has features similar to both *Aquaticola* and *Torrentispora* species. Like *Aquaticola* species, PE0316 has short clavate asci with club-like pedicels, and like *Torrentispora* species it has a peridium of *textura prismatica* in surface view and thick-walled, aseptate ascospores. Phylogenetic analyses do not place PE0316 in a clade with the type of the genus *Torrentispora*, *T. fibrosa*, or with *T. crassiparietis* which were both evaluated in this study. Ascospores are smaller in PE0316 than in *T. crassiparietis* and have more acute ends than in *T. fibrosa*. In addition, PE0316 lacks a fibrillar sheath. In combined three gene analyses, PE0316 is in a clade sister with *Fusoidispora aquatica* and *Aq. miniguttulata* (**Fig. 7, Clade 1f**). PE0316 differs from *Fu. aquatica* in that its ascospores are not long fusiform and from *Aq. miniguttulata* in that its ascospores are much longer and are ellipsoidal fusiform

Fig. 75 A-G. ***Sororofusoidispora quincemilensis*** PE0316. A. Peridium. B. Asci. C Ascus apex. D. Ascus base. E. Paraphysis. F. Ascospores. G. Ascospore. Bars: A-B, D-E = 20 µm; C, F-G = 10 µm.

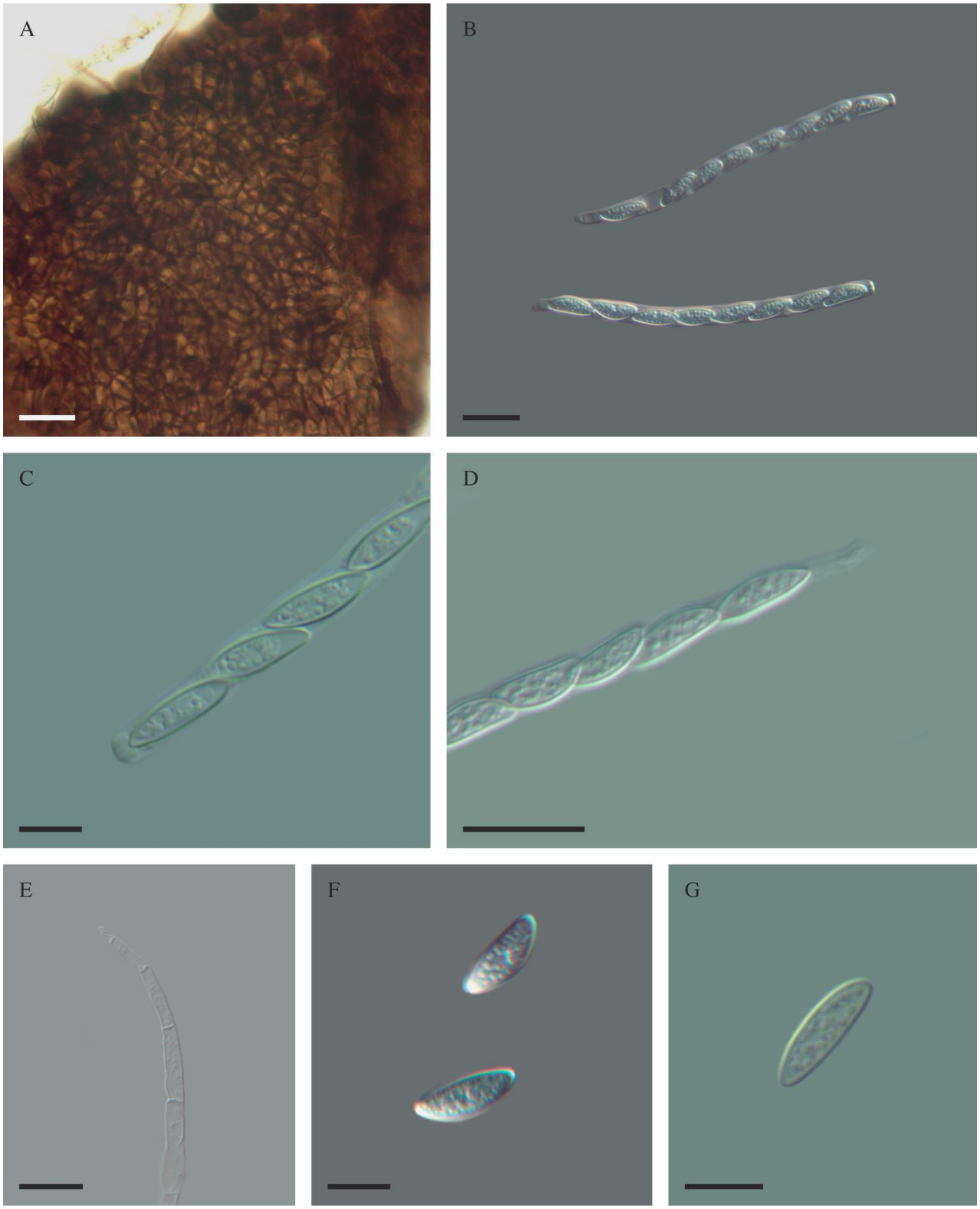


Fig. 75.

rather than ellipsoidal. For these reasons this species warrants a separate genus. PE0316 is not included in Annulatascaceae *sensu stricto* and as it nests in a clade separating Diaporthales and Ophiostomatales, it should be considered Sordariomycetes *incertae sedis*.

***Simileayria peruviana* gen. et. sp. nov. [PE0323]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 76 A-G.**

Ascomata subhyaline to light brown, membranous, superficial, perpendicular to surface, solitary, 400 µm wide × 500 µm high (w:h = .8), globose to subglobose (**Fig. 76 A**). **Ascomal wall** subhyaline with brown pigmentation at junctions of cells (**Fig. 76 A**). **Neck** ill-defined papilla (**Fig. 76 A**). **Paraphyses** hyaline, 5 µm wide at base, tapering toward apex, septate (**Fig. 76 D**). **Asci** unitunicate, 8–spored, 120–140 µm long × 19–25 µm wide ($\bar{x}$ = 130 × 22 µm, l:w = 5.9, n = 20) (**Fig. 76 B,C**). **Annulus** lacking. **Ascospores** hyaline, ellipsoidal, aseptate, overlapping biseriate, 33.5–41.8 µm long × 11–14.1 µm wide ($\bar{x}$ = 38 × 12.7 µm, l:w = 3, n = 30), multiguttulate, with bipolar, long, unwinding appendages, without a sheath (**Fig. 76 E-G**).

Specimens examined:

PERU. Cusco, Camanti Quincemil, 2nd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0323-2;* Madre de Dios, CICRA, stream at Trail 19, 12°34’01.04”S, 70°05’43.24”W, 275m, water 23.7 °C, pH 5.1, on submerged decorticated wood, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0323-1;* C-1777-*4;* CICRA, stream at Trail 19, 12°34’01.04”S, 70°05’43.24”W, 275m, on submerged decorticated wood, water 23.7 °C, pH 5.1, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0323-5.*

Known distribution: Perú.

Comments: This species has the hallmarks of *Ayria* and is morphologically most similar to *Ay. nubispora*. It differs in having smaller ascospores that lack mucilaginous sheaths. It differs from the type of the genus, *Ay. appendiculata,* in having ascospores that are roughly twice as large. Bipolar unfurling appendages are found in the *Halosphaeria* and *Aniptodera* species, as well as in *Pseudoproboscispora caudae-suis*, which is a member of the Annulatascaceae *sensu stricto* clade. Though sharing this feature, PE0323 is not related to *Ay. nubispora*, the Halosphaeriales, or Annulatascaceae *sensu stricto*, but rather is a sister taxon to a new *Annulatascus*-like species (PE0324), which has hyaline ascomata and ascospores lacking appendages and with mucilaginous sheaths (**Fig. 7, Clade 7**). Taxon PE0323 is considered Sordariomycetes *incertae sedis* at present.

***Albumnitidomyces lubricosporus* gen. et. sp. nov. [PE0324]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 77 A-I.**

Ascomata hyaline, membranous, semi-immersed to superficial, perpendicular to surface, covered with loose hyaline hyphal threads, gregarious, 119–185 µm wide × 152–217 µm high ($\bar{x}$ = 148.5 × 174.8 µm, w:h = .85, n=10), globose to obpyriform (**Fig. 77 A**). **Ascomal wall** hyaline, of *textura angularis* in surface view (**Fig**. **77 B,C**). **Neck** hyaline, cylindrical, 108–230 µm long × 48–61 µm wide ($\bar{x}$ = 182 × 55, n = 10), periphysate. **Paraphyses** hyaline, 5 µm wide at base, tapering, septate, as long as asci. **Asci** unitunicate, clavate, 8–spored, 170–290 µm long × 7.5–12 µm wide ($\bar{x}$ = 230 × 9.8 µm, l:w = 23.6, n = 20). **Ascus base** tapering, readily detachable pedicel, exhibiting back door to mid ascus dehiscence, 15 µm from *pars sporifera* (**Fig. 77 F**). **Annulus** bipartite, 2–2.7 µm long × 3.7–4.2 µm wide ($\bar{x}$ = 2.3 × 4, l:w = .58, n = 10) (**Fig, 77 D,E**). **Ascospores** hyaline, fusiform, 3-septate, not constricted at septa, uniseriate to overlapping uniseriate, 16.2–20.4 µm long × 6.6–8.3 µm wide ($\bar{x}$ = 18.6 × 7.4 µm, l:w = 2.5, n = 30), multiguttulate, without appendages with a gelatinous sheath (**Fig. 77 G-I**).

Fig. 76 A-G. ***Simileayria peruviana*** PE0323**.** A. Ascomata. B-C. Asci. D. Paraphysis. E. Ascospore with extended appendages. F-G. Ascospores with coiled bipolar appendages. Bars: A = 100 µm; B-G = 20 µm.

Fig. 76.

HOLOTYPE: PERU. Madre de Dios, CICRA, stream near dam on Trail 19, 12˚34’02.8”S, 70˚05’42.96”W, on submerged woody debris, 26 April 2011, *Zelski S.E. and H.A. Raja,* **PE0324-1.**

Specimens examined:

PERU. Cusco, Camanti, Quincemil, 3rd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0324-3;* 2nd stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, water 21.3 °C, pH 6, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0324-3*; 1st stream along Quincemil Trail 1, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, water 21.7 °C, pH 6.8, on submerged woody debris, 12 Apr 2011, *Zelski S.E. and H.A. Raja, PE0324-6.*Madre de Dios, CICRA, stream at Trail 19, 12°34’01.04”S, 70°05’43.24”W, 275m, water 23.7 °C, pH 5.1, on submerged decorticated wood, 9 Apr 2011, *Zelski S.E. and H.A. Raja, PE0324-1.*

Known distribution: Perú.

Comments:

Taxon PE0324 was initially linked to Annulatascaceae due to the presence of a large J- apical apparatus. A BLAST search of the LSU sequence in GenBank led to hits to taxa closely related to the Annulatascaceae and other members of the Sordariomycetes. The closest match was *Chaetosphaeria* sp., which led to a further investigation of the cause of the sequence similarity of these morphologically dissimilar taxa. The PE0324 28S sequence contained a Type II spliceosomal intron, a non-coding self-splicing element that has been found in 18S and 28S in many members of Ascomyceta (Bhattacharya et al. 2000). Removal of the 54 bp intron and reBLASTing the resulting sequence did not call up *Chaetosphaeria* sp., but matched *Bullimyces* and *Riomyces* closely. These taxa do not resemble PE0324 morphologically. In combined three gene analyses, PE0324 is far apart from them and is most closely related to PE0323, an *Ayria*-like species (**Fig. 7, Clade 7**). These taxa are also not similar

Fig. 77 A-1. ***Albumnitidomyces lubricosporus*** PE0324. A. Ascomata on surface of substrate. B. View of section of ascoma. C. Peridium. D-E. Ascus apices illustrating refractive apical rings. F. Ascus base. G.

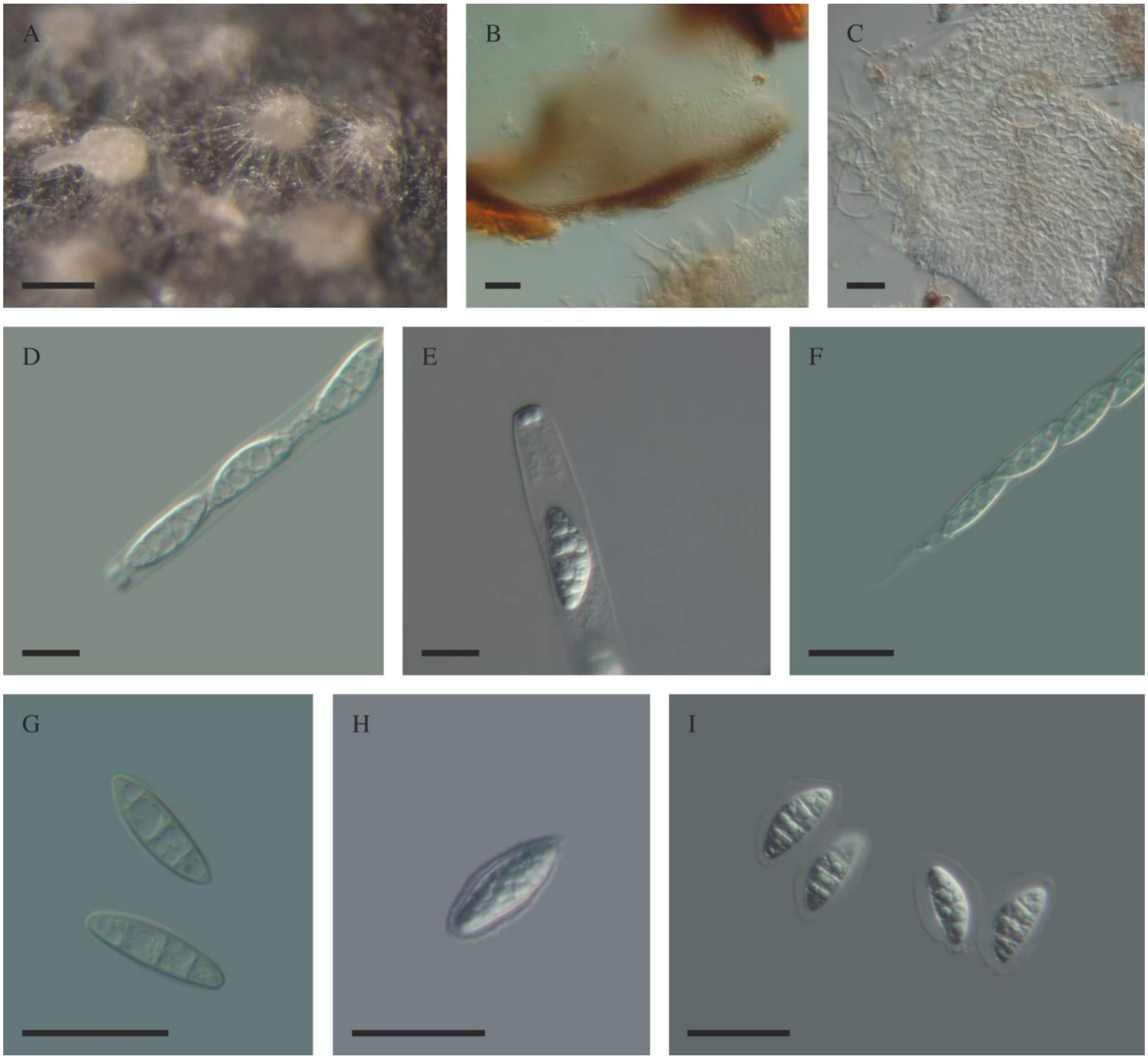


Fig. 77.

morphologically, with PE0323 ascospores having long, unfurling apical appendages and no sheath, while PE0324 ascospores have gelatinous sheaths. At present, PE0324 is best regarded as Sordariomycetes *incertae sedis*.

***Forsitannulatascus nodulosporus* gen. et. sp. nov. [PE0393]** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 78 A-G.**

Ascomata black, coriaceous, immersed, perpendicular to surface, scattered, globose to subglobose (**Fig. 78 A**). **Ascomal wall** black. **Neck** black, erumpent, to 500 µm long × 100 µm wide, periphysate (**Fig. 78 A**). **Paraphyses** hyaline, long, tapering, septate. **Asci** unitunicate, 8–spored, 180-200 µm long × 10-12 µm wide (x̄ = 190 × 11 µm, l × w = 17.3) (**Fig. 78 B-E**). **Ascus base** tapering pedicel, 30 µm from *pars sporifera* (**Fig. 78 E**). **Annulus** bipartite, 2.5 µm long × 5 µm wide (l × w = .5) (**Fig. 78 B-D**). **Ascospores** hyaline, ellipsoidal to ellipsoidal-fusiform, 3-septate, not constricted at septa, uniseriate to overlapping uniseriate, 20-22 µm long × 9-11 µm wide (x̄ = 21 × 10 µm, l × w = 2.1), guttulate, without appendages, with well-defined verrucose protrusions from the ascospore wall, staining in aqueous nigrosin (**Fig. 78 F,G**).

Specimens examined:

PERU. Junin, Central jungle near San Ramon, 118’33.5754”N, 75°20’35.304”W, 916m, on partially decorticated submerged woody debris, water 19 °C, pH 9.4, 147 µS, DO 7.1, *Zelski S.E. and J. Balto, PE0393-1.*

Comments: This species is a good candidate for a new species of Annulatascaceae. Unfortunately, it has not yet been isolated in culture as contamination with bacteria has occurred with germinating single ascospores in two attempts. AWA containing antibiotics other than Streptomycin and Penicillin may be required. Material is old and it is hoped that isolation can be performed from other collections in fresher

Fig. 78 A-G. ***Forsitannulatascus nodulosporus*** PE0393. A. Necks protruding through surface of substrate from immersed venters. B. Ascus apex with annulus seen end on. C. Ascus apex stained in aqueous nigrosin illustrating punctate/tubercular surface texture of ascospores. D. Ascus apex. E. Ascus base. F. Ascospores. G. Ascospore illustrating surface texture in water. Bars: A = 500 µm; B-G = 20 µm.

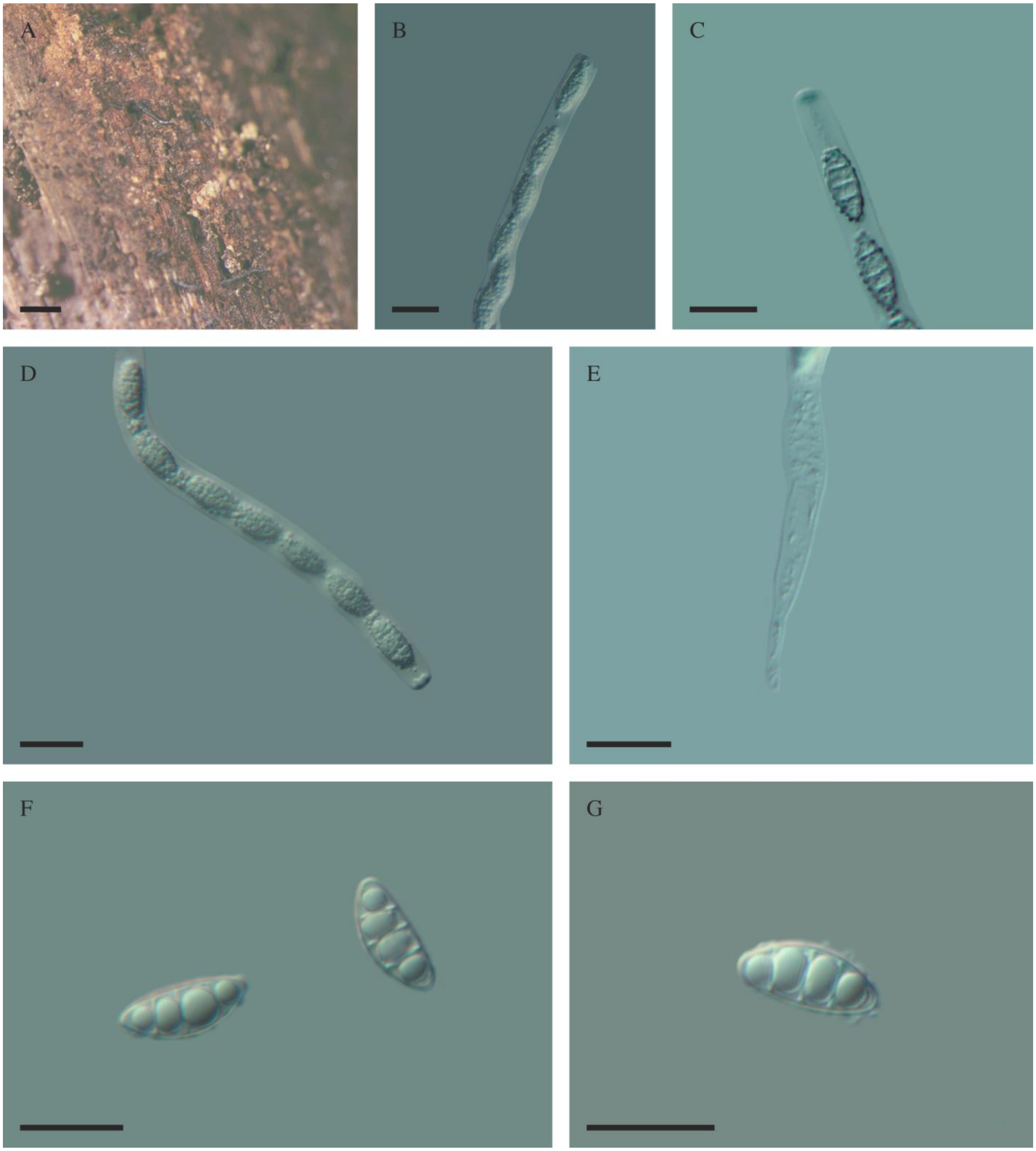


Fig. 78.

condition. The defining feature of this putative *Annulatascus* species is the prominent warty appearance of the ascospores. Ascomal morphology and ascus morphology require corroboration from molecular data to determine a phylogenetic placement for this fungus. At present it should be considered Sordariomycetes *incertae sedis*.

***Vertexicola triseptata* sp. nov.** Zelski, Raja, A.N. Mill. & Shearer. **Fig. 79 A-H.**

Ascomata black, coriaceous, semi-immersed to superficial, perpendicular to surface, solitary, 275–350 µm wide, globose to subglobose. **Ascomal wall** dark brown to black (**Fig. 79 A**). **Neck** black, 95–150 µm long × 40–65 µm wide, periphysate. **Paraphyses** hyaline, 5 µm wide at base, tapering toward apex, septate, constricted at septa. **Asci** unitunicate, cylindrical, 8–spored, 144–171 µm long × 7.5–9 µm wide ($\bar{x}$ = 155.6 × 8.3 µm, l:w = 18.75, n = 20) (**Fig. 79 B**). **Ascus base** tapering pedicel, with a spike-like tail, 20 µm from *pars sporifera* (**Fig. 79 B,C**). **Annulus** bipartite, 3-4 µm long × 4-5 µm wide (l:w = .8, n = 10) (**Fig. 79 E,F**). **Ascospores** hyaline, fusiform, 3–septate, not constricted at septa, uniseriate to slightly overlapping uniseriate, 17.25–22.4 µm long × 6–7.3 µm wide ($\bar{x}$ = 20.4 × 6.7 µm, l:w = 3, n = 60), guttulate, without appendages or sheath (**Fig. 79 G,H**).

Specimens examined: PERU. Cusco, Camanti, Quincemil, Quincemil Trail 1, semi-aquatic habitat, 13°14’22.5594”S, 70°46’12.6114”W, 688m, on submerged woody debris, 26 May 2010, *Zelski S.E. and H.A. Raja, PE0104-1;* Stream at Quincemil Trail 3, 13°18’27.756”S, 70°48’44.9274”W, 757m, on submerged woody debris, water 21.6 °C, pH 6.9, 27 May 2010, *Zelski S.E. and H.A. Raja, PE0104-4;* Semi-aquatic habitat along Quincemil Trail 1, on submerged woody debris, trailhead 13°14’22.5594”S, 70°46’12.6114”W, 688m, Oct 2010, *Zelski S.E. and H.A. Raja, PE0104-2;* Madre de Dios, CICRA, Stream at Trail 28, 12°34’02.81”S, 70°05’42.96”W, 272m, on submerged decorticated wood, water 22.7 °C, pH 5.9, 22 May 2010, *Zelski S.E. and H.A. Raja, PE0104-3.*

Known distribution: Perú.

Comments: Taxon PE0104, *Vertexicola triseptata*, was initially thought to be a collection of *V. caudatus*, though close examination revealed morphological differences. The new species, *V. triseptata*, has shorter, thinner asci than those of *V. triseptata* (144–171 µm long × 7.5–9 µm wide vs. 170–290 µm long × 7.5–12 µm wide). In addition, the ascospores of *V. triseptata* are slightly shorter and thinner on average than those of *V. caudatus* (21 × 7.5 vs. 20.4 ×6.7) and are 3–septate vs. 5–septate. Both morphology and molecular analyses support the establishment of a new species. In combined three gene analyses the two *Vertexicola* species are in a statistically well supported clade with different branch lengths (**Fig. 7, Clade 1e**). ITS analyses mirror this finding (**Fig. 4**). In a clade between Diaporthales and Ophiostomatales, *V. triseptata* should be considered Sordariomycetes *incertae sedis*.

Fig. 79 A-H. ***Vertexicola triseptata*** from specimen PE0104-1. A. Peridium. B. Ascus. C. Ascus base illustrating spike-like tail. D-F. Ascus apices. G-H. Ascospores. Bars: A-H = 20 µm.

Fig. 79.

PROPOSED NOMENCLATURAL CHANGES

Amphibioannulatascus citriosporus **gen. et comb. nov.** (J. Fröhl. & K.D. Hyde) Zelski, Raja, A.N. Mill. & Shearer.

≡ *Annulatascus citriosporus* J. Fröhl. & K.D. Hyd**e,** Palm Microfungi. Fungal Diversity Series 3: 79. 1993.

Insolensaquaticola triseptata **gen. et comb. nov.** (K.M. Tsui, Hodgkiss & K.D. Hyde) Zelski, Raja, A.N. Mill. & Shearer.

≡ *Aquaticola triseptata* K.M. Tsui, Hodgkiss & K.D. Hyde, Nova Hedwigia 77(1-2): 165, 2003.

Ascuslongus biatriisporus **gen. et comb. nov.** (K.D. Hyde) Zelski, Raja, A.N. Mill. & Shearer.

≡ *Annulatascus biatriisporus* K.D. Hyde**,** Nova Hedwigia 61: 120. 1995.

Breviascus miniguttulata **gen. et comb. nov.** (K.M. Tsui, Hodgkiss & K.D. Hyde) Zelski, Raja, A.N. Mill. & Shearer.

≡ *Aquaticola miniguttulata* K.M. Tsui, Hodgkiss & K.D. Hyde, Nova Hedwigia 77(1-2): 162, 2003

Fluviatilomyces ellipsoidea **gen. et comb. nov.** (W.H. Ho, K.M. Tsui, Hodgkiss & K.D. Hyde) Zelski, Raja, A.N. Mill. & Shearer.

≡ *Aquaticola ellipsoidea* W.H. Ho, K.M. Tsui, Hodgkiss & K.D. Hyde, Fungal Diversity 3: 90. 1999.

Pseudosubmersisphaeria rattanicola **gen. et comb. nov.** (J. Fröhl. & K.D. Hyde) Zelski, Raja, A.N. Mill. & Shearer.

≡ *Submersisphaeria rattanicola* J. Fröhl. & K.D. Hyde, Palm Microfungi: 82, 2000.

Pseudotorrentispora crassiparietis **gen. et comb. nov.** (Fryar & K.D. Hyde) Zelski, Raja, A.N. Mill. & Shearer.

≡ *Torrentispora crassiparietis* Fryar & K.D. Hyde, Cryptogamie Mycologie 25(3): 255. 2004.

CHAPTER 6

CONCLUSION

Freshwater ascomycetes are a morphologically and genetically diverse ecological group of fungi. This too holds true in this survey of species referred to the family Annulatascaceae which also appear to have multiple origins from terrestrial taxa. In this case, a natural classification based on morphology and backed by molecular data is problematic because there are few morphological characters or suites of characters with which to group taxa when molecular evidence is included. Suites of characters are problematic as they are subject to overlap. At present, molecular analyses are the gold standard when it comes to determining the monophyly of particular groups and subsequently morphology is layered onto those hypotheses. Using a limited number of genes may present a problem, and perhaps a genomic approach will afford a solution. Even so, the genomic approach may give rise to new challenges in terms of data handling, analyses and interpretation. More information may contribute more confounding factors. The systematics of freshwater ascomycetes with prominent ascus apical rings, be they bipartite or not, appears fraught with homoplasy. It is with further molecular work that phylogenetic relationships will be refined due to the limited morphological characters available for Annulatascaceae-like fungi.

A J- ascus apical ring is a feature that initially suggests possible affinity to Annulatascaceae, but too many other ascomycetes possess this feature for it to be phylogenetically informative. This character must be assessed in combination with the shared features of the family mentioned above, and molecular data is needed for confirmation of that placement. From a morphological perspective, the Annulatascaceae database constructed during the course of this study is an invaluable tool for comparing taxa and performing diagnoses. Molecularly, the newly generated sequences from this study will aid future investigations by filling in gaps in our knowledge about the evolutionary history of these organisms. In future, taxa should not formally be placed in Annulatascaceae (Index Fungorum, GenBank, MycoBank) without accompanying molecular support, rather they should be considered Sordariomycetes *incertae sedis* unless supported at a higher taxonomic level.

TABLES

Table 1. Site details of collections made in the course of this study.

Country	Location	Northing	Easting	Elev. (m)	pH	Temp. (°C)	Date	Collectors
U.S.	IL	40°4’23.124”N	87°49’27.12”W	190	7.5	-	9/1/09	Zelski, Raja, & Miller
U.S.	IL	40°4’23.124”N	87°49’27.12”W	190	8.3	13.0	10/2/09	Zelski & Raja
U.S.	TN	35°35’37.05”N	83°46’19.06”W	520	-	-	10/20/09	Zelski & Hustad
U.S.	TN	35°45’20.90”N	83°12’28.44”W	675	-	-	10/21/09	Zelski & Hustad
U.S.	NC	35°35’50.10”N	83°05’10.25”W	802	-	-	10/21/09	Zelski & Hustad
U.S.	TN	35°39’27.69”N	83°34’49.57”W	659	-	-	10/22/09	Zelski & Hustad
U.S.	TN	35°45’35.48”N	83°06’19.76”W	494	-	-	10/22/09	Zelski & Hustad
U.S.	IL	40°4’23.124”N	87°49’27.12”W	190	7	-	11/5/09	Zelski
U.S.	IL	40°4’23.124”N	87°49’27.12”W	190	10.4	2.1	2/4/10	Zelski
Perú	Madre de Dios	12^{0}42’48.09”S	69^{0}28’11.28”W	242	5.9	23.3	5/20/10	Zelski & Raja
Perú	Madre de Dios	12^{0}34’06.52”S	70^{0}06’04.57”W	281	-	-	5/22/10	Zelski & Raja
Perú	Madre de Dios	12^{0}33’48.95”S	70^{0}05’48.47”W	287	5.6	22.3	5/22/10	Zelski & Raja
Perú	Madre de Dios	12^{0}33’48.95”S	70^{0}05’48.47”W	287	6.3	22.2	5/22/10	Zelski & Raja
Perú	Madre de Dios	12^{0}34’02.81”S	70^{0}05’42.96”W	272	5.9	22.7	5/22/10	Zelski & Raja
Perú	Madre de Dios	12^{0}34’02.81”S	70^{0}05’42.96”W	275	5.2	23.6	5/22/10	Zelski & Raja
Perú	Madre de Dios	12^{0}34’02.86”S	70^{0}04’56.26”W	218	-	-	5/22/10	Zelski & Raja
Perú	Madre de Dios	12^{0}33’34.27”S	70^{0}06’38.00”W	243	-	-	5/22/10	Zelski & Raja
Perú	Madre de Dios	12^{0}34’17.70”S	70^{0}05’23.69”W	241	6.7	23	5/23/10	Zelski & Raja
Perú	Madre de Dios	12^{0}34’17.70”S	70^{0}05’23.69”W	230	6.4	23.2	5/23/10	Zelski & Raja
Perú	Madre de Dios	12^{0}34’03.25”S	70^{0}04’55.92”W	218	7.9	25.3	5/24/10	Zelski & Raja
Perú	Madre de Dios	12^{0}33’51.05”S	70^{0}04’47.42”W	218	7.9	25.3	5/24/10	Zelski & Raja
Perú	Madre de Dios	12^{0}33’46.48”S	70^{0}04’41.81”W	218	7.9	25.3	5/24/10	Zelski & Raja

Table 1 continued.

Country	Location	Northing	Easting	Elev. (m)	pH	Temp. (°C)	Date	Collectors
Perú	Cusco	13^{0}14’22.56”S	70^{0}46’12.61”W	688	6.3	21	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}13’58.26”S	70^{0}46’37.78”W	675	7.2	22.2	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}14’22.56”S	70^{0}46’12.61”W	688	7.1	21.2	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}14’22.56”S	70^{0}46’12.61”W	688	6.8	21.2	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}14’22.56”S	70^{0}46’12.61”W	688	6	21	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}14’22.56”S	70^{0}46’12.61”W	688	5.5	21.2	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}14’22.56”S	70^{0}46’12.61”W	688	-	-	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}13’40.40”S	70^{0}45’14.18”W	659	5.3	22.8	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}13’34.07”S	70^{0}45’12.67”W	669	6.2	21.8	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}13’31.04”S	70^{0}45’10.62”W	653	6.4	21.9	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}13’31.04”S	70^{0}45’10.62”W	653	6.5	21.9	5/26/10	Zelski & Raja
Perú	Cusco	13^{0}18’27.76”S	70^{0}48’44.93”W	757	6	20.7	5/27/10	Zelski & Raja
Perú	Cusco	13^{0}18’27.76”S	70^{0}48’44.93”W	757	6.9	21.6	5/27/10	Zelski & Raja
Perú	Cusco	13^{0}18’27.76”S	70^{0}48’44.93”W	757	7.5	21.3	5/27/10	Zelski & Raja
Perú	Cusco	13^{0}18’27.76”S	70^{0}48’44.93”W	757	7.4	22.3	5/27/10	Zelski & Raja
Perú	Cusco	13^{0}18’27.76”S	70^{0}48’44.93”W	757	7.4	21.1	5/27/10	Zelski & Raja
Perú	Cusco	13^{0}18’43.13”S	70^{0}48’44.82”W	817	7.6	20.3	5/27/10	Zelski & Raja
Perú	Cusco	13^{0}17’07.01”S	70^{0}47’13.63”W	737	7.6	21.7	5/27/10	Zelski & Raja
Perú	Cusco	13^{0}27’04.39”S	70^{0}54’11.38”W	1372	7.6	15	5/28/10	Zelski & Raja
Perú	Cusco	13^{0}35’23.32”S	70^{0}57’21.89”W	2562	8.3	9.7	5/28/10	Zelski & Raja
U.S.	NC	35°30’7.2354”N	83°17’49.56”W	622	8.5	21.8	7/16/10	Zelski & Raja
U.S.	NC	35°34’41.34”N	83°20’43.5474”W	779	7.7	17.8	7/16/10	Zelski & Raja
U.S.	NC	35°35’6”N	83°21’28.7274”W	868	7.4	17.4	7/16/10	Zelski & Raja
U.S.	TN	35°37’15.672”N	83°25’25.3554”W	1330	7.2	14.6	7/17/10	Zelski & Raja
U.S.	TN	35°37’57.108”N	35°37’57.108”W	1144	7.5	15.4	7/17/10	Zelski & Raja
U.S.	TN	35°40’32.8434”N	83°31’37.848”W	536	7.4	20.0	7/17/10	Zelski &

Table 1 continued.

Country	Location	Northing	Easting	Elev. (m)	pH	Temp. (°C)	Date	Collectors
								Raja
U.S.	CO	39°58’5.916”N	105°58’3.1074”W	2832	8.4	5.1	8/13/10	Zelski & Hustad
U.S.	CO	40°15’16.1634”N	106°4’48.252”W	2604	8.5	11.2	8/13/10	Zelski & Hustad
U.S.	CO	40°2’47.976”N	106°2’27.024”W	2818	8.9	6.8	8/13/10	Zelski & Hustad
U.S.	CO	39°53’29.904”N	105°48’44.9274”W	2807	8.5	11.8	8/13/10	Zelski & Hustad
U.S.	CO	39°53’28.68”N	105°49’55.92”W	2822	8.1	6.3	8/14/10	Zelski & Hustad
U.S.	WI	43°46’15.07”N	89°53’10.38”W	340	8	20	9/12/10	Zelski & Hustad
Perú	Madre de Dios	12^{o}34’02.81”S	70^{o}05’42.96”W	275	6.1	23.8	9/30/10	Zelski & Raja
Perú	Madre de Dios	12^{o}34’17.70”S	70^{o}05’23.69”W	241	7.3	25.1	9/30/10	Zelski & Raja
Perú	Madre de Dios	12^{o}34’17.08”S	70^{o}05’47.60”W	244	7.7	25.1	9/30/10	Zelski & Raja
Perú	Madre de Dios	12^{o}33’34.27”S	70^{o}06’38.00”W	243	6.8	31.7	9/30/10	Zelski & Raja
Perú	Madre de Dios	12^{o}34’02.81”S	70^{o}05’42.96”W	272	6.8	23.3	9/30/10	Zelski & Raja
Perú	Madre de Dios	12^{o}33’25.22”S	70^{o}05’59.89”W	288	6.6	23.5	9/30/10	Zelski & Raja
Perú	Madre de Dios	12^{o}33’48.95”S	70^{o}05’48.47”W	287	6.8	23.6	9/30/10	Zelski & Raja
Perú	Madre de Dios	12^{o}34’31.80”S	70^{o}04’28.85”W	218	8	31.4	10/1/10	Zelski & Raja
Perú	Madre de Dios	12^{o}34’13.01”S	70^{o}04’14.77”W	218	8	31.4	10/1/10	Zelski & Raja
Perú	Madre de Dios	12^{o}34’00.34”S	70^{o}04’59.05”W	218	8	31.4	10/1/10	Zelski & Raja
Perú	Cusco	13^{o}13’58.26”S	70^{o}46’37.78”W	675	8.3	19.6	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	688	8	19	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	689	7.7	19.2	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	690	6.7	19.1	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	691	-	-	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	692	5.8	20	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	693	5.8	19.7	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}13’20.87”S	70^{o}44’30.03”W	626	7.2	24.9	10/4/10	Zelski & Raja
Perú	Cusco	13^{o}13’40.40”S	70^{o}45’10.62”W	659	7.4	24	10/5/10	Zelski & Raja

Table 1 continued.

Country	Location	Northing	Easting	Elev. (m)	pH	Temp. (°C)	Date	Collectors
Perú	Cusco	13^{o}13’31.04”S	70^{o}45’10.62”W	653	7.2	24.9	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	757	7.2	20.5	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	757	7.1	21.6	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	757	7.5	21.4	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	757	7.5	21.5	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	757	7.5	21.7	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	757	7.7	21	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}17’07.01”S	70^{o}47’13.63”W	737	7.7	22	10/5/10	Zelski & Raja
Perú	Cusco	13^{o}27’52.20”S	70^{o}53’52.44”W	1463	8.2	15.3	10/6/10	Zelski & Raja
Perú	Cusco	13^{o}34’28.99”S	71^{o}01’13.55”W	3421	8.3	17.9	10/6/10	Zelski & Raja
Perú	Cusco	13^{o}37’40.37”S	71^{o}24’23.94”W	3566	8.4	17.4	10/6/10	Zelski & Raja
Perú	Madre de Dios	12^{o}33’34.27”S	70^{o}06’38.00”W	243	7.9	25.4	4/9/11	Zelski & Raja
Perú	Madre de Dios	12^{o}33’25.22”S	70^{o}05’59.89”W	288	8.3	23.1	4/9/11	Zelski & Raja
Perú	Madre de Dios	12^{o}33’48.95”S	70^{o}05’48.47”W	287	7.8	23.3	4/9/11	Zelski & Raja
Perú	Madre de Dios	12^{o}34’06.52”S	70^{o}06’04.57”W	281	6.7	23.6	4/9/11	Zelski & Raja
Perú	Madre de Dios	12^{o}34’02.81”S	70^{o}05’42.96”W	275	5.1	23.7	4/9/11	Zelski & Raja
Perú	Madre de Dios	12^{o}34’17.08”S	70^{o}05’47.60”W	244	6.7	23.5	4/9/11	Zelski & Raja
Perú	Cusco	13^{o}13’20.87”S	70^{o}44’30.03”W	626	8	25.3	4/12/11	Zelski & Raja
Perú	Cusco	13^{o}13’58.26”S	70^{o}46’37.78”W	675	7.7	21.5	4/12/11	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	688	7.8	21.4	4/12/11	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	689	6	21.3	4/12/11	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	690	6	21.3	4/12/11	Zelski & Raja
Perú	Cusco	13^{o}14’22.56”S	70^{o}46’12.61”W	691	6.8	21.7	4/12/11	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	757	6.8	21.7	4/13/11	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	758	7	21.5	4/13/11	Zelski & Raja
Perú	Cusco	13^{o}18’27.76”S	70^{o}48’44.93”W	759	7.8	21.2	4/13/11	Zelski &

Table 1 continued.

Country	Location	Northing	Easting	Elev. (m)	pH	Temp. (°C)	Date	Collectors
								Raja
Perú	Cusco	13^{0}18’27.76”S	70^{0}48’44.93”W	760	7.1	22.2	4/13/11	Zelski & Raja
Perú	Cusco	13^{0}18’27.76”S	70^{0}48’44.93”W	761	7.2	21.8	4/13/11	Zelski & Raja
Perú	Cusco	13^{0}21’19.91”S	71^{0}36’33.52”W	3182	8.4	13.9	4/14/11	Zelski & Raja
Perú	Cusco	13^{0}21’26.10”S	71^{0}37’59.48”W	3244	8.4	12.4	4/14/11	Zelski & Raja
Perú	Cusco	13^{0}21’02.41”S	71^{0}39’22.00”W	3327	8.3	11.6	4/14/11	Zelski & Raja
Perú	Cusco	13^{0}21’00.92”S	71^{0}40’40.30”W	3571	8.2	14.8	4/14/11	Zelski & Raja
Perú	Cusco	13^{0}21’00.97”S	71^{0}40’40.30”W	3870	8.5	11.6	4/14/11	Zelski & Raja
Perú	Cusco	13^{0}24’21.73”S	71^{0}49’53.29”W	3207	8.6	12.8	4/14/11	Zelski & Raja
U.S.	CO	37°47’1.1754”N	107°50’42.9354”W	3619	9.8	12.6	8/23/11	Zelski
U.S.	CO	37°47’36.6354”N	107°50’49.4154”W	3344	8.9	9.9	8/23/11	Zelski
U.S.	CO	37°47’25.4394”N	107°50’47.5794”W	3317	6.7	5.4	8/23/11	Zelski
U.S.	CO	37°48’17.208”N	107°50’49.452”W	3325	7.7	6.2	8/23/11	Zelski
U.S.	CO	37°49’19.4514”N	107°52’28.272”W	3010	8.4	6.2	8/25/11	Zelski
U.S.	CO	37°49’15.7794”N	107°52’23.3394”W	3058	8.2	6.8	8/25/11	Zelski
U.S.	CO	37°49’11.856”N	107°52’29.4594”W	3040	8.3	9.4	8/25/11	Zelski
U.S.	CO	37°49’19.596”N	107°52’32.592”W	3058	8.3	12.6	8/25/11	Zelski
U.S.	CO	37°49’21.54”N	107°52’37.0554”W	3078	8.2	9.9	8/25/11	Zelski
Perú	Junin	11°5’53.6094”S	74°17’0.564”W	1059	9	19.5	5/20/12	Zelski & Balto
Perú	Junin	10°58’45.8754”S	74°16’14.9154”W	1334	8.3	23.3	5/20/12	Zelski & Balto
Perú	Junin	10°57’43.344”S	74°14’4.4874”W	919	9.3	19.9	5/20/12	Zelski & Balto
Perú	Junin	11°22’33.6318”S	74°32’54.24”W	917	9	21	5/21/12	Zelski & Balto
Perú	Junin	11°23’10.4274”S	74°35’27.3114”W	1085	8.9	17.9	5/21/12	Zelski & Balto
Perú	Junin	11°22’53.58”S	74°35’37.989”W	1116	8.7	21.3	5/21/12	Zelski & Balto
Perú	Junin	11°20’1.7154”S	74°37’36.192”W	891	9	22.1	5/21/12	Zelski & Balto
Perú	Junin	11°24’21.276”S	74°45’49.1034”W	1330	8.9	18.7	5/21/12	Zelski & Balto
Perú	Junin	11°24’0”S	74°28’39.324”W	670	9	23.7	5/23/12	Zelski & Balto
Perú	Junin	11°23’41.748”S	74°26’58.1274”W	637	9.2	22.9	5/23/12	Zelski & Balto
Perú	Junin	11°22’1.056”S	74°26’22.92”W	687	9.4	22.3	5/23/12	Zelski & Balto

Table 1 continued.

Country	Location	Northing	Easting	Elev. (m)	pH	Temp. (°C)	Date	Collectors
Perú	Junin	11°22’40.0074”S	74°24’10.1874”W	1128	9.2	20.9	5/23/12	Zelski & Balto
Perú	Junin	11°22’25.4634”S	74°23’2.292”W	1248	9.3	20.9	5/23/12	Zelski & Balto
Perú	Junin	11°22’15.9954”S	74°22’1.7394”W	1400	9.2	18.1	5/24/12	Zelski & Balto
Perú	Junin	11°28’24.9594”S	74°33’40.6434”W	1300	9.1	18.4	5/24/12	Zelski & Balto
Perú	Junin	11°17’17.268”S	74°32’23.9274”W	696	9.6	21.1	5/25/12	Zelski & Balto
Perú	Junin	11°14’10.932”S	74°41’41.8914”W	650	9.3	22.7	5/25/12	Zelski & Balto
Perú	Junin	11°13’13.6194”S	74°35’36.3474”W	613	9.1	21	5/25/12	Zelski & Balto
Perú	Junin	11°11’47.1474”S	74°42’3.888”W	814	9	21.7	5/25/12	Zelski & Balto
Perú	Junin	11°10’24.42”S	74°42’50.616”W	936	8.9	20.6	5/25/12	Zelski & Balto
Perú	Junin	10°59’28.1034”S	74°46’13.512”W	597	9.4	22.6	5/27/12	Zelski & Balto
Perú	Junin	10°58’35.2194”S	74°47’5.0634”W	527	9.3	22.6	5/27/12	Zelski & Balto
Perú	Junin	10°52’20.352”S	74°59’35.2674”W	529	9.4	23.4	5/27/12	Zelski & Balto
Perú	Junin	11°16’23.00”S	74°28’55.83”W	1803	9.1	20.4	5/29/12	Zelski & Balto
Perú	Junin	10°28’40.8354”S	74°6’35.532”W	1670	9.5	23.4	5/29/12	Zelski & Balto
Perú	Junin	11°5’43.5834”S	75°21’7.668”W	968	9.7	19.1	5/30/12	Zelski & Balto
Perú	Junin	11°8’33.5754”S	75°20’35.304”W	916	9.4	19	5/31/12	Zelski & Balto
Perú	Junin	11°12’17.424”S	75°20’8.196”W	1002	9.5	20.2	5/31/12	Zelski & Balto
Perú	Junin	11°5’42.6474”S	75°26’6.324”W	2117	9.5	14.3	6/1/12	Zelski & Balto
Perú	Junin	11°6’31.0674”S	75°24’42.84”W	1520	9.5	16.9	6/1/12	Zelski & Balto
Perú	Junin	11°6’31.5714”S	75°24’43.02”W	1517	9.4	16.7	6/1/12	Zelski & Balto
Thailand	Chiang Mai	19°7’4.512”N	99°44’2.2194”E	904	7.6	23.5	6/15/12	Zelski
Thailand	Chiang Mai	18°48’24.4794”N	98°54’38.3754”E	1149	7.2	22.4	6/18/12	Zelski
Thailand	Chiang Mai	19°14’48.9834”N	98°39’0.2514”E	994	7.9	23.4	6/22/12	Zelski
Thailand	Chiang Mai	19°4’10.848”N	99°4’46.8834”E	508	7.2	22.6	6/23/12	Zelski
Thailand	Chiang Mai	19°14’32.172”N	98°41’3.408”E	795	-	-	6/26/12	Zelski
Thailand	Chiang Mai	19°06’37.61”N	98°44’17.20”E	1020	6.8	22.8	6/27/12	Zelski
Thailand	Chiang Mai	18°55’31.9074”N	99°21’25.236”E	1137	-	-	7/5/12	Zelski
Laos	Sibouheuang	20°21’12.0954”N	100°4’56.928”E	385	7.8	26	7/9/12	Zelski
U.S.	CT	41°25’54.624”N	72°54’18.0714”W	93	9.7	-	7/17/12	Zelski &

Table 1 continued.

Country	Location	Northing	Easting	Elev. (m)	pH	Temp. (°C)	Date	Collectors
								Hustad
U.S.	PA	41°7’42.744”N	76°21’26.6034”W	191	9.5	-	7/19/12	Zelski & Hustad
U.S.	PA	41°20’1.356”N	76°16’15.4194”W	644	9	-	7/19/12	Zelski & Hustad
U.S.	OH	41°32’12.8034”N	81°31’11.748”W	226	9.4	-	7/20/12	Zelski & Hustad
U.S.	CO	39°23’42.468”N	105°53’58.164”W	2692	8.5	9.8	8/13/12	Zelski & Hustad
U.S.	IL	40°27’17.83”N	90°47’41.75”W	186	6.7	17.2	9/22/12	Zelski & Hustad
U.S.	IL	40°22’23.23”N	91°23’39.52”W	149	7.3	20	9/22/12	Zelski & Hustad
U.S.	IL	40°22’23.23”N	91°23’39.52”W	149	7.3	20	9/22/12	Zelski & Hustad
U.S.	AK	36°26’27.03”N	94°2’9.18”W	585	-	-	10/20/12	Zelski & Hustad
U.S.	AK	36°25’10.63”N	94°3’20.19”W	366	-	-	10/21/12	Zelski & Hustad
U.S.	WI	42°29’18.78”N	87°55’31.83”W	204	-	-	7/28/13	Zelski

Table 2 continued.

Table 2. Herbarium locations of holotypes of Annulatascaceae taxa

Taxon	Herbarium	Details
Annulatascus apiculatus F.R. Barbosa & Gusmão	HUEFS	BRAZIL. Bahia, Santa Terzinha, Serra da Jibóa, stream, on submerged wood, 19 Feb 2008, F.R. Barbosa, (HUEFS 134723).
Annulatascus aquaticus W.H. Ho, K.D. Hyde & Hodgkiss	HKU	CHINA. Hong Kong. Tai Po Forest Stream, on submerged decaying wood, 27 Jun 1996, K.D. Hyde, WH238 (HKU(M) 4526).
Annulatascus aquatorba Boonyuen & Sri-indrasutdhi	BBH	THAILAND. Narathiwat Province, Sirindhorn Peat Swamp Forest, on submerged wood test block of *Erythrophleum teysmannii*, 22 Feb 2003, N. Boonyuen & V. Sri-indrasutdhi (BBH 29936).
Annulatascus biatriisporus K.D. Hyde	BRIP	AUSTRALIA. Northern Queensland, Babinda, The Boulders, on submerged wood, Dec 1991, K.D. Hyde, (880) (BRIP 21481).
Annulatascus citriosporus J. Fröhl. & K.D. Hyde	HKU	BRUNEI DARUSSALAM. Temburong, Batu Apoi Forest Reserve, Sungai Belalong, Kuala Belalong Field Studies Centre (KBFSC), near start of Ashton's Trail, on dead petiole of *Licaula* sp., Dec 1993, J. Fröhlich, (HKU(M) JF LI 9).
Annulatascus fusiformis K.D. Hyde & S.W. Wong	HKU	PHILIPPINES. Mindanao, Bukidnon, Impalutao, Natigbasan Creek, on submerged wood, Jan 1994, K.D. Hyde, (HKU(M) 3102).

Table 2 continued.

Taxon	Herbarium	Details
Annulatascus hongkongensis W.H. Ho, Ranghoo, K.D. Hyde & Hodgkiss	HKU	CHINA. Hong Kong. Plover Cove Reservoir, on submerged decomposing wood, 15 Nov 1996, K D. Hyde & M. Wong, (PC 21) (HKU(M) 4702).
Annulatascus joannae K.M. Tsui, Hodgkiss & K.D. Hyde	HKU	CHINA. Hong Kong, Tai Po, Lam Tsuen River, on submerged wood, Dec 1998, K.M. Tsui, (KM 164) (HKU(M) 12177).
Annulatascus lacteus K.M. Tsui, Hodgkiss & K.D. Hyde	HKU	CHINA. Hong Kong, Tai Po, Lam Tsuen River, on submerged wood, Sep 1996, K.M. Tsui, (KM 313) (HKU(M) 4623).
Annulatascus licualae J. Fröhl. & K.D. Hyde	HKU	AUSTRALIA. North Queensland, Kurunda, Saddle Mountain, palm glade, on dead petiole of *Licuala ramsayi*, Mar 1994, J. Fröhlich, (HKU(M) JF LV 5).
Annulatascus liputii L. Cai & K.D. Hyde	PDD	PHILIPPINES. Negros Occidental, Bario Alegria, Lupit River, on submerged bamboo, 18 Aug 2001, V.A. Hyde, (PDD 75038).
Annulatascus menglensis D.M. Hu, L. Cai & K.D. Hyde	IFRD	CHINA. Yunnan Province, Mengla, Wudaoban Stream, 21°24’N 101°36’E, alt: 660m, 3 April 2009, D.M. Hu, IFRDC 023-002.
Annulatascus nilensis Abdel-Wahab & Abdel-Aziz	IMI	EGYPT. Sohag, River Nile, on decayed submerged stems of *Phragmites australis*, Feb 2006, A.E. Abdel-Aziz, (IMI 397966).
Annulatascus palmietensis Goh, K.D. Hyde & Steinke	HKU	SOUTH AFRICA. Durban, Palmiet River, on submerged wood, Nov 1994, K.D. Hyde &

Table 2 continued.

Taxon	Herbarium	Details
		T.D. Steinke, (SAPR 43) (HKU(M) 2206).
Annulatascus triseptatus S.W. Wong, K.D. Hyde & E.B.G. Jones	HKU	BRUNEI. Temburong, Kuala Belalong Field Studies Centre, Sungai Anak, on submerged wood, Feb 1994, K. D. Hyde and S. W. Wong, (HKU(M) 3129).
Annulatascus tropicalis Ranghoo & K.D. Hyde	HKU	CHINA. Hong Kong, Tai Po, Plover Cove Reservoir, on submerged wood, Jan 1997, V.M. Ranghoo, (HKU(M) 5253).
Annulatascus velatisporus K.D. Hyde	BRIP	AUSTRALIA. North Queensland. Millaa Millaa Falls, on submerged wood, Jul 1990, Hyde K. D., K.D. HYDE 360B BRIP 17373.
Aqualignicola hyalina Ranghoo, K.M. Tsui & K.D. Hyde	HKU	CHINA. Hong Kong, New Territories, Tai Po, the Lam Tsuen River, on submerged wood, 9 Dec 1998, K. M. Tsui, (KM 307) (HKU(M) 12178).
Aqualignicola vaginata D.M. Hu, L. Cai & K.D. Hyde	IFRD	China, Yunnan Province, Mengla, Wudaoban Stream, 21°32’N, 101°29’E, alt. 620m, 2 April 2009, D.M. Hu, IFRDC 021-043.
Aquaticola ellipsoidea W.H. Ho, K.M. Tsui, Hodgkiss & K.D. Hyde	HKU	CHINA. Hong Kong, Tai Po, Tai Po Kau Forest Stream, on *Machilus velutina* wood bait, 29 Mar 1997, W.H. Ho, (HKU(M) 6033).
Aquaticola hongkongensis Ranghoo, K.D. Hyde & E.C.Y. Liew	HKU	CHINA. Hong Kong, New Territories, Plover Creek Reservoir, on submerged wood, Jan 1997, V.M. Ranghoo,

Table 2 continued.

Taxon	Herbarium	Details
		(HKU(M) 5254).
Aquaticola hyalomura W.H. Ho, K.M. Tsui, Hodgkiss & K.D. Hyde	HKU	CHINA. Hong Kong, Tai Po Kau Forest Stream, on submerged decaying wood, 27 Jun 1996, K.D. Hyde, (WH189) (HKU(M) 2969).
Aquaticola longicolla K.M. Tsui, Hodgkiss & K.D. Hyde	HKU	AUSTRALIA. North Queensland, near Ravenshoe, ~ 20 km along Tully Falls Road to Koombooloomba Dam, 17°45’S, 145°35’E, submerged in creek, Mar 1997, K.D. Hyde, (RH 49) (HKU(M) 5159) (HKU(M) 5148, syntype).
Aquaticola miniguttulata K.M. Tsui, Hodgkiss & K.D. Hyde	HKU	CHINA. Hong Kong, Sai Kung, Hang Cho Shui, on submerged wood, Sep 1998, K.M. Tsui & Edmund Shek, (KM52) (HKU(M) 12275).
Aquaticola triseptata K.M. Tsui, Hodgkiss & K.D. Hyde	HKU	CHINA. Hong Kong, Tai Po, Lam Tsuen River, on submerged wood, Dec 1998, K.M. Tsui & Ginimi Chan, (KM111) (HKU(M) 12222).
Ascitendus austriacus (Réblová, Winka & Jaklitsch) J. Campb. & Shearer	PRM	AUSTRIA. Wien 19, Hermannskogel, on submerged wood of *Fagus sylvatica*, 29 Aug 1998, W. Jaklitsch, (WJ 1131-98) (PRM 842991).
Ascolacicola aquatica Ranghoo & K.D. Hyde	HKU	CHINA. Hong Kong. New Territories. Plover Creek Reservoir, on submerged wood, Mar 1997, V.M. Ranghoo, (HKU(M) 5243).

Table 2 continued.

Taxon	Herbarium	Details
Ayria appendiculata Fryar & K.D. Hyde	HKU	BRUNEI. Sungai Kelakas, Tutong River, Site 2, on submerged rotting wood, 1998, S .C. Fryar, (HKU(M) 15553).
Ayria nubispora Raja, Ferrer & Searer	ILL	USA. Florida. Marion County, Ocala National Forest, Fore Lake, 29°16’15”N, 81°55’02”W, water 18 C, pH 7, on submerged woody debris, 9 Feb 2006, Huzefa A. Raja and J.L. Crane, F99-1, ILL 40594.
Brunneosporella aquatica Ranghoo & K.D. Hyde	HKU	CHINA. Hong Kong. New Territories, Plover Cove Reservoir, on submerged wood, Jan 1997, V.M. Ranghoo, (PC 60) (HKU(M) 5251).
Cataractispora appendiculata K.D. Hyde, S.W. Wong & E.B.G. Jones	HKU	BRUNEI. Temburong, Kuala Belalong Field Studies Centre, Sungai Esu, on submerged wood, Feb 1994, K.D. Hyde & S.W. Wong, (HKU(M) 3120).
Cataractispora aquatica K.D. Hyde, S.W. Wong & E.B.G. Jones	HKU	BRUNEI. Temburong, Kuala Belalong Field Studies Centre, Sungai Esu, on submerged wood, Feb 1994, K.D. Hyde & S.W. Wong (HKU(M) 3123).
Cataractispora bipolaris (K.D. Hyde) K.D. Hyde, S.W. Wong & E.B.G. Jones	BRIP	AUSTRALIA. North Queensland, Clohesy River, on wood submerged in a river, Nov 1990, K.D. Hyde, (BRIP 17374). Ex. *Annulatascus bipolaris*.
Cataractispora receptaculorum W.H. Ho, K.D. Hyde & Hodgkiss	HKU	CHINA. Hong Kong, Plover Cove Reservoir, on bamboo submerged in freshwater, 15 Feb 1997, V.M. Ranghoo, (PC38) (HKU(M) 5239).

Table 2 continued.

Taxon	Herbarium	Details
Cataractispora viscosa K.D. Hyde, S.W. Wong & E.B.G. Jones	HKU	CHINA. Hong Kong. New Territories, Tai Po Kau Country Park, on twigs submerged in a stream, Sep 1993, S. W. Wong & K.D. Hyde, (HKU(M) 3130).
Chaetorostrum quincemilense Zelski, Raja, A.N. Mill & Shearer	ILL	PERU, CAMANTI: Stream at Quincemil Trail 1, 13˚14'23"S, 70˚46'13"W, on submerged woody debris, 26 May 2010, Zelski S.E. and H.A. Raja, PE105-1, ILL 40822.
Clohiesia corticola K.D. Hyde	BRIP	AUSTRALIA. North Queensland, Davies Creek National Park, Davies Creek, on submerged wood, Oct 1990, K.D. Hyde, (364b) (BRIP 21485).
Clohiesia curvispora L. Cai & K.D. Hyde	HKU	CHINA. Yunnan, Jinghong, on submerged wood in a small forest stream, 15 Sep 2002, L. Cai, (CAI-9BNA38) (HKU(M) 10854).
Clohiesia lignicola K.M. Tsui, K.D. Hyde & Hodgkiss	HKU	CHINA. Hong Kong, Lantau Island, Tung Chung River, on submerged wood, 28 Jul 1997, K.M. Tsui, (KM 202) (HKU(M) 5539).
Cyanoannulus petersenii Raja, J. Campb. & Shearer	ILL	USA. North Carolina. Great Smoky Mountain National Park, Ocanaluftee River, 35°34’12.06”N, 83°20’7.38”W, water 20 °C, pH 5, on submerged decorticated wood, 3 Jul 2005, H.A. Raja and Nate Hamburger, R44-1, ILL 40101.

Table 2 continued.

Taxon	Herbarium	Details
Diluviicola capensis K.D. Hyde, S.W. Wong & E.B.G. Jones	HKU	BRUNEI DARUSSALAM. Temburong, Kuala Belalong Field Studies Centre, Sungai Esu, on submerged wood, Feb 1994, K.D. Hyde & S.W. Wong, (HKU(M) 3125).
Fluminicola bipolaris S.W. Wong, K.D. Hyde & E.B.G. Jones	HKU	PHILIPPINES. Mindanao, Bukidnon, Impalutao, Natigbasan Creek, on submerged wood, Jan 1994, K.D. Hyde, (HKU(M) 3127), HKUCC 3717.
Frondicola tunitricuspis K.D. Hyde	BRIP	BRUNEI. South China Sea. Tungit Api Api mangrove, on decaying fronds of *Nypa fruticans*, intertidal region, 14 Jun 1987, K.D. Hyde, (BRIP 17110).
Fusoidispora aquatica Vijaykr., Jeewon & K.D. Hyde	HKU	CHINA. Hong Kong, New Territories, Tai Po Kau Country Park, Tai Po Kau Forest Stream, on submerged wood, 26 Oct 2003, D. Vijaykrishna, (HKU(M) 17484).
Longicollum biappendiculatum Zelski, F.R. Barbosa, Raja, A.N. Mill & Shearer	ILL	PERU. Camanti, Stream at Quincemil Trail 1, 13°23960S, 70°77017W, on submerged woody debris, pH 6, water temp 20°C, 3 October 2010, Zelski S. and H.A. Raja, PE0017-2, ILL 40794.
Pseudoproboscispora aquatica (S.W. Wong & K.D. Hyde) Punith.	HKU	AUSTRALIA. North Queensland, near Cairns, Crystal Cascades, on submerged wood, Jun 1996, K.D. Hyde & T.M. Hyde, (CC63) (HKU(M) 2792). Ex. *Proboscispora aquatica*.

Table 2 continued.

Taxon	Herbarium	Details
Pseudoproboscispora caudae-suis (Ingold) J. Campb., Shearer, J.L. Crane & Fallah	IMI	ENGLAND. Anglia, Westmorland, Lake Windermere, on submerged decorticated twigs of *Fraxinus*, 14 Oct 1949, C.T. Ingold, (IMI 38506). Ex. *Ceriospora caudae-suis.*
Rivulicola aquatica Ranghoo & K.D. Hyde	HKU	CHINA. Hong Kong, Tai Po, Plover Cove Reservoir, on submerged wood, Jan 1997, V.M. Ranghoo, (PC 61) (HKU(M) 5214).
Rivulicola cygnea Raja & Shearer	ILL	USA. Florida. Apalachicola National Forest, unnamed lake east of Lost Lake, 30°21’48”N, 84°22’56”W, water 35 °C, pH 7.3, on submerged decorticated wood, 13 Jul 2004, H.A. Raja & Chris Brown, F41-1, ILL40111.
Rivulicola incrustata K.D. Hyde	BRIP	AUSTRALIA. North Queensland, Clohesy River, on submerged wood, Mar 1991, K.D. Hyde, (601) (BRIP 23340).
Submersisphaeria aquatica K.D. Hyde	BRIP	AUSTRALIA. North Queensland, Atherton Tablelands, Mt. Lewis, rainforest stream, on submerged wood, Aug 1992, K.D. Hyde, (1543) (BRIP 22743).
Submersisphaeria bambusicola D.Q. Zhou & K.D. Hyde	HKU	CHINA. Hong Kong, Hong Kong Island, Lung Fu Shan, on dead culm of *Arundinaria hindsii*, 19 Jul 1998, Dequn Zhou, (HKU(M) 9045).
Submersisphaeria palmae Pinnoi	BHH	THAILAND. Narathiwat, Sirindhorn Peat Swamp Forest, on submerged rachis of *Eleiodoxa conferta*, 13 Feb 2002,

Table 2 continued.

Taxon	Herbarium	Details
		A. Pinnoi, (Aom 152 in BHH).
Submersisphaeria rattanicola J. J. Fröhl. & K.D. Hyde	HKU	AUSTRALIA. North Queensland, Palmertson, Palmerston National Park, on dead rattan of *Calamus radicalis*, Mar 1994, J. Fröhlich, (HKU(M) JF 308).
Submersisphaeria vasicola (Ellis & Everh.) Y.Z. Wang, Aptroot & K.D. Hyde	NY	USA. New Jersey, Newfield, in cedar pail in constant use, Dec 1882, Ellis. Ex. *Sphaeria aquatica*.
Teracosphaeria petroica Réblová & Seifert	PDD	NEW ZEALAND. Westlands, Victoria National Park, Lewis Pass, ~30 km from Springs Junction, Riordan's Creek valley, decayed wood of a trunk of *Nothofagus* sp., 20 Feb 2003, M.R. Réblová & K.A. Seifert, (2570/03) (PDD 81436).
Torrentispora crassiparietis Fryar & K.D. Hyde	HKU	BRUNEI. Tutong District, Sungai Kelakas, Site 3, on submerged rotting wood, 27 Oct 1998, S.C. Fryar, (HKU(M) 15667).
Torrentispora fibrosa K.D. Hyde, Wai, H., Ho, E.B.G. Jones, K.M. Tsui & S.W. Wong	HKU	CHINA. Hong Kong, New Territories, Tai Po Kau Forest Reserve, Tai Po Kau Forest Stream, on submerged decomposing wood, 10 Dec 1995, W. H. Ho (HKU(M) 4519).
Torrentispora fusiformis Fryar & K.D. Hyde	HKU	BRUNEI. Tutong District, Sungai Kelakas, a tributary of the Tutong River, Site 1, in submerged rotting wood, 27 Oct 1998, S.C. Fryar, (HKU(M)

Table 2 continued.

Taxon	Herbarium	Details
		16048).
Torrentispora pilosa Shearer & F.R. Barbosa	ILL	COSTA RICA. Heredia, La Selva Biological Station, 10^{o}25.948’N, 84^{o}19.32’W, water 25 C, pH 5, on submerged wood, 18 May 2000, J. Anderson & R. Wulffen, A652-1, ILL 40814.
Vertexicola ascoliberatus Shearer & F.R. Barbosa	ILL	COSTA RICA. Heredia, La Selva Biological Station, Salto 30, 10^{o}25.939’N, 84^{o}09.90’W, water 26 C, pH 7, on submerged wood, 18 May 2000, J. Anderson & R. Wulffen, A 653-1, ILL 40815.
Vertexicola caudatus K.D. Hyde, S.W. Wong & Ranghoo	HKU	PHILIPPINES. Mindanao, Bukidnon. Natigbasan Creek, Impalutao, on submerged wood, Jan 1994, K.D. Hyde, (HKU(M) 3108), HKUCC 3715.

Table 3. Primers used in this study.

Gene	Primer	Sequence (5' -> 3')	Reference
SSU	NS1	GTAGTCATATGCTTGTATC	White et. al 1990
	NS2	GGCTGCTGGCACCAGACTTGC	White et. al 1990
	NS3	GCAAGTCTGGTGCCAGCAGCC	White et. al 1990
	NS4	CTTCCGTCAATTCCTTTAAG	White et al. 1990
ITS	ITS1F	CTTGGTCATTTAGAGGAAGTAA	Gardes and Bruns 1993
	ITS2	GCTGCGTTCTTCATCGATGC	White et al. 1990
	ITS3	GCATCGATGAAGAACGCAGC	White et al. 1990
	ITS4	TCCTCCGCTTATTGATATGC	White et al. 1990
LSU	LROR	GTACCCGCTGAACTTAAGC	Rehner and Samuels 1994
	LR3	GGTCCGTGTTTCAAGAC	Vilgalys and Hester 1990
	LR3R	GTCTTGAAACACGGACC	Vilgalys and Hester 1990
	LR6	CGCCAGTTCTGCTTACC	Vilgalys and Hester 1990
Mcm7	709for	ACIMGIGTITCVGAYGTHAARCC	Schmitt et al. 2009
	1348rev	GAYTTDGCIACICCIGGRTCWCCCAT	Schmitt et al. 2009

A – Adenine; C – Cytosine; D –Adenine, Guanine, or Thymine; G – Guanine; H – Adenine, Cytosine, or Thymine; I – Inosine; M – Adenine or Cytosine; R – Purine (Adenine or Guanine); T – Thymine; V – Adenine, Cytosine, or Guanine; W – Adenine or Thymine; Y – Pyrimidine (Cytosine or Thymine). Abbreviations based on the Nomenclature Committee of the International Union of Biochemistry (NC-IUB 1985a, NC-IUB 1985b, NC-IUB 1985c, NC-IUB 1986a, NC-IUB 1986b, NC-IUB 1986c, NC-IUB 1992).

Table 4. Number of taxa, number of sequences, and alignment length for molecular analyses conducted in this study

Alignment	SSU	ITS	LSU	Mcm7	SSU + LSU	SSU + LSU + Mcm7
Taxa	90	49	148	35	148	148
Sequences	113	73	203	43	203	203
Length	1132	763	632	645	1764	2409

Table 5. jModelTest results for alignments used in this study.

Result	SSU	ITS	LSU	Mcm7	SSU+LSU	SSU+LSU+Mcm7
Model	TIM2+I+G	TIM2ef+I+G	GTR+G	GTR+I+G	TIM3+I+G	TIM1+I+G
AIC	17526.09	34005.168	25340.60	10135.28	60163.188	81212.204
-lnL	8597.046	16829.584	12377.40	10135.28	29671.594	40194.102
F[A]	0.2512	0.2584	0.159	0.1602	0.23	0.22
F[C]	0.2244	0.2657	0.2936	0.3795	0.25	0.27
F[G]	0.27	0.2236	0.3671	0.3105	0.32	0.3
F[T]	0.2544	0.2522	0.1803	0.1498	0.19	0.2
R[AC]	1.687	1.4267	0.9232	6.1553	0.702	1.0
R[AG]	3.7814	2.0577	2.2483	17.0591	2.004	2.8811
R[AT]	1.6712	1.4267	1.5367	9.7229	1.0	1.1522
R[CG]	0.9032	1.0	.4972	2.7383	0.702	1.1522
R[CT]	6.5704	3.9642	6.2735	21.1104	6.288	5.6671
R[GT]	1.0	1.0	1.0	1.0	1.0	1.0
p-inv	0.2672	0.051	-	0.304	0.23	0.267
Γ	0.6542	0.607	0.436	0.483	0.46	0.473

Table 6 continued.

Table 6. Isolate, strain, and/or voucher information (if available) and GenBank accession numbers for taxa and sequences used in this study.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
Aniptodera chesapeakensis	U46882	ATCC 32818	-	-	U46870	ATCC 32818	-	-
Annulatascus aquatorba	JN226107	SS02424.01	-	-	JN226106	SS02424.01	-	-
Annulatascus aquatorba	Sequin	ILL 41216 PE0255-1a	Sequin	ILL 41216 PE0255-1a	Sequin	ILL 41216 PE0255-1a	Sequin	ILL 41216 PE0255-1a
Annulatascus aquatorba	Sequin	ILL 41216 PE0255-1b	Sequin	ILL 41216 PE0255-1b	Sequin	ILL 41216 PE0255-1b	-	-
Annulatascus biatriisporus	AY316352	A464-3	-	-	-	-	-	-
Annulatascus biatriisporus	Sequin	ILL 41217 PE0330-1	-	-	-	-	-	-
Annulatascus citriosporus a	Sequin	ILL 41218 PE0086-2a	Sequin	ILL 41218 PE0086-2a	Sequin	ILL 41218 PE0086-2a	Sequin	ILL 41218 PE0086-2a
Annulatascus citriosporus b	Sequin	ILL 41218 PE0086-2b	-	-	-	-	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
Annulatascus hongkongensis	AF132319	HKUCC 3702	-	-	-	-	-	-
Annulatascus-like	Sequin	ILL 41219 PE0035-4	Sequin	ILL 41219 PE0035-4	-	-	-	-
Annulatascus-like	Sequin	ILL 41220 PE0324-1	-	-	-	-	Sequin	ILL 41220 PE0324-1
***Annulatascus*-like**	Sequin	ILL 41221 A612-3a	Sequin	ILL 41221 A612-3a	Sequin	ILL 41221 A612-3a	-	-
***Annulatascus*-like**	Sequin	ILL 41221 A612-3b	Sequin	ILL 41221 A612-3b	Sequin	ILL 41221 A612-3b	-	-
Annulatascus longisporus	-	-	AF177149	GenBank	-	-	-	-
Annulatascus nilensis	HQ616536	IMI 397966	-	-	-	-	-	-
Annulatascus velatisporus	Sequin	ILL 41222 PE0160-9	Sequin	ILL 41222 PE0160-9	Sequin	ILL 41222 PE0160-9	Sequin	ILL 41222 PE0160-9
Annulatascus velatisporus	AY312320	HKU 3701	AF177150	HKU 3701	-	-	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
Annulatascus velatisporus	AF316355	R047	-	-	-	-	-	-
Annulatascus velatisporus	AF316354	A70-18	-	-	-	-	-	-
Annulatascus velatisporus	Sequin	ILL 41223 TH0011-1a	-	-	-	-	-	-
Annulatascus velatisporus	Sequin	ILL 41223 TH0011-1b	-	-	-	-	-	-
Annulatascus velatisporus	-	-	Sequin	ILL 41224 PE0011-54	Sequin	ILL 41224 PE0011-54	-	-
Annulatascus velatisporus	Sequin	ILL 41225 PE0011-9a	Sequin	ILL 41225 PE0011-9a	Sequin	ILL 41225 PE0011-9a	Sequin	ILL 41225 PE0011-9a
Annulatascus velatisporus	Sequin	ILL 41225 PE0011-9b	-	-	Sequin	ILL 41225 PE0011-9b	-	-
Annulatascus velatisporus	Sequin	ILL 41225 PE0011-9c	-	-	Sequin	ILL 41225 PE0011-9c	-	-
Annulatascus	-	-	Sequin	ILL 41226	-	-	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
velatisporus				PE0011-35a				
Annulatascus velatisporus	-	-	Sequin	ILL 41226 PE0011-35b	-	-	-	-
Annulusmagnus triseptatus	-	-	Sequin	ILL 41227 Z31-1b	-	-	-	-
Annulusmagnus triseptatus	Sequin	ILL 41228 Z31-1824a	Sequin	ILL 41228 Z31-1824a	Sequin	ILL 41228 Z31-1824a	Sequin	ILL 41228 Z31-1824
Annulusmagnus triseptatus	-	-	Sequin	ILL 41228 Z31-1824b	Sequin	ILL 41228 Z31-1824b	-	-
Annulusmagnus triseptatus	AY316353*	R059	-	-	JQ429242	CBS 128831	-	-
Annulusmagnus triseptatus	AY346257	SMH2359	-	-	-	-	-	-
Annulusmagnus triseptatus	AY780049	SMH4832	-	-	-	-	-	-
Annulusmagnus triseptatus	AY590289	A353-1f	-	-	-	-	-	-
Annulusmagnus	AY590288	A325-1d	-	-	-	-	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
triseptatus								
Annulusmagnus triseptatus	AY590287	A54-10e	-	-	-	-	-	-
Annulusmagnus triseptatus	AY590286	A54-10a	-	-	-	-	-	-
Annulusmagnus triseptatus	AY590285	A413-6	-	-	Sequin	-	-	A413-6
Annulusmagnus triseptatus	GQ996540	MR2948	-	-	-	-	-	-
Apignomonia errabunda	AF408334	AR 2813 AFTOL 2120	DQ313525	AR 2813 AFTOL 2120	DQ862045	AR 2813 AFTOL 2120	-	-
Aquaticola ellipsoidea	AY316356	A411-3	-	-	-	-	-	-
Aquaticola ellipsoidea	AY590290	R008	-	-	-	-	-	-
Aquaticola hongkongensis	AF132321	HKUCC 3703	AF177156	HKUCC 3703	-	-	-	-
Aquaticola hyalomura	AY590291	R038	-	-	-	-	-	-
***Aquaticola*-like**	Sequin	ILL 41229 AK-1838a	-	-	-	-	Sequin	ILL 41229 AK-1838a

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
***Aquaticola*-like**	Sequin	ILL 41229 AK-1838b	-	-	-	-	-	-
***Aquaticola*-like**	Sequin	ILL 41230 PE0157-1	Sequin	ILL 41230 PE0157-1	Sequin	ILL 41230 PE0157-1	-	-
***Aquaticola*-like**	Sequin	ILL 41231 PE0159-1a	Sequin	ILL 41231 PE0159-1a	Sequin	ILL 41231 PE0159-1a	-	-
***Aquaticola*-like**	Sequin	ILL 41232 PE0316-1	Sequin	ILL 41232 PE0316-1	Sequin	ILL 41232 PE0316-1	Sequin	ILL 41232 PE0316-1
***Aquaticola*-like**	Sequin	ILL 41233 PE0264-1	-	-	-	-	-	-
Aquaticola miniguttulata	-	-	Sequin	ILL 41234 PE0120-1a	-	-	-	-
Aquaticola miniguttulata	Sequin	ILL 41234 PE0120-1c	-	-	-	-	Sequin	ILL 41234 PE0120-1c
Aquaticola miniguttulata	-	-	Sequin	ILL 41234 PE0120-1d	-	-	-	-
Aquaticola triseptata	Sequin	ILL 41235	Sequin	ILL 41235	Sequin	ILL 41235	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
		PE0164-1b		PE0164-1b		PE0164-1b		
Ascitendus austriacus	AY590294	A324-1f	-	-	-	-	-	-
Ascitendus austriacus	AY590293	A324-1b	-	-	-	-	-	-
Ascitendus austriacus	AY590292	A44-28a	-	-	-	-	-	-
Ascitendus austriacus	AY094186	A413-6b	-	-	-	-	-	-
Ascitendus austriacus	Sequin	ILL 41236 Z57-1688	Sequin	ILL 41236 Z57-1688	-	-	-	-
Ascitendus austriacus	GQ996539	MR 2936	-	-	GQ996542	MR 2936	-	-
Ascotaiwania hughesii	AY316357	P2-6	-	-	-	-	-	-
Ascotaiwania mitriformis	AF132324	HKUCC 3706	-	-	-	-	-	-
Ascotaiwania persoonii	AY590295	A57-14c	-	-	-	-	-	-
Ascotaiwania sp.	Sequin	ILL 41237 PE0059-1	Sequin	ILL 41237 PE0059-1	Sequin	ILL 41237 PE0059-1	Sequin	ILL 41237 PE0059-1
Ascotaiwania sp.	Sequin	ILL 41238 PE0238-2	-	-	-	-	-	-
Ayria-like	Sequin	ILL 41239	Sequin	ILL 41239	Sequin	ILL 41239	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
		PE0323-1a		PE0323-1a		PE0323-1a		
Ayria nubispora	Sequin	F99-1	-	-	-	-	-	-
Balansia henningsiana	AY489715	AEG 96-27a GAM 16112	JN049815	AEG 96-27a GAM 16112	AY489683	AEG 96-27a GAM 16112		
Bertia moriformis	AY695261	SMH 3344	-	-	-	-	-	-
Bertia moriformis	AY695260	SMH 4320	-	-	-	-	-	-
Brunneosporella aquatica	AF132326	HKUCC 3708	AF177154	HKUCC 3708	-	-	-	-
Bullimyces aurisporus	JF775590	AF316-1b	-	-	JF758614	AF316-1b	-	-
Bullimyces communis	JF775585	AF281-3	-	-	JF758617	AF281-3	-	-
Bullimyces costaricensis	JF775592	AF317-1b	-	-	JF758616	AF317-1b	-	-
Camarops amorpha	AY780054	SMH 1450	-	-	-	-	-	-
Camarops lutea	JN673032	Hanson 2008-222	-	-	-	-	JN672974	Hanson 2008-222
Camarops microspora	AY083821	CBS 649.92 AFTOL 1361	-	-	DQ471036	CBS 649.92 AFTOL 1361	-	-
Camarops plana	JN673033	Pouzar s.n.	-	-	-	-	JN672975	Pouzar s.n.

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
Camarops polysperma	JN673030	Hanson 2006-747	-	-	-	-	JN672973	Hanson 2006-747
Camarops scleroderma	JN673034	MP 4464	-	-	-	-	JN672976	MP 4465
Camarops sp.	JN673035	CH 08-570	-	-	-	-	JN672977	CH 08-570
Camarops tubulina	AY346266	SMH 4614	-	-	-	-	-	-
Camarops ustulinoides	DQ470941	DEH 2164 AFTOL 72	-	-	DQ470989	DEH 2164 AFTOL 72	-	-
Cataractispora appendiculata	Sequin	ILL 41240 PE0066-1a	Sequin	ILL 41240 PE0066-1a	Sequin	ILL 41240 PE0066-1a	-	-
Cataractispora appendiculata	Sequin	ILL 41240 PE0066-1b	-	-	Sequin	ILL 41240 PE0066-1b	-	-
Cataractispora receptaculorum	AF132327	HKUCC 3710	AF177153	HKUCC 3710	-	-	-	-
Cataractispora sp. nov		ILL 41241 PE0266-1a	Sequin	ILL 41241 PE0266-1a	-	-	-	-
Cataractispora viscosa	Sequin	ILL 41242	Sequin	ILL 41242	-	-	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
		PE0165-8b		PE0165-8a				
Ceratocystis fimbriata	U17401	TCH C89	-	-	U32418	TCH C89	-	-
Ceratosphaeria lampadophora	AY761084	CBS 117555	-	-	AY761088	CBS 117555	-	-
Cercophora aquatica	JN673036	JF 09314	-	-	-	-	JN672978	JF 09314
Cercophora arenicola	JN673037*	ANM 1080	JN673037*	ANM 1080	-	-	JN672979	ANM 1080
Cercophora sp.	JN673038*	JF 09214	JN673038*	JF 09214	-	-	JN672980	JF 09214
Chaetomium globosum	JX280684	CBS 148.51	KC109754	CBS 155.52	JN546130	YK28	XM_001220296	CBS 148.51
Chaetorostrum quincemilense	Sequin	ILL 40822 PE0105-1a	Sequin	ILL 40822 PE0105-1a	Sequin	ILL 40822 PE0105-1a	Sequin	ILL 40822 PE0105-1a
Chaetorostrum quincemilense	Sequin	ILL 40822 PE0105-1b	Sequin	ILL 40822 PE0105-1b	-	-	-	-
Chaetorostrum quincemilense	Sequin	ILL 40822 PE0105-1c	Sequin	ILL 40822 PE0105-1c	-	-	-	-
Chaetosphaerella phaeostroma	AY346274	SMH 4585	-	-	-	-	-	-
Chaetosphaeria	EF063574	ICMP 15153	-	-	-	-	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
fuegiana								
Chaetosphaeria innumera	AY017375	SMH 2748	AY906956	SMH 2748	-	-	-	-
Chaetosphaeria lateriphiala	JN673039	ANM1079	JN673039	ANM1079	-	-	JN672984	ANM 1079
Chaetosphaeria ovoidea	AF064641	SMH 2605	-	-	-	-	-	-
Chromendothia citrine	AF408335	AR 3445 AFTOL 2121 CBS 109758 BPI 747935	-	-	DQ862046 NG_013200	AR 3446 AFTOL 2121 CBS 109758 BPI 747935	-	-
Chrysoporthe cubensis	AF408338	AFTOL 2122 CBS 101281	-	-	DQ862047	AFTOL 2122 CBS 101281	-	-
Claviceps purpurea	AF543789	GAM 12885	U57669	GAM 12885	AF543765	GAM 12885	-	-
Clohiesia corticola	AF132329	HKUCC 3712	-	-	-	-	-	-
Clypeosphaeria uniseptate	DQ810219	HKUCC 6349	-	-	DQ810255	HKUCC 6349	-	-
Coniochaeta	FJ167399	Jong 54 TNMF	-	-	-	-	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
leucoplaca		17728						
Coniochaeta ostrea	DQ470959	AFTOL 915	-	-	DQ471007	AFTOL 915	-	-
Coniochaeta sp.	JN673041	ANM 1174	JN673041	ANM1174	-	-	JN672986	ANM 1174
Coniochaetidium savory	AY346276	TRTC 51980	-	-	-	-	-	-
Conlarium duplumascospora a	JN936991	CGMCC 3.14938	JN936995	CGMCC 3.14938	JN936987	CGMCC 3.14938	-	-
Conlarium duplumascospora b	JN936992	CGMCC 3.14939	JN936996	CGMCC 3.14939	JN936988	CGMCC 3.14939	-	-
Conlarium duplumascospora c	JN936993	CGMCC 3.14940	JN936997	CGMCC 3.14940	JN936989	CGMCC 3.14940	-	-
Creosphaeria sassafras	JN673042	ANM1978	JQ846063	5482	-	-	JN672987	ANM1978
Cryphonectria parasitica	JN940857	CMW 13749 MAFF 410158	JN942326	CMW 13749 MAFF 410158	JN940857	CMW 13749 MAFF 410158	-	-
Cyanoannulus petersenii	Sequin	ILL 41243 Z60-1824	Sequin	ILL 41243 Z60-1824	Sequin	ILL 41243 Z60-1824	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
Cyanoannulus petersenii	AY316359	R044b	-	-	-	-	-	-
Cyanoannulus petersenii	AY316358	R044a	-	-	-	-	-	-
Diaporthe eres	AF408350	AR 3538 BPI 748435	DQ491514	AFTOL 935 CBS109767	DQ471015	AFTOL 935 CBS109767	-	-
Diaporthe passiflorae	JX069844	PC 19183 CBS 132527	JX069860	PC 19183 CBS 132527	-	-	-	-
Diaporthe phaseolorum	U47830	FAU 458 EU255083	-	-	L36985 AY779326	FAU 458	-	-
Doratomyces stemonitis	DQ836907	AFTOL 1380 CBS 127.22	-	-	DQ836901	AFTOL 1380 CBS 127.22	-	-
Elaphocordyceps capitate	AY489721	OSC 71233	-	-	AY489689	OSC 71233	-	-
Elaphocordyceps ophioglossoides	AY489723	OSC 106405	-	-	AY489691	OSC 106405	-	-
Endothia gyrosa	DQ470972	CBS 112915	-	-	DQ471023	CBS 112915	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
		AFTOL 1223				AFTOL 1223		
Eutypa lata	DQ836903	AFTOL 929	-	-	DQ836896	AFTOL 929	-	-
Fluminicola bipolaris	Sequin	ILL 41244 PE0020-1a	Sequin	ILL 41243 PE0020-1a	Sequin	ILL 41243 PE0020-1a	Sequin	ILL 41243 PE0020-1a
Fluminicola bipolaris	Sequin	ILL 41243 PE0020-1b	Sequin	ILL 41243 PE0020-1b	-	-	-	-
Fluminicola bipolaris	Sequin	ILL 41245 PE0020-3b	-	-	-	-	-	-
Fluminicola coronata	AF132332	HKU 3717	AF177152	HKU 3737	-	-	-	-
Fragosphaeria purpurea	AF096191	CBS 133.34	AB278192	CBS 133.34	AF096176	CBS 133.34	-	-
Fusoidispora aquatica	AY780365	HKU(M) 17484	-	-	-	-	-	-
Gnomonia borealis	EU255169	CBS 799.79 Monod358	EU255000	CBS 799.79 Monod358	-	-	-	-
Gnomonia gnomon	AF408361	CBS 199.53 AFTOL 952	AY818956	CBS 199.53 AFTOL 952	DQ471019	CBS 199.53 AFTOL 952	-	-
Gnomonia petiolorum	EU255070	AR 4082 CBS 121227 BPI	EU254748	AR 4082 CBS 121227	-	-	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
		844274		BPI 844274				
Graphium penicillioides	AF027384	CBS 506.86	-	-	DQ471038	AFTOL 1415 CBS 506.86	-	-
Halosphaeria appendiculata	U46885	CBS 197.60	-	-	U46872	CBS 197.60	-	-
Hyalorostratum brunneisporum	HM191719	A573-2a	-	-	HM191721	A573-2a	-	-
Hyalorostratum brunneisporum	HM191720	A573-2b	-	-	-	-	JN672996	A573-2b
Hydromelitis pulchella	JF775588	AF284-2	-	-	JF758613	AF284-2	-	-
Hypocrea citrina	AY544649	AFTOL 52 OSC 100005	-	-	AY544693	AFTOL 52 OSC 100005	-	-
Hypocrea lutea	AF543791	ATCC 208838	-	-	AF543768	ATCC 208838	-	-
Hyponectria buxi	AY083834	UME 31430	-	-	AF130976	UME 31430	-	-
Jobellisia guangdongensis	JN936990	GD14-4	JN936994	GD14-4	-	-	-	-
Jobellisia luteola	AY346286	SMH 2753	-	-	-	-	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
Lasiosphaeria hirsuta	JN673051*	ANM 1066	JN673051*	ANM 1066	-	-	JN673009	ANM 1066
Lasiosphaeria lanuginose	AY436412	SMH 3819	AY587921	SMH 3819	-	-	JN673006	SMH 3819
Lasiosphaeria lanuginose	JN673049	SMH 4925	-	-	-	-	JN673007	SMH 4925
Lasiosphaeria ovina	AY436413	SMH 4605	-	-	DQ836894	SMH 4605 AFTOL 17	-	-
Lasiosphaeria ovina	JN673050	ANM 1295	JN673050	ANM 1295	EU940082	M176	JN673008	ANM 1295
Lentomitella cirrhosa	AY761085	ICMP 15131	-	-	AY761089	ICMP 15131	-	-
Lentomitella crinigera	AY761086	CBS 113655	-	-	-	-	-	-
Leucostoma niveum	AF362558 NG_027590	AR 3413 BPI 748232	-	-	DQ862050 NG_013203	AFTOL 2125 AR 3413 CBS 109489 BPI 748232	-	-
Longicollum biappendiculatum	Sequin	ILL 40794 PE0017-1a	Sequin	ILL 40794 PE0017-1a	Sequin	ILL 40794 PE0017-1a	Sequin	ILL 40794 PE0017-1a
Longicollum biappendiculatum		ILL 40794 PE0017-1b	-	-	-	-	Sequin	ILL 40794 PE0017-1b

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
Longicollum biappendiculatum		ILL 41246 PE0017-2a	-	-	-	-	-	-
Longicollum biappendiculatum		ILL 41246 PE0017-2b	-	-	-	-	-	-
Mazzantia napelli	AF408368	AR 3498 BPI 748443	-	-	DQ862051	AFTOL 2126 AR 3498	-	-
Melanconis alni	AF408371	AR 3500 BPI 748444	-	-	DQ862052	AFTOL 2127 AR 3500	-	-
Melanconis marginalis	AF408373	AR 3442 BPI 748446	-	-	DQ862053	AFTOL 2128 AR 3442	-	-
Melanconis stilbostoma	AF408374	AR 3501 BPI 748447	-	-	DQ862054	AFTOL 2129 AR 3501	-	-
Melanospora tiffanii	AY015630	ATCC 15515	-	-	AY015619	ATCC 15515	-	-
Melanospora zamiae	AY046579	ATCC 12340	-	-	AY046578	ATCC 12340	-	-
Menispora tortuosa	AY544682	AFTOL 278 DAOM 231154 OSC100094	-	-	AY544723	AFTOL 278 DAOM 231154 OSC100094	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
Microascus longirostris	AF400865	CBS 267.49	-	-	DQ471026	AFTOL 1237 CBS 267.49	-	-
Microascus trigonosporus	DQ470958	AFTOL 914 CBS 218.31	DQ491513	AFTOL 914 CBS 218.31	DQ471006	AFTOL 914 CBS 218.31	-	-
Mirannulata samuelsii	AY578353	SMH 1880	-	-	-	-	-	-
Nectria cinnabarina	U00748	GJS 89-107 CBS 114055	-	-	U32412	GJS 89-107 CBS 114055	-	-
Neurospora crassa	AF286411	MUCL 19026	KF040479	HT-ITV31	KF312458	FH-1	XM_958785	OR74A
Nitschkia grevillea	AY346294	SMH 4663	-	-	-	-	-	-
Nohea umiumi	U46893	JK 5103F	-	-	U46878	JK 5103F	-	-
Ophioceras tenuisporum	AY346295	SMH1643	-	-	AF050475	CS652-1	-	-
Ophiostoma piliferum	DQ470955	CBS 158.74 AFTOL 910	AF221070	CBS129.32	AJ243295	CBS 129.32	-	-
Ophiostoma stenoceras	DQ836904	CBS 139.51 AFTOL 1038	AF484475	CBS 798.73	M85054	UCB 57.013	-	-

Table 6 continued.

Taxon	LSU	Voucher	ITS	Voucher	SSU	Voucher	Mcm7	Voucher
Paoayensis lignicola	EF622535	HKU(M) 17516	-	-	EF622536	HKU(M) 17516	-	-
Papulosa amerospora	DQ470950	AFTOL 748 JK 5547F	-	-	DQ470998	AFTOL 748 JK 5547F	-	-
Petriella setifera	DQ470969	AFTOL 956 CBS 437.75	-	-	DQ471020	AFTOL 956 CBS 437.75	-	-
Pleurostoma ootheca	AY761079	CMU 23858 CBS 115329	HQ878590	CBS 115329	AY761074	CMU 23858 CBS 115329		
Podospora decipiens	AY780073	CBS 258.64	-	-	-	-	-	-
Podospora fibrinocaudata	AY780074	TRTC 48343	-	-	-	-	-	-
Poroconiochaeta discoidea	AY346297	SANK 12878	-	-	-	-	-	-
Pseudoproboscispora caudae-suis	Sequin	ILL 41247 Z32-1822	Sequin	ILL 41247 Z32-1822	Sequin	ILL 41247 Z32-1822	Sequin	ILL 41247 Z32-1822
Pseudoproboscispora caudae-suis	-	-	-	-	Sequin	Z32-1824	-	-
Pseudoproboscispora caudae-suis	Sequin	ILL 41248 A40-39a	-	-	Sequin	ILL 41248 A40-39a	Sequin	ILL 41248 A40-39a

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
Pseudoproboscispora caudae-suis	Sequin	ILL 41248 A40-39b	-	-	Sequin	ILL 41248 A40-39b	-	-
Pseudoproboscispora caudae-suis	AY094192	A336-2d	-	-	-	-	-	-
Pseudoproboscispora caudae-suis	AY094191	A40-1a	-	-	-	-	-	-
Rhamphoria delicatula	AF261068	MR 1396/98	-	-	AF242267	MR 1396	-	-
Riomyces rotundus	JF775589	AF303-1	-	-	JF758612	AF303-1	-	-
Rivulicola incrustata	Sequin	ILL 41249 PE0031-2	Sequin	ILL 41249 PE0031-2	Sequin	ILL 41249 PE0031-2	-	-
Scortechinia acanthostroma	FJ968991	GKM L163N	-	-	-	-	-	-
Scortechinia acanthostroma	FJ968990	SMH 5313	-	-	-	-	-	-
Sordaria fimicola	AY545728	CBSC 15-6291	-	-	AY545724	CBSC 15-6291	-	-
Stachybotrys	AY489712	ATCC 66238	-	-	AY489680	ATCC 66238	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
chartarum								
Submersisphaeria aquatica	Sequin	ILL 41250 Z43-2e	Sequin	ILL 41250 Z43-2e	Sequin	ILL 41250 Z43-2e	-	-
Submersisphaeria aquatica	Sequin	ILL 51251 Z43-1824	Sequin	ILL 51251 Z43-1824	Sequin	ILL 51251 Z43-1824	Sequin	ILL 51251 Z43-1824
Submersisphaeria aquatica	AY094194	A354-1c	-	-	-	-	-	-
Submersisphaeria aquatica	AY094193	A95-1b	-	-	-	-	-	-
Submersisphaeria rattanicola	Sequin	ILL 41252 PE0292-1a	Sequin	ILL 41252 PE0292-1a	Sequin	ILL 41252 PE0292-1a	Sequin	ILL 41252 PE0292-1a
Teracosphaeria petroica	EF063576	ICPM 15111	-	-	-	-	-	-
Togninia minima	AY761082	CBS 6580 CBS 213.31	-	-	AY761068	CBS 6580 CBS 213.31	-	-
Togninia fraxinopennsylvanica	AY761083	ATCC 26664 CBS 101585	-	-	AY761070	ATCC 26664 CBS 101585	-	-
Torrentispora	Sequin	ILL 41253	Sequin	ILL 41253	Sequin	ILL 41253	-	-

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
crassiparietis		PE0068-1		PE0068-1		PE0068-1		
Torrentispora fibrosa	Sequin	ILL 41254 PE0038-2fb	Sequin	ILL 41254 PE0038-2fb	Sequin	ILL 41254 PE0038-2fb	-	-
Valsa ambiens	AF362564	AFTOL 2131 AR 3516 BPI 748237	AY347330	CBS 191.42	DQ862056	AFTOL 2131 AR 3516	-	-
Valsella salicis	AF408389 EU255210	AR 3514 CBS 109754 BPI 748461	-	-	DQ862057	AFTOL 2132 AR 3514	-	-
Vertexicola triseptata	Sequin	ILL 41255 PE0104-1a	Sequin	ILL 41255 PE0104-1a	Sequin	ILL 41255 PE0104-1a	Sequin	ILL 41255 PE0104-1a
Vertexicola triseptata	Sequin	PE0104-1b	Sequin	PE0104-1b	Sequin	PE0104-1b	-	-
***Vertexicola caudatus* (= *V. confusa* in GenBank)**	AF132331	HKUCC 3715	AF177151	GenBank	-	-	-	-
Xylaria acuta	AY544676	AFTOL 63 ATCC 56487	DQ491493	AFTOL 63 ATCC 56487	AY544719	AFTOL 63 ATCC 56487	-	-
Xylaria hypoxylon	AY544648	AFTOL 51	DQ491487	AFTOL 51	AY544692	AFTOL 51	JX000190	F118002

Table 6 continued.

Taxon	**LSU**	**Voucher**	**ITS**	**Voucher**	**SSU**	**Voucher**	**Mcm7**	**Voucher**
		OSC 100004		OSC 100004		OSC 100004		
Xylomelasma sordida	AY761087	CBS 116000	-	-	AY761090	CBS 116000	-	-

Voucher/strain/isolate abbreviations: A = Carol A. Shearer; AF = Astrid Ferrer; AFTOL = Assembling The Fungal Tree Of Life; ANM = Andrew N. Miller, AR = Amy Rossman; BPI = U.S. National Fungus Collections, Systematic Botany and Mycology, Rm. 304, Bldg. 011A, 10300 Baltimore Avenue, Beltsville, MD 20705-2350, USA; CBSC = Carolina Biological Supply Company; CBS = Centraalbureau voor Schimmelcultures; CGMCC = China General Microbial Collection; CH = China; CMW = Forestry and Agricultural Biotechnology Institute (FABI) culture collection; CS = Carol A. Shearer; F = Florida, Huzefa Raja; FAU = F.A. Uecker; GAM = Julian H. Miller Mycological Herbarium; GenBank = GenBank only ID; GKM = George K. Mugambi; HKU = Hong Kong University; HKUCC = Hong Kong University Culture Collection; HT-ITV = Instituto Tecnologico de Veracruz; ICMP = International Collection of Microorganisms from Plants; ILL = University of Illinois; IMI = Herbarium, CABI Bioscience, Bakeham Lane, Egham, Surrey TW20 9TY, UK; JF = Jack Fournier; JK = Jan Kohlmeyer; MAFF = Ministry of Agriculture, Forestry and Fisheries, Japan; MP = Meike Piepenbring; MYA = American Type Culture Collection; MR = Martina Réblová; MUCL = Mycothèque de l'Université Catholique de Louvain, Louvain-la-Neuve, Belgium; OSC = Oregon State University Herbarium; P = Payam M. Fallah; PE = Perú, Carol A. Shearer; R = Huzefa Raja; SMH = Sabine M. Huhndorf; SS = Somsak Sivichai; TCH = T.C. Harrington; TNMF = Herbarium of National Museum of Natural Science, Taichung; TRTC = Royal Ontario Museum Fungarium; UME = Umeå University, Umeå , Sweden; UPS = Uppsala University, Uppsala, Sweden; Z = Zelski S.E.

Table 7 continued.

Table 7. Partial morphological database used in this study. Non-bold taxa are invalid names..

Species	**Ascoma dimensions**	**Neck dimensions**	**Paraphyses dimensions**	**Ascus dimensions**	**Annulus dimensions (l × w)**	**Ascospore dimensions**	**Ascospore shape**	**Ascospore septation**	**Ascospore coloration**	**Appendages**	**Sheath**
Annulatascus apiculatus	400–550 × 240–410 µm	100–250 × 100–180 µm	2.5–6 × 75–100 µm	175–250 × 10–13 µm	6–7.2 × 1.8–2.4 µm	23–36.5 × 8.8–10 µm	fusiform	0–3 septate	hyaline	bipolar apiculi	present, narrow
Annulatascus aquaticus	240–600 × 220–600 µm	400–800 × 60–80 µm	3–4.5 × ? µm	150–175 × 10–12 µm	4.5–5 × 2.5–4 µm	19–24 × 6–7 µm	ellipsoidal	aseptate	hyaline	none	present at ultrastructural level
Annulatascus aquatorba	390–500 × 310–350 µm	400–480 × 100–120 µm	to 2.5 µm wide	102.5–152.5 × 6.25–8.75 µm	5.0–7.5 µm wide	15–22.5 × 5–7.5 µm	fusoid to lunate	3–septate	central cells brown, end cells subhyaline	none	none observed
Annulatascus biatriisporus	195–325 × 390–520 µm	390 × 110 µm	4 × ? µm	210–260 × 12–17 µm	3–4 × 7–8 µm	40–65 × 7.5–10 µm	fusiform	aseptate	hyaline	weakly swollen ends	thin, irregular
Annulatascus citriosporus	350–485 × 224–310 µm	?	6–8 × 72.5–155 µm	137.5–178.8 × 10–12 µm	2.75–3.25 × 5–5.5 µm	22.5–30.4 × 6.4–8.5 µm	citriform	0–3 septate	hyaline	none	thin
Annulatascus crassitunicatus	280–400 × 200–380 µm	550–950 × 75–90 µm	8–14 × ? µm	95–120 × 14–16 µm	4.5–5 × 4–5.5	24–29 × 6–8.5 µm	ellipsoidal	3–septate	hyaline	none	none observed
Annulatascus fusiformis	170–220 × ? µm	"long"	7.5 × ? µm	141–235 × 7.5–10.5 µm	4–5 × 3–4 µm	16.5–25.5 × 6–9 µm	fusiform	1–5 septate	hyaline	bipolar pad–like	none observed
Annulatascus hongkongensis	250–280 × ? µm	140–150 × 35–40 µm	6.5–8.5 × 200–250 µm	250–275 × 25–30 µm	3.5–4.5 × 4.7–6 µm	35–37.5 × 12.5–15 µm	ellipsoidal	3–septate	hyaline	verruculose at SEM level	thick
Annulatascus incrustatus	200–340 × 100–160 µm	200–350 × 50–90 µm	2.5–4.5 µm	100–140 × 7–8 µm	4.5–5 × 3.5–4 µm	16–21 × 5–6 µm	ellipsoidal	1–septate	hyaline	none	thin
Annulatascus joannae	180–200 × 150–250 µm	150–200 × 40–60 µm	4 × ? µm	150–200 × 10–13 µm	2.5–4 × 5–6.5 µm	20–28 × 9–12 µm	ellipsoidal to fusiform	aseptate	hyaline	none	thin
Annulatascus lacicola	220–250 × 225–250 µm	175–200 × 30–40 µm	5–7.5 × 75–88 µm	75–100 × 19–25 µm	2.5–4 × 3–4 µm	35–38 × 13–16 µm	fusiform	2–3 septate	hyaline	none	none observed
Annulatascus lacteus	140–200 × 100–130 µm	50 × 40–50 µm	2–3 × ? µm	130–170 × 9–10.5 µm	3–4 × 4–5 µm	24–28 × 6–8 µm	fusiform	aseptate	hyaline	none	none observed

Table 7 continued.

Species	**Ascoma dimensions**	**Neck dimensions**	**Paraphyses dimensions**	**Ascus dimensions**	**Annulus dimensions (l × w)**	**Ascospore dimensions**	**Ascospore shape**	**Ascospore septation**	**Ascospore coloration**	**Appendages**	**Sheath**
Annulatascus licualae	77.5–95 × 202–460 µm	[? × 56–82 µm]	2.6–3.6 × 74–120 µm	88.8–125 × 5.8–7.8 µm	1.2–2 × 2.5–2.75µm	15–17.8 × 3.8–5 µm	fusoid–rhomboid	aseptate	hyaline	none	thin, inconspicuous
Annulatascus liputii	180–260 × 200–300 µm	250–400 × 45–70 µm	4–5 × ? µm	130–187.5 × 8.5–10 µm	2 × 3.5 µm	15–22.5 × 6.5–7.5 µm	fusiform	0–2 septate	hyaline	none	thin
Annulatascus menglensis	120– 150 µm wide × 180 – 200 µm high	150–340× 40–60 µm	5–6 µm wide	115–163 × 10–13 µm	3 × 4 µm	21–25 × 8–10 µm	fusiform	aseptate	hyaline	none	none observed
Annulatascus nilensis	400–600 × 220–280 µm	240–360 × 96–112 µm	3–12 × ? µm	260–400 × 12–14 µm	3–4 × 5–6 µm	32–52 × 7–10 µm	fusoid	5–9–11 septate	hyaline	none	large, irregular, granular
Annulatascus palmietensis	150–440 × ? µm	200 × 70 µm	3 × ? µm	98–142 × 7–10.5 µm	3–4 × 4–4.5 µm	20–26 × 6–7 µm	short fusiform	3–septate	hyaline	none	none observed
Annulatascus triseptatus	222–353 × ? µm	110 × ? µm	3 × ? µm	140–218 × 9–14 µm	3 × 5.5 µm	18–33 × 6–12 µm	fusiform	3–septate	hyaline	none	thin
Annulatascus tropicalis	263–275 × 250–263 µm	100–150 × 30–50 µm	6–7.5 × 163–200 µm	190–255 × 12–18 µm	3–5 × 3–5 µm	42.5–52.5 × 7.5–10 µm	fusiform	1–3 septate	hyaline	none	none observed
Annulatascus velatisporus	260–410 µm × ? µm	384 × 140 µm	"wide"	150–290 × 10–18 µm	6–9 × 4–6 µm	21–30 × 8–11 µm	fusiform	0–3 septate	hyaline	none	thin, expanding
Annulusmagnus triseptatus	375–660 × 400–700 µm	88–340 × 80–150 µm	4–10 × 165–210 µm	138–283 × 7–14 µm	2–4 × 3–5 µm	16–37 × 5–10 µm	fusoid	3–septate	hyaline to pale brown	none	thin, adpressed
Aqualignicola hyalina	140–150 × 120–125 µm	150–180 × 35–40 µm	3.75–5 × 95–100 µm	137.5–150 × 9–12 µm	3–3.5 × 3.5–4 µm	14–15 × 6.25–7.5 µm	ellipsoidal to fusiform	aseptate	hyaline	none	none observed
Aqualignicola vaginata	100–140 µm × 100–140 µm	200–350 × 40–60 µm	4–6 × 84–100 µm	145–156 × 5.5–6.5 µm	?	11–15 × 5–6 µm	ellipsoidal–fusiform	aseptate	hyaline	none	unipolar sheath
Aquaticola ellipsoidea	150–200 × 150–250 µm	?	5 × ? µm	90–125 × 10–12.5 µm	"small"	12–14 ×5–7 µm	ellipsoidal	aseptate	hyaline	none	none observed

Table 7 continued.

Species	**Ascoma dimensions**	**Neck dimensions**	**Paraphyses dimensions**	**Ascus dimensions**	**Annulus dimensions (l × w)**	**Ascospore dimensions**	**Ascospore shape**	**Ascospore septation**	**Ascospore coloration**	**Appendages**	**Sheath**
Aquaticola hyalomura	160–290 × 160–300 µm	100–150 × 45–65 µm	2–3.5 × ? µm	52–64 × 7–8 µm	2 × 4 µm	10–14 × 5–7 µm	ellipsoidal	aseptate	hyaline	none	thin
Aquaticola hongkongensis	250–275 × 250–263 µm	100–120 × 30–40 µm	6–8 × 143–150 µm	150–175 × 13–15 µm	3–4 × 3–4 µm	23–26 × 7.5–11 µm	ellipsoidal	aseptate	hyaline	none	thin
Aquaticola longicolla	212–248 × ? µm	1200 × ? µm	7.5 × ? µm	75–83 × 9–12 µm	1.5 × 3 µm	12–16.5 × 4.5–6 µm	ellipsoidal to fusiform	aseptate	hyaline	none	none observed
Aquaticola miniguttulata	150–220 × 200–300 µm	150–300 × 25–30 µm	10 × 150 µm	96–130 × 10–12 µm	2–2.5 × 3–4 µm	9–12 × 6–8 µm	ellipsoidal	aseptate	hyaline	none	thin, (spreading wide)
Aquaticola rhomboida	80–140 × 160–260 µm	60–140 × 10–14 µm	3–4.5 × ? µm	80–125 × 8.7–10 µm	3–3.5 × 2–2.5 µm	16–20 × 7–8.5 µm	rhomboid	aseptate	hyaline	none	none observed
Aquaticola triseptata	200–240 × 240–320 µm	240 × 60 µm	5 × ? µm	50–80 × 8–12 µm	3 × 4 µm	15–19 × 5–7 µm	ellipsoidal	3–septate	hyaline	none	thin
Ascitendus austriacus	350–585 × 295–505 µm	85–300 × 50–140 µm	4–10 × 120–190 µm	100–260 × 4–8 µm	2.5–3.5 × 3–3.8 µm	14–27 × 4–9 µm	fusoid	3–septate	pale brown, middle cells darker than end cells	none	none observed
Ascocollumdensa aquatica	210–225 × 470–510 µm	230–250 × 220–250 µm	5.6–6.6 × 120–150 µm	210–215 × 12.5–15 µm	25. × 5 µm	25–30 × 6–10	fusiform–ellipsoidal	5–6 septate	hyaline	none	none observed
Ascolacicola aquatica	250–375 × 225–275 µm	175–200 × 62.5–75 µm	2.5–3.8 × 175–200 µm	125–163 × 10–12.5 µm	2.5–3.7 × 6.2 µm	12.5–16.5 × 4–7.5 µm	ellipsoidal	3–septate	brown with pale end cells	none	none observed
Ayria appendiculata	150 × ? µm	150 × 44 µm	5–5.5 × ? µm	115–150 × 16–29 µm	? µm	21–26 × 7.5–10 µm	ellipsoid	aseptate	hyaline	bipolar unravelling	none observed
Ayria nubispora	250–270 × 158–168 µm	40–50 × 30–60 µm	7–9 × 90–120 µm	212–250 × 34–40 µm	lacking	48–56 × 16–20 µm	ellipsoidal to fusiform	aseptate	hyaline	bipolar unravelling	10–18 µm
Brunneosporella aquatica	250–275 × 175–188 µm	130–150 × 35–40 µm	4–5 × 150–163 µm	150–175 × 10–13 µm	2–3 × 2–3 µm	17.5–20 × 9–10 µm	ellipsoidal to fusiform	1–septate	brown	lacking	lacking

Table 7 continued.

Species	**Ascoma dimensions**	**Neck dimensions**	**Paraphyses dimensions**	**Ascus dimensions**	**Annulus dimensions (l × w)**	**Ascospore dimensions**	**Ascospore shape**	**Ascospore septation**	**Ascospore coloration**	**Appendages**	**Sheath**
Cataractispora appendiculata	224–390 × 128–167 µm	short, lateral	5 × ? µm	129–195 × 11–20 µm	4.5 × 6 µm	23–30 × 7–10 µm	fusiform	aseptate	hyaline	bipolar unravelling	none observed
Cataractispora aquatica	240–370 × ? µm	short	3.3 × ? µm	344–366 × 11–12 µm	5 × 6 µm	36–65 × 8–14 µm	fusiform	0–5 septate	hyaline	bipolar unravelling	none observed
Cataractispora bipolaris	155–235 × ? µm	long black	4 × ? µm	174–205 × 9–10.5 µm	4 × 6 µm	21–30 × 6.5–8.5 µm	fusiform	aseptate	hyaline	pads to strands	none observed
Cataractispora receptaculorum	275–300 × 250–275 µm	250–275 × 113–125 µm	4–5 × 175–200 µm	213–233 × 15–22.5 µm	5–7.5 × 3.8–4 µm	27.5–31 × 7.5–10 µm	ellipsoidal	3–septate	hyaline	pads to strands	none observed
Cataractispora viscosa	160–260 × ? µm	50 µm	2 × ? µm	126–230 × 8–12 µm	5 × 6.5 µm	25–34 × 6–6 .5µm	fusiform	5–septate	hyaline	thin unfurling	none observed
Chaetorostrum quincemilense	200–270 × 800–900 µm	600–700 × 64–70	5–7 wide × 137–162 long	180–240 × 12–15 µm	5–6 long × 7–8 µm wide	30–38 µm × 10–12 µm	broadly ellipsoidal	3–septate	brown central cells and hyaline end cells	gelatinous apiculate appendages	adpressed ephemeral sheath
Clohiesia corticola	200 × ? µm	200 × 120 µm	? µm	125 × 7.5 µm	? µm	18.5–26.5 × 3.7–5 µm	curved fusiform	aseptate	hyaline	none	thin spreading
Clohiesia curvispora	400–600 × 400–500 µm	short	3–4 µm	125–175 × 8.5–10.5 µm	1.5 × 2.5–3 µm	32.5–39 × 3.2–5 µm	curved elongate fusiform	aseptate	hyaline	none	thin
Clohiesia lignicola	440–500 × 400–440 µm	? × 20–30 µm	5–8 × 200 µm	120–140 × 8–12 µm	2 × 3–4 µm	14–28 × 4.5–6 µm	fusoid ellipsoidal	aseptate	hyaline	none	thin
Cyanoannulus petersenii	405–800 × 214–297 µm	340–760 × 25–65 µm	9–12 × 55–110 µm	73–142 × 8–12 µm	2–3 × 4–5 µm	20–26.5 × 5–7 µm	fusiform	3–septate	hyaline	none	3–5 µm
Diluviicola capensis	93–128 × 32–80 µm	short hyaline	4 × ? µm	266–326 × 11–14 µm	3–4 × 2–3 µm	26–34 × 6–11 µm	fusiform	aseptate	hyaline	bipolar unravelling	none observed

Table 7 continued.

Species	**Ascoma dimensions**	**Neck dimensions**	**Paraphyses dimensions**	**Ascus dimensions**	**Annulus dimensions (l × w)**	**Ascospore dimensions**	**Ascospore shape**	**Ascospore septation**	**Ascospore coloration**	**Appendages**	**Sheath**
Fluminicola bipolaris	75–177 × 39–80	short black	11 – 5 × μm ? μm	107–192 × 9–12 μm	3–6 × 1.5–3 μm	15–21 × 6–9 μm	fusiform	1–3 septate	hyaline	apical cups	none observed
Frondicola tunitricuspis	340–620 × 155–280 μm	short	6 × ? μm	144–181 × 7.1–11.3 μm	? μm	17–26 × 4.8–7.1 μm	ellipsoidal	aseptate (3)	hyaline	none	12–13 × 9–12 μm w/ 3 extensions
Fusoidispora aquatica	115–215 × 80–150 μm	75 × 45 μm	4.5 × ? μm	150–178 × 8.5–11 μm	? × 2 μm	42–50 × 4–6 μm	fusoid to sickle shaped	0–5 septate	hyaline	apical pads	none observed
Longicollum biappendiculatum	205–220 tall × 155–160 wide	315–370 long × 74–82 wide	5–7 × 140–170	180–225 × 15–17 μm	2–4 long × 3–5 wide	26–35 × 12–14	broadly ellipsoidal	Aseptate	hyaline	short, ephemeral, bipolar mucilaginous, 3–6 × 6–9	none observed
Mirannulata samuelsii	550–630 tall × 480–525 wide	100–150 tall × 120–130 wide at ape×, 250–300 at base	3–4.5	140–185 × 13–18 μm	4.8–6 long × 5–6.5 wide	33.6–43.3 × 5.4–7.3	fusiform	3–septate	hyaline	none	none observed
Paoayensis lignicola	546–626 high, 520–586 wide	?	3–10 wide	45–130 × 13–35 μm		53–90 × 30–58	lemoniform, with a 10–12 long germ slit	0–1–3 septate	brown to dark brown at maturity	none	none observed
Pseudoproboscispora aquatica	196–280 × ? μm	short hyaline	? μm	135–140 × 14–15 μm	3 × 5 μm	22.5–24 × 7.5–8 μm	fusiform	3–septate	hyaline	bipolar filamentous	none observed
Pseudoproboscispora caudae-suis	400–700 × 300–500 μm	short, 136.5–156 × 109–117 μm	5–6 × 150–200 μm	190–343 × 15–21 μm	? μm	18–36 × 9–18 μm	oval	1–septate, 3–septate	pale yellow	2–3 × 50–120 μm	none observed
Rivulicola aquatica	140–150 × 175–180 μm	50–88 × 50–60 μm	5.6–7 × 120–150 μm	125–162 × 22–25 μm	2.5–4 × 4–5 μm	15–20 × 7.5–12.5 μm	ellipsoidal	1–3 septate	hyaline	thin, fibrillar ornamentation	none observed

Table 7 continued.

Species	**Ascoma dimensions**	**Neck dimensions**	**Paraphyses dimensions**	**Ascus dimensions**	**Annulus dimensions (l × w)**	**Ascospore dimensions**	**Ascospore shape**	**Ascospore septation**	**Ascospore coloration**	**Appendages**	**Sheath**
Rivulicola cygnea	470–680 × 140–170 µm	240–500 × 30–40 µm	3 × ? µm	122–155 × 7–8 µm	? µm	20–24 × 6–7 µm	ellipsoidal	3–septate	hyaline	none	2–3 µm, constricted at first septum
Rivulicola incrustata	160 × 280 µm	green	4 × ? µm	130–205 × 11–12 µm	? × 4.1 µm	21–27 × 8–12 µm	ellipsoidal	3–5 septate	hyaline	none	irregular granular
Submersisphaeria aquatica	180–250 × ? µm	70 × 560 µm	3–4 × ?µm	175–210 × 10–12.5 µm	4–5 × 6–7 µm	23–27 × 7.5–10 µm	ellipsoidal to fusiform	1–septate	brown	hyaline germ pores	none observed
Submersisphaeria bambusicola	420–580 × 350–540 µm	210 × 60 µm	4–8 × ? µm	235–290 × 10–12 µm	5 × 5 µm	28–36 × 6–8 µm	ellipsoidal to fusiform	aseptate	dark brown	hyaline germ pores	bipolar thin
Submersisphaeria palmae	200–300 × ? µm	?	2.5–3.75 × ? µm	100–127.5 × 6.25–8.75 µm	1–2 × 4–5 µm	17.5–22.5 × 5–7.5 µm	ellipsoidal	aseptate	uniseriate	bipolar pads	none observed
Submersisphaeria rattanicola	104–164 × 94–112 µm	?	1.2–1.6 × ? µm	123.8–154.5 × 7–8.5 µm	2–2.75 × 3.25–4.75 µm	14.3–20.8 × 5–6.8 µm	fusiform to ellipsoid	1–septate	brown	hyaline apical germ pores	none observed
Submersisphaeria vasicola	250 × ? µm	?	3.5 × ? µm	140 × 8 µm	?	16–22 × 6–7 µm	cylindro-ellipsoidal	1–septate	brown	none	present
Teracosphaeria petroica	450–600 × 400–550 µm	600–900 × 190–210 µm	5–6 × ? µm	98–132 × 12–17 µm	1.5 × 4–5 µm	15–20 × 5–6 µm	ellipsoidal	3–septate	hyaline	none	none observed
Torrentispora crassiparietis	220–315 × ? µm	long black (200 × 50 µm from micrograph	3 × ? µm	212–300 × 10.5–12 µm	7–8 × 7–9 µm	31–42.5 × 7.5–10 µm	ellipsoidal to fusiform	aseptate	hyaline	none	thin fibrillar
Torrentispora fibrosa	135–255 × ? µm	long black	5.5 × ? µm	154–254 × 6–9 µm	2.5 × 4.5 µm	13.5–19.5 × 5–7 µm	ovoid to fusiform	aseptate	hyaline	none	narrow fibrillar
Torrentispora fusiformis	220–315 × ? µm	long black	4 × ? µm	200–230 × 9–10 µm	4 × 6 µm	24–32.5 × 6–9 µm	fusiform	aseptate	hyaline	none	none observed
Torrentispora pilosa	247–450 × 320–400 µm	44—770 × 55–180 µm	4–6 × ?	164–204 × 7–8 µm	3–4 × 4–5 µm	21–30 × 7–8 µm	ellipsoidal to fusiform	0–2	hyaline	none	none observed

Table 7 continued.

Species	**Ascoma dimensions**	**Neck dimensions**	**Paraphyses dimensions**	**Ascus dimensions**	**Annulus dimensions (l × w)**	**Ascospore dimensions**	**Ascospore shape**	**Ascospore septation**	**Ascospore coloration**	**Appendages**	**Sheath**
Vertexicola aquatica	250–275 × 250–263 µm	100–120 × 30–40 µm	4–5 × 120–250 µm	125–150 × 8–9 µm	4–5 × 3–4 µm	15–20 × 7.5–10	fusiform	3–5 septate	hyaline	none	none observed
Vertexicola ascoliberatus	737–869 × 214–285 µm	500–550 × 100–120 µm	4–9 × ? µm	194–273 × 10–12 µm	4–5 × 6–8 µm	30–34 × 10–12 µm	ellipsoidal to fusiform	0 (3)	hyaline	none	present
Vertexicola caudatus	240–360 × ? µm	80–120 × 40–60 µm	6 × ? µm	170–290 × 7.5–12 µm	4 × 5 µm	18–24 × 6–9 µm	ellipsoidal to fusiform	5–septate	hyaline	none	none observed

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