

# Towards a natural classification and backbone tree for *Pleosporaceae*

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**Abstract** The family *Pleosporaceae* includes numerous saprobic, opportunistic human, and plant pathogenic taxa. The classification of genera and species *Pleosporaceae* has been a major challenge due to the lack of a clear understanding of the importance of the morphological characters used to distinguish taxa as well as the lack of reference strains. Recent treatments concluded that *Pleospora* and some other genera in *Pleosporaceae* are likely polyphyletic. In order to establish the evolutionary relationships and to resolve the polyphyletic nature of *Pleospora* and allied genera, we sequenced the 18S nrDNA, 28S nrDNA, ITS, GAPDH, RPB2 and TEF1-alpha gene regions of *Pleosporaceae* species and phylogenetically

analysed this data. Multigene phylogenies strongly support the monophyletic nature of *Pleosporaceae* among the other families in *Pleosporales*, and the acceptance of the genera *Alternaria*, *Bipolaris*, *Clathrospora*, *Comoclathris*, *Curvularia*, *Dactuliophora*, *Decorospora*, *Diademosia*, *Exserohilum*, *Extrawettsteinina*, *Gibbago*, *Neocamarosporium*, *Paradendryphiella*, *Platysporoides*, *Pleospora*, *Porocercospora*, *Pseudoyuconia* and *Pyrenophora*. *Austropleospora*, *Dendryphion*, *Edenia* and *Macrospora* are excluded from the family based on morphology coupled with molecular data. Two novel species, *Alternaria murispora* in this paper and *Comoclathris sedi*

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are introduced. The sexual morph of *Alternaria alternata* is re-described and illustrated using modern concepts from fresh collections. The paraphyletic nature of *Pleospora* is resolved based on the available morpho-molecular data, but further sampling with fresh collections, reference or ex-type strains and molecular data are needed to obtain a natural classification of genera and the family.

**Keywords** Backbone tree · Dothideomycetes · Pleosporales · Plant pathogens · Saprobes

## Introduction

*Pleosporaceae* is the largest family in the *Pleosporales* and is representative of the order (Zhang et al. 2012; Hyde et al. 2013; Wijayawardene et al. 2014). The classification in the *Pleosporaceae* has primarily been based on the *Pleospora* type of centrum development (Dong et al. 1998) and asci that are interspersed with pseudoparaphyses in the ascomata. These pseudoparaphyses originate above the hymenial layer and grow downward among the asci to fuse at the base of the locule (Wehmeyer 1975). Ascomata are perithecial, initially immersed and become erumpent and are usually black and sometimes hairy or setose. Asci are bitunicate and fissitunicate, cylindrical, with an ocular chamber and the hamathecium comprises cellular pseudoparaphyses. Ascospores are usually brown and phragmosporous or dictyosporous (Dong et al. 1998; Kirk et al. 2001). Species are pathogenic or saprobic on wood and dead herbaceous stems or leaves, as well as pathogenic in humans (Sivanesan 1984; Carter and Boudreaux 2004).

## Historic overview of *Pleosporaceae*

The family *Pleosporaceae* was introduced by Nitschke (1869) based on the immersed ascomata and presence of pseudoparaphyses, and was classified in *Sphaeriales*. The family was transferred to *Pseudosphaeriaceae* and later raised to ordinal rank as the *Pseudosphaeriales* (Theissen and Sydow 1918). *Pleosporaceae*, *Venturiaceae* and *Lophiostomataceae* were assigned under *Pleosporales* by Luttrell (1955), while treating *Pseudosphaeriales* as a synonym of *Pleosporales*. Luttrell (1973) included eight families in *Pleosporales* including *Pleosporaceae*. Preliminary genera added to this family were based on ascospore characteristics, including shape, colour, septation, pigmentation and presence or absence of mucilaginous sheaths (Luttrell 1955, 1973; Wehmeyer 1961, 1975; Eriksson 1981; Sivanesan 1984; Barr

1987b; Abler 2003). Many of these characters were also found in other families, such as *Leptosphaeriaceae*, *Melanommataceae*, *Phaeosphaeriaceae* and *Sporormiaceae*. This led for confusion in the intergeneric and familial classification (Luttrell 1955, 1973; Wehmeyer 1961, 1975; von Arx and Müller 1975; Sivanesan 1984; Barr 1987a, b; Eriksson and Hawksworth 1986, 1991). Barr (1987b) redefined the *Pleosporaceae* to include *Clathrospora* (= *Comoclathris*), *Kirschsteiniothelia*, *Lewia* and *Pleospora* and grouped *Cochliobolus*, *Pyrenophora* and *Setosphaeria* into the family *Pyrenophoraceae*. Berbee (1996) disagreed, suggesting that all those genera belong to the *Pleosporaceae*. Kodsueb et al. (2006) showed the intergeneric relationships and phylogenetic perspectives of the family *Pleosporaceae* based on sequence analyses of partial 28S rDNA and accepted 14 genera in *Pleosporaceae*. Based on multi-gene phylogenetic analysis, Zhang et al. (2009) concluded that some species from *Lewia*, *Cochliobolus*, *Pleospora*, *Pyrenophora* and *Setosphaeria* resided in the *Pleosporaceae*. Lumbsch and Huhndorf (2010) accepted 13 genera in *Pleosporaceae* but Zhang et al. (2012) included only 10 genera by excluding *Monascostroma*, *Kriegeriella*, and *Zeuctomorpha* (see notes below). Hyde et al. (2013) included 23 genera in this family based on morphology coupled with molecular data. Multi-gene phylogenetic studies has shown that the familial placement of *Pleosporaceae* with respect to other families in order *Pleosporales* is valid (Lumbsch and Huhndorf 2010; Zhang et al. 2012; Hyde et al. 2013). Major circumscription changes of the genera in *Pleosporaceae* from 1961 to 2013 are given in Table 1.

## Asexual morphs of *Pleosporaceae*

The asexual morphs of *Pleosporaceae* can be coelomycetous or hyphomycetous (Zhang et al. 2012; Hyde et al. 2013) and some sexual genera in *Pleosporaceae* have been linked with asexual morphs (Zhang et al. 2012; Hyde et al. 2013, 2014). The type species of the family *Pleospora* is linked to *Stemphylium*, which causes leaf disease (Sivanesan 1984). *Bipolaris* was shown to be the asexual morph of “*Cochliobolus*”, and the cause of plant disease or infection in human beings (Khan et al. 2000; Manamgoda et al. 2014). The nomenclatural conflict in this complex was resolved by giving priority to the more commonly used established generic name *Bipolaris* (Manamgoda et al. 2012a, b; 2014). At the same time, Manamgoda et al. (2012a, b) showed that *Curvularia* grouped with “*Pseudocochliobolus*”. The type species of *Pleoseptum* (*P. yuccaesedum* A.W. Ramaley & M.E. Barr) has been linked with *Camarosporium yuccaesedum* Fairm (Ramaley and Barr 1995) and *Pyrenophora* has the asexual morph *Drechslera* (Farr et al. 1989). Ariyawansa et al. (2014c) proposed to

**Table 1** Major circumscription changes of the genera in *Pleosporaceae*

Wehmeyer 1961	Wehmeyer 1975	Eriksson and Hawksworth 1998	Kirk et al. 2001	Kodsueb et al. 2006	Lumbsch and Huhndorf 2010	Zhang et al. 2012	Hyde et al. 2013	Wijayawardene et al. 2014	This study
<i>Clathrospora</i>	<i>Allonecta</i>	<i>Cochliobolus</i>	<i>Cochliobolus</i>	<i>Cochliobolus</i>	<i>Cochliobolus</i>	<i>Cochliobolus</i>	<i>Alternaria</i>	<i>Alternaria</i>	<i>Alternaria</i>
<i>Platyospora</i>	<i>Catharinia</i>	<i>Extrawetsteinina</i>	<i>Extrawetsteinina</i>	<i>Decorospora</i>	<i>Crivellia</i>	<i>Crivellia</i>	<i>Alternaria</i>	<i>Bipolaris</i>	<i>Bipolaris</i>
<i>Pleospora</i>	<i>Clathrospora</i>	<i>Falciformispora</i>	<i>Falciformispora</i>	<i>Extrawetsteinina</i>	<i>Decorospora</i>	<i>Extrawetsteinina</i>	<i>Alternariaster</i>	<i>Clathrospora</i>	<i>Clathrospora</i>
<i>Pyrenophora</i>	<i>Cochliobolus</i>	<i>Kirschsteintothelia</i>	<i>Kirschsteintothelia</i>	<i>Falciformispora</i>	<i>Extrawetsteinina</i>	<i>Lewia</i>	<i>Bipolaris</i>	<i>Curvularia</i>	<i>Comoclathris</i>
	<i>Cucurbitaria</i>	<i>Kriegeriella</i>	<i>Kriegeriella</i>	<i>Kriegeriella</i>	<i>Kriegeriella</i>	<i>Macrospora</i>	<i>Brachycladium</i>	<i>Decorospora</i>	<i>Curvularia</i>
	<i>Didymoplella</i>	<i>Leptosphaerulina</i>	<i>Leptosphaerulina</i>	<i>Lewia</i>	<i>Lewia</i>	<i>Platysporoides</i>	<i>Chalastospora</i>	<i>Decorospora</i>	<i>Dactuliothpora</i>
	<i>Eudartluca</i>	<i>Lewia</i>	<i>Lewia</i>	<i>Macrospora</i>	<i>Macrospora</i>	<i>Pleospora</i>	<i>Clathrospora</i>	<i>Edenia</i>	<i>Decorospora</i>
	<i>Fenestella</i>	<i>Macrospora</i>	<i>Macrospora</i>	<i>Monacostroma</i>	<i>Platysporoides</i>	<i>Pseudoyuconia</i>	<i>Decorospora</i>	<i>Exserohilum</i>	<i>Diademosa</i>
	<i>Gibberidea</i>	<i>?Macroventuria</i>	<i>Macroventuria</i>	<i>Platysporoides</i>	<i>Pleospora</i>	<i>Pyrenophora</i>	<i>Dendryphion</i>	<i>Extrawetsteinina</i>	<i>Exserohilum</i>
	<i>Gilletiella</i>	<i>Monascostroma</i>	<i>Platysporoides</i>	<i>Pleospora</i>	<i>Pseudoyuconia</i>	<i>Setosphaeria</i>	<i>Edenia</i>	<i>Mariellioitia</i>	<i>Extrawetsteinina</i>
	<i>Herpotrichia</i>	<i>Platysporoides</i>	<i>Pleospora</i>	<i>Pseudoyuconia</i>	<i>Pyrenophora</i>	<i>Setosphaeria</i>	<i>Embellisia</i>	<i>Neocamarosporium</i>	<i>Gibbago</i>
	<i>Herpotrichiella</i>	<i>Pleospora</i>	<i>Pseudoyuconia</i>	<i>Pyrenophora</i>	<i>Setosphaeria</i>	<i>Zeuctomorpha</i>	<i>Exserohilum</i>	<i>Paradendryphiella</i>	<i>Neocamarosporium</i>
	<i>Letendrea</i>	<i>Pseudoyuconia</i>	<i>Pyrenophora</i>	<i>Setosphaeria</i>	<i>Zeuctomorpha</i>		<i>Extrawetsteinina</i>	<i>Pleoseptium</i>	<i>Paradendryphiella</i>
	<i>Licopolia</i>	<i>Pyrenophora</i>	<i>Setosphaeria</i>	<i>Zeuctomorpha</i>			<i>Macrospora</i>	<i>Platysporoides</i>	<i>Platysporoides</i>
	<i>Melanomma</i>	<i>Setosphaeria</i>	<i>Teratosphaeria</i>				<i>Mariellioitia</i>	<i>Pseudoyuconia</i>	<i>Pleospora/ Stenphylium</i>
	<i>Melanopsamma</i>	<i>Wetsteinina</i>	<i>Tremateia</i>				<i>Platysporoides</i>	<i>Pyrenophora</i>	<i>Porocerospora</i>
	<i>Metasphaeria</i>	<i>Zeuctomorpha</i>	<i>Wetsteinina</i>				<i>Pleospora</i>	<i>Stemphylium</i>	<i>Pseudoyuconia</i>
	<i>Mycomicrothelia</i>		<i>Zeuctomorpha</i>				<i>Pseudoyuconia</i>	<i>Pyrenophora</i>	<i>Pyrenophora</i>
	<i>Ophiobolus</i>						<i>Pyrenophora</i>	<i>Setosphaeria</i>	
	<i>Othitia</i>						<i>Setosphaeria</i>	<i>Simomyces</i>	
	<i>Paralimyces</i>						<i>Teretispora</i>	<i>Ulocladium</i>	
	<i>Platyospora</i>								
	<i>Pleomassaria</i>								
	<i>Pleospora</i>								
	<i>Pteridiospora</i>								
	<i>Pyrenophora</i>								
	<i>Teichospora</i>								
	<i>Teratosphaeria</i>								
	<i>Thyridaria</i>								
	<i>Thyridium</i>								
	<i>Tomasella</i>								
	<i>Trematosphaeria</i>								
	<i>Trichometasphaeria</i>								
	<i>Zignoella</i>								

conserve *Pyrenophora* over *Drechslera* by giving priority to the oldest name. Based on the combined gene analysis of GAPDH, RPB2 and TEF1, Woudenberg et al. (2013) synonymised *Allewia*, *Brachycladium*, *Chalastospora*, *Chmelia*, *Crivellia*, *Embellisia*, *Lewia*, *Nimbya*, *Sinomyces*, *Teretispora*, *Ulocladium*, *Undifilum* and *Ybotromyces* under *Alternaria*. In same study, Woudenberg et al. (2013) treated the 24 internal clades in the *Alternaria* complex as sections, which is a continuation of a recent proposal for the taxonomic treatment of lineages in *Alternaria*. Furthermore, *Alternariaster*, a genus formerly seen as part of *Alternaria* was transferred to *Leptosphaeriaceae* based on molecular data. Recently, new asexual genera were introduced by different researches based on both morphology and phylogeny. *i.e.* *Porocercospora* was introduced as a new genus in *Pleosporaceae* by Amaradasa et al. (2014) to accommodate the buffalo grass false-smut pathogen, while *Johnalcornia* was introduced to accommodate *Bipolaris aberrans*, which clusters sister to the newly described *Porocercospora* (Tan et al. 2014). *Paradendryphiella* was introduced by Woudenberg et al. (2013) to accommodate two marine *Dendryphiella* species (*D. arenariae* Nicot, *D. salina* (G.K. Sutherl.) Pugh & Nicot) that did not group with the type species *D. vinosa* (Berk. & M.A. Curtis) Reisinger (Jones et al. 2008; Woudenberg et al. 2013).

Taxa in *Alternaria*, *Bipolaris*, *Stemphylium* and phoma like species are more common asexual morphs in *Pleosporaceae* and can be saprobic or parasitic on various hosts. *Phoma betae* A.B... Frank is a notorious pathogen of sugar beet, which causes zonate leaf spot. *Alternaria porri* (Ellis) Cif., *Stemphylium solani* G.F. Weber, *S. botryosum* Wallr and *S. vesicarium* (Wallr.) E.G. Simmons can cause leaf blight of garlic (Zhang et al. 2009). *Phoma incompta* Sacc & Martelli is a pathogen on olive, and *S. botryosum*, the asexual morph of *Pleospora herbarum* (Pers.) Rabenh, causes leaf disease of olive trees (Malathrakis 1979). Some *Curvularia* species have been reported as human pathogens, causing respiratory tract, cutaneous, and corneal infections (Carter and Boudreaux 2004).

We have been studying the families of *Dothideomycetes* based on morphology and molecular phylogeny, in order to provide a natural classification of this large class. Hyde et al. (2013) provide an account of all 105 families of *Dothideomycetes*, while Wijayawardene et al. (2014) provided an outline of *Dothideomycetes* and suggestions on the on the names to use where asexual and sexual genera were linked. The largest order in *Dothideomycetes* is *Pleosporales* and we also have provided a natural classification of this large order (Ariyawansa et al. 2013a, b, c; Ariyawansa et al. 2014a, b, c, d, e, f; Zhang et al. 2012). Zhang et al. (2012) provided an account for 105 genera in *Pleosporales*, however, at that time little verified molecular data were available. The aim of the present study was to delineate the phylogenetic lineages

within *Pleosporaceae*, and to build a robust taxonomy to act as a backbone tree for the family. Phylogenetic analysis were conducted using sequence data from parts of the 18S nrDNA (SSU), 28S nrDNA (LSU), the internal transcribed spacer regions 1 and 2 and intervening 5.8S nrDNA (ITS) and RNA polymerase second largest subunit (RPB2) gene regions of extype, reference and putative strains of all available allied genera in *Pleosporaceae*. We also focused on the phylogenetic lineages within *Alternaria* and allied genera, and to create a stable taxonomy. Phylogenetic inferences were conducted on sequence data of parts of the SSU, LSU, ITS, glyceraldehyde-3-phosphate dehydrogenase (GAPDH), RPB2 and translation elongation factor 1-alpha (TEF1) gene regions of extype and reference strains of *Alternaria* species and all available allied genera.

## Material and methods

### Specimen examination

Fresh material of species of *Pleosporaceae* were collected in Thailand, Germany and Italy during 2011–2014. Specimens were taken to the laboratory in Ziplock plastic bags. The samples were processed and examined following the method described in Ariyawansa et al. (2013a, b). Fresh and herbarium material were examined under a Motic SMZ 168 dissecting microscope to locate and isolate ascomata fruiting bodies. Hand sections of the fruiting structures were mounted in water for microscopic studies and photomicrography. The taxa were examined using a Nikon ECLIPSE 80i compound microscope and photographed with a Canon 450D digital camera fitted to the microscope. Measurements were made with the Tarosoft (R) Image Frame Work program and images used for figures processed with Adobe Photoshop CS3 Extended version 10.0 software (Adobe Systems, USA). Isolations were made from single ascospores, following a modified method of Ariyawansa et al. (2013b) and Chomnunti et al. (2014). Contents of the sectioned fruiting body were transferred to a drop of sterile water on a flame-sterilized slide. Drops of the spore suspension were pipetted and spread on a Petri-dish containing 2 % water agar (WA) and incubated at 25 °C. Germinated ascospores were transferred singly to MEA media (Alves et al. 2006).

Herbarium specimens were obtained on loan from the Swedish Museum of Natural History (S) and the New York Botanical Garden (NY). Voucher specimens are deposited in the herbarium of Mae Fah Luang University (MFLU), Chiang Rai, Thailand, Kunming Institute of Botany (KIB) and New Zealand Fungal Herbarium-Landcare Research (PDD), New Zealand. Living cultures are deposited at the Mae Fah Luang University Culture Collection (MFLUCC), International collection of microorganisms from plants (ICMP) and



Queensland Plant Pathology Herbarium (BRIP), the latter under Material Transfer Agreement No. 4/2010 (MTA). Each genus is listed along with a description of the type species, except in cases where there is only a single species in the genus.

#### DNA extraction, PCR amplification and sequencing

Single ascospore fungal isolates were grown on MEA or PDA for 28 days at 25 °C in the dark. Genomic DNA was extracted from the growing mycelium using the Biospin Fungus Genomic DNA Extraction Kit (BioFlux®) following the manufacturer's protocol (Hangzhou, P.R. China). Otherwise DNA extracted directly from ascospores using a DNA extraction kit (E.Z.N.A.® Forensic DNA kit, D3591-01, Omega Bio-Tek) following Telle and Thines (2008).

The amplification procedure was performed in a 50 µl reaction volume containing 5–10 ng DNA, 0.8 units Taq polymerase, 1X PCR buffer, 0.2 mM dNTP, 0.3 µM of each primer with 1.5 mM MgCl<sub>2</sub> (Cai et al. 2009). The PCR reactions for amplification of the recently ratified universal fungal barcode ITS1-5.8S-ITS2 of the nuclear ribosomal DNA operon (Schoch et al. 2009), were performed under standard conditions (White et al. 1990; Stielow et al. 2010). PCR conditions for amplifying the partial SSU and LSU r-DNA followed the protocol of Phillips et al. (2008). Amplification of GAPDH, RPB2 and TEF1 followed the protocol of Woudenberg et al. (2013). The PCR products were observed on 1 % agarose electrophoresis gels stained with ethidium bromide. Purification and sequencing of PCR products were carried at Shanghai Sangon Biological Engineering Technology and Services Co., (China).

DNA sequence data was obtained from the internal transcribe spacer (ITS), small and large subunits of the nuclear ribosomal RNA genes (SSU, LSU) and the protein coding gene, GAPDH (TUB). Primer sets used for these genes were as follows: ITS: ITS5/ITS4 SSU: NS1/NS4; LSU: LR0R/LR5; RPB2–5F2/rRPB2–7cR (Liu et al. 1999; Sung et al. 2007). The GAPDH region with *gpd1* and *gpd2* (Berbee et al. 1999) and the TEF1 gene with the primers EF1-728F and EF1-986R (Carbone and Kohn 1999). Primer sequences are available at the WASABI database at the AFTOL website (aftol.org). Sequences are deposited at NCBI GenBank under the accession numbers provided in Supplementary Table 1. Alignments are deposited in TreeBASE.

#### Sequence alignment and phylogenetic analysis

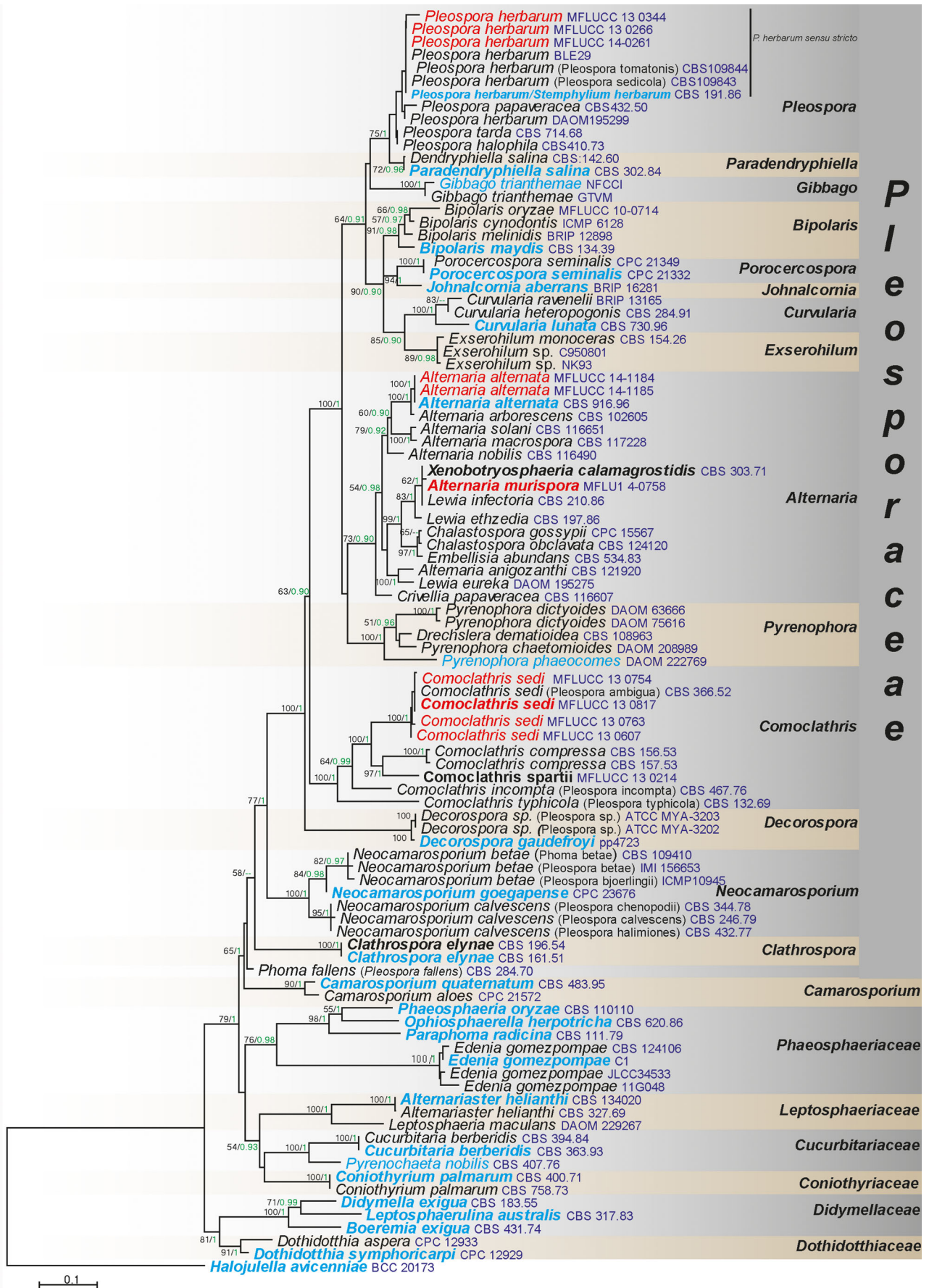
Multiple sequence alignments were generated with MAFFT v. 6.864b (<http://mafft.cbrc.jp/alignment/server/index.html>). The alignments were checked visually and improved manually where necessary. Two different datasets were used to estimate two phylogenies; a *Pleosporineae* family tree and

an *Alternaria* phylogeny. The first tree focuses on phylogenetic placement of *Pleosporaceae* in sub order *Pleosporineae*, the second one was generated to show the placement of newly reported *Alternaria* species and their sexual state. All introns and exons were aligned separately. Regions containing many leading or trailing gaps were removed from the ITS, SSU, LSU, GAPDH, TEF1 and RPB2 alignments prior to tree building. The alignments were checked visually and improved manually where necessary. All sequences obtained from GenBank and used by Ariyawansa et al. (2014a), Boonmee et al. (2014), Hyde et al. (2013), Phookamsak et al. (2014) Schoch et al. (2009), Suetrong et al. (2009), Verkley et al. (2014), and Zhang et al. (2012) are listed in supplementary Table 1.

Maximum likelihood analyses including 1000 bootstrap replicates were run using RAxML v. 7.2.6 (Stamatakis 2006; Stamatakis et al. 2008). The online tool Findmodel (<http://www.hiv.lanl.gov/content/sequence/findmodel/findmodel.html>) was used to determine the best nucleotide substitution model for each partition. The best scoring tree was selected with a final likelihood value of  $-19492.551787$ . The resulting replicates were plotted on to the best scoring tree obtained previously. Maximum Likelihood bootstrap values (ML) equal or greater than 50 % are given below or above each node in black (Fig. 1).

The model of evolution was performed by using MrModeltest 2.2 (Nylander 2004). Posterior probabilities (PP) (Rannala and Yang 1996; Zhaxybayeva and Gogarten 2002) were determined by Markov Chain Monte Carlo sampling (MCMC) in MrBayes v. 3.0b4 (Huelsenbeck and Ronquist 2001). Six simultaneous Markov chains were run for 5,000,000 generations and trees were sampled every 100th generation and 50,000 trees were obtained. The first 10,000 trees, representing the burn-in phase of the analyses, were discarded while the remaining trees were used for calculating posterior probabilities in the majority rule consensus tree (critical value for the topological convergence diagnostic set to 0.01) (Crous et al. 2006). Bayesian Posterior Probabilities (BYPP) equal or greater than 0.90 is given below or above each node (Fig. 1).

In order to determine the species limits in *Pleospora herbarum* species complex, *Alternaria* complex and the newly introduced *Comoclathris sedi*, we applied the criteria of Genealogical Concordance Phylogenetic Species Recognition (GCPSR) (Taylor et al. 2000; Dettman et al. 2003). Dettman et al. (2003) emphasised that species should be recognised if they satisfy one of two criteria: genealogical concordance or genealogical non-discordance. Clades were genealogically concordant if they were present in at least some of the gene trees and genealogically non-discordant if they were strongly supported ( $MP \geq 70\%$ ;  $ML \geq 70\%$ ) in a single gene and not contradicted at or above this level of support in any other single gene tree. This criterion prohibited poorly supported



**Fig. 1** RAxML tree based on a combined dataset of ITS, SSU, LSU and RPB2 of 98 strains representing the Pleosporineae. Bootstrap support values for maximum likelihood greater than 50 % (*black*) and bayesian posterior probabilities greater than 0.90 (*green*) below and above the nodes. *Halojulella avicemniae* is the out group taxon. The original isolate numbers are noted after the species names. Ex-type culture numbers are in bold. Newly generated strains in this study are indicated in *red*. The type species of each genus is indicated in *blue*

non-monophyly at one locus from undermining well-supported monophyly at another locus. Phylogenetic trees and data files were viewed in MEGA v. 5 (Tamura et al. 2011), TreeView v. 1.6.6 (Page 1996) and FigTree v. 1.4 (Rambaut and Drummond 2008).

## Results and discussion

### Phylogeny

#### Phylogeny of Pleosporineae

The final *Pleosporineae* alignment included 98 strains, representing seven families, and consisted of 3170 characters (SSU 941, LSU 863, ITS 493, RPB2 858). In the SSU alignment a large insertion at position 446 in the isolates *Chaetosphaeronema hispidulum* (Corda) Moesz (CBS 216.75), *Phoma fallens* Sacc (*Pleospora fallens*) (CBS 161.78) and *Ophiosphaerella herpotricha* (Fr.) J. Walker (CBS 620.86) was excluded from the phylogenetic analyses. Four different alignments corresponding to each individual gene, and a combined alignment of the four genes were analysed. Comparison of the alignment properties and nucleotide substitution models are provided in Table 2. Results of the partition-homogeneity test ( $P=0.107$ ) indicate that the ITS, LSU, SSU and RPB2 gene trees reflect the same underlying phylogeny. Therefore, these datasets were combined and analyzed by using several tree-building programs, the resulting trees compared and the best tree is presented in Fig. 1. New sequences are deposited in GenBank (Table 1).

The combined ITS, LSU, SSU and RPB2 gene dataset from seven families in the *Pleosporales* sub order *Pleosporaneae* is shown in Fig 1. All trees (ML and BYPP) were similar in topology and not significantly different (data not shown). A best scoring RAxML tree is shown in Fig. 1, with the value of  $-19492.551787$ . Phylogenetic trees obtained

from Maximum Likelihood and Bayesian analysis yielded trees with similar overall topology at subclass and family relationships in agreement with previous work based on Maximum Likelihood analysis (Schoch et al. 2009; Suetrong et al. 2009; Zhang et al. 2012; Ariyawansa et al. 2014a; Boonmee et al. 2014; Hyde et al. 2013, Phookamsak et al. 2014, Verkley et al. 2014; Wijayawardene et al. 2014). The support values for the different phylogenetic methods vary, with the Bayesian posterior probabilities being higher than the RAxML bootstrap support values.

In the multi-locus phylogeny inferred from the combined dataset shown in Fig. 1, several well-supported sub-clades can be identified in the family *Pleosporaceae*, which are interpreted as appropriate for the delimitation of genera, *i.e.* *Alternaria*, *Bipolaris*, *Comoclathris*, *Curvularia*, *Decorospora*, *Dendryphion*, *Exserohilum*, *Gibbago*, *Johnalcornia*, *Neocamarosporium*, *Porocercospora*, *Pleospora* and *Pyrenophora*.

The *Pleospora sensu stricto* clade comprises of eleven strains along with the type strain of *Pleospora herbarum*/*Stemphylium herbarum* (CBS 191.86) including our strains (MFLUCC 14–0261, MFLUCC 13–0344 and MFLUCC 13–0266). In order to determine the species limits of *Pleospora herbarum*, a combine gene tree (Fig. 1.) was used. Nodes that were supported ( $\geq 70\%$ ) in the combined gene phylogeny were recognised as taxa within *Pleospora herbarum sensu stricto*. Thus the putative strains of *P. tomatonis* E.G. Simmons (CBS 109844), *P. sedicola* E.G. Simmons (CBS 109843), which clustered within *Pleospora herbarum sensu stricto* are treated as strains of *Pleospora herbarum* in this study. Further work may yet prove this is a species complex.

A recently introduced genus, *Paradendryphiella* forms a basal clade to the *Pleospora sensu stricto* clade including the type strain of *Paradendryphiella salina* (CBS 302.84) and *Dendryphiella salina* (CBS 142.60). Another poorly-supported clade forming the major part of the ingroup of the tree comprises the putative strains of *Gibbago trianthemae* E.G. Simmons (NFCCI 1886 and GT-VM). The *Bipolaris* clade comprises four strains with *B. oryzae* (MFLUCC 10–0714), *B. cynodontis* (Marignoni) Shoemaker (ICMP 6128), *B. melinidis* Alcorn (BRIP 12898) and *B. maydis* (Y. Nisik. & C. Miyake) Shoemaker (CBS 134.39, previously known as *Cochliobolus heterostrophus*). Two monophyletic genera *Porocercospora seminalis* (Ellis & Everh.) Amaradasa et al.,

**Table 2** Comparison of alignment properties of genes and nucleotide substitution models used in *Pleosporineae* phylogenetic analysis

Genes /loci	LSU	SSU	ITS	RPB2
Alignment strategy (MAFFT v6)	FFT-NS-II	FFT-NS-I	FFT-NS-I+manual	FFT-NS-I+manual
Nucleotide substitution models for Bayesian analysis (determined by MrModeltest)	SYM+I+G	GTR+I+G	GTR+I+G	GTR+I+G



(CPC 21349 and CPC 21332) and *Johnalcornia aberrans* (Alcorn) Y.P. Tan & R.G. Shivas (BRIP 16281) form well-supported clades sister to the *Curvularia*, *Exserohilum* and *Bipolaris* clades. *Curvularia* forms a relatively well-supported clade within the family *Pleosporaceae*. This clade contains three strains with *C. lunata* (Wakker) Boedijn (CBS 730.96), the type species of *Curvularia*, and the highly supported clades of the following species: *C. ravenelii* (M.A. Curtis ex Berk.) Manamgoda et al. (BRIP 13165) and *C. heteropogonis* Alcorn (CBS 284.91). *Exserohilum* forms a well-supported clade sister to *Curvularia* containing the putative strains of *Setosphaeria monoceras* Alcorn (CBS 154.26), *Exserohilum* sp. (C950801) and *Exserohilum* sp. (NK93).

The recently redefined genus *Alternaria* forms a well-supported clade sister to *Pyrenophora* containing 18 strains with *A. alternata* (Fr.) Keissl (CBS 916.96) the type species of *Alternaria*, along with the three sexual strains of *A. alternata* (MFLUCC 14–1184, MFLUCC 14–1185 and ICMP). Apart from *A. alternata* strains, the clade consist of *A. arborescens* E.G. Simmons (CBS 102605), *A. solani* (Ellis & G. Martin) L.R. Jones & Grout (CBS 116651), *A. macrospora* Zimm., (CBS 117228), *A. nobilis* (Vize) E.G. Simmons (CBS 116490), *Lewia infectoria* (Fuckel) M.E. Barr & E.G. Simmons (CBS 210.86), *Lewia ethzedia* E.G. Simmons (CBS 197.86), *Chalastospora gossypii* (Jacq.) U. Braun & Crous (CPC 15567), *Chalastospora obclavata* Crous & U. Braun (CBS 124120), *Embellisia abundans* E.G. Simmons (CBS 534.83), *A. anigozanthi* R.D. Raabe (CBS 121920), *Lewia eureka* E.G. Simmons (DAOM 195275) and *Crivellia papaveracea* (De Not.) Shoemaker & Inderb (CBS 116607). Furthermore the type strain of *Xenobotryosphaeria*, *X. calamagrostidis* Quaedvl et al., (CBS 303.71) also clustered within the *Alternaria* clade.

The *Pyrenophora* clade comprises five strains with *P. phaeocomes* (Rebent.) Fr., (DAOM 222769), the type species of *Pyrenophora*, *P. dictyoides* A.R. Paul & Parbery (DAOM 63666 and DAOM 75616), *Drechslera dematioidea* (Bubák & Wróbl.) Scharif (CBS 108963) and *P. chaetomioides* Speg., (DAOM 208989). The type species of *Comoclathris*, *C. lanata* Clem, was not available for study, but the five verified strains of *Comoclathris* species form a well-supported clade within the family *Pleosporaceae*. *i. e.* *C. compressa* (Harkn.) Shoemaker & C.E. Babc (CBS 156.53 and CBS 157.53) and the novel species *C. sedi* (MFLUCC 13–0817, MFLUCC 13–0754, MFLUCC 13–0763 and MFLUCC 13–0607) along with a putative strain of *Pleospora ambigua* (Berl. & Bres.) Wehm (CBS 366.52) cluster in a well-supported clade within the *Pleosporaceae* outside *Alternaria s. str.* A putative strain of *Pleospora ambigua* (CBS 366.52) also clustered with our new species *Comoclathris sedi* and is thus treated as an additional strain of *C. sedi*. Furthermore, *Comoclathris incompta* (*Pleospora*

*incompta* (Sacc. & Martelli) Gruyter & Verkley) (CBS 467.76) and *Comoclathris typhicola* (*Pleospora typhicola* (Cooke) Sacc. 1875) (CBS 132.69) also forms two distinct clades within the *Comoclathris* clade.

Another well supported clade (Fig. 1) was formed by *Decorospora*, *D. gaudefroyi* (Pat.) Inderb et al., (pp4723) the type species, along with two putative strains of *Pleospora* sp. ATCC MYA-3203 and ATCC MYA-3202 basal to the *Comoclathris* clade. The *Neocamarosporium* clade comprises seven strains with *Neocamarosporium goegapense* Crous & M.J. Wingf. (CPC 23676<sup>T</sup>), the type species of *Neocamarosporium*: *N. betae* (CBS 109410, IMI 156653, ICMP 10945) and *N. calvescens* (CBS 344.78, CBS 246.79 and CBS 432.77). The type species of *Clathrospora*, *C. elyanae*, forms a well-supported clade, located basal to the *Pleosporaceae* (Fig. 1), outside the *Alternaria* complex.

The genus *Edenia*, with *E. gomezpompae* M.C. González et al., as the type (C1c<sup>T</sup>, CBS 124106, JLCC34533 and 11G048) clusters outside *Pleosporaceae* and formed a distinct clade in *Phaeosphaeriaceae*. During preliminary analysis (data not shown), the type strains of *Austropleospora osteospermi* R.G. Shivas & L. Morin (BRIP 51628) and *Dendryphion europaeum* Crous & R.K. Schumach (CPC 23231 and CPC 22943) clustered outside the suborder *Pleosporineae*, thus are excluded from the final analysis (data not shown).

*Pleospora fallens* (Sacc.) Gruyter & Verkley (CBS 161.78) treated here as *Phoma fallens* Sacc., forms a basal clade in family *Pleosporaceae*. Confusion surrounding this species is discussed in the latter part of this study.

#### Phylogeny of *Alternaria*

In order to define the phylogeny and taxonomy of *Alternaria* and allied genera, 114 strains were included in the *Alternaria* complex alignment. For the *Alternaria* tree, the gene boundaries were: 1–533 bp for ITS, 539–1080 bp for GPDH, 1087–1941 bp for LSU, 1947–2811 bp for RPB2 and 2817–3080 bp for TEF. All phylogenies, different phylogenetic methods and gene regions or gene combinations used on this dataset (data not shown, trees and alignments deposited in TreeBASE), show a weak support at the deeper nodes of the tree. The only well-supported node (Bayesian posterior probability of 1.0, RAxML Maximum Likelihood support value of 99) in all phylogenies separates *Pyrenophora* and the *Pleospora/Stemphylium* clade from the *Alternaria* complex (Fig. 1). The overall topology of the concatenated genes analysis of ITS, GPDH, LSU, RPB2 and TEF was in agreement to the phylogenetic trees obtained from Maximum Likelihood and Bayesian analysis yielded trees with previous work based on Maximum Likelihood and Bayesian analysis of Woudenberg et al. (2013) and Lawrence et al. (2013).

In the *Alternaria* clade, six monotypic lineages and 24 internal clades occur consistently in the individual and



combined phylogenies, although positions vary between the different gene regions or combinations used and are similar with Woudenberg et al. (2013) and Lawrence et al. (2013). The support values for the different phylogenetic methods vary, with the Bayesian posterior probabilities being higher than the RAxML bootstrap support values (Fig. 2). The recently introduced genus, *Xenobotryosphaeria*, *X. calamagrostidis* (CBS 303.71) clustered within the *Alternaria* complex in sect. *Infectoriae*, thus giving rise to confusion (further discussed in the **Taxonomy** section of *Alternaria*).

Seven alignments were analysed corresponding to single gene analyses of ITS, SSU, LSU, GAPDH, TEF1 and RPB2 and combined alignments of the both *Alternaria* and *Pleosporineae*. Comparison of the alignment properties and nucleotide substitution models are provided in Tables 2 and 3.

### Taxonomy

**Pleosporaceae** Nitschke, Verh. naturh. Ver. preuss. Rheinl. 26: 74 (1869).

### **Facesoffungi number: FoF 00500**

*Pathogenic* or *saprobic* on wood and dead herbaceous stems or leaves or pathogen of humans. **Sexual morph:** *Ascomata* perithecial, initially immersed and becoming erumpent to nearly superficial, black, globose, subglobose or ovoid, sometimes hairy or setose, ostiolate. *Ostirole* papillate or apapillate, sometimes with a pore-like ostiole, ostiolar canal filled with or lacking periphyses. *Peridium* relatively thin, usually thick at the sides, thinner at the base. *Hamathecium* of hyaline, septate, cellular pseudoparaphyses interspersed with asci. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical, with or without a pedicel, with an ocular chamber. *Ascospores* uniseriate or biseriate, partially overlapping, phragmosporous or muriform, brown or pale brown, with or without mucilaginous sheath. **Asexual morph:** coelomycetous or hyphomycetous, and the conidiogenous cells can be phialidic, annellidic or sympodial blastoc.

*Type:* ***Pleospora*** Rabenh. ex Ces. & De Not., Comm. Soc. crittog. Ital. 1(4): 217 (1863)

### **Genera accepted in Pleosporaceae**

***Alternaria*** Nees, Syst. Pilze (Würzburg): 72 (1816) [1816–17].  
= *Lewia* M.E. Barr & E.G. Simmons, in Simmons, Mycotaxon 25(1): 289 (1986).  
= *Allewia* E.G. Simmons, Mycotaxon 38: 260 (1990).  
= *Brachycladium* Corda, Icon. fung. (Prague) 2: 14 (1838).

= *Chalastospora* E.G. Simmons, CBS Diversity Ser. (Utrecht) 6: 668 (2007).

= *Chmelia* Svob.-Pol., Biológia, Bratislava 21: 82 (1966).

= *Crivellia* Shoemaker & Inderb., in Inderbitzin, Shoemaker, O'Neill, Turgeon & Berbee, Can. J. Bot. 84(8): 1308 (2006).

= *Embellisia* E.G. Simmons, Mycologia 63(2): 380 (1971).

= *Nimbya* E.G. Simmons, Sydowia 41: 316 (1989).

= *Sinomyces* Yong Wang bis & X.G. Zhang, Fungal Biology 115(2): 192 (2009).

= *Teretispora* E.G. Simmons, CBS Diversity Ser. (Utrecht) 6: 674 (2007).

= *Ulocladium* Preuss, Linnaea 24: 111 (1851).

= *Undifilum* B.M. Pryor, Creamer, Shoemaker, McLain-Romero & Hambl., Botany 87(2): 190 (2009).

= *Ybotromyces* Rulamort, Bull. Soc. bot. Centre-Ouest, Nouv. sér. 17: 192 (1986).

*Type species:* ***Alternaria alternata*** (Fr.) Keissl., Beih. bot. Zbl., Abt. 2 29: 434 (1912).

***Bipolaris*** Shoemaker, Can. J. Bot. 37(5): 882 (1959).

= *Cochliobolus* Drechsler, Phytopathology 24: 973 (1934).

*Type species:* ***Bipolaris maydis*** (Y. Nisik. & C. Miyake) Shoemaker 1959.

***Clathrospora*** Rabenh., Hedwigia 1(18): 116 (1857).

*Type species:* ***Clathrospora elynae*** Rabenh., Hedwigia 1: 116 (1857).

***Comoclathris*** Clem., Gen. fung. (Minneapolis): 37, 173 (1909).

*Type species:* ***Comoclathris lanata*** Clem. [as '*Comochlathris*'], Gen. fung. (Minneapolis): 1–227 (1909).

***Curvularia*** Boedijn, Bull. Jard. bot. Buitenz, 3 Sér. 13(1): 123 (1933).

= *Pseudocochliobolus* Tsuda, Ueyama & Nishih., Mycologia 69(6): 1117 (1978) [1977].

*Type species:* ***Curvularia lunata*** (Wakker) Boedijn, Bull. Jard. bot. Buitenz, 3 Sér. 13(1): 127 (1933).

***Dactuliophora*** C.L. Leakey, Trans. Br. mycol. Soc. 47(3): 341 (1964).

*Type species:* ***Dactuliophora tarrii*** C.L. Leakey, Trans. Br. mycol. Soc. 47(3): 343 (1964).

***Decorospora*** Inderb. et al., in Inderbitzin et al., Mycol. Progr. 1(4): 657 (2002).

*Type species:* ***Decorospora gaudefroyi*** (Pat.) Inderb. et al., in Inderbitzin et al., Mycol. Progr. 94(4): 657 (2002).

≡ *Pleospora gaudefroyi* Pat., Tab. analyt. Fung. (Paris)(7): 40 (no. 602) (1886).



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◀ **Fig. 2** RAxML tree based on a combined dataset of ITS, LSU GAPDH, RPB2 and TEF1 of 114 strains representing the *Alternaria*-complex. Bootstrap support values for maximum likelihood greater than 50 % and Bayesian posterior probabilities greater than 0.90 (green) are indicated below or above the nodes. *Pleospora herbarum* is the out group taxon. The original isolate numbers are noted after the species names. Newly generated strains in this study are indicated in red

**Diademosia** Shoemaker & C.E. Babc., Can. J. Bot. 70(8): 1641 (1992).

*Type species: Diademosia californiana* (M.E. Barr) Shoemaker & C.E. Babc. [as ‘californianum’], Can. J. Bot. 70(8): 1641 (1992).

Basionym: *Graphyllum californianum* M.E. Barr, Mem. N. Y. bot. Gdn 62: 40 (1990).

**Exserohilum** K.J. Leonard & Suggs, Mycologia 66(2): 289 (1974).

= *Setosphaeria* K.J. Leonard & Suggs, Mycologia 66(2): 294 (1974).

= *Luttrellia* Khokhr. & Gornostaĭ, in Gornostaĭ in Azbukina et al. (Eds), Vodorosli, Griby i Mkhi Dal’nego Vostoka [Algae, Fungi and Mosses of the Soviet Far-East] (Vladivostok): 80 (1978).

*Type species: Exserohilum turcicum* (Pass.) K.J. Leonard & Suggs, Mycologia 66(2): 291 (1974).

**Extrawettsteinina** M.E. Barr, Contr. Univ. Mich. Herb. 9(8): 538 (1972).

*Type species: Extrawettsteinina minuta* M.E. Barr, Contr. Univ. Mich. Herb. 9(8): 538 (1972).

**Gibbago** E.G. Simmons, Mycotaxon 27: 108 (1986).

*Type species: Gibbago trianthemae* E.G. Simmons, Mycotaxon 27: 108 (1986).

**Neocamarosporium** Crous & M.J. Wingf., Persoonia, Mol. Phyl. Evol. Fungi 32: 273 (2014).

*Type species: Neocamarosporium goegapense* Crous & M.J. Wingf., Persoonia, Mol. Phyl. Evol. Fungi 32: 273 (2014).

**Paradendryphiella** Woudenberg & Crous, Stud. Mycol. 75(1): 207 (2013).

*Type species: Paradendryphiella salina* (G.K. Sutherl.) Woudenberg & Crous, Stud. Mycol. 75(1): 207 (2013).

Basionym: *Cercospora salina* G.K. Sutherl., New Phytol. 15: 43 (1916).

**Platysporoides** (Wehm.) Shoemaker & C.E. Babc., Can. J. Bot. 70(8): 1648 (1992).

*Type species: Platysporoides chartarum* (Fuckel) Shoemaker & C.E. Babc., Can. J. Bot. 70(8): 1650 (1992).

Basionym: *Pleospora chartarum* Fuckel, Jb. nassau. Ver. Naturk. 23–24: 133 (1870) [1869–70].

**Pleospora** Rabenh. ex Ces. & De Not., Comm. Soc. crittog. Ital. 1(4): 217 (1863).

*Type species: Pleospora herbarum* (Pers.) Rabenh., Klotzschii Herb. Viv. Mycol.: no. 547 (1854).

Basionym: *Sphaeria herbarum* Pers., Syn. meth. fung. (Göttingen) 1: 78 (1801).

**Porocercospora** Amaradasa et al., Mycologia 106(1): 81 (2014).

*Type species: Porocercospora seminalis* (Ellis & Everh.) Amaradasa et al., Mycologia 106(1): 81 (2013).

Basionym: *Cercospora seminalis* Ellis & Everh., J. Mycol. 4(1): 4 (1888).

**Pseudoyuconia** Lar.N. Vassiljeva, Nov. sist. Niz. Rast. 20: 71 (1983).

*Type species: Pseudoyuconia thalictri* (G. Winter) Lar.N. Vassiljeva [as ‘thalicti’], Nov. sist. Niz. Rast. 20: 71 (1983).

Basionym: *Leptosphaeria thalictri* G. Winter, Hedwigia 10: 40 (1872)

**Pyrenophora** Fr., Summa veg. Scand., Section Post. (Stockholm): 397 (1849).

= *Drechslera* S. Ito, Proc. Imp. Acad. Japan 6: 355 (1930).

= *Mariellottia* Shoemaker, Can. J. Bot. 76(9): 1559 (1999) [1998].

*Type species: Pyrenophora phaeocomes* (Rebent.) Fr., Summa veg. Scand., Section Post. (Stockholm): 397 (1849).

Basionym: *Sphaeria phaeocomes* Rebent., Prodr. fl. neomarch. (Berolini): 338 (1804).

#### Excluded genera

**Austropleospora** R.G. Shivas & L. Morin et al., Fungal Diversity 40(1): 70 (2010).

**Table 3** Comparison of alignment properties of genes and nucleotide substitution models used in *Alternaria* complex phylogenetic analysis

Genes /loci	LSU	ITS	GAPDH,	TEF1	RPB2
Alignment strategy (MAFFT v6)	FFT-NS-II	FFT-NS-I+manual	FFT-NS-I+manual	FFT-NS-I+manual	FFT-NS-I+manual
Nucleotide substitution models for Bayesian analysis (determined by MrModeltest)	GTR+I+G	GTR+I+G	GTR+I+G	GTR+I+G	SYM+I+G



*Type species: Austropleospora osteospermi* R.G. Shivas & L. Morin, in Morin, Shivas, Piper & Tan, Fungal Diversity 40(1): 70 (2010).

**Dendryphion** Wallr., Fl. crypt. Germ. (Norimbergae) 2: 300 (1833).

*Type species: Dendryphion comosum* Wallr., Fl. crypt. Germ. (Norimbergae) 2: 300 (1833).

**Edenia** M.C. González et al., in González et al., Mycotaxon 101: 254 (2007).

*Type species: Edenia gomezpompae* M.C. González et al., in González et al., Mycotaxon 101: 254 (2007).

**Kriegeriella** Höhn., Annl. mycol. 16(1/2): 39 (1918).

*Type species: Kriegeriella mirabilis* Höhn., Annl. mycol. 16(1/2): 39 (1918).

**Macrospora** Fuckel, Jb. Nassau. Ver. Naturk. 23–24: 139 (1870) [1869–70].

= *Nimbya* E.G. Simmons, Sydowia 41: 316 (1989).

*Type species: Macrospora scirpicola* (DC.) Fuckel, Jb. Nassau. Ver. Naturk. 23–24: 139 (1870) [1869–70].

Basionym: *Sphaeria scirpicola* DC., in Lamarck & de Candolle, Fl. franç., Edn 3 (Paris) 2: 300 (1805).

**Monascostroma** Höhn., Annl. mycol. 16(1/2): 160 (1918).

*Type species: Monascostroma innumerosum* (Desm.) Höhn. [as ‘innumerosa’], Annl. mycol. 16(1/2): 160 (1918).

**Zeuctomorpha** Sivan et al., Bitunicate Ascomycetes and their Anamorphs (Vaduz): 572 (1984).

*Type species: Zeuctomorpha arecae* Sivan et al., in Sivanesan, Bitunicate Ascomycetes and their Anamorphs (Vaduz): 572 (1984).

Herein we accepted 18 genera in family *Pleosporaceae* based on morphology coupled with molecular data and exclude 7 genera, which have previously been classified under the family (*sensu* Hyde et al. 2013; Wijewardana et al. 2014).

#### Treatment of genera in *Pleosporaceae*

**Alternaria** Nees, Syst. Pilze (Würzburg): 72 (1816) [1816–17]

**Facesoffungi number: FoF 00501**

*Pathogenic or saprobic* on wood and dead herbaceous stems or leaves. **Sexual morph:** *Ascomata* small, solitary to clustered, erumpent to (nearly) superficial at maturity, globose to ovoid, dark brown, smooth, apically papillate, ostiolate. *Ostiole* papilla short, blunt. *Peridium* relatively thin. *Hamathecium* of cellular pseudoparaphyses. *Asci* (4–6) 8-spored, bitunicate, fissitunicate, cylindrical to cylindro-

clavate, straight or somewhat curved, with a short, furcate pedicel and minute ocular chamber. *Ascospores* ellipsoid to fusoid, muriform, slightly constricted at septa, yellow-brown, without guttules, smooth-walled. **Asexual morph:** *Stroma* rarely formed, setae and hyphopodia absent. *Conidiophores* macronematous, mononematous, simple or irregularly and loosely branched, pale brown or brown, solitary or in fascicles. *Conidiogenous cells* integrated, terminal becoming intercalary, polytretic, sympodial, or sometimes monotretic, cicatrized. *Conidia* catenate or solitary, dry, ovoid, obovoid, cylindrical, narrowly ellipsoid or obclavate, beaked or non-beaked, pale or medium olivaceous-brown to brown, smooth or verrucose, with transverse and with or without oblique or longitudinal septa (Woudenberg et al. 2013).

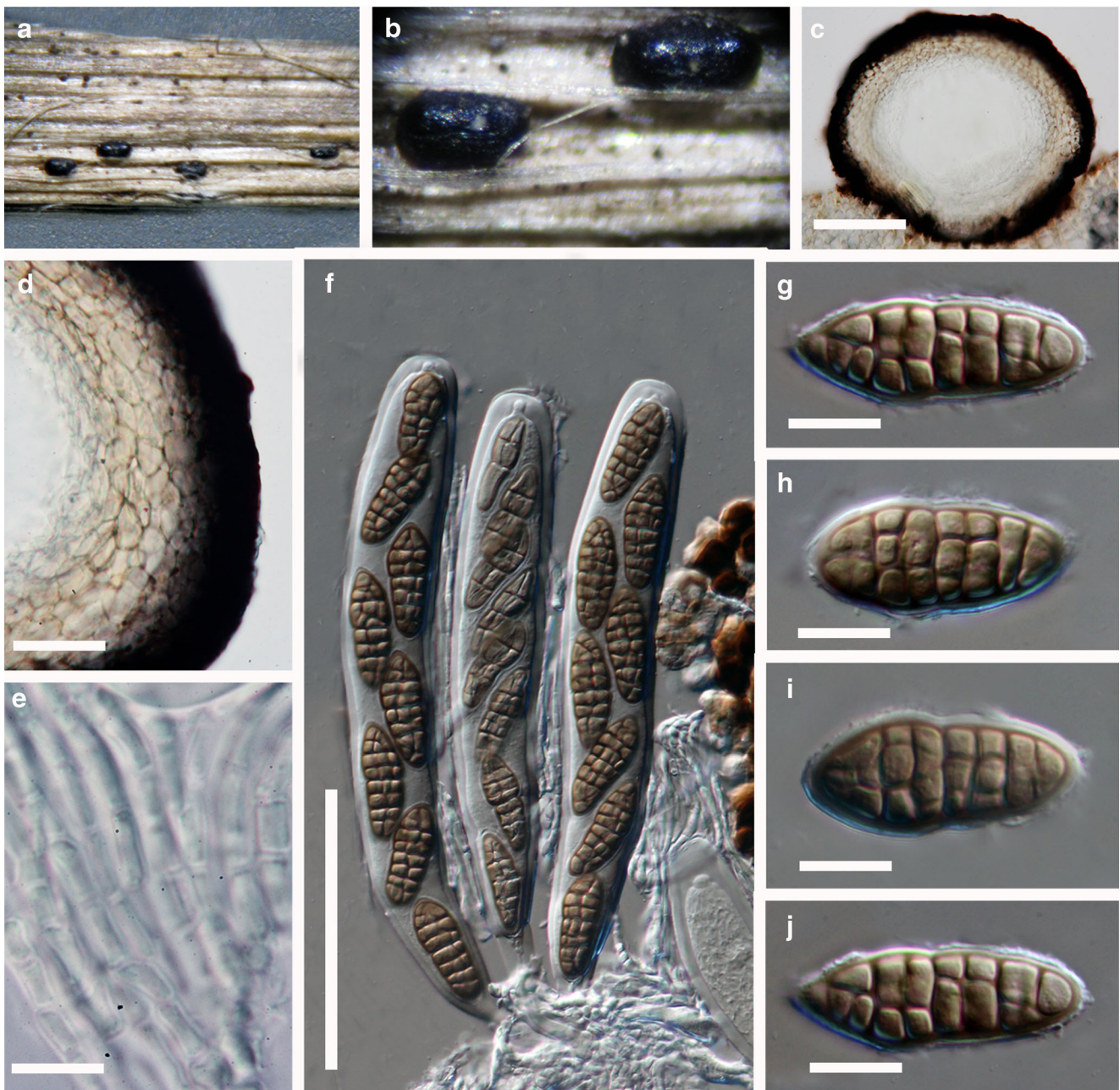
*Type species: Alternaria alternata* (Fr.) Keissl., Beih. bot. Zbl., Abt. 2 29: 434 (1912), Figs. 3 and 4

Basionym: *Torula alternata* Fr., Syst. mycol. (Lundae) 3(2): 500 (1832)

*Pathogen or saprobe* on wood and dead herbaceous stems or leaves. **Sexual morph:** *Ascomata* 170–20×190–240 μm ( $\bar{x}$ =190×440 μm,  $n$ =10), small, solitary to clustered, erumpent to (nearly) superficial at maturity, globose to ovoid, dark brown, smooth, apically papillate, ostiolate. *Ostiole* papilla short, blunt. *Peridium* 38–50 μm ( $\bar{x}$ =45 μm,  $n$ =10) wide, thin, comprising two cell types, outer layer composed of small heavily pigmented, thick-walled cells of *textura angularis*, inner layer composed of lightly pigmented or hyaline, thin-walled cells of *textura angularis*. *Hamathecium* of 2–2.5 μm ( $\bar{x}$ =2.5 μm,  $n$ =10) broad, long, cellular pseudoparaphyses. *Asci* 170–190×23–30 μm ( $\bar{x}$ =180×25 μm,  $n$ =20), (4–6) 8-spored, bitunicate, fissitunicate, cylindrical to cylindro-clavate, straight or somewhat curved, with a short, furcate pedicel and minute ocular chamber. *Ascospores* 37–43×13–14 μm ( $\bar{x}$ =37×13.5 μm,  $n$ =40), ellipsoid to fusoid, muriform, slightly constricted at septa, with 3–7 transverse septa, 1–2 series of longitudinal septa through the two original central segments, end cells without septa, or with 1 longitudinal or oblique septum, or with a Y-shaped pair of septa, brown, without guttules, smooth-walled. **Asexual morph:** *Conidiophores* 40–50×3–6 μm solitary to clustered, simple or branched, straight or flexuous, sometimes geniculate, pale, olivaceous or golden brown, smooth. *Conidia* 20–63×9–18 μm, mostly branched, obclavate, obpyriform, ovoid or ellipsoidal, often with a short conical or cylindrical beak, pale to mid golden brown, up to 8 transverse and usually several longitudinal or oblique septa, smooth or verruculose (Ellis 1971).

**Material examined:** ITALY, Monte Cervarola, Sestola, on the dead stem, 26 March 2012, E. Camporesi IT181-1 MFLU 14-0755, ICMP, living culture (MFLUCC 14-1184). ITALY, Monte Cervarola, Sestola, on the stem of *Achillea* sp. (*Asteraceae*) 15 July 2013, E. Camporesi IT 181-2 MFLU





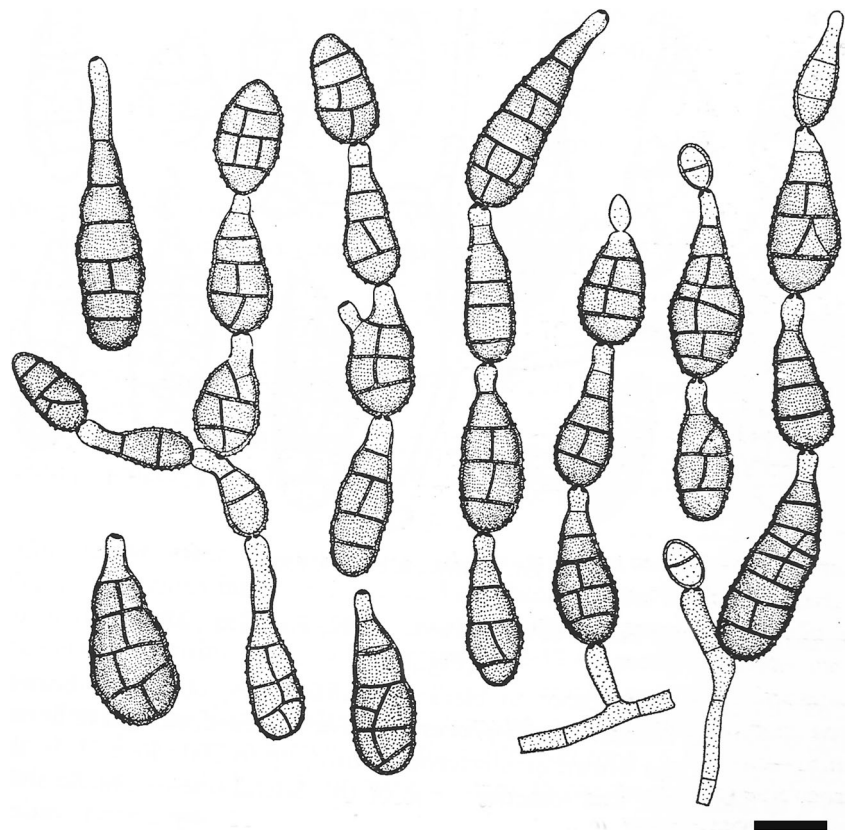
**Fig. 3** *Alternaria alternata* (MFLU 14–0755) **a–b**. Ascomata on host substrate. **c**. Section of ascoma (TS). **d**. Close up of peridium. **e**. Pseudoparaphyses. **f**. Asci with 8-spores. **g–j**. Muriform, brown ascospores. **Scale bar**: **c**=80  $\mu$ m, **d**=30  $\mu$ m, **e–f**=20  $\mu$ m, **g–j**=5  $\mu$ m

14–0756 living culture (MFLUCC 14–1185). ITALY, Monte Cervarola, Sestola, on the stem of *Portulaca* sp. (*Portulacaceae*), 5 September 2012, E. Camporesi IT 466 (MFLU 14–0757); (KIB, PDD).

*Notes:* *Alternaria* was introduced by Nees (1816) and is a ubiquitous genus that includes saprobic, endophytic and pathogenic species associated with a wide variety of substrates (Woudenberg et al. 2013). Recent studies based on DNA data as well as literature reviews revealed multiple paraphyletic genera within the *Alternaria* complex, and *Alternaria* species

clades that do not always correlate to species-groups based on morphological characteristics (Woudenberg et al. 2013). Based on the combined gene analysis of GAPDH, RPB2 and TEF1, Woudenberg et al. (2013) concluded that the *Alternaria* clade contains 24 internal clades and six monotypic lineages, the grouping of which are recognised as *Alternaria* and thus synonymised *Allewia*, *Brachycladium*, *Chalastospora*, *Chmelia*, *Crivellia*, *Embellisia*, *Lewia*, *Nimbya*, *Sinomyces*, *Teretispora*, *Ulocladium*, *Undifilum* and *Ybotromyces* under *Alternaria sensu stricto*. Furthermore, Woudenberg et al. (2013) treated 24 internal clades in the

**Fig. 4** *Alternaria alternata* (Redrawn from *Alternaria alternata* in Ellis (1971), Fig 330.). Scale bars: 650  $\mu\text{m}$



*Alternaria* complex as sections. Our phylogeny based on analysis of ITS, GAPDH, LSU, RPB2 and TEF1 sequence data (Fig. 2) produced similar results as Woudenberg et al. (2013). We therefore follow their recent proposal for the taxonomic treatment of lineages in *Alternaria*.

In the present study we collected the sexual morph of *Alternaria alternata* from Italy in 2013 and 2014. Cultures did not produce the asexual morph but phylogenetically our collections (MFLUCC 14–1185, MFLUCC 14–1184) are 100 % similar with the type strain of *Alternaria alternata* (CBS 916.96). Therefore here we have given the description of the sexual morph of *Alternaria alternata* based on our collections.

We carried out a separate phylogenetic study for the *Alternaria* section to show the placement of some newly introduced species in this study and some sexual reports of *Alternaria* (Fig. 2). During our phylogenetic analysis we observed that the type strain of *Xenobotryosphaeria calamagrostidis* (CBS 303.71) forms a clade within the *Alternaria* section *Infectoriae* and clustered with *Alternaria infectoria*.

The sexual morph of *Xenobotryosphaeria calamagrostidis* is characterised by globose, smooth, superficial ascomata, a peridium with 3–4 layers of brown *textura angularis*, bitunicate, clavate, short pedicellate, 6–8-spored, asci and multiseriate, hyaline, granular, broadly ellipsoid, aseptate

ascospores. This is a clear morphological deviation from the sexual morph of *Alternaria infectoria*. *Xenobotryosphaeria* is typical of genera in the *Botryosphaerales*, but is phylogenetically distinct (Crous et al. 2006; Phillips et al. 2008; Liu et al. 2012). It also resembles species of *Muyocopron* (*Muyocopronaceae*), but the latter genus differs in that it has circular, flattened ascomata, as well as prominent pseudoparaphyses, which are absent in *Xenobotryosphaeria*. Therefore the confusion surrounding *Xenobotryosphaeria* remains unresolved.

During the study we found a novel species of *Alternaria*, *A. murispora* from Germany and this is described below.

*Alternaria murispora* Ariyawansa & K.D. Hyde, *sp. nov.*, Fig. 5

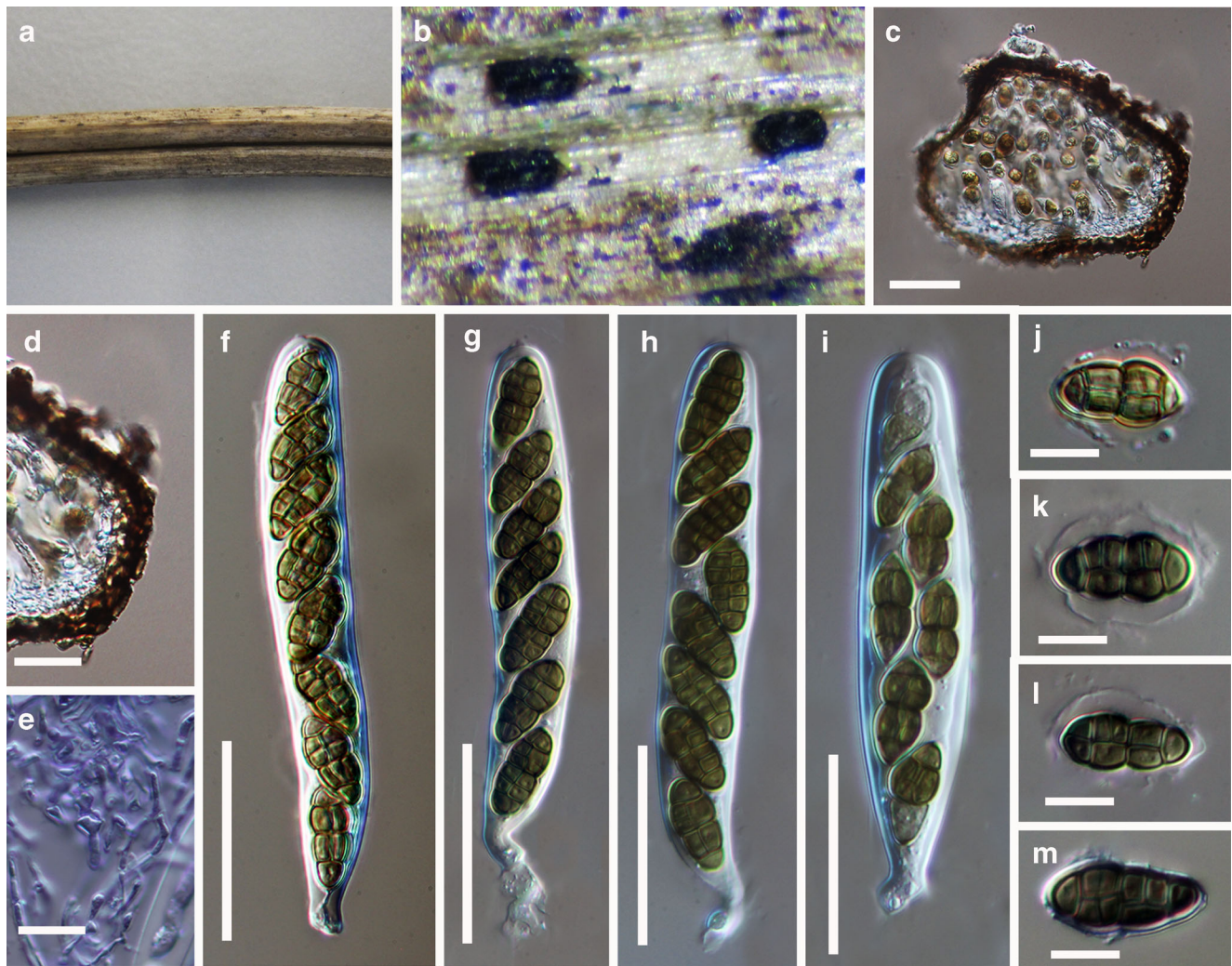
***Index Fungorum number:*** IF 550952, ***Facesoffungi number:*** FoF 00502

***Etymology:*** Named after its muriform ascospores.

***Holotype:*** MFLU 14–0758

***Saprobic on dead stem. Sexual morph:*** *Ascomata* 110–160 × 100–200  $\mu\text{m}$  ( $\bar{x}$  = 130 × 150  $\mu\text{m}$ ,  $n$  = 10), small, scattered, erumpent to (nearly) superficial at maturity, globose or spheroid, dark brown, smooth, apically papillate, ostiolate. *Ostiole* papilla short and blunt. *Peridium* 7–15  $\mu\text{m}$  ( $\bar{x}$  = 12  $\mu\text{m}$ ,  $n$  = 10), thin, comprising two cell types, outer layer composed of small





**Fig. 5** *Alternaria murispora* **a, b.** Ascomata immersed in host substrate. **c.** Section of ascoma (TS). **d.** Close up of peridium. **e.** Cellular pseudoparaphyses. **f–i.** Asci with 8-spores. **j–m.** Brown ascospores. **Scale bars:** c–e=50  $\mu\text{m}$ , f–i=30  $\mu\text{m}$  j–m=10  $\mu\text{m}$

heavily pigmented, thick-walled cells of *textura angularis*, inner layer composed of lightly pigmented or hyaline, thin-walled cells of *textura angularis*. *Hamathecium* of 1–2.5  $\mu\text{m}$  ( $\bar{x}$ =1.8  $\mu\text{m}$ ,  $n$ =20), cellular, septate, pseudoparaphyses branching and anastomosing between and above asci. *Asci* 75–106 $\times$ 11–16  $\mu\text{m}$  ( $\bar{x}$ =92 $\times$ 14  $\mu\text{m}$ ,  $n$ =20), (4–6–) 8-spored, bitunicate, fissitunicate, cylindrical to subcylindrical, straight or somewhat curved, with a short, furcate pedicel and minute ocular chamber. *Ascospores* 16–18 $\times$ 5–7  $\mu\text{m}$  ( $\bar{x}$ =17 $\times$ 6  $\mu\text{m}$ ,  $n$ =40), overlapping seriate, ellipsoid to fusoid, muriform, initially 3-transseptate become 4–5 at maturity, constricted at the central septum, pale brown when immature, dark brown at maturity, without guttules, smooth-walled, with a 1–2.5  $\mu\text{m}$  wide sheath. **Asexual morph:** undetermined.

**Material examined:** GERMANY, Frankfurt, on dead stem, 28 November 2013, A. D Ariyawansa (MFLU 14–0758, **holotype**).

*Notes:* Single spore isolation was not successful and therefore DNA was extracted directly from the fruiting body of the fungus. Therefore no living culture is available.

*Notes:* The novel species *Alternaria murispora* is introduced here based on both morphology and phylogeny. *Alternaria murispora* fits well with the general concept of *Alternaria* in having superficial, globose to ovoid ascomata with short, blunt ostiole, cellular pseudoparaphyses, cylindrical asci with a short, furcate pedicel and ellipsoid to fusoid, brown ascospores which are slightly constricted at the septa. Morphologically *A. murispora* is closely related to *A. conjuncta* E.G. Simmons (= *Lewia scrophulariae* (Desm.) M.E. Barr & E.G. Simmons), but differs in having comparatively smaller asci (75–106  $\mu\text{m}$  versus 120–140  $\mu\text{m}$ ), smaller ascospores (16–18  $\mu\text{m}$  versus 23–25  $\mu\text{m}$ ) and in the number of septa at maturity (3–5  $\mu\text{m}$  versus 6–7  $\mu\text{m}$ ). This is also supported phylogenetically. *i.e.* *A. conjuncta* forms a sister

clade to *A. californica* E.G. Simmons & S.T. Koike, while *A. murispora* forms a sister clade with *A. triticina* Prasada & Prabhu, in section *Infectoriae* (Fig. 2).

***Bipolaris*** Shoemaker, Can. J. Bot. 37(5): 882 (1959)  
= *Cochliobolus* Drechsler, Phytopathology 24: 973 (1934)  
**Facesoffungi number: FoF 00503**

*Pathogenic* or *saprobic* on wood and dead herbaceous stems or leaves. **Sexual morph** *Ascomata* brown or black, immersed, erumpent, partially embedded or superficial, free or on flat stroma, mostly globose to ellipsoidal, sometimes flask-shaped or flattened on hard substrata, smooth or covered with vegetative filaments. *Ostiole* arising centrally, papillate or with a neck. *Peridium* comprising pseudoparenchymatous cells of equal thickness or slightly thickened at apex. *Hamathecium* dense septate, filiform, branched pseudoparaphyses. *Asci* 2–8 spored, bitunicate, fissionate, clavate, cylindrical-clavate or broadly fusoid, straight or slightly curved, thin-walled, often becoming more or less distended prior to dehiscence, short pedicellate, rounded at apex. *Ascospores* fasciculate, filiform or flageliform, hyaline or sometimes pale yellow or pale brown at maturity, septate, helically coiled within ascus, degree of ascospore coiling moderate to very strongly coiled, sometimes with free ends, often with a thin mucilaginous sheath (modified from Manamgoda et al. 2012). **Asexual morph** *Conidiophores* pale to dark brown, single, branched, sometimes arranged in small groups, straight to flexuous or geniculate with smooth or verrucose conidiogenous node. *Conidia* mostly curved, canoe-shaped, fusoid or obclavate, rarely straight, 3–14 pseudoseptate (usually more than 6), hyaline, pale or dark brown, reddish brown or pale to deep olivaceous, germinating by production of one or two germination tubes by polar cells. *Hilum* often slightly protruding or truncate sometimes inconspicuous. *Septum ontogeny* first septum median to sub median, second septum delimits basal cell and third delimits distal cell.

*Type species: Bipolaris maydis* (Y. Nisik. & C. Miyake) Shoemaker, *Canad. J. Bot.* 33: 882 (1959), Fig. 6.

Basionym: *Helminthosporium maydis* Y. Nisik. & C. Miyake, *Journal of Plant Protection, Tokyo* 13: 20 (1926).

= *Drechslera maydis* (Y. Nisik. & C. Miyake) Subram. & B.L. Jain, *Curr. Sci.* 35: 354 (1966)

= *Helminthosporium maydis* Brond., Ill. Iconogr. Microscop. Cryptog. France 15. 1856–1857 (as ‘Helmisporium’), nom. rej. prop. (Rossman, Manamgoda & Hyde, 2013b)

= *Ophiobolus heterostrophus* Drechsler in J. Agric. Res. 31: 701. 1925, nom. rej. prop. (Rossman et al., 2013b)

= *Cochliobolus heterostrophus* (Drechsler) Drechsler, *Phytopathology* 24: 973 (1934).

**Facesoffungi number: FoF 00504**

*Pathogenic* or *saprobic* on wood and dead herbaceous stems or leaves. **Sexual morph of *Bipolaris luttrellii*** on Sach’s agar medium: *Ascomata* 260–370  $\mu\text{m}$  ( $\bar{x}$ =316  $\mu\text{m}$ ,  $n$ =10) diam., superficial or slightly immersed, black, sub globose to ellipsoidal. *Ostiole*, subconical to campanulate, ostiolar beak and upper part of ascomata covered by densely arranged setae. *Pseudoparaphyses* filiform, hyaline, septate. *Asci* 140–205  $\times$  18–26  $\mu\text{m}$  ( $\bar{x}$ =178  $\times$  22  $\mu\text{m}$ ,  $n$ =20), 1–8-spored, bitunicate, fissionate, hyaline, subcylindrical, short pedicellate. *Ascospores* 180–285  $\times$  6–8  $\mu\text{m}$  ( $\bar{x}$ =235  $\times$  7  $\mu\text{m}$ ,  $n$ =20), filiform, hyaline, tapered slightly towards apex and base, tightly coiled inside ascus, sometimes slightly coiled to straight at upper most part, (7–)8(–12)-distoseptate. **Asexual morph of *Bipolaris maydis*** on PDA: *Conidiophores* 105–470  $\times$  5–7  $\mu\text{m}$  ( $\bar{x}$ =286  $\mu\text{m}$ ,  $n$ =20), usually arising singly or in small groups, simple or rarely branched, septate, straight or flexuous, geniculate at upper part, olivaceous brown. *Conidiogenous nodes* dark brown, distinct. Occasionally *secondary sporulation* observed. *Conidia* 66–102  $\times$  14–18  $\mu\text{m}$  ( $\bar{x}$ =94  $\times$  16  $\mu\text{m}$ ,  $n$ =40)  $\mu\text{m}$ , pale to mid dark brown, smooth, slightly curved, fusiform, distoseptate. *Hilum* distinct, 3–5  $\mu\text{m}$  wide, germination tubes arising from both ends of conidia.

**Material examined:** AUSTRALIA, on *Dactyloctenium aegyptium*, 3 June 1985, J.L. Alcorn, (BRIP 14791, **holotype of dried culture of *Cochliobolus luttrellii***) and USA, North Carolina, isolated from *Zea mays* L?, Olin Yoder C5, resulting from six crosses, culture sporulating on *Zea mays* (BPI 892696, **neotype of *Bipolaris maydis***).

*Notes:* *Bipolaris* was introduced by Shoemaker (1959) and it is considered an important plant pathogen associated with over 60 host genera (Sivanesan 1987; Manamgoda et al. 2011; Agrios 2005; Hyde et al. 2014). *Cochliobolus* Drechsler (1934) is the sexual stage of *Bipolaris*. There was no clear morphological boundary between the asexual genera *Bipolaris* and *Curvularia*, and some species show intermediate morphology which thus caused confusion for many plant pathologists and mycologists for their correct identification (Sivanesan 1987; Manamgoda et al. 2011; Hyde et al. 2014). Based on combined gene analysis of rDNA ITS (internal transcribed spacer), LSU, GPDH and EF1- $\alpha$ , Manamgoda et al. (2012) resolved the taxonomic confusion in *Bipolaris* and *Curvularia* complex. Multilocus phylogeny showed that *Bipolaris* and *Curvularia* form two well supported clades in previous studies (Manamgoda et al. 2011, 2012) as well as in the present study. Further, the nomenclatural conflict in this complex was resolved giving priority to the more commonly used established generic names *Bipolaris* and *Curvularia* thus *Cochliobolus* was synonymised under *Bipolaris*. Recently Manamgoda et al. (2014) revised the genus *Bipolaris* based





**Fig. 6** **a-h:** *Bipolaris luttrellii* (holotype of dried culture of *Cochliobolus luttrellii*, **j-n**), *Bipolaris maydis* (neotype of *Bipolaris maydis*, **a-h**) **a.** Ascromata on host substrate. **b.** Section of the ascomata. **c.** Close up of

the peridium **d-e.** Subcylindrical asci with a short pedicle note: helical arrangement of the ascospores **j.** Conidiophore. **k-n.** Conidia. Scale bars:  $b=100\ \mu\text{m}$ ,  $c=10\ \mu\text{m}$ ,  $d-g=60\ \mu\text{m}$ ,  $h-j=30\ \mu\text{m}$

on DNA sequence data derived from living cultures of fresh isolates, available ex-type cultures from worldwide collections and observation of type and additional specimens. They accepted 47 species in the genus *Bipolaris* and clarify the taxonomy, host associations, geographic distributions and

species' synonymies while epi- or neotypes were designated for *Bipolaris cynodontis* (Marignoni) Shoemaker, *B. oryzae* (Breda de Haan) Shoemaker, *B. victoriae* (F. Meehan & H.C. Murphy) Shoemaker, *B. yamadae* (Y. Nisik.) Shoemaker and *B. zeicola* (G.L. Stout) Shoemaker.



In our phylogeny, *Bipolaris* forms a robust clade sister to *Curvularia* and *Porocercospora*. Therefore we accept *Bipolaris* as a well established genus in *Pleosporaceae* based on both morphology and phylogeny.

***Clathrospora*** Rabenh., Hedwigia 1(18): 116 (1857)

**Facesoffungi number: FoF 00505**

*Saprobic* on wood and stems. **Sexual morph:** *Ascomata* semi-immersed, scattered on putrid host stems and foliage, brown to blackish brown, subglobose or nearly globose, with a central sunken ostiole open via a circular lid, asci and pseudoparaphyses forming at the base of the peridium. *Peridium* composed of 3–5 layers of brown, relatively thick-walled cells of *textura angularis*, inner cells flattened, thin-walled and lighter. *Hamathecium* composed of dense, hyaline, filiform, pseudoparaphyses which are longer than the asci. *Asci* 8-spored, bitunicate, fissitunicate, thick-walled, cylindrical to clavate, with a short pedicle and shallow ocular chamber. *Ascospores* biseriolate, fusiform, muriform, 7-transseptate, two or many rows of longitudinal septa, applanate, constricted only at the central septum, dark brown to brown, surrounded by a thin, hyaline mucilaginous sheath. **Asexual morph:** *Alternaria* like (Zhang et al. 2012).

**Type species:** *Clathrospora elyanae* Rabenh., Hedwigia 1: 116 (1857), Fig. 7.

≡ *Clathrospora elyanae* Rabenh. (1857).

≡ *Pleospora elyanae* (Rabenh.) Ces. & De Not., Commentario della Società Crittogamologica Italiana 1 (4): 218 (1863).

**Facesoffungi number: FoF 00506**

*Saprobic* on wood and stems. **Sexual morph:** *Ascomata* 220–140 × 145–175 μm ( $\bar{x}$  = 170 × 150 μm,  $n$  = 10), semi-immersed, scattered on the putrid host stems and foliage, subglobose or nearly globose, brown to blackish brown, with a central sunken ostiole open via a circular lid. *Peridium* 20–55 μm ( $\bar{x}$  = 38,  $n$  = 20), composed of 3–5 layers of brown, relatively thick-walled cells of *textura angularis*, inner cells flattened, thin-walled and lighter. *Hamathecium* composed of dense, 2–3 μm diam ( $\bar{x}$  = 2,  $n$  = 20), hyaline, filiform, pseudoparaphyses, longer than the asci. *Asci* 160–230 × 24–48 μm ( $\bar{x}$  = 190 × 35 μm,  $n$  = 20), 8-spored, bitunicate, fissitunicate, thick-walled, cylindrical to clavate, with a short pedicle and minute ocular chamber. *Ascospores* 40–65 × 18–27 μm ( $\bar{x}$  = 53 × 23 μm,  $n$  = 40), biseriolate, fusiform, muriform, 7-transseptate, two or many rows of longitudinal septa, constricted only at the central septum, applanate, dark brown to brown, surrounded by a thin, hyaline mucilaginous sheath. **Asexual morph:** *Alternaria*-like (Zhang et al. 2012).

**Material examined:** SWITZERLAND, on the stem of *Carex curvula*, September 1898, Winter (BPI 627748, **isotype**).

**Notes:** Shoemaker and Babcock (1992) assigned *Clathrospora* to *Diademaceae* and included an additional nine species and provided a key to the genus based on the number of septa and length of ascospores. *Clathrospora* was characterized by ascomata circular lid-like opening and applanate, muriform ascospores. Currently, 50 *Clathrospora* species are listed in the genus in Index Fungorum (2015). Molecular studies based on combine gene analysis showed that two putative strains of *Clathrospora*, *C. elyanae* (CBS 196.54) and *C. diplospora* (IMI 68086) clustered in *Pleosporaceae* (Ariyawansa et al. 2014a). We obtained similar results in the phylogenetic tree produced from combined ITS, nrLSU, nrSSU and RPB2 sequence analysis (Fig. 1). *Clathrospora elyanae*, the type of *Clathrospora*, formed a separate clade with low bootstrap support (58 %) within *Pleosporaceae*. Based on the phylogenetic result together with the morphological characters (slightly papillate ostiole with applanate, muriform ascospores and *Alternaria*-like asexual morph) we refer *Clathrospora* to *Pleosporaceae*.

***Comoclathris*** Clem., Gen. fung. (Minneapolis): 37, 173 (1909)

= *Platyspora* Wehm., World Monograph of the Genus *Pleospora* and its Segregates: 254 (1961).

**Facesoffungi number: FoF 00507**

*Saprobic* on dead wood or stems. **Sexual morph:** *Ascomata* semi-immersed to superficial, scattered or aggregated, subglobose or nearly globose, brown to blackish brown coriaceous, ascomata opening via a large circular aperture or lid. *Peridium* comprising 3–4 layers of brown, relatively thick-walled cells of *textura angularis*. *Hamathecium* composed of dense, hyaline, filiform, septate pseudoparaphyses. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical to cylindro-clavate with an ocular chamber. *Ascospores* uniseriate or partially overlapping, fusiform, muriform, applanate, brown to reddish-brown, surrounded by a thick, hyaline, mucilaginous sheath. **Asexual morph:** *Alternaria*-like (Zhang et al. 2012).

**Type species:** *Comoclathris lanata* Clem. [as ‘*Comochlathris*’], Gen. fung. (Minneapolis): 1–227 (1909), Fig. 8.

**Facesoffungi number: FoF 00508**

*Saprobic* on dead stem. **Sexual morph:** *Ascomata* 175–245 × 142–197 μm ( $\bar{x}$  = 205 × 159 μm,  $n$  = 20), scattered or aggregated on the host stem, subglobose or nearly globose, superficial, coriaceous, covered with a pale membrane, brown to blackish brown, with a central ostiole. *Peridium* 10–22 μm wide, comprising 3–4 layers of brown, relatively thick-walled cells of *textura angularis*, inner cells flattened, thin-walled and lighter.

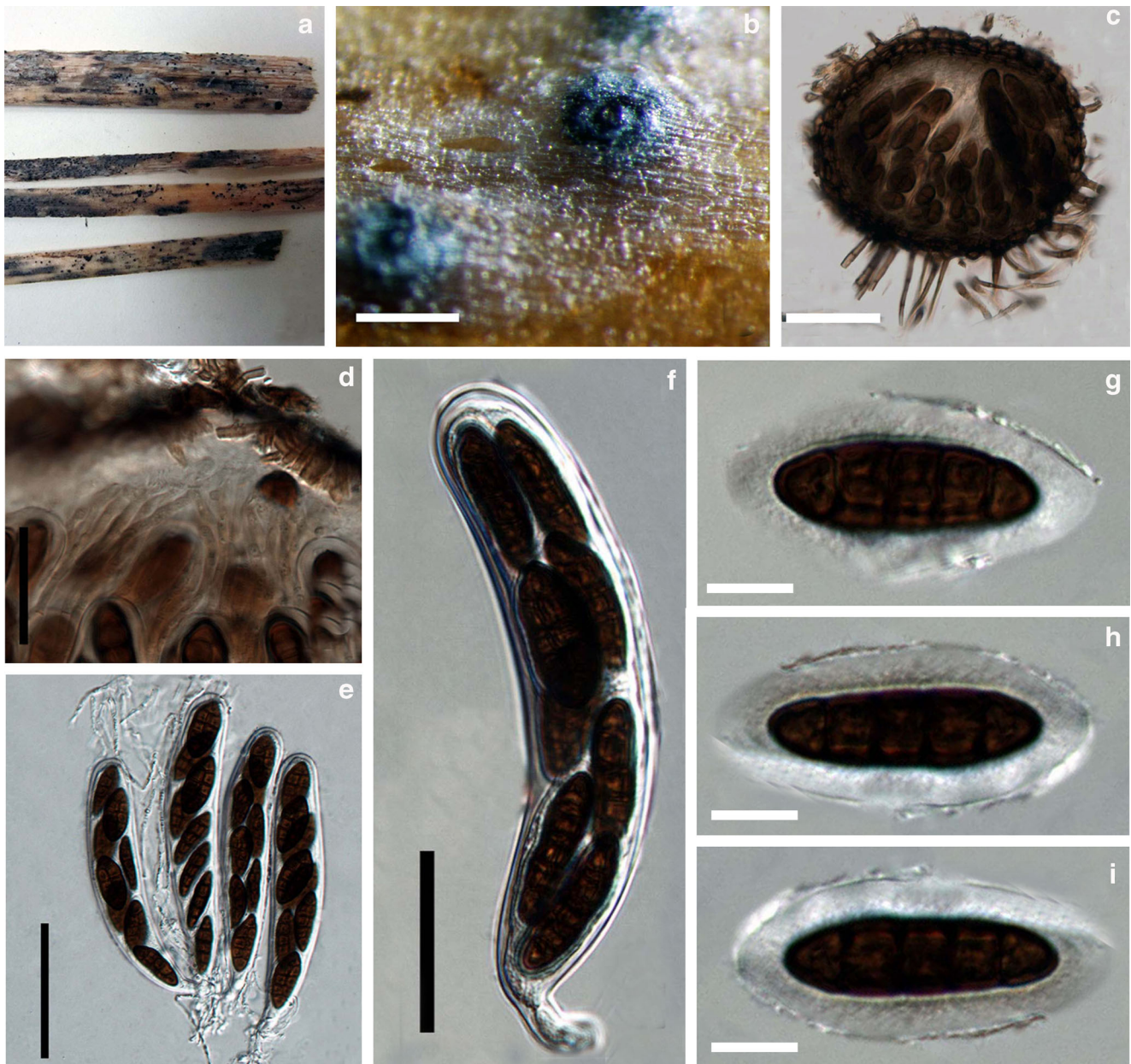




**Fig. 7** *Clathrospora elyanae* (isotype) **a.** Herbarium material. **b.** Close up of ascomata. **c.** Section of the ascoma **d.** Close up of the peridium. **e.** Hyaline, filiform, pseudoparaphyses. **f–h.** Cylindrical to clavate asci with

a short pedicle and ocular chamber. **i–k.** Dark brown to brown muriform ascospores surrounded by a thin, hyaline mucilaginous sheath. **Scale bars:** b=100  $\mu$ m, c=10  $\mu$ m, d–g=60  $\mu$ m, h–j=30  $\mu$ m





**Fig. 8** *Comoclathris lanata* (holotype). **a.** Herbarium material showing habit of fungus on host stem. **b.** Erumpent ascomata. **c.** Section through ascoma. Note the arrangement of asci and external setae. **d.** Section showing peridial cells of ascoma. **e.** Hyaline, filiform,

pseudoparaphyses. **f.** Asci with short knob-like pedicels and shallow ocular chamber. **g–i.** Muriform, applanate ascospores with a thick sheath. **Scale Bars:** b =200  $\mu\text{m}$ , c=40  $\mu\text{m}$ , d–e=10  $\mu\text{m}$ , f=20  $\mu\text{m}$ , g–i=10  $\mu\text{m}$

*Hamathecium* composed of dense 2–4  $\mu\text{m}$  wide, septate, hyaline, filiform, pseudoparaphyses longer than the asci. *Asci* 108–149  $\times$  20–30  $\mu\text{m}$  ( $\bar{x}$ =24  $\times$  125  $\mu\text{m}$ ,  $n$ =20), 8-spored, bitunicate, fissitunicate, thick-walled, cylindrical to cylindro-clavate, with a short knob-like pedicel, and indistinct shallow ocular chamber. *Ascospores* 20–32  $\times$  8–13  $\mu\text{m}$  ( $\bar{x}$ =12  $\times$  28  $\mu\text{m}$ ,  $n$ =20), 1–2 overlapping seriate, fusiform, muriform, with 4–5-transverse septa and 1–2-longitudinal septa, not constricted at the septa, applanate, brown to reddish-brown, surrounded by a distinct, hyaline, mucilaginous 3–8  $\mu\text{m}$  wide sheath. **Asexual morph:** undetermined.

**Material examined:** USA, Colorado, on stem of *Leptotaenia multifida* Nutt (*Umbelliferae*), 8 July 1907, F.E. & E.S. Clements (COLO 62872, holotype).

*Notes:* *Comoclathris*, typified by *Comoclathris lanata*, was introduced by Clements (1909). The genus is characterized by ascomata with circular lid-like openings and applanate reddish-brown to dark reddish-brown, muriform ascospores, with single longitudinal septa (Ariyawansa et al. 2014a). Zhang et al. (2012) tentatively placed *Comoclathris* in the *Pleosporaceae* based on Alternaria-like asexual morphs and



this was followed by Woudenberg et al. (2013). *Comoclathris* shares common characters with *Pleospora herbarum*, the type of *Pleospora*, in having cylindrical to cylindro-clavate asci with an ocular chamber and muriform, brown or pale brown ascospores, with or without sheath. *Comoclathris* and *Pleospora* differ in the opening of ascomata (opening via a large circular aperture or lid versus opening by a central pore and applanate ascospores). *Comoclathris* and *Pleoseptum* share similar characters in having globose, black, ascomata and cylindrical to cylindro-clavate asci with muriform, yellowish to dark brown ascospores. *Comoclathris* differs from *Pleoseptum* in having superficial ascomata with circular lid-like openings composed of comparatively thin peridium and applanate and fusiform ascospores surrounded by a distinct hyaline, mucilaginous thick sheath (Ariyawansa et al. 2014a). In *Pleoseptum* ascomata are immersed, usually with a papillate apex, with a relatively broad peridium and ovoid to fusoid ascospores (Ariyawansa et al. 2014a). *Comoclathris* was considered to differ from *Clathrospora* as in the latter genus species have two or more rows of longitudinal septa as compared with a single row in *Comoclathris* (Ariyawansa et al. 2014a). Shoemaker and Babcock (1992) provided a key to 21 species of *Comoclathris*. Presently 32 epithets are listed for *Comoclathris* in Index Fungorum (2015).

Molecular data for *Comoclathris lanata*, the type species of *Comoclathris*, is not available. Two putative strains of *Comoclathris compressa* (CBS 157.53 and CBS 156.53) however, cluster together in a well-supported clade within the family *Pleosporaceae* (Ariyawansa et al. 2014a). Based on the phylogenetic result, coupled with the morphological characters (*Alternaria*-like asexual morph), we agree with Ariyawansa et al. (2014a), Zhang et al. (2012) and Woudenberg et al. (2013) in placing *Comoclathris* in *Pleosporaceae*, but re-collection of the type species and epitypification or a reference specimen (Ariyawansa et al. 2014a) with molecular data is essential to establish the correct placement of the genus.

During this study we have proposed several new combinations in order to resolve the paraphyletic nature of *Pleospora* and to make a stable taxonomy for the family *Pleosporaceae*. *Pleospora incompta* (Sacc. & Martelli) Gruyter & Verkley (CBS 467.76) was introduced by de Gruyter et al. (2013) in order to accommodate *Phoma incompta* Sacc. & Martelli. In the same study *Pleospora typhicola* Cook was proposed by de Gruyter et al. (2013) to accommodate *Phoma typhina* (Sacc. & Malbr.) van der Aa & Vanev in the family *Pleosporaceae* but during our study *Pleospora typhicola* (Cooke) Sacc. 1875 (CBS 132.69) and *Pleospora incompta* (CBS 467.76) form separate clades within the genus *Comoclathris* (Fig. 1). Therefore we provide two new combinations namely *Comoclathris incompta* and *C. typhicola* to accommodate *Pleospora incompta* and *P. typhicola* in *Comoclathris*.

***Comoclathris incompta*** (Sacc. & Martelli) Ariyawansa & K. D. Hyde, **comb. nov.**,

**Fungorum number: IF 550953**

Basionym: *Phoma incompta* Sacc. & Martelli, Syll. Fung. 10: 146.1892.

≡ *Pleospora incompta* (Sacc & Martelli) Gruyter & Verkley, in Gruyter et al., Stud. Mycol. 75: 25 (2012).

***Comoclathris typhicola*** (Cooke) Ariyawansa & K. D. Hyde, **comb. nov.**,

**Fungorum number: IF 550954**

Basionym: *Sphaeria typhicola* Cooke, Grevillea 5: 121. 1877.

≡ *Pleospora typhicola* (Cooke) Sacc., Reliq. Libert 2: no. 152 (1875)

≡ *Clathrospora typhicola* (Cooke) Höhn., Ann. Mycol. 16: 88. 1918.

≡ *Pyrenophora typhicola* (Cooke) E. Müll., Sydowia 5: 256. 1951.

≡ *Macrospora typhicola* (Cooke) Shoemaker & C.E. Babc., Canad. J. Bot.

70: 1644. 1992.

≡ *Phyllosticta typhina* Sacc. & Malbr., Sacc., Michelia 2: 88. 1880.

≡ *Phoma typhina* (Sacc. & Malbr.) van der Aa & Vanev, A revision of the species described in *Phyllosticta*: 468. 2002.

≡ *Phoma typharum* Sacc., Syll. Fung. 3: 163. 1884.

We also collected a novel species of *Comoclathris*, *C. sedi* from Italy and this is described below.

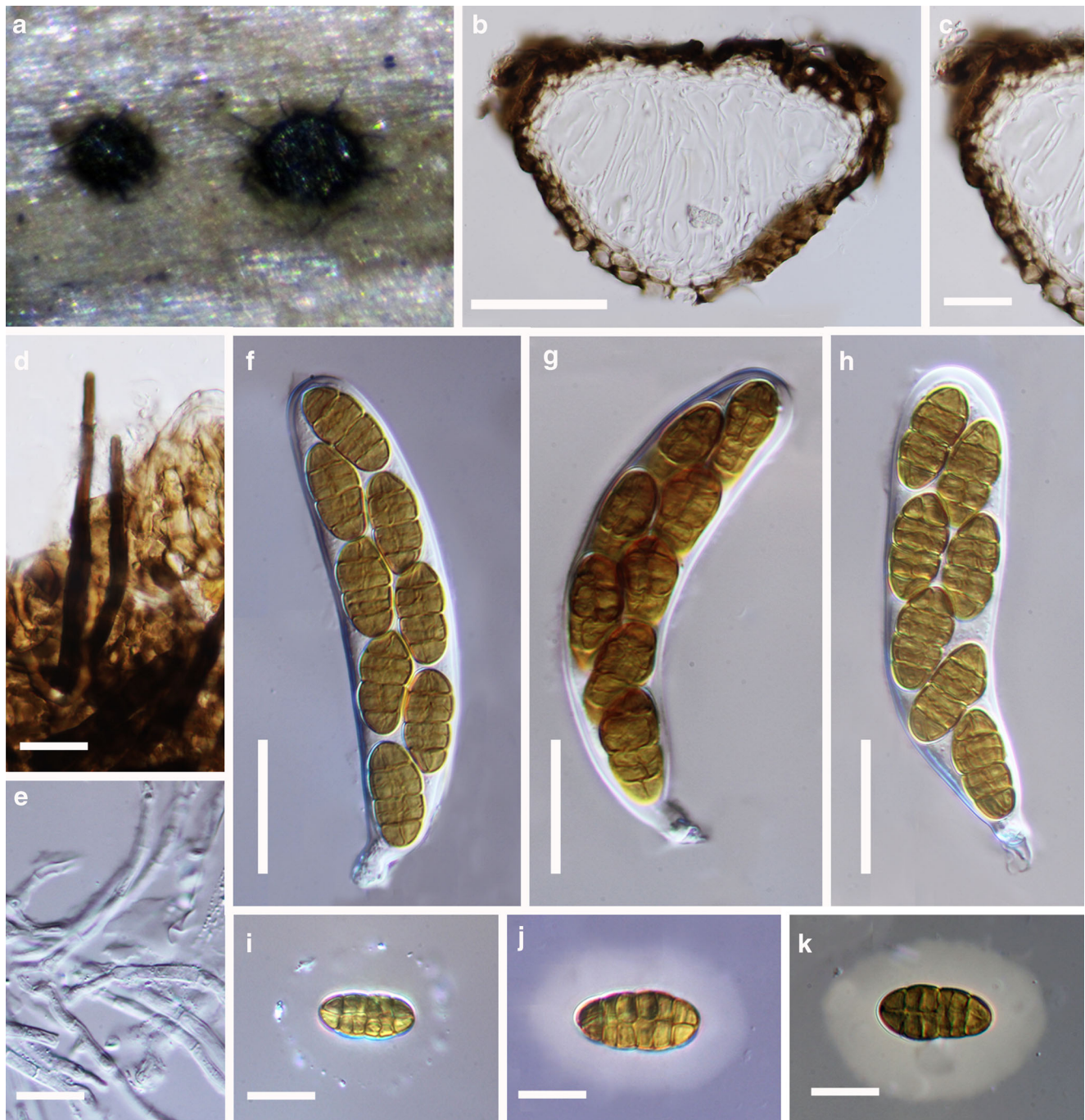
**Type species: *Comoclathris sedi*** Wanasinghe, Ariyawansa, E. Camporesi & K.D. Hyde, **sp. nov.**, Fig. 9.

**Index Fungorum number: IF 550955, Facesoffungi number: FoF 00509**

**Etymology:** The specific epithet *sedi* is based on the host genus from which the fungus was isolated.

**Holotype:** MFLU 14 0758

*Saprobic* on dead stem. **Sexual morph:** *Ascomata* 200–250 × 290–350 μm ( $\bar{x}$ =230 × 320 μm,  $n$ =10), scattered or aggregated on the host stem, subglobose or nearly globose, superficial, coriaceous, brown to blackish brown with a blunt ostiole. *Peridium* 20–38 μm ( $\bar{x}$ =25 μm,  $n$ =10) wide, comprising 3–4 layers of brown, relatively thick-walled cells of *textura angularis*, inner cells flattened, thin-walled and lighter. *Hamathecium* composed of dense 1.5–2.5 μm ( $\bar{x}$ =2 μm,  $n$ =10) wide, septate, hyaline, filiform, pseudoparaphyses longer than the asci. *Asci* 80–110 × 16–18 μm ( $\bar{x}$ =98 × 18 μm,  $n$ =20), 8-spored, bitunicate, fissionate, cylindrical to cylindro-clavate, with a short knob-like pedicel, and indistinct shallow ocular chamber. *Ascospores* 19–20 × 8–10 μm ( $\bar{x}$ =20 × 19 μm,  $n$ =40), 1–2 overlapping seriate, fusiform, muriform,



**Fig. 9** *Comoclathris sedi* (holotype). **a.** Habit of the fungus on host stem. **b.** Superficial ascomata. **c.** Thick section through ascoma. **d.** Dark brown setae. **e.** Hyaline pseudoparaphyses. **f-h.** Asci with shallow

curved, muriform ascospores with thick sheath. **Scale Bars:** b=200  $\mu$ m, c=40  $\mu$ m, f-h=10  $\mu$ m, i-k=20  $\mu$ m

with 4–5-transverse septa and 1–2-longitudinal septa, not constricted at the septa, brown to reddish brown, surrounded by a distinct, hyaline, mucilaginous 5–9  $\mu$ m wide sheath. **Asexual morph:** undetermined.

**Material examined:** ITALY, Stavel, Ortignano-Raggiolo, on stem of *Sedum* sp. (*Crassulaceae*), 6 June 2012, E. Camporesi IT 416, (MFLU 14 0759, **holotype**) – ex-type living culture

(MFLUCC 13 0817, BRIP); ITALY, Monte Cervarola, Sestola, on dead branch of *Clematis vitalba* (*Ranunculaceae*), 21 July 2012, E. Camporesi IT 589, (KIB, **paratype**) – ex-type living culture (ICMP); ITALY, Almazago, on dead stem of *Rosa* sp. (*Rosaceae*), 8 August 2013, N. Camporesi IT 1408 (MFLU14-0760) – living culture (MFLUCC 13 0763).

**Notes:** The novel taxon, *Comoclathris sedi* was initially isolated on dead stem of *Sedum* sp. But later the same species



was isolated from *Clematis vitalba*, *Rosa* sp. and *Digitalis* sp. and they are morphologically and phylogenetically identical. *Comoclathris sedi* shows similarities with *Comoclathris*, in having globose, black, ascumata with setae and cylindrical to cylindro-clavate asci having knob-like pedicel with muriform, yellowish to dark brown appanate ascospores. Our new species differs from other species of *Comoclathris* in having superficial, 200–250×290–350 µm ascumata ( ) with distinct ostioles, , a peridium comprising a single layer of brown, relatively thick-walled cells of *textura angularis*, and fusiform to ellipsoidal, muriform, brown to reddish brown ascospores with 2–5-transverse septa and 1–4-longitudinal septa. The phylogenetic analysis of combined ITS, LSU, SSU nrDNA and RPB2 sequence data provides strong evidence that *Comoclathris sedi* belongs in *Pleosporaceae* and forms a robust clade within the genus *Comoclathris* sister to *Comoclathris compressa* (CBS 157.53 and CBS 156.53) with relatively high bootstrap support (Fig. 1); thus a new species is proposed.

***Curvularia*** Boedijn, Bull. Jard. bot. Buitenz, 3 Sér. 13(1): 123 (1933)

= *Pseudocochliobolus* Tsuda, Ueyama & Nishih., Mycologia 69(6): 1117 (1978) [1977]

= *Curvusporium* Corbetta [as ‘Curvosporium’], Riso 12(3): 28, 30 (1963)

= *Malustela* Bat. & J.A. Lima, Publicações Inst. Micol. Recife 263: 5 (1960)

**Facesoffungi number: FoF 00510**

*Pathogenic* or saprobic on wood and dead herbaceous stems or leaves or humans. **Sexual morph:** *Ascomata* superficial, globose to ellipsoidal, dark brown to black, free or frequently developing from columnar stromata or flat stromata, with a well defined ostiolar neck. *Peridium* coriaceous, carbonaceous, pseudoparenchymatous. *Hamathecium* comprising filiform, septate and sometimes branched pseudoparaphyses. *Asci* 1–8-spored, bitunicate, fissitunicate, cylindrical to cylindrical clavate, pedicel short and minute ocular chamber. *Ascospores* fasciculate, usually parallel, loosely coiled or highly coiled at the extremities of the ascus, filamentous, filiform to flageliform and somewhat tapered at the extremities, 3–20 septate, hyaline or somewhat pigmented at maturity. **Asexual morph:** *Conidiophores* branched or unbranched, straight to flexuous, septate, smooth to verruculose, often geniculate sometimes nodulose. *Conidiogenous cells* polytretic, integrated, sometime when mature becoming intercalary, sympodial, cylindrical with smooth to verrucose conidiogenous nodes. *Conidia* straight oblong, ellipsoidal, clavate, fusiform, subcylindrical or lunate, rounded at the ends or sometimes tapering slightly towards the base, pale brown, medium reddish brown to dark brown, 3–10 distoseptate (usually 3–5), conidial wall smooth to verrucose. *Hilum* protuberant in some

species. *Stromata* formed in some species (obtained from Manamgoda et al. 2012).

*Type species: Curvularia lunata* (Wakker) Boedijn, Bull. Jard. bot. Buitenz, 3 Sér. 13(1): 127 (1933), Fig. 10.

= *Acrothecium lunatum* Wakker, De ziekten van het suikerriet op Java, die niet door dieren veroorzaakt worden: 196 (1898).

= *Helminthosporium curvulum* Sacc., Atti della Accademia Scientifica Veneto-Trentino-Istria 10: 89 (1916).

= *Helmisporium curvulum* Sacc. (1916).

**Facesoffungi number: FoF 00511**

*Pathogenic* or saprobic on wood and dead herbaceous stems or leaves. **Sexual morph** not observed in culture. **Asexual morph:** *Conidiophores* 39–430×4–9 µm, branched or unbranched, straight to flexuous, septate, smooth to verruculose, often geniculate sometimes nodulose. *Conidiogenous cells* 4–20×3–13 µm, polytretic, integrated, sometime when mature becoming intercalary, sympodial, cylindrical, cicatrized or sometimes swollen. *Conidiogenous nodes* smooth to verrucose. *Conidia* 21–31×9–13 µm, straight oblong, ellipsoidal, clavate, fusiform, sub-cylindrical or lunate, rounded at the ends or sometimes tapering slightly towards the base, pale brown, medium reddish brown to dark brown, 3–10 distoseptate (usually 3–5), conidial wall smooth to verrucose.

**Material examined:** THAILAND, Chiang Rai, on stem of *Zea mays*, 8 July 2012, D.S Manamgoda (MFLU 10–0555) – ex-type living culture (MFLUCC 12–0181).

*Notes:* *Curvularia* was introduced by Boedijn (1933) and it is an important pathogen in humans and plants (Sivanesan 1987; Agrios 2005, Manamgoda et al. 2012, 2014). *Bipolaris* and *Curvularia* share many morphological similarities (Sivanesan 1987). There are some *Bipolaris* species with short, straight conidia showing intermediate conidial characters between these two genera (Manamgoda et al. 2012). Confusion surrounding *Bipolaris* and *Curvularia* was resolved by Manamgoda et al. (2012) based on combined genes of ITS, GPDH, LSU, EF1- $\alpha$  sequence data thus Manamgoda et al. (2012) revealed that the traditionally circumscribed *Bipolaris* and *Curvularia* cannot be combined into a single monophyletic genus (Goh et al. 1998; Shimizu et al. 1998; Berbee et al. 1999). A similar phylogeny was found in the present study, where *Bipolaris* and *Curvularia* form two well-supported clades in the family *Pleosporaceae*, thus we accept *Curvularia* as a separate genus in *Pleosporaceae*.

***Dactuliophora*** C.L. Leakey, Trans. Br. mycol. Soc. 47(3): 341 (1964).

**Facesoffungi number: FoF 00512**





**Fig. 10** *Curvularia lunata* (MFLU 10-0555). **a, b.** Fungus on host stem. **c, d.** Conidiophores. **e-i.** Conidia. **j.** Atypical bifurcate conidium. **Scale Bars:** c-j = 10  $\mu$ m

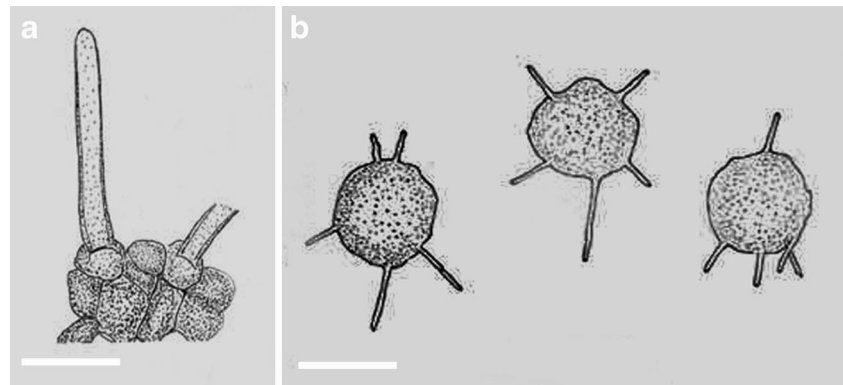
*Parasitic* on leaves. *Mycelium* generally immersed, hyaline and diffuse in the leaf tissues, aggregated irregularly in the epidermal or deeper leaf tissues into plectenchymic masses from which sclerotiphores and sclerotia develop, 'sclerotiphore' remains after the disjunction of the sclerotium as a superficial or occasionally more or less immersed structure, being the external continuation of the immersed aggregation. *Hyphae* at the circumference are dematiaceous and relatively large while those in the centre of the ring so formed are generally paler and relatively smaller. Sclerotia maybe glabrous, hispidulous, hispid or sparsely setose, spherical to

subspherical, ellipsoidal, pyriform or rostrate, wholly or partly composed of dematiaceous cells on the outside, hyaline and undifferentiated, separates from sclerotiphore by the fracture of many thin-walled cells joining the base of the mature sclerotium to the centre of the sclerotiphore. **Note:** No other structures have been observed other than sclerotia (Leakey 1964).

*Type species:* *Dactuliophora tarrii* C.L. Leakey, Trans. Br. mycol. Soc. 47(3): 343 (1964), Fig. 11.

*Facesoffungi number:* FoF 00513

**Fig. 11** *Dactuliophora tarrii*  
(Redrawn from the holotype in  
Leakey 1964, Figs. 1 and 2).  
Scale bars: = 50  $\mu\text{m}$



*Parasitic* on leaves. *Leaf spots* broadly zonate on upper leaf surface, rosette-like altering white and brown bands, lower surface densely covered with sclerotiophores and sclerotia, zonation less distinct. *Mycelium* immersed, hyaline, generally diffused throughout the tissues in the leaf spot and aggregated in plectenchymic masses beneath the sclerotiophores. *Sclerotiophores* consist of a fully erumpent ring of dematiaceous hyphae continuous with a subepidermal plectenchymic mycelial mass. *Sclerotia* 35–135  $\mu\text{m}$  diam., scattered, specially on the paler parts of the leaf spot, generally hypophyllous but occasionally amphigenous, spherical to subspherical, bearing 0–10 (usually 3–7) scattered setae or sometimes glabrous, dematiaceous external cells (8–12  $\mu\text{m}$  diam.), internal cells hyaline, undifferentiated. *Sclerotial setae* 3–4  $\mu\text{m}$  diam., when present more or less cylindrical but sometimes slightly sinuate, slightly attenuated distally to a blunt tip, dematiaceous or hyaline, 0–13 septate (Leakey 1964).

*Notes:* *Dactuliophora* was introduced by Leakey (1964) to accommodate four species of *Dactuliophora*, namely *D. elongata* C.L. Leakey, *D. glycines* C.L. Leakey, *D. harrisiae* C.L. Leakey and *D. tarrii* C.L. Leakey and typified with *D. tarrii* (Leakey 1964). The genus is characterised on the basis of the production of a cup-like ‘sclerotiophores’, which are parasitic on several economically important crops (Leakey 1964). Wijayawardene et al. (2014) placed *Dactuliophora* in *Pleosporaceae* thus we follow this classification. Currently five *Dactuliophora* species are listed in Index Fungorum (2015) including *D. anuae* S.M. Singh., and no molecular data is available for any of these species. Therefore fresh collections of the type species of the genus are needed so that molecular data can be obtained to verify the natural taxonomic affinities of this genus.

*Decorospora* Inderb et al., in Inderbitzin et al., Mycol. Progr. 1(4): 657 (2002).

**Facesoffungi number: FoF 00514**

*Saprobic* on dead wood and maritime plants in marine habitats. **Sexual morph:** *Ascomata* subglobose to ellipsoidal,

immersed, ostiolate, epapillate or short papillate, carbonaceous, black. *Peridium* composed of thick-walled cells with large lumina, forming a *textura angularis* in longitudinal section. *Hamathecium* composed of septate, branch pseudoparaphyses. *Asci* 8-spored, bitunicate, fissitunicate, clavate, short pedicellate, thick-walled, with a clear ocular chamber. *Ascospores* biseriata, ellipsoidal, muriform, brown, covered by a gelatinous sheath that is slightly constricted around the centre and drawn out at each apex into 2 or rarely 3 subconical extensions. **Asexual morph:** undetermined.

*Decorospora gaudefroyi* (Pat.) Inderb. et al., in Inderbitzin et al., Mycol. Progr. 1(4): 657 (2002), Fig. 12

Basionym: *Pleospora gaudefroyi* Pat., Tabl. analyt. Fung. France (Paris) 10: 40 (no. 602) (1886)

= *Pleospora salsolae* Fuckel var. *schoberiae* (Sacc.) Sacc., Syll. fung. (Abellini) 2: 248 (1883)

= *Pleospora schoberiae* (Sacc.) Berl., Icon. fung. (Abellini) 2: 23 (1895)

= *Pleospora lignicola* J. Webster & M. T. Lucas, Trans. Br. mycol. Soc. 44(3): 431 (1961)

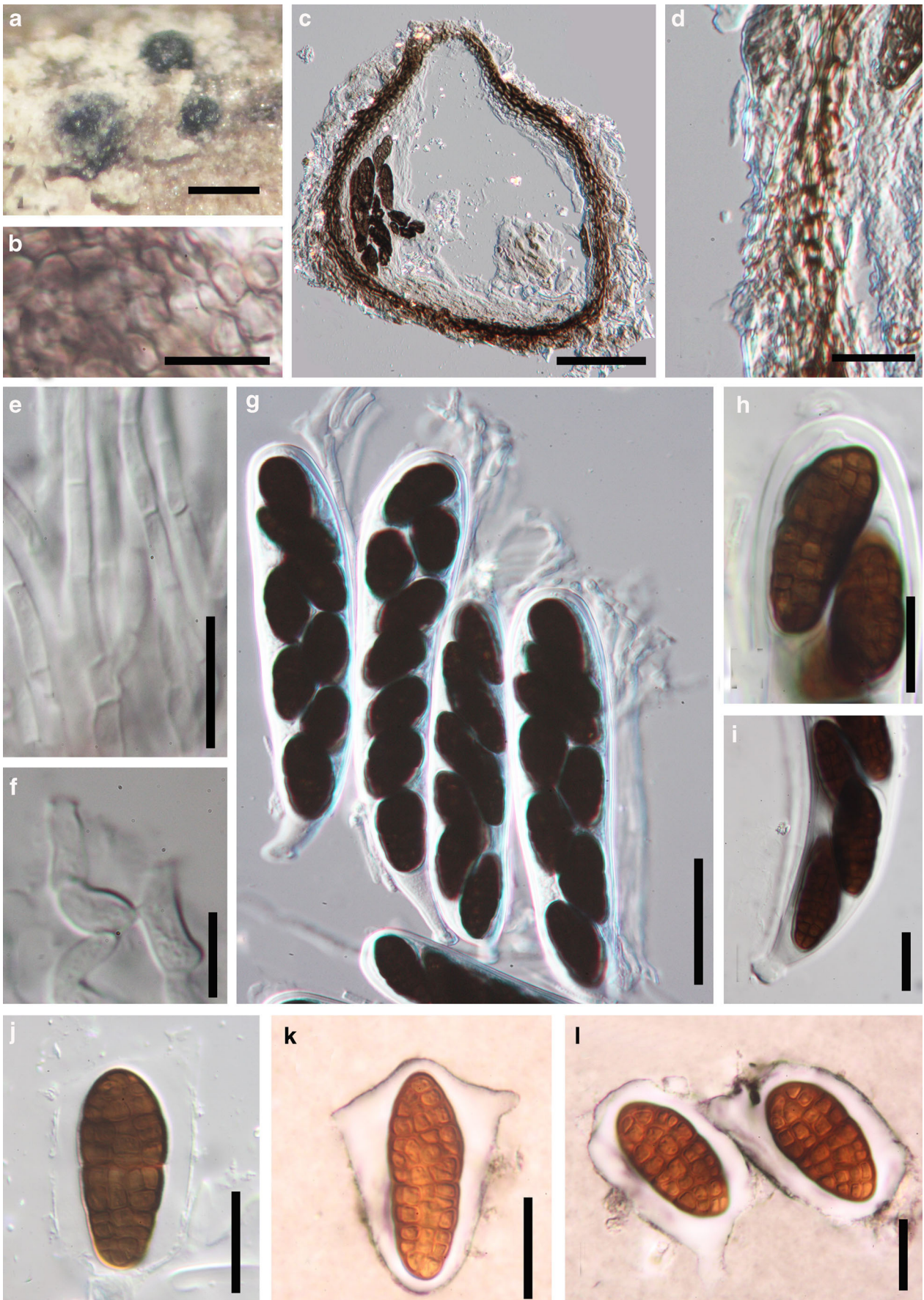
= *Pleospora salicorniae* Jaap, Verh. Bot. Ver. Prov. Brandenburg 49, 16. 1907 (non *Pleospora salicorniae* P. A. Dang. 1888)

= *Pleospora herbarum* var. *salicorniae* Jaap, Annl. mycol. 14(1/2): 17 (1916).

**Facesoffungi number: FoF 00515**

*Saprobic* on dead wood in marine habitats. **Sexual morph:** *Ascomata* perithecioid, small, 200–250  $\mu\text{m}$  in diam., globose to subglobose, immersed to erumpent, becoming superficial on the substrate at maturity, carbonaceous, black, solitary or several clustered, short ostiolate, papillate. *Ostirole* central, short, brown to black. *Peridium* thin, composed of thick-walled cells, forming a *textura angularis* in both surface view and longitudinal section, up to 12  $\mu\text{m}$ . *Hamathecium* composed of 2  $\mu\text{m}$  wide, hypha-like, cellular, septate, branched pseudoparaphyses anastomosing above the asci. *Asci* 80–125  $\times$  15–25  $\mu\text{m}$  ( $\bar{x}$  = 104  $\times$  18  $\mu\text{m}$ ,  $n$  = 10), 8-spored, bitunicate, fissitunicate, clavate, short pedicellate, thick-







◀ **Fig. 12** *Decorospora gaudefroyi* (holotype). **a.** Ascomata on the host surface. **b.** Peridium cells from surface view. **c.** Section of ascoma. **d.** Peridium. **e–f.** Branched pseudoparaphyses. **g.** Young asci amongst pseudoparaphyses. **h.** Tip of the ascus. Note the ocular chamber. **i.** Pedicellate ascus base. **j–l.** Ascospores. **k, l** Ascospores stained in Indian ink. Note the sheath. **Scale bars:** a=300  $\mu\text{m}$ , b=10  $\mu\text{m}$ , c=50  $\mu\text{m}$ , d–e=10  $\mu\text{m}$ , f=5  $\mu\text{m}$ , g=30  $\mu\text{m}$ , h–l=10  $\mu\text{m}$

walled with large lumina, apically rounded with an ocular chamber. *Ascospores* 20–25 $\times$ 7–10  $\mu\text{m}$  ( $\bar{x}$ =21.2 $\times$ 9  $\mu\text{m}$ ,  $n$ =10), biseriate, ellipsoidal, muriform, 6–7 transverse septa and 2–3 longitudinal septa in each segment, slightly constricted at the centre transverse septum, brown, covered by a gelatinous thick sheath with 2 or rarely 3 extensions. **Asexual morph:** undetermined.

**Material examined:** France, Marais de la Pointe de Touquet, Etaples, on *Suaeda maritima* (L.) Dumort, 15 Aug. 1879, O. Hariot (PC 0084490, **holotype**)

*Notes:* *Decorospora gaudefroyi* (Pat.) Inderbn et al., is a marine ascomycete firstly described as *Pleospora gaudefroyi* Pat. from northern France in 1886 and described by Inderbn et al. (2002). Yusoff et al. (1994) described the ascospores of *P. gaudefroyi* at the ultrastructural level without the unfolded sheath extensions. Based on analysis of partial SSU and ITS ribosomal DNA gene data transferred *P. gaudefroyi* to a new genus *Decorospora*, as it did not group in *Pleospora*. Morphological characters of *D. gaudefroyi* include black, erumpent ascomata becoming superficial on the substrate at maturity, septate and branched pseudoparaphyses, fissitunicate, clavate asci with thick wall, as well as yellow-brown ascospores with seven transverse and 1–3 longitudinal septa with a gelatinous sheath with a tripartite outer boundary. The characteristic sheath makes these species easy to distinguish from any other marine ascomycetes. *D. gaudefroyi* was considered to be a synonym of *Pleospora herbarum* (Fr.) Rabh. which is the type species of *Pleospora* (Yusoff et al. 1994). However, the presence of the ascospore sheath with its apical extensions and its marine habitat distinguish it from other marine *Pleospora* species (Inderbn et al. (2002). *Decorospora* is placed in *Pleosporaceae*, which is well-supported by partial SSU and ITS ribosomal DNA sequences.

During our phylogenetic analysis, the type strain of *Decorospora gaudefroyi* (pp4723) forms a robust clade basal to *Comoclathris*. Furthermore the putative strains of *Pleospora* sp. ATCC MYA-3203 and *Pleospora* sp. ATCC MYA-3202 are included. Thus we renamed them as *Decorospora* sp. in this study.

***Diademosia*** Shoemaker & C.E. Babco., Can. J. Bot. 70(8): 1641 (1992).

***Facesoffungi* number: FoF 00516**

*Saprobic* on stems and wood. **Sexual morph:** *Ascomata* immersed, initially erumpent becoming superficial, scattered, depressed-globose, some flattened at the base, opening a disc-like lid of brown prismatic cells with setae. *Peridium* composed of brown pseudoparenchyma cells of *textura angularis*. *Hamathecium* comprising numerous, septate, hyaline, cellular pseudoparaphyses. *Asci* 8-spored, bitunicate, fissitunicate, clavate with short narrow pedicel and minute ocular chamber. *Ascospores* biseriate, partially overlapping, fusiform, straight, frequently circular in section but narrowing to one end, with transverse and vertical septa, pale brown to dark brown, smooth walled. **Asexual morph:** undetermined.

**Type species:** *Diademosia californiana* (M.E. Barr) Shoemaker & C.E. Babco. [as ‘californianum’], Can. J. Bot. 70(8): 1641 (1992), Fig. 13

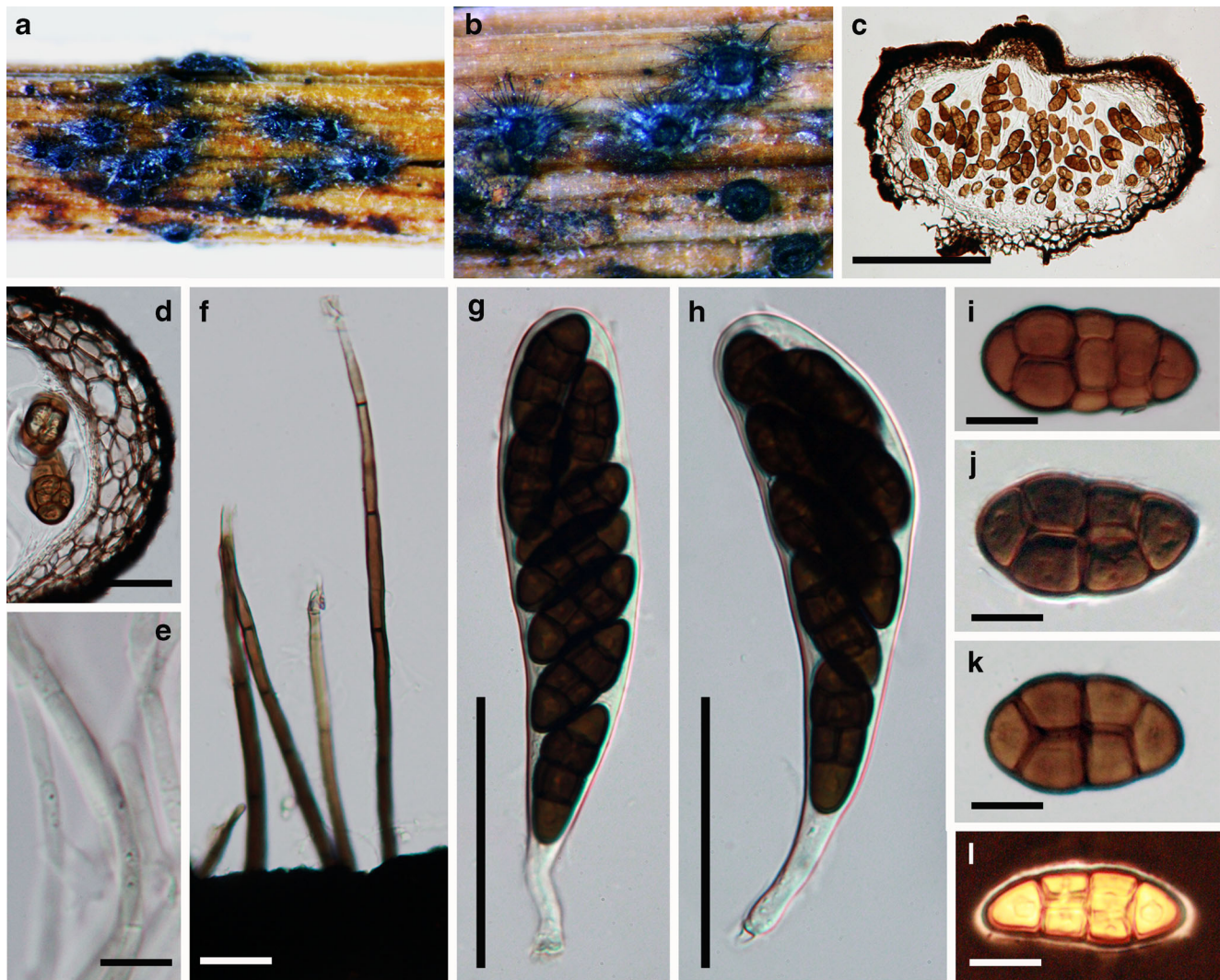
Basionym: *Graphyllum californianum* M.E. Barr, Mem. N. Y. bot. Gdn 62: 40 (1990).

***Facesoffungi* number: FoF 00517**

*Saprobic* on stem and wood. **Sexual morph:** *Ascomata* 200–365 $\times$ 240–425  $\mu\text{m}$  ( $\bar{x}$ =315 $\times$ 275  $\mu\text{m}$ ,  $n$ =10), immersed, initially erumpent becoming superficial, scattered, depressed-globose, some flattened at the base, opening a disc-like lid of brown prismatic cells with setae. *Peridium* 24–59  $\mu\text{m}$  ( $\bar{x}$ =32  $\mu\text{m}$ ,  $n$ =20), composed brown pseudoparenchyma cells of *textura angularis*. *Hamathecium* of dense, 2–3  $\mu\text{m}$  diam. ( $\bar{x}$ =2  $\mu\text{m}$ ,  $n$ =20), numerous, septate, hyaline, cellular pseudoparaphyses. *Asci* 140–175 $\times$ 24–28  $\mu\text{m}$  ( $\bar{x}$ =150 $\times$ 26  $\mu\text{m}$ ,  $n$ =20), 8-spored, bitunicate, fissitunicate, clavate with short narrow pedicel and minute ocular chamber. *Ascospores* 50–65 $\times$ 26–32  $\mu\text{m}$  ( $\bar{x}$ =57 $\times$ 29  $\mu\text{m}$ ,  $n$ =40), biseriate or discontinuously arranged, partially overlapping, fusiform, straight, cylindrical, frequently circular in end view with transverse and vertical septa, muriform, constricted at first septum, pale brown to dark brown, smooth walled with narrow sheath. **Asexual morph:** undetermined.

**Material examined:** USA, Bump-Cold Boiling Lake Trail, Lassen Volcanic National Park, Shasta, California, on branch of *Wyethia* Nutt (*Asteraceae*), 12 July 1966, W.B. Cooke & D.L. Hawksworth. (NY, **holotype**).

*Notes:* *Diademosia* was established by Shoemaker and Babcock (1992) and typified by *D. californiana*, based on the ascomatal opening via a circular lid and ascospores being frequently circular in end view. *Diademosia californiana* was initially introduced as *Graphyllum californianum* by Barr (1990) and referred to *Hysteriaceae* based on the pore or slit like opening. Re-examination of the type specimens by Shoemaker and Babcock (1992) concluded that *Diademosia* opened by a flat lid similar with *Diadema* and assigned it in to *Diademaceae*. The lid is hard to observe in sections unless they are mounted directly in lactic acid



**Fig. 13** *Diademosa californiana* (holotype) **a, b.** Ascomata on host substrate. **c.** Section of ascoma. **d.** Section of peridium. **e.** Septate, hyaline, cellular pseudoparaphyses **f.** Light to dark brown setae. **g, h.**

Ascus with minute pedicel bearing 8 irregularly arranged ascospores. **i–k.** Ascospores. **l.** Ascospore stained in Indian ink and showing narrow sheath. **Scale bars:** d–e=200  $\mu$ m, f–h=10  $\mu$ m, g–h=50  $\mu$ m, i–l=10  $\mu$ m

because excessive swelling occurs in water (Shoemaker and Babcock 1992). *Diademosa* differs from *Comoclathris* in having cylindrical ascospores which are frequently circular in section, but narrow of one end, as compared with the flattened ascospores of *Comoclathris*. Ariyawansa et al. (2014a) placed *Diademosa* in *Pleosporaceae* based on its similarities with other genera in this family, but confirmation of the phylogenetic status of this genus depends on recollecting the fungus and epitypification with molecular sequences.

*Exserohilum* K.J. Leonard & Suggs, *Mycologia* 66(2): 289 (1974)

= *Setosphaeria* K.J. Leonard & Suggs, *Mycologia* 66(2): 294 (1974).

= *Luttrellia* Khokhr. & Gornostaĭ, in Gornostaĭ in Azbukina et al. (Eds), *Vodorosli, Griby i Mkhi Dal'nego*

*Vostoka* [Algae, Fungi and Mosses of the Soviet Far-East] (Vladivostok): 80 (1978)

**Facesoffungi number: FoF 00518**

*Saprobic* or *pathogenic* on leaves or wood. **Sexual morph:** *Ascomata* superficial, globose or ellipsoid with or without an ostiole. *Ostiole* surrounded by simple, short, rigid, brown hairs, similar hairs scattered over surface of upper part of ascomata. *Peridium* coriaceous-carbonaceous, pseudoparenchymous. *Pseudoparaphyses* filamentous. *Asci* 1–8-spored, bitunicate, cylindrical to cylindrical-clavate. *Ascospores* hyaline, fusoid, trans-septate, surrounded by a mucilaginous sheath. **Asexual morph:** *Conidiophores* cylindrical, simple, olivaceous brown, upwardly geniculate. *Conidia* porogenous, acrogenous and pseudopleurogenous, subcylindrical to fusoid or broad obclavate-rostrate,



pseudoseptate, olivaceous to brown, hilum protruding from basal cell, germinating by bipolar germ tubes.

***Exserohilum turcicum*** (Pass.) K.J. Leonard & Suggs, *Mycologia* 66(2): 291 (1974), Fig. 14

≡ *Helminthosporium turcicum* Pass., *Boln Comiz. Agr. Parmense*: 3 (1876).

≡ *Bipolaris turcica* (Pass.) Shoemaker, *Canadian Journal of Botany* 37 (5): 884 (1959).

≡ *Drechslera turcica* (Pass.) Subram. & B.L. Jain, *Current Science* 35 (14): 355 (1966).

≡ *Luttrellia turcica* (Pass.) Khokhr., *Vodorosli, Griby i Mkhi Dal'nego Vostoka*: 81 (1978).

= *Helminthosporium inconspicuum* Cooke & Ellis, *Grevillea* 6 (39): 88 (1878).

= *Trichometasphaeria turcica* Luttr., *Phytopathology* 48(5): 282 (1958).

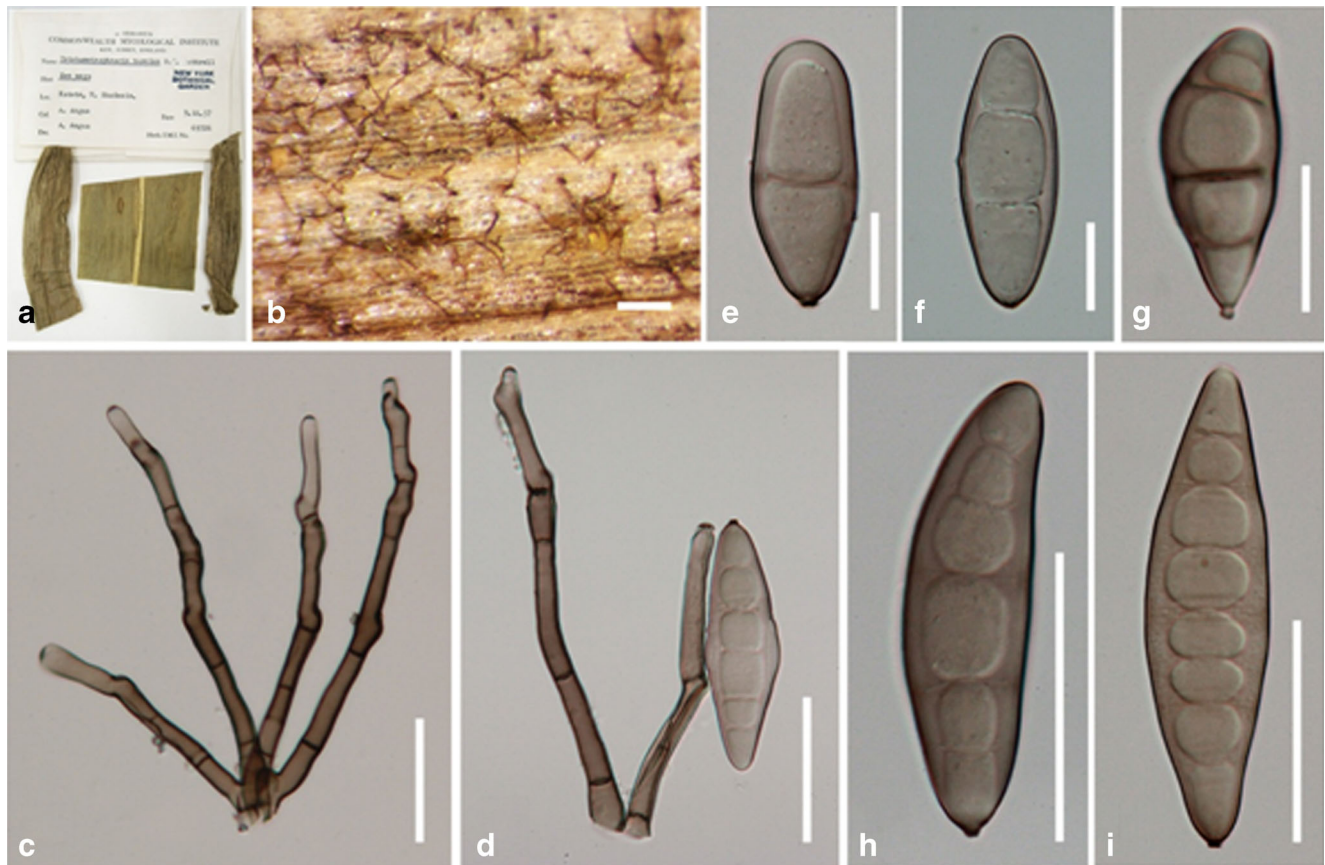
**Facesoffungi number: FoF 00519**

*Pathogenic* on living leaves of *Zea mays*. **Sexual morph:** undetermined. **Asexual morph:** *Conidiophores* (111-)147–164(–215) × 7.3–8.5(–11) μm ( $\bar{x}$ =158 × 8.2 μm,  $n$ =20) erect singly or in groups of 2–5, long, straight or flexuous, cylindrical, unbranched, or branched below, 2–5-septate, grayish brown,

inner wall layers of conidiogenous cell are continuous with the conidial wall, pale to medium brown. *Conidia* (41-)112–127 × (17-)22–23(–29) μm ( $\bar{x}$ =99 × 23 μm,  $n$ =20), ellipsoid-fusiform, widest at the middle and tapering toward ends, long, straight or slightly curved, 2–8-distoseptate, some euseptate, grayish brown, thick-walled, smooth, scars thickened.

**Material examined:** ZAMBIA, Katete, on living leaves of *Zea mays* (*Poaceae*), 9 February 1957, A. Angus, NY (Ex herbarium: IMI 69726).

**Notes:** The genus *Exserohilum* was proposed by Leonard and Suggs (1974) to accommodate species which were previously classified in *Bipolaris*, in which the conidial hilum was distinctly protuberant. In the same study, Leonard and Suggs (1974) introduced a new genus *Setosphaeria* to place the sexual morphs of *Exserohilum*, which is segregated from *Keissleriella* on the basis of lack of a clypeus, lysigenous development of the ostiole, occurrence of setae on the perithecial wall, the absence of periphyses in the ostiole and the hyphomycetous conidial states. As a genus can now only have one name, Wijayawardene et al. (2014) proposed to conserve *Exserohilum* over *Setosphaeria* on the basis having of more epithets (Index Fungorum 2015), and is more widely use in the literature.



**Fig. 14** *Exserohilum turcicum* (holotype). **a.** Herbarium material. **b.** Conidiophores and conidia on leaf surface of *Zea mays*. **c.** Conidiophores. **d.** Conidiophore and conidium. **e.** I Immature and mature conidia. **Scale bars:** b=200 μm, c, d, h, i=50 μm, e–g=20 μm



During our phylogenetic study putative strains of *Exserohilum monoceras* (= *Setosphaeria monoceras* CBS 154.26), *Exserohilum* sp. (C950801) and *Exserohilum* sp. (NK93) formed a well-supported clade sister to *Curvularia*. Therefore based on phylogeny and morphology we accept *Exserohilum* as a separate genus in the family *Pleosporaceae*.

*Extrawettsteinina* M.E. Barr, Contr. Univ. Mich. Herb. 9(8): 538 (1972).

**Facesoffungi number: FoF 00520**

*Saprobic* on dead leaves and wood. **Sexual morph:** *Ascomata* scattered, erumpent to superficial, globose or conical, dark brown to black, carbonaceous. *Papilla* black, with a pore-like ostioles, ostiolar canal filled with periphyses. *Peridium* a single layer composed of compressed, thick-walled cells, fusing with the host at the outside. *Hamathecium* of dense broad, cellular pseudoparaphyses. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical, apex rounded with and a minute ocular chamber with short pedicel. *Ascospores* uniseriate, slightly overlapping, ellipsoid, obovate-clavate, 3-septate, slightly to not constricted at the septum, wall smooth. **Asexual morph:** undetermined.

*Extrawettsteinina minuta* M.E. Barr, Contr. Univ. Mich. Herb. 9(8): 538 (1972), Fig. 15

≡ *Kriegeriella minuta* (M.E. Barr) Arx & E. Müll., Stud. Mycol. 9: 88 (1975)

**Facesoffungi number: FoF 00521**

*Saprobic* on dead leaves of *Juniperus communis*. **Sexual morph:** *Ascomata* 100–150 × 210–350 μm ( $\bar{x}$  = 120 × 280 μm,  $n$  = 10), scattered, erumpent to superficial, globose or conical, dark brown to black, carbonaceous. *Papilla* black, with a pore-like ostioles, ostiolar canal filled with periphyses. *Peridium* 10–25 μm ( $\bar{x}$  = 21 μm,  $n$  = 10), a single layer composed of compressed, thick-walled cells, fusing with the host at the outside. *Hamathecium* of dense 1–1.5 μm ( $\bar{x}$  = 1.25 μm,  $n$  = 20), broad, cellular pseudoparaphyses. *Asci* 75–90 × 6–8 μm ( $\bar{x}$  = 80 × 7 μm,  $n$  = 10), 8-spored, bitunicate, fissitunicate, cylindrical, apex rounded with and a minute ocular chamber with short pedicel. *Ascospores* 10–12 × 4–5 μm ( $\bar{x}$  = 11.5 × 4 μm,  $n$  = 10), uniseriate, slightly overlapping, ellipsoid, obovate-clavate, 3-septate, slightly to not constricted at the septum, wall smooth. **Asexual morph:** undetermined.

**Material examined:** USA, Vermont, Stow Pinnacle trail, on dead leaves of *Juniperus communis* L (*Cupressaceae*), 11 July 1964, H.E and M.E Bigelow (NY, 4278).

**Notes:** During our study we observed the holotype of *Extrawettsteinina minuta* (NY, 01103003) collected by H. E. Bigelow (19 August 1957) from *Juniperus communis* but we

could not find any ascomata in the type material. Therefore we examined another collection of *E. minuta* (NY, 4278), which was collected from same host and by the same collector as present in Fig. 15.

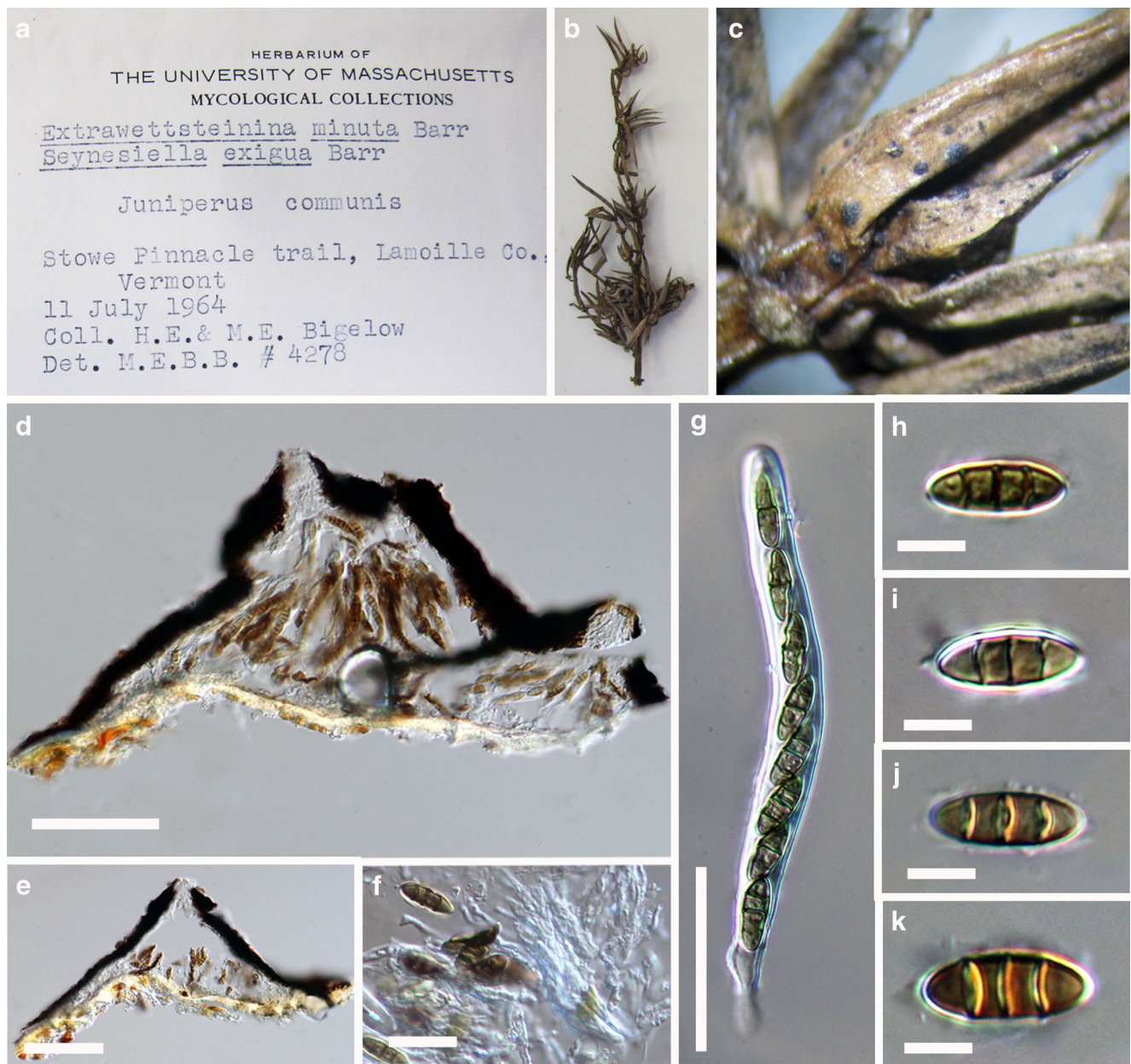
*Extrawettsteinina* was introduced by Barr (1972) and typified by *E. minuta*. The genus was introduced to accommodate three species, i.e. *E. minuta*, *E. pinastri* M.E. Barr and *E. mediterranea* (E. Müll.) M.E. Barr, which are saprobic on the dead leaves of gymnosperms and angiosperms, in North America and Europe (Barr 1972). Subsequently, a fourth species was introduced, viz. *E. andromedae* (Auersw.) M.E. Barr (based on *Wettsteinina andromedae*) by Barr (1987a). *Extrawettsteinina* is characterized by superficial, conical ascomata, containing a few saccate bitunicate asci and ellipsoidal, obovate-clavate, septate, smooth and hyaline ascospores which turn dull brown at maturity (Barr 1972). The diagnostic character of *Extrawettsteinina* is its conic ascomata, which are superficial on the substrate, and radiating arrangement of wall cells, which makes it distinguishable from comparable genera such as *Stomatogone* and *Wettsteinina*. Morphologically, *Extrawettsteinina* is comparable with *Kriegeriella*. In particular, *E. pinastri* could not be distinguished from *K. transiens* or *K. mirabilis*. Thus, *K. transiens*, including *Extrawettsteinina pinastri*, was treated as synonyms of *K. mirabilis*, and was included in the section of *Kriegeriella* (von Arx and Müller 1975; Barr 1975). The other section of *Kriegeriella*, *Extrawettsteinina*, includes two previous *Kriegeriella* species, i.e. *K. minuta* and *K. mediterranea*. Barr (1987b) introduced a family, *Kriegeriellaceae* (*Dothideales*) to accommodate *Kriegeriella* and *Extrawettsteinina*. This proposal has rarely been followed.

In this study we tentatively refer *Extrawettsteinina* in *Pleosporaceae* following Zhang et al. (2012) and Lumbsch and Huhndorf (2010). Currently no molecular data is available for this genus therefore generic type needs to be collected, isolated and sequenced to show the natural classification of this genus.

*Gibbago* E.G. Simmons, Mycotaxon 27: 108 (1986)

**Facesoffungi number: FoF 00522**

*Saprobic* or *pathogenic* on leaves and wood. **Sexual morph:** undetermined. **Asexual morph:** *Conidiophores* solitary or 2–4 loosely fasciculate, erect, rarely distantly branched, simple with a single apical conidiogenous locus, then often proliferating by means of a secondary conidiophore that arises immediately below the apical cell of the existing conidiophore, septate, slightly pigmented. *Conidia* initially solitary, ellipsoid, beakless, pigmented, becoming transversely and longitudinal septate, with apical cells swelling slightly and producing secondary conidia similar to the initial ones (Simmons 1986).



**Fig. 15** *Extrawettsteinina minuta* (NY, 4278). **a, b.** Herbarium material. **c.** Fruiting bodies on the host. **d, e.** Sections of ascomata. **f.** Cellular pseudoparaphyses. **g.** Ascus with short pedicel. **h-k.** Ellipsoid, obovate-

clavate, 3-septate ascospores. **Scale bars:** d=80  $\mu$ m, e=50  $\mu$ m, f=10  $\mu$ m, g=30  $\mu$ m, h-k=5  $\mu$ m

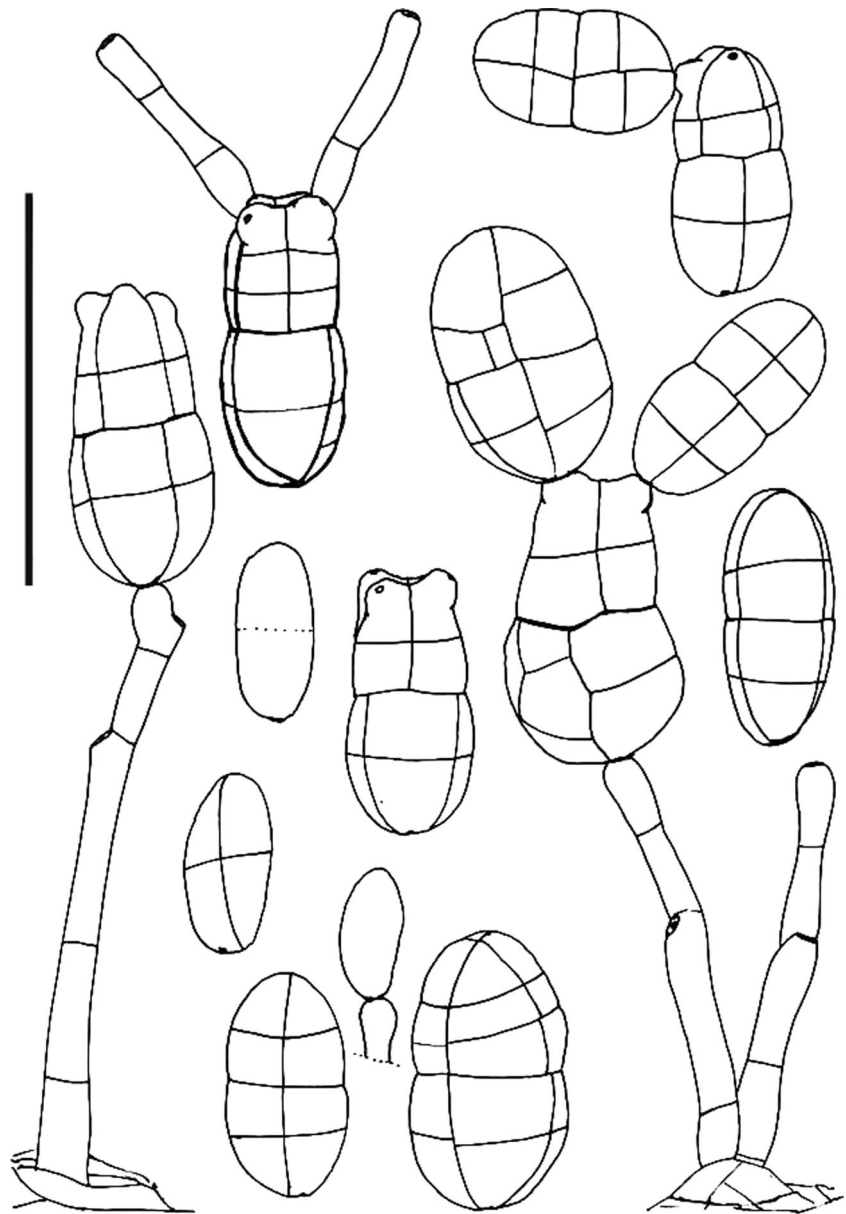
**Type species:** *Gibbago trianthemae* E.G. Simmons, Mycotaxon 27: 108 (1986), Fig. 16  
**Facesoffungi number:** FoF 00523

**Notes:** *Gibbago* was introduced by Simmons (1986) and typified by *G. trianthemae*. The genus is characterised by erect, simple with a single apical conidiogenous locus, septate, slightly pigmented, having solitary conidiophores with ellipsoid, beakless conidia and producing secondary conidia. *Gibbago* was assigned to *Pleosporaceae* by Simmons (1986) based

on its similarities with *Alternaria*, *Embellisia*, *Ulocladium* and *Stemphylium* and this was followed by Wijayawardene et al. (2014). During our phylogenetic analysis, the putative strains of *Gibbago trianthemae* GT-VM and NFCCI 1886 forms a distinct clade basal to *Paradendryphiella* and *Pleospora* clades with low bootstrap support. Therefore based on morphology coupled with molecular data, we tentatively refer *Gibbago* in *Pleosporaceae*, but this needs to be confirmed with further sequences of the monotypic genus and type species *G. trianthemae*.



**Fig. 16** *Gibbago trianthemae*  
(Redrawn from the holotype in  
Simmons 1986, Fig. 1). Scale  
bar: = 50  $\mu$ m



*Johnalcornia* Y.P. Tan & R.G. Shivas, in Tan et al., Australas. Pl. Path.: 10.1007/s13313-014-0315-6 (2014).

**Facesoffungi number: FoF 00524**

*Type species: Johnalcornia aberrans* (Alcorn) Y.P. Tan & R.G. Shivas, in Tan et al., Australas. Pl. Path.: 10.1007/s13313-014-0315-6 (2014), Fig. 17

≡ *Bipolaris aberrans* Alcorn, Mycotaxon 39: 364 (1990)

≡ *Cochliobolus aberrans* Alcorn, Mycotaxon 39: 362 (1990)

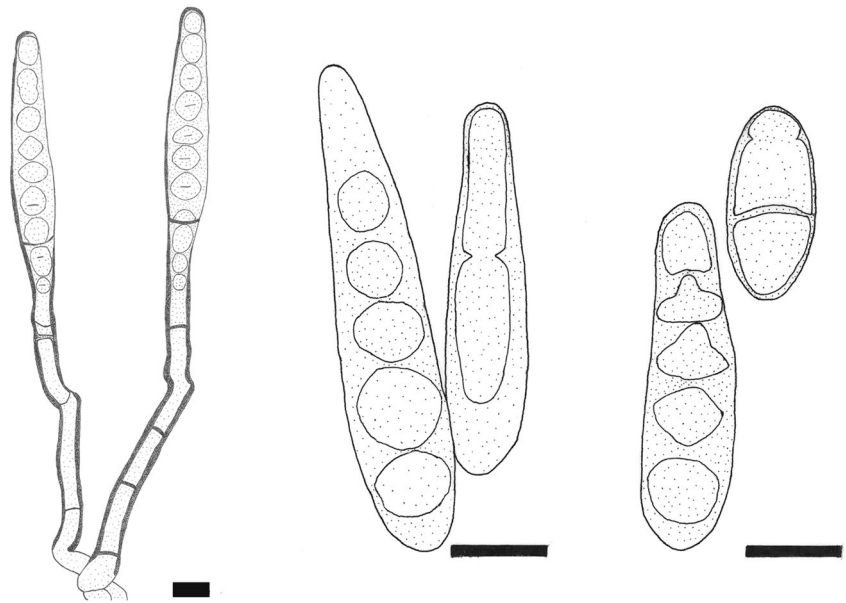
**Facesoffungi number: FoF 00525**

*Parasitic* on leaves. **Sexual morph:** *Ascomata* globose with a short neck. *Asci* cylindrical to fusoid, straight or curved. *Ascospores* hyaline, filiform, tightly coiled. **Asexual morph:**

*Conidiophores* solitary or in fascicles, simple or branched, straight to flexuous, apically geniculate, cylindrical, smooth. *Conidiogenous cells* integrated, cylindrical, proliferating sympodially, smooth to verruculose, with thickened and conspicuous scars. *Conidia* solitary, straight to curved, smooth, hilum inconspicuous, distoseptate, germinating from both polar cells, first conidial septum median or submedian, second conidial septum delimiting the apical cell, third septum basal (Description obtained from Tan et al. 2014).

*Notes:* *Johnalcornia* was introduced by Tan et al. (2014) to accommodate *Bipolaris aberrans* and is monotypic. Combined gene analysis of EF-1 $\alpha$ , GAPDH, ITS and LSU has shown that *J. aberrans* forms a sister clade to the newly described genus *Porocercospora*. *Johnalcornia* differs from *Porocercospora* in having distinctive conidia-like

**Fig. 17** *Johnalcornia aberrans*  
(Redrawn from the holotype in  
Tan et al. (2014), Fig. 2). **Scale**  
**bars:** 10  $\mu$ m



chlamydospores as well as comparatively thick-walled, geniculate conidiophores, with conidiogenous cells that have conspicuous scars. *Johnalcornia* further differs from related genera by forming the second conidial septum in the apical cell. In our phylogenetic analysis we also obtain the similar results where the type strain of *J. aberrans* forms a distinct clade sister to *Porocercospora* thus we accept *Johnalcornia* as a separate genus in the family *Pleosporaceae*.

***Mariellottia*** Shoemaker, Can. J. Bot. 76(9): 1559 (1999) [1998].

**Facesoffungi number:** FoF 00526

**Pathogenic** on cereals, grasses and seeds or saprobic on wood and leaves. **Sexual morph:** undetermined. **Asexual morph:** *Conidiophores* solitary to clustered, erect, septate, straight, brown, closely geniculate from sympodial growth or distantly nodose, apex truncate or inflated, forming conidia through a terminal pore, with flat, non-protruding scars, smooth or sometimes slightly roughened. *Conidiogenous cells* tretic, integrated, terminal, sympodially proliferating, cylindrical or swollen, cicatrized or not cicatrized. One species additionally produces massive, subulate macroconidiophores radiating from a pseudoparenchymatic base. *Conidia* porogenous, first septum formed near base, transversely 3-septate, exceptionally 2-septate when immature in one species and sometimes up to 5-septate in one species, brown, straight, ovoid or obovoid; scar pore-like, basal, dark brown, not protruding, set off by a hyaline zone, atrium type (Alcorn 1983) or flat in a slightly papillate base. Germ tubes usually two from the basal cell, rarely one from the apex, but never from the central cells (Description obtained from Shoemaker 1998).

**Type species:** *Mariellottia biseptata* (Sacc. & Roum.) Shoemaker, Can. J. Bot. 76(9): 1560 (1999) [1998], Fig. 18

≡ *Helminthosporium biseptatum* Sacc. & Roum., Revue Mycol. (Toulouse): 56 (1881).

≡ *Brachysporium biseptatum* (Sacc. & Roum.) Sacc., Sylloge Fungorum 4: 428 (1886).

≡ *Drechslera biseptata* (Sacc. & Roum.) M.J. Richardson & E.M. Fraser, Transactions of the British Mycological Society 51 (1): 148 (1968).

≡ *Pyrenophora biseptata* (Sacc. & Roum.) Crous, CBS Biodiversity Series 13: 257 (2013)

= *Helminthosporium bifforme* E.W. Mason & S. Hughes, Transactions of the British Mycological Society 30: 114 (1948).

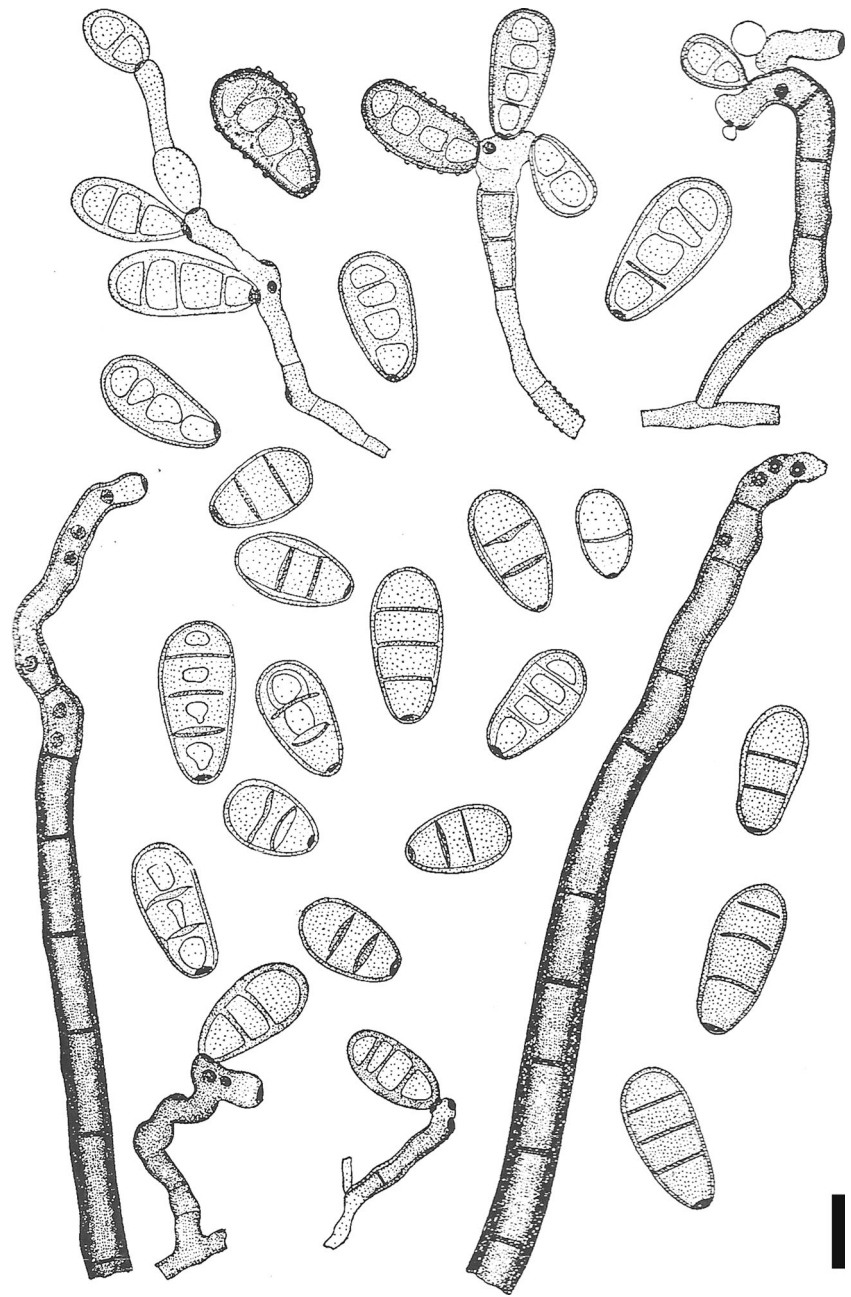
= *Drechslera bifforme* (E.W. Mason & S. Hughes) Subram. & B.L. Jain (1966).

**Facesoffungi number:** FoF 00527

**Saprobic** on stems. **Sexual morph:** undetermined. **Asexual morph:** *Conidiophores* two types. *Macronematous conidiophores* from stroma, single to many in a cluster, erect to divergent, long, broad at base, gradually narrowed towards apex, closely septate at intervals of 10–18 mm, dark chocolate brown; sporogenous area gnarled, frequently bent, with numerous dark brown scars, apex truncate, bearing an “ear” of conidia, 100–800×10–15  $\mu$ m (at base), 9–11  $\mu$ m wide near middle, 6–8  $\mu$ m wide near apex. *Conidiogenous cells* tretic, integrated, terminal, sympodially proliferating, cylindrical, cicatrized, stomata shallow ellipsoidal to depressed globose, 100–250×100–150  $\mu$ m. *Micronematous conidiophores* arising from hyaline to pale yellow hyphae (in culture), slender at base, sparingly septate, short, erect, sporogenous area gnarled, densely scarred, dark chocolate brown, usually simple, rarely



**Fig. 18** *Marielliottia biseptata*  
(Redrawn from *Drechslera*  
*biseptata* in Ellis 1971, Fig 288.).  
Scale bar: 100  $\mu\text{m}$



branched, 15–105  $\times$  5–8  $\mu\text{m}$ . *Conidiogenous cells* tretic, integrated, terminal, sympodially proliferating, cylindrical, cicatrized. *Conidia* obovoid, 3-septate, uppermost septum inconspicuous or lacking in some, rarely constricted at septa, thick walled, light to dark olive brown except lighter basal cell, narrowed to base, bearing a broad black nonprotruding scar 3–4.5  $\mu\text{m}$  wide, (21–)32–45(–49)  $\times$  (9–)11–13(–17)  $\mu\text{m}$ . Germination by one or two (three) oblique tubes near scar, rarely one hypha from apex and very rarely through the scar (Shoemaker 1998).

*Notes:* *Marielliottia* was introduced by Shoemaker (1998) to accommodate three species formerly treated in *Drechslera*

namely *D. biseptata* (Sacc. & Roum.) M.J. Richardson & E.M. Fraser, *D. dematioidea* (Bubák & Wróbl.) Scharif and *D. triseptata* (Drechsler) Subram. & B.L. Jain. The genus is typified with *Marielliottia biseptata*. The species differ from *Drechslera* in having mostly three-septate, obovoid to ovoid conidia that germinate primarily from the basal cell, occasionally from the apical cell, and not from the central cells (Shoemaker 1998). These fungi are occasional parasites of cereals and grasses, are sometimes seed borne, and may infect non-grass plants (Shoemaker 1998).

Currently no sequence data is available for *Marielliottia* species in GenBank. Therefore, fresh collections of the type

species of the genus are needed so that molecular data can be obtained to verify the natural taxonomic affinities of this genus. Based on the morphological similarities with *Drechslera*, we tentatively classified *Marielliotia* in *Pleosporaceae*.

*Neocamarosporium* Crous & M.J. Wingf., *Persoonia* 32: 273 (2014).

**Facesoffungi number: FoF 00528**

*Pathogenic* or *saprobic* on dying leaves and stems. **Sexual morph:** undetermined. **Asexual morph:** *Conidiomata* brown to black, immersed, becoming erumpent, globose with papillate apex and central ostiole; wall of 3–6 layers of brown *textura angularis*. *Conidiophores* reduced to conidiogenous cells. *Conidiogenous cells* lining the inner layer of conidioma, separate, hyaline, smooth, ampulliform; proliferating several times percurrently near apex, or at the same level, giving rise to prominent periclinal thickening. *Conidia* solitary, initially hyaline, aseptate, thick-walled, developing a central septum and then becoming muriformly septate, shape variable from globose to obovoid to ellipsoid, golden brown, finely roughened, thick-walled (Description obtained from Crous et al. 2014).

*Type species: Neocamarosporium goegapense* Crous & M.J. Wingf., *Persoonia*, *Mol. Phyl. Evol. Fungi* 32: 273 (2014), Fig. 19

**Facesoffungi number: FoF 0529**

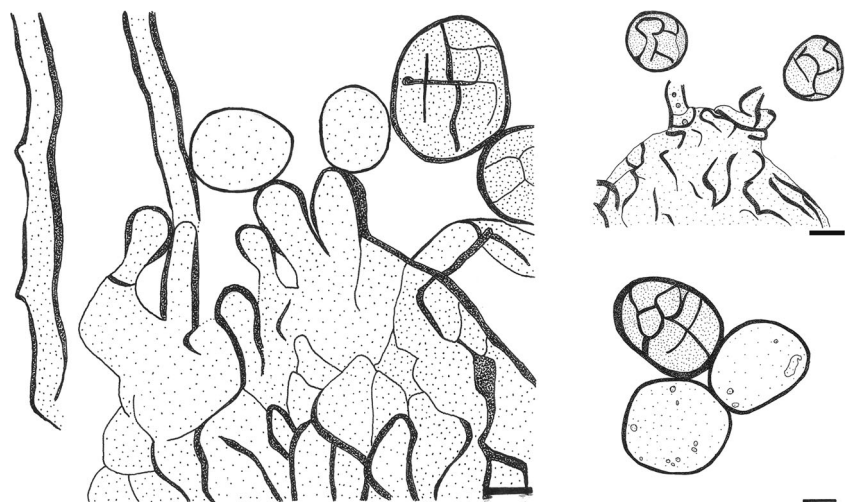
*Pathogenic* on dying leaves of *Mesembryanthemum* sp. (*Aizoaceae*). **Sexual morph:** undetermined. **Asexual morph:** *Conidiomata* 300 µm diam, brown to black, immersed, becoming erumpent, globose with papillate apex and central ostiole. *Conidiomata wall* consists of 3–6 layers of brown *textura angularis*. *Conidiophores* reduced to conidiogenous cells. *Conidiogenous cells* 7–9 × 5–6 µm, lining the inner layer

of conidioma, separate, hyaline, smooth, ampulliform, proliferating several times percurrently near apex, or at the same level, giving rise to prominent periclinal thickening. *Conidia* (15–) 20–22(–24) × 15–17(–19) µm, solitary, initially hyaline, aseptate, thick-walled, developing a central septum and then becoming muriformly septate, shape variable from globose to obovoid to ellipsoid, golden brown, finely roughened, thick-walled. (Description obtained from Crous et al. 2014)

*Notes: Neocamarosporium* was introduced by Crous et al. (2014) and typified with *Neocamarosporium goegapense*. *Neocamarosporium* is morphologically similar to *Camarosporellum*, though the latter appears to have holoblastic conidiogenesis. Based on a megablast search of NCBI's GenBank nucleotide database of LSU and ITS sequences, Crous et al. (2014) referred *Neocamarosporium* to *Pleosporaceae*. Crous et al. (2014) also suggested that phylogenetically, *Neocamarosporium* is allied to a clade containing taxa accommodated in *Phoma*, *Chaetosphaeronema* and *Pleospora*, but morphologically quite distinct. In our phylogenetic analysis we observed that the type strain of *Neocamarosporium goegapense* (CBS 138008) forms a robust clade sister to the *Clathrospora* and *Decorospora* clades. Furthermore, putative strains of *Phoma betae* (CBS 109410), *Pleospora betae* (IMI 156653) and *Pleospora bjoerlingii* (ICMP10945) reside within the *Neocamarosporium* clade. de Gruyter et al. (2013) reported *Phoma betae* (CBS 109410) as the asexual state of *Pleospora betae* (IMI 156653), thus they synonymised *Phoma betae* under *Pleospora betae*. Furthermore based on the morphological and DNA data they synonymised *Pleospora bjoerlingii* under *Pleospora betae*. Therefore in this study we introduce *Neocamarosporium betae* to accommodate these species in genus *Neocamarosporium*.

*Neocamarosporium betae* (Berl.) Ariyawansa & K. D. Hyde, **comb. nov.**,

**Fig. 19** *Neocamarosporium goegapense* (Redrawn from the holotype of *Neocamarosporium goegapense* in Crous et al. 2014). Scale bars: 10 µm





**Index Fungorum number: IF 550956**

Basionym: *Pyrenophora echinella* var. *betae* Berl., Nuovo Giorn. Bot. Ital. 20: 208. 1888.

≡ *Pleospora betae* (Berl.) Nevod., Grib. ross. Exs., No. 247. 1915.

= *Pleospora betae* Björl., Bot. Not. 1944: 218. 1944. (later homonym), nom. illeg.

≡ *Pleospora bjoerlingii* Byford, Trans. Brit. Mycol. Soc. 46: 614. 1963.

= *Phoma betae* A.B... Frank, Z. Rübenzucker-Ind. 42: 904, tab. 20. 1892.

= *Phyllosticta betae* Oudem., Ned. Kruidk. Arch. Ser. 2, 2: 181. 1877.

= *Gloeosporium betae* Dearn. & E.T. Barthol., Mycologia 9: 356. 1917.

During our study the putative strains of *Pleospora chenopodii* (CBS 344.78), *P. calvescens* (CBS 246.79) and *P. halimiones* (CBS 432.77) also forms a separate clade basal to the type species of *Neocamarosporium*, *N. goegapense*. de Gruyter et al. (2013) treated these species as separate species but our phylogeny clearly show that these three species can be treated a one species thus we propose *Neocamarosporium calvescens* to accommodate *Pleospora chenopodii*, *P. calvescens* and *P. halimiones*.

*Neocamarosporium calvescens* (Fr. ex Desm.) Ariyawansa & K.D. Hyde, **comb. nov.**

**Index Fungorum number: IF 550957**

Basionym: *Sphaeria calvescens* Fr. ex Desm., Ann. Sci. Nat., Bot., 2e Sér., 19: 353. 1843.

≡ *Pleospora calvescens* (Fr. ex Desm.) Tul. & C. Tul., Selecta Fung. Carpol. 2: 266. 1863.

≡ *Pyrenophora calvescens* (Fr. ex Desm.) Sacc., Syll. Fung. 2: 279. 1883.

≡ *Chaetoplea calvescens* (Fr. ex Desm.) Clem., Gen. Fung., 2nd Ed.: 275. 1931.

≡ *Leptosphaeria calvescens* (Fr. ex Desm.) Crivelli, Ueber Heterog. Ascomycet. Pleospora: 177. 1983.

= *Phloeospora chenopodii* Ellis & Kellerm., J. Mycol. 4: 26. 1888, [as “Phleospora”].

≡ *Pleospora chenopodii* (Ellis & Kellerm.) Gruyter & Redhead, Index Fung. 205: 1. 2014.

= *Pleospora halimiones* Gruyter & Verkley, Stud. Mycol. 75: 25. 2012.

*Paradendryphiella* Woudenberg & Crous, Stud. Mycol. 75(1): 207 (2013)

**Facesoffungi number: FoF 00530**

*Saprobic* or *parasitic* in terrestrial and marine habitats. **Sexual morph:** undetermined. **Asexual morph:** *Conidiophores*

simple or branched, septate or not, straight or flexuous, often nodose with conspicuous, subhyaline, brown pigmentation at the apical region; at times reduced to conidiogenous cells. *Conidiogenous cells* terminal or lateral, with denticles aggregated at apex, with prominent conidial scars, thickened but not darkened; sometimes proliferating with a new head or a short, inconspicuous sympodial rachis. *Conidia* produced holoblastically, on narrow denticle, smooth, cylindrical to obclavate, straight or slightly flexuous, 1–7 transverse septa, pale to medium brown, often with dark septa (often constricted), and a darkened zone of pigmentation at the apex, and at the hilum, which is thickened, and somewhat protruding, with a minute marginal frill (Description obtained from Woudenberg et al. 2013).

*Type species: Paradendryphiella salina* (G.K. Sutherl.) Woudenberg & Crous, Stud. Mycol. 75(1): 207 (2013), Fig. 20

≡ *Cercospora salina* G.K. Sutherl., New Phytol. 15: 43 (1916).

≡ *Dendryphiella salina* (G.K. Sutherl.) Pugh & Nicot, Transactions of the British Mycological Society 47 (2): 266 (1964).

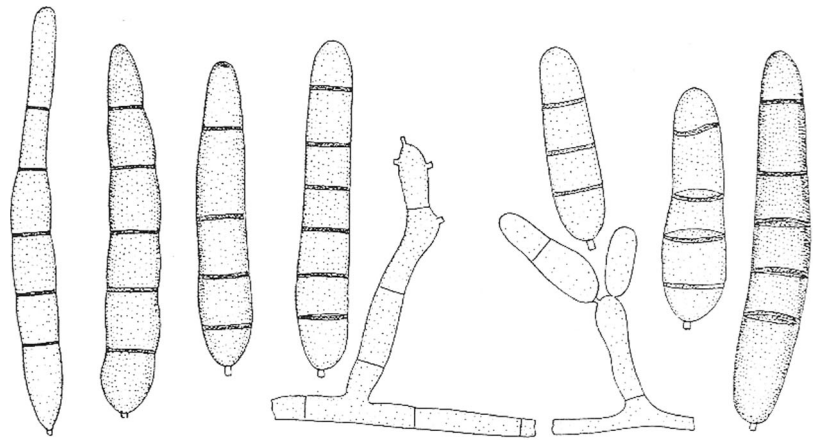
≡ *Scolecobasidium salinum* (G.K. Sutherl.) M.B. Ellis, More dematiaceous Hyphomycetes: 192 (1976).

**Facesoffungi number: FoF 0531**

*Saprobic* or *parasitic* in marine habitats. **Sexual morph:** undetermined. **Asexual morph:** *Conidiophores* 30–40×2–4 μm, simple or branched, septate or not, straight or flexuous, often nodose with conspicuous, brown pigmentation at the apical region; at times reduced to conidiogenous cells. *Conidiogenous cells* terminal or lateral, with denticles aggregated at apex, with prominent conidial scars, thickened but not darkened; sometimes proliferating with a new head or a short, inconspicuous sympodial rachis. *Conidia* 16–65×5–9 μm, produced holoblastically, on narrow denticle, smooth-walled, cylindrical to obclavate, straight or slightly flexuous, 1–7 transverse septa, pale to medium brown, often with dark septa (often constricted), and a darkened zone of pigmentation at the apex, and at the hilum, which is thickened, and somewhat protruding, with a minute marginal frill (Description of *Scolecobasidium salinum* obtained from Ellis 1976).

*Notes: Paradendryphiella* was introduced by Woudenberg et al. (2013) to accommodate two species *Dendryphiella arenariae* and *D. salina* (= *Cercospora salina*). *Paradendryphiella* is characterised by simple or branched conidiophores with a new head or a short, inconspicuous sympodial rachis conidiogenous cells and holoblastically produced cylindrical to obclavate, straight or slightly flexuous, 1–7 transverse septa, pale to medium brown conidia. The phylogenetic trees show that *Paradendryphiella* nested within the family *Pleosporaceae* and clustered sister to *Pleospora*

**Fig. 20** *Paradendryphiella salina* (Redrawn from *Scolecobasidium salinum* in Ellis 1976, Fig 138c.). **Scale bar:** 5  $\mu$ m



*sensu stricto* clade in Jones et al. (2008), Woudenberg et al. (2013) and as well as in this study. Therefore we accept *Paradendryphiella* as separate genus in the family *Pleosporaceae*.

***Platysporoides*** (Wehm.) Shoemaker & C.E. Babcock, Can. J. Bot. 70(8): 1648 (1992).

**Facesoffungi number:** FoF 00532

*Saprobic* in terrestrial habitats. **Sexual morph:** *Ascomata* small, scattered, immersed, semi-immersed to nearly superficial, globose, subglobose, black, smooth; apex with a protruding papilla and pore-like ostiole, without periphyses. *Peridium* thin, composed of a few layers of *textura angularis*. *Hamathecium* of numerous, cellular pseudoparaphyses, anastomosing above the asci, septate. *Asci* bitunicate, fissitunicate, cylindrical to cylindro-clavate, with a short, furcate pedicel and minute ocular chamber. *Ascospores* broadly ellipsoid, reddish brown, muriform. **Asexual morph:** undetermined

**Type species:** *Platysporoides chartarum* (Fuckel) Shoemaker & C.E. Babcock, Can. J. Bot. 70(8): 1650 (1992), Fig. 21

Basionym: *Pleospora chartarum* Fuckel, Jb. Nassau. Ver. Naturk. 23–24: 133 (1870) [1869–70]

≡ *Clathrospora chartarum* (Fuckel) O.E. Erikss., Arkiv for Botanik 6 (4–5): 352 (1967)

**Facesoffungi number:** FoF 00533

*Saprobic* in terrestrial habitats. **Sexual morph:** *Ascomata* 150–230  $\mu$ m  $\times$  180–260  $\mu$ m, scattered, immersed, semi-immersed to rarely superficial, globose, subglobose, black, smooth; apex with a protruding papilla, ostiolate. *Ostiole* comprises apex with a protruding papilla and without periphyses. *Peridium* 8–22  $\mu$ m wide, composed of 2–4 layers of brown cells of *textura angularis*, cells 5–9  $\mu$ m diam., cell wall 1–2.5  $\mu$ m thick. *Hamathecium* of 2–3  $\mu$ m broad, long cellular pseudoparaphyses, anastomosing above the asci, septate. *Asci*

110–140  $\times$  12.5–16.5  $\mu$ m ( $\bar{x}$ =121.5  $\times$  14.7  $\mu$ m,  $n$ =10), (6-)8-spored, bitunicate, fissitunicate, cylindrical to cylindro-clavate, with a short, furcate pedicel, 8–17  $\mu$ m long, and minute ocular chamber. *Ascospores* 20–26  $\times$  8–11  $\mu$ m ( $\bar{x}$ =23.7  $\times$  9  $\mu$ m,  $n$ =10), obliquely uniseriate and partially overlapping, flattened, broadly ellipsoid in front view, reddish brown, muriform, 3 transverse septa, one longitudinal septum in each central cell, 1 oblique septum in each end cell, constricted at all septa, granulate, with a 2–3  $\mu$ m wide mucilaginous sheath. **Asexual morph:** undetermined.

**Material examined:** GERMANY, Rhineland-Palatinate, on dead leaves, 1894, Herbar Barby-Boissier (HUH 00290381, **holotype**).

**Notes:** *Platysporoides* was introduced as a subgenus of *Pleospora* by Wehmeyer (1961) and was typified by *Pleospora chartarum*. Shoemaker and Babcock (1992) raised *Platysporoides* to generic rank and placed it in the *Pleosporaceae* based on its “aplanodictyospore” and “terete pored beak of the ascomata”. Currently, eleven species are included in this genus (Shoemaker and Babcock 1992). Another comparable pleosporalean family is *Diademaceae*, which is distinguished from *Platysporoides* by its ascoma opening as “an intraepidermal discoid lid” (Shoemaker and Babcock 1992). *Aigialus grandis* is another pleosporalean fungus with flattened and muriform ascospores as well as papilla and ostioles, which belongs to *Aigialaceae*, a phylogenetically well supported marine family (Suetrong et al. 2009). Thus, it is highly likely that flattened and muriform ascospores have evolved on separate occasions and not taxonomically informative.

Currently no molecular data is available for this genus and therefore it is essential to epitypify the generic type with fresh collections. Therefore molecular data can be used to resolve the correct natural classification of *Platysporoides*. We tentatively place *Platysporoides* in *Pleosporaceae* based on its





◀ **Fig. 21** *Platysporoides chartarum* (holotype). **a, b.** Specimen and herbarium material. **c.** Ascomata on substrate. **d.** Section through ascoma. **e.** Upper walled cells of ascoma. **f.** Peridium. **g.** Ascus when immature in Melzer's reagent. **h.** Ascus when immature in Cotton Blue reagent. **i.** Ascus at maturity. **j.** Hamathecium in Cotton Blue reagent. **k.** Ocular chamber in Melzer's reagent. **l.** Ascospores at maturity. **m, n.** Ascospore when immature with Sheath. **o.** Ascospore when immature in Cotton Blue reagent. **p.** Ascospore at maturity with sheath in India Ink. **Scale bars:** d=40  $\mu\text{m}$ , e–f=10  $\mu\text{m}$ , g–i=100  $\mu\text{m}$ , j=10  $\mu\text{m}$ , k–p=5  $\mu\text{m}$

similarities with the other genera of this family, such as, in having globose to subglobose ascomata with pore-like ostiole, a peridium composed of a few cell layers of *textura angularis*, cellular pseudoparaphyses, fissitunicate, cylindrical to cylindro-clavate asci with broadly ellipsoid, reddish brown, muriform ascospores.

*Pleospora* Rabenh. ex Ces. & De Not., Comm. Soc. crittog. Ital. 1(4): 217 (1863)

= *Stemphylium* Wallr., Fl. crypt. Germ. (Norimbergae) 2: 300 (1833)

= *Cleistotheca* Zukai, Österreichische Botanische Zeitschrift 53 (5): 163 (1893)

= *Cleistothecopsis* Stevens & True, Illinois Agric. Exper. Stat. Bull. Nr. 220: 507 (1919).

**Facesoffungi number: FoF 00534**

*Habitat terrestrial or aquatic, saprobic or parasitic* on stems and leaves. **Sexual morph:** *Ascomata* small to medium-sized, immersed, erumpent to superficial, base not easy to remove from the substrate, broadly to narrowly oblong and flattened, ostiolate. *Ostiole* papillate, black, smooth, ostiolar canal filled with hyaline cells. *Peridium* thin, usually with two layers, thick at the sides and thinner at the base, outer layer composed of heavily pigmented, thick-walled cells of *textura angularis*, inner layer composed of hyaline thin-walled cells of *textura angularis*, coriaceous. *Hamathecium* of cellular, hyaline, septate, broad, dense pseudoparaphyses. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical to clavate, with furcate pedicel and minute ocular chamber. *Ascospores* uniseriate or partially overlapping, mostly ellipsoidal, muriform, brown or pale brown, with or without sheath. **Asexual morph:** *Stemphylium*. *Mycelium* immersed or nearly superficial, brown. *Stroma* sometimes present. *Conidiophores* macronematous, mononematous, scattered or caespitose, unbranched or rarely loosely branched, straight or flexuous, usually nodose with a number of vesicular swellings, pale to mid brown or olivaceous brown, smooth or in part verruculose. *Conidiogenous cells* monoblastic, integrated, terminal, percurrent, at first clavate or subspherical with the wall at the apex thin. *Conidia* solitary, dry, acrogenous, oblong, rounded at the ends, ellipsoidal, obclavate or subspherical, pale to mid dark or olivaceous brown, smooth, verrucose or echinulate, muriform, often constricted at one or more of the septa, cicatrized at the base.

echinulate, muriform, often constricted at one or more of the septa, cicatrized at the base. *Colonies* effuse, grey, brown, olivaceous brown or black, velvety or cottony (Ellis 1971).

*Type species: Pleospora herbarum* (Pers.) Rabenh., Klotzschii Herb. Viv. Mycol. 2: no. 547 (1854), Fig. 22

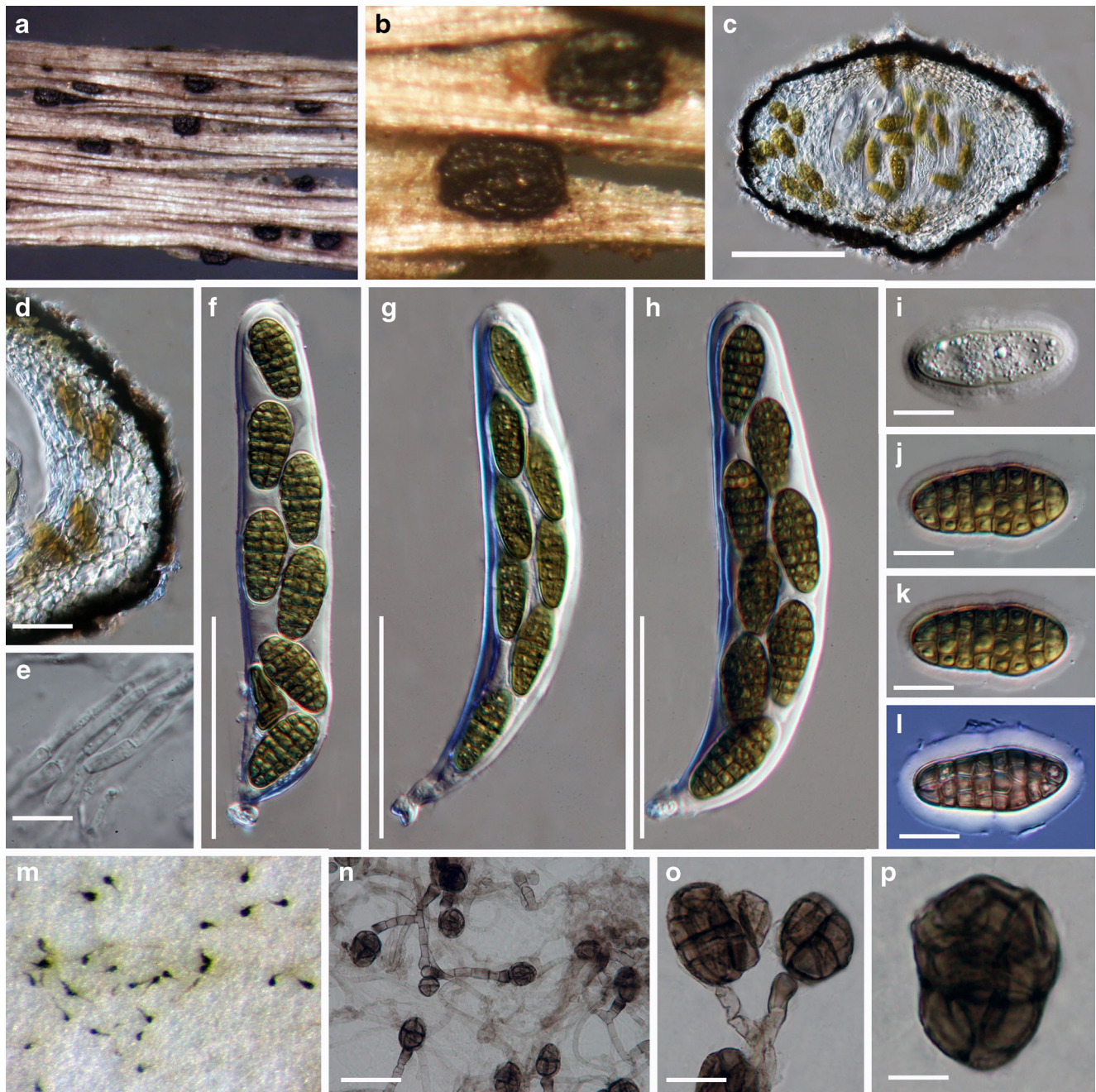
= *Sphaeria herbarum* Pers., Syn. meth. fung. (Göttingen) 1: 78 (1801)

**Facesoffungi number: FoF 00535**

*Habitat terrestrial, marine, saprobic or parasitic* on stems and leaves. **Sexual morph:** *Ascomata* 110–240×250–460  $\mu\text{m}$  small to medium-sized, immersed, erumpent to superficial, base not easy to remove from the substrate, broadly to narrowly oblong and flattened, ostiolate. *Ostiole* papillate, black, smooth, ostiolar canal filled with hyaline cells. *Peridium* 30–50  $\mu\text{m}$  ( $\bar{x}$ =42  $\mu\text{m}$ ,  $n$ =10) thin, usually with two layers, thick at the sides and thinner at the base, outer layer of heavily pigmented thick-walled cells of *textura angularis*, inner layer composed of hyaline thin-walled cells of *textura angularis*, coriaceous. *Hamathecium* of 2–3  $\mu\text{m}$  ( $\bar{x}$ =2.5  $\mu\text{m}$ ,  $n$ =10) cellular, hyaline, septate, broad, dense pseudoparaphyses. *Asci* 100–225×25–30  $\mu\text{m}$  ( $\bar{x}$ =142×28  $\mu\text{m}$ ,  $n$ =20), 8-spored, bitunicate, fissitunicate, cylindrical to clavate, with furcate pedicel and minute ocular chamber. *Ascospores* 25–40×12–15  $\mu\text{m}$  ( $\bar{x}$ =33×14  $\mu\text{m}$ ,  $n$ =40), uniseriate or partially overlapping, mostly ellipsoidal, muriform, brown or pale brown, with or without sheath. **Asexual morph:** *Stemphylium*. *Conidiophores* 4–6  $\mu\text{m}$  ( $\bar{x}$ =5  $\mu\text{m}$ ,  $n$ =10) macronematous, mononematous, scattered or caespitose, unbranched or rarely loosely branched, straight or flexuous, usually nodose with a number of vesicular swellings, pale to mid brown or olivaceous brown, smooth or in part verruculose. *Conidiogenous cells* monoblastic, integrated, terminal, percurrent, at first clavate or subspherical with the wall at the apex thin. *Conidia* 16–22×11–18  $\mu\text{m}$  ( $\bar{x}$ =21×16  $\mu\text{m}$ ,  $n$ =20) solitary, dry, acrogenous, oblong, rounded at the ends, ellipsoidal, obclavate or subspherical, pale to mid dark or olivaceous-brown, smooth, verrucose or echinulate, muriform, often constricted at one or more of the septa, cicatrized at the base. *Colonies* effuse, grey, brown, olivaceous-brown or black, velvety or cottony.

**Material examined:** ITALY, Forli-Cesena, Montevescovo, on dead stem of *Brassica nigra* (*Brassicaceae*), 01 January 2012, E. Camporesi IT 82 (MFLU 12–2216) – living culture (MFLUCC 14–0261); ITALY, Tessello - Cesena, on dead stem of *Cirsium* sp (*Asteraceae*), 16 December 2013, E. Camporesi IT 956 (MFLU 14–0762) – living culture (MFLUCC 13–0344); ITALY, Massera - Predappio, on dead fruits of *Lunaria rediviva* (*Brassicaceae*), 16 December 2013, E. Camporesi IT 958 (MFLU 14–0763) – living culture (MFLUCC 13–0266, ICMP)





**Fig. 22** *Pleospora herbarum* (MFLU12-2216). **a.** Ascomata on host substrate. **b.** Close up of ascoma. **c.** Section of ascoma. **d.** Close up of the peridium. **e.** Cellular, hyaline, septate, broad pseudoparaphyses. **f–h.** Asci with short, broad pedicel bearing 8 ascospores **i–l.** Mature and

immature ascospores with mucilaginous sheath. *Stemphylium* sp. **m–p.** Conidiophores with pale to mid dark or olivaceous brown, smooth conidia. **Scale bars:** c=100  $\mu$ m, d=50  $\mu$ m, e=20  $\mu$ m, f–g=60  $\mu$ m, i–k=10  $\mu$ m, l–o=10  $\mu$ m

*Notes:* *Pleospora* was originally described by Rabenhorst (1857) and is typified by *Pleospora herbarum* (Pers.) Rabenh., (Kirk et al. 2008). Initially the genus was included in *Sphaeriales* and later *Pseudosphaeriales* and *Pleosporales*, respectively (Wehmeyer 1961). All species belonging to *Pleospora* have muriform ascospores (Wehmeyer 1961, 1975). Pseudoparaphyses arrangement (downward growing) within the ascomata of “*Pleospora*-

type” development (Luttrell 1951) is considered to be the main feature for the genus. Various authors had included and excluded different species in *Pleospora* at various times. Barr (1981) placed *Curreya* in *Pleospora* however, von Arx and van der Aa (1983) treated it as separate genus, because of its *Coniothyrium* asexual morph. Petrak (1952) transferred *Graphyllum* to *Pleospora*, and noted that the elongate ascomata and closely grouped rows of

small ascomata are not sufficient to recognize the genus. Barr (1987b, 1990b) supported this proposal. Due to the heterogenous nature of *Pleospora* several subgenera have been included. *i.e.* *Teichosporoides* contains species of *Pleospora* with immersed ascomata, and *Pleospaeria* with superficial and setose ascomata (Wehmeyer 1961). *Pleospora* species have a wide host range, especially on monocotyledons as well as dicotyledonous plants (Wehmeyer 1975).

There are numerous species of *Dothideomycetes* that have muriform ascospores and have at one time or another been placed in *Pleospora* (Wehmeyer 1961). *Pleospora* is however, rather distinctive and should be confined to species with characters that are similar to *Pleospora herbarum*. The ascomata in *P. herbarum* are immersed and usually become erumpent, the peridium is unusual in having three strata, a thin inner layer of thin-walled, hyaline to light brown flattened cells, a relatively wide central layer of thin-walled, hyaline to light brown angular cells, and an outer very thin black amorphous cells, which gives the blackened colour to the ascomata. The asci are broadly clavate and have a distinctive, squarish, wide, ocular chamber and ascospores are muriform with at least three longitudinal septa per transverse row, and surrounded by a mucilaginous sheath. The asexual morph is *Stemphyllium* (Zhang et al. 2012; Hyde et al. 2013). For this reason several *Pleospora*-like species are now transferred to other genera (e.g. *Curreya*).

Based on our phylogenetic assessment we proposed to confine *Pleospora* to one well-supported clade based on the type strains of *P. herbarum* (CBS 191.86) along with the other alternative strains of *P. herbarum* used in this study (*P. herbarum* MFLUCC 13–0344, *P. herbarum* MFLUCC 13–0266 and *P. herbarum* (MFLUCC 14–0261), sister to *Paradendryphiella* clade applying GCPSR (Taylor et al. 2000; Dettman et al. 2003). By implementing this, we treat some putative strains, such as *P. tomatonis* (CBS109844), and *Pleospora sedicola* (CBS109843) as alternative strains of *P. herbarum*. Furthermore we treated the putative strains of *Pleospora papaveracea* (CBS432.50), *Pleospora herbarum* (DAOM195299), *Pleospora tarda* (CBS 714.68) and *Pleospora halophila* (CBS410.73) as separate species which clustered outside the *P. herbarum sensu stricto*.

Wijayawardene et al. (2014) proposed to conserve *Stemphyllium* over *Pleospora* because *Stemphyllium* has priority as it is an older name, is better established in the literature than *Pleospora* (more than double the number of hits on Google Scholar), and is a well-known genus to the plant pathology community, and is well circumscribed on a molecular basis (Lawrence et al. 2012). In this study the first author prefers to keep *Pleospora* because the entire family *Pleosporaceae*, as well as the order *Pleosporales*, is based on the *Pleospora*-type of centrum development.

***Porocercospora*** Amaradasa et al., Mycologia 106(1): 81 (2014)

***Facesoffungi* number: FoF 00536**

*Type species: Porocercospora seminalis* (Ellis & Everh.) Amaradasa et al., Mycologia 106(1): 81 (2013), Fig. 23

≡ *Cercospora seminalis* Ellis & Everh., J. Mycol. 4(1): 4 (1888)

≡ *Sporidesmium seminale* (Ellis & Everh.) U. Braun, Cryptogamie Mycologie 20 (3): 175 (1999).

***Facesoffungi* number: FoF 588.**

*Parasitic* on grasses. **Sexual morph:** undetermined. **Asexual morph:** *Conidiophores* 450–500×5–8 μm, intermingled among hyphae, subcylindrical, medium brown, smooth to finely verruculose, branched above, thin-walled, septate. *Conidiogenous cells* 20–30×5–7 μm, subcylindrical, medium brown, smooth to finely verruculose, apex rounded, monotretic, with a central pore, indistinct, not darkened or thickened. *Conidia* 60–90×7–8 μm solitary, medium brown, thick-walled, finely verruculose, obclavate to cylindro-obclavate, with short conidia obovoid to subcylindrical, transversely multi-distoseptate; apex subobtuse, base obconically truncate, with hila having a distinct central brown pore, thickened and darkened (Description obtained from Amaradasa et al. 2014).

*Notes:* *Porocercospora* was introduced by Amaradasa et al. (2014) to accommodate *Cercospora seminalis* which causes false smut in buffalograss (*Buchloe dactyloides*). The genus is characterised by densely aggregated and repeatedly branched conidiophores arising from a brown stroma, monotretic conidiogenous cells with inconspicuous loci, and scolecosporous conidia with distosepta, and thickened, darkened hila. DNA sequence data in the present study as well as in the previous studies (Amaradasa et al. 2014) indicated that *Porocercospora seminalis* is phylogenetically close to but distinct from the genera *Bipolaris* and *Curvularia*. In our phylogeny the type strain of *Porocercospora seminalis* (CPC 21332) forms a distinct clade sister to *Johnalcornia*. Therefore we accept the placement of *Porocercospora* as separate genus in *Pleosporaceae*.

***Pseudoyuconia*** Lar.N. Vassiljeva, Nov. sist. Niz. Rast. 20: 71 (1983).

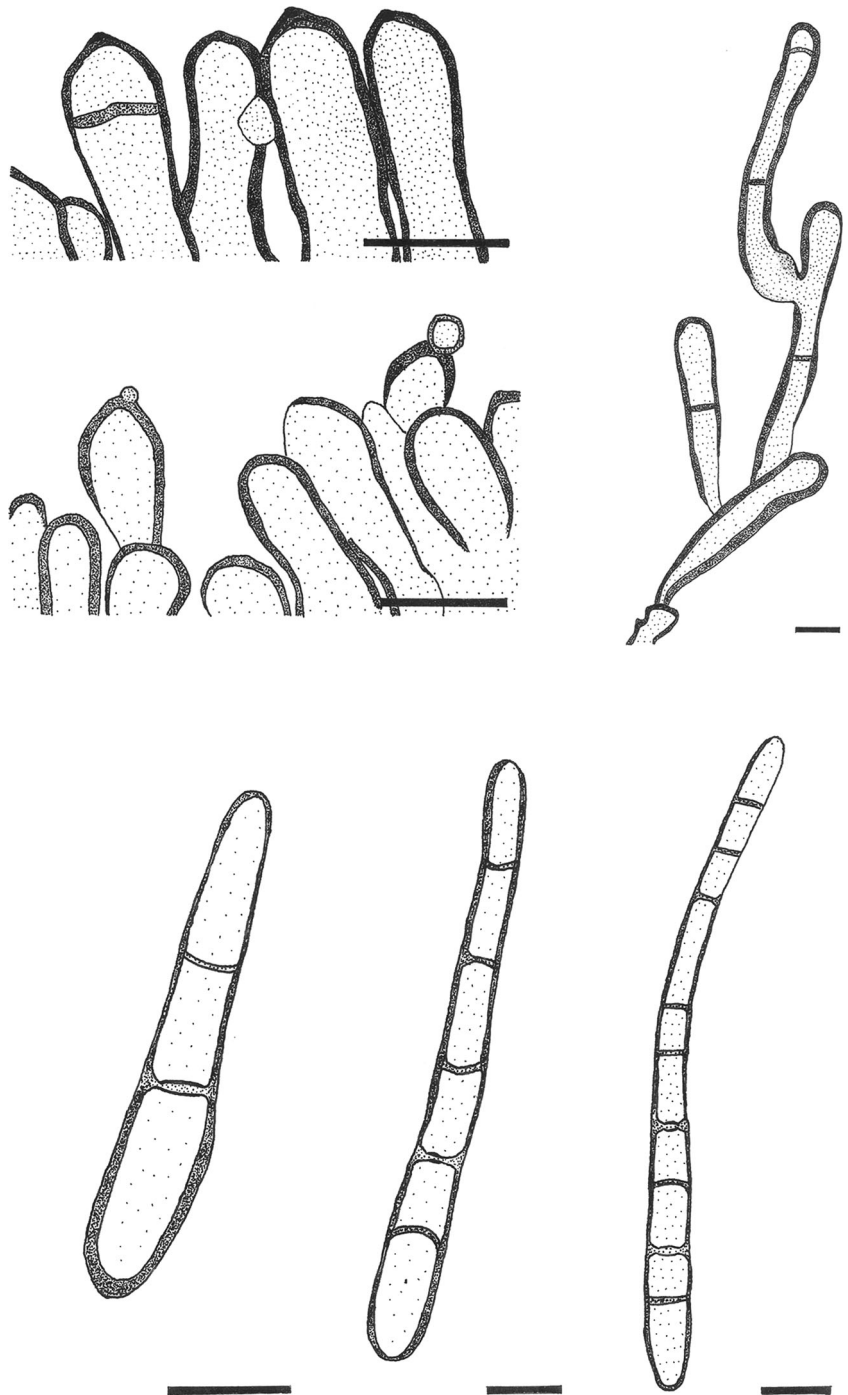
= *Barrella* Ahn & Shearer, Canadian Journal of Botany 73 (4): 578 (1995).

***Facesoffungi* number: FoF 00537**

*Saprobic* on dead wood. **Sexual morph:** *Ascomata* semi-immersed to superficial, globose to subglobose. *Ostiole* central when mature, ostiolar canal filled with periphyses. *Peridium* comprising of thick-walled cell, forming a *textura angularis*. *Hamathecium* comprising hyaline, cellular, septate



**Fig. 23** *Porocercospora seminalis* (Redrawn from Amaradasa et al. 2014, Fig. 2.). Scale bars: 5  $\mu$ m



pseudoparaphyses. *Asci* 8-spored, bitunicate, broadly cylindrical or clavate to obovoid, with a minute pedicel, apically rounded end, with an ocular chamber. *Ascospores* broadly ellipsoidal, pale brown to light brown, 2-septate. **Asexual morph:** undetermined

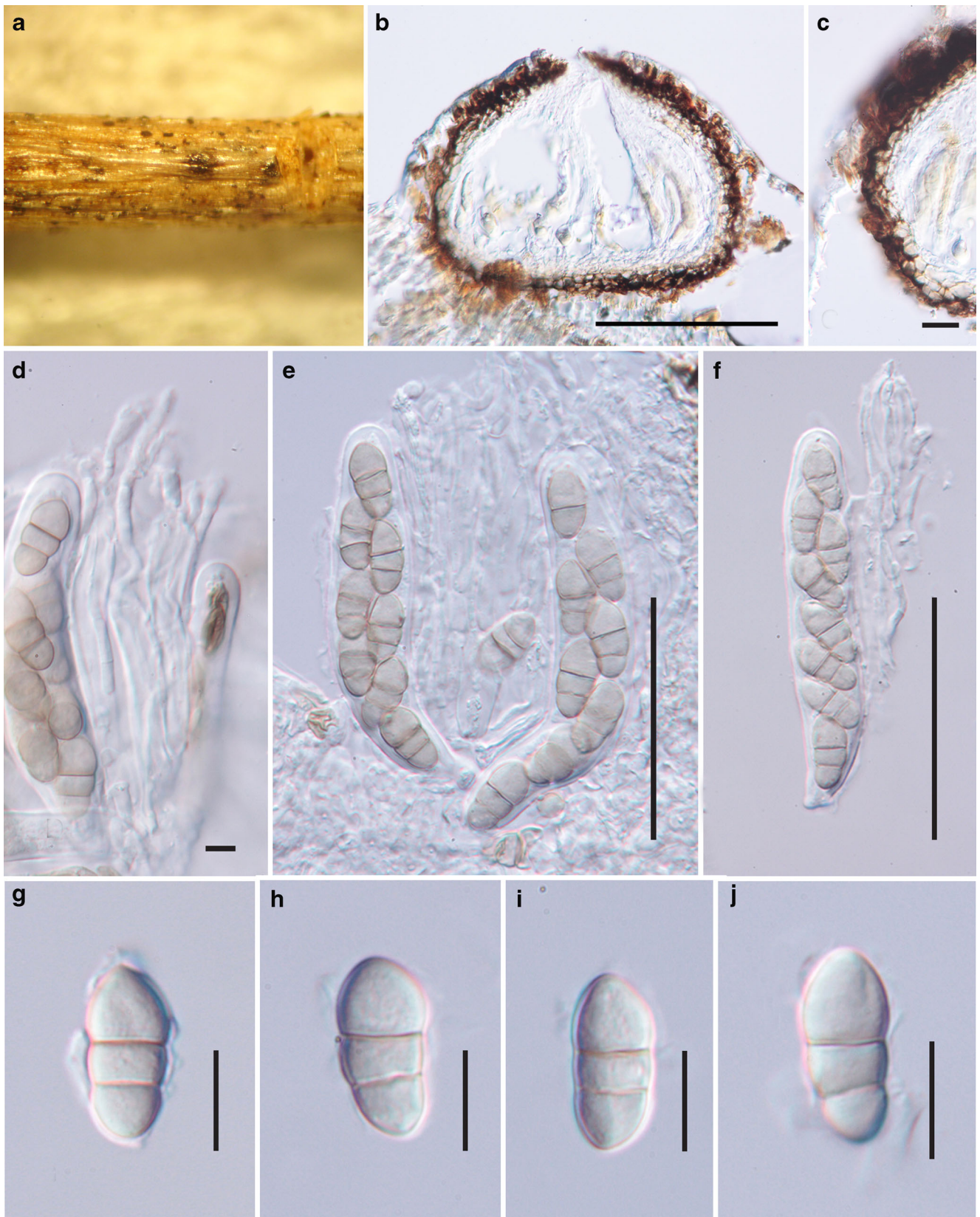
*Type species: Pseudoyuconia thalictri* (G. Winter) Lar.N. Vassiljeva [as ‘thalicti’], Nov. sist. Niz. Rast. 20: 71 (1983), Fig. 24

≡ *Leptosphaeria thalictri* G. Winter, Hedwigia 10: 40 (1872)

≡ *Metasphaeria thalictri* (G. Winter) Sacc., Sylloge Fungorum 2: 156 (1883)

≡ *Scleroplella thalictri* (G. Winter) Höhn., Annales Mycologici 18 (1–3): 76 (1920)

≡ *Buergenerula thalictri* (G. Winter) E. Müll., Sydowia 4 (1–6): 307 (1950)



**Fig. 24** *Pseudoyuconia thalictri* (VLA, P/187) **a.** Fungus on host substrate. **b.** Section through ascoma. **c.** Close up of the peridium. **d.** Pseudoparaphyses. **e, f.** Asci with eight ascospores. **g, j.** Ascospores with two septa. **Scale bars:** b=100  $\mu\text{m}$ , c, g–j=10  $\mu\text{m}$ , d=5  $\mu\text{m}$ , e–f=50  $\mu\text{m}$



≡ *Pseudoyuconia thalicti* (G. Winter) Lar.N. Vassiljeva (1983)

≡ *Barrella thalictri* (G. Winter) Ahn & Shearer, Can. J. Bot. 73(4): 578 (1995)

**Facesoffungi number: FoF 00538**

*Saprobic* on dead wood. **Sexual morph:** *Ascomata* 137–186×221–137 μm semi-immersed to superficial, globose to subglobose, black. *Ostiole* central present when mature, ostiolar canal filled with periphyses. *Peridium* 19–25 μm wide, 3–4 layered, comprising of thick-walled cell, forming a *textura angularis*. *Hamathecium* 2–3 μm wide, comprising hyaline, cellular, septate pseudoparaphyses anastomosing above the asci. *Asci* 80–119×17–23 μm ( $\bar{x}$ =97×20 μm,  $n$ =20), 8-spored, bitunicate, broadly cylindrical or clavate to obovoid, with a minute pedicel, apically rounded end, with an ocular chamber. *Ascospores* 21–25×9–12 μm ( $\bar{x}$ =23×11 μm,  $n$ =20), broadly ellipsoidal, pale brown to light brown, 2-septate, slightly constricted at each septum. **Asexual morph:** undetermined.

**Material examined:** RUSSIA, Magadan region, Magadan vicinity, Snow Valley, 8 July 1974, on *Thalictrum contortum* L. (*Ranunculaceae*), L. Vasilyeva (VLA P/187).

*Notes:* *Pseudoyuconia*, a monotypic genus, was introduced by Vassiljeva (1983) to accommodate *Leptosphaeria thalictri* G. Winter, and was typified by *P. thalictri*. Currently, *Pseudoyuconia* is included in *Pleosporaceae* (Lumbsch and Huhndorf 2010). No molecular data is available for this genus. Therefore, fresh collections of the type species of the genus are needed so that molecular data can be obtained to verify the natural taxonomic affinities of this genus. Based on the similarities with the other genera of the family *Pleosporaceae*, such as, in having globose to subglobose ascomata with central ostiole filled with periphyses, peridium composed cells of *textura angularis* we tentatively place *Pseudoyuconia* in *Pleosporaceae*.

In this study we could not locate the type material therefore our understanding is based on the collection of Vasilyeva (1983), which was observed by Vasilyeva (1983) when she was introduced this genus.

*Pyrenophora* Fr., Summa veg. Scand., Section Post. (Stockholm): 397 (1849).

= *Drechslera* S. Ito, Proc. Imp. Acad. Japan 6: 355 (1930).

**Facesoffungi number: FoF 00009**

*Saprobic* or *pathogenic* on leaves and stems. **Sexual morph:** *Ascomata* immersed, becoming erumpent to near superficial, solitary or scattered, globose to subglobose, broadly or narrowly conical, smooth-walled, ostiolate. *Ostiole* papillate, covered with brown to reddish-brown setae, which are darkened at the base. *Peridium* comprising 2–4 layers of brown,

thick-walled cells of *textura angularis*. *Pseudoparaphyses* not observed. *Asci* 8-spored, bitunicate, fissitunicate, clavate to sub-cylindrical, with a short, broad pedicel, with a distinct ocular chamber surrounded by a large apical ring. *Ascospores* 2–3-seriate, muriform, constricted at the septum, smooth-walled, surrounded by a mucilaginous sheath. **Asexual morph:** hyphomycetous *Conidiophores* macronematous, mononematous, sometimes caespitose, straight or flexuous, often geniculate, unbranched or in a few species loosely branched, brown, smooth in most species. *Conidiogenous cells* polytretic, integrated, terminal, frequently becoming intercalary, sympodial, cylindrical, cicatrized. *Conidia* solitary, in certain species also sometimes catenate or forming secondary conidiophores which bear conidia, acropleurogenous, simple, straight or curved, clavate, cylindrical, rounded at the ends, ellipsoidal, fusiform or obclavate, straw-coloured or pale to dark brown or olivaceous brown, sometimes with cells unequally coloured, the end cells then being paler than intermediate ones, mostly smooth, rarely verruculose, pseudoseptate (description of asexual morph obtained from Ellis 1971).

*Type species: Pyrenophora phaeocomes* (Rebent.) Fr., Summa veg. Scand., Section Post. (Stockholm): 397 (1849), Fig. 25

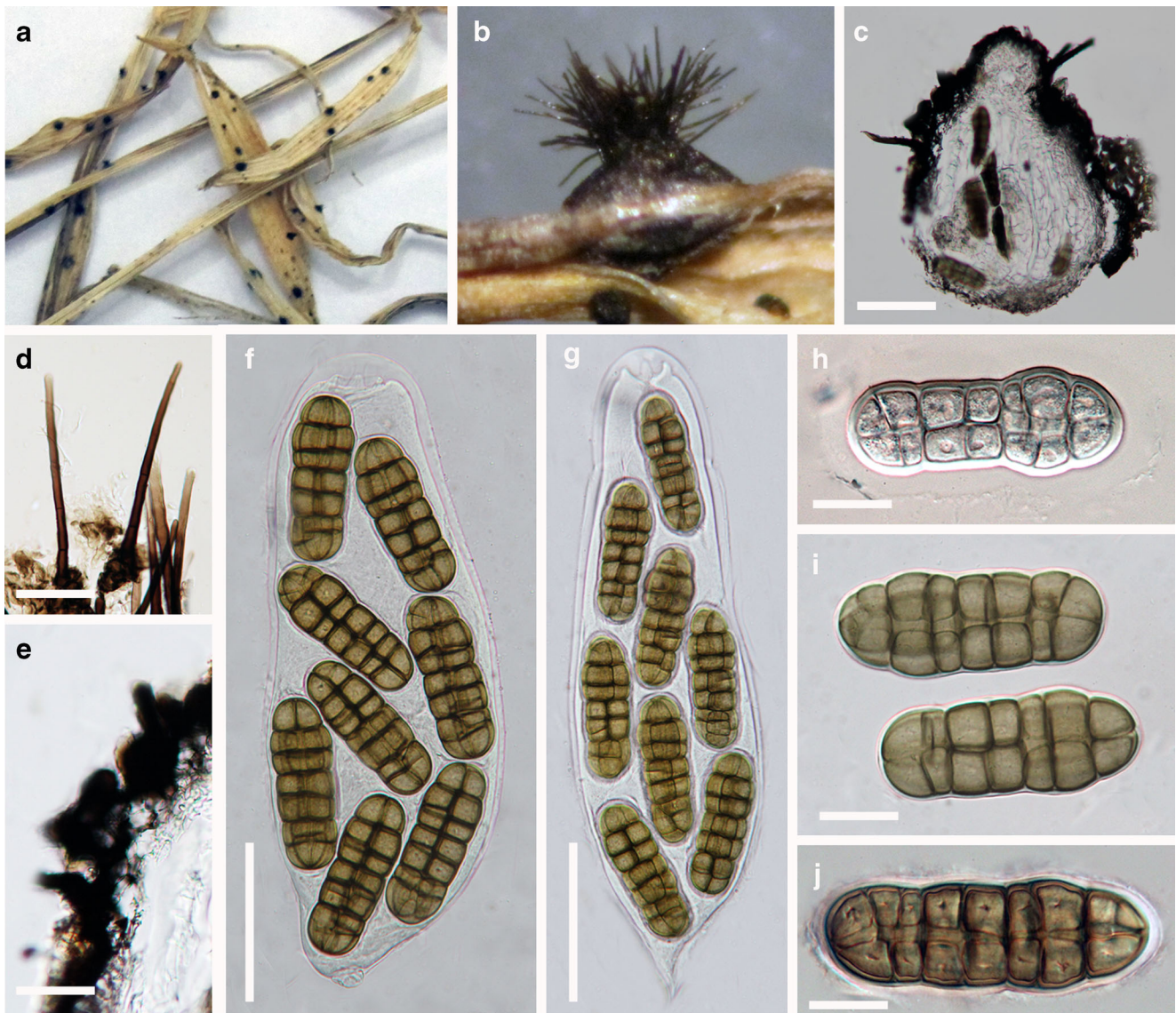
Basionym: *Sphaeria phaeocomes* Rebent., Prodr. fl. neomarch. (Berolini): 338 (1804)

≡ *Ceuthospora phaeocomes* (Rebent.) Fr., J. Taihoku Soc. Agric.: 119 (1825)

≡ *Pleospora phaeocomes* (Rebent.) Fr., Summa veg. Scand., Section Post. (Stockholm): 398 (1849).

**Facesoffungi number: FoF 00010**

*Saprobic* on dead leaves. **Sexual morph:** *Ascomata* 380–450×370–430 μm ( $\bar{x}$ =395×380 μm,  $n$ =10), solitary or scattered, initially immersed, becoming erumpent to near superficial, globose to subglobose, broadly or narrowly conical, coriaceous, smooth-walled, ostiolate. *Ostiole* usually broadly papillate, central ostiolar canal filled with periphyses and covered with setae. *Setae* brown to reddish brown, darkened at the base, septate and tapered towards the apex. *Peridium* 40–70 μm ( $\bar{x}$ =45 μm,  $n$ =20) wide, comprising two types of cells, outer cells of 1–2 layers of heavily pigmented cells of *textura angularis*, inner layer composed of small, light brown to hyaline cells of *textura angularis*. *Pseudoparaphyses* not observed. *Asci* 300–400×130–160 μm ( $\bar{x}$ =345×140 μm,  $n$ =20), 8-spored, bitunicate, fissitunicate, clavate to subcylindrical, with a short, broad pedicel, thickened and rounded at apex with a distinct ocular chamber surrounded by a large, distinct, apical ring. *Ascospores* 78–96×27–34 μm ( $\bar{x}$ =88×30 μm,  $n$ =40), biseriate to overlapping triseriate, ellipsoidal with broadly rounded ends, hyaline to light brown when immature, becoming brown to chestnut



**Fig. 25** *Pyrenophora phaeocomes* (neotype) **a.** Fungus on host substrate. **b.** Ascomata on substrate. **c.** Section through ascoma. **d.** Brown setae. **e.** Peridium. **f, g.** Asci with a distinct ocular chamber

surrounded by a large, distinct, apical ring. **h–j.** Immature and mature ascospores. **Scale bars:** c=100  $\mu\text{m}$ , d–e=50  $\mu\text{m}$ , f–g=40  $\mu\text{m}$ , h–j=10  $\mu\text{m}$

brown when mature, muriform with 5–6 transverse septa, and single longitudinal septa in one or all cells, constricted at the septa, smooth-walled, relatively thick-walled, with a 5–9  $\mu\text{m}$  thick mucilaginous sheath. **Asexual morph of *Drechslera teres*** (Fig. 26); *Conidiophores* 150–200 $\times$ 7–11  $\mu\text{m}$ , solitary or in groups of 2–3, straight or flexuous, sometimes geniculate, often swollen at the base, pale to mid brown or olivaceous brown. *Conidia* 70–160 $\times$ 16–23  $\mu\text{m}$ , straight, cylindrical, rounded at the ends, subhyaline to straw-coloured, smooth, 1–10 (4–6), pseudoseptate, frequently with constriction.

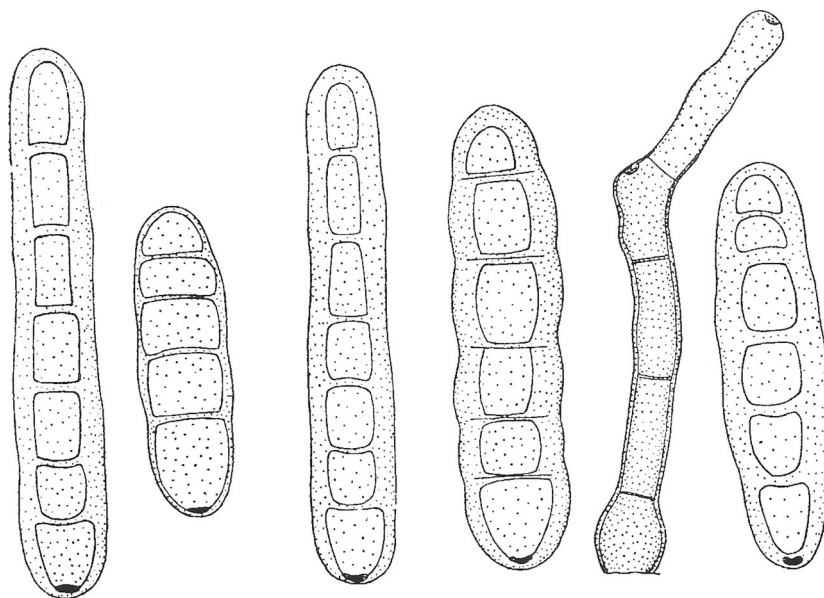
**Notes:** The asexual morph of *Pyrenophora teres*, *Drechslera teres* is described and illustrated here in Fig. 25 because in original description of *P. phaeocomes* only mentored that it has *Drechslera*-like asexual morph without giving a species name (Sivanasen 1984).

**Material examined:** SWEDEN, on leaves of *Anthoxanthum* (*Poaceae*), 7 August 1951, J. Ax. Nannfeldt (UPS 170980, **neotype**).

**Notes:** Based on both morphology and molecular data, *Pyrenophora* has been assigned to *Pleosporaceae* (Zhang et al. 2012). The genus is characterized by immersed, erumpent to nearly superficial ascomata, asci usually with a large apical ring with a clear ocular chamber, lack of pseudoparaphyses and muriform, terete ascospores (Sivanasen 1984). Currently 199 species of *Pyrenophora* are listed in Index Fungorum (2015). Over 3000 hits are found in GenBank including the putative strain of *Pyrenophora phaeocomes* (DAOM 222769), which is the type species for this genus. In the present study the genus *Pyrenophora* clusters in the suborder *Pleosporineae* of the family *Pleosporaceae* with a relatively high bootstrap support



**Fig. 26 Morphology of *Drechslera teres*** (Redrawn from Ellis 1971, Fig 299.). Scale bars: 50  $\mu\text{m}$



(Fig. 1, 60 %). Further phylogenetic analysis shows that sexual *Pyrenophora* morphs cluster with asexual *Drechslera* morphs, i.e. *Pyrenophora dictyoides* (DAOM 75616) clusters with *Drechslera dictyoides* (DAOM 63666). The putative strain of *Pyrenophora phaeocomes* (DAOM 222769), which is the type species of the genus, clusters with other *Pyrenophora* species.

As a genus can now only have one name, Ariyawansa et al. (2014c) proposed to synonymise *Drechslera* under *Pyrenophora* by giving the priority for the oldest name. This was followed by Wijayawardene et al. (2014).

#### Excluded genera

***Austropleospora*** R.G. Shivas & L. Morin, in Morin et al., Fungal Diversity 40(1): 70 (2010).

**Facesoffungi number: FoF 00539**

**Type species: *Austropleospora osteospermi*** R.G. Shivas & L. Morin, Morin et al., Fungal Diversity 40(1): 70 (2010), Fig. 27  
**Facesoffungi number: FoF 00540**

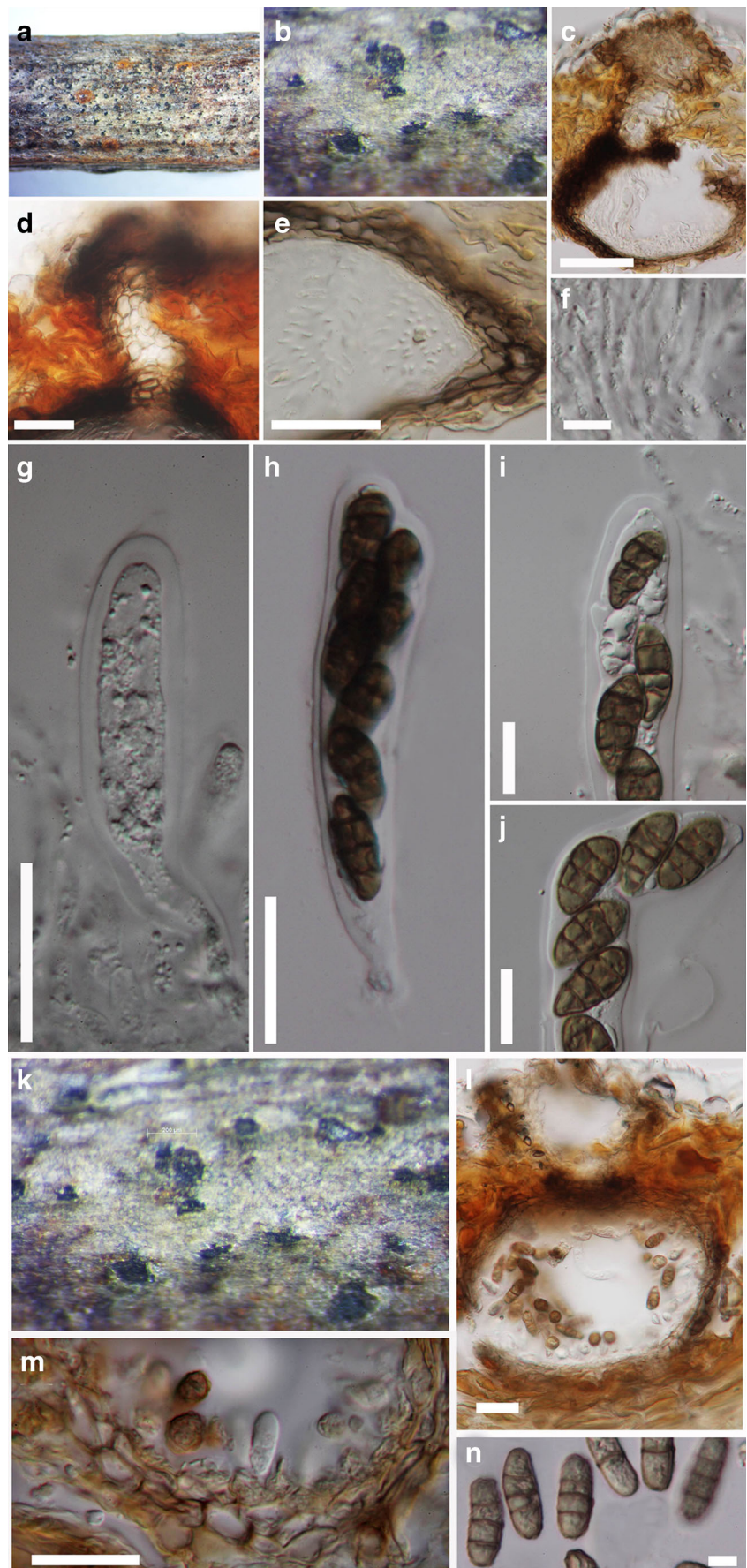
Parasitic on stem and leaves of *Chrysanthemoides monilifera* (Asteraceae). **Sexual morph:** *Ascomata* 75–110  $\times$  130–200  $\mu\text{m}$  ( $x=100 \times 146 \mu\text{m}$ ,  $n=10$ ), subglobose, sometimes slightly flattened, solitary or in groups, scattered, immersed immediately below the stem epidermis, ostiole 60–90  $\mu\text{m}$  long, with a protruding neck. *Peridium* 8–18  $\mu\text{m}$  ( $x=12 \mu\text{m}$ ,  $n=10$ ) wide, composed of dark brown to black cells of *textura angularis*. *Hamathecium* of 2–3  $\mu\text{m}$  wide, dense, filamentous, anastomosing, aseptate, hyaline pseudoparaphyses. *Asci* 75–120  $\times$  13–18  $\mu\text{m}$  ( $x=92 \times 16 \mu\text{m}$ ,  $n=20$ ), bitunicate, fissitunicate, 6–8-spored, cylindrical to clavate, rounded at

the apex and minute ocular chamber with a short, broad, pedicel. *Ascospores* 16.5–21  $\times$  6–8.4  $\mu\text{m}$  ( $x=18 \times 7.7 \mu\text{m}$ ,  $n=25$ ), biseriate to overlapping uniseriate, ellipsoidal, yellowish brown, muriform, mostly with 3-transverse septa, 0–2 longitudinal septa, slightly constricted at median septum, not or very slightly constricted at other septa, apex rounded to slightly tapered, base tapered to rounded, smooth. **Asexual morph:** *Conidiomata* 75–110  $\times$  100–130  $\mu\text{m}$  ( $x=88 \times 115 \mu\text{m}$ ,  $n=10$ ), pycnidial, globose, superficial on stem, immersed in the host tissue and becoming erumpent at maturity, globose, dark brown in the erumpent part, with a single ostiole. *Conidiomata wall* 9–16  $\mu\text{m}$  wide, brown to reddish-brown, thin-walled, comprising several layers with cells of *textura angularis*. *Conidiophores* reduced to conidiogenous cells. *Conidiogenous cells* 10–12  $\times$  2.5–3.5  $\mu\text{m}$  ( $x=11 \times 2 \mu\text{m}$ ,  $n=15$ ), inconspicuously annellidic, discrete, cylindrical. *Conidia* 14–18  $\times$  4.8–6.5  $\mu\text{m}$  ( $x=15.8 \times 5.7 \mu\text{m}$ ,  $n=25$ ), cylindrical to narrowly ellipsoidal, initially hyaline and aseptate, becoming yellowish-brown at maturity, mostly transversely 1–3-septate, ends rounded.

**Material examined:** AUSTRALIA, NSW Bellinger Head, Kalang river mouth, on stem and leaves of *Chrysanthemoides monilifera* ssp. *rotundata* (L.) Norlindh (Asteraceae). 5 March 2008, L. Morin (BRIP 52234, **holotype**).

**Notes:** *Austropleospora* was introduced by Morin et al. 2010 to accommodate *A. osteospermi* on *Chrysanthemoides monilifera* ssp. *rotundata* (Asteraceae) and it is monotypic. In the same study Shivas and Morin (in Morin et al. 2010) observed *Hendersonia osteospermi* Wakef. on the same host and identified it as the asexual morph of *A. osteospermi* both in culture and by DNA sequence analysis. Based on ITS sequence analysis, Morin et al. (2010) placed *Austropleospora*

**Fig. 27** *Austropleospora osteospermi* (holotype) **a.** Herbarium material. **b, c.** Ascomata on host surface. **d.** Section through ascoma. **e.** Section through ostiole. **f.** Peridium. **g.** Pseudoparaphyses **h.** Immature ascus. **i.** Mature bitunicate ascus. **j.** Apex of ascus. **k.** Ascospores. **k.** Pycnidia on host. **l.** Section of pycnidium. **m.** Pycnidial wall. **o.** Conidiogenous cells and developing conidia. **n.** Brown 3-septate conidia. **Scale bars:**  $d=50\ \mu\text{m}$ ,  $e-f=25\ \mu\text{m}$ ,  $g=10\ \mu\text{m}$ ,  $h-i=25\ \mu\text{m}$ ,  $k=15\ \mu\text{m}$ ,  $l, m=20\ \mu\text{m}$ ,  $n=5\ \mu\text{m}$





under *Pleosporales* without assigning it to any family. *Austropleospora* is classified in *Dothideomycetes* genera *incertae sedis* in Index Fungorum (2015). After re-examining the holotype of *A. osteospermi*, Thambugala et al. (2014b) tentatively referred *Austropleospora* in *Pleosporaceae* based on morphological similarities. During our preliminary phylogenetic analysis (data not shown), we observed that the type strain of *A. osteospermi* (BRIP 51628) forms a distinct clade in family *Didymosphaeriaceae*. Therefore we exclude *Austropleospora* from *Pleosporaceae* and refer it to *Didymosphaeriaceae*.

***Dendryphion*** Wallr., Fl. crypt. Germ. (Norimbergae) 2: 300 (1833)

*Dendryphiella* Bubák & Ranoj., in Ranojevic, Anns mycol. 12(4): 417 (1914)

*Dwayamala* Subram., J. Indian bot. Soc. 35: 475 (1956)

*Entomyclium* Wallr., Fl. crypt. Germ. (Norimbergae) 2: 189 (1833)

**Facesoffungi number: FoF 00541**

*Type species: Dendryphion comosum* Wallr., Fl. crypt. Germ. (Norimbergae) 2: 300 (1833)

= *Dendryphion curtum* Berk. & Broome, Ann. Mag. nat. Hist., Ser. 2 7: 176 (1851)

= *Dendryphion curtum* Berk. & Broome, Ann. Mag. nat. Hist., Ser. 2 7: 176 (1851) var. *curtum*

= *Dendryphion curtum* var. *ramosius* Sacc., Syll. fung. (Abellini) 4: 490 (1886)

**Facesoffungi number: FoF 00542**

*Notes: Dendryphion* was introduced by Wallroth (1833) and is typified by *Dendryphion comosum*. The genus *Dendryphion* is characterised by having apically branched conidiophores with polytretic dark scars, and chains of brown, septate (didymo or cheiro) conidia (Crous et al. 2014). The genus *Brachycladium* had been synonymised with *Dendryphion* by Ellis (1971), was resurrected for the asexual morph based on polyphyly within *Dendryphion* and morphological distinction from type species, *D. comosum* (Crous et al. 2014). The type species of *Brachycladium*, *B. penicillatum*, resides in *Alternaria* sect. *Crivellia* in this study as well as in Woudenberg et al. (2013). *Brachycladium* is therefore a synonymy of *Alternaria* instead of *Dendryphion*. Although there are more than 80 names recorded in Index Fungorum (2015), Seifert et al. (2011) confined them to six species. Inderbitzin et al. (2006) showed that *Dendryphion* was polyphyletic thus the natural classification of the genus has become confused. Currently, no sequence data is available in GenBank for the generic type of *Dendryphion*. Recently, Crous et al. (2014) introduced a new species of *Dendryphion*, *D. europaeum* with molecular and morphological data, and placed it species in the suborder *Pleosporineae*, family *Pleosporaceae*.

Considering the morphological key provided by Siboe et al. (1999), Crous et al. (2015) confirmed that *D. europaeum* appears to be distinct from the currently recognised *Dendryphion* species based on its conidial morphology. During our preliminary analysis we observed that *D. europaeum* forms a robust clade outside the suborder *Pleosporineae*. Therefore we tentatively exclude *Dendryphion* from *Pleosporaceae* and place it in *Pleosporales*, genera *incertae sedis*. However, the placement of *Dendryphion* in *Pleosporales* can only be confirmed by phylogenetic analysis, including sequencing the generic type of *Dendryphion* (*D. europaeum*).

***Edenia*** M.C. González et al., in González et al., Mycotaxon 101: 254 (2007)

*Type species: Edenia gomezpompae* M.C. González et al., in González et al., Mycotaxon 101: 254 (2007)

**Facesoffungi number: FoF 00543**

*Notes: Edenia* was introduced by González et al. (2007) and is typified by *Edenia gomezpompae*. This endophytic fungus was isolated from living leaves of *Callicarpa acuminta* Kunth (González et al. 2007). González et al. (2007) placed *Edenia* in *Pleosporaceae* based on the ITS sequence data. In our phylogeny we observed that the type strain of *Edenia gomezpompae* (C1c) along with putative strains JLCC 34533, CBS 124106 and 11G048 form a robust clade in family *Phaeosphaeriaceae* outside *Pleosporaceae*. Therefore we exclude *Edenia* from *Pleosporaceae* and placed it in the *Phaeosphaeriaceae*.

***Kriegeriella*** Höhn., Anns mycol. 16(1/2): 39 (1918).

*Type species: Kriegeriella mirabilis* Höhn., Anns mycol. 16(1/2): 39 (1918).

**Facesoffungi number: FoF 00544**

*Notes: Kriegeriella* was introduced by von Höhnel (1918b) in order to accommodate *K. mirabilis* and *K. transiens* Höhn. The genus *Kriegeriella* was typified with *K. mirabilis* and referred to *Microthyriaceae* by Höhnel 1918b. It is characterised by superficial, subglobose, black thyrothecial-like ascomata, cellular pseudoparaphyses, bitunicate, obpyriform asci bearing hyaline, multi-septate ascospores which turn brown when mature. Subsequently, *Kriegeriella* was assigned to the subfamily of *Aulographiodeae* (*Microthyriaceae*) (Batista et al. 1959), *Asterinaceae* (*Hemisphaeriales*) (Luttrell 1973) and *Pseudosphaeriaceae* (*Dothideales*) (Barr 1975). Barr (1987b) introduced a family, i.e. *Kriegeriellaceae* (*Dothideales*) to accommodate *Kriegeriella* and *Extrawettsteinina*. This scheme is rarely followed, and *Kriegeriella* is generally classified under *Pleosporaceae* (*Pleosporales*) (Eriksson 2006; Lumbsch and Huhndorf 2007, 2010).

Zhang et al. (2012) excluded *Kriegeriella* from the family *Pleosporaceae* and concluded that it is not a typical genus of the family. Furthermore, Zhang et al. (2012) suggested that *Kriegeriella* might belong to *Microthyriaceae*, as it would be unusual in *Pleosporaceae* in having thyrothecium-like ascomata and 5–6-septate ascospores. Therefore we exclude *Kriegeriella* from *Pleosporaceae*, following the opinion of Zhang et al. (2012).

**Macrospora** Fuckel, Jb. nassau. Ver. Naturk. 23–24: 139 (1870) [1869–70].

= *Nimbya* E.G. Simmons, Sydowia 41: 316 (1989)

**Facesoffungi number: FoF 00545**

*Saprobic* on stems or wood. **Sexual morph:** *Ascomata* immersed to nearly superficial, subglobose. *Ostiole* central, present when mature, ostiolar canal filled with periphyses. *Peridium* comprising thick-walled cells, forming a *textura angularis*. *Hamathecium* comprising hyaline, unbranched, septate, pseudoparaphyses. *Asci* 8-spored, bitunicate, broadly cylindrical or clavate to obovoid, with a knob-like pedicel at the base, apically rounded, with an ocular chamber. *Ascospores* broadly ellipsoidal, brown to dark brown, multi septate. **Asexual morph:** undetermined.

*Type species: Macrospora scirpicola* (DC.) Fuckel, Jb. nassau. Ver. Naturk. 23–24: 139 (1870) [1869–70], Fig. 28

Basionym: *Sphaeria scirpicola* DC., in Lamarck & de Candolle, Fl. franç., Edn 3 (Paris) 2: 300 (1805)

≡ *Clathrospora scirpicola* (DC.) Höhn., Anns mycol. 18(1/3): 77 (1920)

≡ *Macrospora scirpicola* (DC.) Fuckel, Jb. nassau. Ver. Naturk. 23–24: 139 (1870)

≡ *Pleospora scirpicola* (DC.) P. Karst., Bidr. Känn. Finl. Nat. Folk 23: 72 (1873)

= *Pleospora scirpicola* var. *scirpicola* (DC.) P. Karst., Bidr. Känn. Finl. Nat. Folk 23: 72 (1873)

≡ *Pyrenophora scirpicola* (DC.) E. Müll., Sydowia 5(3–6): 256 (1951)

= *Sphaeria culmorum* Wallr., Fl. crypt. Germ. (Norimbergae) 2: 770 (1833)

**Facesoffungi number: FoF 00546**

*Saprobic* on dead stems. **Sexual morph:** *Ascomata* 83–101 × 72–79 μm, immersed in host tissues, subglobose, brown to dark brown, ostiolate. *Ostiole* central when mature, ostiolar canal 31–36 μm wide at the base, with periphyses. *Peridium* 9–25 μm wide, comprising 2–3 layers of thick-walled cells, forming a *textura angularis*, brown to dark brown at outside and hyaline at the inner layer. *Hamathecium* of 2–4 μm wide, thick-walled, cellular, hyaline, unbranched, septate, pseudoparaphyses. *Asci* (26–) 64–88 × 9–13 μm, 8-spored, bitunicate, broadly cylindrical or clavate to obovoid, with a

knob-like pedicel at the base, apically rounded, with an ocular chamber. *Ascospores* 16–28 × 8–12 μm., 2–4 overlapping seriate, broadly ellipsoidal, ends rounded, straight or curved, brown to dark brown, 3-septate, constricted at the central cell, dark brown at the septum, second cell from above widest. **Asexual morph:** undetermined.

**Material examined:** FRANCE, collector unspecified, Exsiccati title: Plantes Cryptogames De France, Exsiccati Number: 1778 (NY 0098598, **holotype**).

*Notes:* *Macrospora* had been assigned to *Diademaceae* based on its applanate and muriform ascospores with 1-row of longitudinal septa, with a sheath, 2–3 μm wide and constricted at the first septum and ascospores that are paler and larger than those of *Comoclathris* (Shoemaker and Babcock 1992). *Macrospora* was however, considered as a synonym of *Pyrenophora* by Eriksson and Hawksworth (1991) which was assigned to *Pleosporaceae*, and this proposal has been widely followed (Eriksson 2006; Lumbsch and Huhndorf 2010). Ariyawansa et al. (2014a) suggested that *Macrospora scirpicola* is neither typical of *Diademaceae* nor *Pleosporaceae*. *Macrospora* is compatible with *Thyridaria* in having immersed, subglobose ascomata with 2–3 layers, of thick-walled cells of *textura angularis* and cylindrical asci bearing phragmospores, but differs in having cellular pseudoparaphyses and asci with a knob-like or furcate pedicel.

Currently no molecular data is available for any *Macrospora* species in GenBank. Therefore based on its similarities with *Thyridaria*, we tentatively place *Macrospora* in *Thyridariaceae*, but fresh collections of the type species of the genus are needed so that molecular data can be obtained to verify its natural taxonomic affinities.

**Monascostroma** Höhn., Anns mycol. 16(1/2): 160 (1918).

**Facesoffungi number: FoF 00547**

*Type species: Monascostroma innumerosum* (Desm.) Höhn. [as ‘innumerosa’], Anns mycol. 16(1/2): 160 (1918).

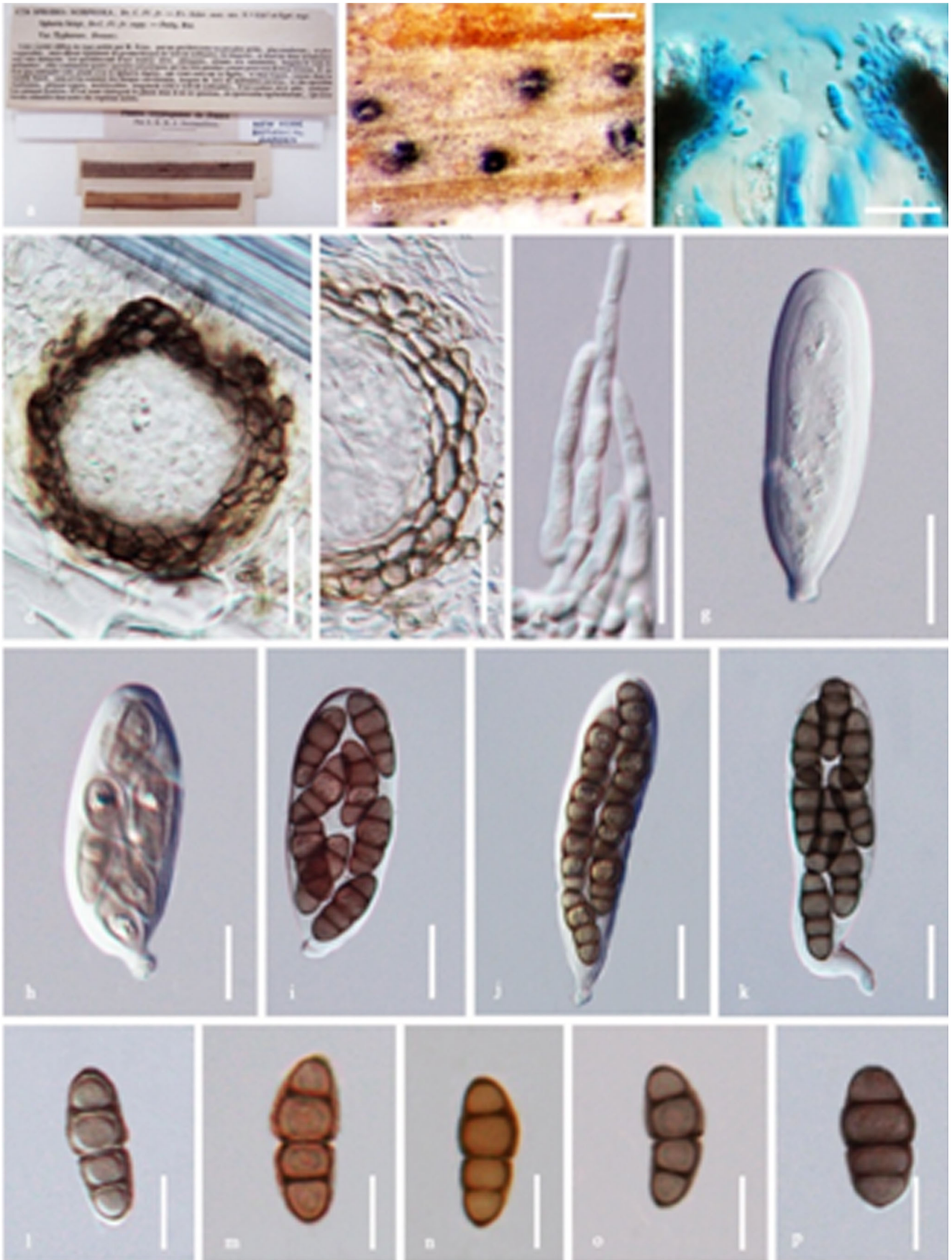
Basionym: *Hendersonia innumerosa* Desm., Anns Sci. Nat., Bot., sér. 3 16: 10 [repr.] (1851)

≡ *Stagonospora innumerosa* (Desm.) Sacc., Sylloge Fungorum 3: 451 (1884)

**Facesoffungi number: FoF 00548**

*Notes:* *Monascostroma* was introduced by Desmazières (1851) to accommodate *M. innumerosum* (= *Hendersonia innumerosa*). Eriksson and Hawksworth (1998) and Kodsueb et al. (2006) included *Monascostroma* in *Pleosporaceae*, while later researchers classified *Monascostroma* in *Didymellaceae* (Zhang et al. 2009, 2012; Schoch et al. 2009; Lumbsch and Huhndorf 2010).





The putative strain of *Monascostroma innumerosum* (CBS 345.50) forms a robust clade in family *Didymellaceae* (Zhang et al. 2009, 2012; Schoch et al. 2009; Hyde et al. 2013). Thus we also accept the genus *Monascostroma* in the *Didymellaceae* following other published work (Zhang et al. 2009, 2012; Schoch et al. 2009; Hyde et al. 2013).

***Zeuctomorpha*** Sivan et al., in Sivanesan, Bitunicate Ascomycetes and their Anamorphs (Vaduz): 572 (1984).

***Facesoffungi* number: FoF 00549**

*Type species: Zeuctomorpha arecae* Sivan et al., in Sivanesan, Bitunicate Ascomycetes and their Anamorphs (Vaduz): 572 (1984).

***Facesoffungi* number: FoF 00550**

*Notes: Zeuctomorpha*, a monotypic genus typified by *Z. arecae*, was formally treated as a member of the *Pleosporaceae* by Sivanesan (1984), based on its superficial setose ascomata, clavate, asci, ellipsoid and 1-septate ascospores, and presence of pseudoparaphyses. Eriksson and Hawksworth (1998) classified *Zeuctomorpha* in *Pleosporaceae* and this was followed by Kirk et al. (2001), Kodsueb et al. (2006) and Lumbsch and Huhndorf (2010).

Zhang et al. (2012) suggested that *Zeuctomorpha* was unusual amongst the *Pleosporaceae* with its hairy superficial ascomata, few pseudoparaphyses, broadly clavate to obclavate asci and 1-septate pigmented ascospores. Furthermore, Zhang et al. (2012) concluded that all of these morphological characters are most comparable with species of *Acantharia*, which might be closely related to *Venturiaceae*, but this needs to be confirmed by molecular data. Therefore, based on the morphology we also recommend to exclude *Zeuctomorpha* from *Pleosporaceae* without assigning it to any family.

**Concluding remarks**

Molecular data coupled with morphology has provided the basis for the modern classification of the kingdom fungi, but have some restrictions in application (Hibbett et al. 2007; Ariyawansa et al. 2014a; Boonmee et al. 2014; Hyde et al. 2014; Nilsson et al. 2014; Schoch et al. 2014; Thambugala et al. 2014a). The most important and disconcerted problem is that the phylogeny inferred from any gene may not divulge the evolution history of the organism (Uilenberg et al. 2004;

Ariyawansa et al. 2014a). Therefore polyphasic taxonomy, including genotypical and phenotypical characteristics are recommended in all future studies (Uilenberg et al. 2004; Ariyawansa et al. 2014a, Udayanga et al. 2014a). The genome also needs to be evaluated (Uilenberg et al. 2004; Ariyawansa et al. 2014a). Another main problem is most early studies is that they focused on techniques and used strains of fungi that in most cases were not carefully referenced thus creating confusion as explained in Ariyawansa et al. (2014a). Most strains in GenBank are named without attached voucher material and it impractical to confirm their characters to ensure correct identification.

There have only been a few molecular analysis of *Pleosporaceae* as compared to morphological studies (Kodsueb et al. 2006). Genera with bitunicate asci and brown muriform ascospores viz *Murispora*, *Pleospora*, *Pyrenophora* and *Tremateia* were generally classified under *Pleosporaceae*. Molecular studies have shown that these particular morphological characters have evolved in different families such as *Aigialaceae*, *Amniculicolaceae*, *Didymellaceae*, *Lophiostomataceae*, *Montagnulaceae* and *Sporormiaceae* (Zhang et al. 2012a, Hyde et al. 2013). Recently published papers such as Zhang et al. (2012a), Woudenberg et al. (2013) and Hyde et al. (2013), did not discuss details of the family *Pleosporaceae* and related genera. Woudenberg et al. (2013) was mainly focused on the genus *Alternaria*, while Zhang et al., (2012a) gave a detail account of sexual morphs only. Hyde et al. (2013) provided a key to the genera in *Pleosporaceae* in work on Dothideomycetes.

Fourteen clades of *Pleosporaceae* have been identified by our analysis based on ITS, LSU, SSU and RPB2 gene data, which are treated as suitable for the delimitation of genera (Figure 5.1). Although relationships among these clades may be weakly supported and some may vary in detail, some tentative conclusions can be drawn. Several current taxonomic hypotheses are supported by our molecular data, and this makes it possible to propose some taxonomic hypotheses about the relationships among the genera in *Pleosporaceae*. By combining multi-locus DNA sequencing with detailed morphological analyses, we were able to define and formally propose two new species, four combinations and several sexual morphs among the taxa in the family *Pleosporaceae*. In our study we accept 16 sexual or asexual genera and exclude three genera from the family *Pleosporaceae* based on morphology coupled with molecular data.

In previous treatments *Pleospora* species were considered as paraphyletic within the family *Pleosporaceae* (Kodsueb et al. 2006, Inderbitzin et al. 2009). Several studies have confirmed this opinion and have led to the removal of *Pleospora* species to other genera (Crivelli 1983, Inderbitzin et al. 2002, 2006; Leuchtman 1984, Zhang et al. 2009b). In the present study we were able to resolve the paraphyletic nature of the genus by following Woudenberg et al. (2013) and

◀ **Fig. 28** *Macrospora scirpicola* (holotype) **a.** Herbarium material. **b.** Appearance of ascomata on host. **c.** Neck with periphyses. **d.** Vertical section through ascoma. **e.** Section of peridium. **f.** Pseudoparaphyses. **g, h.** Immature asci. **i–k.** Mature asci. **l.** Immature ascospore. **m–p.** Mature ascospores. **Scale bars:** b=500 µm, g–k=50 µm, d–e=30 µm, c=20 µm, f=20 µm, l–p=20 µm



Wijayawardene et al. (2014), where they have treated the genus based on the generic type *Pleospora herbarum* (CBS 191.86<sup>T</sup>). Therefore in order to create a stable phylogenetic taxonomy, several putative strains of *Pleospora* which clustered with the type strain of *Pleospora herbarum* (CBS 191.86) were classified under *P. herbarum sensu stricto* (*P. tomatonis* CBS109844 and *P. sedicola* CBS109843). The putative strains of *P. papaveracea* CBS432.50, *P. tarda* CBS 714.68 and *P. halophila* CBS410.73 were treated under the genus *Pleospora* as separate species but this needs to be clarified with morphology because currently these strains are not attached to any herbarium material.

Some putative strains of *Pleospora* species other than species discussed in the above paragraph were assigned to different genera in *Pleosporaceae* based on their molecular data. *Pleospora fallens* (Sacc.) Gruyter & Verkley was introduced by de Gruyter et al. (2013) in order to accommodate *Phoma fallens* Sacc., because during their study the strain of *Phoma fallens* (CBS 284.70) clustered within the family *Pleosporaceae*. Woudenberg et al. (2013) and Ariyawansa et al. (2014a) showed that this strain forms a separate clade in the family *Pleosporaceae* basal to the other genera of the family. Importantly *Pleospora fallens* clustered outside the *Pleospora sensu stricto*. Therefore here we propose to keep the name *Phoma fallens* instead of *Pleospora fallens*. By taking this approach we were able to resolve the paraphyletic nature of *Pleospora* within the family *Pleosporaceae*.

In recent years, DNA based studies revealed multiple non-monophyletic genera within the *Alternaria* complex, and *Alternaria* species clades that do not always correlate to species-groups based on morphological characteristics (Pryor and Bigelow 2003; Inderbitzin et al. 2006; Pryor et al. 2009; Runa et al. 2009; Wang et al. 2011; Lawrence et al. 2012; Woudenberg et al. 2013). A separate phylogeny was prepared to show the placement of the recently revised sections in *Alternaria* (Figure 5.2). The well-supported node for the *Alternaria* clade obtained in the present study, and the low bootstrap support at the deeper nodes within the *Alternaria* complex is similar to previous phylogenetic studies. (Pryor and Bigelow 2003; Inderbitzin et al. 2006; Pryor et al. 2009; Runa et al. 2009; Wang et al. 2011; Lawrence et al. 2012; Woudenberg et al. 2013). We also observed the same result as Woudenberg et al. (2013) where the *Alternaria* clade contains 24 internal clades and six monotypic lineages, the assemblage of which is recognised as *Alternaria*. Furthermore we introduced one new species (*A. sedi*) and several collections of the sexual morph of *A. alternata* from Italy are also introduced in the study.

The explanation of *Pleosporaceae* in the current work is sustained by a well-supported phylogenetic nodes in multiple analyses coupled with morphology. We pursue the precedence introduced by Lawrence et al. (2013) and Woudenberg et al. (2013) to assign the taxonomic status of sections of *Alternaria*

for the different clades found. In here also we show the significance of having accurate DNA sequence data attached with both correct taxonomic names and clearly annotated specimen data. For end-users, this also results in a more stable and understandable taxonomy and nomenclature. Furthermore we gave a well resolved back-bone tree for the family *Pleosporaceae*, representing all the sexual and asexual genera, which currently have DNA sequences.

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