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Article

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New asexual morph taxa in Phaeosphaeriaceae

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Abstract

Species of *Phaeosphaeriaceae*, especially the asexual taxa, are common plant pathogens that infect many important economic crops. Ten new asexual taxa (*Phaeosphaeriaceae*) were collected from terrestrial habitats in Italy and are introduced in this paper. In order to establish the phylogenetic placement of these taxa within *Phaeosphaeriaceae* we analyzed combined ITS and LSU sequence data from the new taxa, together with those from GenBank. Based on morphology and molecular data, *Poaceicola gen. nov.* is introduced to accommodate the new species *Po. arundinis* (type species), *Po. bromi* and *Po. elongata*. The new species *Parastagonospora dactylidis*, *P. minima*, *P. italica*, *P. uniseptata* and *P. allouniseptata*, *Septoriella allojunci* and *Wojnowicia spartii* are also introduced with illustrated accounts and compared with similar taxa. We also describe an asexual morph of a *Nodulosphaeria* species for the first time.

Key words – Asexual morphs – *Phaeosphaeriaceae* – Phylogeny – Taxonomy

Introduction

The order Pleosporales has been of great research interest in the recent years and has undergone considerable revision (Zhang et al. 2009, 2012, Hyde et al. 2013, Ariyawansa et al. 2014, 2015b). Hyde et al. (2013) provided a taxonomic account for 41 families in Pleosporales, in which *Phaeosphaeriaceae* is a large and important family, comprising more than 300 species, and has also undergone considerable revision (Hyde et al. 2013, Phookamsak et al. 2014, Liu et al. 2015). Species in *Phaeosphaeriaceae* can be saprobic or endophytic, but many are pathogenic on economically

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important plants and crops (Kirk et al. 2008, Zhang et al. 2009, Quaedvlieg et al. 2013, Hyde et al. 2013, Wijayawardene et al. 2014, Phookamsak et al. 2014). For example, *Parastagonospora* species are pathogenic on *Poaceae* and are directly or indirectly responsible for significant crop losses in wheat, barley and rye worldwide (Quaedvlieg et al. 2013). In addition, many genera, such as, *Phaeosphaeriopsis*, *Setophoma, Xenoseptoria* and *Wojnowicia* have been found associated with leaf spot disease of various hosts (Arzanlou and Crous 2006, Quaedvlieg et al. 2013; Wijayawardene et al. 2013; Phookamsak et al. 2014).

Barr (1979a) introduced *Phaeosphaeriaceae* based on the type genus *Phaeosphaeria*, and the history has been reviewed by Phookamsak et al. (2014). The latter authors also carried out a revision of *Phaeosphaeriaceae*, based on multi-gene (ITS, LSU, SSU, RPB2 and TEF1) analyses coupled with morphological data. They included 30 genera in the family, in which 17 genera are asexual morphs, Liu et al. (2015) and Ariyawansa et al. (2015c) added four additional genera in *Phaeosphaeriaceae*, and to date, 37 genera are accommodated in the family (Table 1).

In the present study we introduce ten new asexual taxa in *Phaeosphaeriaceae* using combined ITS and LSU sequence data, as well as morphological data.

Material & methods

Collection and examination of specimens

Fresh specimens were collected from terrestrial habitats in Italy. Samples were examined and pure cultures obtained by single spore isolation following the method described in Chomnunti *et al.* (2014). The growing colonies were transferred to 2 % potato-dextrose agar (PDA) and incubated at 25°C. The colony characters and growth rates were determined after one to four weeks. The pure cultures from our study are deposited at Mae Fah Luang University Culture Collection (MFLUCC). Duplicate cultures are deposited in the International Collection of Microorganisms from Plants, Landcare Research, New Zealand (ICMP) and Kunming Institute of Botany Culture Collection (KUMCC). The holotype is deposited at the herbarium of Mae Fah Luang University (MFLU), Chiang Rai, Thailand, and the isotype specimens are deposited at the herbarium of Kunming institute of Botany Chinese Academy of Sciences (KUN). Faces of Fungi numbers and Index Fungorum numbers were obtained as explained in Jayasiri et al. (2015). The asexual morphs were established in culture using the method of Phookamsak et al. (2015).

DNA extraction, PCR amplification and sequencing

Isolates were grown on PDA plates in the darkness at 25°C until completely covering the agar surface. The mycelium (about 50 mg) was scraped off and collected in a 1.5 ml micro centrifuge tube. Genomic DNA was extracted from fresh mycelium, following the specification of Biospin Fungus Genomic DNA Extraction Kit (BioFlux®). For *Poaceicola arundinis* sp. nov. and *Septoriella allojunci* sp. nov., despite several attempts we could not isolate them into culture. Therefore, DNA was extracted directly from the conidiomata of the dried specimens following the method of Li et al. (2015). The primer pairs LROR and LR5 as defined by Vilgalys and Hester (1990) were used to amplify a segment of the large subunit rDNA. Primer pairs ITS4 and ITS5 as defined by White et al. (1990) were used to amplify the internal transcribed spacers (ITS). DNA amplification was performed by polymerase chain reaction (PCR). The sequencing of PCR products were carried at Shang Hai Biological Engineering Technology Co. Ltd (Shang Hai, P. R. China).

DNA sequence data analysis

Sequences were obtained from GenBank mostly following previous literature (Phookamsak et al. 2014, Ariyawansa et al. 2014, 2015c, Crous et al. 2015b, Liu et al. 2015) and are listed in Table 2.

Multiple sequences were aligned using Bioedit v. 7.0.9 (Hall, 1999) and Clustal X v. 1.83 (Thompson *et al.*, 1997). The alignments were checked visually and improved manually wherever necessary. A maximum likelihood (ML) analysis was performed with raxmlGUI version 1.3 (Silvestro & Michalak 2011). The optimal ML tree search was conducted with 1000 separate runs, using the default algorithm of the program from a random starting tree for each run. The final tree was selected among suboptimal trees from each run by comparing likelihood scores under the GTRGAMMA substitution model. The resulting trees were printed with TreeView v. 1.6.6 (Page, 1996).

Results

Phylogenetic analyses

ITS and LSU sequence data were used to resolve the generic placement of the collections of *Phaeosphaeriaceae*. The alignment datasets included 83 taxa of which *Didymella exigua* was used as the outgroup taxon. The combined datasets comprised 1435 characters including gaps. The best scoring RAxML tree was chosen as the backbone tree and is shown in Figure 1.

The phylogenetic analyses show that *Poaceicola elongata* comb. nov. and two other species, *Po. arundinis* and *Po. bromi*, are represented as a lineage distinct from any other genera in *Phaeosphaeriaceae* (Fig. 1). The other strains are distributed in *Nodulosphaeria* (one species), *Parastagonospora* (five species), and *Septoriella* (one species). Based on the morphology and sequence data, these fresh collections are introduced as new asexual taxa in *Phaeosphaeriaceae*. The new taxa are five species of *Parastagonospora*, i.e. *P. allouniseptata*, *P. dactylidis*, *P. minima*, *P. talica* and *P. uniseptata*; *Septoriella allojunci* sp. nov, and an asexual morph of *Nodulosphaeria*, *N. digitalis*.

Taxonomy

Nodulosphaeria digitalis W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Fig. 2

Index Fungorum number: IF551664 *Facesoffungi number*: FoF 01302,

Etymology – Named after the host genus, *Digitalis*.

Holotype – MFLU 15–2716

Sexual morph – Undetermined. Asexual morph – Coelomycetous. Conidiomata 130–150 µm high, 240–270 µm diam., pycnidioid, brown, solitary, aggregated or confluent, semi-immersed, subglobose, unilocular, papillate. Ostiole centrally located, short. Wall of conidiomata 20–60 µm wide, composed thick-walled, brown outer cells of textura angularis, and an inner layers of hyaline textura prismatica. Conidiophores cylindrical, short, hyaline, arrising from the inner layer of conidioma. Conidiogenous cells 7–15 × 2–4 µm, entoblastic, phialidic, cylindrical to subcylindrical, hyaline, smooth-walled. Conidia 9–18 × 2–9 µm (\bar{x} = 15 × 3 µm; n = 30), hyaline, falcate, fusiform, strongly curved towards the apices, rounded at both ends, 0–1-septate, smooth and thick-walled.

Material examined – ITALY, Province of Arezzo [AR], Bagno di Cetica, on dead stem of *Dactylis* sp., 1 October 2012, Erio Camporesi, IT-753 (MFLU 15–2716); *ibid.* (KUN! HKAS 90731).

Notes – *Nodulosphaeria digitalis* clustered with *Nodulosphaeria* species with high bootstrap suppert (99%), but *N. digitalis* formed a separate branch basal to *N. modesta* (Rabenh.) Munk ex L. Holm) and *N. italica* (Fig. 1). This is the first recorded asexual morph for *Nodulosphaeria*.

Table 1 Genera of *Phaeosphaeriaceae*, types species and morphs

Genus name	Types species	Sexual/Asexual morph	Reference
Allophaeosphaeria	Allophaeosphaeria muriformia	Asexual morph: Coelomytous	Liu et al. 2015
Amarenographium	Amarenographium metableticum	Sexual morph unknown	Phookamsak et al. 2014
Amarenomyces	Amarenomyces ammophilae	Asexual morph unknown	Phookamsak et al. 2014
Ampelomyces	Ampelomyces quisqualis	Sexual morph unknown	Phookamsak et al. 2014
Bricookea	Bricookea sepalorum	Asexual morph unknown	Phookamsak et al. 2014
Chaetosphaeronema	Chaetosphaeronema hispidulum	Sexual morph unknown	De Gruyter et al. 2010
Dematiopleospora	Dematiopleospora mariae	Asexual morph unknown	Wanasinghe et al. 2014
Diederichomyces	Diederichomyces xanthomendozae	Sexual morph unknown	Trakunyingcharoe et al. 2014
Dothideopsella	Dothideopsella agminalis	Asexual morph unknown	Wijayawardene et al. 2014b
Entodesmium	Entodesmium rude	Asexual morph unknown	Zhang et al. 2009
Eudarluca	Eudarluca australis	Asexual morph unknown	Wijayawardene et al. 2014b
Galliicola	Galliicola pseudophaeosphaeria	Asexual morph unknown	Ariyawansa et al. 2015 b
Leptospora	Leptospora porphyrogona	Asexual morph unknown	Crous et al. 2006
Loratospora	Loratospora aestuari	Asexual morph unknown	Schoch et al. 2009
Neosetophoma	Neosetophoma samararum	Sexual morph unknown	De Gruyter et al. 2010
Neostagonospora	Neostagonospora caricis	Sexual morph unknown	Quaedvlieg et al.2013
Nodulosphaeria	Nodulosphaeria hirta	Asexual morph: Coelomytous	from this study
Ophiobolus	Ophiobolus disseminans	Asexual morph unknown	Phookamsak et al. 2014
Ophiosphaerella	Ophiosphaerella graminicola	Asexual morph unknown	Zhang et al. 2012
Paraphoma	Paraphoma radicina	Sexual morph unknown	Gruyter et al. 2010
Parastagonospora	Parastagonospora nodorum	Sexual morph = phaeosphaeria-like	Quaedvlieg et al. 2013
Phaeosphaeria	Phaeosphaeria oryzae	Asexual morph = Phaeoseptoria	Quaedvlieg et al. 2013
Phaeosphaeriopsis	Phaeosphaeriopsis glaucopunctata	Asexual morph Coelomycetous	Phookamsak et al. 2014
Phaeostagonospora	Phaeostagonospora nolinae	Sexual morph unknown	Phookamsak et al. 2014
Poaceicola	Poaceicola arundinis	Sexual morph= Phaeosphaeria-like	from this study
Populocrescentia	Populocrescentia forlicesenensis	Sexual morph unknown	Ariyawansa et al. 2015 b
Sclerostagonospora	Sclerostagonospora heraclei	Sexual morph = phaeosphaeria-like	Quaedvlieg et al. 2013
Scolecosporiella	Scolecosporiella typhae	Asexual morph unknown	Phookamsak et al. 2014
Scolicosporium	Scolicosporium fagi	Sexual morph unknown	Wijayawardene et al. 2013
Septoriella	Septoriella phragmitis	Sexual morph unknown	Crous et al. 2015

Genus name	Types species	Sexual/Asexual morph	Reference	
Setomelanomma	Setomelanomma holmii	Sexual morph unknown	Phookamsak et al. 2014	
Setophoma	Setophoma terrestris	The sexual morph of <i>Setophoma</i> is similar to <i>Phaeosphaeria</i> species	Phookamsak et al. 2014	
Stagonospora	-	Sexual morph unknown	Phookamsak et al. 2014	
Sulcispora	Sulcispora pleurospora	Asexual morph unknown	Senanayake et al. 2014	
Tiarospora	Tiarospora westendorpii	Sexual morph unknown	Phookamsak et al. 2014	
Vrystaatia	Vrystaatia aloeicola	Sexual morph unknown	Quaedvlieg et al. 201	
Wojnowicia	Wojnowicia hirta	Sexual morph unknown	Crous et al. 2015	
Wojnowiciella	Wojnowiciella eucalypti	Sexual morph unknown	Crous et al. 2015a	
Xenophoma	Xenophoma puncteliae	Sexual morph unknown	Phookamsak et al. 2014	
Xenoseptoria	Xenoseptoria neosaccardoi	Sexual morph unknown	Quaedvlieg et al. 2013	

Table 2 Collection details and GenBank accession number of isolates includes in this study. The newly generated sequences are indicated in bold, T signifies ex-type/ex-epitype isolates.

Species name	strain	Host	Country	GenBank accession number	
				ITS	LSU
Allophaeosphaeria muriformia	MFLUCC 13-0349 ^T	-	Italy	KP765680	KP765681
Allophaerosphaeria subcylindrospora	MFLUCC 13-0380 ^T	Dactylis glomerata	Italy	KT314184	KT314183
Ampelomyces quisqualis	CBS 129.79	-	Canada	HQ108038	JX681064
Chaetosphaeronema coonsii	CBS 559.78	Malus sylvestris	Japan	-	EU754196
Dematiopleospora mariae	MFLUCC 13-0612 ^T	Ononis spinosa	Italy	-	KJ749653
Didymella exigua	CBS 183.55 ^T	Rumex arifolius	France	GU237794	EU754155
Diederichomyces caloplacae	CBS 129338	Caloplaca cerina	Canada	KP170639	JQ238643
Diederichomyces cladoniicola	CBS 128026	Cladonia sp.	Spain	KP170642	JQ238628
Diederichomyces ficuzzae	CBS 128019	Ramalina fastigiata	France	KP170647	JQ238616
Entodesmium rude	CBS 650.86	Unknown	-	-	GU301812
Galliicola pseudophaeosphaeria	MFLUCC 14-0527 ^T	Galium sp.	Italy	KT326692	KT326693
Leptospora rubella	CPC 11006	Eucalyptus sp.	Colombia	DQ195780	DQ195792
Loratospora aestuarii	JK 5535B	Juncus roemerianus	USA	-	GU301838
Neosetophoma samarorum	CBS 138.96 ^T	Phlox paniculata	Netherlands	FJ427061	KF251664
Neostagonospora caricis	CBS 135092/S616 ^T	Carex acutiformis	Netherlands	KF251163	KF251667
Neostagonospora elegiae	CBS 135101 ^T	Elegia cuspidata	South Africa	KF251164	KF251668

Species name	strain	Host	Country	GenBank accession number	
-				ITS	LSU
Nodulosphaeria aconiti	MFLUCC 13-0728	Aconitum vulparia	Italy		
Nodulosphaeria digitalis	-	Dactylis sp.	Italy	KU058710	KU058720
Nodulosphaeria dolioloides	MFLUCC 15-0065	Achillea sp.	Italy	-	-
Nodulosphaeria hirta	MFLUCC 13-0867	Achillea sp	Italy		
Nodulosphaeria sanguisorbae	MFLUCC 13-0745	Sanguisorba minor	Italy		
Nodulosphaeria scabiosae	MFLUCC 14-1111	Scabiosa sp.	Italy		
Nodulosphaeria senecionis	MFLUCC 15-1297	Senecio sp.	Italy	KT290257	KT290257
Nodulosphaeria spectabilis	MFLUCC 14-1112	Peucedanum cervaria	Italy	-	-
Ophiobolus cirsii	MFLUCC 13-0218	-	Italy	KM014664	KM014662
Ophiobolus disseminans	AS2L14-6	-	- -	KP117305	-
Ophiosphaerella agrostidis	MFLUCC 12-0007	grass	Thailand	KM434272	KM434282
Ophiosphaerella agrostidis	MFLUCC 11-0152	grass	Thailand	KM434271	KM434281
Paraphoma chrysanthemicola	CBS 522.66	Chrysanthemum morifolium	United Kingdom	FJ426985	KF251670
Paraphoma radicina	CBS 111.79 ^T	Malus sylvestris	Netherlands	KF251172	KF251676
Parastagonospora allouniseptata	MFLUCC 13-0386 ^T	Dactylis glomerata	Italy	KU058711	KU058721
Parastagonospora avenae	CBS 289.69	Lolium perenne	Germany	KF251174	KF251678
Parastagonospora avenae	CBS 290.69	Lolium perenne	Germany	KF251175	KF251679
Parastagonospora caricis	CBS 135671/S615 ^T	Carex acutiformis	Netherlands	KF251176	KF251680
Parastagonospora dactylidis	MFLUCC 13-0375 ^T	Dactylis sp.	Italy	KU058712	KU058722
Parastagonospora italica	MFLUCC 13-0377 ^T	Dactylis sp.	Italy	KU058714	KU058724
Parastagonospora minima	MFLUCC 13-0376 ^T	Dactylis sp.	Italy	KU058713	KU058723
Parastagonospora nodorum	CBS 110109	Lolium perenn	Denmark	KF251177	KF251681
Parastagonospora poae	CBS 135091	Poa sp.	Netherlands	KF251179	KF251683
Parastagonospora poae	CBS 135089 ^T	Poa sp.	Netherlands	KF251178	KF251682
Parastagonospora poagena	CBS 136776 ^T	Poa sp.	Netherlands	KJ869116	KJ869174
Parastagonospora uniseptata	MFLUCC 13-0387 ^T	Daucus sp.	Italy	KU058715	KU058725
Phaeosphaeria papayae	CBS 135416/S528 ^T	Carica papaya	Brazil	KF251187	KF251690
Phaeosphaeria alpina	CBS 456.84	Phleum alpinum	Switzerland	KF251181	KF251684
Phaeosphaeria chiangraina	MFLUCC 13–0231 ^T	Oryza sativa	Thailand	KM434270	KM434280
Phaeosphaeria eustoma	CBS 573.86	Dactylis glomerata	Switzerland	AF439479	-
Phaeosphaeria eustoma	CBS 573.86 _	Dactylis glomerata	Switzerland	AF439479	-
Phaeosphaeria musae	CBS 120026 ^T	Unknown	Unknown	DQ885894	GU301862
Phaeosphaeria nigrans	CBS 307.79	Zea mays	Switzerland	KF251184	KF251687
Phaeosphaeria oryzae	CBS 110110 ^T	Oryza sativa	South Korea	KF251186	KF251689
Phaeosphaeria thysanolaenicola	MFLUCC 10–0563 ^T	Thysanolaena maxima Kuntze	Thailand	KM434266	KM434276

Species name	strain	Host	Country	GenBank accession number	
				ITS	LSU
Phaeosphaeria typharum	CBS 296.54	Nardus stricta	Switzerland	KF251192	KF251695
Phaeosphaeriopsis glaucopunctata	MFLUCC 13-0265 ^T	Ruscus aculeatus	Italy	KJ522473	KJ522477
Phaeosphaeriopsis glaucopunctata	MFLUCC 13-0220	Ruscus hypoglossum	Italy	KJ522474	KJ522478
Phaeosphaeriopsis triseptata	MFLUCC 13-0271 ^T	Ruscus aculeatus	Italy	KJ522475	KJ522479
Poaceicola arundinis	-	reed (Juncaceae)	Italy	KU058716	KU058726
Poaceicola bromi	MFLUCC 13-0739 ^T	Bromus sterilis	Italy	KU058717	KU058727
Poaceicola elongate	MFLUCC 12–4444 ^T	Unknown	Italy	KM491546	KM491548
Populocrescentia forlicesenensis	MFLUCC 15–0651 ^T	Populus nigra	Italy	KT306948	KT306952
Sclerostagonospora cycadis	CBS 123538 ^T	Cycas revolut	Japan	FJ372393	FJ372410
Scolicosporium minkeviciusii	MFLUCC 12–0089 ^T	Quercus pubescens	Italy	-	KF366382
Septoriella allojunci	-	Juncus sp.	Italy	KU058718	KU058728
Septoriella hubertusii	CBS 338.86 ^T	Phragmites australis	France	KF251230	KF251733
Septoriella leuchtmannii	CBS 459.84	Phragmites australis	Switzerland	KF251188	KF251691
Septoriella oudemansii	CPC 24116 ^T	Phragmites australis	Netherlands	-	KJ869224
Septoriella phragmitis	CPC 24118 ^T	Phragmites sp.	Netherlands	KR873251	KR873279
Septoriella poae	CBS 136766 ^T	Poa sp.	Netherlands	KJ869111	KJ869169
Setomelanomma holmii	CBS 110217	Unknown	-	_	GU301871
Setophoma chromolaena	CBS 135105T/CPC 18553	Chromolaena odorata	Brazil	KF251244	KF251747
Setophoma sacchari	CBS 333.39	Saccharum officinarum	Brazil	KF251245	KF251748
Setophoma sacchari	MFLUCC 11-0154	Saccharum officinarum	Thailand	KJ476144	KJ476146
Stagonospora neglecta	CBS 343.86	Phragmites australi	France	AJ496630	EU754218
Sulcispora pleurospora	MFLUCC 13-0796	Unknown	Italy	KP271443	KP271444
Vagicola vagans	CBS 604.86 ^T	Calamagrostis arundinacea	Sweden	KF251193	KF251696
Vrystaatia aloeicola	CBS 135107/CPC 20617 ^T	Aloe maculata	South Africa	KF251278	KF251781
Wojnowicia dactylidicola	MFLUCC 13-0738 ^T	Dactylis glomerata	Italy	KP744469	KP684147
Wojnowicia lonicerae	MFLUCC 13-0737 ^T	Lonicerasp	Italy	KP744471	KP684151
Wojnowicia spartii	MFLUCC 13-0402 ^T	Spartium sp.	Italy	KU058719	KU058729
Wojnowicia dactylidis	MFLUCC 13-0735 ^T	Dactylis glomerata	Italy	KP744470	KP684149
Wojnowiciella eucalypti	CPC 25024 ^T	Eucalyptus grandis	Colombia	KR476741	KR476774
Wojnowiciella viburni	MFLUCC 12-0733 ^T	Viburnum utile	China	KC594286	KC594287
Xenophoma puncteliae	CBS 128022 ^T	Punctelia rudecta	USA	JQ238617	JQ238619
Xenoseptoria neosaccardoi	CBS 128665 ^T	Lysimachia vulgaris var. Davurica	South Korea	KF251281	KF251784
Xenoseptoria neosaccardoi	CBS 120.43	Cyclamen persicum	Netherlands	KF251280	KF251783

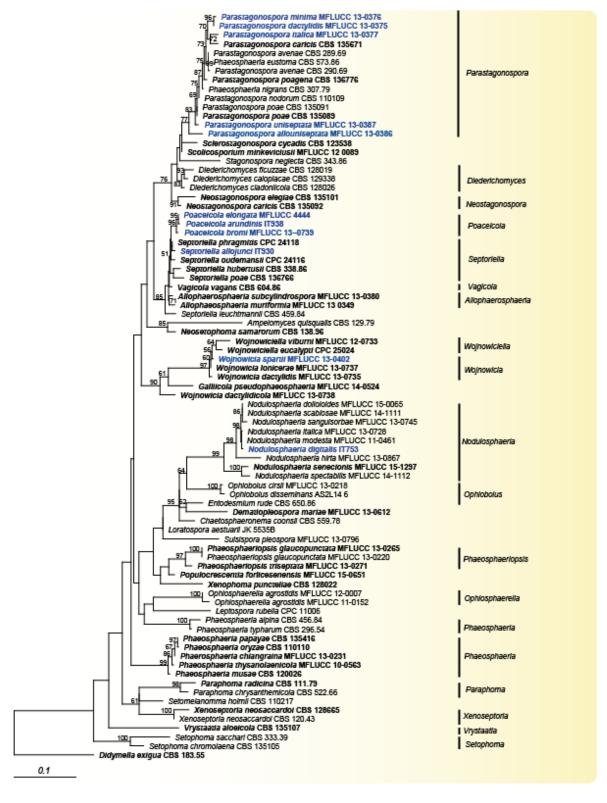


Fig. 1 – Best scoring RAxML tree of *Phaeosphaeriaceae* strains obtained from analysis of ITS and LSU sequence data. RAxML bootstrap support values (equal to or greater than 50% based on 1.000 replicates) are shown at the nodes. The ex-type strains are in bold; the new isolates are in blue. The tree is rooted to *Didymella exigua* CBS 183.55.

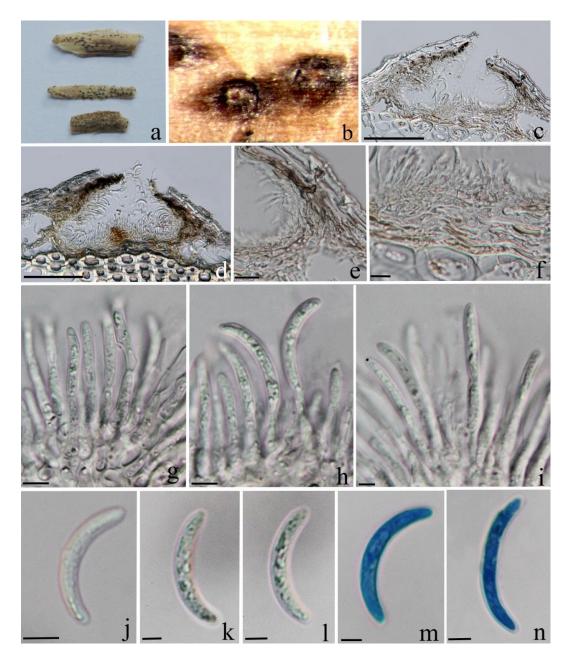


Fig. 2 – *Nodulosphaeria digitalis* (MFLU 15–2716, **holotype**) a Herbarium specimen b Appearance of black conidiomata on the host. c, d Vertical section of conidioma. e, f Section of peridium. g–i Conidiogenous cells and developing conidia. g–n Conidia. – Bars: c–d = 100 μ m; e–f = 20 μ m; g–i = 5 μ m.

Parastagonospora allouniseptata W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Fig. 3

Index Fungorum number: IF551665 Facesoffungi number: FoF 01307

Etymology – Named after its morphological similarity to *Parastagonospora uniseptata* Holotype – MFLU 15–0698

Saprobic on dead stem of Dactylis glomerata L. (Poaceae), forming conspicuous, rounded, black fruiting bodies. **Sexual morph** – Undetermined. **Asexual morph** – Coelomycetous. Conidiomata 60–90 μm high, 70–90 μm diam., pycnidial, black, solitary or gregarious, semi-

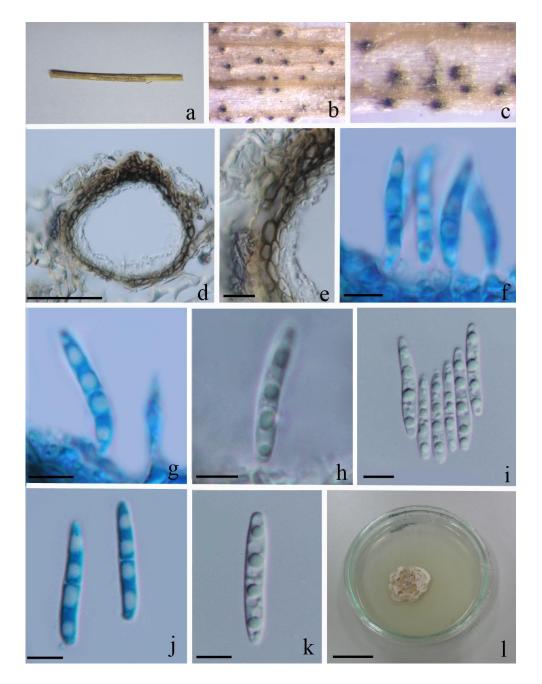


Fig. 3 – *Parastagonospora allouniseptata* (MFLU 15–0698, **holotype**) a Herbarium specimen. b–c Appearance of black conidiomata on the host. d Vertical section of conidioma. e Section of peridium. f–h Conidiogenous cells and developing conidia. i–k Conidia. l Culture on PDA. – Bars: $d = 50 \mu m$; $e = 10 \mu m$; $f-k = 5 \mu m$; l = 25 mm.

immersed, unilocular, globose to subglobose, ostiolate. *Ostiole* centrally located, papillate. *Wall of conidiomata* 5–10 µm wide, comprising outer layer of brown to dark brown, thin-walled cell of *textura globosa*, and an inner layer of light brown to hyaline cell of *textura angularis*. *Conidiophores* reduced to conidiogenous cells, arising from inner layers of stroma. *Conidiogenous cells* 3–5 µm long × 3–5.5 µm wide, phialidic, integrated, short, hyaline, smooth. *Conidia* 16–22 × 2.5–3.5 µm (\overline{x} = 19.5 × 3 µm; n = 30), hyaline, subcylindrical, 1-septate, with narrow and subobtuse apex and truncate base, granular, smooth and thick-walled.

Culture characteristics – Colonies slow growing on PDA, reaching 15 mm diam. after 4 weeks at 20–25°C, circular to irregular, with uneven margins, whitened to pink, flattened, filamentous, sparse, aerial mycelium on the surface, reverse similar in colour.

Material examined – ITALY, Province of Forlì-Cesena [FC], Galeata, Passo delle Forche, on dead stem of *Dactylis glomerata* L. (*Poaceae*), 15 October 2013, Erio Camporesi, IT-719 (MFLU 15–0698, **holotype**); ex-type living culture, MFLUCC 13–0386, KUMCC 15–0134; *ibid*. IT-719A (KUN! HKAS 90735, **isotype**).

Notes – Parastagonospora allouniseptata is similar to P. uniseptata in having pycnidial, globose to subglobose, conidiomata, with hyaline, subcylindrical, 1-septate, granular conidia, but is distinct in having shorter and narrower conidia than P. uniseptata (14–18 × 2–3 μ m). Phylogenetically, P. allouniseptata clusters with Parastagonospora species with 88% bootstrap support. Furthermore, P. allouniseptata forms a distinct branch in the Parastagonospora phylogeny (Fig. 1).

Parastagonospora dactylidis W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Fig. 4

Index Fungorum number: IF551666 Facesoffungi number: FoF 01303

Etymology – Named after the host genus, *Dactylis*.

Holotype – MFLU 15–0693

Saprobic on dead stem of Dactylis sp., forming numerous, conspicuous, dark brown fruiting bodies in a linear series on the surface. **Sexual morph** – Undetermined. **Asexual morph** – Coelomycetous. Conidiomata 50–100 µm high, 100–150 µm diam., pycnidial, brown, separate, ampulliform or globose, immersed, unilocular, thick-walled, ostiolate. Wall of conidiomata 10–15 µm wide, composed of 3–4 layers, with outer 1–2-layers of dark brown and inner 1–2-layers of hyaline cells of textura angularis. Ostiole circular, papillate. Conidiophores reduced to conidiogenous cells. Conidiogenous cells 2–6 µm long × 3–8 µm wide, phialidic, ampuliform, determinate, hyaline, unbranched, and developing only at the base, formed from the inner cells of the pycnidial wall. Conidia 25–40 × 4–5.5 µm (\bar{x} = 30 × 4.5 µm; n = 30), hyaline, fusiform, curved, rounded at both ends, slightly narrower at the base, 3-septate, constricted at the septa, with distinctly granular cytoplasm.

Culture characteristics – Colonies on PDA, reaching 20 mm after 2 wk at 20–25°C, with dense, white, flat, aerial mycelium, with rounded, smooth, margins, reverse similar in colour.

Material examined – ITALY, Province of Arezzo [AR], Passo della Consuma, on dead stem of *Dactylis* sp., 19 June 2012, Erio Camporesi, IT-448 (MFLU 15–0693, **holotype**); ex-type living culture, MFLUCC 13–0375, ICMP 20774, KUMCC15-0131; *ibid*. (KUN! HKAS 90738, **isotype**).

Notes – *Parastagonospora dactylidis* is most closely related to *P. minima* (Fig. 1). However, the individual sequence of *P. dactylidis* as compared with *P. dactylidis* has seven different substitions in the LSU sequence data. Morphologically, these two species also can be easily distinguished in form of conidia. *P. dactylidis* has fusiform conidia with a slightly narrower base, and distinctly granular cytoplasm, whereas *P. minima* has subcylindrical conidia which are wider in the basal half, and narrow at the apex. In addition, the conidiomata and conidia of *P. dactylidis* are larger and longer than those of *P. minima* (conidiomata: 40–70 μ m high, 50–100 μ m diam., conidia: 20–28 \times 3.5–4.5 μ m). Thus *P. dactylidis* is introduced as a novel species.

Parastagonospora minima W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Fig. 5

Index Fungorum number: IF551667 Facesoffungi number: FoF 01304

Etymology – Named after the small conidiomata.

Holotype – MFLU 15–0694

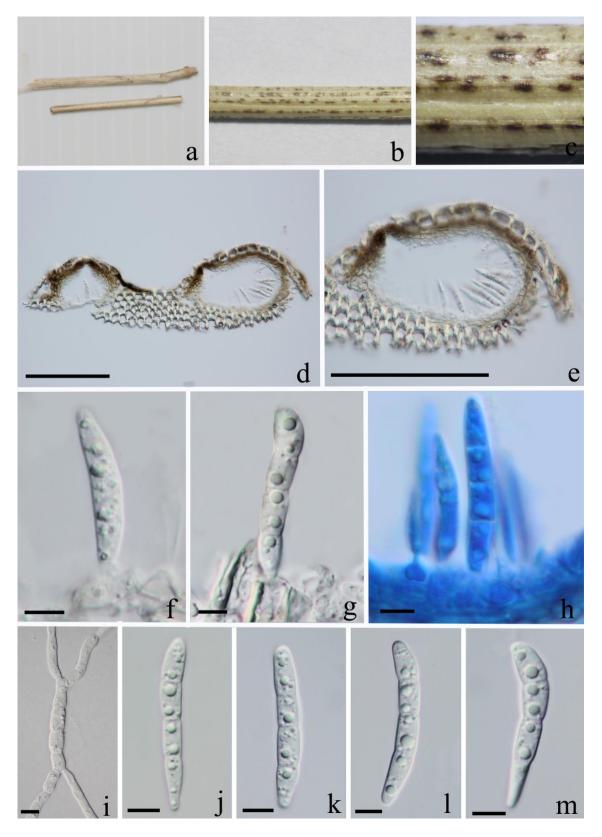


Fig. 4 – *Parastagonospora dactylidis* (MFLU 15–0693, **holotype**) a Herbarium specimen. b, c Appearance of brown conidiomata on the host. d–e Vertical section of conidioma. f–h Conidiogenous cells and developing conidia. i Germinating spore. j–m Conidia. – Bars: d–e = $50 \mu m$; f–m = $5 \mu m$.

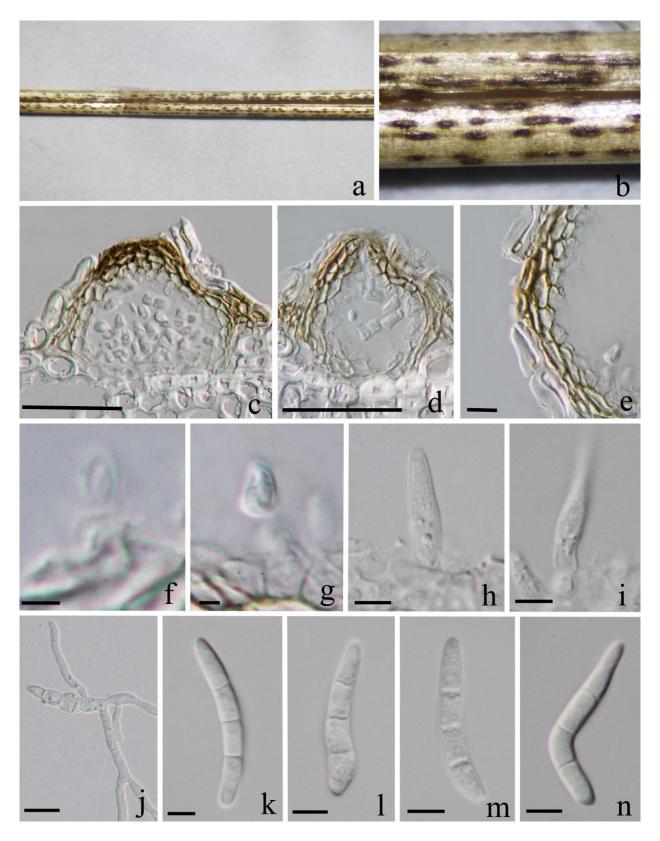


Fig. 5 – *Parastagonospora minima* (MFLU 15–0694, **holotype**) a Herbarium specimen. b Appearance of black conidiomata on the host. c–d Vertical section of conidioma. e Section of peridium. f–i Conidiogenous cells and developing conidia. j Germinating spore. k–n Conidia. – Bars: c–d = 100 μ m; e = 20 μ m; f–i, k–m = 5 μ m; j = 20 μ m.

Sexual morph – Undetermined. Asexual morph – Coelomycetous. Conidiomata 40–70 μ m high, 50–100 μ m diam., pycnidial, brown to black, amphigenous, separate, gregarious or confluent, obpyriform or globose, subepidermal, unilocular, ostiolate, thick-walled. Wall of conidiomata 6–10 μ m wide, composed of dark brown to pale brown, thick-walled cells of textura angularis. Ostiole centrally located, papillate, circular. Conidiophores reduced to conidiogenous cells. Conidiogenous cells 3–6.5 μ m long \times 3–7 μ m wide, phialidic, hyaline, smooth. Conidia 20–28 \times 3.5–4.5 μ m (\bar{x} = 25.5 \times 4 μ m; n = 30), hyaline, subcylindrical, wider at the basal half, narrow and rounded at both ends, slightly curved, 3-euseptate, smooth-walled.

Culture characteristics – Colonies on PDA, reaching 25 mm after 2 wk at 20–25°C, with moderate aerial mycelium, flat, dense, spreading, with rounded, smooth margins.

Material examined – ITALY, Province of Arezzo [AR], Passo della Consuma, on dead stem of *Dactylis* sp., 19 June 2012, Erio Camporesi IT-451 (MFLU 15–0694, **epitype**); ex-type living culture, MFLUCC 13–0376, ICMP 20776, KUMCC15–0132; *ibid*. (KUN! HKAS 90734, **isotype**).

Notes – $Parastagonospora\ minima$ is morphologically similar with the generic type, $P.\ nodorum$, in having subcylindrical, hyaline conidia, with an obtuse apex, as well as being of similar size ($P.\ nodorum\ 13-28\times2.8-4.6\ \mu m$) (Quaedvlieg et al 2013). However, recognizable differences between these two species are in the septation. $Parastagonospora\ minima$ has uniformly 3-septate conidia, whereas $P.\ nodorum\ has\ (0-)1(-3)$ -septate conidia. Phylogenetically, $P.\ minima$ is distinct from any other species of $Parastagonospora\ (Fig.1)$. Based on the above morphological characters together with molecular sequence data, $P.\ minima$ is introduced as a new species.

Parastagonospora italica W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Fig. 6

Index Fungorum number: IF551668 Facesoffungi number: FoF 01305

Etymology – Named after the country of collection.

Holotype – MFLU 15–0696

Saprobic on dead stem of Dactylis sp.. **Sexual morph** – Undetermined. **Asexual morph** – Coelomycetous. Conidiomata 65–80 µm high, (40–) 100–150 µm diam., pycnidial, brown to black, solitary or gregarious, subepidermal, immersed, unilocular, globose to subglobose, thick-walled, black. Wall of conidiomata 10–20 µm wide, composed of 2–4 layers, thick-walled cells of textura angularis, brown. Ostiole centrally located, circular, short, papillate. Conidiophores reduced to conidiogenous cells, arising from inner layers of pycnidia. Conidiogenous cells short, broadly cylindrical, phialidic, hyaline, smooth. Conidia $25-32 \times 3-4 \mu m$ ($\bar{x}=29.5 \times 3-5 \mu m$; n = 20), hyaline, cylindric-fusiform, with narrow and obtuse apex, truncate at base, slightly curved, 3-euseptate, smooth and thick-walled.

Culture characteristics – Colonies on PDA, reaching 20 mm after 10 d at 20–25°C, with crenate, white margins, pink, flattened, with dense, aerial mycelium on the surface; reverse pale-yellowish to pale brown in the middle, whitened at the margins.

Material examined – ITALY, Province of Forlì-Cesena [FC], Santa Sofia, Campigna, on dead stem of *Dactylis* sp., 24 June 2012, Erio Camporesi, IT-471 (MFLU 15–0696, **holotype**); ex-type living culture, MFLUCC 13–0377, ICMP 20777, KUMCC. *ibid*. 15 May 2013, Erio Camporesi, IT-471A (KUN! HKAS 90736, **isotype**).

Notes – In the multi-locus phylogeny, *Parastagonospora italica* clusters with *P. caricis* Quaedvl. et al., but this grouping is not well-supported. Morphologically, these two species can easily be distinguished by dimension and septation of conidia; *P. italica* has 3-septate conidia which are shorter and narrower than the 7–15-septate, $(50–)60–70(–75) \times (5–)6$ µm conidia in *P. caricis*. In addition, the conidiomata of *P. italica* are smaller than those of *P. caricis* which are up to 250 µm.

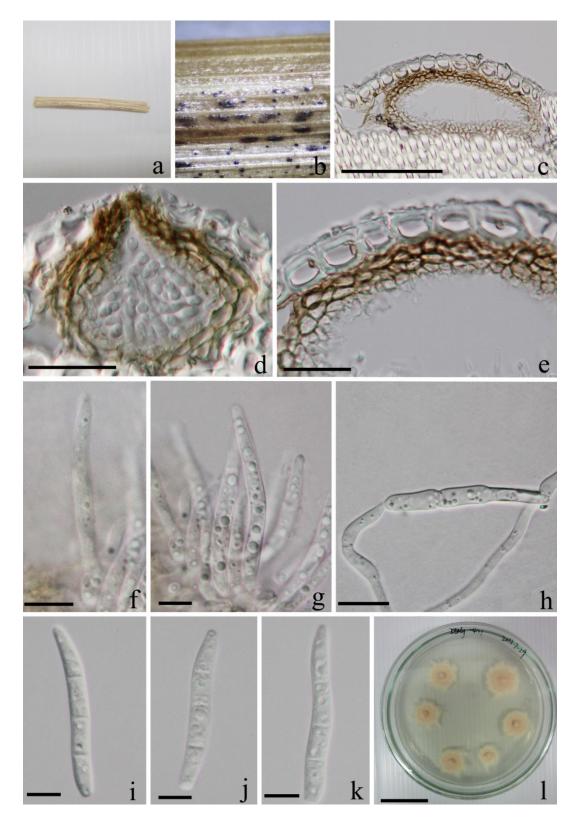


Fig. 6 – *Parastagonospora italica* (MFLU 15–0696, **holotype**) a Herbarium specimen. b Appearance of black conidiomata on the host surface. c–d Vertical section of conidioma. e Section of peridium. f–g Conidiogenous cells and developing conidia. h Germinating spore. i–k Conidia. l Culture on PDA. – Bars: $c = 100 \mu m$; $d = 25 \mu m$; $e = 20 \mu m$; $f-g = 5 \mu m$; $h = 10 \mu m$; $i-k = 5 \mu m$; $l = 25 \mu m$.

Parastagonospora uniseptata W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Index Fungorum number: IF551669 Facesoffungi number: FoF 01306

Etymology – Named after the fact that conidia are 1-septate.

Holotype – MFLU 15–0699

Sexual morph – Undetermined. Asexual morph – Coelomycetous. Conidiomata 60–100 μm high, 70–100 μm diam, pycnidial, brown to black, appearing on upper surface of the stem, solitary, sometimes gregarious or confluent, immersed to semi-immersed, globose or subglobose, unilocular, with a blackened tip above, ostiolate at the centre, thick-walled. Wall of conidiomata 10–20 μm wide, composed of 6-layers, outer 4–5-layers brown, inner 2–3-layers hyaline, upper wall region dark brown, with periclinal and basal wall composed of thick-walled cells of textura angularis. Conidiophores reduced to conidiogenous cells, arising all around the basal region of the conidiomata. Conidiogenous cells 3–6 μm long × 3–6.5 μm wide, ampulliform to broadly conical, phialidic, hyaline, smooth. Conidia 14–18 × 2–3 μm (\bar{x} = 16 × 2.5; n = 30), hyaline, subcylindrical, 1-septate, guttulate, truncate at the base, with obtuse apex, smooth-walled.

Culture characteristics – Colonies slow growing on PDA, reaching 20 mm diam. after 4 weeks at 20–25°C, with a glabrous, circular margin, pale yellow, flattened, filamentous, sparse, with aerial mycelium on the surface, similar in colour from below.

Material examined – ITALY, Province of Arezzo [AR], Modigliana, Bagno di Cetica, on dead stems of *Daucus* sp., 18 September 2012, Erio Camporesi, IT-727 (MFLU 15–0699, **holotype**); extype living culture, MFLUCC 13-0387, KUMCC 15–0134; *ibid.* IT-727A (KUN! HKAS 90730, **isotype**).

Notes – *Parastagonospora uniseptata* should be morphologically compared with stagonosporalike asexual morphs. The results show that *P. uniseptata* is more similar to the type of *Neostagonaspora*, *N. caricis* Quaedvl. et al. than to other species within *Parastogonospora*. However, phylogenetic tree reconstruction based on multigene (LSU and ITS) sequence analyses shows that *P. uniseptata* clusters together with type *P. nodorum*, as well as other species of *Parastagonospora* (BS=88%) (Fig. 1). *Parastogonospora uniseptata* however, forms a discrete branch basal to *P. poae* Quaedvl.et al. (Fig. 1). Thus *P. uniseptata* is introduced as a new species in *Parastagonospora*.

Poaceicola W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, gen. nov.

Index Fungorum number: IF551658 Facesoffungi number: FoF 01298

Etymology – Named after inhabiting grass (*Poaceae*).

Type species – *Poaceicola arundinis* W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, **sp. nov.**

Saprobic on dead stems of plant host. **Sexual morph** – Ascomata solitary to gregarious, black, immersed to semi-immersed, subepidermal, uniloculate, glabrous, globose to subglobose, papillate. Ostiole centrally located, black, smooth, with an ostiole, filled with hyaline periphyses. Peridium thick-walled, brown, composed of cells of textura angularis. Hamathecium comprising cellular, septate, broad, dense pseudoparaphyses. Asci 8-spored, bitunicate, fissitunicate, cylindrical, with a short, broad pedicel. Ascospores tetraseriate or partially overlapping, reddish-brown, fusiform, straight or inequilaterally curved, echinulate, 10-septate, fourth cell from apex swollen towards middle and slightly longer than adjacent cells, with a conspicuous sharply delimited sheath. **Asexual morph** – Coelomycetous. Conidiomata pycnidioid, solitary or aggregated, immersed or semi-immersed, unilocular, globose, papillate, dark brown. Wall of conidiomata comprising inner layers of textura angularis cells, gradually merging with the outer surrounding layers of brown, textura oblita. Ostiole

Fig. 7

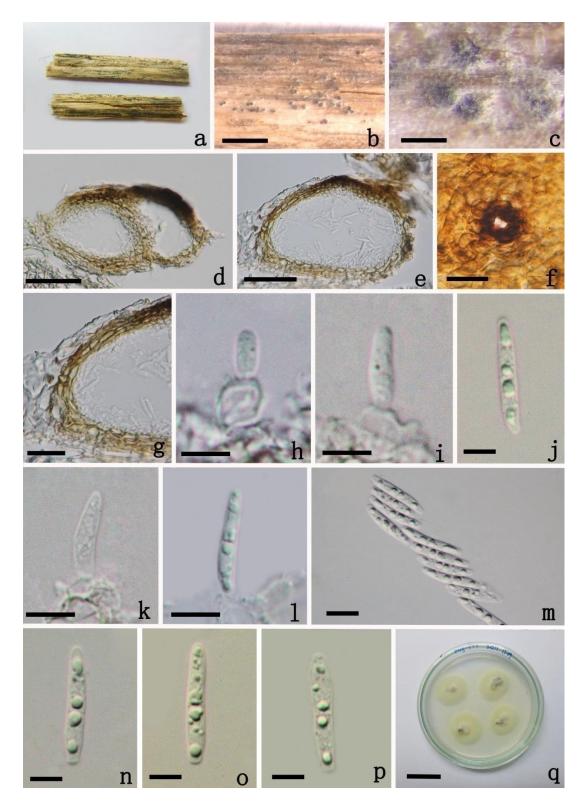


Fig. 7 – *Parastagonospora uniseptata* (MFLU 15–0699, **holotype**) a Herbarium specimen. b–c Appearance of black conidiomata on the host. d–e Vertical section of conidiomata. f Ostiole. g Section of peridium. h–i, k–l Conidiogenous cells and developing conidia. j, m–p Conidia. q Culture on PDA. – Bars: b–c = 200 μ m; d–e = 50 μ m; f = 15 μ m; g = 20 μ m; h–i, k–l= 5 μ m; j, m–p = 5 μ m; q = 25 mm.

central, circular to oval, papillate. *Conidiophores* reduced to conidiogenous cells. *Conidiogenous cells* holoblastic, phialidic, hyaline, smooth-walled, discrete, formed from the inner cells of the stroma. *Conidia* pale brown, cylindrical or sub-cylindrical, up to 7-euseptate, straight or slightly curved, smooth-walled..

Notes – The phylogeny of the family *Phaeosphaeriaceae* is reconstructed based on combined gene (LSU and ITS) analysis, showing that *Poaceicola* species cluster away from any other genera in *Phaeosphaeriaceae* (Fig. 1). Thus the genus *Poaceicola* is introduced to accommodate *Poaceicola elongata* comb. nov. and two asexual morphs introduced in this study i.e. *Po. arundinis* sp. nov. and *Po. bromi* sp. nov.

Poaceicola arundinis W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Fig. 8

Index Fungorum number: IF551659 Facesoffunginumber: FoF 01299

Etymology – Named after the host genus *Arundo*.

Holotype – MFLU 15–0702

Saprobic on dead stems of Arundo plinii Turra, forming conspicuous, rounded, black fruiting bodies. **Sexual morph** – Undetermined. **Asexual morph** – Coelomycetous. *Conidiomata* 100–150 µm high, 100–200 µm diam., pycnidioid, dark brown, solitary or aggregated, semi-immersed, unilocular, globose, papillate. *Wall of conidiomata* 10–30 µm wide, comprising inner layers of cells of *textura angularis*, gradually merging with the outer, surrounding layers of brown, *textura oblita*. *Ostiole* central, circular to oval, papillate. *Conidiophores* reduced to conidiogenous cells. *Conidiogenous cells* 3–6 µm wide, holoblastic, phialidic, hyaline, smooth-walled, discrete, formed from the inner cells of the stroma. *Conidia* 30–40 × 6.5–10 µm (\bar{x} = 35 × 8 µm; n = 30), pale brown, cylindrical, flexuous, up to 8-euseptate, slightly curved, smooth-walled, with middle cells wider than end cells, guttulate, with an acute apex, truncate at the base.

Material examined – ITALY, Province of Arezzo [AR], Montemezzano, on dead on dead stems of *Arundo plinii* Turra, 25 August 2013, Erio Camporesi, IT-938 (MFLU 15–0702); *ibid.* (KUN! HKAS 90732).

Notes – *Poaceicola arundinis* is distinct from *Po. bromi* in conidiomata and conidia form. *Poaceicola arundinis* has globose conidiomata with circular to oval, short ostioles, while *Po. bromi* has pyriform conidiomata with cylindrical, long ostioles. In addition, the conidia of *Po. arundinis* with up to 8-septa are longer and wider than those of *Po. bromi* which are 7-septate and $15-23 \times 3-5 \mu m$.

Poaceicola bromi Wijayawardene, W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov. Fig. 9

Index Fungorum number: IF551660 Facesoffungi number: FoF 01300

Etymology – Named after the host genus, *Bromus*.

Holotype – MFLU 15–2719

Saprobic on dead twigs of Bromus sterilis L. (Poaceae). Sexual morph – Undetermined. Asexual morph – Coelomycetous. Conidiomata 310–350 µm high, 240–280 µm diam., pycnidioid, dark brown, solitary, semi-immersed, unilocular, globose, papillate. Wall of conidiomata 29–49 µm wide, thick-walled, composed of thick-walled cells of textura angularis at the outside, with inner layers lighter and flattened. Setae 20–30 × 5–6 µm, lateral, sparse, subcylindrical, 1–2-septate, dark brown. Ostiole centrally located, cylindrical to subcylindrical, papillate. Conidiophores reduced to conidiogenous cells. Conidiogenous cells $1-2 \times 1-3$ µm, holoblastic, phialidic, hyaline, smoothwalled, discrete, formed from the inner cells of the stroma. Conidia $15-23 \times 3-5$ µm ($\overline{x}=20.5 \times 4$ µm; n = 30), pale brown to dark brown, cylindrical, straight or slightly curved, obtuse at both ends, up to 7-euseptate, constricted at the septa, smooth-walled.

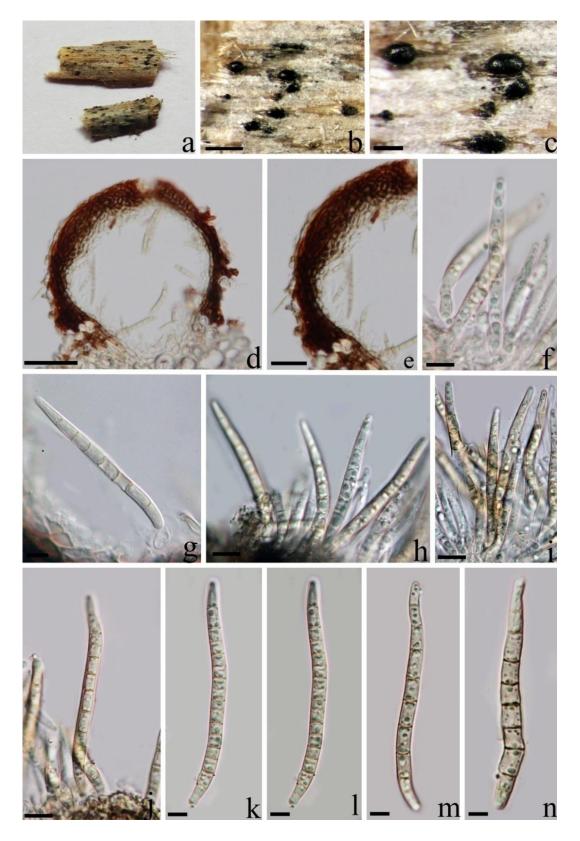


Fig. 8 – *Poaceicola arundinis* (MFLU 15–0702, **holotype**) a Herbarium specimen. b–c Appearance of black conidiomata on the host. d Vertical section of conidioma. e Section of peridium. f–i Conidiogenous cells and developing conidia. j–n Conidia. m Germinating spore. n Culture on PDA. – Bars: $b = 200 \mu m$; $c = 20 \mu m$; $d = 100 \mu m$; $e = 50 \mu m$; $f-i = 10 \mu m$.

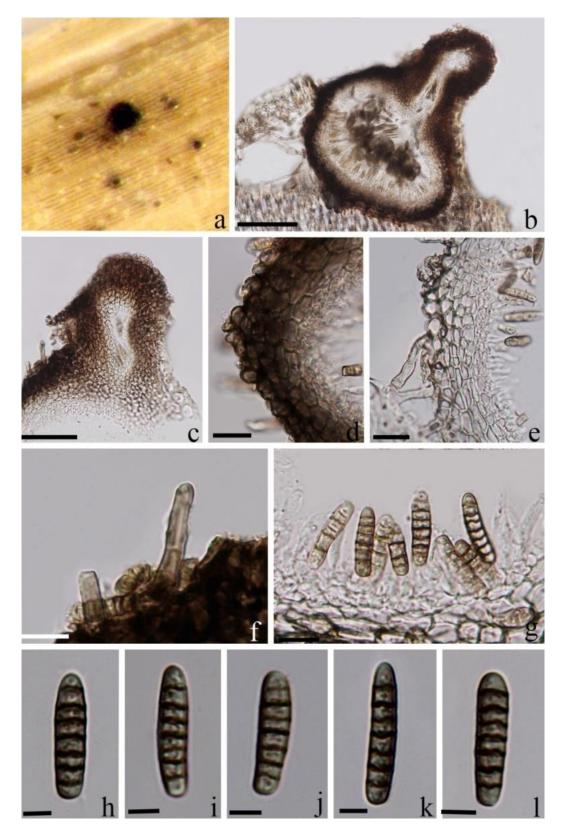


Fig. 9 – *Poaceicola bromi* (MFLU MFLU 15–2719, **holotype**) a Appearance of black conidioma on host. b Vertical section of conidioma. c Ostiole. d–e Section of peridium. f–g Conidiogenous cells and developing conidia. h–l Conidia. – Bars: $b = 100 \ \mu m$; $c = 50 \ \mu m$; $d-e = 20 \ \mu m$; $f-g = 10 \ \mu m$; $h-l = 5 \ \mu m$.

Culture characteristics – On PDA slow growing, attaining 2 cm diam. in 7 days, with circular to slightly wavy margin, greyish white from above, grey from below, with thin mycelium.

Material examined – ITALY, Province of Forlì-Cesena [FC], on leaves of *Bromus sterilis* L. (*Poaceae*), 19 July 2013, Erio Camporesi, NNW IT-1389 (MFLU 15–2719, **holotype**). (KUN! HKAS 90729, isotype), living cultures, MFLUCC 13–0739, GUCC 1389

Notes – *Poaceicola bromi* is morphologically and phylogenetically distinct from *Po. arundinis*; see notes under *Po. arundinis*.

Poaceicola elongata (Wehm.) Shoemaker & C.E. Babc.) W.J. Li, Camporesi & K.D. Hyde, comb. nov.

Index Fungorum number: IF551661 Facesoffungi number: FoF 01301 Holotype – MFLU14–0635

Basionym – *Phaeosphaeria elongata* (Wehm.) Shoemaker & C.E. Babc., Can. J. Bot. 67(5): 1540 (1989)

Synonym – Leptosphaeria elongata Wehm., Mycologia 44: 633 (1952). 633 (1952).

Notes – This specimen was collected from dead wood in Italy. Ariyawansa et al. (2014) designated the collection as a reference specimen of *Phaeosphaeria elongata*. However, *P. elongata* clusters close to *Po. cylindrispora* and *Po. bromi* with high bootstrap support (BS = 96%) (Fig. 1), and is best assigned to *Poaceicola*.

Septoriella allojunci W.J. Li, Camporesi, D.J. Bhat & K.D. Hyde, sp. nov.

Fig. 10

Index Fungorum number: IF551662 Facesoffungi number: FoF 01308

Etymology – Named after its morphological similar to Septoriella junci.

Holotype – MFLU 15–0701

Saprobic on dead stem of *Juncus* sp.. **Sexual morph** – Undetermined. **Asexual morph** – Coelomycetous. *Conidiomata* 90–130 µm high, 100–150 µm diam., stromatic, pycnidial, dark brown, solitary or aggregated, linearly disposed with only the dark spots visible in surface view, immersed, unilocular, globose, papillate. *Ostiole* centrally located, circular to oval, apapillate. *Wall of conidiomata* 10–20 µm wide, composed of brown, thick-walled cells of *textura angularis*, at the base, thin-walled, dark brown cells in upper part and surrounding the ostiole. *Conidiophores* reduced to conidiogenous cells. *Conidiogenous cells* 3–6 µm wide, holoblastic, discrete, phialidic, ampulliform, hyaline, smooth-walled, formed from the inner cells of the conidiomata. *Conidia* 48–70 × 3–6.6 µm (\bar{x} = 60 × 4 µm; n = 30), pale brown, subcylindrical, flexuous, with obtuse ends, 3–5-euseptate, smooth-walled, guttulate, with mucoid cap at the apex.

Material examined – ITALY, Province of Forlì-Cesena [FC], Galeata, Strada San Zeno, on dead stems of *Juncus* sp. (*Juncaceae*), 27 November 2012, Erio Camporesi, IT-930 (MFLU 15–0701, **holotype**); *ibid.* (KUN! HKAS 90733, **isotype**).

Notes – Crous et al. (2015b) fixed the application of the type species of generic name (Septoriella phragmitis Oudem.), and confirmed the placement of Septoriella in Phaeosphaeriaceae. In the present study, we introduced an additional taxon, Septoriella allojunci. Morphologically, S. allojunci shows similarities with S. junci and S. canadensis in the form of conidiomata and conidiogenous cells, but differs in the dimension, and septation of conidia. Septoriella allojunci has 3–5-septate conidia which are longer and wider than those of S. canadenesis which have 3–4-septate conidia (36–56 × 2.5–3.5 μ m), as well as shorter and wider than S. junci (6–7-septate, 49–90 × 2–3 μ m). Phylogenetically, S. allojunci is close to S. phragmitis (Fig. 1).

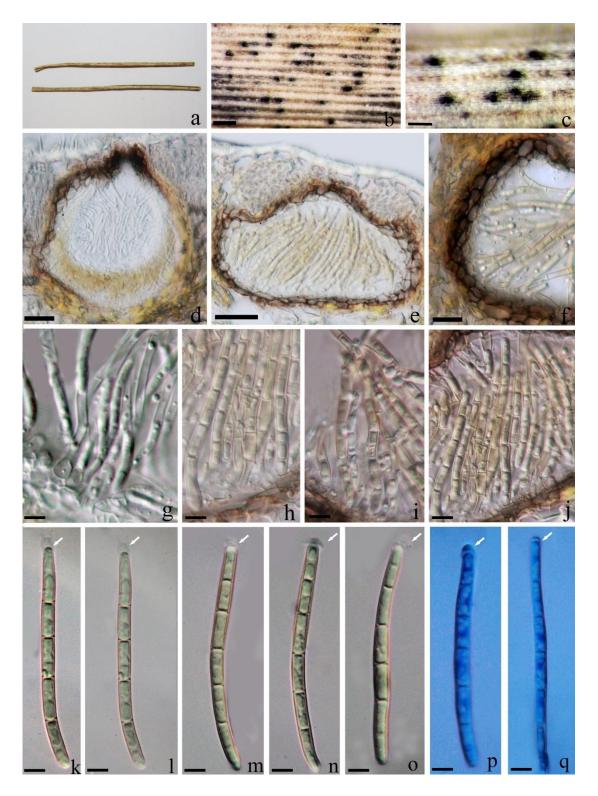


Fig. 10 – *Septoriella allojunci* (MFLU 15–0701, **holotype**) a Herbarium specimen. b–c Appearance of black conidiomata on the host surface. d–e Vertical section of conidioma. f Section of conidioma. g–j Conidiogenous cells and developing conidia. k–q Conidia. – Bars: b–c = 200 μ m; d–e = 100 μ m; f = 50 μ m; g–j = 10 μ m; k–q = 10 μ m.

Index Fungorum number: IF551663 Facesoffungi number: FoF 01309

Etymology – Named after the host genus, Spartium.

Holotype – MFLU 15–0700

Sexual morph – Undetermined. Asexual morph – Coelomycetous. Conidiomata 120–180 µm high, 140–200 µm diam., pycnidial, dark brown, scattered, immersed to semi-immersed, unilocular, globose to subglobose, glabrous, ostiolate. Ostiole centrally located, papillate, cylindrical. Wall of conidiomata 13–28 µm wide, composed of 4–5-cell layers, of thick-walled cells of textura angularis, and inner layers of light brown to hyaline textura angularis. Conidiophores reduced to conidiogenous cells. Conidiogenous cells 2.8–6.5 µm long \times 3–7 µm wide, phiallidic, hyaline, integrated, flask-shaped, thick-walled. Conidia 28–36 \times 5–6 (\overline{x} = 28 \times 5 µm; n = 30), dark brown, fusiform or cylindrical, straight or slightly curved, wide in the middle, 7–12-septate, constricted at septa, subobtuse at each end, thick-walled, smooth-walled, guttulate.

Culture characteristics – Colonies slow growing on PDA, reaching 10 mm diam. after 5d at 20–25°C, circular, flattened, with sparse, grey to olivaceous aerial mycelium, surface patches of olivaceous-grey in the older parts; reverse olivaceous-black.

Material examined – ITALY, Province of Forlì-Cesena [FC], Bagno di Romagna, Passo del Carnaio, on dead stem of *Spartium* sp., 17 November 2012, Erio Camporesi, IT-889 (MFLU 15–0700, **holotype**); ex-type living culture, MFLUCC 13–0402, ICMP 20790, KUMCC 15–0136; *ibid.* (KUN! HKAS 90737, **isotype**).

Notes – *Wojnowicia* was introduced by Saccardo (1892) with *W. hirta* as the type species. Subsequently, the genus was expanded to include two more species, viz. *W. colluvium* D.F. Farr & Bills and *W. ephedrae* Hollós (Sutton 1980, Farr & Bills 1995). According to Index Fungorum (2015) there are 14 species epithets in *Wojnowicia*. However, *W. graminis* (McAlpine) Sacc. & D. Sacc. and *W. tenella* Pat. were synonymised under *W. hirta* by Sutton (1980). *Wojnowicia buxi* Bertault & Malençon was synonymised under *W. ephedrae* Hollós by Farr & Bills (1995). *Wojnowicia bryophila* Racov., *W. exilis* (Corda) Sacc. & Traverso and *W. lophostoma* (Höhn.) Sacc. were excluded from *Wojnowicia* (Sutton 1980, Farr & Bills 1995, Wijayawardene et al. 2013). *Wojnowicia viburni* Wijayaw. et al. was transferred to *Wojnowiciella* (Crous et al. 2015a). Presently, seven species are accepted in *Wojnowicia*, including *W. spartii* described in this paper (Sutton 1980, Wijayawardene et al. 2015).

Wojnowicia spartii should be compared with W. dactylidis Wijayaw. et al. and W. lonicerae Wijayaw.et al. However, conidia of W. spartii are shorter than in W. dactylidis (35–40 × 4–5.5 µm) and W. lonicerae (38–49 × 5–6 µm). Wojnowicia spartii has similar conidial dimensions as W. dactylidicola Wijayaw. et al. (25–35 × 3.5–6.5 µm ($\bar{x} = 28.38 \times 4.87 \text{ µm}$), but can be distinguished using septation (7–12 septa in W. spartii and 3–5 septa in W. dactylidicola). Multigene analyses shows that W. spartii is distinct from any other Wojnowicia species (Fig. 1).

Discussion

Many genera of ascomycetes are pleomorphic and propagate through sexual or asexual reproduction in different geographical locations and at different times, and it is often difficult to understand the whole life cycle of a species (Hyde et al. 2011, Wijayawardene et al. 2012). Observation of sporulating structures from single ascospores on artificial media, the occurrence of two morphs on the same substrate, or mating studies was previously the only way to link morphs (Wijayawardene et al. 2014). However, the occurrence of an asexual morph adjacent to the sexual morph on a host cannot equivocally show they are linked; they may be unrelated species. DNA



Fig. 11 – *Wojnowicia spartii* (MFLU 15–0700, **holotype**) a Herbarium specimen. b–c Appearance of black conidiomata on the host. d Vertical section of conidioma. e Ostiole. f Section of peridium. j–i Conidiogenous cells and developing conidia. j Germinating spore. k–n Conidia. o Culture on PDA. – Bars: $b = 200 \ \mu m$; $c = 200 \ \mu m$; $d = 100 \ \mu m$; $e = 20 \ \mu m$; $f = 20 \ \mu m$; $g-i = 5 \ \mu m$; $j-n = 10 \ \mu m$; $o = 25 \ nm$.

sequence analysis and phylogenetic inference can irrefutably establish asexual and sexual morph connections, for example, the *Mycosphaerella arbuticola / Septoria unedonis*, *Phaeosphaeria papaya / Phaeoseptoria oryzae*, *Leptosphaeria maculans / Plenodomus lingam*, and also provide phylogenetic placements for asexual taxa within the modern taxonomic classification schemes (Quaedvlieg et al. 2013; Ariyawansa et al. 2015b, Crous et al. 2015b). In this study, sequence data together with morphology are used to delimit and propose several new species and one new genus in *Phaeosphaeriaceae* and link the sexual and asexual morph of *Nodulosphaeria*.

The observations presented in this study show that the LSU and ITS sequence data can be used to ditinguish most genera in *Phaeosphaeriaceae* such as *Parastagonospora* and *Nodulosphaeria*. Nevertheless, the circumscription of *Septoriella* is still unclear, and our phylogenetic analysis (Fig. 1) suggests that *Septoriella leuchtmannii* should be excluded from the genus. Future studies should use more isolates and gene regions to resolve the genetic concepts of *Septoriella*.

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