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ARTICLE

Ophiobolus hydei sp. nov. (Phaeosphaeriaceae, Ascomycota) from *Cirsium* and *Phlomoides* in Uzbekistan

Yusufjon Gafforov, Rungtiwa Phookamsak, Hong-Bo Jiang, Dhanushka N. Wanasinghe, and Mukhiddin Juliev

Abstract: We introduce a new fungal species, ***Ophiobolus hydei***, from dead stems of *Cirsium alatum* (Compositae) and *Phlomoides brachystegia* (Lamiaceae), based on morphological and phylogenetic evidence. The species was collected from the Mountains of Western Tien Shan and southwestern Hissar in Uzbekistan. *Ophiobolus hydei* is characterized by globose to subglobose ascomata with short to long papilla, cylindrical to subcylindric-clavate asci, broad pseudoparaphyses, and scolecosporous, yellowish-brown to brown, filiform, multiseptate ascospores that can split into several part-spores at the septa. Multigene phylogenetic analyses using a combined gene analysis of ITS, LSU, SSU, and TEF1-α indicated that the new species has a close affinity to *Ophiobolus ponticus*, but differs from that species in the micromorphological characteristics of the ascomata, asci, and ascospores, as well as biogeographic distribution. A distribution map, morphological descriptions, and illustrations with colour photographs of the novel species are provided.

Key words: new species, ascomycetous microfungi, Central Asia, *Compositae*, GIS, multigene, *Lamiaceae*, *Pleosporales*, phylogeny.

Résumé : Les auteurs présentent une nouvelle espèce, ***Ophiobolus hydei***, isolée de tiges mortes de *Cirsium alatum* (Compositae) et de *Phlomoides brachystegia* (Lamiaceae), identifiée sur la base de données morphologiques et phylogénétiques. L'espèce a été récoltée dans les montagnes du Tien Shan de l'Ouest et des monts Hissar du Sud-ouest, en Ouzbékistan. *Ophiobolus hydei* est caractérisé par des ascomes globuleux à subglobuleux avec des papilles courtes à longues, des asques cylindriques à subcylindriques à clavés, de larges pseudoparaphyses et des ascospores scolécosporeuses, de couleur brune jaunâtre à brune, filiformes, multiseptées qui peuvent se fragmenter en plusieurs spores au septum. Les analyses phylogénétiques multigéniques réalisées par la combinaison de l'analyse des gènes de l'ITS, de la LSU, de la SSU et de TEF1-α ont indiqué que cette nouvelle espèce présente une affinité étroite avec *Ophiobolus ponticus* mais qu'elle s'en distingue par les caractéristiques micromorphologiques des ascomes, des asques, et des ascospores, de même que par sa distribution biogéographique. Une carte de la distribution, des descriptions et des illustrations morphologiques et des photographies en couleur de la nouvelle espèce sont présentées. [Traduit par la Rédaction]

Mots-clés : une nouvelle espèce, microchampignon ascomycète, Asie centrale, *Compositae*, GIS, multigénique, *Lamiaceae*, *Pleosporales*, phylogénie.

Introduction

Phaeosphaeriaceae is one of the largest families in Pleosporales, comprising over 60 genera and more than 400 species (Phookamsak et al. 2014, 2017, 2019; Wanasinghe et al. 2018*a*; Wijayawardene et al. 2018; Yang et al. 2019; Bakhshi et al. 2019; Maharachchikumbura et al. 2019; Marin-Felix et al. 2019). Species of this family are saprotrophs, nectrotrophs, or endophytes. Some species, espe-

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cially in their asexual life stages, are reported as important plant pathogens that have been introduced worldwide (Arzanlou and Crous 2006; Schoch et al. 2009; Zhang et al. 2012; Phookamsak et al. 2014; Farr and Rossman 2019).

The genus *Ophiobolus* was established by Riess based on the type species *Ophiobolus disseminans*, which was found on *Cirsium arvense* in 1854 (Reiss 1854), and this genus belongs to the order Pleosporales (Phookamsak et al. 2014, 2017; Ariyawansa et al. 2015). *Ophiobolus* is one of the most species-rich genera of ascomycetous fungi. They are saprotrophic on dead branches and stems of herbaceous flowering plants; their ecological function is that they degrade dead plants. Most species of *Ophiobolus* have been described in the Northern Hemisphere (Holm 1948; Shoemaker 1976, 1984; Barr 1979; Walker 1980; Shoemaker and Babcock 1989; Winter 1886). Recently, some new species of *Ophiobolus* have been described from Italy and Russia (Ariyawansa et al. 2015; Phookamsak et al. 2017, 2019; Tibpromma et al. 2017; Wanasinghe et al. 2018*a*). Currently, 287 species are recognised in worldwide (Index Fungorum 2019).

Species in *Ophiobolus* are characterized by their large or small size ascomata, which are scattered or clustered, semi-immersed, or erumpent through the host tissue, globose with a long cylindrical erumpent beak lined with hyaline periphyses. The asci are numerous, short or long-stalked, cylindrical to cylindric-clavate, pedicellate, eight-spored, usually in linear fascicles, sometimes tetraseriate, and thin or thick-walled at the apex. The ascospores are variously shaped, parallel or spirally arranged in asci, multiseptate, phragmosporous to scolecosporous, elliptical to fusiform, sometimes bearing globose appendages at each end, sometimes with bandlike or cushion-shaped appendages near the first-formed septum, without a refractive sphere near each end, lacking detectable polar germ pores. The ascospores can be separated into part-spores, or may not break into several spores; light brown to dark brown or reddish brown (Holm 1948; Shoemaker 1976; Phookamsak et al. 2017). Recently, Phookamsak et al. (2017) revisited *Ophiobolus* and demonstrated that *Ophiobolus*-like fungi are polyphyletic within Phaeosphaeriaceae. The type, *Ophiobolus disseminans*, had a close alliance with species of *Entodesmium* and *Premilcurensis*, and therefore those species have been synonymized under *Ophiobolus* (Phookamsak et al. 2017). In addition, *Ophiobolopsis*, *Paraophiobolus*, and *Pseudoophiobolus* were introduced to accommodate other remaining *Ophiobolus*-like taxa within Phaeosphaeriaceae (Phookamsak et al. 2017).

The diversity of fungi in Uzbekistan, especially ascomycetous microfungi, has received little attention. Our aim is to improve the knowledge of the Uzbekistan mycota, especially Ascomycetes, and provide a detailed description and illustrations, as well as information on the molecular phylogeny of this group of fungi. Additionally, we aim to revise the taxonomic limits, and obtain more specific information on the species ecology and geographic distribution. Recently, several new genera, species, and new records of ascomycetous microfungi were reported from Uzbekistan (Solieva and Gafforov 2001, 2002; Gafforov 2002, 2015, 2016*a*, 2016*b*, 2017; Gafforov and Rakhimov 2017; Wanasinghe et al. 2017, 2018*a*, 2018*b*; Samarakoon et al. 2018; Pem et al. 2018, 2019*a*, 2019*b*; Hyde et al. 2019).

Materials and methods

Study site

The Tien Shan and Hissar mountain systems occupy 15% of the territory of Uzbekistan. The unique diversity of flora and landscapes makes this mountainous area one of the most biologically interesting places in Central Asia, with a wide range of habitats from xeric and moist forests, to alpine meadows and semi-arid grasslands. These Central Asian Mountains lie at a biological crossroads at the most westerly part of the Himalayan range, and support both Palearctic species and others representative of more southerly subtropical latitudes. The peculiarity of biota is due to its mixed character: Indo-Himalayan, Mongolian, Eurasian, and Mediterranean species are represented, as well as local endemics. In any geographic region, species richness and endemism are always higher in the mountains than on the surrounding plains, first, because mountains serve as refugia during unfavorable climatic epochs, and second, because of greater habitat diversity.

Collection and preservation of samples

During a survey of ascomycetous microfungi in Uzbekistan, several undescribed *Ophiobolus* specimens were collected from dead stems and leaves of *Cirsium* and *Phlomoides* from Compositae and Lamiaceae in the Surkhandaryo and Tashkent provinces of Uzbekistan. The studied specimens were deposited in the Tashkent Mycological Herbarium (TASM; Institute of Botany, Academy of Sciences of Uzbekistan). Herbarium acronyms used in the paper are from *Index Herbariorum* (Thiers 2019, continuously updated).

Morphological examination

Dried specimens were studied following the methods described in Phookamsak et al. (2017). The specimens were brought to the laboratory and examined under an Olympus SZ61 stereomicroscope (Model SZ2-ILST; Olympus, Tokyo, Japan). Micromorphological characters were examined under a Nikon ECLIPSE 80i (Nikon, Tokyo, Japan) compound microscope, and images were captured using a Nikon ECLIPSE 80i compound microscope with a Canon 550D (Tokyo, Japan) digital camera and illustrated using DIC microscopy. All morphological character measurements were made with Tarosoft Image Frame Work (version 0.9.7), and the images used for figures were processed using Adobe Photoshop CS6 Extended (version 10.0; Adobe Systems). Faces of Fungi and MycoBank numbers are provided as outlined in Jayasiri

et al. (2015). The new species is justified based on the recommendations as outlined by Jeewon and Hyde (2016).

DNA extraction, PCR, and sequencing

The genomic DNA was extracted directly from dried ascomata using a DNA extraction kit (E.Z.N.A Fungal DNA Mini Kit, D3390-02; Omega Bio-Tek) following the manufacturer's protocol. The DNA product was kept at 4 °C for DNA amplification and maintained at –20 °C for long term storage. DNA was amplified by polymerase chain reaction (PCR) for four gene regions, namely the 28S large ribosomal subunit (LSU), the 18S small subunit nuclear rDNA, the internal transcribed spacers (ITS1–5.8S–ITS2) and the translation elongation factor 1-alpha (TEF1-α). The LSU gene region was amplified using the primers LR0R and LR5 (Vilgalys and Hester 1990; Rehner and Samuels 1994); the SSU gene region was amplified using the primers NS1 and NS4 (White et al. 1990); nuclear ITS was amplified using the primers ITS5 and ITS4 (White et al. 1990). The TEF1-α gene region was amplified using primers EF1-983F and EF1-2218R (Rehner and Buckley 2005). PCR was carried out in a volume of 25 μL that contained 8.5 μL of sterilized water, 12.5 μL of 2× Power *Taq* PCR MasterMix, 1 μL each of forward and reverse primers (0.2 μmol/L) and 2 μL of DNA template (50 μg). The PCR thermal cycle program for ITS, LSU, SSU, and TEF1-α gene regions were as detailed by Phookamsak et al. (2017). PCR amplification was confirmed on 1% agarose electrophoresis gels containing Safeview DNA stain. The amplified PCR fragments were sent to a commercial sequencing provider (Tsingke Biological Technology Co.).

Sequence alignment and phylogenetic analyses

Consensus sequences were assembled in BioEdit version 7.0.5.2 (Hall 1999) and additional reference sequences were obtained from GenBank (Table 1). Subsequent alignments for each locus (LSU, SSU, TEF1-α, and ITS) were generated with MAFFT version 7 (http://mafft.cbrc.jp/alignment/server/index.html; Kuraku et al. 2013; Katoh et al. 2017), and manually corrected when necessary in BioEdit version 7.0.9 (Hall 1999). Each locus and the concatenated aligned dataset were analysed separately using Maximum Likelihood (ML) and Bayesian Inference (BI). The best-fit models of evolution for the four loci tested (GTR+I+G for all gene regions) were estimated using MrModeltest version 2.3 (Nylander 2004).

BI analyses were performed using MrBayes version 3.2.1 (Ronquist et al. 2012) based on the models selected by the MrModeltest. The Markov Chain Monte Carlo (MCMC) algorithm of six chains was initiated for 2×10^6 generations in parallel from a random tree topology. The trees were sampled every 200th generation. The distribution of log-likelihood scores was examined to determine the stationary phase for each search and to decide whether extra runs were required to achieve convergence, using the program Tracer version 1.5 (Rambaut and Drummond 2007). All sampled topologies beneath the asymptote (10%) were discarded as part of a burn-in procedure; the remaining trees were used for calculating PP in the majority rule consensus tree. Posterior probabilities values of the BI analyses (BYPP) over 0.95 were considered statistically significant. The ML analyses were conducted with RAxML-HPC BlackBox (version 8.2.8) (Stamatakis et al. 2008; Stamatakis 2014) in the CIPRES Science Gateway platform (Miller et al. 2010) using a GTR+I+G substitution model with 1000 bootstrap replicates. The robustness of the analyses was evaluated by bootstrap support (MLBS).

Phylograms were visualized using FigTree version 1.4.0 program (Rambaut 2012) and a phylogenetic tree was edited in Microsoft Office PowerPoint 2007.

Thirty-six taxa are used as ingroup taxa (including our newly generated sequences) and *Phaeosphaeria chiangraina* (MFLUCC 13-0231), *P. oryzae* (CBS 110110), and *P. thysanolaenicola* (MFLUCC 10-0563) were selected as the outgroup taxa in the final dataset. Sequences generated in this study were deposited in GenBank (Table 1) and the final matrices and trees in TreeBASE (accession number: 24465; Study Accession URL: http://purl.org/phylo/treebase/phylows/study/24465).

Reviewer access URL: http://purl.org/phylo/treebase/phylows/study/TB2:S24465?x-access-code=7ee4c384369905d924a11df642a16d50&format=html.

Data compilation for the GIS map

A point distribution map of new species ***Ophiobolus hydei*** (Fig. 1) was produced using ArcGIS 10.3 desktop software. A global navigation system (GPS) navigation device and Google Earth software were used for georeferencing recently collected samples in study sites for *Ophiobolus* specimens. A WGS84 geographic coordinate system was used as a reference datum. Information for *Ophiobolus hydei* such as collection date, location including latitude and longitude, elevation, and host plants are provided. Landsat satellite data was utilized for the Normalized Difference Vegetation Index (NDVI 2019) and the Advanced Spaceborne Thermal Emission and Reflection Radiometer Digital Elevation Model (ASTER DEM 2019) to prepare the elevation map. Using open source datasets is a way to upgrade remote sensing and GIS skills, especially for researchers in developing countries. Landsat optical satellite images and ASTER DEM are frequently used for different research areas (Lu et al. 2004; Juliev et al. 2019).

Results

Phylogenetic inferences

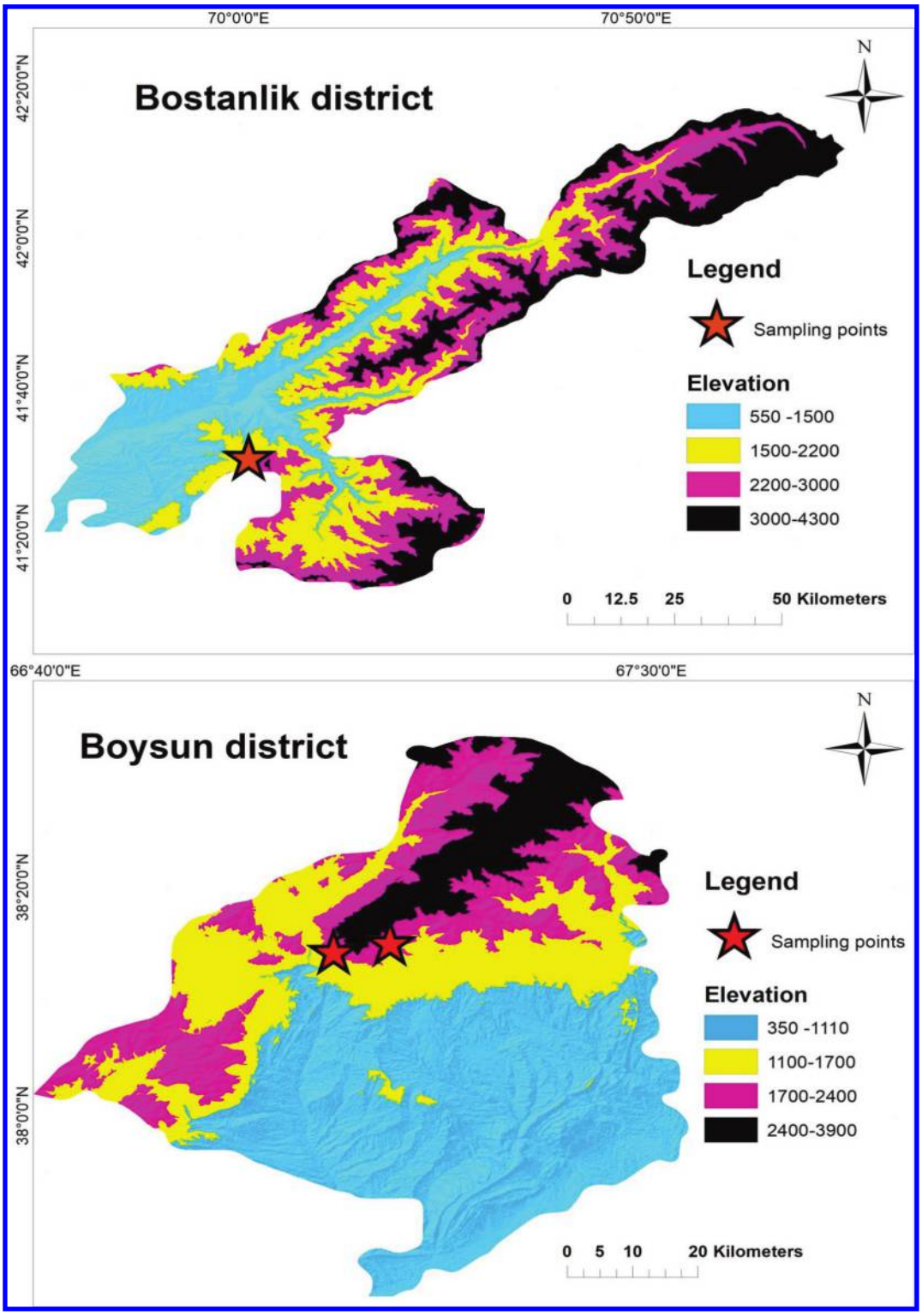
Topologies of trees (under ML and BI criteria) recovered for each gene dataset were visually compared, and the overall tree topology was congruent to those obtained from the combined dataset. The RAxML analysis of the combined dataset yielded a best scoring tree (Fig. 2) with a final ML optimization likelihood value of –11460.866279. The matrix had 636 distinct alignment patterns, with 17.17% proportion of gaps

Table 1. A list of species, specimens, strains, and GenBank accession numbers for the sequences used in the phylogenetic analysis.

Species	Host/Family	Strain/voucher No.	GenBank accession numbers				Reference	Origin
			ITS	LSU	SSU	TEF1-α		
Chaetosphaeronema achilleae	*Achillea nobilis*/Compositae	MFLUCC 16-0476	KX765265	KX765266	KX765265	n/a	Hyde et al. 2016	Russia
C. hispidulum	*Anthyllis vulneraria*/Fabaceae	CBS 216.75	KF251148	EU754144	KF251148	KF253108	Quaedvlieg et al. 2013	Germany
Dematiopleospora cirsii	*Cirsium* sp./Compositae	MFLUCC 15-0615	KX274243	KX274250	KX274243	KX284708	Hyde et al. 2016	Italy
D. fusiformis	*Achillea millefolium*/Compositae	MFLU 15-2133	KY239029	KY239030	KY239029	n/a	Huang et al. 2017	Russia
Dermatiopleospora mariae	*Ononis spinosa*/Fabaceae	MFLUCC 13-0612	KX274244	KJ749653	KJ749652	KJ749655	Wanasinghe et al. 2014	Italy
Dlhawksworthia alliariae	*Alliaria petiolata*/Brassicaceae	MFLUCC 13-0070	KX494876	KX494877	KX494876	n/a	Hyde et al. 2016, Wanasinghe et al. 2018*a*	Italy
D. clematidicola	*Clematis vitalba*/Ranunculaceae	MFLUCC 14-0910	MG828901	MG829011	MG828901	MG829202	Wanasinghe et al. 2018*a*	Russia
D. lonicera	*Lonicera* sp./Caprifoliaceae	MFLUCC 14-0955	MG828902	MG829012	MG828902	MG829203	Wanasinghe et al. 2018*a*	Italy
Muriphaeosphaeria ambrosiae	*Ambrosia artemisiifolia*/Compositae	MFLU 15-1971	KX765267	KX765264	KX765267	n/a	Hyde et al. 2016	Russia
M. galatellae	*Galatella villosa*/Compositae	MFLUCC 14-0614	KT438333	KT438329	KT438331	MG520900	Phukhamsakda et al. 2015	Russia
M. galatellae	*Galatella villosa*/Compositae	MFLUCC 15-0769	n/a	KT438330	KT438332	n/a	Phukhamsakda et al. 2015	Russia
Nodulosphaeria guttulatum	*Scabiosa* sp./Caprifoliaceae	MFLUCC 15-0069	KY496746	KY496726	KY501115	KY514394	Tibpromma et al. 2017	Italy
N. multiseptate	*Sambucus ebulus*/Adoxaceae	MFLUCC 15-0078	KY496748	KY496728	KY501116	KY514396	Tibpromma et al. 2017	Italy
N. scabiosae	*Scabiosa* sp./Caprifoliaceae	MFLUCC 14-1111	KU708850	KU708846	KU708842	KU708854	Mapook et al. 2016	Italy
Ophiobolopsis italica	*Ononis spinosa*/Fabaceae	MFLUCC 17-1791	MG520939	MG520959	MG520977	MG520903	Phookamsak et al. 2017	Italy
Ophiobolus artemisiae	*Artemisia austriaca*/Compositae	MFLU 15-1966	MG520940	MG520960	MG520978	MG520904	Phookamsak et al. 2017	Russia
O. artemisiae	*Artemisia campestris*/Compositae	MFLUCC 14-1156	KT315508	KT315509	MG520979	MG520905	Ariyawansa et al. 2015, Phookamsak et al. 2017	Russia
O. artemisiicola	*Artemisia austriaca*/Compositae	MFLUCC 15-2137	MG828930	MG829039	MG829145	MG829220	Wanasinghe et al. 2018*a*	Russia
O. artemisiicola	*Artemisia santonicum*/Compositae	MFLUCC 15-2140	MG828931	MG829040	MG829146	MG829221	Wanasinghe et al. 2018*a*	Russia
O. disseminans	*Cirsium arvense*/Compositae	MFLUCC 17-1787	MG520941	MG520961	MG520980	MG520906	Phookamsak et al. 2017	Italy
O. italicus	*Lathyrus* sp./Fabaceae	MFLUCC 14-0526	KY496747	KY496727	n/a	KY514395	Tibpromma et al. 2017	Italy
O. hydei	***Cirsium alatum*/Compositae**	**TASM 6143**	**MK981301**	**MK981305**	**MK981303**	**MK993651**	**This study**	**Uzbekistan**
O. hydei	***Cirsium alatum*/Compositae**	**TASM 6144**	**MK981300**	**MK981304**	**MK981302**	**MK993650**	**This study**	**Uzbekistan**
O. malleolus	*Cirsium arvense*/Compositae	MFLUCC 15-1077	MH399730	MH399731	MH399729	n/a	Phookamsak et al. 2019	Russia
O. ponticus	*Centaurea* sp./Compositae	MFLUCC 17-2273	MG520943	MG520963	MG520982	MG520908	Phookamsak et al. 2017	Italy
O. rossicus	*Medicago falcata* subsp. *romanica*/Fabaceae	MFLU 17-1639	MG520944	MG520964	MG520983	MG520909	Phookamsak et al. 2017	Russia
O. rudis	none	CBS 650.86	KY090650	GU301812	n/a	GU349012	Schoch et al. 2009	n/a
O. senecionis	*Senecio* sp./Caprifoliaceae	MFLUCC 13-0575	KT728365	KT728366	n/a	n/a	Tibpromma et al. 2015	Italy
Paraophiobolus arundinis	*Arundo pliniana*/Poaceae	MFLUCC 17-1789	MG520945	MG520965	MG520984	MG520912	Phookamsak et al. 2017	Italy
P. plantaginis	*Plantago* sp./Plantaginaceae	MFLUCC 17-0245	KY797641	KY815010	KY815012	MG520913	Phookamsak et al. 2017	Italy
Pseudoophiobolus achilleae	*Achillea millefolium*/Compositae	MFLU 17-0925	MG520946	MG520966	n/a	n/a	Phookamsak et al. 2017	Italy
P. galii	*Galium* sp./Rubiaceae	MFLUCC 17-2257	MG520947	MG520967	MG520989	MG520926	Phookamsak et al. 2017	Italy
P. italicus	*Onobrychis viciifolia*/Fabaceae	MFLUCC 17-2255	MG520948	MG520968	MG520990	MG520927	Phookamsak et al. 2017	Italy
P. mathieui	*Salvia* sp./Lamiaceae	MFLUCC 17-1785	MG520951	MG520971	MG520991	MG520929	Phookamsak et al. 2017	Italy
Phaeosphaeria chiangraina	*Oryza sativa*/*Poaceae*	MFLUCC 13-0231	KM434270	KM434280	KM434289	KM434298	Phookamsak et al. 2014	Thailand
P. oryzae	*Oryza sativa*/*Poaceae*	CBS 110110	KF251186	KF251689	n/a	n/a	Quaedvlieg et al. 2013	South Korea
P. thysanolaenicola	*Thysanolaena latifolia*/Poaceae	MFLUCC 10-0563	KM434266	KM434276	KM434286	KM434295	Phookamsak et al. 2014	Thailand

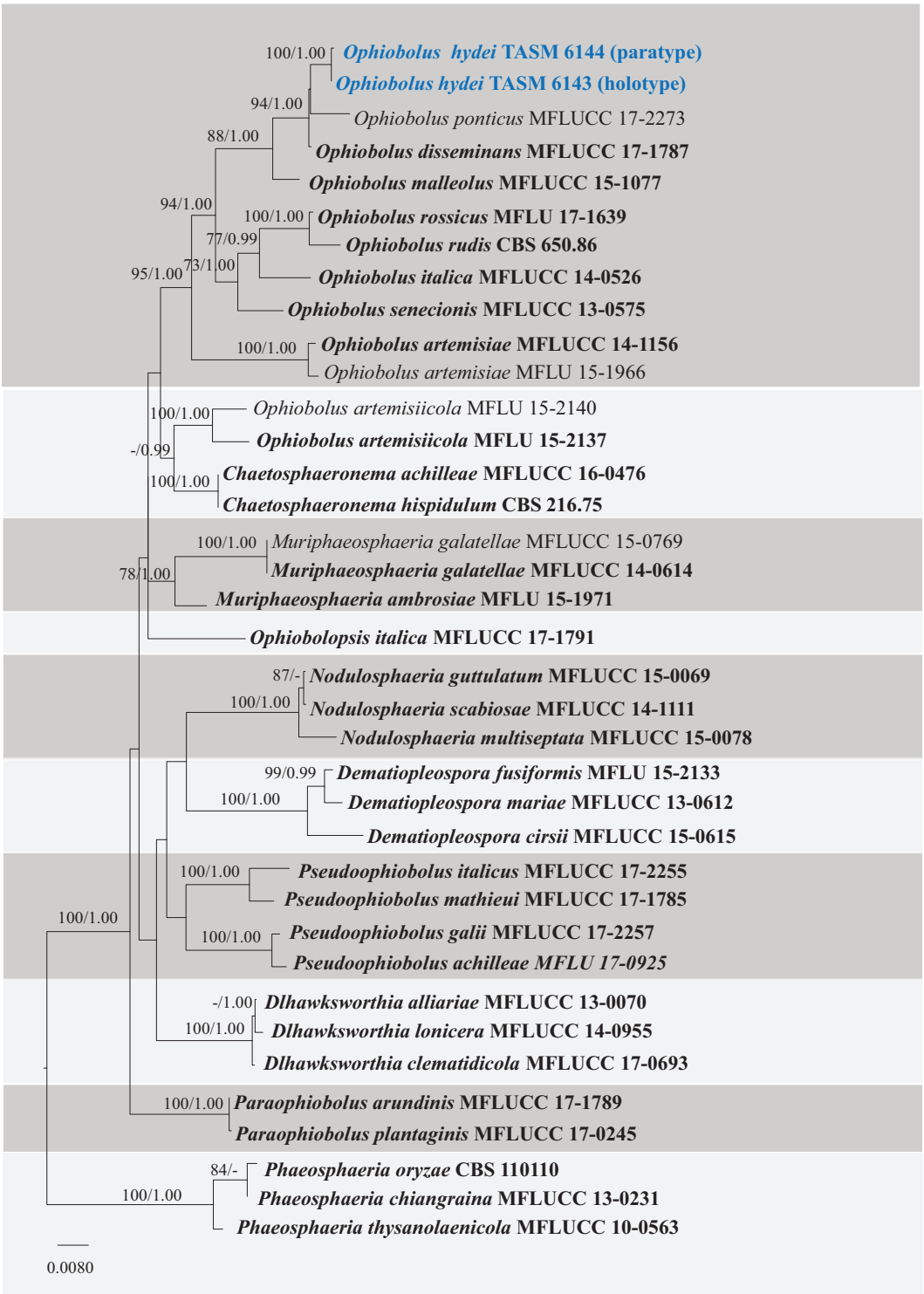
Note: Sequences generated in this study are in bold font.

Fig. 1. Distribution of ***Ophiobolus hydei*** sp. nov. in Uzbekistan. [Colour online.]



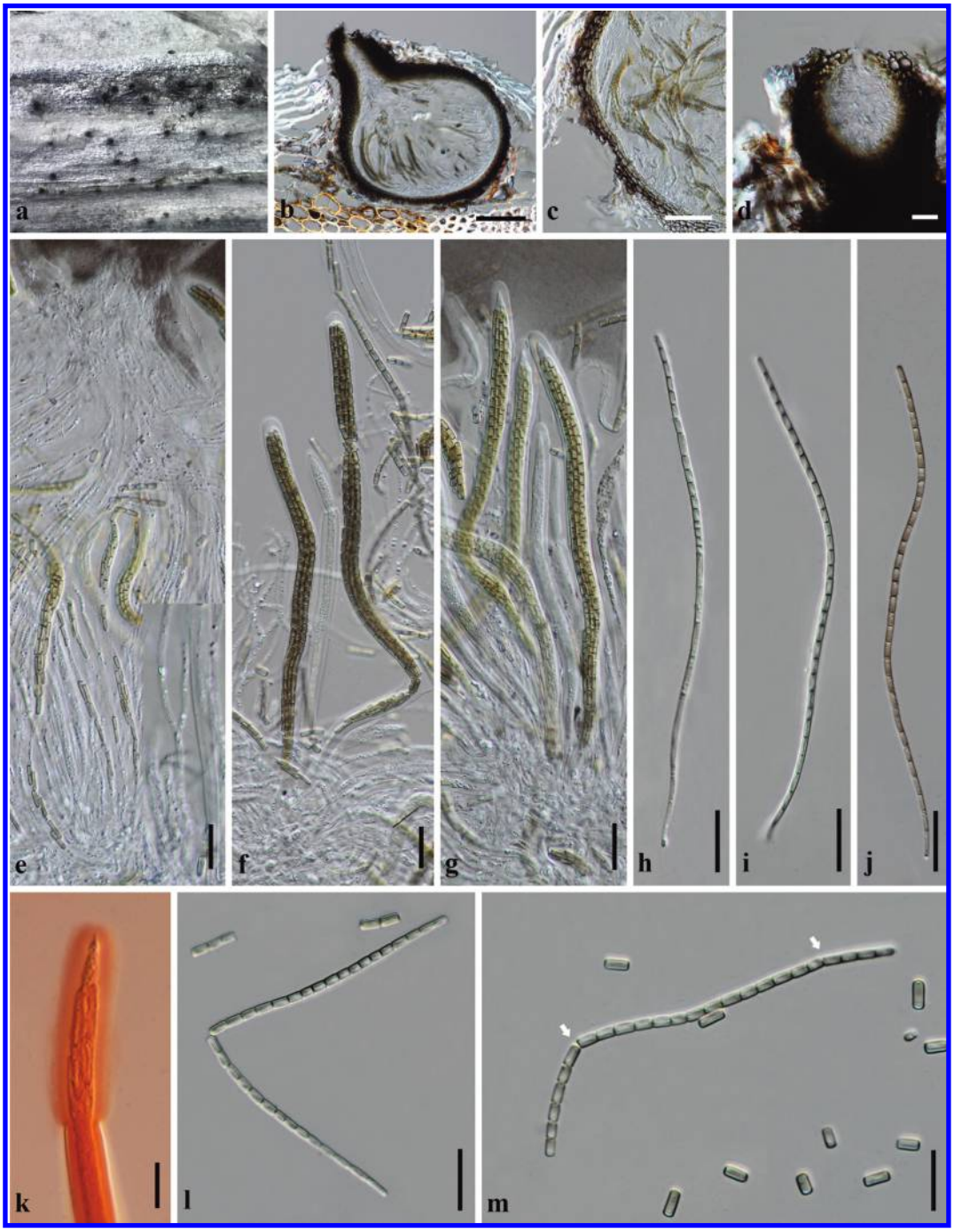
and completely undetermined characters in this alignment. Parameters for the GTR+I+G model of the combined LSU, SSU, TEF1-α, and ITS were as follows: estimated base frequencies: A = 0.243022, C = 0.238618, G = 0.264759, T = 0.253601; substitution rates: AC = 1.084693, AG = 3.131132, AT = 1.890296, CG = 0.922679, CT = 8.054753, GT = 1.000; proportion of invariable sites: I = 0.707451; gamma distribution shape parameter: α = 0.582839. The Bayesian analysis resulted in 10001 trees after 2 M generations. Therefore, the first 1000 trees, representing the burn-in phase of the analyses, were discarded, while the remaining 9001 trees were used for calculating posterior probabilities in the majority rule consensus tree.

Fig. 2. Phylogram generated from maximum likelihood analysis based on the combined LSU, SSU, ITS, and TEF1-α sequence data for *Ophiobolus* species and several related genera in Phaeosphaeriaceae. Bootstrap values for maximum likelihood (ML) equal to or greater than 70% and Bayesian posterior probabilities (BYPP) equal to or greater than 0.95 are listed above the branches, respectively. The newly generated sequences are indicated in blue bold font. Ex-type strains are indicated in black bold font. The tree is rooted with *Phaeosphaeria chiangraina* (MFLUCC 13-0231), *P. oryzae* (CBS 110110), and *P. thysanolaenicola* (MFLUCC 10-0563). [Colour online.]



All available sequence data from *Ophiobolus* species and representative taxa in *Chaetosphaeronema*, *Dematiopleospora*, *Dlhawksworthia*, *Muriphaeosphaeria*, *Nodulosphaeria*, *Ophiobolopsis*, *Paraophiobolus*, and *Pseudoophiobolus* were included in our final dataset. All these genera received high statistical support for their phylogenetic lineages (Fig. 2). Species of *Ophiobolus* are monophyletic with good statistical support, and *Ophiobolus hydei* sp. nov. showed a close phyloge-

Fig. 3. ***Ophiobolus hydei*** sp. nov. (TASM 6143; holotype). (*a*) Appearance of ascomata on the host surface. (*b*) Vertical section through ascoma. (*c*) Section through peridium of textura angularis to textura globulosa. (*d*) Ostiolar canal filled with periphyses. (*e*) Pseudoparaphyses embedded in a hyaline gelatinous matrix (stained with Indian ink). (*f* and *g*) Asci (*g*: stained with Indian ink). (*h–j*) Ascospores (*j*: stained with congo red). (*k*) Apical ascus showing fissitunicate and ocular chamber (stained with Congo red). (*l* and *m*) Ascospores split into part-spores. Scale bars: *b* = 100 μm; *c* = 50 μm; *d–j* and *l* = 20 μm; *k* and *m* = 10 μm. [Colour online.]



netic affinity to *O. disseminans* (MFLUCC 17-1787), *O. malleolus* (MFLUCC 15-1077), and *O. ponticus* (MFLUCC 17-2273).

Taxonomy

Ophiobolus hydei Gafforov, Phookamsak & Wanas, sp. nov. (Fig. 3)

MYCOBANK: MB831142.

ETYMOLOGY: *hydei* (Lat.): in honour of the international mycologist Kevin D. Hyde for his contribution to the taxonomy and phylogeny of ascomycetous fungi.

HOLOTYPE: UZBEKISTAN, Surkhandaryo province, Boysun district, Qizilnaur village, right slope of the Baysuntau Mountains, on a dead stem of *Cirsium alatum* (S.G.Gmel.) Bobrov (Compositae), coll. 14 May 2016, *Y. Gafforov*, YG-

S82-2 (holotype TASM 6143); Surkhandaryo province, Qizilnaur village, in southwestern spurs of the Hissar Range, on dead stem and leaves of *Cirsium alatum*, coll. 14 May 2016, *Y. Gafforov*, YG-S81-1 (paratype TASM 6144).

DESCRIPTION: Saprobic on dead stems of *Cirsium* and *Phlomoides* plant species from Compositae and Lamiaceae, respectively. *Sexual morph*: **Ascomata** 270–450 μm long (excluding papilla), 290–345 μm in diameter, scattered or often grouped under the host tissues, solitary, immersed in host tissue with long neck, visible as black dots when immature, in groups on the host surface, dark brown to black, globose to subglobose, ostiole lined with hyaline periphyses, coriaceous, papillate. **Papilla** 125–160 μm long, 70–160 μm in diameter, with rounded to truncate apex, dark brown to black. **Peridium** 18–33 μm wide, thick-walled cells, pseudoparenchymatous cells, arranged in textura angularis to textura globules, 8.5–10 μm in diameter, comprising 3–5 layers to multilayered, of thin, flattened, dark-brown cells. **Hamathecium** comprising numerous, long, anastomosed pseudoparaphyses embedded in a hyaline gelatinous matrix, septate, 1.2–2.8 μm wide, filamentous, embedded in mucilage. **Asci** (123–)162–258(–290) μm × (7.3–)8.3–12.5 μm ($\bar{x}$ = 210 × 10.2 μm, n = 20), numerous in a broad hymenium, 8-spored, sometimes ascospores overlapping in asci, bitunicate, fissitunicate, cylindrical to subcylindrical-clavate, pedicellate, apically rounded with a well-developed, apical ascus. **Ascospores** fasciculate, scolecosporous, filiform, (110.5–)162–258(–264.2) μm × (1.7–)1.9–2.8(–3.2) μm ($\bar{x}$ = 137 × 2.5 μm, n = 20), hyaline when immature, then becoming slightly pale brown, and becoming brown to yellowish brown at full maturity, multiseptate up to 30-septate, split into part-spores at the septa, with rounded ends, smooth-walled, without sheath, without appendages. **Asexual morph**: Undetermined.

OTHER SPECIMENS EXAMINED: UZBEKISTAN, Tashkent Province, Bustonliq District, Ugam-Chatkal State National Nature Park, Beldersay, the Greater Chimgan Mountain, Chatkal Range of Western Tien Shan Mountains, on dead stems of *Phlomoides brachystegia* (Bunge) Adylov, Kamelin et Makhm. (Lamiaceae), 7 May 2016, *Y. Gafforov*, YG-B9-2 (TASM 6145).

KNOWN DISTRIBUTION AND ECOLOGY: northeast and southern Uzbekistan (Tashkent and Surkhandaryo Province). The new species was collected in two different localities in the mountain systems of Western Tien Shan and Pamir-Alay, and we provided a distribution map for new species in Uzbekistan (Fig. 1). The new species found on two host plant species belonging to two families and two genera. It can be found on other dead plant species from Compositae and Lamiaceae in mountains of Central Asia.

Discussion

Ophiobolus hydei differs from *O. ponticus* by its larger ascomata, with long papilla (125–160 μm long 70–160 μm in diameter) and with rarely truncate apex on host plants surface; larger asci [(123–)162–258(–290) μm × (7.3–)8.28–12.5 μm] and larger ascospores [(110.5–)162–258(–264.2) μm × (1.7–)1.9–2.8(–3.2) μm], and the ascospores can be up to 30-septate and split into several part-spores. *Ophiobolus ponticus* has smaller ascospores [(65–)70–85(–94) μm × (2.5–)3.0–4.5 μm] and the ascospores have 10–12 septa, are swollen at the fourth triangular cell, and do not separate into part-spores (Phookamsak et al. 2017).

Ophiobolus hydei morphologically resembles *O. rossicus* Wanas., Bulgakov., E.B.G. Jones & K.D. Hyde, *O. disseminans* Riess, and *O. rudis* (Riess) Rehm by having a similar shape of ascospores that are broken into pieces (spores) at the septa (up to 30 part-spores). However, these three species differ from *O. hydei* mainly in their smaller ascomata with shorter necks, and smaller asci and ascospores. Multigene phylogenetic analyses of combined LSU, SSU, ITS, and TEF1-α sequence data showed that *O. hydei* clusters with *O. ponticus* and shares a close relationship to *O. disseminans* with support (94% ML and 1.00 PP). However, *O. rudis* and *O. rossicus* are phylogenetically far from *O. hydei*.

Ophiobolus artemisiae (S. Konta, Bulgakov and K.D. Hyde) Wanas., Phookamsak and K.D. Hyde resembles *O. hydei* in morphology, but *O. artemisiae* has smaller ascospores (80–140 μm × 3–5 μm) that do not break into part-spores, and lack a swollen cell; it is saprobic on *Artemisia* species (Phookamsak et al. 2017). In addition, *O. hydei* and *O. artemisiae* are not closely related in the phylogenetic analysis (Fig. 2).

Ophiobolus was previously known from Europe and North America. Regarding the ecological aspects, species of *Ophiobolus* have been reported on various representatives of Caprifoliaceae, Compositae, Fabaceae, and Lamiaceae (Table 1): species of *Artemisia*, *Astragalus*, *Cirsium*, *Coronilla*, *Lathyrus*, *Medicago*, *Senecio*, and *Vicia* (Holm 1948; Shoemaker 1976, 1984; Ariyawansa et al. 2015; Phookamsak et al. 2017, 2019; Tibpromma et al. 2017; Wanasinghe et al. 2018*a*; Farr and Rossman 2019). However, we found *Ophiobolus hydei* on *Cirsium alatum* and *Phlomoides brachystegia* from Compositae and Lamiaceae, respectively, in the Uzbekistan. These two host plants are new substrates for species of the genus *Ophiobolus*. Therefore our new species from Uzbekistan represents a significant addition to both the biogeography and host range of *Ophiobolus* species. A systematic survey of ascomycetes in Central Asia is needed. Given the special geographic position of this central region of Eurasia into consideration, this kind of survey would greatly improve our knowledge on the worldwide diversity of ascomycetous microfungi.

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