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Dematiopleospora mariae gen. sp. nov., from *Ononis spinosa* in Italy

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Abstract – *Dematiopleospora mariae,* isolated from *Ononis spinosa* collected in Forlì-Cesena Province in Italy, is introduced as a new ascomycete genus and species based on morphology and combined LSU and SSU sequence analyses. Phylogenetic analysis based on maximum parsimony (MP), maximum likelihood (ML) and Mr Bayes all support *Dematiopleospora* as being a distinct genus within the *Phaeosphaeriaceae. Dematiopleospora* is distinguished from other genera in this family in having ascospores whose central cells have longitudinal septa with light end cells, ascomata with a thick peridium and necks comprising short, light brown setae. Phylogenetic analysis also separates *Dematiopleospora* from other genera in the *Phaeosphaeriaceae. Dematiopleospora* forms a sister group with *Entodesmium* and *Chaetosphaeronema,* but these three genera are clearly separated in the molecular analysis with relatively high bootstrap support (89% and 99% respectively). The new genus is compared with similar genera of *Phaeosphaeriaceae* and a comprehensive description, and micrographs are provided.

***Phaeosphaeriaceae*/ LSU / new genus/ new species / SSU**

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INTRODUCTION

With an estimated 19,000 species (Kirk *et al*. 2008, Hyde *et al*. 2013) the Dothideomycetes is the largest class in Ascomycota. Known earlier as Loculoascomycetes (Nannfeldt 1932, Luttrell 1955, 1973, Barr 1987), this class is characterized by bitunicate, usually fissitunicate asci (Ariyawansa *et al.* 2013, Schoch *et al.* 2009) and is made up of species from ecologically diverse habitats, occurring as endophytes, pathogens or epiphytes on living plants, and saprobes on dead plant parts. Some are coprophilous and others lichenized or lichenicolous (Schoch *et al*. 2006, Ruibal 2009, Hyde *et al*. 2013).

There have now been several studies using multigene phylogeny providing the groundwork towards a natural classification of Dothideomycetes (Boonmee *et al*. 2011, 2012, Chomnunti *et al*. 2011, 2014, Liu *et al*. 2011, 2012, Schoch *et al*. 2009b, Nelsen *et al*. 2009, 2011b, Zhang *et al*. 2011, 2012, Hyde *et al*. 2013). The most recent arrangement of Dothideomycetes is that of Hyde *et al*. (2013) which included 22 orders incorporating 105 families (249 genera) and a further 23 families which were placed in Dothideomycetes, families *incertae sedis*.

Pleosporales is the largest order of Dothideomycetes (Kirk *et al*. 2008). Molecular data confirmed that *Pleosporales* should comprises 20 accepted families (Boehm *et al.* 2009a, b; Mugambi and Huhndorf 2009a, Hyde *et al*. 2013, Schoch *et al.* 2009b, Shearer *et al.* 2009, Suetrong *et al.* 2009; Tanaka *et al.* 2009, Zhang *et al.* 2009a, b). One of the most speciose families of *Pleosporales* is *Phaeosphaeriaceae* and was introduced by Barr (1979) and currently contains 26 accepted genera (Hyde *et al*. 2013).

The *Phaeosphaeriaceae* was introduced to accommodate some pleosporalean genera that have saprobic, parasitic or hyperparasitic lifestyles and have small to medium-sized, subglobose or conical ascomata, bitunicate asci and hyaline or pigmented ascospores with or without septation (Zhang *et al.* 2012, Barr 1979).

The aim of this paper is to introduce a new genus and species in *Phaeosphaeriaceae* which was discovered as a result of examining saprobic fungi on *Ononis spinosa* L. in Italy. Combined gene (LSU and SSU rDNA) analyses using maximum-likelihood (ML), maximum-parsimony (MP) and MrBayes clearly showed this species groups in *Phaeosphaeriaceae* with high statistical support.

MATERIALS AND METHODS

Sample collection and morphological studies

Samples were collected on 23 May 2013 in Premilcuore (Province of Forlì-Cesena [FC]), Italy and brought to the laboratory in Zip lock plastic bags. Specimens were examined under a Motic SMZ 168 Series stereomicroscope to establish the fungal morphology. Ascomata were removed using a needle, placed in a droplet of distilled water on a clean slide and covered by a cover slip. Ascomata were squashed and examined under a compound microscope (Nikon Eclipse E600 DIC microscope and a Nikon DS-U2 camera or a Nikon Eclipse 80i

compound microscope fitted with a Canon 450D digital camera) to observe the morphological features.

Single ascospore isolation was carried out following the method described in Chomnunti *et al*. (2014). Germinating spores were transferred aseptically to malt extract agar (MEA) plates and grown at 18°C. Colony colour and other characters were observed and measured after a week and again after 1 month. The specimens are deposited in the Mae Fah Luang University (MFLU) Herbarium, Chiang Rai, Thailand with isotypes in Kunming Institute of Botany Herbarium (KUN), Heilongtan, Kunming, China. Living cultures are also deposited at the Culture Collection at Mae Fah Luang University (MFLUCC) and Landcare Research, Auckland, New Zealand (IMCP).

DNA extraction, PCR amplification and sequencing

Genomic DNA was extracted from fresh fungal mycelium grown on MEA media at 18°C for 4 weeks and DNA extraction and PCR reaction were carried out according to Telle and Thines (2008). The amplification of rDNA regions of internal transcribed spacers (5.8S, ITS), small subunit rDNA (18S, SSU), large subunit (28S, LSU) and translation elongation factor 1-alpha gene (TEF 1α) was carried out by using ITS5 and ITS4, NS1 and NS4 (White *et al*. 1990), LROR and LR5 (Vilgalys and Hester 1990) and EF1-983F and EF1-2218R (Rehner, 2001) primers. The procedure of DNA amplification was carried out according to Telle *et al*. (2011) and amplified PCR fragments were sent to a commercial sequencing provider (GATC Biotech, Germany). The nucleotide sequence data acquired were deposited in GenBank (Table 1).

Table 1. Taxa used in the phylogenetic analysis and their corresponding GenBank numbers. The newly generated sequences are indicated in bold

Taxon	*Culture Accession No*	*GenBank Accession*	
		LSU	*SSU*
Bipolaris oryzae Sawada	MFLUCC 10-0694T[1]	JX256381	–
Bipolaris oryzae Sawada	MFLUCC 10-0715T[1]	JX256384	–
Boeremia telephii (Vestergr.) Aveskamp, Gruyter & Verkley	CBS 135415	KF251649	–
Chaetosphaeronema hispidulum (Corda) Moesz	CBS 216.75	KF251652	EU754045
Cochliobolus heterostrophus (Drechsler) Drechsler	CBS 134.39	AY544645	AY544727
Coniothyrium carteri (Gruyter & Boerema) Verkley & Gruyter	CBS 105.91	KF251712	GQ387533
Coniothyrium carteri (Gruyter & Boerema) Verkley & Gruyter	CBS 101633	KF251713	GQ387532
Coniothyrium glycinicola	CBS 124141	KF251714	GQ387537
Cucurbitaria berberidis Fuckel	MFLUCC 11-0384T[2]	KC506793	KC506797
Cucurbitaria berberidis Fuckel	MFLUCC 11-0385T[2]	KC506794	KC506798
Cucurbitaria berberidis Fuckel	CBS 394.84	GQ387605	GQ387544
Cucurbitaria berberidis Fuckel	CBS 363.93	GQ387606	GQ387545
Didymella exigua (Niessl) Sacc.	CBS 183.55T	EU754155	EU754056
Dothidotthia aspera (Ellis & Everh.) M.E. Barr	CPC 12933	EU673276	EU673228

Table 1. Taxa used in the phylogenetic analysis and their corresponding GenBank numbers. The newly generated sequences are indicated in bold *(continued)*

Taxon	*Culture Accession No*	*GenBank Accession*	
		LSU	*SSU*
Dothidotthia symphoricarpi (Rehm) Höhn.	CBS 119687T	EU673273	EU673224
Entodesmium rude Riess	CBS 650.86	GU301812	AF164356
Halojulella avicenniae (Borse) Suetrong, K.D. Hyde & E.B.G. Jones	BCC 20173T	GU371822	GU371830
Halojulella avicenniae (Borse) Suetrong, K.D. Hyde & E.B.G. Jones	BCC 18422T	GU371823	GU371831
Leptosphaeria errabunda (Desm.) Gruyter, Aveskamp & Verkley	CBS 617.75	JF740289	–
Leptosphaeria pedicularis (Fuckel) Gruyter, Aveskamp & Verkley	CBS 390.80	JF740294	–
Leptosphaeria sydowii (Boerema, Kesteren & Loer.) Gruyter, Aveskamp & Verkley	CBS 385.80	JF740313	–
Leptosphaerulina saccharicola Phookamsak, J.K. Liu & K.D. Hyde.	MFLUCC 11-0169T[3]	KF670716	–
Loratospora aestuarii Kohlm. & Volkm.-Kohlm.	JK 5535B	GU301838	GU296168
Melanomma pulvis-pyrius (Pers.) Fuckel	CBS 124080T	GU456323	GU456302
Neosetophoma samarorum (Desm.) Gruyter, Aveskamp & Verkley	CBS 568.94	KF251664	GQ387519
Neosetophoma samarorum (Desm.) Gruyter, Aveskamp & Verkley	CBS 138.96T	KF251665	GQ387517
Neostagonospora caricis Quaedvlieg, Verkley & Crous	CBS 135092T	KF251667	–
Neostagonospora elegiae Quaedvlieg, Verkley & Crous	CPC 16977T	KF251668	–
Ophiosphaerella herpotricha (Fr.) J. Walker	CBS 620.86	DQ678062	DQ678010
Paraphoma radicina (McAlpine) Morgan-Jones & J.F. White	CBS 111.79T	KF251676	EU754092
Paraphoma radicina (McAlpine) Morgan-Jones & J.F. White	CBS 102875	KF251677	EU754091
Parastagonospora avenae (A.B. Frank) Quaedvlieg, Verkley & Crous	CBS 289.69	KF251678	–
Parastagonospora avenae (A.B. Frank) Quaedvlieg, Verkley & Crous	CBS 290.69	KF251679	–
Parastagonospora caricis Quaedvlieg, Verkley & Crous	CBS 135671	KF251680	–
Parastagonospora nodorum (Berk.) Quaedvlieg, Verkley & Crous	CBS 110109	KF251681	EU754076
Dematiopleospora mariae Wanasinghe, Camporesi, E.B.G. Jones & K.D. Hyde	MFLUCC 13-0612	KJ749653	KJ749652
Phaeosphaeria oryzae I. Miyake	CBS 110110T	KF251689	GQ387530
Phaeosphaeria phragmiticola Leuchtm.	CBS 459.84T	KF251691	–
Phaeosphaeria vagans (Niessl) O.E. Erikss.	CBS 604.86	KF251696	–
Phaeosphaeriopsis glaucopunctata (Grev.) M.P.S. Câmara, M.E. Palm & A.W. Ramaley	MFLUCC 13 0220T[4]	KJ522482	KJ522478
Phaeosphaeriopsis triseptata K.M. Thambugala & K.D. Hyde	MFLUCC 13-0271T[4]	KJ522484	KJ522479
Phoma herbarum Cooke	CBS 615.75	KF251715	DQ678014
Pleospora herbarum P. Karst.	CBS 191.86T	DQ247804	DQ247812
Pleospora herbarum P. Karst	MFLUCC 14-0261	KJ790251	KJ90252

Table 1. Taxa used in the phylogenetic analysis and their corresponding GenBank numbers. The newly generated sequences are indicated in bold *(continued)*

Taxon	*Culture Accession No*	*GenBank Accession*	
		LSU	*SSU*
Pyrenochaeta nobilis De Not.	CBS 407.76T	DQ678096	EU754107
Pyrenochaeta nobilis De Not.	CBS 566.75	GQ387616	GQ387555
Pyrenophora phaeocomes (Rebent.) Fr.	DAOM 222769	DQ499596	DQ499595
Scolicosporium minkeviciusii Treigienė	MFLUCC 12-0089T[5]	KF366382	KF366383
Septoria steviae Ishiba, T. Yokoy. & Tani	CBS 120132T	KF251741	–
Setophoma sacchari (Bitanc.) Gruyter, Aveskamp & Verkley	MFLUCC 12-0241T[6]	KJ476147	KJ476149
Setophoma terrestris (H.N. Hansen) Gruyter, Aveskamp & Verkley	CBS 335.87	KF251750	GQ387528
Wojnowicia hirta (J. Schröt.) Sacc.	CBS 295.69	EU754223	EU754124
Wojnowicia viburni Wijayaw., Yong Wang bis & K.D. Hyde	MFLUCC 120733T[7]	KC594287	KC594288
Xenoseptoria neosaccardoi Quaedvlieg, H.D. Shin, Verkley & Crous	CBS 120.43	KF251783	–
Xenoseptoria neosaccardoi Quaedvlieg, H.D. Shin, Verkley & Crous	CBS 128665T	KF251784	–

Abbreviations: **BCC:** Belgian Coordinated Collections of Microorganisms; **CBS:** Centraalbureau voor Schimmelcultures, Utrecht, The Netherlands; **CPC:** Collection of Pedro Crous housed at CBS; **DAOM:** Plant Research Institute, Department of Agriculture (Mycology), Ottawa, Canada; MFLUCC: Mae Fah Luang University CultureCollection, Chiang Rai, Thailand; **T:** ex-type/ex-epitype isolates: **JK:** J. Kohlmeyer;
Sequence data used was published in Gruyter *et al.* 2013, Hyde *et al.* 2013, Quaedvlieg *et al.* 2013 and [1]Manamgoda *et al.* 2012, [2]Doilom *et al.* 2013, [3]Phookamsak *et al.* 2013, [4]Thambugala *et al.* 2014, [5]Wijayawardene *et al.* 2013a, [6]Phookamsak *et al.* 2014, [7]Wijayawardene *et al.* 2013b.

Phylogenetic analysis

The closest taxa to our strain were determined with standard nucleotide blast searches against the nucleotide database in GenBank (http://www.ncbi.nlm.nih.gov/), and recent published phylogenies (Ariyawansa *et al.* 2013, Hyde *et al*. 2013, Phookamsak *et al.* 2013). Combined analysis of LSU and SSU closest relatives in *Coniothyriaceae, Cucurbitariaceae*, *Didymellaceae*, *Dothidotthiaceae*, *Halojulellaceae, Leptosphaeriaceae* and *Pleosporaceae* were used to confirm the phylogenetic placement in suborder Pleosporineae in Pleosporales. *Melanomma pulvis-pyrius* (Pers.) Fuckel was selected as the outgroup taxon. Multiple sequence alignments were generated with MAFFT v. 7.036 (Katoh and Standley, 2013), and adjusted by manually using BioEdit v. 7.2 (Hall, 1999) and ClustalX (Kohli and Bachhawat 2013). Maximum-likelihood (ML) analysis was performed in RAxML (Stamatakis 2006) implemented in raxmlGUI v.0.9b2 (Silvestro and Michalak 2010). Maximum-parsimony (MP) analysis was carried out using PAUP v. 4.0b10 (Swofford 2003). Posterior probabilities (PP) (Rannala and Yang 1996; Zhaxybayeva and Gogarten 2002) were valued by Markov Chain Monte Carlo sampling (BMCMC) in MrBayes v. 3.0b4 (Huelsenbeck and Ronquist 2001). The setup details for the above phylogenetic analysis followed those described in Liu *et al.* (2012), Phookamsak *et al.* (2013) and Hyde *et al.* (2013). Maximum trees were visualized with Tree View (Page 1996).

RESULTS AND DISCUSSION

Phylogenetic analysis

The combined LSU and SSU dataset comprised 58 taxa including our new stain of *Dematiopleospora mariae* with *Melanomma pulvis-pyrius* as the out group taxon (CBS 124080).

This analysis comprised 1968 characters, of which 1708 were constant, 176 parsimony informative and 84 parsimony-uninformative. Six equally parsimonious trees were generated and the first is selected (Figure 1). Bootstrap support (BS) values of ML and MP (equal to or above 50% based on 1000 replicates) are shown on the upper branches. Values of the Bayesian posterior probabilities (PP) from MCMC analyses are shown below the branches. The Kishino-Hasegawa test shows length = 506 steps with CI = 0.579, RI = 0.797, RC = 0.462 and HI = 0.421.

Our strain *Dematiopleospora mariae* (MFLUCC 13-0612) grouped in *Phaeosphaeriaceae*, but separated from the remaining genera of the family in a clade with relatively high bootstrap support (99%, Figure 1).

TAXONOMY

Dematiopleospora Wanasinghe, Camporesi, E.B.G. Jones & K.D. Hyde, *gen. nov.*

Index Fungorum IF550536

Etymology: The generic epithet is from the combination of two words *Dermatio* and *Pleospora* meaning brown spores similar to *Pleospora*.

Type: *Dematiopleospora mariae* Wanasinghe, E. Camporesi, E.B.G. Jones & K.D. Hyde

A genus of *Phaeosphaeriaceae*. *Saprobic* on dead herbaceous branches. Sexual state: *Ascomata* superficial, solitary, scattered, broadly oblong and flattened, dark brown to black, coriaceous, cupulate when dry, ostiolate. *Ostiole* papillate, black, smooth, with ostiolar canal comprising short, light brown setae. *Peridium* thick, comprising 5-6 layers, thick at the sides and thinner at the base, outer layer heavily pigmented, thick-walled, comprising reddish to dark brown cells of *textura angularis*, inner layer composed of hyaline thin-walled cells of *textura angularis*. *Hamathecium* comprising numerous, filamentous, branched septate, pseudoparaphyses. *Asci* 8-spored, bitunicate, fissitunicate, cylindrical to cylindric-clavate, pedicellate, thick-walled at the apex, with minute ocular chamber. *Ascospores* overlapping 1-2-seriate, muriform, ellipsoidal subfusiform, slightly curved, upper part larger than the lower part, 5-9 transversely septate, with 3-6 vertical septa, deeply constricted at the middle septum, initially hyaline, becoming yellowish brown at maturity, ends lighter, conical and narrowly rounded at the ends, without a mucilaginous sheath. Asexual state: Unknown.

Dematiopleospora mariae Wanasinghe, Camporesi, E.B.G. Jones & K.D. Hyde, *sp. nov.* **Fig. 2**

Index Fungorum IF550536

Etymology: The species epithet refers to the departed mother's name of the specimen collector

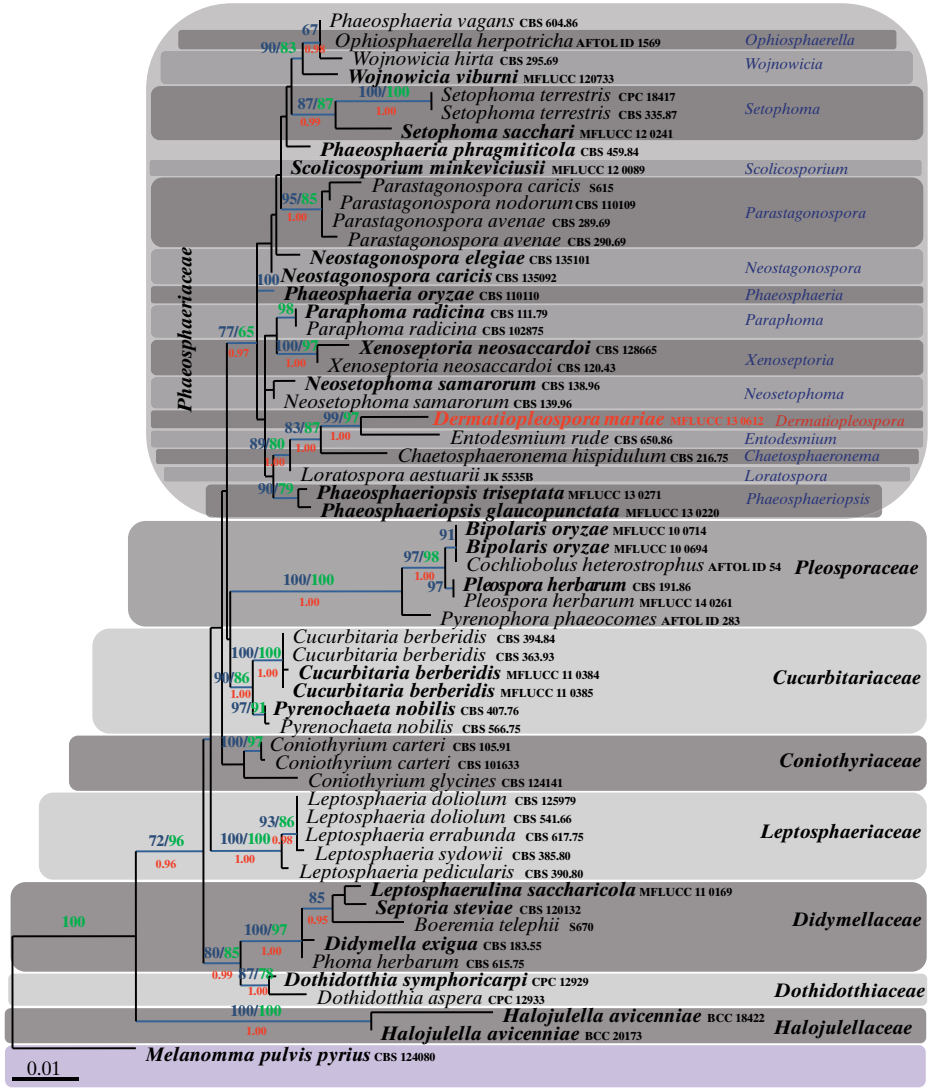

Fig. 1. RAxML tree based on a combined dataset of LSU (910 bp) and SSU (1055 bp) partial equences. Bootstrap support values for maximum parsimony (MP, green) and maximum likelihood (ML, blue) higher than 60% are defined as above the nodes. Bayesian posterior probabilities (BYPP, red) greater than 0.95 are provided below the nodes. The tree is rooted to *Melanomma pulvis-pyrius* (CBS 124080). All ex-type strains are in bold.

Holotype: MFLU 14-0033

Saprobic on dead and hanging branches of *Ononis spinosa*. Sexual state: *Ascomata* 150-210 µm high 200-300 µm diam. ($\bar{x}$ = 186.7 × 233 µm, n = 5) superficial, solitary, scattered, broadly oblong and flattened, dark brown to black, coriaceous, cupulate when dry, ostiolate. *Ostiole* 60-90 µm high 75-90 µm diam. ($\bar{x}$ = 75.3 × 82.4 µm, n = 5) papillate, black, smooth, comprising short, light brown

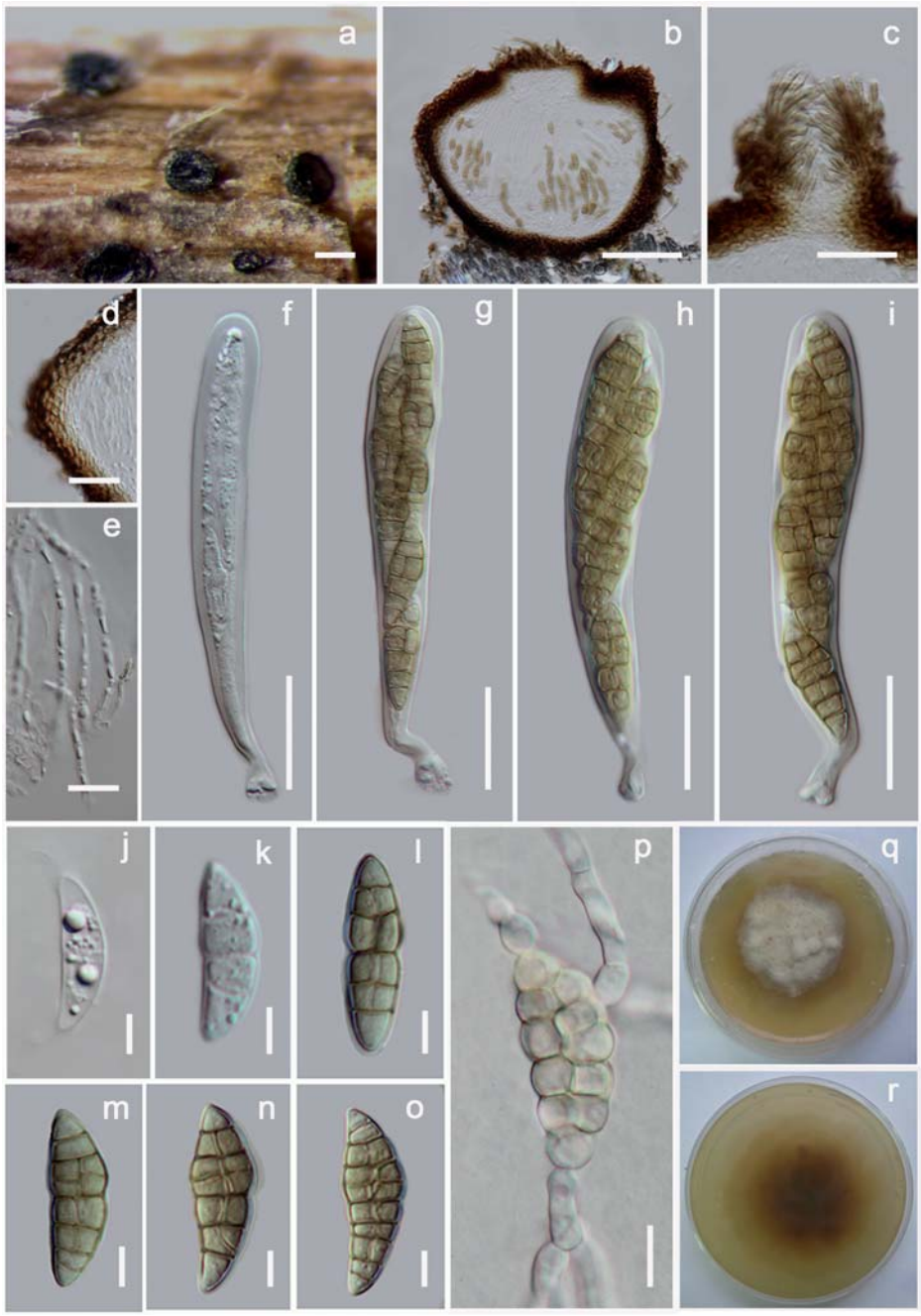

Fig. 2. *Dematiopleospora mariae* (holotype). **a.** Ascomata on host substrate. **b.** Section of ascoma. **c.** Close up of ostiole with short, light brown, setose hyphae. **d.** Peridium. **e.** Pseudoparaphyses. **f-i.** Asci. **j-o.** Ascospores. Note the lighter coloured end cells. **p.** Germinating ascospore. **q,r.** Colonies on MEA (r from below). Scale bars: a = 200 µm, b = 100 µm, c =50 µm, d=30 µm, e=10 µm, f-i=20 µm, j-p=5 µm.

setae. *Peridium* 15-20 µm wide at the base, 11-28 µm wide in sides, thick, with 5-6 layers, outer layer heavily pigmented, thick-walled, comprising reddish to dark brown cells of *textura angularis*, inner layer composed of hyaline thin-walled cells of *textura angularis*. *Hamathecium* comprising numerous, 2.5 µm wide, filamentous, branched, septate, pseudoparaphyses. *Asci* (80-100) × (11-17) µm ($\bar{x}$ = 92.4 × 14.1 µm, n = 30), 8-spored, bitunicate, fissitunicate, cylindrical to cylindric-clavate, pedicellate, thick-walled at the apex, with minute ocular chamber. *Ascospores* (21-27) × (7-9) µm ($\bar{x}$ = 23.8 × 7.8 µm, n = 50), overlapping 1-2-seriate, muriform, ellipsoidal to subfusiform, slightly curved, upper part wider than the lower part, 5-9 transversely septate, with 3-6 vertical septa, deeply constricted at the central septum, initially hyaline, becoming yellowish-brown at maturity, ends remaining lighter and cone-shaped, with rounded ends, without a mucilaginous sheath. Asexual state: Unknown.

Colonies on MEA: slow growing, reaching 1.5 cm diam. after 30 days at 18°C, later with dense mycelium, circular, smooth margin white at first, pinkish ash after 4 weeks (Fig. 2Q-R) flat on the surface, without aerial mycelium. Hyphae septate branched, hyaline, thin, smooth-walled.

Known Distribution: On dead branches of *Ononis spinosa* (Fabaceae) Italy.

Material examined: ITALY, Forlì-Cesena Province: Premilcuore, dead and hanging branches of *Ononis spinosa*, 23 May 2013, E. Camporesi (MFLU 14-0033, **holotype**); (HKAS-81971, **isotype**),— ex-type living culture = MFLUCC 13-0612 = IMCP 20198.

Gene sequence data: ITS (KJ749654), LSU (KJ749653), SSU (KJ749652) and TEF1α (KJ749655).

DISCUSSION

Morphology

Generally phaeosphaeriaceous taxa have cylindrical to fusiform light brown ascospores with transverse septa, and narrow peridia, although muriform ascospores can be seen in two genera (Phookamsak *et al*. 2014, Hyde *et al*. 2013, Zhang *et al*. 2012, Shoemaker and Babcock, 1989). *Dematiopleospora* is characterized by ascospores with light end cells and whose central cells have longitudinal septa, ascomata with a thick peridium and necks with short, light brown apical setae. Our species is similar to *Phaeosphaeria vagans* (Niessl) O.E. Erikss., *P. phragmiticola* Leuchtm., *P. phragmitis* (Hollós) Leuchtm. (Shoemaker and Babcock 1989) and *Pleoseptum yuccaesedum* A.W. Ramaley & M.E. Barr (Zhang *et al*. 2012) in possessing muriform ascospores. However, phylogenetically these species are not closely related (Figure 1) with our new genus/species. Its relationship with *Pleoseptum* cannot be verified as there are no sequences available for comparison, however they are morphologically distinct. In *Pleoseptum yuccaesedum* ascomata are immersed, with a wide peridium of *textura angularis*, a papilla lacking light brown apical setae, and dark brown concolorous ascospores (Zhang *et al*. 2012). *Dematiopleospora* should also be compared with other *Phaeosphaeriaceae* genera, such as *Ammophila,* for which sequence data is lacking. *Amarenomyces ammophilae* (Lasch) O.E. Erikss. which is an intertidal

species on *Ammophila arenaria* (L.) Link has deeply immersed ascomata with a thin peridium, a papilla lacking light brown apical setae, brown ascospores with only transverse septa, and a *Amarenographium* asexual state (Eriksson 1981).

Phylogeny

Phylogenetic analysis (Figure 1) groups *Dematiopleospora* in a clade comprising *Entodesmium rude* Riess, (type species of *Entodesmium*), *Chaetosphaeronema hispidulum* (Corda) Moesz (type species of *Chaetosphaeronema*) and *Loratospora aestuarii K*ohlm. & Volkm.-Kohlm. (type species of *Loratospora*). These genera have significantly different morphologies (Shoemaker 1984, Shoemaker and Babcock 1989, Zhang *et al*. 2012). This clade is an unsupported sister group to *Phaeosphaeriopsis triseptata* K.M. Thambugala & K.D. Hyde and *Phaeosphaeriopsis glaucopunctata* (Grev.) M.P.S. Câmara, M.E. Palm & A.W. Ramaley. *Dematiopleospora* differs from *Entodesmium rude* in having yellowish-brown muriform ascospores without a mucilaginous sheath, whereas *E. rude* has filiform, brown, multi-septate ascospores which break into 22-28 partspores (Zhang *et al*. 2012). *Loratospora aestuarii* differs from *Dematiopleospora* in having hyaline, cylindrical, 3-septate ascospores which are surrounded by a mucilaginous sheath (Zhang *et al*. 2012, Kohlmeyer and Volkmann-Kohlmeyer 1993). *Chaetosphaeronema hispidulum* is an asexual species with no known sexual state and forms a sister group with *Dematiopleospora* with 89% statistical support. *Phaeosphaeriopsis* species differ from *Dematiopleospora* in having cylindrical to fusiform spores with only transverse septation without constriction or slightly constricted at the basal septum and also being surrounded by a mucilaginous sheath (Thambugala *et al.* 2014).

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