Karstenia

ORIGINAL RESEARCH

doi:10.29203/ka.2021.507
Karstenia, Volume 59 (2021), Issue 1–2, pages 13–29
www.karstenia.fi

Fungi causing powdery mildew on plants of a Botanical Garden in Southern Finland

Ville J. Heiskanen[1, 2,*], Jari P.T. Valkonen[1]

[1] Department of Agricultural Sciences, University of Helsinki, Latokartanonkaari 7, FI-00014 Helsinki, Finland
[2] Kumpula Botanic Garden, Jyrängöntie 2, FI-00014 Helsinki, Finland

* Corresponding author:
villejuhaniheiskanen@gmail.com

Keywords: botanic garden, Erysiphales, internal transcribed spacer (ITS), powdery mildew, Southern Finland

Abbreviations: PMCF, Powdery Mildew-Causing Fungi

Article info:
Received: 11 November 2020
Accepted: 05 April 2021
Published online: 07 June 2021
Corresponding Editor: Risto Kasanen

Abstract

Fungi that cause powdery mildew on plants are plant pathogenic parasites (*Erysiphales*) and can significantly reduce the ornamental value of plants and cause significant yield losses among cultivated plants. In this study, 94 plant accessions infected with powdery mildew were observed in Kumpula Botanic Garden, Helsinki, Finland, in 2015. The taxonomic affiliation and species richness of powdery mildew fungi were investigated. Morphological studies by microscope distinguished only 14 fungal species, whereas further comparisons of internal transcribed spacer (ITS) sequences enabled the identification of 28 species. Hence, ITS sequencing improved the reliability of species determination, as compared with the use of morphological characteristics only. The vegetation in an area of six hectares supported a wide range of fungi that cause powdery mildew as well as hyperparasitic microbes, which may balance the impact of pathogens in host plants. The findings of this study emphasize the role of botanical gardens in protecting biological diversity in urban areas.

Introduction

The fungi in the order *Erysiphales* are obligate parasites that cause symptoms of powdery mildew and crop losses in a wide range of cultivated agricultural and horticultural crops, including ornamental plants (Glawe 2008). Globally, ca. 900 powdery mildew–causing fungi (PMCF) have been described (Braun & Cook 2012). However, diversity among PMCF in the Northern Hemisphere is more limited due to the short growing season and early death of host plants due to frost. In fact, diversity of PMCF in the Northern Hemisphere hasn't been studied extensively. Botanical gardens in the Northern Hemisphere offer the possibility for studying the diversity of PMCF in this region, as they contain a wide range of plant species that are adapted to a northern climate. Such studies are relatively rare and have so far focused mainly on single taxa rather than on the analysis of overall diversity of PMCF within a certain area (Korytnanskaya 2010; Mieslerová et al. 2020a; Mieslerová et al. 2020b).

Over 100 PMCF have been identified and are listed in the Finnish Biodiversity Information Facility (Salo et al. 2019), and 256 natural host-fungus combinations associated with powdery mildew have been documented in Finland (Weltzien 1978). Kumpula Botanic Garden of University of Helsinki (subsequently referred to as KBG) hosts a collection of ca. 1500 plant taxa, consisting of plants that have been introduced from different areas of the Northern Hemisphere with bioclimatic habitats resembling Southern Finland (Schulman & Hällfors 2012). Powdery mildew is common and can significantly reduce the ornamental value of plants in the garden. The large and diverse collection of plant species in the garden may host a wide range of non-native PMCF from other countries.

PMCF can be cumbersome to study because hyphae may not be long-lasting, PMCF cannot grow on artificial media and need a living host to survive, and the characteristic structures used for identification by microscopy may be difficult to find and/or maintain. These obstacles can be alleviated by use of DNA-based methods, such as analysis of the internal transcribed spacer (ITS) sequences between the small and large subunit of ribosomal RNA genes. ITS sequences are used as universal barcodes for fungi (Schoch 2012) allowing identification of species based on their ITS sequences and comparisons with those deposited in gene banks (Benson et al. 2013).

The aim of this study was to use morphological characteristics and molecular techniques, such as barcodes based on ITS sequences, to determine the PMCF species infecting plants in KBG, to study species richness of PMCF, and to find out whether the population of PMCF differs from the species previously characterized in Finland.

Materials and methods

Collection of samples for analysis of morphology and ITS sequences

KBG is located at latitude 60.192059(N) and longitude 24.945831(E) in Southern Finland. In total, 94 plant accessions with symptoms of PMCF (**Fig. 1**) were observed during the growth season of 2015 (May 18th to September 30th), and 70 herbarium samples were collected. Plant disease symptoms were surveyed once a week. Samples were taken from symptomatic parts, dried between sheets of blotting paper, and preserved for further examination (**Table 1**). In addition, powdery mildew (mycelia) was scraped off of 42 plants, transferred into Eppendorf tubes, and frozen at -18 °C for DNA analysis.

Morphological examination of fungi

All dried samples from the herbarium were examined and measured with a Researcher Bino II light microscope (Bresser GmbH, Germany) that was equipped with a 3-megapixel camera and MicroCamLab software (v. 7.3.1.8). Fresh mycelia, were transferred onto a microscope slide covered with a piece of adhesive tape and placed on top of a drop of water (Heffer et al. 2006). Characteristics of the mycelium (color, width, growth habit, and shape of appressoria), anamorphs (size and shape of conidia and conidiophores; observation of conidiogenesis) and teleomorphs stages (size and color of chas-

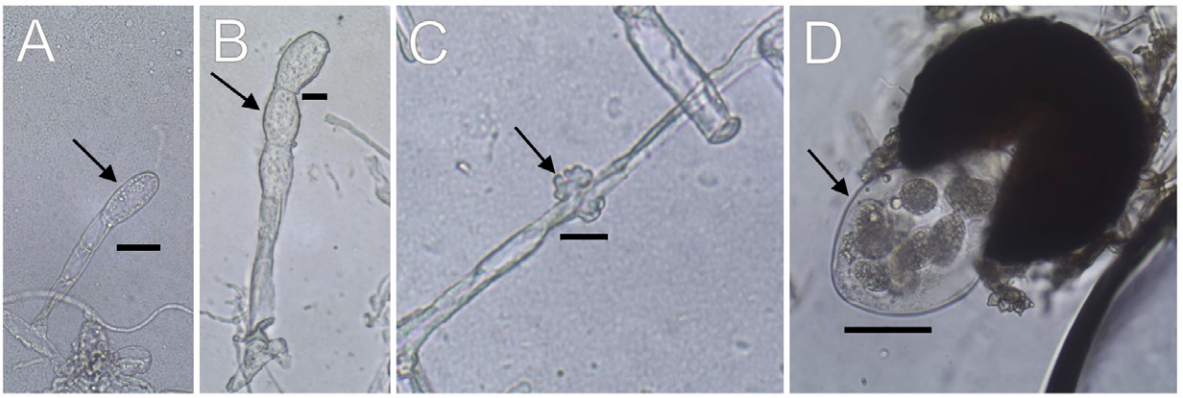


Fig. 1. Symptoms of powdery mildew on different hosts. (a) *Viburnum lantana* (00XX-0153), (b) *Salix caprea* (2010-0939), and (c) *Acer platanoides* (00XX-0004). Numbers in parentheses indicate accession numbers for individual plant specimens.

mothecia; shape of appendages; shape, size, and color of asci and ascospores) were recorded. Species identification of PMCF was based on the descriptions of Braun (1995) and Braun & Cook (2012).

DNA isolation and design of PMCF-specific primers

DNA was extracted from the fungal mycelia using DNeasy Plant Mini Kit (Qiagen GmbH, Hilden, Germany). All 42 samples were screened first by polymerase chain reaction (PCR) using the universal primers ITS5/ITS4 (White et al. 1990) to ensure that samples contained fungal DNA. Subsequently, PMCF-specific primers were designed by downloading rDNA sequences of powdery mildew–causing fungi from NCBI GenBank (Benson et al. 2013): AB022346.1, AB022347.1, AB022349.1, AB022354.1, AB022364.1, AB022365.1, AB022366, AB022369.1, AB022373.1, AB022374.1, AB022398.1, AB022399.1, AB022402.1, AB022404.1, AB022405.1, AB022410.2, AB022411.1, AB022419.1, AB033477.1, AB033479.1, AB033482.1, AB077619.1, AB077671.1, AB080411.1, AB080470.1, AB103069.1, AB103078.1, AB103370.1, AB193465.1, AB237812.1, and AF021796.1. These sequences were aligned with MultAlin software (Corpet 1988) and used to design primers for PCR (18S-2F, 28S-4R, and 28S-6R) and sequencing (5.8S-F, 5.8S-2F, 5.8S-R, and 5.8S-3R) (**Fig. 2**).

PCR reactions

The whole ITS region (ITS1 + 5.8S + ITS2), which included partial 18S and 28S regions (800–900 bp), was amplified by PCR using the primer set 18S-2F/28S-4R or 18S-2F/28-6R (**Fig. 2**). PCR reactions consisted of a final volume of 25 µl containing 1 µl DNA template, 2.5 µl of Optimized 10x DyNAzyme buffer, 0.5 µl of 10 mM dNTPs (Thermo Fisher Scientific Baltics, UAB, Vilnius, Lithuania), 1 µl of 10 µM forward primer (18S-2F), 1 µl of 10 µM reverse primer (28S-4R or 28S-6R), and 0.25 µl of 2 U/µl DyNAzyme II DNA polymerase (Thermo Fisher Scientific Baltics, UAB, Vilnius, Lithuania). Thermal cycling included an initial denaturation at 94 °C for 1 min, followed by 30 cycles of denaturation at 94 °C for 30 s, annealing at 50 °C for 30 s, and elongation at 72 °C for 40 s and a final extension of 8 min at 72 °C. All PCR products were electrophoresed in an agarose gel with a 1% Tris-acetate-EDTA buffer (Sigma-Aldrich Co., St. Louis, MO, USA). Ethidium bromide was added (2.5 mg/ml) to the gel to visualize the PCR products.

Table 1. Ninety-four host plants from KBG (Helsinki, Finland) on which symptoms of powdery mildew were observed and the methods used for sampling.

Host plant a, b	Family	Collected c	Accession
Abelmoschus esculentus	*Malvaceae*		2010-0348
Acer negundo (H) (Amp) (9)	*Sapindaceae*	31.8.	1992-0060
Acer platanoides (H) (Amp)	*Sapindaceae*	30.9.	00XX-0004
Acer platanoides	*Sapindaceae*		00XX-0167
Acer platanoides (H) (Amp)	*Sapindaceae*	30.9.	00XX-0166
Acer platanoides	*Sapindaceae*		00XX-0003
Acer tataricum subsp. *ginnala*	*Sapindaceae*		1994-0891
Acer tataricum subsp. *ginnala* (H) (Amp) (40)	*Sapindaceae*	30.9.	2008-0104
Acer tataricum subsp. *tataricum* (H)	*Sapindaceae*	3.8.	2005-0017
Acer tataricum subsp. *tataricum* (H) (Amp)	*Sapindaceae*	10.9.	2008-0161
Aconitum napellus (H) (13)	*Ranunculaceae*	20.8.	1995-0451
Alchemilla indet.	*Rosaceae*		2008-1029
Arctium lappa var. *edule* (H) (Amp) (25)	*Asteraceae*	20.8.	2010-0296
Avena sativa (H) (37)	*Poaceae*	20.8.	2009-0567
Avena strigosa	*Poaceae*		2014-0350
Brassica oleracea var. *sabellica* (H)	*Brassicaceae*	29.9.	2013-0024
Campanula bononiensis (H)	*Campanulaceae*	30.9.	2012-0735
Centaurea phrygia (H) (Amp) (10)	*Asteraceae*	30.9.	1992-0147
Clematis recta (H) (31)	*Ranunculaceae*	23.9.	2004-0502
Cucumis sativus (H) (35)	*Cucurbitaceae*	20.8.	2013-0042
Cucurbita maxima (H)	*Cucurbitaceae*	23.9.	2015-0123
Cucurbita moschata (H)	*Cucurbitaceae*	23.9.	2015-0125
Cucurbita pepo (H) (Amp)	*Cucurbitaceae*	23.9.	2015-0176
Cucurbita pepo (H)	*Cucurbitaceae*	23.9.	2015-0175
Cucurbita pepo (H) (Amp) (36)	*Cucurbitaceae*	8.9.	2015-0124
Cucurbita pepo (H)	*Cucurbitaceae*	23.9.	2015-0300
Delphinium elatum (H) (38)	*Ranunculaceae*	3.8.	2006-0690
Delphinium elatum (H)	*Ranunculaceae*	31.8.	1995-0091
Delphinium indet. (H) (1)	*Ranunculaceae*	3.8.	2008-0149
Echium maculatum (H) (28)	*Boraginaceae*	3.8.	2012-0627
Echium vulgare	*Boraginaceae*		2011-0721
Euonymus europaeus (H)	*Celastraceae*	3.8.	1992-0099
Euonymus europaeus	*Celastraceae*		1996-0337
Geranium pretense (H) (39)	*Geraniaceae*	10.9.	2001-0068
Geranium sanquineum (H) (7)	*Geraniaceae*	3.8.	1977-0512

Table 1. continues on the next page

Host planta, b	Family	Collected c	Accession
Geum urbanum	*Rosaceae*		1996-0284
Hieracium umbellatum (H)	*Asteraceae*	30.9.	2010-1360
Hordeum vulgare (H)	*Poaceae*	20.8.	2009-0568
Hypericum ascyron (H) (12)	*Hypericaceae*	10.9.	1993-0728
Hypericum maculatum	*Hypericaceae*		2007-0699
Hypericum perforatum (H) (18)	*Hypericaceae*	23.9.	2003-0634
Incarvillea delavayi (H) (26)	*Bignoniaceae*	31.8.	2011-1215
Lonicera tatarica	*Caprifoliaceae*		1997-0172
Monarda didyma (H) (5)	*Lamiaceae*	30.9.	1993-0230
Mycelis muralis (H)	*Asteraceae*	30.9.	2010-1327
Pisum sativum (H)	*Fabaceae*	23.9.	2005-0571
Pisum sativum (H) (27)	*Fabaceae*	8.9.	2012-1008
Pisum sativum (H)	*Fabaceae*	23.9.	2013-0762
Pisum sativum (H)	*Fabaceae*	23.9.	2010-0650
Plantago lanceolata (H) (Amp) (20)	*Plantaginaceae*	20.8.	2006-0888
Plantago major subsp. *major* (H) (Amp) (33)	*Plantaginaceae*	10.9.	2006-0889
Plantago major subsp. *major* (H) (2)	*Plantaginaceae*	20.8.	2008-1028
Pulmonaria montana	*Boraginaceae*		2006-0654
Quercus macrocarpa (H) (15)	*Fagaceae*	30.9.	1997-0448
Quercus macrocarpa (H)	*Fagaceae*	23.9.	1996-0015
Quercus robur (H) (29)	*Fagaceae*	31.8.	00XX-0256
Quercus robur (H)	*Fagaceae*	30.9.	00XX-0035
Rhamnus frangula (H) (Amp) (16)	*Rhamnaceae*	30.9.	1998-0201
Rosa acicularis subsp. *sayi* (H) (6)	*Rosaceae*	3.8.	1969-0232
Rosa amblyotis	*Rosaceae*		1969-0282
Rosa amblyotis	*Rosaceae*		1993-0734
Rosa amblyotis (H) (11)	*Rosaceae*	30.9.	1993-0721
Rosa blanda	*Rosaceae*		1990-0064
Rosa maximowicziana (H) (Amp) (17)	*Rosaceae*	23.9.	1998-0600
Rosa mollis	*Rosaceae*		1994-0443
Rosa nutkana	*Rosaceae*		1992-0531
Rosa villosa	*Rosaceae*		2012-0542
Rosa virginiana (H)	*Rosaceae*	30.9.	1983-0565
Salix caprea (H) (Amp) (30)	*Salicaceae*	3.8.	1987-0943
Salix caprea (H)	*Salicaceae*	23.9.	2010-0939
Salix caprea (H) (Amp)	*Salicaceae*	30.9.	00XX-0095

Host plant a, b	Family	Collected c	Accession
Salix hastata (H) (42)	*Salicaceae*	20.8.	1977-0871
Salix repens subsp. *rosmarinifolia* (H)	*Salicaceae*	31.8.	1980-1929
Salvia tesquicola	*Lamiaceae*		2012-0751
Sambucus racemosa (H)	*Adoxaceae*	23.9.	2010-0946
Sambucus racemosa var. *melanocarpa* (H) (Amp) (41)	*Adoxaceae*	3.8.	1987-1194
Solidago canadensis (H) (Amp) (14)	*Asteraceae*	30.9.	1995-0741
Stachys palustris (H)	*Lamiaceae*	30.9.	2010-1389
Stachys sylvatica (H) (Amp) (21)	*Lamiaceae*	8.9.	2007-0700
Succisa pratensis (H) (Amp) (24)	*Dipsacaceae*	30.9.	2010-1341
Succisa pratensis (H) (19)	*Dipsacaceae*	10.9.	2006-0844
Symphytum officinale var. *bohemicum* (H) (Amp) (8)	*Boraginaceae*	20.8.	1991-0452
Thalictrum aquilegiifolium	*Ranunculaceae*		2013-0911
Thalictrum aquilegiifolium (H) (Amp) (22)	*Ranunculaceae*	23.9.	2008-1011
Thalictrum lucidum (H) (32)	*Ranunculaceae*	8.9.	2006-0610
Tragopogon capitatus	*Asteraceae*		2009-0893
Triticum aestivum	*Poaceae*		2009-0564
Triticum boeticum	*Poaceae*		2015-0361
Triticum compactum	*Poaceae*		2015-0363
Veronica jacquinii (H) (34)	*Plantaginaceae*	23.9.	2012-0760
Veronica longifolia var. *longifolia* (H)	*Plantaginaceae*	30.9.	2010-1391
Veronica spicata (H) (23)	*Plantaginaceae*	8.9.	2010-0870
Viburnum lantana (H) (Amp) (4)	*Adoxaceae*	30.9.	00XX-0153
Vicia sylvatica (H) (3)	*Fabaceae*	8.9.	1991-0137

A samples 1–42 for ITS sequencing. Accession numbers refer to the KBG database (Kotka) and are also used to identify isolates subjected to DNA isolation.

b Hyperparasitic fungus (*Ampelomyces quisqualis*) was observed from the sample with a microscope (Amp).

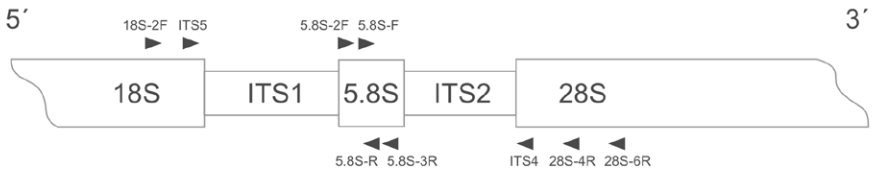
c Dates of collection (in the year 2015) for inclusion in herbarium (H).

Sequencing

PCR products were purified using the E.Z.N.A. Gel Extraction Kit (Omega Bio-Tek, Georgia, USA). The purified DNA products were sequenced by Macrogen Inc. (Amsterdam, The Netherlands). Five samples (isolates 4, 8, 9, 28, and 33) were cloned into pGEM-T Easy vector (Promega Corporation, Madison) using *E. coli* 5α cells and sequenced with universal M13F-pUC primer. This approach was used, because direct sequencing did not produce good-quality sequences (data not shown). All ITS1 and ITS2 sequences were deposited in European Nucleotide Archive (ENA) GenBank (accessions LT794916–LT795001).

Analysis of sequences

All ITS sequences obtained in this study were examined with BioEdit (v. 7.2.5, http://www.mbio.ncsu.edu/BioEdit/bioedit.html) software (Ibis Biosciences, Carlsbad, CA, USA), and all ambiguous regions within the sequences were manually removed. The sequences were processed with the program ITSx (Bengtsson-Palme et al. 2013) to remove conservative rDNA sequences (18S, 5.8S, and 28S) and separate the ITS1 and ITS2 sequences of each isolate. ITS1 and ITS2 of the same sample were joined together using a text editor, followed by comparison with the sequences deposited in GenBank by applying the BLASTn tool (Altschul et al. 1990). Sequences (ITS1 + ITS2) were aligned with AliView software (v. 1.18) (Larsson 2014), and a phylogenetic tree was built using MEGA software (v. 7) (Kumar et al. 2016). The maximum likelihood method with 1000 bootstrap replicates (Felsenstein 1985) was used to estimate branch support. The evolutionary distances were counted with Kimura's two-parameter model (Kimura 1980). The ITS sequences of a closely related fungus, accession no. KT970793 (Baturo-Ciesniewska et al. 2017) from order Helotiales, were used as an outgroup.



PRIMER	PRIMER SEQUENCE (5´→3´)
Forward primers	
18S-2F	TGA ATG GCT AAG TGA GGC
ITS5 *	GGA AGT AAA AGT CGT AAC AAG G
5.8S-F	GTA ATG TGA ATT GCA GAA TTT
5.8S-2F	TAA AAC TTT CAA CAA CGG ATC
Reverse primers	
5.8S-R	GCG CAA TGT GCG TTC AAA G
5.8S-3R	GAA CAG GCA TGC CCC TMG
ITS4 *	TCC TCC GCT TAT TGA TAT GC
28S-4R	CCA GAT TTC AAA TTT GAG CT
28S-6R	AAG CAT CTT CTA CAA ATT ACA

* Universal ITS primers (White et al., 1990)

Fig. 2. Primers used for PCR and sequencing. The approximate binding sites of primers used for amplification of ITS1 and ITS2 regions are shown.

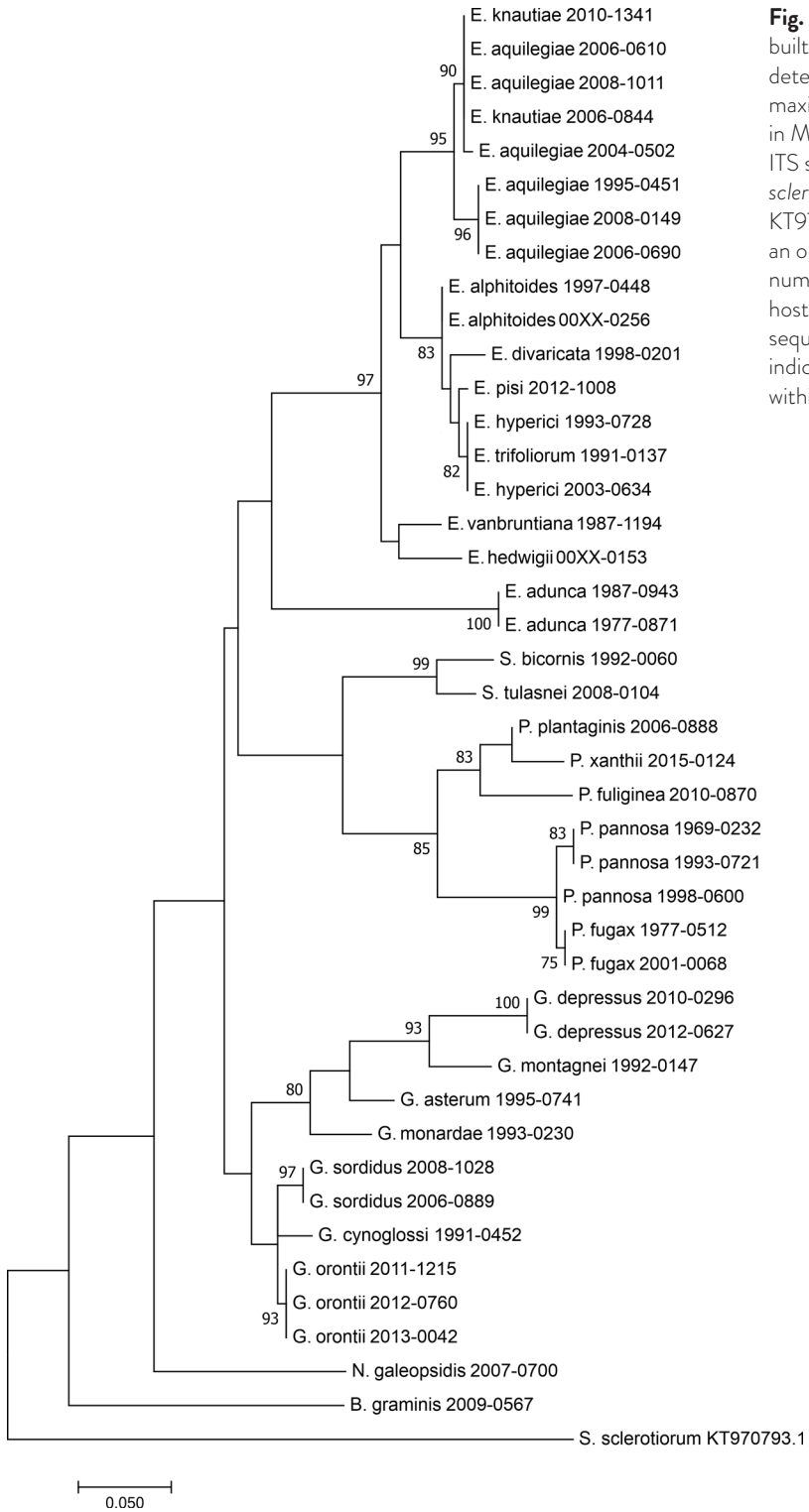


Fig. 3. Phylogenetic tree built from PMCF sequences determined in this study using maximum likelihood method in MEGA 7 software. The ITS sequence of *Sclerotinia sclerotiorum* (accession no. KT970793.1) was chosen as an outgroup. KBG accession numbers are provided for the host plants corresponding to the sequenced specimens. The bar indicates 50 nucleotide changes within 1000 nucleotides.

Results and discussion

Morphological characteristics and phylogenetic analysis

Altogether, 94 observations of powdery mildew were made on different host plants grown in KBG during the growth season of 2015 (**Table 1**), which represented 70 plant species from 24 families. Among the 70 herbarium samples collected, 14 species of PMCF were detected and identified. Their identification was based on morphological characteristics only (Supplementary **Tables 1** and **2**). A hyperparasitic fungus (*Ampelomyces quisqualis* sensu lato) was detected in 23 herbarium samples with a microscope (Table 1). Using the PMCF-specific primers, we were able to amplify rDNA from 37 of the 42 isolates (Table 2). The ITS regions were PCR-amplified from five PMCF samples in which multiple fungi were present and were cloned into pGEM-T Easy vector (Promega Corporation, Madison).

A phylogenetic tree of PMCF including six fungal genera was generated based on the identified sequences (**Fig. 3**) using BLASTn (Altschul et al. 1990). A few isolates lacked polymorphisms within their ITS sequences. They were assumed to be different species based on their minor morphological variations and previous knowledge of their known hosts as mentioned in the literature (Braun 1995; Braun & Cook 2012).

Taxonomy

In this study, 28 species of PMCF from six genera were identified based on their morphological characteristics, ITS sequences, or both (**Table 3**). All PMCF determined in this study were previously known fungi in Finland. One of the identified fungi (*Erysiphe hedwigii*) lacks native hosts in Finland, yet it may thrive on ornamental host plants that are not native to Finland. The results of phylogenetic analyses and species determination based on morphological characteristics were largely consistent. The somewhat ambiguous species are discussed below.

Erysiphe spp

Eleven species of PMCF in the genus *Erysiphe* were identified based on knowledge of their morphological characteristics and analysis of ITS sequences (**Table 3**). A common feature in genus *Erysiphe* is production of a single conidium (**Fig. 4a**), whereas in the majority of other powdery mildew fungi the conidial-producing structures are characterized as catenescent (**Fig. 4b**). The appressoria in *Erysiphe* are typically lobed (**Fig. 4c**), and this feature helps to identify some samples. The fungal species *E. hedwigii* (isolate 00XX-0153) lacks a native host in Finland but may thrive on its ornamental hosts.

Erysiphe aquilegiae sensu lato infects plants within the family *Ranunculaceae* and is found throughout most of the world (Braun & Cook 2012). There are two known variants of the fungus: var. *aquilegiae*, which infects *Thalictrum* and *Clematis*, and var. *ranunculi*, which infects *Aconitum*, *Clematis*, *Delphinium*, and *Thalictrum* (Braun & Cook 2012). In this study, the morphological characteristics were not sufficient to distinguish between these variants on their hosts, but the two clusters of *E. aquilegiae* branches in the phylogenetic tree (**Fig. 3**) support the variants mentioned by Braun & Cook (2012). In contrast, Cunnington et al. (2004) showed that the ITS sequences are not the best candidates for distinguishing between these variants. On the same phylogenetic tree, isolates *Erysiphe knautiae* 2006-0844 and 2010-1341 showed 100% identity with isolates 2006-0610 and 2008-1011 of *E. aquilegiae* and 95% identity with *E. aquilegiae* isolate 2004-0502. Takamatsu et al. (2015) showed that *E. knautiae* belongs to the homogenous clade *E. aquilegia*, which includes at least 15 fungal species that are identical or differ very little among their rDNA sequences. It is assumed that *E. knautiae* does not infect Ranunculaceae hosts but does infect hosts belonging to family *Dipsacaceae* (Braun & Cook 2012). In Finland, *E. knautiae* is known to infect *Knautia arvensis* and *Succisa pratensis* (Braun 1995). The unexpected morphology of the appressoria of *E. knautiae* isolated in this study showed that appressoria in the observed samples were opposite (**Fig. 4c**) rather than single lobed, although they were described as being single lobed by Braun & Cook (2012).

Erysiphe hyperici (Wallr.) S. Blumer and *E. trifoliorum* (Wallr.) U. Braun were identified based on knowledge of their hosts (Braun 1995; Braun & Cook 2012) and ITS sequences. The ITS sequences show

Table 2. The samples identified based on their ITS sequence identities, as compared with previously described fungi using the BLASTn tool.

Fungus	Host plant	Isolate	Sample
Blumeria graminis f. sp. *avenae*	*Avena sativa*	2009-0567	37a
Erysiphe adunca	*Salix caprea*	1987-0943	30a
Erysiphe adunca	*Salix hastata*	1977-0871	42a
Erysiphe alphitoides	*Quercus macrocarpa*	1997-0448	15a
Erysiphe alphitoides	*Quercus robur*	00XX-0256	29a
Erysiphe aquilegiae	*Delphinium indet.*	2008-0149	1a
Erysiphe aquilegiae	*Aconitum napellus*	1995-0451	13a
Erysiphe aquilegiae	*Thalictrum aquilegiifolium*	2008-1011	22a
Erysiphe aquilegiae	*Clematis recta*	2004-0502	31a
Erysiphe aquilegiae	*Thalictrum lucidum*	2006-0610	32a
Erysiphe aquilegiae	*Delphinium elatum*	2006-0690	38a
Erysiphe divaricata	*Rhamnus frangula*	1998-0201	16b
Erysiphe hedwigii	*Viburnum lantana*	00XX-0153	4c
Erysiphe hyperici	*Hypericum ascyron*	1993-0728	12a
Erysiphe hyperici	*Hypericum perforatum*	2003-0634	18a
Erysiphe knautiae	*Succisa pratensis*	2006-0844	19a
Erysiphe knautiae	*Succisa pratensis*	2010-1341	24d
Erysiphe pisi	*Pisum sativum*	2012-1008	27a
Erysiphe trifoliorum	*Vicia sylvatica*	1991-0137	3a
Erysiphe vanbruntiana	*Sambucus racemosa var. melanocarpa*	1987-1194	41a
Golovinomyces asterum var. *solidaginis*	*Solidago canadensis*	1995-0741	14a
Golovinomyces cynoglossi	*Symphytum officinale var. bohemicum*	1991-0452	8c
Golovinomyces depressus	*Arctium lappa*	2010-0296	25a
Golovinomyces depressus	*Echium maculatum*	2012-0627	28c
Golovinomyces monardae	*Monarda didyma*	1993-0230	5a
Golovinomyces montagnei	*Centaurea phrygia*	1992-0147	10a
Golovinomyces orontii	*Incarvillea delavayi*	2011-1215	26e
Golovinomyces orontii	*Veronica jacquinii*	2012-0760	34a
Golovinomyces orontii	*Cucumis sativus*	2013-0042	35a
Golovinomyces sordidus	*Plantago major subsp. major*	2008-1028	2a
Golovinomyces sordidus	*Plantago major subsp. major*	2006-0889	33c
Neoërysiphe galeopsidis	*Stachys sylvatica*	2007-0700	21a
Podosphaera fugax	*Geranium sanguineum*	1977-0512	7a
Podosphaera fugax	*Geranium pratense*	2001-0068	39d
Podosphaera fuliginea	*Veronica spicata*	2010-0870	23a
Podosphaera pannosa	*Rosa acicularis subsp. sayi*	1969-0232	6a
Podosphaera pannosa	*Rosa amblyotis*	1993-0721	11a
Podosphaera pannosa	*Rosa maximowicziana*	1998-0600	17b
Podosphaera plantaginis	*Plantago lanceolata*	2006-0888	20a
Podosphaera xanthii	*Cucurbita pepo*	2015-0124	36a
Sawadaea bicornis	*Acer negundo*	1992-0060	9c
Sawadaea tulasnei	*Acer tataricum subsp. ginnala*	2008-0104	40a

a PCR amplified with primer set 18S-2F/28S-4R and sequenced with primers 5.8SF and 5.8SR
b PCR amplified with primer set 18S-2F/28S-4R and 28S-6R, and sequenced with primers 5.8SF and 5.8S-3R
c PCR amplified with primer set 18S-2F/28S-6R and sequenced with M13F-pUC primer
d PCR amplified with primer set 18S-2F/28S-4R and sequenced with primers 5.8SF and 5.8S-3R
e PCR amplified with primer set 18S-2F/28S-4R and sequenced with primers 5.8S-2F and 5.8S-3R

Accession (ENA)			BLASTn (ITS1 + ITS2)	
ITS1	**ITS2**	**Length (bp)**	**Identity (%)**	**Accession (ENA)**
LT794935	LT794977	324	100	AJ313140.1
LT794916	LT794958	396	100	LC028970.1
LT794917	LT794959	396	100	LC028970.1
LT794918	LT794960	396	100	AB292705.1
LT794919	LT794961	396	100	AB292705.1
LT794922	LT794964	392	99	AY452802.1
LT794920	LT794962	392	99	AB921982.1
LT794923	LT794965	392	100	LC010016.1
LT794921	LT794963	392	99	LC010016.1
LT794924	LT794966	392	100	LC010016.1
LT794925	LT794967	392	100	AY452802.1
LT794926	LT794968	400	99	LC009956.1
LT794934	LT794976	395	100	AF298539.1
LT794928	LT794970	398	100	LC010027.1
LT794927	LT794969	398	100	LC010027.1
LT794929	LT794971	392	100	LC010042.1
LT794930	LT794972	392	100	LC010042.1
LT794931	LT794973	398	100	LC009890.1
LT794932	LT794974	398	100	FJ378884.1
LT794933	LT794975	395	99	LC009909.1
LT794936	LT794978	349	100	KC513763.1
LT794937	LT794979	345	100	AB077684.1
LT794938	LT794980	345	100	AB077675.1
LT794939	LT794981	345	100	AB077675.1
LT794940	LT794982	353	100	LC076842.1
LT794941	LT794983	350	99	AB077656.1
LT794943	LT794985	346	100	AB769464.1
LT794944	LT794986	346	100	AB769464.1
LT794942	LT794984	346	100	AB769464.1
LT794945	LT794987	346	100	AB077658.1
LT794946	LT794988	346	100	AB077658.1
LT794949	LT794991	344	99	KX231842.1
LT794951	LT794993	318	98	AB525922.1
LT794950	LT794992	316	98	AB525922.1
LT794952	LT794994	316	98	AB046986.1
LT794953	LT794995	317	98	AB525937.1
LT794954	LT794996	317	98	AB525937.1
LT794955	LT794997	317	100	KX842352.1
LT794956	LT794998	316	100	JX442063.1
LT794957	LT794999	316	100	KX369541.1
LT794947	LT794989	313	99	AB193380.1
LT794948	LT794990	303	99	AB193385.1

Table 3. Summary of 28 PMCF species identified in the host plant samples collected from KBG and their identification methods. The accession numbers refer to the origin of the samples in the database of the KBG (https://kotka.luomus.fi).

Fungus	Host plant (accession)	Identification	
		Morphology	ITS
Blumeria graminis (DC.) Speer	*Avena sativa* (2009-0567)	×	×
	Hordeum vulgare (2009-0568)	×	
Erysiphe adunca (Wallr.) Fr.	*Salix caprea* (1987-0943)	×	×
	Salix hastata (1977-0871)	×	×
	Salix caprea (00XX-0095)	×	
	S. repens subsp. *rosmarinifolia* (1980-1929)	×	
	Salix caprea (2010-0939)	×	
Erysiphe alphitoides (Griffon & Maubl.) U. Braun & S. Takam.	*Quercus macrocarpa* (1997-0448)	×	×
	Quercus robur (00XX-0256)		×
	Quercus robur (00XX-0035)	×	
Erysiphe aquilegiae DC.	*Aconitum napellus* (1995-0451)	×	×
	Clematis recta (2004-0502)	×	×
	Delphinium elatum (2006-0690)	×	×
	Delphinium indet. (2008-0149)	×	×
	Thalictrum aquilegiifolium (2008-1011)	×	×
	Thalictrum lucidum (2006-0610)	×	×
	Delphinium elatum (1995-0091)	×	
Erysiphe divaricata (Wallr.) Schltdl.	*Rhamnus frangula* (1998-0201)		×
Erysiphe euonymi DC.	*Euonymus europaeus* (1992-0099)	×	
Erysiphe hedwigii (Lév.) U. Braun & S. Takam.	*Viburnum lantana* (00XX-0153)		×
Erysiphe hyperici (Wallr.) S. Blumer	*Hypericum ascyron* (1993-0728)		×
	Hypericum perforatum (2003-0634)		×
Erysiphe knautiae Duby	*Succisa pratensis* (2006-0844)	×	×
	Succisa pratensis (2010-1341)	×	×
Erysiphe pisi DC.	*Pisum sativum* (2012-1008)		×
Erysiphe trifoliorum (Wallr.) U. Braun	*Vicia sylvatica* (1991-0137)		×
Erysiphe vanbruntiana (W.R. Gerard) U. Braun & S. Takam.	*Sambucus racemosa* (2010-0946)	×	
	S. racemosa var. *melanocarpa* (1987-1194)	×	×
Golovinomyces asterum var. *solidaginis* U. Braun	*Solidago canadensis* (1995-0741)		×
Golovinomyces cichoracearum (DC.) V.P. Heluta	*Hieracium umbellatum* (2010-1360)	×	
Golovinomyces cynoglossi (Wallr.) V.P. Heluta	*S. officinale* var. *bohemicum* (1991-0452)	×	×

Fungus	Host plant (accession)	Identification	
		Morphology	ITS
Golovinomyces depressus (Wallr.) V.P. Heluta	*Arctium lappa* (2010-0296)		×
	Echium maculatum (2012-0627)		×
Golovinomyces monardae (G.S. Nagy) M. Scholler	*Monarda didyma* (1993-0230)		×
Golovinomyces montagnei U. Braun	*Centaurea phrygia* (1992-0147)		×
Golovinomyces orontii (Castagne) V.P. Heluta	*Cucumis sativus* (2013-0042)		×
	Incarvillea delavayi (2011-1215)		×
	Veronica jacquinii (2012-0760)		×
Golovinomyces sordidus (L. Junell) V.P. Heluta	*Plantago major* subsp. *major* (2006-0889)		×
	Plantago major subsp. *major* (2008-1028)		×
Neoërysiphe galeopsidis (DC.) U. Braun	*Stachys sylvatica* (2007-0700)	×	×
Podosphaera fugax (Penz. & Sacc.) U. Braun & S. Takam.	*Geranium pratense* (2001-0068)	×	×
	Geranium sanguineum (1977-0512)	×	×
Podosphaera fuliginea (Schltdl.) U. Braun & S. Takam.	*Veronica spicata* (2010-0870)	×	×
Podosphaera pannosa (Wallr.) de Bary	*Rosa acicularis* subsp. *sayi* (1969-0232)		×
	Rosa amblyotis (1993-0721)		×
	Rosa maximowicziana (1998-0600)		×
	Rosa virginiana (1983-0565)	×	
Podosphaera plantaginis (Castagne) U. Braun & S. Takam.	*Plantago lanceolata* (2006-0888)		×
Podosphaera xanthii (Castagne) U. Braun & Shishkoff	*Cucurbita pepo* (2015-0124)		×
Sawadaea bicornis (Wallr.) Homma	*Acer negundo* (1992-0060)		×
Sawadaea tulasnei (Fuckel) Homma	*Acer platanoides* (00XX-0004)	×	
	Acer platanoides (00XX-0166)	×	
	Acer tataricum subsp. *ginnala* (2008-0104)	×	×
	Acer tataricum subsp. *tataricum* (2005-0017)	×	
	Acer tataricum subsp. *tataricum* (2008-0161)	×	

100% similarity among all three isolates (1991-0137 on *Vicia sylvatica*, 1993-0728 on *Hypericum ascyron,* and 2003-0634 on *H. perforatum*) although they were collected from hosts belonging to different plant families. The morphological characteristics were not sufficient to distinguish between species. *E. hyperici* is a common fungus of *Hypericum* species in Asia, the Caucasus, Europe, and North America (Braun & Cook 2012). There are also a few native hosts (*Hypericum* spp.) in Finland (Braun 1995; Mäkinen 1965; Rauhala 1957). In contrast, *E. trifoliorum* is known to infect only Fabacean hosts in Africa, North America, the Caucasus, and Europe. It has spread to South America, Australia, and New Zealand as well (Braun & Cook 2012). The native hosts in Finland for *E. trifoliorum* are *Succisa pratensis* and several *Lathyrus* and *Trifolium* species (Ahti 1967; Braun 1995; Mäkinen 1965; Rauhala 1957). Also, genus *Caragana* is among the host plants in Finland (Braun 1995; Rauhala 1957), but *Erysiphe palczewskii* (Jacz.) U. Braun & S. Takam. is known to have displaced *E. trifoliorum* on *Caragana* host plants in Finland (Huhtinen et al. 2001). Takamatsu et al. (2015) presented the clade *E. trifoliorum* sensu lato, which includes both *E. hyperici* and *E. trifoliorum*.

GOLOVINOMYCES SPP. Eight species of PMCF were identified as *Golovinomyces* with the methods previously mentioned (**Table 3**). A common morphological feature for the fungi in this genus is catenescent conidial formation (**Fig. 4b**). Similar to the *Erysiphe* species, all of the identified *Golovinomyces* species had been previously identified in Finland (Salo et al. 2019). The species *Golovinomyces monardae* (G.S. Nagy) M. Scholler (= *G. biocellatus* Ehrenb.) does not have any native *Monarda* plants as its host in Finland, but it is found on non-native ornamental hosts and on several native or non-native Lamiaceae hosts. In earlier publications, it is usually referred to as *G. biocellatus*, but Scholler et al. (2016) showed in their study that *G. monardae* can be excluded from the wider *G. biocellatus* complex and is classified as its own species.

Golovinomyces depressus (Wallr.) V.P. Heluta was identified in two species (*Arctium lappa* 2010-0296 and *Echium maculatum* 2012-0627) based on ITS sequencing. This may indicate that *Echium maculatum* is a new host for the fungus *G. depressus*, although the morphological characteristics were not enough to confirm this. In addition, pathogenicity was not tested to provide further proof. It is currently understood that *G. depressus* infects *Arctium* and *Centaurea* species in Asia, the Caucasus, Europe, and North America (Braun & Cook 2012), but not Boraginaceae plants such as *Echium* spp. In Finland, the known *G. depressus* observations are from *Arctium*, *Centaurea*, and *Onopordum* acting as host plants (Braun 1995; Rauhala 1957).

BLUMERIA, NEOËRYSIPHE, PODOSPHAERA, AND SAWADAEA SPP. One *Blumeria* sp., one *Neoërysiphe* sp., five *Podosphaera* spp., and two *Sawadaea* spp. were identified (**Table 3**). Among them, *Podosphaera* and *Sawadaea* are the only species that have fibrosin bodies (**Fig. 4e**) inside their conidia, which can be used as a supportive feature for identification based on morphological characteristics. If teleomorphs are present, the species in genus *Podosphaera* are the only ones that have a single ascus inside each chasmothecia (**Fig. 4d**). Among all PMCF, *Blumeria* is the only genus that is capable of infecting monocot plants. *Blumeria* spp. share a common morphological feature of having a bulbous swelling (**Fig. 4f**) that develops from the hyphae and forms the base of the conidium. The species identified in these four genera are PMCF that had been previously identified in Finland (Salo et al. 2019).

Podosphaera fugax (Penz. & Sacc.) U. Braun & S. Takam. was identified in two *Geranium* samples (*G. sanguineum* 1977-0512 and *G. pratense* 2001-0068) based on morphological characteristics as well as ITS sequence similarity. Knowledge of the known hosts of *P. fugax* (Braun & Cook 2012) was also used for species determination. Among ITS sequences from the fungus *Podosphaera pannosa* (Wallr.) de Bary isolated from three *Rosa* spp. (1969-0232, 1993-0721, and 1998-0600) there were differences at only a few nucleotides. All three *P. pannosa* sequences were 317 nucleotides in length but showed minor nucleotide polymorphisms among the analyzed isolates (1969-0232, 1993-0721, and 1998-0600). *P. pannosa* was also identified on a *Rosa virginiana* (1983-0565) herbarium sample based on morphological characteristics. *P. fugax* is known to infect *Geranium* species in Africa, Asia, the Caucasus, Europe, and North America, and it has been introduced into New Zealand (Braun & Cook 2012). *P. fugax* is also a common fungus on *Geranium* spp. in the wild in Finland (Ahti 1967; Braun

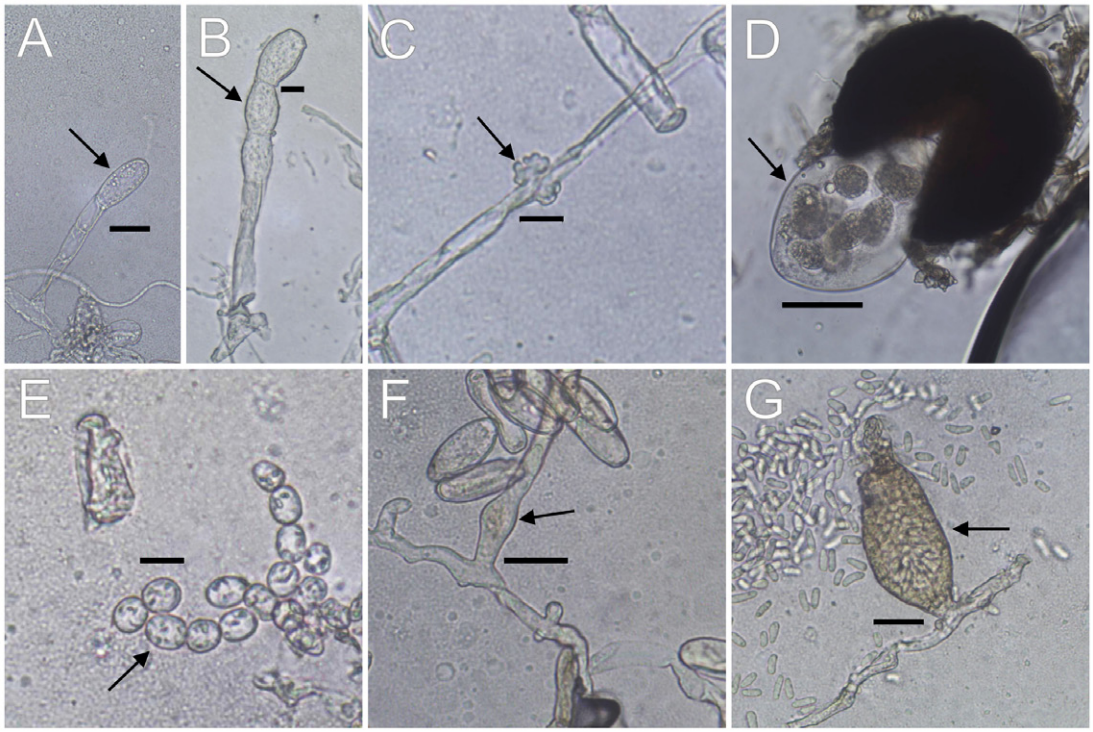


Fig. 4. Morphologically important characteristics of powdery mildew fungi. (**a-g**) The following are examples of structures observed among the 14 morphologically identifiable species of PMCF (host plant accession numbers are included): (**a**) a single conidiospore (1991-0137), bar = 20 µm; (**b**) a catenescent conidial formation (1993-0230), bar = 10 µm; (**c**) a lobed appressorium (2006-0844), bar = 10 µm; (**d**) a chasmothecium and single ascus containing multiple ascospores (1983-0565), bar = 40 µm; (**e**) microconidia and fibrosin bodies inside them (2005-0017), bar = 10 µm; (**f**) a bulbous swelling (2009-0568), bar = 20 µm; and (**g**) a hyperparasite of powdery mildew (1995-0741), bar = 20 µm.

1995; Rauhala 1957). Takamatsu et al. (2010) placed both *P. fugax* and *P. pannosa* in the same Rosoideae group, as they seem to be evolutionarily closely related. *P. pannosa* has spread all over the world (Braun & Cook 2012) and has been found on several *Rosa* spp. in Finland (Braun 1995; Mäkinen 1965; Rauhala 1957).

Podosphaera plantaginis (Castagne) U. Braun & S. Takam. was identified from one plant of *Plantago lanceolata* (isolate 2006-0888) based on ITS sequence similarity. *P. plantaginis* is known to infect plants in Asia, the Caucasus, Europe, and North America (Braun & Cook 2012). The common host for the fungus in Finland is *Plantago lanceolata* (Braun 1995; Rauhala 1957; Jousimo et al. 2014). *Ampelomyces quisqualis* Ces. is a well-studied hyperparasitic fungus that infects *P. lanceolata* (Tollenaere et al. 2014) in Finland. It grows inside the mycelial structures of the powdery mildew fungus (**Fig. 4g**) (Kiss et al. 2004). The ITS sequence of *A. quisqualis* was obtained in this study while sequencing the isolate from *Plantago major* 2006-0889. The *A. quisqualis* ITS sequence was deposited in ENA GenBank (ITS1: LT795000; ITS2: LT795001).

Taken together, the results of this study show that an area of six hectares can support a wide range of fungal populations, such as PMCF, when compatible host plants are present. Furthermore, the hyperparasitic fungus *A. quisqualis* sensu lato was commonly present based on microscope analysis. The 28 PMCF determined in this study are likely to represent just a subset of PMCF in Finland. However, they were isolated by one of the few surveys carried out in a botanical garden using morphological characteristics

and molecular methods for identification. The higher number of PMCF identified with ITS sequences relative to morphological characteristics is supported by a previous study carried out elsewhere (Cunnington et al. 2003). The results of this study also show that botanical gardens maintain not only the genetic diversity of plants but also their pathogens, such as hyperparasitic fungi, which may help to regulate the impact of plants and microbes in gardens.

Acknowledgments

We thank Dr. Marko Hyvärinen and the personnel of Kumpula Botanic Garden for providing advice and space for the study. Financial support from Maiju ja Yrjö Rikala Foundation is gratefully acknowledged.

References

Ahti, T. 1967: Micromycetes (*Peronosporaceae*, *Erysiphales* and *Uredinales*) new to the province of Kuusamo, N. E. Finland. Karstenia 8: 5–8. https://doi.org/10.29203/ka.1968.47

Altschul, S.F., Gish, W., Millew, W., Myers, E.W. & Lipman, D.J. 1990: Basic local alignment search tool. Journal of Molecular Biology 215: 403–410. https://doi.org/10.1016/S0022-2836(05)80360-2

Baturo-Ciesniewska, A., Groves, C.L., Albrecht, K.A., Grau, C.R., Willis, D.K. & Smith, D.L. 2017: Molecular identification of *Sclerotinia trifoliorum* and *Sclerotia sclerotiorum* isolates from the United States and Poland. Plant Disease 101: 192–199. https://doi.org/10.1094/PDIS-06-16-0896-RE

Bengtsson-Palme, J., Ryberg, M., Hartmann, M., et al. 2013: Improved software detection and extraction of ITS1 and ITS2 from ribosomal ITS sequences of fungi and other eukaryotes for analysis of environmental sequencing data. Methods in Ecology and Evolution 4: 914–919. https://doi.org/10.1111/2041-210X.12073

Benson, D.A., Cavanaugh, M., Clark, K., Karsch-Mizrachi, I., Lipman, D.J., Ostell, J. & Sayers, E.W. 2013: GenBank. Nucleic Acids Research 41: D36–D42. https://doi.org/10.1093/nar/gks1195

Braun, U. 1995: The powdery mildews (*Erysiphales*) of Europe. Gustav Fischer Verlag: Jena.

Braun, U. & Cook, R.T.A. 2012: Taxonomic manual of the *Erysiphales* (Powdery mildew), CBS Biodiversity series no. 11, CBS-KNAW Fungal Diversity Centre: Utrecht.

Corpet, F. 1988: Multiple sequence alignment with hierarchical clustering. Nucleic Acids Research 16: 10881–10890. https://doi.org/10.1093/nar/16.22.10881

Cunnington, J.H., Lawrie, A.C. & Pascoe, I.G. 2004: Unexpected ribosomal DNA internal transcribed spacer sequence variation within *Erysiphe aquilegiae sensu lato*. Fungal Diversity 16: 1–10. http://www.fungaldiversity.org/fdp/sfdp/16-1.pdf

Cunnington, J.H., Takamatsu, S., Lawrie, A.C. & Pascoe, I.G. 2003: Molecular identification of anamorphic powdery mildews (*Erysiphales*). Australasian Plant Pathology 32: 421–428. https://doi.org/10.1071/AP03045

Felsenstein, J. 1985: Confidence limits on phylogenies: an approach using the bootstrap. Evolution 39: 783–791. https://doi.org/10.1111/j.1558-5646.1985.tb00420.x

Glawe, D.A. 2008: The Powdery Mildews: a review of the world's most familiar (Yet poorly known) plant pathogens. Annual Review of Phytopathology 46: 27–51. https://doi.org/10.1146/annurev.phyto.46.081407.104740

Heffer, V., Johnson, K.B., Powelson, M.L. & Shishkoff, N. 2006: Identification of Powdery Mildew Fungi anno 2006. The Plant Health Instructor. https://doi.org/10.1094/PHI-I-2006-0706-01

Huhtinen, S., Alanko, P. & Mäkinen, Y. 2001: The invasion history of *Microsphaera palczewskii* (*Erysiphales*) in Finland. Karstenia 41: 31–36. https://doi.org/10.29203/ka.2001.376

Jousimo, J., Tack, A.J.M., Ovaskainen, O., Mononen, T., Susi, H., Tollenaere, C. & Laine, A.L. 2014: Ecological and evolutionary effects of fragmentation on infectious disease dynamics. Science 344: 1289–1293. https://doi.org/10.1126/science.1253621

Kimura, M. 1980: A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. Journal of Molecular Evolution 16: 111–120. https://doi.org/10.1007/BF01731581

Kiss, L., Russell, J.C., Szentiványi, O., Xu, X. & Jeffries, P. 2004: Biology and biocontrol potential of *Ampelomyces* mycoparasites, natural antagonists of powdery mildew fungi. Biocontrol Science and Technology 14: 635–651. https://doi.org/10.1080/09583150410001683600

Korytnanskaya, V.G., Tkachenko, F.P., Tovstuha, N.I. & Rusanov, V.A. 2010: Powdery mildew fungi (*Erysiphales*) of Botanical Garden of Odessa National Mechnikov University. Chornomors'k. bot. z. 6: 259–264. https://doi.org/10.14255/2308-9628/10.62/9

Kumar, S., Stecher, G. & Tamura, K. 2016: MEGA 7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets. Molecular Biology and Evolution 33: 1870–1874.

Larsson, A. 2014: AliView: a fast and lightweight alignment viewer and editor for large data sets. Bioinformatics 30: 3276–3278. https://doi.org/10.1093/molbev/msw054

Mieslerová, B., Kitner, M., Petřeková, V., Dvořáková, J., Sedlářová, M., Cook, R.T.A. & Lebeda, A. 2020a: Golovinomyces powdery mildews on Asteraceae in the Czech Republic. Plant Protection Science 56: 163–179. https://doi.org/10.17221/129/2019-PPS

Mieslerová, B., Sedlářová, M., Michutová, M., Petřeková, V., Cook, R. & Lebeda, A. 2020b: Powdery mildews on trees and shrubs in botanical gardens, parks and urban green areas in the Czech Republic. Forests 11: 967. https://doi.org/10.3390/f11090967

Mäkinen, Y. 1965: On Finnish micromycetes. 7. Recent additions to the distribution of powdery mildews in Finland. Annales Botanici Fennici 2: 243–247. https://www.jstor.org/stable/23724118

Rauhala, A. 1957: Mehltauspilzfunde aus Finnland mit berücksichtigung der bisherigen verbreitungsangaben. Karstenia 4: 14–26. https://doi.org/10.29203/ka.1958.33

Salo, P., Ahti, T. & Salo, V. 2019: *Erysiphales*, in: Suomen Lajitietokeskus 2019: Lajiluettelo 2018. Suomen Lajitietokeskus, Luonnontieteellinen keskusmuseo, Helsingin yliopisto, Helsinki. http://urn.fi/URN:ISSN:2490-0907. Accessed 8 June 2020

Schulman, L. & Hällfors, M. 2011: Botanical garden profile: Kumpula botanic garden, Helsinki, Finland. Sibbaldia: The Journal of Botanic Garden Horticulture 9: 11–28. https://journals.rbge.org.uk/index.php/rbgesib/article/view/119/109

Schoch, C.L., Seifert, K.A., Huhndorf, S., Robert, V., Spouge, J.L., Levesque, C.A., Chen, W. & Fungal Barcoding Consortium 2012: Nuclear ribosomal internal transcribed spacer (ITS) region as a universal DNA barcode marker for Fungi. Proceedings of the National Academy of Sciences of the United States of America 109: 6241–6246. https://doi.org/10.1073/pnas.1117018109

Scholler, M., Schmidt, A., Siahaan, S.A.S., Takamatsu, S. & Braun, U. 2016: A taxonomic and phylogenetic study of the *Golovinomyces biocellatus* complex (*Erysiphales*, *Ascomycota*) using asexual state morphology and rDNA sequence data. Mycological Progress 15: 1–13. https://doi.org/10.1007/s11557-016-1197-5

Takamatsu, S., Ito, H., Shiroya, Y., Kiss, L. & Heluta, V. 2015: First comprehensive phylogenetic analysis of the genus *Erysiphe* (*Erysiphales*, *Erysiphaceae*) I. The *Microsphaera* lineage. Mycologia 107: 475–489. https://doi.org/10.3852/15-007

Takamatsu, S., Niinomi, S., Harada, M. & Havrylenko, M. 2010: Molecular phylogenetic analyses reveal a close evolutionary relationship between *Podosphaera* (*Erysiphales*: *Erysiphaceae*) and its rosaceous hosts. Persoonia 24: 38–48. https://doi.org/10.3767/003158510X494596

Tollenaere, C., Pernechele, B., Mäkinen, H.S., Parratt, S.R., Németh, M.Z., Kovács, G.M., Kiss, L., Tack, A.J.M. & Laine, A.L. 2014: A hyperparasite affects the population dynamics of a wild plant pathogen. Molecular Ecology 23: 5877–5878. https://doi.org/10.1111/mec.12908

Weltzien, H.C. 1978: Geographical distribution of powdery mildews. In: Spencer, D.M. (ed). The powdery mildews, 39–49. Academic Press, London.

White, T.J., Bruns, T.D., Lee, S. & Taylor, J.W. 1990: Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics. In: Innis, M.A., Gelfand, D.H., Sninsky, J.J. & White, T.J. (eds). PCR protocols: A guide to methods and applications, 315–322. Academic Press, London.

SUPPLEMENTARY TABLES

Table S1. Fungal isolates observed at the anamorph stage with a focus on their mycelia and conidia.

Table S2. Fungal isolates observed at the teleomorph stage with a focus on chasmothecia, asci, and ascospores.