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A reexamination and realignment of Peziza sensu lato (Pezizomycetes) species in southern South America

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Abstract

Pfister, D. H.; R. Healy, G. Furci, A. Mujic, E. Nouhra, C. Truong, M. V. Caiafa & M. E. Smith. 2022. A reexamination and realignment of Peziza sensu lato (Pezizomycetes) species in southern South America. *Darwiniana*, nueva serie 10(1): 148-177.

In this study we review recent collections and historical records of epigeous members of the Pezizales formerly placed in the large, heterogenous genus *Peziza* from temperate southern South America. Recent analyses using molecular phylogenetic methods allow placement of these species in several previously described genera in recognition of the heterogeneity of *Peziza*. We include species in nine genera, describe one new species (*Peziza gamundiae* sp. nov.), and propose one new combination (*Phylloscypha nothofageti* comb. nov.). We also demonstrate that *Pustularia microspora* is a synonym of the previously described

taxon *Peziza pseudosylvestris*. Our purpose is to draw attention to these taxa in order to promote their collection and study in a modern framework. **Keywords:** Argentina, Chile, generic assignment, Pezizaceae, phylogeny, subantarctic forests.

Resumen

Pfister, D. H.; R. Healy, G. Furci, A. Mujic, E. Nouhra, C. Truong, M. V. Caiafa & M. E. Smith. 2022. Un reexamen y revisión de las especies de *Peziza* sensu lato (Pezizomycetes) en el cono sur de Sudamérica. *Darwiniana*, nueva serie 10(1): 148-177.

En este trabajo hemos revisado material fresco y colecciones históricas de Pezizales epigeos formalmente incluidos en el amplio y heterogéneo género *Peziza* provenientes de las zonas templadas del sur de Sudamérica. Estudios recientes basados en filogenias moleculares han permitido posicionar estas especies en varios géneros previamente descritos demostrando su heterogeneidad. Incluimos aquí especies de nueve géneros, una especie nueva para la ciencia (*Peziza gamundiae* sp. nov.) y proponemos una nueva combinación (*Phylloscypha nothofageti* comb. nov.). También demostramos que *Pustularia microspora* es sinónimo de un taxón previamente descrito como *Peziza pseudosylvestris*. Nuestro objetivo es llamar la atención sobre la presencia de estos taxa para promover su recolección y estudio en trabajos científicos modernos.

Palabras clave: Argentina, bosque Andino-Patagónico, Chile, delimitación genérica, filogenia, Pezizaceae.

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Introduction

The genus *Peziza* (Pezizaceae, Pezizales, Pezizomycetes) encompasses several often collected, conspicuous species, but the genus has been recognized as representing an heterogeneous assemblage (Hansen et al., 2001; Pfister, 2015). In recognition of this heterogeneity Van Vooren (2020) used molecular characters and morphological characters to segregate various recognized clades (Hansen et al., 2001) into genera. Some of the generic names now used were previously published but were synonymized with *Peziza*; others were newly created by Van Vooren (2020). Van Vooren's work largely focused on species from the northern temperate regions, particularly Europe. In our mycological investigations in the temperate regions of southern South America we have encountered several species of *Peziza* in the

broad sense (*Peziza* sensu lato) that can be assigned to genera as delimited by Van Vooren. The nucleus of the present work are the recent collections for which we have obtained DNA sequences. Analyses of DNA sequences along with morphological examinations have facilitated placement of species in modern genera in consultation with the published literature. We also highlight areas where further study is needed.

The species under consideration display a variety of ecological strategies. Some are saprobes occurring on dung, soil or plant debris. Others form ectomycorrhizae with Nothofagaceae. Recent work on the class Pezizomycetes has revealed a number of the members of the class as endophytic or endolichenic taxa (Healy et al., 2022). To highlight the growing knowledge about the hidden ecological strategies of some of the taxa discussed here, we have included DNA sequences in our phylogenetic analyses of endophytic and endolichenic representatives despite the fact that some of the sequences are derived from specimens from outside southern South America.

Species of *Peziza* sensu lato from southern South America have been documented by Spegazzini, Gamundí and others as summarized by Gamundí et al. (2004). Gamundí's extensive and well-illustrated work on the genus *Peziza* (as *Galactinia* in some publications) and related genera of Pezizales has provided a firm grounding for our studies. She did extensive fieldwork in Argentina that allowed detailed morphological study of specimens comparing collections with those species earlier described by Spegazzini (1887, 1909). Her investigations based on type studies and consultation with experts were well situated in the historical context of earlier workers.

During our field expeditions from 2008-2019 we collected specimens of *Peziza* sensu lato from across southern Chile and Argentina. We observed, photographed, and generated DNA sequences from most collections. The color and form of fresh ascomata can be particularly important for species identifications but photographs of these species have been mostly absent in the published literature. Molecular data have also been mostly missing from southern South American specimens. Based on these recent collections we have provided for the first time DNA sequences for many of the species we discuss. Comparison of our collections with DNA sequences and taxon descriptions from elsewhere in the world give critical insights into the distribution of these fungi.

We address a problematic species, *Pustularia microspora* Raithelhuber (1983). Through morphological and molecular analyses, we show that it is a synonym of the widespread and common *Peziza pseudosylvestris*. A new species, *Peziza gamundiae* is proposed, as is a new combination, *Phylloscypha nothofageti*. In the discussion we take up the broader question of the diversity of the Pezizaceae in the Southern Hemisphere.

Material and methods

Fresh Collections and Fungarium Specimens

Collections used in this study are housed primarily in the fungaria of the University of Florida Herbarium (FLAS-F) and the Farlow Herbarium (FH), Harvard University. The type specimen of *Pustularia microspora* was from the Eidgenössische Technische Hochschule Zürich, Zürich, Switzerland (ZT) with written permission allowing molecular and morphological study. Representative specimens from this study are also deposited at the Herbario del Museo Botánico de Córdoba (CORD) in Córdoba, Argentina and Museo Nacional de Historia Natural de Chile (SGO) in Santiago, Chile.

Fresh specimens were collected in Nothofagaceae forests or in mixed forests from Tierra del Fuego in the south (lat. 56° S) to the Nahuelbuta region in the north (lat. 37° S). In the field, specimens were placed in plastic boxes and transported to a field lab within 8 hours. Specimens were cleaned of debris, photographed while fresh and preserved in CTAB DNA extraction buffer (Gardes & Bruns, 1993) and/or an alkaline extraction buffer (Vandepol et al., 2020). Specimens were dried over low heat in a dehydrator and stored in plastic bags as vouchers for further morphological examination and preservation.

Although the type specimen of *P.pseudosylvestris* was not examined for this study, two internal transcribed spacer (ITS, consisting of ITS1, 5.8S, and ITS2) sequences were available in Genbank from specimens identified as *P. pseudosylvestris* by Irma Gamundí (Lantieri et al., 2016). Both specimens are deposited at the Museum of Evolution in Uppsala, Sweden (UPS). The collections morphologically match the description of *P. pseudosylvestris* by Gamundí (1975) and have almost identical ITS sequences.

Rather than recounting synonymy and information regarding type status for each taxon, we include the Mycobank (mycobank.org) accession number associated with the name under discussion. Herbaria are listed as found in Index Herbariorum (http://sweetgum.nybg.org/science/).

Morphological Analyses

Specimens from Chile and Argentina were examined using light microscopy with morphological characters documented using standard methods (Pfister et al., 2016). Dried material was hand-sectioned with a fresh razor blade and then mounted in water, 3% KOH, cotton blue, or Melzer's reagent. At least 20 measurements of each

morphological feature were recorded in water mounts. Spore measurements exclude ornamentation. We collected and examined both anamorphs and teleomorphs for this work and cite both teleomorph and anamorph collections among the specimens cited. Anamorphs are indicated as such, while all others are teleomorphs. Images were captured using an Olympus BX50 microscope (Tokyo, Japan) with a XC50 digital camera (Olympus, Tokyo, Japan) using the MicroSuite Five software system (Olympus, Tokyo, Japan) or a QImaging MicroPublisher 3.3 RTV digital camera (British Columbia, Canada) mounted on a Nikon Optiphot light microscope (Tokyo, Japan). Images were edited in Adobe Photoshop CS5.1 (San Jose, California). Additional observations were made using a LW Scientific compound microscope and a Zeiss Axio Imager.A2 mounted with a Zeiss Axiocam 305 digital camera (Jena, Germany).

Molecular Protocols

Field-collected material preserved in CTAB and/ or an alkaline extraction buffer were transported to the laboratory for processing. DNA extraction of tissues in alkaline extraction buffer followed the protocols of Vandepol et al. (2020). Tissues in CTAB were extracted using a modified CTAB method (Gardes & Bruns, 1993). Polymerase chain reactions (PCR) of the ITS and 28S rDNA were performed using primer sets ITS1F/ ITS4 (White et al., 1990) for ITS and LROR/LR5 for 28S (Vilgalys & Hester, 1990; Hopple & Vilgalys, 1994) using the Phusion Hot Start Flex DNA Polymerase standard protocol (New England Biolabs, Ipswich, Massachusetts). PCR products were visualized on 1.5% agarose gels stained with SYBR Green I (Molecular Probes, Eugene, Oregon). Amplicons were enzymatically cleaned with EXO (exonuclease I) and AP (Antarctic phosphatase) (New England Biolabs) (Werle et al., 1994) and sequenced by GENEWIZ (South Plainfield, New Jersey). Sequences were then edited with Sequencher 5.0.1 (Gene Codes, Ann Arbor, Michigan).

Phylogenetic analyses

Sequences of epigeous Pezizaceae were obtained from GenBank based on BLAST searches and previous phylogenetic results of Hansen et al. (2002), Lantieri et al. (2016) and Pfister et al. (2016). We separately analyzed one dataset of 28S rDNA Pezizaceae sequences, and four datasets of the ITS rDNA for the species considered in this manuscript. Our 28S alignment was comprised of 136 sequences, including 14 new sequences generated for this study. Our ITS alignments were

comprised of sequences from 178 specimens and 21 sequences from endophyte or endolichenic fungal isolates, including 26 new sequences generated for this study. The datasets were aligned in MAFFT v 7.471 (Katoh & Toh, 2010), and alignments were manually corrected in SeAl v2.0a11 (Rambaut, 2007). Table 1 provides a list of taxa that were analyzed along with their GenBank accession numbers The alignments were analyzed with maximum likelihood (ML) as performed in the Cyberinfrastructure for Phylogenetic Research Science Gateway (CIPRES) 3.1 (Miller et al., 2010). ML was run via RAXML 8.2.10 (Stamatakis, 2014) with 1000 bootstrap iterations and a GTRGAMMA model under the default parameters (Stamatakis, 2015). *Psilopezia* served as the outgroup taxon for the 28S rDNA alignment of Pezizaceae because it belongs to Rhizinaceae. The ITS trees were midpoint rooted. The best ML tree for each analysis was visualized and rooted in FIGTREE 1.4.3 (Rambaut, 2012). Nodes were considered strongly supported if bootstrap values were $\geq 70\%$.

Results

Morphological results

Details of our morphological studies are presented in the section of assignments below. Our morphological review of the type specimen of *Pustularia microspora* revealed amyloid asci and spore ornamentation that were not noted in the original description (Raithelhuber, 1983). The morphology of the type specimen is consistent with placement within *Peziza* sensu stricto. Our combined morphological and molecular analyses indicate that *Pustularia microspora* is a synonym of *Peziza pseudosylvestris* (see below). Other *Peziza* sensu lato taxa were studied and compared with described species. Based on morphological and molecular evidence, one new species of *Peziza* sensu stricto is described (*P. gamundiae* sp. nov.) and a new combination (*Phylloscypha nothofageti* comb. nov.) is proposed. Additional discussion of the various species is found below.

Phylogenetic results

The final 28S alignment had 848 positions with 425 distinct patterns. The *Geoscypha-Malvipezia* ITS alignment had 49 taxa, 784 positions, and 332 distinct patterns; the *Phylloscypha* ITS alignment had 43 taxa, 630 positions, and 250 distinct patterns; the *Peziza* sensu stricto part 1 alignment had 53 taxa, 607 positions, and

218 distinct patterns; the *Peziza* sensu stricto part 2 alignment had 54 taxa, 726 positions, and 370 distinct patterns. The 28S phylogenies placed our recent collections in four genera: Geoscypha, Malvipezia, Peziza sensu stricto, and Phylloscypha (Fig. 1). The ITS phylogenetic analyses identified eight putative species of *Peziza* sensu lato from southern Chile and Argentina (Figs. 2-5). Additionally, our ITS phylogeny of *Peziza* sensu stricto resolved the type specimen of Pu. microspora in a well-supported clade comprised of several specimens of P. pseudosylvestris from southern South America (Fig. 2). This clade includes material identified by Gamundí as P.pseudosylvestris (KC832899 and KC832900, Lantieri et al., 2016) as well as eight new collections that we identified as *P. pseudosylvestris* from sites spread across southern Chile and Argentina. This clade received strong bootstrap support and was phylogenetically differentiated from all Northern Hemisphere species. Sequences from two collections of *Peziza* from Argentina (OM302394 and KY462666) were distinct from any sequenced identified species and are discussed in the taxonomy section. Two additional *Peziza* sensu stricto species were detected among our collections (Fig. 3). One was a putatively new species from Argentina (KY462304) that is molecularly similar to *P.ampliata* and to the *P. fimeti* species complex, but distinct from previously sequenced specimens. The other is a new species we propose as P. gamundiae represented by sequences of two collections from Chile (MT505311 and MT505312) that are allied to but distinct from a strongly supported clade of species from the Northern Hemisphere consisting of P. lohjaoensis, P. montirivicola, P. nordica, P. oliviae, and P. sublaracina.

All of the southern South American clades were distinct from Northern Hemisphere clades and likely represent endemic taxa, except for members of two *Geoscypha* clades (Fig. 4). In a recent study, Van Vooren (2020) resurrected the genus *Geoscypha* and included *G. ampelina* (Gillet) Van Vooren & Dougoud (=*Galactinia ampelina* (Gillet) Boud.), *G. tenacella* (Sacc.) Van Vooren (=*P.tenacella* W. Phillips ex Sacc.) and *G. violacea* (Pers.) Lambotte (=*P. violacea* Pers.). Sequences from ascomata and an anamorph collection in Argentina and Chile cluster with *G. tenacella*; an ascoma sequence from Chile clustered with *G. violacea*. All of our collections of *G. tenacella* were from fire pits. Half of our *G. tenacella* collections were anamorphs but were not collected in the same places as their teleomorphs. Our sequences from both species from Chile and Argentina match in 100% BLAST searches to endolichenic and endophytic ITS sequences from China, Japan, Germany, Russia, Canada and the USA (Alaska and Arizona) (JX298895, LC431567, KX909074, MG098331, HM123160).

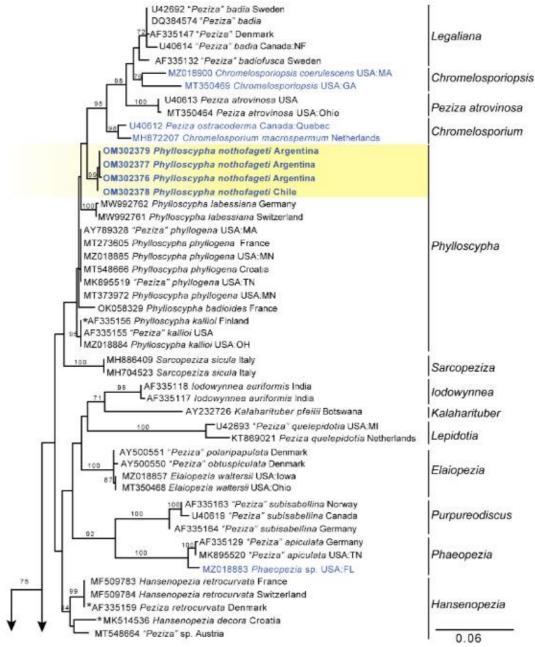


Fig. 1. Maximum likelihood phylogeny based on analysis of 28S sequences of selected species of the family *Pezizaceae* rooted with the outgroup *Psilopezia*

(family Rhizinaceae), and bootstrap support ≥70% shown at branch nodes. Sequenced taxa from southern South America are highlighted in yellow and shown in bold text, sequences from ascomata are in black, asexual sporemats are in blue, sequences from TYPE material are indicated with an asterisk, and material collected

from burn sites are indicated with orange-red circles. The current generic delimitation is indicated by text and bars on the right side of the tree with outdated

species names indicated in quotes. Color version at http://www. ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

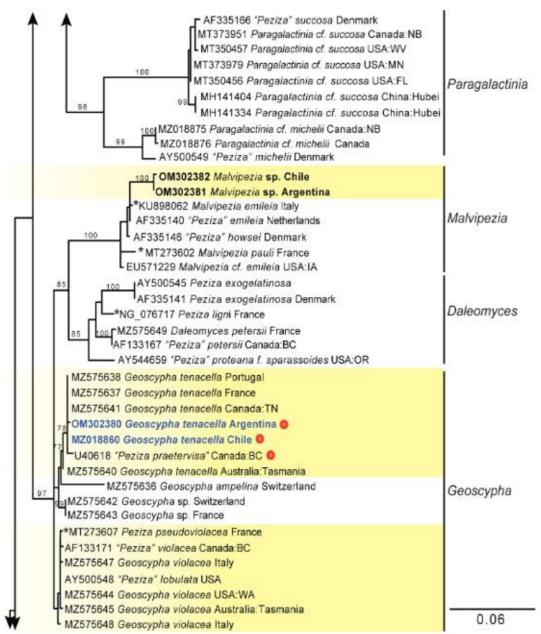


Fig. 1. (Continuation). Maximum likelihood phylogeny based on analysis of 28S sequences of selected species of the family *Pezizaceae* rooted with the outgroup *Psilopezia* (family Rhizinaceae), and bootstrap support ≥70% shown at branch nodes. Sequenced taxa from southern South America are highlighted in yellow and shown in bold text, sequences from ascomata are in black, asexual sporemats are in blue, sequences from TYPE material are indicated with an asterisk, and

material collected from burn sites are indicated with orange-red circles. The current generic delimitation is indicated by text and bars on the right side of the tree with outdated species names indicated in quotes. Color version at http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

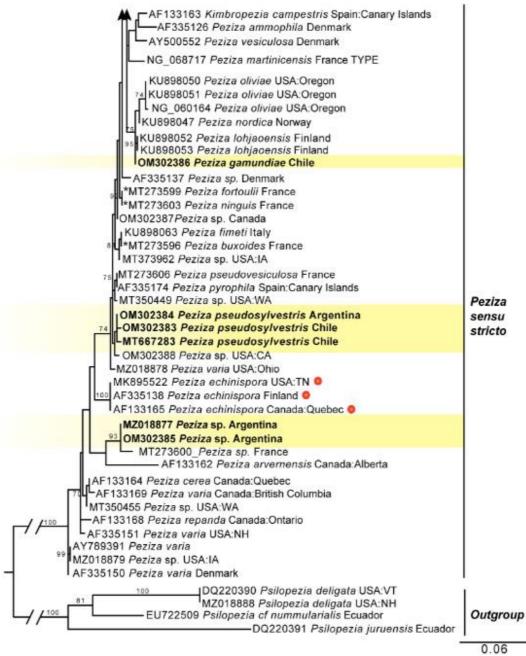


Fig. 1. (Continuation). Maximum likelihood phylogeny based on analysis of 28S sequences of selected species of the family *Pezizaceae* rooted with the outgroup

Psilopezia (family Rhizinaceae), and bootstrap support ≥70% shown at branch nodes. Sequenced taxa from southern South America are highlighted in yellow and shown in bold text, sequences from ascomata are in black, asexual sporemats are in blue, sequences from TYPE material are indicated with an asterisk, and material collected from burn sites are indicated with orange-red circles. The current generic delimitation is indicated by text and bars on the right side of the tree with outdated species names indicated in quotes. Color version at http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

An unknown species of *Malvipezia*, one of the new genera erected by Van Vooren (2020), was detected from two sequences (KY462454 and OM302389) from collections in Argentina and Chile that are molecularly distinct from any other species based on DNA (Fig. 4). Endophytes are also detected within the genus *Malvipezia*. The *Geoscypha-Malvipezia* alignment showed a unique indel in the ITS1 of *Malvipezia* that was approximately 185 bp long in the taxon from South America, 131 bp long in *M. emileia*, and 134 bp long in endophytes and other undetermined *Malvipezia* species. Where the indel aligned, there was an 18% difference between the South American species and *P. emileia*, a 13% difference between the endophytes and *P. emileia*.

One well-supported species from the newly described genus *Phylloscypha* (Van Vooren, 2020) was detected from sequences of both Argentine and Chilean collections (Fig. 5). It is recombined here as *P. nothofageti*.

Assignment of taxa and discussion

Daleomyces

Setch., Mycologia 16(5). 240. 1924.

Daleomyces petersii

(Berk.) Van Vooren, Ascomycete.org 12 (4): 185. 2020. Basionym: *Peziza petersii* Berk. & M.A. Curtis, Grevillea3: 150. 1875. Mycobank no. 835804. Reported by Gamundí (1964, 1971) from burned ground in Chile and Argentina. Some of her collections were confirmed by expert R. W. G. Dennis (Gamundí, 1964). Rifai (1968) also reports specimens from New Zealand. Recent work by Vizzini et al. (2020) treats this species in the genus *Peziza* and suggests several synonyms. There are no DNA sequences from southern South American material.

Geoscypha

(Cooke) Lambotte, Mém. Soc. Roy. Sci. Liège, Série 2 14: 320. 1887 [1888].

Geoscypha tenacella

(Sacc.) Van Vooren, Ascomycete.org 12: 187. 2020. Basionym: Humaria tenacella W. Phillips ex Sacc. Syll. Fung. 8: 145. 1889. Mycobank no. 189696. Fig. 6. This record is based on new collections of both teleomorphs and anamorphs. Our morphological identification of teleomorph specimens agrees with the description by Rifai (1968) who studied the type collection from Melbourne, Australia as well as additional collections from elsewhere in Victoria and Tasmania. Rifai (1968) points out that this was likely confused with G. violacea (=Peziza violacea) in Australia. Geoscypha tenacella differs from G. violacea in having finely ornamented ascospores with two indistinct guttules. The spore dimensions of our collection (11-12 \times 5-6 µm) fall within the range recorded by Rifai (10.5-12 \times 5.4-6.5 µm). He remarks that *P. tenacella* and *P. praetervisa* differ only in minor details of spore ornamentation and curvature of the paraphyses. Following general acceptance of Rifai's opinion, Van Vooren (2020) places P. praetervisa in synonymy with G. tenacella. The reports of P. praetervisa from Chile (Lazo, 2016) and Argentina by Gamundí (1971) and Gamundí and Giaiotii (1998) probably represent G. tenacella.

In our collections the excipulum is up to 200 μ m thick toward the base. The excipulum rehydrates poorly but seems to be composed of two layers. The spores are clearly ornamented with very minute warts. Field collections of the *Oedocephalum* conidial state are also listed below. Webster et al. (1964) described this anamorph from spore culture of a specimen named *Peziza praetervisa*. We report and illustrate this anamorphic state from field collected materials which were encountered in fire pits. This species is frequently isolated as an endophytic or endolichenic fungus from around the world (Healy et al., 2022). We have included sequences of some of these isolates in Fig. 4.

Specimens examined

ARGENTINA. **Río Negro**, Parque National Nahuel Huapi, Los Rapidos near bridge, forest of *Nothofagus dombeyi*, 11-V-2015, Donald H. Pfister MES-1232 (FLAS-F-63191, CORD-C6631, FH 00964505), anamorph; Parque National Nahuel Huapi, Los Rapidos near bridge, *Nothofagus dombeyi*, 13-V-2016, Rosanne Healy MES-1913 (FLAS-F-64622, CORD-C6908), anamorph. CHILE. **Los Lagos**, Prov. Osorno, Puyehue National Park, Anticura, in firepit in *Nothofagus dombeyi* forest, 05-V-2016, Matthew E. Smith MES-1674 (FLAS-F-64458, FH 00965551). CHILE. **Magallanes y Antártica Chilena**, Prov. Magallanes, Puente San Pedro, 53° 41' 57.5" S, 70° 58' 10.5" W, 12 m a.s.l., in a firepit in *Nothofagus betuloides* forest, 5-IV-2017, Donald H. Pfister MES-2448 (FLAS-F-65081, FH 00965553).

Geoscypha violacea

(Pers.) Lambotte, Mém. Soc. Roy. Sci. Liège, Série 2 14:320. 1887 [1888]. Basionym: *Peziza violacea* Pers., Neues Magazin für die Botanik 1: 113 (1794). Mycobank no. 200578. Fig. 7 A, B.

For the complicated nomenclatural history of this taxon see the commentary in Van Vooren (2020). The specimen examined agrees morphologically and phylogenetically with *G. violacea* (Fig. 4) as defined by Van Vooren (2020) who characterizes this species as violet, with smooth spores that lack prominent guttules. Ascospores in our material are $15-17 \times 7-9 \mu m$. Gamundí et al. (2004) reports a collection from Argentina. The species is widespread and as is the case with *G. tenacella* is endophytic and endolichenic (Healy et al., 2022); exemplar sequences are included here.

Specimen examined

CHILE. **Araucanía**, Prov. Cautín, Parque Nacional Villarrica, sector Quetrupillán, on soil under *Nothofagus dombeyi* and *Araucaria araucana*, 15-X-2019, M. Caiafa & M. E. Smith MES-3594 (FLAS-F-66023; FH 0096554).

Legaliana

Van Vooren, Ascomycete.org 12 (4): 188. 2020.

Legaliana badia

(Pers.) Van Vooren, Ascomycete.org. 12: 188. 2020. Basionym: Peziza badia Pers., Observ. Mycol. 2: 78. 1799. Mycobank no. 141966. Gamundí et al. (2004) reports a collection from Argentina (BCRU-1860). We could not gain access to this specimen for morphological studies, but we consider this report to be tentative. This species was first reported by Bertero (1829) from the bank of a ditch in Chile. The spores of *Legaliana badia* are characteristically marked with partial or complete reticulations. Legaliana badia seems to represent a species complex and careful morphological and molecular phylogenetic analyses will be necessary to fully understand the diversity within this group. Importantly, taxa in this group are ectomycorrhizal so mycorrhizal hosts should be documented. Although Rifai (1968) included L. badia (as Peziza badia) in his treatment of Australasian Pezizales, the description is drawn from British material. A sequence (OL653060) establishes the presence of L. badia or a close relative in New Zealand. There are several reports of *L. badia* from the southern regions of Australia (see Atlas of Living Australia). There are no DNA sequences from southern South American material.

Malvipezia

Van Vooren, Ascomycete.org 12(4): 188. 2020.

Malvipezia

sp. Fig. 7 C, D.

Two collections were identified. FLAS-F-63073 (MES-1069) can be referred to this genus based on both ITS and 28S data (Figs. 1, 4). FLAS-F-63073 (MES-1283) is immature but shows the general ascus bluing of walls and at the ascus tip that is characteristic of this group (WT in the notation of Van Vooren, 2021). Collection FLAS-F-63073 has mature spores that are minutely echinulate and measure $(14-)15-15.8(-17) \times 8-9 \ \mu m$ with isolated warts less than 0.5 $\ \mu m$ high. The warts are uniformly dispersed on the spore surface. Paraphyses are mostly inflated and pigmented at the tips. The terminal cells of the paraphyses are up to 10 $\ \mu m$ and filled with brownish globular contents. Species-level identification is not possible based on the single mature specimen we have

available; more collections will allow a thorough study of this species. It is related to *M. emileia*.

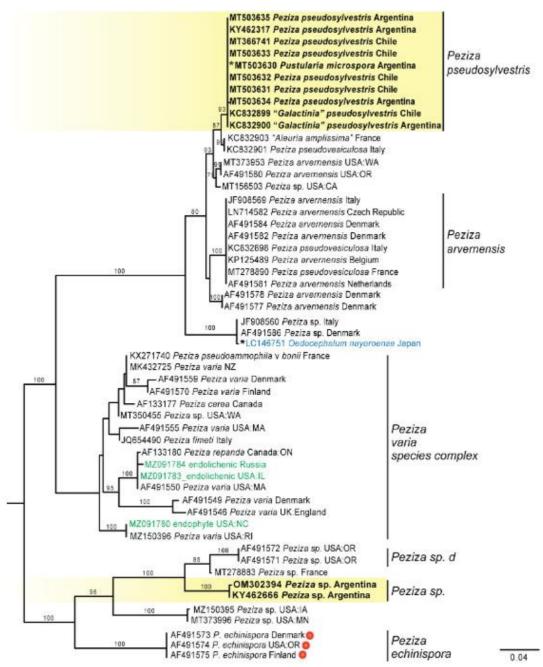


Fig. 2. Maximum likelihood phylogeny based on analysis of ITS sequences from species of *Peziza* sensu stricto, part 2, with bootstrap support \geq 70% shown at branch nodes. Sequenced taxa from southern South America are highlighted in yellow and shown in bold text. Sequences from ascomata are in black, asexual sporemats are in

blue, sequences from TYPE material are indicated with an asterisk, and material collected from burn sites are indicated with orange-red circles. The current generic delimitation is indicated by text and bars on the right side of the tree with outdated species names indicated in quotes. Color version at

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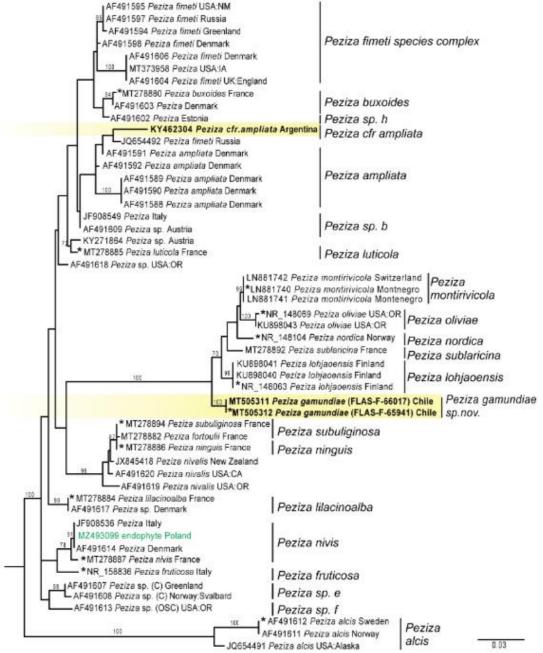


Fig. 3. Maximum likelihood phylogeny based on analysis of ITS sequences from

species of *Peziza* sensu stricto, part 3, with bootstrap support ≥70% shown at branch nodes. Sequenced taxa from southern South America are highlighted in yellow and shown in bold text. Sequences from ascomata are in black, sequences of endophytes are in green, and sequences from TYPE material are indicated with an asterisk. The current generic delimitation is indicated by text and bars on the right side of the tree with outdated species names indicated in quotes. Color version at http://www.ojs. darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

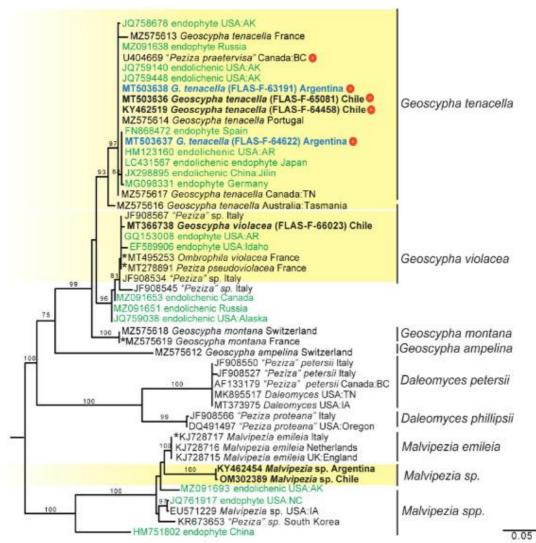


Fig. 4. Maximum likelihood phylogeny based on analysis of ITS sequences from species of *Geoscypha*, *Malvipezia*, and *Daleomyces*, with bootstrap support \geq 70% shown at branch nodes. Sequenced taxa from southern South America are highlighted in yellow and shown in bold text. Sequences from ascomata are in black, sequences of endophytes are in green, asexual sporemats are in blue, and

sequences from TYPE material are indicated with an asterisk. The current generic delimitation is indicated by text and bars on the right side of the tree with outdated species names indicated in quotes. Color version at

http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

Examined specimens

ARGENTINA. **Neuquén**. Los Lagos, Villa la Angostura, Lago Queñi area, Parque Nacional Lanín, *Nothofagus dombeyi, N. nervosa, N. obliqua*, 15-V-2015, Rosanne Healy & Matthew E. Smith MES-1283 (CORD-C6573, FLAS-F-63230); CHILE. **Los Ríos,** Osorno, Puyehue National Park, below Antillanca, ca. 1000 m a.s.l., under *Nothofagus dombeyi*, 6-V-2015, Rosanne Healy MES-1069 (FLAS-F-63073).

Paragalactinia

Van Vooren, Ascomycete.org 12(4): 188. 2020.

Paragalactinia succosa

(Berk.) Van Vooren, Ascomycete.org 12 (4): 189. 2020. Basionym: *Peziza succosa* Berk., Annals and Magazine of Natural History 6: 358. 1841. Mycobank no. 231097.

This species is described and beautifully illustrated in Gamundí (1960) based on collections from Río Negro and Mendoza in Argentina. Gamundí states that a collection was authoritatively determined by Madame Marcelle LeGal. Gamundí noted that it is common along ditches in the fruit growing regions of Mendoza. *Paragalactinia succosa* and *P. succosella* represents a complex awaiting resolution. *Paragalactinia succosa* and related taxa are all considered ectomycorrhizal, so it seems likely that collections from central Argentina are symbionts of introduced *Salix, Quercus* and other Northern Hemisphere plants, but this has not been confirmed. A hypogeous ptychothecial member of the genus *Paragalactinia* has been collected in Argentina (Truong et al. 2017) and will be described in a subsequent publication. Two other hypogeous *Paragalactinia* species, *P. infossa* and *P. eriniae*, have been previously described from North America (Smith, 2014). There are no DNA sequences from southern South

American material of Paragalactinia succosa.

Paragalactinia succosella

(LeGal & Romagn.) Van Vooren, Ascomycete.org 12 (4): 189. 2020. Basionym: *Galactinia succosella* Le Gal & Romagn., Revue Mycol. (Paris) 5: 113. 1940. Mycobank no 297582.

Gamundí (1971) provides a description of a collection from Chile (Valparaíso, Viña del Mar, Botánico National, leg. W. Lazo Vi-26, 28-VII-1966, LPS 34526) for a report by Lazo et al. (1977) that she refers to this species based on morphological similarities with the description of the type specimen. It is illustrated and described by Lazo (2016). The species differs from *P. succosa* in having smaller ascospores that are not as densely ornamented. As is the case with *P. succosa*, these names have been broadly assigned within species complexes that are shown in phylogenetic sequence analyses to be highly diverse (data not shown). There are no DNA sequences from southern South American material.

Peziza

Dill. ex Fr., Syst. Mycol. (Lundae) 2(1): 40. 1822.

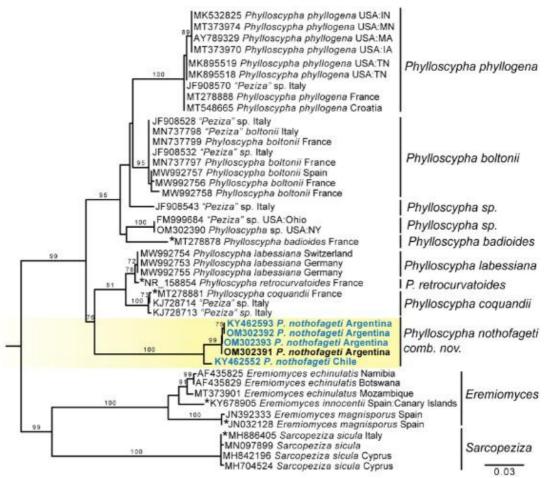
Peziza ammophila

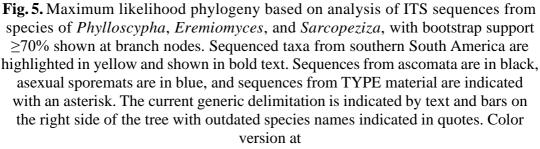
Durieu & Lév., in Durieu, Expl. Sci. Alg., Fl. Algér 1: tab 28, fig. 2 (1848). Mycobank no. 235446.

Gamundí (1966, 1971, 1975, 1983) reported collections from Chile, Argentina, and Uruguay and, because of its semihypogeous habit, treated the species in the genus *Sarcosphaera*. It is described and illustrated by Lazo (2016). Phylogenetic studies have shown that *Sarcosphaera* is assigned to a distinct clade outside the *Peziza* sensu stricto group. Molecular phylogenetic works place *P.ammophila* in *Peziza* sensu stricto. Gamundí states that this species is common on the Atlantic coast of Argentina. The species has a characteristic habit of being partially hypogeous and occurring on dunes, often with beach grasses in the genus *Ammophila*. *Ammophila* species are native to the North Atlantic coasts of North America and Europe, and Great Lakes of North America but have been widely introduced, in Chile and Argentina but also Australia and New Zealand (Wiedemann, 1998). Gamundí (1966) does not mention plant associates of the collections she cites. Although this fungus regularly co-occurs with *Ammophila*, there is no evidence of a strict association.

Recent morphological and phylogenetic studies have shown that collections of *P. ammophila* represent several taxa (Vizzini et al., 2016). The spore size reported by Gamundí (1975) places South American collections within the *P. ammophila* complex, but molecular data are needed to resolve the exact identity of the South American species.

Vizzini et al. (2016) studied the southern hemisphere collections referred to P. ammophila and described *P. oceanica* from New Zealand. This species is distinctive in its reddish pigments and in its cupulate apothecial form that lacks the characteristic star-like splitting found in some other members of the group. Among this complex *P. psammobia* Rifai (1968) should also be reconsidered. It is described as semi-hypogeous on sandy soil in Australia but differs from other described species in the complex by smaller ascospores $(13.8-16.3 \times 10-11.5 \ \mu\text{m})$ that are broadly ellipsoid with a surface marked with low, isolated warts. Yet another Australian dune-inhabiting fungus is *Peziza austrogeaster* (Rodway) Rifai. This species has apiculate ascospores up to 27.3 µm long and 16.2 µm broad. Peziza retispora is another dune dweller from Australia. It has reticulate spores that are $17.2-20 \times 10-11.8 \,\mu\text{m}$ and have a larger oil droplet. This species is probably not part of the *P. ammophila* group but is interesting because of the similar habitat. These additional species are noted here to draw attention to these understudied southern hemisphere taxa and the diversity among these duneinhabiting fungi.





http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

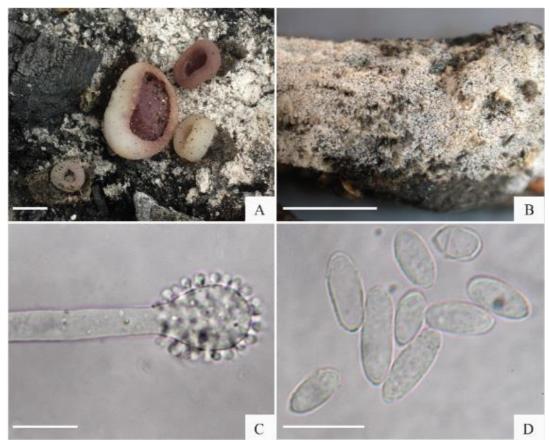


Fig. 6. *Geoscypha tenacella*, teleomorph and anamorph. **A,** fresh field photograph of *G. tenacella* MES-1674 (scale = 1 cm). **B,** field collected anamorph, MES-1913 (scale = 1 cm). **C,** condiophore with conidiogenous loci, MES-1913 (scale = $25 \mu m$). **D,** conidia, MES-1913 (scale = $10 \mu m$). Color version at http://www.ojs.darwin.edu.ar/ index.php/darwiniana/article/view/1019/1252

We have not reexamined specimens of this group. Southern South American specimens of *P. ammophila* are reported from dunes of the Buenos Aires region and from Punta del Este, Uruguay. Given the seeming high diversity among related fungi, South American material should be sought to compare DNA sequences with other taxa in the *P. ammophila* complex. There are no DNA sequences from southern South American material.

Peziza cfr. **ampliata** Pers., in Pant, *Mycol. eur.* (Erlanga) 1: 227 (1822). Mycobank no. 245810. Fig. 7 C, D.

The collection cited here falls in a clade with several collections referred to *Peziza* cfr. *ampliata* (Fig. 3). It is apparent from the sequenced collection that there is considerable molecular variation within this group. Among the sequences, one specimen was identified as *P. fimeti*. We have examined specimen CT4247 (FLAS-F-63345) and consider it as a representative of the *P. ampliata* complex. The

hymenium is maroon-brown. The excipulum is thin, approximately 75 μ m thick. Asci are 110-120 × 12.5 μ m and the suhymenium consists of a layer of inflated cells (up to 12.5 diam. but usually around 5 μ m diam.). Spores are 21-23 (-25) × 9-10 μ m. This spore range is within that accepted for *P. ampliata* by Hansen et al. (2002). We observed an obvious general amyloid response of the asci (WT following Van Vooren, 2021); many of the spores we observed were immature in this specimen. The specimen was fruiting on rich, wet earth.



Fig. 7. *Geoscypha violacea*, *Malvipezia* sp., and *Peziza* cfr. *ampliata*. **A**, **B**, *Geoscypha violacea* field photographs, MES-3594 (scale = 1 cm). **C**, *Malvipezia*

sp. field photograph MES-1069 (scale = 1 cm). **D**, *Malvipezia* sp. MES-1283 (scale = 1 cm). **E**, **F**, *Peziza* cfr. *ampliata*, field photograph CT-4274, (scale = 1 cm). Color version at http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

See also Medardi et al. (2012) and the discussion under *P. fimeti* below.

Examined specimens

CHILE. **Magallanes y de la Antártica Chilena**, Depto. Tierra del Fuego, El Parque Natural Karunkinka, Valle de La Paciencia, S54.36025 W 68.834138, 200 m a.s.l., in *Nothofagus pumilio* forest, 11-III-2015, Camille Truong CT-4247 (FLAS-F-63345, FH 00965549).

Peziza arvernensis see *Peziza sylvestris*. Peziza badia see *Legaliana badia*. Peziza badioconfusa see *Phylloscypha nothofageti*.

Peziza bonaerensis

Speg., An. Soc. Cient. Argent. 10(1):23. 1880. Mycobank no. 587290. This species was described from Buenos Aires and according to Gamundí (1960), who studied the type material, the ascospores are smooth and 15.6-22.7 \times 9.1-14.3 µm with small guttules. The paraphyses are bifurcate. Three collections are cited but one is reported as having smaller spores. The broadly ellipsoid smooth spores and flesh that is uniformly composed of globose cells places this in the *Peziza* sensu stricto. Collections are on humid rich soil. Gamundí (1960) suggests an affinity with *P. domiciliana*. There are no DNA sequences from southern South American material.

Peziza cerea

Sowerby, Col. Fig. Engl. Fung. (London) 1(no 6): tab 3. 1796. Mycobank no. 161555.

Hansen et al. (2002) suggested that *P. cerea* in the sense of most modern authors is synonymous with *P. varia*. Gamundí's (1975) excellent description of *P. cerea* from a collection on soil in Tierra del Fuego is in close agreement with the

information given by Hansen et al. (2002) for *P. varia* except for the smaller ascospores indicated by Gamundí. She records a range from 12-14.4 × 6.7-7.2 µm whereas Hansen et al. (2002) give a range of "14.5-16-17.5 × 8-9.5-10.5." *Peziza varia* represents a complex of species that share often stratified excipular tissues of large globose cells with an intermediate zone of interwoven hyphae (textura intricata). Rifai (1968) also reported *P. cerea* from Australia and New Zealand. He reported spore sizes of 13-15.7 (17.2) × 8-9.5 µm. He included collections on wood as well as on soil, damp cellars and greenhouses. He also recognized *P. varia* as a distinct species, which he primarily characterized by the swollen cells of the paraphyses giving them a monilioid appearance. We consider this feature a product of environmental factors, such as high relative humidity. Spores of *P. varia* were reported as $12.5 \times 16 \times 8-10$ µm (Rifai, 1968). Pegler et al. (1980) recorded the species from South Georgia Island, Husvik Whaling Station, on an old (pre-1964) heap of coal (10 March 1979, specimen M. Smith 12). There are no sequences from southern South American material.

Peziza domiciliana

Cooke, Gard. Chron., N. S. 7: 793. 1877. Mycobank no. 217216. Gamundí (1960) illustrates a collection and cites material from the castings of mushroom cultivation beds, a hot house and other domestic habitats. One collection was confirmed by F. J. Seaver. The interpretation of this species centers on the obscurely guttulate ascospores with finely verrucose walls. Lazo (2016) illustrates and describes a collection referred to *P. domiciliana*. Rifai (1968) discussed *P. domiciliana* under his commentary on *P. cerea*. Hansen et al. (2002) accepted the species, pointing out that it is the only member of the *Peziza* sensu stricto group that has guttules. The small spores ranging only up to 14 μ m are also distinctive. These characters are also discussed by Gamundí (1960). There is no DNA sequence from southern South American material.

Peziza echinispora

"complex".

We have adopted the spelling *P. echinispora* rather than *P. echinospora* for this assemblage of taxa following Hansen et al. (2001). They pointed out that this was the original spell when the name was published. This complex requires further work to resolve the status of several taxa. Two of our collections from soil are placed as sister to "*Peziza d*" in our phylogeny (Fig. 2) which in turn are sister to *P.*

echinispora, a species putatively found worldwide associated with burns. "Peziza d" was a placeholder for collections from Oregon USA in Hansen et al. (2002). "Peziza d" was not indicated as associated with burned areas, but rather on wood chips and among grasses. "Peziza d" agrees in habitat with our collections, which were found on soil and are not associated with burns. This points to the ecological variation in taxa around P. echinispora. More collections will be needed to determine the status of these specimens as well as to determine if "Peziza d" has been described. Our collections from Argentina cited here differ from P. echinispora in having smooth ascospore walls rather than echinulate ascospore markings. Gamundí's (1964, 1975), records from Argentina seem to fit the concept of *P. echinispora*, a widespread species that seems to occur around the world on burned areas and soil. Rifai (1968) reported P. echinispora from Australia. He suggests that the smaller spores of the Australian material may indicate that this represents a new species. Palacios (1966) reported P. anthracophila from Chile. This is generally considered to be a later name for Peziza echinispora.

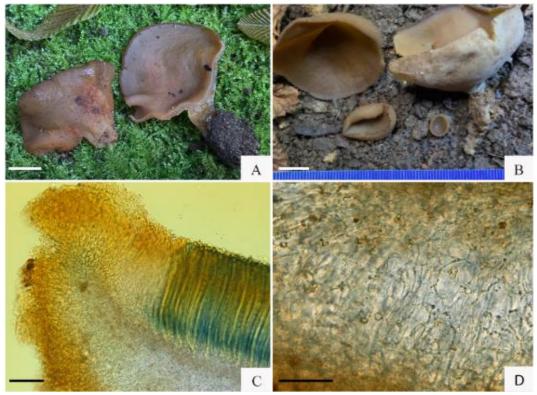


Fig. 8. *Peziza gamundiae*. A, field collection, MES-3587 (scale = 1 cm). B, field collection, MES-3507 (scale = 1 cm). C, portion of the apothecial margin, mounted in Melzer's reagent, MES-3587 (scale = $50 \mu m$). D, outer layer of the excipulum showing globose and elongate cells, MES-3587 (scale = $50 \mu m$). Color version at

http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

Specimens examined

ARGENTINA. **Río Negro**. Nahuel Huapi National Park, 1 km after Lago Hess, on soil under *Nothofagus antarctica*, 15-V-2015, Alija Mujic MES-2089 (FLAS-F-64805, CORD-C6907, FH 00965558); Nahuel Huapi National Park, 1 km before Lago Hess, on soil under *Nothofagus antarctica*, 18-V-2016, Matthew E. Smith MES 2133 (FLAS-F-64754, CORD-C5272, FH 00965559).

Peziza fimeti

(Fuckel) E. C. Hansen, Vidensk. Meddel. Dansk Naturhist. Foren. Kjøbenhavn 59: 267. 1877 [1876]. Basionym: *Humaria fimeti* Fuckel, Jb. *nassau. Ver. Naturk.* 25-26: 338. 1872 [1871]. Mycobank no. 182983. Gamundí et al. (2004) lists several collections all from Argentine Tierra del Fuego. We have not encountered collections referable to *P. fimeti* but see the discussion under *P. granulosa* and results in Fig. 3. One collection from Tierra del Fuego closely agrees with Gamundí's description and falls within a complex surrounding *P. ampliata* in our molecular analyses (see discussion under that name). Gamundí correctly determined that *P. bovina* is a synonym of *P. fimeti*, as was also established by Hansen et al. (2002). See also comments under *Peziza*

granulosa.

Peziza gamundiae

sp. nov. Pfister, Caiafa & M.E. Smith, Fig. 8 A-D. Mycobank no. 843798.

Diagnosis

This species is most similar to *P. oliviae* in spore size, $17-21 \times 8-9 \mu m$ (in *P. gamundiae*) vs. $15.5-24 \times 9-5-12 \mu m$ (in *P. oliviae*), asci J+ with lateral walls bluing and bluing intensely at the tip forming a stained cap-like structure. *Peziza gamundiae* has a shorter stipe and does not occur under water, as does *P*.

oliviae.

Holotypus

CHILE. **Araucanía**, Cordillera de Nahuelbuta, Fundo Rucalemu, on soil in mixed native forest dominated by *Lophozonia obliqua* and in some places *Nothofagus dombeyi*, 04-X-2019, *Marcos Caiafa & Matthew E. Smith* MES 3507 (holotype SGO, isotypes FLAS-F-6594 and FH 00965560).

Description

Apothecia up to 3 cm diam., hymenium pale brown, outside smooth or slightly tomentose, concolorous or slightly lighter, with a short but distinct stipe, $4-6 \times$ 10 mm. When young slightly concave, margin becoming in-rolled, at maturity deep cupulate, margin wavy. Flesh fragile, brown in dried material. Asci up to $325 \,\mu\text{m}$, with a terminal operculum, 8-spored, croziers present, contents of young asci intensely dextrinoid in Melzers, ascus walls intensely blue in Melzers along their entire length, with an intensely bluing area at the tip appearing as a blue cap, no bluing ring present. Ascospores uniseriate, narrowly ellipsoid, smooth, hyaline, (17) 19-21 (22) \times (7) 8-9 (10) μ m, thick-walled when young, thin-walled at maturity, without internal guttules, contents granular, de Bary bubbles sometimes present. Paraphyses only slightly swollen at the tip, to 5-7 μ m, unbranched, with yellow inclusions in terminal cells that are released into the mounting medium. Subhymenium densely interwoven textura intricata of cells 5-6 µm diam., with brownish walls, becoming tightly interwoven toward the hymenium. Excipulum composed of three layers, below the subhymenium is a layer composed of globose cells that are less than 100 µm diam. This is subtended by a zone of interwoven hyphae forming a textura intricata. The outer layer composed of globose cells up to 80 µm diam., some cells of which are compressed and radially arranged.

Etymology

Named in honor of Irma J. Gamundí, distinguished mycologist of Argentina, whose study of cup-fungi has established a foundation for our work.

Ecology, habitat, and distribution

Collected on soil from Nothofagaceae forests in Southern Chile.

Examined specimens

CHILE. **Araucanía**, Villarrica National Park, Sector Puesco, on soil on bank of road, under *N. dombeyi* and *N. alpina*, 14-X-2019, Marcos Caiafa & Matthew E. Smith MES-3587 (FLAS-F-66017, FH 00965556); Cordillera de Nahuelbuta, Fundo Rucalemu, on soil, in mixed native forest dominated by *Lophozonia obliqua* and in some places *Nothofagus dombeyi*, 04-X-2019, Marcos Caiafa & Matthew E. Smith MES-3507 (FLAS-F-65941, FH 00965560), isotype.

Comments

This species falls with a clade that includes *P. oliviae*, *P. montirivicola*, *P. lohjaoensis* and *P. nordica* (Fig. 3). See Pfister et al. (2016) for a comparison of these species.

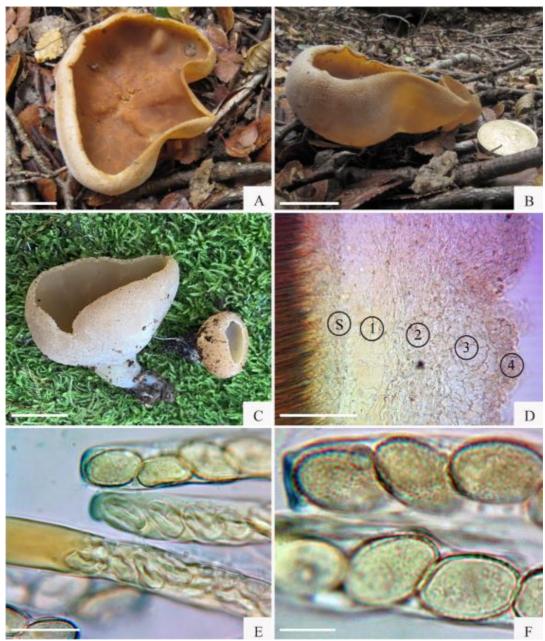


Fig. 9. *Peziza pseudosylvestris*. **A, B,** fresh field photograph of *P. pseudosylvestris* specimen MES-2455 (scale = 2 cm). Photo credit: G. Furci. **C,** fresh field photograph of *P. pseudosylvestris* specimen MES-3614 (scale = 1 cm). **D,** microscopic photograph of *P. pseudosylvestris* specimen MES-3614 showing the subhymenium (S) and four excipular layers (1-4) discussed in the original description of the species by Gamundí et al. (1975) (scale = 500 μ m). **E,** microscopic photograph of the TYPE specimen of *Pustularia microspora* (ZT-60854). Note the ornamented spores, the strong amyloid response of the ascus tips and the dextrinoid response of the young ascus (scale =

20 μ m). **F**, micrograph of the TYPE specimen of *Pustularia microspora* (ZT-60854). Note the amyloid ascus tip and the obvious spore ornamentation (scale = 10 μ m). Color version at http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

Three of these species (P. oliviae, P. montiricola and P. nordica) are unusual in that they occur in or under running water or in areas that are frequently inundated. There is no indication that P. gamundiae fruits in such situations, but this requires further field observations. All of the taxa in this group are characterized by their narrow elliptical, smooth ascospores without guttules and amyloid asci that are intensely blue at the tip, forming almost a cap, but lacking an amyloid ring. *Peziza gamundiae* is the only Southern Hemisphere representative of this clade so far as known; all of the other known taxa in this group (P. oliviae, P. montiricola, P. nordica and P. lohjaoensis) are described from the Northern Hemisphere. Among the known taxa from southern South America, Peziza gamundiae is most likely to be confused with member of the P. varia group in ascomatal coloration and in having smooth ascospore walls. It might be confused with the common *P. pseudosyvestris* in ascomatal color but differs in having unornamented ascospore walls. Microscopically P. gamundiae differs from the Peziza sensu stricto species in having asci with a cap-like region around the apex that becomes blue in iodine and lacks a distinct amyloid apical ring.

"Peziza granulosa Schumach."

Enum. pl. (Kjbenhavn) 2: 415 (1803). Mycobank no. 476746.

Several collections are noted by Gamundí (1964, 1975). *Peziza granulosa* Schumach. is illegitimate as a later homonym of *P. granulosa* Pers., which is *Cheilymenia granulata* (Bull.) J. Moravec. Hansen et al. (2002) considered that *P. granulosa* in the sense used by Gamundí, and other authors may be a synonym of *P. fimeti*. The spore size reported by Gamundí (1975) is slightly larger ($20-23.4 \times 10-12.5 \mu m$) than that reported by Hansen et al. (2002). This form occurs on soil and has yellow-green pigments in the paraphyses. These are characters that Gamundí considered important in defining *P. granulosa*. Since we have not seen recent collections that are morphologically referable to this taxon, additional new specimens are needed to evaluate the identity of this taxon sensu Gamundí (1964, 1975). There are no DNA sequences from southern South American material.

Peziza micropus see comments under *P. varia*.
Peziza nothofageti see *Phylloscypha nothofageti*.
Peziza petersii see *Daleomyces petersii*.
Peziza praetervisa see *Geoscypha tenacella*.

Peziza pseudosylvestris

(Gamundí) Alessio, Micol. Ital. 4(2): 21. 1975. Basionym: *Galactinia pseudosylvestris* Gamundí, Fl. Criptog. Tierra del Fuego (Buenos Aires) 10(3): 37. 1975. Mycobank no. 314256. Fig. 9 A-F.

= Pustularia microspora Raithelh., Metrodiana, Sounderheft 2: 20. 1983. Mycobank no. 107217.

= Tarzetta microspora (Raithelh.) Raithelh. *Nueva Flora Micológica Argentina* (Stuttgart): 421. 2004.

Gamundí described *P. pseudosylvestris* (as *Galactinia pseudosylvestris* Gamundí) as a large, light-brownish species (20-80 mm diam. \times 12-40 mm deep) that typically fruits on the ground, on fallen twigs and wood (Gamundí, 1975; Gamundí et al., 2004). She described it as having a short stipe and crenulate, irregular margin that is exaggerated in age as the ascomata tend to open up and turn darker. She recorded the spores as ellipsoid and 14.1-18.2(-19.2) \times 7.2-9.6 µm with ornamentation of minute warts. The excipular structure was described as complex and composed of four layers. Mature asci were amyloid with a ring at the tip. Young asci were described as dextrinoid ("pseudoamyloid" in the terminology of Gamundí, 1975). Gamundí et al. (2004) reported *P. pseudosylvestris* from a wide geographical range in Argentina, from Tierra del Fuego to Neuquén province near Bariloche. This is the most commonly encountered species of *Peziza* sensu stricto in southern South America. Photographs and a description are in Furci (2016).

We examined type material of *Pustularia microspora* Raithelhuber (1983), which Raithelhuber (2004) later placed in the genus *Tarzetta*. We found it to be identical to *Peziza pseudosylvestris*. The short, abbreviated description of *Pu. microspora* is similar to that of *P. pseudosylvestris* (Gamundí, 1975) but differs from Gamundí's in a few fundamental points. Raithelhuber described *Pu. microspora* as brown and 40-60 mm with a short stipe and spores $15-18 \times 9-10.5 \mu$ m. He mentioned neither the spore ornamentation nor the amyloid reaction of the asci. He noted a dextrinoid reaction of asci in Melzer's reagent ("aurantiaco-fuscis colorantibus"), which we found only in immature asci. In our examination the spore ornamentation and the amyloid asci were clearly visible in the type specimen of *Pu. microspora* (Fig. 9 D, E, F).

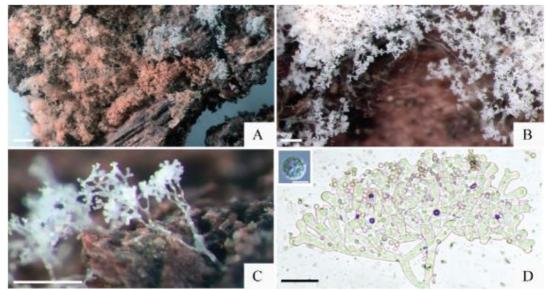


Fig. 10. *Phylloscypha nothofageti*, anamorph all MES-2125. A, habitat in the field, (scale = 200 μ m). B, habitat in the field (scale = 50 μ m) C, magnified view of conidiophores in situ (scale = 50 μ m). D, dichotomously divided conidiophore (scale = 50 μ m), inset is DIC image of mitospore (scale = 5 μ m). Color version at http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

Raithelhuber (1983) deposited the type material in his personal herbarium – "in herb. Raith. cons." with no other indication of the fate of those specimens. We located some of his specimens, including several other type specimens, at the Eidgenössische Technische Hochschule Zürich (ZT) in Zurich, Switzerland. Study of the type specimen of *Pu. microspora* allowed us to compare it with *Tarzetta* specimens from South America. *Pustularia* was the name used previously for species now treated in *Tarzetta. Tarzetta* is included in the Tarzettaceae, species of which lack positive iodine reactions. Recent studies of ectomycorrhizal fungi from Nothofagaceae forests in Chile and Argentina have revealed two distinct species of *Tarzetta* that occur in southern South America (Truong et al., 2017; Van Vooren et al., 2019). The identities and appropriate names for these two *Tarzetta* species still need clarification.

Our ITS phylogeny of *Peziza* sensu stricto from southern South America strongly supported the position of *Pu. microspora* within the *P.pseudosylvestris* clade (Fig. 2) along with specimens identified by Gamundí (e.g., specimen F-570657 sequenced by Lantieri et al., 2016) and our own recent collections (Figs. 1-2). Our morphological analysis of the *Pu. microspora* type specimen revealed noticeably ornamented ascospores, mature asci with a strong amyloid response particularly in the ring area, and young asci that are dextrinoid (Fig. 9). The morphology of the *Pu. microspora* type specimen matches well the original description of *P. pseudosylvestris* and corresponds to the morphology of our fresh collections.

The only exception is that Gamundí (1975) describes the spores as containing two small guttules at the poles or groups of small guttules. We did not detect guttules in our review of the *Pu. microspora* type but this feature is sometimes transient in dried material and difficult to see even in fresh collections. For most specimens we noted small guttules only in young spores in fresh material.

We found P. pseudosylvestris fruiting directly on soil in most cases, but occasionally it was attached to well-decayed woody debris. We never found P. pseudosylvestris in association with fires or burn pits. Gamundí et al. (2004) reported collections from January and February in southern Patagonia but did not provide fruiting dates for other collections (Gamundí et al., 2004). The type specimen of Pustularia microspora was found in late January (Raithelhuber, 1983), and therefore corresponds to this phenology. We collected P. pseudosylvestris both in Spring (e.g., FLAS-F-66043 in October, FLAS-F-63744 in November) and in Fall (e.g., FLAS-F-65087 in April, DHP-CH 2 in March). Our collections also came from a wide geographic area that spanned from Tierra del Fuego in the south to Araucanía, Chile in the north. Our collection FLAS-F-66043 from the Nahuelbuta region in Chile's coastal range is among the northernmost collections that have been reported for this species. Peziza pseudosylvestris may have been confused with several species in the Peziza sensu stricto complex. Gamundí (1975) suggested that confusion might exist with P. sylvestris, Peziza arvernensis and P. cerea (all treated in her publications under Galactinia). If the excipular structure and ascospore ornamentations are not studied, collections could be referred to several ill-defined taxa. See Peziza varia

for further comments on large, smooth-spored taxa.

Specimens examined

ARGENTINA. **Tierra del Fuego**, Dept. Tolhuin, Estancia Ushuaia, cerca de la entrada a Corazon de la Isla, -54.4689 -67.4456, 225 m a.s.l., bosque de lenga, horizonte organico, sotobosque rel. abierto, 23-IV-2015, Camille Truong CT-4274 (FLAS-F-63370, CORD-C6782); Torre Tierra Mayor, 13-III-1975, E. Horak, nro.16, det. Irma Gamundí (FH 00965573); Turbera cercana a Iapataia, 21-II-1974, E. Horak, nro. 74, det. Irma Gamundí (FH 00965572); Dept. Los Lagos, Villa Angostura, site A4.3 near uninvaded site, 818 m a.s.l., on woody debris and soil in a flood plain, 21-XI-2015, Alija Mujic, AM-AR-15-024 (FLAS-F-63744, CORD C00005272); Dept. Río Negro, Nahuel Huapi National Park, Lago Mascardi, 4 km before the lookout area, soil under *Nothofagus dombeyi*, 11-V-2015, Eduardo Nouhra MES-1196 (FLAS-F-63165); Nahuel Huapi National Park, along road to Lago Roca, on moist soil of a cut bank,18-Mar-2012, Donald H. Pfister & Matthew E. Smith DHP-AR-13 (FH 00965569); Cerro Otto Ski

area, on soil and bits of wood, 17-III-2012, Donald H. Pfister DHP AR-1 (FH 00965571); Nahuel Huapi National Park, Ultima Esperanza/Lago Espejo Trail, near Villa La Angostura, Nothofagus dombeyi, on soil, 13-V-2015, Marisol Sanchez MES-1246 (FLAS-F-63199, FH 00965555). CHILE. Magallanes y de la Antártica Chilena, Prov. Magallanes, Punta Arenas, R. Thaxter, n.s. III 1906; Punta Arenas, Río Las Minas near park entrance on trail toward cliff overlook, S 53°8' 45.58 W71° 0' 13.12", on wood and soil, 21-III-2008, Donald H. Pfister & Matthew E. Smith DHP Ch-70 (FH 00284858); Punta Arenas, Club Andino Ski Area, Reserva Nacional Magallanes, Andino Ski Area, Nothofagus forest, on soil by side of trail, 18-Mar-2008 Donald H. Pfister & Matthew E. Smith DHP-CH-2 (FH); Punta Arenas, Club Andino ski area entrance, on soil in forest of Nothofagus betuloides, Nothofagus pumilio, 31-III-2017, Matthew E. Smith MES-2263 (FLAS-F-64945); Río Santa Maria, just south of Reserva San Juan and Fuerte Bulnes, Nothofagus betuloides, 01-IV-2017, Donald H. Pfister & Giuliana Furci MES-2294 (FLAS-64966); Río Santa Maria, just south of Reserva San Juan and Fuerte Bulnes, Nothofagus betuloides dominated forest (some Nothofagus *pumilio*), on disturbed soil, where cows walked, 01-IV-2017, Alija Mujic MES-2306 (FLAS-F-64971); Río Santa Maria, just south of Reserva San Juan and Fuerte Bulnes, on soil, Nothofagus betuloides dominated forest (some Nothofagus pumilio), 05-IV-2017, Alija Mujic MES-2455 (FLAS-F-65087); Punta Arenas, Río Las Minas near park entrance on trail toward cliff overlook, -53.145994 -71.003644, 21-III-2008, D. H. Pfister & M. E. Smith DHP Ch-70 (FH-00284858); Tierra del Fuego, Karukinka Park, La Paciencia Valley, III-2012, G. Furci (FH-00995006); Araucanía, Cordillera de Nahuelbuta, Fundo Rucalemu, in mixed Nothofagus forest, 19-X-2019, Marcos Caiafa & Matthew E. Smith MES-3614 (FLAS-F-66043).

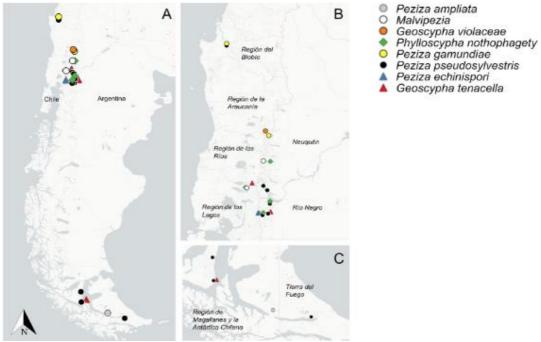


Fig. 11. Map showing the location of sequenced collections included in this study constructed from field recorded GPS locations. Color version at http://www.ojs.darwin.edu.ar/index.php/darwiniana/article/view/1019/1252

Peziza repanda

Wahlenb. ex Fr., Syst. Mycol. (Lundae) 2(1): 51. 1822. Mycobank no. 207813. Although reported in Chile, the concept and application of this name is nebulous. Among the species reported here one might consider *P. varia*, *P. cerea* or *P. pseudosylvestris* as possible names for large specimens with tan or brownish apothecia on wood and woody debris. There is a single record by Palacios (1966) from Chile reporting *P. repanda*. Without sequences the identity remains in doubt.

Peziza retiderma

See *Peziza* cfr. *rifaii*. **"Peziza"** cfr. *rifaii* J. Moravec & B. Spooner, Trans. Br. Mycol. Soc. 90(1): 45. 1988. Mycobank no. 132509. Listed by Gamundí (1966) as *Peziza retiderma* from soil and burned areas in Santa Catalina, Buenos Aires, Argentina. She examined Cooke's specimen of *P. retiderma* collected by Fuller 64 (K), part of the type material collected near Portland, Maine, USA. Moravec & Spooner (1988) revisited the type specimen of *P. retiderma* in their revision of the brown-spored species of *Peziza* and also reexamined the material from New Zealand considered by Rifai (1968) to be *P. retiderma*. They concluded that the New Zealand material represented a new species, *P. rifaii*. Gamundí noted that the spore ornamentation of her collection was generally more tuberculate than reticulate, as is the case for *P. retiderma*. We tentatively use the species name *P. rifaii* for Gamundí's fungus, although the ascospore measurements she provides are slightly smaller than those given by Moravec and Spooner (1988).

The pattern of ascospore ornamentation is similar. Gamundí (1966) reported spore germination and growth in culture, observing that neither conidia nor ascomatal primordia appeared in culture. The South American material of *P. retiderma* was reported on soil in with charcoal and ashes. This is unlike the habitat reported for *P. rifaii*, *P. retiderma* or *P.atrovinosa* (the other brown-spored species treated by Moravec & Spooner, 1988) but recalls *P. vacinii* (Velen.) Svrček. The placement of these brown-spored taxa remains unresolved. They are not part of the *Peziza* sensu stricto group. There is no DNA sequence for material from southern South America. **Peziza succosa** see *Paragalactinia succosa*.

Peziza succosella see Paragalactinia succosella.

Peziza sylvestris

(Boud.) Sacc. & Traverso, Syll. Fung. (Abellini) 20: 317. 1911. Basionym: *Aleuria sylvestris* Boud., Icon. Mycol. 261. 1906. Mycobank no. 166660. Gamundí (1964, 1971) and Lazo (1971) report this species from Argentina and Chile. Gamundí compared her collections with Boudier's type specimen. Her concept is one that encompasses a large species with low isolated warts on the ascospores. She mentions a yellow substance arising from the apothecium in microscopic mounts. Gamundí (1975) distinguishes *P. sylvestris* from *P. pseudosylvestris* by the multilayered excipulum of *P. pseudosylvestris*, which makes it a much more robust species. Hansen et al. (2002) considered *P. sylvestris* a synonym of *P. arvernensis*. See the discussion under *P. varia* for the confusion surrounding species with smooth or nearly smooth-walled ascospores. Most modern treatment of *P. sylvestris* place the name in synonymy of *P. arvernensis*. There is no DNA sequence for material from southern South America. **Peziza tenacella** see *Geoscypha tenacella*.

(Boud.) V. P. Tewari & D. C. Pant, in Pant, J. Mycopathol. Res. 31(1): 22 (1993). Basionym: *Galactinia tosta* Boud. Bull. Soc. Mycol. France 14: 19. 1898. Mycobank no. 189545.

Gamundí (1966) considered this species to be terricolous or carbonicolous. The species was described as medium to large and characterized by ascospores with rounded, somewhat irregular warts. Gamundí included several collections from around Buenos Aires and compared her collections with those identified as *P. tosta* from Europe, including those studied by Boudier. Spores germinated but no anamorphic state was produced in culture (Gamundí, 1966). The identity of this species is unclear. Given the ornamented ascospores with a single guttule it is not part of the *Peziza* sensu stricto group. This collection might be referred to *Phylloscypha*. Further collections and sequencing will be required.

Peziza varia

(Hedw.) Alb. & Schwein., Consp. Fung. (Leipzig.): 311. 1805. Basionym: *Octospora varia* Hedw., Descr. micr. -anal. musc. frond. 2: 22, t.6D 1789 Mycobank no. 226115.

The name *Peziza varia* has been applied in various ways. The reader is referred to Hansen et al. (2002) for a treatment of this complex. A number of species with smooth or nearly smooth-walled ascospores have been reported from southern South America. These include the following: *P.cerea, P. domiciliana, P. micropus* and *P. sylvestris*. All of which have been reported in South America. The complexities surrounding the identity of these species requires detailed study based on morphological, cultural and molecular data to resolve the identities of these taxa. A collection named *P. varia* was reported by Pegler et al. (1980) from the Marble Knolls area of Signy Island in the South Orkney Islands. The collection was cultivated in England on mosses on basic mineral soil (Pegler et al., 1980). This specimen is the basis of the report by Gamundí et al. (2004).

Peziza vesiculosa

Bull., Champ. Fr. pl. 457, fig. 1, 1791; Fries, Syst. Myc. 2: 58. 1821. Mycobank no. 194560.

Gamundí (1960) reported several collections from around Buenos Aires and from Chile (Gamundí et al., 2004) and a collection made by Espinosa is reported by Lloyd (1920).

The first collection from Chile was that reported by Bertero (1829). Hariot (1889)

cites an abundant collection on soil from Punta Arenas and Lebrun's collection from Patagonia. It is listed from Chile by Lazo (1971, 2016). *Peziza vesiculosa* Bull. var. *aparaphysata* Speg. (Spegazzini, 1909) has not been studied and is probably not distinct from *P.vesiculosa*. In several instances Spegazzini reported that paraphyses were lacking in other species of Pezizales. His observation is probably due to the compression of the paraphyses in fully mature specimens; paraphyses are a universal character in the apothecial Pezizales. There is no DNA sequence for material from southern South America.

Peziza violacea see Geoscypha violacea.

Phylloscyphya

Van Vooren, Ascomycete.org 12 (4): 189. 2020.

Phylloscypha nothofageti

(Gamundí) Pfister, Healy & M.E. Smith, comb. nov., Mycobank no 843799. Basionym: *Galactinia nothofageti* Gamundí, *Darwiniana* 13: 578. 1965. Mycobank no. 331044. Fig. 10 A-D.

Gamundí et al. (2004) cites specimens from Neuquén, Río Negro, Argentina and from Tierra del Fuego, Argentina for var. *fuegiana*. Gamundí (1964, 1975) points out that the asci are flattened at the apex and amyloid at the tip and on the lateral walls. The asci have croziers. The species is large and the ascospores are finely warted. Ascospores have one or two small guttules at the poles and measure $16.6-19 \times 7.5$ -9.2 µm. Gamundí reports the species on soil and in burned areas. The outer surface is pustulate; the hymenium is castaneous. Paraphyses have brownish pigments adhering to the outside. Gamundí (1964) describes the apothecium as having a violet base and compares her fungus to Aleuria olivacea of Boudier. The spore markings are similar to those seen in *Phylloscypha phyllogena*, which is an earlier name for Peziza badioconfusa. Peziza badioconfusa was reported in Chile by Mujica et al. (1980) and by Cash (1957). Cash (1957) cites one of Roland Thaxter's collections from Punta Arenas, Chile that was collected in 1906. These collections were found on soil in Nothofagaceae forest near Punta Arenas, Chile (Mar. 1906 (BPI)). Gamundí (1975) described a variety, G. nothofageti var. fuegiana, with larger ascospores, denser ornamentations at the poles and paraphyses that have a brown amorphous external pigment. It seems to be known from a single collection and probably represents an undescribed species.

The new combination is justified on the form of the spore ornamentation, the color

of the hymenium and the violet tinted ascomatal base. This coloration is typical of *P. phyllogena* and related species. Our specimens and DNA sequences from recent collections place the species in *Phylloscypha* and our analyses of specimens agree with the descriptions by Gamundí and allow placement in the genus *Phylloscypha* (Fig. 5).

Several anamorphs of this species were collected on wood, woodland debris, and soil in Nothofagaceae forests (Fig. 10). The spore mats are macroscopically visible as clusters of white hyphae with masses of white spores that mature to light salmon-pink. The hyphae that give rise to conidiophores are horizontal with the substrate, septate, hvaline, and sparsely branching. The conidiophores are smooth, hyaline, mononematous, erect, ≤ 1 mm tall \times 12.2-15.4 µm diam., septate, and serially apically branching in a dichotomous pattern from the top half or third of the main conidiophore. Conidiophore branches hyaline, septate, 8.5-9.4 µm diam. Mitospores are simultaneously holoblastic from short denticles along conidiophore branches, unicellular, mostly globose but some subglobose, finely ornamented with short warts, pink in mass when mature. Mitospores (N=30) are 5.2 ± 0.28 , excluding warts, which are $\leq 0.3 \,\mu m$ tall. Mitospore ornaments are not appreciably darker than the sporoplasm and wall in cotton blue, warts isolated or connected by low ridge with one or two adjacent warts. The conidiophores loosely fit the description and resemble the drawings of the anamorph of Peziza ostracoderma (Hennebert, 1973) and Chromelosporium ochraceum Corda (Hennebert, 2020). Despite the similar anamorph structures, *Peziza ostracoderma* is not closely related to *Phylloscypha*. A major difference in the anamorphs is the warts on the mitospores of *P. nothofageti*, that are connected by a low ridge in places, which is not described for P. ostracoderma mitospores.

There is no teleomorph known for *C. ochraceum*. For the complex history of nomenclature for this species see Hennebert (2020). *Chromelosporium ochraceum* is found in Africa, Asia, and Europe (Hennebert, 2020). The spores are similar in color, shape, size and ornamentation. Perhaps the teleomorph of *C. ochraceum* should be sought among species of *Phylloscypha*. *Chromelosporium arenosum* Hennebert, described from Venezuela, differs in the much shorter conidiophores reported for *C. arenosum* (Hennebert, 2020). Cultures were not attempted.

Specimens examined

ARGENTINA. **Río Negro**. Nahuel Huapi National Park, Colonia Suiza, Arroyo Goye, drainage just upstream, 41 6 25.3 S 71 31 14.3 W, 926 m a.s.l., on soil, under *Nothofagus dombeyi* and *Pseudotsuga menziesii*, 26-IV-2017 Alija Mujic AM-AR17-022 (FLAS-F-63776, CORD-C6689, FH 00965550); anamorph on sticks and soil, in mixed forest of *Nothofagus dombeyi* and *N. pumilio*, 12-V-

2016 Rosanne Healy MES-1852 (FLAS-F-64603, CORD-C6909); anamorph on soil in mixed forest of *Nothofagus antarctica* and *N. dombeyi*, 18-V-2016 R. Healy MES-2125 (FLAS-F-64749, CORD-C6905); **Neuquén**, Lanín National Park, anamorph on soil under *Nothofagus nervosa* and *N. obliqua*, 18-V-2015 R. Healy MES-1355 (FLAS-F-64669, CORD-C6910); CHILE. **Osorno**, Puyehue National Park, below Antillanca, anamorph on water-soaked wood in forest of *Nothofagus pumilio*, 18-V-2016 Matthew E. Smith MES-1765 (FLAS-F-64529).

Rhodopeziza

Hohmeyer & J. Moravec, in Moravec, Czech Mycol. 47: 261 (1995) [1994].

Rhodopeziza tuberculata

(Gamundí) J. Moravec & Hohmeyer, In Moravec, Czech Mycol. 47(4): 261. 1995 [1994]. Basionym: *Aleuria tuberculata* Gamundí, Fl. Criptog. Tierra del Fuego 10: 116. 1975. Mycobank no. 308464.

Gamundí (1975) described a fungus under the name Aleuria tuberculata on soil. The red apothecia range from 20-22 mm diam. The asci were described as lightly amyloid and the paraphyses contained yellow pigments that turned green in Melzer's reagent. The excipulum was composed of globose cells intertwined with hyphae that had granular contents. The spores are ellipsoid, pale yellow, uniguttulate and ornamented with mostly isolated warts. Gamundí compared her fungus to species of *Aleuria* and *Melastiza*, both of which exhibit red, yellow or orange hymenial pigments. She did not mention the discrepancy of finding amyloid asci in these highly carotenoid-pigmented fungi. Carotenoids are uncommon and unverified in the Pezizaceae. There is a single collection, the holotype, which was studied by Moravec (1994) and Hohmeyer. They verify the ascus reaction in Melzer's reagent and describe the bluing as occurring over the entire length of the ascus. They further note that the paraphyses contained orange granules. Further collections of this fungus would greatly aid in a proper placement and understanding its phylogenetic placement. There is no DNA sequence for material from southern South America.

Discussion

The 24 species recorded here from temperate South America compare favorably with findings from Australia and New Zealand (14 species recorded by Rifai, 1968, 18 species from the New Zealand Fungi database, 25 from the Australian Virtual Herbarium) but in these cases the number of species is smaller than reported in north temperate regions (50 species in the Nordic region (Dissing, 2000), 103 reported for the North Temperate regions of the world by Jamoni (1997), and 37 species for China (Zhuang, 1998, 2001, 2003). The number from China certainly represents an undercount. Sánchez-Jardón et al. (2021) recently reported on the fungi of the Aysen Region of Chile. They included five species from their literature search, all of which are included among the species we discuss.

Still the need for further collections is clear as can be seen by our map provided as Figure 11. This indicates the areas where our sequenced material was collected. Large areas remain to be explored. Our presentation here is preliminary and we hope will provide a guide for continued work in the southern regions of South America.

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