



MICROMYCETES ON BARK AND WOOD OF *PODOCARPUS PARLATOREI* (PODOCARPACEAE) FROM ARGENTINA. X. *CHAETOSPHAERIA* (CHAETOSPHAERIACEAE, ASCOMYCOTA)

Micromicetes asociados a la corteza y madera de *Podocarpus parlatorei* (Podocarpaceae) en la Argentina. X. *Chaetosphaeria* (Chaetosphaeriaceae, Ascomycota)

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Summary: Four species of *Chaetosphaeria* were collected during a survey on the biodiversity of microfungi growing on bark and decorticated wood of *Podocarpus parlatorei* in Argentina. A new variety, *Chaetosphaeria hispida* var. *podocarp*, is proposed. *Chaetosphaeria abietis*, *C. montana*, *C. vermicularioides* are reported as new records for Argentina. All species are described and illustrated. Along with data on geographical distribution, habitat and notes on the described fungi, a dichotomous key for the known species in Argentina is provided.

Key words: Argentina, Ascomycota, *Chaetosphaeria*, Chaetosphaeriaceae, Chaetosphaeriales, *Podocarpus parlatorei*.

Resumen: Durante un estudio de la biodiversidad de micromicetes que crecen en la corteza y madera de *Podocarpus parlatorei*, se encontraron cuatro especies de *Chaetosphaeria*. *Chaetosphaeria hispida* var. *podocarp*, se propone como una nueva variedad. *Chaetosphaeria abietis*, *C. montana*, *C. vermicularioides* se citan por primera vez para la Argentina. Se presentan descripciones, ilustraciones, comentarios, datos de distribución geográfica, hábitat y una clave dicotómica para las especies de *Chaetosphaeria* conocidas en Argentina.

Palabras clave: Argentina, Ascomycota, *Chaetosphaeria*, Chaetosphaeriaceae, Chaetosphaeriales, *Podocarpus parlatorei*.

Introduction

The genus *Chaetosphaeria* was established by Tulasne et Tulasne (1863) with *C. innumera* Berk. & Broome ex Tul. & *C. Tul.* as the type species. The genus is rich in species, with 171 records in Index Fungorum (<http://www.indexfungorum.org>). *Chaetosphaeria* had been placed in the Lasiosphaeriaceae (Barr, 1990), and it is currently in the Chaetosphaeriaceae (Réblová et al., 1999) in the order Chaetosphaeriales (Huhndorf et al., 2004; Maharachchikumbura et al., 2015). As a cosmopolitan distributed genus

it has been studied from both temperate and tropical regions (Hyde et al., 1999; Fernández et Huhndorf, 2005; Fernández et al., 2006; Réblová et al., 1999; Perera et al., 2016). It is frequently found growing on wood or bark of different angiosperms and gymnosperms, both in terrestrial and aquatic freshwater and marine habitats (Réblová, 2004; Fernández et al., 2006; Farr et Rossman, 2019). Both states, sexual and asexual are frequently associated (Réblová, 2000; Fernández et Huhndorf, 2005; Huhndorf et Fernández, 2005; Ariyawansa et al. 2015, Perera et al., 2016). There have been few investigations

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on the genus *Chaetosphaeria* in Argentina. Peña et Arambarri (1996) described *Chaetosphaeria chaetosa* Kohlm., this is a marine lignicolous species, now placed in *Marinokulati* E. B. G. Jones & K. L. Pang (Jones et al., 2014). Romero (1999) described *Chaetosphaeria myriocarpa* (Fr.) C. Booth. and *Chaetosphaeria preusii* Gams & Hol.-Jech. on *Eucalyptus viminalis* Labill.

Through our work on the biodiversity of fungal species associated with the native gymnosperm *Podocarpus parlatorei* “pino del cerro” in Argentina (Catania, 2004; Catania et Romero, 2001, 2011, 2014, 2017; Gallo et al., 2018), a new variety and new records of *Chaetosphaeria* species were found growing on dead wood and bark. Here we present descriptions, illustrations and comments regarding the geographic distribution for all the findings. A key to the known species of *Chaetosphaeria* in Argentina is also included.

Materials and Methods

The specimens were collected during mycological explorations in the forests of *P. parlatorei*, in the provinces of Tucumán and Catamarca (Catania et Romero, 2010). The forests belong to the phytogeographic province of the “Yunga” of the Amazonic Dominion (Cabrera et Willink, 1980). Studied specimens are kept at mycological herbarium of Fundación Miguel Lillo (LIL); in addition, a collection kept at the herbarium PRM (Thiers, 2019) was examined.

Observations, digital imaging and measurements of microscopic structures (ascmata, asci, ascospores and asexual structures: conidiophores, conidia) were made on material mounted in distilled water, 5% KOH and 1% phloxine with either an Olympus CX31 microscope and Olympus SP-350 digital camera or a Zeiss Axioskop microscope and a Dage MTI video camera. To observe ascospores and apical rings in the asci, material was mounted in calcofluor and examined with a Zeiss Axioplan epifluorescence microscope according to Romero et Minter (1988). Drawings were made with a camera lucida (Leica MZ6).

Results and Discussion

Chaetosphaeria abietis (Höhnelt) W. Gams & Hol.-Jech., Stud. Mycol. 13: 53. 1976. *Zignoëlla abietis* Höhnelt, in Rehm, Ann. Mycol. 5: 469. 1907. Type: Austria, Dürnwien, Wiener Wald, on inner side of decayed bark, VII-1907, *F. von Höhnelt s/n*, Rehm Ascom. exsicc. N° 1740 (lectotype K 49555).
Fig. 1A-D

Ascomata perithecial, dark brown to black, subglobose, 150-300 × 100-250 µm, superficial, scattered to gregarious, papillate, ostiolate, shining, surrounded by sterile conidiophores. Peridium 35-45 µm thick, with cells of *textura epidermoidea* in surface view; in longitudinal section 2-layered; inner layer composed by elongate to flattened hyaline cells, outer layer of light brown pseudoparenchymatous cells. Asci unitunicate, cylindrical, 110-130 × 8-13 µm, 8-spored. Ascospores fusiform with rounded ends, 3-septate, straight or slightly curved, 25-32 × 3-4 µm, hyaline, each cell with a lipid droplet, smooth-walled.

Distribution and habitat: *Chaetosphaeria abietis* was found in Austria and Czech Republic on wood and bark of conifers: *Abies alba* Mill. and *Picea abies* (Gams et Holubová-Jechová, 1976). Réblová et Gams (1999) cited *C. abietis* on bark of *Abies alba* in Czech Republic, France, Ukraine, Germany and United States of America (www.gbif.org/species). *Podocarpus parlatorei* is recorded here as a new host for the species.

Specimens examined: **ARGENTINA.** **Catamarca:** Dep. Ambato, Las Juntas, in forest of *P. parlatorei*, 28°07'94"S, 65°54'49"W, 1780 m, 26-IX-1999, *Catania 1769* (LIL).

Observations: Detailed descriptions of *Chaetosphaeria abietis* were provided by Gams et Holubová-Jechová (1976) and Réblová et Gams (1999). Our material only differs in the size of the ascomata (ca. 180 µm diám) and of the asci [80-90(-120) × 8-10 µm] which are smaller than those described by Gams et Holubová-Jechová (1976), and in addition to the absence of a hyphal subiculum on which ascomata develop.

Chaetosphaeria abietis is reported for the first time from Argentina.

Chaetosphaeria hispida Réblová & Seifert var. ***podocarpi*** Catania var. nov. Type: Argentina. Tucumán: Taficillo, in forest of *P. parlatoresi*, 26°42'82"S, 65°19'53"W, 29-II-2000, M. del V. Catania 1811 (holotype LIL). IF 555782. Figs. 1E-H, 2A-J

Differs from *Chaetosphaeria hispida* var. *podocarpi* by possessing longer setae (up to 47 µm) and shorter ascospores (40-49.5 µm).

Ascomata perithecial, brown to dark brown, globose to subglobose, 250-450 × 250-400 µm, superficial, scattered to gregarious, slightly papillate, ostiolate, setose. Setae dark brown, aseptate, stiff, pointed, 23-47 × 5.0 µm; abundant setae in young ascomata. Peridium 35-49.5 µm thick, with cells of *textura globosa* to *angularis* in surface view, dark brown; 2-layered in longitudinal section, inner layer of light brown to hyaline, thin-walled flattened elongated cells, outer layer thicker, dark brown, composed of angular-polygonal cells. Paraphyses 2-3 µm wide in the base, unbranched, tapering towards the 1.5 µm wide apex, septate with intervals of 19-22 µm long. Asci unitunicate, cylindrical-clavate, 110.5-156 × 8-9(-10) µm, 8-spored, rounded at the apex, with refractive apical ring. Ascospores cylindrical-filiform, 7-septate, straight or slightly curved, with rounded ends, slightly tapering to base, (35.5-)40-49.5(-51) × 3-4 µm, hyaline, 2-seriate, smooth-walled. Conidiophores macronematous, mononematous, erect, straight or flexuous, cylindrical, unbranched, septate, smooth, brown, becoming light brown towards the apex, 221-240 µm long, 5-9 µm diam in the base, 4-6 µm diam upwards. Conidiogenous cells terminal, integrated. Conidia acrogenous, cylindrical, straight to slightly curved, apically rounded, truncated at the base, 13-17 × 5-8 µm, 3-septate, central cells brown to dark brown and distal cells pale brown to subhyaline, smooth.

Etymology: referring to the associated plant, *Podocarpus*.

Distribution and habitat: *Chaetosphaeria hispida* is known from the type collection from Thailand, on rotten wood (Réblová et Seifert, 2003). Here it is recorded on bark and wood of *P. parlatoresi* from Argentina.

Specimens examined: **ARGENTINA**. **Tucumán**: Dep. Tafí Viejo, Taficillo, in forest of *P. parlatoresi*, 26°42'82"S, 65°19'53"W, 1600 m, 1-III-1999, *Catania 1325* (LIL); idem, 24-V-1999, *Catania 1661, 1662* (LIL); idem, 27-XI-2000, *Catania 1940* (LIL). **THAILANDIA**. **Nakhon Nayok**: Khao Yai National park NE of Bangkok, Bung Phai trail ca. 5 Km NW from Khao Yai forest Headquarters on a way to Pak Chong, 14°28'N 10°23'E, 750 m, MR 2220/01; on decayed wood of a branch, 6-IX-2001, Réblová, Samuels & Nasit (holotype PRM 900543).

Observations: *Chaetosphaeria hispida* var. *podocarpi* occurs on wood and bark. Typically, ascospores are 7-septate and scolecosporous ascospores, and the ascomal wall has a *textura globosa* to *angular* disposition of cells in surface view.

The argentine collections agree with the morphological characteristics of *C. hispida* (PRM 900543!, Réblová et Seifert, 2003) in the setose ascomata and ascospore features such as shape, number of septa and color, but differ because setae are longer (up to 47 µm vs. up to 37 µm) and ascospores shorter (40-49.5 µm long vs. 74-82 µm long), so I thus propose a new variety.

Several species of *Chaetosphaeria* form anamorphs similar to those described by Réblová et al. (1999) and Seifert et al. (2011), and the variety of conidiophores and conidia when they are grown *in vitro* is huge (Réblová et Seifert, 2003).

Asexual structures were found around the ascomata of *C. hispida* var. *podocarpi*. Réblová et Gams (1999) described *C. decastyla* (Cooke) Réblová & W. Gams with similar conidiophores arising from the substratum. The asexual conidiophores in the present material were considered to be related to the ascomata of *C. hispida* var. *podocarpi*. In *C. hispida*, no asexual state was observed on the natural material.

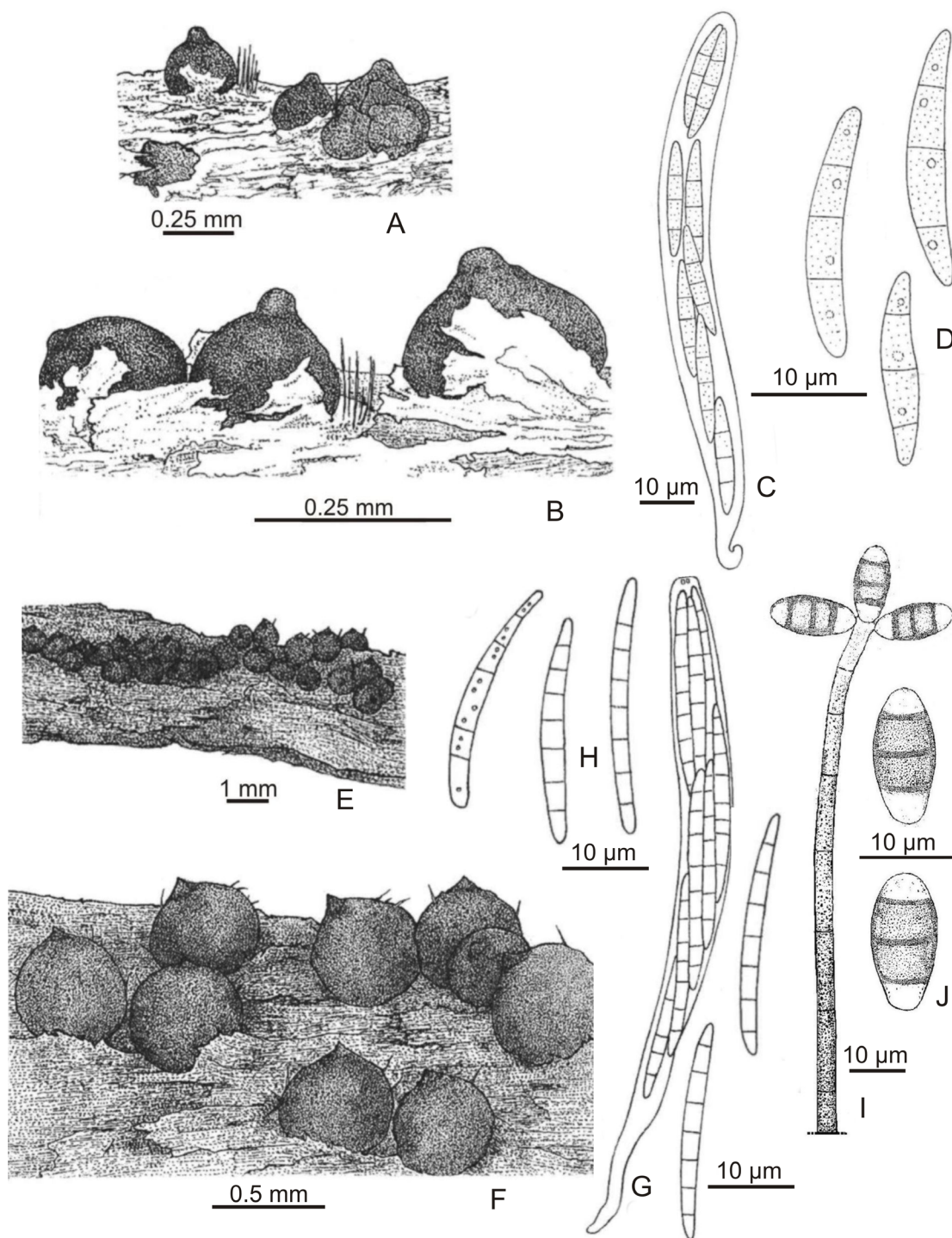


Fig. 1. *Chaetosphaeria abietis*. A-B: Ascomata on substrate. C: Ascus. D: Ascospores. *Chaetosphaeria hispida* var. *podocarpi*. E-F: Ascomata on substrate. G: Ascus. H: Ascospores. I: Conidiophore, conidia. J: Conidia. (A-D, *Catania 1769*, LIL; E-J, *Catania 1811*, LIL).

Fig. 1. *Chaetosphaeria abietis*. A-B: Ascomas sobre el sustrato. C: Asco. D: Ascosporas. *Chaetosphaeria hispida* var. *podocarpi*. E-F: Ascomas sobre el sustrato. G: Asco. H: Ascosporas. I: Conidióforo, conidios. J: Conidios. (A-D, *Catania 1769*, LIL; E-J, *Catania 1811*, LIL).

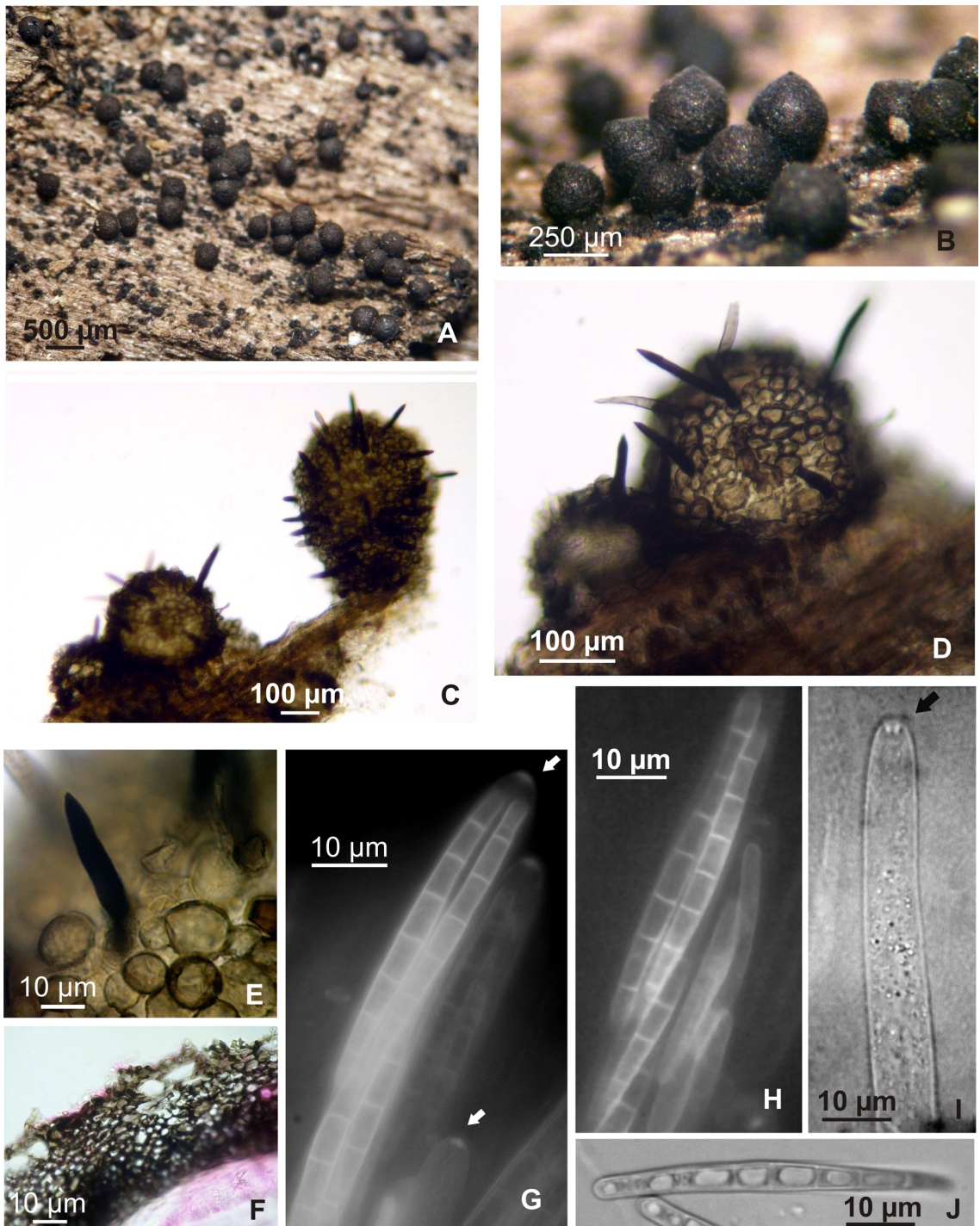


Fig. 2. *Chaetosphaeria hispida* var. *podocarp*i. A-B: Ascomata on substrate. C-D: Young ascomata with setae. E: Seta. F: Peridium, longitudinal section. G: Ascus, detail of apical ring, arrow (EF). H: Ascospores (EF). I: Ascus, refractive apical ring (arrow). J: Ascospore. (A-J, *Catania 1811*, LIL).

Fig. 2. *Chaetosphaeria hispida* var. *podocarp*i. A-B: Ascomas sobre el sustrato. C-D: Ascomas jóvenes con setas. E: Seta. F: Peridio, sección longitudinal. G: Ascus, detalle del anillo apical, flecha (EF). H: Ascosporas (EF). I: Asco, anillo apical refringente (flecha). J: Ascospora. (A-J, *Catania 1811*, LIL).

Chaetosphaeria montana Réblová, Czech Mycol. 50(3): 153. 1998. Type: Czech Republic, Southern Bohemia, Novohradské hory Mts., virgin forest Zofinsky prales near Pivonice; on rotten wood of *Fagus sylvatica*, 20-V-1997, M. Réblová (holotype PRM 842969. Fig. 3A-F

Ascomata perithecial, dark brown, subglobose, 150-250 × 140-200 µm, slightly immersed to superficial, scattered or in small groups, papillate; sterile conidiophores among the ascomata. Peridium 30-40 µm thick, with cells of *textura epidermoidea* in surface view, 2-layered in longitudinal section, inner layer of thin-walled hyaline elongate cells, outer layer of thick-walled, brown to dark brown pseudoparenchymatous cells. Paraphyses 3-4,5 µm wide, numerous, simple, filiform, septate. Asci unitunicate, cylindrical, 65-95 × 8-11 µm, 8-spored. Ascospores ellipsoid to fusiform, 1-septate to 3-septate at maturity, slightly constricted, 17-21 × 5-6,5 µm, hyaline, guttulate, smooth-walled.

Distribution and habitat: *Chaetosphaeria montana* was originally found in the Czech Republic, on decayed wood of *Fagus sylvatica* L. (Réblová, 1998). Up to the present, the species was known only from the type specimen, and from now, growing on bark of *P. parlatoarei* from Argentina.

Specimens examined: **ARGENTINA.** **Catamarca:** Dep. Ambato, Las Juntas, in the forest of *P. parlatoarei*, 28°07'94"S, 65°54'49"W, 1780 m, 16-VI-1999, *Catania 1678* (LIL).

Observations: *Chaetosphaeria montana* was described by Réblová (1998). Our material agrees well with the original description, differing only in that both ascospores [17-21 × 5-6.5 µm vs. (18.5-) 19-25 × 5.5-7 µm] as asci (65-95 × 8-11 µm vs. 100-130 × 10.5-12.5 µm) are slightly smaller. The species is reported for the first time from Argentina, also this record constitutes the second world.

Taking into account the differences in

the geographic location, habitat, host and morphological features, it would not be rare to be in front of a new species. To confirm or deny this hypothesis, more samplings and molecular and phylogenetic studies are needed.

Chaetosphaeria vermicularioides (Sacc. & Roum.) W. Gams & Hol.-Jech., Stud. Mycol. 13: 15. 1976. *Eriosphaeria vermicularioides* Sacc. & Roum., Revue mycol. 5: 235. 1883. Type: Belgium, Malmédy, Fungi Gall. Exs. 2693 (isotype L 910.236-665). Fig. 3G-J

Ascomata perithecial, dark brown to black, subglobose, 130-200 × 120-180 µm, superficial, gregarious, papillate, ostiole slightly conical. Peridium 20-25 µm thick, *textura epidermoidea* in surface view, 2-layered in longitudinal section, inner layer composed of light brown elongate cells, outer layer of dark brown pseudoparenchymatous cells. Asci unitunicate, cylindrical, 45.5-61 × 3-5 µm, 8-spored, thin-walled. Ascospores ellipsoidal-fusiform, (6-)6.5-8 × 2.5-3 µm, hyaline, smooth-walled, septum in the middle, breaking apart in a rather early stage within the ascus. Conidiophores macronematous, mononematous, unbranched, septate, brown, paler towards the top, 65-130 µm × 2.5-4 µm. Conidiogenous cells phialidic, hyaline, 2.5-3 µm wide, collarette inconspicuous. Conidia ellipsoidal, hyaline, unicellular, 3.5-4(-4.5) × 2-2.5 µm.

Distribution and habitat: Belgium, Czechoslovakia, Denmark, France, Great Britain, Netherlands (Gams et Holubová-Jechová, 1976); Australia, Brazil, Germany, New Zealand, Mexico (www.gbif.org/species). Ellis et Ellis (1997) reported it on decay wood of *Alnus glutinosa* (L.) Gaertn., *Fraxinus excelsior* L. and *Quercus* sp. Gams et Holubová-Jechová (1976) mentions different substrates. Romero (1983) described the asexual state from wood and bark of *Eucalyptus viminalis* Labill. in Argentina. In this work, we report *Chaetosphaeria vermicularioides* on wood of branch of *P. parlatoarei* as a new host

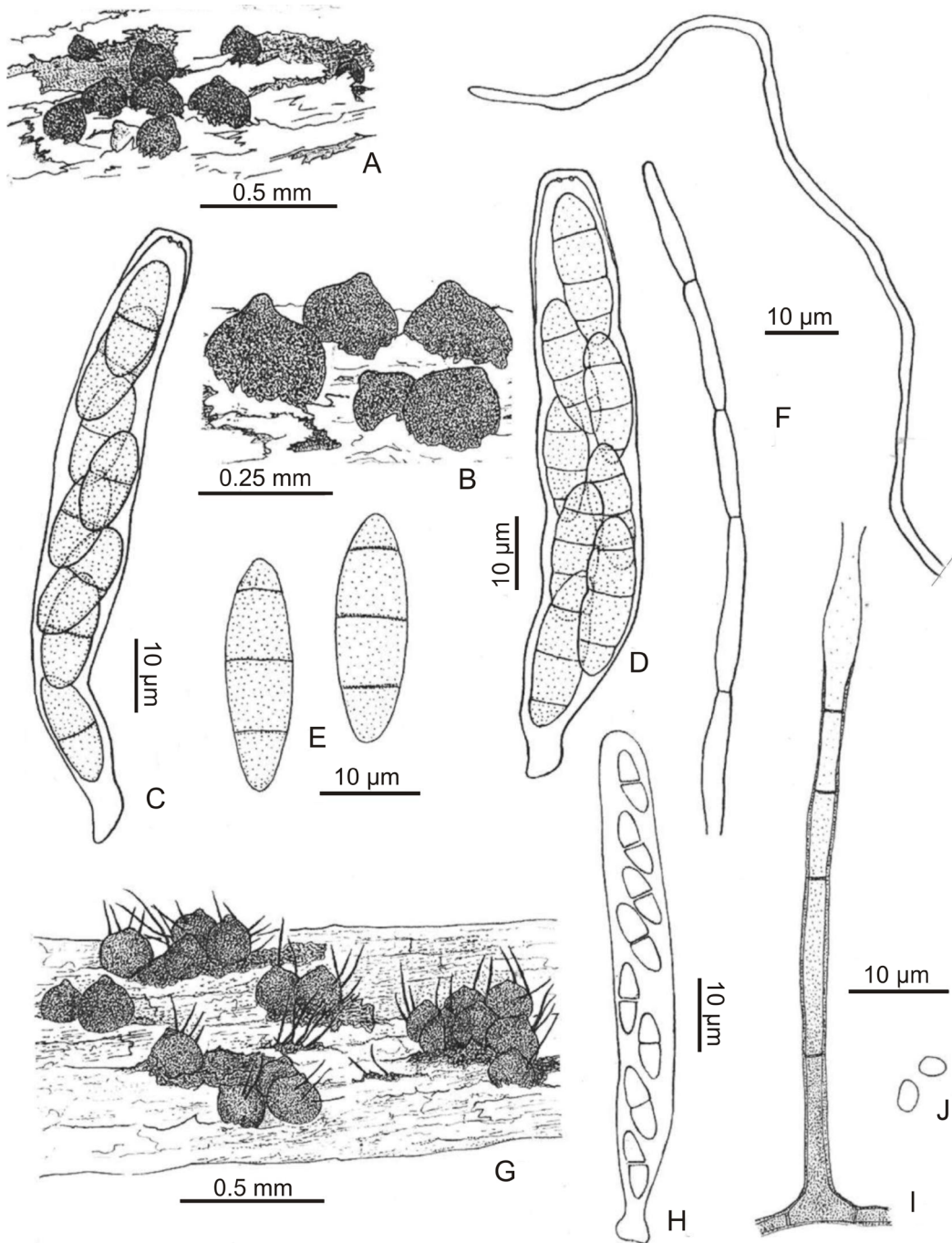


Fig. 3. *Chaetosphaeria montana*. A-B: Ascomata on substrate. C: Ascus with young ascospore, 1-septate. D: Ascus with mature ascospore, 3-septate. E: Ascospore. F: Paraphyses. *Chaetosphaeria vermicularioides*. G: Ascomata, with conidiophore. H: Ascus. I: Conidiophore, conidiogenous cells phialidic. J: Conidia. (A-F, Catania 1678, LIL; G-J, Catania 1935, LIL).

Fig. 3. *Chaetosphaeria montana*. A-B: Ascomas sobre el sustrato. C: Asco con ascospora joven, 1-septada. D: Asco con ascospora madura, 3-septada. E: Ascosporas. F: Paráfisis. *Chaetosphaeria vermicularioides*. G: Ascomas, con conidióforos. H: Asco. I: Conidióforo, célula conidiógena fialídica. J: Conidios. (A-F, Catania 1678, LIL; G-J, Catania 1935, LIL).

plant. The distribution area of the species is extended to the northwestern Argentina.

Specimens examined: **ARGENTINA.**
Tucumán: Dep. Burreyacu, Sierra de Medina, ruta provincial 310, at 31 km from Villa Padre Monti, Aguas Negras, Finca Mansilla, in forest of *P. parlatorei*, 26°22'06"S, 65°03'46"W, 1600 m, 22-XI-2000, *Catania 1935* (LIL). **Buenos Aires:** Pdo. de San Pedro, Gdor. Castro, on wood and bark of *Eucalyptus viminalis*, XI-1982, *Romero 18* (BAFC).

Observations: Our material agrees with the description given by Gams et Holubová-Jechová (1976), but it has slightly longer conidia (3.5-4.0(-4.5) µm vs. 2.5-3.8 µm).

Chaetosphaeria vermicularioides can be confused with *C. inaequalis* (Gove) W.

Gams & Hol.-Jech, but it differs mainly in ascospore size [part spores 3-4.5 µm long. vs. 2.5-3.5(-4.5) µm long].

Conidia described here are slightly larger than those in Romero (1983) (3.5-4 × 2-2.5 µm vs. 2-3 × 1.5-2 µm).

In Argentina, Arambarri et al. (1981) cited *Chloridium lignicola* (Mangenot) W. Gams & Hol.-Jech. growing on leaf litter of *Nothofagus dombeyi* (Mirb.) Oerst., in the Quetrihué Peninsula (Neuquén Province). Currently, this asexual name is considered the asexual state of *Chaetosphaeria*. Godeas et Arambarri (2007) described asexual structures of *Chaetosphaeria lentomitae* W. Gams & Hol.-Jech. on decorticated wood of *Quercus petraea* (Matt.) Liebl., *Fagus sylvatica* L., *Pinus* sp, from Tierra del Fuego Province.

Chaetosphaeria vermicularioides is described for the first time for Argentina.

Key to *Chaetosphaeria* species reported in Argentina

- 1. Ascospores 1-septate or 1-3-septate. 2
 - 2. Ascospores never fragmenting into part-spores. 3
 - 3. Ascospores cylindrical with rounded ends, hyaline, 1-septate, 5-7 × 1.5-2.0 µm. Conidiophores simple, with simple phialides; conidia truncate at the base and rounded at the tip, in short chains. *C. myriocarpa*
 - 3'. Ascospores ellipsoidal to fusiform, 1-3-septate, hyaline, guttulate, 17-21 × 5-6.5 µm. Conidiophores scattered among the sterile setae; conidiogenous cells phialides mono- or polyphialidic; conidia fusiform, falcate with pointed ends. *C. montana*
 - 2'. Ascospores always fragmenting into part-spores. 4
 - 4. Ascospores ellipsoidal-fusiform, 1-septate, part-spores 3-4.5 × 2.5-3 µm. Conidiophores unbranched; conidiogenous cells phialidic with multiple loci apertures; conidia ellipsoidal. *C. vermicularioides*
 - 4'. Ascospores cylindrical to ellipsoidal, 1-septate, part-spores 2.5-3.5 × 1.5-2.0 µm. Conidiophores simple; conidiogenous cells phialidic with single loci aperture; conidia ellipsoidal. *C. preussii*
- 1'. Ascospores 3-septate or more than 3-septate. 5
 - 5. Ascospores fusiform with rounded ends, 3-septate, 25-32 × 3-4 µm, guttulate; ascomata glabrous. Conidiophores simple; conidiogenous cells phialidic; conidia ellipsoidal-cylindrical, base truncate. .. *C. abietis*
 - 5'. Ascospores cylindrical- fusiform, 7-septate, with rounded ends, (35.5-)40-49.5(-51) × 3-4 µm; ascomata setose. Conidiophores unbranched; conidiogenous cells terminal, integrated; conidia cylindrical. *C. hispida* var. *podocarp*

Acknowledgements

I want to thank the financial support by Fundación Miguel Lillo. Also I am grateful for the help of Lic. Inés Jaume (Iconography Section, Fundación Miguel Lillo) for the ink drawings; to the herbarium PRM for providing the loan of material. My special gratitude to Dr. Andrea I. Romero, for her critical reading of the manuscript, and her valuable suggestions.

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