MYCOTAXON

Volume 93, pp. 401–406 *July–September 2005*

A new species of *Leptosphaeria* (Ascomycotina, *Pleosporales*) on *Rosaceae* from Bolivia

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Abstract—*Leptosphaeria polylepidis* is described as a new species on *Polylepis tarapacana* from Sajama National Park in the Bolivian Andes at more than 4,000 m elevation. Diagnostic features are the long asci and the large, brown, 3-septate ascospores.

Key words— biogeography, conservation, rDNA, taxonomy

Introduction

The species of the genus *Polylepis* Ruiz & Pav. (*Rosaceae*: tribe *Sanguisorbeae*) grow naturally at high elevations, usually higher than any other arborescent plant, and occur in South America with the highest number of species in Bolivia, Ecuador, and Peru. It is an important plant in preventing soil erosion and land degradation as well as providing a good source of fuel and building supplies for local communities. Species of this genus are endangered in the Andean high regions and are listed as a primary genus to use in reforestation projects in the Andes (Brandbyge & Holm-Nielsen 1986). However, the regeneration of *Polylepis* largely has been unsuccessful due to cultural and biological factors, and forests continue to disappear at an alarming rate (Kessler & Driesch 1993). Thus, it is important to determine the regenerative needs of this genus as well as to identify pathogens that might reduce vigor and complicate regeneration efforts.

The Sajama National Park is located in the Bolivian Andes (Sajama Province, Oruro Department) and is a small reserve (1002 km^2) created in 1939 to protect the *Polylepis tarapacana* Phil. vegetation formation. This small to medium sized tree or shrub, which is 1-3 m tall and popularly called keñua, is the main component of the world's highest woody plant formation at 5100 m (Jordan 1980; Kessler 1995). The mean annual temperature in the Sajama village is around 10 ºC (range between –30 to 22 ºC) and the mean annual precipitation is near 280 mm (range between 90-400 mm). In the Bosque de Keñua (18º08'S; 68º57'W), at 4300 m and likely with the highest

density of *P. tarapacana* in the region, some of the shrubs had black knots in both the apical and basal parts of branches (Fig. 1a). During a study of the systematics of *Polylepis* in Bolivia many branches were observed that were malformed due to black, irregular growths (Kessler, personal communication). It is likely that this disease killed a number of *P. tarapacana* trees but no published reports of these malformations or disease were found.

The growths on *Polylepis* resemble the black knot on plums and cherries caused by *Apiosporina morbosa* (Schwein.) Arx (≡*Dibotryon morbosum* (Schwein.) Theiss. & Syd) (Ellis 2002). These growths also resemble those caused on the same host by *Grandigallia dictyospora* Barr et al. (1987). However, *Gr. dictyospora* produces very large galls, from 3-14 cm diam, and has larger ascospores that become densely muriform and break down internally, eventually producing numerous conidia inside the ascospore. In this paper, the probable casual agent of black knot on *P. taracapana* is described as a new species and relationships discussed based on molecular analyses.

Materials and methods

Morphological characters were observed macroscopically and microscopically. All measurements of microscopic structures were made on material mounted in water. Light micrographs were captured using a QImaging MicroPublisher digital camara (QImaging, Burnaby, BC, Canada) that was mounted on an Olympus BX51 compound microscope as described in Díeguez-Uribeondo et al. (2003). All material is deposited in the herbarium MA-Fungi (Real Jardín Botánico de Madrid, Spain). Efforts to obtain this species into pure culture were unsuccessful.

The internal transcribed spacer regions of nrDNA (ITS1 and ITS2), including the 5.8S, were amplified using the primer pair ITS 1F (Gardes & Bruns 1993) and ITS 4 (White et al. 1990). All protocols are described in Martín, Raidl & Tellería (2004). Nucleotide BLAST searches with the option Standard nucleotide-nucleotide BLAST of BLASTN 2.26 were used to compare the sequence obtained in this study against other sequences in the National Center for Biotechnology Information (NCBI) nucleotide databases (Altschul et al. 1997). The new consensus sequence has been accessioned in the EMBL database with the Accession Number AJ786644.

Results

Leptosphaeria polylepidis M.J. Macía, M.E. Palm & M.P. Martín **sp.nov.**

Figs. 1-2

Asci cylindrical-clavate, 8 spori, 185–200 x 28–35 µm. Ascosporae fusiformes, brunneae, 3-septatae, 50–55 x 12–14 µm. Parasitatur in Polylepis tarapacana *Phil., loco dicto Parque Nacional Sajama, Bolivia, supra 4300 m, IV/2002, M.J. Macía (Holotypus MA-Fungi 57843).*

Etymology: from the name of the host *Polylepis tarapacana*

Ascomata aggregated, botryose, on well-developed, black stroma intermixed with plant tissue, superficial, black, surface cracked, 310–360 μm diam, 230–320 μm high,

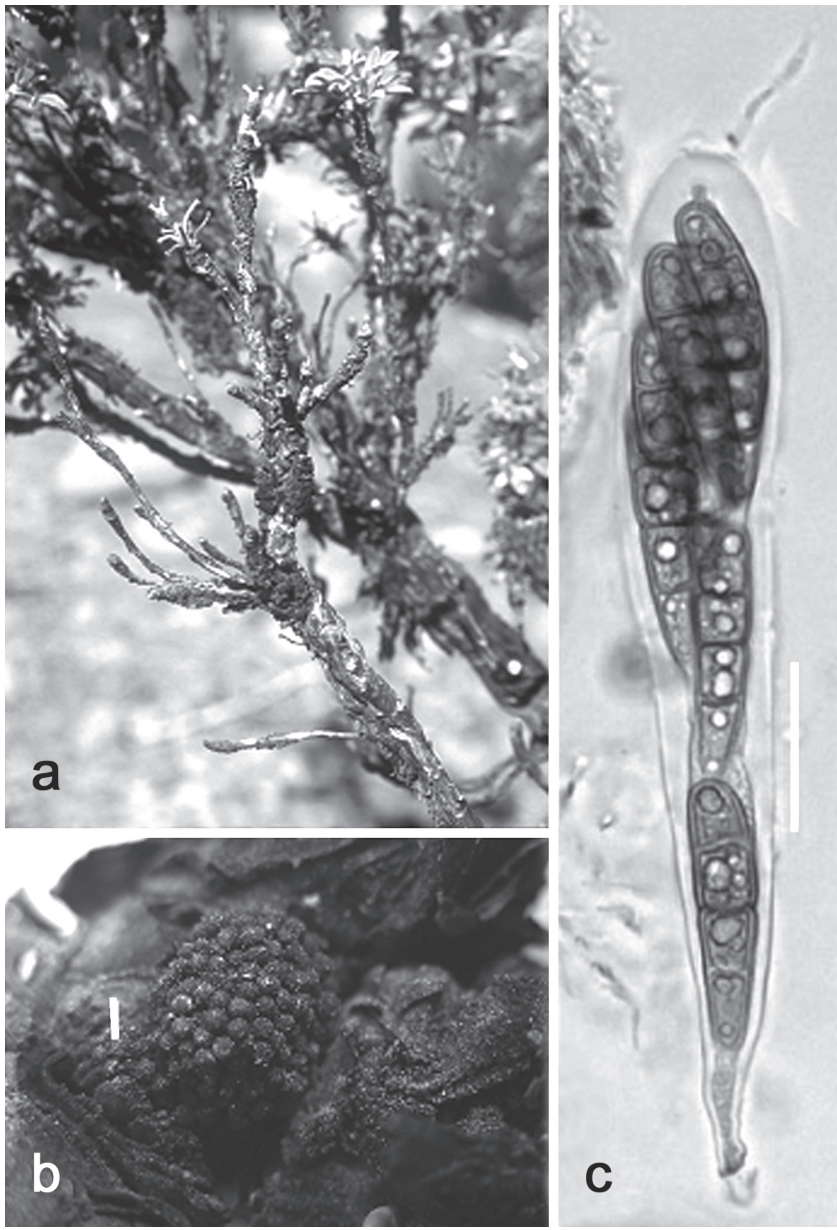

Fig. 1 *Leptosphaeria polylepidis*. a) Stromatic black knots on branches of *Polylepis tarapacana*. b) Papillate ascocarps (MA-Fungi 57843) (Bar=2 mm). c) Mature ascus (MA-Fungi 57843) (Bar=20 μm).

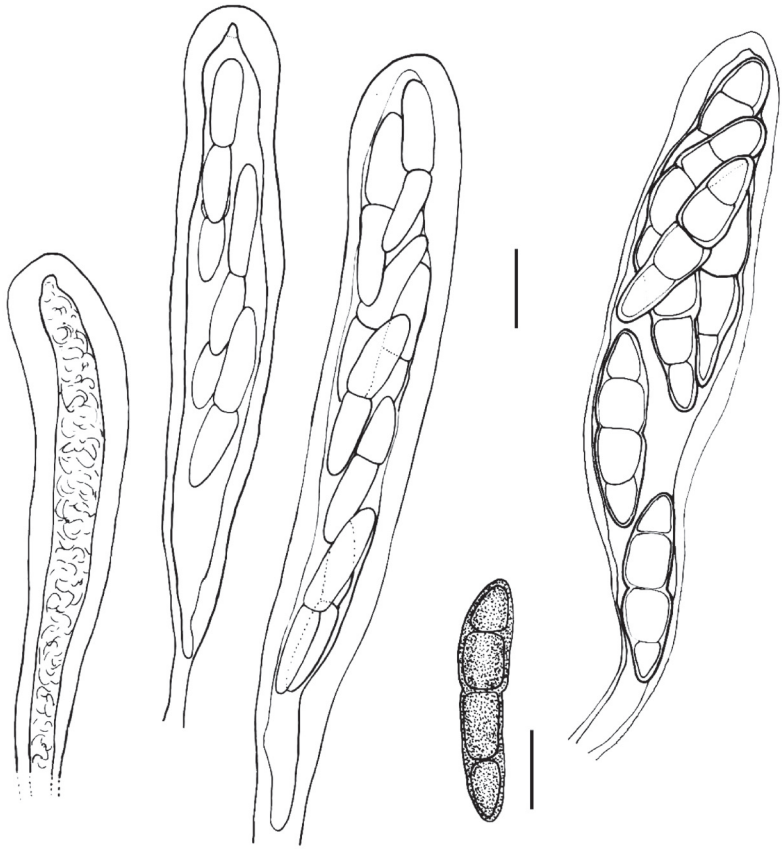

Fig. 2 Line drawings of *Leptosphaeria polylepidis*. a) Asci. b) Ascospore (Bar= 20 μm).

with broadly rounded papilla. Ascomatal wall of *textura angularis* in surface view; in longitudinal section 60–72 μm thick, of 4–5 layers of polygonal, isodiametric to slightly elongate cells, 18–20 x 9–11 μm, all layers with brown-melanized cells of scleroplectenchyma. Pseudoparaphyses 190-210 x 3.0–4.5 μm wide, numerous, narrowly cellular, without gelatinous coating. Asci 185–200 x 28–35 μm, numerous, basal, cylindrical-clavate, with 8 overlapping, uniseriate ascospores. Ascospores when immature 41–50 x 9–10 μm, hyaline to subhyaline, at maturity 50–55 x 12–14 μm, brown, narrowly fusiform, end cells acute, transversely 3-septate.

Material examined: Bolivia, Parque Nacional Sajama, on *Polylepis tarapacana*, 4300 m elev., IV/2002, leg. M.J. Macía 7507 (MA-Fungi 57843) (HOLOTYPE); 4800 m elev., IV/2002, leg. M.J. Macía 7508 (MA-Fungi 57842).

The Blast search of the sequence AJ786644 of *L. polylepidis* shows 92% similarity (392/426) to the sequence AF439461 of *Leptosphaeria dryadis* Rostr. (CBS 643.86) (Câmara et al. 2002).

Discussion

Huhndorf (1992) examined 28 species on *Rosaceae* with names in *Leptosphaeria* and found that five of those species belonged to *Leptosphaeria* Ces. & De Not. sensu Huhndorf. *Leptosphaeria polylepidis* differs from the species treated by Huhndorf (1992) namely *L. cercocarpi* Syd. & P. Syd., *L. doliolum* (Pers.) Ces. & De Not., *L. dryadis (*as *L. dryadophila* Huhndorf), *L. praetermissa* (P. Karst.) Sacc. and *L. umbrosa* Niessl, in the long asci and large, dark brown, 3-septate ascospores that are characteristic of this new species.

Based on comparison of the ITS sequence from this organism with the sequences in Genbank (EMBL), this species is most similar to *Leptosphaeria dryadis*. Chen et al. (2002) pointed out that *L. dryadis* is the correct name for *L. dryadophila* (basionym: *Melanomma dryadis* Johanson). *Leptosphaeria dryadis* occurs on *Dryas octopetala* L. at high latitudes mainly in Europe and is more common in arctic and subarctic areas than alpine zones according to Chlebicki and Suková (2004). *Leptosphaeria polylepidis* is ecologically similar to *L. dryadis* in that it grows at high altitudes.

Acknowledgments

Thanks to Dr. Amy Y. Rossman for her suggestions and comments during the preparation of this manuscript and Dr. Marcos P.S. Câmara for prepublication review. Thank you to Dr. Javier Dieguez-Uribeondo (RJB, Madrid) for Fig. 1c and to Dr. Miguel A. García for Fig. 1b. Thanks to Luis Miguel Monje and Teresa Valdecantos for their help in the fieldwork. This study was supported by Consejería de Educación, Comunidad de Madrid, Spain (to MJM).

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