

Short communication

Morphological features of *Camarosporium pini* – the fungus associated to health state degradation in Austrian and Ponderosa pine

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Abstract

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The subject of this study is escalated occurrence of the pathogenic fungus *Camarosporium pini* in the needle tissue of symptomatic trees *P. nigra* and *P. ponderosa* var. *jeffreyi* growing in urbanized settings and parks. *C. pini* induces severe infections and initiates a blight and premature loss of second-year foliage in pine trees. The fungus was identified microscopically and on base of morphological keys. The affected needles displayed a distinct bluish-grey necrotic band in the centre. On the surface of infected needles, there were formed pycnidia producing brown, oval conidia with three transversal and one or two vertical walls. Disease symptoms, some important characteristics in pure culture, and distinctive morphological features of *C. pini* associated to the health state degradation in Austrian and Ponderosa pine are described and compared. Cumulative effects of these stressful biotic and various abiotic factors may explain the current situation concerning the decline in the *P. nigra* and *P. ponderosa* var. *jeffreyi* in Slovakia.

Keywords

Camarosporium pini, *Pinus nigra*, *P. ponderosa* var. *jeffreyi*

Introduction

Needle and shoot blights in pines may be caused by a number of fungi. Within these, the fungus *Camarosporium pini* (Westend.) Sacc. (Dothidiomycetes), syn. *Hendersonia pini* Westend. induces a severe infection which may result in a significant growth reduction (SUTTON, 1980; FARR and ROSSMAN, 2014; IVANOVÁ, 2015). MINTER (1981) discusses *Camarosporium pini* as a common species, often associated with *Pinus radiata* D. DON. (MINTER, 1977; DENNIS, 1995), *P. sylvestris* L. (FRANCIS, 1975; MINTER et al., 1978; DENNIS, 1995; ELLIS and ELLIS, 1997; KARADŽIĆ and MILJAŠEVIĆ, 2008; PASTIRČÁKOVÁ et al., 2014), *P. nigra* Arnold (FRANCIS, 1975; MINTER, 1979; IVANOVÁ and BERNADOVIČOVÁ, 2010; IVANOVÁ, 2015) and *P. contorta* Dougl. (JAMES, 1984).

This micromycete parasitizes needles of trees debilitated by low temperatures in winters and by drought

periods occurring from spring to summer seasons. The dry needles exhibit little back spots – pycnidia arranged linearly and parallel with the nervure (GROVE, 1937; IVANOVÁ and BERNADOVIČOVÁ, 2010; IVANOVÁ, 2015). Thus, combined effects of stressful abiotic factors predisposing trees health state together with enhanced occurrence of several pathogenic fungi traditionally known as secondary pathogens may explain the current situation in the pine tree decline in Slovakia.

The aim of this study was to investigate distribution, disease symptoms, some important characteristics in pure culture, growth and morphological attributes, distinctive morphological features in *Camarosporium pini* as a causative agent involved to different degrees in the health state degradation of *Pinus nigra* Arnold and *P. ponderosa* var. *jeffreyi* Peter & Charles Lawson in urbanized settings and park.

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Materials and methods

Fresh decaying plant materials were collected from localities in the Nitra town and in the Arborétum Mlyňany. Altogether 30 trees between 35–40 years were sampled. The experiments and morphometric measurements were carried out with implementing a widely accepted methodology (IVANOVÁ and BERNADOVIČOVÁ, 2010; IVANOVÁ, 2015). The identification of fungus was performed using morphological keys according to MINTER et al. (1978); LUO et al. (2010) or morphological studies in JAMES (1984) and BOTELLA et al. (2010).

Results and discussion

Pinus nigra and *Pinus ponderosa* var. *jeffreyi* were disabled by the disease caused by the micromycetous fungus *Camarosporium pini*. This fungus initiates a blight and premature loss in the second-year foliage. The affected needles displayed a distinct bluish-grey necrotic band in the centre. A distinct black line often delimited the necrotic area from green tissues (JAMES, 1984). Pycnidia 180–300 (500) μm in diam. visible on the surface of infected needles were distinctly convex, subglobose, roundish or elongated and black. Brown and oval conidia with three transversal and one or two vertical walls were generally rounded at both ends, often slightly curved and coming out through a pycnidium subpapilliform pore (IVANOVÁ and BERNADOVIČOVÁ, 2010; IVANOVÁ, 2015).

Culture characters: Colonies on PDA of *Pinus ponderosa* var. *jeffreyi* reaching 3 cm diam. after one week at 24 °C. This result is comparable with the data obtained during cultivation of *Pinus nigra* injured needles with colonies on PDA slow growing, 2.5–3 cm after

one week at 24 °C. White mycelium gradually turned to dark grey (Fig. 1a). The reverse side was white, later becoming dark green to olive green from the centre (IVANOVÁ and BERNADOVIČOVÁ, 2010; IVANOVÁ, 2015). Our colonies obtained during cultivation of *Pinus ponderosa* var. *jeffreyi* injured needles formed dense mycelium, circular, with rough margin, white at first, later grey or dark grey, olive-green after 5 days in the centre of the colony, surrounded by white mycelium, flat on the surface, without aerial mycelium. The hyphae were septate branched, hyaline, the pycnidia up to 0.5 mm diam., black, erumpent, globose, separate, ostiolate, papillate, conidiophores short, simple, spherical, swelling the epidermis by pustules (Fig. 1b, c). The spores from pycnidia were produced within necrotic needles on short, hyaline conidiophores. The conidia were oblong, relatively large, dark, golden brown at maturity, ovoid to ellipsoid or elongate to fusoid, non-constricted at the septa, several-celled with 2–3 occasionally 4 transverse septa and 1–2 longitudinal septa 20 (–22) 28–30 \times 10 (–12) 15 μm in size, narrowly rounded at both ends, smooth walled (Fig. 1e). The mature conidia, apparently coated with a self-adhering material, were exuded through minute ostiolar openings situated by one on pycnidia penetrating the host epidermis. When wetted, the individual spores would separate and spread.

On the other hand, the conidia obtained during our cultivation of *Pinus nigra* injured needles were pale brown, thin-walled, with three or four transverse walls and one, sometimes two vertical walls, smooth fusiform to fusiform elliptical, straight, apex subobtuse, base truncate 20–22 \times 6–8 μm (Fig. 1d) (IVANOVÁ and BERNADOVIČOVÁ, 2010). Our results are morphologically similar to *Camarosporium pini* from *Pinus contorta* (JAMES, 1984) and to *C. pini* from *P. sylvestris* (ELLIS and ELLIS, 1997). The comparison of our results with the results of other authors is in Table 1.

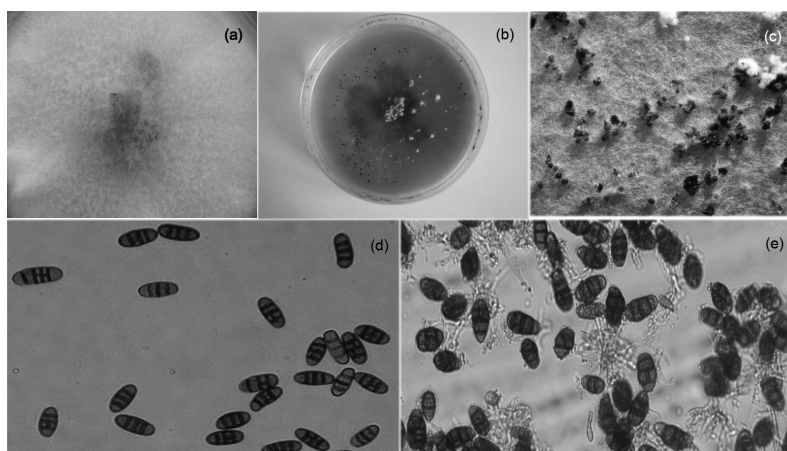


Fig. 1a. Mycelium of *Pinus ponderosa* var. *jeffreyi*; 1b, c. Pycnidia of the fungus *Camarosporium pini*; 1d. Conidia of *C. pini* on *P. nigra*; 1e. Conidia of *C. pini* on *P. ponderosa* var. *jeffreyi*.

Scale bars: d, e = 50 μm .

Table 1. Morphological parameters of the fungus *Camatosporium pini*: comparison of our results with results of other authors

Author	GROVE, 1922	DENNIS, 1964	ELLIS and ELLIS, 1997	JAMES, 1984	IVANOVA and BERNADOVIČOVÁ, 2010	IVANOVA, 2015	Our experiment
Hosts	<i>Pinus nigra</i>		<i>Pinus sylvestris</i>	<i>Pinus contorta</i>	<i>Pinus nigra</i>	<i>Pinus nigra</i>	<i>P. ponderosa</i> var. <i>jeffreyi</i>
Conidiomata	180–300 µm diam.	–	–	120–150 µm in diam., ostiolate, globose, black, papillate, singly or grouped together within black stromata	180–300 µm in diam.	180–300 µm in diam., subglobose arranged linearly, brown, ostiole subpapilliform	Pycnidia 0.5 mm diam., globose, black, erumpent, separate, ostiolate, papillate
Conidiophores	Short and indistinct	Indistinct	–	Short hyaline	Short	Short and indistinct	Short, simple
Conidia size (µm)	15–18 × 7–8	15–25 × 7–9	15–18 × 6–8	15.5–22.0 × 5.5–6.8	20–22 × 6–8	18–20 × 9–10	20 (–22) 28–30 × 10 (–12) 15
Conidia	Red-brown, oval oblong, rounded at both ends, often slightly curved, the central cells often shorter than the terminal ones	Elongate-oval, brown, muriform, non-constricted	Brown	Fusoid-ellipsoid, four-celled, reddish-brown, slightly constricted at each septum	Pale brown, thin-walled, smooth fusiform to fusiform elliptical, straight, apex subobtuse, base truncate	Oval, brown, rounded at both ends, often slightly curved	Oblong, larger, dark, golden brown at maturity, ovoid to ellipsoid or elongate to fusoid, several-celled, non-constricted, narrowly rounded at both ends
Septa transverse	3	3–6 or even 8	3	3	3(4)	3	2–3(4)
Septa longitudinal	1–2	–	1–2	–	1–2	1–2	1–2
Culture characters	–	–	–	Growth very slow, 0.6 mm/day over the agar surface, 13 days at 25°C	Colonies on PDA slow growing, 2.5–3 cm after 1 week at 24°C	–	Colonies on PDA slow growing, 3 cm diam. after 1 week at 24°C
Mycelium	–	–	–	Olivaceous-brown with white margin	Initially white mycelium gradually grey to dark grey	–	White at first, grey and olive-green at maturity
Hyphae	–	–	–	Olivaceous, distinct barrel-shaped cells	–	–	Septate, hyaline, thin

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