## *Phoma fuliginosa* sp. nov., from *Caloplaca trachyphylla* in Nebraska, with a key to the known lichenicolous species

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**Abstract:** Phoma fuliginosa sp. nov. is described and illustrated from apothecia of Caloplaca trachyphylla (Teloschistaceae) collected in Nebraska, USA. The species is distinguished from other lichenicolous fungi referred to the genus in the shape and size of the conidia. It is considered pragmatic to continue to use the generic name Phoma for this group of lichenicolous coelomycetes until cultural and molecular information is available, although these fungi may later prove not to be congeneric amongst themselves nor with the species occurring on plants. A key to the 14 lichenicolous species now accepted in Phoma is provided, and notes on reports of some additional collections referred to the genus are included.

Key words: Coelomycetes, conidial fungi, generic concepts, lichens, mitosporic fungi, *Teloschistaceae*.

## Introduction

The generic name Phoma Sacc. 1880 is a conserved name with P. herbarum Westend. 1852 as the conserved type species (Greuter et al. 2000). Phoma herbarum is a frequently encountered saprobe of dead stems of herbaceous plants, and is particularly commonly encountered on Urtica in Europe; it has an extensive list of synonyms (Boerema 1963, 1970). The taxonomy of the genus has been increasingly based on the comparison of cultures grown under standard conditions of growth, temperature, and light regimes rather than specimens on plant hosts. Around 200 'true Phoma taxa' are currently recognized (Aa et al. 1990), and critical revisions of many groups of those occurring on plants have been prepared during the last decade. Where molecular work has been carried out, there has been a high degree of support for species and varietal concepts based on cultural studies (Abeln *et al.* 2002). The genus has been divided into nine sections and a detailed overview and key to these based on characteristics as seen in culture has been produced (Boerema 1997). Many species do not have known teleomorphs, but most sections have some species with teleomorphs in *Didymella*; in other cases the teleomorphs belong to other loculoascomycete genera, notably *Leptosphaeria, Mycosphaerella*, or *Pleospora* (Boerema 1997).

The generic name *Phoma* has been used for lichenicolous fungi since the late nineteenth century. Twenty-six *Phoma* species had been described on lichens up to 1980, of which five were accepted by Hawksworth (1981). Since that time, eight additional lichenicolous species have been described. All share discrete pycnidial thinwalled conidiomata, often dark brown around a well-formed ostiole, subglobose to

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short-ampulliform to doliiform conidiogenous cells, enteroblastic conidiogenesis, and conidia which are simple, colourless, subglobose to ellipsoid, and lack appendages. No teleomorphs are known amongst the lichenicolous species, and no lichenicolous species of *Didymella* were recognized by Grube & Hafellner (1990); all those previously reported belong to other genera. None of the lichenicolous *Phoma* species has ever been grown in pure culture, and neither is any molecular information available on them.

Having discovered another new uncultured lichenicolous Phoma growing on Caloplaca trachyphylla in the USA, we were challenged as to whether additional species should be described in the genus when they could not be subjected to the protocols developed for the study of Phoma species in culture from plants, i.e. when they could not be compared directly with the currently accepted species on plant substrata. However, the main purpose of scientific names is to provide a means of communication, and that the name conveys accurate phylogenetic information is secondary. As the alternative would be not to have names, leaving the species undescribed, we decided to continue with a pragmatic approach and therefore describe the new taxon in Phoma. We also take the opportunity to present a new key to the currently recognized lichenicolous species, in view of the many species recognized since the key of Hawksworth (1981).

## Methods

The specimen was studied macroscopically with a Nikon dissecting microscope fitted with an eyepiece reticule at magnifications up to  $\times$  80. Microscopic characters were studied in water, 10% potassium hydroxide, Lugol's iodine after pre-treatment with K, unstained and stained in lactophenol with both Cotton blue and fuchsin, and erythrosin B-ammonium solution. Both squash preparations and 12 µm thick sections prepared on a Bright Starlet cryostat freezing microtome were used. Slides were examined in an Olympus BH-2 microscope fitted with Nomarski differential interference contrast optics, a drawing tube, and an automated photographic attachment. In citing measurements, average figures are given in italics where

ten or more measurements could be made. Drawings were prepared using the drawing tube at  $\times$  3200.

#### Taxonomy

## Phoma fuliginosa M. S. Cole & D. Hawksw., sp. nov.

Pycnidia lichenicola, immersa, subglobosa, 50–75  $\mu$ m in diametro. Cellulae conidiogenae breviter ampulliformes vel subglobosae, 5–6·5–7·5 × 3–4·5–6  $\mu$ m. Conidia ellipsoidea, 5–5·5–6·5 × 3–3·5  $\mu$ m.

Typus: USA, Nebraska, Scott's Bluff County, Scott's Bluff National Monument, south bluff on point of ridge south of Coyote Pass, on ridgetop with scattered *Pinus ponderosa* and *Juniperus*, 41°49′20″N 103°42′51″W, alt. 1387 m, on *Caloplaca trachyphylla*, 4 July 1997, *C. M. Wetmore* 77657B (MIN—holotypus).

## (Figs 1 & 2)

Conidiomata pycnidial, immersed in the apothecia of the host lichen at first but becoming partially erumpent at maturity, scattered brown to dark brown, 50-75 µm diam.; pycnidial wall  $9.5-11-12.5 \mu m$  thick, composed of 2-3 layers of pseudoparenchymatous cells, mainly polyhedral but some subglobose, outer cells brown and the inner ones hyaline, outer cells  $6-8-11 \times 4.5-6-$ 7  $\mu$ m, inner cells 5–7 × 4–6  $\mu$ m; ostiolate, ostiole c. 10 µm diam., sometimes up to 15 µm diam when old and splitting open, cells surrounding the ostiole dark brown, the cells somewhat elongated and pointed and to 11 µm long. Conidiogenous cells lining the inner wall of the pycnidial cavity, shortampulliform to subglobose, not proliferating, hyaline, smooth-walled,  $5-6\cdot 5-7\cdot 5 \times 3$ conidiogenesis  $4.5-6 \,\mu m;$ enteroblastic. Conidia abundantly produced, arising singly, narrowly ellipsoid, rounded at the ends, hyaline, simple, smooth-walled,  $5-5\cdot 5 6.5 \times 3 \,\mu m.$ 

*Etymology.* From 'fuliginosus' (Latin), sooty; recalling the appearance of the infected host apothecia.

*Host.* In apothecia of *Caloplaca trachyphylla*, the infected discs becoming discoloured brownish. In section, ascospore production appears to be inhibited in

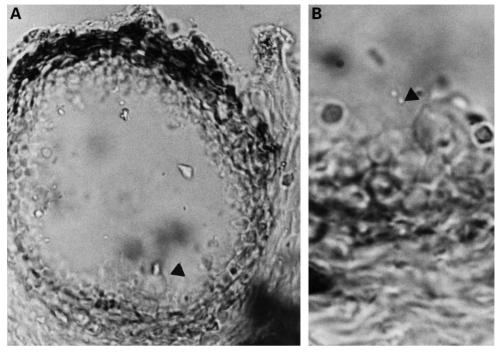


FIG. 1. Phoma fuliginosa (holotype). A, vertical section of pycnidial conidioma, showing the darkly pigmented cells around the ostiole and a conidiogenous cell with attached conidum (arrow), × 800; B, detail of pycnidial wall with a conidiogenous cell and conidium (the point of attachment arrowed), × 2200.

infected parts of the hymenium. However, no extensive bleaching was evident and infections were localized. The species therefore appears to be a mild pathogen.

*Distribution.* USA (Nebraska). Known only from the original collection. The host lichen has a wide distribution in the intermontane area of western North America, occurring in sunny open situations, and is also known from Afghanistan, China, and Greenland (Wetmore & Kärnefelt 1998).

Observations. This new species differs from Phoma caloplacae D. Hawksw. 1981 in the shape of the conidia, which are ellipsoid rather than globose (Hawksworth 1981). Amongst the other lichenicolous species referred to the genus with ellipsoidal conidia, the conidia are broader and(or) longer or shorter than those of the currently known species, from which it can be distinguished using the key presented below.

It would only be possible to compare the species with plurivorous *Phoma* species, known from plants and soil, by obtaining pure cultures or by molecular methods. This possibility must await fresh collections.

## Key to the lichenicolous species of Phoma

1	Conidia broadly to narrowly ellipsoid
	Conidia subglobose
2(1)	Conidiomata mainly less than 50 µm diam
	Conidomata mainly more than 50 $\mu m$ diam $\hfill \ldots \ldots \ldots \ldots \ldots \ldots \ldots \ldots 6$

Vol. 36

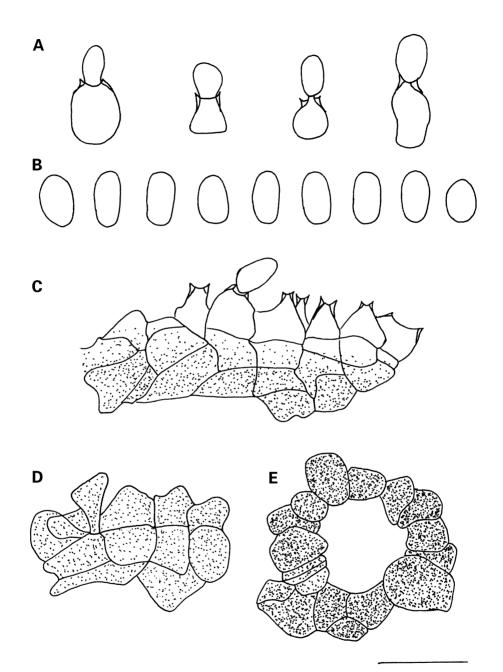


FIG. 2. *Phoma fuliginosa* (holotype). A, conidiogenous cells; B, conidia; C, vertical section of pycnidial wall with conidiogenous cells; D, surface view of pseudoparenchymatous pycnidial wall cells; E, surface view of dark brown pigmented cells around the ostiole. Scale=10 μm.

2004	Phoma fuliginosa—Hawksworth & Cole 11
3(2)	Conidia less than 6 $\mu$ m in length
4(3)	Conidiogenous cells short-ampulliform
5(4)	Conidiomata 15–60 $\mu$ m diam; conidiogenous cells 4–5 × 3–4 $\mu$ m; conidia 3–5 × 1– 1·5 $\mu$ m; on <i>Lecanora expallens</i> ; Luxembourg. See Diederich (1986) <b>P. lecanorina</b> Diederich 1986
	Conidiomata $30-50 \ \mu\text{m}$ diam; conidiogenous cells $4 \cdot 5 - 6 \times 2 \cdot 5 - 3 \cdot 5 \ \mu\text{m}$ ; conidia $3 \cdot 5 - 5 \cdot 5 \times 1 \cdot 5 - 2 \ \mu\text{m}$ ; on <i>Diploschistes ocellatus</i> ; Spain. See Calatayud & Etayo (2001)
6(2)	Conidia ellipsoid, usually more than $2.5 \mu\text{m}$ wide $\dots \dots \dots$
7(6)	Conidiomata exceeding 80 $\mu$ m diam
8(7)	Conidia not exceeding 7 $\mu$ m in length
9(8)	Conidia $4-5\cdot5(-6) \times 2\cdot5-3\cdot5(-4) \mu m$ (length:breadth ratio $1\cdot5-1\cdot7$ ); conidiomata 100–150 $\mu m$ diam; conidiogenous cells 4–7 $\mu m$ diam; on <i>Physcia aipolia</i> and <i>Physconia distorta</i> ; Austria, Canada, Sweden, UK, and USA. See Hawksworth (1981), and Keissler (1911) <b>P. physciicola</b> Keissl. 1911 Conidia $(4-)6-7 \times (2-)3 \mu m$ (length:breadth ratio $2-2\cdot3$ ); conidiomata 80–150 $\mu m$ diam; on <i>Phaeophyscia orbicularis</i> , <i>P. sciastra</i> , and also reported from <i>Xanthoria parietina</i> ; France, Greenland, and Sweden. See Alstrup & Hawksworth (1990) and Vouaux (1914) <b>P. epiphyscia</b> Vouaux 1914
10(6)	Conidia 2–2·5 µm wide
11(10)	Conidiogenous cells 4–6 μm diam; conidia (4–)4·5–6(–7) × 2–2·5(–3) μm; conidiomata (75–)100–150(–200) μm diam; on <i>Peltigera</i> spp.; Austria, Finland, France, Germany, Luxembourg, Norway, and Sweden. See Hawksworth (1981)
	Conidiogenous cells $8-10 \times 1.5-2 \mu m$ ; conidia $5-6(-7) \times 2-2.5 \mu m$ ; conidiomata 120–200 $\mu m$ diam; on <i>Dibaeis baeomyces</i> ; Norway. See Ihlen (1998) <b>P. maculiformans</b> Ihlen 1998
12(10)	Conidia 5–7 × 1·5–2 μm; on a wide range of parmelioid lichens ( <i>Flavoparmelia</i> , <i>Hypotrachyna</i> , <i>Melanelia</i> , <i>Menegazzia</i> , <i>Parmelia</i> , <i>Parmeliopsis</i> , <i>Parmotrema</i> , and <i>Rimelia</i> spp.) and also <i>Ramalina calicaris</i> ; Europe (many countries) and North America. See Hawksworth & Punithalingam (1973), Hawksworth (1981)

# 13(1). Conidia (4–)5–6(–7) μm diam; on *Caloplaca cerina*; Russia. See Hawksworth (1981) Conidia 3–4 × 2·5–3 μm diam; on *Lobaria pulmonaria*; Canada, France, and the UK. See Etayo & Diederich (1995) P. lobariae Diederich & Etayo 1995

In addition to the species keyed out here, and species excluded in Hawksworth (1981), three additional reports of lichenicolous *Phoma* species merit comment.

Hafellner & Wieser (2000) mention *Phoma caperatae* Vouaux 1912 on *Flavoparmelia caperata* from Austria but do not provide any descriptive details. The type material of this taxon was destroyed, and no authentic material has been located (Hawksworth, 1981). According to the original description, the conidia should be  $3-6 \times 2.5-3 \,\mu\text{m}$  and with narrowed bases, something not to be expected in *Phoma*. The Austrian material merits more detailed investigation.

Martínez & Hafellner (1998) and Martínez (1999) report "Phoma epiparmelia Vouaux s. ampl." from two collections of Peltigera horizontalis from Spain. That fungus, the type material of which is also lost (Hawksworth 1981), was described on P. pusilla from Croatia and had elongate conidiogenous cells  $6-10 \times 1.5-2 \,\mu\text{m}$  and conidia  $3.5-4.5 \times 2.3 \,\mu\text{m}$ . The "s. ampl." was used because the Spanish material had slightly larger conidiogenous cells  $(9-12 \times 2 2.5 \,\mu\text{m}$ ) and conidia ( $6-8 \times 2.5-3 \,\mu\text{m}$ ). It seems possible that these collections belong to the same species, and that one of the Spanish specimens could provide an appropriate neotype, but the 'sinuous' conidiogenous cells immediately exclude Phoma as a candidate genus. The position of this taxon clearly requires further study.

An unnamed "*Phoma* sp." forming necrotic black-ringed patches on *Lobaria pulmonaria* in Spain is described by Martínez & Hafellner (1998). It was said to differ from *P. lobariae* in the larger conidia  $(5\cdot5-7\times3\,\mu\text{m})$  and the structure of the conidiomata, and from *P. lobariicola* in the dimensions of the conidiomata  $(122-160\times$  88–141  $\mu$ m), conidogenous cells (5 × 7  $\mu$ m), and conidia, and further in causing different symptoms. The authors preferred to await additional material before formally describing this fungus, but it appears to represent a further lichenicolous species of the genus from the information supplied.

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