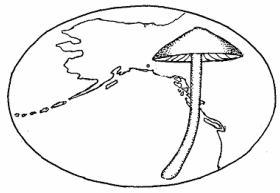
Pacific Northwest Fungi



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Phyllactinia guttata is a Host for *Cladosporium uredinicola* in Washington State

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Abstract: The powdery mildew fungus *Phyllactinia guttata*, parasitic on European hazelnut (*Corylus avellana*), is a host for the fungicolous hyphomycete *Cladosporium uredinicola* in Washington State. Mucilaginous penicillate cells at the apices of the *Phyllactinia* ascocarps are the primary site for colonization and sporulation by *C. uredinicola*. Range of morphological variation in *C. uredinicola* from *P. guttata* was congruent with previous descriptions from hosts in Uredinales.

Key words: *Cladosporium uredinicola*, Erysiphales, fungicolous, mycoparasite, *Phyllactinia guttata*, powdery mildew.

Introduction: *Phyllactinia guttata* (Wallr. : Fr.) Lév. is a powdery mildew (Erysiphales) with a broad host range including many trees and shrubs as well as a smaller number of records from composites and other plants (Braun 1987, 1995; Farr et al. n.d.). Many records are from *Corylus* species (filbert, hazelnut). In the Pacific Northwest, *P. guttata* has been previously recorded from *Corylus avellana* L. (European hazelnut) in British Columbia and Oregon, *C. cornuta* Marsh. (beaked

hazelnut) and *C. californica* (*C. cornuta* var. *californica*) in British Columbia, Oregon and Washington, and *C. maxima* Mill. (giant hazelnut) in Oregon and Washington (Conners 1967; Farr et al. n.d.; Ginns 1986; Glawe n.d.; Shaw 1973). A key to *Phyllactinia* species in North America, synonyms of *P. guttata*, and miscellaneous comments on *Phyllactinia* in North America are found in Braun (1985).

Phyllactinia guttata was identified using the key of Braun (1985) on leaves of *C. avellana* collected 16 September and 2 December 2005 on the campus of University of Washington, Seattle Washington. Examination of both sets of specimens revealed colonization of a high proportion of chasmothecia (also termed ascocarps, perithecia or cleistothecia) by one or more species of *Cladosporium*. We resolved to recover representative *Cladosporium* isolates into culture and identify them to species.

Materials and Methods: Leaves of the 16 September collection were examined at 50X to locate chasmothecia bearing sporulating tufts of *Cladosporium* conidiophores. Sixteen chasmothecia were removed from leaves by dipping the apex of an insect pin in melted glycerin jelly (Dhingra and Sinclair 1985) then gently touching the needle apex to an individual chasmothecium. Individual chasmothecia were transferred to water agar, rolled along the surface or propelled just under the surface of the agar for approximately four to five centimeters to clean adherent spores and debris from the surface, then transferred to Difco malt extract agar (MEA) amended with 50 µg/ml streptomycin sulfate and 50 µg/ml tetracycline hydrochloride (Sigma, St. Louis, Missouri) and incubated at ambient laboratory conditions (ca. 25 ºC). Single-conidium colonies were produced by streaking conidial suspensions of mass isolates onto MEA, and single colonies were transferred to MEA for analysis. The process was repeated for an additional 16 ascocarps from the 2 December collection. All recovered single-colony isolates were incubated at ca. 25 ºC under periodic (12 hr / 12 hr) near ultraviolet and fluorescent lights for ca. 10 days. Species names were assigned by use of keys in Ho et al. (1999) and Heuchert et al. (2005)

Colonized chasmothecia, and conidiophores and conidia from representative single-conidium *Cladosporium* isolates, were photographed at 100-1000X with an Olympus DP-11 camera mounted on a BH-2 Olympus microscope.

Portions of the 2 December collection were deposited at WSP (no. 71183) and HAL. Single-conidium isolates designated as CladexPhyl-1 through CladexPhyl-14 and CladexPhyl-a through CladexPhyl-m from the collections of 16 September and 2 December, respectively, are stored at minus 80 ºC at USDA-ARS Western Regional Plant Introduction Station.

Results: Of 16 chasmothecia transferred to MEA from the 16 September collection, 13 produced only *Cladosporium* colonies, 1 produced both *Cladosporium* and *Aureobasidium*, and two produced no fungal growth on agar. Of 16 chasmothecia transferred to MEA from the 2 December collection, 3 were discarded because the culture plate was contaminated with *Penicillium*. The other 13 chasmothecia produced *Cladosporium* colonies. One *Cladosporium* isolate from the 16 September collection keyed to *C. herbarum* (Pers. : Fr.) Link in Ho et al. (1999). The other *Cladosporium* isolates keyed to *C. uredinicola* Speg. in Heuchert et al. (2005). U. Braun confirmed the identification by examination of cultures and herbarium material deposited with HAL.

Microscopic examination revealed that growth of *Cladosporium* spp. was concentrated in or on the mucilaginous penicillate cells at the apex of a chasmothecium (Fig. 1). Penetration of penicillate cells could be observed (Fig. 2), with most conidiogenesis occurring on or

near the chasmothecial apex (Fig. 3). Variation in dimensions of ramoconidia of different isolates was documented photographically (Figs. 4-6).

Discussion: Both fungicolous and saprobic *Cladosporium* species have been recovered from multiple fungal substrata (Heuchert et al. 2005), including powdery mildews (Heuchert et al. 2005, Kiss 2003). Kiss (2003) lists *C. oxysporum* Berk. & M.A. Curtis and *C. spongiosum* Berk. & M.A. Curtis as occurring on *Phyllactinia corylea* (Pers.) Karst., a synonym of *P. guttata* (Braun 1985). Heuchert et al. (2005) list *Phyllactinia angulata* (Wallr. : Fr.) Lév. and an *Erysiphe* species as powdery mildew hosts for *C. uredinicola*. Most records for *C. uredinicola* indicate rust species (Uredinales) as hosts (Dugan et al. 2004; Heuchert et al. 2005; Morgan-Jones and McKemy 1990). To our knowledge, this is the first report of *P. guttata* as a host for *C. uredinicola*, and the first systematic sampling of chasmothecia of *P. guttata* for colonization by *Cladosporium* species. Hartney et al. (2005) first reported *P. guttata* on *C. avellana* in Washington State.

Morphological variation in *C. uredinicola* is substantial and isolates of *C. uredinicola* have sometimes been assigned to other species (Heuchert et al. 2005). Such variation with substrata and growth conditions was assessed in detail by Morgan-Jones and McKemy (1990), working with isolates from rusts (Uredinales). Variation also was discernable in our isolates (Figs. 4-6), especially with regard to length of ramoconidia. According to Heuchert et al. (2005), ramoconidia sensu stricto are basal within conidial chains, possess a truncate base lacking both a dome and raised periclinal rim, and are rare in *C. uredinicola*. However, long conidia, intercalary in a chain and with two or more apical conidiogenous loci (ramoconidia sensu lato) are common in some isolates of the species (Figs. 4, 5); they are much less frequent or shorter in other isolates. Published illustrations (cited in Dugan et al. 2004, plus those of Heuchert et al. 2005 and Figs. 4-6 here) convey the range of variation in this character.

Kiss (2003) noted that *Cladosporium* species colonize the penicillate cells of *Phyllactinia* species in Hungary and stated that this form of colonization was essentially saprobic. However, Kiss (2003) also noted suppression of maturation and dispersal of *Phyllactinia* ascocarps by colonization with *Cladosporium*, as well as negative impacts on powdery mildew conidia. It remains to be determined whether colonization of the penicillate cell mass, which functions to attach dispersed chasmothecia to new substrata prior to ascospore discharge (Braun 1987), might adversely affect the survival and functioning of chasmothecia in dispersal. *Cladosporium uredinicola* and other *Cladosporium* species recorded from powdery mildews grow well on agar media, but detailed studies of the efficacy of these fungi against powdery mildew are rare. Kiss (2003) summarizes reports of *Cladosporium* species and other fungal antagonists on powdery mildews.

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*Erratum: this reference was inadvertently omitted from the original version of this paper. The correction was made January 30, 2007.

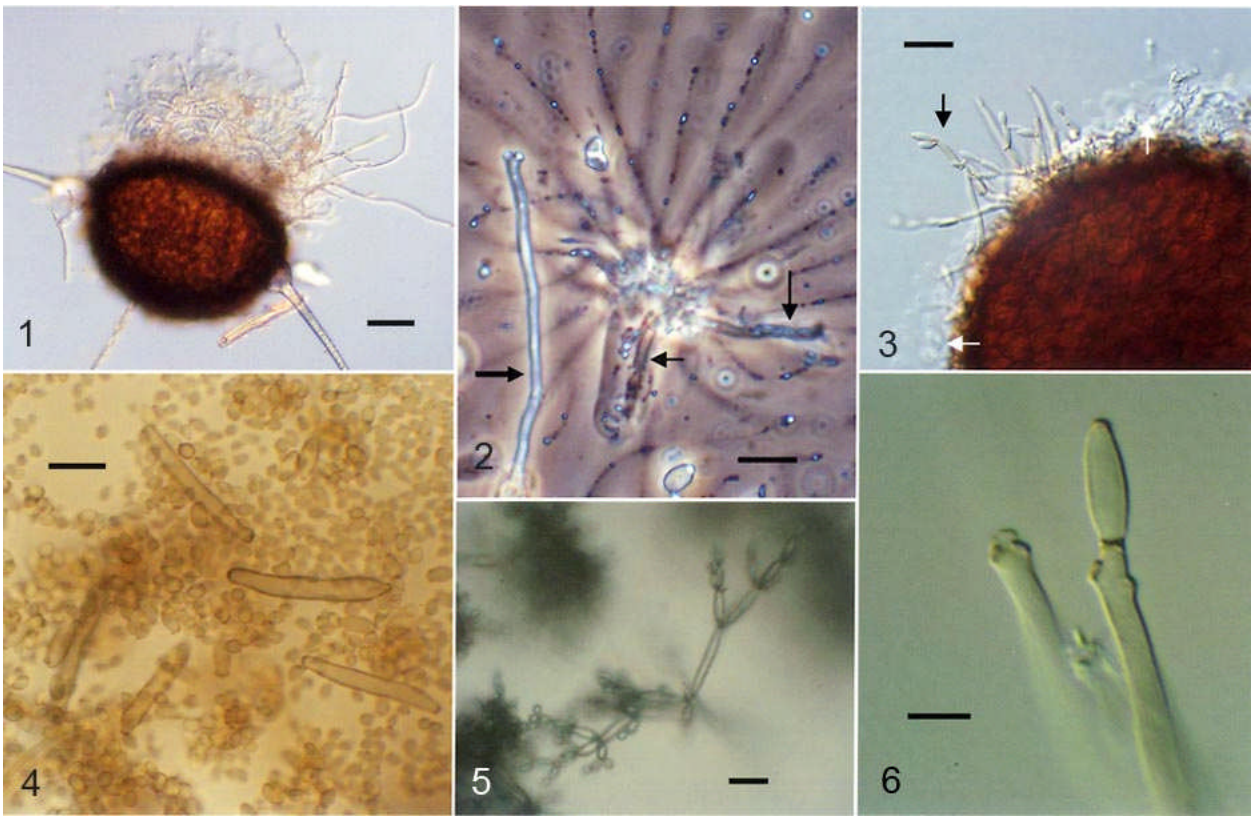

Fig. 1. Hyphae and conidiophores of *C. uredinicola* concentrated amongst translucent penicillate cells at chasmothecium apex. Differential interference contrast. Bar = 50 µm.

Fig. 2. Hypha of *C. uredinicola* internal to base of a penicillate cell (short arrow), and growing amongst mucilaginous appendages (down-pointing arrow). Conidiophore of *C. uredinicola* at left (right-pointing arrow). Phase contrast. Bar = 10 µm.

Fig. 3. Conidiophores and conidia of *C. uredinicola* (black arrow) in proximity to translucent penicillate cells (white arrows). Differential contrast. Bar = 25 µm.

Fig. 4. Conidia and long ramoconidia of *C. uredinicola* seen in lactic acid. Bright field. Bar = 10 µm.

Fig. 5. Conidia and long ramoconidia of *C. uredinicola* in dry mount. Bright field. Bar = 10 µm.

Fig. 6. Apices of *C. uredinicola* conidiophores produced on a chasmothecium; one short ramoconidium still adherent. Differential interference contrast. Bar = 10 µm.