

A New Leaf Disease of *Ilex* spp. Caused by *Cylindrocladium* *avesiculatum* sp. nov.

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ABSTRACT

Cylindrocladium *avesiculatum* sp. nov. causes a severe leaf spotting, defoliation, and twig dieback of *Ilex cornuta*, *I. crenata*, *I. opaca*, and *I. vomitoria*. Small chlorotic spots that became purplish black as they enlarge characterize the disease. Mature lesions are circular to subcircular, up to 15 mm in diam, and frequently zonate, with grey to tan centers and wide, purplish-black margins. In cross-

inoculation tests, *C. avesiculatum* and *C. scoparium* were pathogenic to leaves of *I. cornuta*, *I. crenata*, *I. vomitoria*, *Rhododendron obtusum*, and *Pyracantha coccinea*. The most significant morphological feature of the pathogen is its avesiculate stipe, although on occasion a slight clavate swelling may occur at the apex. *Phytopathology* 61:58-60.

In 1967, a serious and apparently new disease, causing leaf spotting, defoliation, and twig dieback of species of *Ilex*, was discovered in southwest Georgia. Recently, the same disease was found on *I. vomitoria* Ait. 'Nana Compacta' plants at Sanford, Florida. The fungus isolated from diseased leaves and tissues was identified as a species of *Cylindrocladium*. Available literature yielded a single report by Timonin & Self (10) describing a blight and wilt of *I. rotunda* Thunb. caused by *C. scoparium*. The fungus associated with species of *Ilex* in Georgia and Florida, however, was not *C. scoparium*, and appeared to differ from any previously described species of *Cylindrocladium* known to us.

Leaf lesions appear initially as chlorotic spots that become purplish black, surrounded by a light green border. Mature lesions are circular to subcircular, up to 15 mm in diam, often zonate, with wide purplish-black margins and grey to tan centers (Fig. 1). Under very humid conditions the entire leaf may become affected. Leaves abscise readily, frequently with only a single lesion present on the leaf. The fungus sporulates freely on both surfaces of fallen leaves, and frequently on affected foliage before abscission. Twig dieback and death of entire plants may follow heavy infection and subsequent leaf fall.

The fungus has been isolated from container-grown plants of *I. cornuta* Lindl. & Paxt. 'Rotunda' and 'Burfordi', *I. crenata* Thunb. 'Helleri', *I. opaca* Ait. 'Savannah', and *I. vomitoria* 'Nana Compacta'.

In an effort to determine the relationship between the fungus and its suspected hosts, and because of the frequency with which *C. scoparium* has been reported as the cause of diseases of ornamental plants (3, 5, 8, 10), cross-pathogenicity tests using both fungi were initiated. Detailed morphological examination of the fungus was made, and it was compared with descriptions of *C. simplex* and *C. simplex* var. *microchlamydosporum* (6). A preliminary report has been made (4).

MATERIALS AND METHODS.—Inoculum for patho-

genicity tests was prepared by comminuting separately, in distilled water, 15-day-old cultures of *C. scoparium* Morgan and the *Ilex* fungus growing on V-8 juice agar. The mixtures were filtered through two thicknesses of cheesecloth and sprayed on the leaves of test plants. Control plants were sprayed with water. After inoculum was applied, the plants were placed in plastic bags and maintained in a greenhouse where the temp varied from 29-35 C during the day and 18-24 C at night.

In the first series of pathogenicity tests, two groups (each consisting of 23 *I. crenata* 'Helleri' and six *I. vomitoria* 'Nana Compacta' plants in 3-inch peat pots, 10 *Ligustrum japonicum* Thunb. and 10 *Pyracantha coccinea* Roem. 'Lalandi' plants in trays, and three plants each of *Rhododendron obtusum* (Lindl.) Planch. 'Hinodigiri' and 'Coral Bell', respectively) were inoculated with the *Ilex* fungus and with *C. scoparium*. The second series of tests, using the same two fungi, consisted of inoculating 80 unrooted cuttings of *I. vomitoria* 'Nana Compacta', 100 of *I. crenata* 'Helleri', 40 of *I. cornuta* 'Rotunda', and three plants each of the two varieties of *R. obtusum* in 6-inch pots. Appropriate controls of each plant type were employed in all tests.

The description of the fungus is based on a minimum of 500 observations and/or measurements of structures produced on leaves of *I. cornuta* 'Rotunda', *I. opaca* 'Savannah', and *I. vomitoria* 'Nana Compacta'. Morphological comparisons of the fungus from leaves of species of *Ilex* with *C. simplex* and its variety are based on descriptions of the latter by Meyer (6). Cultures of *C. scoparium* were obtained from leaves of azalea collected at Ft. Myers, Florida, in 1965, and from azalea cuttings collected at Omega, Ga., in 1969.

RESULTS.—*Pathogenicity tests.*—In the first series of pathogenicity tests, many of the leaves had fallen from plants of *I. crenata* and *I. vomitoria* 1 week after inoculation with *C. scoparium* and the *Cylindrocladium* from *Ilex*. At the end of the 2nd week, the plants were almost completely defoliated, and dieback of the smaller branches was apparent. Lesions produced by

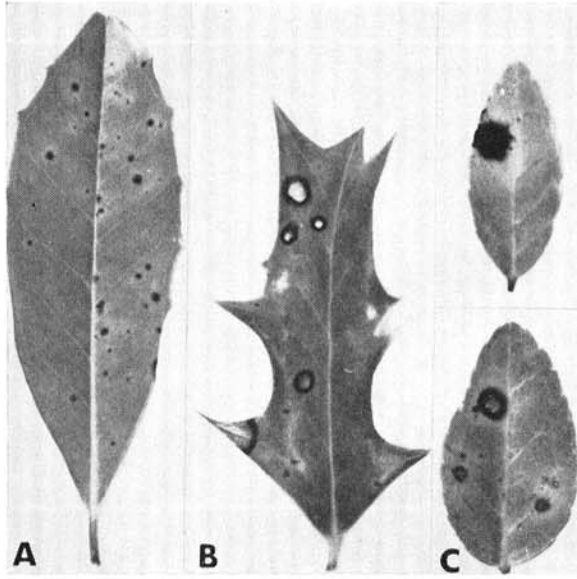


Fig. 1. Lesions caused by *Cylindrocladium avesiculatum* on leaves of A) *Ilex opaca*; B) *I. cornuta*; and C) *I. crenata*.

the two fungi were essentially the same in appearance. Leaves of *P. coccinea* were highly susceptible to *C. scoparium* and slightly susceptible to the *Cylindrocladium* from *Ilex*. Leaves of *L. japonicum* plants were unaffected by the *Ilex* fungus, but were slightly susceptible to *C. scoparium*.

In the second series of pathogenicity tests, cuttings of *I. crenata* and *I. vomitoria* lost most of their leaves during the 1st week after inoculation with *C. scoparium* and the *Ilex* fungus. Plants of *I. cornuta* lost only a few leaves during the same period. By the end of the 2nd week, however, all of the plants, including *I. cornuta*, were almost completely defoliated. Again, both fungi produced lesions that were similar in appearance.

Both fungi produced lesions similar in number and appearance on leaves of the 'Coral Bell' and 'Hinodigiri' varieties of *R. obtusum* in the two tests. Control plants remained unaffected throughout each experiment. Both fungi were reisolated from a representative number of plants.

The pathogen.—Conidiophores arise laterally from a stipe that tapers to an avesiculate, subacute tip (Fig. 2-A, B); or, rarely, terminates in a very slight clavate swelling $12.2-20.4 \times 5.3-7.6 \mu$ (Fig. 2-C). The stipes arise at right angles from the surface of the host or from procumbent mycelia in culture; they are septate, $200-340 \times 3.0-6.5 \mu$, and hyaline throughout, although with age the portion below the conidiophore branches is frequently pale olivaceous. Short lateral stipes often develop from primary, or occasionally from secondary, branches in a manner similar to that described for *C. crotalariae* (2) and *C. floridanum* (9). Conidiophore branching is mostly dichotomous (Fig. 2-A). Primary branches are mostly nonseptate, hyaline, and $17-44 \mu$ in length. Secondary branches and tertiary branches

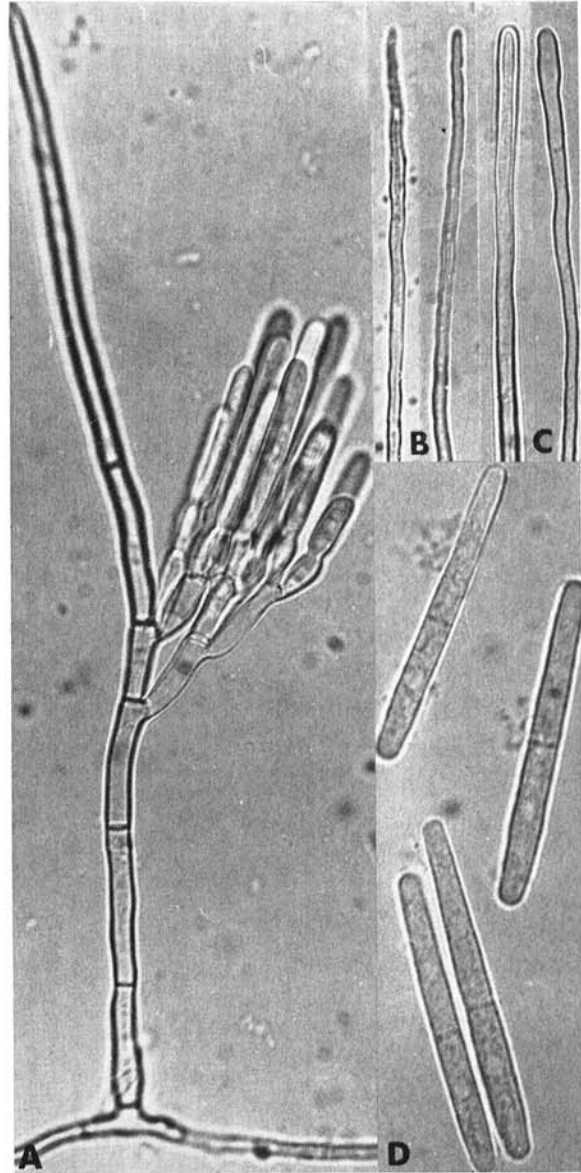


Fig. 2. *Cylindrocladium avesiculatum*: A) conidiophore; B) avesiculate stipe; C) vesiculate stipe; and D) conidia; for size comparison, conidia top left to right 62.6 and 66.3μ , respectively.

are hyaline, nonseptate, and $12-24 \mu$ and $10-19 \mu$ in length, respectively. Two to three or occasionally four hyaline, nonseptate, reniform to doliform phialides, $9-15 \times 4.5-5.9 \mu$, develop at the distal end of the secondary or tertiary branches (Fig. 2-A).

Conidia are hyaline, granular, cylindrical, straight, slightly wider at the base than the apex, rounded at both ends, one-septate, and $51-78 \times 3.5-4.7 \mu$ (Fig. 2-D).

Comparison with other species of *Cylindrocladium*.—There are two references in available literature to species of *Cylindrocladium* without vesicles. Meyer (6) describes the stipe of *C. simplex* and *C. simplex*

var. *microchlamydosporum* as terminating in a point or slight swelling. But the conidia of this species and its variety are $6-9 \times 1.5-2.5 \mu$, as compared with $51-78 \times 3.5-4.7 \mu$ for the *Cylindrocladium* from leaves of *Ilex*.

The *Cylindrocladium* from leaves of *Ilex* spp. is easily distinguished from other species of *Cylindrocladium* by its avesciculate stipe. Sobers & Seymour (9), comparing *C. floridanum* (*Calonectria floridana*) with *C. scoparium*, Bell & Sobers (2), comparing *C. theae* (*Calonectria theae*) with *C. crotalariae* (*Calonectria crotalariae*), and Alfieri et al. (1), comparing *C. ellipticum* with *C. scoparium*, considered the shape of the vesicle to be a primary character by which the species could be distinguished one from the other. Morrison & French (7) consider the vesicle a stable morphological structure, and used it as a basis to distinguish between *C. floridanum* and *C. scoparium*. Since the avesciculate stipe of the *Cylindrocladium* from *Ilex* spp. had been consistent in culture and on its host for 3 years, we propose a new species described as follows:

Cylindrocladium avesciculatum D. Gill, Alf. & Sob., sp. nov.

Conidiophora stipitata a latere oriunda dichotome vel trichotome ramosa; rami plerumque eseptati, primariis $17-44 \mu$ longis, secundariis $12-24 \mu$ longis, tertiariis $10-19 \mu$ longis; phialides reniformes vel doliformes, hyalinae, eseptatae $9-15 \times 4.1-5.9 \mu$; stipites septati, hyalini, $200-340 \times 3.0-6.5 \mu$, plerumque avesciculati vel raro in vesiculis clavatis $12.2-20.4 \times 5.3-7.6 \mu$ terminantes; conidia hyalina, granulata, cylindrata, basibus latioris quam apicibus, utrimque rotundata, uniseptata, $51-78 \times 3.5-4.7 \mu$.

Sejunctum foliis *Ilex vomitoria* Ait. et *Ilex cornuta* Thunb. in Cairo, Ga. In herb. *New York Botanical Gardens* (NY), *The National Fungus Collections* (BPI), et The University of Florida (FLAS). Holotypus: *Ilex cornuta* NY Isotypus: BPI, FLAS.

Cylindrocladium avesciculatum D. Gill, Alf. & Sob.

Conidiophores arise laterally on a stipe, hyaline, dichotomously or trichotomously branched; primary branches mostly nonseptate, $17-44 \mu$ long; secondary branches nonseptate, $12-24 \mu$ long; when present, ter-

tiary branches nonseptate, $10-19 \mu$ long; phialides reniform to doliform, hyaline, nonseptate, $9-15 \times 4.1-5.9 \mu$. Stipes hyaline, septate, $200-340 \times 3.0-6.5 \mu$, mostly avesciculate or rarely terminating in clavate vesicles $12.2-20.4 \times 5.3-7.6 \mu$. Conidia hyaline, granular, cylindrical, straight, slightly wider at the base than the apex, rounded at both ends, one septate, $51-78 \times 3.5-4.7 \mu$.

Isolated from the leaves of *Ilex vomitoria* Ait. and *Ilex cornuta* Thunb. in Cairo, Ga. Deposited in the herbaria of *The New York Botanical Gardens*, *The National Fungus Collections*, and *The University of Florida*.

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