Cylindrocarpon Root Rot of Tulip Poplar

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ABSTRACT

MacDonald, J. D., and Butler, E. E. 1981. Cylindrocarpon root rot of tulip poplar. Plant Disease 65:154-157.

Tulip poplars in a 6-yr-old landscape planting were severely stunted as a result of a root disease characterized by black, scabby lesions in cortical tissues. A pathogen was identified and described as *Cylindrocarpon liriodendri* MacDonald & Butler sp. nov. In greenhouse inoculations, *C. liriodendri* appeared less virulent than *Cylindrocladium floridanum* to tulip poplar, although symptoms produced by the two pathogens were very similar.

Additional key words: *Liriodendron tulipifera* L., tulip tree, yellow-poplar

Tulip poplar, also known as yellow-poplar, or tulip tree (9) (*Liriodendron tulipifera* L.), is a vigorous, attractive tree, widely planted in California landscapes. In March 1978, many trees in a 6-yr-old landscape planting at Davis, CA, appeared severely stunted compared with other tulip poplars at the same site. The root systems of the stunted trees were covered with black, dry, scabby lesions that completely girdled or rotted off distal portions of some roots.

Although the symptoms on the roots of stunted trees resembled those of Cylindrocladium root rot, a serious disease of tulip poplar seedlings in eastern and southern U.S. nurseries (5,6,10), *Cylindrocladium* spp. were not isolated from diseased roots. Instead, a *Cylindrocarpon* sp. was consistently isolated from the margins of the necrotic root tissue.

Because there are no published reports of a *Cylindrocarpon* sp. attacking tulip poplar, an investigation was initiated to determine whether it was the cause of the root disease and to identify the species.

MATERIALS AND METHODS

Isolation and identification. Twelve trees, selected for their stunted appearance, were dug up and removed from the affected landscape planting to allow examination of their root systems. Tree heights ranged from 1.5 to 3.0 m, with stem diameters (30 cm above the soil line) ranging from 3.5 to 7.5 cm. Trees the same age and considered growing normally exceeded 5 m in height and had trunk diameters exceeding 14 cm.

The root systems of the stunted trees were washed to remove adhering soil and examined for cankered tissues. Marginal tissue from the cankers was surface-disinfested in 0.5% NaOCl for 3–5 min and plated on either potato-dextrose agar (PDA), acid PDA (PDA acidified with 25% lactic acid to pH 4.5), or water agar. Isolation plates were incubated at 24 C for 7–10 days, and after colonies grew out, single-conidium transfers were made to slants of PDA incubated at 24 C.

For taxonomic purposes, colonies were started from single conidia and grown on PDA in plastic petri plates at 22–24 C under fluorescent lights (GE "warm white," 48 $W \cdot m^2$) providing 14-hr light periods. Colonies 7–10 days old were examined for conidiophore characteristics; cultures, 12–16 days old, were examined for chlamydospore characteristics. Colony color was estimated in diffuse daylight using Rayner's charts (13), and dried holotype specimens were prepared by Hawksworth's methods (8). Growth rates were determined by incubating PDA plate cultures in darkness at 21 C and measuring colony diameters daily.

Pathogenicity trials. The pathogenicity of *Cylindrocarpon* sp. was evaluated by inoculating healthy tulip poplars of different ages. For seedling tests, seed was purchased from a commercial supplier and stratified for 3 mo at 12 C before planting in flats of pasteurized UC mix

Fig. 1. Root segments from 6-yr-old tulip poplar trees naturally infected by *Cylindrocarpon liriodendri*. Characteristic black, scabby lesions girdle and sever portions of the root system.

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0191-2917/81/02015404/$03.00/0

(1) in a greenhouse. Approximately 1 mo after emergence, seedlings were transplanted individually to 10-cm diameter plastic pots of UC mix, where they were grown an additional 4 mo before inoculation.

Inoculum of the fungus was prepared by growing colonies in plates of PDA under fluorescent lights for 7–10 days and harvesting conidia in distilled water. Conidial suspensions were adjusted to 10^6 conidia per milliliter.

Ten seedlings were inoculated in each of two ways. By one method, 40 ml of inoculum was added to the surface of the soil around each plant and flushed into the undisturbed soil by adding approximately 300 ml of irrigation water. In a second method, 10 ml of inoculum was poured into each of four gashes that were made in the soil with a pot label and were uniformly spaced around the base of each plant. This assured deep penetration of the inoculum into the soil and provided root wounds for entry of pathogens. Groups of control plants were treated in a similar manner but given only distilled water. After inoculation, all plants were maintained in a greenhouse so that symptom development could be observed.

In a separate experiment, 3-yr-old bare-root trees were obtained from a large wholesale nursery and planted in 19-L cans of UC mix. At the time of planting, 100 ml of conidial inoculum was poured over the surface of the exposed roots of six of the plants, which then were placed in a lath house until examination for root lesion development. One-year-old bare-root seedlings from an eastern U.S. nursery also were used in pathogenicity trials. These plants were potted in 15-cm diameter plastic pots of UC mix and inoculated by the second method described for the seedlings.

RESULTS

Isolation studies. Of the 12 trees removed from the landscape planting, 11 had diseased root systems. The exception was apparently stunted due to circled, girdling roots. In the 11 diseased trees, there appeared to be a positive relationship between the severity of root disease and the degree of stunting. The lesions on the roots appeared to be primarily a cortical decay that caused cracks and ruptures in the epidermis and resulted in black, scabby cankers (Fig. 1). The blackened, necrotic tissues were separated from healthy tissue by a fairly sharp, dark brown margin. In severe infections, cankers appeared to coalesce and encircle roots, causing girdling and severing of distal portions of the root system (Fig. 1).

Isolations from cankered roots on each of the diseased trees consistently yielded a species of *Cylindrocarpon*. This fungus was never isolated from the partially excavated root systems of healthy plants or from portions of diseased roots without cankers.

Pathogenicity trials. The seedlings were evaluated for disease 6 wk after inoculation. At that time, plants that were incoulated by the first method of pouring inoculum around the base had developed girdling crown cankers (Fig. 2B), and some were beginning to drop their foliage and appeared near death. Plants inoculated by the second method had large cankers on the taproot and numerous infections on smaller lateral roots (Fig. 2C). All inoculated plants had similar lesions on their roots, and *Cylindrocarpon* was readily isolated from the cankers.

The 3-yr-old trees inoculated as bare-root plants were evaluated for disease symptoms 3 mo later, after they had fully leafed out in the spring. At that time, the shoot systems showed no external symptoms of disease, and inoculated and control plants appeared equally vigorous. When the root systems were washed and examined, however, each inoculated plant had characteristic black, scabby lesions on the roots. Again, *Cylindrocarpon* was easily reisolated from the margins of the cankered tissue. Roots of control plants in these and the seedling inoculations remained free of canker symptoms (Fig. 2A).

After the 1-yr-old, bare-root seedlings were received from the nursery, six of the 50 plants had small, black lesions on their roots. Isolations from these roots showed infection by *Cylindrocladium floridanum*, a common and serious pathogen of tulip poplar seedlings in eastern and southern nurseries (5,6). The affected plants were discarded, and the others were held in isolation for 4 mo after potting and were periodically checked for further evidence of developing infections. During this period, an additional five plants were culled. The remaining plants then were divided into three groups; one served as a control group and the second was inoculated with *Cylindrocarpon*. The third group was inoculated with *C. floridanum*, which was originally obtained from the bare-root plants to provide a direct comparison of *Cylindrocarpon* and *C. floridanum* on tulip poplar seedlings. Inoculum of *C. floridanum* was prepared as described for *Cylindrocarpon*, and the plants were inoculated by the same method.

All plants were harvested and examined 8 wk after inoculation. In this comparison, *C. floridanum* appeared to be more aggressive and virulent on tulip poplar seedlings than did *Cylindrocarpon*. At harvest, some *C. floridanum*-inoculated plants were dead, and all showed severe canker symptoms. The root lesions produced by *Cylindrocarpon* appeared very similar to, although somewhat less severe than, those caused by *C. floridanum*. Despite the similarity of symptoms, the pathogens could readily

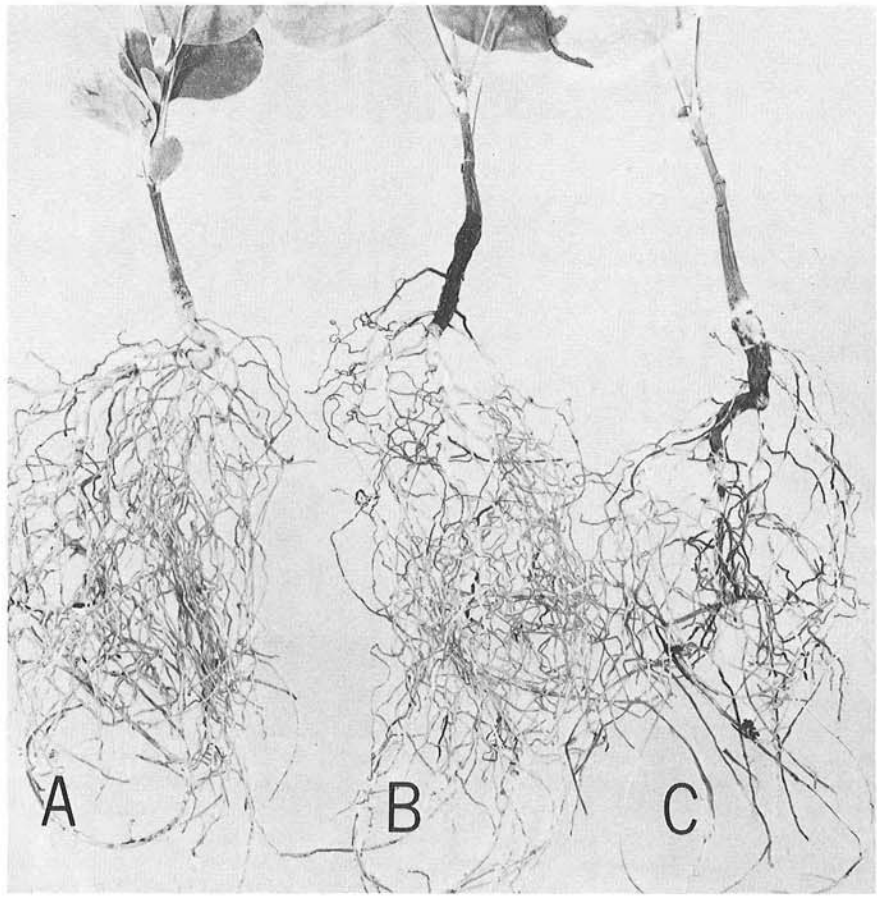

Fig. 2. Tulip poplar seedlings inoculated with *Cylindrocarpon liriodendri*. **(A)** Uninoculated control. Seedling inoculated by method one **(B)** and method two **(C)** described in text.

be distinguished by culturing from the infected tissue.

The pathogen. A review of the literature and study of cultures indicated that the pathogen from tulip poplar was a new species of *Cylindrocarpon* Wollenw.

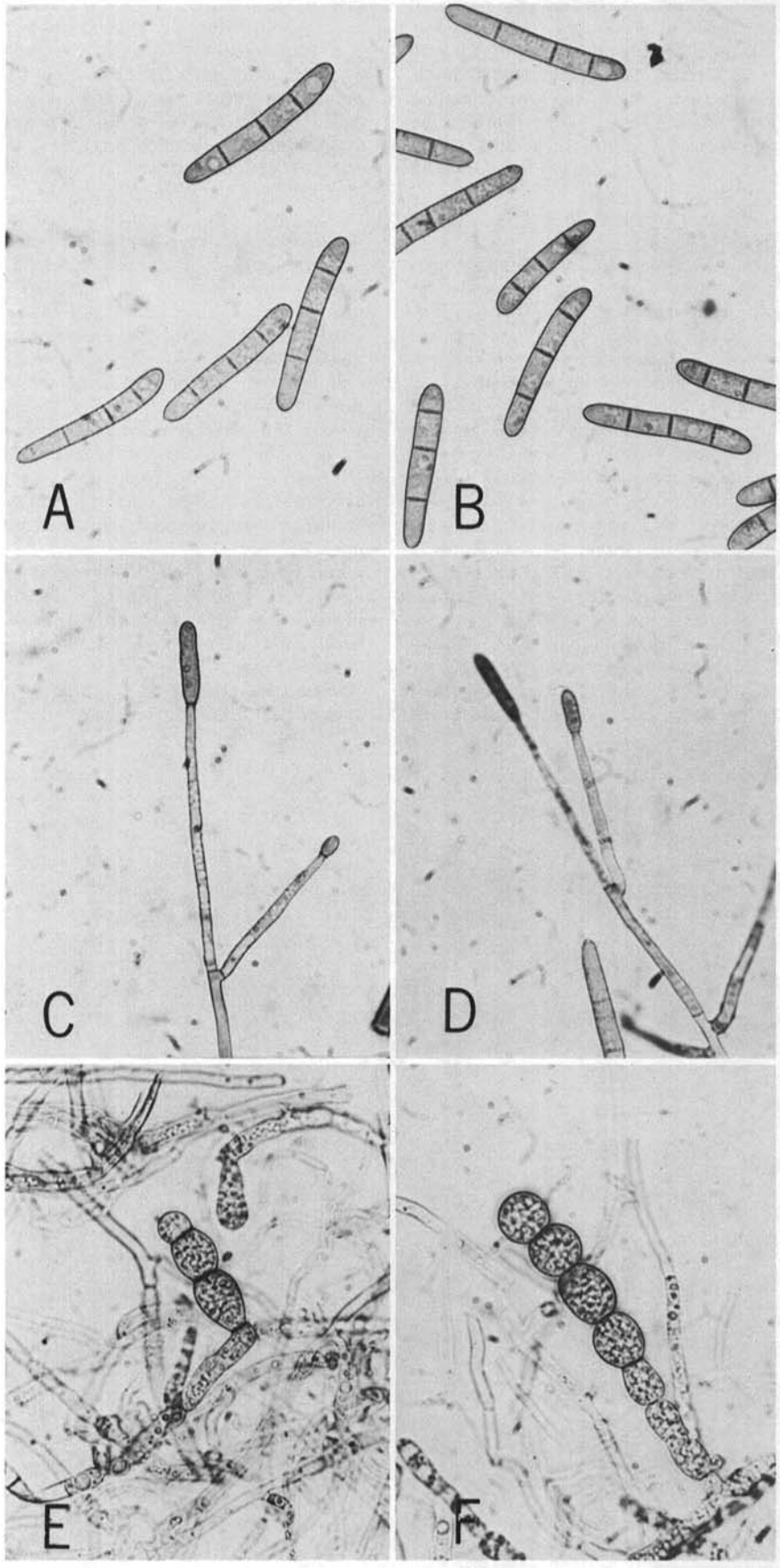

Fig. 3. *Cylindrocarpon liriodendri* MacDonald & Butler sp. nov., magnified × 575. **(A and B)** Macroconidia. **(C and D)** Conidiophores and phialides. **(E and F)** Chlamydospores.

Cylindrocarpon liriodendri MacDonald & Butler sp. nov. (Figs. 3A–F)

Macroconidiophorum parce ramosum, rectum, leve, hyalinum; phialides graciles, 25–38 × 2.5–3.5 μm, attenuatae versus apicem. Macroconidia 0–4, plerumque triseptata, (32–)37–50(–57) × 5–7.5 μm, recta usque ad subcurvata, rotundata in extremis, extremo distali subinflato. Microconidia disunt. Chlaymdosporae intercalares vel terminales, solitariae aut concatenatae trinae usque ad octones, globosae ad ovoideas, 8–12 μm dia, fulvae. Aliquandro intra macroconidia formatae. Telemorphae nullae visae.

Hab. radice morbosa *Liriodendri tulipiferae Holotypus* in herb. Univ. California, Berkeley. No. 1475003.

Macroconidiophore sparsely branched (Fig. 3C and D), straight, smooth, hyaline; phialides slender, 25–38 × 2.5–3.5 μm, tapering toward the apex. Macroconidia 0–4, mostly three septate (Fig. 3A,B), (32–) 37–50 (–57) × 5–7.5 μm, straight to slightly curved, rounded at the ends, distal end slightly enlarged. Microconidia lacking. Chlamydospores intercalary or terminal, single or in chains of three to eight (Fig. 3E and F), globose to ovoid, 8–12 μm dia., golden brown; sometimes formed in macroconidia. Colonies white to pale buff (13), becoming buff to honey with age; reverse pale honey becoming cinnamon to sepia. Colony surface slightly flocculose to powdery, mycelium appressed. Diameter of a colony arising from a single conidium: 45 mm in 7 days at 21 C. Sexual state not observed.

Name: The epithet *liriodendri* refers to the host genus.

Collections; Obtained from diseased roots of *Liriodendron tulipifera* L. in Davis, CA, in March and June 1978. Cultures have been deposited with the ATCC and CBS.

C. liriodendri differs from the similar *C. effusum* Bugn. (3) by its smaller macroconidia, simple branched conidiophore, and slender, tapered phialides. These characters, in addition to the number of septa and the shape of conidia, separate *C. liriodendri* from *C. olidum* (Wollenw.) Wollenw. (3). *C. liriodendri* differs from *C. orchididearum* Schischk. & Tzanava (14), *C. fraxini* Mamuk, (11), and *C. schischkinae* Mamuk. (11) in shape of the macroconidia and morphology of the conidiophore.

DISCUSSION

Although species of *Cylindrocarpon* cause root diseases on a number of plants (2–4,7,9,15), this is the first report of a species attacking tulip poplar. On older trees, *C. liriodendri* causes degeneration of the root system and stunting; young seedlings can be girdled and killed. The ability of *Cylindrocarpon* spp. to infect roots of other hosts has been associated with the presence of plant-pathogenic nematodes (2,4,15), which could cause injuries that allow fungus attack. However, this is not the case with *C. liriodendri* on tulip poplar. An examination of root pieces from plants naturally infected in the landscape failed to implicate any plant-pathogenic nematodes, and our experiments in pasteurized soil showed that the

pathogen could easily attack uninjured roots.

In our inoculation studies, the cankers that developed on roots of tulip poplar inoculated with *C. liriodendri* appeared identical to those caused by *C. floridanum*. Indeed, the only difference in the root diseases caused by these two pathogens appeared to be in severity; *C. floridanum* was more aggressive on and lethal to the young plants. The only means of accurately distinguishing between these pathogens on tulip poplar is by culturing from infected tissue.

The fact that the plants that were removed from the landscape had been on site for 6 yr, during which they had made very poor growth, suggested they were infected with *C. liriodendri* at or shortly after planting. Although *Cylindrocarpon* spp. are well-known as soil inhabitants (12) and the young trees could have been infected by an already present pathogen, they also could have come from a nursery with latent infections. This disease may occur in tulip poplar nurseries and be unrecognized because the symptoms are similar to those of the well-known Cylindrocladium root rot. Although *C. liriodendri* appears less aggressive than *C. floridanum* in attacking tulip poplar, the removal of landscape plants for poor performance is just as important a consequence of disease as plant death. Both result in economic and aesthetic losses.

ACKNOWLEDGMENTS

We thank R. N. Campbell for help and advice, Carl W. Lawhead for technical assistance, and Peter M. Schaeffer, University of California, Davis, for preparing the Latin diagnosis.

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