

## Basidiomycetes Associated with a Patterned Midsummer Wilt of Bluegrass

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### ABSTRACT

Pennypacker, B. W., Sanders, P. L., and Cole, H., Jr. 1982. Basidiomycetes associated with a patterned midsummer wilt of bluegrass. *Plant Disease* 66:419-420.

Histologic examination of crowns from circular areas of wilting Merion Kentucky bluegrass (*Poa pratensis*) and annual bluegrass (*P. annua*) revealed hyphae with clamp connections in the cortical tissue. The wilting grass plants occurred in patterns similar to those described for *Fusarium* blight of turfgrass. Basidiomycetes may play a role in the development of symptoms resembling *Fusarium* blight in turf.

A common manifestation of midsummer disease in bluegrass (*Poa pratensis* L. and *P. annua* L.) stands is roughly circular areas of dead grass. The circular pattern of dead grass, with or without a center tuft of living grass, closely resembles the symptoms of *Fusarium* blight described by Sanders et al (7). Death of affected grass is usually rapid and may be preceded by a short period of wilting. Such circular infection centers are often diagnosed as outbreaks of *Fusarium* (2).

The lack of consensus in the literature concerning *Fusarium* blight (8) and the results of field experiments with the fungicide triadimefon for control of *Fusarium* blight have raised questions about the accuracy of a common diagnosis for all investigated outbreaks. Triadimefon completely controls outbreaks of symptoms resembling *Fusarium* blight on Merion Kentucky bluegrass and annual bluegrass (1,9), although in vitro tests with this fungicide indicate that it has little activity against the cultivars of *F. roseum* associated with the symptom (7). Triadimefon does, however, have activity against many basidiomycetes (7), and Filer (3) has shown that the basidiomycete *Marasmius oreades* (Bolt. ex Fr.) Fr. is capable of infecting grass roots. It is possible that the literature on *Fusarium* blight reflects the investigation

of more than one disease syndrome, with similar hosts, infection center morphology, and colonization of dead or wilted grass by *Fusarium* spp.

Most research aimed at identifying the fungi associated with symptoms resembling *Fusarium* blight has involved grass plants in infection centers after collapse and death of affected plants. The present histologic study was conducted on bluegrass plants in circular wilting areas, 12–20 cm in diameter, in an effort to determine the fungi that were associated with such plants at the onset of visible symptoms of the disease.

### MATERIALS AND METHODS

Plugs (10 cm diameter) of Merion Kentucky bluegrass (*P. pratensis*) showing symptoms of wilt were taken at the Joseph Valentine Turfgrass Research Center at Pennsylvania State University. Similar plugs of wilting annual bluegrass (*P. annua*) were obtained at the university golf course. The sampling was done during an extended period of hot (35 C), dry weather in July 1980. The soil from each plug was examined macroscopically and microscopically for the presence of mycelium. Samples of crowns from symptomless grass plants were collected from New York and Pennsylvania over several summers during outbreaks of symptoms resembling *Fusarium* blight.

Histologic samples consisting of crown areas, including crown and proximal root and stem tissues from individual plants, were fixed in a mixture of formalin, acetic acid, and alcohol (5). Fixed specimens were dehydrated, using a tertiary butyl alcohol schedule, and embedded in paraplast (5). Longitudinal sections were cut at 10  $\mu$ m on a rotary microtome, mounted on chemically clean slides with Haupt's adhesive, and stained with Johansen's quadruple stain (5).

### RESULTS

Macroscopic examination of the soil in the plugs of wilting grass revealed white mycelial strands ramifying through the soil in 35% (19/54) of the samples examined. Clamp connections were found in some mycelial strands under microscopic examination.

Histologic examination of the wilting crown areas revealed abundant mycelium external to the epidermis. The observed hyphae were of two diameters and had numerous clamp connections. Hyphae were also present in the cortical cells of the crown and lower stem (Fig. 1). The hyphae were intracellular and generally restricted to the cortex. Clamp connections were noted on some of the cortical hyphae (Fig. 2), indicating that a basidiomycete had penetrated and colonized the cortical tissue. In addition to clamp-bearing hyphae, hyphae exhibiting right-angle branching characteristic of *Rhizoctonia* spp. were occasionally found in the cortical cells and external to the epidermis (Fig. 3). Limited hyphal penetration and colonization of the vessel elements and tracheids of the xylem were noted. No sporulation was observed.

The fungal hyphae entered the grass crown through breaks in the epidermis associated with emerging roots and leaves. In several cases, external hyphae were tightly appressed to the epidermis (Fig. 4). Intracellular hyphae penetrated the cell walls via pit pairs and exhibited some hyphal swelling adjacent to the penetrated cell walls.

The major host response in the wilting crown tissue was vascular plugging with a chromophyllic material that stained strongly with safranin. Such plugging was present in varying amounts in all crown tissue examined from wilting plants.

Histologic examination of symptomless bluegrass from New York and Pennsylvania over several summers failed to reveal the presence of clamp-bearing hyphae in crown tissue.

### DISCUSSION

The basidiomycetes observed in and around the bluegrass crowns at wilt exhibited many of the characteristics of ectotrophic pathogens. According to Garrett (4), these pathogens are capable of growing over the root surface and through the soil before infecting a host.

Contribution 1268, Department of Plant Pathology, Pennsylvania Agricultural Experiment Station. Authorized for publication 8 July 1981 as *Journal Series Paper* 6266.

Accepted for publication 30 December 1981.

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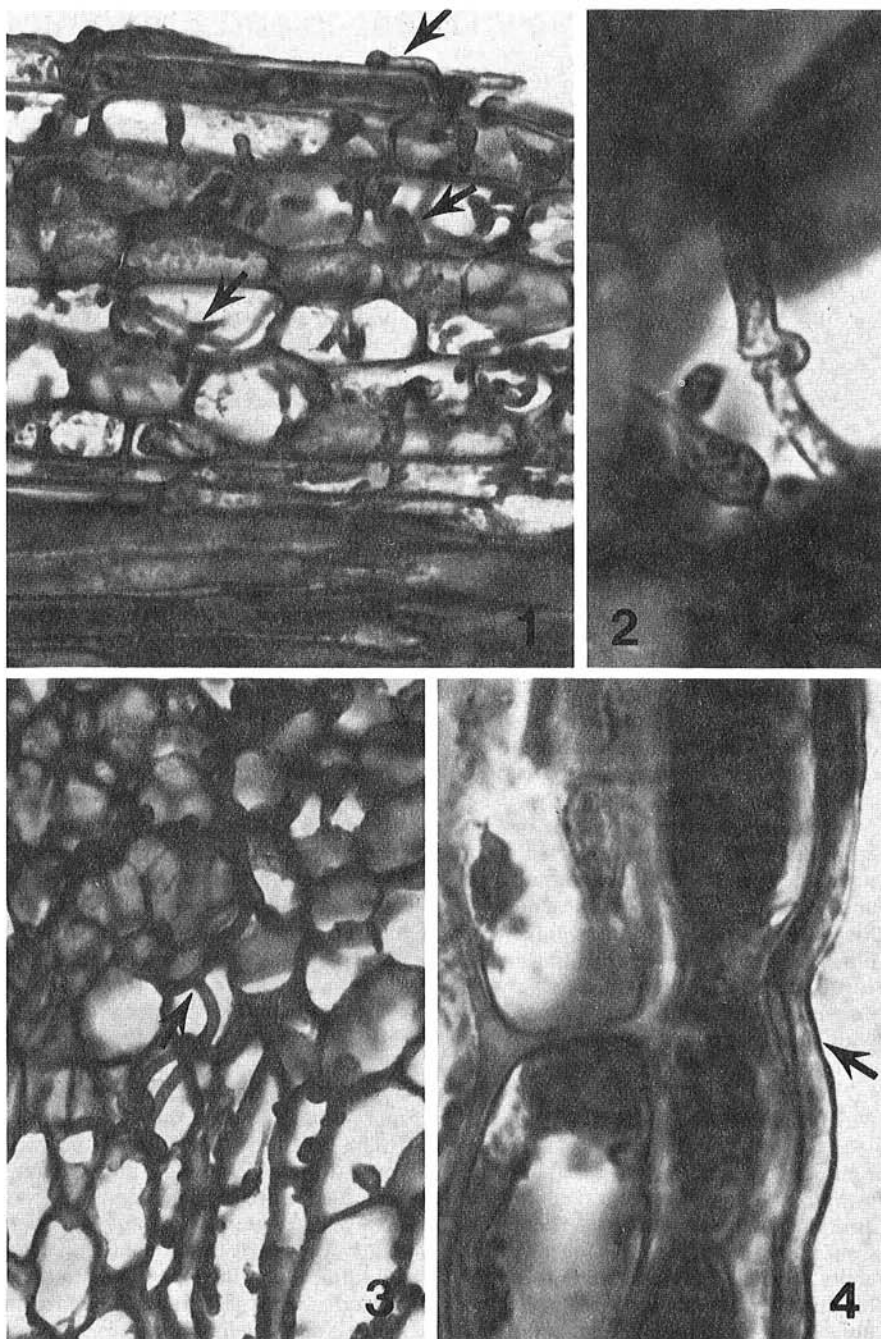


Fig. 1-4. Crown tissue of a wilting bluegrass plant. (1) Longitudinal section showing hyphae (arrows) in the cortical cells and external to the epidermal cells ( $\times 500$ ). (2) Clamp-bearing hypha in a cortical cell ( $\times 1,600$ ). (3) Hypha exhibiting right-angle branching (arrow) present in the cortex ( $\times 500$ ). (4) External hypha (arrow) tightly appressed to the epidermis ( $\times 1,600$ ).

They frequently form mycelial strands and rhizomorphs to facilitate their movement through soil. The basidiomycetes noted in this study were capable of existing outside the host and were also observed tightly appressed to the exterior of the host epidermal cells.

Ectotrophic pathogens are sensitive to changes in the soil environment (4). Such sensitivity could account for the absence of visible signs of basidiomycete colonization of soil and plants in centers of infection resembling Fusarium blight after the collapse and death of affected grass plants. This sensitivity, in addition to slow growth rate or complete inability

to grow on common growth media, probably accounts for the fact that basidiomycetes are rarely isolated from these circular infection centers. Waid (10), in a study of discolored ryegrass (*Lolium perenne* L.) roots, frequently saw clamp-bearing hyphae in cortical tissues, but was able to isolate these fungi on only two occasions.

The histologic evidence presented in this paper associates one or more basidiomycetes with a patterned mid-summer wilt of annual and Kentucky bluegrass. Clamp-bearing hyphae have been reported in both symptomless and discolored grass roots. Warcup (11) has

noted that basidiomycetes are only occasionally present in roots from symptomless pasture grasses, and Waid (10) frequently saw clamp-bearing hyphae in microscopic studies of discolored ryegrass roots. The present study demonstrated the presence of basidiomycetes in crown and lower stem tissue from wilting bluegrass plants.

This study was restricted to grass plants showing initial wilt symptoms, thus we have no evidence that the cortical-infecting basidiomycetes actually caused the ultimate death of the affected plants. In the absence of histologic evidence of destruction of colonized tissue, it is possible that the cortical-infecting basidiomycetes either act as moisture sinks by competing for scarce moisture or produce phytotoxic metabolites, resulting in internal moisture stress and wilt of affected grass plants during dry conditions. The observed vascular plugging may be in response to such a chemical stimulus. After wilting, affected grass plants may be predisposed to infection and colonization by *Fusarium* spp.

Papendick and Cook (6) have reported that *Fusarium* spp. are able to rapidly colonize wheat (*Triticum aestivum* L.) plants under internal moisture stress. Thus, the dead circular areas of bluegrass resembling Fusarium blight may be the result of the action of several pathogens, and the possibility of complexes of *Fusarium* spp. with various basidiomycetes could account for some of the variability in reported research on Fusarium blight.

#### LITERATURE CITED

- Burpee, L. L., Sanders, P. L., Cole, H., and Duich, J. M. 1978. Control of Fusarium blight with fungicides under fairway conditions in mixed Kentucky bluegrass-annual bluegrass, 1977. *Fungic. Nematic. Tests* 33:140.
- Couch, H. B., and Bedford, E. R. 1966. Fusarium blight of turfgrasses. *Phytopathology* 56:781-786.
- Filer, T. H. 1965. Parasitic aspects of a fairy ring fungus, *Marasmius oreades*. *Phytopathology* 55:1132-1134.
- Garrett, S. D. 1970. *Pathogenic Root-Infecting Fungi*. Cambridge University Press, Cambridge, England. 294 pp.
- Johansen, D. H. 1940. *Plant Microtechnique*. McGraw-Hill, New York. 523 pp.
- Papendick, R. I., and Cook, R. J. 1974. Plant water stress and development of Fusarium foot rot in wheat subjected to different cultural practices. *Phytopathology* 64:358-363.
- Sanders, P. L., Burpee, L. L., Cole, H., and Duich, J. M. 1978. Uptake, translocation and efficacy of triadimefon in control of turfgrass pathogens. *Phytopathology* 68:1482-1487.
- Sanders, P. L., and Cole, H., Jr. 1981. The Fusarium diseases of turfgrass. In: *Fusarium: Diseases, Biology and Taxonomy*. P. E. Nelson, T. A. Toussoun, and R. J. Cook, eds. Pennsylvania State University Press, University Park. 457 pp.
- Smiley, R. W., Craven, M. M., O'Knefski, D. C., and Parson, D. 1978. Controlling Fusarium blight of Kentucky bluegrass, 1977. *Fungic. Nematic. Tests* 33:143.
- Waid, J. S. 1957. Distribution of fungi within the decomposing tissues of ryegrass roots. *Trans. Br. Mycol. Soc.* 40:391-406.
- Warcup, J. H. 1959. Studies on Basidiomycetes in soil. *Trans. Br. Mycol. Soc.* 42:45-52.