Infection and Colonization of Bermudagrass by Ophiosphaerella herpotricha, *the Causal Agent of Spring Dead Spot*

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Objectives:

1. To transform *O. herpotricha* to express fluorescent protein genes.
2. Evaluate infection and colonization of bermudagrass cultivars at different temperatures.
3. Evaluate differences in infection and colonization between bermudagrass cultivars that vary in disease susceptibility.

Start Date: 2006
Project Duration: three years
Total Funding: $59,684

Spring dead spot is the most devastating and important disease of bermudagrass that undergoes winter dormancy (Figure 1). The disease is caused by one or more of three fungal species in the genus *Ophiosphaerella* (*O. herpotricha, O. korrae, or O. narmari*). The disease results in unsightly dead patches on fairways, tees, and bermudagrass greens, resulting in the encroachment of weeds and increased management efforts to eliminate weeds and encourage regrowth of bermudagrass into the dead areas.

Despite the identification of the causal agents of the disease in the 1980s, the underlying factors that ultimately lead to death of the plants remain poorly understood. A critical limitation to the study of turfgrass root diseases is the inability of researchers to rapidly and easily study the plant-fungus disease interaction because it happens below ground and often inside of roots. The overall goal of this study is to enhance our understanding of the interaction between *O. herpotricha* and its bermudagrass host and how environmental factors influence this interaction.

Figure 1. *The most devastating and important disease of bermudagrass is spring dead spot.*

Through the insertion of genes into the fungus, transgenic isolates of *O. herpotricha* expressing fluorescent protein genes (visualization genes) have been generated and are currently being used to follow root infection and colonization of various bermudagrass cultivars at different temperatures (conducive and non-conducive) (Figure 2). Root necrosis surrounding fungal hyphae was observed for the susceptible cultivars 'Tifway' and 'Jackpot' 10 days after inoculation. Only minor root discoloration was observed around hyphae of the more resistant 'Midlawn' cultivar.

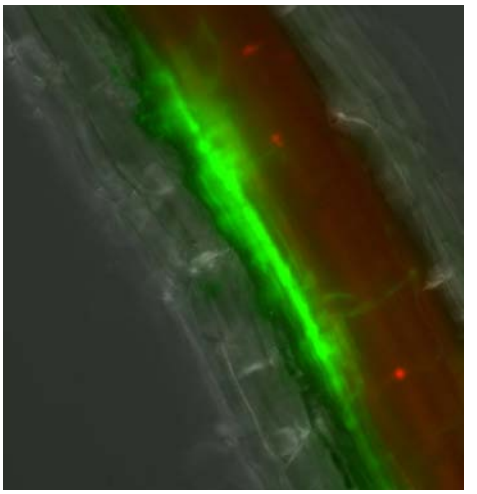

Figure 2. O. herpotricha *expressing fluorescent protein genes (visualization genes) have been generated and are currently being used to follow root infection and colonization of various bermudagrass cultivars at different temperatures.*

Transverse sections revealed extensive internal necrosis and infection of 'Jackpot' and 'Tifway' root cortices (Figure 3). In contrast, infection of 'Midlawn' appeared limited to the outer most cortical cells and these cells did not appear necrotic. No vascular infection by *O. herpotricha* was observed for all cultivars.

Future studies will utilize a confocal scanning laser microscope that can optically 'section' infected roots, producing

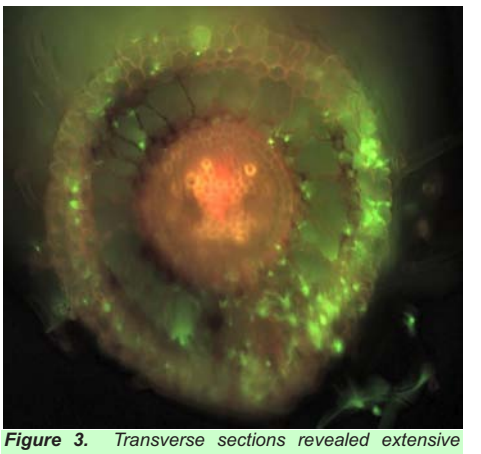

Figure 3. *Transverse sections revealed extensive internal necrosis and infection of 'Jackpot' and 'Tifway' root cortices.*

3-dimentional images of the fungus as it moves in and on bermudagrass roots. We expect to further observe cellular differences in the infection and colonization of bermudagrass cultivars that differ in susceptibility to *O. herpotricha*. This basic information on how the cultivars react to the causal fungus will improve our ability to enhance and deploy host-plant resistance through traditional breeding efforts at Oklahoma State University.

Summary Points

- Fluorescent transgenic fungi have been generated.
- These fluorescent fungi are being used to study the progression of disease in bermudagrass varieties that differ in susceptibility to the disease. Susceptible varieties display more extensive root cortical cell necrosis associated with fungal invasion than that observed in resistant varieties.
- These fluorescent fungi are also being used to study the progression of disease under conducive and non-conducive temperatures regimes.
- This information will be used to enhance host-plant resistance through traditional breeding efforts at Oklahoma State University.