## Identification of *Phoma sclerotioides*, the Causal Agent of Brown Root Rot, in Wisconsin and Minnesota

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Brown root rot (BRR) is a fungal disease, caused by *Phoma sclerotioides*, which is associated with stand decline and reduced yield of forage legumes such as alfalfa, red clover, bird's-foot trefoil, alsike clover, and sweet clover (1). The pathogen was found to be widespread in Alberta's Peace River Valley in the 1980's. More recently the disease has been reported in Wyoming, Montana and Idaho on alfalfa (2). The fungus was identified from diseased alfalfa roots from Shawano and Outagamie counties in Wisconsin in the spring of 2003 (C. Grau, personal communication). To begin to determine the distribution of the fungus in midwestern states, a survey for the pathogen was initiated in the fall of 2003 and continued in spring 2004. Samples of alfalfa plants were requested from university, industry, and extension personnel. During 2003, samples were assayed from Iowa (3 counties), Idaho (1 county), Illinois (4 counties), Minnesota (8 counties), Wisconsin (12 counties), and Wyoming (2 counties). In 2004, samples from 3 additional counties in Wisconsin and 2 counties in Minnesota have been assayed to date. To assay for the presence of the fungus, DNA was extracted from roots of the alfalfa samples and used in PCR reactions with *P. sclerotioides*-specific DNA primers (3). At least five plants were assayed from each location tested.

The survey confirmed the presence of P. sclerotioides in Idaho and Wyoming. This is the first report of *P. sclerotioides* in Natrona County and Goshen County, WY. The survey confirmed the presence of the fungus in Shawano County, WI, although from only one of 19 locations tested. In addition, plants from Columbia, Oconto, Pierce, and St. Croix counties tested positive for the fungus. The survey found the fungus in seven Minnesota counties for the first time. In northwestern Minnesota, plants from Marshall, Pennington, and Red Lake counties were positive by the PCR assay. A majority of the plants tested positive for the fungus from the Pennington and Red Lake County locations. In addition, the fungus was isolated from plants from these counties and from plants obtained from Otter Tail County. PCR positive plants were also obtained from southern locations including Sherburne, Wabasha, Washington, and Winona counties. No positive plants were found in samples submitted from Iowa or Illinois. Plants positive for the fungus originated from both younger (2-year-old) and older (3- to 11-year-old) stands. From plants assayed in fall 2003, the presence of a lesion on the root was not strongly associated with a positive PCR result. Roots with and without lesions were PCR positive. However, this research indicates that plants lacking symptoms can harbor the fungus. The fungus was identified from plants with rotted roots in spring 2004 as well as from plants with healthy roots. PCR reactions with representative DNA samples and primers to the ribosomal DNA internal transcribed spacers indicates that the false negative rate for the test is very low. However, sensitivity of the test may be improved by use of realtime PCR.

## References

1. Davidson, J. G. N. 1990. Brown root rot. In: Compendium of alfalfa diseases, 2nd Edition, Stuteville, D. L. and D. C. Erwin, editors. APS Press, St. Paul, MN.

2. Hollingsworth, C. R., Gray, F. A., Koch, D. W., Groose, R. W. and Heald, T. E. 2003. Distribution of Phoma sclerotioides and incidence of brown root rot of alfalfa in Wyoming, USA. Can. J. Plant Pathol. 25:215-217.

3. Larsen, R. C., Hollingsworth, C. R., Vandemark, G. J., Gritsenko, M. A. and Gray, F. A. 2002. A rapid method using PCR-based SCAR markers for the detection and identification of Phoma sclerotioides: The cause of brown root rot disease of alfalfa. Plant Dis. 86:928-932.