

***Pythium* and *Fusarium* species causing seed rot and damping-off of alfalfa**

Laurine E. Berg¹, Susan S. Miller², Melinda R. Dornbusch², Lorien Radmer,¹ and Deborah A. Samac^{1,2}

¹ Department of Plant Pathology, University of Minnesota, Saint Paul, MN

² USDA-ARS-Plant Science Research Unit, Saint Paul, MN

Seed rot and damping-off is an important disease of alfalfa, severely affecting stand establishment when conditions favor the disease and reducing vigor of adult plants. Globally, 15 *Pythium* species have been found to cause damping-off and seed rot of alfalfa, although surveys of species causing disease on alfalfa in the Midwestern U.S. are lacking. A survey for soil-borne seedling pathogens identified soils from commercial production fields in Minnesota with high levels of damping-off. Pathogens were isolated by a seedling baiting technique from soil of five alfalfa fields. Of the 149 organisms isolated, 93 (62%) were identified as *Pythium* species and 43 (29%) were identified as *Fusarium* species by a combination of morphological characteristics and DNA sequences. *Pythium* species were identified using the ITS and mitochondrial cytochrome oxidase *c* subunit I gene sequences and *Fusarium* species were identified using the ITS and elongation factor 1- α gene. Three species, *P. sylvaticum*, *P. irregulare*, and *P. ultimum* var. *ultimum*, were pathogenic on germinating alfalfa seedlings at 21 °C using a standard agar plate test. Assays with soil infested with individual strains confirmed that these strains cause pre- and post-emergence damping off. Strains of seven species isolated from infected soybean, *P. irregulare*, *P. intermedium*, *P. sylvaticum*, *P. recalcitrans*, *P. conidiophorum*, *P. ultimum* var. *sporangiferum*, and *P. ultimum* var. *ultimum*, were also pathogenic on alfalfa in the plate test. The majority of the *Fusarium* isolates were identified as *F. solani* and *F. oxysporum* with a low number of *F. redolans*, and *F. incarnatum-equisetum*. All species caused seed rot, damping-off, and root rot when tested in the agar plate assay. Strains of *F. oxysporum* and *F. incarnatum-equisetum* were the more aggressive in causing seed rot. Assays with Apron XL (mefanoxam) treated seed showed that sensitivity varied between and within *Pythium* species with approximately 56% of strains insensitive to the fungicide. In Apron XL amended medium, hyphal density was reduced at all concentrations but all strains had a similar growth rate as on non-amended medium. Insensitivity to Stamina seed treatments (pyraclostrobin) occurred in 94% of *Pythium* strains tested. The presence of broad host range species and fungicide resistance of a high percentage of *Pythium* isolates suggests that crop rotation and these widely used seed treatments are not effective tools for managing this disease. These results indicate that resistant cultivars are needed for managing damping-off in alfalfa production systems.