Specific Primers for Detection of Two *Sirococcus* Pathogens of Conifers and Comparison of PCR to Cultural Methods for Detection of *S. conigenus*

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Abstract – A PCR-based method was developed for the specific detection of the conifer shoot blight pathogen *Sirococcus conigenus* and the closely related fungus *Sirococcus tsugae*. Regions of diversity in the internal transcribed spacer (ITS) sequences of *Sirococcus* species were exploited to design primer pairs. Forward primer SirCf and reverse primer SirCr were used for identification of *S. conigenus*, and forward primer SirTf and reverse primer SirTr2 were used for identification of *S. tsugae*. Specificity was tested using multiple isolates of these two species, isolates of *S. piceicola* from spruce, *S. clavigignenti-juglandacearum* from butternut, and isolates of several other fungi obtained from pines. The PCR-based method for detection of *S. conigenus* was tested and results compared to those obtained using a cultural assay using shoots collected at six locations in Wisconsin and Michigan. For needles, bark, and wood of symptomatic shoots, the mean frequencies of detection of *S. conigenus* using the PCR-based methods were consistent ($\geq$ 7.5 out of 10) and always greater than for the cultural assay. For the cultural assays of symptomatic shoots, detection of *Sirococcus* spp. was more frequent from needles than bark or wood. Both the PCR-based method and the cultural assay detected *S. conigenus* in similar frequencies from asymptomatic shoots, though less frequently than from symptomatic shoots. The relative efficiency of our PCR-based method and its utility for direct testing of field-collected host material should make it particularly useful in areas of the western United States and Canada where both *S. conigenus* and *S. tsugae* have been found, and in situations in which other shoot blight pathogens also are commonly encountered.

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