

First report of *Heterobasidion irregulare* causing root rot and mortality of red pines in Minnesota.

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Heterobasidion annosum sensu lato is one of the most important pathogens in conifer forests of North America where it causes root rot and tree mortality (Garbelotto and Gonthier 2013). Although present in the eastern and western regions of the United States, the north central United States had been free of this pathogen until it was found in Wisconsin in 1993 (Stanosz et al. 1995) and the lower peninsula of Michigan in 2011. *Heterobasidion annosum* is now considered a complex of many species and *Heterobasidion irregulare* (Underw.) Garbel. & Ostrosina is the species that attacks pines in Wisconsin, Michigan and other regions of North America. Koch's postulates have been demonstrated for *Heterobasidion irregulare* on red pine (Stanosz et al. 1995). Despite surveys conducted over past decades in Minnesota, *Heterobasidion* root rot had not been found causing tree mortality. Two herbarium collections, identified as *Heterobasidion annosum* from Itasca State Park, Minnesota, were deposited in the U.S.D.A. Forest Service's Center for Forest Mycology Research, Madison Wisconsin in the 1970's but no evidence of the fungus causing root rot or tree mortality has ever been documented. Itasca State Park is located in north central Minnesota and one collection was noted to be from a downed *Picea glauca* and the other from *Abies*. DNA sequencing was attempted from each of these collections but it was not possible to obtain rDNA for analysis from them. The host substrate of spruce and fir suggests these collections may represent *Heterobasidion occidentale*. *Heterobasidion irregulare* has spread widely in Wisconsin and is currently found in 24 counties (Wisconsin Department of Natural Resources) with three of those counties adjacent to the border with Minnesota. Recently a survey conducted in red pine (*Pinus resinosa* Aiton) plantations that had been previously thinned in southeastern Minnesota found groups of dead and dying trees, occurring in a circular pattern, typical of root rot. Samples from the main stem at and below ground line and roots obtained from the dead and dying trees were placed in sterile bags, brought to the laboratory and cultured using a semi-selective media for isolating wood decay fungi (Worrall 1983). Cultures obtained produced asexual fruiting structures and spores typical for *Heterobasidion*. Genomic rDNA was extracted from a pure culture and directly from basidiocarps found at the site using a cetyl trimethylammonium bromide (CTAB) method (Beier et al. 2014) and PCR carried out as described by Arenz and Blanchette (2009) using general fungal primers ITS1F/ITS4 for cultures and basidiomycete specific primers ITS1F/ITS4B for basidiocarps and sequenced. BLASTn, searches of GenBank revealed that the sequences matched *Heterobasidion irregulare* at 100% and were deposited in GenBank with accession numbers KP202876 and KP202875. A voucher specimen of the basidiocarps has been accessioned into the Bell Museum of Natural History University of Minnesota Herbarium as MIN-938396. Red pine is an important tree species in Minnesota with an estimated 662,000 acres of red pine forests. The establishment of this pathogen in the state and knowledge of its spread throughout Wisconsin in the last 20 years demonstrates the need to implement management strategies for controlling the disease in Minnesota.

References:

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