

A COLLABORATIVE RESEARCH APPROACH FOR DIAGNOSING AND EVALUATING THOUSAND CANKERS DISEASE IN WALNUT: CURRENT PROGRESS AND FUTURE DIRECTIONS

Emel Oren¹, William Klingeman², Romina Gazis³, John Moulton¹, Paris Lambdin¹,
Massimo Faccoli⁴, Mark Coggeshall⁵, Jiri Hulcr⁶, Steven J. Seybold⁷,
and Denita Hadziabdic¹

Thousand Cankers Disease (TCD) results from interactions between the canker-producing fungal pathogen *Geosmithia morbida*, an insect vector *Pityophthorus juglandis* (walnut twig beetle), and the susceptible plant hosts, *Juglans* spp. (walnuts) and *Pterocarya* spp (wingnuts). In the past two decades, TCD has expanded from the western to the native range of black walnut (*J. nigra*) in the Eastern United States. In 2013, TCD was discovered in northwestern Italy affecting both black and native English walnuts (*J. regia*). Although TCD has caused significant mortality among native and non-native walnut tree populations, our understanding regarding the genetic diversity of the insect vector and the fungal pathogen remains limited. One factor that may be contributing to the spread of the disease is the difficulty of its diagnosis due mainly to non-unique external symptoms. Based on the speed at which TCD has expanded its range and the potential global spread and invasion of areas where susceptible host plants are commercially grown, there is a critical need to understand the genetic diversity presented by the causal agent and its primary vector. Moreover, to limit the spread of the disease we need to improve our detection methods by utilizing specific and sensitive molecular tools that enable quick identification. Rapid and accurate detection of TCD will facilitate quarantine implementation in infested areas. Collaborative research presented here showcases our current understanding of the population structure of both pathogen and the vector of this disease, as well as a recently developed molecular protocol to rapidly detect *G. morbida* and *P. juglandis* directly from woody tissue samples. Our results indicate high genetic diversity, presence of population structure, and evidence of gene flow among subpopulations of *P. juglandis* and *G. morbida*. In particular, our work reveals that human mediated movement of infested plant material from multiple sources and on multiple occasions, has significantly contributed to TCD range expansion. Our results support an earlier hypothesis that the disease has been established in western TCD-affected areas for a long period of time and can't be considered a recent introduction. An important by-product of our population genetics research was the development of a set of specific microsatellite regions that have been extremely useful in the detection of *G. morbida* directly from infested wood tissue, even at low concentrations. Here, we address how we developed this novel molecular detection tool and explain how this approach can serve as a model for future research on disease outbreaks caused by similar disease complexes.

¹ Department of Entomology and Plant Pathology, University of Tennessee, Knoxville, TN 37996; Current address Diyarbakör Plant Protection Research Institute, Diyarbakör, Turkey, (dhadziab@utk.edu).

² Department of Plant Sciences, University of Tennessee, Knoxville, TN 37996.

³ Department of Plant Pathology, University of Florida, Tropical Research and Education Center, Homestead, FL 33031.

⁴ Department of Agronomy, Food, Natural Resources, Animals and Environment (DAFNAE), University of Padua, Legnaro, PD, Italy.

⁵ USDA Forest Service, West Lafayette, IN 47906.

⁶ School of Forest Resources and Conservation, University of Florida, Gainesville, FL 32611.

⁷ USDA Forest Service, Davis, CA 95616.