

Influence of abiotic and biotic stresses in predisposing eastern black walnut (*Juglans nigra*) to establishment and spread of *Geosmithia morbida* and *Pityophthorus juglandis*, causal agents of Thousand Cankers Disease (TCD), in the eastern U.S.

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Widespread dieback and mortality of eastern black walnut (*Juglans nigra*) been observed since 2010 in PA, TN, and VA where black walnut is native and grows both as planted landscape tree in urban areas and as a naturally-occurring species across numerous forest cover types. The disease complex, known as Thousand Cankers Disease (TCD), involves a recently described fungus (*Geosmithia morbida*) and a walnut twig beetle (WTB, *Pityophthorus juglandis*), both of which are presumed native to the southwestern US, but outside the native range of black walnut. The recent discovery of TCD in Bucks County, Pennsylvania in August 2011 within the native range of black walnut have heightened concerns regarding the future of this species and its viability as a commercial timber species in the east, where the estimated value of walnut growing stock exceeds \$500 billion dollars. The threat of TCD to eastern tree populations appears to be intensifying, emphasizing the need for applied research to help inform management decisions and prioritize control efforts by identifying risk factors associated with establishment and spread of causal agents of TCD. Despite recent efforts to better understand TCD epidemiology, many questions remain.

The overall goal of our my research project research is to elucidate potential links among predisposing biotic and abiotic stresses of black walnut, site factors, and the establishment and spread of *G. morbida* and *P. juglandis* within the recent TCD-confirmed township in Bucks County, Pennsylvania. To this end, the proposed research will pursue this goal through several key objectives: 1) Analyze tree-ring patterns of healthy and diseased/declining BW colonized with ubiquitous canker pathogens outside TCD-confirmed areas to quantify baseline growth responses to biotic stresses and variations in climate, 2) Delineate the extent of TCD within PA and establish permanent monitoring areas to clarify the relationships between site factors, climate, tree health, and TCD establishment and spread, 3) Examine and quantify spatial and temporal relationships between initial colonization sites of *P. juglandis* and the location of pre-existing cankers and/or wounds on symptomatic trees within TCD-confirmed areas, 4) Establish a more succinct timeline for establishment and spread of TCD in the eastern US, and 5) Conduct tree-ring and statistical analyses of black walnut chronologies from both symptomatic and asymptomatic trees within TCD-confirmed areas to elucidate the effect of TCD on annual incremental growth and tree health and determine if decline precedes symptom development 6) Resolve phylogenetic relationships between TCD-associated fusaria and ubiquitous walnut-associated fusaria not associated with TCD areas.

In February 2012, 51 walnut trees were preemptively harvested with the assistance and cooperation of state and federal regulatory agencies as well as Penn State University researchers in Plumstead Township, Bucks County where TCD agents were first reported. Both stem cross-sections and branch sections were removed from felled trees and examined for the presence of WTB and for fungal isolation. Results show that of the 51 trees sampled, 23, only 5 of which were dead, had WTB with varying levels of gallery formation. Of these 23 trees, *Fusarium* sp. was consistently isolated from symptomatic tissue on 14 trees while *G. morbida* was observed at

much lower levels. Future research will focus on tree-ring analysis and phylogenetic studies of cultured fusaria to better understand the role of this fungus as well as predisposing factors on disease development.