E11 An Assessment of the Volatile Organic Compounds (VOCs) of an Agricultural Biothreat Agent, *Raffaelea lauricola*, and Training Aids for Canine Detection

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After attending this presentation, attendees will learn about an agricultural biothreat agent known as the laurel wilt pathogen. Attendees will gain an understanding of how research is combatting the spread of this biothreat by analyzing the Volatile Organic Compounds (VOCs) present in the headspace above the fungus in order to create a mimic training aid to be used in canine detection.

This presentation will impact the forensic science community by presenting a novel method of preventing the spread of a biothreat through canine detection. It will also strengthen the validity of canine detection as it is currently used in the fields of forensics and agriculture by showing the canines' ability to detect biothreats and distinguish diseased trees from uninfected ones.

Invasive biological agents pose a huge threat to the agriculture, environment, and economy of the United States. These biothreats can enter the United States through ports-of-entry accidently, as is the case of the invasive and phytopathogenic fungus *Raffaelea lauricola*. The current research identified the VOCs of the laurel wilt pathogen in order to create a mimic training aid. Using this aid, canines can be trained to detect infected trees before physical symptoms develop so that infected trees can be removed from groves and healthy trees protected. The vector of the *Raffaelea lauricola* fungus is the invasive redbay ambrosia beetle (*Xyleborus glabratus*), which entered the United States from Southeast Asia on infected wood. The symbiosis between the beetle and the fungus is a relationship where the beetle carries the fungus to a tree, bores into the host tree, and farms the fungus for food. The fungus colonizes the xylem of the tree, which then causes the tree to systematically shut down its transpiration mechanism in order to stem the spread of the infection. This leads to the tree's death within four to six weeks. Symptoms include wilting of the leaves and discoloration in the bark. Laurel wilt is advancing through the Lauraceae forests in the southeastern United States, and now is found in commercial avocado groves in Florida.

Because of the rate of advancement of the disease, a method of detection before the development of physical symptoms must be developed. Canines are often used in law enforcement and in ports-of-entry to prevent the entry of substances such known biothreats or banned agricultural items; however, novel biothreats such as *Raffaelea lauricola* have not been stopped at the port-of-entry and are now spreading through the forests and agricultural environments. The current study focuses on identifying the VOCs above the laurel wilt pathogen in infected trees through Solid Phase Microextraction/Gas Chromatography/Mass Spectrometry (SPME/GC/MS). The goal is to create a biological mimic with which to train canines to locate the pathogen in avocado groves so the diseased trees can be removed and healthy trees preserved. Bipolar SPME fibers were used to sample compounds in the headspace above samples of infected wood and uninfected wood. The compounds were then desorbed and separated through GC/MS. Results show that avocado trees infected with the laurel wilt produce different VOCs than trees that are uninfected by the pathogen. Based on these differences, a training aid will be created and subsequently verified through canine trials. This mimic training aid can then be used to combat the biothreat caused by *Raffaelea lauricola* and the skills and practices developed applied to the wider effort of protecting the nation's food supply from foreign invasive pests and other biothreats.

Biothreat, Canine Detection, *Raffaelea lauricola*