EPIDEMIOLOGY OF PIERCE'S DISEASE IN SOUTHERN CALIFORNIA: IDENTIFYING INOCULUM SOURCES AND TRANSMISSION PATHWAYS

Project Leaders:

Donald A. Cooksey
Department of Plant Pathology
University of California
Riverside, CA 92521

Heather S. Costa
Department of Entomology
University of California
Riverside, CA 92521

INTRODUCTION

Previous studies on the epidemiology of Pierce's disease (PD) of grape in Northern California have described systems dealing with different primary vector species and different alternate host plants than those that are found in the Southern California systems. Understanding the role that other plant species in the Temecula area may play in spread of Pierce's disease of grapes could be critical to management decisions. In addition to dealing with different host plants, the feeding habits and host range of the primary vector of the pathogen in Southern California differ from other primary vector species in Northern California. Studies with the insect vector species present in Northern California suggest that the pathogen was primarily spread by vectors moving into vineyards from outside habitats, rather than spreading from vine to vine. There is little information available on the relative ability of the glassy-winged sharpshooter to acquire or transmit the Pierce's disease pathogen from vine to vine, or from alternate hosts to grape. Because in many cases the vineyards of the Temecula area are in close proximity to citrus groves, it is critical to know the relative inoculum pressure that citrus and other plant hosts may provide in that area. Knowledge of the source of disease inoculum from vectors, whether from inside or outside the vineyard, will be critical to development of management strategies for disease control, such as the choice and management of plant species surrounding vineyards. Results of these studies, combined with data on seasonal fluctuations of sharpshooter populations, will also allow us to estimate the time of year and the regions where pathogen pressure is the greatest, and management strategies can be adjusted appropriately.

OBJECTIVES

1. Determine which plant species near vineyards harbor *Xylella fastidiosa (Xf)* and serve as potential reservoirs of inoculum for the spread of Pierce's disease to grapes.

2. Measure the ability of the glassy-winged sharpshooter to acquire and transmit *Xf* to and from grape, citrus, almond, and other plant species identified as potential hosts and sources of inoculum for the spread of Pierce's disease.

3. Comparison of the sensitivity and specificity of various methods to screen large numbers of plant and insect samples for the presence of Pierce's disease.

RESULTS AND CONCLUSIONS

Detection of *Xylella fastidiosa* in various plant species. Our regular sampling of over 60 plant species at 10 sites in the Temecula area is continuing. Samples are macerated in buffer and plated on selective media for *Xf*. All samples are also processed for serological (ELISA) and/or DNA-based (PCR) detection. Thus far we have detected *Xf* in several plant species in Temecula, including grapevine, almond, oleander, and Spanish broom. For these hosts, positive results were found using ELISA, PCR, and culture methods. Detection in mirror plant by ELISA was supported by PCR results, but we were not able to culture *Xylella* from these samples due to culture contamination. Spanish broom gave consistently strong ELISA reactions comparable to that of symptomatic grapevines, suggesting that the bacterium achieves a high titer in this new host. Weak positive results using ELISA were sporadically found with wild mustard, coyote brush, and elderberry; however, we were not able to confirm these results with other methods, suggesting that they could have been false positives.

Transmission studies. We have initiated greenhouse transmission studies testing the relative ability of *Xylella* to infect grape, citrus, almond, oleander, blackberry, bougainvillea, *Vinca* sp., toyon, coyote brush, *Brassica nigra*, brittlebush, mule fat, sage, California buckwheat, sugar bush, and laurel sumac. Our preliminary results suggest that grape-to-grape transmission can occur, as was suspected based on the observed widespread occurrence of PD in Temecula vineyards. We can now rapidly and reliably assay the glassy-winged sharpshooter for the presence of the pathogen, so we can follow it

during each step of the transmission process. We have also initiated experiments testing the ability of insects to transmit *Xylella* from Spanish broom to grape.

Evaluation of detection methods. We are continuing to evaluate the effectiveness of various methods for detecting *Xylella* in plants and in the insect vector. Immunocapture PCR is now incorporated into our regular screening procedures. We also have had success in developing strain-specific detection that allows us to differentiate between infections with either the Pierce's disease or oleander leaf scorch strains of *Xylella*. This was done by designing a new set of PCR primers to amplify a gene involved in xanthan gum biosynthesis that is present in all *Xylella* strains characterized (based on comparative genomic analysis). However, the sequence of the gum gene differs slightly between strains, and this can be easily detected by digesting the amplified gene with specific restriction endonucleases. Amplification of the gum gene from a sample indicates that *Xylella* was present, and subsequent digestion of the amplified product and gel electrophoresis yields two bands for the grape strain and one band for the oleander strain, or vice versa, depending on which one of two restriction endonucleases are chosen. This ability to detect which plants and insects are truly infected with the Pierce's disease strain will be essential for interpretation of our survey results in the Temecula area.