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

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INTRODUCTION

Knowledge of the source of disease inoculum from vectors, whether from inside or outside the vineyard, is critical to development of management strategies for disease control, such as the choice and management of plant species surrounding vineyards. In addition, 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.

OBJECTIVES

1. Determine which plant species near vineyards harbor *Xylella fastidiosa* 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 *Xylella fastidiosa* 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:

We are completing our third and final season of plant host sampling. We are still consistently getting positive detection of *X. fastidiosa* in several plant species in Temecula, including grapevine, oleander, Spanish broom and the few almond trees that remain. We also detected the presence of *X. fastidiosa* in *Brassica nigra* (wild mustard) by ELISA and PCR, but have not yet been able to culture it from this host. We increased the sampling of *Brassica nigra,* coyote brush, and elderberry and other weed and ornamental hosts that either appear symptomatic, or that have occasionally tested weakly positive with ELISA in previous years. We were never able to confirm positive results for coyote brush or elderberry with other methods, suggesting that they could have been false positives.

In other areas of Riverside, San Bernardino and Orange Counties some symptomatic landscape plants have tested positive for *X. fastidiosa.* Thus far, liquidamber, olive, mirror plant, and ornamental plum all tested positive by ELISA and PCR. We have also obtained cultures of *X. fastidiosa* from several samples of ornamental plum, but so far, have only been able to obtain one culture from olive samples. Several landscape plants, including olive and liquidamber, were repeatedly tested in the Temecula valley, but thus far, these species have not tested positive for *X. fastidiosa* in that area. The detection of *X. fastidiosa* does not necessarily mean that the bacterium is causing disease in these hosts; other pathogens or abiotic factors may be causing the observed symptoms. Additional studies will need to be conducted to determine if *X. fastidiosa* alone can cause disease in these species.

We are in the process of sequencing amplification products to identify the strains of *X. fastidiosa* that are infecting these new hosts.

Transmission studies:

Studies were initiated last year to test the ability of GWSS to transmit *Xylella* from infected grape to several species of host plants including: grape, lemon, grapefruit, orange, almond, oleander, blackberry, bougainvillea, toyon, coyote brush, *B. nigra*, brittlebush, mule fat, sage, California buckwheat, sugar bush, laurel sumac, tree tobacco, elderberry, alfalfa, peach, and coast live oak. One year after inoculation, sampling of test plants with ELISA and PCR found that transmission occurred only from infected grape to grape, and from infected grape to *B. nigra* plants. None of the other hosts have been confirmed positive thus far. Transmission experiments were also conducted to see if GWSS could transmit the pathogen from field-infected Spanish broom into grape test plants. In that study, 9/26 grape plants tested positive for the pathogen, indicating that Spanish broom may serve as a source of inoculum for Pierce's disease. Similar studies testing GWSS from greenhouse-infected *B. nigra* plants to grape found 1/9 grape plants became infected. This year, addition replicates of these species and

11 additional species (including Spanish broom) were initiated. Sampling two months after inoculation found only 1 grape plant tested positive so far.

Evaluation of detection methods:

We are continuing to evaluate the effectiveness of various methods for detecting *X. fastidiosa* in plants and in the insect vector. Both ELISA and immunocapture-PCR methods work well for plant samples. An additional method of extracting bacterial DNA from plants and insects using a commercially available kit was successful. This type of extraction can provide enough material for multiple PCR reactions to allow sequencing of DNA products. Strain specific primers have also been identified that can detect the OLS and PD strains of the pathogen. One primer set amplifies the PD but not the OLS strain, the other amplifies the OLS but not the PD strain. Although these primers pairs can be used to distinguish between these two strains, these primer pairs alone cannot necessarily distinguish these strains from all other strains that might be present in the environment.

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