

***XYLELLA FASTIDIOSA* TRANSMISSION BY GLASSY-WINGED SHARPSHOOTERS
AND SMOKETREE SHARPSHOOTERS FROM ALTERNATE HOSTS TO GRAPEVINES**

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ABSTRACT

The goal of this project is to evaluate the importance of many crops, cover crops and common weeds that are found in close proximity to vineyards as sources of *Xylella fastidiosa* (*Xf*) from which glassy-winged (GWSS) and smoketree (STSS) sharpshooters can acquire and transmit Pierce's Disease (PD) into grapevines. Acquisition from non-grapevine hosts and subsequent transmission to grape is of fundamental importance to primary spread of *Xf* in California vineyards. Identifying the plants that are contributing to primary spread enables growers to target these plants around their vineyards as a mechanism to reduce spread.

INTRODUCTION

Over 140 plants are known to host PD strains of *Xylella fastidiosa* (*Xf*) (Freitag 1951, Raju et al. 1980, 1983, <http://www.cnr.berkeley.edu/xylella/temp/hosts.htm>). Many of these plants are found in close proximity to vineyards, and some are even used as cover crops in vineyards. While considerable research has identified *Xf* hosts, little work has been done to determine if sharpshooters can acquire the bacteria from these hosts and transmit it to grapevines. If this does not occur, then the alternate host is of little consequence in PD epidemiology. Conversely, plants that contribute inoculum for sharpshooter acquisition and transmission to grape should be removed if growers wish to reduce primary spread into their vineyards.

To successfully implement a program to remove pathogen sources, we first must identify those sources. The introduction into California of GWSS, an insect with a broad host range, theoretically increases the probability of disease spread from the these alternate host plants to grape. For this to occur, GWSS must feed on the infected plant in such a way to acquire *Xf* from the plant, and successfully transmit the acquired pathogen to grapevines. While studies have shown mechanical and insect transmission to wide variety of alternate hosts (Freitag 1951, Purcell and Saunders 1999) they have demonstrated transmission from only a handful of alternate hosts to grapevines (Hill and Purcell 1995, 1997). We are unaware of research published on transmission of *Xf*, PD strain, from alternate hosts into grapevines using GWSS or STSS, a native California sharpshooter also found in grape growing regions, as the vector.

OBJECTIVES

Using GWSS and STSS vectors:

1. Evaluate the acquisition and transmission of *Xf* to grapevines from agricultural crop plants known to be PD hosts that are grown in the vicinity of vineyards.
2. Evaluate the acquisition and transmission of *Xf* to grapevines from weed plants known to be PD hosts that are grown in the vicinity of vineyards.
3. Evaluate the acquisition and transmission of *Xf* to grapevines from vineyard cover crop plants.

RESULTS

Plants for our studies were selected using the UC IPM lists of common weed and cover crop species found in and around California grape vineyards (Statewide IPM Program 2007) to determine the plants that would likely be found near vineyards. This list of plants then was cross-referenced with previously published inoculation studies and field surveys of *Xf* and we selected plants that developed systemic infections in 50% or more of the plants inoculated by Wistrom and Purcell 2005, and plants identified as *Xf* hosts in almond orchards (Shapland et al. 2006) and in vineyard areas of southern California (Costa et al. 2004). Additional plants were chosen because they are commonly found in or around vineyards, but they have not been previously inoculated with *Xf* or used in transmission studies for GWSS and STSS. Our plant list includes:

Table 1: Selected Plants

| Common Name | Scientific Name | Plant Type |
|-----------------------------|--------------------------------|-------------------|
| Alfalfa | <i>Medicago sativa</i> | Agriculture Crop |
| Bell Pepper | <i>Capsicum annuum</i> | Agriculture Crop |
| Cotton, Upland | <i>Gossypium hirsutum</i> | Agriculture Crop |
| Lima Bean, Fordhook 242 | <i>Phaseolus lunatus</i> | Agriculture Crop |
| Tomato, Rutgers | <i>Solanum lycopersicum</i> | Agriculture Crop |
| Annual Bluegrass | <i>Poa annua</i> | Weed |
| Annual Bursage | <i>Ambrosia acanthicarpa</i> | Weed |
| Black Nightshade | <i>Solanum nigrum</i> | Weed |
| Cheeseweed | <i>Malva parviflora</i> | Weed |
| Chickweed | <i>Stellaria media</i> | Weed |
| Cocklebur | <i>Xanthium strumarium</i> | Weed |
| Common Groundsel | <i>Senecio vulgaris</i> | Weed |
| Common Morning Glory | <i>Ipomoea purpurea</i> | Weed |
| Common Sunflower | <i>Helianthus annuus</i> | Weed |
| Field Bindweed | <i>Convolvulus arvensis</i> | Weed |
| Filaree | <i>Erodium sp.</i> | Weed |
| Goosefoot | <i>Chenopodium sp.</i> | Weed |
| Horseweed | <i>Conyza canadensis</i> | Weed |
| London Rocket | <i>Sisymbrium irio</i> | Weed |
| Poison Hemlock | <i>Conium maculatum</i> | Weed |
| Prickly Lettuce | <i>Lactuca serriola</i> | Weed |
| Shepherd's Purse | <i>Capsella bursa-pastoris</i> | Weed |
| Spanish Broom | <i>Spartium junceum</i> | Weed |
| Speedwell | <i>Veronica sp.</i> | Weed |
| Stinging Nettle | <i>Urtica sp.</i> | Weed |
| Tree Tobacco | <i>Nicotiana sp.</i> | Weed |
| Alyssum | <i>Alyssum sp.</i> | Cover Crop |
| Annual Ryegrass | <i>Festuca sp.</i> | Cover Crop |
| Annual Fescue, Zorro | <i>Lolium multiflorum</i> | Cover Crop |
| Black Mustard | <i>Brassica nigra</i> | Cover Crop |
| Birdsfoot Trefoil | <i>Lotus spp.</i> | Cover Crop |
| Buckwheat | <i>Fagopyrum sp.</i> | Cover Crop |
| Burr Medic | <i>Medicago polymorpha</i> | Cover Crop |
| Cilantro | <i>Coriandrum sativum</i> | Cover Crop |
| Clover, New Zealand White | <i>Trifolium repens</i> | Cover Crop |
| Clover, Hykon Rose | <i>Trifolium hirtum</i> | Cover Crop |
| Cowpea, California Blackeye | <i>Vigna unguiculata</i> | Cover Crop |
| Fava Bean, Windsor | <i>Vicia faba</i> | Cover Crop |
| Field Pea, Miranda | <i>Pisum sativum</i> | Cover Crop |
| Oat, California Red | <i>Avena sativa</i> | Cover Crop |
| Sweetclover | <i>Medicago sp.</i> | Cover Crop |
| Vetch, Cahaba White | <i>Vicia sativa</i> | Cover Crop |

Seeds have been obtained for all of the agricultural and cover crops and we have secured seeds for 11 of the 21 weed species listed in Table 1. We have completed inoculations of 7 plant species (Table 2). From the agricultural crop group, we have worked with bell pepper and tomato. Bell pepper appears to be a non-host for PD. Five of 20 PD-inoculated bell pepper plants tested positive with ELISA at 2-weeks, but they all tested negative at 4-weeks, suggesting a transient infection. Attempts to isolate *Xf* from bell pepper by culturing have been negative. A third culture attempt for all inoculated bell pepper plants is currently underway and final results are pending. Of the 20 tomato plants inoculated with PD, 1 died before the first assay, and 10 have tested positive with ELISA. PD has been successfully isolated from these plants and confirmed with PCR. Insect transmissions with both sharpshooter species have been done for bell pepper and data are being collected on this study. Transmissions from tomato to grapevine will be conducted in late October.

From the weed category, we have studied goosefoot and it appears to be a non-host for PD. Only 1 of the 20 PD-inoculated plants tested positive and all cultures have been negative. A third isolation attempt is underway for all 20 goosefoot test plants and final results are pending. Experiments on four cover crop species (buckwheat, cowpea, fava bean, and field pea) are underway. All four tested positive for *Xf* with ELISA and bacteria was successfully cultured from these plants (Table 2). Two of the 20 buckwheat plants died before the two-week assay. Most of the field pea plants were dying by two-weeks post-

inoculation. Three were dead at four-weeks post-inoculation and at five-weeks post-inoculation there were only six plants alive, of which one was confirmed positive for PD, but it was in poor condition and could not be used for insect transmission. Field pea inoculations will be repeated in the spring 2008. Insect transmissions have been done for buckwheat and cowpea and we are in the process of evaluating those studies. Insect transmission from fava bean to grape is planned for late October 2007.

Table 2: First Test Group Needle Inoculation Results.

| Plant Type | Plant Name | # Inoculated | # Positive 2-Week ELISA | # Positive 4-Week ELISA | Cultured |
|-------------|-------------|--------------|----------------------------|----------------------------|----------|
| Agric. Crop | Bell Pepper | 20 | 5 | 0 | Negative |
| Agric. Crop | Tomato | 19 | 5 | 6 | Positive |
| Cover Crop | Buckwheat | 18 | 5 | 12 | Positive |
| Cover Crop | Cowpea | 20 | 4 | 7 | Positive |
| Cover Crop | Fava Bean | 20 | 11 | 17 | Positive |
| Cover Crop | Field Pea | 20 | 4 | 9 | Positive |
| Weed | Goosefoot | 20 | 0 | 1 | Negative |
| Control | Grapevine | 20 | 9 | 16 | Positive |

CONCLUSIONS

This project addresses the 2006 Scientific Summit category of “*Understanding transmission of the disease,*” and relates directly to the acquisition and transmission of *Xf* by GWSS. It also has relevance to several of the recommendations developed by the National Academy of Science, National Research Council (2004). First and foremost, the definition of the Category 1 research option is that it “holds a reasonable promise of generating successful tools for management of PD/GWSS, either in the short term or the long term.” By determining the plants that truly contribute to primary spread by sharpshooters, we can give growers another strategy (i.e. removing those plants) in an effort to reduce bacterial inoculum around their vineyards. This proposal also meets the general criteria defined in the NRC report in recommendation 2.2, of “contributing to PD/GWSS management and its sustainability,” and it applies specifically to recommendation 3.9 of examining plants “for effective transmission rates from host to grape.”

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