WEB GEOGRAPHIC INFORMATION SYSTEMS FOR PIERCE'S DISEASE AND GLASSY-WINGED SHARPSHOOTER MAPPING IN CALIFORNIA

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ABSTRACT
The establishment of the non-native glassy-winged sharpshooter (GWSS) in California has seriously affected the epidemiology of Pierce's disease (PD) throughout the agricultural landscape. Geographic Information Systems (GIS) offers the opportunity to aid in the management of PD as well as in epidemiological research. We developed a web-based GIS site with spatial and temporal data relating to PD/GWSS based on feedback from a web-based survey emailed to 2005 PD research symposium participants. The survey focused on participants' interests in PD-related data, spatial analysis, and additional comments. The resulting webGIS displays various data layers of value to PD/GWSS researchers, including climatic variables and proximity analyses. Our survey results indicate an interest among PD/GWSS researchers in temporal analyses and some interest in data sharing. In addition, the data survey provides insight to PD researchers' attention to investigating PD patterns at a landscape scale and spatial modeling. However, there still exist some barriers preventing access to all statewide PD and GWSS data that will have to be overcome in order to develop and maintain a comprehensive statewide PD/GWSS webGIS system.

INTRODUCTION
Pierce's disease (PD), caused by the *Xylella fastidiosa* (*Xf*) bacterium, has been present in California for about 100 years. However, the introduction of the non-native glassy-winged sharpshooter (GWSS), *Homalodisca vitripennis*, in recent years has increased the ability for the bacterium to spread. GWSS and PD have the ability to invade areas outside of their natural range and pose serious threats to the health of agricultural crops such as alfalfa, almond, citrus, coffee, grape, peach, plum and oleander (Hoddle, 2004). In particular, the GWSS vector has contributed to above normal grapevine losses in Southern California and could influence grapevine losses in Northern California.

Past studies indicate a linkage between the local environment and PD incidence and spread. For instance, Hoddle (2004) indicated the improbability of GWSS colonizing areas north of California due to the climatic condition of cold stress. Fiel and Purcell (2001) found temperatures below 12 to 17º C and above 34 º C to negatively affect *XF* growth in vitro and in potted grapevines. Also, proximity to citrus crops or riparian habitats appears to influence PD incidence (Perring et al. 2001; Purcell and Saunders, 1999). The intersection of Geographical Systems (GIS) and the Internet has allowed the provision and visualization of geospatial data over the web possible. Web-based GIS (webGIS) provides insight into relationships between environmental variables at multiple scales to aid in natural resource management (Kearns et al., 2003). The ease of web-based access to spatial data is particularly advantageous for individuals to develop epidemiological hypotheses about distribution and spread at several scales – from vineyard to county to regional. This paper documents our progress in developing one for the PD/GWSS community.

OBJECTIVES
The goals for this project were twofold:
1. Provide researchers with a web-based tool to access spatial and temporal data relating to PD/GWSS in California at the landscape scale
2. Provide initial spatial analysis of known crop relationships to GWSS movements.

To achieve these goals, we developed a web-based GIS site and conducted a web-based survey to acquire user input about data needs relating to GWSS/PD research.

METHODS
WebGIS
We developed a webGIS site titled "Pierce's Disease & GWSS Mapping" utilizing ESRI's ArcIMS software (Figure 1). Determining which data to include in the webGIS was based on an initial evaluation of publicly available GIS data relevant to GWSS/PD. The GIS data were then downloaded from the web, processed, and integrated in the webGIS and finalized after survey responses were assessed. In addition to collecting data from the web, we developed and included a "Growing Degree-Days for 2005" and "Weather Stations" spatial data layers (Figure 2) based on a non-spatial degree-days model by UC Statewide Integrated Pest Management Program (http://axp.ipm.ucdavis.edu/WEATHER/ddretrieve.html).

Figure 1. Opening page of PD/GWSS Mapping website, located at:

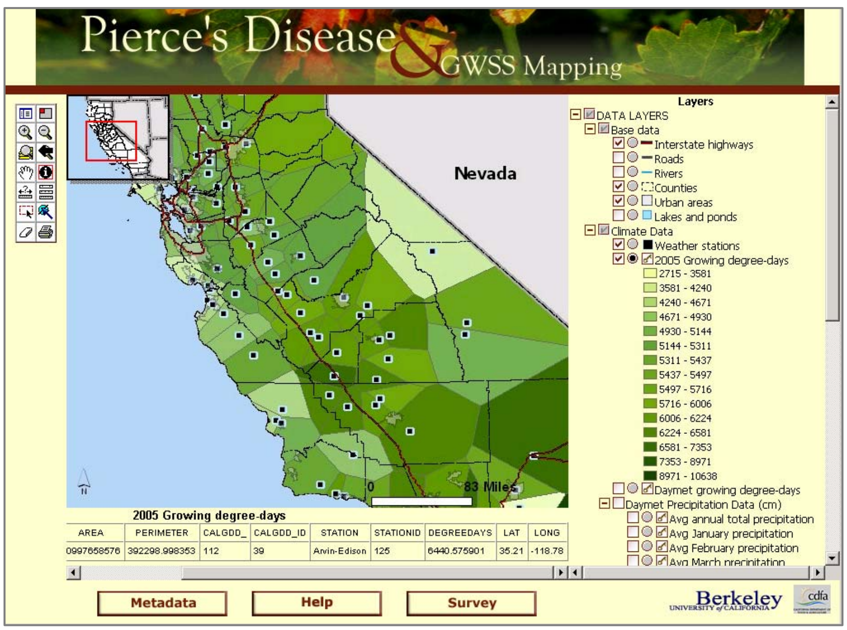

Figure 2. 2005 growing degree-days and weather stations data layers displayed with identify tool results

The data collected for the webGIS were grouped into three categories: base data, climate data, and vegetation data. A fourth category is based on data layers we developed based on a spatial analysis of proximity to crops. A fifth category of PD/GWSS data will be included when the data are publicly available.

The webGIS site is completely functional with zoom tools, navigation tools, measurement tools, feature selection tools and an identify attributes tool. An example of the identify tool is seen in figure 2, which is particularly useful for users to obtain information for an exact area of interest. The webGIS site also has a metadata link for users to view important details about each data layer.

Data Survey

A data survey was developed for participants to answer questions and tell us which data and/or analysis they thought to be potentially useful in understanding PD. The survey provided user input in determining data content to include in the webGIS as well as assessing future research topics.

The data survey was presented to 2005 PD research symposium participants in the form of a website (http://giifserv.cnr.berkeley.edu/website/PD/Survey/PDfeedback.htm). The survey had a total of eight questions, focusing on data interests, possible spatial analysis interests, and text boxes for participants to give additional suggestions at the end of each question. The survey included a link to our webGIS site for participants to view data examples from those listed in the survey. The survey responses were collected and stored in a database.

RESULTS

Data Survey

We had a 20% response rate to our survey. An estimated 175 people were sent out emails requesting their input and a total of 32 responded. All 32 survey participant responses were assessed in early June 2006.

The data with the highest interest is shown in figure 3. We were particularly interested in responses relating to climate data, which generated considerable interest. Climatic factors max/min temperatures, temperatures, growing degree days, number of frost day, precipitation, humidity, and dew point resulted in high levels of interest. Also apparent in figure 3 is the higher interest in monthly data (44%-72%) instead of annual data (38%-47%) when applicable. In general, there was more interest in Daymet data (1km resolution) over PRISM data (4km resolution), except where PRISM offers a dew point, a climatic variable Daymet does not include. In addition, there was a general consensus to have most of the base data listed in the survey included in the webGIS. The survey yielded the highest data interest in riparian vegetation (84%).

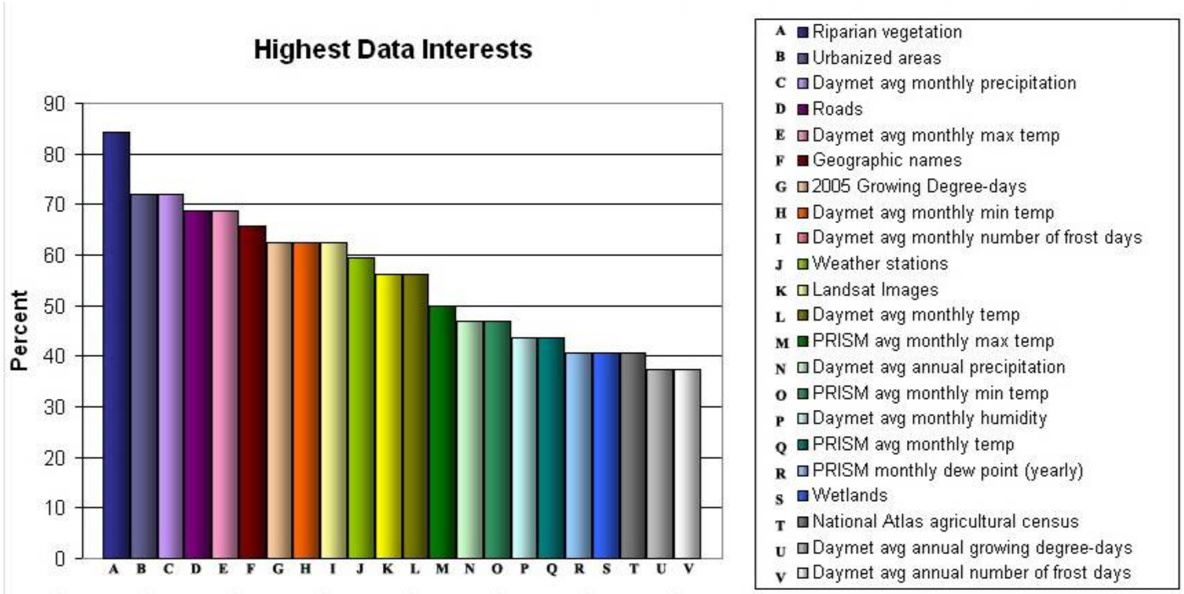

Figure 3. Data with interests levels above 35%

The spatial modeling questions relating to proximity and a change over time analyses received much attention. The respondents' interest in temporal analysis (66%) and proximity analysis (97%) was very high. This high level of interest in spatial analyses directed us to perform a first round proximity analyses.

Spatial Analysis

Due to high interest from survey participants, an initial proximity analysis was performed based on information about GWSS movement and its connection to PD incidence. It has been indicated that vineyards within close proximity to citrus orchards are susceptible to PD incidence (Perring et al. 2001). In addition, Purcell and Saunders (1999) indicated a connection between PD incidence and proximity to riparian vegetation. An initial proximity analysis was conducted in which grape and citrus crops data layers were used for the analysis; the crop data layers, supplied by CDFA, are based on based on 2003 Pesticide Use Report (PUR) and aggregated to PLSS sections. A new data layer was created with all grape sections that intersect with citrus sections. A total of 20 % of the grape sections that intersect citrus sections, known as the citrus-grape interface, indicated a high potential for PD incidence. Another data layer with a 375m buffer from streams was created to

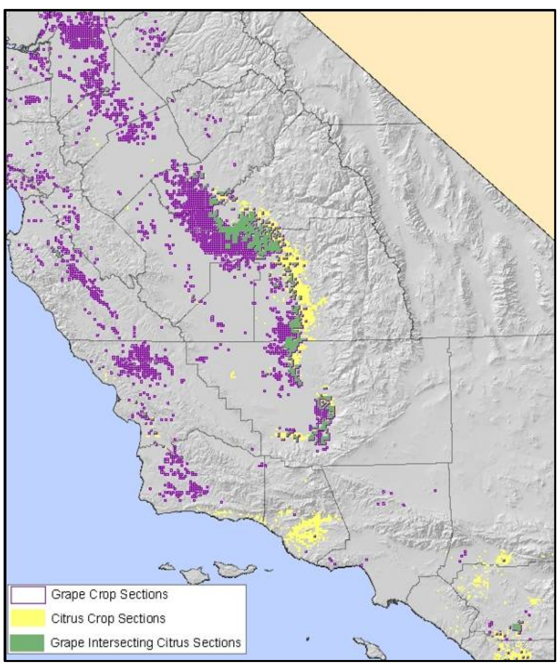

Figure 4. Map highlighting grape crop sections intersecting citrus crop sections (shown in green)

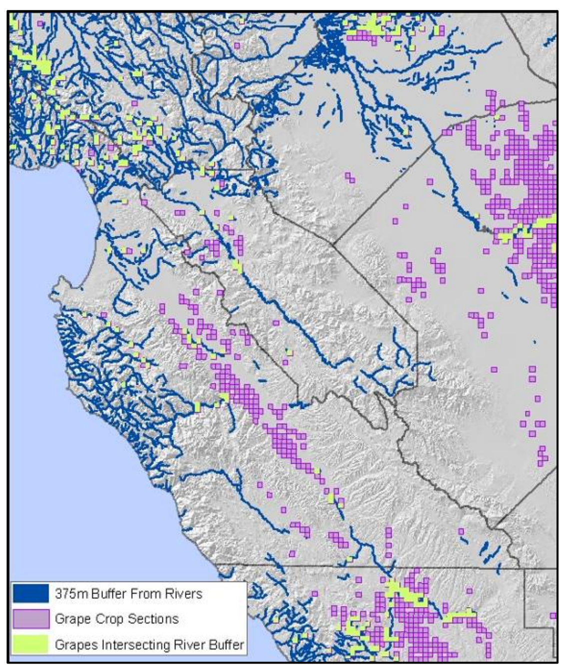

Figure 5. Map highlighting grape crop sections intersecting buffered rivers (shown in light green)

represent possible riparian habitat, a host habitat for GWSS. The results of these data intersections are available to researchers via the webGIS site (http://giifserv.cnr.berkeley.edu/website/PD/viewer.htm).

CONCLUSIONS

Our webGIS site will be a resource of spatial data for researchers to assess spatial phenomenon such as climatic effects on PD/GWSS. The data survey provides insight to PD researchers' interest in investigating PD patterns at a landscape scale and spatial modeling. There is particular interest in temporal analysis, which is a potential topic for future research. In addition, a little over half (54%) of the survey participants are interested in sharing their data with the rest of the research community. This indicates another area of research: the development of tools for researchers to upload their data to our webGIS, making it a central resource for accessing data for PD. We also hope to continue working on the development of the webGIS site by including enhanced query tools such as, select by attribute and allowing the user to define thresholds (eg. growing degree-days threshold). These areas of development would advance the PD/GWSS webGIS site to a more comprehensive statewide resource to assess spatial data at multiple scales.

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ANALYSIS OF THE BACTERIAL COMMUNITY INHABITING GLASSY-WINGED SHARPSHOOTER FOREGUT BY CULTURE-DEPENDENT TECHNIQUES AND DGGE

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ABSTRACT

The glassy-winged sharpshooter (GWSS) is an important vector of *Xylella fastidiosa* (*Xf*), the bacterial pathogen that causes several economically important plant diseases, including citrus variegated chlorosis (CVC), oleander leaf scorch (OLS) and Pierce's disease (PD) of grapevines. In recent years there has been an increasing interest in the potential use of biological control agents to halt the spread of *Xf*. One such strategy is the exploration of symbiotic microorganisms to reduce the spread of the pathogen (symbiotic control). In a symbiotic control strategy a bacterium symbiont that occupies the same niche as the pathogen must be identified. The study of the bacterial community of GWSS foreguts by isolation and DGGE revealed the presence of several potential symbiotic candidates such as *Bacillus* sp., *Pseudomonas* sp., *Methylobacterium* sp. and *Curtobacterium flaccumfaciens* (*C. flaccumfaciens*). Members of genus *Methylobacterium* and *C. flaccumfaciens* are frequently isolated as endophytes from citrus plants with CVC symptoms and without disease symptoms. Recently, an interaction between *Methylobacterium*, *C. flaccumfaciens* and *Xf* was strongly indicated, reinforcing that these bacteria could interact inside the host plant and vector insect. In the future, the genus *Methylobacterium* and *C. flaccumfaciens* could be an interesting candidate in a strategy of symbiotic control to *Xf*.

INTRODUCTION

The glassy-winged sharpshooter (GWSS) is one of the main vectors of *Xylella fastidiosa (Xf)*. It is a xylophagous insect that has a wide array of host plants, including many ornamental and crop plants (Purcell and Hopkins 1996, Purcell and Saunders 1999). One new potential management strategy for Pierce's disease (PD) of grapevine is the use of symbiotic control. Symbiotic control exploits the interactions among a pathogen-transmitting organism, its bacterial symbionts, and the pathogenic organism itself (Beard 2002). For symbiotic control to be effective in limiting the spread of PD, a culturable symbiont that inhabits the pre-cibarium and cibarium of GWSS is required, since these areas are colonized by the pathogen, *Xf*. A previous biochemical analysis of the GWSS foregut microdiversity encountered three bacterial species that meet these requirements: *Chryseomonas* spp, *Ralstonia* spp, and *Alcaligenes* spp (Bextine 2004). The *Alcaligenes* species were of particular interest because they were frequently isolated from wild GWSS (Kuzina 2004). Although *Alcaligenes* spp. can colonize GWSS, this specie does not colonize grapevines well (Bextine 2005). According to Bextine et al (2005), the amount of colonization by *Alcaligenes xylosoxidans* subsp. *denitrificans* decreased in the following order: orange (*Citrus sinensis* ‘‘sweet orange’’) > chrysanthemum (*Chrysanthemum grandiflora* cv. ‘‘White Diamond’’) > periwinkle (*Vinca rosea*) > crepe myrtle (*Lagerstroemia indica*) > grapevine (*Vitis vinifera* cv. Chardonnay). Therefore, steps in a symbiotic control strategy should be directed in finding a bacterial symbiont that colonizes well the pre-cibarium and cibarium of GWSS as well as the target host plant: grapevines. The CelectrophoreticCT separation of PCR products of variable regions of genes encoding 16S rDNA (by use of primers homologous to conserved regions of the gene) in a polyacrylamide matrix over a denaturing gradient (DGGE) is a technique recently introduced in microbial ecology (Muyzer et al. C1993CT). The denaturing gradient can be achieved either chemically with urea and formamide in DGGE. This technique is reported to be interchangeable, giving comparable fingerprints of microbial communities. Recently, Reeson et al. (2003) demonstrated the efficiency of DGGE to study the bacterial communities associated to insects (wasp *Vespula germanica*).

OBJECTIVES

1. The aim of this work was to characterize the bacterial community inhabiting GWSS foreguts by using isolation in culture medium and DGGE techniques.

RESULTS

The study of bacterial community by isolation and DGGE (Figures 1, 2, and 3) revealed the presence of several genera of bacteria such as *Bacillus* sp., *Cryocola* sp., *Microbacterium* sp., *Micrococcus* sp., *Pedobacte* sp.r, *Pseudomonas* sp., *Methylobacterium extorquens*, *C. flaccumfaciens*, *Baumannia cicadellinicola*, and *Wolbachia* spp. Members of genus