



Texas Research and Education Program to Combat Pierce's Disease

Project Summaries for Research planned for 2005

The research summaries in this document were prepared and submitted in January 2005, and represent anticipated continuing and new work on Pierce's Disease by Texas scientists for the period April 2005 through March 2006.

Major Research or Education Activities – February 2005

David N. Appel

TAES – College Station - Dept. of Plant Pathology and Microbiology

1. Compare Real Time PCR, ELISA, and field diagnosis of Pierce's Disease in different grape cultivars – continuing high priority.
 - a. Importance:
 - i. Texas growers need a local diagnostic service for reliable, consistent diagnosis of Pierce's Disease.
 - ii. There is an unacceptably high degree of uncertainty involved in diagnosing Pierce's disease with existing technology.
 - iii. Cultivars and varieties vary in their responses to *Xylella fastidiosa*, confusing diagnostic efforts.
 - iv. Completion of this study will provide growers with a dependable diagnostic service.
 - b. Timeline
 - i. Samples for laboratory analysis will be collected in May from groups of vines in various stages of disease development, as rated in previous surveys - sampled vines will be from different cultivars.
 - ii. The same vines will be monitored biweekly during the growing season for symptoms and additional sampling.
 - iii. The final sampling will be in October, when data analysis will compare the performance for ELISA and real time PCR.
 - iv. Results will be ready for reporting before December.
 - c. Technology transfer
 - i. The results of this work would be best appropriate for the January 2006 growers meeting.
 - ii. Some of the information will be incorporated into the Diagnostic Guide to be finished by March 2006.
2. Epidemiology – sequential analysis of Pierce's disease progress within vineyards – continuing high priority.
 - a. Importance:
 - i. Disease progress is a function of numerous underlying factors that influence the spatial distribution and temporal dynamics of disease incidence and severity.
 - ii. The mechanisms of vine-to-vine spread by *Xylella* are poorly understood.
 - iii. Reliable testing of control methods will require a thorough understanding of disease progress.
 - b. Timeline
 - i. Two years of data are already collected, and have been reported at previous growers meetings.
 - ii. The third year of data, to be finished in October, will provide three data points, the minimum needed to analyze disease progress curves.
 - c. Technology transfer
 - i. Data analysis will be completed in Spring 2006, and will be published in the journal Plant Disease.
 - ii. This work will be presented at the 2005 California PD meeting.
3. Studies on direct control of Pierce's disease with Cambistat[®] - new high priority.
 - a. Importance
 - i. There are currently no means available to treat vines for infection with *X. fastidiosa*.

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- ii. If successful, this compound would extend the function lifespan of vines colonized by the Pierce's disease pathogen, thus increasing productivity and lowering costs of production.
 - b. Timeline
 - i. Success of these treatments will require at least two years to fully evaluate.
 - ii. The rates of disease progress in the treated populations will be compared to untreated controls.
 - c. Technology transfer
 - i. Results of the treatments will be reported at the January 2007 growers meeting.
- 4. Development of a Brochure Depicting Diagnosis of *Xylella* Diseases of Plants in Texas.
 - a. Importance
 - i. There are no diagnostic guides available to identify Pierce's disease and related disorders of plants in Texas.
 - ii. The first step of any plant disease management program begins with diagnosis.
 - iii. Pierce's disease is a difficult problem to identify, and this brochure will provide a guide for helping growers to diagnose the disease and communicate with other growers and resource providers about their scenarios.
 - b. Timeline
 - i. Photographs of symptoms will be taken during the 2005-growing season on grapes, sycamores, red oaks, and oleanders.
 - ii. Information will be derived from the research in Part A of this proposal, and the brochure will be completed by March 2006.
 - c. Technology transfer
 - i. The brochure will be made available to growers through the TCE website and information system.
 - ii. The brochure will be distributed at growers meetings and CEA offices throughout high-risk areas.

Mark C. Black

TCE - Plant Pathology - Uvalde

- 1. Surveys near Texas Hill Country vineyards (two with PD history, two without PD history) for supplemental plant hosts of *Xylella fastidiosa*.
 - a. Importance
 - i. Knowledge of *X. fastidiosa* plant reservoirs near vineyards with PD adds to our understanding of high risk sites, provides growers with criteria for selecting low-risk sites for future vineyards, and allows growers to target certain weed species for control efforts.
 - ii. Supplemental hosts collected to date near vineyards are all in Asteraceae; that four annual weeds and two perennials have no symptoms suggests long-term ecological vector/plant/pathogen relationships in Texas that may be unique from CA.
 - iii. No supplemental hosts in Texas to date in Poaceae, Cyperaceae, and Juncaceae (CA reports one *Juncus* sp. as *X. fastidiosa* host) indicates potential use/management of grasses, sedges, and rushes in and around vineyards for erosion control, prevention of soil compaction, water conservation, and broadleaf weed control through competition.

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- iv. Entomologists should include known *X. fastidiosa* supplemental hosts in their studies on sharpshooter/spittlebug feeding/reproduction hosts.
- v. Beneficial insect research should include searches for egg mass parasitism on supplemental hosts.
- b. Timeline
 - i. Additional plants reacted to *X. fastidiosa*-specific serology in both 2003 and 2004, but were not confirmed with dilution plating. We need to add other techniques in 2005 to resolve *X. fastidiosa*-host status of *Grindelia nuda* var. *nuda* (perennial Asteraceae) and non-Asteraceae *Vitex agnus-castus*, *Salix nigra*, *Lantana* sp., and *Sapindus saponaria* var. *drummondii* and others. Following discussions with A.H. Purcell, we will modify techniques on these problem plant species in 2005. This includes 1) modifying serology (ELISA) techniques with a healthy plant-based sap for one buffer to avoid/minimize false positives; 2) extracting xylem contents for dilution planting with alternative techniques to avoid *X. fastidiosa* inhibiting materials and bacterial/fungal contaminants from non-xylem tissues, and fluorescent light microscopy with commercially available *Xylella* antibody (requires purchase of new microscope). Another option is to purchase standard PCR or RT-PCR for use with *X. fastidiosa*-strain-specific primers on plant material from the field. Currently, *X. fastidiosa* non-grape isolates are being sent to Blake Bextine at UC Riverside for strain determination.
 - ii. We expect to resolve status of some plants by Dec 2005, but probably not all problem species will be resolved that soon.
- c. Technology transfer
 - i. Posters at APS annual meeting Austin, TX 2005 and Dec 2005 PD Research Symposium in San Diego, CA.
 - ii. Updates and summaries posted periodically on Texas PD Working Group Website
 - iii. Presentations to growers at meetings
 - iv. One-on-one contacts by e-mail, telephone, site visits.
2. Comparisons of plant communities near Texas Hill Country vineyards (two with PD history, two without PD history).
 - a. Importance
 - i. This work (4 100-m transects at each of 4 vineyards) complements the supplemental XF host work by characterizing botanical diversity at sites with and without PD.
 - ii. Poaceae species are more frequent at putative low risk (no-PD) sites than at the two PD sites.
 - iii. USDA ARS personnel need these data to assess potential for beneficial insect releases to survive on endemic plants (e.g., one species being studied feeds preferentially on Apiaceae nectar.
 - iv. Isabelle Lauziere needs these data and our expertise/herbarium sheets to identify plants where various sharpshooters are observed feeding-reproducing and where egg masses are parasitised.
 - b. Timeline
 - i. Transects at one point-in-time (fall 04) has been completed, and Joe Patt, USDA ARS has expressed interest in additional plant surveys in spring 05 to have record of spring annuals.

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- ii. Some species were not in vegetation/flower/fruit and were not identified or collected in the fall 04 work, so our plant lists and species frequencies are incomplete.
 - c. Technology transfer
 - i. Posters at APS and PD Research Symposium meetings.
 - ii. Presentations at grower meetings
 - iii. Postings on TX PD website
 - 3. Characterizing *X. fastidiosa* strains recovered from supplemental plant hosts near vineyards, and from urban landscape hosts.
 - a. Importance
 - i. We need to determine which supplemental plant hosts harbor strains of *X. fastidiosa* pathogenic to grape-specific *X. fastidiosa* strains. If certain weeds harbor only *X. fastidiosa* that are not pathogenic to grape, weed control can concentrate better on the weeds infected with grape *X. fastidiosa* pathogens (*Ratibida columnifera* and *Ambrosia psilotachya* may be in this category). Realistically, some weeds probably harbor both grape and non-grape strains, so numerous isolates from each supplemental host should be determined to strain level.
 - ii. In CA, isolates from urban sites were usually not pathogenic to grape. If urban landscape species in TX harbor *X. fastidiosa* pathogenic to grape, we should caution vineyard/winery owners about landscape plant choice.
 - iii. Plants hosting non-grape *X. fastidiosa* strains have potential for future studies on evolution of *X. fastidiosa* in TX and research on biocontrol with bacteriophage.
 - iv. We have grape vines (cv. Chardonnay) in a greenhouse at Uvalde and will be inoculating with selected grape and weed isolates in 2005 in attempts to determine pathogenicity. This is much slower and labor/space intensive than PCR with strain-specific primers.
 - b. Timeline
 - i. We should have some isolates characterized by Dec 05, but new isolates, and isolates from not-yet-resolved plants will need to be characterized in sufficient numbers to estimate risk they pose to vineyards.
 - c. Technology transfer
 - i. Same as above.
 - 4. Study soils from vineyards without and without PD under controlled shade/screen-house conditions (New study in 05 with Kamas).
 - a. Importance
 - i. Test hypothesis that some vineyards have no PD because of some soil factor. If we observe differences, then research would be indicated to determine nature of soil factor, and if it could be affected by vineyard management. If no soil differences occur, site differences may be related to plant communities, e.g. some supplemental hosts that do not grow well on granitic sands, or past land use has altered weed species at some sites.
 - ii. This will help answer questions about site selection, vineyard management, and vegetation management in high-risk areas of TX and CA.
 - b. Timeline
 - i. 2005 will be year 1 of this work, and it should be repeated at least once, so perhaps Dec 06.

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- c. Technology transfer
 - i. Same as above.
- 5. Screening rootstocks commonly used in TX for reaction to *X. fastidiosa* (Black & Kamas, perhaps also Morano)
 - a. Importance
 - i. Rootstocks vary in reaction to *X. fastidiosa* based on work at Cornell/FL, and in CA. We need information on those used in TX, because some are unique, e.g. need for resistance to *Phymatotrichopsis omnivorum* (cotton root rot) in TX.
 - ii. We need to know which rootstocks to avoid in high-risk areas of TX and CA. Resistant scions on susceptible rootstocks would likely fail and cause growers to avoid very useful scion cultivars.
 - b. Timeline
 - i. Starting this project in 2005, with 12 entries (5 reps) in Llano Co. vineyard with long and severe history of PD. Vineyard owner is willing to provide space, irrigation, and all management once vines are planted, so our expense would be primarily for monitoring and sampling plants for *X. fastidiosa* and symptoms for 3 years. Expect to terminate trial in Dec 07.
 - ii. The Llano Co. site probably has cotton root rot (personal communication, G. R. McEachern), so this trial may need to be repeated at a PD site with no history of cotton root rot starting in 2006 or later. Bell Co. grower that works with Morano is a possibility.
 - c. Technology transfer
 - i. Same as above.

Edward W. Hellman

TCE & Texas Tech University – Viticulture - Lubbock

- 1. Texas statewide vineyard survey.
 - a. Importance
 - i. A comprehensive survey of commercial vineyards will provide GPS coordinates for each surveyed vineyard location.
 - ii. For each vineyard, the survey will record grape acreage by variety, soil type, proximity to bodies of water, extent of weed control, surrounding vegetation, presence of supplemental hosts for *Xylella fastidiosa*, and use of the insecticide imidicloprid.
 - iii. The survey will collect data on the presence of Pierce's disease and sharpshooters in each vineyard.
 - iv. Survey data will be entered into a geographic information system (GIS) to analyze spatial relationships of Pierce's disease and sharpshooters with geographical and environmental factors and viticultural practices.
 - b. Timeline
 - i. Survey was initiated in 2004 and will be completed by year 5 (2007).
 - ii. Data will be entered into GIS at the end of each growing season, enabling preliminary evaluation of data.
 - c. Technology transfer
 - i. Preliminary survey results will be reported at the annual Texas Pierce's Disease Research Symposium, other grower meetings, and on the Texas Pierce's Disease website.

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2. Development of a Geographic Information System (GIS).
 - a. Importance
 - i. The GIS will enable spatial analysis of the association of Pierce's disease and sharpshooters with geographical, environmental and viticultural factors in Texas.
 - ii. Spatial analysis may provide insights into the biology and epidemiology of Pierce's disease and the biology of sharpshooter vectors that may lead to improved management practices.
 - b. Timeline
 - i. The GIS work was initiated in 2004. Texas vineyard survey data will be entered into the system as it is collected (survey scheduled for completion in 2007) and existing geographic, climatic, soils, and other relevant data will be incorporated into the database through 2007.
 - ii. Spatial analyses of preliminary data can be conducted and reported on after each season.
 - iii. Additional data will be added to the GIS system for as long as the overall project is active and generating data.
 - c. Technology transfer
 - i. Preliminary survey results will be reported at the annual Texas Pierce's Disease Research Symposium, other grower meetings, and on the Texas Pierce's Disease website.
3. Development of a Texas Pierce's disease program website.
 - a. Importance
 - i. The website will be a major communications tool for reporting progress of the Texas PD program.
 - b. Timeline
 - i. The website was created in 2003 and revised in 2004. <http://piercesdisease.tamu.edu>
 - ii. Research reports, photos, and data will be posted as it becomes available.
 - c. Technology transfer
 - i. Website is live.

James S. Kamas

TCE – Fredericksburg - Horticultural Sciences

1. Genetic analysis of *Xylella fastidiosa* strains in Texas.
 - a. Importance
 - i. Determine genetic variability of *X. fastidiosa* in Texas and compare to diversity in California.
 - ii. Determine evolutionary relatedness of strains in Texas to strains in California.
 - iii. Diversity of strains within single vineyards.
 - iv. Identify strains in plant reservoirs as identical or different from those of infected vineyards.
 - v. To better understand the virulence and potential weaknesses of the genus *Xylella*, it is imperative that the genetic diversity in the pathogen's center of origin be explored. These findings may provide insight into development of bio-rational methods of pathogen management.

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- b. Timeline
 - i. Provide L. Morano with grape and other *X. fastidiosa* infected plant material from across Texas.
 - ii. Morano will grow strains and use genetic analysis with gyrase B gene (for sequencing) and SSR (small sequence repeats banding patterns). Technology and initial comparisons should be possible spring and summer of 2005. Already have Blake Bextine (UC Riverside) and Andy Walker (UC Davis) committed to help.
- c. Technology transfer
 - i. Poster or talk of initial strains at ASEV in June 2005.
 - ii. Present data (even if preliminary tree of *X. fastidiosa* relatedness) at next year's PD meeting in December.
 - iii. Completion will depend on how many strains that can be successfully extracted and cultured.
 - iv. Results will be relayed via semi-annual reports to USDA-APHIS.
2. Impact of hickory soils on survival of *Xylella* in grapevines.
 - a. Importance
 - i. Anomalous absence of Pierce's disease in the Hickory sands area may provide links to the Achilles heel of the vector. This unique area appears somehow to exclude the growth and reproduction of this pathogen in insect vectors, grape and other known plant hosts. Any insight may provide overall management tools for Pierce's disease of grape.
 - ii. Verifying or denying the influence of these soils on *X. fastidiosa* survival may provide important insight on site selection or mitigation of sites to improve grapevine survival.
 - iii. Growers considering grape production need to know specific soil/site considerations that may ameliorate virulence or pathogenicity of *X. fastidiosa*.
 - b. Timeline
 - i. Initial survival or impact should be completed within one year of inoculation.
 - ii. Preliminary findings may give rise to a second set of media studies which should be completed within two years
 - c. Technology transfer
 - i. Work will be reported in appropriate journals such as Plant Disease
 - ii. Results will be described in regular semi-annual reports and presented at extension educational events.
3. Texas statewide vineyard survey.
 - a. Importance
 - i. This work is important in order to better understand the spatial and geographic limitations of Pierce's disease in Texas.
 - ii. These data are needed to help justify the project to stakeholders and legislators.
 - iii. Mapping vineyards with and without Pierce's disease will help develop data necessary for quantification of the impact of the disease on the Texas grape industry and economy.
 - b. Timeline
 - i. Some vineyards have been surveyed and data dictionaries are being revised to accommodate current needs.

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- ii. Vineyards currently existing should be mapped by the end of 2006
 - c. Technology transfer
 - i. Work will be relayed at grower meetings, shared with legislators and included in semi-annual reports as it develops over time.
 - ii. Updated maps will be available for public viewing via the program's website.
 - 4. Pierce's Disease information transfer.
 - a. Importance
 - i. Generation of data without interpretation and presentation to growers is of little value.
 - ii. Understanding PD risk factors in choosing sites will reduce disease impact for the next generation of growers.
 - iii. Understanding cultural practice and chemical inputs that can mitigate disease risk has immediate value currently suffering losses from Pierce's disease.
 - b. Timeline
 - i. Real-time insect activity should be made available to growers via a password protected web-based information system. This information is critical to growers making the best decisions on a timely basis.
 - ii. Details on information transfer activities will be included in semi-annual reports.
 - iii. The creation of a decision-making matrix should be started in 2006
 - c. Technology transfer
 - i. As in past two years, extension education seminars will be conducted to relay the latest research findings to the grape growing community.
 - ii. As true over past few years, presentations on the progress of the Texas Pierce's disease project will be given to University Administrators, Local Community Development Groups, Rural County Judges' Association, the greater research community and at numerous grower educational functions throughout the year.

Isabelle Lauziere

TAES – Entomology - Fredericksburg

- 1. Monitor leafhopper populations in Central Texas vineyards.
 - a. Importance
 - i. We need to know the most common vectors *X. fastidiosa* in Texas, and their respective population dynamics (ends April 2006).
 - ii. This work is an essential precursor to recognizing the percentage of leafhoppers carrying *X. fastidiosa* and thus have the capacity to infect commercial vineyards with Pierce's disease.
 - iii. This work lays the foundation for eventual recommendations to Texas grape producers on which leafhoppers to monitor, and what management strategies are important for minimizing risks for moving *X. fastidiosa* from wild habitats to commercial grapes.
 - b. Timeline
 - i. Data collected in 2004 is being analyzed.
 - ii. This work is being continued in 2005, and results will be shared on a timely basis with cooperating producers.

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- iii. Information from cooperating vineyards should be posted on the Pierce's disease website for use by all.
 - c. Technology transfer
 - i. Initial observations were shared at Texas PD Symposium February 2005.
 - ii. Data will be presented at California PD Symposium December 2005.
 - iii. Understanding of the data will be shared with growers regarding crop management.
 - iv. Publish data in peer-reviewed journals.
 2. Characterize the frequency of presence of *Xylella fastidiosa* in xylem-feeding leafhoppers (collaborative work with Forrest Mitchell and Lisa Morano).
 - a. Importance
 - i. Insect counts alone do not provide adequate information for insect management decisions; must also know percentage of insects in Central Texas carrying the *Xylella* bacterium.
 - ii. This information is critical for explaining why certain vineyards are impacted by Pierce's disease while others have no known record of the disease.
 - iii. This information is critical for developing insect and disease management strategies and tactics.
 - b. Timeline
 - i. This is a new line of research to begin in April 2005, and results will depend on availability of good molecular technologies. Application of electrochemical detector (ECD) instrumentation for evaluating insects was positive, but needs more work. Anzenbio is interested to try to figure out if we want to pursue this line of work.
 - ii. Easy test for positive insects would be invaluable for all growers and scientists.
 - iii. Vineyards appropriate for this work are yet to be selected.
 - iv. Preliminary results will help confirm methodology for pursuit of an expanded insect monitoring effort.
 - c. Technology transfer
 - i. Periodical reports to Texas wine grape producers, and to California Pierce's disease program.
 - ii. Publish data.
 - iii. Submit data to Pierce's disease website.
 3. Survey and culture natural enemies of selected leafhoppers for biological control.
 - a. Importance
 - i. California is seeking to establish a complex of natural enemies.
 - ii. Natural enemies could be useful/important to eventual Texas PD vector management tactics.
 - b. Timeline
 - i. Several years of research on biological control will be necessary before considering this approach as part of a vector and disease management program.
 - c. Technology transfer
 - i. Periodical reports to Texas wine grape producers, and to California Pierce's disease program.
 - ii. Publish data.
 - iii. Submit data to Pierce's disease website.

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Forrest Mitchell

TAES – Stephenville - Entomology

Descriptive studies - These studies concern epidemiology of Pierce's Disease in Texas grape.

1. ELISA analysis of captured leafhoppers and comparison to trapping density across the state.
 - a. Importance: Correlating trapping density with ELISA values may yield insight into importance of the different vector insects to possible epidemics. In California, this is not possible due to the unnatural size of the *H. coagulata* populations.
 - b. Timeline: The majority of this research will proceed during the late spring and early summer, when the *H. coagulata* begin to appear. Other identified leafhoppers and cercopids will also be tested. We hope to test samples from a transect of vineyards extending east and west as well as north and south, coordinated with APHIS-Austin and Isabelle Lauzierre.
2. Determine presence and prevalence of *X. fastidiosa* in the Cercopidae-Clastopteridae (particularly *Clastoptera xanthocephala*) via ELISA
 - a. Importance: There is some indication that this group may be important in west Texas.
 - b. Timeline: Same as above for "a".
3. Develop lab technique for PCR detection of *Xylella fastidiosa* in leafhopper heads.
 - a. Importance: Although ELISA has proven useful, PCR is the more sensitive, as well as more difficult, technique for detection of low levels of bacterial DNA. Further, it may be discriminatory between *X. fastidiosa* strains.
 - b. Timeline: This will probably be initiated in the mid to late summer and concluded by the end of the field season.
4. Culture of *X. fastidiosa* from leafhopper heads, cross referenced with host plant
 - a. Importance: Culturing *X. fastidiosa* from the heads of leafhoppers provides a means of determining which strains are important to economic crops and whether strains detected in wild hosts match what are in the leafhoppers found in crops. *X. fastidiosa* may be transferred amongst some wild hosts by insects that do not infest crops and the importance of these hosts over inflated. Conversely, many of the wild hosts may support economically important strains of *X. fastidiosa*. Understanding the diversity of leafhoppers relative to the diversity of *X. fastidiosa* strains hosts other than grape will also be important to understanding disease dynamics, both in Texas and California.
 - b. Timeline: This will be an ongoing study. Samples from traps and ad hoc samples of suspected insects will be collected as time allows in order to determine if leafhoppers from hosts besides grape are contaminated with *X. fastidiosa*.

Experimental studies: These studies are oriented toward insect management in vineyards.

5. Determination of imidocloprid efficacy by caging *Homalodisca coagulata* on treated plants in the field. Measures are speed of kill and duration of efficacy on different parts of the plant.
6. Identify preferred feeding sites on grapevines and whether the feeding site can be predicted by stem diameter.
 - a. Importance: The objectives "5" and "6" are directed toward understanding management of *H. coagulata* in its native habitat. Management of native vectors of PD in California was by early season insecticide treatment. This may also work in Texas and may work again in California once the outbreak of this species

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- is over and it reaches levels comparable to Texas. Experimentation in Texas may provide target goals for reduction of the hoppers in California.
- b. Timeline: These studies will be initiated during the 2005 field season. The research will be conducted at the TAES-Stephenville vineyard.
7. Relate absolute density of *H. coagulata* to trap catches.
 - a. Importance: This objective will be important to understanding the meaning of the trap catches in both Texas and California.
 - b. Timeline: This will occur throughout the growing season.
 8. Determine if sunflower stands can be used as a trap crop or whether they simply increase leafhopper populations
 - a. Importance: The last objective simply plays a hunch. Populations of the sharpshooters may be manipulated, either up or down, by the presence of sunflowers. They may serve as reservoirs for the hoppers, useful in an experimental situation, or a sink for the hoppers, useful as a trap crop.
 - b. Timeline: The stand will be planted in the early spring and be observed season long.

Technology transfer for all objectives: This will be achieved mainly through TCE and APHIS efforts. Demonstrations will originate in the TAES research vineyard and result-demonstration tests can follow in grower vineyards once the value of the results is certain. The TAMU wine grapes website will also serve as a means of distributing information. Research manuscripts will be published in peer-reviewed journals and presentations made on results at professional meetings. One such was made in November at the Entomology Society of America national meetings in Salt Lake City.

Lisa D. Morano

University of Houston-Downtown - Biology and Microbiology - Houston

4. Genetic Analysis of *Xylella fastidiosa* Strains in Texas (TOP PRIORITY NEW AREA).
 - a. Importance
 - i. Determine genetic variability of *X. fastidiosa* in Texas and compare to diversity in California
 - ii. Determine evolutionary relatedness of strains in Texas to strains in California
 - iii. Diversity of strains within single vineyards.
 - b. Timeline
 - i. Identify strains in plant reservoirs as identical or different from those of infected vineyards.
 - ii. Collect strains from PD infected vines and wild plants via Jim Kamas and Mark Black.
 - iii. Grow strains and use genetic analysis with gyrase B gene (for sequencing) and SSR (small sequence repeats banding patterns). Technology and initial comparisons should be possible spring and summer of 2005. Already have Blake Bextine (UC Riverside) and Andy Walker (UC Davis) committed to help.
 - c. Technology transfer
 - i. Poster or talk of initial strains at ASEV in June 2005
 - ii. Present data (even if preliminary tree of *X. fastidiosa* relatedness) at next year's PD meeting in December.
 - iii. Completion will depend on how many strains I can get (Mark Black's lab is good at this), ability to keep all strains alive and how many total strains we want to do?

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5. Evaluation of American hybrid varieties and surrounding plant reservoirs in the PD Hot Zone (HAVE COMPLETED THIS).
 - a. Importance
 - i. Viticultural information about American hybrids important for those in area with high disease pressure (Texas, Florida, Missouri).
 - ii. Field evidence of American hybrids that are very resistant to PD suggests genetic mechanisms for further study.
 - iii. Evaluation of *X. fastidiosa* positive plant reservoirs in this area more likely to identify all potential native and ornamental sources of inoculum.
 - iv. Evaluation of sharpshooters in this warm southern region may be critical for understanding ecology and epidemiology of the disease.
 - b. Timeline
 - i. Have already completed an initial study of addressing activities above at Austin County Vineyards.
 - ii. Should we continue work at other vineyards in hot zone?
 - c. Technology transfer
 - i. Have just completed a manuscript of this work titled “An American hybrid vineyard in the Texas Gulf Coast: analysis within a Pierce’s disease hot zone.”
 - ii. Plan to submit to American Journal of Viticulture and Enology this week.