

United States Department of Agriculture

Animal and Plant Health Inspection Service

# Safeguarding Through Science

Center for Plant Health Science and Technology

2011 Accomplishments

## **Center for Plant Health Science and Technology**

#### Mission

The Center for Plant Health Science and Technology (CPHST) supports the regulatory decisions and operations of the Animal and Plant Health Inspection Service's (APHIS) Plant Protection and Quarantine (PPQ) program through methods development, scientific investigation, analyses, and technology.

#### **Strategic Goals**

- Enhance PPQ's efforts in pest detection and management
- · Provide timely scientific and technical support required for emergency response and management
- Enhance support for APHIS trade-related plant health issues
- Provide current, relevant scientific and technical information to PPQ decisionmakers
- Enhance PPQ's capacity to anticipate and respond to emerging scientific, technical, and regulatory issues through partnership

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Cover Photo: Asian gypsy moth (USDA/Forest Service, John H. Ghent). CPHST has worked with stakeholders to develop a geospatial model to predict areas that are at the highest risk for Asian gypsy moth introduction.

## Message from the Director

The Center for Plant Health Science and Technology (CPHST) provides scientific support for the regulatory decisions, policies, and operations of the Animal and Plant Health Inspection Service's (APHIS) Plant Protection and Quarantine (PPQ) program in order to safeguard U.S. agriculture and natural resources. CPHST is responsible for ensuring that PPQ has the information, tools, and technology to make the most scientifically valid regulatory and policy decisions possible. In addition, CPHST ensures that PPQ's operations have the most scientifically feasible and practical tools for pest exclusion, detection, and management. This 2011 CPHST Accomplishments Report is intended to offer an in-depth look at the status of our programs and the progress we have made toward the Center's long-term strategic goals.

Our scientists provide leadership and expertise in a wide range of fields, including pest risk assessments that support trade, commodity quarantine treatments, pest survey and detection methods, identification tools and molecular diagnostics, and integrated pest management.

Some highlights of significant CPHST efforts in 2011 include:

**CPHST Project Information Assistant.** In the fall of 2011, CPHST rolled out its new SharePointbased project tracking system called the CPHST Project Information Assistant (CPIA). CPIA replaces the previous Workbench and Adhoc systems and centralizes all CPHST project information into a single system that is accessible to anyone in PPQ. CPIA is used to enter new project requests, approve projects and budgets, and track project workplans, reports, and progress. This new system provides enhanced visibility for our entire portfolio of projects within CPHST and to our stakeholders.

Leadership Structure Review and Reorganization. In December 2010, CPHST began a process of future planning, which was designed to feed into the next CPHST Strategic Plan and to position us to address anticipated resource challenges. An initial workgroup identified priority initiatives that CPHST should address, including evaluating and refining the CPHST leadership structure. In the summer of 2011, a Leadership Structure Review Workgroup was assembled to conduct this evaluation and develop options for strengthening CPHST's leadership and organizational structure. As a first step, the workgroup conducted structured interviews with CPHST and other PPQ staff to gather input on the current structure and recommendations for improvement. The workgroup delivered its final report in September, which provided a summary of interview results and the workgroup's proposed options for CPHST's leadership structure. As a result of this work, CPHST is reorganizing its leadership structure in 2012 so that the Associate Director and six National Science Program Leader positions will be consolidated into four Associate Directors. The Associate Directors will have administrative responsibilities in addition to providing programmatic oversight for CPHST's portfolio of projects. The Associate Directors will play a key role in providing a focal point for communications and coordination with stakeholders, customers, and the scientific and regulatory community, as well as strategic, operational, and fiscal planning. Implementing CPIA and reorganizing the CPHST leadership structure should result in better communications and interactions between CPHST and its valued customers.

Farm Bill Section 10201 Projects. CPHST has continued to provide leadership and to leverage significant additional support through Farm Bill Section 10201, which provides funds to strengthen the Nation's infrastructure for pest detection and surveillance, identification, and threat mitigation, while working to safeguard the nursery production system. In fiscal year 2011, CPHST managed more than \$10.7 million to support 91 projects—conducted mainly through cooperators—in areas such as pest survey technology, identification tools, molecular diagnostics, citrus pest management, and nursery pest management.

**European Grapevine Moth (EGVM).** One of CPHST's major efforts over the past couple years has been to provide support to the EGVM program. EGVM is a key pest of vineyards in the Mediterranean basin that was first detected in California in 2009 after it had reached outbreak status and caused crop losses of nearly 100 percent in core areas of the infested regions. EGVM has regulatory significance for both internal and export markets, which depend on the timely movement of grapes within the State, across the United States, and to foreign trading partners. California wine, table grape, grape juice, and raisin production would be severely impacted if this pest was not controlled. Information from CPHST-initiated projects has provided benefits to all aspects of the EGVM cooperative program, including survey, management, and regulatory efforts. The program has minimized EGVM impacts within California, prevented spread to other areas of the country, and maintained U.S. growers' access to international markets. The effectiveness of this program has resulted in a thousand-fold reduction of EGVM in California (over 100,000 moths captured in 2010 versus about 100 moths in 2011 and about 80 moths in 2012) and reduced the damage and regulatory impact of this pest on grapes.

CPHST is recognized nationally and internationally for its leadership in scientific development to battle plant pests and diseases. We are proud to serve in this role and are issuing this annual report to provide an informative overview of CPHST's many accomplishments and projects.

Dr. Philip Berger, Director Center for Plant Health Science and Technology USDA-APHIS-PPQ

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## National Science Programs

# Agricultural Quarantine Inspection and Port Technology

### National Science Program Leader -Dr. Michael K. Hennessey

The Agricultural Quarantine Inspection and Port Technology (AQI&PT) program provides scientific support for agricultural inspection and commodity treatments to prevent the entry or spread of invasive quarantine pests. This program supports PPQ and the U.S. Department of Homeland Security (DHS) Customs and Border Protection (CBP) inspection and treatment programs. Requests for methods development are received from PPQ programs for port operations, trade, regional operations, and emergency pest programs, as well as CBP's agriculture inspection program. The AQI&PT program directly supports plant health officers and agriculture specialists stationed at 17 plant inspection stations and over 350 cargo ports throughout the United States, as well as numerous international ports that have preclearance programs.

Several CPHST laboratories work with this program, including the Treatment Quality Assurance Unit (TQAU), the Plant Epidemiology and Risk Analysis Laboratory (PERAL), and the Otis, Mission, Gulfport, Fort Collins labs, as well as the newly renovated AQI Laboratory in Miami. CPHST made several advances in commodity treatments in 2011 in the areas of sulfuryl fluoride fumigation, cold treatment, microwave and vacuum steam heat, leaf washes, and a cargo hold spray. These treatments supported exports, imports, and movement of domestic commodities. Farm Bill Section 10201 funding supported most of these projects. CPHST also developed a new risk-based sampling protocol for propagative plant imports at plant inspection stations. Additionally, the National Science Program Leader was active on the USDA Radiation Safety Committee, Quadrilateral Scientific Working Group (Australia, Canada, New Zealand, and the United States), Methyl Bromide Alternatives Outreach, Plant Inspection Station Working Group, and Ag-Screening Tools and Disposal and Decontamination Committees in DHS.

#### **Recent Accomplishments**

- Collaborated with industry to confirm a sulfuryl fluoride fumigation treatment for export logs (TQAU).
- Developed a risk-based sampling protocol for plant inspection stations to inspect propagative plant material (PERAL).

- Supervised renovation of the CPHST AQI Laboratory.
- Conducted a special project on fruit fly baggage inspection to determine which flights represent the highest risk for introduction (AQI).
- Conducted pilot field testing of portable gas chromatography to detect prohibited plant material at the Hawthorne Plant Inspection Station (AQI).
- Tested snail fumigation treatments and phytotoxicity of treatments to propagative material (Otis/AQI).
- Worked to develop postharvest fumigation treatments for *Brevipalpus* mites on fig, European grapevine moth (EGVM) on grape, and Oriental fruit fly on cherry (Otis/AQI).
- Confirmed a cold treatment for peach fruit fly in citrus (Otis).
- Neared completion of work to develop microwave heating of wood as an International Standards for Phytosanitary Measures 15-accepted treatment (Otis).
- Tested candidate knockdown sprays for Japanese beetle in cargo holds (Otis).
- Conducted proof-of-concept tests for vacuum steamheat treatments for large veneer logs for export (Otis).
- Collaborated with CBP on isotope analysis for determining the origin of mangos (Gulfport).
- Conducted proof-of-concept tests on alternatives to incineration to dispose of intercepted quarantine material (Fort Collins).
- Demonstrated an effective leaf wash to remove Asian citrus psyllid (ACP) from kaffir lime leaves in California (Mission).

## **Risk and Pathway Analysis**

#### National Science Program Leader - Dr. Ron Sequeira

The Risk and Pathway Analysis (RPA) program focuses on collecting and interpreting scientific evidence and technical information regarding plant pest risks. RPA products help APHIS design risk-based policies and regulations for import, export, and domestic programs. The work of the RPA staff is essential to identify and assess new pest threats, provide scientific support for regulatory updates and revisions, and help prioritize resources to maximize plant health safeguarding capabilities.

The RPA program is supported primarily by CPHST's Plant Epidemiology and Risk Analysis Laboratory in Raleigh, NC, with satellite staff in Hawaii and Bogotá, Colombia. Additionally, RPA coordinates with TQAU, other CPHST laboratories, other APHIS and USDA groups, academia, foreign counterparts, and stakeholders to develop scientific analyses that are essential to producing risk-based phytosanitary policies and procedures.

The foundation of RPA's analytical capacity is its diverse, high-caliber scientific staff—which includes entomologists, plant pathologists, botanists, ecologists, economists, and other specialists—along with its extensive collection of information on plant pests. RPA uses state-of-the-art tools and methodologies for pest risk assessment. These include sophisticated spatial technology systems that integrate weather, pest distribution, and other databases to analyze pest dynamics; identify pests of greatest concern; identify potential pathways for the introduction of harmful exotic pests; and predict the spread of exotic pests.

#### **Recent Accomplishments**

**Green Approaches for Risk Management.** Collaborated with the University of Florida to characterize the effectiveness of non-fumigant commodity treatment options against citrus and grape mites of regulatory significance. As part of a 3-year plan, CPHST will reexamine risk management for asparagus and grape imports.

**Climate Change.** Led the development of agency-level adaptation plans and guided strategic plan development.

**Information Management.** Began the development of new information management systems to support exports with a new module for the Global Pest and Disease Database. In addition, began to establish protocols for a developmental infrastructure for simulation and epidemiological modeling testing.

**Simulation Modeling.** Led the development of a strategy to adapt existing systems into the next generation of fore-casting models for APHIS. This includes interagency cooperation with the U.S. Geological Service and the Agricultural Research Service (ARS).

**Mentoring and Training.** Hosted and mentored foreign students and international visitors, including two doctoral students. This training builds international capacity in risk analysis and supports PPQ efforts to facilitate trade in agricultural commodities. Offshore Surveys/Emergency Programs. Led the coordination of overseas research and survey efforts to characterize best management practices in order to advance management systems for citrus greening disease or Huan-glongbing (HLB). CPHST also led the development of new pathway and risk analyses for citrus black spot (CBS) disease, including coordination of risk analyses efforts with ARS. In addition, CPHST developed spread models and systems approaches to support the domestic HLB survey and control programs.

**Technical Support for Exports.** Provided support to strawberry producers by developing frameworks to evaluate systems approaches to ensure that the light brown apple moth (LBAM) is not present in commodities for export. Launched a pathway analysis to support exports of logs from the Pacific Northwest to China.

## Domestic Survey, Detection, and Identification

#### National Science Program Leader - Vacant

The Domestic Survey, Detection, and Identification (SDI) program provides scientific and technical support to PPQ for rapid detection, identification, and mitigation of plant pests. SDI consists of three cross-functional groups: early detection, identification, and spatial technology. CPHST scientists work as teams to address the scientific and technical needs of PPQ leadership and operations. Additional support is leveraged through cooperation with academic institutions, industry, and other government agencies.

The early detection team supports pest surveillance through the PPQ Cooperative Agricultural Pest Survey (CAPS) program. Support includes pest prioritization, pest datasheets and survey guidelines, approved methods for the survey and identification of CAPS targets, and trap and lure development. Each year, CPHST develops an updated, prioritized list of pests for the CAPS program. This list consists of exotic, high-impact pests that threaten U.S. agriculture and the environment. In addition, CPHST works with the CAPS program to develop pest lists for commodities and taxon groups recognized as national CAPS targets. Surveys focused on specific crops or types of pests provide an efficient method for detecting a suite of exotic pests at the same time. CPHST provides survey and identification support for pests from the prioritized pest list and national surveys. The Identification Technology Program (ITP) develops digital and Web-based products for detecting and identifying plant arthropods, molluscs, weeds, and diseases. ITP aims to constantly be attuned to its clients' needs, to pursue new avenues and advanced technologies for delivery of products, and to offer its services in multiple ways to better serve PPQ and its cooperators. In 2011, ITP rose to new heights in identification tool production in its ID Tool project area. The ITP team delivered 16 digital identification tools and resources to PPQ and cooperators and continued development of 14 digital tools and resources during the year. ITP officially launched ID Source, a resource containing links to over 1,500 identification-themed Web sites. In addition, ITP has made significant accomplishments in the ID Image and ID Mobile project areas. Looking to 2012 and beyond, ITP plans to make a big push toward sharing its data with other organizations' Web-based tools and greater and stronger mobile delivery of products.

The spatial technologies team integrates data collection, management, analysis, statistics, and modeling to support the broad needs of PPQ. With members located in CPHST headquarters and laboratories across the country, the team contributes to a wide range of programs including pest surveillance, monitoring, and control, as well as supporting emergency programs, global risk assessment, and safeguarding operations. In 2011, the team contributed to numerous plant health programs. The team has continued work on the ACP areawide monitoring study for the Lower Rio Grande Valley in Texas in an effort to better understand how ACP dynamics contribute to the spread of citrus greening disease. In addition, the team supported department-level directives on climate change, supported CPHST scientists using the global positioning system for invasive weed monitoring, and further contributed to analyzing insect population dynamics for grasshoppers in the western United States.

#### **Recent Accomplishments**

#### Early Detection Team Support for CAPS

- Completed the Stone Fruit Commodity-based Survey Reference and Guidelines.
- Enhanced the Exotic Wood Borer/Bark Beetle National Survey Guidelines with new pest maps, updated survey methods, and verified host and distribution references.
- Developed 18 pest datasheets for pests on the CAPS Prioritized Pest List.
- Developed approved survey and diagnostic methods for 22 new CAPS pests.

- Developed a visual survey protocol for cotton seed bug.
- Collaborated with other labs on the development of improved traps.
- Provided technical guidance on survey methods to CAPS field staff.
- Provided technical pest and survey information for the Integrated Plant Health Information System.

### Identification Technology Program Releases

- A Resource for Pests and Diseases of Cultivated Palms: Identifying Commonly Cultivated Palms, http://itp.lucidcentral.org/id/palms/palm-id/
- A Resource for Pests and Diseases of Cultivated Palms: Symptoms of Diseases and Disorders, http://itp.lucidcentral.org/id/palms/symptoms/
- Pink Bollworm and its Look-Alikes, http://itp.lucidcentral.org/id/pbw/
- Xyleborini Ambrosia Beetles: An Identification Tool to the World Genera, http://itp.lucidcentral.org/id/wbb/xyleborini/
- Ironclad ID: Diagnosing Ironclad and Cylindrical Bark Beetles (Coleoptera: Zopheridae) of North America north of Mexico, http://itp.lucidcentral.org/id/wbb/ironcladid/
- Bark Beetle Genera of the United States, http://itp.lucidcentral.org/id/wbb/bbgna
- Oncid ID: Tool for Diagnosing Adult Twig Girdlers, http://itp.lucidcentral.org/id/wbb/oncidid/
- A Resource for Wood Boring Beetles of the World, http://wbbresource.org/
- A Resource for Pests and Diseases of Cultivated Palms: Screening Aid to Pests, http://itp.lucidcentral.org/id/palms/SAP/
- Citrus Resource, http://idtools.org/id/citrus/resource/
- Citrus ID: Hosts and Potential Hosts for Citrus Pests and Diseases in the United States, http://itp.lucidcentral.org/id/citrus/citrusid
- Citrus ID, Edition 2,
   http://idtools.org/citrus/citrusid/
- ID Source, http://idsource.colostate.edu/
- Citrus Diseases, http://www.idtools.org/id/citrus/diseases
- Common Nymphal Grasshoppers of the Western United States, http://itp.lucidcentral.org/id/grasshopper/nymphal/
- Dried Botanical ID, http://idtools.org/id/dried\_botanical

- Identification Tool to Weed Disseminules of California Central Valley Table Grape Production Areas, http://itp.lucidcentral.org/id/table-grape/
- A Resource for California Central Valley Table Grapes, http://itp.lucidcentral.org/id/table-grape/resource
- A Resource for Pests and Diseases of Cultivated Palms, http://itp.lucidcentral.org/id/palms/resource/
- Terrestrial Mollusc Tool, http://idtools.org/id/mollusc/

## Spatial Technologies Team Products

- Developed a prototype database of barberry eradication historical records from the Pacific Northwest.
- Used geospatial technology to analyze ACP populations and develop an efficient trapping grid.
- Developed models to investigate causes of ironwood tree decline in Guam.
- Updated HOPPER software to assist land managers with management decisions for grasshopper control.

## Response and Recovery Systems Technology - Arthropods

## National Science Program Leader - Mike Stefan

The Response and Recovery Systems Technology (RRST) program for arthropod pests provides timely scientific support to regulatory program managers and decisionmakers for high-consequence plant pests. The program emphasizes science-based preventive measures to reduce or inhibit further spread and develops rapid response and recovery technologies geared towards eradicating or containing targeted pests of concern. RRST supports a wide range of programs and oversees the CPHST LBAM Unit and the Fruit Fly Unit. These programs utilize the tools of current integrated pest management strategies, phytosanitary measures, regulatory treatments, and related technologies to mitigate the effects of arthropod pests while minimizing adverse effects on the environment, producers, and consumers.

## **Recent Accomplishments**

## European Grapevine Moth, Lobesia botrana

• A CPHST-led technical working group (TWG) has provided information and feedback on trapping, lures, mating disruption, treatment, hosts, risks associated with commodities, and treatment of regulated articles. These recommendations have largely been adopted by the program. The EGVM program has resulted in a decline in captures (from 100,000 moths in 2010 to less than 100 moths in 2011) as well as deregulation of previously infested portions of 3 counties.

• The Otis Lab has developed rearing systems for EGVM that have supported pesticide testing and commodity treatment development. The lab has also made significant progress in developing mass-rearing methodology in the event that it is needed in the future. In addition, the lab has conducted tests for alternative mating disruption formulations and to use in developing a monitoring tool that can be utilized when an area has been treated with a mating disruption formulation.

## Light Brown Apple Moth (LBAM), *Epiphyas postvittana*

- The LBAM Unit has developed LBAM mass-rearing and collection methods, allowing for the production of more than 1,000 moths per tray and up to 500,000 moths per week in a small, pilot-production facility. The unit also released over 600,000 sterile LBAM in a sterile insect technique (SIT) pilot project in Long Beach, CA, to evaluate the potential for use of SIT to suppress LBAM populations.
- The LBAM Unit and Otis Lab have continued to support development of regulatory treatments to allow movement of commodities from regulated areas. This effort includes cold treatment validation for control of LBAM for movement ofstrawberry plants for planting, as well as testing of methyl bromide and alternative fumigants for LBAM post-harvest control for several commodities including apples, stonefruits, and caneberries.
- CPHST supported research for a mating-disruption treatment for production. The Otis Lab participated in the research that led to caneberry growers' adoption of this technology. Additional testing of novel matingdisruption formulations is nearly complete and should provide the growers with alternatives for treatment.
- CPHST tested a new four-component pheromone lure under California conditions and determined that the new lure would significantly improve trap catch over the current two-component lure.

## Fruit Fly

 During 2011, the Fruit Fly Unit has integrated into the APHIS cooperative fruit fly programs and concentrated on technical and methods support for the field during emergency response quarantine and eradication activities. These efforts include support to improve fruit fly rearing and emergence techniques, aerial sterile fruit fly releases, release equipment calibration, ground pesticide treatments and operational techniques, enhanced detection, modeling of pest colonization and dispersal, and quality assurance of program resources such as lures, diets, and control pesticides.

#### Emerald Ash Borer (EAB), Agrilus planipennis

- The Otis Lab continued to support research on improving traps, lures, control, and management of EAB.
- CPHST has worked in conjunction with USDA's Forest Service and ARS to successfully release several parasites that attack EAB. The Otis Lab continues to monitor the establishment and effectiveness of these parasites at field-release sites and to consult with a PPQ parasite rearing facility in Michigan that was developed based on this work. An additional parasite species collected from Russia is now under evaluation for field release. Field releases of parasites are now being evaluated in almost all States where EAB is known to be established.

## Asian Longhorned Beetle (ALB), Anoplophora glabripennis

- The Otis Lab continues to provide technical support to the ALB program through testing of regulatory treatments, control treatments, and discerning infestation dynamics. The lab continued assessing ALB populations and tree-damage patterns to understand the dynamics and spread of ALB infestations and coordinated research on attractant lures and trapping research with university cooperators and ARS.
- CPHST also conducted pilot tests with the PPQ National Detector Dog Training Center to test the possibility of using detector dogs in ALB surveys. A large study was conducted in Ohio and Massachusetts to compare the efficacy and cost effectiveness of visual survey, traps, and sniffer dogs. Efforts continue to identify additional attractants for ALB adults.

#### Sirex Woodwasp (Sirex noctilio) Biological Control

- CPHST successfully identified a male-produced pheromone from *Sirex notilio* and identified an improved trap design. Additional work continues to determine how to utilize the pheromone for survey or control.
- The Otis Lab conducted controlled release experiments of the nematode biocontrol agent *Beddingia siricidicola* and investigated its native strain.

• In collaboration with the Forest Service, CPHST continued to develop survey tools and evaluate the effectiveness of the biological control agent.

## Emergency Response and Response and Recovery Systems Technology - Plant Pathogen and Weeds

#### National Science Program Leader - Dr. Russ Bulluck

The Emergency Response program's role is to provide scientific support to PPQ during a plant health emergency. The program provides scientifically valid and operationally practical information on the biology of the plant pest, survey techniques, and pest management. During the initial plant health emergency response, a TWG of scientific experts may also be assembled.

The Emergency Response Program Leader is responsible for facilitating and coordinating the interactions of the appropriate response and recovery programs and working in concert with these program leaders to ensure a continuum of scientific support during transitions from emergency response to emergency recovery. During the past year, CPHST provided scientific input for 22 plant health emergencies, including emergencies for red palm weevil, South American palm weevil, annual ryegrass seed gall nematode, Mexican fruit fly (Mexfly), boxwood blight, red ring nematode of palms, and several other pests.

In addition, CPHST prepares New Pest Response Guidelines (NPRG) for new and emerging pests in coordination with PPQ Emergency and Domestic Program (EDP) staff. The Emergency Response program also represents PPQ in the Biological Indications and Warning Analytic Community, which is composed of Government and academic labs committed to improving biosurveillance in the United States.

#### **Recent Accomplishments**

National Ornamental Research Site at Dominican University of California (NORS-DUC). The NORS-DUC is a secure research site that simulates a nursery environment and is used to perform pest and disease studies on nursery stock with high-level containment safeguards. There are currently five research projects underway, with an additional three to four projects in the planning stage.

**NPRG Scientific Team.** The NPRG provide scientific information on the biology of specific pests and an action plan for new plant health emergencies, including control methods, survey methods, ecology of the pest, and identification methods. In addition, environmental and regulatory components are supplied by the EDP staff. These documents are most useful prior to a plant health emergency involving a pest, although in some cases the NPRG are completed after a pest is detected. Through additional funding provided by the Farm Bill, four post-doctoral fellows were hired specifically to develop NPRG. To date, 14 NPRG have been completed or are underway.

Potato Cyst Nematode Programs. CPHST continues to provide scientific support for the Golden Nematode Program in New York and the Pale Cyst Nematode Program in Idaho. CPHST provides scientific information related to the biology and control of these nematodes, as well as information regarding sampling strategies and protocols. CPHST also provided technical input for the 5-year review of the Pale Cyst Nematode Program.

**Management of Technical Working Groups.** Often when new plant health pests and pathogens occur within the United States, very little information is readily available about the organism. This is due, in large part, to the fact that the pests and pathogens are almost always exotic and any scientific expertise resides offshore. CPHST assembles TWGs to obtain the scientific information needed on pest issues. TWGs are composed of scientific experts from around the world who may meet in person or use video or teleconferencing to discuss issues regarding a specific pest. During the past year, CPHST managed six TWGs. Past TWGs were focused on citrus health, *Phytophthora ramorum*, golden nematode, pale cyst nematode, *Plum pox virus*, boxwood blight, red palm weevil, fruit flies, white flies, ALB, EAB, and various other pests.

#### Planning and Preparing for Plant Health Emergencies.

One of the major functions of the Emergency Response national science program is to help the agency prepare for plant health emergencies. The program is involved in the planning and preparation of plant health emergency exercises and participates in the exercises. These exercises range from phone call and email chains and tabletop exercises to full-scale Incident Command System exercises and Area Command exercises. In 2011, the program helped to plan and facilitate eight separate full-scale and tabletop exercises.

## Response and Recovery Systems Technology - Citrus Health

## National Science Program Leader -Dr. Charla Hollingsworth

The RRST-Citrus Health program is responsible for methods development work in support of PPQ operations and policy units to prevent the introduction and establishment of citrus pathogens of regulatory concern in the United States. The program is currently addressing issues arising from established plant diseases such as HLB, citrus canker, sweet orange scab (SOS), and CBS, as well as plant diseases not known to occur in the United States such as citrus variegated chlorosis (CVC) and Citrus leprosis virus (CiLV). The program emphasizes an integrated, multifaceted approach for controlling plant disease through discovery and implementation of effective best management practices and diagnostic tools. Working cooperatively with others, the program supports the development of applied, basic, and technological solutions to accomplish effective response and recovery while preserving the integrity of the production environment.

One of the program's primary missions is to work closely with the Citrus Health Response Program (CHRP) to preserve citrus health in the United States. This is accomplished through interactions with researchers, technical experts, international cooperators, regulatory program managers, stakeholders, and others. Two principal mechanisms are utilized in these interactions: advising the subject matter expert group and methods development support. In 2011, the program continued its leadership role in the National CHRP Science and Technology Research Coordination Group. In this role, the program coordinates citrus research efforts nationally and internationally and organizes an annual meeting that brings together researchers, technical experts, and regulatory programs from across the United States. This effort supports open communication and research coordination across the citrus research community to benefit scientific inquiry and inform stakeholders.

#### **Recent Accomplishments**

 Planned and organized the 2nd Annual Citrus Health Research Forum held October 4–6, 2011, in Denver, CO. The forum brought together researchers, technical experts, regulatory programs, and industry leaders to discuss research accomplishments from the past year and to develop a coordinated action plan for the forthcoming year. A primary outcome from the meeting was the planning and organizational structure needed to develop a multidisciplinary research proposal to submit to the National Institute for Food and Agriculture.

Co-chaired the CPHST Leadership Structure Review
Group. This group interviewed PPQ and National Plant
Board stakeholders to understand their perspectives
regarding CPHST's current leadership structure and
proposed structure options to the CPHST Director for
consideration.

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Reviewed 10 citrus-related cooperative projects for 10201 Farm Bill funding and edited CPHST internal projects for Farm Bill submission.

## Laboratories and Units

## **CPHST Lab, Beltsville, MD**

### Laboratory Director - Dr. Laurene Levy

The Beltsville Laboratory's mission is to develop, validate, and implement advanced biochemical and molecular methods for the detection of high-consequence plant pathogens, including APHIS select agents and plant pathogens in foreign germplasm. Laboratory programs utilize cutting-edge technologies from the fields of plant pathology, molecular biology, and clinical diagnostics to develop, adapt, and improve methods for accurate and rapid diagnosis of plant pathogens. The laboratory's scientists validate plant pathogen diagnostic methods prior to stakeholder release to assure their performance and fit for purpose in regulatory programs.

The Beltsville Lab strives to achieve timely transfer of diagnostic tools that are field deployable for PPQ emergency response and eradication programs. Tools are deployed to stakeholders through clearly written standard operating procedures and hands-on laboratory training for end users within and outside of PPQ. The laboratory also utilizes new diagnostic methods to accurately and rapidly diagnose and differentiate high-consequence and select agent plant pathogens, or pathogens that require Federal confirmation. In these situations, the laboratory conducts sample testing in parallel with the PPQ Molecular Diagnostics Laboratory. The Beltsville Lab is enhancing its forensics capabilities to understand where pathogens originate or are emerging. This effort will support mitigation efforts by aiding in the determination of pathogen incursion pathways.

#### **Recent Accomplishments**

- Developed and adapted polymerase chain reaction (PCR) assays for targeted pathogens for the Germplasm program, including assays for phytoplasmas, *Sweet potato chlorotic stunt virus* (East African and West African groups), *Potato leafroll luteovirus*, and *Nepovirus* subgroup A. Wrote 12 work instructions describing conventional and real-time PCR assays, DNA extraction, and RNA extraction protocols.
- Developed assays for molecular detection and identification of *Citrus leprosis virus-C* (CiLV-C). Wrote four work instructions for conventional and real-time PCR assays and RNA extraction protocol. Validated the three detection protocols using samples from Costa Rica and Panama.
- · Validated molecular diagnostics and developed markers

for forensic studies of the citrus black spot (CBS) fungal pathogen *Guignardia citricarpa*.

- Developed molecular diagnostics methods and conducted pathogenicity testing for the sweet orange scab (SOS) fungal pathogen *Elsinoë australis*.
- Developed and validated PCR assays for rapid and reliable differentiation of *Xylella fastidiosa* strains that cause citrus variegated chlorosis (CVC).
- Completed molecular and serological characterization of *Plum pax virus* strain W (PPV-W) isolate UKR 44189. Analysis of the nucleic amino acid sequence showed more than 99-percent identity with a PPV isolate found in Latvia. High-sequence identity and epidemiological data suggest a possible common origin for PPV-W isolates found so far.
- Evaluated Cellular Analysis and Notification of Antigen Risk and Yield (CANARY), which is a cell-based technology capable of rapidly identifying low levels of plant pathogens. Discovered that a non-specific reaction occurred between individual B cell lines and nontarget pathogens, but that this reaction could be blocked by adding phosphocholine.
- Evaluated three *Xylella* B cell clones for CANARY detection of the *Xylella fastidiosa* CVC strain in infected citrus material.
- Evaluated nine *Phytophthora* specific CANARY B cell lines using *P. ramorum* pure cultures, selected the most sensitive line, and then determined its shelf life. Selected optimized field-deployable and central laboratory sample preparation platforms. Used the optimized central laboratory *Phytophthora* CANARY system to test fieldcollected and laboratory-infected *Phytophthora* samples. As expected, all field-collected and laboratory-infected subsamples tested positive for *Phytophthora*. The *Phytophthora* CANARY assay is very promising and will be further optimized and adapted for use in *Phytophthora* diagnostics.
- Developed domestic and international shipping protocols for CANARY B cells and demonstrated that the cells maintained viability and detection sensitivity for at least 120 hours.
- Evaluated the MIT Lincoln Lab Nucleic-Acid Kit (LiNK) DNA extraction cartridge against a common commercial DNA extraction kit using five economically significant plant pathogens and two sample types. Preliminary results showed that DNA extracted using the LiNK was of higher quantity but lower purity than DNA extracted using the commercial kit. The LiNK system is very promising and will be further optimized and adapted for use in plant pathogen diagnostics.

- Developed proficiency test panels for the PPQ National Plant Protection Laboratory Accreditation Program (NPPLAP). Certified 30 diagnosticians from 18 laboratories for the *P. ramorum* program, and certified 8 diagnosticians from 3 laboratories for the PPV program. Developed and validated the HLB panel.
- Completed diagnoses for 7 different plant pathogenic organisms, which included about 250 total samples.
   Provided 7 training courses that provided hands-on experience for 29 scientists and diagnosticians from PPQ, the National Plant Diagnostic Network, and universities.

## **CPHST Lab, Fort Collins, CO**

### Laboratory Director - Dr. Richard Zink

Work at the Fort Collins Laboratory focuses on five critical areas of methods development for APHIS: pest identification technologies, pest survey protocols and guidelines, risk mapping, agricultural quarantine and inspection (AQI) waste disposal and decontamination, and biological control. The staff at Fort Collins develop and transfer scientifically based methods, innovative tools, and state-of-the-art technologies to APHIS and other Federal and State agencies to reduce the risk associated with potential, new, and established problem species and plants for planting. The laboratory provides extensive scientific support for the Cooperative Agricultural Pest Survey (CAPS) program. The laboratory transfers methodologies and tools to field operations to ensure efficient and effective survey, detection, identification, emergency response, and eradication efforts. As a primary source for the latest technologies, the Fort Collins Laboratory develops electronic, matrix-based identification resources to help support rapid, consistent, and accurate identification and nomenclature of pest species.

The Fort Collins Laboratory delivers highly innovative and cost-effective methods for the management of invasive plants on public lands through the utilization of insect biological control agents as well as chemical and cultural control procedures. PPQ operational programs receive spatial technology support from the laboratory to guide them in the application of new geospatial survey and detection methods. At a satellite location in Albany, CA, efforts continue to improve upon artificial diets for rearing insects for biological control of weeds and for use in eradication programs such as pink bollworm. Albany also develops protocols for biochemical analysis and identification of wild and artificially reared insects.

#### **Recent Accomplishments**

#### Identification Technology Program

• Delivered a record 14 digital identification tools in 2011 and managed 32 identification projects in cooperation with 19 institutions. Launched use of image-based identification and conducted initial exploration of smartphone and tablet PC apps for mobile delivery of products.

## Molecular Diagnostic Technology for Invasive or Regulated Plants

 Developed molecular diagnostic tools, including rapid automated molecular identification tools, for several problematic or federally listed species: Avena sterilis; Commelina benghalensis; Heracleum mantegazzianum; Imperata cylindrica; Mikania spp.; the red rice species Oryza longistaminata, O. punctata, and O. rufipogon; Saccharum spontaneum; Oplismenus hirtellus ssp. undulatifolius; and the Rutaceae genera Murraya, Zanthoxylum, and Citrus.

## Methods Development for Disposal of Agricultural Waste at Ports-of-Entry

• Tested a small-scale electrical incineration system and three large waste disposal machines currently in use by hospitals, research labs, and municipal waste facilities, and found that such machines could be more costeffective alternatives for disposal of regulated international waste at U.S.ports-of-entry.

#### Rearing the Russian Knapweed Gall Midge

• Established a greenhouse-based rearing program for a weed biocontrol agent, the Russian knapweed gall midge (*Jaapiella ivannikovi*): over 3,400 galls were produced, about one-quarter (850) of which were successfully distributed for field releases in several western States.

#### Support of CAPS

 Provided scientific foundations for the CAPS program via product and tool development; visual survey protocol development; survey manual and pest datasheet updates; evaluation of alternative insect trap adhesives and pheromone lures; and daily support for the Integrated Plant Health Information System and for CAPS field staff.

## Observation Sampling Analysis To Support an Areawide Monitoring Study

 Utilized geospatial interpolation, zonal statistical analysis, and time-series data visualization to evaluate the impact of various trap-spacing grids on population observations of ACP, a vector of citrus greening disease (Huanglongbing). Determination of optimal trap spacing will help managers to balance collection of vital pest population information and orchard protection with available fiscal resources. This effort supports an existing areawide ACP monitoring study under the CPHST Mission Lab.

#### Chemical and Cultural Control of Invasive Weeds

Tested and evaluated the efficacy of several traditional and non-traditional weed control methods in field trials against leafy spurge, field bindweed, cogongrass, giant hogweed, and Benghal dayflower; the latter two trials evaluated control of the soil seedbank as well as mature plants.

## **CPHST Lab, Gulfport, MS**

#### Laboratory Director - Anne-Marie Callcott

In 2011, the CPHST Gulfport Laboratory continued to support PPQ programs through the analytical chemistry section and the imported fire ant (IFA) section. The chemistry section primarily processes APHIS-wide pesticide treatment program samples and provides technical support in the form of methods development to address changing program needs. In addition to routine pesticide residue analysis work, the chemistry methods development staff continued to shift resources from routine methods adaptation work to more specialized work supporting the development and verification of lures used by PPQ programs and CPHST scientists conducting projects. The IFA section develops methods and tools for the survey, detection, regulation, and chemical and biological control of the IFA. PPQ, State plant regulatory officials, the nursery industry, chemical industry, farmers, homeowners, and other stakeholders utilize technology developed by the IFA section. The lab's primary focus is on developing quarantine treatment options for growers who move nursery stock and other regulated articles outside the Federal quarantine area, with the current emphasis on grass sod and field-grown nursery stock treatments. The lab also supports the rearing and distribution of phorid flies-an IFA biological control agent-to States.

In March 2011, PPQ management announced the anticipated closure of the Gulfport facility. Consequently, 2011 was the final year of operational work out of the Gulfport facility. Much of 2011 was spent preparing for the facility closure in 2012, including planning for relocation of operations and staff and the outsourcing of other operational work.

#### **Recent Accomplishments**

- Analyzed 721 routine APHIS program-related samples, including environmental monitoring samples and lures, in addition to 173 associated quality control samples. There was significantly less demand this year for analysis of environmental monitoring samples, most notably from the Grasshopper program, which conducted fewer than normal treatments due to lower than anticipated pest populations.
- Redirected all analytical chemistry project work to the AQI Lab in 2011; accordingly, such projects will be reported through that laboratory.
- In support of the ALB program, conducted inhouse method adaptations to program analytical methods in order to improve in-matrix recoveries.
- Through a cooperative agreement with the University of South Alabama, developed an extraction, isolation, and analytical process for the determination of a multicomponent brown spruce longhorn beetle lure, as well as an extraction, concentration, and analytical process for a European spruce bark beetle lure. In addition, the university conducted lure quality control sample analysis and synthesis of the trimedlure.
- Continued an interagency agreement with the DHS-CBP Savannah Lab to develop a model to determine the origins of mangoes by isotope analysis. Collected second season samples of mangoes in Florida and Puerto Rico, as well as conducted Dominican Republic sampling. Conducted isotope analysis on the samples to generate profiles for each location. The second season sampling was evaluated to ensure that agricultural and environmental influences did not adversely affect the model's ability to distinctly identify a known growing region. The statistical model was expanded to include the Dominican Republic growing region and evaluate whether Dominican Republic-grown mango is distinct from sources already in the model.

- Continued the APHIS-funded phorid fly (*Pseudacteon* spp.) rearing and release programto support IFA control efforts in 2011, with multiple releases of a third fly species, *P. obtusus*, and the first releases of a fourth species, *P. cultellatus*. A publication with CPHST co-authors on the establishment and spread success of the first two species released, *P. tricuspis* and *P. curvatus*, was published in the Journal of Insect Science. Both species are now established in more than 50 percent of the IFA quarantined area.
- Label changes on Onyx Pro® Insecticide (bifenthrin) were completed in 2011 to include an application rate effective on IFA in grass sod as a quarantine treatment. When a new environmental assessment for IFA is completed, this treatment will be added to the Treatment Manual. This will provide growers with a treatment that does not include chlorpyrifos, which is difficult to find due to growing EPA restrictions on its use.
- Initiated development in 2011 of a cold treatment for IFA in bulk soil with a focus on contaminated soils destined for burial. The lab demonstrated successful cold treatment in small containers in a lab setting. The treatment results were verified in tests conducted in fullsized refrigerated containers through a cooperative agreement with the University of Tennessee.

#### Laboratory Closing Highlights:

- APHIS-PPQ will be closing the Gulfport Facility in mid-2012.
- Existing PPQ State staff and services will remain in the local commuting area.
- Existing CPHST staff and services will be outsourced or relocated to other facilities.
- CPHST staff to support outsourcing of routine residue analysis work and IFA work will be moving to offices in Biloxi, MS.
- Analytical chemistry changes:
  - Routine pesticide residue analysis of environmental monitoring samples conducted to support routine PPQ programs will be outsourced to USDA-Agricultural Marketing Service's (AMS) National Science Laboratory in Gastonia, NC, at a prenegotiated per sample cost. CPHST staff will oversee and coordinate the program, act as a liaison with PPQ staff, and provide quality assurance reviews and audits. AMS or the CPHST AQI Lab will handle emergency pesticide residue analysis of environmental monitoring samples on a case-by-case basis.

- Project work and staff to support CPHST and PPQ analytical chemistry needs will be relocated to the CPHST AQI Lab.
- CPHST will no longer provide analytical support for IFA soil samples. States may use a State pesticide lab or enter into an agreement with AMS to conduct the analyses
- IFA changes:
  - All methods development work will be outsourced through cooperative and interagency agreements managed by CPHST.

## **CPHST AQI Lab, Miami, FL**

### Acting Laboratory Director – Michael Hennessey

The CPHST AQI Laboratory is co-located with the Agricultural Research Service in Miami at the Subtropical Horticulture Research Station. While the laboratory was being renovated, two CPHST personnel were housed with ARS scientists at the site. These two personnel are members of the Biological Control Unit (BCU) but also have some responsibilities to conduct inspection methods development. These scientists provided technical oversight and expertise to help both DHS-CBP and PPQ port inspectors improve the efficiency and efficacy of domestic inspection and commodity treatment. Specifically, the scientists helped design, conduct, and analyze methods to test rapid screening tools and verify fumigation treatments. The scientists also served as laboratory representatives on the regional CBP and PPQ South Florida Biological Threat Advisory Group, where they helped to design special operations projects, including one on passenger baggage inspection.

#### **Recent Accomplishments**

#### Pilot Field Testing of Portable Gas Chromatography

- Developed a protocol at the Hawthorne Plant Inspection Station using zNose chromatography to distinguish prohibited citrus bonsai species from non-prohibited species.
- Developed and refined methods to distinguish prohibited species of bonsai by testing 14 citrus and non-citrus species.
- Wrote standard operations protocol reviewed by program managers.
- Trained Hawthorne Plant Inspection Station safeguarding officers and identifiers on how to operate the zNose and sample bonsai trees.

## Snail Fumigation and Phytotoxicity of Treatments to Selected Propagative Material

• Evaluated the efficacy of methyl bromide and sulfuryl fluoride fumigation schedules on *Succinea* spp. and the phytotoxicity of the treatments to propagative material on which the snail is commonly intercepted.

### Fruit Fly Baggage Inspection Special Project

- Cooperated with PPQ Eastern Region staff, Fort Lauderdale airport PPQ identifiers, and CBP staff to design a study to determine which flights (airline, country, and port of lading) represent the highest pest risk based on fruit fly (Tephritidae) host material seizures and Tephritidae interceptions entering through passenger baggage. Presented the results to the Florida Agriculture Safeguarding Partnership Council.
- The results of the project helped determine if baggage from countries known to be infested with Tephritidae pose a high risk of pest introduction and if the destination zip code of the baggage interceptions could be used to predict (and possibly trace back) exotic fruit fly outbreaks in Florida.

## **CPHST Lab, Mission, TX**

#### Laboratory Director - Dr. Matt Ciomperlik

The Mission Laboratory supports PPQ's programs by developing pest detection and management methods, mitigation strategies, and molecular diagnostic tools for invertebrate pests. The laboratory identifies, develops, and provides technology transfer for a wide range of scientific methods to PPQ and State departments of agriculture. In addition to these core functions, the laboratory cooperates with stakeholders and researchers in academia and USDA to provide expertise to PPQ regarding epidemiology of plant diseases, remote sensing/geographic information systems, biological control, areawide pest management, and sterile insect technology (SIT) support for the Mexfly eradication program.

In order to provide scientific expertise to PPQ regarding technological advances in molecular biology, the Mission facility has developed advanced strengths in the evaluation and application of DNA technologies and bioinformatics tools. This has enabled the adoption of cutting-edge methods that are used for DNA barcoding of pest species for confirmatory identifications, microsatellite analysis of pest populations to track high-risk pathways of invasive species, and screening of insect genomes for new molecular tools. These in-house techniques and skills are supporting PPQ programs through the development of identification tools and integrative projects to understand pest and vector distributions, dispersal, introduction pathways, and behaviors.

#### Recent Accomplishments Asian Citrus Psyllid/HLB

- Conducted validation trials of a leaf washing process for removing ACP from kaffir lime leaves and other edible citrus leaf products in California. Provided technical support for the writing of the Federal Domestic Quarantine Order on Fresh, Mature Citrus Leaves from Areas Quarantined for Asian Citrus Psyllid.
- Collaborated with Texas A&M University, extension, and the citrus industry to develop an areawide management approach to control ACP in Texas. The areawide management program has reduced insecticide application cost by 50 percent and increased the time before pest resurgence.
- Developed and evaluated the efficacy of insecticides as post-harvest treatments of citrus products to provide options for control of ACP prior to transport.
- Concluded a 2-year collaborative field study with ARS regarding the use of guava inter-plantings with citrus for the management of HLB.
- Completed host-specificity testing of the biocontrol agent *Tamarixia radiata* (Pakistan) and received a permit for field releases in Texas. Received, screened, and established two new colonies of *T. radiata* from Southeast Asia (Yunnan and Singapore), bringing the total to seven foreign collections that may exhibit enhanced efficacy or climatic adaptabilities.
- Developed two methods for mass production of *T. radiata.* Videotaped protocols for mass production, using field insectary cages on mature citrus trees, for use as a tech transfer tool to State departments of agriculture and university researchers.

#### Sweet Orange Scab

• Developed and shared an isolation method for the elusive fungal pathogen *Elsinoë australis*, the causal agent of SOS. Multiple isolates from Texas citrus (lemon, lime, grapefruit, orange, and tangerine) are now available to further the understanding of this pathogen and to help manage this plant disease.

 Initiated studies to evaluate several surface disinfectants for potential mitigation of the fungal pathogen that causes SOS. Interstate shipments of organic citrus were affected by the discovery of SOS in Texas, and the development of mitigation measures is in progress to assist organic citrus producers.

#### Brevipalpus Mites/Citrus Leprosis Virus (CiLV)

• Conducted surveys to document the distribution of *Brevipalpus* mites, a known vector of CiLV, in the citrusgrowing counties in the Rio Grande Valley of Texas. A regional distribution map coupled with a library of preserved specimens and DNA isolates will serve as future reference in case of introduction of the disease.

#### Mexican Fruit Fly Sterile Insect Technique (SIT)

- Supported the Mexfly eradication program by evaluating the survivorship of adult Mexflies released by both aircraft and ground release machines; the effectiveness of lures used in the trapping program; and the efficacy of pesticide treatments used in control efforts.
- Continued support of the Mexfly SIT program by evaluating eggs and pupae, as well as a male predominant strain, produced in Guatemala against the current strain used in the program.
- Developed a Mexfly monitoring tool that allows fruit fly coordinators and decisionmakers to analyze the effectiveness of sterile releases in program areas.

#### Arthropod Quarantine Facility

- Supported the joint ARS and DHS giant reed (*Arundo donax*) biological control program by receiving shipments of the root-feeding armored scale *Rhizaspidiotus donacis* and a leafminer *Lasioptera donacis*.
- Supported the biological control program against ACP by receiving and culturing shipments of the parasitoid *Tamarixia radiata* from China and Singapore.
- Supported the Mexfly eradication program by maintaining and evaluating a wild strain of fruit fly for potential use in the Mexfly SIT mass-rearing facility.

#### Molecular Diagnostics Unit

- Developed procedures for DNA barcode analysis of fruit fly pests and completed a pilot study using the technology to identify *Anastrepha* larvae from intercepted baggage at ports-of-entry in support of PPQ fruit fly programs.
- Developed new molecular procedures to help identify ACP populations within the United States and the possible sources of introduced populations.
- Generated new molecular tools for Medfly pathway analysis and demonstrated their utility for identifying high-risk pathways into the United States in support of PPQ fruit fly programs.

## CPHST Lab, Otis Air National Guard Base, Buzzards Bay, MA

#### Laboratory Director - Vic Mastro

The Otis Laboratory's mission is to identify, develop, and transfer technology for survey, exclusion, control, and risk assessment for APHIS and its cooperators. The lab serves a wide variety of PPQ programs that include: exotic pestdetection programs, such as CAPS; AQI phytosanitary treatments; and emergency response and eradication programs for ALB, EGVM, Asian gypsy moth (AGM), EAB, Sirex noctilio woodwasp, and other pests. Otis personnel identify high-risk exotic pests and develop survey technology to facilitate the early detection of introductions. The lab continues to support the AGM program by developing molecular methods to distinguish among subspecies of gypsy moth, producing the gypsy moth virus product Gypchek, and helping ensure the quality of gypsy moth lures. Recent developments in the use of microsatellites are providing a better definition of the geographic distribution of strains of gypsy moth throughout the world. Additional work is focused on the development of regulatory treatments for various commodities (grapes, citrus, figs, etc.) and the means of their conveyance, such as pallets and containers. The Otis Lab is developing rearing systems for EGVM and ALB and has biological control programs for EAB, S. noctilio, and the winter moth.

To fulfill its mission, the Otis Lab maintains cooperative relationships with other Federal agencies such as ARS and Forest Service, State departments of agriculture, universities, and private industry. These cooperative arrangements extend to government organizations and universities in a number of foreign locations, including Australia, Canada, China, Japan, Korea, New Zealand, Russia, and South Africa. The work includes developing methods to monitor and exclude AGM from North America, predicting invasiveness of organisms by assessing damage on expatriate North American plants in foreign locations, developing and evaluating attractants, and developing control techniques for targeted exotic pests.

#### **Recent Accomplishments**

#### European Grapevine Moth

- Led the EGVM TWG, whose recommendations have largely been adopted by the EGVM program. This program has resulted in a reduction from captures of 100,000 moths in 2010 to less than 100 moths in 2011, as well as deregulation of previously infested portions of 3 counties.
- Developed and evaluated phytosanitary treatments that allowed for intrastate, interstate, and international movement of grapes, *Vitus* nursery stock, grape products, and other commodities. Assessed the efficacy of insecticides for EGVM control programs, and confirmed suitability of attractant-based trapping methods for survey and population monitoring.
- Developed rearing systems for EGVM that support pesticide testing and commodity treatment development, and made significant progress in developing mass-rearing methodology in case it is needed in the future. The pink bollworm diet was found to be highly suitable for EGVM. Conducted tests for alternative mating-disruption formulations to support development of a monitoring tool that can be used when an area has been treated with mating disruption. This is important because the standard monitoring tool for the program is a pheromone-baited trap, which is rendered inoperable under a mating disruption regime.

#### Light Brown Apple Moth

• Continued to support the LBAM program by providing recommendations for survey and regulatory and control treatments. Several crops depend on mating-disruption treatment for production. The Otis Lab participated in the research that led to the adoption of this technology by the caneberry growers. Additional testing of

novel mating-disruption formulations in New Zealand is nearly complete. This work should provide the growers with treatment alternatives.

#### **Emerald Ash Borer**

- Worked in conjunction with USDA's Forest Service and ARS to successfully release several parasites that attack EAB. The lab continues to monitor the establishment and effectiveness of these parasites at field release sites and to consult with a PPQ parasite rearing facility in Michigan that was developed on the basis of this work. An additional parasite species collected from Russia is now under evaluation for field release. Field releases of parasites are now being evaluated in almost all States where EAB is known to be established.
- Continued testing to fine-tune EAB traps and lures that were developed by the Otis Lab and are used nationally by the EAB program for its survey work. The lab developed a dry (non-sticky) trap for EAB and provided a recommendation for its use in future years. The Otis Lab is collaborating with Pennsylvania State University to evaluate aversion of this trap to determine if its performance can be further enhanced by the addition of buprestid models.
- Conducted additional studies with cooperators in Canada to evaluate a modified version of the traps as an auto-dissemination for fungal pathogens.
- Evaluated EAB dispersal and the systemic insecticide emamectin benzoate in West Virginia for EAB management.
- Continued to coordinate the scientific efforts ongoing in Federal and State agencies and universities, including cooperating with the U.S. Forest Service to develop a risk-based model that would optimize placement of traps within the limited program resources available for survey.

#### Asian Longhorned Beetle

- Continued assessing ALB populations and tree-damage patterns to understand the dynamics and spread of ALB infestations in New York, New Jersey, Massachusetts, and the newly discovered population in Bethel, OH. The lab has consulted with the ALB program regarding the age, spread, and survey methodology for the Massachusetts and Ohio infestations.
- Participated in public and management meetings in support of the program in both Massachusetts and Ohio.

- Began studies to develop regulatory treatments for ALB in nursery stock with results expected by the spring of 2013.
- Updated the official host tree list, which will help save program resources used for survey and treatment. The genus *Celtis* has been deleted, and the lab is now evaluating species in the genus *Betula*. This is important because in Massachusetts and Ohio there are many black and yellow birch that could either potentially harbor ALB or be eliminated from survey.
- Conducted a large study in Massachusetts and Ohio to compare the efficacy and cost effectiveness of visual survey, traps, and sniffer dogs. Efforts continue to attempt to identify additional attractants for ALB adults.

### Gypsy Moth

- Continued to perform DNA analysis of gypsy moth specimens submitted by PPQ's domestic monitoring program and intercepted and submitted by PPQ and DHS. In addition, work continued toward developing a worldwide library for microsatellite DNA markers from gypsy moths, which should prove useful in determining likely source populations for intercepted gypsy moths and newly detected populations. Preliminary analysis of this worldwide collection of gypsy moth specimens indicates that within the species there are at least four distinguishable groups. The Otis Lab is nearing completion of the first phase of this work and continues to work cooperatively with Cornell University to find genetic "flight markers."
- Started work toward developing DNA "barcoding" capabilities so that intercepted lymantrids that are not gypsy moth can be readily and accurately identified.

#### Sirex Woodwasp

 Conducted additional experimental releases in New York and Michigan of the nematode *Beddingia siricidicola*, a biocontrol agent for Sirex woodwasp. The Otis Lab has been unable to evaluate the spread and impacts of the released Kamona strain of the nematode under natural conditions because permission has not been granted for an environmental release. However, the lab now has the ability to distinguish between the endemic and the Kamona strains of the nematode and has determined the distribution of the endemic stain in the United States.

- Successfully identified a male-produced pheromone from *Sirex noctilio*, which has eluded other researchers for over 50 years. Additional work will be conducted to determine how to utilize it for survey or control efforts.
- Identified an improved trap design based on preliminary results. This design will undergo additional testing in the summer of 2012.

### Survey Support

- Formulated and provided over 50,000 pheromone dispensers to support the Cooperative Agricultural Pest Survey (CAPS) and other survey efforts. The lab continued to provide advice on traps, lures, and survey design.
- Developed a universal "moth" dry trap for survey and prototypes for testing. The lab continues to evaluate dry-stick adhesives for traps used by APHIS-supported survey programs.

### **Phytosanitary Treatments**

- Continued development of regulatory treatments for solid wood packing, hardwood and softwood logs, and firewood. In particular, good progress was made toward validating the use of radio-frequency waves for phytosanitary treatment of wood. Testing on EAB and ALB has now been completed, and the results are being analyzed. This work supports a petition to accept this treatment for inclusion under International Standards for Phytosanitary Measures-15.
- Conducted tests on the use of vacuum steam treatment for veneer-quality logs. Preliminary work on these techniques is encouraging. If it is successful, it could replace one of the major uses of methyl bromide.
   Cooperative work with the University of Tennessee is underway to develop an effective treatment for walnut wood infested with the walnut twig beetle and thousand canker disease.

## **CPHST Lab, Phoenix, AZ**

#### Laboratory Director - Dr. Richard Zink

The Phoenix Laboratory's mission is to develop, adapt, and implement areawide control technologies for new and existing program pests. Current work includes developing control tools, methods, equipment, and support for pink bollworm (PBW), rangeland grasshopper/Mormon cricket complex, and LBAM. These control technologies include biocontrol, the sterile insect technique (SIT), pheromones, new chemicals, ground and aerial delivery systems, and geographic information system applications. The lab's scientists conduct extensive laboratory and field developmental and operational scale studies to test and validate materials, methods, and equipment. The lab employs specialized equipment, including ground and aerial application technology; environmental chambers and mass-rearing modules; a twin-screw extruder for insect diet development; a room that allows for accurate simulation of aerial applications of sprayed products; a quarantine laboratory for large-scale rearing of genetically modified PBW; greenhouses; laboratory-located mini-rangeland and cotton field plots; and equipment for testing pesticide and pheromone application technology.

The Phoenix Lab's rangeland section works with Federal and State customers to provide technical assistance for the grasshopper and Mormon cricket control programs. This section also develops and implements solutions to program problems and continuously evaluates the technology and tools of the control program to maintain state-of-the-art status. The PBW section supports the PBW eradication program by providing expertise on pheromone mating disruption, custom rearing and mass-rearing of insects, SIT mechanisms, insect population monitoring, and insect behavior. This section works closely with the PBW rearing facility, the CPHST LBAM Unit and Albany facility, the Arizona Cotton Research and Protection Council, ARS' Arid Lands Agricultural Research Center, the University of Arizona's Entomology Department, local cotton growers, the International PBW Eradication Program, and State and regional PPQ offices.

#### **Recent Accomplishments**

#### **Rangeland Section**

- Conducted a replicated aerially applied dose study of Rynaxypyr to determine the dose to be used in the final operational scale evaluation and development for rangeland grasshopper control. This pesticide will be an additional alternative to malathion and carbaryl, the currently used traditional insecticides.
- Conducted a semi-operational scale evaluation of a commercial adjuvant as a replacement for oil diluents in aerial application mixes of diflubenzuron to control rangeland grasshoppers. This study at this scale was successful: if a larger operational scale evaluation proves successful, this method will be recommended for

implementation in to the grasshopper/Mormon cricket program. It potentially will produce significant savings in terms of mixing and loading time, reduced diluent volumes, and aircraft load efficiency.

- Evaluated new carbaryl-based bait for initial effectiveness against rangeland grasshoppers and Mormon crickets. This new bait appears equal to other carbaryl-based baits and will be further tested against other grasshopper species that are typically not highly susceptible to carbaryl bait.
- Conducted an initial bait study to determine a dose of Rynaxypyr that could be used against rangeland grasshoppers. Results indicate that low concentrations will be possible, but further testing will be required to determine the lowest effective concentration. Immature grasshoppers were significantly more susceptible than adults. Successful development of this bait will provide a much-needed alternative active ingredient for baits against rangeland grasshoppers and Mormon crickets.
- Continued work on a project to develop an inventory for the prevalence of naturally occurring pathogens in rangeland grasshopper populations by surveying untreated, field-collected grasshoppers.
- Acquired and catalogued thousands of books, technical bulletins, circulars, university-published documents, pamphlets, Web sites, maps, and articles concerning the PBW, grasshopper, and Mormon cricket.

#### Pink Bollworm Section

- Participated in creating and updating trapping quality assurance protocols for the International PBW Eradication Program, including supplying fresh, un-dyed, fluorescing PBW moths to program managers in four U.S. and two Mexican States.
- Researched PBW trapping methods and demonstrated success with a new modified-funnel trap design, and began implementing new containment procedures for spent PBW rearing facility (PBWRF) diet and laying cages. Despite the capture of un-dyed adult moths that led to these efforts, no evidence of wild infestation was observed, indicating the success of the eradication program (greater than a 98-percent overall population reduction).
- Produced and shipped PBW diet to at least eight U.S. and English cooperators for rearing diverse Lepidoptera species, including PBW, LBAM, EGVM, and obliquebanded leafroller.

- Researched reconstituted freeze-dried PBW diet and found it similar to a fresh diet in terms of rearing success and apparent nutritional content, with the benefits of reduced weight and shipping costs as well as an extended shelf life, while maximizing use of the diet extruder.
- Finished modifications to render the PBWRF's 34-millimeter twin-screw extruder ready for diet production, and collaborated with the PBWRF to write a complete and illustrated standard operating procedures manual for operation of the extruder and its auxiliary equipment.
- Determined that mass-reared PBW from APHIS and University of Arizona colonies maintain the ability to enter diapause, despite not being allowed to do so for many generations.
- Conducted investigations of potential use of the tetracycline-dependent OX3402C PBW strain in a sterilerelease type of program, which seem to indicate that the strain is functioning as designed, with insects dying during early larval stages.
- Continued tests of a custom air cannon as a fast and inexpensive method of releasing healthy, sterile PBW adults safely into the field. Longevity of test insects shot from the cannon was unaffected compared to an untreated control.
- Continued development of a digital bibliographic repository for documents associated with PBW, grasshopper, and Mormon cricket. Crucial components included a metadata descriptor manual, a manual for the digitization of physical collections, and "bit rot" mitigation practices to insure the longevity and stability of data.

## CPHST Plant Epidemiology and Risk Analysis Laboratory, Raleigh, NC

#### Laboratory Director - Robert Griffin

The CPHST Plant Epidemiology and Risk Analysis Laboratory (PERAL) is PPQ's primary unit producing pest risk analyses (PRA). In this laboratory, a diverse group of scientists and professionals provides essential scientific support to risk-based policymaking across a broad range of phytosanitary issues. Staff members use sound science to analyze import, export, and operational issues. PRA are essential to help safeguard American agriculture and plant health from harmful exotic plant pests in both managed and unmanaged ecosystems. More specifically, the analyses help PPQ to design risk-based regulations for import and domestic pest management programs, identify and assess new pest threats, monitor the effectiveness of existing programs, and optimize available resources to enhance protection. PERAL personnel also provide technical support documents that PPQ requires for pests, commodities, and pathways. These products may include risk maps that indicate existing or potential range domestically or internationally or that predict ranges from weather- or climate-matching analyses.

Over the years, PERAL has established itself as a global leader in both productivity and quality management. PERAL is currently the only International Standards Organization (ISO)-certified plant health risk analysis unit in the world. The group contributes significantly to the promotion of international dialogue and increased capacity for science-based management of phytosanitary issues through its Risk Analysis Mentoring Program for visiting scientists. In addition, PERAL provides basic PRA training workshops, with topics covering the spectrum of concepts, methods, and resources associated with pest risk analysis. Furthermore, PERAL promotes regional and international harmonization of plant health regulations by providing scientific support to PPQ in the North American Plant Protection Organization (NAPPO) and the International Plant Protection Convention (IPPC).

#### **Recent Accomplishments**

- PERAL finalized 24 original Q-56 risk assessments and pest lists in 2011. These risk assessments represent new potential imports of 26 commodities from 41 different countries. PERAL also completed 2 original Q-37 risk assessments and revised 41 Q-56 risk assessments and 2 Q-37 risk assessments.
- PERAL processed risk assessment and informational documents for the organism, pathway, citrus, and risk-mapping areas, including 7 organism, 11 pathway, 1 ad-hoc, 7 citrus, and 13 risk-mapping documents.
   PERAL also completed 27 requests for export assistance, one of which helped to facilitate the opening of the Mexican market to peaches from Georgia and South Carolina.
- The New Pest Advisory Group (NPAG) completed 46 full NPAG reports and 35 preassessments for organisms that were not considered NPAG pests in FY 2011. Of the 46 pests that were the subject of full NPAG reports, 17 were deregulated after consultation with the National Plant Board, 21 received port policies of reportable/

actionable, and 8 were sent to Emergency and Domestic Programs for continued discussions with the States.

- The Deregulation Evaluation for Established Pests project analyzed 34 pests, 17 of which were determined to no longer meet the definition of a quarantine pest and were eventually deregulated at U.S. ports-of-entry after consultation with the National Plant Board.
- The PERAL Weed team completed 10 original weed risk assessments and published a paper in Biological Invasions on the development and validation of their weed screening tool.
- The Exotic Pest Information Collection and Analysis team produced weekly notifications containing approximately 200 articles on pests of regulatory significance.
- The Global Pest and Disease Database and Data Archival and Reporting Tool teams added 1,132 pests to the database.
- PERAL staff organized and delivered six risk analysis workshops, including the first annual weed risk assessment workshop and a Spanish-language workshop in Lima, Peru; hosted scientists from Egypt, Brazil, and Vietnam as participants in the Risk Analysis Mentoring Program; and presented at a risk analysis workshop in South Africa.

## **Biological Control Unit, Raleigh, NC**

#### Coordinator - Dr. Kenneth Bloem

The CPHST Biological Control Unit (BCU) is a virtual team of scientists (14–18 depending on project approvals from year to year) located at various CPHST laboratories and plant protection stations, including Fort Collins, CO; Mission, TX; Otis, MA; Phoenix, AZ; Albany, CA; Miami, FL; and Guatemala City, Guatemala. The BCU focuses on developing technologies that support the safe use of parasitoids, predators, herbivores, and pathogens to help mitigate the impacts of introduced invasive weeds and plant pests, while minimizing impacts on the environment and nontarget organisms.

CPHST scientists provide technical oversight and expertise to programs to ensure that gaps in scientific knowledge are identified and addressed, cooperators deliver needed services, and implementation protocols and educational materials are effectively developed and transferred to stakeholders as quickly as possible. More specifically, they provide permitted biocontrol agents collected from established field insectaries for distribution by PPQ and other project cooperators, develop new rearing and monitoring systems, and work to ensure the safety of biocontrol agents by conducting both preand post-release impact studies. Project selection is based on national review recommendations from stakeholders within PPQ, including Emergency and Domestic Programs, Eastern Region, Western Region, and individual State plant health directors, as well as State plant regulatory officials and the National Plant Board.

### **Recent Accomplishments**

#### Canada Thistle

CPHST Fort Collins completed an assessment of several accidentally introduced and native arthropods attacking the exotic weed Canada thistle. The mite *Aceria anthocoptes* and the lace bug *Corythucha distincta* were shown to have a broad host range that includes many native thistles, which precludes their use as biocontrol agents. In addition, a survey for fungal pathogens on Canada thistle was conducted in northwestern China through collaborations with CABI Europe-Switzerland. The most promising pathogen collected to date is a white rust pathogen, *Pustula (Albugo) tragopogonis*. Although additional taxonomic work and host range testing are needed, initial field observations suggest that the rust could be highly specific and damaging to the weed.

#### Hound's-Tongue

In 2011, CPHST Fort Collins continued to participate in a survey of the root weevil *Mogulones crucifer* in Washington and Idaho. *M. crucifer* is a hound's-tongue biocontrol agent that was released in western Canada and has spread into the northwestern United States, but it is not permitted for U.S. release. The survey has documented the spread and nontarget plant utilization by the weevil in these two States.

#### **Russian Knapweed**

A new Russian knapweed biological control agent, the gall midge *Jaapiella ivannikovi*, was permitted for U.S. release in 2009. In 2011, a *Jaapiella* colony was initiated at CPHST Fort Collins, in partnership with Montana State University, and a greenhouse-based rearing program provided galls and insects for field release at 19 sites in 7 States: California, Colorado, Idaho, New Mexico, Oregon, Washington, and Wyoming. These 2011 *J. ivannikovi* releases were the first occurring in these States, with the exceptions of Colorado and Wyoming.

#### Yellow Toadflax

CPHST Fort Collins initiated a greenhouse-based rearing effort with a yellow toadflax-adapted strain of the stem-mining weevil *Mecinus janthinus*, in partnership with Colorado State University. In 2011, weevils were provided for field release in Colorado and West Virginia. PPQ partners in Montana provided additional insects for releases in Colorado, Idaho, North Dakota, Oregon, and South Dakota. This effort will continue through at least 2012 to facilitate releases in a number of additional States in the Western Region and Eastern Region.

#### Asian Citrus Psyllid

CPHST Mission received a field release permit for a Pakistan strain of *Tamarixia radiata*, a parasitoid of ACP (vector of citrus greening disease), after completion of host-specificity tests conducted in collaboration with the University of California–Riverside. It is hoped that the developmental requirements of this new strain of *T. radiata* will more closely match the unique environmental conditions of the citrus-growing areas found in the United States. In addition, two methods for mass production of *T. radiata* were developed. The first method uses large field insectary cages over mature lemon and lime trees. The second method utilizes smaller cages and potted plants to facilitate greenhouse rearing. Baseline population data of the endemic strain of *T. radiata* was gathered to compare against future releases of the new strain.

#### Coffee Mealybug

The coffee mealybug, *Planococcus lilacinus*, has a broad host range and poses a significant threat to ornamental and agricultural crops. In 2011—in cooperation with APHIS' International Services program in the Dominican Republic, the Instituto Dominicano de Investigaciones Agropecuarias y Forestales, and Florida A&M University—CPHST supported a Master's student research project to determine the impact of the coffee mealybug on local agriculture in the Dominican Republic. As an offshore safeguarding effort in case the pest becomes established in the United States, the project is also researching whether natural enemies are providing any control for the pest.

#### **Emerald Ash Borer**

In 2011, CPHST Otis continued to study ways to enhance the establishment of the three Chinese parasitoids approved for release in the United States. Spraying a release site with sugar water as parasitoid food may reduce dispersal and enhance parasitization. Although large-scale releases only began in 2009, CPHST Otis has documented establishment of the parasitoids in five States. CPHST Otis also conducted host-specificity tests with a new *Spathius* species from Russia to determine the likelihood that it might attack nontarget woodborers native to North America. The new species from Russia may be more cold tolerant than *Spathius agrili* from China, which was collected from a considerably warmer climate.

#### Grasshoppers

CPHST Phoenix, under permit and in cooperation with ARS, is conducting a long-term field exposure experiment with *Metarhizium acridum*, a pathogen specific for grasshoppers commercialized and sold as Green Guard and Green Muscle, which are Australian and African strains respectively. In response to public comments on the environmental assessment, experiments are being conducted to determine the fungi's ability to remain viable under field conditions. The first overwintering experiments have been completed and initial results indicate that the fall exposure of *M. acridum* resulted in persistence through 28 days, although final activity was much diminished. Spring exposure experiments and a permit application for a 10-acre field release are being prepared.

#### **Imported Fire Ant**

The APHIS-funded IFA phorid fly (*Pseudacteon* spp.) biological control rearing and release program continued in 2011 with multiple releases of the third fly species, *P. obtusus*, and the first release of a fourth species, *P. cultellatus*. The first two species, *P. tricuspis* and *P. curvatus*, are now established in more than 50 percent of the IFA quarantined area.

#### Mediterranean Fruit Fly

The Moscamed Program and CPHST Guatemala produced and conducted open field releases of the egg parasitoid *Fopius ceratitivorus* in a highly sensitive ecological area around Lake Atitlan with problematic reoccurring medfly infestations. Parasitoids were released at the rate of about 3,000 female parasitoids per hectare in a 100-hectare area during a 32-week period, in combination with sterile insects. The pest appears to have finally been eradicated from this area, demonstrating the utility of parasitoids in a large eradication effort.

#### Passion Vine Mealybug

Passion vine mealybug, *Planococcus minor*, is a pest of over 250 plants including citrus, corn, grape, potato, and soybeans. CPHST developed protocols to survey and monitor populations of P. minor using a recently identified sex pheromone. These methods led to the discovery of the pest in south Florida in 2011. Working with cooperators in the Caribbean, CPHST also showed that *P. minor* and *P. citri* share a common suite of natural enemies that keep them under good biological control. These natural enemies were found to be present and attacking *P. citri* in Florida and have now been shown to be attacking *P. minor* since its establishment. This work resulted in PPQ changing the status of this pest, once a 'top 10' invasive, to non-actionable/non-regulated.

#### Winter Moth

CPHST Otis continued to support cooperators at the University of Massachusetts working on biological control of the winter moth, a leaf-feeding geometrid native to Europe that invaded eastern Massachusetts in around 2000 and is causing widespread defoliation and tree death. Previous invasions by this species in Canada were suppressed by the introduction of the tachinid fly, *Cyzenis albicans*, from Europe. In 2010, establishment of *C. albicans* was documented at five of six releases in Massachusetts and Rhode Island. In 2011, over 7,000 adult *C. albicans* were released compared to about 2,000 in previous years, which allowed the number of release sites to be increased from 6 to 15.

## Fruit Fly Unit, Raleigh, NC

#### **Coordinator - John Stewart**

The CPHST Fruit Fly Unit supports the APHIS Fruit Fly Exclusion and Detection program's major goal to strengthen detection and response capabilities, preventative release programs, and control programs in order to prevent exotic fruit fly populations from becoming established and/or spreading within the United States. This goal also supports USDA's goals to protect agricultural health from pests to assure a safe and plentiful food supply and to strengthen emergency response preparedness.

During 2011, the Fruit Fly Unit has integrated into APHIS' cooperative fruit fly programs and concentrated on technical and methods support for the field during emergency response quarantine and eradication activities. These include: support to improve fruit fly rearing and emergence

techniques; fruit fly aerial sterile releases; release equipment calibration; ground pesticide treatments and operational techniques; enhanced detection; modeling of pest colonization and dispersal; and quality assurance of program resources such as lures, diets, and control pesticides. In 2011, there were major fruit fly quarantines in Florida, Texas, and California.

#### **Recent Accomplishments**

- Developed dispersal and colonization models that support operations.
- Improved identification techniques and tools.
- Developed a host compendium and updated host lists for regulatory quarantine.
- Developed molecular methods to support pathway analysis, regulatory decisions, and identification.
- Developed an SIT monitoring model for sterile release evaluations and regulatory decisionmaking.
- Developed provisional post-harvest treatments for quarantine commodity movement.
- Supported additional pesticide labels for pre-harvest treatments.
- Approved more dilution rates for GF 120 Spinosad applications.
- Improved ground-treatment operational issues with foam fighters and odor controls.
- Developed a Systems Approach Working Group to address quarantine movement of major commodities including walnut, tomato, and cherry.
- Developed lure, diets, and pesticide quality assurance bioassays.
- Developed the CPHST Quality Assurance Survey Program for program lure quality assurance and standard protocols.
- Approved a new solid three-component biolure for efficiency and cost savings.
- Analyzed alternate Mexfly host reservoirs and evaluated over-summering hosts.
- Performed SIT release equipment quality assurance and calibration.
- Evaluated SIT aerial swath tests in Florida and California.
- Evaluated emergence pupae holding temperature and provided recommendations.
- Performed Mexfly Rearing and Emergence Facility quality assurance review and provided recommendations.

- Provided fruit fly rearing and diet microbial contamination analysis and recommended new procedures.
- Developed new egg shipment and bubbling procedures for Mexfly rearing.

## Light Brown Apple Moth Unit, Moss Landing, CA

#### **Coordinator - Dr. Gregory Simmons**

The LBAM Unit coordinates and conducts scientific and technical support activities in response to two invasive species that recently arrived in California, LBAM, Epiphyas postvittana, and EGVM, Lobesia botrana. In 2011, the unit worked to develop technology and to provide technical analysis of program data needed to control and manage these pests and to assist export markets of affected commodities. A major focus of the unit in 2011 was to conduct a pilot evaluation of the sterile insect release technique for LBAM suppression. For the evaluation, sterile LBAM were mass-reared, irradiated, and shipped to an LBAM-infested area of Long Beach, CA, for 2 months of field testing. A coordination of efforts between the LBAM Unit and the Fort Collins, Mission, Otis, and Phoenix laboratories has resulted in advancements in identification, detection, and control of LBAM. The unit also provided technical support and methods development to detect, delimit, and control EGVM in California.

## **Recent Accomplishments**

- Released over 600,000 sterile LBAM in an SIT pilot project in Long Beach, CA, to evaluate the potential for use of SIT to suppress LBAM populations.
- Developed LBAM mass-rearing and collection methods allowing for production of more than 1,000 moths per tray and up to 500,000 moths per week in a small pilot-production facility.
- Validated cold treatment for control of LBAM for movement of strawberry plants for planting.
- Worked with ARS and the CPHST Otis Lab to test methyl bromide and alternative fumigants for LBAM post-harvest control for several commodities including apples, stonefruits, and caneberries. The work resulted in a publication on post-harvest treatments for LBAM.
- Worked with the University of California and CPHST Otis to develop and test regulatory control treatments and provide technical recommendations for LBAM on ornamental nursery stock. This work has resulted in the recent addition of an insect growth regulator

(methoxyfenozide) and the addition of several horticultural oils to the California Department of Food and Agriculture-approved treatment list.

- Analyzed LBAM trapping records and literature to provide a technical recommendation for the size of an effective quarantine area around a new LBAM find.
- Tested a new four-component pheromone lure under California conditions and determined the use of the new lure would significantly improve trap catch over the current two-component lure.
- Supported the EGVM control program by conducting studies of phenology and surveys of alternate host plants in Napa County; conducting a demonstration of mating disruption for control of EGVM on a small isolated population; testing monitoring methods for areas under areawide control using mating disruption; and testing different delta traps and pheromone lure formulations for specification of contract purchases for the EGVM control program.
- Conducted research using EGVM and LBAM to assess the effects of wine-making procedures on the survivability of larvae and pupae during crushing, pressing, and storage of grape must for shipping. This work has allowed refinements to current regulations on treatment of wine-making green waste and pressing procedures and resulted in the recent deregulation of red wine must.
- Established an EGVM quarantine colony and developed efficient mass-rearing methods in support of post-harvest treatment research, pesticide efficacy basic biological studies, and regulatory treatments of green waste and wine-making processes.

## **Treatment Quality Assurance Unit, Raleigh, NC**

## **Director - Scott Wood**

The Treatment Quality Assurance Unit (TQAU) develops, adapts, and supports technology to detect, identify, and mitigate the risk posed by exotic pests in preclearance programs and at ports-of-entry. Commodity treatments are the last line of defense in preventing exotic pest invasions. Currently, core activities within TQAU include developing quarantine treatments, contributing to the treatment manual for ports-of-entry, certifying shipping containers and vessels, maintaining the Commodity Treatment Information System, training-the-trainer for proper application of treatments, certifying domestic and offshore non-routine commodity treatment facilities, and conducting quality assurance audits of treatments performed at domestic and offshore sites.

#### **Recent Accomplishments**

- Continued support and expansion of the Commodity Treatment Information System: Fumigation Form 429 Database, Irradiation Reporting and Accountability Database (IRADS), 556 In-transit Cold Treatment Database, Niger Seed Database, Treatment Index Database, and Certified Vessels and Containers Database.
- Reviewed quarantine treatment research and/or developed research protocols aimed at developing methyl bromide, cold, hot water, irradiation, and vapor heat treatments for high-risk pests, including Queensland fruit fly, Oriental fruit fly, Asian fruit fly, ACP, EAB, LBAM, Chilean false red mite, EGVM, false codling moth, litchi moth, Mediterranean fruit fly, South American fruit fly, spotted wing Drosophila, sweet potato weevil, and West Indian sweet potato weevil.
- Reviewed treatment proposals for the International Plant Protection Convention's Technical Panel on Phytosanitary Treatments.
- Provided support to the Quadrilateral Scientific Collaboration Work Group and its project teams.
- Compiled an extensive methyl bromide usage report for 2011, including data on commodity, country of origin, reporting station, schedule, and contractor. Previous yearly reports were used to develop an APHIS economic study (U.S. Trade and the Use of Methyl Bromide as Phytosanitary Treatment) and an ARS publication (Status of Alternatives for Methyl Bromide in the United States).
- Worked with the Professional Development Center to create training materials for cold treatment vessels inspection, the mango hot water program, the irradiation program, and a fumigation workshop.
- Provided more than 200 treatment recommendations for PPQ officers and stakeholders.
- Edited the phytosanitary treatments chapter for the Handbook of Plant Biosecurity. Authored fumigation, cold treatment, irradiation, and international treatment standards sections.
- Participated in the Agricultural Risk Management initiative.
- Approved three irradiation facility plans (Benebion (Mexico), Mintec Sinagama (Malaysia), and National Center for Electron Beam Research (Texas)). Certified one irradiation facility (FTSI (Florida)).

- Certified 143,043 cold treatment containers (345 container series); certified 50 cold treatment vessels; approved 2 cold treatment warehouse plans; approved 2 equipment installations; and maintained existing ISO 9001:2008 registration for cold treatment.
- Approved 12 fumigation chamber plans, and developed a fumigation chamber application for future ISO certification.
- Approved 3 hot water mango facility plans and approved 10 hot water mango facility modifications (Caribbean, Central America, and South America).
- Approved Niger seed facility plans (India); certified two Niger seed facilities (New Jersey and Maryland); and trained PPQ Regional staff to assume Niger seed facility certifications in the future.
- Continued electronic indexing of documents in the TQAU library in an effort to develop a paperless electronic system for AQI treatments, policies, and research literature. The multiple-year process has accumulated over 17,000 documents.

## National Plant Pathogen Laboratory Accreditation Program, Raleigh, NC

#### **Coordinator - Dr. Patrick Shiel**

The National Plant Protection Laboratory Accreditation Program (NPPLAP) evaluates laboratories to ensure their capability to make accurate molecular diagnostic determinations for regulatory purposes. In addition to ensuring lab capability within PPQ and other agencies in USDA, NPPLAP engages the National Plant Diagnostic Network (NPDN) and State laboratories in this process to increase diagnostic capacity and proficiency to harmonize a dispersed laboratory network. The goal of this program is to establish a state of readiness when needed by PPQ in emergency situations. NPPLAP also fosters the adoption of practices that promote continuous improvement and accreditation standards suitable for use by plant diagnostic labs. In addition, NPPLAP serves to develop a functional Quality Assurance program for plant diagnostic labs. NPPLAP currently accredits laboratories for USDA regulatory molecular diagnostics and provides yearly certifications to diagnose Phytophthora ramorum, Plum pox virus, and the HLB (citrus greening) pathogen. This is accomplished by NPPLAP lab inspections for accreditation of new labs and certification of analysts through a yearly proficiency test program deployed by the CPHST Beltsville Lab Proficiency Test Group.

NPPLAP's current primary initiative is to develop strategic partnerships with laboratories performing plant regulatory diagnostics to develop and deploy a functional quality management system through partnership with established accreditation bodies. This is accomplished by continued alignment with quality management systems used by the National Animal Health Laboratory Network (NAHLN), the American Association of Veterinary Laboratory Diagnosticians, Inc. (AAVLD), and other established laboratory accreditation systems.

#### **Recent Accomplishments**

- Approved 17 labs and 27 analysts for *P. ramorum* testing. There was a 16-percent increase in proficiency test participation over 2010.
- Approved 12 labs and 29 analysts for HLB testing. Proficient test participation increased by 20.8 percent, and participant submission of results decreased in time to an average of 14 days.
- Organized a NAHLN/AAVLD training course to provide an introduction to quality management for NPDN labs.
- Conducted Internal Auditing Training for NPDN.
- Provided leadership in NPDN quality management initiative.
- Organized Plum pox virus TWG meetings.
- Continued participation in the DHS Integrated Consortium of Laboratory Networks (ICLN) and participated in the ICLN *Rathayibacter toxicus* Tabletop Exercise.

## **Project Highlights**

## Molecular Diagnostics Methods and Pathogenicity Testing for the Sweet Orange Scab Fungal Pathogen Elsinoë australis

#### Location: CPHST Beltsville Laboratory Lead Scientist: Kurt Zeller Team Member: Ping Yang Cooperators: Madhurababu Kunta, Mani Skaria (Texas A&M University-Kingsville, Citrus Center)

In July 2010, PPQ confirmed the first discovery of the fungal pathogen Elsinoë australis, causal agent of SOS in the United States, from residential lemon and tangerine trees in Harris County, TX. These citrus samples were initially collected as part of the annual citrus commodity survey performed under a cooperative agreement with Texas A&M University through the Citrus Health Response Program (CHRP). The detections in Harris County are approximately 320 miles from the lower Rio Grande Valley where most of the commercial citrus production in Texas is located. Since these initial findings, additional areas with SOS-positive trees have been found in other Texas counties and in Louisiana. SOS is a serious disease of citrus crops worldwide that causes unsightly lesions that can render fruits unmarketable. State and Federal customers have requested that CPHST develop and validate molecular methods for confirming the presence of Elsinoë australis and for distinguishing it from the closely related species Elsinoë fawcettii that can co-occur on citrus.

In 2011, the CPHST Beltsville Laboratory validated a conventional PCR method to identify *Elsinoë australis* and to differentiate it from *Elsinoë fawcettii* using primers developed by Hyun et al. (2007). The lab wrote work instructions for this method and made them available to the diagnostics community. *Elsinoë australis* and *Elsinoë fawcettii* are difficult for a non-expert to differentiate morphologically and are difficult to isolate from infected fruit because they grow very slowly. The two species overlap in host range among citrus and cause similar types of lesions.

We also developed quantitative PCR primers and probes for a novel real-time PCR diagnostic assay for *E. australis*. Testing and validation of this method are underway, and a work instruction for this method should be available in 2012. We have also provided support to our CPHST Mission colleagues as they work to develop fruit treatment methods for packinghouses to help counter the spread of this disease to uninfested regions of the United States. We also adapted a method used in pathogenicity testing of *Elsinoë* spp. (Timmer et al., 1996) on detached citrus leaves to complete Koch's Postulates for *Elsinoë australis* originating from Texas on orange leaves. This was done in order to confirm some of the unusual disease symptoms and increased host range that had been seen in the field. We were able to successfully inoculate detached citrus leaves in the laboratory, obtain symptoms of the disease, and successfully re-isolate *Elsinoë australis* from those leaves. Scientists at the CPHST Mission Laboratory in Texas performed parallel inoculations on both detached leaves and on fruit.

Finally, we began isolating DNA sequences from representative cultures of *E. australis* to develop population genetics and forensic markers. Over 100 clones have been sequenced, and several candidate markers have been identified. We continue to develop additional markers to begin forensic characterization of the U.S. populations.

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## Modeling Asian Gypsy Moth Introduction for the States of Washington, Oregon, California, and Texas

## Location: CPHST Fort Collins Laboratory Lead Scientist: Lisa Kennaway

Asian gypsy moth (AGM), *Lymantria dispar*, is an exotic pest that has been detected but has not become established in the United States. The threat to American agriculture is significant due to the broad host range of this moth, which includes 500 species of trees and shrubs. The AGM is closely related to the European gypsy moth. However, AGM has a much broader host range and the females are capable of flying up to 25 miles, unlike the flightless female European gypsy moth. This makes it crucial to identify early introductions of the pest to prevent its establishment in North America.

A primary pathway of introduction of AGM into America is via ship and cargo traffic from Asia. These trade patterns place the coastal United States at high risk of AGM introduction. Program managers currently have surveillance systems in place to identify, and eradicate if necessary, any moths prior to establishment. The trapping systems are organized and managed with expert local knowledge, and higher trap densities are placed in and near major shipping ports and transportation routes.

To enhance the placement of these traps, a geospatial model has been developed to predict areas with the highest AGM introduction risk based on transportation, population, and environmental variables. The goal of this model is to improve and/or validate existing trapping locations. From 2009–2011, models were delivered to Washington, Oregon, and California. In 2011, model work for Texas was initiated with the final deliverable planned for 2012. The analysis method uses a raster-based geospatial model and is strongly guided by input from stakeholder groups within each State. For each model, a group was organized that included representatives from PPQ Regions, Emergency and Domestic Programs, and Smuggling Interdiction and Trade Compliance; State departments of agriculture; and DHS-CBP. Each model is unique in its set of data inputs and weighted schema, which were discussed and finalized among each stakeholder group. The model results were delivered to stakeholders through a final report, spatial data, and a quantitative spreadsheet analysis of modeled risk zones and existing traps locations.

The success of this project is heavily dependent on input from the stakeholder group that includes participants from Federal, State, and local levels. The analysis method and technology is driven by the CPHST Fort Collins Lab, but the decisions regarding input data and weighting rely on stakeholders' expertise. By including stakeholders, ownership is shared among the entire group, and value is measured by use in field operations. Interest for this type of analysis continues to grow as additional States face increased risk for AGM introduction. In addition, this approach can be used to predict efficient trap location for a variety of other pests, as managing programs face reductions in resources.

## Washington/Oregon Model

Columbia River High/Low Risk Ports Canada/CA Border Crossings 08 AGM Ship Interceptions

#### **Transportation**

Highways Railways Weigh Stations Rest Areas

#### **Population**

Census Urban Nightlights Commodity Import Volume

#### California Model Introduction/Maritime

Shipping Ports Deepwater Ports 03-09 AGM Trap Interceptions 08-09 AGM Ship Interceptions

#### **Transportation**

Highways Railways Inland Waterway Warehouse Districts Inspection Stations

Population Census

#### Environmental

Wind Zones Parks Forest Density

## Texas Model

Ports Mexico Border Crossings Military Household Shipments Firework Distributors

#### **Transportation**

Highway Railway

#### Population Census

Environmental Vegetation

## Development of Imported Fire Ant Quarantine Cold Temperature Techniques for Certifying Bulk Soil for Movement

#### Location: CPHST Lab, Gulfport, MS Lead Scientist: Anne-Marie Callcott Team Members: Craig Hinton, Lee McAnally Cooperators: Karen Vail (University of Tennessee), Jeremy Shoop (City-State, LLC)

As a federally regulated item under the Federal Imported Fire Ant Quarantine, bulk soil must be treated in an approved manner prior to shipping outside of a regulated area to prevent movement of IFA. Currently, only heat treatment is approved for bulk soil; however, this is not a viable option for chemically contaminated soils due to potential volatility of contaminants. Contaminated soils are packaged for shipping in a variety of containers, such as B-25 boxes, cubic yard boxes, super sacks, and 55-gallon drums. The lack of a treatment alternative for contaminated soils limits disposal options.

The Gulfport Lab conducted preliminary work to develop a cold treatment for IFA in soil. IFA workers with associated nest tumulus were placed in 4-ounce plastic cups with lids and placed at various levels in a 4-foot chest freezer filled with local soil placed in plastic bags. A temperature data logger with four temperature probes attached was used to record temperature changes over time. Once the time interval and temperature to kill all IFA had been determined for the small chest freezer, the experiment was moved to a 10-foot commercial portable walk-in freezer. All trials were conducted on the freezer setting that maintained a cold temperature of between -15 degrees Celsius and -20 degrees Celsius. Three trials were conducted using three 30-gallon plastic drums filled with dirt, and three trials were conducted using two 36-inch square corrugated boxes lined with plastic and filled with soil.

These data indicated that the larger the individual soil mass, the longer it takes for the soil to achieve and maintain temperatures required to kill IFA. The soil in the drums cooled more rapidly than the soil in the chest freezer or the boxes due to the tall cylindrical shape of the drums. In all trials where ants died, the soil maintained temperatures below -5 degrees Celsius for a minimum of 36 hours. In the limited trials where a few ants survived, soil temperatures were below -5 degrees Celsius for less than 24 hours. The number of hours to reach -5 degrees Celsius varied from 30–108 hours. Based on this data, we initiated a cooperative agreement with the University of Tennessee. At this point in the project, we began testing bulk soil packaged in a manner similar to contaminated soils and cooled in full-sized refrigerated truck containers. Four types of containers were filled with soil: 55-gallon steel drums, super sacks (38-inch by 38-inch by 38inch woven polypropylene), B-25 boxes (4-foot by 4-foot by 6-foot steel) and cubic yard cardboard boxes (five-wall construction). Ants were buried 6, 12, 18, and 24 inches from the bottom of each container in the center of the container and also at 18 inches from the bottom on the left and right sides of the B-25 boxes.

A 40-foot cold storage trailer was used for this test and set at -15 degrees Fahrenheit (-26 degrees Celsius). Data loggers with external temperature sensors were used to record soil and ambient air temperatures inside the trailer. Nine loggers with two temperature probes each were buried with each cup of ants, and one logger with two temperature probes was suspended in the air. Soil containers were randomly assigned positions within each of the areas in the trailer. Two trials were conducted.

Results from the Tennessee trials were similar to those in the smaller Gulfport trials in that the larger the individual soil mass or better insulated the soil, the longer it takes for the soil to achieve and maintain temperatures required to kill IFA. The tall cylindrical shape of the drums and the woven material of the super sacks allowed more rapid cooling of the soil than either the cubic yard boxes or the B-25 boxes. The number of hours to reach -5 degrees Celsius varied from 17–151 hours, thus requiring a minimum of 7.8 days to effectively treat for IFA. Additional trials will be conducted to validate efficacy with different soil types and moisture levels.

## Molecular Pathway Analysis of Asian Citrus Psyllid, a Vector of Citrus Greening Disease

#### Location: CPHST Mission Laboratory Lead Scientist: Evan Braswell Team Members: David Bartels, Oscar Obregon, Terrance Todd Cooperators: Robert Shatters (USDA-ARS, Fort Pierce, FL)

Citrus greening disease, also known as "huanglongbing" (HLB) disease, is one of the most devastating diseases of citrus. With approximately 500,000 hectares of citrus orchards located in Florida, Texas, Arizona, and California with an annual value of \$1.3 billion at risk, HLB poses a significant threat to the U.S. citrus industry.

As with any non-native pest or disease, the extent of the threat to U.S. agriculture depends on the mode of introduction and the method of spread. While HLB can be introduced to new areas through transportation of infected plant material, more is required for transmission to new plants. The causative agent of HLB disease is transmitted by ACP. This widespread psyllid is thought to have originated in tropical and subtropical Asia and has spread around the world. The current invasive distribution of ACP includes the Saudi Arabian Peninsula, Africa, Reunion and Mauritius Islands, South America, Central America, Mexico, and the United States. HLB disease spreads in new areas only after an ACP population has been introduced.

Given that the movement of the psyllid mediates movement of the disease, we may be able to identify future introduction pathways if we understand how ACP spreads. This will help APHIS exclude HLB from areas that are currently free of the psyllids. Moreover, by understanding psyllid migration between populations, it is possible to improve management strategies of currently infested areas.

To study ACP movement, we worked with ARS, APHIS' International Services program, university researchers, and foreign government officials to build an international network of collectors. This collaboration has generated an ACP repository of more than 7,000 psyllids from over 150 collections. To utilize these collections, we developed methods to explore DNA-based variation among populations of ACP.

Using these methods, we have identified a number of distinct populations from around the world and determined that multiple invasions have occurred in the United States. Further, our analyses identified a timeline for one of these invasions. We found that populations of ACP from the Caribbean share a genetic variant that was not found elsewhere in the world until recently. This variant was not found in the United States in 2005, but 5 years later it made up 11 percent of the population in Texas, indicating a recent invasion. We are working to trace the pathway of this invasion and, to date, find no evidence of it entering from Mexico, moving northward, or moving west from Florida. Further analysis of Texas populations may clarify this invasion. This will include examining 15 new genetic markers we developed to improve the resolution of our analyses. We are currently testing these markers to acquire a more precise picture of ACP population movement.

## Host Range and Preference of Asian Longhorned Beetle

#### Location: CPHST Otis Laboratory Lead Scientists: Baode Wang, Victor Mastro Cooperators: Ruitong Gao, Chuang Song (Chinese Academy of Forestry); Yan Wang, Xiaoming Yang, Yanfang Jin (Baiyin Municipal Forestry Bureau)

The purpose of this project has been to evaluate host plant range of the ALB, *Anoplophora glabripennis*. The studies include evaluations of the insect's utilization of various tree species for feeding and egg deposition as well as suitability of the trees for completion of the life cycle. This work is conducted in China and complements other ongoing assessments of host quality at the Otis Insect Containment Facility and extensive tracking and analysis of host utilization patterns in U.S. ALB program areas. Results of these projects are used by the ALB National Program for determining tree species to survey and insecticide treatments to use against the beetle.

In studies prior to 2011, we confirmed the host status of Acer (maple) species, *Ulmus* (elm), *Salix* (willow), and several *Populus* (poplar) species and characterized the relative preference of the beetle for those trees. We found several additional tree species that could be used by ALB as hosts, including Russian olive, golden rain tree, grey alder, green ash, Chinese ash, and the Japanese pagoda tree. In 2011, we conducted studies in the following areas: (1) evaluation of host preference and suitability test in "common garden" plots of North American and Asian trees; (2) small-plot testing of host preference among trees transplanted into large screen-houses; and (3) surveys of natural and landscape trees to determine which tree species are utilized by ALB in China.

#### **Common-Garden Studies**

Over several years, we established and have maintained two garden plots that include multiple species of North American and Asian trees. One of these "common gardens" is in Baiyin, Gansu, in northwestern China and contains mostly Asian species. The other is in suburban Beijing and focuses primarily on North American trees. These plots have been used for studies to measure the relative acceptability and suitability of various tree species for ALB.

#### Site One: Baiyin, Gansu Province

In 2011, we sought to determine: whether ALB can complete development in tree species that had active egg sites last fall; whether ALB can complete development on the Japanese pagoda tree; and the suitability of Russian olive as an ALB host. The level of the natural ALB population has been very low in the surrounding area since 2008, and no adult beetles were found during the 2011 observations. New egg sites were found on only three species—*Populus alba* var. *pyramidalis*, *P. tomentosa*, and *Ulmus pumila*—although some of the tree species at this site (e.g., maple, willow, and birch) are known to be preferred hosts of ALB.

One significant finding at this site in 2011 was that one female ALB adult emerged from a Japanese pagoda tree that had been caged with paired beetles in the summer of 2008 and subsequently was found to have an active egg site. This is the first record of *Styphnolobium japonicum* as a physiological host of ALB.

To determine the suitability of Russian olive as a host, 15 Russian olive trees were caged with beetles. A total of 243 egg sites were found on 9 trees, but no active sites were found by mid-September. Adult ALB will feed readily on this species, and quite a few egg sites have been found on Russian olive in areas with high ALB populations. We also found a couple of exit holes on this species in 2000 and 2001, but none have been seen since then.

#### Site Two: Beijing

The trees at this site include 20 species from the United States, such as maples and ash, as well as other North American species and a few Asian species. We looked for new egg sites, larval activity, and adult emergence (exit holes). No new exit holes were found on any trees despite the presence of preferred hosts, probably due to the low natural population of ALB in 2010 and 2011.

#### Testing of Tree Species Planted in Screen Houses To Determine Adult ALB Preferences

In Beijing, adjacent to the "common garden" plot, two large screen houses were set up in 2009 and 2010 respectively to compare host preference and acceptance by ALB in an area that was controlled but allowed relatively free movement of the insects. Initially, seven species of trees were planted in the houses: black walnut, paper birch, Chinese golden-rain tree, little-leaf linden, horse chestnut, European hazelnut, and yellow poplar. In 2011, we added trees of five more species: Norway maple, sugar maple, painted maple, grey alder, and poplar. For testing, one pair (one female and one male) of adult beetles was released into the net house at each of the 36 release points. We made three sets of releases at 7- to 10-day intervals in early, mid-, and late July.

Counts of marked beetles on trees at different times of day indicated that ALB adults spent more time on horse chestnut, painted maple, Norway maple, and little-leaf linden than on other species such as gray alder and yellow poplar. The top four species ranked in terms of feeding areas were painted maple, Norway maple, sugar maple, and little-leaf linden. In terms of egg sites made on individual trees: horse chestnut (148) and Chinese golden rain tree (95) had the most, while poplar (36) and painted maple (32) ranked in the middle. No egg sites were found on black walnut or yellow poplar trees. A few active egg sites were found on Chinese golden rain tree, little-leaf linden, and painted maple. No active sites were found on horse chestnut; however, in a previous test, ALB adults emerged from horse chestnut trees. Use of horse chestnut by ALB in North America has been variable, as it has appeared to be a highly preferred host in some program areas (Illinois) but has not been attacked at all in others (Toronto).

Dissections of ALB egg sites were made for trees of 10 species. In terms of the percentage of viable eggs, the tree species can be grouped as: (1) box elder and Norway maple (both more than 80 percent); (2) poplar, horse chestnut, and grey alder (70–80 percent); (3) Chinese golden rain tree and painted maple (50–69 percent); and (4) paper birch, London plane tree, and little-leaf linden (all less than 50 percent).

### Survey of Different Tree Species To Determine Beetle Preference

In addition to the site testing reported above, we also conducted surveys in several locations in China to evaluate ALB preference of tree species. We focused primarily on tree species whose status as the ALB host had not been determined and on species thought to be potential trap trees. We did not find any ALB exit holes on black locust, although there were some egg sites and feeding sites. No ALB exit holes were found on hackberry or rose of Sharon.

To further evaluate whether painted maple or box elder can be used as trap trees in PPQ's ALB eradication programs, as suggested by some researchers, we visually surveyed the number of ALB adults on painted maple, box elder, and weeping willow (*Salix babylonica*) that were growing in close proximity. Many more beetles were found on weeping willow than on painted maple and box elder trees, which were both smaller than the *Salix* trees in these surveys. The results of the surveys indicate that painted maple and box elder are not effective trap trees in areas where large host trees exist.

In addition, we also evaluated whether the Chinese chestnut is an ALB host. There were anecdotal reports that ALB adults feed on the Chinese chestnut trees. We caged 13 pairs of virgin adults with a few Chinese chestnut trees. Although adults survived for 9–10 days on average on this tree species, they did not make any egg pits, and there were no viable eggs found when females were dissected after they were dead. Further evaluation will be carried out in 2012.

## Determination of Effective Field Dose of Chlorantraniliprole (Rynaxypyr®) for Control of Rangeland Grasshoppers

#### Location: CPHST Phoenix Laboratory Lead Scientists: Larry Jech, R. Nelson Foster Team Members: K. Chris Reuter, Lonnie Black

The current treatment options for APHIS-sponsored grasshopper suppression efforts on rangeland are carbaryl, malathion, and diflubenzuron. Carbaryl and malathion have been used for almost 50 years and diflubenzuron has been used since around 2000. Other materials are available but are unattractive because they require high total-application volumes, have label restrictions on grazing and pre-harvest intervals, and have high costs.

A new class of insecticides may offer some alternatives for rangeland grasshopper control. Rynaxypyr® is a member of the anthranilic diamide class of insecticides with a novel target. The anthranilic diamide activates the ryanodine receptor, which results in impaired regulation of muscle contraction. This insecticide has been shown to exhibit a more than 500-fold differential selectivity toward insect over mammalian receptors. Although Rynaxypyr has some contact activity, it is most effective through ingestion of treated material. Rynaxypyr demonstrates both trans-laminar activity and rain-fastness, each critical for residual activity and a plus on rangeland. After exposure to Rynaxypyr, affected insects exhibit rapid cessation of feeding, lethargy, regurgitation, muscle paralysis, and typically die within 1–3 days. The following study was conducted to help develop new treatment alternatives for suppression and control of grasshoppers on rangeland.

Objectives of the study were to:

(1) Ascertain the lowest effective dose of Rynaxypyr for controlling rangeland grasshoppers.

(2) Appraise the following doses: 1.25, 1.5, and 2 fluid ounces of Rynaxypyr in a total volume of 32 fluid ounces per acre treatment aerially applied for control of rangeland grasshoppers.

(3) Assess efficacy following 2 fluid ounces per acre treatments of Rynaxypyr in 100-percent coverage and 50-percent coverage Reduced Agent Area Treatment (RAAT) applications.

(4) Evaluate Rynaxypyr for ease in mixing, calibration, operational aerial application, and cleanup.

In July 2011, 4 treatments of Rynaxpyr were aerially applied to 16 40-acre plots (4 plots per treatment) of grasshopperinfested rangeland in Custer County, MT. Pretreatment densities from individual plots averaged 21.8 grasshoppers per square meter across all precount plots. At the time of treatment, the population included first instars (15.8 percent), second instars (33.9 percent), third instars (37.6 percent), fourth instars (11.5 percent), fifth instars (1.0 percent), and adults (0.1 percent) with a population maturity index of 0.4141 and a total average instar age of 2.48. This population age structure is considered to be ideal for a grasshopper suppression program treatment. The dominant species were: *Melanoplus sanguinipes* (66.1 percent), *Ageneotettix deorum* (14.9 percent), and *Aulocara elliotti* (6.4 percent).

All doses of Rynaxypyr produced substantial and significant reductions in grasshopper populations at all-time intervals compared to untreated populations, which actually increased. A dose rank order of levels of mortality was not evident as all doses produced similar mortality (unadjusted data). When the data was adjusted for the natural change in the untreated populations, populations treated with Rynaxypyr at 2 ounces resulted in 98-percent mortality at 18–20 days after treatment, which was significantly higher than the 80-percent mortality observed in populations treated with 1.5 ounces of Rynaxypyr. No significant difference in mortality occurred among populations treated with 1.25-ounce and 1.5-ounce full-coverage applications and 2.0 ounces of Rynaxypyr in RAAT applications. RAATs are applied at one-half to three-quarters of the standard dose and are applied to 50–80 percent of the infested area in alternating treated and untreated strips. While not significantly different, the 2 ounces of Rynaxypyr applied at full coverage consistently showed numerically higher mortalities (98-percent adjusted mortality) compared to the same dose applied in RAAT applications (85-percent mortality).

In general, with the exception of Dimilin (diflubenzuron), RAAT applications (reduced dose applied at 50- to 80-percent coverage) result in about 15-percent lower mortality than traditional treatments applied at 100-percent coverage. Full coverage, large-scale traditional treatments produce mortality ranging from 90 to 95 percent (malathion), more than 95 percent (carbaryl), and 98–99 percent (diflubenzuron). RAATs of these same chemicals produce mortality in the range of 80 percent (malathion), 80–85 percent (carbaryl), and 98 percent (diflubenzuron). More recently, Dimilin treatments rely on not only a reduced dose but a reduced total volume (12–13 versus 31 fluid ounces per acre) combined with the reduced coverage and produce 78- to 96-percent mortality.

The 80- to 98-percent control produced in this study was sufficient to compete with existing USDA-sponsored treatments against grasshoppers on rangeland. However, the complete economic costs are yet to be determined, depending on industry-assigned costs. No problems were detected during the mixing, calibration, and clean-up phase of the study, and ease of operation in these aspects is extremely important to the utility of this product. The 1.25-ounce dose of Rynaxypyr produced higher mortality than expected compared to other treatments, and an additional dose study is planned to better understand these results. RAAT applications of the two lowest doses should also be studied. Once a reasonable low dose is identified, large operational scale evaluations should be conducted with RAAT and traditional applications.

**Acknowledgements:** The authors would like to acknowledge Daryl Hill for project support and project champions Charlie Brown, Roeland Elliston, and Gary Adams.

## **Supporting U.S. Agricultural Exports**

#### Location: CPHST Plant Epidemiology and Risk Analysis Laboratory (PERAL) Lead Analyst: Lottie Erikson, Export Coordinator Team members: Robert Ahern, Ignacio Baez, Christina Devorshak, Tara Holtz, Walter Gutierrez, Alison Neeley, Scott Redlin, Jim Smith

In March 2010, President Obama signed Executive Order (EO) 13534 (National Export Initiative) with the goal of doubling exports over a 5-year period—from \$1.57 trillion in 2009 to \$3.14 trillion by 2015. The order established an Export Promotion Cabinet (of which the Secretary of Agriculture is a member) and directed that cabinet to assist first-time and current exporters in identifying new export opportunities in international markets. The EO specifically targets reduction of trade barriers. Though PERAL's export support function pre-dates EO 13534, this initiative has given our export mission increased relevance.

Export analysts provide analytical support required to open new export markets and expand or retain existing markets. For example, these staff help to minimize or prevent potential non-tariff trade barriers by reviewing and challenging citations, assumptions, methodology, and conclusions found in an importing country's risk assessments and pest lists. Export analysts also proactively support domestic agriculture industries' bids for new market access, retention, or expansion by developing export pest risk assessments and export pest lists to assure importing countries that a commodity meets legitimate phytosanitary concerns and may be safely traded. Disputes and disagreements between trading partners regarding export requests typically require significant technical/scientific analysis and support.

The PERAL export team works with trade specialists in APHIS, USDA, State governments, other Federal agencies, and industry. The team has expertise in plant pathology, entomology, trade negotiation, and development and application of international standards for phytosanitary measures, as well as considerable experience in the appropriate use of regulatory science. In FY 2011, the PERAL team completed 27 export analyses, which included export risk assessments, export pest lists, reviews of assessments and pest lists developed by trading partners, and ongoing analytical support for an export trade dispute with a major trading partner. There is often a time lag between the completion of an export analysis and the successful conclusion of an export trade issue. For example, past PERAL analyses supported successful conclusions of several export issues in FY 2011: retention of the Chinese market for alfalfa hay (estimated market value: \$2 million); retention of the Egyptian market for baled cotton (estimated market value: \$240 million); and retention of the Kenyan wheat market (estimated market value: \$13 million). In FY 2011, PERAL analysts also developed a risk assessment that supported the successful opening of the Mexican market to U.S. peaches (estimated market value: \$4.25 million) from Georgia and South Carolina under a systems approach.

### Biological Control of the Imported Fire Ant Using Phorid Flies: A Cooperative Rearing and Release Project - 2001–2011

# Location: Biological Control Unit and CPHST Gulfport Lab

Lead Scientist: Anne-Marie Callcott Team Members: Charlie Brown, Katie Hough, Shaharra Usnick, Ron Weeks

Cooperators: Sanford Porter (ARS), George Schneider and staff (Florida Department of Agriculture and Consumer Services), and other State departments of agriculture

Biological control is a process with multiple steps that may take 5–10 years to complete. The process includes foreign exploration to locate potential agents, pre-release assessments of these agents to determine host range (safety) and impact, permit application for field release, development of rearing and release methodologies, implementation, and post-release evaluations of efficacy and possible unintended consequences. As a result, CPHST—or even PPQ—rarely conducts all of these tasks on their own for a given project, but rather partners with other stakeholders who are also concerned with the target pest or weed. IFA biological control is a success story in the making that illustrates how these cooperative efforts can and should work to deliver a cost-effective program, as well as the persistence that is often required to see biological control efforts come to fruition.

IFA are 5–10 times more abundant in the United States than they are in their South American homeland. Escape from the natural biological control agents left behind in South America—including pathogens, viruses, and various parasitoids including at least 24 species of phorid flies—is the most likely explanation for these intercontinental differences in populations. The APHIS phorid fly rearing and release project is part of an effort by many researchers to introduce multiple species of phorid flies to the United States. As a result of these combined efforts, two species of flies, Pseudacteon tricuspis and P. curvatus, are well established across large areas of the Southeast. They are the first classical or self-sustaining biological control agents ever to be successfully established against invasive ants and are among the first released against any invasive social insect. The impact of these flies on IFA is not yet fully evident, and several non-APHIS projects to assess impacts are underway. However, it is believed that phorid flies do contribute to the significant differences in IFA population levels between North America and South America directly through eliminating a small number of worker ants and indirectly through negative impacts on ant foraging.

The APHIS phorid fly project is dependent on the cooperation and collaboration of numerous Federal, State, and university groups for its success. Preliminary research, preparation of permit applications, and the development of rearing techniques are conducted by ARS' Center for Medical, Agricultural, and Veterinary Entomology (ARS-CMAVE) in Gainesville, FL. This technology is then transferred to the Florida Department of Agriculture and Consumer Services (FDACS), which is responsible for mass-rearing phorid flies and shipping them to cooperators for release. Field releases and monitoring of releases are conducted by a variety of personnel in each State, including PPQ, State departments of agriculture, university, extension, and ARS staff. CPHST oversees direction of the project and coordinates releases. The PPQ Eastern Region provides oversight on the cooperative agreement with FDACS.

Highlights of the APHIS phorid fly project:

- APHIS funding initiated through CPHST in 2001 and supported by PPQ.
- Cooperative agreement initiated with FDACS in 2001 to conduct rearing.
- 2001 Pseudacteon tricuspis rearing initiated.
- 2002 P. tricuspis releases begun.
- 2002 *P. curvatus* rearing initiated.
- 2004 *P. curvatus* releases begun.
- 2006 *P. obtusus* rearing initiated.
- 2008 *P. obtusus* releases begun.
- 2010 P. cultellatus rearing initiated.
- 2011 P. cultellatus releases begun.

From 2002 through 2011, there have been 129 field releases totaling more than 1.4 million phorid flies in IFA-quarantined States and Puerto Rico (no releases in New Mexico and only one species released in California). Of these 129 releases, 67 were P. tricuspis, 42 were P. curvatus, 18 were P. obtusus, and 2 were P. cultellatus. Through APHIS releases, along with other Federal and university releases, P. tricuspis is now well established in the southern United States, covering about 50 percent of the IFA-regulated area. To date, P. tricuspis is not known to be established in California, Oklahoma, or Tennessee. The second species, P. curvatus, is well established in all southern IFA-regulated States and Puerto Rico, covering about 65 percent of the regulated area. P. curvatus has not been released in California. Overwinter establishment of the third species P. obtusus has recently been confirmed. The cooperators on the phorid fly project authored an article on the known U.S.-wide distribution of P. tricuspis and P. curvatus that was published in 2011 in the Journal of Insect Science (http://insectscience.org/11.19/). The publication contains a history of the APHIS program, other Federal/State/university release programs, maps depicting distribution in 2008 and expected distribution in 2011, and a discussion of the future of new species releases.

#### CPHST Sterile Insect Technique (SIT) Monitoring Model: Technology Transfer To Support Fruit Fly SIT Control and Regulatory Decisionmaking

#### Location: CPHST Fruit Fly Unit and CPHST Mission Laboratory Project Lead: Hugh Conway Team members: Pedro Rendon, John Stewart, David Dean

The CPHST fruit fly entomologist at CPHST Mission developed a SIT monitoring model to evaluate the effectiveness of the Mexfly sterile releases in the Lower Rio Grande Valley in support of the Mexican Fruit Fly Eradication Program. SIT is a key component for the control and eradication of this fruit fly pest that threatens the multimillion-dollar citrus industry in the valley. The entomologist, in conjunction with other members of the Fruit Fly Unit, developed the model using formulas to calculate sterile and wild fly captures per day, determine sterile fly over-flooding ratios, and recommend adjustments to the fly release rates in the SIT areas. The program release areas were developed into model blocks. Each block was included in the model and the resulting trap recapture can be evaluated by block in order to make adjustments in resource allocation. Prior to the development of the model, the program was unable to make critical assessments of SIT efficiency or effectiveness. Another key utilization of the model involves regulatory decisionmaking in regards to initiating new quarantines. The model allows for quick assessment of the SIT effectiveness at the detection site, the core square mile, and the first buffer of square miles around the site in order to determine the probability of wild females being mated with sterile or wild flies. The model was utilized numerous times to aid in the quarantine decisionmaking process during the 2012 Mexfly outbreak. The SIT monitoring model evaluation will also be transferred to other SIT release areas in the future.

### Detection and Control Program for European Grapevine Moth

#### Location: CPHST LBAM Unit

Lead Scientist: Greg Simmons Team Members: Candace Messner, Jonathon Bishop, Addie Abrams, Saben Kane, Tom Greene, Meghan Brumgard, Stephen Friedt, Kathleen Harding, Amber Reece, Hannah Nadel, Scott Myers, Dave Lance, Vic Mastro

Cooperators: Lucia Varela, Monica Cooper, Robert Van Steenwyk, Rhonda Smith, Max Suckling

EGVM is a key pest of vineyards in the Mediterranean Basin that has recently invaded California. It was first detected in 2009 after it had reached outbreak status, causing crop losses of nearly 100 percent in core areas of the infested regions. EGVM has regulatory significance for both internal and export markets, which depend on the timely movement of grapes within the State, across the United States, and to foreign trading partners. California wine, table grape, grape juice, and raisin production have a combined value greater than \$50 billion. Information from CPHST-initiated projects has provided benefits to all aspects of the EGVM cooperative program to improve and enhance survey, management, and regulatory efforts in order to minimize impacts within California, prevent spread to other areas of the country, and maintain U.S. grower access to international markets.

#### Research on Monitoring, Field and Post-Harvest Treatments, and Degree-Day Models

Work conducted by the University of California (UC) on insecticides has led to refinements on the use of the most

effective materials and the timing of applications to provide better information for growers and program managers to make areawide applications for EGVM control. UC also led work on degree-day modeling to predict the phenology and occurrence of EGVM at different crop stages, which has led to the coordination of uniform areawide applications of both mating disruption and insecticides. Taken together, this effort has resulted in a thousand-fold reduction of EGVM in California (more than 100,000 moths captured in 2010 versus about 100 moths in 2011) and reduced the damage and regulatory impact of this pest on grapes. A project initiated between CPHST, ARS, and UC led to the establishment of a California quarantine EGVM research colony that be used to develop new post-harvest treatments for EGVM on table grapes and other commodities.

#### Monitoring Non-Crop Areas and Alternate Host Plants

CPHST' work on monitoring of native and urban non-crop areas as well as surveys of alternate hosts demonstrated that there is no evidence that EGVM occurs on plants other than grapes or olives or in areas that are distant from grape or olive production. This information suggests that it is unlikely that there are hidden populations of EGVM, and it has helped the control program to focus its efforts on treating grapes and olives and refining the regulatory program for commodities besides grapes.

#### Survivorship During Winemaking Procedures

Several de-stemming, crush, and press experiments were conducted by overseas cooperators as well as UC and CPHST personnel in California. These experiments showed that while some larvae could survive crushing and de-stemming operations, pressing at 1.8 bars caused 100-percent mortality. This work also highlighted the risk of EGVM surviving on winery equipment and demonstrated the need to disinfest such equipment after processing infested grapes. In addition, the work showed that larvae do not survive in red wine must, which allowed for it to be deregulated.

### Methyl Bromide Alternatives: Sulfuryl Fluoride for Log Exports to China

#### Location: Treatment Quality Assurance Unit, Raleigh, NC Lead Scientist: Laura Jeffers Team Members: Scott Wood, Woody Bailey Cooperators: Ellen Thoms, Marty Morgan (Dow AgroSciences LLC)

After the international adoption of The Montreal Protocol on Substances that Deplete the Ozone Layer, the phaseout of methyl bromide production and worldwide use was initiated. Despite the progress that has been made, PPQ still relies heavily on methyl bromide for mitigating the pest risks associated with imported goods. In 2010, more than half a million pounds of methyl bromide were used in PPQsupervised, port-of-entry fumigations on approximately 150 commodities from nearly 100 countries. During the continuing phaseout of methyl bromide, CPHST is charged with identification, development, and validation of alternative treatments, as well as development of techniques to reduce usage during methyl bromide fumigations. While heat treatments and irradiation may prove to be valuable methyl bromide alternatives in the future, CPHST has already successfully developed a fumigation procedure that significantly reduces methyl bromide usage for log fumigations. In the closed-door container fumigation procedure, the amount of fumigant added is based only on the volume of the container. In conventional tarped container fumigations, the amount of fumigant added is based on the entire volume underneath the tarpaulin.

However, PPQ continues to use large amounts of methyl bromide during log fumigations. In 2010, 150,000 pounds of methyl bromide were used for PPQ-supervised export log fumigations, which accounts for 22 percent of the total methyl bromide usage for all PPQ-supervised fumigations. Recently, in addition to methyl bromide and heat treatments, China announced that they would allow entry of hardwood and softwood logs treated with sulfuryl fluoride (at 5–10 degrees Celsius, logs must be treated with 104 grams per cubic meter for 24 hours; at 10 degrees Celsius and above, logs must be treated with 80 grams per cubic meter for 20 hours). Unfortunately, current labeling for many of the cost-effective sulfuryl fluoride formulations does not allow fumigation of logs. In response to industry requests, Dow AgroSciences filed special local need registrations [24(c)] for Virginia, Maryland, New Jersey, Mississippi, and, more recently, for Alabama to allow the use of ProFume for log fumigations. However, without established operational procedures for ProFume, PPQ was hesitant to monitor any log fumigation until several treatment parameters were established. CPHST was tasked to establish standards for:

- Buffer zone around enclosure during fumigation
- Buffer zones for aeration procedures
- Enclosure types
- Safety and monitoring equipment
- ProFume fumigations in the vicinity of methyl bromide fumigations

CPHST was also tasked to provide a guide outlining operational and safety issues for inclusion in the Phytosanitary Export Database. The guide will ultimately be moved to the PPQ Export Program Manual.

For methyl bromide fumigations, PPQ established a 30-foot buffer zone around fumigation enclosures. To work within 30-foot of the fumigation enclosure, PPQ officials must wear approved respiratory protection. The area outside of the 30-foot perimeter is regarded as a safe distance from the fumigation enclosure. However, due to ProFume labeling restrictions, PPQ cannot establish a boundary around the enclosure within a structure during a fumigation. Fumigators cannot legally enter a structure during a ProFume fumigation without approved respiratory protection unless sulfuryl fluoride concentrations are below 1 parts per million.

After reviewing the available ambient air monitoring data for ProFume fumigations of dried fruit and nuts, CPHST decided the existing methyl bromide standards could be used for aeration of ProFume. CPHST worked with Dow AgroSciences to design research protocols to validate the existing aeration procedures. In August 2011, scientists from Dow AgroSciences supervised the ProFume fumigation of hardwood logs in a container (under tarpaulin with container doors open) at the Dundalk warehouse, Port of Baltimore, MD. At the request of the regional PPQ offices, no PPQ staff was onsite. Because there was no PPQ supervision of the ProFume treatment, no phytosanitary certificate was issued. The logs were re-treated with methyl bromide before export to China. Ambient air was monitored around the perimeter of the warehouse during fumigation and aeration, and ambient air was monitored at the 200-foot boundary during aeration. To maximize the possibility of detecting sulfuryl fluoride at monitoring stations, the higher dosage was used (104 grams per cubic meter). Dow AgroSciences reported that no sulfuryl fluoride was detected at any of the monitoring stations during the fumigation. During the first 10 minutes of aeration, negligible amounts of sulfuryl fluoride were recorded outside of the warehouse, but no fumigant was detected at any of the monitoring stations at the 200-foot boundary. After the first 10 minutes of aeration, no fumigant was detected at any of the monitoring stations.

In January 2012, scientists from Dow AgroSciences returned to Dundalk to repeat the ProFume fumigations during cold weather. Dow AgroSciences fumigated eight containers of hardwood (non-oak) or softwood logs (under tarpaulin with container doors open) and seven containers of hardwood (non-oak) or softwood logs (under tarpaulin with container doors closed), using the cold weather schedule (5–10 degrees Celsius, 104 grams per cubic meter for 24 hours). Since PPQ was onsite to supervise the fumigation, a phytosanitary certificate was issued and the logs were shipped to China. Again, ambient air was monitored around the perimeter of the warehouse during fumigation and aeration and at the 200foot boundary during aeration. For this trial, Dow AgroSciences reported that no sulfuryl fluoride was detected at any of the monitoring stations during the fumigation or aeration. CPHST confirmed that the 200-foot aeration boundary established for methyl bromide for the first 10 minutes of aeration is adequate for ProFume fumigations. CPHST approved open- and closed-door container log fumigations for ProFume (eight containers under a tarp maximum).

After reviewing specifications for fumigation monitoring and personal monitoring devices, CPHST decided that methyl bromide and ProFume fumigations can occur simultaneously within a warehouse. If methyl bromide and ProFume occur at the same time, no buffer zone surrounding the methyl bromide enclosure can be established; fumigators and PPQ officials cannot enter the structure without approved respiratory equipment. Since PPQ is considering using sulfuryl fluoride for imports, additional data was gathered during the two field trials. Gas concentrations were monitored within the enclosure (three leads/container) during the fumigation and aeration. For containers fumigated with closed doors, gas concentrations were also monitored underneath containers. As part of this project, CPHST arranged for PPQ officers in New Jersey and Maryland, PPQ regional staff in Raleigh and Fort Collins, PHP staff in Riverdale, and CPHST staff in Raleigh to receive the Dow AgroSciences ProFume training. This training is a prerequisite for all fumigators for ProFume purchase as part of the Dow AgroSciences stewardship program. CPHST has also worked with the training specialists at the PPQ Professional Development Center to incorporate the new procedures into the fumigation workshop. Efforts are underway to develop standards for outdoor tarped container fumigations. Dow plans on filing additional special local need registrations for ProFume in Washington, Missouri, Georgia, Delaware, Massachusetts, North Carolina, South Carolina, and Indiana.

# **Publications**

The following is a list of scientific articles published by CPHST employees in fiscal and calendar year 2011.

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The following is a list of additional publications by CPHST cooperators based on cooperative agreement work.

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# **Scientific Meetings**

CPHST scientists participate in national and international scientific meetings, workshops, and conferences in order to promote the exchange of the latest scientific information on plant protection methods. The following is a list of such meetings attended in 2011.

- American Phytopathological Society Annual Meeting
- American Society for Testing and Materials
- Bi-National Workshop of the Asian Citrus Psyllid for Texas, Tamaulipas, and Nuevo Leon
- Canadian Food Inspection Agency Science Symposium
- Caribbean Food Crops Society Meeting
- Citrus Health Research Forum
- Citrus New Technology Meeting
- Citrus Research Board Asian Citrus Psyllid Scientific
   Workshop and California Citrus Showcase
- Commission on Phytosanitary Measures of the International Plant Protection Convention Annual Meeting
- Continental Dialogue for Forest Pests
- Entomological Society of America Annual Meeting
- Entomological Society of America Pacific Branch Meeting
- Entomological Society of America Southeastern Branch Meeting
- Environmental Systems Research Institute Federal User Conference
- European Food Safety Authority Scientific Colloquium on Emerging Risks in Plant Health
- FAO/IAEA Coordinated Research Project for Development and Evaluation of Improved Strains of Insect Pests for SIT
- Florida Entomological Society Meeting
- IAEA Technical Working Group for Establishment of Phytosanitary Irradiation Treatments and Generic Doses
- Imported Fire Ant Research Conference
- International Acarology Congress
- International Association for the Plant Protection Sciences Annual Meeting
- International Meeting on Radiation Process
- International Pest Risk Mapping Workshop
- International Research Conference on Huanglongbing

- International Research Conference on Methyl Bromide Alternatives and Emission Reductions
- International Symposium on Biological Control of Weeds
- National CAPS Meeting
- National Grasshopper Board Management Meeting
- National Plant Diagnostic Network Meeting
- National Plant Disease Recovery System Workshop
- National Seed Health System Review Meeting
- NCERA-213: Migration and Dispersal of Agriculturally Important Biota
- New England Kiln Driers Association Meeting
- North American Plant Protection Organization Annual Meeting
- Pacific Northwest Stem Rust Working Group
- Potential Invasive Pests Workshop
- Sirex noctilio Research Updates Conference
- Society of Risk Analysis Annual Meeting
- South Dakota Weed and Pest Annual Conference
- Subtropical Plant Science Society Meeting
- Texas Citrus Showcase
- Tri-National Citrus Health Working Group
- USDA Interagency Research Forum on Invasive Species
- W-2185 Western Regional Biological Control Program Annual Meeting
- Weed Science Society of America Annual Meeting
- World Avocado Congress

# Appendix A. List of International Partners/ Collaborators

International partners and collaborators provide support for safeguarding our borders from invasive plant pests. CPHST collaborated with 32 countries on national and global projects.

Argentina
Australia
Austria
Belgium
Brazil
Canada
China
Chile
Colombia
Dominican Republic
Egypt

Germany Guatemala Japan India Ireland Israel Italy Kenya Malaysia New Zealand Mexico Peru Russia Serbia South Africa South Korea Spain Switzerland Thailand Ukraine United Kingdom

# Appendix B. Funded Projects for Fiscal Year 2011

Funding Category	\$ in thousands
AQI-User Fee	11,345
Farm Bill	10,740
Plant Methods	7,792
Emerald Ash Borer	4,624
Biocontrol	3,473
Sirex	1,447
Light Brown Apple Moth	1,434
Pest Detection	1,191
Fruit Fly	697
Asian Longhorned Beetle	620
European Grapevine Moth	500
Grasshopper	498
Sudden Oak Death	249
Imported Fire Ant	210
Cotton Pests	170
Select Agent	150
Gypsy Moth	75
Miscellaneous Pests	70
Noxious Weeds	45
Golden Nematode	20
Total	45,350

Funding lines for CPHST in fiscal year 2011. Includes allocated funds of \$34.6 million and Farm Bill Section 10201 funds of about \$10.7 million.

## **Beltsville Laboratory**

#### **Foreign Plant Germplasm Diagnostics**

- Nepovirus subgroup A molecular detection
- Phytoplasma conventional molecular detection
- Phytoplasma real time nested molecular detection
- Phytoplasma real time one-tube nested molecular detection
- Potato leafroll virus molecular detection
- Sweet potato chlorotic stunt virus East African group molecular detection
- Sweet potato chlorotic stunt virus West African group molecular detection

#### **Citrus Plant Pathogen Diagnostics**

- Citrus leprosis virus-C molecular detection
- *Xylella fastidiosa* strains causing citrus variegated chlorosis molecular detection
- Citrus black spot fungal pathogen *Guignardia citricarpa* molecular detection

- Sweet orange scab fungal pathogen molecular detection
- Elsinoë australis molecular detection

#### **Biotechnology Platforms Evaluation/Adaptation**

- CANARY technology
- Phosphocholine antibody blocking experimentation
- Xylella fastidiosa CANARY system development
- Phytophthora CANARY technology development
- Shipment of CANARY B cells experimentation
- LiNK cartridge DNA/RNA extraction evaluation

#### Proficiency Testing Program for High-Consequence Plant Pathogens

Test panel development

## **Gulfport Laboratory**

#### **Chemistry Methods Development Projects**

- New lure methods development: extraction, instrumental analysis and/or emission rate studies under Farm Bill projects
- Chlor-tetracycline in insects for CPHST-Phoenix (redirected to AQI Lab in July 2011)
- Grasshopper program field spray mix studies and related methods development (redirected to AQI Lab in July 2011)

#### **Imported Fire Ant Projects**

- Biological control of the imported fire ant using phorid flies: cooperative rearing and release program
- Biological control of the imported fire ant: monitoring of field releases of *Thelohania solenopsae* and *Pseudacteon* spp.
- Grass sod and bait treatments for control of imported fire ants (University of Arkansas)
- Development of quarantine treatments for field-grown, balled-and-burlapped (B&B) nursery stock
- New treatments for containerized nursery stock

#### Farm Bill-Funded Projects

- Development of rapid assay kit to identify IFA from other fire ants and to develop a species-specific IFA trap (ARS-CMAVE)
- Analytical support for traps and lures (University of South Alabama)
- Isotope analysis for fruits and vegetables (DHS-CBP Savannah Lab)

## **Fort Collins Laboratory**

#### Digital Identification Aids, Tools, and Resources

- A Resource for California Central Valley Table Grapes (USDA-Foreign Agricultural Service, California Table Grape Commission)
- A Resource for Pests and Diseases of Cultivated Palms: Beetle Screening Tool of Palms (Florida A&M University)
- A Resource for Pests and Diseases of Cultivated Palms: Identifying Commonly Cultivated Palms (Florida Division of Plant Industries)
- A Resource for Pests and Diseases of Cultivated Palms: Mite Screening Tool for Palms (Florida Division of Plant Industries)
- A Resource for Pests and Diseases of Cultivated Palms: Scale Insect Screening Tool of Palms (University of Florida, Southern Plant Diagnostics Network, Florida Division of Plant Industries)
- A Resource for Wood Boring Beetles of the World (University of New Mexico)
- AphID: Identification Guide for Cosmopolitan and Polyphagous Aphid Species (USDA-ARS Systematic Entomology Laboratory, University of Maryland, University of Montreal)
- Bark Beetle Genera of the United States (Colorado State University, USDA Forest Service Rocky Mountain Research Station)
- Citrus ID: Hosts and Potential Hosts for Citrus Pests and Diseases in the United States (North Carolina State University, University of California Riverside)
- Citrus Pests (University of Florida, Southern Plant Diagnostic Network)
- Citrus Resource (Colorado State University)
- Common Nymphal Grasshoppers of the Western United States (USDA-PPQ)
- Dried Botanical ID (Delaware State University)
- Grasshoppers of the Western United States, Edition 4 (Chadron State College, University of Nebraska)
- ID Source (Colorado State University)
- Identification Resource for the Fruit Fly Species of *Anastrepha* (USDA-ARS Systematic Entomology Laboratory, Universidad de Panama, Universidade de São Paulo)
- Identification Tool to Weed Disseminules of California Central Valley Table Grape Production Areas (California Department of Food and Agriculture, Colorado State University)

- IDpic (University of Georgia, Colorado State University)
- Ironclad ID: Diagnosing Ironclad and Cylindrical Bark Beetles (Coleoptera: Zopheridae) of North America North of Mexico (University of New Mexico, Australia CSIRO, New Zealand Collection of Arthropods)
- Oncid ID: Tool for Diagnosing Adult Twig Girdlers (University of New Mexico, USDA-ARS Systematic Entomology Laboratory)
- Pink Bollworm and its Look-Alikes (University of Arizona, Arizona Cotton Research and Protection Council)
- Potentially Invasive Weevil Species from the Caribbean Countries to the United States (Florida A&M University)
- Table Grape Spider ID (University of California Riverside)
- Terrestrial Mollusc Tool (University of Florida)
- TortAI: Diagnostic Tools for Tortricid Moths of Immediate and Future Concern to United States Agriculture (Colorado State University, California Department of Food & Agriculture)
- Wood Boring Beetles of the World: Genera of the Buprestidae (University of California Davis, California Department of Food & Agriculture, Harvard University)

## Farm Bill-Funded Digital Identification Tools

- Citrus Diseases (University of Florida)
- Citrus ID, Edition 2 (North Carolina State University, Colorado State University, University of California Riverside)
- Flat Mites (Acari: Tenuipalpidae) of the World: *Brevipalpus* and *Raoiella* (USDA-ARS Systematic Entomology Laboratory, University of Maryland, Australia's Queensland Museum)
- Identification Tool to Longhorned Beetle Subfamilies and Tribes (Cerambycidae) (University of New Mexico, USDA-ARS Systematic Entomology Laboratory, Australia CSIRO)
- Invasive Ants of the United States (University of Illinois)
- Synopsis of *Diabrotica* (Coleoptera: Chrysomelidae: Galerucinae) of North and Central America: An Interactive Tool to Species (USDA-ARS Systematic Entomology Laboratory, University of Maryland)

## Farm Bill-Funded Decontamination and Waste Disposal

• Investigation of *Phytophthora ramorum* decontamination methods for greenhouse uses (Colorado State University)

- Study of in-field farm equipment decontamination using oxidant biocides (FB)
- Waste disposal for contaminated agricultural waste at our ports and borders (FB)

### **Biological Control**

- Technology transfer and information services in weed management and biological control
- Development of biological control for management of Canada thistle (University of Belgrade)
- Development of biological control for management of Russian knapweed (CABI-Switzerland, Montana State University)
- Development of biological control for management of field bindweed (CABI-Switzerland)
- Development of biological control for management of yellow toadflax and garlic mustard (CABI-Switzerland)
- Development of biological control for management of hoary cress (CABI-Switzerland)
- Development of biological control for management of hound's-tongue (CABI-Switzerland, University of Idaho)
- Development of biological control for management of saltcedars
- Development of biological control for management of dyer's woad and perennial pepperweed (CABI-Switzerland)
- Development of biological control for management of orange and meadow hawkweed (CABI-Switzerland)
- Survey for natural enemies of Canada thistle (CABI-UK)

#### **Spatial Technologies**

• Modeling Asian gypsy moth (AGM) introduction and establishment in Texas

#### Survey Support

• CPHST support for CAPS community surveys of pathogens, nematodes, and invasive weeds

### Weed Control

- Reducing seedbank populations of Benghal dayflower
- Database development for localized spread rates for three invasive weeds
- Efficacy screening study for post-treated resprouts of giant hogweed
- Herbicide efficacy study for common tansy
- Soil fumigation field study for evaluation of methyl bromide alternatives (University of Florida)

# **AQI Laboratory (Miami)**

### **Analytical Chemistry**

- Improving techniques for detecting prohibited plants and invasive insects at ports of entry (ARS and Oak Ridge National Laboratories)
- Chlor-tetracycline in insects for CPHST-Phoenix (redirected from Gulfport Lab in July 2011)
- Grasshopper program field spray mix studies and related methods development (redirected from Gulfport Lab in July 2011)

### **Biological Control**

- Identifying native natural enemies of the red bay ambrosia beetle
- Developing biological control and survey technologies for passionvine mealybug
- Assessing biological control options for Harrisia cactus mealybug in Florida and Puerto Rico
- Offshore safeguarding in the Caribbean: biological control of the coffee mealybug
- Offshore safeguarding in the Caribbean: developing improved trapping protocols for *Tuta absoluta* and understanding host plant relationships of *Anastrepha grandis* in Panama

#### Farm Bill-Funded Projects

- Real time Internet invasive pest identification training
- Improving techniques for detecting prohibited plants and invasive insects at ports-of-Entry
- Mitigating invasive pests in Puerto Rico: A front line initiative for rapid response

## **Otis Laboratory**

#### Pest Survey and Detection

- *Tuta absoluta* pheromone detection and management tools (Isca Tech)
- Development of a novel insect trap for use in detection of invasive plant pests (Technical Precision Plastics)
- Odor-based detection and monitoring systems for exotic pests (also provides lures for CAPS programs)
- Survey and regulatory technology for European grape berry moth *Eupocelia ambiguella*
- Improved analysis and interpretation of insect trapping data

#### Farm Bill-Funded Pest Detection Projects

- Trap and lure for walnut twig beetle
- Walnut twig beetle pheromone and trapping (Forest Service)
- Development of an aggregation pheromone attractant for the walnut twig beetle (UC Davis)
- Development of sex-pheromone-based systems for detection and control of the fruit-piercing moth (ARS)
- Development of detection tools for exotic buprestid beetles (Pennsylvania State University)
- Development of chemical attractants and improved trap designs to facilitate detection of exotic Cerambycidae (UC Riverside)
- Development of attractants for the juniper bark borer and preliminary behavioral study on the mulberry longhorned beetle (Beijing Forestry University)
- Development of chemical attractants for monitoring Monochamus alternatus and Anoplophora chinensis (Fujian Agriculture & Forestry University)
- Tree injections and other methods to improve trapping of buprestid, cerambycid, and Scolytinae borers (Tennessee State Univerity)
- Generic pheromone blends for early detection of longhorn beetles (Canadian Forest Service)
- Accelerated program to develop behaviorally active semiochemicals for survey and control (Virginia Military Institute)
- Enhancing survey techniques for the goldspotted oak borer in southern California (UC Davis)

#### **Forest Pest Management**

- Species of insects attacking North American tree species planted in China – a common garden study (University of Science and Technology China)
- Behavior, biology and control of wood borers (University of Massachusetts)
- Evaluation of soil applications of systemic insecticides for control of wood-boring beetles
- Evaluation of pesticide residues, levels, and distribution in trees treated with systemic insecticides
- Forecasting pest potential through offshore assessments
- Cooperative projects on management of invasive insects (Beijing Forestry University)
- Production of insect diets and life stages for use in APHIS and other cooperative research programs

#### Sirex

- Assessing synthetic pheromone lure and trap effectiveness for the monitoring of *Sirex noctilio* (University of Pretoria)
- Sirex chemical ecology studies (Cornell)
- Biological control of Sirex using entomopathic nematodes
- Behavior and sensory ecology of Sirex noctilio
- Development of monitoring tools for Sirex

#### **Gypsy Moth**

- Studies on the flight capabilities of Asian gypsy moth females and variations of geographical
- Populations of *Dendrolimus* species in Russia (Russian Academy of Science)
- Molecular analysis of male gypsy moths trapped in U.S. ports and other high-risk sites
- Production of nuclear polyhedrosis virus for gypsy moth control (USDA Forest Service)

#### **Asian Longhorned Beetle**

- Potential of nursery treatments for the ALB Eradication Program (University of Science and Technology China)
- Improved rearing technology for ALB
- Control of ALB using insecticides (Chinese Academy of Sciences)
- Infestation dynamics of ALB and EAB in North America
- Insecticide control of exotic wood borers (EAB & ALB)

#### **Emerald Ash Borer (EAB)**

- Optimization of methods for mass-rearing newly introduced parasitoids for biocontrol of EAB (ARS)
- Developing an improved rearing system for EAB (North Carolina State University)
- An integrated comprehensive program to develop EABresistant ash trees (Ohio State University)
- EAB: alternative control strategies biological control
- Development of transgenic North American ash trees expressing a *Bacillus thuringiensis* protein for management of EAB (Purdue University)
- Exploration for EAB biocontrol agents in East Asia
- Nanofabrication of visual lures for EAB (Pennsylvania State University)
- Distribution of EAB in China (Beijing Forestry University)
- Investigating the ecology and natural control of EAB in the Russian Far East (Russia)

- Investigating the ecology and natural control of EAB in South Korea (University of Seoul)
- Biocontrol of EAB exploration and research in China (Chinese Academy of Forestry)
- Development of survey tools for EAB
- EAB studies in eastern Russia

#### **European Grapevine Moth**

- EGVM survey and control technologies (University of California (UC), Davis)
- Developing management tools for EGVM, *Lobesia botrana* (University of Pisa)
- Support for Lobesia botrana program

#### **Treatment Development**

- Development of a methyl bromide schedule for thousand canker disease and its vector walnut twig beetle in eastern black walnut logs (University of Tennessee)
- Survey, detection, and treatment evaluation on insects associated with wood packaging materials (China Inspection and Quarantine)
- Using steam and vacuum to heat treat hardwood veneer logs for export (Virginia Tech)
- Japanese beetle regulatory treatments
- Methyl bromide alternative wood and log treatments for export (EAB and pathogens)

#### Farm Bill-Funded Treatment Projects

- Development of post-harvest quarantine treatments for *Lobesia botrana* and other significant pests
- Methyl bromide alternative treatments for unprocessed wood and log exports
- Development of phytosanitary and regulatory treatments for exotic tephritid fruit flies (IAEA)

### **Phoenix Laboratory**

#### **Rangeland Grasshopper/Mormon Cricket**

- Field evaluation of commercial and experimental pathogenic fungi applied by ground equipment against grasshoppers on Montana rangeland
- Design enhancements for all-terrain vehicle-mounted ground application system for grasshoppers and Mormon crickets on rangeland
- Development of a new active ingredient in bait and

evaluation of a new formulation of the traditional active ingredient in bait for controlling grasshoppers and Mormon cricket on rangeland

- Determination of effective field dose of chlorantraniliprole (Coragen®) for control of rangeland grasshoppers
- Development of fungal pathogens for control of grasshoppers and Mormon crickets (ARS)
- Isolation and characterization of new insect-pathogenic fungi from U.S. soil with high potential for biological control of many insect pests, including grasshoppers/ crickets (Utah State University)

#### **Pink Bollworm**

- Testing of a custom air canon and projectile carrier system for SIT field releases of PBW
- Determination of diapause capacity in APHIS massreared PBW populations
- Comparison of freeze-dried versus freshly extruded PBW diet
- Preliminary observations of progeny from genetically modified, tetracycline-dependent pink bollworm strain OX3402C (homozygous OX3402C x OX3402C, heterozygous OX3402C x APHIS, and a control APHIS x APHIS)
- Sticky and live trapping to capture and characterize dyed and un-dyed adult PBW in the eradication area
- Fecundity of live-trapped PBW rearing facility moths leading to study of spent diet sent to landfills and study to examine numbers of PBW in spent diet leaving the rearing facility
- Continued support of investigations for definitive identification methods for APHIS mass-reared versus wild PBW
- DNA markers for distinguishing released sterile pink bollworm from wild pink bollworm (University of Arizona)
- Pink bollworm eradication program support and technical transfer (Larry Antilla, Phoenix, AZ)
- Trapping study to model pink bollworm population development and movement from non-regulated cottonproducing areas in Texas and New Mexico (Texas AgriLife Extension)

## Plant Epidemiology and Risk Analysis Laboratory

#### **Commodity Pest Risk Analysis**

- Produce scientific documentation in support of trade decisions regarding the importation of commodities
- Prepare pest risk assessments, identify and evaluate potential mitigations, and review pest risk assessments prepared by other countries
- Respond to scientific and technical issues associated with commodity import rulemaking
- Identify and develop improvements in the pest risk assessment and risk management process

#### Exports

- Provide scientific and analytical support to facilitate new market access for U. S. agriculture exports
- Provide scientific and analytical support to the expansion or maintenance of export opportunities that are blocked by technical barriers
- Prepare export risk analysis products (focused on pest lists of arthropods and plant pathogens) associated with commodities for export
- Provide scientific information and analytical support for trade dispute settlement

#### **Risk Analysis for Individual Organisms**

- Through the New Pest Advisory Group (NPAG), assess new and imminent pest introductions into the United States and make recommendations to PPQ management regarding appropriate agency responses to exotic plant pests, including arthropods, mollusks, pathogens, and weeds
- Perform deregulation evaluations for established pests to support policy decisions on pest status for consistency with import actions

# Accreditation and Certification of Risk Analysis Functions

• Through audits and improvements, maintain ISO certification for the lab's commodity risk assessments and NPAG

# Outreach and Training/Capacity Building/Regulatory Curricula

• Provide instructors for a regulatory science minor at North Carolina State University (NCSU)

- Maintain the strong cooperative relationship established between CPHST, NCSU, and other academic institutions
- Support a regulatory curriculum that provides training to students in relevant fields on key aspects of regulatory plant protection
- Host risk analysts from other countries, pairing visiting scientists with resident analysts as mentors to provide training in risk analysis methods

#### Plants for Planting (Q-37) Analyses and Regulatory Overhaul

- Provide scientific and strategic support to revise and update 7 CFR 319.37, the quarantine that regulates the import of plants for planting
- Advance the regulatory process through the development of methodologies and analyses to support the APHIS decisionmaking processes associated with the evaluation of pest risk prior to authorizing the entry of propagative material into the United States
- Prepare risk assessments for propagative material proposed for importation

#### International Standards: International Plant Protection Convention (IPPC) and North American Plant Protection Organization (NAPPO)

- Lend support, time, and expertise to international organizations, such as the IPPC and NAPPO, by participating on international working groups to write standards and review draft standards and the specifications for new standards as they become available
- Manage and maintain the Web site for the Phytosanitary Alert System (PAS) panel, which provides oversight to early warning initiatives for NAPPO

#### Information Systems and Biosurveillance Analysis Forecasting

- Maintain and expand the Global Pest and Disease Database
- Through the NCSU/APHIS Plant Pest Forecast (NAP-PFAST) system, support the predictive pest mapping needs of the Cooperative Agricultural Pest Survey (CAPS) program and the risk assessment activities of the lab
- Generate global plant hardiness maps and post them on the NAPPFAST Web site (www.nappfast.org/)
- Create risk maps for the CAPS top 50 pests as well as for CAPS 2010, CAPS historical pests, and CAPS commodity surveys

- Support the APHIS PPQ CAPS program by producing pest prioritization lists using the analytic hierarchy process
- Produce and circulate the exotic pest information collection and analysis notifications

#### Weed Risk Assessment

- Generate pest lists for weeds
- Conduct weed risk analysis
- In support of the Q-37 revision, revise weed risk assessment guidelines to improve and streamline the process
- Conduct weed risk assessments of plants that pose a risk to the United States as defined by the Plant Protection Act of 2000
- Develop training for staff and others using new weed risk assessment guidelines

#### **Reference Management**

- Maintain and improve physical library (currently 4,000 scientific references, including publications, large-scale maps, and videos)
- Maintain and improve PRA library in Endnote (currently 600 PRAs that are indexed and text searchable)
- Maintain and improve the digital library of scientific references (currently 14,000 documents in PDF format housed in an Endnote library)
- Maintain and improve the Equal Employment Opportunity library (83 topical books and videos are available to CPHST employees through a SharePoint request site)
- Maintain and improve the health and fitness library (currently two shelves of books and videos)

## **Biological Control Unit**

A full listing of the project titles of CPHST scientists working on biological control can be found under the highlights and funded projects sections for the individual CPHST laboratories, with the exceptions of Amy Roda and Scott Weihman in Miami and Pedro Rendón and Carlos Cáceres at the Guatemala Station, who are administered from the Director's Office in Raleigh, NC.

#### **BCU Coordinator Cooperative Agreements**

- Development of biological control and other safeguarding tools to manage invasive pests (Florida A&M University)
- Development of biological control and pheromone

management options for cactus moth (ARS-Georgia/ Florida)

- Development of biological control technologies for cactus moth (Florida DACS)
- Development of biological control technologies for cactus moth (Mississippi State University)
- Winter moth biological control (University of Massachusetts)
- Laplap bug biological control (University of Georgia)
- Organization and implementation of a Technical Resource Group Webinar series on insect rearing (North Carolina State University)

#### Farm Bill-Funded Projects

- Biological control of the brown marmorated stinkbug (ARS-Delaware)
- Biological control of the brown marmorated stinkbug (University of Delaware)
- Biological control of the brown marmorated stinkbug (Michigan State University)
- Biological control of the brown marmorated stinkbug (Oregon Dept. Ag.)
- Biological control of the brown marmorated stinkbug (Florida DACS)
- Development of effective monitoring tools for brown marmorated stinkbug (ARS-West Virginia)
- Development of effective monitoring tools for brown marmorated stinkbug (Vermont University)
- Improvement of mass-rearing rearing techniques for cactus moth (Florida DACS)
- Strategic research on pest threats in the Caribbean pathway (Florida A&M University)

#### Miami

- Identification of native natural enemies of the red bay ambrosia beetle
- Development of biological control and survey technologies for passionvine mealybug
- Assessment of biological control options for Harrisia cactus mealybug in Florida and Puerto Rico
- Offshore safeguarding in the Caribbean: biological control of the coffee mealybug
- Offshore safeguarding in the Caribbean: developing improved trapping protocols for Tuta absoluta and understanding host plant relationships of Anastrepha grandis in Panama

#### **Miami Farm Bill-Funded Projects**

- Real time Internet invasive pest identification training
- Improvement of techniques for detecting prohibited plants and invasive insects at ports-of-entry
- Mitigation of invasive pests in Puerto Rico: A front line initiative for rapid response

#### Guatemala

- Development of mass-rearing techniques for the Medfly egg parasitoid *Fopius ceratitivorous*
- Field testing augmentative releases of *F. ceratitivorous* in combination with SIT for Medfly control

## **Fruit Fly Unit**

#### Headquarters

- CPHST Quality Assurance Survey Program in support of national fruit fly trap and lure procurement
- Fruit systems approaches and regulatory treatments
- Fruit fly host compendium
- Alternatives to diazinon ground drench for fruit fly control

#### Florida Fruit Fly Methods Support

- Evaluation of protein diet augmentation
- Evaluation of agar feeding block hydration
- Optimal tray pupae seeding densities
- Evaluation of sterile fly eclosion tower holding times
- Optimal sterile fly adult chill times
- Testing of improved trap and lure female fly attractants-Cera trap
- Evaluation of combined three-component Bio-Lure cones versus patches
- Evaluation of solid male fruit fly lures versus liquids in Florida
- Fruit fly invasion modeling and eradication program support

#### **Mexican Fruit Fly Support**

- Mexfly detection, development, and support
- Mexfly eradication support
- Methods development and quality control for Mexfly SIT rearing, eclosion, and release
- Fruit fly molecular identification program to support diagnostic and pathway analysis
- Molecular pathway analysis of Anastrepha fruit flies
- Development of diagnostic tools for identifying species and geographic sources of intercepted dacine fruit flies

- Evaluation of long-distance shipment of Mexfly eggs
- Induction of *tsl* strain for Mexfly by means of the utilization of classic genetics
- Assessment of the longevity and quality control parameters of *A. ludens* black pupae strain, as compared with the standard (both sexes) mass-reared strain

#### Hawaii and California Fruit Fly

- Field testing of solid wafer containing all three major male lures (trimed lure, methyl eugenol, and cue lure) in capturing males of *Ceratitis capitata*, *Bactrocera dorsalis*, or *B. cucurbitae*
- Field testing of melolure plugs and liquid cue lure evaluating the effectiveness in capturing male *B. cucurbitae*
- Field test of effectiveness of pre-release exposure to methyl eugenol to sterile males of *Bactrocera dorsalis*
- Testing effect of chill duration on flight ability of sterile male *Ceratitis capitata* at Los Alamitos, CA
- Comparing effect of container-type on results of longevity under stress tests for sterile male *Ceratitis capitata* performed at eclosion facilities
- Measurement of post-release dispersal and swath width of sterile male *Ceratitis capitata* in southern California
- Assessing effect of methyl eugenol on hybridization between putative species within the *Bactrocera dorsalis* complex
- Mating compatibility evaluation between the *Bactrocera cyrptic* species complex

#### Guatemalan Moscamed Operations Support for Fruit Fly Preventative Release and Eradication Programs

- Evaluation of program contract diet ingredients (evaluation of diet ingredients/companies participating in a qualified manufacturers list prior to the respective bidding process)
- Evaluation of LED lighting for the egging rooms/rearing process
- Validation/implementation for larval collection system with water recycling capabilities
- Validation/implementation of naked pupation for maleonly production in combination with water collection prototypes already developed
- Laboratory characterization of Medfly genetic modified strain
- Evaluation of semi-liquid diets; Gel diet evaluation, and others for med fly (cost-benefit analyses)
- Validate/implement the use of hydrochloric acid addition to agar diet preparation process

- Evaluation of the combined effect of ginger root oil and different sterile: wild ratios
- Assess impact of GRO at different doses and exposure times using Worley towers (validation of California and Florida GRO application protocol)
- Assessment of alternative diet types and presentations (agar/semi-liquid/liquid) for emergence towers
- Comparative evaluation of the Worley emergence tower and the locally developed emergence tower for *A. ludens*
- Assessment of new proposed tests for the Quality Control manual
- Development of novel bait stations for fruit fly control
- Field release evaluation of parasitoids in combination with sterile medfly
- Coordinated research project on harmful and beneficial microbes in laboratory rearing of fruit flies (IAEA)

## **Treatment Quality Assurance Unit**

- Commodity Treatment Information System (NCSU NSF Center for Integrated Pest Management)
- Technical Assistance for Specialty Crops (TASC) grant from Foreign Agricultural Service for irradiation research (Chapman University)
- TASC grant for irradiation research (University of Hawaii)
- Grant for the development and maintenance of ASTM standards (American Society for Testing and Materials)

## **Director's Office**

# Farm Bill-Funded Cooperative Agreements for Citrus Health

- Understanding *Guignardia citricarpa* ascospore production and potential inoculum reduction strategies in Florida (University of Florida)
- Engineering mature citrus with DiseaseBlock® to achieve immunity against citrus greening (Integrated Plant Genetics, Inc.)
- Disease modeling via stochastic simulation to test disease control and mitigation strategies to maximize regulatory intervention (University of Cambridge)
- Monoclonal antibody development for CiLV, CVC, and HLB (University of Florida)
- Transmission of *Xyllela fastidiosa* to sweet orange seedlings through infected seed (ARS)

- Stochastic modeling and the design of early detection surveys for high-risk pathogens (Rothamsted Research, UK)
- The effect of physical and chemical treatment on activity of citrus canker lesions and other pests and diseases on fruit and leaves of citrus (University of Florida)
- Methods development for routine assessment of HLB susceptibility (University of Florida)
- Mortality of ACP exposed to heat treatments (ARS)
- Prevention of the spread of citrus black spot outside of southern Florida (University of Florida)

### Farm Bill-Funded Projects at NORS-DUC

- Use of *Trichoderma* to remediate Phytophthora ramoruminfested soil (ARS)
- The risk of asymptomatic *Phytophthora ramorum* infection on fungicide-treated *Rhododendron* (Washington State University)
- Potential efficacy of a copper fungicide for preventing establishment and dissemination of *Phytophthora ramorum* in ornamental plant nurseries (Clemson University)
- Effect of fungicides and biocontrol agents on sporulation and persistence of *Phytophthora ramorum* on nursery hosts (University of California Cooperative Extension, Washington State University)
- Solarization to eliminate *Phytophthora ramorum* from nursery beds (Oregon State University)
- Episodic abiotic stress and ramorum blight in nursery ornamentals: Impacts on symptom expression and chemical management of *Phytophthora ramorum* in *Rhododendron* (University of California)
- Risk of root-to-root spread of *Phytophthora ramorum* in ornamental production nurseries (Washington State University, University of California Cooperative Extension, ARS)
- Development and application of an epidemiological framework for management of *Phytophthora ramorum* on *Rhododendron* in nursery settings (ARS, Oregon State University, University of Cambridge)

# Appendix C. Acronyms and Abbreviations

AAVLD	American Association of Veterinary Laboratory Diagnosticians
ACP	Asian Citrus Psyllid
AGM	Asian Gypsy Moth
ALB	Asian Longhorned Beetle
AMS	Agricultural Marketing Service
AQI	Agricultural Quarantine Inspection
AQI&PT	Agricultural Quarantine Inspection and Port Technology
APHIS	
ARS	Animal and Plant Health Inspection Service Agricultural Research Service
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BCU	Biological Control Unit Cellular Analysis and Natification of Antiona Rich and Vield
CANARY	Cellular Analysis and Notification of Antigen Risk and Yield
CAPS	Cooperative Agricultural Pest Survey
CBP	Customs and Border Protection
CBS	Citrus Black Spot
CHRP	Citrus Health Response Program
CiLV	Citrus Leprosis Virus
CiLV-C	Citrus Leprosis Virus-Cytoplasmic
CMAVE	Center for Medical, Agricultural, and Veterinary Entomology
CPIA	CPHST Project Information Assistant
CPHST	Center for Plant Health Science and Technology
CVC	Citrus Variegated Chlorosis
DHS	U.S. Department of Homeland Security
EAB	Emerald Ash Borer
EDP	Emergency and Domestic Programs
EGVM	European Grapevine Moth
FAS	Foreign Agricultural Service
FY	Fiscal Year
HLB	Huanglongbing
IAEA	International Atomic Energy Association
IFA	Imported Fire Ant
IPPC	International Plant Protection Convention
ISO	International Standards Organization
ITP	Identification Technology Program
LBAM	Light Brown Apple Moth
LiNK	Lincoln Lab Nucleic-Acid Kit
Medfly	Mediterranean Fruit Fly
Mexfly	Mexican Fruit Fly
NAPPFAST	NCSU/APHIS Plant Pest Forecasting System
NAPPO	North American Plant Protection Organization
NORS-DUC	National Ornamental Research Site at Dominican University of California
NPAG	New Pest Advisory Group
NPDN	National Plant Diagnostics Network
NPRG	New Pest Response Guidelines
NPPLAP	National Plant Protection Laboratory Accreditation Program
PBW	Pink Bollworm
PCR	Polymerase Chain Reaction
PERAL	Plant Epidemiology and Risk Analysis Laboratory

PPQ	Plant Protection and Quarantine
PPV	Plum Pox Virus
PRA	Pest Risk Analysis
RAAT	Reduced Agent Area Treatment
RPA	Risk and Pathway Analysis
RRST	Response and Recovery Systems Technology
SDI	Survey, Detection, and Identification
SIT	Sterile Insect Technique
SOS	Sweet Orange Scab
TASC	Technical Assistance for Specialty Crops
TQAU	Treatment Quality Assurance Unit
TWG	Technical Working Group
UC	University of California
USDA	United States Department of Agriculture

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