

# California Agriculture



**Endemic and Invasive  
Pests and Diseases:**  
*How UC and its collaborators detect,  
contain and manage them*





University of California  
Agriculture and Natural Resources

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**COVER:** Across the United States, endemic and invasive pests are a growing problem that costs billions of dollars each year in direct damage and funding for pest management programs. In California, UC ANR researchers collaborate with state and federal agencies to contain and manage pests such as glassy-winged sharpshooter, European grapevine moth and herbicide-resistant weeds. In this photo, Elizabeth Karn, Ph.D. student (left), and Associate Professor Marie Jasieniuk (right) of UC Davis cross glufosinate-resistant *Lolium* (ryegrass) plants with glufosinate-susceptible plants to determine whether the resistance trait can be transmitted to offspring. The results of their research will help inform resistance management strategies. Photo by Will Suckow.

## About this issue

This special issue of *California Agriculture* highlights the UC Division of Agriculture and Natural Resources' work on endemic and invasive pests and diseases that threaten the state's people, agriculture and natural resources. The commitment to research and outreach profiled inside includes the EIPD Strategic Initiative, the UC Statewide IPM Program and historic and ongoing successful collaborations with regulatory agencies and the agricultural community. *California Agriculture* gratefully acknowledges the EIPD leaders for this special issue: Elizabeth Grafton-Cardwell (Cooperative Extension Specialist and Research Entomologist, Department of Entomology, UC Riverside) and Cheryl Wilen (IPM Advisor, UC Statewide IPM Program and UCCE San Diego County).

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# UC and the state of California team up against invasive species



**Elizabeth E. Grafton-Cardwell**

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California Department of Food and Agriculture



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In recent years, the escalation of world travel has rapidly moved pests and pathogens to new regions and allowed them to invade and establish. This places enormous pressure on existing pest management systems and provides many challenges for research, regulatory agencies and outreach. It is estimated that the direct damage costs combined with the costs of controlling invasive species exceeds \$138 billion per year in the United States. Invasive species also impact natural ecosystems, affecting, in the long term, the diversity of our biological systems.

In 2009, as part of UC Agriculture and Natural Resources' (UC ANR) 2025 Strategic Vision, five strategic initiatives were established to focus UC ANR's research and extension programs and increase its ability to address challenges that face California. The goal of the Endemic and Invasive Pests and Diseases (EIPD) Strategic Initiative is to coordinate and support research and extension efforts on new and emerging biological threats to California's healthy food systems, natural resources and communities. The EIPD Initiative focuses on (1) the exclusion of pests and pathogens, (2) rapid response to emerging problems with pests and diseases and (3) integrated management of established pests.

This issue of *California Agriculture* highlights some of those UC research and extension efforts. UC programs studying crops and animal health have provided critical scientific information and outreach to bring pests (insects, nematodes, weeds and vertebrates) and pathogens (viruses, fungi, protozoans and bacteria) under significant control. However, UC does not conduct these efforts alone. The work to combat invasive and endemic species relies heavily on teamwork between UC and state and/or federal regulatory agencies; these collaborations have proved critical for managing pests and pathogens and protecting the state's agriculture industry, homeowners' gardens, California's wildlands and human health.

In particular, UC and the California Department of Food and Agriculture (CDFA) have together created a powerful team to limit the impact of pests and diseases on the state. Section 403 of the California Food and Agriculture Code states that "the Department [CDFA] shall prevent the introduction and spread of injurious insects or animal pests, plant diseases, and noxious weeds." CDFA operates a pest prevention system that includes pest exclusion, pest detection, pest eradication, public information and education, and pest identification and records. Its Plan for Pest Prevention complements UC's EIPD Strategic Initiative; it too focuses on exclusion of pests and pathogens and rapid response to emerging problems. Once pests become established, UC research and

extension programs continue, supported by grant funds from UC ANR, CDFA, commodity groups and other sources.

**Exclusion.** Exclusion includes diagnostics, detection and interception. With an early detection system in place, the eradication of invading pests and diseases may be feasible. Lack of early detection often results in establishment, high management costs and disruption of trade and commerce. In the case of diseases, a lack of early detection may impact human or animal health. UC and CDFA commit major resources to early detection. It's UC ANR's role to provide science-based information to support exclusion strategies and inform policy and to assist with the detection of new pests. This may take the form of assisting regulatory agencies with risk assessments, developing innovative technologies for rapid identification and trapping, developing models of invasion biology for predictive purposes and studying how the changing climate or a local environment influences the introduction of pests and pathogens.

CDFA's Plant Pest Diagnostics Branch (PPDB) scientists provide identification of plant pests and pathogens in their Sacramento laboratory. The PPDB is the largest state plant diagnostic laboratory in the United States. Samples may be collected by first detectors trained through the Western Plant Diagnostic Network (WPDN) housed at UC Davis (page 117) and submitted to a UC Cooperative Extension office or the county agricultural commissioner. Samples not identified at the county level are sent to the Sacramento laboratory. UC ANR scientists, such as Valerie Williamson at UC Davis, play a role in developing diagnostics tools; Williamson is using mitochondrial DNA to identify invasive and endemic root knot nematodes that attack crops.

UC and CDFA have also worked together closely to ensure growers have access to clean planting stock. UC ANR is home to the Citrus Clonal Protection Program, directed by Georgios Vidalakis, and the Foundation Plant Services program, directed by Deborah Golino. These two programs are part of the National Clean Plant Network, and they work closely with CDFA's nursery stock regulatory program to help ensure that California fruit, nut, strawberry and grape nurseries have access to new varieties of pathogen-free propagative material to supply growers.

CDFA Animal Health Branch (AHB) veterinarians provide the expertise in the detection of foreign animal diseases and zoonotic diseases of concern. Working with the UC ANR California Animal Health and Food Safety (CAHFS) laboratory diagnosticians and researchers, they can provide a rapid detection and diagnostic confirmation of livestock and poultry



## It is estimated that the direct damage costs and the costs of controlling invasive species exceeds \$138 billion per year in the United States.

diseases. In the face of the exotic Newcastle disease outbreak in 2002 to 2003 in Southern California poultry farms, UC's CAHFS developed a rapid assay, which reduced the time from sample submission to diagnosis from 5 to 7 days to 1 day and became the primary diagnostic tool to support the successful eradication effort.

In 2012, USDA, CAHFS and AHB together provided personnel to assist in the investigation of a positive bovine spongiform encephalopathy (BSE) cow in the Central Valley. Part of the USDA National Animal Health Laboratory Network, the CAHFS lab is one of six labs in the United States that performs testing for BSE. CAHFS's surveillance testing identified the initial inconclusive sample for BSE, which was then rushed to the National Veterinary Services Laboratory, where it was confirmed to be positive. The investigation was crucial in isolating the cow, confirming there was no risk to human or animal food supplies, assuring foreign delegations that the U.S. safeguards for BSE are strong and ensuring the safety of the country's beef products. The multibillion dollar U.S. beef export market was preserved through these efforts.

**Rapid response to emerging problems.** UC, using its science, outreach and research capabilities, can speedily and efficiently mount programs to tackle emerging threats from new pests and diseases. For example, when the European grapevine moth (EGVM) was first detected in California in 2009, UC quickly developed degree-day models, lists of effective treatments, and commodity-specific treatments for movement of infested fruit and distributed that information and best management practices to growers and homeowners. Even as a threat is emerging, UC can provide provisional treatment guidelines until research can be conducted to provide science-based integrated pest management (IPM) recommendations.

When quarantines and other regulatory measures are deemed necessary to help manage emerging pest and disease problems, UC scientific expertise and local knowledge guide the regulatory decisions made by CDFA. UC input is provided through USDA's technical working groups, CDFA's science advisory panels and ad hoc committees. UC and the regulatory agencies have worked together on many cases of invasive pests and pathogens, including the Asian citrus psyllid/huanglongbing complex, EGVM, diaprepes root weevil, light brown apple moth, red imported fire ant, glassy-winged sharpshooter/Pierce's disease complex, Mediterranean



Elena Zhukova

fruit fly, palm weevils, polyphagous shot hole borer, foot and mouth disease, BSE, avian influenza, exotic Newcastle disease, anthrax, bovine tuberculosis, equine herpes myeloencephalopathy, livestock toxicosis, bovine brucellosis and both land and aquatic invasive plants.

### **Integrated management of established invasive species.**

Once a pest or pathogen becomes established, its ongoing management is a challenge for homeowners, land managers and the agricultural community, and methods must be developed to contain and control it. If it is a new pest or disease, this might require studying the biology of the organism, sampling techniques, the natural enemies that control it in its country of origin and chemical controls (pheromones and pesticides). The UC ANR network of research and extension personnel develop IPM strategies that take into account effectiveness, sustainability, costs and impacts on the environment, and then communicate them to a wide range of clientele, from large-scale commercial growers and ranchers to homeowners. One form that the communication takes is the UC IPM guidelines for California crops, which provide monitoring methods and treatments that are crucial for managing pests and diseases. In some cases, for example the guidelines for managing Fuller rose beetle in citrus, these guidelines provide best management practices that address phyto-sanitary barriers enacted by foreign countries.

CDFA has made strong contributions to the long-term management of pests and diseases through its grant programs.

The Specialty Crops Block Grant program offers grants for projects that enhance the competitiveness of California specialty crops. CDFA also has input on Farm Bill Section 10201 (Plant Pest and Disease Management and Disaster Preparation) project funding priorities. UC ANR has a competitive grant program that funds research and extension programs from the Strategic Initiatives, including EIPD. These grant programs, in combination with other state and federal funds and commodity boards, support the university's excellent research and extension programs.

UC, through its research and extension programs, and CDFA, through its diagnostics and regulatory activities, work together to address critical plant and animal issues facing California. This special issue of *California Agriculture* highlights these and other collaborations and some of the successes of the many pest and disease programs conducted in California. [CA](#)



Elena Zhukova



# Unwelcome arrivals

California's numerous ports of entry and borders provide plenty of opportunities for exotic and invasive insects, weeds and plant diseases to enter the state. Some of these species are intentionally introduced, while others arrive accidentally. Once here, they can benefit from the lack of natural controls of their native environments and thrive in new plant or animal systems and management practices. Here are five significant and new arrivals to California under investigation by UC scientists.

## Polyphagous shot hole borer

In 2012, a South Gate (Los Angeles County) homeowner was searching the Internet to identify mysterious symptoms in her backyard avocado tree. She found Akif Eskalen, UC Cooperative Extension (UCCE) specialist in the Department of Plant Pathology at UC Riverside, and emailed pictures.

"As soon as I got the pictures, I realized it was something I had never seen before," Eskalen said.

He visited the South Gate residence and identified the polyphagous shot hole borer, a tiny beetle from Asia that bores through bark carrying with it a

harmful fungus (*Fusarium euwallacea*). The fungus attacks the tree's vascular tissue, choking off water, causing branch dieback and eventually killing the tree. Polyphagous shot hole borer and the fungus are now distributed widely in more than 110 types of trees in Los Angeles and Orange counties, and have been observed in San Bernardino, Riverside and San Diego counties. So far, infested trees are mostly urban, but agriculturally important trees like olive and persimmon, as well as avocado, are known hosts.

Eskalen is conducting research aimed at defeating the fungus, UC Riverside entomologist Richard Stouthamer is researching the beetle's origin and UC Riverside entomologist Timothy Paine is studying control measures for the beetle. Until solutions to the problem can be found, the



Geovork Arakelian, Los Angeles County

UC Riverside scientists are researching control measures for the polyphagous shot hole borer, an invasive beetle from Asia that bores into trees and transmits a pathogenic fungus.



USDA ARS

In July 2014, the USDA intercepted a shipment of giant African snails (shown above in an official's hand) in Los Angeles. Although the snails haven't reached the wild in California, UC researchers are working with scientists in Florida, where officials are trying to eradicate an infestation discovered in 2010.

scientists are asking the public to help prevent the spread of this pest.

"If you have heavily infested plant material, remove it, chip it and cover it with a plastic tarp to get rid of the colony," Eskalen said. "Never move infested material to non-infested areas."

## Giant African snail

When 68 giant African snails were intercepted at Los Angeles International Airport in July 2014, the news went viral. Gardeners, farmers and researchers were unnerved by a broadly circulated photo showing the size of grapefruits that had been seized in a shipment from Nigeria.

Giant snails have a wide host range and feed voraciously. Besides causing crop damage, the snails can transmit a parasitic nematode that can lead to meningitis in humans.

The giant snails haven't made it into the wild in California, but UC Riverside entomologist Jocelyn Millar is working closely with scientists in Florida, where officials are trying to eradicate an infestation discovered in 2010. Giant snails are difficult to root out because the small, newly hatched snails are hard to find, and when fully grown they are adept at hiding in foliage and leaf litter. Millar, an expert on pheromone attractants, is looking for natural attractants that will draw them out of hiding and into traps. He is working on the project with UC Riverside research specialist Rory McDonnell, who is an expert on snail and slug biology, and Amy Roda, a U.S. Department of Agriculture Animal Plant Health Inspection Service (USDA APHIS) scientist specializing in invasive species.



## Quagga mussel

In 2010, officials were alarmed to learn that quagga mussel had invaded Lake Piru, a reservoir on the Ventura County–Los Angeles County border. A native of Ukraine, the freshwater pest made its way to the U.S. Great Lakes in 1989. In 2007 they were found in Lake Mead and other lakes on the Colorado River. At this time, it is not known how quagga reached Lake Piru. It is the first appearance of the invasive pest in a California water body that is not fed by the Colorado River.

Quagga mussels multiply rapidly, encrusting boat hulls, engines and equipment, and clogging water pipes up to two feet in diameter. They are voracious filter feeders, clearing plankton from lakes that is needed by native species.



Quagga mussels, native to Ukraine, encrust boat hulls, engines and equipment, and clog water pipes.

Greg Giusti, UCCE advisor in Lake County, is working closely with county officials to

keep quagga mussels out of Clear Lake, the largest natural freshwater lake in California. Giusti helped the county

draft an ordinance requiring regular inspections of boats launched in Clear Lake. The local media, the Internet, the

California Department of Fish and Wildlife and the vast fishing

community were part of an outreach effort to make people aware of the regulation. Failing to comply results in a fine up to \$1,000.

D. Hamilton, CDFG

“One thing in our favor is that surrounding lakes are also uninfested,” Giusti said. “If quagga make it to Shasta, Folsom or Berryessa lakes, we’re toast.”

The most common way mussels are moved is recreational boating, but that’s not the only threat.

For example, it is common practice for fire-fighting agencies to dip equipment into lakes to collect water to fight fire. Sabrina Drill, UCCE advisor for Los Angeles and Ventura counties, briefed firefighting officials about ways to prevent spreading the invasive species.

“It might be OK to use water from Lake Piru to fight a fire in Lake Piru’s own watershed,” Drill said. “I shared best management practices for decontaminating the equipment that was in contact with the infested water — starting with making sure everything is drained and completely dried before it is used in another lake.”

## Brown marmorated stink bug

“They’re here,” wrote UCCE advisor Chuck Ingels in a September 2013 email to Sacramento growers. He had just visited the midtown Sacramento site where hundreds of brown marmorated stink bugs (BMSB; *Halyomorpha halys*) were clustered together on a Chinese pistache street tree.

Comments, emails and calls began pouring in. “These bugs were horrible when I lived in Pennsylvania,” commented Todd Jumper on the UC Agriculture and Natural Resources (ANR) website. “Hundreds of them in our home and (it was) literally almost impossible to stop them.”

BMSB is a pest of agricultural crops and a serious residential problem. It is a strong flyer and also travels long distances by hitching rides in vehicles or inside furniture or other articles when they are moved, often in late summer and early fall. As a result, new infestations pop up in neighborhoods where people travel from infested areas.

A native of China, Japan and Korea, BMSB was first documented in the United States in Pennsylvania in 2001. It is either established or found occasionally in about 41 states.

BMSB feeds on apples, pears, cherries, peaches, corn, beans, tomatoes, berries and many other California crops. Feeding creates pock marks and distortions that make the fruit

**“If quagga make it to Shasta, Folsom or Berryessa lakes, we’re toast.”**

— Greg Giusti

**Brown marmorated stink bug, a pest of fruit and vegetable crops, was first discovered in the United States in Pennsylvania. In California, reproducing populations have been found in Los Angeles County and Sacramento.**



Baldo Villegas



**“In certain respects, [the brown marmorated stink bug] is one of the worst invasive pests we’ve ever had in California.”**

—Chuck Ingels

unmarketable. In grapes, berries collapse and rot increases. Wine tasters have been able to detect stink bug odor in wines made from grapes that had as little as one bug per three clusters.

“In certain respects, this is one of the worst invasive pests we’ve ever had in California,” Ingels said.

In summer 2014, Ingels conducted a monitoring program for BMSB in Sacramento County funded by the California Pear Advisory Board and the Lodi Winegrape Commission. Ingels said his team documented the population fluctuations of BMSB in midtown Sacramento. He found no bugs in traps near farms south of Sacramento along the Sacramento River. UCCE advisors Lucia Varela and Rachel Elkins, also with funding from the Pear Advisory Board, conducted BMSB monitoring programs in Lake and Mendocino counties; none were found.

“They don’t seem to be spreading fast,” Ingels said. “We’ve learned that it takes four or five years for a major reproducing population to build up. That suggests they could be on farms, but we haven’t found them yet.”

In the meantime, problems persist in midtown Sacramento.

“Restaurants are concerned that the stink bugs are visible on the walls and crawling on tables. A couple of apartment managers called me to say their tenants are threatening to move out if they are not controlled,” Ingels said. “They are a nuisance, so they are already having an economic impact.”

### Downy mildew

In the plant disease world, the introduction of new pests is more ambiguous than with insects. In Imperial County, for example, farmers are dealing with a new race of downy mildew in spinach (*Peronospora farinosa* f. sp. *spinaciae*).

“We don’t know where it may have come from,” said Steven Koike, UCCE plant pathologist for Monterey and Santa Cruz counties. “It could have

come in from other countries, or it might have mutated here in California. There’s some suspicion viable downy mildew is carried on seed, but the evidence at this point is all circumstantial.”

They do know an Imperial Valley grower noticed bright yellow blotches staining spinach leaves in 2012. It wasn’t long before samples were delivered to Koike, who operates UCCE’s only county-based plant diagnostic lab.

“We have developed a very strong relationship with industry over the years,” Koike said. “When growers start to see some breakdown in previously resistant varieties, they get in touch with me.”


Downy mildew will not grow in petri dishes, only on living tissue. Koike and his team washed the spores off the sample leaves and sprayed them onto 12 varieties of spinach in the lab. They assign a plus or minus to each variety based on which come down with the disease and which stay healthy.

“This creates a fingerprint of the pathogen,” Koike said. “We match the fingerprint to see if it is a known race. In this case, it was a brand new fingerprint, so we realized we likely had a new race.”

**“When growers start to see some breakdown in previously resistant varieties, they get in touch with me.”**

—Steven Koike

The same test was conducted by James Correll, a plant pathologist at the University of Arkansas and Koike’s frequent collaborator, who reached the same conclusion. In 2014, the International Working Group on *Peronospora farinosa* (IWGP) in The Netherlands designated Koike and Correll’s isolate as race Pfs: 15.

The IWGP promotes consistent and clear communication between public and private entities — scientists, farmers, plant breeders and others — about all resistance-breaking races that affect a wide area and cause significant economic impact. Koike and Correll’s work contributed to this international effort. 

—Jeannette Warnert



Stephen Ausmus, USDA ARS

The new race of downy mildew stains spinach leaves with bright yellow blotches.



Steven T. Koike



# A look at EIPD Strategic Initiative projects

**UC** ANR's five strategic initiatives seek new ways of partnering within and outside the university to tackle emerging issues in California. As part of this strategic vision, the Division annually funds research and extension projects in and across all initiatives. Four recent and ongoing studies in the Endemic and Invasive Pests and Diseases Strategic Initiative are profiled here.

## Thousand cankers disease

**T**housand cankers disease "is a rapidly emerging invasive threat to walnut orchards in California," says Richard Bostock, plant pathologist at UC Davis and a lead researcher on a 3-year study of the disease. "Only in a few places in the state have English walnut trees died due solely to this disease," he says, "but it's on its way to becoming endemic in all the walnut-growing areas, and we are still not sure where this is going to go."

The fungal pathogen, *Geosmithia morbida*, was new to science when it was found in California in 2008 in Northern California black walnut trees growing in Davis. Its vector, the walnut twig beetle (WTB), has been known in California since 1959. The male beetles attack a tree first, releasing a pheromone that attracts thousands of beetles to the phloem

tissue of the tree's trunk and large branches, where they tunnel, feed and produce offspring. Spores of the pathogen are carried on the vector's body and infect the host tissue; the cankers produced by the pathogen coalesce and can girdle a trunk or branch, and in severe cases can cause decline and death of the tree.

The study, reaching completion now and co-directed by Steven Seybold, a USDA Forest Service entomologist, has assessed the distribution of thousand cankers disease in the state's walnut-growing areas, differences among walnut (*Juglans*) species in their reaction to the pathogen and attractiveness to

the beetle, the genetic diversity of the fungus and the biology of the beetle. The disease appears to be present in all areas; in some orchards, there is a very low incidence of the disease, while in others, as many as 90% of the trees have WTB attacks and cankers.

Thousand cankers disease has decimated eastern black walnut, *Juglans nigra*, in landscape and urban plantings in some Western states and parts of Colorado, where, in 2008, the association of the beetle and fungal pathogen in the disease was first discovered and reported by researchers at Colorado State University. In 2010, the disease was reported on eastern black walnut in Tennessee and subsequently in other Eastern states within the native range of this species. In California, the research team has documented the disease in the Southern California black walnut, *Juglans californica*, in English walnut, *Juglans regia*, which is the commercial orchard species, and in Northern California black walnut, *Juglans hindsii*, which is used as a rootstock for *J. regia*, and also in Paradox rootstocks. "About 70% of commercial trees are on Paradox," says Bostock, and the study has discovered that "Paradox is highly susceptible and is very attractive to the beetles." Investigating the population structure of the fungus in different geographic regions in the United States, other researchers have reported "a great deal of diversity in the fungal population in Arizona, California and New Mexico," says Bostock, "and their findings suggest that these states could be the source of the thousand cankers disease epidemic."

The susceptibility of Paradox and the destruction the disease has caused in eastern black walnut are cause for concern, but "we are not raising a lot of

Stacy Hishinuma



Above, a heavily infected Southern California black walnut (*Juglans californica*) showing sap-staining of the bark and dieback. Inset, a canker underneath the outer bark of a Paradox hybrid walnut showing necrosis around walnut twig beetle tunnels.



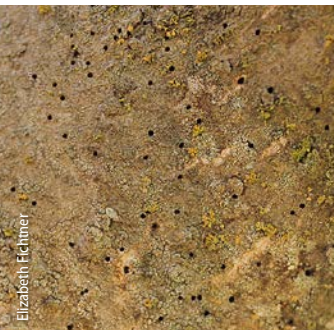
Richard Bostock



Steve Seybold and Stacy Hishinuma  
Tatiana Roubtsova

Top, culture of *Geosmithia morbida*, the fungal pathogen that causes thousand cankers disease. Middle, close-up of walnut twig beetles tunneling in the bark of an infected tree. Bottom, actual size of the beetle in relation to a penny.





Characteristic entry and exit holes of the walnut twig beetle in the trunk of an English walnut tree in Tulare County. The holes are less than 1 millimeter in diameter.

alarm bells at this point," says Bostock. Researchers suspect thousand cankers disease may be contributing to the complex of diseases, including Phytophthora root and crown rot, lethal Paradox canker and crown gall, that cause decline in walnut trees. Thousand cankers disease has been found in the stems of even very young vigorous English walnut trees, and yet it seems the trees can grow out of it. However, when it begins in the crowns of older trees and progresses downward, it is often fatal.

The current suggested treatment for severely diseased trees is removal and burning on site, if burning is allowed. Study team members Elizabeth Fichtner (UCCE Tulare County) and Seybold have found that the majority of WTBs begin to emerge from infested wood by late spring, suggesting that tree removal and sanitation should be conducted by March, or April at the latest. The beetles fly nearly year-round in California, but the primary flights are in May/June and September/October.

Entirely unknown is why thousand cankers disease apparently emerged recently in California and spread so fast. The beetle has been reported in the state for decades but has not been considered a significant pest; however, this insect-pathogen association presents a new challenge for growers, whose 2013 crop was valued at nearly \$1.8 billion according to the USDA National Agricultural Statistics Service. "Something has changed with these beetles and their association with this pathogen that has led to the widespread occurrence of the disease we are now seeing in the state," says Bostock.

The research team also included Mohammad Yaghmour (Plant Pathology, UC Davis), Stacy Hishinuma (Entomology, UC Davis), Tiwonne Nguyen (Plant Pathology, UC Davis), Tatiana Roubtsova (Plant Pathology, UC Davis), Carolyn DeBuse (USDA-ARS), Mary Louise Flint (UC IPM), Janine Hasey (UCCE Sutter-Yuba counties) and Richard Hoenisch (Plant Pathology, UC Davis).

### Asian citrus psyllid and huanglongbing disease

The bacterial pathogen that causes the citrus disease huanglongbing (HLB) is on its way to California; it is vectored by the Asian citrus psyllid (ACP), which is established in Southern California and has moved into citrus orchards in the Central Valley. "There is currently no cure for this disease," explains Beth Grafton-Cardwell, UCCE specialist and research entomologist at UC Riverside and lead researcher on a 5-year project to alert and prepare growers and the public. "HLB kills citrus trees, and it's spreading northward from Mexico fast," she says.

The symptoms of HLB are progressive mottling of leaves, deformed and off-flavor fruit, plant stunting and eventual plant death. HLB was first identified in Florida in 2005, and citrus acreage and fruit yields in that state have dropped precipitously since then, according to Grafton-Cardwell.

ACP spreads the disease quickly; the nymphs of the insect can feed on infected tissue after they hatch and carry the disease to other trees when they emerge as adults. "ACP is a very efficient vector, so not many psyllids are needed to spread the disease," says Grafton-Cardwell. And yet, the disease symptoms don't show up in the trees for 1 to 2 years after infection, by which time infection has already spread. In some areas of Texas, she says, "hot psyllids"

infected with the pathogen have been found, "but the trees are not showing infection yet."

The project team's goals have been to create and keep up to date a "one-stop" online information hub that provides California growers and homeowners with what they need to know to help delay the arrival of HLB. In the first year of the project, 2012, the team set up the ACP/HLB Distribution and Management website, [ucanr.edu/sites/acp/](http://ucanr.edu/sites/acp/). This site provides users with a Google map to locate their property in relation to the reported findings of ACP, HLB and releases of the biological control wasp *Tamarixia*. They are also provided guidelines on how to protect their trees.

The map currently shows a single red circle in Los Angeles, which signifies the only documented instance to date of HLB in California. It was recorded in 2012 in an urban backyard citrus tree. That incidence of the disease "was eliminated," says Grafton-Cardwell, "but we are afraid HLB will turn up somewhere again soon."

Most of the California catches of ACP, plotted by square mile on the website map, are in urban backyard trees in Southern California. It's estimated that 60% of Californians have a citrus tree. Statewide, the number of citrus trees in residential gardens exceeds the number in commercial orchards. The website gives homeowners information on identifying the psyllid,



Symptoms of huanglongbing disease, also known as citrus greening, include mottling of leaves and greening of fruit.



Adult Asian citrus psyllids (actual size).



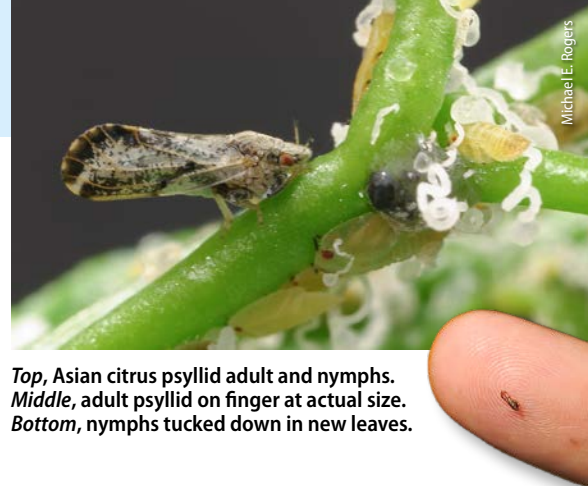
and directions on what to do if an ACP is found: Act fast! Call the California Department of Food and Agriculture (CDFA) hotline, 1-800-491-1899, to report the finding; if it's a finding in a new area, CDFA provides eradication treatment, but if ACP is already established in the area, the homeowner is directed to manage the psyllids on their own.

Economist Karen Jetter (Agricultural Issues Center, UC Davis) has created tables on the website showing the costs of various treatments for both homeowners and growers. A cost calculator helps conventional and organic growers compare the cost per acre of various chemical options. "We are in the process of surveying growers in various regions of the state to determine what chemical choices they have made over the last 4 years, so that we can provide an analysis of costs by region," says Grafton-Cardwell.

For outreach, the project team is providing training to the general public through Master Gardeners

and retail nurseries. The team works with local grower task forces in citrus areas infested with ACP, such as the Imperial Valley, Ventura County and the San Joaquin Valley, helping them to develop ACP management strategies. The website is used as an educational tool during these meetings. Grafton-Cardwell communicates a clear message that insecticide spraying to slow the spread of ACP and keep numbers low is critical for preventing psyllids from finding diseased trees and helps to buy time for researchers to develop a cure for the disease.

*Other members of the project team are Matthew Daugherty (Entomology, UC Riverside) and Robert Johnson (UC ANR Informatics and GIS Program).*



Top, Asian citrus psyllid adult and nymphs. Middle, adult psyllid on finger at actual size. Bottom, nymphs tucked down in new leaves.



Elizabeth Grafton-Cardwell: Above and fingertip

## Bovine respiratory disease

Bovine respiratory disease is one of the leading causes of death in preweaned dairy heifers in the United States. To help dairy farmers diagnose and control the disease, a new scoring system to assess the respiratory health of preweaned dairy calves has been developed in research led by Sharif Aly at the Veterinary Medicine Teaching and Research Center, UC Davis. Called the California Bovine Respiratory Disease Scoring System, it is composed of six questions requiring only yes/no answers and can be easily used on-farm by owners, veterinarians and farmworkers.

The 4-year study began in 2012. The goals of the research were to collect data on current management practices and to develop a simple, low-cost clinical diagnostic scoring system that could be used on farms to identify clinical signs early, avoid unnecessary animal deaths and misuse of antimicrobials, and track the health of calves over time.

A survey questionnaire was sent to 1,450 dairies in 2012 with questions on calf management practices. The data was statistically analyzed to identify the most important factors that may be related to the risk of bovine respiratory disease. Then, a model-based scoring system was developed for six clinical signs: eye discharge (2), nasal discharge (4), ear droop or head tilt (5), spontaneous cough (2), rapid or difficult breathing compared to other calves (2) and temperature at or above 102.5°F (2). A score of 5 or higher suggests a calf may have respiratory disease.

The scoring system has been validated in a study of 500 calves on three dairy farms and two calf ranches in California; results are expected to be published in 2015. One of the advantages of the California scoring system compared to the Wisconsin scoring system, developed in 2008, is that it requires less calf handling and allows easier assessment of clinical signs. A recent study compared the diagnoses of both systems and showed excellent agreement beyond chance.

Aly stresses how easy it is to use the California scoring system. "We've had very good feedback from dairies," he says. "Assessing each clinical sign as present or absent takes the guesswork out of diagnosis. In addition, the scores and cut-off for case status are

UC DAVIS VETERINARY MEDICAL CENTER		UC CE University of California Agriculture and Natural Resources		Cooperative Extension		UC DAVIS ANIMAL SCIENCE	
<b>Bovine respiratory disease scoring system for pre-weaned dairy calves<sup>1,2</sup></b>							
Clinical sign	Score if normal	Score if abnormal (any severity) <sup>3</sup>					
Eye discharge	0		2				
Nasal discharge	0		4				
Ear droop or Head tilt	0		5				
Cough	0	No cough	2	Spontaneous cough			
Breathing	0	Normal	2	Rapid or difficult breathing			
Temperature	0	< 102.5° F	2	≥ 102.5° F			
Add scores for all clinical signs. If total score is ≥ 5, calf may be positive for bovine respiratory disease							
<small><sup>1</sup> Luke J. Leffler, PhD, Ross P. Ross, MS, 1985. Development of a novel clinical scoring system for the diagnosis of bovine respiratory disease in pre-weaned dairy calves. <i>Proc 2012 ISAE Conference</i>.</small>							
<small><sup>2</sup> Aly S, Lohr J, Williams D, Leffler L, Ross P, 2014. Development of a novel clinical scoring system for the diagnosis of bovine respiratory disease in pre-weaned dairy calves. <i>Am J Vet Res</i> 75: 117-121.</small>							
<small><sup>3</sup> Any abnormality (redness, but not listed), the examples shown in the above pictures.</small>							

**The California Bovine Respiratory Disease Scoring System allows owners, vets and farmworkers to easily diagnose respiratory disease in preweaned calves.**



Dairy Epidemiology Lab, UC Davis

model based.” Because it is so simple, the scoring system can be used daily, for example, on sick calves to confirm respiratory disease status prior to treatment.

The team’s current research is focused on developing a risk assessment tool that incorporates the new scoring system and also a questionnaire, based on the statewide survey, that points to risk mitigation strategies. The questionnaire can be completed by a calf raiser and the herd veterinarian to form a disease control plan. The scoring system can be used to monitor the prevalence of respiratory disease in calves once control measures are implemented. The risk assessment tool will be used to monitor 5,000 calves on

different dairies for the risk of bovine respiratory disease in collaboration with dairy farm advisors and UC Cooperative Extension specialists.

The research team also includes Terry Lehenbauer and Alison Van Eenennaam (Animal Science, UC Davis), Randall Anderson (CDEA), Alejandro Castillo (UCCE Merced County), Carol Collar (UCCE Kings County), Christiana Drake (Statistics, UC Davis), Jennifer Heguy (UCCE Stanislaus County), Lindsey Hulbert (Animal Science, UC Davis), Betsy Karle (UCCE Glenn County), Frank Mitloehner (Animal Science, UC Davis), Nyles Peterson (UCCE San Bernadino County) and Noelia Silva-del-Rio (UCCE Tulare County).

### Pest and disease risks in compostable wastes

California municipal waste plans and air quality regulations are shifting green waste processing from composting operations, where materials must be heated to at least 122°F, at which few pathogens survive, to chip and grind operations, where pile temperatures may not exceed 122°F. “The question,” says David Crohn, associate professor of environmental science, UC Riverside, “is whether there’s a potential for pests and diseases, such as the Asian citrus psyllid, to survive chipping and grinding and be spread to disparate locations.”

The preliminary results of a recent research project suggest there is that risk.

Assembly Bill 341, passed in 2012, required state government to plan for 75% of municipal waste to be diverted from landfills by 2020. This will be possible only if millions of tons of green waste are recycled. As stricter environmental regulations increase the cost of opening and operating compost operations, more green waste will be going to chip and grind operations; and the resulting material can be moved out of the facilities within a few days, transported over sometimes long distances and applied in diverse locations: orchards, parks, gardens, farms, commercial landscapes and roadsides. It is usually applied as a surface

mulch, which can allow a pest or disease to be easily spread by wind, water or wildlife.

Crohn, who is a waste management specialist, and a team of other UC specialists, farm advisors and graduate students set up simulated composting and chip and grind environments and compared the survival rates of Asian citrus psyllid, citrus leafminer larvae, tobacco mosaic virus, fusarium in palms, bermudagrass, nutsedge, and clover and tomato seeds. In each environment, pest and disease samples were either placed in mesh bags or in envelopes in the green waste material, and either the material was heated to 122°F (i.e., composted) or kept at 77°F, ambient temperature, for 3 days.

“The results were a mix,” says Crohn. “Citrus leafminer was controlled by chipping and grinding; Asian citrus psyllid, though, had some survival in chipping and grinding but was controlled by composting.” In terms of the weeds, chipping and grinding “was effective at inactivating nutsedge, but clover and tomato seed survived it just fine.”

Crohn stresses this was a small study, and it is impossible to simulate the variety of conditions in commercial green waste operations. But in terms of the risks of invasive pests and diseases being spread more easily and rapidly through chipping and grinding, the preliminary results, he says, “suggest there’s reason for concern.”

The next step of the research is to analyze the economic impact of the state regulatory policies on composting and chip and grind operations. Chip and grind operations are exempt from costly new rules to protect air and water quality; composting facilities are not exempt.

The project team also includes Matthew Daugherty (Entomology, UC Riverside), James Downer and Ben Faber (UCCE Ventura County), Deborah Matthews (Plant Pathology, UC Riverside) and Steven Swain (UCCE Marin County). [CA](#)

—Hazel White



A UC study found that some insects and weeds survived grinding (left) and chipping (right) operations.



Asian citrus psyllid nymphs (left) and citrus leafminer damage (right).



## EXCLUDING PESTS AND PATHOGENS

The optimal way to control new biological threats to agriculture, natural resources, communities and human health is to prevent them from arriving in the first place. If detected and intercepted early enough, a pest or disease may be eradicated completely. This ability to quickly detect and identify a new arrival depends on having diagnostic expertise, infrastructure and resources in place. The articles in this section highlight UC's work and partnerships in this critical stage of protecting livestock and plant systems.



# Diagnostics in animal health: How UC helps exclude and minimize impact of livestock pathogens

by John M. Adaska, Edward R. Atwill and Glenn A. Nader

*UC has a wide reach in the agriculture sector of the California economy and is well recognized for research expertise in plant diseases. Less well known is the role UC plays in animal agriculture. In 2012, the California Animal Health and Food Safety lab at UC Davis performed nearly 980,000 tests on samples from sick livestock, including cattle, horses, pigs, chickens and turkeys. The lab is prepared to respond rapidly to any disease outbreak or identification of a foreign disease. Researchers at the School of Veterinary Medicine at UC Davis are testing novel subunit vaccines to prevent pinkeye in cattle; UC ANR specialists and advisors and the staff at the Sierra Foothill Research and Extension Center were key to the development of best management practices that landowners and resource managers are using to protect their herds and public water sources against the parasite *Cryptosporidium parvum*; and UC veterinary scientists are part of a large team of experts, including state and federal agencies, determined to combat the endemic bluetongue virus, which can affect the state's exports.*

Cash receipts for California's total livestock and livestock products were \$12.3 billion in 2011 (table 1). The responsibility for protecting the livestock industries and, by extension, food safety and security, in the state falls on an integrated system that starts on the farm and extends to a wide array of private and public entities. UC has a large role in outreach, education, surveillance and response to significant animal disease events, and faculty and staff at each of the UC campuses are involved.

### The CAHFS laboratory system

The California Animal Health and Food Safety (CAHFS) laboratory system is funded in large part by the California Department of Food and Agriculture (CDFA) and operates within the UC Davis School of Veterinary Medicine (SVM). CAHFS is the backbone of California's early warning system to safeguard public health from foodborne pathogens, toxins and diseases common to animals and humans. CAHFS also protects the health of California's livestock and poultry populations by providing broad-based surveillance for all catastrophic animal diseases not currently found in the United States (referred to as foreign animal diseases, or FADs).

CAHFS must immediately detect an introduction of a highly contagious disease like avian influenza in poultry and foot and mouth disease in livestock to minimize the devastating effects it would inflict on consumer confidence and California's agricultural economy. As a result, CAHFS is responsible for the vast



UC campuses and UC Cooperative Extension work with the CAHFS lab system, funded largely by CDFA, to research livestock diseases, protect animal health and respond to disease outbreaks.

Online: <http://californiaagriculture.ucanr.edu/landingpage.cfm?article=ca.v068n04p109&fulltext=yes>  
doi: 10.3733/ca.v068n04p109

**TABLE 1. California livestock, dairy, poultry and apiary income, 2010 and 2011**

	2010	2011	Change
	..... \$1,000s.....		%
Aquaculture	58,200	64,036	10
Chickens, all	721,724	702,051	-3
Cattle and calves	2,068,412	2,825,125	37
Eggs, chicken	367,788	391,578	7
Hogs and pigs	36,063	39,196	9
Honey	42,579	28,594	-33
Milk and cream	5,928,150	7,680,566	30
Sheep and lambs	66,060	NA	NA
Turkeys	262,910	287,463	9
Wool and mohair	3,835	5,050	32
Other livestock	218,617	269,345	23
<b>Total</b>	<b>9,819,519</b>	<b>12,357,994</b>	<b>26</b>

Source: USDA NASS 2012.

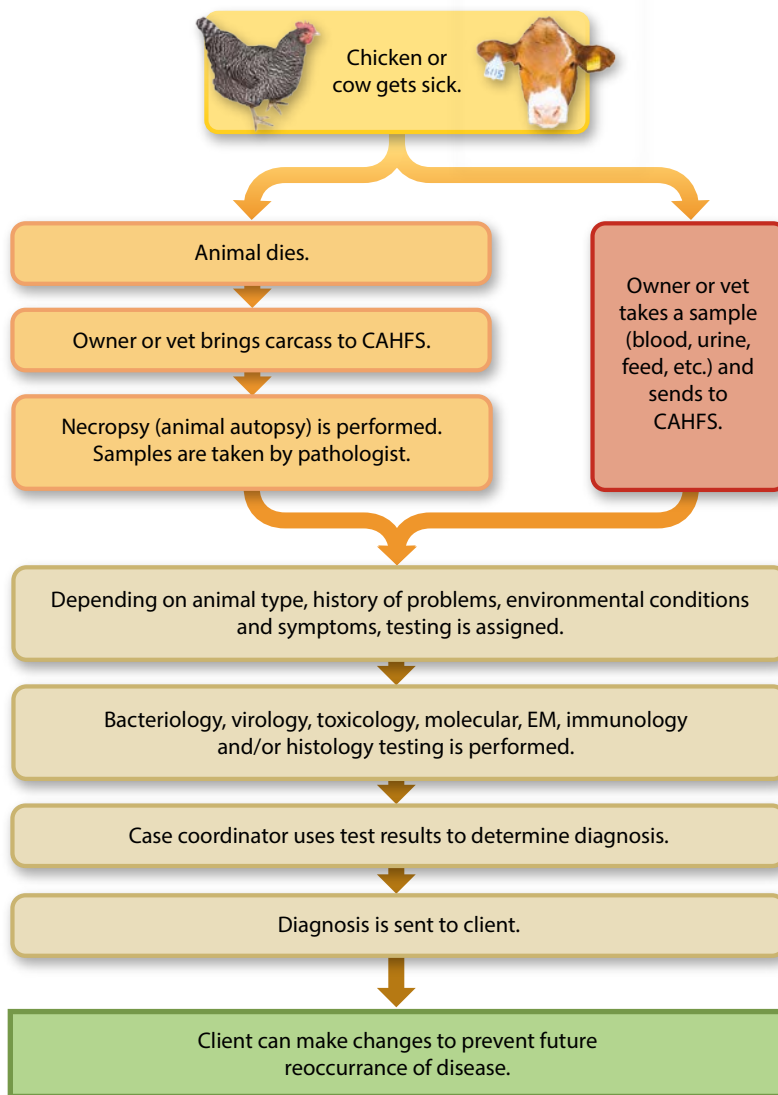
majority of diagnostic testing of livestock samples within the state. In 2012, the lab system had 30,567 cases submitted to it (4,255 equine, 6,952 bovine, 1,855 caprine, 340 porcine, 819 ovine, 2,064 other mammals, 3,063 chicken, 477 turkey, 1,114 other avian, and 9,805 cases in which a species was not provided, involving feed or bedding samples, etc.) and performed nearly 980,000 tests. The case load that year included over 10,800 necropsies (animal autopsies) of whole animals or animal tissues submitted for full diagnostic work-up (E. Sanson-Smith, CAHFS, personal communication). The work-ups typically include post-mortem and microscopic examination, bacterial cultures, testing for viral infections, testing for antibody levels and, in some cases, testing for the presence of toxic agents.

CAHFS is a member of a number of important national diagnostic networks, including the National Animal Health Laboratory Network (NAHLN) (<https://www.nahln.org>) and the Food Emergency Response Network (FERN) ([fernlab.org/](http://fernlab.org/)). NAHLN is a partnership of federal, university and state veterinary diagnostic laboratories across the United States and is a surveillance and emergency response system that provides critical and ongoing resources for disease surveillance testing, information management, quality assurance and the development and validation of new diagnostic tests. FERN is also a federal-state partnership and provides critical and ongoing resources for responding to microbiological, chemical or radiological food contamination incidents, for which the U.S. Department of Agriculture (USDA) and the U.S. Food and Drug Administration (FDA) have regulatory authority.

**How samples get to the labs**

The primary route by which samples get to CAHFS labs is through submissions by private veterinarians, owners and state and federal regulatory authorities (fig. 1). This widely diverse group of people ensures that the surveillance is broad based from both geographical and industry perspectives. The laboratory system has four locations in the state: Davis, Turlock, Tulare and San Bernardino ([cahfs.ucdavis.edu/](http://cahfs.ucdavis.edu/)).

The Davis laboratory is adjacent to the SVM complex on campus and is the largest of the four laboratories. The



Photos: chicken, Mike Poe; calf, Elena Zhukova

**Fig. 1. The route that samples from a diseased animal and the subsequent case follow through the California Animal Health and Food Safety (CAHFS) laboratory system.**





The CAHFS lab in San Bernardino was instrumental in the diagnosis and eradication efforts during the 2002–2003 outbreak of exotic Newcastle disease, which was originally diagnosed in a backyard chicken.

Davis lab receives a variety of avian and mammalian livestock samples for diagnostic evaluation and is the site for both the CAHFS toxicology section and the equine drug testing program. The toxicology section is nationally recognized and has partnerships with a variety of federal agencies, such as the Federal Bureau of Investigation (FBI), the FDA, and the Centers for Disease Control and Prevention (CDC). Faculty and staff in this section routinely test for toxins such as oleandrin and various rodenticides as well as natural substances such as nitrate, copper and lead.

The Turlock laboratory performs testing on avian samples and was instrumental in the identification of very virulent infectious bursal disease (vvIBD) in the state in December 2008. This disease, by infecting an organ important in the immune response in commercial poultry, can cause severe morbidity and mortality. The disease has been present in Europe for a number of years but, until it was recognized by the diagnosticians in Turlock, had not been reported previously in the United States (Pitesky et al. 2013).

The Tulare lab works with both mammalian and avian species and has had a significant role in recent identification and eradication efforts directed toward *Mycobacterium bovis* (bovine

tuberculosis) in a small number of dairy herds in California.

The San Bernardino lab also works with both mammalian and avian species and was instrumental in the diagnosis and eradication efforts directed at the exotic Newcastle disease (END) outbreak in 2002 to 2003 in Southern California (Nolen 2002). The original diagnosis of this economically important disease was made on a backyard chicken that had been referred to the San Bernardino lab by a local small-animal practitioner. Eventually the effort to control and eradicate this disease from California involved nearly 1,700 personnel from numerous agencies and a total cost to the state and federal governments of \$168 million over 10 months (E. Sanson-Smith, CAHFS, personal communication). The CAHFS San Bernardino branch served as the central entry point for birds that were necropsied on-site and for samples that were shipped to the USDA National Veterinary Services Laboratory and the Davis CAHFS lab for testing for the presence of the causal virus.

## Research into livestock diseases

Research conducted within UC, including the Agriculture and Natural Resources (ANR) network of advisors, specialists, and UC Davis College of Agricultural and Environmental Sciences (AES) faculty and the staff at the many ANR Research and Extension Centers (RECs), is vitally important in helping the state's producers control endemic diseases of livestock, safeguard the microbiological safety of food and water and improve the sustainability of production agriculture.

**Pinkeye.** One disease that affects both beef and dairy cattle statewide is pinkeye, also known as infectious bovine keratoconjunctivitis (IBK). IBK is the most common eye disease of cattle worldwide (Angelos 2009). Compared to human pinkeye, bovine pinkeye is a much more devastating disease as it affects not only the conjunctiva surrounding the eye but also the cornea itself, and in severe cases causes blindness following rupture of the eye. IBK is a multifactorial disease, meaning that many different factors contribute to its development, including environmental conditions (plant awns such as foxtails, dust, ultraviolet light), insects (flies), bacteria (*Moraxella bovis*) and viruses (Angelos 2009).

Dr. John Angelos, an SVM professor, is building on the earlier work by Lisle George (retired) in conducting research into the pathogenesis and prevention of this important disease. Their collective work has helped characterize important pathogenic factors associated with *Moraxella bovis* and identified genes encoding these factors (Angelos et al. 2001; Angelos, Ball et al. 2007). In addition, they have discovered *Moraxella bovoculi*,



Bovine pinkeye, the most common eye disease of cattle worldwide, affects not only the conjunctiva surrounding the eye, but also the cornea itself, and in severe cases causes blindness.



another *Moraxella* species believed to play a role in bovine pinkeye (Angelos, Spinks et al. 2007). Current research efforts are directed at developing and testing novel subunit vaccines as well as testing alternate routes of vaccination such as intranasal vaccines to prevent IBK (Angelos et al. 2010, 2012). Collaborative efforts between SVM and UC Davis Department of Animal Science faculty and UC Cooperative Extension (UCCE) personnel and ANR resources such as the Sierra Foothill Research and Extension Center (SFREC) have greatly enhanced research into the pathogenesis and prevention of IBK under naturally occurring conditions typical for California's cattle industries.

***Cryptosporidium parvum*.** One of the leading waterborne infectious diseases that is transmitted between animals and humans throughout the United States is the protozoal parasite *Cryptosporidium*

*parvum*. Following several community outbreaks of this disease in the 1990s, concerns escalated by regulatory agencies and drinking water districts that livestock were loading watersheds with this parasite resulted in ad hoc grazing restrictions on private and public rangeland watersheds (Atwill et al. 2012). To clarify the processes causing this emerging human health risk and to develop practical farm-level solutions for the state's livestock industry, a large collaborative effort was initiated in the late 1990s by ANR specialists, advisors and faculty at UC Davis. Key members of the collaboration included Rob Atwill, SVM; Ken Tate, Thomas Harter and Randy Dahlgren, AES; UCCE advisors and their staff throughout California (e.g., livestock and natural resources, watershed, and dairy specialists); state and federal agencies; local water districts; private landowners; and livestock and agricultural organizations such as Backcountry Horsemen of California, California Cattlemen's Association and the California Wool Growers Association.

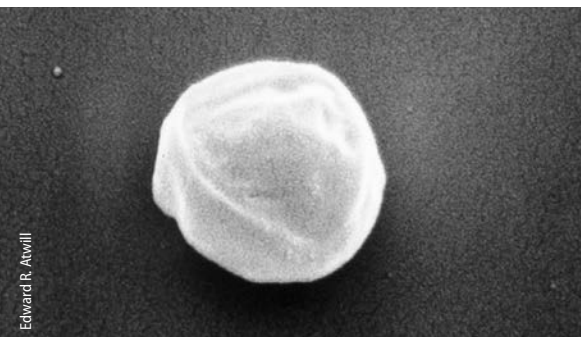
Left, photomicrograph of *Cryptosporidium parvum* oocyst at 10,000x magnification. Below, Lake Kaweah in the Sierra foothills of California, one of the watersheds in which researchers studied comanagement of livestock grazing and water quality.

The research team conducted a series of epidemiological studies in livestock herds on the risk factors for animal infection and also DNA fingerprinting projects to investigate which species of *Cryptosporidium* were being shed by wildlife and livestock and whether these strains were infective for humans (Rochelle et al. 1999; Xiao et al. 2002). Perhaps more importantly, the team conducted a series of experimental soil box trials at the Veterinary Medicine Teaching and Research Center, SVM, followed up by a set of detailed field trials at the SFREC and the San Joaquin Experimental Range, USDA Forest Service, that studied the efficacy of vegetative buffers to remove waterborne pathogens from rangeland runoff (Atwill et al. 2002).

The collaborations on this disease research have created one of the largest and most integrated scientific teams in the United States working on waterborne pathogens in agricultural watersheds. The results are a variety of practical and adoptable best management practices and science-based tools that livestock owners and resource managers can use to reduce the risk of waterborne pathogens from livestock grazing (Atwill et al. 1999). The efforts of ANR, ranging from the specialists and advisors to the staff and resources at the SFREC, were key to the development of these best management practices, which are widely adopted today by both landowners and resource managers.

**Bluetongue.** Bluetongue (BT) is a non-zoonotic disease of certain wild and domestic species of cloven-hoofed ungulates with substantial adverse economic impact on livestock production in California. The causative agent of BT, bluetongue virus (BTV), is an arbovirus that is spread through temperate and tropical regions of the world by biting *Culicoides* midges, which serve as biological vectors. There are several different serotypes of the BTV, and the individual serotypes may be endemic or newly introduced; the newly introduced serotypes often cause outbreaks of disease in naïve animals.

The global distribution of BTV infection has recently altered, likely driven in part by climatic influences on the midge vectors resident in different regions. Similarly, the behavior of BTV infection of livestock has recently altered, with an alarming apparent increase in the







The global distribution of BTV infection has recently altered, likely driven in part by climatic influences on the midge vectors resident in different regions.

occurrence of overt disease in infected cattle (MacLachlan and Mayo 2013). BTV has long been endemic in California and is an ongoing cause of economic loss to livestock producers, with both direct losses caused by reduced production of infected animals and, even more importantly, the adverse impact of nontariff trade restrictions, which close lucrative potential export markets.

Researchers within the UC system target bluetongue in a collaborative effort involving individuals at SVM, UCCE, UC Riverside, CDFA, CAHFS, USDA, and also local veterinarians and local cattle and sheep producers. These extensive interactions and collaborations are critical to the intensive surveillance studies that underpin ongoing efforts to develop accurate and predictive mathematical models relevant to this economically important arboviral disease of livestock. The research group, led by Dr. Jim MacLachlan, has an extramurally funded (currently by NIFA USDA) project to further characterize the ecological drivers of BTV infection among *Culicoides* vectors and ruminant hosts in the state. The long-term objective is to utilize data generated by the



Bluetongue disease, an arbovirus that affects sheep, cattle and other cloven-hoofed ungulates, is spread by a biting midge (*Culicoides sonorensis*) and is an ongoing cause of economic loss to livestock producers in California. UC researchers are part of a collaborative effort to conduct bluetongue virus infection prevalence studies and to develop predictive models. Left, a 1/16 inch long, female, biting midge.

studies for the predictive modeling of this emerging and economically important arboviral infection.

The group's specific objectives are to perform spatial BTV infection prevalence studies to identify environmental, climatic and land-use (and anthropogenic) characteristics associated with regional (local-scale) BTV infection rates of livestock in different regions of California. Initial studies were focused on intensively farmed dairy cattle, and the group is expanding its investigations to free-ranging cattle and sheep. The studies also include extensive entomological investigations to better define the impact of climatic and anthropogenic drivers of the population dynamics of vector midges, and their subsequent impact on the dynamics of BTV infection. Lastly, the group is studying the feeding behaviors of the vector midge species that might be important virus vectors in California and genetically characterizing the evolution of the types of BTVs that circulate in the state, including, most recently, a newly incursive novel virus serotype.

#### Outreach to producers

UCCE has 23 livestock advisors spread across the state ([ucanr.edu/sites/UCCE\\_LR/Beef\\_Cattle/Beef\\_Cattle\\_Advisors/](http://ucanr.edu/sites/UCCE_LR/Beef_Cattle/Beef_Cattle_Advisors/)). They play an important part in the animal disease network, serving a role in

education and applied research with livestock producers in both the management of endemic diseases and in the surveillance for foreign animal diseases. They are supported by three statewide veterinarian specialists, who provide a conduit for information from the campus to livestock producers. This network of advisors and specialists also provides identification of diseases occurring in the field based on observed clinical signs. The CAHFS laboratory system then confirms these observations with laboratory findings. The link provided by UCCE personnel between producers and CAHFS is also used to inform producers about any new diseases in their area. UCCE works with producers and their veterinarians on management actions they can take to minimize the occurrence of the disease in their herd. Applied research may also be required with laboratory analysis to determine the effectiveness of the mitigating management action.

Recently, the CAHFS laboratory database of toxicological findings was mined for use in a UCCE publication on poisonous plants in California (Forero et al. 2011). It reported that for all kinds of livestock the number one reported problem from toxic plants was oleander, with over 555 cases during 17 years (fig. 2). Although this plant is used by

*Continued on page 116*



## Solving the puzzle of foothill abortion in beef cattle

by Glenn A. Nader, Mike N. Oliver, Julie A. Finzel, Myra T. Blanchard and Jeff L. Stott

Foothill abortion, also known as epizootic bovine abortion (EBA), has been a long-standing problem for California beef cattle producers. It is a major source of economic loss for California cow and calf producers, and in the 1990s it was estimated that 5% to 10% (45,000 to 90,000 calves) of the California beef calf crop may be lost each year (Bushnell et al. 1991). UC Cooperative Extension (UCCE) farm advisors, specialists and UC Davis School of Veterinary Medicine (SVM) faculty have worked on this disease for nearly 50 years. This long research process finally moved forward in 2005, when the causative agent was identified.

The Pajaroello (pa-ha-WAY-lo) tick, *Ornithodoros coriaceus*, is responsible for transmitting the causative agent (a deltaproteobacterium) when it feeds on a pregnant cow. The Pajaroello is a soft-bodied tick that resides in dirt or litter under trees and bushes, locations where deer and cattle typically bed down. The Pajaroello does not embed itself in animal flesh, but rather it feeds rapidly (for as little as 20 minutes) and then drops back onto the ground. It can survive for years in a dormant state, without taking a blood meal.

If a cow or heifer is bitten by a tick when 2 to 6 months pregnant, the calf may abort or be born weak. Heifers and cows that have not previously grazed in tick-infested pastures are most susceptible. Once bitten, cows appear to gain some degree of immunity, but ranchers have observed that immunity can be lost if cattle go for a year or more without tick bites, which serve as an immunity booster.

Early UC efforts focused on identifying the vector of the disease. First, mosquitoes were suspected. They were eliminated as a possibility when cattle elevated off the ground (in an area where the disease commonly occurred) carried their calves to term. Additional experiments also eliminated the *Leptoconops* gnat as a possible vector.

It was initially thought that the Pajaroello tick did not live in the most northern areas of California, where EBA occurred. When ticks were subsequently trapped on a northern Lassen County ranch that had experienced abortions, it was confirmed as a potential vector. Ticks were collected and placed to feed on susceptible heifers (on the UC Davis campus), and abortions occurred, confirming the Pajaroello as the vector of the disease.

The next step was to determine the causative agent being transmitted by the bite of the tick. This was difficult because the tick harbors numerous potential causative agents. In the late 1960s

and through the 1970s, chlamydia was considered as a possible causative agent. During this period, a field trial was conducted with cooperated cattle in Lassen County in which susceptible heifers were fed tetracycline crumbles, and the data suggested there might be some protection from the antibiotics. Numerous chlamydia vaccines were prepared and given to susceptible heifers, but this effort was ultimately abandoned when heifers continued to abort following vaccination.

In the 1970s and early 1980s, viruses were considered as possible causative agents. A large research effort was initiated, with over 80 viruses isolated from the tick. After exhaustive work, research on causative agents moved from viruses to spirochete-like organisms; a *Borrelia* species was suggested to be a potential cause of EBA, but further experimental studies essentially eliminated spirochetes and *Borrelia* species as potential causative agents.

With no definitive causal agent of EBA identified, the California Cattlemen's Association gave UCCE a grant from its Livestock Memorial Research Fund to develop educational outreach through a video on how to manage cattle to minimize the impact of the disease. Farm advisors and specialists with knowledge of the tick's feeding habitats and how the abortions developed in cattle used case studies with ranchers to develop management options that ranchers could use to lessen the impact of the disease on their business. Successful practices included pre-exposing sexually mature heifers to known tick areas prior to breeding, avoiding tick areas during the critical 2 to 6 months of pregnancy and shifting from spring to fall calving in the most northern regions of the state.

SVM researchers and the California Animal Health and Food Safety (CAHFS) laboratory system, using a large number of aborted calves, were able to develop methods to identify foothill abortion in aborted calves. This knowledge was extended to practicing veterinarians working with ranches throughout the state.

In 2002, a SVM laboratory developed a reliable challenge system for experimental transmission of EBA that was used to establish that the causative agent was antibiotic susceptible. This report was quickly followed with a positive identification of the agent causing foothill abortion, a bacteria belonging to a very unusual group of slime bacteria; then referred to simply as the agent of EBA, the bacterial pathogen has now been unofficially named *Pajaroellobacter abortibovis*.



Pajaroello tick



Other breakthroughs followed quickly. The cultivation of the bacteria in immunodeficient mice gave new life to research efforts. A vaccine development phase was initiated with over \$200,000 from the California Cattlemen Association's Livestock Memorial Research Fund and financial support from SVM and their collaborators at the University of Nevada, Reno. In 2009, a small group of heifers were protected against experimental infection after they were immunized several weeks prior to breeding with a candidate vaccine that was both live and infectious.

The success of a second and larger trial in 2010 prompted SVM researchers to pursue product licensing with the U.S. Department of Agriculture (USDA) Center for Veterinary Biologics. Vaccine efficacy experiments were conducted in accordance with USDA regulations. University-owned heifers were immunized before breeding and then administered an artificial challenge with virulent bacteria at the peak of fetal susceptibility (100 days gestation). Vaccine field trials that combined USDA-required field safety trials with field efficacy were then initiated at the UC Sierra Foothill Research and Extension Center on UC Davis Department of Animal Science heifers, on heifers at University of Nevada, Reno, and on producer-owned beef herds in California and Nevada. Over 1,600 heifers were enrolled in these trials in 2011 and 2012. Additional funding for such a massive effort was provided to the SVM by UC's Office of the President via a UC Proof of Concept Discovery Grant (grant ID no. 212263).

Although the results of these studies are currently being assembled, preliminary assessment of the experimental vaccine indicates excellent protection against foothill abortion has been successfully demonstrated. All the successes realized to date were a result of SVM collaborations across the UC spectrum with CAHFS's diagnostic laboratory at UC Davis, the UC Davis Department of Animal Science, UCCE, the Sierra Foothill Research and Extension Center, and also with researchers at the University of Nevada, Reno.

Historically, specialty vaccines created for use only in California were licensed through the California Department of Food and Agriculture (CDFA). Currently, CDFA does not process new specialty vaccines, requiring researchers to work with the USDA to get the foothill abortion vaccine licensed. USDA requirements are more stringent than CDFA's requirements, as food animal vaccines must comply with the federal Virus-Serum-Toxin Act requirements. SVM and USDA are charting new territory as they work to certify the safety and efficacy of the vaccine. The developers of the vaccine at SVM are in the process of establishing a USDA-required vaccine seed, determining if production can be scaled up to a commercial level and identifying viable options for commercial production of the vaccine.

As the commercialization efforts proceed, researchers are fine-tuning the vaccination regime to address concerns over the prolonged persistence of the vaccine bacteria and the potential impact

on embryonic mortality in animals bred within weeks following vaccination. These studies are being conducted using a combination of UC and private producer replacement heifers. The vaccine dose is being adjusted downward, and the time from vaccination to breeding is being extended. The vaccine cannot be administered to pregnant cattle. Skin reactions following vaccination suggest that the live bacterial pathogen can persist for up to 2 months. On the positive side, this bacterial persistence induces a solid immunity that likely lasts through the next breeding cycle and possibly beyond. Studies are under way to begin to address length of immunity.

The fact that the vaccine is live and infectious poses several unique challenges. For example, the cryopreserved bacteria must be transported and stored in liquid nitrogen, and the cost of purchasing the vaccine could also be high because of the cost of manufacturing — the live vaccine must be cultured in an immunodeficient mouse. The California Cattlemen's Association is working to develop a regional distribution system for the vaccine, which could become available within a couple of years.

Work is being conducted to develop a recombinant vaccine through genomic research. In a recombinant vaccine, the genomic sequence of candidate bacterial genes must first be established. Next, the genes must be expressed as protein and then combined with adjuvant(s) to construct candidate vaccines. A recombinant vaccine would be far less sensitive to temperature and would not require immunodeficient mice in the manufacturing process, thereby making the finished product much more cost effective and practical for on-ranch use.

Subsequent findings by the SVM researchers have also improved the diagnostic procedures for identifying foothill abortions at the CAHFS diagnostic laboratory at UC Davis and have provided additional important information for UCCE to extend to ranchers to confirm abortions caused by the disease. Researchers are now actively pursuing validation of a diagnostic assay that may allow ranchers in the future to identify cows that have been exposed to the tick. Such an assay could be used to establish susceptibility of naive replacement heifers to foothill abortion or confirm whether the disease is present on a ranch.

Decades of hard work by UC researchers and educators all across the system have allowed the pieces of this difficult disease puzzle to come together in assisting California's cattle ranchers. [CA](#)

## References

- Blanchard MT, Chen CI, Anderson M, et al. 2010. Serial passage of the etiologic agent of epizootic bovine abortion in immunodeficient mice. *Vet Microbiol* 144(1-2):177-82.
- Bushnell R, Oliver M, Nader G, Norman B. 1991. Foothill Abortion. *Veterinary Medicine Extension, Cooperative Extension, School of Veterinary Medicine, University of California, Davis.*
- King DP, Chen C-I, Blanchard MT, et al. 2005. Molecular identification of a novel deltaproteobacterium as the etiologic agent of epizootic bovine abortion (foothill abortion). *J Clin Microbiol* 43(2):604-9.
- Stott JL, Blanchard MT, Anderson M, et al. 2002. Experimental transmission of epizootic bovine abortion (foothill abortion). *Vet Microbiol* 88(2):161-73.

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Caltrans and others because of its drought hardiness, it does have an impact on the animal population, which needs to be highlighted. The publication by Forero et al. (2011) and the information provided also helped producers define the risk of plant poisonings.

The CAHFS laboratory system and UCCE also play an important role in poultry diseases. Many backyard poultry operations allow their birds to roam free and intermix with wild birds, and as a result they represent a potential portal for the transmission of diseases such as avian influenza from wild birds. Due to the relative lack of veterinary clinics with poultry expertise, many backyard producers look to UCCE livestock advisors to help them with routine management and disease identification during a disease outbreak. This partnership between Extension personnel and the CAHFS lab system is an important part of the effort to protect both commercial and noncommercial poultry as well as the general public from diseases originating in wild birds.

The CAHFS laboratory system and UCCE specialists have worked together to define and test new diagnostic procedures that have higher accuracy or shorter turn-around times. For example, sampling of bulls to test for trichomoniasis, a venereal disease of cattle, requires that the animals be gathered and restrained in a chute. When culture-based methods were in use, bulls had to be sampled three times to be certain they were not infected, which led to increased feed costs and potential trauma to the animals from repeated

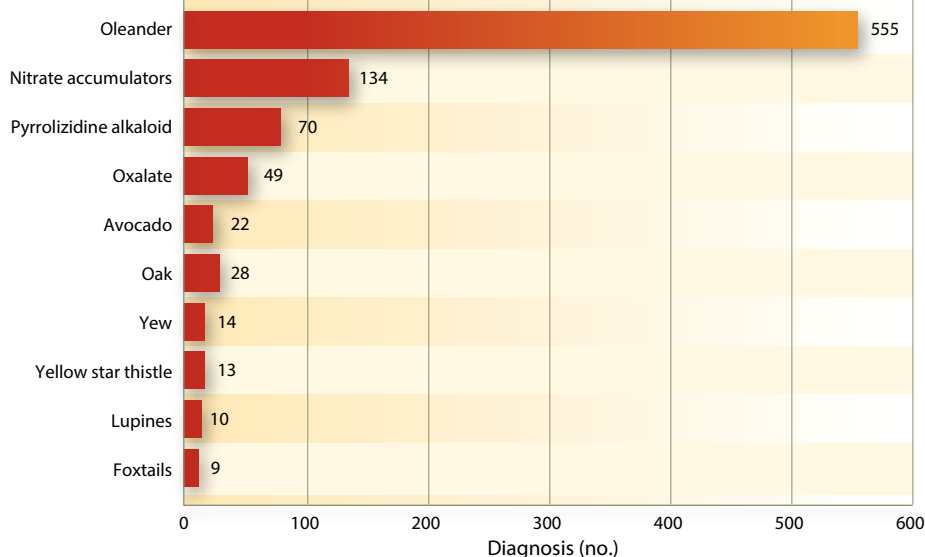


Fig. 2. Livestock plant-poisoning cases, 1990–2007.

handling. In response to requests from the California Cattlemen’s Association, the CAHFS lab system adopted a PCR-based test that had originally been developed by Dr. Bob BonDurant of SVM (Ho et al. 1994). It has subsequently been determined that the same degree of certainty that an animal is free of infection can be provided by testing a single sample via PCR as was formerly gained by culturing three separate samples. This has resulted in a faster test turn-around time and significantly less handling of bulls.

UC continues to demonstrate its value on a daily basis by playing an important and wide-ranging role in the protection of animal agriculture, by being heavily involved in the surveillance for and diagnosis of important diseases of livestock,

by performing fundamental and applied research on those same diseases and by distributing the knowledge gained back to producers. UC is also an important source for expert knowledge about livestock diseases to both federal and state regulatory officials on a regular basis and during disease outbreak emergencies. [CA](#)

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## References

- Angelos JA. 2009. Infectious bovine keratoconjunctivitis. In: BP Smith. *Large Animal Internal Medicine*. St. Louis, Missouri: Mosby Elsevier. p 1286–8.
- Angelos JA, Ball LM, Hess JF. 2007. Identification and characterization of complete RTX operons in *Moraxella bovoculi* and *Moraxella ovis*. *Vet Microbiol* 125:73–9.
- Angelos JA, Gohary KG, Ball LM, et al. 2012. Randomized controlled field trial to assess efficacy of a *Moraxella bovis* pilin-cytotoxin-*Moraxella bovoculi* cytotoxin subunit vaccine to prevent naturally occurring infectious bovine keratoconjunctivitis. *Am J Vet Res* 73:1670–5.
- Angelos JA, Hess JF, George LW. 2001. Cloning and characterization of a *Moraxella bovis* cytotoxin gene. *Am J Vet Res* 62:1222–8.
- Angelos JA, Lane VM, Ball LM, et al. 2010. Recombinant *Moraxella bovoculi* cytotoxin-ISCOM matrix adjuvanted vaccine to prevent naturally occurring infectious bovine keratoconjunctivitis. *Vet Res Commun* 34:229–39.
- Angelos JA, Spinks PQ, Ball LM, et al. 2007. *Moraxella bovoculi* sp. nov., isolated from calves with infectious bovine keratoconjunctivitis. *Int J Syst Evol Micr* 57:789–95.
- Atwill ER, Hou L, Karle BM, et al. 2002. Transport of *Cryptosporidium parvum* oocysts through vegetated buffer strips and estimated filtration efficiency. *Appl Environ Microb* 68(11):5517–27.
- Atwill ER, Johnson E, Das Gracias M, Pereira C. 1999. Association of herd composition, stocking rate, and calving duration with fecal shedding of *Cryptosporidium parvum* oocysts in beef herds. *J Am Vet Med Assoc* 215(12):1833–8.
- Atwill ER, Partyka M, Bond RF, et al. 2012. An Introduction to Waterborne Pathogens in Agricultural Watersheds. Nutrient Management Technical Note No. 9. Natural Resources Conservation Service, US Department of Agriculture. p 1–84.
- Forero L, Nader G, Craigmill A, et al. 2011. Livestock-Poisoning Plants of California. UC ANR Pub 8398. Oakland, CA. <http://anrcatalog.ucdavis.edu/pdf/8398.pdf>.
- Ho MS, Conrad PA, Conrad PJ, et al. 1994. Detection of bovine trichomoniasis with a specific DNA probe and PCR amplification system. *J Clin Microbiol* 32(1):98–104.
- MacLachlan NJ, Mayo CE. 2013. Potential strategies for control of bluetongue, a globally emerging, *Culicoides*-transmitted viral disease of ruminant livestock and wildlife. *Antivir Res* 99(2):79–90.
- Nolen RS. 2002. Exotic Newcastle disease strikes game birds in California. *J Am Vet Med Assoc* 221(10):1369–70.
- Pitesky M, Cataline K, Crossley B, et al. 2013. Historical, spatial, temporal, and time-space epidemiology of very virulent infectious bursal disease in California: A retrospective study 2008–2011. *Avian Dis* Mar 57(1):76–82.
- Rochelle PA, Jutras EM, Atwill ER, et al. 1999. Polymorphisms in the beta-tubulin gene of *Cryptosporidium parvum* differentiate between isolates based on animal host but not geographic origin. *J Parasitol* 85(5):986–9.
- USDA NASS. 2012. US Department of Agriculture National Agricultural Statistics Service, Crop Year 2011. [www.nass.usda.gov/Statistics\\_by\\_State/California/Publications/California\\_Ag\\_Statistics/Reports/2011cas-all.pdf](http://www.nass.usda.gov/Statistics_by_State/California/Publications/California_Ag_Statistics/Reports/2011cas-all.pdf).
- Xiao L, Sulaiman IM, Morgan UM, et al. 2002. Host adaptation and host-parasite co-evolution in *Cryptosporidium*. *Int J Parasitol* 32:1773–85.



# Plant health: How diagnostic networks and interagency partnerships protect plant systems from pests and pathogens

by Richard M. Bostock, Carla S. Thomas, Richard W. Hoenisch, Deborah A. Golino and Georgios Vidalakis

*Early detection and rapid response are crucial in any effort to reduce the risk of new and emerging biological threats to crops and other plant resources. This underscores the importance of having the necessary diagnostic expertise, infrastructure and resources in place. Three programs — the National Plant Diagnostic Network, the National Clean Plant Network and the Citrus Clonal Protection Program — illustrate how accurate and rapid diagnosis plays a critical role in providing healthy plants for growers and in securing production systems for food and fiber. These three programs depend on state-wide, regional and national networking among university, state and federal scientists, regulatory officials and industry members to help mitigate the impacts of plant pests and diseases.*

Plant pathogens and pests present continual challenges to the production and security of food, fiber and forest resources. Introduced biological agents threaten crops and forests locally and regionally with their direct damage to host plants, and their presence has national and international consequences for trade and regulatory policy. Contaminated seed and nursery plants provide efficient introduction pathways for pests, highlighting the critical importance of clean seed and nursery programs for protection of both domestic and export markets. Introduced biological agents, whether they arrive via natural dispersal mechanisms or through human activity, may remain present but below our threshold for detection and perception for years, only to emerge later with seeming suddenness and dramatic destructive intensity (Crooks 2005).

As is the case in human and animal medicine, early detection, accurate diagnosis and rapid response are critical for achievement of successful outcomes when dealing with outbreaks of endemic and newly introduced plant diseases and insect or weed pests. The United States has a long history of pathogen and pest

introductions for which containment and eradication have been unsuccessful, often with devastating consequences to agriculture and natural ecosystems (Pimentel et al. 2005; Rossman 2009). In California, the past two decades have seen the emergence of a number of high-consequence plant pests, including the sudden oak death pathogen, the Pierce's disease pathogen (carried by the glassy-winged sharpshooter) and more recently the Asian citrus psyllid, vector of the citrus greening disease (huanglongbing), and

the European grapevine moth. California is the leading U.S. state for agricultural products, with gross cash receipts in 2011 in excess of \$43 billion (CDFA 2013) for more than 400 commodities, including numerous specialty crops. In fact, seven of the top 10 commodities in terms of cash value in California are specialty crops. All of this presents great opportunities, as well as risks and challenges, for diagnostic, pest management and clean stock and seed programs.

During the past decade, awareness and concern have increased in regard to the threat of economic harm to U.S. crop agriculture, nurseries and forests posed by introduced biological agents, and society has responded with greater investments in biosecurity programs to enhance surveillance and detection. Understandably, exotic and high-consequence agents receive the greatest focus in such programs, but it is important to note that sufficient diagnostic infrastructure and expertise must be continually maintained so we can accurately identify both routine and unusual conditions.



Heavy infestation of Asian citrus psyllids (*Diaphorina citri*) on *Murraya*. Inset, adult psyllid.

Online: <http://californiaagriculture.ucanr.edu/landingpage.cfm?article=ca.v068n04p117&fulltext=yes>  
doi: 10.3733/ca.v068n04p117

Even with recent investments in plant biosecurity programs, the immensity of the task exceeds the resources currently available to adequately address it. For example, the most significant pathway for introduction of unwanted plant pathogens and pests is through live plant imports, which have increased, on average, by 51 million plants per year for the past 43 years (Liebhold et al. 2012; Palm and Rossman 2003). In 2010, more than 2.8 billion plants intended for planting within the United States passed through federal plant inspection stations at U.S. ports of entry. This sheer volume of plants by itself reduces the likelihood that all of the potentially invasive pests and pathogens will be intercepted, and that in turn makes downstream programs for detection, diagnosis and containment all the more important.

### The National Plant Diagnostic Network

The National Plant Diagnostic Network (NPDN) was established in 2002 to provide greater support for and integration of plant diagnostic laboratories in the United States and help thwart the establishment and dispersal of introduced insect and weed pests and pathogens ([npdn.org/](http://npdn.org/)). The NPDN works with state and federal agencies to ensure the quick, accurate and secure conveyance

of information and expertise about new detections in order to minimize their economic and ecological impacts (Stack et al. 2006; Stack et al. 2014).

Federal funding for the establishment of the NPDN and a parallel program for animal agriculture, the National Animal Health Laboratory Network (NAHLN), was largely a response to biosecurity risks to plant and animal agriculture in the United States. Also compelling for decision-makers at the time was a growing awareness that many publicly funded plant diagnostic labs associated with land-grant universities (LGUs) and state departments of agriculture were in fact underfunded and were, in some cases, at risk of closure as a result. In addition, it appeared that a lack of coordination among diagnosticians and experts at university, state and federal laboratories could create bottlenecks in the processing of critical samples and delays in communication of diagnostic results.

The NPDN is supported in part by the USDA's Food and Agriculture Defense Initiative (FADI), a program within the National Institute of Food and Agriculture (NIFA). To further its core missions of diagnostics, training and education, and communication, the NPDN provides resources for diagnostic laboratory infrastructure and supplies,

delivers advanced training for diagnosticians and educational programs for "first detectors," facilitates communication among plant diagnosticians at LGUs, state departments of agriculture and national expert laboratories, and seeks to provide accurate and timely information to state and federal authorities to guide an appropriate response. For greater efficiency in program delivery, the NPDN is divided into five geographic regions, each with a regional center located in the plant pathology department of an LGU (fig. 1), where its directors help identify and coordinate local and regional scientific expertise in plant pathology, entomology and weed/plant science as needed. The NPDN does not have any formal regulatory authority of its own; rather, the network's principal role is to provide a framework that facilitates access to additional expertise in the event of a plant health emergency.

Given the agricultural diversity of the United States, the regionally distributed structure of the NPDN enables each region to tailor the resources and programs it provides to best meet the needs of that particular region. For example, the Western region includes 10 Western states and U.S. Pacific territories that partner to form the Western Plant Diagnostic Network (WPDN; [wpdn.org/](http://wpdn.org/)). The regional center, located at UC Davis,

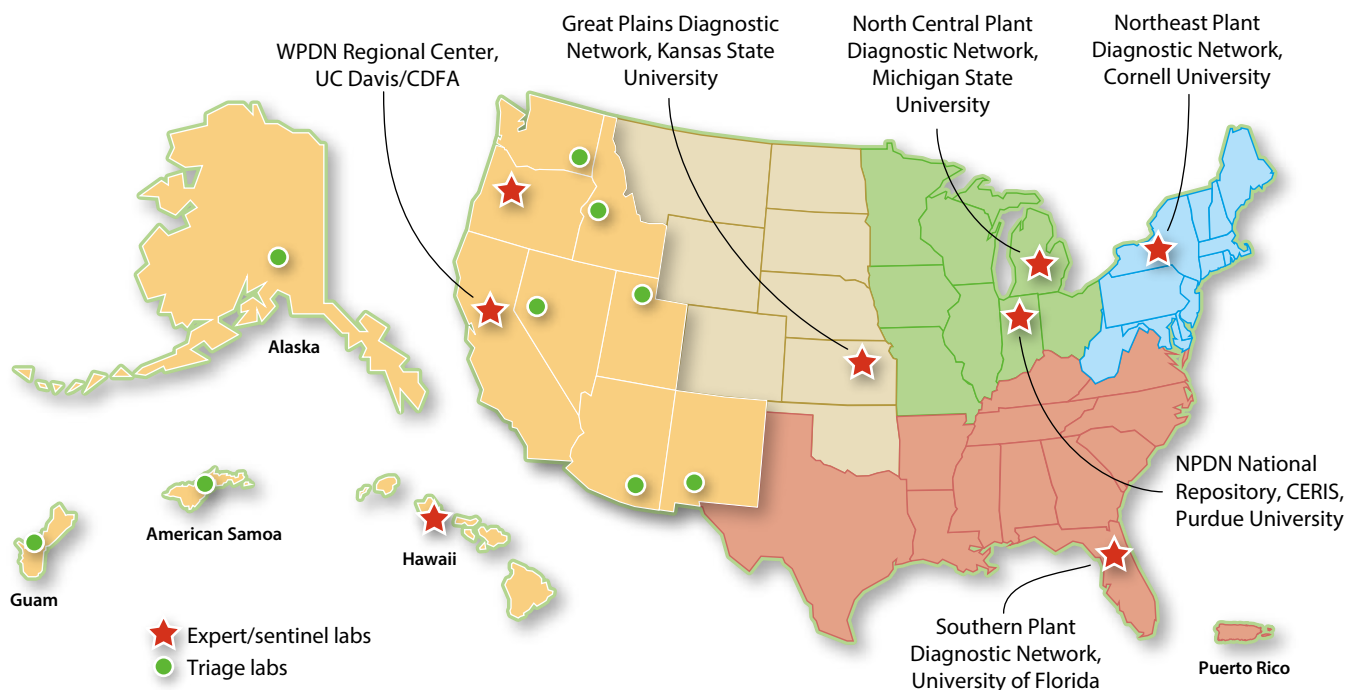


Fig. 1. Organizational map of the National Plant Diagnostic Network with the five regional networks and the NPDN National Repository at Purdue University. The Western Plant Diagnostic Network (WPDN) includes 10 Western states and U.S. territories in the Pacific.



works in partnership with the California Department of Food and Agriculture (CDFA) Plant Pest Diagnostic Center, which is the formal diagnostic reporting authority for California ([cdfa.ca.gov/plant/PPD/](http://cdfa.ca.gov/plant/PPD/)).

The scope and size of agricultural, nursery and forest production systems, as well as infrastructure and expertise in support of plant health programs, vary considerably throughout the WPDN, with three subregional expert laboratories that work together with the various triage laboratories in individual states and territories. The three expert laboratories — the CDFA laboratory in Sacramento and diagnostic laboratories at Oregon State University (Corvallis) and the University of Hawaii (Manoa), working in partnership with the Hawaii Department of Agriculture — can handle most or all types of samples and conduct collaborative diagnostics with other member laboratories in the region or the nation. The CDFA laboratory is the largest and most comprehensive state plant diagnostic laboratory in the United States, with more than 70 scientists and support staff specializing in pests, diseases, weeds, nematodes and seed certification. It receives little funding from the NPDN, relative to the scope and size of the laboratory's activities, drawing its primary support from state funds and other grants.

Similar to the WPDN, the other four regional networks in the NPDN are consortia of LGUs and state departments of agriculture working together to support mission-related activities and provide diagnostic services. Diagnostic results from NPDN member laboratories in all 50 states and three U.S. territories are submitted to the NPDN National Repository at Purdue University's Center for Environmental and Regulatory Information Systems (CERIS; [ceris.purdue.edu/npdn/](http://ceris.purdue.edu/npdn/)). At the time of this writing (September 2014), the repository houses more than 930,000 diagnostic sample records in a database that is updated daily; it receives about 100,000 new records each year from as many as 150 laboratories throughout the nation. Such a comprehensive national database of diagnostic records is unprecedented in plant agriculture. It is available for search and analysis by authorized scientists, regulatory officials and diagnosticians who use it to rapidly identify novel detections and

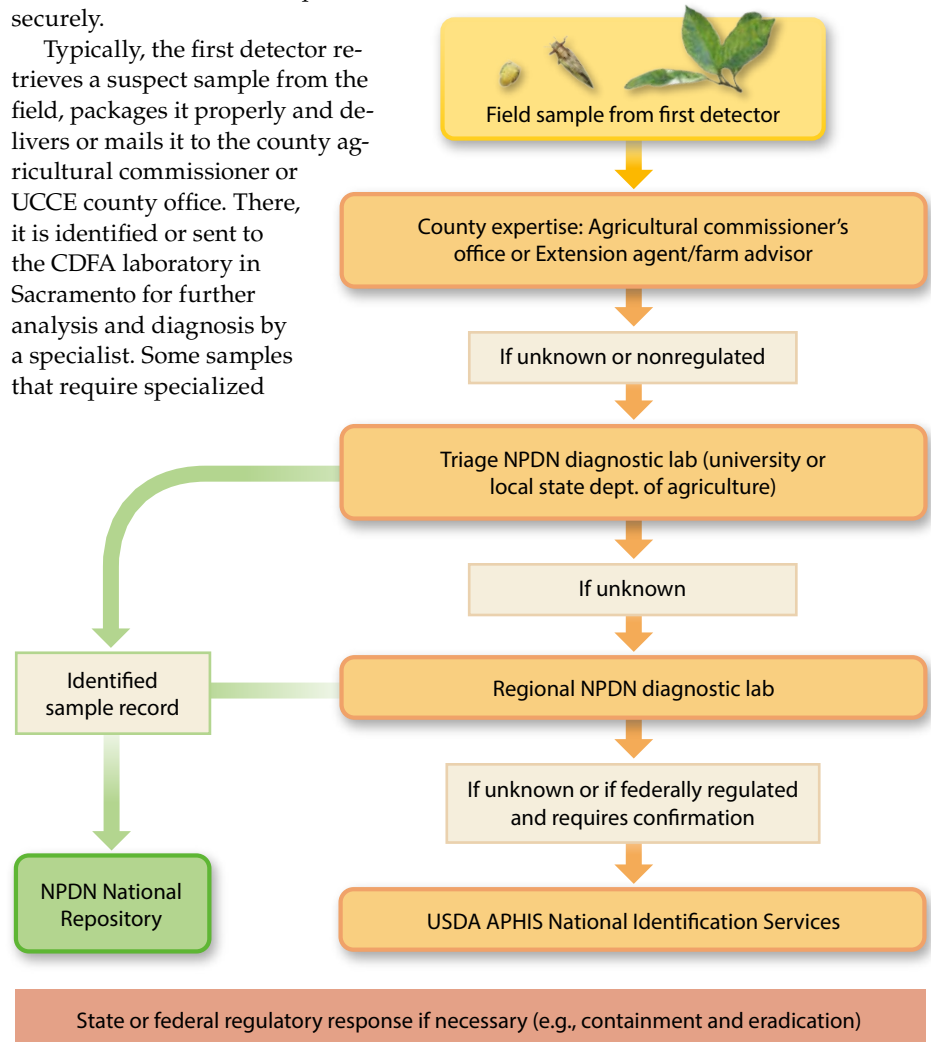
discern patterns of new outbreaks at various scales of resolution and in relation to GIS-based platforms for weather and land use.

**Path of a suspect sample.** An important contribution of the NPDN has been the education of first detectors in the proper procedure to follow when submitting samples to diagnostic labs (fig. 2). In California, a first detector may be a grower, crop consultant, pest control adviser, UC researcher, UC Cooperative Extension (UCCE) farm advisor, biologist within a county agriculture commissioner's office, UC-trained master gardener, employee of a city or county parks department or homeowner — really, any person who has been trained to recognize potential threats to plant health and understands the importance of plant biosecurity, and who knows how to collect and submit a sample securely.

Typically, the first detector retrieves a suspect sample from the field, packages it properly and delivers or mails it to the county agricultural commissioner or UCCE county office. There, it is identified or sent to the CDFA laboratory in Sacramento for further analysis and diagnosis by a specialist. Some samples that require specialized

diagnostics are often sent directly to UC laboratories that have the appropriate expertise. If the pest or pathogen is suspected to be new to California or to the county where it was found, a confirmatory diagnosis must be made by CDFA scientists, and may require further sampling. When a pest or pathogen is suspected to be new to the United States or to North America, samples are sent to the USDA Animal Plant Health Inspection Service (APHIS) National Identification Services, where specialists at federal, state or university laboratories with expert knowledge about the suspect agent are consulted for a confirmatory diagnosis.

**How the NPDN has made a difference.** Perhaps the most important contribution of the NPDN has been to foster an unprecedented level of coordination



**Fig. 2.** Path of a field sample from first detector through the diagnostic process and ultimately to being archived as a record in the NPDN National Repository.

and communication among the nation's diagnostic laboratories. For example, in the event of a regional or national plant health emergency, instead of overwhelming one laboratory with samples, the distributed network structure provides a more flexible surge capacity, eliminating bottlenecks that could hamper the diagnostic and reporting process (see sidebar, page 121). In addition, through its regional networks and in partnership with USDA APHIS, the NPDN has made significant investments in laboratory infrastructure, equipment, supplies and distance diagnosis capabilities, and it has provided partial salary support and advanced training for scientific and technical staff.

Another important accomplishment of the NPDN is the development and delivery of training and education programs for first detectors and diagnosticians. The NPDN's national registry now includes more than 16,000 first detectors who can be alerted quickly to new outbreaks and who receive regular updates via national and regional e-newsletters with useful information about new pests and pathogens of regulatory concern. In California, currently boasting more

than 2,500 registered first detectors, the WPDN training coordinator and UCCE specialists and farm advisors and other UC Division of Agriculture and Natural Resources (ANR) personnel work together to conduct trainings and workshops. These are offered in conjunction with experts at universities, the CDFA and the USDA APHIS and USDA Agricultural Research Service (ARS) to ensure network-wide consistency and preparedness of diagnostic labs.

### The National Clean Plant Network

Healthy planting stock is key to the cost-effective production of horticultural crops such as fruit trees, nut trees and grapevines. Healthy stock is easier to propagate, requires fewer chemical inputs and produces higher crop yields and better crop quality. The U.S. agricultural sector needs healthy planting stock to keep it internationally competitive and economically viable. The most efficient approach to producing healthy planting stock is through programs that screen valuable plant selections for viruses and other diseases that have the potential to be spread through contaminated plant stock. Quarantine services provided by clean stock programs reduce the chances

of introduction of exotic pests that, once introduced, can be difficult and costly to control.

The original impetus for the organization of networks with a focus on clean stock came from the U.S. grape and fruit tree industries, which in 2005 began to explore the formation of a national group devoted to foundation materials that have been tested, treated and maintained as a healthy source of plant materials for growers to use. In 2008, these grape and fruit tree networks were developed by stakeholders, industry members, scientists and other interested parties. The new National Clean Plant Network (NCPN) was included in the 2008 and 2014 Farm Bills, with funding of \$5 million per year to provide reliable sources of propagative material that are free of graft-transmitted pathogens. Congress stipulated that funding go exclusively to existing clean plant centers that were already supported by their home institutions.

The NCPN is a voluntary association made up of specialty crop networks that promote the use of pathogen-tested, healthy plant material for U.S. growers of grapes, fruit trees, hops, berries and citrus. Its formal mission is to "provide high quality asexually propagated plant material free of targeted plant pathogens and pests that cause economic loss to



1. Auburn University, Auburn, Alabama
2. Clemson University, South Carolina
3. Cornell University, Geneva, New York
4. Florida Department of Agriculture and Consumer Service, Gainesville
5. Florida Department of Agriculture and Consumer Service, Winter Haven
6. Florida A&M University, Tallahassee
7. Louisiana State University, Baton Rouge
8. Michigan State University, East Lansing
9. Missouri State University, Mountain Grove
10. North Carolina State University, Raleigh
11. Oregon State University, Corvallis
12. Texas A&M University (TAMUK), Kingsville
13. University of Arizona, Yuma
14. University of Arkansas, Fayetteville
15. University of California, Davis
16. University of California Riverside, in collaboration with USDA ARS National Clonal Germplasm Repository for Citrus and Dates
17. University of Hawaii, Honolulu
18. USDA ARS, Corvallis, Oregon
19. Washington State University-IAREC, Prosser

Fig. 3. The 19 regional centers of the National Clean Plant Network (NCPN).



protect the environment and ensure the global competitiveness of specialty crop producers.” By agreement, it operates under the auspices of three federal agencies — USDA APHIS, ARS and NIFA — which cooperatively support its research, quarantine and outreach activities.

USDA funding supports existing clean plant centers that have the expertise, facilities and appropriate climates to efficiently produce, maintain and distribute healthy planting stock for those crops. Advisory committees that include industry representatives and researchers from throughout the country make up an essential part of the equation for communicating priorities to the NCPN.

**NCPN centers.** As of this writing, the NCPN network includes 19 clean plant centers (fig. 3). Each of the five current crop programs has one program that serves as its administrative lead. A national website ([nationalcleanplantnetwork.org](http://nationalcleanplantnetwork.org)) maintained by the staff of Foundation Plant Services (FPS) at UC Davis has links to a website for each of the NCPN crops maintained by the administrative lead center. UC Davis is administrative home for the grape network. The NCPN berries group — the Berry Crops Testing, Therapy and Diagnostics Development Program — is headquartered at the USDA Horticultural Crops Research Unit, USDA ARS, in Corvallis, Oregon. The citrus network home is with the Citrus Clonal Protection Program (CCPP) at UC Riverside (see below). Both the fruit tree network and the hops network are housed at Clean Plant Center Northwest, Washington State University-IAREC, in Prosser, Washington.

### Citrus Clonal Protection Program

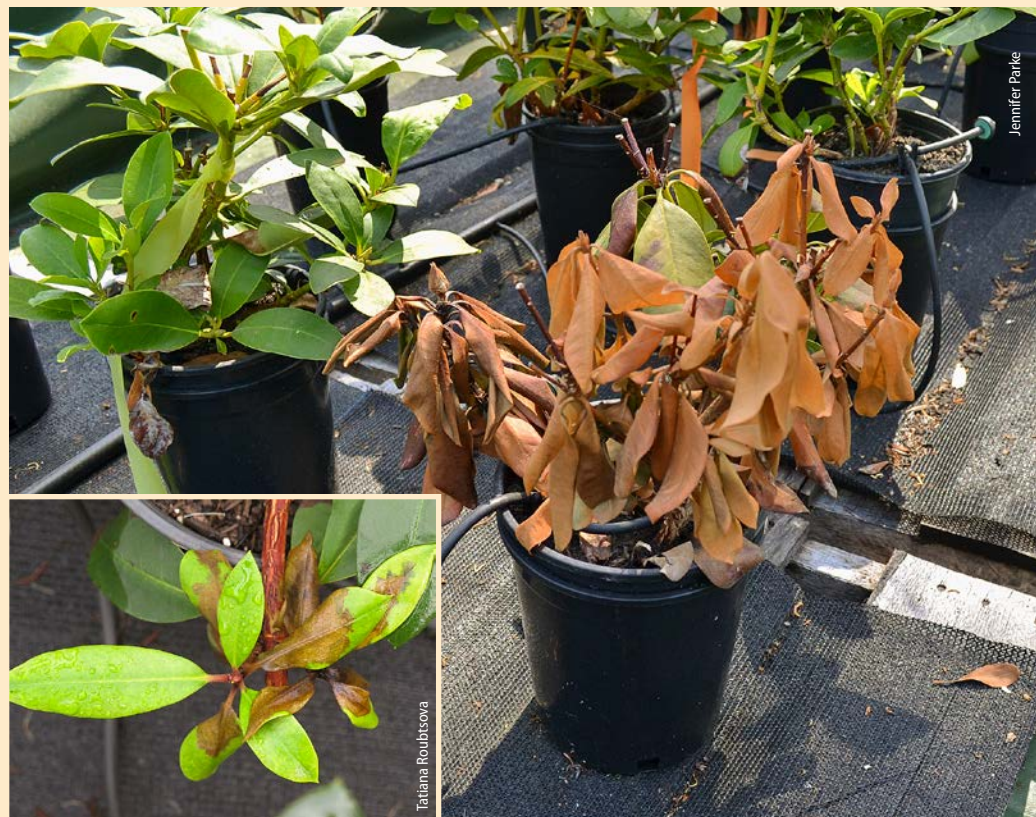
The CCPP has its roots in the 1930s, when Professor Howard Fawcett of the UC Citrus Experiment Station in Riverside discovered the viral nature of the graft-transmissible disease citrus psorosis — the discovery that triggered the establishment of the Psorosis Free Program. Following a request from the citrus industry, UC established what is now the CCPP in 1956, initially calling it the Citrus Variety Improvement Program. Today the CCPP stands as a cooperative program of UC Riverside’s Department of Plant Pathology and Microbiology, CDFR, USDA APHIS and the NCPN, as well as the state’s citrus industry as represented

## A broad lab network means greater flexibility and better response

The “surge capacity” of the National Plant Diagnostic Network (NPDN) was instrumental in providing support for several investigations of *Phytophthora ramorum*, the cause of sudden oak death in coastal forests and ramorum blight of ornamentals (Rizzo et al. 2005). In 2004, *P. ramorum* was unexpectedly detected in several Southern California production and wholesale nurseries that had already shipped 2.3 million potentially infected plants to retail nurseries and other wholesale nurseries in 49 states and the District of Columbia. This triggered a national regulatory response to determine where plants had been shipped and then, once found, to test the plants for *P. ramorum*.

Over the next year, federal and NPDN labs processed more than 100,000 samples. Because of the NPDN laboratories’ advanced coordination and training, they were able to rapidly implement the standardized APHIS diagnostic protocol with little advance notice. One critical element of this protocol is a molecular diagnostic based on the PCR assay, which at the time was newly developed by APHIS and UC scientists. In addition, the NPDN provided support for the purchase of PCR machines and supplies and reagents that enabled many network labs to participate. In total, 171 nurseries in 20 states tested positive for *P. ramorum*, triggering implementation of containment and eradication measures at those sites.

Prior to establishment of the network, it could take 6 weeks or more for samples to go through the system, and nursery plants were quarantined until diagnosis could be completed. With the NPDN labs and the new diagnostic assay, turnaround times from field collection to diagnosis were reduced to as few as 4 days, enabling many nurseries around the country to quickly resume operations once they had been cleared as free from *P. ramorum*. [CA](#)



*Rhododendron* plants declining from ramorum blight caused by *Phytophthora ramorum* in a commercial nursery. Inset, leaf symptoms of ramorum blight on *Rhododendron*.

by the California Citrus Nursery Board (CCNB) and the Citrus Research Board (CRB).

The CCPP operates at three locations: the Rubidoux Quarantine Facility in downtown Riverside, the Citrus Diagnostic Laboratory on the UC

Riverside campus and the Foundation and Evaluation Blocks at the UC Lindcove Research and Extension Center, in Exeter, California. The CCPP is supported by the CRB and CCNB (industry organizations founded in 1968 and 2005, respectively, in response to the California State Handler

Marketing Orders), while UC Riverside offers infrastructure support and scientific expertise. A committee of industry members (growers and nursery people) supports the CCPP activities.

The CCPP is the basic element of a long-term, multilevel program whose

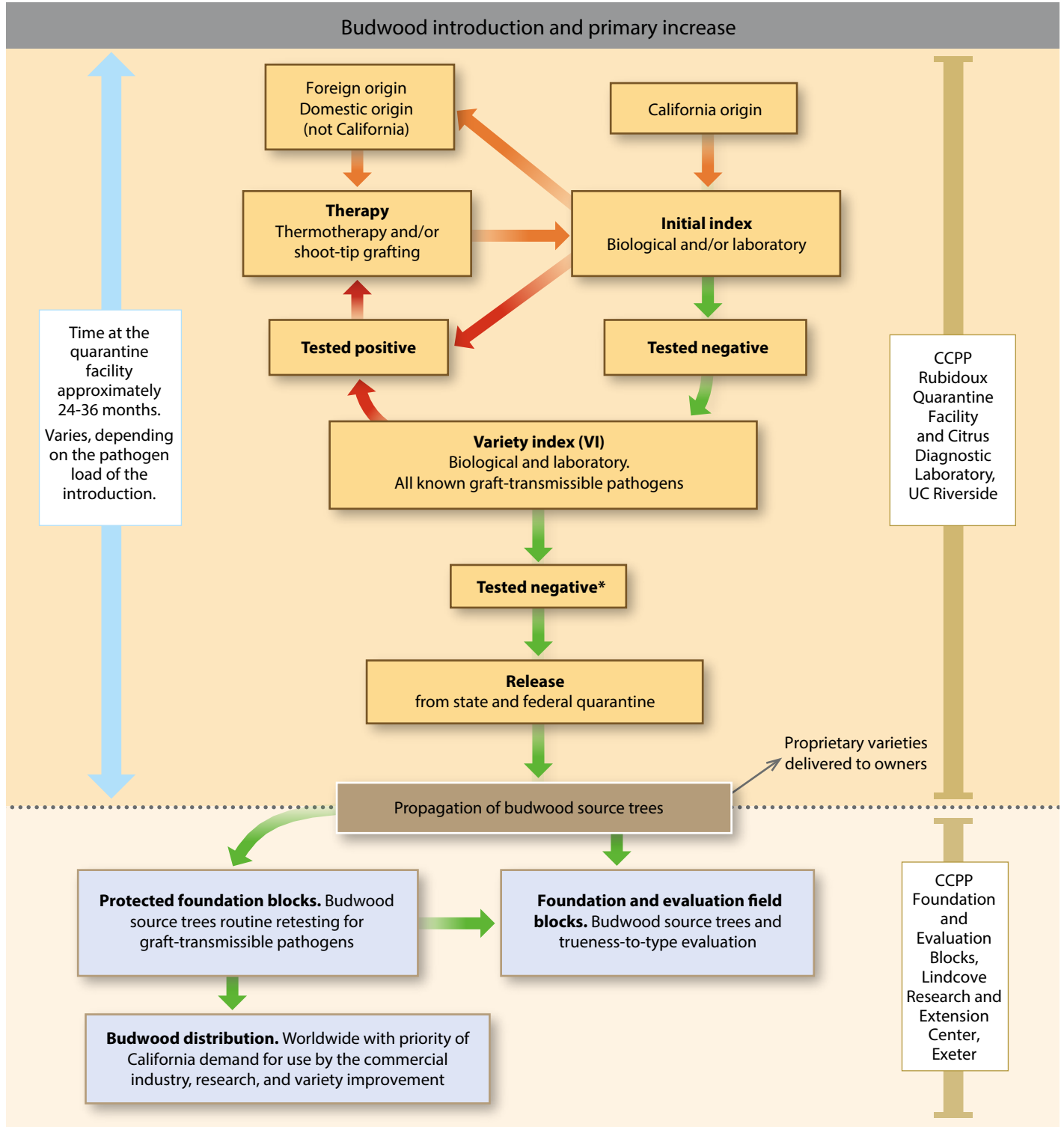


Fig. 4. Flow chart for citrus budwood introduction, propagation of source trees and distribution conducted by the Citrus Clonal Protection Program (CCPP). \* To fulfill movement requirements between quarantine zones, all materials must undergo therapy regardless of the pre-index results.



objectives include avoiding or restricting the spread of bud- and graft-transmitted pathogens of citrus in support of a profitable, competitive and sustainable citrus industry. As such, the CCPP provides a safe mechanism for the introduction of citrus varieties into California from any area around the world. The process for varietal introduction includes disease diagnosis and pathogen elimination followed by maintenance, pathogen retesting and distribution of true-to-type citrus propagative material (fig. 4).

The CCPP program of importation, production and distribution of pathogen-tested propagative materials is based on a comprehensive indexing (testing) program to detect graft-transmissible diseases and pathogens that may arrive in imported budlines. Graft-transmissible diseases may be caused by viruses, viroids or other pathogens (e.g., bacteria, phytoplasmas) and are vegetatively transmitted with an infected budline. Graft-transmissible diseases can seriously harm fruit quality, production and tree health

and longevity. In addition, diseases from infected field propagation may spread to neighboring orchards via insects or contaminated farm equipment (Timmer et al. 2000; Wallace 1978).

Disease diagnosis and pathogen detection take place in the insect-proof greenhouse and the Delfino Plant Laboratory at the Rubidoux Quarantine Facility as well as in the Citrus Diagnostic Laboratory at UC Riverside. Detection of graft-transmissible citrus diseases is based on a comprehensive indexing scheme that involves biological and laboratory diagnostics. For biological indexing, technicians graft tissue from the imported budline onto citrus indicator plants, with a specific citrus indicator for each specific disease. Indicator varieties have been selected for their sensitivity to diseases and their ability to express symptoms. For each index, technicians maintain adequate positive control plants under the same environmental conditions as the test indicators. The controls serve dual purposes: They provide a comparison for the test plant

and also demonstrate that environmental conditions in the greenhouse are optimal for plant growth and symptom expression (Childs 1978; Roistacher 1991; Vidalakis et al. 2004).

Citrus propagative material (i.e., budwood) distributed to homeowners, hobbyists, citrus growers and nurseries draws from tree sources of the CCPP foundation block. All CCPP- and nursery-owned citrus budwood tree sources must be established and propagated from citrus material that has been through the CCPP introductory, therapy and diagnostic protocols, regularly and routinely tested for several citrus pathogens, and registered as a budwood source with the CDFA. In May 2010, CDFA filed regulations for a mandatory Citrus Nursery Stock Pest Cleanliness Program as an emergency action. Under this new mandatory program, more than 8,000 registered budwood source trees were tested for several citrus pathogens in the first 2 years. The CCPP and UC Riverside researchers have been instrumental in the development of a



Elizabeth Grafton-Cardwell



Therese Kapatun



David J. Gumpf

**Fig. 5.** Panoramic view of the Citrus Clonal Protection Program Protected Foundation Block in the UC Lindcove Research and Extension Center in Tulare County. (A) The first screenhouse (40,000 sq ft) was constructed between 1998 and 1999, (B) the second screenhouse (30,000 sq ft) was completed in 2010 and (C) the positive pressure greenhouse (5,700 sq ft) was completed in 2011. The interior of the screenhouses with both (D) container and (E) in-ground budwood source trees.

high-throughput nucleic acid extraction and purification procedure optimized for citrus budwood tissues as well as molecular diagnostic methods for detection of citrus pathogens. These efforts have enabled the successful implementation of the new § 3701 CDFA Citrus Nursery Stock Pest Cleanliness Program (Vidalakis and Wang 2013; CDFA permits QC 1354 and QC 1388, [www.cdfa.ca.gov/plant/pe/nsc/nursery/citrus.html](http://www.cdfa.ca.gov/plant/pe/nsc/nursery/citrus.html)).

Since 2009, CCPP has supplied 154,000 buds from 301 different citrus varieties. A citrus nursery can produce as many as 300 trees within a year to 18 months from each CCPP bud. That means that an estimated 231 million citrus trees have been produced from pathogen-tested CCPP material in the past 5 years alone. If a single pathogen had been present in any of the buds used for this tree propagation, it would have been transmitted to millions of trees, with unknown horticultural, production and economic effects.

Diseases can sometimes become so important in citrus production that it may be necessary, for example, to change from a long-used rootstock to one that is more tolerant or resistant to a disease agent. Such was the case when citrus tristeza disease became a limiting factor for citrus production in California. Millions of trees growing on sour orange rootstock, which was susceptible to tristeza quick decline, had to be replaced with trees grown on tristeza-tolerant rootstocks of trifoliolate and trifoliolate hybrids. The availability of pathogen-tested tristeza-tolerant rootstocks and scions (e.g., mandarins) from the CCPP was critical to the industry's transition and subsequent economic success (Barnier et al. 2010; Calavan et al. 1978). The availability of a wide selection of citrus species, varieties and selections for evaluation and experimental use is essential to our ability to address new and emerging problems such as citrus greening.


Another important factor for the health and sustainability of the commercial citrus industry is maintenance of our capacity to import new varieties having different or improved fruit qualities or different maturity dates and organoleptic characters to satisfy ever-changing consumer demands. A capacity to import and maintain new pathogen-free citrus germplasm is imperative to keeping risk to the industry at a minimum and enabling

research to move forward. The only way we can meet these goals is through a program like the CCPP that works collaboratively with state and federal regulators, the citrus industry and UC to release pathogen-tested citrus varieties.

### Partnerships for plant health

The three programs described here demonstrate how investments in diagnostics, germplasm screening and development, and related training and education are helping to safeguard agriculture and plant resources at statewide, regional and national levels. These programs provide the agricultural industry with healthy planting stock as well as highly coordinated systems to deal efficiently with disease and pest outbreaks. Their success is due to the commitment of resources by the USDA, state agencies, industry and LGUs.

Especially critical to the success of these programs in California are the contributions of ANR and CDFA scientists and support staff. Yet recent limitations in state and federal funding for agricultural science have put the continuation of these programs in serious jeopardy. Reduced funding will weaken interagency collaboration and sharing of expertise, limit training and education programs, reduce

the speed with which samples can be processed and put our general preparedness to address plant health emergencies at risk. Increases in agricultural trade and plant importation, together with the importance of sustainable production systems for food and fiber, mean that the demand for integrated programs to provide accurate, rapid diagnoses in support of plant health has never been greater. 

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### References

- Barnier JB, Grafton-Cardwell E, Polek M. 2010. Citrus tristeza virus (CTV): Then and now. *Citrograph* 6:16–23.
- Calavan CE, Mather SM, McEachern EH. 1978. Registration, certification, and indexing of citrus trees. In: Reuther W, Calavan EC, Carman GE (eds.). *The Citrus Industry (Vol. IV): Crop Protection*. Berkeley, CA: University of California, Division of Agricultural Sciences. p 185–222.
- [CDFA] California Department of Food and Agriculture. 2013. California Agricultural Statistics Review 2012–2013. California Department of Food and Agriculture, Sacramento, CA. 131 p.
- Childs JFL (ed.). 1978. Indexing Procedures for 15 Virus Diseases of Citrus Trees. Agricultural Research Service–United States Department of Agriculture, Washington, DC.
- Crooks JA. 2005. Lag times and exotic species: The ecology and management of biological invasions in slow-motion. *Ecoscience* 12(3):316–29.
- Liebholt AM, Brockerhoff EG, Garrett LJ, et al. 2012. Live plant imports: The major pathway for forest insect and pathogen invasions of the US. *Front Ecol Environ* 10(3):135–43.
- Palm ME, Rossman AY. 2003. Invasion pathways of terrestrial plant-inhabiting fungi. In: Ruiz JM, Carleton JT (eds.). *Invasive Species: Vectors and Management Strategies*. Washington, DC: Island Press. p 31–43.
- Pimentel D, Zuniga R, Morrison D. 2005. Update on the environmental and economic costs associated with alien-invasive species in the United States. *Ecol Econ* 52(3):273–88.
- Rizzo DM, Garbelotto M, Hansen EA. 2005. *Phytophthora ramorum*: Integrative research and management of an emerging pathogen in California and Oregon forests. *Annu Rev Phytopathol* 43:309–35.
- Roistacher CN. 1991. Graft-transmissible diseases of citrus. Handbook for detection and diagnosis. Rome: Food and Agriculture Organization of the United Nations (FAO). [www.fao.org/docrep/t0601e/t0601e00.HTM](http://www.fao.org/docrep/t0601e/t0601e00.HTM).
- Rossman A. 2009. The impact of invasive fungi on agricultural ecosystems in the United States. *Biol Invasions* 11(1):97–107.
- Stack JP, Bostock RM, Hammerschmidt R, et al. 2014. The National Plant Diagnostic Network: Partnering to protect plant systems. *Plant Dis* 98:708–15.
- Stack J, Cardwell K, Hammerschmidt R, et al. 2006. The National Plant Diagnostic Network. *Plant Dis* 90(2):128–136.
- Timmer LW, Garnsey SM, Graham JH. 2000. Graft-transmissible, systemic diseases. In: *Compendium of Citrus Diseases*. St. Paul, Minnesota: APS Press. p 51–69.
- Vidalakis G, Garnsey SM, Bash JA, et al. 2004. Efficacy of bioindexing for graft-transmissible citrus pathogens in mixed infections. *Plant Dis* 88(12):1328–34.
- Vidalakis G, Wang J. 2013. Molecular method for universal detection of citrus viroids. US Patent No. 20,130,115,591.
- Wallace JM. 1978. Virus and viruslike diseases. In Reuther W, Calavan EC, Carman GE (eds.). *The Citrus Industry (Vol. IV): Crop Protection*. Berkeley, CA: University of California, Division of Agricultural Sciences. p 69–72.



When new pests or diseases become established, they often lack the natural control of their native environments and develop into more serious problems when interacting with local plant or animal systems or local management practices. The response to constrain and manage them must be complex and coordinated between research institutions and scientists, government agencies and the agricultural community. The articles in this section profile two successful multi-partner responses to newly established threats to one of California's premier commodities.

# Growers, scientists and regulators collaborate on European grapevine moth program

by Monica Cooper, Lucia Varela, Rhonda Smith, David Whitmer, Gregory Simmons, Andrea Lucchi, Roxanne Broadway and Robert Steinhauer

*The first detection of the European grapevine moth in North America triggered the establishment of federal and state regulatory programs that (1) identified the insect's geographic range in California, (2) developed and implemented detection and management programs, (3) regulated the movement of plant material and equipment to minimize the threat of dispersal, (4) incorporated research-based information developed by subject-matter experts into policy decisions and (5) promoted a wide-reaching educational program for grape growers, the public and local officials. The action plan, developed and carried out through a coordinated program that included multiple government agencies, university scientists and the agricultural community, drastically reduced insect populations and limited the distribution in California vineyards such that some previously infested areas were removed from quarantine regulation.*

Invasive species increasingly threaten agricultural sustainability in a global economy. If an invasive species is a known pest or assessed as potentially threatening, its detection in California may trigger a regulatory response coordinated by the U.S. Department of Agriculture (USDA), the California Department of Food and Agriculture (CDFA) and agricultural commissioners. Regulatory programs often encompass activities such as trapping, quarantine and treatment protocols, and depend on reliable scientific information generated by university researchers. European grapevine moth, *Lobesia botrana* (Denis & Schiffermüller), is endemic to Mediterranean Europe, has invaded portions of the Palearctic region (Europe, west Asia and North Africa) and East Africa, and was detected for the first time in the Americas in Chile, April 2008; in California, September 2009; and in Argentina, April 2010 (Ioriatti et al. 2012; Varela et al. 2013a). In the United States,

the first detections were in Napa County, California, where this invasive pest and associated fungal rot caused significant crop damage in 2009. A coordinated program by USDA, CDFA, county agricultural commissioners, UC, international

scientists and grape growers aimed to control, contain and potentially eradicate insect populations. The cooperative effort to develop and implement this program is the focus of this article.

### Seasonal biology in California vineyards

The European grapevine moth (EGVM) may complete two to five annual generations, as determined by latitude, climate and microclimate (Ioriatti et al. 2011); temperature models predict three generations per year in the Napa Valley, which has been validated through ground observations (Gutierrez et al. 2012).

Pupae overwinter in diapause (a resting state) inside silken cocoons in protected locations, such as under the bark of the vine. The first male flight generally begins slightly before budbreak and may continue for 10 to 14 weeks. Adults generally fly at dusk when temperatures



Jack Kelly Clark

In 2009, European grapevine moth was first detected in the United States in Napa County, where it caused significant crop damage.

Online: <http://californiaagriculture.ucanr.edu/landingpage.cfm?article=ca.v068n04p125&fulltext=yes>  
doi: 10.3733/ca.v068n04p125

Monica Cooper



Feeding damage by European grapevine moth larva to a grape flower before bloom. Note the characteristic hole in the flower, webbing and larval excrement.

Andrea Lurchi



Feeding damage to grape berries by second- and third-generation larvae exposes them to fungal infections that can be economically damaging.

are above 53.5°F (12°C). During the first flight, the female glues single eggs to flat surfaces on or near the flower cluster; second- and third-generation eggs are laid on grape berries. Larvae form webbed nests; the first-generation larvae feed on flowers before and during bloom in Northern California; second-generation larvae feed on green berries; and third-generation larvae feed inside ripening berries. Larvae create distinctive round holes in prebloom flowers and ripening fruit, which distinguishes their feeding damage from that of other common Lepidoptera larvae found in California vineyards (photo, left). Feeding damage to berries by second- and third-generation larvae exposes them to infection by *Botrytis* and other secondary fungi (photo, below left) that can be economically damaging.

### Quarantine programs

On October 7, 2009, USDA confirmed the presence of EGVM in Napa County. A federal order issued by USDA in June 2010 initiated a quarantine area within 5 miles (8 kilometers [km]) of all detections (USDA 2010). Detections were defined as two or more adult moths trapped within 3 miles (5 km) of each other during the same life cycle or immature stages confirmed to be EGVM by DNA analysis. The order indicated plant host species as well as plant parts, products, farming and processing equipment, and green waste residues as regulated articles that could not be transported interstate from a quarantine area except under specific conditions. The state interior quarantine (CDEA 2012) enforced restrictions parallel to those in the federal order for intrastate movement of regulated articles within or from quarantine areas. In 2012, the quarantine buffer was reduced from 5 miles to 3 miles (5 km) around detections. This change accommodated the program’s need to reduce the cost of implementation while acknowledging the adult moths’ short-distance natural dispersal (Boller 1993; Schmitz et al. 1996).

### Statewide survey and detection program

Immediately after the first confirmed detection in 2009, USDA, CDEA and the Napa County agricultural commissioner deployed 248 sex pheromone-baited traps to delimit the population. However, very few moths were caught because traps were deployed at the end of the third flight (table 1). In addition to the trap captures, ground surveys in 2009 recorded 26 larvae, eight

TABLE 1. European grapevine moths captured in pheromone-baited traps, presented by California county and by year (2009–2014)

Year	Napa	Sonoma	Solano	Mendocino	Fresno	Merced	San Joaquin	Santa Cruz	Santa Clara	Monterey	Nevada
..... Number of male moths (number of traps) .....											
2009	5 (248)										
2010	100,831 (3,882)	59 (6,932)	11 (1,514)	36 (1,594)	11 (8,648)	4 (860)	2 (3,522)	1 (449)	3 (596)	1 (1,733)	0 (55)
2011	113 (4,930)	9 (9,048)	0 (2,644)	0 (2,237)	0 (11,013)	0 (1,502)	0 (7,537)	1 (552)	19 (1,346)	0 (2,651)	4 (1,902)
2012	77 (4,706)	0 (8,393)	0 (1,844)	0 (1,432)	0 (8,630)	0 (86)	0 (4,714)	0 (318)	0 (658)	0 (2,033)	0 (920)
2013	40 (11,621)	0 (6,906)	0 (1,383)	0 (1,430)	0 (7,651)	0 (1,265)	0 (1,301)	0 (202)	0 (267)	0 (1,998)	0 (60)
2014*	0 (11,574)	1 (7,011)	0 (ND)	0 (1,468)	0 (2,169)	0 (828)	0 (4,243)	0 (ND)	0 (ND)	0 (1,978)	0 (ND)

\* These are preliminary values, as of July 30, 2014; final values will be available in November 2014; ND = no preliminary data available.



pupae and one female at multiple sites in two distinct areas of Napa County. As a result, the state interior quarantine was established in March 2010, over an area totaling 162 square miles (420 square kilometers [km<sup>2</sup>]) in Napa County (fig. 1).

In February 2010, trapping efforts expanded throughout all grape-growing regions of California — roughly 803,000 acres (325,000 hectares [ha]). Traps were deployed at densities of nine to 16 or 25 traps per square mile of planted vineyard (three to six or 10 traps per km<sup>2</sup>) outside and inside the regulated area, respectively (table 1). In select urban areas, traps were placed on potential EGVM host plants at a density of five traps per square mile (two traps per km<sup>2</sup>) (Mastro et al. 2010). By the end of 2010, the quarantine area included portions of eight California counties, totaled 2,091 square miles (5,416 km<sup>2</sup>) and contained approximately 150,760 acres (61,010 ha) of vineyards (fig. 2, table 2). Subsequently, in 2011, traps detected moths in two additional counties, bringing the total number of regulated counties to 10 and a peak quarantine area of 2,335 square miles (6,048 km<sup>2</sup>) (fig. 2, table 2). Trap captures in Napa County indicated a large, widely distributed population, whereas populations in other counties were significantly smaller and more contained (table 1).

The EGVM regulatory program has relied heavily on the use of pheromone-baited sticky traps to detect moth populations. UC scientists evaluated the efficacy and longevity of four pheromone lures in replicated field experiments in Napa during the first and second moth flights of 2010 (Varela et al. 2013b). All lures were

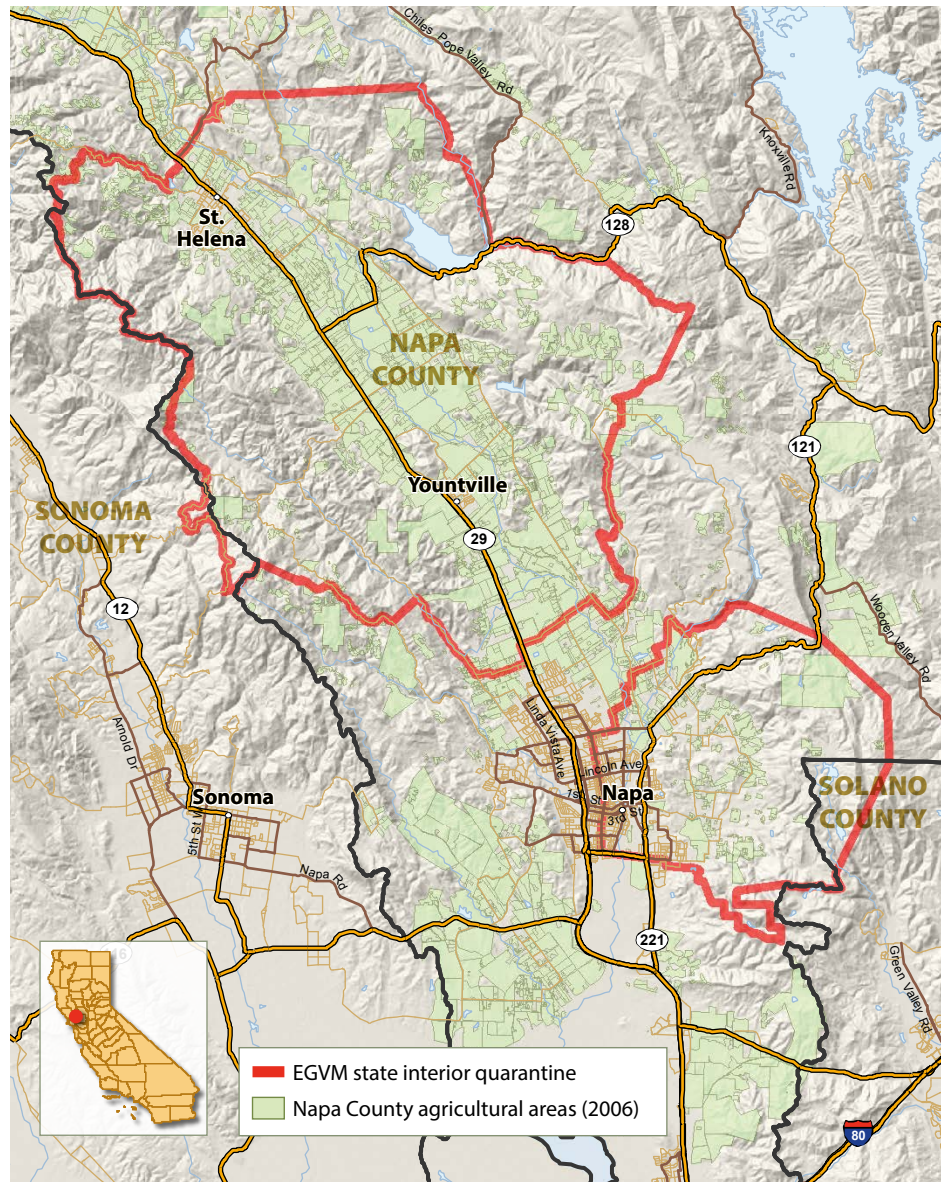


Fig. 1. The state interior quarantine established in Napa County, California, following the first detection of EGVM (September 29, 2009) in North America. The area encompassed all areas within a 5-mile (8 km) radius of all known EGVM populations at that time, as determined by trap captures and ground surveys.

TABLE 2. Quarantine area in square miles (vineyard acres inside quarantine) by county and year

Year	Napa	Sonoma	Solano	Mendocino	Fresno	Merced	San Joaquin	Santa Cruz	Santa Clara	Nevada	California total
.....Square miles (vineyard acres).....											
2010	597 (43,139)	664 (52,000)	237 (2,397)	179 (5,860)	96 (24,769)	108 (1,432)	96 (20,544)	0 (0)	94 (619)	0 (0)	2,091 (150,760)
2011	597 (43,452)	664 (52,000)	237 (2,397)	179 (5,860)	96 (24,769)	108 (1,432)	96 (20,544)	87 (310)	94 (619)	176 (345)	2,335 (151,728)
2012	575 (43,078)	458 (46,500)	124 (2,289)	Removed from quarantine 3/8/2012				34 (231)	38 (552)	74 (99)	1,302 (92,749)
2013	554 (42,703)	78* (5,600)	55† (1,009)	Removed from quarantine 3/8/2012				Removed from quarantine 12/21/2012			687 (49,312)
2014	554 (42,703)	78 (5,600)	55 (1,009)	Removed from quarantine 3/8/2012				Removed from quarantine 12/21/2012			687‡ (49,312)

\* In Sonoma County, 380 square miles were removed from quarantine on 12/21/2012.

† In Solano County, 69 square miles were removed from quarantine on 8/24/2012.

‡ Effective August 2014: the quarantine area measures 446 square miles, following the removal of Solano County and portions of Napa and Sonoma counties.



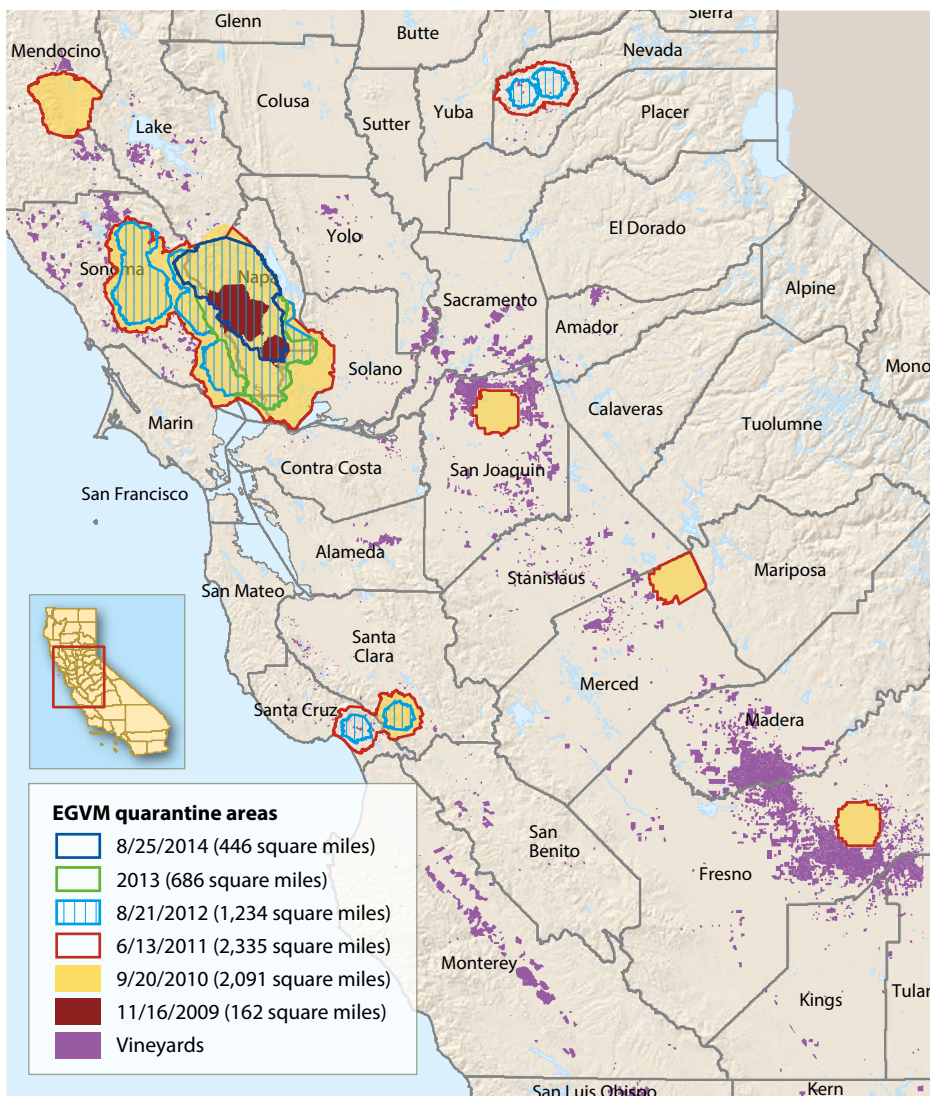


Fig. 2. Regulated areas of California for EGVM, defined by USDA, 2009 to 2014.



The distinctive color pattern on EGVM wings make them easily recognizable in pheromone-baited traps.

effective for monitoring EGVM male moth populations during and beyond the period recommended by the manufacturer (fig. 3).

### International technical working group

In November 2009, USDA assembled a technical working group (TWG) of subject-matter experts to provide urgent scientific recommendations to regulatory program managers in California. The TWG included university scientists from Italy, France, Germany, Spain, Chile and California, as well as USDA scientists (and chair) and a representative of the wine grape industry. TWG members continue to meet annually to review program activities and provide technical expertise on topics as diverse as insect biology, detection strategies, handling of harvested fruit and winery waste, and management activities.

Since 2010, TWG members have agreed that eradication of EGVM from California remains a realistic goal as long as (1) the population did not become substantially more widespread than was known at that time, (2) the grape industry remained supportive of the effort and (3) effective control methods were available for use by the program (Mastro et al. 2010). Over the course of the EGVM program, TWG members evaluated research data and program developments to formulate recommendations based on the insect's biology. Government agencies matched these recommendations to political and fiscal analyses to formulate and deliver the EGVM program.

### Management tools

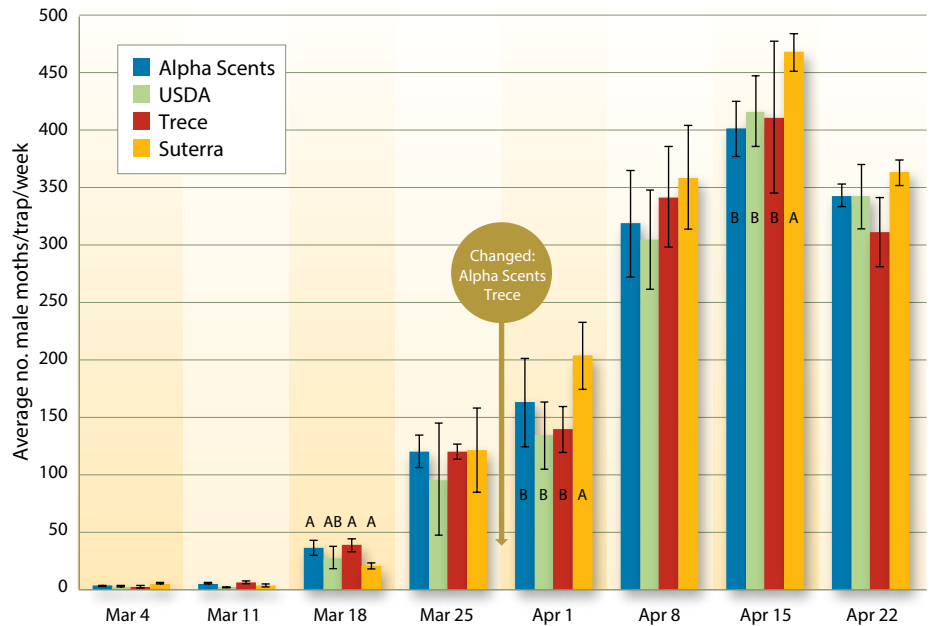
**Treatment areas.** Treatment areas were defined within a requisite distance from all detections and included agricultural, residential and commercial properties. In 2010, the distance was not standardized, so the size of the treatment areas varied by county. Following a review of the scientific literature suggesting that EGVM are short-distance fliers, the TWG recommended a treatment area within 1,640 feet (500 meters [m]) of all detections regardless of when the detection occurred. From 2013 onward, the TWG modified this recommendation to account for timing of the detection: Treatment areas came to be defined as within 500 m of detections occurring in the current year and previous 2 years. Within the treatment areas,



grape and olive were the targeted hosts of concern, and total vineyard acreage varied considerably by county and year (table 3). The specific combination of tools (insecticides, mating disruption and host removal) used in treatment areas differed by land use and occurrence of potential host plant species.

**Insecticide program.** The recommended insecticide program for EGVM in California vineyards targets the eggs and larvae and includes at least one application of a conventional insecticide or at least two applications of an organic insecticide for each of the first two generations (and for the third generation in extenuating circumstances). Although treatment of the first generation is not typical in the Palearctic regions, the TWG determined that treating the first two generations in California would provide the greatest opportunity to eradicate populations. In early 2010, UC scientists provided an exhaustive list of potential insecticides for EGVM management based on a review of the scientific literature. From this list, EGVM program leaders made a concerted effort to identify and recommend products that would provide selective control of EGVM while minimizing risks to non-target organisms and the environment. The availability of organic treatment options ensured that growers could maintain organic certification while complying with the eradication effort. Pesticide use reporting (PUR) data for Napa County (2010 to 2014) indicate that growers used a combination of the recommended materials (table 4).

**Insecticide efficacy trials.** UC scientists conducted field trials in commercial vineyards in Napa in 2010 to evaluate the efficacy of registered insecticides for EGVM.



**Fig. 3.** Average number of male moths caught in traps baited with commercial pheromone lures and a USDA lure monitored from February 25 to April 22, 2010. Alpha Scents, USDA and Trece lures are rubber septa, and Suterra is a membrane lure. Letters indicate homogenous groups at 95% least significant difference. Per the manufacturer's recommendation, the Alpha Scents and Trece lures were changed 4 weeks into the experiment.

Chlorantraniliprole, methoxyfenozide, spinosad and *Bacillus thuringiensis* provided control of young larvae; abamectin, indoxacarb and spinetoram provided the best control of mature larvae (Van Steenwyk et al. 2011). These results were widely distributed to the program team and grape growers.

**Insecticide treatment.** Selective insecticides are most effective if applied when the pest is at its most susceptible stage (Ioriatti et al. 2011). In Napa County, UC scientists monitored the male flight, egg and larva development and calculated degree-days (lower and upper thresholds of 50°F and 86°F [10°C and 30°C], respectively) from a biofix of January 1 for

each life stage (Caffarelli and Vita 1988; Touzeau 1981). Referencing these observations and calculations to local weather data and vine phenology, UC scientists then determined the optimal timing for insecticide applications for all affected locations in California. For each generation, a 3-week treatment window minimized application costs by combining the insecticide with preventative treatments for powdery mildew. This information was widely disseminated via conference calls with government agencies, a UC electronic newsletter, industry associations' e-news blasts, and grower liaisons in Napa, Sonoma, Fresno and Mendocino counties. An analysis of the PUR data

**TABLE 3.** Insecticide-treated acres and reported use of Isomate EGVM pheromone dispensers in counties affected by quarantine regulations

Year	Napa	Sonoma	Solano	Mendocino	Fresno	Merced	San Joaquin	Santa Cruz	Santa Clara	Nevada
..... Insecticide-treated acres (acres under mating disruption) .....										
2010*		~8,000 (0)	594 (0)	94 (13)	928 (0)	620 (0)	364 (0)	N/A	N/A	N/A
2011†	23,700 (13,300)	2,395 (1,533)	785 (0)	201 (100)	540 (0)	200 (0)	83 (0)	16 (0)	114 (0)	8 (8)
2012‡	28,000 (23,071)	2,395 (0)	N/A	N/A	N/A	N/A	N/A	16 (0)	114 (0)	8 (0)
2013§	11,800 (2,800)	23 (0)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A

\* Treatment areas are within 1,000 m of any detection in 2010 (except Napa and Solano [200 m]).

† Treatment areas are within 500 m of any detection in 2010 or 2011.

‡ Treatment areas are within 500 m of any detection in 2010, 2011 or 2012.

§ Treatment acres are within 500 m of any detection in 2011, 2012 or 2013.

from 2011 and 2012 shows a high level of compliance for treatment timing among Napa County growers (table 5).

**Mating disruption.** Mating disruption (MD) programs deploy synthetic (*E,Z*)-7,9-dodecadienyl acetate — the main component of the female sex pheromone — in hand-applied dispensers (Pasquier and Charmillot 2005). When applied as an area-wide control strategy in Palearctic

regions, MD has provided sustained control of EGVM populations while decreasing reliance on insecticides and reducing conflict between agricultural and urban populations (Ioriatti et al. 2011). Although MD does not completely inhibit EGVM mating, delayed mating reduces populations because older females produce fewer eggs than younger females (Torres-Vila et al. 2002). TWG scientists strongly

supported the use of MD as a control tool (Ioriatti et al. 2004; Lucchi et al. 2012), and by the second EGVM flight of 2010, Isomate EGVM pheromone dispensers (Pacific Biocontrol, Vancouver, WA) became widely used (table 3).

After the MD and insecticide programs were implemented, trap catches and visual inspections revealed dramatic decreases in population size in Napa County (table 1). Beginning in 2012, all Napa County vineyards within the defined treatment areas received pheromone dispensers through federal, state and local funding programs. Because MD limits the reliability of sex pheromone-baited traps and makes it very difficult to detect residual populations, the EGVM program has avoided the use of MD in treatment areas as they transition to deregulation.

**Urban and residential treatment programs.** CDEA personnel used multiple strategies to manage EGVM populations in noncommercial grapevines in urban and residential areas. The organic product *B. thuringiensis* was applied during the first and second generations if the crop was to be harvested; if not, flower and/or fruit clusters were removed in the spring or early summer (table 6). MD was also used in certain areas and select counties.

#### Alternate host surveys

Polyphagy by EGVM has been documented in the literature: Larvae may feed on up to 40 hosts in 27 plant families (Ioriatti et al. 2011; Lucchi and Santini 2011). However, larvae are rarely found on hosts other than *Daphne gnidium* and *Vitis*; exceptions seem to result from adaptations to local climate and flora (Ioriatti et al. 2011) or elevated pest pressure and presence of ripe fruit (Maher 2002). UC and USDA personnel used pheromone-baited traps and visual surveys to monitor a variety of plant species in Napa County that are reported to be EGVM hosts (table 7); no EGVM life stages were found during these surveys, suggesting that these species currently pose little risk in California. In separate surveys of olive orchards, 10 eggs and 12 EGVM larvae were found, indicating that olive flowers were a minor host in Napa during the first EGVM generation, but olive fruit did not host the second or third generations. The main host of consequence in California continues to be cultivated grape, *Vitis vinifera*.

**TABLE 4. Pesticide use report data collected by the Napa County agricultural commissioner for applications targeting EGVM during the first and second generations of 2011 and 2012 within defined treatment areas (all vineyards within 500 m of all EGVM detections)**

First generation	Second generation	Year	Acres treated	Percent of total acres	Sites treated	Percent of total sites
Conventional	Conventional	2011	11,111	46.9	593	48.4
		2012	11,597	60.9	444	52.5
Organic	Organic	2011	2,073	8.7	113	9.2
		2012	2,492	13.0	115	14.0
Conventional or organic	Conventional or organic	2011	41	0.17	4	0.33
		2012	195	1.0	15	1.8
Insecticide	None	2011	5,290	22.3	194	15.8
		2012	1,063	5.6	83	9.8
None	Insecticide	2011	1,783	7.5	73	6.0
		2012	814	4.3	51	6.0
None	None	2011	3,399	14.3	249	20.3
		2012	2,869	15.1	138	16.3

Conventional insecticides: abamectin, chlorantraniliprole, and methoxyfenozide; organic insecticides: spinosad and *Bacillus thuringiensis*. "None" indicates that no insecticide targeting EGVM was reported.

**TABLE 5. Timing of reported insecticide treatments in Napa County targeting the first and second generations of 2011 and 2012 categorized as: recommended timing, before or after**

Product	Year	Acres treated, by treatment timing for first generation (percent of total)			Acres treated, by treatment timing for second generation (percent of total)		
		Before	Recommended	After	Before	Recommended	After
Chlorantraniliprole (Altacor)	2011	—	444 (87)	65 (13)	1,109 (10)	9,639 (89)	68 (1)
	2012	—	2,803 (91)	286 (9)	989 (11)	6,254 (67)	2,072 (22)
Methoxyfenozide (Intrepid)	2011	3 (< 1)	13,085 (90)	1,558 (10)	165 (10)	1,432 (89)	7 (< 1)
	2012	299 (2)	11,771 (86)	1,606 (12)	181 (4)	2,853 (63)	1,509 (33)
<i>Bacillus thuringiensis</i> and/or spinosad	2011	0.3 (< 1)	2,752 (93)	195 (7)	225 (10)	1,968 (88)	35 (2)
	2012	210 (6)	3,146 (85)	346 (9)	745 (22)	2,464 (73)	152 (5)
Abamectin (Agri-Mek)	2011	—	66 (89)	8 (11)	—	324 (100)	—
	2012	12 (9)	110 (91)	—	16 (41)	23 (59)	—
Flubendiamide	2011	—	—	—	—	—	—
	2012	—	—	91 (14)	540 (86)	—	—



**TABLE 6. Treatments for EGVM in noncommercial grapevines in urban areas included flower/fruit removal, applications of *Bacillus thuringiensis* (Bt) and mating disruption (MD)**

Year	Treatment	Number of properties by California county									
		Fresno	Mendocino	Merced	Napa	Nevada	San Joaquin	Santa Clara	Santa Cruz	Solano	Sonoma
2010	Fruit removal	24	168	1			7			115	
	Bt	2		1			1				
2011	Fruit removal	23	148	1	189		1	9	14	306	165
	Bt	7	42	1			12	13	4	63	34
	MD				2,651	601					
2012	Fruit removal					112		8	11	294	200
	Bt						36	14	6	75	147
	MD				860						
2013	Fruit removal				157						
	Bt				121						
	MD				373						

### Compliance agreements

To prevent the movement of EGVM on regulated articles grown inside quarantine areas, CDFA required businesses to sign compliance agreements that mandated specific activities prior to and during harvest, transport, processing and waste handling. Compliance agreements were also required for similar activities in raisins and regulated fruit other than grape, including olive, persimmon, pomegranate, most stone fruit (*Prunus* spp.) and specific caneberries. Based on recommendations provided by the TWG, in 2012 the USDA revised the list of regulated articles to exempt olive fruit and *Rubus* spp., and limited the acreage of *Prunus* spp. affected.

### Management of winery waste

The possibility that EGVM could survive in unfermented winery waste was addressed by requiring that waste be composted on site, transported to an approved compost facility or returned to the vineyard of origin, depending on where the fruit was sourced relative to the quarantine area and county. Alternatively, if grapes were pressed to a minimum of 2 bars (0.2 MPa) or 28 psi, then movement of waste was unrestricted within California. Investigations provided evidence that EGVM larvae could survive on unpressed green waste after destemming and determined that in general the processing equipment was likely a greater source of contamination than unfermented waste (Smith et al. 2013). Mature larvae may move from clusters to protected locations, emphasizing the need for thorough

washing of all equipment used to harvest, transport and process infested loads of grapes.

### Outreach and educational program

EGVM program leaders provided transparent, consistent, timely and coordinated communication to parties directly and indirectly affected by the EGVM program. USDA led international communications and jointly with CDFA coordinated statewide communication. County agricultural commissioners and UC advisors directed local communication and assisted in other efforts. The outreach program also collected relevant information from stakeholders to ensure the appropriateness of regulatory requirements and adapted the program to local conditions and concerns. This openness and flexibility to change fostered the development of trust, respect and cooperation among all parties. The proactive, local campaign to communicate, educate and collaborate with a diverse community became a hallmark of the EGVM program (Zalom et al. 2013).

**Industry outreach and education.** Many methods were used to communicate information to growers and winery personnel. Educational materials included two tri-fold brochures, a poster and training videos developed and distributed through UC Cooperative Extension. Mass marketing campaigns raised public awareness using postcards, door-hangers, magnets, billboards and campaign signs, public service announcements, and online and social media; cross-linked websites provided comprehensive and current

**TABLE 7. Surveys of alternate host plants along the Napa River, 2010–2011**

Plant type	Number of fruit/flowers inspected
Blackberry	55,625
Elderberry	230,615
Olive	3,937 (and 4,837 leaves)
Wild rose	2,962
Wild or domestic grape	28,471
Wild or domestic plum	2,582
Peach	83
Nectarine	83
Walnut	226
Blueberry	305
Gooseberry	342

information. The online UC IPM Pest Alert presented information on pest identification, biology and management.

Grower liaison/outreach coordinators in Napa, Sonoma, Mendocino and Fresno counties played an important role in outreach and educational efforts. In each county, the EGVM grower liaison conducted targeted outreach and was a trusted, independent source of information for growers and winery personnel. Program information was communicated at meetings, seminars and field days, as well as through individual communications and a UC Cooperative Extension Napa County newsletter. Farmworkers — particularly important “first responders” capable of identifying potential threats during their daily work — were reached through Spanish language presentations, field days and outreach materials. Partnerships with local industry groups



The public campaign "Kick the Moth Out" raised awareness of the EGVM program in Napa County.

to deliver program information contributed to the educational efforts. Growers also shared information and related their experiences through peer networks that strengthened the formal educational program.

**Public outreach and education.** An estimated 2.5 million acres of California farmland are adjacent or in close proximity to nonfarm residences (Hammond et al. 2010). To address potential areas of urban-agricultural conflict, county agricultural commissioners supported by USDA, CDFA, UC and industry groups worked within established local networks of community leaders to develop a public outreach campaign. County supervisors and city council members, environmental, community and commercial organizations, residents and tourists were the targets of the outreach efforts, which built trust between program and community leaders, growers and the public.

#### Program update

The conditions set forth by the TWG specified that in areas attempting to qualify for deregulation (1) no moths must be captured during five consecutive generations, (2) insecticide treatments must continue to target the first and

second generation, (3) MD may not be used during the final two generations that the area is under regulation and (4) during these two generations, trap density must increase to 100 traps per square mile (39 traps per km<sup>2</sup>) (one trap per 6 acres) in all vineyards within 1,640 feet (500 m) of previous detections. Under California conditions, only the first and second are considered full generations because a proportion of the second generation enters diapause (L. Varela and M. Cooper, personal observation). Four counties (Fresno, Mendocino, Merced and San Joaquin) were removed from regulation at the beginning of 2012, and by the end of 2012, five additional counties (Nevada, Santa Clara, Santa Cruz, Solano and Sonoma) had been deregulated partially or in full.

Napa County and portions of neighboring Sonoma and Solano counties remained under regulation in 2013, an area encompassing 686 square miles (fig. 2). Due to the historically large and widely distributed populations in Napa County, the TWG recommended a revised approach to deregulation: High-density traps (100 per square mile) deployed county-wide must be free from detections during four full flights before areas will be considered for deregulation (Mastro

et al. 2012). Any moth captured will trigger the delimitation and establishment of treatment areas within 1,640 feet (500 m) of the detection. Consequently, trap density nearly tripled in Napa County in 2013 (table 1) and the area under MD shrank from 23,000 to 2,800 acres in 2013 and approximately 1,907 acres in 2014. The remaining EGVM population in Napa County has been drastically reduced in size and distribution (fig. 2; table 1).

By mid-August 2014, southern Sonoma County, the remainder of Solano County, and a portion of southern Napa County were removed from quarantine. The remainder of Napa County and a portion of northern Sonoma County, an area totaling 446 square miles, will continue to be regulated in 2015. Pheromone-baited trap surveys continue in other grape-growing areas of California and the United States as part of CDFA/USDA early detection programs. These surveys detected one moth in Sonoma County in 2014; this was not sufficient to trigger establishment of a new quarantine boundary.

#### A model collaborative effort

Development and implementation of a successful regulatory program in response to the detection of an invasive species in California agricultural systems requires a concerted and coordinated effort to address the pest while balancing the needs of the agricultural industry, trading partners and the general public. Regulatory, fiscal, environmental and biological aspects must be weighed to develop goals and determine program activities. The leaders of the California EGVM program were sensitive to these issues and gained credibility and trust by involving a diverse community for dialogue, responding to the needs of local communities, considering the impact on the environment, adopting new scientific information, investing in relationships and networks, and ensuring the appropriateness of regulatory requirements (Zalom et al. 2013). Factors such as the insect's limited host range, which allowed the program to focus on commercial vineyards; the use of management tools that minimized the impact to nontarget organisms and are compatible with organic and backyard vineyards; and the long history of EGVM management in the Palearctic region also contributed to the successes of the program. The EGVM program



leaders were recognized with the USDA Administrator's Award in September 2012, acknowledging the program's successful collaborative approach. The program has clear direction and well-defined goals, is grounded in biology, engages a diverse community and is responsive to the changing needs of participants. The participation of USDA, Cdfa, agricultural commissioners, UC and other university scientists, growers, industry groups, community leaders and the general public resulted in a model effort that has reduced EGVM populations to a few areas of Napa and Sonoma counties. [CA](#)

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Field day training sessions with growers and farmworkers were held in Oakville, CA, in April 2010.

## References

Boller EF. 1993. Wie weit fliegt der Traubenwickler? Deut. Weinbau Jahrbuch 44, Waldkircher Verlag, Waldkirch, Germany.

Caffarelli V, Vita G. 1988. Heat accumulation for timing grapevine moth control measures. Bulletin SROP 11:24-6.

[Cdfa] California Department of Food and Agriculture. 2012. European grapevine moth (*Lobesia botrana*) state interior quarantine. In: California Department of Food and Agriculture Plant Quarantine Manual 3437:422.1-12. <http://pi.cdfa.ca.gov/pqm/manual/pdf/422.pdf>.

Gutierrez AP, Ponti L, Cooper ML, et al. 2012. Prospective analysis of the invasive potential of the European grapevine moth *Lobesia botrana* (Den. & Schiff) in California. *Agr Forest Entomol* 14:225-38.

Hammond SV, Norton MV, Schmidt EE, Sokolow A. 2010. California communities deal with conflict and adjustment at the urban-agricultural edge. *Calif Agr* 64:121-8. doi: 10.3733/ca.v064n03p121.

Ioriatti C, Anfora G, Tassin M, et al. 2011. Chemical ecology and management of *Lobesia botrana* (Lepidoptera: Tortricidae). *J Econ Entomol* 104:1125-37.

Ioriatti C, Bagnoli B, Lucchi A, et al. 2004. Vine moths control by mating disruption in Italy: Results and future prospects. *REDIA* 87:17-28.

Ioriatti C, Lucchi A, Varela LG. 2012. Grape berry moths in western European vineyards and their recent movement into the New World. In: Bostanian NJ, Vincent C, Isaacs R (eds.). *Arthropod Management in Vineyards: Pests, Approaches, and Future Directions*. Springer Science + Business Media B.V. p 339-59. doi: 10.1007/978-94-007-4032-7\_14.

Lucchi A, Bagnoli B, Cooper M, et al. 2012. The successful use of sex pheromones to monitor and disrupt mating of *Lobesia botrana* in California. Working group meeting on pheromones and other semio-chemicals in integrated production, October 1-5, 2012, Bursa, Turkey. IOBC-WPRS Bulletin 99:45-8.

Lucchi A, Santini L. 2011. Life history of *Lobesia botrana* on *Daphne gnidium* in a natural park of Tuscany. IOBC-WPRS Bulletin 67:197-202.

Maher N. 2002. Oviposition site selection by *Lobesia botrana* (Lepidoptera: Tortricidae): Influence of non-volatile chemical cues from host plants fruits [in French]. PhD thesis, Université de Bordeaux 2, France. 109 p.

Mastro V, Cardé R, Cooper M, et al. 2012. Report of the Technical Working Group for the European Grapevine Moth Program in California. October 30-November 1, 2012, Sacramento, CA. [www.aphis.usda.gov/plant\\_health/plant\\_pest\\_info/eg\\_moth/downloads/TWG-Report-Dec2012.pdf](http://www.aphis.usda.gov/plant_health/plant_pest_info/eg_moth/downloads/TWG-Report-Dec2012.pdf).

Mastro V, Cardé R, Lance D, et al. 2010. Second Report of the USDA International Technical Working Group for the European Grapevine Moth in California. May 14, 2010. [www.aphis.usda.gov/plant\\_health/plant\\_pest\\_info/eg\\_moth/downloads/TWG%20report-5-14-2010.pdf](http://www.aphis.usda.gov/plant_health/plant_pest_info/eg_moth/downloads/TWG%20report-5-14-2010.pdf).

Pasquier D, Charmillot PJ. 2005. Survey of pheromone emission from different kinds of dispensers used for mating disruption in orchards and vineyards. IOBC-WPRS Bulletin 28:335-40.

Schmitz V, Roehrich R, Stockel J. 1996. Dispersal of marked and released adults of *Lobesia botrana* in an isolated vineyard and the effect of synthetic sex pheromone on moth movements. *J Int Sci Vigne Vin* 30: 67-72.

Smith RJ, Cooper ML, Varela LG. 2013. Determining the threat of dispersal of *Lobesia botrana* larvae in infested grapes processed for winemaking. *Practical Winery and Vineyard* August 2013:73.

Torres-Vila LM, Rodríguez-Molina MC, Stockel J. 2002. Delayed mating reduces reproductive output of female European grapevine moth, *Lobesia botrana* (Lepidoptera: Tortricidae). *B Entomol Res* 92:241-9.

Touzeau J. 1981. Modelisation de l'évolution de l'Eudémis de la vigne pour la région Midi-Pyrénées. *Bol Zool Agr Bach* 11 16:26-8.

USDA. US Department of Agriculture. 2010. Establishment of Quarantine Areas in California for European Grapevine Moth, *Lobesia botrana*. APHIS Federal Domestic Quarantine Order DA-2010-25, June 22, 2010. 6 p. [www.aphis.usda.gov/plant\\_health/plant\\_pest\\_info/eg\\_moth/downloads/spro/DA-2010-25.pdf](http://www.aphis.usda.gov/plant_health/plant_pest_info/eg_moth/downloads/spro/DA-2010-25.pdf).

Van Steenwyk RA, Varela LG, Cooper ML. 2011. Monitoring and control of European grapevine moth, *Lobesia botrana* (Lepidoptera: Tortricidae) in California vineyards. Abstracts of the IOBC-WPRS meeting in Lacanau, France. October 2-5, 2011.

Varela LG, Cooper ML, Smith RJ. 2013a. Can European grapevine moth, *Lobesia botrana* (Lepidoptera: Tortricidae) be eradicated from California? In: Calonne C, Duso C, Gessler C, et al. (eds.). IOBC/WPRS Bulletin 85:92-102.

Varela LG, Cooper ML, Smith RJ. 2013b. Trapping for European grapevine moth. CAPCA Adviser 16:20-5.

Zalom F, Grieshop J, Lelea M, et al. 2013. Community Perceptions of Emergency Responses to Invasive Species in California: Case Studies of the Light Brown Apple Moth and the European Grapevine Moth. Report submitted to USDA in fulfillment of Cooperative Agreement #10-8100-1531-CA.

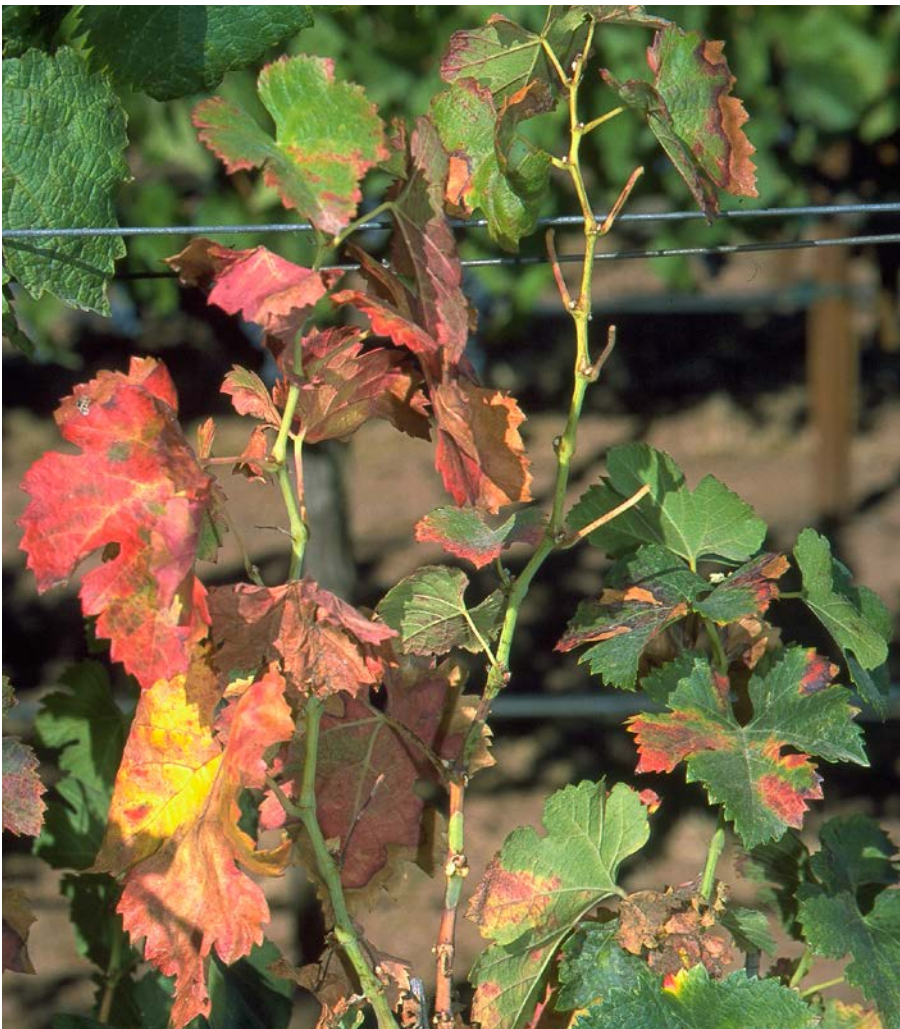


# Cooperative efforts contained spread of Pierce’s disease and found genetic resistance

by George Bruening, Bruce C. Kirkpatrick, Thomas Esser and Robert K. Webster

*An outbreak of Pierce’s disease of grapevine in the Temecula Valley in the late 1990s was one in a decades-long series of sporadic appearances of this infection in California. However, the new outbreak was qualitatively different because of the rapidity with which it spread in the vineyard and its appearance almost simultaneously at distant locations. The causative agent of Pierce’s disease is the bacterium *Xylella fastidiosa*, and the distinct characteristics of the Temecula Valley outbreak were traced to the establishment of a new insect vector in California, the glassy-winged sharpshooter. Intensive and collaborative efforts among government agencies, industry and research institutions over 15 years have successfully contained the disease, and given scientists time to discover promising long-term potential solutions through genetic resistance.*

Jack Kelly Clark



PD symptoms on a red grape variety, showing progressive foliage discoloration, irregular bark maturity and petioles without leaf blades. The Temecula Valley PD outbreak in 1999 triggered an emergency response to save the wine and grape industries.

Pierce’s disease (PD) was not new to California in 1999, when its characteristic scorching symptoms suddenly and severely devastated several hundred acres of grapevines in the Temecula Valley, affecting a quarter of the region’s vineyards. In the early 1880s, PD, then referred to as Anaheim disease, decimated 30,000 acres of vines near Anaheim and Pomona. Newton B. Pierce, a special agent of the U.S. Secretary of Agriculture, responded to this event as the first professional plant pathologist in California. He skillfully eliminated explanations other than disease but was not able to identify the causative agent. In the late 1880s, the disease damaged vineyards in Napa County, and there were severe outbreaks in Southern California in the 1930s and 1940s. Between 1994 and 2000, grapevines on over 1,000 acres in Northern California were affected, resulting in \$30 million in damage (Webster and Nation 2000).

The causative agent of PD was long thought to be a virus. The disease actually is caused by a strain of the bacterium *Xylella fastidiosa* (Davis et al. 1978). Strains of this bacterium also inflict damage on many other plant species, including almond, citrus, elm, maple, oak, oleander, stone fruit and sycamore. The 1999 outbreak in the Temecula Valley was unique, alarming and characterized by rapid and long-distance spread of PD in the vineyard, resulting from the establishment of a new vector in California, the glassy-winged sharpshooter (GWSS; *Homalodisca vitripennis*). Further spread of GWSS would place at risk California’s \$3 billion grape crop, \$60 billion in associated activity, and \$3 billion in fruit, nut and ornamental crops.

## Epidemiology of PD

*X. fastidiosa* is transmitted in the field by xylem-feeding insects, especially

Online: <http://californiaagriculture.ucanr.edu/landingpage.cfm?article=ca.v068n04p134&fulltext=yes>  
doi: 10.3733/ca.v068n04p134



sharpshooters (family Cicadellidae, subfamily Cicadellinae) and related insects such as spittlebugs (family Cercopidae) (Hopkins and Purcell 2002). Insect vectors acquire *X. fastidiosa* from infected plants; some plants are much more effective sources of the bacterium than others, depending on vector feeding preferences and patterns of *X. fastidiosa* accumulation and spread in the plant (Purcell and Saunders 1999). Ingested *X. fastidiosa* attaches to the sharpshooter's mouthparts, where it multiplies. Transmission to healthy plants occurs when the infected vector feeds and egests the bacterium into the plant's xylem. Plant symptoms, if any, appear weeks to months later. All nymphal stages of the sharpshooter can acquire *X. fastidiosa*, but nymphs lose the bacterium when they molt, because the mouth lining is shed during molting. If the sharpshooter acquires *X. fastidiosa* as an adult, it remains infective for the remainder of its life (Hopkins and Purcell 2002; Purcell and Finley 1979).

PD is most prevalent in grape-growing regions of the Southern U.S. states, Mexico and Central America, in areas that have mild winter temperatures. Research has shown that *Vitis vinifera* vines experimentally infected with *X. fastidiosa* will become pathogen-free if the vines are subjected to cold temperatures (Feil and Purcell 2001; Lieth et al. 2011). This cold curing phenomenon likely accounts for the lack of PD in viticultural regions such as New York, Oregon, Washington and higher-elevation vineyards in California, where vines experience cold winter temperatures. A recent model based on cold curing data by UC Davis professor Johan Lieth showed good predictive correlations between winter temperatures and the presence or absence of PD in viticultural regions in California (Lieth et al. 2011). Global warming is expected to alter crop geographical distributions (Weare 2009) and could reduce the areas currently subject to cold curing. Where winter temperatures have warmed in the Southeastern states, PD distribution has increased (Anas et al. 2008).

All cultivars of European grapevine, *V. vinifera*, are susceptible to *X. fastidiosa* infection. While most varieties eventually die from the infection, some varieties are more susceptible and succumb more

quickly than others (Hopkins and Purcell 2002). Muscadine varieties (*V. rotundifolia*) and certain *Vitis* hybrids, especially those that are endemic to North America, such as *V. labrusca* X *V. vinifera* (Concord grape), support only very low *X. fastidiosa* titers, which do not affect fruit quality or kill the vine.

The incidence and severity of PD in California vineyards have been cyclic since the first record of PD in Anaheim and Pomona in the early 1880s. A PD epidemic occurred from 1933 to 1940 in the Central Valley, where diseased vineyards were often located next to weedy alfalfa fields, which were breeding sites for green (*Draeculacephala minerva*) and red-headed (*Xyphon fulgida*) sharpshooters (Hopkins and Purcell 2002). The blue-green sharpshooter (BGSS; *Graphocephala atropunctata*), a very efficient vector of PD, was largely responsible for the severe outbreaks of PD that have occurred in the coastal regions of Northern and Southern California and in the interior areas of Napa and Sonoma counties (Redak et al. 2004; Webster and Nation 2000).

None of these sharpshooters is a strong flyer, so the PD infections they cause are typically limited to an area of a few hundred meters from their overwintering sites, which for the BGSS are weedy riparian areas and ornamental landscapes. Vineyards can be protected by treating them with insecticides in the spring in response to BGSS counts in yellow sticky traps (Varela et al. 2001). When the emerging shoots are short, BGSS-vectored *X. fastidiosa* can reach the permanent woody tissues of the vines and cause PD that persists and kills the vines. Inoculation of the green tips of longer canes later in the season tends to not result in systemic disease, since the infected portions of the vines are pruned off (Feil et al. 2003).

### The glassy-winged sharpshooter

GWSS probably entered Southern California on imported plant material in the late 1980s or early 1990s. While GWSS is not considered to be an



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**GWSS adults caught in a yellow sticky trap. In areas infested with GWSS, trap counts indicate when insecticides should be applied to citrus groves located near vineyards.**

economically important pest of grapevines in its own right (Andersen et al. 2003), its establishment greatly increased the incidence and severity of PD in the viticultural regions of Southern California and the lower San Joaquin Valley. To date, GWSS has not become permanently established north of Fresno County, although incipient infestations have occurred sporadically and have been successfully eradicated thus far (CDFA 2013).

The epidemiology of PD transmitted by GWSS differs considerably from the epidemiology of PD spread by the BGSS and other native sharpshooters. GWSS, surprisingly, is a less efficient vector of

**GWSS flies farther than other sharpshooters, has a polyphagous diet and can spread PD vine to vine. It initiates infections on a nearly year-round basis.**



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*X. fastidiosa* than native California sharpshooters (Daugherty and Almeida 2009). However, GWSS is a highly polyphagous insect, feeding on a variety of plants to fulfill its nutritional requirements (Varela et al. 2001). It also flies much greater distances than the native sharpshooters. The severe PD losses that occurred in the Temecula region in the late 1990s and early 2000s often occurred in vineyards located adjacent to citrus orchards, which are preferred feeding and breeding hosts of GWSS. PD transmitted by GWSS can rapidly encompass an entire vineyard, and significant PD losses occurred in vineyards located a mile or more from citrus orchards.

Unlike native PD vectors, GWSS can acquire and transmit *X. fastidiosa* from woody tissues such as 1- to 2-year-old lignified canes (Almeida and Purcell 2003; Almeida et al. 2005), and even spurs that remain when the dormant vines

are pruned. Thus, GWSS spreads *X. fastidiosa* on a nearly year-round basis and initiates chronic infections of vines. Extensive disease mapping done in the early stages of the Temecula epidemic showed that GWSS can spread *X. fastidiosa* vine to vine. Other PD vectors tend to introduce the bacterium from infected nongrape-

vine sources outside the vineyard rather than from chronically infected vines (Varela et al. 2001).

### Preventing spread of GWSS

After the magnitude of the threat from the Temecula PD epidemic was realized, the California Department of Food and Agriculture (CDFA) put in place a successful four-pronged program: containment, detection, rapid response and outreach.

**Containment.** Within the infected areas, the CDFA promulgated regulations aimed at interdicting likely pathways of GWSS spread from infested areas of Southern California. This involved regulating shipments of general nursery stock, bulk grapes and (later) bulk citrus. The



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During hot weather, PD causes leaves to turn brown, typically first at the leaf margins. New infestations are met with a rapid response and have been eradicated in Northern California counties.

regulations required inspection, treatment, monitoring and certification of shipments from infested areas. Enforcement of the regulations is coordinated by the CDFA and usually performed by the staff of county agricultural commissioners (CDFA 2001).

The certification program, established to ensure that shipments of nursery stock were free of GWSS, was effective but proved to be labor intensive, expensive and a significant burden to many growers (Redak and Bethke 2003). In response, concerted efforts were made to find a treatment that could be relied upon to minimize the risk of spreading GWSS on shipments of plant material from infested areas. While adult and nymphal GWSS were found to be fairly susceptible to insecticide treatments, the egg masses of the insect, embedded in the foliage of plants, were not (Grafton-Cardwell et al. 2003; Redak and Bethke 2003). Trials conducted by researchers at UC Riverside eventually identified two insecticides effective against all GWSS stages. Based on the trial results and beginning in 2008, the Nursery Stock Approved Treatment Protocol (Kabashima et al. 2011) became an optional substitute for the certification program.

In large grape production areas that were already infested with GWSS, such as portions of Kern, Riverside and Tulare counties, area-wide trapping and treatment programs for reducing GWSS numbers were implemented and evaluated. In these programs, coordinated insecticide treatments were applied primarily to citrus and, on occasion, to windbreaks, when trap counts revealed GWSS populations at or above damage thresholds (CDFA 2005). Similar programs were later

implemented in other infested viticultural areas. Removal of diseased vines helped reduce the incidence and sources of disease in these areas.

**Detection.** In addition to the GWSS containment activities, annual detection surveys in agricultural and urban areas were begun to search for new GWSS infestations and verify that uninfested areas remained free of GWSS. Surveys rely primarily on sticky yellow panel traps deployed in areas deemed at risk of GWSS introduction. Trapping is conducted every year during the warm-weather months, with some locations being monitored year-round.

**Rapid response.** Discovery of one or more GWSS in a new area triggers rapid response activity by the CDFA, which consists of intensive visual survey and deployment of more traps to determine if a new infestation exists and, if so, where its boundaries lie. New infestations are treated with insecticides applied primarily to preferred host plants of GWSS to eradicate the infestation (CDFA 2013).

**Outreach.** Outreach plays an important role in GWSS containment and suppression efforts. Outreach activities, including meetings, mailings and media pieces prepared by CDFA, UC, counties and outside contractors, are aimed at the affected industries and the general public to inform them of the new pest threat in California and the new programs and activities being conducted to address it. These efforts have helped gain compliance with new shipping requirements and enlisted assistance from the public in finding new infestations and preventing further spread of GWSS.

Success of the current four-pronged program is evidenced by two

See also: Jetter KM, Morse JG, Kabashima JN. 2014. The cost of the glassy-winged sharpshooter to California grape, citrus and nursery producers. *Calif Agr* 68(4): 161-167. <http://californiaagriculture.ucanr.edu/landingpage.cfm?article=ca.E.v068n04p161&fulltext=yes>



observations: The GWSS-infested area in California this year is only slightly larger than the area infested in 2002 (fig. 1), and 17 infestations found outside this area to date have been eradicated. The success of the GWSS control program shows it can be a model for responding to other pest invasions. However, the potential remains for GWSS populations to flare up, as evidenced by increased trap counts, and spread to new areas or resistance to current control methods could develop, requiring that program measures continue and prompting recent initiation of studies on resistance management.

While insecticide treatments have proven critical to keeping GWSS populations low and preventing the spread and establishment of GWSS in new areas of California, biological control is also used as part of an integrated management approach. Biological control efforts began in the late 1990s, when explorations for natural enemies were conducted in the native range of GWSS (Triapitsyn et al. 1998). The Temecula PD outbreak stimulated additional searches for biological control agents, and testing, rearing, release and monitoring of natural enemies are now

being conducted on an ongoing basis (CDFA 2001, 2013).

The natural enemies predominantly used against the sharpshooter are tiny mymarid wasps in the genus *Gonatocerus*, which lay their eggs inside the eggs of the sharpshooter. Approximately 2.4 million of these wasps have been released in California to date (CDFA 2013). These efforts have involved a collaborative response from CDFA (Larry Bezark, David Morgan, Charlie Pickett), UC Riverside (Ali Al-Wahibi, Mark Hoddle, Nick Irvin, Rodrigo Krugner, Joseph Morse, Leigh Pilkington, Serguei Triapitsyn) and USDA (Greg Simmons).

### Genetic approaches to combating PD

Genetic resistance is generally regarded as the most reliable control measure for a readily transmitted plant disease. Although there is apparent variation in the susceptibility of European grapevine (*V. vinifera*) varieties to *X. fastidiosa*, all succumb to PD (Hopkins and Purcell 2002). The research group of Andrew Walker at UC Davis discovered both apparently single-gene and multiple-gene resistance in other *Vitis* species

collected in the Southern U.S. states and northern Mexico (Walker and Riaz 2012). Progeny of interspecific (i.e., interspecies) crosses and backcrosses to *V. vinifera* were evaluated for a spectrum of horticultural and quality traits in addition to resistance to *X. fastidiosa* (Viana et al. 2011). Aggressive vine training, which forced flowering of 2-year-old seedlings, and application of DNA marker technology generated progeny of the fourth backcross, expected to be genetically 97% *V. vinifera*, in only 10 years. Test-size lots of wine have been made from grapes of previous generations and currently are being made from fourth backcross generation selections. Release of PD-resistant varieties with a range of wine styles is anticipated (Walker and Tenschler 2012). The breeding program continues.

Efforts in other laboratories have transferred specific genes from nongrapevine sources, referred to as transgenes, into *V. vinifera*. The advantage of the transgenic approach is that it greatly increases the number of sources of resistance beyond what is available in conventional breeding. Ten years ago, only a few sketchy notions were available on how to create

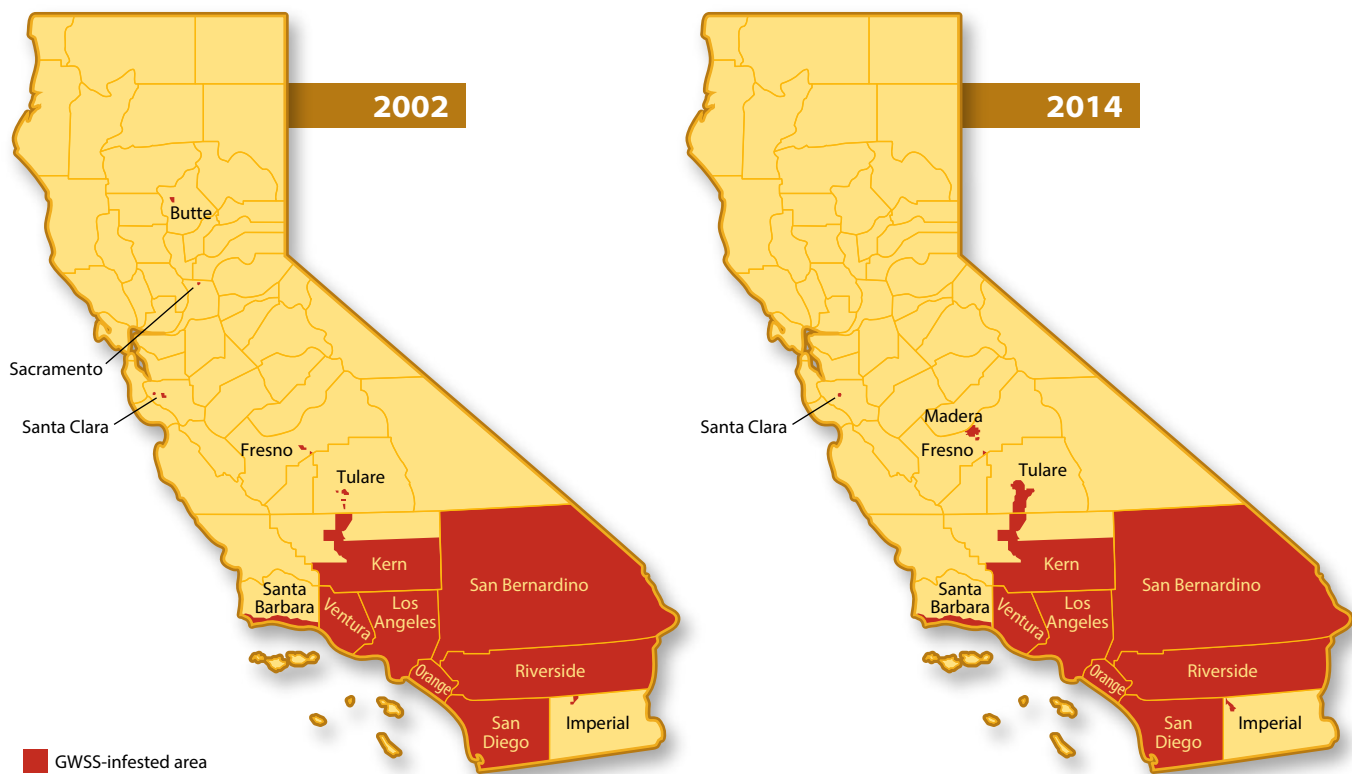


Fig. 1. Areas of California infested by GWSS in 2002 and 2014. At the end of 2002, GWSS was confined mostly to Southern California counties, with minor infestations in three Northern California counties. The Southern California infestation in 2014 is slightly expanded compared to the infestation in 2002, but some Northern California infestations have been eradicated. Other infestation maps are available at [cdfa.ca.gov/pdcp/map\\_index.html](http://cdfa.ca.gov/pdcp/map_index.html).

transgenes capable of interfering with *X. fastidiosa* infection of grapevine or the mechanisms on which they might operate. Infecting *X. fastidiosa* bacterial cells reside not in the living cells of the plant, where a transgene product normally would accumulate, but in the xylem, the array of dead cells that form the vascular elements responsible for conducting water and minerals from the roots into the aerial parts of the plant. Responding to this challenge, researchers not only identified various genetic elements that encode molecules that restrain the accumulation and/or spread of *X. fastidiosa* but also elements that deliver these molecules to the xylem. Importantly, several of the molecules were shown to cross graft unions and to confer, as a transgenic rootstock, protection to a grafted nontransgenic scion. Rootstock-conferred resistance has obvious versatility: Varietal scions now in

use could be grafted onto the transgenic rootstock line and be at least partially protected against *X. fastidiosa*.

The transformed lines that have been examined closely are indistinguishable from the untransformed lines from which they were derived (Dandekar et al. 2011; Lindow et al. 2014), suggesting that the vines have the quality and horticultural traits of the untransformed lines. This result is expected because the transformation process introduces only a minute change in the plant genome. In commercialization, combinations of several transgenes acting through distinct biochemical mechanisms of action likely would be introduced into a grapevine line to obtain at least additive resistance effects and to greatly reduce the chance that a variant *X. fastidiosa* would be able to overcome resistance. The following paragraphs summarize five mechanistically diverse and

clever experimental approaches for developing transgenic resistance to *X. fastidiosa* in grapevines.

**Fusion of two proteins.** One straightforward approach to reducing the titer of *X. fastidiosa* in grapevine xylem is to discover or develop a protein capable of killing or preventing the increase of *X. fastidiosa* cells and then to deliver that protein to the xylem. A research group headed by Abhaya Dandekar at UC Davis and including Goutam Gupta of Los Alamos National Laboratory conceived the approach of fusing two proteins, each with some activity against *X. fastidiosa*, into a single, xylem-targeted protein. One protein of the fusion pair binds to a specific protein that is abundant on the surface of the *X. fastidiosa* cell. The other protein breaches the *X. fastidiosa* cell membrane, thereby killing the bacterial cell. Both proteins killed *X. fastidiosa* cells

## Coordinated response to PD involves growers, scientists and government

The academic and government research on GWSS and PD received support from the state's viticulture and enology industries and even local government before and during the Temecula Valley outbreak. Industry organizations partnering with CDFA and UC Division of Agriculture and Natural Resources (UC ANR) in these efforts included the American Vineyard Foundation, the California Table Grape Commission, the California Raisin Board and the California Rootstock Commission. UC ANR and CDFA provided coordination to prioritize research, avoid duplication and maximize the collective benefits of the programs. Since 1999, USDA has provided about 75% of the funding for PD control and research, and the state of California, industry and UC have provided the rest. California legislation initiated a statewide assessment on wine grapes to support research and related activities, raising about \$46 million to date (Tumber et al. 2014; Wiggins 2001). The timeline here highlights some of the key events in the efforts to contain and find solutions to the PD outbreak in California.

### 1996

UC ANR forms Viticulture Consortium (VC), funded by a grant from USDA Cooperative State Research, Education, and Extension Service (USDA CSREES, now USDA NIFA, National Institute of Food and Agriculture).

Report of GWSS establishment in Southern California is published.

### 1997

Industry-matched state funds initiate the California Competitive Grant Program for Research in Viticulture and Enology (CCGPRVE).

### 1998

Search for biological control agents for GWSS begins.

### 1999

Temecula Valley vineyards show severe damage from PD; GWSS is recognized as the vector.

Riverside County declares emergency; the county and city of Temecula each contribute \$125,000 for research.

UC appoints Pierce's Disease Research and Emergency Response Task Force to identify strategies for combating PD.

CDFA appoints task force with members from UC, California State University, CDFA, county agricultural commissioners, USDA and industry to identify PD/GWSS research priorities.

CA Assembly Bill 1232 provides \$2.25 million over 3 years to be matched by \$0.75 million in industry funds for PD/GWSS research; the CDFA competitive grants program is funded.



in culture, but the fusion protein was significantly more potent than either protein component alone. Grapevines expressing the fusion protein showed significant protection against PD (Dandekar, Gouran et al. 2012).

**HxfA protein.** Bruce Kirkpatrick's laboratory at UC Davis identified a protein of *X. fastidiosa*, HxfA, which, when inactivated, surprisingly led to increased virulence of the bacterium (Guilhabert and Kirkpatrick 2005). This result suggests that, when active, HxfA suppresses virulence. Perhaps a grapevine line that produces HxfA could reduce PD severity by decreasing the virulence of infecting *X. fastidiosa*, and infecting *X. fastidiosa* would be less successful at inducing PD than in unaltered vines. Grapevine plants producing full-length or truncated versions of HxfA were constructed. The genes were designed to cause the HxfA

protein to be secreted into the xylem, where infecting *X. fastidiosa* cells colonize the vine. Greenhouse-grown plants were found to have reduced disease symptoms compared to unaltered grapevines when inoculated with a high dose of *X. fastidiosa* (Kirkpatrick et al. 2012).

**XfDSF, pathogen confusion.** Steven Lindow and his research associates at UC Berkeley intensely investigated intercellular communication of *X. fastidiosa* and applied their findings to suppress the spread of the bacterium in the plant using a phenomenon they refer to as pathogen confusion. *X. fastidiosa* secretes a specific 14-carbon fatty acid and possibly other similar molecules as diffusible signal factors (XfDSF). As *X. fastidiosa* populations grow, the concentration of XfDSF around the cells increases. Compared to the unaltered bacterium, *X. fastidiosa* mutants that fail to produce XfDSF spread more readily

in the plant, whereas spread of mutants that overproduce XfDSF is severely restricted (Chatterjee et al. 2008).

Transfer of a gene whose product catalyzes XfDSF synthesis should result in accumulation of XfDSF even in the absence of *X. fastidiosa* and, after inoculation with *X. fastidiosa*, should constrain movement and accumulation of *X. fastidiosa* and reduce PD symptom extent and intensity. These expected results have been observed (Lindow et al. 2012, 2014).

**Programmed cell death.** The characteristic scorching symptoms of PD were shown by the laboratory of David Gilchrist at UC Davis to be an example of the phenomenon known as genetically programmed cell death, where the symptoms resulted from a reaction of the plant to the presence of a bacterium (*X. fastidiosa*) rather than directly to an action of that bacterium (Gilchrist et al. 2007). This

## 2000

UC is awarded USDA CSREES grant of \$2 million to establish a competitive grants program for PD/GWSS research; annual funding was renewed eight times.

UC establishes Pierce's Disease Competitive Grants Program.

CA Senate Bill 671 provides funds for a coordinated statewide effort against PD/GWSS.

Federal government declares emergency and provides \$22.3 million for GWSS containment and PD research.

First GWSS infestation is found outside the generally infested Southern California area, in Contra Costa County; eradication efforts begin.

CDFA adopts emergency regulations to prevent spread of GWSS on shipments of nursery stock, bulk grapes and (later) citrus.

## 2001

CA Assembly Bill 1394 creates the Pierce's Disease and Glassy-Winged Sharpshooter Board and an assessment on wine grapes to fund PD/GWSS research and related activities; this assessment raised \$46 million through February 2014.

First area-wide management program begins in Kern County, testing methods for controlling PD and GWSS in large and diverse agricultural areas.

First annual PD/GWSS research symposium takes place.

## 2002

GWSS infestation found in Contra Costa County in 2000 is declared eradicated, marking the first successful eradication of a localized GWSS infestation.

## 2004

CDFA and UC competitive PD/GWSS research programs combine their proposal review processes.

CDFA-requested report "California Agricultural Research Priorities: Pierce's Disease" is published by National Research Council of the National Academies.

Number of GWSS biological control agents released in California since 1999 exceeds 1 million.

## 2005

California wine grape growers vote to continue statewide PD/GWSS wine grape assessment.

## 2008

Nursery Stock Approved Treatment Protocol established to reduce need for GWSS inspections.

## 2010


California wine grape growers vote again to continue statewide PD/GWSS wine grape assessment, with option to support research and outreach on other wine grape pests and diseases.

## 2011

Last CCGPRVE-supported project completed, and the program ends.

## 2013

Last projects funded by USDA-UC and VC are completed; these programs end.

Industry-funded CDFA competitive grants program continues, with its proposal review process now conducted under UC ANR's Unified Grant Management for Viticulture and Enology. 



UC Davis plant scientist Abhaya Dandekar and colleagues have fused two genes to engineer resistance to Pierce's disease of grapevines.

research group searched a large collection of grapevine genes for the ability of specific nucleotide sequences to confer protection against programmed cell death in tomato roots. Two of these grapevine nucleotide sequences, when transferred back into grapevine, suppressed PD symptoms and prevented death of the *X. fastidiosa*-inoculated plants (Harvey et al. 2008). Moreover, the bacterial titer was reduced by 4 to 6 orders of magnitude relative to the titer in untransformed control vines, which died in 2 to 3 months. In the transformed plants, *X. fastidiosa* titers were reduced to a level that should result in only rare or no acquisition by the sharpshooter. One of the nucleotide sequences, when introduced into the rootstock only, was able to protect untransformed scions (Gilchrist and Lincoln 2009).

**PG-inhibiting protein (PGIP).** The *X. fastidiosa* genome has one gene for the enzyme polygalacturonase (PG), an enzyme that cleaves homogalacturonan, a major component of the intercellular matrix that joins the walls of adjacent plant cells. A *X. fastidiosa* strain with an inactive PG was unable to move long distance in the inoculated plant or to induce PD symptoms (Roper et al. 2007). UC Davis researchers Abhaya Dandekar, John Labavitch and Ann Powell lead research groups that have demonstrated interference with *X. fastidiosa* accumulation in plants by the action of a PG-inhibiting protein (PGIP). The PGIP from pear fruit was found to be effective in inhibiting bacterial PG. The pear PGIP gene was moved into grapevine, where it provided

scion was demonstrated to accumulate PGIP from its PGIP-transgenic rootstock, suggesting that transported PGIP is the actual agent of protection in the scion (Dandekar, Gilchrist et al. 2012; Labavitch et al. 2012).

Tests of transformed grapevine lines were performed first in the greenhouse, and outdoor trials of these newly developed grapevine materials were begun in March 2010. The field trials are expected to yield valuable information on the feasibility of developing these approaches into commercial applications (Gilchrist et al. 2012; Miller et al. 2012).

### Research advances

The seriousness of the GWSS-borne *X. fastidiosa* threat to the California wine and grape industries was realized early in the Temecula Valley PD outbreak, and effective cooperation between federal, state and county agricultural agencies, industry groups, and UC followed. Actions taken to combat the Temecula Valley PD epidemic and prevent its spread were rapidly mounted, and funding from federal, state and industry sources has supported both control measures and basic and applied research. A summary of current research and control costs associated with PD appeared recently (Tumber et al. 2014), and the sidebar below indicates some of the research expenditures.

Of the biological actors in the epidemic (*X. fastidiosa*, GWSS and grapevine), none was regarded in 1999 as being well characterized in research terms, and all seemed at the time to be almost

## Varietal scions now in use could be grafted onto the transgenic rootstock line and be at least partially protected against *X. fastidiosa*.

protection against *X. fastidiosa*. Protection and level of PGIP accumulation were positively correlated in different transformed grapevine lines (Aguero et al. 2005). PGIP is naturally targeted to the xylem, and PGIP-generating rootstocks protected grafted nontransgenic scions. The untransformed

intractable in molecular terms. Over the years, research projects were supported at 16 universities and at several government laboratories, and the continued funding from various sources, including grower-voted assessments (see sidebar, pages 138–9), resulted in numerous advances in our understanding of the biology of the disease and the development of effective control measures.

The entire genome sequence of *X. fastidiosa* became available, and procedures for creating mutants of the bacterium and introducing new genes were developed and applied (Guilhabert et al. 2001; Guilhabert and Kirkpatrick 2003; Newman et al. 2004; Van Sluys et al. 2003). Genetic variation in *X. fastidiosa* was characterized as to its origins and its determination of host range (Nunney 2011; Nunney et al. 2013). Individual events in the association of *X. fastidiosa* with the sharpshooter and the establishment of feeding sites by the sharpshooter have been revealed (Almeida and Purcell 2003; Almeida et al. 2005; Son et al. 2012). The arrays of grapevine messenger RNAs (responsible for directing the synthesis of proteins) that accumulate under various situations were characterized and revealed grapevine genes that specifically respond to *X. fastidiosa* infection but not to drought (Choi et al. 2010, 2013). In cold curing, *X. fastidiosa* was found to be cleared not by the effects of low temperatures alone but by the grapevine's response to the cold (Feil and Purcell 2001; Lieth et al. 2011). Obviously, these research advances would not have been possible without the sources of research support indicated above. The new discoveries have elevated the bacterium, the insect vector and the plant host close to the level of a model research system and created a foundation for new management strategies in the future. [CA](#)

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## References

- Agüero CB, Uratsu SL, Greve C, et al. 2005. Evaluation of tolerance to Pierce's disease and *Botrytis* in transgenic plants of *Vitis vinifera* L. expressing the pear PGIP gene. *Mol Plant Pathol* 6(1):43–51. doi:10.1111/j.1364-3703.2004.00262.x.
- Almeida RPP, Purcell AH. 2003. Transmission of *Xylella fastidiosa* to grapevines by *Homalodisca coagulata* (Hemiptera:Cicadellidae). *J Econ Entomol* 96(2):264–71. doi:10.1603/0022-0493-96.2.264.
- Almeida RPP, Wistrom C, Hill BL, et al. 2005. Vector transmission of *Xylella fastidiosa* to dormant grape. *Plant Dis* 89(4):419–24. doi:10.1094/pd-89-0419.
- Anas O, Harrison UJ, Brannen PM, et al. 2008. The effect of warming winter temperature on the severity of Pierce's disease in the Appalachian mountains and Piedmont of the southeastern United States. *Plant Health Progress*. www.plantmanagementnetwork.org/pub/php/research/2008/pierces. doi:10.1094/PHP-2008-0718-01-RS.
- Andersen PC, Brodbeck BV, Mizell RF. 2003. Plant and insect characteristics in response to increasing density of *Homalodisca coagulata* on three host species: A quantification of assimilate extraction. *Entomologia Experimentalis Et Applicata* 107(1):57–68. doi:10.1046/j.1570-7458.2003.00036.x.
- [CDFA] California Department of Food and Agriculture. 2001. Pierce's Disease Control Program, Report to the Legislature, January 1, 2001 (2000 Report). www.cdfa.ca.gov/pdcp/ReportsToLegislature.html. 33 p plus appendices.
- CDFa. 2005. Pierce's Disease Control Program, Report to the Legislature, April 2005 (2004 Report). www.cdfa.ca.gov/pdcp/ReportsToLegislature.html. 28 p plus appendices.
- CDFa. 2013. Pierce's Disease Control Program, Report to the Legislature (2012 Report). www.cdfa.ca.gov/pdcp/ReportsToLegislature.html, www.cdfa.ca.gov/pdcp/map\_index.html. 24 p.
- Chatterjee S, Wistrom C, Lindow SE. 2008. A cell-cell signaling sensor is required for virulence and insect transmission of *Xylella fastidiosa*. *P Natl Acad Sci USA* 105:2670–5.
- Choi HK, da Silva FG, Lim HJ, et al. 2010. Diagnosis of Pierce's disease using biomarkers specific to *Xylella fastidiosa* rRNA and *Vitis vinifera* gene expression. *Phytopathology* 100(10):1089–99. doi:10.1094/phyto-01-10-0014.
- Choi HK, landolino A, da Silva FG, et al. 2013. Water deficit modulates the response of *Vitis vinifera* to the Pierce's disease pathogen *Xylella fastidiosa*. *Mol Plant Microbe In* 26(6):643–57. doi:10.1094/mpmi-09-12-0217-r.
- Dandekar AM, Gilchrist D, Miller T, et al. 2012. Chimeric antimicrobial protein and polygalacturonase-inhibiting protein transgenic grapevine field trial. In: Esser T (ed.). *Pierce's Disease Research Progress Reports*. Sacramento: California Department of Food and Agriculture. p 94–103. www.cdfa.ca.gov/pdcp/research.html.
- Dandekar AM, Gilchrist D, Miller T, et al. 2011. Chimeric antimicrobial protein and polygalacturonase-inhibiting protein transgenic grapevines field trial. In: Esser T (ed.). *Pierce's Disease Research Progress Reports*. Sacramento: California Department of Food and Agriculture. p 101–6. www.cdfa.ca.gov/pdcp/research.html.
- Dandekar AM, Gouran H, Ibanez AM, et al. 2012. An engineered innate immune defense protects grapevines from Pierce disease. *P Natl Acad Sci USA* 109(10):3721–5. doi:10.1073/pnas.1116027109.
- Daugherty MP, Almeida RPP. 2009. Estimating *Xylella fastidiosa* transmission parameters: Decoupling sharpshooter number and feeding period. *Entomologia Experimentalis Et Applicata* 132(1):84–92. doi:10.1111/j.1570-7458.2009.00868.x.
- Davis MJ, Purcell AH, Thomson SV. 1978. Pierce's disease of grapevines — isolation of causal bacterium. *Science* 199(4324):75–7. doi:10.1126/science.199.4324.75.
- Feil H, Feil WS, Purcell AH. 2003. Effects of date of inoculation on the within-plant movement of *Xylella fastidiosa* and persistence of Pierce's disease within field grapevines. *Phytopathology* 93(2):244–51. doi:10.1094/phyto.2003.93.2.244.
- Feil H, Purcell AH. 2001. Temperature-dependent growth and survival of *Xylella fastidiosa* in vitro and in potted grapevines. *Plant Dis* 85(12):1230–4. doi:10.1094/pdis.2001.85.12.1230.
- Gilchrist DG, Lincoln JE. 2009. Systemic control of Pierce's disease by altered expression of anti-apoptotic genes or their RNA-based regulatory elements. In: Esser T (ed.). *Pierce's Disease Research Symposium*. CDFa, Sacramento. p 10–11. www.cdfa.ca.gov/pdcp/research.html.
- Gilchrist DG, Lincoln JE, Cook DR. 2007. Resistance to Pierce's disease by transgenic expression of plant-derived anti-apoptotic genes. In: Esser T (ed.). *Pierce's Disease Research Symposium*. CDFa, Sacramento, CA. p 252–5.
- Gilchrist D, Lincoln J, Kominek T, et al. 2012. Field evaluation of grape plants expressing potential protective DNA sequences effective against Pierce's disease. In: Esser T (ed.). *Pierce's Disease Research Progress Reports*. CDFa, Sacramento. p 109–11. www.cdfa.ca.gov/pdcp/research.html.
- Grafton-Cardwell EE, Reagan CA, Ouyang Y. 2003. Insecticide treatments disinfest nursery citrus of glassy-winged sharpshooter. *Calif Agri* 57(4):128–31.
- Guilhabert MR, Hoffman LM, Mills DA, et al. 2001. Transposon mutagenesis of *Xylella fastidiosa* by electroporation of Tn5 synaptic complexes. *Mol Plant Microbe In* 14(6):701–6.
- Guilhabert MR, Kirkpatrick BC. 2003. Transformation of *Xylella fastidiosa* with broad host range RSF1010 derivative plasmids. *Mol Plant Pathol* 4(4):279–85. doi:10.1046/j.1364-3703.2003.00175.x.
- Guilhabert MR, Kirkpatrick BC. 2005. Identification of *Xylella fastidiosa* antivirulence genes: Hemagglutinin adhesins contribute to *X. fastidiosa* biofilm maturation and colonization and attenuate virulence. *Mol Plant Microbe In* 18(8):856–68.
- Harvey JJW, Lincoln JE, Gilchrist DG. 2008. Programmed cell death suppression in transformed plant tissue by tomato cDNAs identified from an *Agrobacterium rhizogenes*-based functional screen. *Mol Genet Genomics* 279(5):509–21. doi:10.1007/s00438-008-0329-2.
- Hopkins DL, Purcell AH. 2002. *Xylella fastidiosa*: Cause of Pierce's disease of grapevine and other emergent diseases. *Plant Dis* 86(10):1056–66. doi:10.1094/pdis.2002.86.10.1056.
- Kabashima J, Gutman G, Redak R, et al. 2011. GWSS Nursery Approved Treatment Best Management Practices. CDFa. 23 p.
- Kirkpatrick BC, Lincoln JE, Roper C, et al. 2012. Evaluation of Pierce's disease resistance in transgenic *Vitis vinifera* grapevines expressing either grape thaumatin-like protein or *Xylella fastidiosa* hemagglutinin protein. In: Esser T (ed.). *Pierce's Disease Research Symposium*. CDFa, Sacramento. p 130–6.
- Labavitch JM, Powell ALT, Bennett A, et al. 2012. Optimizing grape rootstock production and export of inhibitors of *Xylella fastidiosa* polygalacturonase activity. In: Esser T (ed.). *Pierce's Disease Research Symposium*. CDFa, Sacramento. p 147–53.
- Lieth JH, Meyer MM, Yeo K-H, et al. 2011. Modeling cold curing of Pierce's disease in *Vitis vinifera* 'Pinot Noir' and 'Cabernet Sauvignon' grapevines in California. *Phytopathology* 101:1492–500.
- Lindow SE, Ionescu M, Baccari C, et al. 2012. Enhancing control of Pierce's disease by augmenting pathogen signal molecules. In: Esser T (ed.). *Pierce's Disease Research Progress Reports*. CDFa, Sacramento. p 154–66. www.cdfa.ca.gov/pdcp/research.html.
- Lindow S, Newman K, Chatterjee S, et al. 2014. Production of *Xylella fastidiosa* diffusible signal factor in transgenic grape causes pathogen confusion and reduction in severity of Pierce's disease. *Mol Plant Microbe In* 27(3):244–54. doi:10.1094/mpmi-07-13-0197-fi.
- Miller T, Daugherty M, Mauk P. 2012. Field trial for resistance to Pierce's disease. In: Esser T (ed.). *Pierce's Disease Research Progress Reports*. CDFa, Sacramento. p 175–7. www.cdfa.ca.gov/pdcp/research.html.
- Newman KL, Almeida RPP, Purcell AH, et al. 2004. Cell-cell signaling controls *Xylella fastidiosa* interactions with both insects and plants. *P Natl Acad Sci USA* 101(6):1737–42.
- Nunney L. 2011. Homologous recombination and the invasion of a new plant host by the pathogenic bacterium, *Xylella fastidiosa*. *Phytopathology* 101(6):S130.
- Nunney L, Vickerman DB, Bromley RE, et al. 2013. Recent evolutionary radiation and host plant specialization in the *Xylella fastidiosa* subspecies native to the United States. *Appl Environ Microb* 79(7):2189–200. doi:10.1128/aem.03208-12.
- Purcell A, Finley AH. 1979. Evidence for noncirculative transmission of Pierce's disease bacterium by sharpshooter leafhoppers. *Phytopathology* 69:393–5.
- Purcell AH, Saunders SR. 1999. Fate of Pierce's disease strains of *Xylella fastidiosa* in common riparian plants in California. *Plant Dis* 83(9):825–30.
- Redak R, Bethke J. 2003. Toward a standardized treatment protocol to eliminate glassy-winged sharpshooter egg masses in commercial nursery stock. In: Esser T (ed.). *Pierce's Disease Research Symposium*. CDFa, Sacramento. p 308–10.
- Redak RA, Purcell AH, Lopes JRS, et al. 2004. The biology of xylem fluid-feeding insect vectors of *Xylella fastidiosa* and their relation to disease epidemiology. *Annu Rev Entomol* 49:243–70. doi:10.1146/annurev.ento.49.061802.123403.
- Roper MC, Greve LC, Warren JG, et al. 2007. *Xylella fastidiosa* requires polygalacturonase for colonization and pathogenicity in *Vitis vinifera* grapevines. *Mol Plant Microbe In* 20(4):411–9. doi:10.1094/mpmi-20-4-0411.
- Son Y, Backus EA, Groves RL, et al. 2012. Pattern of stylet penetration activity by *Homalodisca vitripennis* (Hemiptera: Cicadellidae) adults in relation to environmental temperature and light conditions. *Environ Entomol* 41(5):1215–30. doi:10.1603/en11319.
- Triapitsyn SV, Mizell RF, Bossart JL, et al. 1998. Egg parasitoids of *Homalodisca coagulata* (Homoptera: Cicadellidae). *Fla Entomol* 81:241–3.
- Tumber KP, Alston JM, Fuller KB. 2014. Pierce's disease costs California \$104 million per year. *Calif Agr* 68(1–2):20–9.
- Van Sluys MA, de Oliveira MC, Monteiro-Vitorello CB, et al. 2003. Comparative analyses of the complete genome sequences of Pierce's disease and citrus variegated chlorosis strains of *Xylella fastidiosa*. *J Bacteriol* 185(3):1018–26. doi:10.1128/jb.185.3.1018-1026.2003.
- Varela LG, Smith RJ, Phillips PA, et al. 2001. Pierce's Disease. *UC ANR Pub* 21600. Oakland, CA. p 20.
- Viana AP, Riaz S, Walker MA. 2011. Evaluating genetic diversity and optimizing parental selections in a segregating table-grape population. *Am J Enol Viticult* 62(3):285–90. doi:10.5344/ajev.2011.10073.
- Walker A, Riaz S. 2012. Genetic mapping of *Xylella fastidiosa* resistance gene(s) in grape germplasm from southern United States. In: Esser T (ed.). *Pierce's Disease Research Symposium*. CDFa, Sacramento. p 241–7.
- Walker A, Tenschler A. 2012. Breeding to Pierce's disease resistant winegrapes. In: Esser T (ed.). *Pierce's Disease Research Symposium*. CDFa, Sacramento. p 233–40.
- Weare BC. 2009. How will changes in global climate influence California? *Calif Agri* 63(2):59–66.
- Webster RK, Nation SP. 2000. Report of the University of California Pierce's Disease Research and Emergency Response Task Force. *UC Office of the President*. 62 p.
- Wiggins P. 2001. Food and Agriculture Code, Amend. California State Assembly Bill 1394. *Pests: Pierce's disease and the Glassy-winged Sharpshooter*. [http://www.leginfo.ca.gov/pub/01-02/bill/asm/ab\\_1351-1400/ab\\_1394\\_bill\\_20010725\\_chaptered.html](http://www.leginfo.ca.gov/pub/01-02/bill/asm/ab_1351-1400/ab_1394_bill_20010725_chaptered.html).

Once pests and diseases become established, their interactions with crops, landscapes or animals are in a continuous state of flux, depending on environmental conditions and changes in pest control practices. Their long-term management is never static; it relies on a combination of techniques and strategies. The articles in this section take the long view and present how UC scientists tackle the evolution of a pest problem — herbicide resistance — and how the UC Statewide IPM program has managed pests while minimizing environmental risks for 35 years.

# Herbicide-resistant weeds challenge some signature cropping systems

by Bradley D. Hanson, Steven Wright, Lynn M. Sosnoskie, Albert J. Fischer, Marie Jasieniuk, John A. Roncoroni, Kurt J. Hembree, Steve Orloff, Anil Shrestha and Kassim Al-Khatib

*Invasive and endemic weeds pose recurring challenges for California land managers. The evolution of herbicide resistance in several species has imposed new challenges in some cropping systems, and these issues are being addressed by UC Cooperative Extension farm advisors, specialists and faculty. There are currently 24 unique herbicide-resistant weed biotypes in the state, dominated by grasses and sedges in flooded rice systems and, more recently, glyphosate-resistant broadleaf and grass weeds in tree and vine systems, roadsides and glyphosate-tolerant field crops. Weed scientists address these complex issues using approaches ranging from basic physiology and genetics research to applied research and extension efforts in grower fields throughout the state. Although solutions to herbicide resistance are not simple and are affected by many biological, economic, regulatory and social factors, California stakeholders need information, training and solutions to address new weed management problems as they arise. Coordinated efforts conducted under the Endemic and Invasive Pests and Disease Strategic Initiative directly address weed management challenges in California's agricultural industries.*

Endemic and invasive weeds are important management concerns in California due to their direct and indirect costs to agriculture, the environment and society. Pimentel et al. (2005) estimated that weeds cost U.S. crop producers and pasture managers over \$30 billion in control-related expenses and reduced productivity. Although specific data are not available for California's portion of these losses, weed management costs for the state's 40 million acres of crop and grazing lands, as well as the remaining 60 million acres of land area, amount, undoubtedly, to several billion dollars annually. In addition to the direct cost of weed control and lost agricultural productivity, weeds also affect ecosystem quality and function, reduce recreational access and degrade aesthetics in natural areas, change wildland fire regimes and severity, and impede water flow through rivers and canals, among other negative impacts.

Although crop weeds are seldom considered as being "invasive" in the traditional sense, novel biotypes can develop, spread and subsequently occupy a greater proportion of crop acreage than might normally be expected. For example, when a weed population evolves resistance to an herbicide or any other control measure, a "routine" pest can become a new and serious problem. The first case of an herbicide-resistant weed in California was reported in 1981 by UC scientists (Holt et al. 1981); in recent years, additional species have evolved resistance to various herbicide chemistries (table



A stone fruit orchard in Fresno County is dominated by glyphosate-resistant horseweed. Reliance on one method of weed control imposes selection pressure, which can lead to population shifts to tolerant species or selection of resistant biotypes.

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doi: 10.3733/ca.v068n04p142





1) used in some of California's signature cropping systems, including flooded rice, orchards and vineyards as well as nearby noncrop areas.

### How do weeds become resistant to herbicides?

Environmental factors and production practices influence species composition at any location, a phenomenon known as selection pressure. Under constant conditions, the weed community will become dominated by species that thrive under those conditions. If this steady state is upset by a change in management practices, a weed shift may occur, resulting

in a community dominated by different species adapted to the new conditions (Hanson et al. 2013). This weed shift can be caused by agronomic and horticultural practices (tillage, fertility, irrigation, etc.) or by the use of herbicides, which are very strong selective agents. Some species will be less susceptible (more tolerant) than others to any management practice, and repeated use of the same control strategy can shift weed populations to become dominated by naturally tolerant species (fig. 1A).

Herbicide resistance, on the other hand, implies that a genetic change has caused a formerly susceptible population

of a species to become resistant to an herbicide. Herbicide resistance arises from the process of adaptive evolution, whereby mutations change the physiology of plants in such a way that the herbicide is less effective. Under the continued selection pressure exerted by the herbicide(s), resistant plants with the new genotype are not controlled, and their offspring build up in the population (fig. 1B). Depending on the initial frequency and genetic basis of resistance, the regularity and rate of herbicide applications, and the reproductive system of the weed, it may take from a few to many generations for resistance to become

TABLE 1. Important herbicide modes of action

Mode of action	WSSA group	Target site and effects	Herbicide examples
ACCase inhibitors	1	Several important classes include aryloxyphenoxypropionates, cyclohexanediones and phenylpyrazolin. These herbicides inhibit the enzyme acetyl coenzyme A carboxylase (ACCase), which leads to the disruption of lipid synthesis at the growing point of susceptible grasses.	Clethodim, cyhalofop, diclofop, fluzafop, pinoxaden, sethoxydim, many others
ALS inhibitors	2	Several herbicide classes including the imidazolinones and sulfonylureas and others inhibit the enzyme acetolactate synthase (ALS), which disrupts synthesis of branched-chain amino acids.	Bensulfuron, chlorsulfuron, halosulfuron, imazamox, imazethapyr, metsulfuron, rimsulfuron, sulfometuron, many others
Carotenoid synthesis inhibitors	11, 12, 13, 27	Several unrelated chemical classes block enzymes important in the synthesis of carotenoids and/or chlorophyll. Because carotenoids protect plants from excess oxidative energy, lack of carotenoids usually results in membrane and protein damage from free radicals.	Amitrole, clomazone, fluridone, mesotrione, norflurazon, topramezone, others
Cellulose inhibitors	20, 21, 27	Several chemical classes inhibit aspects of cell wall (cellulose) synthesis.	Dichlobenil, indaziflam, isoxaben, quinclorac
EPSPS inhibitors	9	The glycine herbicides inhibit the enzyme 5-enolpyruvylshikimate-3-phosphate synthetase (EPSPS), which is important in the synthesis of aromatic amino acids.	Glyphosate
Fatty acid and lipid synthesis inhibitors	8, 16, 26	Several chemical classes, including the thiocarbamates, inhibit processes important in the synthesis of fatty acid and lipids, impacting production of membranes, proteins, hormones and other cellular components.	Bensulide, butylate, EPTC, molinate, triallate, vernolate, others
Glutamine synthetase inhibitors	10	Phosphonic acid herbicides inhibit the enzyme glutamine synthetase. Blocking of this process leads to buildup of ammonia in the plant and also inhibits PSII and PSI.	Glufosinate
Mitosis inhibitors	3, 15, 23	Several different chemical families affect various processes important in cell division. The most widely used include chloroacetemides (Group 3) and dinitroaniline (Group 15) herbicides.	Alachlor, dimethenamid, metolachlor, oryzalin, pendimethalin, pronamide, trifluralin, many others
Photosystem I inhibitors (PSI)	22	PSI inhibitors divert electrons during photosynthesis and create free radicals that quickly degrade cell membranes and lead to cell and tissue desiccation.	Paraquat, diquat
Photosystem II inhibitors (PSII)	5, 6, 7	Herbicide classes including the triazines, uracils, amides and several others disrupt photosynthesis by blocking electron transport in PSII. Plant death usually occurs from protein and lipid oxidation caused by free radicals.	Atrazine, bromacil, diuron, hexazinone, linuron, propanil, simazine, tebuthiuron, others
Synthetic auxins	4	Benzoic acids, phenoxycarboxylic acids, pyrachlor and pyridine carboxylic acids mimic endogenous auxins. At high concentrations, these growth regulator herbicides lead to uncontrolled cell division and growth and can stimulate ethylene production.	2,4-D, aminocyclopyrachlor, aminopyralid, clopyralid, dicamba, MCPA, quinclorac, triclopyr, others

For a more complete listing and description of herbicide modes of action, refer to the Weed Science Society of America (WSSA) website at <http://wssa.net/wp-content/uploads/WSSA-Mechanism-of-Action.pdf>.

apparent (Jasieniuk et al. 1996; Maxwell et al. 1990).

### Current status of herbicide resistance

The strongest selection pressure for herbicide-resistant weeds tends to be in modern, high-intensity agricultural cropping systems due to a high reliance on herbicides. According to the International Survey of Herbicide Resistant Weeds (weedsscience.org), since the first confirmed report of a resistant biotype in

1957, herbicide-resistant weed biotypes have been reported in at least 60 countries and include more than 400 unique species-herbicide group combinations (fig. 2A). The United States has more herbicide-resistant biotypes (162) than any other country (fig. 2B), and California accounts for 21 of these (fig. 2C, table 2). Worldwide, resistance to acetolactate synthase (ALS)-inhibiting herbicides and photosystem II (PSII)-inhibiting herbicides (Groups 5, 6 and 7) are the most

commonly occurring among weedy species. However, in recent years, glyphosate (glycine herbicide) resistance and multiple resistances (resistance to two or more herbicides with dissimilar modes of action) have also emerged as major problems in some cropping systems. Interestingly, while herbicide resistance in the United States as a whole is primarily found in broadleaf weeds, California has more herbicide-resistant grasses or sedges (15) than broadleaf species (6) (table 2).

Due to the extensive use of preplant and in-season tillage in some agronomic crops in California, along with the use of pre- and postemergence herbicides, herbicide resistance is not as widespread as it is in other parts of the country where no-till and minimum-till systems have been widely adopted. Reduced tillage systems are heavily reliant on a few herbicide modes of action (e.g., glyphosate) and have correspondingly larger problems with herbicide resistance (Culpepper 2006).

In contrast to the rest of the United States, where herbicide resistance problems are centered on agronomic crops, the greatest problems with herbicide-resistant weeds in California are in orchards, vineyards, flooded rice, roadsides and irrigation canal banks. Herbicide-resistant weeds have become especially



Glyphosate-resistant horseweed in a raisin vineyard near Parlier, left, and glyphosate-resistant ryegrass in a walnut orchard near Davis.

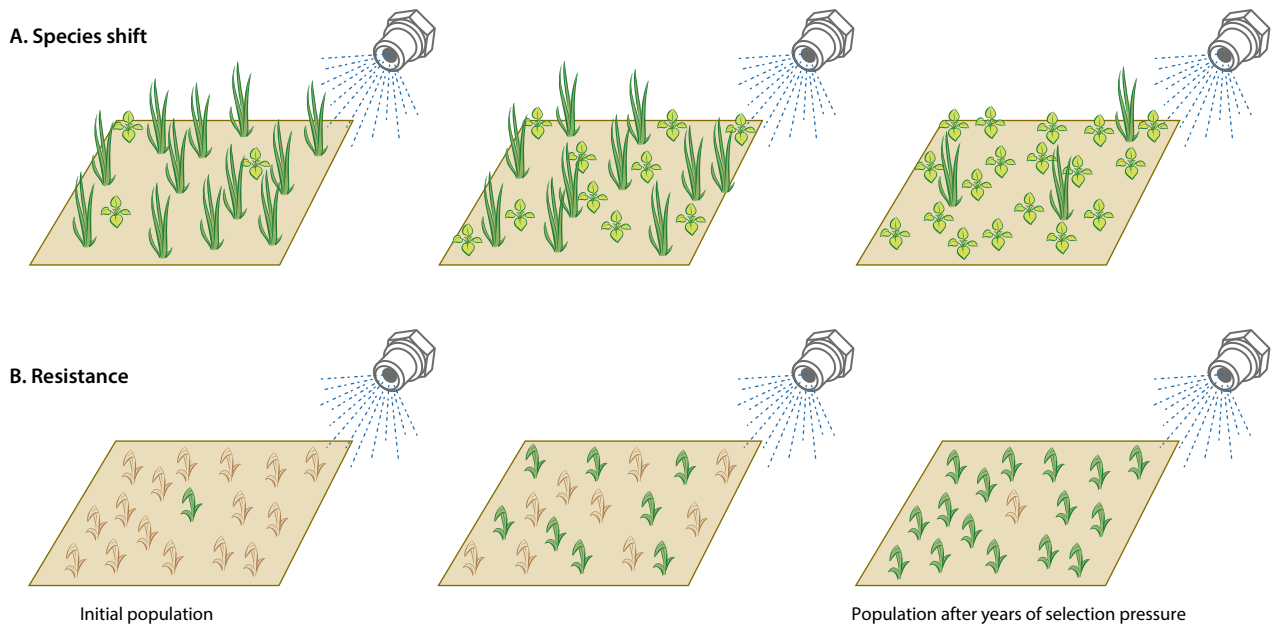


Fig. 1. Herbicides impose selection pressure and can lead to weed species shifts, resulting in populations dominated by more-tolerant species (A). Occasionally, an individual weed has a mutation that confers resistance to an herbicide or group of herbicides, and this individual survives and reproduces despite being treated with herbicide (B). In both cases, after several generations and repeated selection with the same or similar herbicides, the tolerant species or resistant biotype can become dominant in the population. (Modified from Orloff et al. 2009 with permission.)

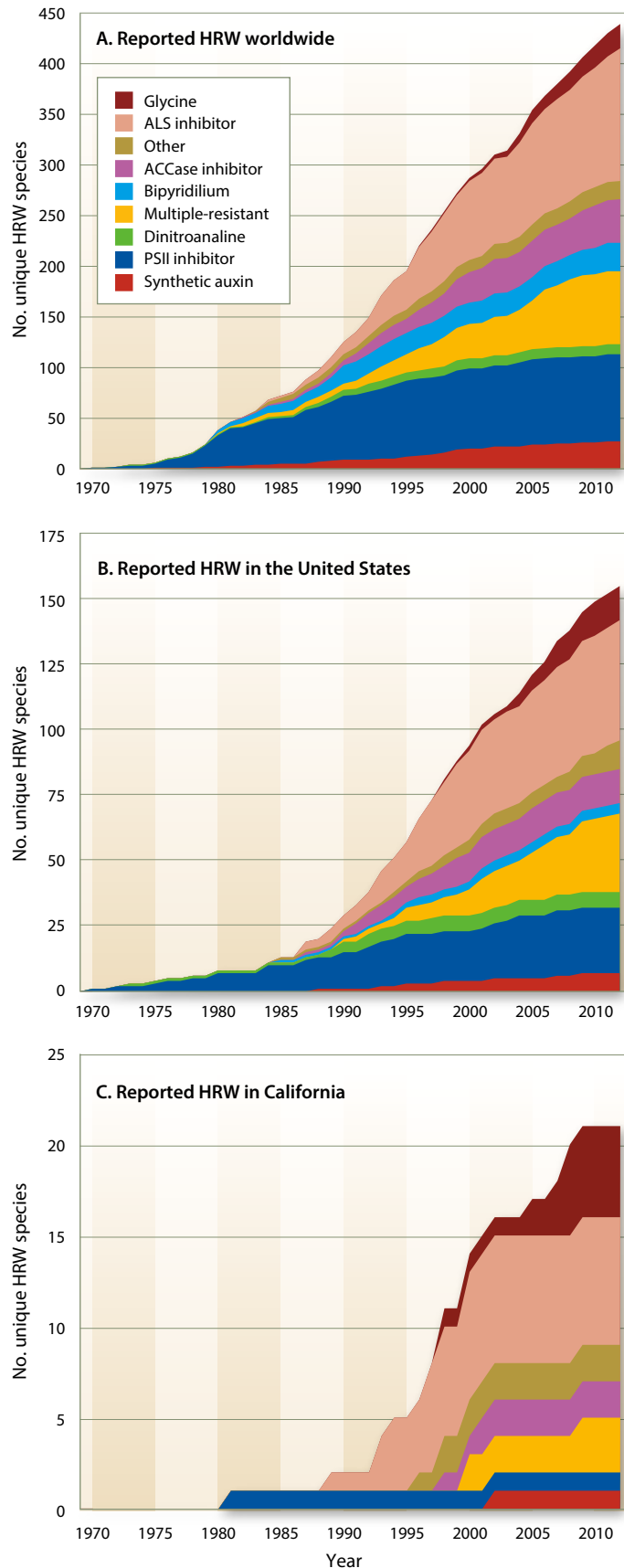


challenging problems in California's signature cropping systems, which are characterized by little or no crop rotation due to soil limitations (rice) or long cropping cycles (orchards and vineyards) and relatively few opportunities for mechanical weed control. Although large by specialty crop standards, the approximately 3 million acres devoted to orchard, vineyard and rice production in California is a small market for herbicide manufacturers; thus, herbicide options are somewhat limited. Combined, these factors have led to a high degree of selection pressure for herbicide-resistant weed biotypes as well as weed population shifts to naturally tolerant species (Hanson et al. 2013; Prather et al. 2000).

**TABLE 2. Confirmed cases of herbicide-resistant weeds in California**

Scientific name	Common name	Year	Mode of action*
<i>Senecio vulgaris</i>	Common groundsel	1981	PSII inhibitor
<i>Lolium perenne</i>	Perennial ryegrass	1989	ALS inhibitor
<i>Cyperus difformis</i>	Smallflower umbrella sedge	1993	ALS inhibitor
<i>Sagittaria montevidensis</i>	California arrowhead	1993	ALS inhibitor
<i>Salsola tragus</i>	Russian-thistle	1994	ALS inhibitor
<i>Avena fatua</i>	Wild oat	1996	Pyrazolium (difenzoquat)
<i>Ammannia auriculata</i>	Eared redstem	1997	ALS inhibitor
<i>Schoenoplectus mucronatus</i>	Ricefield bulrush	1997	ALS inhibitor
<i>Echinochloa phyllopogon</i>	Late watergrass	1998	Thiocarbamate
<i>Echinochloa phyllopogon</i>	Late watergrass	1998	Multiple (ACCase inhibitor, ALS inhibitor, thiocarbamate and clomazone)
<i>Lolium rigidum</i>	Rigid ryegrass	1998	Glycine
<i>Ammannia coccinea</i>	Redstem	2000	ALS inhibitor
<i>Echinochloa crus-galli</i>	Barnyardgrass	2000	Multiple (ACCase inhibitor and thiocarbamate)
<i>Echinochloa phyllopogon</i>	Late watergrass	2000	Thiocarbamate
<i>Echinochloa oryzoides</i>	Early watergrass	2000	Multiple (ACCase inhibitor and thiocarbamate)
<i>Phalaris minor</i>	Small-seeded canarygrass	2001	ACCase inhibitor
<i>Digitaria ischaemum</i>	Smooth crabgrass	2002	Synthetic auxin
<i>Conyza canadensis</i>	Horseweed	2005	Glycine
<i>Lolium perenne</i> ssp. multiflorum	Italian ryegrass	2005	Glycine
<i>Conyza bonariensis</i>	Hairy fleabane	2007	Glycine
<i>Echinochloa colona</i>	Junglerice	2008	Glycine
<i>Conyza bonariensis</i>	Hairy fleabane	2009	Multiple (glycine and bipyridylum)
<i>Cyperus difformis</i>	Smallflower umbrella sedge	2013	PSII
<i>Poa annua</i>	Annual bluegrass	2013	Glycine

\* PSII = photosystem II, ALS = acetolactate synthase, ACCase = acetyl coenzyme A carboxylase.



**Fig. 2. Chronological increase in reports of herbicide-resistant weeds (HRW) worldwide and in the United States and California. Data compiled in August 2013 from the International Survey of Herbicide Resistant Weeds (weedscience.org).**

## UC weed scientists address herbicide resistance in weeds

In order to combat complex issues such as herbicide resistance, organized collaborations between weed scientists and other agricultural researchers with a wide array of expertise are required. This includes the activities of UC Cooperative Extension farm advisors and specialists, Agricultural Experiment Station faculty, support scientists, research staff and graduate students, as well as faculty from other universities and agricultural industry representatives (for a list of UC weed scientists, visit the Weed Research and Information Center at [wric.ucdavis.edu](http://wric.ucdavis.edu)). Current herbicide-resistant weed management efforts range from applied research and extension efforts to basic plant biology and evolutionary ecology studies. Although the specifics vary, these efforts can be grouped into three general areas: (1) applied management of herbicide-resistant plants, (2) physiology and mechanisms of resistance and (3) biology, ecology and evolution of herbicide resistance.

**Applied management of herbicide-resistant plants.** Many cases of herbicide resistance in weeds are identified after growers, land managers or pest control advisers observe weed control failures with treatments that were once effective. These weeds are generally brought to the attention of local or statewide Cooperative Extension personnel. If the herbicide application method is ruled out as the cause of poor weed control (i.e., incorrect product, rate, timing, placement,

etc.), researchers often conduct field or greenhouse tests to verify and quantify the level of resistance. Plants from the suspected herbicide-resistant population are treated with the herbicide of interest at rates ranging from below normal doses to doses well above those legally allowed in the field (see photos, below). The response (i.e., plant growth or mortality) of the putative resistant population is then compared with the response of the known susceptible, or wild-type, population. Resistance is confirmed if the herbicide affects the two (or more) populations of the same species in markedly different ways with respect to plant growth and survival. In many cases, an estimate of the level of resistance also is made from these data. For example, if the susceptible population is controlled at one-half the field rate, but the resistant population survives at twice the field rate, it would be described as having a fourfold ( $2 / 0.5 = 4$ ) level of resistance.

**Physiology and mechanisms of herbicide resistance.** Identifying and verifying herbicide resistance and developing alternative management strategies provides short-term solutions for weed managers. Researchers often conduct further studies to determine the underlying molecular and physiological causes of resistance and to compare the biology, growth and competitive ability of herbicide-resistant species and biotypes. The mechanism(s) and fitness costs of herbicide resistance can have important ramifications on the selection, spread and competitive ability of herbicide-resistant biotypes, in addition to directly impacting their management. The

goal of these efforts is to help growers and pest control advisers recognize the importance of taking a proactive approach to preventing the evolution of a resistant population, rather than a reactive approach to managing herbicide resistance after it occurs.

Target-site resistance occurs when the enzyme that is the target of the herbicide becomes less sensitive, or fully insensitive, to the herbicide, often due to a physical change in the target enzyme's structure. These physical changes can impair the ability of the herbicide (or other herbicides) to attach to a specific binding site on the enzyme, thus reducing or eliminating herbicidal activity. Target-site resistance is sometimes evaluated at the tissue level using portions of plants such as leaves, leaf disks or roots (see photos, next page). In some cases, a functioning target enzyme (e.g., ALS or acetyl coenzyme A carboxylase [ACCase]) can be extracted and its function evaluated in laboratory in vitro experiments in the presence or absence of the herbicide. Recently, overproduction or enhanced activity of the target enzyme has been shown to confer herbicide resistance in certain cases (Gaines et al. 2011).

Several mechanisms of nontarget-site resistance confer resistance to herbicides in plants without involving the target sites of the herbicides. This can result in unpredictable resistance to unrelated herbicides (Délye 2013; Délye et al 2013). Of these, the best-known cases involve resistance in which herbicide-resistant plants have an enhanced ability to metabolically degrade the herbicide to less- or



Orchard-collected junglerice plants 21 days after treatment in a greenhouse dose-response experiment. The pot at the farthest left in each photo was untreated, and the remaining plants were treated with glyphosate rates ranging from (left to right)  $\frac{1}{32} \times$ ,  $\frac{1}{16} \times$ ,  $\frac{1}{8} \times$ ,  $\frac{1}{4} \times$ ,  $\frac{1}{2} \times$ ,  $1 \times$ ,  $2 \times$  and  $4 \times$  of the labeled use rate. The glyphosate-susceptible population was controlled with a  $\frac{1}{4}$  use rate, while the resistant population had some survivors at the  $4 \times$  rate.

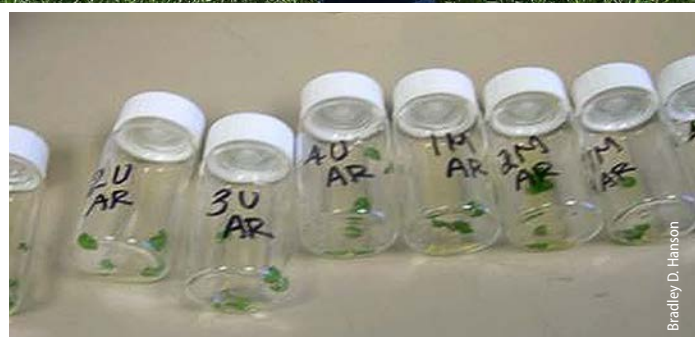


nontoxic forms. Many processes can be involved in metabolic resistance, but the most well-understood cases are due to changes in three groups of isozymes (cytochrome P450 monooxidases, glutathione transferases and glycosyltransferases) and changes in ATP-binding cassette (ABC) transporters (Yuan et al. 2007). This type of resistance is most commonly evaluated using nonherbicide inhibitors of the various isozymes in the presence or absence of the herbicide and comparing metabolic degradation of the herbicide in laboratory or greenhouse assays.

**Biology, ecology and evolution of herbicide resistance.** Many factors influence the evolution of herbicide resistance in weed populations (reviewed in Jasieniuk et al. 1996). To design effective resistance management strategies for the long term, UC and other scientists are conducting basic research on weed biology and on ecological and evolutionary processes in weed populations.

In a few cases, the mechanisms that confer resistance to herbicides have altered the fitness (i.e., survival, growth and/or seed production) of resistant plants, as compared with susceptible plants of the same species in the absence of herbicide treatment. Differential plant fitness among biotypes can affect the rate at which herbicide resistance can spread. For example, if resistant and susceptible plants have equal fitness, the number of resistant plants in the population would not change relative to the number of susceptible plants during periods when the herbicide was not being applied (Jasieniuk et al. 1996). In contrast, if resistant plants are less fit than susceptible plants, the number of resistant plants may decrease during periods when herbicide is not applied. Fitness is usually evaluated by growing resistant and susceptible plants in direct competition with one another, or with the crop of interest, and comparing relative productivity or fecundity.

Similar to efforts for other invasive weeds, insects and disease pathogens, surveys are sometimes used to delineate the extent of population growth or the expansion of new herbicide-resistant weed biotypes. Because there often are a few escaped weeds in herbicide-treated fields, herbicide resistance may not be recognized until the resistant biotype makes up a significant portion of the local population (Vencill et al. 2012). Surveys



In some cases of herbicide-resistant weeds, enzyme- or tissue-level assays can be used to understand and quantify resistance. Above, a lab assistant collects leaves from suspected glyphosate-resistant horseweed; left, leaf disks from the intact leaves are cut for an *in vivo* assay; right, disks are incubated overnight in the laboratory in buffer solutions containing various concentrations of glyphosate in order to evaluate activity of the EPSPS enzyme.

can help inform growers of emerging herbicide-resistant weed populations while they are still localized; surveys are also often used to encourage adoption of resistance mitigation measures to minimize economic and environmental impacts. Further, surveys combined with population genetic research can determine the evolutionary and geographic origins, and routes of spread, of resistance across an agricultural landscape (e.g., Okada et al. 2013; Okada et al. 2014).

### Herbicide resistance in California

Herbicide resistance has been an important management concern in California flooded rice production for several years (Busi et al. 2006). Weeds with resistance to the ALS inhibitors (Group 2), thiocarbamates (Group 8) and ACCase inhibitors (Group 1) are the dominant weed management problems in most of the Sacramento Valley rice production region. In orchards and vineyards, herbicide resistance is a more recent development and is dominated by resistance to the broad-spectrum postemergence herbicide

glyphosate. This herbicide is, by far, the most widely used herbicide in the state in perennial crop production systems, as well as in many roadsides, canal banks and residential and industrial areas. Glyphosate-tolerant (Roundup Ready) cotton, alfalfa and corn are becoming widely adopted in the state, which will further increase selection pressure for additional glyphosate-resistant and -tolerant species.

### Herbicide resistance in flooded rice.

Most California rice is produced in monoculture systems due to impeded soil drainage, which limits rotation to other upland crops (Hill et al. 2006). Rice fields are kept under continuous flood conditions during the growing season, primarily for the control of grass weeds (Strand 2013). Although this system favors sedges and other water-tolerant weeds, selective herbicides such as molinate and bensulfuron provided highly effective weed control for several years. However, in the early 1990s, after repeated use, resistance to the ALS-inhibiting herbicide bensulfuron became widespread among weedy species in rice. By 2000, several additional

weed biotypes with resistance to ALS inhibitors, thiocarbamates or ACCase inhibitors had evolved and were causing significant weed management, economic and environmental issues in the rice cropping system. UC researchers, extension personnel and industry partners have devoted considerable efforts to understanding and managing herbicide-resistant weeds in rice.

**Smallflower umbrella sedge** (*Cyperus difformis*) and **California arrowhead** (*Sagittaria montevidensis*) resistance to ALS-inhibiting herbicides was first reported in California rice fields in 1993 following repeated use of bensulfuron (Hill et al. 1994). Field research has shown that California arrowhead is a fairly weak competitor in rice systems (Gibson et al. 2001) and that the ALS-resistant biotypes can be adequately controlled with other registered herbicides. Recently, smallflower umbrella sedge biotypes with multiple resistance to the PSII herbicide propanil and to several ALS-inhibiting herbicides were identified in the Sacramento Valley (Valverde et al. 2014), and research is ongoing to elucidate the mechanisms of resistance and any cross resistance to other rice herbicides.

**Eared redstem** (*Ammannia auriculata*) and **ricefield bulrush** (*Schoenoplectus mucronatus*) resistance to ALS inhibitor herbicides in rice was reported in 1997. Redstem research has focused on intra- and interspecific competition in an effort to develop agronomic solutions to reduce its competition with rice (Caton et al. 1997; Gibson et al. 2003). Studies have shown that California populations of ricefield bulrush are resistant to most registered ALS inhibitors, whereas populations from other regions are resistant only to one chemical family, the sulfonylureas, in the ALS inhibitor group (Busi et al.

2006). Recently, ricefield bulrush biotypes

with multiple resistance to propanil and bensulfuron were identified in the Sacramento Valley (Abdallah et al. 2014).

**Late watergrass** (*Echinochloa phyllopogon*) populations resistant to ACCase inhibitors, ALS inhibitors and the thiocarbamate herbicides in rice systems were reported in 1998 (Fischer, Ateh et al. 2000; Fischer, Bayer et al. 2000). This resistance to multiple herbicides within an individual plant indicated that using herbicides with different modes of action would be unlikely to provide satisfactory control of the species in the long term. Further complicating the situation in rice, populations of late watergrass and **barnyardgrass** (*Echinochloa crus-galli*) with resistance to both ACCase inhibitors and thiocarbamates, and thus exhibiting multiple resistance, were reported in 2000. Later research confirmed that the mechanisms of multiple resistance to several herbicide classes are due to metabolic degradation of these compounds (Yasuor et al. 2008, 2009).

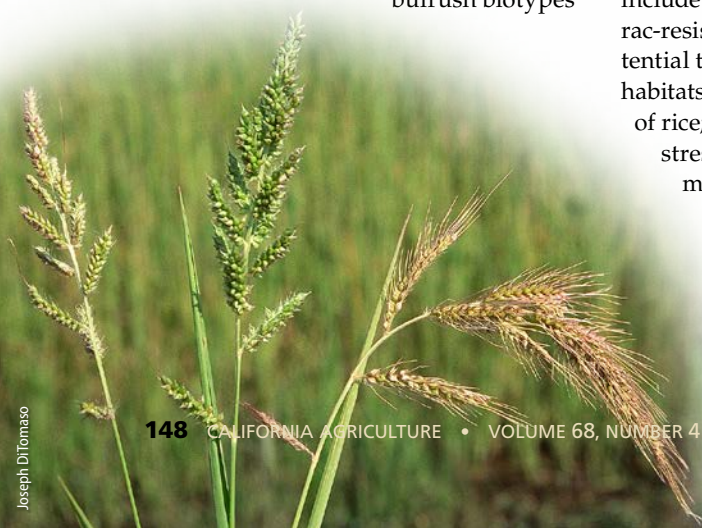
**Smooth crabgrass** (*Digitaria ischaemum*) resistance to the synthetic auxin herbicide quinclorac was reported in 2002. Detailed research into the mechanisms of resistance suggested that the cause was an altered sensitivity in the auxin response pathway, leading to ACCase activity, ethylene synthesis and enhanced ability to detoxify cyanide (a byproduct of ethylene biosynthesis) (Abdallah et al. 2006). Although crabgrass is not an important rice weed, quinclorac is used in rice systems for control of other weeds, and resistance to it has been reported in *Echinochloa* species of rice in California (Yasuor et al. 2011) and from other regions. Most importantly, the observed changes in ethylene synthesis and production of toxic byproducts may also relate to the plant's ability to tolerate abiotic stress. Two implications of this finding include the possibilities that (1) quinclorac-resistant smooth crabgrass has the potential to invade a more diverse range of habitats and become an important weed of rice; and (2) adaptation to the abiotic stress of the flooded environments may predispose *Echinochloa phyllopogon* or other major rice weeds to evolve resistance to quinclorac in the future.

**Barnyardgrass** (*Echinochloa crus-galli*).

**Herbicide resistance in orchard and vineyard cropping systems.** The first herbicide-resistant weed in orchard cropping systems was perennial ryegrass, *Lolium perenne* (now named *Festuca perennis* spp. *perenne*), reported in 1989 (Heap 2013). This ALS inhibitor-resistant biotype was selected on roadsides by the use of sulfometuron and, thus far, has not been a major problem in orchards or vineyards because relatively little of this class of herbicides is used in these crops. However, several ALS inhibitors, including rimsulfuron, penoxsulam, halosulfuron and flazasulfuron, are becoming more widely used in tree and vine crops, and selection pressure for ALS inhibitor resistance may increase in the future.

The first case of glyphosate resistance in California was reported in a population of **rigid ryegrass** (*Lolium rigidum*, now *Festuca perennis* spp. *rigidium*) in 1998 (Simarmata and Penner 2008). However, most confirmed glyphosate-resistant ryegrass populations have been identified as **Italian ryegrass** (*Lolium multiflorum*, now *Festuca perennis* spp. *multiflorum*) (Sherwood and Jasieniuk 2009). Glyphosate-resistant ryegrasses have become widespread and are a major weed problem in orchards, vineyards and roadsides of Northern California (Jasieniuk et al. 2008). Research indicated that resistance in ryegrass is not due to metabolism of the herbicide and is instead due to an altered EPSPS enzyme (Jasieniuk et al. 2008; Simarmata and Penner 2008). Glyphosate resistance in these areas has been largely driven by decreases in grower use of other herbicides, especially those under increasing regulatory pressure because of pesticide contamination of ground or surface water. The use of glyphosate-based herbicide programs also increased when the patent on Roundup expired in 2000 and low-cost, generic glyphosate herbicides became readily available. Today, glyphosate accounts for over 60% of all herbicide-treated acreage in California orchard and vineyard systems (DPR 2013).

Glyphosate-resistant **horseweed**, or mare's tail (*Conyza canadensis*), was reported in 2005 and is one of the dominant weeds in and around raisin and tree fruit production areas of the San Joaquin Valley, as well as on roadsides and canal banks in the region (Hanson et al. 2009; Hembree and Shrestha 2007; Shrestha,





# Importance of herbicide resistance in weeds of natural areas

by Joseph M. DiTomaso

Worldwide, the majority of the plant species that are developing herbicide resistance are those that occur as weeds in agricultural environments, on roadsides and in other rights-of-way. In contrast, herbicide resistance is not nearly so common in weeds of natural areas or rangelands. A search of the International Survey of Herbicide Resistant Weeds ([weeds-science.com](http://weeds-science.com)) revealed no herbicide-resistant weeds (i.e., invasive nonnative species) listed for terrestrial natural areas anywhere in the world, and only two resistant weeds listed for aquatic areas, both of them in Florida. In pastures, 15 species worldwide have developed resistance, eight of which are considered primarily as agricultural weeds. Only two of those 15 are found in pastures within the United States, and none occurs in any Western state.

The reason more weeds develop herbicide resistance in agricultural and right-of-way systems has to do with factors associated with characteristics of specific weeds, herbicides and weed management practices. For example, high seed production increases the opportunity for genetic variation, and with it the probability that a resistance adaptation will occur. It so happens that all of the major weeds that have developed resistance to herbicides are annuals. In an agricultural system, annual species make up the vast majority of problematic weeds. Annuals can have high seed production, rapid turnover of the seedbank (due to a high percentage of seed germination each year) and, in some cases, several reproductive generations per growing season. This increases the selective pressure for herbicide-resistant biotypes. In natural areas of California, the California Invasive Plant Council (Cal-IPC) lists 214 flowering plants as invasive ([cal-ipc.org](http://cal-ipc.org)). Of these, only 27.5% are annual species; the remainder (and the majority) are either woody species or herbaceous perennials or biennials. Perennial weeds, and particularly those with vegetative reproductive tissues, are less likely than annuals to evolve herbicide resistance.

The choice of herbicide can also increase or decrease the likelihood that weeds will develop herbicide resistance. In most natural areas, herbicides are not used as intensively as in croplands, where it is common to repeat herbicide applications within a single year or over several consecutive years. In addition, fewer herbicides are available for use in natural areas of California, and the most widely used compounds (e.g., 2,4-D, aminopyralid, dicamba, triclopyr or clopyralid) belong to the growth regulator chemical families. Resistance to these herbicides does not develop as commonly as resistance to other herbicide families, despite their having been available and extensively used for a long time. Glyphosate is also commonly used in natural areas, and although glyphosate resistance is on the rise in cropping systems, its development is often associated with repeated applications over multiple years, a strategy not generally used in natural areas.

Weed management practices are often the most important contributing factors leading to the selection of herbicide-resistant biotypes. In general, a land manager's complete and repeated reliance on a single herbicide or mode of action for weed control can greatly enhance the occurrence of herbicide-resistant weeds. This is particularly true when the manager uses no other

weed control option, such as mechanical or cultural control practices. For a number of reasons, including economic feasibility and the potential for damage to desirable (nontarget) vegetation, it is uncommon for a land manager to reapply the same herbicide for several consecutive years in a natural area.

Because the evolution of herbicide resistance is typically the result of intensive, persistent selective pressure on a rapidly regenerating weed population (i.e., annual species), the incidence of herbicide-resistant species would be expected to be much higher in a cropping system with limited rotations or in other systems,



**Yellow starthistle (*Centaurea solstitialis*) infestation, left; aerial spraying to control yellow starthistle near Sierra Foothill Research and Extension Center, right.**

such as rights-of-way, that are continuously managed with herbicides. In many natural areas, the effort to manage invasive plants can involve several different control strategies besides, or instead of, herbicide application. These can include mechanical means such as mowing, cultural methods including grazing management or prescribed burning and, when available, biological control agents. Furthermore, even when herbicides are used, they are rarely applied repeatedly over a long period of time. The total area of noncropped lands treated with herbicides is far smaller than the total acreage of agricultural land treated with herbicides. It is hardly surprising, then, that the incidence of herbicide resistance in natural areas and rangelands is low — in fact, it is not even reported as present in California.

Regardless of the vegetative environment, whether natural or agricultural, prevention of herbicide resistance and management of established resistant weed populations could be accomplished more effectively if we put a greater reliance on integrated weed management approaches. Although the likelihood that resistance will develop in natural areas is already low, management strategies that employ rotation of herbicides with different modes of action, the use of competitive species in restoration programs, and a combination of mechanical, biological and cultural control options in an integrated management program will further reduce the selective pressure on invasive plant populations and with it the potential that weeds will develop herbicide resistance.

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Jack Kelly, Clark

Hembree, Wright 2008; Shrestha et al. 2010). The level of glyphosate resistance in horseweed is relatively low, and resistant plants are usually injured to some degree following glyphosate applications, which suggests that resistance is not due to an altered target enzyme. Genetic comparisons of horseweed accessions from around the state suggest that there have been multiple, independent origins of resistance in this species, rather than the spread of resistance from a single-source population (Okada et al. 2013).

**Hairy fleabane** (*Conyza bonariensis*) populations resistant to glyphosate were reported in 2007 (Shrestha, Hanson,

Hembree 2008). Glyphosate resistance in hairy fleabane appears to be similar to resistance in horseweed in that (1) selection has occurred in response to similar management strategies in perennial crops and surrounding areas (Hembree and Shrestha 2007); (2) multiple origins of resistance are suspected (Okada et al. 2014); and (3) growth stage and environmental conditions affect the level of resistance (Moretti, Hanson et al. 2013; Shrestha et al. 2007). The discovery by Moretti, Hanson et al. (2013) of hairy fleabane resistant to both glyphosate and paraquat raises questions about whether a common physiological mechanism is helping to

confer resistance to these dissimilar herbicides, and research is ongoing to elucidate these factors.

**Junglerice** (*Echinochloa colona*) resistant to glyphosate was first identified in 2008 in a Roundup Ready corn field in the Sacramento Valley (Alarcon-Reverte et al. 2013); since then, glyphosate-resistant junglerice has become widespread in orchards and field crops throughout California (Moretti, Garcia et al. 2013). Resistance appears to be due to mutations in the EPSPS target site (Alarcon-Reverte et al. 2013), although some populations also appear to have enhanced EPSPS activity (A.J. Fischer, unpublished data).

## Herbicide-resistant weeds unlikely in vegetable crops

by Steve Fennimore, Richard Smith and Michelle Le Strange

**W**eed management systems in California vegetable crops can be described as robust, complex, multitactic and integrated. Vegetable herbicides generally make up just one component in a multicomponent weed management system. With California's seasonally dry weather and growers' ability to control soil moisture by means of irrigation scheduling, it becomes possible for the grower to apply effective cultural and physical control practices, such as preparation of stale seedbeds and inter-row cultivation. Redundancy is designed into the weed management system to minimize weed emergence in the crop. The key tools that make up an integrated vegetable weed management system are field selection, sanitation, crop rotation, land preparation, stale seedbeds, herbicides and physical weed control (UC IPM 2009). Growers who carefully apply these practices are able to manage weeds effectively and reduce the presence of weed seeds in the soil seedbank.

**Field selection.** Farmers often grow vegetable crops on fields that have low weed pressure so their weed control operations can be more efficient and economical. They use translocated herbicides during fallow periods to control troublesome perennial weeds like field bindweed.

**Sanitation.** Growers often keep vegetable fields and the surrounding areas as weed-free as possible to keep the weeds from going to seed. Some operations that utilize a "zero weed seed" philosophy have successfully reduced weed pressure in subsequent vegetable crops by eliminating weed seed inputs to the soil seedbank. Other measures such as cleaning all field equipment when moving it from a weedy field or into a clean field are also employed.

**Rotation.** By growing vegetable crops in rotation with crops that normally have more intensive weed control programs, growers can help keep a field clean of weeds. Because field conditions are constantly changing under a rotation system, no one weed species is likely to become dominant.

**Land preparation.** Direct-seeded vegetable crops require well-prepared seedbeds free of large clods to facilitate precision planting and allow rapid and uniform emergence of vegetable seedlings. A uniform seeding depth is critical to uniform crop emergence

and improved tolerance to herbicides. Mechanical cultivation is facilitated with smooth seedbeds and good tillth, which allows the cultivation equipment to remove weeds that are close to the crop row. Increasingly, growers are using precision guidance systems to improve the speed and accuracy of cultivation.

**Preirrigation and use of a stale seedbed.** Preirrigation before final seedbed preparation is a common practice, as it stimulates a weed flush a few days after watering. As soon as the weeds have emerged and the field is dry enough to enter, the grower uses shallow cultivation, flaming or a nonselective herbicide to remove the new weeds. Research has shown this technique to provide 15% to 50% control of weeds in crops like lettuce (Shem Tov et al. 2006). The combination of stale seedbed technique and both herbicides and cultivation often results in good weed control.

**Herbicides.** One category of herbicide used in vegetable crops is fumigants, such as metam potassium, which is applied 14 to 21 days before planting to kill weed seeds and germinating seedlings. After planting, soil-active herbicides like pronamide (used in artichokes and head lettuce) and S-metolachlor and trifluralin (used in tomatoes and peppers) are applied to provide preemergence control of weeds. Postemergence herbicides are utilized in some crops; examples include clethodim, used to control emerged grass weeds in many broadleaf vegetable crops, and oxyfluorfen and bromoxynil, used to control broadleaf weeds. Many vegetable herbicides were developed in the 1960s and 1970s and include products like DCPA (used in broccoli and onion), napropamide (used in tomatoes and peppers) and linuron (used in asparagus and celery). Given the complexity of the vegetable weed control program and the extensive use of cultivation and hand-weeding, the selective pressure on weeds from vegetable herbicides is very light, despite their decades of use.

**Physical weed control.** Vegetable growers make extensive use of physical weed control. One example is inter-row cultivation or shallow cultivation between the crop rows to control weeds. Inter-row cultivation is a very old but effective method that buries, cuts or uproots weeds. Hand-weeding by workers with hoes is the last



Target-site mutations appear to be the most frequent mechanism among the accessions so far collected in California; however, additional research is ongoing to determine whether the same is true with populations selected in orchards and in other regions of the Central Valley.

Several other common weeds in orchards and vineyards, including **Palmer amaranth** (*Amaranthus palmeri*), **three-spike goosegrass** (*Eleusine tristachya*) and **witchgrass** (*Panicum capillare*), are suspected to have evolved resistance to glyphosate; preliminary research trials by UC researchers and California State University, Fresno, collaborators have

been initiated to verify and characterize the putative resistant populations.

### **California herbicide resistance research: Locally applied research and extension with national and international implications**

Since the discovery of herbicide-resistant weed biotypes in California, UC research and Cooperative Extension personnel, as well as university and non-university cooperators and students, have conducted locally relevant weed management research in the state. Research and extension efforts have included alternative chemical management techniques

using various postemergence and pre-emergence herbicides along with mechanical control measures in an integrated approach. Applied research integrating agronomy, weed control, spray application technology and water management have been useful to regulatory agencies (e.g., California Department of Pesticide Regulation and California Environmental Protection Agency) and have had positive impacts on water and air quality, wildlife habitat and water use (Pittelkow et al. 2012).

Information on the underlying mechanisms and genetic basis of resistance provides useful information to California

line of defense against weeds in vegetable crops. Among the hoeing crew, manual dexterity and good depth perception allow the workers to carefully remove weeds from the vegetable crop in the row and near the crop plant. Hand-weeding is expensive and can cost \$300 or more per acre in organic vegetable plantings and high-density plantings (e.g., spinach and baby lettuces) — sometimes a lot more.

**Integrated weed management in lettuce.** In a typical lettuce weed control program, the crop is grown on a field with a light weed population, so one tool growers use is field selection. Sometimes the soil is fumigated with metam potassium before planting to control weeds and soilborne diseases, but most lettuce is grown on nonfumigated land. Prior to planting, the soil is irrigated to stimulate weed emergence and then shallow-tilled to kill weeds and form a smooth seedbed for planting. At time of seeding, pre-emergence herbicides such as pronamide or bensulide are applied, to be activated with the initial sprinkler irrigation. About 4 weeks after emergence, the lettuce is hand-thinned and weeded by a hoeing crew to its final stand. Inter-row cultivation in furrows and on bed tops is conducted one or more times, also removing weeds. Finally, about 6 weeks after lettuce emergence, the field is hand-weeded to remove any remaining weeds. After harvest, the field is quickly tilled under, killing any remaining weeds before the field is rotated to another crop.

**Integrated weed management in tomatoes.** Virtually all California tomatoes are transplanted, and 75% are grown using subsurface drip irrigation buried 8 to 10 inches deep. Fields with low weed populations, especially those free of field bindweed and dodder, are most often sought for tomato production. Beds are preirrigated to germinate weeds, then cultivated and shaped prior to planting. Typically only two herbicide applications are made: one just prior to planting or at planting, and the other at layby. Herbicides such as halosulfuron, pendimethalin, rimsulfuron, S-metolachlor, sulfentrazone and trifluralin are used, depending upon the site and weed spectrum. Just prior to layby application, beds and furrows are mechanically cultivated.

These practices significantly reduce weed emergence and competition against the young tomato crop. Hoeing crews may hand-weed once or twice before or after layby, depending on weed

populations. Adding to the cost for growers who practice “zero weed seed tolerance” is the physical removal of troublesome weeds such as flowering nightshades and dodder to prevent seeding and further field contamination. The harvest operation undercuts all plants growing on the bed top, and after harvest the field is quickly tilled under, killing any remaining weeds before the field is rotated to another crop.

The lettuce and tomato weed management systems are intensive and redundant, made up of many operations conducted in sequence with the aim of minimizing weed emergence. In practice, these weed management systems are not always as flawless as the above descriptions might suggest. Crops like broccoli and cauliflower are grown during winter months, when extended rain and wet field conditions prevent cultivation and hand-weeding. Other complications are high-density plantings such as those used for spinach, which limit the grower’s ability to cultivate.

Overall, the chances are low that weeds will develop herbicide resistance in a vegetable crop planting. Technology is evolving that will allow intelligent robotic cultivators to remove weeds from the intra-row space without the use of herbicides, so there is reason for optimism that the development of herbicide-resistant weeds in California vegetable fields will remain low for the foreseeable future.

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### **References**

- Shem-Tov S, Fennimore SA, Lanini WT. 2006. Weed management in lettuce (*Lactuca sativa*) with pre-plant irrigation. *Weed Technol* 20:1058–65.
- UC IPM. 2009. Integrated weed management. In: Spinach: UC IPM Pest Management Guidelines. UC ANR Pub 3467. Oakland, CA. [www.ipm.ucdavis.edu/PMG/r732700111.html](http://www.ipm.ucdavis.edu/PMG/r732700111.html).

weed managers in the longer term. This information is broadly applicable to the biology, physiology, evolution and control of weeds in other crops and regions at the local, national and international level. Although this paper has focused on the efforts of UC weed scientists and collaborators, the basic and applied scientific information developed in California supports similar research being conducted in other regions of the country and world.

Like many other areas encompassed by the Endemic and Invasive Pests and Diseases Strategic Initiative, solutions to

herbicide resistance are not simple and are affected by many biological, economic, regulatory and social factors. The diverse network of weed scientists and collaborators in a land-grant university system is well positioned to address these complex issues and respond to stakeholder concerns through applied and basic research, extension and outreach to affected agricultural industries, and education of future scientists and leaders. Ultimately, the goal of weed science research is to help growers maintain agricultural productivity and economic stability while increasing environmental sustainability. **CA**

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## References

- Abdallah I, Fischer AJ, Elmore CL, et al. 2006. Mechanism of resistance to quinclorac in smooth crabgrass (*Digitaria ischaemum*). *Pestic Biochem Physiol* 84:38–48.
- Abdallah IS, Garcia A, Fischer AJ. 2014. Rice field bulrush (*Schoenoplectus mucronatus* (L.) Palla) evolved multiple resistances to propanil and bensulfuron herbicides. *J Biol Chem Res* 31:788–99.
- Alarcon-Reverte R, Garcia A, Urzua J, Fisher AJ. 2013. Resistance to glyphosate in junglerice (*Echinochloa colona*) from California. *Weed Sci* 61:48–54.
- Busi R, Vidotto F, Fischer AJ, et al. 2006. Patterns of resistance to ALS herbicides in smallflower umbrella sedge (*Cyperus difformis*) and ricefield bulrush (*Schoenoplectus mucronatus*). *Weed Technol* 20:1004–14.
- Caton BP, Foin TC, Hill JE. 1997. Mechanisms of competition for light between rice (*Oryza sativa*) and redstem (*Ammannia* spp.). *Weed Sci* 45:269–75.
- [DPR] California Department of Pesticide Regulation. 2013. Pesticide use reporting. [www.cdpr.ca.gov/docs/pur/pur11rep/11\\_pur.htm](http://www.cdpr.ca.gov/docs/pur/pur11rep/11_pur.htm) (accessed on March 1, 2013).
- Culpepper SA. 2006. Glyphosate-induced weed shifts. *Weed Technol* 20:277–81.
- Délye C. 2013. Unraveling the genetics bases of non-target-site-based resistance (NTSR) to herbicides: A major challenge for weed science in the forthcoming decade. *Pest Manag Sci* 69:176–87.
- Délye C, Jasieniuk M, Le Corre V. 2013. Deciphering the evolution of herbicide resistance in weeds. *Trends Genet* 29:649–58.
- Fischer AJ, Ateh CM, Bayer DE, Hill JE. 2000. Herbicide-resistant *Echinochloa oryzoides* and *E. phyllopopogon* in California *Oryza sativa* fields. *Weed Sci* 48:225–30.
- Fischer AJ, Bayer DE, Carriere MD, et al. 2000. Mechanism of resistance to bispyribac-sodium in an *Echinochloa phyllopopogon* accession. *Pestic Biochem Physiol* 68:156–65.
- Gaines TA, Shaner DL, Ward SM, et al. 2011. Mechanism of resistance of evolved glyphosate-resistant Palmer amaranth (*Amaranthus palmeri*). *J Agr Food Chem* 59:5886–9.
- Gibson KD, Fischer AJ, Foin TC. 2001. Shading and the growth and photosynthetic responses of *Ammannia coccinea*. *Weed Res* 41:59–67.
- Gibson KD, Fischer AJ, Foin TC, Hill JE. 2003. Crop traits related to weed suppression in water-seeded rice (*Oryza sativa* L.). *Weed Sci* 51:87–93.
- Hanson B, Fisher A, Shrestha A, et al. 2013. Selection Pressure, Shifting Populations, and Herbicide Resistance and Tolerance. UC ANR Pub 8493. Oakland, CA. 6 p.
- Hanson BD, Shrestha A, Shaner DL. 2009. Distribution of glyphosate-resistant horseweed (*Conyza canadensis*) and relationship to cropping systems in the Central Valley of California. *Weed Sci* 57:48–53.
- Heap I. 2013. The International Survey of Herbicide Resistant Weeds. [www.weedscience.com](http://www.weedscience.com) (accessed August 30, 2013).
- Hembree K, Shrestha A. 2007. Control of glyphosate-resistant marestail and hairy fleabane in orchards and vineyards. *California Weed Science Society Proc* 59:113–7.
- Hill JE, Smith RJ, Bayer DE. 1994. Rice weed control: Current technology and emerging issues in temperate rice. *Aust J Exp Agr* 34:1020–9.
- Hill JE, Williams JF, Mutters RG, Greer CA. 2006. The California rice cropping system: Agronomic and natural resource issues for long-term sustainability. *Paddy Water Environ* 4:13–9.
- Holt JS, Stemler AJ, Radosevich SR. 1981. Differential light responses of photosynthesis by triazine-resistant and triazine-susceptible *Senecio vulgaris* biotypes. *Plant Physiol* 67:744–8.
- Jasieniuk M, Ahmad R, Sherwood AM, et al. 2008. Glyphosate-resistant Italian ryegrass (*Lolium multiflorum*) in California: Distribution, response to glyphosate, and molecular evidence for an altered target enzyme. *Weed Sci* 56:496–502.
- Jasieniuk M, Brule-Babel AL, Morrison IN. 1996. The evolution and genetics of herbicide resistance in weeds. *Weed Sci* 44:176–93.
- Maxwell BD, Roush ML, Radosevich SR. 1990. Predicting the evolution and dynamics of herbicide resistance in weed populations. *Weed Technol* 4:2–13.
- Moretti ML, Garcia AM, Fischer AJ, Hanson BD. 2013. Distribution of glyphosate-resistant junglerice (*Echinochloa colona*) in perennial crops of the Central Valley of California. *Proc Western Soc Weed Sci* 66:19.
- Moretti ML, Hanson BD, Hembree KJ, Shrestha A. 2013. Glyphosate resistance is more variable than paraquat resistance in a multiple-resistant hairy fleabane (*Conyza bonariensis*) population. *Weed Sci* 61:396–402.
- Okada M, Hanson BD, Hembree KJ, et al. 2013. Evolution and spread of glyphosate resistance in *Conyza canadensis* in California. *Evol Appl* 6:761–77.
- Okada M, Hanson BD, Hembree KJ, et al. 2014. Evolution and spread of glyphosate resistance in *Conyza bonariensis* in California and a comparison with closely related *C. canadensis*. *Weed Res* (in press).
- Orloff SB, Putnam DH, Canevari M, Lanini WT. 2009. Avoiding Weed Shifts and Weed Resistance in Roundup Ready Alfalfa Systems. UC ANR Pub 8362. Oakland, CA.
- Pimentel D, Zuniga R, Morrison D. 2005. Update on the environmental and economic costs associated with alien-invasive weeds in the United States. *Ecol Econ* 52:273–88.
- Pittelkow CM, Fischer AJ, Moechnig MJ, et al. 2012. Agronomic productivity and nitrogen requirements of alternative tillage and crop establishment systems for improved weed control in direct-seeded rice. *Field Crop Res* 130:128–37.
- Prather TS, DiTomaso JM, Holt JS. 2000. Herbicide resistance: Definitions and management strategies. UC ANR Pub 8012. Oakland, CA. 12 p.
- Sherwood AM, Jasieniuk M. 2009. Molecular identification of weedy glyphosate-resistant *Lolium* (Poaceae) in California. *Weed Res* 49:354–64.
- Shrestha A, Hanson BD, Fidelibus MW, Alcorta M. 2010. Growth, phenology, and intraspecific competition between glyphosate-resistant and glyphosate-susceptible horseweeds (*Conyza canadensis*) in the San Joaquin Valley of California. *Weed Sci* 58:147–53.
- Shrestha A, Hanson BD, Hembree KJ. 2008. Glyphosate-resistant hairy fleabane documented in the Central Valley. *Calif Agr* 62:116–9.
- Shrestha A, Hembree KJ, Va N. 2007. Growth stage influences level of resistance in glyphosate-resistant horseweed. *Calif Agr* 61:67–70.
- Shrestha A, Hembree K, Wright S. 2008. Biology and management of horseweed and hairy fleabane in California. UC ANR Pub 8314. Oakland, CA. p 9.
- Simarmata M, Penner D. 2008. The basis for glyphosate resistance in rigid ryegrass (*Lolium rigidum*) from California. *Weed Sci* 56:181–8.
- Strand L. 2013. *Integrated Pest Management for Rice* (3rd ed.). UC ANR Pub 3280. Oakland, CA. 98 p.
- Valverde BE, Boddy LG, Pedroso RM, et al. 2014. *Cyperus difformis* evolves resistance to propanil. *Crop Prot* 62:16–22.
- Vencill WK, Nichols RL, Webster TM, et al. 2012. Herbicide resistance: Toward an understanding of resistance development and the impact of herbicide-resistant crops. *Weed Sci* 60 (Special Issue):2–30.
- Yasuor H, Milan M, Eckert JW, Fischer AJ. 2011. Quinclorac resistance: A concerted hormonal and enzymatic effort in *Echinochloa phyllopopogon*. *Pest Manag Sci* 68:108–15.
- Yasuor H, Osuna MD, Ortiz A, et al. 2009. Mechanisms of resistance to penoxsulam in late watergrass (*Echinochloa phyllopopogon*). *J Agr Food Chem* 57:3653–60.
- Yasuor H, TenBrook PL, Tjeerdema JS, Fischer AJ. 2008. Response to clomazone and 5-ketochlomezone by *Echinochloa phyllopopogon* resistant to multiple herbicides in Californian rice fields. *Pest Manag Sci* 10:1031–9.
- Yuan JS, Tranel PJ, Stewart Jr CN. 2007. Non-target-site herbicide resistance: A family business. *Trends Plant Sci* 12:6–13.



# Over 35 years, integrated pest management has reduced pest risks and pesticide use

by Peter B. Goodell, Frank G. Zalom, Joyce F. Strand, Cheryl A. Wilen and Karey Windbiel-Rojas

*Pests and their interactions with crops, ecological landscapes and animals are in continuous flux — they are never static. Pest severity increases or decreases depending on environmental conditions and changes in production or pest control practices. Pest management is made even more challenging by exotic and newly invasive pests. Over its 35-year history, the UC Agriculture and Natural Resources Statewide IPM Program has supported research and extension that has decreased risks of crop losses, improved treatment programs for invasive and endemic pests, and reduced the use of pesticides and their impact on the environment and human health. Its publications are widely used among growers, pest control advisers, research institutions, state agencies, agricultural organizations and gardeners; and integrated pest management has been adopted statewide in agriculture, as well as in managed landscapes and urban areas.*



Elena Zhukova

In 2001, UC IPM and the Center for Invasive Species Research established a grant program to support research of exotic and invasive pests and diseases such as European grapevine moth, glassy-winged sharpshooter and Pierce's disease. Lab assistant Emily Kuhn checks a pheromone trap for European grapevine moth in a vineyard at UC Davis Oakville Research Station, Napa County. The moth, which is endemic to Mediterranean Europe, was first discovered in California in 2009.

Integrated pest management (IPM) is a systems approach to pest management. Because of the diverse situations in which pests occur, what constitutes IPM best practices may vary with time, location and other circumstances. IPM considers each available control tactic — for example, cultural, biological, chemical — and often applies a combination of tactics to enhance overall effectiveness and reduce reliance on any single tactical approach. It relies on extensive knowledge of the pest, the crop and the environment in which it exists. Regularly and frequently monitoring the status of a pest, its natural enemies and the site is fundamental to IPM decision making.

In 1979, the California legislature provided funding to the UC Division of Agricultural Sciences (now Agriculture and Natural Resources, ANR) to establish the UC Statewide Integrated Pest Management Program (UC IPM). The broad goals of the original program have remained consistent:

- Increase use of ecologically based integrated pest management programs.
- Provide leadership in IPM, including building coalitions and partnerships that link with communities and public agencies.
- Increase the predictability and effectiveness of pest management techniques.
- Develop science-based pest management programs that are economically and environmentally sustainable and socially appropriate.
- Protect human health and the environment by reducing risks caused by pests and pest management practices.

Since 1980, UC IPM scientists have worked with other ANR scientists to

Online: <http://californiaagriculture.ucanr.edu/landingpage.cfm?article=ca.v068n04p153&fulltext=yes>  
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conduct research on specific pests and pest systems and develop economically feasible and environmentally sound IPM programs. These programs have been extended to California growers by IPM advisors and other UC Cooperative Extension (UCCE) advisors.

Experience has shown that developing and implementing an IPM program is only the first challenge. Maintaining an IPM program in which the pest and its damage are managed economically and with minimum risk to the environment and human health is often not easily achieved. One goal of a mature IPM system is to establish equilibrium within the ecosystem, such that frequent chemical intervention is not required. This goal is rarely achieved, however, due to the dynamic nature of pests, horticultural practices, crop values, new pest control technologies, new regulations and the range of possible ecological landscape interactions.

A stable IPM system can also be upset by the introduction of an exotic pest. Opportunities for UC scientists to conduct research on the management of exotic pests in California are extremely limited due to regulated early response programs by federal and state agencies. However, once these invaders become established, they can be studied as part of the ecosystems they have invaded, and managed in an IPM systems framework. While virtually all of the pests for which UC IPM has guidelines are established in California, a significant number of them, 40% or more, are not endemic but were invaders that accompanied the movement of people, food or plant material into the state. Some of these pests, including sudden oak death, thousand cankers disease, giant reed, Sahara mustard, ash whitefly, sweetpotato whitefly biotype B, glassy-winged sharpshooter, olive fruit fly and spotted wing drosophila, were first detected and became problematic after 1980, within the

timeframe of UC IPM. Most of the invaders, however, have been established for a half century or more.

Development of IPM programs for key pests, whether endemic, invasive but long established, or more recently introduced, shares many commonalities. It requires an understanding of the pest's biology and interactions with the crop to develop an integrated approach that favors the crop over the pest. To be effective, program development must include the skills and knowledge of other UC researchers and the results must be distributed widely. This challenge is largely met through competitive grant funds, production of educational materials and demonstration of new practices in local fields and orchards.

### UC IPM overview

UC IPM was built upon the successful land-grant university research and extension model. Beginning in 1979, UCCE

## Publications provide a foundation of IPM practices

UC IPM's publications and website ([ipm.ucdavis.edu](http://ipm.ucdavis.edu)) have greatly contributed to the statewide adoption of IPM practices by growers, landscape professionals and gardeners. To date, there are 19 books in print, and the website has multiple layers of information, including pest management guidelines for 65 crops. Together, the materials present a foundation of IPM practices for California crops and urban settings. They are broadly cited in technical


### Pest Management Guidelines (PMGs), Pest Notes, Quick Tips and Pest Alerts.

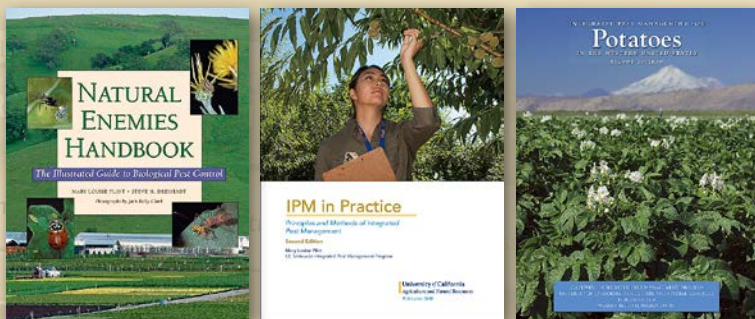
There are 47 PMGs representing 65 crops and crop groups available on the UC IPM website. The guidelines provide brief descriptions of a pest's biology; damage symptoms; and monitoring support; plus biological, cultural and chemical control practices; options for organic production; and illustrations for diagnostic purposes.

Since 2000, the Pest Notes have addressed pests in urban and landscape settings, with 166 Notes currently available. Pest Alerts are brief overviews that highlight new pests invading California. Quick Tips are based on Pest Note subjects but provide summaries for easy reference.

The PMGs and Pest Notes also contain information on pests of quarantine concern for exports, such as the oriental fruit moth for stone fruit exports to Mexico, and the Fuller rose beetle for citrus exports to Japan; IPM management options are included for these insects.

### Pest identification and monitoring cards.

Available for grapes, tree crops, vineyards and residential landscapes, these pocket-sized, laminated cards have high-quality photos and a brief description to aid in identifying pests. 



journals, agency reports and nontechnical articles, and the practices they detail have also been adopted by agricultural organizations and used in many self-assessment and certification programs (e.g., Lodi-Woodbridge Wine Grape Commission, San Joaquin Sustainable Farming Project). To see the full range of UC IPM products, visit [ipm.ucdavis.edu/IPMPROJECT/pubs.html](http://ipm.ucdavis.edu/IPMPROJECT/pubs.html).

**Manuals and books.** IPM manuals have been developed and updated for 17 crops, providing information on pest biology and nonchemical management of important pests, as well as other management tactics. Other reference books include *IPM in Practice* and *Handbook of Natural Enemies*.



professionals were hired to consolidate, test and deliver applied research findings on pest management to end-users. The program included a computer network to process and disseminate data and information required for effective IPM programs, and a designated writing staff to produce IPM manuals and other documents that contained practical information for growers and other pest management decision makers.

Until 2009, UC IPM sponsored a competitive research grants program, which generated new IPM knowledge and practices to address gaps in pest management systems and improve upon existing practices; it supported more than 450 IPM-related projects. By 1990, at the conclusion of UC IPM's first decade, 578 publications had been produced based on project-funded research. A 1989 survey indicated that 36% of the principal investigators found evidence of a reduction in pesticides as a result of UC IPM-funded research (Grieshop and Pence 1990).

Some of the notable successes from UC IPM-sponsored research involved then-recent invaders, for example, the ash whitefly. A pest of ornamental trees first detected in California in 1988, the ash whitefly became a considerable nuisance for homeowners and businesses because of the sticky film that covered everything underneath an infested tree. A competitive grant was provided to researchers



Jack Kelly Clark

initially worked with ANR scientists to develop models to predict potential damage (e.g., the alfalfa weevil model) based on pest and weather scenarios. To improve disease and insect forecasting in the field, statewide computing networks were introduced in the mid-1980s to incorporate data from California weather networks into the models. In recent years, web designers have created platforms for delivering IPM information and promoting IPM practices statewide, nationally and internationally.

To ensure IPM practices are adapted to local conditions, IPM advisors have

**Ash whitefly (*Siphoninus phillyreae*), a pest of ornamental trees, was first detected in California in 1988. UC IPM-sponsored research at UC Riverside resulted in a successful biological control program. Above, researcher Tom Bellows examining an ash tree for ash whitefly. Right, *Clitostethus arcuatus* adult (bottom), a natural predator of ash whitefly, and ash whitefly adult (top) on leaf, UC Davis Arboretum.**



Jack Kelly Clark

## In recent years, web designers have created platforms for delivering IPM information and promoting IPM practices statewide, nationally and internationally.

at UC Riverside to study its biology and support foreign exploration for biological control agents. The research resulted in a permanent and successful management program utilizing biological control as its primary management approach.

In the early 1980s, technical writers and editors developed comprehensive IPM manuals for 15 crops and created pragmatic and easily updatable guidelines for managing pests of specific crops. Other innovative publications followed, including additional books, Pest Notes, and pest identification and monitoring cards (see sidebar, page 154).

Computing and network technology have been critical to UC IPM's information delivery. Computer programmers

been distributed throughout the state to implement IPM through field research and extension. One of their key roles is to support local production farm advisors in integrating the latest pest management practices into local cropping systems. Equally important, IPM advisors communicate locally identified pest management needs to campus-based researchers.

### Foundation of UC IPM, 1980 to 1986

During the formative years of UC IPM, nine major crops were highlighted for IPM research and extension: alfalfa, almond, cereals, citrus, cotton, grapes, rice, tomatoes and walnuts. The selection of these crops was largely based on their value, acreage and pesticide use. Research

and extension projects were developed by cross-disciplinary teams of key scientists from all three UC campuses with colleges of agriculture (Davis, Berkeley and Riverside) and UCCE academics from critical counties in which the crops were produced. Each team focused their research efforts on the biology of the pests and natural enemies in the cropping system and the crop plant's development to better understand the relationship among those three key elements of pest population growth. Key research topics included improved decision making, better timing of pesticide intervention, increased understanding of crop and pest interactions, and alternative pest control approaches.

Crop modeling was emphasized to increase knowledge of environmental influences on pest and crop dynamics. A crop model is a mathematical simulation of the growth and development of a plant



## UC IPM increases its urban and community IPM footprint

The original focus of UC IPM's research and extension was on food and fiber crops. However, as California cities grew, pesticide use by structural pest control operators, landscape maintenance professionals, and home and garden nonprofessionals increased as well. Recognizing the impacts on human health and air and water quality, UC IPM began increasing resources to educate professionals and nonprofessionals alike.

Officially established in 2007, the Urban and Community IPM program codifies UC IPM's urban IPM resources and reinforces the program's commitment to this area. Issues currently addressed include reduction of pesticide use, protection of natural enemies of pests, impact on water quality from residential pesticide runoff in surface waters, IPM in structures and landscapes, and invasive pests. The UC IPM website ([ipm.ucanr.edu](http://ipm.ucanr.edu)) contains information on nearly 1,000 home and landscape pests, and other products



developed for urban audiences include videos and training materials for UC Master Gardeners, schools and retail nursery staff. To keep audiences current on IPM news, articles and updates are frequently posted on the Pests in the Urban Landscape blog ([ucanr.edu/blogs/UCIPMurbanpests/index.cfm](http://ucanr.edu/blogs/UCIPMurbanpests/index.cfm)) and in newsletters for retail nursery and garden center staff, landscapers and structural pest management professionals. 📺



Jack Kelly Clark

**A survey of IPM research grant awardees found that 38% of projects conducted between 1989 and 1999 focused on biological control. Farm Advisor Janet Caprile reaches into trellised apples to release *Aphidoletes* predatory midge for control of rosy apple aphid in Contra Costa County.**

(alfalfa, cotton, grape) that allows for stress caused by the environment and pests to be incorporated. Such models greatly aid the understanding of crop-pest interactions by predicting the impact pests can have on yield.

Sampling methodology was refined to be reliable, accurate and easy to use, including binomial sampling for spider mites in cotton and almonds and for caterpillar pests in tomato. The 1989 survey conducted by Grieshop and Pence revealed that 43% of growers and 43% of pest control advisers (PCAs) were using publications and information developed by UC IPM-sponsored programs. A complete listing of research and extension projects supported can be found in Pence (1990).

### New focus, new pests, 1986 to 2000

In 1986, UC IPM convened a meeting of stakeholders and UC IPM staff to review the program's research and extension focus. Stakeholders consisted of PCAs, growers, UC campus faculty from Berkeley, Davis and Riverside, and public agencies such as California Department of Pesticide Regulation and California Department of Food and Agriculture. In order to address pest issues in a wider set of crops, UC IPM's direction was changed from crop-based projects to practice-based projects. This change expanded the opportunity for studying pests and diseases on many more crops, and in noncrop sites, such as public spaces, natural areas and animal

agriculture. During this period, a wide variety of projects were funded on topics including improving pest monitoring and treatment decision support, increasing the understanding of relationships between pests, crops and natural enemies (applied field ecology) and improving the use of nonchemical management approaches such as biological control, cultural control and biorational use of biotic agents or chemicals.

A survey of IPM research grant awardees by Klonsky and Shouse (2000) reported almost three-quarters of the projects between 1989 and 1999 were directed toward reducing pesticide use and two-thirds of the projects were undertaken to improve the efficacy of pest control. Reflecting changes in the regulatory climate toward pesticides during the 1990s, 38% of the projects focused on biological control and use of indigenous natural enemies and 13% investigated microbial and botanical pesticides. Overall, 30% of the projects produced entirely nonchemical pest control procedures.

It was during this period that UC IPM began to address, through its competitive grants program and with local collaborations with PCAs and growers, the management of new pests that were affecting existing IPM programs. For example, the appearance of sweetpotato whitefly biotype B, a genetic variant of *Bemisia tabaci*, in the early 1990s, created a crisis for the production of cotton and other crops, such as melons, in the



Imperial Valley and the southern San Joaquin Valley. The existing management program was insufficient to prevent late-season outbreaks of the new biotype that resulted in unacceptable cotton lint covered in sticky honeydew. Research by scientists from UC Davis, UC Riverside and UCCE, partially funded by UC IPM, and extension of new management tactics coordinated by IPM advisors ultimately resulted in the whitefly's successful management. Other newly discovered invaders targeted by UC IPM funding included blue gum psyllid, Russian wheat aphid and giant whitefly on urban plants.

### Changing regulations, more new pests, 2000 to 2012

In 2001, UC IPM expanded its role in addressing exotic and invasive pests by collaborating with the Center for Invasive Species Research at UC Riverside through its UC Exotic/Invasive Pests and Diseases Research Program to establish a competitive grant program funded through the predecessor of USDA's National Institute for Food and Agriculture. From 2001 to 2009, 102 projects were supported that addressed specific exotic and invasive pests and diseases, including European grapevine moth, glassy-winged sharpshooter and Pierce's disease (see pages 125–141). Since the emergence of a new pest may result in pesticide applications that are detrimental to an established IPM system, the funded projects studied the effects of those applications, with results often leading to useful revisions of the UC IPM guidelines. Information on exotic and invasive pests, including management guidelines, is now disseminated in all UC IPM materials and at [ipm.ucdavis.edu/EXOTIC/index.html](http://ipm.ucdavis.edu/EXOTIC/index.html).

Concern about the impact of pesticides on humans and the environment intensified during the 1990s. For example, legislation such as the Food Quality Protection Act of 1996 resulted in increasingly stringent enforcement of environmental regulations. UC IPM worked closely with growers and regulatory agencies to identify and implement appropriate management strategies to decrease risks posed from pesticide use. Of particular note was the reduction of the use of organophosphate insecticides as dormant sprays in orchards during sensitive periods of rainfall and fog to protect surface water quality, air quality and wildlife, including

raptors. Since IPM is only one component necessary for environmental research on pesticide mitigations, a diverse group of scientific expertise needed to be assembled. UC IPM provided leadership in coordinating projects to identify and extend mitigation practices and alternatives to the high-risk organophosphate insecticides. The use of these insecticides subsequently declined dramatically across California orchard crops as growers turned to winter orchard floor management, reduced-risk insecticides and alternative treatment timings.

Publication of the Natural Resources Defense Council white paper "More IPM Please" (Hamerschlag 2007) focused public attention on the role of IPM in the conservation of natural resources. In response to the white paper, UC IPM began working closely with USDA Natural Resources and Conservation Service (NRCS) to develop an IPM training program for NRCS staff. The training emphasized IPM practices that mitigate the impacts of pest management activities on soil, water, air, plants and animals. This partnership with NRCS in California provided an opportunity to increase IPM adoption by linking activities to NRCS-recognized practices for cost sharing (Brewer and Goodell 2012). For example, as part of NRCS whole farm resource planning, the inclusion of an IPM plan was encouraged to identify potential mitigation activities related to pest management.

Now in its fourth decade, UC IPM continues to collaborate successfully with UC campuses, UC Cooperative Extension, commodity organizations and governmental agencies. It provides a platform for the organization, coordination and leadership needed for addressing pests and pest-related issues that threaten California crops, rangeland, public spaces and residential landscapes. [CA](#)

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### References

- Brewer MJ, Goodell PB. 2012. Approaches and Incentives to Implement Integrated Pest Management which Addresses Regional and Environmental Issues. *Ann Rev Entomol* 57:41–59.
- Grieshop JI, Pence RA. 1990. Research results: State-wide IPM's first 10 years. *Calif Agr* 44(5):24–6.
- Hamerschlag K. 2007. More Integrated Pest Management Please: How USDA Could Deliver Greater Environmental Benefits from Farm Bill Conservation Programs. Natural Resources Defense Council Issue Paper, Feb 2007. 23 p. [www.nrdc.org/health/pesticides/ipm/contents.asp](http://www.nrdc.org/health/pesticides/ipm/contents.asp).
- Klonsky K, Shouse B. 2000. IPM research profiled: 10-year trends. *Calif Agr* 54(6):20–1.
- Pence B. 1990. Bibliography of Research, 1980–1990. Publication 10. University of California Statewide Integrated Pest Management Project. 57 p.

**Over 40% of the pests for which UC IPM has guidelines were invaders that accompanied the movement of people, food or plant material into California. Bagrada bug (*Bagrada hilaris*), an invasive pest species native to Africa, attacks vegetable crops and ornamental plants. It was first found in Los Angeles County in 2008, and since then has spread north to 19 counties.**



# The cost of the glassy-winged sharpshooter to California grape, citrus and nursery producers

by Karen M. Jetter, Joseph G. Morse and John N. Kabashima

*In the late 1990s, widespread outbreaks of Pierce's disease in grapevines were linked to transmission via the glassy-winged sharpshooter (GWSS), threatening California's multibillion-dollar table, raisin and wine grape industries. Government agencies responded to the crisis by implementing two control programs to manage GWSS. We analyzed the long-term economic impact of these two programs on citrus, grape and nursery producers. The net economic effects on all citrus producers and on grape producers in the southern San Joaquin Valley were insignificant, while all grape producers in the Temecula Valley saw an average increase in annual production costs of about \$13.04 an acre. Based on our survey of nurseries in Southern California, approximately 11% had an infestation in 2008 and 2009, but only 3.0% in 2010. Average losses to nurseries per GWSS infestation were about \$12,238. Nursery producers also undertook a variety of pest control methods to prevent infestations and plant losses, and to meet quarantine regulations. Average annual per-acre costs of these approaches were \$2,975 for barrier methods to prevent GWSS from entering a premises, \$1,032 in pesticide controls and \$1,588 for in-house monitoring.*

In 1989, a pest new to California, the glassy-winged sharpshooter (GWSS), was collected in Irvine. Since then, it has been identified throughout Southern California and has spread into the southern San Joaquin Valley, including Kern County, parts of Fresno and southern Tulare counties, and to the coastal counties of Ventura, Santa Barbara and San Luis Obispo. The main commercial hosts for GWSS are citrus, grapes, almonds and alfalfa. GWSS overwinters in citrus, avocados, riparian vegetation and on several ornamental plants, such as crape myrtle.

As grapevines and almond trees leaf out in the spring and summer, GWSS moves onto those hosts. Feeding by GWSS generally does not result in economic losses, and it was initially treated as a harmless pest. However, in the early 1990s, widespread outbreaks of oleander leaf scorch in Southern California, followed by significant outbreaks of Pierce's disease (PD) in table, raisin and wine grapes in the Temecula Valley in



**Glassy-winged sharpshooter (*Homalodisca vitripennis*) caused widespread outbreaks of Pierce's disease in Southern California. Initially, before it was identified as a vector of the disease, it was treated as a harmless pest.**



**To contain the spread of PD, the California Department of Food and Agriculture placed a quarantine on the movement of plant material from GWSS-infested areas to GWSS-free areas within California. Above, discoloration of grape foliage caused by PD and sunburn on fruit in a Sonoma County vineyard.**

the late 1990s, were linked to transmission of the causal bacterium, *Xylella fastidiosa*, via GWSS.

At the height of the outbreak in the Temecula Valley, about 200 acres, or 10% of the total grape acreage in the Temecula Valley, was lost to PD (Siebert 2001). These outbreaks were followed in 2001 by widespread PD outbreaks in grapes in parts of the southern San Joaquin Valley, raising concerns for the health and economic viability of California's nursery industry and multibillion-dollar grape industry (USDA NASS 2011). Shortly thereafter, the California Department of Food and Agriculture (CDFA) changed GWSS's rating from a "C" rated pest (an organism not subject to state-enforced action except to provide for pest cleanliness in nurseries) to a "B" rated pest (an organism of known economic importance subject to eradication, containment, control or other holding action at the discretion of the individual county agricultural commissioner [CAC]).

To read full text of this peer-reviewed article, go to the current issue at <http://californiaagriculture.ucanr.edu>

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# 2014 index

The following peer-reviewed research articles, and news and editorial coverage, were published in *California Agriculture*, Volume 68, Numbers 1 to 4 (January–June, July–September, October–December), 2014. E = Article initially published online. Back issues are \$5 per copy, while supplies last. To subscribe to the journal, order back issues, search the archives or download PDFs of all research articles in full, go to:

<http://californiaagriculture.ucanr.edu>.



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## Research and Review Articles

### Economics and public policy

Jetter KM, Morse JG, Kabashima JN. The cost of the glassy-winged sharpshooter to California grape, citrus and nursery producers. 68(4):161–7. **E, EI**

Tumber KP, Alston JM, Fuller KB. Pierce's disease costs California \$104 million per year. 68(1,2):20–9.

SIDEBAR: Editors. Early conclusions on Pierce's disease. 68(1,2):21.

### Food and nutrition

Blackburn ML, Bruhn CM, Soederberg Miller L, Ganthavorn C, Ober B. Seniors, and their food handlers and caregivers, need food safety and nutrition education. 68(1,2):30–7.

## Collaborations

Adaska JM, Atwill ER, Nader GA. Diagnostics in animal health: How UC helps exclude and minimize impact of livestock pathogens. 68(4): 109–16. **EI**

SIDEBAR: Nader GA, Oliver MN, Finzel JA, Blanchard MT, Stott JL. Solving the puzzle of foothill abortion in beef cattle. 68(4):114–5. **EI**

Bostock RM, Thomas CS, Hoenisch RW, Golino DA, Vidalakis G. Plant health: How diagnostic networks and interagency partnerships protect plant systems from pests and pathogens. 68(4):117–24. **EI**

SIDEBAR: Bostock RM, Thomas CS, Hoenisch RW, Golino DA, Vidalakis G. A broad lab network means greater flexibility and better response. 68(4):121. **EI**

Bruening G, Kirkpatrick BC, Esser T, Webster RK. Cooperative efforts contained spread of Pierce's disease and found genetic resistance. 68(4):134–41. **EI**

SIDEBAR: Bruening G, Kirkpatrick BC, Esser T, Webster RK. Coordinated response to PD involves growers, scientists and government. 68(4):138–9. **EI**

Cooper M, Varela L, Smith R, Whitmer D, Simmons G, Lucchi A, Broadway R, Steinhauer R. Growers, scientists and

SIDEBAR: Editors. The Nutriture of People: From The Yearbook of Agriculture (USDA 1959). 68(1,2):33.

### Land, air and water sciences

Arnold BJ, Upadhyaya SK, Roach J, Kanannavar PS, Putnam DH. Water advance model and sensor system can reduce tail runoff in irrigated alfalfa fields. 68(3): 82–8. **WE**

SIDEBAR: Coats WJ. Placement of tensiometers as guides to irrigation practices. 68(3):87. **WE**

Platts BE, Grismer ME. Chloride levels increase after 13 years of recycled water use in the Salinas Valley. 68(3):68–74. **WE**

SIDEBAR: Coats WJ. 2014. Can irrigation with municipal wastewater conserve energy? 68(3):73.

Platts BE, Grismer ME. Rainfall leaching is critical for long-term use of recycled water in the Salinas Valley. 68(3):75–81. **WE**

SIDEBAR: Coats WJ. 2014. Improved leaching practices save water, reduce drainage problems. 68(3):77.

Weber E, Grattan SR, Hanson BR, Vivaldi GA, Meyer RD, Prichard TL, Schwankl LJ. Recycled water causes no salinity or toxicity issues in Napa vineyards. 68(3):59–67. **WE**

SIDEBAR: Coats WJ. Irrigation of deciduous orchards and vineyards influenced by plant-soil-water relationships in individual situations. 68(3):63.

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### Pest management

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## Special issue/section key

**EI** = Endemic and invasive pests and diseases

**UC** = UC Cooperative Extension centennial

**WE** = Water efficiency

**E** = Article published online

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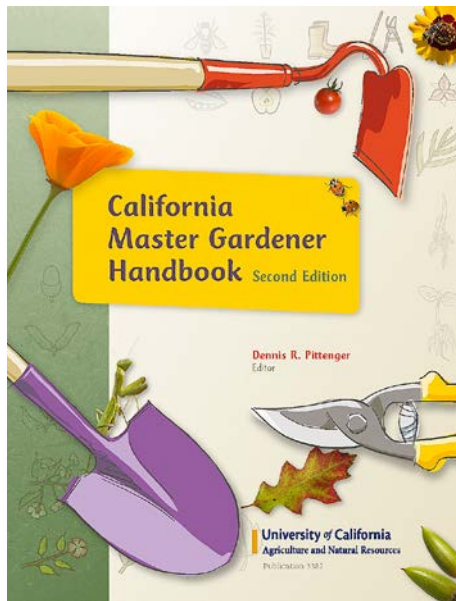
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# The cost of the glassy-winged sharpshooter to California grape, citrus and nursery producers

by Karen M. Jetter, Joseph G. Morse  
and John N. Kabashima

*In the late 1990s, widespread outbreaks of Pierce's disease in grapevines were linked to transmission via the glassy-winged sharpshooter (GWSS), threatening California's multibillion-dollar table, raisin and wine grape industries. Government agencies responded to the crisis by implementing two control programs to manage GWSS. We analyzed the long-term economic impact of these two programs on citrus, grape and nursery producers. The net economic effects on all citrus producers and on grape producers in the southern San Joaquin Valley were insignificant, while all grape producers in the Temecula Valley saw an average increase in annual production costs of about \$13.04 an acre. Based on our survey of nurseries in Southern California, approximately 11% had an infestation in 2008 and 2009, but only 3.0% in 2010. Average losses to nurseries per GWSS infestation were about \$12,238. Nursery producers also undertook a variety of pest control methods to prevent infestations and plant losses, and to meet quarantine regulations. Average annual per-acre costs of these approaches were \$2,975 for barrier methods to prevent GWSS from entering a premises, \$1,032 in pesticide controls and \$1,588 for in-house monitoring.*

In 1989, a pest new to California, the glassy-winged sharpshooter (GWSS), was collected in Irvine. Since then, it has been identified throughout Southern California and has spread into the southern San Joaquin Valley, including Kern County, parts of Fresno and southern Tulare counties, and to the coastal counties of Ventura, Santa Barbara and San Luis Obispo. The main commercial hosts



Glassy-winged sharpshooter (*Homalodisca vitripennis*) caused widespread outbreaks of Pierce's disease in Southern California. Initially, before it was identified as a vector of the disease, it was treated as a harmless pest.

for GWSS are citrus, grapes, almonds and alfalfa. GWSS overwinters in citrus, avocados, riparian vegetation and on several ornamental plants, such as crape myrtle. As grapevines and almond trees leaf out in the spring and summer, GWSS moves onto those hosts. Feeding by GWSS generally does not result in economic losses, and it was initially treated as a harmless pest. However, in the early 1990s, widespread outbreaks of oleander leaf scorch in Southern California, followed by significant outbreaks of Pierce's disease (PD) in table, raisin and wine grapes in the Temecula Valley in the late 1990s, were linked to transmission of the causal bacterium, *Xylella fastidiosa*, via GWSS.

At the height of the outbreak in the Temecula Valley, about 200 acres, or 10% of the total grape acreage in the Temecula Valley, was lost to PD (Siebert 2001). These outbreaks were followed in 2001 by widespread PD outbreaks in grapes in parts of the southern San Joaquin Valley, raising concerns for the health and economic viability of California's nursery industry and multibillion-dollar grape industry (USDA NASS 2011). Shortly thereafter, the California Department of Food and Agriculture (CDFA) changed GWSS's rating from a "C" rated pest (an organism not subject to state-enforced action except

to provide for pest cleanliness in nurseries) to a "B" rated pest (an organism of known economic importance subject to eradication, containment, control or other holding action at the discretion of the individual county agricultural commissioner [CAC]).

Federal and state agencies responded to the crisis by implementing control programs to manage and contain GWSS. In 2000, CDFA placed a quarantine on the movement of plant material from GWSS-infested areas to GWSS-free areas within California. Plant material can be fruit, twigs or leaves collected during fruit harvesting, or nursery stock. The quarantine required that all bulk citrus and grapes moved from an infested area to a clean one meet one of the following criteria: surveys show that (1) the grove or vineyard is free of GWSS, (2) fruit harvest was done in compliance with methods that will eliminate vectors or (3) postharvest treatments were completed to remove the vector (CDFA 2013b).

The CDFA quarantine for nurseries required that all plants transported from areas with known GWSS populations to

Online: <http://californiaagriculture.ucanr.edu/landingpage.cfm?article=ca.E.v068n04p161&fulltext=yes>  
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GWSS-free areas of the state be shipped under a Blue Tag protocol (Kabashima et al. 2008). This protocol required that the plant material be visually inspected by the CAC's office prior to shipment and be held for inspection by the destination county's agricultural commissioner's office. Counties enforcing the Blue Tag restriction were Alameda, Contra Costa, Fresno, Lake, Mendocino, Monterey, Napa, San Joaquin, Sonoma and Tulare. Under the Blue Tag protocol, if GWSS were detected in the load at the destination site, at the discretion of the CAC, it could be sprayed, rejected and/or reloaded on the truck to be transported out of the county back to the sender, or to another county not requiring the Blue Tag protocol (but only with the permission of the new destination county), or destroyed. Shippers and receivers who violated the quarantine's nursery stock regulations were also subject to fines. In 2010, 99.99% of the 50,600 shipments of nursery stock shipped under the Blue Tag protocol were free of any viable life stages of GWSS. Only six shipments were intercepted due to egg masses (four), nymphs (one) or adults (one).

In 2001, the U.S. Department of Agriculture Animal and Plant Health

Inspection Service (USDA APHIS) implemented an area-wide treatment program that involves the coordinated control of GWSS on citrus in infested areas where citrus is grown in proximity to grapes. In these areas, GWSS are monitored, and when they exceed treatment thresholds, GWSS populations are controlled before they can move into vineyards and transmit the PD bacterium. Any citrus grove located within one-quarter mile of a trapped grapevine (i.e., a trap that has captured a GWSS) is treated. The exception is a grove located along the northern boundaries of GWSS infestation in the San Joaquin Valley and the California coast. In this case, the barrier is one-half mile from a trapped vine. Areas with active area-wide programs include Kern County, the Temecula and Coachella valleys, and parts of Ventura, Fresno, Madera and Tulare counties (CDFA 2013a). While some citrus producers may benefit from the control of GWSS in their groves, it was thought chemical treatments might also disrupt integrated pest management control practices, imposing additional costs on the citrus industry. Currently, CDFA manages the federally funded area-wide program as part of its Pierce's Disease Control Program in coordination

with USDA APHIS, infested counties, and treatment coordinators.

In addition to the public programs instituted by USDA APHIS and CDFA, the grape producers and nurseries most affected by PD and GWSS quarantine undertook their own private measures to prevent the spread of the disease and vector. In 2001, grape producers assessed themselves a fee of \$3 per \$1,000 in revenues to fund research and other activities. As part of this effort, in 2002, the PD/GWSS Board funded a nursery treatment reimbursement program in Ventura County; Riverside County was added in 2008. Both counties' programs are currently active, with eight to 12 nurseries in Ventura County and one nursery in Riverside County receiving reimbursements twice a year. To receive reimbursement, the CAC must verify that the nursery is an active in-state shipper, has a GWSS compliance agreement and has a high enough GWSS population to justify treatment. In 2005, CDFA implemented the Nursery Stock Approved Treatment Program (NSATP), a 3-year pilot that evaluated whether nursery stock that was treated just before shipping, with insecticides that provided 100% control in research experiments (carbaryl or



Oleander is a GWSS host. The yellow, brown, dying leaf margins are a symptom of bacterial leaf scorch vectored by GWSS.



A yellow sticky trap monitors the GWSS population in an almond orchard. GWSS overwinters in citrus, avocados and riparian vegetation and moves into almond orchards and grape vineyards as they leaf out in spring.



fenpropathrin), would provide the same level of efficacy in real-world conditions. The NSATP pilot was 100% effective, with no viable life stages of GWSS detected during the 3 years of the project, which resulted in qualifying nurseries utilizing the NSATP protocol to ship to noninfested areas without an origin inspection.

Even with these control programs, grape producers and nurseries often needed to treat or otherwise manage GWSS, affecting crop production costs. How GWSS control affected private control costs depended on the actual pest treatment costs, plus any changes in cultural practices such as pruning, irrigation or nursery management. The net effect of private control costs, however, depended on how any additional control measures affected the frequency with which they substituted for or reduced current pest control treatments. Finally, private control costs for grape producers may be affected by increases in the rate of PD compared to the pre-infestation levels, even with GWSS control.

### Measuring the costs of GWSS control

To estimate the effects of CDFA and USDA APHIS control programs on producer management decisions and costs, data was needed on changes in cultural practices and the costs of those changes, the effect on other pest control decisions, and changes in nursery costs in infested regions. In November and December 2008, we held meetings in the southern San Joaquin Valley and the Temecula Valley with a UC Cooperative Extension farm advisor, six pest control advisers (PCAs) and producers, and two program managers to discuss the area-wide program and how the establishment of GWSS affected pest control materials applied, costs and nontarget pests. We obtained additional information on pest control practices and the current rates of PD in infested areas through phone interviews in 2009 and 2010 of 12 PCAs in Southern California and the southern San Joaquin Valley and UC Cooperative Extension county advisors. The costs estimated from these meetings were compared to UC Cooperative Extension budgets for grapes to determine how pest control treatments changed as a result of the treatments required for GWSS (Hashim-Buckley et al. 2007; Peacock et al. 2007a, 2007b; Vasquez et al. 2006, 2007). Costs and prices used



Applications of insecticides in late fall have avoided disrupting biological control and causing secondary pest outbreaks. This GWSS egg mass has been parasitized by a *Gonatocerus* species parasite.

in this analysis reflect those current for 2007–2008.

In 2011, to determine the economic effects of GWSS on the ornamental nursery industry, we sent an online survey to the 114 nursery producers in Southern California who ship under the Blue Tag protocol. The survey included questions on what type of nursery products they produced, whether they had ever had a GWSS outbreak, how outbreaks were treated and what preventative measures they took to prevent GWSS from entering their premises. The survey was pre-tested with select nursery operators in Southern California and final adjustments made. A total of 37 nursery producers responded.

Compared to nurseries that produce for the Southern California market, the nurseries that ship under the Blue Tag protocol need to be large and more diverse in order to supply the mass merchandiser box stores, retail nurseries and landscape installers. Thus, they have outdoor, shade house and greenhouse production at one location. Nurseries that do not ship to counties enforcing the Blue Tag protocol do not face the same regulations; consequently, they do not typically treat for GWSS. By sales, the nurseries that ship under the Blue Tag protocol represent about 40% to 50% of total sales by Southern California nurseries. The producers who responded to our survey have a product mix typical of those nurseries that ship under the Blue Tag protocol.

### GWSS control costs for citrus

The first line of defense against the spread of PD by GWSS is the USDA APHIS area-wide control program whereby citrus producers treat orchards during the fall and/or spring to prevent the buildup of GWSS populations. To control for GWSS in citrus, an application of acetamiprid is typically made in late fall, followed by an application of systemic imidacloprid in the spring. Imidacloprid is applied at a rate of 0.5 pounds of active ingredient (AI) per acre through the irrigation system. Treatments and the breadth of the area to be treated are determined based on monitoring and trapping results. Under the public program, citrus producers are reimbursed for the total cost of their GWSS treatments, including all materials, labor and application costs, and participation in the program is voluntary.

Total costs of production for citrus producers under the area-wide program may be affected if treating for GWSS allows producers to forego an existing treatment, as GWSS control may control other citrus pests. Alternatively, treatments for GWSS may cause secondary pest outbreaks of nontargeted pests. In conversations with the authors, local producers, PCAs and managers of the area-wide program reported that there have been neither substantial cost savings nor secondary pest outbreaks. One reason is due to timing: Late fall control of GWSS

in citrus typically occurs after beneficial insects have exerted control, and pesticide residues decline to negligible levels by the following year; thus, few secondary pest outbreaks are observed. Those who were interviewed felt that minor additional control of pests by spring treatments was balanced by the negative impacts of these treatments on beneficial species — thus, there was little net impact.

The CDFA quarantine program requires that fruit from infested areas be inspected and treated before leaving a quarantine area to be packed in a GWSS-free county. If GWSS are found in a producer's orange shipments, the producer bears the cost of treating GWSS in his or her grove if the producer did not participate in the area-wide program plus postharvest treatments. This aspect of the public control program is believed to encourage greater participation by citrus producers in helping to control GWSS. At the time of our meetings and interviews with PCAs, no citrus producers were incurring these costs in the San Joaquin and Temecula valleys.

#### GWSS control costs for grapes

**The southern San Joaquin Valley.** The second line of defense against the spread of PD is for individual grape producers to treat their grapevines for GWSS. Producers typically apply treatments to provide immediate control *in case* GWSS enters the field, not because it has been identified in their fields. This treatment

consists of one application of systemic imidacloprid annually, right before the first leaves appear in the spring. Applications of systemic imidacloprid are typically completed at the maximum rate of 0.5 pounds AI per acre through the irrigation system. The cost of applying imidacloprid was about \$50 to \$60 an acre in 2007. Since then, the cost of treating with imidacloprid has fallen due to competition from generic products.

Treatments for one pest often influence how another pest is treated. On grapes, GWSS treatments also control the variegated leafhopper and western grapeleaf skeletonizer. Treatments for these two pests were no longer typically needed, as the annual application of imidacloprid reduced these pest populations below economically damaging levels. We estimate that the cost savings by producers was \$62 an acre, or about the same amount as the costs to apply imidacloprid in the southern San Joaquin Valley in 2007–2008. Insecticides used to prevent GWSS establishment in vineyards also control the vine mealybug, which was first found in the Coachella Valley in 1994 and has since spread throughout several grape-growing counties in California, including Kern County in the southern San Joaquin Valley.

Grape producers may also suffer losses if the incidence of PD is higher than it was before GWSS became established. According to the PCAs we interviewed during August 2010, the incidence of PD

in the southern San Joaquin Valley is at about the same level, or slightly less, than it was before GWSS invaded. As a result, we estimate that producers in southern San Joaquin County had no additional costs due to changes in the incidence of PD in their area.

Finally, grape producers interviewed by the authors reported that they did not incur additional costs due to the quarantine on the movement of grapes. Grapes destined for the fresh market are hand-harvested and field packed, meeting the quarantine regulations that no plant material be transported with the fruit. The movement of bulk grapes did not require postharvest treatments, as bulk grape producers within an infested area can meet quarantine regulations through shipping grapes to processing centers within the infested areas, or by treating vineyards if surveys show the presence of GWSS.

**The Temecula Valley.** Private treatment of GWSS in grapes in the Temecula Valley also consists of an annual application of imidacloprid. With the imidacloprid treatments, producers in the Temecula Valley no longer needed to treat for the grape twig borer. As was the case in the San Joaquin Valley, the cost savings for treating secondary pests just about offset the additional costs of imidacloprid.

Because there is greater GWSS pressure in this region, however, some grape producers located near citrus groves in the Temecula Valley treat vineyards with one or two additional sprays of fenpropathrin in about 5% of the grape acreage per year. Fenpropathrin is applied at a rate of 11 ounces per acre, with the cost per ounce equal to \$1.62. With two treatments per year, the cost to treat with fenpropathrin was \$35.64. Custom application costs were an additional \$25 per acre per application. The average costs per acre to apply fenpropathrin, prorated to the 5% of acreage that was treated, were \$4.28 (table 1).

Furthermore, the Temecula Valley has a drier climate than the San Joaquin Valley. In order for producers to apply imidacloprid when it can do the most good, a separate irrigation may be required. We held meetings with farm managers and PCAs, who estimated that half the time they need to complete a separate irrigation in order to apply imidacloprid. The additional irrigation was estimated at 2 gallons of water applied per vine per

TABLE 1. Added average annual costs per acre to treat GWSS in the Temecula Valley, 2009

	Amount
	<i>\$ per acre</i>
<b>Cost of fenpropathrin</b>	
Insecticide applied at 11 ounces per acre two times per year at \$1.62 per ounce	35.64
Application by a custom applicator at \$25 per acre	50.00
Total cost if fenpropathrin applied	85.64
<b>Cost of extra irrigation</b>	
Water applied on 850 vines per acre × 2 gallons per vine × 6 hours of irrigation at a cost of \$400 per acre-foot	12.52
Labor per acre	5.00
Total cost if an additional irrigation needed	17.52
<b>Costs prorated</b>	
Irrigation prorated to 50%	8.76
Fenpropathrin prorated to 5%	4.28
<b>Total additional costs</b>	<b>13.04</b>



hour over 6 hours (with an average of 850 vines per acre). At a cost of \$400 per acre-foot during the time period covered by this study, the total additional cost to irrigate was estimated to be \$17.52 per acre (table 1). Prorated to 50% of the time an additional irrigation was needed, the additional cost was estimated to be \$8.76 per acre. We estimated that the total cost of the extra fenpropathrin on 5% of the acreage and extra irrigation on 50% of the acreage was \$13.04 per acre per year (table 1).

Grape producers in the Temecula Valley had a slightly higher incidence of PD in 2007, at 2% to 3%, up from about 1% before the establishment of GWSS. While there were a few large plots that were infested, PD in grape vineyards tended to be localized. Producers typically pulled vines and replanted instead of removing a whole plot; based on UC Cooperative Extension wine grape budgets for the San Joaquin Valley, the cost to replant about 2% of vines a year was \$65 per acre (Hashim-Buckley et al. 2007; Peacock et al. 2007a, 2007b; Vasquez et al. 2006, 2007). We estimate that the total cost of GWSS and PD management was about \$78 an acre per year in the Temecula Valley.

There were no market effects for grape producers and consumers from GWSS establishment in California. The success of the GWSS area-wide control program caused few changes in the costs of production for producers in the southern San Joaquin Valley, where much of California's table grape and raisin production occurs. Additional costs to treat GWSS were offset by cost savings on other pests. Although producers in the Temecula Valley incurred higher costs of production, with about 1,300 acres planted in wine grapes, their share of total grape production in California was less than 3%, a share too small to influence markets significantly. As a result, the higher costs of production were not offset by increases in market prices, and the net change in producer revenues was equal to the changes in the costs of production. Given that only producers in the Temecula Valley incurred higher costs of production, we estimate the total increase in the annual direct costs to the entire grape industry in California to be about \$103,000 per year.

#### **Additional costs to grape producers.**

Grape producers of wine, wine vineyards



Jack Kelly Clark

**To contain the spread of PD, the California Department of Food and Agriculture placed a quarantine on the movement of plant material from GWSS-infested areas to GWSS-free areas within California. Above, discoloration of grape foliage caused by PD and sunburn on fruit in a Sonoma County vineyard.**

and juice concentrates pay an assessment to the PD/GWSS board (PD/GWSS Research Board 2013). In 2001, the assessment was \$3 per \$1,000 in farm revenues. By 2008 and 2009, the rate had fallen to \$1 per \$1,000 in farm revenues; in 2010, it fell to \$0.75. As of 2009, this assessment had raised a total of \$34 million (PD/GWSS Research Board 2013). While cumulatively the assessment represents a significant funding source to address the GWSS problem, the assessment rate represents less than 0.5% of revenues, and again there are no market effects to producers and consumers due to the cost of the assessment.

#### **GWSS control costs for nurseries**

Out of the 37 nursery operators who responded to our survey, eight operations had to destroy plants due to the presence of GWSS in 2008 and 2009 (table 2).

Four nurseries (or 11%) of the respondents destroyed plants in 2009, the year with the greatest number of nurseries

that had to destroy plants. In contrast, only one nursery had to destroy plants in 2010. The total cost of the destroyed plants, including wholesale value of lost plants, labor and materials, was \$97,899, for an average loss of \$12,238 per infested operation (table 2). However, costs varied widely by nursery from a low of \$1,500 in 2010 to a high of \$35,000 for plants destroyed in 2008 (table 2). The plants that were destroyed included broadleaf evergreens, deciduous flowering trees, deciduous shade trees, perennial herbaceous bedding plants, ornamental shrubs and annual bedding plants. Annual bedding plants were the plants listed most often by the respondents as the type of plant that needed to be destroyed either in the nursery or at the destination.

Over 50% of the nursery operators who responded ( $n = 20$ ) to the question on GWSS management applied pesticides to manage GWSS (table 3). GWSS control occurred both during plant growth and as a preshipment treatment.

**TABLE 2. Nursery costs due to GWSS infestations by place and year, 2008–2010**

		Respondents ..... number .....	Total observations	Costs		
				Average	Minimum	Maximum
				..... \$ per infestation .....		
<b>2010</b>						
In nursery	Were plants destroyed?	37	1			
	Value of plants destroyed	37		1,500	1,500	1,500
	Labor and material costs	37		0	0	0
	Total average costs	37		1,500		
At point of sale	Were plants destroyed?	37	1			
	Value of plants destroyed	37		0	0	0
	Labor and material costs	37		0	0	0
	Total average costs	37		0	0	0
Both	Were plants destroyed?	37	1			
	Value of plants destroyed	37		1,500	1,500	1,500
	Labor and material costs	37		0	0	0
	Total average costs	37		1,500		
<b>2009</b>						
In nursery	Were plants destroyed?	37	4			
	Value of plants destroyed	37		7,500	0	15,000
	Labor and material costs	37		1,950	0	5,500
	Total average costs	0		9,450		
At point of sale	Were plants destroyed?	37	4			
	Value of plants destroyed	37		2,500	0	5,000
	Labor and material costs	37		125	0	500
	Total average costs	37		2,625		
Both	Were plants destroyed?	37	4			
	Value of plants destroyed	37		10,000	0	20,000
	Labor and material costs	37		2,075	0	6,000
	Total average costs	37		12,075		
<b>2008</b>						
In nursery	Were plants destroyed?	37	3			
	Value of plants destroyed	37		13,000	0	35,000
	Labor and material costs	37		1,200	0	3,600
	Total average costs	37		14,200		
At point of sale	Were plants destroyed?	37	3			
	Value of plants destroyed	37		1,667	0	5,000
	Labor and material costs	37		167	0	500
	Total average costs	37		1,833		
Both	Were plants destroyed?	37	3			
	Value of plants destroyed	37		14,667	0	40,000
	Labor and material costs	37		1,367	0	4,100
	Total average costs	37		16,033		
<b>Average over all unique infestations</b>				<b>12,238</b>		

For both types of control, nursery producers used a variety of chemical treatments, including fenpropathrin, carbaryl, acetamiprid and abamectin. The average cost to treat nursery stock on the premises was \$1,032 an acre. Costs again varied widely with a minimum cost of \$175 an acre and a maximum of \$2,159. The pre-shipment treatments were a lower cost per acre. The average cost was \$229 per acre, with a minimum cost of \$39 per acre and a maximum cost of \$850 (table 3).

Our survey also asked questions on other methods used to prevent GWSS infestations and plant loss, including barrier methods to prevent the entry of GWSS (or otherwise manage GWSS) and the inspections of traps installed by CDFR. Almost 30% of the operators who responded to the question about methods ( $n = 22$ ) used some type of barrier method. The methods used were shade cloths (three), an insect screen (one), sticky traps (one), oleander hedge (one) or a combination of all methods (one). Because oleanders are a GWSS host, it is probable that the nurseries who use oleander hedges are using them as a “trapping” hedge to monitor and treat for GWSS. With a diversity in methods, there was a diversity in costs. Costs ranged from \$750 to \$4,660 an acre, with an average of \$2,975 (table 3). Both the minimum cost and maximum cost for the barrier methods were for shade cloths. Most of the barrier methods used provided additional protection against other pests; for example, insect screens protect against aphids and thrips.

Finally, nursery operators were asked about in-house monitoring of GWSS traps installed by CDFR. About half of the operators who responded to this question ( $n = 22$ ) did some in-house monitoring. Monitoring varied though, from as frequently as once a week to as little as every other month. The average cost per acre to monitor was \$1,588. The minimum cost incurred by a nursery was \$24 per acre, and the highest was \$7,680 (table 3). Among the nurseries who responded yes to this question, the two with the highest cost per acre to monitor traps also produced the greatest diversity of plants. The number of different plant categories produced by these nurseries was twice that of the nursery with the next highest number of plant categories. The nursery that produced plants from only one category also



had the lowest cost per acre to monitor GWSS traps.

### Impacts on producers

The establishment of GWSS and spread of PD fundamentally changed commercial agricultural practices for several table, raisin and wine grape producers, as they needed to carefully monitor fields for both the pest and disease to prevent vine death. With low GWSS populations, preventive measures undertaken by producers caused a negligible net change in the profitability of producing grapes.

Nursery producers experienced higher costs in some cases. Recurring costs were incurred for monitoring and preventing the entry of GWSS, and certain nurseries needed to treat when infestations were found. Nursery producers also experienced increased costs due to paperwork and coordination of treatments, inspections and notifications required by the Blue Tag and NSATP protocols (table 4).

The largest impact of GWSS on agricultural producers occurred in relation

to the area-wide management programs (San Joaquin Valley, Temecula, Ventura and Coachella). These programs allowed the coordinated treatment of a grape pest by citrus producers in order to prevent the spread of a deadly grape disease. As a result, the net effects in changes in pest treatments by commercial grape producers were negligible.

The area-wide programs have not been cost-free, however. These programs are managed and coordinated by the USDA and CDFA out of public monies. The cost of the GWSS area-wide treatment program was greater than \$20 million a year in 2010. In comparison, the average farm-gate value in 2010 of the grape and nursery commodities produced in California was \$3.2 billion for all grapes and \$1 billion for floriculture production (USDA NASS 2011). While the benefits of the area-wide program are beyond the scope of this analysis, had the program not effectively reduced GWSS populations to current levels, much of the state's multibillion-dollar grape industry would

have been at risk for the spread of GWSS and Pierce's disease. [CA](#)

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**TABLE 3. Select average annual nursery costs per acre for GWSS management, 2008–2010**

	Observations ..... number .....	Yes responses	Costs		
			Average	Minimum	Maximum
			..... \$ per acre .....		
Treat GWSS on premises	20	11			
<i>Total costs</i>	20	8	1,032	175	2,159
Treat for GWSS preshipping	15	9			
<i>Total costs</i>	15	6	229	39	850
Use barrier methods	22	7			
<i>Total costs</i>	22	4	2,975	750	4,660
Have traps used by inspectors	22	18			
Do in-house monitoring of traps	22	10			
<i>Total costs</i>	22	9	1,588	24	7,680

Average number of traps used, three; minimum, one; maximum, six.

**TABLE 4. Select average annual costs per nursery operation to meet regulatory requirements, 2008–2010**

	Observations ..... number .....	Yes responses	Costs		
			Average	Minimum	Maximum
			..... \$ per acre .....		
Do own inspections of traps	9	7	111	12	250
Number of inspections per year	9	7	62	2	244
<i>Total costs</i>	9	7	5,444	24	16,800
Staff training costs	9	7	1,605	15	10,000
Time spent on paperwork per year	9	6	57	4	150
Cost of paperwork	9	6	1,433	50	5,000

### References

- [CDFA] California Department of Food and Agriculture. 2013a. Pierce's Disease Control Program Maps. Sacramento, CA. [www.cdca.ca.gov/pdcp/map\\_Index.html](http://www.cdca.ca.gov/pdcp/map_Index.html) (accessed July 2013).
- CDFA. 2013b. Pierce's Disease Control Program. Plant Quarantine Manual. Sacramento, CA. <http://pi.cdca.ca.gov/pqm/manual/pdf/454.pdf> (accessed July 2013).
- Hashim-Buckley JM, Peacock WL, Vasquez SJ, et al. 2007. Sample Costs to Establish and Produce Table Grapes: Redglobe in the San Joaquin Valley South. Number GR-VS-07-3. Davis, CA: University of California Cooperative Extension. 22 p.
- Kabashima J, Gutman G, Redak R, et al. 2008. Glassy-winged sharpshooter nursery approved treatment best management practices. December 2008. In: Kabashima J, Morris G (eds.). Glassy-Winged Sharpshooter Nursery Stock Approved Treatment Manual. 20 p. [www.cdca.ca.gov/pdcp/guidelines.html#Nursery](http://www.cdca.ca.gov/pdcp/guidelines.html#Nursery).
- PD/GWSS Research Board. 2013. Area-wide FAQs. [pdgwss.net/background/area-wide-faqs/](http://pdgwss.net/background/area-wide-faqs/) (accessed November 2012).
- Peacock WL, Vasquez SJ, Hashim-Buckley JM, et al. 2007a. Sample Costs to Establish and Produce Table Grapes: Crimson Seedless in the San Joaquin Valley South. Number GR-VS-07-2. Davis, CA: University of California Cooperative Extension. 22 p. <http://coststudies.ucdavis.edu/current.php>.
- Peacock WL, Vasquez SJ, Hashim-Buckley JM, et al. 2007b. Sample Costs to Establish and Produce Table Grapes: Flame Seedless in the San Joaquin Valley South. Number GR-VS-07-4. Davis, CA: University of California Cooperative Extension. 22 p. <http://coststudies.ucdavis.edu/current.php>.
- Siebert J. 2001. Economic impact of Pierce's disease on the California grape industry. In: Proceedings of the Pierce's Disease Research Symposium. Sacramento, CA: CDFA. p 111–6. [cdca.ca.gov/pdcp/research.html](http://www.cdca.ca.gov/pdcp/research.html).
- USDA NASS. 2011. Agricultural Statistics, 2011. Washington, DC: United States Government Printing Office.
- Vasquez SJ, Fidelibus MW, Christensen LP, et al. 2006. Sample Costs to Produce Grapes for Raisins: Continuous Tray Dried Raisins in the San Joaquin Valley. Number GR-SJ-06-2. Davis, CA: University of California Cooperative Extension. 22 p. <http://coststudies.ucdavis.edu/current.php>.
- Vasquez SJ, Hashim-Buckley JM, Peacock WL, et al. 2007. Sample Costs to Establish and Produce Table Grapes: Thompson Seedless in the San Joaquin Valley South. Number GR-VS-07-1. Davis, CA: University of California Cooperative Extension. 22 p. <http://coststudies.ucdavis.edu/current.php>.