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Advancing Biopesticide Technologies for Managing Pierce's Disease

Of the many threats to grapevines, Pierce's disease (PD) stands out as an especially formidable foe. Growers who have encountered PD don't need to be reminded of its perils, they have devastating losses to show for it. It's time PD met its match.



Dr. Anika Kinkhabwala administers Xylphi-PD, one of the biological treatments the research team is evaluating to control Pierce's disease.

A team of researchers, led by Dr. Akif Eskalen at University of California, Davis, has been diligently testing eight combinations of four biological treatments for controlling *Xylella fastidiosa* (*Xf*), the bacterial pathogen that causes PD, in the field. Less than one year into a three-year study, they observed promising results and hosted a field day to share them.

"I'm really happy that the project got funded by the Pierce's Disease and Glassy-Winged Sharpshooter Board because we can compare treatments of all the biopesticides," said research collaborator Dr. Philippe Rolshausen. "This is a major step forward."

In September 2022, a crowd gathered at the research site, UC Davis' 11-year-old Cabernet Franc vineyard, which, crucially, has no history of PD. Eskalen explained that the team has spent the past several months observing symptoms in

controls versus eight treatments of experimental biopesticides along, and in combination, with a bacteriophage. The results will offer valuable insight into which methods growers could use and how they should use them.

All treatments were administered via a Xyleject, an application device used to inject the product directly into the grapevine, and grapevines were artificially inoculated with *Xf* one week later. The team followed up with a second round of treatments the following week and closely observed the vineyard for symptoms of PD.

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PD/GWSS Board Strengthening its Grapevine Virus Research Strategy "The inoculated control grapevines at the field site showed traditional PD symptoms, including leaf scorching and poor cane lignification. Comparatively, there was a clear reduction of symptoms in treated vines," said Dr. Kristin Lowe, PD/GWSS Board research coordinator. "The promise of getting these tools into growers' hands is very exciting."

Dr. Anika Kinkhabwala introduced the first control method: a sophisticated cocktail of bacterium-killing viruses (bacteriophages) named Xylphi-PD. Unlike antibiotics that obliterable the plant's microbiome, Xylphi-PD is a precise tool that targets and kills only Xf. Certified Organic and non-GMO, the product is currently the only commercially available control method.

Dr. Steven Lindow introduced an equally sophisticated control method: the biological control agent *Paraburkholderia phytofirmans*. This naturally occurring organism grows alongside *Xf*, spreading quickly through the vine, but unlike *Xf*, it's recognized by the plant as a threat and triggers a defensive reaction. Though a preventive application is recommended, the team found in prior studies that this method can even be somewhat curative, successfully treating vines when applied up to

five weeks after *Xf* was introduced. Substantial levels of disease control have been observed in treated plants.

Two additional experimental biological control agents are being evaluated in the trial, both alone and in combination with XylPhi-PD. Preliminary results suggest all the control methods provide some reduction in PD symptoms, but the greatest reductions were seen when the combination treatments were used. Collaboration among various entities is making this breakthrough in PD research possible.

Though this experiment is far from complete, it offers encouraging results even in its early stages. "This is not the final version of our experiment. We are going to be repeating this experiment for two more years," said Eskalen. "And after that, we are hoping that we will be able to provide some concrete options to our growers to manage, or even prevent or control PD."

The field trial presentation is available online for free CEUs at bit.ly/3wj2JKz. There will be another field day in fall 2023 to assess the next round of results, and researchers are hopeful that it won't be long before they can get a proven, potent product to control PD into growers' hands.

Meet the New Pierce's Disease Control Program Statewide Coordinator



New Pierce's Disease Statewide Coordinator Joseph Damiano brings a unique perspective as a winegrape grower with a wide range of experience in California's wine and agricultural industries.

Joseph Damiano joined the Pierce's Disease Control Program as the new statewide coordinator in November 2022, bringing a unique perspective to the program with a wide range of experience in the public and private sectors in the state's wine and agricultural industries.

Damiano is a California native, grew up on a small family farm in the Sierra Foothills, and now manages and operates a 6-acre family vineyard in the Sierra Foothills in addition to his new role at the California Department of Food and Agriculture.

"As a California winegrape grower, I understand the devastating effect that pests and diseases can have on agriculture. Having a perspective of both industry and government and being able to engage in meaningful discussions with the PD/GWSS Board and other stakeholders will help move the program forward in a positive manner," said Damiano.

Damiano has a degree in Plant Science-Viticulture from the University of California, Davis, and extensive knowledge of the wine and agriculture industries, with previous positions at Gallo Winery, Kendall Jackson Winery, Nevada County Department of Agriculture, Colusa County Department of Agriculture, and California Department of Pesticide Regulation.



Red arrows point to the death of canes inoculated with *Xylella fastidiosa* in 2021.

Transgenic Rootstock-Mediated Protection of Grapevine Scion Against Pierce's Disease by Dual Stacked DNA Constructs

Project leaders: David Gilchrist and James Lincoln, University of California, Davis

The goal of this field research is to determine if pairs of five genes transformed into adapted rootstocks will act across a graft union to suppress Pierce's disease symptoms in non-transgenic Chardonnay scions. This year, control vines and the most impacted treatments showed poor bud break at the point of *Xylella fastidiosa* inoculation. Following another round of inoculations this spring, data collection over the summer included ratings of percent leaf scorch, vine growth, and other visual symptoms and tracking the concentration and movement of the pathogen across tissues. Learn more at bit.ly/3oyv91d.

Ecology of Grapevine Red Blotch Virus

Project leaders: Marc Fuchs and Keith Perry, Cornell University

Greenhouse studies showed that the threecornered alfalfa hopper (TCAH) can transmit grapevine red blotch virus (GRBV) and the team has now confirmed field transmission in vineyards in New York and California. To better understand potential vector dynamics, the team is investigating alternate food sources and overwintering plant hosts. Analysis of the gut content of insects caught in vineyards show they have a wide and varied diet. The team also learned that wild, "free-living" grapevines in riparian areas around vineyards can host both GRBV and TCAH over the winter. Learn more at bit.ly/3Bicsmc.



The threecornered alfalfa hopper is a primary vector of grapevine red blotch virus.



Protoplast-derived Thompson Seedless somatic embryos germinating after transfection with a plasmid containing a plant codon-optimized Cas9 gene.

Protoplast-Mediated Gene Editing for Disease Resistance

Project leaders: David Tricoli and Juan Debernardi, University of California, Davis

The team is investigating CRISPR-Cas9, a gene editing technology that allows for precise changes in a plant's genetic code by treating protoplasts with DNA or ribonucleoproteins (RNPs) prior to reforming whole plants. They are testing this method to create disease-resistant grapes by editing a gene to make the plants more resistant to powdery mildew. This proof-of-concept study will lay the groundwork for creating grapes with other disease-resistant traits using protoplast-mediated gene editing. Learn more at bit.ly/3rKaNCN.

PD/GWSS Board Strengthening its Grapevine Virus Research Strategy

To ensure growers' dollars continue to be invested wisely in research to find solutions to winegrape pests and diseases, the Pierce's Disease and Glassy-Winged Sharpshooter Board will conduct a comprehensive review of its grapevine viruses research program with the National Academy of Sciences.

"This review will help the Board continue to focus growers' dollars where it counts, zeroing in on the most promising research and opportunities to advance progress on combatting grapevine viruses," said Board Chair Will Drayton.

The committee will review current knowledge on grapevine red blotch virus and grapevine leafroll-associated virus type 3, research outcomes and gaps, and approaches for future research. They will also assess the Board's current virus research projects, proposals for new funding, and the Board's process for reviewing and selecting research proposals to fund. The final report will guide the Board's strategy for research funding.

"The National Academy of Sciences brings a valuable outside perspective to our program, and will help us identify grapevine virus knowledge gaps, opportunities for collaboration, and new technologies and techniques," said Dr. Kristin Lowe, Board research coordinator.

The Board conducted a similar review with the National Academy of Sciences focused on PD and GWSS in 2004, which paved the way for many of the PD research successes over the past two decades. "California Agricultural Research Priorities — Pierce's Disease" is available online at <a href="https://bitsubsciences.org/b

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