Research Updates

Prophage diversity of “Candidatus Liberibacter asiaticus” strains in California

**Submitted to:** Phytopathology


Huanglongbing (HLB) is a deadly citrus disease and is associated with a non-culturable bacterium “Candidatus Liberibacter asiaticus” (CLas). HLB has reduced Florida citrus production by two-thirds and more than doubled production costs. The first CLas detection in California occurred in 2012 in an urban garden in Hacienda Heights (Los Angeles County), and CLas has now been detected in multiple urban locations in Southern California. Knowledge of genetic diversity of California CLas strains is important to ascertain pathways of entry as well as HLB management. In this study, ten Southern California CLas strains from six urban locations were analyzed for genomic diversity using next generation sequencing. Prophage sequences, the most variable part of the genome, were analyzed. Four prophage typing groups (PTGs) were identified and profiled. Results suggest multiple exotic introductions, possibly from Asia, have occurred in California. This study provides baseline information for further HLB epidemiology research and management in California.

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Fungicide resistance in Botrytis cinerea populations in California and its influence on control of gray mold on stored mandarin fruit

**Submitted to:** Plant Disease

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In recent years, the acreage of mandarin citrus fruit has increased significantly in California in response to consumer demand for "easy-to-peel" citrus fruit, and storing mandarin fruit in cold facilities before packing has become a common practice to retain fruit quality and extend marketing opportunities. Extended storage, however, can increase the risk for development of postharvest fruit rots. Gray mold caused by the fungus *Botrytis cinerea* is an emerging fruit rot disease affecting stored mandarin fruit in California. Control of *B. cinerea* has largely been dependent on the use of fungicides. However, *B. cinerea* is a high risk fungal pathogen for the development of resistance to certain groups of fungicides. Fungicide resistance in the pathogen often results in the failure of disease control. In this study, we tested resistance of *B. cinerea* isolates obtained from mandarin fruit to four citrus postharvest fungicides. We found that 83-98% of the isolates were resistant to azoxystrobin, 71-93% of the isolates were resistant to pyrimethanil, 63-68% of the isolates were resistant to thiabendazole, and no fludioxonil resistance was detected in a 2-year survey. Of the 200 *B. cinerea* isolates tested, 5, 24, and 62% were resistant to 1, 2, or 3 classes of fungicides, respectively. Inoculation tests were conducted to evaluate if the fungicides at label rates control various resistant phenotypes on fruit. Most fungicides failed to control gray mold on mandarin fruit inoculated with respective fungicide-resistant phenotypes. Our
results suggest that alternative control methods need to be integrated into existing decay control programs to target this emerging disease on mandarin fruit.

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**Physiological response of ‘Fuji’ apples to irradiation and the effect on quality**

**Submitted to:** LWT - Food Science and Technology

**Authors:** N. Kheshti, A. Melo, A. Bacquero, D. Obenland, A. Prakash

Irradiation is approved for quarantine treatment of apples exported from California to Mexico but the effects of treatment on apple quality had previously not been thoroughly described. ‘Fuji’ apples were irradiated at 377 or 1148 Gy, and then stored for 7 days at 1°C to simulate ground transportation to Mexico plus a further 7 days at ambient temperature to mimic retail and commercial storage. Irradiation suppressed ethylene production but increased the rate of respiration. The concentration of sugars was unaffected while that of acids decreased. Flavor volatiles were relatively unaffected at 377 Gy but were diminished in concentration by the 1148 Gy treatment. Higher electrolyte leakage in treated apples indicated that some damage occurred due to irradiation but firmness was the only major quality factor impacted, although the loss of firmness was far less at 377 Gy than 1148 Gy. An irradiation treatment of 377 Gy is sufficient for quarantine purposes and, given its minimal impact on apple quality, can be a viable alternative to conventional phytosanitary treatments for ‘Fuji’ apples. Since prior treatments either take too long or cause damage this helps enable a new export market for this variety.

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**Grapevine phenolic compounds bind Xylella fastidiosa lipopolysaccharide and influence cell surface adhesion**

**Submitted to:** Journal of Bacteriology

**Authors:** S. Lee, L. Burbank, C. Wallis, E. Rogers

Many chemical compounds produced by plants are known to influence growth and colonization of bacterial pathogens, either because of antimicrobial activity, or by acting as signals that trigger a pathogen response. *Xylella fastidiosa* colonizes the xylem tissue of grapevines, an environment that is generally low in nutrients but contains a range of plant-produced compounds classified as phenolics. Specific phenolic compounds found in grapevines were tested for influence on growth and cell-surface attachment of *X. fastidiosa*. Several of these compounds inhibited adhesion of *X. fastidiosa* cells to a solid surface during extended growth, likely due to binding of the compounds to molecules on the outside of the bacterial cell. Identifying plant-produced compounds that can inhibit effective colonization of *X. fastidiosa* in grapevine will improve the ability to identify and screen for novel sources of host resistance.

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**Detection of a single-copied plasmid, pXFSL21, from Xylella fastidiosa strain Stag’s Leap with two toxin-antitoxin systems using a next generation sequencing approach**

**Submitted to:** Phytopathology

**Authors:** C. Van Horn, F. Wu, Z. Zheng, Z. Dai, J. Chen

*Xylella fastidiosa* is a bacterial pathogen causing Pierce’s Disease (PD) of grapevine resulting in significant economic loss in California. Strain Stag’s Leap of the pathogen was originally isolated from Napa Valley, California, and is highly virulent. Little is known about the virulence mechanism(s) of this bacterium, but genome sequence analyses could provide initial clues. In this study, DNA samples of strain Stag’s Leap were extracted from a pure culture and a PD-symptomatic grapevine in the greenhouse. Next generation sequencing (NGS) and subsequent computational analyses were performed. A circular plasmid was found and designated as pXFSL21. The plasmid existed as a single copy per bacterial cell and encoded 27 genes, including four genes forming two toxin-antitoxin (T-A) systems. The double T-A systems may facilitate maintenance of the single-copy plasmid in the bacterial population.

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Substrate-mediated feeding and egg-laying by spotted wing drosophila: waveform recognition and quantification via electropenetrography

Authors: R. Guedes, F. Cervantes, E. Backus, S. Walse

Substrate suitability is paramount for arthropods in general and agricultural pest species in particular. Therefore, we electronically monitored the interaction between adults of the spotted wing drosophila with two substrates, artificial diet and strawberry fruits, using electropenetrography (EPG). The objectives were 1) to assess the suitability of EPG for monitoring substrate-mediated activities of spotted wing drosophila; 2) to develop methods for recognizing the waveforms descriptive of the main behaviors observed, and 3) to determine the qualitative and quantitative waveform differences in both substrates. Three substrate-associated phases were recognized in adults: non-probing, feeding, and egg-laying. Dabbing and ingestion constituted the feeding phase, while abdominal probing and egg-laying constituted the egg-laying phase. The egg-laying phase was similar on diet and strawberry. In contrast, non-probing events were more frequent, but shorter, leading to less overall non-probing on diet compared with strawberry. Dabbing was more frequent and lasted longer overall on diet, but ingestion events lasted longer on strawberry. Therefore, although the flies fed (dabbed and ingested) for longer overall on diet, each ingestion event was longer on strawberries. Our results suggest that strawberry fruits are a more suitable and preferred food source because they led to extended periods of sustained ingestion. These findings indicate the potential of EPG for rigorous observation and quantification of feeding and egg-laying in phytophagous species beyond hemipteroids, and should allow exploration of how substrate differences may alter such activities.

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Quantitative differences in feeding behavior of 3rd instar nymphal Lygus lineolaris (Hemiptera: Miridae) occur on transgenic Bt cotton compared with non-Bt cotton

Authors: F. Cervantes, E. Backus, L. Godfrey, W. Akbar, T.L. Clark, M. Rojas

Cotton production in the mid-southern United States is severely affected by direct damage to cotton squares during feeding by the tarnished plant bug, Lygus lineolaris, especially immature, nymphal life stages. Cotton plants carrying the protein Cry51Aa2.834_16 derived from a soil-dwelling microbe, Bacillus thuringiensis (Bt), were recently shown to have long-exposure insecticidal effects on nymphs of L. lineolaris. However, development of lygus-resistant varieties of crops depends upon more than just killing the insects, but also upon changing the types and amounts of short-term feeding performed on cotton. One of the most important tools for studying insect feeding is electropenetrography (EPG), in which an electrical signal is applied to a plant on which a gold wire-tethered insect is feeding. The present study compared feeding behavior of nymphal L. lineolaris on pin-head cotton squares of non-Bt vs. Bt plants using AC-DC EPG. Overall, insects performed the same types of behaviors on both non-Bt and Bt-cotton squares. However, differences in numbers and duration of behaviors were observed between treatments. Nymphs on cotton increased their testing/tasting behaviors, made more frequent events of salivation, and decreased ingestion, supporting that Bt cotton plants were less palatable and/or digestible to L. lineolaris nymphs than non-Bt cotton. Results show that Bt cotton can productively contribute to an integrated pest management program for lygus bugs in cotton.

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Carbon and nitrogen dynamics affected by different irrigation and fertilization practices in a pomegranate orchard

Authors: R. Tirado-Corbala, S. Gao, J. Ayars, D. Wang, C. Phene, R. Phene

Soil carbon and nitrogen dynamics can partially reflect soil health status or how agricultural practices impact the sustainability of an agronomic production system. This research investigated the effect of high frequency drip irrigation and different levels of nitrogen application on total or...
soluble carbon and nitrogen concentration changes in soil and leaf tissues as well as total N uptake in fruits in a 1.4-ha pomegranate orchard in the San Joaquin Valley of California. Results clearly show the advantage of using subsurface drip in comparison with surface drip irrigation in total N uptake with different patterns of soil C and N distribution. This research concluded that the proper amount of N needed for a 4–6 year old pomegranate orchard is in the range of 166–263 kg/ha, and demonstrated that high frequency subsurface drip irrigation can lead to higher N use efficiency and minimized leaching loss while increasing water use efficiency. The results are applicable to other orchards in the same region or other regions with similar irrigated agriculture.

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The National Academies of Sciences, Engineering, and Medicine recently released a comprehensive review of the status of research on citrus greening (Huanglongbing) with recommendations for future research. The report can be downloaded for free at the link below.