

# CITRUS ADVANCED TECHNOLOGY PROGRAM

**PREPROPOSAL FORM:** Control of Citrus Greening, Canker & Emerging Diseases of Citrus



Proposal Title

**Novel formulations and application methods for bactericides to control systemic HLB infection**

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## PURPOSE & RATIONALE OF STUDY

HLB caused by phloem-limited *Liberibacter asiaticus* (Las) is unlikely to be contacted by bactericides applied to the leaf surface. Objective 1 is to evaluate trunk injections of soluble Cu (Magna-Bon, M-B) and oxytetracycline (OTC) that have already been shown to be effective for systemic control of HLB. Trunk injection and rates of bactericides will be modified to facilitate ease of application and minimize the cost while maximizing residual activity. These trials will be conducted in HLB affected groves with trees that are healthy, asymptomatic and mildly declined. Objective 2 will test topical applications of bactericides to the trunk with pentra-bark, a product already proven effective for trunk applications of systemic insecticides and fungicides. Trunk applications of bactericides will be applied to young trees and their systemic activity quantified *in vitro* with a bioassay against *Xanthomonas citri* subsp. *citri* (Xcc), in greenhouse plants inoculated with LAS, and in the field with young HLB infected trees. PCR will be used to monitor Las infection.

## TECHNOLOGY & EXPERTISE

In a field trial, 3-5 ml trunk injections of M-B (copper sulfate pentahydrate) at 500-1000 ppm were made at 1 and 3 mo intervals to 5-yr old Hamlin orange that were healthy (PCR-), asymptomatic (PCR+ without HLB symptoms) and PCR+ with mild decline. Trees were assayed by PCR and HLB canopy symptoms were visually assessed before and at 5 mo. after injections began in April 2009. All M-B treatments produced changes in canopy ratings that reflected positive response compared to no change for the non-treated trees (NTC). Ct values for the NTC trees indicated positive HLB status, whereas Ct's for M-B treated trees were higher and indicated HLB- or threshold bacterial infection. SAR inducing Admire (imidacloprid) at 1X rate was applied twice to the trunk or soil surface of 4-yr old Hamlin trees with a hand sprayer and effectiveness against citrus canker evaluated in comparison to 6 copper sprays. Trunk application control matched that of soil application or copper sprays confirming systemic activity through the bark.

## TIMELINE & MILESTONES

In greenhouse, trunks of sweet orange plants will be painted with a range of concentrations of M-B, OTC or non-treated. After uptake for 2wk, leaves will be assayed for Cu by nutrient analysis and with leaf bioassay using injection infiltration with Xcc. Systemic activity will be assessed as *reduction in canker* lesions at 2wk after inoculation. Trees with bactericidal activity will be graft-inoculated with HLB-infected budwood or left noninoculated. Starting at 1 mo post inoculation, leaf samples will be taken every month to test for Las titer. A trial with trunk paints will be conducted in the field with 3 yr-old Hamlin trees that are already HLB+ or HLB-. A trial will also be initiated in 7-yr old Valencia trees that are healthy (PCR-), asymptomatic (PCR+ without HLB symptoms) and PCR+ with mild decline. The trial will compare trunk treatments made by infiltration of solutions of M-B and OTC at concentrations that approximate those in previous trials. In both trials, trees will be assayed by PCR to estimate Las titer and HLB canopy symptoms visually assessed before and at 6 mo. after treatments begin.

## EXPECTED RESULTS & PRACTICAL APPLICATION

Nutritional sprays of trees in Florida groves are purported to suppress symptoms of HLB, but thus far have had no effect on bacterial titer. The major concern is that because nutritional supplements do not limit Las titer, their use will promote area-wide inoculum build-up and HLB spread. Bactericidal treatments under evaluation in this proposal are expected to not only suppress HLB symptoms, but also control bacterial titer and reduce inoculum spread. The greenhouse and field trials are likely to provide answers quickly enough that rates and application methods can be optimized in a few years time. The viability of bactericides for long-term therapy for reducing pathogen spread and/or damage to the citrus tree will require a multi-year field study. If the systemic activity of bactericides prevents or slows bacterial replication and spread in the tree or prevents pathogen-induced damage to the phloem tissue, it may have promise as an inexpensive long term chemotherapy for HLB.

**SUBMIT PROPOSAL**