

CALIFORNIA CITRUS NURSERY BOARD

Project Plan/Research Grant Proposal

Project Title: Annual citrus tristeza virus index at the Lindcove Research & Extension Center.

Project Year: 2013 Anticipated Duration of Project: 10+ years

This project is: New or X Ongoing (Year 10 of 15)

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Additional sources of funding: The Citrus Clonal Protection Program tests the screenhouse trees and the Tulare County Pest Control District tests trees for CTV and MCA13 reactivity in a 1 mile radius around LREC.

Executive Summary

To protect the Citrus Clonal Protection Program (CCPP) field plantings and other research programs from citrus tristeza virus infections (CTV), an annual index of all trees is conducted each year at the 175 acre University of California Lindcove Research & Extension Center (LREC). During the period of 1990-2006, an average of 3 CTV-positive infected trees were found and removed each year in the research blocks and for a 14 year period, no CTV-positive trees were found in the CCPP foundation blocks, demonstrating that infected tree removal at the Center provided protection of the foundation block from this disease. This low level of CTV incidence at the field station and lack of CTV in the foundation block allowed budwood to be released from the field trees in the foundation block 2-3 times/year. During 2007, 4 CTV-infected trees were found in the foundation blocks and 48 CTV-infected trees were found in the research blocks and in 2008 an additional 83 CTV-infected trees were found (8 in the foundation blocks and 75 in the research blocks). The epidemic of CTV at LREC had two consequences. In the foundation blocks caused the CCPP program to stop releasing budwood from the foundation blocks and instead release budwood from the LREC screenhouse. In the research blocks, many infected trees were removed affecting research results, especially in scion and rootstock trials. Thus the epidemic of CTV affected both the budwood program and research at LREC.

The sudden increase in the incidence of CTV-infected trees at the field station is thought to be due to several factors. First, there has been no CTV-infected tree removal in the commercial citrus orchards surrounding LREC since 1998. Surveys of the citrus orchards in the ½ mile surrounding LREC indicate that the incidence of CTV has increased from an estimated 0.14% of trees in 1998 to 1.2% in 2007. Thus, there was a 10 fold increase number of CTV-infected trees surrounding the station. Secondly, a number of growers have planted pomegranates among the citrus orchards within ½ mile of the field station and these orchards produce heavy populations of winged cotton aphids in the spring, the primary vector of CTV. The presence of more aphids during the spring when virus titer is high increases the likelihood of transmission of the virus. Finally, weather conditions have been cool and wet in the spring for the past few years, favoring aphid survival and lengthening the time that the flush, that is attractive to aphids, is present on the trees.

After a series of meetings in 2007 and 2008, the Tulare County Pest Control District (TCPCD) voted and provided funding for aphid pesticide treatments in a 1-2 mile radius around LREC in an effort to reduce the

aphid vectors and so reduce the incidence of CTV at LREC since 2008. The Kern, Southern Tulare and Central Valley Pest Control Districts also contribute funding to this effort because the entire citrus industry benefits from protecting LREC from CTV. Citrus around LREC is being treated in the spring with Assail (acetamiprid) and in the fall with Admire or generics (imidacloprid) and pomegranates are being treated in the fall with imidacloprid. Citrus and pomegranates in backyards in the town of Lindcove are also being treated with imidacloprid in the fall. Treatments started in the fall of 2008 and with the latency period of CTV, we did not expect to see significant success in the form of reduced CTV infection of LREC tree immediately. However, in 2009, the number of infected trees dropped to 52, and in 2010, 2011, and 2012 that number dropped further to 20, 12 and 20 trees respectively. Thus, we have evidence that aphid treatments are successfully reducing the incidence of CTV at LREC and protecting the foundation field trees and research program there. The TCPCD also provides funding for surveying for MCA13 reactive, potentially severe strains of CTV in a 1 mile radius around LREC and 47 trees have been detected and have been voluntarily removed by the growers to further protect LREC.

Project's Benefit to the Citrus Nursery Industry:

CTV infection of trees at Lindcove Research and Extension Center (LREC) has shifted the budwood release program from the outdoor foundation blocks to the protected structure foundation blocks to ensure that the budwood is disease free. Continued testing and removal of CTV positive trees in the outdoor trees is necessary because these trees are needed for assessing true-to-type characteristics. Four Pest Control Districts are providing funding to treat for aphids in a 2 mile radius around LREC during spring and fall and to conduct testing for severe strains of CTV in a 1 mile radius around LREC in an effort to reduce CTV spread to LREC. CCNB funding to detect and remove CTV-infected trees at LREC is critical for protecting LREC foundation block trees (valuable resource for true-to-type characteristics) and the research plots.

Objectives:

Provide funds for the leaf collection and direct tissue blot immunoassay (DTBIA) testing by LREC to detect CTV positive trees on the Lindcove Research & Extension Center with tree removal to follow detection.

Workplans and Methods

1) CTV testing:

During 2012, 50% of the trees were tested using DTBIA and 50% by the CCTEA ELISA. During the spring in 2013, we plan to test all of the field trees with DTBIA and utilize the CCTEA testing only for confirming positives and providing data on MCA13 reactivity. Four new flush leaves with petioles are collected/tree and clipped with a cigar cutter. The petioles are then each pressed to a membrane. The field number and the start of each row is marked with a pencil. Blots for about 120 trees can be fit onto one membrane. Positive controls on the membrane are provided by Ray Yokomi's laboratory. Groups of membranes are processed using a CTV ImmunoPrint kit from Agida. Membranes are processed until the positive controls develop a bluish-purple color. After drying, the membranes are examined for dark spots indicating a reaction using a microscope. Trees that have one or more positive reactions are retested with DBTIA, and tested with ELISA by the CCTEA and PCR by Ray Yokomi's laboratory. The CCTEA and Yokomi labs confirm the DTBIA and test for MCA13 (severe strain) reactivity.

2) Tree removal:

CTV positive trees are treated with a 0.25 lb AI rate of foliar Assail (acetamiprid) to kill any aphids that may be infesting the tree and removed 24 hours later. These treatments and removals will occur a short period (within a few weeks) after the tree is confirmed to be CTV-infected.

Project Management and Evaluation

Dr. Grafton-Cardwell will report the results of the CTV testing to the Citrus Clonal Protection Program director, Georgios Vidalakis, the nursery industry and to the citrus industry in general. Kurt Schmidt will oversee the leaf collection and submission of samples to the CCTEA. Jill Barnier will oversee the testing of samples at the CCTEA and provide the results of the testing.

