



Request for Proposals

CRDF Study on Preharvest Fruit Drop Prevention Using Plant Growth Regulators (PGRs)

Objectives of the Project: To determine the efficacy and impacts on fruit yield, fruit quality, fruit drop, canopy health and root density when using Citrus Fix™ and ProGibb® in the use patterns prescribed herein.

Situation Statement: This is a one-year study to evaluate the efficacy of 2,4-D and gibberellic acid (GA) applications to prevent or reduce HLB-induced preharvest fruit drop in round oranges in Florida. The possibility for a second year of this trial will be determined following the analysis of the first year's data. Numerous Florida citrus growers have reported positive results when applying Citrus Fix™ (2,4-D) and ProGibb® (gibberellic acid) – alone and in combination - in the months preceding harvest. Different use patterns for these products have been implemented. With this study, these use patterns with these products will be tested in side-by-side, multiple-site trials to better understand the impacts on fruit yield, fruit quality, fruit drop, canopy health and root density.

Narrative:

This study will test the hypothesis that Citrus Fix™ and ProGibb®, when used alone or in combination, may significantly reduce or prevent preharvest fruit drop. Both products are commercially available and have industry acceptance. It is hoped that after one season of the prescribed spray applications and data collection from multiple trial sites there will be enough evidence to draw tentative conclusions regarding the impacts of these treatments.

Description of the Project: Nine foliar applied treatments will be evaluated plus an untreated control (10 plots). The trees used in this trial should be of similar visual health and must be the same rootstock, scion, and age. The 10 treatment plots will be replicated five times with each plot containing seven trees. Evaluations will be focused on the center five trees of the seven-tree plot. Quality horticultural care should continue throughout the trial.

Treatments for 2,4-D and gibberellic acid trial

All treatments shall be applied with an air blast sprayer at a spray volume of 150-200 gallons per acre at a sprayer speed of no more than 1.5 mph. Consistent and sufficient spray coverage is a priority.

1. Water treated control
2. Label Rate - 3.2 oz/acre Citrus Fix™ single application

- a. No surfactant or spray oil
- b. Hamlin application in October
- c. Valencia application end of December
3. 2.5 oz/acre Citrus Fix™ divided into 3 applications + water
 - a. 0.5 oz/acre, 1.5 oz/acre, 0.5 oz/acre
4. 2.5 oz/acre Citrus Fix™ divided into 3 applications + water + Tactic™
 - a. 0.5 oz/acre, 1.5 oz/acre, 0.5 oz/acre
5. 2.5 oz/acre Citrus Fix™ divided into 3 applications + water + 435 spray oil
 - a. 0.5 oz/acre, 1.5 oz/acre, 0.5 oz/acre
6. 3.2 oz/acre Citrus Fix™ + water (2 applications at full label rate)
7. 3.2 oz/acre Citrus Fix™ + water + Tactic™ (2 applications at full label rate)
8. 3.2 oz/acre Citrus Fix™ + water + 435 spray oil (2 applications at full label rate)
9. 20 oz/acre ProGibb® LV + Tactic™ (1 application at night)
10. 20 oz/acre ProGibb® LV + 1 oz/acre Citrus Fix™ + Tactic™ (1 application at night)

Timing:

Citrus Fix™ - Split applications with an application in the months of July, September, November (treatment 3, 4 and 5, regardless of variety)

Citrus Fix™ - two full-rate applications with an application in August and an application in October (treatment 6, 7 and 8, regardless of variety)

ProGibb® – a single nighttime application when the fruit transitions from dark green to light green (treatment 9)

ProGibb® + Citrus Fix™ – a single application at night (Hamlin – September, Valencia – November) (treatment 10)

Proposed Start and End Date: Treatments shall begin after normal June drop. Treatment application and data collection will continue through harvesting.

All tasks outlined herein shall be performed by the Researcher unless otherwise specifically stated.

Site Selection:

Field trial sites shall be chosen by the Researcher, subject to approval by the Citrus Research and Development Foundation, Inc. (CRDF).

Trial design:

At each site there will be five replicates of the 10 treatments with seven trees per plot.

10 treatments x 5 reps x 7 trees per replicate = 350 trees at each site.

Studies will focus on a subset of measurement trees in each plot. **Measurement trees** shall be five uniform trees in the center of the seven-tree plot. 10 treatments x 5 reps x 5 trees = **250 measurement trees** at each trial site. The two end trees within the seven-tree main plots will be treated but not evaluated and serve as buffer trees.

All products shall be applied as described in this proposal. Spray applications shall be applied with an airblast sprayer at a volume of 150-200 gallons per acre. Consistent and sufficient spray coverage is important for this trial. Evaluations will be focused on the center five trees of each treatment plot. The trial design should allow for enough space between treatment plots to minimize the chances of spray drift affecting the other treatment plots.

Data collection:

Pre-treatment tree and grove evaluation shall include scion, rootstock, soil type, soil pH, and cultural practices, including irrigation, fertility programs, and pest/psyllid control, and yield and fruit quality records from the past two years.

DI Rating:

All measurement trees must be initially visually rated using eight canopy sectors for **Disease Index** (DI= 0 to 40, Gottwald et al., 1989, see below). Only trees with a DI score of **less than or equal to 24** will be chosen and flagged as measurement trees. All measurement trees (250 at each site) will receive a unique ID code that will allow for easy identification of treatment, replication, and tree number within the plot. GPS locations for each tree should be noted if available. ID codes will be built into Excel data sheets for DI data entry.

Measurement trees shall be **DI rated** at the start and end of the trial. Excel spreadsheets with the DI rating data shall be sent to the CRDF Research Coordinator in a timely fashion.

Root density:

Root density samples (dry wt/vol) from 20 soil cores per tree shall be collected at the start and end of the trial for root density evaluations. These cores shall be collected and combined into a one-gallon sized ziplock plastic sample bag with the unique ID code for each measurement tree (250 trees/site). Each soil core shall be taken from the wetted zone under the tree. Each soil core shall be one inch in diameter by nine inches in depth. Soil samples shall be stored in a dark cooler and promptly delivered to the Syngenta laboratory (LaBelle, FL) for analysis.

Fruit yield and quality:

At **harvest**, a 65-fruit sample shall be collected from each treatment plot, placed into a labeled mesh bag and delivered to the CRDF Research Coordinator at CREC for **fruit quality** analysis (since there are 50 treatment plots, there shall be a total of 50 mesh bags delivered, each with 65 fruits). Harvest lead time will be anticipated and communicated to the CRDF Research

Coordinator for scheduling. **Total yield** shall be collected from the 50 treatment plots. Fruit weight per plot shall be submitted on Excel data sheets and sent to the CRDF Research Coordinator in a timely fashion. Weight of the 65-fruit quality samples shall be added to calculate total yield per treatment plot.

Scheduling for the collection of all fruit quality and soil samples shall be coordinated with the CRDF Research Coordinator.

Fruit Drop:

Fruit drop shall be assessed by the Researcher starting three months prior to the expected harvest time for the scion being evaluated at each trial site. Prior to the start of evaluating fruit drop, all fruit under the measurement trees shall be removed but not counted. From that point on, fruit drop shall be measured once every two weeks. Dropped fruit are to be counted and removed from underneath each measurement tree. Fruit drop counts shall continue until harvesting. At harvest, a 100-fruit subsample shall be counted and weighed by the Researcher. Fruit drop counts will be reported for each measurement tree and the data shall be delivered to the CRDF Research Coordinator on an Excel spreadsheet.

Data interpretation and analysis: Data will be analyzed by the CRDF Research Coordinator at CREC with assistance from the contracted statistician.

1. Yield per plot (weight in Kg)
2. Fruit quality (50 samples per site, 65 fruits per sample)
3. Fruit drop per tree (calculate percentage of fruit drop) (total fruit drop/(total fruit drop + # of harvested fruit))*100
4. Visual ratings per tree (DI Rating)
5. Root density (dry wt/vol)

Proposal Submission:

Full Proposals must be submitted to catp@citrusrdf.org, and received by 5:00 p.m. Eastern time on Friday, April 22, 2022

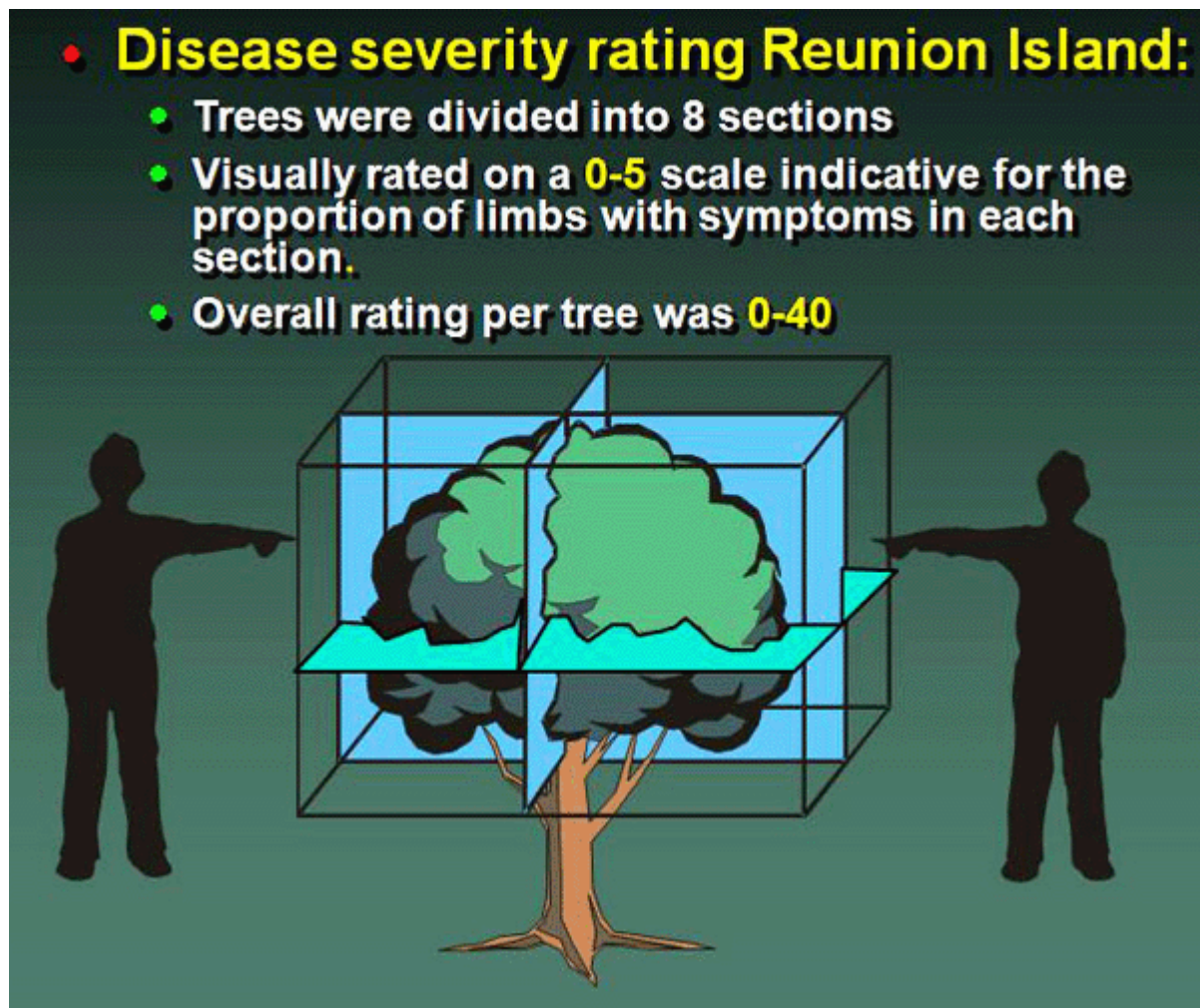
The following documents will comprise the Full Proposal package:

1. **Full Proposal Cover Page and Budget** - Form CB-21 -link to download form: <http://citrusrdf.org/wp-content/uploads/2021/03/CB-21-Cover-Page-and-Budget.pdf> (5 pages; pdf file).
2. **Budget Justification** (Word file)
3. **Subcontract Budget** - Form SC-21 -link to download form: <http://citrusrdf.org/wp-content/uploads/2021/03/SC-21-Subcontract-budget.pdf> (4 pages; pdf file. SC-21 forms do

not count in the 7-page limit) - complete if applicable, using a separate form for each proposed subcontract. See detailed instructions later in this file.

All questions should be directed to catp@citrusrdf.org.

- **Disease severity rating Reunion Island:**
 - Trees were divided into 8 sections
 - Visually rated on a **0-5** scale indicative for the proportion of limbs with symptoms in each section.
 - Overall rating per tree was **0-40**



Tree canopy decline index (DI) score: Each canopy hemisphere will be subdivided into four equal quadrants by two imaginary perpendicular planes (vertical and horizontal at mid canopy height) passing through the axis of the tree trunk. The resulting eight sections are scored individually on a 0-5 scale indicative of the proportion of limbs expressing HLB disease symptoms within each section (0 = no limbs, 5 = all limbs). The summation of the eight scores for each tree will result in a severity rating of 0-40 for each tree on each survey date. Trees that were severely declined with initial DI scores greater than 24 (average $DI\ 3 \times 8 = 60\%$ declined with symptoms), will not be chosen as measurement trees within each plot.

Gottwald, T. R., Aubert, B., and Xue-Yuan, Z. 1989. Preliminary analysis of citrus greening (Huanglongbing) epidemics in the People's Republic of China and French Reunion Island. *Phytopathology* 79:687-693.