



GROWER RESEARCH REPORT

JANUARY 2022

Moving Forward in a New Year

by Rick Dantzler, COO of CRDF



Several months ago, CRDF staff wrote a document titled Pathway to a Sustainable Florida Citrus Industry. We did it for several reasons. First, we wanted to honestly assess where we were in our research effort. Second, we wanted to make sure we had a plan in place to address what needs to be done and weren't simply chasing the latest shiny thing. And finally, we wanted growers and state policymakers

(funders) to know that we weren't funding everything that came along, that we had a strategy in place that would drive our overall research funding effort.

I shared many of its components with you in the October 2021 Grower Report. Since it will drive much of what CRDF funds, much of our work over the last quarter has involved laying the groundwork for the research that will put that document into action.

We also operate pursuant to a philosophical position that, while there is still a need for long-term research that will put HLB behind us once and for all, CRDF is attempting to use its funding on work that will provide more immediate help to you, leaving many of the long-range projects to Uncle Sam. We are also placing a premium on improving juice quality because we understand that preserving your ability to produce outstanding not-from-concentrate juice is critical to profitability.

We are also continuing our oversight of projects already funded. The reality is that research priorities sometimes change, and with funding becoming tighter it is necessary to terminate projects or change them in mid-stream, making sure that your money is working for you as best it can. This is terribly disruptive to researchers, though, so CRDF works with them to minimize negative impacts when possible. Still, we can't always mitigate these impacts, so it is not something we do lightly. Nevertheless, making these tough calls is required in this environment of reduced research funding and difficult market conditions for growers.

In the last three months I have also had several interesting meetings with groups of growers. A question I sometimes ask is, "If you were me, what would you have CRDF do?" I've gotten some interesting answers.

One answer I received not long ago was to do more research on 2,4-D as a way to reduce drop. This has made its way into our project plans.

Another response was to embark upon a crash planting program of Sugar Belle or other orange-like hybrid for the purpose of growing fruit for processing, not fresh, as a way to blend with juice from citrus sinensis and improve quality. Sugar Belle has its problems, not the least of which is a tight harvest window and softness, but it is an intriguing idea. We had an interesting discussion about this at a joint meeting of our research committees just last week.

A third interesting topic was a theory posited by a grower that budwood grown from trees that had not gone through the budwood cleanup program of DPI did better than trees grown from budwood which had, that there was something about the cleanup process that eliminated not just bad stuff but good stuff, too. He further opined that the threat from the bad stuff was not as great as the threat of HLB, and faster turnaround of promising budwood to the field was more important at this point in the industry's existence than guarding against the bad stuff. I'd be curious to know what you think about this.

Last but certainly not least, the Grower Referendum that funds CRDF is up for renewal this year. Florida statutes govern this process, and the Florida Department of Agriculture and Consumer Affairs conducts the vote. CRDF has no formal role, and because it is a 501c3 corporation we are limited to helping educate growers on questions they may have. If you wish for me to meet with you or a group you are a part of, please let me know and I will be there.

NEW RESEARCH FUNDED

CRDF made nine funding decisions for new research since my last Grower Report. They are:

PROJECT NUMBER	PRINCIPAL INVESTIGATOR	PROJECT TITLE	INSTITUTION
21-005	Albrecht	Comparison of field performance of citrus trees on rootstocks propagated by seed, cuttings, and tissue culture	UF/IFAS SWFREC
21-012	Dewdney	Evaluating the role of greasy spot and peel disorders in the greasy green defect on citrus fruit	UF/IFAS-CREC
21-013	Duncan	Integrated management of sting nematode in newly planted citrus trees.	UF/IFAS-CREC
21-014	El Mohtar	CTV-T36 vectors as a tool to induce efficient flowering in citrus seedlings	UF/IFAS-CREC
21-021	Pelz-Stelinski	CLas Inhibition with Antisense Oligonucleotides for Management of Citrus Greening Disease	UF/IFAS-CREC
21-024	Schumann	Determine optimal timing for application of fertilizer to improve fruit quality and reduce preharvest drop	UF/IFAS-CREC
21-032	Albrecht	Assistance with Phase 3 Rootstocks Trials	UF/IFAS SWFREC
18-004	Bowman	Development of SuperSour and other outstanding rootstocks with tolerance to HLB (bridge funding through April 2022)	USDA/ARS-USHRL
18-058C	Stover	Fort Pierce Field Test Site for Validating HLB and/or ACP Resistance (bridge funding through April 2022)	USDA/ARS-USHRL

BAYER UPDATE

As you know, CRDF embarked upon a large project with Bayer Crop Science in 2017. In fact, it's the most expensive project in CRDF's history. Two years ago, the project was transitioned to a federal grant.

It has three objectives: One, to construct a high-throughput system that can screen antimicrobials against liberibacter (done). Two, to develop a commercially available antimicrobial(s) that has efficacy against liberibacter (in progress). Three, to develop a commercially available plant growth modulator(s) that has efficacy against liberibacter (in progress).

Several plant growth modulators fell by the wayside recently because of health and environmental concerns, but many more in the same class are showing equal efficacy against liberibacter and don't have the health and environmental concerns. This is an advantage of working with a private company; registration personnel work in tandem with researchers to make sure that what stays in the pipeline can survive the regulatory process. Work is progressing so well that Uncle Sam has awarded the project the third year of the grant.

PEPTIDE UPDATE

CRDF has been working with three companies that have proprietary peptides: Elemental Enzymes, Invaio and Southern Gardens Citrus. I've written about this before; here's an update:

ELEMENTAL ENZYMES:

Elemental Enzymes (EE) is a midwestern company that has conducted more than 40 field trials in Florida of a novel peptide and applied for EPA approval to market the product. CRDF has been in touch with EPA to encourage an early decision, and just last week I spoke with EPA again to ask that the decision be made quickly. I am not confident that the decision is imminent, which is unfortunate because the product works best when applied during the spring flush, which is nearly upon us. Nevertheless, we will keep pressing.

INVAIO:

Invaio is the company that is developing the finger lime peptide product to attack the HLB bacterium in two different ways: to directly inhibit its multiplication and to induce the plant defense against the disease. CRDF maintained constant contact with Invaio for a year after news of the peptide broke. We offered to pay to test the product against the hairy root assay at Texas A&M University and provided university and USDA researchers with \$200,000 worth of the peptide to test on trees of fruit-bearing age as well as greenhouse trees.

SOUTHERN GARDENS CITRUS:

Southern Gardens Citrus has numerous peptides they have been testing and is also using the CTV vector to apply spinach defensins (also peptides) to citrus trees to reduce losses due to HLB. This work has been underway for more than a dozen years and is approaching approvals by EPA and FDA for this biological pesticide.

These peptides are now in their second generation. Southern Gardens is aggressively testing them, and several appear to be highly effective at killing *Liberibacter*. Trees containing the new defensins were recently planted to see how they hold up in a commercial growing environment. CRDF is maintaining a constant dialogue with Southern Gardens to help where it can in order to bring these peptides and CTV vectoring to the marketplace as soon as possible.

In the last 10 months, Invaio has not provided CRDF with any new information, but I expect to hear results from either the company or researchers announcing their findings shortly, certainly within the next few months.

TREE INJECTION OF PEPTIDES AND ANTIMICROBIALS

CRDF is continuing its work with a novel type of injection device that makes it economical to inject antimicrobials and peptides into the vascular system of citrus trees. First tests to gauge uptake were not encouraging, but we believe it was because what we were injecting was in a powdered form. We certainly haven't given up on this and are working with the company that makes the device to see how we can make it work more effectively.

I was recently invited to attend a tour of a field trial in which oxytetracycline (oxy) had been injected into trees using muriatic acid and oxy at a pH of 7 and at a pH of 2. Other acids were tried, including citric acid and oxy at a pH of 7 and a pH of 2. I was given the chance to bring a few growers with me, which I did.

To my eye, the trees receiving oxy and muriatic acid, and oxy and citric acid were markedly healthier looking than the trees that received no treatments. I'm told that data indicates that muriatic acid works better than citric acid, but both looked better than the control trees.

These trials, which were replicated, have been done entirely with private sector funding, although work CRDF previously funded laid a portion of the predicate for these advancements. What distinguishes this work is that the company behind the effort intends to apply for a Section 3 registration with EPA and possibly a Section 24C with the State in February. CRDF has asked to see more data, which is being provided. Expect to hear more about this in the coming weeks.

FIELD TESTING NEW ROOTSTOCKS AND SCIONS

CRDF has led a grower effort to revise the breeding requirements of citrus breeding programs and has begun an effort to independently field-test new rootstocks and scions. What's more, UF/IFAS has hired a citrus horticulturist with a breeding background to, among other things, help implement these guidelines.

CRDF is putting in three new Stage 3 field trials for the 12 most promising new rootstocks (two that are used by some growers but haven't been thoroughly tested), and 20 of the most promising fruit scions (separate trials). We still need one Ridge site for the rootstock trials and three grower-cooperators for the scion trials. Please contact us if you are interested.

CRDF has also convened nine plant breeding experts from across the country to help it determine where to spend what appears to be a diminishing amount of money for plant breeding. We simply must get this right since a resistant or sufficiently tolerant tree is probably our best shot at getting this disease behind us.

TRANSGENIC CITRUS

A number of transgenic scions and rootstocks have shown tolerance or resistance to HLB and are now in the field. Some of these trees are expressing a gene to increase plant defenses and others produce a peptide against CLAs.

These transgenic trees, which would be considered GMOs, are expected to provide resistance or tolerance to HLB for their lifetime without any extra labor input. CRDF believes the development of these GMO cultivars and their approvals should be finished while the industry debates the policy question of bringing such fruits to market. At the very least, these cultivars could identify desirable traits against HLB that could be duplicated in a non-GMO manner. CRDF is helping to fund Dr. Zhonglin Mou and Dr. Nian Wang to perform such work.

CRISPR

CRDF has funded groups that have developed new technologies that now allow the development of citrus varieties resistant or tolerant to HLB without being considered GMOs.

A new technology – clustered regularly interspaced short palindromic repeats - referred to as "CRISPR," allows the introduction of small deletions in the genes needed for CLAs infection. The value of this process is that it does not insert foreign nucleotides into the tree, resulting in a non-GMO, HLB-resistant tree. The difficulty is knowing which gene or genes to silence.

However, Dr. Zhonglin Mou and his team are bypassing the ambiguity of what gene to silence with CRISPR by duplicating the phenotype of a transgenic tree that has been shown to be tolerant to HLB. By understanding the relationship between the multiple genes involved in plant defense, they are knocking out a gene that blocks the induction of tolerance to HLB. CRDF is funding the field trial testing of these plants.

COMMITTEE REORGANIZATION

Earlier in this report I mentioned a joint meeting of our two research committees, the Research Management Committee and the Commercial Products Delivery Committee. The board recently made the decision to merge the committees into one. The new committee will be allowed to have up to 17 members and will likely be divided into three sub-committees. Our plan for the new committee will be to use committee members to not just help with funding decisions, but to assist in monitoring projects. I will have more to say on this later.

