Title:	**Rear and Release Psyllids as Biological Control Agents - An Economical and Feasible Mid-Term Solution for Huanglongbing (HLB) Disease**

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Project Director
Thomas Turpen
469-371-2608
catp@citrusrdf.org

Recipient Organization
CITRUS RESEARCH AND DEVELOPMENT
700 EXPERIMENT STA RD
Lake Alfred, FL 338502243
DUNS No. 961745697

Performing Department
{NO DATA ENTERED}

Co-Project Directors	**Departments**
Burns, Jacqueline	Dean for Research (IFAS)
Polek, Mary Lou	{NO DATA ENTERED}
Browning, Harold	Agricultural Research Service
Patt, Joseph	Subtropical Insects Research
Shatters, Robert	
Pelz-Stelinski, Kirsten	

Non-Technical Summary

This proposal presents research targeting the elimination of Huanglongbing (HLB) as an economic threat to US citrus production by blocking the ability of the psyllid insect to move the causative agent of this disease between infected and healthy trees. The primary long term goal of this project is to interfere with the spread of HLB within citrus orchards where HLB disease is established and to interfere with the invasion of disease organism into areas where the insect that transmits the causal agent is established, but in which HLB has not been detected, by strategically releasing a nuPsyllid population that is incapable of moving the disease. A further goal is to ensure the necessary adoption of the method by the social system of growers, and understanding and acceptance by consumers and the general public. Once established, this novel system of biological control would be operationally transferred to the citrus industries of U.S states (Florida, California, Texas and Arizona). Other ongoing support, if necessary, will be provided by the stakeholder organizations. We believe current management practices are not sustainable, and in any event psyllid vector eradication has never been achieved, except on small islands. Alternative HLB-management approaches must be developed as a mid-term solution to the HLB problem. Without control measures in hand, citrus growers have no incentive to replace infected trees or to replant entire orchards. The uncertainties associated with HLB will undermine the stability of the industry in currently HLB-free areas. A mid-term solution is crucial to maintain a profitable industry until citrus varieties with resistance to HLB can be developed and released. Therefore, we present a novel and more environmentally friendly alternative strategy, which we will convey to growers and the public. Grower response to this disease has resulted in a mix of increased costs, modifications to long-successful production management systems and acceptance of at least short-term yield and/or quality reductions. Total orchard loss is increasing as HLB spreads. The current situation suggests that without development of an adequate control strategy, commercial citrus production will become economically unfeasible. This disease also is impacting the millions of citrus trees grown in homeowner yards.

Accomplishments

Major goals of the project

The primary long term goal of this project is to interfere with the spread of HLB within groves where HLB is endemic and to interfere with the invasion of CLas into areas where ACP is established but HLB has not been detected. Once released and established, the nuPsyllid population will naturally penetrate and displace the wtPysllid population. A further goal of this proposal is to ensure the necessary adoption of the method by the social system of growers, consumers and the general public in citrus states. To achieve these goals, we propose a three-fold approach: 1). Develop a psyllid management strategy based on the development of psyllid populations incapable of transmitting CLas (nuPsyllid) and strategically release the nuPsyllid population to displace current ACP populations that have invaded the US. 2). Provide optimized management strategies for integration of the proposed population displacement technique into current management practices: a. Southeast and Southern U.S. (FL and TX) where both the ACP and CLas are endemic. b.Western U.S. (CA, AZ) where ACP is present and spreading while there is currently no detection of HLB. 3). Integrate the management strategies with monitoring strategies to continually assess effectiveness and provide outreach education to the grower stakeholders and citizens about the control strategy. The feasibility of the approach proposed here is supported by the experience with HLB management in Florida through the creation of Citrus Health Management Areas (CHMAs). The CHMA is based on the recognition that HLB has an important edge effect. Although insecticide applications can control ACP populations within the grove, without effective ACP management in the surrounding areas, CLas-bearing ACP rapidly returns. A CHMA coordinates the insecticide sprays and other management activities over a large area, thus greatly reducing the edge effect of ACP re-invasion. Thus, we are proposing that most nuPsyllid releases will be focused at the periphery of CHMAs and other smaller management areas to displace the endemic population at the periphery. Vigorous ACP control measures would be continued temporarily in the interior of the management area but would be gradually tuned down to allow populations of CLas-transmission-deficient ACP populations to become established

What was accomplished under these goals?

The purpose of this NIFA-CAPS is to create options for management of HLB by replacing the wild type ACP with a population that is unable to transmit the bacterial causative agent CLas. Achieving this outcome will require progress in the following three areas of emphasis - An Effector Mechanism, A Driver System, and Diffusion. At the end of the fourth year of NIFA support significant progress has been accomplished in each of these areas as summarized below.
Effector Mechanism - Initial assessments have not identified the required variation in CLas transmission to occur naturally in ACP populations. However the prospects for engineering a mechanism to achieve the desired phenotype are under active investigation. The <u>effector is the content</u> of the phenotypic change we aim to introduce. Candidate effectors are being identified through multiple parallel methods of investigation.

- Extensive transcriptome data sets (Transcriptome Computational Workbench; TCW) have been determined for whole adults, adult salivary glands, adult guts, and nymphs, infected or uninfected with CLas or CLso, and published datasets are made available to the research community, at www.sohomoptera.org/ACPPoP.
- The TCW is being updated to allow for protein sequence and spectral count data, in addition to the transcript and expression count data to allow for better identification of RNAi targets from protein data.
- Data from ongoing yeast two-hybrid, specific bait-prey co-transformations, co-immunoprecipitation, in silico transcriptional and proteomic profiling experiments support findings from previous TEM/SEM studies, and strongly suggest an "invasion model" in which CLas/CLso transforms the endocytic/exocytic host pathways to facilitate internalization, infection, and circulation in the psyllid host and vector.
- Based on proteome and transcriptome expression profiling, and/or yeast-two hybrid analyses in conjunction with a literature review of other pathosystems, 31 genes were selected as candidates for analysis. RNAi of 3 of these genes have caused significant psyllid mortality, compared to untreated controls whereas 12 genes have shown some reduced CLso transmission. All have predicted functions that corroborate the invasion model.
- In order to find additional targets for RNAi analysis, 10 putative phage-encoded effectors are being analyzed by expression profiling in psyllid adults and nymphs and an RNASeq study was conducted using CLas-infected and -uninfected ACP instars and adults.
- A system for screening molecules for binding to psyllid digestive tract epithelium has identified several candidate hexameric peptide ligands that bind specifically at submicromolar concentrations with different binding kinetics and also a potent bactericidal peptide. These sequences are ideal for the design of single gene products that could be used to block the ACPs ability to acquire/transmit CLas when expressed either within citrus or ACP.
- CLas acquisition transmission studies demonstrate that CLas multiplies in both nymphs and adults of ACP, but attains much higher levels in a shorter period of time post-acquisition when acquired by nymphs than when acquired by adults, and that adults may require longer access to infected plants compared to nymphs for CLas to reach higher levels in the psyllid.
- A comprehensive analysis of the ultrastructure of the Asian citrus psyllid stylet sheaths, salivary glands and alimentary

canal was completed.

• Single chain antibody fragments (ScFvs) encoding genes have been isolated and are expressed in transgenic citrus and are being evaluated for their effects on CLas acquisition and transmission.

Driver System - A new trait will not spread efficiently upon release within an existing population without a genetic bias of some kind. The driver is the medium of spread of the introduced phenotype--lack of CLas transmission. The drivers under investigation are viral, endosymbiont and chromosomal.

• An ACP picorna-like virus (DcPLV) was discovered in a worldwide search of ACP collections and is a leading candidate vector that might be of use for a paratransgenesis delivery system. Efforts continue to clone the complete genome as cDNA.

• Other ACP viruses being assessed as potential vehicles for transgenesis include ACP densovirus (DcDNV) and Diaphorina citri flavi-like virus (DcFLV). Unlike for DcPLV, there is evidence that these three viruses are in some, but not all, U.S. ACP populations.

• Experiments are also in progress to determine to determine if infection of insect cell lines, as opposed to whole insects, may offer a rapid approach for evaluating the efficacy of various effectors and if a well characterized ssRNA nodavirus (Flock House Virus) may be useful for this purpose.

• The genetic diversity of Wolbachia in ACP, was compared across selected gene sequences. Two wDi strains were detected among the samples tested. Some populations were co-infected with multiple wDi strains. These findings suggest that Wolbachia-induced cytoplasmic incompatibility may exist in ACP.

• ACP was successfully transfected with a supergroup A Wolbachia of Drosophila melanogaster by microinjection. Due to the low number of individuals produced, establishing co-infected isolines for use in experiments is ongoing.

• Proof of concept has been established for several chromosomal-based gene drive systems for population replacement in the psyllid. DNA vectors for a preferred system, engineered translocations, have been constructed and implemented in a model system predicted to yield a relatively high threshold system that will feature genetic containment and likely public acceptance advantages.

• This system shows great potential for ACP-HLB control because it should be readily transferrable once ACP can be transformed and is robust to mutations anticipated to inactivate drive while genes of interest can be easily linked to the translocation breakpoint.

• Use of this technology in ACP requires a transgenesis system of gene constructs and transformation of the psyllid germline. This priority is being pursued with both embryo injection and injection of adult males and females using a variety of transfection reagents and methods including the gene gun and electromagnetic rail gun. A number of constructs that carry different promoters, a reporter gene and a source of transposase have been generated and injected into nymphs and adults. There is evidence for transient expression in somatic tissues, but no evidence for germline transmission yet. Additional efforts include attempts to modify the psyllid genome using Cas9-based direct integration of foreign DNA at DNA breaks created by direct injection of a Cas9- and guide RNA-encoding plasmid.

Diffusion - Once a nuPsyllid population is developed, its successful use will depend on series of factors based on the overall phenotype and fitness of the population in the environment and most importantly, will depend on human adoption, including the behavior of regulatory agencies, growers and consumers. All of these attributes must be modeled accurately for a nuPsyllid release to be used effectively. As for any other innovation, diffusion is the rate of change.

Several aspects of the technical and communication plan for diffusion of this proposed HLB solution can be addressed most effectively only when an actual candidate nuPsyllid is available for release. The ability to rear, release and monitor psyllids has been initiated and is of immediate use in HLB disease management applications outside of this proposal.

• USDA-APHIS field response personnel in Southern California were interviewed to capture factors affecting the success of regulatory programs and to transfer this information to the nuPsyllid modeling team.

Super-stimulant attractants based on methyl-salicylate and methyl-jasmonate are under development to improve trapping efficiencies to monitor psyllid populations.

What opportunities for training and professional development has the project provided?

Elements of this project are being conducted in University and USDA laboratories in a number of states. These sub-projects are providing considerable professional training to undergraduate and graduate students through direct involvement in the nuPsyllid project. In addition, the project employs a number of Post-Doctoral trainees in the labs, whose contributions to the research objectives serve also to provide them additional professional training. All involved in this project are being exposed to the approaches and mechanics of team research on a large scale, and team meetings involve shared experience on how component research objectives fit into the larger picture.

B Falk Team:

Emilyn Matsumura was a visiting graduate student from Brazil. She worked on this project for 1 year from July 1, 2015 to June 30, 2016.

Luca Nerva is a visiting graduate student from Italy. He is currently working on this project, he arrived in February, 2016.

B. Hay Team:
Omar Akbari, a former postdoctoral fellow funded by this work is now an Assistant Professor in the department of Entomology, UC Riverside.
Tobin Ivy, a graduate student funded by this work, has advanced to candidacy (achieved the equivalent of a Masters degree), and is in progress on his PhD.

N. McRoberts Team:
Brianna McGuire (Junior Specialist) and Carla Thomas (Senior Analyst) have been added to the project team at UC Davis. Brianna is helping with statistical analysis of psyllid catch data and is learning relevant programing skills for integrating R and GIS as part of this work. Carla will work on the cognitive map modeling of the social risks for diffusion. She has been learning cognitive mapping theory to prepare for this and more recently has started to familiarize herself with the software we currently use for capturing and representing cognitive maps.

How have the results been disseminated to communities of interest?

The annual Team meeting was held on May 9th, 2016 at the Citrus Research and Education Center in Lake Alfred, FL including stakeholders, administrative management and advisors. This was a great opportunity for the Driver and Effector teams, who are primarily involved in attempting to isolate or build a nuPsyllid colony to present results to those responsible for the ultimate downstream release and adoption of this technology. Presentations on the project goals and objectives, as well as progress to date have occurred at meetings of the citrus growers in California, Texas and Florida numerous times during the year.
There is a substantial effort to rear and release any type of nuPsyllid under development:

- Florida, Texas, and California will each develop and maintain its own colony to provide nuPsyllids, if available, for initial greenhouse studies and pilot field releases within its borders. The decision as to where to house nuPsyllid colonies within each state will be likely have to be made at several administrative levels.
- Regulatory agencies will likely require that nuPsyllid colonies be housed in a controlled/quarantine facility. Potential sites in each state were identified.
- An estimated population size for a nuPsyllid required for testing cannot be provided until the driver mechanism is selected. The effector mechanism may have associated fitness costs, as well, and these will have to be figured into rearing effort estimates.
- The initial plan is to piggyback nuPsyllid rearing efforts onto that of the existing parasitic wasp programs (Tamarixia) for initial testing with care to control for Tamarixia contamination.
- Modeling efforts to include better climate suitability GIS layer for ACP are in progress.
- An initial economic model was developed and captures the longer-term effects of HLB on the citrus supply response and the unique complexity associated with new planting decisions for this type of perennial crop.

Details of the nuPsyllid project have been made available for public consumption through inclusion in trade journal articles, through the nuPsyllid web page and through other mechanisms, including newsletters of the CRDF.

What do you plan to do during the next reporting period to accomplish the goals?

At the Annual Team meeting the entire group of participants and stakeholders reviewed budget performance against milestones and adjusted spending plans towards the priority objectives for the remaining term of the grant. The entire group discussed revisions to focus the current technical plan based on the progress achieved, as well to address budget to plan variances. These contract modifications are now being implemented. All of the budget modifications are under the proposed scope of work. The administrative team has in several cases been able to propose redirecting some cash flow from within the program toward these more recently focused objectives. The priorities in the coming year are:

- accelerate development of a viral vector based on DCPLV or any other of the candidate viral vectors because this is likely to be the first tool for genetic manipulations and would be immediately useful for effector prioritization;
- analyze the phenotypes of non-native Wolbachia introduced into ACP;
- develop ACP transformation capacity at any level of efficiency because of the impact of success with this bottleneck on the ability to create the desired nuPsyllid colony;
- engage the grower community in a broad educational outreach to raise awareness of the alternatives for genetic technologies in the management of HLB.

Participants

Actual FTE's for this Reporting Period

Role	Non-Students or faculty	Students with Staffing Roles			Computed Total by Role
		Undergraduate	Graduate	Post-Doctorate	
Scientist	4.3	0	0	0	4.3
Professional	0.5	0	0	0	0.5
Technical	12.3	0	0	0	12.3
Administrative	0.3	0	0	0	0.3
Other	0	0	0	0	0
Computed Total	17.4	0	0	0	17.4

Student Count by Classification of Instructional Programs (CIP) Code

{NO DATA ENTERED}

Target Audience

Target audiences include the primary benefactors of the research, the U.S. citrus growers. In addition, the target is the scientific community who is engaged in developmental research that has allowed this research project to be envisioned, and on whose progress we will continue to move forward. The general public is a target of our outreach. As consumers, they are interested and concerned about how research solutions are implemented to solve practical problems, and have shown interest in the foundations of this research project. Finally, policy-makers who often are involved in funding research for Florida citrus, need to be appraised of the project, its goals, and expectations that come from progress. The Outreach Team has determined that 1) because the effector and driver systems are all progressing equally it will not be possible to eliminate one or more from the outreach efforts and 2) an educational program should focus on the context of genetic technologies in general so that the nuPsyllid option for disease management is contrasted for example with a genetically modified citrus host and other technology options. Together with molecular biologist and extension expert Peggy Lemaux, Powerpoint presentation materials based on 3 grower interview events in California have been drafted, adapted as handouts and incorporated into the website "Science for Citrus Health."

Products

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Published	2014	YES

Citation

Fisher, T.W., and Brown, J.K., 2014. Transcriptomic and proteomic analysis of Candidatus Liberibacter asiaticus and solanacearum during psyllid infection and vector-mediated transmission. APS-CPS Joint Meeting, Minneapolis, MN. August 9-13, 2014.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Published	2016	YES

Citation

Cicero, J. M., Fisher, T.W., and Brown, J.K. 2016. Localization of ‘Ca. Liberibacter solanacearum’ and evidence for surface appendages in the potato psyllid vector. Phytopathology 106:142-154.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Published	2015	YES

Citation

Russell, C.W. and K.S. Pelz-Stelinski. 2015. Development of an artificial diet and feeding system for juvenile stages of the Asian citrus psyllid, Diaphorina citri. Entomologia Experimentalis et Applicata 154:171-176

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Published	2015	YES

Citation

Hoffmann M., M.R. Coy, H.N. Kingdom-Gibbard, and K.S. Pelz-Stelinski. 2014. Wolbachia infection density in populations of the Asian citrus psyllid (Hemiptera: Liviidae). Environmental Entomology 43: 1215-1222.

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2015	YES

Citation

Pelz-Stelinski, K.S. 2015. Battling the Asian citrus psyllid and citrus greening in Florida. 8th International Integrated Pest Management Symposium. 23-26 March, Salt Lake City, UT

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2015	YES

Citation

Hoffmann, M., C. Russell, M. Coy, and K.S. Pelz-Stelinski. 2015. Endosymbiotic control of the Asian citrus psyllid, Diaphorina citri) (Hemiptera: Liviidae): Diversity and Ecology of Wolbachia in Florida ACP Populations. The 4th International Research Conference on Huanglongbing, 8-13 February, Orlando, Florida.

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2015	YES

Citation

Hoffmann, M., M. Coy, C.W. Russell, and K.S. Pelz-Stelinski. 2014. Molecular characterization and ecological survey of Wolbachia in Florida Diaphorina citri (Hemiptera: Liviidae) populations. The 62nd Annual Meeting of the Entomological Society of America, 16-19 November, Portland, Oregon.

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2015	YES

Citation

Pelz-Stelinski, K.S. 2015. Incapacitating psyllids to host the greening bacteria or transmit the disease. International Research Conference on Huanglongbing Grower Day. Florida State College, Avon Park, FL. South 7 April (370 Attendants).

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2015	YES

Citation

Pelz-Stelinski, K.S. 2015. Developing new solutions for HLB management. Florida Citrus Growers’ Institute. Citrus Research and Education Center, Lake Alfred, FL. 12 March (309 Attendants).

Type	**Status**	**Year Published**	**NIFA Support Acknowledged**
Journal Articles	Published	2014	YES

Citation

Fisher, T.W., M. Vyas, R. He, W. Nelson, J.M. Cicero, M. Willer, R. Kim, R. Kramer, G.A. May, J.A. Crow, C.A. Soderlund, D.R. Gang, and J.K. Brown. 2014. Comparison of potato and Asian citrus psyllid adult and nymph transcriptomes identified vector transcripts with potential involvement in circulative, propagative Liberibacter transmission. Pathogens 3: 875-907; doi: 10.3390/pathogens3040875.

Type	**Status**	**Year Published**	**NIFA Support Acknowledged**
Journal Articles	Published	2015	YES

Citation

Vyas, M., Fisher, T., He, R., Nelson, W.H., Yin, G., Cicero, J.M., Willer, M., Kim, R., Kramer, R., May, G.A., Crow, J.A., Soderlund, C.A., Gang, D.R., and Brown, J.K. 2015. Asian citrus psyllid expression profiles suggest Candidatus Liberibacter asiaticus-mediated alteration of adult nutrition and metabolism, and of nymphal development and immunity. PLoS One 10(6), e0130328.

Type	**Status**	**Year Published**	**NIFA Support Acknowledged**
Journal Articles	Published	2016	YES

Citation

Cicero, J.M., Fisher, T.W., Qureshi, J.A., Stansly, P.A., and Brown, J.K. 2016. Colonization and intrusive invasion of potato psyllid by ‘Ca. Liberibacter solanacearum’. Phytopathology ‘First look: doi: http://dx.doi.org/10.1094/PHYTO-03-16-0149-R’.

Type	**Status**	**Year Published**	**NIFA Support Acknowledged**
Journal Articles	Published	2016	YES

Citation

Ammar, E.D., Ramos, J.E., Hall, D.G., Dawson, W.O. and Shatters Jr, R.G., 2016. Acquisition, Replication and Inoculation of Candidatus Liberibacter asiaticus following Various Acquisition Periods on Huanglongbing-Infected Citrus by Nymphs and Adults of the Asian Citrus Psyllid. PLoS One, 11(7), p.e0159594.)

Type	**Status**	**Year Published**	**NIFA Support Acknowledged**
Journal Articles	Published	2015	YES

Citation

Ammar, E.D., Hall, D.G. and Shatters, R.G., 2016. Ultrastructure of the salivary glands, alimentary canal and bacteria-like organisms in the Asian citrus psyllid, vector of citrus huanglongbing disease bacteria. Journal of Microscopy and Ultrastructure; http://www.sciencedirect.com/science/article/pii/S2213879X16000067

Type	**Status**	**Year Published**	**NIFA Support Acknowledged**
Journal Articles	Published	2015	YES

Citation

Ammar, E.D., Hall, D.G. and Shatters Jr, R.G., 2015. Ultrastructure and development of the new stylets inside pre-molting first instar nymphs of the Asian citrus psyllid Diaphorina citri (Hemiptera: Liviidae). Florida Entomologist, 98(1), pp.373-376.)

Accession No. 230893 **Project No. FLAW-2012-01527**

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Published	2016	YES

Citation

Nouri, S., Salem, N., Nigg, J. C., Falk, B. W. 2016. Diverse array of new viral sequences identified in worldwide populations of the Asian citrus psyllid (Diaphorina citri) using viral metagenomics. J. Virology 90: 2434 – 2445).

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Awaiting Publication	2016	YES

Citation

Nigg, J.C., Nouri, S., and Falk, B.W. 2016. Complete genome sequence of a putative densovirus of the Asian citrus psyllid, Diaphorina citri. Genome Announcements, In Press.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Awaiting Publication	2016	YES

Citation

Nouri, S., Salem, N., Falk, B.W. 2016. Complete genome sequence of Diaphorina citri Associated C virus (DcACV), a novel putative RNA virus of the Asian citrus psyllid. Genome Announcements, In Press.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Published	2016	YES

Citation

Matsumura, E. E., Nerva, L., Nigg, J. C., Falk, B. W., Nouri, S. Complete genome sequence of the largest known flavi-like virus, Diaphorina citri Flavi-Like Virus, a novel virus of the Asian citrus psyllid, Diaphorina citri. American Society for Microbiology; Genome Announcements. In Press. Volume 4 Issue 5, Sept/Oct 2016

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2016	YES

Citation

Nouri, S. Diverse array of new viral sequences identified in worldwide populations of the Asian citrus psyllid (Diaphorina citri) using viral metagenomics. Society for Invertebrate Pathology, July 27, 2016, Tours, France.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Published	2016	YES

Citation

Chu, C., M. Hoffman p, and Pelz-Stelinski, K.S. 2016. Inter-population variability of endosymbiont densities in the Asian citrus psyllid, Diaphorina citri. Microbial Ecology. 71: 999-1007.

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2016	YES

Citation

Hay, B. Scheduled presentation at International Congress of Entomology, 2016

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2015	YES

Citation

Patt, J.M. Annual Meeting of the Entomological Society of America, Minneapolis, MN, November 2015

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2016	YES

Citation

Patt, J.M. Annual Florida Citrus Show, Fort Pierce, FL, January 2016

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2016	YES

Citation

Patt, J.M. Annual Meeting of the American Chemical Society, Philadelphia, PA, August 2016

Type	Status	Year Published	NIFA Support Acknowledged
Conference Papers and	Other	2016	YES

Citation

Patt, J.M. International Congress of Entomology, Orlando, FL, September 2016

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Submitted	2016	YES

Citation

Mitchell, P.D., McRoberts, N., Brown, Z. (submitted). Economic and social factors in deploying gene drive technology. Journal of Responsible Diffusion.

Type	Status	Year Published	NIFA Support Acknowledged
Journal Articles	Published	2015	YES

Citation

Citrograph: Teiken, C., P. Lemaux, B. Grafton-Cardwell, and N. McRoberts. 2015. Genetic Engineering to protect citrus from HLB Citrograph 6(1): 24-31.

Other Products

Product Type

Other

Description

Technology assessments have suggested a near term application of this research for the protection of new plantings. This concept “Psyllid Shield” is being evaluated for field trials. While it is not full insect replacement, it is based in part on research progress in the search for Effectors. CRDF has supplemented funding to model and assess the minimum field trial plot size and time required to demonstrate efficacy in protecting new solid block plantings from HLB with RNAi. Extensive modeling efforts have been accomplished to date and a regulatory strategy is in progress. RNAi might be delivered genetically by CTV vectors or through transgenic citrus. If exogenous delivery technology is developed and the cost of goods is inexpensive, it may be possible to delivery the actives exogenously. There is one licensee interested in pursuing this approach and discussions with others are progressing. Additional RNAi molecules and peptides were discovered and characterized in the course of this research in the past year.

Changes/Problems

Despite the complexity of communication and reporting between the 15 institutions, the project is on target with its timeline of objectives. There are no major changes to the project in terms of approach and none are anticipated. However, in this continuation year 4 planning we have spent significant administrative effort in cash flow management to direct funds to the most important current objectives within the original proposed scope of work. CRDF, as the primary on this project, requires quarterly written progress reports on its funded projects, and we have included this term in all nuPsyllid project subcontracts. Consequently, we are receiving and posting these brief progress reports that are generated by each participant and coordinated through the team leaders up to a collective quarterly progress report submitted by the Project Director. We feel this keeps the team members focused on the goals and allows us to communicate regularly on progress. The gene drive system could likely be used to deliver a effector mechanism and create the desired nuPsyllid population if ACP could be transformed and therefore this is one current priority area of emphasis.