CITRUS ADVANCED TECHNOLOGY PROGRAM

QUARTERLY & FINAL PROGRESS REPORT FORM: Control of Citrus Greening, Canker & Emerging Diseases of Citrus

PROJECT QUARTER END June 2024 ◉ *Quarterly Report* ○ *Final*

Proposal Title

Getting to the root of the problem: Managing Diaprepes root weevil on trees with HLB

Today's Date 07/14/2024

Sponsoring Organization (drop-down) Citrus Research and Development Foundation

Category (drop down) Other

ABSTRACT (Executive Summary-public report-do not disclose proprietary information or intellectual property)

1. Please state project objectives and what work was done this quarter to address them:

Objective 1: Evaluate currently available registered insecticides in Florida citrus against DRW

In this quarter, we have continued to evaluate the use of Btt against Diaprepes larvae. A laboratory experiment was conducted in preparation for field experiments to determine the effects of concomitant infection of insects by Btt and entomopathogenic nematodes. To optimize conditions, DRW larvae of two ages (6 or 12 wk old) were fed on artificial diet inoculated with either of three different concentrations of *Bacillus thuringiensis* subsp. t*enebrionis* (Novodor 3% [AI], [30 mg spores and δ-endotoxin crystals per ml product]) (0, 300, or 3000 ppm) for 10 or 21 days. After Btt feeding exposure, beetles were transferred individually to soil, which was inoculated with the entomopathogenic nematode (EPN), *Steinernema riobrave*, at a rate of 15 injective juveniles (IJs) /cm^2. The mortality of 6- or 12-week-old DRW larvae was recorded after three days of exposure to the EPNs based on symptoms of infection, which are presented in Fig. 1. Cadavers of dead DRW larvae were transferred individually to White traps to confirm infection with EPN IJs by detecting emergence of nematodes from killed larvae.

No mortality of DRW larvae was observed in the control. Mortality of DRW larvae that were pre-exposed to Btt at both concentrations evaluated (75-87%) was higher than that of larvae exposed to diet free of Bt (54%). There was a trend suggesting that larvae exposed at the higher rate of Bt exhibited more specific symptoms of nematode infection than larvae exposed to the lower rate of Bt. EPN infection of DRW larvae exposed to the higher rate of Btt was higher after 10 (89%) than 21 (72%) days of exposure to Btt. Six-week-old larvae were much more susceptible to EPN than 12-week-old larvae, but are difficult to evaluate under in field conditions due to their small size. Therefore, based on the results of this laboratory investigation, we will expose 12-week-old DRW larvae to Btt at a concentration of 3000 ppm concentration for 10 days in upcoming field trials.

Also, nematode symptoms and the presence of infective juveniles in the White traps indicated that larvae infected with Bt provided better conditions for nematode population growth compared to control insects that were not exposed to Btt. More IJs were produced in larvae exposed to 300 ppm of Btt than 3000 ppm Bt. Overall, our results indicate that exposure of DRW larvae to Btt renders the beetle larvae more susceptible to EPN infection. These results suggest that combining Btt with EPN may be more effective for EPM management than either factor alone. Furthermore, the results suggest that the use of Btt against DRW larvae may render beetle larvae more susceptible to naturally occurring EPN in citrus groves

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Organization CREC

Sponsor Project Number 22-013

Project Duration (years) 3 *Year of Project* 2

% Completion of Objectives (FDACS requirement) 50.00%

CITRUS RESEARCH AND DEVELOPMENT FOUNDATION, INC.