

**Elvenia J. Slosson Endowment Foundation Final Report
2007 to 2008**

I. Title: Entomopathogenic Nematodes for Complying with Quarantine Restrictions of Nursery Containerized Plants and Controlling *Diaprepes* Weevil Larvae

Summary of the project proposal: The *Diaprepes* root weevil, *Diaprepes abbreviatus*, has been discovered in southern California in a number of localities in Los Angeles, Orange and San Diego counties and is a major threat to citrus orchards, landscape plants and nursery production. This exotic pest probably came into California in infested ornamental plants from Florida. It has over 270 plant species in 59 plant families as hosts with the larval stage spending 8 to 12 months feeding on roots. There are very few control options. In Florida, the entomopathogenic nematodes, *Steinernema riobrave* and *Heterorhabditis indica*, have proven to be effective in sandy soils in citrus orchards. As nurseries use sandy soils for their containerized plants, this situation provides an ideal habitat for the nematodes to infect and kill weevil larvae. Our objectives are to use commercially available nematodes, *S. riobrave* and *H. indica*, to determine whether they can provide complete control of larvae in containerized plants and to compare the efficacy of the commercial nematode products against nematode-infected cadavers. The efficacy of the nematodes will be compared with the synthetic pyrethroid, bifenthrin.

Final report on trials

Summary: Our objectives are to use commercially available nematodes, *S. riobrave* and *H. indica*, to determine whether they can provide complete control of larvae in containerized plants and to compare the efficacy of the commercial nematode products against nematode-infected cadavers. We planned this work for two years, but due to a misunderstanding, did not apply in a separate proposal for the second year of funding. Thus, the work is not complete, but we have made some significant progress toward meeting our stated objective. A series of experiments were conducted testing the efficacy of the entomopathogenic nematode *S. riobrave* in different concentrations alone and in combination with the entomopathogenic fungus *Metarhizium anisopliae* against the invasive weevil pest, *Diaprepes abbreviatus*. Findings to date suggest that the nematodes provide good, but not complete control using standard methods of application. *M. anisopliae* was not effective controlling *D. abbreviatus*, and further seemed to have an antagonistic interaction with the nematodes. We are continuing greenhouse trials currently to address the efficacy of entomopathogenic nematodes applied while inside the insect host cadaver.

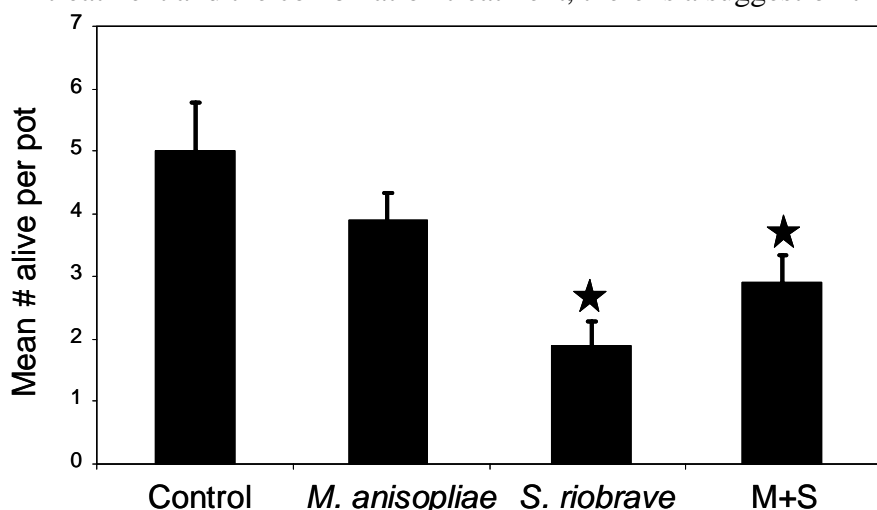
Trial Results: The results from two complete trials are reported here. We attempted several trials where we had difficulties with larval survival in control treatments, invasion of the pots in the trial by ants which killed the *D. abbreviatus* larvae and other problems. Now that we have overcome most of these problems, the trials are going well.

The same methods apply to each of the trials described except for the application of treatments. Indian hawthorn bushes donated from commercial nurseries were used as host plants. They were obtained as bare-root plants and grown in commercial soil-less medium in the quarantine greenhouse facility of the University of California, Riverside. Plants were allowed to grow for 6 weeks before infestation. After 6 weeks, each 1 gallon pot was infested with 10 3rd instar *D. abbreviatus*, after which larvae were allowed to develop and feed on plant roots for 4 weeks before treatments were added. There were 10 pots per treatment.

Results from Trial 1.

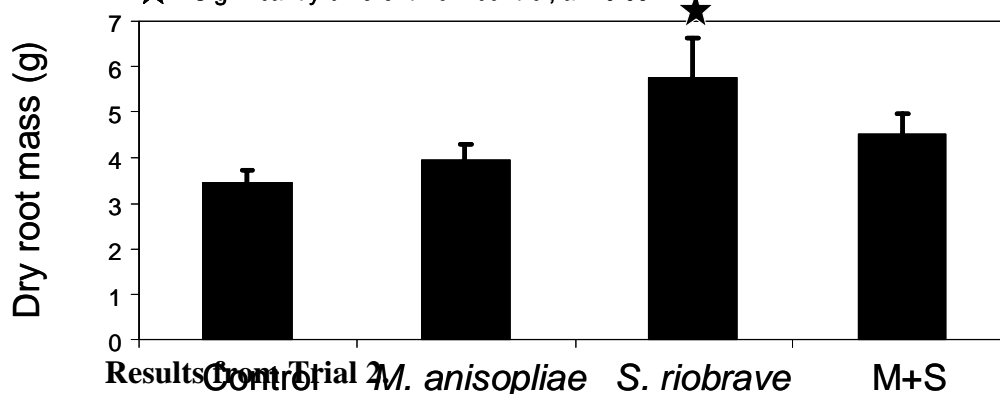
In the first trial, three treatments were compared with the control; 5,000 *S. riobrave* per pot, 0.02 ml *M. anisopliae* per pot, which is approximately the label rate for the product (Tick-Ex, donated by Novozymes, Inc.), and both of the products combined. After 8 weeks, surviving larvae were counted and root mass for each plant was recorded.

Survival of weevils is presented below. *S. riobrave* provided the best control of the treatments tested. Though there was not a significant difference between the nematode treatment and the combination treatment, there is a suggestion that the two materials do not



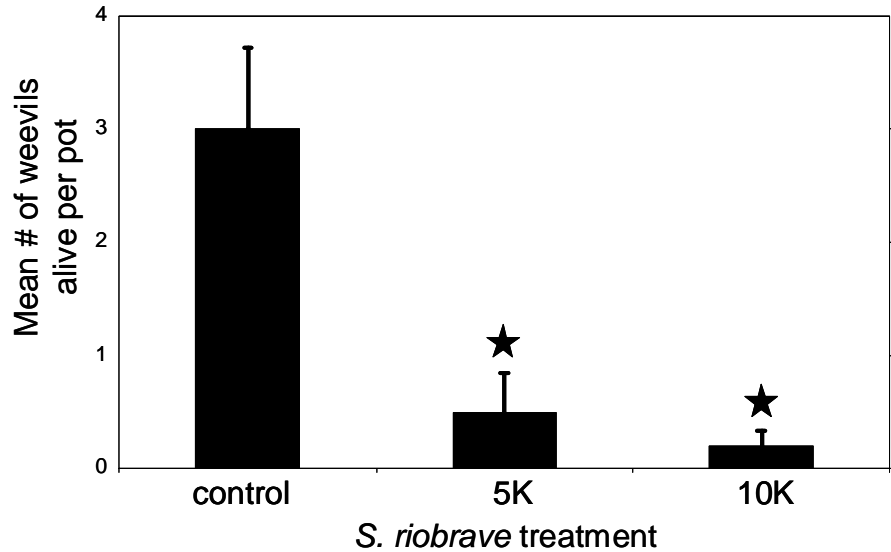
work well combined. We plan to repeat this trial to better document the interaction between the nematodes and the fungus. The root mass data agree with the efficacy data in that the *S. riobrave* plants had a greater mass than any of the other treatments.

★ - Significantly different from control; $\alpha = 0.05$

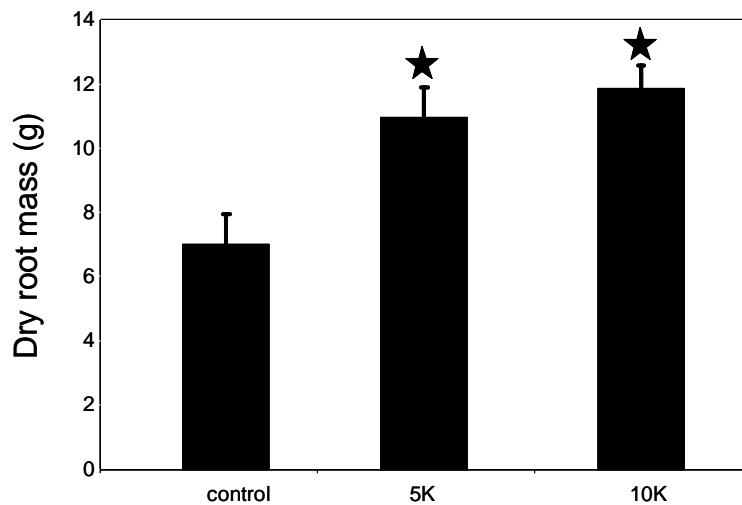


★ - Significantly different from control; $\alpha = 0.05$

In the second trial, three treatments were compared; control, 5,000 *S. riobrave* and 10,000 *S. riobrave*. The same data were collected as in trial 1 and are shown below. Again, *S. riobrave* treatments were effective in controlling *D. abbreviatus*. However, 10,000 IJs did not out-perform the 5,000 IJ treatment. Our next trials will include treatments with entomopathogenic nematodes applied while inside their host cadavers.



★ - Significantly different from control; $\alpha = 0.05$



★ - Significantly different from control; $\alpha = 0.05$

Publications

Spence, K.O., Lewis, E.E., Bethke, J. and Kaya, H.K. Potential for biocontrol of *Diaprepes abbreviatus* larvae in nurseries in southern California. Presented to the 41st Annual Meeting of the Society for Invertebrate Pathology and 9th International Conference on *Bacillus thuringiensis*, August 3-7, 2008 University of Warwick, Coventry, United Kingdom.