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Comparison of Two Steinernematid Species for Control of the Root Weevil *Diaprepes abbreviatus*

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Abstract: *Steinernema carpocapsae* Weiser All strain was compared to *Steinernema riobravis* Cabanillas, Poinar, and Raulston for control of the root weevil, *Diaprepes abbreviatus* (L.), in the laboratory and in potted citrus. In the laboratory bioassay, *D. abbreviatus* larvae were exposed to 30, 60, and 120 nematodes/cm^3 in sand. Insect mortality 1 week after application was greater ($P \leq 0.05$) for *S. riobravis* than for *S. carpocapsae* in the laboratory bioassay. In the greenhouse bioassay, *D. abbreviatus* larvae were exposed to 3 and 9 nematodes per cm^3 of soil in potted citrus. Again, at each rate, mortality was greater ($P \leq 0.05$) in pots treated with *S. riobravis* than in pots treated with *S. carpocapsae.* The results of this study suggest that *S. riobravis* is a better biological control agent against *D. abbreviatus* larvae in potted plants than *S. carpocapsae.*

Key words: biological control, citrus, *Diaprepes abbreviatus,* entomopathogenic nematode, nematode, *Steinernema carpocapsae, Steinernema riobravis,* Steinernematidae.

The citrus root weevil complex consists of five species: the Fuller rose beetle, *Asynonychus godmami* (Crotch); the little leaf notcher, *Artipus floridanus* Horn; the citrus root weevils, *Pachnaeus litus* (Germar) and *P. opalus* (Oliver); and the sugarcane rootstalk borer weevil *Diaprepes abbreviatus* (L.). The life cycle of the five species is similar. Eggs are deposited in the canopy of the tree, and neonate larvae fall to the ground, enter the soil, and feed on roots. Major injury to citrus, sugarcane, ornamental plants, and vegetable crops in Florida and the Caribbean results from larval feeding damage to roots (5,11). *Diaprepes abbreviatus* is potentially the most destructive because it is the largest of the five species (11). Potted citrus is also a major concern to the industry and is considered one of the main methods for movement of *D. abbreviatus.*

Rhabditid nematodes of the family Steinernematidae are obligate parasites of insects that are characterized by a mutualistic relationship with *Xenorhabdus* spp. bacteria. They are lethal to a broad range of economically important insect pests (4,6). The nematode *Steinernema carpocapsae* (Weiser) has been evaluated for control of larvae of *D. abbreviatus* in Florida (9) and Puerto Rico (8). In one study, application of *S. carpocapsae* in the citrus grove reduced *D. abbreviatus, P. opalus,* and *P. litus* adult weevil emergence by 70% compared with the check (10). Subsequently, the commercial product BioVector®, containing the nematode *S. carpocapsae* All strain, was introduced as a biological agent for control of the citrus root weevil complex in Florida.

Recently, *Steinernema riobravis* Cabanillas, Poinar, and Raulston (2) was isolated from the lower Rio Grande Valley in Texas. It is a parasite of the corn earworm, *Helicoverpa zea* (Boddie), and the fall armyworm, *Spodoptera frugiperda* (Smith) (7). This study compares *S. riobravis* with *S. carpocapsae* as biological agents for control of the larvae of *D. abbreviatus* under controlled conditions.

MATERIALS AND METHODS

Nematodes: Steinernema carpocapsae All strain and *S. riobravis* were obtained from Biosys, Palo Alto, California. Additional nematode generations were produced from infected *D. abbreviatus* larvae using the method described by Dutky et al. (3).

Weevil larvae: D. abbreviatus were reared on diet (1). The average weight of the

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3-month-old larvae was 0.516 g (range 0.310–0.901 g).

Laboratory bioassay: Each 3.5-cm-d bioassay cup contained 16 cm^3 (25 g) of sterile dry Astatula fine sand (hyperthermic, uncoated typic quartzipsamments). The moisture of the sand was adjusted to 10% wt/wt with deionized water. A single *D. abbreviatus* larvae was placed in each cup. Nematodes were added at 30, 60, and 120 per cm^3 of sand, and a slice of carrot was placed on the sand as food for the larvae. The cups were maintained at a temperature of 26 C for 1 week. After 1 week, all weevil larvae were dissected to confirm nematode infection. There were 10 cups per replication and eight replications per treatment. A bioassay was conducted simultaneously without food to determine if this was a factor in nematode infection. Also, comparison of the efficacy of F1 and F2 generations of *S. carpocapsae* and *S. riobravis* was done to eliminate variables in shipping, formulation, and storage that might have affected each nematode species differently.

Potted plant bioassay: Sour orange *Citrus aurantium* (L.) seedlings were established in 15-cm-d pots with 2 liters of soil. The potting soil media was three parts Florida peat and one part coarse builder's sand (v/v). Ten *D. abbreviatus* larvae were placed 5 cm below the soil in each pot. After 2 weeks, nematodes were added to the pots at the rate of three and nine nematodes/cm^3 of soil. The study was conducted from October through March with ambient weather conditions (5–28 C), and plants were watered once a week. The soil was removed from the pot after 2–4 weeks and the number of live larvae determined. There were 20 plants per treatment for a total of 100 plants.

Arcsine-transformed data were subjected to an analysis of variance (ANOVA) and means were separated by a Student-Newman-Keuls multiple-range test.

Results and Discussion

Bioassay: The results of the laboratory study comparing *S. riobravis* with *S. carpocapsae* at 30, 60, and 120 nematodes/cm^3 are shown in Table 1. More *D. abbreviatus* larvae were killed by *S. riobravis* than by *S. carpocapsae* at each of the three rates tested ($P \leq 0.05$). In the bioassay that was conducted simultaneously with or without food, it was determined that food was not a factor in infection of weevil larvae. Apparently, nematodes that were consumed with the carrot did not affect ($P \leq 0.05$) the mortality of the larvae. Therefore, food was not included in the laboratory bioassay when nematode generations were compared.

When the parent, F1, and F2 generations of *S. riobravis* and *S. carpocapsae* were compared, mortality by *S. riobravis* was different ($P \leq 0.05$) from *S. carpocapsae* (Table 2). This difference in activity indicates

TABLE 1. Mortality of *Diaprepes abbreviatus* caused by *Steinernema riobravis* or *S. carpocapsae* with and without food in a laboratory bioassay.

Nematode species and treatment	% mortality (nematodes per cm^3)				
	0	30	60	120	Total
S. riobravis					
No food	02	60	57	75	63 a
Food	03	57	63	60	60 a
S. carpocapsae					
No food	03	13	22	35	26 b
Food	02	32	48	27	36 b

Means within the same column followed by the same letter are not significantly different ($P \geq 0.05$; Student-Newman-Keuls multiple-range test).

TABLE 2. Mortality of *Diaprepes abbreviatus* caused by *Steinernema riobravis* or *S. carpocapsae* in a laboratory bioassay.

Nematode species and generation	% mortality (nematodes per cm^3)				
	0	30	60	120	Total
S. riobravis					
Parent	0	43	50	50	48 a
F1	0	78	68	80	75 b
F2	7	55	63	80	66 b
S. carpocapsae					
Parent	5	20	23	25	23 c
F1	0	20	35	48	34 c
F2	0	30	48	20	33 c

Means within the same column followed by the same letter are not significantly different ($P \geq 0.05$; Student-Newman-Keuls multiple-range test).

TABLE 3. Mortality of *Diaprepes abbreviatus* larvae caused by *Steinernema riobravis* or *S. carpocapsae* in potted citrus.

Nematode species	Nematodes per cm^3	Larvae per plant		% mortality
		Mean	Range	
S. riobravis	3	1.4	0–5	86 a
	9	2.3	0–7	77 a
S. carpocapsae	3	6.8	2–10	32 bc
	9	5.8	2–10	42 b
Check	0	7.8	5–10	23 c

Means within the same column followed by the same letter are not significantly different ($P \geq 0.05$; Student-Newman-Keuls multiple-range test).

that *S. riobravis* is a more virulent biocontrol agent compared with *S. carpocapsae.*

Potted plant bioassay: This study evaluated treatment effects under field conditions for plants with soil attached to the roots. Mortality of *D. abbreviatus* larvae in plants treated with *S. riobravis* was greater ($P \leq 0.05$) than for plants treated with *S. carpocapsae* (Table 3). Mortality of larvae in the check plants was apparently due to cannibalism by other larvae.

The results of this study suggest that the entomopathogenic nematode, *S. riobravis,* is a more effective biological control agent against *D. abbreviatus* larvae than *S. carpocapsae.* This was evident in the laboratory bioassay and in potted citrus. Citrus was the larval host used in this study; however, the data should apply to other potted plant species that are infested with *D. abbreviatus.*

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