Control of Diaprepes abbreviatus (Coleoptera: Curculionidae) with Steinernema riobrave (Rhabditida: Steinernematidae) in Plasticulture Florida Strawberry

Authors: Renkema, Justin M., Krey, Karol L., and Lahiri, Sriyanka

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Control of *Diaprepes abbreviatus* (Coleoptera: Curculionidae) with *Steinernema riobrave* (Rhabditida: Steinernematidae) in plasticulture Florida strawberry

Justin M. Renkema[1,2], Karol L. Krey[1,3], and Sriyanka Lahiri[1,]*

Abstract

Diaprepes abbreviatus L. (Coleoptera: Curculionidae) is an occasional root pest of plasticulture strawberry in central Florida, USA. There are few chemical insecticide options for larval *D. abbreviatus* in strawberry. Therefore, we tested soil-applied aqueous *Steinernema riobrave* Cabanillas, Raulston and Poinar (Rhabditida: Steinernematidae), which is used for *D. abbreviatus* control in citrus. When *S. riobrave* was applied (100 infective juveniles per cm^2) to the root zones of plants in a *D. abbreviatus*-affected area of a commercial strawberry field, less than 12% of plants were severely wilted or dead 17 d after treatment, whereas 23% of plants in control plots were wilted or dead. In research plots, peripheral plants and a central plant in each plot were infested with 4 late-instar *D. abbreviatus* and treated with 1 or 2 applications of *S. riobrave* (25 infective juveniles per cm^2), 1 application of imidacloprid or water (control). Dead *S. riobrave*-infected *D. abbreviatus* larvae were recovered from plots to which *S. riobrave* was applied, but there was no effect of treatment on numbers of live larvae recovered 1, 2, and 3 wk post-application in the peripheral plants. At the end of the experiment, no live *D. abbreviatus* larvae were recovered from the central plants or the plants proximal to the central plants in plots treated once or twice with *S. riobrave*. Plant wilting and death due to larval *D. abbreviatus* root feeding was minimal (averages of 1–1.4 on a 1–5 rating scale) in plants proximal and distal to the central *D. abbreviatus*-infested plant. Treatment did not affect plant wilting and death rates in the proximal plants, but death of distal plants occurred only in control plots. Our results showed *S. riobrave* infected and killed late-instar *D. abbreviatus* in plasticulture Florida strawberry, and further research should be conducted to optimize *S. riobrave* applications and develop it into a management strategy for *D. abbreviatus*.

Key Words: entomopathogenic nematodes; imidacloprid; plant damage; larval movement; rescue application

Resumen

Diaprepes abbreviatus L. (Coleoptera: Curculionidae) es una plaga ocasional de la raíz de la fresa en plasticultura en el centro de la Florida, EE. UU. Hay pocas opciones de insecticidas químicos para las larvas de *D. abbreviatus* en fresa. Por lo tanto, probamos *Steinernema riobrave* Cabanillas, Raulston y Poinar (Rhabditida: Steinernematidae) acuoso aplicado al suelo, que se utiliza para el control de *D. abbreviatus* en cítricos. Cuando se aplicó *S. riobrave* (100 juveniles infecciosos por cm^2) a las zonas de las raíces de las plantas en un área afectada por *D. abbreviatus* de un campo comercial de fresas, menos del 12% de las plantas estaban severamente marchitas o muertas 17 días después del tratamiento, mientras que el 23% de las plantas en las parcelas de control estaban marchitas o muertas. En las parcelas de investigación, se infestaron plantas periféricas y una planta central en cada parcela con larvas del 4 estadio tardíos de *D. abbreviatus* y se trataron con 1 o 2 aplicaciones de *S. riobrave* (25 juveniles infecciosos por cm^2) y 1 aplicación de imidacloprid o agua (control). Se recuperaron larvas muertas de *D. abbreviatus* infectadas con *S. riobrave* de las parcelas a las que se aplicó *S. riobrave*, pero no hubo efecto del tratamiento sobre el número de larvas vivas recuperadas 1, 2, y 3 semanas después de la aplicación en las plantas periféricas. Al final del experimento, no se recuperaron larvas vivas de *D. abbreviatus* de las plantas centrales o de las plantas próximas a las plantas centrales en parcelas tratadas una o dos veces con *S. riobrave*. El marchitamiento y muerte de la planta debido a la alimentación de las raíces de las larvas de *D. abbreviatus* fue mínima (promedios de 1–1,4 en una escala de calificación de 1–5) en plantas proximales y distales a la planta central infestada con *D. abbreviatus*. El tratamiento no afectó las tasas de marchitez y muerte de las plantas en las plantas proximales, pero la muerte de las plantas distales ocurrió solo en las parcelas de control. Nuestros resultados mostraron que *S. riobrave* infectó y mató a *D. abbreviatus* en estadio tardío en la fresa en plastificultura en la Florida, y por ello se deben realizar más investigaciones para optimizar las aplicaciones de *S. riobrave* y desarrollarlas dentro de una estrategia de manejo para *D. abbreviatus*.

Palabras Claves: nematodos entomopatógenos; imidacloprid; daño a la planta; movimiento de larvas; aplicación de rescate

*Diaprepes abbreviatu*s L. (Coleoptera: Curculionidae) is a polyphagous root weevil and pest of approximately 300 agricultural, ornamental, and wild plants (Simpson et al. 1996; Mannion et al. 2003). In citrus (*Citrus* spp. L.; Rutaceae), adults may damage trees by scoring young leaves; however, larval root feeding causes significant tree damage, leading to yield reductions and eventually tree death (Woodruff 1985; McCoy 1999; Knapp et al. 2000; Stuart et al. 2006). Adults are active from spring to autumn; an adult female lays an average of 11,000 eggs in her life span (Nigg et al. 2004), and neonates drop from foliage to the soil where they feed on roots and develop over several mo.

[1]University of Florida, Gulf Coast Research and Education Center, 14625 County Road 672, Balm, Florida 33598, USA; E-mails: lahiris@ufl.edu (S. L.)
Current affiliations:
[2]London Research and Development Centre – Vineland Campus, Agriculture and Agri-Food Canada, Vineland Station, Ontario, Canada;
E-mail: justin.renkema@canada.ca (J. M. R.)
[3]ISK Biosciences, 7470 Auburn Road, Suite A, Concord, Ohio 44077, USA; E-mail: kreyk@iskbc.com (K. L. K.)
*Corresponding author; E-mail: lahiris@ufl.edu

Florida, USA, is a major source of winter strawberries (*Fragaria* × *ananassa* Duchesne; Rosaceae) in the USA, with 4,000 planted ha and a production value of nearly $300 million USD (USDA/NASS 2018). Development of effective pest management practices will help enable Florida producers to be viable economically in an increasingly competitive market space (Suh et al. 2017). While the primary arthropod pests in Florida strawberry are twospotted spider mite (*Tetranychus urticae* Koch; Trombidiformes: Tetranychidae), flower thrips (*Frankliniella occidentalis* [Pergande]; Thysanoptera: Thripidae), and *Frankliniella bispinosa* Morgan; Thysanoptera: Thripidae), chilli thrips (*Scirtothrips dorsalis* Hood; Thysanoptera: Thripidae), and fall armyworm (*Spodoptera frugiperda* [J.E. Smith]; Lepidoptera: Noctuidae), secondary pests such as *D. abbreviatus*, occasionally require control (Caruso et al. 2019). Root feeding by larval *D. abbreviatus* causes foliar wilting often followed by plant death. Eggs are laid on young strawberry plants shortly after planting in early autumn by late-flying female *D. abbreviatus*, and damage typically is concentrated in small patches near field edges. However, plant damage was more widespread in a few fields adjacent to old, neglected, or recently uprooted citrus groves (J. Renkema, personal observation). Insecticides are available for weevils and grubs in Florida strawberry (Renkema et al. 2019), but they either are not labelled for application through drip irrigation to control root-feeding larvae or have long pre-harvest intervals not compatible with in-season harvest schedules. For insecticides that may be chemigated, growers typically are not able to restrict applications to areas of a field where damage occurs, and treating entire fields is not economical or sustainable. A tactic for control of *D. abbreviatus* in small areas of strawberry fields is needed.

Diaprepes abbreviatus has been managed successfully with the entomopathogenic nematode *Steinernema riobrave* Cabanillas, Raulston and Poinar (Rhabditida: Steinernematidae) in Florida citrus (Dolinski et al. 2012). *Steinernema riobrave*, an indeterminate forager, was first found in southern Texas, USA, and is acclimated to warm soils (Kaspi et al. 2010). Applications of commercially produced *S. riobrave* have caused larval mortality rates of *D. abbreviatus* in the field of 50% to greater than 90%, depending on application rates and other factors such as soil conditions (Duncan et al. 1996, 2003; Bullock et al. 1999). Over a yr, adult *D. abbreviatus* emergence was reduced by half by *S. riobrave* in one study (Duncan et al. 2007). In citrus, *S. riobrave* is applied to the soil using tractor-mounted sprayers or through drip irrigation (Duncan et al. 1999), but more targeted application methods used in other cropping systems for other entomopathogenic nematode species may be better suited to or adjusted for small applications in strawberry (Shapiro-Ilan & Dolinski 2015). Here we tested aqueous preparations of *S. riobrave* applied as a soil drench around the base of individual strawberry plants, but formulation of entomopathogenic nematodes may affect efficacy and ease-of-application, with pelletized baits showing promise in some scenarios (e.g., Hiltpold et al. 2012).

Our goal was to provide new knowledge on a potential management strategy for *D. abbreviatus* in Florida plasticulture strawberry production. Control methods that are effective, sustainable, reduce insecticide use, and are practical for treating only small, affected areas of strawberry fields are needed for mitigating losses due to *D. abbreviatus* root feeding. Therefore, the objectives of the research were to determine the efficacy of *S. riobrave* for control of late-instar *D. abbreviatus* in combination with determining the movement of larvae among strawberry plants.

Materials and Methods

COMMERCIAL FIELD

In late Dec 2016, wilting plants were noticed in a small area (about 0.15 ha) of a commercial, raised-bed, plasticulture strawberry (cv. 'Radiance') field (27.9386111°N, 82.1836111°W) near Dover, Florida, USA. Large, late-instar *D. abbreviatus* larvae were found in the root zone of wilted plants. To test the efficacy of *S. riobrave* for control of *D. abbreviatus*, 7 plots of 32 plants each (plot = 1 raised bed, 6.5 m long) were marked out in the affected field area. Each plot was randomly assigned a treatment of *S. riobrave* (4 replications) or water (3 replications).

Steinernema riobrave (Nemasys®R, BASF Corp., Florham Park, New Jersey, USA) were prepared in distilled water in the laboratory according to the manufacturer's instructions (rate of 100 IJ per cm^2) and transported to the field in 1 L bottles (Thermo Scientific™ Nalgene™, Thermo Fisher Scientific, Waltham, Massachusetts, USA). The bottles were shaken by hand to redistribute the *S. riobrave* before pouring aliquots into plastic containers (120 mL, Fisherbrand™, Thermo Fisher Scientific, Waltham, Massachusetts, USA). On 3 Jan 2017, the plastic mulch around the base of each plant was pulled back, and *S. riobrave* or water (60 mL aliquots) was poured evenly and slowly onto an 80 cm^2 area of the soil surface around the base of all plants in all plots.

To determine an effect of *S. riobrave*, plants in each plot were categorized as either: (1) healthy, showing no visual wilting symptoms; (2) wilting, with leaves and stems lacking rigidity as result of low turgor pressure; (3) dead, with plants entirely brown; or (4) missing, where empty planting holes occurred in the plastic mulch. Plants were rated pre-treatment (3 Jan) and 10 and 17 d after treatment (13 and 20 Jan). Plants were not removed to determine *D. abbreviatus* mortality at the end of the experiment by request of the grower.

RESEARCH PLOTS

An experiment was conducted during the 2017 to 2018 strawberry season in research plots at the Gulf Coast Research and Education Center, Balm, Florida, USA (27.7619444°N, 82.2272222°W). Bare-root strawberry transplants (cv. 'Radiance') were set on 9 Oct 2017 (38 cm in-row spacing) in 2 rows in a raised, double-pressed, black plastic/virtually impermeable film (Blockade™, Berry Plastics, Evansville, Indiana, USA) mulched beds (1.2 m spacing) that were fumigated with Telone®C-35 (1,3-dicholoroprepene + chloropicrin) at bed formation (early Aug 2017). Plants were overhead irrigated for 7 to 10 d following transplanting and drip fertigated with 0.27 and 0.34 kg N per d from Nov to mid-Jan and mid-Jan through Apr, respectively. Herbicides Round-up® (glyphosate) and Chateau® (flumioxazin) were applied to row aisles before transplanting. Plants received regular applications of fungicides to control prominent strawberry diseases and early-season applications each yr of DiPel® DF (*Bacillus thuringiensis*, subsp. *kurstaki* Berliner; Bacillaceae) to control lepidopteran larvae.

Plots (3 m × 1 raised bed) were arranged in a randomized complete block design with 6 replications in a checkerboard pattern, so that the raised beds adjacent to each plot were unplanted and the total plot area was 12 m × 12 raised beds. In each plot, there were 13 contiguous plants, with 1 infested plant, and 4 peripheral, infested plants, separated from the 13 plants by unplanted space (Fig. 1).

Larval *D. abbreviatus* were from a laboratory colony maintained at the US Department of Agriculture, Agricultural Research Service facility in Fort Pierce, Florida, USA. The larval cohort used in both experiments were from eggs obtained 9 Sep 2017, with neonate larvae transferred to artificial diet in small cups on 15 Sep and older larvae transferred singly to diet cups on 17 Oct. Larvae were removed from diet and placed singly in empty cups, and the cups (n = 4 per plant) were overturned onto the soil around the base of the plants in the evening of 2 Nov. The following morning, the cups were removed and dead larvae that did not burrow into the soil were replaced with healthy larvae. On 14 Nov, dead leaves and ripe and ripening fruit were removed from plants, and ripe fruit was harvested for the duration of the experiment.

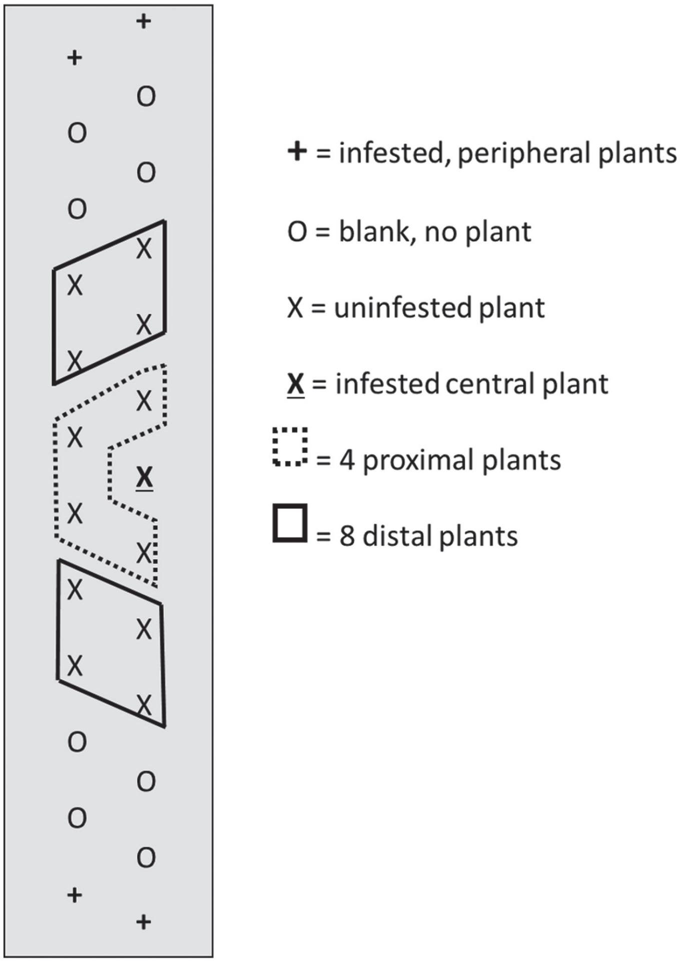

Fig. 1. Strawberry plot layout for testing applications of *Steinernema riobrave* for controlling late-instar *Diaprepes abbreviatus* placed around the root zones of infested plants. Strawberries (38 cm in-row spacing) were planted into raised, earthen beds (1.2 m width) covered with black plastic mulch in research plots at the Gulf Coast Research and Education Center, Balm, Florida, USA.

Treatments were *S. riobrave* applied once, *S. riobrave* applied twice, imidacloprid applied once and water. On 16 Nov, *S. riobrave* (Nemasys®R), prepared in distilled water in the laboratory according to the manufacturer's instructions (25 IJ per cm^2), imidacloprid (Admire®Pro, Bayer CropScience LP, Research Triangle Park, North Carolina, USA; 767 mL per ha) or water were applied as a drench (50 mL) to 100 cm^2 around the base of each plant in a plot using a hand-held CO_2-powered backpack sprayer (0.34 atm.) (R&D Sprayers, Opelousas, Louisiana, USA) and a single wand with the nozzle removed. The end of the wand was used to pull up the plastic around the base of the plant as the drench was being made. *Steinernema riobrave* were prepared and reapplied to plots on 22 Nov.

Effects of drenches were evaluated on 16, 22, and 30 Nov, and 7 and 14 Dec by categorizing each of the 13 contiguous plants in all plots as either (1) healthy, (2) minor wilting, (3) major wilting, (4) mix of brown and green leaves, or (5) dead with all brown leaves. A randomly selected peripheral plant with a core of soil from its central root zone (11 cm diam × 30 cm depth) was removed with a golf-cup cutter from each plot on each of the first 4 evaluation dates. The soil was hand-sorted in the field for live and dead larvae. Dead or moribund larvae were placed in white traps (reference) in the laboratory to determine infection by *S. riobrave*. On 14 Dec, the root-zones of all 13 contiguous plants in each plot were removed and assessed for larvae in the same manner.

DATA ANALYSIS

For the commercial field experiment, the distribution of plant categories was compared between *S. riobrave*-treated and control plots with a Fisher's exact test for small sample sizes. For the research plot experiment, a mixed-model analysis of variance (ANOVA) was used to compare the total number of larvae recovered from peripheral plants on each sample date among treatments, with the block included as a random effect. Data transformation was not necessary. The same model was used to determine the effect of treatments on numbers of live larvae recovered from the central, proximal, and distal plants (14 Dec). For the 4 proximal and 8 distal plants, the average number of larvae per plant were calculated for analysis. The distribution of plant damage categories was compared among treatments separately for each sample date, and central, proximal, or distal plants in each plot with a Fisher's exact test for small sample sizes. All analyses were conducted using JMP® 15.0.0 (SAS 2019) at $\alpha = 0.05$.

Results

COMMERCIAL FIELD

Categorization of strawberry plant health was not affected by *S. riobrave* 10 d after treatment but was affected 17 d after treatment (Table 1). There was 16% more healthy plants in plots with *S. riobrave* compared to those treated with water at 17 d after treatment. Numbers of healthy plants declined by 8% in water-treated plots and increased by 6% in *S. riobrave*-treated plots from pre-treatment to 17 d after treatment (Table 1).

RESEARCH PLOTS

The number of live larvae recovered from peripheral infested plants was similar among treatments pre-application ($F = 0.70$; df = 3,15; $P = 0.57$) and was not affected by treatment at 1 ($F = 1.16$; df = 3,15; $P =$

Table 1. Healthy compared to wilting, dead, or missing strawberry plants due to *Diaprepes abbreviatus* root feeding in a commercial field near Dover, Florida, USA, treated 3 Jan 2017 with soil drenches of *Steinernema riobrave* nematodes (80,000 infective juveniles per plant) (*n* = 128 plants) or water (*n* = 96 plants) and evaluated 10 and 17 d after treatment.

	Pre-treatment		10 d after treatment		17 d after treatment	
Categories	Water	*S. riobrave*	Water	*S. riobrave*	Water	*S. riobrave*
Healthy	78.1 (3.6)	81.3 (1.8)	72.9 (6.8)	83.6 (2.3)	69.8 (5.2)	85.9 (2.0)
Wilted	9.4 (1.8)	11.7 (2.7)	8.3 (3.8)	6.3 (1.3)	5.2 (2.8)	0.8 (0.8)
Dead	8.3 (2.8)	6.3 (2.2)	14.6 (1.0)	7.8 (2.7)	17.7 (4.2)	10.9 (2.7)
Missing	4.2 (2.8)	0.8 (0.8)	4.2 (2.8)	2.3 (1.5)	7.3 (2.1)	2.3 (1.5)
Fisher's exact test	*P* = 0.353		*P* = 0.241		*P* = 0.013	

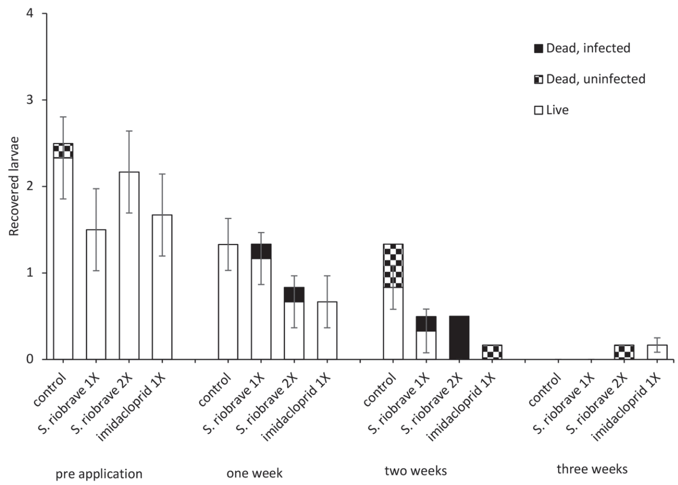

Fig. 2. Mean live (± SEM) and dead *Steinernema riobrave*-infected or uninfected, late-instar *Diaprepes abbreviatus* from soil in the root-zone of a randomly selected peripheral strawberry plant (*n* = 6) (see Fig. 1) after 1 or 2 applications of infective juvenile *Steinernema riobrave* (25 per cm^2 in 50 mL water), 1 application of imidacloprid (Admire® Pro, 767 mL per ha), or water (control) were made 16 and 22 Nov 2017 around the base of each plant. Plants were in raised, plasticulture earthen beds in research plots at the Gulf Coast Research and Education Center, Balm, Florida, USA.

0.36), 2 (F = 2.41; df = 3,15; P = 0.13), or 3 wk (F = 1.00; df = 3,15; P = 0.42) post-application. Dead larvae infected with *S. riobrave* were recovered only from plants to which *S. riobrave* was applied (Fig. 2).

The number of live larvae recovered from centrally infested plants was similar among treatments (F = 1.28; df = 3,15; P = 0.32) and similar for the distal plants (F = 1.43; df = 3,15; P = 0.27). For the proximal plants, larval recovery varied by treatment (F = 6.07; df = 3,15; P = 0.007) with fewer larvae found in plots treated once or twice with *S. riobrave* than in control plots (Table 2).

Categorization of strawberry plant health was not affected by treatments in central, infested plants at pre-application, (P = 0.80), at 1 wk after application (P = 0.31), 2 wk (P = 0.60), 3 wk (P = 0.55), or 4 wk (P = 0.55) (Fig. 3A), or in proximal plants at pre-application (P = 0.90), 1 wk (P = 0.31), 2 wk (P = 0.58), 3 wk (P = 0.75), or 4 wk (P = 0.90) (Fig. 3B). For distal plants, plant health categorization did not differ among treatments at pre-application (P = 0.99), 1 wk (P = 0.26), or 2 wk (P = 0.57), but it did differ at 3 wk (P = 0.01) and 4 wk (P = 0.01) (Fig. 3C).

Discussion

Based on our results, aqueous applications of *S. riobrave* provided a modest level of control of *D. abbreviatus* in plasticulture strawberry production in Florida. In the grower field experiment, there were 17% more healthy and 7% fewer dead plants by 17 d after an application of *S. riobrave* compared to an application of water. In the research plot experiment, no live *D. abbreviatus* were recovered from plants proximal to the *D. abbreviatus*-infested plants when *S. riobrave* was applied to all plants, and almost all dead larvae that were recovered were infected with *S. riobrave*. In proximal and distal plants, there were low rates of plant wilting in all treatment plots, up to about 10% in control plots. Wilting was reduced by 1 or 2 applications of *S. riobrave* in distal plants, but a similar effect was not clear in proximal plants. Applications of *S. riobrave* may not be able to rescue a wilting plant with significant root feeding by *D. abbreviatus*, but preventative applications to visually healthy plants minimized further plant wilting and loss.

There were no significant differences in the efficacy of 1 or 2 applications of *S. riobrave* and 1 application of imidacloprid for control of *D. abbreviatus*. In citrus, the efficacy of imidacloprid and *S. riobrave* on *D. abbreviatus* also was similar (Bender et al. 2014). Admire® Pro (42.8% imidacloprid) is labelled for soil and chemigation application in strawberry for aphids and whiteflies. However, the pre-harvest interval for Admire® Pro is 14 d, limiting use in Florida strawberry to the early season before plants produce ripe fruit, because the harvest interval typically is 3 or 4 d once fruit begins to ripen. Similar efficacy between 1 and 2 applications of *S. riobrave* may be due to most larval infection occurring after the first application, and to high *S. riobrave* survival rates in soil in irrigated strawberry beds protected by black plastic mulch. Entomopathogenic nematodes are susceptible to desiccation and have reduced virulence and viability in dry soil conditions (Patel et al. 1997). Less than optimal soil temperatures may have reduced the efficacy of *S. riobrave*, because average hourly Jan nighttime soil temperatures under black plastic in strawberry root zones were less than 21 °C (Deschamps et al. 2019), and applications in citrus are recommended when soil temperatures are consistently above 21 °C (Duncan & Mannion 2021).

In plants infested with *D. abbreviatus* (central plants), there were similar numbers of dead and wilted plants in plots with 2 applications of *S. riobrave* as in control plots (mean plant ratings of about 4) by the last sample date, but more healthy plants on average in plots with 1 application of *S. riobrave*. However, there was already 50% plant death in plots with 2 applications of *S. riobrave* before the first application (mean plant rating of about 2.5), which was more than in other treatments. Conversely, plots with 1 application of *S. riobrave* had more dead and wilting proximal plants than other treatments, meaning *D. abbreviatus* larvae may have moved away from central plants to proximal plants more quickly in plots with 1 application of *S. riobrave* than other in other plots. From this experiment, the value of a second application of *S. riobrave* is not clear, but a second application at a longer interval, greater than a wk may be warranted, if wilting progresses to greater levels on plants adjacent to the initially infested and highly wilted plant.

In addition to determining efficacy of *S. riobrave* in plasticulture strawberry, we designed our research plot experiment to assess larval movement of *D. abbreviatus*. In untreated control plots, larval re-

Table 2. Mean (± 95% CI) numbers of late-instar *Diaprepes abbreviatus* recovered from soil in the root-zone of strawberry plants when 1 or 2 applications of infective juvenile *Steinernema riobrave* (25 per cm^2 in 50 mL water), 1 application of imidacloprid (Admire® Pro at 767 mL per ha), or water (UTC: untreated control) were made 16 and 22 Nov 2017 around the base of each plant. Plants were in raised, plasticulture earthen beds in research plots at the Gulf Coast Research and Education Center, Balm, Florida, USA. See Figure 1 for location of central, proximal, and distal plants in plots. Means followed by the same letter are not significantly different (Tukey's HSD test, P < 0.05).

Treatments	Central plant		Proximal plants		Distal plants	
Control	0.111	(0.003–0.378)	0.135	(0.046–0.272) a	0.003	(0.000–0.033)
S. riobrave 1×	0.000	(0.000–0.079)	0.000	(0.000–0.023) b	0.000	(0.000–0.000)
S. riobrave 2×	0.000	(0.000–0.079)	0.000	(0.000–0.023) b	0.003	(0.000–0.033)
Imidacloprid 1×	0.028	(0.000–0.200)	0.007	(0.000–0.056) ab	0.014	(0.000–0.058)

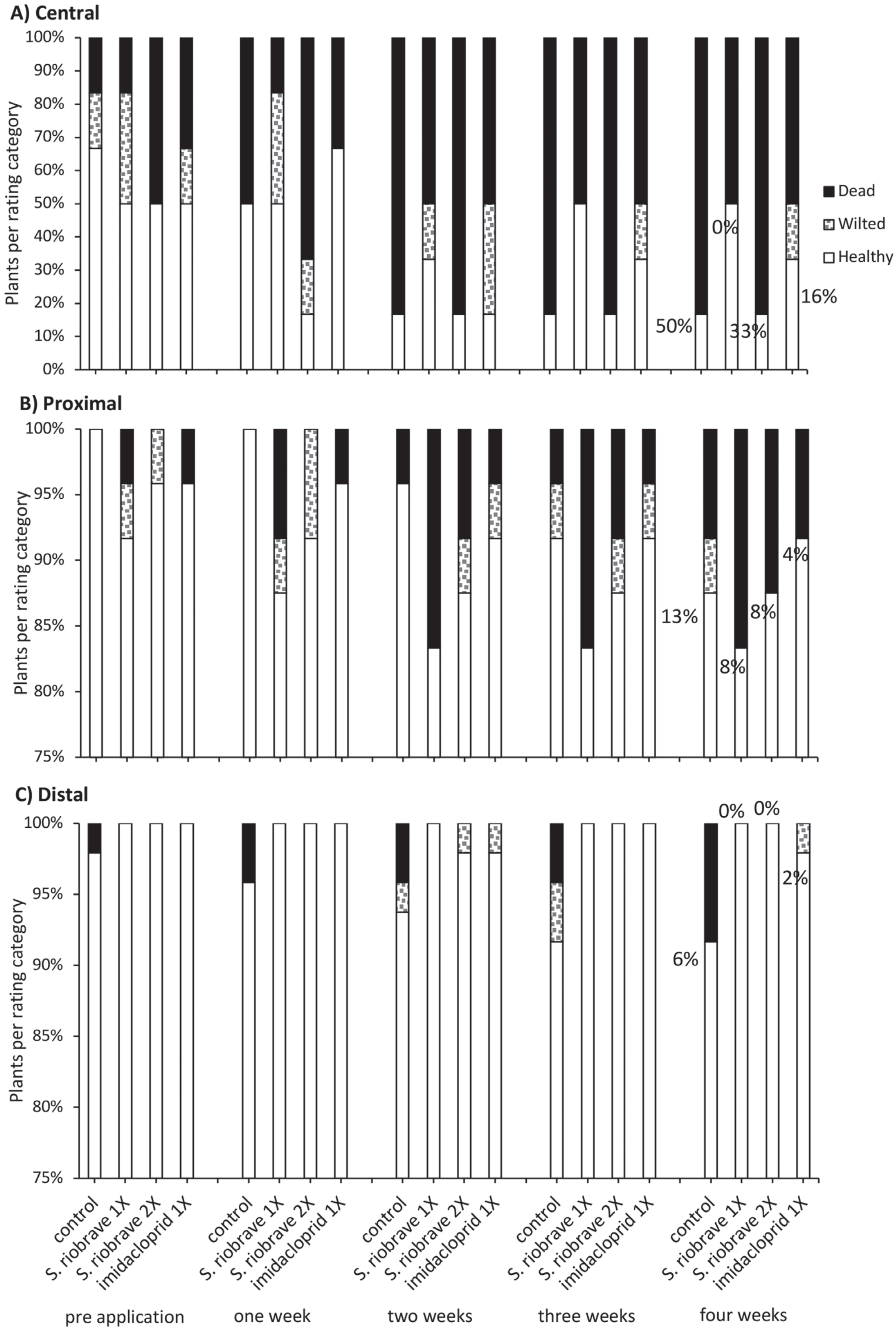

Fig. 3. Progression of damage to strawberry plants (*n* = 13 per plot) when a central plant in each plot was infested with 4 late-instar *Diaprepes abbreviatus*. All plants in a plot received 1 or 2 soil applications of infective juvenile *Steinernema riobrave* (25 per cm^2 in 50 mL water), 1 application of imidacloprid (Admire® Pro, 767 mL per ha), or water (control) on 16 and 22 Nov 2017, and plants were categorized weekly from pre-application (16 Nov) up to 4 wk post-application (14 Dec) using a rating scale of (1) healthy, (2) minor wilting, (3) major wilting, (4) mix of brown and green leaves, or (5) dead with all brown leaves. Strawberry plants were in raised, plasticulture earthen beds in research plots at the Gulf Coast Research and Education Center, Balm, Florida, USA.

covery after 7 wk (2 Nov to 14 Dec) was similar between central and proximal plants, which were about 40 cm apart. Movement likely was motivated by crowding at the central plant and a resulting search for new food resources, because most of the central plants were severely wilted or dead by the end of the experiment. Vertical movement of *D. abbreviatus* neonates has been studied (Quintela & McCoy 1998), and larvae typically remain in about 1 m radius of citrus tree trunks (Bates et al. 2015), but little is known about horizontal movement of late-instar *D. abbreviatus* in soil. A couple *D. abbreviatus* larvae were recovered from distal plants, suggesting movement of almost 1 m in 7 wk is possible but not common.

Overall, larval *D. abbreviatus* recovery rates were low at an average of 0.7 out of 4 larvae from central plants in control plots, and no larvae were found in peripheral plants from control plots by 3 wk after treatment applications. We searched only for larvae in plant root zones and missed any larvae moving between plants. In addition, some larvae likely died, because uninfected dead larvae were found in the peripheral untreated control plants. *Diaprepes abbreviatus* late-instars are capable of moving from 1 strawberry plant to another but caused a relatively low level of injury to plants to which they moved.

In the research plot experiment, *S. riobrave* were applied to all plants in a plot 2 wk after placing *D. abbreviatus* on 1 central plant. No live *D. abbreviatus* were recovered from proximal plants treated with *S. riobrave* even though minimal plant wilting occurred. It is likely that uninfected larvae moved from the central to proximal plants, began feeding on plant roots, were infected by *S. riobrave*, but ceased feeding and died before significant plant damage was caused. Since we allowed *D. abbreviatus* 2 wk to begin feeding and moving before applying *S. riobrave*, an earlier application to central plants only as soon as wilting was noticed, may have produced similar or even better results. This possibility needs to be tested in a future experiment. From a practical perspective, it will be more economical and thus feasible to selectively apply *S. riobrave* in large strawberry fields to wilting plants only and not to large numbers of plants adjacent to them.

There is increasing interest in managing *D. abbreviatus* through entomopathogenic nematode conservation, as abundant and suppressive entomopathogenic nematode communities were found in some Florida citrus orchards (Duncan et al. 2013). Conventional strawberry fields are fumigated annually and will require annual *S. riobrave* augmentation, but entomopathogenic nematode communities should be evaluated in organic strawberry production where cover crops and physical methods are used in place of chemical fumigation, and may result in conserved and suppressive entomopathogenic nematode communities. Because it appears that *D. abbreviatus*-damaged plants are localized to certain fields or in field areas, yearly monitoring of larval patches and plant wilting will determine whether or not they are seasonally stable and, if so, then *S. riobrave* applications can be consistently applied to these fields or field areas. More knowledge about *D. abbreviatus* flight and oviposition periods in strawberry is needed to guide *S. riobrave* application timing decisions, and persistence of *S. riobrave* in plasticulture earthen beds should be determined to guide timing of repeat applications. Currently, *S. riobrave* applications may not be cost-effective as they are only sold in large quantities for use in citrus; localized applications would require a change in marketing and production of *S. riobrave*. In conclusion, *S. riobrave* successfully infected and killed late-instar *D. abbreviatus* in strawberry and was as effective as imidacloprid in reducing plant wilting. With further research to determine optimal application strategies, *S. riobrave* can be successfully developed into safe and relatively easy-to-use biological control method to reduce impacts of *D. abbreviatus* in strawberry.

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References Cited

Bates LM, Bethke JA, Bender GS, Morse JG, Godfrey KE. 2015. Seasonal adult emergence patterns and soil larval distribution of *Diaprepes abbreviatus* (Coleoptera: Curculionidae) in southern California. Journal of Entomological Science 50: 326–334.

Bender GS, Bates LM, Bethke JA, Lewis E, Tanizaki G, Morse JG, Godfrey KE. 2014. Evaluation of insecticides, entomopathogenic nematodes, and physical soil berries of control of *Diaprepes abbreviatus* (Coleoptera: Curculionidae) in citrus. Journal of Economic Entomology 107: 2137–2146.

Bullock RC, Pelosi RR, Killer EE. 1999. Management of citrus root weevils (Coleoptera: Curculionidae) on Florida citrus with soil-applied entomopathogenic nematodes (Nematoda: Rhabditida). Florida Entomologist 82: 1–7.

Caruso NA, Smith HA, Leppla NC, Liburd OE, Funderburk JE. 2019. Common pests of Florida strawberry. Integrated Pest Management Florida. University of Florida, Institute of Food and Agricultural Science, Gainesville, Florida, USA. https://ipm.ifas.ufl.edu/pdfs/Strawberry_Pests_Poster.pdf (last accessed 16 Mar 2021).

Deschamps SS, Whitaker VM, Agehara S. 2019. White-striped plastic mulch reduce root-zone temperatures during establishment and increases early season yields of annual winter strawberry. Scientia Horticulturae 243: 602–608.

Dolinski C, Choo HY, Duncan LW. 2012. Grower acceptance of entomopathogenic nematodes: case studies on three continents. Journal of Nematology 44: 226–235.

Duncan LW, Mannion CW. 2021. 2020–2021 Florida citrus production guide: citrus root weevils. Publication #ENY-611. University of Florida, Institute of Food and Agricultural Science, Gainesville, Florida, USA. http://edis.ifas.ufl.edu/cg006 (last accessed 16 Mar 2021).

Duncan LW, McCoy CW, Terranova AC. 1996. Estimating sampling size and persistence of entomogenous nematodes in sand soils and their efficacy against the larvae of *Diaprepes abbreviatus* in Florida. Journal of Nematology 28: 56–67.

Duncan LW, Shapiro DI, McCoy CW, Graham JH. 1999. Entomopathogenic nematodes as a component of citrus root weevil IPM, pp. 69–78 *In* Polavarapu S [ed.], Optimal Use of Insecticidal Nematodes in Pest Management. Rutgers University, New Brunswick, New Jersey, USA.

Duncan LW, Graham JH, Dunn CD, Zellers J, McCoy CW, Nguyen K. 2003. Incidence of endemic entomopathogenic nematodes following application of *Steinernema riobrave* for control of *Diaprepes abbreviatus*. Journal of Nematology 35: 178–186.

Duncan LW, Graham JH, Zellers J, Bright D, Dunn DC, El-Borai FE. 2007. Food web responses to augmentation biological control using entomopathogenic nematodes in bare and composted-manure amended soil. Journal of Nematology 39: 176–189.

Duncan LW, Stuart RJ, El-Borai FE, Campos-Herrera R, Pathak E, Giurcanu M, Graham JH. 2013. Modifying orchard planting sites conserves entomopathogenic nematodes, reduces weevil herbivory and increases citrus tree growth, survival and fruit yield. Biological Control 64: 26–36.

Hiltpold I, Hibbard BE, French BW, Turlings TCJ. 2012. Capsules containing entomopathogenic nematodes as a Trojan horse approach to control the western corn rootworm. Plant and Soil 358: 11–25.

Kaspi R, Ross A, Hodson AK, Stevens GN, Kaya HK, Lewis EE. 2010. Foraging efficacy of the entomopathogenic nematode *Steinernema riobrave* in different soil types from California citrus groves. Applied Soil Ecology 45: 243–253.

Knapp JL, Simpson SE, Peña JE, Nigg HN. 2000. Diaprepes root weevil: what Floridians need to know. Publication #ENY-640. University of Florida, Institute of Food and Agricultural Science, Lake Alfred, Florida, USA. https://ufdcimages.uflib.ufl.edu/IR/00/00/28/01/00001/IN11800.pdf (last accessed 21 Mar 2021).

McCoy CW. 1999. Arthropod pests of citrus roots, pp. 149–156 *In* Timmer LW, Duncan LW [eds.], Citrus Health Management. APS Press, St. Paul, Minnesota, USA.

Mannion CM, Hunsberger A, Peña JE, Osborne LS. 2003. Oviposition and larval survival of *Diaprepes abbreviatus* (Coleoptera: Curculionidae) on select host plants. Florida Entomologist 86: 165–173.

Nigg HN, Simpson SE, Stuart RJ, Yang LK, Adair RC, Bas B, UR-Rehman S, Cuyler NW, Barnes JI. 2004. Reproductive potential of Florida populations of *Diaprepes abbreviatus* (Coleoptera: Curculionidae). Journal of Entomological Science 39: 251–266.

Patel MN, Perry RN, Wright DJ. 1997. Desiccation survival and water contents of entomopathogenic nematodes, *Steinernema* spp. (Rhabditida: Steinernematidae). International Journal for Parasitology 27: 61–70.

Quintela ED, McCoy CW. 1998. Synergistic effect of imidacloprid and two entomopathogenic fungi on the behavior and survival of *Diaprepes abbreviatus* (Coleoptera: Curculionidae) in soil. Journal of Economic Entomology 91: 110–122.

Renkema J, Devkota S, Nagle C. 2019. Pesticide options for insect, mite, and mollusk management in commercial strawberry production in Florida. Publication #ENY-689. University of Florida, Institute of Food and Agricultural Sciences, Gainesville, Florida, USA. https://edis.ifas.ufl.edu/pdffiles/IN/IN48600.pdf (last accessed 16 Mar 2021).

SAS Institute. 2019. JMP® software version 15.0.0. SAS Institute, Cary, North Carolina, USA.

Shapiro-Ilan D, Dolinski C. 2015. Entomopathogenic nematode application technology, pp. 231–254 *In* Campos-Herrera R [ed.], Nematode Pathogenesis of Insects and Other Pests. Springer, Basel, Switzerland.

Simpson SE, Nigg HN, Coile NC, Adair RA. 1996. *Diaprepes abbreviatus* (Coleoptera: Curculionidae): host plant associations. Environmental Entomology 25: 333–349.

Stuart RJ, McCoy CW, Castle WS, Graham JH, Rogers ME. 2006. *Diaprepes*, *Phytophthora*, and hurricanes: rootstock selection and pesticide use affect growth and survival of 'Hamlin' orange trees in a Central Florida citrus grove. Proceedings of the Florida State Horticultural Society 119: 128–135.

Suh DH, Guan Z, Khachatryan H. 2017. The impact of Mexican competition on the US strawberry industry. International Food and Agribusiness Management Review 20: 591–604.

USDA/NASS. 2018. State Agricultural Overview – Florida. https://www.nass.usda.gov/Quick_Stats/Ag_Overview/stateOverview.php?state=FLORIDA (last accessed 16 Mar 2021).

Woodruff RE. 1985. Citrus weevils in Florida and the West Indies: preliminary report on systematics, biology, and distribution (Coleoptera: Curculionidae). Florida Entomologist 68: 370–379.