Insect-Parasitic Nematodes for Management of Black Vine Weevil in Commercial Centers

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The black vine weevil (BVW), *Otiorhynchus sulcatus*, long recognized as a serious pest of container-grown nursery plants, ornamentals in landscape, and field vine crops, causes damage through larval feeding on roots and girdling of stems at the soil surface. Among the weevil's favored host plants are azaleas, cyclamens, escallonias, euonomyus, grapes, ivy, sweet gum, junipers, and rhododendrons.

Recent studies by other researchers (Parella & Keil, Stimmann et al. and Phillips) have documented the life history of BVW and its associated feeding damage. Briefly, the flightless parthenogenetic females (males are unknown in this species) emerge in early spring and feed at night on foliage leaving small characteristic notches along the leaf margins. Within 20 to 30 days after emergence, the weevils begin to lay eggs with each capable of producing up to 500 eggs. The eggs, deposited at the soil surface near the roots, hatch within 10 days at soil temperatures of 75°F. The white, legless larvae feed on the roots and under high population densities will girdle the plant at the soil surface causing its death. BVW larvae overwinter in the soil with pupation occurring in the spring.

Managers of shopping and commercial centers and buildings beautify their surroundings with plants, many of which are favored hosts for BVW. Infested plants from nurseries are often the source of BVW for those centers. Because all BVWs are females, an infestation can start with one individual and spread throughout the plantings by adult BVW movement or by human manipulation of infested plants or soil containing eggs, larvae or adults. The nocturnal habit of BVW adults, which hide in the leaf litter during the day, and the nonvisibility of larvae and pupae in the soil make detection difficult. Once a foothold is established, significant BVW damage may not occur for several years, and an infestation often is recognized only when plants start to show dieback.

BVW infestations in nurseries, outdoor landscaping, and field vine crops can be controlled using several options including chemical insecticides. However, in commercial buildings that cater to shoppers and outdoor diners, applying chemical control to containerized landscape plants is difficult if not impossible. Therefore, biological control of this insect is an attractive alternative.

BVW larvae and pupae have been shown to be highly susceptible to insect-parasitic nematodes in the families Steinernematidae and Heterorhabditidae. These nematodes possess many attributes of successful biological control agents including their active host-seeking ability in soil, availability from commercial sources, safety to nontarget organisms including humans, and ease of application.

Our objective was to establish these insect-parasitic nematodes as long-term control agents against BVW. We were alerted to an infestation in containerized plants in a commercial financial building in San Francisco and initiated a year-long biological control program against BVW larvae and pupae using insect-parasitic nematodes.

Black vine weevil adult and feeding damage to leaf.

Two black vine weevil pupae (right) infected with *Heterorhabditis bacteriophora*. Pupa (left) is healthy.

Experimental Design

Our approach was to determine the persistence and recycling ability of two insect-parasitic nematodes and hence the long-term control of BVW in soil planted with kangaroo ivy (*Cissus antarctica*) vines. The building's 7-by-24-by-7-inch fiberglass planters (168-square-inch surface area) with 3 drainage holes in the bottoms contained a commercial potting soil and three ivy vines. The soil and plants had been in those containers for less than one year at the start of the experiment, and black vine weevil was known to be present in the adjacent plantings.

The nematodes we used in the experiments were *Steinernema feltiae* (also known as *bibionis*) Finland strain and *Heterorhabditis bacteriophora* NC1 strain and were obtained commercially from Biosys (Palo Alto, CA) and Bioenterprises (Roseville, Australia), respectively. These and related nematode species carry a mutualistic bacterium, *Xenorhabdus* spp., in their intestines and together they attack and kill their insect hosts. The small infective nematodes, measuring about .02 inches in length, enter their insect host through the host's mouth, anus, or breathing pore. Once inside the host's intestine or breathing pore, the nematode penetrates the body cavity and releases the bacterium. The bacterium multiplies rapidly and kills the insect within 48 hours. The nematodes feed on the bacterial cells and insect tissues and become adults in a few days. The adult females produce progeny that develop into a second or third generation of adults which produce the infective nematodes. The entire life cycle from infection to production of a new infective generation takes 10 to 14 days.

To determine if the presence of an alternate non-feeding insect host for the nematode would bolster BVW control, half of the treatments received 10 *Galleria mellonella* (greater wax moth) larvae which were buried one inch deep and equally spaced around the perimeter of each planter. The following six treatments were made: (1) control with *Galleria*, (2) control without *Galleria*, (3) *S. feltiae* with *Galleria*, (4) *S. feltiae* without *Galleria*, (5) *H. bacteriophora* with *Galleria*, and (6) *H. bacteriophora* without *Galleria*. Each treatment was replicated three times with four planters per replicate. The plants were watered, fertilized, and maintained by the grounds personnel at the center.

Before treatment, the soil from each planter was bioassayed for presence of naturally occurring nematodes. Three soil subsamples from around the base of each vine within a planter were taken, placed into one plastic cup, returned to the laboratory, and baited with six *Galleria* larvae. The cups were capped and stored at room temperature. One week later, the *Galleria* larvae were removed from the cups and examined for the presence of nematodes. No insect-parasitic nematodes were found in any of the planters before nematode application. Throughout the experiment nematodes were never recovered from control planters with or without the addition of *Galleria* larvae.

Using a hand-held two-gallon Hudson sprayer, we applied *Steinernema feltiae* at a rate of 320 per square inch, and *H. bacteriophora* at 480 per square inch on the morning of February 3, 1989, between 6:00 and 8:00. *Galleria* larvae were placed into the soil prior to spray application and again on May 26, 1989.

Using the same bioassay technique described above, the soil from each of the treatment containers was sampled separately for nematode persistence at prescribed intervals (2 to 3 weeks) after treatment. Nematode presence and persistence in the soil was measured by the percentage of *Galleria* larvae infected with nematodes. Dead *Galleria* larvae were dissected for presence or absence of nematodes. All soils were sterilized and returned to the planters after each sample period. At the end of the experiment on February 15, 1990, containers were meticulously examined as we removed all soil for final sampling and searched for BVW larvae and pupae.

Persistence of Nematodes

In the *S. feltiae* plots, there was no difference in nematode persistence between planters receiving *Galleria* or not for the first 100 days after application. However, after the second addition of *Galleria* larvae (112 days after nematode application) in the planters with alternate hosts, the percentage of bioassayed hosts infected with nematodes was always higher than in the planters not receiving the alternate host.

Similar trends were observed with *H. bacteriophora*, albeit at a lower level of nematode infection compared with *S. feltiae*. There was no difference in nematode persistence between planters receiving *Galleria* or not for the first 70 days. After this period, the percentage of *H. bacteriophora*-infected bioassay insects dropped drastically, rebounded slightly between 80 and 100 days, and then dropped to less than 10 percent between 100 and 140 days. In the treatment with the second addition of *Galleria* larvae, the soil showed a marked increase in bioassayed insects infected with the nematode from 160 days onward. The planters not receiving *Galleria* remained at a low level (less than 15 percent bioassay insects infected) until 240 days after nematode application when the percentage of bioassayed hosts infected with the nematode increased to 50 percent.

Both *S. feltiae* and *H. bacteriophora* treatments with alternate hosts had higher nematode populations than treatments without alternate hosts, suggesting that the nematodes used them to recycle. Yet, the long persistence of both nematodes in treatments without alternate hosts is uncharacteristic, suggesting that they were recycling also in weevil larvae and pupae.

Upon examination of all planters at the end of the experiment, BVW larvae were recovered from all treatments. The control planters had a significantly higher population of larvae than the nematode treatments. There was no significant difference between nematode treatments receiving alternate hosts or not, although the nematode treatments with *Galleria* had less BVW larvae than the nematode treatments without *Galleria*.

Practical Applications

Our experiment demonstrated that *S. feltiae* and *H. bacteriophora* can persist and recycle in potted soil when hosts are present. More significantly, those nematodes reduced black vine weevil infestations to a very low level. The periodic addition of alternate hosts to boost nematode populations is not practical for those commercial buildings where large number of planters are maintained for several years. Rather, it seems that a periodic check for BVW as suggested by Stimmann et al. (*California Agriculture*, January-February, 1985) may be a more prudent method of managing this insect. If BVW are found, nematodes can be augmented as needed to bolster the existing nematode population to suppress this pest. We conclude that the safety feature and the degree of long-term control by insect-parasitic nematodes of BVW provide significant advantages compared with chemical control in buildings with containerized landscape.

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