Using Beneficial Nematodes to Control Black Vine Weevil and Rhododendron Borer

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Rhododenron borer (*Synanthedon rhododendri)* and the black vine weevil (*Othioranchus sulcatus)* are two major pests of rhododendron. Research conducted by the University of Maryland Cooperative Extension has shown that these pests can be greatly reduced in number by using entomopathogenic nematodes. Our work was conducted over several years at the Central Maryland Research and Education Center and at commercial nurseries.

Rhododendron Borer

Early symptoms of damage from rhododendron borer appear like drought stress with foliage los-ing color and dying back. This clearwing moth borer, a member of the family Sesiidae, damages rhododendrons, azaleas, and mountain laurel. The female moth lays eggs at the base of branches or near the crown of the plant. On close examination frass may be found being expelled from the holes as the larvae excavate a gallery. The borer has a one year life cycle with the larvae being present in the plant for about 11 months of the year. Adults emerge in mid-summer, mate, and then females lay eggs on susceptible plants. The larval stage is susceptible to entomopathogenic nematodes.

Photo Stanton Gill

▲ Adult Rhododendron Borer

A traditional control for insects boring in plants such as rhododendrons is the application of broad spectrum chemical insecticides applied to the trunk and branches just before the borer eggs hatch. Insecticidal bark sprays are effective only if a lethal residue is present during the brief interval between the time when larvae hatch and before the larvae enter the tree.

The use of pheromone traps has made timing applications of synthetic insecticides more efficient (1, 8, 9). Unfortunately, pheromone traps are under utilized by many people, and pesticide applications are often made on predetermined schedules. This approach can result in borer larvae successfully penetrating the bark before a pesticide application is made or after the chemical has broken down to a sub-lethal level. Once the larvae are under the bark, pest control options are severely limited.

Black Vine Weevil

The flightless black vine weevil damages rhododendrons, yews, and many herbaceous perennials. The adult weevils, which all are females, are active in mid-summer and feed on the margins of leaves, leaving a characteristic notching of foliage. The females reproduce parthenogenetically and lay eggs into the soil. The larva is a legless grub with a white body and red head capsule. The larvae, present in the soil 10 -11 months of the year, feed on roots and at the base of plants, causing girdling of rhododendron plants with resulting death. Soil drench insecticides such as imidacloprid have provided good levels of control but some rhododendron enthusiasts wish to use a biological control, over an insecticide. Fortunately, the larvae of black vine weevil are susceptible to entomopathogenic nematodes.

Photo John Davidson

▲ Black Vine Weevil Adult and Larva

Beneficial Nematodes

An alternative control method for dealing with borers and ground infesting weevil larvae is through the use of entomopathogenic nematodes (5). Entomopathogenic nematodes infect only insects or related arthropods. As a biological control agent, entomopathogenic nematodes offer two major advantages. In the case of rhododendron borer, the first is their ability to attack borer larvae after they have entered the plant. The second advantage is that nematodes are safe for the pesticide applicator and have no adverse impact on non-target sites (10). In the case of black vine weevil the larvae provide a safe alternative for people wishing to avoid use of synthetic chemical applications.

Several commercial companies are marketing nematodes for use in controlling insects in the landscape and nursery. These products contain infective juvenile nematodes in the "dauer" or J-3 stage and are formulated for applications as sprays or drenches. The J-3 stage nematodes enter through the mouth, anus, or breathing tubes (spiracles) of the insect (2). After penetrating the insect, the nematodes release bacteria that enter the insect's blood stream. The nematodes then feed on these bacteria as they multiply and the insect dies of bacterial septicemia. These entomopathogenic nematodes and their associated bacteria, *Xenorhabdus nematophilus* and *X. luminescens*, have been extensively

Photo Home and Garden Information Center/University of Maryland

▲ Rhododendron Borer damage to a rhododendron.
▼ Close up of Rhododendron Borer damage.

Photo Home and Garden Information Center/University of Maryland

tested for toxicity to non-target organisms, and they are considered to be nontoxic and nonpathogenic to plants and mammals (10).

Beneficial nematodes are microscopic, but their benefit to growers and plant enthusiasts is sizable. These nematodes feed within insects, releasing bacteria in their fecal waste that causes the insect to become sickened and die. The entomological term for this is "entomopathogenic," and these beneficial nematodes are referred to as entomopathogenic nematodes. Entomopathogenic nematodes infect only insects or related arthropods. Entomopathogenic nematodes are exceptionally lethal to many soil and cryptic insect pests, yet pose no danger to plants and animals. Entomopathogenic nematodes kill insects by entering through a body opening such as the insects' mouth, spiracles, or anus and releasing a bacterium. In the case of *Heterorhabditis* species, the structure of the strong head allows it to enter through the host's soft, sidewall tissue.

Only the third instar juvenile of this nematode is capable of entering insects. If the environment is warm (50–85° F [10–30° C]) and moist, these nematodes will complete their life cycle within the infected insect. Using entomopathogenic nematodes is ideal for control of insects that have life stages in soil.

Once the nematode is inside the insect, the nematode sheds a protective cuticle and begins feeding and defecating. A symbiotic bacterium lives inside the nematode and is excreted in its waste material. The nematode's feeding does not kill the insect, but the bacterium it excretes does. The bacterium also contributes anti-immune proteins to assist the nematode in overcoming host defenses. The bacterium in the fecal waste of *Heterorhabditis bacteriophora* nematodes is *Photorhabdus luminescens*; the steinernematid nematodes release *Xenorhabdis nematophilus* bacterium. The insect dies of bacterial septicemia in 24 to 48 hours.

The nematode's life span is six to eight weeks for *H. bacteriophora* and 12-15 weeks for *Steinernema carpocapsae.* The nematodes progress through four immature stages called J-1, J-2, J-3, and J-4, before reaching adulthood. The first two J-stages are spent usually inside the host (in vivo), but when they hit the J-3 stage they search for prey. The J-4 stage and adulthood is spent inside the host. Their life cycle cannot progress beyond J-3 until they find a new host. The conditions for optimum success with most species of steinernematid and heterorhabditid entomopathogenic nematodes are when soil temperatures are 60 to 70° F (16 to 21° C) and the soil is slightly moist. *H. bacteriophora* are active at low soil temperatures of 45° F (7° C), but the *Photorhabdus* bacterium inside the nematode requires higher temperatures, usually of 60° F or greater to break dormancy. *Steinernema* spp. show some activity at low temperature of 40° F (4° C), but the *Xenorhabdus* bacterium generally needs temperatures above 55° F (13° C) to break dormancy. If the nematodes are placed into the soil when the pest is active, the bacterium will become active when

the minimum temperature is reached. Nematodes such as steinernematids can generally be maintained for six to fifteen weeks. Shorter performance is influenced by the soil media drying out or the nematodes' being exposed to ultraviolet light, which is not likely in most container production. Cooler and warmer temperatures and insufficient moisture levels in the soil will hamper development and reproduction to a certain degree.

Nematodes such as *S. carpocapsae* sit and wait for prey, standing on their tail and nictating (wiggling) until prey moves close, when they respond quickly to carbon dioxide given off by the host insect. Other nematodes such as *S. glaseri*, and *Heterorhabditis bacteriophora* are highly mobile and actively searching for host insects.

Small bio-technical companies are producing and marketing a variety of nematodes, and many "cottage" industries have grown up to supply growers' needs. A list of suppliers of beneficial nematodes is provided at the end of this article. Marketing of entomopathogenic nematodes has focused on *Steinernema* and *Heterorabditis spp. Steinernema spp.* occur most commonly in the soil, are generally easier to rear on artificial media, and tend to be more persistent after application. The Steinernema nematodes are best suited to control soil pests commonly found in the top 1-2 inches of the media or for control of clearwing moth borers. *Heterorhabditis spp.* are usually more mobile and have superior host-searching activity; they are the nematode of choice for insects that inhabit deeper regions of the soil profile, such as black vine weevil and scarab beetles. Several of the *Heterorhabditis spp.* nematodes have been labeled "Cruisers" since they cruise about in search for their insect prey.

When a susceptible host is located, the infective nematodes enter the insect's body through natural openings such as the mouth, spiracles, and anus. There, they feed and release bacteria, which live symbiotically in the gut of the nematode. The bacteria rapidly multiply, killing the insect. The nematodes will then reproduce and colonize the cadaver. Once the food supply is exhausted, thousands of infective stages leave the insect's body and return to the soil in search of additional host larvae. Host death occurs within 27-72 hours of host penetration, depending on the nematode and pest species, and ambient conditions.

Nematodes are commonly formulated as slurries, water dispersible granules within an inert clay carrier, gels, or in sponges. All need to be soaked to release nematodes into suspension and to ensure their rehydration and "activation."

Of the nearly 30 steinernematid and heterorhabditid nematodes identified to date, seven species are commercially available. When purchasing nematodes a comparison-shopping approach is recommended as prices vary greatly among suppliers. One billion nematodes per acre is the rule of thumb against most soil insects such as fungus gnat larvae, black vine weevil, and scarab beetle larvae. Rates for containerized plant material tend to treated at slightly higher rates.

Steinernema carpocapsae

S. carpocapsae is the first nematode that we started testing in Maryland back in the late 1980's. It is easy to mass rear and can be formulated in a partially desiccated state that provides several months of room-temperature shelf life. This nematode stands on its tail in an upright position and attaches to passing hosts. They are highly responsive to carbon dioxide once a host has been contacted. The nematodes generally enter through the insect's spiracles. The nematodes are most effective at temperatures ranging from 70 – 85 F. *S. Carpocapsae* has been effectively used for dogwood borers, banded ash clearwing borer, lilac borer, oak borer, rhododendron borer, and peach tree borer larvae. It has also been used for cutworm control and armyworms.

Steinernema kushidai

This is the new kid on the block. This nematode was isolated in Japan and has shown to be a very effective parasite of scarab beetle larvae such as Japanese beetle and oriental beetle larvae. If the companies can resolve how to mass reproduce this nematode, you should see it on the market in a couple of years.

Phasmarbditis hermaphrodita

This is a nematode that causes mortality on a wide range of slugs and snails, but it harmless to other invertebrates. It can be readily reared in culture, formulated and stored for use as a biocontrol agent. The nematode enters the dorsal pore, just behind the mantle on slugs, and once inside caused the slug to swell up and die rapidly. This nematode is label in England and distributed by MicroBio. We would love to test this nematode in Maryland, but this species has to be found existing naturally in our state before it can be imported from the United Kingdom.

Heterorhabditis bacteriophora

This nematode is fairly versatile, attacking lepidopterous (moths and butterflies) and Coleopterous (beetle) larvae, among other insects. This nematode species is labeled a "cruiser" that actively searches for its prey. The best control we have obtained with this species as been against black vine weevil larvae in container grown plants. The only drawback is this nematode needs warm soil temperatures of at least 50 F and has shown reduced efficacy at temperatures below 70 F. Shelf life of this nematode species is a problem and nematodes that are shipped to a nursery should be used shortly after arriving. If they must be stored, refrigerating will keep them for two to three months. Most infective juveniles persist only a few days following a release in field conditions. This nematode species is the most sensitive among entomopathogenic nematodes to physical stress.

Nematodes are usually applied to the soil by drenching, using coarse sprays and even overhead irrigation systems. The goal is to distribute them evenly over and through the media. Once in the soil, nematodes begin to search for a suitable host. While the nematodes can move to reach a host,

Suppliers of Entomopathogenic Nematodes

Beneficial Insect Company
244 Forrest Street, Fort Mill, SC 29715.
BioLogic Company
P.O. Box 177, Willow Hill, PA 17271
Bioshield
P.O. Box 9068, Fresno, CA 93790
Bountiful Gardens
18001 Shafer Ranch Rd, Willits, CA 95490
Bozeman Bio-Tech
P.O. Box 3146, Bozeman, MT 59772
Crop King, Inc.
P.O. Box, Medina, OH 44258
Ecogen Inc
2005 Cabot Boulevard West,
Langhorne, PA 19047-1810
Gardener's Supply Company
128 Intervale Road, Burlington, VT 05401
Green Spot, Department of Bio-Ingenuity
93 Priest Road, Nottingham, NH 03825
Harmony Farm Supply
P.O. Box 460, Graton, CA 95444
Hydro-Gardens, Inc.
P.O. Box 25845, Colorado Springs, CO 80936
MicroBio
17 Street, Whittlesford, Cambridge CB2 4LT,
England. Sold through E.C. Geiger, Box 285,
Route 63, Harleysville, PA 19438-0332

the distances they can move are limited, so placement in the vicinity of the target pest(s) will promote parasitization and control. The soil must be kept moist to allow the nematodes to move, but should not be over watered as this will wash the nematodes from the "target zone." If the soil dries, nematodes can die through desiccation, or they will not be able to move in search of a suitable host.

Most nematodes can be satisfactorily stored under refrigeration for some time prior to use. The length of time varies according to their method of production and formulation. Typically, nematodes are either produced on an artificial diet (*in vitro*) or in an insect host (*in vivo*). Those produced *in vitro* are often formulated in a dormant state that can be stored for up to six months without any loss of viability. Such products need to be "re-activated" prior to application (see list). Nematodes produced *in vivo* may be stored for two to three months without any problems, living off fat reserves; healthy nematodes will not be unaffected but the initial pre-storage health of the shipment should be confirmed.

Used correctly, these biocontrol agents are highly effective management tools, providing control as effective as, as cost-comparable to, conventional chemical treatments. Nematodes are available for greenhouses and nurseries, and have been successfully used in a wide variety of media, including soil, peat-lite mixes, bark products, and rockwool. For example, in trials run in Maryland, fungus gnat control in a poinsettia crop with *S. feltiae* was as effective as that obtained using Knox-Out®.

Detailed recommendations on dose and use are generally provided with each shipment of nematodes or can be readily obtained by contacting the producer or supplier. Applications must be made when susceptible stages of the target pest are present; if multiple and overlapping generations occur, susceptible stages will always be present and several applications will be required to bring the pest population under control. Otherwise, applications must be made at specific times to target susceptible developmental stages during the pest's life cycle.

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