Entomopathogenic Nematode use in Nurseries and Greenhouses

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Entomopathogenic nematodes in the genus *Steinernema* and *Heterorhabditis* and their associated bacteria (*Xenorhabdus* spp.) have shown great potential as biological control agents for a variety of insect pests including several curculionid species, or so-called "root weevils" and sciarid flies, such as fungus gnats. The nematodes are able to kill hosts rapidly, are easy to apply, and are exempted from federal and state registration requirements in most countries because of their safety to mammals and plants. Difficulties in production, storage, formulation, quality control, and application technology had limited their success for market introductions in the past. Recent public pressure to limit environmental contamination associated with chemical insecticide use has resulted in a dramatic increase in research conducted by scientists in government, universities, and industry to overcome some of these technological difficulties. Industry has now seen the development of three major biotechnology companies which have been successful in introducing nematode-based products into some commercial markets.

Production. Since their discovery as biological control agents, nematodes have been produced *in vivo,* in which an insect host serves as the medium for nematode-bacterial growth and production. This method has limitations because it requires a constant source of healthy insects, is sensitive to biological variation, and costs of production are high in terms of equipment and man-hours. More efficient methods of production using *in vitro* methods have been and are being developed.

Currently, both heterorhabditid and steinernematid nematodes are produced in monoxenic solid phase systems. However, there are economic limitations to this approach such as labor costs, consistency of production, and sensitivity to contamination. *Steinernema* spp. are now commercially produced in monoxenic liquid culture systems which utilize fermentation tank technology. This approach is the most economical of all known methods. Nematode production is taking place in tanks of up to 80,000 liters in volume, which has lowered costs considerably, allowing successful introductions into markets requiring large numbers of nematodes, or markets of low cash crop value.

Formulation. The successful market introduction of an entomopathogenic nematode product requires a reliable and stable formulation. This has been a difficult task because most larger markets are demanding a product with a minimum shelf-life of 6 months when stored at room temperatures (20-25° C). Nematode products contain living animals which have certain temperature, oxygen, and moisture requirements necessary for their survival and effectiveness as control agents. While no nematode formulation has been completely successful in reaching these goals, some have been developed in certain market segments. A few commercial products use moist substrates such as sponge, vermiculite and peat. These materials require refrigeration because warm temperatures increase nematode metabolic activity thus reducing pathogenicity and virulence. Other

materials such as alginate gels, clays, activated charcoal, and polyacrylamide gels immobilize or partially desiccate the nematodes. This reduces their metabolism and improves their tolerance to temperature extremes. Commercial products using alginate materials can now provide a viable product for 5 months at temperatures as high as 25°C

Application Technology. Strategies must also be developed which insure the successful delivery of the nematode to the target site and target insect, thereby increasing the probability of nematode-insect interaction. One strategy is the selection of proper equipment for application. Entomopathogenic nematodes can be applied through most common agricultural liquid application equipment including irrigation systems (via chemigation which is preferred for certain crops) They can withstand pressures of up to 300 lb/in^2 and can be applied through most common nozzles with openings as small as 50 microns. Another consideration is proper spray volume and sufficient irrigation. This can vary tremendously depending on soil type, crop, relative humidity and rainfall. Most nematode applications are targeted against life stages found in the soil environment and sufficient spray volume must be used to insure adequate coverage and movement of the nematode into the target area. Adequate irrigation is also crucial to insure nematode movement, survival, and persistence in the soil. Target pest populations and behavior play a role in timing and location of application. Different insect species may require different field dosage rates and optimum entomopathogenic nematode species need to be assessed.

Compatibility with a wide range of agrochemicals has been demonstrated which has allowed nematodes to be introduced successfully into many existing Integrated Pest Management programs With certain root weevil species, high population pressures have been effectively controlled with a combined approach of using nematodes against the immature stages and insecticides against the adults stages

In North America two entomopathogenic nematode species have been introduced commercially for control of certain insect pests associated with nursery and/or greenhouse production These are *Steinernema* (=*Neoaplectana*) *carpocapsae* (Weiser, 1955) and *Heterorhabditis bacteriophora* (=*heliothidis*) (Poinar, 1976). By far the most successful large scale introductions have been with *S. carpocapsae* because of improved production and formulation technologies Insect pests listed on current labels are. 1) black vine weevil (*Otiorhynchus sulcatus)*, 2) strawberry root weevil (*O. ovatus*) and 3) fungus gnats (*Bradysia* spp.). Future market possibilities are 1) sugarcane rootstalk borer (*Diaprepes abbreviatus*), 2) blue green weevil (*Pachneus litus*), 3) shore fly (*Scatella stagnalis*), 4) leafminers (*Liriomyza* spp.) and 4) western flower thrips (*Frankliniella occidentalis*).