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PROGRESS IN THE USE OF NATIVE STRAINS OF ENTOMOPATHOGENIC
NEMATODES FOR PLUM CURCULIO MANAGEMENT

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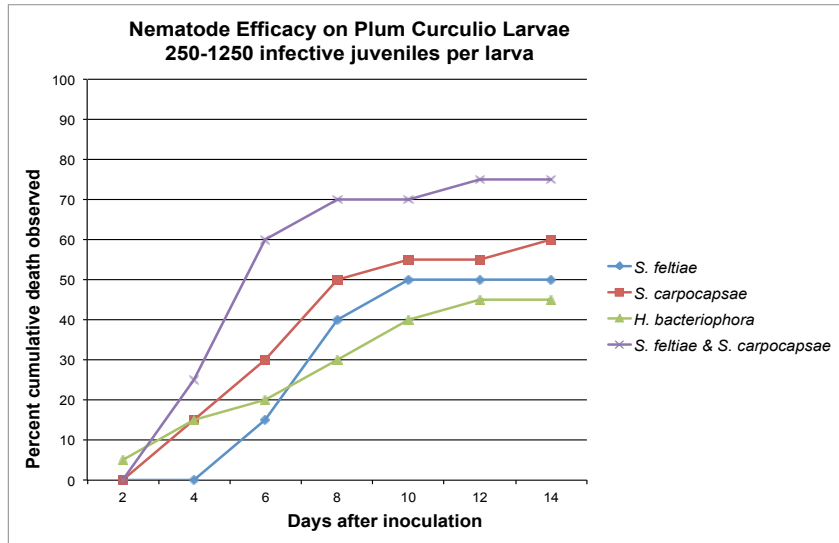
Plum curculio is a key pest in eastern US apple orchards, and is considered to be one of the primary pests limiting organic apple production in this region. Growers typically apply 2–3 insecticide treatments to manage plum curculio, but fruit damage levels and fruit yield losses in conventional commercial apple production can range from 0.5–3%, even with a complete insecticide control program. Current organic production often relies on multiple applications of kaolin clay, which acts as a physical barrier to the insect's attack. However, plum curculio treatment costs in an organic program can easily range between \$150–\$450/acre/year, with fruit damage often remaining at 5–20%. A biological control program for the plum curculio currently does not exist. The purpose of this project is to evaluate the potential of NY cold-adapted entomopathogenic (insect-attacking) nematodes to reduce the impact of plum curculio on apple production, reduce the cost of organic apple production, and provide a higher degree of marketable fruit and a higher profit for all apple producers.

In laboratory bioassays, entomopathogenic nematodes, particularly strains of *Steinernema riobrave*, *S. feltiae*, *S. carpocapsae*, and *Heterorhabditis bacteriophora* have shown efficacy against last instar plum curculio larvae. In the field, various strains of these species have reduced populations by as much as 70–97%, depending on the nematode strain, insect stage, treatment timing and field conditions. Effective control of larval populations has been achieved if the correct nematode species and strain was selected and effectively introduced into the system.

The majority of current research has been directed toward the inundative release (or "biopesticide approach") using commercially reared nematode strains. However, commercial nematode strains tested under NY field conditions have not persisted at any effective population level after 6 months, and the use of these commercial strains would require an annual application. Over the last two years, we have been testing nematode strains that are native to NY, adapted to persist under NY conditions, and have been explicitly propagated to avoid some of the problems with persistence displayed by commercially available nematodes.

NY native strains of the nematode species *S. carpocapsae* (strain 'NY 001') and *H. bacteriophora* (strain 'Oswego') were isolated during the 1990s and used extensively during the past 20 years in field research in NY alfalfa plantings (Shields et al. 2009). In 2004, *S. feltiae* (strain 'NY 04') was isolated from snout beetle-infested fields in northern NY and used in field trials since that time. These NY native nematode strains have been shown to kill mature larvae of Japanese beetle, *Popillia japonica*, and European chafer, *Rhizotrogus majalis*, at varying levels of effectiveness, with *H. bacteriophora* 'Oswego' killing 50% of late-instar Japanese beetle larvae and 8% of European chafer larvae. The native strains show excellent pathogenicity because late-instar larvae are the most difficult life stage for nematodes to attack and kill. These strains have been documented to persist in field sites for many years after inoculation.

In the laboratory, we conducted trials to determine the potential of *S. carpocapsae* ‘NY 001’, *S. feltiae* ‘NY 04’, and *H. bacteriophora* ‘Oswego’ as plum curculio (PC) biocontrol agents. Petri plates were filled with autoclaved soil and inoculated with water solutions at concentrations of 250–1250 nematode infective juveniles (IJs) per ml, and PC larvae and adults were confined in the plates to determine the ability of different nematode strains to kill these two life stages. We found that exposure of PC larvae to the nematodes was more effective than for PC adults, with the highest mortality obtained using a combination of *S. feltiae* and *S. carpocapsae* (up to 75%); mortality of PC larvae was seen within 14 days (Fig. 1). There was no effect of increasing the concentration of IJs above 250/ml.



For testing in the field, we selected a young Idared apple planting and an established block of Empire apples, both located at a NYSAES research farm, as sites for establishment of long-term entomopathogenic nematode (EPN) populations within designated treatment areas. In the spring of 2012, soil cores from each block were bioassayed for presence of EPNs prior to inoculation of the soil surface with native NY strains. No existing nematode populations were found in any of the soil cores. A population of nematode infective juveniles (IJs) of *S. feltiae* and *S. carpocapsae* was applied to the soil surface on May 30 using a modified herbicide sprayer mounted on an ATV. The nematodes were reared in a lab colony using wax moth larvae as hosts, which were brought to the field, dissolved and mixed in water in the spray tank, and applied shortly before sunset in order to avoid excessive sun or heat exposure.

To assess the level of nematode establishment in these plots, we conducted a larval exposure trial using laboratory-reared PC larvae. In August 2012, we dug micro-plot arenas (4.5" diam x 5" deep) using a soil corer in an inoculated row in the Idared block. In each replicate, 10 mature PC larvae were introduced into the arena, which was covered with an emergence trap top, and checked for emerged PC adults over a 5-week period. The same setup was repeated in an untreated row, to serve as a negative check, as well as in an untreated row in which the arenas were first hand-inoculated with laboratory-mixed nematode populations (containing 7,500-9,000 *S. feltiae* IJs per arena), to serve as a positive check. We repeated this trial in 2013, in both the Idared and Empire research plantings, and also in two commercial organic apple orchards in the Hudson Valley (Milton and Fishkill, NY) that had been inoculated in June of that year.

In these field assays, because of very dry soil conditions beginning in late May 2012, coupled with the lack of natural hosts (other insect larvae) in the orchard soil, the establishment of EPN populations in the 2012-treated rows was significantly hampered. In the 2012 micro-plot arena assays, the average percent adult emergence in field-inoculated plots (70%) was greater than in both the hand-inoculated (31%) and check (41%) plots (Fig. 2), indicating that adequate field establishment of the EPN population had not yet occurred. In the 2013 Geneva micro-plot assays, survival of larvae to the adult stage was much lower in all treatments than the levels seen in 2012 (maximum of 26.3%); however, poor survival was also seen in the checks, so that no significant treatment differences were found (Fig. 3). The same trend was seen in the Hudson Valley 2013-inoculated sites, but at least in one orchard (Milton), survival in the hand-inoculated plots was significantly lower than in the checks (Fig 4). The progression of soil core samples in each of the treated orchards taken at successive times after being inoculated shows that gradual field establishment of EPN populations is taking place at all of the sites, with greater levels being seen for *S. feltiae* (Fig. 5); by the end of the 2013 season, most sites showed this species present in 20–30% of samples taken. It is anticipated that levels will ultimately stabilize at 30–40%, which would correspond to an optimal level for effective biocontrol to begin occurring.

Fig. 2. PC emergence in Geneva, 2012

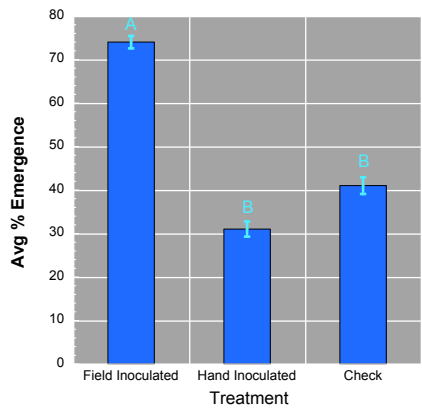


Fig. 3. PC emergence in Geneva, 2013

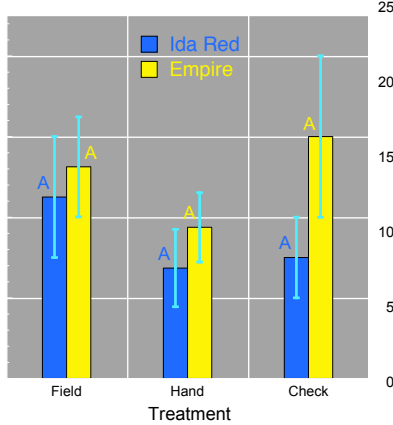


Fig. 4. PC emergence in Hudson Valley, 2013

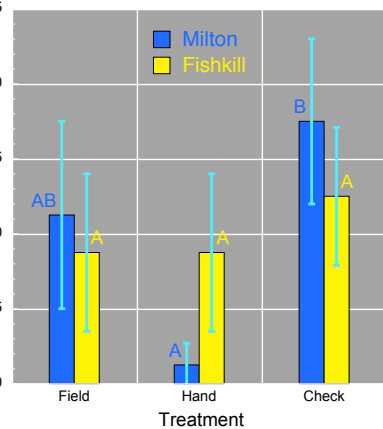
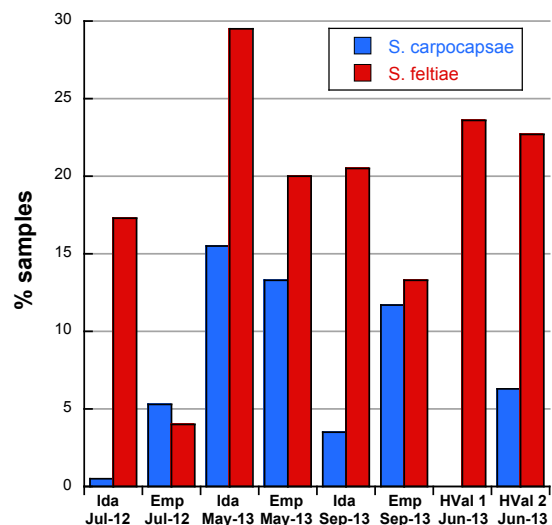


Fig. 5. Progression of nematode establishment in field sites as seen from percent soil samples showing presence of nematodes over time in Geneva (Idared and Empire) and Hudson Valley orchard sites, July 2012 to September 2013.



As another method of assessing overall impact of the nematode treatments on PC populations, fruitlet samples were taken from each of the (2012-inoculated) Geneva research orchards during June of each of the two years. In each row of each plot sampled, 100 fruits from each of 5 trees were assessed for PC oviposition damage (scars). Damage levels in 2013 were reduced at each site from those seen in 2012 (Table 1). At the Empire site, damage in the treated rows decreased 52% from 2012; however, damage in the untreated check rows decreased 76% from 2012, so damage in the check (8.6%) was roughly half of that seen in the treated rows (14.1%). At the Idared site, results were more along the trend anticipated: fruit damage in the untreated rows decreased ~63% from 2012, and in the treated rows, it decreased ~55% from 2012, with damage marginally lower in the nematode treatment (1.7%, vs 2.7% in the check).

Table 1. Plum curculio oviposition damage to fruitlets in nematode-treated orchards

Idared Block	% Damage		Empire Block	% Damage	
	2012	2013		2012	2013
Untreated Rows	7.2	2.7	Untreated Rows	35.9	8.6
Treated Rows	3.8	1.7	Treated Rows	33.7	14.1

We feel that organic apple plantings tend to have greater PC infestations because of fewer effective management options, and that an established, persistent EPN population could help reduce the number of plum curculio larvae residing in the orchard floor that originate from dropped infested fruits.

Acknowledgments

We would like to thank Steve Clark (Prospect Farm, Milton, NY), Josh Morgenthau (Fishkill Farms, Hopewell Junction, NY), and Fabio Chizzola (Westwind Orchard, Accord, NY), for allowing us to work on their farms. Invaluable technical lab and field assistance was provided by Dave Kain, Kristin McGregor, Cortni McGregor, Dylan Tussey, Tessa Lessord, Chrissy Dodge, and Danielle Carolie. We are grateful to Tracy Leskey, Starker Wright, and Torri Hancock (USDA-ARS, Kearneysville, WV) for supplying the plum curculio starter population, and to the NYS Specialty Crops Block Grants Program for funding support.