The Great Lakes Entomologist

Volume 54
Numbers 3 & 4 - Fall/Winter 2021 *Numbers 3 & 4 - Fall/Winter 2021*
Article 4

December 2021

Management of Wireworms in Sweet Potatoes with Persistent NY Entomopathogenic Nematodes

Elson Shields
Cornell University, es28@cornell.edu

Antonio Testa
Cornell University, at28@cornell.edu

Teresa Rusinek
Cornell University

Charles Bornt
Cornell University

Recommended Citation

Shields, Elson; Testa, Antonio; Rusinek, Teresa; and Bornt, Charles 2021. "Management of Wireworms in Sweet Potatoes with Persistent NY Entomopathogenic Nematodes," *The Great Lakes Entomologist*, vol 54 (2)
DOI: https://doi.org/10.22543/0090-0222.2401
Available at: https://scholar.valpo.edu/tgle/vol54/iss2/4

Management of Wireworms in Sweet Potatoes with Persistent NY Entomopathogenic Nematodes

Cover Page Footnote

We thank USDA Speciality Block Grant, NY Farm Viability Institute and Cornell University for providing funds for this study.

Management of Wireworms in Sweet Potatoes with Persistent NY Entomopathogenic Nematodes

Elson Shields*, Antonio Testa, Teresa Rusinek, and Charles Bornt

Department of Entomology, Cornell University, Ithaca, NY 14853
* Corresponding author: (e-mail: es28@cornell.edu)

Abstract

Wireworms are the larval stages of click beetles (Coleoptera: Elateridae) and common polyphagous soil dwelling pests feeding on different plant parts including seeds, roots, stems, and tubers inhibiting plant growth eventually leading to plant death. With the ban of persistent synthetic insecticides such as lindane in 2009 due to negative effects on the environment, no effective control tactics (chemical or biological) are available for wireworms. Some entomopathogenic nematode species/strains have been reported to attack wireworms in the soil, causing death. Non-native EPN species have the advantage of being easily available commercial products for insect pest control. However, these strains do not persist and require annual application as a biopesticide. Native species provide an advantage because application is a single event with multi-year persistence and pest suppression.

In the first harvest, the EPN combination of *Steinernema feltiae* × *Heterorhabditis bacteriophora* had significantly less wireworm feeding damage than the untreated check irrespective of whether the plants were located in the outside rows or the inside rows. The EPN combination of *S. carpocapsae* × *S. feltiae* were numerically different from the untreated checks, but the fewer wireworm feeding wounds were not statistically different from the untreated check. Inoculated EPNs were still present in 30% of the soil samples in all treated plots 1076 days after application/inoculation.

Keywords: Biological Control, EPNs, Wireworms, Persistent EPNs

---

Wireworms are the larval stages of click beetles (Coleoptera: Elateridae) and common polyphagous soil dwelling pests feeding on different plant parts including seeds, roots, stems, and tubers inhibiting plant growth eventually leading to plant death (Parker and Howard 2001, Traugott et al. 2015, Knodel and Shrestha 2018). Crop losses in the United States, Canada, and United Kingdom due to wireworms can reach up to 25% (Parker et al. 1990).

With the ban of persistent synthetic insecticides such as lindane in 2009 due to negative effects on the environment (Vernon et al. 2009), no effective control tactics (chemical or biological) are available for wireworms. Neonicotinoids as seed treatment suppress the wireworm damage to a limited extent, but resurgence can occur, causing more crop damage (Vernon et al. 2009, Barsics et al. 2013). These insecticides might not always be effective because even low wireworm populations can cause plant damage (Parker and Howard 2001, La Forgia and Verheggen 2019). Recently, Labrie et al. (2020) reported the effectiveness of neonicotinoid seed treatments in only 5% of the corn and soybean crop fields treated by neonicotinoids in Quebec and suggested to not to use these insecticides prophylactically due to increasing evidence regarding to negative effects on pollinators (Paquet-Walsh et al. 2019, Labrie et al. 2020).

As a result, there is a need for alternative methods of wireworm control/suppression to reduce plant damage from feeding. Some entomopathogenic nematode (EPN) species/strains have been reported to attack wireworms in the soil, causing death. Non-native EPN species have the advantage of being easily available commercial products for insect pest control (Kaya et al. 2006, Lacey et al. 2015). However, these strains do not persist and require annual application as a biopesticide. Native species provide an advantage because application is a single event with multi-year persistence and pest suppression (Shields et al. 2018). Persistent EPNs strains from a single application are present to attack soil insects for the entire growing season, resulting in greater biological control efficacy (Shields et al. 2018). While concerns of potential non-target effects have been raised by a couple of researchers (Rojht et al. 2009, Abate et al. 2017), local and native EPNs were observed to suppress pests with no additional non-target effects (Shapiro-Ilan et al. 2002, Duncan et al. 2003,

Dillon et al. 2006, Lewis et al. 2006, Campos-Herrera 2015, Sandhi and Reddy 2019).

Recently, Sandhi et al (2020) reported on two Montana native EPN species, *Steinernema feltiae* Filipjev and *Heterorhabditis bacteriophora* Poinar against wireworms in both laboratory and shade house. *S. feltiae* was reported to kill up to 50% of *Limonius californicus* (Mannerheim) larvae, while *H. bacteriophora* did not kill more than 30% of larvae tested. Greater virulence of *S. feltiae* against *L. californicus* in the laboratory is similar to the reports of Toba et al. (1983), who also observed 50% *L. californicus* mortality from *S. feltiae* in laboratory experiments. However, wireworm mortality did not exceed 25% in shade house studies with *S. feltiae* against and array of wireworm species (Ester and Huiting 2007, Ansari et al. 2009, Campos-Herrera and Gutierrez 2009, Sandhi et al. 2020). While these results are disappointing through the lens of a biopesticide insecticide application, these results do not reflect the long-term effectiveness of the season-long efficacy across a several month growing season.

The focus of this study was to examine the efficacy of persistent native NY EPNs against wireworms in an organic NY production system.

Materials and Methods

This set of experiments was located on the Hudson Valley Farm Hub located near Hurley, NY. The field was a sandy loam and was planted to a rye cover crop the previous season. The experiment was established with three treatments and four replicates. Each experimental plot measured 3.7 m wide by 30 m long. The placement of each treatment within each replicate was randomized.

Each plot within a replicate was separated by 15 m and replicates were separated by 10 m to reduce potential cross contamination from entomopathogenic nematode (EPN) movement within and across growing seasons. In the initial year, four soil hills were formed on 0.9 m centers the entire length of the plot (30 m), prior to the application of EPNs. After EPN application for the 3-year duration of the experiment, all tillage work was regulated. The untreated checks were tilled first across all replicates before the EPN plots to reduce the probability of contamination from the tillage equipment. Each EPN treatment was then tilled across replicates with the equipment cleaned between treatments.

The EPN species/strains used in this study were *Steinernema carpocapsae* (Weiser) 'NY 01', *Steinernema feltiae* (Filipjev) 'NY 04' and *Heterorhabditis bacteriophora* Poinar 'Oswego' Poinar. *H. bacteriophora* 'Oswego' was initially isolated from soil samples collected in 1990 from Oswego County, NY, *S. carpocapsae* "NY 01" was initial isolated from soil samples in 1990 from Jefferson County, NY and *S. feltiae* 'NY 04', was initially isolated from soil samples collected from Jefferson County, NY in 2004. To maintain the ability of these strains to persist under NY conditions, each species was re-isolated from the field every second year beginning in 2007, and used to reinitiate the laboratory culture (Shields and Testa 2015). The EPN strains used in this trial were re-isolated from Northern NY agricultural fields in 2016. Greater wax moth, *Galleria mellonella* (L.), larvae (Woodring and Kaya 1988) were used as hosts to maintain the nematode cultures (Vanderhorst Wholesale, St. Mary, OH 45885). Between field isolations, culturing protocols have been modified to preserve the genes for persistence in the population during the two years of laboratory culturing (Shields 2015). A *Galleria* based non-white trap rearing system (Testa and Shields 2017) was used for the production of IJs for field application.

Prior to the application of EPNs, the experimental areas were pre-sampled for the presence of native EPNs (27 April 2017) in the same manner as the post treatment samples. Entomopathogenic nematode treatments were applied on 23 May 2017. Treatment one was a species mix of *S. carpocapsae* 'NY 01' (Sc) + *S. feltiae* 'NY 04' (Sf) at a rate of 250 million Sc infective juveniles (IJs) per ha and 170 million Sf IJs per ha. Treatment two was a species mix of Sf + *H. bacteriophora* 'Oswego' (Hb) at a rate of 170 million Sf IJs per ha and 250 million Hb IJs per acre. In both treatments, the total 420 million IJs per ha were applied. Treatment three was an untreated check.

EPNs were applied to the soil surface using a modified ATV small plot sprayer with all screens and filters removed and calibrated to apply 945 L per ha through fertilizer stream nozzles (TeeJet™ 0010, Springfield, IL) mounted 30 cm apart. Application timing was late in the day to allow the UV sensitive IJs to enter the soil with limited UV exposure. After Treatment 1 was applied, the sprayer was thoroughly washed before being used to apply treatment 2.

EPN sampling protocol: All individual plots (including the untreated control plots) were sampled for EPNs 30d (22 June 2017), 150 d (20 Oct. 2017), 390 d (19 June 2018), 487 d (24 Sept. 2018), 694 d (19 April 2019), 860 d (10 Oct. 2019) and 1,076 d (5 May 2020) after application. At each sampling date, a total of 25 soil cores (2 cm × 20 cm) were collected from each plot and returned to the

laboratory to be bioassayed for the presence of EPNs. At the time of collection, the top 7 cm was placed in a 100 ml plastic cup with lid and the lower 13 cm was placed in a 240 ml cup with lid. Each container had a tight fitting lid. Soil cores were divided in this manner to isolate Sc in the upper layers from Sf in the lower layers in the Sc + Sf treatment (Trt 1) for the bioassay. Likewise, soil cores were divided in this manner to isolate Sf in the upper layers from Hb in the lower layers in the Sf + Hb treatment (Trt 2) for the assay (Ferguson et al. 1995). A similar procedure was followed for the untreated checks to detect any EPN contamination.

All soil samples were laboratory bioassayed using *G. mellonella* larvae as indicator hosts (5 larvae per 7 cm core, 10 larvae per 13 cm core). Before insect larvae were added, soil dryness was checked and the soil sample was misted if too dry. Samples were incubated at room temperature (23 °C), on shelves in the laboratory for 7 d. Dead *G. mellonella* were examined for nematode infection by observing the condition and color of the cadaver (Poinar 1984). Cadaver coloration between Sc, Sf and Hb is uniquely different (light brown, dark brown, brick red respectively) and cannot be confused. In some instances, the cadaver coloration between Sc and Sf looks similar, so cadavers were then placed on moist plaster of Paris disks in Petri dishes (White 1927) ("White trapped"), and observed for IJ emergence. Isolated IJs were then used to infect *G. mellonella* larvae, dissecting out the adult males and verifying the EPN species with the shape of the male spicule head, a separating characteristic between Sc and Sf (Neumann 2007).

Wireworm sampling: During the first two weeks of June 2017, in areas of rye cover crop adjacent to each plot (untreated and EPN treated), wireworm larval bait stations consisting of eight cut potato pieces in a mesh bag were buried to the depth of 30 cm and left in place for 14 days. After retrieval, contents of each bait station was examined for the presence of wireworm larvae and the larvae present were identified.

Crop procedures.

2017–2018: Sweet potato, *Ipomoea batatas* (L.) Lam 'Covington', slips (20–30 cm) were planted into EPN trial plots on 30 May 2017 and 31 May 2018. In both years, slips were planted into ridges by hand at 10" in-row spacing. Fertilizer was applied at rate of 91 Kg N, 23 Kg P2O5, 23 Kg K2O /hectare. Weed control in planted ridges included two mechanical cultivations in June and hand pulling weeds the remainder of the season. Buffer zones around plots were over-seeded in rye cover crop and mowed twice during growing season. There were no applications of pesticides and no supplemental irrigation to plots or buffer zones in the field.

Sweet potatoes (SP) were harvested on 26 Sept. 2017 and 24 Sept. 2018. SPs were mechanically lifted out of the ridges which placed the SP on top of the ridge it grew in. In each plot/rep a total of 200 potatoes were harvested. Fifty potatoes were randomly harvested from each of the four ridges in a plot. SPs from the two outer ridges were collected and binned separately from the SPs collected from inner ridges. SPs were cured for one week and stored at ~ 55 °F during the damage assessment period. Damage assessments took place the week of 23 Oct. 2017 and 15 Oct. 2018. In 2017, a total of 200 SPs from each plot rep were evaluated including 100 "inner ridge" SP samples and 100 "outer ridge" SP samples. Within each treatment and grouping of SP, damage was incidence of wireworm feeding (0 = none, 1 = observed), number of wireworm mines and weight of the SP in each plot. In 2018, assessment for white grub damage was added to the data set and was recorded as the number of inches of grub channels observed on the surface of the SP. Wireworm damage assessment remained the same as 2017 except weight was recorded for each 100 SP within each treatment.

2019: In 2019, Irish potatoes, *Solanum tuberosum* L.'Eva' were planted in the research plots rather than sweet potatoes due to increased attractiveness to wireworms. The potatoes were hand planted on 7 May 2019. Plots were slightly modified; three rows of potatoes (15 spuds per row) 10 feet in length that were used in observations. Fertilizer program was similar to the one used in 2017 & 2018. No supplemental irrigation was used and no pesticides were used in the plots. Plots were hand weeded after planting. Plots were harvested on 15 August 2019. On this date, 30 potatoes were dug from within each plot and examined for wireworm feeding.

Statistical Analysis: The study was designed as a randomized complete block design with four replications using three treatments (EPN species mix 1, EPN species mix 2, & untreated). Wireworm feeding damage was evaluated using analysis of variance for a Random Complete Block Design (ANOVA) with post-hoc t-test applying Bonferroni correction (Systat Software Inc. 2009).

EPN population levels expressed in percent of soil samples with a positive bioassay for the presence of EPNs were normalized with Arcsine transformation before analysis. Significant differences in populations between years was tested using analysis of variance for a Random Complete Block Design (ANOVA) with post-hoc t-test

Table 1. Percent of soil cores bioassayed as positive for the presence of entomopathogenic nematodes after a single application (% ± SE).

Days after application	30d	150d	390d	490d	694d	860d	1076d
Sf × Hb							
Sf	30±3 a	28±3 a	28±2 a	33±4 a	30±2 a	33±3 a	28±1 a
Hb	1±1 a	1±1 a	3±3 a	2±1 a	2±1 a	1±1 a	3±2 a
Sc × Sf							
Sc	2±1 a	1±1 a	2±2 a	0	0	6±3 a	1±1 a
Sf	29±4 a	24±2 b	34±4 a	28±4 a	35±5 a	27±3 a	28±3 a

Numbers followed by the same letter within a row are not significantly different (p= 0.05)

applying Bonferroni correction (Systat Software Inc. 2009).

Results

The wireworms collected from the plot area using subsurface bait stations were identified as a mix of the Eastern field wireworm, *Limonius agonus* (Say), corn wireworm, *Melanotus communis* (Gyllenhal) and *Glyphonyx inquinatus* (Say). There was little relation between the low number of wireworm collected in the subsurface bait stations and resulting damage to the sweet potatoes. As a result, wireworm baiting was discontinued during the remainder of the study.

EPN persistence: Throughout the duration of the experiment (3 years), no EPNs were detected in the untreated control plots.

Sf × Hb: Bioassay results for Sf ranged from 28–33% of the soil samples positive for the presence of Sf across the 1076 days (3 yrs) of the study. The levels of Sf remained significantly unchanged throughout the duration of the study ($F = 0.37$; df = 6; $P = 0.05$). The levels of Hb also were not significantly different across the 1076 days with the results ranging from 1–3% of the soil samples positive for the presence of Hb ($F = 0.48$; df = 6; $P = 0.05$). (Table 1, Fig. 1).

Sc × Sf: Bioassay results for Sf ranged from 24–35% of the soil samples positive for the presence of Sf across the 1076 days (3 yrs) of the study. The levels of Sf remained significantly unchanged across the duration of the study with the exception of the 150 d bioassay ($F = 0.67$; df = 6; $P = 0.05$) where the level of Sf dipped significantly below the mean level (24% vs 30%). This may have been a sampling issue since the levels increased to the former level for the remainder

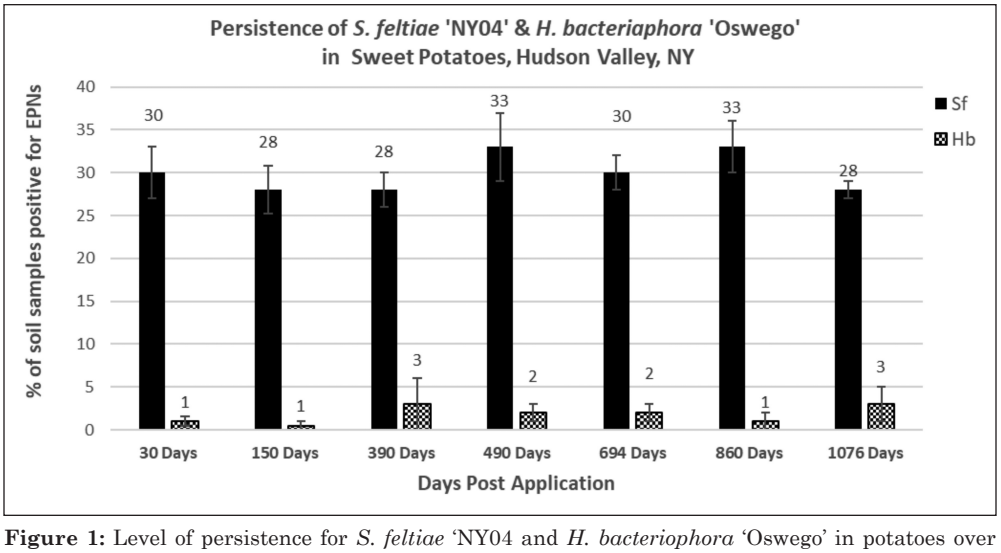

Figure 1: Level of persistence for *S. feltiae* 'NY04 and *H. bacteriophora* 'Oswego' in potatoes over multiple growing seasons in Hudson Valley, NY (% ± SE).

Table 2: Wireworm feeding wounds on sweet potatoes at harvest during year 1 and year 2. The damage was separated by the position of the 4 rows of the plot. Inside = the two inside rows and Outside = the two outside rows adjacent to the rye cover crop (# of feeding wounds ± SE).

	Year 1		Year 2	
Treatment	**Outside rows**	**Inside rows**	**Outside rows**	**Inside rows**
Sf × Hb	30 ± 2 a	41 ± 19 a	14 ± 4 a	11 ± 3 a
Sc × Sc	62 ± 19 b	55 ± 17 ab	24 ± 5 b	10 ± 3 a
UTC	73 ± 37 b	73 ± 31 b	27 ± 6 b	9 ± 4 a

Numbers followed by the same letter within a column are not significantly different (p= 0.05)

of the study. The levels of Sc also were not significantly different across the 1076 days with the results ranging from 0–6% of the soil samples positive for the presence of Sc ($F = 0.58$; df = 6; $P = 0.05$). (Table 2, Fig. 2).

In addition, the levels of Sf were statistically identical when comparing levels of Sf across the two treatments ($F = 0.35$; df = 13; $P = 0.05$).

Wireworm feeding damage: In the first harvest (2017), the EPN combination of Sf × Hb had significantly less wireworm feeding damage than the untreated check irrespective of whether the plants were located in the outside rows or the inside rows ($F = 2.39$; df = 23; $P = 0.01$). The EPN combination of Sc × Sf was numerically different from the untreated checks but the fewer wireworm feeding wounds were not statistically different from the untreated check ($F = 0.95$; df = 7; $P = 0.05$). When comparing the outside rows between the two EPN combinations, the Sf × Hb combination had significantly fewer feeding wounds than the Sc × Sf combination ($F = 2.15$; df = 11; $P = 0.05$). However, when comparing the inner rows between the two EPN combinations, the numerical difference was not statistically different ($F = 1.05$; df = 11; $P = 0.05$).

At the second year harvest, the level of wireworm feeding wounds across all treatments were reduced from year 1. Comparing the outside rows across treatment, only the EPN combination of Sf × Hb has significantly less damage than either the untreated control plots or the Sc × Sf combination. ($F = 2.05$; df = 11; $P = 0.05$). At the third year harvest, no wireworm damage was recorded in any of the treatments (Fig. 3).

Discussion

EPN levels: The levels of Sf in both of the nematode species combinations were not significantly different from each other

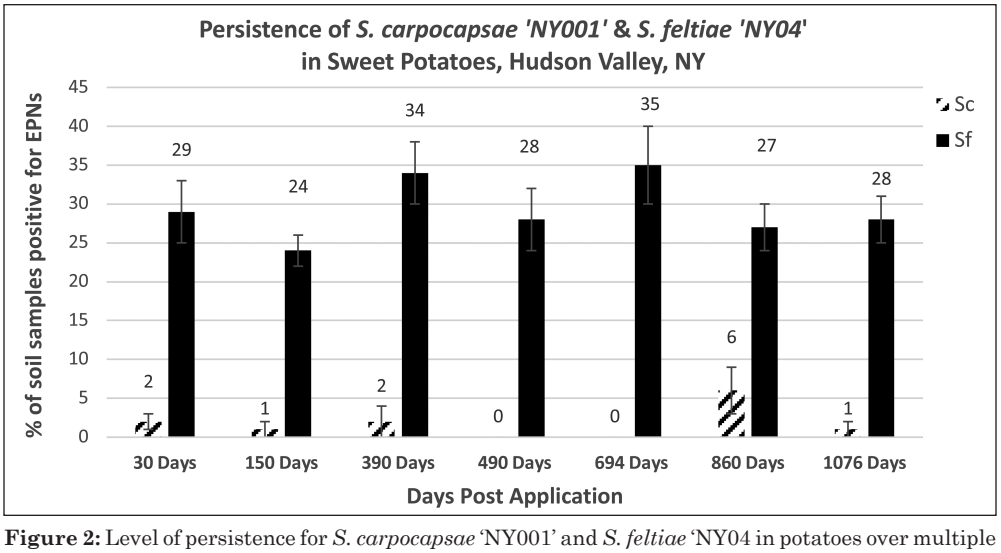

Figure 2: Level of persistence for *S. carpocapsae* 'NY001' and *S. feltiae* 'NY04 in potatoes over multiple growing seasons in Hudson Valley, NY (% ± SE).

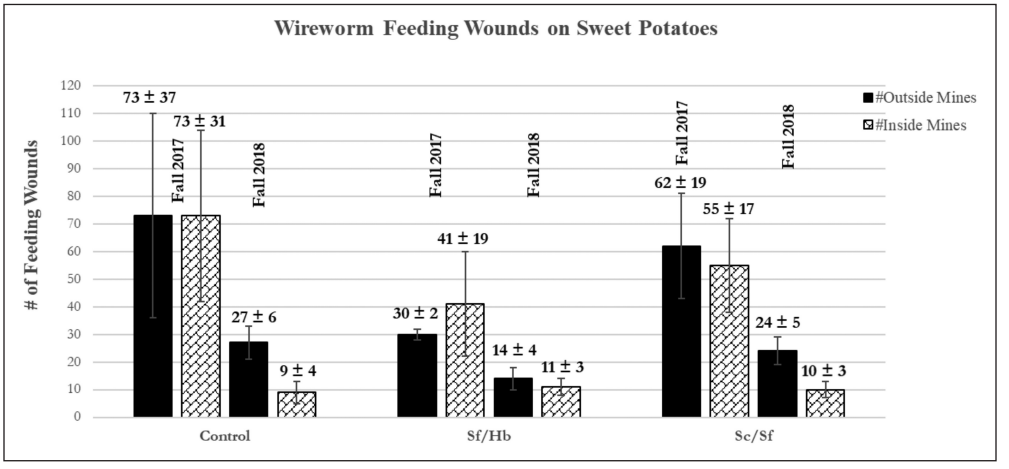

Figure 3: Wireworm feeding wounds on sweet potatoes at harvest during year 1 and year 2. The damage was separated by the position of the 4 rows of the plot. Inside = the two inside rows and Outside = the two outside rows adjacent to the rye cover crop (# of feeding wounds ± SE).

and the level of Sf (24–35%) is very similar to the long-term persistence levels reported by Shields et al. (2018) across 75 fields ranging from clay loam to sandy loam. In the multi-year and multi-field study reported in Shields et al. (2018), the long-term persistence level of Sf (NY04) is suggested to be in the 20–30% range under NY agricultural conditions. Results reported in this study were in line with levels reported in Shields et al. (2018). The levels of Sc (NY01) were lower in this study (0–6%) than reported by Shields et al. (2018) in alfalfa fields (8–13%), but more closely matched the levels reported in continuous corn (1–14%) or alfalfa following corn (1–6%). Ferguson et al. (1995) reported Sc preferred the top 5–7 cm of the soil profile and this zone can become very dry in sandy loam soils when row crops are grown. This may explain the lower level of Sc in this study along with the reported levels in continuous corn in Shields et al. (2018). In addition, the ambush nature of Sc along with limited dispersal behavior (Kaya and Gaugler 1993) often results in Sc hotspots separated by areas without Sc, resulting in a lower reported level of Sc than actually is present in the field. When Sf is matched with Sc, Sf ranges deeper in the soil and is less effected by the dry upper soil layers (Ferguson et al. 1995; Neumann and Shields 2006, 2008, 2011) coupled with a hybrid searching behavior using both ambush and cruising strategies. When these two species are mixed, data suggests that Sf fills in the gaps between the Sc areas of concentration (hotspots) resulting in a more complete coverage of the soil environment.

The levels of Hb in this study range from 1–3% of the soil samples across the duration of the study. With the relatively low density of hosts in this study, these low levels are not unexpected. Hb is a cruising nematode resulting in two issues; 1) this behavior matched with the bioassay technique of removing a soil sample for laboratory bioassay significantly underestimates the presence of Hb in the soil profile searching for hosts, 2) Hb numbers rise after the host has increased to economic numbers and 3) Hb prefers to attack larger larvae, often after damage has occurred to the crop (Shields et al. 1999). Hb numbers can rise to 100% of the soil samples in the presence of large numbers of hosts (Shields et al. 1999), but a more typical range under moderate host densities are 2–10% (Shields and Testa 2020). The very presence of Hb 1076 d after inoculation indicated that Hb is established in the soil and available to respond to host invasion.

Acknowledgments

We thank USDA Specialty Block Grant, NY Farm Viability Institute and Cornell University for providing funds for this study.

Literature cited

Abate, B. A., M. J. Wingfield, B. Slippers, and B. P. Hurley. 2017. Commercialisation of entomopathogenic nematodes: should import regulations be revised? Biocontrol Science and Technology 27: 149–168.

Ansari, M. A., M. Evans, and T. M. Butt. 2009. Identification of pathogenic species of ento-

mopathogenic nematodes and fungi for wireworm control. Crop Protection 28: 269–272.

Barsics, F., E. Haubruge, and F. J. Verheggen. 2013. Wireworms' management: an overview of the existing methods, with particular regards to Agriotes spp. (Coleoptera: Elateridae). Insects 4: 117–152.

Campos-Herrera, R. 2015. Nematode pathogenesis of insects and other pests. Series: sustainability in plant and crop protection. Springer International Publishing, Switzerland.

Campos-Herrera, R. and C. Gutiérrez. 2009. Screening Spanish isolates of steinernematid nematodes for use as biological control agents through laboratory and greenhouse microcosm studies. Journal of Invertebrate Pathology 100: 100–105.

Dillon, A. B., D. Ward, M. J. Downes, and C. T. Griffin. 2006. Suppression of the large pine weevil *Hylobius abietis* (L.) (Coleoptera: Curculionidae) in pine stumps by entomopathogenic nematodes with different foraging strategies. Biological Control 38: 217–226.

Duncan, L. W., J. H. Graham, D. C. Dunn, J. Zellers, C. W. McCoy, and K. Nguyen. 2003. Incidence of endemic entomopathogenic nematodes following application of *Steinernema riobrave* for control of *Diaprepes abbreviatus*. Journal of Nematology 35: 178–186.

Ester, A. and H. Huiting. 2007. Controlling wireworms (*Agriotes* spp.) in a potato crop with biologicals. Insect Pathogens and Insect Parasitic Nematodes IOBC wprs Bulletin 30: 189–196.

Ferguson, C. S., P. C. Schroeder, and E. J. Shields. 1995. Vertical distribution, persistence and activity of entomopathogenic nematodes (Nematoda: Heterorhabditidae and Steinernematidae) in alfalfa snout beetle (Coleoptera: Curculionidae) infested fields. Environmental Entomology 24: 149–158.

Kaya, H. K and R. Gaugler. 1993. Entomopathogenic Nematodes. Annual Review of Entomology 38:181–206.

Kaya, H. K., M. M. Aguillera, A. Alumai, H. Y. Choo, M. de la Torre, A. Fodor, S. Ganguly, S. Hazir, T. Lakatos, A. Pye, M. Wilson, S. Yamanaka, H. Yang, and R.-U. Ehlers. 2006. Status of entomopathogenic nematodes and their symbiotic bacteria from selected countries or regions of the world. Biological Control 38: 134–155.

Knodel, J. J. and G. Shrestha. 2018. Pulse crops: pest management of wireworms and cutworms in the Northern Great Plains of United States and Canada. Annals of the Entomological Society of America 111: 195–204.

La Forgia, D. and F. Verheggen. 2019. Biological alternatives to pesticides to control wireworms (Coleoptera: Elateridae). Agri Gene. 11: 100080. Available from https://doi.org/10.1016/j.aggene.2018.100080

Labrie, G., A. È. Gagnon, A. Vanasse, A. Latraverse, and G. Tremblay. 2020. Impacts of neonicotinoid seed treatments on soil-dwelling pest populations and agronomic parameters in corn and soybean in Quebec (Canada). Plos One. 15: e0229136. Available from https://doi.org/10.1371/journal.pone.0229136

Lacey, L. A., D. Grzywacz, D. I. Shapiro-Ilan, R. Frutos, M. Brownbridge, and M. S. Goettel. 2015. Insect pathogens as biological control agents: back to the future. Journal of Invertebrate Pathology 132: 1–41.

Lewis, E. E., J. Campbell, C. Griffin, H. Kaya, and A. Peter. 2006. Behavioral ecology of entomopathogenic nematodes. Biological Control 36: 66–79.

Neumann, G. 2007. Dual-strategy biological control of the alfalfa snout beetle, *Otiorhynchus ligustici* L. (Coleoptera: Curculionidae), using persistent entomopathogenic nematodes in a single vs multispecies natural enemy approach. Ph.D Thesis. Cornell University, Ithaca, NY. pp. 113.

Neumann, G. and E. J. Shields. 2006. Interspecific interactions among three entomopathogenic nematodes, *Steinernema carpocapsae*, *S. feltiae*, and *Heterorhabditis bacteriophora*, with different foraging strategies for hosts in multi-piece sand columns. Environmental Entomology 35: 1578–1583.

Neumann, G. and E. J. Shields. 2008. Multiple-species natural enemy approach for the biological control of the alfalfa snout beetle, *Otiorhynchus ligustici* L. (Coleoptera: Curculionidae), using entomopathogenic nematodes. Journal of Economic Entomology 101: 1533–1539.

Neumann, G. and E. J. Shields. 2011. Long term field persistence and coexistence in three entomopathogenic nematodes, *Steinernema carpocapsae* Weiser, *S. feltiae* Filipjev, and *Heterorhabditis bacteriophora* Poinar. The Great Lakes Entomologist 44: 42–52.

Paquet-Walsh, A., A. Bertolo, C. Landry, L. Deschamps, and M. Boily. 2019. Interactive effects of neonicotinoids and natural ultraviolet radiation on yellow perch (*Perca flavescens*) larvae. Science of the Total Environment 685 (2019) 690–701. Available from https://doi.org/10.1016/j.scitotenv.2019.05.113

Parker, W. E. and J. J. Howard. 2001. The biology and management of wireworms (*Agriotes* spp.) on potato with particular reference to

the U.K. Agricultural and Forest Entomology 3: 85–98.

Parker, W.E., A. Clarke, S.A. Ellis, and J.N. Oakley. 1990. Evaluation of insecticides for control of wireworms (*Agriotes* spp.) on potato. Tests of agrochemicals and cultivars No. 11. Annals of Applied Biology 116 (Supplement), 28–29.

Poinar, G. O. 1984. The natural history of nematodes. Prentice-Hall, Englewood Cliffs, NJ.

Rojht, H., M. KaČ, and S. Trdan. 2009. Non-target effect of entomopathogenic nematodes on larvae of twospotted lady beetle (Coleoptera: Coccinellidae) and green lacewing (Neuroptera: Chrysopidae) under laboratory conditions. Journal of Economic Entomology 102: 1440–1443.

Sandhi, R. K. and G. V. P. Reddy. 2019. Effects of entomopathogenic nematodes and symbiotic bacteria on non-target arthropods, pp. 247–273. In Md. A. Khan and W. Ahmad (eds.), Microbes for sustainable insect pest management. Springer, Cham. Sandhi.

Sandhi, R.K., D. Shapiro-Ilan A. Sharma, and G. V.P. Reddy. 2020. Efficacy of entomopathogenic nematodes against the sugarbeet wireworm, *Limonius californicus* (Mannerheim) (Coleoptera: Elateridae). Biological Control 143: Available from https://doi.org/10.1016/j.biocontrol.2020.104190

Shapiro-Ilan, D. I., D. H. Gouge, and A. M. Koppenhöfer. 2002. Factors affecting commercial success: case studies in cotton, turf and citrus, pp. 333–356. In R. Gaugler (ed.), Entomopathogenic nematology. CABI Publishing, New York.

Shields, E. J. 2015. Ch 6—Utilizing persistent EPNs in a conservation or a more classical biological control approach. In: R. Campos-Herrera (ed). Nematode pathogenesis of insects and other pests–ecology and applied technologies for sustainable plant and crop protection, Series: Sustainability in Plant and Crop Protection, Vol. 1 (R. Campos-Herrera ed). Springer. 2015. Pp. 165–184.

Shields, E. J., A. Testa, J. M. Miller, and K. L. Flanders. 1999. Field efficacy and persistence of the entomopathogenic nematodes *Heterorhabditis bacteriophora* 'Oswego' and *H. bacteriophora* 'NC' on alfalfa snout beetle larvae (Coleoptera: Curculionidae). Environmental Entomology 28: 128–136.

Shields, E. J. and A. M. Testa. 2015. Ch 11—New York Case Study: Biological Control of *O. ligustici* with native persistent EPNs using a more classical approach. In: R. Campos-Herrera (ed). Nematode pathogenesis of insects and other pests–ecology and applied technologies for sustainable plant and crop protection, Series: Sustainability in Plant and Crop Protection, Vol. 1 (R. Campos-Herrera ed). Springer. 2015. pp 285–307.

Shields, E. J., A. M. Testa, and W. J. O'Neil. 2018. Long-term persistence of native NY entomopathogenic nematode isolates across crop rotation. Journal of Economic Entomology 111: 2592–2598.

Shields, E. J. and A. M Testa. 2020. Multi-year Biological Control of Black Vine Weevil, *Otiorhynchus sulcatus*, with Persistent Entomopathogenic Nematodes. The Great Lakes Entomologist 53: 117–123.

Systat Software Inc. 2009. SigmaPlot 11.2. User's guide, part 2 statistics. Systat Software Inc., San Jose, CA.

Testa, A. M. and E. J. Shields. 2017. Low Labor "in vivo" Mass Rearing Method for Entomopathogenic Nematodes. Biocontrol 106: 77–82.

Toba, H. H., J. E. Lindegren, J. E. Turner, and P. V. Vail. 1983. Susceptibility of the Colorado potato beetle and the sugarbeet wireworm to *Steinernema feltiae* and *S. glaseri*. Journal of Nematology 15: 597–601.

Traugott, M., C. M. Benefer, R. P. Blackshaw, W. G. van Herk, and R. S. Vernon. 2015. Biology, ecology, and control of elaterid beetles in agricultural land. Annual Review of Entomology 60: 313–334.

Vernon, R. S., W. G. van Herk, M. Clodius, and C. Harding. 2009. Wireworm management I: stand protection versus wireworm mortality with wheat seed treatments. Journal of Economic Entomology 102: 2126–2136.

White, G. F. 1927. A method for obtaining infective nematode larvae from cultures. Science. 66: 302–303.