Title: **Efficacy Evaluation of Biological Control Agents Against Wireworms in Organic Production**

Principal Investigator:
Arash Rashed, Associate Professor, Department of Entomology, Plant Pathology and Nematology, University of Idaho, Moscow, ID, arashed@uidaho.edu, 208-885-5977.

Co-Principle Investigator:

Diane Green, Producer Advisor, Greentree Natural Certified Organic Farm, Sandpoint, ID, diane@greentreenaturals.com, 208-263-8957.

Collaborators:
Atoosa Nikoukar, PhD Student, Department of Entomology, Plant Pathology and Nematology, University of Idaho, Moscow, ID, anikoukar@uidaho.edu, 205-207-0345.

Greg Sempel, Producer, Pokey Creek Certified Organic Farm, Saint Maries, ID, pokeycreekfarm@gmail.com.

Kyle Nagy, Field Manager, University of Idaho Sandpoint Organic Agriculture Center, Sandpoint, ID, knagy@uidaho.edu, 208-304-7815.

Report is prepared by A. Nikoukar

Project Summary:

Wireworms are subterranean herbivores and the larval stage of click beetle (Coleoptera: Elateridae) species. There are several species of wireworms which can cause serious damage to crops in both conventional and organic production systems. Their long-life cycle, subterranean habitat, wide host range and their ability to survive in the absence of host plants make their management challenging. Although there are a few insecticides to control wireworms in conventional cropping systems, organic producers are left without effective alternative options to manage this pest. The main objective of this proposal was to evaluate, in a series greenhouse and on-farm trials, the effectiveness of several biological control agents, applied either alone or in combination (mixed application), in reducing wireworm damage and numbers. Unique to this proposal was to collect, rear and reapply field-collected entomopathogenic nematodes (EPN) to field plots, along with treatments including commercial EPNs and entomopathogenic fungi (EPF).

The sugar beet wireworm, *L. californicus*, was the species used in all greenhouse assays. Our greenhouse treatments included the EPN, *Steinernema feltiae* (both commercial and field-collected), and the EPF, *Beauveria bassiana* and *Metarhizium brunneum*. Local and commercial EPNs, *M. brunneum*, and combined EPN/*Metarhizium* resulted in significantly higher wireworm mortality than the nontreated controls. Our greenhouse findings also indicated that combining EPN/EPF applications does not consistently result in increased wireworm mortality.

On-farm trials were conducted in three certified organic vegetable farms in the northern Idaho, where a pre-planting survey confirmed the presence of wireworm species *Limonius californicus* (aka., sugar beet wireworm) and *L. infuscatus* (aka., western field wireworm). The treatment *M. brunneum* was not included in the field trials. Combining the application of *S. feltiae* and *B. bassiana* did not improve the efficacy of biological control against wireworms when applied at-planting. However, the application of local and commercial *S. feltiae*, and also *B. bassiana*, significantly reduced the number of wireworms. We also demonstrated that EPN and EPF may also persist in the environment, especially if applied alone.

Introduction:

Pest management is one of the most important actions in crop production. Wireworms which are the immature stage of click beetles (Coleoptera: Elateridae) are considered as one of the most damaging pests in a variety of crops in the Pacific Northwest region of the USA. Depending on species and environmental conditions, the larval stage can take up to 11 years (Vernon et al. 2008, Traugott et al. 2015), during which they continue to feed on belowground plant tissues like root, stem, and seed. Wilting, stunt growth, and plant death can be caused by wireworm feeding (Traugott et al. 2015). The damaged underground tissues will also become susceptible to secondary pathogen infections which can result in reduced crop quality, especially in vegetable crops (Traugott et al. 2015). Historically, wireworms were controlled using broad spectrum insecticides; many of those highly toxic insecticides are no longer available in the USA because of their harmful effects on human health, environment and non-target organisms (Vernon et al. 2008). Over the years and as residues of those environmentally persistent broad-spectrum insecticides are fading away (Vernon et al. 2008), wireworms have re-surged as a serious pest in most of agricultural crops (Parker and Howard 2001). There is a critical need for identifying alternative (to insecticides) effective control measures to mitigate the impact of this major pest.

Besides some cultural practices like alfalfa rotation (Shirck 1945), applying biological control agents have been recommended as a practical method against some species of wireworms (Ansari et al. 2009, Razinger et al. 2013, Reddy et al. 2014). Some recent field and laboratory studies have shown the effectiveness of the EPN, *Steinernema carpocapsae,* and the EPF, *Metarhizium anisopliae,* against sugar beet wireworm (e.g., Milosavljević et al. 2016). In a series of field and laboratory experiments conducted by Sufyan et al. (2017), application of the EPF, *Beauveria bassiana*, caused sufficient wireworm mortality and reduced damage in both laboratory and field (Sufyan et al. 2017). In spite of some promising results in biological control against wireworms, there are also inconsistencies in the reported effectiveness of EPNs. The efficacy of EPN can vary with their species, the species of the host, and environmental conditions (Grewal et al. 2005, Lewis et al. 2006). Field-collected EPNs which are expected to be adapted to the environmental conditions and prey types in their local habitat, may be better candidates to be used as biological control agents in a given field (Campos-Herrera and Gutiérrez 2014, Morton and Garcia-del-Pino 2017, Nikoukar et al., in press). In a series of recent laboratory bioassays, we found that field-collected isolates of *S. feltiae* can be more effective against sugar beet wireworms than the commercially obtained species of *Heterorhabditis bacteriophora* and *Steinernema carpocapsae* (Nikoukar et al., in press). However, we also demonstrated that field-collected isolates of *S. feltiae* may have different efficacies against sugar beet wireworms, with some exceeding the efficacy offered by the commercial isolate of the same EPN species (Nikoukar et al., in press). This finding of the effectiveness of local EPNs against sugar beet wireworm, the most common wireworm species in Idaho (Rashed et al. 2015), motivated us to propose this project in organic farms with the aim to reduce wireworm pressure using local field-collected EPNs along with the available commercial EPNs and EPFs. We also evaluated the efficacy of a combination of EPF and EPN against the sugar beet wireworm to understand whether the interaction between the two biocontrol agents could increase, or reduce, the efficacy of our biological control approach. Through two years of monitoring of three experimental organic farms, *L. californicus* and *L. infuscatus* were identified as the predominant wireworm species in our experimental plot areas. These two species are also the most commonly found wireworms in conventional production systems of Idaho (Rashed et al. 2015).

Objectives Statement: The overall goal of this project was to explore biological control as an alternative management tactic to control wireworms in organic farms. There were two complementary objectives to achieve this goal:

Objective 1) Evaluate and compare the effectiveness of the entomopathogenic fungus (EPF), *Metarhizium brunneum* (but see below), the field-collected EPN *S. feltiae* (local EPN, collected from the same field) and the commercial EPN *S. feltiae* (commercial-EPN), against wireworms; our sub-objectives were the following:

> *1a.* Determine efficacies of EPF, local and commercial EPNs, and mixed EPF/EPN application against wireworms in field soil under controlled greenhouse conditions,
>
> *1b.* Quantify and compare efficacies of EPF, local and commercial EPNs, and mixed EPF/EPN application in wireworm-infested organic farms.
>
> *1c.* Examine the survival of the applied local and commercial EPNs at the end of the trial.

Objective 2) Disseminate findings to regional organic producers.

Modifications to the original objectives: In the second subobjective, we initially proposed to apply *M. brunneum* as our EPF treatment, however since *M. brunneum* is not allowed in certified organic production (and is no longer available on market shelves), we replaced that treatment with *B. bassiana* in our on-farm trials. However, both *M. brunneum* and *B. bassiana* were included in our greenhouse assays (one additional treatment).

Materials and Methods:

The main goal of this objective was to determine the efficacies of the EPFs, *M. brunneum* and *B. bassiana*, as well as local EPN (*S. feltiae*) and commercial EPN (*S. feltiae*), and the two mixes of local EPN/EPFs and commercial EPN/EPFs against wireworms.

The local EPNs were extracted from the soil in each farm, using Galleria traps (*Galleria mellonella* L.; Lepidoptera: Pyralidae) (White 1927). For that, three subsamples of 250 ml of mixed soil per location were removed and mixed, and 7 *Galleria* larvae were placed on top of the soil in a plastic container. After 2-3 days, the dead larvae were placed on White traps to recover the infective juveniles (IJs) (Bedding and Akhurst 1975) (Fig. 1). The collected IJs were incubated in 10 °C until application.

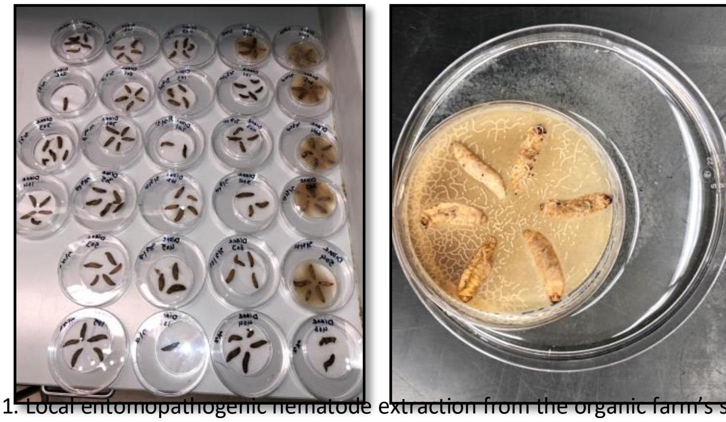

Figure 1. Local entomopathogenic nematode extraction from the organic farm's soil. To collect EPNs, infected waxworms were placed in the White trap, which is a 5cm petri dish placed inside a 10 cm petri dish filled with distilled water. Infective Juveniles (IJs) after emergence from waxworms' cadaver swim into the water which would then be collected and used in experiments.

Subobjective 1a. *Greenhouse trials*: The greenhouse study was conducted at the University of Idaho's Manis Entomological Laboratory greenhouses in Moscow, ID. Only the sugar beet wireworm, *L. californicus*, was used in this experiment. The experimental pots were cone-shaped and 4.2 × 20.32 cm (diameter × height) in size. Each pot was filled with soil from one organic field (Greentree Naturals). Before filling pots, soil was baked in an oven (82°C) for 30 min to reduce the chance of natural infestations with nematodes. A single wireworm was placed in each pot, 7 cm deep. A single organic black bean seed was also planted in each pot. There was a total of 10 treatments in each of the three time-blocks. Our 10 treatments included: 1) EPF, *M. brunneum*, 2) EPF, *B. bassiana*, 3) local EPN (*S. feltiae*), 4) commercial EPN (*S. feltiae*), 5) local EPN/EPF, *M. brunneum*, 6) commercial EPN/EPF, *M. brunneum*, 7) local EPN/EPF, *B. bassiana*, 8) commercial EPN/EPF, *B. bassiana*, 9) non-treated wireworm control, and 10) no-wireworm control. There were 12 pot-replicates per treatment per time-block. Treatments were arranged in a completely randomized design and kept in a greenhouse chamber with the average temperature of 25 ± 3 °C for 30 days (Fig. 2). All entomopathogen mixtures were suspended in 10 ml distill water and applied to the soil surface after placing the seed in ~2.5 cm depth. The 10 ml suspension volume per pot was used to prevent losing applied treatments through drainage. *M. brunneum* (EPF) (Met 52 EC, Evergreen Growers Supply LLC., Clackamas, OR) was applied at the rate of 1 ml per pot based on the highest rate recommended for drench application. *B. bassiana* (BotaniGard 22WP, ARBICO Organics CO., Oro Valley, OR), was applied at the rate of 0.16 gr EPF per pot. The application rate of commercial (Evergreen Growers Supply LLC., Clackamas, OR) and field-collected EPNs was ~ 8200 IJs per pot (suspended in 10 ml of distilled water). The mixed treatments of EPN/EPF were also first suspended in 10 ml water and then applied onto the surface before covering seed. Pots were watered no more than the soil water capacity. The non-treated control pots were inoculated with 10 ml distill water, only. The no-wireworm treatment was included as a control for emergence (germination) rate and wireworm damage. Wireworms were checked for mortality 30 days after treatment application, and the dead insects were removed and dissected under a stereomicroscope to confirm the presence of nematode and/or fungus infection. Live insects were individually placed in containers filled with autoclaved sand and were inspected after one additional week. Wireworm mortality, plant emergence and damage rate (percent seedlings with feeding damage) were compared using generalized linear mixed models. All statistical analyses were conducted in IBM-SPSS Statistics version 27.

Figure 2. Experimental setup for the greenhouse study.

Subobjective 1b. *On-farm trials*:

Study sites: There were three certified organic vegetable field locations: 1) University of Idaho Sandpoint Organic Agriculture Center (SOAC), Sandpoint, ID, 2) Greentree Naturals Organic Farm, Sandpoint, ID, and 3) Pokey Creek Organic Farm, Saint Maries, ID.

Wireworm pressure was assessed before planting and one month after treatment application (mid-season), using two solar bait traps per experimental plot. Each solar bait trap consisted of a mixture of soaked untreated organic wheat and barley seeds, buried 15-cm deep in the ground and covered with soil and a black plastic bag, according to Rashed et al. (2015). After two weeks, traps were removed and transported to the laboratory to collect and record the captured wireworms (Rashed et al. 2015). The number of experimental plots in each location varied with space availability (and the size of the infested area). We established 36, 30, and 24 black bean plots (10 ft-long row, each) in Greentree Naturals Organic Farm, University of Idaho SOAC, and Pokey Creek Farm, respectively. A total of six treatments were included in our on-farm trials: 1) EPF, *B. bassiana*, 2) local EPN (*S. feltiae*), 3) commercial EPN (*S. feltiae*); 4) local EPN (*S. feltiae*)/EPF (*B. bassiana*), 5) commercial EPN (*S. feltiae*)/EPF (*B. bassiana*) and 6) non-treated control. *B. bassiana* (EPF) was applied at the rate of 10 gr per plot (10 sq.ft.). Both commercial and local field-collected EPNs (*S. feltiae)* were applied at the rate of ~1,000,000 EPN per plot. All treatments were suspended in one gallon water and then applied in-furrow (on top of the planted seed). Plots were arranged in a randomized complete block design, with four, five, or six replicates per treatment, depending on space availability in each location (Fig. 3, 4). Black beans were planted in all locations. Stand count (2-week post emergence and at harvest), wireworm count (before planting and one month post emergence), and final yield were measured to estimate and compare effectiveness of the examined biological control treatments in managing wireworms. Generalized linear mixed models (repeated measures for wireworm and stand counts) were used to conduct comparisons (IBM-SPSS Statistics ver. 27).

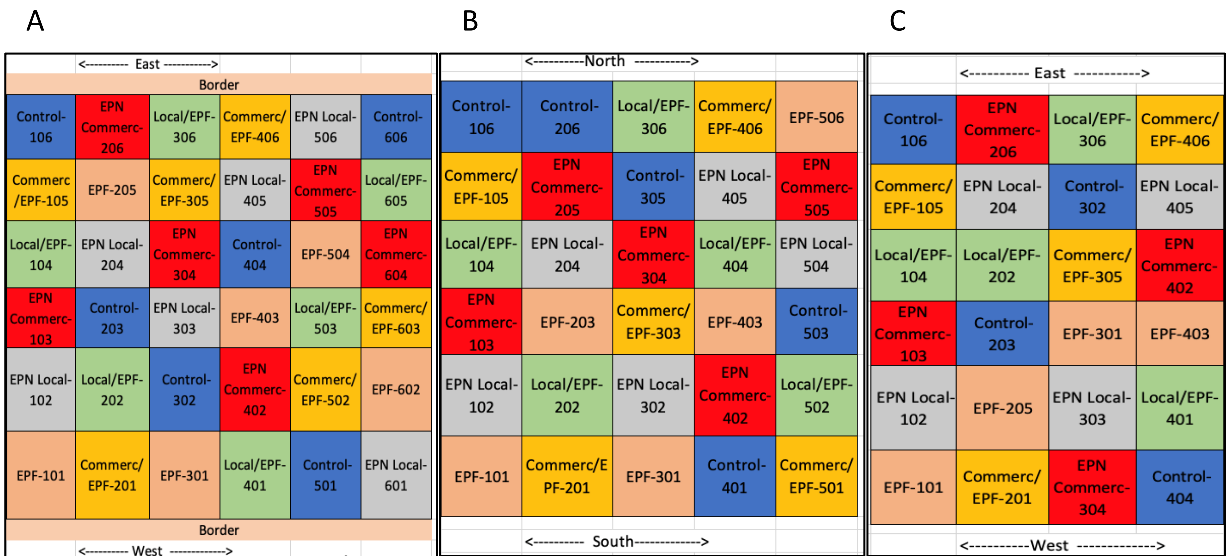

Figure 3. Experimental plot maps for the three study sites. Treatments were randomized within each row in each location. A) Greentree Naturals Organic Farm, Sandpoint, ID, B) University of Idaho Sandpoint Organic Agriculture Center (SOAC), Sandpoint, ID, and C) Pokey Creek Organic Farm, Saint Maries, ID.

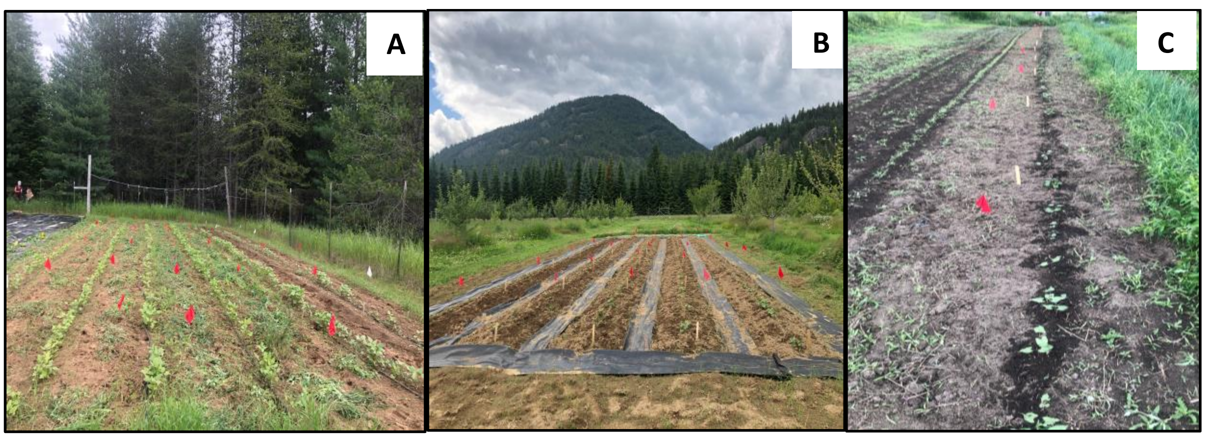

Figure 4. Experimental plots in three study sites. A) Greentree Natural Organic Farm, Sandpoint, ID, B) University of Idaho Sandpoint Organic Agriculture Center (SOAC), Sandpoint, ID, C) Pokey Creek Organic Farm, Saint Maries, ID.

Subobjective 1c. *Persistence of applied EPNs:*

To estimate the persistence of our applied biological control agents in the field, we collected two soil sample per plot using a 6-inch auger prior to planting, 3 weeks after planting, after harvest, and then prior to planting in the subsequent season in two field locations in Sandpoint, ID. In this report, we only presented EPN and EPF incidence in plots in the subsequent season (approximately a year after application). In each sampling, the two collected subsamples from each plot were mixed and 250 ml of the mixed soil (per plot) was placed in a plastic container. Five Galleria larvae were placed on the soil surface in each container and kept for 2-3 days in the room temperature. After 3 days, dead larvae were washed with distill water and transferred onto white traps to check for EPN (or EPF) infection (Fig. 5). Emerging EPN IJs from Galleria cadaver or EPF infections were used to determine the persistence of the applied biological control agents in the soil over time.

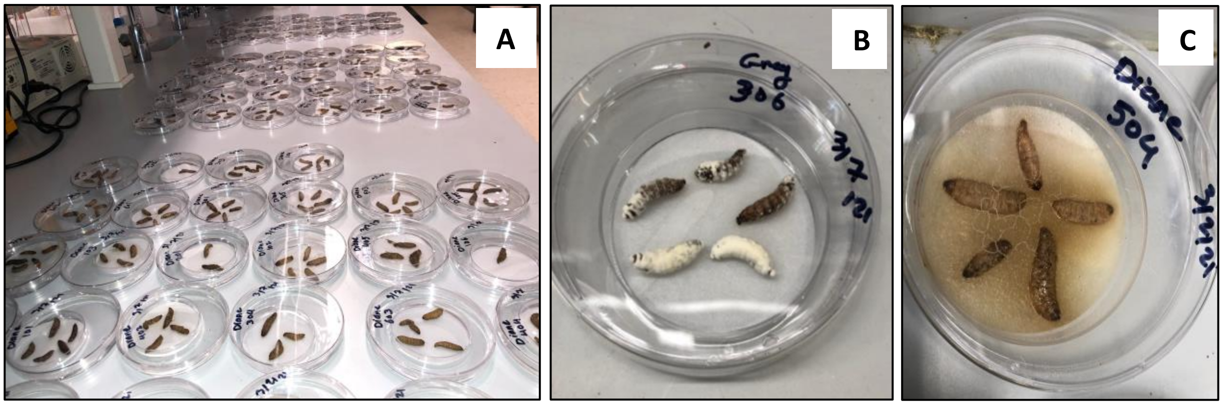

Figure 5. Evaluating the persistence of the applied entomopathogens: A) infected waxworms from soils collected from the three study sites one year after application, B) waxworms infected with entomopathogenic fungi (EPF), C) Waxworms infected with entomopathogenic nematodes (EPN).

Results:

<u>*Greenhouse study:*</u>

Wireworm mortality: Overall, a significant difference in wireworm mortality was detected among treatments (F= 2.07, df = 8, 306, P = 0.039). The commercial EPN (43%) and *M. brunneum* (43%) caused significantly higher mortality on sugar beet wireworm compared to control, *Beauveria bassiana* and combined EPN/*B.bassiana* treatments (Fig. 6). Between the two EPF species, *M. brunneum* was more effective against wireworm than *B. bassiana* individually and in combination with local EPNs. The application of the local EPN also resulted in 34% mortality which was significantly higher than the non-treated controls.

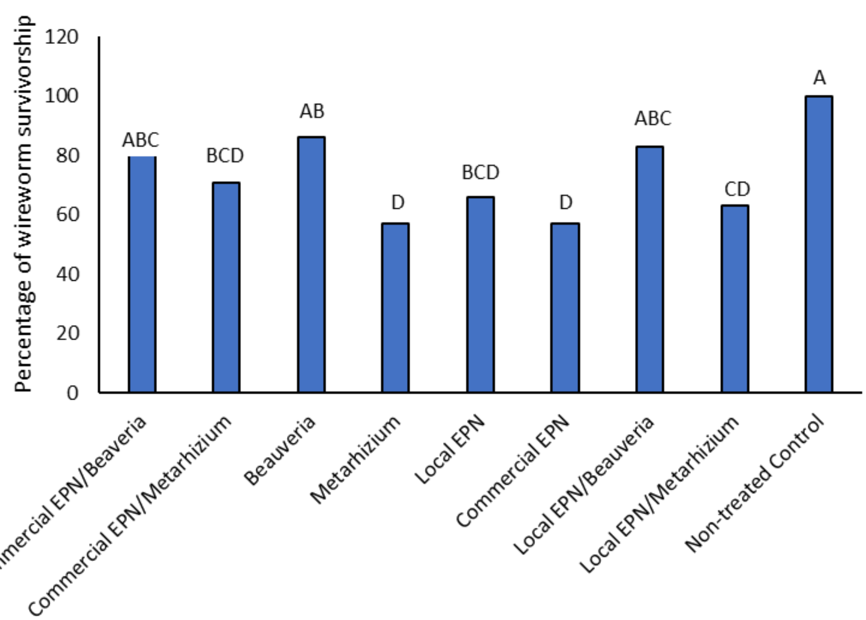

Figure 6. Wireworm survival after exposure to different entomopathogenic treatments (and water treated control) in our greenhouse study.

Percent plant emergence: Plant emergence was significantly affected by treatment (F= 4.75, df = 8, 306, *P* <0 .001). The combination of the EPF, *B. bassiana*, with both local and commercial EPNs resulted in higher plant emergence compared to control and the mixed treatments of local and commercial EPNs with the EPF, *M. brunneum*. Plant emergence was not different between local and commercial EPN treatments. However, between the two EPF species, applying *M. brunneum* resulted in higher plant emergence than *B. bassiana* (Fig. 7).

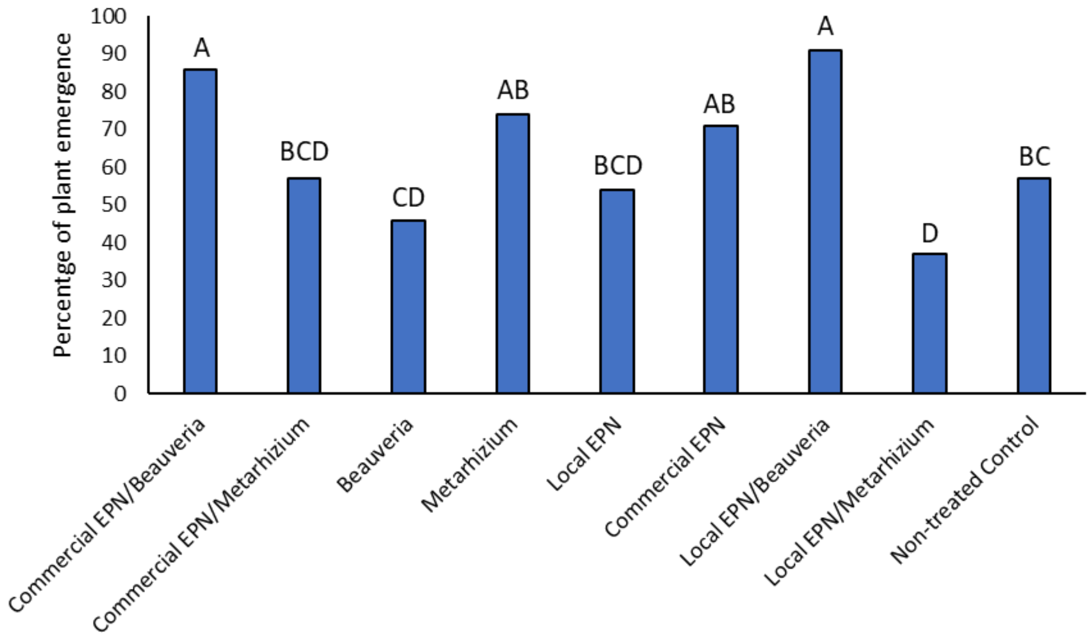

Figure 7. Plant emergence among treatments in our greenhouse study. No wireworm controls (not included) had 100% emergence and were not included in the analyses.

<u>*On-farm trial:*</u>

Number of wireworms: Overall, the number of wireworms were significantly different before and after treatments, with fewer wireworms recovered after the application of biocontrol agents (F= 4.90, df = 1, 173, *P* = 0.028). The number of wireworms also varied in response to treatment (F= 4.28, df = 5, 173, *P* = 0.019), with the least number of wireworms recovered from the commercial EPN treatment (Fig. 8). There were no significant differences in number of wireworms in plots treated with commercial EPN, local EPN, EPF (*B. bassiana*), as well as combination of EPF (*B. bassiana*)/commercial EPN. The mixture of local EPN and EPF was not effective in further reducing wireworm numbers.

Stem count: No significant differences was found in the number of plants before and after treatment application in each plot (F= 2.50, df = 1, 173, *P* = 0.116) and the stem counts were not influenced by treatment (F= 1.21, df = 5, 173, *P* = 0.306; Fig. 9).

Crop yield: Final yield was not affected by treatment (F= 0.172, df = 5, 54, *P*=0.972). Unfortunately, however, we missed crop yield in one of the experimental sites, due to an accidental harvest by a collaborator.

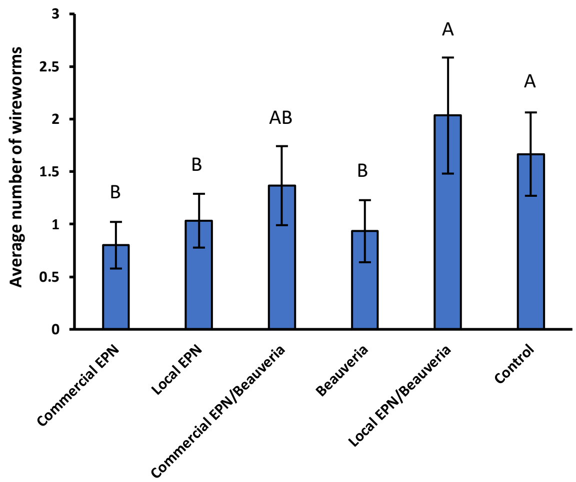

Figure 8. Average number of wireworms collected from each plot after treatment application.

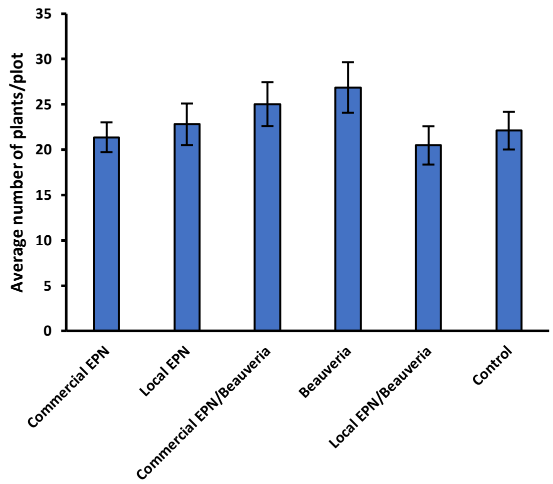

Figure 9. Average number of plants in each plot after applying treatments.

Persistence of applied EPNs:

EPN was detected more frequently in the experimental plots that included EPN in the previous year than the nontreated controls (F = 2.6, df = 4, 51, *P* = 0.043; Fig. 10). Significant variation in EPF detection was also present among plots which were treated with *B. bassiana* in the previous year (F = 3.7, df = 3, 40, *P* = 0.022) (Fig. 11), where EPF was detected at a significantly higher rate than those treated with a mix of EPN/EPF and the non-treated controls.

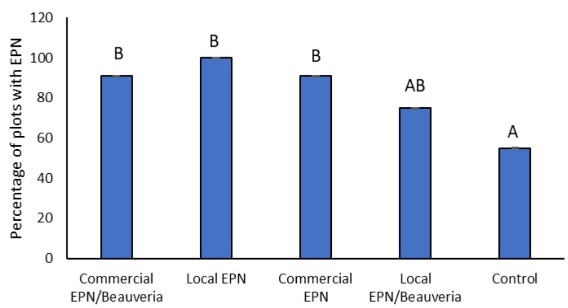

Figure 10. Percentage EPN detection in plots treated with EPN in the previous year, and the nontreated controls.

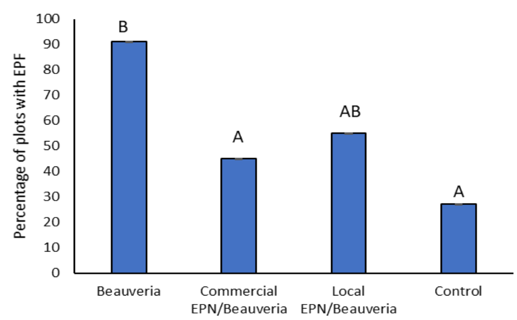

Figure 11. Percentage of EPF detection in plots treated with EPF in the previous year, and the nontreated controls.

Conclusions and Discussion:

Overall, our field and greenhouse results suggested that using a combination of EPF and EPN would not consistently improve the effectiveness of biological control against wireworms.

In the greenhouse the variation in wireworm survival was affected by treatment (*P* = 0.039). The survival rate of wireworms exposed to the commercial EPN/EPF (*S. feltiae/B. bassiana*) treatment was estimated to be 82.8%, and a 71.4% survival was recorded for the wireworms treated with a combination of *M. brunneum* and the commercial EPN. The commercial EPN, *M. brunneum*-only, and the local EPN/*M. brunneum* treatments resulted in

42.9%, 42.9%, and 37.2% mortality, respectively, which were relatively higher than the observed mortality in the rest of the evaluated treatments. Interestingly, however, the highest emergence rates were observed in the commercial EPN/*B. bassiana* (85.7%) and local EPN/*B. bassiana* (91.4%) treatments. Another inconsistency was where the local EPN/*M. brunneum* treatment, with one of the lower wireworm survivorship rates (62.8%), had the least rate of emergence success (37.1%). While one might argue that seed damage could have been caused prior to wireworm mortality, emergence rates of 91.4, 85.7, and 71.4% were reported for local EPN/*B. bassiana*, commercial EPN/*B. bassiana*, and commercial EPN-only treatments. The observed low emergence in local EPN/*M. brunneum* treatment is likely to be a coincidence.

In our on-farm trials, plots treated with *B. bassiana*, local EPN and commercial EPN alone had fewer wireworms than the nontreated controls and plots which were treated with a combination of local EPN/*B. bassiana*. Although, these results supported findings by some of the previous studies (Barberchek and Kaya 1990, Shapiro-Ilan et al. 2004), there are also research demonstrating synergistic effects in using a combination of EPN and EPF (Ansari et al. 2006, Wakil et al. 2017). Important to note is that all EPN and EPF applications in this study were applied at-planting; the time and order of EPN and EPF application, insect developmental stage (Wakil et al. 2017), species/strain (Acevedo et al. 2007), and environmental conditions (Barberchek and Kaya 1990) can influence the success of the approach.

From the wireworm management perspective, our results did not show a significant advantage of using either commercial or locally collected *S. feltiae*, even though the locally collected EPNs are likely adapted to their wireworm prey and the field conditions (Campos-Herrera and Gutiérrez 2014, Morton and Garcia-del-Pino 2017). However, differences in the pathogenicity of local EPNs are also reported among geographic isolates and strains (Kaya and Gaugler 1993, Nikoukar et al. in press).

A higher incidence of waxworm infection was observed in soils collected from plots treated with EPN (commercial and local *S. feltiae*) and EPF/EPN in the previous year than those from the nontreated controls. However, the difference between EPN/EPF and the nontreated controls was not statistically significant. Our results suggest that both commercial and local EPN have the potential to establish within fields. Likewise, the infection rate by EPF was significantly higher in the waxworms exposed to the soil form plots which were treated with *B. bassiana*-only in the previous year. Interestingly, however, no significant difference in waxworm infection rate was detected between nontreated controls and plots, which were treated with the EPF/EPN mix in the previous year.

In conclusion, combining the application of *S. feltiae* (EPN) and *B. bassiana* (EPF) did not improve the efficacy of biological control against sugar beet and western field wireworms when applied at-planting in our field trials. However, the application of local and commercial *S. feltiae* (and also *B. bassiana*) significantly reduced the number of wireworms in our bait traps. We also showed that EPN and EPF have the potential to persist in the environment, especially if applied alone. Further studies are needed to evaluate the effects of timing of combined applications of EPN and EPF (i.e., additive applications) and different species and isolates of EPN (and EPF) on the efficacy of the management approach against wireworms.

Outreach: This proposal included a separate objective (objective 2) for outreach. To disseminate findings of this project to regional organic producers, an online field day (March 2021) was organized and led by Co-PI Mrs. Diane Green (Greentree Naturals Organic Farm), where PI

Rashed and PhD student Nikoukar talked about wireworms, wireworm management, and also presented results of this project. Our goal of organizing an on-site field day was not achieved due to COVID-19 restrictions. A report of our field research is available on the Greentree Naturals Organic Farm website. This report is also made available on Idaho IPM Laboratory website. Research findings were also promoted in Master Gardener classes taught by PI Rashed throughout the state.

Financial accounting: The financial report is enclosed.

Leveraged resources: In addition to OFRF funds, we have secured funding form *Idaho Wheat Commission* and *Idaho Barley Commission*. Nikoukar also secured student support from *USDA-NIFA-Western SARE* to study the effectiveness of mustard rotation in reducing wireworm damage.

References

Acevedo, J. P. M, R. I. Samuels, I. R. Machado, and C. Dolinski. **2007.** Interaction between isolates of the entomopathogenic fungus *Metarhizium anisopliae* and the entomopathogenic nematode *Heterorhabditis bacteriophora* JPM4 during infection of the sugar cane borer Diatraea saccharalis (Lepidoptera: Pyralidae). J. Invertebr. Pathol. 96: 187–192.

Ansari, M. A., M. Evans, and T. M. Butt. **2009**. Identification of pathogenic strains of entomopathogenic nematodes and fungi for wireworm control. Crop Prot. 28: 269–272.

Ansari, M. A., F. A. Shah, L. Tirry, and M. Moens. **2006**. Field trials against *Hoplia philanthus* (Coleoptera: Scarabaeidae) with a combination of an entomopathogenic nematode and the fungus *Metarhizium anisopliae* CLO 53. Biol. Control. 39: 453–459.

Barberchek, M. E., and H. K. Kaya. **1990**. Interactions between *Beauveria bassiana* and the entomogenous nematodes, *Steinernema feltiae* and *Heterorhabditis heliothidis*. J. Invertebr. Pathol. 55: 225–234.

Bedding, R. A., and R. J. Akhurst. **1975**. A simple technique for the detection of insect paristic rhabditid nematodes in soil. Nematologica. 21: 109–110.

Campos-Herrera, R., and C. Gutiérrez. **2014**. *Steinernema feltiae* intraspecific variability: infection dynamics and sex-ratio. J. Nematol. 46: 35–43.

Grewal, P. S., R.-U. Ehlers, and D. I. Shapiro-Ilan. **2005**. Nematodes as Biocontrol Agents. CABI.

Kaya, H. K., and R. Gaugler. **1993**. Entomopathogenic Nematodes. Annu. Rev. Entomol. 38: 181–206.

Lewis, E. E., J. Campbell, C. Griffin, H. Kaya, and A. Peters. **2006**. Behavioral ecology of entomopathogenic nematodes. Biol. Control. 38: 66–79.

Milosavljević, I., A. D. Esser, N. A. Bosque-Pérez, and D. W. Crowder. **2016**. The identity of belowground herbivores, not herbivore diversity, mediates impacts on plant productivity. Sci. Rep. 6: 39629.

Morton, A., and F. Garcia-del-Pino. **2017**. Laboratory and field evaluation of entomopathogenic nematodes for control of *Agriotes obscurus* (L.) (Coleoptera: Elateridae). J. Appl. Entomol. 141: 241–246.

Nikoukar, A., P. Ensafi, E. E. Lewis, D. W. Crowder, and A. Rashed. **In press**. Efficacy of naturally occurring and commercial entomopathogenic nematodes against sugar beet wireworm (Coleoptera: Elateridae). J. Econ. Entomol.
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. Agric. For. Entomol. 3: 85–98.
Rashed, A., F. Etzler, C. W. Rogers, and J. M. Marshall. **2015**. Wireworms in Idaho cereals: Monitoring and identification. Univ. Ida. Ext. Bull. 898.
Razinger, J., M. Lutz, H. Schroers, G. Urek, and J. Grunder. **2013**. Laboratory testing of insect associated fungi for the control of wireworms (*Agriotes* sp. L.). IOBC-WPRS Bull. 90: 103–107.
Reddy, G. V., K. Tangtrakulwanich, S. Wu, J. H. Miller, V. L. Ophus, J. Prewett, and S. T. Jaronski. **2014**. Evaluation of the effectiveness of entomopathogens for the management of wireworms (Coleoptera: Elateridae) on spring wheat. J. Invertebr. Pathol. 120: 43–49.
Shapiro-Ilan, D. I., M. Jackson, C. C. Reilly, and M. W. Hotchkiss. **2004**. Effects of combining an entomopathogenic fungi or bacterium with entomopathogenic nematodes on mortality of *Curculio caryae* (Coleoptera: Curculionidae). Biol. Control. 30: 119–126.
Shirck, F. H. 1945. Crop rotations and cultural practices as related to wireworm control in Idaho. J. Econ. Entomol. 38: 627–633.
Sufyan, M., A. Abbasi, M. D. Gogi, M. Arshad, A. Nawaz, and D. Neuhoff. **2017**. Efficacy of *Beauveria bassiana* for the management of economically important wireworm species (Coleoptera: Elateridae) in organic farming. Gesunde Pflanz. 69: 197–202.
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. Annu. Rev. Entomol. 60: 313–334.
Vernon, R. S., W. Van Herk, J. Tolman, H. Ortiz Saavedra, M. Clodius, and B. Gage. **2008**. Transitional sublethal and lethal effects of insecticides after dermal exposures to five economic species of wireworms (Coleoptera: Elateridae). J. Econ. Entomol. 101: 365–374.
Wakil, W., M. Yasin, and D. Shapiro-Ilan. **2017**. Effects of single and combined applications of entomopathogenic fungi and nematodes against *Rhynchophorus ferrugineus* (Olivier). Sci. Rep. 7: 1–11.
White, G. F. 1927. A method for obtaining infective nematode larvae from cultures. Sci. Wash. 66.