The effect of Mustard Seed Meal, Enterra Natural Fertilizer, Spent Compost Mushroom Manure, or *Metarhizium brunneum* on wireworm damage in Hakurei turnip

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Abstract

Wireworms are soil-dwelling pests that feed on vegetable, fruit, and cereal crops. They are the larvae of click beetles (Coleoptera: Elateridae. Management against wireworms is difficult because the larvae can stay in the soil for 2-6 years. *Metarhizium brunneum* has previously shown positive results as biocontrol against wireworms. Use of organic soil amendments, such as Enterra Natural fertilizer (ENF) and Brassica Defatted mustard Seed Meal (DSM), have also shown to help reduce wireworm damage in crops. In this study, *M. brunneum*, ENF, DSM, and Spent Mushroom Compost (SMC) effects on wireworms were tested. This experiment had four treatments plus a control, with four replicates per planting date (early and late planting dates). No significant effect was found in any treatments or planting dates.

1. Introduction

Wireworms are the larvae of click beetles (Coleoptera: Elateridae) and are soil dwelling pests that damage and cause destruction to fruit, cereal, and vegetable crops worldwide (Barsics et al, 2013). The wireworm life cycle can range from 2-6 years, depending on the species and location in which they live, and have three stages they go through: adult, larva, and pupal stages (Andrews et al, 2008). After adults mate, females can lay eggs around the roots of grasses and grains, laying an average of 80 eggs (Andrews et al, 2008).

Adult click beetles are slender, hard-shelled beetles. They range from tan to dark brown, can be 8–20 mm long, and get their name from the clicking sound they make (Andrews et al, 2008). After hatching, wireworm larvae are usually white, though they can darken with age. Depending on species, wireworm larvae range from about 2 mm long after hatching to 4 cm long (Andrews et al, 2008).

Larvae of wireworms feed underground on seeds and roots. They are attracted to germinating seeds by the CO_2 given off during germination (Furlan et al, 2010). Larval wireworms are among the most destructive of soil insect pests. Wireworms that cause the most economic impact in the south coastal region of British Columbia are of the genera

Aeolus, Agriotes, Ctenicera, Dalopius, Limonius, and Melanotus (Vernon and van Herk, 2013). Wireworms of the *Aeolus* spp., *Melanotus* spp. and *Limonius* spp have been known to target turnip plants (Penn State University).

Figure 1. Wireworm collected from Richmond, BC

There are several species in British Columbia that cause damage. In recent years, European species in the genus *Agriotes* have become more prominent pests in B.C (Vernon & van Herk, 2017). Control or management techniques of wireworms are crop rotations, frequent tilling, and use of chemical pesticides (Andrews et al, 2008).

The use of chemical pesticide control for management of wireworms has been popular, however, many older, broad-spectrum products that have been used for control of wireworm such as carbamates and organophosphates are no longer available or are being phased out. Other products, such as clothianidin, which has been approved by the Pest Management Regulatory Agency (Government of Canada, 2021), would not be available to organic farmers.

Hakurei turnips, *Brassica rapa* var. *rapa* are cold hardy vegetables which can be seeded in the spring for a summer harvest or sown in the summer for harvesting in the fall. Hakurei turnips are a fast-growing brassica and have both edible greens and roots (Liu et al, 2019). Turnips are usually direct seeded and can be sown as soon as the soil is workable. Common pests of turnips are diseases such as black rot, club foot, and downy mildew or insects that include cabbage aphids, root maggots, and wireworms (Andrews et al, 2008).

Numerous studies have been performed on the effect of wireworm damage on potatoes and cereal crops. However, few have studied the effect wireworms have on turnips and ways to control that damage. Hakurei turnips were chosen for this experiment due to damage done to them by wireworms as well as their short maturation cycle, allowing for an early planting and late planting.

Metarhizium brunneum is an entomopathogenic fungus that occurs naturally in the soil and has already been tested with varying success against wireworm damage and has been found to contribute to the natural control of a wide range of insects (Eckard et al, 2014). *Metarhizium brunneum* has also shown a reduction in wireworm damage in potatoes using an attract-and-kill strategy (Kabaluk et al, 2015). The timing of the application of *Metarhizium brunneum* may influence effectiveness on wireworms (Eckard et al, 2014). Some studies have shown limitations to using *Metarhizium brunneum*, due to timing, temperature, and soil moisture (Ericsson et al., 2007).

Adding amendments to the soil has shown to have added protection to crops against wireworm damage (Temple et al, 2013). Defatted seed meal has shown to mitigate damage by wireworms (Furlan et al, 2010). Defatted seed meals derived from Brassicaceae plant tissues with high glucosinolate content and have been used as an alternative to control soil-borne plant pathogens and pests (Mataeo et al, 2018). Defatted seed meal from brassicas such as mustard, rapeseed, and canola may protect plants from wireworms by releasing glucosinolates, which break down into gasses that are toxic to insects (Furlan et al, 2010).

Enterra Natural Fertilizer is food waste digested by black soldier fly larvae and converted into organic fertilizer. The frass of black soldier fly larvae has been shown to protect plants from wireworms. Enterra Natural Fertilizer contains plant nutrients such as nitrogen, phosphorus, and potassium, and is a good source of beneficial microbes and minerals to protect plants. (Enterra, 2014).

Mushroom compost is produced from chopped straw, chicken manure, gypsum and water (Uzun, 2004). Spent mushroom compost is the by-product of mushroom production industries (Fidanzaet al. 2010). Fresh spent mushroom compost contains a lot of salts (Uzun, 2004), with an electrical conductivity around 13 dS/m (Fidanza et al, 2010). Mushroom manure does not contain any pests or weed seeds, because of the high temperatures associated with the composting and pasteurization processes, as well as contains very low levels of pesticides and heavy metals (Uzun, 2004).

The purpose of this experiment was to assess, analyze, and compare defatted mustard seed meal, Enterra Natural Fertilizer, spent mushroom manure compost, and *Metarhizium brunneum* applications to soil seeded to Hakurei turnips, to measure the effect of wireworm damage.

Methods and Materials

2.1 Growing Site

This experiment was conducted at the Kwantlen Polytechnic University Orchard on Gilbert Road in south Richmond. The experiment took place from June to October of 2021.This experiment was done in partnership with Kwantlen Polytechnic University, and Agriculture and Agri-Food Canada.

2.2 Experimental Design

The experimental design was a randomized complete block with four replicates. The five soil amendment treatments were: 1) Defatted Seed Meal (DSM); 2) spent mushroom manure compost (SMC); 3) *Metarhizium brunneum* (MB); 4) Enterra Natural Fertilizer (ENF), consisting of black soldier fly frass; and 5) an untreated control. Two planting dates were tested: An early planting (July 9th) and a late planting (August 20th). The combination of four replicates, two planting dates, and five soil amendments resulted in 40 plots, each 9.0 m^2 (2.5 x 3.6 m). Each replicate block contained 10 plots and covered 90 m^2 (5.0 x 18 m). Ten rows of turnips were planted in each plot. There was a one-meter buffer between the blocks.

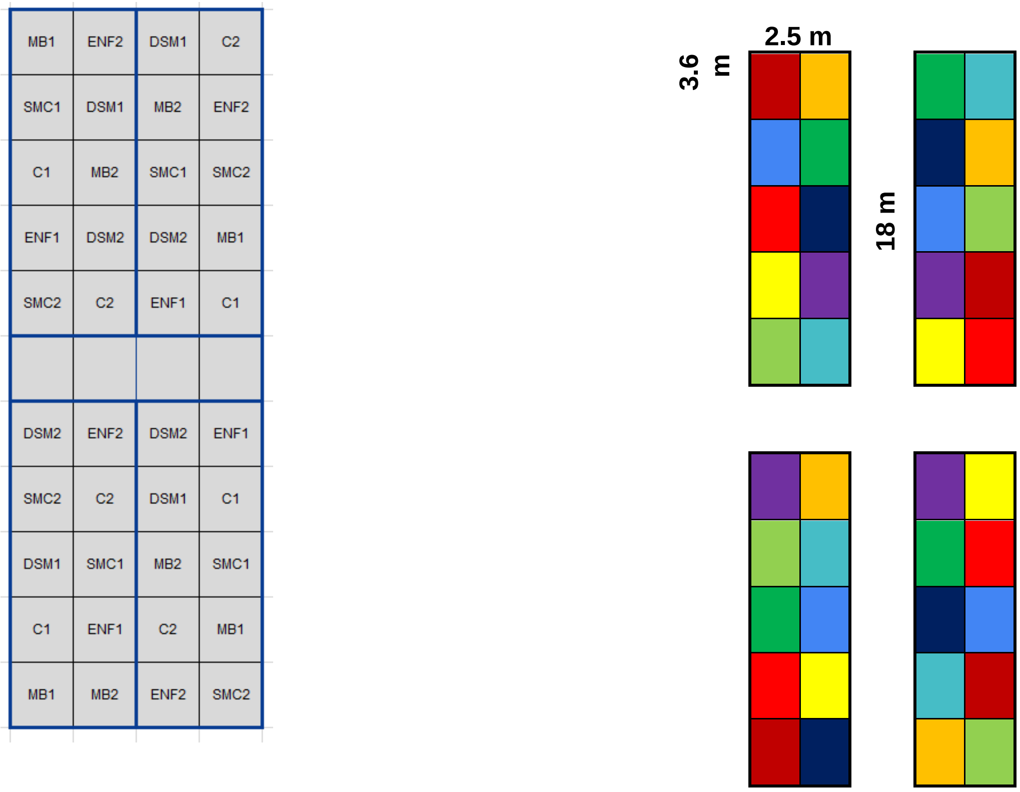

Figure 2. Randomized complete block design, showing four replicates of the five treatments as well as early and late plantings and field setup.

2.3 Materials

The field was tilled May 8 and again June 20 to incorporate the cover crop. Defatted seed meal and *Metarhizium brunneum* were applied two weeks before planting. The spent mushroom manure compost and Entera Natural Fertilizer was applied at seeding.

The Enterra Natural fertilizer was applied at 100 g/m^2, which equals .9 kg/plot. It was supplied by Enterra and was spread throughout the plot and incorporated by raking into the soil.

Spent mushroom compost manure was applied at a rate of 6 kg/m^2, which was equal to 54 kg/plot. Spent mushroom compost, made of wheat straw, chicken manure, mined gypsum, peat moss, lime, and feather meal, was supplied by Highline Mushrooms (Langley, BC). The mushroom compost was added as a treatment due to its higher saline content than other fertilizers or composts. It was incorporated into the soil by raking it in.

The defatted seed meal was applied at 1.3 kg/m^2, coming to 11.5kg per plot. Hakurei turnips seeds were purchased from West Coast Seeds and planted in 10 rows per plot.

Figure 3. *Metarhizium brunneum* before combined with rolled oats.

Metarhizium brunneum was applied at 46.6 g/plot mixed with 74 g of rolled oats for the carbon dioxide source. *Metarhizium brunneum* sourced from wireworm cadavers and was supplied by Agri-Food Canada (Aggasiz, BC). MB and oats were divided between four 10 cm deep trenches running the length of each plot, 50 cm spacing which were re-filled with soil after treatment application. Irrigation was provided by sprinklers as needed.

2.4 Methods

The first planting took place on July 9. Twenty plots were seeded with Hakurei turnips, 10 rows per plot. Plots were weeded three times a week and irrigated every other day for two hours from 3-5 am. Due to low germination, the early planting was re-seeded August 2. On August 6, *M. brunneum* and the DSM were applied and incorporated for the late planting. The late planting was seeded on August 20. On September 23, a single potato tuber without wireworm feeding holes was dug into the centre of each plot, 25 cm deep to help measure wireworm pressure.

Figure 4. Application of *Metarhizium brunneum* in block 4.

Turnips were harvested from both plantings on October 11. The number of turnips and the proportion of turnips damaged by wireworms was recorded for each plot. Potato traps were removed October 23. The number of wireworm feeding holes in each potato was recorded.

Statistical analysis was conducted using Jamovi R software. Analysis of Variance (ANOVA) was used to analyze the data. The Shapiro-Wilk test was used to check for normality. ANOVA was used to test for treatment effects. Analyses were conducted using P-value of 0.05.

Results

No significant effects of soil amendment or planting date were detected on wireworm damage. Table 1 shows the range of turnips grown per plot. SMC and DSM grew a mean of over 200 turnips per plot. MB had the least number of turnips grown per plot with a mean of 71. Five plots had zero results due to lack of germination and turnip growth.

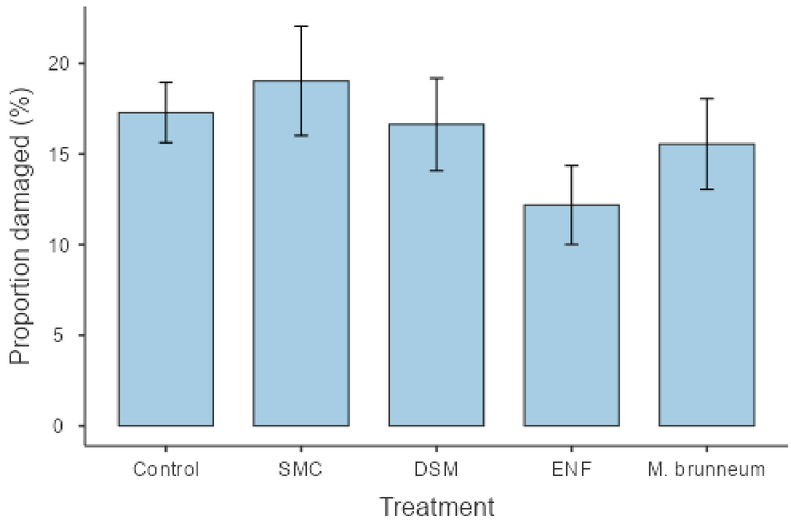

Figure 5. Proportion of turnips damaged by treatment (± S.E.).

Figure 5 shows the proportion of turnips damaged per treatment. SMC had the highest proportion of turnips damaged and ENF had the lowest proportion damaged, but these treatments did not differ significantly.

Potato traps were placed in September due to the lack of turnips in some plots. The potato traps also showed no significant effect for treatments.

Table 3. Monthly mean temperature and total precipitation in Richmond, BC from April-October, 2021 (Environment Canada).

Month	Total Precipitation (mm)	Mean Temperature (C°)
April	38.0	9.4
May	30.2	12.5
June	37.2	17.8
July	0	19.4
August	0	19.4
September	155.0	15.4
October	148.4	9.7

Discussion

Wireworms have proven to be well established in the field that this study took place, with multiple studies having taken place since 2015, with varying results of success. No treatment proved to be successful in this study. In block 1 the treatment of DSM in the early planting, 40% of the turnips in the plot had wireworm damage as well as the potato trap having holes of damage in it. However, the southeast corner of that plot is where most of the damaged turnips were harvested from. *M. brunneum* of the late planting in both block 1 and 2, also had the majority of the damaged turnips come from the corner of the plots. In block 4, SMC from the early planting had almost 25% of turnips with the potato trap showing 8 holes. The majority of the turnips with wireworm damage came from the southwest portion of the plot, alongside the control of the late planting as well as *M. brunneum* of the early planting.

The northern most plots from block 1 and 2, showed the least amount of turnip growth. Two of the four plots had zero turnip germinations (MB early planting and DSM early planting). There was a distinct difference of moisture in these four plots compared to the rest of the field, even with constant irrigation.

Turnip germination was poor throughout this study, even after the early planting was reseeded. The first treatments of *M. brunneum* and DSM were applied three days before the southern coast of British Columbia suffered a heat dome, with temperatures reaching almost 40°C in Richmond. The first early planting was seeded shortly after the heat dome. As shown in table 3, temperatures in July and August were high, with a mean temperature of 19.4°C, almost two degrees higher than average (Environment Canada, 2021). Germination from the August 2 and August 20 plantings were also sporadic in certain plots. High nightshade, thistle, and purslane weed pressure, and flea beetle damage (Figure 6) contributed to poor turnip growth in the eastern plots

Figure 6. Flea beetle damage to turnip plants.

Conclusion

No significant treatment effects were detected in this study for any treatments. The lack of significant treatment effects does not necessarily imply that *M. brunneum* was inactive. *M. brunneum* has been applied to different areas on this site in different experiments since 2015 and could have persistent effects. Future studies may include testing the site for lasting *M. brunneum* residue in the soil or studying if previous applications have a residual effect to determine how long it could persist. Other studies could also include other treatments being used on different crops and explore the effect of timing of the applications.

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Appendix – Raw Data

Table 2 – Raw turnip data from early and late plantings.

Block	Treatment	Planting	No. of Turnips	Wireworm Damage
1	C	1	101	21
1	MB	2	49	5
1	C	2	119	24
1	DSM	1	120	48
1	ENF	1	121	26
1	ENF	2	171	7
1	DSM	2	176	28
1	SMC	2	208	30
1	MB	1	-	-
1	SMC	1	-	-
2	MB	1	8	2
2	ENF	1	15	2
2	ENF	2	43	4
2	C	2	46	4
2	SMC	1	47	14
2	MB	2	119	17
2	DSM	2	174	37
2	SMC	2	177	28
2	DSM	1	-	-
2	C	1	-	-
3	MB	1	27	2
3	MB	2	76	17
3	ENF	2	116	13
3	C	2	133	23
3	DSM	2	244	39
3	SMC	2	315	18
3	C	1	334	71
3	ENF	1	365	32
3	SMC	1	375	70
3	DSM	1	389	86
4	DSM	1	51	4
4	C	2	60	9
4	MB	1	64	4
4	DSM	2	76	14
4	C	1	107	19
4	ENF	2	123	21
4	SMC	1	139	34
4	MB	2	152	31
4	SMC	2	173	42
4	ENF	1	-	-

Block	Treatment	Planting	Wireworm Holes
1	MB	1	0
1	SMC	1	8
1	C	1	6
1	ENF	1	6
1	M	2	0
1	ENF	2	1
1	DSM	1	6
1	MB	2	8
1	DSM	2	4
1	C	2	6
2	DSM	1	0
2	C	2	0
2	MB	2	4
2	SMC	1	0
2	DSM	2	4
2	ENF	1	3
2	ENF	2	4
2	SMC	2	6
2	MB	1	7
2	C	1	7
3	DSM	2	1
3	SMC	2	0
3	DSM	1	5
3	C	1	4
3	MB	1	1
3	ENF	2	3
3	C	2	1
3	SMC	1	8
3	ENF	1	1
3	MB	2	2
4	DSM	2	1
4	DSM	1	1
4	MB	2	0
4	C	2	2
4	ENF	2	1
4	ENF	1	1
4	C	1	4
4	SMC	1	8
4	MB	1	10
4	SMC	2	3