

The Pennsylvania State University

The Graduate School

**EVALUATING AGROECOSYSTEM QUALITY THROUGH ARTHROPOD INTERACTIONS IN
PENNSYLVANIAN CASH CROP PRODUCTION SYSTEMS**

A Thesis in Entomology

by

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ABSTRACT

During the last 60 years in the United States, agricultural production and demand has increased, while total acres farmed has remained relatively constant. The increased demand has led to environmentally damaging agricultural and pest management strategies. This thesis examines the effects of multiple crop production practices on pests and natural enemies. To do this, I have split my thesis into three chapters, each of which examines an aspect of agroecosystem resiliency and quality in modern Pennsylvanian crop production systems. Here, resiliency refers to an agroecosystem's ability to withstand stresses (e.g., pest populations) without the use of preventative-pest management strategies while practicing IPM.

In Chapter 1 of my thesis, I evaluate the effects of planting into a living cereal-rye (*Secale cereal* L.) cover crop on conservation biological control of multiple pests within corn (*Zea mays*) and soybean (*Glycine max*). In the mid-Atlantic, United States, Chesapeake Bay watershed region, the wide use of preventative-pest-management strategies (E.g., seed treatments and tillage) can have serious negative effects on the environment. To mitigate environmental risks, minimal tillage, no-till, and cover cropping practices have been evaluated by extension personnel and adopted by row-crop growers in the region and the United States. While these practices can be effective in reducing run off, reducing pest pressure, increasing diversity, and bolstering resiliency of an agroecosystem, little work has been done to evaluate the role planting into a living cover crop (planting green; PG) has on pest-plant-predator interactions. My goal in this chapter was to determine if planting green can lower overall pest pressure, reduce slug populations, increase predation by natural enemies, and increase yield. I recorded minimal changes in overall pest abundance or pressure, where the multiple-pest damage was the only instance to have more damage in the planting-green treatment. Slug populations only increased with cover-crop biomass in corn plots during the spring of 2021. For predation, higher cover-crop biomass treatments had higher levels of attacked prey, with the highest predation levels occurring later in the season. Lastly, I recorded no differences in yield among treatments. This study demonstrates that planting green can be an effective – conservation based– approach to pest management for row-crop growers.

In Chapter 2, I evaluate the role of generalist-spider predators and their contributions to pest control in soybeans. Integrated Pest Management (IPM) within soybeans relies on natural enemies, typically generalist predators, to keep invertebrate-pest populations below economic thresholds. Due to the complex nature of agroecosystems, seldom can one natural enemy contribute to the bulk of pest control. Spiders are ubiquitous insect-feeding predators present in almost all terrestrial ecosystems. Depending on the taxa, spiders deploy several means of prey capture which allows them to exploit a variety of niches within habitats. As a result, spiders have potential to contribute positively to pest control within modern soybean systems managed with Integrated Pest Management (IPM). In this chapter, my goal is to determine the most abundant natural enemies within modern Pennsylvanian soybean systems. Furthermore, I attempt to quantify the role that spiders play in invertebrate-pest control through the manipulation of pest and predator populations in the field. I determined that spiders are the most abundant predators of soybeans in Pennsylvania and that the family Thomisidae had the greatest abundance of all spiders. While pest populations did not decrease with an increase in spider populations, it appears that spider populations respond proportionally to prey abundance. Time of season influenced spider populations and most individuals sampled came from mid-late season sampling periods (July-August). This study highlights spiders as the most abundant predators within modern soybean systems and that current row-cropping systems do not support their populations during the early season.

In Chapter 3 of my thesis, I evaluate the biological quality of soils under different crop management practices using soil invertebrates and the Biological Soil Quality index-arthropod (QBS-ar). We rely on soils for many things including food production and environmental improvement. Soils of course support crop production, but also extend to the improvement and support of environmental quality. One way environmental quality can be improved by soils is through support of soil-dwelling fauna. To support biological diversity of soil-dwelling fauna soils must be of high quality. Soil quality refers to measurable attributes that relate to the capacity of soil to function without a negative interaction with the environment. Current tests for soil quality use multiple chemical, physical, and biological attributes (e.g.,

bulk density, nitrogen availability, and microbial respiration). These attributes often reveal important information to growers on how to maintain and increase crop quality and yield. What these tests do not cover are direct links to biological quality. Specifically, the biological assessment of soils subject to different crop management practices. The QBS-ar index is a reliable method for evaluating the biological quality of an ecosystem. The QBS-ar index scores biological quality based on the morphological adaptations of soil-dwelling (edaphic) invertebrates within. My goal was to determine which suite of row-crop practices produced the highest QBS-ar values and to determine which factors most influenced soil-invertebrate abundance and QBS-ar values. Along with this, I set out to test if the termination timing of a cover-crop influenced QBS-ar values and overall soil-invertebrate abundance. I found no change in QBS-ar value based on cover-crop biomass or system type (organic vs. conventional). I did, however, record differences in QBS-ar values based on tillage practices. Fields subject to no-till had higher QBS-ar values than fields that underwent any form of tillage pre-cash-crop planting. Additionally, twenty-six of the total twenty-eight recorded QBS-ar scores fell below the high-quality threshold. My results display a trend that the sampled row-crop-management practices seldom aid in bolstering biological quality of soils.

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To my friends and companions, it has been a pleasure. From dumpster diving to fungus walks to cheese parties, the support and comradery was unequivocal. At the start of my degree, I did not think I had space for any more friends, but it turns out I was wrong. I am happy to say I have made lifelong friends during this two-year period, and I will not forget you soon.

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Andy Deans – who gave me an A- in his seminar – taught me more about arthropod and insect identification and life history than I could have imagined. Your passion for taxonomy and love of all

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To all the arthropods that lost their lives to my research, it was an honor and I hope my work has meaning. It is difficult to kill so many wonderful invertebrates, but regrettably, such is the nature of a degree in entomology. The total tally of *killed and counted* arthropods was 13240 and an additional 600 (13840) waxworms used as bait in sentinel prey assessments. These values do not include bycatch and lost samples.

A special thanks to the Ski Patrol team at Tussey Mountain. While I am unsure if any of you will read this, it was an honor to work alongside you all.

Lastly, to Rothrock State Forest. How lucky we are to be but a stone's throw from one of the most technically challenging and beautiful state forests in the state of Pennsylvania.

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Chapter 1

Conservation Biological Control: Managing Pests, Increasing Predators, and Improving Agroecosystem Quality through Cover-Crop Termination Timing

1. INTRODUCTION

In U.S. since the 1960s, cereal production acres have remained constant while cereal production yield has increased from 15.1 million t to 31.7 million t in 2014 (Kopittke et al., 2019). This increase in production has emerged from an intensification that involves greater reliance on fertilizers and pesticides (Barzman et al., 2015). Inherent to many intensified agricultural systems is tillage, which is one of the oldest and still widely used means of pest control (Lal et al., 2007). Newer approaches to pest control, like preventative strategies, such as pesticide-seed treatments or adding insecticides or fungicides to necessary herbicide applications, have been highly adopted in modern, annual crop production systems that grow corn and soybean (Douglas & Tooker, 2015; Johnston-Fennell et al., 2021). The increase in synthetic chemical use and more general intensification of agricultural systems comes with growing concerns of environmental degradation (Goulson, 2013; Hladik et al., 2014; USDA, 2011).

Widespread use of preventative-management strategies (Sparks, 2013) in conjunction with soil-disturbance intensification, like tillage, can have strong negative effects on the agroecosystem and the environment (Goulson 2013), especially in the Chesapeake Bay watershed located in the mid-Atlantic United States (USDA, 2011). The Chesapeake Bay watershed comprises many land use types, which include hayland (5.9%), pasture and grazing land (12.6%), urban land (8.6%), forest and other (62.5%), and cultivated cropland (10.5%) (USDA, 2011). This 10.5% of cultivated land is responsible for 21.5% of total sediment run off, 30.8% of nitrogen leaching, and 28.3% of total phosphorus delivery; all of which run into rivers and streams (USDA, 2011).

Tillage can help manage invertebrate pests that overwinter in soil or lay eggs on crop residues while also disrupting life cycles of weeds (Lal et al., 2007; Rowen et al., 2020; Wallace et al., 2018). While tillage can provide benefits, this practice also increases soil erosion potential, decreases soil structure and stability, decreases water retention potential, and decrease nutrient retention (USDA, 2015). Due to environmental concerns of soil erosion and nutrient transport within the Chesapeake Bay, there has been a concerted effort to improve the sustainability of agroecosystems in the region (USDA, 2015). Conservation tillage, and no-till farming in particular, in agronomic systems are increasing in popularity in the United States, but the Mid-Atlantic region started adopting these conservation practices decades ago to help protect the Chesapeake Bay. Conservation tillage minimizes soil disturbance and thus builds soil structure (USDA, 2015). Improving soil structure through no-till can reduce erosion, reduce tillage-

induced equipment emissions, maintain or increase soil organic matter, and increase efficiency of water use and storage (USDA, 2015). Reducing or eliminating tillage can also aid in the control of invertebrate pests by protecting natural-enemy populations (Bohnenblust et al., 2014; Douglas & Tooker, 2012; Finke & Denno, 2002; Hammond & Stinner, 1987; Landis et al., 2000; Rowen et al., 2020; Thorbek & Bilde, 2004). Increasing natural-enemy populations is of particular interest for managing slug (Phylum: Mollusca) populations due to their potential for crop damage in no-till row-crop systems (Godan, 1983; South, 1992).

Another means of improving agroecosystem functions is using cover crops (Gyssels et al., 2005). Cover crops can reduce erosion from wind and water, increase soil organic matter content, capture and recycle residual nutrients in the soil profile, manage soil moisture, and minimize or reduce soil compaction (USDA, 2015). Furthermore, adding cover crops can improve agroecosystem diversity by increasing habitat for natural enemies (Landis et al., 2000). However, a potential downside to adding cover crops is creating cooler and wetter environments (Blevins et al., 1971; Imholte & Carter, 1987). With these conditions, there is concern among growers that increasing overall residue and biomass on the soil surface will lead to an increase in invertebrate pests (Douglas & Tooker, 2012; Willson & Eisle, 1992) and diseases. While cover crops may increase pests, there is likely an increase in both invertebrate pest and predator abundance and diversity (Finke & Denno, 2002; Schmidt et al., 2008). Additionally, the combination of cover crops and conservation practices, like no-till, can further increase agroecosystem resiliency and environmental quality (Duiker, 2023) by increasing habitat for natural enemies, increasing organic matter in the field (USDA, 2015), and help mitigate and adapt agroecosystems to changes to the climate, such as drought (Berrand et al., 2022).

Traditionally, cover crops have been terminated 7-28 days prior to cash-crop planting. This range in termination timing can promote soil warming, allow for cover crop drying and decomposition, and can reduce pest pressure carry over from the living cover crop. However, terminating cover crops early in the season can reduce the potential for agroecosystem benefits, whereas delaying termination to cash crop planting can increase potential benefits associated with cover crops (USDA, 2015). Delaying termination of cover crops to shortly after cash-crop planting is a practice known as planting green (PG). This increasingly popular management practice allows cover crops to grow longer and increase overall biomass is of interest to growers looking to increase agroecosystem quality. For example, increasing cover-crop biomass can increase water retention, reduce the potential for soil erosion than no-till alone (Gyssels et al., 2005; Kenneth G. Renard et al., 1991), improve soil quality metrics (Gyssels et al., 2005), reduce nutrient leaching into watersheds (Alonso-Ayuso et al., 2014; Farsad et al., 2011), and decrease overall weed and invertebrate pest pressures (Finke & Denno, 2002; Mischler et al., 2010).

Planting green, however, comes with several concerns: 1) a reduction in seed contact with the soil (Reed et al., 2019), 2) an increase in abundance of invertebrate pests (Douglas & Tooker, 2012), 3) and a reduction of overall yield (Reed et al., 2019) due to the combination of delayed germination from poor soil seed contact, cooler and drier soils at planting, and an increase in pest abundance.

With growing popularity in mid-Atlantic states of delaying cover-crop termination and planting green, there is a need to understand effects of delayed cover-crop termination on pest control in no-till agronomic systems. In this chapter, I quantify the effects of differential termination timing of a cereal-rye (*Secale cereal L.*) cover crop on the pest and predator communities in a no-till corn and soybean system within Pennsylvania. I hypothesized that by increasing overall cover-crop biomass (ecosystem complexity) and using IPM (i.e., avoiding preventative insecticides), I would see *i*) lower invertebrate pest damage, *ii*) higher natural-enemy populations and more prey killed, *iii*) and higher crop yield in the late-terminated, high-biomass treatments.

2. MATERIALS AND METHODS

2.1. Study location and experimental design

I conducted a three-year field experiment (2021-2023) at Penn State's Russell E. Larson Agricultural Research Center (Pennsylvania Furnace, Pennsylvania, U.S.A.; 40.720064, -77.934317). In a long-term no-till field (>10 yr.), I established a single-factor experiment, arranged in a randomized complete block design with four cover crop treatment levels and five replicates. Plot size was 12 x 15 m. Cover crop treatments varied by termination timing of the cereal-rye cover crop and included 1) a no cover-crop control (Referred to here as No CC) and cover crop terminations of: 2) 14-28 days pre-planting (DPP; Referred to here as 'Early terminated'), 3) 3-7 DPP (Referred to here as 'Late terminated'), and 4) 1-3 days after planting (DAP; Referred to here as 'Planting green') of the cash crop. Cover crops were terminated using glyphosate (1.27 kg ae ha⁻¹) and ammonium sulfate (2.5% v/v) using water as a carrier at 140 L ha⁻¹. Based on abiotic conditions, planting dates of cover and cash crops, and termination dates varied annually (Table 1-1; For soil type information, see Table 1-1a). I established a 3 m border of cover crop around the experiment and 9 m alleys between blocks. The border and the alleys follow the planting-green schedule for termination and cash-crop planting (Fig. 1-1).

Table 1-1. Cover-crop and cash-crop planting and termination dates and seeding rates.

Crop year	Corn field cover crop planting date	Soybean field cover crop planting date	Cereal rye seeding rate (kg/ha)	Treatment termination			Planting green	Corn planting date	Corn seeding rate (seeds/ha)	Soybean planting date	Soybean seeding rate (seeds/ha)
				No Cover Crop	Early	Late					
2021	8 Sept. 2020	-	67.5	-	27 April	12 May	24 May	21 May	81,250	-	-
2022	10 Nov. 2021	21 Oct. 2021	67.5	-	29 April	17 May	1 June	31 May	81,250	31 May	450,000
2023	26 Oct. 2022	10 Nov. 2022	67.5	-	5 May	12 May	23 May	23 May	81,250	23 May	450,000

Table 1-1a. Soil type information for the three field sites and the associated crops and year. All soil data retrieved from the Web Soil Survey (Soil Survey Staff, n.d.).

Field number	Crop	Year	Crop	Year	Soil Type	Slope	%	Slope	%
1	Corn	2021	Soybean	2022	Hagerstown silt loam	0 to 3	66.7	3 to 8	33.3
2	Corn	2022	Soybean	2023	Murrill channery silt loam	0 to 3	100	-	-
3	Corn	2023	-	-	Murrill channery silt loam	0 to 3	100	-	-

To characterize influences of termination date on cover-crop productivity, I harvested aboveground biomass from each plot one to two days prior to termination. From four randomly positioned quarter meter quadrats per plot, I cut cover crop biomass roughly 2.54 cm above the soil surface. After collection, samples were dried in an oven (70°C) for 5-7 days, and then weighed.

To evaluate legacy effects associated with a corn-soybean rotation, I followed corn in 2021 with soybean in 2022 as well as corn in 2022 followed by soybean in 2023. In both instances, I applied the

same treatment to plots as were used in the previous year. I did not collect pitfall samples in corn in 2021. The legacy pairs 2021 corn – 2022 soybean and 2022 corn – 2023 soybean both share the following data types: cover crop biomass, yield, slug counts, and sentinel prey. Additionally, 2022 corn – 2023 soybean share pitfall sampling.

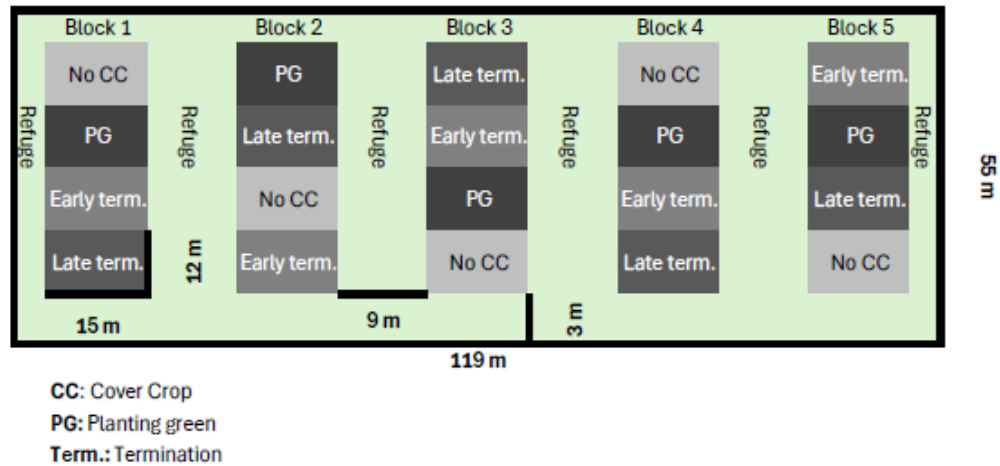


Figure 1-1. Schematic diagram of the design showing treatments, blocks, and buffer areas. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

2.2. Damage from pests and pest abundance

To understand the influence of insect and slug populations on corn establishment and growth, I evaluated corn population and early season herbivory in all three years of the experiment (2021-2023). Due to time constraints, I did not collect similar data in soybean. When corn plants were in growth stage V3 and V5, I established 3-meter long transects in three randomly selected rows within each plot. For each plant along this transect, I characterized the amount of damage caused by invertebrates and the type of damage (slugs, true armyworm, black cutworm, stink bug, multiple, or other) for each plant. I rated amount of damage using a scale from 0-4: 0 (no damage), 1 (<25% defoliation), 2 (25-50% defoliation), 3 (50-75% defoliation), 4 (>75% defoliation; Douglas, Rohr, and Tooker 2015).

In addition to assessing amounts of damage they caused, I also quantified slug populations weekly. I conducted these evaluations in corn plots in 2021, and corn and soybean plots in 2022 and 2023. Between crop rows 6-7 and 10-11, I deployed two shingles per row (approximately 5 and 10 m from the western edge of each plot; total of 4 singles per plot). Shingle size was 0.093 m² and were laid flat on the ground between crop rows (Busch et al., 2020; Byers & Calvin, 1989). In cover crop plots, I clipped away foliage to allow shingles to rest on soil, and then placed the foliage on top of the shingle to best match the moisture and temperature conditions of the matted cover crop. Assessments began immediately following

crop planting until corn growth stage V6 and then began again from R5 through harvest. Early season sampling was done to monitor slug populations throughout the vulnerable corn seedling and early growth stages. For ease of sampling, I matched soybean sampling with corn. To sample during the coolest parts of the day, I conducted assessments as early in the morning as possible (6:00-8:00). For each evaluation, I counted all slugs on top of and underneath the shingle.

2.3. Predator community and predation

To evaluate the influence of the timing of cover-crop termination on abundance and diversity of ground-dwelling predators, during 2022 and 2023 in corn and soybeans I deployed two pitfall traps near the center of each plot (5 and 10 m from the western edge of each plot). Traps consisted of 7.6 x 7.6 cm plastic cups filled with a 50:50 mixture of propylene glycol and water and were covered with plastic plates perched on nails (7.6-cm tall) to prevent rain and debris from falling in (Busch et al., 2020; Carmona & Landis, 1999). I left pitfalls in the field for 48 h and then collected them and returned the contents to the lab to sort arthropods. In 2022, I deployed pitfall traps monthly, starting shortly after crop planting (28 May 2022) and extending until crop maturity (growth stage R5-6; final trapping date for corn: 1 July 2022; final trapping date for soybeans: 18 August 2022). Final sampling dates in 2022 differed between corn and soybean due to sampling errors. In 2023, I conducted this assay monthly in both corn and soybean, starting post-plant (corn stage V3; 26 June 2023) until crop maturity (growth stage R5-6; 15 September 2023). Due to high levels of in-field vertebrate disruption, I excluded 15 September 2023 samples from analysis.

To assess the influence of the timing of cover crop termination on predation by generalist invertebrate predators, I conducted sentinel prey assays in corn and soybean across corn growth stages (Lundgren et al. 2006; Busch et al. 2020). I used waxworms (*Galleria mellonella*; Timberline Fisheries, Marion, IL) as sentinel prey. I pinned waxworms through their terminal segment into balls of modelling clay. I deployed six waxworms per plot in vertebrate exclusion cages with three between rows 6-7 and 11-12 (approximately 4, 8, and 12 m from the western edge of each plot; total of 6 cages per plot). I deployed waxworms at ~8:00 and checked them at ~20:00 and again the next day at ~8:00, recording presence/absence and noting any predators (e.g., wolf spider, carabid beetle, ant, etc.) that were present. Missing waxworms were not replaced with living ones. To improve efficiency of our assessment of predation, I deployed sentinel prey in corn and soybean on the same days (Table 1-2).

Table 1-2. Sentinel prey assessment dates with corn and soybean growth stages.

Date	Corn Growth Stage	Soybean Growth Stage
6/16/2021	V3	V2-3
6/24/2021	V5	V4
8/24/2021	R3	R4
6/22/2022	V3	V2-3
7/12/2022	V5	V4
9/1/2022	R3	R4
6/20/2023	V3	V2-3
7/15/2023	V5	V4
9/15/2023	R3	R4

3. STATISTICAL ANALYSIS

I conducted all statistical analyses in R (version 4.3.1). To analyze effects of timing of cover-crop termination, I ran a generalized linear mixed-effect model (GLMM) for the following data collection types: cover-crop biomass (Gaussian distribution), yield (Gaussian distribution), damage incidence (binomial distribution), damage score (binomial distribution), damage type (binomial distribution), slug counts (negative binomial distribution), and sentinel prey (binomial distribution). Cover-crop biomass and yield were run with a fixed effect term of treatment*year to capture the main effects and their interaction. For both data types, the random effect term was block.

When conducting repeated measures for damage incidence, damage score, damage type, and sentinel prey, to capture the main effects of both treatment, timing (i.e., growth stage), and their interaction, the fixed effect term was treatment*timing. To account for non-independence in the random structure, terms were nested. Timing (i.e., growth stage) was chosen as the random effects expression in the nested random effects term to account for the repeated measure of sampling within the same plot several times throughout the season. I ran the following data collection types with a repeated measure: damage score, damage type, sentinel prey, and slug counts. For damage score, the random effects term was block nested within year. For damage incidence, damage type and sentinel prey, the random effect term was plot nested within block within year. For slugs, the fixed effect term was structured to capture the main effects and interactions between treatment and year (treatment*year) and the random effect term of was block. Slug data were averaged among years, blocks, and treatments and split between spring (planting-V6) and fall (R3-crop maturity) due to an imbalance in sampling observations.

To determine the significance of fixed main effects and interactions, the “anova” function was used to obtain log-likelihood ratio tests and the Wald X^2 test statistic. The anova output allowed a

comparison of null models to fully-fitted models, adding one effect at a time. For linear models, I ran a post hoc analyses with the ‘emmeans’ package (Lenth, 2023).

To analyze changes of arthropod communities by crop, sampling date, and treatment sampled with pitfall traps, I used permutational multivariate analysis of variance (PERMANOVA; ‘vegan’ package; Oksanen et al. 2022). I used Bray-Curtis dissimilarity to analyze count data and set permutations to 999. I aggregated predators into four main groups: Araneomorph, Carabid, Ensifera, and Formicidae. These groupings comprise the four most common predator groups recorded in 2022 and 2023. I also analyzed Lycosidae populations within the Araneomorph suborder.

I analyzed predator and slug interactions with a generalized linear model (GLM, Negative binomial distribution; ‘MASS’ package Venables and Ripley 2002). The values used in this model were averages of populations by treatments over the two years of sampling. The response variable was slug populations and the fixed effect was predator populations.

To analyze the legacy effects of cover-crop termination treatments, I ran a GLMM for the following data collection types: slug counts, sentinel prey, and pitfall samples. To account for non-independence, I nested plot within block. To capture the repeated measures effect of overlaying the same treatments in subsequent years, I chose crop as my random term. In all these models, the fixed effect term was treatment*crop to capture the main effect of both, and their interaction.

To analyze the differences in populations of Carabidae, Araneomorphae, and Lycosidae between crops, I ran GLM (Negative binomial distribution) models with crop as the fixed effect term.

4. RESULTS

4.1. Cover-crop biomass

In corn, I found differences among all treatments except between late-terminated and planting-green treatments in 2023 (Fig. 1-2). It is important to note the cover crop planting dates differed (Table 1-1).

In soybean, all cover-crop treatments differed in 2022 and in 2023 the early-terminated treatment had significantly lower biomass than both late-terminated and planting-green treatments (Fig. 1-3).

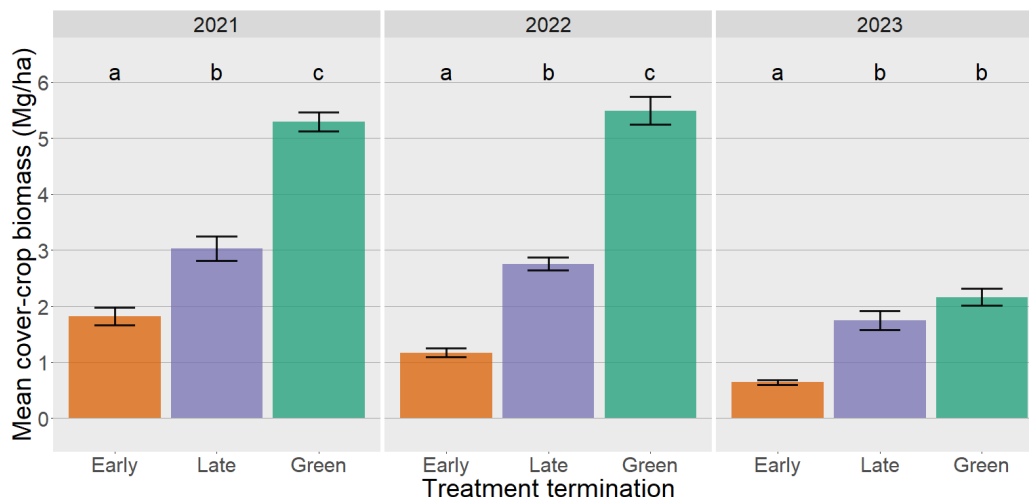


Figure 1-2. Cover-crop biomass from corn plots split by treatment and year. In 2021 and 2022, all cover-crop treatments differed. In 2023, early-terminated was lower than the late-terminated and planting-green treatments, but late-terminated and planting-green treatments did not differ. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

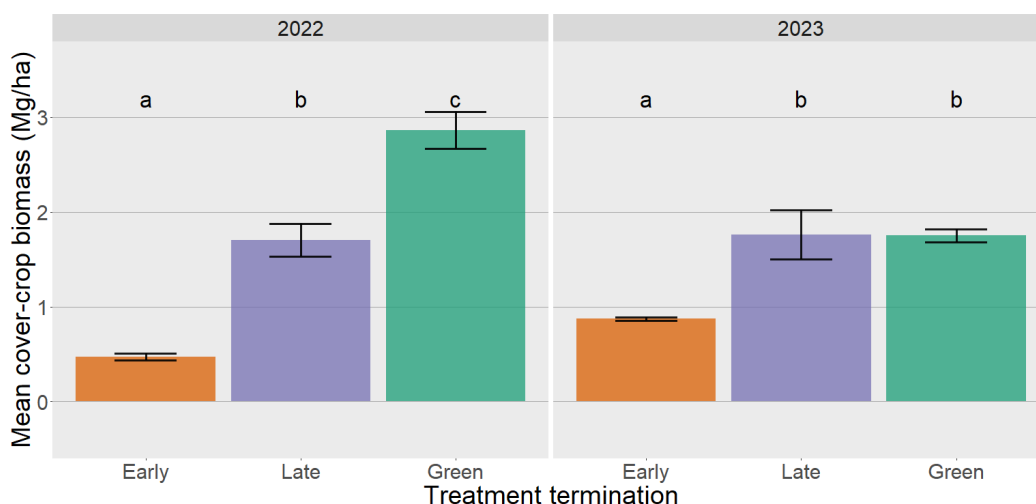


Figure 1-3. Cover-crop biomass from soybean plots split by treatment and year. In 2022, all cover-crop treatments differed. In 2023, early-terminated was lower than the late-terminated and planting-green treatments, but late-terminated and planting-green treatments did not differ. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

4.2. Damage Incidence and score (Corn only)

I recorded a significant main effect of treatment ($df=3$, $X^2=11.9$, P -value=0.008) and the interaction of treatment and growth stage ($df=3$, $X^2=8.9$, P -value=0.03) on the proportion of damaged plants (Fig. 1-4). During both V3 and V5 growth stages, the planting-green treatment had the highest

proportion of plants damaged. During growth stage V3, the no-cover-crop treatment was lower than the early-terminated and planting-green treatments. During growth stage V5, the early-terminated and late-terminated treatments were lower than the planting-green treatments.

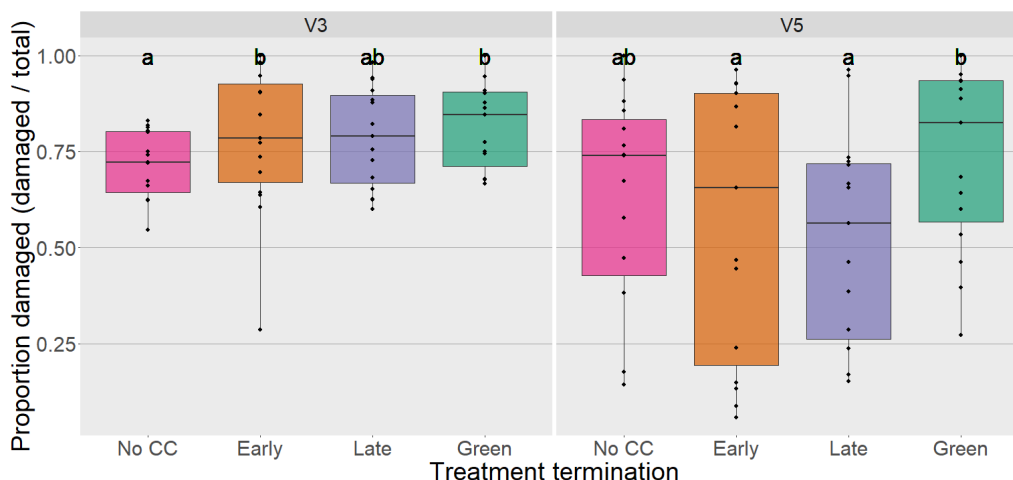


Figure 1-4. There was a significant main effect of treatment ($df=3$, $X^2=11.9$, P -value=0.008) and the interaction of treatment and growth stage ($df=3$, $X^2=8.9$, P -value=0.03) on the proportion of damaged plants. During both V3 and V5 growth stages, the planting-green treatment had the highest proportion of plants damaged. During growth stage V3, the no-cover-crop treatment was lower than the early-terminated and planting-green treatments. During growth stage V5, the early-terminated and late-terminated treatments were lower than the planting-green treatments. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

I recorded a significant main effect of treatment ($df=3$, $X^2=75.3$, P -value<0.001) and the interaction of treatment and growth stage ($df=3$, $X^2=26$, P -value<0.001) on average damage. During growth stage V3, the highest average damage scores were in the planting-green and early-terminated treatments, followed by the late-terminated and no-cover-crop treatments. The highest average damage score recorded in V5 was the planting-green treatment, which was higher than the other three treatments (Fig. 1-5).

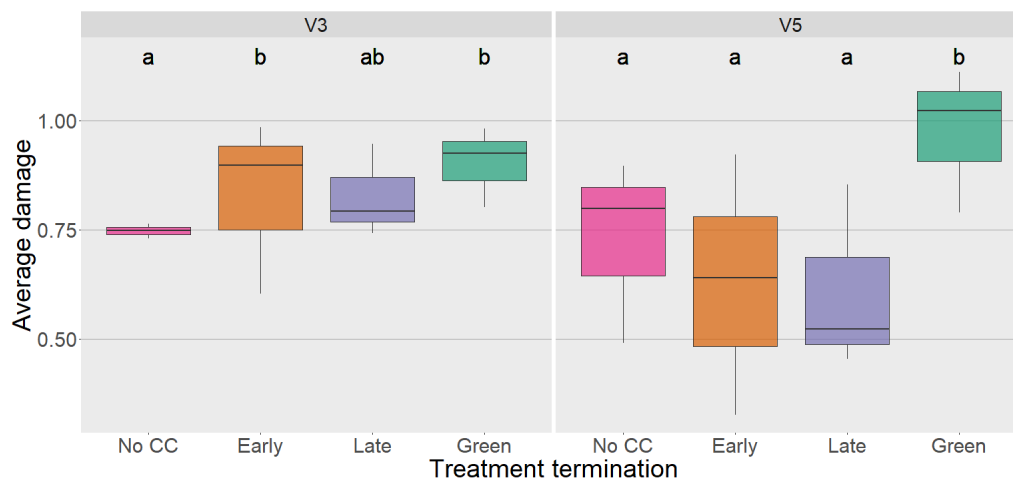


Figure 1-5. Average damaged recorded from 2021-2023 in corn plots. I recorded a significant main effect of treatment ($df=3$, $X^2=75.1$, $P\text{-value}<0.001$) and the interaction of treatment and growth stage ($df=3$, $X^2=26.2$, $P\text{-value}<0.001$) on average damage. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

4.3. Damage Type (Corn only)

There was a significant main effect of the treatment and growth stage interaction ($df=3$, $X^2=10.4$, $P\text{-value}=0.02$) for slug damage. However, I recorded no differences between growth stages or among treatments in overall slug, true armyworm, or stink bug damage. For black cutworm damage, there was a marginal main effect of treatment ($df=3$, $X^2=7.2$, $P\text{-value}=0.06$). During growth stage V3, the no-cover-crop treatment had higher damage than the late-terminated treatment (Fig. 1-6). Black cutworm damage did not differ among treatments during growth stage V5. For multiple pest damage, I recorded a significant main effect of treatment ($df=3$, $X^2=22.5$, $P\text{-value}<0.001$). During both V3 and V5 growth stages, the planting-green treatment had higher damage levels than the other three treatments (Fig. 1-7). For other pest damage, the data I recorded was too low to create a meaningful model.

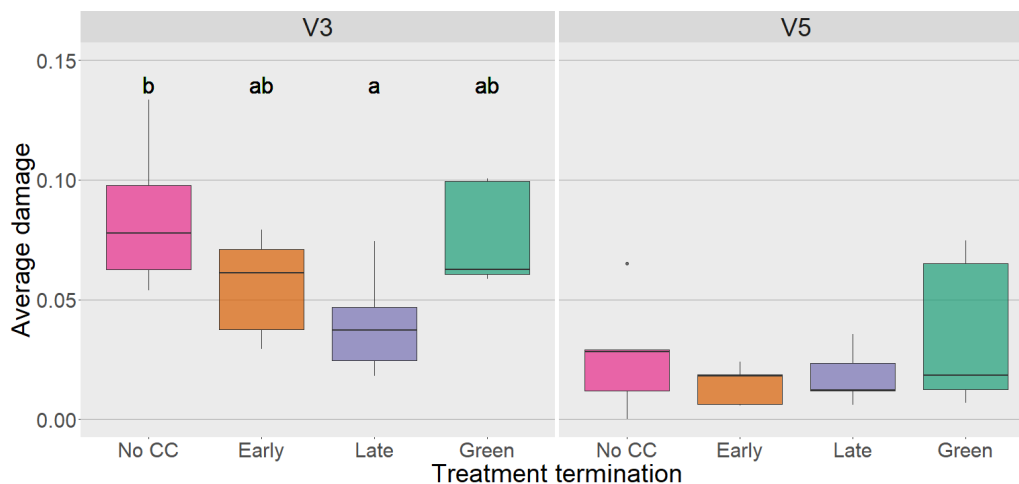


Figure 1-6. Total Black cutworm damage by growth stage and treatment in corn plots. There was a marginal main effect of treatment ($df=3$, $X^2=7.2$, $P\text{-value}=0.06$). During growth stage V3, the no-cover-crop treatment had higher damage than the late-terminated treatment Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

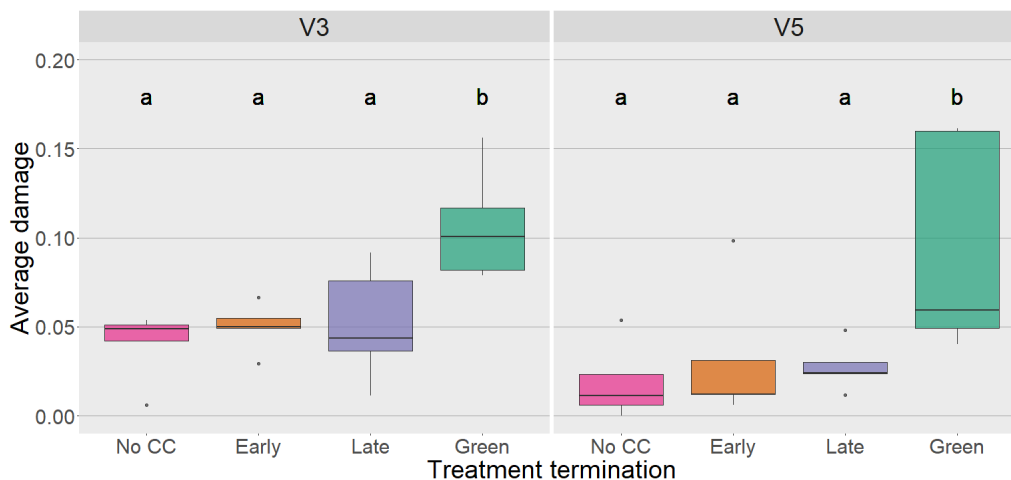


Figure 1-7. Total multiple pest damage by growth stage in corn plots. There was a main effect of treatment ($df=3$, $X^2=22.5$, $P\text{-value}<0.001$). During both V3 and V5 growth stages, the planting-green treatment had higher damage levels than the other three treatments Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

4.4. Slugs

In corn, spring slug populations had a significant main effect of year ($df=2$, $X^2=211$, $P\text{-value}<0.001$). The 2021 season had greater slug populations than the 2022 season but did not differ from 2023 (Fig. 1-8). During 2021, the planting-green treatment had greater slug populations than the no-cover-crop check treatment but did not differ from either the early-terminated or the late-terminated treatments. For slug populations during the fall in corn, there was a significant main effect of year ($df=2$, $X^2=67.4$, $P\text{-value}<0.001$) and the interaction of treatment and year ($df=6$, $X^2=21.6$, $P\text{-value}=0.001$). All three years differed from one another in total slug populations with 2022 having the greatest populations, followed by 2023, then 2021 (Fig. 1-9). During the 2022 season, the no-cover-crop check and the early-terminated treatment had greater slug populations than the planting-green treatment. Additionally, the no-cover-crop check had greater slug populations than the late-terminated treatment. The early-terminated and late-terminated treatments did not differ from one another.

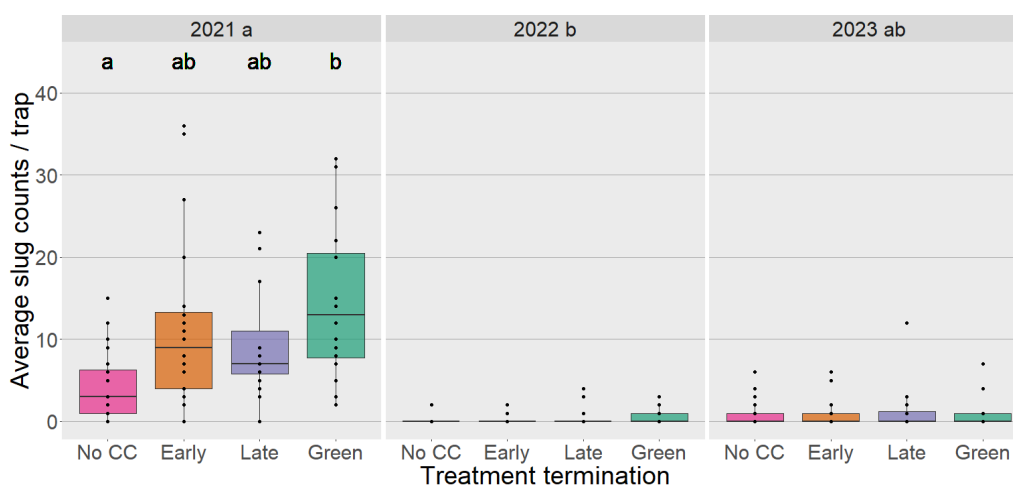


Figure 1-8. Slugs populations by year and treatment from the spring season in corn plots. There was a significant main effect of year ($df=2$, $X^2=211$, $P\text{-value}<0.001$). The 2021 season had greater slug populations than the 2022 season but did not differ from 2023. During 2021, the planting-green treatment had greater slug populations than the no-cover-crop check treatment but did not differ from either the early-terminated or the late-terminated treatments. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

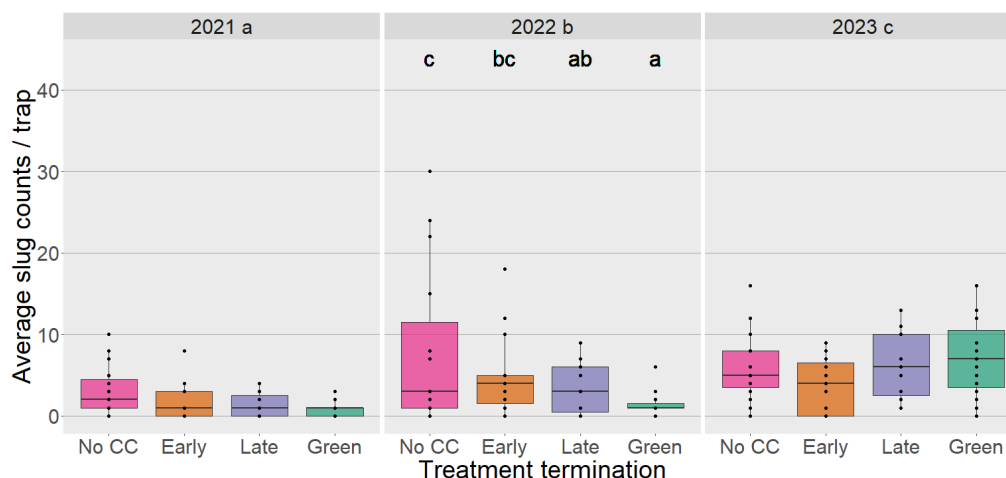


Figure 1-9. Slugs populations by year and treatment from the fall season in corn plots. There was a significant main effect of year ($df=2$, $X^2=67.4$, $P\text{-value}<0.001$) and the interaction of treatment and year ($df=6$, $X^2=21.6$, $P\text{-value}=0.001$). All three years differed from one another in total slug populations with 2022 having the greatest populations, followed by 2023, then 2021. During the 2022 season, the no-cover-crop check and the early-terminated treatment had greater slug populations than the planting-green treatment. Additionally, the no-cover-crop check had greater slug populations than the late-terminated treatment. The early-terminated and late-terminated treatments did not differ from one another. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

In soybeans, there was a significant main effect of year ($df=1$, $X^2=6$, $P\text{-value}=0.01$) for slug populations sampled during spring. Slug populations were greater in 2023 than 2022 (Fig. 1-S1). For slugs in sampled in the fall there was a significant main effect of year ($df=1$, $X^2=181$, $P\text{-value}<0.001$). Populations in 2022 were greater than in 2023 (Fig. 1-S2).

4.5. Predator community and predation

4.5.1. Sentinel prey

In corn, there was a significant main effect of treatment ($df=3$, $X^2=12.4$, $P\text{-value}=0.006$). The no-cover-crop treatment had lower predation levels than the late-terminated treatment (Fig. 1-10). The planting-green, early-terminated, and no-cover-crop check treatments did not differ.

In soybeans, there was a significant main effect of treatment ($df=3$, $X^2=15.6$, $P\text{-value}=0.001$) and growth stage ($df=2$, $X^2=6.7$, $P\text{-value}=0.04$). The no-cover-crop treatment had lower predation levels than the planting-green treatment (Fig. 1-11). The early-terminated and late-terminated treatments did not differ from either the planting-green or no-cover-crop check treatments. It is important to note that while the no-cover-crop check had the lowest predation, 81% predation is still high. Growth stage V3 had fewer prey attacked than growth stage V5 (Fig. 1-12). Growth stages V3 and R3 did not differ. While there was variability in overall predation, it is important to note that 80% predation during growth stage V3 is high.

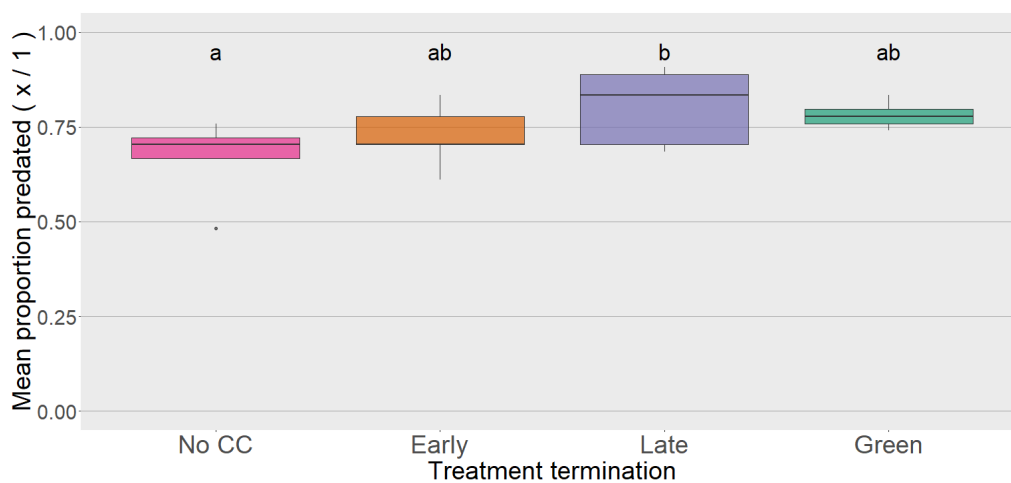


Figure 1-10. Average predation by treatment from corn plots. There was a significant main effect of treatment ($df=3$, $X^2=12.4$, $P\text{-value}=0.006$). The no-cover-crop treatment had lower predation levels than the late-terminated treatment. The planting-green, early-terminated, and no-cover-crop check treatments did not differ. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

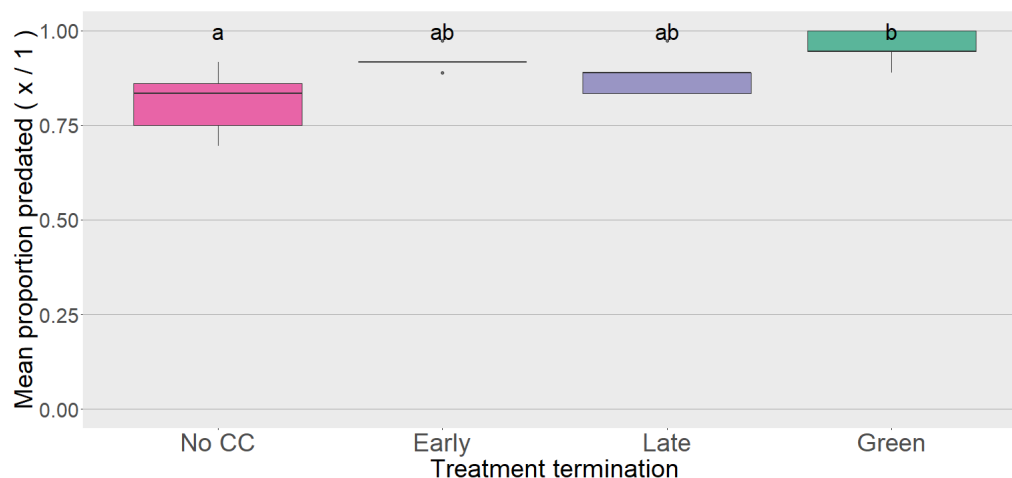


Figure 1-11. Average predation by treatment from soybean plots. There was a significant main effect of treatment ($df=3$, $X^2=15.6$, P -value=0.001). The no-cover-crop treatment had lower predation levels than the planting-green treatment. The early-terminated and late-terminated treatments did not differ from either the planting-green or no-cover-crop check treatments. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

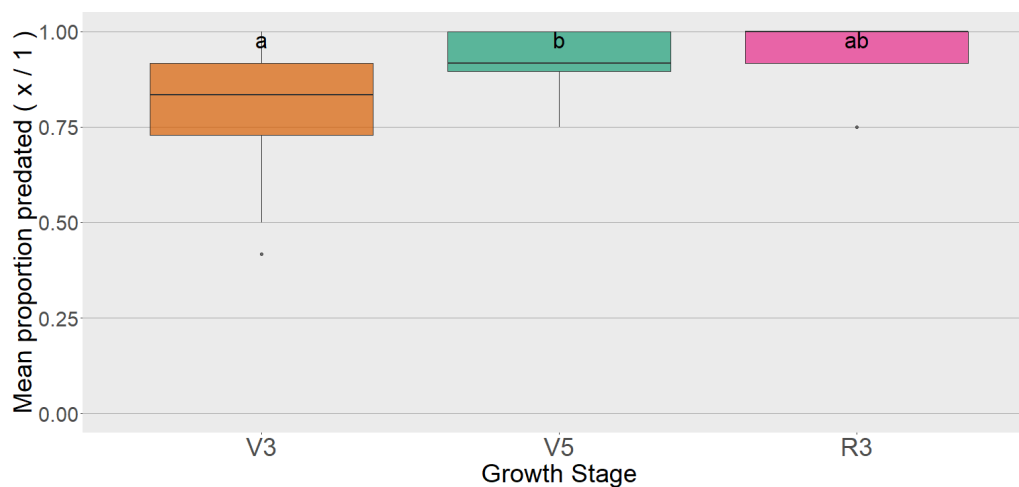


Figure 1-12. Average predation by growth stage from soybean plots. There was a significant main effect of growth stage ($df=2$, $X^2=6.7$, P -value=0.04). Growth stage V3 had fewer prey attacked than growth stage V5. Growth stages V3 and R3 did not differ. Letter change denotes significance.

4.5.2. Pitfall traps

I recorded similar overall arthropod populations per plot in 2022 (Mean \pm SE: 10.5 ± 0.7 , $n = 80$) and 2023 (10.1 ± 1.30 , $n = 80$). Araneomorph (40%) and carabid (30%) populations accounted for most of the predators collected. Lycosidae account for 83% of the araneomorph populations while carabid is dominated by the *Chlaenius* (56%), *Poecilus* (11%), and *Pterostichus* (26%) genera.

Both crop ($F_{1,79} < 0.001$) and sampling date ($F_{2,79} < 0.001$) influenced arthropod populations. Soybean harbored greater populations of arthropods than corn (Figure 1-13). When evaluating carabids, total araneomorphs, and Lycosidae abundance by crop, all three groups had greater abundance in soybean than corn (Figure 1-14).

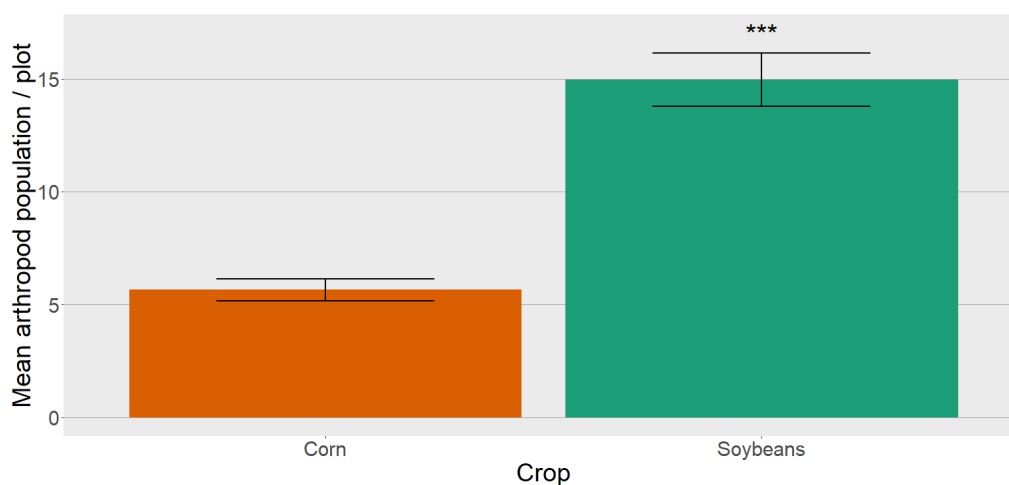


Figure 1-13. Average arthropod populations per plot by crop. Soybean harbored greater overall populations than corn. Asterisks denote significance. (* = $P \leq 0.05$; ** = $P \leq 0.01$; *** = $P \leq 0.001$)

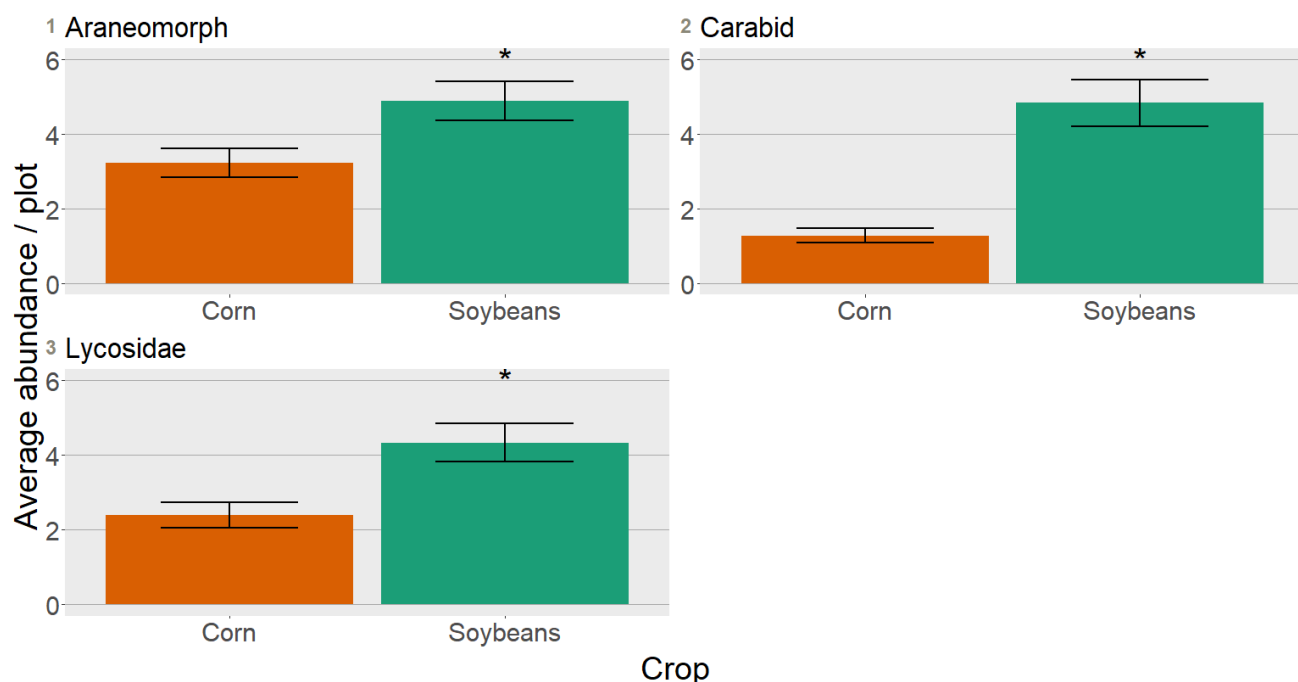


Figure 1-14. ¹Araneomorph, ²Carabid, and ³Lycosidae abundance by crop. Soybean harbored greater abundance of all three groups. Asterisk denotes significance. (* = $P \leq 0.05$; ** = $P \leq 0.01$; *** = $P \leq 0.001$)

4.5.2.1. Corn

The main effects of year and sampling date were significant in explaining the distribution of arthropod populations. Both effects had an $F_{1,79} < 0.001$. Treatment had no effect on arthropod composition or populations.

In both 2022 and 2023, I recorded no differences in araneomorph or carabid populations by sampling date. When comparing total populations of araneomorph and carabids from 2022 and 2023, I found lower ($P \leq 0.001$) araneomorph populations in 2023 (Table 1-6).

4.5.2.2. Soybean

The main effects of year and sampling date were significant in explaining the distribution of the arthropod populations. Both effects had an $F_{3,99} \leq 0.001$. Treatment had no effect on arthropod composition or populations.

In 2022, I recorded no differences in araneomorph populations by sampling date. Carabids had greater abundance during the last sampling date, 18 August 2022, than the earlier two, 28 May 2022 and 1 July 2022 (Table 1-7). In 2023, araneomorph populations did not change between sampling dates. Carabids had greater abundance during the last sampling date, 28 July 2023, compared to the first

sampling date, 26 June 2023 (Table 1-8). When comparing total populations of carabid and araneomorph from 2022 (May 28 and July 1) and 2023 (June 26 and July 28), I found variation in abundance between years (Table 1-9).

Table 1-3. Araneomorph and carabid averages by sampling date from corn pitfalls. This table comprises both 2022 corn sampling dates (28 May and 1 July) and both 2023 sampling dates (26 June and 28 July). Araneomorphs had greater populations in 2022 than 2023.

CLD; Compact letter display: Letter change denotes significance.

N = 20

Sampling date	Araneomorph mean \pm SE	Araneomorph CLD	Carabid mean \pm SE	Carabid CLD
28 May 2022	4.6 \pm 0.6	a	1.4 \pm 0.5	a
1 July 2022	6.7 \pm 0.7	a	0.6 \pm 0.2	a
26 June 2023	0.7 \pm 0.2	b	1.15 \pm 0.3	a
28 July 2023	0.9 \pm 0.3	b	2.00 \pm 0.4	a

Table 1-4. Araneomorph and carabid averages by 2022 sampling dates of pitfalls from soybean plots. Carabid populations increase among the three sampling dates from 2022.

CLD; Compact letter display: Letter change denotes significance.

N = 20

Sampling date	Araneomorph mean \pm SE	Araneomorph CLD	Carabid mean \pm SE	Carabid CLD
28 May 2022	6.5 \pm 1.3	a	3.7 \pm 0.7	a
1 July 2022	8.75 \pm 0.9	a	4.6 \pm 0.6	a
18 August 2022	11.90 \pm 4.9	a	17.5 \pm 2.8	b

Table 1-5. Araneomorph and carabid averages by 2023 sampling dates of pitfalls from soybean plots. Carabid populations increase between the two sampling dates from 2023.

CLD; Compact letter display: Letter change denotes significance.

N = 20

Sampling date	Araneomorph mean \pm SE	Araneomorph CLD	Carabid mean \pm SE	Carabid CLD
26 June 2023	1.8 \pm 0.3	a	2.5 \pm 1.5	a
28 July 2023	2.55 \pm 0.5	a	8.1 \pm 1.8	b

Table 1-6. Araneomorph and carabid averages by sampling date of pitfalls from soybean plots. This table comprises two of the 2022 sampling dates (28 May and 1 July) and both 2023 sampling dates (26 June and 28 July). Araneomorphs had greater abundance in 2022 than 2023. Carabids had the highest abundance during 28 July 2023.

CLD; Compact letter display: Letter change denotes significance.

N = 20

Sampling date	Araneomorph mean \pm SE	Araneomorph CLD	Carabid mean \pm SE	Carabid CLD
28 May 2022	6.5 \pm 1.3	a	3.7 \pm 0.7	a
1 July 2022	8.8 \pm 0.9	a	4.6 \pm 0.6	ab
26 June 2023	1.8 \pm 0.3	b	2.5 \pm 1.5	a
28 July 2023	2.6 \pm 0.5	b	8.1 \pm 1.8	b

4.5.3. Predators and slugs

In corn, I recorded a positive relationship between slug counts and total predator counts, with an $R^2=0.29$ and $P < 0.01$ (Fig. 1-15). In soybean, I recorded a negative relationship between slug counts and total predator counts, with an $R^2=0.83$ and $P < 0.001$ (Fig. 1-16).

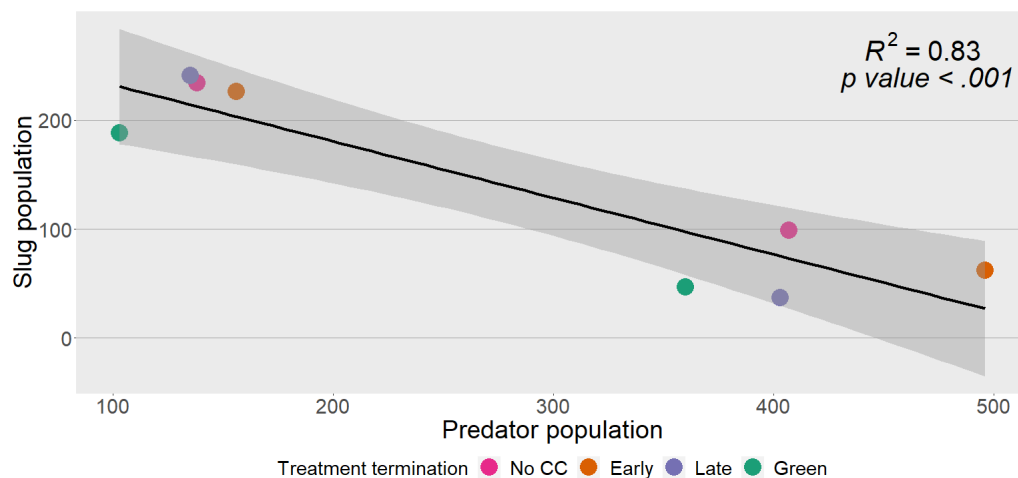


Figure 1-15. Slug and predator population regression from soybean plots. Negative relationship between slug and predator interactions with an $R^2=0.83$ and $P < 0.001$. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

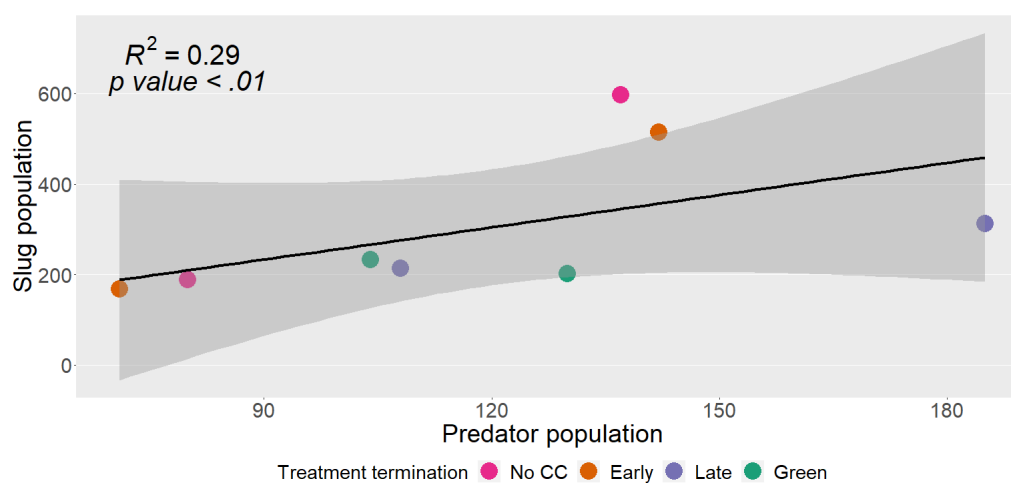


Figure. 1-16. Slug and predator population regression from corn plots. Positive slope between predators and slugs with an $R^2=0.29$ and $P < 0.01$. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

4.6. Yield

In corn, there was a significant main effect of treatment ($df=3$, $X^2=13.7$, $P\text{-value}=0.003$), the interaction between treatment and year ($df=6$, $X^2=18$, $P\text{-value}=0.006$), and year ($df=2$, $X^2=8.5$, $P\text{-value}=0.014$). In years 2021 and 2022 I recorded no differences in yield among treatments (Fig. 1-17). In 2023, the no-cover-crop check was lower than the other three treatments.

For soybean yield, there was a significant main effect of year ($df=1$, $X^2=5.9$, $P\text{-value}=0.015$). The 2022 season had lower overall yields than 2023 (Fig. 1-18).

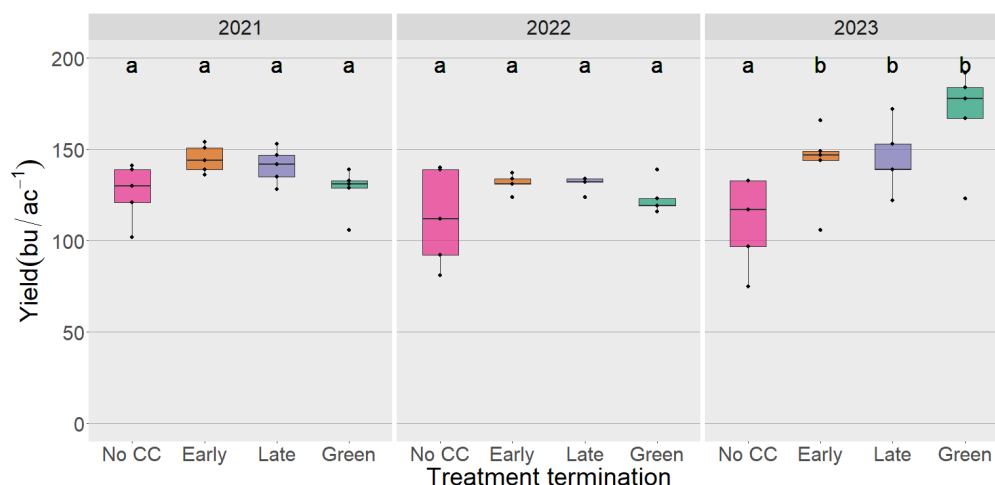


Figure 1-17. Yield in bushels / acre from corn plots. there was a significant main effect of treatment ($df=3$, $X^2=13.7$, $P\text{-value}=0.003$), the interaction between treatment and year ($df=6$, $X^2=18$, $P\text{-value}=0.006$), and year ($df=2$, $X^2=8.5$, $P\text{-value}=0.014$). In years 2021 and 2022 I recorded no differences in yield among treatments. In 2023, the no-cover-crop check was lower than the other three treatments. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

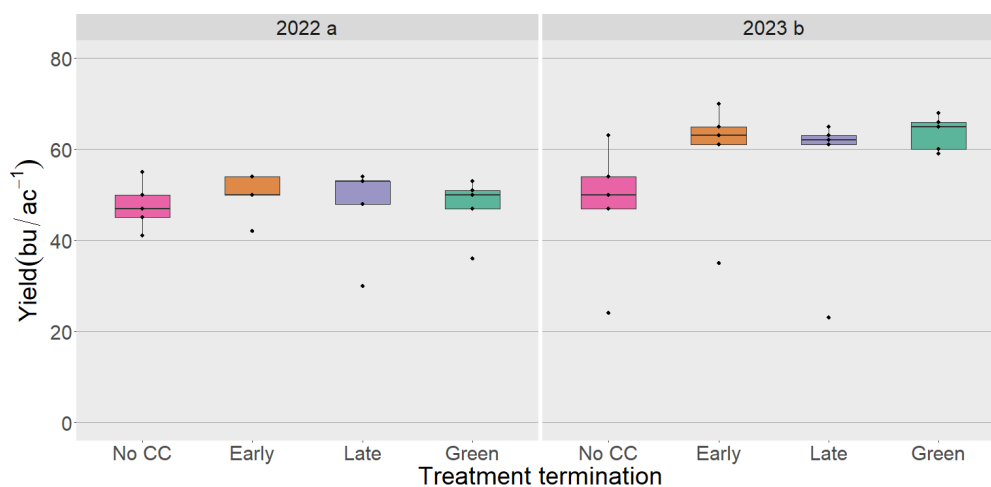


Figure 1-18. Yield in bushels / acre from soybean plots. There was a significant main effect of year ($df=1$, $X^2=5.9$, $P\text{-value}=0.015$). The 2022 season had lower overall yields than 2023. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

4.7. Corn – Soybean sequence

For spring slug counts in the 2021 corn – 2022 soybean sequence, there was a main effect of treatment ($df=3$, $X^2=20.7$, $P\text{-value}<0.001$) and crop ($df=1$, $X^2=9.7$, $P\text{-value}=0.002$). Corn plots had higher slug counts than soybean plots and all pairwise comparisons of treatment and crop differed (Fig. 1-19).

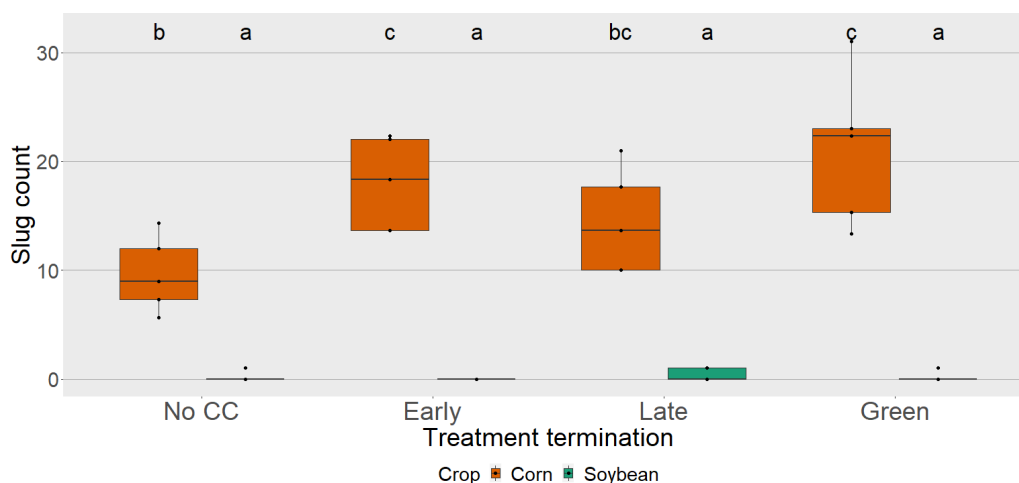


Figure 1-19. Slug populations from the spring 2021 corn-2022 soybean legacy pair. There was a main effect of treatment ($df=3$, $X^2=20.7$, $P\text{-value}<0.001$) and crop ($df=1$, $X^2=9.7$, $P\text{-value}=0.002$). Corn plots had higher slug counts than soybean plots and all pairwise comparisons of treatment and crop differed. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

For the fall counts in the 2021 corn – 2022 soybean sequence, there was a significant main effect of treatment ($df=3$, $X^2=17.6$, $P\text{-value}<0.001$). Soybean slug populations were higher in the no-cover-crop check, early-terminated, and planting-green treatments (Fig. 1-20). Corn had higher slug populations in the late-terminated treatment.

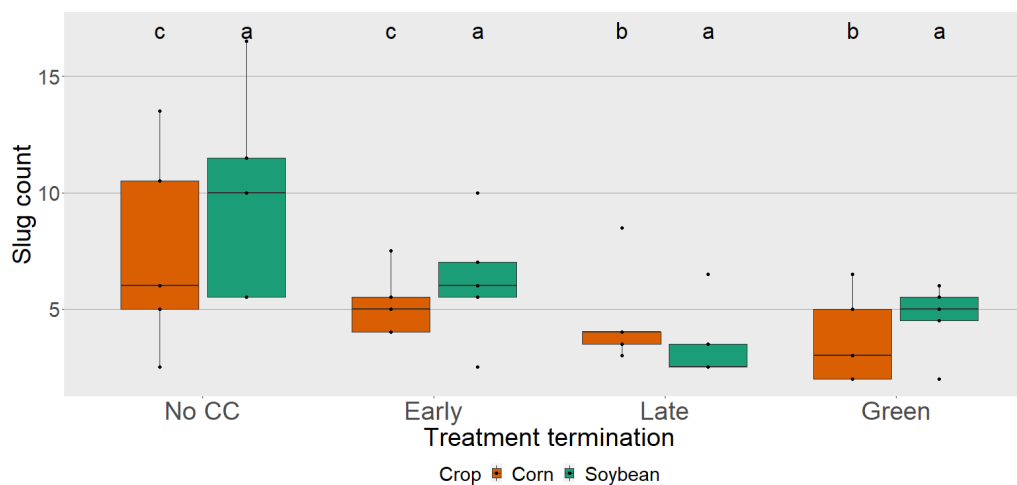


Figure 1-20. Slug populations from the fall 2021 corn-2022 soybean legacy pair. There was a significant main effect of treatment ($df=3$, $X^2=17.6$, $P\text{-value}<0.001$). Soybean slug populations were higher in the no-cover-crop check, early-terminated, and planting-green treatments. Corn had higher slug populations in the late-terminated treatment. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

For spring slug counts in the 2022 corn – 2023 soybean sequence, there was a significant main effect of crop ($df=1$, $X^2=10.6$, $P\text{-value}=0.001$). Soybean plots had higher slug counts than corn plots and all pairwise comparisons of treatment and crop differed (Fig. 1-21).

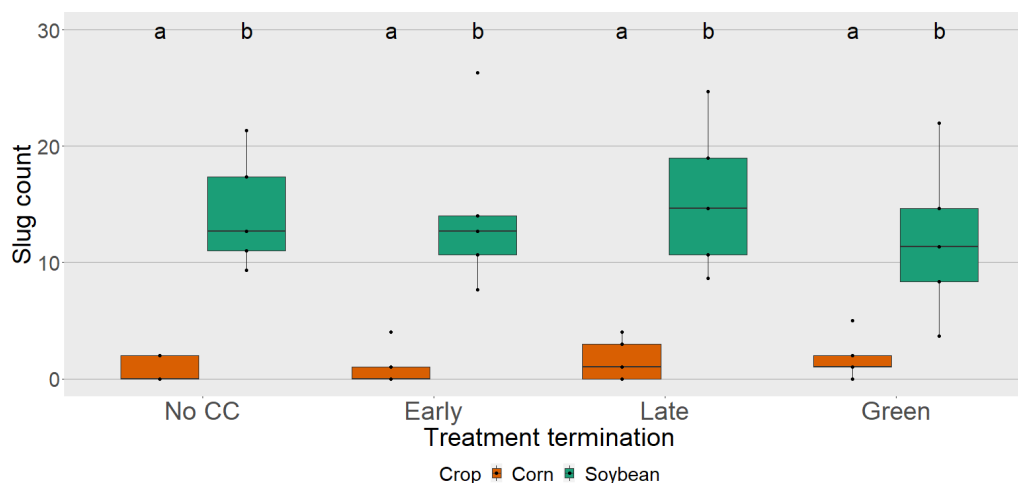


Figure 1-21. Slug populations from the spring 2022 corn-2023 soybean legacy pair. There was a significant main effect of crop ($df=1$, $X^2=10.6$, $P\text{-value}=0.001$). Soybean plots had higher slug counts than corn plots and all pairwise comparisons of treatment and crop differed. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

For fall slug counts in the 2022 corn – 2023 soybean sequence, there was a significant main effect of treatment ($df=3$, $X^2=36.2$, $P\text{-value}<0.001$) and crop ($df=1$, $X^2=20.1$, $P\text{-value}<0.001$). Corn plots had higher slug counts than soybean plots and all pairwise comparisons of treatment and crop differed (Fig. 1-22).

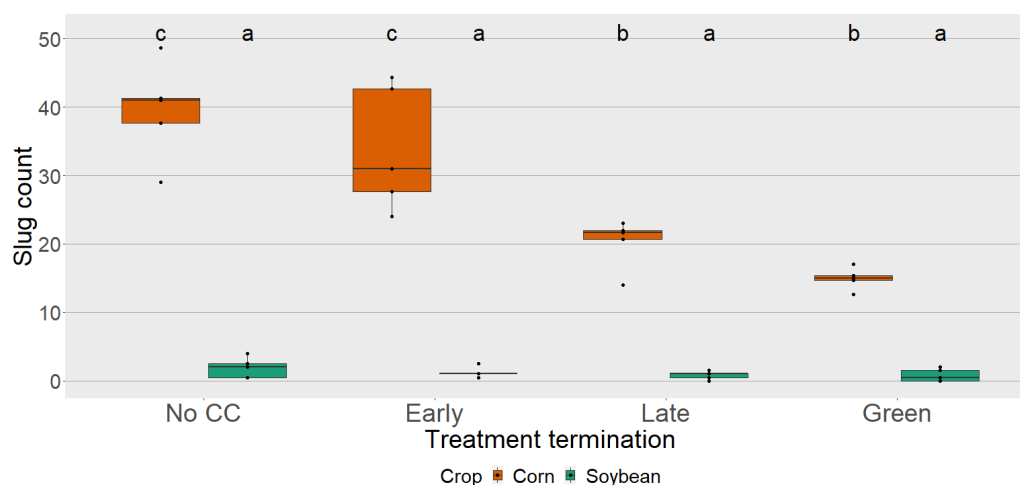


Figure 1-22. Slug populations from the fall 2022 corn-2023 soybean legacy pair. There was a significant main effect of treatment ($df=3$, $X^2=36.2$, $P\text{-value}<0.001$) and crop ($df=1$, $X^2=20.1$, $P\text{-value}<0.001$). Corn plots had higher slug counts than soybean plots and all pairwise comparisons of treatment and crop differed. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

For the 2021 corn – 2022 soybean and 2022 corn – 2023 soybean sequences, I recorded high levels of variation in total prey consumption among treatments.

In the sentinel prey 2021 corn – 2022 sequence, there was a significant main effect of treatment ($df=3$, $X^2=11$, $P\text{-value}=0.01$) and crop ($df=1$, $X^2=8.4$, $P\text{-value}=0.004$). Both the no-cover-crop and early-terminated treatments in soybean had higher levels of predation (Mean \pm Standard Deviation) at $90\% \pm 3\%$ ($n = 90$) and $97\% \pm 2\%$ ($n = 90$), compared to corn at $66\% \pm 5\%$ ($n = 90$) and $71\% \pm 5\%$ ($n = 90$), respectively (Fig. 1-23). I found no differences in the late-terminated or planting-green treatments between crops.

In the sentinel prey 2022 corn – 2023 soybean sequence, there was significant main effect of the interaction between treatment and crop ($df=3$, $X^2=8.5$, $P\text{-value}=0.04$). The only treatment pair with

differences in overall predation was no cover crop, in which corn had higher levels of predation ($90\% \pm 3\%$; $n = 90$) than soybean ($73\% \pm 5\%$; $n = 90$; Fig. 1-24). I recorded no differences in overall predation in the early-terminated, late-terminated, or planting-green treatments between crops.

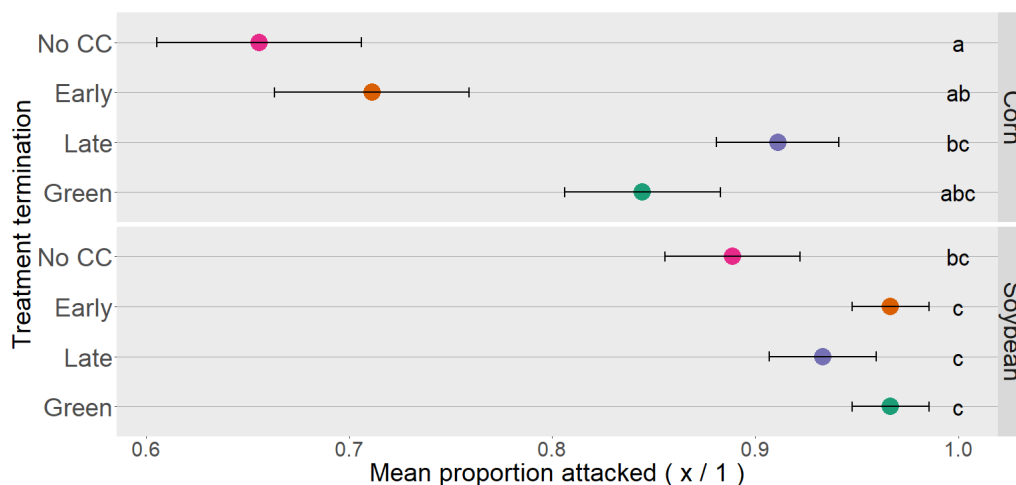


Figure 1-23. Average seasonal proportions of prey consumption by treatment and crop from 2021 corn – 2022 soybean. There was a significant main effect of treatment ($df=3$, $X^2=11$, P -value=0.01) and crop ($df=1$, $X^2=8.4$, P -value=0.004). Both the no-cover-crop and early-terminated treatments in soybean had higher levels of predation (Mean \pm Standard Deviation) at $90\% \pm 3\%$ ($n = 90$) and $97\% \pm 2\%$ ($n = 90$), compared to corn at $66\% \pm 5\%$ ($n = 90$) and $71\% \pm 5\%$ ($n = 90$), respectively. I found no differences in the late-terminated or planting-green treatments between crops. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

In the 2022 corn – 2023 soybean legacy arthropod abundance counts, I recorded a significant main effect of crop ($df=1$, $X^2=9.4$, P -value=0.002). For total arthropod counts by crop, corn had lower ($P < 0.001$) overall counts (Mean \pm Standard Error; 7.62 ± 0.72 , $n = 40$) than soybean (16.5 ± 2.1 , $n = 40$).

I recorded high levels of variation in carabid and araneomorph populations between crops. Carabids had greater abundance in soybean ($P < 0.001$; Soybean: 5.5 ± 1.2 , $n = 40$; Corn: 1 ± 0.3 , $n = 40$) and araneomorphs had greater abundance in corn ($P < 0.001$; Corn: 5.65 ± 0.51 , $n = 40$; Soybean 2.17 ± 0.3 , $n = 40$). When investigating Lycosidae abundance, corn ($P < 0.001$; 4.4 ± 0.5 , $n = 40$) had a greater abundance than soybean (1.72 ± 0.3 , $n = 40$).

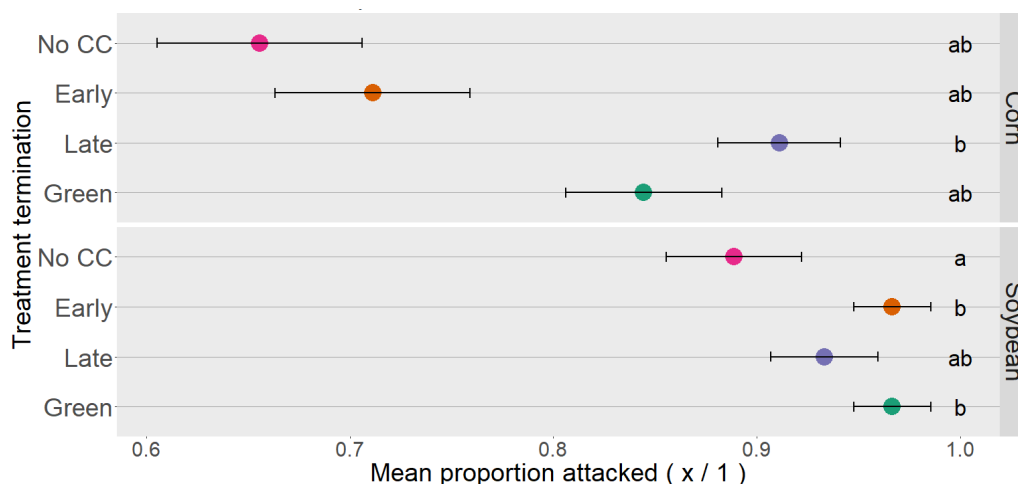


Figure 1-24. Total seasonal proportions of prey attacked by treatment from 2022 corn – 2023 soybean. there was significant main effect of the interaction between treatment and crop ($df=3$, $X^2=8.5$, $P\text{-value}=0.04$). The only treatment pair with differences in overall predation was no cover crop, in which corn had higher levels of predation ($90\% \pm 3\%$; $n = 90$) than soybean ($73\% \pm 5\%$; $n = 90$). I recorded no differences in overall predation in the early-terminated, late-terminated, or planting-green treatments between crops. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

5. DISCUSSION

The aim of this study was to evaluate the effects of increasing cover-crop biomass on pests and predators within corn and soybean systems in Pennsylvania. The results are nuanced and do not entirely corroborate my hypotheses that increasing cover-crop biomass results in an increase in yield and reduction of pests and their damage.

With greater cover-crop biomass, I found little to no increase in damage by invertebrate pests. While I hypothesized that there would be less damage as cover-crop biomass increased, I generally recorded few significant changes. For proportion of plants damaged within a plot, the planting-green treatment was among the highest for both V3 and V5 growth stages (Fig. 1-4). For average damage, the planting-green treatment was again among for both V3 and V5 growth stages (Fig. 1-5).

For individual pest species, I found no difference among treatments in damage incidence by true armyworm. While there was a significant effect of the interaction between treatment and growth stage for slug damage, I recorded no differences between growth stages or among treatments in overall slug damage.

For black cutworm, I detected variation among treatments during the V3 growth stage (Fig. 1-6) which could be attributed to potential early season damage by black cutworm larvae. These results are opaque and unexpected. The planting-green, early-terminated, and no cover-crop treatments shared similar levels of damage. I expected there to be an increase in damage with more cover-crop biomass since black cutworm are early-season pests and preferentially feed on grasses, but I recorded no such trend. When multiple pest species attacked corn plants, however, termination timing influenced amounts of damage corn plants received (Fig. 1-7). This trend contradicts my hypothesis and findings from previous studies (Finke & Denno, 2002), but is consistent with studies that highlight potential for greater pest damage with more cover-crop biomass (Douglas & Tooker, 2012; Reed et al., 2019; Willson & Eisle, 1992).

While increases in slug abundance and their damage are a common concern among growers, I found no such trend. Rather, I found that greater cover-crop biomass did not increase slug populations. In corn plots, rather than cover-crop biomass influencing slug populations, it appears that year had the greatest effect on slug populations. This could be attributed to annual abiotic changes, like precipitation timing, which has been reported frequently as a driver in slug populations (Douglas & Tooker, 2012; Godan, 1983; South, 1992). It is important to note that slug populations varied considerably between spring and fall (Figs. 1-8, 9 and 1-S1, S2). While slugs had greater populations in corn during spring 2021, I detected no significant change in slug damage among treatments. This seasonal variation did not contribute to increased slug damage in either crop species but is a potential factor of poor stands during early season cereal and row-crop production (Godan, 1983; South, 1992). During early vegetative stages, row crops are vulnerable to pest populations, often leading to growers to adopt preventative-pest-management strategies, like seed treatments (Douglas & Tooker, 2015). These results suggest that slug populations can be kept under control by natural-enemy populations in planting-green systems managed with IPM (Le Gall & Tooker, 2017).

In both corn–soy crop sequences, I recorded high levels of variation in slug populations among treatments and between crops (Figs. 1-22-24). While my results demonstrate that no-till and cover crops did not increase slug populations and damage, slugs remain a concern and farmers should track their populations to understand the risk they pose to their fields, especially during early-season vegetative growth stages (Douglas & Tooker, 2012; Godan, 1983; South, 1992).

In both corn and soybean, predation of sentinel caterpillars generally increased with more cover-crop biomass (Fig. 1-10, 11). Indeed, predation during my sampling period at soybean growth stage R3 was nearly 100% in the planting-green treatment, whereas the no-cover-crop treatment predation rate was roughly 80%. This finding confirms that increasing complexity in agricultural fields can increase predator activity (Finke & Denno, 2002; Landis et al., 2000). Due to increases in predation rates with more cover-

crop biomass, these results corroborate the hypothesis of predator response to ecosystem complexity (Finke & Denno, 2002). These results suggest that preventative-pest-management strategies, like insecticidal seed treatments, may not be warranted in high-cover biomass systems.

In addition to greater prey consumption with higher cover-crop biomass, predation also increased throughout the season. In soybean plots, predation at growth stage V3 (~16-22 June) was significantly lower than at growth stages V5 (~24 June-15 July). This increase in predation as the season progressed may be driven by carabid beetles in the genus *Pterostichus*, the adults of which are active mid-summer into autumn and can account for a majority of predation in our region (Busch et al., 2021); however, carabid populations appeared equal for my 28 May 2022, 1 July 2022, 26 June 2023, and 28 July 2023 sampling dates in corn in 2022 and 2023 (Table 1-6). This relatively even distribution of carabid beetles in corn plots could explain why there was no increase in predation rates in corn plots as the season progressed. Having detected no changes in carabid populations in pitfall traps leads me to hypothesize that cover-crop biomass increased a broader diversity of predatory invertebrate taxa, not just the genus *Pterostichus*, but further experimentation will need to explore this hypothesis.

In the 2022 corn – 2023 soybean rotation, predator abundance increased in soybean. This response by natural enemies may be attributed to the legacy effects of the cover-crop treatments. My results do not directly corroborate this theory, but rather present that soybeans in the Northeast may be more hospitable for natural enemy populations. Corroborating an increase in predators due to the legacy effect, carabids had higher populations in soybean. Contradicting the previously mentioned legacy effects, araneomorphs (dominant family: Lycosidae) had higher populations in the preceding corn crop. This finding is interesting, but somewhat expected because these two groups represent the most abundant predators sampled and it is unlikely that they would dominate the same space. While it is uncommon for spiders to consume adult Coleoptera due to their hardened elytra and exoskeletons (Pekár et al., 2012; Riechert & Łuczak, 1982), intraguild consumption may be occurring, which is a highlighted concern among generalist predators (Snyder & Wise, 2001).

In corn plots, predator populations increased with slug populations. This population trend may be due to the nature of the corn agroecosystem. Corn fields are less hospitable than soybean fields because of the lower number of plants per acre (around 30,000 vs. 140,000) and their time to canopy closure is longer, which allows more sun to reach the soil surface which can lead to lower biodiversity (Landis et al., 2000). I hypothesized that by growing a cover crop before corn, late-terminated crops, including those planted green, gain complexity in ground-cover, which can increase pests (slugs) and predators, but also increase pest consumption by natural enemies in response to the increase in pests ((Riechert & Lockley, 1984). With this increase in pest consumption, one could expect similar crop damage by slugs among treatments. Furthermore, although I saw a positive slope in the regression between corn slugs and

predators (Fig. 1-16), I did not find more slug damage in the late-terminated or planting-green treatments, consistent with my hypothesis.

In soybean, however, I found a negative relationship between slug abundance and predator populations (Fig. 1-15). I attribute this response to soybean harboring invertebrate-predator populations 2-3 times higher than corn. With more foliage that overlaps within and between rows, soybean fields have higher humidity under their shaded canopy, a condition more hospitable for arthropod communities, which can sustain higher populations of natural enemies. By adding cover crops for ground cover, soybean systems could see a further increase in natural-enemy populations rendering slug pests unable to maintain populations.

The relationship between soybean and corn slug – predator interactions is one of further consideration. Due to the compounds (cellulose and lignin) in corn, it can be generally less palatable than soybean to a rich diversity of invertebrate taxa (Martin et al., 1991; Prins & Kreulen, 1991), and thus I would expect to see higher populations of herbivores in soybean. For slugs, this was not the case. The greater plant population per acre in soybean may be contributing to the decline in slugs in soybean systems, because higher levels of complexity can improve habitat for natural enemies (Landis et al., 2000). Perhaps, these relationships are attributable to the response of predators to agroecosystem complexity (Finke & Denno, 2002), but further investigation into this would be prudent.

For yield, I found that high-biomass treatments did not reduce yield. Instead, the highest yielding treatment of 2023 was the planting-green treatment. Similar yields across the four treatments in both corn and soybean are consistent with the findings of previous research from the same region (Reed et al., 2019). While this trend does not support my hypothesis that yield would be higher with increases in cover-crop biomass, reporting no changes in yield may help increase adoption of planting-green practices. Although there was slightly more damage in the planting-green treatment, there was minimal, if any, yield drag.

Further investigation into the complexity of the pest and predator interactions in high cover-crop biomass systems may be considered useful to further corroborate these findings. My results support the combination of no-till planting and high-biomass-cover cropping to increase natural enemy populations and lower the reliance on preventative-pest-management strategies.

6. CONCLUSIONS

Despite concerns in agronomic systems of increasing pest populations and reducing yield by planting green (Douglas & Tooker, 2012; Willson & Easley, 1992), my results suggest that planting green: *i*) may not increase the amount of pest damage or damage incidence, *ii*) can promote predation, and *iii*) may not

limit yield in either corn or soybean. In the mid-Atlantic region, studies have shown the benefits that come from planting green and practicing IPM (USDA, 2015). These benefits include, but are not limited to a, reduction in the potential for soil erosion (Renard et al. 1991; Gyssels et al. 2005), improved soil quality metrics (Gyssels et al., 2005), reduction of nutrient leaching into watersheds (Alonso-Ayuso et al., 2014; Farsad et al., 2011; USDA, 2015), and decrease of overall weed and invertebrate pest pressures, which is supported by findings and previous studies (Bohnenblust et al., 2014; Douglas & Tooker, 2012; Finke & Denno, 2002; Hammond & Stinner, 1987; Landis et al., 2000; Mischler et al., 2010; Rowen et al., 2020; Thorbek & Bilde, 2004). My results suggest that these practices be adopted by row-crop growers across the United States looking to decrease invertebrate pest damage and incidence and increase natural-enemy populations.

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Chapter 2

An Evaluation of Benefits by Generalist Spiders in Modern Soybean (*Glycine max.*) Production

1. INTRODUCTION

Preventing pests from causing economic damage to crops is a primary agronomic challenge. Historically, pest management was achieved through integrated pest management (IPM), which involved understanding pest biology and introducing cultural-, mechanical-, and biological-control measures before chemical control (Barzman et al., 2015; Kogan, 1998). While the IPM definition is relatively flexible, the key points remain the same. “Integrated pest management emphasizes the growth of a healthy crop with the least possible disruption to the agroecosystem and encourages natural pest control mechanisms”, as defined by the European Union Framework Directive on the Sustainable Use of Pesticides (Barzman et al., 2015). The means of natural pest control using a living organism, whether introduced or manipulated, to control another (typically a pest) is referred to as *biological control* (Smith, 1919).

A form of biological control that can be used in agricultural systems is augmentative biological control (Waage et al., 1988). Augmentation refers to increasing populations of natural enemies (Rabb et al., 1976) by breeding and releasing (Collier & Van Steenwyk, 2004). In annual cropping systems, this form of pest control historically has lower rates of use because of the associated in-field disturbances (Landis et al., 2000). Due to the complex nature of agroecosystems, seldom can one natural enemy or biological control organism attribute to the bulk of the control (Huffaker & Messenger, 1976). Because of this, increasing habitat suitable for a suite of generalist natural enemies within annual systems, or conservation biological control, may be considered the most practical approach to leveraging biological control.

Although contributions of spiders in pest control of annual agricultural systems is somewhat unclear, studies have shown that population responses of generalist spider predators appears to be proportional to that of prey populations (Riechert & Lockley, 1984). Of the 400-800 million metric tons of prey killed by spiders, roughly 95% is attributed to spiders of forests and grasslands (Nyffeler & Birkhofer, 2017). Spiders of annual cropping systems (agrobiont, Luczak, 1979) only account for 2% of annual prey killed by spiders (Nyffeler & Birkhofer, 2017), likely due to the highly disruptive nature of annual cropping systems (Landis et al., 2000).

In terrestrial ecosystems, spiders (Arachnida: Order Aranea) are among the most ubiquitous predatory animals (Riechert & Lockley, 1984; Turnbull, 1973). As insects diversified over evolutionary

time, it appears that spiders, their main predators, did the same (Selden, 2016). Each year, spiders consume 400-800 million metric tons (fresh weight) of prey, 90% of which are insects and other arthropods (Nyffeler & Birkhofer, 2017). Although the main food source of spiders is insects, they also exhibit araneophagy and cannibalism (Nyffeler, 1999). The most diverse suborder in Aranea is Araneomorphae and includes three main hunting strategies (Humenik et al., 2011; Tonhasca Jr., 1993). These include web building (e.g., Araneidae; Humenik et al., 2011; Schmidt et al., 2008), vagabond hunters (e.g., Salticidae; Turnbull, 1973), and ambush hunters (e.g., Thomisidae; Turnbull, 1973). The hunting modality attributes of araneomorph spiders allow them to exploit multiple prey types within different ecosystems and throughout different times of the year. This prey exploitation makes their potential for pest consumption in agricultural systems of increasing interest.

Agricultural systems continue to intensify while available arable land is diminishing (Kopittke et al., 2019). This intensification can lead to a deficit in natural-enemy populations (Finke & Denno, 2002; Landis et al., 2000). Furthermore, agricultural intensification is the leading cause of biodiversity losses, which are predicted to increase during the first half of the 21st century (Tilman et al., 2001). Loss of biodiversity can be related to the highly disruptive nature of agricultural systems (Goulson, 2013) and the conversion of natural systems to agroecosystems (Tilman et al., 2001). These agroecosystems, in turn, are over-simplified and semi-artificial in their design, and are generally not very hospitable to beneficial invertebrates (Landis et al., 2000). When compared to organic farming systems, natural-enemy populations, including spiders, in modern agricultural systems harbor significantly lower abundance and diversity of natural enemies (Bengtsson et al., 2005; Hole et al., 2005; Rhmann, 2011).

In response to these trends of biodiversity loss, finding ways to improve agroecosystem quality and support natural-enemy populations has been growing in interest (Kassam et al., 2019). By increasing suitable habitats within crop fields, these agroecosystems hold potential to harbor more natural enemies that can contribute to pest control (Landis et al., 2000; Rowen et al., 2020; Tonhasca Jr., 1993). One method of increasing habitat within the field is by adding non-cash-crop habitats (Birkhofer et al., 2013), like cover crops. When evaluating spider abundance by landscape composition, it was found that with an increase in non-cash-crop habitat came an increase populations of the majority of the spider taxa evaluated (Schmidt et al., 2008). With this increase in cover-crop habitat, one would expect to see more spiders and higher levels of associated pest control (Riechert & Lockley, 1984; Turnbull, 1973).

With this understanding of density-dependent spider and pest populations, I set out to evaluate the populations and potential control that can be provided by foliage-dwelling spiders in soybean (*Glycine max*) fields. In two complementary experiments, I asked several questions: 1) Which generalist predator group(s) is/are the most abundant? 2) How will a cereal-rye (*Secale cereal* L.) cover crop affect foliage

dwelling and web building spider populations? 3) Does a cover crop maximize the amount of time agrobiont spiders spend in the field? 4) Can I attribute numerical values of pest control to spiders? 5) If I can artificially increase foliage-dwelling spider populations, will I see a decrease in invertebrate herbivore assemblages?

I hypothesized that *i*) spiders are the most abundant generalist predators in the leaf canopy of soybean, *ii*) spider populations will be greater in cash crops established into living cover crops when the agroecosystem has more cover-crop biomass, and *iii*) foliage-dwelling spiders can limit herbivorous pest populations.

2. MATERIALS AND METHODS

2.1. Study location and experimental design

To understand the value of spiders for pest control in Pennsylvania fields crops, I conducted two studies over two years (2022-2023) at Penn State's Russell E. Larson Agricultural Research Center (Pennsylvania Furnace, Pennsylvania, U.S.A.; 40.720064, -77.934317). The first explored the influence of the timing of cover-crop termination on spider populations in subsequent corn and soybean crops (referred to here as 'Cover-crop termination'). The second involved manipulating spider populations in soybean plots to provide insight on the role spiders play in pest control (referred to here as 'Augmentation').

2.2. Cover-crop termination experiment

In a long-term no-till field (>10 yr.), I established a single-factor experiment, arranged in a randomized complete block design with four cover crop treatment levels and five replicates. Plot size was 12 x 15 m. Cover crop treatments varied by termination timing of the cereal-rye cover crop and included 1) a no cover-crop control (Referred to here as No CC) and cover crop terminations of: 2) 14-28 days pre-planting (DPP; Referred to here as 'Early terminated'), 3) 3-7 DPP (Referred to here as 'Late terminated'), and 4) 1-3 days after planting (DAP; Referred to here as 'Planting green') of the cash crop. Cover crops were terminated using glyphosate (1.27 kg ae ha⁻¹) and ammonium sulfate (2.5% v/v) using water as a carrier at 140 L ha⁻¹. Based on abiotic conditions, planting dates of cover and cash crops, and termination dates varied annually (Table 2-1). I established a 3 m border of cover crop around the experiment and 9 m alleys between blocks. The border and the alleys follow the planting-green schedule for termination and cash-crop planting (Fig. 2-1). Soybeans were sown on 76 cm row spacing.

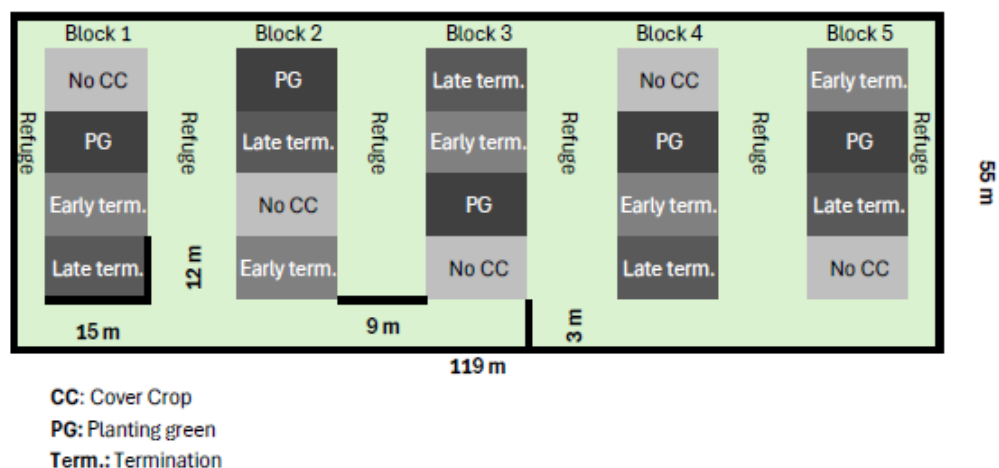


Figure 2-1. Schematic diagram of the design for the cover crop termination experiment, showing treatments, blocks, and buffer areas.

Table 2-1. Cover-crop planting date, seeding rate, termination timing, soybean planting date, and seeding rate from the cover-crop termination experiment.

Year	Cover-crop planting date	Cereal-rye seeding rate	Termination dates				Soybean planting	Soybean seeding rate
			No Cover Crop	Early	Late	Planting green		
2022	21 Oct. 2021	67.5 kg/ha	NA	29 April	17 May	1 June	31 May	450,000 seeds/ha
2023	10 Nov. 2022	67.5 kg/ha	NA	5 May	12 May	23 May	23 May	450,000 seeds/ha

To characterize influences of termination date on cover-crop productivity, I harvested aboveground biomass from each plot one to two days prior to termination. From four randomly positioned quarter meter quadrats per plot, I cut cover-crop biomass roughly 2.54 cm above the soil surface. After collection, samples were dried in an oven (70°C) for 5-7 days, and then weighed.

To understand the influence of cover-crop termination on foliage-dwelling predators, I sampled their communities with a 48-cm muslin sweep net. In 2022, I completed 10 sweeps covering approximately 7.5 m between rows 6-7 of each plot. A sweep was one pass of the net over an arc of approximately 180° with the net dipping into plant canopies over most of the arc. In 2023, to increase the

numbers of predators captured, I doubled my effort to two transects of 10 sweeps per plot from rows 3-4 and 11-12. To account for this difference, I report the number of predators per sweep. I collected samples around midday to avoid dew-covered foliage. I began sampling around growth stage V2-3 and continued until crop maturity (growth stage R3-4; final sampling date: 12 August 2022 and 26 July 2023).

For analyses, I aggregated arthropods by functional groups and several individual families. These included Araneomorphae (e.g., Linyphiidae), predatory Coleoptera (e.g., Cicindellidae), other Coleoptera (e.g., Chrysomelidae), predatory Hemiptera (e.g., Reduviidae), other Hemiptera (e.g., Auchenorrhyncha), Caelifera (e.g., Acrididae), Ensifera (e.g., Gryllidae), Syrphidae, Formicidae, and Hesperiiidae. For araneomorph analyses, I aggregated araneomorphs by family.

2.3. Augmentation experiment

During the 2023 growing season to quantify the influence of spiders in pest control of soybean, I established in each of six no-till fields a randomized block design that included three treatments that were each replicated twice and organized into two blocks, each with three plots (12 x 15 m; total of 36 plots across the six fields; Fig. 2-2). Planting dates ranged from 15-18 May 2023 and each field received 450,000 seeds/ ha with 76-cm row spacing. The three treatments differed in how I altered the predator and pest communities; they included: 1) depletion or 2) augmentation of communities, or 3) the communities remained unchanged (control). For the “depletion” treatment, I depleted arthropod communities by sampling pests and predators from canopies of all plants in each plot. I conducted this sampling with a sweep net (details above) swung in 180° arcs without stopping until I had covered the entire plot. I then randomly dumped the contents of the net (i.e., all the captured arthropods) into the “augmentation” plot in the same block, making a point to scatter arthropods in all portions of the plot. To control for the influence of walking through the plots and sweeping plants, I also disturbed control plots by “sweeping” the canopy of plants with the handle of a sweep net. I imposed these treatments when soybean plants were large enough to sweep (Growth stage range R1-R6; 11 August, 22 August, 4 September, and 22 September 2023) until soybean started to senesce (4 October 2023). On the last sampling date, sweep net contents were collected and brought back to the lab for identification and counting.

Upon returning arthropods to the lab, I categorized them into functional groups, including: Araneomorphae, pestiferous hemipterans (e.g., Aphidae, Cicadellidae, Pentatomidae), pestiferous coleopterans (e.g., Chrysomelidae), caeliferans (e.g., Acrididae), and all lepidopteran larvae (e.g., Hesperiiidae). I identified pestiferous and predatory Pentatomidae using, ‘Stinks Bugs of Oregon’

(Hedstrom, 2019) and spider families using ‘Spiders of North America, an Identification Manual’ (Ubick et al., 2017).

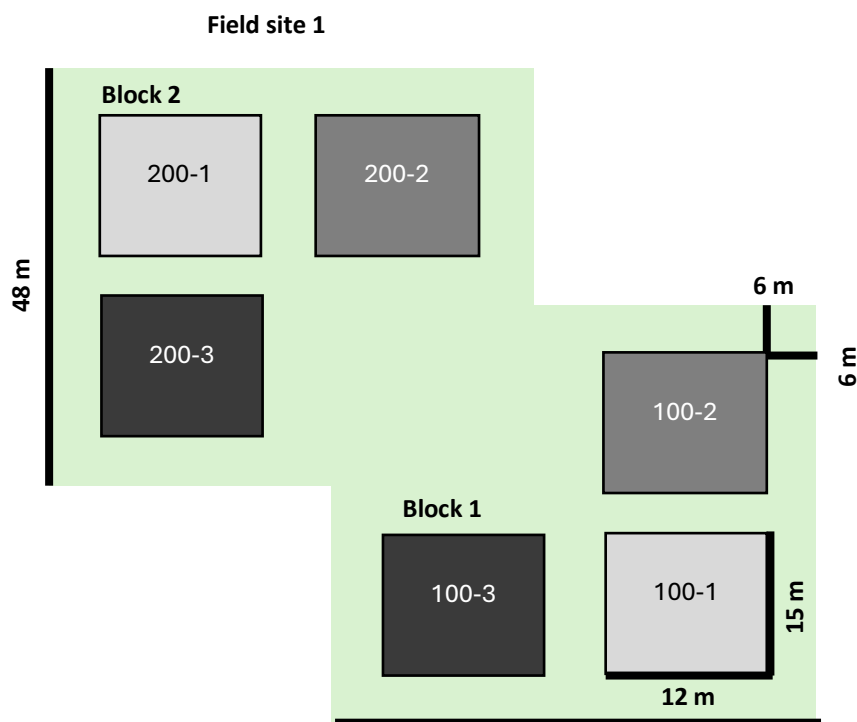


Figure 2-2. Schematic diagram of the design for the augmentation experiment. In each of six fields, I established two blocks each with one replicate of three treatments. Each plot had 6-m alleys on each side.

To assess the influence of depleting and augmenting arthropod communities on defoliation, I evaluated damage to soybean leaves from each plot. I randomly selected two plants from every fourth row within each plot (eight plants per plot). From the younger foliage of each plant, I collected five tri-lobate leaves (forty leaves per plot), totaling 120 leaflets per plot. To quantify defoliation from each treatment, I followed an established protocol (O’Neal et al., 2002). Briefly, I scanned each leaflet into a digital format using an Epson Perfection V39 scanner (Epson America, Los Alamitos, CA) with Scanner Driver and EPSON Scan Utility software (*EPSON Scane Utility*, n.d.). I then analyzed images with ImageJ software (Rasband, 1997). Methodology for this process was derived from O’Neal et al. (2002).

3. STATISTICAL ANALYSIS

I conducted all statistical analyses in R (version 4.3.1).

3.1 Cover-crop termination experiment

To analyze effects of timing of cover-crop termination, I ran a generalized linear mixed-effect models (GLMM) on the following data collection types: cover-crop biomass (Gaussian distribution), spider

populations (Negative binomial distribution), and pest populations (Negative binomial distribution). For cover-crop biomass, the fixed effect term was treatment*year to capture the main effect of each, but also their interaction. The random effect term for cover-crop biomass was block. For spider and pest populations, I chose a fixed effect of treatment*year and a random effect plot within block to account for non-independence ('lme4', Mächler et al., 2015). The data used for the spider and pest models was averaged across year and block to account for an imbalance in sampling dates.

To analyze relationships, I ran generalized linear models (GLM) on the following interactions: spider and pest (Negative binomial distribution); spider and date (Negative binomial distribution; Package: 'MASS', Venables & Ripley, 2002).

To compare the average populations of arthropod groups and spider families, I ran Dunn's Tests. To evaluate differences by treatment, I used analysis of variance (ANOVA) followed by Tukey HSD tests on the following data: spider populations and arthropod group populations.

3.2 Augmentation experiment

To analyze the effects of the control, depletion, and augmentation treatments on spider and pest populations, I ran generalized linear-mixed models (GLMM). Both distributions were negative binomial with a fixed effect term of treatment and a random effect term of field site. To compare the average populations of spider families and arthropod groups, I ran Dunn's Tests. For changes by year, sampling date, and treatment in spider and pest populations, I ran non-parametric multivariate statistical permutation tests (PERMANOVA) using the 'vegan' package (Oksanen et al., 2022). Because I analyzed count data, I used Bray-Curtis dissimilarity and set permutations to factorial 10 (3628800). Due to the smaller size of the data set and my available computational power, I chose a higher permutation count to ensure accuracy of the F statistic. To evaluate differences in soybean leaf damage by treatment, I used analysis of variance (ANOVA) followed by Tukey HSD tests.

For linear models from both experiments, to determine the significance of fixed main effects and interactions, the "anova" function was used to obtain log-likelihood ratio tests and the Wald X^2 test statistic. The anova output allowed a comparison of null models to fully-fitted models, adding one effect at a time. I ran a post hoc analyses with the 'emmeans' package (Lenth, 2023) for linear models.

4. RESULTS

4.1. Cover-crop termination experiment

4.1.1. Cover-crop biomass

All cover-crop treatments differed in 2022 and in 2023 the early-terminated treatment had significantly lower biomass than both late-terminated and planting-green treatments (Fig. 2-3).

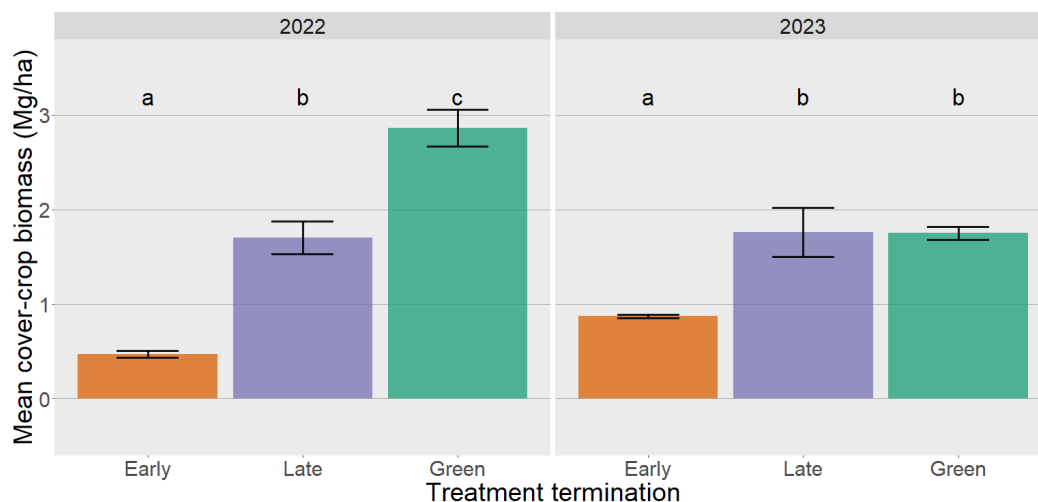


Figure 2-3. Cover-crop biomass from soybean plots split by treatment and year. In 2022, all cover-crop treatments differed. In 2023, early-terminated was lower than the late-terminated and planting-green treatments, but late-terminated and planting-green treatments did not differ. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

4.1.2. Spider and pest populations

There was a significant main effect of year for spider populations ($df=1$, $X^2=25.3$, $P\text{-value}<0.001$) and pest populations ($df=1$, $X^2=12.8$, $P\text{-value}<0.001$). In both instances, populations of pests and spiders were greater in 2022 than in 2023.

Of all the groups analyzed, I recorded a difference between Hemiptera and araneomorph ($P=0.03$; Fig. 2-4: A). Tetragnathid abundance was greater in the no cover crop ($P=0.04$) than the early-terminated treatment and marginally more abundant in no-cover-crop ($P=0.064$) than the planting-green treatment (Table 2-S1). I recorded a significant difference between Mysmenidae and Thomisidae populations ($P=0.048$; Fig. 2-4: B).

In the PERMANOVA, Araneomorph abundance was correlated to both year ($F_{1,44} < 0.001$) and sampling date ($F_{2,44} \leq 0.001$). When evaluating the relationship between araneomorph abundance and sampling date, abundance of araneomorph taxa was greater at the second sample date, but only significantly greater in 2022 (Fig. 5).

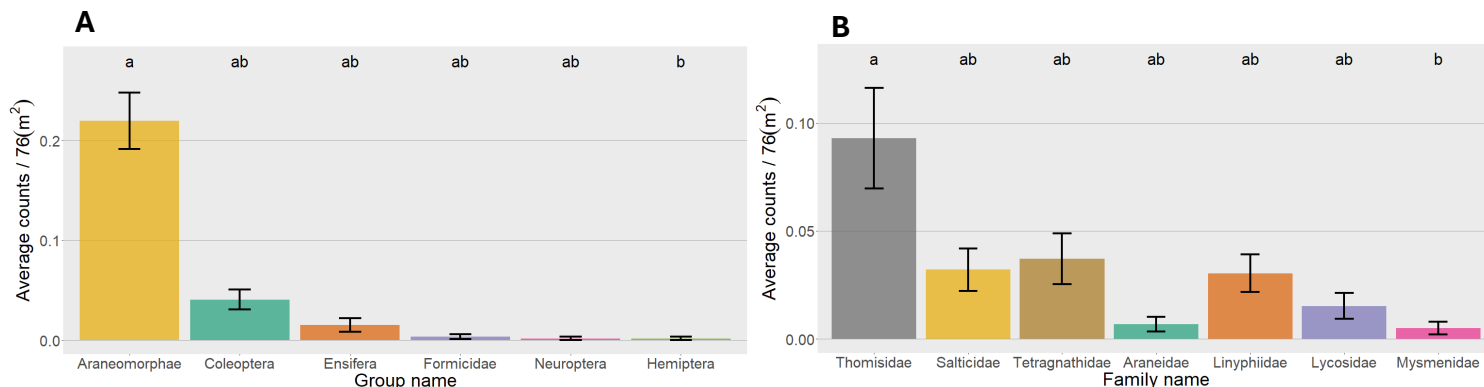


Figure 2-4. A. Abundance of spiders and insects collected in soybean established after different termination timings of a cereal-rye cover crop from the cover-crop termination experiment. Statistical differences were derived from a Dunn's Test to analyze the mean values of each group population. Letter change denotes significance. Differences derived from Dunn's output when $P \leq \alpha / 2$. Araneomorph populations were the highest among all groups, but only statistically higher than Hemiptera.

Figure 2-4. B. Abundance of araneomorph families collected in soybean established at different termination timings of cereal-rye cover crop from the cover-crop termination experiment. Statistical differences were derived from a Dunn's Test to analyze the mean values of each family population. Letter change denotes significance. Differences derived from Dunn's output when $P \leq \alpha / 2$. Thomisidae had the highest populations among all spider families sampled but was only statistically higher than Mysmenidae.

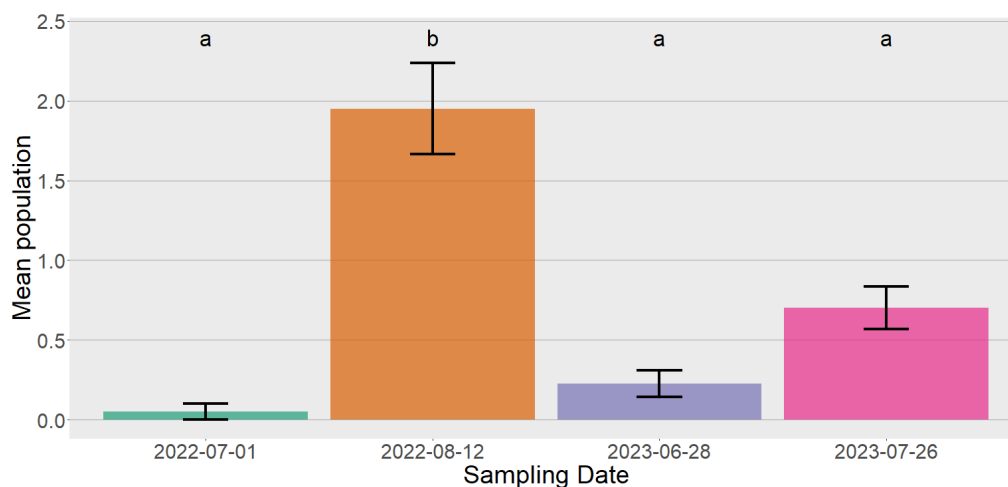


Figure 2-5. Abundance of total spider populations per plot by sampling date in soybean established after different termination timings of a cereal-rye cover crop from the cover-crop termination experiment. Statistical differences derived from a generalized linear model. Letter change denotes significance. Sampling date 12 August 2022 had significantly higher populations than the other three sampling dates.

4.2. Augmentation experiment

4.2.1. Spider and pest populations

There was a significant main effect of treatment for both spider ($df=2$, $X^2=20.5$, $P\text{-value}<0.001$) and pest populations ($df=2$, $X^2=14.5$, $P\text{-value}<0.001$; 2-S1, S2). In both cases, the depletion treatment had lower populations than the control and augmentation treatments. The control and augmentation treatments did not differ.

Manipulations successfully increased the abundance of four families (Oxyopidae, Salticidae, Tetragnathidae, and Thomisidae) with all four being significantly more abundant in augmented plots compared to depleted plots. For oxyopid spiders, augmented and control plots had higher populations than depleted plots (Fig. 2-6). Abundance of other araneomorph families were lower and similar to each other (Fig. 2-7).

In the PERMANOVA, communities differed significantly among treatments ($F_{2,32}<0.001$; Fig. 2-8). The depletion treatment had significantly lower populations of araneomorph spiders and pests than both the control and the augmentation ($P<0.05$; Fig. S1, S2). Across treatments, abundance of araneomorph spiders was positively correlated with pest abundance (Fig. 2-9).

4.2.2. Soybean damage

Plant defoliation was equal among treatments (Fig. 2-S3).

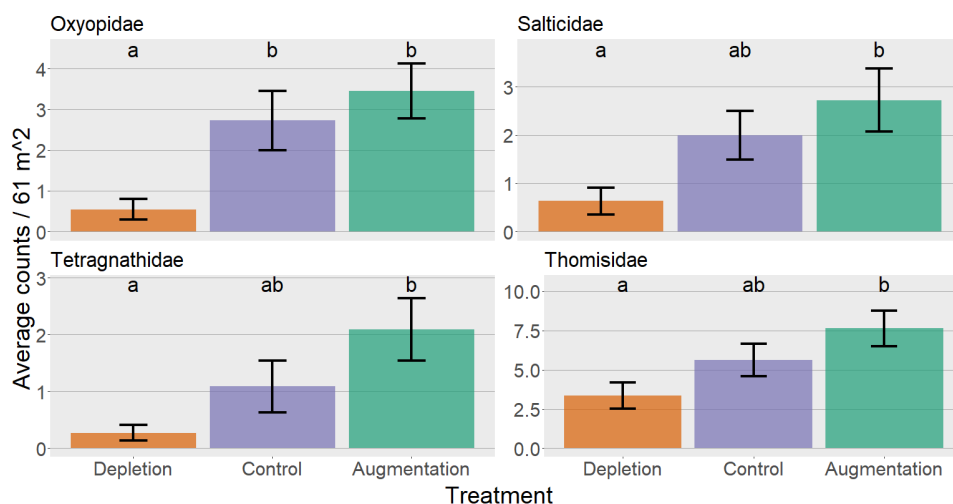


Figure 2-6. Abundance counts of the spider four families that had significant differences in population among treatment from the augmentation experiment. They include Oxyopidae, Salticidae, Tetragnathidae, and Thomisidae. Statistical differences derived from an ANOVA of spider family by treatment. Letter change denotes significance. For Salticidae, Tetragnathidae, and Thomisidae, the depletion populations were lower than the augmentation, but the control did not differ from either. For Oxyopidae, both the control and augmentation treatments differed from the depletion.

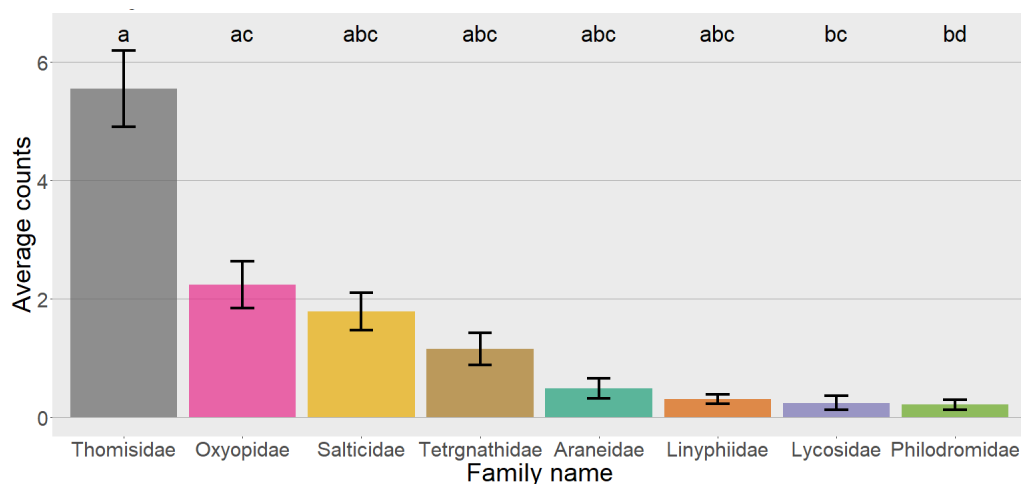


Figure 2-7. Abundance counts of araneomorph family populations from the augmentation experiment. Statistical differences derived from a Dunn's Test of mean family populations. Letter change denotes significance. Differences derived from Dunn's output when $P \leq \alpha / 2$. Thomisidae had the highest overall populations but was only significantly higher than Lycosidae and Philodromidae. Oxyopidae had significantly higher populations than Philodromidae.

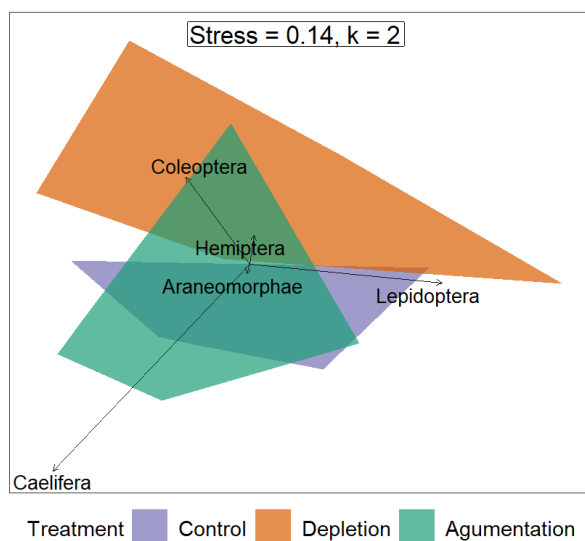


Figure 2-8. NMDS plot displaying the distribution of arthropod groups from the augmentation experiment. Each polygon represents a treatment. Overlap of the polygon denotes similarity among populations. Arrow direction and length refers to each arthropod group's distribution among the treatments. For example, Caelifera appears to correlate with the control and augmentation treatments, but not depletion treatment. The stress level was 0.14 and dimensions were set to $k = 2$.

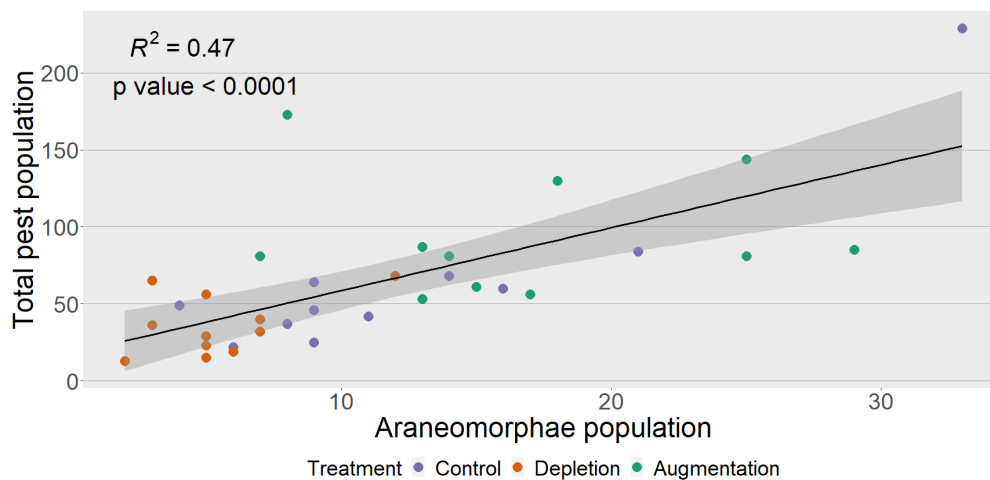


Figure 2-9. Linear regression of total pest by total araneomorph populations from the augmentation experiment. Statistical values were derived from a generalized linear model. Recorded an increase in pest populations with an increase in spider populations. $R^2 = 0.47$ and $P < 0.001$.

5. DISCUSSION

The aim of this study was to determine the most abundant generalist predators of Pennsylvanian soybean systems and determine if, and how much, spiders are contributing to pest control in these systems. The results do not entirely corroborate my hypotheses of higher cover-crop biomass resulting in higher spider populations.

Spiders were the most abundant predators in the cover-crop termination experiment (Fig. 2-3A), and thomisids were the most abundant spider family. Of the spiders I collected, the three most abundant families –Thomisidae, Oxyopidae, and Salticidae– are mobile or ambush hunters. The hunting and cursorial movement strategies (Nyffeler, 1999) of these spiders could be the reason for their colonizing of the field. For example, if web building spiders balloon into a field (Schmidt et al., 2008) with inadequate architecture or prey, they may balloon again. Whereas spiders that rely on cursorial movement may not be able to balloon, or balloon as far.

Notably, spider abundance did not increase with a later-terminated-cover crop. While increasing non-crop habitat can increase abundance of natural enemies (Finke & Denno, 2002; Landis et al., 2000; Tonhasca Jr., 1993), I saw no residual effects of increasing cover-crop biomass in the following soybean crop. The unexpected result of not increasing spiders makes sense because while the late-terminated and

planting-green treatments had higher biomass pre-cash-crop planting, there was no increase in habitat throughout the season. Because biomass and habitat complexity was greater in the later-terminated treatments, perhaps ground-dwelling predators were influenced (Finke & Denno, 2002) but foliage-dwelling predators were not. Furthermore, a lack of continuous vegetation from cover-crop termination may have influenced spider populations by temporally decrease suitable habitat for spiders (e.g. Nyffeler et al., 1994). When the cover crop died, spiders did not have enough resources for their communities to persist locally until the soybeans started to grow well. It is also possible that spiders did not colonize the cereal-rye cover crop prior to termination and soybean planting. While there is potential for toxicological effects of herbicides on spider, it is unlikely that the glyphosate herbicide application played a role in the depletion of spider populations (Schmidt-Jeffris et al., 2022). Spider abundances were greater later in the season in 2022, and although the trend was not significant in 2023, populations were still higher later in the season (Fig. 2-4), suggesting that abundance of foliage-dwelling spiders increases over the growing season.

In the augmentation experiment, I successfully manipulated spider populations (Fig. 2-7). Four spider families had higher populations in augmented plots than depleted ones (Fig. 2-5), with thomisids being the most abundant family. Despite increasing abundance of some spider taxa, neither pest abundance nor their defoliation was lower in augmented plots (Fig. 2-7). Absence of the expect relationship could be explained because augmentative strategies seldom work when pest populations are not high (Riechert & Lockley, 1984). This hypothesis is further supported by a lack of statistical differences in leaf damage among the three treatments (Fig. 2-S3). It is also possible that I saw no changes in amounts of defoliation because I sampled leaves too late in the season, so there was little opportunity for spiders to make a difference.

While I was unable to augment spider populations by increasing early-season biomass via cover crops, previous research suggests that it is feasible. For example, increasing non-cash-crop plants can lead to higher spider populations (Schmidt et al., 2008). By increasing in-season non-cash-crop plants, like planting cover crops between cash crop rows (intercropping; Brooker et al., 2015), there may be more suitable habitats throughout the season for spiders. Nevertheless, cover crops are an important resources for promoting ground-dwelling predators (Inveninato Carmona et al., 2021; Landis et al., 2000), but implementing another means—like intercropping—of early-season habitat could intercept and harbor spider populations until the cash crop has created adequate in-field habitat.

Further research on promoting spider populations in early-season soybean production is advisable. If spider populations can be bolstered throughout the season, rather than only later in the season, there may be a reduction in early-season pest incidence.

6. CONCLUSION

My results revealed that spiders are the most abundant generalist predators in soybean grown in central Pennsylvania. Additionally, time of year was a key driver of populations, which is likely in response to habitat availability (Finke & Denno, 2002; Landis et al., 2000; Schmidt et al., 2008). Of the spiders I collected, the most dominant families were ambush or mobile hunters, rather than web builders, which makes sense given the disruptive nature and crop phenology of the annual growing season (Landis et al., 2000; Prieto-Benítez & Méndez, 2011). The cover-crop termination experiment illustrates that spiders are not supported throughout the season in annual cropping systems and farming and pest control practices could be adapted with spiders in mind, which can be done by increasing non-cash-crop habitats throughout the season (Schmidt et al., 2008). This study is integral to continued research on bolstering spider populations in annual systems because I recorded that spiders are the most abundant predators and that cover cropping alone is not enough to increase the diversity of foliage-dwelling invertebrate fauna in modern soybean systems. More research is necessary to understand how to support spider populations within annual cropping systems.

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Chapter 3

Assessing Biological Quality of Soils Using Soil Invertebrates

1. INTRODUCTION

Soil quality is critical to crop production and maintenance of environmental quality (Doran & Parkin, 1994; USDA, 2015). Environmental quality can be improved by soils in many ways (e.g., water retention), but acting as a habitat for soil-dwelling fauna is of particular importance (Blum, 2005; USDA, 2015). This is because higher richness of soil-dwelling fauna can lead to increases in soil functionality in process like nutrient cycling (Kampichler & Bruckner, 2009; Lavelle et al., 2006; Pulleman et al., 2012). To support biological diversity and improve environmental quality, soils must be high quality and able to perform ecosystem functions.

Soil quality refers to measurable attributes that relate to the capacity of soil to function and interact positively (e.g., filter waste) with the environment (Idowu et al., 2009). Understanding the role agricultural practices have on the quality of soil ecosystems is imperative with a global rise in population and agricultural demand. The condition of agroecosystems and the soils within determine the sustainability of our agricultural systems, quality of the environment, and directly correlate to plant, animal, and human health (Doran & Parkin, 1997). For example, without high quality soils, food production would be nearly impossible. Furthermore, understanding how different agricultural practices affect soil quality can allow for more informed decision making by growers looking for ways to improve soil and environmental quality. For these reasons, when considering agroecosystem quality, soil quality should be considered (Doran & Parkin, 1997; Glanz, 1995).

Researchers evaluate soils for two reasons: diagnosis and prognosis (Peverill et al., 1999). When considering agricultural land, the former is used to identify constraints of a soil to produce specific crops and achieve desired yields (Peverill et al., 1999). Current quantitative evaluations of soil quality use multiple chemical, physical, and biological attributes (Morrow et al., 2016). Readily available and commonly used tests include the Cornell Soil Health Tool (CSHT; Idowu et al., 2009), the Soil Management Assessment Framework (SMAF; Andrews et al., 2004), the Agroecosystems Performance Assessment Tool (AEPAT; Wienhold et al., 2006), and the NRCS-USDA soil health assessment tool (Seybold et al., 2002). These tests measure soil metrics such as carbon and nitrogen cycling and bulk density which can be used to inform farmers of the effects of their management practices on soil quality. Presently, there are two common tests for soil biological quality which include the Autoclaved Citrate Extractable (ACE) Protein Index (Soil Protein) (van Es et al., 2020) and the respiration of the metabolic

activity of the soil microbial community (van Es et al., 2017). These tests, while insightful on microbial activity within the soil, are indirect measures of soil biological quality. What all afore-mentioned tests lack are direct links to the biological quality of soils. Monitoring the biological quality of soils within agricultural systems can provide insight into the sustainability of crop-management practices. The need to understand if crop-management practices are improving soil-ecosystem quality within a field has emerged with an increased awareness of ecosystem health and the associated risks of different land uses on the environment (Doran & Parkin, 1994; Suter, 2007).

A useful approach to assessing ecosystem quality is through bioindicators. Bioindicators can be organisms whose habitat preference or abundance help reveal different qualities of an ecosystem (e.g., pollution; Adams et al., 2001; Bartell, 2006). Due to the complex nature of most ecosystems, it is common that a suite of indicators has been identified for some ecotypes (Burger, 2006). For example, in streams, researchers will often collect macroinvertebrates and count abundance of insects in the orders Ephemeroptera, Plecoptera, and Trichoptera, taxa that are good indicators of high water quality (Barbour et al., 1999).

Soil invertebrates, such as mites and collembolans, are diverse and can be used to evaluate the influence of agricultural practices (Cao et al., 2011). Estimates suggest that edaphic (soil-living) animals compose 23% of all described animal species, of which 80% are insects, 12% are arachnids, and 5% are other invertebrates (Decaëns et al., 2006; Wolters, 2001). These soil-dwelling invertebrates live in almost all ecosystems (Giller, 1996) and have a large contribution to ecosystem services such as decomposition and cycling of nutrients and organic matter (Kampichler & Bruckner, 2009; Lavelle et al., 2006; Puleman et al., 2012; Wolters, 2000). Arthropods within this broad group of soil invertebrates can have distinct morphological adaptations to different soil qualities and levels of disturbances (Menta et al., 2020). Due to the specialization of edaphic invertebrates, some groups are highly sensitive to changes in soil conditions, making them excellent bioindicators (Parisi et al., 2005).

Originating in Italy, the Biological Soil Quality index-arthropod (QBS-ar) is a reliable method using edaphic soil invertebrates to assess the biological quality of an ecosystem (Parisi et al., 2005). This method –which is based on scoring groups of soil invertebrates with the eco-morphological index (EMI)– can be used to evaluate invertebrate groups based on their adaptations to the soil environment, and does not require researchers to identify specimens to species level (Testi et al., 2012). Each apparent taxon or morphotype receives a score associated with specializations to edaphic life, such as reduction or loss of pigmentation, reduced appendages, and reduction or loss of flying (Parisi et al., 2005; Testi et al., 2012). This method of evaluating biological quality of an ecosystem has been used to investigate multiple ecosystem types, including cropland (Parisi et al., 2005). For example, studies have used this approach to

reveal that high levels of disturbance (e.g., herbicide, insecticide, and fertilizer applications, tillage, and soil type) can alter populations of soil invertebrates (Boutin et al., 2009; Cortet et al., 2002; Diekötter et al., 2010; Fiorini et al., 2020; House & Parmelee, 1985), which corroborate their use in assessing the biological quality of soils (Parisi et al., 2005).

In this chapter, I used the QBS-ar index to investigate the influence of cover crop use and a suite of crop-management practices on the biological quality of fields used to grow field crops. In two complementary experiments in Pennsylvania, I asked three questions: 1) How does a cover crop of cereal-rye (*Secale cereal* L.) affect relative abundance of soil invertebrates and QBS-ar scores? 2) Does timing of cover-crop termination affect relative abundance of soil invertebrates and QBS-ar scores? 3) How do legacy effects of production practices associated with corn affect relative abundance of soil invertebrates and QBS-ar scores? I hypothesized that compared to a no-cover-crop treatment soil-invertebrate abundance and QBS-ar scores would be higher *i*) in cash crops planted into living cover crops, *ii*) in late-terminated cover crops, and *iii*) in agricultural fields with fewer disturbance events (e.g., herbicide applications, tillage, synthetic fertilizing).

2. MATERIALS AND METHODS

2.1. Experiments

This project comprises two experiments. The first explored the influence of the timing of cover-crop termination on soil invertebrates in subsequent corn and soybean crops (referred to here as ‘Cover-crop experiment’). The second explored soil invertebrates in Rodale’s Farming Systems Trial (Referred to here as ‘Rodale experiment; FST; *Farming Systems Trial: 40-Year Report*, n.d.).

2.2. Cover-crop experiment

2.2.1. Study location and experimental design

To understand the influence of delayed cover-crop termination on soil invertebrate communities in Pennsylvania crop fields, I conducted a three-year field experiment (2021-2023) at Penn State’s Russell E. Larson Agricultural Research Center (Pennsylvania Furnace, Pennsylvania, U.S.A.; 40.720064, -77.934317). In a long-term no-till field (>10 yr.), I established a single-factor experiment, arranged in a randomized complete block design with four cover crop treatment levels and five replicates. Plot size was 12 x 15 m. Cover crop treatments varied by termination timing of the cereal-rye cover crop and included 1) a no cover-crop control (Referred to here as No CC) and cover crop terminations of: 2) 14-28 days pre-planting (DPP; Referred to here as ‘Early terminated’), 3) 3-7 DPP (Referred to here as ‘Late terminated’), and 4) 1-3 days after planting (DAP; Referred to here as ‘Planting green’) of the cash crop. Cover crops were terminated using glyphosate (1.27 kg ae ha⁻¹) and ammonium sulfate (2.5% v/v) using

water as a carrier at 140 L ha^{-1} . Based on abiotic conditions, planting dates of cover and cash crops, and termination dates varied annually (Table 3-1; For soil type information, see Table 3-1a). I established a 3 m border of cover crop around the experiment and 9 m alleys between blocks. The border and the alleys follow the planting-green schedule for termination and cash-crop planting (Fig. 3-1). Soybeans were sown on 76 cm row spacing.

To evaluate the legacy effects associated with a corn-soybean sequence, I followed my 2021 corn with soybean in 2022 as well as 2022 corn followed by soybean in 2023. In both instances, I overlaid the same treatments as the previous year.

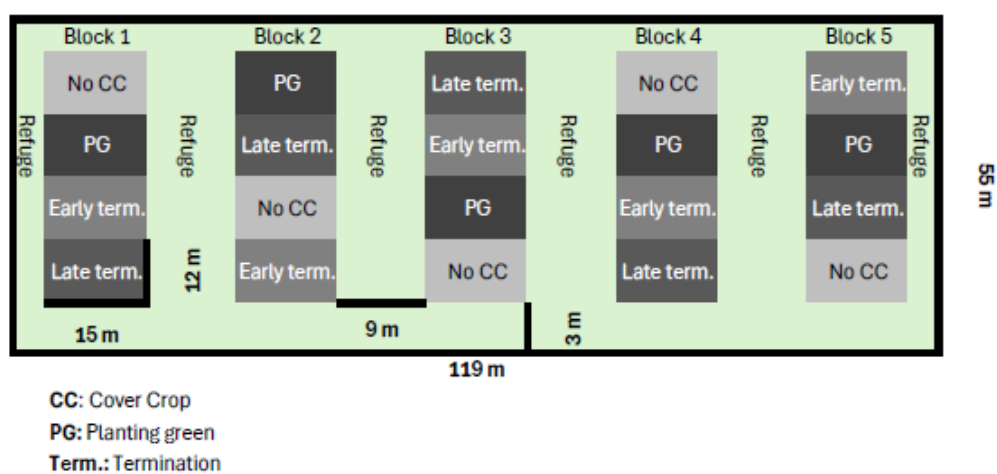


Figure 3-1. Schematic diagram of the design for the cover crop assay experiment, showing treatments, blocks, and buffer areas from the cover-crop experiment.

Table 3-1. Cover-crop and cash-crop planting and termination dates and seeding rates from the cover-crop experiment.

Crop year	Corn field cover-crop planting date	Soybean field cover-crop planting date	Cereal rye seeding rate (kg/ha)	Treatment termination			Planting green	Corn planting date	Corn seeding rate (seeds/ha)	Soybean planting date	Soybean seeding rate (seeds/ha)
				No Cover Crop	Early	Late					
2021	8 Sept. 2020	-	67.5	-	27 April	12 May	24 May	21 May	81,250	-	-
2022	10 Nov. 2021	21 Oct. 2021	67.5	-	29 April	17 May	1 June	31 May	81,250	31 May	450,000
2023	26 Oct. 2022	10 Nov. 2022	67.5	-	5 May	12 May	23 May	23 May	81,250	23 May	450,000

Table 3-1a. Soil type information for the three field sites and the associated crops and year from the cover-crop experiment. All soil data retrieved from the Web Soil Survey (Soil Survey Staff, n.d.).

Field number	Crop	Year	Crop	Year	Soil Type	Slope	%	Slope	%
1	Corn	2021	Soybean	2022	Hagerstown silt loam	0 to 3	66.7	3 to 8	33.3
2	Corn	2022	Soybean	2023	Murrill channery silt loam	0 to 3	100	-	-
3	Corn	2023	-	-	Murrill channery silt loam	0 to 3	100	-	-

2.2.2. In-field sampling

To characterize influences of termination date on cover-crop productivity, I harvested aboveground biomass from each plot one to two days prior to termination. From four randomly positioned quarter meter quadrats per plot, I cut cover-crop biomass roughly 2.54 cm above the soil surface. After collection, samples were dried in an oven (70°C) for 5-7 days, and then weighed.

To evaluate soil-invertebrate abundance and QBS-ar scores in a cover-crop termination gradient, I collected four soil cores of 23 cm² from each plot at roughly 5 m intervals starting from the western edge of each plot. Soil sampling was specifically targeting soil invertebrates and done spatially to capture variations throughout the plot. I placed the soil from each plot into one plastic bag and transported the samples back to the lab for extraction. This sampling regime was done on 1 July 2021, 1 September 2021, 22 June 2022, 23 September 2022, 18 July 2023, and 4 November 2023. These dates capture seasonal trends in invertebrate abundance (Jernigan et al., 2020).

2.3. Rodale experiment

2.3.1. Study location and experimental design

This study was conducted during one field season in 2023 in the Farming Systems Trial at the Rodale Institute (Kutztown, Pennsylvania, U.S.A. 40.550467, -75.722164).

The Farming Systems Trial (FST) –started in 1981– was designed to address the barriers to the adoption of organic farming by farmers (*Farming Systems Trial: 40-Year Report*, n.d.). Located on a 6.1 ha field, the soil is well drained Comly silt loam with small areas of Berks shaley silt loam and Duffield silt loam (Ryan, et al. 2009). This trial compares three core systems, which include conventional synthetic (CNV), organic legume (LEG), and organic manure (MNR). The focus of this trial is on corn and soybean production because 70% of the U.S. acreage is devoted to growing grain (*Farming Systems Trial: 40-Year Report*, n.d.).

FST is a split-plot randomized complete block with eight replicates. Four treatments are represented in eight blocks with three main plots split into nine sub plots (Fig. 3-2). Main plot systems comprise one CNV and two organic systems (MNR and LEG), which measure 18 x 92 m with 1.5 m strips of grass between.

For this study, I sampled subplots producing corn. All four treatments were represented in the thirty-two subplots producing corn, including organic manure (MNR), organic legume (LEG), conventional without cover crop (CWC), and conventional with cover crop (CCC).

Along with the treatments, there was an additional tillage component. Two tillage regimes were implemented, comprising till and no-till. Of the thirty-two subplots, sixteen were subject to till and the other sixteen to no-till. Subplot dimensions are 6 x 92 m.

All field operations including tillage, spraying, mowing, and seeding were completed by Rodale Institute staff (Table 3-2). Based on abiotic conditions, planting dates of cover and cash crops, and termination dates varied annually (Table 3-2a). Insufficient cover-crop data were taken to conduct any statistical analyses.

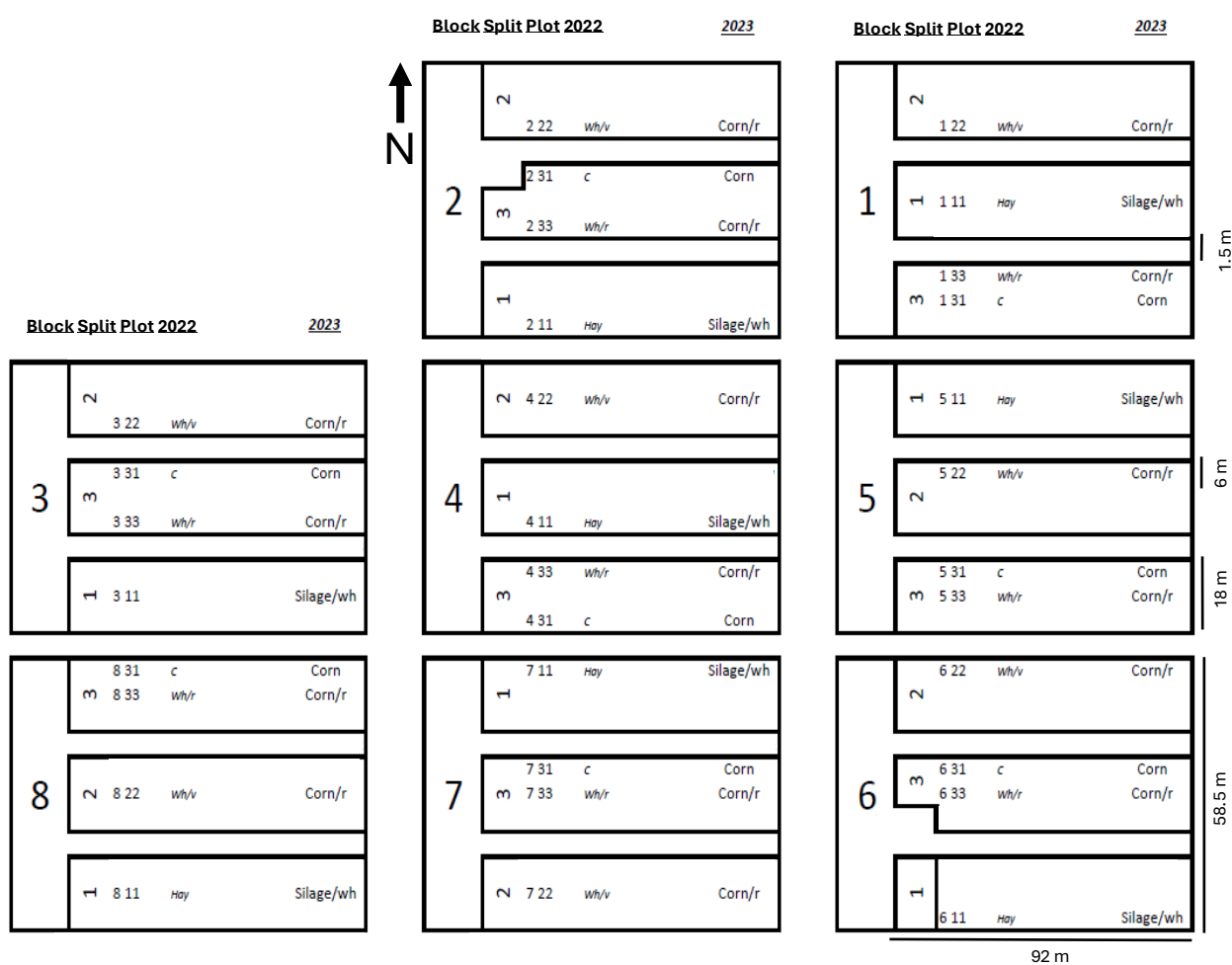


Figure 3-2. Schematic of the Farming Systems Trial for the Rodale experiment, showing corn plots, treatments (2022 and 2023), blocks, split plots, sub plots, and buffer areas.

2.3.2. In-field sampling

I collected six soil cores of 23 cm² from each subplot at roughly 15 m intervals starting from the eastern edge of each plot. Soil sampling was specifically targeting soil invertebrates and done spatially to capture variations throughout the plot. I placed the soil from each subplot into one plastic bag and transported the samples back to the lab for extraction. I took one soil moisture and temperature reading from each plot.

This sampling regime was carried out on two dates: 28 July 2023 and 11 October 2023. These dates capture seasonal trends in invertebrate abundance (Jernigan et al., 2020).

Table 3-2. Cover-crop planting, seeding rate, corn seeding rate, corn variety, and till and no-till planting dates from the Rodale experiment.

MNR: Organic manure

LEG: Organic legume

CWC: Conventional without cover crop

CCC: Conventional with cover crop

Treatment	Cover crop planting date	Cover crop	Seeding rate (kg/ha)	Corn seeding rate (seeds/ha)	Variety	Tillage planting date	No-till corn planting date	Harvest date
MNR	-	-	-	87,500	0.48-08PGS: Local Seed Co.	24 May 2023	24 May 2023	5 Oct. 2023
LEG	9 Sept. 2022	Purple Bounty Vetch	39	87,500	51T59: Albert Lea Seed	2 June 2023	5 June 2023	9 Nov. 2023
CWC	-	-	-	87,500	LC0297 SSXRIB: Local Seed Co.	10 May 2023	10 May 2023	9 Nov. 2023
CCC	16 Sept. 2022	Cereal Rye	189	87,500	LC0297 SSXRIB: Local Seed Co.	10 May 2023	10 May 2023	9 Nov. 2023

Table 3-2a. Cover-crop termination timings, termination methods, and fertilizer applications from the Rodale experiment.

CCC: Conventional cover crop

CNC: Conventional without cover crop

CC: Cover crop

Treatment	Conventional cover crop				Conventional without cover crop	
Date	9 April 2023	10 May 2023	14 May 2023	23 June 2023	14 May 2023	23 June 2023
Tillage	No-till	Tilled and No-till	Tilled	Tilled and No-till	Tilled	Tilled and No-till
Purpose	CC burndown	Fertilizer with planting	Pre-emergent	Side dress N	Pre-emergent	Side dress N
Product (L/ha)	Roundup PowerMax (1.6)	-	Lumax EZ (6.4)	-	Lumax EZ (6.4)	-
Product (L/ha)	2,4-D LVE (2.25)	-	Atrazine (2.25)	-	Atrazine (2.25)	-
Product (L/ha)	Lumax EZ (6.4)	-	-	-	-	-
Product (L/ha)	Infantry (2.25)	-	-	-	-	-
Product (kg/ha)	AMS (2.25)	UREA: 46-0-0 (73)	-	UAN (169)	-	-
Product (kg/ha)	-	Potash: 0-0-60 (169)	-	-	-	-

2.4. Soil invertebrate extractions and scoring

In the lab, I subsampled the collected soil from both experiments using an 8 x 11 cm cup (88 cm²), obtained a wet weight, and placed each sample on a Berlese-Tullgren funnel to extract all arthropods. Edaphic soil invertebrates were extracted following the methodology described in Parisi, et al. (2005). Upon removal from the funnel, I obtained a dry weight of the soil. This method was used to collect relative soil moisture (Reynolds, 1970). Scoring was complete following the methodology described in

Lisa, et al. (2022). Scores ranged from 1-20, with higher scores attributed to fauna with more specialized adaptations to soil life (e.g., reduction or loss of pigmentation, reduced appendages, reduction, or loss of flying and jumping appendages, etc.; Lisa et al., 2022; Parisi et al., 2005). Total scores per system are based on richness of the sample. For example, if there are five soil-invertebrates collected with score of 1, 5, 5, 10, and 20, the total score for this system is 41. The threshold score that determines a system with high biological quality is 93.7, where any score above this value is considered to be of high biological quality (Menta et al., 2018).

3. STATISTICAL ANALYSIS

I conducted all statistical analyses in R (version 4.3.1).

3.1. Cover-crop experiment

To analyze soil invertebrates, I ran a generalized linear mixed-effect model (GLMM) on the following data types: invertebrate abundance and QBS-ar score. Both models had a negative binomial distribution and a fixed effect term of treatment*crop to capture the main effect of each as well as their interaction. The random effect structure was plot nested within block to account for non-independence of the samples. Additionally, crop was used as the random term to account for repeated measures. Models were conducted using the ‘lme4’ package (Package: Mächler et al., 2015).

To analyze relationships, I ran generalized linear models (GLM) on the following interactions: cover-crop biomass and QBS-ar scores; cover-crop biomass and soil-invertebrate abundance; yield and QBS scores; yield and soil-invertebrate abundance.

3.2. Rodale experiment

To analyze soil invertebrates, I ran a generalized linear mixed-effect model (GLMM) on the following data types: invertebrate abundance and QBS-ar score. Both models had a negative binomial distribution and a fixed effect term of treatment*tillage*sampling date to capture the main effect of each as well as their interaction. To account for non-independence, the random effect term was plot.

For linear models from both experiments, to determine the significance of fixed main effects and interactions, the “anova” function was used to obtain log-likelihood ratio tests and the Wald X^2 test statistic. The anova output allowed a comparison of null models to fully-fitted models, adding one effect at a time. I ran a post hoc analyses with the ‘emmeans’ package (Lenth, 2023) for linear models.

4. RESULTS

4.1. Cover-crop experiment

4.1.1. Cover-crop biomass

In corn, I found differences among all treatments except between late-terminated and planting-green treatments in 2023 (Fig. 3-3). It is important to note the cover crop planting dates differed (Table 3-1).

In soybean, all cover-crop treatments differed in 2022 and in 2023 the early-terminated treatment had significantly lower biomass than both late-terminated and planting-green treatments (Fig. 3-4).

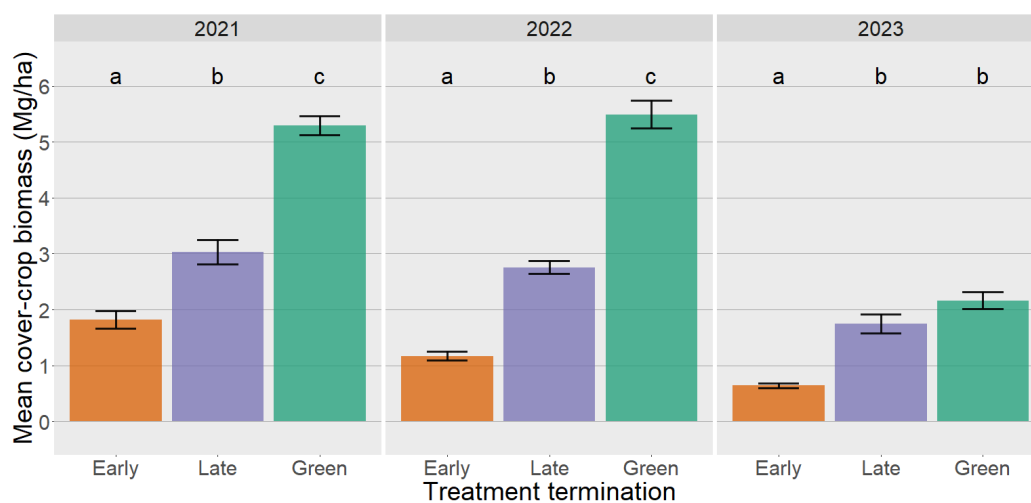


Figure 3-3. Cover-crop biomass from corn plots split by treatment and year. In 2021 and 2022, all cover-crop treatments differed. In 2023, early-terminated was lower than the late-terminated and planting-green treatments, but late-terminated and planting-green treatments did not differ. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

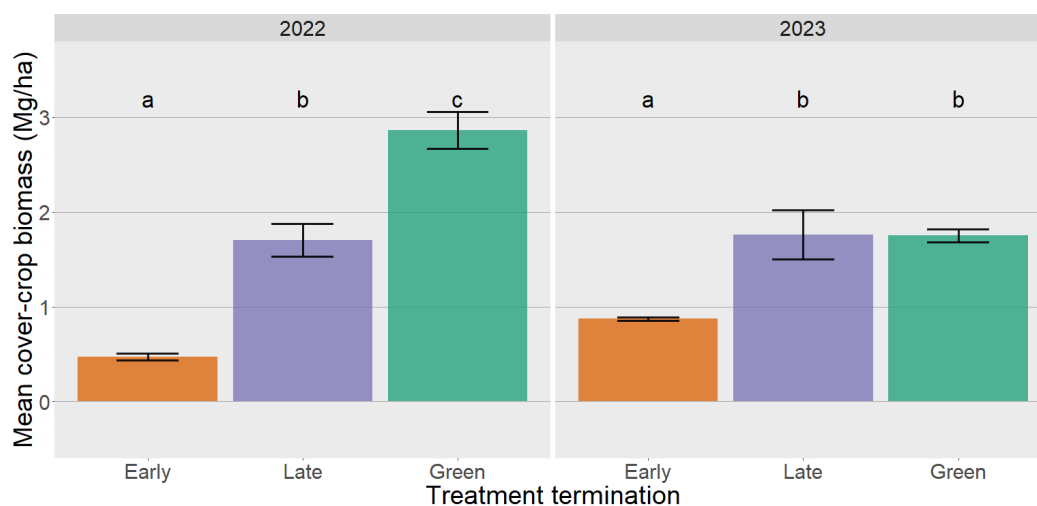


Figure 3-4. Cover-crop biomass from soybean plots split by treatment and year. In 2022, all cover-crop treatments differed. In 2023, early-terminated was lower than the late-terminated and planting-green treatments, but late-terminated and planting-green treatments did not differ. Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

4.1.2 Abundance and QBS-ar scores

The total soil-invertebrate count for this experiment was 3709. The most abundant groups were Acari (52%), Collembola (29%), Annelids (5%), other larvae (3.6%), Coleoptera larvae (2%), Chilopoda (1.4%), Diptera (1.4%), and Formicidae (1.2%). See table 3-6 for the remaining taxa collected.

I recorded no differences in abundance counts or QBS-ar scores among treatments or between crops. See Table 3-3 for QBS-ar scores by treatment and crop.

4.1.3 Interactions

I recorded no relationship between cover-crop biomass or yield and soil-invertebrate abundance or QBS scores in either corn or soybean.

Table 3-3. Average QBS-ar scores per 88 cm² sample by treatment and crop from the cover-crop experiment.

Recorded no differences in scores by treatment or by crop.

CLD: Compact letter display; Letter change denotes significance.

N = 5

Treatment	Crop	Mean ± SE	CLD
No CC	Soybean	51 ± 5.4	a
No CC	Corn	44.6 ± 4.1	a
Early terminated	Soybean	48.5 ± 6.3	a
Early terminated	Corn	43.6 ± 3.9	a
Late terminated	Soybean	50.4 ± 6	a
Late terminated	Corn	50.9 ± 4.5	a
Planting green	Soybean	49.4 ± 6.6	a
Planting green	Corn	53.2 ± 5.3	a

4.2. Rodale experiment

4.2.1. Abundance and QBS-ar scores

The total soil-invertebrate count for the Rodale experiment was 2556. The most abundant taxa were Acari (60%), Collembola (21%), Annelids (4%), Formicidae (3%), Coleoptera larvae (2.4%), and Chilopoda (1.2%). See table 3-7 for the remaining taxa collected.

There was a significant main effect of sampling date ($df=1$, $X^2=13.8$, $P\text{-value}<0.001$) and the interaction between tillage and sampling date ($df=2$, $X^2=14.7$, $P\text{-value}<0.001$) for microarthropod abundance. On average, no-till has greater abundance of microarthropods and in both tillage types the 28 July 2023 sampling date had higher abundance counts than 11 October 2023 (Fig. 3-5).

There was a significant main effect of sampling date ($df=1$, $X^2=9.4$, $P\text{-value}=0.002$) and the interaction between tillage and sampling date ($df=2$, $X^2=11.8$, $P\text{-value}=0.002$) for QBS-ar scores. In the no-till plots, 28 July 2023 had higher scores than 11 October 2023. In the tilled plots, however, there was no difference in score between dates (Fig. 3-6, Table 3-4). There was high variability in QBS-ar scores by treatment, tillage, and sampling date (Table 3-5). Soil moisture and management type (organic vs conventional) had no effect on either abundance or QBS-ar scores (Fig. 3-S1, 3-S2).

Table 3-4. Average QBS-ar scores per 88 cm² sample by tillage and date from the Rodale experiment. Sampling date 28 July 2023 had higher QBS-ar scores than 11 October 2023.

CLD: Compact letter display; Letter change denotes significance.

N = 16

Sampling date	Tillage	Mean ± SE	CLD
7/28/2023	No-till	85.4 ± 6.2	ab
7/28/2023	Till	65.5 ± 5.4	a
10/11/2023	No-till	51.1 ± 6.5	c
10/11/2023	Till	53.1 ± 5.2	bc

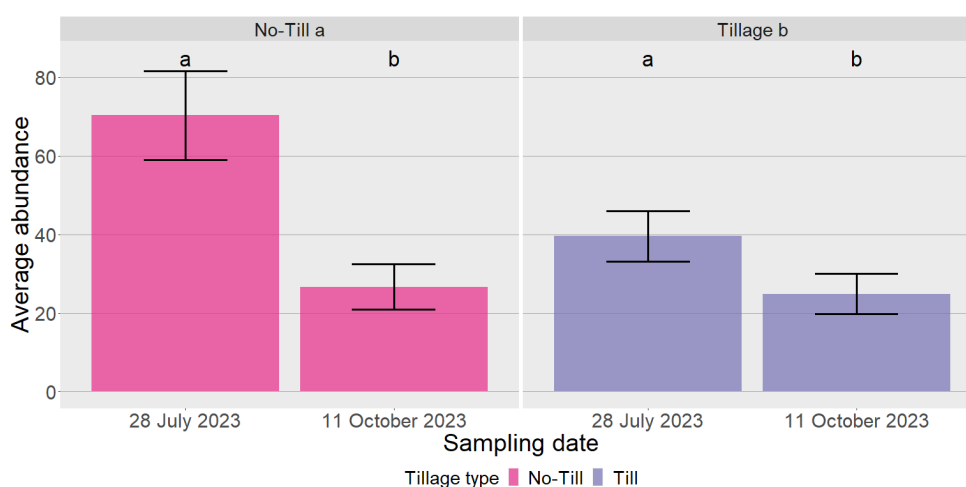


Figure 3-5. Soil-invertebrate abundance counts per 88 cm² sample by sampling date and tillage from the Rodale experiment. There was a significant main effect of sampling date ($df=1$, $X^2=13.8$, $P\text{-value}<0.001$) and the interaction between tillage and sampling date ($df=2$, $X^2=14.7$, $P\text{-value}<0.001$) for microarthropod abundance. On average, no-till has greater abundance of microarthropods and in both tillage types the 28 July 2023 sampling date had higher abundance counts than 11 October 2023. Letter change denotes significance.

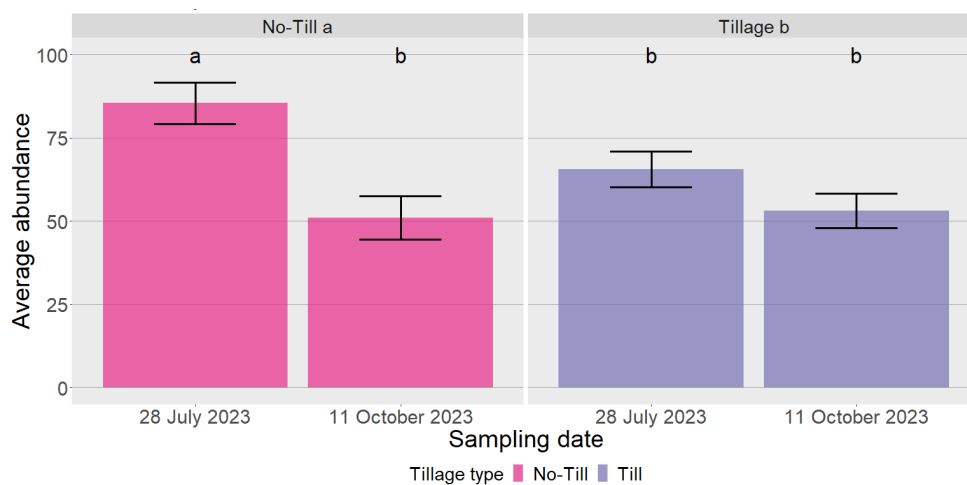


Figure 3-6. QBS-ar scores per 88 cm² sample by sampling date from the Rodale experiment. There was a significant main effect of sampling date ($df=1$, $X^2=9.4$, $P\text{-value}=0.002$) and the interaction between tillage and sampling date ($df=2$, $X^2=11.8$, $P\text{-value}=0.002$) for QBS-ar scores. In the no-till plots, 28 July 2023 had higher scores than 11 October 2023. In the tilled plots, however, there was no difference in score between dates. Letter change denotes significance.

Table 3-5. Average QBS scores by sampling date, treatment, and tillage from the Rodale experiment.

CCC: Conventional with cover crops; **CWW:** Conventional without cover crops; **OL:** Organic legume; **OM:** Organic manure

NT: No-till; **T:** Tillage

CLD: Compact letter display; Letter change denotes significance.

N = 4

Sampling date	Treatment	Tillage type	Mean \pm SE	CLD
7/28/2023	CCC	NT	109.3 \pm 14.1	b
7/28/2023	CCC	T	60 \pm 3.9	ab
7/28/2023	CWW	NT	65 \pm 10.8	ab
7/28/2023	CWW	T	76.6 \pm 10.5	ab
7/28/2023	OL	NT	92.5 \pm 4.8	ab
7/28/2023	OL	T	42.3 \pm 8.3	a
7/28/2023	OM	NT	75 \pm 7	ab
7/28/2023	OM	T	83 \pm 6.7	ab
10/11/2023	CCC	NT	40.7 \pm 12.5	a
10/11/2023	CCC	T	54.5 \pm 5.2	ab
10/11/2023	CWW	NT	61.3 \pm 13.3	ab
10/11/2023	CWW	T	59.8 \pm 3.9	ab
10/11/2023	OL	NT	50 \pm 14.3	ab
10/11/2023	OL	T	47 \pm 18.8	ab
10/11/2023	OM	NT	52.6 \pm 15.9	ab
10/11/2023	OM	T	51 \pm 10.8	ab

Table 3-6. Total soil-invertebrate abundance counts by crop from the cover-crop experiment.																
Crop	Acari	Annelid	Archaeognatha	Carabidae	Chilopoda < 5mm	Chilopoda > 5mm	Cocomorpha	Coleoptera larvae	Dermoptera	Diplopoda < 5mm	Diplopoda > 5mm	Diptera	Epigeic Collembola	Eudaphic Collembola	Hemi-Eudaphic Collembola	Formicid
Soybean	527	83	1	0	0	2	1	43	0	8	7	28	251	94	72	7
Corn	1,403.6	95.3	1	2	1	47.6	1	29.3	1	26	5.6	22.3	285.6	202.3	161.6	36.6
Crop	Hemiptera	Hymenoptera	Isopoda	Lepidoptera	Other Coleoptera	Other larvae	Paupoda	Protura	Pseudoscorpion	Psocodea	Siphonaptera	Spider	Thysanoptera	Symphyla		
Soybean	5	4	0	1	8	82	0	3	2	4	2	5	2	3		
Corn	13.6	4.6	0	0	15.3	53	8	8	4	6.6	3	5.3	3	17		

Table 3-7. Total soil-invertebrate abundance counts by sampling date and tillage type from the Rodale experiment.

Sampling date	Tillage	Acari	Annelid	Carabidae	Chilopod < 5mm	Coleopter larvae	Diplopod < 5mm	Diptera	Diptera	Epigeic Collembola	Eudaphic Collembola	Formicid	Hemi-Eudaphic Collembola	Hemiptera	Hymenoptera	Lepidoptera	Other Coleoptera	Other larvae	Paupoda	Protura	Pseudoscorpion	Siphonaptera	Spider	Thysanoptera
28 July 2023	T	379	52	1	5	13	0	1	6	19	68	1	36	10	1	0	16	18	0	1	0	0	2	1
28 July 2023	NT	624	22	1	14	32	3	4	5	28	140	73	100	7	3	1	18	18	0	18	0	1	1	0
11 October 2023	T	283	7	0	5	15	1	1	3	12	40	0	12	1	0	0	6	7	0	2	0	0	1	1
11 October 2023	NT	262	22	1	6	1	2	5	2	8	63	2	12	1	0	0	2	4	1	4	0	1	0	1

5. DISCUSSION

The aim of this study was to evaluate within different field management practices soil-invertebrate abundance and QBS-ar scores. The results did not corroborate my expectation that higher cover-crop biomass and less disturbance would result in higher soil-invertebrate abundance counts and higher QBS-ar scores.

Of all arthropods sampled for both experiments, Acari and Collembola were most abundant (Tables 3-8, 3-9), as expected (Curry, 1979; Ferraro & Ghera, 2007; Menta et al., 2020; Tabaglio et al., 2009). In the experiment testing the influence of cover crops on soil invertebrates, crop, year, and sampling date had the strongest effects on arthropod abundance, rather than cover-crop biomass. Previous research, however, suggest that invertebrate abundance should increase with habitat complexity (Boutin et al., 2009; Cortet et al., 2002; Diekötter et al., 2010; House & Parmelee, 1985). Importantly, the cover-crop treatments were only replicated in the same field twice (corn 2021-soybean 2022 and corn 2022-soybean 2023). While soil invertebrates can move through the landscape (Tuck et al., 2014), it is possible that two years is not enough time for highly adapted edaphic fauna to colonize and their populations to build. Additionally, previous crops planted in fields and their management likely influence their colonization of fields (Cortet et al., 2002). However, abundance or QBS-ar scores were similar between either corn and soybean legacy pair (corn 2021-soybean 2022 and corn 2022-soybean 2023). Finding that there was no change in abundance or QBS-ar score in the corn-soy rotations may be further explained by the amount of time that practices were used to see changes in abundance and adaptations to local conditions of life.

In the cover-crop experiment, soil-invertebrate populations differed between corn and soybean. Identity of crop species tends to influence soil-invertebrate populations (Boutin et al., 2009; Jernigan et al., 2020), but average QBS-ar scores did not differ between crop species (corn = 48.1; soybean = 49.8; Table 3-5). Both of these scores are lower than the threshold score (i.e., 93.7), which separates high quality and low quality soils (Menta et al., 2018). It is unclear why the scores that I calculated are roughly 50% lower than the threshold score. I hypothesized that soybean would harbor higher abundance and QBS-ar scores due to the differences in microclimate, branching complexity, ground cover from direct sunlight (Landis et al., 2000). Additionally, due to differences in canopy complexity, soybean generally harbor more biodiversity than corn (Finke & Denno, 2002). This hypothesis was not supported by my findings. While soil and abiotic conditions are factors, it appears the fields sampled in the cover-crop experiment did not support populations of highly adapted soil invertebrates. Furthermore, higher cover-crop biomass, at least in the short term, had no effect on QBS-ar scores.

In the Rodale experiment, management type (organic vs conventional) had no statistical effect on soil-invertebrate abundance or QBS-ar scores. Prior studies suggest that organic agricultural practices can increase species richness, but abundance of decomposers remains relatively constant across management practices (Tuck et al., 2014). My findings were similar because soil-invertebrate richness –comprising mostly decomposers– and QBS-ar scores did not change between organic and conventional practices. Previous studies also reported no changes in soil invertebrates by management type (Boutin et al., 2009; Menta et al., 2020).

In contrast to management type, sampling date, and tillage influenced abundance of soil invertebrates and QBS-ar scores. For example, in the Rodale experiment, I found mite abundance was 40% higher earlier in the season (28 July 2023) than later (11 October 2023). Changes in populations between dates may be attributed to seasonal trends, like precipitation and available organic matter (Cortet et al., 2002; Curry, 1979). In both sampling dates, tilled plots had numerically lower QBS-ar scores than no-till (Fig. 3-4; Table 3-6). This result is supported in previous research which found that mites are highly sensitive to tillage (Ferraro & Ghera, 2007; Tabaglio et al., 2009), which would ultimately affect QBS-ar scores. Of the sixteen samples I collected (Table 3-7), only two had QBS-ar scores near or above the high quality threshold of 93.7 (Menta et al., 2018); these samples included no-till conventional cover crop (109.3 ± 14.1) and no-till organic legume (92.5 ± 4.8). Previous research supports my results of higher QBS-ar scores within no-till (Menta et al., 2018, 2020; Sapkota et al., 2012; Tabaglio et al., 2009). Although two of the tillage-treatment combinations from the Rodale experiment had high quality QBS-ar scores, they account for 12.5% of scores recorded in the Rodale experiment and 8.3% of the total scores recorded from both experiments. These results are consistent with previous work that show how management type (organic vs conventional), sampling date, and tillage can affect soil-invertebrate populations (Cortet et al., 2002; Curry, 1979; Tuck et al., 2014). While any of these variables can influence soil invertebrates, within the suite of practices sampled in the Rodale experiment, tillage appeared to have the most influence.

Decreases over time in abundance of soil invertebrates could have resulted from diminishing amounts of residue in the field (Curry, 1979). Mite and collembola abundances have been shown to persist throughout the season with regular additions of cattle manure (Curry, 1979). Additionally, differences by sampling date may be attributed to weather. Spring and early summer conditions are typically warmer with higher levels of precipitation, which are favorable for increasing soil invertebrate abundance (Cortet et al., 2002). While precipitation can affect soil-invertebrate abundance, soil moisture did not significantly influence abundance or QBS-ar score.

Further investigation into the complexity of soil microarthropods in high cover-crop systems is prudent. High cover-crop biomass treatments appeared to have little influence on microarthropod communities in the short term (2 years). A longer term (>5 years) investigation into the effects of high cover-crop biomass on microarthropod abundance and QBS-ar scores would be insightful for researchers interested in investigating the legacy cover crop effects within agronomic systems on soil-invertebrate populations. It is advisable that this method of assessing crop management practices on soil invertebrates be used as new conservation agricultural practices are recommended to growers. This method of evaluating the biological quality of an ecosystem can be highly effective and may provide novel insight. Combining the QBS-ar index with traditional soil quality tests may provide additional information for growers on their crop management practices. Furthermore, research comparing the QBS-ar index and current soil quality tests and metrics may be insightful.

6. CONCLUSION

My results suggest that agricultural practices and crop species influenced soil-invertebrate abundance (Menta et al., 2018). Tillage and sampling date are the main factors in both abundance and QBS-ar scores (Menta et al., 2010, 2018; Sapkota et al., 2012; Tabaglio et al., 2009). In the cover-crop experiment, crop species influenced microarthropod abundance, but not QBS-ar scores. Additionally, twenty-six of the total twenty-eight QBS-ar scores fell below the high-quality threshold of 93.7, suggesting that the communities I sampled were degraded, perhaps by regular disturbance from herbicide and insecticide applications and/or tillage. Soil invertebrate communities may also be influenced by soil type and plant communities, but my experiments did not take these factors into account.

A study combining multiple soil quality tests with the QBS-ar index would be insightful to further evaluate the reliability of microarthropods as biological indicators in agricultural systems. Along with this, sampling in multiple cover-crop species and crop-management practices would allow for a broader understanding of how biological quality changes within a suite of crop management strategies. A study like this would allow for direct comparisons of current soil quality assessments and the QBS-ar index. Furthermore, developing a means to use soil-quality assessments and the QBS-ar index together could provide more meaningful information on soil quality. With this information, growers could make more informed decisions about their management strategies that include increasing yield and agroecosystem quality.

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Conclusion

This thesis covered multiple facets of corn and soybean agroecosystems of Pennsylvania including evaluating the influence of cover crops on pests and predators, determining the most abundant invertebrate predator groups in modern soybean production systems, attempting to quantify the contributions of spiders in pest control, and the evaluation of soil biological quality within a suite of crop and field-management practices. While the results from this thesis may be more consistent with mid-Atlantic trends, the practices evaluated can be adopted in other regions. Additionally, further research from each chapter may be insightful.

In chapter one, I evaluated the effects of delayed termination of a cover crop and planting green on pest and predator populations. Despite concerns in agronomic systems of increasing pest populations and reducing yield by planting green (Douglas & Tooker, 2012; Willson & Easley, 1992), my results suggest that planting green: *i*) does not increase the amount of pest damage or damage incidence, *ii*) can promote predation, and *iii*) does not limit yield in either corn or soybean. In the mid-Atlantic region, studies have shown the benefits that come from planting green and practicing IPM (USDA, 2015). These benefits include, but are not limited to a, reduction in the potential for soil erosion (Renard et al. 1991; Gyssels et al. 2005), improved soil quality metrics (Gyssels et al., 2005), reduction of nutrient leaching into watersheds (Alonso-Ayuso et al., 2014; Farsad et al., 2011; USDA, 2015), and decrease of overall weed and invertebrate pest pressures, which is supported by findings and previous studies (Bohnenblust et al., 2014; Douglas & Tooker, 2012; Finke & Denno, 2002; Hammond & Stinner, 1987; Landis et al., 2000; Mischler et al., 2010; Rowen et al., 2020; Thorbek & Bilde, 2004). My results suggest that these practices be adopted by row-crop growers across the United States looking to decrease invertebrate pest damage and incidence and increase natural-enemy populations.

In chapter two, I set out to determine the most abundant generalist predators of modern soybean systems. Additionally, I attempted to quantify the contributions of spiders in pest control. My results revealed that spiders are the most abundant generalist predators in soybean grown in central Pennsylvania. Additionally, time of year was a key driver of populations, which is likely in response to habitat availability (Finke & Denno, 2002; Landis et al., 2000; Schmidt et al., 2008). Of the spiders I collected, the most dominant families were ambush or mobile hunters, rather than web builders, which makes sense given the disruptive nature and crop phenology of the annual growing season (Landis et al., 2000; Prieto-Benítez & Méndez, 2011). The cover-crop termination experiment illustrates that spiders are not supported throughout the season in annual cropping systems and farming and pest control practices could be adapted with spiders in mind, which can be done by increasing non-cash-crop habitats throughout the season (Schmidt et al., 2008). This study is integral to continued research on bolstering

spider populations in annual systems because I recorded that spiders are the most abundant predators and that cover cropping alone is not enough to increase the diversity of foliage-dwelling invertebrate fauna in modern soybean systems. More research is necessary to understand how to support spider populations within annual cropping systems.

In chapter three, I used the QBS-ar index to investigate the influence of cover crop use and a suite of crop-management practices on the biological quality of fields used to grow field crops. My results suggest that agricultural practices and crop species influenced soil-invertebrate abundance (Menta et al., 2018). Tillage and sampling date are the main factors in both abundance and QBS-ar scores (Menta et al., 2010, 2018; Sapkota et al., 2012; Tabaglio et al., 2009). In the cover-crop experiment, crop species influenced abundance, but not QBS-ar scores. Additionally, twenty-six of the total twenty-eight QBS-ar scores fell below the high-quality threshold of 93.7, suggesting that the communities I sampled were degraded, perhaps by regular disturbance from herbicide and insecticide applications and tillage. Soil invertebrate communities may also be influenced by soil type and plant communities, but my experiments did not take these factors into account. A study combining multiple soil quality tests with the QBS-ar index would be insightful to further evaluate the reliability of microarthropods as biological indicators in agricultural systems. Along with this, sampling in multiple cover crop and crop management practices would allow for a broader understanding of how biological quality changes within a suite of crop management strategies. A study like this would allow for a direct comparison of current soil quality assessments and the QBS-ar index. Furthermore, developing a means to use soil quality assessments and the QBS-ar index in conjunction could provide more meaningful information on soil quality. With this information, growers could make more informed decisions about their management strategies that include increasing yield and agroecosystem quality.

Appendix A

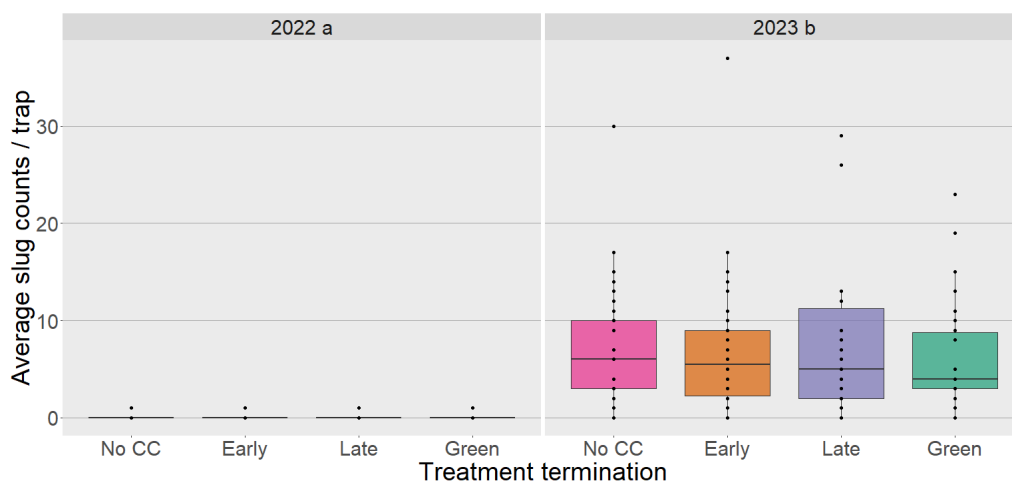


Figure 1-S1. Slugs populations by year and treatment from the spring season in soybean plots. There was a significant main effect of year ($df=1$, $X^2=6$, P -value=0.01) for slug populations sampled during spring. Slug populations were greater in 2023 than 2022 Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

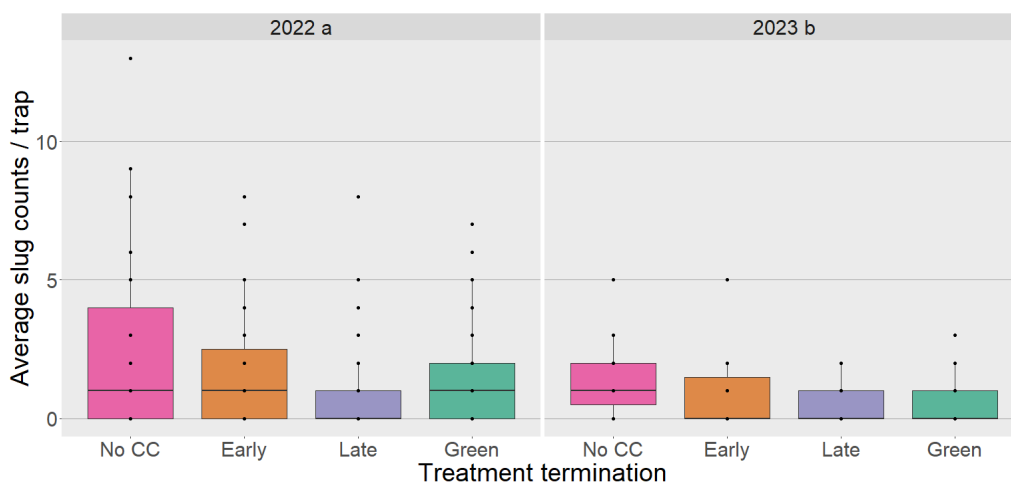


Figure 1-S2. Slugs populations by year and treatment from the fall season in soybean plots. There was a significant main effect of year ($df=1$, $X^2=181$, P -value<0.001). Populations in 2022 were greater than in 2023 Letter change denotes significance. (Early: 14-28 DPP; Late: 3-7 DPP; Green: 1-3 DAP)

Appendix B

Table 2-S1. Average abundance of tetragnathids by treatment from the cover-crop experiment. Abundance was greater in the no cover crop than the planting green treatment. CLD; Compact letter display: Letter change denotes significance.

Treatment	Mean \pm SE	CLD
No CC	0.093 \pm 0.04	a
Early terminated	0.0006 \pm 0.0006	b
Late terminated	0.05 \pm 0.03	ab
Planting green	0.012 \pm 0.012	ab

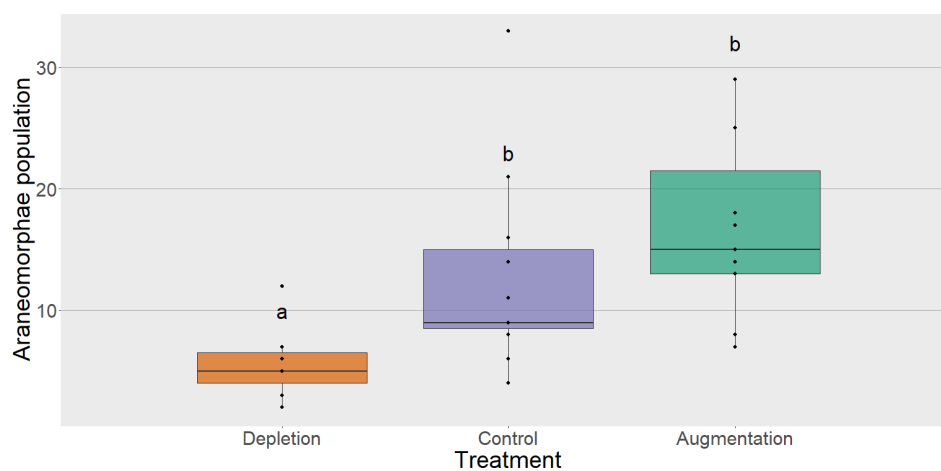


Figure 2-S1. Total araneomorph counts per treatment from the augmentation experiment. The depletion had lower counts than both the control and the augmented treatment. Letter change denotes significance.

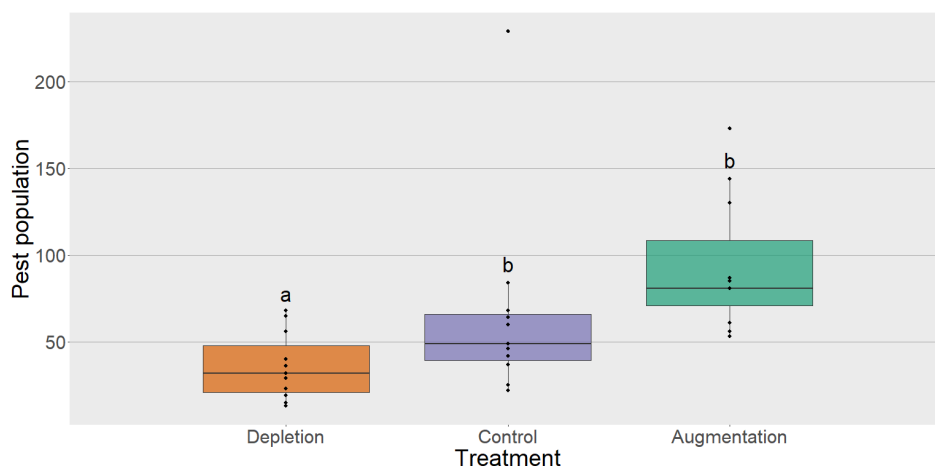


Figure 2-S2. Total pest counts per treatment from the augmentation experiment. The augmented treatment had higher pest populations than both the depletion and the control. Letter change denotes significance.

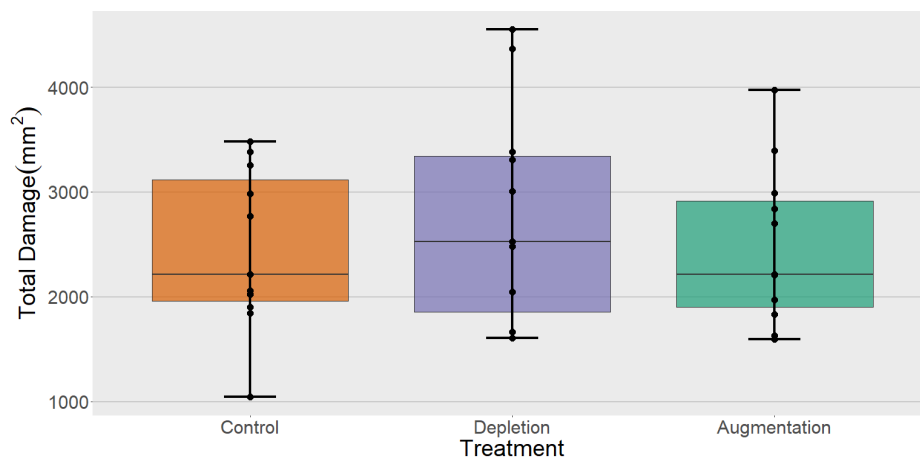


Figure 2-S3. Total defoliation of soybean plants by treatment from the augmentation experiment. No recorded differences among treatments.

Appendix C

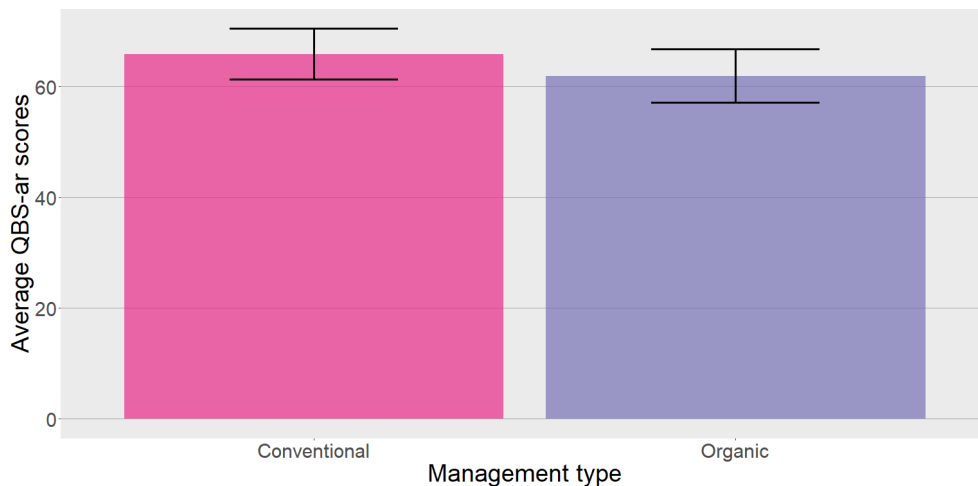


Figure 3-S1. QBS-ar scores per 88 cm² sample by management type from the Rodale experiment. No differences recorded between conventional and organic. Letter change denotes significance.

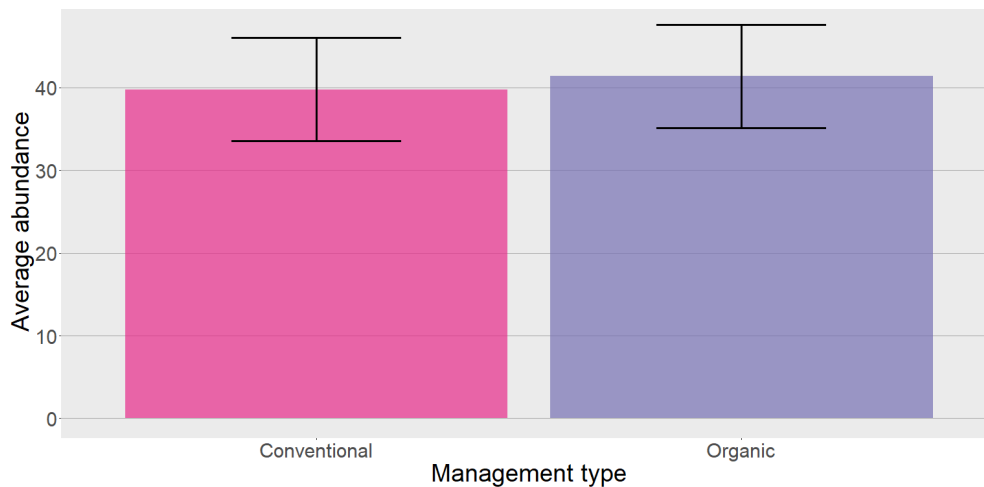


Figure 3-S2. Soil invertebrate abundance counts in per 88 cm² sample by management type from the Rodale experiment. No differences recorded between conventional and organic. Letter change denotes significance.