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Effects of cover crops, rotation, and biological control products on soil properties and productivity in organic vegetable production in the Northeastern US

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Abstract The potential benefits of different cover crops, biological control amendments, and vegetable rotation on soil chemical and biological properties, crop development and yield, and disease development in organic vegetable production, as represented by legume (green bean, *Phaseolus vulgaris*), cucurbit (yellow summer squash, *Cucurbita pepo*), and solanaceous (sweet pepper, *Capsicum annuum*) vegetable crops, were evaluated in a multi-year field trial in Maine, USA. Cover crops evaluated included winter rye (*Secale cereale*)/hairy vetch (*Vicia villosa*), mustard (*Brassica juncea*) green manure, a multi-species mixture (8 crops), and a fallow control. Overall, cover crops had only marginal effects on soil chemical properties, but all cover crops improved biological properties (microbial activity, populations, respiration) compared to fallow soil. The multi-species mixture and rye/vetch cover crops were associated with earlier emergence in beans and squash. All cover crops improved yield in beans and squash by 7–13%, but only the cover crop mixture increased yield for pepper (by 7–11%). Minimal crop diseases were observed throughout these studies, and biological control amendments, which included commercial formulations of *Streptomyces*, *Trichoderma*, and *Bacillus* sp., in general, did not positively affect yield, but reduced powdery mildew on squash and leaf necrosis on beans by 10–28%. The vegetable rotation also had significant effects, with beans yielding 8% higher following squash vs. pepper, squash yielding 15% higher following beans vs. pepper, and pepper yielding 11% higher following beans vs. squash. These results help define specific management practices to improve organic vegetable production and provide useful information and options for growers.

Keywords Cover crop mixtures · Snap bean · Summer squash · Sweet pepper · Mustard green manure · Biological control

Mention of names or commercial products in this article is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

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Introduction

Growing interest in organic farming and farming practices, by growers and consumers alike, has greatly increased the demand for and production of fresh market organic vegetables in recent years. Here in the Northeastern US, just in the New England area (comprising Connecticut, Maine, Massachusetts, New Hampshire, Rhode Island, and Vermont), the number of farms and acres producing organic vegetables increased by over 50% from 2011 to 2016 (USDA NASS 2012, 2017). However, successful sustainable organic vegetable farming faces many production challenges. In this region, organic vegetable production tends to be small-scale but highly diversified, involving several production crops and requiring knowledge and expertise on the

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management of numerous crops and crop types. The effective use of cover crops and rotations, which help build soil fertility and health and the management of pests and diseases, are the cornerstone of organic production and are especially critical for developing effective sustainable vegetable production systems.

There are many very useful and helpful general resources documenting the benefits, importance, and applications of cover crops and rotations in organic farming (Lu et al. 2000; Sarrantonio and Gallandt 2003; Snapp et al. 2005; Clark 2007; Magdoff and van Es 2009; Mohler and Jonson 2009; Robacer et al. 2016). However, there are relatively few sources of information detailing specific combinations of practices and how they may or may not work together to improve or optimize production and productivity within a system appropriate for the Northeastern US. There are many available options for cover crops, and choices are usually made based on the primary goals of the cover, but it is not always clear what may work best with specific types of crops and conditions. Standard cover crops such as winter rye or rye/legume combinations have well-established benefits (Teasdale and Abdul-Baki 1998; Griffin et al. 2000; Fageria et al. 2005). More recent uses of multi-species cover crop mixtures show potential for addressing multiple soil health needs but have not always clearly demonstrated benefits over single crop covers (Wortman et al. 2012; Finney et al. 2016; Appelgate et al. 2017). *Brassica* family cover crops, such as mustard green manures, have weed- and disease-suppressive properties, but also may or may not be a good fit within specific production systems (Matthiessen and Kirkegaard 2006; Larkin and Griffin 2007; Larkin 2013).

Regarding management of crop diseases, although good soil health management practices should reduce soilborne diseases, these approaches may not be sufficiently effective and supplemental management practices may be needed. Biological control using applications of microorganisms known to reduce populations or the disease-causing activity of plant pathogens is an approach available to organic growers that has not been very widely adopted but may provide additional or supplemental control options. Although the specific problem diseases may vary greatly with different vegetable crops, overall, there are several general disease problems that occur on a wide variety of crop types, such as damping-off and root diseases (caused by *Rhizoctonia*, *Pythium*, and *Phytophthora* spp.), Fusarium and Verticillium wilts, gray mold (*Botrytis cinerea*), anthracnose (*Colletotrichum* spp.), and others, and can be managed using similar approaches. A variety of microorganisms have been shown to have activity against multiple soilborne diseases of vegetables, including *Trichoderma* spp., *Streptomyces* spp., and *Bacillus* spp. (Yuan and Crawford 1995; Sabaratnam and Traquair 2002; Jacobsen et al. 2004; Bonaldi et al. 2011; Sharon et al. 2011; Li et al. 2015; Vurukonda et al. 2018; Sharma et al. 2017), several of which have been developed into commercially available biocontrol products. The use of multiple biocontrol agents with differing mechanisms of action and target pathogens has been associated with improved disease management (Guetsky et al. 2001, 2002; Roberts et al. 2005).

Crop diversity is also important for soil health, and vegetable production systems should include different types of vegetable crops in their rotations. Three main families of vegetable crops, cucurbits (cucumber, squash, melons, pumpkins), legumes (beans, peas, lentils, soybeans), and solanaceous crops (tomato, potato, eggplant, peppers) cover many of the most commonly grown vegetable crops. Each group has its own characteristics, requirements, and management issues, but within each group, they are quite similar. In this study, we included a representative crop from each of these family groups to provide a balanced rotation and assess cover crop effects for each type.

Weed management is another critical area of organic production. Black plastic mulch has several benefits and is often used as a weed barrier in organic vegetable production (Nachimuthu et al. 2017). However, there are numerous problems associated with its removal, disposal, and potential soil health issues that make its use less than ideal for organic agriculture (Steinmetz et al. 2016). Various forms of paper mulch (which does not have to be removed and disposed, as it breaks down in the soil) have been used as an alternative with varying success (Miles et al.2007; Coolong 2010; Haapala et al. 2014). In this study, we used a commercial paper mulch weed barrier (OMRI listed) to assess its usefulness within an organic vegetable production system.

The purpose of this research was to assess how different cover crops would affect a multiple crop organic vegetable production system, as well as determine whether biological control amendments for soilborne diseases would provide a beneficial addition to the system. Thus, in this research, three cover crops (in addition to no cover crop) and two biological control treatments were assessed for their effects on soil properties, crop growth and yield, and disease development in three organic vegetable crops (snap bean, yellow squash, and sweet peppers) in an organic vegetable production system over two full cropping seasons in central Maine. In addition, vegetable rotation options were compared, and the use of a paper mulch weed barrier was evaluated for its feasibility within the system. The ultimate goal of these studies is to develop and optimize improved production systems for organic vegetables that maximize sustainability and productivity.

Materials and methods

Field design and management

Field experiments were conducted at the USDA Research Farm in Newport, ME, over three consecutive growing seasons (2014–2016), same plot locations each season, as a split-block design (with cover crop as main plot and biocontrol treatment as the sub-plot) with four replicate blocks for each of three vegetable crops. Each main plot was 9.1 × 7.3 m, consisting of 3 subplots (3.0 × 7.3 m) containing 2 rows (6.1 m length) of vegetable crops each. The soil type was a Nokomis sandy loam, a coarse-loamy, mixed, frigid, Typic Haplorthod. This field location had a previous history of conventional potato production up through 2008, but after that, the field was planted to ryegrass and transitioned over the next few years to organic production only. Tillage for all plots at the start of the study consisted of primary tillage with a chisel plow and then secondary tillage with a disc harrow prior to planting. Soil for all vegetable plots was prepared by addition of composted dairy manure at the rate of 46 m^3/ha (~ 27 Mg/ha), which was tilled into the soil prior to planting. Compost composition (fresh weight basis) was ~ 32% solids, 8.8% C, 0.67% N, 0.30% K, 0.18% P, and a C:N ratio of 13. Vegetable plots were also fertilized with a commercial organic fertilizer (Fertrell Feed-n-Grow, Fedco seeds), with 3–2-3 NPK content (from blood meal, fish meal, feather meal, alfalfa, rock phosphate, greensand, and kelp) applied to soil at 290 kg/ha prior to vegetable planting, based on prior soil tests.

Weeds were managed through use of a paper mulch barrier within plots, as well as by hand cultivation between plots. Fields were dependent on natural rainfall for all watering needs. Environmental conditions (air temperature, relative humidity, and rainfall) were monitored at an on-site weather station and used to determine daily, weekly, and monthly average conditions throughout the cropping season. In 2016 only, additional environmental conditions of air temperature and relative humidity were monitored within plots at canopy level, and soil temperature and soil moisture, measured by Watermark sensors (Spectrum Technologies, Plainfield, IL), were monitored within plots in each block at 15-cm depth for (a) under the paper weed barrier, (b) outside the paper barrier, and (c) under paper in squash plots where insect netting was used, using Watchdog data loggers (model 450, Spectrum Technologies, Plainfield, IL) throughout the season.

Soil chemical and biological properties

Soil samples were collected throughout each block in July 2014 prior to planting cover crops to assess soil properties at the beginning of the experiment. In each of three zones in each block, 6 to 8 soil cores (2.5 × 15 cm) were collected and combined into one composite sample, sieved through a 2-mm screen, air-dried, and used for soil physical and chemical analyses. Likewise, soil samples were collected within each cover crop plot in spring of 2015, prior to planting vegetable crops, for subsequent analyses. Soil samples were also collected within each cover and vegetable crop plot in the spring of 2016 (prior to 2nd year vegetable crop), and again in spring of 2017 as a follow-up to assess the previous year's effects on soil properties. Soil properties measured included pH, organic matter content, cation exchange capacity, and concentrations of nutritionally important elements and compounds. Potentially available N, as nitrate (NO_3^-) and ammonium (NH_4^+), was determined using cold water bath KCl extractions. Soil concentrations of P, K, Ca, Mg, Al, B, Fe, Mn, Na, S, Cu, and Zn were estimated using Modified Morgan extraction procedures (Helmke and Sparks 1996) and analyzed using inductively coupled plasma optical emission spectroscopy (ICP-OES) by the University of Maine Analytical Lab (Orono, ME). Values were expressed as mg/kg soil. Average soil properties for initial samples collected in 2014 prior to the start of the study included soil texture of 50.1% sand, 41.0% silt, and 8.9% clay, 6.15 pH, 4.9% organic matter (OM), and 7.3 mg/kg P, 310 mg/kg K, 201 mg/kg Mg, and 917 mg/kg Ca.

For 2016 soil samples, additional testing on biological properties was conducted, including CO_2

respiration using the Solvita® CO_2 burst assay (Woods End Laboratories, Mt. Vernon, ME) (Brinton 2015), soil dilution plating for enumeration of culturable bacteria and fungi populations (Larkin 2003), and microbial activity based on the BIOLOG (Biolog, Inc., Hayward, CA) substrate utilization assay (using BIOLOG GN2 plates) (Larkin 2003).

Cover crop treatments

Cover crop treatments consisted of (1) cereal rye (*Secale cereale*) mixed with hairy vetch (seeded at 67 and 22 kg/ha, respectively), (2) mustard blend ('Caliente 199') green manure (9 kg/ha), (3) a multi-species cover crop mixture consisting of legumes (field peas [*Pisum sativum*], hairy vetch, and crimson clover [*Trifolium incarnata*], at 10, 6, and 4.5 kg/ha), grains (cereal rye and annual ryegrass [*Lolium multiflorum*], 16 and 5 kg/ha), Brassicas (oilseed radish [*Raphanus sativus*] and 'Dwarf Essex' rapeseed [*Brassica napus*], 3.5 and 9 kg/ha, respectively), and buckwheat (*Fagopyrum esculentum* at 9 kg/ha) planted using a cone seeder drill (Gandy Company, Owatonna, MN), and (4) fallow (no cover crop). Cover crops were seeded in July of 2014. The mustard green manure treatment was flail mowed, then immediately incorporated with a rotovator in late September 2014, whereas the rest of the cover crops were allowed to grow through winter. All plots were tilled in spring of 2015, first with a chisel plow, then with a Perfecta harrow (Unverferth manufacturing, Kalida, OH) as needed to prepare the field for vegetable planting. Prior to planting, each plot was covered with a biodegradable paper mulch (organic heavyweight grade, WeedGuardPlus, Aurora, CO) for weed control in 2, 0.91 × 6.4 m strips and held down by burying with soil around the edges. Prior to planting, holes (~ 7.6 cm diam.) were cut in the paper in rows at the appropriate spacing for each crop.

Biological control treatments

Two biological control treatments, each consisting of two commercial biocontrol products, were used in addition to a nontreated control for each vegetable crop and cover crop combination. The combination of two biocontrol agents was used to maximize potential disease control capabilities. The first treatment, or BC1, was a combination of the actinomycete *Streptomyces lydicus* WYEC108 (Actinovate AG, Novozymes BioAg Inc., Brookfield, WI) and *Trichoderma virens* GL-21 (SoilGard, Certis USA, Columbia, MD, added to soil around each plant hole at a rate of 1 g and 2.5 g/plot, respectively, as a liquid suspension (in 0.5 L water) at the time of planting. Both these agents have activity against damping-off and root rot pathogens, and *S. lydicus* also provides suppression of foliar fungal pathogens responsible for powdery mildew and various leaf spots (Yuan and Crawford 1995; Sharon et al. 2011). BC2 was a combination of the bacteria *Bacillus amyloliquefaciens* D747 (Double Nickel 55, Certis USA, Columbia, MD), and the fungus *Trichoderma harzianum* T-22 (RootShield Plus WP, Bioworks, Victor, NY) also added to soil at rate of 1 g and 2.5 g/plot as a liquid (0.5 L water) at the time of planting. In this combination, *B. amyloliquefaciens* has activity against foliar diseases such as powdery mildew, *Alternaria*, *Botrytis*, and bacterial leaf spots, and *T. harzianum* has activity primarily against damping-off and root rot fungal pathogens (Sharon et al. 2011; Li et al. 2015). All biocontrol products used are OMRI listed and NOP certified for use in organic production. A nontreated control consisted of no biological treatment of any kind. No pathogen inoculum was added to the field, with development of any diseases or pest issues dependent on natural inoculum and infestation.

Vegetable crops

Vegetable crops grown were organic green snap beans (*Phaseolus vulgaris* L.), yellow summer squash (*Cucurbito pepo* L.), and sweet peppers (*Capsicum annuum* L.), as representative examples of legume, cucurbit, and solanaceous vegetable crops. The snap bean was variety 'Provider' (Johnny's Selected Seeds, Winslow, ME), a bush-type green snap bean, planted at a 15-cm spacing in 2, 6.1-m rows (0.91 m between rows) per plot for a total of 40 plants per row. The yellow squash was 'Success PM' straightneck summer squash (High Mowing Organic Seeds, Wilcott, VA) planted at a spacing of 0.61 m (between row spacing 0.91 m), but plants staggered, with 2–3 seeds planted per hole, and later thinned to one plant, for a total of 10 plants per row. The type of pepper used was an organic orange-colored sweet pepper 'Lunchbox Orange' (Johnny's Selected Seeds, Winslow, ME). The pepper was not direct-seeded but germinated and seedlings were grown in the greenhouse in light potting mix for 6 weeks before transplanting into the field plots at a within-row spacing of 46 cm and between rows of 0.91 m, for a total of 13 plants per row. Each vegetable crop was grown each year in separate sub-blocks, following a rotation of either squash-bean-pepper or squash-pepper-bean. Due to technical and logistic issues, planting was delayed in 2015, with beans and squash planted in the first week of July, and peppers planted in the second week of July 2015. In 2016, all crops were planted in the first week of June. However, due to dry conditions and poor germination, a substantial portion of the beans and squash were replanted on June 20, 2016. After planting and prior to emergence, squash plots were covered by an insect netting (ProtekNet, Johnny's Seeds, Winslow, ME) laid over steel hoops to protect plants from squash bugs and other insects during early growth stages. Netting was removed when plants were at the flowering stage to allow pollination and fruit development. Emergence (as percentage of emerged seedlings relative to total seeds planted) was assessed for squash and bean crops periodically through the first 35 days after planting (DAP). All seed, products, equipment, inputs, and methodologies used throughout these trials were certified organic and/or approved for use in organic production.

Crop growth, yield, and disease development evaluations

All crops were monitored in the field for signs and symptoms of foliar and soilborne diseases. Crops were also scouted for insect activity and damage. Squash bugs (*Anasa tristis*), cucumber beetles (*Acalymma vittatum*), and occasional findings of tomato hornworms (*Manduca quinquemaculata*) on pepper were removed by hand as needed. All vegetables were harvested by hand as they ripened to maturity and weighed and recorded by row. On each harvest date, all ripe (ready to eat) vegetables within each row were harvested. In 2015, bean and squash harvests began on August 17, with beans harvested approximately once per week and squash twice per week through September 18, 2015, for a total of four harvests for beans and seven harvests for squash. Pepper harvests were once a week, beginning August 31 and ending September 22, 2015, for a total of four harvests. In 2016, bean and squash harvests began on July 25 (due to earlier planting dates) and continued over a total of 8 weeks, ending on September 15, 2016. Pepper harvests ran from August 8 to September 30, 2016. During the final month of harvest in both years, visual assessments of any symptoms of disease development, such as powdery mildew, various leaf spots, or necrosis, were recorded (as the percentage of leaf area affected) for each plot periodically as needed. Yield was evaluated as the total weight of harvested vegetables per 6.1-m harvest row in each plot at each harvest date and as the total for all harvest dates and converted to the equivalent value expressed as Mg/ha.

Statistical analyses

Soil properties, yield estimates, and disease assessment data were analyzed using standard analysis of variance (ANOVA). Data from each crop year were analyzed separately, and then data from both years were also combined and analyzed together (with year as an additional factor) to evaluate average and overall effects over the course of the study. Significance was evaluated at $P < 0.05$ for all tests. Mean separation was accomplished with Fisher's protected LSD test. All analyses were conducted using the Statistical Analysis Systems ver. 9.4 (SAS Institute, Cary, NC).

Results

Environmental conditions and overall crop growth

Environmental conditions varied during the vegetable growing seasons over the 2 years of the study. Overall, temperatures tended to average above normal for the summer in each year, with notably higher temperatures for the summer months in 2016, compared with the long-term (30-year) averages for the area (Table 1). Rainfall was highly variable from month to month and year to year, with 2015 being wetter than normal in June

Table 1 Average daily temperature and total rainfall for the months of May through September at the Newport, ME, research site for 2015 and 2016 compared with long-term (30-year) average (LTA) conditions

Treatment	Average daily temperature (°C)			Rainfall (cm)		
	2015	2016	LTA	2015	2016	LTA
May	14.0	14.3	12.0	7.2	5.1	9.5
June	15.4	18.1	17.0	14.0	7.5	9.9
July	19.7	22.3	19.8	6.7	5.6	8.7
August	20.9	22.1	18.9	10.1	6.7	8.6
September	18.0	16.6	14.2	7.5	3.8	9.6
Season avg	17.6	18.7	16.4	9.1	5.7	9.3

and August, but slightly below normal for July and September, resulting in an average rainfall season overall (Table 1). In 2016, however, lower than normal rainfall throughout the summer resulted in dry conditions and an overall rain deficit for the summer of 18 cm relative to the long-term average conditions (Table 1).

Environmental conditions were also measured within and around plots throughout the 2016 growing season, and differences in soil moisture (measured at 15 cm depth) in relation to rainfall were observed among measurements taken outside the paper mulch, under the paper mulch, and under squash plots (under mulch and with additional insect netting) (Fig. 1). Early in the growing season, when the spring rains were more plentiful and soil was fairly moist, soil outside the paper mulch tended to be slightly wetter than under the mulch, but as the season progressed and soils began to dry out in mid-July through the rest of the season, soil under the paper mulch retained moisture better than outside the mulch (Fig. 1). Also, during the mid-July dry period, soil under the squash plants showed drier conditions than either under paper or outside the paper. After some August rain, soils quickly began drying and remained somewhat dry through September, but soil under the paper mulch maintained better soil moisture than either the soil outside the paper or under the squash plots throughout the remainder of the season (Fig. 1). Soil temperature did not vary substantially among the three location types throughout the season (data not shown).

Due to the late planting in 2015, crop growth was delayed resulting in a shortened growing season and lower than normal yields, particularly for peppers. In 2016, earlier emergence, higher temperatures, and a longer growing season resulted in greater yields for all three vegetable crops relative to 2015. Insect and disease pressure were minimal throughout both years, with little indication of disease problems, other than powdery mildew (caused by *Podosphaera xanthii*) on squash and some general leaf chlorosis and necrosis (etiology undetermined) emerging late in the season both years.

Cropping effects on soil properties

Cover crop treatments did not greatly affect soil physical and chemical properties in the spring of 2015 prior to planting the vegetable crops. However, there were some significant differences among treatments. Soil pH following the rye/vetch cover and multi-species mixture was higher than following mustard or no cover crop (fallow), and NO_3-N was lower, whereas NH_4-N was higher following rye/vetch than all other treatments (Table 2). There were no differences among cover crop treatments for organic matter content or major element concentrations (Table 2) or for all other nutrients tested (Al, B, Cu, Fe, Mn, Na, S, Zn, or CEC—data not shown). Cover crop was also not a significant factor on soil properties in the spring 2016 sampling (Table 3). The previous year's vegetable crop, however, did significantly affect most soil properties measured. Soil pH was higher following all three vegetable crops than the fallow control. Squash and bean maintained higher K and Mg concentrations than fallow soil, squash also resulted in higher NO_3 than pepper, whereas bean kept higher P levels than fallow soil, and fallow soil showed lower Ca concentration than following all three vegetable crops (Table 3). In addition, fallow soil demonstrated lower nutrient concentrations than all or some

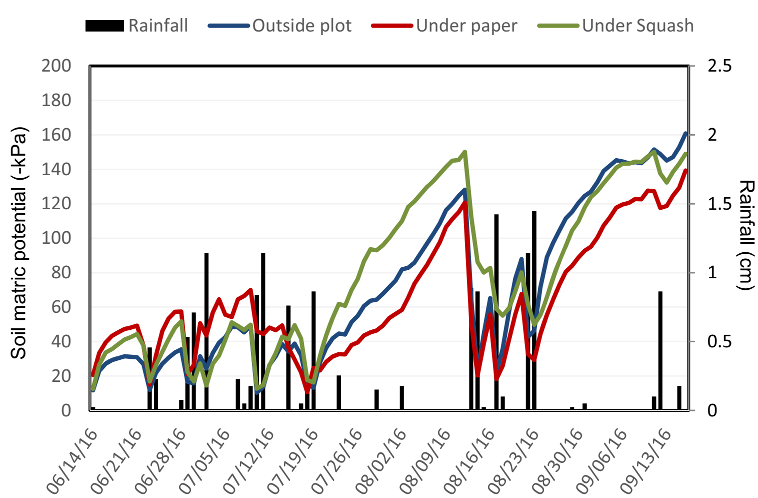

Fig. 1 Daily rainfall totals and daily average soil moisture (soil matric potential) readings throughout the 2016 growing season at field site in Newport, ME, as measured within plots under a paper mulch weed barrier (under paper), bare soil outside the paper (outside plot), and under paper mulch in squash plots fitted with insect netting (squash mulch)

Table 2 Selected soil chemical properties and nutrient concentrations in spring 2015 following different cover crop treatments prior to planting vegetable crops

Cover crop	pH	OM (%)	(mg/kg soil)					
			NO_3	NH_4	P	K	Mg	Ca
Mixture[y]	6.30 a [z]	5.28 a	4.0 b	3.3 b	6.73 a	211 a	168 a	1037 a
Rye/vetch	6.33 a	4.98 a	4.0 b	6.0 a	5.92 a	180 a	162 a	923 a
Mustard	6.10 b	4.45 a	19.0 a	3.5 b	6.02 a	187 a	153 a	942 a
Fallow	6.11 b	4.89 a	15.0 a	2.0 b	6.45 a	212 a	155 a	919 a
LSD	0.12	0.78	5.8	2.0	2.40	55	29	171

[y] Cover crop mixture consisted of eight crops (field peas, hairy vetch, crimson clover, cereal rye, annual ryegrass, oilseed radish, rapeseed, and buckwheat) planted together

[z] Values within columns followed by the same letter for each parameter are not significantly different from each other based on ANOVA and Fisher's protected LSD test ($P < 0.05$)

of the vegetable crops for B, S, Zn, and CEC, but showed higher concentrations of Al, Cu, and Fe (data not shown).

Soil microbial biomass and activity, as represented by CO_2 respiration (Solvita test) and substrate utilization (BIOLOG) assays in 2016 soil samples, tended to be higher in soil from all previous cover crop treatments than in fallow soil, with CO_2 respiration significantly higher in the cover crop mixture soil than fallow soil (Table 4). Microbial activity based on average well color development in the substrate utilization assay indicated higher activity in all the previous cover crop treatments relative to previously fallow plots (Table 4). Total culturable soil bacterial populations were also higher in soil from all the previous cover crop treatments relative to fallow soil, but there were no differences in fungal populations among cover crop treatments (Table 4). In addition, soil from plots previously planted to all the vegetable crops demonstrated higher CO_2 respiration than fallow soil, and with soil from squash and bean registering higher CO_2 respiration than pepper

Table 3 Selected soil chemical properties and nutrient concentrations in spring 2016 as affected by earlier cover crop treatment or previous year vegetable crop

	pH	Organic matter (%)	(mg/kg soil)					
			NO_3	NH_4	P	K	Mg	Ca
Cover Crop								
Mixture[y]	6.41 a[z]	4.63 a	6.75 a	3.67 a	6.55 a	195 a	157 a	1048 a
Rye/vetch	6.38 a	4.92 a	5.08 ab	2.67 a	6.85 a	208 a	162 a	1023 a
Mustard	6.39 a	4.95 a	4.25 b	5.25 a	7.33 a	208 a	156 a	1018 a
Fallow	6.46 a	4.69 a	4.25 b	3.67 a	7.54 a	212 a	164 a	1064 a
LSD	0.10	0.43	1.94	4.26	1.49	26	17	95
Previous crop								
Squash	6.46 a	4.79 a	5.88 a	5.50 a	7.17 ab	213 a	167 a	1048 a
Pepper	6.39 a	4.97 a	3.88 b	3.13 a	6.74 ab	184 b	152 ab	1020 a
Bean	6.37 a	4.45 a	5.50 ab	2.63 a	7.30 a	220 a	161 a	1048 a
Fallow	6.15 b	4.81 a	4.92 ab	2.92 a	5.81 b	161 c	138 b	841 b
LSD	0.09	0.39	1.75	3.84	1.34	24	16	85

[y] Cover crop mixture consisted of eight crops (field peas, hairy vetch, crimson clover, cereal rye, annual ryegrass, oilseed radish, rapeseed, and buckwheat) planted together

[z] Values within columns followed by the same letter for each crop category are not significantly different from each other based on ANOVA and Fisher's protected LSD test ($P < 0.05$)

Table 4 Effect of previous cover crop treatments on selected soil microbiological properties in 2016, including average well color development (AWCD) from substrate utilization assays indicating microbial activity, culturable bacteria and fungal populations from soil dilution plating, and CO_2 respiration (Solvita test) indicating microbial biomass

Treatment	Soil biological properties			
	AWCD (optical density)	Bact. pops. ($\times 10^6$/g soil)	Fungal pops. ($\times 10^4$/g soil)	CO_2 resp. (mg/kg soil)
Mixture[y]	0.427 a[z]	55.8 a	33.6 a	127.0 a
Rye/vetch	0.447 a	57.8 a	37.2 a	117.4 ab
Mustard	0.440 a	61.4 a	34.1 a	118.1 ab
Fallow	0.374 b	50.2 b	34.6 a	110.3 b
LSD ($P = 0.05$)	0.036	5.6	4.0	12.6

[y] Cover crop mixture consisted of eight crops (field peas, hairy vetch, crimson clover, cereal rye, annual ryegrass, oilseed radish, rapeseed, and buckwheat) planted together

[z] Values within columns followed by the same letter for each parameter are not significantly different from each other based on ANOVA and Fisher's protected LSD test ($p < 0.05$)

soil (127.5 and 124.0 mg/kg CO_2 vs 111.4 for pepper and 80.6 for fallow soil, $p < 0.01$).

In spring 2017, following the 2016 cropping year, previous cover crop still showed some effects on soil properties, with all or most cover crops maintaining higher NO_3, K, Mg, and Ca concentrations than previously fallow soil (Table 5). Fallow soil also demonstrated lower concentrations of B, Na, S, Zn, and CEC than some or all the vegetable crops, as well as higher levels of Al and Fe (data not shown). The previous vegetable crop grown also was a significant factor on soil properties, with fallow soil averaging lower levels of NO_3, P, K, Mg, and Ca than following all vegetable crops, and lower organic matter content than following squash (Table 5).

Fallow soil also showed lower concentrations of B, S, Zn, and CEC than all vegetable crops and lower Fe, Mn, and Na concentrations than soil from squash plots (data not shown).

Table 5 Soil properties in Spring 2017 as affected by original cover crop treatments or previous year vegetable crop

	pH	Organic matter (%)	(mg/kg soil)					
			NO_3	NH_4	P	K	Mg	Ca
Cover crop								
Mixture[y]	6.23 a[z]	5.73 ab	14.0 a	1.67 a	13.8 a	244 a	185 ab	1245 a
Rye/vetch	6.22 ab	5.88 a	13.3 a	2.00 a	14.1 a	270 ab	194 a	1214 a
Mustard	6.21 ab	5.73 ab	13.9 a	1.33 a	14.7 a	286 a	190 a	1232 a
Fallow	6.13 b	5.41 b	10.6 b	1.58 a	11.7 a	250 b	169 b	1089 b
LSD	0.09	0.42	2.6	0.82	3.3	32	18	102
Previous crop								
Squash	6.26 a	5.86 a	11.9 b	1.50 a	12.9 a	261 a	193 a	1255 a
Pepper	6.16 b	5.76 ab	12.3 b	1.56 a	15.0 a	264 a	182 a	1190 ab
Bean	6.17 b	5.44 ab	14.8 a	1.88 a	12.7 a	263 a	178 a	1140 b
Fallow	6.25 a	5.42 b	8.0 c	1.83 a	7.5 b	214 b	160 b	1034 c
LSD	0.08	0.42	2.3	0.74	2.9	29	17	92

[y] Cover crop mixture consisted of eight crops (field peas, hairy vetch, crimson clover, cereal rye, annual ryegrass, oilseed radish, rapeseed, and buckwheat) planted together

[z] Values within columns followed by the same letter for each parameter are not significantly different from each other based on ANOVA and Fisher's protected LSD test ($P < 0.05$)

Table 6 Effect of previous cover crop treatments on early emergence of green bean and yellow squash seedlings in 2015 and 2016 plantings

Cover crop	Crop emergence (% of seeds planted)			
	Green bean		Yellow squash	
	2015 (7 DAP)	2016 (15 DAP)	2015 (7 DAP)	2016 (15 DAP)
Mixture[y]	75.9 a[z]	91.7 a	77.5 a	96.7 a
Rye/vetch	51.8 b	89.4 ab	70.8 a	99.2 a
Mustard	46.1 bc	86.2 bc	67.5 a	97.1 a
Fallow	42.0 c	83.9 c	54.2 b	99.2 a
LSD (P = 0.05)	7.3	3.5	12.1	2.6

[y] Cover crop mixture consisted of eight crops (field peas, hairy vetch, crimson clover, cereal rye, annual ryegrass, oilseed radish, rapeseed, and buckwheat) planted together

[z] Values within columns followed by the same letter for each parameter are not significantly different from each other based on ANOVA and Fisher's protected LSD test ($P < 0.05$)

Cropping effects on yield and disease

Cover crop treatment significantly affected crop emergence for bean and squash (since peppers were transplanted, no emergence data obtained). In beans, the cover crop mixture was associated with earlier and higher emergence than other cover crop treatments, and the fallow treatment was associated with the lowest emergence early in the season in both years (Table 6). The rye/vetch cover also resulted in earlier emergence than fallow soil in both years. For squash, all three cover crop treatments had higher emergence after 7 DAP in 2015, but there were no differences among cover treatments by 15 DAP in 2016 (Table 6).

Cover crop treatment significantly affected yield for all three vegetable crops in both production years. In 2015, the cover crop mixture and rye/vetch treatments were associated with higher green bean yields than fallow plots, with increases of 11–13%, and all cover crops increased total yield of yellow squash relative to fallow plots, lead by the cover crop mixture with a yield increase of 16% (Table 7). For sweet pepper, the cover crop mixture increased yield over all other treatments by 16–24%. In 2016, a previous cover crop of mustard or rye/vetch increased green bean yield by 7–12%, all cover crops increased yellow squash yield by 7–11%, and the cover crop mixture increased pepper yield by 9%, all relative to fallow plots (Table 7). When results from both years are combined, all three cover crops were associated with overall yield increases relative to fallow plots for green beans and yellow squash, by 6–

Table 7 Total vegetable yield of green bean, yellow squash, and sweet pepper crops as affected by previous cover crop in 2015 and 2016 growing seasons and previous vegetable crop in 2016

Previous crop	Total yield (Mg/ha)					
	Green bean		Yellow squash		Sweet pepper	
	2015	2016	2015	2016	2015	2016
Cover crop						
Mixture[y]	14.15 a[z]	14.83 bc	33.17 a	47.38 a	7.36 a	15.24 a
Rye/vetch	14.44 a	15.18 ab	32.16 a	46.40 a	6.27 b	14.51 ab 9.9
Mustard	13.50 ab	15.91 a	30.93 a	45.90 a	5.93 b	14.84 ab
Fallow	12.74 b	14.18 c	28.71 b	42.73 b	6.31 b	13.93 b
Vegetable crop						
Green bean		–		49.58 a		15.41 a
Yellow squash		15.84 a		–		13.93 b
Sweet pepper		14.58 b		43.03 b		–
Fallow		14.22 b		42.94 b		14.50 b

[y] Cover crop mixture consisted of eight crops (field peas, hairy vetch, crimson clover, cereal rye, annual ryegrass, oilseed radish, rapeseed, and buckwheat) planted together

[z] Values within columns followed by the same letter for type of crop type are not significantly different from each other based on ANOVA and Fisher's protected LSD test ($P < 0.05$)

10% and 7–13%, respectively (Fig. 2a, b). For pepper, only the cover crop mixture significantly increased yield overall relative to all other treatments, by 7–11% (Fig. 2c).

The vegetable crop rotation used also showed significant effects on yield in 2016. Green beans that followed squash averaged 8% higher yield than beans that followed peppers or fallow soil (Table 7). Yellow squash that followed green beans averaged 15% higher yield than those that followed pepper or fallow, and the pepper crop that followed beans averaged 11% higher yield than when followed squash or fallow (Table 7).

Although indications of disease development in all three crops were minimal and the only specific disease that was identified was powdery mildew on squash late in the season, there were some signs of late-season browning or leaf necrosis (etiology undetermined) in both beans and squash that may have been disease-related, and was measured as the overall percentage of leaf tissue affected per plot. Cover crop treatments of mustard and fallow resulted in the lowest levels of leaf necrosis on green beans in both 2015 and 2016, and the combined data for both years showed significantly less necrosis than either the cover crop mixture or rye/vetch, with reductions of 15–27% (Fig. 3a). On squash, leaf necrosis was lowest in the mustard and cover crop

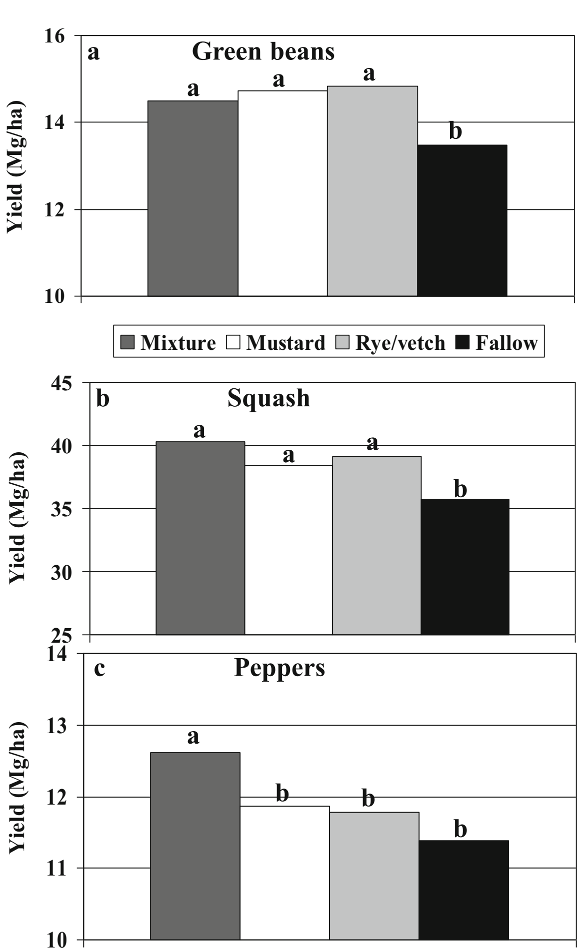

Fig. 2 Effect of cover crop treatments on total harvested yield of **a** green snap beans, **b** yellow summer squash, and **c** sweet lunchbox peppers averaged over two growing seasons (2015–2016). Bars topped by the same letter for each vegetable are not significantly different based on Fisher's protected LSD test ($P < 0.05$). Mixture = multi-species (8 crops) mixture; Mustard = mustard blend green manure; Rye/vetch = winter rye/hairy vetch combination; Fallow = no cover crop

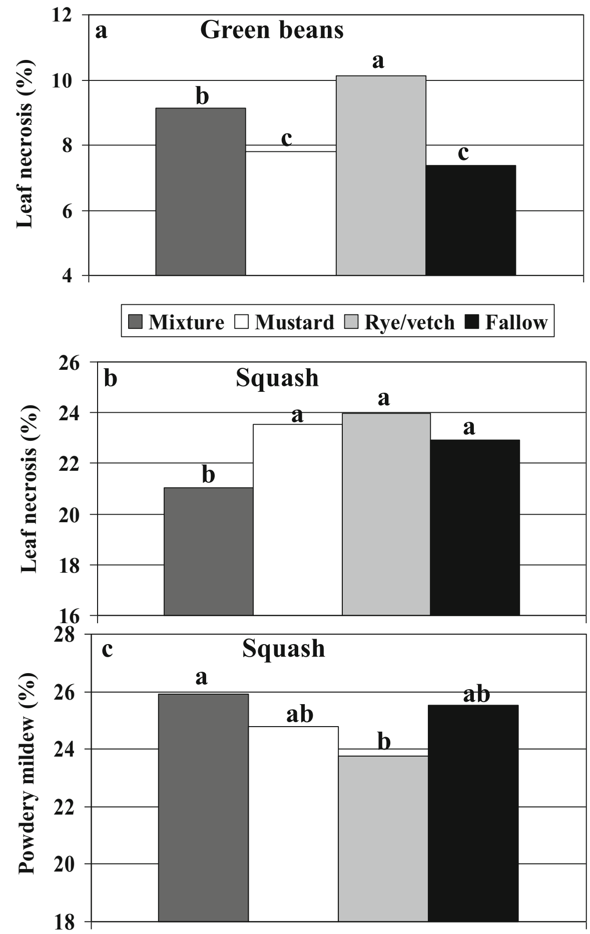

Fig. 3 Effect of cover crop treatments on disease development for **a** late season leaf necrosis on green snap beans, **b** late season leaf necrosis on yellow summer squash, and **c** powdery mildew on yellow summer squash averaged over two growing seasons (2015–2016). Bars topped by the same letter for each graph are not significantly different based on Fisher's protected LSD test ($P < 0.05$). Mixture = multi-species (8 crops) mixture; Mustard = mustard blend green manure; Rye/vetch = winter rye/hairy vetch combination; Fallow = no cover crop

mixture treatments in 2015, and fallow and cover crop mixture in 2016, with the combined data for both years showing reduced leaf necrosis for the cover crop mixture relative to all other cover crop treatments, with reductions of 8–12% (Fig. 3b). Powdery mildew on squash was lowest for rye/vetch in 2015 and showed no difference among cover crop treatments in 2016, but the combined data for both years indicated reduced mildew in the rye/vetch treatment relative to the mixture, by 8% (Fig. 3c).

Biological control effects on yield and disease

Both biocontrol treatments resulted in lower yield on green bean than the nontreated control in 2015, but not 2016 (Table 8), and the overall combined data for both years showed no overall effect on yield (Fig. 4a). On squash, biocontrol treatment BC1 resulted in higher yields in both 2015 and 2016, as well as in the overall combined data for both years, with an increase of ~ 7% (Table 8, Fig. 4b). On pepper, there were no differences in yield among biocontrol treatments in 2015, but BC1 resulted in lower yield than the other treatments in 2016, and overall results for both years combined also showed a slight reduction in yield for BC1, averaging 5% less than the nontreated control (Table 8, Fig. 4c). Biocontrol treatments did not have any significant effects on crop emergence in either year (data not shown).

Both biocontrol treatments reduced leaf necrosis on green bean in both 2015 and 2016, and when both years were combined, with average reductions of 24–28% compared to the nontreated control (Fig. 5a). On squash, BC1 reduced the severity of powdery mildew in both 2015 and 2016 relative to the nontreated control, as well as for both years combined, with an average reduction of 10% over both years (Fig. 5b). Although BC1 reduced the percentage of leaf necrosis relative to the nontreated control (by 15%) in 2015, there was no biocontrol effect in 2016, and the results from the combined data also showed no overall significant effect of the biocontrol treatments on leaf necrosis in squash (Fig. 5c).

Discussion

In addition to other factors, finding the right cover crop and rotation combinations are critical to establishing an effective, productive, and sustainable organic vegetable production system. In this research, different types of cover crops had some differing effects on soil properties and disease development across three vegetable crops, but overall, all three cover crops were superior to no cover crop (fallow) for most parameters. The vegetable rotation was also important, as the previous vegetable crop also significantly affected many parameters.

Overall, cropping effects on most soil chemical and nutritional properties were marginal, with relatively minor effects and changes observed due to cover or vegetable crop throughout the study. There was no difference observed in organic matter content between the cover crop soils and fallow, and interestingly, there was no apparent N benefit in the form of NO_3-N observed in the rye/vetch or cover crop mixture cropped soils. Overall, the most substantial effects were related to cropped vs. not cropped (fallow) soil, indicating that the presence of living plants and roots throughout the year is most important for improving overall soil health (as outlined by USDA-NRCS soil health management initiatives—http://www.nrcs.

Table 8 Total vegetable yield of green bean, yellow squash, and sweet pepper crops as affected by biological control treatments in 2015 and 2016 growing seasons

Biocontrol	Total yield (Mg/ha)					
	Green bean		Yellow squash		Sweet pepper	
	2015	2016	2015	2016	2015	2016
Control	14.49 a[z]	15.01 a	28.91 b	45.83 ab	6.59 a	14.68 a
BC1	13.27 b	14.97 a	33.23 a	47.13 a	6.38 a	13.81 b
BC2	13.39 b	14.92 a	31.59 ab	43.58 b	6.42 a	15.40 a
Average	13.72	14.97	31.24	45.52	6.46	14.63

[z] Values within columns followed by the same letter for each parameter are not significantly different from each other based on ANOVA and Fisher's protected LSD test ($P < 0.05$)

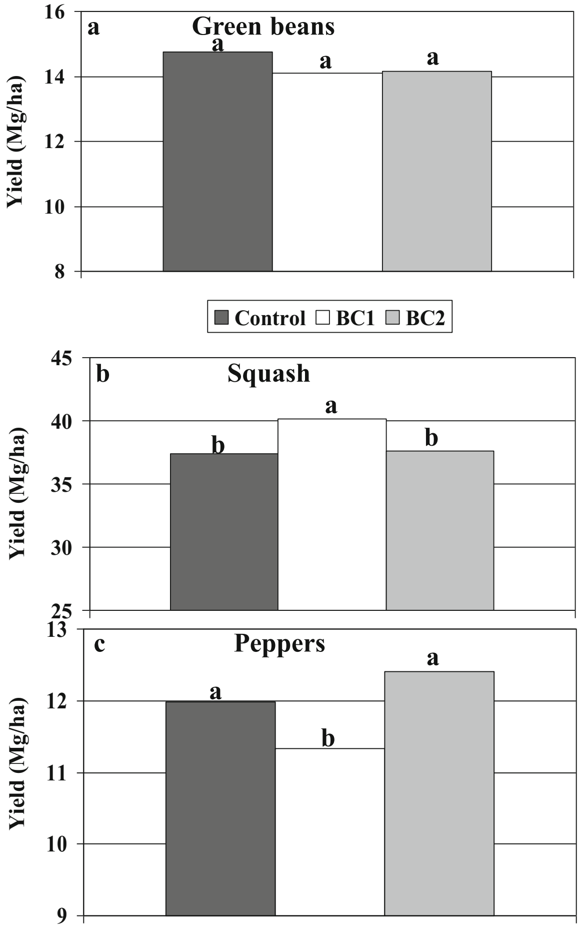

Fig. 4 Effect of biocontrol treatments on total harvested yield of **a** green snap beans, **b** yellow summer squash, and **c** sweet lunchbox peppers averaged over two growing seasons (2015–2016). Bars topped by the same letter for each vegetable are not significantly different based on Fisher's protected LSD test ($P < 0.05$). Control = no biocontrol treatment added; BC1 = combination of *Streptomyces lydicus* (Actinovate) and *Trichoderma virens* (SoilGard) added to soil at the time of planting; BC2 = combination of *Bacillus amyloliquefaciens* (Double Nickel) and *Trichoderma harzianum* (RootShield) added to soil at the time of planting

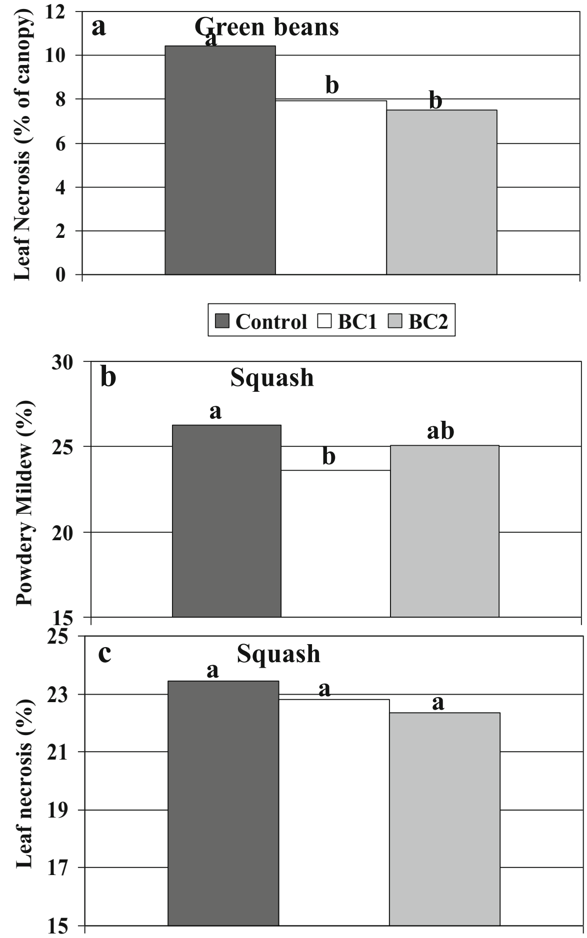

Fig. 5 Effect of biocontrol treatments on disease development for **a** late season leaf necrosis on green snap beans, **b** late season leaf necrosis on yellow summer squash, and **c** powdery mildew on yellow summer squash averaged over two growing seasons (2015–2016). Bars topped by the same letter for each graph are not significantly different based on Fisher's protected LSD test ($P < 0.05$). Control = no biocontrol treatment added; BC1 = combination of *Streptomyces lydicus* (Actinovate) and *Trichoderma virens* (SoilGard) added to soil at the time of planting; BC2 = combination of *Bacillus amyloliquefaciens* (Double Nickel) and *Trichoderma harzianum* (RootShield) added to soil at the time of planting

usda.gov/wps/portal/nrcs/main/soils/health/mgnt/). Although early effects on soil properties related to cover crop were minor, there were indications of increasing effects over time, as at the 2017 soil sampling at the end of the trial several parameters, including the concentrations of OM, NO_3, P, K, and Mg, increased for all cover crop systems relative to the initial values from the 2015 sampling. This is similar to results observed by others, in that changes in cover or rotation often have little effect on soil physical and chemical properties in the first year or two, as it generally takes several years before these types of changes have a substantial effect on soil properties, and the greatest effects generally require large additions of organic matter (Delate et al. 2008; Griffin et al. 2009; Pritchett et al. 2011). In a trial on organic peppers, Delate et al. (2003) also observed generally small effects due to cover crops and compost in the first few years of the study, but anticipated greater effects on soil health after several years. This has been borne out in longer-term studies, where cover crops have had more prominent effects over time (Sainju et al. 2002; Larkin et al. 2017). Generally, biological properties respond more quickly to cropping changes than soil physicochemical properties, and that was also observed in this study, with all three cover crops resulting in significant effects on microbial activity and bacterial populations, although soil respiration was only significantly affected by the multi-species mixture. Brennan and Acosta-Martinez (2017), in a long-term trial in an intensive commercial-scale organic vegetable operation, determined that whereas compost amendments had the greatest effect on soil organic C, it was cover cropping frequency more so than cover crop type that was the primary driver of changes in soil microbial biomass and composition.

Cropping effects were more apparent on yields for all three vegetable crops. In general, cover cropping was associated with greater yield than fallow soil. All three cover crops increased yield for bean and squash, whereas only the cover crop mixture increased yield for peppers. Thus, the multi-species mixture was the only cover crop that significantly increased yield in all three vegetable crops, with increases ranging from 7 to 14%. Although there were no clear indications in the soil properties measured, the presence of the multiple types of crops in the cover crop mixture may have improved soil conditions, resulting in earlier germination and yield benefits across all three crops. By 2016, in addition to effects due to cover crop treatments, vegetable rotation also affected yield with both squash and pepper showing a yield increase when following bean (11–15% increase) vs. the other vegetable, and bean yielding better following squash (8% increase). This potentially reflects nutritional or biological changes in the soil related to the previous crop. Better yield following bean would presumably be due to the potential N boost provided by the legume crop, but may also be related to other soil factors, such as soil microbial biomass. Soil CO_2 respiration was higher following either bean or squash relative to pepper or fallow in 2016. Based on these yield results, the better cover crop and rotation would be cover crop mixture and squash-bean-pepper rotation, to maximize yield opportunities, although all three cover crops performed equally well for most attributes. However, further study is required for verification, as these results are based on limited data in a short-term trial.

Yields observed were quite high for all crops (with the exception of the late-transplanted peppers in 2015) in both years, despite a shortened season in 2015 and drier than normal conditions in 2016. Our average yields (across all treatments) were 13.7 and 15.0 Mg/ha for beans in 2015 and 2016, respectively, which were well above the US national average of 9.5 Mg/ha for snap beans in 2016 (USDA NASS 2018b), and substantially above the New England and Maine averages of 3.8 and 3.5 Mg/Ha (USDA NASS 2018a). Our squash yields averaged 31.2 and 45.56 Mg/ha in 2015 and 2016, respectively, which were substantially greater than the US average of 18.8 Mg/ha for summer squash in 2016 (USDA NASS 2018b), and New England and Maine average of 11.5 and 9.5 Mg/ha (USDA NASS 2018a). For pepper, logistical problems and difficulty readying the plants for transplant lead to extreme late entry into the field in 2015 and a subsequent low yield average of 6.5 Mg/ha. In 2016, a more normal yield of 14.6 Mg/ha was observed. The New England average yield for "other" peppers in 2016 was of 9.2 Mg/ha (USDA NASS 2018a).

Little evidence or occurrence of either foliar or soilborne diseases, except for powdery mildew on squash late in the season, was observed through these studies. This was a field location that had not been cropped to any vegetable crops in recent years (last previously cropped to potatoes in 2008), and thus few disease problems. Although this was good for vegetable production, it did not allow adequate assessment of the efficacy of the biocontrol amendments in reducing disease. Both biocontrol treatments did reduce late season leaf necrosis on beans, although damage levels were generally low, potentially indicating a reduction in root-infecting organisms that may have contributed to late season decline. Biocontrol treatment BC1, which was a combination of *Streptomyces* and *Trichoderma* agents, also reduced powdery mildew on squash, but the reduction was somewhat marginal (10%) and, again, the overall disease levels were not very high. Thus, it is not clear whether the biological controls would have provided disease control if more disease was present.

The biocontrol treatments had varying effects on yield. On beans, although there was no overall significant effect on yield, the biocontrol treatments were associated with lower yields in 2015. This may indicate that in the absence of disease pressure (mitigated by the biocontrols), there could be a slight yield reduction due to the biocontrol amendments. Treatment BC1 increased yield on squash in both years. It is doubtful that the 10% reduction in powdery mildew was responsible for the observed increase in yield (most harvests occurred before powdery mildew appeared). However, overall growth improvement provided by the biocontrol may have been responsible. Previous research has indicated both induced resistance and growth promotion in these agents, and that may account for the increased yield (Yuan and Crawford 1995; Sharma et al.2017). On pepper, BC2, which was a combination of *Bacillus* and *Trichoderma*, produced the highest yield in 2016 and over both years, but was not significantly greater than the control, only greater than BC1. These agents have also been observed to provide some induced resistance to disease and plant growth promotion in previous studies (Sharon et al. 2011; Li et al. 2015). So, overall results from the biocontrol amendments were inconclusive, there was not enough disease present to adequately assess disease management potential, and yield effects varied depending on the crop. Overall, in the absence of any substantial disease pressure, there does not appear to be any advantage or benefit in the application of these biological control agents, and in some cases (bean, BC1 on pepper) could depress yield. Previous research has indicated that biological control treatments are affected by different cropping systems and rotations and disease control may be enhanced or diminished depending on interactions with different rotations (Larkin 2008). Several studies have indicated that combining biocontrol isolates, particularly those having different mechanisms of action, can provide improved performance (Guetsky et al. 2001, 2002; Roberts et al. 2005).

This research confirmed the feasibility of using a paper mulch weed barrier for organic production, as it worked well through the entire growing season in both years, with no substantial breakdown or problems. The paper held up well with minimal intrusions and no weed issues through the entire season. The paper mulch retained soil moisture better than outside the mulch during dry periods, and soil temperatures were not appreciably affected by the paper mulch (no difference under or outside paper). Most previous studies assessing paper mulches used variations on kraft paper or shredded newspaper and forms that may not have been specifically designed for agricultural use (Miles et al. 2007; Coolong 2010; Haapala et al. 2014). The paper barrier used in this study seemed to be of a higher quality and specifically made for this purpose, producing favorable results.

This research assessed the potential benefits of different types of cover crops and biocontrol amendments within a diversified organic vegetable production system. Although the results are tentative and subject to additional testing in longer-term trials, several interpretations can be made. Of greatest importance was the presence or inclusion of a cover crop in the system, as all three cover crops contributed to significant benefits in improving soil properties and yield relative to no cover crop (fallow) for all vegetable crops in this system. With that said, the multi-species mixture did perform somewhat better overall than the other two covers for several parameters, including earlier emergence and improved yield in all three vegetable crops. Due to low disease levels throughout the trial, no conclusions regarding the efficacy or utility of the biocontrol amendments could be made. However, in the absence of notable disease problems, there did not seem to be any justification for including them in the system, as they provided no significant yield or quality benefit overall. The vegetable rotation used also affected yield, with squash and pepper following bean associated with improved yield and bean following squash providing an improvement over pepper. In addition, the use of a paper mulch weed barrier was feasible in this system, resulting in no weed issues, not interfering with production, and allowing high yields in all vegetable crops. These results emphasize the importance of cover crops and cropping sequence and provide specific management practices and approaches to help improve the sustainability and productivity of organic vegetable production.

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