

# Influence of cover crops on arthropods, free-living nematodes, and yield in a succeeding no-till soybean crop



Alan W. Leslie<sup>a,\*</sup>, Koon-Hui Wang<sup>b</sup>, Susan L.F. Meyer<sup>c</sup>, Sharad Marahatta<sup>d</sup>, Cerruti R.R. Hooks<sup>a</sup>

<sup>a</sup> Department of Entomology, University of Maryland, College Park, MD, United States

<sup>b</sup> Department of Plant and Environmental Protection Sciences, University of Hawaii, Manoa, HI, United States

<sup>c</sup> United States Department of Agriculture, Agricultural Research Service, Mycology and Nematology Genetic Diversity and Biology Laboratory, Beltsville, MD, United States

<sup>d</sup> Kauai Community College, University of Hawaii, Lihue, HI, United States

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## ABSTRACT

Production practices that incorporate fall-planted cover crops into no-till agronomic crop rotations have become increasingly popular across the Northeastern United States for weed suppression and enhancing environmental stewardship. Field experiments were conducted in 2011 and 2012 to investigate effects of rotating cereal (barley, *Hordeum vulgare*), legume (Austrian winter pea, *Pisum sativum* subsp. *arvense*), cereal/legume cover crop mixture, and a fallow (bare-ground) control on above- and belowground fauna in a succeeding soybean crop. Free-living nematodes and soybean foliar arthropods were sampled through time to determine effects of cover crops on soil food web structure and complexity and herbivorous and beneficial arthropods, respectively. Our hypotheses were that organic matter from cover crop biomass would provide energy and nutrients to the soil food web and that increased habitat complexity from cover crop residue would provide habitat for more predatory arthropods aboveground. In general, cover crops in this no-till system had a stronger influence on the below- than aboveground fauna. There was no consistent, positive effect of cover crops on beneficial foliar arthropods or on soybean yield. Cover crops increased the soil food web structure and complexity as determined by nematode community indices. Specific effects of different cover crop types on the free-living nematode community varied within the growing season and between study years. Probable causes for differences encountered among cover crop treatments and years are discussed.

## 1. Introduction

Cover cropping has long been used as a practice for reducing soil erosion, increasing soil quality, and suppressing weeds (Colla et al., 2000; Sainju and Singh, 1997; Teasdale, 1996; Yenish et al., 1996). Cover crops have a history of also being used as green manures and providing animal feed during periods of drought (Lu et al., 2000). With concerns over environmental stewardship and sustainability issues, cover crops have become of greater interest for their potential to provide additional agroecosystem services. Cover crops can impact several factors within a cropping habitat contemporaneously such as influencing soil quality, health and fertility, water quality, above- and belowground organisms, and crop yield. For example, cover crops have been shown to increase soil organic matter, reduce carbon loss, improve soil structure and tilth, conserve nitrogen resources, and suppress weeds in a wide range of cropping systems (Hartwig and Ammon, 2002). In addition, integration of cover crops into vegetable crops can

increase natural enemies and reduce pest insect abundances (Hinds and Hooks, 2013; Hooks et al., 2013) and influence the health of neighboring ecosystems (Snapp et al., 2005). However, different cover crops can enhance, decrease, or have no effect on yield, arthropods, and other organisms associated with cropping systems according to how they are managed. As such, predicting their influence on a cropping system can be challenging.

Over the years, the adoption of conservation practices in the form of cover crops has increased sharply in the Northeastern US. Cover crops are currently being grown on hundreds of thousands of hectares of arable land throughout this area as part of soil conservation plans. In 2015, cover crops were planted on a total of 199,204 ha in Maryland alone as part of the Maryland Department of Agriculture's Cover Crop Program (MDA, 2016). Producers can receive from 62 to 235 USD per ha to grow winter cover crops in the state of Maryland, which is enough to cover most or all expenses required to grow most cover crops (Pelton, 2010). In the Northeastern US, winter cover crops are planted in early

Abbreviations: AWP, Austrian winter pea; BG, bare ground; MI, Maturity index; EI, Enrichment index; SI, Structure index; CI, Channel index; F/B, Fungivore to bacterivore ratio

\* Corresponding author.

E-mail address: [aleslie@umd.edu](mailto:aleslie@umd.edu) (A.W. Leslie).

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fall and generally allowed to grow until mid-spring, at which time they are incorporated by tillage or killed and left as a surface mulch into which a crop is planted. In Maryland, soybean (*Glycine max*) producers typically follow a corn (*Zea mays*)-soybean rotation scheme and plant cover crops in the fall after corn is harvested, then terminate the cover crop with a post-emergent herbicide prior to planting a soybean crop.

Though soybean producers grow cover crops mainly for soil conservation benefits, cover cropping can modify the fauna of arthropods within field crop systems and these changes may differ according to the amount of cover crop residue (Smith et al., 1988) and species of cover crop grown. For example, herbivorous arthropod pests and associated plant damage were reduced in soybean plots which contained rye (*Secale cereale*) cover crops prior to planting (Koch et al., 2015). Conversely, rye cover crops can increase pest abundance within the following corn (*Zea mays*) crop, if they serve as a host for polyphagous pest insects (Dunbar et al., 2016). Fall-planted grass cover crops have been shown also to increase natural enemy abundances in corn and soybeans (Hooks et al., 2011; Lundgren and Fergen, 2010).

In addition to influencing organisms above the soil surface, residues from killed cover crops cover the soil surface and release organic carbon and nutrients that provide subsidies to the soil food web, which can alter soil biodiversity (Hooks et al., 2011; Lundquist et al., 1999; Norris et al., 2016; Quintanilla-Tornel et al., 2016). Free living nematodes have often been used as indicators of how land management practices (e.g., tillage, cover cropping, solarization, etc.) in agroecosystems impact biodiversity below the soil. This is because nematodes are ubiquitous, well classified into functional groups, and functionally diverse (Bongers and Bongers, 1998). Further, they are easy to sample and play an important role in soil nutrient cycling. Free-living nematodes directly influence soil processes and reflect the structure and function of many other taxa within the soil food web (Ferris et al., 2001). Their food chain ranges from fast-growing, fast-breeding, bacteria-feeding nematodes at the bottom (colonizers) to slow-growing, slow-reproducing predatory nematodes (persisters) at the top of the food web. Further, nematode communities respond readily to changes in soil physical and chemical conditions and have a direct linkage to ecological processes (Neher, 2010). Some nematodes can survive disturbed environments better than others and some have short life cycles and respond rapidly to environmental changes (e.g., colonizers). Thus, they can provide an early sign of how crop husbandry practices are affecting soil organisms (Hinds et al., 2013; Wang and McSorley, 2005).

Specific effects of cover crops on above- and belowground communities can vary depending on what types of species are used and how they are managed (Gill et al., 2011; House and Alzugaray, 1989). For example, functional groups of plants (i.e., legumes, forbs, grasses) have dissimilar rooting patterns that create habitats more congenial to some species of nematodes than others (Neher, 2010). Djigal et al. (2012) found that grass and legume cover crops increased populations of beneficial nematodes in a banana plantation compared to bare ground, but grass cover crops supported lower populations of plant-feeding nematodes while legume cover crops supported greater numbers of predacious nematodes. In addition to physical features, biochemical compositions of cover crops such as carbon to nitrogen ratio (C:N) will influence the pattern and timing of mineral decomposition and N release of cover crop residues (Kuo and Sainju, 1998). This will in turn mediate the activity of decomposers including the free-living nematode fauna. Barley, *Hordeum vulgare*, and Austrian winter pea, *Pisum sativum* subsp. *arvense* are popular grass (Poaceae) and legume (Fabaceae) winter cover crop species, respectively, in the US. Barley has a relative high C:N ratio and can produce a tremendous amount of biomass in a short time compared to several other grass cover crops and has a thick root system, which can improve soil structure and water infiltration (Creamer et al., 1996; Overland, 1966). Austrian winter pea (AWP) is shallow-rooted and slower to establish than barley, but is capable of rapid biomass accumulation (Norsworthy et al., 2010) and because of

high N content, it can readily provide N to subsequent crops (Mahler and Auld, 1989). Barley has been found to inhibit weed emergence through allelopathic chemicals and by providing a physical barrier to weed establishment (Creamer et al., 1996; Overland, 1966). Though AWP is capable of producing a large amount of biomass, it is prone to rapid decay, and as such is not known for providing prolonged surface mulch and weed suppression benefits (Norsworthy et al., 2010).

Limited research has been conducted to investigate impacts of winter cover crops jointly on above- and belowground communities of organisms (Hooks et al., 2011). However, the fact that cover crops can influence these organisms concurrently warrants concerted investigations on these disparate communities. Barley and AWP are often grown as winter cover crops, especially in the northeastern US, and agricultural producers in Maryland can receive incentive payment to plant barley and barley/AWP mixes. As such, if it is determined that these cover crops can have a positive influence on beneficial organisms within the crop field, this will provide an ecological incentive for producers to plant these cover crops as a standard land management practice. Thus, the objective of this study was to compare effects of cereal (barley), legume (AWP), and cereal/legume mixture (barley + AWP) fall planted cover crop and no cover crop (fallow) treatment on the above- and belowground fauna within a subsequent soybean crop. The specific focus was on communities of herbivorous pests and beneficial arthropods within the soybean canopy and free-living nematodes below the soil surface. Our hypotheses were that the additional organic inputs from cover crops and their resulting residue would lead to an increase in the complexity of the soil food web belowground, and the added habitat structure provided by the cover crop would lead to a larger population of predacious arthropods aboveground.

## 2. Materials and methods

### 2.1. Experimental site, treatment, and plot layout

Field trials were conducted in 2011 and 2012 in separate fields (2011: 39.0252°, −76.8412°; 2012: 39.0121°, −76.8314°) at the University of Maryland Beltsville Research and Education Center in Beltsville, MD. Different fields were used in each year of the experiment to follow standard farming practices for the area; farmers rarely plant soybeans in the same field two years in a row. Soils for this study were mesic Aquic Hapludults, and were mapped within the Russett-Christiana complex (Soil Survey Staff, 2016). The soil texture ranged from fine sand to loamy sand with mineral fractions ranging from 81.1 to 88.7% sand, 7.1 to 14.1% silt, and 3.5 to 4.8% clay. Prior to initiation of the experiment, the field site for the 2011 trial was rye (*Secale cereale*) followed by soybean that was mowed prior to reaching maturity, whereas the field for the 2012 trial was planted with wheat (*Triticum aestivum*). Both field sites had been under no-till practices for several years. No-till cropping is defined as planting directly into the residue of the previous crop without performing any tillage operations that disturb the soil prior to planting (Stubbs et al., 2004). The entire experimental area was 0.45 ha with four treatments, arranged in a randomized complete block design, with four replications. Each treatment plot measured 11 m × 12 m, and was separated by 7 m of bare-ground between plots. The four treatments were soybeans planted into (1) AWP, (2) barley, (3) AWP + barley mixture, or (4) bare-ground (BG). Cover crops were planted on 9 October 2010 and 19 September 2011 for the 2011 and 2012 field trials, respectively. Barley and AWP were planted at 112 kg seeds/ha, whereas AWP + barley mixture was planted at 44.8 and 67.2 kg seeds/ha of AWP and barley, respectively. Plant biomass was collected from four 0.1 m<sup>2</sup> quadrats randomly placed in each plot just prior to cover crop termination. Samples were dried at 65 °C and weighed. Subsets of biomass samples were ground into powder (< 1 mm) and analyzed for total C and N (A & L Laboratories, Memphis, TN). Soil total C and N were also measured from each plot at

**Table 1**

Cover crop (CC) C:N, biomass and total nitrogen (TN) across treatments in 2011 and 2012, indicating organic C and N input differences between treatments. AWP = Austrian winter pea, B = barley, BG = bare ground.

Year	CC trt	Soil C:N		CC C:N	CC biomass (Mg/ha)	CC TN (kg/ha)
		Pre-plant	Pre-harvest			
2011	AWP	3.5 (0.5) aA <sup>†</sup>	3.1 (0.3) aA	17.2 (1.2) c	5.2 (0.7) a	118.4 (17.4) a
	AWP + B	2.8 (0.3) aA	2.6 (0.4) aA	25.6 (2.7) c	5.5 (0.6) a	88.7 (16.0) a
	B	3.4 (0.7) aA	2.4 (0.5) aA	61.1 (4.0) a	5.0 (0.4) a	32.6 (4.4) b
	BG <sup>‡</sup>	3.3 (0.9) aA	3.0 (0.4) aA	39.0 (2.7) b	1.8 (0.5) b	17.8 (4.9) b
2012	AWP	5.3 (0.6) aA	4.0 (0.7) aB	11.6 (0.4) b	1.1 (0.5) a	37.6 (15.6) a
	AWP + B	5.0 (0.4) aA	4.3 (0.3) aB	23.4 (1.9) ab	2.2 (0.5) a	38.1 (8.2) a
	B	4.8 (0.3) aA	3.5 (0.3) aB	33.4 (6.5) a	2.0 (0.6) a	26.0 (8.7) a
	BG	5.0 (0.7) aA	4.0 (0.7) aB	19.5 (3.8) b	1.2 (0.6) a	32.7 (23.7) a

<sup>†</sup> Means (n = 4) followed by the same lower case letter are not different among treatments within each year (Tukey-Kramer adjusted  $P > 0.05$ ). Means (n = 4) followed by the same upper case letter are not different between sampling dates ( $P > 0.05$ ) in the linear mixed model.

<sup>‡</sup> BG values are measured from winter-germinated weeds.

**Table 2**

Summary output of linear mixed models testing the effects of cover crop treatment, sampling period, and their interaction on nematode community indices from 2011 and 2012 field seasons.

Year	Parameter	Treatment			Sampling Period			Treatment × Sampling Period		
		Wald- $\chi^2$	DF	P	Wald- $\chi^2$	DF	P	Wald- $\chi^2$	DF	P
2011	Richness	2.13	3	0.55	26.78	2	< 0.001	4.88	6	0.56
	Diversity	7.98	3	0.05	1.63	2	0.44	2.96	6	0.81
	FG Div <sup>†</sup>	4.29	3	0.23	0.39	2	0.82	7.52	6	0.28
	F/B	1.77	3	0.62	4.11	2	0.13	13.77	6	0.03
	MI	7.29	3	0.06	31.31	2	< 0.001	8.79	6	0.19
	EI	3.53	3	0.32	12.93	2	0.002	16.69	6	0.01
	SI	5.77	3	0.12	14.79	2	0.001	7.98	6	0.24
	CI	0.75	3	0.86	12.75	2	0.002	17.55	6	0.01
2012	Richness	1.55	3	0.67	7.42	2	0.02	14.44	6	0.03
	Diversity	7.27	3	0.06	3.99	2	0.14	18.24	6	0.01
	FG Div.	4.45	3	0.22	53.77	2	< 0.001	30.51	6	< 0.001
	F/B	43.41	3	< 0.001	29.89	2	< 0.001	49.79	6	< 0.001
	MI	45.64	3	< 0.001	16.75	2	< 0.001	20.72	6	0.002
	EI	49.97	3	< 0.001	474.98	2	< 0.001	5.91	6	0.43
	SI	33.58	3	< 0.001	40.20	2	< 0.001	16.91	6	0.01
	CI	6.91	3	0.07	1161.44	2	< 0.001	47.13	6	< 0.001

<sup>†</sup> FG Div = feeding guild diversity, F/B = abundance of fungivorous/bacterivorous nematodes, MI = Maturity index, SI = Structure index, CI = Channel index.

the beginning (pre-plant) and end (pre-harvest) of each experiment to determine whether different cover crop treatments affect soil C:N and to calculate N contribution of residue from each treatment following decomposition.

All plots were mowed using a flail mower (John Deere 25 A, Moline, IL) two days prior to soybean planting. Soybean line '92Y51' (DuPont Pioneer, Johnston, IA) was planted on 25 May 2011 and 26 May 2012 using a no-till coulters (John Deere Corn Planter Model 7200, Moline, IL) at 56,656 seeds per ha with a 76 cm inter-row spacing. During the study, Z-Glyphosate 41% Max (Fuzion Technologies, LLC., Fort Collins, CO) was applied at a rate of 15.6 mL/L of water to terminate the cover crop and any germinated weeds following planting. Yield was estimated by harvesting all rows in each plot with a small-plot combine.

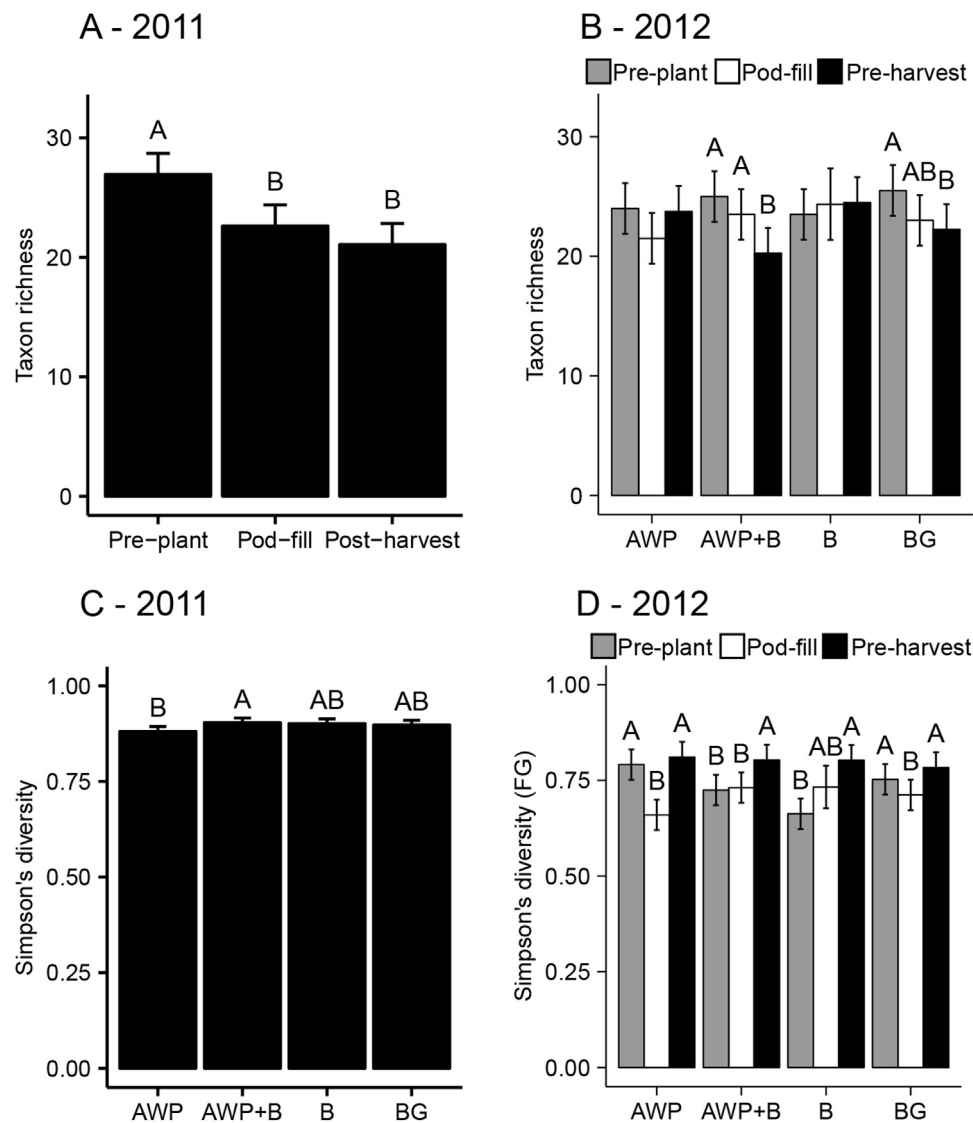
## 2.2. Nematode sampling

After initiation of the experiment, each treatment plot was sampled at pre-planting shortly before cover crops were mowed (24 May 2011 and 25 May 2012), mid-season during the pod-filling period (22 August 2011 and 26 July 2012), and at soybean maturity just prior to soybean harvest (04 October 2011 and 01 October 2012). At each sampling, 20 soil cores (2.5 cm diameter × 20 cm deep) were collected randomly from each plot with a soil auger from the intra-row area approximately 10 cm from soybean plants. Soil cores collected from each plot were discharged into a bucket, thoroughly mixed, and a 100 cm<sup>3</sup> subsample

was taken for nematode faunal analysis. Subsamples were placed into a Ziploc plastic bag and transferred to the laboratory for nematode extraction. Nematodes were extracted using sieving and centrifugal flotation methods (Jenkins, 1964). All nematodes were identified and recorded to the genus level whenever possible except for individuals in the family Rhabditidae, which were identified to family level under an inverted microscope (DM IL LED, Leica Microsystems, Wetzlar, Germany).

## 2.3. Nematode community analysis

Free-living nematodes were assigned to one of the six trophic groups: algivores, bacterivores, fungivores, herbivores, omnivores, or predators (Yeates et al., 1993). The feeding habit of Tylenchidae (mainly *Filenchus* and *Tylenchus*) was classified as fungivore (McSorley and Frederick, 1999; Okada and Kadota, 2003). *Prismatolaimus* was grouped as a bacterivore in this study as opposed to substrate ingestor suggested by Yeates et al. (1993). Total numbers of each trophic group in the community were calculated. Nematode richness was calculated as the total number of taxa recorded per sample. Simpson's index of diversity (Simpson, 1949) was calculated as  $1/\lambda$ , where  $\lambda = \sum (p_i)^2$ , and  $p_i$  is the proportion of each of the  $i$  taxa. Specimens identified only to the order level were excluded from this calculation. The fungivore to bacterivore (F/B) ratio was calculated to characterize decomposition and mineralization pathways (Freckman



**Fig. 1.** Effects of cover crop treatments (AWP = Austrian winter pea, B = barley, BG = bare ground) and sampling periods on (A & B) taxon richness, (C) Simpson's diversity, and (D) Simpson's diversity of nematode feeding guilds in 2011 and/or 2012. Bars labeled with different letters indicate significant differences (Tukey-Kramer adjusted  $P \leq 0.05$ ) among pairwise contrasts of (A) sampling periods, (C) cover crop treatments, or (B & D) sampling period within cover crop treatment. Error bars = 95% CI.

and Ettema, 1993). Total maturity index (MI) as defined by Yeates (1994) was calculated as  $\sum (p_i c_i)$ , where  $c_i$  is the colonizer-persister (c-p) rating of taxon  $i$  according to the 1 to 5 c-p scale of Bongers and Bongers (1998). Simpson's diversity was also calculated for nematode functional guilds as represented by combinations of trophic group and c-p rating. The nematode fauna was also analyzed by a weighting system for nematode functional guilds in relation to enrichment and structure of the soil food web. The enrichment index (EI) assesses soil food web responses to available resources, and structure index (SI) reflects the degree of trophic connection in soil food webs of increasing complexity as the system matures (Ferris et al., 2001). These indices were calculated as  $EI = 100 \times [e/(e + b)]$  and  $SI = 100 \times [s/(s + b)]$  where  $e$ ,  $s$ , and  $b$  are enrichment, structure, and basal food web components calculated as suggested by Ferris et al. (2001). They are the sums of weighted abundance of nematodes in guilds representing those components, whereas the channel index (CI) represents the decomposition pathway in the soil food web, calculated as  $CI = 100 \times [0.8Fu_2/(3.2Ba_1 + 0.8Fu_2)]$ , where  $Fu_2$  and  $Ba_1$  are the numbers of fungivores with a c-p rating of 2 and bacterivores with a c-p rating of 1, respectively. In general, higher EI and lower CI suggest greater bacterial activity.

#### 2.4. Foliar sampling of pests and beneficial arthropods

Soybean foliage arthropods were sampled weekly with the use of a 38 cm diameter sweep net for relative population estimates. A sample consisted of two sets of five sweeps, each in separate soybean rows. Two samples were taken per treatment plot. Arthropod collections were transferred into sealed Ziploc plastic storage bags and temporarily stored on ice in a portable cooler then stored in a freezer in the laboratory for later species identification and recording. Sampling commenced on 21 July 2011 and 03 July 2012, and was terminated on 05 September 2011 and 15 August 2012, respectively, following soybean crop senescence. For analysis, foliar arthropods were first divided broadly into pests (plant-feeding taxa) and beneficials (predacious and parasitic taxa) and subsequently divided into different feeding guilds. These groups were parasitoids, chewing predators, sucking predators, spiders, chewing herbivores, sucking herbivores, and plant-feeding stink bugs. Stink bugs were analyzed separately because of the economic importance of the brown marmorated stink bug *Halyomorpha halys* and other stink bugs as soybean pests in the Northeastern US.



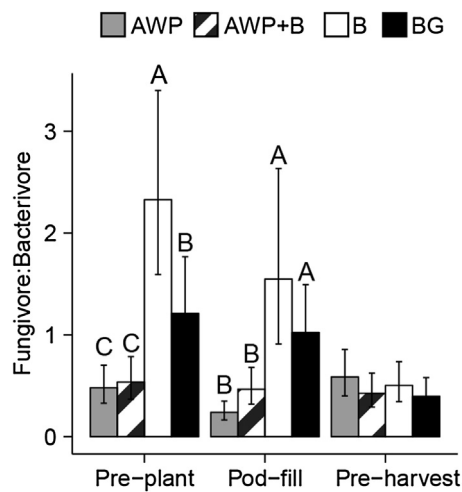


Fig. 2. Effects of cover crop treatments (AWP = Austrian winter pea, B = barley, BG = bare ground) on fungivorous to bacterivorous nematode ratios (F/B) at three sampling periods in 2012. Means of treatment ( $n = 4$ ) at each sampling period labelled with different letters indicate significant differences among pairwise contrasts of treatment (Tukey-Kramer adjusted  $P \leq 0.05$ ). Error bars = 95% CI.

## 2.5. Statistical analysis

Linear mixed-effects models (LMMs) were used to test for differences in soil and cover crop C:N, cover crop biomass, and total N contribution of cover crops among cover crop treatments. A random factor was included in the model to account for the blocked design and the repeated measurement of the soil C:N at the beginning and the end of the growing season. Data were analyzed separately by year.

Nematode community indices were analyzed using LMMs to test for differences among cover crop treatments and sampling dates. A random factor was included in the model to account for the blocked treatment design and the repeated measurement of the nematode community over time. Data were analyzed by year. Plots of residuals were checked for homogeneity of variances and normal distribution. Log-transformations were used whenever necessary to correct for deviations from normality. When either sample date or treatment effect was significant ( $P < 0.05$ ) based on the LMM, all combinations of pairwise comparisons were made for levels of the significant factor using Tukey-adjusted  $P$  values. When the LMM indicated a significant interaction term along with a significant main effect, separate LMMs were run for each level of the non-significant factor. When both main effects and their interaction were significant, only the effects of cover crop treatment were tested separately at the levels of sample date.

Generalized linear mixed models (GLMM) were used to test for differences in abundances of feeding guilds of foliar arthropods between cover crop treatments. Abundances were pooled across weekly samples. A random factor was included in the model to account for the blocked treatment design. A Poisson distribution with a log-link function was used to account for the error distribution of the count data for the foliar arthropods. When the GLMM indicated a significant difference between treatment means, the same contrasts were performed as described above.

Linear mixed models were used to test for differences in yield between cover crop treatments. A random factor was included in the model to account for the blocking structure of the experiment. Separate analyses were performed for each year of the study.

All statistical analyses were performed using the program 'R' v. 3.2.3 (R Core Team, 2016). Linear mixed effects models and generalized linear mixed effects models were built using the package 'lme4' (Bates et al., 2015). Post-hoc means comparisons were performed using the package 'multcomp' (Hothorn et al., 2008). Treatment means were extracted from models using the package 'effects' (Fox, 2003).

## 3. Results

### 3.1. Cover crops

Differences in cover crop biomass and C:N values are summarized in Table 1. There was no effect of cover crop treatment or sampling period on soil C:N in 2011. In 2012 there was a significant effect of sampling period (Wald- $\chi^2 = 96.33$ ,  $df = 1$ ,  $P < 0.001$ ), with pre-harvest having a lower C:N than pre-plant, but there was no significant effect of treatment. Significant differences in the C:N of different treatments were observed in 2011 (Wald- $\chi^2 = 144.63$ ,  $df = 3$ ,  $P < 0.001$ ) and 2012 (Wald- $\chi^2 = 21.33$ ,  $df = 3$ ,  $P < 0.001$ ). In 2011, barley had the highest C:N, and BG had higher C:N than AWP and AWP + barley. In 2012, barley had greater C:N than AWP and BG. Although not analyzed statistically, there was a large difference in cover crop biomass production between study years, with 2011 having mean biomass values 2.5 to 4.5 times greater than 2012. There were significant differences in biomass produced by different treatments in 2011 (Wald- $\chi^2 = 63.44$ ,  $df = 3$ ,  $P < 0.001$ ) but not 2012 ( $P = 0.08$ ). In 2011, all cover crop treatments had greater biomass than BG with no significant differences among individual cover crop treatments. Differences in biomass and C:N presumably resulted in different amounts of total N released following residue decomposition in different treatments in 2011 (Wald- $\chi^2 = 82.03$ ,  $df = 3$ ,  $P < 0.001$ ) but not 2012. In 2011, the extrapolated N released from AWP and AWP + barley were greater than barley and BG treatments (Table 1).

### 3.2. Nematode community

In 2011 and 2012, 54 and 43 nematode taxa were collected, respectively (see Appendix in Supplementary material). Summaries of outputs from LMM of indices calculated from nematode community data are presented in Table 2. Cover crop treatment had no effect on taxon richness in either study year, but there was a significant effect of sampling period in 2011 (Wald- $\chi^2 = 26.78$ ,  $df = 2$ ,  $P < 0.001$ ) and 2012 (Wald- $\chi^2 = 7.42$ ,  $df = 2$ ,  $P = 0.02$ ) and the interaction between sampling period and treatment was significant (Wald- $\chi^2 = 14.44$ ,  $df = 6$ ,  $P = 0.03$ ) in 2012. In 2011, nematode richness was higher at pre-plant than pod-fill and pre-harvest (Fig. 1a). The BG and AWP + barley treatments showed a similar pattern in 2012, where richness was lower during pre-harvest than pre-plant. In the AWP + barley treatment, there was no difference between pre-plant and pod-fill, and in the BG treatment, pod-fill did not differ significantly from pre-plant or pre-harvest (Fig. 1b). There was no significant effect of sampling period on richness in barley, while sampling period was almost significant in AWP ( $P = 0.053$ ).

There was a significant effect of treatment (Wald- $\chi^2 = 7.98$ ,  $df = 3$ ,  $P = 0.05$ ) but not sampling period on diversity in 2011 (Fig. 1c). In 2011, AWP had lower diversity compared to AWP + barley. There was no effect of cover crop or sampling period on diversity in 2012. In addition, there was no effect of cover crop or sampling period on diversity of nematode feeding guilds in 2011. In 2012, there was a significant effect of sampling period (Wald- $\chi^2 = 53.77$ ,  $df = 2$ ,  $P < 0.001$ ) and the interaction of sampling period and treatment (Wald- $\chi^2 = 30.51$ ,  $df = 6$ ,  $P < 0.001$ ) on diversity of functional guilds (Fig. 1d). In AWP and BG treatments, diversity of functional guilds was significantly higher at pre-plant and pre-harvest than pod-fill. In AWP + barley, diversity was significantly greater during pre-harvest than pre-plant and pod-fill. In the barley treatment, diversity was significantly greater at pre-harvest than pre-plant while diversity at pod-fill was not significantly different from either other sampling period. Consequently, the overall trend was greater diversity of functional guilds at pre-harvest regardless of values earlier in the season.

There was no effect of sampling period or cover crop treatment on the fungivore to bacterivore ratio (F/B) in 2011. In 2012, sampling

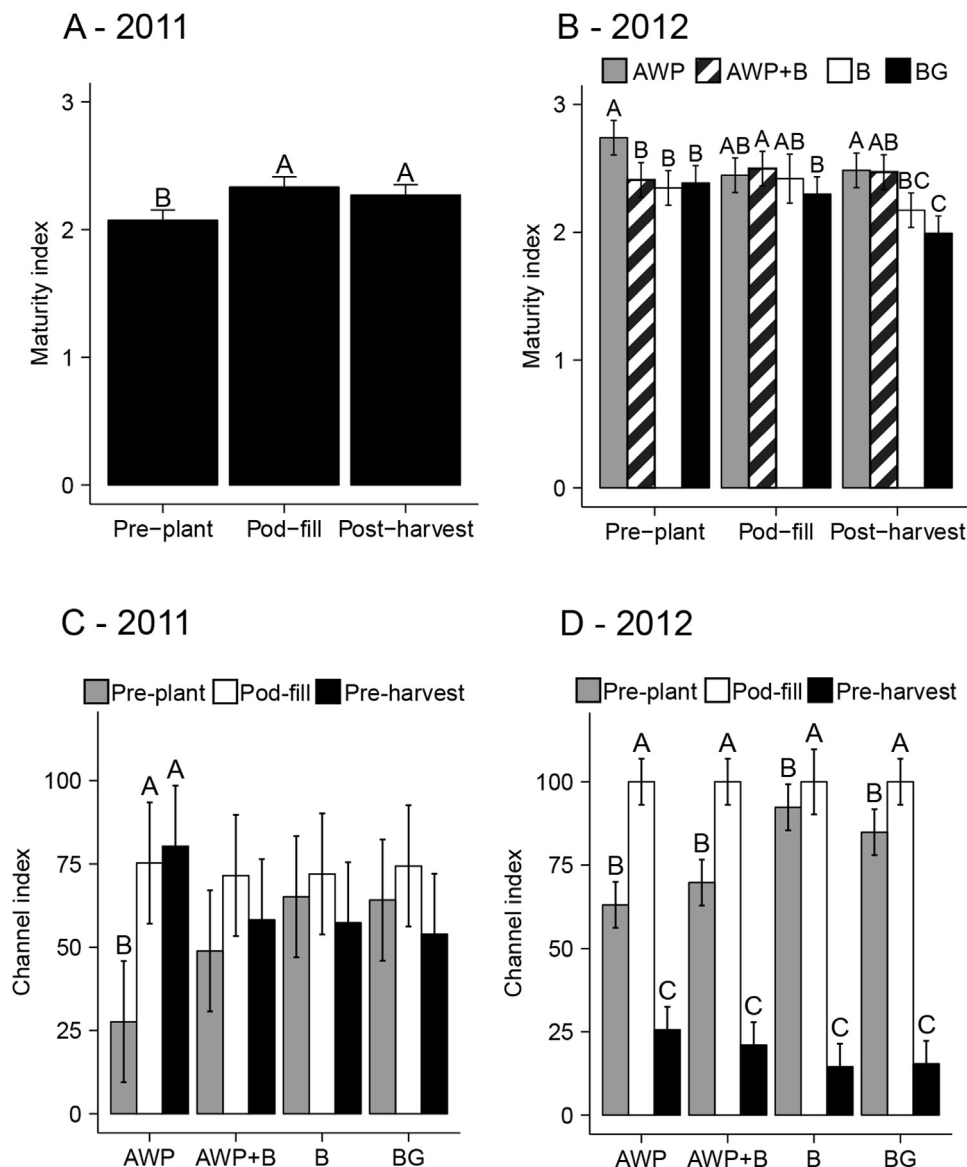


Fig. 3. Effects of cover crop treatments (AWP = Austrian winter pea, B = barley, BG = bare ground) and sampling periods on (A & B) maturity index and (C & D) channel index of nematodes. Bars labeled with different letters indicate significant differences (Tukey-Kramer adjusted  $P \leq 0.05$ ) among pairwise comparisons of (A) sampling period, (B) cover crop treatment within sampling periods, or (C & D) sampling period within cover crop treatment. Error bars = 95% CI.

period (Wald- $\chi^2 = 29.89$ ,  $df = 2$ ,  $P < 0.001$ ), treatment (Wald- $\chi^2 = 43.41$ ,  $df = 3$ ,  $P < 0.001$ ), and their interaction (Wald- $\chi^2 = 49.79$ ,  $df = 6$ ,  $P < 0.001$ ) had significant effects on F/B. At pre-plant, barley had a significantly higher F/B than other treatments, and BG had a higher F/B than AWP and AWP + barley (Fig. 2). At pod-fill, BG and barley had higher F/B values than AWP and AWP + barley (Fig. 2). There was no significant effect of cover crop treatment on F/B at pre-harvest.

There was a significant effect of sampling period (Wald- $\chi^2 = 31.31$ ,  $df = 2$ ,  $P < 0.001$ ) on maturity index (MI) in 2011 and sampling period (Wald- $\chi^2 = 16.75$ ,  $df = 2$ ,  $P < 0.001$ ), treatment (Wald- $\chi^2 = 45.64$ ,  $df = 3$ ,  $P < 0.001$ ), and their interaction (Wald- $\chi^2 = 20.72$ ,  $df = 6$ ,  $P = 0.002$ ) in 2012 (Fig. 3a, b). In 2011, MI at pre-plant was significantly lower than at pod-fill or pre-harvest. In 2012, AWP had higher MI than all other treatments at pre-plant (Fig. 3b). At pod-fill, AWP + barley had higher MI than BG, and at pre-harvest, AWP and AWP + barley had higher MI values than BG, and AWP had higher MI values than barley.

Sampling period (Wald- $\chi^2 = 12.75$ ,  $df = 2$ ,  $P = 0.002$ ) and the interaction of sampling period and treatment (Wald- $\chi^2 = 17.55$ ,

$df = 6$ ,  $P = 0.01$ ) had significant effects on the channel index (CI) in 2011 (Fig. 3c). For AWP, CI was significantly lower at pre-plant than pod-fill or pre-harvest. There were no significant differences among sampling periods for other treatments. In 2012, there was also a significant effect of sampling period (Wald- $\chi^2 = 1161.44$ ,  $df = 2$ ,  $P < 0.001$ ) and the interaction of sampling period and treatment (Wald- $\chi^2 = 47.13$ ,  $df = 6$ ,  $P < 0.001$ ) on CI (Fig. 3d). Channel index values differed among treatments during each sampling period. For all treatments, CI increased from pre-plant to pod-fill and then decreased to lowest values at pre-harvest.

There was a significant effect of sampling period (Wald- $\chi^2 = 12.93$ ,  $df = 2$ ,  $P = 0.002$ ) and the interaction of sampling period and treatment (Wald- $\chi^2 = 16.69$ ,  $df = 6$ ,  $P = 0.01$ ) on the enrichment index (EI) for 2011 (Fig. 4a). Significant differences among sampling periods were only found in AWP. Within AWP, pre-plant had higher values for EI than pod-fill or pre-harvest. In 2012, there were significant main effects for sampling period (Wald- $\chi^2 = 474.98$ ,  $df = 2$ ,  $P < 0.001$ ) and treatment (Wald- $\chi^2 = 49.97$ ,  $df = 3$ ,  $P < 0.001$ ) (Fig. 4b). Post-hoc comparisons of main effects show that EI decreased from the pre-plant to pod-fill and then increased at pre-harvest, and that AWP and

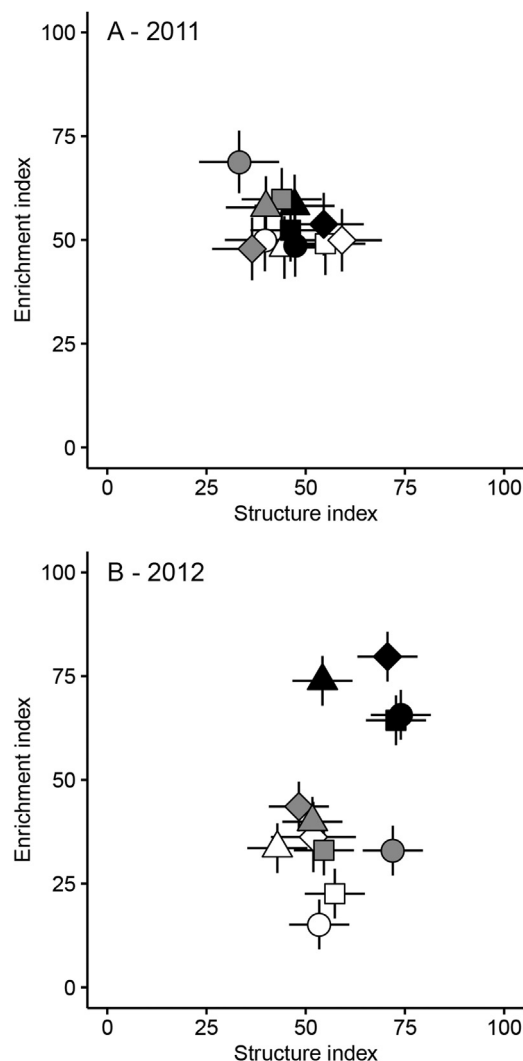


Fig. 4. Mean values of Enrichment index plotted against mean values of Structure index for 2011 and 2012 as calculated from nematode sampling. Color represents sampling period: gray = pre-plant, white = pod-fill, black = pre-harvest. Shapes represent cover crop treatment: circle = AWP, square = AWP + B, diamond = B, triangle = BG. Error bars = 95% CI.

AWP + barley had lower EI values than barley and BG.

There was a significant effect of sampling period ( $\text{Wald-}\chi^2 = 14.79$ ,  $\text{df} = 2$ ,  $P = 0.001$ ) on structure index (SI) in 2011 (Fig. 4a). Structure index was significantly lower at pre-plant than pod-fill or pre-harvest. In 2012, sampling period ( $\text{Wald-}\chi^2 = 40.20$ ,  $\text{df} = 2$ ,  $P < 0.001$ ), treatment ( $\text{Wald-}\chi^2 = 33.58$ ,  $\text{df} = 3$ ,  $P < 0.001$ ), and their interaction ( $\text{Wald-}\chi^2 = 16.91$ ,  $\text{df} = 6$ ,  $P = 0.01$ ) had a significant effect on SI (Fig. 4b). At pre-plant, AWP had higher SI than other treatments. At pod-fill, AWP + barley had higher SI than the BG treatment, and at the post-harvest period, BG had lower SI than other cover crop treatments.

### 3.3. Foliar arthropods and yield

In 2011, 11 arthropod taxa were collected from seven weeks of sweep samples, with a total of 549 individuals (Table 3). In 2012, 24 arthropod taxa were collected from six weeks of sweep samples, with a total of 1449 individuals (Table 3). The most abundant pest species in 2011 were the brown marmorated stink bug *Halyomorpha halys* and the green cloverworm *Hypena scabra*, which together accounted for 32.4% of the foliar arthropod community. The main pests in 2012 were *H. scabra* and plant bugs in the family Miridae, which together accounted for 32.7% of the foliar arthropod community. In both years, main

predators by abundance in the foliar arthropod community were spiders and big-eyed bugs in the genus *Geocoris*, which together accounted for 43.7% (2011) and 24.4% (2012) of the total foliar arthropod community. Three groups within the foliar arthropod community were influenced by cover crop treatment. In 2011, cover crop treatment had a significant effect on sucking predator abundance ( $\text{Wald-}\chi^2 = 14.66$ ,  $\text{df} = 3$ ,  $P = 0.002$ ). Means comparisons showed that AWP + barley had significantly more sucking predators than the barley treatment (Fig. 5a). In 2011, cover crop also had a significant effect on spider abundance ( $\text{Wald-}\chi^2 = 9.04$ ,  $\text{df} = 3$ ,  $P = 0.03$ ), with significantly more spiders in BG than AWP + barley (Fig. 5b). In 2012, total predators showed a significant response to cover crop treatment ( $\text{Wald-}\chi^2 = 9.50$ ,  $\text{df} = 3$ ,  $P = 0.02$ ). There were significantly more predacious arthropods in AWP plots than the AWP + barley plots (Fig. 5c).

There was no effect of cover crop treatment on soybean yield in 2011 or 2012 (data not shown). Mean soybean yield in 2011 was 1013.4 kg/ha and in 2012 was 2787.6 kg/ha. Total precipitation was 584 mm during the 2011 growing season, and 326 mm during the 2012 growing season. There was minimal rainfall during the early part of the 2011 growing season, with less than 10 mm of rain recorded during the first 21 days after planting. In 2012, approximately 53 mm of rain had accumulated over the same amount of time.

## 4. Discussion

Overall results from this study supported the hypothesis that winter cover crops could affect above- and belowground organisms in a subsequent soybean crop. However, effects of cover cropping were more profound on the nematode fauna than the arthropod community within the soybean foliage. Nematodes are diverse fauna, represent a wide range of functional groups, and are sensitive to land management practices (Wang and McSorley, 2005). Thus, quantification of the free-living nematode community provides an indication of how the soil biota and food web respond to crop husbandry practices (Bongers and Bongers, 1998). As such, it was not unforeseen that they would be more responsive to varying cover crop types than the arthropod community. Still, responses of the nematode fauna to cover crop types varied over time within seasons and between study years, and the cover crop effect was stronger in 2012 than 2011, despite lower cover crop biomass production.

Cover crops were anticipated to add sources of nutrients and energy to the soil that would subsequently support higher microbial activities (Dabney et al., 2011; Lehman et al., 2015) and nematodes were anticipated to respond positively to this comestible energy source. Generally, the nematode communities on arable land planted with cover crops are expected to reflect a more diverse fauna indicative of a more resilient, healthier soil community compared to bare-ground fields, which are more emblematic of early successional habitats (Gruver et al., 2010; Wang et al., 2011). This generally occurs because bare-ground or fallow fields add limited organic material to the soil that can be utilized as a source of food and energy. Results from the 2012 experiment supported the hypothesis regarding beneficial effects of cover crops on the soil food web. However, findings in 2011 indicate a neutral response to cover cropping despite having much greater cover crop biomass in 2011 than 2012. The winter of 2011/2012 was unusually warm, and unseasonably warm temperatures may have caused cover crops to break dormancy before the final frost date, which would have caused an overall reduction in growth relative to the 2010/2011 winter.

Lack of cover crop effects on nematode communities in 2011 could be due to the cropping history of that field, which was preceded by a soybean crop in 2010. Relatively high EI ( $\geq 50\%$ ) in all treatments including BG at pre-plant in 2011 indicate a nutrient enrichment condition throughout the entire field. This could have been caused by the soybean plant residues that were left on the field after mowing, with

**Table 3**

Foliar arthropod total abundances and proportions, calculated from sweep net samples of all treatments.

Common name	Taxonomic group	Feeding guild	Abundance	Proportion
2011				
Spider	Araneae	Spider	180	0.328
Brown marmorated stink bug	<i>Halyomorpha halys</i>	Sucking herbivore	93	0.169
Green cloverworm	<i>Hypena scabra</i>	Chewing herbivore	85	0.155
Big-eyed bug	<i>Geocoris</i> spp.	Sucking predator	60	0.109
Minute pirate bug	<i>Orius</i> spp.	Sucking predator	47	0.086
Brown stink bug	<i>Euschistus</i> sp.	Sucking herbivore	25	0.046
Lady beetle	Coccinellidae	Chewing predator	22	0.040
Caterpillar	Lepidoptera	Chewing herbivore	10	0.018
Green lacewing	Chrysopidae	Chewing predator	10	0.018
Green stink bug	<i>Chinavia hilaris</i>	Sucking herbivore	9	0.016
Damsel bug	<i>Nabis</i> spp.	Sucking predator	8	0.015
2012				
Green cloverworm	<i>Hypena scabra</i>	Chewing herbivore	280	0.193
Plant bug	Miridae	Sucking herbivore	194	0.134
Spider	Araneae	Spider	183	0.126
Big-eyed bug	<i>Geocoris</i> spp.	Sucking predator	171	0.118
Japanese beetle	<i>Popillia japonica</i>	Chewing herbivore	94	0.065
Threecornered alfalfa hopper	<i>Spissistilus festinus</i>	Sucking herbivore	55	0.038
Brown marmorated stink bug	<i>Halyomorpha halys</i>	Sucking herbivore	54	0.037
Damsel bug	<i>Nabis</i> spp.	Sucking predator	52	0.036
Spotted cucumber beetle	<i>Diabrotica undecimpunctata</i>	Chewing herbivore	40	0.028
Ground beetle	<i>Lebia</i> spp.	Chewing predator	39	0.027
Minute pirate bug	<i>Orius</i> spp.	Sucking predator	35	0.024
Green stink bug	<i>Chinavia hilaris</i>	Sucking herbivore	33	0.023
Parasitoid wasp	Hymenoptera	Parasitoid	30	0.021
Caterpillar	Lepidoptera	Chewing herbivore	27	0.019
Green lacewing	Chrysopidae	Chewing predator	26	0.018
Mexican bean beetle	<i>Epilachna varivestis</i>	Chewing herbivore	23	0.016
Click beetle	Elateridae	Chewing herbivore	21	0.014
Pigweed flea beetle	<i>Disonychia glabrata</i>	Chewing herbivore	18	0.012
Lady beetle	Coccinellidae	Chewing predator	17	0.012
Cereal leaf beetle	<i>Oulema melanopus</i>	Chewing herbivore	17	0.012
Brown stink bug	<i>Euschistus</i> sp.	Sucking herbivore	15	0.010
Assassin bug	Reduviidae	Sucking predator	12	0.008
Blister beetle	<i>Epicauta</i> spp.	Chewing herbivore	7	0.005
Spined soldier bug	<i>Podisus maculiventris</i>	Sucking predator	6	0.004

no nutrient removal from harvesting. This may explain why the subsequent cover crop treatments did not further improve the soil condition in 2011. Berkelmans et al. (2003) also reported that after a uniform management with a healthy oat cover crop for one year, the differences in nematode community indices in conventional cropping versus organic agricultural systems for the previous seven years disappeared.

In 2012, the structure index (SI) and to a lesser extent, maturity index (MI) of at least one cover crop treatment was higher compared to the BG control throughout the three sampling periods. Although AWP took the lead in increasing SI initially, B and AWP + B resulted in similar SI as AWP towards the soybean pre-harvest period. Increased MI and SI indicate the nematode community has become more structured and populated by nematodes with longer persistency (Ferris, 2001). A more structured soil food web indicates more linkages in the soil food web, which can provide more ecosystem services such as soil nutrient mineralization and pest suppression by predatory nematodes (Sylvain and Wall, 2011). DuPont et al. (2009) did not find an effect of cover crops on SI within one cropping cycle in no-till system. The current study was conducted in fields that had been under no-till agriculture for several years, which may have allowed the colonization of nematode guilds with higher colonizer-persister (c-p) values (Freckman and Ettema, 1993). Shifting of nematode communities to higher c-p values indicate a shift from species adapted to colonizing disturbed habitats (lower c-p values) to longer-lived species that compete well in more stable habitats (Bongers and Bongers, 1998). The SI and MI values in B and AWP + B may not have responded significantly to cover crop treatments during the pre-plant sampling period in 2012 because their higher C:N materials are slower to be decomposed by bacteria and fungi

right after the cover crop is terminated (Wang et al., 2004). However, low C:N of AWP lead to a faster decomposition rate and resulted in higher SI values than other treatments at pre-plant sampling in 2012. Management practices that reduce or eliminate soil cultivation within crop fields and maintain vegetation throughout the year will favor agricultural soils that more closely resemble soils of natural ecosystems, which are in favor of establishment of free-living nematodes (Neher, 1999; Fu et al., 2000; Mendoza et al., 2008).

Cover crop species with different C:N are predicted to impact the belowground community discordantly (Gruver et al., 2010; Sylvain and Wall, 2011). As a legume, Austrian winter pea (AWP) has lower C:N than barley. Differences in the stoichiometry of the breakdown products of legumes or grasses would provide different basal resources for the soil food web (Waggoner et al., 1998). This effect was evident in the ratio of fungivore to bacterivore (F/B) nematodes in 2012 but not 2011. In 2012, at the pre-plant and pod-fill stages, the barley treatment had significantly higher F/B than the AWP or AWP + barley treatments. This indicates a prevalence of fungal decomposition in the barley treatment. An increase in fungal decomposition is often associated with crop residues with higher C:N (Lundquist et al., 1999). AWP produced lower C:N residues than barley, which resulted in lower F/B at pre-plant and at pod filling. Although CI and F/B are similar indices, the cover crop treatments did not affect CI in this study. CI is the abundance of fungivores with c-p 2 divided by the same fungivores plus bacterivores with c-p 1 value, whereas F/B is total fungivores divided by total bacterivores. The CI did not differ among cover crop treatments in 2012 possibly due to the exclusion of fungivores with c-p values above 2 and bacterivores with c-p values above 1 in the calculation.

As expected, AWP provided greater total N to plots than barley and



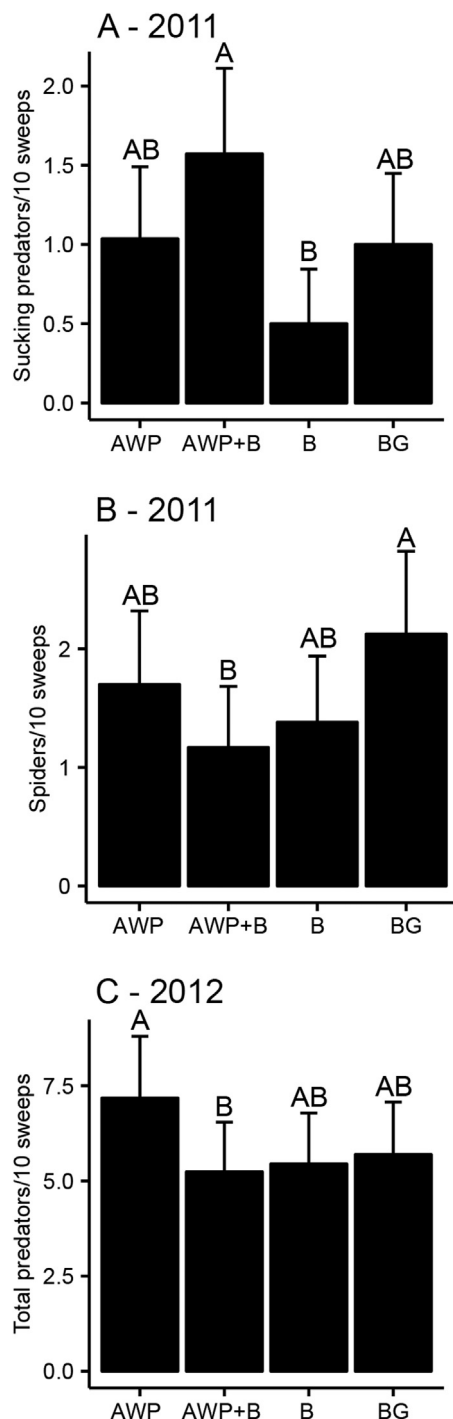


Fig. 5. Effects of cover crop treatments (AWP = Austrian winter pea, B = barley, BG = bare ground) on mean abundance of foliar arthropod (A) sucking predators in 2011, (B) spiders in 2011, and (C) total predators in 2012 between cover crop treatments. Different letters indicate significant differences (Tukey-Kramer adjusted  $P \leq 0.05$ ) from among pairwise comparisons of levels of cover crop treatments. Error bars = 95% CI.

BG in 2011, and AWP was associated with higher EI only during the pre-plant period. In contrast, EI was lower in AWP and AWP + barley treatments compared to barley throughout the three sampling periods in 2011. This is most likely because AWP and AWP + barley did not increase total N addition to plots in 2012. Patterns in EI values could also be related to overall cover crop biomass as reported by DuPont et al. (2009). In 2011, CI was also lower at pre-plant than pod-fill or pre-harvest for the AWP treatment, indicating increased bacterial decomposition in response to higher N levels with a legume cover crop

at the pre-plant sampling period. These results indicate a transient effect of soil N enrichment by a legume cover crop on nematode functional group composition (Wang et al., 2004).

In general, cover crop treatments did not have a significant effect on the foliar arthropod community. Abundances of sucking predators and spiders in 2011 and total predators in 2012 were the only groups to respond differently to cover crop treatments. In 2011, spiders and sucking predators had similar abundances, but showed nearly the opposite patterns in abundance between cover crop treatments. Spider abundance was lower overall in the cover crop treatments than BG, although the only significant comparison was between AWP + barley and BG. This result is inconsistent with other studies that show a positive response of spiders to cover crops (Hooks et al., 2011; Nyffeler and Sunderland, 2003). Overall, there was no consistent response of arthropod predators to cover cropping. Similarly, other studies have found variable responses of foliar arthropods to cover crop presence (Bottenberg et al., 1997; Koch et al., 2012; Masiunas et al., 1997; Smith et al., 1988). The lack of a consistent effect in this study may be due in part to the low number of arthropods encountered during sampling events. Further, cover crops remained on the soil surface as organic mulch for several weeks after soybeans were planted. As such, sampling the epigeal (ground dwelling) arthropod community as opposed to the foliar community may have revealed more notable effects of cover crops on aboveground arthropods (House and Alzugaray, 1989). In addition, cover crops may be of conservation value by serving as overwintering quarters for natural enemies that eventually move into the soybean canopy. As such, measuring the abundance of soil-hibernating arthropods at spring emergence could help elucidate how different cover crop systems impact the abundance and diversity of overwintering arthropods.

Overall, this study provides direct evidence that planting winter cover crops as part of a no-till farming operation can have positive effects on belowground organisms during one cropping cycle; and that the response of organisms within the cropping system may differ depending on cover crop functional group. However, there was no cover crop effect on yield. The absence of an effect of cover crops on soybean yield is consistent with findings in other studies (Hooks et al., 2011; Koch et al., 2012; Reddy et al., 2003; Ruffo et al., 2004). Benefits of cover crops to soybean yields may vary with environmental conditions, or may not occur until several continuous years of cover cropping (Dabney et al., 2001; Sainju et al., 2002). The lack of a consistent effect of cover crop presence on above- and belowground organisms and no effect on yield may be indicative of field history. Each of the study sites has a history of no-till practices, and reducing the frequency of cultivation is one primary method to increase trophic diversity in arable soils (Hendrix et al., 1986). The nematode trophic structure should improve under long term no-till conditions as all trophic groups may become more diverse under no-till regimes (Fu et al., 2000). As such, it is conceivable that the prevalence and history of no-till practices at both study sites partially negated the cover crop treatments and subsequent influence on the nematode fauna as well as yield. Thus, it is conjectured that if field studies were conducted on lands that were subjected to more frequent disturbances (e.g., from tillage or heavy equipment), with identical land management histories and similar environmental conditions occurred during both years, cover crop treatment influence on the nematode fauna would have been more marked and less variable. Though there was a lack of consistency, findings suggest that there may still be some benefits of cover cropping on the community of organisms within a no-till soybean production system.

In conclusion, this study provides further evidence of the benefits of cover cropping in no-till agronomic systems. Contrary to finding from other studies, cover crop residue did not consistently increase the abundance of beneficial arthropods or reduce the abundance of herbivorous arthropods in the soybean canopy. Rather, the main effect of incorporating cover crops into a corn-soybean rotation in this study is

greater diversity and complexity of free-living nematodes, indicating enhanced cycling of energy and nutrients belowground. The specific effects of cover crops on the soil food web likely depend on environmental contexts. Effects of cover crops on the soil food web will need to be evaluated across a broader range of environmental conditions and cover crop species before more specific conclusions can be made. Nonetheless, the results of the soil health analysis from this study support previous findings that the benefits of cover cropping on below ground communities are more apparent if the soil was not previously enriched with nutrients.

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## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at <http://dx.doi.org/10.1016/j.apsoil.2017.04.003>.

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