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Ecosystem services of the soil food web after long-term application of agricultural management practices

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ABSTRACT

The structure of soil nematode assemblages was assessed in field plots in the San Joaquin Valley of California which have 16-year management system histories. Attributes of the ecosystem functions of the assemblages were determined in laboratory studies. The four agricultural management systems were no tillage (minimum tillage) with cover crops in the intervals between economic crops, standard tillage with cover crops, minimum tillage without cover crops and standard tillage without cover crops. The economic crops were sorghum and garbanzo beans. A soil column system was used in laboratory studies to evaluate the nitrogen mineralization ecosystem service associated with nematode assemblages in soils from the four management systems compared to that in defaunated soil. In an additional comparison, defaunated soil was amended with mineral fertilizer solution for comparison with the mineralization service of the soil fauna. Management systems using cover crops, which created a continuity of both photosynthetic production and roots in the soil, strongly enhanced the nematode assemblages in the field soil. Management systems with cover crops had greater total abundance, measured as numbers, biomass and metabolic footprints, of nematodes, and also of the functional guilds of nematodes considered important in soil fertility and as prey for predators. Leachates from soil columns with intact nematode assemblages had greater total mineral nitrogen and supported greater plant growth than those from defaunated columns. Soil carbon levels in field plots were strongly affected by the management systems. The biomass and diversity-weighted footprint of bacterivore and microbivore (bacterivores plus fungivores) nematodes, in turn, were correlated with levels of soil carbon.

1. Introduction

Ecosystem services provided by soil organisms include the cycling of mineral nutrients and the regulation of pest species; they are important contributors to the availability of plant nutrients and to supporting food and fiber production (de Vries et al., 2013; Ciobanu et al., 2015). The below-ground decomposer system provides the basis for soil fertility through recycling plant material and mineralizing soil nutrients. The rates and magnitudes of these ecosystem services are determined by the composition, abundance and diversity of the soil biota (Ruess and Ferris, 2004; de Vries et al., 2013; Ferris and Tuomisto, 2015).

Soil nematodes, as a major mesofauna component of soil food webs, play important roles in ecosystem functioning, including organic material decomposition, nutrient turnover and energy transfer (Hättenschwiler et al., 2005; Wall et al., 2012; Coleman and Wall, 2015). Among soil nematodes, microbivores (bacterivores and fungivores) are key intermediaries in decomposition processes and nutrient cycling; they increase bacterial turnover and accelerate decomposition of soil organic matter (Bardgett and Chan, 1999; Neher, 2001; Yeates, 2003). Also, nematodes transport bacteria to new resources, further accelerating the rates and magnitude of decomposition activity (Brown et al., 2004; Fu et al., 2005). Since microbivore nematodes ingest more of certain nutrients than required, the excesses are excreted in a mineral or readily-mineralizable form, and thus may enhance plant growth (Ferris et al., 1997; Ingham et al., 1985; Neher et al., 2012). Specialist predator and omnivore nematodes are also involved in nutrient cycling through a more indirect process of predation on microbial grazers and herbivore species (Ferris et al., 2012a; Holtkamp et al.,

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2011).

Besides their direct contributions to ecosystem services, nematodes are also useful bioindicators of the abundance and activity of other soil organisms providing similar services (Ferris et al., 2012b; Georgieva et al., 2005; Sánchez-Moreno et al., 2008, 2009; Yeates et al., 2009). Based on their abundance and turnover rates, soil nematodes can account for up to 25% of nitrogen mineralization in the soil (Ferris et al., 2012a). Metabolic footprints of nematode functional guilds, and the diversity of species comprising the guilds, provide a quantitative assessment of the magnitude of ecosystem functions (Ferris, 2010; Ferris et al., 2012c; Ciobanu et al., 2015; Ferris and Tuomisto, 2015; Zhang et al., 2015).

Soil ecosystem services, supported by organic resources, are the foundation for agricultural production systems that do not rely on high inputs of mineral fertilizer or synthetic pesticides. Agricultural management practices impact soil organic matter levels and, directly and indirectly, the biomass and diversity of soil nematodes, with consequent effects on nutrient cycling (Balota and Filho, 2004; DuPont et al., 2009; Sánchez-Moreno et al., 2009). They also affect the resilience of soil food webs and ecosystem functions (Zhang et al., 2013, 2016). Most previous research has been focused on the effect of agricultural management practices on the structure and composition of soil nematode assemblages (Li et al., 2009; Sánchez-Moreno et al., 2009; Zhang et al., 2012, 2016). But what is the relationship between the functions and ecosystem services of the faunal assemblage and its structure? There are voids in our validation of the relationships between the structure and functions of ecosystems. The soil provides wonderful opportunities for examining such relationships without the cost of landscape-scale experimentation.

We analyzed the effects of agricultural management strategies on soil nematode assemblages in a field experiment and conducted laboratory studies to determine the relationship between structure of those assemblages and the associated ecosystem services as reflected in soil fertility and plant growth. We hypothesized that: 1) after 16 years of different agricultural practices, the soil ecosystems at the field site had transitioned to new states; 2) the resultant differences in nematode assemblages and soil food webs in soils under different management systems would be reflected in rates of nutrient mineralization; and 3) that mineral fertilizer applications can be reduced in management systems in which the soil food web is providing adequate plant nutrition.

2. Materials and methods

2.1. Field experiment

2.1.1. Experimental site

The field study site is located at the University of California's West Side Research and Extension Center in Five Points, CA (36°20′29″N, 120°7′14″W). The mean maximum and minimum annual air temperature in the area is 24 °C and 8 °C, respectively. Annual precipitation is about 180 mm. The soil is classified as Panoche clay loam (fine-loamy, mixed superlative, thermic Typic Haplocambids) (Veenstra et al., 2007). A field comparison of conservation and standard tillage, with and without cover crops between the economic crops, was established in 1999 (16-year management history as of the end of 2015; Mitchell et al., 2017). The farming systems in this experiment are described in detail elsewhere (Veenstra et al., 2007). The only soil disturbance operations in the conservation tillage systems were shallow cultivation during establishment of a tomato (*Solanum lycopersicum* L.) crop, used as the economic crop, during the first eight years of the experiment. In subsequent years, sorghum (*Sorghum bicolor* (L.) Moench) or garbanzo beans (*Cicer arietinum* L.) have been used as economic crops. In 2012, the conservation tillage treatments became true no-tillage systems with the only soil disturbance occurring at the time of seeding in the sorghum plots or garbanzo plots. Consequently, herein that treatment is referred to as no tillage which was its condition during the current study.

The field experiment is in a randomized complete block design with four replicates. Management treatments include two factors, i.e. tillage level and presence or absence of cover crops in the intervals between economic crops. The four treatments are no tillage and cover crops (NTCC), standard tillage and cover crops (STCC), no tillage and no cover crops (NTNO) and standard tillage and no cover crops (STNO). The treatment philosophies are that cover crops maximize the time during which there are roots in the soil and resources available to the soil food web while no tillage minimizes disruption of the soil environment and the habitat provided for soil organisms. Treatment plots, 9.1 m × 82.3 m, each consist of six beds. A six-bed buffer area separates tillage treatments to enable system-specific equipment operations without disturbing neighboring plots.

The cover crop mixture of Juan triticale (*Triticosecale* Wittm.), Merced rye (*Secale cereale* L.) and common vetch (*Vicia sativa* L.) is applied at 19 cm row spacing on the beds at a rate of 89.2 kg ha^{-1} (30% triticale, 30% rye and 40% vetch by weight) in late October in the STCC and NTCC plots. After 2012, the basic cover crop mixture was changed to include a greater diversity of species, including pea (*Pisum sativum* L.), faba bean (*Vicia faba* L.), radish (*Raphanus sativus* L.), and phacelia (*Phacelia tanacetifolia* Benth.) (Mitchell et al., 2015). The cover crop plots, which are planted in advance of winter rains, were irrigated once with 10 cm of water in 1999 but not in subsequent years. Plots have been planted with either sorghum or garbanzo beans as summer or winter economic crops, respectively. Therefore, the field experiment is comprised of 32 plots (2 crops × 4 management systems × 4 replicates).

2.1.2. Field sampling for nematode assemblage assessment

Soil samples were collected in the 32 plots of the field experiment on April 27 and Oct 20, 2015. Each sample was a composite of 20 sub-samples collected randomLy with a 2.5 cm diameter auger at 0–20 cm depth and then mixed. The samples for nematode assemblage assessment were stored at 4 °C until analysis.

2.1.3. Soil fertility

Soils were sampled for total C and N in December 20, 2014 after harvest of the economic crop. From each plot, six to eight 7.6-cm-diameter cores at 0–15 cm depth were composited, sieved through a 2-mm screen, pulverized to pass through a 60-mesh screen, and dried to constant weight according to protocols of the University of California, Davis Analytical Laboratory (http://anlab.ucdavis.edu/sampling/soil-sampling-and-preparation). Total soil carbon was measured using a combustion C analyzer (CE Elantech, Inc., Lakewood, NJ). Bulk density was measured by the compliant cavity method (USDA NRCS, 2004). Total soil C levels were adjusted for bulk density and are reported as mg cm^{-3} dry soil (Mitchell et al., 2017).

2.2. Ecosystem function experiments

2.2.1. Soil column microcosms

Soil-column microcosms were constructed from 30-cm lengths of 4-cm i.d. polyvinyl chloride pipe with a drain hole in the basal cap. A stainless steel mesh with 0.24 mm apertures was placed in the bottom of each column to prevent soil loss. A 50 g layer of sand was placed above the mesh to facilitate drainage. Alfalfa leaf and stem tissue was dried and ground to a coarse powder. Alfalfa powder (5 g) was gently incorporated into 250 g of soil from each of the field site tillage and crop cover plots and the soil placed in the columns. The soil columns were tapped gently to settle the soil into a compact continuum without air voids. Soil columns were arranged vertically in a rack.

2.2.2. Experimental design

Three treatments were applied to soils from each of the four field management systems. In one treatment (F), soil and its biological component were maintained in field condition; in the second treatment (DF), the soil was defaunated by heating the columns in an oven at 80 °C for 2 h and then replacing weight loss with an equivalent weight of distilled deionized water; and in the third treatment (DFF) columns were defaunated and received commercial fertilizer solution during the subsequent 30-day incubation period. Prior to the start of the daily leaching regimen, each column received 100 mL of distilled deionized water and was allowed to drain so that the soils were all at field capacity. During the incubation period, each soil column was leached daily by adding 50 mL distilled deionized water (treatments F and DF) or 50 mL of dilute commercial fertilizer solution (DFF) to the top of the column. The leachate was collected in beakers placed under the holes in the base of the columns. In essence, the difference between treatment F and DF measured the ecosystem service of nutrient mineralization provided by the soil biota and the difference between DFF and DF measured the difference between the ecosystem service and the effect that could be achieved with mineral fertilizer. The concentration of fertilizer solution used to leach the DFF columns was calculated as the commercially recommended application divided by the anticipated number of days of the experiment. The fertilizer applied to the DFF columns was a dilute Miracle Gro solution (1 g/L, consisting of 3.5% NH_4NO_3, 20.3% urea-N, 8% P_2O_5, 16% K_2O, 0.02% B, 0.07% Cu, 0.15% chelated Fe, 0.05% chelated Mn, 0.0005% Mb). The column experiment was established with the three laboratory treatments (F, DF and DFF) and the four field management systems (NTCC, STCC, NTNO and STNO) with four replications, a total of 48 columns. Each day, 1 mL of leachate from each column was transferred by pipette to a capped glass vial and frozen prior to further processing for N analysis. Each day, 20 mL of each leachate was used to irrigate radish bioassay plants. The experiment was repeated in order to fine-tune the procedures; the procedures and data reported are those from the second iteration.

2.2.3. Radish bioassay

Pots (6.5 cm × 6.5 cm × 8.5 cm) with 50 g sand were autoclaved and eight radish seeds were sown in each pot. After 5 days, the germinated seedlings were thinned to five per pot for bioassay of the nutrient status of the leachates. Plants were grown at room temperature (±24 °C) with 12 h light per day. After 30 days of irrigation with leachate from the soil columns, plant height, root length, fresh and dry weight of radish seedlings were measured, mineral N concentrations from the frozen vials, and nematode assemblages in the soil columns were determined.

2.3. Biological and chemical analyses

2.3.1. Soil nematodes

Nematodes were extracted from the field samples and from the columns at the end of the 30-day incubation period using a combination of decanting and sieving and Baermann funnel methods (Barker, 1985). All nematodes in a sample were counted, and then the sample was centrifuged and nematodes identified in the small water drop remaining after removal of the supernatant (Ferris et al., 2004). At least 100 specimens per sample were identified to genus level, according to Bongers (1994), using an inverted compound microscope. Nematodes were assigned to a functional guild matrix of feeding habit (bacterivores, fungivores, herbivores, omnivores-predators) and colonizer-persister value of life-course strategies (1–5) (Ferris et al., 2001; http://plpnemweb.ucdavis.edu/nemaplex). We included the abundance of microbivores (bacterivores plus fungivores) as an indicator of the total activity in decomposition channels. Nematode abundance was expressed per 100 g dry soil.

2.3.2. Nitrogen

Frozen leachate samples were thawed. The NO_3^--N and NH_4^+-N were extracted using the methods of Doane and Horwath (2003). The concentrations of soil NO_3^--N and NH_4^+-N in the leachates were determined using a microplate reader. Standard curves were developed using known concentrations of NH_4Cl and KNO_3. The concentrations of NH_4^+-N and NO_3^--N in the samples was determined by comparison with the known standards. The total amount of mineral N (NH_4^+-N and NO_3^--N) was expressed as μg N mL^{-1} leachate.

2.4. Statistical analyses

Data from field samples were averaged over the two sampling times (April and October 2015) to capture the aggregate effects on the nematode assemblages of the 16 years under each management system. Nematode metabolic footprints, or nematode ecological indices, and the assignment of nematode functional guilds and colonizer-persister (cp) values were all calculated by the Ninja application (Sieriebriennikov et al., 2014) and as provided in http://plpnemweb.ucdavis.edu/nemaplex. Species diversity within functional guilds and diversity-weighted metabolic footprints were calculated per Ferris and Tuomisto (2015). Where necessary and appropriate, nematode data were ln (x+1) transformed to achieve normality prior to statistical analysis. The software package SPSS 16.0 for Windows (SPSS Inc., Chicago, IL, USA) was used for data analysis. Effects of crop, tillage and cover crops on nematode assemblages in the field experiment, and those of tillage, cover crop and fauna in the soil column experiment were analyzed by the general linear model (GLM) procedure and by the statistical procedures provided in Microsoft Excel spreadsheets.

3. Results

3.1. Field experiments

3.1.1. Effects of management systems on soil carbon levels

In data from the field plots described by Mitchell et al. (2017), the effect of management systems (tillage level and presence of absence of cover crops) on soil organic matter and soil organic carbon was significant ($P < 0.05$). The level of total soil carbon in NTCC plots was greater than those in STCC and STNO ($P < 0.05$; Fig. 1).

3.1.2. Nematode assemblage composition

In total, 31 genera nematode were identified across both sampling periods in the management system field plots, 18 in the first sampling and 28 in the second (Table 1; Supplementary Table S1 and S2). Among these genera, the predominant trophic group in numerical abundance was the bacterivores (50.2%), followed by fungivores (21.9%), herbivores (18.3%) and all predators (9.6%). The bacterivore, *Acrobeloides*, was the dominant genus in all management systems.

The numerical and biomass abundance of the total soil nematode assemblages in field plots were significantly greater where cover crops were included in the management systems ($P < 0.01$; Fig. 2). The inclusion of cover crops enhanced the diversity of

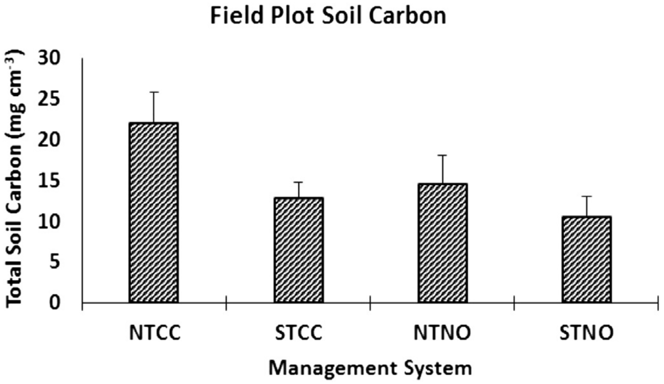

Fig. 1. Soil organic carbon (mg cm^{-3}) in field plots after 16 years under the following management systems: NTCC = no tillage and cover crop; STCC = standard tillage and cover crop; NTNO = no tillage and no cover crop; STNO = standard tillage and no cover crop (data from Mitchell et al., 2017).

Table 1
Nematode genera encountered in soil samples from the management system plots of the field experiment.

Bacterivores	Fungivores	All predators[a]	Herbivores
Acrobeles	*Aphelenchoides*	*Aporcelaimellus*	*Aglenchus*
Acrobeloides	*Aphelenchus*	*Discolaimium*	*Longidorus*
Chiloplacus	*Ditylenchus*	*Eudorylaimus*	*Merlinius*
Chronogaster	*Filenchus*	*Epidorylaimus*	*Pratylenchus*
Heterocephalobus	*Tylencholaimus*	*Mesodorylaimus*	*Psilenchus*
Mesorhabditis		*Microdorylaimus*	*Trichodorus*
Metateratocephalus		*Thonus*	*Tylenchus*
Monhystera		*Tripyla*	
Panagrolaimus			
Rhabditis			
Wilsonema			

[a] Nematode genera considered to be either specialist predators of nematodes or generalist predators (omnivores) of prey organisms that include nematodes.

bacterivore and herbivore nematodes, and enhanced the biomass and metabolic footprints of bacterivores and fungivores ($P < 0.05$; Table 2). The abundance of nematodes in all functional guilds, the diversity of bacterivores, and the metabolic footprints of bacterivores and fungivores were significantly higher in NTCC and STCC than in STNO and NTNO in plots with a garbanzo bean winter economic crop ($P < 0.05$) (Table 2). There were significant relationships between the diversity-weighted metabolic footprints of bacterivore and microbivore nematodes and soil carbon ($P < 0.01$, Fig. 3).

3.1.3. The relationship between nematode assemblages and total soil carbon

Soil carbon is considered the main source of energy for soil microorganisms while soil organic matter is a heterogeneous, dynamic substance that varies in particle size, C content, decomposition status, decomposition rate, and turnover time (Karlen et al., 2001; Franzluebbers, 2010). We expected that the abundance of soil microorganisms, and therefore resources for microbivore nematodes, will be related to levels of soil carbon. Therefore, we examined the relationships between the Enrichment Index and soil carbon and between the diversity-weighted abundance of microbivore nematodes and soil carbon. Since we were interested in examining the effects of carbon on nematode diversity and abundance, irrespective of the management system that resulted in the carbon, and since the data for carbon and the data for nematodes were from the same plots but not the same samples or sampling times, we ranked the pairs of observations on the basis of levels of carbon (mg cm^{-3}) irrespective of management system. Then, to reduce variability and reveal underlying trends, we produced pseudoreplicates based on three-lag moving averages of the ranked dataset (Fig. 3).

The Enrichment Index, based on proportions of nematodes in different functional guilds (Ferris et al., 2001) is an indicator of the nature of the relationship between opportunistic bacterivore and fungivore nematodes and available resources. The Enrichment Index across the field plots was strongly correlated with levels of total soil carbon generated by the field management strategies, reflecting the response of opportunistic bacterivore and fungivore nematode to organic enrichment ($r = 0.57$, $P < 0.01$; Fig. 3A). However, since it is based on proportional abundances, the Enrichment Index is not a useful indicator of the magnitude of the activities of a functional guild of nematodes. Metabolic footprints are measures of carbon usage in biomass accumulation and respiration of functional guilds of nematodes and as such are considered measures of the magnitudes of ecosystem services provided (Ferris, 2010). Ferris and Tuomisto (2015) suggested that the species diversity of nematodes in functional guilds is also a contributor to the magnitude of the ecosystem service. They proposed the use of metabolic footprints weighted by the species diversity of the contributing nematodes. The diversity-weighted metabolic footprint of enrichment opportunistic nematodes was correlated with the level of total soil carbon ($r = 0.50$, $P < 0.01$; Fig. 3B).

3.1.4. Predator-prey relationships in the field plot nematode assemblages

Bacterivores and fungivores were more abundant in the cover-cropped systems (Fig. 2) and the abundance of all predator nematodes (specialists and generalists) was significantly related to the abundance of those functional guilds ($r = 0.55$, $P < 0.01$; $r = 0.46$, $P < 0.01$). When one outlier data point was removed from the dataset and the 3-lag moving average pseudoreplication technique was used, a positive relationship emerged between predator biomass and the diversity-weighted footprint of all potential nematode prey taxa ($r = 0.70$, $P < 0.01$; Fig. 4). A large majority of the predator taxa was actually generalist predators or omnivores. Consequently, their abundance was probably also responding to non-nematode prey organisms.

3.2. Soil column experiments

3.2.1. Nematode assemblage composition

At the end of the soil column incubation period, the defaunated treatments (DF and DFF) remained essentially nematode-free and there were no significant field management system effects on total nematode abundance in the columns. The nematode assemblage in the faunated columns (F) consisted of 15 genera at the end of the incubation period (Table S3). Their proportional abundances were 52% bacterivores, 30% fungivores, 21% generalist and specialist predators and 35% herbivores. The enrichment-opportunist bacterivore, *Mesorhabditis* was the dominant genus in all faunated management system columns.

3.2.2. Plant growth in response to leachates from soil columns

Fresh and dry weights of radish plants used to bioassay the effects of leachates from the soil columns were significantly affected by the management systems applied to the soils over the 16-year period of the field experiment. They were greater in plants irrigated with leachates from faunated than defaunated column treatments. Plant height and root length in the columns with nematodes were greater than those in defaunated treatments ($P < 0.05$) and ranked as F > DF > DFF (Fig. 5). The mineral fertilizer content of the DFF treatments was apparently excessive and had an adverse or even toxic effect on the plant growth.

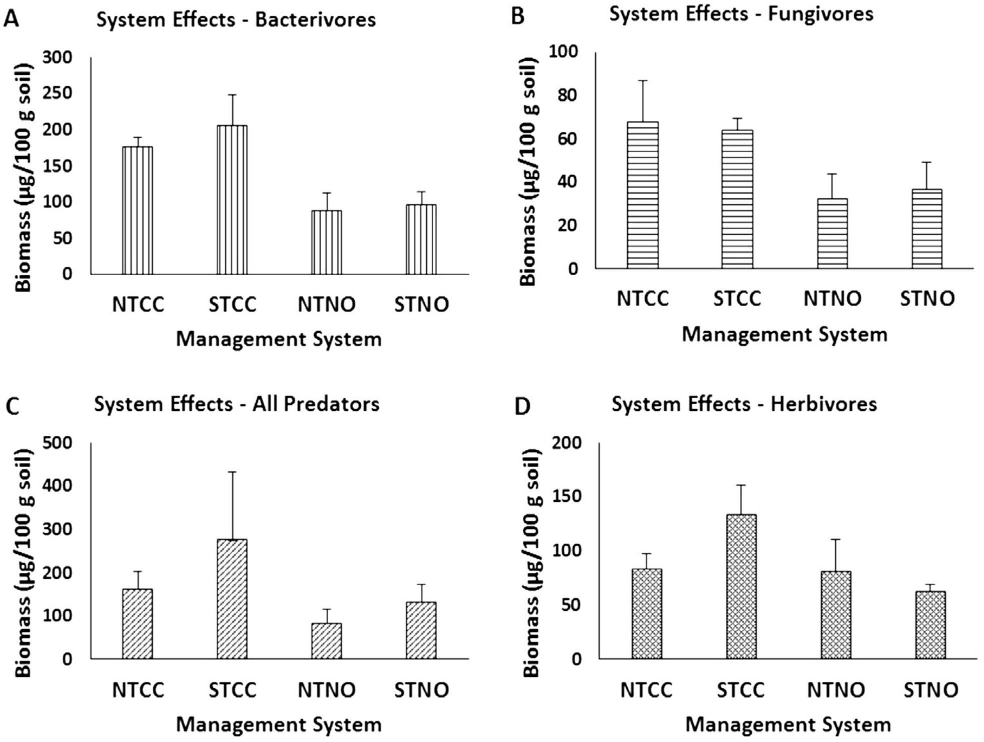

Fig. 2. Biomass of functional guilds of the soil nematode assemblage after 16 years under the following management systems: NTCC = no tillage and cover crop; STCC = standard tillage and cover crop; NTNO = no tillage and no cover crop; STNO = standard tillage and no cover crop. A. Bacterivores, B. Fungivores, C. All predators (omnivores and specialist predators), D. Herbivores.

Table 2
Numerical abundance (individuals 100 g^{-1} dry soil), species diversity (effective number of species) and metabolic footprints (µg C 100 g^{-1} dry soil) of soil nematode functional guilds in soils after 16 years of different management systems.

	Garbanzo Economic Crop				Sorghum Economic Crop			
	NTCC	STCC	NTNO	STNO	NTCC	STCC	NTNO	STNO[a]
Numerical Abundance								
Bacterivores	766.	624	326	186	641	619	539	446
Fungivores	378	397	86.1	78.0	315	323	297	258
All Predators	16	247	42.6	46.5	97.0	116	87.0	71.4
Herbivores	306	294	100	89.3	286	205	127	252
Species Diversity								
Bacterivores	3.84	3.64	2.4	3.08	3.07	4.78	3.22	3.21
Fungivores	1.76	1.68	1.64	1.69	1.74	1.71	1.73	1.75
All Predators	2.06	2.57	2.54	2.04	2.58	3.07	2.13	2.65
Herbivores	1.67	2.06	1.32	1.69	2.35	1.87	1.15	1.54
Metabolic Footprints								
Bacterivores	86.6	72.7	29.2	19.2	79.7	74.1	54.8	55.2
Fungivores	31.5	34.2	7.9	6.92	26.6	27.4	25.0	22.3
All Predators	75.0	47.7	15.3	10.8	37.3	37.8	42.2	27.3
Herbivores	52.5	34.8	20.6	15.0	40.7	30.3	23.3	45.5

[a] NTCC = no tillage and cover crop, STCC = standard tillage and cover crop, NTNO = no tillage and no cover crop, STNO = standard tillage and no cover crop.

3.2.3. *Nitrogen concentrations in leachates*

Cumulative mineral N was determined from the aggregated samples of 1 mL of leachate from each column collected each day. The concentrations of total mineral nitrogen in leachates of the faunated soil from no tillage and cover crop field management system were significantly greater than those from either tillage system without cover crops ($P < 0.05$; data not shown). In the F treatment, where the field nematode assemblage was not altered by defaunation, and the nitrogen in the leachate not altered by fertilizer application, we tested the relationship between cumulative mineral N and the species-diversity-weighted metabolic footprint of microbivore nematodes. Cumulative N provided a measure of the degree of mineralization of the alfalfa feedstock in the columns during the 30-day incubation period. The cumulative N concentration was correlated with the diversity-weighted metabolic footprint of the microbivore guild in the field plot soil placed in each column at initiation of the incubation period ($r = 0.73$; $P < 0.01$; Fig. 6), and with the diversity-weighted footprint of bacterivores ($r = 0.69$; $P < 0.01$).

4. Discussion

4.1. *The effect of agricultural practices on soil nematode assemblages*

Total nematode abundances, both numbers of individuals and biomass, in four functional guilds were greater in field plots with 16 years of history of cover crops grown in the intervals between economic crops. That is, in plots where the supply of resources to the soil food web through rhizodeposition and crop residues was continuous rather than punctuated. The increased availability of carbon in those plots provided resources for soil biota (Zhang et al., 2012; Shaw et al., 2016) and that was reflected in the magnitude and diversity of functional guilds of the nematode assemblages.

The abundance of nematode predators was linked to that of the prey nematodes (bacterivores, fungivores and herbivores). From the viewpoint of bottom-up forces, high abundance of prey favored the maintenance of higher abundance of predatory nematodes (Sánchez-Moreno et al., 2011). Predatory nematodes may regulate the prey nematodes by feeding, thus channeling resources from lower to higher trophic groups (Wardle and Yeates, 1993; Neher, 2001; Ferris et al., 2012b).

Bacterivores and fungivores, near the entry level of decomposition channels, are more direct beneficiaries of resource subsidy

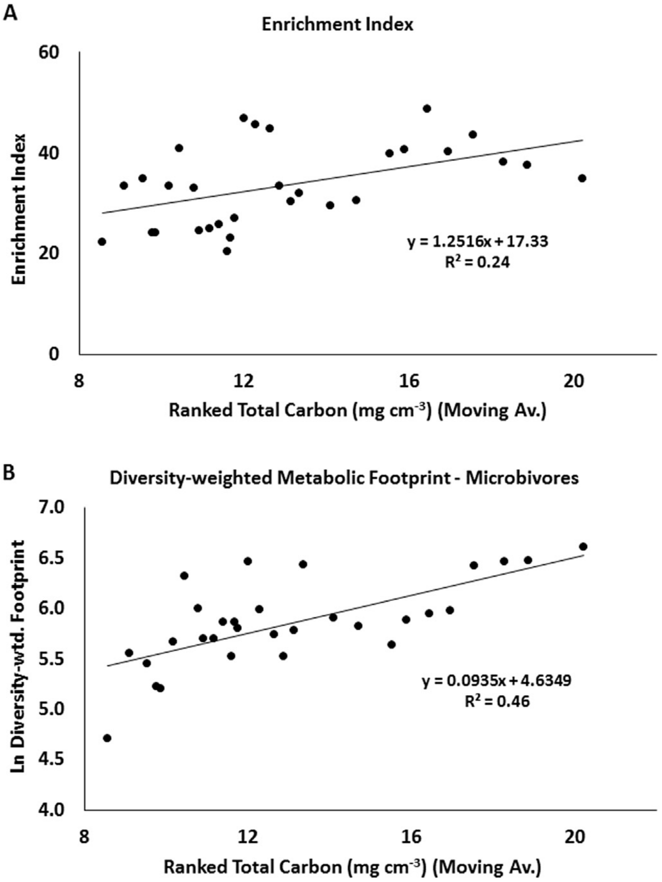

Fig. 3. The relationship between indicators and metrics of soil fertility provided by nematode faunal analysis and total soil carbon resulting from the four management systems. A. The Enrichment Index indicates that opportunistic bacterivore and fungivore nematode species respond to organic enrichment; B. The metabolic footprint weighted by species diversity of the enrichment opportunist nematodes in relation to soil organic carbon. Data points are averages of pseudoreplicates based on 3-lag moving averages of soil carbon levels averaged across 0–15 cm and 15–30 cm depths.

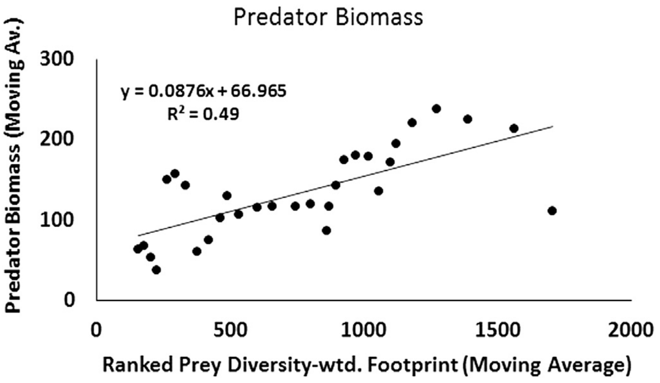

Fig. 4. The relationship between the biomass of generalist and specialist predator nematodes and the diversity-weighted abundance of the nematode component of their prey (bacterivores, fungivores and herbivores). Samples are ranked on the basis of nematode abundance and the regression is calculated between 3-lag moving averages of data on the ordinate and abscissa axes.

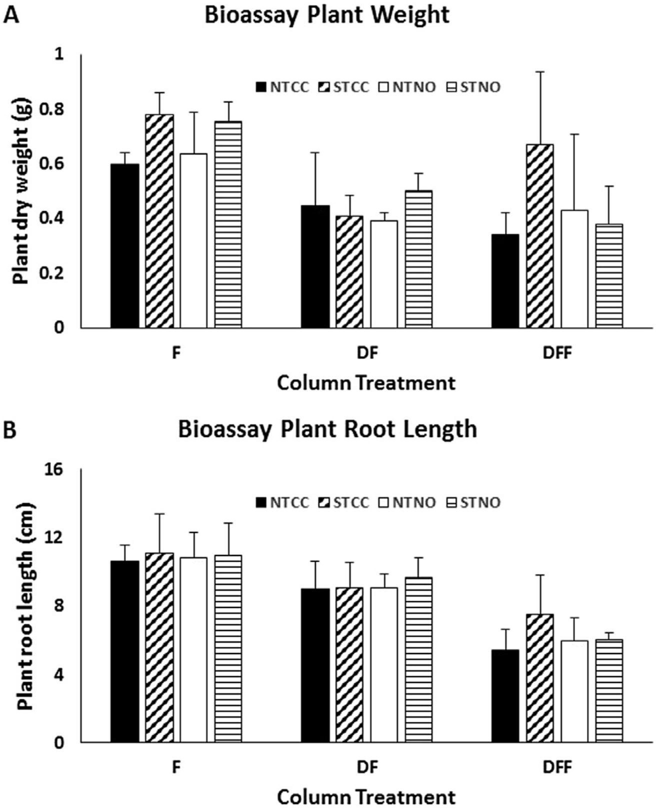

Fig. 5. Bioassay plant response to daily irrigation for 30 days with leachate from columns of soil from field plots subjected to 16 years of different management systems (NTCC = no tillage and cover crop; STCC = standard tillage and cover crop; NTNO = no tillage and no cover crop; STNO = standard tillage and no cover crop). A. Mean dry weights of total 5 seedlings and B. Mean root lengths per radish plant. In F columns the soil fauna remained intact, in DF columns the soil was defaunated, and in DFF columns the soil was defaunated and received a daily application of mineral fertilizer solution.

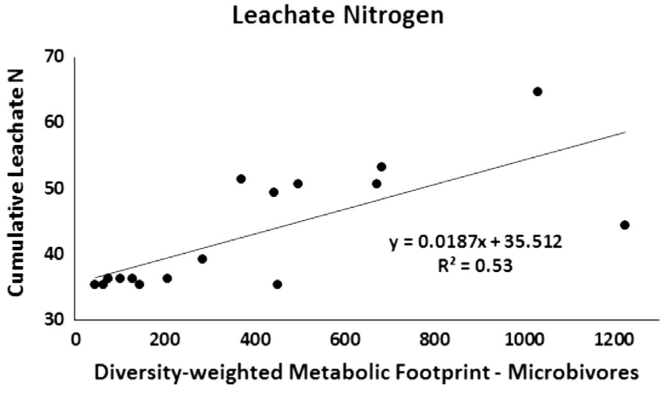

Fig. 6. The relationship between cumulative mineral N (μg mL^{-1}) in leachate from soil columns and the diversity-weighted metabolic footprint of microbivore nematodes at initiation of the incubation period. The mineral N was collected from the leachate daily across 30 days of incubation.

than other functional guilds. Greater biomass and metabolic footprints of nematode guilds under cover-cropped systems was also related to mechanical incorporation of organic resources in the upper layers of the soil profile, as occurred with standard tillage operations. While mechanical incorporation of organic material through tillage may benefit opportunistic taxa, it is generally considered detrimental to large-bodied predator species that are more sensitive to environmental disturbance (Ferris et al., 2001; Zhang et al., 2012). However, in this study, omnivore and predator nematodes in the field plots were not negatively affected by the disturbance effects of soil tillage. It is conceivable that disturbance-tolerant taxa of omnivores and predators have been selected during the 16 years of exposure to farming systems with standard tillage.

Cover crop quantity and resource quality are important determinants of the magnitude of soil food web services (DuPont et al., 2009; Andrés et al., 2016). The incorporation of plant residues or other organic matter increases the supply of resources to the soil food web and fuels the ecosystem (Li et al., 2009). In the current study, continuous input of resources into the soil food web in management systems that include cover crops had greater impact on soil nematode communities than the incorporation effects associated with tillage. Zhang et al. (2012) also found that the responsiveness of opportunistic soil nematode species offset any negative effects of tillage disturbance.

4.2. *Metabolic footprints and energy flows through nematode assemblages*

Nematode metabolic footprints convey more information on allocation of C into growth and egg production, and to its utilization in metabolic activity, than numerical abundance or even biomass of individuals. They are useful indicators of the magnitude of soil food web functions (Ferris, 2010; Bhusal et al., 2015). In the present study, metabolic footprints of microbivore nematodes were greater in cover-cropped than non-cover-cropped systems, which reflected the greater amount of C flowing through food web decomposition channels in those systems.

Additional resources (such as cover crops and organic amendments) directly affect organisms at the entry level of the food web, and bacterivore and fungivore nematodes are always the first responders to resource enrichment (Ferris et al., 2012c; Ferris and Tuomisto, 2015; Steel and Ferris, 2016). Thus, besides considering their ecosystem services in nutrient mineralization, it is instructive to consider the role of these nematodes in the indirect service of providing prey to predator nematodes and thus enhancing the regulation of opportunistic target species, including parasites of plants. Ferris et al. (2012b) termed bacterivore and fungivore nematodes the “amplifiable prey” because their abundance can be enhanced by managed resource inputs. They theorized that such increase would provide resources for predator nematode which would also increase and enhance the regulation of “target prey” plant-parasitic species. The regulation of target prey was not determined in this study but there was a clear relationship between predator biomass and the biomass of amplifiable prey.

Weighting of the metabolic footprints by species diversity was proposed by Ferris and Tuomisto (2015) in recognition that the magnitudes of ecosystem services of functional guilds of nematodes and other organisms must be related to, besides their total metabolic, the differences in size, behavior and activity that is provided by greater species diversity. The diversity-weighted footprints of bacterivores and other microbivores were significantly positively related to soil organic carbon. Similarly, a high species diversity of prey nematodes would provide a range of resources with different size and activity to sustain higher-order predator guilds (Ferris and Tuomisto, 2015).

4.3. *The effect of mineralization roles of nematodes on plant growth*

Fungal and bacterial-feeding nematodes, and other organisms that graze on primary decomposers, accelerate the decomposition of soil organic matter and enhance the mineralization rates of nutrients available for plant growth. The grazing is considered to stimulate microbial growth turnover while movement of the microbivores transports microbes to new resources. Differences in C:N ratio between nematodes and their prey, and the excess organic N associated with C used in respiration, result in the excretion of excess N in mineral form by the nematodes (Ingham et al., 1985; Ferris et al., 1997, 2012c, 1998; Chen and Ferris, 1999; García-Palacios et al., 2013; Gebremikael et al., 2015). In the current experiments, all component parts of plants irrigated with leachate from columns with the soil fauna intact were greater than those irrigated with leachate from defaunated columns. Also, the cumulative concentrations of mineral N were greater in leachates from the faunated than from the defaunated columns.

Our idea with the defaunated columns receiving dilute fertilizer solution each day was to allow us to determine whether the N mineralized by the soil fauna would allow reduction in fertilizer application rates in field sites with high metabolic footprints of appropriate functional guilds of nematodes. In fact, the mineral fertilizer leachate, although applied at rates calculated from label recommendations, was apparently too concentrated and had a phytotoxic effect on the bioassay plants. However, plants irrigated with leachate from the faunated soil columns supported robust growth of radish plants.

5. Conclusions

Through the current research and other cited studies, the abundance and species diversity of microbivore nematodes, and organisms of which they are bioindicators, are strongly linked to levels of total soil carbon. Those levels can be enhanced by long-term and consistent application of farming systems that maximize uninterrupted C-fixation or C-amendment and which reduce soil disturbance. The physiological, behavioral and activity differences among diverse microbivore species facilitates successional changes in food web structure without compromising ecosystem services. In such functional succession, the players change but the game continues. The evolution of our understanding of the functional attributes of the soil food web continues to underscore the importance of soil carbon in soil quality and soil health.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.soilbio.2017.03.017.

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