

Conservation agriculture affects arthropod community composition in a rainfed maize–wheat system in central Mexico



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ABSTRACT

As a system of practices involving crop rotations, reduced soil disturbance, and the retention of organic matter at the soil surface, conservation agriculture (CA) increases soil quality, reduces erosion, and provides a favorable habitat for beneficial soil-dwelling organisms which may provide improved pest control. To determine the effect of CA on generalist arthropod predators and pests, we assessed the ground-dwelling arthropod assemblage prior to crop planting and shortly after crop emergence in a long-term CA trial at the International Maize and Wheat Improvement Center (CIMMYT) in central Mexico. We used pitfall traps and in-field sentinel insect assay arenas to evaluate arthropod activity-density and predation, respectively, in a maize–wheat rotation, planted under CA (zero tillage, retention of residues) and conventional agriculture (tillage and no surface residue). In maize, activity-density of generalist predators (excluding ants) was higher in conventional agriculture treatments than in CA treatments prior to crop planting ($P = 0.03$), but no significant differences were apparent in arthropod activity-densities at the treatment level at any other time. In multivariate analyses, the arthropod community was affected by tillage in maize at both sampling dates ($P \leq 0.05$), and by residue after crop emergence in wheat ($P = 0.03$). Spiders trended toward a greater association with no-till treatments in maize and treatments with residue retained in wheat. In wheat, predation (biological control potential) was significantly lower in conventional compared with CA treatments ($P \leq 0.05$). According to multiple linear regressions, higher levels of soil cover significantly explained predation before and after planting in maize, and before planting in wheat ($P \leq 0.05$). Our results indicate that the type and amount of residue that remains at the soil surface may influence arthropod community dynamics. This first report of the effects of CA on arthropods in this long-term trial indicates that CA in central Mexico may contribute to conservation of certain arthropod predators and biological control of insect pests.

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1. Introduction

Globally, soil degradation is one of the many constraints contributing to low yields in subsistence agriculture, and thus a significant contributor to food insecurity (Greenland and Nabhan, 2001; Lal, 2009). Conventional agricultural practices involving frequent and intensive tillage and crop residue removal have been associated with degradation of soil resources by causing erosion and compaction, reducing nutrient and water holding capacities, and reducing habitat for beneficial soil organisms (Henneron et al., 2015; Nyamangara et al., 2014; Thierfelder and Wall, 2010). As an alternative to conventional agricultural production, conservation agriculture (CA) includes the retention of crop residues on the soil

surface, an increase in crop diversity through rotations, and minimizing tillage used for various cultural practices, such as weed management (Erenstein et al., 2012; Hobbs et al., 2008; Knowler and Bradshaw, 2007; Palm et al., 2014; Verhulst et al., 2010). These practices together augment soil quality and reduce erosion, increase and stabilize yields, and provide a more complex and favorable habitat for soil-dwelling organisms (Govaerts et al., 2005; Henneron et al., 2015; Nyamangara et al., 2014; Pineda et al., 2012; Rendon et al., 2015), but many challenges within regional contexts still need to be addressed in CA systems.

Decreasing the frequency and intensity of tillage and retaining crop residues on the soil surface can contribute to an increase in herbivorous insects, some of which may be crop pests of economic importance (Brévault et al., 2007; Hammond, 1991; Henneron et al., 2015; Kladienko, 2001). An increase in the prevalence of insect pests may be a risk factor associated with CA, but arthropod natural enemies, e.g., generalist predators, may help to suppress these

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insect pests (Henneron et al., 2015; Schmidt and Rypstra, 2010; Wyckhuys and O'Neil, 2007). Generalist predators, such as spiders (Araneae) and carabid beetles (Coleoptera: Carabidae) non-selectively feed on other arthropods, and have been cited as contributing to lower plant damage and a reduced number of herbivores in vegetable systems, for example (Riechert and Bishop, 1990). In CA, the practices that may contribute to increased numbers of insect pests, namely residue retention and reduced tillage, may also contribute to the conservation of generalist predators (Rendon et al., 2015; Schmidt and Rypstra, 2010). Any potential increases in pest numbers because of these practices may then be mitigated by an increase in the abundance of generalist predators, but the total effect of CA on the interactions between herbivorous and predatory arthropods is an area that warrants further study. An understanding of the arthropod community at the soil surface is also important in informing interactions beyond plant–herbivore–predator, as some non-predatory and non-herbivorous arthropods present in the system may serve as supplemental prey to retain generalist predators in the field prior to pest outbreaks (Mémott et al., 2007).

Since 1991, the International Maize and Wheat Improvement Center (CIMMYT) has maintained a long-term trial in El Batán, Mexico, to evaluate and refine CA-based practices. As compared to practices considered conventional for the area (the removal of crop residues from the field and the use of inversion tillage for soil preparation and weed control), a maize–wheat rotation and retention of crop residues in combination with no-till management have contributed to stabilizing yields (Govaerts et al., 2006; Verhulst et al., 2011). Additionally, CA practices, in particular no-till and crop residue retention in combination, resulted in higher numbers of bacteria and fungi indicative of soil health, low to moderate prevalence of root rot and plant-parasitic nematodes, and maintenance of a high level of soil microbial biomass as compared to the treatments classified as conventional (Govaerts et al., 2008, 2007, 2006).

The risk of increased insect pests with CA, coupled with the high use of pesticides in Mexico and the significant damage caused annually by the fall armyworm in maize, *Spodoptera frugiperda* (J.E. Smith) (Blanco et al., 2014; Bolaños-Espinoza et al., 2001; Wyckhuys et al., 2013), are reason to study the effects of CA on the arthropod community in the long-term trial located at CIMMYT, where such research has not previously been a focus. By determining how CA and conventional tillage and residue management practices affect the beneficial arthropod community and predation rates in this agroecosystem, we can gain a better understanding of how these practices could contribute to in-field biodiversity and biological control potential (Wyckhuys et al., 2013). Specifically, we hypothesized that in a no-till system where the previous year's crop residue had been retained in the field (full CA), we would observe the following as compared to a tilled system with the residue removed (full conventional agriculture): (1) higher activity-densities and a greater diversity of generalist arthropod predators at the soil surface; (2) fewer herbivores at the soil surface; (3) higher in-field predation (and thus, biological control potential); and (4) lower crop damage caused by chewing insects early in the cropping season.

2. Materials and methods

2.1. Site description

We conducted our research during the May–November, 2013 growing season at CIMMYT's experimental station in El Batán, Mexico (19°31'55"N, 98°50'51"W). El Batán is located in the central Mexican highlands at an elevation of 2250 masl, with a mean annual precipitation of 625 mm between 1991 and 2013, and a

mean of 542 mm of precipitation during the growing season of May through October. Rainfall during the growing season in 2013 was above average, at 645 mm. Mean monthly minimum and maximum temperatures were 6.3 and 24.4 °C, respectively, in the years 1991 through 2013 (data recorded from CIMMYT's on-site weather station). According to the Food and Agriculture Organization of the United Nations (FAO) soil classification system, the soil is a Haplic Phaeozem, described as a moderately well drained, light clay (FAO et al., 2012).

2.2. Experimental design and field operations

In the long-term, rain-fed trial, conservation and conventional agricultural practices have been implemented at various levels at the same site since 1991 (Govaerts et al., 2005). The trial consists of a randomized complete block design, with two repetitions, and each plot measuring 7.5 m by 22 m. Of the 32 total treatments in the long-term trial, 8 were selected for the research reported here: a full entry, maize (*Zea mays* L.) and wheat (*Triticum aestivum* L.) rotation, with either no-till or tilled plots, and retention or removal of the previous years' crop residues. Tillage consisted of a single pass with a chisel plow after harvest in the previous year, to a depth of 30 cm, followed by a disk harrow at a depth of 20 cm. Residue was incorporated into the soil when retained in tilled plots, and left on the soil surface in no-till plots. For the purposes of this research, we consider the residue retained, no-till treatments as full CA treatments, and tilled plots with residue removed as full conventional agriculture treatments.

Both crops were planted in the first week of June; maize at a rate of 25 kg seed ha⁻¹ in 75 cm rows, and wheat at a rate of 110 kg seed ha⁻¹, in 20 cm rows, both with recommended crop cultivars commonly used in the area. All treatments received the same rate of fertilizer (150 kg N ha⁻¹ as urea), which was disked into the soil at the time of planting in maize. In wheat, urea was disked into the soil prior to planting in zero tillage, and incorporated through tillage in conventional tillage. Maize seed was treated with an insecticide with an active ingredient of clothianidin prior to planting, at a rate of 0.3 mg/kernel of active ingredient. Both crops received 20 mm of sprinkler irrigation after planting to ensure uniform germination, and both crops emerged during the second week of June. Weeds were controlled with applications of a post-emergence herbicide as appropriate. On July 3 and again on July 24, maize received an insecticide treatment with an active ingredient of chlorpyrifos (240 g of active ingredient per hectare) in response to high numbers of *Spodoptera frugiperda* J. E. Smith (Lepidoptera: Noctuidae) and a weevil pest complex (*Nicentrites testaceipes* Champion and *Geraeus senilis* Gyllenhal, Coleoptera: Curculionidae) (Blanco et al., 2014; Bolaños-Espinoza et al., 2001). Historically, the experiment has received similar treatments of insecticides in response to pest incidence as needed, typically once or twice per growing season.

The two center maize rows of each plot were hand-harvested on November 26, and the 8 center rows (1.6 m width) were harvested in wheat on October 8 with a combine. Grain was dried and shelled, and yield is reported as dry weight of grain in kg ha⁻¹.

2.3. Characterization of ground-dwelling arthropods

2.3.1. Pitfall traps

To characterize the local assemblage of ground-dwelling arthropods, we employed pitfall traps (at a depth of 129 mm, and with a 114 mm diameter), using ethylene glycol as a killing agent. Traps remained open in the field for 72 h (Bestelmeyer et al., 2000). Arthropods were preserved in 70% ethanol, counted and identified to at least order, with some groups identified to family, and species in the case of ants, according to established keys. We

assigned arthropod groups to a specific trophic group (predator, herbivore, decomposer, or omnivore) based on review of the literature. Trapping occurred twice during the growing season, in the week prior to planting and approximately two weeks after crop emergence, to isolate treatment effects on recruiting and retaining generalist predators in the field during crop establishment (Landis et al., 2000; Wyckhuys and O'Neil, 2006).

We placed two pitfall traps in a transect in each treatment plot during each sampling date, outside of the center yield rows and approximately 1.5 m from the plot edge. We determined total activity-densities per plot by averaging the numbers of arthropods captured in each trap. After crop emergence, we pitfall sampled at maize growth stage V3, and wheat at Feeke's stage 3 (Nafziger, 2009a,b). We obtained measurements of environmental variables within each plot at the time of pitfall sampling, including soil temperature at 5 cm below the soil surface, soil moisture at a depth 0 to 20 cm, depth of crop residues, proportion of soil covered by crop residues, and crop height (when present) in order to classify the local microenvironment which could affect arthropod mobility and presence (Andersen, 2000).

2.3.2. Visual assessments

To further characterize the arthropod community and potentially identify arthropods not captured by pitfall traps, we conducted timed visual assessments to identify live invertebrates at the soil surface (Bestelmeyer et al., 2000). We conducted the assessments at a time corresponding with the risk of a mid-season pest outbreak of the true armyworm, *Pseudaletia unipuncta* Haworth in mid-August (Lepidoptera: Noctuidae) (Bolaños-Espinoza et al., 2001). In maize, we searched two areas of 2.25 m² per treatment plot for live individuals. In wheat, we searched two areas of 0.1 m² in the same manner, with different sizes of areas in the maize and wheat due to the higher density of plants in a smaller area in wheat (Bestelmeyer et al., 2000). We searched the two areas in each crop for five minutes, and we identified arthropods in the field to various taxonomic levels: to order (Araneae and Chilopoda), family (most groups), genus (*Diabrotica* sp.), and species (*P. unipuncta*). For each crop, we averaged the arthropod abundances to obtain a mean value per treatment plot prior to analysis.

2.4. Biological control potential

To determine the biological control potential of generalist arthropod predators on populations of early-season pests, we deployed assay arenas baited with live, last-instar larvae of the greater waxworm (*Galleria mellonella* Fabricius) as sentinels (Grieshop et al., 2012). Each assay arena consisted of a round cage made of 19-gauge plastic hardware cloth placed at the soil surface, which excluded larger, vertebrate predators while permitting access by arthropods. In each assay arena we placed a card affixed with five waxworm larvae, with four assay arenas in each treatment plot. Assays occurred in the 24 h prior and the 24 h after pitfall trapping (Grieshop et al., 2012). The before- and after-pitfall sentinel assays were combined as one predation "sampling event", with the number of damaged waxworms reported as a proportion of total waxworms deployed in each plot (20 waxworms before and after pitfalling) for a total of 40 waxworms deployed per plot per predation sampling event.

2.5. Crop damage and yield

In both maize and wheat treatment plots, we assessed plant damage approximately two weeks after crop emergence, and dry grain yield in kg ha⁻¹ at time of harvest. The total number of plants, and the number of plants with chewing damage (e.g., by

caterpillars) were counted in two areas of 2.25 m² and 0.1 m² in maize and wheat, respectively, with different areas assessed due to the difference in plant density for each crop. Damage by the fall armyworm, *Spodoptera frugiperda* (J.E. Smith), was also assessed in maize in mid-August. We assessed every maize plant within the plot for damage, considering the plot as a representative of the entire maize population in the area, and plants were counted as either damaged, with feeding damage in the maize whorl, or with no damage (Wyckhuys and O'Neil, 2006).

2.6. Data analysis

All analyses were conducted using R (R Core Team, 2013), with specific packages used as described. Crops and sampling dates were kept separate for all analyses because of the differences anticipated in pitfall captures due to seasonality in insect phenology, and due to the strong crop effect. In the case of analysis of variance (ANOVA) and mixed model analyses, we used Tukey's honest significant difference test to conduct post hoc tests of means (Gotelli and Ellison, 2004; R Core Team, 2013).

Differences in means of activity-density of arthropod functional groups, predator group richness (the number of groups present in a plot), predator evenness (the relative contribution of each predator to total predator activity-density) (Smith and Wilson, 1996), proportion of crop damage by chewing insects and crop height (measured at the time of pitfall sampling after crop emergence) and yield were subjected to ANOVA, with treatments as the explanatory variables. Data were log or square root transformed to ensure assumptions of normality and equality of variances were met (Ives, 2015). In the case of the timed observations completed in mid-August, we used the non-parametric, one-way Kruskal–Wallis rank sum test to identify the differences between the pairs of treatment levels (Kutner et al., 2005). Due to a difference in the foraging behavior of ants, and their potential antagonistic relationships with other predatory taxa, we analyzed the total activity-density of the predatory ants separately from other predators (Benckiser, 2010; Mestre et al., 2012; Philpott and Armbrecht, 2006).

We conducted tests of nonmetric multidimensional scaling (NMDS) to summarize relationships within the arthropod community as a whole using the vegan package in R (Borcard et al., 2011; Oksanen et al., 2015; R Core Team, 2013). Vegan is a statistical package designed for use in community ecology, with multiple functions developed for the purposes of identifying patterns and relationships between different taxa within local assemblages of organisms (Oksanen et al., 2015). We used post hoc tests of environmental regression fits (envfit function) to interpret the ordination axes using treatment and environmental variables measured at the time of pitfall trapping (Borcard et al., 2011; Oksanen et al., 2015). Envfit identifies correlations between the variables and ordination scores through bootstrapping (we used 10,000 permutations), but cannot test for interactions, so the residue by tillage treatment was included as a treatment variable (Borcard et al., 2011). We show only significant vectors and treatment variables in the results ($P \leq 0.05$) to eliminate noise in biplots.

To determine the effect of treatment on predation (biological control potential), we employed mixed models using the lme4 package in R, with treatments as fixed effects, and time (the two sampling dates of pre-planting and post-emergence) as a random effect (Bates et al., 2015; Gotelli and Ellison, 2004; R Core Team, 2013). To further understand the effects driving sentinel predation, we used multiple linear regression with sentinel predation as the response variable and various environmental and arthropod values as explanatory variables, and used backward stepwise selection using the stepAIC function in the MASS package

in R to reduce the complexity of the full models (Schmidt and Rypstra, 2010; Venables and Ripley, 2002). Models were compared by Akaike's Information Criteria (AIC), with the final model having the lowest AIC (Kutner et al., 2005; Murtaugh, 2009).

3. Results

3.1. Characterization of ground-dwelling arthropods

3.1.1. Pitfall traps

In both maize and wheat, predatory ants, *Pheidole pilifera* Roger, *Pheidole hirtula* Forel, and *Dorymyrmex insanus* Buckley (Hymenoptera: Formicidae), dominated the ground-dwelling predator community (Tables 1 and 2), with ants representing 75.7% of the predator activity-density in maize and 76.3% in wheat, for both dates and all treatments combined. Activity-densities of predatory ants were consistently high in the full conventional treatments (tilled, residue removed) in maize at both sampling dates, and more variable among treatments in wheat, but in neither crop were any main effects significant.

Melyridae (Coleoptera) and Araneae comprised the bulk of the remaining predator activity-densities in both crops, representing 12.1 and 7.7% of the predator activity-density in maize pitfall traps, and 10.9 and 8.9% in wheat pitfall traps, respectively. Carabidae (Coleoptera), Staphylinidae (Coleoptera), Anthocoridae (Hemiptera), Cantharidae (Coleoptera), Chilopoda, Reduviidae (Hemiptera), and Solifugae, represented less than 2% each of total pitfall captures in both crops. In maize, post hoc test of means indicated a significant difference between the two no-till treatments prior to crop planting, with a higher activity-density of predators (excluding ants) in the residue removed treatment than in the residue retained treatment ($P=0.03$, Table 1). In wheat, after crop emergence, only the main effect of residue was significant for the total predator activity-density ($P=0.04$, Table 2).

Predator richness, the number of taxonomic predatory groups with ants counted at the family level, was relatively constant (Tables 1 and 2), and only the main effect of tillage was significant in maize prior to crop planting ($P=0.03$), with more species captured in the tilled treatments. Predator evenness was

consistently high in the full CA treatments in maize (Table 1), but no differences between treatments were significant.

Herbivore numbers were very low in pitfall traps in both crops, both prior to crop planting and after crop emergence (Tables 1 and 2). With both dates combined and in all treatments, unidentified hemipteran nymphs comprised the bulk of the herbivores (42% in maize, 36% in wheat), followed by Thysanoptera (19% in maize, and 22% in wheat). In both crops, herbivore activity-density was lower in treatments with residue retained compared to treatments with residue removed (not significant, Tables 1 and 2).

Nonmetric multidimensional scaling (NMDS) indicated that arthropod groups captured in the pitfall traps varied in the growing season (Figs. 1 and 2). In maize, of the predatory arthropods, spiders (Araneae) and Melyridae (soft-winged flower beetles) trended toward greater activity-densities in no-till treatments at both sampling dates, while Carabidae and Staphylinidae trended toward tilled treatments (Fig. 1). Some herbivorous groups were associated with specific treatments, e.g., Acrididae (Orthoptera) trended toward an association with tilled treatments on both dates, and lepidopteran larva trended toward an association with no-till treatments after crop emergence. According to the tests of environmental fit, the main effect of tillage was significantly correlated with the arthropod community in maize on both dates ($P \leq 0.05$, Fig. 1), and treatment (the variable tested for the residue by tillage interaction) was significant prior to maize planting ($P=0.01$). The amount of soil cover at the soil surface correlated with the community at both sampling dates ($P \leq 0.05$), indicating a strong correlation between quantity of soil cover and the full CA (residue retained, no-till) treatments, due to the direction of the vector (Fig. 1).

In wheat, the main effect of residue is significantly related to the arthropod community after crop emergence ($P=0.03$, Fig. 2), with some predators (Araneae) associated with residue retention, and others associated with residue removal (Carabidae). Herbivore activity-density was low in wheat after crop emergence, but Aphididae and Cicadellidae were associated with treatments where residue had been removed (Fig. 2b). Crop height after wheat emergence was significantly related to activity-density of the community ($P=0.04$), with the vector from the centroid of the

Table 1
Mean activity-density, richness, and evenness (\pm SEM) for arthropod trophic groups in maize prior to planting and after crop emergence, and mid-season visual assessments of arthropods on the soil surface.

	Residue retained		Residue removed	
	Tilled ($n=2$)	No-till ($n=2$)	Tilled ($n=2$)	No-till ($n=2$)
<i>Pre-planting pitfall traps</i>				
Predatory ant activity-density	19.00 (3.50)	26.00 (0.00)	55.25 (35.25)	17.50 (3.00)
Non-ant predator activity-density	13.25 (2.25)	6.75 (0.25)	10.25 (1.75)	16.25 (0.75)*
Total predator activity-density	32.25 (5.75)	32.75 (0.25)	65.50 (33.50)	33.75 (3.75)
Predator group richness**	6.00 (2.00)	2.50 (0.50)	5.00 (0.00)	3.00 (0.00)
Predator evenness	0.55 (0.13)	0.70 (0.04)	0.40 (0.16)	0.34 (0.05)
Herbivore activity-density	2.00 (0.50)	2.75 (2.25)	5.00 (2.50)	2.25 (0.25)
<i>Post-emergence pitfall traps</i>				
Predatory ant activity-density	17.00 (5.00)	17.25 (6.75)	49.00 (20.50)	9.50 (2.00)
Non-ant predator activity-density	5.00 (2.00)	7.50 (0.50)	3.25 (0.75)	5.00 (1.00)
Total predator activity-density	22.00 (3.00)	24.75 (6.25)	52.25 (19.75)	14.50 (3.00)
Predator group richness	5.50 (0.50)	4.00 (1.00)	4.00 (0.00)	5.00 (0.00)
Predator evenness	0.39 (0.12)	0.47 (0.01)	0.20 (0.05)	0.41 (0.03)
Herbivore activity-density	1.50 (1.00)	2.50 (2.00)	2.25 (0.25)	2.00 (1.50)
<i>Mid-season visual assessment</i>				
Predator abundance**	0.50 (0.50)	3.25 (0.75)	0.50 (0.50)	3.75 (2.75)
Herbivore abundance	0.50 (0.50)	0.75 (0.75)	0.50 (0.50)	1.50 (0.50)

n indicates the number of repetitions of each treatment.

* Post hoc tests of means by Tukey's honestly significant difference test indicated significantly different values at $P \leq 0.05$.

** Means significantly different for the main effect of tillage ($P \leq 0.05$).

Table 2

Mean activity-density, richness, and evenness (\pm SEM) for arthropod trophic groups in wheat prior to planting and after crop emergence, and mid-season visual assessments of arthropods on the soil surface.

	Residue Retained		Residue Removed	
	Tilled ($n=2$)	No-till ($n=2$)	Tilled ($n=2$)	No-till ($n=2$)
<i>Pre-planting pitfall traps</i>				
Predatory ant activity-density	30.50 (22.00)	25.00 (7.00)	24.00 (3.00)	35.00 (11.00)
Non-ant predator activity-density	10.50 (0.50)	8.25 (3.25)	10.50 (0.50)	6.50 (2.00)
Total predator activity-density	41.00 (22.50)	33.25 (3.75)	34.50 (2.50)	41.50 (9.00)
Predator group richness	3.50 (0.50)	3.50 (0.50)	4.50 (0.50)	4.00 (1.00)
Predator evenness	0.69 (0.18)	0.79 (0.11)	0.53 (0.02)	0.70 (0.11)
Herbivore activity-density	2.25 (0.25)	1.50 (0.00)	1.25 (0.75)	2.75 (2.75)
<i>Post-emergence pitfall traps</i>				
Predatory ant activity-density	30.25 (10.25)	18.25 (4.75)	7.75 (2.75)	13.00 (5.00)
Non-ant predator activity-density	5.25 (2.25)	8.25 (1.75)	5.00 (1.00)	2.75 (0.75)
Total predator activity-density [#]	35.50 (8.00)	26.50 (6.50)	12.80 (1.75)	15.80 (5.75)
Predator group richness	5.00 (1.00)	5.00 (1.00)	5.50 (0.50)	3.20 (0.50)
Predator evenness	0.25 (0.06)	0.37 (0.03)	0.54 (0.12)	0.37 (0.10)
Herbivore activity-density	1.25 (0.25)	1.25 (0.25)	1.00 (0.00)	3.50 (2.00)
<i>Mid-season visual assessments</i>				
Predator abundance	1.50 (0.50)	2.00 (0.00)	1.00 (0.00)	1.50 (0.50)
Herbivore abundance	1.00 (0.00)	11.25 (2.75)	0.75 (0.75)	0.00 (0.00)

n indicates the number of repetitions of each treatment.

[#] Means significantly different for the main effect of residue ($P \leq 0.05$).

biplot directed toward the residue retained treatments, indicating a strong correlation between a taller wheat crop where residue had been retained (Fig. 2b).

3.1.2. Visual assessments

In the visual assessments, the bulk of the predators observed in maize were Coccinellidae (Coleoptera, 30%), followed by Araneae (27.5%), Staphylinidae (Coleoptera, 17.5%), Cantharidae larva (Coleoptera, 7.5%), Chilopoda (7.5%), Melyridae (Coleoptera, 5%), Anthocoridae (Hemiptera, 2.5%) and ants (Hymenoptera: Formicidae, 2.5%). *Diabrotica* sp. (Coleoptera: Chrysomelidae) was the primary herbivore observed in maize (46.9%), followed by Curculionidae (Coleoptera, 18.8%), true armyworm, *Pseudaletia unipuncta* Haworth (Lepidoptera: Noctuidae, 15.6%), Acrididae (Orthoptera, 12.5%), and Scarabaeidae (Coleoptera, 6.3%). The main effect of tillage was significant in maize ($P=0.04$, Table 1), with more predators observed in no-till compared with tilled treatments.

In wheat, we observed fewer groups of predators, the bulk of which were Coccinellidae (50%), followed by Araneae (33.3%), Melyridae (8.3%) and Anthocoridae (8.3%). We also observed fewer groups of herbivores than in maize, the majority of the observations were of true armyworm (96.2%), and we observed a single *Diabrotica* sp. individual (3.8% of total observations) in a tilled plot with residue. While more true armyworms were observed in the residue retained, no-till treatments (full CA treatment), differences were not significant between treatments (Table 2).

3.2. Biological control potential

The biological control potential, as measured by sentinel predation assays, was relatively high in the long-term trial, with all treatments exhibiting some level of predation of live *G. mellonella* larvae. In maize, as measured by proportion of damaged waxworms, the full CA treatments (no-till, residue retained) had the highest proportion of mean predation (Fig. 3a). No main effects were significant in maize. In wheat, treatment had a greater effect on sentinel predation (Fig. 3b), with predation suppressed in the full conventional treatments (tilled, residue removed), with a significant main effect of tillage ($P=0.02$). In wheat, predation is

significantly higher in each of the no-till treatments than in the full conventional treatment (tilled, residue removed, $P \leq 0.05$, Fig. 3b).

Backward selection in the multiple regression models revealed associations between predation of sentinel waxworms, and activity-densities of various arthropod groups and certain environmental variables. In maize, four variables were present in all three of the best fitting models: predator richness, predator evenness, soil cover, and soil temperature (Table 3). The amount of residue at the soil surface (soil cover) explains the greatest amount of variance in predation in all three models, with a higher mean proportion of predation where the amount of residue is higher at the soil surface. Predator richness and evenness are both represented in all three models, but it is only after crop emergence and prior to planting, respectively, that each explains a large portion of the variance in waxworm predation.

In wheat, three variables were present in all three of the best fitting regression models: predator evenness, soil cover, and soil temperature (Table 4). Prior to crop planting, soil cover explains the greatest amount of variance in sentinel predation in wheat, with tillage explaining the bulk of the remainder of the variance. After wheat emergence, soil temperature captures most of the variance in predation, with no treatment effects present in the model at that time. In the model incorporating both sampling dates (repeated measures, Table 4), variables that were not in the models for each individual date are present, i.e., crop height and herbivore activity-density. However, the treatment effects of tillage and residue explain the most variance in sentinel predation in the repeated measures model.

3.3. Crop damage and yield

In both maize and wheat, early in the season, the crop exhibited minor damage by chewing insects (data not shown). In maize, plant damage by chewing was moderately higher in treatments where the residue had been retained, and only the main effect of residue is significant ($P=0.05$). At the time of the mid-season assessment of damage by fall armyworm (*Spodoptera frugiperda* J.E. Smith) in maize, tilled treatments experienced higher damage (Fig. 4a), with a significant main effect of tillage ($P=0.01$) and a significant interaction between residue and tillage ($P=0.05$). Post hoc tests of means indicated a significant difference between the

two no-till treatments and the tilled, residue retained treatment, with both of the no-till treatments exhibiting significantly less damage than the tilled treatment ($P \leq 0.05$). Mean early-season crop height and dry weight (kg ha^{-1}) of grain in maize were highest in the residue retained, no-till treatments (full CA), although there were no significant effects for either (Fig. 4b).

No significant differences or trends were apparent in wheat for damage by insects or for crop height. Wheat grain yield was highest in the full CA treatments (Fig. 4c), although the benefit of no-till was negated if the residue is removed, as the no-till, residue removed treatments had the lowest mean grain yield. However, these differences were not significant.

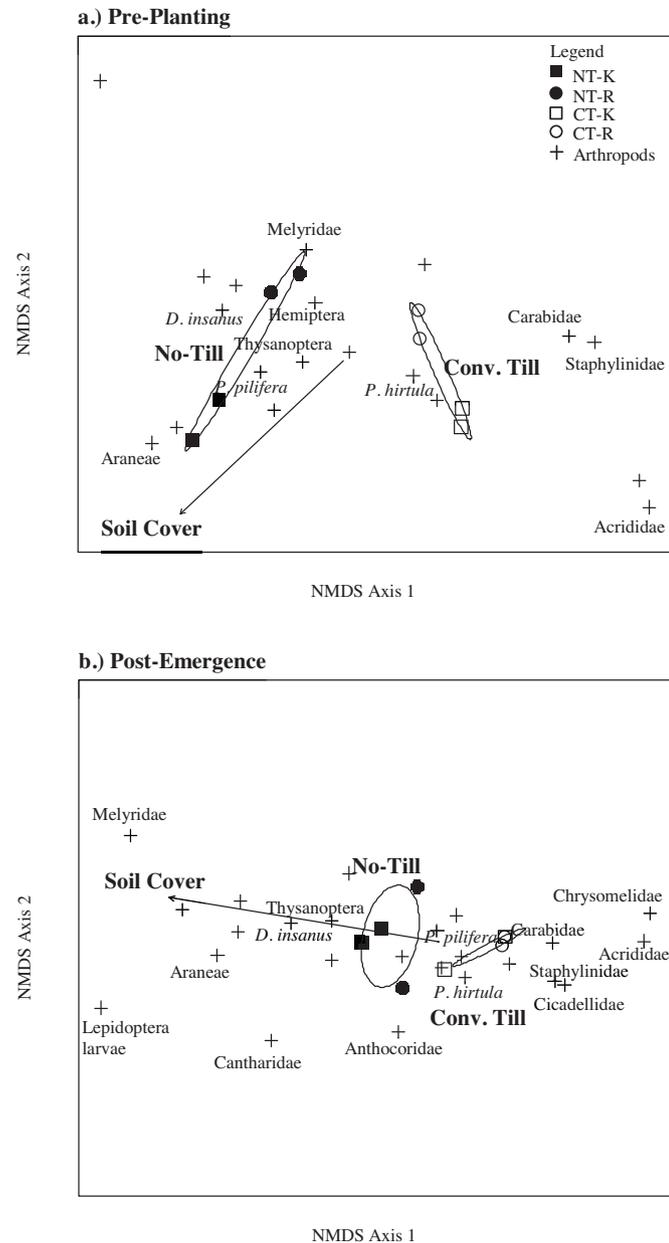


Fig. 1. Nonmetric multidimensional scaling (NMDS) ordination plots (3 dimensions with the first two axes shown, Bray–Curtis distance) for the maize arthropod community captured by pitfall trap prior to crop planting (a) and after crop emergence (b). Only predatory and herbivorous groups and significant environmental variables ($P \leq 0.05$) are shown. Ellipses represent 95% confidence intervals for significant main effects ($P \leq 0.05$). NT: no-till; CT: conventional tillage; K: residue retained in field; R: residue removed from field. Full CA treatments are NT-K; full conventional are CT-R.

4. Discussion

In accordance with our hypotheses, our results indicate that residue retention in tandem with no-tillage and crop rotations (CA) has potential for conserving certain ground-dwelling predators, e.g., Araneae (spiders). Brévault et al. (2007) identified a similar pattern to that suggested here; compared with conventional practices in that in a mulched, no-till cotton (*Gossypium hirsutum* L.) cropping system in Cameroon, Araneae were associated with CA and Carabidae with conventional tillage practices (Brévault et al., 2007). Soil cover, which is largely determined by the implementation of CA practices, contributed significantly to explaining variance in predation (biological control potential) at both sampling dates in maize and prior to crop planting in wheat, indicating that predation in this long-term trial may also benefit from CA practices. Our hypothesis regarding reduced plant damage was in part confirmed in maize, in that damage by fall armyworm (*Spodoptera frugiperda* J.E. Smith) was significantly lower in the full CA treatment compared to the full conventional treatment (tilled, no residue) in our mid-season assessments. In both crops, CA treatments provided a non-significant advantage to grain yield, although the effect was significant in previous years in the long-term trial (Govaerts et al., 2005; Verhulst et al., 2011).

Arthropods are highly mobile, and with a small plot size, there is the potential for movement between experimental treatments; however, because of the age of the long-term trial, there is a strong chance that arthropod populations associated with specific treatments have stabilized through time (Henneron et al., 2015; Prasifka et al., 2005; Sabais et al., 2011). Cantelo (1986) studied a large matrix of plot sizes, in the range of 4–4,000 m^2 , and suggests that minimum plot sizes of 100 m^2 and 30 m^2 are necessary to determine effects of insecticides on potato leafhopper (*Empoasca fabae* Harris) and corn earworm (*Helicoverpa zea* Boddie), respectively. Prasifka et al. (2005) suggests that the effect of plot size is taxon specific, based on the behavior of the organism (e.g., due to size and relative immobility, Collembola are less likely to move between plots). These researchers avoided the recommendation of a minimum plot size for studying nontarget effects of pest management (transgenic crops), but suggest that plots with a size of less than 81 m^2 may underestimate effects of pest management treatments (Prasifka et al., 2005). Perner (2003) also proposes that community parameters, e.g., evenness, are a sound estimate of population dynamics when an achievable level of precision in sample size may not be possible. Similarly, Wyckhuys and O'Neil (2006) were able to identify positive and significant effects of natural enemies, including spiders and ants, in suppressing *S. frugiperda* in smallholder maize in Honduras. The field sizes were larger than those used here (an approximate range of 0.24 to 1.17 ha in the two areas they studied), but their research suggests that even at a small scale, generalist predators are important for suppressing insect pests in subsistence and smallholder agriculture (Wyckhuys and O'Neil, 2006). Thus, in spite of the small plot size (165 m^2) and low number of replicates in the long-term trial, the trends isolated in this research may be indicative of trends we might observe at a field scale in CA in Mexico.

While we did not examine a year-to-year effect of a crop rotation in this system, we see strong differences between the arthropod communities in maize and wheat when grown in close proximity to, and in rotation with, each other. Predator activity-densities were comparable between the two crops; however, herbivore activity-density varied between maize and wheat. Avoiding crop pests is a strong impetus for rotations within a CA system, and rotations have long been established as a beneficial integrated pest management (IPM) tactic (Prasifka et al., 2006; Thierfelder and Wall, 2010). However, the benefit of the rotation in

CA may also be related to the type of crop residue that has been retained at the soil surface from the previous year, as this will in part dictate the habitat for beneficial insects at the time of planting of the following crop (Abro et al., 2011; Schmidt and Rypstra, 2010). This difference in the structure and composition of the maize and wheat residues at the soil surface may explain the difference between the arthropod communities in each crop prior to planting, i.e., the strong treatment effect observed where maize was to be planted (Fig. 1) with no effect in wheat.

The residue treatment in maize does not significantly affect the arthropod community according to our multivariate analyses at either sample time, but the amount of residue at the soil surface

(soil cover) does prior to crop planting, indicating that the residue treatment itself (retained or removed) is not particularly important so much as the type and amount of residue that remains at the soil surface. Retaining residue at the soil surface may be important in preserving an early-season predator assemblage that can protect the crop as it establishes and in early developmental stages (Wyckhuys and O'Neil, 2006). The additional complexity provided to generalist predators by residue at the soil surface – be it habitat, alternative prey items, or intraguild predators which may warrant avoidance of a habitat patch – is of particular importance in the early-season as these factors may affect establishment of predator populations for the duration of the growing season (Landis et al., 2000; Schmidt and Rypstra, 2010; Wyckhuys and O'Neil, 2006). Schmidt and Rypstra (2010) identified the importance of different mulches in retaining the wolf spider, *Pardosa milvina* (Araneae: Lycosidae), with the identity of the mulch driving the activity-densities of the spiders more so than the availability of prey items. Caballero-López et al. (2012) also identified a significant relationship between aphidophagous predators and the type of plant cover present in the field, with a significant and positive relationship between legumes and foliar predators.

The relationship between habitat and predator abundance is not constant through time, as we observed for predatory ants. While CA may provide a specific benefit to ground-nesting ants (Brévault et al., 2007), some ant species prefer warmer soils with less obstructions at the soil surface (Andersen, 2000; Grieshop et al., 2012; Thompson, 1990). This may be the case with the ants identified as predatory in this system, as the activity-densities of these three ant species combined were highest in the full conventional agriculture treatments (residue removed, tilled) in maize at both sampling dates. Little information exists on the feeding ecology of these individual ant species, but many *Pheidole* species are omnivorous and *Dorymyrmex* are generalist scavengers (Andersen, 2000; Fisher and Cover, 2007; Thompson, 1990), and all three were observed feeding on sentinel waxworms in the field (data not shown). These three ant species may thus be foraging for the food resources available in the conventional agriculture system, e.g., preying on herbivores or Collembola, both of which are present in the full conventional treatments in higher numbers in pitfall traps than in the CA treatments in maize (Carroll and Janzen, 1973; Perfecto, 1990). Ants have a relatively large foraging range and species within these two genera are known to be stress tolerant. As such, they may be able to withstand the disturbance (i.e., tillage) associated with the conventional treatments or foraging in areas where residue would not interfere with their foraging efficiency (Andersen, 2000; Benckiser, 2010; Carroll and Janzen, 1973; Evans et al., 2011). Pitfall traps also have the potential for underestimating ground-dwelling populations in high-residue environments, especially for arthropods with unique foraging habits like ants (Bestelmeyer et al., 2000; Lang, 2000; Melbourne, 1999).

While identifying the activity-densities of these predators through pitfall captures is essential in understanding probable habitat effects on specific groups, the functional role that predators and other organisms play in the environment is of equal or more importance as their presence at a specific location and time. In particular, we are interested in the biological control potential of these predators—their ability to not only reduce herbivore numbers, but also to reduce the potential for those herbivores to cause crop damage (Landis et al., 2000; Wyckhuys et al., 2013). Where sentinel predation (biological control potential) is suppressed in tilled maize with residue retained compared to the full CA treatments, activity-densities of predators in pitfalls are comparable between those two treatments. However, the evenness of the predator assemblage is always numerically highest in the full CA treatments in maize (although the trend is

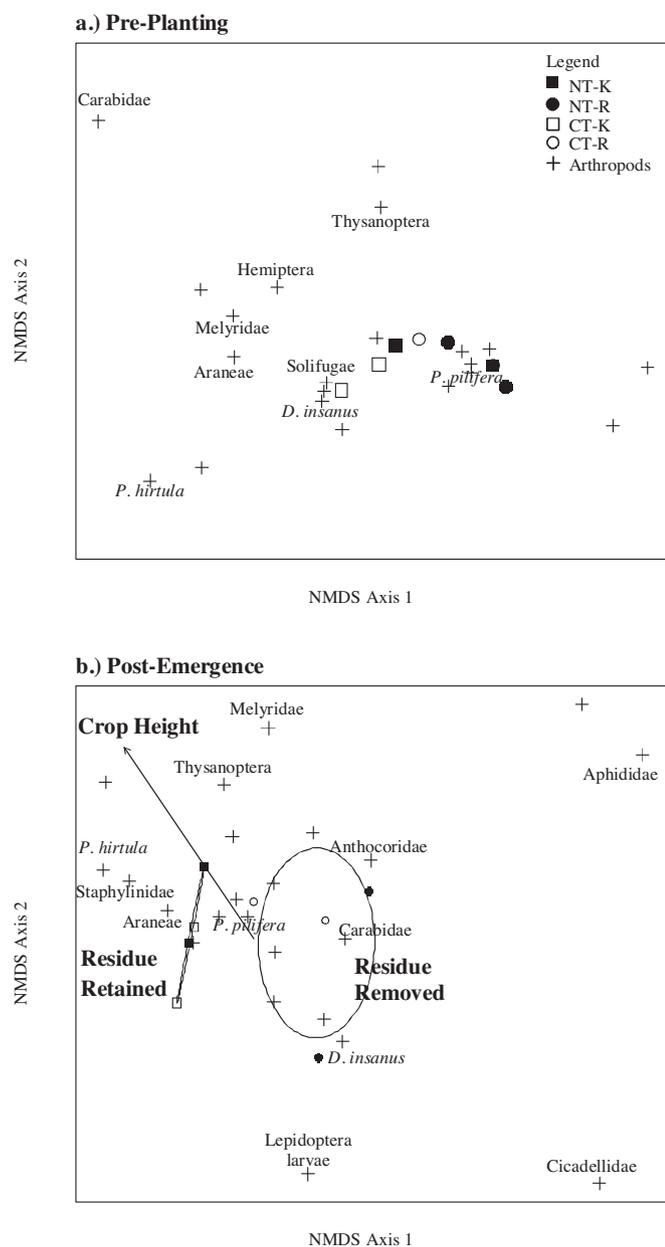


Fig. 2. Nonmetric multidimensional scaling (NMDS) ordination plots (3 dimensions with the first two axes shown, Bray–Curtis distance) for the maize arthropod community captured by pitfall trap prior to crop planting (a) and after crop emergence (b). Only predatory and herbivorous groups and significant environmental variables ($P \leq 0.05$) are shown. Ellipses represent 95% confidence intervals for significant main effects ($P \leq 0.05$). NT: no-till; CT: conventional tillage; K: residue retained in field; R: residue removed from field. Full CA treatments are NT-K; full conventional are CT-R.

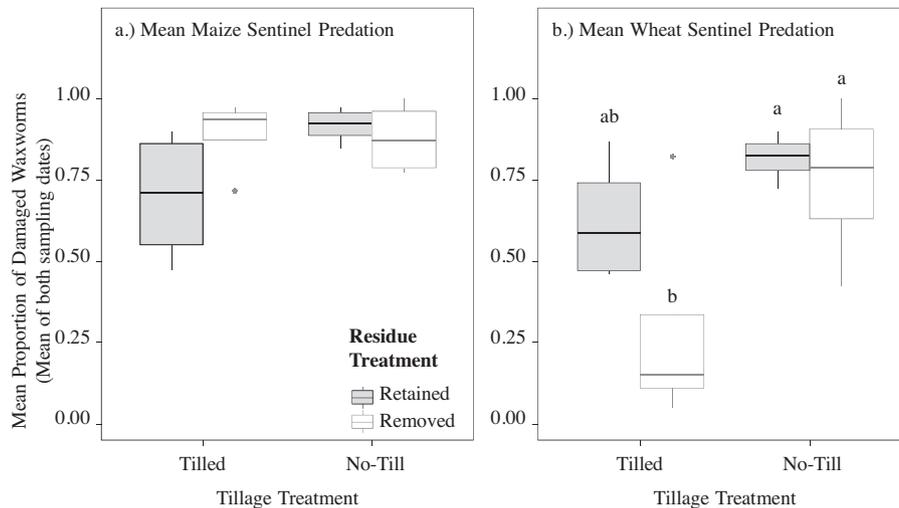


Fig. 3. Mean proportion of predator-damaged sentinel waxworms for both sampling dates ($n=80$) in maize (a) and wheat (b). Treatments with different letters are significantly different by post hoc comparisons of means with Tukey's honest significant difference test at $P \leq 0.05$. Single dots represent potential outliers. Full CA treatments are no-till with residue retained; full conventional are conventional tillage with residue removed.

nonsignificant at $0.05 < P < 0.10$), and evenness is reflected in the multiple logistic regressions as an important variable in predicting predation in both maize and wheat. A highly abundant, dominant and stress tolerant predator group, such as ants, may be important in influencing biological control potential in some cases (i.e., the conventional agriculture treatments in maize), but a more even community may be the most important predictor of biological control potential in the full CA treatments (Crowder et al., 2010). This relationship between CA, predator evenness, soil cover, and biological control potential warrants further study in large-scale CA experiments, as the nonsignificant trend may be indicative of results that could scale up to the commercial field level.

One of the concerns of a high residue environment at the soil surface is the potential for increased incidences of pests and a resulting effect on yield (Henneron et al., 2015; Mischler et al., 2010). We observed higher numbers of true armyworm (*Pseudaletia unipuncta* Haworth) in full CA treatments in wheat at the time of our mid-season assessments, but the increased presence of the pest in those treatments did not correspond with an effect on yield. Likewise, we observed a reduced number of fall armyworm (*S. frugiperda*) in CA treatments in maize. A number of factors may contribute to yield in between the time when we sampled the arthropod community and predation, e.g., rainfall was above

average in 2013 for the period 1991–2013, potentially resulting in a reduced benefit to yield of CA in 2013 as compared to conventional treatments and to the results in previous years of the long-term trial (Govaerts et al., 2005; Verhulst et al., 2011). However, the results presented here are a promising indication that CA treatments may provide enough of an agronomic benefit to the crop that it is able to withstand potential damage by insect pests if they are increased by any aspect of CA (Thierfelder and Wall, 2010), as they were in this year in wheat. The increased activity-density of the predators at times, and their provision of biological control services, may also be an additional ecosystem service of CA in protecting the crop at key times of pest infestations.

5. Conclusion and recommendations

While the experimental design in this system is not ideal for studying the arthropod community due to the low number of replications and small plot size, we were still able to isolate trends worth further exploration. In light of the high rate of pesticide use in Mexico, and the need for promoting integrated pest management within the country, the results have broad implications for both small- and large-scale producers of maize and wheat. The

Table 3

ANOVA table for the explanatory variables for the best fitting models in maize for predicting in-field sentinel predation as selected by Akaike's information criteria (AIC) prior to planting and after crop emergence, and with time included as a random variable.

Explanatory variable	Pre-planting			Post-emergence			Repeated measures		
	df	F	P	df	F	P	df	F	P
Collembola	1	676.13	0.02				1	17.80	0.01
Crop height				1	297.87	0.00	1	7.94	0.04
Herbivores							1	2.03	0.21
Predatory ants							1	5.30	0.07
Predator richness	1	121.46	0.06	1	2,965.13	0.00	1	2.24	0.19
Predator evenness	1	377.30	0.03	1	466.05	0.00	1	2.03	0.21
Soil cover	1	691.24	0.02	1	3,672.62	0.00	1	3.08	0.14
Soil moisture	1	82.95	0.07						
Soil temperature	1	337.52	0.03	1	95.79	0.01	1	0.80	0.41
Residue							1	4.00	0.10
Tillage							1	7.26	0.04

Table 4

ANOVA table for the explanatory variables for the best fitting models in wheat for predicting in-field sentinel predation as selected by Akaike's information criteria (AIC) prior to planting and after crop emergence, and with time included as a random variable.

Explanatory variable	Pre-planting			Post-emergence			Repeated measures		
	df	F	P	df	F	P	df	F	P
Collembola				1	648.58	0.02			
Crop height							1	2.06	0.20
Herbivores							1	2.85	0.14
All predators				1	171.39	0.05			
Predatory ants							1	1.22	0.31
Predator richness	1	138.74	0.05	1	233.75	0.04			
Predator evenness	1	295.70	0.04	1	89.36	0.07	1	2.20	0.19
Soil cover	1	1,147.54	0.02	1	67.57	0.08	1	1.68	0.24
Soil temperature	1	140.96	0.05	1	845.55	0.02	1	1.11	0.33
Residue	1	193.65	0.05				1	3.92	0.10
Tillage	1	685.22	0.02				1	10.24	0.02
Tillage × residue							1	1.49	0.27

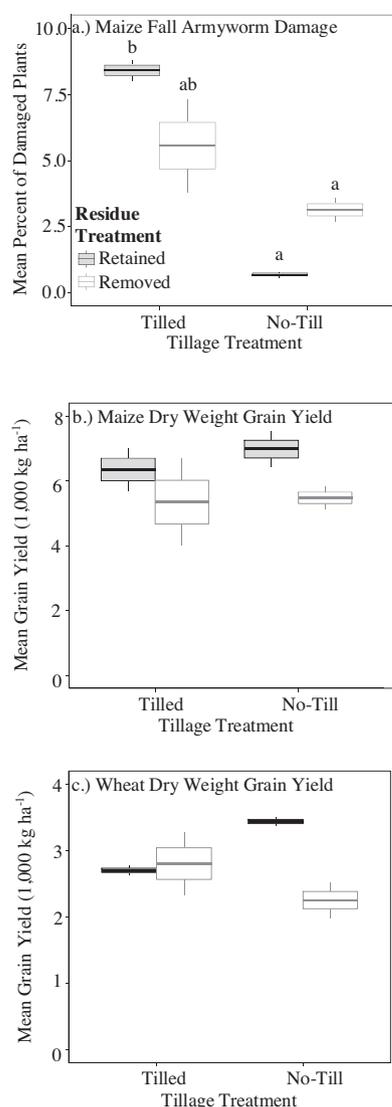


Fig. 4. Mean percent of damage by fall armyworm in maize (a), and mean dry weight grain yield (kg ha^{-1}) in maize (a) and wheat (b). Means with different letters are significantly different at the treatment level at $P \leq 0.05$ according to Tukey honestly significant different test of means. Full CA treatments are no-till with residue retained; full conventional are conventional tillage with residue removed.

established trends might be a result of the age of the trial (22 years), which ensures that the different systems have stabilized over time. In particular, the greater activity-densities of certain predators and predator evenness, a relationship between high soil cover and biological control potential, and no effect on yield of higher numbers of true armyworm in wheat, may all indicate the potential benefit of conservation agriculture to the ground-dwelling arthropod community and their beneficial activities.

The research initiated here could be expanded upon with on-farm assessments of the benefits of conservation agriculture to predator-prey interactions and mitigation of pest populations. Including manipulative experiments with known densities of predators and pests, as well as exploring the landscape level factors affecting arthropod populations could provide additional value in understanding the mechanisms affecting these populations. Additionally, expanding crop rotations to include other types of crops, e.g. legumes, whose residue may provide an additional subsidy to predators is of interest, especially in regards to how these residues may affect predators of the key lepidopteran pests in this system. In particular, identifying levels and types of residue

that may benefit multiple taxa of predators, e.g. both spiders and ants, may be a way to maximize predation efficiency and predator community evenness.

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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.2015.12.004>.

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