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## RESEARCH ARTICLE - ANTS

### Ant Community in Neotropical Agrosystems: A Four-Year Study in Conventional and No-Tillage Systems

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#### Abstract

Studies comparing agricultural practices that maintain a better quality and a healthy soil fauna consider the no-tillage farming as the most effective practice when compared to other planting techniques. In order to evaluate the influence of the no-tillage and conventional tillage methods (with and without manipulation of the soil before planting, respectively) on ant communities, we monitored two areas with these two types of practice (conventional and no-tillage) over the period of four years. We collected ants once per month along 10 transects randomly distributed using three pitfall traps in each area. In addition, we collected the dead plant biomass present at each point sampled as a parameter for measuring the environmental complexity of the areas. In total, we captured 27,480 individuals belonging to 26 species in the no-tillage area and 24,570 individuals belonging to 24 species in the conventional tillage area. The generalised linear model analysis showed that the no-tillage system had the highest abundance of individuals, as well as richness and diversity of species, during most of the study period, as compared to conventional tillage areas. We also found a significant positive correlation between species richness and dead plant biomass. Thus, it is possible to infer that the no-tillage area is a more complex environment with a greater diversity of ants and, therefore, a more sustainable agrosystem as compared to conventional tillage areas.

#### Introduction

Agriculture is a major cause of biodiversity loss around the world (McLaughlin & Mineau, 1995; Urrutia-Escobar & Armbrrecht, 2013). While agricultural expansion has increased the productivity of farmland, the number of animal and plant species has decreased (Isselstein et al., 1991). This scenario does not occur only in countries with extensive cultivated areas, but also in small countries with limited farmland (McLaughlin & Mineau, 1995; Büchs, 2003).

The global scenario of transformation of the natural environment in agrosystems has stimulated researchers to look for alternatives, to minimize the negative effects of agriculture on biodiversity. In this sense, agricultural practices that have a reduced impact on nature, but are economically competitive,

has been the goal of sustainable agriculture in recent decades (Tilman et al., 2002; Sani, 2011).

In searching for sustainable agricultural practices that are able to maintain the soil quality and the diversity of fauna, the no-tillage farming practice is currently been considered as the most efficient method (Sani, 2011). Unlike conventional tillage, in which the dry plant biomass of the previous crop is incorporated to the soil by a mechanization process, the no-tillage is a reduced form of soil preparation, where essentially no manipulation of the soil is performed before planting (Stinner & House, 1990). In no-tillage systems, the dry plant biomass on soil after harvest provides protection against soil degradation and improves microclimate conditions for survival of the edaphic fauna (Doran et al., 1994; Hawksworth, 1991).



Several studies have evaluated the effect of tillage on soil fauna, using groups of arthropods, such as beetles (Stinner & House, 1990; Bohac, 1999; Ashford et al., 2013), collembolans and spiders (Stinner & House, 1990; Ashford et al., 2013) and ants (Stinner & House, 1990; Booij & Noorlander, 1992; Harvey & Eubank, 2004; Sharley et al., 2008; Ashford et al., 2013). Other studies have also investigated communities of bacteria, fungi, mites and nematodes (Behan-Pelletier, 1999; Treonis et al., 2010). All of these organisms are affected by the type of soil management and have been used as bioindicators (Ashford et al., 2013).

In addition to their role as bioindicators of environmental change, most ant species are efficient predators of other insects, contributing to pest control in crops (Perfecto, 1991; Agarwal et al., 2007; Rico-Gray & Oliveira, 2007; Lange et al., 2008). According to Booij and Noorlander (1992), ants, beetles and spiders are the most effective generalist predators in agrosystems. Several authors have shown that more complex environments exhibit higher predation rates by ants (Harvey & Eubanks, 2004; Philpott & Armbrrecht, 2006; Lange et al., 2008). According to Mansfield et al. (2003), effective biological pest control is one of the goals of sustainable agriculture.

During the present study, ants were monitored over the period of four years in two areas treated with different soil management practices, one with mechanized conventional tillage and the other with no-tillage. We hypothesised that the area with no-tillage will have a more diverse ant community when compared to the area with conventional tillage. This result should be associated with the greater structural complexity of the no-tillage system, because of greater availability of food, less competition between ant species, and therefore greater diversity. Studies on ants communities in agrosystems such as this is aimed to understand the effect of tillage systems on edaphic fauna and inform stakeholders that aim to implement agricultural practices that have a lower impact on biodiversity.

## Material and Methods

### *Study Area*

We carried out the study in Dourados, Mato Grosso do Sul, Brazil (22°13'16" S and 54°48'20" W) from November 1999 to December 2003. The region is part of the Cerrado biome, with an altitude ranging between 449 and 477 m. The climate, according to Köppen, is Cwa (humid mesothermal), which is characterized by the presence of two seasons: rainy (September-February) and dry (March-August), average annual rainfall of 1,500 mm and the annual average temperature of 22°C ( $\pm$  2.3). The soil of the region is the type dystroferric hapludox clayey with flat topography (EMBRAPA, 1999).

We monitored the ant communities of two contiguous areas of 1.5 ha each, both belonging to the Universidade Federal da Grande Dourados. The two areas were kept

under conventional tillage from 1997 until the beginning of this study (1999), when a no-tillage method was established in one of the areas. During the study, three annual harvests were carried out in both areas. The summer crops, sown in October or November and harvested in February or March, were soybeans and corn; in the winter, corn and wheat sown after the harvest of summer and harvested in June or July. The sorghum, oats, sunflower and turnip crops were sown in August or September to preserve the plant cover on the ground, without harvesting them. All seeded crops, as well as planting dates, were the same in both areas evaluated (tillage and no-tillage system) throughout the four years of study.

### *Data collection*

We used pit-fall traps in containers of 80 ml and seven centimetres of diameter, baited with sardines and honey (15 g and 5 g, respectively) to capture the ants. On the sides of each container, fluon-sliding liquid kept the ants in the trap. We installed 10 linear transects with three pitfall traps keeping a distance of three meters between traps of the same transect and 15 m between transects. The location of transects within the areas was randomised on a monthly basis, maintaining a distance of 10 m from the edge of areas previously sampled. The traps remained exposed in the field for 24 hours. After captured, we placed the ants in 70% alcohol and returned to the laboratory for identification. For taxonomic identification, we used dichotomous keys from Bolton (2006), as well as comparisons with specimens from the reference collection of the Insects Ecology Laboratory of the Faculdade de Ciências Biológicas e Ambientais of Universidade Federal da Grande Dourados. In addition to ants, to represent the presence or not of soil management, we also collected the dead biomass on soil in areas of 1 m<sup>2</sup> in the same location and time of removal of the ant traps. We put the dead biomass into paper bags and kept it in an oven at 40°C for 48 h. After this period, we weighted it using a balance that is accurate to 0.01 g.

### *Data analysis*

To describe and compare the ant communities of the two areas evaluated, we used data of individual abundance, species richness and diversity (Shannon-Wiener index). We used the data of dry plant biomass for inference about the structural complexity of the areas. To compare the ant abundance and the amount of dry plant biomass between the tillage systems and during the four years after implementation of each tillage system, we used the average values of the three sampled points in each transect, totalling 10 independent monthly samples in each area. For comparisons of the richness and diversity between areas and among the years after the implementation of each tillage system, we used one monthly value from the 10 monthly samples. In these later comparisons, each year had 24 samples (corresponding to 12 months for each area), with the exception of the fourth year, with 28 samples (14 months for each area).

For these comparisons, we used Generalized Linear Models (GLM). We chose this approach because the assumptions of the model related to a completely randomised design with two factors (type of planting and year of implementation), in a split plot design, were not attended. In this study, we modelled continuous variables, dry plant biomass and diversity, from the Gamma distribution with the logarithmic link function, and discrete variables (abundance and richness) from the Poisson distribution with the logarithmic link function. We defined “linear” for this study’s GLM as a completely randomised design with two factors (area and year of implementation) in a split-plot design predictor. For comparison among means, we

used the Wald test at a significance level of 5%. We conducted these analyses using the software SPSS 17.0. In addition, we related the monthly values of abundance, richness and diversity with the monthly values of dry plant biomass, using the Spearman correlation.

In order to verify the similarity of the composition of ant species throughout the months ( $n = 100$ ; comprising the 50-month study in both areas), we performed the Non-Metric Multidimensional Scaling (NMDS), and later, the “joint plot”. Then, we related these sorts of scores, through Pearson’s correlation, to the data regarding the monthly mean biomass. Values were considered significant only if

**Table 1.** Relative frequency of ant species and rank of species by year in areas of no-tillage (NT) and conventional tillage (C) ( $n = 500$  per area) during the period from November 1999 to December 2003 in Dourados, MS, Brazil. Numbers from 1 to 4 represent the four years evaluated. Species with the same frequency have the same hierarchical placement in the rank.

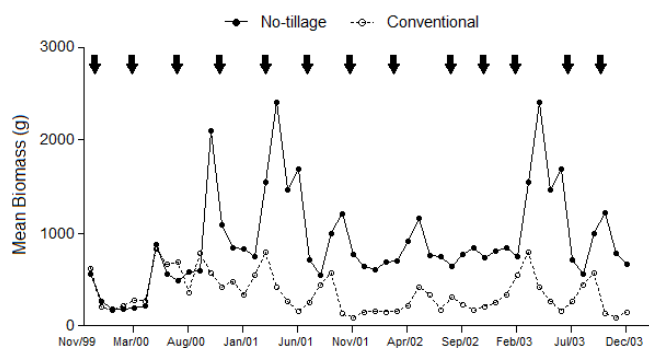
Subfamilies/Species	Relative Frequency (%)		Rank of species by year							
	NT	C	No-tillage				Conventional			
			1	2	3	4	1	2	3	4
<b>Formicinae</b>										
<i>Brachymyrmex</i> sp1	1.64	0.95	13	14	12	7	13	-	9	12
<i>Camponotus</i> sp1	0.82	0.47	14	12	15	12	13	12	-	13
<i>Camponotus</i> sp2	0.50	0.16	-	-	13	12	-	12	-	14
<i>Camponotus</i> sp3	0.57	0.24	-	16	16	11	13	12	-	14
<b>Myrmicinae</b>										
<i>Atta</i> sp1	3.15	3.16	8	8	11	5	10	9	11	5
<i>Crematogaster</i> sp1	1.14	1.18	11	11	-	10	9	12	12	12
<i>Pheidole</i> sp1	15.14	15.01	2	3	2	2	2	2	3	2
<i>Pheidole</i> sp2	8.20	6.48	4	4	6	6	5	4	7	7
<i>Pheidole</i> sp3	6.12	5.77	5	5	8	9	6	5	5	6
<i>Pheidole</i> sp4	0.82	0.95	14	15	13	11	-	10	-	9
<i>Pheidole</i> sp5	5.55	4.27	7	7	5	6	8	6	6	10
<i>Solenopsis</i> sp1	23.22	30.73	1	1	1	1	1	1	1	1
<i>Solenopsis</i> sp2	2.21	0.95	9	10	10	8	11	12	12	11
<i>Myocepurus goeldii</i> (Forel, 1893)	0.95	0.47	14	13	13	11	13	12	10	14
<i>Hylomyrma</i> sp1	1.20	0.32	12	14	17	8	13	-	-	14
<i>Carebara</i> sp1	0.06	-	-	-	-	13	-	-	-	-
<b>Ectatomminae</b>										
<i>Ectatomma planidens</i> Borgmeier, 1939	13.19	12.09	3	2	3	3	3	3	4	3
<i>Ectatomma tuberculatum</i> (Olivier, 1792)	4.29	3.32	6	9	7	9	7	8	8	8
<b>Ponerinae</b>										
<i>Neoponera obscuricornis</i> Emery, 1890	0.44	0.08	15	16	15	13	13	-	-	-
<i>Odontomachus haematodus</i> (Linnaeus, 1758)	0.50	0.24	13	-	14	-	12	-	-	-
<b>Pseudomyrmicinae</b>										
<i>Pseudomyrmex</i> sp1	0.32	-	15	-	17	11	-	-	-	-
<b>Dorylinae</b>										
<i>Nomamyrmex</i> sp1	0.63	0.95	15	14	15	12	-	12	12	-
<i>Neivamyrmex</i> sp1	0.06	0.24	-	-	17	-	-	-	10	-
<b>Dolichoderinae</b>										
<i>Dorymyrmex</i> sp1	6.88	10.35	7	6	4	4	4	7	2	4
<i>Linepithema</i> sp1	2.33	1.50	10	8	9	10	13	11	8	12
<i>Tapinoma</i> sp1	0.06	0.16	-	-	17	-	13	-	12	-
<b>Species richness</b>	<b>26</b>	<b>24</b>	<b>21</b>	<b>20</b>	<b>24</b>	<b>23</b>	<b>20</b>	<b>18</b>	<b>17</b>	<b>19</b>

$r^2 \geq 0.3$ , according to McCune and Mefford (2006). The Bray-Curtis' similarity index was used in this analysis, from an array of values with a monthly relative frequency of species in the samples. For these analyses, we used 250 randomisations, and the significance was tested with the proportion of randomisations, with stress less than or equal to the observed stress. For this analysis, the software PC-ORD 5.10 was used (McCune & Mefford, 2006).

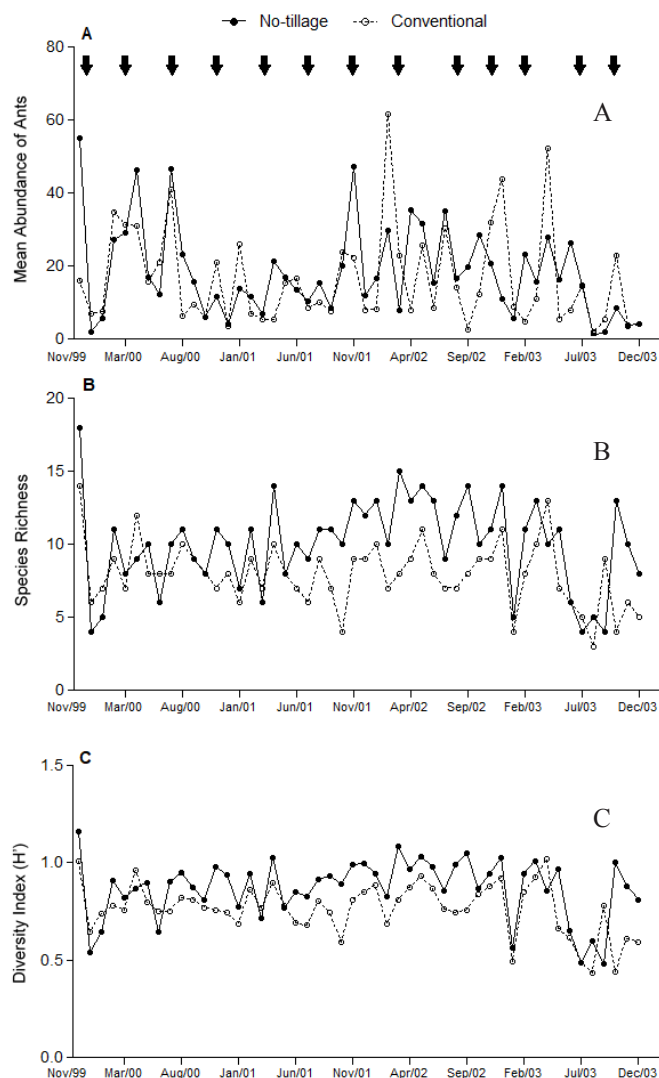
## Results

A total of 52,050 individuals were collected (27,480 in no-tillage and 24,570 in conventional tillage). These individuals represented 26 species, 18 genera and seven subfamilies located in the two areas evaluated during the four years of this study. The number of species found in the two areas were similar (26 in no-tillage, and 24 species in conventional) for most of the months evaluated (Table 1). However, some species, such as *Oligomyrmex* sp1 and *Pseudomyrmex* sp1, were found only at the no-tillage site. Other species, such as *Pachycondyla obscuricornis* (Emery, 1890) and *Odontomachus haematodus* (Linnaeus, 1758), initially found in two areas, were only present in the area with no-tillage after the second year of evaluation. According to the rank of species shown in Table 1, we found a predominance of species of the subfamily Myrmicinae during the four years; among them, the genera *Solenopsis* and *Pheidole* were the most abundant and frequent in both areas (Table 1).

Due to the interaction between factors, the type of planting and the year of implementation were significant for the variables dead plant biomass and ant abundance. As a result, we evaluated the interaction effect by unfolding the type of planting within the year of implementation and vice-versa (Table 2). From the results shown in Table 2, using the Wald test and a significance level of 5%, it is possible to verify that the mean biomass in no-tillage sites was higher than the found in conventional tillage, with the exception of the first year of implementation, which showed no significant difference between areas (Fig 1 and 2). The mean abundance



**Fig 1.** Monthly variation of mean dead plant biomass during the four years of study in the area of no-tillage and conventional tillage in Dourados, MS, Brazil. The black balls and solid lines indicate no-tillage, and the white balls and dashed lines indicate conventional tillage. The arrows indicate the months when the cultures were harvested.



**Fig 2.** Monthly variation, mean abundance of ants (A), species richness (B) and diversity index (C) during the four years of study in the area of no-tillage and conventional tillage in Dourados, MS, Brazil. The black balls and solid lines indicate no-tillage, and the white balls and dashed lines indicate conventional tillage. The arrows indicate the months when the cultures were harvested.

of individuals was higher in the no-tillage area every year, except the last year. The highest values of abundance of individuals for conventional tillage were found in the first and third years, respectively; and for no-tillage also, the first and third years had highest abundance of individuals.

Due to the interaction between factors, the type of planting and year of implementation were not significant for the variables species richness and diversity; we evaluated the effect of the factors of type of planting and year of implementation in an isolated way. The monthly mean richness and diversity of species in conventional tillage were lower than those of no-tillage, and the mean monthly richness in the third year of implementation was higher for both types of planting (Table 3). Moreover, there is no significant difference between the mean richness for the first, second and fourth years of

**Table 2.** Mean of dead plant biomass and ant abundance based on planting type and year since implementation of no-tillage system. Lower case letters represent differences in years and capital letters represent differences between planting types. Wald test at 5% significance.

Response variable	Planting type	Year since implementation			
		First	Second	Third	Fourth
Dead plant biomass	Conventional	170.40aA	146.38abB	84.83cB	12325bB
	No-tillage	202.24cA	403.42aA	269.39bA	373.07aA
Ant abundance	Conventional	54.24aB	35.71cB	53.52aB	44.64bA
	No-tillage	70.79bA	38.01cA	73.09aA	38.23cB

implementation. For diversity, the mean value was higher in the third year of implementation and lower in the fourth year, without any significant difference between diversity values for the first and second years of implementation.

When analysing the correlation between the abundance of individuals and dry plant biomass, there was no correlation observed (Spearman’s  $r = -0.003$ ;  $p = 0.97$ ;  $n = 100$ ). However, a significant positive correlation was found between species richness and dry plant biomass (Spearman’s  $r = 0.39$ ;  $p < 0.01$ ;  $n = 100$ ). We also observed that the highest values for dead plant biomass, total abundance, species richness and diversity were not concentrated at, or near, the harvest period (Fig 1 and 2).

We did not find relationships between the distribution of monthly samples formed by the composition and frequency of ant species (Fig 3) and the mean monthly total quantity of dead plant biomass (Pearson’s  $r = 0.041$ ;  $p = 0.045$  for axis 1 and  $r = 0.074$ ;  $p = 0.69$ ;  $n = 100$  for axis 2).

**Discussion**

*Community composition*

The composition of the two communities evaluated in this study was similar. The subfamily Myrmicinae (12 species) was the most abundant and frequent in all sampled months. According to Hölldobler and Wilson (1990), this group is the largest subfamily of Formicidae in number of species and variety of food and nesting habits, with wide adaptation to disturbed environments. Furthermore, many authors have shown that most Myrmicinae species are aggressive and they have efficient and massive recruitment, being of ten sampled

**Table 3.** Mean of species richness and diversity on planting type and year since implementation for no-tillage system. Lower case letters represent differences between years and capital letters represent differences between planting types. Wald test at the 5% significance.

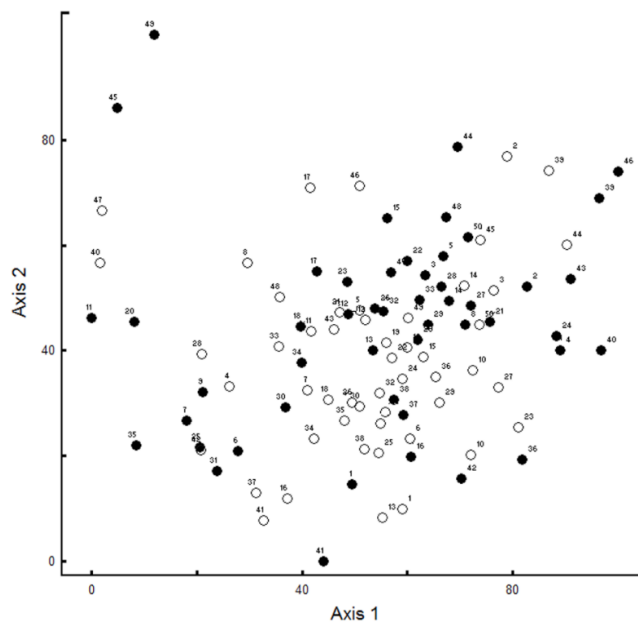
Response Variable	Factor	Factor Level			
Richness	Planting type	Conventional	No-tillage		
		2.52b	3.16a		
	Year since implementation	First	Second	Third	Fourth
		2.78b	2.76b	3.21a	2.58b
Diversity	Planting type	Conventional	No-tillage		
		0.76b	0.87a		
	Year since implementation	First	Second	Third	Fourth
		0.81b	0.82b	0.91a	0.73c

in agrosystems (Castro & Queiroz, 1987; Perfecto, 1991; Lange et al., 2008; Falcão et al., 2015). They are considered to be effective agents in the biological control of pests in crops (see Fernandes et al., 1994, 2012).

In this study, species of the genera *Solenopsis* and *Pheidole* were most frequent in both areas, occupying 38.36% of the samples in no-tillage and 45.74% in conventional farming. These two genera may be considered as dominant in this study. Agrosystems, exhibiting lower structural complexity than natural environments, may be predisposed (Castro & Queiroz, 1987) because of their vulnerability to colonization by more generalist and aggressive species, such as species of the genera *Pheidole*, *Solenopsis* and *Ectatomma*.

On the other hand, the low frequency of species of the genera *Nomamyrmex* and *Neivamyrmex* may be explained by their nomadic behaviour (Hölldobler & Wilson, 1990). The genus *Brachymyrmex*, which was also found in low frequencies, is common in Neotropical forests, and their nests may be found in small cavities under epiphytes, or even in leaf litter. Individuals of *Brachymyrmex* have small body sizes and are hardly dominant species, but they can get resources because even in the presence of larger species, they show an opportunistic behavior (Agosti et al., 2000).

Species of the genera *Camponotus*, *Crematogaster* and *Pseudomyrmex* are commonly found foraging on vegetation where they can nest and establish mutualistic associations with their host (Rico-Gray & Oliveira, 2007). As the herbaceous cropping system employed in this study is not considered an appropriate environment for these species, they were also



**Fig 3.** Ordination in two dimensions (Axis 1 indicate dimension 1, and Axis 2 dimension 2) of monthly samples using the Non-Metric Multidimensional Scaling (NMDS) of Bray-Curtis distances at relative frequencies of ants species. The black balls indicate conventional tillage and the white balls indicate the no-tillage. The 50 monthly observations of each area were numbered in chronological order, from November 1999 to December 2003.

found in small number in this study. Other genera, such as *Pachycondyla* and *Odontomachus*, have solitary foraging, and exhibit nests with few individuals (Hölldobler & Wilson, 1990). Therefore, these species are often found at low frequencies in studies that use traps. In addition, they were found only in the first year in both areas, and posteriorly only in the no-tillage. This fact may be related directly to the increased availability of food offered by the no-tillage. Because of the solitary strategy, foraging individuals of these genera hardly dominate the resources and are considered to be opportunistic (Silvestre et al., 2003).

#### *Ant communities and habitat complexity*

The high similarity of the two ant communities evaluated in this study may be related to the fact that before the implementation of no-tillage, the two areas were conventional tillage. However, with the implementation of no-tillage, and increasing the amount of dry plant biomass in the soil in the no-tillage area, the properties of the communities (abundance of individuals and species richness) were being changed from the second year of implementation, becoming significantly differences in the third year of the study.

The highest total abundance of individuals was found in the area with no-tillage. According to Kladvko (2001), there is a wider variety of species with higher abundance on dead plant biomass in no-tillage than in conventional methods. The same authors showed that most organisms are sensitive to the soil preparation, due to physical changes and the amount of water that result from the incorporation of residues from the previous crop into the soil. Even though the abundance of ants is not a metric used in studies about community structure due to variation in species behaviour (Longino et al., 2002), in this study, this metric (abundance of ants) should be taken into consideration because the impact of soil management directly affects the survival of colonies and individuals. Thus, the abundance of ants, as well as other organisms, relates to the impact of soil management on the fauna in agrosystems. This fact may be evidenced in this study during the first year. However, when we observe the abundance of ants in the traps over the months evaluated, it is impossible to demonstrate a clear difference between areas. This result may be related to the enormous biological variation of the species and their seasonal variations in resource foraging (see Lach et al., 2009).

However, when we compare the richness and diversity of species between the two areas, we observe a clear pattern of higher values for the no-tillage area. Initially, both areas had similar richness, which were differentiated after the first year of study, reaching significant values in the third year. This result supports our hypothesis that dry plant biomass present in the soil, derived from the previous cultures, provides a more complex habitat when compared to conventional tillage.

The relationship between the diversity of ant species and the structural complexity of the habitat was also evidenced in many studies (Lassau & Hochuli, 2004; Ribas et al., 2003; Dáttilo & Izzo, 2012; see also Rico-Gray & Oliveira, 2007).

The main factors influencing the increase in ant diversity include a variety of nesting sites, the amount of food available, the foraging area and the competitive interactions (Kaspary et al., 2000; Ashford et al., 2013). The increased availability and variety of food enables the establishment of the richest ant communities and with more co-occurrence among species (i.e., less competition) (Sharley et al., 2008; Lange et al., 2008). This result was shown in this study based on the data on ant abundance and richness found in the samples.

In a broader scenario, there are good reasons to relate the abundance and diversity of predatory insects, such as ants, to the natural conservation in agrosystems (see Cividanes & Yamamoto, 2002; Kladvko, 2001; Mas & Verdú, 2003). This is because the greater abundance and richness of predators decrease populations of pest insects (Hooper et al., 2005; Christiaans et al., 2007) and reduces the use of pesticides (Booij & Noorlander, 1992; French et al., 1998). The greater the number of predator species with different diet and diverse foraging strategies, stronger will be the effects on herbivores and plants (e.g. Hooper et al., 2005; Naeem & Li, 1998; Schmitz & Suttle, 2001). A meta-analysis of studies in natural and agricultural systems showed that the greater the diversity of predators, the lower the herbivore community, and consequently, the growth and reproduction of plants increase (Schmitz et al., 2000). Besides affecting the community of herbivores, ants may also reduce fungal pathogens, eliminating spores (de la Fuente & Marquis, 1999) or restricting the interactions between plants and disease vectors (Khoo & Ho, 1992).

The increase in dry plant biomass in the soil led to a gradual increase in the richness and abundance of ants in no-tillage area when compared to conventional tillage. Our study brings important information on ant communities in two cropping systems over a long period, providing interesting results that have not yet been reported in other studies. We showed that the no-tillage system results in an environment that promotes the greater diversity of ant species when compared to conventional tillage. Additional similar studies should be conducted in order to better understand the dynamics of communities, both in agrosystems and natural systems. In addition, more focus should be given to the areas with agrosystems, since they are often left out, making studies in protected areas (natural) more common (Martin et al., 2012).

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