



# Effect of Good Agricultural Practices under no-till on litter and soil invertebrates in areas with different soil types



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## ABSTRACT

Good Agricultural Practices (GAPs) under no-till (NT) includes a mixed crop rotation; cover crops; integrated pest, weed and disease management; nutrient restoration; and a rational use of agrochemicals. When applied all together, GAPs promotes high productivity, while maintaining the production capacity of resources. In the Pampas region of Argentina, there is a need to assess the effects of these practices on soils, particularly on soil fauna, as they play an important role in soil functioning. The aim of this study was to evaluate the effect of the application of GAPs under NT on invertebrates and to assess whether this effect is different between soil types. We hypothesized (1) that GAP will produce an increase in the abundance, as well as changes in the faunal composition of litter and soil invertebrates; (2) that the effects will differ with soil type, and (3) that the changes in soil invertebrate fauna will be explained by soil properties. We compared two contrasting NT treatments –with and without GAP application–, replicated in three agricultural areas, on different soil types (Entic Haplustolls to Typic Argiudolls) situated across a west–east transect in the Pampas region of Argentina. A positive (Natural environment) and a negative (Conventional tillage) reference sites were included in the comparison. Litter and soil invertebrates and soil properties were assessed at each sampling site. Overall, our results indicated that the application of GAPs in productive NT fields increases litter and soil invertebrate abundance and modifies faunal composition. In the litter layer, four of the five taxa present were favoured by GAPs with an increase in the abundances of ants, prostigmatid mites, earthworms and collembolans. GAPs also induced changes in invertebrate faunal composition, from the initial NO-GAP situation to the present state under GAP system. The observed changes in litter and soil invertebrates, changes in faunal abundance and composition can be expected to translate to changes in soil functioning. Our last hypothesis was partially confirmed in that soil properties have to be considered in the examination of differences in fauna between treatments with there are only subtle differences in practices, as in the present study.

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## 1. Introduction

No-till (NT) has been widely adopted across the entire Pampas region of Argentina, including areas previously considered not highly productive. NT currently accounts for over 78% of the total area cultivated with soybean (*Glycine max*), maize (*Zea mays*), sunflower (*Helianthus annuus*), wheat (*Triticum aestivum*) and sorghum (*Sorghum bicolor*) (AAPRESID, 2012; Albertengo et al., 2013). Soybean and maize are the dominant crops, with more than 20 and 6 million hectares cultivated in the 2012/2013 crop cycle,

respectively (MAGyP, 2014). Interestingly, even when soil cover crops and appropriate rotation schedules under NT are recognized as necessary to achieve all the NT benefits for soil quality, most farmers only grown single species crops, use NT seeders and a chemical winter fallow. These practices have led to physical, chemical and biological soil degradation even under NT (Díaz-Zorita et al., 2002; Parra et al., 2009; Domínguez et al., 2010; Bedano et al., 2011). In response to a decline in soil quality, a group of farmers organized the Argentine No-till Farmers Association (AAPRESID) and started to adopt and promote crop species rotation; cover crops; integrated pest, weed and disease management; nutrient restoration; and a rational use of agrochemicals as an integral part of a NT system. Together these practices are called

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“Good Agricultural Practices” (GAPs), in accordance with the definition of the Food and Agricultural Organization (FAO) described in [Poisot et al. \(2004\)](#). Nowadays, AAPRESID considers that only when all the GAPs are implemented is management considered a sustainable “no-till system” achieving high productivity, while maintaining the production capacity of resources ([Albertengo et al., 2011](#)). Farmers from AAPRESID have reported higher yields when GAPs are applied as an integral part of the NT system.

The important role of soil fauna in soil functioning is well known, in particular in the formation of stable soil aggregates, pore size and function, the production and decomposition of organic matter, and population stability of the various soil inhabiting organisms ([El Titi, 2003](#)). Soil fauna can be separated into mesofauna and macrofauna according to their body width. Soil mesofauna (0.1 and 2 mm) is dominated by mites (Acari) and springtails (Collembola), which are among the most abundant and widespread soil arthropods in most soils and both have important roles in soil organic matter cycling through their feeding activities ([Bedano et al., 2006a,b](#)). Soil macrofauna (>2 mm) includes earthworms, ants, termites, and beetles, and are important in both organic matter cycling and soil structure formation ([Lavelle et al., 2006](#)).

With sustainable agricultural practices becoming a priority for farmers and the general public alike, a more complete understanding of the soil ecosystem is needed ([Stubbs et al., 2004](#)). In the Pampas region, the dissemination of GAP's benefits under NT by AAPRESID, does not include an assessment of the effects of these practices on soils and particularly on soil fauna. In this region, there is evidence showing a negative effect of NT -without GAP- on soil macrofauna (e.g., [Domínguez et al., 2010](#)) and mesofauna ([Arolfo et al., 2010](#)). To date there has been no evaluations of GAP applications on reversing the decline in soil quality.

Soil development is governed by five different soil-forming factors, namely climate, vegetation, relief, parent material, and time ([Jenny, 1941](#)). Within a continuum of possibilities, there are recognizable soil types that originate, depending on variations in these factors, which largely determine the dominant physical and chemical soil properties ([Kibblewhite et al., 2008](#)). Evaluation of GAPs on soil must recognize the natural differences between soil types. Here, we compared two contrasting NT treatments, which were replicated in three agricultural areas with different soil types, across a west-east transect in the Pampas region of Argentina. The treatments consist of a “no-till system”, where NT with Good Agricultural Practices is applied (GAP), and a NT system without GAP application (NO-GAP). For both treatments the management history of each plot is well documented by farmers.

This study is part of BIOSPAS project (Biology of Soil and Sustainable Agricultural Production, [www.biospas.org](http://www.biospas.org)), a multi-disciplinary research project aiming to find biological indicators of sustainability under NT farming by means of a polyphasic description ([Wall, 2011](#)). Previous studies ([Duval et al., 2013](#)) found no differences in SOM concentration in the top 10 cm between GAP and NO-GAP treatments. There were also no differences in soil bulk density. A biological indicator was developed that discriminate between GAP and NO-GAP soils. This was the ratio between the abundance of a selected group of bacteria within the GP1 group of the phylum Acidobacteria and the genus *Rubellimicrobium* of the phylum Alphaproteobacteria ([Figuerola et al., 2012](#)). Agricultural management was also found to have a strong influence on  $\beta$ -diversity patterns, with the NO-GAP having a significantly lower  $\beta$ -diversity and narrower breadth compared with GAP, because of loss of endemic taxon groups ([Figuerola et al., 2014](#)). Soil fatty acid profiles from Phospholipids (PLFA) and Neutral Lipids (NLFA) fractions clearly discriminated between GAP and NO-GAP, whereas GAP soils were particularly

characterised by higher concentrations of the fatty acid 20:0 and total NLFAs concentration in winter ([Ferrari et al., 2015](#)).

To the best of our knowledge, the effects of GAPs on litter and soil invertebrates have not been systematically investigated. Therefore, the objective of this study was to evaluate the effect of the application of GAPs under NT on litter and soil invertebrates and to assess whether this effect differs between soils types. We hypothesized that (1) GAP will produce an increase in the abundance, as well as changes in the faunal composition of litter and soil invertebrates; (2) that the effects will differ between soil types, and (3) that the changes in soil invertebrate fauna will be explained by associated changes in soil properties.

## 2. Materials and methods

### 2.1. Study sites

The study sites were located in the most productive zone of the Pampas Region of Argentina, at Bengolea (Córdoba province; 33° 01' 32.9" S, 63° 37' 36.4" W), Monte Buey (Córdoba province; 32° 58' 17.0" S, 62° 27' 02.4" W) and Pergamino (Buenos Aires province; 33° 56' 42.6" S, 60° 33' 35.6" W) ([Fig. 1](#)). In Bengolea and Monte Buey the climate is temperate subhumid with a mean annual temperature of 17°C; in Pergamino the climate is temperate humid with a mean annual temperature of 16°C. Mean annual precipitation is 870, 910 and 1000 in Bengolea, Monte Buey and Pergamino, respectively. The slope in all sites is lower than 0.5% and the altitude is on average 223, 110 and 66 m a.s.l. in the three areas, respectively.

The sites were selected according to soil type, from the Entic Haplustolls (sandy loam) in Bengolea, Typic Argiudolls (silty loam) in Monte Buey, to the Typic Argiudolls (silty clay loam) in Pergamino. The three sites have soil types with increasing clay and decreasing sand concentration from Bengolea (west) to Pergamino (east).

### 2.2. Treatments

The treatments were defined according to a set of definitions of GAPs provided by FAO ([www.fao.org/prods/GAP/index\\_en.htm](http://www.fao.org/prods/GAP/index_en.htm)) and AAPRESID (<http://www.aapresid.org.ar/ac/wp-content/uploads/sites/4/2013/02/manual.pdf>), described in [Poisot et al. \(2004\)](#) and [Albertengo et al. \(2011\)](#). The final treatments and study sites were defined after thoughtful discussion between the scientists and the farmers participating of the BIOSPAS project. Four treatments were defined: (1) Good agricultural practices under NT (GAP): subjected to intensive crop rotation (including winter cover crops), nutrient replacement, and minimized agrochemical use (herbicides, insecticides and fungicides) ([Table 1](#)); (2) No-till management without good agricultural practices (NO-GAP): high crop monoculture (soybean), low nutrient replacement and high agrochemical use (herbicides, insecticides and fungicides) ([Table 1](#)); (3) Conventional tillage (CT): Mouldboard and disc ploughing, low nutrient replacement and high agrochemical use; (4) Natural Environment (NA): undisturbed natural grassland adjacent to the cultivated treatments (less than 5 km), where no cultivation was practiced for (at least) the last 30 years. Both the NA and CT reference sites, were located near the NT treatment.

The treatments were replicated three times in the three agricultural regions with different soil types, situated across the west-east transect described previously, with the exception of CT, which was not available in Bengolea. [Table 1](#) summarizes the information on the agricultural practices and crop yields of the different study sites. All sites had been under NT for at least five years before sampling (100% of NT), with the exception of a chisel

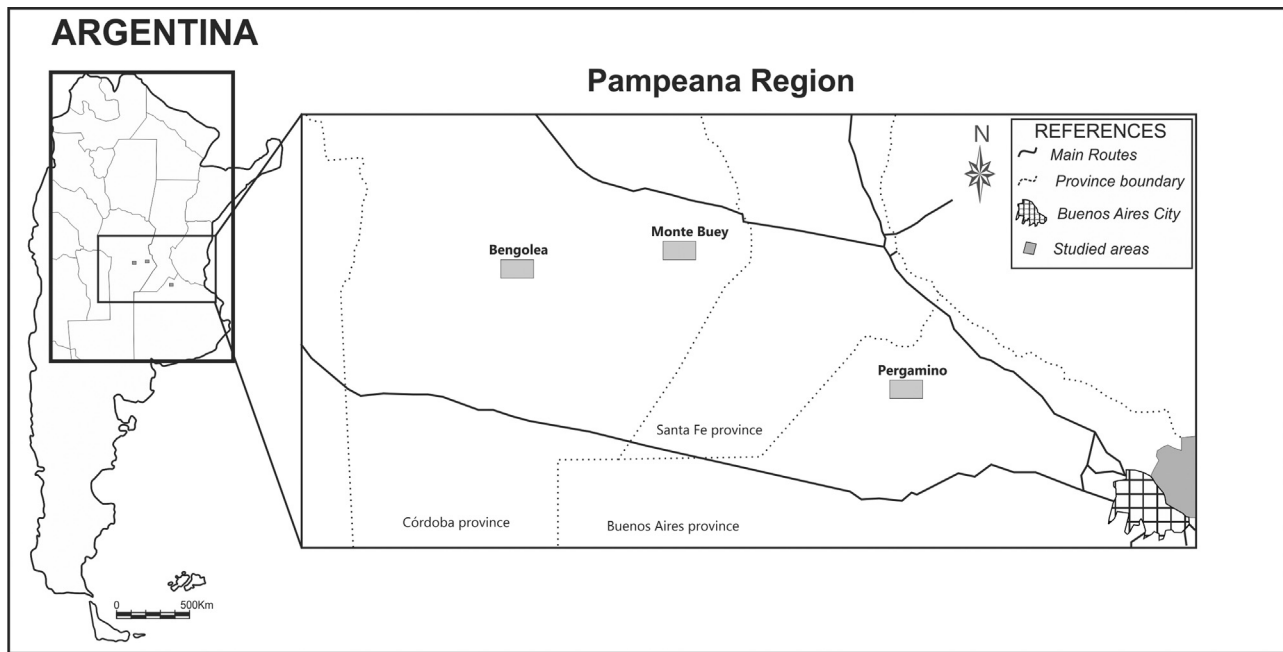


Fig. 1. Study area (see text for details).

plough application in 2004/2005 in the NO-GAP site of Bengolea (80% of NT). In the three regions, GAP had a lower rate of soybean in the crop rotation than the NO-GAP. GAP also had on average 50% of the winters with crop in the last five years, whereas NO-GAP had only 20%. GAP and NO-GAP also differed in the amount of herbicides used, with an average of 27.4 L and 43.0 L, respectively (Table 1). Yield of both crops, soybean (*Glycine max*) and maize (*Zea mays*), was on average higher in GAP (soybean: 3055 kg ha<sup>-1</sup>; maize: 10850 kg ha<sup>-1</sup>) than in NO-GAP (soybean: 2758 kg ha<sup>-1</sup>; maize: 5350 kg ha<sup>-1</sup>). Plot size was 82, 23 and 25 ha in Bengolea, Monte Buey and Pergamino, respectively.

### 2.3. Litter and soil invertebrates

Invertebrate densities were assessed in late summer 2010. On each sampling site, five sampling points were defined every 20 m along a transect with a random starting point. At each sampling point, a soil sample was taken to extract mesofauna and another

one to extract macrofauna. Samples for mesofauna were obtained using a soil corer of 10 cm in diameter and 10 cm in depth. Samples were separated into litter (from 0.5 to 2 cm in depth, depending on the treatment) and soil (10 cm in depth) and then carefully conducted to the laboratory, where mesofauna (mites and collembolans) was extracted with a Berlese apparatus for 10 days, using 40 W bulbs suspended 10 cm above the top of the samples. The organisms obtained were stored in 70% alcohol. Mites were sorted into the following suborders: Oribatida, Mesostigmata, Prostigmata and Astigmata, and counted with a stereomicroscope.

Samples for macrofauna were obtained following the TSBF method (Anderson and Ingram, 1993), by digging a soil monolith of 25 cm × 25 cm, to a depth of 30 cm, that was then separated into two layers: litter (from 0.5 to 2 cm in depth, depending on the treatment) and soil (10 cm in depth). In the laboratory, soil and litter samples were hand-sorted to collect and count invertebrates larger than 2 mm. All macrofauna was preserved in 70% alcohol except earthworms, which were fixed and preserved in 4%

Table 1

Management and crop yield in each agricultural management system and soil type.

Soil type	Sandy loam		Silty loam			Silty clay loam			
	GAP	No-GAP	GAP	No-GAP	CT	GAP	No-GAP	CT	
Management system									
% No-tillage	100	80	100	100	0	100	100	0	
Soybean ( <i>Glycine max</i> )/maize ( <i>Zea mays</i> ) ratio	1.5	4	0.67	4	– <sup>d</sup>	1.5	5	– <sup>d</sup>	
% Winter with wheat ( <i>Triticum aestivum</i> ) <sup>a</sup>	60	40	60	20	0	40	0	0	
% Winter cover crops <sup>b</sup>	20	0	40	0	0	0	0	0	
Herbicide (L) used <sup>c</sup>	27.7	43.8	25.2	38.9	NA	29.3	46.5	NA	
Fertilizer (kg ha <sup>-1</sup> ) <sup>e</sup>									
	N	57.4	2.8	81.4	19.6	NA	37.4	0	16.2
	P	30.4	3.2	40	7.8	NA	34	0	5
	S	8.2	0	0	1.6	NA	1.2	0	0
Soybean yield (kg ha <sup>-1</sup> )	3067	2775	3167	2675	NA	2933	2825	NA	
Maize yield (kg ha <sup>-1</sup> )	10500	2700	12550	8000	– <sup>f</sup>	9500	– <sup>f</sup>	– <sup>f</sup>	

Data are average for 5 years (2005–2009).

<sup>a</sup> Percentage of winters that wheat was planted as a winter crop.

<sup>b</sup> Percentage of winters that a cover crop (*Vicia* sp., *Melilotus alba* or *Lolium perenne*) was planted. Cover crops are chemically burned before summer crops are planted.

<sup>c</sup> Calculated as liters of low-toxicity herbicides plus liters of moderate-toxicity herbicides weighted by two. Toxicity was defined according to EPA Toxicity Categories. The most frequently used herbicides were glyphosate, 2,4-d amine and atrazine.

<sup>d</sup> Because no maize was planted in the previous 5 years, the rate is the maximum.

<sup>e</sup> Calculated as the average of the previous 5 years. 6: no maize was planted in the previous 5 years. NA: data not available. Modified from Figuerola et al. (2012).

formalin. Organisms were sorted into the following taxa: earthworms (Oligochaeta: Lumbricina), potworms (Oligochaeta: Enchytraeina: Enchytraeidae), ants (Hexapoda: Insecta: Formicidae), beetles (Hexapoda: Insecta: Coleoptera), termites (Hexapoda: Insecta: Isoptera), insect larvae (Hexapoda: Insecta), Millipedes (Myriapoda: Diplopoda), Centipedes (Myriapoda: Chilopoda) and spiders (Arachnida: Araneae).

#### 2.4. Soil physical, chemical and physicochemical properties

Next to each faunal sampling point, an undisturbed soil core (0–10 cm) was extracted to measure soil bulk density and water content. In the laboratory, soil cores were weighed to obtain first humid and then dry weight (until constant weight, at 105 °C) and soil water content and bulk density were calculated. From the remaining soil of each faunal sample, an aliquot was used to measure soil organic matter (SOM) concentration (using the Walkley–Black method) and soil pH (using the potentiometric method, soil–water relationship 1:2.5).

#### 2.5. Statistical analyses

A generalized linear mixed model (GLMM) was used to assess the effect of treatments on invertebrate abundances. Poisson error distribution and log link function were used, according to the distribution of abundance data. The management system was the assessed fixed factor, and soil type used as a random parameter. A posteriori tests were performed using the DGC test (Di Rienzo et al., 2002). The random coefficients and the estimated best linear unbiased predictors (BLUPs) of the random effects were used to account for the influence of the random factor (Pinheiro and Bates, 2000). The abundance of the four identified mesofaunal taxa (Oribatid, mesostigmatid and prostigmatid mites and collembolans) and the three most abundant macrofaunal taxa (Earthworms, ants and beetles) were statistically analysed.

To evaluate the significance of the differences in soil properties among treatments a number of general linear models (GLMs) were performed, and Akaike's information criterion was used to determine the best predictive model. The fixed factor was management system. In the best-fit model, error variance structure was modelled using management system as grouping criterion and VarIdent of R's *nlme* library as variance function. A posteriori tests were performed using the DGC test (Di Rienzo et al., 2002).

A multivariate Discriminant Analysis based on the 14 invertebrate taxa (including mesofauna and macrofauna) was performed to explore in detail the differences in soil invertebrate fauna

between GAP and NO-GAP treatments in relation to soil type. This statistical procedure allowed us to discriminate samples from GAP and NO-GAP and to represent them in a space where the differences between groups are maximal (Balzarini et al., 2008). Finally, to study the association of physical and chemical soil properties with treatments and all invertebrate taxa, the Canonical Correspondence Analysis (CCA) (ter Braak and Verdonschot, 1995) was used. InfoStat software (Di Rienzo et al., 2014) was used to perform statistical analyses.

### 3. Results

#### 3.1. Litter and soil invertebrates

Earthworms and ants were not present in the litter layer. Effects of cultivation practices were significant for all invertebrate groups in both litter and soil layers (Table 2). In the litter layer, beetles were more abundant in NA, followed by both NT treatments ( $p < 0.05$ ), and were absent in CT (Fig. 2). Oribatid and prostigmatid mite abundance decreased in the following order: NA>GAP>NO-GAP>CT ( $p < 0.05$ ) (Fig. 2). Mesostigmatid mites and collembolans showed a similar pattern, but the abundance of the former in CT was higher ( $p < 0.05$ ) and the abundance of the latter was not different from that of NO-GAP ( $p > 0.05$ ).

The abundances of oribatid, mesostigmatid and prostigmatid mites and Collembola in litter were affected by soil type, shown by the high variance in the random component of the model (Table 2). Oribatid mite abundance was lower in the coarse textured soil, whereas mesostigmatid and prostigmatid mite abundances were lower in the silty clay loam soils, and collembolan abundance was lower in both extremes of the soil gradient (Table 2).

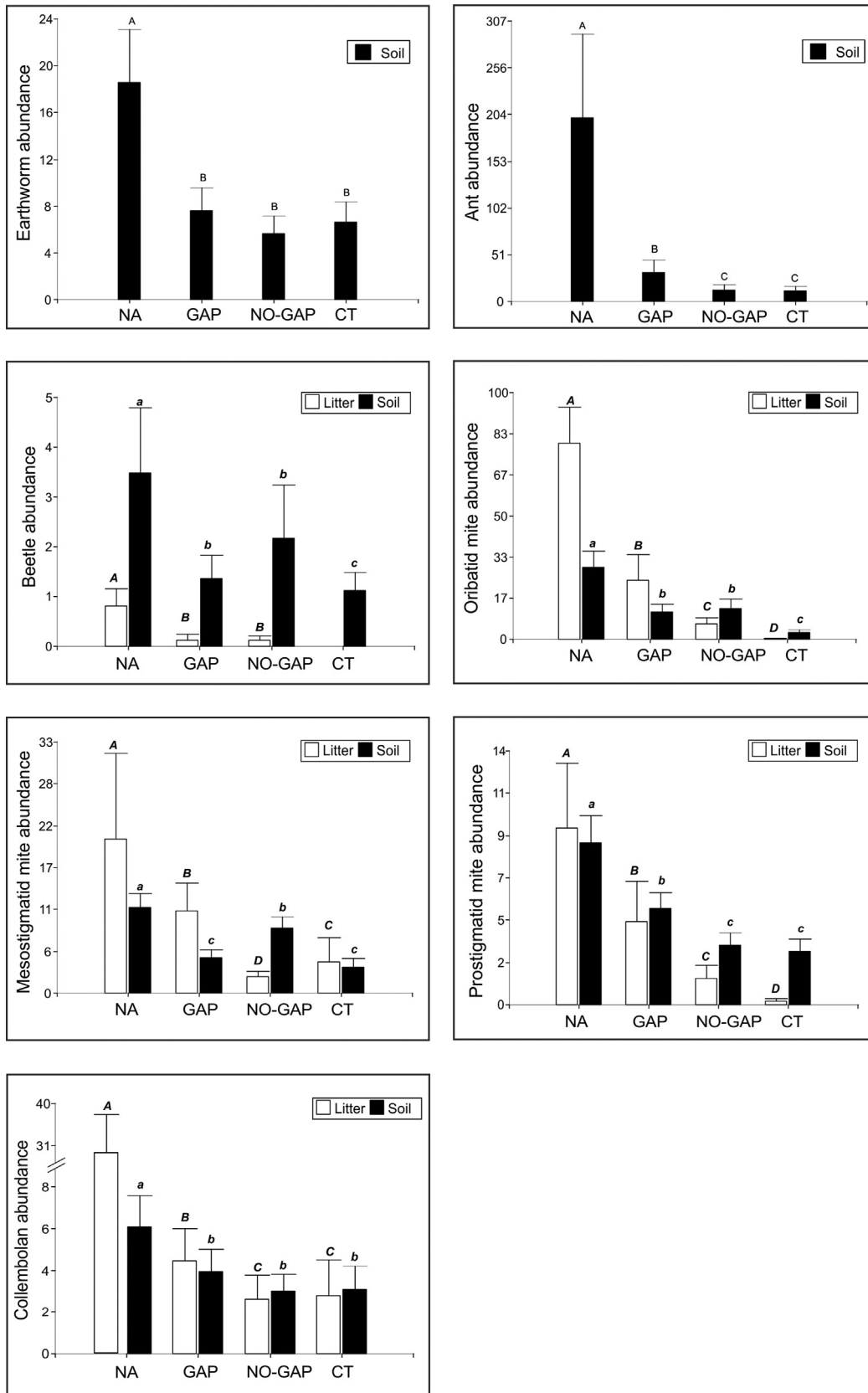
The pattern of change of soil invertebrate abundances as a function of GAP use was less clear in soil samples than in litter (Fig. 2). Earthworms and collembolans were more abundant in NA than in the cultivated sites ( $p < 0.05$ ). Both taxa tended to be more abundant in GAP than in NO-GAP, but differences were not statistically significant. Ants and Prostigmatid mites were also more abundant in NA than in the agricultural sites, and among them, were more abundant in GAP than in NO-GAP and CT ( $p < 0.05$ ). Beetles and oribatid mites were more abundant in NA, followed by both NT treatments, and were less abundant in CT ( $p < 0.05$ ) (Fig. 2). Mesostigmatid mites were more abundant in NA and less abundant in GAP and CT, with intermediate values in NO-GAP ( $p < 0.05$ ).

In soil, abundances of earthworms, ants and, to a lesser extent, mesostigmatid mites and Collembola were affected by soil type (Table 2). Earthworm and Collembola abundances were lower in

**Table 2**

Parameter estimates from the Generalized Linear Mixed Model with management (M) system as fixed factor and soil type as random factor affecting soil faunal abundances.

Faunal group	Depth	Fixed factor ( <i>p</i> value) Management system	Random factor (variance) Soil type	BLUPs for random parameter		
				Sandy loam	Silty loam	Silty clay loam
Earthworms	Soil	<0.0001	0.41	−0.56	0.34	0.23
Ants	Soil	<0.0001	0.78	0.79	−1.07	0.28
Beetles	Litter	0.0013	$3.3 \cdot 10^{-11}$	$4.5 \cdot 10^{-11}$	$4.5 \cdot 10^{-11}$	$−8.9 \cdot 10^{-11}$
	Soil	<0.0001	$1.2 \cdot 10^{-12}$	$9.1 \cdot 10^{-12}$	$−4.5 \cdot 10^{-12}$	$−4.5 \cdot 10^{-12}$
Oribatid mites	Litter	<0.0001	0.32	−0.75	0.59	0.16
	Soil	<0.0001	0.09	0.01	−0.36	0.36
Mesostigmatid mites	Litter	<0.0001	0.8	0.11	1.03	−1.13
	Soil	<0.0001	0.12	0.40	−0.42	0.03
Prostigmatid mites	Litter	<0.0001	0.18	0.06	0.48	−0.51
	Soil	<0.0001	0.01	0.04	−0.11	0.08
Collembolans	Litter	<0.0001	1.19	−0.90	1.53	−0.60
	Soil	<0.0001	0.17	−0.41	−0.10	0.53



**Fig. 2.** Litter and soil invertebrate abundance (number of individuals/site) in Natural Environments (NA), Good Agricultural Practices (GAP), No-Good Agricultural Practices (NO-GAP) and Conventional tillage (CT). Bars represent the standard deviation (SD). Different upper-case letters indicate significant differences in soil invertebrates among treatments and different lower-case letters indicate significant differences in litter invertebrates among treatments ( $p < 0.05$ ).



the coarse textured soils, whereas ant and mesostigmatid mite abundances were lower in the silty loam (Table 2).

The differences in soil invertebrate fauna between GAP and NO-GAP treatments between soil types were examined in more detail by means of a multivariate discriminant analysis based on all invertebrate taxa (Fig. 3). Axes 1 and 2 accounted for 78% of the total variation among groups (43% and 35%, for axes 1 and 2, respectively). The results showed a combined effect of treatment and soil type on faunal composition. In each soil type, samples from GAP system formed a separate group from NO-GAP samples, showing that the application of GAP produced a change in the invertebrate faunal composition. The confidence ellipses for the two treatments in each soil type were well separated, confirming the ability of this approach to discriminate between treatments. The trajectory of change of the faunal composition from the NO-GAP treatments to the GAP ones for each soil type is shown in Fig. 3.

The discriminant analysis plot grouped the high GAP with the medium NO-GAP samples as a mixed group; however, no overlapping between GAP and NO-GAP samples from the same soil type was observed (Fig. 3).

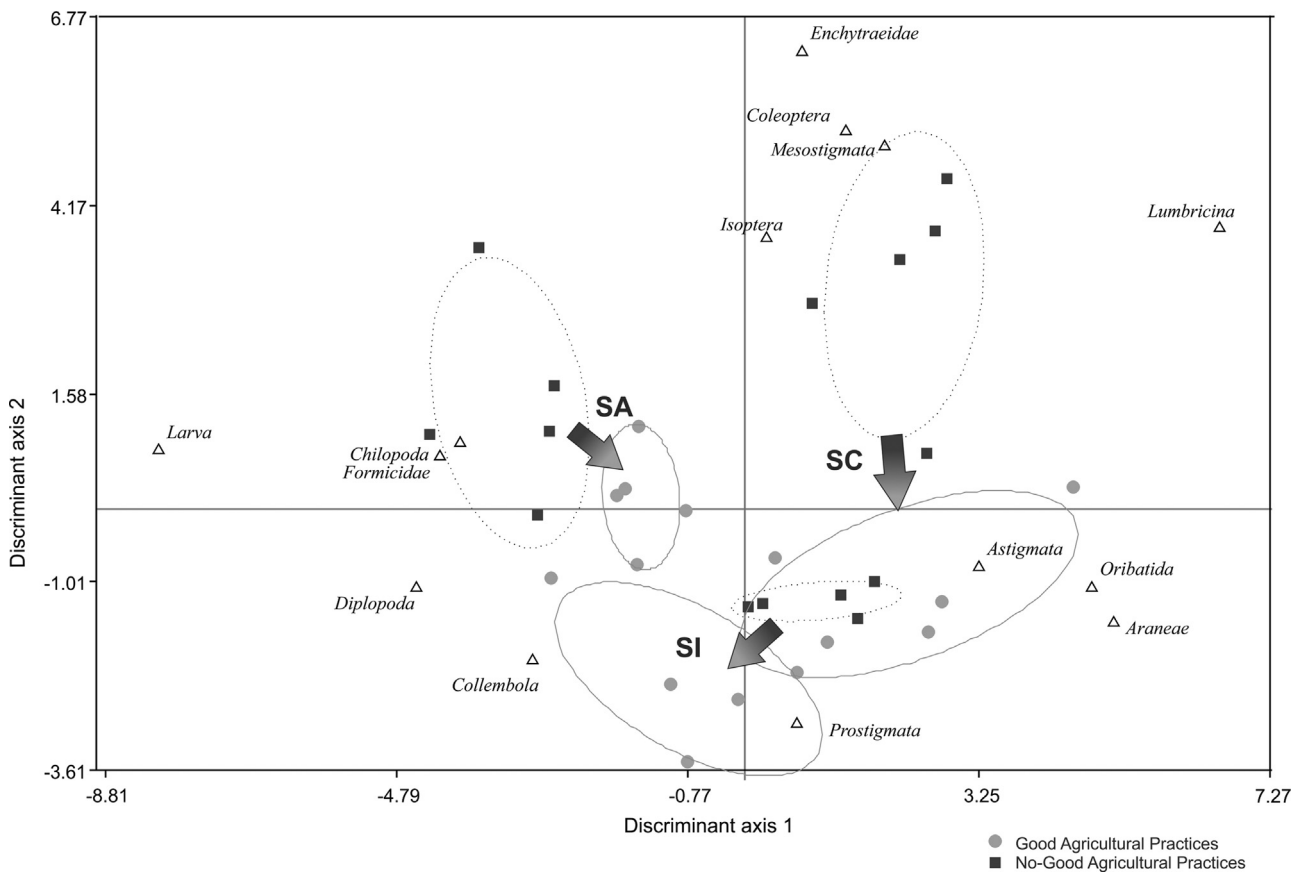
### 3.2. Soil physical and chemical properties

Cultivation had a significant effect on SOM concentration in all three soil types ( $p < 0.05$ ), but only in the sandy loam was SOM concentration significantly higher in the GAP than in the NO-GAP treatment (Table 3). There were higher SOM levels in NA than in the cultivated treatments for the silty loam and silty clay loam

(Table 3). Soil bulk density was significantly affected by treatments in the sandy and silty loam soils ( $p < 0.05$ ), showing lower values in NA than in the managed soils in both cases, with no significant differences between them. Water content of the silty loam was higher in NA than in the managed sites ( $p < 0.05$ ) and in the silty clay loam, soil water content was higher in NA and GAP than in the other treatments ( $p < 0.05$ ). Soil pH was lower in GAP than in the other treatments regardless of soil type ( $p < 0.05$ ), except in the silty loam, where pH did not differ from that of NA (Table 3).

### 3.3. Invertebrates and soil properties

The eigenvalues were 0.300 for axis 1 and 0.182 for axis 2 (Fig. 4). Environmental variables explained 68% of the variance of the data. Of this variation, 84% was explained by the first two axes, indicating a strong correlation between environmental variables and soil invertebrate composition. The first ordination axis was negatively correlated to SOM ( $-0.61$ ) and soil water content ( $-0.27$ ), and the second axis was negatively correlated to BD ( $-0.87$ ) and SOM concentration ( $-0.56$ ). The ordination of sites was influenced by both canonical axes, with axis 1 mainly separating the NA sites from the agricultural ones and axis 2 separating GAP from NO-GAP sites. Among NA sites, the effect of the soil type across axis 2 was also observed. Among agricultural systems, GAP and NO-GAP were separated, with GAP sites located closer to the NA than NO-GAP. Among GAP, sandy loam site was closer to the NA than sites from silty loam and silty clay loam, whereas among NO-GAP, silty clay loam was closer to NA.



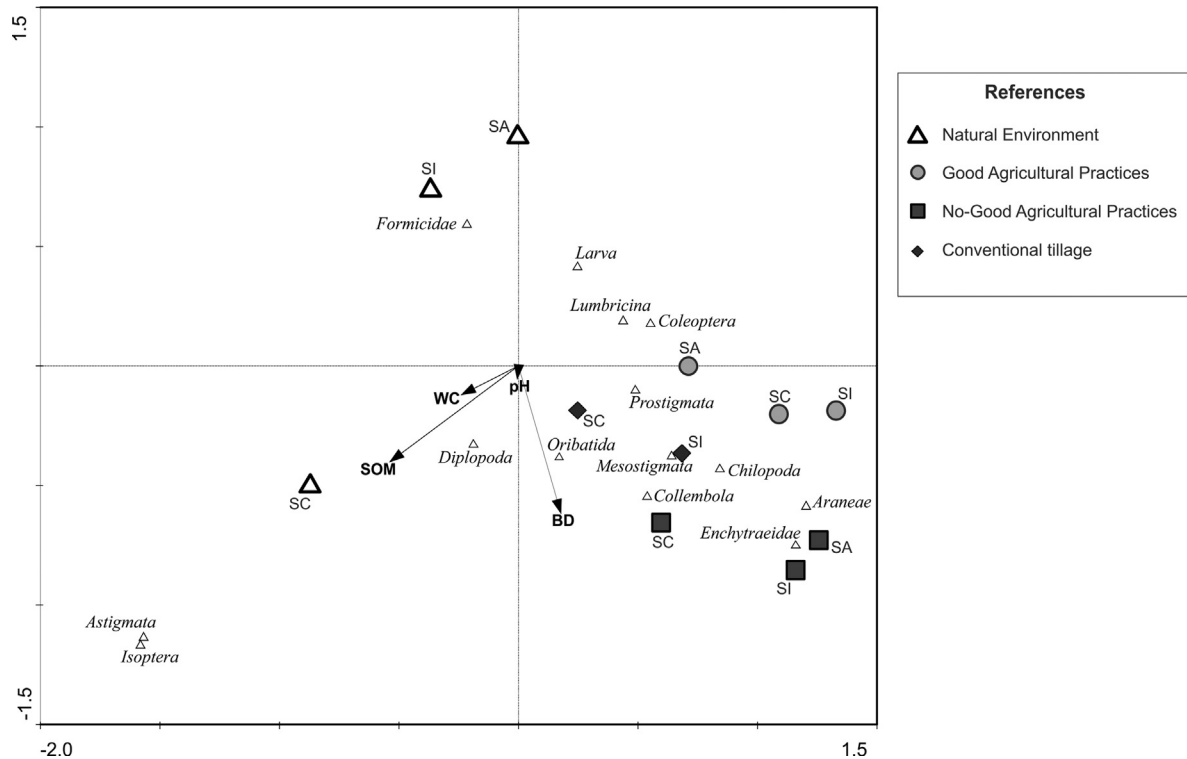
**Fig. 3.** Multivariate Discriminant Analysis of Good Agricultural Practices (GAP) and No-Good Agricultural Practices (NO-GAP) samples based on all invertebrate taxa. Treatments are identified with the following symbols and colours: GAP: light grey circles; NO-GAP: dark grey squares. Invertebrate taxa are indicated by empty triangles. The 90% confidence ellipse for each treatment in each soil type is plotted. Arrows indicate the trajectory of change of the faunal composition from NO-GAP to GAP, for each soil type. SA, SI and SC: Sandy loam, Silty loam and Silty clay loam, respectively.

**Table 3**

Physical, chemical and physicochemical soil properties in each agricultural management system and soil type.

Soil type	Sandy loam			Silty loam				Silty clay loam			
	NA	GAP	No-GAP	NA	GAP	No-GAP	CT	NA	GAP	No-GAP	CT
Soil classification <sup>a</sup>	Entic Haplustoll			Typic Argiudoll				Typic Argiudoll			
Texture	Sandy loam			Silty loam				Silty clay loam			
Organic mater (%)	1.95 b	2.88 a	2.30 b	4.25 a	2.80 b	2.58 b	2.72 b	5.43 a	3.35 b	3.36 b	3.18 b
Bulk density (g/cm <sup>3</sup> )	1.03 a	1.23 b	1.30 b	1.11 a	1.27 b	1.28 b	1.21 b	1.22a	1.30 a	1.28 a	1.21 a
Water content (g kg <sup>-1</sup> )	206.7 a	169.5 a	174.9 a	356.5 a	326.8 b	309.1 b	309.9 b	269.5 a	263.9 a	196.2 b	179.3 b
pH	5.75 a	5.52 b	5.82 a	5.20 a	5.35 a	5.86 b	5.82 b	5.66 a	5.26 b	5.73 a	5.74 a

<sup>a</sup> According to Soil Taxonomy. For each soil type different letters for each parameter indicate statistically significant differences between management systems ( $p < 0.05$ ), DGC test. NA: Natural Environments, GAP: Good Agricultural Practices, NO-GAP: No-Good Agricultural Practices, CT: Conventional tillage.



**Fig. 4.** Ordination diagram from Canonical Correspondence Analysis of soil physical and chemical properties, sampling sites and soil invertebrate taxa. Soil properties are indicated with arrows. Treatments are indicated with the following symbols and colours: Good Agricultural Practices: light grey circles; No-Good Agricultural Practices: dark grey squares; Natural Environment: empty triangles; Conventional tillage: black diamonds. Invertebrate taxa are indicated with small empty triangles. SA, SI and SC: Sandy loam, Silty loam and Silty clay loam, respectively. SOM: soil organic matter; WC: soil water content; BD: bulk density.

#### 4. Discussion

We studied two NT systems, with one, NO-GAP, the most widely used by farmers during approximately the first 15 years of application of NT agriculture in Argentina. The GAP system is nowadays still only practiced by a minority of farmers, even though it is considered to be more sustainable by the association of NT farmers of Argentina (Albertengo et al., 2011). Accordingly, the GAP sites in this study were previously managed as NO-GAP no-till for at least 15 years, according to the detailed management information provided by the farmers. These progressive farmers practicing GAP, combined with the availability of unmanaged soils in the study area, offered us the opportunity to evaluate if the use of GAP produces a change, as well as the trajectory of that change, in litter and soil invertebrates from a known non-sustainable initial situation.

The findings of the study support our first hypothesis that GAP will increase the abundance and a change in the faunal composition of litter and soil invertebrates. In the litter layer,

four of the five taxa present, oribatid, mesostigmatid and prostigmatid mites and collembolans, increased under GAP. This is explained mainly by the greater mass and heterogeneity of the litter in the GAP system, as a consequence of the combination of crop rotation, winter cover crops and fertilization inputs. The permanent vegetation and litter cover provides those faunal groups with a more regulated and stable habitat for shelter or feeding. That the presence of a permanent soil cover moderates extremes soils temperatures and reduces water content loss rates from the soil surface (Edwards and Lofty, 1975; Fox et al., 1999). Plant residue cover provides a readily available food source for invertebrates (Donegan et al., 2001; Coleman et al., 2002; El Titi, 2003). Oribatid mites and collembolans that both feed on fungi and dead organic matter (Behan-Pelletier, 2003; Coleman et al., 2004) found more suitable conditions under GAP. Previous studies (Bardgett et al., 1993) indicated that residue presence promotes proliferation of soil Collembola due to favourable microhabitat conditions.

The higher litter cover under GAP may also provide predators (Mesostigmata and most Prostigmata) with more abundant and diverse prey than the almost bare NO-GAP soil surface.

Higher predator abundance can be explained by the establishment of species that were able to exploit the prey-rich food resources available in the litter layer of GAP. For example, the high density of mesostigmatid mites may be related to the high abundance of Collembolans, which are one of their preferred food items (Hopkin, 1997; Koehler, 1999).

Our results from soil sampling show an increase in the abundances of ants and prostigmatid mites as well as an increase of earthworm and collembolan numbers produced by the use of GAP. These results suggest that the improvements of surface soil conditions mentioned earlier for GAP system also influenced the mineral soil layer. Prostigmatid mites are predators that feed mainly on nematodes (Kethley, 1990), small euedaphic organisms that may be benefited from the microclimate of the first soil centimetres produced by soil cover in GAP, and therefore facilitate the increase in the abundance of Prostigmata.

Earthworm numbers in the three managed systems were significantly lower than in the undisturbed natural reference site. There were few differences between both NT systems and the ploughed system. This result is not in agreement with much of the literature that suggest that as residues build up on the soil surface, so do earthworms numbers (Chan, 2001; Errouissi et al., 2011). Earthworms tended to be more abundant in GAP than in NO-GAP, but the differences were not significant. We propose two possible explanations for this different result: (1) The time of response of earthworms to GAP is longer than the period of GAP application in the study sites, considering that the initial condition was unfavourable for earthworms; (2) the subtle differences we found between GAP and NO-GAP require an analysis of earthworm by species. For example, edoigeic species may be less sensitive to changes in soil cover -which is one of the main differences between GAP and NO-GAP- than anecic or epigeic species.

Most of the literature on the effects of agricultural managements on soil fauna compare two or more contrasting management systems, such as conventional tillage vs NT (e.g., Tabaglio et al., 2009; Errouissi et al., 2011), conventional tillage vs minimum tillage (e.g., Kennedy et al., 2013), or organic vs conventional agriculture (e.g., Suthar, 2009; Domínguez et al., 2014). In this study, two variants of NT management were compared, were the magnitude of the practice differences were small. While we detected clear changes in litter fauna abundance the picture on the effects of GAP was less clear in soil, partly because one of the more important practices from GAP -crop rotation- had a stronger effect on the litter layer than in mineral soil. To better assess the effects of GAP on soil invertebrates, we conducted a detailed analysis of soil invertebrate data from GAP and NO-GAP treatments, considering the data from all the taxa sampled. In that analysis we found a clear change in faunal composition produced by the introduction of GAP. Results showed that GAP changed invertebrate faunal composition in all three soil types. For each soil type, the trajectory of change of faunal composition from the initial in NO-GAP situation to the present state under GAP system was also monitored. This finding agrees with the proposed hypothesis on the effects of GAP on soil invertebrate composition, that is soil invertebrate composition is sensitive to the relatively subtle changes in agricultural management systems, including GAP and NO-GAP.

We also hypothesized that the effects of GAP on soil invertebrates will be more significant in coarse textured soils, as different soil types may be more or less resilient to the agricultural practices (Kibblewhite et al., 2008) and may have different effects on soil fauna (Kováč, 1994). The three soils we studied are all Mollisols, but differ in texture, from sandy loam to silty clay loam and in SOM concentration, from ~2 to ~5 % (Table 3) and in

physical structure, with greater aggregate and matrix structure in the silty clay loam than in the sandy loam soil.

In the soil layer, abundances of earthworms, collembolans, ants and mesostigmatid mites differed between soils, with the first two taxa having lower abundance and the last two taxa higher abundances in the sandy loam. These results can be explained in part by de fact that earthworms and collembolans are trophically dependent on soil organic matter concentration (Lavelle, 1997; Coleman et al., 2004; Brennan et al., 2006) while ants and mesostigmatid mites are not (Koehler, 1999; Lobry de Bruyn, 1999).

The effect of soil type on invertebrate faunal composition was apparent for both GAP and NO-GAP treatments, with the composition from the sandy loam soil separated from the samples from the other two soil types. Samples from NO-GAP have different faunal composition according to soil type. Samples from GAP in the silty and silty clay loam appear to be more similar in terms of composition. Only in one case was a partial overlap observed between GAP and NO-GAP samples from different soil types noted, highlighting the need to consider soil type in the analysis of GAPs on soil fauna.

The soil properties with stronger influence on fauna and on site differentiation were SOM, water content and bulk density (BD). Regarding the ordination of sites according to soil properties, the sites from GAP were separated from sites from NO-GAP, which were characterized, as a whole, by higher soil compaction (high BD) and lower SOM and soil water content. Even when differences in these soil properties between the two systems were not consistent when analysed separately, by taking them together in the CCA it was possible to separate both NT variants.

Regarding the effect of soil properties on soil invertebrate fauna, SOM was positively linked to Astigmata, Isoptera and Diplopoda; the occurrence of most of the taxa present in agricultural sites can also be considered to be linked to a reduction of BD. Soil compaction has a negative effect on earthworms (Edwards and Bohlen, 1996; Chan, 2001; Domínguez et al., 2010), collembolans (Bedano et al., 2006a) and mites (Bedano et al., 2006b). However, no other soil properties emerged as good explanatory variables of differences in faunal composition between GAP and NO-GAP. Therefore, taking into account the relatively recent and subtle differences between GAP and NO-GAP previously mentioned, we propose considering more specific and therefore more responsive soil properties. Among physical properties, the type and size of pores, obtained from micromorphological studies, has been used to explore differences of fauna between the NT variants (Shiso Toma et al., 2013a,b). Fractions of organic matter can be more sensitive to changes in soil management than total SOM concentration (Galantini et al., 2008). For example Duval et al. (2013) found that while GAP and NO-GAP had no effect on total SOM, particulate fractions did show differences. For example, in the sandy loam the labile organic matter fractions were in greater proportion under GAP (Duval et al., 2013). This increase was undetectable by the total SOM analysis. Some of the differences in soil fauna composition observed between GAP and NO-GAP in this study might reflect the build up on the labile pool. Bacterial groups like Acidobacteria have been tend to be more abundant in GAP (Figuerola et al., 2012) and bacterial  $\beta$ -diversity was also higher in GAP (Figuerola et al., 2014). Because of the close trophic relationships between invertebrates and microorganisms (Lavelle et al., 2006; Coleman et al., 2004), these differences could also explain the differences in invertebrate faunal composition.

Finally, the effect of agrochemicals on invertebrates deserves attention. One of the practices included in the GAP system is a reduction in agrochemical use, and this can also have had an influence on the differences observed in litter and soil invertebrates between GAP and NO-GAP, mainly in the higher abundances of litter fauna in GAP.



The benefits of maintaining abundant populations of litter and soil invertebrates in agricultural systems are well known (El Titi, 2003; Brussaard et al., 2007), especially under NT systems (Kladivko, 2001; Arolo et al., 2010; Domínguez et al., 2010). Therefore, as a consequence of the linkages between invertebrates and soil processes, the increase in abundance of most invertebrate taxa in GAP is expected to favour soil functioning in comparison with the non-application of GAP. However, the effects of GAP application on key soil processes like litter decomposition and soil structure maintenance, needs further research.

## 5. Conclusions

Application of GAPs in productive NT fields increases litter and soil invertebrate abundance and modifies faunal composition. The plant residue cover generated by the GAP system contributed to increase the numbers of litter invertebrates by generating a food-rich and microclimatic suitable environment. In the soil layer, most invertebrate taxa and, to a lesser extent, earthworms were more abundant in GAP than in NO-GAP.

The functional implications of the observed changes in litter and soil invertebrates were not directly measured, but potentially translate to changes in soil function.

We demonstrated that the effect of GAP differs with soil type, meaning that differences are needed to be analysed considering the pedological context at the landscape scale.

Although GAP represents an improvement in litter and soil fauna when compared with NO-GAP, both NT systems are significantly different from the natural soils of the region, both in terms of faunal abundances and composition and of soil physical and chemical properties. GAP as a soil management strategy, may be improved by increasing and diversifying crop rotation intensity; these changes could be measured using biological parameters such as soil fauna.

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