



Soil macrofauna diversity as a key element for building sustainable agriculture in Argentine Pampas



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ABSTRACT

The agricultural activity in the Argentine Pampas, characterized by an important trend towards no-till soybean monocropping, has completely transformed the original Pampas landscape into a monotonous scenario with a continuous succession of farms of very low crop diversity. This process has led to soil physical, chemical and biological degradation in those systems. The increase of crop rotation rates in no-till and reduced tillage systems has been proposed as an alternative with reduced negative impact on soils in the context of conventional agriculture. On the other hand, extensive organic farming is also suggested as an alternative to high-input agriculture systems. In this article, we aim to explore how different variations of farming practices and systems impact soil macrofauna, along an edaphoclimatic gradient in the Pampas region. We studied the following systems: natural grassland (Gr) as indicator of the original community, extensive organic farming (Org), conventional agriculture with no-tillage and three crop rotation levels (Nt-R1, Nt-R2 and Nt-R3), and reduced tillage with two levels of crop rotation (Til and Til-R). We assessed soil macrofauna, with emphasis on earthworm, beetle and ant communities; and soil physical and chemical properties. Macrofaunal taxa composition was significantly affected by both management systems and edaphoclimatic conditions. The Gr community had pronounced differences from all the agricultural systems. The earthworm community from Gr had distinctive features from those of most agricultural systems, with Org and Nt-R3 being the most similar to Gr in native and exotic earthworm species, respectively. The beetle community in Org was the most different one, and the communities from the other systems did not show a pattern related to management. Ant community composition was not determined by management systems, but it was affected by edaphoclimatic conditions. All the studied macrofauna groups had a significant co-variation with soil physical and chemical properties, showing that both the characteristics of each soil relative to the geographic location and the effect of management on abiotic soil attributes have an important effect on soil macrofauna. This study confirms that biodiversity is being lost in Pampas soils, which implies a possible threat to the soil capacity to perform the processes that sustain soil functioning and hence plant productivity. Further considerations about the sustainability of the current agricultural model applied in the Argentine Pampas are needed.

1. Introduction

Soils are non-renewable resources, meaning that their loss and degradation are not recoverable within a human lifespan. However, soils of all around the world are being exploited, mostly neglecting this essential feature. Important soil threats have been described but the extent, severity, and consequences of soil degradation remain poorly documented (Brevik et al., 2015).

The main region devoted to agricultural land use in Argentina is the Pampas region; however, in the last years, agricultural boundaries have

been moving to other regions where soils are less developed and more susceptible to degradation. Cereal and oilseed production covered 37.4 Mha. in the 2015/2016 crop season, with 68.2% of that area being cropped with soybean (*Glycine max*) and 23% with maize (*Zea mays*), meaning that 91.2% of the land sown with cereal and oilseeds was cultivated with only two crops. Most crop production in Argentina follows a production model initiated after the “green revolution”, in the 70s. That model was then reinforced with the incorporation of transgenic crops, most of them with resistance to herbicides, and with the widespread use of a synthetic package of fertilizers, herbicides,

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insecticides and fungicides in the region. Therefore, agricultural activity has completely reshaped the Pampas landscape, generating a monotonous scenario with a continuous succession of farms cropped using similar conventional practices and with very low crop diversity, typically with absence of timberline, partly because many tree species are susceptible to herbicides used in annual cropping.

Previous research in the area has warned about the loss of biodiversity, especially of soil biodiversity, associated with that agricultural model (Bedano et al., 2016; Bedano and Domínguez, 2016; Domínguez et al., 2010, 2014; Domínguez and Bedano, 2016). There is growing concern about this situation, given that soil biodiversity is thought as one of the resources that require the greatest attention, since the soil capacity to sustain crops ultimately relies on soil biology. Actually, soil biology sustains or regulates many of the soil functions that are needed to keep resilient soils, those able to sustain ecosystem services in time and withstand to perturbation whether anthropogenic or not (Tittone, 2016).

Soil biota includes an enormous diversity of organisms and addressing them as a whole would pose an arduous challenge. However, some groups of organisms, such as soil macrofauna, can be assessed as indirect indicators of the whole soil community as well as direct indicators of soil functioning. Soil macrofauna includes invertebrates with body diameter greater than 2 mm, inhabiting surface litter or digging galleries in the soil (Lavelle and Spain, 2003). Macrofauna comprises organisms belonging to two functional groups: ecosystem engineers and litter transformers. The former directly or indirectly modulate the availability of resources to other species by causing physical state changes in biotic and abiotic materials, and in so doing, they modify, maintain, and create habitats (Jones et al., 1994). Earthworms, termites, ants and some beetle larvae are the most important examples (Lavelle et al., 2006, 2007; 2016; Stork and Eggleton, 1992). In the Pampas region, earthworms are by far the most important ecosystem engineers, strongly linked to processes like soil structure formation and nutrient cycling. Numerous litter transformers, like isopods, millipedes, many beetles, larval insects, and some earthworm and enchytraeid species, are important in litter decomposition through comminution of organic residues, facilitating and enhancing decomposing process mediated by bacteria and fungi (Lavelle and Spain, 2003). Furthermore, a diverse community of predators dwell in litter, acting as regulators of soil invertebrate populations and ecosystem processes (Moya-Laraño, 2011).

While there is a wide consensus about the role of soil fauna in soil functioning and therefore in achieving sustainable agriculture production, in Argentina these organisms are rarely considered by the most important actors in deciding which agricultural models and practices are used: international companies involved in the agricultural businesses, the governmental agricultural agencies and farmers. Aware of this situation, farmers have proposed different approaches based on different agricultural paradigms that intend to promote soil biodiversity conservation in the Pampas region.

One of the approaches is organic agriculture, which is based on ecological and biological processes and involves soil biodiversity conservation as an inherent goal (IFOAM, 2012). Organic agriculture is not merely limited to farming without using chemical inputs (Jiménez, 2007). Rather, it implies understanding the farm as an organism, in which all the components, living or not, interact to create a coherent, self-regulating and stable whole; organic farming implies a degree of awareness of the functioning of, and inter-relationships (between animals, plants, and the environment) within the farm system (Jiménez, 2007). However, extensive organic farming in Argentina often lacks this holistic approach and practices related to improve agroecosystem biodiversity are not applied evenly. Mixed farming with alternation of crop and livestock is generally adopted; however, cover crops, green manure, intercropping, agroforestry, and management practices in the

environment surrounding the agricultural plots, such as the use of windbreaks, shelterbelts, and living fences, are very scarcely used in extensive organic farms in the Pampas region. A wide variety in the tillage system applied for weed controlling is also observed. Therefore, in many cases, the main measure in favour of biodiversity conservation is the non-use of synthetic agrochemicals and the inclusion of pasture in the crop rotation, usually every 3–4 years.

There is still controversy in the scientific community regarding the benefits of organic farming to soil organisms. Several studies show that organic farming favours them compared to conventional systems (e.g. Bengtsson et al., 2005; Hole et al., 2005; Mäder et al., 2002) with earthworms seeming to be the group most consistently benefited (Bettiol et al., 2002; Crittenden and de Goede, 2016; Domínguez et al., 2014; Domínguez and Bedano, 2016; Siegrist et al., 1998; Suthar, 2009). However, Flohre et al. (2011) found that the effects of organic farming on soil biota are greatly influenced by the landscape context. In a recent meta-analysis, Tuck et al. (2014) observed a lack of positive effects on decomposers, which are mostly soil fauna, although they remarked that organic farming effects on soil organisms are ambiguous and in general understudied. Some studies have also found neutral or even negative effects. Specific practices, such as the use of manure, green manure, fertilization, different tillage intensities, and different pesticides, are very variable and hinder identification of the specific aspects of organic farming that produce positive effects. Therefore, research articles usually find different results because they assessed systems that vary in specific practices. Nonetheless, the bias towards the study of organic farming systems adopted in Europe and, to some extent, in USA, has enormous proportions. Latin American countries, especially Argentina, lack deep research in organic agriculture and its effect on soil biology.

On the other hand, following the general principles of conventional agriculture, which involves a wide use of machinery, transgenic crops and synthetic agrochemicals, several schemes with different levels of crop rotation and tillage intensities are being used by Argentinean farmers. Reducing tillage intensity and enhancing crop diversity, with higher crop rotation or with the use of cover crops, have been recognized as practices with a strong positive effect on soil biology (e.g. Blanchart et al., 2006; Brevault et al., 2007; de Aquino et al., 2008; House and Parmelee, 1985; Lavelle et al., 2001). Moreover, those practices have been linked to a general improvement of soil physical and chemical properties, such as organic matter content, aggregation, and nitrogen content (Caviglia and Andrade, 2010; Lal et al., 2007). The improvement of those soil habitat characteristics has a great importance in soil biology as well. Thus, the positive effect of increasing crop diversity and reducing tillage intensity has been proven to have several beneficial effects on ecosystem processes; however, studies addressing this issue in an applied agronomic context are very scarce (Bender et al., 2016). Thus, it is interesting to assess if soil biota conservation is improved when crop rotation intensity is increased and if that improvement is then translated into higher yields.

Therefore, we aimed to study how different variations in farming practices and systems, belonging to different agricultural paradigms, impact on soil macrofauna along an edaphoclimatic gradient in the Pampas region. Since agricultural practices which preserve soil biodiversity while maintaining crop production are intended, we were not interested in comparing different land uses, i.e. forests or pastures versus monocultures, but in comparing changes in specific management practices in the agricultural land use. We are aware that those practices present subtle differences in crop rotations or in tillage intensity and therefore we do not expect to find the kind of major differences in soil macrofauna expected, for example, when comparing different and contrasting land uses. However, considering the sensitivity of many of the macrofauna taxa to changes in soil, litter and microenvironmental conditions, produced by changes in land management, we expected

macrofauna to consistently differ among treatments. Natural grasslands were also studied as a reference system containing the ideally expected macrofauna community.

Thus, we investigated, first, the effect of different management systems on macrofauna composition and earthworm, beetle and ant communities along an edaphoclimatic gradient; and second, the covariation between soil macrofauna and physicochemical properties in those different management systems and edaphoclimatic locations. We aim to answer the following questions: does the taxonomic composition of soil macrofauna change with different management strategies? Are those changes independent of the edaphoclimatic conditions? Does extensive organic farming or the inclusion of crop rotation in conventional farming promote a macrofauna composition similar to that of grasslands? Are changes in macrofauna composition linked to changes in soil physical and chemical properties?

Our results will allow to analyse how different agricultural systems change soil macrofauna composition but also how key organisms for soil ecosystem functioning are affected. This analysis may contribute to the knowledge about the provision of ecosystem services and therefore to the performance of the analysed agricultural systems within the ecological domain of sustainable development.

2. Materials and methods

2.1. Study region

The Argentine Pampas region is a wide plain covering more than 52 Mha of lands suitable for cropping and cattle rearing; the high fertility and productivity characteristic of the area provide significant comparative advantages for agricultural production. 25 sampling sites (Fig. 1) were selected from a wide geographical range along an edaphoclimatic gradient; in order to understand if the effect of management strategies on the macrofauna is mainly due to agricultural management, regardless of soil and climate variations.

2.2. Farming systems

The selected sites are briefly described in Table 1. The agricultural sites were at least 100 ha in area and were managed using similar

agricultural practices for about 10 years before sampling. Conventional agriculture sites were selected to represent the main farming systems used in the Pampas region. Among them, systems with no-till were classified according to crop rotation intensity into three levels: treatments Nt-R1, Nt-R2 and Nt-R3. For that, an index between the number of crops and the number of years was calculated. In Nt-R1 the index ranged from 1 to 1.14; in Nt-R2 from 1.28 to 1.43; and in Nt-R3 from 1.57 to 1.86. Systems under tillage were classified into those with a strong trend to soybean monocropping, named Til, and those with a higher level of crop rotation, named Til-R. In general terms, although with some variation among sites, fertilization consisted of a mean of 70 kg/ha/crop season of urea for maize crop; soybean crop was not fertilized. Glyphosate was the most widely used herbicide, at rates of about 4–8 l/ha/crop season, depending on the crop and the farm; 2,4-D and atrazine were also occasionally used. Chlorpyrifos, endosulfan and lambda-cyhalothrin were the most widely used insecticides. Two extensive organic agricultural sites under tillage were also sampled (Org); no fertilizer, herbicides, insecticides or fungicides were used in these sites. Grasslands (Gr) located near the agricultural sites were sampled to characterize the reference community of soil macrofauna. All of them have been undisturbed and covered with natural grass species during the last 30 years. Plant cover was 100% and the litter layer was approximately 1 cm thick. These sites were not managed and were occasionally subjected to extensive cattle grazing, trampling, or grass cutting. The 25 sites selected to represent the mentioned systems were located in 5 different localities along a west-east edaphoclimatic gradient of increasing precipitation and degree of soil development as follows: Bengolea – Cabrera-Deheza – Alejandro Roca – Monte Buey – Pergamino (see Fig. 1).

2.3. Macrofauna and soil sampling

In each of the 25 sites, five soil monoliths of 25 cm × 25 cm × 20 cm were delimited and then excavated (ISO, 2006). Soil cover percentage was visually estimated *in situ* as the percentage of soil covered by litter or crop residues within the 25 × 25 cm frame. Monoliths were gently taken to the laboratory and carefully hand-sorted to collect all the macroinvertebrates visible to the naked eye. Invertebrates were counted and preserved, and then identified into the following high-

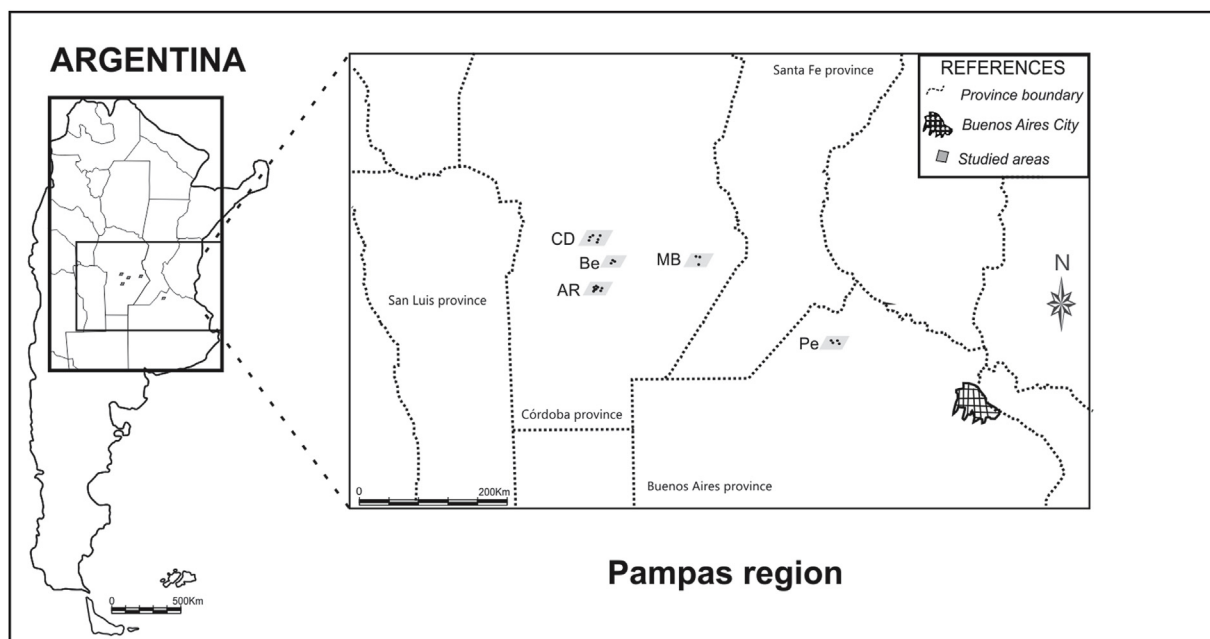


Fig. 1. Distribution of 25 sites in the study area of the Pampa region, centre Argentina. Locations: CD: Cabrera-Deheza, Be: Bengolea, AR: Alejandro Roca, MB: Monte Buey and Pe: Pergamino. From west to east there is an increasing in mean precipitation and soil development (see Table 1 for details).

Table 1
Climatic, soil description, management system, practices and crop rotation of the 25 sampling sites.

Location	AMP (mm)	AMT (°C)	MA (m)	Soil classification* Soil texture Soil use classes	Management system	Site	Management practices and crop rotation
Alejandro Roca	840	17	200	Typic Haplustoll Sandy loam III	Grassland	A1-Gr	–
						A2-Gr	–
						A1-Til	Conventional tillage with low rotation
						A2-Til	Conventional farming with agrochemical use, mechanical tillage with disk harrow and roller, crops in the last five years: Sy/Sy/Sy/Sy/Sy.
						A1-Nt-R1	No-till with low rotation
						A2-Nt-R1	Conventional farming with agrochemical use, mechanical tillage with disk harrow, roller and plough, crops in the last five years: Sy/Sy/Sy/Sy/Mz.
Cabrera-Deheza	695	17	291	Typic Haplustoll Coarse-loamy III	Grassland	A1-Org	Organic farming, mechanical tillage with sub-tiller, disk harrow, roller and weeder, crops in the last five years: Oa-Sf/So/Oa-Sy/Mz/Oa-Sy.
						A2-Org	Organic farming, mechanical tillage with sub-tiller, disk harrow, roller and weeder, crops in the last five years: Mz/Sf/Sy/Sf/Sy.
						C1-Gr	–
						C2-Gr	–
						C1-Nt-R1	Conventional farming with agrochemical use, crops in the last five years: Wh-Sy/Mz/Wh-Sy/Sy/Sy.
						C1-Nt-R2	Conventional farming with agrochemical use, crops in the last five years: Mz/Sy/Sy/Mz/Pe-Ry.
Bengolea	852	17	223	Entic Haplustoll Sandy loam IV	Grassland	C1-Til-R	Conventional farming with agrochemical use, mechanical tillage with disk harrow and roller, crops in the last five years: Wh-Sy/Pe/Sy/Mz/Sy.
						C2-Til-R	Conventional farming with agrochemical use, mechanical tillage with disk harrow and roller, crops in the last five years: Mz/Sy/Mz/Sy/Mz.
						B-Gr	–
						B-Nt-R2	Conventional farming with agrochemical use, crops in the last five years: Sy/Wh-Sy/Sy/Sy/Sy.
						B-Nt-R3	Conventional farming with agrochemical use, crops in the last five years: Wh-Sy/Ve-Mz/Wh-Sy/Sy/Mz.
						M-Gr	–
Monte Buey	910	17	110	Typic Argudoll Silt loam II	Grassland	M-Nt-R2	Conventional farming with agrochemical use, crops in the last five years: Mz/Sy/Sy/Sy/Wh-Sy.
						M-Nt-R3	Conventional farming with agrochemical use, crops in the last five years: Wh-Sy/Ve-Mz/Mz/Wh-Sy/Ve-Mz.
						M-Til	Conventional farming with agrochemical use, mechanical tillage with plough and disk harrow, crops in the last five years: Sy/Sy/Sy/Sy/Sy.
						P-Gr	–
						P-Nt-R1	Conventional farming with agrochemical use, crops in the last five years: Sy/Sy/Sy/Sy/Sy.
						P-Nt-R2	Conventional farming with agrochemical use, crops in the last five years: Mz/Sy/Wh-Sy/Mz/Sy.
Pergamino	1000	17	66	Typic Argudoll Silt loam I	Grassland	P-Til	Conventional farming with agrochemical use, mechanical tillage with disk harrow and roller, crops in the last five years: Sy/Sy/Sy/Sy.
						P-Gr	–
						P-Nt-R1	Conventional farming with agrochemical use, crops in the last five years: Sy/Sy/Sy/Sy/Sy.
						P-Nt-R2	Conventional farming with agrochemical use, crops in the last five years: Mz/Sy/Wh-Sy/Mz/Sy.
						P-Til	Conventional farming with agrochemical use, mechanical tillage with disk harrow and roller, crops in the last five years: Sy/Sy/Sy/Sy.
						P-Til	Conventional farming with agrochemical use, mechanical tillage with plough and disk harrow, crops in the last five years: Sy/Sy/Sy/Sy/Sy.

Mz: maize; Oa: Oat; Pe: peanut; Ry: rye; Sf: sunflower; So: sorghum; Sy: soybean; Ve: vetch; Wh: wheat. AMP: annual mean precipitation, AMT: annual mean temperature, MA: mean altitude above sea level. * According to Soil Survey Staff (2010).

range taxa: Araneae, Chilopoda, Coleoptera, Diplopoda, Enchytraeidae, Formicidae, Isoptera, Insecta larvae and Lumbricina, hereafter referred to as “macrofaunal taxa”. Clitellate earthworms were identified to species level using James et al. (2015), Mischis (1991), and Righi (1971, 1979) taxonomic keys. Ants were identified to genus level (Bolton, 1994; Palacio and Fernandez, 2003) and beetles to family level (Lawrence et al., 2002), and morphospecies were defined in both groups. Hereafter, ant and beetle morphospecies will be referred to as species. Soil samples from each monolith were preserved to determine soil organic matter content (Walkley-Black method, Jackson, 1976) and pH (potentiometric method, soil–water ratio 1:2.5). In the field, next to each macrofauna monolith, an undisturbed soil core was extracted to further determine bulk density and soil moisture. In the laboratory, immediately after sampling, soil cores were weighed first to obtain moist weight and then oven-dried to a constant weight at 105 °C. Soil moisture percentage (gravimetric method) and soil bulk density (cylinder method) were then calculated.

2.4. Statistical analysis

As the biological database contained many zeros, a Euclidean-based transformation that allows the use of ordination methods was used (Legendre and Gallagher, 2001). For that, abundance data for all invertebrate groups were transformed with the Hellinger transformation, according to Legendre and Gallagher (2001) and Legendre and Legendre (2012) by using the *decostand* function from vegan R package (Oksanen et al., 2017). Since data of soil properties were not dimensionally homogenous, they were standardized using *decostand* function (Legendre and Legendre, 2012).

Two multivariate approaches were used: a linear discriminant analysis (LDA) and co-inertia analyses (CoIA). LDA was used to answer the questions about changes in macrofauna community composition under different agricultural managements and edaphoclimatic conditions. The function *discrimin* from the *ade4* package (Dray and Dufour, 2007) in R (R Core Team, 2017) was used. Monte-Carlo permutation test was used to assess the significance of the analyses via the *rtest* function from *ade4* package in R (10,000 permutations). CoIA was used for macrofaunal taxa, earthworms, ants and beetles, to analyse if changes in macrofauna composition were linked to changes in soil physical and chemical (SPC) properties, and to explore whether their covariation is related to the different managements systems. CoIA is a global measure of the co-structure of sites in the environmental and species hyperspaces (Dolédec and Chessel, 1994; Dray et al., 2003). It maximizes the co-inertia between the variables of two tables. Unlike the commonly used canonical correspondence analysis (CCA), which uses a correlation matrix, co-inertia uses a covariance matrix and is especially appropriate when the number of species is higher than the number of sampled sites (Dolédec and Chessel, 1994). CoIA avoids the multicollinearity problem associated with CCA and is simple and robust for matching two tables (Dolédec and Chessel, 1994). When variables are correlated, CCA becomes unstable and CoIA is appropriate (Dray et al., 2003). PCA on both transformed fauna and environmental data tables were performed. Then, the *coinertia* function in the *ade4* package in R was applied. Randomization procedures are available to test the association between two tables like the R_v coefficient. Heo and Gabriel (1998) developed a test for the significance of R_v coefficient with Monte-Carlo randomization procedure which was performed using the *randtest* function from *ade4* (10,000 permutations). The null hypothesis of the test was that the two data sets were no more related than random data sets would be (Legendre and Legendre, 2012). A summary of soil physical and chemical and macrofauna data used for the analyses is presented in Tables 2 and 3.

3. Results

3.1. Macrofaunal taxa

Total macrofauna abundance (Table 2) had the highest value in Gr, and it was reduced in more than three times in the agricultural system with the greatest maximum macrofauna abundance (NtR3). To assess how the whole macrofaunal community change under different management systems and whether those changes are independent of edaphoclimatic conditions, we conducted an LDA for management system and geographical location. Sites were ordinated by the management systems, according to the composition of macrofaunal taxa (Fig. 2a, $p=0.02$). As expected, the grassland was the most different system. None of the agricultural management systems had a macrofaunal taxa composition similar to that of grasslands. However, macrofauna composition, even at this low taxonomic resolution, was affected by contrasting management systems. Each one of the three different no-till systems with different rotation intensities had a macrofauna composition with distinctive features. However, Nt-R3 was, to some extent, overlapped with Til-R and Nt-R1 with Til. Nt-R2 was strongly associated with spiders, and Nt-R3 and Til-R with ants. The macrofauna composition of organic farming system had the lowest within-variance of the studied systems and was overlapped with the Til system.

We also tested the effect of the geographical location on macrofauna composition via the LDA (Fig. 2b). The results showed that the changes in macrofauna composition due to the different systems were also deeply related to edaphoclimatic conditions. Sites belonging to the sampled locations were plotted almost following the edaphoclimatic gradient, from less developed soils and lower precipitations in Cabrera-Deheza through soils of intermediate development and precipitations in Alejandro Roca and Bengolea to highly developed soils and high precipitations in Monte Buey and Pergamino. The sites from Pergamino and Monte Buey, with similar soil and climate characteristics, were grouped close to each other and shared a macrofauna composition characterized by the high abundance of earthworms, ants, millipedes and potworms. Beetles, spiders and centipedes were strongly associated with Bengolea sites. Cabrera-Deheza had the most different macrofauna composition, followed by Alejandro Roca.

Significant covariation according to CoIA ($R_v = 0.27$, $p = 0.064$, Monte Carlo test) was observed among macrofaunal taxa and soil properties (Fig. 3). The first two canonical axes explained 85% of the total co-inertia. Fig. 3c shows the 25 sites projected in the co-inertia space according to both datasets. The circle indicates a group of sites with similar characteristics for both sets of properties, in which most of the grasslands are included, together with both organic farming sites and three NT sites (B-NtR3, B-NtR2 and A-NtR1). Grasslands from Monte Buey and Pergamino were not included in this grouping of sites, mainly due to their different soil properties. Higher SOM content was especially marked in Pergamino grassland and higher moisture in Monte Buey grassland (Table 3). Macrofauna composition in both Gr are close to the other grasslands as indicated by the arrow heads pointing to the circle. All the sites from Monte Buey and Pergamino were grouped in quadrant III, except for the grassland from Pergamino.

In Fig. 3a and b, the long arrows in both faunal and environmental variables indicate that both aspects highly contributed to the distribution of the sites in the co-inertia plane. The first axis (explaining 51% of variance) associates high abundances of Lumbricina with high moisture content (quadrant III of both Fig. 3a and b), and with sites from Monte Buey and Pergamino in quadrant III of Fig. 3c. Araneae and Diplopoda are linked to high bulk density values, and Formicidae and Immature Hexapoda are related to high soil pH. The second axis (explaining 34% of total variance) links Chilopoda and Isoptera abundances with higher values of cover and organic matter, whereas Enchytraeidae and Coleoptera inversely covaried with both properties (Fig. 3a and b).

Table 2
Macrofauna groups mean abundance (ind/m²) for the 25 sampled sites.

Site	Lumbricina		Formicidae		Isoptera		Coleoptera		Araneae		Diplopoda		Chilopoda		Larvas		Enchytraeidae		Macro	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
A1-Gr	272	243.44	2851.2	2005.25	169.6	312.64	64	89.8	32	40.79	0	0	0	0	134.4	24.27	166.4	204.15	3827.2	2045.82
A2-Gr	134.4	197.46	1808	4042.81	19.2	42.93	60.8	59.22	0	0	0	0	6.4	8.76	140.8	109.34	486.4	946.95	2720	4060.71
B-Gr	140.8	144.18	4076.8	2828.75	0	0	80	72.44	12.8	13.39	25.6	57.24	12.8	20.86	278.4	177.66	278.4	356.44	4944	2872.3
C1-Gr	99.2	109.34	3126.4	2459.41	0	0	22.4	24.27	0	0	35.2	52.34	3.2	7.16	246.4	123.73	0	0	3571.2	2521.14
C2-Gr	134.4	85.12	1049.6	1339.35	0	0	16	19.6	6.4	14.31	0	0	48	29.93	284.8	272.33	12.8	28.62	1568	1507.19
M-Gr	454.4	146.9	700.8	1108.3	0	0	76.8	136.42	3.2	7.16	0	0	9.6	14.31	99.2	68.26	51.2	72.8	1561.6	1062.08
P-Gr	147.2	96.27	4572.8	6612.87	1766.4	3949.79	12.8	17.53	9.6	8.76	41.6	35.05	22.4	18.24	140.8	78.71	89.6	116.81	6905.6	10461.39
A1-NR1	0	0	124.8	226.95	0	0	25.6	26.77	6.4	8.76	0	0	28.8	42.93	147.2	34.69	537.6	253.64	908.8	372.56
A2-NR1	0	0	25.6	35.05	0	0	9.6	14.31	6.4	8.76	0	0	3.2	7.16	51.2	34.69	908.8	513.3	1004.8	484.96
C2-NR1	3.2	7.16	489.6	1068.15	0	0	0	0	3.2	7.16	0	0	0	0	86.4	97.72	6.4	8.76	624	1067.27
P-NR1	166.4	161.43	76.8	77.89	6.4	14.31	22.4	24.27	6.4	14.31	3.2	7.16	6.4	14.31	25.6	40.16	483.2	409.39	838.4	432.39
B-NR2	0	0	560	1026.25	0	0	12.8	7.16	12.8	20.86	3.2	7.16	32	22.63	73.6	52.58	476.8	442.45	1209.6	872.59
C1-NR2	60.8	44.4	3.2	7.16	0	0	0	0	6.4	8.76	3.2	7.16	25.6	21.47	0	0	0	0	102.4	64.6
M-NR2	76.8	49.83	9.6	14.31	0	0	22.4	24.27	12.8	20.86	6.4	8.76	0	0	44.8	38.2	105.6	107.69	368	128
P-NR2	240	197.59	54.4	79.68	0	0	9.6	14.31	12.8	17.53	3.2	7.16	0	0	19.2	20.86	182.4	43.23	668.8	218.62
B-NR3	6.4	8.76	1203.2	2045.42	0	0	41.6	36.83	6.4	14.31	3.2	7.16	25.6	24.27	54.4	29.07	454.4	266.68	1814.4	2058.46
M-NR3	73.6	33.18	672	1379.67	0	0	3.2	7.16	19.2	20.86	9.6	8.76	12.8	20.86	25.6	40.16	86.4	78.06	1110.4	1332.36
A1-Or	28.8	13.39	0	0	0	0	64	91.21	6.4	8.76	0	0	3.2	7.16	249.6	90.93	480	315.16	854.4	448.8
A2-Or	28.8	17.53	9.6	21.47	0	0	41.6	18.24	9.6	21.47	0	0	48	33.94	284.8	115.6	800	499.98	1334.4	623.55
A1-TI1	6.4	8.76	0	0	0	0	57.6	46.1	3.2	7.16	0	0	0	0	19.2	13.39	179.2	92.88	348.8	99.53
A2-TI1	3.2	7.16	3.2	7.16	0	0	22.4	24.27	3.2	7.16	0	0	3.2	7.16	89.6	191.53	540.8	260.31	726.4	425.82
M-TI1	179.2	152.38	9.6	14.31	0	0	22.4	21.47	3.2	7.16	0	0	6.4	8.76	60.8	44.4	521.6	498.39	1001.6	613.51
P-TI1	60.8	65.39	140.8	305.97	0	0	16	19.6	12.8	20.86	9.6	8.76	6.4	8.76	57.6	41.72	556.8	551.76	1344	947.72
C1-TI1R	9.6	14.31	44.8	70.11	0	0	3.2	7.16	9.6	8.76	3.2	7.16	28.8	20.86	44.8	20.86	28.8	20.86	179.2	98.89
C2-TI1R	0	0	499.2	1063.39	0	0	22.4	14.31	6.4	14.31	6.4	14.31	48	45.25	57.6	43.23	6.4	8.76	665.6	1078.23

SD: standard deviation.

Table 3
Mean soil physical and chemical properties for the 25 sampled sites.

Sites	Cover (%)		Organic matter (%)		Bulk density (g/cm ³)		Moisture (%)		ph	
	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
A1-Gr	100	0	2.37	0.18	1.29	0.02	17.96	0.65	6.04	0.1
A2-Gr	58.5	24.34	3.78	0.53	1.12	0.11	18.24	3.91	5.75	0.19
B-Gr	100	0	1.43	0.24	1.12	0.06	19.03	3.62	6.01	0.08
C1-Gr	42.5	29.01	3.69	0.53	1.25	0.05	19.08	1.7	6.17	0.16
C2-Gr	27.4	26.1	4.24	0.81	1.28	0.04	23.94	2.19	5.87	0.17
M-Gr	100	0	3.25	0.57	1.23	0.05	31.93	1.73	5.39	0.24
P-Gr	100	0	4.55	0.35	1.3	0.04	24.07	1.69	5.87	0.17
A1-NtR1	86	7.2	2.09	0.23	1.3	0.1	20.38	1.47	5.91	0.09
A2-NtR1	51	25.9	2.82	0.27	1.27	0.07	21.36	1.1	5.78	0.13
C2-NtR1	100	0	3.2	0.39	1.42	0.01	16.06	1.8	6.06	0.07
P-NtR1	71	18.17	3.29	0.12	1.36	0.05	20.1	1.84	5.78	0.12
B-NtR2	63	13.96	1.78	0.59	1.36	0.1	15.82	5.99	5.92	0.08
C1-NtR2	100	0	3.28	0.89	1.39	0.04	23.1	1.23	5.79	0.1
M-NtR2	30	15.81	2.3	0.4	1.37	0.04	28.22	1.42	5.86	0.07
P-NtR2	21.5	10.84	2.88	0.34	1.39	0.06	24.08	1.6	5.45	0.07
B-NtR3	100	0	2.22	0.18	1.28	0.06	15.94	0.97	5.75	0.12
M-NtR3	100	0	2.42	0.54	1.39	0.06	27.82	1.29	5.64	0.08
A1-Or	100	0	3.28	0.21	1.27	0.07	18.41	1.39	6.04	0.06
A2-Or	88	17.89	3.13	0.53	1.24	0.01	19.85	2.27	5.86	0.17
A1-Til	10	7.91	2.77	0.54	1.27	0.07	19.48	0.87	5.84	0.05
A2-Til	9	4.18	2.99	0.2	1.26	0.07	22.16	1.39	6.02	0.07
M-Til	6.4	6.11	2.89	0.48	1.27	0.05	28	2.39	5.81	0.27
P-Til	12	4.47	3.03	0.26	1.27	0.08	18.35	2.27	5.73	0.04
C1-TilR	100	0	3.83	0.26	1.38	0.07	14.94	3.54	5.85	0.1
C2-TilR	59.6	25.46	3.85	0.18	1.27	0.05	15.45	1.54	6.16	0.11

SD: standard deviation.

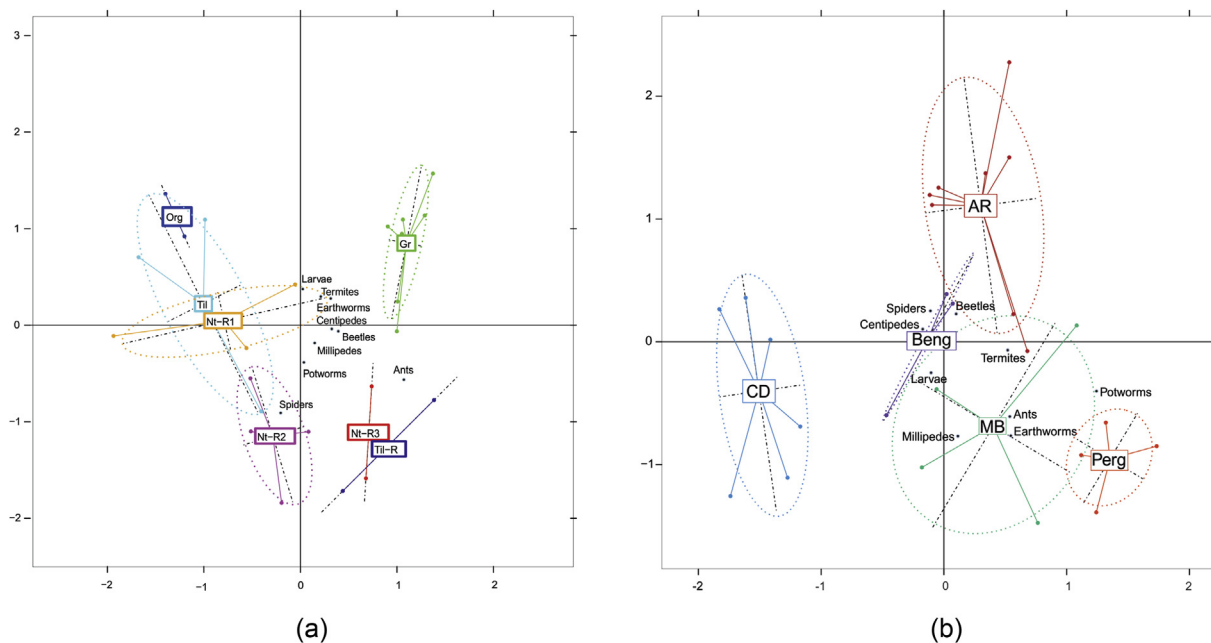


Fig. 2. Projections of the sites in the plane defined by the LDA axes based on macrofaunal taxa data and showing the grouping of the sites according to: a) management systems ($p = 0.02$), and b) geographical location ($p = 0.006$). Gr: Grassland; Org: Organic farming; Nt-R1, Nt-R2 and Nt-R3: No-tillage with low, intermediate and high rotation, respectively; Til and Til-R: Conventional tillage with low and intermediate rotation, respectively. AR: Alejandro Roca, CD: Cabrera-Deheza, Beng: Bengolea, MB: Monte Buey, Perg: Pergamino.

3.2. Earthworms

Earthworm abundance also had the highest value in Gr while the highest value in agriculture was in NtR2, reduced in almost two times regarding grasslands (Table 2). Species composition was not significantly different among agricultural systems or geographical locations, according to the LDA. However, sites from Cabrera-Deheza were

plotted far from all the other sites; then, a new LDA was performed in which all the sites from that location were removed, showing significant differences among systems (Fig. 4). Thus, in all the localities but Cabrera-Deheza, grasslands were related to high abundance of most of the earthworm species. Among agricultural systems, extensive organic farming had the most distinctive earthworm community and was the most related to grassland sites of all the agricultural systems.

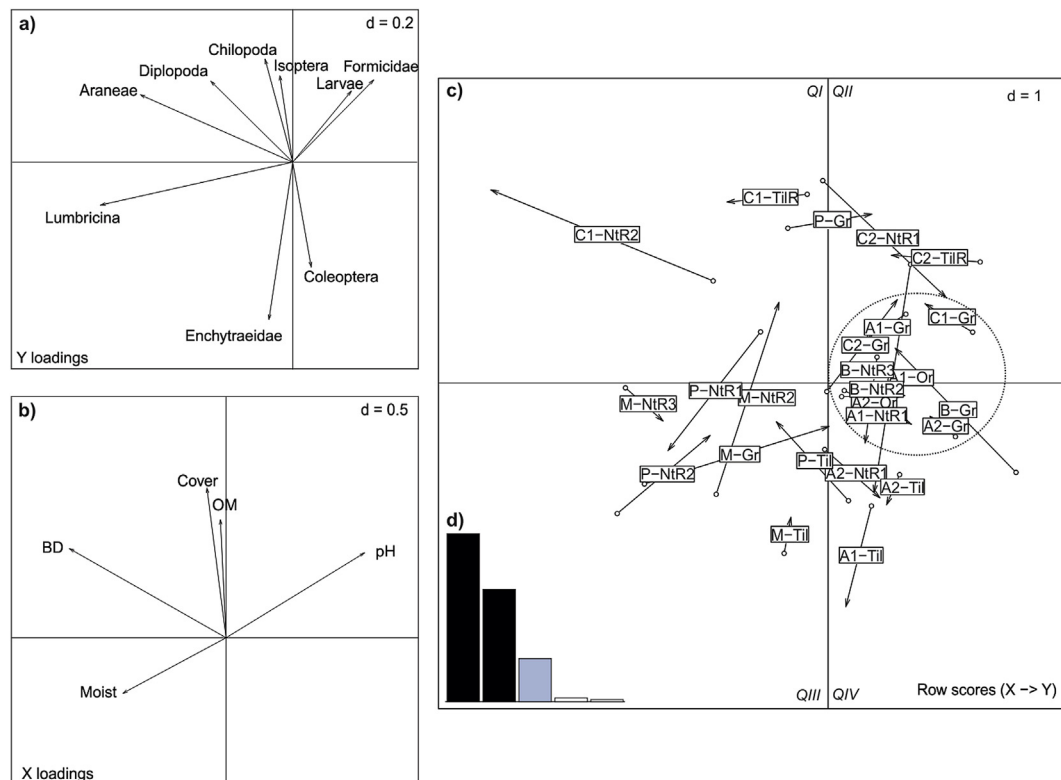


Fig. 3. Co-inertia analyses between macrofaunal taxa and soil properties (p-value = 0.064, Monte Carlo permutation test). (a) Projection of macrofauna high range taxa and (b) of soil properties onto the co-inertia plane. (c) Joint site plot depicting the ordination of sites as a function of both macrofauna composition and soil properties, represented in the plane of canonical axes 1 and 2, which account for 85% of the total co-inertia; arrow length is proportional to the difference between the ordination of the two data sets; position of the arrow tails is determined by soil properties, and the head by macrofauna composition. (d) eigenvalues diagram. Q: quadrant. Soil properties: BD: bulk density (g/cm^3), OM: soil organic matter content (%), Moist: soil moisture content (%), Cover: soil cover (%). Site abbreviations are indicated in Table 1 in the main text.

Moreover, the proximity of organic sites to grasslands was related to native species, such as *Belladrilus* sp. and *Microscoclex phosphoreus*. Regarding conventional agricultural systems, a trend to communities more similar to those of the grasslands was observed in Nt-R3, which is in agreement with results for macrofaunal taxa; the community of this site was related to exotic species of *Aporrectodea* genus. Earthworm composition in the Til system was similar to that of Nt-R3, whereas communities in Nt-R2 and Nt-R1 were very similar between them but different from the other systems.

The correlation between earthworm community and soil properties showed significant coordinated changes (Fig. 5, CoIA: $R_V = 0.38$, $p = 0.0013$, Monte Carlo test). The first two canonical axes explained 74.62% of the total co-inertia. Fig. 5b shows a little contribution of soil cover to the distribution of sites in the co-inertia plane, as depicted by the short arrow, whereas the remaining properties and all the earthworm species (Fig. 5a) highly contributed to the description of the sites, as indicated by the long arrows. Axes 1 and 2 of the CoIA analyses explain 50.3% and 24.27% of total co-inertia, respectively. Fig. 5c shows the 25 sites projected in the co-inertia space for both datasets; a pattern in the distribution of the sites according to their geographical location is observed. Sites from Monte Buey, Pergamino and Cabrera-Deheza tend to be on the left side of the plot, and those from Alejandro Roca and Bengolea tend to be on the right side. There is not a clear effect of agricultural management systems on the ordination of sites. Those sites grouped together in the first quadrant are related to *Microscoclex dubius* and *Eukerria* sp. and to high values of soil bulk density (greater than $1.36 \text{ g}/\text{cm}^3$, see also Table 3). High abundances of *Aporrectodea caliginosa* are associated with sites in quadrant III and with high values of organic matter and soil moisture. *Aporrectodea rosea* and *A. trapezoides* are almost exclusively linked to one of the natural

grasslands from A. Roca and to low values of bulk density. Finally, sites plotted in quadrant II (A1-Gr, A1-Or, A2-Or, B-Gr, C1-Gr) are mainly related to high abundances of *Belladrilus* sp., *Microscoclex phosphoreus* and *Glossodrilus parecis*, that belong to Ocnoderilidae, Acanthodrilidae and Glossoscolecidae families respectively and all of them can be considered native earthworms (Brown and Fragoso, 2007).

3.3. Beetles

Beetles were most abundant in grasslands, followed by Org management with a slighter reduction of 1.25 times (Table 2). Beetle community structure was not significantly different among agricultural systems, but it did differ among geographical locations (Fig. 6a). Community composition in Pergamino was markedly different from those in the other locations; thus, a new LDA was performed excluding Pergamino sites. In this case, beetle community composition was significantly different among systems (Fig. 6b). Grasslands were linked to a higher number of species than agricultural sites. Organic sites were situated far from the other agricultural and grassland sites, and had the most distinctive beetle community, strongly characterized by Carabidae species. The systems Til, Nt-R2 and Nt-R3 were grouped near one another, associated with Staphylinidae sp.2. Nt-R1 and Til-R were both located near the grasslands and linked to Staphylinidae sp. 3 and Polyphaga sp. 1.

The co-structure between beetle species and soil properties was also significant (Fig. 7, CoIA: $R_V = 0.36$, $p = 0.003$, Monte Carlo test). The first two canonical axes represented 81.8% of the total co-inertia. Fig. 7a shows that several beetle species made a little contribution to the distribution of the sites into the co-inertia plane, as indicated by the short arrows, with species from Staphylinidae family together with one

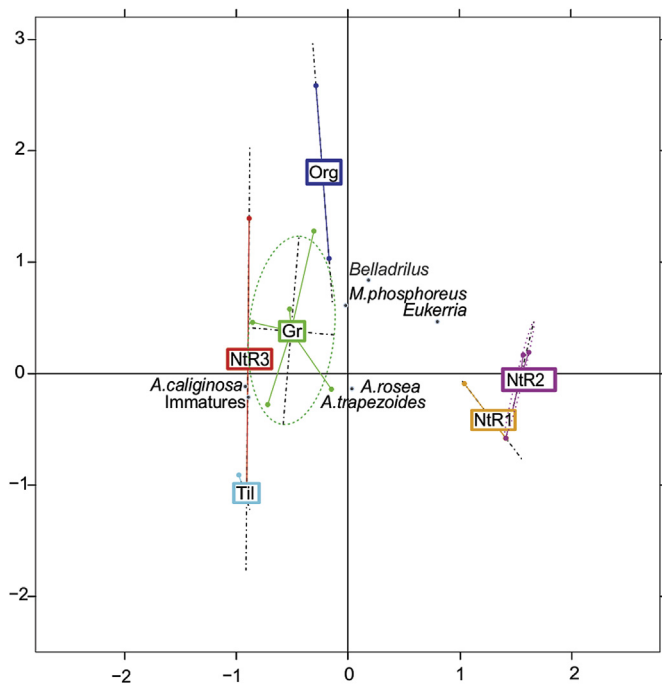


Fig. 4. Projections of the sites in the plane defined by the axes of the LDA based on earthworm species data, showing the grouping of the sites according to the agricultural systems ($p = 0.0165$), excluding the sites from Cabrera-Deheza location. Gr: Grassland; Org: Organic farming; Nt-R1, Nt-R2 and Nt-R3: No-tillage with low, intermediate and high rotation, respectively; Ttl: Conventional tillage with low rotation.

species of Carabidae and one from Cicindelidae being the most important species in defining the characteristics of the sites. Instead, all soil properties made a great contribution to the characterization of sites (Fig. 7b).

The first axis of the CoIA explained 48.8% of the total co-inertia and strongly associated soil moisture content with Scarabidae sp. 1, Staphylinidae sp. 1 and Staphylinidae sp. 5. The second axis (33% of the total co-inertia) shows a positive association between soil organic matter content and Staphylinidae sp. 4, Cicindelidae sp. 1, Polyphaga sp. 2 and Staphylinidae sp. 2 abundances, whereas an inverse covariation was observed among SOM and the abundance of Staphylinidae sp. 3 and Carabidae sp.1, which were also positively related to soil cover in axis 1.

Fig. 6c shows a strong association among sites with similar edaphoclimatic conditions, especially indicated by the beetle composition (arrow heads). Thus, all sites belonging to Cabrera, except for one grassland, are grouped in the solid line circle; all sites from Alejandro Roca and Bengolea, except for Bengolea grassland, are grouped in the dashed line circle; and all sites from Monte Buey and Pergamino, except for the grassland from Pergamino, are grouped in the dotted line circle. Grasslands from Bengolea, Cabrera and Pergamino had more different beetle communities and soil properties than those from the agricultural sites in the same location. These results indicate that in those sites, agricultural systems had a greater impact on beetle community composition than in Alejandro Roca and Monte Buey, where grasslands had similar characteristics to those of agricultural sites.

3.4. Ants

Ants presented the maximum abundance in grassland, followed by NtR3 but with a reduction of almost four times in abundance (Table 2). The LDA for ant community was significant ($p = 0.02$) for discriminating only geographical location, with ant community not differing significantly among agricultural systems. Each location had a

distinctive ant community and it was not correlated with the geographical gradient.

A significant correlation between ant species and soil properties was observed (Fig. 8, CoIA: $R_v = 0.36$, $p = 0.006$, Monte Carlo test). The first two canonical axes explained 79.75% of the total co-inertia. Most of the species made an important contribution to the distribution of sites (Fig. 8a) as well as to the soil properties, except for bulk density (Fig. 8b). The first axis explained 51% of the total inertia and related soil pH to two species: *Brachymyrmex* sp.1 and *Linepitema* sp.1. Moreover, high soil moisture content was related to high abundances of *Ponerinae* sp., *Cheliomyrmex* sp., *Acanthostichus* sp.1, and *Pheidole* sp. The second axis explained 28.7% of the total inertia and linked high abundances of *Solenopsis* sp.2 with soil cover. High SOM content was related to *Solenopsis* sp. 1, *Formicinae* sp.1 and *Myrmicinae* sp. 1, mainly related to C2-Gr (SOM 4.24%, see Table 3). As shown in Fig. 8c, most of the sites are grouped together near the origin of the plot, and in those sites the arrows point to the centre of the plot and are highly overlapped, whereas the arrow tails are more separated from one another. This indicates that most of these sites are more differentiated by the soil properties than by the ant community. The exception to this result are five grasslands (one from Alejandro Roca, Bengolea and Monte Buey and the two from Cabrera), whose arrows point from the origin to the sides of the plot; these sites have different ant communities from those of the agricultural sites of the same geographical location. Both no-till sites from Bengolea also had singular ant and soil features, and are plotted closer to their grassland than to the other farming sites.

4. Discussion

Regarding the four questions we aimed to answer, we found evidence about the relevance of management systems in driving macrofauna communities, the importance of edaphoclimatic conditions in regulating the influence of agricultural managements on macrofauna, and the strong relationship among soil physical and chemical properties and soil macrofauna community. Earthworms were the most responsive to the different management strategies. Our results shows that the systems more similar in earthworm communities to grassland were extensive organic farming and the no-tillage with the highest rotation rate.

Macrofauna composition was deeply changed in all management systems compared to the grassland (reference) systems. This result is outstanding, since grassland sites are relicts from the original landscape, mostly small in size, and conserving only partially the native flora of the region. They are also often exposed to some degree of anthropogenic impact, like occasional cattle trampling and grass cutting. However, macrofauna composition was clearly different in all the agricultural systems, despite the high dispersion observed among sites from the same system, except for organic sites. Among no-till systems, a gradient from sites with lower to higher crop rotation schemes (R1 - > R2 - > R3) was observed, with sites from Nt-R3 having a faunal composition more similar to that in the grassland, and showing that the inclusion of winter crops such as vetch (*Vicia villosa*) and wheat (*Triticum aestivum*) improved conditions for soil arthropod communities. No-till per se provides a more favourable environment for soil organisms than the other systems, by reducing moisture loss, ameliorating temperature extremes and fluctuations, and supplying a relatively continuous substrate for decomposers (House and Parmelee, 1985); however, the inclusion of pastures or legumes in the rotation schemes is also recognized as a strategy to improve biological activity (Lavelle et al., 2001). Accordingly, Blanchart et al. (2006) found that maize intercropped with the bean *Mucuna pruriens* var. *utilis* promoted higher macrofauna density and biomass than maize monocropping. Similar findings were obtained by Brevault et al. (2007) and de Aquino et al. (2008) when comparing no-till with high rotation schemes to conventional systems.

Crop rotation scheme had a greater impact on macrofauna

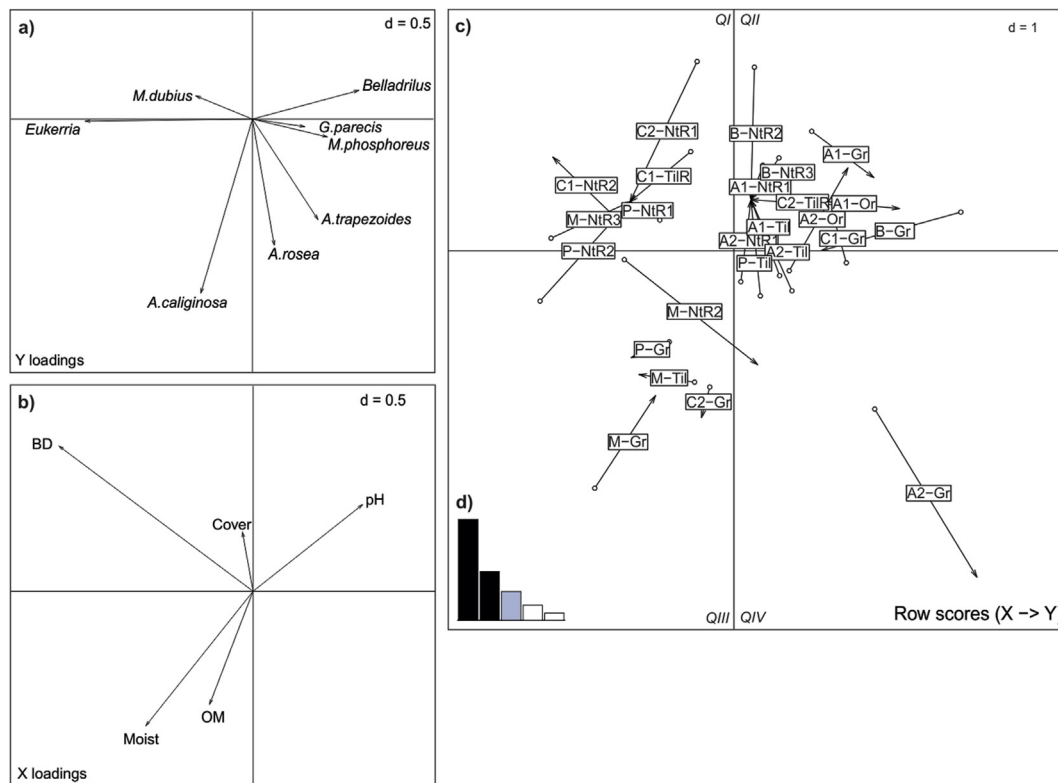


Fig. 5. Co-inertia analyses between earthworm species and soil properties (p-value = 0.0013, Monte Carlo permutation test). (a) Projection of the earthworm species and (b) of the soil properties in the co-inertia plane. (c) Joint site plot depicting the ordination of sites as a function of both earthworm community composition and soil properties, represented in the plane of canonical axes 1 and 2, which account for 74.62% of the total co-inertia; arrow length is proportional to the difference between the ordination of the two data sets; position of the arrow tails is determined by soil properties, and the head by macrofauna composition. (d) eigenvalues diagram. Q: quadrant. Earthworm species: A. caliginosa: *Aporrectodea caliginosa*; A. rosea: *Aporrectodea rosea*; A. trapezoides: *Aporrectodea trapezoide*; Belladrilus: *Belladrilus sp. 1*; Eukerria: *Eukerria sp. 1*; G. parecis: *Glossodrilus parecis*; M. dubius: *Microsclex dubius*; M. phosphoreus: *Microsclex phosphoreus*. Soil properties: BD: bulk density (g/cm³), OM: soil organic matter content (%), Moist: soil moisture content (%), Cover: soil cover (%). Site abbreviations are indicated in Table 1 in the main text.

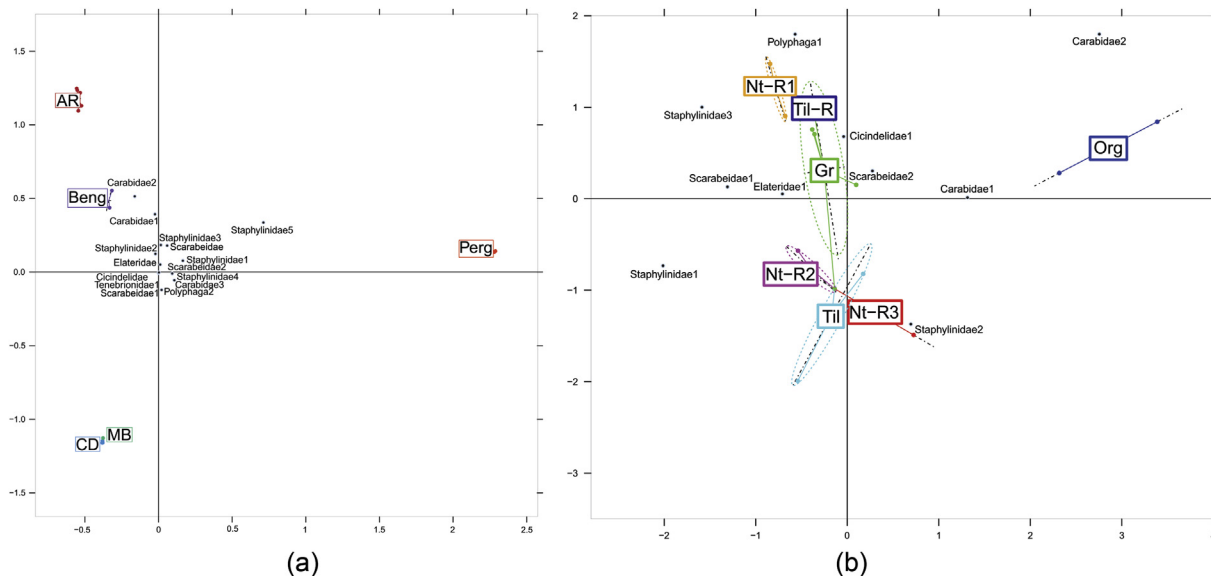


Fig. 6. Projections of the sites in the plane defined by the axes of the LDA based on beetle species abundance, showing the grouping of the sites according to: a) geographical location ($p = 0.00009$), and b) agricultural systems ($p = 0.03$), excluding sites from Pergamino location. Gr: Grassland; Org: Organic farming; Nt-R1, Nt-R2 and Nt-R3: No-tillage with low, intermediate and high rotation, respectively; Til and Til-R: Conventional tillage with low and intermediate rotation, respectively. AR: Alejandro Roca, CD: Cabrera-Deheza, Beng: Bengolea, MB: Monte Buey, Perg: Pergamino.

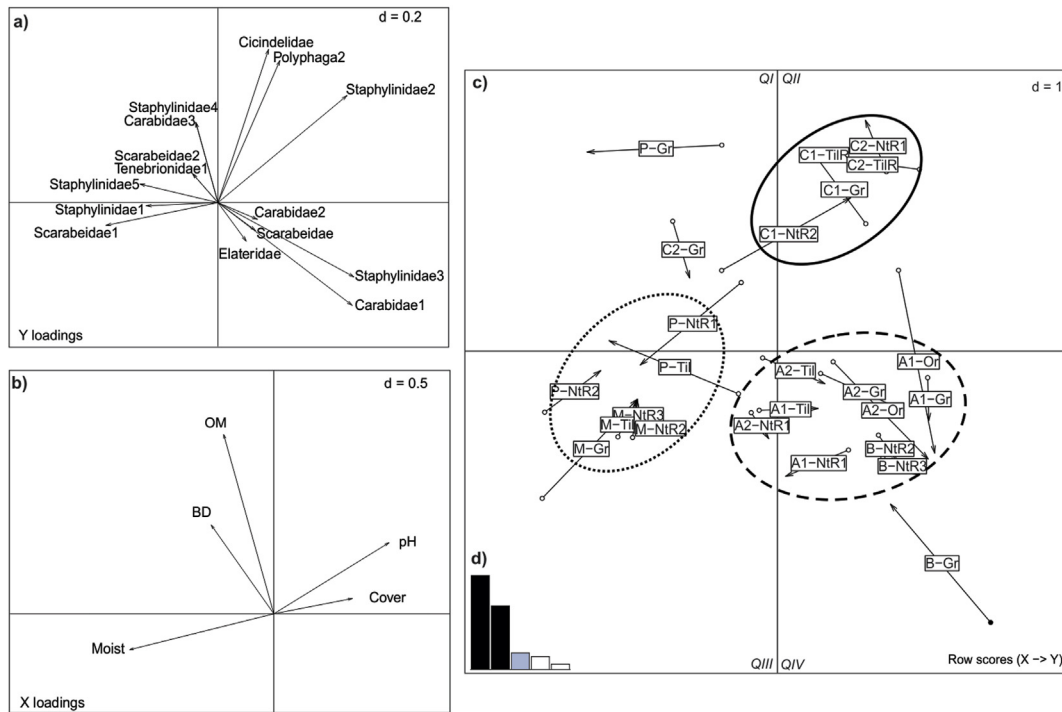


Fig. 7. Co-inertia analysis between beetle species and soil properties (p-value = 0.0032, Monte Carlo permutation test). (a) Projection of the beetle species and (b) of the soil properties onto the co-inertia plane. (c) Joint site plot depicting the ordination of sites as a function of both beetle community composition and soil properties, represented in the plane of canonical axes 1 and 2, which account for 81.8% of the total co-inertia; arrow length is proportional to the difference between the ordination of the two data sets; position of the arrow tails is determined by soil properties, and the head by beetle composition. (d) eigenvalues diagram. Q: quadrant. Soil properties: BD: bulk density (g/cm^3), OM: soil organic matter content (%), Moist: soil moisture content (%), Cover: soil cover (%). Site abbreviations are indicated in Table 1 in the main text.

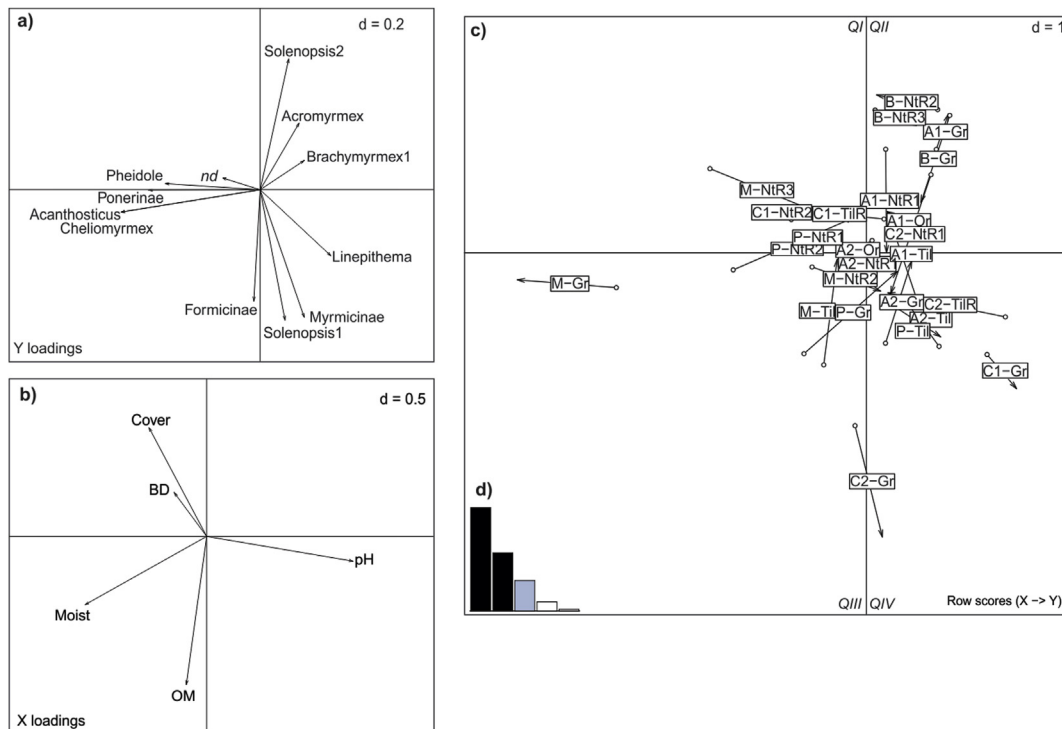


Fig. 8. Co-inertia analysis between ant species and soil properties (p-value = 0.006, Monte Carlo permutation test). (a) Projection of ant species and (b) of soil properties onto the co-inertia plane. (c) Joint site plot depicting the ordination of sites as a function of both ant community composition and soil properties, represented in the plane of canonical axes 1 and 2, which account for 79.75% of the total co-inertia; arrow length is proportional to the difference between the ordination of the two data sets; position of the arrow tails is determined by soil properties, and the head by ant composition. (d) eigenvalues diagram. Q: quadrant. Soil properties: BD: bulk density (g/cm^3), OM: soil organic matter content (%), MOIST: soil moisture content (%), Cov: soil cover (%). Site abbreviations are indicated in Table 1 in the main text.

composition than mechanical tillage. Thus, Til-R sites were similar to Nt-R3, which were both characterized by a strong dominance of ants in their macrofauna composition. Results are also probably linked to the use of shallow tillage tools together with higher crop rotation in Til-R sites. Minimum tillage tools are regarded as having a less negative impact on soil fauna than conventional tillage tools, such as mouldboard plough (e.g. Birkás et al., 2004; Bertrand et al., 2015; Chan, 2001; Kladvik, 2001; Radford et al., 1995; Wilson-Rummenie et al., 1999). However, Robertson et al. (1994) found that no-till had consistently higher abundances of soil-inhabiting macrofauna than either reduced or conventional tillage. Instead, Domínguez and Bedano (2016) found that only spider abundance was favoured in no-till compared to reduced tillage, whereas the remaining macrofauna was equally affected by no-till and reduced tillage. Thus, our results highlight the importance of crop rotation in preserving soil macrofauna composition features.

On the other hand, soil macrofauna composition in organic sites was similar to that in conventional agriculture sites with tillage and low crop rotation. Tillage tools in extensive organic systems are applied to higher depths and with a higher frequency than in Til-R. Tillage in Til conventional systems also includes mouldboard plough in Monte Buey site. This factor seemed to have filtered the macrofauna composition according to the ability of organisms to inhabit highly physically disturbed soils in both Org and Til sites, and offsets the positive effect of the non-use of agrochemicals in Org. However, as will be discussed below, this pattern was not observed in all the groups that compose soil macrofauna.

Although macrofauna community had a different composition for each management, regardless of localities, it was also different among all localities. This indicates the need to be cautious when broad conclusions from local studies are made. In our study, differences in macrofauna composition among localities seem to be linked to soil and climatic conditions, since the organization of the sites in the plane of the discriminant analysis somehow followed their geographical distribution. Macrofauna was also closely related to soil physical and chemical properties. Most grassland sites shared similar physical, chemical and macrofauna features. Instead, according to co-inertia analyses, macrofauna and soil physical and chemical features in agricultural sites altogether were more influenced by the locations of origin than by management system. This phenomenon was especially marked for sites from Monte Buey and Pergamino.

Earthworm species distribution pattern was differently shaped by the agricultural systems. Grasslands were strongly linked to most of the earthworm species, showing that they had the highest richness, in agreement with previous studies (Decaens et al., 2008; Postma-Blaauw et al., 2012; Tsiafouli et al., 2015). The earthworm community in extensive organic farming systems was also more similar to that of grasslands and strongly differentiated from those of all the sites with conventional agriculture. This result may be explained by the non-use of pesticides, which has been linked to the decrease of earthworm populations, especially given that the species that inhabit in or near the litter layer –as those favoured by NT– are the most sensitive to chemical pollution by pesticide use (Bertrand et al., 2015). Our results highlight not only that organic management favours earthworm communities, which are more similar to those of grasslands than to those of the other systems, but also that earthworm composition in organic farming systems is very different from that in conventional managements, even from those nearest the grasslands. This phenomenon can be a consequence of the non-use of pesticides filtering and favouring species more sensitive to those products, especially the native species, such as *Belladrilus* sp. and *M. phosphoreus*, which were linked to the organic system. Our findings agree with those of Henneron et al. (2014), who observed that organic system enhanced the abundance of earthworms, irrespective of their functional group.

Among conventional agricultural systems, the NT system with higher rotation, which includes wheat or vetch in winter, had an

earthworm community more similar to those of grassland sites than to the other sites. Cover crops or increased rotation improves the quality and diversity of organic residues added to the soil, and therefore improves soil as habitat for earthworms (Bertrand et al., 2015; Blanchart et al., 2006), especially for endogeic ones, such as *A. caliginosa*, which was the most abundant species in NT-R3. However, tillage systems were characterized as similar to Nt-R3 sites but without a strong association with any of the species. This result is mostly unexpected, since Til sites had rotation schemes with very low diversity. The earthworm communities in NtR1 and NtR2 were very similar between them but different from those of all the other systems, and not strongly associated with a particular species.

In addition, the relationship between soil physical and chemical properties and earthworm community composition shows interesting results. In contrast to our hypothesis, the percentage of soil cover had little influence in explaining differences among sites. For future research, it will be interesting to measure variables like cover heterogeneity, richness or chemical quality, which are better factors to describe the rotation schemes and more important determinants of earthworm composition. Soil moisture has been proved to be an important factor determining earthworm community. In our study, this factor together with organic matter content was outstanding for the three *Aporrectodea* species but mainly for *A. caliginosa*, which has been already demonstrated to be benefited by both properties (e.g. Didden, 2001; Eriksen-Hamel and Whalen, 2006). The relationship between *A. caliginosa* and soil moisture and OM content triggers the link between this species and three grasslands (M-Gr, C2-GR, P-Gr), where both soil properties were favoured by the absence of anthropogenic interventions. The species *M. phosphoreus*, *G. parecis*, and *Belladrilus* sp. were all inversely related to bulk density, evidencing their preference for less compacted soils, and also positively related to higher pH values. These disturbance-sensitive native species (Momo and Falco, 2009) were characteristic mainly of the grasslands and the organic sites, and in terms of locations, of Bengolea and Alejandro Roca. On the other hand, *Eukerria* and *M. dubius* showed a greater tolerance to compacted soils and were mainly linked to Pergamino and Cabrera-Deheza locations. Historical and biogeographical factors together with management factors can be determining the presence and the abundance of those species. Decaens (2010) indicated that, unlike for above-ground taxa, spatiotemporal patterns of soil biota are still poorly understood. However, this author noticed a relationship between certain soil properties and species richness, but indicated the need for further experimental assessments to identify the specific factors. On the other hand, it has also been observed that, regardless of resource availability, there is an upper limit to the number of earthworm species that can coexist, although functional richness may follow a different pattern (Decaens et al., 2008; Lavelle et al., 1995). Accordingly, competitive exclusion may play a very important role in determining earthworm spatial pattern distribution together with soil quality and land use and management (Decaens et al., 2008; Decaens, 2010; Jimenez et al., 2006).

Moreover, changes in the composition of earthworm community due to improvement of management conditions are necessarily slow, since the natural rate of dispersal of most earthworm species seems to be low, in general less than 10 m per year (Lavelle and Spain, 2003). Slow dispersal of earthworms strongly affects their ability to recolonize soils and therefore to respond to changes in management systems with changes in composition. The conditions for earthworm recolonization in agricultural soils are worsened in the Pampas region because of the large plot areas, usually about 100–200 Ha. The availability of natural habitats and their proximity to agricultural ones is a key feature to ensure a source of species when earthworm community composition recovery through management is aimed. This is also a usually underestimated and rarely considered aspect in the Pampas region; very few relicts of the original landscape persist and very large zones with only agricultural soils are most frequent. There is a crucial need for considering systems and practices beneficial to earthworm conservation

and development given their relevance in agroecosystems. Earthworms are able to improve soil aggregation and macroporosity, stabilize soil organic matter, and accelerate nutrient mineralization, and may decrease the negative impact of some pests and pathogens (Bertrand et al., 2015; Lavelle et al., 2007). Furthermore, earthworms are involved not only in improving physical and chemical conditions for plant growth, but also in driving ecosystem processes and upgrading the ecosystem performance (Lavelle et al., 2016).

Beetles are a very diverse species group; therefore, understanding the changes in community composition produced by agricultural management generally requires a deeper taxonomic approach. The studied grasslands had the highest beetle richness of all the studied systems, which agrees with previous findings (e.g. Purtauf et al., 2005). Beetle composition in organic sites was completely different from that in all the other systems and strongly dominated by Carabidae species. Changes in beetle community after transition from conventional to organic farming have been observed by other authors (Henneron et al., 2014; Shah et al., 2003). As well as we found the highest abundance in Org sites (among agricultural ones), and linked to Carabidae species, Shah et al. (2003) found higher abundances of Carabidae in organic than in conventional farming, but they found lower diversity in organic farming, which was likely related to a high dominance of one species. Their study agrees with the results presented here about the compositional change in beetle community and the dominance of one family in the organic farming. Pffner and Luka (2003) found changes in carabid community between organic farming and other agricultural management systems, but they also discussed that the effect of management practices seems to interact with many other factors, such as landscape characteristics and years of application; therefore, linking certain management practices with certain responses in the soil beetle communities is not a simple task. Purtauf et al. (2005) did not find a significant effect of organic management compared to conventional either on species richness or in activity density of carabids. Instead, they highlight the importance of the landscape context in shaping carabid communities, irrespectively of management type.

Among conventional agriculture systems, we observed two groupings of sites. One of them consists of Nt-R1 and Til-R, which were strongly associated with Staphylinidae sp. 3 and Polyphaga sp. 1. The other grouping was linked to Staphylinidae sp. 2, and included Nt-R2, Nt-R3 and Til systems. Staphylinid beetles are mostly predators but there are also fungal feeder species (Lavelle and Spain, 2003). Clough et al. (2007) did not find differences in species richness between organic and conventional sites; they also found that the management effects strongly depend on the feeding group. Thus, predatory abundance was favoured by conventional farming but detritivores were more abundant in organic fields. Our results agree with those reported by Shah et al. (2003), who found very similar diversity indexes for staphylinid beetle communities in conventional and organic systems, but higher abundances in the conventional one. There is evidence that some soil decomposers and predators are more abundant but not more species-rich in organically managed soils (Bengtsson et al., 2005; Tuck et al., 2014). Higher taxonomic resolution for the complex family Staphylinidae would allow us to better understand the link between their feeding habits and the effect of the management systems here analysed. In fact, for Staphylinidae and Carabidae, Andersen and Eltun (2000) found clear differences and sometimes opposite effects of organic or conventional agriculture on species belonging to the same family, and therefore emphasize the need for studying species of those diverse groups individually.

The relationship between soil properties and beetle community composition was strongly influenced by geographic location, as observed in the co-inertia analysis. Staphylinidae and Carabidae species were the most important in determining the relationship with soil properties together with Cicindelidae and Polyphaga sp.2. The link between soil cover and staphylinid species was weaker than expected. The grouping in the co-inertia plane related to the geographic location

seems to indicate that beetle community composition was more associated with the surrounding environment than with the farming systems. Purtauf et al. (2005) observed that landscape context (i.e., percent cover of surrounding grassland) had a strong effect on species richness, regardless of conventional or organic management type. It is likely that environmental factors that operate at levels higher than the plot scale are also strongly affecting beetle community in our study.

Ant species composition was not different among systems, and each location had a characteristic community composition. Accordingly, in a comparison of ground-dwelling arthropods among different systems of mulching, tillage, herbicide application and fertilization, Miñarro et al. (2009) found that ants was the only group not affected by management practices. However, Ponce et al. (2011) found significantly higher abundances in organic vs. conventional management.

Co-inertia analysis showed a significant correlation between ant species and soil properties. The omnivorous *Linepithema* sp.1 was related to low values of soil bulk density and high values of soil pH. All predator species were grouped together (*Acanthostichus* sp.1, *Cheliomyrmex* sp.1, *Pheidole* sp.1 and *Ponerinae* sp. 1) and were strongly related to high soil moisture content and to the M-Gr site. *Acromyrmex* sp.1 (leaf-cutting and fungus-growing ants), *Brachymyrmex* sp. 1 (cryptic) and *Solenopsis* sp.2 (omnivorous) were related to higher soil cover and to grassland and no-till sites. A third grouping includes *Solenopsis* sp. 1 (omnivorous), *Myrmicinae* sp. 1 and *Formicinae* sp.1, associated with high soil organic matter content. Ants are recognized as ecosystem engineers with the ability to modify soil physical and chemical attributes (Domínguez-Haydar and Armbricht, 2011; Frouz and Jilkova, 2008); at the same time, however, there is also strong evidence that they are reciprocally affected by soil characteristics. Nevertheless, the relationship between soil-dwelling ants and soil properties depends on the type of habitat and the ant species present. In grasslands, Boulton et al. (2005) found an important association between soil physical and chemical properties and both richness and abundance of the whole ant community and the dominant ant species. In mixed forests, Wang et al. (2001) found a negative relationship between soil moisture and ant abundance and diversity. Hill et al. (2008) studied the relationship between ant communities and environmental variation in four habitat types: prairie, actively grazed pasture, oak-hickory forest, and pine-oak flatwoods. They found that the influence of soil attributes varies according to the ant species; for example, Camponotini species were related to soils with high organic matter content. Instead, Dolichoderinae ants showed a relative negative association with soils with high organic matter, since more sandy soils provide better nesting sites. Peck et al. (1998) and Perner et al. (2005) observed a significant correlation between species assemblages and soil properties, such as cation exchange capacity, soil pH, moisture, organic carbon and N content, and sand and clay content. However, Ekschmitt et al. (2003) found that ant richness in grasslands was best explained by topographic factors than by soil quality parameters.

5. Conclusions and outlook

Overall, agricultural management strategies highly conditioned the composition of soil macrofauna community. Moreover, the edaphoclimatic conditions also had an important influence in shaping the macrofauna community composition, likely linked to the effect of soil chemical and physical properties on soil macrofauna community. The natural grasslands had the most distinctive and diverse communities compared to the different crop systems. These results emphasize an ongoing process of continuous loss of soil biodiversity in Argentine agricultural soils. This phenomenon should be more seriously addressed by farmers and policy makers. Indeed, this low diversity in agricultural soils is of especial concern because the loss of key species may easily hamper ecosystem functions (Bender et al., 2016).

Extensive organic agriculture stood out by its ability to promote an earthworm community that was, in terms of abundance and richness,

more similar to grasslands than to all the studied systems of conventional agriculture. This result is outstanding, since it could be marking the initial point to improve earthworm diversity conservation and associated self-functioning of the soil. In the studied soils, earthworms are the most important ecosystem engineers and are involved in most of the soil processes that enhance plant production. Maintaining diverse and active earthworm communities in the long term is crucial for achieving sustainable agricultural systems. Extensive organic farming would be accomplishing that goal while reducing the human health issues linked to agrochemical air, soil and water pollution.

On the other hand, the negative impact of conventional agriculture on soil biology decreases when high rotation schemes with the inclusion of winter cover crops are used, as has been demonstrated here as well as in other studies. The persistence of farmers in maintaining soybean monocropping and the lack of governmental policies to prevent it need to be reconsidered, since soybean cropping can be a profitable business today but in the mid and long term will result in biodiversity loss that will be hardly recovered.

The path towards more sustainable agricultural systems in the Pampas region is a complex one. Environmental, social, political, economic, and, certainly, agronomical aspects need to be considered when dealing with that goal. However, one of the first steps is to promote agricultural management systems with the ability to restore the functional biodiversity of the agricultural landscape (Altieri, 1999). The present work contributes to the knowledge about the practices favouring macrofauna diversity, which in turn should favour soil functioning. Further studies are necessary to assess not only diversity but also soil ecological processes.

Author contributions

A. Domínguez and J.C. Bedano conceived and performed the experimental design and the field research to obtain the data which support the manuscript. A. Domínguez performed the taxonomic determinations. J.J. Jiménez suggested the statistical approach and all the authors contributed in the statistical procedures for analysing the database. A. Domínguez wrote the draft and J.C. Bedano, J.J. Jiménez and C. Ortiz critically reviewed and contribute to the final manuscript. All authors have approved the final article.

Conflicts of interest

The authors declare no conflict of interest.

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