Long-term effect of tillage, rotation and nitrogen fertilizer on soil quality in a Mediterranean Vertisol

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Abstract

Studies of the impacts of the interactions of soil agricultural practices on soil quality could assist with assessment of better management to establish sustainable crop production systems. Our objective was to determine the effects of tillage system, crop rotation and N fertilisation on soil total N and organic C (SOC), labile fractions of organic matter (water soluble carbon, WSC, and active carbon, AC), nitrate content, and soil enzymatic activities (dehydrogenase (DHA), β-glucosidase (Glu) and alkaline phosphatase (AP)); within the framework of a long-term located in a Mediterranean dryland Vertisol in SW Spain.

The highest SOC, total N, WSC, AC and Glu contents were obtained under NT in the WW and in WFb rotations at 0-5 cm depth and at 150 kg N ha^{-1} rate; the lowest SOC was obtained with the WF rotation in both tillage systems. The rate of 150 kg N ha^{-1} increased SOC, WSC, total N, nitrate, Glu and AP contents compared to 50 and 0 kg N ha^{-1}, which means that precautions should be taken with long-term N fertilisation to avoid leaching of nitrates below the tillage layer. Combination of NT with any biannual rotation except fallow could be an adequate

sustainable management, under our conditions. Positive effects on organic C fractions, total N and enzymatic activities were found, indicating that these agriculture practices can improve soil quality of Vertisols.

Key-words: rain-fed agriculture; soil organic matter; conservation tillage; soil enzymatic activities.

1. Introduction

Cultivation practices such as tillage and crop rotation can modify soil organic matter (SOC). Impacts of tillage on soil organic matter in surface soils have been well documented, but results vary due to soil type, cropping systems, residue management, and climate (Paustian et al., 1997). Tillage produces a reduction of organic matter content due to enhanced mineralisation of crop residues, disruption of soil aggregates and increasing aeration (Sainju et al., 2006). By contrast, conservation tillage (reduced and no-till practices) increases soil organic carbon in the surface layer (Six et al., 1998; Sainju et al., 2006; Melero et al., 2009; López-Bellido et al., 2010), improves soil aggregation (Coulombe et al., 1996), and preserves soil resources better than conventional tillage practices (Six et al., 1998). Soil carbon sequestration in tilled and non-tilled areas can be influenced by crop management practices, due to differences in plant C inputs and their rate of mineralisation (Sainju et al., 2006). Crop rotations have shown positive effects on enhancing SOC (Campbell et al., 1996; López-Bellido et al., 2010), especially in crop rotations that include a leguminous species (Omay et al., 1997; Potter et al., 1998; Ashraf et al., 2004; López-Bellido et al., 2010). Legumes add both organic matter and N to the soil (Omay et al., 1997; Sainju et al., 2003) and increase soil fertility. Espinoza et al. (2007) reported that the changes observed in total organic C and N in response to tillage and rotation were significantly related to the quantity and quality of plant residues returned to the soil. The C/N

ratio of a crop is a determining factor for soil N availability, regardless of placement of their residues in the soil (Upendra et al., 2005). Leguminous crops have low C/N ratios while non-leguminous ones, such as rye and wheat, have low N content and thus favour N immobilisation over soil N mineralisation by microorganisms (Upendra et al., 2005). N fertilisation can also increase SOC by increasing crop biomass production and the amount of residue returned to the soil (Gregorich et al., 1996). Therefore, crop rotations and N fertilisation can influence SOC sequestration in tilled and non-tilled soils due to differences in mineralisation rates of crop residues and soil organic matter. Several authors, including Sainju et al. (2003), observed that legume cover crops and N fertilisation can increase yields and biomass production compared with non-legume or no cover crops and N fertilisation, due to increased C and N supply. In addition, as much as 7 to 43% of the total plant biomass C can be contributed by roots (Kuo et al., 1997). Sandeep et al. (2010) argued that enhancement of root development helps to improve soil physical properties and carbon sequestration. Roots supply C to soils through quantity and quality of root detritus, release of root exudates and C transfer to root symbionts (Tresder et al., 2005). Kong et al. (2010) suggested that root C contributes more to overall C stabilisation than C from residues, which supports a nascent body of research demonstrating greater retention of root-derived than residue-derived C in soil organic matter (SOM). Several authors (Allmaras et al., 2004) have reported that corn roots contributed 1.6 to 3.5 times more C to SOC than did stover. SOM decomposition always occurs with plant roots, thereby inevitably becoming entangled with both the soil component and the plant component. Any environmental conditions that affect either the plant functions or the soil functions, or both, inevitably modulate root effects on SOM decomposition (Cheng and Kuzyakov, 2005).

Although SOC is an important component of soil quality and productivity, measurement of SOC alone does not adequately reflect changes in soil quality and nutrient status (Franzluebbers et al., 1995). Measurement of more labile fractions of SOC, which change rapidly over time,

such as active carbon (AC) and dissolved organic carbon (WSC), could better reflect changes in soil quality due to changes induced by soil management practices (Weil et al., 2003; Melero et al., 2009). However, the contribution of crop residues and root exudates to the WSC and AC pool is not entirely understood. Thus, assessment of the distribution of soil C and N pools with depth may provide an account of sequestration potential.

Moreover, microbial activity-based indicators of soil quality, such as assessments of the annual effects of field labours on soil fertility, may respond to disturbances over a shorter period of time than those based on physical or chemical properties, such as SOC. Therefore, microbiological properties such as soil enzyme activities have been suggested as potential indicators of soil quality because of their essential role in soil biology, ease of measurement and rapid response to changes in soil management (Kandeler et al., 1999). Generally, tilled soils present lower soil enzymatic activity values than do non-tilled soils (Roldán et al., 2005; Melero et al., 2008 and 2009). Comparisons between crop rotations and monoculture systems have shown that enzymatic activities are more sensitive to positive effects of polyculture (Acosta-Martinez et al., 2003). The inclusion of a leguminous crop in the crop rotation increases soil enzymatic activity values because the rhizosphere of leguminous crops may more actively secrete higher amounts of exudates than non-leguminous crops (Sainju et al., 2005; Sainju et al., 2006).

There are some studies on the effects of different tillage management systems on soil quality under different climatic conditions (Kandeler et al., 1999; Roldán et al., 2005; Melero et al., 2009). However, information on changes produced by the interaction of tillage system, crop rotation and rate of N fertilisation in semiarid Mediterranean agroecosystems is very scarce. To address this issue, the objectives of this research were: (i) to assess the long-term impact of no tillage and conventional tillage on soil organic fractions and enzymatic activities and their distribution in the soil profile of a dryland Vertisol and (ii) to determine whether different crop

rotation systems and N fertilisation rates influenced the soil organic fractions and soil biochemical status in tilled and non-tilled soils. We hypothesised that the effect of different tillage systems on chemical and biochemical properties of a Vertisol would be influenced by crop rotations and N fertilisation rates; the inclusion of a leguminous species in the rotation would have more effect than other rotations and monoculture on soil quality.

2. Material and Methods

2.1 Experimental site and management of tillage systems

A long-term field experiment was begun in 1986 in Córdoba, Southern Spain (37°46 N and 4°31 W, 280 m above sea level), on a Vertisol (Typic Haploxerert) typical of the Mediterranean region (water holding capacity about 350 mm m^{-1}), where rain-fed cropping is the standard practice. The experiment was designed as a randomised complete block with a split-split-plot arrangement and three blocks. The main plots were divided by tillage system (non-tillage, NT and conventional tillage, CT). Subplots were divided by crop rotation, with four different two-year rotations [wheat-sunflower (*Triticum aestivum* L.- *Helianthus annuus* L.) (WS), wheat-faba bean (*Vicia faba* L.) (WFb), wheat-chickpea (*Cicer arietinum* L.) (WC), wheat-fallow (WF)], and a monoculture of wheat-wheat (WW); sub-subplots were divided by N-fertilisation rates (0, 50 and 150 kg N ha^{-1}), applied to wheat only. The area of each subplot was 50 m^2 (10 m × 5 m). NT plots were seeded with a no-till drill (Great Plains). Weeds were controlled with glyphosate and *(*4-chloro-2-methylphenoxy*)* acetic acid (MCPA) at a rate of 0.5 + 0.5 l a.i. ha^{-1} prior to seeding. The CT treatment included mouldboard ploughing and disk harrowing and/or vibrating tine cultivation to provide a proper seedbed. Hard red spring wheat (cv. Gazul) was planted in rows 18 cm apart in December of each year at a rate of 150 kg ha^{-1}. Faba bean (cv. Alameda) was planted in 50 cm-wide rows in November at a seeding rate of 170 kg ha^{-1}. Chickpea (cv. Zoco) was planted in 35 cm-wide rows in March at a seeding rate of 80 kg ha^{-1}. Sunflower (hybrid cv. Pioneer PR63A76) was planted in 50 cm-wide rows in March of each

year at a rate of 5 kg ha^{-1}. Nitrogen fertiliser was applied to wheat plots as ammonium nitrate. At all fertiliser application rates, half was applied before sowing (incorporated by disk harrowing in CT plots and surface-broadcast in NT plots). The remaining N was topdressed at the beginning of wheat tillering. Every year, wheat plots were also supplied with P fertiliser at a rate of 65 kg P ha^{-1}. The fertiliser was incorporated in CT soil and banded with a drill in NT plots.

2.2 Soil sampling

Bulk soil samples were collected at four depths: 0-5, 5-10, 10-30 and 30-50 cm, prior to soil tillage and sowing of the wheat crop of each crop rotation, in October 2008. From each subplot, three soil scores were taken for further analysis. Each soil score sample was a composite taken by mixing within each subplot. The field-moist soil was sieved (2 mm), homogenised and then divided into two subsamples: one was air-dried for various chemical analyses, while a second subsample was stored at 4ºC in plastic bags, loosely tied to ensure sufficient aeration and to prevent moisture loss, before biological analysis.

In air-dried subsamples, total C, inorganic C and total N of the soil samples were determined by the dry combustion method (Nelson and Sommers, 1996) in an elemental analyser (EA 3000 Eurovector SpA, Milan, Italy). The SOC concentration was calculated by subtracting inorganic C from total C. Soil NO_3^-–N was determined by the colorimetric method of Griess-Illosvay modified by Markus et al. (1985) and Kempers (1974). A continuous flow colorimeter (QUATRO, Bran Luebbe, Norderstedt, Germany) was used. WSC was determined in a 1:10 aqueous extract using a TOC V-CSH Shimadzu analyser. Active carbon (AC) was determined according to Weil et al. (2003).

In field-moist subsamples, dehydrogenase activity (DHA) was determined according to Camiña et al. (1998), β-glucosidase activity (Glu) was measured as indicated by Eivazi and

Tabatabai (1988) and alkaline phosphatase activity (AP) according to Tabatabai and Bremner (1969).

For each microbiological analysis, three replicates per collected sample were analysed. Results were based on the oven-dried weight of the soil.

2.3 Statistical analysis

Annual data for each variable were subjected to analysis of variance (ANOVA), using a randomised complete block design combined with the error term according to McIntosh (1983). Tillage systems and blocks (replications) were considered random effects, while crop rotation, N fertiliser and soil depth were considered fixed effects. Means were compared using Fisher's protected least significant difference (LSD) test at $P < 0.05$. LSDs for the different main effects and interaction comparisons were calculated using the appropriate standard error terms. The Statistix v. 8.1 (Analytical Software, 2005) package was used for this purpose.

3. Results

3.1 Effect of tillage, N fertilisation rate and crop rotation on soil organic carbon fractions (SOC, AC, WSC) and distribution in soil profile

3.1.1 Total organic carbon

Soil organic carbon (SOC) was significantly affected by tillage x crop rotation type x soil depth and by tillage x N fertiliser rate x soil depth interactions (Table 1). Differences between tillage systems (NT and CT) were seen only with WFb and WW rotations at 0-5 cm depth (Fig. 1). The highest SOC values were obtained in wheat-wheat (WW) and in wheat-faba bean (WFb) rotations with 150 kg N ha^{-1} at 0-5 cm depth under NT (Figs. 1, 2), whilst the lowest SOC were obtained with the wheat-fallow (WF) rotation in both tillage systems (Fig. 1). In general, SOC contents decreased with depth in all crop rotations and in both CT and NT systems (Fig. 1).

3.1.2 Labile organic fractions (AC and WSC)

Tillage x crop rotation x soil depth interaction had a significant influence ($p<0.01$) on active carbon (AC) content (Table 1). Differences between tillage systems were not found in either crop rotation or soil depth (Fig. 3). N fertiliser rates did not influence soil AC concentrations (Table 1). However, significant differences in AC between WF and WW and WFb rotations were reported, with the lowest AC mean values in the WF rotation (Fig. 3). In most of the crop rotations, a significant decrease in AC with increasing soil depth was reported, except in the wheat–sunflower (WS) rotation (Fig. 3).

Water soluble carbon (WSC) values were affected by tillage x crop rotation x N fertiliser rate x soil depth interaction (Table 1). Concentrations of WSC were statistically higher in NT than in CT with WW, WFb and WS rotations at 0-5 cm depth and 50 kg N ha^{-1} rates and also with WW at the 150 kg N ha^{-1} rate (Fig. 4). WSC contents were statistically higher in WW than in WF and WC rotations, especially at 0-5 cm depth and 50 and 150 kg N ha^{-1} rates under NT (Fig. 4). WF and WC rotations showed the lowest WSC values (Fig. 4). In general, a significant reduction of WSC values was observed with increasing depth in WW and WFb rotations, with the greatest WSC contents seen in the upper layers under NT (Fig. 4).

3.2 Effect of tillage, N rate and crop rotation on total nitrogen and nitrate contents and distributions in the soil profile

Total N content was strongly affected by tillage x crop rotation x soil depth interaction ($p< 0.001$) (Table 1). Total N content was significantly higher under NT than under CT in all crop rotations studied, especially at 0-5 cm depth (Fig. 5). Total N was also higher under NT at 30-50 cm in WF, WS and WW rotations (Fig. 5). However, the highest total N contents were reported for WW and WFb rotations, with the lowest ones for WF and WS (Fig. 5).

Soil nitrate contents were greatly affected by tillage x crop rotation x N fertiliser rate x soil depth interaction (Table 1). Tilled soils showed higher nitrate contents than non-tilled soils at 150 kg N ha^{-1} with WW rotation and at 50 kg N ha^{-1} with WFb, WS and WC rotations, especially below a depth of 30 cm (Fig. 6). The lowest soil nitrate content was reported with the WS rotation and the highest with WFb rotation (Fig. 6). In general, increased nitrate concentration was observed with increasing depth (Fig. 6).

3.3 Effect of tillage, N rate and crop rotation on soil enzymatic activities and distribution in the soil profile

Both dehydrogenase (DHA) and alkaline phosphatase (AP) activities were affected by tillage x depth and by N fertiliser rate x depth interactions (Table 1).

DHA values did not show significant differences among tillage systems (CT and NT). DHA values were significantly different among WF and other crop rotations, with the WF rotation having the lowest DHA activity (Fig. 7). N fertilisation rate had no significant effect on DHA (Fig. 7).

On the other hand, a significant increase of AP values was seen only in non-tilled soils compared to tilled soils at 0-5 cm depth (Fig. 8). The highest AP values were obtained with the WW rotation at 50 kg N ha^{-1}. Also, it was found that AP activity was higher at 50 kg N ha^{-1} than without N fertilisation at all depths except the deeper layer (30-50 cm) (Fig. 8). A decrease of DHA and AP values with increasing depth, especially under no tillage, was observed (Figs. 7, 8).

A significant interaction among tillage x crop rotation x N fertiliser rate x soil depth was observed for β-glucosidase activity (Glu) (Table 1). In general, higher Glu values were found in non-tilled than tilled soils at 0-5 cm depth, at both N rates (50 and 150 kg N ha^{-1}), in all crop rotations studied (Fig. 9). In addition, a significant reduction of Glu values in deeper layers

compared to upper layers was observed in all crop rotations (Fig. 9). The lowest Glu values were reported for WF rotations (Fig. 9).

4. Discussion

The effects of tillage system on SOC fractions and total N were strongly influenced by crop rotation. Total N content was higher in the surface layer (0-5 cm depth) in non-tilled than in tilled soils, in all crop rotations, although the highest total N concentrations were seen in WW and WFb rotations. The SOC content was also higher at 0-5 cm depth in NT compared with CT, but only in WFb and WW rotations, which reached higher values than other crop rotations. Similar results were reported by Wright et al. (2007), who reported a significant increase in SOC and total N in NT compared with CT systems for both wheat monoculture and wheat-soybean rotation. Friedel et al. (1996) reported higher SOC contents in 0-10 cm depth with rotary cultivation than with ploughing, especially with rape-cereal rotation compared with legume-cereal rotation, which could be related to the amount and distribution of crop residues in the soil profile.

In our study, the WSC was highest in the WW rotation under NT in the surface layer. Wright et al. (2007) also found that WSC content was dependent on both tillage and cropping intensity, although a more pronounced increase was found under crops that produced more residues and under intensive cropping sequences than under continuous wheat. By contrast, Friedel et al. (1996) did not find an effect of either tillage system or crop rotation on WSC content in upper layers. McGill et al. (1986) reported that C turnover rates of the soil microbial biomass decreased with increasing crop residue inputs in the rotation, reflecting a relatively lower C demand by microorganisms.

On the other hand, Wright and Hons (2005) reported that there was greater soil C and N storage in surface soils under crop rotations than under monocultures. By contrast, in our case, similar results were found under WW and WFb rotations. Residues of cereal monocultures (high C/N ratio) may diminish OM degradation, contributing to more similar SOC levels than other crop rotation systems (including legumes, with a low C/N ratio) (Sisti et al., 2004). Blair et al. (1998) also found that a wheat-legumes rotation increased labile C fractions but had no effect on total C compared with continuous wheat.

In general, most variables (SOC, AC, WSC, nitrates and total N) reached lower values under WC rotation than under WFb ones. This may be due to the greater previous wheat yield in the WFb rotation (4819 and 3910 kg ha^{-1} in NT and CT, respectively) than in WC rotation (3734 and 2648 kg ha^{-1} in NT and CT, respectively). Also, in 2007, the WFb rotation generated greater amounts of residues than the WC rotation did. In addition, faba bean residues had a greater N concentration (0.98%) than *Cicer* residues (0.67 %).

Comparatively low values of these variables (SOC, AC, WSC, nitrate and total N) were also recorded in the wheat-sunflower rotation, which might be related to the low degradation rate of the great quantity of sunflower residue produced, with high contents of barely-degradable components (Corbeels et al., 2000).

In general, reductions of SOC, labile organic fractions and total N contents were seen with increasing soil depth. The stratification of SOC fractions with depth was greater under conservation tillage than under conventionally tilled soils. Salinas-Garcia et al. (2002) obtained similar results, which were attributed to the mixing of crop residues and soil by intensive tillage. They also reported that crop residues are distributed more uniformly throughout the tillage zone under conventional tillage than under conservation tillage, in which residues remain at the soil surface.

Studies related to the effects of tillage systems below the plough layer (30 cm depth) are scarce (Poirier et al., 2009). Some authors (Baker et al., 2007; Gal et al., 2007) have shown that the carbon sequestration advantage of conservation tillage in upper layers (0-15 cm) disappears in deeper layers, with greater accumulation of SOC and total N under CT at 20-60 cm depth. In our study, there were no differences in SOC between tillage systems below the plough layer in any rotation. Kern and Johnson (1993) and Angers et al. (1997) also reported similar organic carbon contents between NT and CT below 15 cm depth. In our study, the only significant difference was found for WW, in which a greater SOC content was detected at 50 cm depth under NT. The general absence of differences could be related to the soil type studied, a Vertisol, which contains expandable mineral clays that shrink upon drying, forming wide cracks into which topsoil and organic materials tend to fall, allowing a better mixing of organic materials and soil particles and reducing concentration gradients typically found in NT systems (Dalal et al., 1991). This seemed to be corroborated by the trend seen in nitrate contents, which was opposite those of SOC fractions, with higher concentrations in deeper layers than upper ones; this could indicate consistent leaching, favoured by the wide cracks typical of Vertisols.

Nitrogen fertilisation had an influence on SOC and total N content in both tilled and non-tilled soils. Nevertheless, lower C and N stocks were measured in tilled soils than in non-tilled soils, especially with the highest N fertiliser rate; this was probably due to the greater decomposition of crop residues and soil OM under CT with the addition of N fertilisers (Poirier et al., 2009). Other sources have reported different responses in this context. Knorr et al. (2005) reported that N addition stimulated the decomposition of residues with low lignin content but inhibited the decomposition of those with high lignin content. On the other hand, Halvorson et al. (2002) reported an absence of effect of N fertiliser on SOC sequestration in a WF and in a spring wheat-winter wheat-sunflower rotation.

Nitrogen fertilisation, rotation and tillage systems had no great effect on nitrate contents in upper layers (0-10 cm depth). However, Roldan et al. (2005) found lower soil nitrate concentrations in non-tilled soil than in tilled soil at 0-20 cm depth, which could be related to higher losses of N through denitrification under NT. By contrast, we found higher nitrate contents in tilled soils compared with non-tilled soils in deeper layers (>30 cm depth), especially with the WW rotation with the highest N rate (150 kg N ha^{-1}). These results could reflect combined effects of intensive tillage, increasing the amount of N mineralised in tilled soils due to better aerobic conditions than in non-tilled soils, with major nitrate leaching in these soils. Similar results were reported by Halvorson et al. (2001), who observed a higher accumulation of nitrate below the root zone with CT and reduced tillage, than with NT with increasing N fertiliser rate. The authors argued that NT with annual cropping may reduce the quantity of soil nitrate available for leaching compared with CT.

Soil enzymes can be affected by soil management (tillage, crop rotation, fertilisation, and crop residues) (Klose et al 1999). In general, our study showed the lowest enzymatic activities in the WF rotation. Acosta-Martinez et al. (2007) reported that fallow periods and CT negatively affected to soil enzymatic activities, such as Glu and AP activities, especially at 0-5 cm depth. Both enzymes' activities increased with the decrease of fallow periods in the rotation. In general, soils under crop rotations receive a high input and diversity of organic materials, which is usually conducive to higher levels of microbial biomass and enzymes, compared with monoculture systems (Klose and Tabatabai et al. 1999).

We detected higher Glu and AP activities in NT than in CT in surface layers. Several studies under different climatic conditions have reported the effects of different tillage systems on soil enzymes, showing higher contents under NT than under CT (Roldan et al., 2007; Mina et al., 2008; Melero et al., 2009). Salinas-García (2002) and Eivazi et al. (2003) reported that microbial activity was more uniformly distributed along the plough layer under intensive

tillage, whereas in non-tilled soils it tended to be concentrated in the surface layer. Eivazi et al. (2003) reported that changes in the enzyme activities in the profiles of tilled and non-tilled plots might be a consequence of the large relative changes in the populations of aerobic and facultative anaerobic microorganisms. Also, differences in the amounts and qualities of residues (e.g., carbohydrate and lignin contents) left on soils by different rotations can also influence decomposition rates (Klose and Tabatabai, 2000). Acosta-Martinez et al. (2007) reported that Glu tended to be higher under wheat residues in wheat-corn-fallow and wheat-corn-millet rotations than under corn residue in corn-fallow-wheat and corn-millet-wheat rotations.

The effects of inorganic fertilisers on soil enzyme activities may vary according to the composition and amount of applied fertiliser, soil properties and enzyme assayed (Eivazi et al., 2003). We have not found any effect of different N fertilisation rates on DHA, whilst Glu and AP activities were increased at high N rates (50 and 150 kg N ha^{-1}). However, other authors (Simek et al., 1999; Kanchikerimath and Singh, 2001; Bohme and Bohme, 2006) have reported an inhibitory effect of long-term applications of large amounts of mineral fertiliser on DHA and AP activities. Saha et al. (2008) also observed a strong decrease in DHA and AP activities upon application of mineral fertilisers in comparison with organic fertilisers or with combined inorganic and organic fertilisation. It is possible that differences in soil characteristics had some influence on the different behaviour of the assayed enzymes.

In general, long-term conservation tillage combined with a basic biannual wheat-crop rotation, especially with legumes, increased the soil quality of the dryland Vertisol studied.

Conclusions

Under our experimental conditions of dryland Mediterranean agriculture, tillage system was not the main factor in determining the changes in soil C and N pools and in its biochemical

status. In the studied soil, a Vertisol, the results were more dependent on the interaction of tillage system with different factors such as crop rotation and N fertilisation rates.

In general, long-term non-tillage enhanced soil C and N storage and soil enzymatic activities in the superficial layer (0-5 cm depth), mainly with legume rotation or with cereal monoculture. Below the ploughing layer, the positive effect of non-tillage on soil C and N accumulation in the upper layer, compared to traditional tillage, was only maintained in the wheat monoculture.

Nitrogen fertilisation had no effect on soil C sequestration and dehydrogenase activity, although a high rate of 150 kg N ha^{-1} led to the highest total nitrogen, water soluble carbon, β-glucosidase and alkaline phosphatase contents in non-tilled soils. Nevertheless, precautions must be taken in Vertisols because N fertilisation, especially at a high rate (e.g., 150 kg N ha^{-1}), can raise the risk of nitrate leaching below the tillage layer in both conservation and traditional tillage systems.

Implementation of fallowing as biannual rotation with wheat significantly reduced organic carbon fractions, total nitrogen and enzymatic activities in both tillage systems, especially without N fertilisation. Therefore, mouldboard ploughing and fallowing are not adequate agricultural managements because these favour C, N and nutrient losses and decrease soil biochemical activity.

Acknowledgements

Authors want to thank to Commission Interministerial de Ciencia y Tecnología (CICYT) through projects (AGL2005- 2423 and AGL2006-02127), which allowed this work to be carried out. S. Melero thanks to CSIC for her contract through program "JAE-Doc". The authors would like to thank ABECERA for providing the land and allowing us to use their field

facilities. Special thanks are also expressed to Joaquín Muñoz and José Muñoz for their field work.

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Table 1. The properties of the Vertisol used in field experiments. Córdoba (Spain)

		Depth (cm)		
		0-30	30-60	60-90
Fine sand (g kg^{-1})		127	143	187
Silt (g kg^{-1})		179	152	26
Clay (g kg^{-1})		694	705	787
pH (1:2.5 soil-water)		7.7	7.6	7.6
Organic matter (g kg^{-1})		10.2	7.4	5.3
Calcium carbonate equivalent (g kg^{-1})		75	93	71
CEC (cmol kg^{-1})		46.5	36.6	30
Bulk density	No tillage	0.95	1.06	1.10
(t m^{-3}):	Conventional tillage	0.90	1.05	1.04

CEC: cation exchangeable capacity

Table 2. Significant effects of tillage system, crop rotation, N fertilizer, and soil depth on different variables after 20 yr of continuous experiment in a Vertisol under rainfed Mediterranean conditions.

	Parameters							
Source	OC	AC	WSC	Total N	Nitrate	DHA	AP	β-glu
Tillage system (T)	ns	ns	ns	**	*	ns	ns	ns
Crop rotation (R)	**	*	***	***	***	***	**	***
T × R	**	ns	ns	ns	*	ns	ns	ns
N fertilizer (N)	*	ns	*	*	***	ns	*	**
T × N	*	ns	ns	*	*	ns	ns	*
R × N	ns	ns	*	ns	***	ns	ns	*
T × R × N	ns	ns	**	ns	*	ns	ns	ns
Depth (D)	***	***	***	***	***	***	***	***
T × D	***	***	***	***	***	***	***	***
R × D	***	***	***	***	***	ns	ns	***
N × D	*	ns	ns	ns	***	***	*	***
T × R × D	ns	**	*	**	*	ns	ns	**
T × N × D	*	ns	ns	***	*	ns	ns	***
R × N × D	ns	ns	ns	ns	***	ns	ns	***
T × R × N × D	ns	ns	*	ns	*	ns	ns	**

OC: organic carbon, AC: active carbon, WSC: water soluble carbon, DHA: dehydrogenase, AP: alkaline phosphatase: β -Glu: β-glucosidase

* Significant at the 0.05 probability level.

** Significant at the 0.01 probability level.

*** Significant at the 0.001 probability level.

ns, not significant.

Table 3. Above-ground residues (in 2007) of different crops in rotation with wheat in the no tillage and conventional tillage plots. Means followed by the same letters are not significantly different at $p < 0.05$ according to LSD within each rotation.

	Above-ground residues (kg ha^{-1})	
Crop rotation	No tillage	Conventional tillage
Chickpea	1808a	1356b
Sunflower	5399a	4762a
Faba bean	2269a	1559b
Wheat	3565a	2823b

Table 4. Graind yield and above-ground residues (wheat in 2008) influenced by rotation (WF: wheat-fallow; WC: wheat-chickpea; WS: wheat-sunflower; WFb: wheat-faba bean; WW: wheat-wheat), tillage system (NT: no tillage; CT: conventional tillage) and N rate

Rotations	Grain yield (kg ha^{-1})					Above-ground residues (kg ha^{-1})				
	Tillage		N rate (kg ha^{-1})			Tillage		N rate (kg ha^{-1})		
	NT	CT	0	50	150	NT	CT	0	50	150
WF	4571b	5216a	4316b	5068a	5297a	7048a	8427a	7181a	7591a	8439a
WC	3482a	2463b	2411b	3311a	3196a	5004a	3974a	3377b	4802a	5288a
WS	2066a	1859a	1140c	2128b	2619a	3045a	3026a	1716c	3120b	4277a
WFb	4698a	3926b	4157a	4443a	4335b	7061a	6076a	6158a	6746a	6802a
WW	3748a	3296a	3065b	3754a	3747a	5591a	5220a	4950a	5504a	5763a

Means followed by the same letter for tillage system and N rate are not significantly different at $P < 0.05$ according to LSD within each rotation.

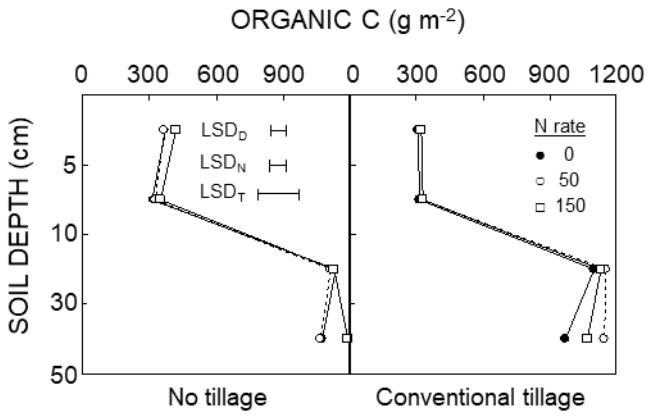

Fig. 1 Organic carbon influenced by tillage system (NT, no-tillage; CT, conventional tillage) and nitrogen for different soil depths. Horizontal bars represent LSD for comparison: LSDT, different level of tillage; LSDN, the same level of tillage; LSDD, the same level of tillage and nitrogen.

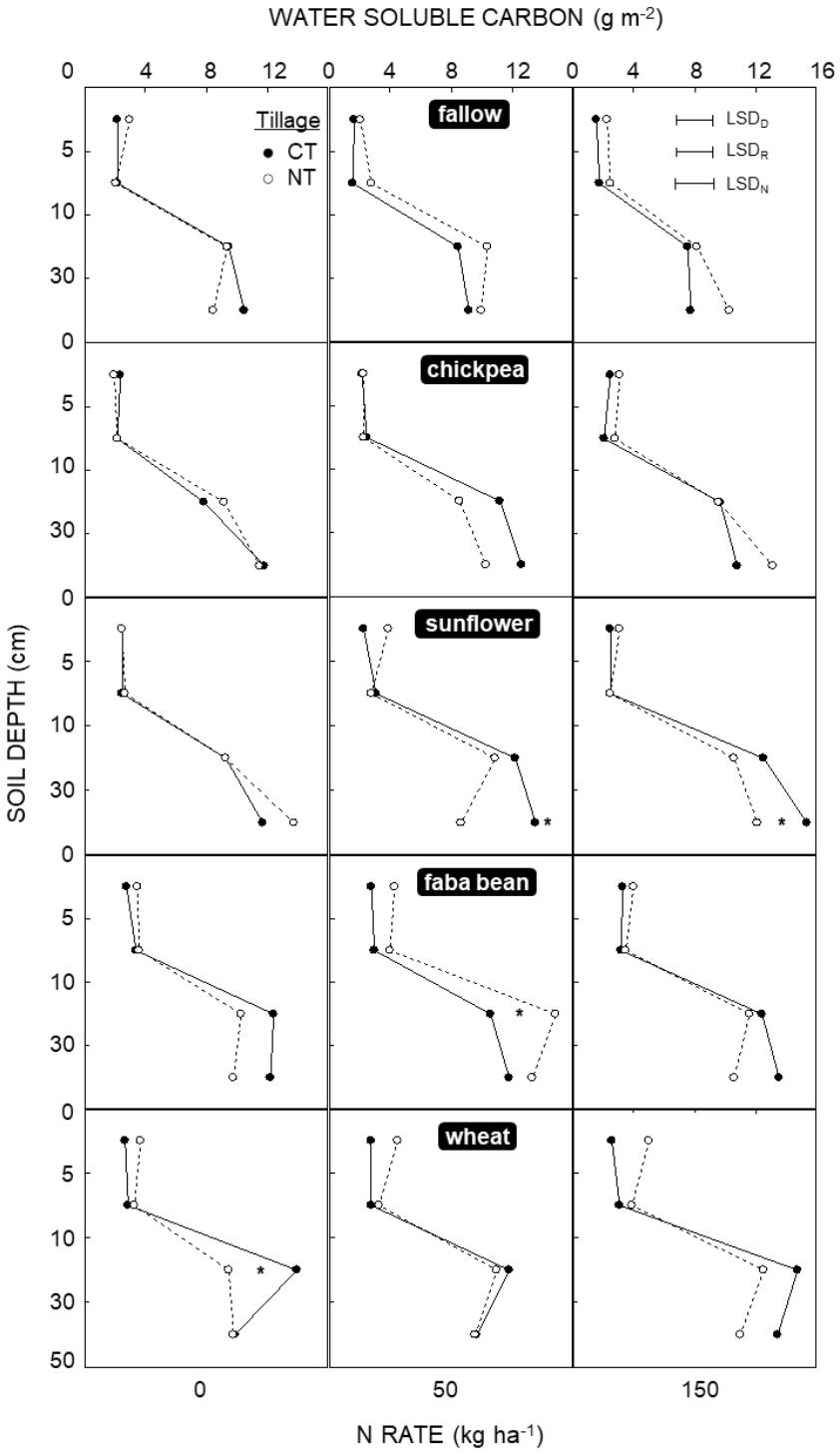

Fig. 2 Water soluble carbon influenced by tillage system (NT, no-tillage; CT, conventional tillage) and rotation for different soil depths and N rates. The asterisk (*) represents significant differences between tillage systems. Horizontal bars represent LSD for comparison: LSDR, the same level of tillage; LSDD, the same level of tillage and rotation.

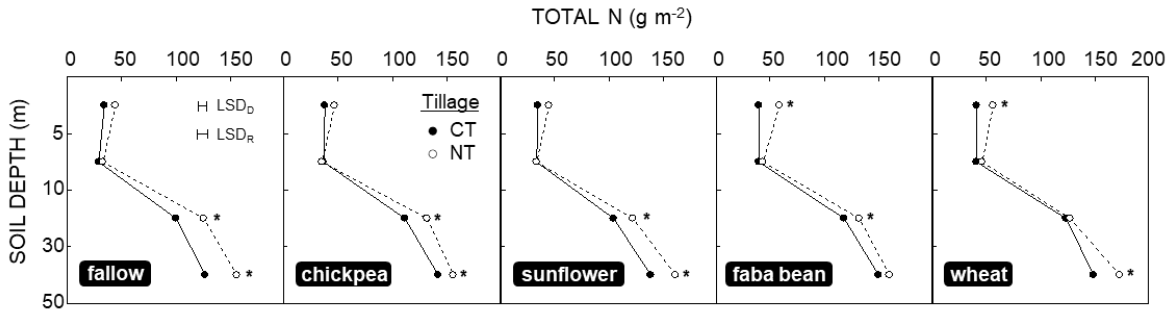

Fig. 3 Total nitrogen influenced by tillage system (NT, no-tillage; CT, conventional tillage) and rotation for different soil depths. The asterisk (*) represents significant differences between tillage systems. Horizontal bars represent LSD for comparison: LSDR, the same level of tillage; LSDD, the same level of tillage and rotation.

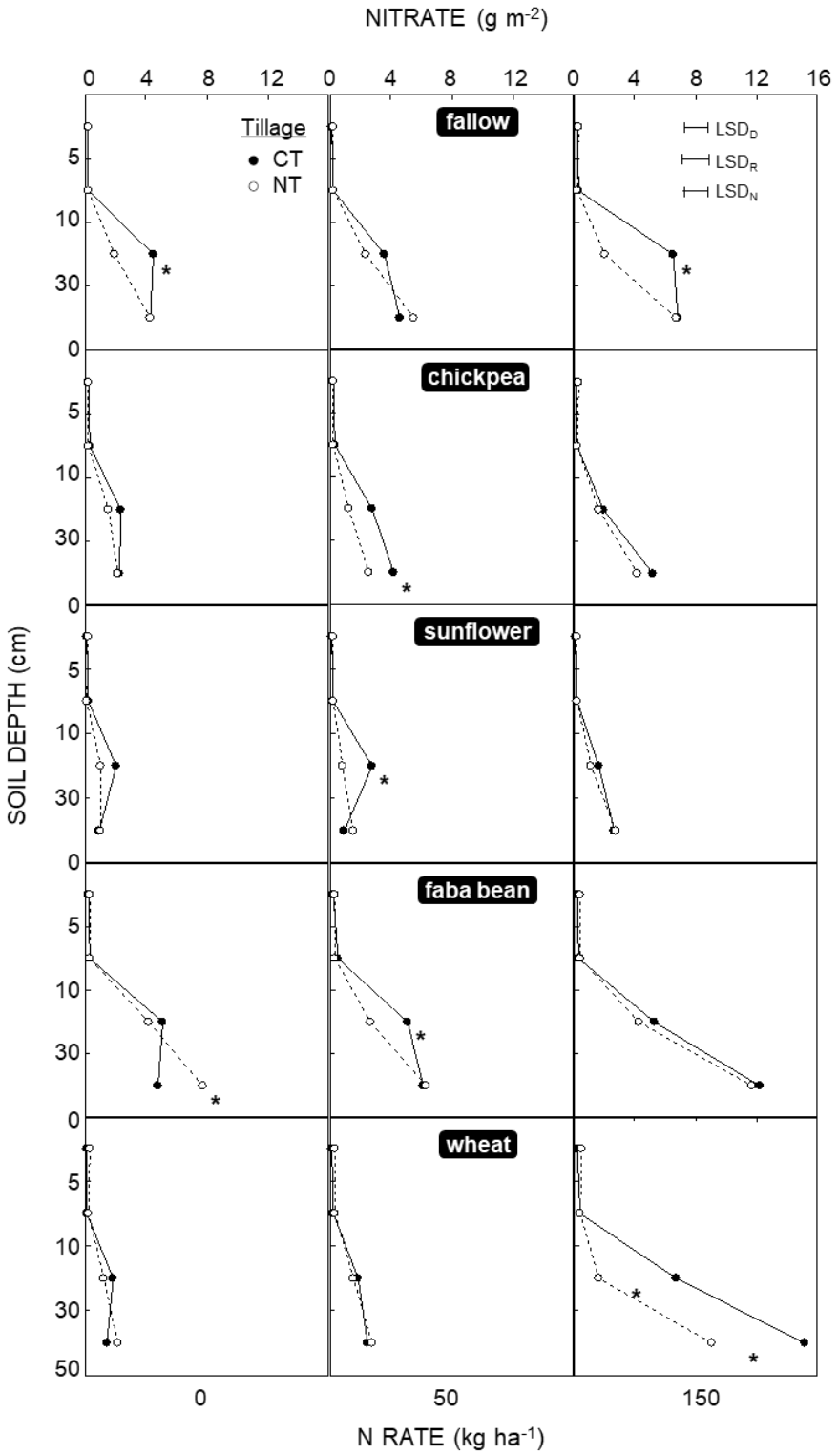

Fig. 4 Nitrate influenced by tillage system (NT, no-tillage; CT, conventional tillage) and rotation for different soil depths and N rates. The asterisk (*) represents significant differences between tillage systems. Horizontal bars represent LSD for comparison: LSDR, the same level of tillage; LSDN, the same level of tillage and rotation; LSDD, the same level of tillage, rotation and nitrogen.

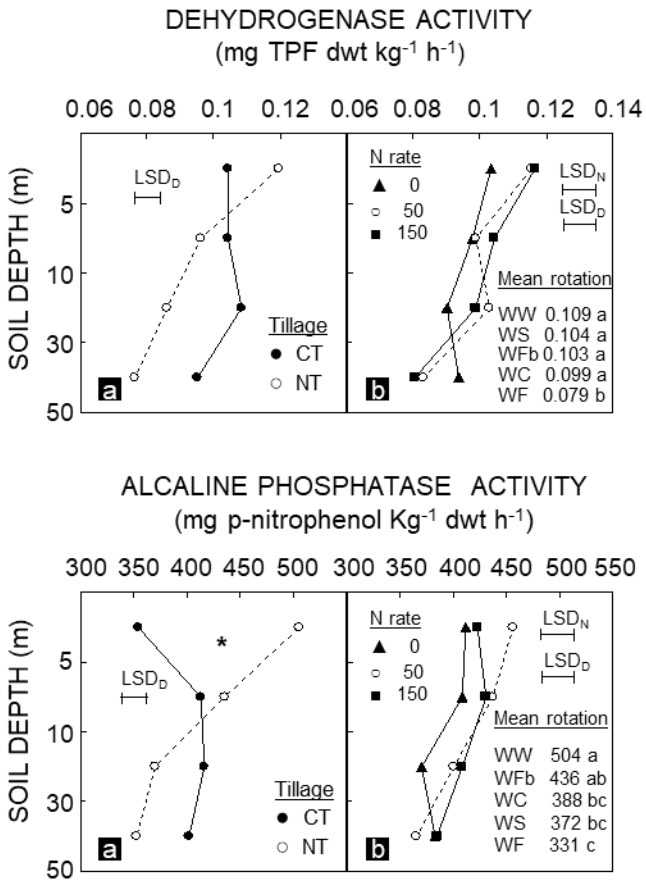

Fig. 5 Dehydrogenase an alcaline phospatase activity influenced by a) tillage system (NT, no-tillage; CT, conventional tillage) for different soil depths. Horizontal bar represent LSD for comparison: LSDD, the same level of tillage; b) N rate for different soil depths. Horizontal bars represent LSD for comparison: LSDN, different level of N rate; LSDD, the same level of N rate.

Mean rotation: for each tillage system, means followed by the same letters are not significantly different at $p<0.05$ according to LSD (WW, continuous wheat; WS, wheat-sunflower; WFb, wheat-faba bean; WC, wheat-chickpea; WF, wheat-fallow).

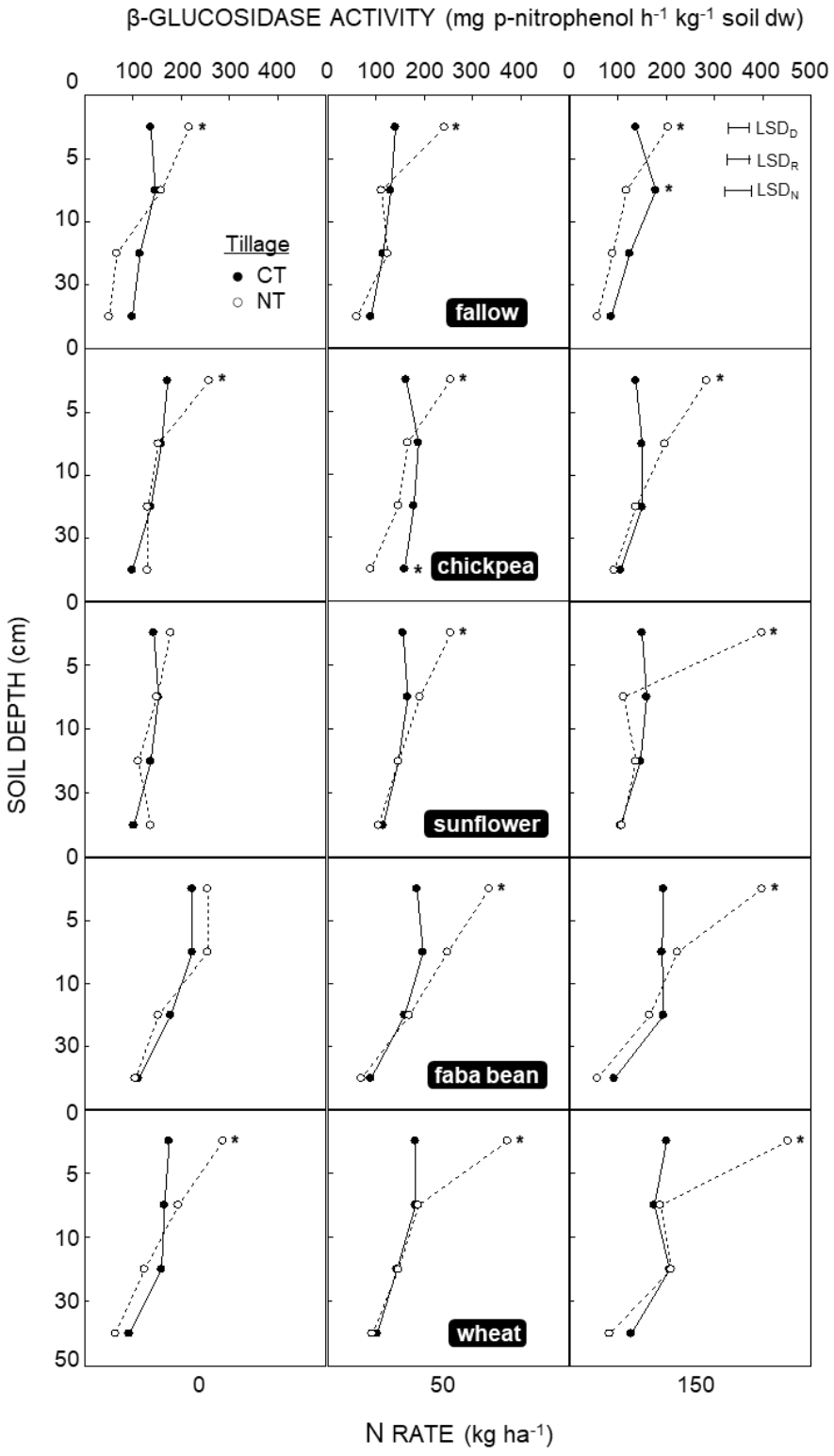

Fig. 6 β-glucosidase activity influenced by tillage system (NT, no-tillage; CT, conventional tillage) and rotation for different soil depths and N rates. The asterisk (*) represents significant differences between tillage systems. Horizontal bars represent LSD for comparison: LSDR, the same level of tillage; LSDN, the same level of tillage and rotation; LSDD, the same level of tillage, rotation and nitrogen.