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Long-term effect of tillage, nitrogen fertilization and cover crops on soil organic carbon and total nitrogen content

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ABSTRACT

No-tillage, N fertilization and cover crops are known to play an important role in conserving or increasing SOC and STN but the effects of their interactions are less known.

In order to evaluate the single and combined effects of these techniques on SOC and STN content under Mediterranean climate, a long term experiment started in 1993 on a loam soil (Typic Xerofluvent) in Central Italy.

The experimental variants are: conventional tillage (CT) and no-tillage (NT), four N fertilization rates (N0, N1, N2 and N3) and four soil cover crop (CC) types (C – no cover crop; NL – non-legume CC; LNL – low nitrogen supply legume CC, and HNL – high nitrogen supply legume CC).

The nitrogen fertilization rates (N0, N1, N2 and N3) were: 0, 100, 200, 300 kg N ha^{-1} for maize (*Zea mays*, L.); 0, 60, 120,180 kg N a^{-1} for durum wheat (*Triticum durum* Desf.); 0, 50, 100, 150 kg N ha^{-1} for sunflower (*Helianthus annuus* L.).

From 1993 to 2008, under the NT system the SOC and STN content in the top 30 cm soil depth increased by 0.61 and 0.04 Mg ha^{-1} $year^{-1}$ respectively. In the same period, the SOC and STN content under the CT system decreased by a rate of 0.06 and 0.04 Mg ha^{-1} $year^{-1}$ respectively.

During the experimental period, N1, N2 and N3 increased the SOC content in the 0–30 cm soil layer at a rate of 0.14, 0.45 and 0.49 Mg ha^{-1} $year^{-1}$. Only the higher N fertilization levels (N2 and N3) increased STN content, at a rate of 0.03 and 0.05 Mg ha^{-1} $year^{-1}$.

NL, LNL and HNL cover crops increased SOC content by 0.17, 0.41 and 0.43 Mg C ha^{-1} $year^{-1}$ and −0.01, +0.01 and +0.02 Mg N ha^{-1} $year^{-1}$.

Significant interactions among treatments were evident only in the case of the N fertilization by tillage system interaction on SOC and STN concentration in the 0–10 cm soil depth in 2008.

The observed SOC and STN variations were correlated to C returned to the soil as crop residues, aboveground cover crop biomass and weeds (C input).

We conclude that, under our Mediterranean climate, it is easier to conserve or increase SOC and STN by adopting NT than CT. To reach this objective, the CT system requires higher N fertilization rates and introduction of highly productive cover crops.

1. Introduction

Changes in soil organic carbon (SOC) and soil total nitrogen (STN) are largely responsible for the variations of soil physical, chemical and biological properties (Monaco et al., 2008) and have great influence on crop productivity and environmental quality (Sainju et al., 2008). No-tillage (NT) cropping systems are known to provide many benefits to soils mainly due to increased SOC and STN (Alvarez and Steinbach, 2009; Grandy et al., 2006). NT systems maintain and/or increase SOC and STN both by reducing their loss through decomposition and erosion and by sequestering C and N into the soil (Sainju et al., 2002). Previous studies have reported higher concentrations of SOC and STN under NT systems compared with conventional tillage (CT) systems in various top-soil layers (Mahboubi et al., 1993; Alvarez et al., 1995; Sainju et al., 2010) whereas the results obtained for the subsoil are less definite (Dalal, 1989; Baker et al., 2007; Blanco-Canqui and Lal, 2008).

Nitrogen fertilization can influence soil C and N inputs through increased biomass production. Higher biomass return to the soil can increase SOC and STN (Gregorich et al., 1996; Halvorson et al., 1999; Omay et al., 1997; Sainju et al., 2002; Sainju et al., 2003). On the other hand, increased N application to soil can stimulate mineralization, thus potentially increasing N losses through leaching and gaseous emissions (Malhi and Lemke, 2007).

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Cover crops are beneficial in agro-ecosystems in many ways (Dinesh, 2004; Kuo et al., 1997; McCracken et al., 1994; McVay et al., 1989; Reicosky and Forcella, 1998). Besides prevention of soil erosion, cover crops have gained attention also for other environmental services they provide, such as the preservation or increase in SOC and STN (Kuo et al., 1997; Sainju et al., 2003; Villamil et al., 2006), which reduces their losses to the environment and enhance the soil C sink capacity (Reicosky and Forcella, 1998). Furthermore, no-tillage, N fertilization and cover crops can interact positively to improve SOC and STN. While NT conserves or maintains SOC and STN by reducing the rate of residue decomposition and aggregate degradation and, in some cropping systems, by increasing productivity and thus C input (Franzluebbers et al., 1995; Havlin et al., 1990), N fertilization and cover crops can increase soil C input by increasing biomass production and residue addition to the soil (Kuo et al., 1997; McVay et al., 1989; Sainju et al., 2002). As the changes in SOC and STN occur slowly, long-term studies are necessary to determine the magnitude of the effects of management factors on SOC and STN. The combined long-term effects of tillage systems, N fertilization and cover crops on SOC and STN are not well-known, particularly under Mediterranean conditions. To evaluate the effects of these management techniques on soil characteristics and crop productivity, a long-term experiment was established in Central Italy in 1993. This paper examines the single and combined effects of tillage systems, N fertilization rates and cover crops on SOC along with STN and total C input to the soil (crop residues, weed and cover crops biomass). We hypothesized that the NT system, coupled with the use of cover crops and an optimum level of N fertilization would increase the content of C and N in the soil.

2. Materials and methods

2.1. Study site, treatments and experimental design

A long-term experiment was established in 1993 at the Interdepartmental Centre for Agro-Environmental Research (CIRAA) 'Enrico Avanzi' of the University of Pisa, Central Italy (Lat. 43°40′ N; Long. 10°19′ E) to study the effects of tillage system, N fertilization and cover crops on soil quality and crop productivity. The experimental field has a Typic Xerofluvent (Soil Survey Staff, 2010) loam soil (443, 402 and 155 g 1000 g^{-1} of sand, silt and clay respectively and pH 8.2 in the 0–30 cm soil layer).

Two tillage systems, four N fertilization rates and four soil cover types were factorially combined in a split-split-plot design with four replications, giving a total of 128 sub-sub-plots of 21 m length and 11 m width each. The tillage systems (main plots) were: conventional tillage (CT), i.e. mouldboard ploughing of the cover crops down to 30–35 cm depth, and no-tillage (NT), i.e. surface mulching of cover crop residues. The nitrogen fertilization rates (sub-plots: no nitrogen, N0; low nitrogen, N1; medium nitrogen, N2; and high nitrogen, N3) differed upon the main crop in rotation (0, 100, 200 and 300 kg N ha^{-1} for maize (*Zea mays* L.); 0, 60, 120 and 180 kg N ha^{-1} for durum wheat (*Triticum durum* Desf.); and 0, 50, 100, and 150 kg N ha^{-1} for sunflower (*Helianthus annuus* L.).

The cover types treatments (sub-sub-plots) consisted of three cover crops plus a control (C) without cover crop (i.e. residues of the previous main crop plus weeds). The three types of cover crops included in the systems were: a non-legume cover crop and two species of legume cover crop. Since the experiment focused more on the system effect than on the effect of individual cover crops, the design allowed freedom to change the cover crop species in order to optimize the efficiency of the systems in terms of nutrient cycling and soil fertility conservation. Therefore, the cover crop species were changed periodically to include the best cover crops for the experimental site conditions based on our experience (Mazzoncini et al., 1997; Antichi et al., 2008). Until 2002, the cover crops of the experimental site were: rye (*Secale cereale* L.), crimson clover (*Trifolium incarnatum* L.) and subterranean clover (*Trifolium subterraneum* L.). In 2003, rye was substituted by brown mustard (*Brassica juncea* L.) and subterranean clover by a 50:50% mixture of hairy vetch (*Vicia villosa* Roth) and rye. In 2005, this mixture was replaced by hairy vetch alone and crimson clover was substituted by squarrosum clover (*Trifolium squarrosum* L.). These species have showed different biomass productivity and different nitrogen concentration in the above-ground biomass during the experimental period; on average, 43.5 Mg d.m. ha^{-1} and 12.6 g of N kg^{-1} d.m., for rye and brown mustard; 36.0 Mg d.m. ha^{-1} and 24.2 g of N kg^{-1} d. m. for crimson and squarrosum clover and 42.4 Mg d.m. ha^{-1} and 25.9 g of N kg^{-1} d.m. for subterranean clover and hairy vetch. As a consequence, the average N accumulation potential in the above ground biomass was: 55, 87 and 110 kg of N ha^{-1} for rye and brown mustard, crimson and squarrosum clover and for subterranean clover and hairy vetch respectively. On this basis, the cover types treatments have been identified as: a control without cover crop (C); a non-legume cover crop (NL) with a low N accumulation potential in the above ground biomass (rye and brown mustard); a low nitrogen supply legume cover crop (LNL) with a medium N accumulation potential in the above ground biomass (crimson and squarrosum clover), and a high nitrogen supply legume cover crop (HNL) with a high N accumulation potential in the above ground biomass (subterranean clover and hairy vetch).

2.2. Crop rotation and management

The study was based on a maize (*Zea mays* L.) continuous crop from 1994 until 1998. In 1998 the cropping system was changed into a two-year crop rotation with maize in 1998 and durum wheat (*Triticum durum* Desf.) in 1999, continued for some years. Since 2005, the experiment is hosting a four-year crop rotation of durum wheat–maize–durum wheat–sunflower (Table 1). Within crop rotations, cover crops were always cultivated in winter, when the soil is generally bare (between maize and maize, wheat and maize and wheat and sunflower) (Table 1). Cover crops were sown manually in autumn after harvesting maize until 1997 and wheat (from 1999 onwards) (Table 1). In the next spring cover crops were killed off at the early flowering stage. In the CT system, cover crops were incorporated into the soil with a mouldboard plough to a depth of 30–35 cm after mechanical killing with a disk harrow or a shredder. In the NT system, cover crops were sprayed with glyphosate, flailed and retained on soil surface as dead mulch. The main crops in the CT and NT systems were seeded with a common seed drill and a NT drill, respectively. All plots were fertilised before main crop sowing with 20 kg ha^{-1} of P (as superphosphate) and 42 kg ha^{-1} of K (as potassium sulphate). N fertilizers were applied to the main crops according to the planned rate in just one (for maize and sunflower as urea) or two (for wheat as nitrate ammonium) split applications of 50% rate each. Cover crops were not fertilized. Weed control was based only on post-emergence herbicides application in the conventional tillage system while in the no-tillage system pre-sowing glyphosate was also applied.

2.3. Crop and weather measurements

In order to measure the amount of dry biomass returning to the soil each year in the different systems and then to estimate the C input to the soil, main crops above-ground residue, weed biomass and cover crop biomass were measured yearly at harvest time in the case of maize, sunflower and wheat, or immediately before their killing in the case of cover crops. Biomass was measured by collecting samples including weeds from two quadrates of 4 m^2

Table 1
Crop rotation timing. (CC = cover crop; M = maize; W = wheat; S = sunflower.)

Crop rotation	Years	CC killing time	Spring sowing time	Harvest time	Autumn sowing time
M monoculture	1993	–	–	–	December 22 – CC
	1994	April 21	May 9 – M	September 14 – M	October 11 – CC
	1995	April 18	May 10 – M	September 25 – M	October 6 – CC
	1996	May 6	June 21 – M	October 14 – M	November 4 – CC
	1997	May 2	May 26 – M	October 15 – M	November 17 – CC
	1998	May 11	June 1 – M	November 4 – M	December 3 – W
W–M 2-yr rotation	1999	–	July 13 – W	September 17 – CC	
	2000	April 13	May 10 – M	September 27 – M	December 28 – W
	2001	–	–	July 9 – W	October 2 – CC
	2002	May 13	May 22 – M	October 7 – M	December 30 – W
	2003	–	–	July 1 – W	September 26 – CC
	2004	April 21	May 11 – M	September 26 – M	December 14 – W
W–M–W–S 4-yr rotation	2005	–	–	July 14 – W	September 22 – CC
	2006	April 13	May 4 – M	October 16 – M	November 30 – W
	2007	–	–	June 28 – W	October 22 – CC
	2008	March 31	April 26 – S	September 21 – S	November 16 – W

within each plot in the case of maize and sunflower, and of 2 m^2 within each plot in the case of wheat and cover crops. The aboveground dry matter biomass returned to the soil was calculated by summing up the dry matter biomass of crop residues, cover crop and weed of each year over the experimental period (1994–2008, i.e. 15 years). The C input was estimated considering a concentration of 0.45 kg C kg^{-1} dry matter biomass according to our findings (data not shown) and other (Vanotti et al., 1997; Johnson et al., 2006). Monthly minimum (T min) and maximum (T max) air temperature and total rainfall were recorded at the CIRAA meteorological station and are presented in Table 2 as averages for the testing period (1994–2008) and for a longer term period (1983–2008).

2.4. Soil sampling and laboratory analyses

Soil sampling and analyses were carried out in 1993 (before the start of the experiment), in 1998 and 2008 at the end of September, immediately after harvest of maize in 1998 and sunflower in 2008, before soil tillage for the subsequent wheat crop. In each plot, two soil sub-samples were taken at each of two depths (0–10 and 10–30 cm) using a manual soil probe. The sub-samples were analysed separately in the laboratory and values were averaged plotwise for further statistical analysis. Upon arrival in the laboratory, the soil samples were dried, ground and sieved (2 mm mesh) for subsequent analysis. SOC concentration was determined by oxidising organic carbon with $K_2Cr_2O_7$ (1.5 N) in an acid environment with H_2SO_4 and subsequent reading with a spectrophotometer (Nelson and Sommers, 1982). STN concentration was determined with the Kjeldhal method (Bremner and Mulvaney, 1982). SOC and STN content in the whole layer (0–30 cm) were calculated using SOC and STN concentrations and soil bulk density values obtained by annual samplings. These were carried out by taking six soil cores in each plot at three different depths (0–10 cm, 10–20 cm and 20–30 cm) with a manual steel probe equipped with an inner metal cylinder of 22.287 cm^3 volume. Soil samples were collected at the end of spring (April–May), when soil moisture was intermediate between the wilting point and field capacity, in order to reduce deformation of soil cores during sampling. Soil cores were oven dried (105 °C for 24 h) and weighted; the soil bulk density was calculated as the ratio between the weight of oven-dried soil and the bulk volume of the soil.

2.5. Data analysis

Analysis of variance (ANOVA) for a split-split-plot design was performed using the CoStat Software (CoHort, 2002). Before analysis, the Bartlett test was performed to test the homogeneity of error variances. Differences between treatment means were compared using a Fisher's protected LSD test at $P < 0.05$ (Gomez and Gomez, 1984). The relationships between C input and SOC or STN variation between 1993 and 2008 were determined by linear regression analysis.

3. Results

3.1. Weather

At the experimental site, air temperatures and rainfall patterns indicate a typical Mediterranean climatic conditions characterized by a major rainfall pick in the autumn and a lower one in the spring and rainfall shortage in summer coupled with high temperatures reaching the maximum values (about 30 °C) in July and August.

Table 2
Monthly minimum and maximum air temperature (°C) and rainfall (mm) averaged over the 15 years of the research period (1994-2008) and over a longer-term period (1983–2008).

	Months												Mean temp. and total rainfall
	I	II	III	IV	V	VI	VII	VIII	IX	X	XI	XII	
Period of research (1994–2008)													
Min temp.	2.5	2.3	4.3	6.9	10.5	14.0	16.1	16.8	13.5	11.1	6.9	3.6	9.0
Max temp.	11.7	13,1	15.6	18.5	22.6	26.7	29.3	29.5	25.9	21.9	15.7	12.1	20.2
Rainfall	78	51	52	64	58	38	13	39	92	115	156	70	826
Longer-term period (1983–2008)													
Min temp.	2.4	2.4	4.4	7.3	10.6	13.9	16.5	17.0	14.2	11.2	6.6	3.8	9.2
Max temp.	11.8	13.0	15.5	18.3	22.3	26.1	29.5	29.7	26.1	21.7	15.8	12.3	20.2
Rainfall	66	48	54	75	62	44	23	47	93	124	142	86	864

Table 3
Significance of the effects of experimental factors and their interactions on above-ground dry biomass yield ($Mg\ ha^{-1}$) in the 1994–2008 period, soil organic carbon and soil total nitrogen concentration ($g\ kg^{-1}$) in 1998 and 2008, as resulting from analysis of variance. (ANOVA.)

Sources of variation	df	Aboveground biomass yield				Soil organic carbon concentration				Soil total nitrogen concentration			
		Over 15 years (1994–2008)				0–10 cm		10–30 cm		0–10 cm		10–30 cm	
		Main crop residues	Cover crops	Weed	Total	1998	2008	1998	2008	1998	2008	1998	2008
Tillage system (T)	1	***	*	**	**	**	***	NS	NS	**	***	NS	NS
N fertilization (N)	3	***	***	***	***	NS	**	NS	NS	*	***	NS	NS
N × T	3	*	NS	***	NS	NS	*	NS	NS	NS	***	NS	*
Cover type (C)	3	***	***	***	***	*	***	NS	**	NS	***	NS	*
C × T	3	**	**	*	*	NS	NS	NS	*	NS	NS	NS	NS
C × N	9	***	***	***	***	NS	NS	NS	NS	NS	NS	NS	NS
C × N × T	9	NS	*	**	NS	NS	NS	NS	NS	NS	NS	NS	NS

NS, not significant.
* Significant at $P < 0.05$.
** Significant at $P < 0.01$.
*** Significant at $P < 0.001$

The climatic conditions during the experimental period (1994–2008) did not substantially differ from the longer-term period (1983–2008) pattern (Table 2). The more appreciable differences may be ascribed to lower summer (in June, July and August) rainfall during the experimental period than in the longer-term one (Table 2).

3.2. Biomass returns to the systems

The tillage system caused significant differences in main crop residue, cover crop and weed biomass production, thus resulting in significant effects on the total above-ground biomass returned to the soil over 15 years (Table 3). Specifically, CT increased biomass yield of main crop residues and cover crops and decreased weed biomass as compared to the NT system (Table 4). On average, total above-ground biomass yield was 17% higher in CT than in NT (Table 4). Similarly, N fertilization resulted in significant differences in total above-ground biomass yield and in biomass yield of each component (Table 3), which increased with increasing rate of N fertilization (Table 4). On average, compared to N0, total biomass yield over 15 years was 24, 47 and 66% higher in N1, N2 and N3 respectively. The effect of cover types was also significant on each measured parameter (Table 3). Main crop residue yield was highest in HNL cover crop systems followed by LNL systems (Table 4). Cover crop biomass was significantly higher in HNL and NL than in LNL. In contrast, weed biomass was significantly higher in the system without cover crop followed by LNL and HNL. As a 15-year sum, total biomass yield was 23, 35 and 41% higher in NL, LNL and HNL respectively than in the system without cover crop. Averaged over N fertilization rates, the above-ground biomass yield was higher in CT than in NT and increased progressively from the system without cover crops to NL, LNL and HNL under both tillage systems (Fig. 1a). However, the degree of increment between LNL and HNL was higher in CT than in NT (Fig. 1a), leading to a significant tillage system by cover type interaction (Table 3). The N0 and N1 fertilization rates yielded higher total above-ground biomass with use of leguminous cover crops (LNL and HNL) than with use of a non-legume cover crop (NL) or without any cover crop (Fig. 1b). However, in the system with medium N rate (N2) biomass yield differed only slightly among the three cover crop types, only the difference between NL and HNL being significant. In contrast, the system with the highest nitrogen rate (N3) produced the highest total above-ground biomass when coupled with the NL cover crop (Fig. 1b).

Table 4
Effect of tillage system, N fertilization and cover type on total above-ground dry biomass yield and its components (main crop residues, cover crops and weeds).

Factors and treatment	Total dry biomass yield ($Mg\ ha^{-1}$) over 15 years (1994-2008)[†]			
	Main crop residues	Cover crops	Weeds	Total
Tillage system				
CT	71.5 a	32.1 a	12.3 b	115.9 a
NT	49.1 b	28.9 b	20.9 a	98.9 b
N fertilization				
N0	42.5 d	27.0 c	10.6 d	80.1 d
N1	56.9 c	27.8 c	14.7 c	99.4 c
N2	67.1 b	32.0 b	18.2 b	117.3 b
N3	74.8 a	35.2 a	22.9 a	132.9 a
Cover type				
C	57.9 c	-	28.2 a	86.1 d
NL	55.8 c	43.5 a	6.6 d	105.9 c
LNL	62.5 b	36.0 b	17.5 b	116.0 b
HNL	65.1 a	42.4 a	14.2 c	121.7 a

CT, conventional tillage; NT, no-tillage.
N0, N1, N2 and N3 are respectively no nitrogen, low nitrogen, medium nitrogen and high nitrogen fertilization rate.
C, no cover crop; NL, non-legume cover crop; LNL, low nitrogen supply legume cover crop; HNL, high nitrogen supply legume cover crop. See Section 2 for further details.
† Within each factor, means in the same column followed by different letters are significantly different at $P < 0.05$ (LSD test).

3.3. Soil bulk density

The NT system showed a significantly lower soil bulk density than the CT system in the upper 10 cm, but the contrary was observed in the deeper soil layers (Table 5). Nitrogen fertilization effects on soil bulk density were not significant in all soil depths, likewise the effects of the interactions among treatments (Table 5). HNL cover crops led to a significant decrease in bulk density in the 10–20 cm depth compared to NL.

3.4. Soil organic carbon

The effect of tillage system on SOC concentration was significant in both 1998 and 2008 in the surface (0–10 cm) soil layer but not in the deeper (10–30 cm) layer (Table 3).

SOC concentration in the upper soil layer was 29% and 44% higher in NT than in CT in 1998 and 2008, respectively (Table 6).

Table 7 shows changes across time in SOC content in the 0–30 cm soil layer. Between 1993 and 2008 SOC content increased by 21% in NT, corresponding to a gain of 0.61 $Mg\ SOC\ ha^{-1}\ year^{-1}$ whereas it decreased by 2% in CT, corresponding to a loss of 0.06 $Mg\ SOC\ ha^{-1}\ year^{-1}$ (Table 7).

The effect of N fertilization on SOC concentration was significant only in 2008 in the 0–10 cm layer (Table 3), indicating

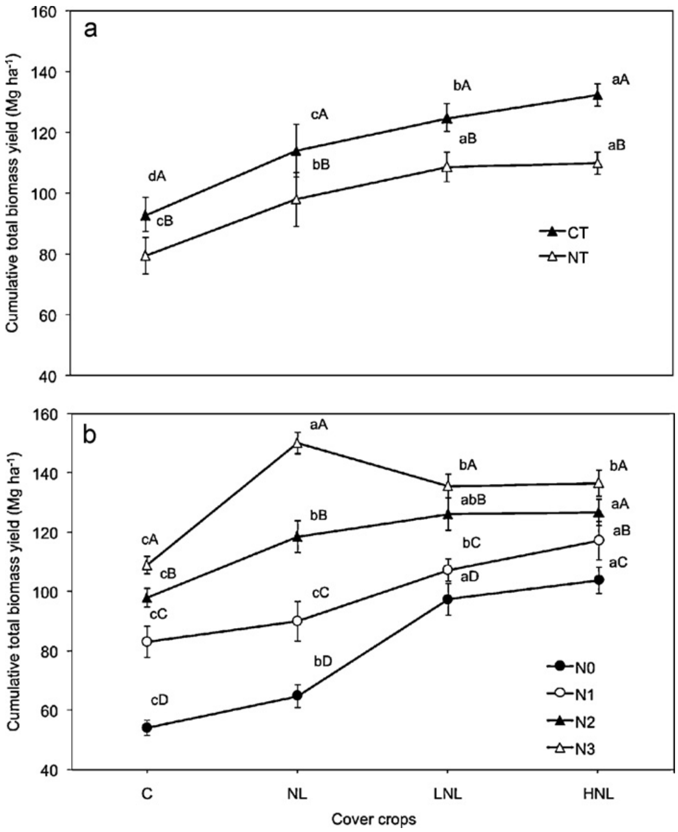

Fig. 1. Total above-ground dry biomass yield under two tillage systems averaged over N-fertilization (a) and four N fertilization rates averaged over tillage systems (b) as affected by different cover types. C = no cover crop, NL = non-legume cover crop, LNL = low nitrogen supply legume cover crop, HNL = high nitrogen supply legume cover crop. Means within the same tillage system in (a) and within the same N fertilizer rate in (b) followed by the same lower case letter are not significantly influenced by cover type. Within the same cover type, significant effects of tillage (a) or N fertilization (b) are shown by different upper case letters. P was always <0.05 (LSD test). Vertical bars show standard errors of the means.

that nitrogen supply to the system did not affect SOC concentration as fast as tillage system did. In 2008, SOC concentration in the 0–10 cm soil layer did not differ between the lower nitrogen dose (N1) and the unfertilized plots (N0) (Table 6). In contrast, medium (N2) and high (N3) nitrogen rates significantly increased SOC concentration as compared with N0 and N1 (Table 6).

Across time, different N fertilization rates produced different rates of change in SOC content in the 0–30 cm soil layer as compared with the unfertilized treatment (Table 7). Compared with the initial year (1993), the low nitrogen rate (N1) poorly increased SOC content as compared with the control (0.14 *vs* 0.03 Mg ha^{-1} year^{-1}), while the medium (N2) and high (N3) rates increased it by 0.45 and 0.49 Mg ha^{-1} year^{-1}, respectively (Table 7).

Cover types significantly affected SOC concentration in both soil layers in 2008, but only in the shallowest (0–10 cm) soil layer in 1998 (Table 3). In 1998, only HNL significantly increased SOC concentration in this layer (by 9%) as compared to the control (Table 6). Only 15 years after the onset of the long-term experiment (2008) the repeated cultivation of cover crops led to significant increases in SOC concentration in the 0–10 and 10–30 cm layers. In particular, SOC concentration was significantly higher in HNL and LNL than in the control without cover crop (Table 6). On average, the leguminous cover crops increased SOC concentration by 10% in the shallower layer and by 8% in the deeper layer as compared to the control. The SOC concentration increase by NL cover crops was lower (6% and 3% in the two layers respectively) and not significantly different from the control (Table 6).

Table 5
Significance of the effects of experimental factors and their interactions on soil bulk density in different soil layers (averaged over the 1994–2008 period), as resulting from analysis of variance. (ANOVA.)

Source of variation	0–10 cm	10–20 cm	20–30 cm	0–30 cm
Soil bulk density ANOVA				
Tillage system (T)	*	*	*	*
N fertilization (N)	NS	NS	NS	NS
Cover type (C)	NS	*	NS	NS
N × T	NS	NS	NS	NS
C × T	NS	NS	NS	NS
C × N	NS	NS	NS	NS
C × N × T	NS	NS	NS	NS
Soil bulk density (g cm^{-3})				
Treatments	0–10 cm	10–20 cm	20–30 cm	0–30 cm
CT	1.41 a	1.44 b	1.43 b	1.43 b
NT	1.37 b	1.51 a	1.54 a	1.47 a
N0	1.40	1.47	1.48	1.45
N1	1.40	1.47	1.49	1.45
N2	1.38	1.48	1.49	1.45
N3	1.39	1.47	1.49	1.45
C	1.40	1.47 ab	1.48	1.45
NL	1.39	1.49 a	1.48	1.46
LNL	1.40	1.47 ab	1.50	1.46
HNL	1.38	1.46 b	1.50	1.45

CT, conventional tillage; NT, no-tillage.
N0, N1, N2 and N3 are respectively no nitrogen, low nitrogen, medium nitrogen and high nitrogen fertilization rate.
C, no cover crop; NL, non-legume cover crop; LNL, low nitrogen supply legume cover crop; HNL, high nitrogen supply legume cover crop.
* Significant at $P < 0.05$; NS, non-significant. Within each factor, means in the same column followed by the same letter are not significantly different at $P < 0.05$ (LSD test).

The cover type effect on SOC content during the whole study period is shown in Table 7. The introduction of different cover crops increased SOC content at various rates across time as compared with the control. NL, LNL and HNL cover crop systems increased SOC content by 0.08, 0.32 and 0.34 Mg ha^{-1} year^{-1}, respectively, as compared with system without cover crop (Table 7).

Analysis of variance showed significant 'N fertilization by tillage system' and 'cover type by tillage system' interactions in 2008 (Table 3). The 'N fertilization by tillage system' interaction for SOC concentration was significant in the 0–10 cm layer. In this layer SOC concentration was not affected by nitrogen fertilization level

Table 6
Mean effects of tillage system, N fertilization and cover type on soil organic carbon concentration in the 0–10 cm and 10–30 cm soil layers in 1993, 1998 and 2008.

Factors and treatment	Soil organic carbon (g kg^{-1})†					
	0–10 cm			10–30 cm		
	1993	1998	2008	1993	1998	2008
Tillage system						
CT	11.0	10.1 b	10.8 b	10.8	10.6	10.6
NT	11.1	13.0 a	15.5 a	9.8	9.7	10.2
N fertilization						
N0	11.6	11.3	12.6 b	11.0	9.8	10.3
N1	11.1	11.3	12.5 b	10.2	10.1	9.9
N2	10.7	11.4	13.6 a	10.2	10.5	10.7
N3	10.8	12.0	14.0 a	10.0	10.1	10.6
Cover type						
C	11.2	11.0 b	12.4 c	10.3	10.2	9.9 c
NL	11.3	11.7 ab	13.1 bc	10.5	10.1	10.2 bc
LNL	10.8	11.4 ab	13.5 ab	10.2	10.1	10.6 ab
HNL	10.9	12.0 a	13.7 a	10.2	10.2	10.8 a

N0, N1, N2 and N3 are respectively no nitrogen, low nitrogen, medium nitrogen and high nitrogen fertilization rates.
C, no cover crop; NL, non-legume cover crop; LNL, low nitrogen supply legume cover crop; HNL, high nitrogen supply legume cover crop.
CT, conventional tillage; NT, no-tillage.
† Within each factor, means in the same column followed by the same letters are not significantly different at $P < 0.05$ (LSD test).

Table 7
Mean effects of tillage system, N fertilization and cover type on C input, soil organic carbon (SOC) and soil total nitrogen (STN) content in the 0–30 cm soil layer in 1993 and 2008 and difference (Δ) between SOC and STN values in 1993 and 2008.

Factors and treatments	Total biomass	C input	SOC ($Mg\,ha^{-1}$)[†]				STN ($Mg\,ha^{-1}$)[†]			
	($Mg\,ha^{-1}$)	($Mg\,ha^{-1}$)	1993	2008	Δ	$Mg\,ha^{-1}\,y^{-1}$	1993	2008	Δ	$Mg\,ha^{-1}\,y^{-1}$
Tillage system										
CT	115.86 a	52.14 a	46.35	45.43 b	−0.92 b	−0.06	5.55	5.01 b	−0.54 b	−0.04
NT	98.94 b	44.52 b	43.78	52.98 a	9.20 a	0.61	5.31	5.86 a	0.55 a	0.04
N fertilization										
N0	80.02 d	36.01 d	47.58	48.06 bc	0.48 b	0.03	5.80	5.12 b	−0.68 b	−0.05
N1	99.41 c	44.74 c	44.72	46.76 c	2.04 b	0.14	5.84	5.34 b	−0.50 b	−0.03
N2	117.36 b	52.81 b	44.19	50.89 ab	6.70 a	0.45	5.15	5.63 a	0.48 a	0.03
N3	132.82 a	59.77 a	43.76	51.08 a	7.32 a	0.49	4.93	5.67 a	0.74 a	0.05
Cover type										
C	86.03 d	38.71 d	45.21	46.60 c	1.39 b	0.09	5.43	5.14 c	−0.29 c	−0.02
NL	105.86 c	47.64 c	45.94	48.51 bc	2.57 b	0.17	5.52	5.40 b	−0.12 bc	−0.01
LNL	115.99 b	52.20 b	44.43	50.51 ab	6.08 a	0.41	5.36	5.50 ab	0.14 ab	0.01
HNL	121.73 a	54.78 a	44.67	51.18 a	6.51 a	0.43	5.41	5.71 a	0.30 a	0.02
Tillage system (T)	**	**	NS	**	**		NS	*	**	
N fertilization (N)	***	***	NS	*	**		NS	**	**	
T × N	NS	NS	NS	*	NS		NS	**	NS	
Cover type (C)	***	***	NS	**	**		NS	**	*	
T × C	*	*	NS	NS	NS		NS	NS	NS	
N × C	***	***	NS	NS	NS		NS	NS	NS	
T × N × C	NS	NS	NS	NS	NS		NS	NS	NS	

CT, conventional tillage; NT, no-tillage; NS, not significant.
N0, N1, N2 and N3 are respectively no nitrogen, low nitrogen, medium nitrogen and high nitrogen fertilization rates.
C, no cover crop; NL, non-legume cover crop; LNL, low nitrogen supply legume cover crop; HNL, high nitrogen supply legume cover crop.
* Significant at $P < 0.05$.
** Significant at $P < 0.01$.
*** Significant at $P < 0.001$
[†] Within each factor, means in the same column followed by the same letters are not significantly different at $P < 0.05$ (LSD test).

under the CT system, while it increased significantly at medium and high N fertilization rates under the NT system (Fig. 2). The 'cover type by tillage system' interaction for SOC was significant only in 10–30 cm soil layer in 2008 (Table 3). Averaged over N fertilization rates, NT and CT in this layer attained similar SOC concentrations with NL and LNL cover crops (Fig. 3); without cover crops, the SOC concentrations were lower but again similar in NT and CT, whereas with HNL cover crops the SOC concentration in CT exceeded that in NT (Fig. 3).

3.5. Soil total nitrogen

In 1998 STN concentration was affected by tillage systems and N fertilization only in the upper soil layer, while interactions among experimental factors were not observed (Table 3). Only in 2008, the main effects of the three factors (tillage system, N fertilization and cover type) and the interaction between tillage system and N fertilization became evident, being significant in the 0–10 cm layer (Table 3). In the deeper layer (10–30 cm) only the effects of cover type and the interaction between tillage system and N fertilization were significant (Table 3).

In 2008, the STN concentration in the 0–10 cm layer was 41% higher in NT than in CT (Table 8). Compared with 1993, under NT the STN concentration in the 0–10 cm soil layer increased by 8 and 24% in 1998 and 2008 respectively, whereas values in CT decreased by 11 and 13% in the same periods.

The changes in STN content in the 0–30 cm soil layer under CT and NT in the 1993–2008 period are shown in Table 7. On average,

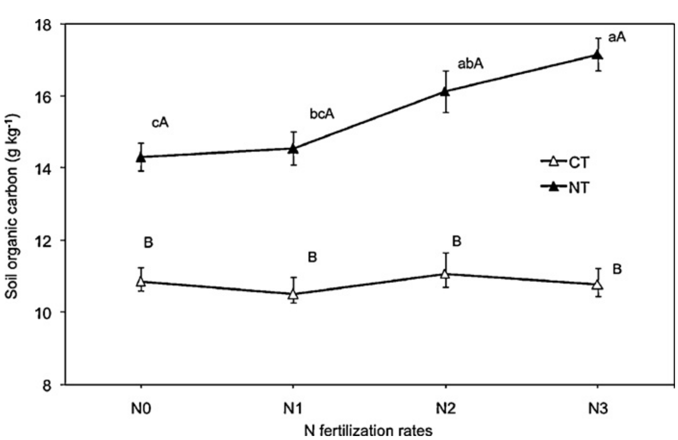

Fig. 2. Year 2008: soil organic carbon in the 0–10 cm soil depth under the conventional tillage (CT) and no-tillage (NT) systems as affected by different N fertilization rates. N0, N1, N2 and N3 are respectively no nitrogen, low nitrogen, medium nitrogen and high nitrogen fertilization rates. Means within the same tillage system followed by the same lower case letter are not significantly influenced by N fertilization. Within the same N fertilization rate, significant effects of tillage are shown by different upper case letters. P was always <0.05 (LSD test). Vertical bars show standard errors of the means.

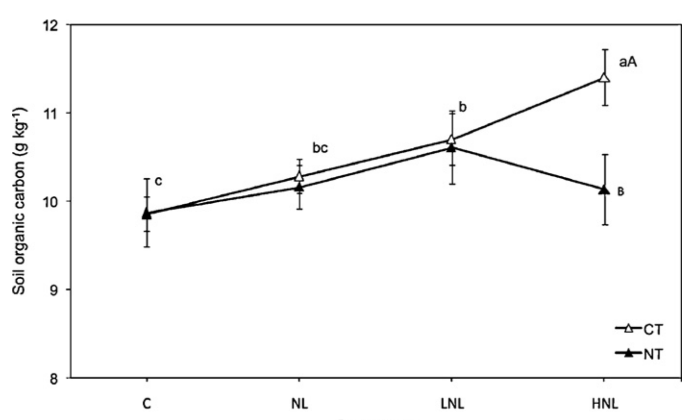

Fig. 3. Year 2008: soil organic carbon in the 10–30 cm soil layer under the conventional tillage (CT) and no-tillage (NT) systems as affected by different cover types. C = no cover crop, NL = non-legume cover crop, LNL = low nitrogen supply legume cover crop, HNL = high nitrogen supply legume cover crop. Means within the same tillage system followed by the same lower case letter are not significantly influenced by cover type. Within the same cover type, significant effects of tillage are shown by different upper case letters. P was always <0.05 (LSD test). Vertical bars show standard errors of the means.

Table 8
Mean effects of tillage system, N fertilization and cover type on soil total nitrogen concentration in the 0–10 cm and 10–30 cm soil layers in 1993, 1998 and 2008.

Factors and treatments	Soil total nitrogen (g kg $^{-1}$)[†]					
	0–10 cm			10–30 cm		
	1993	1998	2008	1993	1998	2008
Tillage system						
CT	1.37	1.22 b	1.19 b	1.26	1.26	1.17
NT	1.35	1.46 a	1.68 a	1.19	1.14	1.15
N fertilization						
N0	1.43	1.27 c	1.32 b	1.32	1.18	1.11
N1	1.48	1.32 bc	1.38 b	1.31	1.20	1.15
N2	1.29	1.36 ab	1.52 a	1.16	1.22	1.18
N3	1.25	1.41 a	1.51 a	1.10	1.22	1.19
Cover type						
C	1.35	1.30	1.34 c	1.23	1.20	1.10 b
NL	1.41	1.34	1.43 b	1.25	1.19	1.15 ab
LNL	1.31	1.35	1.46 ab	1.20	1.22	1.18 a
HNL	1.37	1.37	1.52 a	1.22	1.20	1.21 a

CT, conventional tillage; NT, no-tillage.
N0, N1, N2 and N3 are respectively no nitrogen, low nitrogen, medium nitrogen and high nitrogen fertilization rate.
C, no cover crop; NL, non-legume cover crop; LNL, low nitrogen supply legume cover crop; HNL, high nitrogen supply legume cover crop.
[†] Within each factor, means in the same column followed by the same letters are not significantly different at $P < 0.05$ (LSD test).

NT increased the STN content by 0.04 Mg ha^{-1} year^{-1} whereas CT decreased it by the same rate.

The effect of N fertilization on STN concentration was significant for the 0–10 cm soil layer in both 1998 and 2008 but was never so in the deeper layer (Table 3). In this layer, STN concentration was never influenced by the lower nitrogen dose (N1) as compared with the unfertilized treatment (N0), whereas the medium (N2) and high (N3) nitrogen rates increased significantly STN concentration by 7 and 11% in 1998 and by 15 and 14% in 2008 (Table 8). Averaged over soil depths, the rate of change in STN content over time differed upon N rates: N1 did not increase STN over time (Table 7) whereas N2 and N3 increased it by 0.08 and 0.10 Mg ha^{-1} year^{-1} respectively against the annual change in the unfertilized treatment (Table 7).

The effect of cover type on STN concentration was significant only in 2008 in both soil depths (Table 3). In the shallower soil layer, all cover crops significantly increased STN concentration as compared with the system without cover crop (Table 8). In particular, HNL increased STN significantly more than NL. In the 10–30 cm soil layer, only the systems with LNL and HNL cover crops showed higher STN concentration than the system without cover crop (Table 8). Table 7 also shows the effect of different cover types as relative to the control on STN content in the 0–30 cm soil layer during the study period. NL did not increase STN over time whereas LNL and HNL increased it by 0.03 and 0.04 Mg ha^{-1} year^{-1} respectively against the annual change in the system without cover crop (Table 7).

The 'tillage system by N-fertilization' interaction was significant in 2008 in both soil layers (0–10 cm and 10–30 cm) (Table 3). In the shallowest layer, STN concentrations increased with higher rates of N fertilization (N2 and N3) in the NT system but did not change significantly in the CT system (Fig. 4a); in the deeper layer, STN concentration showed a similar trend (Fig. 4b). All the other interactions ($C \times T$, $C \times N$ and $C \times N \times T$) were not significant (Table 3).

4. Discussion

Under our experimental conditions, CT increased significantly crop residues and cover crop biomass production providing a higher C input to the soil (Table 4), even if weed biomass

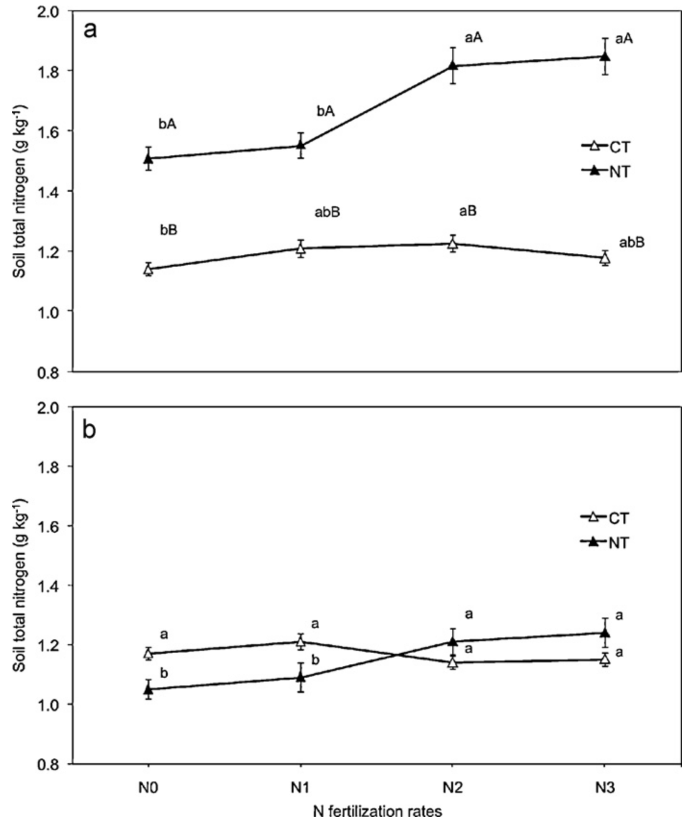

Fig. 4. Year 2008: soil total nitrogen in the 0–10 cm (a) and 10–30 cm (b) soil depths under the conventional tillage (CT) and no-tillage (NT) systems as affected by different N fertilization rates. N0, N1, N2 and N3 are respectively no nitrogen, low nitrogen, medium nitrogen and high nitrogen fertilization rates. Means within the same tillage system followed by the same lower case letter are not significantly influenced by N fertilization. Within the same N fertilization rate, significant effects of tillage are shown by upper case letters. P was always <0.05 (LSD test). Vertical bars show standard errors of the means.

production was lower. The beneficial effect of CT on crop productivity has been observed in the same environment and attributed to better physical and hydrological soil conditions and weed control compared to NT (Mazzoncini et al., 2008).

Despite the lower total C input under NT, higher content of SOC and STN has been observed under NT than CT after 15 years (Table 7) which is in agreement with other studies (Alvarez et al., 1995; Alvarez and Steinbach, 2009; Halvorson et al., 2002; Mahboubi et al., 1993; Sainju et al., 2002). This shows the importance of studying SOC and STN changes due to agricultural management practices in long term experiments. In fact, it is difficult to monitor SOC and STN changes because of their large pool size in the soil and, from the methodological point of view, because of spatial variability (Salinas-Garcia et al., 1997).

In our agro-environmental conditions, changing from CT to NT can sequester 0.61 Mg C ha^{-1} y^{-1} after 15 years; in contrast 0.06 Mg C ha^{-1} y^{-1} can be lost using CT corresponding to a difference of 0.67 Mg C ha^{-1} y^{-1} (Table 7). Regarding the capacity of NT to increase SOC content, our data are similar to those calculated by West and Post (2002) that indicated an average SOC increment of 0.57 Mg C ha^{-1} y^{-1} in converting arable systems in NT systems at the global scale. The review of So et al. (2001) also indicates that SOC accumulation rate under NT in Australian soils may varies from 0.11 to 1.57 Mg C ha^{-1} y^{-1}. The latter value has been indicated by Franzluebbers et al. (2009) as the C sequestration rate under NT in Alabama. For European soils, Freibauer et al. (2004) indicated a potential SOC sequestration of 0 to 0.80 Mg C ha^{-1} y^{-1} changing from CT to NT. The recent review of Franzluebbers (2010) indicates a SOC sequestration rate of 0.55 Mg C ha^{-1} y^{-1} for U.S. agricultural systems including cover crops (like ours) at global scale. This author has also calculated the yearly difference in SOC accumulation rate between CT and NT, which was 0.66 and 0.59 Mg C ha^{-1} y^{-1} in Alabama and Georgia respectively, two regions where rainfall and air temperature annual patterns are similar to ours. In any case, the authors of these reviews underlined that SOC accumulation under NT is site-specific and hence has to be evaluated locally by taking into account climatic conditions, soil characteristics and C returns to the soil (C input) as result of crop productivity and system management (crop rotation including or not cover crops, bare fallow, N fertilization, manure application, etc.). In Europe these conditions vary considerably between the continental regions (France, Germany, England, Switzerland, Denmark, Holland, inland Spain) and the Mediterranean ones (Portugal, coastal Spain, Italy, Greece). The latter have generally higher air temperature, less rainfall in summer, higher SOC mineralization rate and, as a consequence, crop productivity and SOC content are lower. In agreement with Franzluebbers (2010), CT replacement with NT may cause poor SOC increments under the drier climate of typical Mediterranean regions as well as under the colder climates of continental regions of Europe. In Italy, climate varies considerably between the North and South and hence NT application may lead to different results in terms of SOC conservation. In Sicily (South Italy), due to the low annual C input and high mineralization rate, Barbera et al. (2010) found no significant differences in SOC stock between NT and CT after 19 years. These authors estimated on average 0.39 Mg C ha^{-1} y^{-1}, as C requirement to conserve SOC equilibrium against an average C input of 0.36 Mg C ha^{-1} y^{-1}. On the contrary, in North-East Italy (Veneto), Borin et al. (1997) found that, after just 3 years, the difference in SOC content between NT and CT systems was 0.77 Mg C ha^{-1} y^{-1}. These findings indicate that under humid and temperate conditions NT can improve SOC (Alvarez, 2005).

Beside this, our results show that carbon input and SOC and STN are also affected by N fertilization, being positively related to N supply from N0 to N3 (Table 7). Many experiments have shown that N fertilization results in SOC increase over time not only by promoting crop biomass production but also by chemically stabilizing SOC (Paustian et al., 1992; Snyder et al., 2009). Generally, higher fertilization provokes higher SOC increments (Halvorson et al., 2002; Jagadamma et al., 2007; Reid, 2008; Sainju et al., 2010).

Alvarez, in his extensive review (2005), observed a positive effect of N fertilization on SOC content at global scale when residues were returned to the soil. In his study the effect of N fertilization was observed when crop residues were removed or burned. Others studies showed a positive effect on SOC accumulation in time when organic matter is added and N is supplied in both organic and inorganic forms (Paustian et al., 1992). These findings indicate that SOC accumulation is strictly related to soil C input. Using data coming from many long-term experiments, Alvarez (2005) estimated also that no significant SOC increments occur when the cumulative nitrogen rate applied did not exceed 1 Mg ha^{-1}. Above this value, SOC tended to increase by approximately 2 Mg C ha^{-1} for each 1 Mg N ha^{-1} applied, with respect to the unfertilized control. This estimate has been partially confirmed by our results: 1.2, 2.5 and 1.8 Mg C ha^{-1} per 1 Mg N ha^{-1} applied during the research period (1.25, 2.50 and 3.75 Mg N ha^{-1} applied with N1, N2 and N3 respectively).

It is important to observe that in our case SOC content increased only slightly over time under N0 and N1 (according to the lower amount of crop residue retained in the soil), while STN decreased appreciably (Table 7). The different temporal patterns of C and N dynamic could be explained by the N shortage provoked at the cropping system level by these N fertilization rates. In fact, considering that on average 100 to 120 kg N ha^{-1} y^{-1} were removed from the soil by the crops under N0 and N1 (data not shown) and taking into account N losses through leaching, and volatilization, the N requirements of these systems seem higher than their relative N inputs: only 0 and 83 kg N ha^{-1} y^{-1} for N0 and N1 respectively.

In our study, the positive effects of N fertilization on SOC and STN were mainly observed in the upper 10 cm of NT soil (Figs. 2 and 4). Blevins et al. (1977) and Salinas-Garcia et al. (1997) also showed that increased N fertilization resulted in higher SOC concentration in the surface soil of NT only. This is probably because the biomass returned to the soil under NT is subject to more natural processes (including biomass concentration on the soil surface and a poorer contact of biomass with the soil particles) able to reduce the mineralisation rate and to increase humification.

The introduction of cover crops in a rotation generally significantly increases SOM as reported by Smith et al. (1997), Drinkwater et al. (1998) and Lal (2004).

In our study, higher SOC and STN concentration and content were found in the systems with cover crops (Tables 6–8). These results confirm the importance of introducing winter cover crops in crop rotations for maintaining or increasing SOC and STN on loam soils even under the water demanding condition of a Mediterranean climate as also suggested by Paustian et al. (1997). Our findings are in agreement with previous ones from different climatic conditions: in Georgia (USA) under Orthic Luvisols (Sainju et al., 2002) and Rhodic Kandiudults (Sainju et al., 2003) and in Illinois under Aquic Argiudolls (Villamil et al., 2006). In our long term experiment, the highest increases in STN and SOC content has been achieved with legume winter cover crops (HNL and LNL) (Table 7). Others authors observed that including legume cover crops or legume pasture in a crop rotation may easily conserve or increase SOM and STN (Drinkwater et al., 1998; So et al., 2001). In our case, this may be related to the higher biomass yielding capacity of HNL and LNL cover crops system (cover crops + weeds) respect to NL and C systems. Similarly to the effect of N0 and N1 on SOC and STN accumulation, the soil without cover crop (C) and that with NL showed a SOC increase but a STN decrease over time (Table 7), and effect likely due to the lower N content in the biomass produced by these two systems, coupled with lack of N fixation. The low N quantity supplied by C and NL systems (53 and 62 kg N ha^{-1} y^{-1} respectively as average over year and N rates) proved to be inadequate to meet the N requirement of these systems, as indicated by crop N removals (95–120 kg N ha^{-1} y^{-1}) and by N leaching and volatilization losses, which have been estimated as 5–40 kg N ha^{-1} y^{-1} for our environment (Masoni, 2010).

Many authors indicated the existence of a strong relationship between soil C input and SOC and STN changes in the long term (Barbera et al., 2010; Buyanovsky and Wagner, 1998; Halvorson et al., 2002; Paustian et al., 1992). This relationship has been used to explain the positive effect of N fertilization, cover crop cultivation, crop rotation and reduced fallowing time on SOC and STN content through the increase in C input generated by crop residues, cover crop, weed and root biomass and rhizodepositions (Alvarez, 2005). Similarly, in our experiment the variations ($\pm$) in SOC and STN content across time were directly correlated, within each tillage system, to the total C supplied to the soil through the addition of crop residues, cover crop and weed biomass between 1998 and 2008 (Figs. 5 and 6). According to this relationship, the adoption of N2 and N3 fertilization rates and the introduction of cover crops in the system proved to be a good strategy to increase SOC and STN content in the 0–30 cm soil layer. However, soil C storage is not only determined by the C return to the soil but also by the balance between C input and C output represented essentially by SOC mineralization,

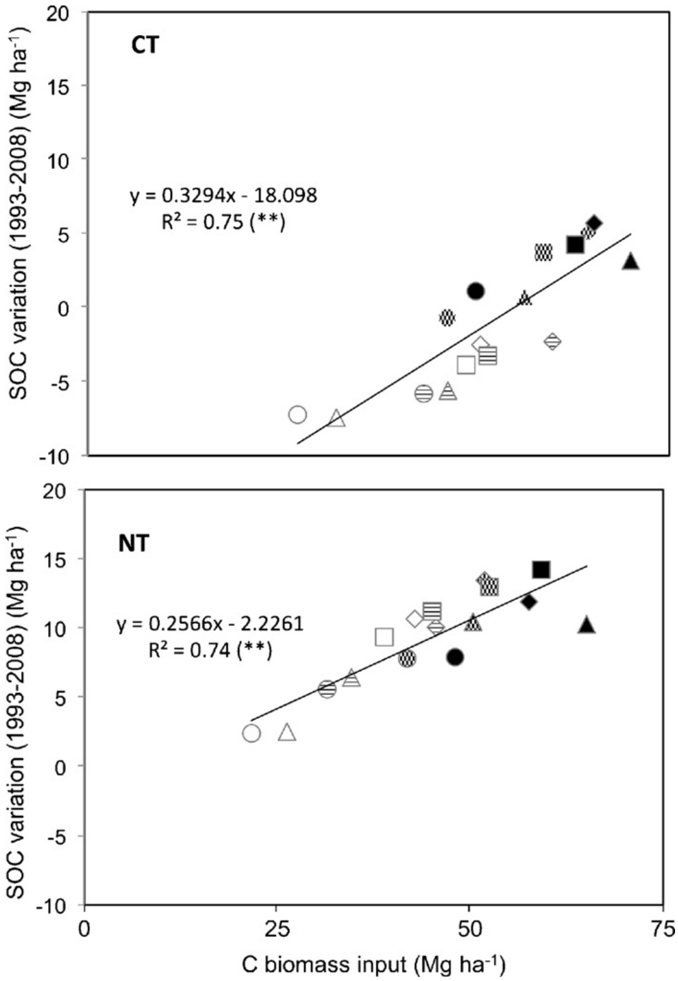

Fig. 5. Results of linear regression analysis of C biomass input on soil organic carbon (SOC) variation between 1993 and 2008 under the conventional (CT, down) and the no-tillage (NT, top) systems. Circles, triangles, squares and diamonds symbols represent no cover crop (C), non-legume cover crop (NL), low nitrogen supply legume cover crop (LNL) and high nitrogen supply legume cover crop (HNL), respectively. Empty, horizontally hatched, checked and full symbols represent no nitrogen (N0), low nitrogen (N1), medium nitrogen (N2) and high nitrogen (N3) fertilization rates, respectively. (**) significant at $P < 0.01$.

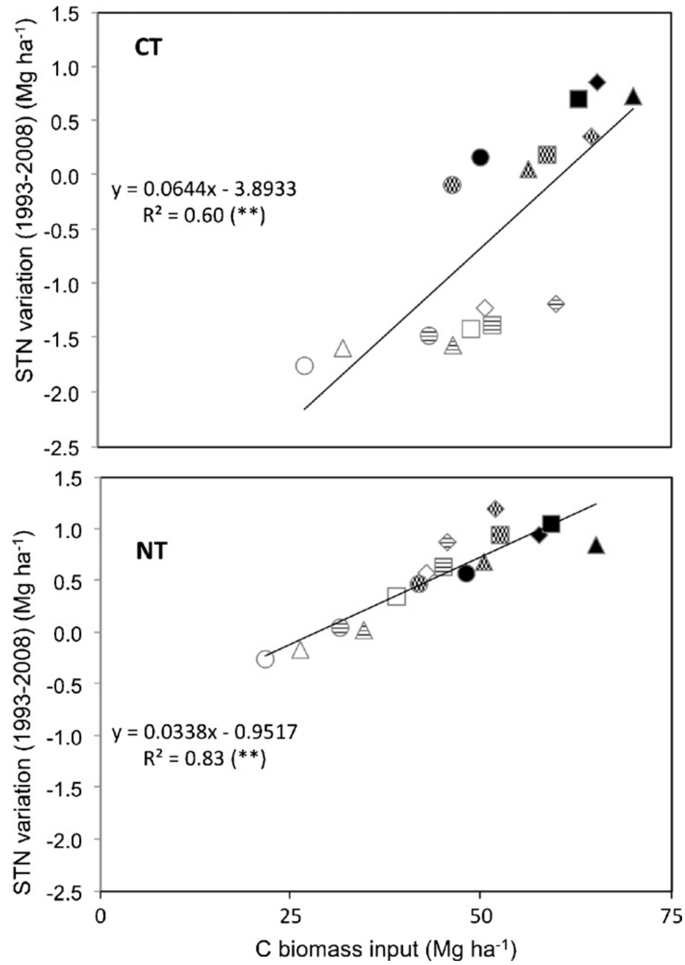

Fig. 6. Results of linear regression analysis of C biomass input on soil total nitrogen (STN) variation between 1993 and 2008 under the conventional (CT, down) and the no-tillage (NT, top) systems. Circles, triangles, squares and diamond symbols represent no cover crop (C), non-legume cover crop (NL), low nitrogen supply legume cover crop (LNL) and high nitrogen supply legume cover crop (HNL), respectively. Empty, horizontally hatched, checked and full symbols represent no nitrogen (N0), low nitrogen (N1), medium nitrogen (N2) and high nitrogen (N3) fertilization rates, respectively. (**) significant at $P < 0.01$.

SOC erosion, runoff and leaching (Sainju et al., 2010). In fact, despite the higher biomass production of main crop residues and cover crops observed under CT than NT (Tables 4 and 7), SOC conservation in CT was not ensured in our Mediterranean climatic conditions, likely because of the higher mineralization rate expected in the conventional tillage system. In our experimental conditions, to maintain the SOC equilibrium under CT it would be necessary to apply 54.9 Mg C ha^{-1} across 15 years (3.7 Mg C ha^{-1} year^{-1}), while under NT 8.7 Mg C ha^{-1} across 15 years (0.6 Mg C ha^{-1} year^{-1}) would be sufficient (Fig. 5). This means that under NT even the lowest C input, observed with the lower N fertilization rates (N0 and N1) and with NL and the system without cover crop, was able to conserve or slightly increase SOC content. In contrast, to meet the higher mineralisation rate of CT, it is necessary to increase C input by applying higher levels of N fertiliser (N2 and N3) and to cultivate highly productive cover crops (Fig. 5).

5. Conclusion

The aim of this research was to evaluate the single and combined effects of tillage system, N fertilization and cover type on the long-term dynamics of SOC and STN under a Mediterranean climate. Based on our results, we can conclude that the use of NT may reduce crop residue production and hence C input respect to CT. Nevertheless, NT is able to conserve SOC in the long run even without N fertilization and without introducing cover crops because of the lower soil organic matter mineralization rate than under CT. Significant increases in SOM and STN can be achieved under NT when medium-high and high (N2 and N3) N fertilization rates are applied to cash crop, and/or when LNL and HNL cover crops are introduced in the rotation. In contrast, under CT it was necessary to apply the medium-high N fertilization rate (N2) just to conserve SOC. Appreciable SOC increments can be obtained under CT with high N fertilization rate (N3) and cover crop (green manure) cultivation or with the use of LNL and HNL green manures coupled with N2. This means that the introduction of cover crops (as green manure) in CT systems may help maintain an adequate C input which compensates the higher soil mineralization rate.

As a whole, C and N accumulation in soil increases the sequestration of atmospheric C into the soil, thereby helping to mitigate the negative effects of cropping on climate change.

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