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Switching from conventional tillage to no-tillage: soil N availability, N uptake, ^{15}N fertilizer recovery, and grain yield of durum wheat

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Abstract

This 2-year study, performed in a typical Mediterranean environment on three soil types (two Inceptisols and one Vertisol), aimed to improve understanding of the factors that play a major role in determining crop response when soil management shifts from conventional tillage (CT) to no-tillage (NT). The effects of NT on the soil nitrogen (N) availability, N uptake, ^{15}N fertilizer recovery, and grain yield of durum wheat were evaluated in comparison to CT under five different N fertilization rates (0, 40, 80, 120, and 160 kg N ha^{-1}).

Compared to CT, NT negatively affected grain yield in one of the two years but only in the two Inceptisols. On average, a considerable grain yield advantage of CT over NT (approximately +0.6 Mg ha^{-1} of grain) was observed with no N fertilization. This benefit decreased progressively when N fertilizer rate increased to the point that at 120 kg ha^{-1} of N applied differences between CT and NT were negligible. The differences between the two tillage systems in both grain yield and N uptake were attributable more to differences in the native soil mineral N (that materialized already during the vegetative phase of the crop cycle) than to differences between CT plants and NT plants in efficiency in taking up N from fertilizer. The differences between CT and NT for many of the traits observed in durum wheat plants increased with decreasing soil fertility and in particular with decreasing soil total N. In conclusion, the shift from CT to NT, which should be accompanied in any case by an increase in the N fertilization rate to take into account the reduction in soil N available for the crop, was less problematic in the Vertisol, which is more fertile and better structured than the two Inceptisols.

Keywords: Conventional tillage; No-tillage; N fertilization; Soil N availability; ^{15}N-fertilizer recovery

Abbreviations: NT, no-tillage; CT, conventional tillage; OM, organic matter; %^{15}N$_{REC}$, percentage of ^{15}N fertilizer recovery; CNRG, contribution of N remobilization to grain N yield; N0, no N fertilization; N40, N80, N120, and N160 fertilization with 40, 80, 120, and 160 kg N ha^{-1}; CDA, canonical discriminant analysis

1. Introduction

No-tillage (NT) is widely recognized as a viable soil management technique in sustainable agriculture (Derpsch, 2008). Compared to conventional tillage (CT; usually based on moldboard plowing), NT helps to protect the soil from erosion (Scopel et al., 2005); enhances aggregation and aggregate stability (Madari et al., 2005); improves soil hydraulic characteristics (Kay and VandenBygaart, 2002); preserves soil macro- and microfauna (Uri et al., 1999); enhances soil microbial activity (Sharifi et al., 2008); reduces fuel consumption, and saves labor and time (Kirkegaard, 1995). Moreover, NT tends to preserve soil water better than CT, which results in huge advantages for the cropping systems in arid and semiarid areas; this is generally attributed to the change in the soil porosity (into more small pores and fewer large pores), to the creation of a more continuous pore system (from decaying roots and soil macrofauna activity), and above all to the minor soil water evaporation in NT as a consequence of both the presence of crop residues on the soil surface and the minor soil surface roughness generated by soil cultivation (Blevins and Frye, 1993; Lampurlanés and Cantero-Martínez, 2006). Such potential benefits suggest that NT is advantageous for cereal-based systems in Mediterranean environments, where water scarcity during the spring is often the main factor limiting the growth and productivity of rainfed crops (Lampurlanés et al., 2002) and where soils are particularly prone to erosion because of their characteristics and morphology (45% of the Mediterranean region has slopes greater than 8%), of cultivation even in steep slopes, and of the high frequency of intense rainfall events in fall and winter (García-Ruiz et al., 2013; Raclot et al., 2016). Several studies carried out under Mediterranean conditions have confirmed the benefits of NT over CT in terms of both a reduction in soil erosion (García-Orenes et al., 2009) and a crop yield advantage, particularly in dry areas/years (Amato et al., 2013; Ruisi et al., 2014). Despite these benefits, however, NT systems are used rarely in the Mediterranean, being practiced on approximately 2% of the total cropland (FAO AQUASTAT, 2013). There are several reasons for this, such as a lack of policies encouraging the adoption of NT and likely also the resistance on the part of farmers, as its positive effects are often not immediately apparent but can only be seen after a new equilibrium in the soil has been established (Stubbs et al., 2004). Such benefits in fact are directly or indirectly attributable to the increase in soil carbon sequestration and storage that, under NT, occurs gradually over time (West and Marland, 2002; West and Post, 2002; González-Sánchez et al., 2012; Badagliacca et al., 2018) depending on several factors, including climatic conditions, soil characteristics, crop rotation, and other crop management practices. As a consequence, the shift from CT to NT can be thorny; the farmer often has to completely reorganize the production system to resolve the problems that will inevitably arise before a new equilibrium is reached. For example, application of NT can markedly affect the population of weeds (Sosnoskie et al., 2006; Giambalvo et al., 2012; Ruisi et al., 2015) and the incidence of pests and diseases (Paulitz et al., 2002), requiring adjustments to control strategies. At the same time, even from the first years of application, NT can result in considerable changes in organic matter (OM) mineralization rates, nitrogen (N) immobilization, N availability, and N-use efficiency of the crop (Gao et al., 2009; Stagnari et al., 2014). Obviously, these effects can vary greatly depending on the context in which NT is implemented (e.g., in terms of climatic conditions, soil type and fertility, crop rotation, crop management, and duration of application); this explains the inconsistent findings in the literature (Franzluebbers et al., 1995; McCarty et al., 1998; Peigné et al., 2007).

Although several studies have been conducted in the Mediterranean environment on the effects of the continuous application of NT over a high number of years on soil N dynamics and the crop response (e.g., Ruisi et al., 2016), relatively few studies have evaluated the effects of NT on soil N dynamics and the fate of the N fertilizer applied during the first year switch from CT to NT, which, as already said, can be thorny so much to induce in some cases the farmer to abandon NT. This knowledge is important to plan cropping management strategies sustainable from both the agronomic and environmental standpoints. Thus, the present study aimed to address the following questions: i) How, and to what extent, does switching from CT to NT alter the availability of soil N for crops and the fate of the N fertilizer applied? ii) How much do these changes vary by soil type and climatic conditions? To this end, we studied the effects of NT on soil N dynamics and crop growth and yield in comparison with CT by applying these two techniques to three soil types differing in physical and chemical characteristics. The experiment was conducted in a semiarid Mediterranean environment and replicated over 2 years. Durum wheat (*Triticum durum* Desf.) was used as the model plant because of its importance as a crop plant in arid and semiarid areas of the Mediterranean basin.

2. Materials and Methods

2.1. Site characteristics

Field experiments were performed during two growing seasons (2011–2012 and 2012–2013; hereafter referred to as 2012 and 2013, respectively) at three sites (highly representative of arable soils of the Sicilian inland), all located within the Pietranera farm, which is located about 30 km north of Agrigento, Sicily, Italy (37°30'N, 13°31'E; 178 m a.s.l.). The farm covers approximately 700 ha and includes a variety of soil types, morphologies, and orographies. The cropping systems of the farm are based on cereal crops (mainly durum wheat) in rotation with legumes (grain and fodder crops). Soil tillage management is based on moldboard plowing (followed by secondary tillage operations) for cereal crops or on minimum tillage for legume crops. Soil characteristics (referring to the 0- to 0.40-m layer) of the three experimental sites are reported in Table 1. The first soil, classified as Typic Calcixerept (Soil Survey Staff, 2006), is deep, with a clayey texture and a low to moderate OM content; it has a sub-angular structure and a sub-alkaline reaction. The second soil is a Vertic Haploxerept evolved on recent alluvial deposits. The soil is deep, with a sandy clay texture and a very low OM content; it has a granular structure, good drainage, and a sub-alkaline reaction. The third soil, classified as Chromic Haploxerert, is a fine-clayey, calcareous, mixed, xeric Vertisol that developed on Mio-Pliocenic clayey substrata. It is especially rich in montmorillonitic clays that promote swelling and shrinking in the soil; it can be considered the most productive soil because of its high natural fertility.

The climate is semiarid Mediterranean, with a mean annual rainfall of 581 mm, mostly in autumn/winter (74%) and in spring (18%), and a mean annual PET of about 1100 mm (calculated using the Penman–Monteith method). The dry period is from May to September. The mean air temperature is 15.9°C in autumn, 9.8°C in winter, and 16.5°C in spring. The average minimum and maximum annual temperatures are 10.0°C and 23.3°C, respectively.

Table 1. Physical and chemical characteristics of the top layer (0–40 cm) of the three soil types where the experiment was conducted.

		Soil type (St)		
		St 1	St 2	St 3
	Unit	Typic Calcixerept (Inceptisol)	Vertic Haploxerept (Inceptisol)	Chromic Haploxerert (Vertisol)
Altitude	m a.s.l.	245	150	175
Particle size analysis:				
Clay	g kg^{-1}	558	267	525
Silt	g kg^{-1}	197	247	227
Sand	g kg^{-1}	245	486	248
pH (1:2.5 H_2O)	–	8.0	8.0	8.2
Total C (Walkley Black)	g kg^{-1}	10.6	6.3	16.8
Total N (Kjeldahl)	g kg^{-1}	0.61	0.86	1.78
Available P (Olsen)	mg kg^{-1}	28.8	55.9	40.1
Cation exchange capacity	cmol+ kg^{-1}	28.1	26.8	35.0
Water content at:				
field capacity	cm^3 cm^{-3}	0.35	0.28	0.37
permanent wilting point	cm^3 cm^{-3}	0.19	0.19	0.20

2.2. Experimental design and crop management

The experiments were set up as a split-plot design with four replications. The main plots (90 m^2 each) were the soil tillage techniques: CT or NT. Each subplot received a different level of N fertilizer: 0, 40, 80, 120 or 160 kg N ha^{-1} (hereafter referred to as “N0”, “N40”, “N80”, “N120” and “N160,” respectively). The size of each subplot was 18 m^2 (16 rows, each 6.0 m long, spaced at 0.1875 m). In both 2012 and 2013, the previous crop was berseem clover (*Trifolium alexandrinum* L.) in all three sites; before berseem clover was sown, soils were always managed with one shallow harrowing operation to prepare a proper seedbed. Berseem clover was managed by cutting plants to an ~8 cm stubble height in the first decade of April and allowing plant regrowth to produce seeds. Standing straws were always left in the soil, and loose residues (3−4 Mg DM ha^{-1}, on average) were spread uniformly throughout the plot. In 2013, the experiments were performed in the same three sites in surfaces adjacent to those used in 2012, to maintain “first year switch conditions”.

CT consisted of one moldboard plowing to a depth of 0.30 m in the summer, followed by two shallow harrowing operations before sowing to control weeds and prepare a suitable seedbed. NT consisted of sowing by direct drilling; before sowing, weeds were controlled with glyphosate (N-[phosphonomethyl] glycine) at rates of 720 to 1440 g a.i. ha^{-1} depending on the development of the weeds; sowing was performed by direct drilling with a no-till seed drill. In CT treatments, residues of previous crops were incorporated into the soil, whereas in NT treatments they were left on the soil surface (to ensure that soil coverage was always more than 30%). Cultivation and tillage treatments were performed with commercial farm equipment. Phosphorus fertilizer was applied to the surface of each plot before planting as triple superphosphate at a rate of 69 kg P_2O_5 ha^{-1}. Nitrogen fertilization was done by hand-applying in each plot the appropriate amount of N fertilizer (as $[NH_4]_2SO_4$) according to the experimental design. The total amount of N fertilizer was always split applied, with 50% applied at crop emergence and 50% applied at the end of tillering. Durum wheat (cv Vertola) was planted (for all treatments) at 350 viable seeds m^{-2} always in the second decade of December. Vertola (released in 2003) is characterized by short plant height, very early heading and maturity, a high yield potential, and good pasta-making quality. Spontaneous weeds were controlled at an early growth stage with application of thifensulfuron-methyl (25 g a.i. ha^{-1}) and tribenuron-methyl (12.5 g a.i. ha^{-1}).

Each year and at each site, soon after crop emergence, two sampling areas of 2.25 m^2 each (8 rows 1.5 m long) were identified within each subplot for subsequent measurements. In N80 treatments, a ^{15}N fertilizer (as $[NH_4]_2SO_4$) with an isotopic enrichment of 1.33 atom% was added to the two sampling areas (50% at crop emergence and 50% at the end of tillering) following the application procedure described by Høgh-Jensen and Schjoerring (1994); the rest of the subplots outside of the ^{15}N-labeled areas received equivalent amounts of unlabeled fertilizer.

Soil samples (0- to 0.40-m layer) were collected from each subplot immediately before sowing and soon after the harvesting of wheat and analyzed for 2M KCl-extractable NH_4^+–N and NO_3^-–N (10 g of soil in 100 mL of extractant) using a Bran+Luebbe AutoAnalyzer 3 (Norderstedt, Germany).

In both years, heading (stage 59, Zadoks scale; Zadoks et al., 1974) was reached in the third decade of April. Both at heading and at maturity (stage 92, Zadoks scale), a sample of total aboveground plant material from a 0.50 m segment of the two central rows of each sampling area was taken, oven-dried at 60°C for 48 h, weighed, ground to a fine powder (sieved using a 0.1-mm mesh) in a fast-running mill, and analyzed for total N and, limited to N0 and N80, for ^{15}N enrichment. Concentrations of total N were assessed using the Dumas method (flash combustion with automatic N analyzer; DuMaster D-480, Büchi Labortechnik AG, Flawil, Switzerland), and ^{15}N concentrations were determined using a Roboprep-CN and 20-20 isotope ratio mass spectrometer (Europa Scientific Ltd, Crewe, UK). In the rest of each sampling area, biomass production was determined. All samples were oven-dried at 60°C for 48 h and weighed. For each subplot, at harvest, grain yield was recorded after hand-cutting plants from a sampling area of 2.25 m^2 and threshing with a laboratory thresher for cereals. Grain N content (g kg^{-1}) was also recorded. The weather data were collected from a weather station located within 500 m of the sites.

2.3. Calculations and statistical analyses

Data on ^{15}N enrichment of biomass, taken both at heading and at maturity, were used to calculate labeled-fertilizer N recovery ($^{15}N_{REC}$) on an area basis (kg N ha^{-1}) and percentage basis according to Hauck and Bremner (1976):

$$^{15}N_{REC} = N_t \times \frac{^{15}N_{fp} - {}^{15}N_{nfp}}{^{15}N_{fert} - {}^{15}N_{nfp}} \qquad [1]$$

and

$$\%\,^{15}N_{REC} = \frac{^{15}N_{REC}}{f} \times 100 \qquad [2]$$

where N_t is the plant N uptake (kg N ha^{-1}), $^{15}N_{fp}$ is the atom% ^{15}N in the fertilized plants (N80 treatments), $^{15}N_{nfp}$ is the atom% ^{15}N in the nonfertilized plants (N0 treatments), $^{15}N_{fert}$ is the atom% ^{15}N in the fertilizer, and f is the fertilizer rate (kg N ha^{-1}).

According to Huggins and Pan (1993), the soil N potentially available for the crop (i.e., N supply, N_s, kg N ha^{-1}) was estimated as the amount of applied N (f) plus N_t plus residual postharvest N in the soil (kg N ha^{-1}), the latter two determined from control subplots (no applied N).

Moreover, the following parameters related to N accumulation and remobilization were calculated according to Cox et al. (1986) and Arduini et al. (2006):

- postheading N accumulation (kg N ha^{-1}), as the difference between the amount of N uptake (kg N ha^{-1}) in the aboveground biomass at heading and at maturity ;
- N remobilization (kg N ha^{-1}), as the amount of N uptake (kg N ha^{-1}) in the aboveground biomass at heading minus the difference between the amount of N uptake (kg N ha^{-1}) in the aboveground biomass and the grain N yield (kg N ha^{-1}) at maturity;
- N remobilization efficiency, as the ratio of the amount of N remobilization (kg N ha^{-1}) to the amount of N uptake (kg N ha^{-1}) in the aboveground biomass at heading;
- contribution of N remobilization to grain N yield (CNRG), as the percentage ratio of N remobilization (kg N ha^{-1}) to grain N yield (kg N ha^{-1}).

Data were analyzed using a linear model for split-plot design (Montgomery, 1997), with tillage system as the main plot and N fertilization rate as the subplot, replicated four times and in three different sites. Besides the site, two more factors could be considered—soil and year—but these and their interaction were confounded with site. To establish which of the two (soil or year) primarily influenced the variability in site and its interaction with tillage system and N fertilization rate, we used two different models to analyze the data with respect to each output variable: i) a model with site, tillage system, and N fertilization rate and their interactions; and ii) a model with soil, year, tillage system, and N fertilization rate and their interactions. In comparing the analysis of variance tables of the two models, we partitioned the effect of site and of its interaction with tillage system and N fertilization rate between a part corresponding to year and a part corresponding to soil. Means differences were compared using the Tukey test ($P < 0.05$ and 0.01). The entire analysis was carried out using R (2011).

Finally, canonical discriminant analysis (CDA) was performed (SAS Institute, 2008) to determine i) whether multivariate statistically significant differences existed between tillage systems across year, N fertilization rate, and site based on some selected plant traits; and ii) which of these traits accounted the most for these differences. Canonical variable means (centroid values) were calculated for each combination (tillage system/year, tillage system/N fertilization rate, and tillage system/site), and the significance between means was determined using the Mahalanobis distance.

3. Results

The total rainfall during the first growing season was 596 mm (Fig. 1), which was very close to the long-term average. Rainfall was well distributed over the growing season and allowed for satisfactory plant growth and yield for the trial environment. The mean temperature for the year was slightly lower than the normal mean temperature, especially during the winter season. During the second growing season, the total rainfall was 866 mm, about 50% greater than the long-term average. Rainfall mainly occurred in October (180 mm) and between January and March (430 mm), with a peak in January (185 mm), causing severe water logging during the vegetative phase of the crop cycle. The mean temperature was similar to the normal mean temperature.

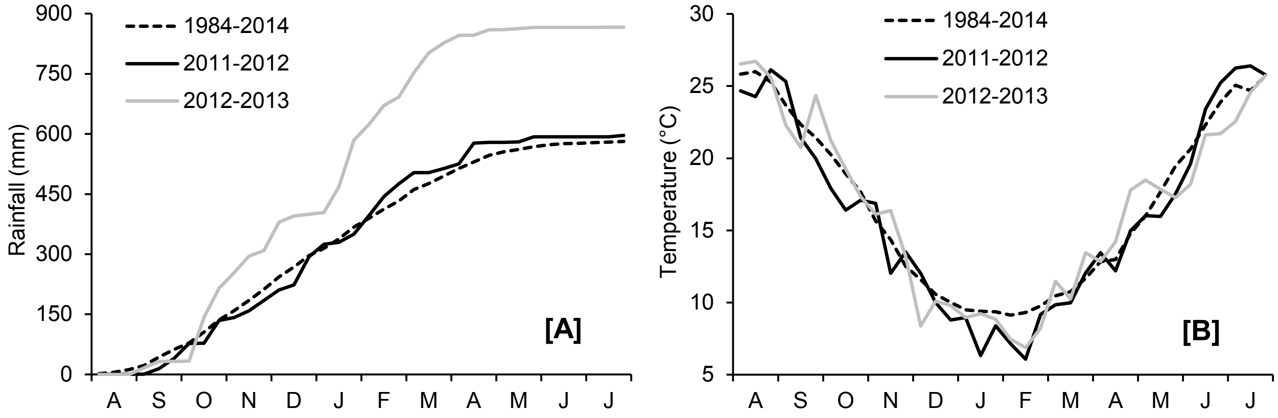

Fig. 1. Accumulated rainfall [A] and 10-day mean air temperature [B] at the experimental site during the two growing seasons (2011–2012 and 2012–2013). 30-year average 10-day temperatures and accumulated rainfall are also included.

Table 2 shows the results of the statistical analysis of the effects of the applied treatments (and their interactions) on wheat yield and N parameters. The great variability in the climatic conditions between the two experimental years markedly influenced crop growth and productivity, with the grain yield approximately twice as great in 2012 as in 2013 (on average 4.61 and 2.34 Mg ha^{-1}, respectively). The effects of tillage system on most of the observed traits varied greatly by site. The greatest contribution to the significance of this interaction was almost always attributable to year; in contrast, the contribution of soil type was generally low. For some traits, the effects of tillage system also varied by N fertilization.

In 2012, compared to CT, NT negatively affected grain yield in the two Inceptisols (–32% and –17%, respectively, in St1 and St2) but not in the Vertisol (St3; Fig. 2), whereas in 2013 no differences were observed between CT and NT in grain yield.

Table 2. Analysis of variance: *P*-values for the effects of the applied treatments on the traits measured on durum wheat plants. Percentage values in brackets represent the proportion of the variance explained by the factor.

	D.f.	Grain yield	N uptake at heading	N uptake at maturity	N uptake heading to maturity	Grain N content	Grain N yield	$^{15}N_{REC}$ at heading¶	$^{15}N_{REC}$ at maturity¶	N remob.‡	N remob. efficiency	N supply¶	CNRG§
		Mg ha^{-1}	kg N ha^{-1}	kg N ha^{-1}	kg N ha^{-1}	g kg^{-1}	kg N ha^{-1}	%	%	kg N ha^{-1}	—	kg N ha^{-1}	%
Site	5	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	0.002
Year (Y)	1	*** (73%)	*** (96%)	*** (84%)	*** (31%)	*** (78%)	*** (77%)	*** (90%)	*** (89%)	*** (89%)	ns (4%)	*** (75%)	** (39%)
Soil type (St)	2	*** (14%)	** (2%)	*** (6%)	*** (24%)	** (11%)	*** (10%)	*** (9%)	*** (9%)	*** (11%)	*** (81%)	*** (13%)	* (33%)
Y × St	2	*** (13%)	* (2%)	*** (10%)	*** (45%)	ns (11%)	*** (13%)	ns (1%)	† (2%)	ns (0%)	† (15%)	*** (12%)	* (28%)
Tillage (T)	1	<0.001	0.004	0.012	*ns*	*ns*	<0.001	*ns*	*ns*	0.018	*ns*	<0.001	*ns*
T × Site	5	<0.001	0.003	0.009	*ns*	*ns*	<0.001	0.049	0.056	0.049	*ns*	0.013	0.068
T × Y	1	*** (77%)	*** (93%)	*** (77%)	ns (13%)	ns (65%)	*** (75%)	** (75%)	** (88%)	** (74%)	ns (2%)	** (71%)	ns (0%)
T × St	2	ns (7%)	ns (4%)	ns (6%)	† (78%)	ns (16%)	ns (9%)	ns (0%)	ns (2%)	ns (11%)	† (57%)	ns (17%)	* (90%)
T × Y × St	2	* (16%)	ns (3%)	* (17%)	ns (9%)	ns (19%)	* (16%)	ns (25%)	ns (10%)	ns (10%)	ns (41%)	ns (12%)	ns (10%)
Fertilization (F)	4	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	—	—	<0.001	0.002	—	*ns*
F × T	4	0.002	0.001	0.013	*ns*	0.043	*ns*	—	—	*ns*	0.037	—	*ns*
F × Site	20	*0.098*	<0.001	<0.001	*ns*	0.003	<0.001	—	—	<0.001	<0.001	—	<0.001
F × Y	4	* (33%)	*** (91%)	*** (86%)	ns (31%)	*** (55%)	*** (60%)	—	—	*** (47%)	ns (9%)	—	* (18%)
F × St	8	* (53%)	ns (5%)	ns (4%)	ns (33%)	ns (26%)	* (29%)	—	—	** (40%)	*** (71%)	—	*** (59%)
F × Y × St	8	ns (14%)	ns (4%)	ns (10%)	ns (36%)	ns (19%)	ns (11%)	—	—	ns (13%)	† (20%)	—	ns (23%)
Site × T × F	20	*ns*	*ns*	*ns*	*ns*	*ns*	*ns*	—	—	*ns*	*ns*	—	*ns*
Y × T × F	4	ns (24%)	ns (44%)	ns (25%)	ns (14%)	ns (20%)	ns (12%)	—	—	ns (36%)	ns (15%)	—	ns (8%)
St × T × F	8	ns (24%)	ns (26%)	ns (20%)	ns (38%)	ns (19%)	ns (25%)	—	—	ns (8%)	ns (39%)	—	ns (38%)
Y × St × T × F	8	ns (52%)	ns (30%)	ns (55%)	ns (47%)	† (60%)	ns (63%)	—	—	ns (55%)	ns (46%)	—	ns (54%)

†, *, **, and *** indicate that the factor is significant at $P \leq 0.1$, 0.05, 0.01, and 0.001, respectively; *ns*, not significant at $P \leq 0.1$. ¶ For the ^{15}N fertilizer recovery (both at heading and maturity) and for the N supply the analysis of variance was performed only on the data obtained from N80

and N0, respectively. ‡N remob. is N remobilization. § CNRG is the contribution of N remobilization to grain N yield (as percentage ratio of N remobilization to grain N yield).

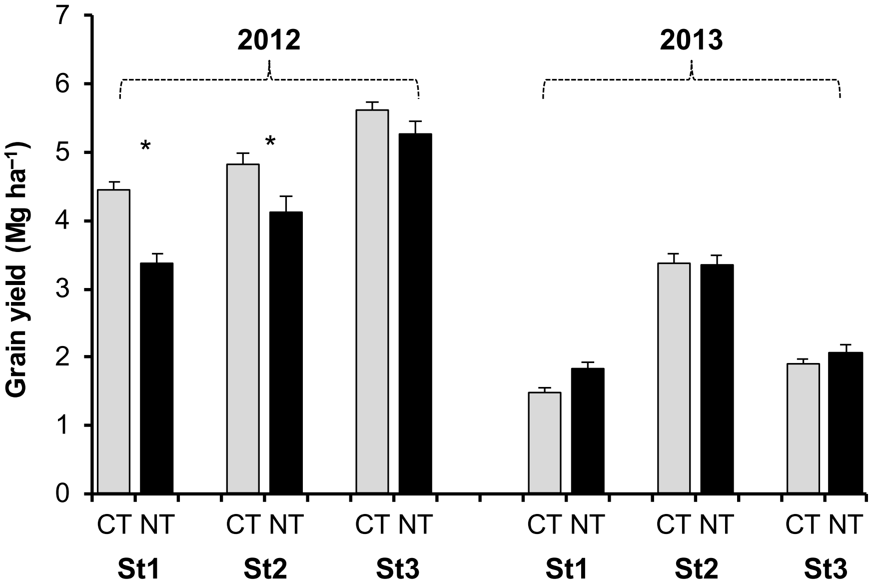

Fig. 2. Grain yield of wheat as affected by tillage system and soil type, separately for year. CT, conventional tillage; NT, no-tillage. St1= Typic Calcixerept; St2= Vertic Haploxerept; St3= Chromic Haploxerert (see Table 1). Each value is a mean of 4 data (4 replicates). Vertical bars indicate standard errors of each mean value. Asterisks in the top of the histograms indicate that tillage system effect was significant at $P \leq 0.05$.

On average, a strong grain yield advantage of CT over NT (approximately 0.6 Mg ha^{-1} of grain) was observed with no N fertilization (Fig. 3A). This benefit decreased progressively with increases in the N fertilizer rate, to the point that at 120 kg ha^{-1} of N differences between CT and NT were negligible. On average, grain N content increased with the N fertilization rate (from 24.0 g kg^{-1} in N0 to 25.8 g kg^{-1} in N160); the effects on N fertilization were higher in CT compared to NT, and the differences between the two tillage systems were significant at the higher N fertilizer rates (N120 and N160; Fig. 3B). On the whole, grain N yield varied in the same way as the grain yield, but, unlike the latter, CT and NT showed a similar response to the increase in the N fertilizer rate (Tillage System × N Fertilization Rate not significant).

N supply was on average markedly higher in 2012 than in 2013 (141 vs. 65 kg N ha^{-1}, respectively). Nitrogen supply was significantly higher under CT than NT only in 2012 (Fig. 4). In this year, the superiority of CT over NT was clear in the two Inceptisols (+52% and +32% in St1 and St2, respectively) and moderate in the Vertisol (i.e., St3; +9%).. The grain yield of wheat increased proportionally with increasing N supply, with no significant differences observed in this relationship by tillage system (Fig. 5A). Moreover, the variation in N supply due to the tillage system adopted explained about 90% of the variation observed in the grain yield between NT and CT systems in N0 (Fig. 5B).

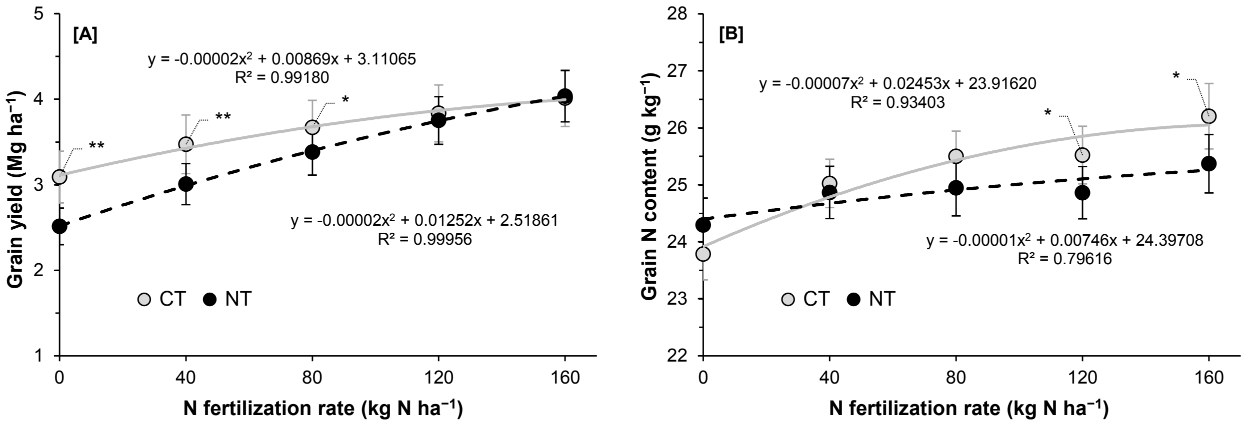

Fig. 3. Grain yield [A] and grain N content [B] in wheat grown under conventional tillage (CT) and no-tillage (NT) at five N fertilization levels (0, 40, 80, 120, and 160 kg N ha^{-1}). Each value is a mean of 24 data (2 years × 3 soil types × 4 replicates). Vertical bars indicate standard errors of each mean value. Asterisks indicate that tillage system effect was significant at $P \leq 0.05$ (*) or $P \leq 0.01$ (**).

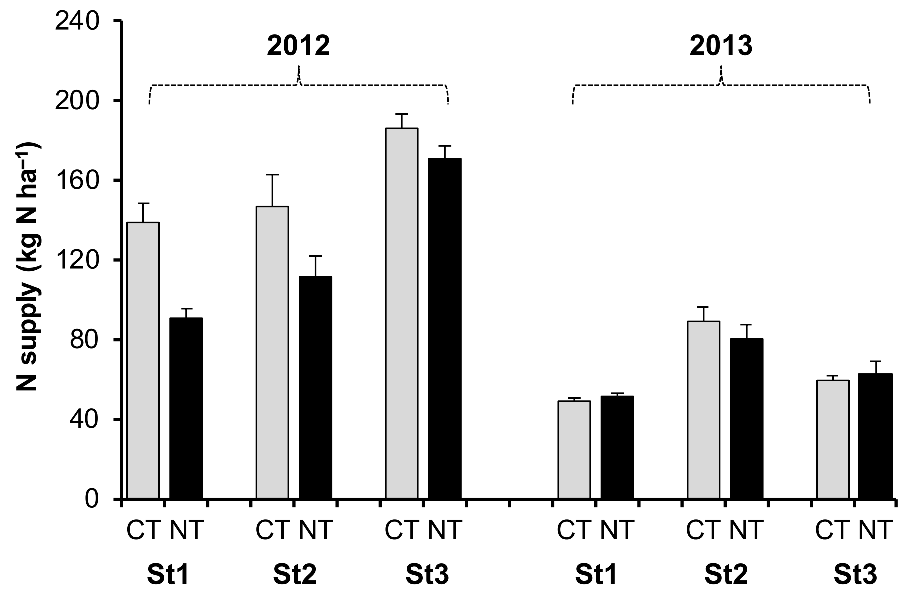

Fig. 4. N supply measured in non-N fertilized plots (N0) as affected by tillage system and soil type, separately for year. CT, conventional tillage; NT, no-tillage. St1= Typic Calcixerept; St2= Vertic Haploxerept; St3= Chromic Haploxerert (see Table 1). Each value is a mean of 4 data (4 replicates). Vertical bars indicate standard errors of each mean value.

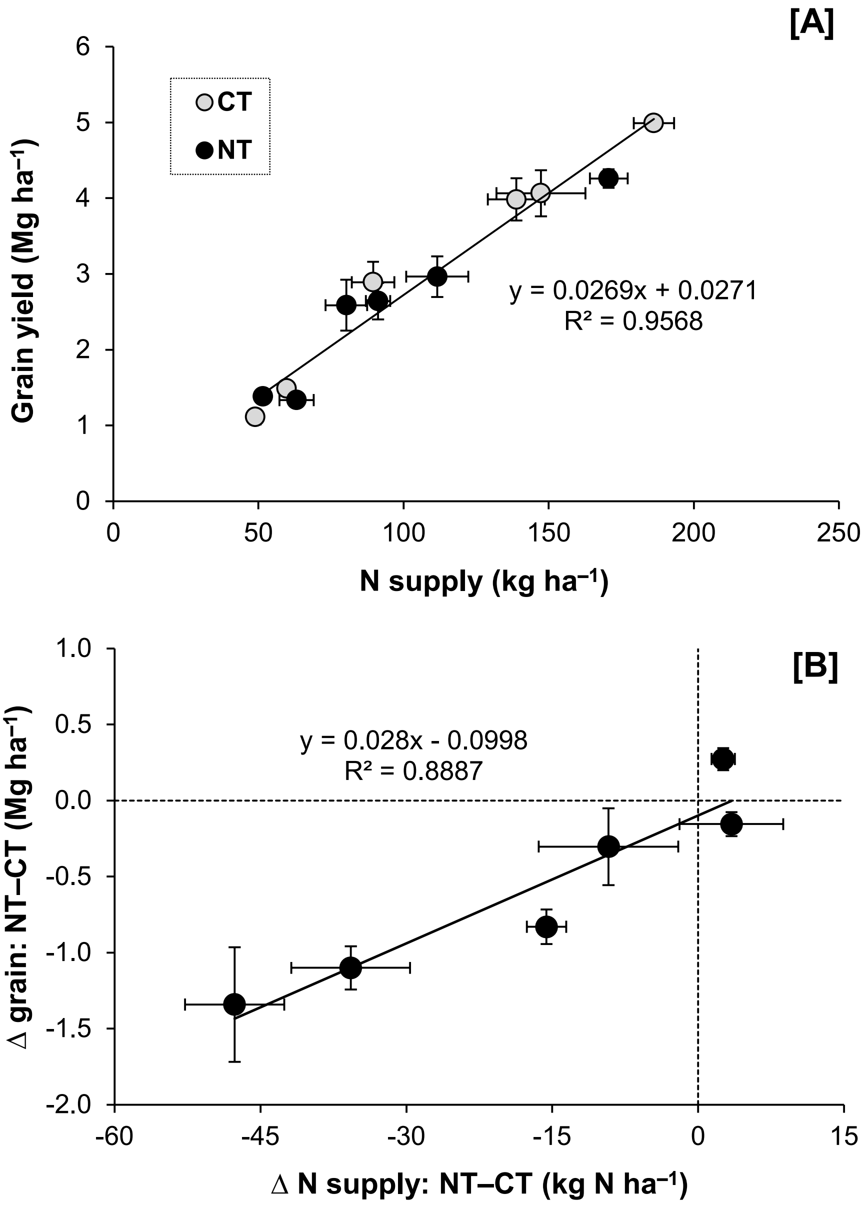

Fig. 5. [A]: Relationship between the N supply and the grain yield of wheat; each value is a mean of 24 data (2 years × 3 soil types × 4 replicates); since no significant differences between NT and CT were found in the regression equations obtained, a unique regression line (and relative equation) was reported. [B]: Relationship between the differences in the N supply between no-tillage (NT) and conventional tillage (CT) and the differences in the grain yield of wheat between NT and CT in the various trial conditions (n = 24; 2 years × 3 soil types × 4 replicates). Both the relationships displayed refer to the non-N fertilized condition (N0). In both figures, horizontal and vertical bars indicate the standard errors of each mean value.

Tillage system influenced N uptake (both at heading and at maturity) differently in the 2 years (Fig. 6A and 6C). In 2012, application of NT instead of CT led to a decrease in N uptake (on average

−11% both at heading and at maturity), whereas in 2013, no differences between NT and CT were observed for N uptake (both at heading and at maturity). On average, the advantage for CT over NT for N uptake, large and significant under N0 conditions, decreased progressively as the rate of N fertilizer increased so that starting from N80 and from N120 no significant differences were observed between the two tillage systems, respectively at heading and at maturity (Fig. 6B and 6D). For N uptake at maturity the effects of tillage varied by soil type, but only in 2012 (Fig. 7). Differences due to tillage system were not observed in the amount of N taken up by wheat from heading to maturity (Table 2).

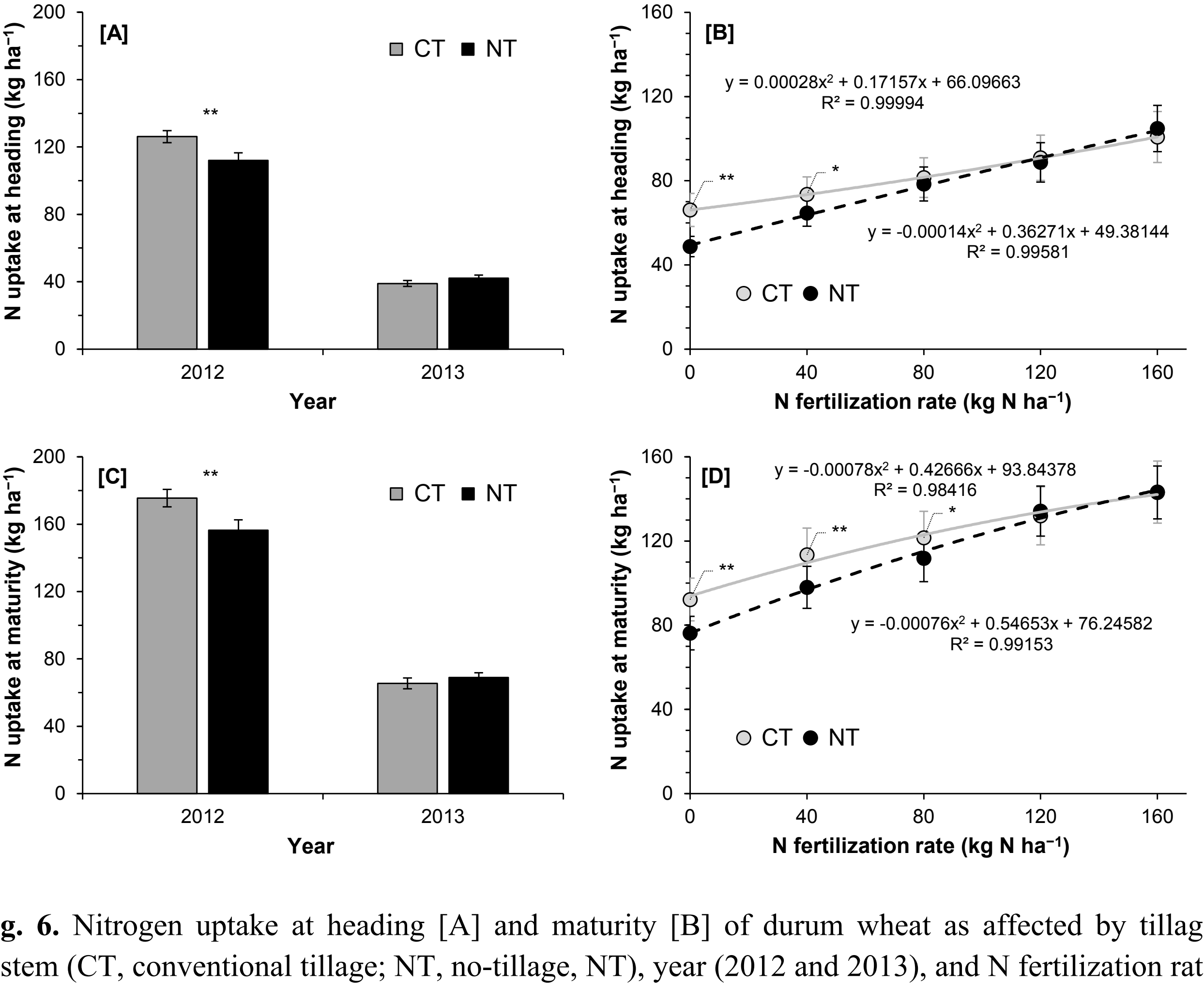

Fig. 6. Nitrogen uptake at heading [A] and maturity [B] of durum wheat as affected by tillage system (CT, conventional tillage; NT, no-tillage, NT), year (2012 and 2013), and N fertilization rate (0, 40, 80, 120, and 160 kg N ha^{-1}). Each value is a mean of 60 data (5 N fertilizer rates × 3 soil types × 4 replicates) in [A] and [C], and of 24 data (2 years × 3 soil types × 4 replicates) in [B] and [D]. Vertical bars indicate standard errors of each mean value. Asterisks indicate that tillage system effect was significant at $P \leq 0.05$ (*) or $P \leq 0.01$ (**).

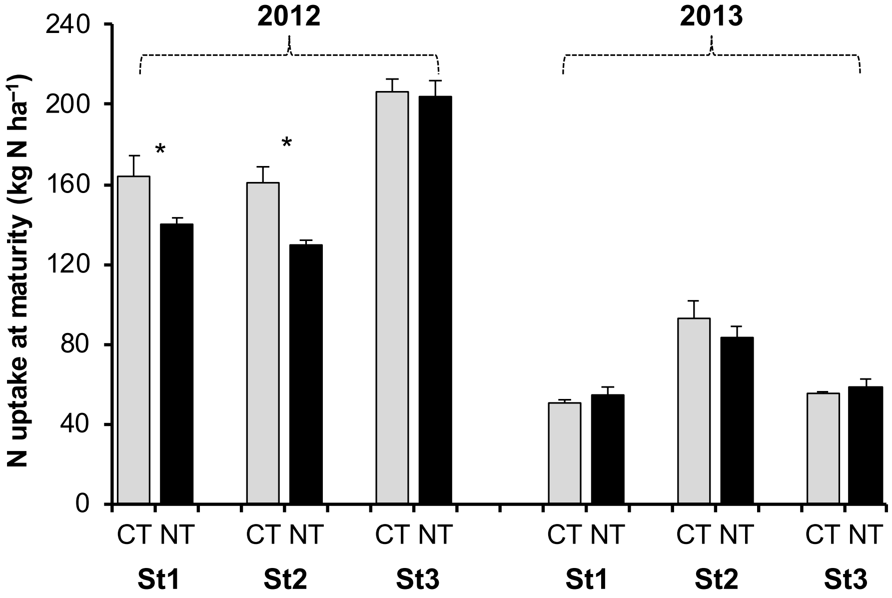

Fig. 7. N uptake of wheat at maturity as affected by tillage system and soil type, separately for year. CT, conventional tillage; NT, no-tillage. St1= Typic Calcixerept; St2= Vertic Haploxerept; St3= Chromic Haploxerert (see Table 1). Each value is a mean of 4 data (4 replicates). Vertical bars indicate standard errors of each mean value. Asterisks in the top of the histograms indicate that tillage system effect was significant at $P \leq 0.05$.

On average, the recovery of ^{15}N fertilizer (%^{15}N$_{REC}$) was markedly higher in 2012 than in 2013 (Fig. 8); at heading %^{15}N$_{REC}$ was 31.2% and 6.5% in 2012 and 2013, respectively, whereas at maturity it was 41.0% and 11.1%, respectively. The %^{15}N$_{REC}$ at heading was significantly lower in NT than in CT in 2012. The opposite was observed in 2013, when %^{15}N$_{REC}$ was significantly higher in NT than CT both at heading and at maturity. Moreover, the advantage for CT over NT for %^{15}N$_{REC}$ was canceled out in 2012 from heading to maturity, whereas in 2013 the advantage for NT over CT increased from heading to maturity.

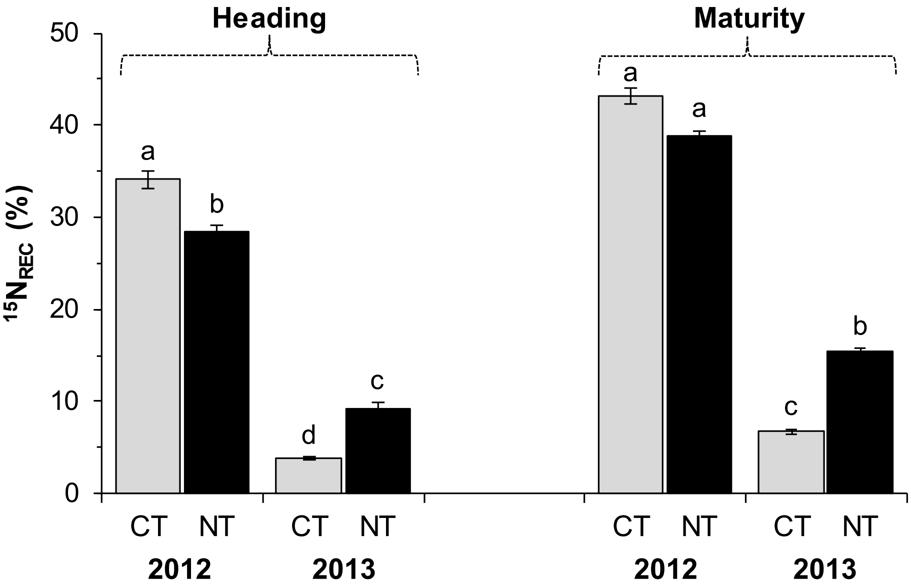

Fig. 8. Percentage of the recovery of the ^{15}N-fertilizer (%$^{15}N_{REC}$) at heading and maturity by durum wheat as affected by tillage system in the two experimental years. CT, conventional tillage; NT, no-tillage. Each value is a mean of 12 data (3 soil types × 4 replicates). Vertical bars indicate standard errors of each mean value. Within each phenological stage, different letters denote significant differences ($P \leq 0.05$).

The amount of N remobilized during the grain filling was markedly higher in 2012 than in 2013 (on average 78.0 vs. 28.4 kg N ha^{-1}, respectively). Application of NT instead of CT resulted in an important decrease in N remobilization only in 2012 (Fig. 9). The effects of tillage system on both N remobilization efficiency and CNRG varied by soil type, being slightly higher in CT than NT only in the Vertisol (St3) (data not shown).

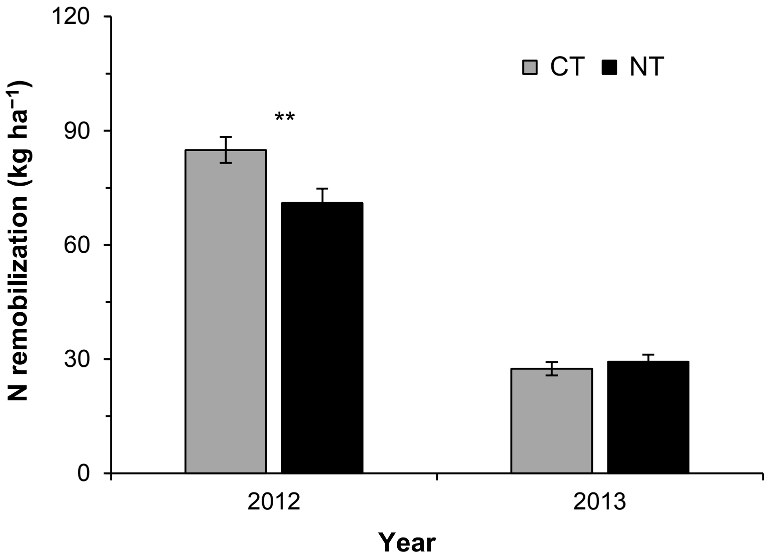

Fig. 9. Nitrogen remobilization in wheat grown under conventional tillage and no-tillage in the two experimental years. CT, conventional tillage; NT, no-tillage. Each value is a mean of 60 data (5 N fertilizer rates × 3 soil types × 4 replicates). Vertical bars indicate standard errors of each mean value. Asterisks in the top of the histograms indicate that tillage system effect was significant at $P \leq 0.01$.

The CDA performed for tillage system and year and based on data for eleven selected traits clearly discriminated the treatments (Fig. 10A). First canonical variable (CAN1) mostly varied according to grain yield, N uptake (both at heading and at maturity), and the $\%^{15}N_{REC}$ (both at heading and at maturity), whereas CAN2 was mostly influenced (negatively) by N remobilization efficiency and grain yield. CAN1 discriminated clearly 2012 by 2013, whereas CAN2 discriminated NT by CT only for 2012. Specifically, NT-2012 and CT-2012 plotted in the upper right and bottom right quadrants, respectively (highly significant differences based on Mahalanobis squared distances), whereas NT–2013 and CT–2013 plotted near each other with no significant differences. In the CDA based on tillage system and N fertilization (Fig. 10B), distances between NT and CT systems decreased as the N fertilizer rate increased, so that differences based on Mahalanobis squared distances were significant only at N0 and N40. Both CAN1 (which was influenced by grain and grain N yields, N uptake both at heading and at maturity) and CAN2 (which varied mainly according to the grain N content and the CNRG) contributed to discriminating the treatments.

Finally, the CDA based on tillage system and soil type also discriminated the treatments (Fig. 10C). CAN1 was positively influenced by grain and grain N yields and N remobilization, whereas the traits with the greatest influence on CAN2 were the N uptake (both at heading and at maturity) and the grain N content. As can be seen from the figure, distances between CT and NT systems varied according to soil type (St1 > St2 > St3).

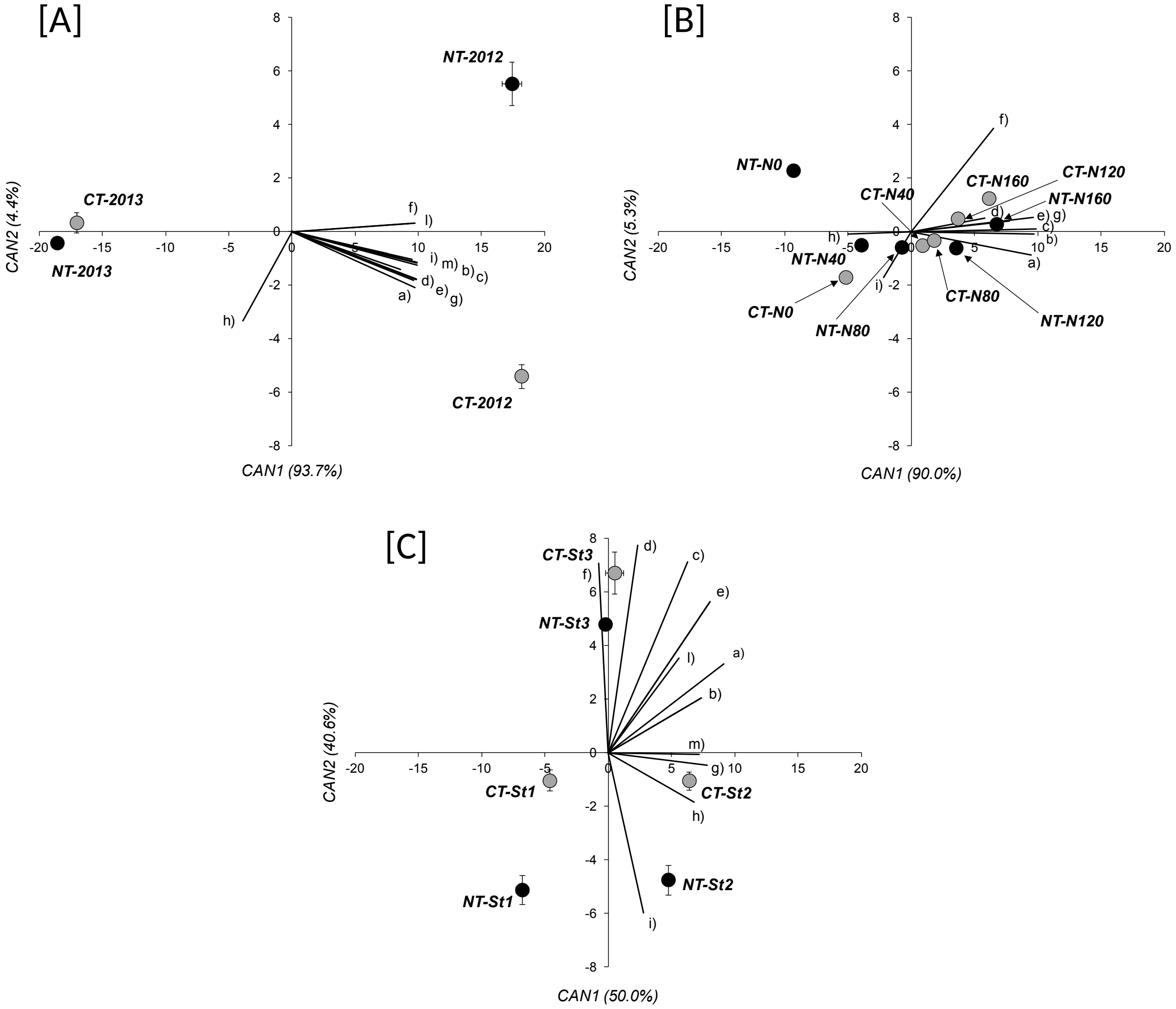

Fig. 10. Canonical discriminant analysis (CDA). Canonical variable means (centroid values) were calculated for each combination: [A]= tillage system/year; [B]= tillage system/N fertilization rate; and [C]= tillage system/soil type. CAN1, first canonical variable; CAN2, second canonical variable. CT, conventional tillage; NT, no-tillage. N0, N40, N80, N120, and N160 represent the following fertilization rates: 0, 40, 80, 120, and 160 kg N ha^{-1}, respectively. St1, Typic Calcixerept; St2,

Vertic Haploxerept; St3, Chromic Haploxerert (see Table 1). CDAs were performed on the basis of only 11 selected traits measured on plants: a) grain yield; b) N uptake at heading; c) N uptake at maturity; d) N uptake from heading to maturity; e) grain N yield; f) grain N content; g) N remobilization; h) N remobilization efficiency; i) ratio between N remobilization and grain N yield(CNRG); l) recovery of ^{15}N-fertilizer at heading; m) recovery of ^{15}N-fertilizer at maturity. Within each diagram, the direction and length of each line indicate the canonical loadings of the measured traits on the first two canonical variables.

4. Discussion

The results of the present study suggest that switching from CT to NT influences crop response through effects on soil N dynamics and in particular on the soil N potentially available for the crop. Soil cultivation, which increases soil aeration and mixes and incorporates crop residues into the soil, generally increases the decomposition of OM by altering the soil structure and temperature, by modifying the degree of physical protection of OM from microorganisms (or their enzymes), and by stimulating microbial activity and modifying the rate of microbial biomass turnover (Silgram and Shepherd, 1999; Watson et al., 2002; Mikha and Rice, 2004). This leads to a faster release of N from both the native soil OM and the crop residues under CT compared to NT systems and thus to an increase in the N supply. The decrease in N supply in NT compared to CT can also be the result of an increased N immobilization rate under NT (Rice and Smith, 1984; Drinkwater et al., 2000). In fact, many studies have shown that surface-managed crop residues (such as those generally found in NT systems) have a greater potential for N immobilization than incorporated residues (Holland and Coleman, 1987; Van Den Bossche et al., 2009).

In the present study, N supply was markedly lower in NT than CT in 2012 (on average about 125 and 160 kg N ha^{-1} in NT and CT,respectively), whereas no differences were observed by tillage system in 2013 (about 65 kg N ha^{-1} for both CT and NT). The differences in N supply between CT and NT affected durum wheat grain yield and N uptake, which both varied significantly in 2012 (with higher values in CT than NT) but not in 2013 (CT = NT). Moreover, we found a strong positive relationship between the differences in N supply and the differences in grain yield between CT and NT, showing that a high proportion (about 90%) of the variation observed for grain yield between NT and CT was explained by the variation in N supply between these two tillage systems. The hypothesis that the lower N supply under NT was the determining factor in reducing grain yield and N uptake is corroborated by the fact that the differences between CT and NT for these two traits progressively decreased as the N fertilizer rate increased. This was confirmed by the results from the CDA that showed how the distances between NT and CT, which were significant in N0 and

N40, decreased as the N fertilizer rate increased, so much so that starting from N80 the distances between the two tillage systems were canceled out. Our data are in line with the findings of Lundy et al. (2015), who performed a meta-analysis to evaluate the influence of crop and environmental variables on NT productivity and found that N fertilization reduces yield declines following the adoption of NT.

The detrimental effects of the adoption of NT on durum wheat varied according to soil type as highlighted by the CDA and, in particular, these detrimental effects seemed to increase with decreasing soil fertility and in particular when soil total N decreased (St1 > St2 > St3). The differences between CT and NT for grain yield and N uptake were indeed much lower in the Vertisol (the N-richest soil with a total N content > 1.5 g kg^{-1} of soil) than in the two Inceptisols (both with a total N content < 1 g kg^{-1} of soil). Compared to NT, CT promotes an increase in soil OM decomposition (and hence the release of N) through increase in soil aeration (Six et al., 2000), in addition to the mechanisms listed above. Thus, in the Vertisol, which was characterized by a good structure due to the high contents of montmorillonitic clays and soil OM, the omission of soil cultivation likely did not reduced the soil OM decomposition rate, as was the case for the two Inceptisols, in which, in the absence of tillage, oxygen could have become a limiting factor for the soil OM decomposition process. The differences in N supply observed between CT and NT in the three soils used in the present study, which were significant and consistent in the two Inceptisols but very low in the Vertisol, support this hypothesis. Clearly, improvements in soil biological activity, porosity, OM content, and structure induced by the continuous application of NT over a number of years should progressively counteract the negative effects highlighted during the shift from CT to NT, as suggested by Rice et al. (1986) and Salinas-García et al. (1997). However, the results of an experiment carried out in the same environment as present study, in which NT was applied continuously for several years, while showing a progressive increase in NT with respect to CT for many of the above mentioned parameters (soil biological activity, OM content, etc.; Badagliacca et al., 2018), they also showed higher values of N supply under CT compared to NT, thus failing to support the hypothesis of occurrence of a transient effect. (Ruisi et al., 2016).

Some authors have attributed the differences in grain yield and N uptake between CT and NT to the differing efficiency of CT plants and NT plants in taking up N and other nutrients from the soil (Fox and Bandel, 1986; López-Bellido and López-Bellido, 2001). However, our research does not confirm this hypothesis. In fact, no differences were observed between the two tillage systems in either the amount of residual N in the soil at harvest (data not shown) or the ^{15}N fertilizer recovery fraction (%$^{15}N_{REC}$). Hence, the differences between CT and NT in N uptake have to be attributed to differences in the N supply between the two tillage systems, similar to what was observed in the above mentioned long-term tillage experiment (Ruisi et al., 2016).

In 2013, no differences were found between CT and NT in any of the observed traits, in contrast with the findings of 2012. This lack of differences can be explained by the excessive rainfall in the 100 days after sowing (more than 400 mm of rain), which most likely caused abundant leaching of both native soil N and N from fertilizer applied at crop emergence. Several studies have shown that in Mediterranean environments N losses through leaching can be considerable, particularly in years characterized by rainy autumns or winters. As reported by Demurtas et al. (2016) in a study carried out under Mediterranean conditions, about 70% of total annual NO_3^- leaching occurs between October and February, the period in which about 60% of the annual percolation occurs in Mediterranean areas as a consequence of the imbalance between rainfall and evapotranspiration (Arregui and Quemada, 2006; Carneiro et al., 2012). This most likely occurred in our experiment in 2013, and this hypothesis is furthermore supported by the values found for the ^{15}N fertilizer recovery fraction (%$^{15}N_{REC}$ = about 10% on average), which were much lower than both those observed in 2012 (%$^{15}N_{REC}$ = about 40% on average) and those usually reported in the literature for wheat grown in the same environment (%$^{15}N_{REC}$ = 25%–30%; Giambalvo et al., 2010; Ruisi et al., 2014, 2015). Previous studies have shown inconsistent findings for the influence of tillage system on N losses via leaching. In particular, the effect of tillage system seems to be highly dependent on soil N availability and pedo-climatic and agronomic factors (Hansen and Djurhuus, 1997; Constantin et al., 2010). Some research has highlighted the fact that adoption of NT increases NO_3^--N drainage losses through leaching, encouraging the formation of continuous soil macro-pores, which may increase NO_3^--N preferential flow (Paul and Clark, 1989; Patni et al., 1998). In contrast, other studies have reported less NO_3^--N leaching under NT due to the reduced amount of N available for leaching because of the lower N mineralization rate compared to CT (Levanon et al., 1993; Mkhabela et al., 2008). Melero et al. (2011) came to similar conclusions in an experiment carried out in a Mediterranean Vertisol. We think that in the present study, N leaching was likely much more intense under CT than under NT during 2013, thus nullifying the advantage of soil cultivation in terms of increased N supply with respect to NT, so much so that no differences were observed in crop response between the two tillage techniques. The hypothesis that N leaching could have been more intense in CT than NT finds further confirmation in the analysis of $\%^{15}N_{REC}$ recorded in 2013, which, although quite low overall, was significantly higher in NT than CT, implying that a greater amount of N fertilizer could have been lost via leaching in CT compared to NT.

Our results highlight the fact that the advantages for CT over NT for plant growth materialized already during the vegetative phase of the crop cycle, as shown by the values of N uptake at heading (on average higher in CT than NT). Thus, the adoption of CT could have led to more favorable conditions for more rapid plant early growth (tillering and first phases of stem elongation) compared to NT, with a consequential increase for plants under CT in N crop demand and a greater chance that the crop would absorb the N provided by the fertilizer. Obviously, in addition to the higherN supply, other factors, such as the higher soil temperatures and aeration under CT, could have contributed to this result (Mielke et al., 1986; Giller et al., 2009; Rusinamhodzi et al., 2011).

Moreover, it is interesting to note that in the present research no differences between CT and NT were observed for the amount of N taken up by durum wheat from heading to maturity. The negative effects on initial crop growth associated with NT were evidently canceled out or partially counteracted by other factors during the reproductive phase of the crop cycle. For instance, compared to CT, in NT more water is generally available for the crop during the spring (i.e., during wheat grain filling), a fact that is attributable to reduced soil water evaporation with NT (Blevins and Frye, 1993; Lampurlanés and Cantero-Martínez, 2006) and to deeper soil water storage under this tillage system (Lampurlanés et al., 2016). This would also explain the higher N remobilization efficiency observed under NT compared to CT when N fertilizer was applied. Indeed, Ercoli et al. (2008) observed in durum wheat that N remobilization efficiency decreased with increasing water stress during grain filling and also that the magnitude of this effect increased with increasing availability of N, thus highlighting the fact that plant N status can influence plant tolerance to water stress. Moreover, it is reasonable to assume that the improved vegetative plant growth under CT could have led to an increased demand for transpiration and a faster depletion of soil water reserve in CT compared to NT, thus resulting in an amplification of water stress conditions during grain filling, which could have adversely affected N remobilization efficiency. On the other hand, in semiarid Mediterranean environments, which are characterized by low and erratic rainfall patterns during the growing season, it is particularly important to adopt crop management strategies that mitigate water stress during grain filling. These strategies should aim to limit excessive water consumption during the initial phase of crop growth and to maximize the availability of water during grain filling. From the above, it follows that adopting NT, which influences N and water availability patterns, represents a solution particularly suitable for Mediterranean environments, as it can enhance the efficient use of these resources (Cantero-Martínez et al., 2007; Morell et al., 2011). Therefore, it is not surprising that several studies carried out in Mediterranean areas have shown that NT generally results in higher grain yields than CT in dry years (Lampurlanés et al., 2002; Amato et al., 2013). Under these conditions, the adoption of NT instead of CT is often advantageous because of the benefits of NT in terms of soil water storage and reduced soil water evaporation (Lampurlanés and Cantero-Martínez, 2006). In our experiment, rainfall was normal or consistently above the long-term average, so the availability of water was certainly not the key factor that limited crop productivity (thus nullifying the potential advantage of NT over CT), whereas N supply was. We believe that if the growing seasons had been drier the results could have been different.

On the whole, the results of the present study show that the shift from CT to NT can be problematic, as it can negatively influence crop performance with effects more or less intense in relation to climate (mainly the amount and distribution of rainfall) and soil type. The effects of climate (which in this experiment were large and intense) and soil type (which were less intense) appeared to be connected to the influence that these two factors exerted on the soil N dynamics (N releases from the soil OM decomposition, N losses, etc.) and hence on the N supply in the soil, so much so that N fertilization progressively reduced or even canceled out the differences observed between the two soil tillage systems.

Moreover, results from the present study also show that i) the advantages of CT over NT for N supply and N uptake by the crop materialized already during the vegetative phase of the crop cycle; ii) the differences between CT and NT in N recovered by the crop from fertilizer were on average small but variable over the years; and iii) the shift from CT to NT was less problematic in the Vertisol, which is more fertile and better structured than the two Inceptisols.

In conclusion, switching from CT to NT can induce, already in the first year of transition, a significant reduction of the soil N potentially available for crops. The magnitude of such a reduction can vary in relation to climatic conditions and soil type. In particular, the differences between CT and NT for many of the traits observed in durum wheat plants increased with decreasing soil fertility and reduced progressively as the rate of N fertilization increased.

Our study suggests that the shift from CT to NT, which is desirable given the numerous agronomic and environmental benefits provided by NT (if continuously applied for a consistent number of years), must be accompanied by a reorganization of crop management, in particular the strategies of N fertilization, especially when the soil is poor in N. In particular, compared to CT, under NT conditions it would be appropriate to increase slightly the rate of N fertilizer applied in the early tillering stage of growth to take into account the reduction in the release of N from OM decomposition during the first phases of the growing season. Clearly, all of this should be done keeping in mind that Mediterranean environments can see rainy years leading to increases in N losses (especially via leaching). Our results may be useful in helping Mediterranean farmers to plan appropriate N fertilization strategies to make the shift from CT to NT more successful.

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Declaration of interest

Conflicts of interest: none.

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