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Consequences on macroporosity and bacterial diversity of adopting a no-tillage farming system in a clayish soil of Central Italy

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ABSTRACT

Conservation agricultural practices, such as no-tillage, crops rotation and balanced fertilization are increasingly adopted for maintaining soil fertility, improving crops health and reducing soil erosion.

The aim of this study was to evaluate the effects of the long-term adoption of contrasting tillage (no-tillage or conventional tillage) and N-fertilization (0 or 90 kg/ha N) practices on soil porosity and active bacterial communities in cropping system plots (sunflower–wheat or maize–wheat rotation) established on a clayish soil and under Mediterranean climate. Soil porosity was evaluated by micromorphological observations of soil thin sections. The composition and structure of the active bacterial communities were estimated by a culture-independent approach (reverse transcription – polymerase chain reaction – denaturing gradient gel electrophoresis, RT-PCR-DGGE) exploiting the 16S rRNA of bacteria and *nirK*, *nirS* and *nosZ* transcripts of denitrifiers. Finally, canonical correspondence analysis (CCA) was used to correlate microbial data with soil physical and chemical characteristics.

When repeated for a long period, no-tillage has significantly increased soil compaction compared to the conventional tilled soil. Soil compaction was likely responsible for creating a selective environment for active bacterial species. On the other hand, tillage favoured the richness and diversity of active soil bacteria by increasing the rate of diffusion of O_2 and the energy sources availability. A wide variability of active *nirK* denitrifiers was found in each soil management, while *nirS* denitrifiers were more closely related to lower porosity conditions. N fertilizer management seemed to affect mainly the active *nosZ* denitrifiers.

Our results suggested that conservation tillage practices on heavy clayish soils are not free of relevant side effects on soil porosity and bacterial soil communities.

1. Introduction

Concerns about the environmental effects of intensive agricultural practices, in particular the loss of soil organic matter, are shifting the focus away from conventional (high-input) farming systems to more ecologically sustainable systems (Lupwayi et al., 1998). Conservation agricultural practices, such as no-tillage and direct seeding, crop rotation and balanced fertilization are believed to be more sustainable than conventional intensive practices, mainly because they improve the nutrient status and quality of soil (Kaschuk et al., 2006) and then because they allow farmers to reduce costs and save time and fuel (Chèneby et al., 2009).

No-tillage, in conjunction with crop residues retention, is being increasingly adopted worldwide and has been strongly encouraged by the European Union agricultural policy for the expected agronomic and environmental benefits. Encouraging no-tillage practice may be a wise policy decision, but it still requires a robust and holistic scientific evaluation to understand induced changes in the short and long term, since there is the potential for promoting unproductive or even counterproductive changes in agricultural policies under different environment and management regimes (Six et al., 2004).

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Many studies have been conducted on the effects of different tillage practices on physical and chemical properties of the soil and on the composition of the resident microbial communities and their diversity (Ceja-Navarro et al., 2010; Mallory et al., 2011; Soane et al., 2012). Many authors report that soil total carbon, microbial biomass and activity significantly increase under no-tillage when compared with the conventional tillage systems (Kaschuk et al., 2006; Hungria et al., 2009; González-Chávez et al., 2010). The increment of soil organic matter affects the capacity of the soil to sustain crop growth and contributes to increase the stability of soil aggregates and reduce soil erosion (Kaschuk et al., 2006; Peixoto et al., 2006). Increasing soil carbon sequestration is also recommended in the perspective of mitigating climate changes by increasing carbon soil sink. However, in certain circumstances (i.e. in poorly aerated soils), no-tillage and N fertilization may lead to increased emissions of N_2O (Almaraz et al., 2009; Chèneby et al., 2009), or, it could result in the trade-off of one greenhouse gas (CO_2) for another (N_2O) (Six et al., 2004), with undesirable implications for global warming (Rochette et al., 2008).

Global research results regarding tillage and fertilizer management effects on soil N_2O emissions have been largely inconsistent (Venterea et al., 2005; Rochette et al., 2008; Omonode et al., 2011; Pelster et al., 2011) and the different responses may be due to the variability of many factors, such as weather, soil type, soil water content, types of fertilizer, timing and application method (Baggs et al., 2003; Bavin et al., 2009; Snyder et al., 2009).

Contrasting results have been obtained also about soil porosity: studies conducted by some authors (Pagliai and De Nobili, 1993; Tebrügge and Düring, 1999) highlighted that soil density was greater under no-tillage than under conventional tillage; Thomas et al. (1996) and Mallory et al. (2011) showed lower bulk density values under no-tillage, whilst Strudley et al. (2008) observed inconsistent effects of the tillage system on soil bulk density and total porosity. The different soil behaviour strongly depends on soil type and climatic conditions (Blevins et al., 1985).

The occurrence of soil compaction and wet condition can impact soil aeration and therefore induce obvious changes in the composition and activity of the microbial community and consequently, have significant effects on the soil fertility status, crop sustainability and greenhouse emissions (Ceja-Navarro et al., 2010). High soil microbial diversity confers an insurance against ecosystem malfunctioning under stress or disturbance (Brussaard et al., 2007). Although more detailed knowledge is required to determine the effects of microbial diversity on ecosystem functioning and stability, it is safer to adopt agricultural practices able to preserve and restore microbial functional diversity than practices that destroy or otherwise threaten it (Lupwayi et al., 1998).

Long-term experiments provide opportunities for improving our understanding of the effects of no-tillage systems on soil biological and physical properties. Nevertheless, the long-term effects of different tillage management systems have not been studied enough under Mediterranean conditions (Pagliai and De Nobili, 1993; De Vita et al., 2007; Casa and Lo Cascio, 2008; De Sanctis et al., 2012) and in particular the long-term impact of no-tillage on active bacterial communities is a topic which has not yet been sufficiently evaluated.

The aim of this study was to assess the modifications induced on soil porosity and active microbial communities (bacteria and denitrifiers) by comparing two tillage practices (no-tillage *vs*. conventional full inversion tillage) and two N fertilization regimes (with *vs*. without nitrogen fertilizer) in a long-term field experiment established on silty-clay soil in a coastal hill of Marche region (Central Italy).

2. Materials and methods

2.1. Experimental site and soil sampling

The long term experimental site was located at the farm "Pasquale Rosati" (43° 32′N, 13° 22′E) of the Polytechnic University of Marche, in Agugliano (Ancona, Italy) in the coastal hills (slope 10–15%) at 100 m a.s.l. (Farina et al., 2011; De Sanctis et al., 2012). The mean annual air temperature of the site is 15.3 °C, with average maximum temperatures in July–August (30.5 °C) and average minimum temperatures in January (2.9 °C). The mean annual precipitation is 786 mm (standard deviation 157 mm) concentrated between October and January and an aridity index (total rainfall/total reference evapotranspiration) of 0.76. The soil was classified as Calcaric Gleyc Cambisol (FAO, 2006) with a silty-clayey texture (Table 1).

The field experiment was set up in 1994 to test the multiple effects of long-term application of different tillage managements (conventional-, minimum-, or no-tillage) and three different rates of N fertilization in the context of a crop rotation based on durum wheat (*Triticum durum* Desf.) and sunflower (*Helianthus annuus* L.) (from 1994 to 2001) or maize (*Zea mais* L.) (from 2002 to 2007), using a factorial randomized complete block design with two replicates for each crop and repeated in the same plots every year. A duplicate set of plots was set up to allow for all crops to be present each year in the rotation.

In this study we analysed the effects of tillage (CT, conventional tillage with 40 cm deep full inversion ploughing; NT, no-tillage with sod seeding following chemical desiccation and chopping) and N fertilization (−N, unfertilized; +N, 90 kg ha^{-1} $year^{-1}$ N) on microbial communities and soil porosity. For each of the four treatments under comparison (CT −N; CT +N; NT −N; NT +N), duplicate plots were sampled in the durum wheat phase of the crop rotation.

In the sampled plots durum wheat cv. Grazia (ISEA) was sown on November 6, 2006 (250 kg ha^{-1} of seeds) and harvested on June 28, 2007. Mouldboard ploughing was conducted on October 10, 2006 followed by two harrowing on October 16 and November 6, 2006. No-tilled plots were sprayed with 2 L ha^{-1} of glyphosate (*N*-(phosphonomethyl)glycine active ingredient of the herbicide Roundup, Monsanto) on October 17, 2006. Where required, nitrogen fertilizer as ammonium nitrate (NH_4NO_3) was broadcasted in two rates on February 5 and April 17, 2007. Phosphorus fertilizer, as

Table 1
Soil physical and chemical properties of the topsoil (0–20 cm) in 2006. Means ($n = 4$) were calculated on the basis of tillage system (NT; CT) and fertilization regime (−N; +N). Values quoted are referred to kg air-dry soil or are expressed as % air-dry soil; numbers in parenthesis refer to standard errors. Different letters in a column between tillage or N-fertilization indicate significant differences at $P < 0.05$ between means. NT, no-tillage; CT, conventional tillage; −N, without nitrogen fertilization; +N, with nitrogen fertilization; FC, field capacity; WP, wilting point; SOC, soil organic carbon; CEC, cation exchange capacity.

	Clay (g kg^{-1})	Sand (g kg^{-1})	$CaCO_3$ (%)	FC (% vol 33 kPa)	WP (% vol 1500 kPa)	Tot N (g kg^{-1})	SOC (g kg^{-1})	CEC (meq 100 g^{-1})	pH (H_2O)	Olsen P (mg kg^{-1})
CT	467 (13)	95 (4)	30.9 (0.4)	41.7 (0.4)	27.6 (0.7)	0.86 (0.03) b	6.8 (0.7) b	24.3 (1.8)	8.3 (0.0) a	10.5 (1.0) b
NT	467 (13)	114 (4)	29.5 (0.4)	41.5 (0.4)	27.8 (0.7)	1.25 (0.03) a	10.7 (0.7) a	23.0 (1.8)	8.1 (0.0) b	16.3 (1.0) a
−N	445 (15)	107 (25)	29.7 (0.1) b	40.9 (0.4) b	26.5 (0.9)	1.01 (0.10)	8.3 (0.8)	23.6 (1.1)	8.2 (0.1)	13.5 (3.7)
+N	490 (15)	103 (25)	30.7 (0.1) a	42.3 (0.4) a	28.9 (0.9)	1.09 (0.10)	9.2 (0.8)	23.6 (1.1)	8.2 (0.1)	13.2 (3.7)