

Effects of tillage and residue management on soil microbial communities in North China

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ABSTRACT

The impacts of tillage system (conventional tillage and no-tillage) and residue management (0, 50, and 100%) on soil properties and soil microbial community structure were determined in the Fengqiu State Key Agro-Ecological Experimental Station, North China. The microbial community structure was investigated by phospholipid fatty acid (PLFA) profiles. The results showed that tillage had significant effects on soil properties and soil microbial communities. In no-tillage (NT), microbial biomass carbon (MBC), total N, microbial biomass carbon/soil organic carbon (MBC/SOC), total microbes, and arbuscular mycorrhiza fungi increased, while actinomycetes, G⁺/G⁻ bacteria ratio and monounsaturated fatty acids/saturated fatty acids (MUFA/STFA) decreased, compared with those in conventional tillage (CT). Residue had a significant positive effect on C/N ratio and MUFA/STFA. Canonical correspondence analysis indicated that tillage explained 76.1%, and residue management explained 0.6% of the variations in soil microbial communities, respectively. Soil microbial communities were significantly correlated with MBC, total N, C/N ratio and MBC/SOC. Among the six treatments, NT with 100% residue application obviously improved soil microbiological properties, and could be a proper management practice in the Huang-Huai-Hai Plain of China.

Keywords: arbuscular mycorrhiza fungi; microbial biomass carbon; no-tillage; PLFA; soil organic carbon

Conventional tillage consisting of plowing or disking can induce water and nutrient losses, and then soil degradation with low organic matter content and a fragile physical structure (Wang et al. 2007). However, conservation tillage, particularly no tillage, had positive effects on soil properties. No tillage could increase soil organic matter, preserve soil moisture, reduce erosion, moderate soil temperature and promote aggregate stability (Peigné et al. 2007, Alguacil et al. 2008, Ceja-Navarro et al. 2010). Compared with conventional tillage, conservation tillage can lead to the improvement of soil physical, chemical, and biological properties and play important roles in maintaining and improving soil quality.

Tillage and residue management affect not only soil properties but also soil microbial community. Soil microorganisms play essential roles in agro-ecosystem, and their changes will influence soil

nutrient cycling (He et al. 2007). No-tillage with residue application was proved to increase the soil microbial community (Gouaerts et al. 2007). In many cases, both bacteria and fungi were more abundant under no-tillage than conventional tillage (Helgason et al. 2009). In no-tillage systems, fungi domination was frequently found and the residue was mainly decomposed by the fungal community (Spedding et al. 2004, Gouaerts et al. 2007). Bacteria were generally considered to be the predominant decomposers of incorporated crop residues under conventional tillage (Nicolardot et al. 2007). However, more information is necessary to understand the effect of different tillage system combined with different residue quantity on soil microbial community. Therefore, the objectives of this research were to investigate the effects of tillage system (conventional tillage and no-tillage) and residue management (0, 50 and 100%) on

Supported by the Knowledge Innovation Project of the Chinese Academy of Sciences, Project No. KZCX2-YW-445, and by the National Key Basic Research Support Foundation of China, Project No. 2011CB100504.

soil properties and soil microbial communities, and to determine the relationships between soil properties and soil microbial communities in the Huang-Huai-Hai Plain of North China.

MATERIAL AND METHODS

Experimental site. The field experiment was established at the Fengqiu State Key Agro-ecological Experimental Station (35°01'N, 114°32'E), Henan province, located in the Huang-Huai-Hai Plain of China in 2007. The experiment was practiced as a rotation of winter wheat (early-October to mid-May) and maize (early-June to mid-September). Soil type is a fluvo-aquic soil with 11.13 g/kg organic matter, total nitrogen 1.39 g/kg, pH (H₂O) 8.24 and bulk density 1.16 g/cm² (Zhu et al. 2009, Zhang et al. 2012).

Experimental design. The experiment was a split-plot design which included conventional tillage (CT) and no-tillage (NT) as main plots and three subplots (no residue, 50% residue and 100% residue). Total 7.5 t/ha dry straws were applied as 100% residue treatment in wheat field. Chopped crop residues were ploughed into a depth of 23 cm in CT and mulched on the soil in NT, respectively. Each treatment plot (4 m × 100 m) was arranged randomly and replicated four times (Wang et al. 2011, Zhang et al. 2012).

Soil sampling and analyses. Ten random subsamples per plot were collected from the 0–20 cm depth and combined into one soil sample before wheat harvest on June 10, 2010. A total of 24 samples (6 treatments × 4 replicates = 24 samples) were collected. Soil samples were sieved (2 mm) and then were stored at 4°C and –20°C until analysis, respectively.

Soil organic carbon and total nitrogen were determined according to Ryan et al. (2001). Microbial biomass carbon (MBC) was measured by the chloroform fumigation and extraction method (Vance et al. 1987).

Analysis of soil microbial community structure was performed by phospholipid fatty acid (PLFA) profile based the method of White et al. (1979). Briefly, 2.0 g lyophilized and ground soil samples were used for extracting fatty acids. The extracted fatty acids were separated on a solid phase extraction column. Phospholipids were methylated, and the resulting fatty acid methyl esters were analyzed by Agilent 6850 (G2630A). The peaks were identified by MIDI identification software (MIDI Inc., Newark, DE), and quantified based on internal

standard methyl nonadecanoate (19:0). Biomarkers included 18:2 ω 6, 9 for fungi; i14:0, i15:0, a15:0, i16:0, i17:0, and 17:0 for Gram-positive bacteria; 18:1 ω 5c, 16:1 ω 7c, cy17:0, 18:1 ω 7c, and cy19:0 for Gram-negative bacteria; 10 Me16:0, 10 Me17:0 and Me18:0 for actinomycetes; and 16:1 ω 5c for arbuscular mycorrhizal fungi (AMF) (Bossio and Scow 1998, Bausenwein et al. 2008). The following fatty acids were chosen for calculating MUFA (mono-unsaturated fatty acids)/STFA (saturated fatty acids) ratio: 15:1 ω 6c, 16:1 ω 9c, 16:1 ω 7c, 17:1 ω 8c, 18:1 ω 9c, 18:1 ω 9t, 20:1 ω 9, 22:1 ω 9, 14:1, 15:1, 16:1, 17:1, 18:1, 20:1, and 22:1 for MUFA; and 10:0, 12:0, 14:0, 15:0, 16:0, 17:0, 18:0, 20:0, 21:0, 22:0, 23:0, and 24:0 for STFA (Bausenwein et al. 2008).

Statistical analysis. All data were calculated based on oven-dried (105°C) weight. The influence of tillage and residue managements on soil properties and PLFA profiles were evaluated with General linear model (GLM) procedure for a split-plot design. Pearson correlation analysis was used to analyze the correlation between PLFA profiles and soil properties. All statistical analyses were performed with SPSS software package release 17.0 (SPSS Inc., Chicago, USA). Canonical correspondence analysis (CCA) was used to analyze the data of PLFA profiles with the Canoco software (Ter Braak and Smilauer 2002).

RESULTS AND DISCUSSIONS

Soil properties. As shown in Table 1, tillage treatments had significant effects on MBC, total N, and MBC/SOC ($P < 0.01$). Compared with CT, MBC, MBC/SOC, and total N increased in NT. Residue treatments had a significant effect on C/N ratio, and CT with 100% residue significantly increased C/N ratio ($P < 0.05$). In NT, SOC increased slightly but not significantly with increasing quantity of residue.

The beneficial effects of conservation tillage on the accumulation of soil C and N were proved by Spedding et al. (2004). Our results showed that NT increased about 2 times MBC and slightly increased soil total N, compared with CT, but did not significantly increase SOC, which was different with previous studies. It could be caused by many factors such as the no-tillage years (Zhang and Song 2003), soil depth (Bausenwein et al. 2008, Treonis et al. 2010) and soil type (Helgason et al. 2009). Zhang and Song (2003) claimed that SOC needed a longer period to response to no-tillage and residue amendment as compared to MBC. Because

Table 1. Soil properties of different tillage and residue treatments

Treatments	SOC (g/kg)	MBC (mg/kg)	MBC/SOC (%)	Total N (g/kg)	C/N ratio
NT 0	6.77	134.87 ^a	1.74 ^a	0.86 ^{ab}	7.91 ^b
NT 50	7.08	113.15 ^a	1.27 ^b	0.94 ^a	7.59 ^b
NT 100	7.33	112.31 ^a	1.15 ^b	0.92 ^a	7.90 ^b
CT 0	7.80	63.49 ^b	0.61 ^c	0.84 ^{ab}	9.28 ^{ab}
CT 50	5.39	55.50 ^b	0.54 ^c	0.77 ^b	6.93 ^b
CT 100	8.05	62.67 ^b	0.60 ^c	0.83 ^{ab}	9.73 ^a
T (tillage)	ns	***	**	**	ns
R (residue)	ns	ns	ns	ns	*
T × R	ns	ns	ns	ns	ns

NT – no-tillage; CT – conventional tillage; SOC – soil organic carbon; MBC – microbial biomass carbon. Values with different letters represent significant difference. *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns – non-significant

of its quicker turnover rate, the changes of MBC in the short term are more apparent than SOC. Although residue and no-tillage had no significant effect on SOC, an increasing trend of SOC with increasing residue quantity was observed in NT soils, which suggested the beneficial co-effects of no-tillage and residue.

Microbial community structure. PLFA profiles showed that tillage treatments had significant effects on soil total microbes, AMF, G^+/G^- bacteria ratio and MUFA/STFA ($P < 0.05$) (Table 2). NT increased the total microbes and AMF, but decreased the actinomycetes, MUFA/STFA and the G^+/G^- bacteria ratio compared with CT. Residue treatments had no significant effect on the PLFA profiles except MUFA/STFA ratio. In CT, the MUFA/STFA ratio significantly increased in 100% residue application compared with oth-

ers ($P < 0.05$). Interaction of tillage and residue had no significant effect on PLFA profiles. Total microbes, total bacteria, G^+ bacteria, G^- bacteria, and actinomycetes in NT gradually increased with increasing residue quantity.

The PLFA data from six treatments were subjected to canonical correspondence analysis (CCA) (Figure 1). 76.1% and 0.6% of the total variance could be explained by the first axis and the second axes, respectively. Both the first canonical axis ($P = 0.002$, $F = 57.19$) and the sum of all canonical axes ($P = 0.002$, $F = 11.90$) were significant by the Monte Carlo test. There was an obvious separation between NT and CT by the first axis. In Figure 1, it was obviously shown that fungi, especially arbuscular mycorrhizal fungi (AMF) enriched in NT, but bacteria (G^+ bacteria, G^- bacteria and actinomycetes) in CT.

Table 2. PLFA (phospholipid fatty acid) profiles under different tillage and residue treatments

PLFA (nmol/g soil)	NT 0	NT 50	NT 100	CT 0	CT 50	CT 100	T	R	T × R
Total microbes	99.60 ± 3.01 ^{ab}	105.89 ± 11.20 ^{ab}	113.02 ± 3.44 ^a	87.86 ± 2.59 ^b	79.07 ± 9.59 ^b	80.80 ± 3.77 ^b	**	ns	ns
Total bacteria	18.06 ± 1.27	20.56 ± 2.86	24.42 ± 1.97	22.24 ± 1.13	19.16 ± 2.89	22.43 ± 0.23	ns	ns	ns
G^+ bacteria	8.19 ± 0.89	9.56 ± 1.43	11.63 ± 0.91	11.55 ± 0.54	10.26 ± 1.66	11.92 ± 0.24	ns	ns	ns
G^- bacteria	9.87 ± 0.69	11.01 ± 1.48	12.79 ± 1.08	10.69 ± 0.70	8.91 ± 1.27	10.51 ± 0.14	ns	ns	ns
Actinomycetes	3.06 ± 0.34 ^b	3.70 ± 0.73 ^b	4.57 ± 0.39 ^{ab}	5.05 ± 0.30 ^{ab}	4.30 ± 0.77 ^{ab}	5.23 ± 0.09 ^a	*	ns	ns
Fungi	1.87 ± 0.08	2.28 ± 0.52	2.18 ± 0.13	1.80 ± 0.08	1.59 ± 0.30	1.71 ± 0.04	ns	ns	ns
AMF	7.50 ± 0.70 ^b	12.89 ± 2.89 ^a	10.53 ± 2.55 ^{ab}	3.56 ± 0.91 ^{bc}	2.29 ± 0.52 ^c	2.68 ± 0.15 ^{bc}	**	ns	ns
G^+/G^- ratio	0.84 ± 0.08 ^b	0.86 ± 0.05 ^b	0.91 ± 0.02 ^b	1.09 ± 0.06 ^a	1.15 ± 0.06 ^a	1.14 ± 0.03 ^a	***	ns	ns
MUFA/STFA	0.37 ± 0.01 ^c	0.48 ± 0.05 ^c	0.51 ± 0.05 ^c	0.70 ± 0.06 ^{bc}	0.72 ± 0.10 ^b	0.94 ± 0.10 ^a	**	*	ns

AMF – arbuscular mycorrhiza fungi; G^+/G^- ratio – G^+ bacteria/ G^- bacteria ratio; MUFA/STFA mono-unsaturated fatty acids/saturated fatty acids; NT – no-tillage; CT – conventional tillage; R – residue; T – tillage. Values with different letters represent significant difference; *** $P < 0.001$; ** $P < 0.01$; * $P < 0.05$; ns – non-significant

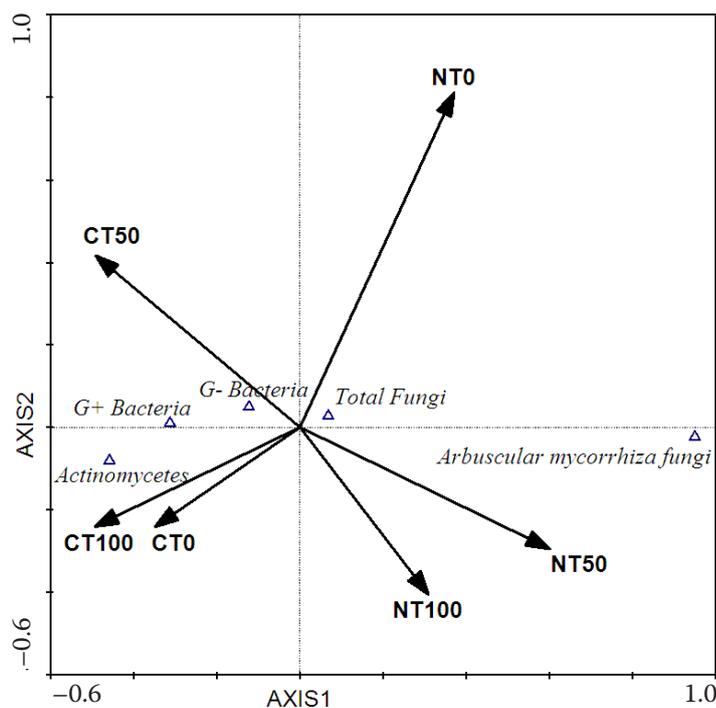


Figure 1. Canonical correspondence analysis of soil PLFA profiles and different treatments. NT0, NT50, NT100: no-tillage with no residue, 50% and 100% residue, respectively; CT0, CT50, CT100: conventional tillage with no residue, 50% and 100% residue, respectively

PLFA profile was adopted to assess the soil microbial community composition under different field management. Simmons and Coleman (2008) indicated that NT with residue application had positive effects on soil microbes, among which fungi were dominant. It was reported that tillage could significantly decrease soil fungi by physically disrupting their hyphal networks and/or by affecting soil moisture regime, resulting in a decreased fungal biomass (Simmons and Coleman 2008, Helgason et al. 2009). In this study, the abundance of fungi especially AMF increased significantly in NT, which was in agreement with previous studies (Spedding et al. 2004, Alguacil et al. 2008).

AMF are a main component of soil microbes in most agro-ecosystems and are necessary for many plants (Parniske 2008). The AMF abundance was often correlated with the mass of macroaggregates which physically protect SOC (Six et al. 2006). AMF can produce glycoprotein (e.g., glomalin) to bind soil aggregates, or use their hyphae acting as a web to encase macroaggregates. Therefore, the soil management practices that can increase or maintain soil AMF abundance, such as NT, could result in greater soil C sequestration (White and Rice 2009). Our results also proved that the AMF abundance had a significant positive correlation with MBC, MBC/SOC and total N.

Many reports argued that residue could increase the abundance of bacteria in NT (Hammesfahr et al. 2008, Ceja-Navarro et al. 2010). However, we did not find any significant positive effects of residue on the abundance of bacteria and there was only

an increasing trend for the bacteria in NT with increasing residue quantity. Our observation was agreed with other researchers, who found that tillage affected the distribution of residue and promoted their mixing with soil, which also enhanced labile substrate. Then bacteria dominate due to their more efficient ability to break down labile carbon sources than fungi (Gouaerts et al. 2007). On the other hand, soil condition with lower temperature on the soil surface was formed in NT, which was not comfortable for bacteria growth (Simmons and Coleman 2008, Helgason et al. 2009).

MUFA/STFA ratio was adopted to identify soil aeration condition, and found to be a sensitive indicator for agricultural management (Bossio and Scow 1998). The significantly low MUFA/STFA ratio in NT soils indicated that an anaerobic condition was formed, which might give stress to soil bacteria and restrain their growth (Spedding et al. 2004, Gouaerts et al. 2007). G^+/G^- bacteria ratio was used as an indicator of soil starvation stress (Hammesfahr et al. 2008). The higher G^+/G^- bacteria ratio in CT suggested that poor nutrients in CT, while fertile in NT.

Correlations between PLFA profiles and soil properties. Pearson correlation analysis between soil PLFA profiles and all soil properties showed that soil PLFA profiles significantly correlated with MBC, total N, C/N and MBC/SOC ($P < 0.05$) (Table 3). Total microbes, G^- bacteria and AMF had positive correlation with soil total N ($P < 0.05$). MBC positively correlated with total microbes and AMF, but negatively with actinomycetes

Table 3. Correlation analysis between soil PLFA (phospholipid fatty acid) profiles and soil properties

PLFA (nmol/g soil)	MBC	Total N	C/N ratio	MBC/SOC
Total microbes	0.511*	0.542**	-0.665**	0.544**
G ⁺ bacteria	ns	ns	ns	-0.477*
G ⁻ bacteria	ns	0.506*	ns	ns
Actinomycetes	-0.498*	ns	0.451*	-0.570**
Fungi	ns	ns	-0.467*	0.432*
AMF	0.674**	0.596**	-0.708**	0.704**
MUFA/STFA	-0.579**	ns	0.724**	-0.662**
G ⁺ /G ⁻ ratio	-0.804**	ns	0.819**	-0.845**

MBC/SOC – microbial biomass C/soil organic C; C/N ratio – SOC/Total N; AMF – arbuscular mycorrhiza fungi; G⁺/G⁻ ratio – G⁺ bacteria/G⁻ bacteria ratio; MUFA/STFA – monounsaturated fatty acids/saturated fatty acids; ** $P < 0.01$; * $P < 0.05$; ns – non-significant

($P < 0.05$). MBC/SOC had a positive correlation with total microbes, fungi and AMF, and a negative correlation with G⁺ bacteria and actinomycetes ($P < 0.05$). For soil C/N, there was a negative correlation with total microbes, fungi and AMF, but a positive correlation with actinomycetes ($P < 0.05$). Both MUFA/STFA and G⁺/G⁻ bacteria ratio positively correlated to C/N, and negatively correlated to MBC and MBC/SOC ($P < 0.01$).

In conclusion, among the six treatments, NT with 100% residue application obviously improved soil microbiological properties, and could be a proper management practice in the Huang-Huai-Hai Plain of China.

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Received on July 22, 2011

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