



Soil microbial community dynamics over a maize (*Zea mays* L.) growing season under conventional- and no-tillage practices in a rainfed agroecosystem

Bin Zhang^{a,b}, Hongbo He^a, Xueli Ding^a, Xudong Zhang^{a,c,*}, Xiaoping Zhang^d, Xueming Yang^e, Timothy R. Filley^f

^aState Key Laboratory of Forest and Soil Ecology, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110164, PR China

^bGraduate University of Chinese Academy of Sciences, Beijing 100049, PR China

^cNational Field Observation and Research Station of Shenyang Agroecosystems, Shenyang 110016, PR China

^dNortheast Institute of Geography and Agroecology, Chinese Academy of Sciences, Changchun 130012, PR China

^eGreenhouse and Processing Crops Research Centre, Agriculture and Agri-Food Canada, Harrow NOR 1G0, Canada

^fDepartment of Earth and Atmospheric Sciences, Purdue University, West Lafayette, IN 47907, USA

ARTICLE INFO

Article history:

Received 11 November 2011

Received in revised form 17 May 2012

Accepted 19 May 2012

Keywords:

Microorganism

Temporal change

Conservation tillage

Culture-independent technique

Mollisol

ABSTRACT

Tillage practices affect soil microorganisms, which in turn influence many processes essential to the function and sustainability of soil. In this study, the changes in soil microbial biomass and community composition in response to conventional tillage (CT, moldboard plowing and post-harvest residue removal) and no-tillage (NT) practices were examined during a maize (*Zea mays* L.) growing season in a clay loam soil (Typic Hapludoll) in northeastern China. Soil samples were taken in May, June, July, August, and September of 2008 at 0–5, 5–10, and 10–20 cm depths. Microbial communities were characterized by phospholipid fatty acid (PLFA) analysis. While microbial biomass increased at the beginning then decreased toward the end of the growing season in CT soils, it showed the opposite trend in NT soils. Microbial community structure showed better distinction among sampling months than between tillage practices. These results suggest that seasonal variations in soil microbial communities could be greater than changes associated with tillage treatments. However, microbial biomass accumulation was tillage dependent. On average, NT treatment resulted in 21% higher microbial biomass in 0–5 cm depth than CT treatment ($P < 0.05$). Higher fungi to bacteria ratio was also observed under NT than CT treatment at both the 0–5 and 5–10 cm sampling depths. These data demonstrate that examining the effect of management practices on soil quality based on soil microbial communities should consider seasonal changes in the environmental properties. It is strongly recommended that NT practice should be adopted as an effective component of an overall strategy to improve soil quality and sustainability in northeastern China.

© 2012 Elsevier B.V. All rights reserved.

1. Introduction

Soil microorganisms are of great importance to agroecosystem function and sustainability through their contributions to the decomposition and dynamics of soil organic matter (SOM) and biogeochemical cycling of soil nutrients (Paul and Clark, 1989), all of which are fairly related to soil management practices which alter soil environment and affect soil processes (Schimel, 1995). Conventional tillage (CT), including moldboard plowing and post-harvest crop residue removal, has been the dominant management practice in northeastern China during the 20th century. However,

these intensive cultivation and residue removal practices have led to a significant loss of SOM (55%) and have severely degraded the soils (Liu et al., 2003, 2010). Moreover, the on-going soil degradation in this region has threatened sustainable crop production and even national food security (Liu et al., 2010). Therefore, no-tillage (NT) was introduced in this region as an alternative to maintain, or possibly improve soil quality.

The impact of management practices on the flow of carbon (C) through ecosystems is largely mediated by soil microorganisms (van Groenigen et al., 2010). Since microbial communities have the ability to respond rapidly to changing environmental conditions by modifying biomass and community composition (Schloter et al., 2003), microbial characteristics are potentially valuable indicators for assessing the effect of agricultural management practices (Zelles, 1999; Gil-Sotres et al., 2005). The surface accumulation of microbial biomass under NT has been demonstrated in many studies (Feng et al., 2003; Spedding et al., 2004; Minoshima et al.,

* Corresponding author at: State Key Laboratory of Forest and Soil Ecology, Institute of Applied Ecology, Chinese Academy of Sciences, Shenyang 110016, PR China. Tel.: +86 24 83970375; fax: +86 24 83970376.

E-mail addresses: hehongbo@iae.ac.cn (H. He), xdzhang@iae.ac.cn (X. Zhang).

2007; Helgason et al., 2009). However, this phenomenon is not observed in some reports (Drijber et al., 2000; Carpenter-Boggs et al., 2003), indicating that the underlying mechanisms that drive biomass accumulation in NT are linked to a wider range of factors than simple tillage treatment. Microbial community composition can also resist simple prediction as soil fungi and bacteria respond differently to agricultural management practices. In general, CT promotes bacterial growth in soil microbial communities while NT, with crop residues retained on soil surface, encourages fungal growth and the temporary immobilization of nutrients (Pankhurst et al., 2002). A number of studies have demonstrated such an increase in the proportion of fungal biomass in NT system (Beare et al., 1997; Frey et al., 1999); however, in other studies there were either no effects or decreases in the relative fungal abundance under NT treatment (Spedding et al., 2004; Helgason et al., 2009).

Examining the seasonal fluctuations in soil microbial biomass and composition could provide a more complete perspective in understanding the influence of tillage practices on soil microbial communities. It is suggested that soil microbial communities are primarily influenced by plant input during the crop cycle, but with important controls governed by the physicochemical environment resulting from tillage treatments during the fallow period (Drijber et al., 2000; Feng et al., 2003). Petersen et al. (2002) conducted studies during the growing season of spring wheat (*Triticum aestivum*) and found limited tillage effects on soil microbial communities. Spedding et al. (2004) concluded that the frequent changes in soil conditions throughout the maize (*Zea mays* L.) growing season could promote shifts in soil microbial communities. However, different climates and soil types could lead to substantially different conditions in water and resource availability in the soil rooting zone, which may have resulted in different microbial community dynamics.

The objective of this study was to evaluate the variations in soil microbial biomass and community composition in response to CT and NT practices during a maize growing season at varying soil depths in a rainfed agroecosystem in northeastern China. Phospholipid fatty acid (PLFA) analysis was used because it is widely accepted as a sensitive tool to indicate viable microbial biomass and provide a fingerprint for the microbial community composition (Frostegård and Bååth, 1996). Examining the dynamics of soil microbial communities at several intervals throughout the maize growing season allows for the determination of how, and at what magnitude, seasonal fluctuations in soil temperature, water content, and crop growth influence soil microbial communities. We hypothesized that: (1) NT could result in higher microbial biomass than CT and a community shift toward fungi would occur in the surface layer; (2) differences in microbial biomass and community composition between CT and NT treatments would be consistent with respect to time of sampling.

2. Materials and methods

2.1. Site description

This study was conducted on a long-term tillage trial initiated in fall 2001 at the experimental station (44°12'N, 125°33'E) of

Northeast Institute of Geography and Agroecology, Chinese Academy of Sciences, in Dehui County, Jilin Province, China. According to the Köppen–Geiger climate classification, the climate of this region is humid continental (Peel et al., 2007). The mean annual temperature is 4.4 °C. The mean monthly temperatures range from –19.5 °C in January to 24.5 °C in July from 2001 to 2007. The mean annual precipitation is 520 mm, with about 70% occurring in June, July, and August, and an average monthly rainfall of 82 mm during crop growing season. The clay loam soil was classified as Typic Hapludoll according to the USDA Soil Taxonomy (Soil Survey Staff, 2010). Selected soil properties at the initiation of the experiment were presented in Table 1. Before the establishment of this study site, the land had been used for monoculture maize production under CT management for over ten years.

The experiment was a randomized complete block design with four blocks and twelve treatments consisting of different tillage and cropping systems. Individual treatment plots were 5.2 m × 20 m. There was a 5 m × 62.4 m buffer zone at both length sides of each block and a 5-m laneway between blocks. Two monoculture maize systems subjected to CT and NT treatments were included in this study. CT treatment involved one moldboard plowing (to a depth of 20 cm) in early October after crop harvest, one disking (7.5–10 cm depth) and field cultivation in late April prior to planting, and one manual hoeing to control weeds. All aboveground crop residues were removed before plowing in the CT plots. Under NT practices, there was no soil disturbance except for planting, crop residues were retained on soil surface, and weeds were controlled by herbicides such as Acetochlor and 2,4-dichlorophenoxyacetic acid. Maize was planted on May 2nd, 2008 at a rate of 49,000 seeds ha⁻¹ in 75-cm row width and harvested on September 30th, 2008. Each year, 100 kg N ha⁻¹, 45.5 kg P ha⁻¹, and 78 kg K ha⁻¹ were applied to the maize as a starter fertilizer during the planting, and 50 kg N ha⁻¹ was used as the top dressing at the sixth leaf stage for maize. Average maize yields from 2001 to 2007 were 10,627 and 10,106 kg hm⁻² for NT and CT treatments, respectively (Ding et al., 2011).

2.2. Soil sampling and methods

Seven randomized soil cores (2.64 cm in diameter) per plot were sampled from the 0 to 20 cm depth with a hand auger on the following dates in 2008: May 31st, June 29th, July 31st, August 30th, and September 30th. The soil cores were cut into three segments: 0–5, 5–10, and 10–20 cm. The soil segments in the same plot were composited on a depth basis, placed in plastic bags in the field, and kept cool until processed in the laboratory within 10 h after collection. The soils were passed through a 2-mm sieve, homogenized, and stored at 4 °C. All visible root and fresh litter material were removed from samples prior to sieving. The field-moist subsamples used for PLFA analysis were freeze-dried and stored in a desiccator for no more than three weeks prior to extraction.

Soil total C and nitrogen (N) were determined by dry combustion on ground samples (100-mesh) on a CN2000 analyzer (LECO Corporation, MI, USA). Because these soil samples are free of carbonates, the total C content is equivalent to the soil organic

Table 1
Characteristics of the soil at the initiation of the experiment (fall 2001).^a

Depth (cm)	Soil organic carbon (g kg ⁻¹)	Particle size distribution (g kg ⁻¹)			pH	Bulk density (g cm ⁻³)
		Clay	Silt	Sand		
0–5	16.5	360	240	400	6.48	1.24
5–10	16.3	358	238	404	6.45	1.38
10–20	16.1	357	243	400	6.51	1.36

^a Data from Liang et al. (2007).

carbon (SOC) content. Soil microbial biomass C (MBC) was determined by chloroform fumigation–extraction technique (Vance et al., 1987). The organic C in the K_2SO_4 extracts was measured as CO_2 by infrared absorption after combustion at $850^\circ C$ using a TOC analyzer (Analytik Jena AG, Germany). Microbial biomass C was calculated as the difference in C concentration between the fumigated and unfumigated samples with an efficiency constant (k_{EC}) of 0.45 (Wu et al., 1990). Soil temperature at 10-cm depth was measured with a bent stem thermometer. This thermometer is similar to regular alcohol filled glass thermometers except that the stem is bent approximately 45° allowing the bulb to be placed horizontally in the soil, with the graduated section protruding from the soil to allow the temperature to be read manually. Volumetric soil water content at 0–22 cm depth was measured by a portable time-domain reflectometer (TDR) probe (Hydrosense System, IMKO, Germany).

2.3. Phospholipid fatty acid analysis

PLFA extraction was conducted for each sample following the procedure of Bligh and Dyer (1959) after modifications by Bossio et al. (1998). Briefly, lipids were extracted in a single-phase chloroform-methanol-citrate buffer system. Phospholipids were separated from neutral lipids and glycolipids on solid phase extraction columns (Supelco Inc., Bellefonte, PA). After methylation of the polar lipids, PLFA methyl esters were analyzed on an Agilent 6890A gas chromatography (GC, Agilent Tech. Co., USA) equipped with an HP-5 capillary column ($30\text{ m} \times 0.32\text{ mm} \times 0.25\text{ }\mu\text{m}$) and a flame ionization detector (FID). Nonadecanoic acid methyl ester (19:0, Sigma) was added as an internal standard when the samples were dissolved in $150\text{ }\mu\text{L}$ of hexane before GC analysis. Super purified N_2 was used as the carrier gas with a flow rate of 0.8 mL min^{-1} . The Supelco 37 Component FAME Mix and Supelco Bacterial Acid Methyl Esters (Supelco, Bellefonte, USA) were used for peak identification and quantification. PLFAs that were detected in less than 1% of the samples were excluded from the data set. Thirteen PLFAs (14:0, i15:0, a15:0, 15:0, i16:0, 16:1 ω 7c, 16:0, i17:0, cy17:0, 18:2 ω 6,9, 18:1 ω 9c, 18:1 ω 9t, 18:0) were used for data analysis. The fatty acid signatures i15:0, a15:0, i16:0, 16:1 ω 7c, i17:0, and cy17:0, which are considered to be of bacterial origin (Frostegård and Bååth, 1996), were used to represent bacterial PLFAs. The fatty acid signature 18:2 ω 6,9, which is known to correlate well with ergosterol, was used as an indicator of fungal PLFA (Frostegård and Bååth, 1996). The summation of all PLFAs was used to represent the total microbial lipid biomass. The ratio of fungal to bacterial PLFAs (F/B) was used as an indicator of changes in the relative abundance of these two microbial groups.

2.4. Statistical analysis

Repeated measures analysis of variance (RMANOVA) was used to determine the effect of tillage and sampling time on soil MBC as

well as the sums and ratios of PLFAs of various microbial groups. Bonferroni's test was used for mean comparisons at a 5% probability level. Student's *t*-test was used to illustrate the differences between the two tillage treatments on the figures. To explore the variations in soil microbial community composition, the standardized mole percentages of individual PLFAs were subjected to principal component analysis (PCA) with Kaiser's rule to select the components. The factor loading scores for the individual PLFAs were used to assess the relative importance of individual PLFAs in the calculation of the principal component axes. Statistical analysis was performed with the software package SPSS 13.0 for Windows (SPSS Inc., Chicago, USA).

3. Results

3.1. Soil characteristics

Because SOC and total N were fairly constant throughout the growing season, we presented the mean values among sampling months for both treatments (Table 2, $n = 20$). On average, NT soils contained 34% and 30% higher SOC and total N, respectively, than CT soils at the 0–5 cm depth ($P < 0.05$, Table 2). No differences were found in SOC and total N between the two tillage treatments at the 5–10 and 10–20 cm depths (Table 2). Soil water content was significantly higher in six of the nine measurements under NT than CT treatment ($P < 0.05$, Fig. 1). Soil temperature was slightly lower under NT practice than CT treatment (Fig. 1).

3.2. Soil microbial biomass C and total PLFAs

Tillage treatments, months and their interactions significantly affected the MBC content in the 0–5 cm depth (Table 3). NT soils contained 45%, 49%, 17%, and 34% higher MBC than CT soils in May, June, August, and September, respectively (Table 2). The tillage \times sampling month interaction at the 0–5 cm depth apparently resulted from the soils sampled in July where CT had greater MBC levels than NT (Tables 2 and 3). At the 5–10 cm depth, higher MBC content was found in NT soils than CT soils sampled from June and September, while the opposite trend was observed in other sampling months (Tables 2 and 3). At the 10–20 cm depth, MBC was generally higher in NT than CT soils in all sampling months except August, and increased significantly at the end of the growing season under both tillage treatments (Tables 2 and 3).

Total PLFAs were significantly affected by tillage and sampling month at the 0–5 cm depth as well having a significant interaction between tillage and month (Table 3). Total PLFAs in NT soils were 80% higher in May, 20% higher in June, 17% lower in July, and 60% higher in September than the corresponding CT soils ($P < 0.05$, Fig. 2). Total PLFAs increased from the beginning of the growing season to the maximum in July and then decreased toward the end of the growing season under CT treatment; however, they were higher in May and September than other sampling months under

Table 2

Soil organic carbon and total nitrogen averaged across a maize growing season and soil microbial biomass carbon sampled at different months of the growing season under conventional- and no-tillage practices.

Depth (cm)	Tillage ^a	Soil organic carbon (g kg^{-1})	Total nitrogen (g kg^{-1})	Soil microbial biomass carbon (mg kg^{-1})				
				May	June	July	August	September
0–5 cm	CT	15.9 ^a	1.43a	145	144	229	173	163
	NT	21.3 ^b	1.86b	210	216	180	203	218
5–10 cm	CT	16.1a	1.42a	132	137	152	145	154
	NT	16.8a	1.50a	122	155	121	123	185
10–20 cm	CT	15.6a	1.22a	104	117	105	138	144
	NT	16.2a	1.31a	117	135	129	137	181

^a CT, conventional tillage; NT, no-tillage.

^b Values followed by different letters between tillage treatments are significantly different ($P < 0.05$).

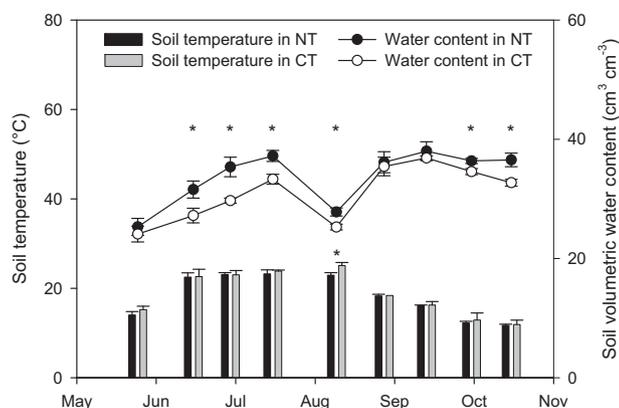


Fig. 1. Soil temperature (10 cm depth) and volumetric water content (0–22 cm depth) under conventional tillage (CT) and no-tillage (NT) practices from May 23rd to October 14th in 2008. Asterisks indicate significant differences ($P < 0.05$) between tillage treatments. Error bars represent standard deviations ($n = 3$).

NT treatment (Table 3, Fig. 2). The total PLFA concentrations were lower in the 5–10 cm relative to 0–5 cm depth, with an average decrease of 33% in NT soils and 23% in CT soils. At the 5–10 cm soil layer, the overall treatment effect was not significant despite NT soils contained 26% and 37% higher total PLFAs than the CT soils in August and September, respectively (Table 3, Fig. 2). The greatest concentrations of total PLFAs in the 5–10 cm depth were found in July for both CT and NT soils. Under CT management, the seasonal dynamic pattern in total PLFAs at 5–10 cm depth was similar to that at 0–5 cm depth. For the 10–20 cm depth, total PLFAs showed further decline, and stayed almost constant regardless of tillage treatments or sampling months (Table 3, Fig. 2). Total PLFAs were significantly correlated with MBC ($r = 0.78$, $P < 0.001$).

3.3. Fungal and bacterial PLFAs

Tillage treatment, sampling month and their interaction had significant influences on the concentrations of fungal and bacterial PLFAs (Table 3, Figs. 3 and 4). The NT soils contained about 214%, 94%, 25%, and 155% higher fungal PLFAs than the CT soils at the 0–5 cm depth in May, June, August, and September, respectively ($P < 0.05$, Fig. 3). The concentrations of bacterial PLFAs at 0–5 cm depth were also significantly higher under the NT than CT treatment in May, June, and September, with an increase of 74%, 25%, and 47%, respectively ($P < 0.05$, Fig. 4). Both fungal and bacterial PLFAs exhibited similar temporal trends as that of the total PLFAs at 0–5 cm depth (Figs. 2–4). Additionally, both fungal

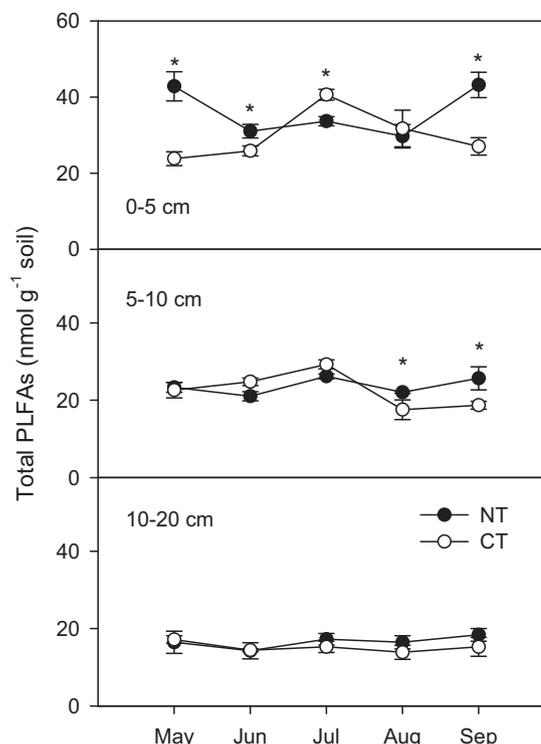


Fig. 2. Total phospholipid fatty acids (PLFAs) under conventional tillage (CT) and no-tillage (NT) practices in 0–5, 5–10, and 10–20 cm soil depths over a maize growing season. Asterisks indicate significant difference ($P < 0.05$) between tillage treatments. Error bars represent standard deviations ($n = 4$).

and bacterial PLFAs declined significantly at the 5–10 cm relative to 0–5 cm depth under NT treatment ($P < 0.05$), with the largest decrease of 78% for fungal PLFAs in September and 41% for bacterial PLFAs in May when compared to the 0–5 cm depth (Figs. 3 and 4). At 5–10 cm depth in May, NT soils contained twice the concentration of fungal PLFAs as the CT soils ($P < 0.05$, Fig. 3). However, greater bacterial PLFAs were found in NT soils than CT soils collected from August and September whereas the reverse pattern was observed in June ($P < 0.05$, Fig. 4). At the 5–10 cm depth, fungal PLFAs in NT soils showed a gradual decline through the maize growing season (Fig. 3). No significant differences were found in the concentrations of fungal and bacterial PLFAs between the two tillage practices at the 10–20 cm soil depth except September sampled bacterial PLFAs (Figs. 3 and 4).

Table 3

Significance of tillage and sampling month on soil microbial biomass carbon, total phospholipid fatty acids (PLFAs), fungal and bacterial PLFAs, and the ratio of fungal to bacterial PLFAs (F/B) in three soil depths.

Source of variation	Microbial biomass carbon	Total PLFAs	Fungal PLFAs	Bacterial PLFAs	F/B
0–5 cm					
Tillage	***	**	***	**	***
Month	**	***	***	*	***
Tillage × month	***	***	***	***	**
5–10 cm					
Tillage	ns	ns	**	ns	**
Month	**	***	***	***	***
Tillage × month	*	***	***	***	***
10–20 cm					
Tillage	*	ns	ns	*	ns
Month	***	ns	ns	ns	***
Tillage × month	*	ns	ns	ns	*

ns, not statistically significant at $P < 0.05$.

* Statistically significant at $P < 0.05$.

** Statistically significant at $P < 0.01$.

*** Statistically significant at $P < 0.001$.

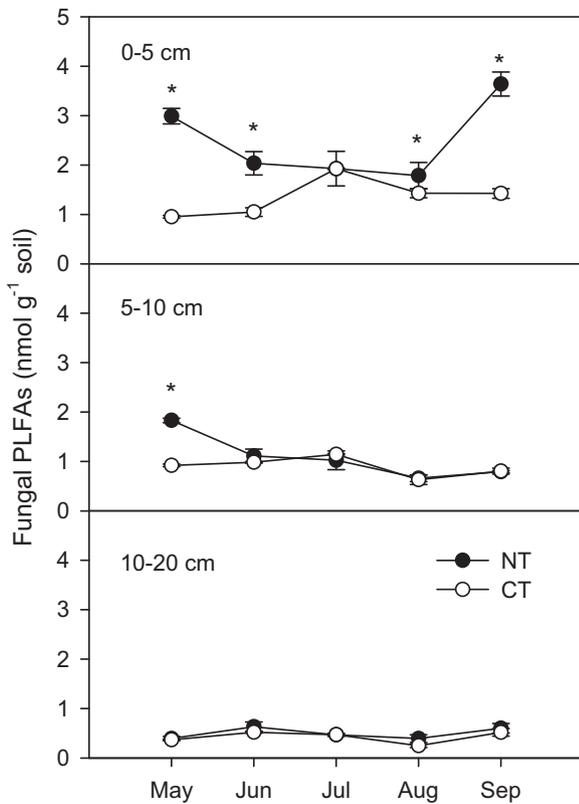


Fig. 3. Fungal phospholipid fatty acids (PLFAs) under conventional tillage (CT) and no-tillage (NT) practices in 0–5, 5–10, and 10–20 cm soil depths over a maize growing season. Asterisks indicate significant difference ($P < 0.05$) between tillage treatments. Error bars represent standard deviations ($n = 4$).

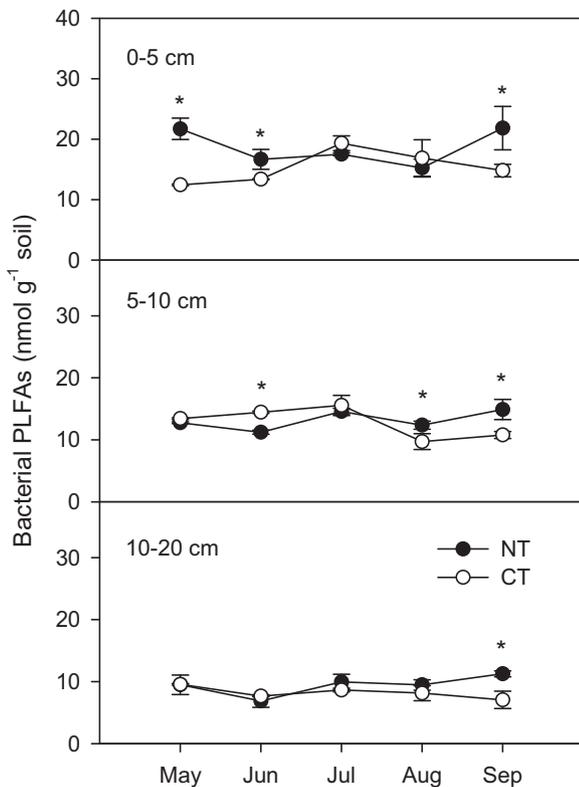


Fig. 4. Bacterial phospholipid fatty acids (PLFAs) under conventional tillage (CT) and no-tillage (NT) practices in 0–5, 5–10, and 10–20 cm soil depths over a maize growing season. Asterisks indicate significant difference ($P < 0.05$) between tillage treatments. Error bars represent standard deviations ($n = 4$).

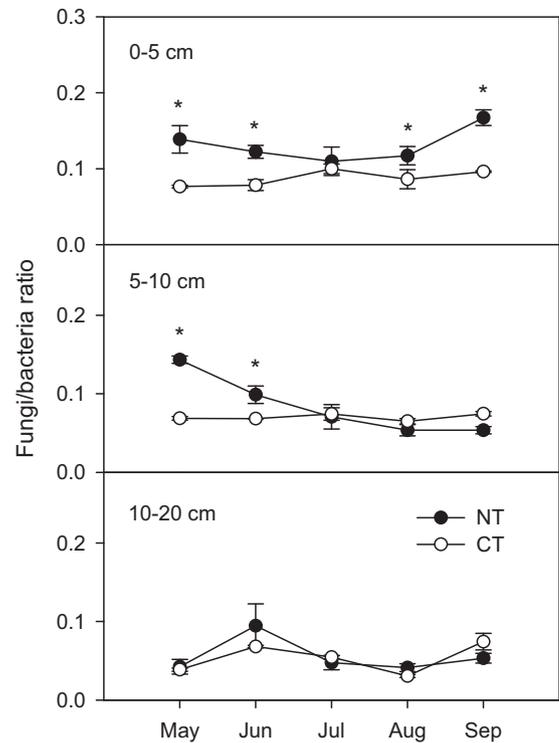


Fig. 5. The ratios of fungal to bacterial PLFAs under conventional tillage (CT) and no-tillage (NT) practices in 0–5, 5–10, and 10–20 cm soil depths over a maize growing season. Asterisks indicate significant difference ($P < 0.05$) between tillage treatments. Error bars represent standard deviations ($n = 4$).

Tillage treatments had a significant effect on F/B ratio at both 0–5 and 5–10 cm depths (Table 3). In the 0–5 cm depth, NT soils had significantly ($P < 0.05$) higher F/B ratios than the corresponding CT soils over the entire growing period except July (Fig. 5). For the 5–10 cm depth, the F/B ratios were significantly higher in May and June under NT practice than CT treatment ($P < 0.05$, Fig. 5). The F/B ratios varied significantly over the maize growing season at all sampling depths (Table 3).

3.4. Soil microbial community structure

PCA of PLFA data from all plots revealed a total sample variance of 40% and 17% for the first two principal components (PCs). The PCA plot showed that PLFA profiles were separated by depth in the PC1 with 10 data points for the 0–5 cm depth showing negative scores, 10 data points for the 5–10 cm depth showing medial scores, and 9 data points for the 10–20 cm depth showing positive scores (Fig. 6). The PCA plot also indicated that PLFA profiles were separated by sampling month in the PC2 with 6 data points for July showing negative scores, 9 data points for June and August showing medial scores, and 10 data points for May and September showing positive scores. Further PCA was performed only for 0–5 and 5–10 cm depth since there were no differences in total, fungal, and bacterial PLFAs at the 10–20 cm depth. The plots showed that PC1 accounted for 39% and 33% of the total variance at 0–5 and 5–10 cm depth, respectively. PLFA profiles grouped together in June, July, August, and September at both 0–5 and 5–10 cm depths (Fig. 7a and b). The loading plots revealed that the cyclopropane fatty acid cy17:0 was most important for the separation of July, the saturated fatty acids 14:0, 15:0, and 16:0 for August, and the fatty acid 18:2 ω 6,9 for June (Fig. 7c and d).

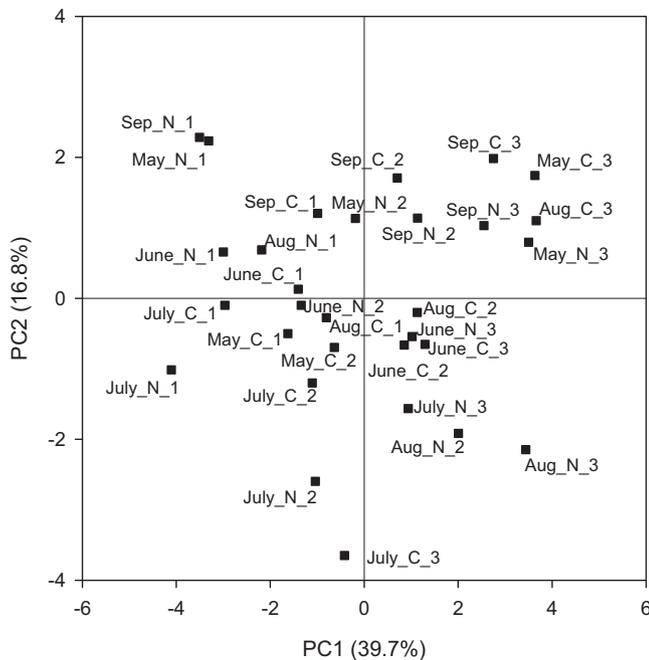


Fig. 6. Principal component analysis (PCA) of phospholipid fatty acids (PLFAs) for conventional- and no-tillage practices at three depths sampled over a maize growing season. Labels for data stand for Month_Tillage treatment_Depth (C, conventional tillage; N, no-tillage; 1, 0–5 cm; 2, 5–10 cm; 3, 10–20 cm).

4. Discussion

4.1. Seasonal variation

This study aimed to examine soil microbial community dynamics over a maize growing season under CT and NT practices. We found PLFA concentration and composition varied significantly during the growing season in both tillage treatments. However, distinct dynamic patterns of microbial biomass were found in CT and NT. Interestingly, the seasonal dynamics in soil microbial community did not follow the changes in soil temperature and water content. In addition, the distinct temporal trends were unrelated to the differences in soil temperature and water content between CT and NT practices. These findings indicate that crop growth and residue return were most important in regulating microbial community dynamics. Seasonal crop growth could play an important role in soil microbial dynamics by competing with microorganisms for substrates and altering the temporal and spatial distribution of organic inputs from crop roots and residues (Franzluebbers et al., 1995). The higher maize yields in NT soils suggest that crops require greater amounts of available substrates and nutrients, which might inhibit the growth and activity of microorganisms. This is supported by the relative lower PLFA concentration in NT soils in June, July, and August when maize was in rapid growth stages. Crop residues retained on the soil surface may have influenced the microbial dynamics greatly in NT plots. The residues in NT plots were mainly decomposed in autumn and spring periods and provided substrates to microbes, which probably resulted in higher PLFA concentration in May and September. This phenomenon was not observed in CT plots because all aboveground residues were removed after harvest. We could not conclude with certainty to any root effects since root development was not monitored in this study. However, it is likely that PLFA concentration was affected by the rhizodeposition throughout the growing season and the presence of crop roots at the end of the growing season.

PLFA analysis which measures the concentration of phospholipids in intact microbial cell membranes might induce unintentional variations in soil microbial biomass compared with measurements of MBC (Tunlid and White, 1992; Bardgett et al., 1999). However, the significant correlation between total PLFA and MBC in our study ($r = 0.78$, $P < 0.001$) implies that the detected seasonal variations in soil microbial communities were not caused by the analytical methods used. In addition, generally constant SOC and total N contents through out the growing season (Table 2) suggest that the factors responsible for the large seasonal variations in soil microbial communities were at least possibly related to spatial variability.

PCA on PLFA profiles indicated that there was more variability in soil microbial community structure due to sampling month than to tillage practices during the maize growing season. This is consistent with Bossio et al. (1998) who found that changes in community structure over time were of greater magnitude than those associated with management regimes (fertilizer type and crop rotation). Our result is also supported by the work of Drijber et al. (2000) which indicated that community structure was clearly distinct between CT and NT, but only during the fallow phase of a long-term wheat-fallow rotation. The PCA indicated cy17:0 characterized the PLFA profiles in July (Fig. 7) and the proportion of cy17:0 was higher in July than in other months. It is reported that cy17:0, which is indicative of Gram-negative bacteria, increased with increasing temperature (Zogg et al., 1997). This concurs with our data of higher temperature in July sampled soils (Fig. 1). Thus, temperature may be partially responsible for the shift in microbial community structure. In addition, the lower soil water content in July (Fig. 1) may contribute to the separation of PLFA profiles to some extent. Soil water content was found as the major factor influencing microbial community structure across seven biogeoclimatic zones in western Canada (Brockett et al., 2012), but it is not totally true in our site.

4.2. Tillage effect

In this study, microbial biomass accumulation under NT treatment at the surface layer is consistent with investigations from various climatic conditions and soil types (Frey et al., 1999; Feng et al., 2003; Spedding et al., 2004; Helgason et al., 2009). It is well known that changes in the microbial biomass pools coincide with changes in substrate availability. In this study, the NT treatment resulted in a resource rich layer near the soil surface (Table 2) which could promote the growth and activity of microbial communities when maize was not in rapid growth stages. In addition, NT practice provided the soil with a wetter plow layer (Fig. 1) and less fluctuation in water content and temperature (Zhang et al., 2005). These environmental conditions are also favorable for soil microorganisms to grow. Our previous study revealed significantly higher content of amino sugars derived from microbial cell wall in NT plots than CT plots at the surface layer (Ding et al., 2011), which supports the present findings.

Since the total microbial biomass alone could not fully capture the soil microbial community characteristics, the relative abundance of fungi to bacteria was examined. As we expected, higher F/B ratio was found at the surface of NT soils, which agreed with other reports (Beare et al., 1997; Frey et al., 1999). One possible reason for this observation is the reduced soil disturbance under NT practices. Soil fungi are very sensitive to physical disturbances due to their fragile hyphae (Kabir et al., 1999). NT practice is therefore more favorable than CT treatment for fungi forming hyphal networks that could promote the translocation of nutrients and resources (Klein and Paschke, 2004). Another factor that contributes to the higher F/B ratio in NT soils may be the soil water

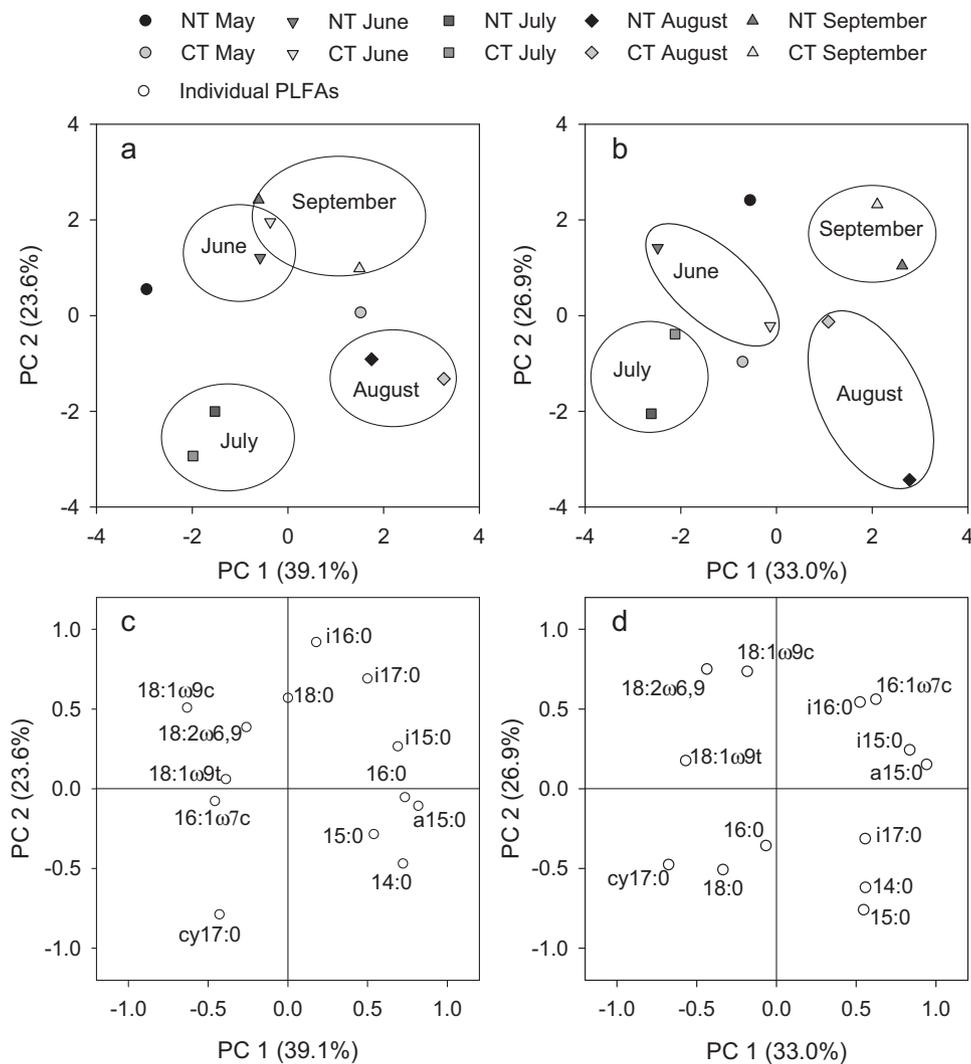


Fig. 7. Principal component analysis (PCA) of phospholipid fatty acids (PLFAs) at (a) 0–5 and (b) 5–10 cm depth in a clay loam soil under conventional tillage (CT) and no-tillage (NT) practices. (c) and (d) are PCA loading plots of the first two components of the individual PLFAs at 0–5 and 5–10 cm depths, respectively.

content since Frey et al. (1999) reported that fungi responded positively to increases in soil water content. We did find that NT practices resulted in higher soil water content at the surface layer than the CT treatment (Fig. 1), but we do not know to what extent soil water affected the F/B ratio.

4.3. Soil depth effect

Depth as a significant determinant of soil microbial communities has been observed in many studies (Fierer et al., 2003; Bausenwein et al., 2008; Helgason et al., 2009). Soil depth provided a gradient of changing habitat conditions as reflected by changes in PLFA profiles (Fig. 6). In addition, soil microbial communities at 10–20 cm depth were not affected remarkably by tillage practices (Figs. 2–4). In NT soils, the decrease in SOC and total N with increasing depth could be an important reason for the pronounced decrease in soil microbial biomass at depths. In contrast, the microbial biomass decrease was less pronounced under the CT treatment since chisel plowing and disking practices homogenize the substrates and resources across the plow layer (Table 2). Besides substrate availability, other factors such as soil water content (Schimel et al., 1999), temperature (Zogg et al., 1997), and aeration may collectively contribute to the variation in microbial communities across soil depths.

5. Conclusion

We found higher microbial biomass in NT soils than CT soils and a community shift toward fungi under NT practices, which supports our first hypothesis. However, the differences in microbial biomass and community composition between CT and NT practices were not consistent during the maize growing season. We found distinct dynamic patterns in microbial biomass under the two tillage treatments. These findings indicate that crop growth and residue return were the key factors in determining soil microbial community dynamics, whereas soil temperature and water content play a less important role. We concluded that NT practice can be used as part of an overall strategy to improve soil quality in rainfed agricultural ecosystem of northeastern China. This study also emphasizes the temporal variability of soil microbial communities in agroecosystem, and thus, it is recommended that samples should be collected both within and between seasons and years to best assess management impacts on microbial dynamics for a given landscape.

Acknowledgements

This work was financially supported by the Natural Science Foundation of China, (41130524) the Natural Basic Research

Program of China (2009CB118600) and the International Partnership Program of Chinese Academy of Science (KZCX2-YW-T06). The authors want to thank Wenny Ng for her kind help in polishing this manuscript. We appreciate Dr. Craig Drury from Agriculture and Agri-Food Canada for his valuable editorial comments. The two anonymous reviewers are gratefully acknowledged for their constructive comments and critical review of this manuscript.

References

- Bardgett, R.D., Lovell, R.D., Hobbs, P.J., Jarvis, S.C., 1999. Seasonal changes in soil microbial communities along a fertility gradient of temperate grasslands. *Soil Biology and Biochemistry* 31, 1021–1030.
- Bausenwein, U., Gättinger, A., Langer, U., Embacher, A., Hartmann, H.P., Sommer, M., Munch, J.C., Schloter, M., 2008. Exploring soil microbial communities and soil organic matter: variability and interactions in arable soils under minimum tillage practices. *Applied Soil Ecology* 40, 67–77.
- Beare, M.H., Hu, S., Coleman, D.C., Hendrix, P.E., 1997. Influences of mycelial fungi on soil aggregation and organic matter storage in conventional and no-tillage soils. *Applied Soil Ecology* 5, 211–219.
- Bligh, E.G., Dyer, W.J., 1959. A rapid method of total lipid extraction and purification. *Canadian Journal of Biochemistry and Physics* 37, 911–917.
- Bossio, D.A., Scow, K.M., Gunapala, N., Graham, K.J., 1998. Determinants of soil microbial communities: effects of agricultural management, season, and soil type on phospholipid fatty acid profiles. *Microbial Ecology* 36, 1–12.
- Brockett, B.F.T., Prescott, C.E., Grayston, S.J., 2012. Soil moisture is the major factor influencing microbial community structure and enzyme activities across seven biogeoclimatic zones in western Canada. *Soil Biology and Biochemistry* 44, 9–20.
- Carpenter-Boggs, L., Stahl, P.D., Lindstrom, M.J., Schumacher, T.E., 2003. Soil microbial properties under permanent grass, conventional tillage, and no-till management in South Dakota. *Soil and Tillage Research* 71, 15–23.
- Ding, X.L., Zhang, B., Zhang, X.D., Yang, X.M., Zhang, X.P., 2011. Effects of tillage and crop rotation on soil microbial residues in a rainfed agroecosystem of northeast China. *Soil and Tillage Research* 114, 43–49.
- Drijber, R.A., Doran, J.W., Pankhurst, A.M., Lyon, D.J., 2000. Changes in soil microbial community structure with tillage under long-term wheat-fallow management. *Soil Biology and Biochemistry* 32, 1419–1430.
- Feng, Y., Motta, A.C., Reeves, D.W., Burmester, C.H., van Santen, E., Osborne, J.A., 2003. Soil microbial communities under conventional-till and no-till continuous cotton systems. *Soil Biology and Biochemistry* 35, 1693–1703.
- Fierer, N., Schimel, J.P., Holden, P.A., 2003. Variations in microbial community composition through two soil depth profiles. *Soil Biology and Biochemistry* 35, 167–176.
- Franzluebbers, A.J., Hons, F.M., Zuberer, D.A., 1995. Tillage and crop effects on seasonal soil carbon and nitrogen dynamics. *Soil Science Society of America Journal* 59, 1618–1624.
- Frey, S.D., Elliott, E.T., Paustian, K., 1999. Bacterial and fungal abundance and biomass in conventional and no-tillage agroecosystems along two climatic gradients. *Soil Biology and Biochemistry* 31, 573–585.
- Frostegård, A., Bååth, E., 1996. The use of phospholipid fatty acid analysis to estimate bacterial and fungal biomass in soil. *Biology and Fertility of Soils* 22, 59–65.
- Gil-Sotres, F., Trasar-Cepeda, C., Leiros, M.C., Seoane, S., 2005. Different approaches to evaluating soil quality using biochemical properties. *Soil Biology and Biochemistry* 37, 877–887.
- Helgason, B.L., Walley, F.L., Germida, J.J., 2009. Fungal and bacterial abundance in long-term no-till and intensive-till soils of the northern great plains. *Soil Science Society of America Journal* 73, 120–127.
- Kabir, Z., O'Halloran, I.P., Hamel, C., 1999. Combined effects of soil disturbance and fallowing on plant and fungal components of mycorrhizal corn (*Zea mays* L.). *Soil Biology and Biochemistry* 31, 307–314.
- Klein, D.A., Paschke, M.W., 2004. Filamentous fungi: the indeterminate lifestyle and microbial ecology. *Microbial Ecology* 47, 224–235.
- Liang, A.Z., Zhang, X.P., Fang, H.J., Yang, X.M., Drury, C.F., 2007. Short-term effects of tillage practices on organic carbon in clay loam soil of northeast China. *Pedosphere* 17, 619–623.
- Liu, X.B., Han, X.Z., Song, C.Y., Herbert, S.J., Xing, B.S., 2003. Soil organic carbon dynamics in black soils of China under different agricultural management systems. *Communications in Soil Science and Plant Analysis* 34, 973–984.
- Liu, X.B., Zhang, X.Y., Herbert, S.J., 2010. Feeding China's growing needs for grain. *Nature* 465, 420.
- Minoshima, H., Jackson, L.E., Cavagnaro, T.R., Sánchez-Moreno, S., Ferris, H., Temple, S.R., Goyal, S., Mitchell, J.P., 2007. Soil food webs and carbon dynamics in response to conservation tillage in California. *Soil Science Society of America Journal* 71, 952–963.
- Pankhurst, C.E., Kirkby, C.A., Hawke, B.G., Harch, B.D., 2002. Impact of a change in tillage and crop residue management practice on soil chemical and microbiological properties in a cereal-producing red duplex soil in NSW, Australia. *Biology and Fertility of Soils* 35, 189–196.
- Paul, E.A., Clark, F.E., 1989. *Soil Microbiology and Biochemistry*. Academic Press, San Diego.
- Peel, M.C., Finlayson, B.L., McMahon, T.A., 2007. Updated world map of the Köppen–Geiger climate classification. *Hydrology and Earth System Sciences* 11, 1633–1644.
- Petersen, S.O., Frohne, P.S., Kennedy, A.C., 2002. Dynamics of a soil microbial community under spring wheat. *Soil Science Society of America Journal* 66, 826–833.
- Schimel, J.P., 1995. Ecosystem consequences of microbial diversity and community structure. *Ecological Studies* 113, 239–254.
- Schimel, J.P., Gullledge, J.M., Clein-Curley, J.S., Lindstrom, J.E., Braddock, J.F., 1999. Moisture effects on microbial activity and community structure in decomposing birch litter in the Alaskan taiga. *Soil Biology and Biochemistry* 31, 831–838.
- Schlöter, M., Dilly, O., Munch, J.C., 2003. Indicators for evaluating soil quality. *Agriculture Ecosystems and Environment* 98, 255–262.
- Soil Survey Staff, 2010. *Keys to Soil Taxonomy*, 11th ed. USDA-Natural Resources Conservation Service, Washington, DC, USA.
- Spedding, T.A., Hamel, C., Mehuys, G.R., Madramootoo, C.A., 2004. Soil microbial dynamics in maize-growing soil under different tillage and residue management systems. *Soil Biology and Biochemistry* 36, 499–512.
- Tunlid, A., White, D.C., 1992. Biochemical analysis of biomass, community structure, nutritional status and metabolic activity of microbial communities in soil. In: Stotzky, G., Bollag, J.M. (Eds.), *Soil Biochemistry*, vol. 7. Marcel Dekker, New York, pp. 229–262.
- van Groenigen, K.J., Bloem, J., Bååth, E., Boeckx, P., Rousk, J., Bodé, S., Forristal, D., Jones, M.B., 2010. Abundance, production and stabilization of microbial biomass under conventional and reduced tillage. *Soil Biology and Biochemistry* 42, 48–55.
- Vance, E.D., Brookes, P.C., Jenkinson, D.S., 1987. An extraction method for measuring soil microbial biomass C. *Soil Biology and Biochemistry* 19, 703–707.
- Wu, J., Joergensen, R.G., Pommerening, B., Chaussod, R., Brookes, P.C., 1990. Measurement of microbial biomass C by fumigation–extraction – an automated procedure. *Soil Biology and Biochemistry* 22, 1167–1169.
- Zelles, L., 1999. Fatty acid patterns of phospholipids and lipopolysaccharides in the characterization of microbial communities in soil: a review. *Biology and Fertility of Soils* 29, 111–129.
- Zhang, X.P., Fang, H.J., Yang, X.M., Liang, A.Z., Wu, S.H., 2005. Effects of no-tillage practices on temperature and moisture of a black soil in the spring and early summer. *Chinese Journal of Soil Science* 36, 313–316 (in Chinese).
- Zogg, G., Zak, D., Ringelberg, D., Macdonald, N., Pregitzer, K., White, D., 1997. Compositional and functional shifts in microbial communities due to soil warming. *Soil Science Society of America Journal* 61, 475–481.