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Temperature sensitivity of soil respiration: Synthetic effects of nitrogen and phosphorus fertilization on Chinese Loess Plateau

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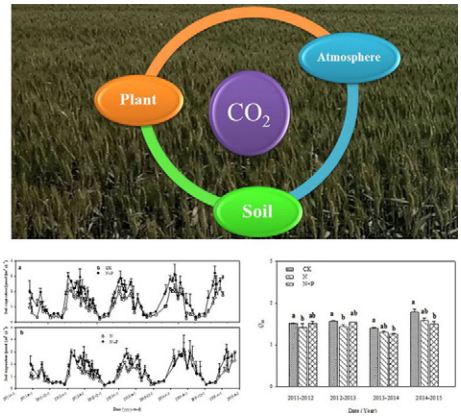
HIGHLIGHTS

- Soil respiration was positively related to SMBC, DOC and photosynthesis rate.
- Variation in Q_{10} maybe related to the increased soil mineral N content and the relative abundance of Acidobacteria.

GRAPHICAL ABSTRACT



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ABSTRACT

Nitrogen (N) and phosphorus (P) fertilization has the potential to alter soil respiration temperature sensitivity (Q_{10}) by changing soil biochemical and crop physiological process. A four-year field experiment was conducted to determine how Q_{10} responded to these biochemical and physiological changes in rain-fed agro-ecosystems on the semi-arid Loess Plateau. Soil respiration, as well as biotic and abiotic factors were measured in winter wheat (*Triticum aestivum* L.), with three fertilization treatments: (no fertilization (CK), 160 kg N hm^{-1} (N), and 160 kg N ha^{-1} with 39 kg P ha^{-1} (N + P). Mean annual soil respiration rate (calculated by averaging the four years) in the N treatment and N + P treatment was 18% and 48% higher than that in the CK treatment, respectively; and it was increased by 26% (14%–48%) in the N + P treatment as compared with that in the N treatment. The decrease of Q_{10} in the N and N + P treatments against the CK treatment was not stable for each year, ranging from 0.01 to 0.28. The maximum decrease of Q_{10} in the N and N + P treatments was 10% and 15% in 2014–2015, while in other years the decrease of Q_{10} was numerical but not significant. Soil microbial biomass carbon (SMBC) was increased by 10% and 50%, dissolved organic carbon (DOC) was increased by 6% and 21%, and photosynthesis rate was increased ranging from 6% to 33% with N and N + P fertilization. The relative abundance of Acidobacteria, Actinobacteria and Chloroflexi were significantly higher by 32.9%–54.1% in N addition soils (N and N + P) compared to CK treatment, whereas additional P application into soils increased the relative abundance of the family Micrococcaceae, Nocardioidaceae and Chitinophagaceae. Soil respiration was positively related to SMBC, DOC and photosynthesis rate ($p < 0.05$). However, variation in Q_{10} may be related to the increase of

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soil mineral N content and variation of the relative abundance of soil microbial community in our study. Nitrogen and additional phosphorus fertilization regimes affect soil respiration and temperature sensitivity differently.

1. Introduction

Temperature sensitivity of soil respiration (often expressed as Q_{10}) is regarded as an important mechanism for the possible feedback between the carbon cycle in terrestrial ecosystem and the climate system (Davidson and Janssens, 2006; Jiang et al., 2015a, 2015b, 2015c). Small variation in Q_{10} can cause a large bias in estimating soil CO_2 release into atmosphere (Xu and Qi, 2001). There is increasing evidence suggesting that Q_{10} is not constant but influenced by many factors. For instance, soil temperature, soil moisture (Janssens and Pilegaard, 2003; Kirschbaum, 1995; Qi and Xu, 2001; Schleser, 1982), and soil nutrient availability (Johnson et al., 2000; Jiang et al., 2015a) by changing soil microbial growth, composition and activity (Jia et al., 2010; Li et al., 2013; Yu et al., 2013; Contosta et al., 2015), and growth of above-ground and root biomass (Pregitzer et al., 2000; Jiang et al., 2015a). Chemical fertilization, as a common management practice in agro-ecosystems, can extensively change soil biological processes and crop primary productivity (Vitousek et al., 2010). However, the mechanism underlying the responses of soil respiration and Q_{10} to chemical fertilization regimes remains unclear.

Application of nitrogen (N) and phosphorus (P) fertilizer into soil-crop system could induce changes in crop physiological processes and soil biochemical processes (Beauregard et al., 2010; Tully et al., 2015; Wood et al., 2015). N fertilization directly or indirectly alter soil microbial community structure (Wakelin et al., 2012; Turlapati et al., 2013; Pan et al., 2014; Yao et al., 2014), microbial growth and activity (Jia et al., 2010; Li et al., 2013; Yu et al., 2013; Contosta et al., 2015). It has context-dependent consequences, with possible outcomes on the microbial community of inhibition, stimulation, or no effect (Hopkins et al., 2008; Ramirez et al., 2012; Ball and Virginia, 2014). In addition, the availability of N is shown to have a significant effect on the photosynthesis rate and the allocation of photosynthetic products between aboveground and belowground components (Bobul'ska et al., 2015; Shukla et al., 2015), root biomass and root tissue N content (Majdi, 2001; Burton et al., 2002; King et al., 2002), and dissolved organic carbon (DOC) content (Zhong et al., 2015). DOC, as an indicator of carbon availability to soil microorganisms (Boyer and Groffman, 1996), it is both a substrate for microbial activity and a byproduct of the subsequent microbial metabolic processes (Marschner and Kalbitz, 2003; Straathof et al., 2014). In addition, soil N: P ratio can influence soil microbial biomass carbon (SMBC), DOC, photosynthesis rate and some other biochemical processes (Ball and Virginia, 2014). Therefore, additional phosphorus fertilization, via influencing in situ soil N: P ratio, crop growth, microbial organisms activity (Liu et al., 2012), may also cause changes in soil respiration and Q_{10}.

However, the effects of chemical fertilization regimes on soil respiration and Q_{10} remain poorly understood. In this study, soil respiration, soil properties, biological indicators were measured in winter wheat systems on the semi-arid Loess Plateau. The main purposes of this study were to: 1) identify the responses of soil respiration and temperature sensitivity to N and N + P fertilization; 2) determine the changes in SMBC, DOC, photosynthesis rate and soil microbial community structure among fertilization regimes; and 3) explore the factors potentially influencing the temperature sensitivity of soil respiration in winter wheat systems on the semi-arid Loess Plateau under N and N + P treatments.

2. Materials and methods

2.1. Site description

On the Loess Plateau, the area of arable land is about 145, 000 km^2, and >70% of crops are planted in rain-fed areas and thus are particularly susceptible to the impacts of climate change (Jiang et al., 2015a). A long-term field experiment was established in September 1984 at Changwu State Key Agro-Ecological Experimental Station located in Wangdonggou (35°13′N, 107°40′E; 1220 m a.s.l), Changwu Country, Shaanxi Province, China (Fig. 1). The study area is a typical tableland-gully region in the southern Loess Plateau in the middle reaches of the Yellow River. It has a continental monsoon climate characterized by hot summers and cold winters. Annual mean precipitation is 560 mm, 60% of which occurs between July and September. Annual mean air temperature is 9.4 °C, and the ≥10 °C accumulated temperature is 3029 °C, and the annual sunshine duration is 2230 h with a total radiation of 484 kJ cm^{-2}, and frost-free period of 171 days. All meteorological data during the experiment time were provided by Changwu State Key Agro-Ecological Experimental Station (Fig. 2).

The soil is a uniform loam of loess deposits belonging to Cumulic Haplustolls according to American Soil Classification System and originated from parent material of calcareous loess. Soils collected at 0–20 cm depth are characterized by: pH of 8.3 (1:1 soil/H_2O suspension), clay content (<0.002 mm) of 24%, field capacity of 22.4%, permanent wilting point of 9.0%, and $CaCO_3$ of 10.5% (Wang et al., 2015a, 2015b; Zhang et al., 2015).

2.2. Experimental design

Given the fact that single P fertilization is not commonly applied on the Loess Plateau but more to use N or N + P fertilization regimes, we attempted to exactly follow the local agriculture management when designing the experimental treatments. Therefore, three fertilization regimes, no fertilization (CK), 160 kg N hm^{-1} (N), and 160 kg N hm^{-1} and 39 kg P hm^{-1}(N + P), were applied to winter wheat (*Triticum aestivum* L., cv. Changwu 89 (1) 3–40). All treatments were arranged in a randomized block design with three replicates per treatment. Each plot was 18 m long by 5.5 m wide and spaced 0.5 m apart, and blocks were separated by a 1.0 m strip. N fertilizer (urea, 46.0% N) and triple superphosphate fertilizer (46% P_2O_5) were broadcasted 5–7 days prior to sowing. Tillage was performed to prepare a seed bed prior to sowing, and wither wheat was planted in September in 20 cm wide rows at a seeding rate of 150 kg ha^{-1}. Weeds were removed manually and plant protection measures were applied as needed. The wheat was harvested manually by cutting close to the ground, and all harvested biomass was removed from the plots at physiological maturity (late June) each year.

2.3. Measurements of soil respiration, soil temperature, and soil moisture

Soil respiration was measured using an automated soil CO_2 flux system equipped with a portable chamber of 20 cm in diameter (Li-8100, Lincoln, NE, USA). Before the measurement, all visible living organisms were artificially removed. At least two measurements were taken for each plot, with a 90 s enclosure period and a 30 s delay between the two measurements, and the average of the two measurements was taken as the soil respiration. However, if the variation between these

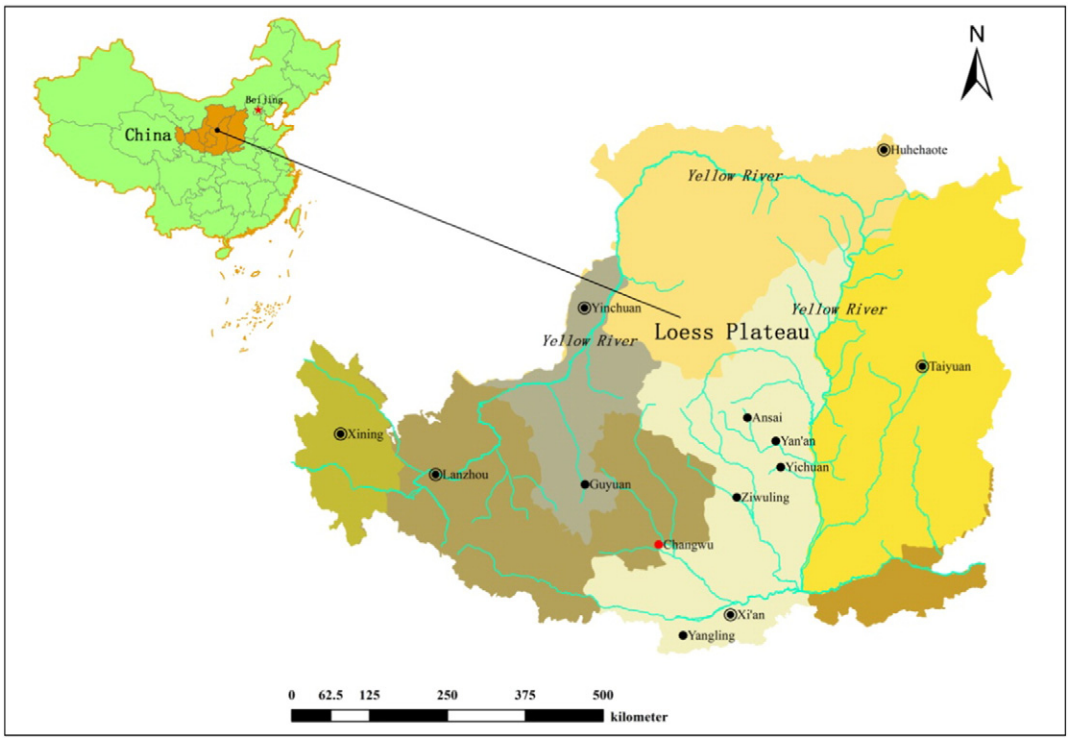

Fig. 1. A sketch map of the Loess Plateau, China.

two measurements was larger than 15%, one or more measurements were taken until the variation between two consecutive measurements was <15%. The measurement was conducted from 09:00 am to 11:00 am (Iqbal et al., 2010) about every 15 days from July 2011 to June 2015. Soil bulk density at 0–5 cm depth was measured using a cutting ring of 5 cm in both depth and diameter.

Along with the soil respiration measurements, soil temperature and moisture at 5-cm depth were also measured (3 and 4 replicates per collar, respectively) in different directions, each 10 cm distant away from the collar. The soil temperature was measured using a Li-Cor thermocouple probe, and the soil moisture was determined by a Theta Probe ML2X with an HH2 moisture meter (Delta-TDevices, Cambridge, England). Soil water-filled pore space (WFPS) was calculated by the following equation (Ding et al., 2007):

$$\text{WFPS}\ (\%) = [\text{volumetric water content}/100 \times (2.65 - \text{soil bulk density})/2.65] \quad (1)$$

2.4. Sampling and analysis

Soil samples were collected using a soil auger of 3 cm in diameter both during the growing and fellow season in the last experimental year (2014–2015), and each sample consisted of six subsamples which randomly collected at top soil (0–20 cm). Each sample was passed through a 2.0-mm sieve and allocated into three subsamples: one part stored at −80 °C for DNA extraction, one part stored at 4 °C for the measurement of SMBC and soil mineral N content (NO_3—N + NH_4—N), and the last part was air dried and then crushed to pass through a 0.15 mm sieve. Soil organic carbon (SOC) was determined using the K_2CrO_7—H_2SO_4 oxidation method (Sparks et al., 1996); the N concentrations in soil samples were determined by acid digestion according to the Kjeldahl method (Grimshaw et al., 1989); DOC was determined using a total organic carbon analyzer (TOC-VCSH, Shimadzu, Japan) (Fujii et al., 2011); and extractable NO_3—N and NH_4—N were extracted with KCl and determined by colorimetry using a Bran & Luebbe II AutoAnalyser (Fernandez-Escobar et al., 2009).

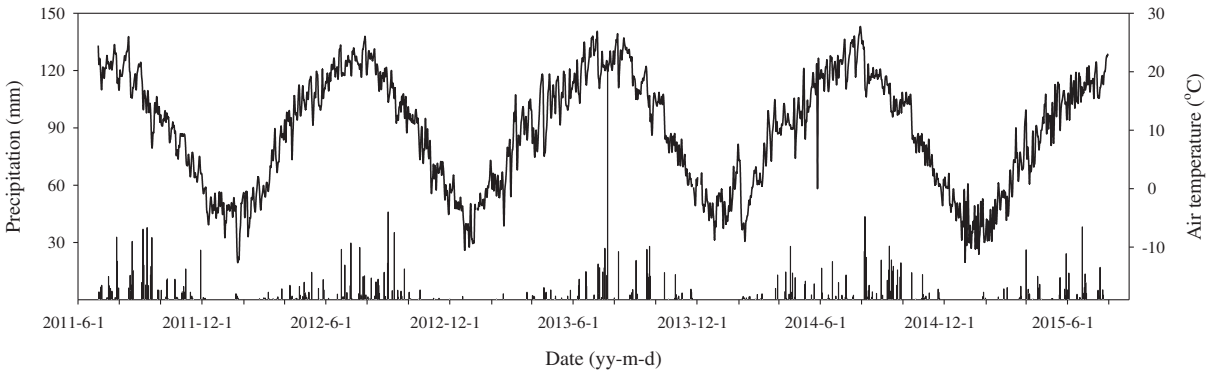

Fig. 2. Variation of precipitation (mm) and air temperature (°C) over the experimental period from 2011 to 2015.

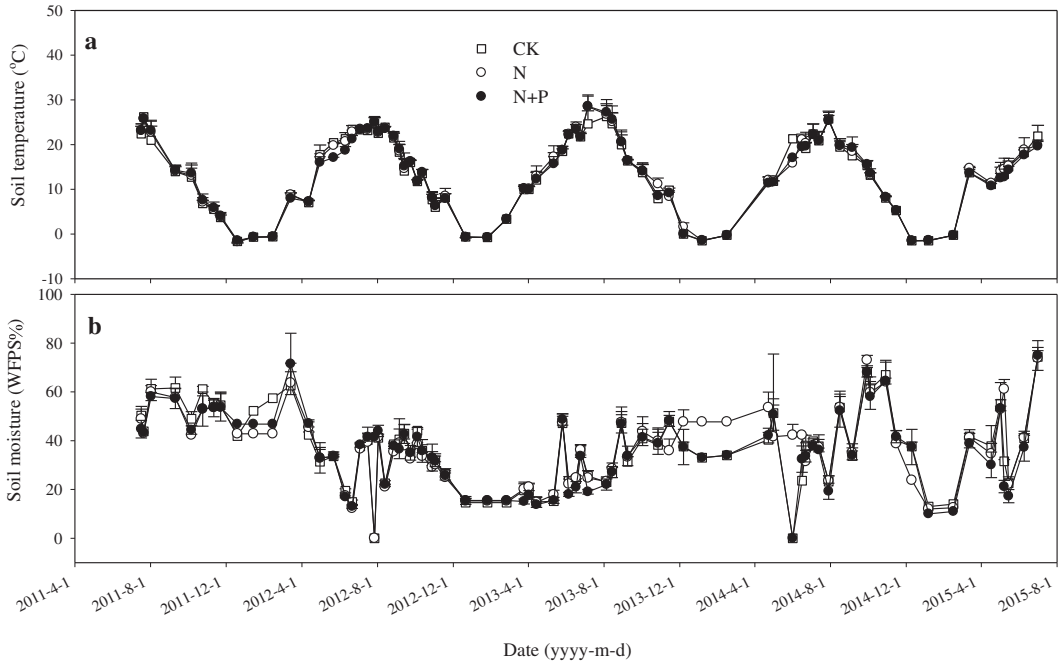

Fig. 3. Variation of soil temperature (°C) and soil moisture (%WFPS) over the experimental period from 2011 to 2015.

SMBC was determined by the chloroform fumigation-extraction method (Vance et al., 1987), a well-recognized method to estimate SMBC in various ecosystems (van Gestel et al., 2011). Photosynthesis rate was measured with a LI-6400 Portable Photosynthesis System (LI-COR, Lincoln, Nebraska, USA) under natural conditions in different phenological phases in 2014–2015, and all the measurements were taken from 9 am to 11 am to avoid the decrease in photosynthesis at midday (Wang et al., 2015a, 2015b).

Soil DNA was extracted from 0.5 g soil using the FastDNA® Spin Kit for Soil (MP Biomedicals, Cleveland, OH, USA) according to the manufacturer's instructions. The purified DNA was diluted with 50 μl sterilized water and checked for quality and quantity using a NanoDrop Spectrophotometer.

DNA was amplified using the primers 515F (50-GTGCCAGC MGCCGCGGTAA-30) and 806R (50-GGACTACHVGGGTWTCTAAT-30) designed to be universal for bacteria and archaea (Caporaso et al., 2011). Primers were tagged with unique barcodes for each replicate DNA sample. PCR reactions were carried out in a 30-μl mixture with 15 μl of Phusion® High-Fidelity PCR Master Mix (New England Biolabs), 0.2 μM of each primer, and about 10 ng template DNA. The thermal cycling was as follows: 98 °C for 1 min; 30 cycles of 98 °C for 10 s, 50 °C for 30 s, and 72 °C for 1 min; and 72 °C for 5 min. Negative controls using sterilized water instead of soil DNA were included to avoid primer or sample DNA contamination. Each DNA sample was amplified in three technical replicates and then quantified with electrophoresis and mixed in one tube. All samples were pooled together with equal molar amounts from each sample and purified with the GeneJET gel extraction kit (Thermo Scientific). The purified library was generated using NEB Next® Ultra™ DNA Library Prep Kit for Illumina (NEB, USA) and mixed with the index codes. The library quality was assessed in the Qubit® 2.0 Fluorometer (Thermo Scientific) and Agilent Bioanalyzer 2100 system. Then, the library was sequenced on an Illumina MiSeq platform by which 250 bp/300 bp paired-end reads were generated.

All sequence reads were merged using FLASH (Magoč and Salzberg, 2011) and assigned to each sample according to their barcodes. Sequence analysis was performed by UPARSE software package using the UPARSE-OTU and UPARSE-OTUref algorithms (Edgar, 2013). Sequences with ≥97% similarity were clustered into operational taxonomic units (OTUs). The aligned 16S rRNA gene sequences were used for a

Table 1
Mean soil moisture, mean soil temperature and cumulative respiration over the study period 2011–2015.

Year	Treatment	Soil moisture (%)	Soil temperature (°C)	Soil respiration (μmol m^{-2} s^{-1})
2011–2012	CK	45.79 ± 14.59a	12.82 ± 9.39a	0.99 ± 0.62c
	N	43.18 ± 13.80a	12.90 ± 9.44a	1.16 ± 0.75b
	N + P	44.08 ± 14.44a	12.53 ± 9.02a	1.49 ± 0.84a
2012–2013	CK	28.37 ± 12.64a	14.17 ± 7.85a	1.14 ± 0.49b
	N	27.35 ± 11.57a	14.40 ± 7.86a	1.15 ± 0.47b
	N + P	29.67 ± 11.77a	14.33 ± 7.89a	1.70 ± 0.74a
2013–2014	CK	33.29 ± 3.50a	15.10 ± 9.02a	1.22 ± 0.62c
	N	38.45 ± 9.17a	15.44 ± 9.10a	1.48 ± 0.86b
	N + P	33.06 ± 12.20a	15.17 ± 9.30a	1.69 ± 0.84a
2014–2015	CK	41.74 ± 17.67a	13.21 ± 8.07a	1.23 ± 0.75c
	N	42.03 ± 18.84a	13.39 ± 8.02a	1.62 ± 0.85b
	N + P	39.10 ± 18.93a	13.08 ± 7.97a	1.86 ± 0.94a
Mean value	CK	37.30 ± 7.91a	13.83 ± 1.02a	1.15 ± 0.11c
	N	37.75 ± 7.22a	14.03 ± 1.13a	1.35 ± 0.24b
	N + P	36.48 ± 16.40a	13.78 ± 1.20a	1.69 ± 0.15a

Note: Different letters represent significant differences between the treatments ($p < 0.05$), values are means of three replicates ± SE.

chimera check using the Uchime algorithm (Edgar et al., 2011). Taxonomy was assigned using the Ribosomal Database Project classifier (Wang et al., 2007). Each sample was rarefied to the same number of reads (28,318 sequences) for both alpha-diversity (Chao1 estimator of richness, observed species and Shannon's diversity index) analyses. The original sequence data are available at the European Nucleotide Archive (ENA) with accession number PRJEB11700 (http://www.ebi.ac.uk/ena/data/view/PRJEB11700).

2.5. Root biomass

To minimize root heterogeneity, six soil cores (0–20 cm) were taken in each plot (three cores at the middle of two rows, and the other three cores at rows) using a sharp iron tube (9 cm in diameter), and mixed well for the measurement of root biomass. Roots were separated from soils by soaking in water and gentle washing over a 0.25 mm mesh. Wet roots were oven dried at 60 °C for 48 h to a constant weight.

2.6. Data analysis

A univariate exponential function model was used to characterize the relationship between soil respiration and soil temperature (Davidson et al., 1998):

$$y = \beta_0 e^{\beta 1 T} \quad (2)$$

where y is the measured soil respiration (μmol m^{-2} s^{-1}), T is the measured soil temperature (°C) at a certain depth, and β_0 and β_1 are constants fitted by the least squares method.

The Q_{10} values were calculated by Eq.(3) (Xu and Qi, 2001):

$$Q_{10} = e^{10\beta 1} \quad (3)$$

Data (mean ± SD, $n = 3$) were subjected to ANOVA, followed by a LSD test for post hoc comparisons of means. Statistical significance was defined as $p \le 0.05$. The seasonal soil respiration rate and the variance of cumulative respiration were analyzed using mixed and general linear models, respectively; and the relationship between soil respiration and temperature, SMBC, DOC, and photosynthesis rate was determined by regression analysis. All analyses were performed using SAS software.

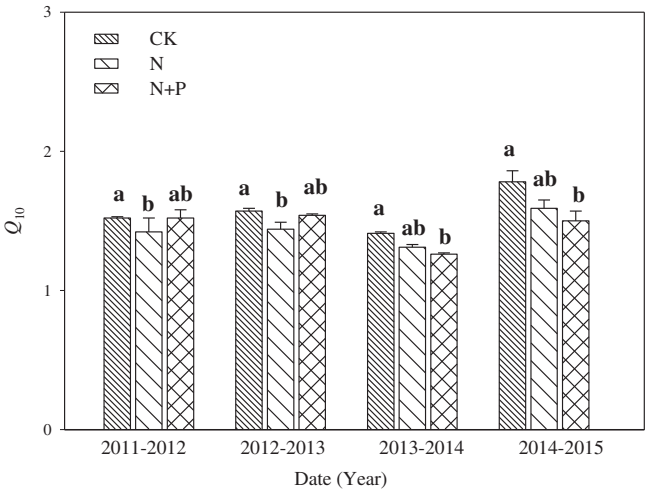

Fig. 5. Variation of Q_{10} among different fertilization regimes.

3. Results

3.1. Soil temperature and soil moisture

Soil temperature at 5-cm depth in the CK, N and N + P treatments showed very similar seasonal and annual variations (Fig. 3a), which was in good agreement with the variation of air temperature (Fig. 2). The lowest soil temperature was recorded in spring and winter, whereas the highest one was recorded in summer. The mean soil temperature over the study period was 13.83 °C, 14.08 °C and 13.78 °C in the CK, N and N + P treatments, respectively (Table 1).

Soil moisture at 0–5 cm depth fluctuated significantly in response to the irregular rainfall (Figs. 2 and 3b). The mean annual soil moisture over the study period was 37.30%, 37.75% and 36.48% WFPS in the CK, N and N + P treatments, respectively (Table 1).

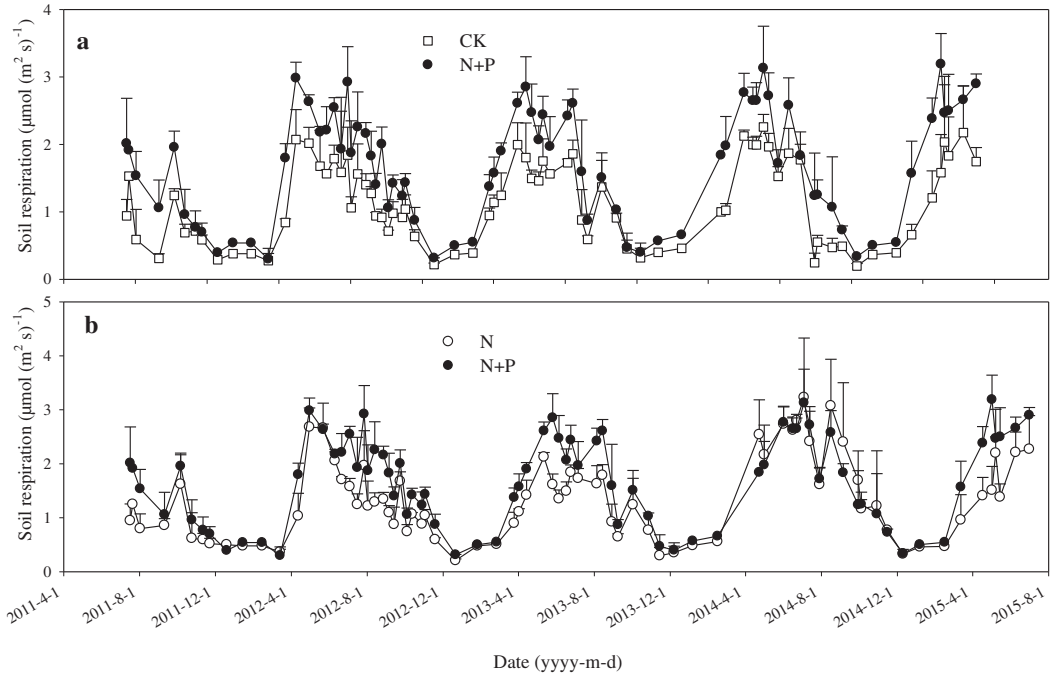

Fig. 4. Dynamics of soil respiration (μmol m^{-2} s^{-1}) from 2011 to 2015 in wheat systems under different N fertilization treatments on the semi-arid Loess region.

Table 2
Effects of fertilization on SMBC, DOC, and photosynthesis rate during different growing seasons.

Treatment	Returning green	Elongating	Flowering	Filling	Ripening	Mean
SMBC (mg kg^{-1})						
CK	91.8 ± 21.3a	91.5 ± 20.7b	182.0 ± 25.5b	136.8 ± 18.1b	252.1 ± 13.1b	150.4 ± 67.92b
N	96.8 ± 17.9b	109.6 ± 18.1b	202.9 ± 10.4b	143.2 ± 21.8b	250.6 ± 7.2b	158.6 ± 67.49b
N + P	110.5 ± 20.6a	205.7 ± 13.4a	252.7 ± 23.7a	160.3 ± 45.9a	340.0 ± 14.2a	213.8 ± 88.09a
DOC (mg kg^{-1})						
CK	15.7 ± 0.7b	13.5 ± 3.1a	30.7 ± 4.1b	38.7 ± 7.7b	24.8 ± 3.7c	24.7 ± 10.5b
N	16.5 ± 1.4b	14.4 ± 2.5a	33.4 ± 2.5b	40.2 ± 6.1b	29.2 ± 6.5b	26.6 ± 11.3b
N + P	19.3 ± 3.2a	15.4 ± 1.2a	42.5 ± 5.5a	61.1 ± 15.2a	31.7 ± 3.3a	34.0 ± 18.5a
Photosynthesis (μmol CO_2 m^{-2} s^{-1})						
CK	15.8 ± 1.0b	16.1 ± 1.1c	18.2 ± 0.4a	17.2 ± 1.3b	/	16.8 ± 1.1b
N	16.7 ± 0.4b	18.3 ± 1.1b	17.6 ± 0.3b	17.4 ± 0.8b	/	17.8 ± 0.9b
N + P	18.1 ± 0.4ba	24.5 ± 1.1a	19.8 ± 0.8a	18.5 ± 2.5a	/	20.2 ± 3.0a

Note: Different letters represent significant differences between the treatments during the same period ($p < 0.05$), values are means of three replicates ± SE.

3.2. Responses of soil respiration to N and N + P fertilization

Soil respiration showed similar seasonal and annual variations in the CK, N and N + P treatments. It increased gradually with increasing temperature from March to June, and decreased quickly as the temperature declined after October. The highest soil respiration was recorded in May (Fig. 4).

There was a significant difference in soil respiration rate among the CK, N and N + P treatments (Table 1). The mean annual soil respiration rate (calculated by averaging the four years) in the N treatment (1.35 μmol m^{-2} s^{-1}) and the N + P treatment (1.69 μmol m^{-2} s^{-1}) was 18% and 48% higher than that in the CK treatment (1.15 μmol m^{-2} s^{-1}). It was on average increased by 26% (14% − 48%) in the N + P treatment as compared to that in the N treatment (Table 1).

3.3. Responses of Q_{10} to N and N + P fertilization

Nitrogen and additional phosphorus in the N + P treatment in general decreased temperature sensitivity of soil respiration (Fig. 5), but the decrease was not stable for each year, ranging from 0.01 to 0.28. The maximum decrease of Q_{10} in the N and N + P treatments was 10% and 15% in 2014–2015, but in other years the decrease of Q_{10} was numerical but not significant. After averaging the four years, the mean annual Q_{10} in the N and N + P treatments slightly decreased by 8% (N: 1.44) and 7% (N + P: 1.46) when compared with that in the CK treatment (1.57). It was also noted that additional phosphorus in the N + P treatment, although not significantly ($p < 0.05$), ($p < 0.05$), resulted in an increase in Q_{10} when compared with N fertilization alone (Fig. 5).

3.4. Responses of SMBC, DOC, and photosynthesis to N and N + P fertilization

Compared with the CK treatment, N fertilization alone had no significant effect on SMBC, DOC and photosynthesis during the experimental period; whereas N + P fertilization resulted in a significant increase in SMBC, DOC, and photosynthesis ($p < 0.05$). More specifically, SMBC in the N treatment (158.6 mg kg^{-1}) and the N + P treatment (213.8 mg kg^{-1}) were 6% and 42% higher than that in the CK treatment (150.8 mg kg^{-1}); DOC in the N treatment (26.1 mg kg^{-1}) and the N + P treatment (34.6 mg kg^{-1}) were 8% and 37% higher than that in the CK treatment (24.7 mg kg^{-1}); and photosynthesis rate in the N treatment (20.0 μmol CO_2 m^{-2} s^{-1}) and the N + P treatment (22.5 μmol CO_2 m^{-2} s^{-1}) were 5% and 20% higher than that in the CK treatment (18.4 μmol CO_2 m^{-2} s^{-1}). In addition, SMBC, DOC and photosynthesis rate in the N + P treatment were 33%, 32% and 14% higher than that in the N treatment, respectively (Table 2). When compared to the CK treatment, root biomass significantly reduced by 33.9% with N fertilization alone, whereas increased by 15% with N + P fertilization ($p < 0.05$) (Table 3).

3.5. Responses of SOC and soil mineral N to N and N + P fertilization

N + P fertilization resulted in an increase in SOC and soil mineral N content (Table 3). The SOC content was 6.56, 6.65 and 7.16 g kg^{-1} in the CK, N and N + P treatments, respectively. However, N fertilization alone resulted in a 75% increase in soil mineral N content when compared to the CK treatment (22.33 vs. 12.72 mg kg^{-1}; $p < 0.05$). It was noted that the soil mineral N content in the N + P treatment (15.19 mg kg^{-1}) was higher than that in the CK treatment, but lower than that in the N treatment.

3.6. Soil respiration normalized by root biomass, SOC and SMBC

The soil respiration rates per unit root biomass in the N and N + P fertilized soils were significantly increased by 77.9% (2.42 vs. 1.36 t t^{-1}) and 27.9% (1.74 vs. 1.36 t t^{-1}) against the CK treatment ($p < 0.05$). However, the increase of soil respiration rates per unit SOC in the N (0.27 g g^{-1}) and N + P (0.32 g g^{-1}) treatments against the CK treatment (0.29 g g^{-1}) was numerical but not significant. Soil respiration rates per unit SMBC was barely changed, ranging from 11.60 mg g^{-1} in the N + P treatment to 12.17 mg g^{-1} in the N treatment (Table 4).

3.7. Responses of microbial communities to N and N + P fertilization

The Chao1 richness, observed species and OUT numbers were lowest in CK soil, whereas the Shannon's diversity index was no significant different among CK, N and N + P (Table 5). The relative abundances of phylum Acidobacteria, Actinobacteria, and Chloroflexi were

Table 3
Effects of fertilization on SOC, soil total N (STN) and soil mineral N (NO_3—N + NH_4—N) and soil total phosphorous (STP).

Treatment	SOC (g kg^{-1})	STN (g kg^{-1})	NO_3—N + NH_4—N (mg kg^{-1})	STP (g kg^{-1})	Root biomass(t ha^{-1})
CK	6.6 ± 0.2b	0.82 ± 0.02 a	12.72 ± 3.32 b	0.68 ± 0.02 a	3.19 ± 0.52 b
N	6.7 ± 0.2 b	0.85 ± 0.05 a	22.33 ± 8.92 a	0.68 ± 0.01 a	2.11 ± 0.13 c
N + P	7.9 ± 0.4 a	0.94 ± 0.05 a	15.19 ± 4.32 b	0.91 ± 0.03 a	3.67 ± 0.16 a

Table 4
Soil chemical and microbial properties in situ field experiment.

Items	CK	N	NP
Rs rate (g CO_2 – C kg^{-1} $year^{-1}$)	1.75 ± 0.04 b	1.93 ± 0.09 b	2.48 ± 0.07 a
Rs/Root biomass (t t^{-1})	1.36 ± 0.21 c	2.42 ± 0.23 a	1.74 ± 0.31 b
Rs/SOC (g g^{-1})	0.27 ± 0.20 a	0.29 ± 0.05 a	0.32 ± 0.08 a
Rs/MBC (mg g^{-1})	11.63 ± 0.59 a	12.17 ± 1.33 a	11.60 ± 0.80 a

Note: Rs rate: soil respiration; MBC: microbial biomass C; Values with different letters in a column mean significant difference at $p < 0.05$, values are means of three replicates ± SE.

significantly higher 45.4%, 46.8%, 49.0% (N), and 46%, 32.9%, 54.1% (N + P) compared with that in the CK treatment, respectively. However, additional phosphorus in the N + P treatment application into soils increased the relative abundance of the family Micrococcaceae, Nocardioidaceae and Chitinophagaceae (Table 6).

4. Discussions

4.1. Responses of soil respiration to N and N + P fertilization

The increased mean annual soil respiration (by 18%) after long-term N fertilization (Table 1), clearly illustrates the stimulating effects of N fertilization to soil respiration. This is possibly because long-term N fertilization had increased the soil mineral N content on the Loess Plateau where normally has low availability of natural N in soils (Zhu, 1989). The mean annual soil respiration was also significantly positively correlated with SMBC, DOC and photosynthesis rate ($p < 0.05$) (Fig. 6). This was probably because the higher soil mineral N content under fertilized soils stimulated photosynthesis rate, soil microbial population size and microbial activity (Table 2), via either directly or through increased organic matter quality and decomposition, or enhanced carbon availability (Pregitzer et al., 2000; Uselman et al., 2000; Ball and Virginia, 2014).

Additional phosphorus in the N + P treatment significantly stimulated soil mineral N uptake and result a higher crop growth (Table 3). N + P fertilization resulted in a 26% increase in soil respiration as compared with the N fertilization, which is similar to previous study (Liu et al., 2012). Higher SMBC, DOC and photosynthesis rate in the N + P treatment contributed much to soil respiration (Table 2 and Fig. 6). In addition, the Chao1 richness which could represent the total species was highest, 36.3% and 37.4% higher than that in the N and CK treatments (Table 5). The mechanism of the effect of additional phosphorus on soil respiration may be because the increased P availability could influence multiple cellular pathways (Stitt and Hurry, 2002; Plaxton and Podesta, 2006). The increased in situ N:P ratio (Table 3), resulting in different responses to additional N, just as previous study: soil communities may inhibit soil respiration when N > P, but stimulate soil respiration when N < P (Ball and Virginia, 2014). Further investigation with isotope labeling method can be done to explore the mechanism of synthetic effects of N and additional phosphorus fertilization on soil respiration.

Significantly increased soil respiration rates per unit root biomass in the fertilized soils is possibly because of the higher soil N availability in fertilized soils (Tables 3 and 4), which probably induced greater metabolic activity per unit root mass (Burton et al., 2000). However, the hardly changed soil respiration rates per unit SOC or soil SMBC in our study was not consistent with the results for the global data set on microbial respiration per unit microbial biomass about soil litter layers reported by Spohn (2015), and the cultivated experiment during long term composting of straw by Eiland et al. (2001). In fact, these values must be interpreted with great cautions to the complex of soil respiration in situ, as the observed soil respiration rates were actually integrative values from root respiration, microbial respiration as well as SOC decomposition. Our results call for a further investigation with specific isotope analysis to effectively identify the relative contributions from microbial respiration or root respiration to soil CO_2 emission.

Table 6
Relative abundances of soil bacterial communities changed by fertilization at phylum, class, order, family, and genus levels.

Prokaryotic communities	CK	N	N + P
Phylum			
Acidobacteria	12.5 ± 1.1 b	18.17 ± 0.4 a	18.25 ± 0.81 a
Actinobacteria	19.67 ± 0.43 c	28.87 ± 0.33 a	26.14 ± 0.46 ab
Bacteroidetes	2.14 ± 0.48 a	2.19 ± 1.16 a	2.73 ± 0.01 a
Chloroflexi	4.82 ± 0.56 c	7.18 ± 0.31 ab	7.43 ± 0.15 a
WS3	0.55 ± 0.01 a	0.31 ± 0.04 b	0.39 ± 0.02 ab
Class			
Acidobacteria subdivision 6	4.58 ± 0.65 b	8.77 ± 0.36 a	8.55 ± 0.56 a
Actinobacteria	7.59 ± 0.08 b	11.83 ± 0.9 a	11.15 ± 0.33 a
Thermoleophilia	5.46 ± 0.83 b	9.69 ± 0.15 a	8.4 ± 0.18 a
Chloracidobacteria	2.85 ± 0.14 b	5.01 ± 0 a	5.03 ± 0.01 a
Thermomicrobia	0.53 ± 0.1 c	1.85 ± 0.03 a	1.37 ± 0 b
Gemmatimonadetes	4.31 ± 0.08 a	3.67 ± 0.08 a	3.07 ± 0.01 b
Gemm-3	0.51 ± 0 c	1.12 ± 0.04 a	0.86 ± 0.02 b
Order			
iii1–15	3.09 ± 0.31 b	6.4 ± 0.24 a	5.71 ± 0.05 a
Actinomycetales	7.38 ± 0.08 b	11.57 ± 0.92 a	10.86 ± 0.3 a
Acidimicrobiales	3.08 ± 0.01 a	2.92 ± 0.08 a	2.86 ± 0.07 a
Family			
Geodermatophilaceae	2.15 ± 0.26 b	3.89 ± 0.42 a	2.72 ± 0.21 ab
Gaiellaceae	1.65 ± 0.29 b	2.42 ± 0.08 a	2.43 ± 0.02 a
Micrococcaceae	1.33 ± 0.08 c	1.36 ± 0.11 b	2.22 ± 0.11 a
Nocardioidaceae	0.75 ± 0.06 c	1.38 ± 0.06 a	1.25 ± 0.04 b
Rubrobacteraceae	1.35 ± 0.11 a	0.82 ± 0.02 b	0.93 ± 0.05 b
Solirubrobacteraceae	0.67 ± 0.07 b	1.45 ± 0.03 a	11.6 ± 0.06 a
Chitinophagaceae	2.22 ± 0.22 a	0.84 ± 0.05 c	1.29 ± 0.03 b
Xanthomonadacea	1.26 ± 0.14 b	1.84 ± 0.16 a	1.96 ± 0.09 a
Sinobacteraceae	1.69 ± 0.05 a	1.09 ± 0.03 b	1.34 ± 0 b
Genus			
Actinomadura	0.07 ± 0.01 a	0.09 ± 0.01 a	0.08 ± 0.01 a
Arthrobacter	1.31 ± 0.08 b	1.31 ± 0.1 b	2.2 ± 0.1 a
Modestobacter	0.21 ± 0 b	0.44 ± 0.05 a	0.31 ± 0.02 ab
Streptomyces	0.45 ± 0.02 b	0.64 ± 0.03 a	0.64 ± 0.02 a
Modestobacter	0.21 ± 0 b	0.44 ± 0.05 a	0.31 ± 0.02 ab

Note: Values with different letters in a row mean significant difference at $p < 0.05$, values are means of three replicates ± SE.

4.2. Shifts of Q_{10} under N and N + P addition

In our study, Q_{10} was decreased in the N and N + P treatment against unfertilized soil, indicating that N fertilization could decrease sensitivity of soil respiration to temperature changes, while additional P fertilization had no significant influence. This is probably because N fertilization increased soil carbon availability through improved photosynthesis and crop growth (Table 2). This could further lead to a decrease in the C:N ratios in the residues and ease mineralization with lower activation energy required for chemical and microbial decomposition (Leifeld and

Table 5
Prokaryotic diversity indices at 97% sequence similarity of 16S rRNA gene sequence calculated based on 28,313 sequences for each sample.

Treatment	Chao1 estimator of richness	Observed species	Shannon's diversity index	OUT number
CK	3341 ± 103 b	2951 ± 99 c	9.82 ± 0.08 a	3398 ± 103 c
N	3370 ± 13 4b	3098 ± 63 ab	9.83 ± 0.05 a	4213 ± 53 ab
N + P	4592 ± 68 a	3279 ± 32 a	9.88 ± 0.04 a	4578 ± 128 a

Note: Values with different letters in a column mean significant difference at $p < 0.05$, values are means of three replicates ± SE.

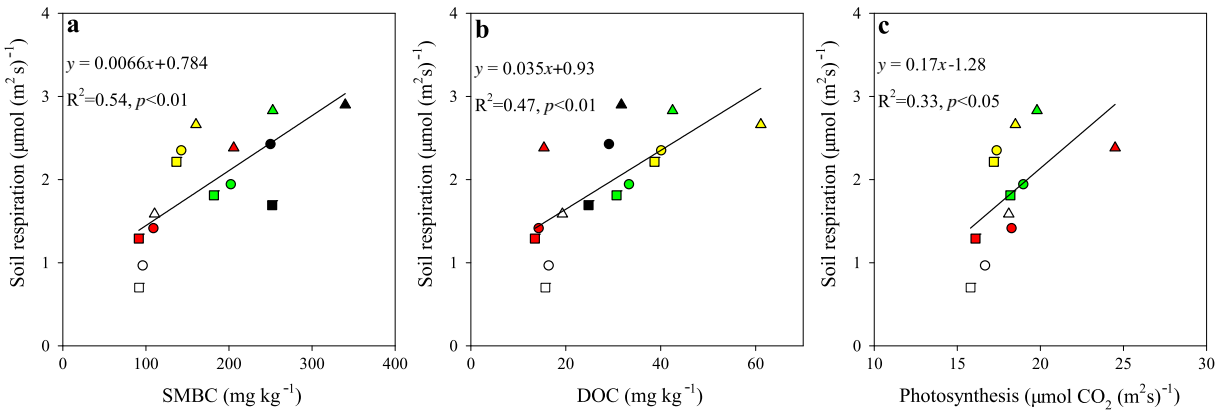

Fig. 6. Relationship between soil respiration and SMBC (a), DOC (b) and photosynthesis rate (c).

von Lutzow, 2014; Jiang et al., 2015b). With the N + P treatment, the soil mineral N content was decreased compared to N fertilization (Table 3), suggesting that the additional phosphorus in the N + P treatment significantly stimulated soil mineral N uptake and result a higher crop growth. This may further lead to greater C:N ratios in soil, which consequently resulted in increased Q_{10} (Wang et al., 2015a, 2015b). Therefore, our results were consistent with the enzyme-kinetic hypothesis which predicted degradation of low-quality substrate has a higher Q_{10} proposed by Bosatta and Agren (1999).

4.3. Potential role of microbial community in soil respiration and Q_{10}

While fertilization can affect soil respiration and Q_{10} by changing the quantity and quality of soil organic matter, N and N + P treatments also stimulated varying responses of different microbial communities (Table 6). The increased relative abundance of Acidobacteria in N application soils (N and N + P), when compared to the unfertilized soil (CK) at phylum level (Table 6), may be responsible for their greater cellulase or hemicellulose [25.54 (CK) vs. 28.66–32.40 nmol g^{-1} h^{-1} (N and N + P), unpublished data]. Those increased cellulase and hemicellulose in the fertilized soils (N and N + P) were often associated with organic carbon catalytic, which required lower total activation energy and thus potentially accelerated mineralization of low-quality carbon (Shimizu et al., 1998; Grammelis et al., 2008; Amin et al., 2014; (Dionisi et al., 2015;Wang et al., 2015a, 2015b). However, the simulative effects of additional phosphorus in soil microbial communities were not equally significant, as the relative abundance of Micrococcaceae, Nocardioidaceae and Chitinophagaceae between N and N + P treatments was only observed at the family level (Table 6). This may possibly explain the slight differences of Q_{10} between N and N + P treatments. Overall, soil microbial community structure can cast a new light into the mechanism of temperature sensitivity of soil respiration to fertilization regimes. Further investigation is required to elucidate the effects of soil microbial community on temperature sensitivity of soil respiration.

5. Conclusion

Fertilization regimes can differently affect soil respiration and temperature sensitivity. Both N fertilization alone and N + P fertilization result in an increase in soil respiration and a decrease in temperature sensitivity of soil respiration. Soil respiration was positively related to SMBC, DOC and photosynthesis rate ($p < 0.05$). Variation in Q_{10} may be related to the increase of soil mineral N content and the relative abundance of Acidobacteria in our study, Micrococcaceae, Nocardioidaceae and Chitinophagaceae might be no effect on Q_{10}. It is necessary to take the effects of fertilization regimes on Q_{10} into account when estimating CO_2 feedback to agro-ecosystem in future climate conditions.

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References

Amin, B.A.Z., Chabbert, B., Moorhead, D., Bertrand, I., 2014. Impact of fine litter chemistry on lignocellulolytic enzyme efficiency during decomposition of maize leaf and root in soil. Biogeochemistry 117, 169–183.
Ball, B.A., Virginia, R.A., 2014. Microbial biomass and respiration responses to nitrogen fertilization in a polar desert. Polar Biol. 37, 573–585.
Beauregard, M.S., Hamel, C., Nayyar, A., St-Arnaud, M., 2010. Long-term phosphorus fertilization impacts soil fungal and bacterial diversity but not AM fungal community in Alfalfa. Microb. Ecol. 59, 379–389.
Bobul'ska, L., Fazekasova, D., Angelovicova, L., Kotorova, D., 2015. Impact of ecological and conventional farming systems on chemical and biological soil quality indices in a cold mountain climate in Slovakia. Biol. Agric. Hortic. 31, 205–218.
Bosatta, E., Agren, G.I., 1999. Soil organic matter quality interpreted thermodynamically. Soil Biol. Biochem. 31, 1889–1891.
Boyer, J.N., Groffman, P.M., 1996. Bioavailability of water extractable organic carbon fractions in forest and agricultural soil profiles. Soil Biol. Biochem. 28, 783–790.
Burton, A.J., Pregitzer, K.S., Hendrick, R.L., 2000. Relationships between fine root dynamics and nitrogen availability in Michigan northern hardwood forests. Oecologia 125, 389–399.
Burton, A., Pregitzer, K., Ruess, R., Hendrick, R., Allen, M., 2002. Root respiration in North American forests: effects of nitrogen concentration and temperature across biomes. Oecologia 131, 559–568.
Caporaso, J.G., Lauber, C.L., Walters, W.A., Berg-Lyons, D., Lozupone, C.A., Turnbaugh, P.J., Fierer, N., Knight, R., 2011. Global patterns of 16S rRNA diversity at a depth of millions of sequences per sample. Proc. Natl. Acad. Sci. U. S. A. 108, 4516–4522.
Contosta, A.R., Frey, S.D., Cooper, A.B., 2015. Soil microbial communities vary as much over time as with chronic warming and nitrogen additions. Soil Biol. Biochem. 88, 19–24.
Davidson, E.A., Janssens, I.A., 2006. Temperature sensitivity of soil carbon decomposition and feedbacks to climate change. Nature 440, 165–173.
Davidson, E.A., Belk, E., Boone, R.D., 1998. Soil water content and temperature as independent or confounded factors controlling soil respiration in a temperate mixed hardwood forest. Glob. Chang. Biol. 4, 217–227.
Ding, W., Cai, Y., Cai, W., 2007. Soil respiration under maize crops: effects of water, temperature, and nitrogen fertilization. Soil Sci. Soc. Am. J. 71, 944–951.
Dionisi, D., Anderson, J.A., Aulenta, F., Mccue, A., Paton, G., 2015. The potential of microbial processes for lignocellulosic biomass conversion to ethanol: a review. J. Chem. Technol. Biotechnol. 90, 366–383.
Edgar, R.C., 2013. UPARSE: highly accurate OTU sequences from microbial amplicon reads. Nat. Methods 10, 996–998.
Edgar, R.C., Haas, B.J., Clemente, J.C., Quince, C., Knight, R., 2011. UCHIME improves sensitivity and speed of chimera detection. Bioinformatics 27, 2194–2200.
Eiland, F., Klamer, M., Lind, A.M., Leth, M., Baath, E., 2001. Influence of initial C/N ratio on chemical and microbial composition during long term composting of straw. Microb. Ecol. 41, 272–280.
Fernandez-Escobar, R., Marin, L., Sanchez-Zamora, M.A., Garcia-Novelo, J.M., Molina-Soria, C., Parra, M.A., 2009. Long-term effects of N fertilization on cropping and growth of olive trees and on N accumulation in soil profile. Eur. J. Agron. 31, 223–232.
Fujii, K., Hartono, A., Funakawa, S., Uemura, M., Kosaki, T., 2011. Fluxes of dissolved organic carbon in three tropical secondary forests developed on serpentine and mudstone. Geoderma 163, 119–126.
Grammelis, P., Malliopoulou, A., Basinas, P., Danalatos, N.G., 2008. Cultivation and characterization of *Cynara cardunculus* for solid biofuels production in the Mediterranean region. Int. J. Mol. Sci. 9, 1241–1258.

Grimshaw, H.M., Allen, S.E., Parkinson, J.A., 1989. Nutrient elements. In: Allen, S.E. (Ed.), Chemical Analysis of Ecological Material. Blackwell Scientific, Oxford, p. 81e159.
Hopkins, D., Sparrow, A., Shillam, L., English, L., Dennis, P., Novis, P., Elberling, B., Gregorich, E., Greenfield, L., 2008. Enzymatic activities and microbial communities in an Antarctic dry valley soil: responses to C and N supplementation. Soil Biol. Biochem. 40, 2130–2136.
Iqbal, J., Hu, R.G., Feng, M.L., Lin, S., Malghani, S., Ali, I.M., 2010. Microbial biomass, and dissolved organic carbon and nitrogen strongly affect soil respiration in different land uses: a case study at three gorges reservoir area, South China. Agric. Ecosyst. Environ. 137, 294–307.
Janssens, I.A., Pilegaard, K., 2003. Large seasonal changes in Q_{10} of soil respiration in a beech forest. Glob. Chang. Biol. 9, 911–918.
Jia, S.X., Wang, Z.Q., Li, X.P., Sun, Y., Zhang, X.P., Liang, A.Z., 2010. N fertilization affects on soil respiration, microbial biomass and root respiration in *Larix gmelinii* and *Fraxinus mandshurica* plantations in China. Plant Soil 333, 325–336.
Jiang, J., Shi, P.L., Zong, N., Fu, G., Shen, Z.X., Zhang, X.Z., Song, M.H., 2015a. Climatic patterns modulate ecosystem and soil respiration responses to fertilization in an alpine meadow on the Tibetan plateau. China. Ecol. Res. 30, 3–13.
Jiang, J.S., Guo, S.L., Zhang, Y.J., Liu, Q.F., Wang, R., Wang, Z.Q., Li, N.N., Li, R.J., 2015b. Changes in temperature sensitivity of soil respiration in the phases of a three-year crop rotation system. Soil Tillage Res. 150, 139–146.
Jiang, J.S., Guo, S.L., Wang, R., Liu, Q.F., Wang, Z.Q., Zhang, Y.J., Li, N.N., Li, R.J., Wu, D.F., Sun, Q.Q., 2015c. Effects of nitrogen fertilization on soil respiration and temperature sensitivity in spring maize field in semi-arid regions on Loess Plateau. Environ. Sci. 36, 1802–1809.
Johnson, D.W., Cheng, W., Ball, J.T., 2000. Effects of CO_2 and nitrogen fertilization on soils planted with ponderosa pine. Plant Soil 224, 99–113.
King, J.S., Albaugh, T.J., Allen, H.L., Buford, M., Strain, B.R., Dougherty, P., 2002. Belowground carbon input to soil is controlled by nutrient availability and fine root dynamics in loblolly pine. New Phytol. 154, 389–398.
Kirschbaum, M.U.F., 1995. The temperature dependence of soil organic matter decomposition: and the effect of global warming on soil organic C storage. Soil Biol. Biochem. 27, 753–760.
Leifeld, J., von Lutzow, M., 2014. Chemical and microbial activation energies of soil organic matter decomposition. Biol. Fertil. Soils 50, 147–153.
Li, F.L., Liu, M., Li, Z.P., Jiang, C.Y., Han, F.X., Che, Y.P., 2013. Changes in soil microbial biomass and functional diversity with a nitrogen gradient in soil columns. Appl. Soil Ecol. 64, 1–6.
Liu, L., Gundersen, P., Zhang, T., Mo, J.M., 2012. Effects of phosphorus addition on soil microbial biomass and community composition in three forest types in tropical China. Soil Biol. Biochem. 44, 31–38.
Magoč, T., Salzberg, S.L., 2011. FLASH: fast length adjustment of short reads to improve genome assemblies. Bioinformatics 27, 2957–2963.
Majdi, H., 2001. Changes in fine root production and longevity in relation to water and nutrient availability in a Norway spruce stand in northern Sweden. Tree Physiol. 21, 1057–1061.
Marschner, B., Kalbitz, K., 2003. Controls of bioavailability and biodegradability of dissolved organic matter in soils. Geoderma 113, 211–235.
Pan, Y., Cassman, N., de Hollander, M., Mendes, L.W., Korevaar, H., Geerts, R., van Veen, J.A., Kuramae, E.E., 2014. Impact of long-term N, P, K, and NPK fertilization on the composition and potential functions of the bacterial community in grassland soil. FEMS Microbiol. Ecol. 90, 195–205.
Plaxton, W.C., Podesta, F.E., 2006. The functional organization and control of plant respiration. Crit. Rev. Plant Sci. 25, 159–198.
Pregitzer, K.S., Zak, D.R., Maziasz, J., DeForest, J., Curtis, P.S., Lussenhop, J., 2000. Interactive effects of atmospheric CO_2 and soil-N availability on fine roots of *Populus tremuloides*. Ecol. Appl. 10, 18–33.
Qi, Y., Xu, M., 2001. Separating the effects of moisture and temperature on soil CO_2 efflux in a coniferous forest in the sierra Nevada mountains. Plant Soil 237, 15–23.
Ramirez, K.S., Craine, J.M., Fierer, N., 2012. Consistent effects of nitrogen amendments on soil microbial communities and processes across biomes. Glob. Chang. Biol. 18, 1918–1927.
Schleser, G., 1982. The response of CO_2 evolution from soils to global temperature changes. Z. Naturforsch. 37, 287–295.
Shimizu, K., Sudo, K., Ono, H., Ishihara, M., Fujii, T., Hishiyama, S., 1998. Integrated process for total utilization of wood components by steam-explosion pretreatment. Biomass Bioenergy 14, 195–203.
Shukla, S.K., Singh, P.N., Chauhan, R.S., Solomon, S., 2015. Soil physical, chemical and biological changes and long term sustainability in subtropical India through integration of organic and inorganic nutrient sources in sugarcane. Sugar Tech 17, 138–149.
Sparks, D.L., Page, A.L., Helmke, P.A., Loeppert, R.H., Soltanpour, P.N., Johnston, C.T., Sumner, M.E., 1996. Methods of soil analysis. Part 3: Chemical Methods. Soil Science Society of America Inc., Madison, WI.
Spohn, M., 2015. Microbial respiration per unit microbial biomass depends on litter layer carbon-to-nitrogen ratio. Biogeosciences 12, 817–823.
Stitt, M., Hurry, V., 2002. A plant for all seasons: alterations in photosynthetic carbon metabolism during cold acclimation in Arabidopsis. Curr. Opin. Plant Biol. 5, 199–206.
Straathof, A.L., Chincarini, R., Comans, R.N.J., Hoffland, E., 2014. Dynamics of soil dissolved organic carbon pools reveal both hydrophobic and hydrophilic compounds sustain microbial respiration. Soil Biol. Biochem. 79, 109–116.
Tully, K.L., Wood, S.A., Almaraz, M., Neill, C., Palm, C., 2015. The effect of mineral and organic nutrient input on yields and nitrogen balances in western Kenya. Agric. Ecosyst. Environ. 214, 10–20.
Turlapati, S.A., Minocha, R., Bhiravarasa, P.S., Tisa, L.S., Thomas, W.K., Minocha, S.C., 2013. Chronic N-amended soils exhibit an altered bacterial community structure in Harvard Forest, MA. USA. FEMS Microbiol. Ecol. 83, 478–493.
Uselman, S.M., Qualls, R.G., Thomas, R.B., 2000. Effects of increased atmospheric CO_2, temperature, and soil N availability on root exudation of dissolved organic carbon by a N-fixing tree (*Robinia pseudoacacia* L.). Plant Soil 222, 191–202.
van Gestel, N.C., Schwilk, D.W., Tissue, D.T., Zak, J.C., 2011. Reductions in daily soil temperature variability increase soil microbial biomass C and decrease soil N availability in the Chihuahuan Desert: potential implications for ecosystem C and N fluxes. Glob. Chang. Biol. 17, 3564–3576.
Vance, E.D., Brookes, P.C., Jenkinson, D.S., 1987. An extraction method for measuring soil microbial biomass-C. Soil Biol. Biochem. 19, 703–707.
Vitousek, P.M., Porder, S., Houlton, B.Z., Chadwick, O.A., 2010. Terrestrial phosphorus limitation: mechanisms, implications, and nitrogen-phosphorus interactions. Ecol. Appl. 20, 5–15.
Wakelin, S., Mander, C., Gerard, E., Jansa, J., Erb, A., Young, S., Condron, L., O'Callaghan, M., 2012. Response of soil microbial communities to contrasted histories of phosphorus fertilisation in pastures. Appl. Soil Ecol. 61, 40–48.
Wang, Q., Garrity, G.M., Tiedje, J.M., Cole, J.R., 2007. Naive Bayesian classifier for rapid assignment of rRNA sequences into the new bacterial taxonomy. Appl. Environ. Microbiol. 73, 5261–5267.
Wang, R., Guo, S., Jiang, J., Wu, D., Li, N., Zhang, Y., Liu, Q., Li, R., Wang, Z., Sun, Q., Du, L., Zhao, M., 2015a. Tree-scale spatial variation of soil respiration and its influence factors in apple orchard in Loess Plateau. Nutr. Cycl. Agroecosyst. 102, 285–297.
Wang, X., Shi, Y., Guo, Z.J., Zhang, Y.L., Yu, Z.W., 2015b. Water use and soil nitrate nitrogen changes under supplemental irrigation with nitrogen application rate in wheat field. Field Crop Res. 183, 117–125.
Wood, S.A., Bradford, M.A., Gilbert, J.A., McGuire, K.L., Palm, C.A., Tully, K.L., Zhou, J., Naeem, S., 2015. Agricultural intensification and the functional capacity of soil microbes on smallholder African farms. J. Appl. Ecol. 52, 744–752.
Xu, M., Qi, Y., 2001. Spatial and seasonal variations of $Q_{(10)}$ determined by soil respiration measurements at a Sierra Nevadan forest. Global Biogeochem. Cy. 15, 687–696.
Yao, M., Rui, J., Li, J., Dai, Y., Bai, Y., Heděnec, P., Wang, J., Zhang, S., Pei, K., Liu, C., 2014. Rate-specific responses of prokaryotic diversity and structure to nitrogen deposition in the *Leymus chinensis* steppe. Soil Biol. Biochem. 79, 81–90.
Yu, W.T., Bi, M.L., Xu, Y.G., Zhou, H., Ma, Q., Jiang, C.M., 2013. Microbial biomass and community composition in a Luvisol soil as influenced by long-term land use and fertilization. Catena 107, 89–95.
Zhang, Y.J., Guo, S.L., Liu, Q.F., Jiang, J.S., Wang, R., Li, N.N., 2015. Responses of soil respiration to land use conversions in degraded ecosystem of the semi-arid Loess Plateau. Ecol. Eng. 74, 196–205.
Zhong, Y.Q.W., Yan, W.M., Shangguan, Z.P., 2015. Soil carbon and nitrogen fractions in the soil profile and their response to long-term nitrogen fertilization in a wheat field. Catena 135, 38–46.
Zhu, X.M., 1989. Soils and Agriculture in the Loess Plateau. Agriculture Press, Beijing, pp. 286–297.