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Response of Soil Respiration to Nitrogen Addition in Two Subtropical Forest Types*1

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ABSTRACT

Anthropogenic activities have increased the quantity of nitrogen (N) deposition in terrestrial ecosystems, which directly and indirectly affected soil biogeochemical processes including soil respiration. However, the effects of increasing N availability on soil respiration are still not fully understood. In this study, soil respiration was measured using an infrared gas analyzer system with soil chambers under four N addition level treatments (0, 5, 15 and 30 g N m^{-2} yr^{-1} as control, low N addition (LN), mediate N addition (MN) and high N addition (HN), respectively) in Camphor tree and Slash pine forests in subtropical China. Results showed that soil respiration rates were reduced in the fertilized plots respect to control plots on average by 37% in the Camphor tree forests and 27% in the Slash pine forests on an annual base, respectively. No significant differences were found in soil respiration between the LN, MN and HN treatments in both forest types because these fertilized plots reached an 'adequate N content zone'. In addition, soil microbial biomass carbon (C) content and fine root biomass were declined in N addition plots compared to the control during the study period. Our results indicated that elevated N deposition might alter the patterns of tree growth, C partitioning and microbial activity, which further affect soil C sequestration through reducing soil respiration in

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subtropical forests of China.

Key words: nitrogen, soil moisture, soil respiration, soil temperature, fine roots, soil microbial biomass

INTRODUCTION

Anthropogenic activities have caused significant changes in a number of biological, ecological and hydrological processes at global scale (IPCC, 2007). Such global environmental change has led to a considerable alteration in soil processes in terrestrial ecosystems, such as soil CO_2 efflux (i.e., soil respiration) (Borken and Matzner, 2009), carbon (C) sequestration and nitrogen (N) mineralization (Barnard *et al*., 2006; Tian *et al*., 2010). In return, the changed soil status may positively or negatively feedback to the global environmental changes. Understanding of belowground dynamics response to global change factors, such as elevated temperature, altered precipitation pattern and N deposition, is critical in assessing soil C cycling and long-term sustainability of forest soils (Lee and Jose, 2003).

Nitrogen deposition has been rapidly increasing across many terrestrial ecosystems worldwide in the past century due primarily to intensive agriculture and fossil fuel combustion (Vitousek *et al*., 1997). In some parts of the European and the American forests, for example, it is estimated that the N deposition are nowadays between 5- and 50-fold higher compared to the pre-industrial levels (Dentener *et al*., 2006). The increase of N deposition is particularly true in Asia due to expansion of industrial and agricultural activities. Zheng *et al*. (2002) reported that the use and emission of N increased from 14 Tg N yr^{-1} in 1961 to 68 Tg N yr^{-1} in 2000 and were expected to reach 105 Tg N yr^{-1} in 2030. Annual atmospheric N deposition through precipitation has also been significantly increasing in southern China, from about 13.5 kg N ha^{-1} yr^{-1} (Gholz *et al*., 1985) to 30 - 73 kg N ha^{-1} yr^{-1} (Ma, 1989; Xu *et al*., 2001). A wide range of studies have been conducted to investigate the linkage between the increasing N deposition and soil respiration processes, but the studies have yielded conflicting results. Increasing, decreasing, and stable in soil respiration in response to N addition have been reported (Fisk and Fahey, 2001; Fernandes *et al*., 2002; Bowden *et al*., 2004; Burton *et al*., 2004; Olsson *et al*., 2005; Mo *et al*., 2008). Therefore, more data and comprehensive research are needed to address the response of soil respiration process to the increasing N deposition, including the application of N fertilizers.

Camphor tree (*Cinnamomum camphora*) is one of the most common tree species in both urban and natural forests in southern China. The species is an important tree component in the evergreen broadleaves forests which are the climax vegetation community in sub-tropical region of the country (Tian, 2008). Slash pine (*Pinus elliottii* Engelm) is an alien tree species from American. Due to advantage characteristics of fast-growth and tolerance ability to drought and infertile lands, Slash pine has widely planted in China since 1980s and has become one of the major silvicultural conifer species for timber-production in southern China. So far, more than two million hectares of Slash pine plantations have been established in the country (Ma *et al*., 2011).

In this study, we conducted an experiment to investigate the effects of changes in N deposition on soil respiration in Camphor tree and Slash pine plantations in subtropical region of China. The overall objective of the project was to examine the response of soil respiration to environmental changes due to anthropogenic activities. We hypothesized that: (1) N fertilizer additions in soils would reduce soil respiration; (2) Low and high N additions may have different effects on seasonal patterns of soil CO_2 efflux; and (3) The feedbacks of soil respiration to N fertilizer would differ in camphor tree and in slash pine stands because of different life-forms of the two tree species.

MATERIALS AND METHODS

Study site

The study site was located at Hunan Forest Botany Park in Changsha, Hunan Province, China (113°02′~01′E, 28°06′~07′N). Elevation was 46-114 m with site slope of 5-15^{o}. The study area was a typical moist subtropical zone with a mean annual temperature of 17.2^{0}C. The minimum and maximum mean monthly temperatures were 4.7^{0}C and 29.4^{0}C in January and July, respectively. Mean annual rainfall was 1422 mm, most of which occurred from April to August. Mean annual relative humidity was higher than 80%. The soil was a typical yellow earth with an acidic pH on the top soil (0-10 cm) ranging between 4.0 and 5.0. The mean wet N deposition rate for study area between 2006 and 2016 was 39.2 kg N ha^{-1} yr^{-1}, with minimum and maximum values of 25 and 55.1 kg N ha^{-1} yr^{-1}, respectively, with roughly 6 : 1 NH_4^+ to NO_3^- molar ratio (Authors' unpublished data). The forests in the park were mainly artificially planted as pure or mixed forests in 1982 with an initial tree density of 2m × 2m or 2m × 3m. The major tree species of the plantations include Masson pine (*Pinus massoniana*), Chinese fir (*Cunninghamia lanceolata* (Lamb. Hook.), Chinese sweet gum (*Liquidambar acalycina*), Camphor tree (*Cinnamomum camphora*) and Slash pine (*Pinus elliottii*). Understory consisted of *Sassafras tsumu* Hemsl.; *Symplocos caudata* Wall. ex A. DC.; *Clerodendron cyrtophyllum* Turcz; *Nephrolepis auriculata* Trimen; *Lophantherum gracile* Brengn.; *Miscanthus floridulus* Warb and *Phytolacca acinosa* Roxb.

Experimental design

In the present study, two types of forests, Camphor tree forest (a broad-leaves forest type) and Slash pine forest (a conifer forest type) were chosen in the study site. Three forest stands (50 m × 50 m in size each) were selected for each forest type. Four plots (20 m × 20 m in size each) were set up in each of the forest stands. N fertilizer was supplied as NH_4NO_3 at four treatment levels: 0 (control), 5 (low N input, LN), 15 (mediate N input, MN) and 30 g m^{-2} (high N input, HN). Each fertilizer treatment level had three replications. The fertilizer was dissolved in 20 L of water and was evenly spread in each of the plots. Two sampling locations were established at each plot. There were thus a total of 48 measurement points in the study (2 forest types × 3 stands × 4 plots (representing 4 fertilizer treatment levels) × 2 measurement points).The fertilizer treatment was performed in May of 2010, as one-time fertilizer application. The initial soil characteristics of the experiment site are shown in Table 1.

Table 1

Soil respiration measurements

Soil respiration was measured on a biweekly basis from August 2010 to July 2011 using a portable infra-red gas analyzer (LI-COR 8100) with soil chamber (LI-COR inc, Lincoln, Nebraska, USA) (Yan *et al*., 2013). At each sampling location, a PVC collar (11.7 cm in diameter, 4.4 cm in height) was installed, leaving 2.5 cm protruding above the soil surface. In order to minimize soil disturbance from the deployment of the flux chamber, PVC collars were placed into the soil at least one week prior to first field measurement and remained in place through the study. The soil respiration value at each measurement point was the mean of the three sequential flux estimates at each sampling interval. Values were express as μmol $CO_2 m^{-2} s^{-1}$, and converted to mg C $m^{-2} h^{-1}$. For each plot, data collected from the two measurement points were averaged, and then the three replicate forest stands were averaged for each measurement time (biweekly) and calculated for each month in the study period of time. The annual soil respiration was accumulated based on the monthly basis.

Soil microbial biomass carbon and nitrogen measurements

For the measurements of soil microbial biomass carbon (SMBC) and soil microbial biomass nitrogen (SMBN), soil samples were collected from each of the four fertilized plots in July and October in 2010 and April and July in 2011, respectively. Soil was sampled to a depth of 20 cm from each of the plots. Litter on the soil surface was removed before sampling. Rocks and roots were carefully removed from the soil samples by hand. Samples were sieved (2 mm) and stored at 4^0C until analyzed. The SMBC and SMBN were measured using the chloroform fumigation extraction method (Brookes *et al*., 1985; Vance *et al*., 1987). Microbial biomass C was calculated as EC/ kEC, where EC= (organic C extracted from fumigated soil) – (organic C extracted from non-fumigated soil) and kEC = 0.45. Microbial biomass N was calculated as EN/ kEN, where EN = (total N extracted from fumigated soil) – (total N extracted from non-fumigated soil) and kEN = 0.54 (Brookes *et al*., 1985; Vance *et al*., 1987).

Fine root biomass measurement

For fine roots (< 2 mm) biomass measurement, a sharp-edge steel soil corer, 10.0 cm diameter, was employed to collect soil samples to a depth of 20 cm. Soil cores were taken at three random locations within each plot. Cores were immediately delivered to the laboratory and roots were extracted from the cores by careful hand-sorting. Fine roots were dried at 70^0C until they reached a constant weight.

Soil temperature and water content measurements

Soil temperature (T_{soil}) and soil volumetric water content (W_{soil}) were measured at the same time during the measurements of soil respiration using a soil thermocouple probe (LI-COR 8100-201 Omega) and a water content probe (LI-COR 8100-202, LI-COR Inc, Lincoln, Nebraska, USA) at 10 cm below the soil surface.

Statistical analysis

Statistical tests for the effects of two forest types and N fertilizer vs. control plots on soil respiration, SMBC, SMBN, fine root biomass, T_{soil}, and W_{soil} were performed using analysis of variance (ANOVA). The original soil respiration data were log-transformed to satisfy the normality and homoscedasticity assumptions of ANOVA; no transformation was necessary for SMBC, SMBN, fine root biomass, T_{soil} and W_{soil}. Multiple comparisons were conducted to identify levels of N fertilizer treatments differences in soil respiration and T_{soil} and W_{soil}. Pair-wise t-tests were used to examine differences between two forest types. Multiple regression analysis was employed to examine relationships between soil respiration and T_{soil} and W_{soil}. Statistical analyses were conducted using the SAS statistical package (SAS Institute, Inc., Cary, NC 1999-2001), with a confidence level of $P < 0.05$.

RESULTS

A significant difference was found in terms of soil respiration between the forest types during the study period ($P < 0.0001$). Nitrogen addition had significant effects on soil respiration ($P = 0.0029$), but no significant differences were found in soil respiration between the LN, MN and HN treatments in both forest types ($P > 0.05$). The interaction of forest types coupled with N fertilizer did not significantly affect soil respiration ($P = 0.9035$). No significant differences were found in mean annual T_{soil} (P > 0.05) and W_{soil} ($P > 0.05$) among the N addition treatments in both forest types.

On average, soil respiration rate in control plots was higher in camphor tree forests than in slash pine forests (Table 2). Nitrogen addition treatments significantly reduced soil respiration rates in the two forest types. In Camphor tree forests annual soil respiration was 10.94 ± 1.29, 6.89 ± 0.87, 6.67 ± 0.67 and 6.98 ± 0.80 t C ha^{-1} yr^{-1} in the control, LN, MN and HN plots, respectively, meaning that N addition significantly reduced soil respiration rates by 37.0%, 39.0% and 36.2% at the LN, MN and HN plots (with a mean of 37%) compared with the control plots ($P < 0.05$). In Slash pine forests the annual soil CO_2 efflux was 6.02 ± 0.81, 4.35 ± 0.52, 4.68 ± 0.52 and 4.23 ± 0.40 t C ha^{-1} yr^{-1} at the control, LN, MN and HN plots, respectively, meaning that soil respiration rates were reduced by 27.7%, 22.3% and 29.9% in LN, MN and HN treatments (with a mean of 27%) when compared with control treatments. Such reduction of soil respiration was significant ($P < 0.05$).

Table 2

Soil respiration showed a strong seasonal pattern, with relatively low rates during winter months and high rates in summer times in both forest stands (Figure 1). In the control plots, soil respiration rates varied from about 1.28 μmol m^{-2} s^{-1} during winter months to 4.83 μmol m^{-2} s^{-1} during the summer months in Camphor tree stands and ranged from 0.82 μmol m^{-2} s^{-1} in winter times to 2.45 μmol m^{-2} s^{-1} in summer times in slash pine stands during the study period. Nitrogen additions did not change the seasonal pattern of soil respiration, but decreased its values almost in the corresponding months in both studied forests. For instance, in the fertilized plots, the seasonal variation of soil respiration rates ranged from 0.72 μmol m^{-2} s^{-1} in winter, to 3.23 μmol m^{-2} s^{-1} in summer in Camphor tree forests and from 0.75 μmol m^{-2} s^{-1} to 1.65 μmol m^{-2} s^{-1} in summer in slash pine forests, respectively (Figure 1). Soil respiration rates always higher in Camphor tree stands than the slash pine stands. Soil respiration was strongly correlated with T_{soil} ($P < 0.0001$) but not W_g ($P > 0.05$). The strong relationship between soil respiration and T_{soil} can be found in both forests in Figure 2.

Figure 1 **Figure 2**

Soil microbial biomass carbon (SMBC) content in the control plots ranged from 87.6 to 155.5 mg kg^{-1} in Camphor tree forests, and from 76.2 to 226.1 mg kg^{-1} in Slash pine forests. Nitrogen application initially significantly increased the SMBC content in Camphor tree stands ($P < 0.05$), but such effect was vanished one year later (Apr. 2011) when compared to the control plots (Figure 3A). The SMBC contents were reduced by about 20, 12 and 7% in LN, MN and HN treatment plots in the Camphor tree forests one year post-treatment. No significant differences were observed for SMBC content between the three N level treatment plots in Camphor tree forests ($P > 0.05$), except HN treatment in Oct. 2010. The seasonal changes in SMBC content in Slash pine forests were similar with that in Camphor tree forests. Nitrogen fertilizer significantly increased the SMBC content after the N application ($P > 0.05$), except in LN plots where SMBC content was still kept lower than that in control plot. One year later after the treatment, SMBC content was significantly lower than the control plots by approximately 23, 39 and 26% in the Slash pine forest in LN, MN and HN treatment plots, respectively (Figure 3B). The SMBC contents did not significantly differ among the fertilized stands in the Slash pine forests during the study period, except in Oct. 2010 when the SMBC content was significantly lower in LN plots than in MN and HN plots.

Figure 3

In Camphor tree forests N addition significantly increased SMBN content in fertilized plots two months later after fertilization compared to the control plots ($P < 0.05$). The SMBN content remained higher in fertilized plots until one year after fertilization (Figure 4A). No significant differences were found in SMBN content between fertilized plots and control plots in Camphor tree stands after Apr. 2011 (Figure 4A). On the contrary, N addition did not change the seasonal pattern of the SMBN content in in Slash pine forests (Figure 4B). Although the SMBN contents were low in N treated plots most of the study time compared to the control, there were no significant differences in terms of the SMBN contents between the fertilized plots and control plots in Slash pine forests during the course of the study ($P > 0.05$), except LN plot in Jul. 2010 when the SMBN content was significant higher in LN plot than all other studied plots (Figure 4B).

Figure 4

Fine root biomass was lower in fertilized plots than in control plots in both examined forests (Figure 5). However, the reduced-effect was significant only in the Camphor tree forests ($P < 0.05$) and not in the Slash pine forests ($P > 0.05$). When compared to the control plots, fine root biomass in Camphor tree forests was reduced by 14, 15 and 20% in LN, MN and HN treatments, respectively. The corresponding reduction was 5, 6 and 9% in LN, MN and HN treatments in Slash pine forests. There were no significant treatment effects among the fertilized treatment plots in both species.

Figure 5

DISCUSSION

In the present study, our results showed that at N addition rates of 50 to 300 Kg N ha^{-1} yr^{-1}, soil respiration was significantly declined by 36-39% in Camphor tree forests and 22-30% in Slash pine forests on an annual basis. The reduction of soil respiration occurred at the same order of magnitude in the three N application levels over the entire period of observation and was greater in summer times than in winter (Figure 1). Additionally, N addition treatments induced the decrease of SMBC content by 7-20% and 23-39%, and fine root biomass by 14-20% and 5-9% in Camphor tree forests and Slash pine forests, respectively (Figure 3 and 5). The decrease of soil respiration under N addition treatments was in agreement with earlier findings (Phillips and Fahey, 2007). For instance, compared to the control experiments, N addition resulted in a reduction of soil respiration rates by 14% in evergreen broadleaf forests (Mo *et al*., 2008), by 8 and 14% and 8% in a montane forest (Koehler *et al*., 2009), by 18% in a Panamanian forest (Giardina *et al*., 2004) and by 30% and 24% in larch and ash plantations, respectively (Jia *et al*., 2010).

The decrease of soil respiration due to N fertilization may be explained by several potential mechanisms. For example, such reduction might be related to the suppression of fine root growth following N addition (Mo *et al*., 2008; Liu *et al*., 2009). It was generally recognized that increasing soil N availability often leads to decline in fine root biomass (Nadelhoffer *et al*., 1985). Bowden *et al*. (2004) reported that root biomass in upper soil layer (0–20 cm) was higher in the control plots than in N addition plots in a temperate forest. Using in-growth core method, Liu *et al*. (2009) investigated the impact of fertilization on fine root growth in larch and ash plantations and found that fine root production was declined about by 50-70% in the two forest types due to the fertilization. Mo *et al*. (2008) indicated that fine root biomass was significantly lower in N application plots compared with the control plots and exhibited a negative response to increasing level of N addition. Fertilizer application can promoted seedlings and tree growth and likely allocated more carbohydrate production into belowground parts when the forests were in a young and fast-growing phase (Pangle and Seiler, 2002). Therefore, a relatively high soil respiration was determined by a high root growth and biomass accumulation in seedlings and young forest stands (Pangle and Seiler, 2002; Peng and Thomas, 2010). The phenomenon was also observed in a six young seedlings experiment in southern China (Deng *et al*., 2010). They found that N addition facilitated root growth and root biomass production, and therefore enhanced soil respiration rates (Deng *et al*., 2010). However, when forests were in a mature stage, additions of limiting nutrients are likely to drive the partition of less carbohydrate to belowground compartments, resulting in a reduction in soil respiration (Boxman *et al*., 1998; Peng and Thomas, 2010). By analyzing the patterns of annual C allocation for 63 forests from tropical, temperate and boreal regions, Litton *et al*. (2007) reported that enhanced nutrient availability generally caused more C partitioning to aboveground components and decreased partitioning to the belowground annual production. It is likely a common phenomenon that a shift in carbon allocation between aboveground and belowground parts in the fertilized plots (Giardina *et al*., 2004; Koehler *et al*., 2009). Furthermore, when N becomes a non-limiting element for plant growth, N addition may decrease tree growth (Magill *et al*., 2000) and the excess of N may damage forests by causing nutrient imbalances in the forested ecosystems (Erisman and de Vries, 2000).

The second mechanism explaining the decline of soil respiration might partly be attributed to suppression of microbial activity due to N fertilization (Foster *et al*., 1980; Mo *et al*., 2008; Jia *et al*., 2010). In our study, it seemed that N application initially significantly increased the SMBC contents in both Camphor tree and Slash pine forests until Oct. 2010, and SMBN contents in the two studied forest types until Jul. 2010 as well. But then the SMBC and SMBN contents were lower in fertilized plots than in unfertilized plots in both species, except the SMBN content in Camphor tree stands (Figure 3 and 4). Our results were consistent with other findings that showed N addition reduced soil microbial biomass over time (Compton *et al*., 2004; Mo *et al*., 2008). Aber *et al*. (1989) indicated that N additions to the N deficient soils would initially motivated soil microbial activity, but over time would lead to a C limited state after microbial demand for N was satisfied. It was reported that when compared to the control, the reduced of SMBC and SMBN due to N addition were about by 34% and 37 in six northern hardwood forests stands (Fisk and Fahey, 2001), and 29-39% and 42-47% in larch and ash stands (Jia *et al*., 2010). The reduction in SMBC and SMBN under N fertilizer application treatments might be due to the high level of mineral N availability (Lovell *et al*., 1995), changes in substrate quality and variation in microbial competition and community structure, repression of enzyme activity and the build-up of recalcitrant and toxic compounds (Fog, 1988). Nitrogen fertilizer application often led to soil acidification which limited soil microbial activity and soil organic matter decomposition rates (Malhi *et al*., 1998). The decreased decomposition rates further resulted in the reduction of microbial respiration in soils (Burton *et al*., 2004). Additionally, there are indications that input of mineral N retards decomposition rates of old litter and recalcitrant SOM by suppression of ligninolytic enzymes of soil microbes and by chemical stabilization. Nitrogen stimulates the initial decomposition of fresh litter, but suppresses humus decay in later stages (Ladd *et al*., 1994).

The third mechanism explaining the decrease of soil respiration due to N fertilization application might also be attributed to the fertilizer-induced impacts on microbial diversity, metabolic activity, mycorrhizal colonization, litter substrate quantity and quality, and production of extracellular enzymes by soil microbes (Boxman *et al*.. 1998; Allison *et al*., 2009; Samuelson *et al*., 2009). The initial nutrient level in soils might influence the response of soil respiration to increasing N deposition (Mo *et al*., 2008). The response of soil respiration to fertilizer application was different due to forest ages and stand growth and development phases (Huang *et al*., 2011). Additionally, the different sensitivities of roots and microbial communities to changes in their surrounding environmental variables resulted in the different response of autotrophic respiration and heterotrophic respiration to increasing global N deposition (Tian *et al*., 2011). As a result, comprehensive studies are necessary for a thorough evaluation of the response of the soil respiration process under global environmental changes and forest management practices.

Surprisingly, there was no significant difference in soil respiration between the LN, MN and HN treatments in the studied forest types. The mechanism for the similar soil respiration among the fertilized plots in our experiment remains unclear, but one of the probable reasons may be a result that all the fertilized plots reached an ‘adequate N content zone’. We thought that when an ‘adequate soil N content zone’ was reached, further addition of N in soils was no longer related to decreases in soil respiration. Some other investigators have found similar ‘adequate soil N content zone’ phenomenon. For example, Bowden et al. (2004) reported that N fertilization significantly decreased annual soil respiration rates in a red pine forest, but the treatment effect did not differ significantly between low N addition (5 g m^{-2} yr^{-1}) and high N addition (15 g N m^{-2} yr^{-1}) plots. Lee and Jose (2003) measured soil respiration along a N fertilization gradient (0, 5.6, 11.2, and 22.4 g N m^{-2} yr^{-1}) in 7-year-old loblolly pine plantation in northwest Florida and found that there was no significant treatment effect on these fertilized plots. Understanding the mechanisms related to belowground processes under 'adequate soil N content zone' will be essential to predict responses of forest ecosystems to elevated N deposition. Clearly, the causes and effects of soil respiration versus N limitation and adequation in soil require further study.

Although N addition significantly reduced soil respiration rates in both Camphor tree and Slash pine forests (Figure 1), the belowground response to N fertilization varied between the both species. Nitrogen fertilization had a significantly negative effect on fine root biomass in Camphor tree species, but no effect was observed in Slash pine species (Figure 5). The effects of N addition on seasonal patterns of soil microbial community (represented by SMBC and SMBN) were likely similar in both species (Figure 3 and 4), but the response of the ratio of SMBC to SMBN to N addition was totally different in these two forest types. Compared with the control, the ratio of SMBC to SMBN was always lower in the fertilized plots in Camphor tree stand. In contrast, the SMBC: SMBN ratio mostly increased in the fertilized plots in Slash pine stands (Figure 6). The different response to N fertilization in terms of the ratio of SMBC to SMBN between the both species might reflect the different composition and activity of microbial community in these two forest soils (Tian *et al*. 2008)

In our study, soil respiration rates exhibited an obvious seasonal change. Nitrogen addition did not altered seasonal pattern of soil respiration but modify the seasonal minimum and maximum values of the respiration in the two forest types (Figure 1). In addition, changes in soil respiration tightly related to the seasonal patterns of T_{soil} when compared to the annual dynamics of soil respiration and T_{soil} (Figure 2). The results indicated that T_{soil} explained a significant fraction of the variance in soil respiration process. The regression analyses confirmed the strong influence of T_{soil} on soil respiration (Figure 2). This finding was consistent with our previous study (Yan *et al*., 2014) as well as other previous reports (Mo *et al*., 2008; Deng *et al*., 2010; Zheng *et al*., 2009). Soil respiration was not related to W_{soil} content in the study site (data not showed). Also in our previous studies for different forest communities in the same site, we found that the correlations between soil respiration and W_{soil} were not significant ($P > 0.05$) (Allison *et al*., 2009; Tian *at al*., 2011; Yan *et al*., 2013).

CONCLUSIONS

In summary, forest soils are considered to have a considerable potential as C sinks. The hypothesis that N fertilization would cause reduced soil respiration in the Camphor tree and Slash pine forests was supported by the results of our studies. The mechanisms in reduction of soil respiration rates by N fertilizer application were complex and depended on forest types. For Camphor tree forests, the reduction of soil respiration was caused by decreasing microbial activity and reducing fine root biomass. For Slash pine forests, the decrease of soil respiration was mainly attributed to the decline of microbial activity. The hypothesis that various N additions would cause different effects on seasonal patterns of soil CO_2 efflux was not supported. The mechanisms dealing with no significant treatment effect among the N fertilization plots remained unclear, it appeared that all fertilized plots reached an 'adequate N content zone' where further addition of N in soils was no longer related to decreases in soil respiration. Although reduction of soil respiration were observed in N addition plots in both Camphor tree and Slash pine forests, the belowground response to N fertilization varied between the both species. Our results suggested that the increased anthropogenic N deposition resulting from either climate change or forest management practice might affect the belowground processes such as soil respiration and microbial activity in subtropical forests of China.

The alteration of belowground processes has a potential to affect C sequestration ability in this region, and this potential strongly depends on forest species.

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Table 1 Soil physical and chemical properties in Camphor tree and Slash pine forests in study site*

Forest types	Soil organic carbon （mg g^{-1}）	Total N （mg g^{-1}）	C/N	pH (1:2.5)	Soil bulk density （g cm^{-3}）
Camphor tree	13.65±3.26	1.31±0.27	10.42	3.98±0.22	1.48
Slash pine	15.67±4.76	1.46±0.41	10.73	4.01±0.17	1.51

*Value is mean±SE.

Table 2 Soil respiration rate (μmol m^{-2} s^{-1}) at different N addition treatments in Camphor tree and Slash pine forests in study site*

Forest type	Nitrogen addition treatment			
	Control	LN	MN	HN
Camphor tree	3.05±0.36a	1.99±0.25b	1.92±0.19b	2.00±0.22b
Slash pine	1.67±0.22a	1.21±0.16a	1.29±0.14a	1.17±0.11a

*Value is mean±SE. LN: low N addition treatment; MN: medium N addition treatment; HN: high N addition treatment. Different letters at the same row represent significant differences between the N addition treatments ($P < 0.05$).

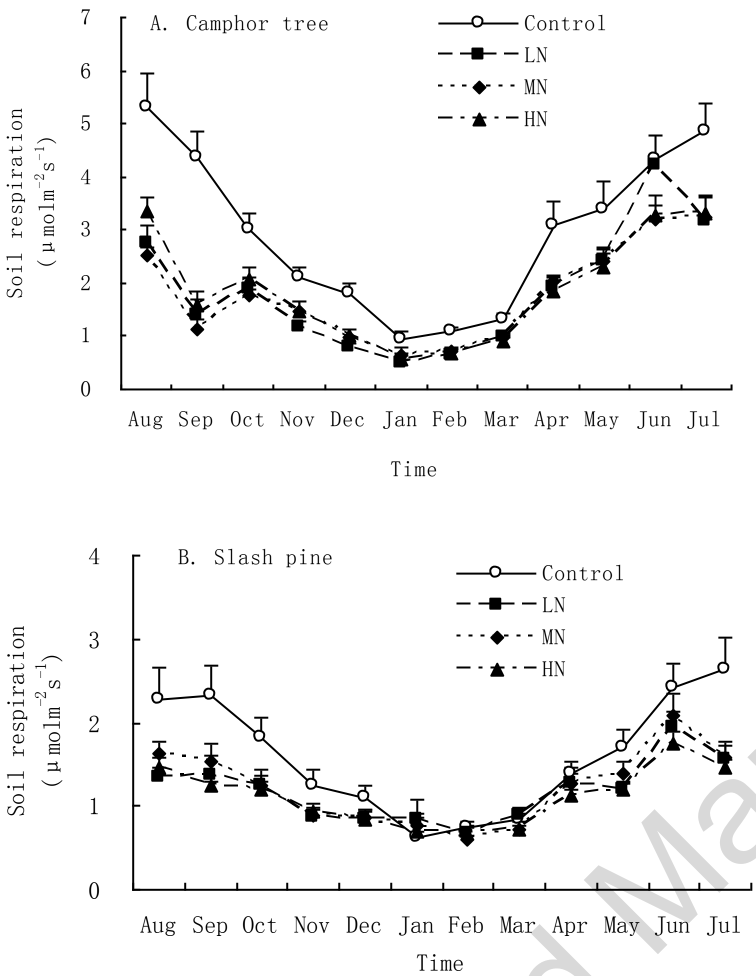
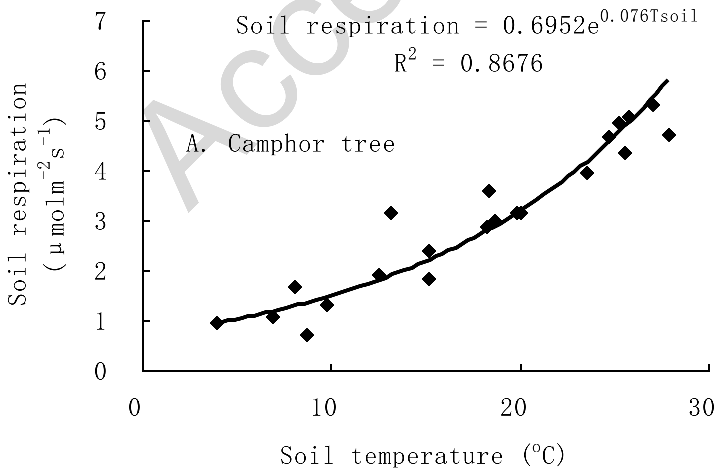


Figure 1. Seasonal patterns of soil respiration at different N addition treatments in Camphor tree forests (A) and Slash pine forests (B). Error bar indicates + 1SE. LN: low N addition treatment; MN: mediate N addition treatment; HN: high N addition treatment.



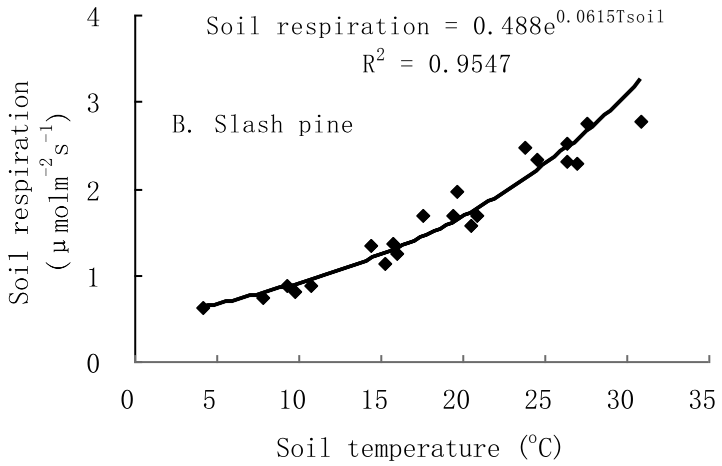


Figure 2. Relationship between soil temperature and soil respiration rates derived from control plots in Camphor tree forests (A) and Slash pine forests (B).

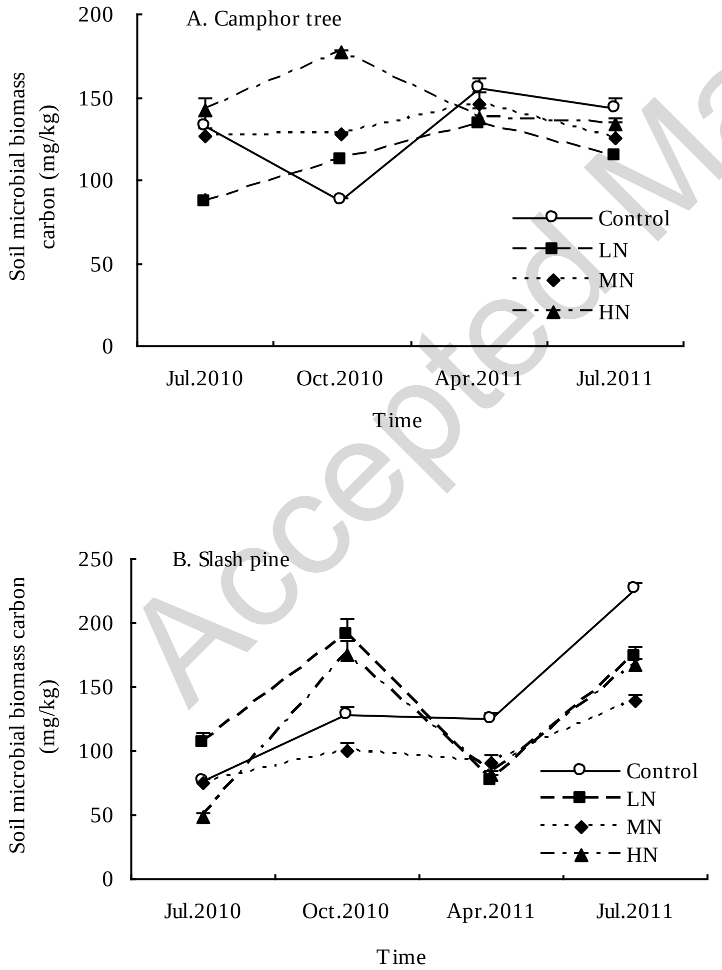
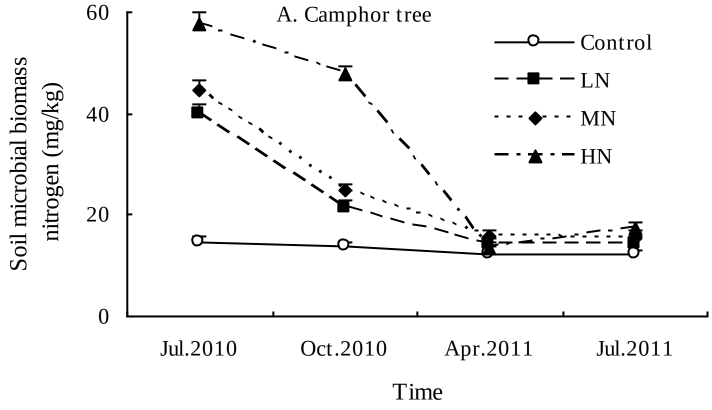


Figure 3. Seasonal patterns of soil microbial biomass carbon at different N addition treatments in Camphor tree forests (A) and Slash pine forests (B). LN: low N addition treatment; MN: mediate N

addition treatment; HN: high N addition treatment.



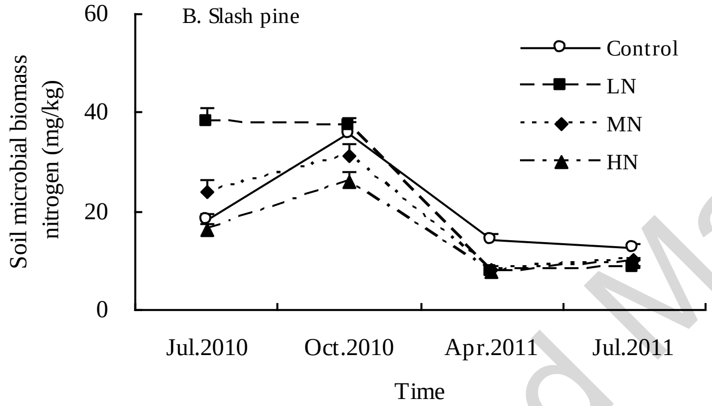
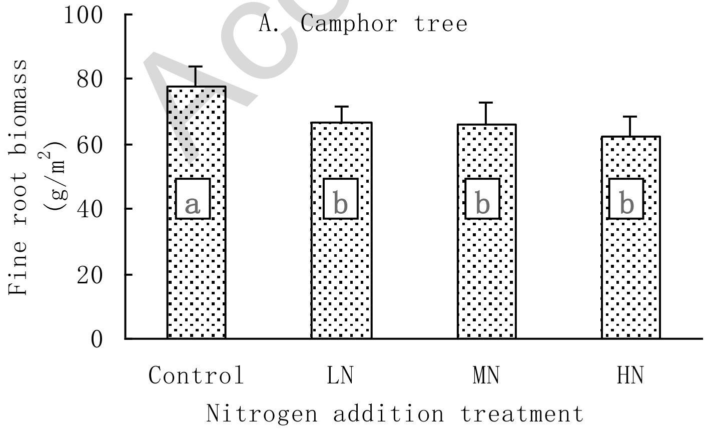


Figure 4. Seasonal patterns of soil microbial biomass nitrogen at different N addition treatments in Camphor tree forests (A) and Slash pine forests (B). LN: low N addition treatment; MN: mediate N addition treatment; HN: high N addition treatment.



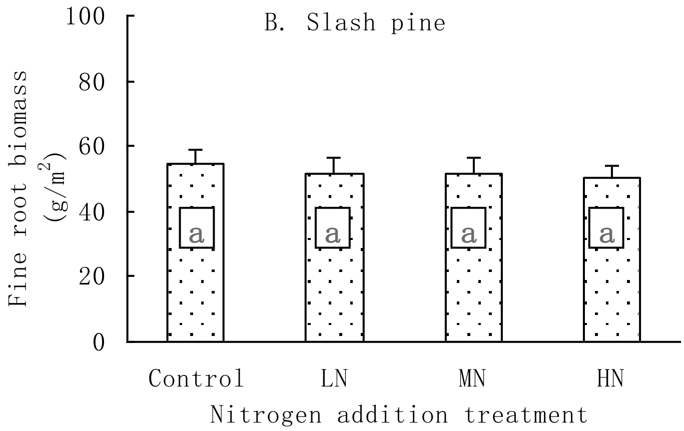


Figure 5. Fine root biomass at different N addition treatments in Camphor tree forests (A) and Slash pine forests (B). Different letters represent significant differences between the N addition treatments ($P < 0.05$). LN: low N addition treatment; MN: mediate N addition treatment; HN: high N addition treatment.

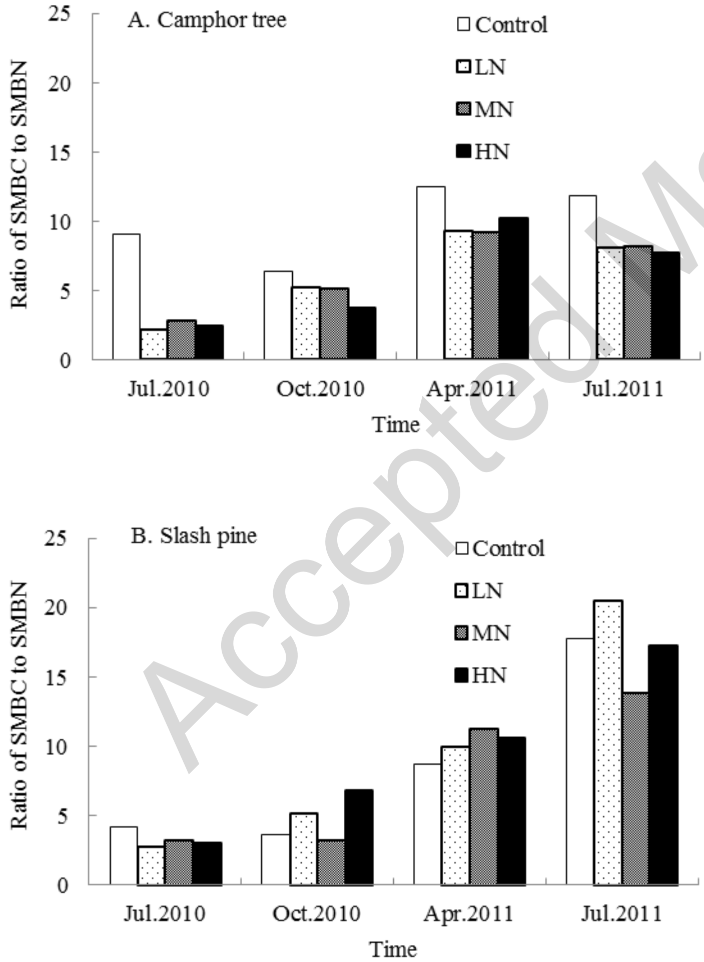


Figure 6. Seasonal patterns of the ratio of soil microbial biomass carbon (SMBC) to soil microbial biomass nitrogen (SMBN) at different N addition treatments in Camphor tree forests (A) and Slash pine forests (B). LN: low N addition treatment; MN: mediate N addition treatment; HN: high N addition treatment.