Biogeosciences Discuss., 12, 10359–10387, 2015
www.biogeosciences-discuss.net/12/10359/2015/
doi:10.5194/bgd-12-10359-2015

Biogeosciences
Discussions
Open Access

Responses of soil microbial communities and enzyme activities to nitrogen and phosphorus additions in Chinese fir plantations of subtropical China

W. Y. Dong[1], X. Y. Zhang[1], X. Y. Liu[2], X. L. Fu[1], F. S. Chen[3], H. M. Wang[1], X. M. Sun[1], and X. F. Wen[1]

[1]Key Laboratory of Ecosystem Network Observation and Modelling, Institute of Geographic Sciences and Natural Resources Research, Chinese Academy of Sciences, Beijing, 100101, China
[2]College of Geographic Science, Northeast Normal University, Changchun, 130024, China
[3]College of Life Sciences, Nanchang University, Nanchang, 330031, China

Received: 17 March 2015 – Accepted: 18 June 2015 – Published: 08 July 2015

Correspondence to: X. Y. Zhang (zhangxy@igsnrr.ac.cn)

Published by Copernicus Publications on behalf of the European Geosciences Union.

Abstract

Nitrogen (N) and phosphorus (P) additions to forest ecosystems are known to influence various above-ground properties, such as plant productivity and composition, and below-ground properties, such as soil nutrient cycling. However, our understanding of how soil microbial communities and their functions respond to nutrient additions in subtropical plantations is still not complete. In this study, we added N and P to Chinese fir plantations in subtropical China to examine how nutrient additions influenced soil microbial community composition and enzyme activities. The results showed that most soil microbial properties were responsive to N and/or P additions, but responses often varied depending on the nutrient added and the quantity added. For instance, there were more than 30 % greater increases in the activities of β-Glucosidase (βG) and N-acetyl-β-D-glucosaminidase (NAG) in the treatments that received nutrient additions compared to the control plot, whereas acid phosphatase (aP) activity was always higher (57 and 71 %, respectively) in the P treatment. N and P additions greatly enhanced the PLFA abundanceespecially in the N2P treatment, the bacterial PLFAs (bacPLFAs), fungal PLFAs (funPLFAs) and actinomycic PLFAs (actPLFAs) were about 2.5, 3 and 4 times higher, respectively, than in the CK. Soil enzyme activities were noticeably higher in November than in July, mainly due to seasonal differences in soil moisture content (SMC). βG or NAG activities were significantly and positively correlated with microbial PLFAs. There were also significant relationships between gram-positive (G^+) bacteria and all three soil enzymes. These findings indicate that G^+ bacteria is the most important microbial community in C, N, and P transformations in Chinese fir plantations, and that βG and NAG would be useful tools for assessing the biogeochemical transformation and metabolic activity of soil microbes. We recommend combined additions of N and P fertilizer to promote soil fertility and microbial activity in this kind of plantation.

1 Introduction

Nutrient availability, one of the most important factors controlling tree growth in forest plantations, can be significantly modified by fertilizer applications (Tumer and Lambert, 2008). Nitrogen (N) is generally believed to be the key growth-limiting element that controls the species composition, diversity, and productivity of forest ecosystems (Weand et al., 2010). N additions to forest ecosystems can influence a number of plant and soil processes, such as litter decomposition, carbon (C) storage and greenhouse gas fluxes (Cusack, 2013). In recent decades, N inputs into forest ecosystems from atmospheric deposition have increased at both regional and global scales, especially in Asia (Lu et al., 2009; Zechmeister-Boltenstern et al., 2011). This has raised the concern that forest ecosystems on nutrient poor soils may be at threat from imbalanced nutrition inputs (Vesterdal and Raulund-Rasmussen, 2002; Weand et al., 2010).

Phosphorus (P) is another primary limiting factor in many systems, especially in subtropical and tropical regions (Esberg et al., 2010). As a result, increased N deposition in these regions will cause a greater imbalance between N and P than in other regions. Exogenous P inputs to forests in these regions can lead to fast tree growth (Chen et al., 2010). However, to date it remains unclear how soil microbial properties respond to these nutrient additions, as N and P are rarely added simultaneously to forest ecosystems (Elser et al., 2007). An improved understanding of how nutrient additions influence soil microbial properties will be beneficial to support development of effective and sustainable management strategies for these forest ecosystems.

Just as different functional groups of microorganisms respond differently to prevailing environmental conditions, forest management practices will influence the composition of the soil microbial community in a specific way (Hackl et al., 2005; Chen et al., 2013). Phospholipid fatty acids (PLFAs) are widely accepted as biomarkers that help us to unravel the composition of soil microbial communities by indicating viable microbial biomass and providing a microbial community "fingerprint" (Zelles, 1999).

Studies have suggested that nutrient additions can significantly impact the population, composition, and function of soil microorganisms (Mandal et al., 2007; Hopkins et al., 2008; Geisseler and Scow, 2014), and that mineral fertilizer amendments can result in increases in soil microbial activity in subtropical forests (Cao et al., 2010; Geisseler and Scow, 2014). However, other studies have demonstrated that mineral fertilizers have either had no, or a negative effect on soil microbial diversity and activities (Moore-Kucera and Dick, 2008; Feng et al., 2009). Frey et al. (2004) found that active fungal biomass was lower in the fertilized plots compared to control plots in pine stands. The response of deciduous forests may be different from that of coniferous forests, and Nilsson et al. (2003) reported that the total soil fungal biomass may not be influenced by nutrient addition. In contrast, N additions led to a significant overall increase in fungal biomass in a northern hardwood forest ecosystem (Weand et al., 2010). In addition, some studies have found that nutrient addition have the opposite effects on soil bacteria in forest ecosystem (Demoling et al., 2008). Clearly, the response of the microbial community composition to nutrient additions appears to be substrate-specific in subtropical forests (Weand et al., 2010; Chang et al., 2011).

Soil microbial communities produce extracellular enzymes to acquire energy and resources from complex soil environments. These enzyme activities are also useful for detecting changes in soil quality, as they underpin nutrient cycling, and also serve as signals of altered microbial activity caused by environmental impacts (Li et al., 2009). Hydrolytic enzymes control the decomposition of many biological macromolecules that are abundant in plant litter and soil such as cellulose, hemicellulose, chitin, and protein (Allison et al., 2007). For our study we chose three enzymes that are related to the C cycle, β-Glucosidase (βG) mainly releases glucose from cellulose. N-acetyl-β-D-glucosaminidase (NAG) mainly releases N-acetyl-β-D-glucosamine residue from the terminal non-reducing ends of chitooligosaccharides and plays an important role in N cycling. Acid phosphatase (aP) mainly releases phosphate groups, and plays an essential role in P cycling (Stone et al., 2012). The production of such enzymes by microbes is closely related to the balance between the availability of and the demand for nutrients.

Mineral fertilizers have been reported to have positive, negative, and neutral effects on soil C-, N-, and P-acquiring enzyme activities (Wang et al., 2011; Stone et al., 2012; Qian et al., 2014). It has also been pointed out that the response of soil enzymes to nutrient additions is highly context-dependent and that it varies with environmental and management related factors (Geisseler and Scow, 2014). Therefore, further studies about the effects of different fertilizers across a range of soil types and environmental conditions are needed to provide an improved understanding of these complex interactions. In recent years, the influence of nutrient additions on soil microbial communities has been intensively studied (Weand et al., 2010; Cusack, 2013). However, most studies have been carried out in subtropical broad-leaved forests (Wu et al., 2011; Tu et al., 2013; Huang et al., 2014). Since coniferous forests are a specific type of subtropical forest (Lv et al., 2014), it is important to study how N and P additions influence nutrient cycling functions in soil microbial communities in subtropical coniferous forest.

Different seasons may have a strong influence on the life cycle of microbes in subtropical forests through changes in biotic and abiotic factors. In spring, the vegetation starts to produce shoots and leaves, followed by a photosynthetically active period in summer. The growth period ends when the litter falls in autumn, providing a wealth of material for the soil decomposer community. During winter, vegetation is generally inactive, and decomposition processes are also slow because of the decelerating effect of low temperatures on soil microbial metabolism (Thoms et al., 2013). There is also an almost complete turnover of the microbial community between winter and summer, with different functions occurring in both seasons (Bardgett et al., 2011). Soil microbial communities are likely to change as the soil temperature and moisture change (Moore-Kucera et al., 2008). July and November were two contrasting periods with hot and humid, and cold and dry conditions. The sharp contrast between the conditions in the two months suggests that the microbial communities may be different, and so findings from this study may reflect seasonal soil microbial diversity. Therefore, because we studied soils from two different months, we have obtained a limited insight into the influence of Chinese fir plantations on soil microorganisms in two seasons with very different climatic conditions.

Chinese fir (*Cunninghamia lanceolata*), an important native conifer, has been extensively planted in subtropical China. It covers 9.11 million hectares and accounts for more than 18 and 5 % of Chinese and global forest plantations, respectively (Huang et al., 2013). Over the past few years, Chinese fir plantations have received attention because of the decline in soil fertility and related yields; these declines are the result of successive planting, short rotation times, whole-tree harvesting, and poor site preparation (Yang et al., 2005). In order to improve soil quality and forest productivity, a number of management practices have been attempted, such as litter management, forest fertilization, and planting of broadleaved tree species (Zhang et al., 2004). Out of these measures, fertilization is the most effective and feasible. Many studies have reported findings about the effects of nutrient additions to Chinese fir plantations, but most of them were focused on the influence of nutrients on soil C, N sequestration, and nutrient cycling (Liao et al., 2014), and few studies have examined soil microbial properties and enzymes.

This study was conducted to determine the response of soil enzyme activities and microbial communities to N and P additions in different seasons in Chinese fir plantations, and to examine the linkages between soil properties, microbial community composition and soil enzyme activities. We hypothesized that soil hydrolytic enzyme activities and microbial biomass would increase under nutrient additions because of increased availability of resources from complex sources; we would also expect to find significant relationships between hydrolytic enzyme activities involved in C, N, and P transformations, soil C, N, and P contents, and the composition of the microbial communities.

Title Page
Abstract | Introduction
Conclusions | References
Tables | Figures
◀| |▶
◀ ▶
Back | Close
Full Screen / Esc
Printer-friendly Version
Interactive Discussion

2 Materials and methods

2.1 Site description

The study was conducted in the Qianyanzhou Forest Experimental Site, in Jiangxi Province, South China (26°44′52′′ N, 115°04′13′′ E, at an elevation of 102 m a.s.l.). The Chinese fir plantation was established in 2000. Average tree height and diameter at breast height were about 15 m and 13 cm, respectively. The site is characterized by a subtropical monsoon climate, with a mean annual temperature and precipitation of 17.9 °C and 1471.2 mm, respectively (Wen et al., 2010). The mean soil temperature and precipitation in July 2013 were 29.6 °C and 171.0 mm, respectively, while the mean soil temperature and precipitation in November 2013 were 14.0 °C and 118.6 mm, respectively (Fig. 1). The soil is classified as Ultisols using the USDA-NRCS soil taxonomy (1996). The soil bulk density was 1.31 g cm^{-3}, the pH value was 4.6, the soil organic carbon (SOC) content was 17.68 g kg^{-1}, total N content was 1.12 g kg^{-1}, and total P was 0.1 g kg^{-1}.

2.2 Experimental treatments

Thirty 20 m × 20 m plots, each with an area of 400 m^2 and a buffer zone of more than 10 m between the plots were established in November 2011. Six different treatments were used on five randomly distributed replicates as follows: control (CK), low N addition (N1: 50 kg ha^{-1} yr^{-1} of N), high N addition (N2: 100 kg ha^{-1} yr^{-1} of N), P addition (P: 50 kg ha^{-1} yr^{-1} of P), low N and P addition (N1 + P: 50 kg ha^{-1} yr^{-1} of N +50 kg ha^{-1} yr^{-1} of P) and high N and P addition (N2 + P: 100 kg ha^{-1} yr^{-1} of N +50 kg ha^{-1} yr^{-1} of P). N was added as NH_4NO_3 and P was added as NaH_2PO_4. The amount of N applied in the lower N treatment matched observed rates of N deposition in southern China (Lü et al., 2007), and the amount of P added was at a 1 : 1 ratio of the amount of the lower N application. The amount of N added for the higher N application was double the amount added for the lower application. Fertilizers were mixed with sand and were hand-scattered once every three months from March 2012 and until December 2013. Application varied according to the season, each application in the growing season accounted for 30 % of the total annual application, while each application in the non-growing season accounted for 20 % of the total annual application. Understory plants were removed manually at regular intervals and no herbicide was applied, so that potential impacts on soil organisms were avoided.

2.3 Soil sampling and analysis

Soils were sampled twice in 2013, at the end of July and November. Five soil cores (5 cm inner diameter) were collected randomly from each plot from the 0 to 10 cm soil layer, and were combined to form a composite sample. The litter layer was carefully removed before sampling. Soil pH was measured on a soil-water suspension (1 : 2.5 *v* : *v*) using a pH digital meter (Iovieno et al., 2010). Soil moisture content (SMC) was measured gravimetrically on 20 g fresh soil oven dried at 105 °C to constant weight (Liu et al., 2012). SOC and total N were measured with an elemental analyzer (Elementar, Vario Max, Germany). Total P was analyzed with a flow injection auto analyzer following digestion with H_2SO_4-$HClO_4$ digestion (Huang et al., 2011).

The soil microbial community was characterized by phospholipid fatty acids (PLFAs) analysis. PLFAs were extracted from the soil using the procedure of Bossio and Scow (1998). After mild alkaline methanolysis to form fatty acid methyl esters (FAMEs), samples were then dissolved in hexane and analyzed with a DB-5 column in a gas chromatography mass spectroscopy (GC-MS) system (Thermo TRACE GC Ultra ISQ). Total amounts of the different PLFA biomarkers were used to represent the different groups of soil micro-organisms. The following combinations of PLFA biomarkers were considered to represent the bacterial origin: (Gram-positive bacteria were represented by i15:0, a15:0, i16:0, i17:0, Gram-negative bacteria by 16:1ω7c, cy17:0, cy19:0, and total bacteria were represented by the sum of the two types) (Frostegård and Bååth, 1996). The PLFA 10Me18:0 and 10Me16:0 were used as a measure of actinomycic biomass. The PLFA 18:2ω6 and 18:1ω9c were used as markers for fungal biomass.

Taken together, the combination of bacterial, fungal and actinomycic PLFAs biomarkers was considered to represent the total PLFAs of the soil microbial community. The enzyme activities of βG, NAG and aP were determined using 96-well microplates following the methods of Saiya-Cork (2002). Assay plates were incubated in the dark at 20 °C for 4 h. Fluorescence was measured at an excitation wavelength of 365 nm and a 450 nm emission cutoff filter by a microplate fluorometer (SynergyH4 BioTek, USA).

2.4 Statistical analysis

One-way analysis of variance (ANOVA) and Duncan's multiple comparisons were performed to identify the differences between the fertilizer treatments because of N and P additions. The paired-sample t test was used to compare the seasonal variation in soil PLFAs and enzyme activities. Pearson correlations were used to determine the significance and strength of any relationships between soil properties, soil PLFAs, and enzyme activities. All statistical analyses were performed using SPSS version18.0 (Statistical Graphics Crop, Princeton, USA). The level of significance was $P < 0.05$.

3 Results

3.1 Soil properties

Comparison shows that, relative to the CK treatment, soil pH declined significantly after fertilizer applications (Table 1). The N2P treatment had the lowest soil pH (4.4 and 4.1 for both sampling times). Further comparison with the CK treatment shows that N and P fertilizer applications resulted in improvements in SOC, and total N and total P contents compared with the CK ($P < 0.05$).The average SOC, total N and total P contents in N1P were highest in July, and were approximately 26, 44 and 127 % higher than those of the CK treatment, respectively. In addition, SOC, total N, and total P concentrations in November were highest for the N2P treatment, and were 18, 35 and 60 % higher than those in the CK. However, compared with the CK, the P treatments had no significant influence on soil properties in either July or November ($P > 0.05$). Seasonally, the SMC was higher in November (25.6–27.9 %) than in July (18.1–21.4 %).

3.2 Soil hydrolytic enzyme activities involved in C, N and P transformations

βG enzyme activity was significantly influenced by fertilizer applications ($P < 0.05$), and the highest activities in both July and November were observed in the N2P treatment, both of which were about 93 % higher than those in the CK, respectively (Fig. 2). In addition, compared with the CK, βG activity was not influenced by P fertilizer applications ($P > 0.05$).

In July, NAG activity was significantly higher in fertilized plots than in the CK ($P < 0.05$), and was about 2 times greater in the N1 treatment, and 3 times greater in the N2 treatment, than in the CK. In November, NAG activity was significantly enhanced in the N1 and N2 treatments compared with the CK. However, applications of P fertilizer inhibited NAG activity, and NAG contents were 12 % lower in N1P than in N1, and 29 % lower in N2P than in N2, respectively. The NAG content was lowest in the P treatment. In contrast to NAG, aP activity was strongly influenced by the P treatment. Compared to the control, aP activities were always higher (57 and 71 %, respectively) in the P treatment. In particular, aP activity tended to be greater in the N1, N2P and P treatments in July, and in the N2 and P treatments in November (Fig. 2).

When the activities in the different sampling months are compared, the βG, NAG, and aP activities were significantly higher in November than in July ($P < 0.05$, Table S1).

3.3 Soil microbial community composition

Soil total PLFAs (totPLFAs) were significantly higher in the fertilized treatments than in the CK ($P < 0.05$). The totPLFAs were about 2.5 times greater in the N2P treatment than in the CK, and about 1.5 times higher in the N2 treatment than in the CK (Fig. 3). Bacterial PLFAs (BacPLFAs), Fungal PLFAs (funPLFAs) and Actinomycic PLFAs (act-

PLFAs) (Fig. 3) were influenced by the treatments in the same way as totPLFAs, that is, there were larger increases in the fertilized soils than in the CK ($P < 0.05$). BacPLFAs, funPLFAs and actPLFAs were highest in N2P, and were about 2.5, 3 and 4 times higher, respectively, than in the CK. G^+ PLFAs were higher than G^- PLFAs and both were significantly influenced by different treatments, and were greatest in the N2P treatments (Fig. 3).

The fungal/bacterial ratio was only significantly higher in the N2P treatments in July ($P < 0.05$, Fig. 4). The G^+/G^- ratio was not significantly influenced by fertilizer treatments ($P < 0.05$); values of this ratio were close to 2.5 (Fig. 4).

The seasonal patterns of total, bacterial, and fungal PLFAs for all soils were similar, and there were no significant differences between July and November ($P > 0.05$, Table S2). However, the F/B ratio was markedly higher in July than in November ($P < 0.05$, Table S2).

3.4 Relationships between soil enzyme activities, PLFAs profiles, and measured soil properties

Figure 5 shows the significance and strength of the relationships between microbial community composition, enzyme activities, and soil properties. Soil pH was significantly and positively correlated with aP activity, and negatively correlated with funPLFAs. The SMC was positively correlated with all soil enzyme activities and total, bacterial, G^+, and actinomycic PLFAs. Total N and total P were positively correlated with enzyme activities and soil PLFAs, while SOC was mainly responsible for the soil microbial community composition ($P < 0.05$).

Figure 6 shows the relationships between soil PLFAs and enzyme activities. βG and NAG activity were positively correlated with totPLFAs, bacPLFAs, actPLFAs, G^+ PLFAs, and G^+/G^-. AP activity was only positively correlated with G^+ PLFAs and G^+/G^-. However, there was no significant correlation between the funPLFAs and all soil enzyme activities.

4 Discussion

Numerous studies have reported decreases in soil pH after nutrient additions due to leaching of magnesium and calcium, as well as mobilization of aluminum (Wang et al., 2011). In line with these observations, we demonstrated that the soil pH decreased to a certain extent in the N and NP treatments, but not in the P treatment. This suggests that N deposition will lead to soil acidification in this region. The relationship between fertilization and soil carbon sequestration has been examined in previous studies (Khan et al., 2007; Wei et al., 2012). Khan et al. (2007) observed a net decline in soil C after 40 to 50 year of synthetic fertilization. Conversely, our study indicated that nutrient additions may have a positive influence on the amount of C stored in forests. These contrasting results may be attributed to the factor that, unlike agricultural systems, nutrient additions to forest ecosystems often lead to changes in the composition and diversity of plant species, which in turn have an influence on the forest litter. Consistent with our research, Wei et al. (2012) reported that nutrient additions led to a significantly enhancement of soil C sequestration and nutrient status in Chinese fir forest soils. Huang et al. (2011) also considered that soil nutrient enrichment, especially N, could reduce SOC decomposition. Moreover, nutrient-induced increases in forest litter and subsequent inputs of organic matter to the forest floor, and ultimately to the mineral soil, could lead to increases in soil nutrient concentrations (Moorhead and Sinsabaugh, 2006). The litter on the forest floor acts as input–output system of nutrient and the rates at which forest litter falls and subsequently, decomposes contribute to the maintenance of soil fertility in forest ecosystems (Wang et al., 2011). Zeng et al. (2015) found that while exogenous N and P additions could promote forest ecosystem biomass and could also lead to increases in the litter on the forest floor in the form of root exudates and aboveground residues, P addition had no influence on forest biomass. Therefore, total P did not change when only P was added, there were however significant increases in total P in response to combined applications of N and P. Besides, other related unpublished studies at our study site have demonstrated that, after P additions, P concentrations in leaves and twigs increased significantly. Soil P was largely absorbed by plants, and soil P remained unchanged.

Several previous studies have reported that nutrient additions can have both positive and negative influences on C-, N- and P- acquiring enzyme activities depending on tree species (Stursova et al., 2006; Piotrowska and Wilczewski, 2012). Consistent with our hypothesis, our study showed that βG and NAG activity levels were obviously higher after N and NP applications than the other treatments, which demonstrates that these enzymes were easily stimulated by substrates. This is the result of increased SOC and total N from the N and NP treatments, which were significantly and positively correlated with βG and NAG activities in our study (Fig. 6). Similar results were also reported by Mandal et al. (2007) and Liang et al. (2014), and they attributed the higher enzyme activity levels to higher organic matter contents and enhanced microbial activity. N additions to both labile and recalcitrant substrates are thought to allow microbes to invest N in enzyme production, which often results in increased activity of enzymes responsible for cellulose degradation (e.g., βG), for acquisition of organic N (NAG). Soil organic matter not only provides substrates for enzymes, but also plays a vital role in protecting soil enzymes by forming complexes with clay and humus (Saha et al., 2008).

The βG and NAG activities in the P fertilized plots were generally equal to or lower than those in the CK. Our results showed that higher total soil N could stimulate βG and NAG activity, but P additions had no influence on total soil N. Secondly, Turner and Wright (2014) found that P additions could lead to increases in soil microbial C and N which, in turn, would mean that microbes could reduce their investment in C- and N-acquiring enzymes such as βG and NAG. When a resource is limiting, microbes may benefit from producing enzymes to obtain it, but could be constrained by the availability of C and N required for enzyme synthesis. Similarly, aP was higher in fertilized treatments than in the control suggesting that fertilization improved soil microbial activity, which, in turn, produce enzymes to mobilize resources from complex sources (Keeler et al., 2009).

Our results clearly demonstrate that the two-season investigated microbial communities (July and November) differed in their functional response to nutrient additions. The November microbes demonstrated a higher capacity to degrade substrates (cellulose, plant cell walls) than the July communities, as indicated by the enhanced βG, NAG and aP activity. This was due to the higher SMC in November, which was significantly and positively correlated with soil enzyme activities in the present study (Table 3). Similar results have been observed previously for other tropical forest sites, in which they considered that low soil moisture would strongly limit soil enzyme activities (Liu et al., 2012; Steinweg et al., 2012; Schaeffer et al., 2013). Furthermore, McDaniel (2013) found that simulated warming decreased forest soil activity of both βG and NAG enzymes by 19 and 21 %, respectively. In our study, the annual temperature in July was close to 30 °C, which suggests that the soil enzyme activity was lower in July than November (Fig. 1).

A meta-analysis based on 107 datasets from 64 trials around the world showed that, compared to control unfertilized treatments, mineral fertilizer applications led to a 15.1 % increase in microbial biomass (Geisseler and Scow, 2014). Allen and Schlesinger (2004) suggested that increases in SOC and total N corresponded with increases in soil microbial biomass. Similarly in this study, we observed that, relative to CK, fertilizer applications enhanced bacterial, fungal, and actinomycic populations. Girvan et al. (2003) reported that soil properties could be a key control on the general composition of the microbial community. Studies have demonstrated that nutrient addition can increase forest productivity (Thomas et al., 2010). The higher productivity can lead to increased inputs of organic resources in the form of root exudates, decaying roots and aboveground residues, which would alleviate the C and N limitations for soil microbes (Keeler et al., 2009). The soil totPLFAs were highest in N2P and lowest in the P treatment, suggesting that the combined additions of N and P promoted synergistic positive effects on the soil microbial community.

Data from our experiment also showed that G^+ bacteria PLFAs tended to be more abundant in fertilized plots than G^- bacteria. Similar results were observed in other fertilized areas in spruce forests, and they suggested that G^+ bacteria were more adaptable to environmental conditions than G^- bacteria in the bulk soil (Demoling et al., 2008).

High values of the F/B biomass ratios are thought to indicate a more sustainable ecosystem with lower environmental impacts, in which organic matter decomposition and N mineralization are the main sources of soil nutrients for plant growth (Chen et al., 2013). In our study, nutrient addition to mineral soil led to significant increases in bacterial and fungal biomass. Similar results were found by Weand et al. (2010). He et al. (2008) suggested that fertilizer applications had less impact on soil bacterial community than fungi. Likewise, the higher F/B ratio in the N2P treatment was due to the degree of fungal increase was greater than that of bacteria under this treatment. Hackl et al. (2005) found that soil moisture was an important driver of overall microbial activity. Using multivariate analysis, Steinweg et al. (2012) reported that SMC was the most closely correlated with bacterial community structure. We also found that SMC was significantly and positively correlated with bacterial PLFA signatures, and the abundance of soil bacteria biomass was higher in November compared to July. This suggests that the significantly lower F/B ratio in November was attributable to the higher SMC.

The correlations between enzyme activities and soil PLFAs were not consistent for all the enzymes assayed. The activities of βG and NAG were correlated strongly with the totPLFAs, bacPLFAs, actPLFAs and G^+ PLFAs, but only aP was correlated with G^+. Therefore, soil βG and NAG activities are more useful for reflecting the metabolic activity of soil microbes in our study region than aP. There were no consistent correlation between fungal PLFAs and enzyme activities in this study. Šnajdr et al. (2008) obtained similar results, which they speculated to be due to the fungal biomass, of which the hyphal cords used for nutrients translocation were metabolically inactive. There was a significant relationship between G^+ and all three enzyme activities, which indicates that some of the changes to the soil hydrolases enzyme activities were regulated by the G^+ biomass.

5 Conclusions

N additions increased soil nutrient contents, with more pronounced effects with combined N and P applications. The average SOC, total N and total P contents in N1P were highest in July, and were approximately 26, 44 and 127 % higher than those of the CK treatment, respectively. Soil pH tended to decrease when nutrients were added, indicating that nutrient inputs, especially N deposition, were the main cause of soil acidification in this region.

The C (βG) and N (NAG) related hydrolase were more sensitive to N and NP additions than the P (aP) related hydrolase, and their contents were higher in the fertilizer applied plots compared to the CK. P additions stimulated the aP activity and inhabited βG and NAG activity. Compared to the control, aP activities were always higher (57 and 71 %, respectively) in the P treatment. The three enzyme activities were obviously higher in November than in July, and reflect the higher SMC in November.

The response of the soil microbial community composition was more significant for the combined N and P additions than for single additions of either N or P. Fertilizer applications resulted in increased bacterial, fungal, actinomycic, and total PLFAs in this study region, especially in the N2P treatment, the bacterial PLFAs (bacPLFAs), fungal PLFAs (funPLFAs) and actinomycic PLFAs (actPLFAs) were about 2.5, 3 and 4 times higher, respectively, than in the CK. However, there were no significant differences between the response for July and November.

The βG and NAG were strongly correlated with different soil PLFAs, and so would be useful tools for assessing the biogeochemical transformation and metabolic activity of soil microbes. G^+ bacteria were significantly related to all three hydrolytic enzyme activities, indicating that it may be the most important microbial community in C, N and P transformations. Since microbial activities are considered to be important components of soil biological activity, we would recommend simultaneous additions of N and P fertilizer to promote soil fertility in Chinese fir plantations.

The Supplement related to this article is available online at doi:10.5194/bgd-12-10359-2015-supplement.

Author contributions. H. M. Wang, F. S. Chen and X. L. Fu designed the measurement strategy for the experiment. The field measurements and soil analysis were carried out by X. Y. Liu, W. Y. Dong and X. Y. Zhang prepared the manuscript with contributions from all of the co-authors. The study was carried out under the supervision of X. M. Sun and X. F. Wen.

Acknowledgements. The authors sincerely acknowledge the financial support provided by the National Basic Research Program of China (973 Program, 2012CB416903) and the state key, major and general program of the National Natural Science Foundation of China (Nos. 31130009, 31290222, 41171153).

References

Allen, A. S. and Schlesinger, W. H.: Nutrient limitations to soil microbial biomass and activity in loblolly pine forests, Soil Biol. Biochem., 36, 581–589, 2004.

Allison, S. D., Hanson, C. A., and Treseder, K. K.: Nitrogen fertilization reduces diversity and alters community structure of active fungi in boreal ecosystems, Soil Biol. Biochem., 39, 1878–1887, 2007.

Bardgett, R. D., Bowman, W. D., Kaufmann, R., and Schmidt, S. K.: A temporal approach to linking aboveground and belowground ecology, Trends Ecol. Evol., 20, 634–641, 2005.

Bossio, D. A., Scow, K. M., Gunapala, N., and Graham, K. J.: Determinants of soil microbial communities: effects of agricultural management, season, and soil type on phospholipid fatty acid profiles, Microb. Ecol., 36, 1–12, 1998.

Cao, Y. S., Fu, S. L., Zou, X. M., Cao, H. L., Shao, Y. H., and Zhou, L. X.: Soil microbial community composition under Eucalyptus plantations of different age in subtropical China, Eur. J. Soil Biol., 46, 128–135, 2010.

Chang, E. H., Chen, C. T., Chen, T. H., and Chiu, C.Y: Soil microbial communities and activities in sand dunes of subtropical coastal forests, Appl. Soil Ecol., 49, 256–262, 2011.

Chen, F. L., Zheng, H., Zhang, K., Ouyang, Z. Y., Lan, J., and Li, H. L.: Changes in soil microbial community structure and metabolic activity following conversion from native Pinus massoniana plantations to exotic Eucalyptus plantations, Forest Ecol. Manag., 291, 65–72, 2013.

Chen, F. S., Fahey, T. J., Yu, M. Y., and Gan, L.: Key nitrogen cycling processes in pine plantations along a short urban–rural gradient in Nanchang, China, Forest Ecol. Manag., 259, 477–486, 2010.

Cusack, D. F.: Soil nitrogen levels are linked to decomposition enzyme activities along an urban-remote tropical forest gradient, Soil Biol. Biochem., 57, 192–203, 2013

Demoling, F., Ola Nilsson, L., and Bååth, E.: Bacterial and fungal response to nitrogen fertilization in three coniferous forest soils, Soil Biol. Biochem., 40, 370–379, 2008.

Elser, J. J., Bracken, M. E. S., Cleland, E. E., Gruner, D. S., Harpole, W. S., and Hillebrand, H.: Global analysis of nitrogen and phosphorus limitation of primary producers in freshwater, marine and terrestrial ecosystems, Ecol. Lett., 10, 1135–1142, 2007.

Esberg, C., du Toit, B., Olsson, R., Ilstedt, U., and Giesler, R.: Microbial responses to P addition in six South African forest soils, Plant Soil, 329, 209–225, 2010.

Feng, W. T., Zou, X. M., and Schaefer, D.: Above-and belowground carbon inputs affect seasonal variations of soil microbial biomass in a subtropical monsoon forest of southwest China, Soil Biol. Biochem., 41, 978–983, 2009.

Frey, S. D., Knorr, M., Parrent, J. L., and Simpson, R. T.: Chronic nitrogen enrichment affects the structure and function of the soil microbial community in temperate hardwood and pine forests, Forest Ecol. Manag., 196, 159–171, 2004.

Frostegård, Å., Tunlid, A., and Bååth, E.: Changes in microbial community structure during long-term incubation in two soils experimentally contaminated with metals, Soil Biol. Biochem., 28, 55–63, 1996.

Geisseler, D. and Scow, K. M.: Long-term effects of mineral fertilizers on soil microorganisms–A review, Soil Biol. Biochem., 75, 54–63, 2014.

Girvan, M. S., Bullimore, J., Pretty, J. N., Osborn, A. M., and Ball, A. S.: Soil type is the primary determinant of the composition of the total and active bacterial communities in arable soils, Appl. Environ. Microb., 69, 1800–1809, 2003.

Hackl, E., Pfeffer, M., Donat, C., Bachmann, G., and Zechmeister-Boltenstern, S.: Composition of the microbial communities in the mineral soil under different types of natural forest, Soil Biol. Biochem., 37, 661–671, 2005.

He, J. Z., Zheng, Y., Chen, C. R., He, Y. Q., and Zhang, L. M.: Microbial composition and diversity of an upland red soil under long-term fertilization treatments as revealed by culture-dependent and culture-independent approaches, J. Soil. Sediment., 8, 349–358, 2008.

Hopkins, D. W., Sparrow, A. D., Shillam, L. L., English, L. C., Dennis, P. G., and Novis, P.: Enzymatic activities and microbial communities in an Antarctic dry valley soil: responses to C and N supplementation, Soil Biol. Biochem., 40, 2130–2136, 2008.

Huang, X. M., Liu, S. R., Wang, H., Hu, Z. D., Li, Z. G., and You, Y. M.: Changes of soil microbial biomass carbon and community composition through mixing nitrogen-fixing species with Eucalyptus urophylla in subtropical China, Soil Biol. Biochem., 73, 42–48, 2014.

Huang, Z. Q., Clinton, P. W., Baisden, W. Troy, D., and Murray, R.: Long-term nitrogen additions increased surface soil carbon concentration in a forest plantation despite elevated decomposition, Soil Biol. Biochem., 43, 302–307, 2011.

Huang, Z. Q., He, Z. M., Wan, X. H., Hu, Z. H., Fan, S. H., and Yang, Y. S.: Harvest residue management effects on tree growth and ecosystem carbon in a Chinese fir plantation in subtropical China, Plant Soil, 364, 303–314, 2013.

Iovieno, P., Alfani, A., and Bååth, E.: Soil microbial community structure and biomass as affected by Pinus pinea plantation in two Mediterranean areas, Appl. Soil Ecol., 45, 56–63, 2010.

Keeler, B. L., Hobbie, S. E., and Kellogg, L. E.: Effects of long-term nitrogen addition on microbial enzyme activity in eight forested and grassland sites: implications for litter and soil organic matter decomposition, Ecosystems, 12, 1–15, 2009.

Khan, S. A., Mulvaney, R. L., Ellsworth, T. R., and Boast, C. W.: The myth of nitrogen fertilization for soil carbon sequestration, J. Environ. Qual., 36, 1821–1832, 2007.

Li, Y. T., Rouland, C., Benedetti, M., Li, F. B., Pando, A., and Lavelle, P.: Microbial biomass, enzyme and mineralization activity in relation to soil organic C, N and P turnover influenced by acid metal stress, Soil Biol. Biochem., 41, 969–977, 2009.

Liang, Q., Chen, H. Q., Gong, Y. S., Yang, H. F., Fan, M. S., and Kuzyakov, Y.: Effects of 15 years of manure and mineral fertilizers on enzyme activities in particle-size fractions in a North China Plain soil, Eur. J. Soil Biol., 60, 112–119, 2014.

Liao, Y. C., McCormack, M. L., Fan, H. B., Wang, H. M., Wu, J. P., and Tu, J.: Relation of fine root distribution to soil C in a Cunninghamia lanceolata plantation in subtropical China, Plant Soil, 381, 225–234, 2014.

Liu, L., Gundersen, P., Zhang, T., and Mo, J. M.: Effects of phosphorus addition on soil microbial biomass and community composition in three forest types in tropical China, Soil Biol. Biochem., 44, 31–38, 2012.

Lü, C. and Tian, H.: Spatial and temporal patterns of nitrogen deposition in China: synthesis of observational data, J. Geophys. Res., 112, 10–15, 2007.

Lu, X. K., Mo, J. M., Gundersern, P., Zhu, W. X., Zhou, G. Y., and Li, D. J.: Effect of simulated N deposition on soil exchangeable cations in three forest types of subtropical China, Pedosphere, 19, 189–198, 2009.

Lv, Y. N., Wang, C. Y., Jia, Y. Y., Wang, W. W., Ma, X., and Du, J. J.: Effects of sulfuric, nitric, and mixed acid rain on litter decomposition, soil microbial biomass, and enzyme activities in subtropical forests of China, Appl. Soil Ecol., 79, 1–9, 2014.

Mandal, A., Patra, A.K, Singh, D., Swarup, A., and Ebhin Masto, R.: Effect of long-term application of manure and fertilizer on biological and biochemical activities in soil during crop development stages, Bioresource Technol., 98, 3585–3592, 2007.

McDaniel, M. D., Kaye, J. P., and Kaye, M.W: Increased temperature and precipitation had limited effects on soil extracellular enzyme activities in a post-harvest forest, Soil Biol. Biochem., 56, 90–98, 2013.

Moore-Kucera, J. and Dick: R. P. PLFA profiling of microbial community structure and seasonal shifts in soils of a Douglas-fir chronosequence, Microb. Ecol., 55, 500–511, 2008.

Moorhead, D. L. and Sinsabaugh, R. L.: A theoretical model of litter decay and microbial interaction, Ecol. Monogr., 76, 151–174, 2006.

Nilsson, L. O. and Wallander, H.: Production of external mycelium by ectomycorrhizal fungi in a Norway spruce forest was reduced in response to nitrogen fertilization, New Phytol., 158, 409–416, 2003.

Piotrowska, A. and Wilczewski, E.: Effects of catch crops cultivated for green manure and mineral nitrogen fertilization on soil enzyme activities and chemical properties, Geoderma, 189, 72–80, 2012.

Qian, X., Gu, J., Sun, W., Li, Y. D., Fu, Q. X., and Wang, X. J.: Changes in the soil nutrient levels, enzyme activities, microbial community function, and structure during apple orchard maturation, Appl. Soil Ecol., 77, 18–25, 2014.

Saha, S., Prakash, V., Kundu, S., Kumar, N., and Mina, B. L.: Soil enzymatic activity as affected by long term application of farm yard manure and mineral fertilizer under a rainfed soybean–wheat system in NW Himalaya, Eur. J. Soil Biol., 44, 309–315, 2008.

Saiya-Cork, K. R., Sinsabaugh, R. L., and Zak, D. R.: The effects of long term nitrogen deposition on extracellular enzyme activity in an Acer saccharum forest soil, Soil Biol. Biochem., 34, 1309–1315, 2002.

Schaeffer, S. M., Sharp, E., Schimel, J. P., and Welker, J. M.: Soil-plant N processes in a High Arctic ecosystem, NW Greenland are altered by long-term experimental warming and higher rainfall, Glob. Change Biol., 19, 3529–3539, 2013. Šnajdr, J., Valášková, V., Merhautová, V., Herinková, J., Cajthaml, T., and Baldrian, P.: Spatial variability of enzyme activities and microbial biomass in the upper layers of Quercus petraea forest soil, Soil Biol. Biochem., 40, 2068–2075, 2008.

Soil Survey Staff: National Soil Survey Handbook, USDA/NRCS, US Government Printing Office, Washington, DC, 1996.

Steinweg, J. M., Dukes, J. S., and Wallenstein, M. D.: Modeling the effects of temperature and moisture on soil enzyme activity: linking laboratory assays to continuous field data, Soil Biol. Biochem., 55, 85–92, 2012.

Stone, M. M., Weiss, M. S., Goodale, C. L., Adams, M. B., Fernandez, I. J., and German, D. P.: Temperature sensitivity of soil enzyme kinetics under N fertilization in two temperate forests, Glob. Change Biol., 18, 1173–1184, 2012.

Stursova, M., Crenshaw, C. L., and Sinsabaugh, R. L.: Microbial responses to long-term N deposition in a semiarid grassland, Microb. Ecol., 51, 90–98, 2006.

Thomas, R. Q., Canham, C. D., Weathers, K. C., and Goodale, C. L.: Increased tree carbon storage in response to nitrogen deposition in the US, Nat. Geosci., 3, 13–17, 2010.

Thoms, C. and Gleixner, G.: Seasonal differences in tree species' influence on soil microbial communities, Soil Biol. Biochem., 66, 239–248, 2013.

Tu, L. H., Hu, T. X., Zhang, J., Li, X. W., Hu, H. L., and Liu, L.: Nitrogen addition stimulates different components of soil respiration in a subtropical bamboo ecosystem, Soil Biol. Biochem., 58, 255–264, 2013.

Tumer, J. and Lambert, M. J.: Nutrient cycling in age sequences of two Eucalyptus plantation species, Forest Ecol. Manag., 255, 1701–1712, 2008.

Turner, B. L. and Wright, S. J.: The response of microbial biomass and hydrolytic enzymes to a decade of nitrogen, phosphorus, and potassium addition in a lowland tropical rain forest, Biogeochemistry, 117, 115–130, 2014.

Vesterdal, L. and Raulund-Rasmussen, K.: Availability of nitrogen and phosphorus in Norway spruce forest floors fertilized with nitrogen and other essential nutrients, Soil Biol. Biochem., 34, 1243–1251, 2002.

Wang, C. Y., Han, G. M., Jia, Y., Feng, X. G., Guo, P., and Tian, X. J.: Response of litter decomposition and related soil enzyme activities to different forms of nitrogen fertilization in a subtropical forest, Ecol. Res., 26, 505–513, 2011.

Weand, M. P., Arthur, M. A., Lovett, G. M., McCulley, R. L., and Weathers, K. C.: Effects of tree species and N additions on forest floor microbial communities and extracellular enzyme activities, Soil Biol. Biochem., 42, 2161–2173, 2010.

Wei, X., Blanco, J. A., Jiang, H., and Kimmins, J. H.: Effects of nitrogen deposition on carbon sequestration in Chinese fir forest ecosystems, Sci. Total Environ., 416, 351–361, 2012.

Wen, X. F., Wang, H.-M., Wang, J.-L., Yu, G.-R., and Sun, X.-M.: Ecosystem carbon exchanges of a subtropical evergreen coniferous plantation subjected to seasonal drought, 2003–2007, Biogeosciences, 7, 357–369, doi:10.5194/bg-7-357-2010, 2010.

Wu, J. P., Liu, Z. F., Wang, X. L., Sun, Y. X., Zhou, L. X., and Lin, Y. B.: Effects of understory removal and tree girdling on soil microbial community composition and litter decomposition in two Eucalyptus plantations in South China, Func. Ecol., 25, 921–931, 2011.

Yang, Y. S., Guo, J. F., Chen, G. S., Xie, J. S., Gao, R and Li, Z.: Carbon and nitrogen pools in Chinese fir and evergreen broadleaved forests and changes associated with felling and burning in mid-subtropical China, Forest Ecol. Manag., 216, 216–226, 2005.

Zechmeister-Boltenstern, S., Michel, K., and Pfeffer, M.: Soil microbial community structure in European forests in relation to forest type and atmospheric nitrogen deposition, Plant Soil, 343, 37–50, 2011.

Zelles, L.: Fatty acid patterns of phospholipids and lipopolysaccharides in the characterisation of microbial communities in soil: a review, Biol. Fert. Soils, 29, 111–129, 1999.

Zeng, W. and Wang, W.: Combination of nitrogen and phosphorus fertilization enhance ecosystem carbon sequestration in a nitrogen-limited temperate plantation of Northern China, Forest Ecol. Manag., 341, 59–66, 2015.

Zhang, X. Q. Kirschbaum, M. U. F., Hou, Z. H., and Guo, Z. H.: Carbon stock changes in successive rotations of Chinese fir Cunninghamia lanceolata(lamb) hook plantations, Forest Ecol. Manag., 202, 131–147, 2004.

Table 1. Response of soil properties to N and P additions to Chinese fir plantations in July and November (means ± standard error).

	Treatment	pH	SMC (%)	Total N (g kg^{-1})	SOC (g kg^{-1})	Total P (g kg^{-1})
Jul	CK	4.6 (0.06) a	18.1 (1.5) ns	0.9 (0.03) b	21.6 (0.75) b	0.11 (0.00) c
	N1	4.2 (0.06) b	18.7 (2.4) ns	0.9 (0.01) b	24.3 (0.15) ab	0.12 (0.01) c
	N2	4.2 (0.13) b	20.8 (2.3) ns	1.1 (0.06) a	25.8 (1.20) a	0.16 (0.01) abc
	N1P	4.2 (0.05) ab	21.4 (2.1) ns	1.3 (0.11) a	27.2 (0.70) a	0.25 (0.03) a
	N2P	4.1 (0.06) b	19.9 (1.7) ns	1.3 (0.07) a	26.7 (1.28) a	0.18 (0.01) ab
	P	4.4 (0.07) a	20.4 (1.4) ns	0.9 (0.02) b	22.1 (0.95) b	0.16 (0.03) bc
Nov	CK	4.8 (0.11) a	25.0 (0.92) ns	1.1 (0.04) b	22.9 (0.51) b	0.15 (0.01) b
	N1	4.4 (0.05) b	27.9 (0.82) ns	1.3 (0.07) b	23.5 (0.63) ab	0.16 (0.01) ab
	N2	4.4 (0.16) b	25.6 (0.67) ns	1.6 (0.02) a	25.8 (1.47) a	0.18 (0.01) ab
	N1P	4.6 (0.04) ab	25.9 (1.16) ns	1.6 (0.06) a	24.5 (1.35) a	0.22 (0.01) ab
	N2P	4.4 (0.06) b	30.2 (1.25) ns	1.7 (0.07) a	27.0 (2.61) a	0.24 (0.02) a
	P	4.8 (0.07) a	26.1 (1.07) ns	1.6 (0.06) a	23.3 (0.58) b	0.18 (0.01) ab

Note: Numbers in brackets represent the standard error of the means. Different lower-case letters in the same column indicate significant differences when $P < 0.05$; ns: no significant difference between treatments.

Title Page
Abstract Introduction
Conclusions References
Tables Figures
◀◀ ▶▶
◀ ▶
Back Close
Full Screen / Esc
Printer-friendly Version
Interactive Discussion

Title Page
Abstract | Introduction
Conclusions | References
Tables | Figures
◂◂ | ▸▸
◂ | ▸
Back | Close
Full Screen / Esc
Printer-friendly Version
Interactive Discussion

Table 2. Pearson correlations between soil properties, soil enzyme activities and microbial variables.

	pH	SMC	Total N	SOC	Total P
βG	0.31 ns	0.82^{b}	0.72^{b}	0.16 ns	0.37^{b}
NAG	0.24 ns	0.71^{b}	0.71^{b}	0.12 ns	0.36^{a}
aP	0.59^{b}	0.73^{b}	0.71^{b}	0.05 ns	0.30 ns
Tot PLFAs	−0.24 ns	0.39^{b}	0.67^{b}	0.65^{b}	0.60^{b}
BacPLFAs	−0.17 ns	0.49^{b}	0.71^{b}	0.62^{b}	0.61^{b}
FunPLFAs	−0.44^{b}	−0.17 ns	0.18 ns	0.49^{b}	0.27 ns
ActPLFAs	−0.07 ns	0.50^{b}	0.67^{b}	0.57^{b}	0.55^{b}
G+PLFAs	−0.10 ns	0.59^{b}	0.73^{b}	0.55^{b}	0.60^{b}
hG-PLFAs	−0.34 ns	0.14 ns	0.53^{b}	0.68^{b}	0.52^{b}
F/B	−0.36 ns	−0.47^{b}	−0.27 ns	−0.10 ns	−0.12 ns
G^{+}/G^{-}	0.20 ns	0.59^{b}	0.34^{b}	0.10 ns	0.15 ns

Note: The values are correlation coefficients. ^{a} $P < 0.05$, ^{b} $P < 0.01$; ns: no significant differences. SMC: soil moisture content, SOC: soil organic carbon, βG: β-Glucosidase, NAG: N-acetyl-β-glucosaminidase, aP: acid phosphatase; Tot PLFAs: Total PLFAs, BacPLFAs: Bacterial PLFAs, ActPLFAs: Actinomycete PLFAS, G^{+} PLAs: Positive gram bacterial PLFAs, G^{-} PLFAs: Negative gram bacterial PLFAs, the same below.

Table 3. Pearson correlations between soil enzyme activities and microbial PLFAs.

	TotPLFAs	BacPLFAs	FunPLFAs	ActPLFAs	G^+PLFAs	G^-PLFAs	F/B	G^+/G^-
βG	0.39^a	0.51^b	−0.22 ns	0.49^b	0.59^b	0.17 ns	-0.59^b	0.57^b
NAG	0.35^a	0.46^b	−0.19 ns	0.43^b	0.56^b	0.09 ns	-0.53^b	0.62^b
aP	0.23 ns	0.33 ns	−0.26 ns	0.37 ns	0.42^b	0.07 ns	−0.53 ns	0.54^b

Note: The values are the correlation coefficients. [a] $P < 0.05$, [b] $P < 0.01$. ns: no significant differences.

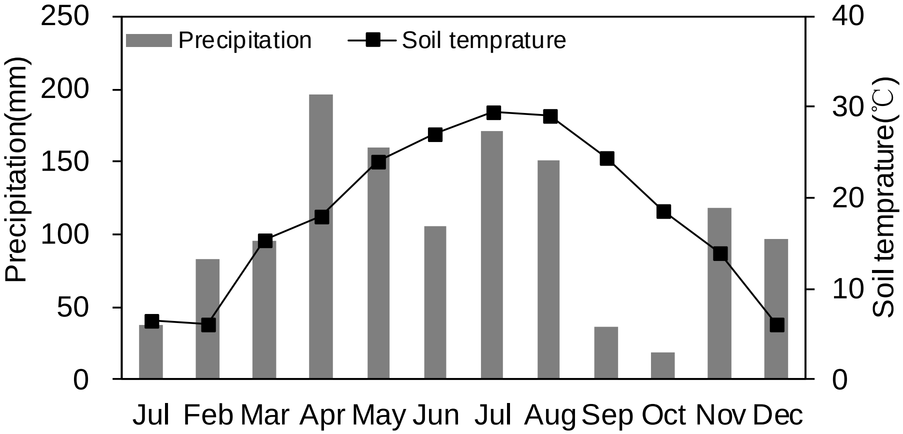

Figure 1. Mean monthly soil temperature and precipitation in the study area during 2013.

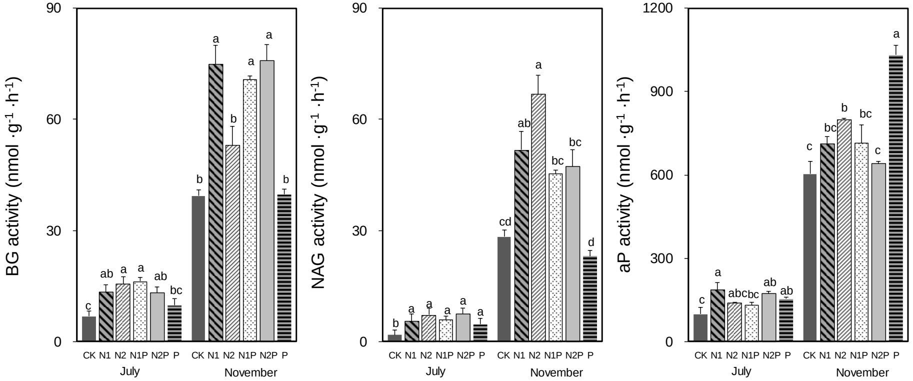

Figure 2. Response of soil enzyme activities to N and P additions in Chinese fir plantations in July and November.

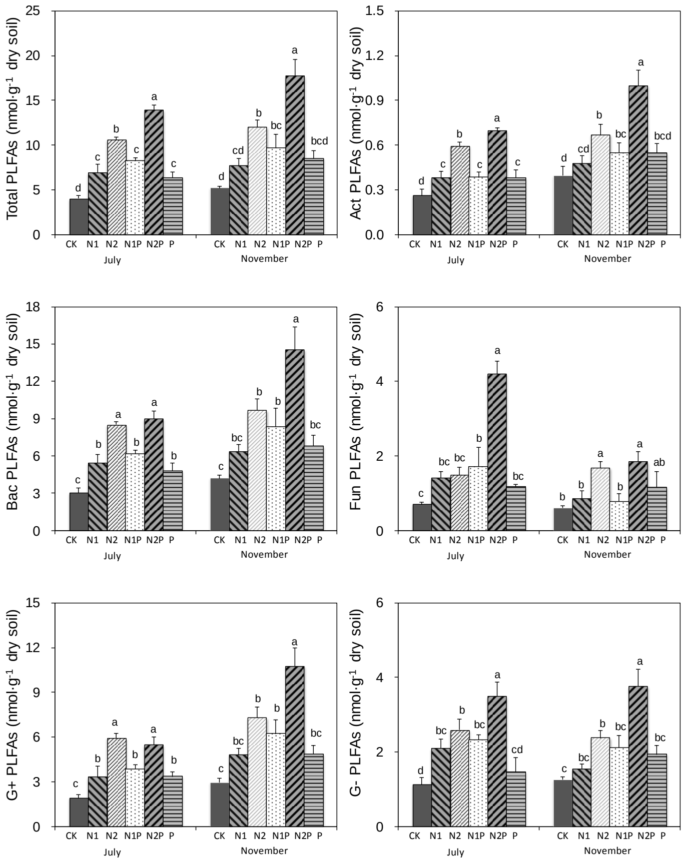

Figure 3. Responses of soil microbial PLFAs to N and P additions to Chinese fir plantations (Tot PLFAs: Total PLFAs, BacPLFAs: Bacterial PLFAs, ActPLFAs: Actinomycete PLFAS, G+ PLAs: Positive gram bacterial PLFAs, G- PLFAs: Negative gram bacterial PLFAs, the same below).

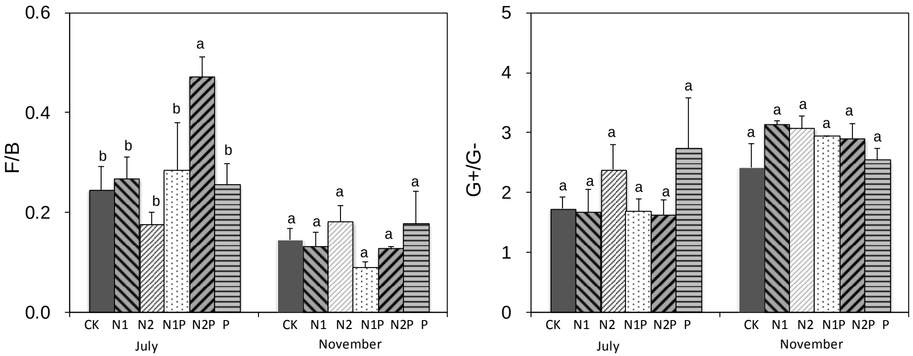

Figure 4. Ratio of F/B and G^+/G^- to N and P additions to Chinese fir plantations.