Stable soil organic carbon is positively linked to microbial-derived compounds in four plantations of subtropical China

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Stable SOC is positively linked to microbial-derived compounds

H. Wang et al.
Abstract

Indigenous broadleaf plantations are increasingly being developed to substitute pure coniferous plantations to increase biodiversity and soil fertility in subtropical China. To assess how plantation types affect soil organic carbon (SOC) chemical composition, we used the solid-state $^{13}$C nuclear magnetic resonance spectroscopy with cross-polarization and magic-angle spinning (CPMAS-NMR) technique to analyze SOC and litter C chemical compositions in a coniferous (Pinus massoniana) and three broadleaf (Castanopsis hystrix, Michelia macclurei and Mytilaria laosensis) plantations in subtropical China. Soil microbial community composition and biomass were investigated with the phospholipid fatty acids (PLFAs) and chloroform fumigation-extraction methods, respectively. The SOC chemical composition varied with plantation type, with 34% of the SOC found in the alkyl C fraction in the P. massoniana plantation compared to <28% in the broadleaf plantations. The amount of total PLFAs, bacterial and particularly the gram-positive bacterial population size, and microbial C/N ratio were correlated with the alkyl C content and alkyl C/O-alkyl C ratio. However, the soil alkyl C content was not correlated with the recalcitrance of leaf litter or fine roots. We thus suggest that the stable SOC composition could be attributable to the contribution of microbial-derived C compounds, rather than leaf litter or fine root quality or a direct flux of C from recalcitrant litter materials to more stable SOC pools.

1 Introduction

Soil organic matter (SOM) is a complex mixture of carbon (C)-rich organic molecules (Solomon et al., 2007; Crow et al., 2009), including labile C forms such as carbohydrates and polysaccharides (O-alkyl C), and recalcitrant C forms such as aliphatic (alkyl C) and lignin (aromatic C) compounds that are resistant to decomposition due to their intrinsic molecular properties (Lorenz et al., 2007; Shrestha et al., 2008). Recent evidence shows that aliphatic compounds often accumulate in the soil, thus contribut-
ing to the increased stable soil organic C (SOC) pools (Nierop, 1998; Lorenz et al., 2007).

The SOC chemical composition in forest soils is affected by the type of vegetation; for example, carbonyl C dominated soils under oak (*Quercus*), O-alkyl C prevailed in soils under manzanita (*Arctostaphylos*), and alkyl C was prominent in soils under coniferous vegetation (Quideau et al., 2001). Conversion from natural to hoop pine forests decreased O-alkyl C and increased alkyl C contents (Chen et al., 2004). Aromatic C content was higher and carbonyl C content was lower in the forest floor of spruce (*Picea glauca* (Moench) Voss) than in aspen (*Populus tremuloides* Michx.) stands in boreal mixed wood forests of Alberta, Canada (Hannam et al., 2004).

Plant litter and soil microbial biomass are two of the major sources of SOM (Kögel-Knabner, 2002), some of which are precursors of stable SOC fractions, formed through leaching, fragmentation and chemical alteration (Chapin et al., 2002). The chemical composition of above- and belowground plant tissues can influence litter decomposability and the association of decomposition products with soil minerals (Kögel-Knabner, 2002). Crow et al. (2009) showed that root-derived aliphatic compounds were a source of SOC with greater relative stability than leaf-derived C in soils of deciduous forests, whereas root-derived lignin and needle-derived aliphatic compounds were preferentially preserved in soils of coniferous forests, indicating that the dominant source of SOC can differ substantially between forest types.

Although most soil C is ultimately derived from plant material (Kögel-Knabner, 2002), a large proportion of it is transformed from microbial biomass into SOM (Simpson et al., 2007; Liang et al., 2011; Miltner et al., 2012). Soil microbial biomass represents a significant source of SOC and biochemical precursors that contribute to the maintenance of SOM (Kindler et al., 2006; Fu, 2007; Simpson et al., 2007). The turnover of microbial non-living biomass was estimated to contribute as much as 80% to the accumulation of SOM (Liang and Balser, 2010). The cell wall compounds, metabolites and C use efficiencies differed among microbial communities (Six et al., 2006). About 50% of the bacterial biomass-derived C remained in the soil, mainly as the non-living component.
of SOM in a 224 day soil incubation experiment studied with $^{13}$C-labelled bacterial cells (Miltner et al., 2012). Fungal biomarkers indicate impaired degradation and preservation of fungal residues in late successional forests, and the dynamics of root-associated fungi is an important regulator of ecosystem C accumulation in boreal forests (Clemmensen et al., 2013). Soil microbial communities can be greatly affected by afforestation and reforestation as such activities change the litter type and the environment under canopy (Hackl et al., 2005; Liu et al., 2012). In particular, the SOC chemical composition can be influenced by the vegetation through the different organic compounds they produce and through their interaction with microbial communities (Kögel-Knabner, 2002; Lorenz et al., 2007).

Plantations are being established at an increasing rate throughout much of the world, which now account for 5% of the global forest cover (FAO, 2001), primarily for the production of wood fiber. There is also growing recognition of the conservation value of plantations in reducing logging pressure on natural forests, in sequestering C and in restoring degraded lands (Kelty, 2006). In China, the total plantation area reached $6.2 \times 10^7$ ha, accounting for 31.8% of the total forest area of the country, ranked first in the world in terms of the total plantation area (SFA, 2010). With the abundance of solar radiation and water resources, southern China makes up 63% of the plantation areas in the country (SFA, 2007). However, most of these plantations were planted with single coniferous tree species (e.g., *Pinus massoniana* or *Cunninghamia lanceolata*) or exotic tree species such as *Eucalyptus* (SFA, 2007), leading to reductions in biodiversity, ecosystem stability and soil fertility (Peng et al., 2008). Plantations of indigenous broadleaf species with high economic value can supply good quality timber while enhance biodiversity and ecosystem services (Carnevalea and Montagnini, 2002; Liang, 2007). They are increasingly developed as an alternative to replace coniferous plantations in many countries (Borken and Beese, 2006; Vesterdal et al., 2008). Few studies, however, have examined relationships among litter C quality, soil microbial community composition and SOC chemical properties in plantation types of different tree species.
In our previous study, we found that soils in the broadleaf plantations contained more decomposable C fractions, as indicated by the lower percentage of alkyl C, higher percentage of O-alkyl C and lower alkyl C/O-alkyl C ratio as compared to those in the pine plantation (Wang et al., 2010a), whereas the controlling factors driving the variations in SOC compositions among the plantations were not clearly evaluated. In this paper we report the differences in litter C chemical composition, soil microbial biomass and microbial community composition among those same four subtropical plantation types. The purpose of this study was to explore the key factors affecting SOC chemical composition in different plantation types. Our hypotheses are that: (i) litter C chemical composition, soil microbial biomass and microbial community composition differ among the four subtropical plantations of different species; (ii) soil organic carbon composition is linked to litter-derived and/or microbial-derived compounds in the four subtropical plantations of different species and (iii) such linkages between bacterial or fungal biomass and stable SOC composition would be different among the four subtropical plantations of different species.

2 Materials and methods

2.1 Site description

The study area was located at the Experimental Center for Tropical Forestry, Chinese Academy of Forestry (22°10’ N, 106°50’ E), located in Pingxiang City, Guangxi Zhuang Autonomous Region, China. The study area was located in the subtropical region. The annual rainfall was 1202.9 mm from May 2006 to April 2007, falling primarily from May to August. The mean annual temperature was 22.5 °C from May 2006 to April 2007 (Lu et al., 2009). The sandy textured soil at the study site was formed from a granitic parent geological material and is classified as a Red soil in the Chinese system of soil classification, equivalent to an Oxisol in the USDA Soil Taxonomy (Liang and Wen, 1992; State Soil Survey Service of China, 1998; Soil Survey Staff of USDA, 2006). Originally,
the study site was a subtropical evergreen broadleaf forest, and a *C. lanceolata* plantation was established in the 1950s after a clearcut of natural forests. Four plantations were randomly established in 1983 as mono-specific plantations after a clearcut of the *C. lanceolata* plantation.

The four plantations were located at an elevation of 550 m and were selected based on their similar topography, soil texture, stand age, and management history. The four plantations include a coniferous plantation (*P. massoniana*) and three broadleaf plantations (*Castanopsis hystrix*, *Michelia macclurei* and *Mytilaria laosensis*). These four tree species are the main indigenous but not N fixing species for afforestation and reforestation in the study area. In each plantation, four plots (each 20 m × 20 m) were randomly delineated for sampling. The stand characteristics in this study were reported in Wang et al. (2010a, b).

### 2.2 Sample collection and analyses

Litterfall was collected monthly in March through September 2008 using five litter traps (1 m × 1 m, 1 mm mesh size) in each plot and sorted into categories of leaf, small woody material, and miscellaneous material (everything other than leaf or small woody material) (Fang et al., 2007). Those samples were oven dried at 65 °C to constant weight and weighed.

To determine root biomass, a total of twelve soil cores (0–10 cm) were collected from each plot in August 2008 using an 8.7 cm diameter stainless steel corer, following the method advocated by Hendricks et al. (2006). We focused on fine roots because of their more rapid turnover rates compared to coarse roots, fine roots represent a substantial proportion of total tree root productivity (Eissenstat et al., 2000; Gill and Jackson, 2000; Guo et al., 2008). We opted to use fresh roots because they best represent roots that have not yet begun to decompose as described by Hobbie et al. (2010). Fine root samples were also oven dried at 65 °C to constant weight and weighed.

Soil samples (0–10 cm) were collected in August 2008 (Wang et al., 2010a). The organic horizons were not separately analyzed due to their very thin layers (∼1 cm) in...
the studied plantations. A total of six soil cores were collected using an 8.7 cm diameter stainless steel corer from each plot and bulked to form one composite sample. Soil samples were passed through a 2 mm sieve to remove coarse roots and gravel. A sub-sample of the soil was air dried at room temperature (25°C), and then was ground with a mill to pass through a 0.25 mm sieve before physicochemical analysis. The samples used for microbial community analysis were immediately stored at −20°C, without drying, for further analysis.

We used the solid-state $^{13}$C nuclear magnetic resonance spectroscopy with cross-polarization and magic-angle spinning (CPMAS-NMR) technique to study organic C chemical composition. This technique has frequently been used to directly and non-destructively study the complex structure (at molecular level) of SOC in terrestrial ecosystems (Kögel-Knabner, 2000; Schnitzer, 2001; Fontaine et al., 2007; Solomon et al., 2007). The solid state $^{13}$C CPMAS NMR spectra of soil, leaf litter and fine root samples were obtained at a frequency of 100.64 MHz on a Bruker AVANCE 400 spectrometer. Soil samples were pretreated with 10 % (v/v) hydrofluoric acid (HF) before NMR spectroscopy (Schmidt et al., 1997). To do that, approximately 10 g of ground sample were shaken with 50 mL HF for 2 h. After centrifugation (3000 rpm or 705 g) for 10 min, the supernatant was removed. The procedure was repeated five times. The remaining sediment was washed with 50 mL deionized water five times to remove residual HF and freeze-dried. The pretreatment removes a substantial amount of Fe$^{3+}$ and Mn$^{2+}$ in the soil, concentrates the organic C of the whole soil sample, and improves the signal/noise ratio of NMR spectroscopy (Schmidt et al., 1997).

To do the NMR analysis, samples were packed into a ZrO$_2$ rotor (o.d. = 7 mm) and spun at 5 kHz at the magic angle. Single contact time of 1 ms was used with an acquisition time of 42 ms, and a recycle delay of 1 s. Transients (20 000) were collected for all samples and a Lorentzian line broadening function of 50 Hz was applied to all spectra. Chemical shift values were referenced externally to glycine at 176.03 ppm, which is equivalent to tetramethylsilane at 0 ppm.
The $^{13}$C CPMAS NMR spectra were divided into four chemical regions that are assigned to specific organic C groups (Kögel-Knabner, 2002; Spielvogel et al., 2006; Wang et al., 2010): 0–45 ppm, alkyl C (lipids, cutin and suberin); 45–110 ppm, O-alkyl C (carbohydrates, cellulose, hemicellulose and methoxyl C); 110–160 ppm, aromatic C (lignin, tannin, olefins and aromatic compounds); and 160–220 ppm, carbonyl C (carboxylic acid, amide and ketone groups). The corresponding areas under the curve of the four regions were quantified by integration. The alkyl C/O-alkyl C ratio, which has been used as an index of the extent of decomposition of SOM or substrate quality for microbes (Baldock and Preston, 1995), or as an indicator of SOC chemical stability (Chen et al., 2004; Huang et al., 2008). In addition, the aromatic C/O-alkyl C ratio was calculated to characterize the extent of humification of SOM, under the assumption that organic matter becomes aromatic during decomposition (Dai et al., 2001; Dieckow et al., 2009).

Soil microbial biomass C (MBC) and N (MBN) were measured by the chloroform fumigation-extraction method (Brookes et al., 1985; Vance et al., 1987). Soil samples were also analyzed for phospholipid fatty acids (PLFAs) using the method described in Bossio and Scow (1998). The abundance of individual fatty acids was determined as nmol per g of dry soil using standard nomenclature (Tunlid et al., 1989). In our study, the concentration of each PLFA was calculated based on the 19 : 0 internal standard concentrations. Bacteria were identified by the following PLFAs: i14 : 0, i15 : 0, a15 : 0, 15 : 0, i16 : 0, a17 : 0, i17 : 0, 15 : 0 3OH, 16 : 1 2OH, cy17 : 0, 17 : 0, 16 : 1 ω7c, 18 : 1 ω7c and 18 : 1 ω9c (Frostergård and Bååth, 1996; Zelles, 1999). We calculated the sum of i14 : 0, i15 : 0, a15 : 0, 15 : 0, i16 : 0, a17 : 0 and i17 : 0 as the gram-positive bacteria (Kourtev et al., 2002; Liu et al., 2012), and the sum of 15 : 0 3OH, 16 : 1 2OH, cy17 : 0, 17 : 0, 16 : 1 ω7c, 18 : 1 ω7c and 18 : 1 ω9c as the gram-negative bacteria (Zelles, 1999). The fungi were identified by the PLFAs 18 : 2 ω6, 9c (Frostergård et al., 1993). Taken together, all of the above PLFAs and others such as 16 : 0, 18 : 0, cy 19 : 0 ω8c, 17 : 1 ω8c, 16 : 1 ω5c, 16 : 0 10methyl and 17 : 0 10methyl were considered to be part of the total PLFAs of the soil microbial community.
2.3 Statistical analysis

The initial C chemical compositions of leaf litter and fine roots, soil MBC, MBN, microbial C/N ratio, and soil microbial total, fungal, bacterial, gram-positive bacterial and gram-negative bacterial PLFAs were analyzed by one-way ANOVAs to determine differences among the four plantations. Multiple comparisons of means among the different plantations were performed using Duncan’s test. We compared soil alkyl C concentration and alkyl C/O-alkyl C ratio to soil microbial biomass, PLFAs and initial chemical fractions of leaf litter and fine roots using bivariate linear regressions. A logarithm transformation was performed on the data of soil alkyl C/O-alkyl C ratio and gram-negative bacterial PLFAs to meet the assumption of normal distribution prior to analysis. Twenty-three individual PLFAs (mol %) from the PLFA analysis of soil samples were subjected to principal component analysis (PCA) after standardisation for equal unit variance. Statistically significant differences were set at $\alpha = 0.05$ level. All analyses were performed using SPSS 19.0 for windows.

3 Results

3.1 SOC and litter C chemistry

Across the four plantations, the following SOC compositions were found: 43–49 % O-alkyl C, 24–34 % alkyl C, 14–17 % aromatic C, and 9–11 % carboxyl C. The SOC chemical composition varied with plantation type, with 34 % of the SOC found in the alkyl C fraction in the $P. massoniana$ plantation compared to $< 28 %$ in the broadleaf plantations. The leaf litter had 42–54 % O-alkyl C, 19–34 % alkyl C, 14–19 % aromatic C, and 8–9 % carboxyl C (Table 1). The fine root samples had 44–60 % O-alkyl C, 13–26 % alkyl C, 17–27 % aromatic C and 6–10 % carboxyl C (Table 1). The O-alkyl C, alkyl C, aromatic C and carboxyl C contents, and alkyl/O-alkyl C ratio of leaf litter and fine roots were also significantly different among the four plantations.
root samples were significantly different among the four plantations of different species (Table 1).

3.2 Soil microbial biomass and community composition

Soil MBC was lower in the *C. hystrix* than in the other three plantations ($p < 0.05$; Fig. 1a). Soil MBN was higher in the *M. macclurei* and *M. laosensis* than in the other two plantations ($p < 0.05$; Fig. 1b). Differences between the *M. macclurei* and *M. laosensis* plantations or between the *P. massoniana* and *C. hystrix* plantations were not significant (Fig. 1b). Soil microbial biomass C/N ratio was lower in the *M. macclurei* plantation than in the other three plantations ($p < 0.05$; Fig. 1c).

The amount of soil microbial PLFAs were significantly different among the four plantations (Fig. 2). Total PLFAs, bacterial PLFAs and gram-negative bacterial PLFAs were lower in the *C. hystrix* than in the other plantations ($p < 0.05$; Fig. 2b, e, and d). Fungal PLFAs were higher in the *M. laosensis* than in the other plantations ($p < 0.05$; Fig. 2a). Gram-positive bacterial PLFAs were significantly higher in the *P. massoniana* than in the other plantations ($p < 0.05$; Fig. 2c). Soil MBC was positively correlated with the amount of soil microbial total PLFAs ($R^2 = 0.39$, $p < 0.05$).

The first (PC1) and second (PC2) principal components using PLFAs accounted for 28.8 and 24.9 %, respectively, of the variation in the control plots (Fig. 3a and b). Principal component axis 2 clearly separated the *P. massoniana* from the other plantations. The PC2 was negatively correlated with soil O-alkyl C content ($R^2 = 0.31$, $p < 0.05$).

3.3 Relationships among litter C, soil microbial community and SOC chemical compositions

There was no significant relationship between stable SOC content and leaf litter or fine root stable C content across the four plantations. The soil alkyl C/O-alkyl C ratio was not correlated with the leaf litter alkyl C/O-alkyl C ratio ($R^2 = 0.14$, $p = 0.16$) but negatively correlated with the fine root alkyl C/O-alkyl C ratio ($R^2 = 0.45$, $p < 0.05$).
Soil microbial total PLFAs, microbial biomass C/N ratio, bacterial PLFAs and gram-positive bacterial PLFAs were positively correlated with soil alkyl C content \( (p < 0.05; \text{Fig. 4a–c, and e}) \). Soil bacterial PLFAs and gram-positive bacterial PLFAs were also positively correlated with soil alkyl C/O-alkyl C ratio \( (p < 0.05; \text{Fig. 4d and f}) \). Soil fungal PLFAs were not correlated with soil alkyl content and alkyl C/O-alkyl C ratio \( (\text{Fig. 5g and h}) \).

4 Discussion

A significantly higher soil alkyl C/O-alkyl C ratio in the *P. massoniana* than in the broadleaf plantations in this study indicates that a greater amount of relatively stable C accumulated in the soil of the *P. massoniana* than that of the broadleaf plantations, as the alkyl C/O-alkyl C ratio is an index of the extent of decomposition (Baldock and Preston, 1995). This would suggest that soil C in the pine plantation would be strongly buffered from environmental changes. Similar results were reported between pine plantations and oak or natural forests in temperate regions (Quideau et al., 2001; Chen et al., 2004).

A complex mixture of C-rich organic compounds including O-alkyl C (e.g., cellulose), aromatic C (e.g., lignin and tannins) and alkyl C (e.g., waxes, suberin, cutin) comprises all plant litter (Crow et al., 2009). The amount of plant litter, its chemical composition and properties are some of the key factors that affect the formation of SOM (such as the humification process) in terrestrial ecosystems (Scholes et al., 1997). The effects of vegetation type on SOC chemical composition could be attributed to the diversity in the C chemical fraction of litter materials and variations in the process of decomposition and humification (Quideau et al., 2001; Hannam et al., 2004). Previous studies pointed to the input and quality of litter as important regulators of C and N sequestration (Wardle et al., 2003, 2012; Cornwell et al., 2008; Brovkin et al., 2012).

Soil C chemical composition was not correlated to that of the initial leaf litter or fine roots in this study, suggesting that plant litter chemical composition may be not the
primary driving force determining the differences in SOC chemical composition among the four plantations. This is supported by a three-year manipulation experiment where the biochemical recalcitrance of the added litter had limited influence on the long-term stabilization of SOC (Gentile et al., 2011). It has been shown that aboveground plant litter dynamics on its own cannot explain the increasing rate of organic matter accumulation with time (Clemmensen et al., 2013). Therefore, our study suggests that the formation and stabilization of SOC was likely controlled by soil microbial populations, rather than plant litter C chemical composition. Tree species selection aiming to increase the input of recalcitrant litter may not be an effective practice to increase the stable SOC content.

The presence/absence of certain groups of decomposer organisms as affected by litter chemistry and environmental conditions could influence SOC chemical composition under different vegetation types (Baldock and Preston, 1995; Quideau et al., 2001; Hannam et al., 2004). Microbial residues in the soil are an important source for humus formation (Haider, 1992). Soil microorganisms are regarded as catalysts for plant residue transformation, and these organisms utilize the plant material as a C source, they therefore transform it to CO$_2$, metabolites, and biomass (Miltner et al., 2012). In this study, the significant difference in soil microbial biomass and the quantity of total PLFAs among the four plantations (Figs. 1 and 2), and the positive relationships between soil alkyl C content and total PLFAs and microbial biomass C/N ratio (Fig. 4a and b), indicate that the high soil microbial biomass maybe linked to the accumulation of alkyl C. Webster et al. (2000) also revealed that soil alkyl C/O-alkyl C ratio as well as microbial activity increased during a 28 day incubation. In contrast, lower soil alkyl C but higher soil microbial biomass existed under a natural forest compared with a first rotation hoop pine plantation in Australia (Chen et al., 2004). That may be accounted for by a combination of factors including chemical composition of litter materials, microbial community structure, and organomineral interaction, because the natural forest was a mixture of different tree and shrub species, including broad- and needle-leaved species (Chen et al., 2004).
The contribution of soil microbial populations to stable soil C is often affected by the biochemical fraction and macromolecular structure of microbial groups (Throckmorton et al., 2012). Both fungal and bacterial necromass can be stabilized in the soil (Miltner et al., 2012). The resistance of soil humic acids to microbial degradation was related to differences in their chemical structure and was also microbial species dependent (Yanagi et al., 2002). Melanin, waxes, terpenoids, and tetapyrrole pigments produced by bacteria maybe biochemically recalcitrant and resistant to biodegradation in the soil (Gleixner et al., 2001); the non-hydrolyzable melanin of fungi consists of proteins, carbohydrates, lipids, and phenolic polymers (Kögel-Knabner, 2002). In most cases the contribution of fungi was higher than that of bacteria (Six et al., 2006). However, in this study soil fungal PLFAs were not correlated with soil alkyl content and alkyl C/O-alkyl C ratio (Fig. 5g and h), indicating that the stable soil organic C composition was not linked to fungi-derived compounds. The significant correlations among soil bacterial, gram-positive bacterial PLFAs, soil alkyl C content and alkyl C/O-alkyl C ratio in this study (Fig. 5c–f) demonstrate that the high abundance of soil bacteria and gram-positive bacteria was linked to the high stable C content in the soil. This is because that bacteria contained more alkyl C but less O-alkyl C than fungi (Baldock et al., 1990), and unique to gram-positive bacteria is the presence of teichoic acids (containing lipid components) in the cell wall (Madigan and Martinko, 2006). Thus more aliphatic compounds of gram-positive bacteria than gram-negative bacteria could have been accumulated in soils in this study. Such linkages were supported by the greater soil retention of gram-positive bacteria compared with gram-negative bacteria necromass at a California forest site (Throckmorton et al., 2012).

The clear separation of *P. massoniana* from the broadleaf plantations on the PC2 axis (Fig. 3a), and the negative correlation between the PC2 axis and soil O-alkyl C content indicates that the higher abundance of gram-positive bacteria in the *P. massoniana* plantation could be one of the reasons for the differences in soil O-alkyl C composition among the four plantations in this study. Thus the specific microbial communities may
play a key role in the formation of characteristic SOC chemical compositions in forest ecosystems.

5 Summary

There was no significant linkage between chemical composition of SOC and initial chemical fraction of plant litter C in the four plantations of indigenous tree species in southern China. The stable SOC composition was attributable to the microbial-derived C compounds rather than plant leaf litter or fine root quality or a direct flux of C from recalcitrant litter materials to more stable SOC pools. The findings suggest that C input from recalcitrant litter does not directly contribute to increases in stable SOC through forest management practices in terms of afforestation, reforestation and litter management; soil microbial communities may play a more important role in determining the stable SOC pool. Future research should focus more on determining the dominant sources of stable SOC and their association with the microbial community.

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H. Wang et al.
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H. Wang et al.


Table 1. Distribution of leaf litter C and fine root C (%) among the chemical groupings of carbon in the four plantations (means with SE in brackets).

<table>
<thead>
<tr>
<th></th>
<th>Distribution of organic C</th>
<th>P. massoniana</th>
<th>C. hystrix</th>
<th>M. macclurei</th>
<th>M. laosensis</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Leaf litter C</strong></td>
<td></td>
<td></td>
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<td></td>
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<tr>
<td>Alkyl C (%)</td>
<td>19.3 (0.4) a</td>
<td>22.9 (0.3) b</td>
<td>26.9 (0.3) c</td>
<td>34.0 (0.3) d</td>
<td></td>
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<tr>
<td>O-alkyl C (%)</td>
<td>53.7 (0.1) d</td>
<td>49.0 (0.2) b</td>
<td>52.0 (0.1) c</td>
<td>42.3 (0.2) a</td>
<td></td>
</tr>
<tr>
<td>Aromatic C (%)</td>
<td>18.5 (0.3) c</td>
<td>19.3 (0.2) d</td>
<td>13.6 (0.1) a</td>
<td>14.6 (0.2) b</td>
<td></td>
</tr>
<tr>
<td>Carbonyl C (%)</td>
<td>8.6 (0.3) b</td>
<td>8.9 (0.2) b</td>
<td>7.5 (0.4) a</td>
<td>9.2 (0.2) b</td>
<td></td>
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<tr>
<td>Alkyl/O-alkyl C</td>
<td>0.36 (0.01) a</td>
<td>0.47 (0.01) b</td>
<td>0.52 (0.01) c</td>
<td>0.80 (0.01) d</td>
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<tr>
<td><strong>Fine root C</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Alkyl C (%)</td>
<td>13.5 (0.5) a</td>
<td>25.7 (0.4) c</td>
<td>21.0 (0.4) b</td>
<td>14.5 (0.7) a</td>
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<tr>
<td>O-alkyl C (%)</td>
<td>59.8 (0.4) c</td>
<td>44.0 (0.3) a</td>
<td>51.9 (0.2) b</td>
<td>50.9 (0.8) b</td>
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<td>Aromatic C (%)</td>
<td>21.2 (0.4) b</td>
<td>22.0 (0.3) b</td>
<td>17.0 (0.4) a</td>
<td>27.3 (0.7) c</td>
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<tr>
<td>Carbonyl C (%)</td>
<td>5.5 (0.3) a</td>
<td>8.3 (0.3) b</td>
<td>10.2 (0.1) c</td>
<td>7.3 (0.6) b</td>
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</tr>
<tr>
<td>Alkyl/O-alkyl C</td>
<td>0.23 (0.01) a</td>
<td>0.58 (0.01) d</td>
<td>0.40 (0.01) c</td>
<td>0.28 (0.01) b</td>
<td></td>
</tr>
</tbody>
</table>

Significant differences among species are identified with a, b, c and d at p < 0.05 level (ANOVA analysis, n = 4).
Fig. 1. Soil microbial biomass C, N and C/N in the four plantations. Error bars are standard errors ($n = 4$). Different letters indicate significant differences among stand types at $p < 0.05$. 

**Stable SOC is positively linked to microbial-derived compounds**

H. Wang et al.
Fig. 2. Soil microbial PLFAs in the four plantations. Error bars are standard errors (n = 4). Different letters indicate significant differences among stand types at p < 0.05.
Stable SOC is positively linked to microbial-derived compounds

H. Wang et al.

Fig. 3. Principal component analysis of the soil microbial PLFAs in the four plantations: (a) PCA scores for each of the plantations, and (b) PC loadings of the individual PLFAs. Each abbreviated code in Fig. 3b represents a biomarker lipid group. Bacteria were identified by the following PLFAs: i14 : 0, i15 : 0, a15 : 0, 15 : 0, i16 : 0, a17 : 0, i17 : 0, 15 : 0 3OH, 16 : 1 2OH, cy17 : 0, 17 : 0, 16 : 1ω7c, 18 : 1ω7c and 18 : 1ω9c. Fungi were identified by the PLFAs 18 : 2ω6, 9c. Other PLFAs such as 16 : 0, 18 : 0, cy19 : 0ω8c, 17 : 1ω8c, 16 : 1ω5c, 16 : 0 10methyl and 17 : 0 10methyl were also considered to be part of the soil microbial community (Frostergård et al., 1993; Frostergård and Bååth, 1996; Zelles, 1999).
Stable SOC is positively linked to microbial-derived compounds

H. Wang et al.

Fig. 4. Relationships among soil alkyl C content, soil alkyl C/O-alkyl C ratio and soil microbial biomass C/N ratio and PLFAs.