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Report

Variation of Soil Labile Organic Carbon Pools along an Elevational Gradient in the Wuyi Mountains, China

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Abstract: Soil labile organic carbon (LOC), a group of dynamic chemical compounds, is important in global carbon (C) cycling due to its short turnover time and sensitivity to environmental changes. However, variation of LOC along elevational gradients in subtropical forests is not fully understood. In this study, we investigated LOC groups, i.e. microbial available carbon (MAC), microbial biomass carbon (MBC), easily oxidation carbon (EOC), water-soluble organic carbon (WSOC), light fraction carbon (LFC) in three soil layers (0-10, 10-25 and 25-40 cm) in different communities along an elevation gradient in the Wuyi Mountains in southeastern China. We also examined plant litter mass (LM), soil temperature and moisture in three soil layers in all communities. We found that MAC, MBC, EOC and WSOC content increased along the elevation gradient across all soil depths, whereas LFC was higher in communities with low elevations compared to others across all soil depths. Soil temperature and moisture mainly regulated MAC, MBC, EOC and WSOC, and plant litter controlled LFC. Positive correlations were found among soil organic carbon (SOC) pools (MAC, MBC, EOC, WSOC, and SOC) (P<0.001) across communities, except for LFC. LFC was positively correlated to other pools at low elevations and high elevations, respectively. Overall, LOC pools decreased with increasing soil depth across communities. Our results suggest that LOC content principally dependeds on the amount of SOC and LOC groups are good indicators for predicting minor changes of SOC in the C cycle.

Key words: soil organic carbon; labile organic carbon; fractionation methods; soil depth; elevational gradient (vegetations)

1 Introduction

Soil organic carbon (SOC), a key component of the global C pool, plays an important role in C cycling. SOC is derived from a complicated mixture of fresh organic materials from plants, soil fauna, root exudates, microbial residues and chemically or physically protected substrates (Davidson and Janssens 2006), which generally consist of labile and recalcitrant pools (Lützow *et al.* 2007; Cheng *et al.* 2008). The soil labile organic carbon (LOC) pool is usually termed microbially available carbon (MAC), microbial biomass carbon (MBC), easily oxidation carbon (EOC), dissolved organic carbon (DOC), water-soluble organic carbon (WSOC), and light fraction carbon (LFC).

Different soil labile C pools usually positively correlate with each other (McLauchlan and Hobbie 2004), and they function as good indicators for predicting minor changes in SOC. Although LOC accounts for a small part of the SOC pool, it is vital in regulating nutrient availability to plants and microbes, as well as catalyzing the transformations of these nutrients in the soil (Haubensak *et al.* 2002). Moreover, it is very sensitive to changes in the vegetation community and soil microclimate, which can result from environmental changes (Cheng *et al.* 2008). Any changes in these factors would substantially alter LOC.

It is well known that vegetation is a major factor in regulating soil LOC (e.g. Cheng *et al.* 2008; Fissore *et al.* 2008). Changes in plant communities lead to alterations in

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soil microclimate, quality and quantity of aboveground C input, and the content, transformation and decomposition of organic C (Garten Jr et al. 1999). For example, surface soil always contains more LOC compared to that in deep soil layers (Cheng et al. 2008). Species composition of the vegetation may greatly influence the quality of litter (Hobbie and Gough 2004), which eventually incorporates into SOC. The vegetative community also controls the activities of microbes (Zak et al. 1990), through which detritus-related C is incorporated into various SOC factions (Loya et al. 2004). Variation in LOC is a main component in the flux of CO₂ between the terrestrial ecosystem and the atmosphere, which, in turn, will affect global climate change (Bardgett et al. 2008). Understanding the present state of LOC inventories allow us to qualitatively predict responses of SOC to climate change in the future.

The Wuyi Mountains, with a clear vertical zonation of vegetation communities (Wang *et al.* 2009), are located in the subtropics in the southeast of China. The complex interactions among plants, soil and microbes over centuries (Garden Jr. *et al.* 1999) provide an ideal model ecosystem to investigate changes in LOC along the elevational gradient. The objectives of this study are to: (i) estimate soil LOC contents in different vegetation communities and different soil layers along the elevational gradient; (ii) compare the relationships among SOC and LOC pools: MAC, MBC, EOC, WSOC and LFC.

2 Materials and methods

2.1 Site description

The experimental sites were located in the Wuyishan National Reserve Area in Fujian Province, China (27°33′– 27°54′N, 117°27′–117°51′E), a 56 527 ha forested area in the southeast of China. The annual mean temperature,

Table 1 Site conditions.

relative humidity, fog days, precipitation were $15.0 \,^{\circ}$ C, 83.5%, more than 100 days, and 2000 mm, respectively. There were four different vegetation types along the elevation gradient: evergreen broad-leaf forest (EBF), coniferous forest (CF), sub-alpine dwarf forest (SDF), alpine meadow (AM). Detailed site description is shown in Table 1.

2.2 Experimental design and soil sampling

Four plots (50×60 m) with different vegetation types (EBF, CF, SDF and AM) were set along an elevational gradient in the Wuyi Mountains. Each 50 m×60 m plot was divided into four 25 m×30 m subplots. Soil samples were randomly collected (0–10 cm, 10–25 cm and 25–40 cm depths) from all the subplots in November, 2006 using a 2 cm-diameter soil corer. Twenty soil cores were taken from each subplot at each soil depth and pooled together, as a replicate. Samples were immediately sieved (<2 mm) to remove soil fauna, rocks and fine roots, thoroughly hand-mixed, placed in plastic bags and transported in several coolers to the ecological laboratory at the Nanjing Forestry University. Small portions of the samples were naturally air-dried, and then prepared for measurement of soil chemical and physical properties, EOC, and LFC.

2.3 Soil carbon methods

Soil microbially available carbon (MAC) was determined using a sequential fumigation-incubation method according to Zou *et al.* (2005). A modified chloroform fumigationincubation technique (Jenkinson and Powlson 1976; Liu and Zou 2002) was used to measure microbial biomass carbon (MBC). Brief fumigation-incubation procedures were as follows: 30 g of fresh soil was fumigated for 36 hours with purified chloroform in a desiccator with moist

Site	Eleva- tion (m)		MAP (mm)	and	Sampling depth (cm)	g SOC (mg g ⁻¹)	$\frac{\text{TN}}{(\text{mg g}^{-1})}$	C:N	pН	Dominant species	Litter mass (t hm ⁻² y ⁻¹)
EBF	7 500	18.5	1700	Mountainous yellow earth, >100	0–10 10–25 25–40	48.67±1.94 28.32±1.77 27.50±6.36	5.50±0.19 4.70±0.17 4.54±0.42	8.91±0.67 6.00±0.16 5.85±0.88	4.78 ± 0.04	Castanopsis carlesii, Castanopsis eyrei	5.63±0.51
CF	1150	14.5	2000	Mountainous yellow earth, 90	0–10 10–25 25–40	50.12±5.80 29.57±2.60 29.02±1.72	5.52±0.46 4.70±0.70 4.58±0.42	9.36±1.67 6.49±0.44 6.41±0.37	4.68±0.13	Pinus taiwanensis, Oligostachyum oedogonatum	8.08±0.65
SDF	F 1750	11.2	2200	Mountainous yellow earth, >80	0–10 10–25 25–40	74.00±4.24 44.92±1.67 31.27±2.66	8.06±0.06 6.06±0.51 5.05±0.24			Symplocos paniculata, Stewartia sinensis	2.92±0.46
АМ	2150	9.7	3100	Alpine meadow earth, 45	0–10 10–25 25–40	129.56±12.35 103.61±14.37 40.32±5.79	11.43±1.36 9.77±0.73 5.44±0.59	10.97±2.19	5.06±0.25	Calamagrostis brachytrich, Miscanthus sinensis, Lycopodium clavatum	1.94±0.22

Note: Values are mean \pm SE (n=4). MAT, mean annual temperature; MAP, mean annual precipitation. Litter mass values are mean \pm SE. EBF: evergreen broad-leaf forest; CF: coniferous forest; SDF: sub-alpine dwarf forest; AM: alpine meadow. Datasets of MAT and MAP are obtained from He *et al.* (1994) and Zheng and Fang (2004).

paper towels. Unfumigated soil samples were placed in another desicator for 36 hours. Chloroform was excluded from the fumigated soil samples and all soil samples were inoculated with 1 g of unfumigated soil. Each fumigated soil sample, control and blank, together with a small plastic bottle (50 ml, without stopple) contained 15 ml of 1 M NaOH was placed in a 1 L Mason jar, and incubated at 25 °C for 10 days. The amount of CO₂ was determined by titration of the NaOH with 1 M of HCl to pH 8.3 in the presence of BaCl₂. Mason jars were flushed with compressed air to allow replenishment of O₂ between each interval and deionized water was added to maintain moisture at 60% of field capacity. The sequential fumigation-incubation procedure had a total of 8 cycles. Easily oxidation carbon (EOC) was measured by a KMnO4 (333 mM) oxidation procedure (Liginow et al. 1987; Blair et al. 1995). KMnO₄ solution (333 mM) and standard KMnO₄ solutions ranging from 0 to 333 nM were prepared. Soil samples containing approximately 15 mg of C were added to 50 ml plastic screw-top centrifuge tubes. The 25 ml KMnO₄ solution was added into each tube to react with the soil. The soil suspension was shaken on a reciprocating shaker for 1 hour at 12 rpm and then centrifuged for 5 min at 2000 rpm. A 1 ml aliquot of the solution supernatant was diluted to 250 ml and absorbance was measured on a spectrophotometer (Cecil Instruments Ltd., Cambridge, England) at 565 nm. The change in the concentration of KMnO₄ is used to estimate the amount of C oxidized, assuming that 1mM MnO₄ is consumed in the oxidation of 9 mg of C. Water-soluble organic carbon (WSOC) was extracted from 30 g of fresh

soil with an addition of 60 ml of deionized water. The mixture was shaken for 0.5h with 250 rpm at 25 °C, and centrifuged for 10 min at 15,000 rpm (Jiang *et al.* 2006). Then the supernatant liquid was filtered through a 0.45 μ m membrane. WSOC in extracts was measured by an automated TOC Analyzer (Shimazu, TOC-Vcph, Japan). Air-dried soil samples were separated into light and heavy fractions using NaI adjusted to a density of 1.70±0.02 g cm⁻³ (Strickland & Sollins 1987; Janzen *et al.* 1992). The light fraction was washed with deionized water, oven-dried at 50 °C for 24 hours, weighed, and analyzed for total C as light fraction carbon (LFC) by Elemental Analyzer vario EL III (Model CNS, ELEMENTAR Analysensysteme Ltd., Germany).

Soil organic carbon (SOC) and total nitrogen (TN) were determined by combustion with an elemental analyzer (Model CNS, Elementar Analysen Systeme GmbH, Germany). Soil pH values were measured with a Calomel electrode on a paste of 1:1 (w:v) of fresh soil and deionized water.

2.4 Statistical analysis

Two-way ANOVA was performed to examine the impact of vegetation and soil depth on different SOC pools. Linear regression analyses were used to evaluate the relationships between each two types of organic C pools. All statistical analyses were conducted using SPSS 15.0 software (SPSS Institute Inc., Chicago, IL, USA).

3 Results

The amount of MAC, MBC, EOC, WSOC and SOC



Fig. 1 Labile organic carbon pools along the elevational gradient. (EBF: evergreen broad-leaf forest; CF: coniferous forest; SDF: sub-alpine dwarf forest; AM: alpine meadow. MAC: microbially available carbon; MBC: microbial biomass carbon; EOC: easily oxidation carbon; WSOC: water-soluble organic carbon; LFC: light fraction carbon.)

Table 2 Results of two-way ANOVA of LOC pools in the four elevational vegetation communities.

	MAC	MBC	EOC	WSOC	LFC	SOC
Source of variation						
Vegetation	***	***	***	***	***	***
Soil depth	***	***	***	***	***	***
Vegetation	***	***	***	*	***	***
×Soil depth						

Note: ***: P<0.001; *: P<0.05. EBF: evergreen broad-leaf forest; CF: coniferous forest; SDF: sub-alpine dwarf forest; AM: alpine meadow. MAC: microbially available carbon; MBC: microbial biomass carbon; EOC: easily oxidation carbon; WSOC: water-soluble organic carbon; LFC: light fraction carbon.

significantly increased (P<0.001) along the elevational gradient, and decreased (P<0.001) with increasing soil depth (Table 2, Fig. 1). For example, MAC content in AM was 3.45 times larger that in EBF in the 0-10 cm soil layer (Fig. 1a). Different vegetation communities and soil depths

had a substantial influence on LOC pools and SOC (Table 2). LFC content, however, was highest in CF followed by that in EBF; and then in SDF and AM (Fig. 1e). But in the soil depth gradient, the LFC content also decreased significantly (Table 2). Estimates of EOC were much higher than MAC, MBC and WSOC, while WSOC content was the smallest. On average, the amount of EOC was 45-fold larger than that of WSOC. LFC content in the 0–10 cm soil layer was high, and the values were even larger than the EOC content in the communities of EBF and CF; in contrast, however, the amount of LFC decreased substantially with the soil depth. The LFC contents in the 10–25 and 25–40 cm soil layers were even smaller than the MAC contents.

Five empirical measurements of SOC pools, SOC, MAC, MBC, EOC and WSOC, positively correlated with one another across vegetation communities (Fig. 2k–t); while no positive correlation was found between LFC and other organic carbon pools. However, further analysis



Fig. 2 Relationships among microbially available carbon (MAC), microbial biomass carbon (MBC), easily oxidation carbon (EOC), water-soluble organic carbon (WSOC), light fraction carbon (LFC) and SOC (***P<0.001).

found that LFC was positively correlated to other pools at low elevation (EBF, CF) and high elevation (SDF, AM), respectively (Fig. 2a–j).

4 Discussion

Previous studies have reported that the state of organic C is closely related to natural changes, including biological and non-biological environmental conditions (e.g. Garten Jr. et al. 1999). Our results indicated that differences in the vegetation communities in the Wuyi Mountains were a major factor in determining the spatial variation of LOC pools and SOC along the elevation gradient and their relative vertical distribution (Fig. 1). Vegetation may affect soil C pools by influencing the quality and quantity of detritus supplied, soil physical and chemical properties (Loya et al. 2004; Jiang et al. 2006). We found that the content of MAC, MBC, EOC, WSOC and SOC significantly increased in different vegetation along the elevational gradient (Fig. 1). Thus, our finding supports previous studies indicating that plant species will influence C accumulation and cycling in soils through variation in microcliamte and microbial activities (Sun et al. 2002; Cheng et al. 2008; Sarkhot et al. 2008). Vegetation communities greatly affected the belowground structure of biological communities, so as to affect ecological processes, like soil C cycling and litter decomposition (Ruan et al. 2004). The content of MAC, MBC, EOC and WSOC in AM was significantly higher than those in the EBF, CF and SDF (Fig. 1). It was consistent with previous results (Insam and Domsch 1988; Smith and Paul 1990; Bauhus and Cote 1998; Kautz 2004) that showed soils in meadow had larger soil microbial biomass than that in forest. For example, grassland is better for the growth of soil microbes than forestland (Insam and Domsch 1988; Bauhus and Cote 1998). Meanwhile, herbaceous plants have a well-developed root system, which is densely distributed in topsoil. Furthermore, the exudates and decomposition of the roots are important C sources in the soil (Smith and Paul 1990).

More importantly, vegetation has a substantial effect on the content of light fraction carbon (LFC) (e.g. Murage *et al.* 2007; Cheng *et al.* 2008). We found that the amount of LFC in CF was the highest, followed by EBF, and it was the lowest in AM (Fig. 1). LFC consists mainly of partially decomposed plant debris (Crow *et al.* 2007), and is considered an intermediate stage between plant residues and stable organic matter (Gregorich and Janzen 1996). The dynamics of LFC content were found to be consistent with the variation f annual litter mass in different vegetation communities along the elevational gradient (Table 1).

SOC pools (MAC, MBC, EOC, WSOC, LFC, and SOC) decreased with increasing soil depth across communities (Fig. 1), which supported greater SOC favoring the growth

of microbes. A decrease of SOC with soil depth was found in the global analysis of the vertical distribution of SOC in shrublands, grasslands, and forests (Jobbágy and Jackson 2000). In addition, litter on the soil surface in forests is a C source as well as providing optimal conditions (Hendrick and Pregitzer 1996) for growth of soil microbes. Microbial and fungal activities declined significantly with increasing soil depth (Ralte *et al.* 2005). Moreover, the degree of C and nitrogen limitation to C mineralization increased with soil depth (Fierer *et al.* 2003a). Therefore, the amount of LOC was much higher in the 0-10 cm soil layer.

EOC and LFC pools were relatively larger compared with MAC, MBC and WSOC across different communities, especially in the 0-10 cm soil layer (Fig. 2). High EOC content in different communities may result from the nature of EOC which includes slower turnover than other labile C. LFC does not always consist of plant residues and animal remains, it may contain some charcoal and wood debris (Crow et al. 2007), which decay slowly. In addition, it may be closely related to our soil sampling date, which was in November. November is the late fall in the Wuyi Mountains and there was much plant litter. WSOC accounted for 0.06%–0.39% of SOC in this study. WSOC content was the smallest because WSOC typically accounted for less than 2% of SOC in forest soils (Lützow et al. 2007), and was always smaller than EOC, LFC, MBC, and MAC. Soil MBC usually constitutes 1%-5% of SOC (Sparling 1997). MBC pools were smaller than MAC (Fig. 2), implying that MBC was only a portion of MAC, which was consistent with the findings of Zou et al. (2005). In other words, some of the C remained in the soils after measuring MBC and was ready for mineralization by microbes (McLauchlan and Hobbie 2004).

The decomposition of plant litter through microbial mineralization transfers fixed C into the SOC pool in forests (Dam et al. 1997). The importance of terrestrial plant litter in C cycling has attracted considerable research (Lova et al. 2004). LFC is mainly composed of partially decomposed plant debris (Murage et al. 2007), which plant litter accounts for a large portion. At a regional scale, LFC is largely determined by the quality and quantity of plant litter. Litter is one of the major C sources in natural forests, and detritus-related microbes control important processes in soil ecosystems (Sanzone et al. 2001). How litter quality affects the transformation of litter into SOC pools is a key question in ecosystem science. The decomposability of litter primarily determined C loss from litter decomposition into the soil C pool (Fierer et al. 2005), though the annual LM in EBF and CF communities was higher (Table 1). Vegetation largely governed litter quality and litter decomposition rates (Hobbie 1996; Yadav and Malanson 2007). For instance, litter in CF was hard to decompose due to the large amount of waxes, resins, and lignin contained (Swift et al. 1979).

Significant correlations were found among LOC pools (MAC, MBC, EOC, WSOC and LFC) and SOC (Fig. 2f-t). Though there were large differences in the pool size of LOC, LOC content was largely dependent on the amount of SOC in the soil (McLauchlan and Hobbie 2004). Ajwa et al. (1998) found that the amount of potential mineralization C in the topsoil of alpine meadow with relatively greater SOC content was much higher than that in the farmland with smaller SOC content. Our finding of positive correlations among SOC and LOC pools was consistent with the findings of McLauchlan and Hobbie (2004), who measured MBC, acid-hydrolyzable C, C respired after 12 d of a laboratory incubation, and LFC in 33 restored grasslands. Chen et al. (2007) also found these positive correlations among SOC, MAC and MBC in a secondary oak forest and loblolly pine plantation in Xiashu forest in China. MAC and MBC correlated well (R^2 =0.960, Fig. 2r), likely because both MAC and MBC were determined using biological methods through measuring CO₂-C released during the course of laboratory incubation. The positive correlations among the labile C pools suggested that though the definitions and methods varied from one study to another, all the LOC could represent a portion of SOC with a relatively rapid turnover rate, and they were important food and energy sources for the belowground ecosystem. McLauchlan and Hobbie (2004) suggested that different LOC pools are appropriate for use as indices of the quality of SOC, and each method provides an accurate index of relative changes in soil labile C pool. Positive correlation between EOC and SOC indicated that they were all measured by chemical oxidation after the soils were air-dried, while MAC, MBC and WSOC were determined with fresh soil samples, and LFC was estimated by density fractionation.

In all sites along the elevational gradient, it seemed that LFC did not correlated with MAC, MBC, EOC and SOC, however, further analyses unmasked positive correlations (Fig. 2a–j) between LFC with MAC, MBC, EOC, WSOC, and also SOC after the C pools were separated into two groups: lower elevation group (EBF and CF) and higher elevation group (SDF and AM). This was because labile C pools, except LFC, increased along the elevational gradient, while LFC content was higher in EBF and CF with lower elevation; in each community, however, both LOC and SOC decreased with increasing soil depth, which resulted in the positive correlations between LFC and others.

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武夷山沿海拔梯度土壤活性有机碳库的变化

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摘要:土壤活性有机碳 (LOC) 是一组活跃的化学物质,由于其较短的周转时间和对环境变化的敏感性在全球碳循环中发挥着 重要的作用。但是,对活性有机碳在亚热带森林沿海拔梯度的空间变异还缺乏了解。在本研究中,我们测定了福建武夷山自然保 护区不同海拔高度具有代表性的中亚热带常绿阔叶林 (500 m)、针叶林 (1150 m)、亚高山矮林 (1750 m)以及高山草甸 (2150 m)土壤 不同土层(0-10, 10-25 和 25-40 cm) 中微生物可利用碳 (MAC)、微生物量碳 (MBC)、易氧化碳 (ROC)、水溶性碳 (WSOC)和轻组 碳 (LFC),并观测了相应的植物凋落物质量 (LM),土壤温度和湿度。结果表明:沿海拔梯度的植被变化和土层深度变化对土壤活 性有机碳有显著的影响。微生物可利用碳、微生物量碳、易氧化碳和水溶性碳在不同土层均沿海拔高度的增加而显著增加;而低 海拔的常绿阔叶林和针叶林中轻组碳的含量高于亚高山矮林和高山草甸的轻组碳的含量。各土壤活性有机碳库均随土层的加深而 减小。除轻组碳外,各碳库间存在着极显著的正相关 (p<0.001)。轻组碳分别在低海拔 (常绿阔叶林和针叶林)和高海拔 (亚高山矮林 和高山草甸)与各碳库显著相关。

关键词:土壤有机碳;活性有机碳;分离方法;土层厚度;海拔梯度植被变化