Ethiop. J. Biol. Sci. **16(2): 171–188, 2017**
 ISSN: 1819-8678

SOIL MICROBIAL BIOMASS AND SOIL RESPIRATION IN MUNESSA FOREST, ETHIOPIA: A COMPARISON BETWEEN NATURAL AND ADJACENT PLANTATION FOREST

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ABSTRACT: Conversion of natural forests to monocrop plantations is a common forest management, yet its impact on ecosystem processes has rarely been evaluated and some earlier studies along the same line were inconsistent and contradictory. The present study compared the difference in soil microbial biomass and soil respiration between natural and adjacent *Cupressus lusitanica* plantation in Munessa forest. Weekly measurements of soil CO_2 efflux were carried out using LI-8100 infrared gas analyzer at 20 randomly selected locations from May 2009 to April 2010. On both forest stands, organic C, total N and microbial biomass declined significantly with increasing soil depth ($p<0.05$). There were no major differences in microbial composition and dominance between the two forest types. However, the total phospholipids fatty acid (PLFA) tended to be significantly larger in soils under the natural forest stand than adjacent plantation forest stand ($p = 0.032$). Mean soil CO_2 efflux rates was found to range from 2.23 and 7.22 µmol $m^{-2}s^{-1}$ versus 2.39 to 6.84 µmol $m^{-2}s^{-1}$ in the natural and plantation forest stand, respectively. Soil moisture appeared to be the major variable controlling soil CO_2 efflux rate. Unlike soil moisture, soil temperature had weak relation with soil CO_2 efflux and was better explained in the plantation forest stand using exponential function ($r^2 = 0.12$; $p<0.01$). The results indicate that the natural forest is better in sustaining soil microbial biomass and nutrients than monocrop plantation. We conclude that the natural forest played a significant role in the annual forest ecosystem carbon budget.

Key words/phrases: PLFA, Soil CO_2, Soil moisture, Soil temperature.

INTRODUCTION

Forests play a decisive role in the global carbon cycle and provide diverse services and values to human society. In Ethiopia, however, land clearing for agriculture coupled with high wood demands have led to increased deforestation. Realizing this fact, in the early 1970s large scale forest plantation programs were started with the objective to meet the ever-increasing demand of forest products, to rehabilitate degraded land and at the same time to promote soil and water conservation (Pohjonen, 1989). In some parts of the country (e.g., Munessa forest), plantations were usually

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established after natural forests have been logged and then burned to clear the land for planting (Chaffey, 1982). Currently, *Cupressus lusitanica* plantation forests have become the dominant source of domestic inputs for the Ethiopian forest industry.

There is a widespread perception that conversion of natural forest to managed land use can have a marked effect on variety of soil properties in forests, which may in turn alter soil microbial community structure, and soil physicochemical properties and soil respiration (Araújo *et al*., 2010; Chiti *et al*., 2018). After removal of the natural forest and transformation into forest plantation, Yeshanew Ashagrie *et al*. (2003; 2005) observed a significant decrease in organic carbon and nitrogen storage for soils of the Munessa forest. Most of them occurred in the organic layer and the particulate organic matter fraction of the mineral soil. Owing to difference in the genetic makeup of the host plant and differences in the intimacy of root inhabiting microbes, differences in abundance of microbial community are likely under the rhizosphere of different tree species (Ayres *et al*., 2009). Soil microbial biomass is a living component of organic matter and plays a critical role in organic matter decomposition, nutrient cycling and energy transfer processes in forest ecosystem (van der Heijden *et al*., 2008). Forest types influence soil microbial biomass. As a result, microbial biomass of soil is being increasingly recognized as a potential early and sensitive indicator of soil organic carbon changes (Cookson *et al*., 2007). The relative contribution of autotrophic and heterotrophic respiration to total soil respiration varies widely depending on the types of vegetation. On the other hand, soil temperature and soil moisture content are the key known environmental variables affecting soil respiration. Therefore, the information regarding the impacts of vegetation types on soil respiration is fundamental to understand the future terrestrial carbon balance in the context of climate change.

Understanding the role humans play in forest management practice would be useful for developing a clear policy advice related to forest conversion and more importantly in addressing our current and emerging environmental challenges like climate change. Except few related studies (e.g., Ambachew Demessie, 2009; Mehari Alebachew, 2015), little is known about the effects of natural forest conversion to plantation on microbial biomass and soil CO_2 efflux under Ethiopian forest vegetation. Consequently, the present study was undertaken in natural forest and adjacent plantation at the Munessa forest, southern Ethiopia. The objectives of the study were (i) to determine the difference in microbial biomass between natural and adjacent plantation

forest stands under the canopy of the selected tree species; (ii) to quantify and compare soil CO_2 flux rate between the two forest stands, and (iii) to elucidate the relationship between soil temperature, soil moisture and soil CO_2 efflux rate in the two forest stands.

MATERIALS AND METHODS

Site description and weather data

The study was carried out in Munessa-Shashemene Forest, West Arsi Zone, Oromia regional state (07°25'56" and 07°25'58" North latitudes and 38°51'58" and 38°51'11" East longitudes). The forest is located 250 km south of Addis Ababa and covers the northwest exposed slopes of the Rift Valley, north of Shashemene and east of Lake Langano (Fig. 1).

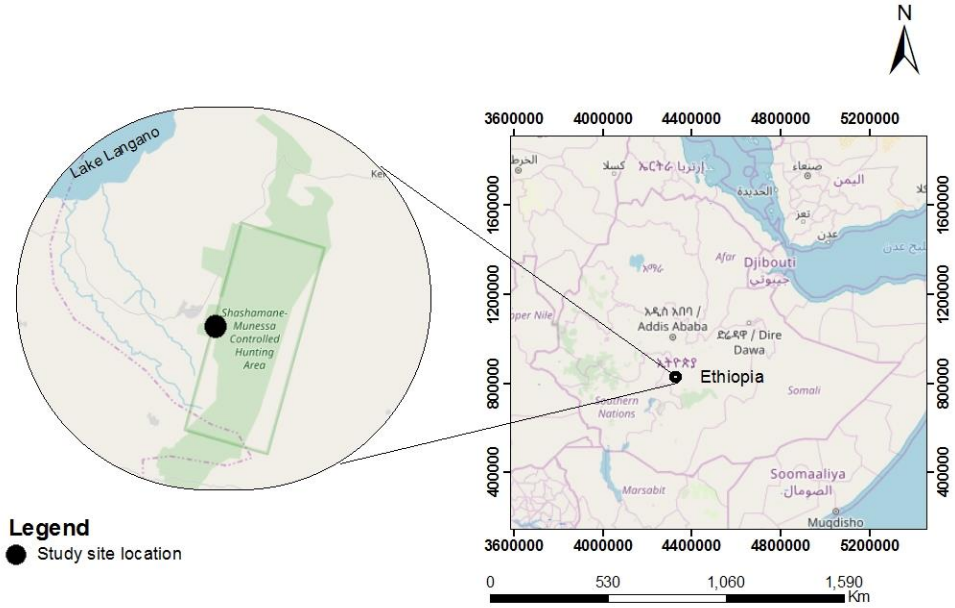

Fig. 1. Location map of the study area.

The forest has an estimated area of 23,000 ha of both natural and plantation forest. It is divided into three blocks; namely Gambo, Sole and Degaga. For the present study, the experimental sites were established in the Degaga block. The annual average precipitation is about 1500 mm, of which the major rainy season falls between July and November with a small rainy season occurring from March to May. Mean annual air temperature is about 15.3°C. Vegetation of the natural forest is dominated by the canopy species *Podocarpus falcatus* (Thunb.) and *Croton macrostachyus* Hochst. The

plantation stand (*Cupressus lusitanica*) was established in 2003 as part of commercial forestry activity and administered by the Oromia Forest and Wildlife Enterprise. Meteorological parameters were continuously recorded by weather stations installed for this purpose close to the study site. Air temperature was measured using µMETOS SMT 160–30 sensor, Pessl Instruments GmbH, Werksweg, Germany. Bulk precipitations were determined on a weekly basis using five randomly distributed polyethylene funnels with a 120 mm upper diameter. The mean monthly temperature and precipitation during the study periods are shown in Fig. 2. The wet and dry periods were separated following the approaches of Gibbs and Maher (1967), where the distribution of precipitation events over a long-term record is divided into sections for each ten percent of the distribution. Such rainfall deciles were calculated from ten years historical rainfall data (1998 to 2007) obtained from a nearby meteorological station (Degaga town 07° 26' 00'' N and 038° 50' 26'' E).

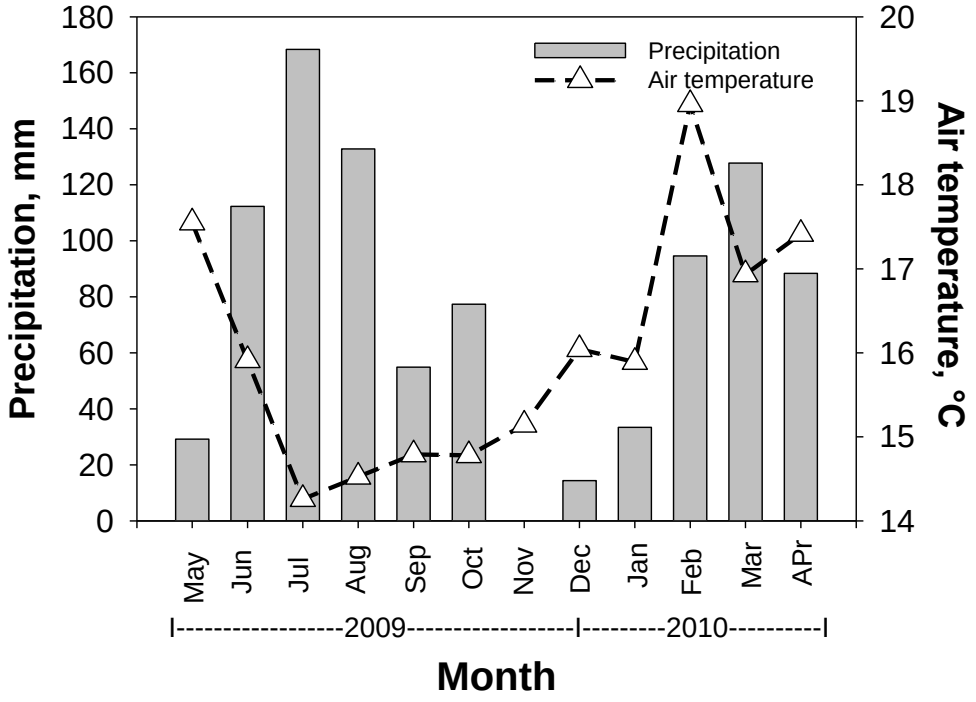

Fig. 2. Monthly total precipitation (bar graph) and mean monthly air temperature (line graph) in the study area from May 2009 to April 2010.

Stand selection and plot establishment

In order to select candidate trees that are comparable in height class with that of the plantation forest stand, the third height class trees of two indigenous trees namely *C. macrostachyus* and *P. falcatus* were considered in the natural forest stand (Getachew Tesfaye *et al*., 2010). All trees with a third height class were first identified and given random numbers. From those trees, two individual trees that were available at the distance of ca. 100 m were randomly selected using simple random sampling techniques. Under the canopy of these trees, a total of four experimental plots (3 x 3 m) were established. Similarly, four experimental plots with a same plot size were also established under the plantation forest stand. The two indigenous trees species were selected because of their relative abundance and ecological importance. *Cupressus lusitanica* (Mill.) was selected because of the vicinity to the natural forest and it is also the dominant planted tree species. Out of 6973 ha, *C. lusitanica* accounted for 60% of the total plantation area (Tesfaye Teshome and Petty, 2000). The *C. lusitanica* stand was planted in July 2003 at a spacing of 2.5 x 2.5 m. In January 2008, selective thinning was done by removing the competitor trees next to trees assigned as potential crop. The average height and diameter at breast height in 2010 were 6.1 m and 8 cm, respectively, and density of trees is 560 trees/ha. The eight plots had a similar topographical feature and the underlying soils are developed from the same parental materials rich in clay evolved from volcanic parent material. At the experimental plots they were classified as Mollic Nitisols according to the WRB system (FAO, 1998). More detailed information on soil and local climatic conditions can be found in Fritzsche *et al*. (2007).

Soil sampling and laboratory evaluation

From each plot, soil samples were collected from three random points during wet and dry seasons separately. The samples were collected using soil cores (equipped with a detachable cylindrical steel core, 4.0 cm diameter, 40 cm length) from 0–10 and 10–25 cm depths. Soil samples collected under the respective sampling plots were bulked and passed through a 2 mm sieve to remove all visible roots, macro-fauna and fresh litter. During sieving, roots were removed from the soil by forceps on the day of sampling. From the homogenized sample, sub-samples were collected separately for phospholipids fatty acid (PLFA) and neutral lipid fatty acid (NLFA) analysis. These samples were transferred into separate glass vials and kept frozen until analysed. The remaining soil samples were

air dried and stored until analysed for physicochemical characteristics.

For physicochemical characterization, dried soil samples were finely grinded with a steel ball mill (Mixer Mill, Retsch MM 200, Haan, Germany) and oven dried overnight at 105°C. Dried powder sample was packaged in tin capsules and analysed for total carbon and nitrogen measured by dry oxidation or combustion method using a Vario EL III elemental analyzer (Elementar Analysensysteme GmbH, Germany). As the soils did not contain carbonates, total carbon was assumed to represent organic carbon. Soil texture was determined by the pipette method. Soil pH was analyzed potentiometrically in 1 M KCl [1:2.5 (m/v)]. Cation exchange capacity was determined with the $BaCl_2$ compulsive exchange method (Gillman and Sumpter, 1986).

PLFA extraction and quantification

PLFAs were extracted in three steps using a modified Bligh and Dyer (1959) procedure. In brief, lipids were extracted overnight from frozen soil samples (1.5 g) using a one phase mixture of citrate buffer, chloroform, and methanol at a ratio of 0.8:1:2 (v/v/v). The samples were then fractionated into neutral- glycol- and phospholipids using solid phase extraction with silicic acid column (Bond Elut LRC-Si, Varian Agilent Technologies, Santa Clara, CA). Thereafter, the neutral and phospholipids were subjected to a mild-alkali methanolysis, and the resulting fatty acid methyl esters were separated by gas chromatography using an Agilent 7890A GC-MS (Varian Agilent Technologies, Santa Clara, CA). The fatty acids were identified based on retention times relative to that of the internal standard (fatty acid methyl ester 19:0).

Standard nomenclature was used to refer to the PLFAs according to the designation described in Zelles (1999). In the present study, fourteen fatty acids were identified using the retention times determined for soil PLFAs by gas chromatography-mass spectrometry (Frostegård *et al*., 1993). Phospholipid fatty acids i15:0, a15:0, i16:0, 18:1ω7c and cy19:0 considered to be of bacterial origin only were summed to give an index of the proportion of bacterial biomass (Frostegård *et al*., 1993; Zelles, 1999). Whereas, the PLFAs 18:2ω6 and 18:1ω9 were considered to have fungal origin (Frostegård and Bååth, 1996; Zelles, 1997), and the Neutral Lipid Fatty Acid (NLFA) 16:1ω5 was used as marker for arbuscular mycorrhizal fungi (Olsson, 1999). Finally, the sum of the fatty acid indicator of bacteria and fungus plus the six additional fatty acids (16:1ω9, 16:1ω7c, 16:1ω5, 16:0, 10Me16:0a and 10Me16:0b) were used to represent microbial biomass

(Federle, 1986; Frostegård *et al*., 1993). Molar amounts (nmol PLFA g^{-1} dry soil) of the individual PLFAs were used as an estimation of microbial biomass.

Soil respiration measurement

In each plot, five polyvinylchloride (PVC) collars (20.3 cm in diameter) were gently inserted two weeks before the soil CO_2 efflux measurement campaigns. The PVC collars were randomly distributed into the soil around the neighbouring trees at a distance of about 0.5 to 1.3 m from the bole. The insertion depth was kept 1–2 cm to minimize severing of roots and the mycorrhizal system, while ensuring that no leaks occurred in the chamber-soil system. The soil respiration chamber was set on top of these collars, allowing an undisturbed measurement of soil CO_2 flux rates. CO_2 release from the forest soil was measured using an Infrared Gas Analyzer Li-8100 supplied by LI-8100-103 soil survey flux chamber (LI-COR, Lincoln, NE, USA). Each flux chamber has an internal volume of 4076.1 cm^3 with an exposed soil area of 317.8 cm^2. Soil CO_2 efflux measurements were taken every week in the morning from May 2009 to April 2010. Soil collars were remained in place throughout the duration of the experiment. Simultaneous to the soil CO_2 efflux measurements, soil temperature (°C) was also measured adjacent to each PVC collar at a depth of 0.1 m using a thermocouple probe (Li-8100-201) connected to the Li-8100. The volumetric soil water content (v/v%) was measured with a handheld theta probe (ML2, Delta-T Device Ltd, Cambridge, UK) inserted into 0.06 m depth at three positions adjacent to each PVC collar.

Statistical analysis

Differences between the two forest stands in physical and chemical properties of soil were analyzed with a One-way Analysis of Variance (ANOVA) and post hoc tests using Tukey's honestly significant difference (HSD) methods. The relative proportions of PLFAs (nmol g^{-1} dry weight of soil) expressed as arithmetic means ± standard deviations were used to describe the microbial community structure. A Three-way Analysis of Variance (Three-way ANOVA) with Holm-Sidak's test ($p<0.05$) was used to determine the significant differences of forest stand, sampling season, and soil depth on soil microbial biomass. Both linear and nonlinear regression models were used to regress soil respiration against soil moisture and/or soil temperature. Each equation was fit using a repeated trials approach and the performance of the equation was evaluated using root mean square error (RMSE), and the coefficient of determination (r^2). Based on preliminary

graphical analysis, the dependence of soil respiration on soil moisture was best fitted using the following Gaussian model both in terms of maximal r^2 values as well as fitting the downturn of respiration rates for high soil moisture values:

$SR = a*\exp(-0.5*((SM-x_0)/b)^2)$

where SM is soil moisture (%) and a, x_0 and b are fitted parameters.

The inclusion of soil temperature effect in the Gaussian model was tested as additional predictor using different functions. In all types of linear and nonlinear regression model tested, the combination of this variable leading to insignificant change in root mean square error (RMSE), and the coefficient of determination (r^2). All graphing and/or statistical analyses were conducted using SigmaPlot version 11 (Systat Software Inc., San Jose, CA, USA) and/or R 3.1 (R Foundation for Statistical Computing, Vienna, Austria).

RESULTS AND DISCUSSION

Soil physical and chemical properties

The soil texture of the two forest stands was found to be comparable, indicating that the soils developed from the same parent material. Soils of the natural forest stand were characterized by a higher (though not statistically significant) soil organic carbon (SOC) and total nitrogen (TN) concentrations compared to the plantation stand at the depth of 0–10 cm (Table 1). A high concentration of soil organic carbon and total nitrogen were reported for the site (Yeshanew Ashagrie *et al*., 2003), and are attributed due to high rates of litter input in these productive stands and large contents of reactive minerals (i.e., Fe- and Al-(hydroxides)), which stabilize SOC by chemically interacting with minerals (Fritzsche *et al*., 2007).

There was no significant difference in soil pH as well as base saturation (BS) between the two forest stand ($p>0.05$), except that soil pH under the plantation forest tend to be slightly more acidic (Table 1). A similar trend was observed in cation exchange capacity (CEC). The CEC was larger under the natural forest stand, driven primarily by differences in exchangeable Ca^{++}. On the other hand, SOC concentration and other parameters decreased with depth (higher in 0–10 cm) significantly ($p<0.05$) owing to the fact that leaf litter enters the soil surface and root biomass is the largest in the topsoil (Asferachew Abate, 2004). Concurrent to the decline in SOC with soil depth, CEC decreased with soil depth showing the

impact of soil organic matter on exchangeable nutrients in these forest soils (Table 1). At a soil depth of 10–25 cm, SOC was significantly different between natural and plantation forest stand ($p<0.001$).

Table 1. Basic characteristic of the soils from 0–10 cm and 10–25 cm soil depths from respective forest stand.

Stand/ Soil depth	Sand	Silt	Clay	C	N	pH	CEC	BS
		–g kg^{-1} –			– g kg^{-1} –		mmol$_{(+)}$ kg^{-1}	%
Natural forest								
0–10 cm	201±22	384±10	414±206	127±22a	11±1.0a	6.6±0.1a	645±54a	100
10–25 cm	219±2	301±24	480±25	61±6b	6±0.4b	6.4±0.2a	457±63b	100
Plantation forest								
0–10 cm	210±15	395±62	395±67	114±27a	10.2±2.3a	6.0±0.5a	638±92a	100
10–25 cm	189±8	371±65	440±70	39±15c	4.3±1.6b	5.7±0.6a	449±69b	100

Values are mean and standard deviation (n = 3). Different letters in each column indicate significant differences ($p<0.05$) with one-way ANOVA, post hoc Tukey's test. BS stands for base saturation

The quality and quantity of input by net primary production and its decomposition rate are the two most important factors influencing the amount of SOC storage (Lutzow *et al*., 2006). The higher concentration of SOC and TN in the natural forest could have resulted from higher litter mass accumulation (Asferachew Abate, 2004). Moreover, the observed difference between the two forest stand can be related to differences in root litter input. In a previous study, a higher live fine root biomass (up to 1 m soil depth) was recorded for the same natural forest (dominated by *P. falcatus*) compared to *C. lusitanica* (Asferachew Abate, 2004). The history of the plantation site might have also contributed to differences in soil quality between the two forest stands. Before reforested, part of the plantation stand has been used for agriculture (for about three years). Agricultural land use is known to deplete soil organic matter as reported earlier for the same forest (Mulugeta Lemenih *et al*., 2005).

Phospholipid fatty acid concentration

In general, the sum of all PLFA concentration followed the patterns of soil organic C and TN content. The normal saturates 16:0 and monosaturate 18:1ω17c, methyl branched 10:Me16b and branched chain saturate i15 were the most abundant PLFAs, accounting for ca. 54% of the total concentration (Table 2). Monounsaturate 16:1ω7, 18:1ω9, and Cyclopropyl fatty acid cy19:0 were found to be the second most predominant group (24%; Table 2). Branched-chain saturates (a15 and i16), monosaturate 16:1ω5 and methyl branched 10:Me16a were 19% of the total concentration (Table 2). A 18:2ω6 and 16:1ω9 were 3% of the total PLFA concentration (Table 2). There were no statistical differences ($p = 0.320$) in mole percentage of individual PLFAs between the forest stands, which indicates the lack of

noticeable effects of soil origin. In general, soil microbial biomass tended to be larger under the natural forest than the plantation forest stand (Table 2). At a depth of 0–10 cm, biomass of fungal marker indicator estimated as sum of PLFA concentration 18:2ω6 and 18:1ω9 (Frostegård and Bååth, 1996; Zelles, 1997) were: 9.5 ± 1.6 and 9.3 ± 0.3 (natural forest) vs. 10.0 ± 0.8 and 9.5 ± 1.1 nmol fungal PLFA g^{-1} dry soil (plantation forest) for wet and dry seasons, respectively (Table 2). At the same soil depth, the values for bacterial marker indicator (Zelles, 1999) in the natural forest stand were: 45.5 ± 1.6 and 45.0 ± 2.3 vs. 40.0 ± 0.8 and 43.9 ± 1.4 nmol bacterial PLFA g^{-1} dry soil for wet and dry seasons, respectively (Table 2). From the depth 10–25 cm soil layer, biomass of bacterial or fungal marker indicators in the respective sampling seasons were: 47.2 ± 5.4 and 46.6 ± 3.4 nmol bacterial PLFA g^{-1} dry soil or 9.7 ± 0.6 and 9.4 ± 0.69 nmol fungal PLFA g^{-1} dry soil (natural forest) vs. 42.4 ± 3.3 and 46.4 ± 5.6 nmol bacterial PLFA g^{-1} dry soil or 9.6 ± 1.9 and 10.0 ± 1.4 nmol fungal PLFA g^{-1} dry soil (plantation forest) for wet and dry seasons, respectively (Table 2).

The sum of all PLFA concentration expressed in nmol PLFA g^{-1} dry soil (sum of soil microbial biomass) significantly affected by forest type [ANOVA: $F(1,40) = 13.349$, $p<0.001$]. Similarly, soil microbial biomass was significantly affected by soil sampling depth [ANOVA: $F(1,40) = 67.610$, $p<0.001$]. On the other hand, soil microbial biomass was not affected by sampling season [ANOVA: $F(1,40) = 0.363$, $p<0.274$]. There were no significant differences in two-way or three-way interaction effects on soil microbial biomass (Table 3). In both forest types and sampling season microbial biomass decreased from 0–10 to 10–25 cm soil depth. The declining PLFAs concentration with soil depth is the result of accompanied decline in organic matter and is a common pattern across many ecosystems (Fall *et al*., 2012). Soil microbial biomass was higher in wet season than in the dry season. This is most common in tropical forest soils where soil microbial biomass over time is closely associated with changes in water availability and temperatures that do not vary greatly (Pabst *et al*., 2013). This suggests that rains enhanced soil CO_2 flux by stimulating both soil microbial activity and growth.

Table 2. Mole percentage of individual fatty acids and total PLFA from 0 to 10 and 10 to 25 cm soil depth in the natural and the plantation forest.

Forest stand	Soil depth (cm)	Season	mol% of individual fatty acid													Total PLFA*
			i15	a15	i16	16:1ω9	16:1ω7c	16:1ω5	16:0	10:Me16a	10:Me16b	18:2ω6	18:1ω9	18:1ω7c	Cy19	
PF	0–10	Wet	10.6	5.4	5.2	1.5	7.6	4.5	16.7	3.9	12.9	1.7	8.4	13.4	8.3	143.8 (33.4)
		Dry	11.3	5.7	5.3	1.4	7.6	4.3	16.4	3.8	13.0	1.6	7.9	13.6	8.0	132.5 (3.7)
	10–25	Wet	10.1	5.0	6.0	1.0	6.6	3.9	18.8	3.5	14.2	2.5	7.1	12.2	9.1	66.6 (32.4)
		Dry	11.4	5.9	6.1	1.5	6.6	4.0	17.5	2.7	11.3	2.1	7.9	13.4	9.6	71.3 (20.6)
NF	0–10	Wet	10.7	5.7	5.1	1.4	7.6	4.7	17.1	3.6	10.9	1.7	7.8	15.1	8.6	185.4 (27.3)
		Dry	11.6	6.1	5.2	1.5	7.9	4.9	15.0	4.0	11.9	1.5	8.0	13.8	8.8	148.0 (32.4)
	10–25	Wet	11.2	6.1	5.9	1.5	6.8	4.4	14.3	3.4	12.6	1.8	7.9	14.4	9.8	101.9 (10.5)
		Dry	12.0	6.4	6.1	1.5	7.0	4.3	14.8	3.7	12.8	1.5	7.7	12.8	9.4	94.8 (17.6)

* nmol PLFA g^{-1} dry soil. Values shown in Table 2 are averages of three samples and the values in parentheses represent standard deviation. PF stands for plantation forest; NF stands for natural forest

Table 3. Summary of three-way analysis of variance (ANOVA) of forest type, season and soil depth effect on soil microbial biomass.

Source of variation	df	Mean square	F-values	P-values
Forest type	1	9743.12	13.349	<0.001
Season	1	265.23	0.363	0.274
Soil depth	1	49345.92	67.610	<0.001
Forest type x Season	1	697.56	0.956	0.336
Forest type x Depth	1	58.06	0.0795	0.780
Season x Depth	1	4.29	0.00587	0.939
Forest type x Season x Depth	1	518.89	0.711	0.405
Residual	40	23355.73		
Total	47	85537.24		

Generally, microbial biomass can offer a means in assessing the soil quality in different vegetation types (Groffman *et al*., 2001). It is clear that there must be a positive relationship between the substrate and those who are using the substrate (e.g., Wardle, 1992; Lee and Jose, 2003). Soil microbial biomass can be influenced by differences in the quantity and quality of substrate inputs via varying litter and root types and associated nutrient (Feng *et al*., 2009). For example, Dawit Solomon *et al*. (2002) have found for the same forests significantly higher production of microbial metabolites below the natural forest as indicated by higher ratios of microbial to plant-derived sugars. The higher microbial biomass in the natural forest in comparison with the plantation forest stand is in agreement with a study carried out in tropical and subtropical forest ecosystems (Behera and Sahani, 2003). These results corroborate the notion that the microbial biomass is a good indicator of soil quality since microbes respond against changes in vegetation cover more noticeably than the physicochemical properties of the soil.

Soil respiration and abiotic factors

Soil CO_2 efflux rates were larger in the natural forest stand ($p<0.05$) than in the plantation forest (Fig. 3). Across the parallel measurement campaign, mean soil CO_2 efflux rates in the natural forest stand ranged from 2.23 to 7.22 μmol $m^{-2}s^{-1}$ (Fig. 3). Corresponding values at the plantation forest stand ranged from 2.39 to 6.84 μmol $m^{-2}s^{-1}$. The higher rates of soil CO_2 efflux in the natural forest stand may be associated with synergistic biological activities of soil microbes and plants; and larger organic C concentration that could accelerate both the plant root and microbial activities (Dawit Solomon *et al*., 2002; Asferachew Abate, 2004). Hence, both components of the autotrophic continuum (mycorrhizospheric) may have enhanced soil respiration at the natural forest stand due to higher energy demand by increasing the autotrophic respiration (Ma *et al*., 2004).

In addition, higher soil microbial biomass (the sum of all PLFAs) at the natural forest stand is suggestive of fuelling of heterotrophic activity by higher substrate supply, such as above ground litter, below ground litter (fine root turnover) and rhizodeposition, or both supply in combination. Soil CO_2 efflux rates followed changes in the precipitation and the resulting changes in soil moisture content. When the weekly data were collated into seasonal patterns, soil CO_2 efflux was the highest for the rainy season but the lowest for the dry season (Fig. 3).

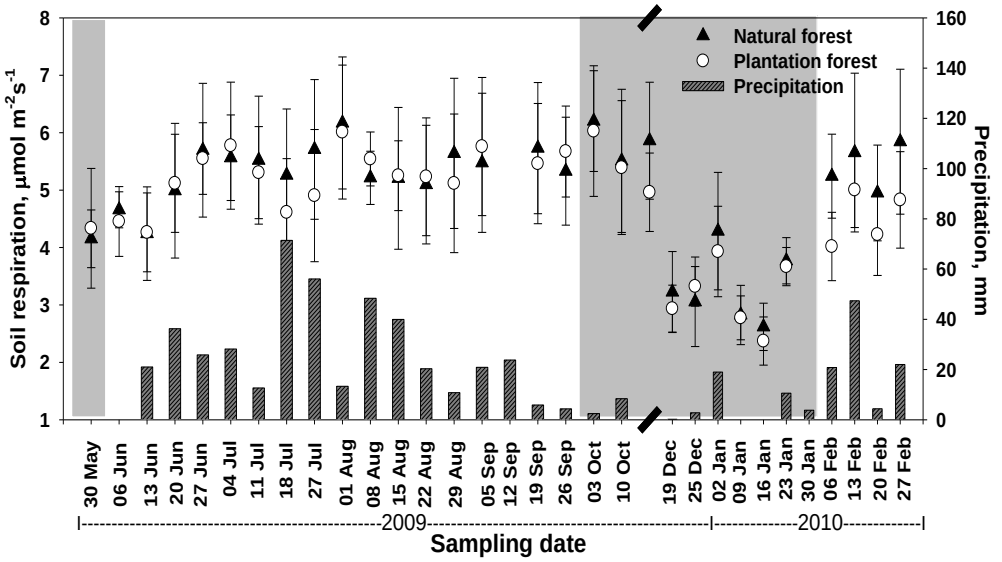

Fig. 3. Patterns of soil respiration rate and precipitation across the measurement campaign (data missed from 19 October to 19 December resulted from instrument failure). Periods with light grey background indicate dry periods as calculated by the approaches of Gibbs and Maher (1967).

The relationship between soil respiration and soil environmental variables was more influenced by soil moisture than soil temperature (Fig. 4). When fitted into a Gaussian function defined by three parameters, soil moisture explained about 44.8% and 42.5% of the variation in soil respiration in natural and plantation forest stands, respectively (Fig. 4). Soil respiration rates steadily increased with increasing volumetric soil water content up to a certain threshold level, after exceeding this threshold values it declined (Fig. 4). It is interesting to note that the optimal volumetric soil moisture at which soil CO_2 efflux measured the highest, was greater for the plantation stand (29.2%) than the natural forest (24.5%). This result indicates that *C. lusitanica* was more moisture tolerant than the natural vegetation, which

could probably relate with the shallow root system with large fine root biomass that enables it to utilize water from the topsoil effectively (Fritzsche *et al*., 2006).

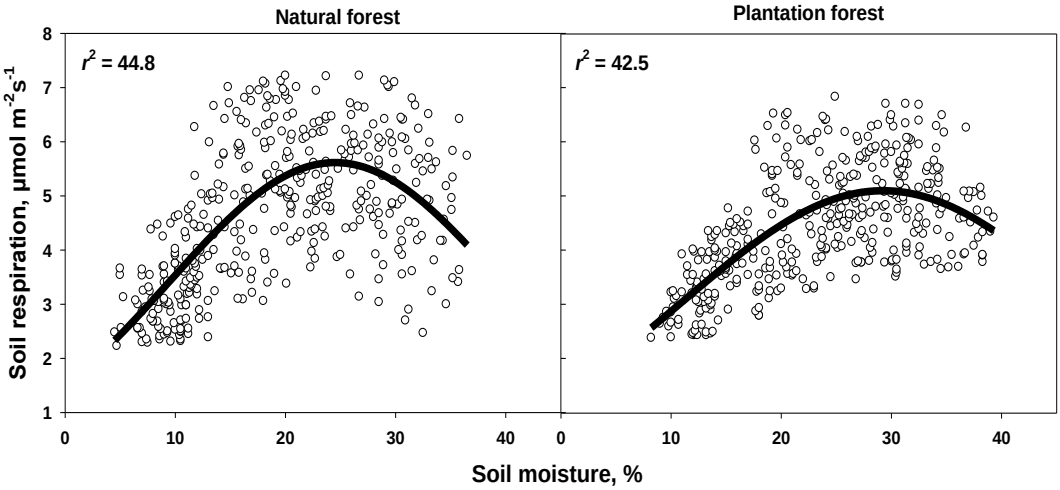

Fig. 4. Relationship between soil respiration rate (SR) and volumetric soil water content (SM) at 0.06 m soil depth. The parameters of the Gaussian function SR=a*exp(-0.5*((SM-x_0)/b)^2) are: a=5.61, b=15. 80, x_0=24.49 (natural forest); a=5.10, b=17.99, x_0=29.25 (plantation forest).

In tropical forest ecosystems, several studies have demonstrated that moisture content is an important factor driving soil respiration where the majority of biological processes coincide with moisture dynamics (e.g., Davidson *et al*., 2000; Hashimoto *et al*., 2004; Epron *et al*., 2006). In this regard, there is a wealth of information that microbial activity is restricted when soils contain too much or too little moisture (Herron *et al*., 2009 and references cited therein). Excess soil moisture may limit soil respiration by limiting oxygen availability due to reduced air diffusion for decomposition and root maintenance and growth, and thus CO_2 diffusivity (Linn and Doran, 1984; Janssens and Pilegaard, 2003; Adachi *et al*., 2006). This may be particularly the case for soil type in the present study where water molecules could be adsorbed onto the surface and interlayer of clay minerals. Low soil water content (drought) on the other hand may also limit soil respiration by having negative impact on soil microbial communities and plant root activities (Hashimoto *et al*., 2004; Epron *et al*., 2006).

The influence of soil temperature on the overall variation of soil CO_2 efflux was very weak; compared to that of soil moisture. Comparing the two forest types, a minor positive (r^2 = 0.12; $p<0.01$) exponential relationship between soil respiration and soil temperature was observed under the plantation

forest stand. Because of the small variation in soil temperature, no clear relationship was found under the natural forest stand (data not shown). Generally, weak correlation of soil temperature with soil respiration is also a feature widely reported in other tropical forest ecosystem studies where most of the year soil temperature difference in the forest floor remained negligible (Wanyama *et al*., 2019). In times of dry period where soil temperature was higher, soil moisture became a limiting factor by affecting soil respiration negatively. This result further highlights the importance of soil moisture in determining rate of C flux from soils which is also supported by the previous studies in tropical forest ecosystem (e.g., Davidson *et al*., 2000; Hashimoto *et al*., 2004; Nsabimana *et al*., 2009). Unlike tropical forest ecosystem, temperature represents the most limiting abiotic factor that regulates soil CO_2 efflux in boreal and temperate forest ecosystems (Shibistova *et al*., 2002).

CONCLUSION

Soil microbial biomass represents an important labile pool of nutrients and carbon. Unlike total organic C, microbial biomass C responds quickly to management changes. Microbial biomass was significantly high in the organic layers and decreased gradually with the depth of the layers. Despite comparable soil organic C, and total N, this study demonstrated that soil microbial biomass was substantially decreased under *Cupressus* plantations. This result implies that change in vegetation types can alter soil microbial biomass with corresponding consequences for ecosystem C balance. On the other hand, the close relationship between soil CO_2 efflux and soil moisture is an important implication when evaluating soil respiration data at the study site. The effect of soil moisture must always be taken into consideration as an important explanation of changes in soil CO_2 efflux. Together with supporting evidence from similar forest ecosystem, this research highlights that carbon mineralization could be shaped by the forestry management practices and ongoing climate change, typically change in precipitation pattern. Therefore, greater thought must be given to the ways in which new plantation forests are established within the landscape in order to maximize their ecological and economic benefits.

ACKNOWLEDGEMENTS

Funding for this research was provided by Deutsche Forschungsgesellschaft (DFG).

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