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Emission of CO_2 from the soil and immobilization of carbon in microbes in a subtropical mixed oak forest ecosystem, Manipur, Northeast India

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Emission of CO_2 from the soil and immobilization of carbon in the microbes were studied in two forest stands of a subtropical mixed oak forest located at Langol hills near Imphal city, Manipur (24°45′N lat. and 93°55′E long.) at an altitude ranging from 780 to 910 m amsl, using alkali absorption method and chloroform fumigation extraction method. The CO_2 emission rate was lowest during the winter season (149.00 and 138.49 mg CO_2 m^{-2} h^{-1}) and highest during the rainy season (250.94 and 220.48 mg CO_2 m^{-2} h^{-1}) in both the forest stands. The immobilization of carbon in the microbes was maximum during the rainy season (1182.6 and 740.73 μg g^{-1}) followed by summer (738.32 and 392.92 μg g^{-1}), and minimum during the winter season (465.14 and 382.58 μg g^{-1}) across the two stands. Out of the total soil organic carbon, maximum immobilization of microbial C occurs in the rainy season (2.7%) and minimum in the summer season (1.2%). Thus, emission of CO_2 from the soil and immobilization of carbon in the microbes are strongly influenced by the seasons.

Keywords: Carbon, immobilization, microbial biomass, oak forest.

SOIL respiration consists of autotrophic root respiration as well as heterotrophic respiration associated with the decomposition of litter, roots and soil organic matter. Hanson *et al.*[1] reported that root respiration contributed 10–90% of total *in situ* soil respiration depending on vegetation type and season of the year. Soil surface carbon dioxide (CO_2) flux, i.e. soil respiration exceeds all other terrestrial atmospheric carbon exchanges with the exception of gross photosynthesis[2]. Almost 10% of CO_2 from the atmosphere passes through the soil each year, which is more than ten times the CO_2 released from fossil-fuel combustion[3]. Due to the magnitude of this high soil-to-atmosphere CO_2 flux and the large pool of potentially mineralizable C in the soils, any increase in soil CO_2 emissions in response to climate change has the potential to exacerbate increasing atmospheric CO_2 levels and to provide a positive feedback to global warming[2–4]. Therefore, identifying the environmental factors that control soil CO_2 emissions and their effects on emissions rates is a necessary step in assessing the potential impacts of

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environmental change. Seasonal changes in soil microclimate play an important role in defining seasonal differences in soil CO_2 emissions within sites, and climatic differences generate different soil respiration rates among distant sites. The microbes also immobilize organic carbon present in the soil as microbial biomass. The microbial biomass accumulates nutrients which are released in the soil and is an important parameter linking the plants to the soil. The ratio of microbial biomass carbon to total organic carbon in the soil might serve as a quantitative indicator of carbon dynamics in the soil.

Limited information is available on the annual soil CO_2 flux[5] and microbial biomass carbon[6] from the northeastern region of India. Therefore, the present study was undertaken to evaluate (i) the monthly and seasonal soil CO_2 flux and soil microbial biomass carbon; (ii) the effect of abiotic factors on soil respiration rate and microbial biomass carbon, and (iii) the relationship between rate of soil respiration and microbial biomass carbon in a subtropical mixed oak forest ecosystem of Manipur.

The study site is located at 24°45′N lat. and 93°55′E long. in Langol hills, at a distance of 7 km from Imphal city, at an altitude ranging from 780 to 910 m amsl. Climate of the area is monsoonic with warm moist summer (March to May), monsoon (June to October) and winter (November to February) seasons. The mean monthly maximum temperature ranges from 24.3°C to 32.7°C and the mean monthly minimum temperature ranges from 3.2°C to 21.1°C. Average annual rainfall of the area is 1089.7 mm, with 68–70% of the rains occurring during the monsoon season. The study was conducted in two experimental forest stands. Forest stand I dominated by *Quercus serrata* and *Schima wallichi* is exposed to the sun for a short period during the morning hours, while forest stand II dominated by *Q. serrata* and *Lithocarpus dealbata* is exposed to the sun directly throughout the day.

Soil texture was analysed using the pipette method[7]. Soil temperature was determined using a soil thermometer and soil moisture was determined by the gravimetric method. Soil pH (1 : 5 water suspensions) was determined by a pH meter (Systronics). Soil organic C, total N and total P were determined using the methods of Anderson and Ingram[8], Bremner and Mulvaney[9] and Sparling *et al.*[10] respectively.

Soil respiration rate was measured by alkali absorption method[11] using open-ended cylinders of 13 cm diameter and 25 cm height, which were inserted into soil up to 15 cm depth. Six cylinders were randomly placed in each of the forest stands. All the vegetation was removed from the cylinder. Next, 50 ml of 0.25 N NaOH solution was kept in each cylinder and made airtight. After 24 h the alkali was titrated with 0.25 N HCl solution using phenolphthalein indicator. CO_2 absorbed from the soil was calculated as follows[8]:

$$\text{mg } CO_2 = V \times N \times 22$$

where V is the volume of the acid and N its normality.

Microbial biomass carbon was determined by fumigation and extraction method[8]. For the estimation of microbial biomass three soil samples were collected randomly from the upper layer 0–10 cm in depth from each of the forest stands I and II. The soil samples were sieved (<2 mm) to remove coarse roots, stones and plant debris and were kept at room temperature for a day. Microbial biomass carbon was determined by modified Walkley Black method and calculated as follows[12]:

Microbial biomass C = $K_{EC} \times 2.64$, where K_{EC} is the difference between C extracted from fumigated and unfumigated soils. Linear regression, multivariate ANOVA was analysed using STATISTICA.

The soil is silt loamy with 51.6% sand, 13.3% clay and 22.7% silt in forest stand I and 61.4% sand, 14.8% clay and 30.7% silt in forest stand II. It is acidic in nature, the parent material being derived from shale and sandstone. The soil moisture ranged from 24.74 to 28.34%, soil temperature from 16.83°C to 17.08°C and soil pH from 4.2 to 6.1. Soil organic carbon ranged from 2.6 to 4.4%, soil total N from 0.33 to 0.54%, total P from 0.042 to 0.082% and bulk density from 1.38 to 1.46 g cm^{-3}. C/N ratio was between 7.0 and 8.7 across the two stands (Table 1).

In forest stand I, soil CO_2 emission ranged from 120.26 to 324.47 mg CO_2 m^{-2} h^{-1} and in forest stand II it ranged from 112.12 to 267.67 mg CO_2 m^{-2} h^{-1} in different months throughout the year. Minimum soil CO_2 emission rates were recorded in March and then consistently increased till August, attaining a maximum value and thereafter decreasing till December (Figure 1). Seasonally, maximum soil respiration rate was recorded during the rainy season, followed by summer and winter seasons respectively.

Table 1. Abiotic variables and physico-chemical characteristics of soils in forest stands I and II

Abiotic variables	Stand I	Stand II
Soil temperature (°C)	16.83	17.08
Soil moisture (%)	28.34	24.74
Relative humidity (%)	73.59	73.59
Mean air temperature (°C)	22.43	22.43
Rainfall (mm)	137.48	137.48
Soil physico-chemical characteristics		
Texture		
Sand (%)	51.6	61.4
Silt (%)	30.7	22.7
Clay (%)	14.8	13.3
Bulk density (g cm^{-3})	1.38 ± 0.32	1.46 ± 0.27
Soil pH	4.2–5.8	4.5–6.1
Soil organic C (%)	2.75–4.4	2.6–4.34
Soil total N (%)	0.39–0.54	0.33–0.50
Soil available P (%)	0.07–0.082	0.042–0.069
C : N	7.0–8.1	7.8–8.7

Table 2. Correlation coefficient (*r*) for the relationship of soil respiration rate and microbial biomass C with abiotic variables

	Forest stand I		Forest stand II	
Parameters	Forest soil CO_2 released	Microbial biomass C	Forest soil CO_2 released	Microbial biomass C
Soil moisture (%)	0.55*	0.79**	0.51*	0.85**
Soil temperature (°C)	0.82**	0.82**	0.83**	NS
Relative humidity (%)	0.56*	0.80**	0.56*	0.69*
Mean air temperature (°C)	0.72**	0.82**	0.87**	0.63
Rainfall	0.87**	0.79**	0.80*	NS

NS, Not significant; *$P < 0.05$; **$P < 0.01$.

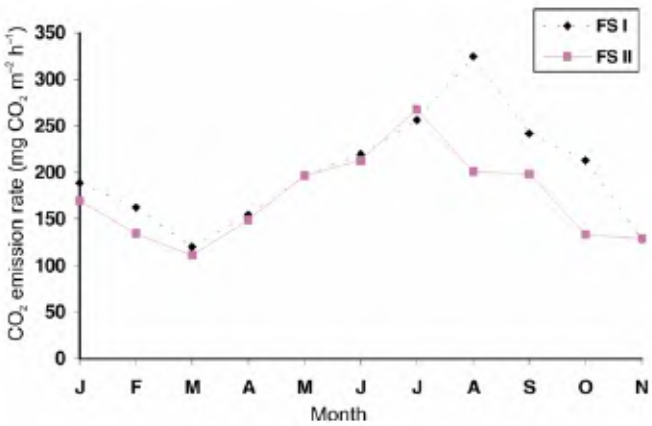

Figure 1. Monthly CO_2 emission rate in forest stands I and II.

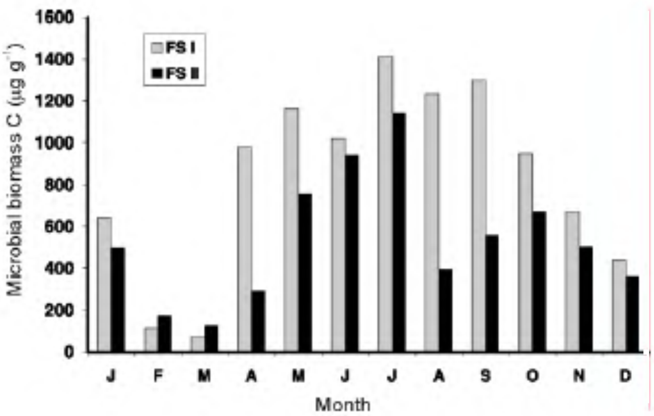

Figure 2. Monthly variation of soil microbial biomass C in forest stands I and II.

In forest stand I the microbial biomass C ranged from 71.1 to 1412.6 µg g^{-1}, while in forest stand II it ranged from 128.5 to 1141.5 µg g^{-1} in different months throughout the year (Figure 2). Maximum value of microbial biomass C was recorded during July and minimum during March in both the stands. The contribution of microbial biomass C to the total organic C varied from 1.7 to 2.7% and 1.2 to 1.7% in forest stands I and II respectively. Maximum value was recorded during the rainy season and minimum during the winter and summer seasons respectively, in both the stands. The multivariate ANOVA indicated a significant difference ($P < 0.001$) in soil respiration and microbial biomass carbon between the two stands. There was significant difference between the months and sites also ($P < 0.001$). The rate of soil respiration was significantly and positively correlated with the abiotic factors, i.e. soil moisture ($P < 0.01$), soil temperature ($P < 0.01$), relative humidity ($P < 0.01$), mean air temperature ($P < 0.01$), and rainfall ($P < 0.01$) in both the forest stands, while microbial biomass C was positively significant with the abiotic variables, except with soil temperature and rainfall in forest stand II (Table 2).

Low rate of CO_2 release from the soil in March in both the forest stands may be due to low moisture content of the soil, temperature and relative humidity, thereby inhibiting the microbial activity and decomposition[13], thus leading to low CO_2 evolution from the soil. However, emission of CO_2 was highest during the rainy months of July and August in forest stands I and II respectively. This coincides with a high soil and air temperature, high soil moisture and relative humidity favouring the activity of soil microflora and microfauna, leading to the enhancement in the decomposition of litter materials and thus contributing to more CO_2 release from the soil. The high rate of CO_2 released during the rainy season may be attributed to a congenial environment for the microorganisms dwelling in the soil and decomposing organic matter. Besides, the rate of decomposition of the litter material is also high during this period[14]. Decomposition of litter and microbial activity during the cool and dry winter season declined, thus leading to low emission of CO_2 from the soil. Several workers also reported a high soil respiration rate during the wet period, which is in conformity with our report[13,15,16]. Most of the studies on soil respiration reported maximum rate of soil respiration in spring or early summer[5,17]. However, our study is contradictory to the findings reporting a maximum rate during the rainy season. This may be due to the frequent periodic drying and wetting of the soil during the rainy season, enhancing the activity of the microbes and displacement of CO_2-enriched air in soil pores by infiltrating rainwater, thus contributing to an increase in CO_2 evolution. The coefficient of correlation (*r*) in the present study shows that soil temperature, mean air temperature and rainfall, soil moisture and relative humidity have a significant positive effect on soil respiration rate in both the forest stands. Adachi *et al.*[18] also reported significant seasonal differences in different tropical ecosystems. Several studies reported that temperature was the single most important variable for predicting the soil CO_2 flux[19,20]. Chapman and Thurlow[21] also reported that a rise in the mean annual temperature of 5°C could potentially increase CO_2 emission by a factor of 2–4.

Immobilization of microbial C was highest in the rainy season, which may be due to the availability of more decomposing plant debris during this season, as the microbial activity and decomposition rate were at a peak during this season. Further, the growth of fungi also increased due to high relative humidity, contributing to soil microbial biomass[22]. In dry tropical deciduous forest, peak value of microbial biomass C was reported in early spring or summer[23] and in subtropical humid forest, maximum value was reported in the winter season[6], which may be due to differences in litter quality and rainfall pattern. However, in the present study the low rate of microbial C immobilization in the winter season may be due to lesser activity of microorganisms and slow rate of decomposition of litter in the cool and dry period. According to Diaz-Ravina *et al.*[24], lack of water seems to limit the microbial biomass more than temperature, since lower microbial biomass contents were observed in the dry period than in the wet period. Several studies on microbial biomass reported a close relationship between soil moisture and microbial biomass[25], where a maximum value was obtained in the wet period and minimum in the dry period, which is in conformity with our study. Similar observations were also reported[25] in different ecosystems (forest, grasslands and arable soil of Greece). The contribution of microbial biomass C to total organic C was maximum during the rainy and minimum in the summer season, thereby indicating high rate of immobilization in the rainy season and a low rate in the summer season. The significant positive relation ($r = 0.76$; $P < 0.01$ and $r = 0.80$; $P < 0.01$) between soil CO_2 release rate and microbial biomass C in the forest stands shows that CO_2 flux from the soil is highly influenced by C immobilization in the microbial biomass.

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