B-6.3.1 Quantification of Soil Microbial Activity on Carbon Cycling in the Cool Temperate Forest

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Abstract This study aimed to model soil microbial decomposition activity, and quantify the carbon flow through soil microbial biomass. For this purpose, changes in the microbial biomass and correlations between temperature and metabolic quotient (MQ: respiration rate per unit of microbial biomass) were measured from April 1997 to March 1999 in two cool temperate deciduous forests. Using *in situ* soil temperature, CO_2 flux though the microbial biomass and CO_2 flux from the forest floor (microbial respiration + root respiration) based on past studies were estimated. The change of microbial biomass affects CO_2 flux through the microbial biomass, but the influence is much lower than that of temperature dynamics. The percentage of microbial CO_2 flux of total annual forest floor CO_2 flux was equivalent to past studies.

Key Words Microbial Biomass, Soil Respiration, Forest Ecosystem, Carbon Cycling

1. Introduction

Soil microorganisms play an essential role in the decomposition of organic material in forest ecosystems. Their function in carbon cycling and their capacity to serve as a relatively labile source of carbon in soils are well recognized[1]. Therefore, estimates of the pool size of microbial C and its decomposition (respiratory) activities are important for the analysis of carbon cycling mechanisms, but few studies have focused on microbial activity in forest ecosystem carbon cycling. In this study, we hypothesized that CO_2 released by microbial activity depends on microbial biomass and its temperature-dependent metabolic activity. We intended to clarify the contribution of microbial activity and biomass to CO_2 flux from the forest floor.

2. Materials and methods

2.1 Sites and soils

We selected sampling sites (20 m • 25 m) at Takayama Experimental Station of Gifu university, Institute of Basin Ecosystem, in Takayama, Gifu (Takayama) and National Grassland Research Institute, in Nishinasuno, Tochigi (Fujinita). Vegetation was dominated by birches (*Betura tauschii*) and oaks (*Quercus mongolica*) in Takayama and chestnuts (*Castanea crenata*) and oaks (*Quercus serrata*) in Fujinita. Soil texture was HC (Takayama) and LiC (Fujinita). Altitude, mean annual temperature and annual precipitation were 1400 m, 7.2℃ and 2439 mm, respectively, in Takayama and 310 m, 11.0℃ and 997 mm (Kuroisho AMeDAS 1996), respectively, in Fujinita.

2.2 Sampling

At each sampling site, to obtain the vertical distribution of microbial biomass, a soil profile was dug to a 80 cm depth and samples were collected from the A_0, 0-5 cm, 5-10 cm, 10-20 cm, 20-40 cm, 40-60 cm and 60-80 cm soil layers.

Next, to obtain the seasonal change of microbial biomass and its respiration rate, samples of the A_0, 0-5, 5-15, 15-35 cm (from Nov. 1997) soil layers were taken 9 times from

April 1997 to March 1999 at 12 points (6 points during snowing season or winter) at each sampling site.

Samples of the A0 layer were cut by kitchen knife and passed through a 8 mm sieve. Soil samples were passed through a 8mm sieve and fine roots were removed. Then samples were stored at 5℃ until analysis.

2.3 Microbial biomass and respiration rate

Microbial biomass in the stored samples were measured using the chloroform fumigation-extraction method according to Vance *et al.*[2] for soil samples, and with some modifications for A0 layer[3].

The respiratation rate of the samples was measured with a continuous air flow system using an infrared gas analyzer under dark conditions. Water content was adjusted to 55% of WHC for soil samples and 100% of WHC for the A0 layer. Respiration rate of each sample was measured at 25℃, 15℃ and 5℃ temperatures in order. Before measurement at each temperature, preincubation was carried out for 2 weeks. Subsamples were also incubated in the same way, and microbial biomass was measured when respiration rate was measured. Microbial respiration rate at each temperature was expressed as MQ (metabolic quotient: μg CO_2-C mg^{-1} biomass C h^{-1}). Through these measurements, hysteresis was not observed (data not shown).

Microbial biomass and respiration rate were measured triplicate and duplicate, respectively.

2.4 Soil temperature

To estimate CO_2 flux though microbial biomass, *in situ* soil temperature was measured at depths of 0, 5, 10, 40 cm from the soil surface.

2.5 Estimation of CO_2 flux though microbial biomass and CO_2 flux from forest floor

CO_2 flux though microbial biomass was estimated for each day as follows:

$$R = MB \cdot aEXP(bT) \quad [1]$$

R: respired CO_2 (mg CO_2-C m^{-2} h^{-1})
MB: microbial biomass (g m^{-2})
T: temperature
a, b: constant

We assumed that the microbial biomass was constant between half interval separately the sampling days. The formula [aEXP(bT)] is a regression curve of MQ temperature dependence. R was estimated in each layer (A0, 0-5 cm, 5-15 cm, 15-35 cm), then the total CO_2-respired by microbial biomass was estimated.

CO_2 flux from forest floor (microbial respiration + root respiration) was estimated for each day as follows:

Takayama	$SR(g\ CO_2\ m^{-2}\ day^{-1}) = 0.0012T^2+0.1748T+0.2544$	[2]	(Nishimura[5])
Fujinita	$SR(g\ CO_2\ m^{-2}\ h^{-1}) = 33.285 \cdot EXP(0.12916T)$	[3]	(Matsumoto[6])

SR: CO_2 flux from the forest floor
T: soil surface temperature

Formulas [2] and [3] were obtained from same study sites of ours.

Using these 3 formulas, annual CO_2 flux in 1998 was estimated and compared.

3. Results and Discussion

3.1 Vertical distribution and seasonal changes of microbial biomass

Microbial biomass existed mainly in the upper soil layer at both sampling sites (Fig. 1). Total microbial biomass per area basis up to a 80 cm depth was 312 and 152gC m^{-1} in Takayama and Fujinita, respectively. The microbial biomass in the A0 layer and soil up to a 35 cm depth corresponded to approximately 80% of total microbial biomass up to a 80 cm depth.

Seasonal changes in the microbial biomass are shown in Fig. 2. There was a significant seasonal change in microbial biomass in each layer at each site. In Takayama, microbial biomass in soil became larger in winter, then decreased to autam. In Fujinita, there was no consistent tendency: rather, the biomass seemed relatively constant in the 0-5 and 5-15 cm layers. In the A0 layers at both sites, no consistent tendency was observed.

Ross[7] reported seasonal changes in the microbial biomass under pastures, though the cause of fluctuations were not explained. Seto and Yanagitani[8] reported positive correlations between water-soluble organic carbon content and respiration rates in soil and carbohydrates in water-soluble organic carbon are easily utilized by microbes[9]. These components may affect the size of the microbial biomass. In this study, there was no positive correlation between microbial biomass content and 0.5M K_2SO_4 extractable carbon. This result supports the result of Walters and Jorgensen[10]. West and Sparling[11] suggested that soil water content affects the size of the microbial biomass. In Takayama, the A0 layer and soil did not freeze when covered by snow in the winter and the A0 layer and soil were very wet. This wet condition might make the microbial biomass larger.

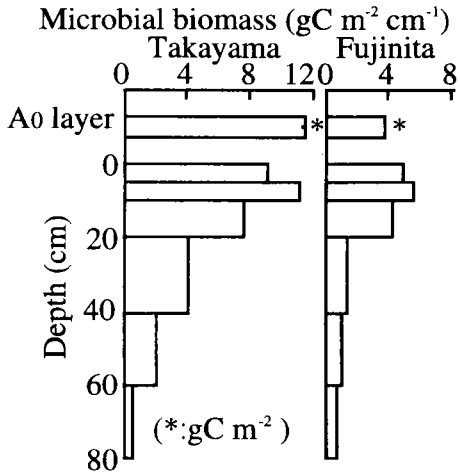


Fig.1 Vertical distribution of microbial biomass

The microbial biomass content of the A0 layer correlated with carbohydrate content[3], though the quantity of the A0 layer also affects microbial biomass on per-area basis. Therefore, a consistent tendency was not observed.

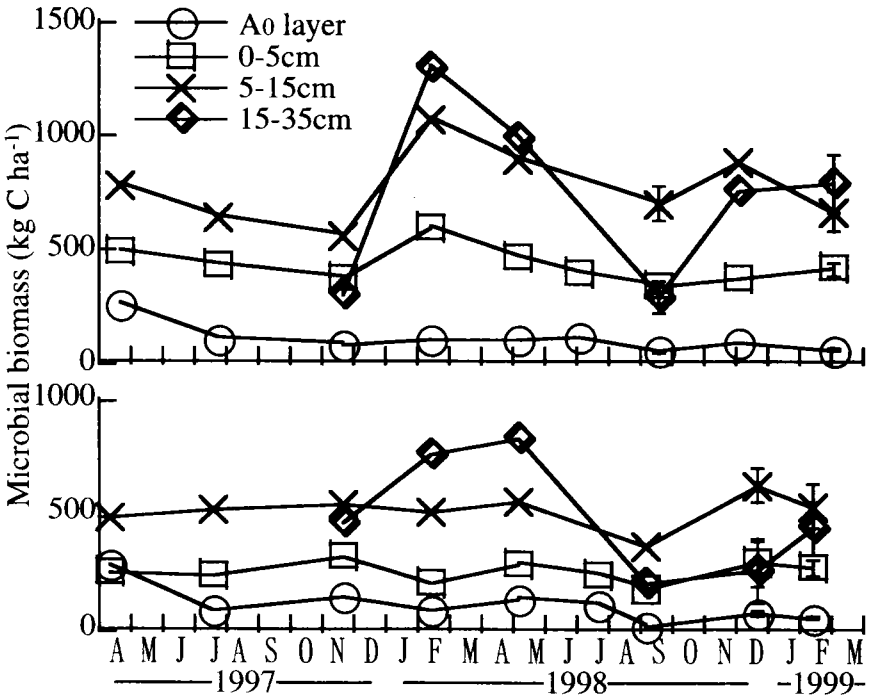


Fig. 2 Dynamics of microbial biomass during sampling priond

3.2 MQ temperature dependence

The regression curve of MQ on temperature did not differ significantly among sampling times. Therefore, a single regression curve adapted to MQ temperature dependence was expressed for each sample, except for one A0 layer sample collected in April 1997 in Fujinita (Fig. 3).

3.3 Estimation of CO_2 flux through microbial biomass and CO_2 flux from forest floor

3.3.1 Takayama

CO_2 flux through microbial biomass was estimated with Formula [1] using microbial biomass and soil temperature measured in 1998 in each soil layer. Total CO_2 flux through microbial biomass in 1998 was estimated to be 3.73t ha^{-1}. The contribution of the A0 layer was 28% while the remaining 72% was respired from the soil. CO_2 flux from the forest floor was estimated using Formula [2] and the annual CO_2 flux was estimated to be 7.45t ha^{-1}. Therefore, the contribution of the microbial respiration on CO_2 flux from the forest floor was 48%. This

percentage varied from 32% to 69% according the month (Fig. 4). This is due to the seasonal change in microbial biomass.

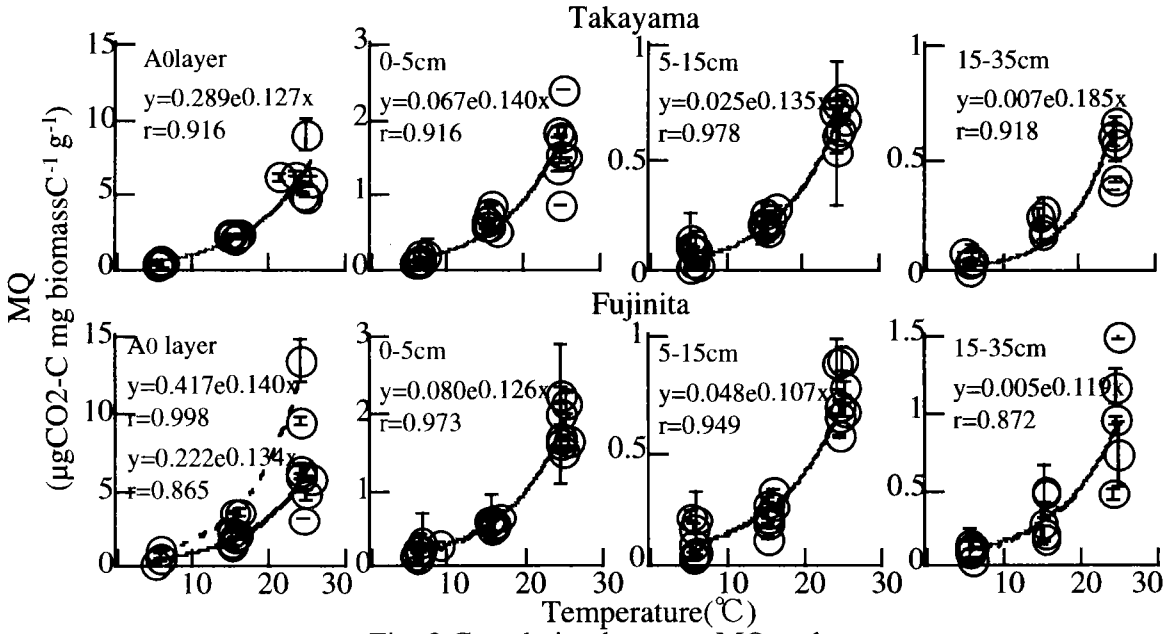


Fig. 3 Correlation between MQ and temperature

To clarify the contribution of changes in microbial biomass and temperature on CO_2 flux through microbial biomass, multiple regression analysis was carried out (Table 1). The results show that the dynamics of soil temperature affect more than the changes in microbial biomass.

3.3.2 Fujinita

Total CO_2 flux through microbial biomass in 1998 was estimated to be 4.52t ha^{-1}. The contribution of the A_0 layer was 28% while the remaining 72% was respired from the soil. CO_2 flux from the forest floor was estimated using Formula [3] and the annual CO_2 flux was estimated to be 5.86t ha^{-1}. Therefore, the contribution of microbial respiration on CO_2 flux from the forest floor is 72%. This percentage ranged from 53% to 129% according the month (Fig. 5). In April to July, a larger microbial biomass in the 15-35 cm layer increased the CO_2 flux through

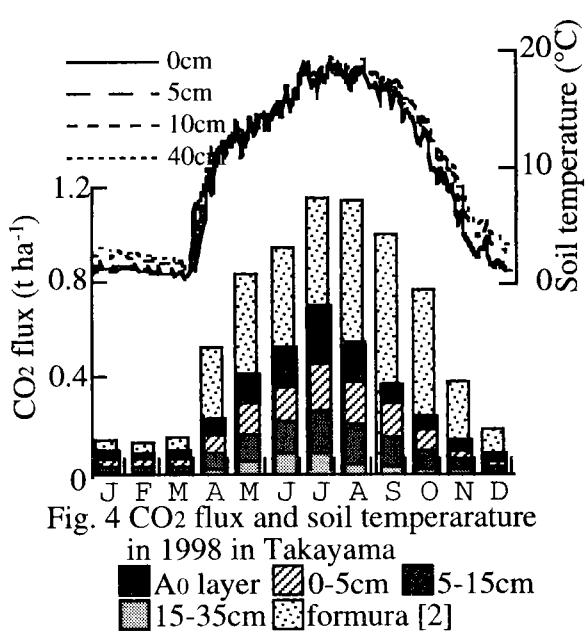


Fig. 4 CO_2 flux and soil temperarature in 1998 in Takayama

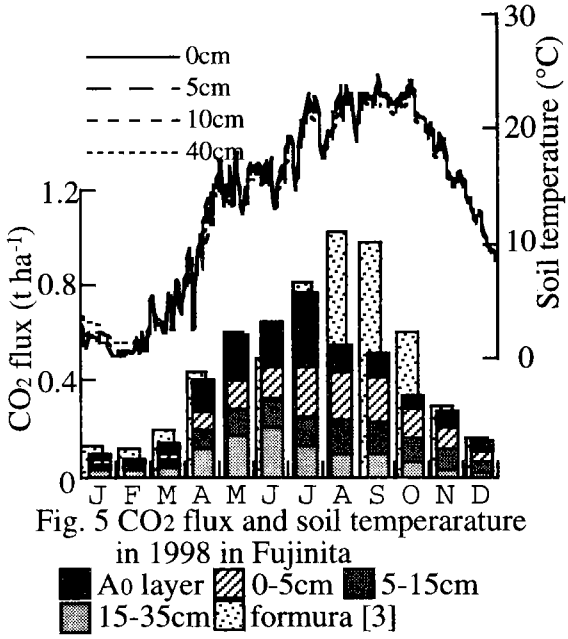
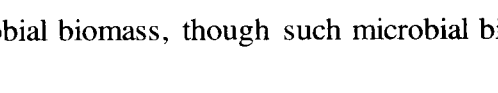
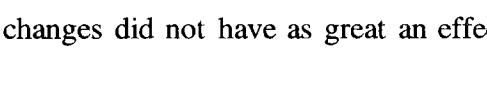


Fig. 5 CO_2 flux and soil temperarature in 1998 in Fujinita





microbial biomass, though such microbial biomass changes did not have as great an effect as

temperature dynamics (Table 1).

Table 1. Contribution of microbial biomass and soil temperature to CO_2 flux

		A0 Layer	0-5cm	5-15cm	15-35cm
Takayama	Temperature	88.93%	95.19%	97.86%	77.27%
	Biomass	7.58%	37.54%	38.09%	27.07%
Fujinita	Temperature	68.60%	96.46%	93.96%	47.72%
	Biomass	0.71%	0.01%	14.63%	4.73%

3.4 General discussion

Although the microbial biomass in Fujinita was approximatery half that in Takayama on a per-area basis, CO_2 flux through microbial biomass in Fujinita was larger than that in Takayama. This was due to the higher soil temperature and short snowfall period.

Through field research in 3 different types of manure forests, Nakane[12] shows that half of the CO_2 flux from the forest floor came from root respiration while the other half came from the degradation of litter and soil organic matter. On the other hand, Tate *et al.* [13] show that root respiration accounted for 23% while the other part came from the degradation of organic matter through an *in vitro* experiment. Kelting *et al.* [14] point out that CO_2 respired from the forest floor by root, rhizosphere and root-free soil accounted for 32%, 20% and 48%, respectively, through *in situ* research. On an annual CO_2 flux basis, our results, on the contribution of the CO_2 flux by microbial activity measured *in vitro* experiments from the CO_2 flux from the forest floor, agreed with those of the past studies.

Nakane[12], Tate *et al.*[13] and Ross and Tate[1] have stated that CO_2 flux from the A0 layer is as much as from soil. In our study, CO_2 from the A0 layer occupied 28% of the total CO_2 flux at both sites. This rate is smaller than past studies.

In our study, respiration was measured under ambient CO_2 concentration conditions. The authers believe that CO_2 concentration of the air in *in situ* soil is higher than that of ambient air. Koizumi *et al.*[15] and Santruckoba and Simek[16] point out that higher CO_2 concentrations suppress microbial respiration, and the degree of suppression differes among soils. Therefore, there is a possibility that our estimation of CO_2 flux through microbial biomass may be and overestimate. Incorporation of this factor would be needed for more accurate estimation of the CO_2 flux through the microbial biomass.

4. Acknowledgments

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