

## Examination of the method for measuring soil respiration in cultivated land: Effect of carbon dioxide concentration on soil respiration

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An acceleration of soil respiration with decreasing CO<sub>2</sub> concentration was suggested in the field measurements. The result supports that obtained in laboratory experiments in our previous study. The CO<sub>2</sub> concentrations in a chamber of the alkali absorption method (the AA-method) were about 150–250 parts/10<sup>6</sup> lower than that in the atmosphere (about 350 parts/10<sup>6</sup>), while those observed in the open-flow IRGA method (the OF-method) were nearly equal to the soil surface CO<sub>2</sub> levels. The AA-method at such low CO<sub>2</sub> levels in the chamber appears to overestimate the soil respiration. Our results showed that the rates obtained by the AA-method were about twice as large as those by the OF-method in field and laboratory measurements. This finding has important consequences with respect to the validity of the existing data obtained by the AA-method and the estimation of changes in the terrestrial carbon flow with elevated CO<sub>2</sub> concentrations.

**Key words:** alkali absorption method; CO<sub>2</sub> concentration; open-flow IRGA method; soil respiration.

### INTRODUCTION

Watson *et al.* (1990) estimated that the total amount of organic carbon stored in biomass and soil organic matter in terrestrial ecosystems in the world is 2050 Pg, nearly three times as much as that in the atmosphere. Since 70% of this organic carbon exists in soil, soil plays a great role in global carbon cycles.

In studies of carbon cycles in terrestrial ecosystems, the rate of CO<sub>2</sub> evolution from the soil provides a useful parameter of biological activity in the soil. Thus the measurement of the CO<sub>2</sub> evolution from soil has been carried out by many researchers in forests and grasslands. The most accurate estimates of the CO<sub>2</sub> evolution can be obtained by direct measurements of soil respiration rates. Methods for measuring soil respiration in the field can be classi-

fied into two groups; an open-flow infra-red gas analyzer (IRGA) method (OF-method) and an alkali absorption method (AA-method). The OF-method has not been used very much in forests (Reiners 1968; Edwards & Sollins 1973) or grasslands (Kucera & Kirkham 1971; Mathes & Schriefer 1984). On the other hand, the AA-method is so simple that it has been used widely to measure *in situ* CO<sub>2</sub> release in forest ecosystems (Witkamp 1966; Schulze 1967; Witkamp 1969; Kirita 1971b; Nakane 1975; Edwards & Ross-Todd 1983; Carlyle & U Ba Than 1988; Rout & Gupta 1989; Maggs & Hewett 1990), in grasslands (Kucera & Kirkham 1971; Wildung *et al.* 1975; Gupta & Singh 1981) and in cultivated areas (Buyanovsky *et al.* 1986; Singh *et al.* 1988).

It is well known that the soil respiration rate depends particularly upon soil temperature (Kucera & Kirkham 1971), water content (Rout & Gupta 1989), and a combination of the two factors (Witkamp 1966; Reiners 1968; Wildung *et al.* 1975; Kowalenko *et al.* 1978; Gupta & Singh 1981;

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Mathes & Schriefer 1984; Buyanovsky *et al.* 1986; Carlyle & Than 1988). In a previous study (Koizumi *et al.* 1991), however, we found that soil microbial activity, which is one of the main sources of soil respiration, was suppressed by increasing CO<sub>2</sub> concentration under laboratory conditions. The soil microbial respiration rate at 150 parts/10<sup>6</sup> CO<sub>2</sub> was about 1.6 times that at 350 parts/10<sup>6</sup> (a normal atmospheric CO<sub>2</sub> concentration).

The purpose of this study is to compare the soil respiration rates measured by the OF-method and the AA-method and to investigate the effect of CO<sub>2</sub> concentration on the soil respiration rate. Effects of soil temperature and CO<sub>2</sub> concentration on soil respiration is also discussed based on the data obtained in the OF-method.

## METHODS

### Study site

The study site is located on experimental fields of the National Institute of Agro-Environmental Sciences at Tsukuba Science City, Ibaraki Prefecture in central Japan (36°01' N, 140°07' E). The topsoil is volcanic ash soil (Kuroboku soil). The experimental fields (barley and corn) have been supplied with about 400 g organic carbon m<sup>-2</sup> (stubbles of barley or corn) every year since 1989. Carbon content of the soil (0–70 cm deep) was about 21.7 kg m<sup>-2</sup> in 1989.

### Measurements of soil respiration

#### *Open-flow IRGA method (OF-method)*

A flow diagram of the OF-method is illustrated in Fig. 1. This system is the same as that described in the previous study (Koizumi *et al.* 1991) except for the addition of an air pump to equilibrate the air pressure between inside and outside the chamber. The atmospheric CO<sub>2</sub> concentrations were measured every hour by changing the pass route using an electromagnetic valve. The air flow rate in the chamber was regulated at 1.0 L min<sup>-1</sup> and the flow speed was 5.9 cm min<sup>-1</sup>. The bottom edge of this chamber, 21.6 cm in diameter and 13 cm in height, was inserted into the soil to a depth of 3 cm. An

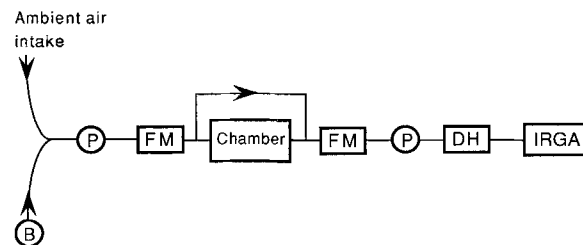


Fig. 1. Flow diagram of the open-flow infra-red gas analyzer method (OF-method) for determination of CO<sub>2</sub> evolution from soil. B: air balloon; P: air pump; FM: flow meter; DH: dehumidifier, electric cooler (COS, Model GC-11) for field experiment or perma pure dryer (Perma Pure Product Inc., Model ZBJ) for laboratory experiment; IRGA: infra-red gas analyzer. Soil respiration was determined from the difference of CO<sub>2</sub> concentrations between input and output of the chamber. Ambient air was taken at a height of 2 m above soil surface.

infra-red gas analyzer (Fuji Electric, Model ZFP5 or ZRC) was used to measure CO<sub>2</sub> concentrations of the air.

#### *Alkali absorption method (AA-method)*

The AA-method developed by Kirita (1971a) was used in the present study. The cylindrical chamber was 12.6 cm in diameter and 23 cm in height, and its bottom edge was pushed into the soil 5 cm in depth. A sponge containing 25.0 mL of 1.0 N KOH solution was placed on a wire holder (12 cm above soil surface) in a chamber immediately before the start of the measurement. At collection, the alkali solution in the sponge was squeezed and stored in a plastic vial and carried to the laboratory. The solution of 5.0 mL was titrated with 0.1 N HCl using phenolphthalein and methyl orange as indicators.

In laboratory experiments, CO<sub>2</sub> concentrations in three chambers of the AA-method were monitored at 1 h intervals during soil respiration measurement. The chamber was equipped with two rubber stoppers at 1 cm and 10 cm high above the soil surface. Air was sampled by a microsyringe from the rubber stoppers in 2.0 mL amounts. The sampled air was inserted into a polyvinyl tube in which CO<sub>2</sub>-free air streamed. An infra-red gas analyzer (IRGA) (Fuji Electric, Model ZRC) was used to measure the CO<sub>2</sub> concentrations of the air.

## Experiments

### *Comparison between the OF-method and the AA-method*

Both the OF-method and the AA-method were applied to measure soil respiration rates in the field and under laboratory conditions.

In the field experiments, soil respiration rates were measured for 24 h in a barley field in May, and in a corn field in August 1990. There was one measurement plot for the OF-method and six for the AA-method. Soil surface temperature was monitored both inside and outside the chamber with copper-constantan thermocouples.

In the laboratory, comparisons between the two methods were carried out at 25 °C. One chamber for the OF-method and four chambers for the AA-method were set up. A 'Kuroboku' soil sample taken from the experimental field was screened through a 3 mm mesh sieve, mixed thoroughly and put in a polyvinyl chloride container (63 cm length, 38 cm width, and 15 cm depth). Soil moisture content was maintained at about 40% on dry weight basis by sprinkling water every day. The measuring period of soil respiration ranged from 6 to 8 h. The ambient air stored in a balloon was passed through the chamber in the OF-method.

## RESULTS

Results of simultaneous field measurements of soil respiration rate obtained by the OF-method and the AA-method are shown in Fig. 2. Daily changes of soil temperature were from 15 to 25 °C in May and 25 to 35 °C in August. The ambient CO<sub>2</sub> concentrations ranged from 350 to 450 parts/10<sup>6</sup> in May and 300 to 400 parts/10<sup>6</sup> in August. The soil respiration rates from the AA-method are the averages of the rates during 24 h because it provides the integrated values. On the other hand, the OF-method can detect the daily patterns of soil respiration rates. The soil respiration rates obtained by the AA-method were 998 and 1143 mg CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup> in May and August, respectively, while the mean rates of those by the OF-method were 484 and 388 mg CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup>, respectively. The rates obtained by the AA-method were about 2.1 times as large as that by the OF-method in May, and about 2.9 times in August.

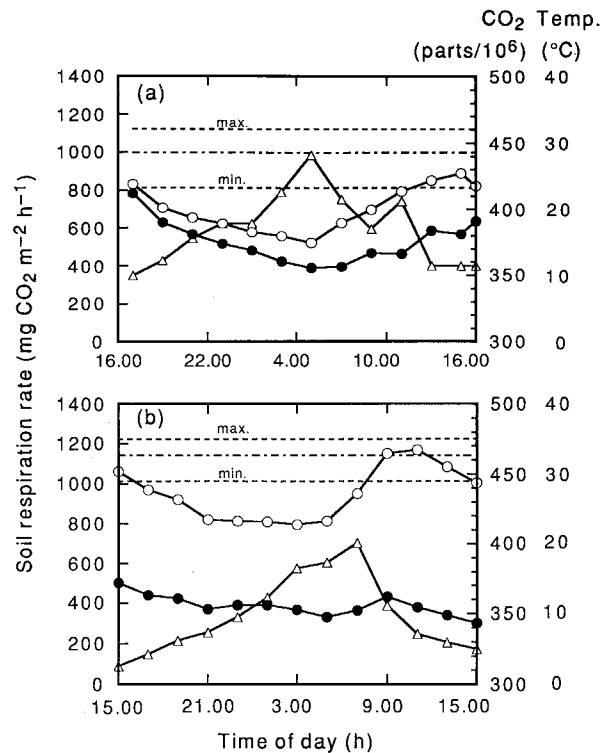
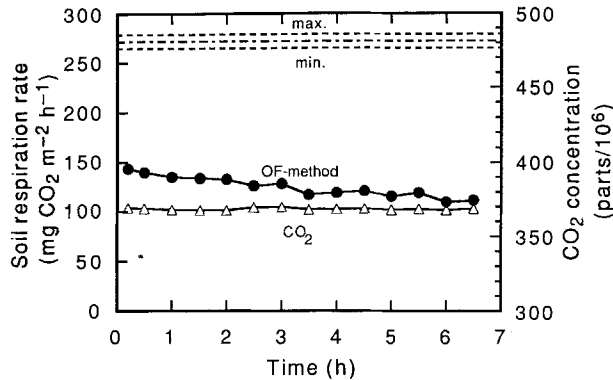


Fig. 2. Daily changes in soil respiration rates measured simultaneously by the OF-method (open-flow IRGA method) and the AA-method (alkali absorption method). Parts (a) and (b) refer to the measurements on 10–11 May (barley field) and 17–18 August (corn field) 1990, respectively. (●) Values obtained by the OF-method; (○) soil surface temperature in the chamber of the OF-method; (△) CO<sub>2</sub> concentrations of ventilated air that were taken at a height of 2 m above the soil surface. Values obtained by the AA-method are shown chain line (— · — ·; average) and dotted lines (---; max. and min.).

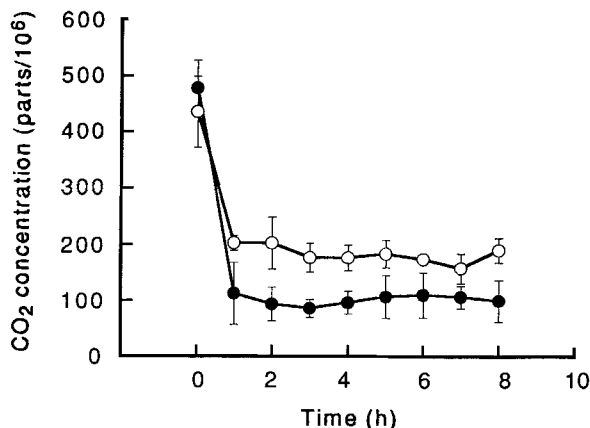
Figure 3 shows comparisons of soil respiration rates measured by the two methods in the laboratory at 25 °C. The CO<sub>2</sub> concentrations of the ventilated air remained at about 370 parts/10<sup>6</sup> because the ambient air was homogenized in a balloon. The rates obtained by the AA-method are the averages of the soil respiration rates during the measurement. Soil respiration rates obtained by the OF-method decreased slightly with time and did not show a daily change because the environmental factors (temperature and CO<sub>2</sub> concentration) were controlled at fixed levels in the laboratory experiment. The mean soil respiration rate obtained by the AA-method was 272 mg CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup>, while that by the OF-method was 124 mg CO<sub>2</sub> m<sup>-2</sup> h<sup>-1</sup>. Thus, the value from the AA-method was about 2.2 times as large as



**Fig. 3.** Comparisons of soil respiration rates obtained by the OF-method (open-flow IRGA method) and the AA-method (alkali absorption method) in the laboratory. Temperature was controlled at 25 °C during the experiment. (●) Values obtained by the OF-method; (△) CO<sub>2</sub> concentration of the ventilated air. Values obtained by the AA-method are also shown chain line (— · — ·; average) and dotted lines (---; max. and min.).

that from the OF-method. The difference between the two methods in the laboratory experiment was similar to those in the field experiments (cf. Fig. 2).

As shown in Fig. 4, the CO<sub>2</sub> concentrations in the chamber of the AA-method were about 400–500 parts/10<sup>6</sup> before the start of the experiment. The CO<sub>2</sub> levels dropped within 1 h to about 100 parts/10<sup>6</sup> at a height of 10 cm (beneath the sponge) and 200 parts/10<sup>6</sup> at 1 cm from the soil surface. These low CO<sub>2</sub> levels were kept throughout the experiment.



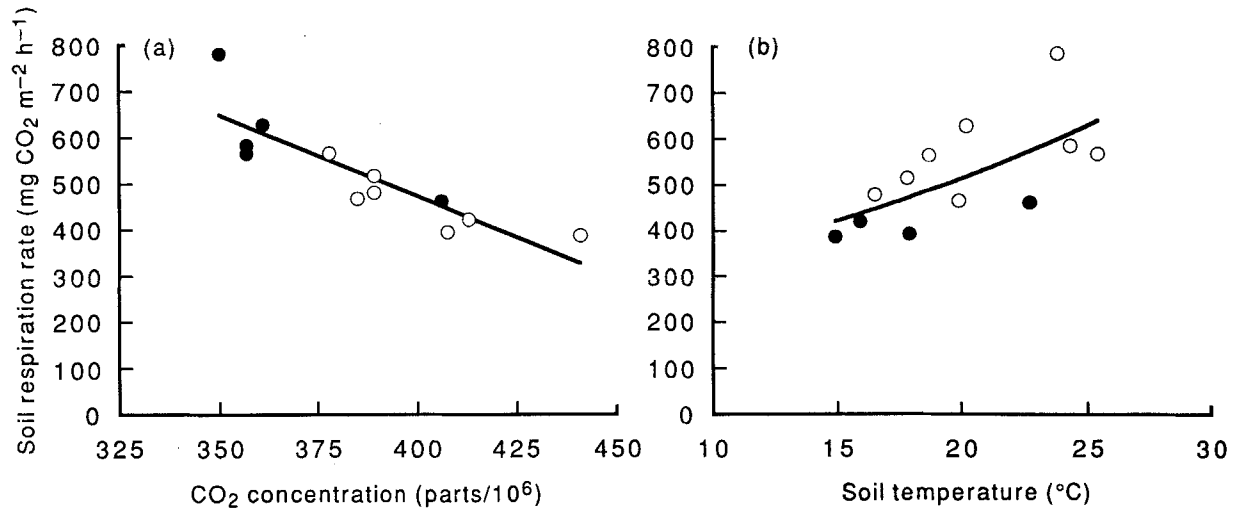
**Fig. 4.** Time course of mean CO<sub>2</sub> concentrations in the chamber of the AA-method (alkali absorption method). There are three chambers. Vertical bars indicate ± SD (●) At 10 cm high beneath CO<sub>2</sub> absorbent (sponge); (○) at 1 cm high from the soil surface.

The effect of environmental factors on soil respiration was considered using data obtained by the OF-method. As shown in Fig. 5 (cf. Fig. 2a), the soil respiration rate in May was correlated more closely with the ambient CO<sub>2</sub> concentration ( $r = 0.87$ ) than with the soil surface temperature ( $r = 0.64$ ). On the other hand, as shown in Fig. 6 (cf. Fig. 2b), the soil respiration rate in August was correlated better with the soil surface temperature ( $r = 0.88$ ) than with the CO<sub>2</sub> concentration ( $r = 0.78$ ).

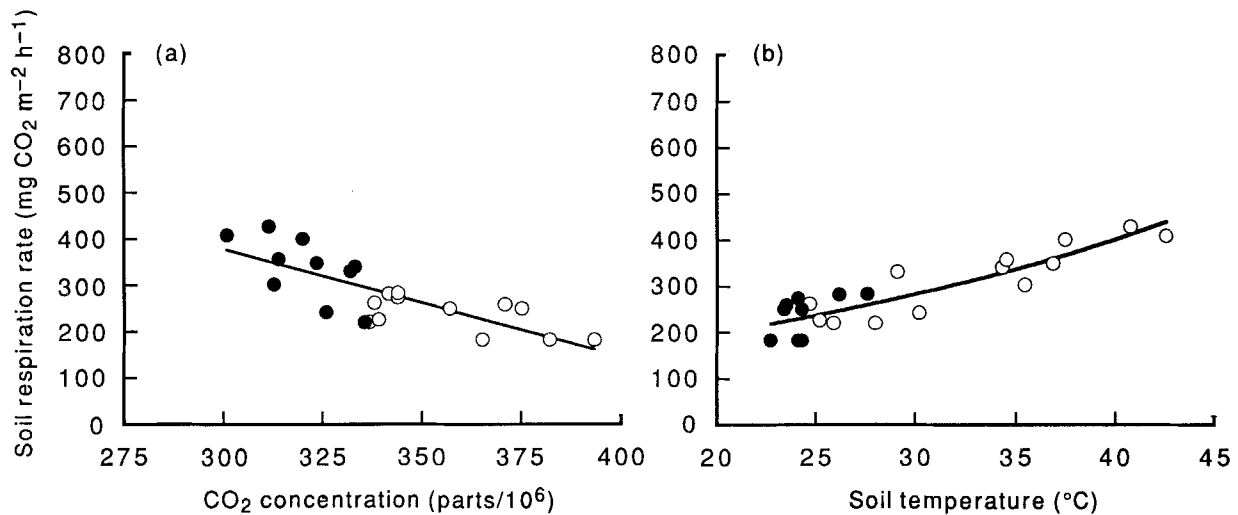
## DISCUSSION

The present study verified that the soil respiration rates obtained by the AA-method were always higher than those by the OF-method both in the field (Fig. 2) and laboratory experiments (Fig. 3). However, some researchers demonstrated that the rates by the AA-method were about 60% of those by the OF-method, in a grassland (Kucera & Kirkham 1971), and about 63–90% in a forest (Edwards & Sollins 1973). They suggested that the AA-method might be inadequate because the CO<sub>2</sub> absorption efficiency of the alkali solution decreased as it was neutralized, while the validity of the values determined by the OF-methods was open to question. On the other hand, other researchers reported that soil respiration might have been overestimated by the AA-method. Soil carbon loss estimated by the AA-method exceeded input of carbon through litter deposition in forests (Witkamp 1966; Kirita 1971b; Rout & Gupta 1989) and in a wheat field (Buyanovsky *et al.* 1986). Kirita (1971b) noted that some branches untrapped as a litter component could result in great differences (about 2.5 to 4.0 times) between the carbon loss and input in evergreen broad-leaf forest. But other authors considered that the differences (about 1.02 to 1.71 times) were attributed to root respiration (Witkamp 1966; Buyanovsky *et al.* 1986; Rout & Gupta 1989).

The rates of CO<sub>2</sub> evolution from soil depend upon both temperature and water content (Witkamp 1966; Reiners 1968; Wildung *et al.* 1975; Kowalenko *et al.* 1978; Gupta & Singh 1981; Mathes & Schriefer 1984; Buyanovsky *et al.* 1986; Carlyle & Than 1988). As shown in Fig. 3, however, soil respiration rates obtained by the AA-method were about twice as large as those by the OF-method



**Fig. 5.** Relationships between soil respiration rate (Rs) and CO<sub>2</sub> concentration (C) or soil surface temperature (Ts) in barley field on 10–11 May 1990. (●) Values above 20°C for (a) and above 400 parts/10<sup>6</sup> CO<sub>2</sub> for (b). The regressions are  $Rs = 1897 - 3.6C$ ,  $r = 0.87$  for (a);  $Rs = 235\exp(0.039Ts)$ ,  $r = 0.64$ , for (b).



**Fig. 6.** Relationships between soil respiration rate (Rs) and CO<sub>2</sub> concentration (C) or soil surface temperature (Ts) in corn field on 6–8 August 1990 (●) Values above 28°C for (a) and above 340 parts/10<sup>6</sup> CO<sub>2</sub> for (b). The regressions are  $Rs = 1075 - 2.3C$ ,  $r = 0.78$  for (a);  $Rs = 98.6\exp(0.035Ts)$ ,  $r = 0.88$ , for (b).

under laboratory conditions where temperature, soil moisture and soil organic matter contents were controlled at constant levels. Koizumi *et al.* (1991) reported that the soil microbial respiration rate was curvilinearly inhibited by increasing CO<sub>2</sub> concentration and the rate at 150 parts/10<sup>6</sup> was about 1.6 times that at the ambient CO<sub>2</sub> level (350 parts/10<sup>6</sup>). In this study, the CO<sub>2</sub> concentration in the chamber of the AA-method was readily decreased to about 100–200 parts/10<sup>6</sup> and kept at low CO<sub>2</sub> levels during the experiment (Fig. 4). Moreover, the

soil respiration rates by the OF-method were high at low CO<sub>2</sub> levels in the field measurements (Figs 5a, 6a). From the regression lines shown in Figs 5a and 6a, the soil respiration rates at 150 parts/10<sup>6</sup> CO<sub>2</sub> (a usual CO<sub>2</sub> level in the AA-method chambers) were extrapolated to be 2.1 and 2.7 times higher than those at 350 parts/10<sup>6</sup> CO<sub>2</sub>, respectively. The accelerating effect of low CO<sub>2</sub> concentration explains the discrepancy in soil respiration between the AA- and OF-methods (Figs 2, 3).

The ambient CO<sub>2</sub> concentration in the field

generally changes from a low level in the day to a high level at night. As shown in Fig. 5b, the soil respiration rates are relatively low above 400 parts/10<sup>6</sup> CO<sub>2</sub>. This result suggests that the soil respiration may be inhibited by the higher CO<sub>2</sub> concentration in the night-time. Therefore, the daily trend of soil respiration should be measured under natural fluctuation of ambient CO<sub>2</sub> concentration. For this purpose, the OF-method used in the present study is useful because the air in the OF-chamber is ventilated by the atmosphere in the field. Furthermore, the verification of the OF-method will be reported in a subsequent study (Beck, in prep.).

To evaluate the carbon cycle in an ecosystem, precise measurements of CO<sub>2</sub> evolution from soil are essential. Many researchers have applied the conventional AA-method for the measurement of soil respiration rate in their studies of carbon dynamics in ecosystems (Nakane 1975; Buyanovsky *et al.* 1987; Singh *et al.* 1988). However, the soil respiration rates in these studies should be re-examined whether CO<sub>2</sub> concentrations in the chamber were at a natural level or not.

The atmospheric CO<sub>2</sub> level has been increasing in recent years. Elevated CO<sub>2</sub> concentrations may lead to global warming. In the estimation of CO<sub>2</sub> emission from soil (Jenkinson *et al.* 1991), the effect of CO<sub>2</sub> concentrations on soil respiration was not taken into account.

Other analyses of changes in terrestrial carbon storage under doubled CO<sub>2</sub> climate (Prentice & Fung 1990; Schlesinger 1990) estimated that the redistribution of vegetation types would act as a larger carbon sink than the accumulation of soil organic matter. These authors assumed that soil respiration rates will be accelerated by the increase of soil temperature, without noticing that the soil respiration rates may be inhibited by higher CO<sub>2</sub> concentrations.

Koizumi *et al.* (1991) pointed out that the effect of CO<sub>2</sub> concentration on soil respiration might be changeable among a variety of microbial groups. The present study showed that the soil respiration rate in May was much more affected by the CO<sub>2</sub> concentration than the soil surface temperature (Fig. 5), while in August it was more affected by the soil surface temperature than the CO<sub>2</sub> concentration (Fig. 6). The different pattern between May and August may be caused by the difference in the species compositions of soil microbes. To predict

ecosystem responses to elevated CO<sub>2</sub> concentrations we must investigate the effect of elevated CO<sub>2</sub> on the soil carbon storage using different soil types and microbes from various areas.

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