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Measurement of soil CO_2 efflux using soda lime absorption: both quantitative and reliable

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Abstract

Measurement of soil CO_2 efflux using a non-flow-through steady-state (NFT-SS) chamber with alkali absorption of CO_2 by soda lime was tested and compared with a flow-through non-steady-state (FT-NSS) IRGA method to assess suitability of using soda lime for field monitoring over large spatial scales and integrated over a day. Potential errors and artifacts associated with the soda lime chamber method were investigated and improvements made. The following issues relating to quantification and reliable measurement of soil CO_2 efflux were evaluated: (i) absorption capacity of the soda lime, (ii) additional and thus artifactual absorption of CO_2 by soda lime during the experimental procedure, (iii) variation in the CO_2 concentration inside the chamber headspace, and (iv) effects of chamber closure on soil CO_2 efflux. Soil CO_2 efflux, as measured using soda lime (with a range of quantities: 50, 100, and 200 g per 0.082 m^2 ground area enclosed in chamber), was compared with transient IRGA measurements as a reference method that is based on well-established physical principles, using several forms of spatial and temporal comparisons. Natural variation in efflux rates ranged from 2 to 5.5 g C m^{-2} day^{-1} between different chambers and over different days. A comparison of the IRGA-based assay with measurement based on soda lime yielded an overall correlation coefficient of 0.82. The slope of the regression line was not significantly different from the 1:1 line, and the intercept was not significantly different from the origin. This result indicated that measurement of CO_2 efflux by soda lime absorption was quantitatively similar and unbiased in relation to the reference method. The soda lime method can be a highly practical method for field measurements if implemented with due care (in terms of drying and weighing soda lime, and in minimizing leakages), and validated for specific field conditions. A detailed protocol is presented for use of the soda lime method for measurement of CO_2 efflux from field soils.

Keywords: Soil CO_2 efflux; Soil respiration methods; Soda lime absorption

1. Introduction

Efflux of CO_2 from soil respiration is a major contributor to net carbon exchange in terrestrial ecosystems, second only in magnitude to photosynthesis by plants (Rustad et al., 2000). Carbon storage in soils globally is three times larger than in the atmosphere and five times that in vegetation (Schlesinger and Andrews, 2000). Soil CO_2 efflux is sensitive to temperature, moisture and disturbance. Any change in these factors could thus change the soil carbon pool in a region from being a net store to a net source of carbon (Raich and Schlesinger, 1992; Valentini et al., 2000). Furthermore, uncertainties in measuring these fluxes can cause significant errors in estimating net ecosystem exchange. Predicted effects of climate change need to include the effects of changes in temperature and moisture conditions on release of CO_2 from terrestrial carbon pools, particularly soils. In regional and global terms, even a small change in the soil CO_2 efflux can thus represent a large change in carbon flux from the land.

Selecting a technique for measuring soil CO_2 efflux is thus a major issue in the study of terrestrial carbon cycling (Raich and Schlesinger, 1992; Rochette and Hutchinson, 2004). Many different methods have been used to measure soil CO_2 efflux. Choice of an appropriate method is guided by: needs for spatial and temporal sampling, resources and equipment available, accuracy required of the measurement, assumptions and measurement artifacts, and evidence that the method can be

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applied reliably in the field (Norman et al., 1997; Lund et al., 1999). Methods are described using the nomenclature of Livingston and Hutchinson (1995). There is, at present, no standard method by which to compare methods quantitatively or test accuracy.

Soil efflux needs to be measured in many replicates over large areas in the field to be representative of an ecosystem, and measured in many regions globally. The primary source of uncertainty in estimation of soil CO_2 efflux is spatial heterogeneity. Additional sources include chamber artifacts and temporal variation in respiratory activity (Norman et al., 1997). A reliable method must minimize uncertainty from these sources. This can be achieved by assessing the source and magnitude of errors and variation due to each factor. Maximizing replication and spatial coverage of the field site is of paramount importance to sample spatial variability adequately. Enclosed chambers can induce uncertainties in measures of soil CO_2 efflux, and it is important that there is no systematic bias in the measurement. However, greater spatial replication will reduce overall uncertainty to a greater extent than will increased accuracy from fewer chambers. Hence, a method is required for measuring soil CO_2 efflux that allows spatial replication, temporal integration and a robust technique for field application.

Our aim was to test and evaluate a method of measuring soil CO_2 efflux in chambers that could be replicated in the field, and hence applied to large (100 s ha) forest areas for deriving annual budgets and seasonal patterns of carbon flux. The NFT-SS (non-flow-through steady-state) chamber technique using chemical absorption of CO_2 by soda lime appeared to be the most practical method for field measurements. It provides mean daily fluxes that can be monitored on a monthly basis for a year or more across a large, heterogeneous site. It is inexpensive and simple, equipment can be used in remote field locations under any weather conditions and can be left in forest areas used by the public, many samples can be measured allowing spatial replication, quite large chambers can be used allowing integration of micro-scale soil variability, and measurement over 24 h provides good temporal integration. However, there were questions about accuracy of this absorption method.

Previous studies have found results from different methods correlated, but often with discrepancies, non-linearity, and site-specificity (Cropper et al., 1985; Ewel et al., 1987; Rochette et al., 1992; Nay et al., 1994; Haynes and Gower, 1995; Jensen et al., 1996; Norman et al., 1997; Janssens and Ceulemans, 1998; Le Dantec et al., 1999; Janssens et al., 2000; Pumpanen et al., 2004). Non-flow-through chambers often had relatively lower values at high efflux, which are thought to be associated with lack of air movement through the chamber, and/or limitation in the rate of chemical absorption, and higher values at low efflux due to absorption of CO_2 in the headspace to below ambient concentrations. Likely reasons for these differences include use of artificial or porous media and soils that have different diffusion characteristics to undisturbed field soils (Nay et al., 1994; Rayment, 2000; Widen and Lindroth, 2003; Pumpanen et al., 2004). Differences in measured fluxes due to fine and coarse sand, and wet and dry sand demonstrated the effect of the medium on diffusion rates (Pumpanen et al., 2004), and hence the likely differences in results between methods that may occur in different soil types. Inaccuracies can occur in absorption of CO_2 by soda lime due to insufficient moisture and inadequate measurement of blanks. Such problems with absorption of CO_2 could artificially reduce the measured efflux (such as those described by Nay et al., 1994, and Knoepp and Vose, 2002). A linear relationship between CO_2 efflux measured by absorption in KOH solution and flow-through steady-state IRGA measurements was shown by Hartigan (1980). Reasons for differences among many methods have not been demonstrated experimentally in soils (Rochette et al., 1997).

These questions were addressed by systematically checking the experimental procedure in the soda lime method, and verifying the measured flux by comparing with an IRGA technique. Potential limitations addressed in our experiments were: (1) the capacity and efficiency of soda lime to absorb all the evolved CO_2, (2) the effects of CO_2 absorbed by soda lime during the experimental procedure, (3) the effects of changing the CO_2 concentration in chamber headspace, and (4) the effects of prolonged closure of chambers (e.g. for 24 h) potentially altering soil temperature and air movement, and hence CO_2 efflux. Soil CO_2 efflux measured by the soda lime method was compared with transient IRGA measurements as a reference method. This reference method was also tested to provide an integrated estimate of daily efflux. Use of an automated chamber, where the lid is closed automatically and the flux measured at intervals over a day, would provide a more accurate integrated daily estimate. However, this equipment was not available for this experiment and would not be feasible for wide-scale use in many regions. Measurements were made in field soil (rather than artificial or porous media), so that tests and verification of methods were under similar conditions to those experienced in field monitoring of soil CO_2 efflux.

2. Materials and methods

2.1. General description of the soda lime method

Soda lime granules consist of NaOH and $Ca(OH)_2$ and about 20% absorbed water. Water is required for chemical absorption of CO_2 to form Na_2CO_3 and $CaCO_3$. Carbonate formation is reflected in weight gain of granules. Weight gain of soda lime must be measured on oven-dried granules so that differences in water content of the initial batch of soda lime, and water absorption during exposure do not interfere with measured weight gain of CO_2.

The chamber technique using soda lime absorption of evolved CO_2 that we describe was based on the techniques described by Monteith et al. (1964); Edwards (1982); Zibilske (1994). A comprehensive description is given in Appendix A, including notes where modifications may be required depending on field conditions. The intention is that this can be used as a reference protocol for field application. Briefly, 50 g moist soda lime is weighed into a petri dish (for a ground area within chamber of 0.08 m^2), then oven-dried at 105 °C for 14 h. The dry weight of the soda and the petri dish is recorded and the petri dish is sealed with PVC electrical insulation tape. At the field site, the soda lime is rewetted with 8 mL of water applied as a spray before being placed in a sealed 6.9 L chamber for 24 h. After collection, dishes are again sealed with tape, transported to the laboratory, dried in a fan-forced oven and re-weighed. Chambers remain open in the field except for the 24 h of incubation. Soil CO$_2$ efflux is measured as a daily integrated value and expressed in units of gram C m^{-2} day^{-1}. Atmospheric CO$_2$ can also be absorbed by soda lime during weighing and drying, and due to chamber leakage, which are not part of the soil CO$_2$ efflux. To assess this, blank chambers, with a sealed base, were treated in the same manner as the sample chambers in the soil.

2.2. Tests of the soda lime method

Experiments were designed to test each of the potential limitations of the soda lime method and to assess the importance of blanks. (1) The capacity and efficiency of the alkali to absorb CO$_2$ depends on the amount and surface area of soda lime granules exposed in chambers. A range in amounts of soda lime was tested to determine the amount required to ensure complete absorption of CO$_2$. The effect of surface area for absorption was tested using fine and coarse grain sizes. (2) A small but significant gain in weight occurs due to CO$_2$ absorption by soda lime during the experimental procedure, which does not represent efflux from the soil. Tests of these experimental procedures compared fan-forced and vacuum ovens, weight gain during cooling and weighing of petri dishes, sealing petri dishes for transport, and leakage through the chamber lid that is sealed with silicone grease. (3) The effect of headspace CO$_2$ concentration on total CO$_2$ absorbed in sample chambers was tested using a range of weights of soda lime (50, 100, 200 g) exposed in sample chambers. Measurements of headspace CO$_2$ concentration were made by removing two bungs from the lid and quickly replacing them with two tubes that were connected to an IRGA in closed circulation. Readings were taken after 50 s to allow a standard mixing time. (4) The potential effect of 24 h closure of chambers in altering soil temperature was tested by measuring soil temperature at 1 and 5 cm depths inside and outside chambers after closure. Wind speed was measured with cup anemometers at 10 cm height outside chambers. The effect of air movement inside chambers was tested by attaching fans (40 mm computer chip cooling fan with air volume 160 L/min.) on the inside of the lid.

2.3. Description of the IRGA method

We used a flow-through, non-steady-state method, using the same chambers as above, and an IRGA from a portable photosynthesis system (LI-6200, Li-Cor, Inc., Lincoln, NE, USA). For each measurement, chamber concentration started close to ambient (approximately 370 μmol mol^{-1}), and readings were taken every 20 s over 2 min, while CO$_2$ concentration increased by approximately 200 μmol mol^{-1}. Efflux was calculated from the slope of the curve of increase in CO$_2$ concentration over time:

$$F_c = \frac{\delta C}{\delta t}(V/A)$$

where F_c is the rate of accumulation (μmol m^{-2} s^{-1}), C is concentration (μmol mol^{-1}), V is chamber volume (mol dry air), A is chamber area (m^2). Linear regression equations between headspace concentration and time were calculated and correlation coefficients were 0.999–1.000. Measurements were taken periodically over the day, and daily efflux was calculated from spot IRGA measurements by integrating the area under the curve of CO$_2$ efflux versus time. For practical field applications, spot IRGA measurements cannot be taken over the whole diel cycle. CO$_2$ efflux was related to soil temperature as a means of providing integration over the diel cycle.

Pressure within an enclosed chamber could potentially increase during closure of the lid and affect the efflux of CO$_2$. As a precaution the chamber was vented through a hole in the lid while the lid was placed on the chamber, and then a rubber bung was put in place. Pressure inside the chamber with the lid on and IRGA operating was measured using a differential pressure transducer (Validyne Engineering Corp., Northbridge CA 91324, Model DP 45, range ± 254 Pa). Pressure was measured as the difference between outside atmospheric pressure and inside chamber pressure, and the range of differential pressure was ± 1 Pa = 0.1 mm water column. This pressure fluctuation is due mostly to the circulation pump of the Li-Cor 6200. Measured soil CO$_2$ efflux is expressed in units of gram C m^{-2} day^{-1} as a daily integrated value to compare with values from the soda lime method, and expressed in units of μmol C m^{-2} s^{-1} when measuring an instantaneous flux.

2.4. Experimental site

Experiments were conducted in the field at the Australian National University campus in Canberra, which has a generally cool temperate climate (mean minimum temperature 7 °C and mean maximum temperature 20 °C, and mean annual rainfall 633 mm). Soil at the site is a Kandosol (Isbell, 1996) with a gradational profile of a loam textured A horizon and sandy clay to light clay B horizon. Soil pH is 5.4 and contains 2.4% C and 0.12% N. We chose an undisturbed field soil (measurements in situ without disrupting the soil profile), similar to that at the site of an extensive field-monitoring program. These field soils have high diffusion resistance due to their fine texture and high moisture content. Measurements were made during summer when conditions were unusually hot (air temperature range from 17 to 42 °C), and dry, so the soil was irrigated and kept moist. Ground layer vegetation was clipped so that there was no CO$_2$ uptake by plants. Eight sample chambers were inserted into the soil and eight blank chambers (with sealed base) placed on the surface within an area of 60 m^2. Chambers were allowed to equilibrate for more than a month prior to measurements.

3. Results and discussion

3.1. Improvements in methodology

The method described in the protocol specifies the amount of soda lime, its grain size, surface area as a proportion of ground area in the chamber, water required for maximum chemical absorption to occur, duration of incubation, and duration of drying. We found that complete absorption of CO_2 efflux from soil occurred using this method under the soil and efflux conditions tested.

(1) *Absorption of CO_2*. Complete absorption of the efflux of CO_2 from the soil enclosed in the chamber depends on adequate capacity (amount), efficiency and surface area (size) of soda lime granules. Tests with a range of weights of soda lime, from 20 to 100 g, over a range of efflux from 1 to 10 g C m^{-2} day^{-1}, showed that weight gain reached a plateau by 50 g. This indicated that 50 g soda lime was sufficient to allow maximum absorption of CO_2 in the chamber volume used. This amount is theoretically sufficient to absorb all the CO_2 evolved in a 24-h period, while remaining well below CO_2 absorption capacity of the soda lime of about 28%. The weight increase of exposed soda lime is $<3\%$, which is well below the recommendation of $<7\%$ by Edwards (1982), and $<10\%$ by Janssens et al. (2000). The amount of soda lime may need to be modified for application in soils with a different range of efflux, but must ensure complete absorption of CO_2 for a given efflux.

Efficiency of absorption of CO_2 by soda lime was tested by Pongracic (2002) using injection of a known amount of CO_2 gas into a sealed chamber over 4 h. CO_2 concentration in the headspace approached zero approximately 2.5 h after CO_2 inflow to the chamber ceased. The amount of CO_2 added (1.1 g per 0.08 m^2 chamber) is similar to a 24-h efflux from field soil, and soda lime absorbed $98\pm4\%$ ($n=12$) of the CO_2 added without any air movement in the chamber. This demonstrated that soda lime is able to absorb the efflux of CO_2 without limitation due to exhaustion. In a simulation of absorption by alkali in a chamber using a gas diffusion model, Rochette and Hutchinson (2004) showed that CO_2 absorption efficiency was complete after 12–24 h deployment times under a range of alkali:chamber area ratios, although small ratios resulted in initially high headspace CO_2 concentrations. During long (up to 24 h) deployment times, there is sufficient time for the CO_2 concentration gradient to adjust to any difference between the rate of CO_2 production and the rate at which CO_2 is absorbed by the alkali trap above the soil surface.

Area of petri dish with soda lime is 11.5% of the area of the soil surface within the chamber. The surface area of soda lime recommended by Edwards (1982) and Zibilske (1994) is greater than 5%, and 6% by Raich et al. (1990). Actual granule surface area for absorption is much higher. The effect of granule surface area on absorption efficiency was tested by comparing weight gain by granules of 4–8 mesh size (2.4–4.8 mm diameter), which is normally used in field experiments, with mesh size 10–16 (1.2–2.0 mm diameter). There was no significant difference in the amount of CO_2 absorbed; fine granules 664.1 mg CO_2 m^{-2} h^{-1} and coarse granules 647.3 mg CO_2 m^{-2} h^{-1} (Least Significant Difference = 106.5 using a 2-sample *t*-test (Zar, 1984)). The finer grain size is more expensive and difficult to use for routine field measurement because dust is produced and results in lost weight.

(2) *Blanks*. Subtracting blank values from the efflux measured in sample chambers takes account of weight gain due to drying in a fan-forced oven, and leakage from chambers and petri dishes. Hence, it is important to have replicated blanks in a field experiment. The average (± standard error) total weight gain of soda lime enclosed in blank chambers was 0.376 ± 0.020 g CO_2/50 g moist soda lime. The proportion of this weight gain attributable to various sources is summarized in Table 1. Weight gain during drying in a fan-forced oven constituted the majority of the blank value. This amount was tested by drying in a fan-forced oven and subtracting a blank compared with drying in a vacuum oven where no CO_2 is absorbed. The increased accuracy of the vacuum oven method is illustrated by relating IRGA and soda lime measurements, where the regression line using drying in the vacuum oven ($y=0.42+0.93x$, in g C m^{-2} day^{-1}, $r^2=0.81$) is not significantly different from the 1:1 line, whereas the regression line using drying in the fan-forced oven ($y=-1.37+1.42x$, in g C m^{-2} day^{-1}, $r^2=0.80$) has a greater deviation from the 1:1 line. Drying in the vacuum oven is a useful procedure for testing the method, but not practical for routine measurement involving large numbers of samples. The effect of drying time on weight gain was tested with duration up to 24 h. A drying time of 12 h was found sufficient to remove moisture and achieve a constant weight, and 14 h was selected to be an optimal and convenient time for field measurements. Edwards (1982) used 24 h drying time but stated that 8 h was adequate.

Leakage from sealed petri dishes was tested using rewetted soda lime placed in the field for varying lengths of time (2, 5, 8 h) and then dried and weighed. Weight gain was not significantly different to soda lime that was dried in the oven immediately and weighed. This indicated that absorption of CO_2 from the atmosphere in sealed petri dishes was not significant. Leakage from chambers was tested by measuring the change in headspace CO_2 concentration in the blank chamber and the weight gain in soda lime. The change in headspace CO_2 concentration was from 370 μmol mol^{-1} initially to 10 μmol mol^{-1} after 24 h absorption by soda lime, and the difference in weight of CO_2 is 4.4 mg in the chamber air (volume 6.9L). The weight gain by 50 g of soda lime exposed for 24 h was 53.5 mg CO_2 (dried in a vacuum oven with no weight gain). The difference of 49.1 mg represents the CO_2 that

Table 1
Sources of weight gain in blank chambers during the experimental procedure

Source of weight gain	Weight gain (g) by 50 g soda lime	Percent of blank weight gain
(1) Oven-drying (105 °C for ≧ 12 h)		
Fan-forced oven	0.3384	90.0
Vacuum oven	n.s.	n.s.
(2) Weighing soda lime		
Exposed while cooling	0.0004	0.1
Covered while cooling	n.s.	n.s.
(3) Amount of soda lime		
Complete absorption with ≥ 50 g soda lime tested for fluxes up to 1500 mg CO_2 m^{-2} h^{-1}	n.s.	n.s.
(4) Grain size		
Mesh size up to 4–8 allowed maximum surface area	n.s.	n.s.
(5) Sealed petri dishes during transport	n.s.	n.s.
(6) Chamber leakage	0.0353	9.4
(7) Headspace CO_2 concentration (at 370 μmol mol^{-1})	0.0019	0.5

Values are means ($n=13$) with average total weight gain in blanks of 0.376 ± 0.020 g/50 g moist soda lime. Tests producing no significant weight gain are shown as n.s.

leaked into the blank chamber during the 24 h. The average weight gain per sample chamber over soil was 1500 mg. Therefore, the leakage rate through the lid seal was a small 2.6% of the average weight gain during a soil CO_2 efflux measurement. This is likely to be a maximum leakage rate for the chambers because of the large CO_2 concentration gradient between the blank chamber and the atmosphere.

(3) *Headspace CO_2 concentration.* Carbon dioxide absorption by soda lime in the chamber is via slow diffusion. Concentration of CO_2 near the surface of the soda lime granules is near zero due to the large boundary layer resistance. The rate of CO_2 absorption by soda lime in an un-stirred chamber increases with the increase in headspace CO_2 concentration. A steady headspace CO_2 concentration is reached when CO_2 efflux equals the rate of absorption by soda lime. For a given spatial geometry, larger quantities of soda lime provide a larger surface area for CO_2 absorption and result in lower headspace CO_2 concentration.

This principle is illustrated in Fig. 1. The measured headspace CO_2 concentration was lower in chambers with increasing weights of soda lime, with mean (± standard error) concentrations for 50 g of 1013 μmol mol^{-1} (±24), for 100 g of 646 μmol mol^{-1} (±32), and for 200 g of 349 μmol mol^{-1} (±21) (Fig. 1). This increase in concentration above ambient for the standard 50 g soda lime in the chamber headspace volume represents a CO_2 mass of 7.4 mg, equivalent to approximately 0.5% of the CO_2 efflux from soil. The effect of higher headspace concentration on efflux was tested with IRGA measurements of headspace CO_2 concentration increasing to 1200 μmol mol^{-1} over 10 min accumulation time (Fig. 2). Efflux was linear under these conditions, which indicated that short-term changes in CO_2 concentration do not affect the efflux.

(4) *Effects of chamber enclosure.* Chambers with white reflective surfaces and under a forest canopy did not affect temperature conditions during closure for 24 h measured to one decimal place inside and outside chambers. Chamber

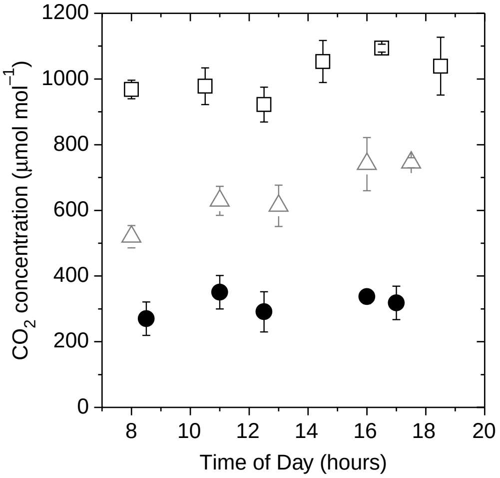

Fig. 1. Concentration of CO_2 in the headspace of chambers measured with different amounts of soda lime(□ 50, △ 100, ● 200 g). Each point represents the mean and standard error of 4 days of measurements at intervals over a day, and each measurement time using four chambers.

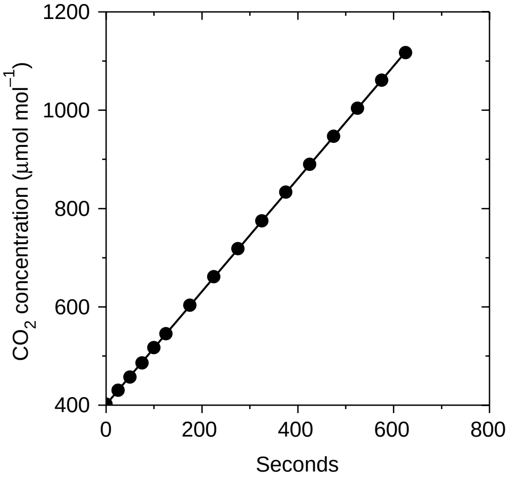

Fig. 2. Rate of soil CO_2 efflux is linear ($y=401.6+1.147x$, $r^2=0.999$) as inferred from headspace CO_2 concentration which increases up to 1200 μmol mol^{-1} over 600 s.

closure does not allow rainfall entry during a measurement period; however, the water collects on the lid and can be added to the soil when the lid is removed. Height of chambers (8 cm above ground) is minimized to reduce interruption of surface airflow as this affects turbulence and mass flow around the chamber. Reduced air movement in a chamber may decrease mass flow of CO_2 from the soil. However, wind speeds at the forest floor were always less than 1 m s^{-1}, and often less than 0.4 m^{-1}, particularly at night. Additionally, the thick litter layer and fine textured soil would minimize the effect of turbulence at ground level on soil efflux. Le Dantec et al. (1999) suggested that the turbulence caused by wind speeds less than 0.4 m s^{-1} at the soil surface had little effect on mass flow of soil CO_2 efflux, and Ohasi et al. (1995) found that efflux from bare soil was increased at 2.5 m s^{-1}. We found that introducing air turbulence into closed chambers using fans appeared to overestimate efflux rates (Fig. 3). The response of soil efflux to turbulence by fans varied among chambers depending on soil properties, particularly the gradient in soil moisture (range 18–25% gravimetric moisture content). Higher efflux with fans occurred in chambers with moderate to high efflux but not in chambers with low efflux. Differing results have been reported about the use of fans in chambers (Norman et al., 1997; Rochette and Hutchinson, 2004). Air circulation by fans may be extracting more CO_2 out of the soil or from surrounding air underneath a chamber. Our result suggests reservations about flow-through steady-state techniques that alter air movement and pressure. Similarly, Rochette et al. (1997); Hutchinson et al. (2000); Pumpanen et al. (2004) caution the use of fans because the turbulence can influence the efflux.

(5) *Temporal and spatial variability.* The instantaneous measurement of CO_2 efflux by an IRGA has a high degree of accuracy. However, this method of IRGA measurement in a non-steady-state chamber cannot be made continuously over 24 h in a large, replicated field situation, can only be made at a limited number of time points during the day, and it is difficult to do measurements at night. Accuracy of daily estimates depends on the method of temporal integration. An example of spot IRGA measurements for eight chambers integrated over 1 day is shown in Fig. 4. These spot measurements may be a poor representation of the integrated daily value of soil CO_2 efflux. For example, the difference between single mid-morning measurements and the daily integrated value derived from the temperature response ranged from −29 to +65%. Similarly, Savage and Davidson (2003) found that extrapolation of single mid-morning measurements to daily flux values were on average 13% lower than daily estimates obtained from summing hourly measurements over 24 h. Irvine and Law (2002) found that measurements of daily minimum and maximum fluxes overestimated the mean daily flux by 7%.

Soils enclosed by the eight chambers varied in their magnitude and amplitude of CO_2 efflux over the day; this is likely due to variability in soil characteristics, such as moisture content, temperature, organic matter quantity and composition, and biological activity. This degree of variability occurred even though chamber size is relatively large in terms of ground area covered (0.08 m^2) to spatially integrate heterogeneous soil conditions and minimize edge effects, and in terms of volume to minimize CO_2 concentration gradients in the headspace. In comparison, the standard chamber used with the commercially available IRGA system is smaller (two versions with area = 0.00837 or 0.0393 m^2 for a Li-Cor). Efflux is calculated on a surface area not volume basis, and so the problem of determining the effective volume of air in the chamber including air-filled spaces in the soil, which is required for

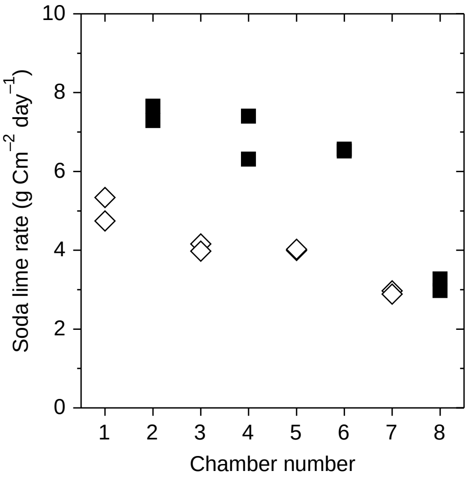

Fig. 3. Soil CO_2 efflux measured by soda lime over 24 h with (■) and without (◇) fans in the chamber lid on 2 days. A gradient in soil moisture occurred along the line of chambers from wetter near chamber 1 to drier near chamber 8.

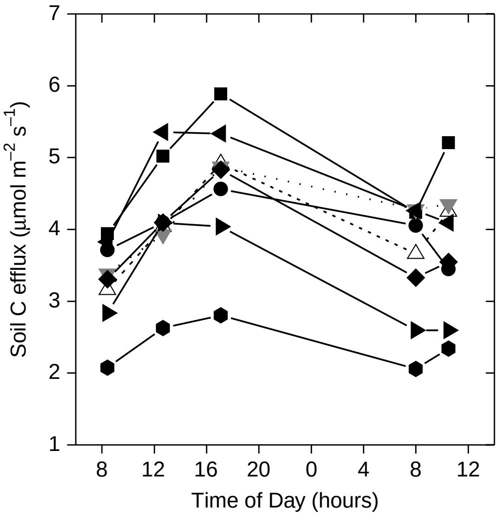

Fig. 4. Daily time course of soil CO_2 efflux obtained from transient IRGA measurements at five times during the day. Daily efflux is calculated by integrating under the curve. Eight chambers within a 20 m^2 area differ in their rate of efflux and amplitude over the day, illustrating the spatial heterogeneity of soil CO_2 efflux.

flow-through steady-state methods, is not an issue with this method.

3.2. Verification of the soda lime method against the IRGA method

Accuracy of the soda lime method for measuring soil CO_2 efflux was verified by comparison with transient IRGA measurements. There is no ideal way to measure soil efflux within the same chamber by two methods simultaneously. Therefore, we used four ways of comparing the two methods based on various spatial and temporal separations (Figs. 5a–c and 7). We describe the sequence of procedures to demonstrate how we resolved problems with each method and finally derived a reliable comparison.

(a) Soda lime and IRGA measurements were performed over 24 h in two sets of four chambers. On day 1, four chambers were measured with soda lime and four chambers with the IRGA, and on day 2 the methods were reversed in each set of four chambers. IRGA measurements were taken periodically during the day and the values integrated over 24 h as described in the Methods. The lids of the four chambers for IRGA measurements were left open in between measurements. Fig. 5a shows that soda lime values were consistently higher, but the intercept was not significantly different from the origin, and the slope was not significantly different from the 1:1 line. A potential problem with this comparison is that both efflux from individual chambers and conditions on each day vary.

(b) All chambers were measured on the same day using both methods by taking IRGA measurements at midday before putting soda lime into chambers, and then again at midday the following day after soda lime was removed. The IRGA rate was calculated using the average of these two measurements. Fig. 5b shows that the IRGA values were higher than those for soda lime across the eight chambers, probably because the measurements were made during the warmer part of the day. The correlation between the two methods was poor, the intercept was not significantly different from the origin, and the slope of the regression line was not significantly different from the 1:1 line.

(c) All chambers were measured concurrently over the same 24-h period with both IRGA and soda lime. A set of transient IRGA measurements was performed on all eight chambers in the morning. Then petri dishes with soda lime were placed in chambers. At five intervals during the day, chamber lids were taken off, petri dishes removed and placed in a gas tight container with a nitrogen gas purge to ensure minimum uptake of CO_2 while outside the chambers. Chambers were opened for 10 min and fanned to allow chamber air to equilibrate with the atmosphere (after testing that the concentration had returned to ambient). Efflux rate was then measured with the IRGA to check that the flux rate was steady before commencing measurements. A set of IRGA measurements was then performed on all chambers. Petri dishes of soda lime were replaced in chambers and lids sealed. Calculation of the absorption of CO_2 by soda lime accounted for the time petri dishes were out of the chambers plus 15 min equilibration time for headspace CO_2 concentration after dishes were returned to chambers. The equilibration time was established by measuring the time to reach stable headspace CO_2 concentration after soda lime was returned to chambers and the chambers sealed. Fig. 5c shows that the correlation was reasonable, the intercept was not significantly different from the origin, and the slope of the regression line was not significantly different from the 1:1 line.

(d) Transient IRGA measurements were integrated over the

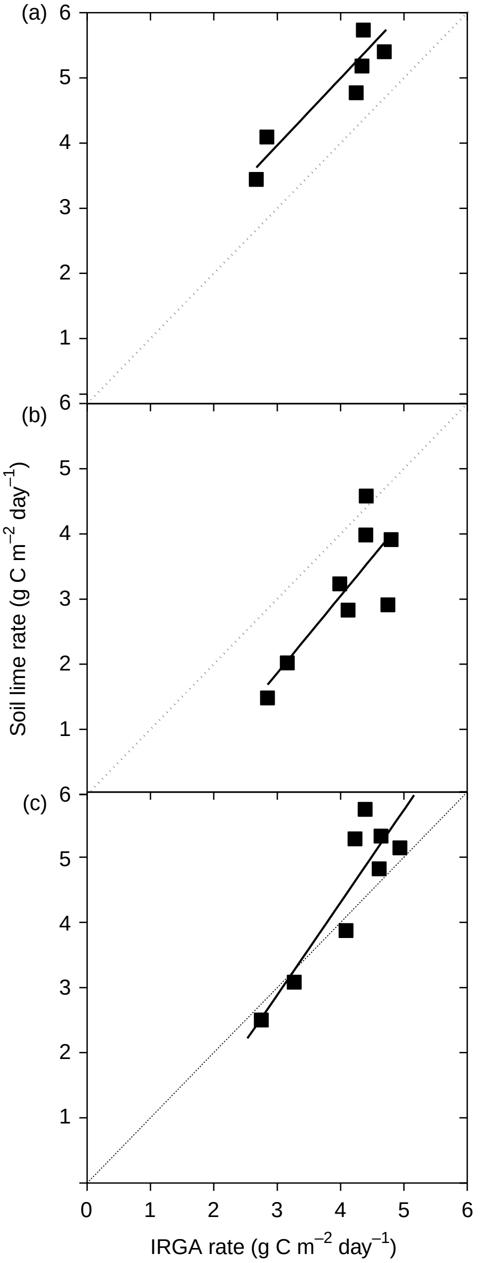

Fig. 5. Comparison of daily soil efflux (g C m^{-2} day^{-1}) measured by soda lime and IRGA, where each point represents a chamber. Dotted line represents the 1:1 line and the solid line is the regression line. (a) Measurements on 2 days alternating chambers with each method ($y=0.87+1.03x$, $r^2=0.84$). (b) IRGA measurements made before and after the 24-h incubation with soda lime ($y=-1.69+1.18x$, $r^2=0.66$). (c) Measurements by soda lime and IRGA simultaneously on the same chambers and day ($y=-1.37+1.42x$, $r^2=0.80$).

day by relating soil efflux to soil temperature, to account for the diurnal variation in temperature (in this experiment air temperature range was 16 to 36 °C). Soil temperature was logged continuously over several days at depths of 2, 5 and 10 cm. Soil CO_2 efflux was measured over two daily time courses using transient IRGA measurements five times during a 24-h period. The efflux from each chamber was related to soil temperature, and a linear temperature response equation derived for each chamber. The temperature response varied among the eight chambers due to variation in soil properties, and on different days depending on soil moisture conditions. The range in the temperature coefficient was from 0.161 to 0.539 μmol C m^{-2} s^{-1} per °C on day 1, and from 0.218 to 0.552 μmol C m^{-2} s^{-1} per °C on day 2, a week later. Relationships were tested between efflux rate and soil temperature at soil depths of 2, 5 and 10 cm. The average temperature from depths 5 and 10 cm provided the best correlation, with $r^2 > 0.94$ for each chamber, whereas other depths and combinations had lower r^2 values.

Rate of soil CO_2 efflux was calculated using the temperature response equation and the recorded soil temperature at 5-min intervals and summed over 24 h to provide a daily flux. An example of the diurnal pattern of soil and air temperature and CO_2 efflux of one chamber is shown in Fig. 6. The area under the efflux curve was integrated over 24 h to give the daily flux. Soil CO_2 efflux was measured with soda lime (using weights 50, 100, 200 g) over the same two daily time courses. The daily efflux was compared between measurements from soda lime and IRGA values interpolated from the temperature response function (Fig. 7). The slope of the regression line was not significantly different from the 1:1 line, the intercept was not significantly different from the origin, and the correlation coefficient was high, all of which indicate that the results of the two methods were similar and there was no bias in the comparison. The 1:1 relationship between the two methods was the same irrespective of soda lime weight and consequent differences in headspace CO_2 concentrations.

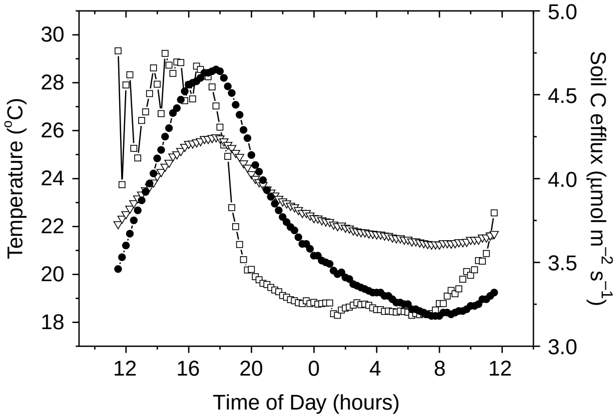

Fig. 6. Diurnal pattern of air temperature(□), soil temperature (▽,averaged over 5–10 cm depth), and soil CO_2 efflux (●) derived from the soil temperature response function for the chamber.

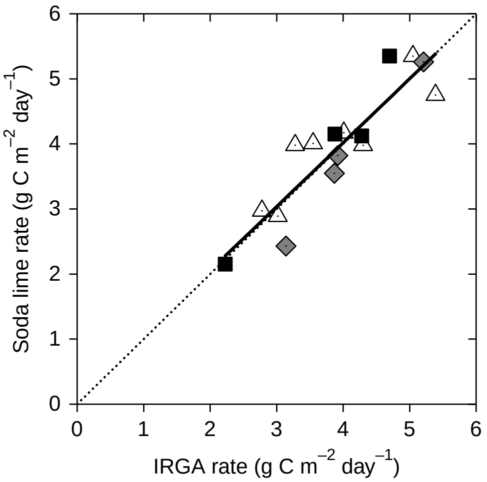

Fig. 7. Comparison of daily soil efflux (g C m^{-2} day^{-1}) measured by soda lime and IRGA, where each point represents a chamber. Dotted line represents the 1: 1 line and the solid line is the regression line. Measurements by soda lime and IRGA simultaneously on the same chambers and day, with IRGA measurements integrated over 24 h using temperature response functions ($y = 0.10 + 0.98x$, $r^2 = 0.82$). Different amounts of soda lime (△ 50 g, ◇ 100 g, ■ 200 g) resulted in different chamber headspace CO_2 concentration.

This success in using the soda lime chamber method for measuring soil CO_2 efflux was likely dependent on using soil with high diffusion resistance, as occurs in most undisturbed field soils with some clay content. Rate of diffusion is determined by the concentration gradient of CO_2 between the soil and atmosphere. The rate varies with the diffusion resistance of the soil, which is controlled by soil attributes such as structure, porosity, and moisture content. In undisturbed soils, diffusion is driven by a very large difference in concentration between low values in the atmosphere and high values in the soil (one to two orders of magnitude higher) (Hamada and Tanaka, 2001; Schwendenmann, 2002). Changing the atmospheric concentration, for example in the headspace of a chamber to 1000 μmol mol^{-1}, has a relatively small effect on the concentration gradient, and hence on the transient rate of CO_2 efflux. The large gradient in CO_2 concentration between soil and atmosphere implies that upward diffusion of CO_2 is governed by the diffusive resistance of soil and is almost independent of atmospheric mixing (Monteith et al., 1964). Additionally, soil CO_2 efflux is driven by production of CO_2 by root and microbial respiration, and this is likely to be the primary factor controlling efflux in organic field soils.

4. Conclusions

Chamber methods that are properly designed and deployed can provide reliable means of accurately measuring soil CO_2 efflux in terrestrial ecosystems (Davidson et al., 2002). A conclusion from a comparison of 20 methods was that reliability of the chamber system was not related to the measurement principle, so that good results could be achieved with steady-state or non-steady-state systems (Pumpanen et al., 2004). The soda lime method has been recommended by Cropper et al. (1985); Ewel et al. (1987); Rochette et al. (1997); Janssens and Ceulemans (1998); Janssens et al. (2000); ESF

(2000), and Rochette and Hutchinson (2004) as a useful technique to give an integrated estimate of the mean flux over a time period at heterogeneous sites where a large number of measurements is required simultaneously to integrate the spatial variation.

Our experiments have demonstrated that the soda lime absorption method can be made reliable, and verified by an IRGA method, for measuring soil CO_2 efflux accurately in undisturbed field soils. We have addressed the issues concerning accuracy of the soda lime method in terms of chamber design and experimental procedure, and have developed a protocol for field measurement (see Appendix A). Variation in CO_2 concentration that occurs in the chamber headspace does not affect efflux over a 24 h measurement period. The relationship between soda lime and IRGA measurements was found to be 1:1, and there was no evidence of limitation in the rate of absorption. This method has advantages for large-scale field experiments that require spatial replication, temporal integration and long-term monitoring. The chamber design and experimental procedure described in Appendix A worked in our environment with medium textured field soil and a given range in efflux, however, some tests would be advisable if used under different conditions, especially where soils are highly porous. The main factor to determine is whether a change in headspace CO_2 concentration affects the soil CO_2 efflux for a specific soil type, especially the flux averaged over 24 h.

A combination of techniques may be a useful approach for field applications, such as initial comparison of soda lime with IRGA in the field soil, monitoring using soda lime (e.g. 24 h measurements every month over a year or more), continuous logging of soil temperature and moisture to allow daily interpolation of soil CO_2 efflux from periodic measurements, and use of an automated flow-through steady state chamber to monitor diurnal variation and short-term temporal patterns during campaign measurements at a single site. Such a combination of methods, based on evaluation, long-term monitoring, and campaign measurements, will provide good spatial and temporal resolution of the soil CO_2 efflux. Application of the soda lime method for determining soil CO_2 efflux will allow broad-scale, replicated measurements in many regions globally, which will form an integral part of ecosystem carbon studies and contribute to interpretation of net ecosystem carbon exchange.

Appendix A: Protocol for the soda lime method

A.1. The Chamber:

(1) Chambers are large (e.g. internal diameter 32.4 cm, internal height below lid 8.4 cm, surface area 0.08246 m^2), with a well-sealed lid, for example, polyethylene plastic buckets with the base cut off (see Fig. 8). Chamber size can be modified to suit specific site conditions, but should be sufficiently large to cover micro-scale soil heterogeneity. White plastic is used to minimize heating within the chamber. Greater protection from radiation may be required in an open environment, such as a reflective coating or shield.

(2) The bucket lid has a 2 cm depth around the edge and this fits inside the rim of the bucket top to form a good seal. The chamber rim is coated with vacuum grease to ensure a tight seal between chamber and lid, and this is re-applied regularly.

(3) Chambers are inserted into the ground several weeks before measurements commence to avoid effects of soil disturbance. Chambers are left uncovered and in place for the entire course of the experiment. Placement of the chamber can cause mechanical disturbance that may affect the diffusion coefficient of the soil and possibly stimulate microbial activity to produce a flush of CO_2.

(4) A circular metal template with the shape of the chamber is used to cut through the litter layer and surface soil to a depth of approximately 1 cm. Chambers are then inserted into the litter and surface soil, to provide a good seal and anchorage but not to sever the root mat. Chambers are pegged into the ground for stability.

(5) All live vegetation inside the chamber is clipped at ground level prior to measurement to prevent CO_2 uptake or evolution. This may have to be repeated during the year.

(6) When the chamber is in use during a measurement period, a weight (e.g. stone) is placed on the lid to secure it firmly.

(7) Blank chambers consist of the same cut-off bucket with well-sealed lid, but with a Perspex base plate sealed to the base with silicone. The blank chamber is the same volume as the sample chambers, but they are not exposed to soil.

A.2. Soda lime:

(1) Soda lime granules are exposed in an inert dish (e.g. glass petri dishes of 11 cm diameter).

(2) 50 g soda lime is weighed into the petri dish (i.e. 0.06 g cm^{-2} of soil surface in chamber).

(3) Soda lime granules have a mesh size of 4– 8 (2.4 –

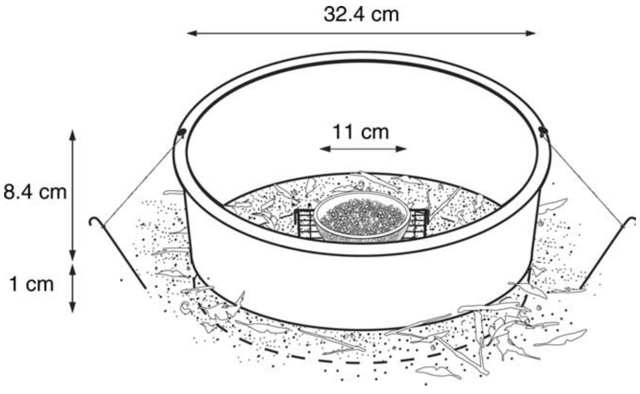

Fig. 8. Diagram illustrating the chamber design inserted in the soil, with petri dish of soda lime on wire stand. A lid is fitted on the chamber during incubation periods.

4.8 mm diameter).

(4) Soda lime is oven-dried at 105 °C for 14 hrs to constant weight (e.g. 6 p.m. to 8 a.m.), and weighed to the accuracy of 0.1 mg. The oven is fan-forced and well-vented to allow rapid and even drying. Petri dishes are covered while cooling, and preferably placed in a desiccator, for a standard time before weighing.

(5) Petri dishes with dried soda lime are sealed with electrical PVC insulation tape and placed in a box for transport to the field site.

(6) At each chamber in the field, petri dishes are untaped and water (8 mls for 50 g) is added to soda lime using a fine spray that wets all the granules evenly. This amount of water is equivalent to the difference in moisture content of soda lime due to drying. A source of moisture is required with the exposed soda lime because the reaction between hydroxide and CO_2 requires the presence of water.

(7) The lid is placed underneath the petri dish, and dishes placed on wire stands suspended above the litter surface to avoid obstructing CO_2 efflux from the soil surface in the chamber. The lid is secured on the chamber and the soda lime exposed to the soil inside for a 24-h period. Time of chamber closure and recollection are recorded so that the exact time of incubation can be calculated.

(8) The next day, dishes are collected, lids taped on, and transported back to the lab. Dishes are oven-dried for 14 h, cooled in a desiccator and then weighed.

(9) Blank measurements are made to account for CO_2 absorbed by soda lime during the experimental procedure. Petri dishes with 50 g soda lime, that undergo the same process of drying and weighing, are placed in blank chambers and left for 24 h in the field to simulate conditions of the incubating chambers. One blank chamber was used per five sample chambers and located randomly, with a minimum of three blanks for a field site. The number of blanks required for an experiment should be tested so that a tightly constrained mean blank value is obtained.

A.3. Calculations:

(1) The method is based on the adsorption of CO_2 by soda lime that is measured by a weight gain. The following absorption reactions occur:

$$2NaOH + CO_2 \Rightarrow Na_2CO_3 + H_2O$$

$$Ca(OH)_2 + CO_2 \Rightarrow CaCO_3 + H_2O$$

For every mole of CO_2 that chemically reacts with soda lime, one mole of water is formed that is subsequently evaporated during drying. Thus, the increase in dry mass after exposure underestimates CO_2 adsorbed by a factor of $18/44 = 40.9\%$. The correction factor used to account for water formed during chemical absorption of CO_2 by soda lime and released during drying is 1.69 as recognized by Grogan (1998) (not 1.41 as originally stated by Edwards (1982)).

(2) Weights recorded include: petri dish, petri dish + soda lime, petri dish + dried soda lime, petri dish + exposed dried soda lime.

(3) Calculations include: percent moisture content in soda lime, soda lime weight gain (g) after exposure, soil CO_2 efflux (g C m^{-2} day^{-1}).

$$\text{Soil } CO_2 \text{ efflux(g C m}^{-2}\text{day}^{-1}) = \left\{ \frac{[(\text{sample weight gain(g)} - \text{mean blank weight gain(g)}) \times 1.69]}{\text{chamber area(m}^2)} \right\} \times \left[\frac{24(\text{h})}{\text{duration of exposure(h)}} \right] \times \left[\frac{12}{44} \right]$$

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References

Cropper Jr.,W.P., Ewel, K.C., Raich, J.W., 1985. The measurement of soil CO_2 evolution in situ. Pedobiologia 28, 35–40.

Davidson, E.A., Savage, K., Verchot, L.V., Rosa Navarro, L.V., 2002. Minimizing artifacts and biases in chamber-based measurements of soil respiration. Agricultural Forest Meteorology 113, 21–37.

Edwards, N.T., 1982. The use of soda-lime for measuring respiration rates in terrestrial systems. Pedobiologia 23, 321–330.

ESF (European Science Foundation), 2000. Investigating the Role of Soils in the Terrestrial Carbon Balance-Harmonizing Methods for Measuring Soil CO_2 Efflux. LESC Exploratory Workshop, Edinburgh, 6–8 April 2000.

Ewel, K.C., Croppe Jr., W.P., Gholz, H.L., 1987. Soil CO_2 evolution in Florida slash pine plantations. I. Changes through time. Canadian Journal Forest Research 17, 325–329.

Grogan, P., 1998. CO_2 flux measurement using soda lime: correction for water formed during CO_2 adsorption. Ecology 79, 1467–1468.

Hamada, Y., Tanaka, T., 2001. Dynamics of carbon dioxide in soil profiles based on long-term field observation. Hydrological Processes 15, 1829–1845.

Hartigan, R.J., 1980. Soil Respiration as an Index of Forest Floor Metabolism. PhD Thesis, University of New England.

Haynes, B.E., Gower, S.T., 1995. Belowground carbon allocation in unfertilized Red pine plantations in Northern Wisconsin. Tree Physiology 15, 317–325.

Hutchinson, G.L., Livingston, G.P., Healy, R.W., Striegl, R.G., 2000. Chamber measurement of surface-atmosphere trace gas exchange: numerical evaluation of dependence on soil, interfacial layer, and source/sink properties. Journal Geophysical Research 105, 8865–8875.

Irvine, J., Law, B.E., 2002. Contrasting soil respiration in young and old-growth ponderosa pine forests. Global Change Biology 8, 1183–1194.

Isbell, R.F., 1996. The Australian soil classification. Australian Soil and Land Survey Handbook Series, vol. 4. CSIRO Australia.

Janssens, I.A., Ceulemans, R., 1998. Spatial variability in forest soil CO_2 efflux assessed with a calibrated soda lime technique. Ecological Letters 1, 95–98.

Janssens, I.A., Kowalski, A.S., Longdoz, B., Ceulemans, R., 2000. Assessing forest soil CO_2 efflux: an in situ comparison of four techniques. Tree Physiology 20, 23–32.

Jensen, L.S., Mueller, K.R., Tate, D.J., Magid, J., Nielsen, N.E., 1996. Soil surface CO_2 flux as an index of soil respiration in situ: a comparison of two chamber methods. Soil Biology & Biochemistry 28 (10/11), 1297–1306.

Knoepp, J.D., Vose, J.M., 2002. Quantitative Comparison of In Situ Soil CO_2 Flux Measurement Methods. Research Paper SRS-28. US Department of Agriculture, Forest Service, Southern Research Station, Asheville, NC, 11 pp.

Le Dantec, V., Epron, D., Dufrene, E., 1999. Soil CO_2 efflux in a beech forest: comparison of two closed dynamic systems. Plant & Soil 214, 125–132.

Livingston, G.P. and Hutchinson, G.L., 1995. Enclosure-based measurement of trace gas exchange: applications and sources of error. In: Matson P.A., Harris R.C. (Eds.), Methods in Ecology. Biogenic Trace Gases: Measuring Emissions From Soil and Water. Blackwell Scientific Publ, pp. 14–51.

Lund, C.P., Riley, W.J., Pierce, L.L., Field, C.B., 1999. The effects of chamber pressurization on soil-surface CO_2 flux and the implications for NEE measurements under elevated CO_2. Global Change Biology 5, 269–281.

Monteith, J.L., Szeicz, G., Yabuki, K., 1964. Crop photosynthesis and the flux of carbon dioxide below the canopy. Journal Applied Ecology 1, 321–337.

Nay, S.M., Mattson, K.M., Bormann, B.T., 1994. Biases of chamber methods for measuring soil CO2 efflux demonstrated with a laboratory apparatus. Ecology 75 (8), 2460–2463.

Norman, J.M., Kucharik, C.J., Gower, S.T., Baldocchi, D.D., Crill, P.M., Rayment, M., Savage, K., Striegl, R.G., 1997. A comparison of six methods for measuring soil-surface carbon dioxide fluxes. Journal Geophysical Research 102, 771–777.

Ohasi, M., Gyokusen, K., Saito, A., 1995. Effects of carbon dioxide concentration and wind speed using the chamber method on soil respiration. Journal Japanese Forest Society 77 (6), 599–601.

Pongracic, S., 2002. Influence of Irrigation and Fertilization on the Belowground Carbon Allocation in A pine Plantation. PhD thesis, University of New South Wales, Sydney, Australia, 207 pp.

Pumpanen, J., et al., 2004. Comparison of different chamber techniques for measuring soil CO_2 efflux. Agricultural Forest Meteorology 123, 159–176.

Raich, J.W., Schlesinger, W.H., 1992. The global carbon dioxide flux in soil respiration and its relationship to vegetation and climate. Tellus 44B, 81–99.

Raich, J.W., Bowden, R.D., Steudler, P.A., 1990. Comparison of two static chamber techniques for determining carbon dioxide efflux from forest soils. Soil Science Society of America Journal 54, 1754–1757.

Rayment, M.B., 2000. Closed chamber systems underestimate soil CO_2 efflux. European Journal Soil Science 51 (1), 107–110.

Rochette, P., Hutchinson, G.L., 2005. Measurement of soil respiration in situ: chamber techniques. In: Hatfield J.L., Baker J.M. (Eds.), Micrometeorology in Agricultural Systems ASA-CSSA-SSSA. Agron. Monogr. 47, pp 247–286.

Rochette, P., Gregorich, E.G., Desjardins, R.L., 1992. Comparison of static and dynamic closed chambers for measurement of soil respiration under field conditions. Canadian Journal of Soil Science 72, 605–609.

Rochette, P., Ellert, B., Gregorich, E.G., Desjardins, R.L., Pattey, E., Lessard, R., Johnson, B.G., 1997. Description of a dynamic closed chamber for measuring soil respiration and its comparison with other techniques. Canadian Journal Soil Science 77, 195–203.

Rustad, L.E., Huntington, T.G., Boone, R.D., 2000. Controls on soil respiration: implications for climate change. Biochemistry 48, 1–6.

Savage, K.E., Davidson, E.A., 2003. A comparison of manual and automated systems for soil CO_2 flux measurements: trade-offs between spatial and temporal resolution. Journal Experimental Botany 54, 891–899.

Schlesinger, W.H., Andrews, J.A., 2000. Soil respiration and the global cycle. Biogeochemistry 48, 7–20.

Schwendenmann, L.C., 2002. Belowground Carbon Dynamics as a Function of Climate Variability in Undisturbed Soils of a Neotropical Rainforest. PhD Thesis, University of Gottingen, Germany, 133 pp.

Valentini, R., et al., 2000. Respiration as the main determinant of carbon balance in European forests. Nature 404, 861–865.

Widen, B., Lindroth, A., 2003. A calibration system for carbon dioxide-efflux measurement chambers: description and application. Soil Science Society America Journal 67, 327–334.

Zar, J.H., 1984. Biostatistical Analysis, second ed. Prentice-Hall, Englewood Cliffs, NJ.

Zibilske, L.M., 1994. Carbon mineralization. In: Mickelson, S.H. (Ed.), Methods of Soil Analysis Part 2 — Microbiological and Biochemical Properties. Publ. Soil Science Society of America, Inc., p. 836.