Evaluation of CO₂ Exchange Rates in a Wetland Ecosystem Using the Closed Geosphere Experiment Facility

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ABSTRACT

To evaluate annual CO₂ exchange rates in a wetland ecosystem, ecosystem respiration rate (Re), net ecosystem productivity (NEP), and gross primary productivity (GPP) were investigated using the Closed Geosphere Experiment Facility (CGEF) located in northeastern Japan. The Cgef is highly airtight and equipped with a Geosphere Module (GM). The GM has a ground area of 5.8 × 8.7 m² and an average height of 11.9 m, including a soil depth of 3.1 m. A wetland ecosystem dominated by *Phragmites australis* was introduced into the Cgef. Air temperature and CO₂ concentration in the GM were controlled automatically. The hourly nighttime Re increased exponentially with the hourly average air temperature. Both hourly NEP and GPP depended on hourly photosynthetic photon flux density (PPFD). In addition, daily ecosystem CO₂ exchange rates (Re, NEP, and GPP) were influenced by above-ground plant biomass. The annual NEP was found to be 64.2 ± 19.2 g C m⁻² yr⁻¹ and it resulted from the annual GPP of 555.8 ± 17.0 g C m⁻² yr⁻¹ and annual Re of −491.6 ± 15.6 g C m⁻² yr⁻¹. Therefore, the wetland ecosystem behaved as a CO₂ sink for the entire year. The annual CO₂ exchange rates obtained were reasonable values compared to the findings of published studies in *P. australis*-dominated wild wetlands using the eddy covariance technique and the combined method of internal gas pressures and flow measurements and harvesting.

1. Introduction

In research on patterns of carbon cycling in terrestrial ecosystems, much attention has been paid to investigate whether an ecosystem is a net source or sink for atmospheric CO₂, to examine factors affecting this source or sink, and to estimate the trends in carbon (C) balance for the future (Mooney et al. 1991; Ryan 1991; Shaver et al. 1992; Schimel 1995; Goulden et al. 1998; Malhi et al. 1999; Valentini et al. 2000; Brix et al. 2001; Flanagan et al. 2002; Houghton 2003; Dunn et al. 2007; Waddington and Roulet 2008). Wetlands are expected to contribute to global carbon dynamics through the exchange of CO₂ and CH₄. Long-term carbon accumulation in terrestrial ecosystems may be relevant either to forests that produce large amounts of recalcitrant lignified tissue or wetlands where the anaerobic conditions in soils reduce decomposition by soil microbes (Brix et al. 2001). The organic carbon reserve in wetlands is estimated to represent around 15% of the reserve of terrestrial organic carbon on earth (Sabine et al. 2004).

The common reed *Phragmites australis* (*P. australis*) is widespread throughout wetlands worldwide; this species is found on every continent, except Antarctica, from subarctic to warm temperate zones (Osada 1993; Clevering and Lissner 1999; Brix et al. 2001). It is a C₃ plant and a perennial grass, which typically forms closed and monodominant stands in the littoral zone of lakes, ponds, along rivers, and in marshes (Brix 1999). Throughout the twentieth century, the distribution of the species was enlarged and *P. australis* was reported in all of the lower 48 U.S. states and across southern Canada (Chambers et al. 1999; Saltonstall 2002). The species is regarded as an

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aggressive invader in North America (Chambers et al. 1999; Rice et al. 2000). On the other hand, in Europe, the balance between the progression and regression of the species has recently shifted toward a process of die back, resulting in setting up of the European research project on reed die-back and progression (EUREED) project to investigate the mechanisms of such phenomena (Brix 1999). Besides, *P. australis*-dominated wetlands are important vegetation types because the species has pressurized gas-flow aeration functions that enhance the transfer of CO$_2$ and CH$_4$ from sediments to the atmosphere (Armstrong and Armstrong 1990; Brix et al. 1992, 1996). Furthermore, the wetlands have high annual net primary productivity (Brix et al. 2001). Therefore, it is valuable to investigate carbon dynamics of *P. australis*-dominated wetlands.

A main focus of many research studies in terrestrial carbon dynamics is on CO$_2$ exchange rates of ecosystems. To estimate ecosystem carbon balance, continuous gas exchange measurements are essential on hourly to yearly time scales. The eddy covariance technique is now widely used to assess CO$_2$ exchange rates of an entire ecosystem across a spectrum of time scales, ranging from seconds to years, by measuring the covariance between the fluctuations in vertical wind velocity and CO$_2$ mixing ratio (Baldocchi 2003). Quantifying the temporal variation in ecosystem CO$_2$ exchange rates at one site helps to clarify the effects of environmental variation on ecosystem physiological processes. Data gathered during fluctuations in environmental conditions are useful to develop physiological response curves for whole-ecosystem response to changes in temperature, light, and so on (Margolis et al. 2006). The net CO$_2$ exchange rate of terrestrial ecosystems is actuated by the balance between the CO$_2$ input by photosynthesis and its output by emission through plant and soil respiration. The carbon dynamics model infers that terrestrial ecosystems will ultimately shift from a net sink to a source of CO$_2$ because respiration rate increases faster than photosynthetic rate as ecosystems acclimate to climate change (Cox et al. 2000). Therefore, it is very important to validate the model under simulated environmental changes such as artificial warming and increased air CO$_2$ concentration at the ecosystem level.

The Closed Geosphere Experiment Facility (CGEF) located in northeastern Japan (40°53′N, 141°16′E) was designed to investigate carbon dynamics in terrestrial ecosystems. A wetland ecosystem with *P. australis*-dominated vegetation was introduced along with soil into the Cgef from a wild wetland in December 2006 and the ecosystem was verified to have been properly reconstructed from physicochemical and biological viewpoints (Suzuki and Endo 2007). It is highly water-tight so that no leaching of dissolved organic carbon occurs from it. Air leakage from the facility is very small and its contribution to carbon balance has been found to be less than 0.5% (Suzuki et al. 2010b). As such, the Cgef has a number of advantages that make it useful in understanding ecosystem behavior within an environment with regulated variables. Its regulated environmental factors include air temperature and CO$_2$ concentration that influence the carbon cycles of ecosystems (Suzuki et al. 2010a). In counterpoint to these benefits, the controlled environment does not represent a natural environment, which introduces its own disadvantages.

Using the Cgef, we made plans to investigate carbon dynamics of *P. australis*-dominated wetlands and to examine the responses of the wetlands to artificial environmental changes through the facility’s capability for controlling wetland environmental conditions. The objectives in this paper are 1) to estimate CO$_2$ exchange rates [net ecosystem productivity (NEP), gross primary productivity (GPP), and ecosystem respiration rate ($R_e$)] of the introduced wetland in the Cgef and 2) to investigate the factors governing the CO$_2$ exchange rates.

2. Materials and methods

a. Introduction of a wetland ecosystem into the Cgef

The Cgef includes a geosphere module (GM) and a geosphere material circulation system (GMCS). The roof and three (south, east, and west) sides of the GM are covered with glass panels so that natural light can be introduced. The GM is airtight and its glass panels are set in special silicone gaskets to prevent both air outflow and inflow. The GM has a ground area of 5.8 × 8.7 m$^2$ and 11.9-m average height including a soil depth space of 3.1 m (Fig. 1a). The GMCS is composed of two subsystems: an air process subsystem and a freshwater process subsystem. The air process subsystem has a pressure controller, a CO$_2$ and an O$_2$ scrubber, a CO$_2$ and an O$_2$ tank, an air composition adjuster, and an air conditioner. This subsystem controls air temperature and CO$_2$ concentration in the GM. The freshwater process subsystem has a freshwater processor and an artificial rain generator. This subsystem supplies water to soil and plants in the GM (Suzuki et al. 2010a).

A wetland ecosystem dominated by the common reed *P. australis* was introduced into the GM in December 2006 from a wild wetland (40°93′N, 141°36′E). A preliminary study was conducted in the wild wetland. A 3-m soil core was taken and soil characteristics were investigated to determine the soil depth for introduction from the wild wetland to the GM. The vertical profile of the total C content in the soil had the following values: 8.4, 3.0–4.2, and 2.4–3.8 mg C g$^{-1}$ soil at 0–0.2-, 0.2–1.0-,
and 1.0–3.0-m depths, respectively. A similar trend for the vertical profile was observed for microbial biomass carbon content: 422 mg C g⁻¹ soil at 0–0.2-m depth, 11–95 mg C g⁻¹ soil at 0.2–1.0-m depth, and almost none detected at 1.0–3.0-m depth (Suzuki and Endo 2007).

The preliminary study showed that soil below 1-m depth was expected to contribute little to the carbon cycle in the wetland ecosystem. Blocks of rhizospheric soil were removed from the wild wetland using a 1.5 × 1.0 × 0.45 m³ iron frame to minimize damage to the rhizomes and roots. Commercial sea sand was washed with water and used to fill the soil depth space from the bottom at 3.1–1 m from the space top height. Sand collected from the wild wetland was placed on top of the sea sand to form a layer from 1 to 0.45 m from the space top height. Twenty-seven blocks of rhizospheric soil were tightly configured on the second layer to finish filling of the soil depth space. Rhizomes and roots were mostly present up to a depth of 0.3 m from the surface, indicating that the blocks sufficiently included the rhizosphere. All layers were hydraulically filled with groundwater collected from the wild wetland. From the next growing season (i.e., May 2007), P. australis grew well, and its growth (about 2-m height) was similar to that in the wild wetland (Fig. 1b). The initial value of below-ground biomass carbon was 978 g C m⁻² obtained using a core sampling method.

Subsequently, soil and groundwater characteristics and carbon fluxes were investigated to estimate the effects of disturbance on the newly introduced wetland ecosystem. Soil redox potential (−193 to −227 mV), which affects CH₄ production, in the GM was similar to that of the wild wetland. Particle size distribution (dominant size: 106–250 μm) of the soil profile in the GM was also similar to that of the wild wetland. Free groundwater characteristics were also investigated. No statistically significant differences (p > 0.05; Mann–Whitney U test) were found for pH (8.11–8.41) and electrical conductance (373–442 mS m⁻¹) between the GM and the wild wetland. Furthermore, no statistically significant difference (p > 0.05; Mann–Whitney U test) in fluxes of CO₂ (9.0–12.8 mg C m⁻² h⁻¹) and CH₄ (0.11–0.23 mg C m⁻² h⁻¹) from the soil between the GM and the wild wetland was observed by a closed chamber method. These results suggested that the wetland ecosystem was properly reconstructed in the GM from physicochemical and biological viewpoints (Suzuki and Endo 2007).

b. Control and measurement of environmental conditions

The experiment for the measurement of the ecosystem CO₂ exchange rates was conducted from 1 January to 31 December 2009. During the experiment, environmental conditions were controlled according to the following procedure. During the entire experimental year, we configured air temperatures in the GM to recreate the daily shifts and seasonal variations in air temperature based on the 5-yr (2001–05) average hourly air temperature at the location (40°53′N, 141°16′E) near the wild wetland. The CGEF was able to control the average air temperature within 1°C of a set value during the plant growing season (Suzuki et al. 2010a); however, during the winter period from December to February, when the above-ground shoots were completely senesced, it was difficult to control the air temperature in the GM to match that in the wild wetland. Relative humidity was controlled to approximately 60%. Light intensity was a passive condition because the GM had glass areas in the roof and walls that admitted natural light, and no electrical lighting was present (Fig. 1a).
The CGEF had a CO₂ scrubber comprising the following: CO₂ canisters that used solid amine as a CO₂ absorber, vacuum pumps; a compressor, hot/cold water pumps and tanks, an accumulator, and a postsorber that removed odors generated from the amine. The CO₂ was scrubbed continuously by switching (adsorb/regenerate) two CO₂ canisters. Scrubbed CO₂ was stored in an accumulator and subsequently transferred to a CO₂ tank. Therefore, the CO₂ scrubber provided continuous CO₂ removal from the GM. When the CO₂ concentration increased above an upper set value (420 ppm), the CO₂ scrubber was turned on. When it decreased below a lower set value (360 ppm), CO₂ was supplied (Suzuki et al. 2010a). In this study, we tried to control air CO₂ concentration in GM to approximately 370 ppm.

Meteorological variables in the GM were continuously measured and automatically logged. As required, air was circulated by fans inside the GM to maintain steady air temperature and CO₂ concentration, which were measured every minute using a platinum resistance sensor (HM119; Oyo Electronics, Ltd., Tokyo, Japan) and an infrared gas analyzer (IRGA) (ZKJ-SZ; Fuji Electric Systems Co., Ltd., Tokyo, Japan), respectively. The CO₂ concentrations were automatically corrected by the software in the IRGA instrument using simultaneously measured air temperature, pressure, and water vapor content. Calibration of the IRGA was automatically carried out once a week throughout the experiment period. The accuracy of CO₂ concentration was 20 ppm, evaluated as the root-mean-square error (RMSE) in the range of the CO₂ concentration in the experiment using standard CO₂ gas. Photosynthetic photon flux density (PPFD) was evaluated with a quantum sensor [PAR-01 (L); Prede Co., Ltd., Tokyo, Japan], which measured global solar radiation from 400 to 700 nm and approximated the spectral band active in photosynthesis; this was done at 30-min intervals above the vegetation canopy (4.5-m height above the ground surface). Soil temperatures at 50 mm below the soil surface (10 replications) were also recorded using a thermal sensor with a built-in datalogger (Stowaway Tidbit; Onset Computer, Pocasset, Massachusetts). The water table was monitored and values were logged automatically every minute using a submersion level detector (SL-710C; JFE Advantech Co., Ltd., Nishinomiya, Japan). Artificial rain was simulated by a sprinkler in the GM that was turned on almost every day from late June to December, and the precipitation was found to be equivalent to the decline in the water table in the wetland due to evapotranspiration.

The site in the GM experienced seasonal variation in environmental conditions (Fig. 2). Mean monthly air temperature ranged from 5.7°C to 19.5°C, with the lowest temperature in January and the highest temperature in August (Fig. 2a). Soil temperature showed a similar trend to air temperature; however, the temperature range in soil (6.8°C–15.2°C) was smaller than that in air (Fig. 2b). The water table was stable and had high values from January to early June. During that period, we could not provide artificial rain because the freshwater process subsystem had technical problems, so the water separated from the air in the GM for controlling air humidity was immediately put back into the wetland soil through
a tube. During late June–December, we provided artificial rain almost once a day, resulting in a large fluctuation in the water table (Fig. 2c). Daily PPFD was highest in April but values were not high during the summer (Fig. 2d). The sunshine durations during the plant growing season at the nearest weather station (40°53’N, 141°16’E) were 224.5, 195.0, 121.8, 113.4, 87.7, 163.7, 173.4, and 84.2 h per month for April, May, June, July, August, September, October, and November, respectively (Japan Meteorological Agency 2009). The decline in the sunshine duration during summer resulted in a lower PPFD per month. The phenomenon may be caused by the cold northeasterly wind, called the “Yamase wind,” which sometimes blows over northeastern Japan and induces clouds from spring to summer (Kanno 1997). Lessmann et al. (2001) investigated the photosynthetic rate against the light intensity and showed that the photosynthetic rate was saturated in the range of PPFD from 1000–1500 μmol m⁻² s⁻¹ for four P. australis populations distributed throughout Europe. In the GM, PPFD attained a level of 1500 μmol m⁻² s⁻¹ above the vegetation canopy (data not shown). The amount of diffuse light at the canopy height of the wetland may be higher in the GM than in the wild wetland because of reflection by the surrounding glass panels of the GM. Air CO₂ concentrations were higher during the nongrowing season for above-ground plants (January–March and December). During that period, we did not need to maintain air CO₂ concentration at ca. 370 ppm because no carbon assimilation was occurring by plant photosynthesis. Moreover, we could not use the CO₂ scrubber because of maintenance work in December 2009. We controlled air CO₂ concentration during the remaining period, resulting in a range from 376 to 433 ppm of the average concentration during the daytime (Fig. 2e). Soil cores (0.3-m depth) were sampled at nine points three times (July, September, and November 2009) in the GM. Each soil core was divided into three depths (0–0.1, 0.1–0.2, and 0.2–0.3 m). The soil carbon concentration was measured (Sumigraph NC22F; Sumika Chemical Analysis Service, Tokyo) and found to be 2.34 ± 0.40% (mean ± SE, n = 81).

c. Estimation of above-ground biomass carbon of P. australis and other minor species

To estimate above-ground biomass carbon of P. australis per shoot, we used the relationship between leaf or culm biomass carbon per shoot and culm length described by a power function, according to the curve-fitting technique by Peng et al. (2008) as follows (Fig. 3):

\[ C_b = a \times L^b, \]  

where \( C_b \) (g C shoot⁻¹) is leaf or culm biomass carbon per shoot, \( L \) is culm length per shoot (m shoot⁻¹), and \( a \) (g C m⁻¹) and \( b \) (dimensionless) are parameters of the function. To estimate the above-ground biomass carbon of P. australis per unit ground area, we measured culm heights of P. australis monthly at 143 fixed points on each approximate 0.5 × 0.5 m² grid, except at boardwalks that were placed above the ground surface to avoid disturbing plants and soil during the measurements and sampling (Fig. 1b). At the same time, we recorded the number of all P. australis shoots in the wetland. Leaf and culm biomass carbon of P. australis per unit ground area (\( B; \) g C m⁻²) were calculated as follows:

\[ B = \frac{N}{143} \sum_{i=1}^{143} C_{b,i}, \]  

where \( N \) is the number of all P. australis shoots, \( S \) (m²) is the ground area of the wetland ecosystem (50.5 m²), and \( C_{b,i} \) (g C shoot⁻¹) is the leaf or culm biomass carbon per shoot \( i \).

To estimate above-ground biomass carbon of the other underlayer species per unit ground area, we monthly set six 0.5 × 0.5 m² quadrats in the wetland. All above-ground parts were harvested in the quadrats every month.
The samples were oven dried (80°C, 72 h) and ground into powder, and then the total carbon content of the samples was analyzed (Sumigraph NC22F; Sumika Chemical Analysis Service, Tokyo, Japan). The aboveground biomass carbon of plants between observation dates was estimated by linear interpolation every day.

d. Estimation of hourly CO2 exchange rates by the CGEF

Air CO2 concentration in the GM fluctuated on a 1-day cycle, resulting from nighttime respiration and daytime photosynthesis. When the CO2 concentration increased past an upper set value (420 ppm), the CO2 scrubber was turned on. When it decreased below a lower set value (360 ppm), CO2 was supplied (Fig. 4a). Hourly CO2 exchange rates were calculated using the data collected every minute. With respect to the sign convention, we considered carbon gain by the ecosystem as positive and carbon loss to the atmosphere as negative.

Without sunlight, air CO2 concentration in the GM increased because of the Re (Fig. 4a). When PPFD was less than 5 μmol m⁻² s⁻¹, hourly Re (g C m⁻² h⁻¹) between time \( t_i \) and \( t_{i+1} \) during nighttime was calculated as follows:

\[
R_{e,i} = \frac{\Delta CO_{2,\text{air}} - \Delta CO_{2,\text{scrubber}}}{S}, \tag{3}
\]

where \( \Delta CO_{2,\text{air}} \) (g C h⁻¹) is the observed change in carbon equivalent CO2 in the air of the GM for 1 h, \( \Delta CO_{2,\text{scrubber}} \) (g C h⁻¹) is the scrubbed carbon equivalent CO2 by the CO2 scrubber compartment for 1 h, and \( S \) (m²) is the ground area of the wetland ecosystem (50.5 m²):

\[
\Delta CO_{2,\text{air}} = \frac{12.0V}{22.4 \times 10^{-3}} \times \frac{273P_i C_i \times 10^{-6} - 273P_{i+1} C_{i+1} \times 10^{-6}}{1013(273 + T_i)} - \frac{273P_{i+1} C_{i+1} \times 10^{-6}}{1013(273 + T_{i+1})}, \tag{4}
\]

and

\[
\Delta CO_{2,\text{scrubber}} = \frac{12.0}{22.4 \times 10^{-3}} \times \sum_k F_{s,k}(C_{GM,k} - C_{s,k}) \times 10^{-6}. \tag{5}
\]

Here, \( V \) (m³) is the volume of air space in the GM (575 m³); \( C_i \) (ppm) is the air CO2 concentration corresponding to time \( t_i \); \( P_i \) (hPa) and \( T_i \) (°C) are air pressure and temperature at time \( t_i \) (h), respectively; \( C_{GM,k} \) (ppm) and \( C_{s,k} \) (ppm) are air CO2 concentrations before and after passing through the CO2 scrubber at time \( t_i \) (min), respectively; and \( F_{s,k} \) (m³ min⁻¹) is the volume of air passed through the CO2 scrubber at time \( t_k \) (min) that is normalized to the standard temperature and pressure of 0°C and 1 atm, respectively. The accuracies of the mass flow controller (JTD920A; Yamatake Corporation, Tokyo, Japan) and the IRGA (ZKJ-SZ; Fuji Electric Systems Co., Ltd., Tokyo, Japan) were 1.8 × 10⁻⁵ m³ min⁻¹ and 20 ppm, respectively. The errors in \( \Delta CO_{2,\text{air}} \) and \( \Delta CO_{2,\text{scrubber}} \) were estimated hourly and minutely based on the error propagation (Taylor 1997) in Eqs. (4) and (5), respectively. In addition, the errors in \( \Delta CO_{2,\text{scrubber}} \) related to the accuracy of hourly CO2 exchange rate only when the CO2 was scrubbed. Then, the error in hourly \( R_e \) was estimated based on the error propagation in Eq. (3).

Hourly daytime \( R_e \) was determined using the predictive relationships developed for nighttime periods under the assumption that daytime \( R_e \) was of similar magnitude and responsiveness as nighttime \( R_e \). The relationship was described by an exponential function using a curve-fitting...
technique (Schedlbauer et al. 2010) based on the data for 8 months together as follows (Fig. 5a):

\[ R_{e,d} = d \exp(\lambda T_i) \tag{6} \]

where \( d \) (g C m\(^{-2}\) h\(^{-1}\)) and \( \lambda \) (°C\(^{-1}\)) are parameters of the function and \( T_i \) is the 1-h average of nighttime air temperature. Nighttime air temperatures in the GM were controlled to exceed the daily maximum air temperatures six times a month to obtain a wide range of the regression variables. Using Eq. (6), we determined hourly daytime \( R_e \) with the 1-h average of daytime air temperature. The errors in \( R_e \) estimates by a nonlinear regression [i.e., Eq. (6)] were obtained as the standard deviation of \( R_e \) calculated by the Monte Carlo method under the assumption that parameter uncertainties of the regression were normally distributed. The changes in air CO\(_2\) concentration under sunlight indicated NEP, which represented the difference between released CO\(_2\) by respiration and assimilated CO\(_2\) by photosynthesis (Fig. 4a). When PPFD was larger than 5 \( \mu \)mol m\(^{-2}\) s\(^{-1}\), hourly NEP (NEP\(_j\); g C m\(^{-2}\) h\(^{-1}\)) between time \( t_j \) and \( t_{j+1} \) during the daytime was calculated as follows:

\[ \text{NEP}_j = (\Delta \text{CO}_2 \text{air} - \Delta \text{CO}_2 \text{scrubber} + \Delta \text{CO}_2 \text{injection})/S, \tag{7} \]

where the former two variables are the same as in the calculation of \( R_{e,d} \) during nighttime and \( \Delta \text{CO}_2 \text{injection} \) (g C h\(^{-1}\)) is the carbon equivalent CO\(_2\) injected into the GM for 1 h:

\[ \Delta \text{CO}_2 \text{injection} = \frac{12.0}{22.4 \times 10^{-3}} \left[ \sum_1 \left( \frac{C_{T,l}}{100} F_{T,l} \right) \right], \tag{8} \]

Here \( C_{T,l} \) and \( F_{T,l} \) (m\(^3\) min\(^{-1}\)) are the injected CO\(_2\) concentration and volume (normalized at 0°C and 1 atm) into the GM at time \( l \) (min). The accuracies of the mass flow controller (F-202S; OVAL Corporation, Tokyo, Japan) and the IRGA (IR-200; Yokogawa Electric Corporation, Tokyo, Japan) for these measurements were 1.0 \( \times \) 10\(^{-4}\) m\(^3\) min\(^{-1}\) and 3.7%, respectively. [Different instruments than before were used for the measurements of CO\(_2\) injected because of its higher concentration (ca. 95% CO\(_2\)).] The errors in \( \Delta \text{CO}_2 \text{injection} \) were estimated minutely based on the error propagation in Eq. (8). These errors related to the accuracy of hourly CO\(_2\) exchange rate only when the CO\(_2\) was injected. Then, the errors of hourly NEP were estimated based on the error propagation in Eq. (7). Hourly NEP was estimated by a rectangular hyperbola function. Then, a regression was obtained between hourly NEP and hourly PPFD for each month from April to November as follows:

\[ \Delta \text{CO}_2 \text{injection} = \frac{12.0}{22.4 \times 10^{-3}} \left[ \sum_1 \left( \frac{C_{T,l}}{100} F_{T,l} \right) \right], \tag{8} \]

where \( I \) (mmol m\(^{-2}\) h\(^{-1}\)) is hourly PPFD; \( \alpha_{\text{NEP}} \) (mol C mol\(^{-1}\) photon) is the initial slope of the rectangular hyperbola, which is also called “the apparent quantum yield” or “the maximum light use efficiency”; \( \text{NEP}_{\text{max}} \) (g C m\(^{-2}\) h\(^{-1}\)) is the maximum NEP at the
saturated light level; and \( R \) (g C m\(^{-2}\) h\(^{-1}\)) is the NEP-axis intercept when hourly PPFD equals zero, which is also called “the dark respiration rate.” The errors in NEP estimates by nonlinear regressions (April–November) [i.e., Eq. (9)] was obtained by the same method with \( R_e \).

GPP was assumed to equal zero when PPFD was less than 5 \( \mu \text{mol m}^{-2} \text{s}^{-1} \). Hourly GPP was defined as hourly NEP minus hourly \( R_e \). Then, hourly \( R_e \), NEP, and GPP values were each summed to determine not only daily but also annual \( R_e \), NEP, and GPP, respectively (Fig. 4b). The errors in hourly GPP were estimated as zero during nighttime when PPFD was less than 5 \( \mu \text{mol m}^{-2} \text{s}^{-1} \). On the other hand, those during daytime were estimated based on the errors in hourly \( R_e \) and NEP. Hourly GPP was also estimated by the similar function with NEP as follows (Fig. 6):

\[
GPP = \frac{\alpha_{\text{GPP}} I_{\text{GPP}} \text{max}}{\alpha_{\text{GPP}} I + \text{GPP} \text{max}}.
\]

\[ (10) \]

\textbf{e. Gap filling of CO}_2 \text{ exchange rates}

Unfortunately, data gaps are inevitable when constructing long data records. In our study, gaps were caused by an accidental disconnection of a signal cable, the disturbance in air CO\(_2\) concentration due to entry of researchers into the GM for measuring and sampling plants and soil, and facility maintenance. Falge et al. (2001) suggested three representative gap-filling methods, the mean diurnal variation method, the nonlinear regression analysis, and the lookup-tables method. In this study, the former two methods were applied for gap filling of data of CO\(_2\) exchange rates. The gap filling was performed separately for \( R_e \) and NEP. Data were divided into two groups to fill gaps in the hourly \( R_e \); one was the growing season of above-ground plants (April–November) and the other was for the winter months (January–March and December). For the growing season, data gaps of a few to several hours were filled with hourly \( R_e \) by Eq. (4). For the winter months, data gaps in hourly \( R_e \) for both day and nighttime were filled by linear interpolation. Average hourly \( R_e \) in December 2009 was used to fill the large gaps during the facility maintenance from 13 to 26 December 2009. To fill daytime NEP gaps, we used the relationship between NEP and PPFD expressed as Eq. (9) for each month.

In our study, the data gaps occurred during 26.8% of the experimental year. Other studies in the wild have reported data gaps of 39% and 52% during April and September in 1999 and 2000, respectively (Harazono et al. 2003); about 29% and 41% at two sites (Guo et al. 2009); 22.0%, 30.0%, and 17.4% at three sites (Yan et al. 2010); and about 44% and 46% at two sites (Yan et al. 2010). The lower ratio of gap filling at the CGEF was expected, as it was a controlled environment rather than a facility out in the wild.

\textbf{3. Results and discussion}

\textit{a. Effects of environmental conditions on the CO}_2 \text{ exchange rates}

Temperature is often responsible for most of the variability in CO\(_2\) fluxes (Bridgham and Richardson 1992; Bubier et al. 2003; Marsh et al. 2005). In our study, nighttime \( R_e \) increased exponentially with air temperature \((r^2 = 0.732; \text{Fig. 5a})\), indicating a predictor of daytime \( R_e \). The coefficient of determination between nighttime \( R_e \) and air temperature for 8 months together was higher than those for each month level. Because nighttime air temperature seldom reaches the maximum air temperature obtained during the daytime in fields, a prediction of daytime \( R_e \) in the higher temperature region must be derived from the extrapolation by the eddy covariance technique. In contrast, our prediction of nighttime \( R_e \) did not depend on the extrapolation in the higher temperature region to make the regression because of the daily maximum air temperature being realizing by artificial warming during the nighttime. On the other hand, regressions were not successful for the relationship between hourly \( R_e \) and hourly PPFD for the whole year \((r^2 = 0.096; \text{Fig. 5b})\). Guo et al. (2009) reported a distinct relationship between fluxes and water table. On the contrary, Bonneville et al. (2008) reported no significant relationship between them. Also, Jones et al. (1999) found no correlation between the temperature in winter and CO\(_2\) flux. In our study, data gaps of hourly \( R_e \) were filled by linear interpolation or the average value in December 2009 (winter period) because no distinct relationship during winter was observed between \( R_e \) and environmental variables such as air and soil temperatures, PPFD, and the water table. There was also no distinct relationship between \( R_e \) and water table from April to November. During winter in our study \( R_e \) was relatively smaller than the plant growing season. Therefore, estimated \( R_e \) errors because of gap filling would contribute less toward the annual \( R_e \).

Hourly GPP increased with hourly PPFD by a rectangular hyperbola function (Fig. 6). The values of both parameters \((\alpha_{GPP} \text{ and } \text{GPP}\text{max})\) for the function increased initially, reached a peak, and then decreased at the end of the growing season (Table 1). Some studies have reported that the variation in parameters among different growth periods may be caused by changes in plant characteristics such as physiological activity, leaf thickness, leaf area index, and environmental conditions (Bubier et al. 2003; Harazono et al. 2003; Zhao et al. 2006, 2009). The parameter \( \text{GPP}_{\text{max}} \) had a positively linear relationship with air temperature \((r^2 = 0.799)\). The regression curve for
FIG. 6. Rectangular hyperbolic response curves between hourly GPP and hourly PPFD from April to November 2009.
NEP was also obtained for each month ($r^2$ varied from 0.161 to 0.637). Parameters for the function between hourly NEP and hourly PPFD showed similar trends with those for the GPP function. Hourly NEP increased with hourly gross primary productivity ($r^2 = 0.812$; Fig. 7a). On the other hand, there was no distinct relationship between hourly NEP and $R_e$ ($r^2 = 0.005$; Fig. 7b). This could be understood from the fact that NEP depended on both light and temperature, while $R_e$ depended only on temperature. Therefore, the ecosystem CO$_2$ balance was strongly affected not by the output of CO$_2$ by emission through plant and soil respiration but by its input by photosynthesis.

b. Diurnal and seasonal patterns in CO$_2$ exchange rates and above-ground plant biomass

Diurnal CO$_2$ exchange rates showed distinct courses—that is, CO$_2$ uptake during the daytime and CO$_2$ release during the nighttime (Fig. 4b), although we showed data for only 2 days during the whole year. Hourly $R_e$ corresponded with temperature, and showed a peak around 12 h and had lower fluxes during the night. After sunrise, hourly GPP began to increase with PPFD and reached a peak at around 12 h. Then, GPP decreased in the afternoon and was zero during the night (Fig. 4b). Hourly NEP became negative (carbon source) in the early morning and evening as well as during the night, and then showed positive values (carbon sink) as PPFD increased sufficiently (Fig. 4b).

Daily CO$_2$ exchange rates exhibited clear seasonal patterns corresponding to the environmental and plant conditions (Figs. 8a–c). Daily $R_e$ ranged from $-0.06$ to $-3.58$ g C m$^{-2}$ day$^{-1}$, increased from April to August, and then decreased from September to November (Fig. 8c). A small amount of CO$_2$ emission was observed during the nongrowing season of plants. Cumulative $R_e$ was $-53.7$ and $-437.9$ g C m$^{-2}$ during the nongrowing and growing seasons, respectively, and the emissions during the nongrowing season accounted for 10.9% of the annual emissions. Daily GPP ranged from 0 to 6.48 g C m$^{-2}$ day$^{-1}$ (Fig. 8a) with larger variations relative to $R_e$ because photosynthesis depended on light intensity as well as temperature (Fig. 6). GPP occurred from April to November. The peak value of daily GPP was observed around mid-August, which is the active period for vegetation. Plants emerged and the above-ground biomass of plants started to increase from early April, peaked in September, and mostly senesced by late November. The seasonal GPP trend coincided with the above-ground

<table>
<thead>
<tr>
<th>Month</th>
<th>$GPP_{\text{max}}$ (g C m$^{-2}$ h$^{-1}$)</th>
<th>$\alpha_{\text{GPP}}$ (mol C mol$^{-1}$ photon)</th>
<th>n</th>
<th>$r^2$</th>
</tr>
</thead>
<tbody>
<tr>
<td>April</td>
<td>0.168</td>
<td>0.0185</td>
<td>305</td>
<td>0.379</td>
</tr>
<tr>
<td>May</td>
<td>0.339</td>
<td>0.0262</td>
<td>364</td>
<td>0.552</td>
</tr>
<tr>
<td>June</td>
<td>0.471</td>
<td>0.0361</td>
<td>333</td>
<td>0.657</td>
</tr>
<tr>
<td>July</td>
<td>0.559</td>
<td>0.0493</td>
<td>317</td>
<td>0.587</td>
</tr>
<tr>
<td>August</td>
<td>0.834</td>
<td>0.0477</td>
<td>308</td>
<td>0.661</td>
</tr>
<tr>
<td>September</td>
<td>0.368</td>
<td>0.0754</td>
<td>311</td>
<td>0.261</td>
</tr>
<tr>
<td>October</td>
<td>0.259</td>
<td>0.0488</td>
<td>311</td>
<td>0.285</td>
</tr>
<tr>
<td>November</td>
<td>0.171</td>
<td>0.0292</td>
<td>223</td>
<td>0.267</td>
</tr>
</tbody>
</table>

### Table 1. Nonlinear parameter estimates for rectangular hyperbolic response curves between hourly GPP and hourly PPFD.

![Fig. 7. Relationship between hourly NEP and (a) hourly GPP, and (b) hourly ecosystem $R_e$ of the wetland ecosystem in the GM.](image)
plant biomass. Daily NEP ranged from $-1.10$ to $3.34$ g C m$^{-2}$ day$^{-1}$ (Fig. 8b) because of the balance between daily GPP and daily $R_e$. The maximum daily NEP was observed in mid-August. NEP started to show positive values around late April to early May and negative values around late October to early November, indicating that the wetland switched from a net CO2 sink to a net CO2 source.

Above-ground biomass carbon of $P. australis$ per shoot was estimated by a power function (Figs. 3a,b). Using the regression curves, above-ground plant biomass carbon was evaluated nondisruptively for $P. australis$-dominated wetlands. Plant shoots started to grow from early April and had fully senesced by late November (Fig. 8d). Above-ground biomass of $P. australis$ increased until early August and then remained relatively constant until senescence. Above-ground biomass of other species also showed a trend similar to that of $P. australis$. Total peak biomass carbon was $449.4$ g C m$^{-2}$ in mid-September (leaf and culm biomass carbon of $P. australis$ and above-ground biomass carbon of other species were $146.2$, $172.6$, and $130.6$ g C m$^{-2}$, respectively; Fig. 8d). The daily CO2 exchange rates depended on above-ground plant biomass as well as environmental variables ($r^2$ varied from 0.155 to 0.494; Fig. 9). Absolute values of daily GPP, NEP, and $R_e$ was evaluated nondisruptively for $P. australis$-dominated wetlands. Plant shoots started to grow from early April and had fully senesced by late November (Fig. 8d). Above-ground biomass of $P. australis$ increased until early August and then remained relatively constant until senescence. Above-ground biomass of other species also showed a trend similar to that of $P. australis$. Total peak biomass carbon was $449.4$ g C m$^{-2}$ in mid-September (leaf and culm biomass carbon of $P. australis$ and above-ground biomass carbon of other species were $146.2$, $172.6$, and $130.6$ g C m$^{-2}$, respectively; Fig. 8d). The daily CO2 exchange rates depended on above-ground plant biomass as well as environmental variables ($r^2$ varied from 0.155 to 0.494; Fig. 9). Absol
increased with above-ground plant biomass during development of the biomass. Furthermore, the values rapidly decreased while the plants continued to senesce. We found that the seasonal trend of daily CO₂ exchange rates was affected by plant biomass more strongly at the senesced stage than the growing stage.

c. Annual CO₂ exchange rates and comparison with other studies

In our study, the accumulated daily NEP values provided an annual carbon budget of the *P. australis*-dominated ecosystem in the GM of 64.2 g C m⁻² yr⁻¹, which resulted from annual CO₂ uptake of 555.8 g C m⁻² yr⁻¹ (annual GPP) and annual CO₂ release of −491.6 g C m⁻² yr⁻¹ (annual Re). Thus, the wetland ecosystem behaved as a CO₂ sink (Table 2). The errors in NEP, Re, and GPP were estimated as ±19.2, ±15.6, and ±17.0 g C m⁻² yr⁻¹ for a whole year, being ±16.7, ±12.5, and ±17.0 g C m⁻² during 8 months for the growing season and ±9.4, ±9.4, and zero g C m⁻² during 4 months for the winter period (Table 2). Contributions of the errors in ∆CO₂,air, ∆CO₂,scrubber, ∆CO₂,initiation, and Re and NEP by nonlinear regressions were 97.8, 1.0 × 10⁻², 0, 2.2, and 0% to the total errors in the annual Re and 65.0, 6.8 × 10⁻⁴, 7.3 × 10⁻⁵, 0, and 35.0% to those in the annual NEP. The errors in ∆CO₂,scrubber and ∆CO₂,initiation were considerably smaller than those in ∆CO₂,air because CO₂ was scrubbed or injected at nighttime or daytime, respectively, during only actively growing periods.

The CO₂ exchange rates in our study were comparable with those in other studies conducted in *P. australis*-dominated wetlands using the eddy covariance technique and the combined method of internal gas pressures and flow measurements and harvesting (Table 2). The annual NEP in our study was consistent with that in *P. australis*-dominated wetlands at similar latitude in Shenyang, China (Zhou et al. 2009). On the other hand, the annual GPP in our study was 2.3-fold smaller than that in Shenyang. We considered the reasons for this. First, GPP positively depended on above-ground biomass (Fig. 9). In addition, the above-ground biomass in our study was 1.5-fold smaller than that in Shenyang (Table 2). Second, GPP increased with PPFD and attained a maximum value (GPPₘᵃₓ) (Fig. 6). Although there were no data on GPPₘᵃₓ in Shenyang, NEPₘᵃₓ values for all months there were higher than those in our study. In addition, monthly PPFD for the above-ground active period was higher in Shenyang (952 mol m⁻² month⁻¹) than that in our study (467 mol m⁻² month⁻¹). Thus, physiological aspects of *P. australis* and the light condition raised the GPP in Shenyang. Third, GPPₘᵃₓ had a positive linear relationship with air temperature ($r^2 = 0.799$). Average air temperature for the above-ground active period was higher in Shenyang (17.9°C) than that in our study (14.2°C). Therefore, warmer temperature enhanced the GPP in Shenyang.

The annual Re was 2.4-fold smaller in our study than that in Shenyang (Table 2). As well, the soil respiration rate in the Vejlerne Reserve, Denmark (Brix et al. 2001), was comparable to the Re for the winter period in our study and in Shenyang because the above-ground shoots were senesced in those wetlands and Re was the same as the soil respiration rate for winter. The soil respiration rate for winter in our study was smaller than those rates in the Vejlerne Reserve and in Shenyang (Table 2). This respiration rate consists of below-ground autotrophic and heterotrophic respiration rates; therefore, we expected it to depend on below-ground biomass and soil organic carbon concentration. Soil organic carbon concentration in our study (2.34%) was similar to that in Shenyang (2.2%–2.5%). Therefore, the smaller below-ground biomass in our study (978 g C m⁻²) than in the other sites (Denmark: 3052 g C m⁻²; China: 1325 g C m⁻²) may result in lower soil respiration rate for winter, although there were no data on soil organic carbon concentration in the Vejlerne Reserve. We suggest that it is useful to investigate the above- and below-ground plant biomass and soil organic carbon concentration as well as climatic conditions for the comparison of CO₂ exchange rates with other sites.

4. Conclusions

The annual CO₂ exchange rates in our study were comparable with those of other wetland ecosystems obtained using the eddy covariance technique and the combined method of internal gas pressures and flow measurements and harvesting. The annual CO₂ budget was found to be 64.2 ± 19.2 g C m⁻² yr⁻¹, indicating that the wetland ecosystem behaved as a CO₂ sink for the entire year. The CO₂ balance of *P. australis*-dominated wetland was strongly affected not by the output of CO₂ through plant and soil respiration but by its input by photosynthesis. It is also important to investigate whether or not the wetland ecosystem will behave as a CO₂ sink for a long period. Such an investigation is linked to clarification of the impact of wetland ecosystems on the global carbon budget of terrestrial ecosystems. For that purpose, multiyear studies are necessary to understand the temporal variations in carbon dynamics and feedback systems among various biotic and abiotic factors. The CGEF is very advantageous for studying the effects of temperature and atmospheric CO₂ concentration on carbon dynamics at the ecosystem level because the facility is large and has the ability to produce the conditions similar to those induced by global warming. Furthermore,
## Table 2. Comparison of this study with other studies on CO2 exchange rates of *P. australis*-dominated wetlands.

<table>
<thead>
<tr>
<th>Location</th>
<th>Method</th>
<th>NEP (g C m⁻² per given period)</th>
<th>Rₚ (g C m⁻² per given period)</th>
<th>GPP (g C m⁻² per given period)</th>
<th>Biomass (g C m⁻²)</th>
<th>Soil carbon concentration (%)</th>
<th>Period</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aomori, Japan (40°53'N, 141°16'E)</td>
<td>Closed experiment facility</td>
<td>64.2 ± 19.2</td>
<td>−491.6 ± 15.6</td>
<td>555.8 ± 17.0</td>
<td>449[^a]</td>
<td>2.34 ± 0.40</td>
<td>January–December 2009</td>
<td>This study</td>
</tr>
<tr>
<td></td>
<td></td>
<td>−53.7 ± 9.4</td>
<td>−53.7 ± 9.4</td>
<td>0</td>
<td>978[^b]</td>
<td></td>
<td>Above-ground active period</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Winter period (January–March and December)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Shenyang, China (41°08'N, 121°54'E)</td>
<td>Eddy covariance</td>
<td>65</td>
<td>−120[^c]</td>
<td>1300[^e]</td>
<td>657[^d,a]</td>
<td>2.2–2.5</td>
<td>January–December 2005</td>
<td>Zhou et al. (2009)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>400[^c]</td>
<td>−900[^d]</td>
<td>1300[^e]</td>
<td></td>
<td></td>
<td>Above-ground active period</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>−300[^c]</td>
<td>−300[^d]</td>
<td>0</td>
<td>1325[^d,b]</td>
<td></td>
<td>Winter period (January–March and November–December)</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>No data</td>
<td>−400[^e]</td>
<td>1200[^f]</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td></td>
<td>No data</td>
<td>−100[^e]</td>
<td>0[^f]</td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

[^a]: Above-ground biomass.  
[^b]: Below-ground biomass.  
[^c]: Values were quantified on graphs using a vernier caliper.  
[^d]: Values were calculated from the dry weight data as 45% of carbon concentration.  
[^e]: Soil respiration rate.  
[^f]: Net primary productivity.
transfer model is essential to consider the carbon dynamics in an ecosystem. A mathematical compartment model consisting of photosynthesis and decomposition processes with nine reservoir compartments (i.e., three plant, two litter, three soil, and one groundwater compartments) was developed to estimate carbon dynamics in the wetland ecosystem (Suzuki et al. 2008). Data obtained in the CGEF in this study are highly valuable to determine the parameters in the compartment model because of less missing carbon and they will be helpful for other carbon dynamics studies in wetland ecosystems.

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