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Design and Testing of a Novel Gas Exchange Chamber

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A transparent closed-system, non-steady-state chamber was developed that allowed measurements of net carbon exchange (NCE). The chamber was 0.85-m x 0.85-m with a height of 0.25-m and total weight of 10.9 kg. The sides were constructed of clear Plexiglas while the top of the chamber was Propafilm-C. Minimizing alterations to the chamber microclimate were of primary concern. To quantify alterations to the microclimate, the chamber was tested for two years over a range of climatic, leaf area, and biomass levels on an ungrazed tallgrass prairie near Manhattan, KS. The chamber had a minimal effect on microclimate; average chamber air temperature increased 2.9° C over 40 s, while chamber pressure increased only 0.3 Pa, and photosynthetically active radiation attenuation was 10%. Also, a method for selecting the appropriate model (linear or quadratic) to calculate the rate change of CO2 and water vapor from time-series data collected by the chamber was developed. The method utilized a computer program that determined whether the quadratic model, based on shape of the CO2 vs. time curve. Using this method, the quadratic flux estimate was utilized for 80.1% of all NCE flux calculations, 14.2% of the RE flux calculations, and 99.5% of water vapor flux calculations.

Key words: Key Words: Net Carbon Flux, Closed Gas Exchange Chamber, Water Vapor Flux, Tallgrass Prairie

INTRODUCTION

Field observations of carbon and water vapor fluxes are critical to understanding how ecosystems respond to different land management practices. Advantages of using chambers over other micrometeorological techniques (i.e. eddy correlation) include the ability to measure responses at the plot scale rather than field scale, replicate treatments in relatively small, uniform areas, and portability. Chamber portability is an essential design criterion to allow easy maneuverability among research plots. Early chamber designs were large, requiring forklifts or tractors to position or relocate them (Reicosky and Peters, 1977). Recent designs have stressed lightweight materials that are manipulated more easily (Risch and Frank 2006). However, chambers can alter the microclimate and architecture of the canopy.

Microclimate perturbations caused by chambers have been well addressed in the literature and include the creation of pressure gradients and alteration of CO_2 gradients, turbulence regimes, air and leaf temperatures, radiation attenuation, and vapor pressure deficit (Wagner and Reicosky, 1992; Steduto et al., 2002; Bremer and Ham, 2005). Alterations to the chamber microenvironment are the main objections to using closed chamber methods. Minimizing microclimate modifications within the chamber has been accomplished by actively controlling air temperature and humidity within the chamber [6] and by performing measurements of short duration (Steduto et al., 2002).

When using the non-steady state method, the rate of change in concentration of CO_2 ($\delta CO_2/\delta t$) and water vapor ($\delta w_c/\delta t$) must be calculated to estimate gas exchange. Both linear and quadratic models have been used to calculate $\delta CO_2/\delta t$ and $\delta w_c/\delta t$ (Wagner et al., 1997; Risch and Frank, 2006). Wagner and Reicosky (1992) found that even when $r^2>0.97$, CO_2 flux was underestimated by 10%. Wagner et al. (1997)reported an average difference of 47% between linear and quadratic evapotranspiration calculations. In non-steady state systems, Wagner et al. (1997) and Studeto et al. (2002) stated that the quadratic model could be considered the most effective. Furthermore, a non-linear least squares method is utilized for calculating fluxes from data collected by the LI-8100 (LiCor Industries, Lincoln, NB). The goal with each model is to calculate the slope of $\delta CO_2/\delta t$ and $\delta wc/\delta t$ at the moment chamber conditions approximate ambient or before the chamber has significantly changed canopy microclimate (i.e., the very start of the measurement, t=0).

Other chamber designs have been utilized on rangelands (Angell and Svejar, 1999; Wilsey et al., 2002; Risch and Frank, 2006). However, some of the chambers are designed to remain on the plot being vented by doors between readings and are heavy (> 20 kg) while other chambers are climate-controlled, but remain on the plot for up to 180 s during a reading. An objective of this research was to design and fabricate a lightweight, portable chamber to measure CO_2 and water vapor fluxes of rangeland in a closed, non-steady-state environment with minimal alterations to canopy microclimate. A transparent chamber was used to measure net carbon exchange (NCE) in the light and a tight fitting opaque box was placed over the chamber to measure ecosystem respiration (R_E) in the dark. Field testing examined: 1) how quickly could measurements be made, 2) what was the temperature, pressure, vapor pressure deficits (VPD), and light effects, and 3) what types of correction for leaks and the buildup of water vapor are required. Another objective was to develop a method of selecting the most appropriate model (linear or quadratic) to calculate CO_2 and water vapor fluxes.

METHODS AND RESULTS

Chamber Design

The chamber sample area was 0.85-m x 0.85-m with a height of 0.25-m and total weight of 10.9 kg. Sides were constructed of 4.59-mm acrylic-FE (Acrylite, Cyro Industries, Clifton, NJ) (Figure. 1). The top was made from heat-stretched Propafilm-C (ICI Americas Inc., Wilmington, DE) with high thermal and visible transmittance (Hunt, 2003). A closed-cell foam gasket (Nomapack-WS, NomacoInc, Zebulon, NC) provided a tight seal between the bottom edges of the chamber sides and the soil collar. The soil collars were constructed of 5.1 cm x 7.6 cm x 0.5 cm angle iron that were pressed into the soil until 5 cm was exposed above ground. Two fans (700 L min^{-1}, BD 12A3, Comair Rotron, San Diego, CA) circulated air through a perforated plenum that was attached near the base of the entire chamber. The plenum was 2.1 cm x 4.3 cm electric raceway (PN10L08V, Wiremold, West Hartford, CT) with two rows of 1.8 mm holes drilled 1.3 cm from the top and bottom of the raceway and spaced 1.3 cm apart. Two additional fans (V571M, Micronel, Fallbrook, CA) were positioned near the top of the chamber, opposite the Comair fans, to promote heat transfer across the propafilm and increase air mixing. Four vents 0.25-m long by 14.3-mm inside diameter were installed on the chamber walls to equilibrate chamber and atmosphere pressure.

A closed path infrared gas analyzer (IRGA, LI-840, Li-Cor Industries, Lincoln, NE) was used to measure CO_2 and water vapor concentrations every second during a test. A continuous air sample was supplied to the IRGA at 1.0 L min^{-1} by a rotary pump (model 50095, Thomas Pumps, Sheboygan, WI); rotary pumps minimize pressure fluctuations in the optical bench of the IRGA. Air was sampled from the plenum to obtain a well-mixed sample. A 0.4-mm-diameter thermistor (10K3MCD1, Betatherm Corp., Shrewburg, MA) placed inside the plenum, 5 cm below one of the Comair Rotron fans, measured chamber air temperature. Photosynthetically active radiation (PAR) flux was measured outside the chamber with a quantum sensor (LI-190, Li-Cor Inc., Lincoln, NB). A CR10X (Campbell Scientific, Logan, UT) logged data from all instruments at 1 Hz. Both the IRGA and CR10X were mounted on a 3.2-mm-thick aluminum plate that was painted flat gray. A sun shield was mounted over the top of the instruments to further shield the instruments from radiation. The system was powered by a 12-V, 12 amp-hr battery, which could run the chamber for 2 hours after a full charge.

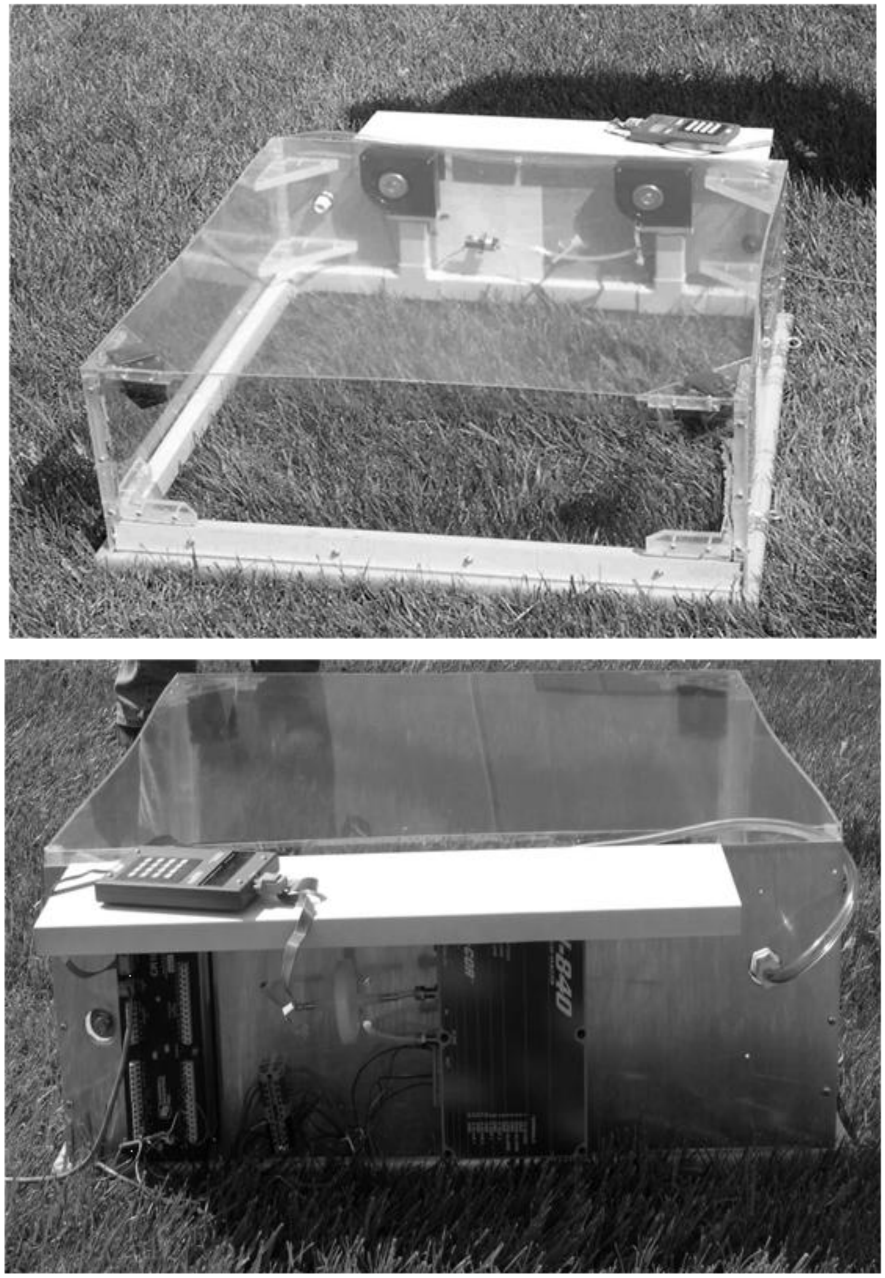

Figure 1.Picture taken of the chamber from opposite the IRGA. The light colored raceway along chamber bottom is air plenum. The foam strip along bottom seals the chamber to the soil frame. Picture taken from IRGA side of chamber showing the IRGA and the CR10X.

Field setup

The chamber was tested in an ungrazed, annually burned pasture located in the Rannells Flint Hills Prairie Preserve, 5 km south of Manhattan, KS (39.11°N, 96.34°W, 324 m above sea level) that was burned during the last 10 days of April. The dominant vegetation consisted of the native C_4 species *Andropogon gerardii* Vitman and *Sorghastrumnutans* (L.) Nash while subdominants included *A. scoparius* Michx. and*Boutelouacurtipendula* (Michx.) Kunth. The remainder of the vegetation consisted of various sedges (C_3), and C_3 forbs including *Vernoniabaldwinii* (Small) Schub.,*Ambrosia psilostachya* DC., *Artemesialudoviciana*

Nutt., and *Psoreleatenuiflora* var. *floribunda* (Nutt.) Rydb. The soil was a Dwightsilty clay (Fine, smectitic, mesicTypicNatrustolls) on a 1-3% slope. Average annual precipitation (1971-2000) was 884 mm with 542 mm occurring from May through September.

Eighteen plots, 0.85 m x 0.85 m, were established in mid-May of 2005 and 2006. At six different dates during June through August, three of the plots were clipped, resulting in the chamber being used over a wide range of canopy sizes and biomass levels. Net carbon exchange was measured with the chamber exposed to ambient light. All readings occurred between 1030-1500 (DST) during clear days. Readings were taken on 24 dates between June to September and June to October in 2005 and 2006. Chamber measurements were initiated by recording ambient conditions above the canopy for 20 seconds. Data acquisition was then paused for five seconds while the chamber was positioned over the canopy and placed onto the soil frame. Following the five-second pause, data acquisition resumed for a 40-second period with the chamber positioned over the canopy. Following each reading, the chamber was removed from the canopy and the CO_2 and water vapor concentrations and air temperature within the chamber were allowed to equilibrate with ambient conditions.

Environmental tests

Leak Test

Considerable effort was made in the design and fabrication of the chamber to minimize entry of ambient air into the chamber. The sheets of Plexiglas that formed the chamber walls were fused with trichloroethylene and further sealed with silicone at each joint. The Propafilm top was sealed to the chamber walls with clear packing tape. However, completely eliminating the exchange of air between the chamber and the ambient environment was extremely difficult. To quantify this potential source of error in the flux calculations, leak tests were conducted prior to each sampling date. This test consisted of positioning the chamber on a flat aluminum surface, in the field, and injecting 24 ml of pure CO_2 into the chamber to create a CO_2 concentration gradient >100 $\mu l\ l^{-1}$ across the chamber walls. This gradient is considerably greater than those created during normal chamber readings (average measurement gradient was typically 35 $\mu l\ l^{-1}$). The gradual decrease of CO_2 within the chamber was monitored and used to calculate a leakage rate of CO_2 from the chamber. The CO_2 leakage rates averaged 0.12 $\pm$ 0.005 $\mu mol\ m^{-2}\ s^{-1}$ (mean $\pm$ SE, n=261) and 0.09 $\pm$ 0.02 $\mu mol\ m^{-2}\ s^{-1}$ (n=401) over the 2005 and 2006 growing seasons, respectively. These rates were considered negligible and the final flux calculations were not corrected for leaks.

Pressure Test

Increased pressure within a chamber has the ability to partially suppress the efflux of CO_2 from the soil, which could create an artificially high net canopy CO_2 flux (Owensby et al., 1997; Lund et al., 1999; Bremer and Ham, 2005). Because the chamber is operated in a closed state, pressure increases could result from either air handling or evapotranspiration (ET). Air moving over the soil surface in response to the forced ventilation of the mixing fans increases pressure and chamber designs that have excessive air flow likely depress soil CO_2 flux. The perforated air plenum was designed to evenly distribute air throughout the chamber and reduce air speed. Evapotranspiration adds additional water vapor to the chamber headspace. Boyle's law predicts pressure must increase as additional gas is added to a sealed, constant volume chamber. Pressure increases were attenuated by venting the chamber to the atmosphere. The initial chamber design incorporated a single vent constructed from flexible tubing measuring 0.45-m long by 14.3-mm inside diameter. However, a numerical model of the chamber showed that ET from large, actively growing canopies might cause pressure increases. Thus, the chamber was redesigned incorporating four vents 0.25-m long by 14.3-mm inside diameter.

Chamber pressure was measured in both the laboratory and field. In the lab, a differential pressure transducer (Furness PPC-500, Indian Trail, NC) measured pressure within the empty chamber that was placed on an elevated platform with nine sampling points positioned in a single line down the center of the chamber. The pressure transducer was connected to one of the nine sampling points and while the chamber was operated in measurement mode the pressure at each sampling point was recorded for 60 seconds. Field measurements also utilized a differential pressure transducer (Setra 264, Setra, 0 – 0.1 in H_2O range, Boxborough, MA). One sampling tube was suspended near the center of the chamber approximately 50-mm above the soil surface, while the other tube was positioned outside the chamber within the grass canopy at approximately the same height.

Average chamber pressure measured in the lab was 0.58 $\pm$ 0.08 Pa (mean $\pm$ SE, n = 9) above ambient. The average chamber pressure recorded during field sampling was 0.32 $\pm$ 0.04 Pa (mean $\pm$ SE, n = 418) above ambient. This very minimal increase in chamber pressure during a reading (Figure 2) was less than that reported from many other chamber designs. The venting system described above, and the perforated air plenum, minimized pressure increases caused by ET and air movement in the chamber to the extent that corrections to flux calculations were not indicated.

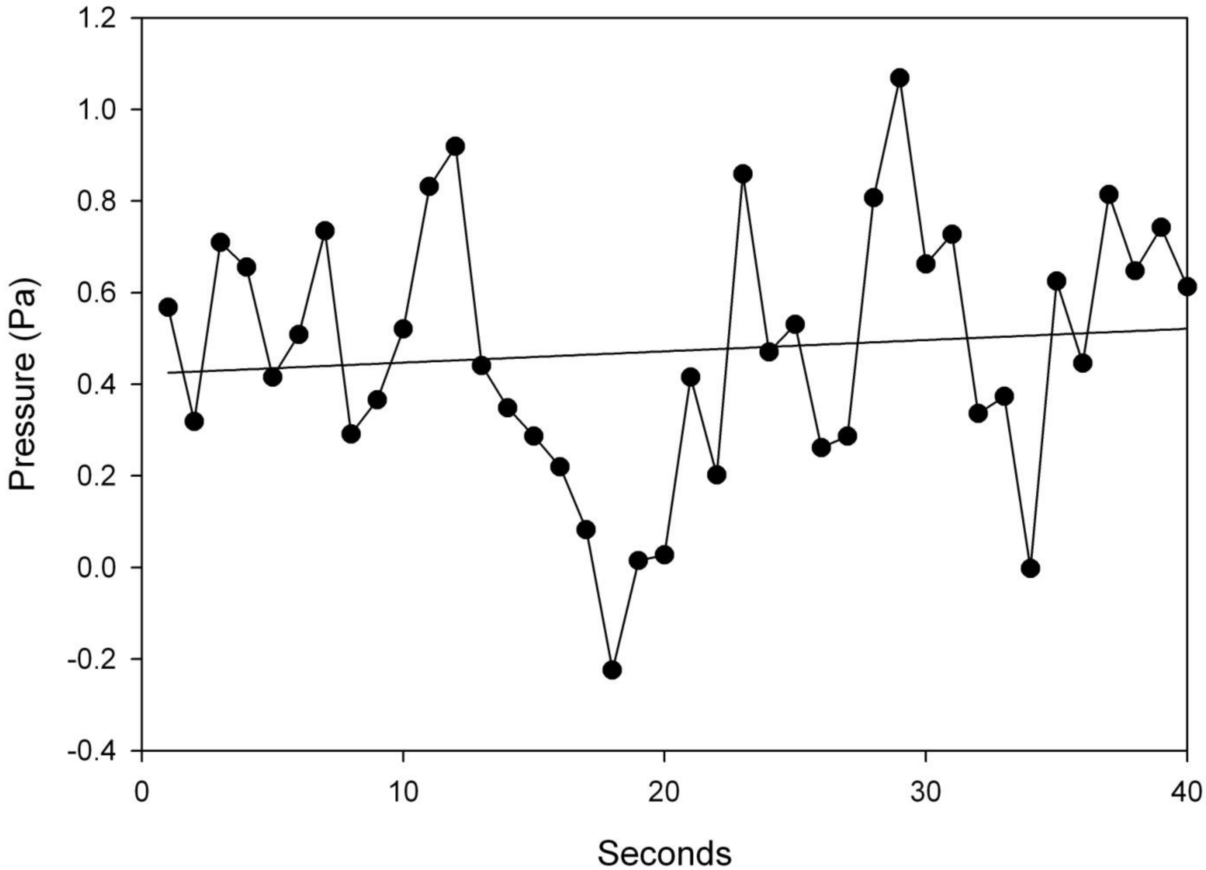

Figure 2. A typical example of chamber pressure observed during the 40 s measurement in the field. The solid black line represents the linear regression equation ($y = 0.0025x + 0.4226$ $r^2 = 0.01$).

Temperature test

Increased temperature can modify carboxylation rates by altering ribulose-1-5-bisphosphate carboxylase (Rubisco) activity [12]. Rubisco catalyzes the reaction that reduces CO_2 to carbohydrate during photosynthesis. Altering the rate of carboxylation directly affects measurements of NCE. One of the main reasons for attempting a short duration measurement (40 s) was to minimize heating. Throughout both years of the study, the average temperature increased 2.9 $\pm$ 0.024°C (mean $\pm$ SE, n = 883) during the 40 s sampling interval (Figure 3). Chamber temperature increases of 2-4°C are commonly reported in the literature (Wagner and reicosky, 1992; Steduto et al., 2002).

Photosynthetically Active Radiation Attenuation

Chamber materials were selected to minimize attenuation of photosynthetically active radiation (PAR). Photosynthetically active radiation (400-700 nm) is the driving force of photosynthesis providing energy to photosystems I and II. Artificial reductions in PAR directly affect measurements of canopy photosynthesis.

Attenuation of PAR by the chamber was determined using a 0.5 m light bar with six quantum sensors (LQS506-SUN, Apogee, Logan, UT). The measurements occurred on a level surface during a cloudless day. A detailed map of PAR attenuation was constructed by measuring PAR at 100-mm increments within the chamber beginning 20-mm from the side of the chamber opposite the IRGA. A narrow shadow, 5-mm wide, was measured at the 20-mm distance, which decreased overall transmittance (Figure 4). This narrow shadow resulted from the joining of the Plexiglas sides and propafilm top. Other than the 5-mm wide shadow, the chamber design allowed for approximately 90% PAR transmittance. The 10% attenuation by the chamber material was less than that reported by Pickering et al.

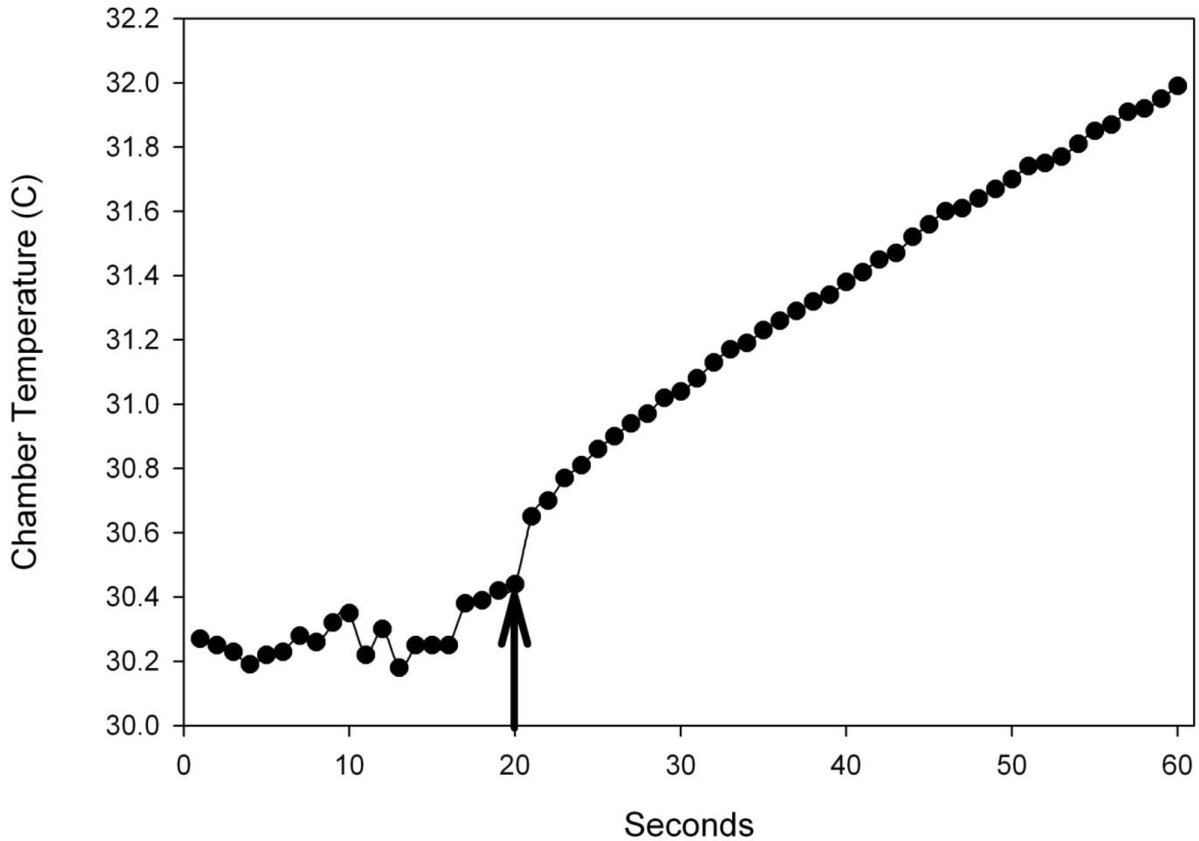

Figure 3. Typical air temperature inside the clear chamber observed during measurements in the field. The solid black arrow at 20 seconds indicates lowering of the chamber over the grass canopy.

(1993) and Steduto et al. (1992) of 20% and 12%, respectively. Overall, the chamber allowed for higher transmittance of PAR than that of previously reported chambers. The higher transmittance values probably derived from the use of propafilm as a top instead of Plexiglas. This result suggested that the current chamber construction minimized modifications to the PAR regime.

Examples of CO_2 and VPD rate changes

The chamber was tested over a wide range of canopy sizes and climatic conditions. Substantial rate changes of CO_2 and decreasing VPD's were associated with periods of high soil water content and large canopies (Figure 5 a,b). However, the rate change of CO_2 and VPD's remained mostly unchanged during measurements on plots following removal of leaf area by clipping (Figure. 6 a,b).

The basic formulas for computing CO_2 (J_c) and water vapor (J_w) fluxes are:

$$J_c = \rho_m \frac{V}{A} \frac{\delta CO_2}{\delta t}$$ [eq 1]

$$J_w = \rho_m \frac{V}{A} \frac{\delta w_c}{\delta t}$$ [eq 2]

Where J_c and J_w are flux density (mol m^{-2} s^{-1}), ρ_m is the molar density of air (mol$_{air}$ m$^{-3}{}_{air}$) calculated from the ideal gas law, V is the chamber volume (m$^{3}{}_{air}$), A is the chamber area (m$^{2}{}_{land}$) and $\delta CO_2/\delta t$ and $\delta w_c/\delta t$ are the rate changes over time of CO_2 and water vapor concentrations within the chamber (mol$_{CO_2}$ or $_{H_2O}$ mol$^{-1}{}_{air}$s^{-1}) respectively. The calculation of J_c is corrected for water vapor dilution as follows:

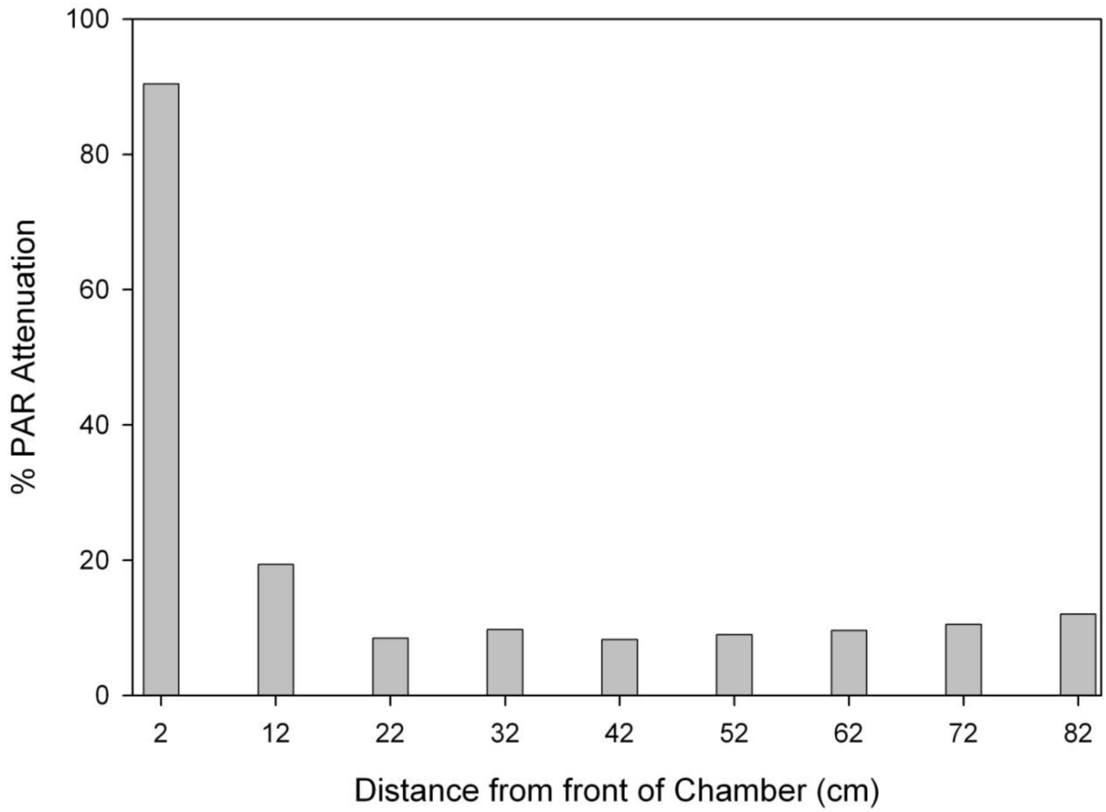

Figure 4. Percent of photosynthetically active radiation (PAR) attenuated by the clear chamber construction materials. Measurements of PAR attenuation began 20-mm from the side of the chamber opposite the IRGA with subsequent measurements taken every 100-mm.

$$J_c = \rho_m \frac{V}{A}\left(\frac{\delta CO_2}{\delta t} + \frac{CO_2}{(1-w_c)} \div \frac{\delta w_c}{\delta t}\right)$$

[eq 3]

where w_c is the mole fraction of water vapor within the headspace (mol mol^{-1}) and $\delta w_c / \delta t$ is the rate change of w_c (mol mol^{-1} s^{-1}). Corrections due to water vapor dilution were substantial during the two years of the study. The average difference between corrected and uncorrected NCE fluxes was 12.9% with a maximum correction difference of 29.2%. The results indicate the significant impact of correcting for water vapor dilution in closed chambers.

Determination of Flux

The preferred models for calculating $\delta CO_2/\delta t$ and $\delta w_c/\delta t$ have been discussed by Wagner and Reicosky(1992), Wagner et al. (1997), and Steduto et al. (2002). These studies mainly utilized either a linear or quadratic regression equation. The goal in selecting a model is to accurately predict flux when conditions within the closed chamber match ambient conditions, or before the chamber significantly influences the canopy environment.

A weakness of the linear model is that the rates of photosynthesis and transpiration are assumed to remain constant with decreasing concentration of CO_2 and increasing concentration of water vapor in the chamber. However, this is unlikely as shown by Fick's first law of diffusion

$$F_j = -D_j \frac{\delta c_j}{\delta x}$$

where F_j is the flux of j, D_j is the diffusion coefficient, and $\delta c_j / \delta x$ is the gradient of j between the chamber air and intercellular leaf air. This equation predicts the rate

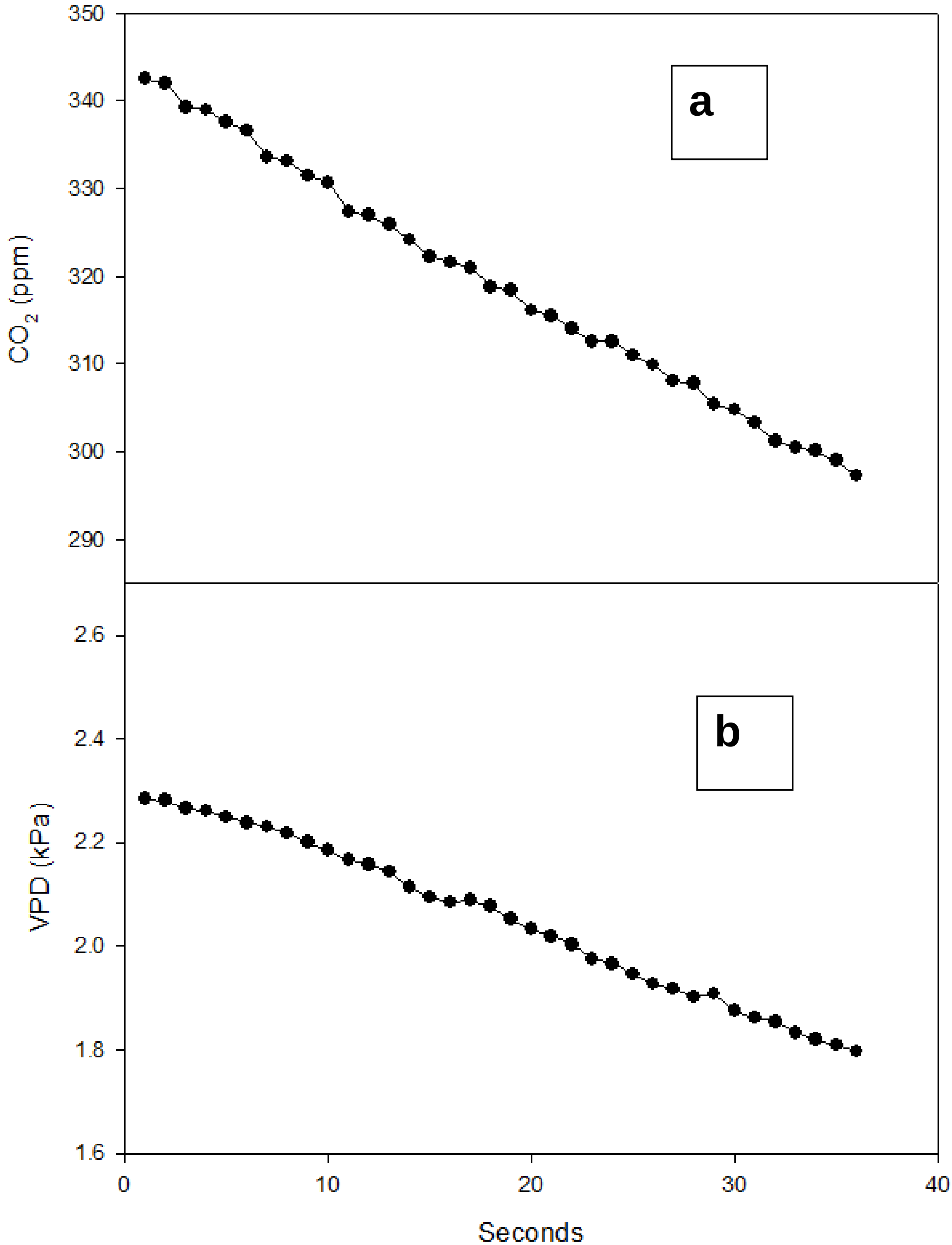

Figure 5. The rate change of CO_2 (a) and VPD rise (b) measured by a clear chamber over a well watered, full grass canopy.

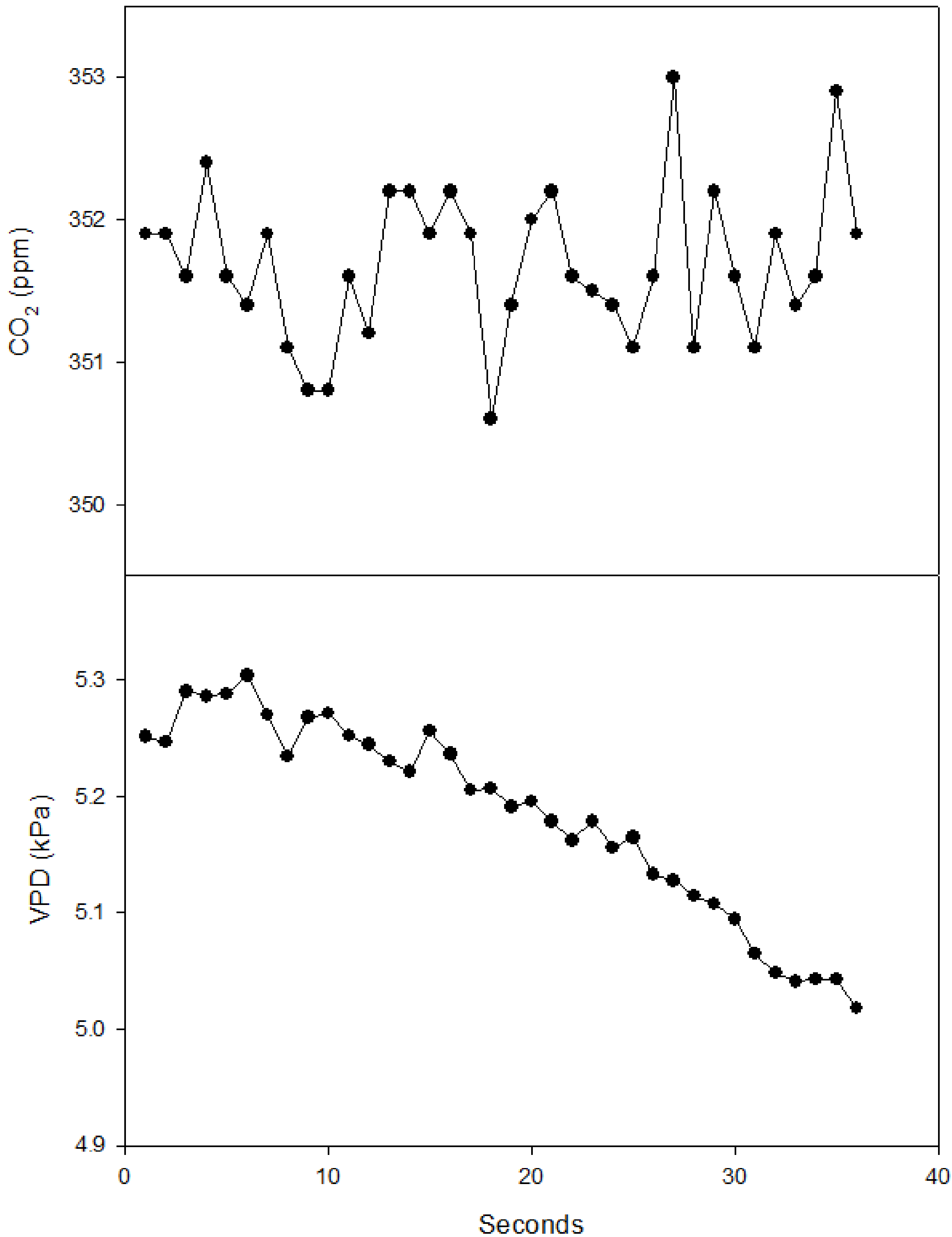

Figure 6. The rate change of CO_2 and VPD rise measured by a clear chamber on a plot with the leaf area recently removed by clipping all vegetation to a height of 5 cm.

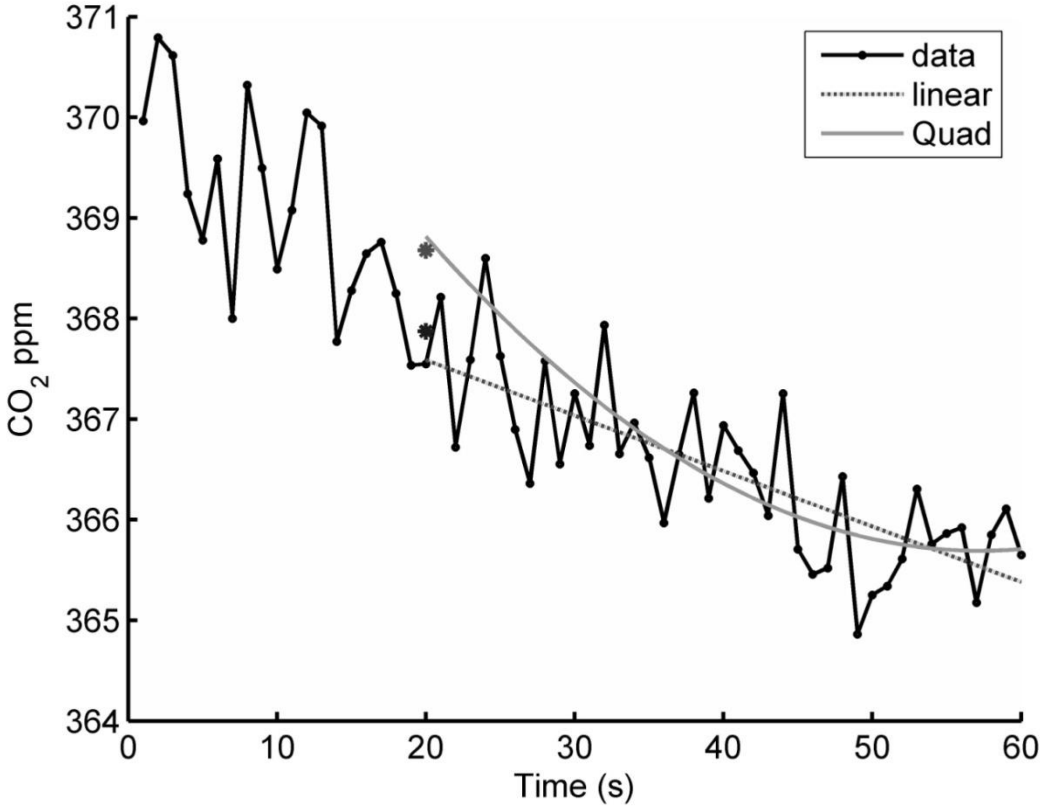

Figure 7. Actual CO_2 concentration recorded by the IRGA (data) and rate of change of CO_2 as calculated by the linear and quadratic (quad) models. The quadratic model predicted that the minimum value should occur at 50 s so a linear model was utilized in calculating the flux.

of diffusion of CO_2 into and water vapor out of the leaf will decrease as concentrations of CO_2 decrease and water vapor increase, respectively, within the chamber.

Wagner et al. (Wagner et al. 1997) and Studeto et al. (2002) stated that the quadratic model is preferred for closed systems. A quadratic model expressing CO_2 and H_2O concentrations as a function of time since closing the system can be written as:

$$CO_2 = a + bt + ct^2 \quad \text{[eq 5]}$$

$$H_2O = a + bt + ct^2 \quad \text{[eq 6]}$$

$\delta CO_2/\delta t$ and $\delta H_2O/\delta t$ are obtained by differentiating these equations with respect to time (t):

$$\frac{\partial CO_2}{\partial t} = b + 2ct_0 \quad \text{[eq 7]}$$

$$\frac{\partial H_2O}{\partial t} = b + 2ct_0 \quad \text{[eq 8]}$$

where t_0 is the time when ambient conditions were predicted to occur within the chamber. The slope is computed by solving the first derivatives at the time = t_0.

While a quadratic model is generally the best choice compared to a linear model for calculating $\delta CO_2/\delta t$ and $\delta H_2O/\delta t$, situations exist where the flux calculated using this model might be incorrect. For this study, a quantitative method for discriminating between regression models was developed. A program was written (MATLAB, The Mathworks Inc., Natick, MA) to quantitatively decide between a linear or quadratic model. Because the quadratic model is highly prevalent in the literature, the program initially accepted all quadratic models. The quadratic models were then tested using three criteria. First, the shape of the quadratic curve was tested. Specifically, the program determined if the quadratic model predicted a local minimum or maximum value within the 40-s sampling period (Figure. 7). Also, the derivatives at 30s and 60s were compared (Figure 8). If the minimum or maximum CO_2 or water vapor concentration; respectively, was observed within the 40-s time period, or if the derivative at 30-s was less than the

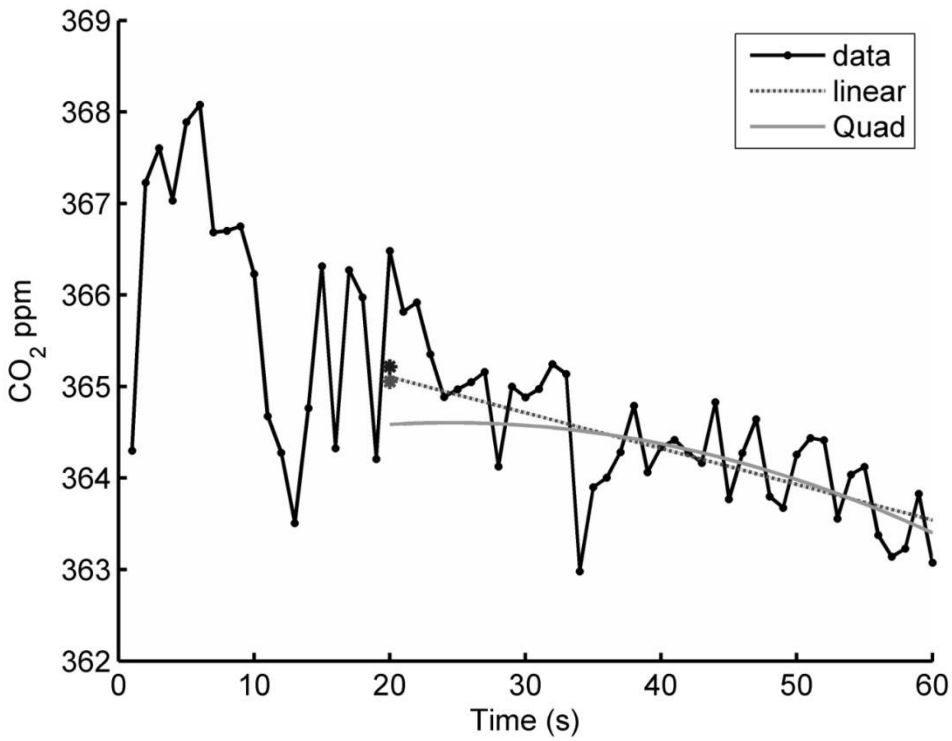

Figure 8. Actual CO_2 concentration recorded by IRGA (data) and rate of change of CO_2 as calculated by the linear and quadratic (quad) models. The quadratic model predicted that the slope of the quadratic model was less at 30 s than at 60 s therefore a linear model was used to calculate the flux.

derivative at 60-s, a linear model flux was utilized. Quadratic models that predicted a minimum or maximum value during sampling were discarded as the minimum and maximum values of CO_2 and water vapor, respectively, would be expected to occur at the end of the reading. The final check was testing if the quadratic model predicted that ambient conditions were achieved inside the chamber within the range of 15 s of placing the chamber on the soil frame. The time that conditions within the chamber matched ambient conditions was used for t_0 in predicting $\delta CO_2/\delta t$ and $\delta H_2O/\delta t$. If the predicted time that conditions within the chamber matched ambient conditions was outside this time frame, a linear model was utilized. Using these three criteria, the quadratic flux estimate was utilized for 80.1% of all NCE flux calculations, 14.2% of the ecosystem respiration flux calculations, and 99.5% of water vapor flux calculations (Table 1).

Comparison of Chamber and Eddy Correlation Fluxes

The definitive confirmation of the gas exchange chamber was the comparison of carbon fluxes measured with the clear chamber to other common methods of flux measurements specifically eddy correlation. The comparisons were conducted in an ungrazed, annually burned, native tallgrass prairie(Owensby et al. 1997). While soil types differed between the sites, similar vegetation dominated each site. The chamber readings were conducted over a 40 s sampling interval, while the eddy correlation values are 30 min averages. Overall, seasonal trends of NCE measured by both methods were similar (Figure 9 a,b). Differences between the two methods are probably due to soil types, size of sample area, and length of measurement.

CONCLUSIONS

The non-steady-state chamber described in this chapter couples a novel chamber design with precision instruments capable of fast sampling rates that in combination minimize microclimate disturbance. Throughout the two-year study, average chamber temperature increased 2.88° C, while chamber pressure

Table 1 The percent of net carbon exchange, ecosystem respiration, and water vapor fluxes calculated with quadratic models that were rejected for each year of the study (2005, 2006) due to violations of the criteria for shape of the predicted curve (Shape) or outside of the time frame for conditions within the chamber to match ambient conditions (Range).

	Net Carbon Exchange		Ecosystem Respiration		Water Vapor	
Criteria	**2005**	**2006**	**2005**	**2006**	**2005**	**2006**
Shape	20.7%	16.5%	89.0%	82.3%	0.24%	0.63%
Range	2.4%	0.2%	0.0%	0.3%	0.24%	0.0%

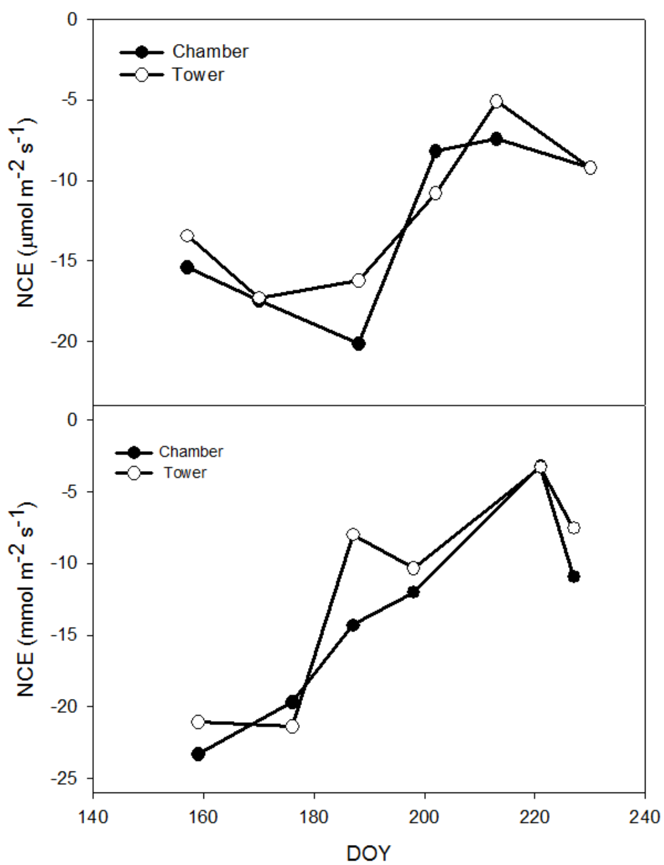

Figure 9. Net Carbon Exchange (NCE) of a tallgrass prairie measured by a clear chamber and eddy correlation on six dates during 2005 (a) and 2006 (b).

increased only 0.32-Pa during measurements, and PAR attenuation was approximately 10%. One key component of minimizing microclimate changes during a sampling period was to perform rapid measurements. The current configuration samples considerably faster than many other chambers, which remain on plots for as long as 3 minutes (Wagner and Reicosky 1992, Pickering et al. 1993, Wilsey et al. 2002). Also, a logical framework for determining the correct regression model also was developed. The method allowed for a non-biased decision regarding the appropriateness of the quadratic model based on a series of predefined criteria.

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