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TECHNICAL REPORT

EcoCELLs: tools for mesocosm scale measurements of gas exchange

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ABSTRACT

We describe the use of a unique plant growth facility, which has as its centerpiece four 'EcoCELLs', or 5×7 m mesocosms designed as open-flow, mass-balance systems for the measurement of carbon, water and trace gas fluxes. This system is unique in that it was conceived specifically to bridge the gap between measurement scales during long-term experiments examining the function and development of model ecosystems. There are several advantages to using EcoCELLs, including (i) the same theory of operation as leaf level gas exchange systems, but with continuous operation at a much larger scale; (ii) the ability to independently evaluate canopy-level and ecosystem models; (iii) simultaneous manipulation of environmental factors and measurement of system-level responses, and (iv) maximum access to, and manipulation of, a large rooting volume.

In addition to discussing the theory, construction and relative merits of EcoCELLs, we describe the calibration and use of the EcoCELLs during a 'proof of concept' experiment. This experiment involved growing soybeans under two ambient CO_2 concentrations (~360 and 710 μmol mol^{-1}). During this experiment, we asked 'How accurate is the simplest model that can be used to scale from leaf-level to canopy-level responses?' in order to illustrate the utility of the EcoCELLs in validating canopy-scale models.

Key-words: ecosystem carbon flux; elevated CO_2; gas exchange; global change; greenhouses; photosynthesis; soil respiration.

INTRODUCTION

Development of a predictive understanding of ecosystem responses to global change depends on identifying the key processes that control the exchange of material, energy, and information at a variety of spatial and temporal scales (molecules, cells, organs, individuals, communities, ecosystems and the biosphere). At each succeeding level of biological organization, diverse interactions between organisms and their environment exist. Related to each of these scales of biological organization is an inherent temporal scale and relaxation time (Jarvis 1995). Therefore, the impact of spatial and temporal characteristics of global change on ecosystems must also be considered (Ojima *et al.* 1991). In a review of critical research needs for understanding global climate change, Mooney (1991a) identified the lack of an integrated understanding of plant and ecosystem responses across spatial and temporal scales as limiting our ability to predict the response of ecosystems and the biosphere to global change.

To a large extent, our ability to understand system-level responses to environmental variables depends on available technologies. Just as technological advances such as leaf-level gas exchange systems have furthered our understanding of the biochemistry and physiology of photosynthesis (Mooney 1991b), an ecosystem-scale technology is needed to further our understanding of how systems respond to environmental variables and perturbations. For example, human-induced increases in atmospheric CO_2 are clearly documented in the direct measurements of CO_2 from historical records and ice cores (Barnola *et al.* 1987; Jouzel *et al.* 1993; Keeling *et al.* 1995). Scientific research concerning the effects of increasing CO_2 on plants has resulted in the development of technologies such as CO_2-controlled growth chambers and glass houses (Helmers & Giles 1979), open-top chambers (Rogers, Heck & Heagle 1983; Drake *et al.* 1989; Leadley & Drake 1993; Daudet & Claustres 1995), branch bags (Teskey, Dougherty & Wiselogel 1991; Barton, Lee & Jarvis 1993; Dufrene, Pontailler & Saugler 1993), controlled microcosms (Oechel *et al.* 1992; Körner & Arnone 1992) and FACE (Free Air Carbon Enrichment facilities; Allen 1975; McLeod & Fackrell 1983; McLeod, Alexander & Hatcher 1983; Hendrey, Lewin & Nagy 1993; Reece *et al.* 1995; technologies reviewed in Drake, Rogers & Allen 1986; Strain *et al.* 1991; Allen *et al.* 1992; Kimball 1992; Strain & Thomas 1992; Schulze & Mooney 1993). These technologies provide the capacity for long-term (weeks to years) CO_2 exposure of plants at a variety of scales and under a variety of conditions. However, these technologies often still have limited capabilities to study the mechanisms of ecosystem-level responses in manipulated environments.

Despite the global nature of current environmental change and the development of larger scale CO_2 dosing technologies, the majority of our mechanistic knowledge is limited to the whole plant down to the biochemical, and often molecular, scales. At these scales knowledge is

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obtained primarily from pot studies on individual plants, and is increasingly falling short of predicting system responses in modelling and scaling experiments (Körner, Arnone & Hilti 1993; Körner 1995a). Controlled environment facilities that can provide the capacity for larger spatial scale response measurements may be the only means of studying the effects of single factors or a small suite of factors on model systems in an attempt to bridge the gap between growth chamber/pot studies and the inherently more complex natural system-level experiments such as FACE (Schulze & Mooney 1993).

To date, only a few studies of system-level responses of ecosystems to CO_2 enrichment have been carried out (reviewed in Strain & Thomas 1992). These include microcosm studies of the Alaskan tundra (Billings *et al.* 1982, 1983, 1984; Tissue & Oechel 1987; Grulke *et al.* 1990; Oechel *et al.* 1993; 1994), pasture systems (Overdieck & Reining 1986), shortgrass prairie (Hunt *et al.* 1990), tallgrass prairie (Owensby *et al.* 1993), Texas rangeland (Johnson, cited in Strain & Thomas 1992), serpentine and greenstone grasslands (Field *et al.* 1995), alpine grassland (Schäppi & Körner 1996), a Mediterranean grassland (Navas *et al.* 1995) and a wetland (Drake 1992). Field studies have provided, and will continue to provide, valuable information and insight into the carbon dynamics of natural systems. One of the most significant contributions to date from elevated CO_2 research in semi-natural systems has been the quantification of biomass accumulation of several tree species during long-term CO_2 enrichment and the attempt to obtain a carbon mass balance, accounting for the inputs, outputs and storage. Mass balancing, or carbon budgeting, is particularly difficult in field situations where natural variability and environmental complexity are often extreme. Use of the mass balance approach in field situations often requires a search for very small signals against very large background fluctuations. Furthermore, the complexity of the system often makes the identification of key mechanisms difficult. Field studies have thus been unable to account entirely for the fate of fixed carbon. Mass balance techniques, similar to those used in leaf-level gas exchange systems but scaled to the system level, are required for measurements of whole-ecosystem carbon balance and flux.

We have developed a system capable of quantifying carbon, water and trace gas fluxes from large mesocosms. This system is unique in that it was conceived specifically to bridge the gap between measurement scales during long-term experiments examining the function and development of model ecosystems. Ecological research on global change uses a spectrum of technologies and covers a continuum of spatial and temporal scales, and it is important to assess the relative merits of each technology. EcoCELLs have several unique advantages.

(1) The EcoCELLs have the same theory of operation as leaf-level gas exchange systems but work at a much larger scale, and measure fluxes continuously.

(2) By providing ecosystem-level measurements under controlled environmental conditions, the EcoCELLs can provide data to evaluate independently canopy-level and ecosystem models simultaneously under ambient and non-ambient conditions, something that to our knowledge has never been done before.

(3) Relative to other technologies such as open-top chambers and FACE, the EcoCELLs can both manipulate environmental factors and measure system-level responses with a scale of resolution similar to that of well-designed leaf-level gas exchange systems.

(4) The EcoCELLs maximize access to, and manipulation of, the rooting volume which is much larger than the rooting volume in most experimental systems.

Furthermore, the resolution of the system and the ability to manipulate the environmental variables allow repeatable single-factor manipulations to study system components and mechanisms. General disadvantages of controlled-environment facilities like the EcoCELLs include limited size (although the EcoCELLs are among the largest controlled environments), the structural simplicity of the model systems contained within the facility, and the need for extrapolation of the data to natural environments (Lawton 1995).

The purpose of this paper is 3-fold: first, to introduce the scientific community to this new, controlled-environment plant growth facility (Ecologically Controlled Enclosed Lysimeter Laboratory, or EcoCELL); secondly, to present the data from a 'proof of concept' experiment utilizing this new facility, and thirdly, to suggest how the EcoCELLs can be used to further our understanding of plant response to global change across a wide range of spatial and temporal scales. The 'proof of concept' experiment involved growing soybean (*Glycine max*) in the EcoCELLs under two ambient CO_2 concentrations (≈360 and 710 μmol mol^{-1}). During this experiment we asked the question, 'How accurate is the simplest model that can be used to scale from leaf-level to canopy-level responses?'

EcoCELLs DESCRIPTION AND TEST

An EcoCELL is a large environmentally controlled and naturally lit plant growth chamber capable of making continuous mesocosom-scale measurements of mass and energy fluxes during long-term experiments (months to years). This mesocosm-scale facility is part of the Great Basin Environmental Research Laboratory (GBERL) at the Desert Research Institute (DRI) in Reno, Nevada. The mesocosms form the centrepiece of a coordinated and cooperative research effort designed to study plant processes in an integrated, cross-scale fashion in order to bridge the gap between measurement scales. This facility contains four EcoCELLs and is complemented by a set of smaller chambers (1m × 1m), that can be used as microcosms. The dimensions and volumes of the EcoCELLs are listed in Table 1. The EcoCELLs are comprised of four main systems: (1) rhizotron/weighing lysimeters (soil

Table 1. EcoCELL specifications. Each of the four EcoCELLs is a separate mesocosm designed as an open-flow, mass-balance system for the measurement of trace gas exchange. Air temperature and humidity, trace gas concentrations, and incoming air flow rate are strictly controlled as well as being accurately and precisely measured

Physical specifications	
EcoCELL	Dimensions, 7·3 × 5·5 × 4·5 m (L × W × D) Total volume, 183·5 m^3 (6313 mol) Circulating volume, 162·5 m^3 (5590 mol)
Soil containers	3 per EcoCELL Dimensions, 2·85 × 1·3 × 1·8 m (L × W × D) Total volume, 6·7 m^3 per pot or 20·1 m^3 per EcoCELL Surface area, 11·2 m^2 per EcoCELL Approximately 11 000 kg soil per pot (sandy loam top soil)
Operational specifications	
Achievable temperature setpoints	Hottest day: 18–50 °C ± 1 °C at plant surface Hottest night: 5–30 °C ± 1 °C at plant surface Coldest day: 10–35 °C ± 1 °C at plant surface Coldest night: 5–30 °C ± 1 °C at plant surface
Achievable humidity setpoint	10–80% RH at all temperatures (± 5% at steady state) Humidity controlled by varying the dew point and flow rate of incoming outside air
Air Circulation	Incoming air flow: 500 to 5000 mol min^{-1} Within chamber circulation rate: 25 000 mol min^{-1} Minimum residence time: 70·7 s (5 passes over canopy) Maximum residence time: 700 s Wind speed over plant canopy: variable from < 1 to 2 m s^{-1}
Trace gas concentrations	CO_2 setpoints from ambient to 1000 μmol mol^{-1} Measurement of most trace gases possible Air filtration with charcoal and/or 99·5% HEPA filters

containers), (2) enclosures, (3) environmental control systems, and (4) instrumentation and data acquisition systems. All materials used in the construction of the EcoCELLs were carefully selected to minimize the gas sorption properties for CO_2 and other trace gases in order to maximize the signal-to-noise ratio of the system and to reduce the system hysterisis. For example, because concrete surfaces can exchange CO_2, all concrete surfaces within the EcoCELLs were sealed with an epoxy paint to prevent CO_2 exchange (0·25-mm-thick application of TNEMEC series 104, TNEMEC Company, Inc., Kansas City, MO).

Rhizotron/weighing lysimeters

Within each EcoCELL there are three soil containers designed to function as weighing lysimeters. The dimensions, total volume, surface area and approximate mass of these soil containers during the preliminary experiments are also given in Table 1. Each lysimeter (soil container) consists of a steel tube frame coated with epoxy paint and lined with 1·9-cm-thick acrylic plastic sheets. Suction candles are installed at the bottom of each container to provide control over soil water potential. Chilled water is pumped through a manifold installed near the bottom of the soil column to provide control of soil temperature (measured via an array of copper-constantan thermocouples). Other devices or instrumentation (e.g. time domain reflectometry probes, gas wells and mini-rhizotron tubes) may be installed as appropriate to the question posed.

Enclosures

Each EcoCELL is constructed using a 5·5 × 7·3 × 2·4 m steel channel frame (see Table 2 for equipment/materials and manufacturers) assembled over a 5·5 × 7·3 × 2·4 m concrete pit. The steel channel frame is glazed with 6·3-mm-thick tempered glass. The roof of the structure is a low-density polyethylene membrane supported by the internal air pressure of the EcoCELL. Other materials may be easily substituted for this low density polyethylene if required for a particular experimental protocol (i.e. enhanced transmissivity). The entire EcoCELL is fully enclosed within a large outer glass house glazed with a two-layer acrylic. The EcoCELLs are housed in this larger outer glass house to improve environmental control and to prevent the formation of condensate on the EcoCELL glazing. The design of the outer greenhouse and EcoCELL enclosure minimizes structural shading of the plant canopy. Outer greenhouse transmission is 85% and EcoCELL skin transmission is 92%. Uniformity of the light environment is quite high (≈20%).

Environmental control

A flow diagram of the system is presented in Fig. 1. The system relies on outside air to supply the relatively high flow rates into each EcoCELL (500 to 5000 mol min^{-1}). The climate of Reno, NV is such that outside air is very dry and can be used to balance the increasing water vapour

Table 2. Suppliers of materials and equipment

Equipment/Materials	Make/model	Supplier (US)
Greenhouse skin	Exolite	Cyro Industries, Orange, CT
Steel frame	Unistrut	Unistrut, Wyane, MI
Air handlers and delivery system		McQuay – Snyder General, Minneapolis, MN
Circulating fans		Barry Blower, Minneapolis, MN
Variable speed drives	Lancer GP 502	Magnetck Drives & Systems, Neuberlin, WI
Direct evaporative cooler	Norpack	Norsaire Systems, Denver, CO
Valves and fittings	various	Clippard Inst., Cincinnati, OH
Mass flow controllers	Side Track – 2, 5, 15 & 100 dm^3 min^{-1}	Sierra Instruments Inc., Monterey, CA
Multipoint hot-wire anemometer	Steel Track	Sierra Instruments Inc., Monterey, CA
Temperature control – hardware	various	Steafa Control System Inc., San Diego, CA
Temperature control – software	Stefa Access	Steafa Control System Inc., San Diego, CA
Data loggers and electronics	CR10T, MD9, SDM-CD16, SDM-A04, AM-416	Campbell Scientific Inc., Logan, UT
System software	RTMS	Campbell Scientific Inc., Logan, UT
IRGA	Li-6262	Li-Cor Inc., Lincoln NE
Quantum sensor	9901-013	Li-Cor Inc., Lincoln NE

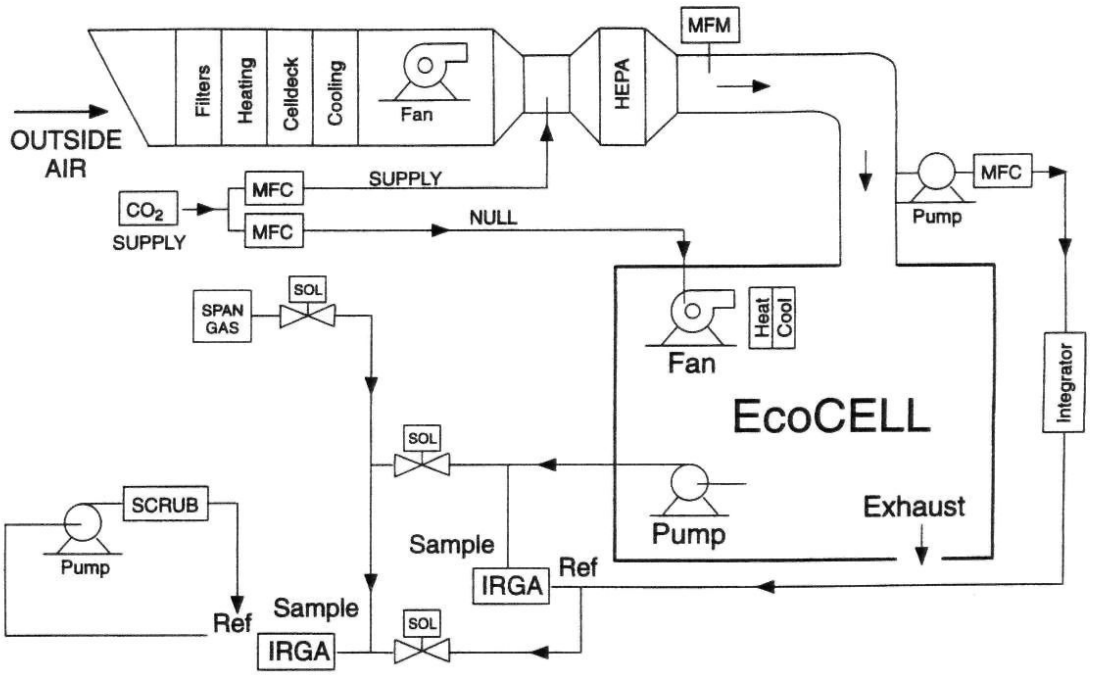

Figure 1. Diagram of EcoCELL flow, control and monitoring systems. All components other than the absolute IRGA are replicated four times, once for each EcoCELL. The absolute IRGA samples gases from all four EcoCELLs and a span gas. Celldeck = humidification, HEPA = high efficiency particulate air filter, MFM = mass flow meter, MFC = mass flow controller, SOL = solenoid valve, Supply MFC = three staged system for controlling the ambient CO_2 concentration of the air entering the EcoCELL, Null MFC = a single mass flow controller functioning to compensate for CO_2 uptake by the plant canopy, and IRGA = infrared gas analyser.

content of the air resulting from transpirational water loss. Total air flow through the system is regulated to maintain a desired water vapour pressure within the cell. Although it would be desirable to mix the incoming air stream from stocks of N_2, O_2 and the desired trace gases, it is not technically or economically feasible to supply these constituents at the required rates. Ambient air introduced into the system is first filtered [High Efficiency Particulate Air (HEPA) and/or charcoal filters] to remove particulates and some trace gases. The air can be heated by moving it across heating coils (hot water), humidified by passing it over a direct evapourative cooler, and cooled/dehumidified by passing it over a chilled water coil.

Once the dew-point of the incoming air is set by conditioning the air for temperature and saturation vapour pressure, the CO_2 concentration is set. As stated previously, the system is dependent on outside air and thus the lower-end set-point is constrained by the global average CO_2 concentration and local anthropogenic source emissions (typically 36 to 38 Pa, but can be as high as 45 Pa on very still winter

days). Elevated carbon dioxide concentrations are set by a three-stage system. First a needle valve is used to inject a constant amount of CO_2, approximating 80 to 90% of the required addition. Secondly, a 100 dm^3 min^{-1} mass flow controller is used in a series of 15 dm^3 steps for coarse control, and finally a 15 dm^3 min^{-1} mass flow controller is used for fine control. Using this three-stage approach we are able to obtain CO_2 concentrations well within 2% of the desired set-point. The system may be configured to maintain either a constant set-point or a constant differential above ambient. The mass flow rate of air entering the EcoCELL is measured directly with a multipoint hot-wire anemometer. The accurate and precise measurement of the air flow entering the EcoCELL is crucial to the system flux measurements and may be the most difficult variable to quantify. Air flow is not laminar and thus each mass flow meter was individually calibrated *in situ* using a trace gas addition technique (Field, Ball & Berry 1991).

The conditioned gas enters the EcoCELLs below the soil containers and is circulated within the chamber at a rate of 25 000 mol min^{-1}. As the air circulates in the EcoCELL it is again passed over heating and cooling coils to control the final ambient air temperature to a set-point of 5 to 50 °C ± 1 °C at the plant surface. The system was designed to ensure that cooling is achieved without the formation of condensate on the cooling coils. Detailed achievable set-points and flow rates are catalogued in Table 2. As the air is circulated within the EcoCELL it can be vectored over the canopy by a series of nozzles, influencing the wind speed over the canopy and thus the physical characteristics of the boundary layer. Finally, air is exhausted from the EcoCELL via a series of weighted dampers installed in an air lock, ensuring that the entire EcoCELL is maintained under a constant positive pressure.

Instrumentation and data acquisition

The entire EcoCELL mass balance system control is obtained by two separate but coordinated computer systems. Temperature set-points, vapour pressure control, air speed and circulation are accomplished with Steafa Control System hardware and Steafa Access software. Temperature control is achieved by a combination of straight proportional and PID (Proportional + Integral + Derivative) algorithms. Humidity is controlled by adding back to the EcoCELL enough dry air to balance the addition of water by the plant canopy, tracking the desired set point. This control strategy works very well in Reno where the average ambient vapour pressure of the air is low. Variable frequency drivers control the fans used to introduce air into the EcoCELLs and can vary the incoming mass flow rate from 500 to 5000 mol min^{-1}, maintaining the EcoCELL to within ±5% of the desired humidity set-point. Control of trace gas addition and sampling is accomplished by a separate computer network consisting of micro data loggers and control software. The amount of CO_2 added to the incoming air stream is calculated from the measurement of the ambient air CO_2 partial pressure and the incoming flow rate, and is set by the three-step valve and mass flow meter system described above. A separate 2 dm^3 min^{-1} mass flow is used to compensate plant canopy CO_2 uptake by adding back an equivalent molar flow. This compensating technique is known as null balancing.

Since the measurement of CO_2 flux is accomplished as a differential measurement of CO_2 across the EcoCELL (see below), the control system must also ensure that the timing of incoming and outgoing gases is matched at the infrared gas analyser. This is accomplished by using an integrating volume and varying the volume-to-flow ratio into this integrating volume in step with changes in the volume-to-flow ratio into the EcoCELL. A 5 dm^3 min^{-1} mass flow is used to accomplish this integration, ensuring that air samples reach the sample and reference sides of the IRGA simultaneously, minimizing the effects of any short-term variations or fluctuations in the ambient CO_2 concentrations.

Whenever possible, the measurement and control systems have been kept completely separate. For example, the relative humidity of the EcoCELL is controlled by STEAFA while the measurement of H_2O vapour flux is accomplished with an IRGA monitored by RTMS. Five IRGAs are dedicated to the monitoring system. Four IRGAs are continuously run in differential mode to record the flux of CO_2 and H_2O across each EcoCELL. The fifth IRGA is run in absolute mode, and sequentially samples a standard gas, and the gas entering and exiting each EcoCELL. The reference gas for this IRGA is a closed loop continuously scrubbed of CO_2 and H_2O by soda-lime and magnesium perchlorate. All five IRGAs average over a 30 s interval are sampled at 5 s intervals, and are recorded as 60 s averages. Light levels in each of the EcoCELLs are monitored with a quantum sensor mounted parallel to the surface of the pots, well above the plant canopy. Ambient air temperature, incoming air temperature and soil temperature at three depths are measured with copper-constantan thermocouples. All aforementioned devices are hardwired to Campbell data loggers and data collection is handled via RTMS.

Flux measurements

The flux of carbon and water through the mesocosm is continuously monitored in real time from the start of an experiment. Carbon and water flux calculations are made either as open system differential measurements or as null balance compensating system measurements as described by Field *et al.* (1991) and expressed on a unit surface area basis. All calculations are suspended during periods of time when personnel are required to enter the EcoCELLs. Thus, these times are coordinated and kept to a minimum.

'Proof of concept' experiment

After thorough calibration and empty chamber testing an initial 'proof of concept' experiment was conducted. The goals of this experiment were 2-fold; first, to test the

functioning and sensitivity of the system, and secondly, to ask a simple scaling question. The primary requirement of this experiment was a fast-growing, highly photosynthetically active plant canopy. For this reason soybean was chosen as a model system. Similarly, in order to test the ability of the system to control the trace gas environment, the experiment was conducted under two CO_2 partial pressures, ≈36 and 71 Pa.

In early October 1994 all three soil containers within EcoCELLs 1 and 2 were filled with a sandy loam top-soil containing 30% organic matter. In order to facilitate settling, the soil was flooded and then the pot was drained and topped off with soil. The highly disturbed nature of the soil, the flooding treatment and the high percentage of organic matter contributed to large CO_2 efflux from the soil surface.

Seeds of *Glycine max* were planted on 5 cm centres and then thinned to 10 cm centres in EcoCELL 1 and 2 on 5 February 1995. EcoCELL 1 was maintained at an ambient CO_2 concentration ~360 μmol mol^{-1}, and EcoCELL 2 was maintained at 710 μmol mol^{-1} throughout the duration of the experiment (5 February to 15 June 1995, 131 d). Both EcoCELLs had a day/night temperature regime of 28/20 °C. The day/night temperature regime followed the photoperiod and therefore was adjusted to day length. Initially the relative humidity of the EcoCELLs was set quite low, at approximately 15%. However, this proved to be limiting to plant growth and after 6 weeks the relative humidity was increased to 45%. Plants were watered with a drip irrigation system as needed.

On 12 and 13 January 1995, prior to planting, point measurements of soil CO_2 efflux were made with a Li-Cor 6000-09 soil respiration chamber and infrared gas analysis system (Li-6200, Li-Cor Inc., Lincoln, NE). Each half pot was divided into 100 10×10 cm plots and a random number generator was used to pick 25 plots in one half pot, and 10 plots in each of the other five half pots. In total, 150 separate measurements of CO_2 efflux in EcoCELL 1 were made over the 2 d period.

During the 3 d period of 1 to 3 June, leaf-level light response curves were recorded in EcoCELL 1 (38 Pa ambient CO_2 partial pressure) with an open flow infrared gas analysis system (Li-6400, Li-Cor Inc., Lincoln, NE). All measurements were made at a constant air temperature of 28 °C, the ambient CO_2 concentration, and a relative humidity of 45%. The photosynthetic response to incident light intensity was measured by varying the light intensity of a LED light source (Li-6400-02, Li-Cor Inc., Lincoln, NE) and recording the steady-state photosynthesis rates at eight light intensities: 10, 50, 100, 200, 500, 1000, 1500 and 2000 μmol quanta m^{-2} s^{-1}. Photosynthesis was considered to have reached steady state when the total coefficient of variation for both the net photosynthetic rate and stomatal conductance was less than 1% over a 45 s time period. In total, 25 light response curves were obtained in the 3 d period. Measurements spanned the photosynthetically active hours of the day, from approximately 0930 to 1600 h. Leaves were randomly selected and represented leaves of different ages, locations within the EcoCELL and within the canopy, and were measured at different times of day. In general all leaves were close to the edge of the canopy.

Leaf area index was estimated with a Li-2000 plant canopy analyser (Li-Cor Inc., Lincoln, NE). Estimates were made on 2 June, a cloudy afternoon. For the purpose of these measurements each pot was divided into eastern and western halves, giving six canopy sections. Two measurements of LAI each consisting of the mean of four randomly chosen points below the canopy were made in each of these sections.

RESULTS

The fundamental calibration challenge to any gas exchange system is to add a known amount of CO_2 directly to the cuvette and compare this to the observed increase in the calculated flux. We performed this experiment, adding the equivalent of 38·8 μmol s^{-1} of CO_2 directly to EcoCELL 1 via a flow controller, and found the corresponding increase in the calculated flux to match to within 1·1 μmol s^{-1} (Fig. 2).

The 150 point measurements of soil CO_2 efflux were variable (from approximately 4 to 25 μmol m^{-2} s^{-1}; Fig 3). This variation was not correlated to soil surface temperature measured at a depth of 10 cm (data not shown, $R^2 = 0{\cdot}05$). The mean of these 150 measurements was quite high, averaging 17·1 ± 0·67 μmol CO_2 m^{-2} soil surface area s^{-1}. EcoCELL flux measurements made just prior to or immediately following the point measurements ranged from 16·3 to 17·2 μmol m^{-2} s^{-1} averaging to within 0·25 μmol m^{-2} s^{-1} of the point measurements. The extremely high rate of respiration from this soil was probably a result of the high percentage of organic mater, the high degree of disturbance and the flooding regime used to settle the soil.

Instantaneous ambient air temperature measurements fluctuated over a 1·5 °C range during the daylight hours and by less than 0·1 °C during the night-time hours (Fig. 4).

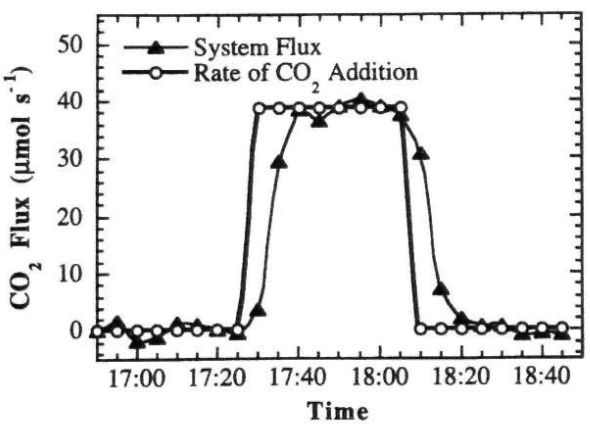

Figure 2. CO_2 calibration of EcoCELL precision. A flow of 38·8 μmol s^{-1} of CO_2 was added directly to an empty EcoCELL. The calculated flux from the EcoCELL exchange increased by a similar amount. The CO_2 addition and calculated flux lines are slightly offset for illustration; the resulting time constant is <15 min.

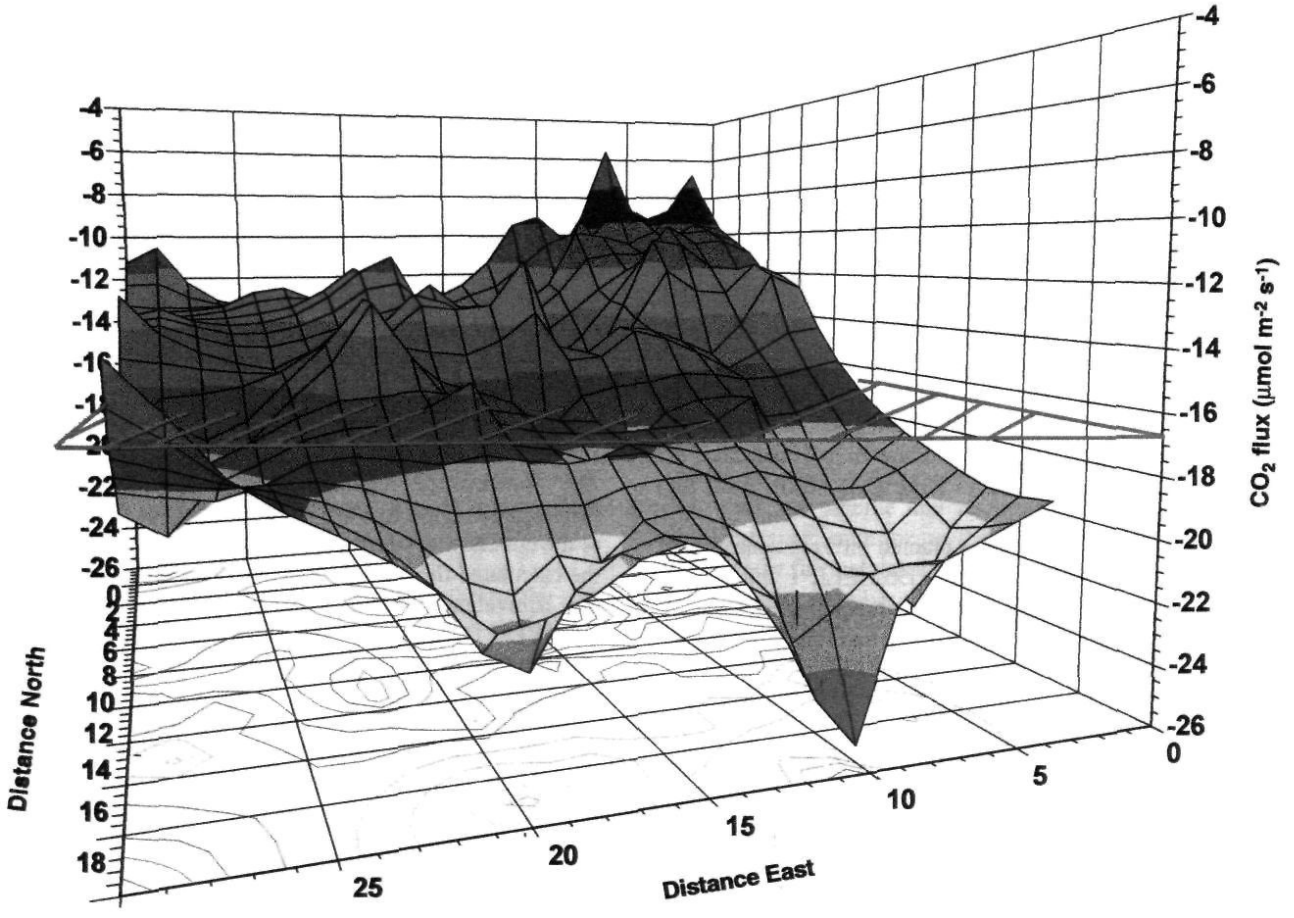

Figure 3. Surface plot of soil CO_2 efflux compiled from 150 point measurements. The red plane represents both the average flux rate from these 150 point measurements and the EcoCELL measurement of whole system flux made immediately prior to and immediately following the point measurements.

During the same 24 h period ambient outside air temperatures fluctuated by more than 20 °C and the mean radiative flux exceeded 900 W m^{-2} for extended periods of time. Manual adjustments in the temperature set-point made the difference between the cells <0·3 °C over any 24 h period. Similarly, we were able to control RH to ±5% and CO_2 to ± 2 μmol mol^{-1}.

Following planting, whole-system flux was initially nearly constant over the course of the day and was dominated by high CO_2 efflux rates from the soil. As the plant canopy matured, there developed a diurnal trend of high night-time CO_2 efflux followed by diminishing whole system efflux as canopy uptake balanced the respired CO_2 from the soil. Fifty-two days after planting, the CO_2 uptake by the canopy at midday closely matched the CO_2 efflux from the soil and the net differential across the EcoCELL was zero (Fig. 5, upper panel). Another common trend displayed in this data set is the increase in CO_2 efflux in the dark period following a sunny day of carbon gain with declining respiration rates over the duration of the dark period. Interestingly, there were no significant differences between the two EcoCELLs on this day. The initial portion of the light response can be seen in the expanded portion of these data from 0600 to 0900 h (Fig. 5, lower panel).

Light-saturated photosynthetic rates from randomly selected leaves ranged from 2·0 to 14·0 μmol m^{-2} s^{-1} (Fig. 6, upper panel). The mean response of all 25 curves had a light-saturated rate of 6·35 ± 0·86 μmol m^{-2} s^{-1}, a dark respiration rate of 0·85 ± 0·1 μmol m^{-2} s^{-1} and a light compensation point of 22 μmol of quanta (Fig. 7, lower panel). For comparison with the whole-system-level flux, we scaled

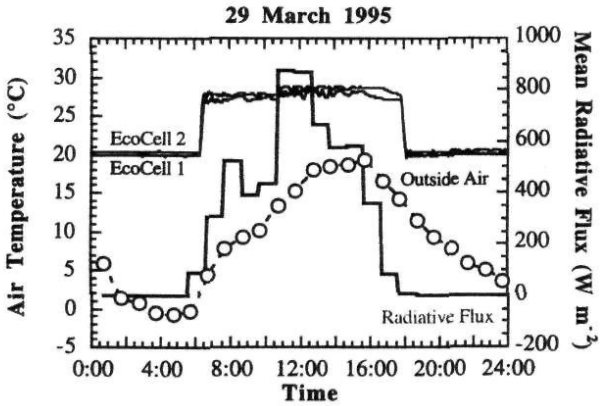

Figure 4. EcoCELL temperature (°C), outside air temperature (°C) and radiative flux (W m^{-1}) on 29 March 1995.

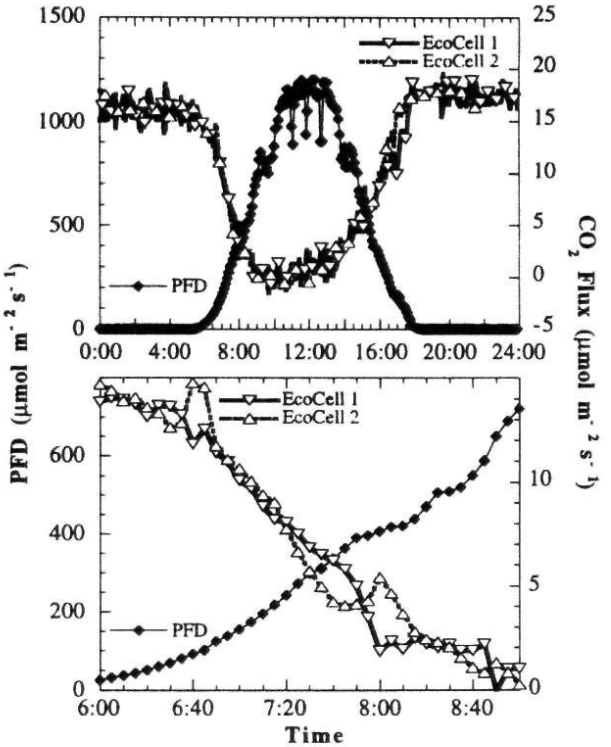

Figure 5. Whole-system flux and photon flux density (PFD) measured inside EcoCELL 1 and 2 on 29 March 1995 (upper panel). Expanded whole-system flux and PFD during sunrise (0600 to 0900, lower panel).

this average response by the average leaf area index of the canopy (2·595 m^2 m^{-2}). This scaled response had a light saturation rate of 16·5 μmol m^{-2} s^{-1} and a dark respiration rate of –2·2 μmol m^{-2} s^{-1} (Fig. 6, lower panel).

On 4 June (the day following the completion of the leaf-level light response curves), whole-system-level fluxes were measured, uninterrupted for the entire day (Fig. 7). During this 24 h period, night-time CO_2 flux was higher than on 29 March. This is at least in part due to increased respiration from the rapidly senescing vegetation of the fully developed canopy on 4 June as compared to the expanding, lower leaf area canopy on 29 March. For comparison with the average leaf-level light response, the light response of the entire system was calculated (Fig. 8). This calculation required three steps. First, the diurnal course of light intensity was smoothed to account for shading of the single quantum sensor by structural members of the outer greenhouse. Secondly, the average CO_2 flux rate from midnight to sunrise was calculated, and thirdly, the average rate of 'system respiration' was assumed to continue throughout the daylight hours. The net uptake of carbon was then calculated as the absolute value of the difference between the system respiration and the carbon flux in the light.

The EcoCELL light response of CO_2 fixation closely matched the scaled leaf-level response for the first 3 h of the day, particularly in the quantum yield portion of the curve. The maximum light-saturated rate of 'system photosynthesis' was 7·8% higher than the leaf-level maximum (17·8 versus 16·5 μmol m^{-2} s^{-1}). As incident radiation approached 1000 μmol m^{-2} s^{-1} the system-level response started to decline, averaging 15·0 μmol m^{-2} s^{-1} for much of the afternoon. The reasons for this decline are unclear but may include a midday depression of photosynthesis due to stomatal closure, carbohydrate accumulation or mild photoinhibition. As the sun began to set, late afternoon system photosynthetic rates showed a large, nearly 30% historesis in the light response. Integration of the measured carbon flux over the entire 24 h period of 4 June results in a total carbon flux of 17·45 mol. If the average scaled leaf-level response is used to estimate the carbon flux on this day an integrated daily flux of 19·4 mol, or an 11% overestimate, is derived.

DISCUSSION

The design of the plant growth facility described here allows a larger scale approach to understanding the response of terrestrial systems to global change. Although the system is complex and technically demanding, the

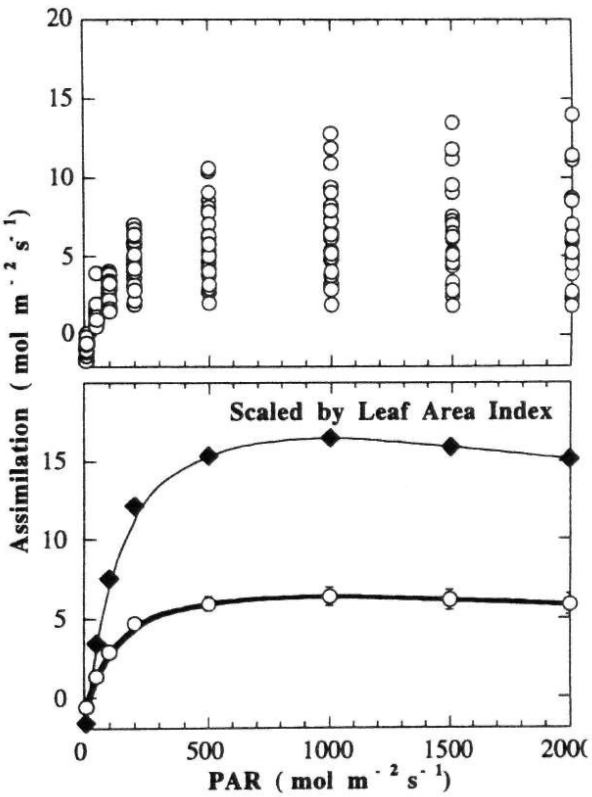

Figure 6. Photosynthetic light response of 25 randomly selected soybean leaves grown for 118 d in EcoCELL 1, with an ambient CO_2 concentration of ≈ 360 μmol mol^{-1} (outside air, upper panel). The average response from these 25 randomly selected leaves is shown (plotted as the mean value ± 1 SEM, open symbols and thick line in lower panel).For comparison with the whole-system flux, this average value was scaled by multiplying each point by the average leaf area index (2·6 m^2 m^{-2}, solid symbols and thin line in lower panel).

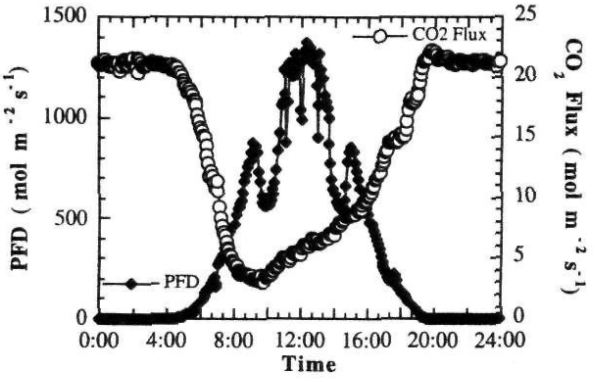

Figure 7. Whole system flux and photon flux density (PFD) measured inside EcoCELL one on June 4, 1995.

theory of operation is simple. Mass balance flux measurements have been the staple of physiological measurements of plant response for several decades. Here we apply this theory to a much larger spatial scale and introduce the ability to make these measurements continuously over the lifetime of the plants or the experiment. Both the CO_2 addition experiment and the soil respiration measurements suggest that whole-system-level flux measurements can be made with a high degree of accuracy even at this large scale. Similarly, the responsiveness of the whole-system flux to light demonstrates the ability of the system to measure meaningful real time carbon fluxes.

Ultimately, one of the many uses for this facility should be to ask complex scaling questions in order to assess how the vast amount of process-level data available at the leaf and lower levels can be applied to higher levels of organization. We took a very simplistic approach to this issue in the 'proof of concept' experiment, by asking the question 'What is the simplest possible model for scaling leaf-level response and how close does it come?' The simplest possible model was to multiply an average leaf response by the leaf area index. Surprisingly this model did quite well. The agreement between the scaled light response and system measurements was within 10%. Perhaps this is most surprising since we did not expect this simplistic model to work. The simplest intelligent model would include at a minimum light extinction through the canopy, and perhaps leaf area display and canopy architecture (Baldocchi 1993). Thus, the model used should be a significant overestimate, particularly in the quantum yield portion of the curve. However, Norman (1993) suggested that models that consider a plant canopy as one large horizontal leaf work reasonably well for canopies with a LAI near 2, and Terashima & Hikosaka (1995) report several canopy-level adjustments that optimize canopy light response.

Interpretation of this finding is challenging. It may suggest that we have all the information we need from the scores of leaf-level data reported in the literature, but given the complexities and non-linearities inherent to natural systems (Jarvis 1995) we find this unlikely. Alternatively, the correlation between the leaf-level and system-level measurements could be coincidental. The scaling process is complex (Field & Ehleringer 1993; Jarvis 1995) and we do not suggest that our simple model is anything more than a limited observation. We present this data as a means of illustrating the capabilities and potential of this measurement system. EcoCELLs allow direct comparison between integrative upscaling and direct measurements at the scale of interest. The correct interpretation of these results demands a carefully defined question. For example, if the ultimate question is related to carbon sequestration, an 11% overestimation on a daily basis, when integrated over several growing seasons or the life-time of the species of interest, is completely unacceptable. Obviously there is a large amount of work yet to be done in this area, and ease of comparison between leaf and canopy fluxes make the EcoCELLs an outstanding tool for pursuing these questions, particularly at a mechanistic and process model level. To our knowledge, this facility is the first that will allow the direct collection of validation data for canopy process models, and thus represents a unique opportunity for this type of research.

The need for process-level models of plant response to environmental change was one of the fundamental motivations for the construction of the EcoCELLs. Focussing on the underlying mechanisms of plant response to environmental variation may lead to the generalizations needed to predict the biosphere response to global change (Field & Ehleringer 1993). Recently, field studies with open-top chambers have made significant progress in the measurement of whole-system fluxes (Drake 1992; Ham *et al.* 1995; Körner 1995b as reported in Körner 1995a). Although these studies can monitor overall responses quite well, information on processes and mechanisms is often limited or lacking. This occurs because open-top-chamber measurements of fluxes are often limited by precision, time and space integration, signal-to-noise ratio, the degree of environmental control and often an inability to isolate soil processes. The EcoCELLs were designed to address these limitations, and the results of the initial testing and

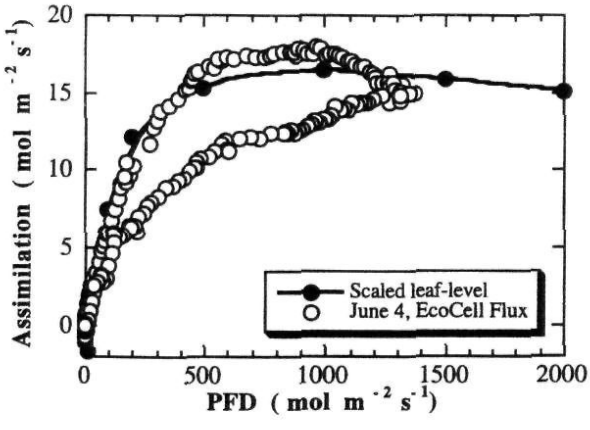

Figure 8. Comparison of the scaled leaf-level light response (from Fig. 7, lower panel) and the whole-system level response measured on a clear day immediately following the leaf level measurements (4 June 1995).

the 'proof of concept' experiment suggest that to a large extent these limitations are greatly reduced or eliminated.

Much of our knowledge of leaf-level physiological responses at the process level has come from careful experimentation with single-factor manipulations. For example, photosynthetic CO_2 response curves are widely used in CO_2 research to investigate the role of photosynthetic regulation in the CO_2 response of individual plants. Response curves provide a useful tool for breaking down the general response into its mechanistic components, furthering our understanding of the problem. Undoubtedly, as the interest in larger spatial scale processes drives future research, unforeseen complexities and interactions will be found, and again response curves, obtained under carefully controlled conditions at the canopy and system scales, can further our understanding. The ability of the EcoCELLs to produce such response curves is a significant advantage of this technology. The EcoCELLs are currently capable of light, temperature, relative humidity or vapour pressure deficit and CO_2 response curves (from ambient CO_2 partial pressures and up). Furthermore, the nature of the controlled-environment facility and the natural climate (light environment) of Nevada allow these measurements to be repeated day after day.

One limitation of the EcoCELLs is the lack of natural soils and the synthetic nature of the systems studied. The system does have the ability to mimic natural systems in real time, tracking environmental conditions via telecommunications from a remote weather station. There are several other possibilities for innovative studies of soils and below-ground processes. For example, if natural soils are seen as the highest priority, the possibility exists to place large soil monoliths into the soil containers. Another alternative is to use low-carbon soils, such as decomposed granite in conjunction with plant communities naturally occurring on such soils, such as aspen. This soil system facilitates the study of below-ground carbon dynamics, since the low-carbon soil would increase the biological signal relative to the soil background. Yet another alternative is to use the isotopic signature of the carbon in the soil to distinguish the signal from the background. Recent advancements have been made in this area using soils from C_4 grasslands in conjunction with experiments on C_3 species (Balesdent & Balabane 1992; Becker-Heidmann & Scharpenseel 1992; McPherson, Boutton & Midwood 1993; Wedin *et al.* 1995). The synthetic nature of the soil system represents one of the many trade offs of controlled-environment facilities. Although the system is synthetic, it is advantageous for direct manipulations of the soil and has greater accessibility.

The above-ground portion of the system is also subject to potential limitations or complications when trying to mimic 'natural' conditions. For example, technologies such as the EcoCELLs that enclose leaves, canopies or systems can dramatically alter the coupling between the atmosphere and the vegetation. Enclosure of the canopy within a highly stirred 'cuvette' influences the physical properties of the boundary layer, changing the diffusion gradients for CO_2, H_2O and other trace gases of interest. Ultimately these properties influence mass and energy exchange between the vegetation and its environment. Furthermore, many of these parameters, and therefore their effects, are difficult to measure. Fortunately, within the EcoCELLs these variables can be manipulated by adjusting the total air flow through the system and differentially vectoring the air over the canopy. Single-factor manipulations of these variables allow the quantification of their effects, and then in conjunction with the other lower impact technologies (e.g. eddy flux correlation and FACE) a predictive understanding of natural systems may be developed.

In a recent review, Lawton (1995) predicts that the use of model systems in controlled-environment facilities is likely to increase as the distinctions between laboratory mesocosms and field experiments continue to diminish as a result of technological advances. Acting as a bridge between theory and nature, Lawton further suggests that controlled-environment facilities can challenge our understanding of the complexities of natural systems. The EcoCELLs represent another technology that can play a vital role in this effort, joining the continuum of technologies from leaf-level studies, through pot and growth chamber studies, to greenhouse, open-top chambers and eventually Free Air Carbon Exchange studies. The strength of the EcoCells is in studying process-level problems under controlled-environmental conditions at a scale previously untractable. The EcoCELLs are particularly well suited to measure the flux of carbon and water for the construction of mass and energy budgets. Interactions between carbon, water and nitrogen are undoubtedly regulating ecosystem performance and response to global change. The EcoCELLs should prove to be an invaluable tool for the development and testing of process-level models to explain these interactions, and ultimately for scaling these mechanistic responses from the leaf and pot levels to the canopy and system levels.

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REFERENCES

Allen L.H., Jr (1975) Line-source carbon dioxide release. III. Prediction by a two dimensional numerical diffusion model. *Boundary-Layer Meteorology* **8,** 39–79.

Allen L.H. Jr, Drake B.G., Rogers H.H. & Shinn J.H. (1992) Field techniques for exposure of plants and ecosystems to elevated CO_2 and other trace gases. *Critical Reviews in Plant Sciences* **11,** 85–119.

Baldocchi D.D. (1993) Scaling water vapour and carbon dioxide exchange from leaves to a canopy: Rules and tools. In *Scaling Physiological Processes: Leaf to Globe* (eds J. R. Ehleringer & C. B. Field), pp. 77–114. Academic Press, New York.

Balesdent J. & Balabane M. (1992) Maize root-derived soil organic carbon estimated by natural ^{13}C abundance. *Soil Biology and Biochemistry* **24,** 97–101.

Barnola J.M., Raynaud R., Korotkevich Y.S. & Lorius C. (1987) Vostock ice core provides 160,000 year record of atmospheric CO_2. *Nature* **329,** 408–414.

Barton C.V.M., Lee H.S.J. & Jarvis P.G. (1993) A branch bag and CO_2 control system for long-term CO_2 enrichment of mature Sitka spruce (*Picea sitchensis* (Bong.) Carr.): Technical report. *Plant, Cell and Environment* **16,** 1139–1148.

Becker-Heidmann P. & Scharpenseel H. (1992) The use of natural ^{14}C and ^{13}C in soils for studies on global climate change. *Radiocarbon* **3,** 535–540.

Billings W.D., Luken J.O., Mortensen D.A. & Peterson K.M. (1982) Arctic tundra: A source or sink for atmospheric carbon dioxide in a changing environment? *Oecologia* **53,** 7–11.

Billings W.D, Luken J.O., Mortensen D.A. & Peterson K.M. (1983) Increasing atmospheric carbon dioxide: Possible effects on Arctic tundra *Oecologia* **58,** 286–289.

Billings W.D, Peterson K.M., Luken J.O. & Mortensen D.A. (1984) Interaction of increasing atmospheric carbon dioxide and soil nitrogen on the carbon balance of tundra microcosms. *Oecologia* **65,** 26–29.

Daudet F.-A. & Claustres J.P. (1995) Continuous monitoring of gas exchange in open-top chambers in pollution experiments: methodological aspects. *New Phytologists* **130,** 81–88.

Drake B.G., Rogers H.H. & Allen L.H. Jr (1986) Methods of exposing plants to elevated carbon dioxide. In *Direct Effects of Increasing Carbon Dioxide on Vegetation* (eds B. R. Strain & J. D. Cure), pp. 11–13. U.S. Department of Energy, Carbon Dioxide Research Division, Washington D.C.

Drake B.G. (1992) A field study of the effects of elevated CO_2 on ecosystem processes in a Chesapeake Bay wetland. *Australian Journal of Botany* **40,** 579–595.

Drake B.G., Leadley P.W., Arp W.J., Nassiry D. & Curtis P.S. (1989) An open top chamber for field studies of elevated atmospheric CO_2 concentration on saltmarsh vegetation. *Functional Ecology* **3,** 363–371.

Dufrene E., Pontailler J.Y. & Saugler B. (1993) A branch bag technique for simultaneous CO_2 enrichment and assimilation measurements on beech (*Fagus sylvatica* L.). *Plant, Cell and Environment* **16,** 1131–1138.

Field C.B. & Ehleringer J.R. (1993) Introduction: Questions of Scale. In *Scaling Physiological Processes: Leaf to Globe* (eds J. R. Ehleringer & C.B. Field), pp. 1–4. Academic Press, New York.

Field C.B., Ball J.T. & Berry J.A. (1991) Photosynthesis: principles and field techniques. In *Plant Physiological Ecology: Field Methods and Instrumentation.* (eds R. W. Pearcy, J. Ehlringer, H. A. Mooney & P. W. Rundel), pp. 209–253. Chapman and Hall, London.

Field C.B., Jackson R.B. & Mooney H.A. (1995) Stomatal responses to increased CO_2: Implications from the plant to the global scale. *Plant, Cell and Environment* **18,** 1214–1225.

Grulke N.E., Riechers G.H., Oechel W.C., Hjelm U. & Jaeger C. (1990) Carbon balance in tussock tundra under ambient and elevated atmospheric CO_2. *Oecologia* **83,** 485–494.

Ham J.M., Owensby C.E., Coyne P.I. & Bremer D.J. (1995) Fluxes of CO_2 and water vapour from a prairie ecosystem exposed to ambient and elevated atmospheric CO_2. *Agricultural and Forest Meteorology* **77,** 73–93.

Hellmers H. & Giles L.J. (1979) Carbon dioxide: critique I. In *Controlled Environment Guidelines for Plant Research* (eds T. W. Tibbitts & T. T. Kozlowski), pp. 229–234. Academic Press, New York.

Hendry G.R., Lewin K.F. & Nagy J. (1993) Free air carbon dioxide enrichment: development, progress, results. *Vegetatio* **104/105,** 17–31.

Hunt H.W., Detling J.K., Elliott E.T., Monz C.A. & Strain B.R. (1990) The effects of elevated carbon dioxide and climate change on grasslands. I. Response of above ground primary production in intact sods of native shortgrass prairie. *Bulletin of the Ecological Society of America* **71(Suppl.),** 196.

Jarvis P.G. (1995) Scaling processes and problems. *Plant, Cell & Environment* **18,** 1079–1089.

Jouzel J., Barkov N.I., Barnola J.M., Bender M., Chappellaz J., Genthon C., Kotlyakov V.M., Lipenkov V., Lorius C., Petit J.R., Raynaud D., Raisbeck G., Ritz C., Sowers T., Stievenard M., Yiou F. & Yiou, P. (1993) Extending the Vostock ice-core record of palaeoclimate to the penultimate glacial period. *Nature* **364,** 407–412.

Keeling C.D., Whorf T.P., Wahlen M. & van der Plicht, J. (1995) Interannual extremes in the rate of rise of atmospheric carbon dioxide since 1980. *Nature* **375,** 666–670.

Kimball B.A. (1992) Cost comparison among free-air CO_2 enrichment, open-top chamber and sunlit controlled-environment chamber methods of CO_2 exposure. *Critical Reviews in Plant Sciences* **11,** 265–270.

Körner Ch. & Arnone J.A. III. (1992) Responses to elevated carbon dioxide in artificial tropical ecosystems. *Science* **257,** 1672–1675.

Körner Ch., Arnone J.A. & Hilti W. (1993) The utility of enclosed artificial ecosystems in CO_2 research. In *Design and Execution of Experiments on CO_2 Enrichment* (eds E. D. Schulze & H. A. Mooney), pp. 407–413. Ecosystems Research Report 6, Environmental Research Programme. Commission of the European Communities, Brussels.

Körner Ch. (1995a) Towards a better experimental basis for upscaling plant responses to elevated CO_2 and climate warming. *Plant, Cell and Environment* **18,** 1101–1110.

Körner Ch. (1995b) The response of complex multispecies systems to elevated CO_2. In *Global change and Terrestrial Ecosystems* (eds B. H. Walker & W. L. Steffen). Cambridge University Press, Cambridge, in press.

Lawton J.H. (1995) Ecological experiments with model systems. *Science* **269,** 328–331.

Leadley P.W. & Drake B.G. (1993) Open top chambers for exposing plant canopies to elevated CO_2 concentrations and for measuring net gas exchange. *Vegetatio* **104/105,** 3–16.

McLeod A.R. & Fackrell, J.E. (1983) *A Prototype System for Open-Air Fumigation of Agricultural Crops. 1: Theoretical Design (TPRD/L/2475/N83).* UK Central Electricity Research Laboratories.

McLeod A.R., Alexander K. & Hatcher P. (1983) *A Prototype System for Open-Air Fumigation of Agricultural Crops. 2: Construction and Description (TPRD/L/2475/N83).* UK Central Electricity Research Laboratories.

McPherson G.R., Boutton T.W. & Midwood A.J. (1993) Stable carbon isotope analysis of soil organic matter illustrates vegetation changes at the grassland/woodland boundary in southeastern Arizona, USA. *Oecologia* **93,** 95–101.

Mooney H.A. (1991a) Biological response to climate change: An agenda for research. *Ecological Applications* **1,** 112–117.

Mooney H.A. (1991b) Plant physiological ecology-determinants of progress. *Functional Ecology* **5,** 127–135.

Navas M.L., Guillerm J.L., Fabregettes J. & Roy J. (1995) The influence of elevated CO_2 on community structure, biomass and carbon balance of Mediterranean old-field microcosms. *Global Change Biology* **1**, 325–335.

Norman J. M. (1993) Scaling processes between leaf and canopy levels. In *Scaling Physiological Processes: Leaf to Globe* (eds J. R. Ehleringer & C. B. Field), pp. 41–76. Academic Press, New York.

Oechel W.C., Riechers G., Lawrence W.T.. Prudhomme T.I., Grulke N. & Hastings S.J. (1992) 'CO_2LT' an automated, null-balance system for studying the effects of elevated CO_2 and global climate change on unmanaged ecosystems. *Functional Ecology* **6,** 86–100.

Oechel W.C., Hastings S.J., Vourlitis G., Jenkins M., Reichers G. & Grulke N. (1993) Recent change of Arctic tundra ecosystems from a net carbon dioxide sink to a source. *Nature* **361,** 520–523.

Oechel W.C., Cowles S., Grulke N., Hastings S.J., Lawrence B., Prudhome T., Reichers G., Strain B., Tissue D. & Vourlitis G. (1994) Transient nature of CO_2 fertilization in Arctic tundra. *Nature* **371,** 500–502.

Ojima D.S., Kittel T.G.F., Rosswall T. & Walker B.H. (1991) Critical issues for understanding global climate change effects on terrestrial ecosystems. *Ecological Applications* **1,** 316–325.

Overdieck D. & Reining E. (1986) Effect of atmospheric CO_2 enrichment on perennial ryegrass (*Lolium perenne*) and white clover (*Trifolium repens* L.) competing in managed model-ecosystems. I. Phytomass production. *Acta Oecologia* **7,** 26–35.

Owensby C.E, Coyne P.I., Ham J.M., Auen L.M. & Knapp A.K. (1993) Biomass production in a tallgrass prairie ecosystem exposed to ambient and elevated CO_2. *Ecological Applications* **3,** 644–653.

Reece C.F., Krupa S.V., Jager H.J., Roberts S.W., Hastings S.J. & Oechel W.C. (1995) Evaluating the effects of elevated levels of atmospheric trace gases on herbs and shrubs: A prototype dual array field exposure system. *Environmental Pollution* **90,** 25–31.

Rogers H.H., Heck W.W. & Heagle A.S. (1983) A field technique for the study of plant responses to elevated carbon dioxide concentration. *Journal of the Air Pollution Control Association* **33,** 42–44.

Schäppi R. & Körner Ch. (1996) Growth responses of an alpine grassland to elevated CO_2. *Oecologia* **105,** 43–52.

Schulze E.-D. & Mooney H.A. (1993) Comparative view on design and execution of experiments at elevated CO_2. In *Design and Execution of Experiments on* CO_2 Enrichment (eds E. D. Schulze & H. A. Mooney), pp. 407–413. Ecosystems Research Report 6, Environmental Research Programme. Commission of the European Communities, Brussels.

Strain B.R., Allen L.H. Jr, Baldocchi D., Bazzaz F., Burke J., Dahlman R., Denmead T., Hendrey G., McLeod A., Melillo J., Oechel W., Risser P., Rogers H., Rozema J. & Wright R. (1991) Available technologies for field experimentation with elevated CO_2 in global change research. In *Ecosystem Experiments*, SCOPE/ICSU Vol. 45(eds. H.A. Mooney, E. Medina. D.W. Schindler, E.-D. Schulze & B.H. Walker), pp. 245–261. John Wiley and Sons, New York.

Strain B.R. & Thomas R.B. (1992) Field measurements of CO_2 enhancement and change in natural vegetation. *Water, Air and Soil Pollution* **64,** 45–60.

Teskey R.O., Dougherty P.M. & Wiselogel A.E. (1991) Design and performance of branch chambers suitable for long-term ozone fumigation of foliage in large trees. *Journal of Environmental Quality* **20,** 591–595.

Tissue D.T. & Oechel W.C. (1987) Physiological response of *Eriophorum vaginatum* to elevated CO_2 and temperature in the Alaskan tussock tundra. *Ecology* **68,** 401–410.

Terashima I. & Hikosaka K. (1995) Comparative ecophysiology of leaf and canopy photosynthesis. *Plant, Cell and Environment* **18,** 1111–1128.

Wedin D.A., Tieszen L.L., Dewey B. & Pastor J. (1995) Carbon isotope dynamics during grass decomposition and soil organic matter formation. *Ecology* **76,** 1383–1392.

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