

# A Field Technique for the Study of Plant Responses to Elevated Carbon Dioxide Concentrations

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The global rise in carbon dioxide (CO<sub>2</sub>) is a well established phenomenon. The preindustrial level of CO<sub>2</sub> was probably around 295 ppm; that level has risen to over 335 ppm and projections indicate it will reach 600 ppm by the year 2030. This has enormous implications for plants which form the base of the Earth's biosphere. The starting point for any assessment of CO<sub>2</sub> effects on the biosphere should be plant responses. Carbon dioxide is a chief input to photosynthesis, the solar energy trapping process of plants. This key role makes it imperative that plant responses to elevated CO<sub>2</sub> levels be well understood as energy policies are set.

There is a paucity of field data on this important topic. Therefore, systems that permit the long-term study of plant response in the field are needed. Recent field work in our laboratory has led to the development of an approach which is described here.

The general approach for this work was to develop a plant exposure system for the generation of large-scale CO<sub>2</sub> test atmospheres in the field in order to continuously expose plants within the system over their growing season. The system has to be cost effective and reliable over long time periods, and had to provide an environment as identical to field conditions as possible.

The open top field chamber as described by Heagle *et al.*<sup>1,2</sup> was selected for use as the plant exposure unit. Much experience with this chamber design has been accumulated from air pollution/plant effects studies in the field. In our opinion, this type of chamber represents the best available technology and is currently in use at numerous laboratories throughout the country.

The specific aim of this investigation, then, was the development of field techniques that would permit the study of elevated CO<sub>2</sub> levels on plants. The design, construction, and testing of a system for dispensing and monitoring CO<sub>2</sub> in the number of open top chambers are discussed below.

## System Development

### Liquid Carbon Dioxide Supply

A 14 t (12.7 metric t) liquid receiver (Airco Industrial Gases) served as a CO<sub>2</sub> supply reservoir. This storage unit operates between -23°C and -16°C, which corresponds to pressures

of 242.8 psig (17.1 kgcm<sup>-2</sup>) and 306.8 psig (21.6 kgcm<sup>-2</sup>). Besides having a low temperature air cooled condensing unit, the receiver is also equipped with a vaporizer. These are automatically controlled and the vaporizers have delivery capacities up to 6000 lb (2722 kg)/h. The vessel pressure and contents are read out on gauges. Safety relief valves are either direct spring loaded or pilot operated. Electrical requirements are usually 208/220/460, 60 cycle, 3 phase, but conversion to single phase is possible.

Delivery from the chemical preparation plant to the receiver is by transfer tank truck. (Such delivery could present a problem in remote locations.) The CO<sub>2</sub> vapor is pumped out while liquid is pumped in.

### High Volume Dispensing Manifold

Carbon dioxide gas from the receiver was delivered through an 8 m run of 1.27 cm copper tubing (Type K) to the dispensing manifold. Figure 1 shows the CO<sub>2</sub> dispensing pathway. The tubing was wrapped with 15 m of conventional unthermostated heat tape (16 w/m). The prevented condensation on the manifold components which would otherwise become quite heavy during cool periods when humidity was high. Between the copper delivery tube and the manifold was a 1.59 cm normally closed solenoid valve. This valve stopped CO<sub>2</sub> flow during power failure.

The custom-made high volume dispensing manifold (Figure 1; Airco Industrial Gases, Research Triangle Park, NC) consisted of three high pressure/stainless steel regulators (Airco Model 57-300), each feeding (at 150 psig) three single-stage line regulators (Airco Model 806-9975). These nine regulators fed (at 30 psig) nine dual range flow meters (Airco Model 805-1606). Each flow meter provided two calibrated ranges 0.025 to 0.400 scfm and 0.256 to 1.280 scfm. A handwheel on each simplified the changing of ranges. A 0.58 cm Impolene (polyallomer, black; Gould Imperial-Eastman) tube led from each flow meter to the inlet of each exposure chamber. The entire unit was assembled and pre-tested before being put into use.

### Plant Exposure Chamber

The basic exposure unit of our system is an open top chamber (Figure 2). These chambers, which are essentially open ended cylindrical baffles, were developed in our labo-

ratory<sup>1,2</sup> and have been recommended for elevated CO<sub>2</sub> research in agriculture by Hardy and Havelka.<sup>3</sup> Tests at our laboratory have shown that plant growth variables measured within these enclosures are similar to those of plants grown outside them.

The chambers, which are 3 m in diameter and 2.4 m in height, are constructed of a structural aluminum frame covered by an 8 mil clear PVC plastic film (Roll-A-Glass, Tenneco; Livingston Coating Corporation, Charlotte, NC). The bottom half of each chamber cover is doubled-walled; the inside wall is perforated with 2.5 cm diameter holes and serves

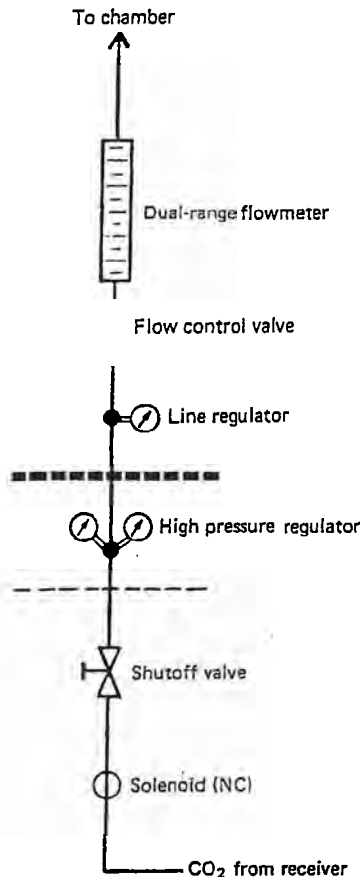


Figure 1. This schematic shows the system that regulates the flow of CO<sub>2</sub> to the plant exposure chambers.

as a duct that distributes air uniformly into the chamber (Figure 2). Air to this duct is supplied from a 0.75 hp axial fan that is mounted in a sheet metal plenum box with a particulate filter. Carbon dioxide from the receiver is injected into the plenum box ahead of the fan to assure thorough mixing. The CO<sub>2</sub> empties about 1/4 way across the vertical center line of the plenum box to enhance mixing. Additional information on the open top chamber can be found in Heagle *et al.*<sup>1</sup>

#### Monitoring

Fifteen test plots were used in a preliminary study, 12 with chambers and 3 without (open field plots). Three replicates of 4 desired CO<sub>2</sub> concentrations (daytime values) were set up: ambient (~335 ppm), ambient + 200 ppm (~535 ppm), ambient + 400 ppm (~735 ppm), and ambient + 600 (~935 ppm). The level of CO<sub>2</sub> in each of the 15 plots was monitored in a 30 min cycle, each of the plots being sampled for 2 min. Sampling intakes [4.2 cm clear polystyrene (Gelman 4338 two-piece air-sampling field monitor) disc filter holder with a glass fiber filter] were located at chamber center about 1 m from the

ground. The disc filters were custom-made. Impolene lines (0.64 cm O.D., polyallomer, black; Gould Imperial-Eastman) delivered sample air at 5 L/min from the intakes to the sampling manifold.

Carbon dioxide was monitored around the clock for the duration of the study. The CO<sub>2</sub> sampling pathway is schematically presented in Figure 3. A time-shared sampling manifold was used to sequentially sample the study plots. Samples were drawn through 3-way 24-VDC stainless steel solenoids (Versa Valves ESM-8302) with 0.24 cm orifices and normally closed. A Metal Bellows pump (MB-302) supplied vacuum to this solenoid bank so that 5 L/min were continuously pulled through each and into a flow meter with built-in adjustment valve (Dwyer Model VFA-BV-23, Visi-float, 0.6 to 5.0 L/min).

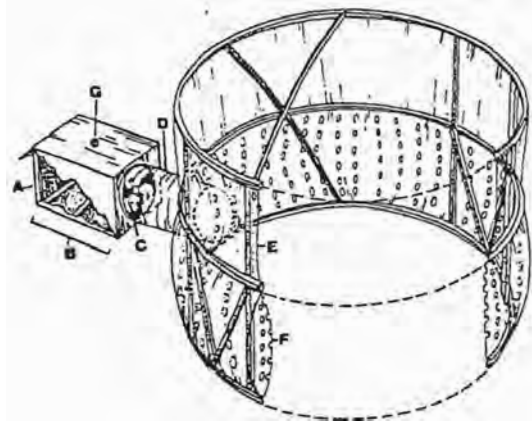


Figure 2. This cylindrical chamber consists of aluminum frame covered by the clear PVC plastic film Roll-A-Glass. A. Fiberglass particulate filter, B. Sheet metal box, C. 0.75 HP Axial fan, D. Connecting duct, F. Upper panel, and G. CO<sub>2</sub> injection port.

The solenoids were actuated sequentially for 2 min each by a custom-made digital timer. A safety relay stopped power to the pump and solenoids when a power failure had occurred. This assured that at no time would the pumps be pulling against a dead wall of closed solenoids. Of the 15 sample points, one was always being sampled while the remaining ones were exhausted. This assured that minimal line flushing has to occur before an accurate sample was read. The sample being measured was diverted to the sampling instruments. A Metal Bellows pump (MB-41) pulled the sample stream and a Neptune Dynapump (Universal Electric Model 3) diverted part of this stream through a dew point hygrometer (EG&G) and an infrared CO<sub>2</sub> analyzer (Horiba).

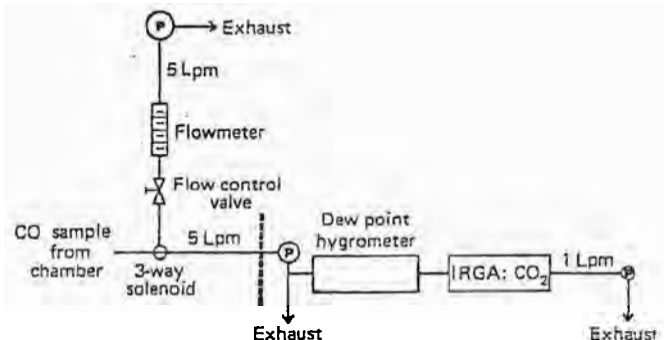


Figure 3. Schematic of CO<sub>2</sub> monitoring system. There are 15 units of each component to the left of the vertical dashed line. A digital clock controls the actuation of the 3-way solenoids to sequentially sample the 15 plots, air flow being constantly maintained to assure sample delivery as near real-time as possible.

## Performance of the System

The system performed satisfactorily in the generation of large-scale CO<sub>2</sub> test atmospheres in the field. Once in place, maintenance and operation of the system presented no major difficulties.

No special problems were encountered with the receiver. It is large (2.6 × 5.6 × 2.3 m) and heavy (7 metric t, empty), so transport to remote study areas could be difficult. Some sealing of minor leaks and replacement of fuses had to be done. Also for long term studies, provision for the delivery of large amounts of liquid CO<sub>2</sub> (e.g., 12.7 metric t to fill receiver) would have to be made. In some areas, electrical power for the CO<sub>2</sub> receiver would be a problem. However, in many areas that should be studied, a liquid CO<sub>2</sub> receiver would be the best method by which to store and supply CO<sub>2</sub> to test chambers.

In using a receiver, pressure drops leading to solidification of the liquid CO<sub>2</sub> must be avoided. Mechanical and electrical systems prevent this, but daily checks of pressure are still good practice.

performance and to identify and solve associated system problems.

Our work with soybean, corn, pine, and sweetgum over a full growing season has shown substantial increases in biomass, changes in plant form and leaf anatomy, and increases in water use efficiency (mg C fixed/g H<sub>2</sub>O consumed); yield increases were observed for the crops.<sup>4,5</sup>

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Table I. Seasonal CO<sub>2</sub> concentrations: means ± standard deviations (ppm) for each of three replications; derived from values read at 2 h intervals on every 10th day over a 100 day period.

Rep	Time	Ambient plot	ppm CO <sub>2</sub> added to chambers			
			+0	+200	+400	+600
1	Day <sup>a</sup>	344 ± 18	343 ± 17	521 ± 30	714 ± 52	905 ± 45
	Night <sup>b</sup>	367 ± 28	367 ± 29	567 ± 66	766 ± 67	942 ± 64
2	Day	339 ± 9	338 ± 8	524 ± 32	707 ± 48	919 ± 61
	Night	369 ± 31	369 ± 30	570 ± 57	757 ± 69	984 ± 53
3	Day	336 ± 7	340 ± 10	515 ± 30	733 ± 58	905 ± 54
	Night	367 ± 31	374 ± 34	549 ± 55	777 ± 88	934 ± 68

<sup>a</sup> Day: 6:00 A.M. to 8:00 P.M.

<sup>b</sup> Night: 8:00 P.M. to 6:00 A.M.

Carbon dioxide concentrations at 15 test points (3 ambient and 12 in open top chambers) were monitored throughout the study period of 3 mo. Means and standard deviations for 3 replicates of the 5 treatments are given in Table I. Variation is seen to increase at night and with rising CO<sub>2</sub> level. Good separation between the exposure levels was obtained.

No operational problems were encountered with the dispensing manifold. Higher resolution flow meters would be desirable. In the case of the sampling system, some condensation was observed in the delivery lines within the air conditioned lab trailer, but this was solved by stripping the lines with unthermostated heat tape (16 w/m).

Calibration of the infrared CO<sub>2</sub> analyzer was accomplished with a series of high pressure tanks of CO<sub>2</sub> in air. Six cylinders, one of nitrogen which served as a zero, were equipped with two-stage regulators and Kwik-connects (Gould Imperial-Eastman). This made switches between the various calibration tanks convenient so that little time was needed for calibration.

## Conclusion

The open top chamber system designed for dispensing and monitoring of CO<sub>2</sub> into open top chambers performed satisfactorily in the generation of large-scale CO<sub>2</sub> test atmospheres in the field. Good separation was obtained for each of the treatment levels. Concentrations of CO<sub>2</sub> in the ambient plots and in the chambers to which no CO<sub>2</sub> was added were essentially the same. Although CO<sub>2</sub> levels were manually controlled, levels obtained were near those desired.

Work on the system in our laboratory is continuing. The work has a dual purpose, to understand open top chamber

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