Seed yield of soybeans with daytime or continuous elevation of carbon dioxide under field conditions

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Abstract

Some studies of responses of plants to elevated concentrations of carbon dioxide (EC) added CO_2 only in the daytime, while others supplied CO_2 continuously. I tested whether these two methods of EC treatments produced differences in the seed yield of soybeans. Tests were conducted for four growing seasons, using open top chambers, with soybeans rooted in the ground in field plots. One third of the chambers were flushed with air at the current ambient [CO_2] (AC), one third had [CO_2] 350 µmol mol^{-1} above ambient during the daytime (EC_d), while one third had [CO_2] 350 µmol mol^{-1} above ambient for 24 h per day (EC_{dn}). EC_{dn} increased seed yield by an average of 62 % over the four years compared with the AC treatment, while EC_d increased seed yield by 34 %. Higher seed yield for EC_{dn} compared with EC_d occurred each year. In comparing years, the relative yield disadvantage of EC_d decreased with increasing overall seed yield. On days with high water vapor pressure deficits, soybean canopies with EC_d had smaller midday extinction coefficients for photosynthetically active radiation than canopies with EC_{dn}, because of a more vertical leaf orientation. Hence the seed yield of soybean at EC varied depending on whether EC was also provided at night, with much greater yield stimulation for EC_{dn} than for EC_d in some years.

Additional key words: *Glycine max*; photosynthetically active radiation; precipitation; rainfall; stem; vapor pressure deficit.

Introduction

There have been numerous field studies of the response of the seed yield of soybean to elevated concentrations of carbon dioxide (EC) which have been conducted using open top chambers (reviewed in Ainsworth *et al.* 2002). Because open top chambers provide somewhat unnatural conditions of air speed, radiation, and air temperature, free air carbon dioxide enrichment systems have been developed, which eliminate many of the chamber effects. However, in environments with low wind speeds at night, providing EC at night in free air systems requires artificial air movement to distribute the CO_2 across the plot, which diminishes one of the advantages of the free air system. It might also seem unnecessary, since night time [CO_2] are often quite high without any addition of CO_2 (Ziska *et al*. 2001). Additionally, because providing EC more hours per day is expensive, many free air CO_2 enrichment systems provide EC only during the daytime (*e.g.* Luscher *et al.* 1998, Caemmerer *et al.* 2001, Lee *et al.* 2001, Takeuchi *et al.* 2001, Leakey *et al.* 2004). The present study was designed to test whether the seed yield of soybean would be the same whether EC was provided continuously (EC_{dn}) or only in the daytime (EC_d).

Several studies have indicated that plant growth may respond to [CO_2] during the dark (reviewed in Bunce 2003). In soybean, this seems to be particularly true when the daytime [CO_2] is elevated (Bunce 2001). Heagle *et al.* (1999) reported a trend toward increased growth and seed yield in soybean with EC_{dn} compared with EC_d, but the data were inconclusive. One possible mechanism by which [CO_2] in the dark may affect plants is through effects on respiration (Reuveni *et al.* 1997), although changes in respiration did not occur in all studies in which plant growth was affected by night time [CO_2] (Bunce 2003). In red maple seedlings, [CO_2] at night affected leaf angle during the daytime (Bunce 2003), which could affect photon interception. In this study, radiation penetration through the soybean canopies was also compared for the EC_{dn} and EC_d treatments.

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Materials and methods

Studies were conducted using open top chambers at the South Farm of the Beltsville Agricultural Research Center, Beltsville, Maryland, for four years, 2000–2003. Soybean, *Glycine max* L. Merr. cv. Clark was grown in 30 cm rows, at an overall density of 25 plants per square meter. Seeds were planted near mid-June in all years except 2001, when they were planted on 15 May. Plants were inside open top chambers made of clear acrylic, which covered 1.3 or 1.9 m^2 of ground, depending on year, and were 1.8 m high. All chambers were flushed continuously at a flow rate of 6 m^3 per minute with air pulled from outside the chambers by blowers. One third of the chambers received no additional CO_2 (AC), and two thirds of the chambers had pure CO_2 injected into the inlet air stream at a flow rate sufficient to increase the $[CO_2]$ by 350±50 µmol mol^{-1} above that of the ambient air (EC). Half of the EC chambers received the added CO_2 continuously (EC_{dn}), and half received added CO_2 for only 14 h per day centered on noon (EC_d). Plots were fertilized with potassium and phosphate at recommended rates, but no nitrogen was added. Plots were weeded by hand, and received normal precipitation. The $[CO_2]$ of each chamber was sampled every 45 min by an absolute infrared analyzer located in an adjacent air conditioned building, and CO_2 flow rates were adjusted daily as needed. The $[CO_2]$ of the ambient air averaged 374 µmol mol^{-1} in the daytime and 450 µmol mol^{-1} at night. Night-time values ranged from 355 to 602 µmol mol^{-1}, with lower values at higher wind speeds.

Near mid season, midday measurements of photosynthetically active radiation (PAR) above and below the canopies were combined with leaf area index (LAI) measurements to determine the extinction coefficient for PAR for each carbon dioxide treatment, using the following equation: (PAR below/PAR above) = exp(–K×LAI), where K is the extinction coefficient. Measurement dates were chosen for contrasting water vapor pressure deficits (VPD), and a total of four high VPD (2.5–3.0 kPa) and four low VPD (1.0–1.5 kPa) days were compared, over two seasons. PAR below the canopy was measured using a 1 m long sensor (*LI-191SA* Line Quantum Sensor, *LI-Cor*, Lincoln, NE, USA). LAI was determined the following morning, before sunrise, using an optical plant canopy analyzer (*LAI 2000* Plant Canopy Analyzer, *LI-Cor*, Lincoln, NE, USA).

Midday rates of net carbon dioxide assimilation of mature, fully exposed upper canopy leaves were determined for the AC, EC_{dn}, and EC_d treatments about every two weeks on clear days. Measurements were made at the ambient conditions of temperature and humidity and at the daytime $[CO_2]$. PAR exceeded 1 200 µmol m^{-2} s^{-1}. Measurements were made with a *CIRAS-1* portable photosynthesis system with automatic CO_2 control and a broad leaf cuvette (*PP Systems*, Haverhill, MA, USA). At least four leaves were measured for all treatments on each occasion.

When leaves had fallen and pods were mature, interior rows from each chamber were harvested to determine seed and stem dry mass per unit of ground area.

In 2001 there were four chambers per treatment in a randomized design. The harvest data from that year were analyzed using analysis of variance. In the other years, there was one chamber per treatment. Linear regression and *t*-tests of *arcsin* transformed percentages of seed yield for the different treatments were used to compare treatment effects over years, with one sample per year. Effects of the $[CO_2]$ treatments on extinction coefficients for photosynthetically active photon flux were analyzed separately for the high and low water vapor pressure deficit days, using analysis of variance, with one chamber measured per treatment on each of four days within each vapor pressure deficit group. Statistical tests were implemented using *JMP* v. *5* (*SAS Institute*, Cary, NC, USA).

Results

In 2001, the highest seed yield occurred in the EC_{dn} treatment, with lower yield for the AC and EC_d treatments (Table 1). Stem mass was highest in the EC_{dn} treatment and lowest for the AC treatment. The seed mass was a higher percentage of the total mass for the AC treatment than for either EC treatment (Table 1).

There was considerable variation in seed yield among years (Fig. 1). Yields were lowest in 2000, which was dry during pod filling in September (Table 2), and highest in 2003, which had abnormally abundant precipitation (42 % above normal) throughout the season. Despite the large year to year variation in yield, the rank order of seed yield of the $[CO_2]$ treatments was the same each year, with the EC_d yield greater than for the AC treatment, and the highest yield with EC_{dn} (Fig. 1). The EC_{dn}

Table 1. Seed yield and stem mass at final harvest in 2001 for soybeans grown at ambient $[CO_2]$ (AC), and elevated $[CO_2]$ provided continuously (EC_{dn}) or only in the daytime (EC_d). There were four replicate chambers per treatment. Means±SE; within columns, means followed by different letters were significantly different at p=0.05 using analysis of variance, and Tukey's HSD test. The seed % was transformed using *arcsin* before analysis.

$[CO_2]$ treatment	Seed [g m^{-2}]	Stem [g m^{-2}]	Seed % of total
AC	432±15 b	213±12 c	67±1 a
EC_{dn}	609±48 a	417±12 a	59±1 b
EC_d	475±41 b	306±39 b	61±3 b

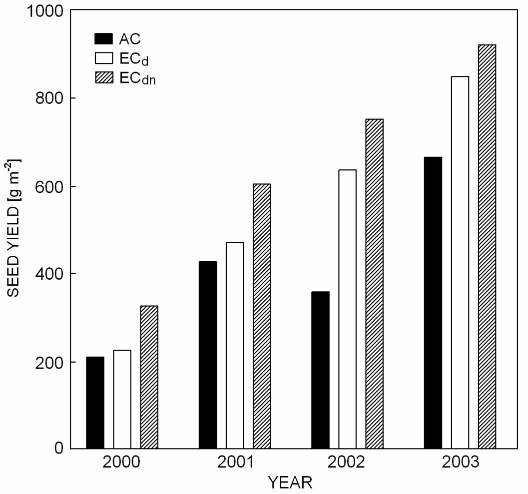

Fig. 1. Seed yield of soybean grown in open top chambers flushed with air at the ambient [CO_2] (AC), or with air elevated in [CO_2] either continuously (EC_{dn}) or only in the daytime (EC_d). EC was 350 µmol mol^{-1} above the AC.

increased seed yield by an average of 62 % over four years compared with AC, while EC_d increased seed yield by an average of 34 % compared with AC. The seed yield with EC_d averaged 81% of that for EC_{dn} over the four years. The probability of obtaining this observed mean percentage over four years under the hypothesis of a percentage of 100 % was <0.01, using a *t*-test on the *arcsin*-transformed percentages. The seed yield of the EC_d treatment increased as a percentage of that of the EC_{dn} treatment with increasing seed yield averaged over all treatments (Fig. 2).

The relative yield increase above AC for the EC_{dn} treatment only ranged from 41 to 55 % in 2000, 2001, and 2003, but was 110 % in 2002. The 2002 season had the highest temperatures among the four years for each month of the growing season (Table 1).

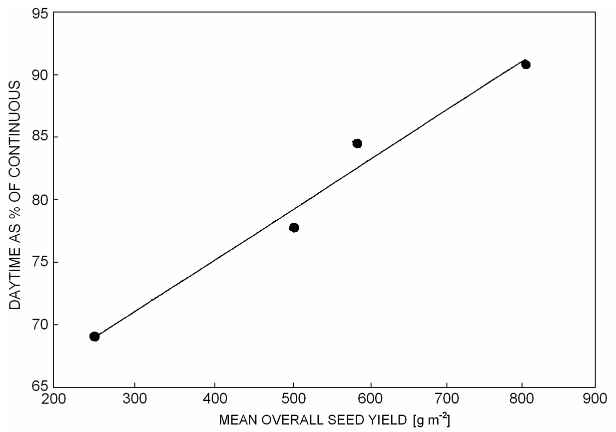

Fig. 2. The seed yield of the daytime elevation of [CO_2] (EC_d) treatment as a percentage of that of the continuously elevated [CO_2] treatment (EC_{dn}) in each of four years as a function of the mean seed yield over all [CO_2] treatments (AC, EC_d, EC_{dn}) each year. The linear regression shown has the equation: % = 58.400±0.042×mean yield, with r^2 = 0.978.

Table 2. Mean air temperature, T [°C], total precipitation, PPT [cm], and maximum days between rainfall events, D [d] during the 2001–2003 soybean growing seasons, at the South Farm of the Beltsville Agricultural Research Center, Beltsville, Maryland. "na" indicates not applicable to the soybean crop.

Year	June			July			August			September		
	T	PPT	D	T	PPT	D	T	PPT	D	T	PPT	D
2000	na	na	na	22.3	12.7	5	22.7	9.1	10	18.6	9.7	18
2001	22.7	10.2	8	22.4	12.3	8	24.3	11.2	10	18.3	6.2	9
2002	na	na	na	25.4	4.8	8	25.0	6.4	15	20.6	7.5	10
2003	na	na	na	24.1	12.5	8	24.3	8.1	8	19.8	13.2	7

Table 3. Extinction coefficients for photosynthetically active photon flux for soybean canopies exposed to ambient [CO_2] (AC), or elevated [CO_2] continuously (EC_{dn}) or only in the daytime (EC_d). Measurement days were grouped by the water vapor pressure deficit of the air into high (2.5–3.0 kPa) and low VPD (1.0–1.5 kPa) days. There were four days per VPD group. Means±SE; within a column, means followed by different letters were significantly different at *p*=0.05, using analysis of variance, and Tukey's HSD test.

[CO_2] treatment	High VPD days	Low VPD days
AC	0.700±0.022 b	0.648±0.029 b
EC_{dn}	0.768±0.010 a	0.837±0.025 a
EC_d	0.601±0.014 c	0.872±0.036 a

There were no significant differences between the EC_d and EC_{dn} treatments in net CO_2 assimilation rates measured at midday on any occasion. Mean values averaged over ten measurement dates were 42.4 and 42.2 µmol m^{-2} s^{-1} for the EC_d and EC_{dn} treatments, respectively, compared with the mean of 24.7 µmol m^{-2} s^{-1} for the AC treatment.

On days with high VPD, the EC_d treatment averaged lower extinction coefficients than the AC treatment, while the EC_{dn} treatment had slightly higher extinction coefficients than the AC treatment (Table 3). On low VPD days, both EC treatments averaged higher extinction coefficients than the AC treatment (Table 3).

Discussion

The results indicated that soybeans provided EC only during the daytime had substantially lower seed yields than plants provided with elevated [CO_2] continuously, especially in years when overall yields were low. In an earlier study using semi-open top chambers I found greater leaf area and shoot dry mass at flowering in soybeans given EC_{dn} than in plants given EC_d (Bunce 2001). Apparently this pattern sometimes continues on through seed production. It is common for the seed mass to be a smaller percentage of the total shoot mass at harvest at EC (Ainsworth *et al*. 2002), and this occurred in this study both with EC_d and with EC_{dn}.

I noticed more nearly vertical leaves on plants in the EC_d treatment than in the EC_{dn} treatment on many afternoons, and this was reflected in lower measured values of PAR extinction coefficients. Lower extinction coefficients with EC_d only occurred on days with high water vapor pressure deficits, for reasons that are not known. Soybean has pulvini capable of changing leaf orientation. Leaf orientation is partly under circadian control, but is also influenced by water status. Increasing water deficits result in a more vertical leaf orientation in soybean, which reduces the radiation load when transpiration is reduced by water stress. The lower extinction coefficients sometimes found in the EC_d treatment indicate reduced canopy absorption of radiation, and could contribute to lower canopy photosynthesis, total biomass production, and seed yield. I do not have enough information on the frequency of the occurrence of lower extinction coefficients with EC_d in the different years to determine how it might relate to the observed year to year variation in effects of EC_d *versus* EC_{dn} on seed yield.

Typical soybean seed yields in Maryland are about 300 g m^{-2}. Our yields for the AC treatment ranged from about two-thirds to twice this value, depending on the year. The relationship between the relative yields of the EC_d and EC_{dn} treatments suggests that in seasons with average yields at AC, the yield with EC_{dn} would be expected to be about 25 % higher than with EC_d. It is not known whether this effect of night time [CO_2] might differ with the level of [CO_2] elevation or for different cultivars, but these results suggest that caution should be used in interpreting soybean yield data obtained using systems in which EC is provided only in the daytime.

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