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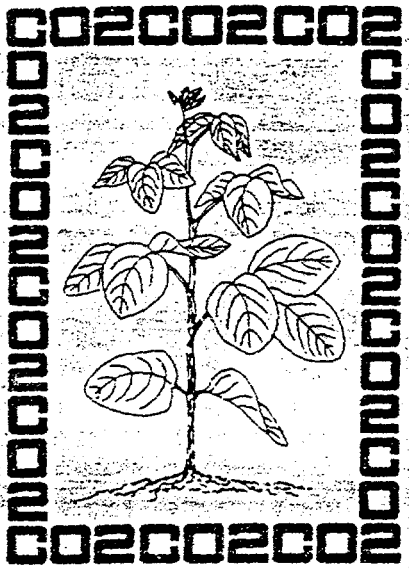

Response of Vegetation to Carbon Dioxide

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Field Studies of Plant Responses to Elevated Carbon Dioxide Levels 1984

DISCLAIMER

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I. INTRODUCTION

A. Rationale

The global carbon dioxide (CO_2) concentration is gradually increasing due to fossil fuel consumption, rapid advancement in industrialization and deforestation. It has been estimated that atmospheric CO_2 concentration will be doubled by 2025 (Clark et al., 1982). Extensive literature are now available to demonstrate that elevated CO_2 results in the increase of dry matter accumulation in both vegetative and reproductive components of the plants (Wittwer, 1980, 1983; see Kimball, 1983 for references).

Although there are a number of reports on the influence of atmospheric CO_2 enrichment on photosynthesis and short-term growth, very few reports are available on crops which have been subjected to CO_2 enrichment during their entire growing season. Furthermore, little information is available on plant responses to CO_2 enrichment under field conditions (Kramer, 1981; Strain et al., 1984). It is, therefore, imperative to have a better understanding of the effects of CO_2 enrichment on crop plants.

Reports on the influence of elevated CO_2 levels on tuber crops like radish (Raphanus sativus L. 'Whitetip'; Knecht, 1975) and potato (Solanum tuberosum, 'Kennebec'; Collins, 1976; Goudriaan and deRuiter, 1983) did not consider the effects of long-term exposure to elevated CO_2. Therefore, more information is needed to understand the overall effects of CO_2 enrichment on growth, development, and yield potential of root crops.

Sweet potatoes (Ipomoea batatas L.) and cowpeas (Vigna unguiculata L.) were selected as experimental plants because they represent major agricultural crops in southern United States. In addition, sweet potato is one of the world's major food crops. Currently, little is known about the responses of sweet potatoes and cowpeas to elevated CO_2 (Strain et al., 1984). Both crops have widespread growth ranges, and consequently can be easily grown for experimental purposes. In the present study, CO_2 enrichment has been applied to sweet potatoes and cowpeas in order to investigate its effect on their growth, physiology, and yield under field condition.

B. Objectives

1. To establish at Tuskegee Institute the facilities for growing crops in the field under enriched CO_2 atmospheric conditions..
2. To obtain field data on the morphological, physiological, biochemical, growth and yield responses of sweet potatoes and cowpeas to elevated levels of CO_2.
3. To determine the effects of elevated CO_2 in the rate of nitrogen fixation of cowpeas.
4. To provide data for a generalized crop growth model for predicting yield of both sweet potatoes and cowpeas as a function of atmospheric CO_2 enrichment.

C. Approach

The main aim of this investigation was to utilize a plant exposure unit for the generation of a large-scale CO_2 test environment system in

the field (Rogers et al., 1980), and to grow plants continuously within the system during an entire growing season. Heagle et al. (1973) described the design of open top chambers suitable for this kind of study. A similar type of design was adopted for the plant exposure study at Tuskegee Institute.

II. CO_2 EXPERIMENTAL SYSTEM

A. Carbon Dioxide Dispensing and Monitoring System

This section describes the design of the open top chambers and of the equipment used in dispensing, monitoring, and sampling carbon dioxide at Tuskegee Institute during the 1984 study.

1. Carbon Dioxide Dispensing System

Liquid carbon dioxide supply: A 14-ton liquid receiver (Carbonic Industries) served as a CO_2 supply reservoir. The receiver maintained the temperature of the CO_2 between $-23^{\circ}C$ and $-19^{\circ}C$, which corresponded to pressures between 250 psig and 310 psig. In addition to the low-temperature, air-cooled condensing unit, the receiver was also equipped with a vaporizer. The vaporizer and cooler were automatically controlled with delivery capacities up to 6000 pounds per day. The contents and pressure of the receiver were displayed on gauges. The CO_2 was delivered from the chemical preparation plant to the receiver by tank truck. The CO_2 vapor was pumped out while the liquid was pumped in. Mechanical and electrical safety devices on the receiver helped to reduce pressure drops which could lead to solidification of the liquid CO_2.

High volume dispensing manifold: Delivery of the carbon dioxide gas from the receiver was accomplished through a 10m underground run of 1.27 cm (1/2") copper tubing (Type K) to the dispensing system, situated in the CO_2 research laboratory. The indoor portion of the tubing was wrapped with conventional unthermostated heat tape. This was done to avoid condensation on the manifold components which would otherwise be quite heavy during periods of high humidity. Between the dispensing manifold

system and the copper delivery tube was a 1.59 cm (5/8") closed solenoid valve. During power failures this valve prevented CO_2 flow.

The high-volume, dispensing manifold (Fig. 1) was custom made (Airco Industrial Gases, Research Triangle Park, NC). The CO_2 from the receiver was fed into six high-pressure, single-stage, line regulators (Airco Model 040-30500) at 100 psig. Three of these high-pressure regulators each fed three single-stage line regulators (Airco Model 048-50101); the other three high-pressure regulators each fed two single-stage line regulators (Airco Model 048-50101). These fifteen regulators fed (at 35 psig) 15 dual flow meters (Airco Model A75). Control valves on the flow meters served to control the flow rates. A 0.95 cm (3/8") Impolene tube (polyallomer, black; Gould Imperial Eastman) led from each of the flow meters to the inlet valves on each of the exposure chambers in the field.

Plant exposure system: Open top field chambers (Figs. 2,3) were used to expose plants to elevated concentrations of CO_2. The open top chambers which are essentially open-ended cylindrical baffles were developed at the Air Pollution Laboratory at North Carolina State University (Heagle et al., 1973, 1979; Davis and Rogers, 1980). Each chamber was 3 m in diameter and 2.4 m in height. They were made of a structural aluminum frame covered with 8 mil clear PVC plastic film (Roll-A-Glass [formerly known as Krene], Tenneco, Livingston Coating Corporation, Charlotte, NC). Each chamber consisted of three parts: (1) the bottom half, (2) the top half, and (3) a 45 degree fustrum that partially reduced the top opening (Fig. 2). The bottom half of each chamber cover was double-walled. The inside wall was perforated and served as a duct to distribute air uniformly into the chamber (Figs. 2,3). Air

was supplied to this duct by a 0.75 HP axial fan, mounted in a sheet metal plenum box with a particulate filter. Carbon dioxide from the dispensing manifold was injected into the plenum box ahead of the fan to assure thorough mixing. The CO_2 emptied about 1/4 way down the vertical center line of the plenum box to enhance mixing. A door in the bottom panel, on the side opposite the fan, permitted convenient access to the inside of the chamber.

2. Monitoring of Carbon Dioxide Concentrations

Gas sampling system: Monitoring of carbon dioxide was done 24 hours a day for the entire duration of the study (Figs. 4,5). The CO_2 concentration of all 15 sweet potato plots was sampled in a 32-min cycle with each of the 15 plots being sampled for 2 min. The remaining 2 minute sampling period was used for monitoring the cowpea chambers. Sampling lines were inserted inside 2-liter, inverted, polystyrene jugs mounted 1 meter above the ground at the center of each chamber. Sample air was drawn from the intake to the sampling manifold through Impolene tubing (0.635 cm, polyallomer, black; Gould Imperial Eastman).

In the sampling manifold, each sample passed through an adjustable flow meter (Dwyer VFA-BV-23) and then through a 3-way 24 VDC stainless steel solenoid (Versa Valves ESM-8302) with a 0.24 cm (3/32") orifice which was normally closed. A vacuum to the solenoid bank was provided by a Metal Bellows pump (MB-602) so that 5 liter/min were continuously pulled through each sample line. The actuation of the solenoids was done sequentially, each for 2 min, by a custom made timer (Rogers et al., 1980). While the CO_2 concentration of one sample was being measured the remaining samples were being exhausted. This minimized the line flushing that had to occur before an accurate sample could be read. The sample being measured

was diverted by a Metal Bellows pump (MB-41) through two infrared gas analyzers.

Carbon dioxide measurement: The monitoring of CO_2 concentrations was done by two Leybold Heraeus (Binos Model 43-200 1/2) non-dispersive, infrared gas analyzers (IRGAs). This type of instrument is based on the absorption of infrared energy by CO_2. The amount of infrared energy passing through the sample cell that is not absorbed is sensed by an analogue indicator. Nitrogen (a non-absorbing gas) served as the reference gas and was sealed in the reference cell. The sample stream passed through a filter cell before entering the sample cell. Outputs from the IRGAs were continuously recorded on two Hewlett Packard (7132A) dual-pen, strip-chart recorders.

Calibration procedures: The infrared gas analyzers were calibrated twice daily using a series of high-pressure tanks of known CO_2 concentrations, ranging from 0 to 1000 ppm of CO_2. These tanks were calibrated against a series of similar tanks that were calibrated by Livermore National Laboratory. The output of the Binos IRGAs was supposed to be linear with respect to CO_2 concentration. However checks against the standard tanks showed a departure from linearity that increased throughout the summer. Therefore, it was necessary to correct the CO_2 readings by fitting quadratic regressions to the calibration curves.

After calibration, the chamber scanner was disabled and manually set to an open field plot to determine the ambient CO_2 concentration. The target CO_2 concentrations were calculated, and the chamber concentrations were adjusted to the target values by manually adjusting the flow tube settings on the dispensing manifold. The new flow tube settings were calculated as follows:

$$\frac{\text{New setting}}{\text{Desired Concentration}} = \frac{\text{Current Setting}}{\text{Current Concentration}}$$

where:

New setting = New, calculated setting of flow tube.

Desired Concentration = Appropriate target CO_2 concentration for each chamber.

Current Setting = Present flow tube setting.

Current Concentration = Concentration of CO_2 in chamber (from IRGA reading).

After the flow rates were adjusted, the scanner was reset to sample each chamber and open field plot automatically.

The system for dispensing and monitoring of CO_2 into open top chambers performed satisfactory in the generation of large-scale test environments in the field. The maintenance and operation of the system presented no major problems.

B. Statistical Analysis of Carbon Dioxide Data

In order to determine the actual concentration of CO_2 inside the chambers, a sub-sample of the entire CO_2 data set was obtained and analyzed as follows. Carbon dioxide was dispensed to and monitored from 15 test points (3 in open field plots and 12 within open top chambers) from June 8 to August 21 (Julian days 160-234). Individual CO_2 readings were continuously recorded on strip chart recorder as described in the section on system development. Beginning on June 9 (Julian Day 161) chart readings were taken every 9 days. This yielded CO_2 values for nine 24-hour periods. The data for two of the nine selected days could not be used. In one case a thunderstorm and high winds caused large fluctuations in the CO_2 concentrations. In the other case the chamber doors were open during parts of the day for sampling purposes. In those two cases, the closest acceptable day was substituted for the unacceptable day. Using these data, hourly means and standard deviations of CO_2 concentrations were calculated for each treatment. These data were separated into daytime, nighttime, and 24-hour group means (Table 1). Daytime was between 0700 and 1700 central standard time (CST). Nighttime was between midnight and 0700 and between 1700 and 2400 CST. Means and standard deviations for CO_2 concentrations for individual chambers are shown in Tables 2-4.

Table 1. Seasonal CO_2 concentrations in sweet potato study plots. Values represent means (± S.E.) of 324 readings (162 daytime, 162 nighttime) taken every two hours from nine selected days between Julian day 160 and Julian day 234.

CO_2 Added (ppm)		Mean CO_2 Concentration (ppm)		
Nominal Treatment	Actual Daytime	Daytime+	Nighttime+	24 Hours*
Ambient	0	354 ± 15	440 ± 50	397 ± 57
+0	0	354 ± 15	437 ± 48	396 ± 54
+75	+77	431 ± 16	519 ± 46	475 ± 56
+150	+152	506 ± 18	597 ± 47	552 ± 58
+300	+305	659 ± 26	759 ± 54	709 ± 66

+N = 162

*N = 324

Table 2. 24 hour CO_2 concentrations in sweet potato study plots. Values represent means (± S.E.) of 108 readings for each chamber (324 for each treatment) taken every two hours from nine selected days between Julian day 160 and Julian day 234.

	Ambient	+0	+75	+150	+300
Target (ppm)	---	397	472	547	697
Treatment Means					
CO_2 (ppm)	397 ± 57	396 ± 54	475 ± 56	552 ± 58	709 ± 66
Target	---	-1	+3	+5	+12
Chamber Means					
Chamber number	2	5	3	4	1
CO_2 (ppm)	401 ± 61	398 ± 57	476 ± 58	553 ± 61	707 ± 64
Mean	+4	+2	+1	+1	-2
Chamber number	7	6	8	9	10
CO_2 (ppm)	396 ± 57	394 ± 54	475 ± 55	553 ± 58	709 ± 66
Mean	-1	-2	0	+1	0
Chamber number	11	14	12	13	15
CO_2 (ppm)	394 ± 53	396 ± 52	474 ± 55	549 ± 55	711 ± 68
Mean	-3	0	-1	-3	+2
Range of means between chambers	7	4	2	4	4

Table 3. Daytime CO_2 concentrations in sweet potato study plots. Values represent means (± S.E.) of 54 readings for each chamber (162 for each treatment) taken every two hours from nine selected days between Julian day 160 and Julian day 234.

	Ambient	+0	+75	+150	+300
Target (ppm)	---	354	429	504	654
Treatment Means					
CO_2 (ppm)	354 ± 15	354 ± 15	431 ± 16	506 ± 18	659 ± 26
Target	---	0	+2	+2	+5
Chamber Means					
Chamber number	2	5	3	4	1
CO_2 (ppm)	356 ± 16	355 ± 16	432 ± 15	506 ± 21	658 ± 27
Mean	+4	+2	+1	+1	-2
Chamber number	7	6	8	9	10
CO_2 (ppm)	354 ± 15	354 ± 16	432 ± 15	507 ± 17	659 ± 26
Mean	0	0	+1	+1	0
Chamber number	11	14	12	13	15
CO_2 (ppm)	352 ± 14	355 ± 14	430 ± 1	506 ± 17	660 ± 26
Mean	-2	+1	-1	0	+1
Range of means between chambers	4	1	2	1	2

Table 4. Nighttime CO_2 concentrations in sweet potato study plots. Values represent means (± S.E.) of 54 readings for each chamber (162 for each treatment) taken every two hours from nine selected days between Julian day 160 and Julian day 234.

	Ambient	+0	+75	+150	+300
Target (ppm)	---	440	515	590	740
Treatment Means					
CO_2 (ppm)	440 ± 50	437 ± 48	519 ± 46	597 ± 47	759 ± 54
Target	---	-3	+4	+7	+19
Chamber Means					
Chamber number	2	5	3	4	1
CO_2 (ppm)	446 ± 55	440 ± 51	521 ± 50	600 ± 51	757 ± 50
Mean	+6	+3	+2	+3	-2
Chamber number	7	6	8	9	10
CO_2 (ppm)	439 ± 51	435 ± 48	518 ± 47	599 ± 47	758 ± 55
Mean	-1	-2	-1	+2	-1
Chamber number	11	14	12	13	15
CO_2 (ppm)	435 ± 44	436 ± 45	517 ± 44	592 ± 45	762 ± 58
Mean	-5	-1	-2	-5	+3
Range of means between chambers	11	5	4	8	5

irregularly plowed was set aside for the cowpea study.

Planting and assembly of chambers: Sweet potato plants (Ipomoea batatas L. 'Georgia Jet') that were three months old and about 20-25 cm in length were obtained from the Macon County Sweet Potato Cooperative. The plants were transplanted into the field by hand, with the help of wooden stakes, on May 21, 1984. Plants were placed about 30 cm apart in rows that were about 90 cm apart. The rows were raised about 20 cm above the surrounding soil. The chambers were assembled and placed on the field during the period of May 23-27. On May 28 some plants were transplanted into the chambers from parts of the field not being used in the experiment. This was done to replace plants that had died and to assure uniformity in the number and size of plants in the chambers. There were two rows of 10 plants inside each chamber. From May 28 to June 7 necessary adjustments were made in the CO_2 dispensing and monitoring systems. Dispensing and monitoring of CO_2 in the chambers began on June 8. The amount of CO_2 required to maintain 9 sweet potato chambers and 2 cowpea chambers at elevated levels of CO_2 was about 1000 pounds per day.

Maintenance of the crop: Water was applied to the field by drip irrigation as necessary to supplement natural rainfall. There was little rainfall in the first part of the growing season, and irrigation was needed frequently. During the second part of the growing season there was an above-average amount of rainfall and irrigation was rarely needed. Fertilizer was applied at the rate of 14.7 kg of nitrogen, 14.7 kg phosphorus and 22.0 kg of potassium per ha, according to the recommendations of the soil tests. One-half of the fertilizer was applied in bands along the rows at the time of planting; the other half was sidedressed four weeks after planting. Weeding in and around the chambers

and open field plots was done manually. Parts of the field not involved in the experiment were weeded manually and by tractor hoeing. Problems with insects were minor; thus no insecticide was used. Nematode counts were taken at the middle and at the end of the growing period. Nematode counts were insignificant.

Weather monitoring: A Taylor Weatherscope Thermivolt Thermometer with thermistor, barometric pressure indicator, anemometer and wind direction indicator was used to measure the ambient temperature, barometric pressure, wind velocity and wind direction. Measurements were taken twice each day, at the time of calibration of the infrared gas analyzers. The thermistor was shielded from direct sunlight. Two rainfall gauges were placed in the field and checked daily.

2. Harvest of Sweet Potato Plants

Shoots: The vegetative portion of sweet potatoes from 15 plots were harvested during August 21-24. Five sweet potato plants from both the left and right row of each plot were randomly harvested to give ten plants for each plot. The first and tenth plants of each row were never harvested due to their close proximity to the edge of the chamber. The five plants from the left row were harvested as a group first, then the five plants from the right row were harvested as a group.

In each group of five plants, the stems were cut 1 cm above the ground. Green and dead leaves were separated from the runners; petioles remained with the runners. Fresh weights green leaves, and dead leaves and runners were determined. Stem diameters were measured by a caliper at the basal end of the main stem of each plant. Runner lengths were measured, and a total runner count was taken.

In order to measure leaf area and count leaf number within a

reasonable time period, one half of the total green leaf fresh weight was utilized as a subsample to measure leaf area and leaf number. The green leaves were run through a LI-COR Area Meter (LI-3100) and were counted as they passed out of the meter. Totals for leaf area and number of leaves were calculated by multiplying the measured leaf area and number of leaves by 2.

The measurements for the left and right rows of each plot were either averaged or totaled, as appropriate, to give values for the entire 10 plants that were harvested from each plot.

At the end of each harvest day (August 21-24), individual leaf and runner sample bags were placed in ovens at Auburn University's E.V. Smith Research Farm at a temperature of 55 $\pm$ 5°C. The leaf and runner samples were checked every seven days and turned over in order to obtain uniform drying throughout each bag. All leaf and runner samples were obtained from the dryers on September 9. Dry weights were recorded for dead leaves, green leaves, and runners separately.

Roots: The sweet potato tubers were harvested during August 27-29. Tubers were dug for the same ten plants in each plot that were used in the vegetative harvest. The tubers were placed on a board, lightly rinsed with water in order to remove clumps of soil adhering to the potatoes, then air-dried outdoors for 10 to 15 minutes. The fresh weight, diameter, length, and volume were taken for each individual tuber. Any tubers which were odd-shaped were noted. Diameter was taken with calipers at the greatest dimension measured at right angles to the longitudinal axis. The length was taken at the greatest dimension measured in a line between points at each end. Volumes were measured by the displacement of known amounts of water in several different size graduated cylinders and beakers.

The total fresh weight and number of potatoes, as well as the mean length, diameter and volume of the potatoes, were calculated for each plot. Individual tuber fresh weights and volumes were used to calculate the density of each tuber.

On August 30, two potato samples of each grade from each plot were selected and fresh weights were taken. Jumbo grade potatoes were present in only 4 plots and were used when available. Each tuber was cut into slices less than 0.5 cm in width. These samples were placed in paper bags and dried in ovens at a temperature of $55 \pm 5^{o}C$. The remainder of the tubers were cured and some of them were used in studies on nutritive composition and acceptability that are described elsewhere in this report. The sliced tuber samples were taken out of the ovens on September 9, and dry weights were recorded. Sample fresh weights and dry weights from the sliced samples were totaled for each plot. Percent dry matter was calculated as follows:

$$\text{Percent dry matter} = \frac{\text{sample dry weight}}{\text{sample fresh weight}} \times 100$$

3. Potato Grades

The individual tuber measurements for fresh weight, diameter, length, and volume were entered in the computer. A code for odd shaped tubers was entered also. The size requirements for each grade generally followed those of the North Carolina Department of Agriculture and Rogers et al. (1983a). Based on these size requirements, the individual tubers in each plot were placed in a specific grade using SAS (Statistical Analysis System).

The criteria used to place individual tubers in a grade are as follows:

Grade	Length (cm)	Diameter (cm)	Weight (gm)
1	4.0 <= length <= 23	4.0 <= diameter <= 9.0	< 1022
2		diameter > 3.8	< 1022
Canner		2.5 <= diameter <= 3.8	< 1022
Cull		diameter < 2.5	< 1022
Jumbo			>= 1022

Grade 2 potatoes are similar in size to #1's but fall into another grade due to their being odd shaped.

4. Statistical Analysis of Sweet Potato Data

Statistical analysis of sweet potato growth data was performed using standard analysis of variance techniques (Snedecor and Cochran, 1967) and by utilizing SAS (Statistical Analysis System, SAS Institute, Cary, NC). In the analyses, a complete randomized block design was used with five treatments (four open top chambers at 354, 431, 506 and 659 ppm CO_2 and the open field plot) in each block. There were three replicate blocks. An analysis of variance was run on all data with the model shown below:

Source	Degrees of Freedom
Block	2
CO_2	4
Error	8
Total	14

Tests for significance were done with the Block * CO_2 error mean square from the ANOVA. This error term had 8 degrees of freedom. The coefficient of variation (CV) was calculated by the ANOVA procedure. The $S_{\bar{x}}$ (standard error of the mean) was calculated as follows:

$$S_{\bar{x}} = \sqrt{\frac{\text{Error Mean Square}}{N}}$$

where N = 3. The LSD (least significant difference) is a test for all main-effect means.

B. Growth, Yield and Nitrogen Content of Sweet Potatoes

Sweet potato plants grown in the field under elevated levels of CO_2 in open top chambers showed large increases in root biomass and small increases in shoot biomass. In addition, there were large decreases in shoot biomass associated with the presence of the open top chambers.

1. Effects of Open Top Chambers

Results: A comparison of the plants grown in the open top chambers at ambient CO_2 with the plants grown in the open field plots indicates that there were significant effects on the plants due of the presence of the chambers. Plants grown in open top chambers had lower total fresh weights than plants grown in open plots (Table 5). Roots and shoots showed similar decreases in fresh weight, and the root/shoot ratio did not change significantly (Table 5). The presence of the chamber also affected numerous parameters associated with shoot size. Plants grown in ambient chambers had lower total runner fresh and dry weights (Tables 6,7), lower total leaf dry weight (Table 7), and lower total runner length (Table 8) than plants grown in open field plots.

Discussion: These results suggest that plants grown in open top chambers at ambient CO_2 had lower rates of photosynthesis and allocated a greater percentage of their photosynthate to below-ground parts than did plants grown in the open field. Sweet potatoes generally grow well under conditions of high temperature, high light intensity and considerable rainfall, provided there is good drainage during the growing period. However, moderately dry weather is favorable for the formation and development of tuberous roots. Thus, decreases in light and soil moisture might be expected to have effects on the growth of sweet potato plants.

The plants grown in the open top chambers were exposed to a constant wind from the fan, to slightly reduced light levels, and probably to higher temperatures (Rogers et al., 1983a). These conditions most likely contributed to a more rapid evaporation of soil moisture inside the chambers. The lower soil moisture and greater wind contributed to reduced stomatal conductances in the plants grown in the chambers (Table 23). The partial closure of stomates in the plants of ambient CO_2 chambers probably decreased the CO_2 uptake and thus photosynthesis. The partial closing of stomates under water stress conditions has been reported by a number of investigators (Wong et al., 1978; Sharkey and Raschke, 1981; Sionit et al., 1981, 1984).

The decreased shoot biomass in plants grown in chambers may have led to less self shading of leaves. This greater exposure of leaves to sunlight could lead to an increase the overall efficiency of light utilization of plants grown in chambers. This potential increase in light utilization efficiency may partly offset decrease in carbon gain caused by stomatal closure. Leaf area has an important effect on net assimilation rate and dry matter production in sweet potatoes (Hahn 1977). In sweet potatoes (Ipomoea batatas L., 'Koganesengan') the crop growth rate has been shown to correlate with the net assimilation rate and the leaf area index (Agata, 1980). Therefore, the effect of the chamber on shoot biomass needs to be considered when analyzing the effects of CO_2 on sweet potatoes.

2. Effects of Carbon Dioxide

Results: Elevated levels of CO_2 were associated with large increases in the roots of sweet potatoes grown in open top chambers. However, shoots were not greatly affected. Sweet potato plants grown at

506 ppm CO_2 had greater total fresh weights than did plants grown in ambient chambers (Table 5, Fig. 9). Most of this difference was due to differences in root fresh weights. Plants grown at 506 and 659 ppm CO_2 had greater total tuber fresh weights than did plants grown in ambient chambers (Table 5). (The differences in tuber fresh weight were significant according to the LSD test, even though the CO_2 effect in the ANOVA was not significant; P=0.057.) Root/shoot ratios were significantly greater at 659 ppm CO_2 than at 354 ppm CO_2 (Table 5, Fig.10). The number of tubers increased significantly with increasing CO_2 (Table 10, Fig. 12), but the sizes of the individual tubers were not affected (Table 13). Total leaf dry weight was greater in plants grown at 431 ppm CO_2 than in plants grown in chambers at ambient (354 ppm) CO_2 (Table 7, Fig. 11). However, no other morphological shoot parameter was significantly affected by CO_2 and most shoot parameters did not show any appreciable non-significant changes. Leaf nitrogen content was significantly at lower at 659 ppm CO_2 than at 354 ppm CO_2 (Table 19, Fig. 13).

Discussion: Plants response to CO_2 enrichment in the field depends on the type of crop, the meteorological conditions, the distribution of CO_2 flux into the vegetation (Anderson, 1975; Wittwer, 1978a; Allen, 1979) and the stage of development (Krenzer and Moss, 1975). The results presented in this investigation confirm earlier studies in Ipomoea batatas 'Travis' (Rogers et al., 1983a) and Ipomoea batatas 'Georgia Jet' (Bhattacharya et al., 1985) at North Carolina State University, Raleigh and Duke University Phytotron, Durham, respectively. The lack of a significant CO_2 effect on shoot growth of sweet potatoes differs from the results reported for most non-tuberous plants grown under

increased CO_2 (Rogers et al.,1980, 1982, 1983a; Sionit et al., 1981, 1982). Bhattacharya et al. (1985) found that differences in the growth of main stems and branches due to elevated CO_2 were apparent in the early stages (28 to 35 days) of growth of sweet potatoes. However, in the later stages (60 to 65 days) of growth, these differences were greatly reduced due to the fact that shoot growth at elevated CO_2 levels ceased earlier that at ambient CO_2.

The increased dry matter production at high CO_2 level has been reported in a number of plants (Mauney et al., 1978; Krizek, 1970; Krizek et al., 1968, 1970, 1974; Allen, 1979; Kramer, 1981) and it was associated with increased rate of photosynthesis at high CO_2 concentration. The increasing rate of photosynthesis with increasing concentration of CO_2 has been well documented in the literature (Akita and Tanaka, 1943; Cooper and Brun, 1967; Ford and Thorne, 1967; Huber et al., 1984). Sionit et al. (1982) reported increases in total dry matter production in soybean (Glycine max L. Merr), radish (Raphanus sativus L., 'White Tip'), sugar beet (Beta vulgaris L., 'F58-554HI, MS of NBIX x NB4), and corn (Zea mays L., 'Silvercross') at all growth stages with increased CO_2 concentration light intensity. Sionit et al. (1984) also studied the interaction of enriched CO_2 and light in soybeans grown in the field and in containers placed in the field. The field grown plants responded to a lesser extent to increasing CO_2 concentration than container grown plants. The photosynthesis-irradiance relationships were linear in the field grown plants and curvilinear in the pot grown plants. At the highest irradiance level tested (1400 umol $m^{-2}s^{-1}$) the field grown plants had greater net photosynthetic rates than the container grown plants.

The significant increase in the number of sweet potato tubers (Table 10) at elevated CO_2 levels suggests greater partitioning of photoassimilates for the growth tubers than the vegetative organs. The increases in root biomass and root shoot-ratio reported here have also been observed in other root crops exposed to elevated levels of CO_2. Sionit et al. (1982) showed increased root/shoot ratio in sugar beets and radishes with increase in atmospheric CO_2 concentration. Knecht (1975) measured the response of radishes (Raphanus sativus L., 'Cherry Belle') to high CO_2 and found that growth of both shoots and roots, as well as the root-shoot ratio, increased under elevated CO_2 conditions in plastic-enclosed, environmentally-controlled environments. Imai and Coleman (1983) studied the effect of an elevated CO_2 atmosphere on dry matter production of konjak (Amorphophallus konjak K. Koch). They demonstrated that at 670 ppm CO_2 the yield of the corm increased two-fold because the net assimilation rate doubled and, due to the simple source-sink relationship, the increased production was partitioned to the corm. The greater partitioning of photoassimilates to roots during the growth and development of tubers in sweet potatoes has also been discussed by Bhattacharya et al. (1985). They have concluded that CO_2 enrichment modified the source-sink relationship so as to enhance the production of tubers in sweet potatoes.

Collins (1976) studied the effect carbon dioxide enrichment on the growth of white potato plants (Solanum tuberosum L., 'Kennebec') in the greenhouse and reported an increased yield of potatoes in response to high CO_2 concentrations. This enhancement in yield was attributed to an increase in the net assimilation rate of carbon dioxide enriched plants and a corresponding increase in the relative growth rate of plants. In

contrast, Goudriaan and deRuiter (1983) investigated the effect of CO_2 enrichment in white potatoes (Solanum tuberosum L., 'Alpha') and reported slightly negative responses of CO_2 enrichment on the dry matter production of leaves, stems, roots and tubers. The decreases in the potato plants in response to high CO_2 concentrations were attributed to increase in the size of starch grains in the leaves resulting in damage to the chloroplasts. Bhattacharya et al. (1985) however reported an increase in starch concentration in the leaves of sweet potatoes (Ipomoea batatas 'Georgia Jet') at elevated CO_2 levels but did not observe any damage to the chloroplasts.

Fujise and Tsuno (1967) showed that high K_2O/N ratios led to increases in water content, respiration rate and tuber growth, leading to accelerated translocation of photosynthates from leaves and to higher photosynthetic rates. These authors proposed that high potassium supply and K_2O/N ratios in tubers might also be associated with increased protein synthesis leading to enhanced tuber growth. This postulation find supports from our present investigation that percentage of nitrogen and protein nitrogen of the leaves (Table 19) decreased significantly with increased concentration of carbon dioxide and it was most pronounced at 659 ppm CO_2 grown plants. The decrease in total nitrogen and protein nitrogen in leaves may be associated with increased dry matter accumulation in leaves (Table 17(?)). With increasing CO_2, it is possible that less energy for nitrate reduction was available in the leaf and more nitrate reduction occurred in the root, due to an increased supply of sucrose. According to Huber et al. (1984) all of the extra carbon input due to enhancement of photosynthesis by CO_2 enrichment was partitioned into starch. Perhaps lack of responsiveness of whole plant nitrogen

accumulation to atmospheric CO_2 enrichment is related to the initial partitioning of fixed carbon into starch and subsequent slow remobilization of that carbon for transport to other plant parts including nodules in soybean (Glycine max, 'Bragg').

Table 5. Effect of different CO_2 concentrations on fresh weight and root-shoot ratio (R/S) of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Fresh Weight (gm)			
	Total	Tubers	Shoot	R/S
354+	24184[a]	13398	10786[a]	1.25[c]
354	15745[b]	9447	6298[b]	1.58[bc]
431	20311[ab]	12638	7673[b]	1.67[bc]
506	21410[a]	13961	7449[b]	1.88[ab]
659	19875[ab]	13829	6047[b]	2.30[a]
$S_{\bar{x}}$*	1555	981	838	0.15
CV (%)*	13.3	13	19	14.6
LSD	5070	3201	2733	0.48
	S	N.S.	S	S

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

Table 6. Effect of different CO_2 concentrations on the total fresh weight of leaves and runners of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Total Fresh Weight (gm)			
	Green Leaves	Dead Leaves	Total Leaves	Runners
354[+]	2628	184	2812	7974[a]
354	2189	101	2290	4008[b]
431	2388	176	2564	5109[b]
506	2329	134	2463	4986[b]
659	1970	98	2068	3979[b]
$S_{\bar{x}}$[*]	229	25	240	620
CV (%)[*]	17.2	30.8	17.1	21
LSD	745	80	78	2023
	N.S.	N.S.	N.S.	S

[+]The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

[*]$S_{\bar{x}}$ and CV (%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

Table 7. Effect of different CO_2 concentrations on the total dry weight of leaves, runners and shoots of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Total Dry Weight (gm)				
	Green Leaves	Dead Leaves	Total Leaves	Runners	Shoots
354+	386	108	494[a]	815[a]	1309[a]
354	305	60	365[c]	452[b]	817[b]
431	340	106	446[ab]	569[b]	1015[b]
506	337	88	425[abc]	584[b]	1009[b]
659	324	57	381[bc]	529[b]	910[b]
$S_{\bar{x}}$*	18	14	23	55	76
CV (%)*	9	30	10	16	13
LSD	60	47	75	181	249
	N.S.	N.S.	S	S	S

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

Table 8. Effect of different CO_2 concentrations on the mean stem diameter, and on the total number and length of runners of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Mean Stem Diameter (cm)	Total Number of Runners	Total Runner Length (cm)
354+	1.14	196	11506[a]
354	1.00	152	6433[b]
431	1.09	189	8652[b]
506	1.11	174	7702[b]
659	1.02	155	7017[b]
$S_{\bar{x}}$*	0.05	12.9	794
CV (%)*	7.5	12.9	16.6
LSD	0.15	42	2588
	N.S.	N.S.	S

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

Table 9. Effect of different CO_2 concentration on the total leaf area and the total number of leaves of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Total Leaf Area (cm^2)	Total Number of leaves
354+	135071	2090
354	107489	1871
431	115586	1886
506	110105	1927
659	88810	1789
$S_{\bar{x}}$*	11792	174
CV (%)*	18	16
LSD	38457	567
	N.S.	N.S.

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 10. Effect of different CO_2 concentrations on the total number of tubers and the number of tubers for each grade of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Total number of tubers	U.S. #1	U.S. #2	Canner	Cull	Jumbo
354+	70[c]	21	24	14	10	2
354	67[c]	16	27	15	9	0
431	73[bc]	22	29	11	11	0
506	93[ab]	25	30	19	19	1
659	97[a]	28	37	19	12	1
$S_{\bar{x}}$*	6.6	6.6	5.6	2.5	3.5	--
CV (%)*	14	50	32	31	49	--
LSD	22	21	18	19	11	--
	S	N.S.	N.S.	N.S.	N.S.	--

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

Table 11. Effect of different CO_2 concentrations on the total fresh weight of different grades of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Total Fresh Weight (gm)				
	#1	#2	Canner	Cull	Jumbo
354+	6025	5782	593	111	1330
354	3247	5348	738	114	0
431	5828	6312	373	125	0
506	6009	6124	1052	301	1425
659	5701	6831	765	165	1098
$S_{\bar{x}}$*	1274	1189	239	65	---
CV (%)*	41	34	59	69	---
LSD	4155	3879	781	211	---
	N.S.	N.S.	N.S.	N.S.	

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 12. Effect of different CO_2 concentrations on the percent by fresh weight of different grades of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Percent by Fresh Weight				
	#1	#2	Canner	Cull	Jumbo
354+	45	43	4	1	7
354	34	57	8	1	0
431	46	50	3	1	0
506	43	44	8	2	3
659	41	49	6	1	3
$S_{\bar{x}}$*	3.2	3.0	6.6	0.5	---
CV (%)*	40	32	60	68	---
LSD	11	10	2	0.5	---
	N.S.	N.S.	N.S.	N.S.	

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 13. Effect of different CO_2 concentrations on diameter, length, and volume of tubers of sweet potatoes plants grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Diameter (cm)	Length (cm)	Volume (cm^3)
354+	4.99	9.88	192.0
354	4.82	9.43	144.7
431	4.99	10.01	170.7
506	4.61	9.63	151.4
659	4.74	9.86	147.0
$S_{\bar{x}}$*	0.29	0.37	14.7
CV (%)*	10.5	6.6	15.8
LSD	0.95	1.22	47.9
	N.S.	N.S.	N.S.

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 14. Effect of different CO_2 concentrations on the mean diameter of different grades of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Mean Diameter (cm)				
	#1	#2	Canner	Cull	Jumbo
354[+]	6.3	6.1	3.3[a]	1.8	11.1
354	6.0	5.9	3.3[ab]	2.0	---
431	6.2	5.9	3.1[bc]	1.8	---
506	6.3	5.6	3.2[c]	1.8	10.3
659	5.6	5.7	3.1[c]	2.0	13.0
$S_{\bar{x}}$[*]	0.3	0.3	< 0.1	0.1	---
CV (%)[*]	7.8	8.1	2.2	9.4	---
LSD	0.9	0.9	0.1	0.3	---
	N.S.	N.S.	S	N.S.	

[+]The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

[*]$S_{\bar{x}}$ and CV (%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

Table 15. Effect of different CO_2 concentrations on the mean length of different grades of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Mean Length (cm)				
	#1	#2	Canner	Cull	Jumbo
354[+]	12.9	10.2	8.3	5.3	17.5
354	11.6	10.0	8.6	5.4	---
431	13.2	10.8	7.5	5.3	---
506	13.3	10.0	8.2	6.1	20.0
659	12.4	10.4	8.1	5.4	14.0
$S_{\bar{x}}$[*]	0.9	0.6	0.8	0.6	---
CV (%)[*]	11.8	9.2	15.9	19.1	---
LSD	2.8	1.8	2.4	2.0	---
	N.S.	N.S.	N.S.	N.S.	

[+]The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

[*]$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 16. Effect of different CO_2 concentrations on the mean volume of different grades of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Mean Volume (cm^3)				
	#1	#2	Canner	Cull	Jumbo
354+	304	235	48	13	1375
354	231	197	46	16	---
431	281	207	36	12	---
506	289	174	58	17	1500
659	204	195	47	16	1020
$S_{\bar{x}}$*	34	19	9	2	---
CV (%)*	22	17	32	24	---
LSD	110	63	28	7	---
	N.S.	N.S.	N.S.	N.S.	

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 17. Effect of different CO_2 concentrations on the density and dry matter content of all grades of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Density (g/cm^3)	Percent Dry Matter
354+	1.00	17.6
354	1.03	19.3
431	1.00	18.9
506	0.98	19.0
659	0.99	19.6
$S_{\bar{x}}$*	0.03	0.4
CV (%)*	4.9	3.7
LSD	0.09	1.3
	N.S.	N.S.

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 18. Effect of different CO_2 concentrations on the density of the different grades of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Density (gm/cm^3)				
	#1	#2	Canner	Cull	Jumbo
354+	1.01	1.03	0.99	0.91	0.97
354	1.05	1.03	1.12	0.88	---
431	1.03	1.00	1.01	0.90	---
506	1.00	1.01	0.93	0.94	0.95
659	0.99	1.00	1.02	0.85	1.08
$S_{\bar{x}}$*	0.02	0.03	0.07	0.07	---
CV (%)*	3.9	5.5	11.9	14.3	---
LSD	0.08	0.11	0.23	0.24	---
	N.S.	N.S.	N.S.	N.S.	

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

Table 19. Effect of different CO_2 concentrations on the percentage of total nitrogen, protein nitrogen, and non-protein nitrogen of leaves of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Total Nitrogen (%)	Protein N (%)	Non-Protein N (%)
354+	3.88[a]	24.2[a]	75.8[a]
354	3.77[ab]	23.6[ab]	76.4[ab]
431	3.67[ab]	23.0[ab]	77.0[ab]
506	3.44[b]	21.5[b]	78.5[b]
659	2.86[c]	17.9[c]	82.1[c]
$S_{\bar{x}}$*	0.11	0.7	0.7
CV (%)*	5.4	5.4	1.5
LSD	0.36	2.3	2.3
	S	S	S

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

Table 20. Effect of different CO_2 concentrations on the nitrogen and on the percent of nitrogen of stems of sweet potatoes grown in open plots and within chambers at day 92 harvest, n = 3.

CO_2 (ppm)	Total N (%)	Protein N (%)	Non-Protein N (%)
354+	0.90	5.7	94.4
354	0.95	5.9	94.1
431	0.89	5.6	94.4
506	0.85	5.3	94.7
659	0.76	4.7	95.3
$S_{\bar{x}}$*	0.05	0.3	0.3
CV (%)*	9.7	9.7	0.6
LSD	0.16	1.0	1.0
	N.S.	N.S.	N.S.

+The first CO_2 value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*$S_{\bar{x}}$ and CV (%) are from ANOVA.

C. Nutritive Composition and Acceptability of Sweet Potato Tubers

Experiments have been conducted to determine the effects of four levels of CO_2 on the nutrient content of 'Georgia Jet' sweet potatoes. The nutrients studied included: protein, carbohydrates, oil, ash, vitamins A and C; dry matter content and dietary fiber were also measured. A taste panel was also conducted to ascertain differences in appearance and taste qualities of the sweet potatoes treated with the four levels of CO_2.

Dry matter (DM) content decreased significantly ($P<0.05$) with the higher CO_2 levels. This finding agreed with those of other researchers. The findings on DM was the complete opposite (?) of those findings for the protein content of the sweet potatoes treated with higher levels of CO_2, i.e. a significant decrease ($P<0.05$) in protein levels were measured for the sweet potatoes receiving the highest CO_2 (506 and 659 ppm) exposure. This finding also agrees with other findings in the literature.

The carbohydrate content data was not consistent, but elevated levels of CO_2 were associated with an increase both in glucose and maltose. The same treatment was associated with a significant decrease (?) in carotenoid pigments. The same decrease was not measured for the ascorbic acid, oil and ash contents.

Total dietary fiber levels were not significantly different, but there was a significant decrease in the insoluble dietary fiber components. Soluble dietary fiber was also unaffected. Total dietary fiber ranged from 3 to 3.5%, insoluble dietary fiber ranged from 1 to 2% and soluble dietary fiber was about 1%.

Color changes were noted among those sweet potatoes treated with CO_2. Hue angle measurements indicated a deeper orange color (?). This finding agreed with those obtained from the taste panel, i.e. treated sweet potatoes had higher rating (thus indicating a preference) scores than the controls (Table 21). All potatoes rated for appearance or taste received ratings not less than 5.80 on a hedonic rating scale ranging from 1 = dislike extremely to 9 = like extremely.

The present research does indicate that an increase in atmospheric CO_2 is associated with a decrease in carotenoid, protein and insoluble dietary fiber. These findings suggest that these negative effects can possibly affect the nutrient intake of these essential nutrients. Unlike the effect on nutrient content (?), the acceptability of the treated products would likely remain the same.

Table 21. Effect of different CO_2 concentrations on the appearance and evaluation of tubers of sweet potatoes grown in open plots and within chambers at day 92 harvest, n=3.

	Appearance		Taste			
CO_2 (ppm)	Eye Appeal	Color Intensity	Texture	Smoothness	Flavor	Moistness
354+	5.87[b]	5.77[bc]	6.58[ab]	6.75[a]	6.20	6.55[ab]
354	6.40[ab]	6.39[ab]	6.00[b]	6.03[b]	5.98	6.18[b]
431	7.02[a]	6.93[a]	6.85[a]	6.84[a]	6.58	6.54[ab]
506	5.87[b]	5.68[c]	6.42[ab]	6.49[ab]	6.38	6.63[ab]
659	6.59[a]	6.51[a]	6.85[a]	6.88[a]	6.64	6.79[a]
S_x*	0.70	0.80	0.60	0.60	0.70	0.70
CV(%)*	19	21	16	16	20	18
LSD	0.68	0.70	0.60	0.60	0.70	0.60
	S	S	S	S	N.S.	S

+The first value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

*S_x and CV(%) are from ANOVA.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

D. Stomatal Conductance of Sweet Potato Leaves

1. Methods

Measurements of stomatal conductance on leaves of sweet potatoes were made on July 11-13, 51 to 53 days after planting. The last major rainfall prior to the measurements occurred on July 1 (1.3 cm). However, the field was irrigated on July 8 and 9, and 0.2 cm of rain fell on July 10. On July 12, the measurements were interrupted by a rainstorm of 2.2 cm. Thus on July 11 and 12 the soil was moderately dry, and on July 13 the soil was quite wet. No mid-day wilting of leaves was observed during the measurements.

On each day, measurements were restricted to one block, consisting of one replicate of each of the five treatments (ambient, 354, 431, 506 and 659 ppm). The blocks were sampled on successive days in numerical order. Because both environmental conditions and stomatal conductance values change throughout the day, the five treatments were sampled in a repeating cycle. Each cycle took approximately one hour to complete. Measurements began as soon as the dew evaporated from the leaves and continued for eight cycles (approximately 0730 to 1530 hours CST). On July 12, only five cycles were completed before the rainstorm. Within a cycle, the treatments were sampled in numerical order, since the order of the treatments in each block had been determined randomly.

The selection of plants and leaves for measurement was designed to maximize the sampling of the variation among the plants and to minimize the variation among the leaves within the plants. For each treatment in each cycle, one leaf was measured on each of four plants. (Three plants were used for the first four cycles of July 11.) The plants for cycles 1

through 4 were chosen randomly, except that plants adjacent to the chamber wall were omitted. Measurements for cycles 5 through 8 were conducted on the same plants that were used on cycles 1 through 4. This procedure minimized the disturbance to the plants by sampling each plant only twice during the day.

Morphological observations indicated that the first three (counting back from the shoot tip) unfolded leaves on a growing runner were still expanding. Thus, whenever possible, measurements were made on the fifth unfolded leaf on a growing runner. Only leaves that were relatively unshaded by other leaves were selected, but no attempt was made to select leaves with any particular orientation with respect to the sun. Leaves were measured in their natural orientation. If no unshaded fifth leaf could be found on a plant, the adjacent leaves were examined (in the order 4, 6, 7, 8) until an acceptable leaf was located. Most measurements were conducted on the 4th, 5th and 6th leaves.

Measurements of stomatal conductance were made with a LI-COR LI-700 transient porometer. Because the porometer measures only one leaf surface at a time, and because sweet potatoes are amphistomatous, separate conductance measurements were made for the two leaf surfaces. The abaxial surface was measured first, followed by the adaxial surface, using a different part of the leaf. Simultaneous measurements of photosynthetically active radiation were made using a LI-COR quantum sensor attached to the porometer cup. Notes were also made on the amount of cloud cover in the sky and on whether or not the sun was obscured by clouds during the measurement. Leaf temperature measurements were made using a fine-wire thermocouple built into the porometer cup. During measurements

inside open top chambers, the chamber door was closed so as to maintain the desired carbon dioxide concentration.

The instruction manual for the LI-COR LI-700 porometer indicated that three to five drying cycles would be required to achieve a stable reading. Preliminary tests with sweet potatoes indicated that a stable reading was achieved with some leaves in two to three cycles. With other leaves, the reading continued to drift for at least five cycles and showed no sign of converging on a stable reading. Apparently these leaves were responding to the presence of the porometer cup. The porometer cup blocked the sunlight from reaching the portion of the leaf being measured. In addition the shading by the porometer cup most likely caused a lowering of the leaf temperature. Either the stomates on these leaves were closing rapidly, or the rate of evaporation of water off of the mesophyll cell surfaces was decreasing in response to the decreasing temperature. To minimize this problem and to standardize the measurements, all measurements were made using the reading from the second complete drying cycle. Preliminary tests indicated that when the readings did stabilize, the difference between the second and stabilized reading was about 5%.

Stomatal density was determined on separate leaf samples harvested on July 24. Counts were made on the fifth unfolded leaf (counting back from the shoot tip) from one runner from each of four randomly selected plants in each chamber. Five counts were made on each surface of each leaf.

Analyses of variances were performed on the total (adaxial plus abaxial) stomatal conductance readings and on the stomatal densities. Analyses of stomatal conductance were performed on each day's readings separately, and on the combined data from all three days.

2. Results and Discussion

There was no difference in the density of stomates among any of the treatments, including the ambient plots (Table 22). In all treatments, the density of stomates on the abaxial surface was slightly more than twice the density of stomates on the adaxial surface.

On July 11 and 12 (the two drier days) stomatal conductances were highest early in the morning (Figs. 14,15). The fact that conductances began at lower values on July 12 than on July 11 was probably due to the fact that one more day had elapsed since irrigation. On July 11, a mostly sunny day, stomatal conductances decreased rapidly during the first few hours, then levelled off. July 12 began as a sunny day, but turned mostly cloudy around 1200 hours CST. Rainfall began about 1400 hours CST. Stomatal conductances decreased during the sunny morning hours, increased somewhat when the cloud cover increased, and then showed some signs of decreasing again just before the rainfall started. On July 13 was partly cloudy. The soil was quite wet from the rainfall of the previous afternoon. Stomatal conductances were higher and more variable than on the two previous days (Fig. 16).

Analyses of variance performed on each day separately indicated a significant effect of treatment on stomatal conductance for all three days when the ambient plot (no chamber) was included in the analyses (Table 23). Stomatal conductance values for the ambient plot (no chamber) were about 35% higher than the values for the 354 ppm chamber indicating that the chamber caused a large decrease in stomatal conductance. Conditions inside the chamber were generally warmer, windier and drier than in the ambient plots. All of these factors probably combined to lead to the development of leaves with lower stomatal conductances.

When the ambient plot was omitted from the analyses, there was a significant effect of CO_2 on July 12 and 13, but not on July 11. However, there was no consistent trend in the effects of CO_2 on stomatal conductance. On July 12, conductance was higher at 431 ppm and lower at 659 ppm than at 354 ppm. On July 13, conductance was higher at 506 ppm than at 354 ppm. When all three days data were combined, there was no effect of CO_2 on stomatal conductance. The significant CO_2 effects on July 12 and 13 might be related to the fact that the samples in the different chambers were not true replicates. Specifically, the light conditions varied with changes in cloud cover and with the orientations of the selected leaves. These changing conditions might have been responsible for the significant differences in stomatal conductance observed on July 12 and 13.

Most previous studies have reported decreases in stomatal conductance with increasing CO_2 (Bingham et al., 1981; Sionit et al., 1981, 1984; Patterson and Flint 1982; Rogers et al., 1982, 1983a, 1983b, 1984; Huber et al., 1984). The lack of a consistent effect of CO_2 on stomatal conductance in this study may be partly related to water stress. In soybeans and in wheat, the effect of CO_2 on stomatal conductance was reduced as water stress increased (Sionit et al., 1981; Rogers et al., 1983a, 1984). In our study with sweet potatoes, soil moisture and the amount of water applied were not measured. Thus, possible variations in soil moisture among the chambers and the fact that the field was not well watered on July 11 or 12 may have combined to obscure any CO_2 effect on stomatal conductance. Alternatively, stomates of sweet potatoes may not respond as strongly to CO_2 as the stomates of other plants. More measurements of stomatal conductance need to be made under

more tightly controlled conditions of soil moisture. In addition, simultaneous measurements of photosynthesis and transpiration need to be made in order to clarify the effects of stomtal behavior on sweet potatoes grown under elevated carbon dioxide.

Table 22. Stomatal densities for sweet potatoes grown under elevated levels of carbon dioxide in open top chambers. Values are means ± standard errors, n = 12.

CO_2 (ppm)	Stomatal Density (number mm^{-2})		
	Adaxial	Abaxial	Total
354[+]	73 ± 5	174 ± 10	246 ± 12
354	82 ± 8	178 ± 10	260 ± 18
431	85 ± 8	176 ± 11	261 ± 18
506	69 ± 4	175 ± 9	245 ± 12
659	81 ± 5	197 ± 11	278 ± 15
P[*]	0.41	0.50	0.52
LSD[#]	19	29	42

[+]The first value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

[*]P values are the significance levels for the CO_2 effect from ANOVA.

[#]LSD = least significant difference.

Table 23. Mean stomatal conductances (adaxial + abaxial) for three days for sweet potatoes grown under elevated levels of carbon dioxide in open top chambers.

CO_2 (ppm)	Stomatal Conductance (sec cm^{-1})			
	July 11	July 12	July 13	Mean
354[+]	1.031[a]	1.163[a]	1.466[a]	1.220[a]
354	0.834[b]	0.906[b]	0.940[c]	0.893[b]
431	0.809[b]	1.060[a]	0.942[c]	0.937[b]
506	0.738[b]	0.818[bc]	1.169[b]	0.908[b]
659	0.739[b]	0.731[c]	1.001[bc]	0.824[b]
P[*] (with Ambient)	0.0003	0.0001	0.0001	0.0001
P (without Ambient)	0.32	0.0001	0.01	0.17
LSD[#]	0.145	0.147	0.186	0.123

[+]The first value (354) is from the open plots (no chambers); other values are from within chambers. Values for CO_2 are daytime means.

[*]P values are the significance levels for the CO_2 effect from ANOVA.

[#]LSD = least significant difference.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

To be multiple
✓ Open field plot
✓ Ambient chamber

IV. COWPEA STUDIES

A. Row Crop Study: Growth and Yield in Cowpeas

1. Experimental Design and Methodology

Carbon dioxide exposure system: This study was conducted at the Tuskegee Institute farm site on a Norfolk sandy loam (Typic Paleudt) on which cowpeas (Vigna unguiculata [L.] Walp., 'Pinkeye Purple Hull') plants were grown in open top chambers. The cowpeas were grown in one open field plot with no chamber and in three open top chambers with CO_2 ~~levels of~~ at 0, 150 and 300 ppm above the ambient CO_2 concentration. The design of the open top chambers is described elsewhere in this report. The equipment and procedures for dispensing and monitoring of CO_2 were identical to those used in the sweet potato study with one exception. The sampling timer was designed for a maximum of 16 different samples. Fifteen of these sampling periods were used by the sweet potato study. Therefore, the open field plot and the chamber with no added CO_2 were not sampled. The one remaining sampling period was used to sample the two chambers with elevated levels of CO_2, one at a time, on alternate days. The switch from one chamber to the other was made after the morning calibration. After both the morning and evening calibrations, the sample timer was temporarily disabled, both of the chambers with elevated CO_2 were sampled, and the flow rates were adjusted as described elsewhere in this report.

Data on actual CO_2 levels in the two chambers with elevated CO_2 were selected for analysis in a manner similar to that used for the sweet potato study, with the exception that nine pairs of adjacent days were selected in order to provide data on both chambers. Since the open

field plot and the chamber with no added CO_2 were not monitored, the values for the similar sweet potato plots were used. Analysis of this data yielded actual daytime mean CO_2 concentrations of 354 ppm in the open field plot and 354, 506 and 655 ppm in the chambers (Table 24).

Plant material: Cowpea seeds were sown on June 13 in continuous rows oriented in a northwest-southeast direction. There were two rows in each plot with 90 cm between them. When the plants were about 10 cm in height, they were thinned to a spacing of 7.5 cm between plants, thus giving 36 plants per row. Water was supplied to each plot by drip irrigation whenever the sweet potato irrigation system was turned on. In addition, the cowpea plants were watered by hand twice each day. The soil had been fertilized prior to planting at a rate of 7.3 kg of nitrogen, 7.3 kg of phosphorus and 11.0 kg of potassium per ha. The daytime temperatures during the growing season averaged approximately 31°C (approximate range: 23°C to 39°C). In general, the weather conditions were dominated by sunny, hot and humid days with sporadic clouds, and occasional showers.

Morphological measurements: During the growing season, observations were made on morphological and physiological parameters, and samples of plant parts were collected for chemical analyses. The measurements of the different parameters were done on randomly selected plants from both rows.

Fresh and dry weights, leaf area, stomatal density and the number of nodules were measured on 10 plants 41 days after sowing. The dry weights were determined after drying in an oven at 65°C for 48 hours. Leaf area was measured using a LI-COR LI-3100 area meter. Stomata were counted on epidermal peels from both adaxial and abaxial surfaces of lateral leaflets from the third unfolded leaf (counting back from the tip). The

epidermal peels were taken from a point midway between the midrib and the leaf margin and halfway between the apex and base of the leaflet. The counts were made using a binocular microscope connected to video camera and a monitor. The area of the monitor screen correlated to 0.0567 mm^2 of leaf surface. Five counts were made on each surface of each leaflet.

At maturity (79 days after sowing), plant height, the number of seeds and the total fresh weight of seeds were measured on 24 random plants.

Nitrogen content: Nitrogen content was determined at 41 and 79 days using the same plants that were used for the morphological measurements. The plant parts were separated into roots, shoots, grains and dried in an oven at 65°C for 48 hours. The samples were then milled separately to a 40 mesh size. The nitrogen content of the ground samples was estimated by the Kjeldahl method using Tecator Digestion System 40, 1016 Digester and Kjeltec System 1002 Distilling Unit. In order to find the total of mg nitrogen content, a factor 0.7 was multiplied to the titration values (0.05N H_2SO_4 was used for titration). Bremmer and Mulvaney (1982) found that 1 ml 0.01N H_2SO_4 = 0.14 mg NH_4^+-N. The conversion of mg nitrogen to protein nitrogen was done by multiplying the total mg nitrogen value by a factor 6.25.

Statistical analyses: A complete randomized design was used to analyze the data. This design was decided according to previous experience and topographic aspect of the experimental site. This site did not show any significant agronomic variations or slopes. Steel and Torrie (1980) advised the use of a complete randomized design when the experimental units are essentially homogeneous, that means, the variation among them is small, and grouping them in blocks would be little more than random procedure.

The effects of CO_2 concentrations on cowpea plants were examined

for each parameter separately, using analysis of variance. When the analysis of variance yielded significant values at the 0.05 level, significant differences among treatment means at 0.05 level were determined with LSD test (Least Significant Different). The statistical analysis was done by using SAS (Statistical Analysis System, SAS Institute Cary, NC) and MMSTAT (a statistical analysis program written and utilized at Tuskegee Institute).

2. Growth, Yield and Nitrogen Content

Most of the parameters studied during this investigation showed significant growth responses of cowpeas to CO_2 enrichment.

The fresh and dry weights of whole plants grown in the open plot (354 ppm CO_2) and in the ambient chamber (354 ppm CO_2) increased 25% and 19% respectively (?) (Table 25), but the increment was not significant. However, the fresh weight increased to 92% in 655 ppm CO_2 as compared to the chamber 354 ppm CO_2 grown plants. The dry matter also increased to 84% and 90% in 655 ppm and 506 ppm CO_2 respectively as compared to those grown at 354 ppm CO_2 in chamber. That means that CO_2 enrichment had a positive response to water storage and accumulation of dry matter in cowpea. Wittwer and Robb (1964) reported increase in fresh weight of lettuce, cucumbers, and tomatoes grown in greenhouse. Kramer (1981) and Krizek (1984) also reported dry matter accumulation in some plants exposed to CO_2 enrichment.

The number of leaves, leaf area, and plant height were significantly greater at 655, 506, 354 ppm CO_2 in chambers than in the open plot 354 ppm CO_2 grown plants 41 and 79 days after planting. The increase in the number of leaves was about 46% higher in 655 ppm CO_2 as compared to 354 ppm CO_2 in chamber. Sionit et al. (1984b) indicated that, stem

length, basal stem diameter, and number of branches and leaves increased at higher CO_2 than at lower CO_2 concentrations for species: Liquidambar styraciflua and Pinus taeda.

Plants grown in the ambient chamber had leaflets that were 29% larger than, but not significantly different from, leaflets from plants grown in the open field plot. Similar response was observed in three different CO_2 concentrations (655, 506 and 354 ppm CO_2 grown plants) although 26% larger leaf area was found in 506 ppm CO_2 than in 655 ppm CO_2. This decrease in leaf area at 655 ppm CO_2 may be associated with an increased population of aphids in this chamber during the growing season of cowpea. Patterson et al.(1984) and Margaret and Thorne (1967) reported an increase of leaf area in response to increased CO_2 concentrations.

The plant height showed significant increase with increased CO_2 concentrations at 41 and 79 days after sowing (Table 26). A comparison of the open plot 354 ppm CO_2 with the chamber 354 ppm CO_2 grown plants did not show any significant differences, but the chamber 354 ppm CO_2 was 11% higher than the open plot 354 ppm CO_2 grown plants at 41 days after sowing. At the juvenile stage (41 days) the plant height in 655 ppm was 107% higher than in the open plot 354 ppm CO_2. At maturity (79 days) the plant height in 655 ppm CO_2 was 58% higher as compared to the chamber 354 ppm CO_2 grown plants. Thus, it is interesting to note that the CO_2 effect was more significant during the juvenile stage than at maturity. Studies conducted with white pine (Funsch et al., 1970) and crab apple seedlings (Krizek et al., 1970, 1971) showed that increased CO_2 concentration resulted in the increase in height and production of lateral branches.

The effect of enriched CO_2 concentrations was also significant in

the nodule numbers, pod numbers and total fresh weight of seeds. The nodule numbers did not differ in the open plot 354 ppm CO_2 as compared to the chamber 354 ppm CO_2 grown plants. But greater increase of about 190% was observed in 655 ppm CO_2 grown plants than in the chamber 354 ppm CO_2 grown plants. The CO_2 enrichment acted on the number and size of the nodules and that resulted in a positive growth response or an increased plant biomass. Gary and Brun (1981) reported an increase in nodule mass and indicated that the increased total nodule activity in response to CO_2 enrichment was a consequence of a general growth response of the plant.

A similar response of pod numbers was observed with increase in CO_2 concentrations. The pod numbers in cowpeas increased 19% more in the open field plot as compared to the ambient chamber. The pod numbers increased 76% more at 655 ppm as compared to the ~~354 ppm CO_2~~ ambient chamber. Rogers et al. (1982) reported an increase in pod numbers of 'Bragg' soybeans grown under two watering regimes with increased CO_2 concentrations.

The CO_2 enrichment enhanced the total fresh weight of seed significantly (Table 27). The effect of chamber however was not significant even though the open plot 354 ppm CO_2 had 13% more seeds than the chamber 354 ppm CO_2 grown plants. A great increase of about 79% was found in 655 ppm CO_2 as compared to the chamber 354 ppm CO_2 grown plants. Gifford (1979) pointed out that an increase of CO_2 concentration 255 ppm above the ambient open plot concentration increased the yield of water-stressed wheat above that of stressed plants grown at the normal concentration of CO_2, resulting in increase water use efficiency.

~~Strain et al. (1984) also reported increases in yield of some plant~~

species including field crops in response to an increase in carbon dioxide concentration.

Nitrogen content in cowpea plants was also studied at 41 and 79 days after sowing under increased CO_2 concentrations. During this study, the chamber effect was not carried out significantly for both roots and shoots at 41 days after sowing. Protein nitrogen was 14% greater in shoots of plants grown in the ambient chamber than in the open field plot. No significant differences were found among the means of the three chambers 655, 506 and 354 ppm CO_2 for both roots and shoots during the juvenile stage (41 days after sowing).

At maturity (79 days after sowing), the nitrogen in roots and seeds varied significantly with increased CO_2 concentrations. The nitrogen content in the roots increased 91% in the ambient chamber as compared to plants grown in the open field plot. However no significant difference in nitrogen content was found for shoots and seeds in the open field plot or in the chamber grown plants. The plants grown in the ambient chamber had 25% and 16% more protein nitrogen in the shoots and seeds respectively. The protein nitrogen in roots decreased significantly in the three chambers 655, 506 and 354 ppm CO_2 with increased CO_2 concentrations (Table 28). In the ambient chamber 354 ppm CO_2 grown plants, the protein nitrogen in roots was about 23% higher than the protein nitrogen in 655 ppm CO_2 grown plants. The protein nitrogen content in shoots was not found to be significant in 655, 506 and 354 ppm CO_2 chambers including open field plot. However, the protein nitrogen in seeds was 25% higher in the 506 ppm than in the open plot (354 ppm of CO_2). Madsen (1968) found that the total contents of carbohydrates measure in the leaves increased in proportion to the concentration of CO_2 in the

surrounding air, until a maximum content was reached at a concentration of 2200 ppm CO_2. Allen (1982) also found an increase of total N harvested, when given on a basis of individual plant (mg N per plant), 25, 26 and 45% over the control 330 ppm CO_2.

3. Stomatal Conductance

Methods: Measurements of stomatal conductance on leaflets of cowpeas were made on July 19, 37 days after planting. The last major rainfall prior to the measurements occurred on July 12 (2.2 cm), and July 13 (2.8 cm), in addition to the rainfall, plants were watered twice a day to minimize moisture stress

Because both environmental conditions and stomatal conductance values change throughout the day, the four treatments were sampled in a random, repeating cycle. Each cycle took approximately one hour to complete. Measurements began as soon as the dew evaporated from the leaves and continued for eight cycles (approximately 830 to 1630 hours CST).

The selection of plants and leaves for measurement was designed to maximize the sampling of the variation among the plants and to minimize the variation among the leaves within the plants. For each treatment in each cycle, one leaf was measured on each of six plants. The plants for cycles 1 through 4 were chosen randomly. Measurements for cycles 5 through 8 were conducted on the same plants that were used on cycles 1 through 4. This procedure minimized the disturbance to the plants by sampling each plant only twice during the day.

Morphological observations indicated that the first (counting back from the shoot tip) unfolded leaf on a growing branch was still expanding. Thus, whenever possible, measurements were made on a lateral leaflet from the third unfolded leaf. Only leaves that were relatively unshaded by

other leaves were selected, but no attempt was made to select leaves with any particular orientation with respect to the sun. Leaves were measured in their natural orientation. If no unshaded lateral leaflet on the third leaf could be found on a plant, the second and then the fourth leaves were examined until an acceptable lateral leaflet was located. If no lateral leaflets were acceptable, a terminal leaflet was used. Most measurements were conducted on the lateral leaflets of the 2nd and 3rd leaves.

Measurements of stomatal conductance were made with a LI-COR LI-700 transient porometer. Because the porometer measures only one leaf surface at a time, and because cowpeas are amphistomatous, separate conductance measurements were made for the two leaf surfaces. The abaxial surface was measured first, followed by the adaxial surface, using a different part of the leaf. All readings were taken using the second complete drying cycle of the porometer. Simultaneous measurements of photosynthetically active radiation were made using a LI-COR quantum sensor attached to the porometer cup. Notes were also made on the amount of cloud cover in the sky and on whether or not the sun was obscured by clouds during the measurement. Leaf temperature measurements were made using a fine-wire thermocouple built into the porometer cup. During measurements inside open top chambers, the chamber door was closed so as to maintain the desired carbon dioxide concentration.

Analyses of variances and LSD tests were performed on the total (adaxial plus abaxial) stomatal conductance readings.

Results and discussion: Conductance and stomatal densities were studied on adaxial and abaxial surfaces separately (Table 29). Stomatal conductances of adaxial and abaxial surfaces in plants from the ambient chamber were not different from conductances in plants from the open field

plot. However, the conductance for both adaxial and abaxial surfaces decreased 0.1% and 1% respectively in the open field plot (354 ppm CO_2), than as compared to the ambient chamber, 354 ppm CO_2 grown plants. The stomatal conductance of adaxial layers decreased 23% more in 655 ppm CO_2 than in the chamber 354 ppm CO_2 grown plants. In abaxial layers the conductance decreased in 655 ppm CO_2 than in the chamber 354 ppm CO_2 grown plants. Meidner and Mansfield (1968), Raschke (1975) and Sheriff (1979) reported that stomata obviously respond to CO_2. Katellapper (1963) showed that response of stomata to the CO_2, was mediated by the CO_2 concentrations of the intercellular airspaces in the leaf, rather than directly by the ambient CO_2 concentration. In general, stomata open as the intercellular CO_2 concentration falls, and close as it rises. However, Mansfield (1971) and Zelitch (1969) observed that at some instances, variations in the intercellular CO_2 concentrations might not substantially affect stomatal apertures.

The stomatal densities of adaxial and abaxial surfaces showed significant differences when the open field plot was compared to the ambient chamber grown plants. In the adaxial surface of the leaves, about 50% more stomata were found in the open field plot than in the ambient chamber. The abaxial surface also showed 21% more stomata in the open field plot than in the ambient chamber grown plants. No significant responses in stomatal densities were found ~~among the three chambers~~ at 655, 506 and 354 ppm CO_2. However, we observed increases in stomatal densities of about 23% and 11% for adaxial and abaxial surfaces respectively in 655 ppm CO_2 over the 354 ppm CO_2 chamber grown plants. O'Leary and Knech (1981) worked on beans (Phaseolus vulgaris) leaves, but they did not report significant difference in stomatal density on the adaxial surface.

However, they did find a significant difference in stomata density on the abaxial surfaces of leaves, which decreased with increased CO_2 concentration.

Conclusion is needed

Table 24. Seasonal mean CO_2 concentrations in cowpea study plots. Values represent means (± S.E.) of 108 readings (54 daytime, 54 nighttime) taken every two hours from nine selected days between Julian day 165 and Julian day 232.

CO_2 Added (ppm)		Mean CO_2 Concentration (ppm)		
Nominal Treatment	Actual Daytime	Daytime+	Nighttime+	24 Hours*
Open Field	0	354 ± 15	440 ± 50	397 ± 57
+0	0	354 ± 15	437 ± 48	396 ± 54
+150	152	506 ± 17	593 ± 48	549 ± 56
+300	301	655 ± 18	729 ± 37	692 ± 47

+N = 54

*N = 108

Table 25. Effect of different CO_2 concentrations on the fresh and dry weights of whole plants of cowpeas grown in an open plot and within chambers after 41 days. The values are means (± S.E.); N = 10 plants.

CO_2 (ppm)	Fresh Weight of Whole Plant (gm)	Dry Weight of Whole Plant (gm)
354[+]	123.2 ± 17.4[b]	16.7 ± 2.2[b]
354	98.4 ± 15.5[b]	14.0 ± 1.9[b]
506	179.3 ± 22.4[a]	26.6 ± 3.5[a]
655	188.6 ± 24.0[a]	25.8 ± 3.8[a]
LSD	50.0	8.5
	S	S

[+]The first value (354) is from the open plot (no chamber); other values are from within chambers. Values for CO_2 are daytime means.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

S = Significant

Table 26. Effect of different CO_2 concentrations on the number of leaves, leaflet area and plant height of cowpeas grown in an open plot and within chambers. Values for number of leaves and leaflet area were taken at 41 days. The values are means (± S.E.); n = 10 (leaf number and leaflet area) or 24 (height).

CO_2 (ppm)	Number of leaves	Leaflet Area (cm^2)	Plant Height (cm)	
			After 41 days	After 79 days
354[+]	10.3 ± 0.7[ab]	48.7 ± 5.4[b]	25.7 ± 0.6[c]	122.7 ± 10.7[b]
354	8.1 ± 0.4[c]	63.0 ± 2.2[ab]	28.4 ± 1.1[c]	158.0 ± 14.0[b]
506	9.9 ± 0.5[b]	73.4 ± 8.0[a]	43.2 ± 1.5[b]	224.1 ± 14.1[a]
655	11.8 ± 0.8[a]	58.2 ± 4.9[ab]	58.8 ± 3.0[a]	249.1 ± 14.4[a]
LSD	1.8	15.9	5.1	37.9
	S	S	S	S

[+]The first value (354) is from the open plots (no chamber); other values are from within chambers. Values for CO_2 are daytime means.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

S = Significant

Table 27. Effect of different CO_2 concentrations on the number of nodules (at 41 days) and on the number of pods per plant and the total fresh seeds (at 79 days) of cowpeas grown in an open plot and within chambers. The values are means (± S.E.); n = 10 plants (nodules) or 24 plants (pods and seeds).

CO2 (ppm)	Number of Nodules	Numbers of Pods	Total Fresh Weight of Seeds (gm)
354[+]	3.1 ± 1.9[b]	9.9 ± 0.8[bc]	11.4 ± 1.1[b]
354	3.1 ± 1.2[b]	8.3 ± 0.8[c]	10.1 ± 1.3[b]
506	6.4 ± 2.1[ab]	12.5 ± 1.3[ab]	18.3 ± 2.4[a]
655	9.0 ± 2.4[a]	14.6 ± 1.3[a]	18.1 ± 2.3[a]
LSD	5.6	3.1	5.3
	S	S	S

[+]The first value (354) is from the open plots (no chamber); other values are from within chambers. Values for CO_2 are daytime means.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

S = Significant

Table 28. Effect of different CO_2 concentrations on the protein nitrogen content of cowpeas grown in an open plot and within chambers at maturity (79 days). The values are means (± S.E.); n = 2 determinations from pooled samples representing 10 plants (41 days) or 24 plants (79 days).

	Protein Nitrogen (%)				
	After 41 Days of Growth		After 79 Days of Growth		
CO_2 (ppm)	Roots	Shoots	Roots	Shoots	Seeds
354[+]	9.2 ± 0.4	17.4 ± 2.6	4.5 ± 0.3[c]	5.8 ± 1.1	19.5 ± 0.5[b]
354	8.9 ± 2.5	19.8 ± 2.6	8.5 ± 0.3[c]	7.3 ± 0.4	22.7 ± 0.5[ab]
506	7.5 ± 1.8	15.3 ± 1.6	6.0 ± 0.4[b]	5.5 ± 0.6	24.4 ± 1.4[a]
655	8.8 ± 0.1	20.1 ± 0.0	7.0 ± 0.2[b]	6.5 ± 0.0	23.3 ± 0.4[a]
LSD	6.0	6.0	1.0	2.6	3.2
	N.S.	N.S.	S	N.S.	S

[+]The first value (354) is from the open plots (no chamber); other values are from within chambers. Values for CO_2 are daytime means.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

S = Significant, NS = Nonsignificant

Table 29. Mean (± S.E.) stomatal conductances (between the hours of 8:41 and 16:21 on July 19) and stomatal densities for cowpeas grown in an open plot and within chambers at elevated levels of CO_2. n = 48 (conductance) or 10 (density).

CO_2 (ppm)	Stomatal Conductance (sec cm^{-1})			Stomatal Density (number mm^{-2})		
	Adaxial	Abaxial	Total	Adaxial	Abaxial	Total
354+	0.312 ± 0.02[b]	0.452 ± 0.02[b]	0.780 ± 0.04[a]	112 ± 11[b]	290 ± 13[b]	402 ± 21[b]
354	0.312 ± 0.02[b]	0.458 ± 0.03[ab]	0.770 ± 0.04[a]	74 ± 9[a]	239 ± 12[a]	314 ± 19[a]
506	0.342 ± 0.02[b]	0.568 ± 0.04[c]	0.910 ± 0.06[b]	85 ± 5[ab]	257 ± 12[ab]	342 ± 14[ab]
655	0.254 ± 0.01[a]	0.412 ± 0.02[a]	0.666 ± 0.03[a]	92 ± 13[a]	265 ± 20[ab]	357 ± 31[ab]
LSD	0.051	0.083	0.124	28	43	??
	S	S	S	S	S	S

+The first value (354) is from the open plots (no chamber); other values are from within chambers. Values for CO_2 are daytime means.

[a]Within each column, values sharing the same letter are not significantly different at the 0.05 level, according to the LSD test.

S = Significant

B. Pot Study: Growth and Nitrogen Fixation in Cowpeas

1. Experimental Design and Methodology

Reports from the National Academy of Science has indicated that the CO_2 in our atmosphere has increased by 7% over the last 25 years (ref ?). Indications are that this atmospheric CO_2 will continue to rise, perhaps at a more rapid rate. It is therefore evident that there is a need to know what possible effect this increased level of CO_2 will have on the productivity of crop plants. Thus, as part of a project funded by the United States Department of Energy, Tuskegee Institute initiated a study to determine the effects of various CO_2 concentrations above ambient on the growth of cowpeas.

Seeds of the cowpea cultivar 'Pinkeye purplehull' were planted 13 x 13 cm polyethylene pots using a Norfolk sandy loam fertilized according to soil test recommendations. After emergence, plants were thinned to three per pot. Groups of 10 pots were placed in open top chambers with the following CO_2 treatments (daytime means).

(a) 354 ppm CO_2 (ambient)

(b) 506 ppm CO_2

(c) 655 ppm CO_2

(d) An additional check treatment was added in which one group of 10 pots were placed in the open field.

The open top chambers were more or less 3 x 3 m cylindrical aluminum frames open at both ends and covered with polyethylene. CO_2 as desired was piped into each chamber through a delivery system from a central tank. The CO_2 experimental system is described in detail elsewhere in this report. Throughout the growing season pots were watered to keep the soil moisture at field capacity (-1/3 bar). Insects were controlled by period

spraying with ? or Malathion.

When plants were 7 days old, leaves were counted and plant height determined at 7 day intervals thereafter. At the 10% bloom stage one plant was removed from each pot, roots were washed and nodules counted: Intact roots with nodules were placed in a 125 ml flask and the rate of N_2 fixation was determined, using the acetylene reduction technique (ref ?). The top portion of each plant was dried at 70°C for 48 hours and then weighed for dry matter determination. Plants not used for N_2 fixation were left in the pots until maturity for seed yield determination.

2. Growth, Yield and Nitrogen Fixation

Both plant height and leaf number tended to increase with increase in plant age (days after planting) at all levels of CO_2 treatment (Table 30). Generally, plants in chambers were taller than those in the open field. It was also noted that chambers with 506 and 655 ppm CO_2 produced approximately three fewer leaves than those in the open field (354 ppm CO_2), and those in the ambient chamber (354 ppm CO_2) had approximately six fewer leaves (Table 30). In addition, plants in both chambers were taller (?) than those in the open field (Table 30). At 49 days after emergence plants in chambers with 506 and 655 ppm were 7.4 and 10.7 cm, respectively taller than those in the open field (Table 30). Rate of growth was not significantly different between chambers, indicating that CO_2 concentration had no effect on rate of growth (Fig 26). However, at 49 days after planting, plants in chambers with 506 and 655 ppm CO_2 had a growth rate of approximately 50% faster than those in the open field (Fig 26).

Number of nodules, though not significantly so, were generally lower in chambers with CO_2 concentrations higher than ambient (Table 31).

The trend in rate of N_2 fixation was opposite to that of N_2 fixation (?). The rate of N_2 fixation in chambers with CO_2 concentrations above ambient was higher than that in the chamber with the ambient CO_2 level, though not significantly so (Table 31).

Biomass yield as indicated by plant dry weight was highest for plants in the open field (Fig. 27). While biomass production was highest in the open field, seed yield was greatest in chambers with 655 ppm (Fig 27). Seed yield in the 655 ppm treatment was only slightly more than that in the open field but was up to 27% higher than in the 506 ppm treatment and 75% more than in the ambient CO_2 chamber. It should be noted that seed yield was highest in chambers with the highest rate of CO_2. Thus, CO_2 concentration up to 655 ppm did not adversely affect seed production in cowpeas.

Table 30. Effect of elevated levels of CO_2 on plant height and leaf number of cowpeas.

Days After Planting	CO_2 Treatment							
	Open Field		354		506		655	
	Plant Height (cm)	Number of Leaves	Plant Height (cm)	Number of Leaves	Plant Height (cm)	Number of Leaves	Plant Height (cm)	Number of Leaves
7	3.9^a	2.0^a	3.6^a	2.0^a	3.9^a	2.0^a	3.9^a	2.0^a
14	5.2^b	5.1^b	5.3^b	5.0^b	5.7^b	5.6^b	5.0^b	5.6^b
21	7.0^c	9.7^c	7.3^c	9.9^c	7.6^c	10.4^c	7.9^c	9.8^c
28	12.8^d	18.7^d	12.0^d	16.1^d	11.1^d	17.2^d	11.8^d	17.8^d
35	13.3^e	24.0^e	15.5^e	18.8^e	15.3^e	12.6^e	16.3^e	23.1^e
42	17.3^e	29.3^f	21.0^f	21.0^f	21.6^f	26.4^f	21.8^f	24.6^f
49	21.4^g	28.9^f	32.1^h	22.4^f	28.8^h	26.7^e?	32.1^h	26.0^h

[a]Within rows and columns means followed by the same letters are not significantly different at the 0.05 probability level by Duncan's Multiple Range Test.

Table 31. Effect of elevated levels of CO_2 on the rate of N_2-fixation of cowpeas. All values are from plants within chambers.

CO_2 Fixed (ppm)	Number of Nodules per Plant	Rate of N_2 (um plant-1 hr-1)
354	12	432
506	7	662
655	8	612
	NS	NS

NS = not significantly different.

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APPENDICES

Growth and Yield Response of Sweet Potato (Ipomoea batatas) to Atmospheric CO_2 Enrichment

N. C. Bhattacharya, P. K. Biswas, Sheila Bhattacharya, Nasser Sionit, and B. R. Strain

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