THESIS

EFFECT OF SULFUR DIOXIDE AND HIGH LEVELS OF CARBON DIOXIDE ON CARNATIONS

Submitted by

Roderick J. McKeag

In partial fulfillment of the requirements for the Degree of Master of Science

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Chapter I INTRODUCTION

Goldsberry and Holley (8) have observed that CO_2 levels in a closed greenhouse filled with plants may vary from 400 ppm at night to slightly more than 100 ppm during high light periods. Outdoor CO_2 levels may vary 250 to 350 ppm, but the fluctuations are not diurnal. Since CO_2 is essential for photosynthesis and CO_2 levels tend to drop during the daytime in a closed greenhouse, experiments to show the effect of CO_2 on carnation yield and quality were initiated. It has been established that additional CO_2 , up to 550 ppm, results in higher quality and yield of carnations (9). Today many crops are grown in atmospheres that contain added CO_2 at concentrations approximately twice atmospheric. Additional benefits from high levels of CO_2 are in question.

 CO_2 can be obtained from many processes. In Colorado, dry ice converters, liquid CO_2 and charcoal burning have been tested (14), but less expensive sources have been found such as the Tectrol¹/ CO_2 generator. This unit involves the burning of gaseous fuel, natural gas or propane, with the release of the combustion products CO_2 and water. Through an efficient burning of pure natural gas (methane) high purity CO_2 can be obtained in a 1:1 ratio based on volume. The

^{1/} Tectrol Division, Whirlpool Corporation, St. Joseph, Michigan.

purity of CO_2 obtained is directly proportional to the purity of the fuel burned (23, 28).

Since sulfur is a contaminant of natural gas (23), and its combustion product, SO_2 , is injurious to some plants in very small concentrations (1, 17, 25, 30, 32, 40, 42), it is essential to detect SO_2 in the greenhouse before it reaches dangerous concentrations.

The problem

As information regarding CO_2 saturation in carnations is theoretical, data on the effects of high levels of CO_2 in relation to total productivity would be beneficial.

There is some skepticism in the use of natural gas as a CO_2 source due to its sulfur content. Will the sulfur content of natural gas create injurious conditions that may damage carnations? Can a plant be found and used as an indicator of the presence of SO_2 before it reaches levels damaging to the carnation?

Problem analysis

Since levels of CO_2 above 600 ppm are of questionable value for carnations, this experiment was designed to compare carnation growth at average levels of 1200 ppm with 600 ppm and with 300 ppm (atmospheric). Kean grade of flowers, yield, and quality were used to compare the three levels of CO_2 .

In order to find a plant indicator for SO_2 injury (a plant more sensitive to SO_2 than the carnation) a visible injury index system was used to compare plant sensitivity to SO_2 .

Delimitations

The study dealing with levels of CO₂ was limited to 3

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greenhouses with 4 blocks of carnations of the following varieties: White No. 88 and Safari (2-year-old plants), and Pink Coquette and Elliott's White (young plants) in each greenhouse. Data was collected from June, 1964 to June, 1965. CO_2 was measured periodically by a ESA Lira infrared CO_2 analyzer.

According to Holley and Juengling (15), a higher temperature is probably beneficial when CO_2 levels are increased. Gaastra (6) states that the optimum temperature may rise slightly with increased levels of CO_2 ; therefore, the temperature was set as follows:

CO2	Greenhouse	Temperatures		
level	number	Night	Day	Cooling above
1200 ppm	l	54°	60 ⁰	69°
600 ppm	2	54 ⁰	60 ⁰	69 ⁰
Atmospheric	3	52 ⁰	60 ⁰	65°1/
		52 ⁰	60 ⁰	69°2/

1/ November 15 to March 15.

2/ Rest of season.

All other factors such as moisture, nutrition, and light were maintained approximately equal in all greenhouses.

The portion of the study dealing with SO_2 sensitivity was limited to a chamber 3 x 3 x 6 feet set in an area of low light (200-400 ft-c) during January, February, and March, 1965.

Plants used were planted in pots and moved from greenhouse conditions to the test area. They included tomato varieties Manapal, Fireball, Marglobe; petunias, single and double white; snapdragon (white greenhouse varieties); chrysanthemums, Jupiter, Princess Ann, Shasta (white); and carnation, White Sim.

Definition of terms

Injury index was obtained by assigning numbers according to degree of damage as follows: (1) slight visible damage; (2) approximately one-half of the leaf damaged; (3) all but veins damaged; (4) complete leaf damaged.

Air pollution is appropriately defined by Tetzloff et al. (29) as "the presence in the outdoor atmosphere of one or more air contaminants or combinations thereof in such quantities and of such duration that they are or may tend to be injurious to human, plant, or animal life or property or unreasonably interfere with the comfortable enjoyment of life or property or the conduct of business."

Scrubbing refers to the process of passing exhaust gases through a solvent in order to rid those gases of one or more air pollutants.

Invisible injury is the term used to describe a reduction in growth rate of plants without visible indications.

Chapter II

REVIEW OF LITERATURE

Part I.

CO₂ research

 CO_2 in relation to photosynthesis has been studied for 200 years (18). Wittwer (37) reported that Kreusler, in 1885, performed a quantitative study and related the rate of photosynthesis to the concentration of CO₂. Work during the early twentieth century generally showed increased productivity (3, 27, 37).

Work progressed sporadically up to the present time but results were inconsistent probably due to impurities in the CO_2 source from burning charcoal, coal gas, or paraffin (37). Some early sources of CO_2 included acid on sodium bicarbonate, burning patented fuels, and mulch manure (27). Later, Holley and Goldsberry (14) added dry ice and bottled CO_2 to the list. Today burning of propane is favored because of economy and purity (23) although widespread use of natural gas seems inevitable.

Contemporary workers and their results

Holley (16) in a recent review of CO_2 has summed up some of the general advantages of using CO_2 : (a) increased lateral growth, (b) decreased bud abortion, (c) increased size, (d) increased yield and (e) decreased time of maturity. Experiments in support of the above advantages appear in numerous places in the literature (14, 28,

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37, 38, 39). Goldsberry and Holley (8) increased dry matter, number of lateral growths, and yield of carnations by additional CO_2 . Goldsberry (9) stated that using CO_2 at 550 ppm as compared with 200 ppm increased yield 38%, increased dry matter, and shortened production time of carnations by two weeks. In a later article on the effect of CO_2 on roses (10) Goldsberry and Holley report a 25% increase in yield and increased stem length.

Wittwer et al. (37, 38, 39) showed that tomatoes, lettuce, and cucumbers were very responsive to CO_2 . They demonstrated that yield increased more than 100% in some cases. Plots with 800-2000 ppm of CO_2 were compared with plots receiving 125-500 ppm.

Other investigators (26) burned propane for CO_2 and raised CO_2 levels to 500 ppm (calculated). Roses and chrysanthemums increased in growth, but no significant differences were found with carnations.

Environmental relations

Under simulated field conditions, with 300 ppm of CO_2 , 70 to 76F, and light to 6000 ft-c; CO_2 limited photosynthesis.(6). By keeping temperature at 70 to 76F and light intensity up to 6000 ft-c and raising the CO_2 level to 1000 ppm, CO_2 saturation was reached in sugar beets, turnips, and tomatoes but not in cucumbers (6). For sugar beets, turnips, and tomatoes, light, temperature, genetics or some other factor probably limited growth in this environment. In the same study (6) the temperature was varied on tomatoes and turnips. When the temperature was raised to 86-93F, an increase in photosynthesis occurred showing that, at light of 6000 ft-c and CO_2 saturation at 1000 ppm, temperature limited photosynthesis. If temperature limited photosynthesis at higher CO_2 concentrations, this could be explained on the basis of a biochemical process and might indicate that a genetic mechanism limits the photosynthetic rate of plants under optimum conditions of light, temperature, and CO_2 . Another indication of a genetic mechanism for light saturation is the quantum efficiencies shown by Gaastra's graphs (6) of photosynthetic rate for several crop plants. In summary of the above work, a generalization was made that for many higher plants, CO_2 saturation seems to be reached at concentrations of 1000 ppm and temperatures of 70 to 76F. At 450 ft-c, light limits photosynthesis. At 2000 ft-c, atmospheric CO_2 limits photosynthesis, and at 5000 ft-c and CO_2 saturation at 1000 ppm, temperatures between 70 and 76F limit photosynthesis.

In recognition of these relationships, Holley and Juengling (15) postulated that improved carnation growth from added CO_2 has probably been limited by low temperature. With added CO_2 there is a need for evaluation of all other environmental factors (26). Goldsberry and Holley (8) concluded that nutrient requirements will increase with CO_2 levels. Optimal temperature, light, and other environmental factors will also increase with added CO_2 (8, 39). Wittwer et al. (38) suggested that the optimum temperature for growing greenhouse lettuce will vary with CO_2 levels, light, age of plant, and variety. Working with vegetables with relatively low quantum efficiencies, they (39) stated that added CO_2 will benefit plant growth even when sunlight is a limiting factor.

CO₂ saturation

Gaastra (6) postulated that the influence of CO₂ may be in direct relation to the photosynthetic process or by degree of diffusion

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through the stomata. Since CO_2 diffuses through the stomata and into the aqueous portion of the leaf cells, it is plausible that this diffusion rate could be the limiting factor in CO_2 saturation. It has been demonstrated (22) that the aperture width of the stomatal opening decreases with increased concentration of CO_2 , and CO_2 concentration inside the leaf increases as outside CO_2 concentration increases but at a decreasing rate. CO_2 utilization depends, among other things, upon stomatal movement and biochemical "capacity" (21). Scarth and Shaw (24) studied stomatal movement and photosynthesis and observed that aperture width depends on photosynthesis and reduction of CO_2 concentration in the leaf. Fick's diffusion law:

$$Q = \frac{D(C_0 - C_1) \cdot a}{1}$$

Where D is a constant

 $C_o - C_l$ is the concentration of substance at the extremes of the gradiant.

a is the width of diffusion tube and

1 is the length of diffusion tube, (6)

involves the concentration differences (in the cell sap and outside leaf), width (aperture diameter), and length of tube (thickness of leaf) as the variables in diffusion. Therefore, the stomatal aperture could affect diffusion of CO_2 into the leaf as could concentration of CO_2 or thickness of leaves. Thick leaves under very low concentrations of CO_2 reassimilated 56% of the CO_2 respired (7). At these low CO_2 levels photosynthesis depends on respiration rate and rate of CO_2 escape from the leaves. Part II.

Air pollution research

In the United States, concern over air pollution damage to plants, animals, and property dates back to the Report of the Selby Smelter Commission (17). SO_2 was found to be the main effluent causing the damage. As much as 7.1 ppm of SO_2 was measured several miles from the manufacturing plant (17). The results in this report were: (a) Plant tolerance of SO_2 was determined to be about 0.5 ppm. (b) The response of plants in their range of sensitivity to SO_2 was in proportion to concentration and time of exposure with 5 ppm for one hour doing as much damage as 1 ppm for 5 hours. (c) It was realized that the atmosphere is a natural resource and must be safeguarded from contamination.

Following the work of this commission, other researchers experimented with SO_2 to learn the symptoms of SO_2 damage on plants, the environmental factors favorable to injury, the possibility of retarded growth without visible signs of damage, the benefit from SO_2 for sulfur requirements of sulfur-deficient plants, and the mechanism for SO_2 entry into the plant and its damage.

Symptoms

The visible symptoms are striking and constant (42). In general, agreement on symptomology exists (1, 4, 32, 33, 35, 41). These symptoms are categorized into two degrees of damage: severe (acute) injury and sub-lethal (chlorotic) injury. Severe injury occurs on the leaves marginally or intercostally. The leaf appears to be water-soaked, drying to an ivory color in most species but may be brownish-red in species with high anthocyanin content. The damage does not cross the leaf veins except in extreme poisoning (32). Sublethal damage causes chlorotic leaf spots. The leaf becomes prematurely senescent and is shed. In either case, the middle-aged leaves, or the youngest leaves that are fully expanded, are the most sensitive in all species (32). Thomas et al. (33) observed that the ivory color is sometimes delayed 3-6 days. Some nongaseous damage that might be confused with SO₂ damage are white spot in alfalfa, frost injury, salt injury, and chlorosis and tip burn associated with nutritional problems (32).

Zimmerman (41) compared SO_2 damage with damage from other gaseous products such as hydrogen fluoride, chlorine, hydrogen sulfide, ammonia, mercury vapors, and ethylene. With damage from hydrogen fluoride, which is toxic at very low concentrations, the leaf tips and margins turn gray-green and may absciss. Hydrogen sulfide damage is categorized by itself since the damage is only on very young leaves. Chlorine, ammonia, and mercury vapor damage resemble SO_2 damage so closely that it is difficult, if not impossible, to distinguish them by visible signs. Ethylene at 1 ppm on roses will cause chlorosis and abscission of leaves, and in this way resembles sub-lethal damage from SO_2 .

Further comparisons and distinctions have been between SO_2 damage and smog and ozone damage by various researchers (2, 13, 20, 35). Hill et al. (13) reported that ozone can be responsible for flecking, necrotic areas and chlorosis on leaf areas between veins. However, Thomas (35) demonstrated with colored plates of ozone damage and SO_2 damage that there was a great similarity between visible

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symptoms. Smog damage has been described as brown-black mottling in tomato leaves (2), while spinach has silver-gray areas (35). Middleton (20) describes smog damage as a silvering, bronzing or blistering of the middle leaves. A discrepancy is noted here, but smog damage is not easily confused with SO₂ damage.

Environmental relations

Environmental relations are an important consideration in SO_2 poisoning. Setterstrom et al. (25) worked with this aspect of SO_2 injury in growth chambers and small controlled greenhouses. They designed experiments in which eight factors were studied. Their work is summarized as follows:

Factor	Most tolerant	Most susceptible
Temperature	Below 40F	65-105F
Humidity	Low	High
Soil moisture	Plants grown dry	Wet
Nutrition	Good levels	Poor
Pretreatment with SO_2	No effect	No effect
Light pretreatment	High light	Low light
Age of plant	Young or old	Middle age
Koist leaves	No effect	No effect

The relation of environment to SO₂ damage

Holmes et al. (17) reported that plants become more severely damaged at high relative humidity than at low relative humidity. His work was done under field conditions, and whether this holds true under greenhouse conditions is questionable. Setterstrom et al. (25) concludes that humidity is difficult to replicate, hence a difficult factor to study. In agreement with Holmes is Thomas (35), who stated that high relative humidity increased sensitivity of alfalfa. In an earlier article (32), he stated that relative humidity is not important as long as the plants are in a turgid condition. Observing that smog damage was increased by scrubbing the air to free SO₂ from the smog complex, Thomas et al. (33) attributed this to one of the following or combinations thereof: SO₂ counteracting smog activity, increased relative humidity, or hydrolysis of smog.

The statement that temperatures below 40F (41), or 5C (33), or moderate temperatures (35), cause tolerance to SO_2 is consistent with the findings of Setterstrom et al. (25). Soil moisture effects were studied by Thomas et al. (33), Zimmerman (41), and Setterstrom et al. (25); and consistent results were found. Turgid plants are susceptible while those in a flacid or wilted condition are resistent. Thomas (35) explains this in terms of stomata being open when turgid and closed when wilting.

Invisible damage theory

The invisible damage theory states that the physiological aspects of the plant are hindered or slowed down without outward (visible) manifestation. Visible damage was early associated with SO_2 , but invisible damage was debatable. Todd and Arnold (36) defined the extent of SO_2 damage on the basis of visual damage rating, reduction in fresh weight, and reduction of chlorophyll. They concluded that visual damage rating is fairly sensitive at the lower

range of injury. It is the consensus that invisible damage does not occur (4, 11, 12, 19, 30, 32, 35, 36, 41).

Bleasdale (1), however, experimented with rye grass using SO_2 at .01 to .06 ppm and found a decrease in the growth rate in the absence of visible damage. Thomas (34) concedes that at low SO_2 concentration plants may develop sulfate toxicity. In support of this excess sulfur in plants is Frazer (5), who found several times as much sulfur in plants from SO_2 -polluted areas.

SO2 for sulfur-deficient plants

Three workers investigated the possibility of SO_2 from the air being used by plants to replace SO_4 from the soil. All had positive results, but each one had a slightly different explanation. Katz and Ledingham (19) claimed that SO_2 will partly fill the requirement of sulfur for sulfur-deficient plants. Setterstrom et al. (25) state that SO_2 can increase the growth of sulfur-deficient plants. Thomas et al. (31) reported that SO_2 does not meet the sulfur requirement as well as SO_4 , but a 48% increase in alfalfa yields was due to SO_2 on sulfur-deficient plants.

Mechanism of SO2

 SO_2 is a colorless gas of pungent odor and acid taste. Fifty percent of the people in an experiment could detect its presence at 4 ppm, and at 5 ppm all could tell it was SO_2 (17).

Thomas (32) reported that the phytotoxicity of a gas depends upon: (a) absorbability, (b) acidity or alkalinity, (c) oxidation or reduction properties, (d) hormonal properties, and (e) toxicity of the element itself. He showed that SO₂ enters through the stomata to cause damage inside the plant (35). Alfalfa and potato are highly sensitive to SO_2 , and were subjected to SO_2 during the night hours with the knowledge that alfalfa stomata close in darkness while potato stomata remain open. The potato was damaged but not the alfalfa.

Thomas further showed that the damage was due to SO₂ having oxidizing properties and not to its acidity (35). Hydrogen chloride, a much stronger acid, did not produce damage while sulfuric acid did.

Most of the SO_2 is converted to SO_4 inside the plant, but some is converted to organic sulfide (35). Radiosulfur and chromatography have established that the final fate of SO_2 is in cystine and methionine (32). This has been substantiated by other workers (19, 25, 29).

Chapter III

METHODS AND MATERIALS

This study is divided into Part I which includes tests to determine the effects of high levels of CO_2 , and Part II which deals with tests to determine plant sensitivity to SO_2 from impure natural gas.

Part I: Effects of high levels of CO2

Greenhouse conditions

The three greenhouses used were oriented east-west, having dimensions of 15 x 18 feet with height of 7 feet at the eave and 10 feet at the ridge. The framework was wood, and the covering of clear fiberglass. $\frac{1}{2}$

A fan and evaporative pad cooling system was used in summer cooling and plenum tube cooling in winter. The cooling system came on at 69F in the two houses where CO₂ was added. In the control house from November 15 to March 15 cooling was at 65F and from March 15 to November 15 cooling was at 69F. The heating system was operable at 54F at night and 60F during days for all houses. Temperature controls were housed in a shelter that allowed free air passage while minimizing stray radiation. A Foxboro 24-hour hygrothermograph was used in each house to set and adjust temperatures.

[⊥] Structoglas, Inc., 11701 Shaker Boulevard, Cleveland 20, Ohio.

Irrigation was on demand with a standard nutrient solution.1/ Two applications of 6-10-4 fertilizer were applied dry on April 2 and 23 at the rate of 4 pounds per 100 square feet to insure adequate nutrition.

Thimet miticide was dusted at the time of plant mulching. Further insect control was maintained with periodic fumigations and sprays applied as needed.

Experimental plants

Each house had two raised benches with dimensions of 4 x 13 feet. Eighty plants of four different varieties and two different ages were planted in each house. Two-year-old plants of White No. 88 and Safari were in place at the start of the experiment, and rooted cuttings of Elliott's White Sim and Pink Coquette were planted June 23, 1964, in steamed soil. After the plants were pinched, a mulch of leaves was applied.

<u>Analysis</u>

Records were kept of the quantity and quality of flowers cut. Quality was determined by a grading system having four grades: (1) fancy - flowers of 24-inch stems and 25 grams in weight with no defect; (2) standard - flowers with no defects, a length of 20 inches and weight of 15 grams; (3) short - flowers with no defects but either weight or stem length limiting it from being graded higher; (4) designs - all flowers not in any of the above three categories.

A MSA Lira infrared gas analyzer was used to set the equipment for metering CO_2 to houses 1 at 1200 ppm and 2 at 600 ppm, and

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^{1/} Colorado State University, Fort Collins, Colorado.

used periodically to check these levels.

Part II: Tests to determine plant sensitivity to SO2 Chamber conditions

Natural gas used in combustion passed from an 8 psi line through a meter, and a pressure regulator reducing it to 6 psi. It, then, passed over liquid mercaptan to receive its sulfur contamination before being combusted by a small "bunsen type" burner. To cool the gas, a 2-inch pipe system of about 4 feet in length was used. A chamber to hold the plants was constructed of wood with the dimensions of $3 \times 3 \times 6$ feet. This structure was covered with polyethylene and set on a table. Four-inch fans were used on each end to dilute the combustion products. The fan connected between the chamber and the cooling pipe pulled the combustion products and air into the chamber. Temperatures of approximately 85F and CO₂ levels of 800-840 ppm could be maintained by this equipment. This system was placed in a shaded area and received only north radiation. This was measured at 200-400 ft-c by a Weston sunlight meter.

Experimental plants

Plants were grown under greenhouse conditions and moved into the chamber to be tested. All plants were watered at the beginning of each experiment.

Analysis

Plants were analyzed by a visible injury index. CO₂ measurements were made with a KSA Lira infrared gas analyzer. Temperature was measured by a laboratory thermometer. Sulfur contaminants

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in the gas and air mixture were measured by the Public Service Company of Colorado with a Model 300 iodine-bromine titrator. Total gas analysis on a set of samples was done by Tectrol, St. Joseph, Michigan; and SO_2 was checked by Mr. John B. Pate of the National Center for Atmospheric Research, Boulder, Colorado.

Chapter IV

RESULTS AND DISCUSSION

Part I.

The effects of high levels of CO_2 (1200 ppm) were compared with 600 ppm and atmospheric (300 ppm) on yield and grade of carnations. Table I shows the yield for the three treatments and indicates that 1200 ppm CO_2 concentration does not significantly increase total yield over 600 ppm. Since total flower production is in proportion to the number of lateral breaks that return from a flower stem, this analysis indicates that the optimum CO_2 level for the production of laterals is around 600 ppm. 300 ppm reduces laterals as does 1200 ppm when compared to 600 ppm.

The mean grade of flowers from the first-year plants, Elliott's White Sim and Pink Coquette, are shown in Table II. Grade data collected on the two-year-old plants was not compared since lack of support of the plants resulted in downgrading many flowers due to crooked stems. Kean grade is significantly higher under 1200 ppm of CO_2 as compared to 300 ppm. There were no significant differences in mean grade between 1200 ppm and 600 ppm, or between 600 ppm and 300 ppm. This suggests a linear relationship between grade and CO_2 concentration.

Measurements made on the flowers from the variety Elliott's White Sim further corroborate this relationship. Table III indicates that average weight and average number of leaf pairs increases with

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	1200 ppm	600 ppm	300 ppm
Safari	1103	972	957
White No. 88	976	1110	988
Total for 2-year- old plants	2079	2082	1945
Elliott's White	1229	1346	1185
Pink Coquette	828	1033	863
Total for first- year plants	2057	2379	2048
Total all plants	4136	4461	3993
5 over control	4	12	0

Table I. Yield of four carnation varieties grown at three CO₂ concentrations from October 2, 1964 to June 12, 1965.

Table II. Effects of three levels of CO₂ on mean grade of Elliott's White and Pink Coquette from October 2, 1964 to June 12, 1965

Source	D of F	Kean grade	F value	Probability
1200 ppm vs 600 ppm	1	4.15 vs 4.04	1.064	n s
1200 ppm vs 300 ppm	1	4.15 vs 3.95	7.716	.02
600 ppm vs 300 ppm	1	4.04 vs 3.95	2.493	N S

CO2 concentrations up to 1200 ppm.

Table III.	Physical measurements of flowers and stems cut at the
	origin for Elliott's White Sim from May 15, 1965 to
	May 31, 1965.

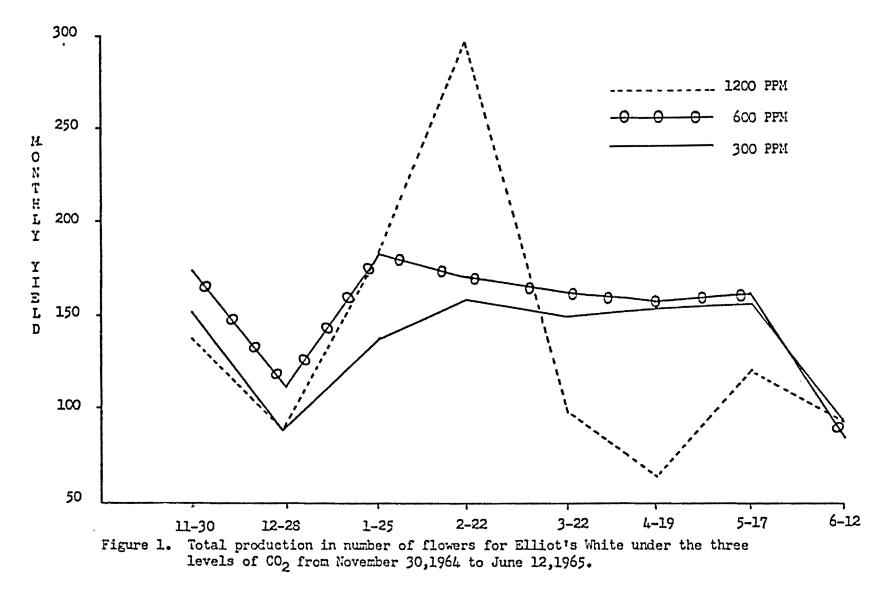
617	No. stems	Ave. weight	Ave. length	Ave. no. leaf pairs
1200 ppm	52	52.1	31 10"	18.1
600 ppm	66	48.1	4ª 0 ¹¹	17.0
300 ppm	77	41.8	31 311	16.3

Figure 1 illustrates the effect of CO_2 concentration on the timing of crops. An extremely fast second crop followed by a period of low production occurred under 1200 ppm of CO_2 . This fast second crop occurred on Elliott's White Sim and not on Pink Coquette; therefore, the response appears to be varietal. The control and 600 ppm produced more typical production curves. The response of young growth, such as laterals, to higher CO_2 is not known but appears to be very favorable at 1200 ppm. Normally this crop should return in April or May from 2-4 inch shoots. In this experiment, it returned in February, some 2 months earlier than expected. In order to return 2 months early, the shoots would have to be 4 inches longer than normal at the time first crop is cut or the growth response of these shoots is great.

Part II.

Plants were fumigated with combustion products from natural gas high in sulfur content to determine sensitivity and detrimental effects on various plants including the carnation and chrysanthemum.

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Stages of development and varietal symptomology were also studied.

Early in the study, three plants were selected for their symptomology and growth characteristics to be used as indicator plants: petunia, tomato, and snapdragon. A graphic representation of relative sensitivity appears in Figure 2.

Plates 1-5 show typical symptoms of SO_2 damage on the carnation, chrysanthemum, and indicator plants. These symptoms were consistent throughout the study and agree with other researchers (1, 4, 32, 33, 35, 41, 42).

Plates 6-8 show 5 stages of development of the indicator plants and the corresponding damage at each stage. Sensitivity of tomato plants to SO_2 is reduced when the plant matures to the open flower stage, although it is quite sensitive throughout the activegrowing period. Petunia fails to respond to SO_2 when the plant is very young but it is very sensitive throughout the growing and reproductive stages. Snapdragon was insensitive to SO_2 at both ends of its growth cycle indicating a correlation between sensitivity and the theoretical growth-rate curve. Response to SO_2 in the snapdragon is in proportion to growth rate.

The threshold concentration of sulfur-contaminated atmosphere causing damage to the most sensitive plant tested, tomato 'Karglobe' was 0.5 grains sulfur per 100 cubic feet.

Whether this concentration or higher concentrations would ever occur in Colorado greenhouses is doubtful. This statement is based on data supplied by Public Service Company of Colorado. Their tests of total sulfur content of the main gas line showed 0.3 to 0.4 grains sulfur per 100 cubic feet.

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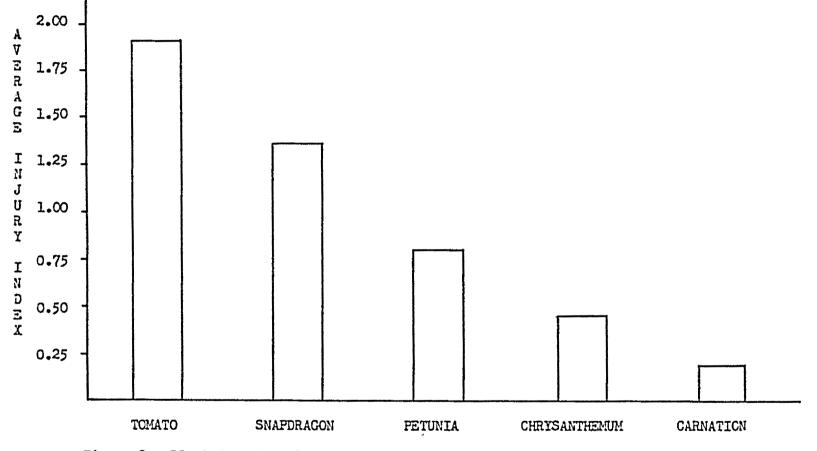


Figure 2. SO2 injury for plants based on average rating of twelve tests.

Plate 1. SO₂ injury on tomato 'Marglobe' showing slight damage from 0.5 grains sulfur per 100 cu. ft. (right), severe damage (left) and normal leaf (center).

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Plate 2. SO₂ injury on snapdragon 'Snowman' showing slight damage (right), severe damage (left) and normal leaf (center).



Plate 3. SO₂ injury on petunia 'White Magic' showing slight damage (right), severe damage (left) and normal leaf (center).

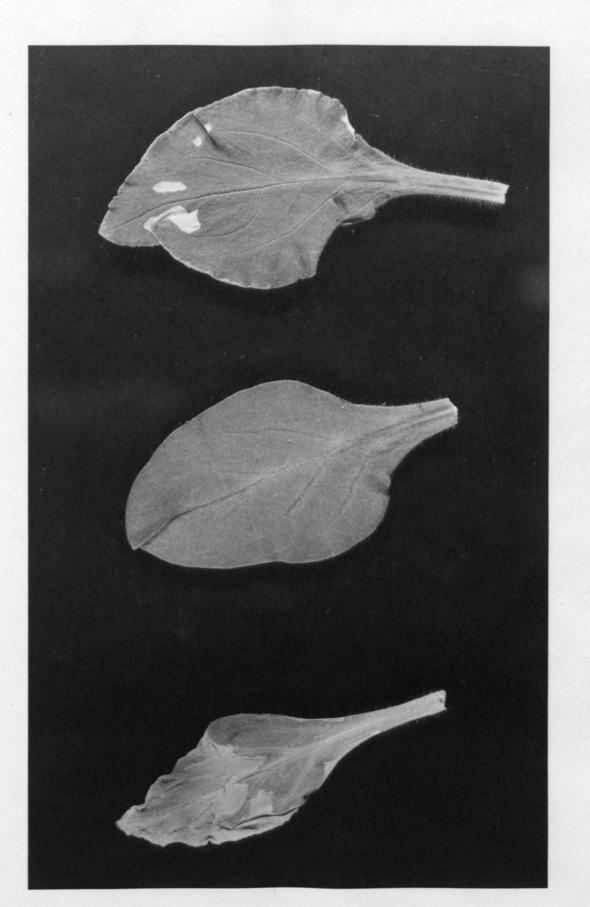


Plate 4. SO₂ injury on chrysanthemum 'Jupiter' showing slight damage (right), severe damage (left) and normal leaf (center).



Plate 5. SO₂ damage on carnation 'White Sim' showing slight damage from 1.4 grains sulfur per 100 cu. ft. (bottom), severe damage (top) and normal leaf (center).

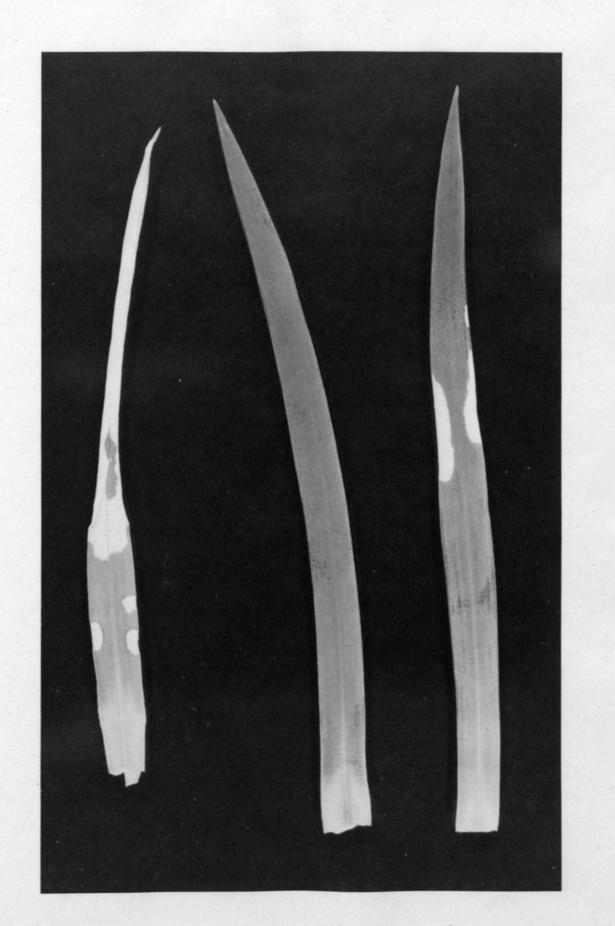


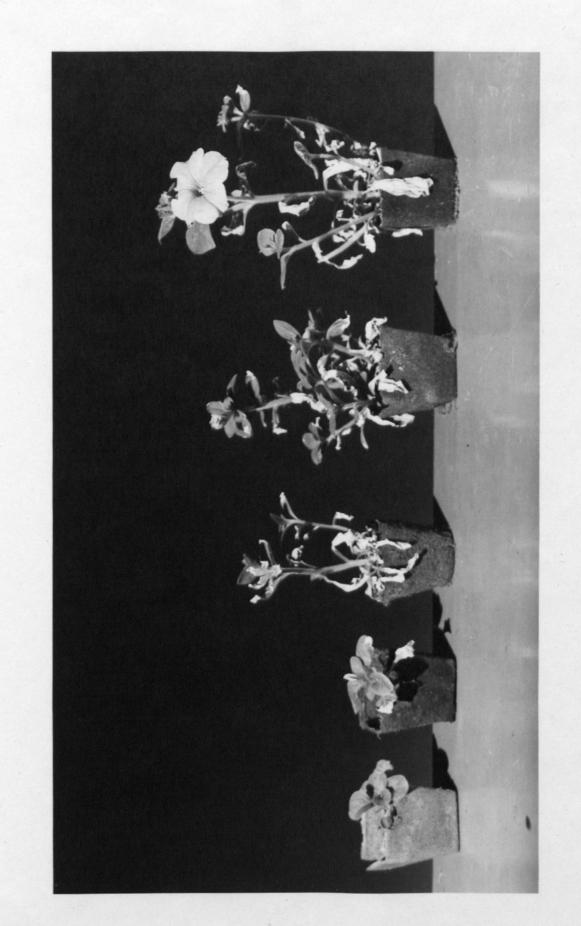
Plate 6. Tomato 'Marglobe' is most susceptible to SO₂ injury in rapidly growing stages, less in seedling stage.



Plate 7. Snapdragon 'Snowman' is most susceptible to SO₂ injury in rapidly growing stages, less in seedling stage.



Plate 8. Petunia 'White Magic' is most susceptible to SO₂ injury in rapidly growing stages, less in seedling stage.



Formulas that can be used to determine theoretical SO₂ potential for natural gas are:

> Kg = P K = constant; g = grains of total sulfur; P = ppm SO₂ potential.
> <u>b</u> = <u>1</u> b = ppm CO₂ at burner; a = ppm CO₂ above atmospheric; D = dilution factor.
> <u>1</u> = C <u>D</u>P D = dilution factor; P = ppm SO₂ potential; C = SO₂ contamination.

In our tests:

- 1) $16 \times 0.4 = 6.4 \text{ ppm } SO_2$.
- 2) 119,000/500 = 238 times.
- 3) $\frac{1}{238} \times 6.4 = 0.027.$

Since 0.5 grains sulfur per 100 cubic feet is required to damage tomato and 1.4 grains per 100 cubic feet is required to damage carnation, natural gas is a relatively safe source of CO_2 as long as the sulfur content does not exceed the above levels.

Plate 9 shows varietal differences to SO₂ injury in chrysanthemum varieties, Jupiter (sensitive) and Princess Ann (resistant).

Since varietal differences were noticed in chrysanthemum, tests were run to determine if varietal differences would be evident in other species such as tomato. A range of growth stages for the 3 varieties: Marglobe, Manapal, and Fireball were tested and replicated but no differences were evident. Since the tomato is a more sensitive species than chrysanthemum, perhaps the varietal differences, if any, were in amplitude below detection. Plate 9. Chrysanthemum 'Jupiter' (left) and 'Princess Anne' (right) showing varietal differences to SO₂ injury.



Need for further study

Additional work in the following areas should be done:

l) A detailed investigation of the effects of $\rm CO_2$ on rate of growth of carnations.

2) The effects of high CO₂ atmospheres on other environmental requirements, particularly nutrition, water and temperature.

3) The effects of sublethal concentrations of SO_2 on carnation.

4) The effects on carnation of other pollutants from fuel combustion, such as oxides of nitrogen, alkenes, and unsaturated hy-drocarbons.

Chapter V

SUMMARY

The effects of high levels of CO₂ (1200 ppm) on carnation were compared to 600 ppm and atmospheric (300 ppm). Data were collected from July, 1964 to June, 1965. The results were:

a. 1200 ppm increased yield 4 percent compared to 300 ppm, while 600 ppm increased yield 12 percent over 300 ppm.

b. Mean grade of first-year plants was significantly increased by 1200 ppm compared to 300 ppm. Differences in mean grade between 1200 ppm and 600 ppm, or 600 ppm and 300 ppm were not significant.

c. 1200 ppm CO₂ accelerated the second crop on Elliott's White Sim by 2 months when compared to 600 ppm or the control.

Five species of plants were fumigated with the combustion products of natural gas. Metered amounts of mercaptan were added to the gas to control concentrations of SO_2 in the fumigation chamber. The results of these experiments were:

a. Typical easily identifiable damage symptoms occurred on all plants if SO₂ concentration was sufficient.

b. Plants were more sensitive to SO₂ during their most rapid growth periods.

c. Species differ in their sensitivity to SO₂. A gradient response was found from the most sensitive plant, tomato, to snapdragon, petunia, chrysanthemum, and the least sensitive carnation.

d. Varieties of chrysanthemum differ in their response to SO2. Jupiter was found more sensitive than Pink Princess Ann.

e. Natural gas containing less than 0.5 grains sulfur per 100 cubic feet was a safe source of $\rm CO_2$ when concentrations to 840 ppm were used.

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APPENDIX

Carbon dioxide	Grade	Total production	% of control	% of distributior of grade
1200 ppm	Design	55	-38	4
	Short	60	-22	5
	Standard	792	+ 3	65
	Fancy	316	+21	26
	Total	1223	+ 4	100
600 ppm	Design	<u>9</u> 0	+ 9	7
	Short	86	+18	6
	Standard	800	+ 3	59
	Fancy	377	+1;1;	28
	Total	1353	+14	100
300 ppm	Design	76	0	7
	Short	73	0	6
	Standard	774	0	65
	Fancy	263	0	22
	Total	1186	0	100

APPENDIX A.	TOTAL PRODUCTION OF FLOWERS AT THREE LEVELS OF CO2 SHOW-
	ING PERCENT OF CONTROL AND DISTRIBUTION OF GRADE FROM
	JUNE, 1964-JUNE, 1965. VARIETY (Elliott's White Sim).

Carbon dioxide	Grade	Total production	% of control	% of distribution of grade
1200 ppm	Design	73	-32	9
	Short	57	-14	7
	Standard	386	- 3	48
	Fancy	285	+ 4	36
	Total	801	- 3	100
600 ppm	Design	71	31	7
	Short	85	+31	9
	Standard	414	+ 4	42
	Fancy	408	+49	42
	Total	978	+18	100
300 ppm	Design	96	0	11
	Short	65	0	8
	Standard	399	0	48
	Fancy	273	0	33
	Total	833	0	100

APPENDIX B. TOTAL PRODUCTION OF FLOWERS AT THREE LEVELS OF CO₂ SHOW-ING PERCENT OF CONTROL AND DISTRIEUTION OF GRADE FROM JUNE, 1964-JUNE, 1965. VARIETY (Pink Coquette).

Carbon dioxide	Grade	Total production	% of control	% of distribution of grade
1200 ppm	Design	65	+10	4
	Short	243	+21	17
	Standard	983	+ 1	67
	Fancy	175	-69	12
	Total	1466	- 3	100
600 ppm	Design	83	+41	5
	Short	197	- 2	13
	Standard	1020	+ 5	64
	Fancy	287	- 3	18
	Total	1587	+ 4	100
300 ppm	Design	59	o	4
	Short	201	0	13
	Standard	972	0	64
	Fancy	295	0	19
	Total	1527	0	100

APPENDIX C. TOTAL PRODUCTION OF FLOWERS AT THREE LEVELS OF CO₂ SHOW-ING PERCENT OF CONTROL AND DISTRIBUTION OF GRADE FROM JUNE, 1964-JUNE, 1965. VARIETY (Safari).

Carbon dioxide	Grade	Total production	え of control	% of distribution of grade
1200 ppm	Design	136	-23	10
	Short	438	+45	35
	Standard	587	-18	45
	Fancy	134	- 4	10
	Total	1295	- 1	100
600 ppm	Design	139	-20	10
	Short	315	+ 4	22
	Standard	822	+18	58
	Fancy	154	+10	10
	Total	1430	+10	100
300 ppm	Design	167	0	13
	Short	303	0	23
	Standard	694	0	53
	Fancy	140	0	11
	Total	1304	0	100

APPENDIX D. TOTAL PRODUCTION OF FLOWERS AT THREE LEVELS OF CO₂ SHOW-ING PERCENT OF CONTROL AND DISTRIBUTION OF GRADE FROM JUNE, 1964-JUNE, 1965. VARIETY (White No. 88).

1200 ppm CO2 production	600 ppm CO ₂ production	300 ppm CO2 production
185	166	146
125	168	139
84	111	79
187	195	146
297	174	162
94	153	135
70	155	145
109	155	148
78	75	85
	production 185 125 84 187 297 94 70 109	production production 185 166 125 168 84 111 187 195 297 174 94 153 70 155 109 155

APPENDIX E. MONTHLY DISTRIEUTION OF PRODUCTION OF ELLIOTT'S WHITE SIM FOR THREE CO2 LEVELS.

	1200 ppm CO ₂		600 ppm CO ₂		300 ppm CO2	
Month	Elliott's White	Pink Coquette	Elliott's White	Pink Coquette	Elliott's White	Pink Coquette
Nov. 2, 1964	3.91	3.92	3.85	3.90	3.80	3.92
Nov. 30, 1964	3.78	3.79	3.40	3.87	3.76	3.65
Dec. 28, 1964	3.81	3.36	3.92	2.60	3.62	3.05
Jan. 25, 1965	4.10	4.11	3.86	3.83	3.77	3.09
Feb. 22, 1965	4.03	4.41	4.01	4.37	3.78	4.03
Mar. 22, 1965	4.20	4.21	4.16	4.09	4.00	4.24
Apr. 19, 1965	4.66	4.67	4.65	4.46	4.48	4.47
May 17, 1965	4.93	4.43	4.87	4.78	4.86	4.65
Average per CO2 level	4.]	4	4.0)L	3.9	95

APPENDIX F. MEAN GRADE OF FIRST-YEAR PLANTS UNDER THREE LEVELS OF CO2.

		In	jury index		
Test	Tomato	Snapdragon	Petunia	Mum	Carnation
l	2	0	1,0,0ª/	0	0
2	4	1,3 ^{a/}	1,1 ^{ª/}	0,0,0 ^{a/}	0
3	2	l	l	0	0
4	3	0,2,2 ^{a/}	2	l	0
5	2	l	-	0	0
6	3	3	2	2	l
7	l	1,1,1 ^{ª/}	l	1	0
8	0,1 ^{ª/}	0	0	0	0
9	l	0	-	0	-
10	3	0	-	0	0
11	3	3,2 ^{ª/} 1,1,3 ^{ª/}	0,0 ^{a/}	l	l
12	1,1 ^{ª/}	1,1,3 ^{ª/}	1	0,2 ^{ª/}	0
Total	27	26	10	7	2
Number o plants	of	20	13	15	11
Average	1.93	1.30	•77	•47	.18

APPENDIX G. TWELVE TESTS SHOWING INJURY INDEX OF SO2 PLANT DAMAGE IN FUMIGATION CHAMBER.

 \underline{a} More than one plant in test.

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ABSTRACT OF THESIS

EFFECT OF SULFUR DIOXIDE AND HIGH LEVELS OF CARBON DIOXIDE ON CARNATIONS

Since it has been established that CO_2 concentration up to 550 ppm results in higher quality and yield of carnations, the question of additional benefit from higher levels arises. An experiment designed to compare yield and quality of carnations under three levels of CO_2 was executed. Three small greenhouses were used; and CO_2 levels were set at 1200 ppm, 600 ppm, and atmospheric (300 ppm). Records of yield and quality from July, 1964 to June, 1965 reveal the following effects:

a. 1200 ppm increased yield 4 percent compared to 300 ppm, while 600 ppm increased yield 12 percent over 300 ppm.

b. Mean grade of first-year plants was significantly increased by 1200 ppm compared to 300 ppm. Differences in mean grade between 1200 ppm and 600 ppm, or 600 ppm and 300 ppm were not significant.

c. 1200 ppm CO₂ accelerated the second crop on Elliott's White Sim by 2 months when compared to 600 ppm or the control.

Burning of natural gas appears to be an economical source of CO_2 ; however, since sulfur is a contaminant of natural gas and its combustion product, SO_2 , is injurious to some plants in very small concentrations, it is essential to detect SO_2 in the greenhouse before it reaches dangerous concentration.

Therefore, an experiment was designed in which five species of plants were fumigated with the combustion products of natural gas. Metered amounts of mercaptan were added to the gas to control concentrations of SO_2 in the fumigation chamber. A visible injury index was used to compare plants in their sensitivity to SO_2 . The results of these experiments were:

a. Typical easily identifiable damage symptoms occurred on all plants if SO₂ concentration was sufficient.

b. Plants were more sensitive to SO₂ during their most rapid growth periods.

c. Species differ in their sensitivity to SO_2 . A gradient response was found from the most sensitive plant, tomato, to snap-dragon, petunia, chrysanthemum, and the least sensitive carnation.

d. Varieties of chrysanthemum differ in their response to SO₂. Jupiter was found more sensitive than Pink Princess Ann.

e. Natural gas containing less than 0.5 grains of sulfur per 100 cubic feet was a safe source of CO_2 when concentrations to 840 ppm were used.

Roderick J. McKeag Department of Horticulture Colorado State University August, 1965