

C. Z.
LIBRARY
AIR RESOURCES BOARD
P. O. BOX 2815
SACRAMENTO, CA 95812

THE INTERACTION OF LOW LEVELS OF OZONE AND
RELATIVE HUMIDITY ON LEAFY VEGETABLES

Final Report
to the
California Air Resources Board
Research Division

under
Agreement No. A6-194-30

J. P. Bennett
Principal Investigator
Dept. of Vegetable Crops
University of California
Davis, CA 95616

March 6, 1979

LIBRARY
AIR RESOURCES BOARD
P. O. BOX 2815
SACRAMENTO, CA 95812

ABSTRACT

The effects of O_3 on the yield of crops are dependent on the conditions of the environment where the crop is grown. Varying levels of humidity are known to affect the development of visible injury produced by O_3 but nothing is known about how O_3 and humidity affect yield. The purpose of this study was to determine the effects of varying levels of O_3 and humidity on the yields of leaf lettuce and spinach.

The crops were grown in controlled environment chambers and fumigated 6 hours daily with 0, .08 and .18 ppm O_3 at 30, 50 and 80% relative humidity (RH) for 32-35 days. Results indicated that spinach yield losses due to O_3 were greater at 50% than at 80% RH while lettuce yield losses followed the opposite pattern. This means that generalizations about yield losses in more humid vs. dry environments are not possible because responses are crop specific. Lettuce plants were also significantly smaller and more tender, rendering them unmarketable regardless of the weight loss. This occurred at a concentration below the California one-hour O_3 standard of .10 ppm and suggests that the standard does not adequately protect this crop. Yield losses in both crops were not correlated with visible injury symptoms and even occurred without injury in the case of lettuce.

This report was submitted in fulfillment of Agreement No. A6-194-30 by J. P. Bennett under the sponsorship of the California Air Resources Board. Work was completed as of 15 October 1978.

TABLE OF CONTENTS

	Page
Abstract	1
List of Tables	3
Acknowledgments and Disclaimer	4
Conclusions and Recommendations	5
Introduction	6
Materials and Methods	8
Results	14
Discussion	24
References	27

LIST OF TABLES

	Page
Table 1. Schedules of experimental runs.	13
2. Ozone values for each experimental run.	15
3. Light values for each experimental value.	16
4. Day and night temperatures for each experimental run.	17
5. Effects of O_3 and RH on lettuce.	18
6. Effects of O_3 and RH on spinach.	19
7. Effects of O_3 and RH on chlorophyll in lettuce.	22
8. Effects of O_3 and RH on chlorophyll in spinach.	23

ACKNOWLEDGMENTS

The execution and completion of this project would not have been successful without the diligent assistance of P. Taillon, K. Barnes and K. Hatridge-Esh, J. Drager, J. Gafney, C. Lindquist, and V. Ventre, who all helped in the monitoring of the experiment and the harvests. I am also indebted to D. Paige for designing special equipment and modifying the chambers, and L. Thorpe and T. Long for developing the software for analyzing the chamber data. I am also grateful to Bob Reynolds of the ARB for his patience and time spent in monitoring our work.

DISCLAIMER

"The statements and conclusions in this report are those of the contractor and not necessarily those of the California Air Resources Board. The mention of commercial products, their source or their use in connection with material reported herein is not to be construed as either an actual or implied endorsement of such products."

CONCLUSIONS AND RECOMMENDATIONS

1. Lettuce plants fumigated with O_3 were significantly smaller and more tender, rendering them unmarketable, regardless of a weight loss and without visible injury symptoms. This occurred at average one-hour O_3 concentrations of .08 and .18 ppm for 6 hours daily for 32-35 days. The first concentration is below the California one-hour O_3 standard of .10 ppm. Thus the standard does not protect this crop from significant yield losses if the same O_3 concentrations and humidity conditions were to occur in the field.
2. Under growing conditions favorable for lettuce, spinach grew poorly and showed a greater O_3 response. Therefore air pollution effects studies should be done under optimal conditions for the crop. In addition, it is evident that unfavorable growth conditions make plants more susceptible to O_3 .
3. Spinach yield losses due to O_3 were greater at 50% RH than at 80% RH while lettuce yield losses followed the opposite pattern. If this interaction is real and not confounded by conclusion No. 1 above, then generalizations about losses in more humid versus dry environments are not possible because responses are crop specific. Thus lettuce growers in more humid areas and spinach growers in drier areas may suffer higher yield losses due to O_3 .
4. A significant increase in leaf number and area occurred only in lettuce fumigated with .08 ppm O_3 at 50% RH. The lack of a similar response in spinach makes these results ambiguous. Further work is needed on this problem.
5. If we assume that senescent leaves are the only visible manifestation of O_3 injury, there does not appear to be any correlation between percent senescent leaves and dry weight losses in either crop. In lettuce, a tolerant crop using injury as a criterion, there was no effect of O_3 on senescent leaves yet yield losses ranged from 25 to 43% depending on humidity. In spinach, percent senescent leaves increased 3.6 to 47 times due to O_3 yet yield losses ranged from 16 to 54%. Therefore, it does not seem reasonable to assume that if no visible injury occurs on a plant there will be no effect on yield. Yield losses can occur with or without any visible symptoms of O_3 injury.

INTRODUCTION

Statement of problem. High levels of ozone (O_3) are known to adversely affect the growth of plants by causing visible injury symptoms on the leaves. These effects are modified by interactions with other environmental factors, one of the most important of which is relative humidity (RH). It is generally accepted that given the same level of O_3 , a high RH will cause more injury than a low RH. Two problems in the $O_3 \times RH$ interaction have never been examined. These are the interaction at low levels of O_3 which do not cause visible injury and the effect on economic and biological yield subsequent to the fumigation. This report addresses these two problems.

Objectives. The experiments and analyses described herein were designed to determine:

1. The effect of varying levels of O_3 and RH on the yield of leaf lettuce and spinach.
2. The appearance of leaf injury and mortality under varying levels of RH and O_3 for the same species.

A more general objective was to test the assumption that California crops are less likely to be damaged by O_3 than crops in other parts of the country because of prevailing lower levels of RH.

Previous work on RH and lettuce and spinach. Heggstad and Heck (1971) have reviewed the effects of RH on plant response to air pollutants. Injury generally increases with increasing humidity in many species, but no studies of lettuce or spinach have been made. Leone and Brennan (1969) have published results of the most comprehensive O_3 and RH experiments on injury but unfortunately they did not quantify their results. Low light levels (~ 0.1 full sunlight, typical of many growth chambers) will also increase the $O_3 \times RH$ interaction effect on injury (Dunning and Heck, 1973). The sensitivity of the cultivar to O_3 is also important in $O_3 \times RH$ interaction studies and must be known before starting a study (Ting and Heath, 1975). Another problem is the period of RH change relative to O_3 exposure. Studies of pre-, during, and post-exposure RH changes have produced ambiguous results (Davis and Wood 1973, Dunning and Heck, 1973) and may not be realistic. No controlled studies have been done of long-term O_3 exposures with varying RH on total growth of a plant. Oshima et al. (1976 and

1977) concluded from their gradient analysis in the field that the influence of RH on the O_3 sensitivity of alfalfa and tomatoes was far less significant than the magnitude of the O_3 dose above. Another model relating tobacco weather flecking to environmental weather factors was unsuccessful at introducing RH into the relationship (Mukammal, 1965). The conclusion one reaches at this time is that although it appears that high humidity increases O_3 injury due to high O_3 doses on some species, there is no well-designed research that has shown whether or not this affects yield under low level ambient concentrations.

The sensitivity of lettuce to high levels of O_3 has been studied. Several kinds of head and leaf varieties ranged from 46 to 10% leaf injury after exposure to .70 ppm/1.5 hr at 27°C (which is too warm for lettuce) and Dark Green Boston was the most sensitive (Reinert et al., 1972). Oshima (1974) observed leaf injury, a 9% reduction in head size, no effect on dry weight, and a reduction in fresh weight due to dead leaves in the same variety after exposure to 0.15 and 0.25 ppm for 6% of the total growing period. Two head varieties showed 43-49% reduction in head weight by ambient air in Riverside, California (Thompson, 1975). Millecan (1976) has observed that Romaine lettuce is no longer grown in Los Angeles County due to air pollution. Most of the crop is now grown in the Imperial and Salinas Valleys where air pollution is not a serious problem today. Given that the air quality in these valleys may degrade in the future, there is a possibility that this highly important crop could be seriously threatened. In addition, spinach is a more sensitive leaf crop, and is included in this study to check on the cross-species response to O_3 xRH interactions.

MATERIALS AND METHODS

Environmental Control

Three 15 ft² Western Environmental HL-14 plant growth chambers were used in rotation. The humidity control system, which originally used a wet bulb-dry bulb sensor, was modified to accept the output of a Vaisala HMP-13 Humidicap proportional capacitance humidity probe. For the 80% RH regime the heated water bath humidifier of the growth chamber was augmented by a centrifugal atomizer placed in the blower discharge air stream under the plant bed.

Ozone control was implemented in only two of the growth chambers. Two 1/4" teflon sample tubes per chamber (four in all) were run to four teflon solenoid valves feeding a common manifold which was connected to the sample inlet of a Dasibi Model 1003 PC Ozone meter. The four valves were opened sequentially by a custom built timing circuit with selectable dwell times of one, two, or four minutes. As each valve was opened in turn, the Dasibi's internap pump was connected to the corresponding tube and sampled the air at that location. A response time of about 30 sec. was observed for stabilization of the reading. The analog output of the Dasibi was connected to four National Semiconductor LM 395 sample and hold amplifiers, each associated with one sample tube. After a 30 second delay for each channel, the sample and hold was activated by the timing circuit until the end of the dwell period, at which time the voltage was held until the next acquisition cycle. This voltage was continuously available for data monitoring and channels 2 and 4 were also connected to controllers which compared the ozone-proportional voltage to an adjustable reference voltage. The controllers employed triac outputs to energize a Sanders Model 3 Ozonizer in each chamber when the ozone level fell below the set value. In addition, the triacs were each associated with two potentiometers which set a low threshold for "off" power, and a high threshold for "on" power for the ozonizer. In this way control oscillations were minimized by maintaining a constant low production of ozone while limiting the rate of production when the controller was on. This also provided some ozone concentration limits if the Dasibi or control circuiting malfunctioned. (See Fig. 1) Fumigations were set at 0.08 and 0.18 ppm for 6 hours every day starting at noon.

Data Monitoring

All environmental data was monitored and logged with a Autodata Nine data acquisition system which included a Techtran Model 8010 data cassette magnetic

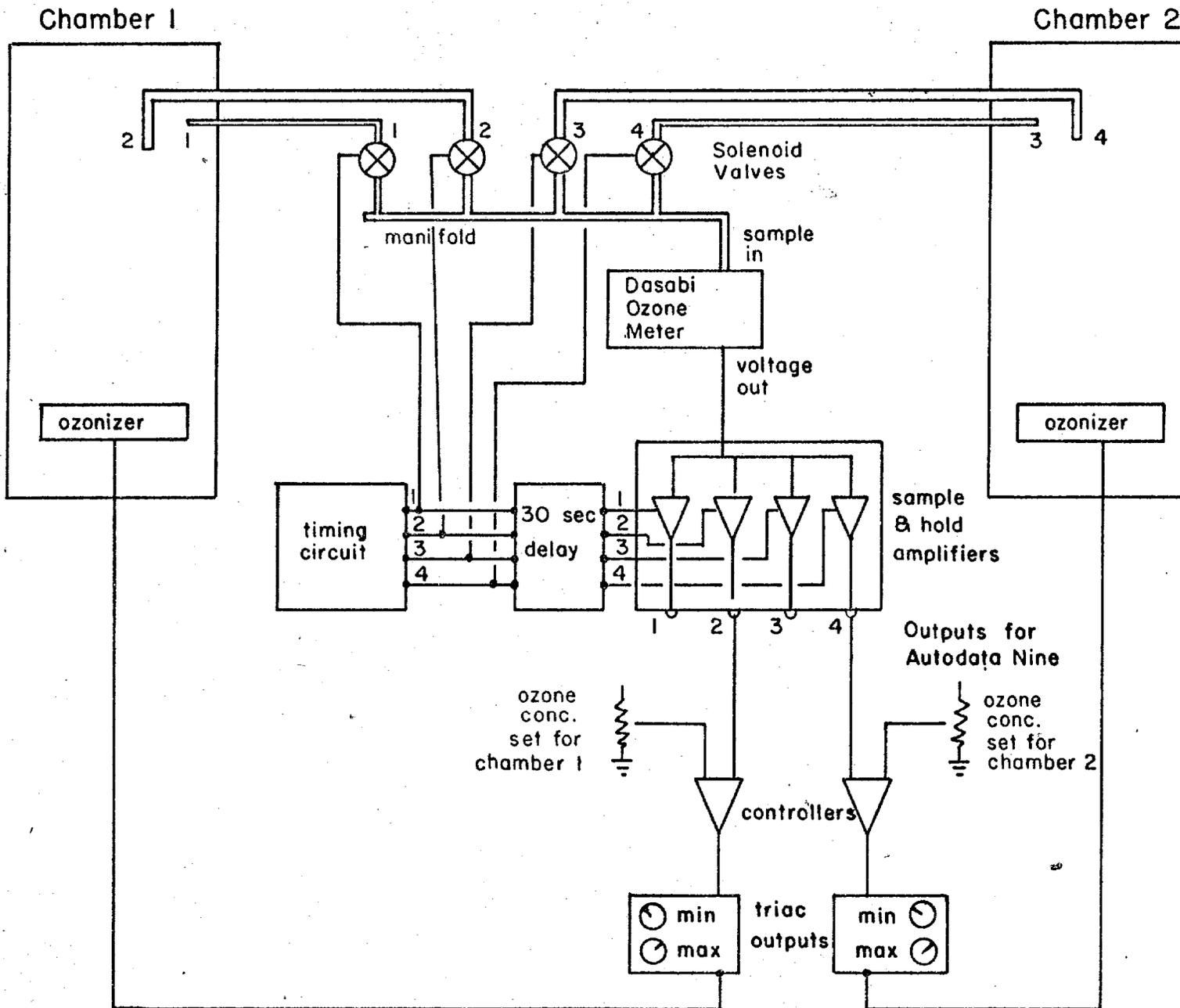


Figure 1. OZONE CONTROL SYSTEM

tape recorder for data storage. Each chamber was monitored continuously for humidity, temperature, light, and ozone. Values were logged every 30 minutes.

Humidity measurements were made using two Vaisala HMP-13 Humidicap probes per chamber housed separately in an aspirated enclosure placed at shelf level in the center of the chamber. Each probe sampled the air stream drawn in through 1/2" nominal white PVC pipe cut at the desired sampling height and fitted with an elbow covered with cheesecloth to exclude particulate matter and reduce the ozone concentration reaching the sensors (see Fig. 2). One probe output was connected to the chamber humidity control system, as detailed above and both probes were monitored by the Autodata Nine.

Temperature profiles were measured with six thermocouples in each chamber; one within the aspirated humidity probe enclosure, four at plant level near each end of the chamber, and the remaining one 8 cm above plant height. All temperatures were monitored and logged by the Autodata Nine.

Each chamber had a Lambda Quantum photon light sensor placed at plant height and shunted by a 200 ohm resistor to convert current to millivolts. Outputs were connected to the Autodata Nine.

As detailed above, the sample-and-hold amplifier associated with each ozone sampling tube stored and updated the ozone reading for that channel, and each voltage was monitored and logged separately by a channel of the Autodata Nine. Each of the two ozonated chambers had two sample tubes placed at different heights; one at plant level and one 18 cm above plant level.

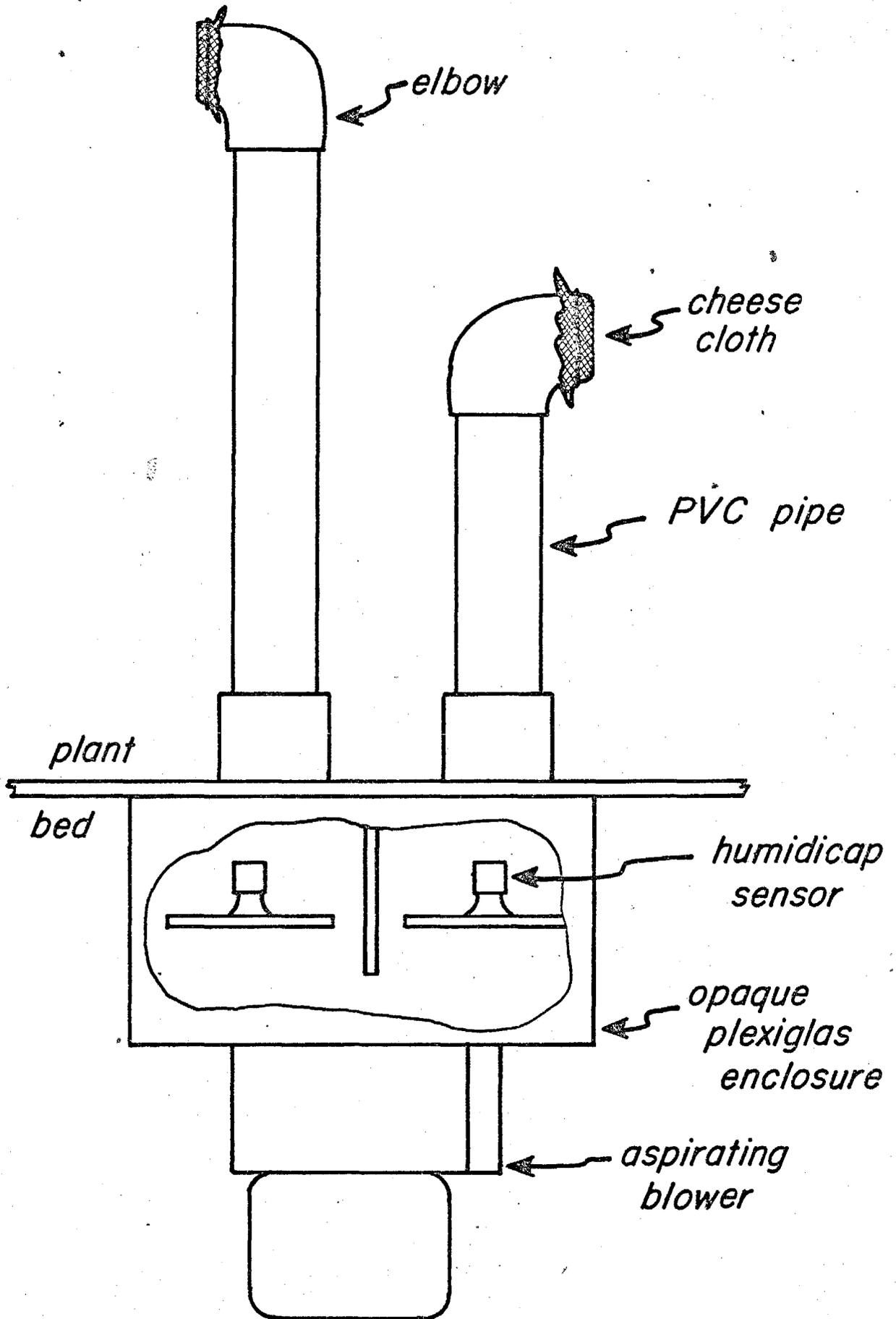
Cultural Practices

A standard UCD soil mix consisting of 1/3 Yolo sandy loam, 1/3 peat moss and 1/3 sand was used throughout the experiment. No fertilizer was mixed in with the soil. New soil mix was put into 2 gal. black nursery containers (8" diameter) a few days before each run. Four seeds of each species were planted in each of ten pots of each species in the growth chamber. Following emergence, plants were allowed to grow to the first true leaf stage* before thinning and the start of fumigation. Pots were kept at or near field capacity by periodic watering every 1 or 2 days with distilled water. Once a week the pots were watered with .50% Hoagland's solution. No pests or diseases were observed during any run.

The lettuce variety used was Dark Green Boston and the spinach variety was Bloomsdale. UCD Vegetable Crops Extension provided a single lot of seed of each variety which was stored under the proper conditions and used throughout the experiment

* "True" leaves are those that appear after the cotyledons emerge from the ground.

Figure 2. HUMIDITY PROBES



The planting and harvest dates of each run, along with their duration, are shown in Table 1. At each harvest, measurements were taken on leaf fresh weight (LFW), total leaf area (LA), stem length, number of senesced leaves, total number of leaves, and stem fresh weight. After drying in a forced-air oven for 48 hours @ 65°C, leaf, stem and root dry weights were recorded. Senesced leaves were not included in measuring LA, which was done on an electronic planimeter. Senesced leaf dry weight was recorded separately in Runs 2 and 3.

Lettuce specialists Ted Welch, Vincent Rubatzky, Leonard Morris and Charles Cheyney observed two harvests and made observations on the marketability of the plants. Their comments are incorporated into the results and discussion.

Two leaves from each plant were stored at -30°C after weighing fresh and analyzed later for chlorophyll content using acetone extraction according to the methods of Yoshida et al. (1972). Measurements included the two most important chlorophylls, a and b, total chlorophyll and the a/b ratio. The chlorophyll analyses were not originally planned or funded as part of the research contract.

Personnel problems and technical difficulties in a fourth run of the experiment, as described in an earlier progress report, caused anomalous results which were excluded from the analyses and discussion. In addition, a tape recorder failure in Run 3 forced data acquisition to digital printing on paper mode. This data has not been analyzed due to a personnel shortage. Spot checks of environmental parameters in the chamber indicated they were functioning satisfactorily.

Data were analyzed by means of 2 x 3 analyses of variances and Duncan's multiple range tests, assuming there were no chamber differences. The latter tests are designed to determine how many subsets of means in a group of means are not significantly different at a given level of probability. Means in each subset are labelled with the same letter to better distinguish them from other subset means. The test is not perfect and it is not always able to determine which subset a particular mean belongs to and is then labelled with two or more letters in the tables.

TABLE 1. Schedules of experimental runs

Dates	<u>Run</u>		
	1	2	3
Planting	14 Nov.	10 Jan.	13 March
Emergence	22 Nov.	14-17 Jan.	22 March
O ₃ begins	10 Dec.	30 Jan.	3 April
Fertilizer	28 Dec. NH ₄ (SO ₄) ₂	25 Feb.	no record
Harvest	11 Jan.	6 March	8 May
days, planting/ harvest	58	55	56
days, O ₃ /harv.	32	35	35

RESULTS

Growth Chambers

The overall mean, standard deviation, and lowest and highest single, instantaneous ozone concentrations for each chamber and each run are shown in Table 2. These values are calculated from points that were extracted from digitizing the strip chart that ran continuously recording the O₃ analyzer output. The 0.08 ppm O₃ treatment was off only in Run 2 where it was low by 0.02 ppm. The 0.18 ppm O₃ treatment was also low in the same run. Standard deviations averaged about 0.020 ppm in the low treatment and about 0.035 ppm in the high treatment.

Table 3 shows the average light values for each chamber in Runs 1 and 2. These did not deviate significantly from the specified range of 500-550 $\mu\text{E m}^{-2}\text{s}^{-1}$. Average temperatures shown in Table 4 were also significantly close to the set objectives of 20° day/12° night. Standard deviations averaged 1-2° except for Chamber 7, Run 2, which reached 2-3°C.

Precise humidity data from the Autodata 9 was not recorded. It was found that calibration of the probes drifted constantly, possibly due to the O₃ treatments. Spot checks were performed daily, weekly and monthly on all components of the system, and usually indicated proper performance. We also knew intuitively that it was impossible to have a 30% humidity regime when the humidifiers were on and vice versa. The greatest difficulty in regulating humidity occurred in the first run, which was set for 30% RH. Control was better in the 50% and 80% RH runs which followed. The degree of uncertainty about the set humidity levels for the latter runs is no greater than 5% RH, based on means from some of the data logger channels which appeared stable. In view of this outcome, the biological results of the 30% RH run will be tabulated in this report, but no further comment or discussion will be made about them. In addition, they have been excluded from the analyses of variance.

The biological yield data for the remaining eight treatments for lettuce and spinach are shown in Tables 5 and 6 respectively.

Shoot Fresh Weight. Both species responded to treatment with a significant O₃ x RH interaction. While lettuce yields without O₃ were the same at 50 and 80% RH, those at 80% were decreased significantly by both .08 and .18 ppm O₃

TABLE 2. Ozone values for each experimental run.

Run	O ₃	Mean (ppm)	Standard Deviation	Min.*	Max.*	Chamber
1	Low	.080	.019	.000	.132	5
	High	.179	.031	.003	.282	6
2	Low	.062	.018	.015	.115	6
	High	.134	.040	.037	.222	5
3	Low	.081	.024	.003	.187	5
	High	.173	.044	.015	.343	6

* Single, instantaneous observations.

TABLE 3. Light values for each experimental value

Run	Chamber	Mean $\mu\text{Em}^{-2}\text{s}^{-1}$	Standard Deviation	Min.*	Max.*
1	5	513.2	14.0	431.5	546.4
	6	534.6	15.8	452.2	595.6
2	5	517.3	18.0	462.3	553.1
	6	540.2	47.4	508.4	608.8
	7	511.0	16.8	456.7	550.1

* Single, instantaneous observations

TABLE 4. Day and night temperatures for each experimental run

Run	Chamber	<u>DAY</u>			
		Mean (°C)	Standard Deviation	Min.*	Max.*
1	5	18.7	1.69	7.6	21.4
	6	20.1	1.35	11.2	23.9
2	5	19.6	0.59	18.2	21.4
	6	20.9	0.97	16.5	23.6
	7	20.2	3.23	0.9	47.2
<u>NIGHT</u>					
1	5	12.1	1.52	8.5	19.6
	6	11.4	2.15	7.8	21.8
2	5	12.6	0.66	11.6	19.2
	6	12.3	1.36	10.3	33.0
	7	11.5	2.41	6.0	17.0

* Single, instantaneous observations

TABLE 5. Effects of O₃ and RH on Lettuce

RH (%)	O ₃ (ppm)	Shoot fresh weight (g)	Root dry weight (g)	Shoot dry weight (g)	Total dry weight (g)	Leaf area (cm ²)	Number of leaves	Percent senesced leaves
30	.08	106.0	1.23	10.4	11.6	2577	51.5	10.8
	.18	90.3	0.94	8.3	9.2	2271	47.3	11.7
50	0	169.7x	2.76y	13.2y	16.0y	3664y	54.8z	10.9z
	.08	188.3x	1.45z	13.8y	15.2y	5044x	68.8xy	8.7z
	.18	127.0y	1.38z	10.5z	11.9z	3544y	51.5z	11.4z
80	0	174.1x	1.91w	15.0y	16.9y	3948y	77.3x	10.2z
	.08	122.0y	1.24x	10.8z	12.0z	2882z	60.2yz	10.0z
	.18	98.5z	0.97x	8.9z	9.9z	2567z	59.5yz	9.7z
Coefficient of variation		18.9	30.0	17.0	16.9	16.4	19.6	26.5

Means followed by different letters are significantly different at the .05 probability level using Duncan's multiple range test. Means for the 30% RH treatment were not included in the analyses (see p. 14). Each number is the mean of ten plants.

TABLE 6. Effects of O₃ and RH on Spinach

RH (%)	O ₃ (ppm)	Shoot fresh weight (g)	Root dry weight (g)	Shoot dry weight (g)	Total dry weight (g)	Leaf area (cm ²)	Number of leaves	Percent senesced leaves
30	.08	27.2	1.42	3.27	4.68	361	35.0	5.0
	.18	19.1	1.28	2.78	4.06	287	36.5	10.1
50	0	56.7v	3.27x	6.38w	9.65w	695v	59.1y	0.2z
	.08	39.1w	1.19y	3.93x	5.12x	528w	35.3z	3.4y
	.18	26.2x	0.94yz	3.08y	4.02y	395x	30.3z	9.4x
80	0	23.6xy	1.18y	2.99y	4.17y	323xy	42.1y	5.5z
	.08	19.8y	1.05y	2.86y	3.91y	298y	38.5z	9.6y
	.18	12.8z	0.62z	2.05z	2.67z	206z	30.5z	19.9x
Coefficient of variation		19.9	27.9	19.3	18.5	20.4	36.6	53.8

Means followed by different letters are significantly different at the .05 probability level using Duncan's multiple range test. Means for the 30% RH treatment were not included in the analyses (see P. 14). Each number is the mean of ten plants.

(-30 and -43% respectively) but those at 50% were reduced significantly only by the .18 ppm O₃ (-25%). Spinach yields were significantly reduced by both O₃ treatments at both 50 and 80% RH, but yield losses at 50% RH were greater than at 80% RH, the opposite of the lettuce. Overall, spinach yields at 80% RH were about half of the yields at 50% RH.

Root Dry Weight. At 50% RH, spinach roots at .18 ppm O₃ were 71% less than the controls while lettuce roots were 50% less in the same treatment. At 80% RH, lettuce roots were decreased by O₃ about the same amount as at 50% RH. Spinach roots at 80% RH, however, were only 11% less at .08 ppm O₃ but 44% less at .18 ppm O₃. Root weights were comparable between the two species and the overall humidity effect was to cut growth by about 50%.

Shoot Dry Weight. A clear interaction occurred with this variable. Lettuce shoots grown at 50% RH showed no O₃ effect until the .18 ppm treatment (a 20% decrease) but at 80% RH, where the control yield was higher, both O₃ treatments significantly reduced dry weights 28 and 41% respectively. Spinach shoots did the reverse: yields were higher at 50% RH and both the O₃ treatments reduced yields 38 and 52% respectively. At 80% RH, however, only the .18 ppm treatment had a significant effect (a 31% decrease).

Total Dry Weight. The pattern of response for this variable followed that of shoot dry weight, reflecting the fact that shoot dry weight accounted for an average of 88% and spinach 74% of the total dry weight. The pattern of O₃ effects followed that of total fresh weight.

Leaf area. At 50% RH, lettuce leaves were significantly larger at .08 ppm O₃ compared to the control, but not affected by .18 ppm O₃. At 80% RH, both O₃ levels decreased leaf area a comparable amount, even though humidity did not significantly change leaf area in the non-fumigated treatments. Spinach leaves, which were much smaller than the lettuce leaves, were more affected by O₃ at 50% RH than at 80% RH, a pattern following the dry weight responses. No increases in leaf area were observed in any O₃ treatment. Both O₃ levels at both humidity treatments significantly reduced leaf area.

Number of leaves. On average there were 14 more leaves on the lettuce plants fumigated at .08 ppm O₃, 50% RH compared to the control plants. Leaf number at .08 ppm O₃ was not significantly reduced. At 80% RH, both O₃ treatments reduced leaf number 22-23% and more leaves were produced at 80% than at 50% RH. Spinach leaf numbers in the O₃ treatments were similar in both humidities, but a greater number were produced at 50% RH without O₃. Consequently, greater reductions were observed at 50% than at 80% RH.

Percent Senesced Leaves. Neither O_3 nor RH significantly affected leaf senescence in lettuce, but in spinach, senescence increased significantly at both O_3 levels in both humidity treatments, indicating no interaction. Many more senesced leaves were observed at 80% RH than at 50% RH.

Variability of Response. In lettuce, the coefficients of variation (CVs) ranged from 16.4 to 30.0 in the following ascending order: leaf area, total dry weight, shoot dry weight, shoot fresh weight, number of leaves, percent senesced leaves and root dry weight. Spinach CVs were higher and ranged from 18.5 to 53.8. In ascending order these were total dry weight, shoot dry weight, shoot fresh weight, leaf area, root dry weight, number of leaves, and percent senesced leaves.

Chlorophyll Content. Tables 7 and 8 show the effects of O_3 and RH on chlorophyll a, b, total chlorophyll and the a/b ratio for lettuce and spinach respectively. For lettuce significant effects were observed only for the a/b ratio. Both O_3 and humidity appeared to decrease the ratio and no interaction occurred. Humidity significantly affected the chlorophyll a, b and total content in spinach. The amounts were consistently lower at 50% RH than at 80%, but no interaction with O_3 appeared. No consistent O_3 pattern emerged for any chlorophyll variable for both species.

TABLE 7. Effects of O₃ and RH on Chlorophyll in Lettuce

RH	O ₃	Chlorophyll a	Chlorophyll b	Total Chlorophyll	a/b ratio
(mg/gm fresh weight)					
30	.08	.725	.277	1.002	2.595
	.18	.592	.226	.819	2.560
50	0	.660	.249	.910	2.632
	.08	.460	.196	.656	2.384
	.18	.625	.242	.867	2.576
80	0	.555	.219	.775	2.499
	.08	.566	.251	.816	2.245
	.18	.507	.217	.724	2.319
Significance O ₃ (.05)	RH	NS	NS	NS	.036
		NS	NS	NS	.026

NS = no significant differences

TABLE 8. Effects of O₃ and RH on Chlorophyll in Spinach

RH	O ₃	Chlorophyll a	Chlorophyll b	Total Chlorophyll	a/b ratio
(mg/gm fresh weight)					
30	.08	1.162	.430	1.593	2.698
	.18	.859	.352	1.211	2.428
50	0	.688	.262	.949	2.630
	.08	.620	.241	.861	2.579
	.18	.871	.328	1.199	2.645
80	0	.888	.350	1.238	2.535
	.08	1.066	.412	1.478	2.593
	.18	.948	.335	1.283	2.844
Significance O ₃		NS	NS	NS	NS
(.05) RH		< .001	< .001	< .001	NS

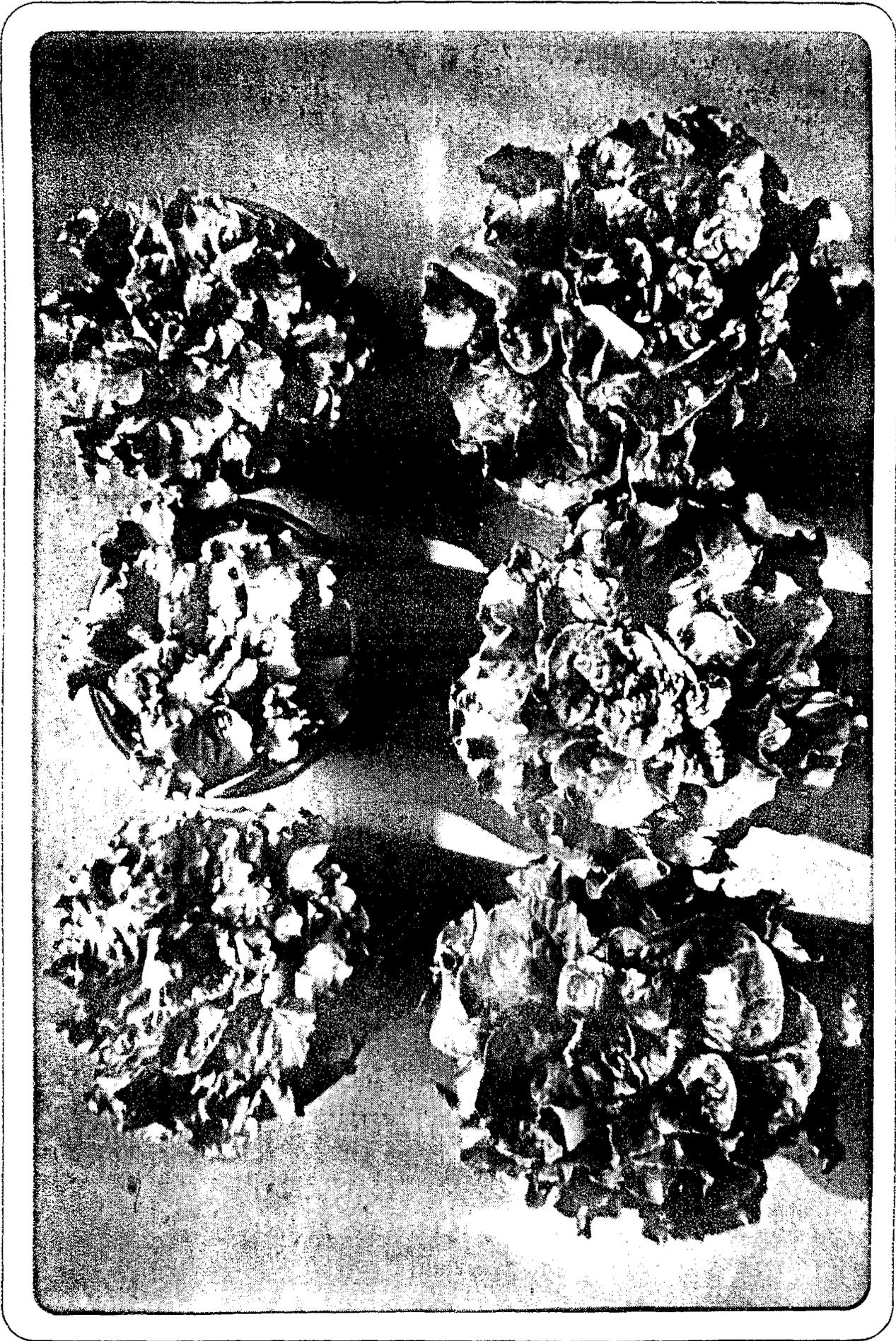


Fig. 3. Lettuce plant exposed to 0.08 ppm (lower row) and 0.18 ppm O_3 (upper row) at 50% RH.

that more plants will be needed to form a bunch if they are grown in areas where O_3 is present. In addition, the presence of senescent leaves and the paler green color of live leaves will detract from their marketability. The low yields of the spinach compared to the lettuce is probably due to the fact that the growing conditions were predetermined and set to match conditions ideal for lettuce growth. It was readily apparent that the spinach did not grow well under these conditions and were thus under additional stress.

Yields were highest at 50% RH and lowest at 30 and 80% RH. Spinach was more sensitive to O_3 at 50% RH rather than 80% RH, the opposite of the lettuce response. Most fresh market spinach is grown in Ventura Co., an area relatively drier than Monterey Co., where a great deal of the canning spinach is grown. Thus, fresh market growers in Ventura Co. are likely to suffer greater losses due to O_3 than growers in Monterey Co., assuming the same O_3 levels are found there. The only probable explanation for the response being the opposite of lettuce lies in using environmental conditions ideal for lettuce. No physiological reason can be put forth at this time.

REFERENCES

- Davis, D. D. and F. A. Wood. 1973. The influence of environmental factors on the sensitivity of virginia pine to ozone. *Phytopath.* 63: 371-376.
- Dunning, J. A. and W. W. Heck. 1973. Response of pinto bean and tobacco to ozone as conditioned by light intensity and/or humidity. *Environ. Sci. & Tech.* 7: 824-826.
- Heggstad, H. E. and W. W. Heck. 1971. Nature, extent and variation of plant response to air pollutants. *Adv. Agron.* 23: 111-145.
- Leone, I. A. and E. Brennan. 1969. The importance of moisture in ozone phytotoxicity. *Atmos. Environ.* 3: 399-406.
- Millecan, A. A. 1976. A survey and assessment of air pollution damage to California vegetation 1970 through 1974. Calif. Dept. of Food & Agric., Sacramento.
- Mukammal, D. D. 1965. Ozone as a cause of tobacco injury. *Agr. Meteorol.* 2: 145-165.
- Oshima, R. J. 1974. Final report to the California Air Resources Board. II. Yield study. IIA. Prototype ozone dosage-crop loss conversion function. Calif. Dept. of Food & Agric., Sacramento.
- Oshima, R. J. et al. 1976. Ozone dosage-crop loss function for alfalfa: a standardized method for assessing crop losses from air pollutants. *J. Air. Poll. Cont. Assoc.* 26: 861-865.
- Oshima, R. J. et al. 1977. Reduction of tomato fruit size and yield by ozone. *J. Amer. Soc. Hort. Sci.* 102: 289-293.
- Reinert, R. A., D. T. Tingey, and H. B. Carter. 1972. Ozone induced foliar injury in lettuce and radish cultivars. *J. Amer. Soc. Hort. Sci.* 97: 711-714.
- Thompson, C. R. 1975. Economic effects of smog injury to agricultural crops. Final report, CAL ARB Contract 2-650.
- Ting, I. P. and R. L. Heath. 1975. Responses of plants to air pollutant oxidants. *Adv. Agron.* 27: 89-121.
- Yoshida, S. et al. 1972. Laboratory Manual for Physiological Studies of Rice, Second Edition. Los Banos, Philippines: The International Rice Research Institute.