MORPHOLOGICAL RESPONSES AMONG CROP SPECIES TO FULL-SEASON EXPOSURES TO ENHANCED CONCENTRATIONS OF ATMOSPHERIC CO_2 AND O_3†

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Abstract: Field studies using open-top chambers were conducted at USDA-BARC involving the growth of soybeans ('89 & '90), wheat ('91 & '92), and corn ('91), under increased concentrations of atmospheric CO_2 and O_3. Treatment responses were compared in all cases to plants grown in charcoal-filtered (CF) air (seasonal 7-h mean = 25±3 n mol O_3 mol^{-1}) having 350 or 500 μ mol CO_2 mol^{-1}. Elevated seasonal O_3 levels for the soybean, wheat, and corn studies averaged 72.2±4, 62.7±2, and 70.2 n mol O_3 mol^{-1}, respectively. Results presented were obtained for plants grown in silt loam soil under well-watered conditions. Grain yield increases in response to elevated CO_2 in the absence of O_3 stress averaged 9.0, 12.0, and 1.0 % for soybean, wheat, and corn, respectively. Reductions in grain yields in response to the elevated O_3 treatments at 350 μ mol CO_2 mol^{-1} averaged 20.0, 29.0 and 13.0% for soybean, wheat, and corn, respectively. Reductions in grain yields in response to elevated O_3 at 500 μ mol CO_2 mol^{-1} averaged 20.0, 8.0, and 7.0% for soybean, wheat, and corn, respectively. Dry biomass and harvest index in wheat were significantly reduced by O_3 stress at 350 μ mol mol^{-1} CO_2 but not at 500 u mol mol^{-1} CO_2. Seed weight 1000^{-1} for soybeans and wheat was significantly increased by CO_2 enrichment and decreased by O_3 stress. Seed weight 1000^{-1} in corn was increased by O_3 stress suggesting that O_3 affected pollination resulting in fewer kernels per ear.

Key words: Global climate change, photochemical oxidants.

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1. Introduction

Progressive changes currently taking place in the earth's atmosphere regarding the increasing concentrations of gases such as CO_2, CH_4, O_3, N_2O, and chlorofluorocarbons (CFC's) have prompted concerns about future impacts such changes may have on the earth's food and fiber crops (Krupa and Kickert, 1989; Kimball, 1986; Kimball et al., 1990; Adams, et al., 1990; Allen, 1990; Mulchi, et al., 1992; and Stockle, et al., 1992). Factors being linked to changes in the atmospheric composition include: acidic deposition; shifts in the solar radiation spectra reaching the soil surface with particular emphasis on ultraviolet B; and increased ambient temperatures which will affect relative humidity, evapotranspiration, clouds and precipitation patterns (White, 1989; Kimball, et al., 1990).

Over the past century, atmospheric CO_2 levels have risen from approximately 270 μ mol CO_2 mol^{-1} during the preindustrial period of the late 1800's to about 355 μ mol CO_2 mol^{-1} in the mid 1990's. Based on the current rate of increase in CO_2 of 0.4% per year, the earth's atmosphere will likely double in CO_2 concentration by the middle of the 21st century. The major causes for the rapid rise in CO_2 concentrations include the burning of fossil fuels as a primary energy source in combination with deforestation practices (Burke and Lashof, 1990). Emissions from liquid fossil fuels are primary sources of VOC's (volatile organic compounds) and the burning of fossil fuels are major sources for

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NO_x, precursors which form O_3 in the presence of ultraviolet radiation. The depletion of O_3 in the stratosphere associated with the buildup of CFC's are partially being offset by the increase in tropospheric O_3 concentrations (Krupa and Kickert, 1989).

The beneficial effects of atmospheric CO_2 enrichment on crop productivity have been documented by numerous investigators as reviewed by Krupa and Kickert (1989). Among the positive effects from enhanced CO_2 concentrations on plants include: increased photosynthesis, leaf area, dry matter accumulation and yield in C3 crops; decreased stomatal conductance and increased water use efficiency in both C3 and C4 plants and increased specific leaf weight and crop maturation rate.

The effects of increased exposures to chronic O_3 air pollution on crop species have likewise been extensively examined since O_3 was identified as among the most damaging components in photochemical smog (Heggestad and Bennett, 1984; Heck, et al., 1988; Krupa and Kickert, 1989). Among the negative impacts on crops attributed to chronic O_3 exposure include: decreased photosynthesis, leaf area, dry matter production and yield. Crops sensitive to O_3 stress exhibit increased specific leaf weight,decreased stomatal conductance and water use efficiency. Crops typically become less sensitive to O_3 during periods of moisture stress but more sensitive to O_3 under mineral deficiency such as nitrogen (Krupa and Kickert, 1989).

Mulchi et al., (1992) grew soybeans (<u>Glycine</u> <u>max</u>. Merr.) full-season in open-top chambers in 1989 at 350, 400 or 500 μ mol CO_2 mol^{-1} under three air quality regimes: charcoal filtered (CF) air, nonfiltered (NF) air, and nonfiltered air + 40 n mol O_3 mol^{-1} ($NF+O_3$). They reported: a) leaf photosynthesis rates were stimulated by increased CO_2 concentration even in the presence of high O_3 exposure; b) plant biomass, pods per plant, and grain yields were likewise stimulated by increased CO_2 in the presence of high O_3 exposure; c) the negative impact of current ambient O_3 concentrations on growth and productivity of soybeans were largely counteracted by increasing the CO_2 concentrations by 150 μ mol CO_2 mol^{-1}; and d) the effect of enhanced CO_2 in combination with O_3 exposure on stomatal conductance appeared to be additive. The objectives of the present investigations were to examine the effects of increased atmospheric CO_2 and O_3 on winter wheat (<u>Triticum</u> <u>aestivum</u> L.) and corn (<u>Zea</u> <u>mays</u> L.). Also, results from a sequel soybean investigation in 1990 were combined with data from the 1989 study.

2. Materials and Methods

Open-top chambers (OTC's) described by Heagle, et al., (1973) were used in all investigations Table 1. The general procedures followed in all instances were similar to that described for the 1989 soybean study (Mulchi et al., 1992). The studies were carried out at the USDA Beltsville Agricultural Research Center (BARC) at Beltsville, Maryland on a Codorus silt loam soil. The soil was amended with fertilizers at rates recommended for the individual crops and pre- or post-emergence herbicides were applied to control weeds. Sprinkler irrigation units were utilized to maintain soil moisture levels near field capacity during periods of below normal rainfall amounts or distribution. Due to the high moisture holding capacity in the silt loam soil, no symptoms of moisture stress were observed on any of the plants. No irrigations were necessary during the two wheat studies (Rudorff, 1993).

2.1 Carbon Dioxide and Ozone Treatments.

The chambers were installed over the plots soon after seedling stands were insured for

TABLE 1

Summary of species, cultivars, and chamber air quality treatments from 1989-1992. USDA-BARC.

Year	Species	Cultivar	Design	Reps	Chamber Treatments
1989	Soybean	Clark	3 x 3 Factorial	2	350,400 & 500 μ mol CO_2 mol^{-1}; CF, NF & NF+40 n mol O_3 mol^{-1}
1990	Soybean	Clark	3 x 3 Factorial	2	350,400 & 500 μ mol CO_2 mol^{-1}; CF, NF & NF+40 n mol O_3 mol^{-1}
1991	Wheat	Massey	2 x 2	4	350 & 500 μ mol CO_2 mol^{-1}; NF & NF+40 n mol O_3 mol^{-1}
1992	Corn	Pioneer 3714	2 x 2 Factorial	3	350 & 500 μ mol CO_2 mol^{-1}; CF & NF+40 n mol O_3 mol^{-1}
1992	Wheat	Saluda	2 x 2 Factorial	4	350 & 500 μ mol CO_2 mol^{-1}; CF & NF+40 n mol O_3 mol^{-1}

the soybean and corn; however, the wheat plots were chambered in late March immediately following the application of ammonium nitrate at 60 kg ha^{-1} N prior to the beginning of spring growth. Gas treatments were initiated normally within 10 days following chamber installation and extended until physiological maturity. Carbon dioxide was injected into the blowers at rates sufficient to raise the chamber air concentrations to the levels listed in Table I for 12 h day^{-1} (0700-1800 h EST). The flow of CO_2 to the OTC's was metered through flowmeters and individual rates were checked daily.

Ozone was injected into blowers supplying air to the NF + O_3 treatments (Table I) only 5 day wk^{-1} for 7 h day^{-1} (1000-1600 h EST). No O_3 additions were made on weekends or during periods of rain; however, the plants in the NF + O_3 treatments were exposed to ambient O_3 concentrations during the weekends. The O_3 concen- trations in the high O_3 treatments were increased by an average of 40 n mol O_3 mol^{-1} above ambient levels; however, the maximum O_3 levels were limited to 120 n mol O_3 mol^{-1}, the current national secondary air quality standard for O_3 in the USA. During the daily fumigation periods, air samples were continuously collected from approximately 10 cm above the crop canopy using Teflon® tubes (6.4 mm O.D.) attached through 3-way solenoid valves to a central vacuum system. The O_3 levels in all chambers were monitored on an hourly basis using one of several O_3 monitors calibrated to U.S.E.P.A. standards by the Maryland Department of Environment (Mulchi et al., 1992; Rudorff, 1993).

3. Results and Discussion

3.1 Chamber Air Quality Treatments.

Summaries of the seasonal 7-h (1000-1600 EST) mean O_3 concentrations for each of the studies conducted over the period 1989-1992 are listed in Table II. The charcoal filters typically reduced the chamber O_3 concentrations to levels 50% below the existing ambient levels. The resulting O_3 concentrations in the carbon filter (CF) treatments were below threshold concentrations (i.e. 40 n mol O_3 mol^{-1}) which cause phytotoxic effects on the crops (Heck et al., 1988; Krupa and Kickert, 1989) being investigated. The

TABLE II

Ambient and chamber seasonal 7-h mean O_3 concentrations (n mol O_3 mol^{-1} for experiments at USDA-BARC 1989-1992.

Year	Crop	Chamber			Ambient Air
		Charcoal Filter (CF)	Nonfilter (NF)	Nonfilter $+O_3$ $(NF+O_3)$	
1989	Soybean	24.1	42.5	68.8	43.8
1990	Soybean	27.8	54.8	76.6	55.5
1991	Wheat	18.6	—	60.7	45.5
1991	Corn	28	—	70.2	60.8
1992	Wheat	20.2	—	64.8	40.7

$NF+O_3$ treatments were $\geq$ to the threshold levels where phytotoxic effects from O_3 exposures have been reported. The seasonal 7-h mean O_3 concentrations for the NF treatments during the summer of 1989 and 1992 was slightly below the 55 $\pm$ 5 n mol O_3 mol^{-1} range typically found for Maryland conditions (Mulchi, 1993).

3.2 Soybeans.

Combined over years, grain yield and oil content increased and grain protein content decreased in soybeans in response to atmospheric CO_2 enrichment (Table III). The grain yield increases were likely caused by the combination of increased pod $plant^{-1}$ and seed $wt.1000^{-1}$, both of which trended upward with increased levels of atmospheric CO_2. Increased exposure to O_3 caused significant reductions in seed $wt.1000^{-1}$ and pod $plant^{-1}$ which resulted in progressively lower grain yields. Grain oil content was lower in the high O_3 treatment compared to the CF control. Increasing the atmospheric CO_2 concentration by 150 μ mol CO_2 mol^{-1} negated the negative impact of O_3 in the NF treatments on pods $plant^{-1}$, grain yield, oil and protein contents thereby confirming the initial report by Mulchi et al.,(1992). Recent studies by Pausch (1993), using ^{13}C-labled CO_2, showed that soybean plants exposedto chronic high O_3 concentrations retained larger quantities of the ^{13}C in their leaves and thereby transported smaller quantities of ^{13}C-labeled photosynthate to roots and nodules compared to plants grown in CF air. The stimulation in pod counts in response to increased CO_2 concentration, especially under high O_3 exposure, suggests that the added CO_2 may have a role in protecting the plant's ability to transport photosynthate from source to sinks. Studies designed to provide greater insights into the mechanism(s) involved are in progress.

3.3 Wheat.

The added CO_2 likewise stimulated significant increases in grain yields, plant biomass, straw, harvest index and seed wt. 1000^{-1} in wheat (Table IV). As was reported by Rudorff (1993), these results can be attributed to several factors including: a) the

TABLE III

Summary of the interactive effects of atmospheric CO_2 enrichment and chronic O_3 air pollution on soybean combined over years. USDA-BARC 1989-1990.

Chamber Treatments		Grain Yields	Seed Wt.	Seed Protein	Seed Oil	Pods Plant^{-1}
CO_2	O_3	g/m^2	g/100	%	%	
	-			Year Means		
	1989	204.3	17.8	39.6	21.9	56.8
	1990	191.8	12.1	41.8	20.1	54.6
	Stat. Sign ($P \leq 0.05$)	*	*	*	*	NS
				CO_2 Treatment Mean s		
Ambient		184.1b	14.4a	41.1a	20.6b	53.8a
+50 μ mol mol^{-1}		200.9a	14.6a	40.6ab	21.2a	56.3a
+150 μ mol mol^{-1}		208.9a	15.8a	40.5b	21.1a	57.0a
	LSD (0.05)	8.4	NS	0.6	0.4	NS
				O_3 Treatment Means		
—	CF	220.7a	16.2a	40.6a	21.0a	57.5a
—	NF	205.4b	15.1ab	40.6a	21.1a	56.0ab
—	NF + O_3	167.8c	13.6b	41.0a	20.7b	53.5b
	LSD (0.05)	8.4	1.8	NS	0.3	3.7
				CO_2 x O_3 Treatment Relative Means$^+$		
Ambient	CF	1.00bc	1.00bc	1.00abc	1.00bc	1.00ab
	NF	0.93d	1.09abc	1.00abc	1.00bc	1.02ab
	NF+O_3	0.75e	0.96bc	1.01a	0.98c	0.94b
Ambient +50 μ mol CO_2 mol^{-1}	CF	1.12a	1.15ab	1.00abc	1.01abc	1.10a
	NF	1.00bc	1.03bc	0.98bc	1.05a	1.06ab
	NF+O3	0.80e	0.91c	1.00abc	1.00bc	0.94b
Ambient +150 μ mol CO_2 mol^{-1}	CF	1.09a	1.26a	0.98bc	1.03ab	1.07a
	NF	1.06ab	1.07abc	1.00abc	1.00bc	1.01ab
	NF+O_3	0.89d	1.01bc	1.00abc	1.02ab	1.06ab
	LSD (0.05)	0.06	0.21	0.02	0.03	0.12

+Means are relative to ambient ambient CF air treatment means.
NS, not statistically significant at $P \leq 0.05$; *, statistically significant at $P \leq 0.05$. Column means having similar letters are not significantly different at $P \leq 0.05$.

combination of increased leaf photosynthesis and reduced photorespiration which results in an increased supply of photosynthate from the source under elevated CO_2; and b) an increase in productive tillers per plant thereby increasing the number of spikes per unit area. In the absence of O_3 stress, the added CO_2 stimulated seed wt. 1000^{-1} by 7% and grain yield by 12%. Grains per spike (data not shown) were not greatly influenced, but straw yields showed a modest 4% increase in response to the added CO_2.

Combined over CO_2 treatments, the increased exposure to O_3 produced significant reductions in grain yields, plant biomass, straw, harvest index and seed wt. 1000^{-1}. Under ambient CO_2 conditions, exposures to high O_3 under ambient CO_2 caused reductions in seed wt. 1000^{-1}, plant biomass, and grain yield equal to 10, 15 and 20% respectively compared to the CF control. However, the effects from high O_3 exposures under elevated CO_2 were greatly diminished and were not significantly different from the ambient CF control except for seed wt. 1000^{-1}. Compared to CF air, harvest index values were reduced by 7% in response to the high O_3 treatments under ambient CO_2. Harvest index values were raised in response to elevated CO_2 under both O_3 regimes with the differences between the two O_3 regimes being nonsignificant. As was noted for soybeans, the added CO_2 would appear to protect the ability of the plant to partition photosynthate to the developing grain thereby negating the negative impact of O_3 stress

TABLE IV

Summary of the interactive effects of atmospheric CO_2 enrichment and chronic O_3 air pollution on winter wheat and corn. USDA-BARC 1991 and 1992.

Chamber Treatments		Wheat (1991 and 1992)					Corn (1991)				
		Grain Yield	Dry Biomass	Straw	Harvest Index	Seed wt.	Grain Yield	Dry Biomass	Stalk	Harvest Index	Seed wt.
CO_2	O_3	g/m^2	g/m^2	g/m^2	(%)	g/1000	g/m^2	g/m^2	g/m^2	(%)	g/1000
		CO_2 Treatment Means									
ambient	---	476	1400	923	34.0	31.0	1167	2005	858	58.1	333
+150 µmol mol^{-1}	---	574	1570	996	36.6	33.7	1210	2079	870	58.1	327
	Stat. Sign. ($P \leq 0.05$)	**	**	*	**	**	NS	NS	NS	NS	NS
		O_3 Treatment Means									
---	CF	562	1560	1000	36.0	33.8	1244	2103	859	59.1	325
---	NF + O_3	488	1405	919	34.6	30.9	1133	1981	849	57.2	335
	Stat. Sign. ($P \leq 0.05$)	**	**	*	*	**	*	NS	NS	*	*
		CO_2 x O_3 Treatment Relative Means+									
ambient	CF	1.00 b	1.00 b	1.00 b	1.00 b	1.00 b	1.00 b	1.00 a	1.00 ab	1.00 ab	1.00 ab
	NF + O_3	0.80 a	0.85 a	0.88 a	0.93 a	0.89 a	0.87 a	0.91 a	0.94 a	0.97 b	1.02 b
ambient	CF	1.12 c	1.07 bc	1.04 b	1.05 b	1.07 c	1.01 b	1.00 a	0.99 ab	1.01 a	0.98 a
+150 µmol mol^{-1}	NF + O_3	1.04 bc	1.01 b	0.99 b	1.03 b	0.99 b	0.94 ab	0.98 a	1.02 b	0.97 b	1.01 b
	LSD (0.05)	0.09	0.07	0.09	0.07	0.07	0.13	0.10	0.07	0.04	0.03

+ Means are relative to ambient CF air treatment means.

*, ** Statistically significant at $P \leq 0.05$ or 0.01, respectively (Rudorff, 1993).

Column means followed by similar letters are not significantly different at $P \leq 0.05$.

on harvest index and grain yield in wheat. Followup studies to identify the mechanisms by which CO_2 reduces the effects of O_3 stress on wheat are also in progress.

3.4 Corn.

Corn, a C4 species, showed no significant differences in response to the elevated CO_2 treatment when combined over O_3 treatments or in the absence of chronic O_3 exposures. (Table IV); however, significant effects of O_3 treatments on grain yield, harvest index and seed wt. 1000^{-1} were found when combined over CO_2 treatments. Under ambient CO_2 levels, the high O_3 treatment reduced grain yield by 13% and harvest index by about 3% compared to the CF controls. Under elevated CO_2, grain yield was reduced only 6% by the elevated O_3 with the differences between the means for the two O_3 regimes being nonsignificant. Although the effects of CO_2 enrichment regarding yields for corn were less spectacular than were found for the C3 species soybeans and wheat, the added CO_2 again exhibited a role in reducing the negative impact of the high O_3 treatment. It is noteworthy that the high O_3 treatment resulted in slightly higher seed wt. 1000^{-1} compared to the ambient CF control. These results were attributed by Rudorff (1993) to a slight reduction in kernels per ear likely caused by reduced pollination in the high O_3 treatments.

4. Conclusions

A series of investigations were conducted at USDA-BARC over the period 1989 to 1992 which involved full-season exposures of soybeans, wheat, and corn to single and combined atmospheric enrichments of CO_2 and O_3. The C3 species exhibited significant gains in biomass and yields in response to the elevated CO_2 both in the presence or absence of O_3. No such gains in response to CO_2 enrichment were observed for corn; however, the added CO_2 did reduce the negative effects from the high level O_3 exposures. Results initially reported by Mulchi, et al.,(1992) concerning the role of elevated CO_2 in partially counteracting the negative effects caused by current ambient levels of O_3 on soybeans were confirmed and expanded to include wheat and corn. The mechanism(s) involved concerning a possible protective role for CO_2 against O_3 are largely unknown. In addition to previously recognized stomatal actions, the results from the present studies support a hypothesis that the added CO_2 somehow protects the plant's ability to partition photosynthates to developing sinks such as grains.

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