Published July, 1990

Effects of Ozone, Sulfur Dioxide, Soil Water Deficit, and Cultivar on Yields of Soybean[1]

Howard E. Heggestad* and Virginia M. Lesser

ABSTRACT

The effects of ozone (O_3) stress on bean yields and seed size of four soybean cultivars [*Glycine max* (L.) Merr.] grown in open-top chambers in 1981, 1982, and 1983 are presented. The O_3 treatments included charcoal filtered (CF) and nonfiltered (NF) air, and 0.03, 0.06, and 0.09 $\mu L\ L^{-1}$ O_3 added 7 h d^{-1} to NF air. The effects of SO_2 in concentrations from 0.005 to 0.224 $\mu L\ L^{-1}$ (4 h d^{-1}, 5 d wk^{-1}) from the seedling stage to maturity were studied in 1981 and 1982. In 1982 and 1983 the effects of soil moisture stress (SMS) and well-watered (WW) soil conditions on the response of soybean to O_3 stress were determined. The primary objective was the evaluation of 3 yr of soybean data from our site using the nonlinear Weibull and the polynomial dose-response models to relate yield responses to O_3 exposure doses. The variables also included SO_2, soil moisture, and cultivar. The homogeneity of the response equations were compared to permit development of the smallest set of homogeneous equations over years. Both O_3 and SO_2 negatively impacted bean yields and seed size. No interactions between O_3 and SO_2 were indicated. With the Weibull model, interactions between O_3 and soil moisture were observed with 'Forrest' in 1982 and 'Williams' in 1983. With an O_3 level considered typical in soybean production areas (seasonal 7 h d^{-1}, 0.055 $\mu L\ L^{-1}$) compared to background O_3 (0.025 $\mu L\ L^{-1}$) and using all data from 3 yr of experiments, the Weibull model predicted the same (15%) mean yield loss under both SMS and WW regimes.

H.E. Heggestad, Climate Stress Lab., USDA-ARS, Beltsville, MD 20705 (present address, 3112 Castleleigh Road, Silver Spring, MD 20904); V.M. Lesser, Dept. of Biostatistics, Univ. of North Carolina, Chapel Hill, NC 27599-7400. Research partly supported by inter-agency agreement between USEPA and the USDA, no. 12-14-7001-1282. Received 30 May 1989. *Corresponding author.

Published in J. Environ. Qual. 19:488–495 (1990).

The National Crop Loss Assessment Program (NCLAN), conducted from 1980–1987, estimated benefits to agriculture from a 10, 25, or 40% reduction of O_3 levels to be \$0.8, \$1.9, and \$2.8 billion (of 1982), respectively (Wilhour, 1988). The effects of different regions of the country, SO_2 dose, cultivar, and soil moisture stress (SMS) were involved to varying extent in the 41 NCLAN field experiments designed to determine the O_3-dose yield response of major agricultural crop species (Lesser et al., 1990). Of these factors, variations in species susceptibility to O_3 had greatest influence, e.g., corn (*Zea mays* L.) was less sensitive than soybean. The effects of SMS seemed to raise more questions about the applicability of the NCLAN dose-response results than other environmental variables (Heagle et al., 1988) and perhaps more than any other factor tested. The influence of SO_2 and cultivar were relatively easy to assess; however, the experiments were limited to one or at most four cultivars of a species at any site due to space limitations in the open-top chambers.

We are summarizing 3 yr of NCLAN experiments conducted on soybean at the Agricultural Research Center, Beltsville, MD. The influences of SO_2, cultivar, and SMS on the O_3 dose yield response were assessed.

[1] The use of trade names in this article does not imply endorsement by the USDA. Although this research was partly funded by the USEPA, it has not been subjected to USEPA review, therefore it does not necessarily reflect their views and no official endorsement should be inferred.

Similar experiments on soybean in North Carolina and Illinois showed no evidence of interaction between O_3 and SO_2 (Heagle et al., 1983; Kress et al., 1986). Sulfur dioxide had no measurable effect on seed yield except at concentrations above those known to exist regionally in the USA, i.e., seasonal 4 h d^{-1} mean of 0.026 $\mu L\ L^{-1}$, according to Heagle et al. (1983). By contrast, at ambient O_3 concentrations there were yield losses. In Illinois, with the cultivars 'Amsoy-71' and 'Corsoy-79', the estimated O_3 induced yield losses were 5 to 15% when exposed for 7 h d^{-1} to a seasonal mean ranging from 0.045 to 0.065 $\mu L\ L^{-1}$ O_3 (Kress et al., 1986). In North Carolina, with the cultivar 'Davis', exposure to a seasonal 7 h d^{-1} O_3 mean of 0.055 $\mu L\ L^{-1}$ reduced yields an estimated 20% compared to yields at 0.025 $\mu L\ L^{-1}$ (Heagle et al., 1983).

Significant interactions have been reported between soil moisture and O_3 in soybean experiments in Maryland (Heggestad et al., 1985, 1988), North Carolina (Heagle et al., 1987), and Illinois (Heagle et al., 1988). The nature of these interactions varied. In North Carolina in 1983 there was a significant linear relation of O_3 dose and soybean yield loss when the soil was well-watered (WW), but not with soil moisture stress (SMS) in a year with severe SMS. However, in 1984, with less SMS there was a linear relation between O_3 dose and soybean yield in both soil moisture regimes. In the 1982–1983 Maryland studies there was evidence that the combined effects of SMS and ambient O_3 on yields were more than additive for the Williams, Forrest, and Corsoy cultivars (Heggestad et al., 1985).

This paper presents parameter estimates of the polynomial and Weibull homogeneous equations relating seed yields to O_3 stress. In 1981 and 1982, yield responses due to exposure from O_3 and SO_2, singularly and in combinations, were studied. In 1982 and 1983, the effects of SMS and WW soil conditions on the yield response of soybean to O_3 were investigated.

MATERIALS AND METHODS

Experimental Design

Split-plot designs were used each year, where the subplot unit was a cultivar and the whole plot unit consisted of the O_3/SO_2/moisture combinations. Two cultivars were investigated each year: 'Williams-79' was paired with 'Essex', Forrest, and Corsoy-79 in 1981, 1982, and 1983, respectively. Essex and Forrest are late maturing cultivars (Group V) and determinant in flowering habit. Williams (Group III) and Corsoy (Group II) mature earlier and are indeterminate forms. The whole-plot arrangement in both 1981 and 1982 were single replicate studies of two-factor and three-factor factorials, respectively, while in 1983 there were three replications in a randomized complete block arrangement. Thirty open-top chambers (Heagle et al., 1973) were used each year.

All the experiments were on a Codorus silt loam soil (fine-loamy, mixed, mesic Fluvaquentic Dystrochrepts), which is deep, permeable to roots, and well drained. There is a water table at about 1.2 m, even after long dry periods. The experiment was on the same site in 1981 and 1982, but moved in 1983 to a nearby larger area that for the 2 previous years had been in corn and rye (*Secale cereale* L.) cover crops. The cover crops were plowed under when the plants were immature. Consequently, soil conditions were excellent for the growth of soybean as revealed by the very high yields of beans.

In 1981, the factorial experiment included all 24 treatment combinations of six SO_2 treatments: 0.0, 0.03, 0.06, 0.12, 0.24, and 0.48 $\mu L\ L^{-1}$ SO_2 added from 1000 to 1400 h EDT from 14 July to 22 September, and four O_3 treatments: charcoal filtered (CF), nonfiltered (NF), NF plus 0.03, and NF plus 0.06 $\mu L\ L^{-1}$ O_3 added from 1000 to 1700 h EDT from 20 July to 22 September. There were three replications of the CF and NF air treatments without SO_2 added and two replications of CF and NF air with 0.03 $\mu L\ L^{-1}$ SO_2 added but no replications of the remaining 20 treatments. The chambers were in place and blowers started 2 July. The pollutants were added each week, Monday through Friday, except on days with rain. The SO_2 treatments were made on 62 d and O_3 on 55 d.

The plots were irrigated to maintain about 0.06 MPa soil water potential (ψ_s) at depths of 0.25 and 0.45 m using tensiometers (Irrometer Co., Riverside, CA) for monitoring. During July, August, and September, 1.9, 12.1, and 7.6 cm of water were applied, respectively. Rain amounts were 11.0, 7.8, and 7.6 cm for July, August, and September, respectively. Rainfall was below average only in August.

In 1981, the cultivars, Essex and Williams, were placed in each chamber in one of two rows 0.91 m apart with the order of cultivars chosen at random. Seeding was on 27 May with sufficient seeds to allow thinning of stands to an average distance of 5 cm. Harvest was made when the plants in each plot reached maturity. Plants in plots with highest pollutant dose matured earlier than those in CF air and were harvested first. All plots of Williams, the earliest maturing cultivar, were harvested by 23 September, whereas those of Essex were harvested about 1 mo later. A 2-m center section of each 3-m row plot was harvested in 0.5-m sections to permit an assessment of position effects in the chamber. The mature plants were cut at ground level and stored about 4 wk at room temperature until dry. The seeds were removed from the pods by hand and further dried until a 6.5% moisture content was achieved. Seed weights were adjusted to 14% moisture. Seed size was determined by weight per 100 seeds.

Methods used in 1981 for O_3 generation, dispensing, and monitoring were the same as those described (Heagle et al., 1979, Heggestad et al., 1985). A TECO Model 49 (Thermo Electron Corp., Hopkinton, MA) and Bendix Model 8002 (Bendix Corp., Lewisburg, WV) were used to monitor O_3, and a Dasibi Model 1003 PC (Dasibi Environmental Corp., Glendale, CA) was used for calibration. Monitoring of SO_2 was with a TECO Model 43, and a TECO Model 143 was used for calibration. The EPA approved quality assurance system was followed to maintain quality control in pollutant monitoring, including an independent audit of instruments at the field site.

Materials and methods utilized in the 1982 and 1983 experiments, except for the SO_2 treaments in 1982, were described previously (Heggestad et al., 1985, 1988). In 1982, except for changes in SO_2 dose, the methodology was the same as in 1981. The 1982 factorial experiment involved all combinations of three levels of SO_2, five levels of O_3, and two moisture levels. The levels of SO_2 were 0.0, 0.03, and 0.12 $\mu L\ L^{-1}$ SO_2 added daily from 1000 to 1400 h EDT from 8 July to 1 October; the five O_3 treatments: CF, NF, and NF with 0.03, 0.06, and 0.09 $\mu L\ L^{-1}$ O_3 added daily from 1000 to 1700 h EDT each day from 14 July to 22 September. The 15 $SO_2 \times O_3$ treatments were conducted under two soil moisture conditions, i.e., WW and SMS with ψ_s averaging about -0.05 and -0.40 MPa, respectively, at 0.25- and 0.45-m depths (Heggestad et al., 1985). The ψ_s in each WW plot was monitored with tensiometers, while the ψ_s in each SMS plot was monitored with soil psychrometers (WESCOR PCT-55-15, Wescor Co., Logan, UT). The ψ_s values in each WW plot were read each morning to determine the amout of irrigation to apply on each plot, if any. The soil psychrometers were read each morning, except when plots were more moist than targeted ψ_s (-0.40 MPal) recorded the previous day. Water was applied by trickle irrigation (AGRI-

FIM Drip Irrigation, Shemin Nurseries, Burtonsville, MD.) After midseason in the WW regime, daily irrigation was often required to maintain targeted ψ_s.

In 1983, there were three replications of the same O_3 and soil moisture treatments as used in 1982, and also, three replications of SMS and WW plots without chambers (Heggestad et al., 1985, 1988). The O_3 treatments were made from 23 July to 23 September. For each year, the pollutant treatments were continued at least until all plants reached physiological maturity. Rain was excluded from two-thirds of the plot area by placing plastic sheeting supported on wood strips about 10 cm above the soil between the rows (Heggestad et al., 1985). The plastic extended on the soil surface beneath the chamber and for a meter beyond to permit removal of collected water. The first irrigation in the WW series was on 18 July and in the SMS series on 3 August.

ANALYSIS

The dependent variables analyzed were crop yield (kg ha^{-1}) and seed size (g/100 seeds). The independent variables were O_3 and SO_2 (μL L^{-1}), and moisture level. For the dose-response models, moisture level was incorporated into the model only as an indicator variable, with 1 and 0 defining the WW and SMS conditions, respectively. Ozone and SO_2 were treated as quantitative variables.

For each year, the validity of the least squares assumptions was checked. The residuals for each study were plotted against the predicted values, and normal plots of the residuals were used to investigate patterns of normality and heterogeneous variances. Bartletts chi-square test was used to test homogeneity of variances between cultivars and moisture levels within each year and over years. In order to obtain the estimates of experimental error, the predicted values, and the residuals for the single replicate studies, a full response polynomial model was fit removing all possible significant effects. Analysis of variance was run on the 1983 studies.

Both polynomial and Weibull dose-response models were used to relate the yield responses to the effects of O_3. Predicted values at two O_3 concentrations are presented to compare differences in the two models. The application of the Weibull model to NCLAN data has been described (Rawlings and Cure, 1985; Lesser et al., 1990) The model is defined as:

$$y = \alpha \exp[-(x/\sigma)^c]$$

where y is the yield response and x is the O_3 concentration. The three parameters to be estimated are α, σ, and c. The effects of the other treatment factors (blocks, SO_2, moisture level, and cultivars) were incorporated into this model by expanding the scaling parameter, α, into a sum of the relevant effects. Blocks, moisture levels, and cultivars are incorporated into the polynomial model as additional linear terms. The primary effects of O_3, SO_2, and moisture level were kept in the model regardless of significance. The degree of polynomial model fit was determined by tests of significance ($P < 0.05$) of the lack-of-fit sums of squares. For both the polynomial and Weibull models, difference in the residual sums of squares between full and reduced models were used to test homogeneity of response to O_3 and SO_2 over other treatment factors (cultivars and moisture levels). The full model allowed a different response to O_3 (or SO_2) for each level of the other factors, while the reduced model forced a common response over all levels. The test of homogeneity were used to combine responses into a minimum number of homogeneous equations for each year. These responses were then tested for homogeneity over the 3 yr of experiments to obtain the smallest set of homogeneous response equations. All test were based on $P < 0.05$.

RESULTS

In each year the plants exposed to the highest O_3 and/or SO_2 dose and SMS were mature about 3 wk before plants grown in CF air and WW. Data are provided from the experiments in each of the 3 yr, beginning with the 1981 season.

Yield and Seed Size

The effects in 1981 of SO_2 and O_3 singularly and in combinations, on the yields of the cultivars Essex and Williams are presented in Table 1. A regression analysis, not shown, revealed significant effects on yield due to exposures from O_3 and SO_2, but no significant interactions. The cultivar responses to both pollutants were linear, although the measured yield of Williams was about the same following exposure to 0.022, 0.035, and 0.068 μL L^{-1} SO_2. Exposure to the highest doses of these pollutants, 0.24 μL L^{-1} SO_2 and 0.064 μL L^{-1} O_3 for the exposure times indicated, each reduced the cultivar yields by about 30%. These differences in pollutant doses provide some measure of the greater toxicity of O_3 over SO_2. The effects of SO_2 and O_3 on soybean yields were independent of each other.

In 1982, the regression analyses showed significant linear effects on yield of Williams from O_3 and SO_2 but only significant effects of O_3 on Forrest. The relatively high negative correlations presented in Table 2 for O_3 and yield indicate that the linear model was quite effective in relating yield response to O_3, and moderately effective in relating seed size to O_3 in all cases except WW Forrest. The correlations between SO_2 concentrations and both yield and seed size were not significant so they are not shown in Table 2. The reduction in yield, compared to CF, from exposure of Williams to 0.067 μL L^{-1} O_3 in 1982 was about the same as that from 0.064 μL L^{-1} O_3 in 1981. Increasing

Table 1. Seed yields and seed size in 1981 for cultivars Essex and Williams in response to increasing doses of SO_2 and O_3 with plants grown in open-top field chambers.

	Essex		Williams	
Pollutant concentration	Seed yield†	Seed size	Seed yield†	Seed size
μL L^{-1}	kg ha^{-1}	g 100 seeds^{-1}	kg ha^{-1}	g 100 seeds^{-1}
SO_2‡				
0.005	4061	13.5	4351	18.0
0.022	3944	13.3	3867	18.1
0.035	3419	13.0	3893	18.6
0.068	3372	13.1	3852	17.9
0.120	3049	12.5	3370	16.3
0.224	2678	11.4	3059	16.4
r§	−0.61	−0.67	−0.57	−0.42
O_3‡				
0.015 (CF)¶	4088	13.3	4328	18.8
0.039 (NF)¶	3445	13.2	4052	17.9
0.054	3089	12.6	3194	17.1
0.064	2881	11.8	3127	16.0
r§	−0.65	−0.63	−0.77	−0.65

† Yield values for SO_2 treatments are means over four O_3 treatments; yield values for O_3 treatments are means over six SO_2 treatments.
‡ Means: SO_2 = 4 h d^{-1} and O_3 = 7 h d^{-1}, 5 d wk^{-1}, from mid-July to 22 Sept. 1981. Values are seasonal means.
§ Pearson correlations were computed using the individual plot values.
¶ CF = charcoal filtered, NF = nonfiltered.

the mean O_3 level to 0.099 $\mu L\ L^{-1}$, 7 h d^{-1} resulted in about 40% reduction in seed yield based on the comparsion of yield in CF air.

For 1983, only some comments about the 1983 yield data are presented since they are available in Heggestad et al., (1988). Plant growth was excellent and seed yields were very high in 1983 compared to the previous 2 yr. As explained under methods, the experiment was moved to an adjacent area that was much improved in soil texture and fertility. For example, in CF air and under WW conditions, Williams yielded 4328 and 5130 kg ha^{-1} in 1981 and 1982, respectively (Tables 1 and 2); whereas in CF air and under WW conditions, Williams in 1983 yielded 7595 kg ha^{-1} (Heggestad et al., 1985). The effects of O_3 on seed yield for five levels of O_3 in 1983 are available in Heggestad et al., (1988). Rather than yield in kilograms per hectare as in this report, the data are presented on a gram per plant basis. However, for conversion purposes, plants per hectare are provided in a table footnote.

An analysis of variance (Heggestad et al., 1984) for Williams revealed a significant effect of O_3 ($P < 0.0001$) and an effect of moisture on yield ($P < 0.002$). There was also a significant interaction ($P < 0.004$) between O_3 and soil moisture. With Corsoy the analysis revealed an O_3 effect on yield ($P < 0.001$). However, there was no moisture effect, although an interaction between O_3 and moisture ($P < 0.04$) was revealed as shown by Fig. 4 in Heggestad et al., (1985). In both of the above regression analyses the seasonal mean 7-h O_3 concentration for each plot and mean ψ_s for each plot were used rather than, as previously described, only the indicator variables one and zero to define WW and SMS conditions, respectively.

Seed Size vs. Seed Yield

Although not shown, Pearson correlations of seed yields and seed size (g 100 $seeds^{-1}$), based on the individual plot data, were determined for each cultivar in each year and under both moisture regimes. All correlations were highly significant ($P < 0.001$), except for Forrest in the WW plots in 1982 ($P = 0.42$). In1983 the correlations, which ranged between 0.87 and 0.92, were higher than in 1981 and 1982, except for Williams WW in 1982 having a value of 0.96. Polynomial regression models (also not shown) relating seed size to the independent variables produced equations that were similar to those of yield (Table 3).

Table 2. Seed yield and seed size in 1982 for Forrest and Williams in response to increasing doses of SO_2 and O_3 with plants grown in open-top field chambers under well-watered and water-stress conditions.

	Forrest		Williams	
Pollutant concentrations	Seed yield†	Seed Size	Seed yield†	Seed size
$\mu L\ L^{-1}$	kg ha^{-1}	g 100 $seeds^{-1}$	kg ha^{-1}	g 100 $seeds^{-1}$
SO_2‡				
		Well-watered		
0.005	3677	11.7	4466	17.6
0.017	3637	12.3	3441	15.5
0.059	3965	11.6	4010	16.7
		Water-stress		
0.005	3873	13.2	3425	16.9
0.017	3338	13.0	2881	16.2
0.059	3703	13.2	3592	17.2
O_3§				
		Well-watered		
0.017 (CF)	4101	11.8	5130	18.7
0.049 (NF)	4730	13.0	4790	18.0
0.067	3614	12.3	3964	16.5
0.084	3441	11.4	3064	15.4
0.099	2912	11.1	2914	14.3
r¶	−0.62	−0.37#	−0.85	−0.85
		Water-stress		
0.017 (CF)	5046	14.5	4482	19.5
0.049 (NF)	3528	13.7	3193	17.8
0.067	3396	13.1	3195	16.6
0.084	3337	12.4	3093	15.5
0.099	2881	11.9	2534	14.4
r¶	−0.86	−0.88	−0.83	−0.94

† Yield values for SO_2 treatments are means over five O_3 treatments; yield values for O_3 treatments are means over three SO_2 treatments.
‡ Seasonal means 4 h d^{-1}, 1000 to 1400 h EDT, from 8 July to 1 October.
§ Seasonal means 7 h d^{-1}, 1000 to 1700 h EDT, from 8 July to 1 October.
¶ Pearson correlations were computed using the individual plot values; the correlations for SO_2 and seed yield were not significant so they are not shown.
# This correlation is not significant, whereas all other correlations are significant at $P \leq 0.05$.

Table 3. Parameter estimates of the polynomial homogeneous equations relating seed yield (kg ha^{-1}) to O_3 and to SO_2 in ($\mu L\ L^{-1}$).

			Slope		
Year	Cultivar	Intercepts†	O_3‡	Other(s)‡	
			By year		
1981	Essex	4 848 (134)	−24 657 (2 792)	SO_2:	−4 926 (715)
	Williams	5 131 (134)			
1982	Forrest	5 177 (199)	−22 528 (2 315)	SO_2:	−7 568 (2 467)
	Williams	5 114 (199)		Moisture:	390 (133)
1983	Corsoy	5 898 (252)	−23 382 (2 328)	Moisture:	985 (287)
	Williams	7 055 (252)		O_3 × moisture:	−11 910 (3 256)
			Over years§		
1981	Essex	4 424 (166)	−23 180 (1 734)	SO_2:	−5 303 (769)
1981	Williams	4 707 (166)		Moisture:	390 (118)
1982	Forrest	5 145 (150)			
1982	Williams	5 082 (150)			

† Estimated standard errors are in parentheses.
‡ Applies to both cultivars within years, and the four cultivars over years.
§ Neither the response of Corsoy nor Williams in 1983 were homogeneous with the 1981 and 1982 responses, so they are not included.

Table 4. Parameter estimates of the Weibull homogeneous equations by year relating seed yield (kg ha^{-1}) to O_3 ($\mu L\ L^{-1}$), and in some years also SO_2 ($\mu L\ L^{-1}$) and soil moisture.†

Cultivar‡	Alpha (O_3)	Alpha (x) label	Alpha (x)	Sigma	c
			1981		
E	4 526 (180)	SO_2	−5 569 (888)	0.106 (0.017)	2.01 (0.63)
W	4 862 (189)				
			1982		
W	4 964 (322)	SO_2	−15 102 (3 715)	0.137 (0.014)	1.56 (0.45)
		Moisture	944 (204)		
F (SMS)	5 738 (322)	SO_2	−10 173 (5 052)	0.153 (0.020)	1.00 (0.0)§
F (WW)	4 313 (381)	SO_2	823 (6 819)	0.126 (0.023)	3.71 (2.57)
			1983		
C	6 524	Moisture	−118 (323)	0.148 (0.013)	1.14 (0.38)
W (SMS)	6 193			0.190 (0.021)	1.65 (0.48)
W (WW)	7 879			0.145 (0.008)	1.60 (0.35)

† Standard errors are in parentheses. E = Essex, W = Williams, F = Forrest, and C = Corsoy. SMS = soil moisture stress, WW = well-watered.
‡ Forrest in 1982 and Williams in 1983 were heterogeneous with respect to moisture. Moisture was not a treatment in 1981.
§ Due to convergence problems in fitting these data, the c parameter for 1982 Forrest under SMS was fixed to one, which best approximates this parameter and the other parameters were then estimated.

Table 5. Parameter estimates of the Weibull homogeneous equations over years relating seed yield (kg ha^{-1}) to O_3 ($\mu L\ L^{-1}$), SO_2 ($\mu L\ L^{-1}$), and soil moisture stress, and used to develop the response curves in Fig. 1.†

Year/cultivar	Alpha (O_3)	Alpha (x) label	Alpha (x)	Sigma	c
			Curves A (Fig. 1)‡		
1982 F	5 767 (492)	SO_2	−14 089 (3 276)	0.150 (0.014)	1.08 (0.34)
1982 W	5 266 (454)	Moisture	1 004 (222)		
			Curves B (Fig. 1)§		
1981 E	4 793 (318)	SO_2	−6 027 (1 013)	0.146 (0.006)	1.27 (0.16)
1981 W	5 138 (325)	Moisture	−107 (249)		
1983 C	6 361				
1983 W	8 420				
			Curve C (Fig. 1)¶		
1982 F	4 401 (316)	SO_2	25 (5 835)	0.178 (0.021)	1.94 (0.59)
1983 W	6 436				

† Estimated standard errors are in parentheses. E = Essex, W = Williams, F = Forrest, and C = Corsoy. SMS = soil moisture stress and WW = well-watered. When plotting response curves in Fig. 1, the appropriate cultivar alpha (O_3) values were averaged, and the moisture value included only for curves labeled adequate moisture.
‡ Curves A includes: 1982 W (SMS and WW) and 1982 F (SMS).
§ Curves B includes: 1981 W and E (WW), 1983 C (SMS and WW) and 1983 W (WW).
¶ Curve C includes: 1982 F (WW) and 1983 W (SMS).

Dose-Response Analyses for Yield

Inspection of the residual and normal plots showed no patterns of nonnormality or heterogeneous variances. Bartlett's test showed no significant differences in the estimates of variances within or over studies. Tests of homogeneity show similar responses to O_3 for both cultivars within each study. The polynomial regression equations are presented in Table 3. All 3 yr showed significant negative impacts of O_3 on crop yield. In 1983, the moisture by O_3 interaction on seed yield was significant for both cultivars. The response over years of yield to O_3 was homogeneous for 1981 and 1982 but not for 1983.

The Weibull equations obtained for each year are shown in Table 4. For 1981, the response of yield to O_3 was homogeneous over cultivars. For 1982, the response of the cultivar Williams was homogeneous with regard to moisture level, but it was heterogeneous to moisture level for the cultivar Forrest. Consequently, separate equations for moisture levels are presented for Forrest. In 1983, Williams was heterogeneous with regard to moisture level while Corsoy was homogeneous. Subsets of the data were homogeneous, as indicated by the five homogeneous equations shown in Table 5. Curve A, adequate soil moisture, and curves B, with and without adequate soil moisture, overlap over most of the range of O_3 concentrations (Fig. 1). Curve A with inadequate soil moisture shows 16% less yield than curve A having adequate soil moisture. The slope for inadequate soil moisture is the same as for adequate soil moisture since they represent homogeneous equations. Curve C, which includes Forrest WW in 1982 and Williams SMS in 1983, is relatively flat compared to the curves A and B (Fig. 1)

To compare differences in the polynomial and Weibull models, predicted yield losses at 0.055 and 0.07 $\mu L\ L^{-1}$ O_3 7 h d^{-1} for the season are presented in Table 6. Moisture in the analyses were treated as indicator variables, with one and zero defining the WW and SMS conditions, respectively. The polynomial equations in Table 3 and the Weibull equations in Table 4 were utilized in the computations. Although some variation in the response to cultivars and years was shown, at 0.055 $\mu L\ L^{-1}$ O_3 the predicted mean yield losses using the Weibull model were the same (15%) for WW and SMS conditions. The mean percent yield losses for the WW and SMS regimes predicted by each model were in close agreement (Table 6), indicating the models were quite similar in predicting yield loss.

Table 6. Predicted yield losses from O_3 exposure obtained with polynomial and Weibull model equations shown in Tables 3 and 4.†

Year	Cultivar	Polynomial		Weibull		Cultivar	Polynomial		Weibull	
		0.055	0.07	0.055	0.07		0.055	0.07	0.055	0.07
		Soil well-watered. Percent loss in bean yield and O_3 concentrations								
1981	Williams	−17	−25	−19	−32	Essex	−18	−26	−19	−32
1982	Williams	−14	−21	−16	−24	Forrest	−14	−20	−4	−10
1983	Williams	−15	−22	−14	−22	Corsoy	−18	−26	−17	−26
Means		−15	−23	−16	−26		−17	−24	−13	−23
		Soil moisture stress. Percent loss in bean yield and O_3 concentrations								
1982	Williams	−15	−22	−16	−24	Forrest	−15	−22	−18	−25
1983	Williams	−11	−16	−9	−15	Corsoy	−13	−20	−17	−25
Means		−13	−19	−13	−20		−14	−21	−18	−25

† To obtain yield losses, the models utilized 0.055 and 0.07 $\mu L\ L^{-1}\ O_3$, 7 h d^{-1} for the season, as compared to yields with O_3 at 0.025 $\mu L\ L^{-1}$. We assumed SO_2 at 0.005 $\mu L\ L^{-1}$. In making the computations well-watered soil was represented by one and soil moisture stress by zero.

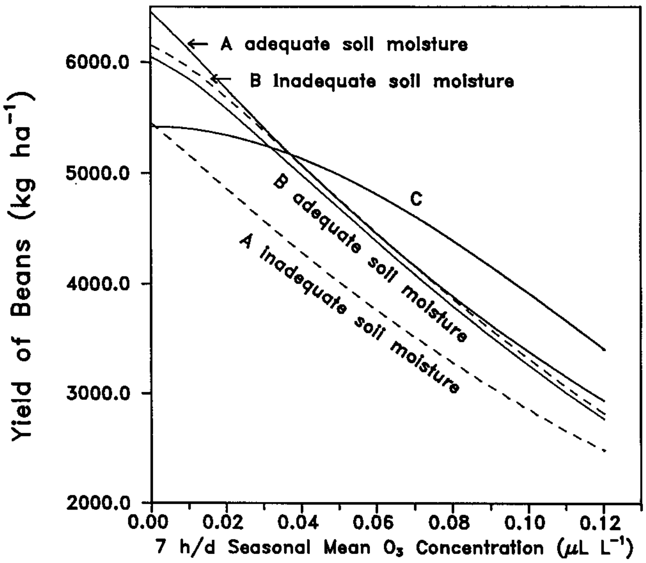

Fig. 1. Minimum number of yield curves over years derived from Weibull homogeneous equations (Table 5) for soybean experiments. Includes (A) 1982 Williams well-watered (WW) and water-stressed (SMS) and 1982 Forrest SMS; (B) 1981 Essex and Williams WW, 1983 Corsoy WW and SMS, and 1983 Williams WW; and (C) 1982 Forrest WW and 1983 Williams SMS. Curves A and B are plotted with and without the alpha moisture factor (Table 5) to show yield response when soil moisture is adequate and inadeqaute.

DISCUSSION

Current levels of O_3 have a negative impact on soybean seed yields. At an O_3 dose of 0.055 $\mu L\ L^{-1}$ 7 h d^{-1} compared to 0.025 $\mu L\ L^{-1}$ (background O_3 level), the Weibull model predicted an average yield loss of 15% under both WW and SMS regimes (Table 6). This includes all data for cultivars and years. Using only data from the 1982 and 1983 seasons, having both WW and SMS treatments, the average losses from exposure to 0.055 $\mu L\ L^{-1}\ O_3$ were 12.8 and 15.0%, respectively. In the same years, at the 0.07 $\mu L\ L^{-1}$ level of O_3, the mean yield losses were 20.5 and 22.3% for WW and SMS, respectively. Although variability exists due to cultivar and year (Table 6), there were relatively little differences in the results comparing losses in the SMS and WW regimes. Consequently, there is no evidence from these data that the NCLAN protocol involving use of irrigation to avoid possible complexities due to drought caused increases in O_3 sensitivity and greater yield losses with soybean, as suggested by Brennan et al., (1987). It is also questionable that there is justification for the 20% predicted reduction in crop sensitivity to O_3 when drought causes yield reductions as utilized in modeling studies by King (1988). Even with 16% reduction in yield due to SMS, slopes A in Fig. 1, for adequate and inadequate soil moisture, were the same.

The Weibull equations in Table 4 were tested for homogeneity over years, which resulted in the equations in Table 5. Each of the equations, which include both SMS and WW treatments, suggested no differences in the shape of the response surface over moisture level. Figure 1 suggests that the differences between curves A and B were not pronounced. Since the slopes of all Weibull curves in Fig. 1 except C were about the same, the estimated percentage yield losses from O_3 exposure were not changed due to an indicated presence or absence of adequate soil moisture.

The effect of SMS on the yield of each cultivar grown in 1982 and 1983 can be compared using the Weibull models shown in Table 4. Two of the four cultivars (Forrest in 1982 and Williams in 1983) showed a heterogeneous yield response to O_3 over moisture level. Heagle et al. (1983) in North Carolina predicted a 20% decrease in seed weight of soybean for a 1981 experiment; however, at a comparable O_3 dose and WW conditions losses were 12 and 14% in 1983 and 1984, respectively (Heagle et al., 1987). Consequently, the mean yield loss in North Carolina for WW plots over the same 3 yr period was also 15%. Their predicted loss for water-stressed plants in 1984 was 12%, but in 1983, a much drier season there was not a significant effect of O_3 on yield. In 1983 the very severe drought reduced yields 49% compared to a 20% reduction in 1984, a year when drought was not as prolonged or as severe, as indicated by ψ_s.

In 1983 Williams showed an interaction between O_3 and SMS (Table 4). Soil water potential data for each plot were available in 1982 and 1983 but not for 1981. In order to analyze data combined over years, moisture in the anaylses was treated as an indicator variable with one and zero defining the WW and SMS conditions. An eariler analysis of the 1983 data using the ψ_s for each plot is presented in Heggestad et al., (1984). The response was almost the same whether 1 and 0 represented WW and SMS, respectively, or plot ψ_s were used. With Williams, the interaction reveals that the yield effects of SMS were dependent on O_3 concentration. At 0.02 $\mu L\ L^{-1}$, SMS reduced yields 10%

while at 0.06 $\mu L\ L^{-1}$, the yield reduction was 5%, but at 0.10 $\mu L\ L^{-1}$ the yields were 5% greater in the SMS plots than in WW plots. That is, at relatively high O_3 concentrations SMS gave measurable protection from O_3 damage.

For Corsoy the analysis using 1 and 0 to represent WW and SMS did not reveal an interaction between soil moisture and O_3, whereas an interaction was revealed using ψ_s for each plot (Heggestad et al., 1985). With the latter analysis the combination of SMS and O_3 in the ambient range was more severe on yield than WW and O_3, but above ambient O_3 concentrations plants grown under SMS conditions showed less yield loss due to O_3 exposure than WW plants. It seems that soil moisture may alter soybean yield response to O_3 in at least four ways, depending on the cultivar, degree of SMS, and soil conditions. The latter includes a range of properties that influence the nature and extent of root penetration and development that indirectly, also influences final bean yield. The NCLAN soil moisture-O_3 experiments (Heagle et al., 1988) indicate that there may be (i) no measureable effect of moisture stress on the yield response of plants to O_3, depending upon the severity of SMS; (ii) reduced sensitivity to O_3 and therefore reduced yield losses; (iii) additive effects of O_3 stress and SMS on yields; and (iv) under some conditions more than additive effects of O_3 stress and SMS on yields.

Soil conditions at our site greatly increased root growth in response to SMS, especially at ambient O_3 exposure as in NF air (Fig. 2, Heggestad et al., 1988). At highest levels of O_3 (NF + 0.06 and NF + 0.09 $\mu L\ L^{-1}\ O_3$) there were no significant differences in root growth between SMS and WW conditions. At lower levels of O_3 exposure (CF, NF, and NF + 0.03 $\mu L\ L^{-1}$) there was significantly more root growth under SMS than under WW conditions. Apparently this decreased the amount of photosynthate available for seed production and may account, at least in part, for the more than additive reduction in measured yields in 1982 and 1983 due to combined stress from ambiant O_3 and SMS shown by comparing yields in NF and CF air (Heggestad et al,, 1985).

Under SMS conditions there was reduced root growth as well as top growth as the O_3 exposure dose increased; however, there was no measureable effect of O_3 on root growth when the plants were in the WW regime (Heggestad et al., 1988). The impact of O_3 on above- and below-ground biomass was similar under SMS conditions, but only top growth, including seed yield, was reduced under the WW conditions as managed in our experiment. The plants were watered, if necessary, every day using trickle irrigation with amounts needed to keep the ψ_s in each plot at about -0.05 MPa at depths of 0.25 to 0.45 m.

The unusual low yield of Forrest in CF air in the WW series in 1982 was attributed to significant cold injury only to plants in the latest maturing plots (Heggestad et al., 1985). The plants in the SMS series and in plots exposed to ambient or higher levels of O_3, in both moisture regimes, were more mature and therefore damaged only slightly, if at all. The cold injury and subsequent reduced yield in the CF-WW plots of Forrest may also account for the lack of a significant correlation between seed size and yield of Forrest in the WW series, and the distinctly nonlinear Weibull response curve C (Fig. 1) representing Forrest WW in 1982 and Williams SMS in 1983.

Plant growth was excellent and seed yields were very high in 1983 compared to the previous 2 yr. Although yields were very high, the predicted percentage yield loss, at an ambient level of O_3, for Williams under WW conditions was about the same as in 1981 and 1982 (Table 6).

There was a significant interaction between soil moisture level and yield respone to O_3 in 6 out of 11 NCLAN experiments (Heagle et al., 1988). In addition to soybean, this occurred with cotton (*Gossypium hirsutum* L.) in California (Temple et al., 1985). The interaction for cotton under water-stressed and normally irrigated plots in the 1981 experiment was similar to that described for Williams in 1983. That is, losses from SMS were dependent upon O_3 concentration, being greatest when O_3 concentrations were low. In all experiments, except soybean in Maryland in 1982 and 1983, SMS reduced the amount of yield loss due to exposure to O_3.

Yield was decreased by increasing levels of O_3 for all of our experiments. For the range of response curves examined in this paper, we believe the nonlinear Weibull model characterizes the shape of the O_3 dose-response curve better than the polynomial response function. Rawlings and Cure (1985) and Lesser et al., (1990) discuss the advantages of choosing the Weibull model as the response function to fit the type of data analyzed in this paper. There are uncertainties even with 3 yr of data on soybean from a single site. The effects of SO_2 and moisture level, in addition to the negative impact due to O_3, have varying impacts on yield, depending upon the cultivar and year. These seemingly inconsistent results of the additional impact of SO_2 and moisture level with O_3, and differences with similar studies at other sites, e.g., Heagle et al., (1987) in North Carolina, suggest additional research in this area, including studies of roots under field conditions, is needed to fully understand the combined effects of these stress factors on soybean.

ACKNOWLEDGMENTS

The authors wish to thank E. H. Lee and T. J. Gish for professional contributions, and K. L. Tuthill for technical assistance. The data used in these analyses were in data storage facilities. Air Quality Research Program, North Carolina State Univ., Raleigh, NC.

REFERENCES

Brennan, E., I. Leone, B. Greenhalgh, and G. Smith. 1987. Chlorophyll content of soybean foliage in relation to seed yield and ambient ozone pollution. J. Air Pollut. Control Assoc. 37:1429–1433.

Heagle, A.S., D.E. Body, and W.W Heck. 1973. An open-top field chamber to assess the impact of air pollution on plants. J. Environ. Qual. 2:365–368.

Heagle, A.S., R.B. Philbeck, H.H. Rogers, and M.B. Letchworth. 1979. Dispensing and monitoring ozone in open-top chambers for plant effects study. Phytopatholgy 69:15–20.

Heagle, A.S. W.W. Heck, J.O. Rawlings, and R.B. Philbeck. 1983. Effects of chronic doses of ozone and sulfur dioxide on injury and yield of soybeans in open-top field chambers. Crop Sci. 23:1184–1191.

Heagle, A.S., R.B. Flagler, R.P. Patterson, V.M. Lesser, S.R. Shafer, and W.W. Heck. 1987. Injury and yield response of soybean to chronic doses of ozone and soil moisture deficit. Crop Sci. 27:1016–1024.

Heagle, A.S., L.W. Kress, P.J. Temple, R.J. Kohut, J.E. Miller, and H.E. Heggestad. 1988. Factors influencing ozone dose-yield response relationships in open-top field chamber studies. P. 141–179. *In* W.W Heck et al. (ed.) Assessment of crop loss from air pollutants. Proc. Int. Conf., Raleigh, N.C. 25–29 Oct. 1987. Elsevier Applied Science, London.

Heggestad, H.E., T.J. Gish, E.H. Lee, J.H. Bennett, and L.W. Douglass. 1985. Interaction of soil moisture stress and ambient ozone on growth and yields of soybeans. Phytopathology 75:472–477.

Heggestad, H.E., E.L. Anderson, T.J. Gish, and E.H. Lee. 1988. Effects of ozone and soil water deficit on roots and shoots of field-grown soybeans. Environ. Pollut. 50–259–278.

Heggestad, H.E., T.J. Gish, E.H. Lee, and L.W. Douglass. 1984. The effects of O_3 and soil moisture stress on the productivity of two soybean cultivars. p. 74–102. *In* National Crop Loss Assessment Network (NCLAN) 1983 Annual Report. EPA 600/3-85-061. USEPA, Corvallis, OR.

King, D.A. 1988. Modeling the impact of ozone × drought interactions on regional crop yields. Environ. Pollut. 53:351–364.

Kress, L.W., J.E. Miller, H.J. Smith, and J.O. Rawlings. 1986. Impact of ozone and sulfur dioxide on soybean yield. Environ. Pollut. 41:105–123.

Lesser, V.M., J.O. Rawlings, S.E. Spruill, and M.C. Somerville. 1990. Ozone effects on agricultural crops: Statistical methodologies and estimated dose-response relationships. Crop Sci. 30:148–155.

Rawlings, J.O., and W.W Cure. 1985. The Weibull function as a dose-response model to describe ozone effects on crop yields. Crop Sci. 25:807–814.

Temple, P.J., O.C. Taylor, and L.F. Benoit, 1985. Cotton yield response to ozone as mediated by soil moisture and evapotranspiration. J. Environ. Qual. 14:55–60.

Wilhour, R. G. 1988. Introduction. p. 1–5. *In* W.W Heck et al. (ed.) Assessment of crop loss from air pollutants. Proc. Int. Conf., Raleigh, N.C. 25–29 Oct. 1987. Elsevier Applied Science, London.