# Influence of Chronic Sulfur Dioxide Exposures on Early Blight of Tomato

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## **ABSTRACT**

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Potted tomato plants were grown outdoors in the presence of airborne Alternaria solani inoculum and were either sprayed with chlorothalonil fungicide or left unsprayed. Plants were moved into controlled-environment chambers and exposed to  $393~\mu g^{-3}$  (0.15 ppm) SO<sub>2</sub> or charcoal-filtered air for 72 continuous hours per week for 4 wk. After exposures, plants were returned to their original outdoor positions. Interactions were observed between SO<sub>2</sub> exposure and fungicide treatment for the number of acceptable fruits per plant and mean individual fruit weight per plant. The combined exposure to SO<sub>2</sub> and infection by A. solani resulted in a decrease in fruit number and an increase in mean individual weight of remaining fruit.

Atmospheric pollutants can increase, decrease, or have no effect on the incidence or severity of plant disease (6,13). Weinstein et al (18) found that

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sulfur dioxide (SO<sub>2</sub>) had no significant effect on early blight severity when gaseous exposures of immature Bonny Best tomato plants (Lycopersicon esculentum Mill.) occurred before and after inoculation with Alternaria solani (Ell. & G. Martin) Sor. However, because early blight severity is directly correlated to physiological maturity and fruit load (1,7,14), the potential interaction between SO<sub>2</sub> and A. solani should be tested on more mature plants. The objective of this study was to determine whether SO<sub>2</sub> would affect early blight severity on mature tomato plants, as determined by observations on fruit yield and plant biomass.

## MATERIALS AND METHODS

Seeds of Merit, a processing tomato

grown in Pennsylvania, were sown one to three per cell in Ball AC-4/8 cell packs (Ball Seed Co., West Chicago, IL) containing a 1:1:1 mixture of steamtreated peat-perlite-soil. Soil was amended based on tests conducted by the Penn State Merkle Soil Testing Laboratory (9). Cell packs were placed in controlled-environment chambers maintained at 24  $\pm$  0.5 C, 75  $\pm$  5% relative humidity (RH), and 415  $\mu$ Em<sup>-2</sup> sec<sup>-1</sup> light intensity with a 14-hr photoperiod beginning at 0600 hours. Plant emergence occurred about 1 wk after seeding. One month after emergence, plants were placed in cold frames where they were maintained for 10 days. Seedlings were transplanted, one per 6-L plastic pot containing the medium described. Plants were staked and placed outdoors on a plastic-covered ground bed in a randomized complete block design (Fig. 1).

Every 7-10 days throughout the season, about 400 ml of 20-19-18 watersoluble fertilizer (Robert B. Peters Co., Inc., Allentown, PA) at a rate of 15 g/3.8 L of water was applied to each pot. Insects were controlled with malathion (15 ml/3.8 L  $H_2O$ ) as needed.

In order to maintain plants free of early blight, 18 treatment plants of each replicate (Fig. 1) were sprayed to runoff with chlorothalonil fungicide at a rate of 15 ml/3.8 L  $H_2O$  on 11 June 1981. Subsequent fungicide applications for control of early blight were based on spray recommendations from a forecasting system (11) and were made on 2, 10, 21, and 30 July and 13 August. An equal number of treatment plants was not sprayed.

An additional group of 64 tomato plants was inoculated with A. solani on 11 June 1981 as follows. Two-week-old cultures of A. solani on potato-dextrose agar were homogenized in a Sorvall

blender (DuPont Co., Instrument Products, Newton, CT) with distilled water and sprayed to runoff on tomato plants. Inoculated plants were covered with a clear plastic bag for 16 hr, then randomly identified and distributed around treatment plants in the outdoor bed (Fig. 1). Because of variability in lesion number, artificially inoculated plants were randomized and redistributed within the bed three more times at 2-wk intervals.

Beginning 11 June 1981, treatment

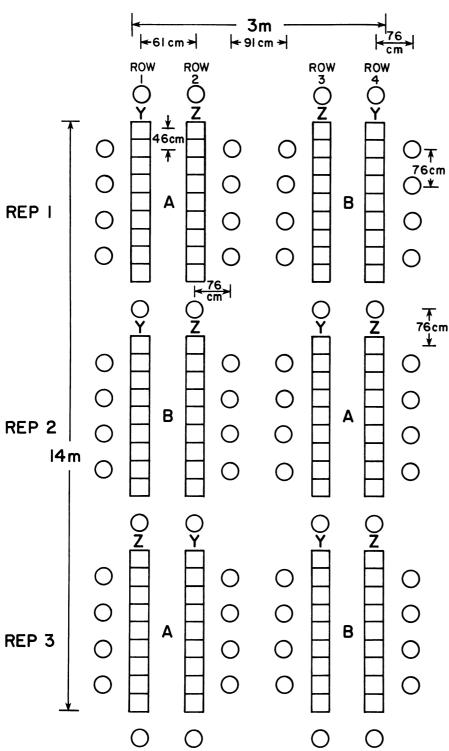


Fig. 1. Plot design for  $SO_2 \times$  early blight interaction study: A = plants sprayed with fungicide, B = unsprayed plants, Y = plants exposed to  $SO_2$ , and Z = unexposed plants.  $\square =$  Treatment plants and O = plants used as sources of inoculum.

plants from rows 1 and 4 (fungicide and unsprayed treatments, respectively) in the first replicate (Fig. 1) were placed in a controlled-environment chamber and exposed to a mean daytime concentration of 393  $\mu g^{-3}$  (0.15 ppm) SO<sub>2</sub> for 72 continuous hours at  $24 \pm 0.5$  C,  $77 \pm 5\%$ RH, and 415  $\mu \text{Em}^{-2} \text{sec}^{-1}$  light intensity with a 14-hr photoperiod starting at 0600 hours. The pollutant was injected into the chamber and monitored as described by Biggs and Davis (2). An equal number of plants from rows 2 and 3 was brought indoors simultaneously and exposed to charcoal-filtered air for 72 hr in a matched controlled-environment chamber.

After exposure, plants of the first replicate were returned to their original positions in the outdoor beds and plants of the second replicate were moved into the respective chambers. This procedure was carried out sequentially for the three replicates until 7 July 1981, when four 72-hr SO<sub>2</sub> exposures had been completed for each group, ie, each of three replicates received four 72-hr exposures to SO<sub>2</sub> or charcoal-filtered air once every 6 days. In summary, the four treatments were fungicide plus SO<sub>2</sub>, SO<sub>2</sub> only, fungicide only, and untreated.

Four days after the final  $SO_2$  exposure, one leaf from each plant in replicate 3 was removed for determination of post-exposure sulfur content. A mature leaf was removed at the stem-petiole junction four or five nodes from the soil surface. The unwashed leaf was placed in a paper bag, dried at 80 C for a minimum of 72 hr, and sulfur content was determined using a LECO sulfur analyzer (LECO Corp., Warrendale, PA) connected to an automatic titrator (8).

The atmospheric spore concentration was estimated by counting spores in the center of the plot with a battery-operated Rotorod Spore Sampler (Ted Brown Associates, Los Altos Hills, CA) with clear acrylic Type I rods coated with silicone gel on the collection surfaces. The sampler was operated for 2 or 3 days per week from 14 July to 6 August 1981. Rods were placed on a microscope slide and trapped spores were counted at ×100.

On 5, 12, and 19 August 1981, fruit in the "turning" to "red" stages (USDA Visual Aid TM-L-1, John Henry Co., Lansing, MI) were harvested from all plants and counted and weighed. Regardless of color, fruit with a diameter greater than 3 cm were denoted as "acceptable size" fruit; fruit with diameters less than 3 cm were termed "culls." On 19 August, remaining acceptable-size green fruit were harvested, counted, and weighed; culls were harvested separately, counted, and discarded. On 20-22 August, foliage and vines were weighed, then dried at 100 C for 7-10 days for dry-weight determination.

Analysis of variance in a general linear

models program (SAS Institute Inc., Cary, NC) was used to determine if fruit yield or tissue mass were affected by fungicide treatments, SO<sub>2</sub> exposures, or the interaction between the two variables.

#### RESULTS

Disease development. Seven to 10 days after inoculation, plants to be used as sources of airborne inoculum developed small dark pinpoint lesions on the most mature foliage. The lesions expanded into the dark concentric rings typical of early blight symptoms on tomato. As the season progressed, lesions increased in number and size and defoliation was extensive. Unsprayed treatment plants showed similar symptoms. Numbers of spores gradually increased in the experimental plot during late July, reaching an apparent peak of 70-100 spores/10<sup>4</sup> L during 1 and 2 August.

Sulfur analysis. Foliage exposed to  $SO_2$  had significantly (P < 0.01) greater sulfur accumulations (1.31% sulfur) than leaves of unexposed plants (0.68-0.82% sulfur). Fungicide applications did not alter the sulfur status of foliage. Also, SO<sub>2</sub> exposures did not result in visible foliar injury.

Biomass. Fungicide applications resulted in significantly (P < 0.05) greater leaf mass but did not influence vine weight or total plant weight (Table 1). SO<sub>2</sub> exposures did not alter plant biomass. Statistically significant interactions between fungicide application and SO<sub>2</sub> exposures were not evident for biomass measurements.

Fruit yield. Number and weight of acceptable-size fruit were significantly (P <0.05) greater on fungicide-treated plants (Table 2). Fungicide-treated plants also had significantly (P = 0.01) greater numbers and weight of green fruit at harvest. However, the yield of "turning" to "red" fruit harvested over the 3-wk period before final harvest was not affected by fungicide treatment. Fungicide application likewise did not affect the number of culls per plant nor the mean individual weight of acceptable-size fruit per plant.

SO<sub>2</sub> exposures did not affect fruit yield parameters (Table 2). However, significant (P = 0.05) interactions between fungicide applications and SO<sub>2</sub> exposures were observed for the total number of acceptable-size fruits per plant, number and weight of green fruits per plant, and the mean individual fruit weight per plant. In all cases, there were no significant differences between means of fungicide treatments in the absence of

Table 1. Effect of fungicide and SO<sub>2</sub> singly and in combination on tomato plant biomass

Treatment		Leaf weight (g) <sup>w</sup>		Vine weight (g)		Total weight (g)	
Fungicide	SO <sub>2</sub>	Fresh	Dry	Fresh	Dry	Fresh	Dry
+		122.6 a*	18.8 a	199.3	31.1	321.9	49.9
_		104.2 b <sup>x</sup>	15.8 b	194.5	29.0	298.7	44.9
	+	114.7 <sup>y</sup>	17.5	200.6	30.1	315.3	47.7
	-	112.3 <sup>y</sup>	17.1	193.3	30.0	305.6	47.1
+	+	124.9 <sup>z</sup>	19.4	205.5	30.8	330.5	50.2
+	_	119.5 <sup>2</sup>	18.3	192.5	31.2	312.1	49.4
_	+	105.2 <sup>2</sup>	16.0	196.7	29.3	301.9	45.4
	_	104.4 <sup>z</sup>	15.9	194.0	28.8	298.4	44.7

<sup>&</sup>lt;sup>v</sup> Fungicide = chlorothalonil, 15 ml/3.8 L H<sub>2</sub>O; + = treatment given, − = treatment not given.

Table 2. Effect of fungicide and SO<sub>2</sub> singly and in combination on tomato fruit yield

Treatment		Green fruit/plant*		Turning-red fruit/plant <sup>w</sup>		Total acceptable sized fruit/plant <sup>w</sup>		Culls/plant*	Mean individual fruit wt/plant (g)
Fungicide	SO <sub>2</sub>	No.	Wt (kg)	No.	Wt (kg)	No.	Wt (kg)	(no.)	(not inc. culls)*
+		17.5 a <sup>x</sup>	1.053 a	11.9 a	1.037 a	29.4 a	2.087 a	6.5 a	71.0 a
****		14.8 b <sup>x</sup>	0.922 ь	12.2 a	1.055 a	27.0 b	1.975 b	6.6 a	73.2 a
	+	16.0 a <sup>y</sup>	0.964 a	12.1 a	1.061 a	28.1 a	2.025 a	7.2 a	72.2 a
	_	16.3 a <sup>y</sup>	1.011 a	12.1 a	1.026 a	28.4 a	2.037 a	5.9 a	71.8 a
+	+	18.3 a <sup>z</sup>	1.082 a	12.1 a	1.037 a	30.4 a	2.121 a	7.1 a	69.0 b
+	_	16.7 b <sup>2</sup>	1.028 a	11.8 a	1.023 a	28.5 a	2.051 a	6.0 a	72.3 ab
_	+	13.8 cd <sup>2</sup>	0.859 ь	12.1 a	1.088 a	25.9 b	1.944 a	7.5 a	75.5 a
_	_	16.0 bc <sup>2</sup>	0.998 ab	12.1 a	1.017 a	28.2 a	2.015 a	5.9 a	71.3 ab

Fungicide = chlorothalonil, 15 ml/3.8 H<sub>2</sub>O; + = treatment given, - = treatment not given.

SO<sub>2</sub>. In the presence of SO<sub>2</sub>, fungicide treatment was a significant (P = 0.05)variable. Plants exposed to SO<sub>2</sub> but not treated with fungicide had the lowest number of acceptable-size fruit and the lowest number and weight of green fruit but produced the highest mean individual fruit weight per plant.

# DISCUSSION

There was adequate inoculum in the experimental plot to cause early blight on the treatment plants. This was reflected by the significantly higher leaf weight of fungicide-treated plants. Levels of A. solani spores trapped in the plot during late July to early August (see [9] for additional data) were comparable to those in unsprayed tomato plots at our experimental farm in Rock Springs, PA (11).

As expected, SO<sub>2</sub> exposures resulted in significant accumulations of sulfur in the foliage. The sulfur levels, however, were within the "intermediate" range of values used to indicate sulfur status in tomato plants; sulfur levels in plants not exposed to SO<sub>2</sub> were in the "low" range (3).

Significant interactions were observed between fungicide treatments and SO<sub>2</sub> exposures for measurements of fruit yield but not foliar weight. This may indicate that the effects of SO<sub>2</sub> on early blight were indirect. A direct effect of SO2 on the pathogen would have resulted in an alteration of disease symptoms, namely defoliation; this did not occur. The interaction between SO<sub>2</sub> stress and increased plant susceptibility to early blight resulted in a 15% decrease in the number of acceptable-size fruit. There are two possible perspectives on this interaction: 1) the number of acceptablesize fruit was significantly affected by fungicide treatment (ie, two levels of disease severity) in the presence of SO<sub>2</sub>, or 2) fruit number was affected by SO<sub>2</sub> only in the absence of fungicide (ie, the presence of disease).

Reduced fruit number may be attributed either to a decrease in the number of flowers produced or fertilized or to an increase in flower abortion. Fruit

 $<sup>^{\</sup>mathrm{w}}$  Values in columns followed by the same letter are not significantly different (P = 0.05) according to Pdiff (Statistical Analysis Systems, Cary, NC); all other means within columns are not significantly different.

<sup>&</sup>lt;sup>x</sup> Values in rows are means of 54 plants in three replicates; SO<sub>2</sub> treatment data were pooled.

y Values in rows are means of 54 plants in three replicates; fungicide treatment data were pooled.

<sup>&</sup>lt;sup>2</sup> Values in rows are means of 27 plants in three replicates.

<sup>&</sup>quot;Values in columns followed by the same letter are not significantly different (P = 0.05) according to Pdiff (Statistical Analysis Systems, Cary, NC).

<sup>&</sup>lt;sup>x</sup> Values in rows are means of 54 plants in three replicates; SO<sub>2</sub> treatment data were pooled.

YValues in rows are means of 54 plants in three replicates; fungicide treatment data were pooled.

<sup>&</sup>lt;sup>2</sup> Values in rows are means of 27 plants in three replicates.

abortion was not observed at any time during the experiment. There are several reports that SO<sub>2</sub> inhibits photosynthesis (12,15,17,19). It is possible that reduced photosynthate in conjunction with increased disease and defoliation resulted in increased flower abortion or decreased flower production. Decreasing photosynthate necessary for fruit development or limiting other resources could induce these effects (16).

Significant interactions were not observed regarding total fruit weight. This may be attributed to a significant (P = 0.05) increase in mean individual fruit weight of plants that had a decrease in fruit number (Table 2). Removing developing fruit may lead to an increase in size attained by the remaining fruit (4.5).

Yield measurements may not be sensitive enough to measure the effects of low levels of ambient SO<sub>2</sub>. The results presented in this paper indicate that tomato fruit production can be affected by combined stresses of SO<sub>2</sub> exposure and early blight. This may reflect an increased need for careful disease monitoring and control measures in areas of high ambient SO<sub>2</sub>. Environmental factors in central Pennsylvania during the

summer of 1981 were considered conducive to average early blight incidence (10). Environmental conditions favoring higher than average early blight levels may increase the potential for interaction between SO<sub>2</sub> and disease to decrease yields.

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