

## Response of Pinto Bean to Simultaneous Exposure to Ozone and PAN

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### ABSTRACT

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Ten-day-old pinto bean plants were exposed for 4 hr to either 588  $\mu\text{g}/\text{m}^3$  (0.30 ppm) ozone, 247  $\mu\text{g}/\text{m}^3$  (0.05 ppm) PAN, or the two pollutants combined at these concentrations. Ozone induced adaxial fleck whereas PAN produced mainly abaxial bronzing on the first trifoliolate leaf. Plant response to the combined pollutants was either

additive or synergistic on the adaxial leaf surface and antagonistic on the abaxial leaf surface. The antagonistic abaxial response resulted in a nearly complete suppression of symptoms. The percentage injury for the combined leaf surfaces indicated an overall antagonistic response to the pollutants.

*Additional key words:* *Phaseolus vulgaris*, air pollution.

Ozone ( $\text{O}_3$ ) and peroxyacetylnitrate (PAN) induced a synergistic increase in symptom severity on a clone of hybrid poplar (4), even though the clone was resistant to PAN. This study was initiated to examine the interaction of ozone and PAN on a plant species susceptible to both pollutants.

Pinto bean (*Phaseolus vulgaris* 'Pinto 111') is sensitive to both ozone (1, 3) and PAN (7) and produces a distinctive response to each pollutant. Ozone produces adaxial fleck, and PAN produces primarily abaxial bronzing. The spatial separation and visual distinctiveness of the symptoms on pinto bean are useful characteristics in evaluating the pollutant interaction.

### MATERIALS AND METHODS

Pinto bean seeds were planted in perlite on three consecutive days to provide plant material of the same chronological age for three sequential exposures. One day after emergence, the plants were transplanted into individual pots containing a steam-treated mixture of peat, perlite, and soil (1:1:1, v/v). After transplanting, all plants were maintained in growth chambers at 23 C, 75% RH with a 14-hr photoperiod of 25 klux light intensity. Preliminary exposures had shown that the first fully expanded trifoliolate leaf on plants 10 days old from emergence was sensitive to both ozone and PAN. Therefore, the plants were exposed at this age.

The pollutant production and monitoring procedures

were as previously described (4). The environmental conditions during exposure were the same as those at which the plants had been grown. All exposures were 4 hr in duration and commenced at approximately 1000 hours, after the plants had received at least 4 hr of light. Plants were exposed to either 588  $\mu\text{g}/\text{m}^3$  (0.30 ppm) ozone, 247  $\mu\text{g}/\text{m}^3$  (0.05 ppm) PAN, or the two combined at these concentrations. Twenty plants were utilized in each exposure. The plants were selected from the uniform age group on the basis of uniformity of height and foliar development. The series of three exposures (ozone, PAN, and combined) was conducted on consecutive days and replicated five times. A total of 300 plants were exposed in the study. Control plants were maintained in another growth chamber at the same environmental conditions during the exposure. After the exposure, all plants were returned to growth chambers.

The first trifoliolate leaf on each plant was evaluated for injury 3 days after exposure. The adaxial and abaxial surfaces of each leaflet were evaluated separately. The symptom types and the percentage of the leaflet surface affected by each type were recorded. The procedure used to determine the percentage of injured leaf surface was the two-factor system previously described (4). The adaxial and abaxial evaluations of the leaflets were used to calculate an average percent plant injury and treatment injury for each leaf surface. A treatment injury level for the total leaf surface was obtained by averaging the adaxial and abaxial injury levels. The statistical procedure employed to evaluate the plant response to the combined pollutants was the contrast analysis technique previously described (4). The significance of the interaction was determined with an F-test ( $P = 0.05$ ).

## RESULTS

The macroscopic symptom types produced by the individual pollutant exposures were those commonly associated with each pollutant on bean foliage. Ozone produced a light tan fleck on the adaxial leaf surface whereas PAN produced primarily abaxial bronzing with some light tan bifacial necrosis. The symptoms produced by the combined pollutants were restricted mainly to the adaxial leaf surface with a nearly complete suppression of symptoms on the abaxial surface. The adaxial symptom produced by the combined pollutants was a fleck similar in appearance to that produced by ozone, but slightly more yellow. In the one replication where abaxial symptoms were produced in response to the combined pollutant exposure, the symptom was a bronzing similar to that produced by PAN.

Three of five replications indicated a synergistic response on the adaxial surface from the combined pollutant exposures, whereas the response was additive in the other two replications (Table 1). Injury levels on the abaxial leaf surface indicated an antagonistic response in all five replications. When the combined leaf surface response was evaluated, all of the replications indicated an antagonistic response.

## DISCUSSION

The responses of pinto bean to ozone and PAN were easily differentiated visually and separated spatially, and

TABLE 1. Visual evaluations of the percentages of foliar injury on the primary leaves of pinto bean produced by 4-hr exposures to either 588  $\mu\text{g}/\text{m}^3$  (0.30 ppm) ozone, 247  $\mu\text{g}/\text{m}^3$  (0.05 ppm) PAN or the two pollutants combined at these concentrations

Rep.	Leaf surface	Injury <sup>a</sup>			Response <sup>b</sup>
		Ozone (%)	PAN (%)	Ozone/PAN (%)	
1	Adaxial	20 ± 2	4 ± 1	25 ± 3	NS
	Abaxial	0	66 ± 6	0	ANT
	Combined	10 ± 1	35 ± 4	13 ± 2	ANT
2	Adaxial	12 ± 2	13 ± 4	24 ± 3	NS
	Abaxial	0	57 ± 8	4 ± 1	ANT
	Combined	6 ± 1	35 ± 6	14 ± 2	ANT
3	Adaxial	25 ± 3	3 ± 1	42 ± 4	SYN
	Abaxial	0	35 ± 6	0	ANT
	Combined	12 ± 1	18 ± 3	21 ± 2	ANT
4	Adaxial	21 ± 3	5 ± 1	45 ± 2	SYN
	Abaxial	0	53 ± 7	0	ANT
	Combined	10 ± 1	29 ± 4	22 ± 1	ANT
5	Adaxial	28 ± 2	4 ± 1	45 ± 4	SYN
	Abaxial	0	23 ± 7	0	ANT
	Combined	14 ± 1	13 ± 4	22 ± 2	ANT

<sup>a</sup>The mean and standard error of the percentage of visible foliar injury for each treatment, based on 20 plants per exposure.

<sup>b</sup>Response was evaluated for each leaf surface. NS = no significant interaction at  $P = 0.05$ ; ANT = antagonistic interaction; SYN = synergistic interaction.

provide a unique opportunity to evaluate the pollutant interaction. The combined pollutant exposures produced an ozone-type symptom on the adaxial leaf surface. The predominance and accentuation of the adaxial ozone-type response has been observed in interaction studies with ozone and sulfur dioxide (2, 5, 6, 8, 9, 10) and in an interaction study with ozone and PAN on hybrid poplar (4).

The results of the statistical evaluations of the adaxial leaf-surface response to the combined pollutants were not consistent enough to allow a general conclusion to be made concerning their combined effects. In contrast, the antagonistic response on the abaxial leaf surface was very consistent. In four of five replications, the combined pollutant exposures did not produce abaxial injury while in one replication the incidence of bronzing averaged 4%. Bronzing induced by PAN alone ranged from 23 to 66%.

Although the concentrations of pollutants employed in this study may be slightly higher than those found in the urban photochemical complex, these results indicate that phytotoxic concentrations of PAN may interact with ozone and result in the suppression of the characteristic abaxial bronzing associated with PAN injury on certain species of vegetation.

Evaluation of the pollutant interactions were based on the estimated percentage of visible symptoms on the adaxial, abaxial, and combined leaf surfaces. When an evaluation was performed for either the adaxial or abaxial surface, the comparison was being made within a similar symptom type; either fleck on the adaxial or bronzing on the abaxial surface. Because the symptoms were similar, they could be compared quantitatively. However, when the adaxial and abaxial injury levels were used to calculate an injury level for the combined leaf surfaces, comparisons within similar symptom types were no longer possible. Although these comparisons can be made strictly on the basis of the percentage of the leaf surface affected, the physiological impact of equal percentages of fleck and bronzing may be different. For these reasons, the final evaluation of the ability of ozone and PAN to interact may require an injury evaluation system which is independent of macroscopic symptom description.

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