Ozone Toxicity in Tomato as Modified by Phosphorus Nutrition

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ABSTRACT

Ozone phytotoxicity in Rutgers tomato increased with increase in phosphorus supply, and hence with the phosphorus content of tomato foliage. Exposure to ozone was followed by increased foliar contents of phosphorus, potassium, calcium, and total nitrogen at all levels of phosphorus applied and a decrease in total carbohydrate and starch content in the foliage of low phosphorus plants. It is suggested that the carbohydrate reserve accumulated in phosphorus-deficient plants may partially explain the resistance of such plants to ozonation. Phytopathology 60:1521-1524.

Brewer et al. (3) found that increases in phosphorus supply from 20 to 150 ppm without potassium at medium and high-nitrogen ranges resulted in poor top growth somewhat resistant to oxidant injury, but when both phosphorus and potassium were added, top growth and severity of injury were significantly increased. When added phosphorus increased growth, it also caused increased susceptibility to oxidants. Brennan et al. (2) reported similar results with respect to phosphorus nutrition when tomato plants were fumigated with hydrogen fluoride. Whereas it appeared that optimum nitrogen and calcium nutrition levels favored fluoride injury in tomato, injury was more severe in plants having a phosphorus supply greater than optimum. Apparently, phosphorus was able to mobilize fluoride within the plant. Later Leone et al. (6) reported that tobacco plants receiving optimal supply of nitrogen were more susceptible to ozone injury than those receiving a deficient supply, probably because of excess carbohydrates in nitrogen-deficient plants. This study was initiated to determine the effect of increasing levels of phosphorus on the response of tomato plants to ozone fumigations.

MATERIALS AND METHODS.—Tomato seeds (*Lycopersicon esculentum* L. 'Rutgers') were sown in vermiculite and 11 days later, after the appearance of the first true leaves, uniform seedlings were transplanted to washed quartz sand, one plant/pot to be treated according to the experimental plan in Table 1. They were flushed daily for 5 days with a dilute nutrient solution, after which time they were supplied with a complete standard nutrient solution in which the phosphorus was varied (1.5, 15.5, and 62 ppm) to cover a range from deficiency to luxury supply. Nutritional treatment was continued for 1 month or just prior to inflorescence when the plants were in a 7- to 9-leaf stage, at which time duplicate plants from each nutrient treatment were subjected to 3-hr ozone fumigations of varying concn. The fumigations were conducted in a glass chamber 6 × 6 × 8 ft at concn of 0.15, 0.18, 0.22, 0.25, 0.28, 0.30, and 0.45 ppm O_3.

Since the number of replicates in each fumigation was limited by the available space on a turntable used to ensure uniform exposure, twelve experimental fumigations were conducted, four in spring, six in summer, and two in fall.

Following treatment, the plants were observed for ozone injury during a 24 to 48-hr period. All plants continued to receive their original nutrient supply until harvest. All leaves on each plant were rated for ozone symptoms as follows: 0 = absence of ozone symptoms; T (trace) = injury involving less than 15% of the leaf; Sl (slight) = injury involving 15 to 25% of the leaf; M (moderate) = injury involving 30 to 65% of the leaf; and S (severe) = injury involving 70% or more of the leaf. An average rating for each plant was calculated. After the fresh wt of total plant and leaf portions was recorded, the leaves were minced, dried overnight in a forced draft oven at 21 C, and ground through a 40-mesh screen in a semimicro Wiley mill for subsequent chemical analyses.

The ozone was evolved by passing a metered stream of pure dry oxygen through a commercial ozone generator (Instrument Development Co.). The resulting ozone was dispersed into a charcoal-filtered air stream which then passed through a mixing chamber before entering the fumigation chamber. The air in the chamber was exchanged once every 45 sec. Ozone concn was monitored continuously by a Mast Ozone Meter which was compared periodically with determinations made by the potassium iodide titration method (5). The temp in the chamber was kept at 27-30 C, the relative humidity at 50-60%.

Analytical methods.—Phosphorus was determined by the method of the Official Agricultural Chemists (1), and nitrogen by the Pepkowitz & Shive (9) modification of the micro-Kjeldahl method. Potassium and calcium were determined by means of a Coleman Flame Photometer, carbohydrates by the semimicromethod of Wildman & Hansen (11), and starch by the method of Nielson (8). Results were expressed as per cent of dry wt, and were analyzed for statistical significance by determining the standard error of the difference between means using Fischer's Table of t values.

RESULTS.—*Effect of phosphorus supply on growth of tomato.*—After 25 to 30 days of treatments at varied phosphorus levels, the foliage of the low phosphorus (1.5 ppm) plants exhibited typical phosphorus deficiency symptoms, dark green with a purplish tinge on the lower surface. Plants grown at the opt phosphorus level (15.5 ppm) had normally green foliage and were larger than the former, while those grown at the luxury level (62 ppm) were lighter in color but showed little or no further increase in size. Figure 1 presents the

TABLE 1. Molecular concn of nutrient solutions[a] for varied phosphorus nutrition of tomato plants prior to ozone treatment

Nutrient salts	*P* levels		
	1.5 ppm	15.5 ppm	62 ppm
$Ca(NO_3)_2$	.004	.004	.004
$MgSO_4$	.002	.002	.002
KH_2PO_4	.000048	.00048	.002
KCl	.001952	.00152	

[a] Trace elements supplied in the following forms: B, 0.10 ppm (H_3BO_3); Fe, 1.00 ppm ($FeSO_4$); Mn, 0.25 ppm ($MnCl_2$); Zn, 0.10 ppm ($ZnSO_4$); Cu, 0.01 ppm ($CuSO_4$); and Mo, 0.01 ppm (Na_2MoO_4).

green wt of plant tops plotted against phosphorus content of the nutrient supply. Slight variations in growth during different seasons were nonsignificant, due in part to the low light intensity during the month of July when heavy rainfall caused the light intensity to be reduced by about 25%. The point of the most economical phosphorus utilization, hence the optimum level, occurred at 15.5 ppm phosphorus.

Phosphorus content of composite leaf samples was directly proportional to phosphorus concn in the nutrient solution in both seasons (Fig. 2). The slight variation in phosphorus content between the spring- and summer-grown plants was normal and nonsignificant. There was, however, an increase in phosphorus content in both seasons as a result of ozone treatment. This increase was not statistically significant in the low or opt phosphorus plants, but significant at $P = .05$ in plants grown with the highest phosphorus supply.

Effect of phosphorus nutrition on ozone phytotoxicity.—Injury of tomatoes grown in summer at increasing phosphorus levels and exposed to 3-hr fumigations was more severe at increasing ozone concn (Table 2). In addition, within each ozone exposure, injury was increased as the phosphorus supply was increased (Fig. 3). Even at ozone concn above 0.22 ppm, where opt and luxury level phosphorus plants were severely injured, the low phosphorus plants did not show more than slight injury (15-20% necrosis).

Injury was generally more severe in summer than during spring and fall (Table 3). Symptoms also varied according to the phosphorus nutrition of the plant. Whereas opt and luxury level phosphorus plants tended to exhibit injury as a tan necrosis on the upper surface of the older true leaves, injury to low phosphorus plants consisted of a barely perceptible dark fleck at the upper surface of leaves.

TABLE 2. Degree of injury caused by 3-hr ozone exposures to foliage of tomatoes grown at increasing phosphorus levels

P level (ppm)	Ozone concn (ppm) and phytotoxicity rating[a]					
	0.15	0.18	0.22	0.28	0.30	0.45
1.5	0	T	Sl	Sl	Sl	Sl
15.5	0	Sl	M	S	S	S
62.0	Sl	M	S	S	S	S

[a] 0 = none; T = trace; Sl = slight; M = moderate; S = severe.

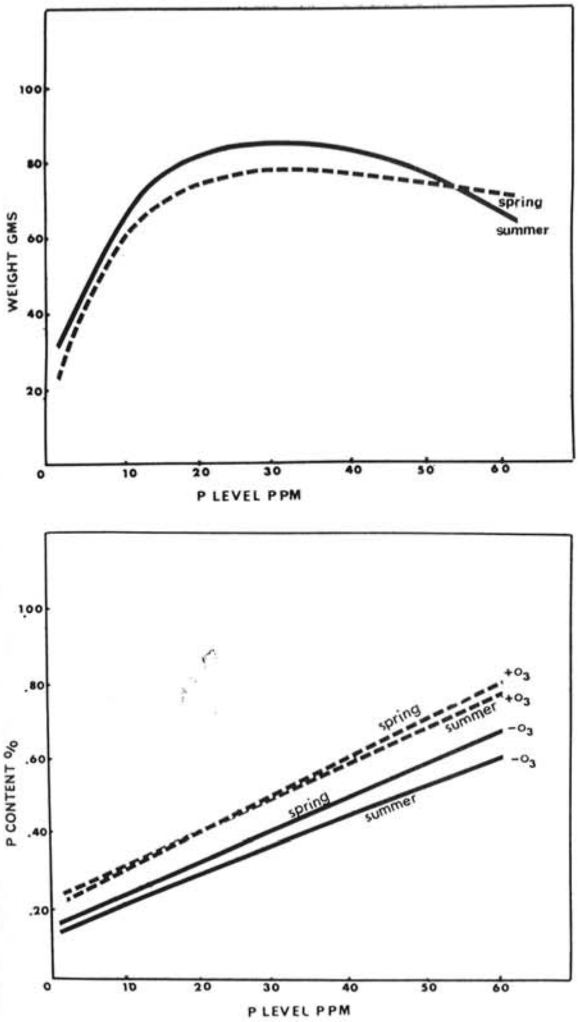

Fig. 1-2. **1)** Green wt of tops of tomato plants grown at increasing phosphorus levels. **2)** Per cent total phosphorus in composite leaf sample of ozonated and control tomatoes grown at increasing phosphorus levels in spring and summer. Increase in phosphorus was significant at the $P = .05$ level in high phosphorus plants after ozonation.

Effect of phosphorus supply and ozone treatment on chemical constituents of tomato foliage.—Analyses of leaf samples from each plant (Table 4) indicated the usual decrease in nitrogen content with increasing phosphorus increments. As a result of ozone fumigation, there was an increase in foliage nitrogen at all phosphorus levels.

Total hydrolyzable carbohydrates also showed a decrease with an increase in phosphorus supply above the optimum. As a result of ozone fumigation, there was a significant decrease in total carbohydrate and starch content ($P = .01$) in the low phosphorus plants.

Data in Table 4 indicate a decrease in potassium content with increase in phosphorus nutrition, and an in-

TABLE 3. Degree of injury caused by 3-hr ozone exposures to foliage of tomatoes grown at increasing phosphorus levels during different seasons

	Ozone concn and phytotoxicity rating[a]						
	0.15 ppm		0.25 ppm			0.30 ppm	
P level (ppm)	Spring	Summer	Spring	Summer	Fall	Spring	Summer
1.5	0	0	T	Sl	T	T	Sl
15.5	0	0	M	M	M	T	S
62.0	Sl	M	M	S	M	M	S

[a] 0 = none; T = trace; Sl = slight; M = moderate; S = severe.

TABLE 4. Total nitrogen, total carbohydrate, starch, calcium, and potassium content[a] of untreated and ozone-treated foliage of tomatoes grown at increasing phosphorus levels[b]

P level (ppm)	Treatment	Total N %	Total Carbohydrate %	Starch %	Potassium %	Calcium %
1.5	Control	2.95	47.3	13.25	2.15	1.45
1.5	Fumigated	3.45[d]	26.8[c]	4.09[c]	2.70	1.84
15.5	Control	2.26	49.7	22.00	0.97	1.25
15.5	Fumigated	2.62	38.0	12.07[c]	1.55	1.48
62.0	Control	2.10	42.3	16.25	0.80	1.43
62.0	Fumigated	2.65[d]	40.5	16.00	1.18	1.83

[a] Per cent of dry wt.

[b] Each figure is the average of four fumigation experiments at varied ozone exposures.

[c] Difference between control and fumigated foliage significant at $P = .01$.

[d] Difference between control and fumigated foliage significant at $P = .05$.

crease in both calcium and potassium in tomato foliage at all phosphorus levels as a result of ozone fumigation.

DISCUSSION.—These results agree in part with previous work done by Brewer et al. (3). Phosphorus deficiency generally causes increased accumulation of carbohydrates in young plants, as does nitrogen deficiency (4, 7). In our experiments, plants containing low levels of phosphorus did not necessarily contain a greater percentage of total hydrolyzable carbohydrate or starch than plants containing the optimal level, but the actual content may have exceeded that required by plants with an obviously low rate of metabolism. The decrease in carbohydrate, however, reported for low nitrogen plants exposed to ozone (6) also resulted from ozone-exposure of low phosphorus plants. The decrease was highly significant in these plants (at $P = .01$), whereas plants supplied opt or luxury amt of phosphorus showed no appreciable carbohydrate decrease on being fumigated. This trend was also observed with respect to starch content.

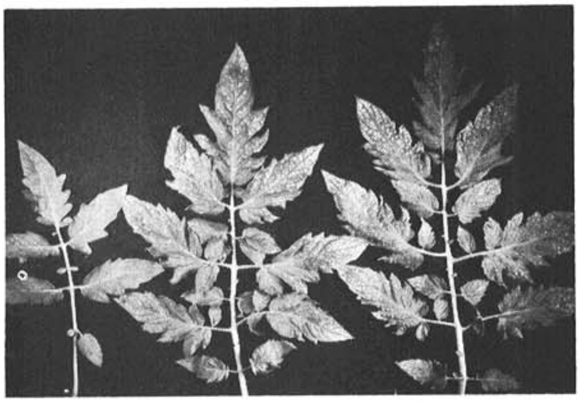

Fig. 3. Effect of increasing phosphorus on ozone toxicity. Fourth oldest true leaf, left to right, of plants grown at 1.5, 15.5, and 62 ppm phosphorus.

Avoidance of the tendency to overfertilize tomatoes in the seedling stage might tend to offer protection from ozone injury during growth stages prior to inflorescence. In an earlier paper, Leone & Shive (7) reported that tomato seedlings low or deficient in phosphorus were more able to undergo the shock of transplanting than were those on an opt or luxury phosphorus supply. Again, there was a decrease in carbohydrate content coincident with the ability of the plant to withstand the unfavorable treatment. Apparently, an available carbohydrate reserve serves to enable the plant to survive a variety of adverse conditions. This carbohydrate reserve may be obtained by withholding either nitrogen or phosphorus, as has been shown in the previous experiments.

The effect of phosphorus deficiency on stomatal opening may provide a further explanation for the protection offered by low phosphorus plants to ozone injury. Wallace & Frolich (10) have reported that stomata failed to open on old leaves of plants from which phosphorus was withheld. Porometer readings during our experiment have corroborated these findings, and further investigation of the role of stomata in air pollution injury to plants is being conducted.

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