# The Effect of Photoperiodic Pretreatments on Symptom Development in Plants Exposed to Ozone

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## Introduction

Plant response to ozone is dependent on the physiological condition of plants during exposure. This physiological condition depends upon pre-exposure environmental growth factors which include temperature, light intensity, soil moisture, relative humidity and nutrition (2, 3, 6, 7). It also depends on biological rhythms which are endogenous or cued by external environmental factors such as photoperiod. The role of photoperiod in cueing the flowering response in many plant species has been well documented. Less well known are changes in other physiological processes which may result prior to or in the absence of the flowering response. It was the object of this study to determine whether photoperiodic pretreatments induced physiological changes which affected sensitivity and symptom expression in plants subsequently exposed to ozone.

## Materials and Methods

Seedlings of tomato *(Lycopersicon esculentum)* cultivar Rutgers and peas *(Pisum sativum)* cultivar Alaska were planted in 10 cm diameter plastic pots containing a soil medium of loam, peat and perlite (1:1:1) supplemented with a 12:12:12 water soluble fertilizer. These seedlings were grown in an environmental chamber with a 27/17°C day/night temperature regime. Relative humidity was uncontrolled, ranging from 60 to 80% during the light period and 80 to 100% during the dark period. A light intensity of 8.93 x $10^4$ ergs $cm^{-2}$ $sec^{-1}$ at the plant surface was provided by cool white fluorescent and incandescent lamps. Tomato plants were grown to the second nine-foliate stage prior to being exposed. They were refertilized with a 12:-12:12 granular fertilizer mix 2 weeks after transplanting. Pea plants were grown to the six-leaf stage prior to being exposed. Because of the short growth time to reach this stage of development, pea plants were not refertilized prior to exposure.

Five photoperiodic pretreatment regimes were employed. These included 8, 10, 12, 14, and 16 hours of light. In each case the themoperiod corresponded to the photoperiodic regime.

Plants of similar physiological age were used for exposures. Physiological age was determined by leaf and leaflet expansion in tomato and by the number of fully mature leaves in peas.

Plants of the appropriate physiological age were exposed to 35 pphm (v/v) $0_3$ for 4 hrs., from 10:00 a.m. to 2:00 p.m. in 10 cubic feet volume plexiglass chambers. In tomato exposures, four plants of a given photoperiodic pretreatment were exposed at a given time. The number of plants exposed was limited by chamber area and volume.

Each exposure was repeated so that a total of eight plants from a photoperiodic pretreatment were exposed to ozone. Since individual pea plants are smaller in size, 3 seeds were planted per pot. Depending on germination percentage, as many as 29 pea plants were exposed to ozone at one time. For peas only one ozone exposure was conducted for a given photoperiodic pretreatment.

Air flow through the chambers was approximately 15 cubic feet per minute. During exposures, chamber temperature was maintained at 27±1° C and a relative humidity of 70±2%. Light intensity of $3.56 \times 10^4$ ergs $cm^{-2}$ $sec^{-1}$ was provided by cool white fluorescent and incandescent lamps above the exposure chamber. Ozone was metered to the exposure chamber from an Alron high voltage ozone generator. Ozone concentrations were monitored during exposure by a Mast oxidant meter calibrated with the Potassium Iodide-Boric Acid Method (4).

After exposure plants were returned to the environmental chamber. Symptom development was evaluated 96 hours after exposure. Symptom severity was determined by the % leaf area exhibiting flecking, mottling and bifacial necrosis. Data were evaluated for significant differences using the Completely Randomized Design Analysis of Variance and Duncan's Multiple Range Test. An alpha level of .05 was accepted as significant.

**Results**

In both tomato and pea cultivars significant differences in ozone-induced symptoms were observed in plants receiving different photoperiodic pretreatments. The effect of these pretreatments on symptom severity on tomato and pea plants are summarized in Table 1. Several important conclusions can be drawn from these data. For tomato plants the 12 hr. photoperiodic pretreatment resulted in greater leaf injury than any other treatment. The 8-hour photoperiodic pretreatment, on the other hand, resulted in the least leaf area injured. For pea plants the most extensive injury was observed with the 14-hour photoperiodic pretreatment. Plants were least sensitive to 8 and 10 hour photoperiodic pretreatments. These observations are significant at the .05 alpha level.

In addition to differences in symptom severity observed on tomato plants, differences in symptom expression were apparent. For the 12-hour photoperiod the dominant symptom type was bifacial necrosis. Necrotic

TABLE 1. *Response of tomato cv. Rutgers and pea cv. Alaska to ozone following photoperiodic pretreatments.*

| Photoperiod (Hrs.) | % Leaf Area Injured Tomato | Pea |
|---|---|---|
| 8 | <1.0 c | 7.6 c |
| 10 | 18.2 a | 3.7 c |
| 12 | 34.5 b | 21.1 a |
| 14 | 12.5 a | 40.4 b |
| 16 | 10.0 a | 16.9 a |

Treatment means followed by the same letter are not significantly different at the 5% level.

areas tended to be continguous along the midrib and interveinal leaflet areas. For the 10, 14, and 16-hour photoperiods the dominant symptoms were flecking and mottling with occasional small pinpoint bifacial lesions. An interesting symptom expression was manifested in tomato plants pretreated with a 16-hour photoperiod. Light, diffuse chlorotic islands were observed. These chlorotic islands were located in the interveinal areas where flecking and mottling were normally observed. This apparent loss of chlorophyll was not included in the % leaf area injured since the experimental design was based on symptom severity due to flecking, mottling, and bifacial necrotic injury.

Flecking, mottling and bifacial necrosis were also observed on pea plants exposed to ozone. Although bifacial necrosis was more extensive on plants pretreated with a 14-hr. photoperiod, differences in qualitative symptom expressions similar to those observed on tomato were not apparent.

**Discussion**

Results presented in this report on the effects of photoperiodic pretreatments on ozone sensitivity of plants are significantly different from the lone previous report on this subject. Juhren *et al* (5) studied the effects of photoperiodic pretreatments on sensitivity of pinto beans exposed to oxidants (presumably ozone). They reported that pinto bean plants were most sensitive to oxidants under short photoperiods (8 hours) and least sensitive to long photoperiods (16 hours). In studies of tomato and peas presented in this report, minimum sensitivity for tomato was observed under the 8 hour pretreatment; for peas minimum sensitivity was observed for the 8 and 10 hour pretreatments. Maximum sensitivity for tomato was observed for the 12-hour photoperiodic pretreatment; peas showed maximum sensitivity under the 14-hour photoperiod. The apparent contradiction between the results of the two studies can be readily explained by differences in experimental methods. In this study care was taken to expose plants of the same physiological age, since sensitivity to ozone is known to be strongly influenced by the developmental stage (7). Because of differences in the total quantity of light received under different photoperiods, plants of the same chronological age will differ significantly in the degree of growth and development. This was true in the studies of Juhren *et al.* It is apparent that the results that they reported for photoperiodic pretreatments could not be separated from differences in sensitivity which may have resulted from differences in development.

Differences in sensitivity of tomato and pea plants under various photoperiodic pretreatments may be explained in part by differences in soluble carbohydrate (sucrose, glucose, and fructose) levels prior to exposure. Dugger *et al* (1) have reported that both low and high soluble plant carbohydrate levels were associated with decreased ozone sensitivity. Intermediate soluble carbohydrate levels are optimum for ozone sensitivity. Although soluble carbohydrate levels were not measured in this study, results reported by Dugger *et al* indicate that soluble carbohydrate levels would be lowest under short photoperiods and build up with increasing photoperiodic length. Under these circumstances the low ozone sensitivity observed for tomato and pea plants under short photoperiods and maximum sensitivity at intermediate photoperiodic lengths correlate well with observations relative to soluble carbohydrate levels.

In this study photoperiod could not be programmed independently of the thermoperiod. Consequently results reported for photoperiodic pretreatments have not excluded possible changes caused by thermoperiod. This possibility is of theoretical interest, although in nature photoperiod and thermoperiod are not independent of each other.

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