PHOTOPERIODIC CHLOROSIS IN TOMATO

A L I C E P . W I T H R O W and R O B E R T B . W I T H R O W

(WITH THREE FIGURES)

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Introduction

The leaves of certain plants have been shown to contain less chlorophyll when subjected to continuous artificial radiation than when given shorter photoperiods. GUTHRIE (3) reported that tomato, transferred from the greenhouse to continuous artificial radiation, after a period of several days developed an interveinal chlorosis and a yellowing of the leaves with a decrease in chlorophyll and a consistent lowering of the a/b ratio as compared to plants transferred to an 18-hour photoperiod. There also was a decrease in total carotenoids and in the carotene to xanthophyll ratio. With the decrease in chlorophylls, there was an increase in brown pigments, which he postulated as an indication that the chlorophyll decrease was due to decomposition rather than to decreased synthesis. ARTHUR and HARVILL (2) did further work on plant growth under continuous irradiation, using sodium lamps and combinations of sodium and mercury arcs. They irradiated geranium, cotton, buckwheat and other species. They found that with geranium and cotton, continuous irradiation from the sodium lamp brought about a marked chlorosis, especially of the younger leaves developed under the sodium irradiation. When the plants were given two hours daily of irradiation from a mercury vapor lamp to replace the sodium for this length of time, the chlorosis disappeared. More rapidly growing plants as buckwheat did not develop the chlorosis very markedly.

ROODENBURG (4) reports an interveinal chlorosis in tomato resulting when daylight was supplemented with eight hours irradiation from neon lamps to lengthen the photoperiod. ÅBERG (1) also reports chlorosis in leaves of tomato plants grown under continuous neon irradiation. Neither Åberg nor Roodenburg presents chlorophyll data and neither appears to have secured completely chlorotic leaves as were observed by the Boyce Thompson workers, but the interveinal chlorosis described by them appears to be similar to that observed under continuous artificial irradiation by Guthrie and Arthur and Harvill.

The present investigation was undertaken to ascertain further aspects of the photoperiodic chlorosis in tomato and the interrelationship of photoperiod and temperature on the chlorophyll content. It was also desired to

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determine whether this reaction has any characteristics in common with other photoperiodic responses in higher plants.

Procedure

Indiana Baltimore tomato was used throughout the investigation. The plants were seeded in subirrigation gravel culture in constant-condition rooms. The nutrient solution used consisted of the following salts given in millimolar concentrations: KNO₃, 10; K₂SO₄, 5; CaSO₄, 8; MgSO₄, 4; Ca(H₂PO₄)₂, 2. The solution was applied three times daily. Twice weekly a micronutrient supplement of 1 p.p.m. of Fe as FeSO₄ and 0.3 p.p.m. of Mn as MnSO₄ was added. Other essential micronutrients had been found previously in adequate supply in the gravel and the fertilizer salts used for the macronutrients. The pH was adjusted to six daily. The solution was changed at weekly intervals throughout the investigation.

The indicated temperature was held to within 1°C. Incandescent lamp irradiation, filtered through 10 cm. of water to remove the near infrared, was used for all treatments except for the irradiances of 4 and 20 fc in which case the lamps were unfiltered. For chlorophyll data, one cm² leaf punchings were taken from the youngest mature leaves close to the center of the leaf and including the midrib. Twenty punchings were used per sample and weighed immediately. They then were extracted in a Waring blender for five minutes in 80 per cent. acetone. The acetone extract was filtered through a Büchner funnel with two thicknesses of Whatman No. 1 filter paper. The sample was made up to 100 ml. with 80 per cent. acetone and placed in a 125 ml. flask, corked, sealed with tape, and stored immediately in the dark at 4°C. All samples were taken in triplicate. The absorption coefficients of the extracts were obtained within 48 hours after extraction with the Beckman spectrophotometer, in some cases at intervals from 380 to 680 mμ and in others at 660 mμ when only relative chlorophyll concentrations were desired. The extent of the chlorosis was determined in relation to the following three variables: (1) length of photoperiod, (2) irradiance, and (3) temperature.

Length of Photoperiod

The plants were transplanted 14 days from seeding to the plots for variable treatment. The temperature was held at 20°C. The plants received 325 fc for 15, 18, 21, and 24 hours per day respectively and the radiation variables were begun at the time of transplanting. Prior to this, the plants had been kept in a 15-hour photoperiod at 325 fc. The plants were photographed for figure 1 after 23 days of treatment and the samples for chlorophyll determination taken on the same day.

Irradiance

The plants were seeded, transplanted, the irradiation variables begun and harvest for chlorophyll determination made at the same time as in the
above experiment. The radiation variables consisted of 15 hours per day at 325 fc supplemented with the following four conditions for the remaining nine hours: 0, 4, 20, and 325 fc respectively.

**FIG. 1.** Above: Response of Indiana Baltimore tomato plants to length of photoperiod. Incandescent sources at 325 footcandles were employed with the temperature controlled at 20° C and photoperiods as indicated.

Below: Leaves of tomato become chlorotic in photoperiods longer than 18 hours at a temperature of 20° C.

**TEMPERATURE**

The plants were transplanted to the plots for variable treatments 20 days from seeding. The irradiation and temperature variables were begun 31 days from seeding and harvest for chlorophyll samples was made after 12 days of treatment, 43 days from seeding. Prior to the application of the treatments, the temperature was 18° C, and the irradiation approximately
500 fc with a photoperiod of 15 hours. The four variable treatments included the following conditions of photoperiod and temperature with an irradiance of 325 fc in all cases: (1) 15 hours at 12° C, (2) 15 hours at 24° C, (3) 24 hours at 12° C, and (4) 24 hours at 24° C. The absorption coefficients of the acetone extracts were obtained at 10 to 20 mµ intervals from 380 to 680 mµ.

Results and discussion

The results are presented in figure 1 through figure 3. It can be seen from the photographs in figure 1 that, after 23 days of treatment, the plants were tallest under an 18-hour photoperiod and markedly decreased in height as the photoperiod was further increased. The decrease under the longer photoperiods is probably directly related to a decreased rate of photosynthesis due to the lowering of the chlorophyll content. Under a 21-hour photoperiod, the leaves acquired interveinal chlorotic areas which later developed a brown flecking markedly similar to that occurring in manganese deficiency. A 24-hour photoperiod caused the new leaves frequently to become almost entirely chlorotic, and they were small and definitely thicker than those from the shorter photoperiods as shown by area-weight relationships.

Because of the similarity of the response to certain mineral deficiency symptoms, in one experiment the leaves of plants under a 24-hour photoperiod were sprayed with a water-polyvinyl alcohol solution of the following micronutrient salts at a pH of 5: 0.1 per cent. ferrous ammonium citrate, 0.05 per cent. manganese sulphate, 0.5 per cent. magnesium sulphate, and water alone as a control. In addition a complete micronutrient spray was used consisting of 0.1 per cent. ferrous ammonium citrate, 0.05 per cent. manganese sulphate, 0.05 per cent. boric acid, 0.005 per cent. copper sulphate, 0.01 per cent. zinc sulphate, and 0.001 per cent. molybdenum trioxide. After ten days of treatment with the above solutions, none of the applications had caused any observable effect on the chlorosis. In addition to this, analysis of leaves from 15-hour and 24-hour photoperiods showed little difference in manganese content and there were several indications that in the very chlorotic leaves, a manganese content considerably higher than in the green leaves obtained.

The chlorosis was incipient at about four long photoperiods and quite noticeable by seven long photoperiods. After 14 to 21 long photoperiods, the youngest leaves were often almost entirely chlorotic, and the marbled pattern had disappeared. The older leaves which had developed under 15-hour photoperiods before transfer never showed the marked chlorotic symptoms evidenced by the younger leaves developing under the 21-hour or 24-hour photoperiods.

One pattern of development observed several times was the apparent recovery of the plant while still subjected to a long photoperiod. For three
or four weeks, the young developing leaves were very chlorotic and remained so. However, the leaves developed later than this entirely lacked

![Graph showing chlorophyll concentration vs. photoperiod] (https://www.plantphysiol.org)

**Fig. 2.** Above: Relative chlorophyll concentration of acetone extracts of tomato leaves of plants grown under varying photoperiods of incandescent irradiation. A marked decrease in concentration occurs in photoperiods longer than 18 hours. Data taken at 660 mJ.

Below: Relative chlorophyll concentration of acetone extracts from leaves of plants grown under 15-hour periods at 325 footcandles of incandescent irradiation supplemented for nine hours with 0, 4, 20 and 325 footcandles to achieve a 24-hour photoperiod. A decrease in concentration based on unit area is observed at the lowest supplemental irradiance used.

the chlorosis, although the older chlorotic leaves already present did not recover. At first this was thought to be due to the higher irradiance incident to the plants as they grew closer to the lamps, but in an experiment
where an irradiance of 1500 fc was used, young plants developed the same typical chlorosis under 24-hour photoperiods as previously evidenced at 325 fc. The relative chlorophyll concentrations of the acetone extracts for various photoperiods are shown in the upper graph of figure 2. The curves indicate that the maximum chlorophyll concentration occurred under 18 hours, a marked decrease under 21 and a very low concentration in the 24-hour photoperiod. The trend of the graph is similar whether the data are plotted on a fresh weight or unit area basis, the greatest difference occurring in the 18-hour photoperiod. The relative chlorophyll concentra-

Fig. 3. Absorption spectrum of acetone extracts from plants grown under 15-hour and 24-hour photoperiods and at 12 and 24°C. Data were taken from 380 to 680 μm as indicated. At the low temperature, there was relatively little difference in concentration of extracts from plants grown under the two photoperiods, but at 24°C, the difference was very great.

The relative chlorophyll concentration, on a unit area basis with the irradiance plotted on a log scale, shows a decrease with a 24-hour photoperiod as compared with a 15, even when a relatively low irradiance of 4 fc was used for nine hours to supplement a 15-hour period of 325 fc. On a unit fresh weight basis, no difference in chlorophyll concentration was shown when this low irradiance was used, although a definite mottling of the leaves was present. When an irradiance of 20 fc was used for the nine-hour period, there was a marked drop in chlorophyll concentration and a still further decrease at 325 fc.

The data presented in figure 3 on the interrelationship of temperature and photoperiod indicate that at a temperature of 12°C, photoperiod has
little effect on chlorophyll concentration. At 24° C there was a very great difference in pigment content between the 15-hour and 24-hour photoperiods. The highest concentration occurred in the 15-hour photoperiod at 24° C, with a very marked decrease in the photoperiods with the 12° C temperature and a very low concentration in the 24-hour photoperiod at 24° C. Very little mottling was observed in the plants held at 12° C although the leaves were the pale yellowish green characteristic of tomato plants held at a low temperature.

The results of this series of investigations on the effect of radiation and temperature variables on chlorophyll concentration in the leaves of tomato show several points of similarity to the effect of similar variables on the flowering of plants. First, occurrence of the chlorosis is controlled by the length of the photoperiod. It further occurs in response to a relatively low irradiance applied as a supplement to a short high irradiance photoperiod. In addition, a definite interrelationship between photoperiod and temperature on the development of the chlorosis occurs, with a failure of the chlorosis to appear at relatively low temperatures. Further investigations are underway on wavelength and other radiation variables on the synthesis and decomposition of chlorophyll in relation to photoperiod.

Conclusions

A typical chlorosis in the leaves of tomato appears at photoperiods longer than 18 hours when incandescent lamps are used as the sole source of radiant energy. This chlorosis occurs at 20° C when a 15-hour photoperiod is lengthened to 24 hours with relatively low irradiances of 4 and 20 fc. When the plants are grown at temperatures of 12° C, the length of the photoperiod has relatively little effect on the chlorophyll concentration of the leaves and the typical severe chlorotic pattern developed at higher temperatures under 24-hour photoperiods is absent.

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LITERATURE CITED