

of the endocarp of "fruit outside" in the sunlight was 3.3 to 5°C higher than that of the endocarp of the "shade" fruit (Table 2). The endocarp temperature of the "shade" fruit and the air temperature of the "outside" and "shade" differed by less than 0.8°C. The small differences in temperature of the "outside" fruit and the "shade" fruit were not sufficient to cause a significant change in lycopene content.

The data indicate that lycopene synthesis in the 41.8°C/36.1°C fruit was either 1) inhibited or reduced, 2) or that lycopene was rapidly degraded, or 3) a combination of both. The rate of carotene synthesis during "outside" and "shade" and 41.8°C/36.1°C treatments remained the same. The effect of cooler temperature on the "outside" fruit is shown by the higher concn of carotene in the fruit at the end of the experiment. Previous work (4, 7) demonstrated that cool temperatures increased the rate of carotene synthesis.

Table 2. Average maximum and minimum temperatures.

Days	Fruit Endocarp				Air			
	Outside		Shade		Outside		Shade	
	Max°C	Min°C	Max°C	Min°C	Max°C	Min°C	Max°C	Min°C
0-15	44.0	27.6	38.8	27.7	38.7	27.4	37.9	27.4
15-30	42.5	28.1	38.5	28.6	38.4	28.1	38.5	28.1
30-45	41.4	27.4	37.7	27.4	36.6	27.1	36.9	26.9
45-60	40.4	27.4	36.9	27.4	35.9	27.2	36.4	27.0

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Effect of Light on Cold-Hardening of Citrus Seedlings¹

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Abstract. Cultivars of citrus, having a wide range in cold tolerance, were conditioned in controlled-environment rooms with and without light: Plants conditioned in the light were injured less than plants conditioned in the dark during subsequent freeze tests. The effect of light was more pronounced at a conditioning temperature of 15°C than at 5°C.

Cold weather for a month or more preceding a natural freeze minimizes injury to citrus trees (1, 2). Under artificial conditions, citrus seedlings survive freezing better after exposure to 10°C or lower for a few weeks (9). In controlled environment rooms, continuous light is usually provided by banks of fluorescent and incandescent lights (3). This light environment adds considerably to the total cost and makes temperature control more difficult. Temperature is well-established as the important factor in cold-hardening of citrus trees, but little is known about the need for light in conditioning treatments. This report compares the effect of continuous light and of no light during hardening treatments of citrus seedlings.

The cultivars of citrus tested represent a wide range in cold hardiness.

These were, in order of increasing cold-hardiness: 'Rough' lemon (*Citrus limon* [L.] Burm. f.), 'Duncan' grapefruit (*C. paradisi* Macf.), 'Orlando' tangelo (*C. paradisi* X *C. reticulata* Blanco), 'Valencia' orange (*C. sinensis* [L.] Osb.), 'Rusk' and 'Carrizo' citranges (*C. sinensis* X *Poncirus trifoliata* [L.] Raf.), and trifoliolate orange (*P. trifoliata* [L.] Raf.). Seedlings were grown in metal cans (15.4 cm diam, 16.7 cm ht) filled with a 1:1:2 mixture of sand, vermiculite and peat. Hardening and freezing resistance were tested when seedlings were 10 to 14 months old. Stem diam ranged from 0.35 to 0.55 cm at 8 cm above soil level, and heights ranged from 55 to 90 cm. For each test, seedlings of uniform size and vigor were selected.

All tests were conducted in 3 X 3 X 1.8 m controlled-environment rooms. Two rooms were used simultaneously to condition the seedlings prior to freeze tests in a third chamber. In both conditioning rooms, air temperature was maintained at 15°C ± 0.6° or 5° ± 0.6° and relative humidity (RH) at 60% ± 5%. Air circulation was about 10 linear m/min velocity. One chamber had continuous light and the other no light. Light intensity was approximately 2,000 ft-c at the top of the seedlings, and the spectrum in microwatts per square centimeter per nanometer (μw/cm²/nm) (Fig. 1) was obtained with

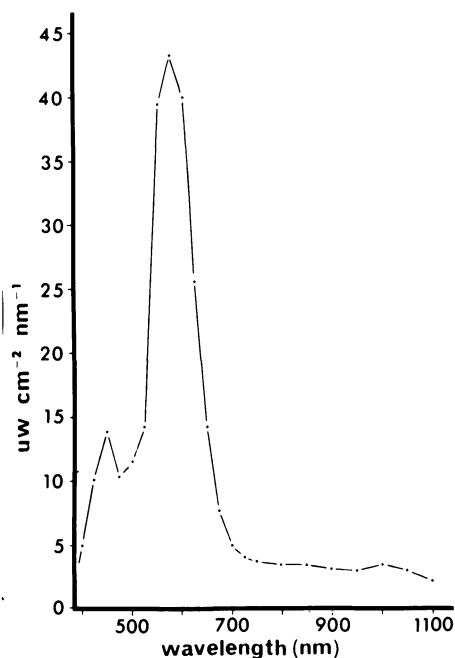


Fig. 1. The light intensity at indicated wavelengths (microwatts/square cm/nanometer) at the top of citrus seedlings exposed to conditioning treatments in controlled-environment rooms. The 40 fluorescent cool-white and 16 incandescent lights were about 1.4 m above the tops of the plants.

a portable spectrophotometer. A group of 7 or more seedlings per cultivar was assigned to one of the 2 conditioning rooms or kept under glasshouse conditions prior to freeze testing. Conditioning treatments were of less than 7 days' duration in this study, although preliminary work indicated that seedlings can tolerate as many as 18 days without light at 5° without serious injury.

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Immediately following conditioning treatments, seedlings were transferred to the freeze room which was preset at 1.7°C, 50% RH, and no lights. After 1 hr, air temperature in the room was decreased 1.2°C/hr to -6.7°C. After exposure to cold, seedlings were returned to glasshouse conditions for 5 weeks before a rating of % defoliation and stem dieback.

The data indicate that light increases cold-hardening. Plants conditioned in the light were injured less than plants conditioned in the dark (Table 1). However, light had a greater effect at 15°C than at 5°C. At 5°C, considerable hardening occurred in the plants kept in darkness. Better hardening developed when the plants were kept in light and at low temperature. Increased hardening in light has been postulated (4, 5) to be the result of sugar accumulation from photosynthesis. Previous observations (6) indicated that light increases the cold resistance of 'Rough' lemon and sour orange seedlings. However, the report (8) that grapefruit seedlings do not harden in the dark is not entirely correct, since there is some hardening in the dark when the temperature is low. At high temperatures, it is possible that prolonged darkness could prevent hardening, although there was still some hardening of the different cultivars at 15°C in the dark in the present study. Plants of the same cultivars were 95% killed when exposed to freeze tests without any hardening treatment.

Under certain conditions, citrus plants can rapidly lose cold tolerance in the dark. This is shown in Table 2. Rusk citrange seedlings were used in this test, and they apparently had acquired some hardening in the greenhouse. When placed in a relatively cold T (6.7°C ± 0.6°C), changes occurred in cold tolerance. Increased hardening occurred within 1 day in the lighted chamber, and decreased hardening occurred in the dark.

Table 2. The average percentage of leaves killed (L_k) and dieback of the main stem (S_d) of 1-year-old Rusk citrange seedlings exposed to -6.7°C for 4 hr after hardening in light and dark at 6.5°C.

Prior hardening (days)	Light ²		Dark	
	L_k	S_d	L_k	S_d
0	30.0	10.1	30.1	10.1
1	29.4	4.3	46.9	12.6
2	5.8	2.6	82.5	42.5
4	7.8	1.0	85.1	45.8
7	8.0	0.0	87.0	50.0

²Differences in damage following light and dark conditioning are significant at the 1% level.

Table 1. The average percentage dieback of the main stem of citrus seedlings exposed to -6.7°C for 4 hr after 7 days at indicated temperature-light treatments.

Cultivar and species	% dieback			
	15°C		5°C	
	Light	Dark	Light	Dark
Rough lemon	36.6 ²	79.5	20.8	37.0
Duncan grapefruit	27.1	68.9	19.0	26.0
Orlando tangelo	26.4	82.3	---	---
Trifoliate orange	18.6	67.5	---	---
Valencia orange	---	---	4.0	19.0
All	27.2	74.5	14.6	27.3

²All differences between light and dark and between temperatures are significant at the 1% level.

Our finding that light *per se* increased hardening of citrus seedlings is not in complete agreement with the report of Young (7) that cold hardiness was increased by an 8-hr day but decreased by a 16-hr day. It is definite, however, that light must be taken into consideration in such hardening studies.

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Effect of Subfreezing Temperature on the Viability of Persian Walnut Pollen¹

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Abstract. Viability of walnut, *Juglans regia* L., pollen was not diminished by storage at subfreezing temperature, as previously indicated. Pollen stored 20 days at -19°C effected high percentages of fruit set in the orchard in 1969. Fruit set of the bagged flowers was relatively low in 1970, but the set effected by pollen stored a year at -19°C was not significantly different from that effected by fresh pollen. Laboratory tests indicated less than 1% germination for both freshly dehiscid and stored pollen, and were unreliable for indicating the ability of walnut pollen to effect fertilization.

Growers and breeders of Persian walnuts, *Juglans regia* L., and persons interested in the commercial aspects of artificial pollination, are concerned with the viability and storage of walnut pollen because pollen is frequently

insufficient at the time pistillate flowers are receptive. This situation results from the dichogamous nature of the walnut tree and from cultivar differences in bloom periods, accentuated by variations in response to different winter chilling conditions (1, 5, 11). A satisfactory method for storing walnut pollen from year to year would enable breeders to use pollen from late-blooming cultivars to pollinate early blooming ones, and would also assure a supply of viable pollen for commercial application (5, 8) during years when conditions for natural pollination are unfavorable.

Pollen of many deciduous fruit trees will maintain germinability for 1 or more years when kept in a freezer at approx -19°C, with no attempt to control humidity (6, 10). Little information is available regarding walnut pollen storage. Lack of a reliable

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