

# Chilling Requirement and Post-rest Heat Accumulation as Related to Difference in Time of Bloom Between Peach and Western Sand Cherry

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**Abstract.** The basis for the difference in time of bloom between 'Redhaven' peach [*Prunus persica* (L.) Batsch] and western sand cherry (*Prunus besseyi* Bailey) clone 'Cornell-Geneva (CG) 3-24' was investigated. Based on 3 years of field observation, average bloom date (50% of buds) of 'Redhaven' was 9 days earlier than 'CG 3-24'. Greenhouse forcing of field-collected shoots and artificially chilled trees suggested that the flower bud chilling requirement of these two clones is similar. Forcing of artificially chilled trees under different post-rest temperature regimes revealed that the base temperature for flower bud heat accumulation was lower in 'Redhaven' than in 'CG 3-24' (4.3° vs 7.0°C, respectively). Values of  $Q_{10}$  for flower bud development were 2.2 and 2.8 for 'Redhaven' and 'CG 3-24', respectively. These results suggest that the basis for difference in time of bloom is due to a difference in the base temperature for heat accumulation and is not related to chilling requirement.

Crop loss due to freeze injury of flower buds from late winter or early spring freezes is common in many temperate-zone fruit crops. Time of bloom is controlled by the interaction of the chilling and heat accumulation requirements (9). A better understanding of the determinants of bloom date in temperate-zone fruit crops and their wild relatives would be beneficial to plant breeders who are interested in developing late-blooming germplasm. Considerable variation exists in the genus *Prunus* for time of bloom, and such variation may prove useful in developing late-blooming cultivars in crops such as peach. The western sand cherry, a native American species, blooms considerably later than peach in most environments. This species is a genetic bridge in *Prunus* (16), hybridizing with many other *Prunus* species. Hence, it may be an important genetic resource in breeding late-blooming *Prunus*. The objective of this investigation was to assess the relative contribution of presumed differences in chilling requirement and/or post-rest heat accumulation requirement to the observed difference in time of bloom between 'Redhaven' peach and western sand cherry (clone 'CG 3-24').

## Materials and Methods

**Field study.** Greenhouse forcing of field-collected shoots was used to determine the approximate time of chilling fulfillment. Terminal shoots,  $\approx 0.25$  to 0.30 m in length, of 'Redhaven' peach and clone 'CG 3-24', were collected at  $\approx 7$ -day intervals during the dormant period from the Sandhills Research Station, Jackson Springs, N.C. Four shoots were collected from each of three 'Redhaven' trees and from one tree of 'CG 3-24'. Shoots were placed in pails of water containing 3% sucrose (w/v) and 0.3%  $Al_2(SO_4)_3$  (w/v) and forced in a greenhouse maintained at  $\approx 21^\circ$  day/ $18^\circ$ C night. Basal ends of the shoots were recut

weekly. Chilling was considered satisfied when 50% of the flower buds had broken after 21 days of forcing (17). Field-collected shoots were forced in a greenhouse during the winters of 1983-84 and 1985-86. Shoots were not sampled in 1984-85 due to a freeze that killed the majority of 'Redhaven' flower buds.

Bloom dates for both clones were recorded in 1984 through 1986. Bloom was defined as the date on which 50% of the flower buds were open.

**Greenhouse study.** Artificially chilled trees of both clones were forced in a greenhouse in Winter 1985-86 to provide a better estimate of chilling requirements. One-year-old trees of both clones propagated on 'Lovell' seedling rootstock were grown at the Sandhills Research Station during Summer 1985. Trees were removed from the nursery after leaf drop and chilled artificially at 6°C. Three trees of each clone were removed from chilling at about 200-hr intervals, potted in 2-liter containers containing sterile sand and perlite (1:1), and forced in a greenhouse maintained at 18° day/13° night. Chilling treatments ranged from 0 to 1700 hr. Since the trees used in this experiment were only 1-year old, only vegetative budbreak could be monitored on 'Redhaven', but trees of 'CG 3-24' bore sufficient flower buds for evaluation. Since the chilling requirement of lateral vegetative and flower buds of 'Redhaven' is equal (10), comparison of 'Redhaven' vegetative bud chilling requirement with that of flower buds of 'CG 3-24' should provide a valid chilling requirement comparison between the two. Number of vegetative buds showing visible leaf emergence on 'Redhaven' and the number of flower buds reaching stage 2 of plum flower bud development (2) on 'CG 3-24' were recorded after 45 days of greenhouse forcing. The plum flower bud development rating scale was used for 'CG 3-24' because it has a flowering habit similar to that of plum. The percentage of budbreak for each chilling treatment within each clone was calculated as the percentage of maximum budbreak in that clone after maximum chilling exposure (1700 hr). Data were subjected to quadratic regression and equations were generated to predict the number of chill units necessary to reach 80% of maximum budbreak for each clone.

**Phytotron study.** Forcing of artificially chilled trees under controlled environmental conditions was conducted to examine the response of both clones to various post-rest temperature regimes. This experiment was conducted in 1985-86 using 2-year-old trees of 'CG 3-24' and in 1986-87 using 3-year-old

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trees of 'Redhaven' and 'CG 3-24'. In the first study, 2-year-old trees of 'CG 3-24' were removed from the Sandhills Research Station nursery after leaf drop, and artificially chilled for 1300 hr at 6°C. Trees were then planted in 8-liter pots, transferred to the North Carolina State Univ. phytotron, and forced under a 9-hr photoperiod at 10°, 15.5°, 21°, or 26.5°, with three trees at each temperature. Flower budbreak was monitored daily in the 21° and 26.5° chambers and every other day in the 10° and 15.5° chambers. Flower buds were considered broken when stage 2 (side white) of development was reached (3). In the 1986–87 study, 3-year-old trees of both clones were grown outside in 15-liter pots during Summer 1986, and removed from the field after leaf drop. Tree were artificially chilled at 6° for 1400 hr, transferred to the phytotron, and forced at the same temperature and photoperiod as in 1985–86. Four trees of each clone were placed at each temperature. Flower buds of peach were considered broken at stage 4 (first pink) of bud development (3). In 'CG 3-24', the time that flower buds reached stage 2 and stage 5 (first white) was recorded (2). The chambers used in both years of this experiment provided  $\approx 645 \mu\text{mol}\cdot\text{s}^{-1}\cdot\text{m}^{-2}$  of light. Only cool-white fluorescent light was used to minimize the differential between actual bud temperature and that of ambient air.

In both years, the number of days (W) necessary to reach 80% budbreak of those buds that broke was determined for each clone at each temperature. A reciprocal transformation,  $100/W$ , was used in data analyses. This transformation has been used by Arnold (1) and Campbell (4), and represents the percentage daily average rate of development (DARD) toward budbreak. Thus, if it takes 25 days to reach 80% budbreak at a particular temperature, the  $\text{DARD} = 100/25 = 4\%$  per day = 4 DARD (4).

After calculation of DARD values for each clone at each temperature, the base temperature for heat accumulation was calculated using the X-intercept method of Arnold (1). To document the response to temperature over the range of temperatures used in this experiment,  $Q_{10}$  values were calculated (11).

## Results and Discussion

**Field study.** Based on greenhouse forcing of field-collected shoots, chilling requirement was fulfilled on 17 Jan. for both clones in 1983–84. In 1985–86, chilling was fulfilled on 15 Jan. and on 28 Jan. for 'CG 3-24' and 'Redhaven', respectively, suggesting that the chilling requirement for flower buds of 'Redhaven' is slightly greater than that for flower buds of 'CG 3-24'. The average date of 50% bloom from years 1984 through 1986 was 21 Mar. and 30 Mar. for 'Redhaven' and 'CG 3-24', respectively. These observations suggest that the basis for 'Redhaven' blooming before 'CG 3-24' is not a function of a difference in chilling requirement, since the time of chilling fulfillment, as suggested by greenhouse forcing of shoots, was later for the earlier-blooming 'Redhaven'.

**Greenhouse study.** Quadratic prediction equations generated by regressing percent maximum budbreak on chilling hours at 6°C (CU) showed that the chilling requirements for 80% maximum budbreak were 1180 CU for vegetative buds of 'Redhaven' and 1185 CU for flower buds of 'CG 3-24' (Fig. 1). These results confirm that the chilling requirements of 'Redhaven' and 'CG 3-24' are similar, and that chilling requirement cannot explain the difference in time of bloom between the two clones.

**Phytotron study.** Average DARD values for 'CG 3-24' flower bud development in 1985–86 ranged from 2.9 DARD at 10°C to 15.9 DARD at 26.5°. Linear regression of temperature vs.

DARD revealed that the base temperature for heat accumulation for development up to bud stage 2 was 7.7°.

Linear regression of temperature vs. DARD for 'CG 3-24' in 1986–87 revealed that the base temperature for heat accumulation for development up to bud stage 2 was 7.8°C, comparable to the calculated value of 7.7° obtained in the 1985–86 experiment. Base temperature calculation for 'Redhaven' peach (stage 4) and 'CG 3-24' (stage 5) flower bud development yielded values of 4.3° and 7°, respectively (Fig. 2). The value obtained for peach in this study agrees closely with the value of 4.5° commonly used in heat accumulation models for peach, originally proposed by Richardson et al. (9). Examination of the regression lines calculated from these data shows that the slopes of the lines for the two clones are significantly different (0.01 level) and that the lines intersect at  $x = 12.5^\circ$ . Values of  $Q_{10}$  calculated from the regression formula over the range of 12.5° to 22.5° were 2.2 and 2.8 for 'Redhaven' and 'CG 3-24', respectively.

The results of these experiments suggest that the basis for the difference in time of bloom between these two clones is related to the difference in the base temperature for heat accumulation and not to chilling requirement. Under field conditions, 'Redhaven' bloomed, on average, 9 days earlier than 'CG 3-24'. However, under controlled forcing conditions, 'Redhaven' bloomed later than 'CG 3-24' at 21° and 26.5°C, about the same time as 'CG 3-24' at 15.5°, and earlier than 'CG 3-24' at 10°. This suggests that these two clones are responding differentially to temperature, which is confirmed by the difference between the calculated  $Q_{10}$  values. That 'Redhaven' blooms earlier under field conditions in North Carolina may be explained by its lower base temperature for heat accumulation, since significant flower bud development occurs after rest fulfillment and between 4.5° to 12.5°. At higher temperatures, 'CG 3-24' would be expected to bloom earlier than 'Redhaven' due to the difference in  $Q_{10}$  for bud development.

The results of these experiments can also be interpreted relative to Levin's hypothesis (7), which proposed that temperature, or any other environmental signal, acts not only as an immediate variable modifying growth, but also as information relative to predicting future environmental conditions. Thus, the rate of bud development at a given temperature reflects not only the metabolic reaction to that temperature, but also reflects expected future growing conditions, given that particular temperature (4). Western sand cherry is native to the upper midwestern United States (18), and has undergone natural selection at more northerly latitudes than peach. Present-day commercial peach cultivars were derived from races of peach indigenous to southern China. Thus, the difference in response to temperature between the two species may reflect not only a difference in metabolic reaction to temperature, but also a difference in response to temperature as information, a function of place of origin.

Various authors have discussed the importance of a high post-rest heat unit requirement relative to late blooming in temperate-zone tree fruits. Spiegel-Roy and Alston (12) documented differences in heat-unit requirement among various pear species and cultivars and found that heat-unit requirement was positively correlated with time of bloom. Other studies have documented heat-unit differences in plum (13, 19) and almond (8, 14). Comparison and interpretation of heat-unit values must be done with caution, since the often-used base temperature of 4.5°C has not been experimentally confirmed for many woody species. Base temperature and heat unit total are related, in that

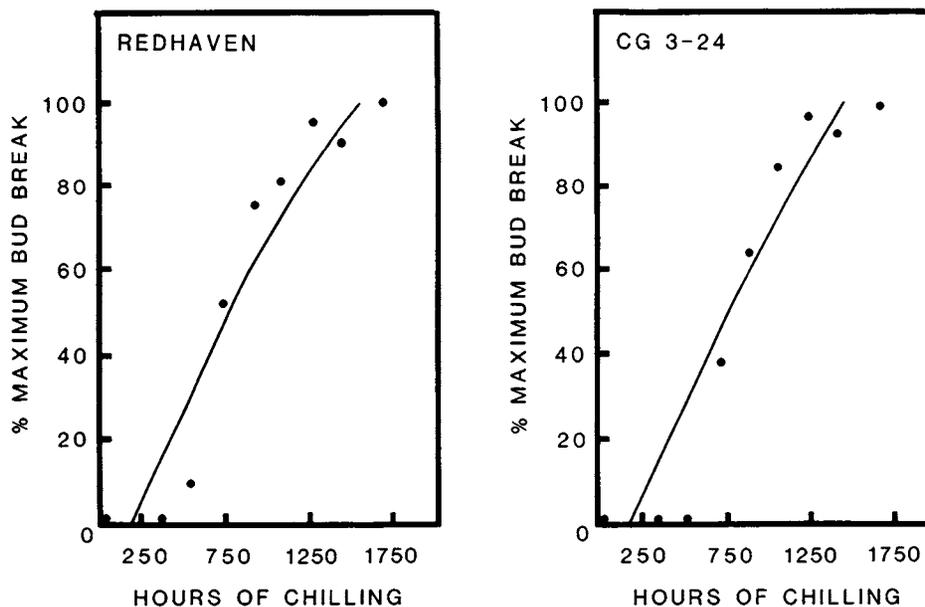


Fig. 1. Percent maximum vegetative budbreak (BB) on 'Redhaven' peach and percent maximum flower budbreak on *P. besseyi* clone 'CG 3-24' after various durations of controlled chilling at 6°C (CU) and greenhouse forcing for 45 days. Prediction lines were generated from the following formulas obtained through parabolic regression: 'Redhaven' % BB =  $-15.9 + 0.10(\text{CU}) - 1.6 \times 10^{-5}(\text{CU})^2$ ,  $R^2 = 0.88^{**}$ . 'CG 3-24' % BB =  $-18.7 + 0.10(\text{CU}) - 1.0 \times 10^{-5}(\text{CU})^2$ ,  $R^2 = 0.82^{**}$ . Data points are means of three replications.

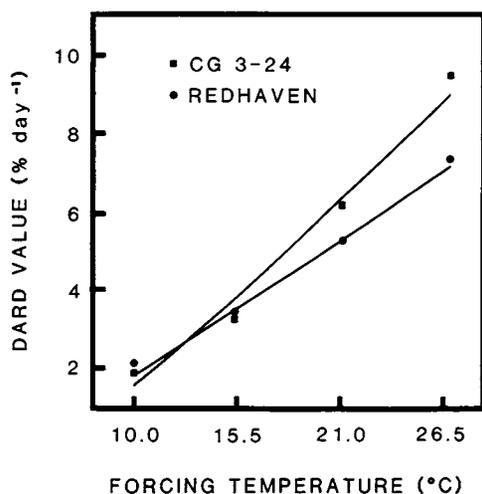


Fig. 2. Daily average rate of development (DARD) vs. forcing temperature after 14 hr of chilling for flower buds of 'Redhaven' peach and *P. besseyi* clone 'CG 3-24'. Linear regression lines shown were generated from the formulas: 'Redhaven' DARD =  $-1.36 + 0.32$  (forcing temperature),  $R^2 = 0.99^{**}$ . 'CG 3-24' DARD =  $-3.19 + 0.46$  (forcing temperature),  $R^2 = 0.97^{**}$ . Data points are means of four replications.

fewer heat units are accumulated as the base temperature is raised (15). Thus, reported differences in "heat-unit requirement" may actually be a function of differences in the base temperature. Gianfagna et al. (6) showed that late-blooming apple clones were less responsive to 10°C forcing temperature than earlier-blooming clones, suggesting that base temperatures for flower-bud development were higher in late-blooming clones. Our results suggest that the difference in base temperature for heat accumulation may be an important determinant of time of bloom in peach and western sand cherry.

Although interspecific hybridization has been accomplished between peach and western sand cherry (5), sterility in these hybrids will have to be overcome before introgression of genes

from western sand cherry into peach is possible. At this time, the use of late-blooming accessions of exotic peach germplasm appears to be the most feasible route for developing late-blooming peach cultivars through breeding. However, interspecific hybrids between western sand cherry and the Japanese plum (*P. salicina*) are highly fertile (5). Hence, the goal of developing later-blooming Japanese plums through interspecific hybridization with western sand cherry appears feasible.

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## Effects of External Ethylene on the Production of Endogenous Ethylene in Olive Leaf Tissue

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**Abstract.** Olive (*Olea europaea* L.) leaves are characterized by their ability to respond to exogenous ethylene by a 100- to 400-fold enhanced ethylene production irrespective of leaf age or time of year when sampled. The autoenhancement of ethylene production from intact or detached leaves is positively correlated with the concentration of external ethylene. A lag time of 72 to 120 hr occurred before the autoenhancement of ethylene production could be observed. An autoinhibition of ethylene production was usually observed during the first 24 to 48 hr. The effect was, however, much less pronounced. This autoinhibition of ethylene production apparently does not involve wound ethylene. Olive fruit normally produce only negligible amounts of ethylene, and the enhanced ethylene evolution, which was observed after the fruits were exposed to exogenous ethylene, was found to be exogenous ethylene that was trapped by the fruit tissue during its exposure to ethylene. In leaves, however, autoenhancement of ethylene production evidently is a physiological response that may induce a senescing process in the leaves rather than abscission.

Mechanical harvesting has been one of the major objectives of the olive industry for many years, and the major factors involved have been summarized by Martin et al. (21) as follows: a) costs of hand-harvest procedures are high and labor availability is unreliable; b) the rate of hand harvest is slow; and c) better results are envisioned when growers can initiate harvest as deemed most appropriate and proceed at a rate that maximizes the use of handling facilities.

The main problem with commercial mechanical harvest is the need to reduce the retention force of the fruit and yet retain the leaves that are necessary for flower bud formation. Many fruit-abscising chemicals have been tested (8-10), with those releasing ethylene showing the greatest potential (4, 10, 13-15, 19, 20).

2-(Chloroethyl)phosphonic acid (ethephon) was found to be rather stable on fruit and leaf surfaces in its acid form (15), but decomposed as pH (1) and temperature (11, 24) increased. Ethephon has not been widely used because of the inconsistency of treatments in loosening fruit satisfactorily without promoting excessive leaf loss (10, 21). One major obstacle to the wide use of ethephon in this connection is the lack of understanding of

the mechanism by which ethylene is released and its relation to the abscission process (7, 13).

The uptake of ethephon is rather slow (5, 15), its translocation from the site of application is limited (5, 15, 22, 25), it does not accumulate in the treated tissue (19), and the response to ethephon is usually localized and often limited to the organ treated (5, 15). Thus, the short-term response of the tissue is not surprising, especially since it was reported (13) that the first peak of ethylene production is obtained 18 hr after ethephon treatment. However, several questions were raised concerning the long-term response of plants to released ethylene (13, 18-20). Lang and Martin (13) compared the release of ethylene in fruits treated with ethephon and 2-(chloroethyl)methylbis-(phenylmethoxy)-silane (CGA-15281). In addition to the first peaks of ethylene release, they observed a second peak of ethylene 30 hr after application of ethephon. No such peak was observed when fruits were treated with CGA-15281, although ethylene release from CGA-15281 was much greater than from equimolar concentrations of ethephon throughout the application time. Although the characterization of the two peaks of ethylene derived from ethephon in treated olive fruits also has been based on field studies (1, 10), the nature of the second peak of ethylene evolution was questioned. One interpretation suggested that the second peak is an autocatalytic production of endogenous ethylene induced by ethylene that initially was released by ethephon (1, 3). The long-term response of the tissue to ethephon, including the occurrence of the second peak of ethylene pro-

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