The Effect of LAB 173711 and Ethephon on Time of Flowering and Cold Hardiness of Peach Flower Buds

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Abstract. Peach flowers are often killed during bloom by spring frosts. LAB 173711, a compound with abscisic (ABA)-like activity, and ethephon delayed flowering in peach trees. In greenhouse experiments, LAB 173711, at concentrations of 10⁻³- 10^{-2} M, was most effective in delaying bloom when applied after a 5°C cold storage period, rather than before the dormancy breaking treatment. In contrast, ethephon delayed bloom most effectively when applied before 5°C cold storage; ethephon caused flower bud abscission when treatments were made after the chilling requirement had been satisfied. In field experiments, ethephon delayed flowering by 6-7 days, which reduced bud injury after a spring frost during bloom. No flower bud injury was found on ethephon-treated trees after temperatures of -4.3° C; whereas without ethephon 25% of the flower buds were frost damaged. LAB 173711 delayed the time to 50% bloom by 2-3 days. However, this was not long enough to avoid low-temperature injury to the flower buds.

LAB 173711 (5-(2,6,6-trimethyl-1-hydroxy-4-(propylene-1,2-dioxy)-cyclohex-2-en-1-yl)-3-methvlpent-2-en-4-yn-1-al-dimethylacetal) is a synthetic analog of abscisic acid (ABA) (Fig. 1) which induces responses in plants similar to ABA. LAB 173711, for example, promotes stomatal closure in isolated barley and sunflower leaves, reduces transpiration in whole plants, promotes senescence of detached leaf segments, and abscission in petiole explants (Flores and Dorffling 1990, Grossmann and Jung 1984, Jung and Grossmann 1985). Since ABA may also play a role in bud dormancy and break (El-Antably et al. 1967), as well as increase resistance to low-temperature stress (Chen and Gusta 1983, Flores et al. 1988), I began to investigate the effect of LAB 173711 on flower bud break

and cold hardiness in peach (*Prunus persica* L. Batsch). Since low-temperature injury to flower buds is a significant problem in most peach-producing regions worldwide, we were particularly interested in determining whether this compound could delay bloom and thereby allow flower buds to avoid or resist spring frost injury. I was also interested in comparing its potential effect on flowering with ethephon, a compound known to delay bloom and increase winter flower bud cold hardiness (Durner and Gianfagna 1988, Proebsting and Mills 1972).

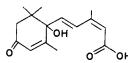
Materials and Methods

Bloom Delay Screen

A screening procedure was developed to discover bloom delay compounds by using container-grown peach trees propagated from adult stem cuttings (Couvillon et al. 1975). This method of propagation bypasses the 2–3 year nonflowering juvenile growth phase, and provides a small tree (approximately 40×40 cm in height and width) with 3–5 branch units containing 20–40 flower buds, which can be used for screening and other experiments after 1 year. These trees are moved between the greenhouse and cold rooms to simulate the seasonal temperature cycles which induce bud dormancy and provide the rest breaking conditions for subsequent bud growth. For the "Redhaven" variety used in this experiment, a 9-week cold storage period in the dark at 5°C was used to break rest of the flower buds, after which trees were maintained in a greenhouse at 25°C daytime and not less than 10°C night temperatures.

Compounds were applied with surfactants as foliar sprays to each of at least five trees at five rates: (A) after flower bud differentiation, but before the plants were placed in 5° C storage, (B) after the plants received the 5° C storage treatment, and (C) both before and after the cold storage period.

Flower bud development was assessed using a numerical scoring system (Gianfagna et al. 1986) in which 1 = dormant bud, 2 = green stage, 3 = red calyx, 4 = pink, and 5 = open flower. Each flower was individually rated at each sampling date, and an average value for each plant was obtained. These data were analyzed statistically by ANOVA and LSD test.



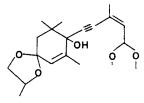


Fig. 1. The chemical structures of abscisic acid (upper) and LAB 173711 (lower).

LAB 173711 was formulated as a 10% solution by cyclohexanone:Emulan EF (4:1). Approximately 30 ml of a 0, 10^{-1} , 10^{-2} , 10^{-3} , 10^{-4} , or 10^{-5} LAB 173711 treatment solution with Lutensol surfactant (0.1%) was applied to each tree using the protocol described above. Since the best results were obtained with the 10^{-2} M treatment, this experiment was repeated twice at this rate only.

Ethephon (2-chloroethyl phosphonic acid, 0.85 mM) and ABA (10^{-2} M) were applied using the standard protocol described above.

Field Experiments

LAB 173711 (2.5 mM) with Lutensol surfactant (0.1%) was applied to 11-year-old "Cresthaven" peach trees in a volume of 3 L per tree with a power hand gun sprayer, to five single tree replicates, in a complete randomized block experimental design. Treatments were made at about the time the buds were in the green stage of development. Weather conditions at the time of application were overcast and windy, with an air temperature of $22^{\circ}C$.

Ethephon (0.85 mM) with Regulaid surfactant (0.25%) was applied as described above, after flower bud differentiation in the fall. Weather conditions at the time of application were clear and calm, with an air temperature of 15° C.

Ten twigs were tagged on each treatment replicate and flower buds were rated for bud development on nine dates during the period of bud expansion, using the numerical scoring system. Flower bud mortality due to low-temperature stress was determined by recording the number of live flower buds present on each tagged twig on each of nine dates, beginning with bud expansion, until about 10 days after full bloom.

Fruit were harvested, graded by size, and weighed after the ground color changed from green to yellow.

Table 1. Relative bloom delay response of greenhouse grown "Redhaven" peach flower buds to five rates of LAB 173711.

Application rate (-log M)	Application time		
	Before 5°C storage	After 5°C storage	Before and after 5°C storage
1	Toxic	Toxic	Toxic
2	No effect	+ + a	+ +
3	No effect	+ ^b	+
4	No effect	No effect	No effect
5	No effect	No effect	No effect

N = 6.

^a Significant at 1% level; ^bsignificant at 5% level.

Results

Bloom Delay Screen

The preliminary peach screen factorial experiment indicated that LAB 173711 delayed flowering in peach at the 10^{-2} and 10^{-3} M rates when applied after the 5°C storage period (Table 1). Lower application rates, or application of LAB 173711 before the 5°C cold storage period, had no effect on the rate of flower bud expansion and bloom date. The 10^{-1} M treatment was toxic at all application times; treated buds either failed to develop or simply abscised after treatment.

The rate of flower bud development was significantly slower for the treatments which received 10^{-2} M LAB 173711 and ABA after 5°C storage, than for those which did not (Fig. 2). After 15 days in the greenhouse there was no measurable growth; however, for the control, and the treatment which received LAB 173711 before cold storage, the flower buds had developed from the green to red calyx stage. These treatments were close to full bloom 22 days after transfer to the greenhouse, whereas the after-cold storage, and the combination treatment, had not yet reached the red calyx stage of growth.

In contrast, ethephon delayed flowering most effectively when applied prior to the 5°C cold storage period (Fig. 3). After 25 days in the greenhouse, buds treated with ethephon had developed only to the red calyx stage, whereas untreated flower buds were close to full bloom. The ethephon applications made after the trees had received the 5°C cold storage period induced flower bud abscission within several days of treatment.

Field Experiments

Application of LAB 173711 in the spring resulted in

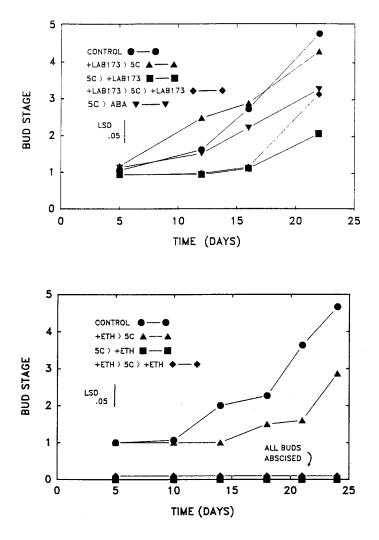


Fig. 2. The effect of application time of ABA and LAB 173711 on the rate of flower bud growth of greenhouse grown "Redhaven" peach trees (N = 6).

a 2 to 3-day delay to 50% bloom (4.5 on the scoring system) and a 4-day delay to full bloom (Fig. 4). Ethephon provided a greater delay in flowering (6 days), and the combination treatment of ethephon in the fall and LAB 173711 in the spring resulted in a 7-day delay in bloom.

The number of live flower buds declined significantly during the period of flower bud expansion (Fig. 5). Flower bud mortality was correlated with two periods of low-temperature stress. The first on April 12 (-4.3° C) reduced the number of live buds on both the control and LAB 173711-treated plants by about 25%, but had little effect on the ethephontreated plants, and the combined ethephon-LAB 173711-treated buds. Flower buds from treatments not receiving ethephon grew more rapidly, and about 50% of the buds were fully opened at the time of low-temperature stress, whereas there were no open flower buds on the more slowly developing ethephon-treated plants. The second incidence of low-temperature stress occurred on April 22

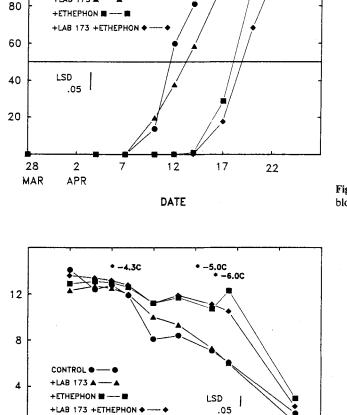
Fig. 3. The effect of application time of ethephon on the rate of flower bud growth of greenhouse grown "Redhaven" peach trees (N = 6).

 $(-5.0^{\circ}C)$ and April 23 $(-6.0^{\circ}C)$, and caused significant bud losses on all treatments. At the time of the second frost period, flower buds on all treatments were in their most cold-sensitive growth stages.

Nevertheless, fruit yield was significantly greater for treatments receiving ethephon, although not for the LAB 173711 treatments (Table 2). There were no significant treatment effects on fruit size or harvest date (data not shown).

Discussion

Flowering was delayed by 5–8 days in the greenhouse screening experiment with LAB 173711 (Fig. 2), but only when this compound was applied after the chilling requirement for bud break had been satisfied. The results suggest that the compound acts as a growth inhibitor of either cell division or enlargement within nondormant or quiescent floral primordia. LAB 173711 would appear to have little



effect on the intensity or duration of the rest period, since it had no effect on time of bloom when applied to buds before transfer to 5°C. The role of ABA in flower bud dormancy is unclear. Early work suggested a role in the induction and maintenance of the rest period. However, there is little evidence that the levels of ABA in flower buds change in response to environmental conditions, which either induce or break bud dormancy. Lenton et al. (1972) found higher amounts of ABA in actively growing buds maintained in long days compared to buds in which dormancy was induced by short photoperiods. Balboa-Zavala and Dennis (1977) found that ABA levels in peach seeds declined during stratification at 5°C, which breaks dormancy, but that ABA levels declined even more rapidly at 20°C, conditions which do not break the rest period. There are numerous reports, however, demonstrating that ABA applied to seeds in which dormancy has been broken will prevent or inhibit germination (Zigas and Coombe 1977).

Fig. 4. The effect of LAB 173711 and ethephon on time of bloom of field grown "Cresthaven" peach trees (N = 5).

Fig. 5. The effect of LAB 173711 and ethephon on flower bud survival after low-temperature stress in field grown "Cresthaven" peach trees (N = 5).

 Table 2. Effect of LAB 173711 and ethephon on fruit yield in field grown "Cresthaven" peach.

Treatment	Fruit yield (kg/tree)	
Control	87.5	
Ethephon	104.3	
LAB 173711	79.9	
Ethephon + LAB 173711	102.2	
Significance		
Ethephon	а	
LAB 173711	NS⁵	
Ethephon × LAB 173711	NS	

N = 5.

^a Significant at 5% level; ^bNS, not significant.

Ethephon, under the same screening test also delays flowering (Fig. 3), but probably by a different mechanism, as it was only active when applied before the 5°C storage treatment. We have shown that ethephon acts to prolong the period of flower bud

PERCENT BLOOM

BUD SURVIVAL (BUDS/30 CM)

CONTROL ● +LAB 173 ▲

7

APR

12

17

DATE

22

27

2

MAY

dormancy by reducing the effectiveness of 5° C storage to break the rest period (Durner and Gianfagna 1991). This increases the length of the dormant period and delays bloom. Another apparent effect of the 5°C cold storage period is that it causes the floral bud abscission zone to become sensitive to abscission induction by ethephon, but apparently not to ABA or its analogue.

In the field experiment with "Cresthaven," flowering was delayed by both LAB 173711 and ethephon. LAB 173711 delayed flowering (50% bloom) by 2–3 days. This was not enough, however, to significantly prevent low-temperature injury to the buds. It may be possible to extend the delay in bloom date by LAB 173711 by increasing the concentration and/or by earlier or multiple applications. The field rate of 2.5 mM could be increased by perhaps 4–10-fold without toxic effects, especially if some breakdown of the compound occurs by photodecomposition in the field. However, cost of application at higher rates may be commercially prohibitive.

Application at the green bud stage may not have been the most LAB 173711-sensitive stage of bud development. Flower buds may indeed respond better to the growth inhibitory effects of LAB 173711 prior to visible bud growth, which was the stage at which LAB 173711 was applied in the greenhousescreening experiments.

Ethephon delayed bloom by about 6 days and this did allow the avoidance of low-temperature injury due to the frost on April 12. It has been shown that flower buds progressively lose cold hardiness as they develop in the spring (Proebsting and Mills 1978), including those treated with ethephon (Durner and Gianfagna 1988); however, trees treated with ethephon were at the more coldresistant red calvx to pink growth stages, compared to 40% bloom for the LAB 173711, and 60% bloom for the control treatments, at the time the -4.3° C temperatures were experienced. Ethephon did not provide much protection, however, from the frosts of April 22 and 23; at this point all treatments were at full bloom or in the equally frost-sensitive stages of early fruit set.

Harvest data indicated that despite the highly significant loss of flower buds in April, yield was not reduced to any considerable extent (Table 2); since from past records with these trees, 100 kg per tree yields were rarely exceeded. Only about 15% flower bud survival is actually required for a full crop in peach, and flower bud survival after the two frost periods averaged 20% for the ethephon-treated plants, and 13% for the plants receiving LAB 173711 (Table 2) which is around the threshold level for a full crop. In the field, the effectiveness of any chemical treatment in delaying flowering to avoid frost injury (and thereby maintaining fruit yields) will only be clearly observable when bud losses occur in excess of these levels.

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