

Labavitch

ANNUAL REPORT
December 29, 1983

PROJECT NO. 83-17: Tree & Crop Research, Almond Development

Objectives

To develop information about various physiological aspects of almond fruit development: (1) The effects of light treatment (interruption of night) on fruit set (Martin and Weinbaum), (2) the effects of temperature on pollen tube growth (Weinbaum, Parfitt and Polito), (3) the effects of cross and self pollen tube growth on ethylene production, ovule receptivity and fruit set (Polito, Weis and Labavitch), and (4) the role of ethylene in normal almond abscission (Labavitch and Weis).

Interpretive Summary

1. An experiment to determine the effect of midnight orchard illumination on almond fruit set was conducted. No effect (positive or negative) on set was measured although wet weather created a lot of problems. We plan to repeat this work, at Davis this time, and have been promised full cooperation by GTE.
2. Pollen germination and pollen tube growth were studied in six cultivars of almond and four cultivars of peach. Almond pollen germinated much more effectively at low temperatures (down to 35°F) than did that of peach. Pollen tube elongation rates for almond and peach were similar for similar temperatures. Germination of pollen from low-chilling almond cultivars at low temperatures was no greater than that of pollen from cultivars with a high chilling requirement.
3. Embryo sac development and production of the gaseous hormone ethylene were measured in pollinated and unpollinated 'Non-pareil' almond flowers. A strong correlation between embryo sac growth (which indicates successful pollination) and floral ethylene production was demonstrated.
4. Application of ethylene gas (3 parts per million) to growing almond fruits led rapidly (within a few days) to dehiscence and abscission while treatment of fruits with agents which prevent ethylene action tended to retard fruit maturation. Spray application of ethephon (5 ppm) also hastened fruit maturation. These results indicate that the natural production of ethylene by almond fruit controls nut abscission. Thus, if a means to control tree/fruit ethylene production can be found then effective regulation of fruit abscission may be possible.

Experimental Procedures

1. This work was carried out in an orchard in the vicinity of Hughson, CA. GTE had established light fixtures in a part of the orchard. The lights were turned on for 30 minutes each night (beginning at midnight) from February through April.

Trees in the illuminated and control (unlighted) regions of the orchard were selected, limbs were marked, and blossoms on each of the selected limbs were counted (late February). In April fruits on the selected limbs were counted and the percentage of fruit set was calculated.

2. Pollen was collected at anthesis from flowers ("popcorn" stage) of six cultivars of almond and four of peach. Pollen was incubated at various temperatures in Petri dishes containing an agar/sugar medium. After 16 hours of incubation the preparations were fixed (killed) and examined for pollen germination using a microscope. Following germination in a defined medium, pollen germination and tube growth (at various temperatures) were assessed using a microscope in conjunction with computer-interfaced measuring devices.
3. 'Nonpareil' flowers in the "popcorn" stage were emasculated and either cross-pollinated ('Ne Plus Ultra' pollen) or left unpollinated. Samples of pistils were taken daily and prepared for microscopic examination (measurement of embryo sac size) or enclosed in glass tubes in order to measure ethylene production (gas chromatography).

Samples of 'Nonpareil' pollen were washed with water and the material washed from the pollen was assayed for the presence of 1-aminocyclopropane-1-carboxylic acid (ACC), the substance which is converted to ethylene in plant tissues.

4. On July 13 clusters of 'Nonpareil' almonds growing on trees at the U.C. Davis orchard were enclosed in plastic bags fitted with inlet and outlet tubes. Gases could then be directed into the bags so as to control the atmosphere surrounding developing fruits. Bagged clusters were then gassed with air (control) or air to which 3 ppm ethylene had been added. A third group of clusters was treated with 3 ppm ethylene for one day and then removed from the bags for two days. This one day ethylene/2 days air cycle was repeated continuously. The effects of these treatments on nut dehiscence and abscission were visually assessed at regular intervals.

Developing fruits of 'Ne Plus Ultra' almond (growing in Winters, CA) were subjected to weekly spray applications of a variety of chemicals beginning on July 22. Applications were made with hand-held sprayers and care was taken to thoroughly wet fruit surfaces. Spray materials used were:

- a. Ethephon (1, 5, 10 and 20 ppm in water)

- b. Silver thiosulfate (2 mM) in a 0.1% water solution of Tween 20
- c. Water
- d. 10 mM aminooxyacetic acid (AOA) in 0.1% Tween 20
- e. Tween 20 (0.1%) in water

The effects of spray treatments on nut dehiscence and abscission were assessed twice weekly and compared to a group of nuts that had received no spray treatments.

Similar tests of spray applications were carried out on "Mission" almonds growing at the Nickel's Estate in Arbuckle, CA beginning on August 2. Treatments were similar to those described above (the 1 and 5 ppm ethephon treatments were eliminated and AOA concentration was raised to 10 mM) and evaluations of nut maturation were made twice weekly.

Results

1. There appeared to be no effect of midnight orchard illumination on almond fruit set in our trials although inclement weather could easily account for this.
2. Germination of almond pollen at low temperatures was much greater than for pollen of peach (Fig. 1). Maximal germination of pollen from the six almond cultivars tested ('Harriott', 'Bigelow', 'Hybrid A', 'Ne Plus Ultra', 'Nonpareil', and 'Texas') occurred at 16°C while the maximum germination for the four peach cultivars occurred at 23°C. Pollen tube growth with respect to temperature (Fig. 2) was similar for peach and almond. (See appended paper by Weinbaum, Parfitt and Polito for complete details.)
3. Beginning at about three days following pollination a divergence in average embryo sac length could be seen when cross- and non-pollinated flowers were compared (Fig. 3) and by 9 days post pollination embryo sacs in the cross-pollinated flowers averaged twice the length of those in non-pollinated flowers. Ethylene emanation from excised pistils was low and variable but by seven days following pollination the pollinated pistils were clearly producing more ethylene (Fig. 4).

Materials washed from the surface of stored 'Nonpareil' pollen were shown by chemical test to contain the ethylene biosynthetic precursor ACC.

4. 'Nonpareil' almonds treated with 3 ppm ethylene in an air stream rapidly began to split (Fig. 5) and abscise. Samples treated with ethylene for one day out of every three also matured much more rapidly than "air-treated" or unbagged nuts.

When samples which had been air-treated were switched to ethylene treatment their development was also accelerated.

Sprays with ethephon accelerated dehiscence of fruits of 'Ne Plus Ultra' (Fig. 6) and 'Mission' (Fig. 7). STS treatment delayed development in these two varieties. AOA did not affect maturation of 'Ne Plus Ultra' almonds when used at a concentration of 1 mM and thus was used at 10 mM on the 'Mission' fruits in Arbuckle. Dehiscence was slowed at this higher concentration (Fig. 7). Sprays with the wetting agent Tween 20 had a slightly promotive effect on dehiscence (data not shown) and so we presume that the actual retardation caused by STS and AOA (which contained Tween 20) might have been even greater.

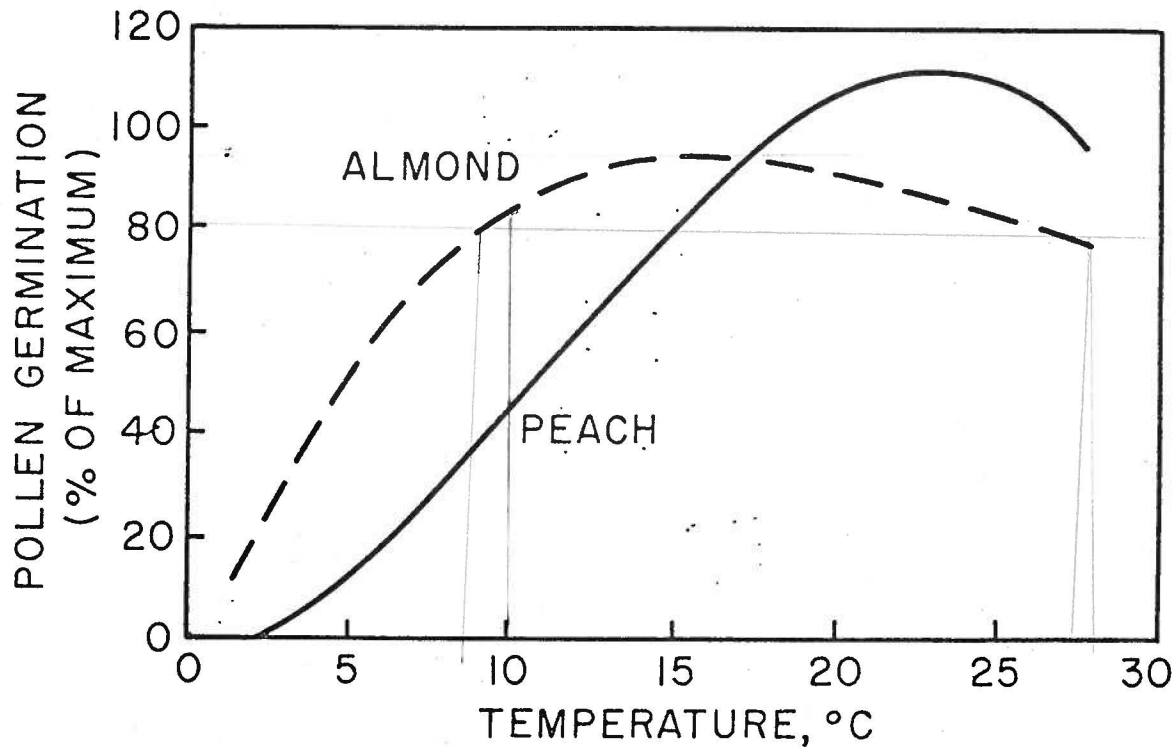


Fig. 1. Effect of temperature on pollen germination.

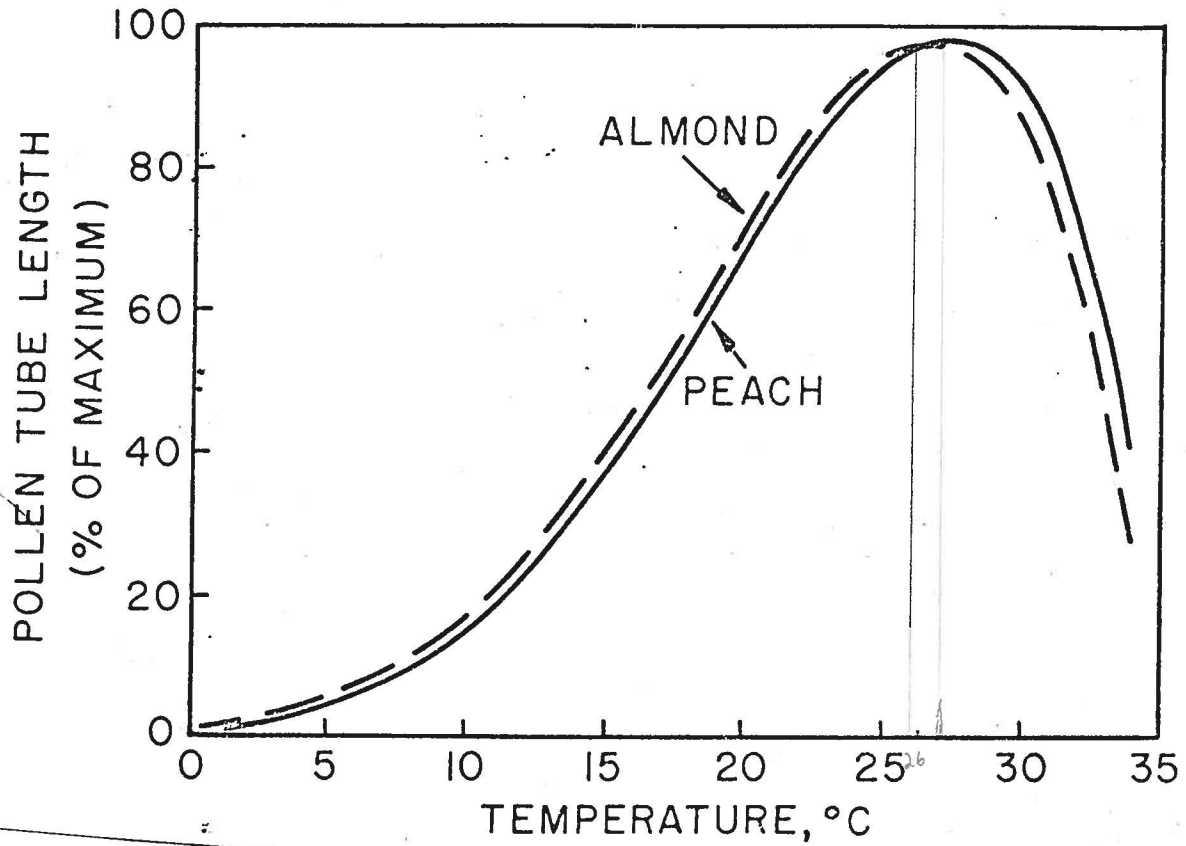


Fig. 2. Effect of temperature on pollen tube elongation.

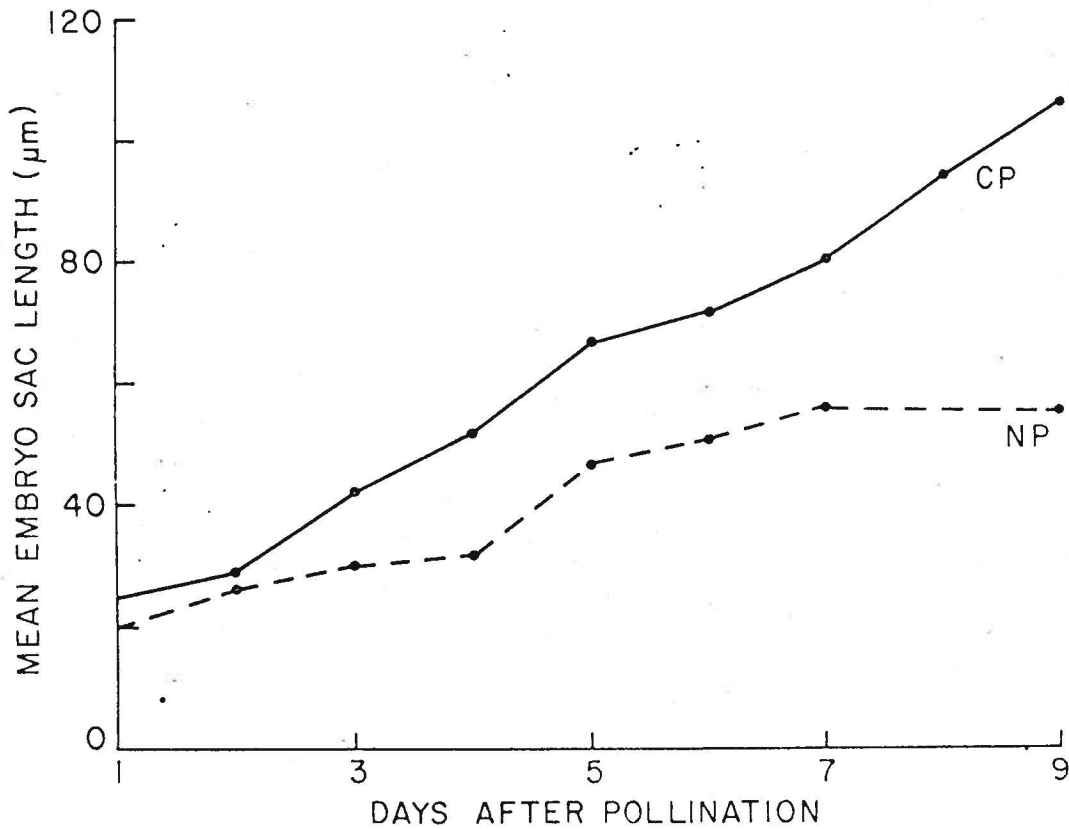


Fig. 3. Embryo sac growth in cross-pollinated ("CP") and non-

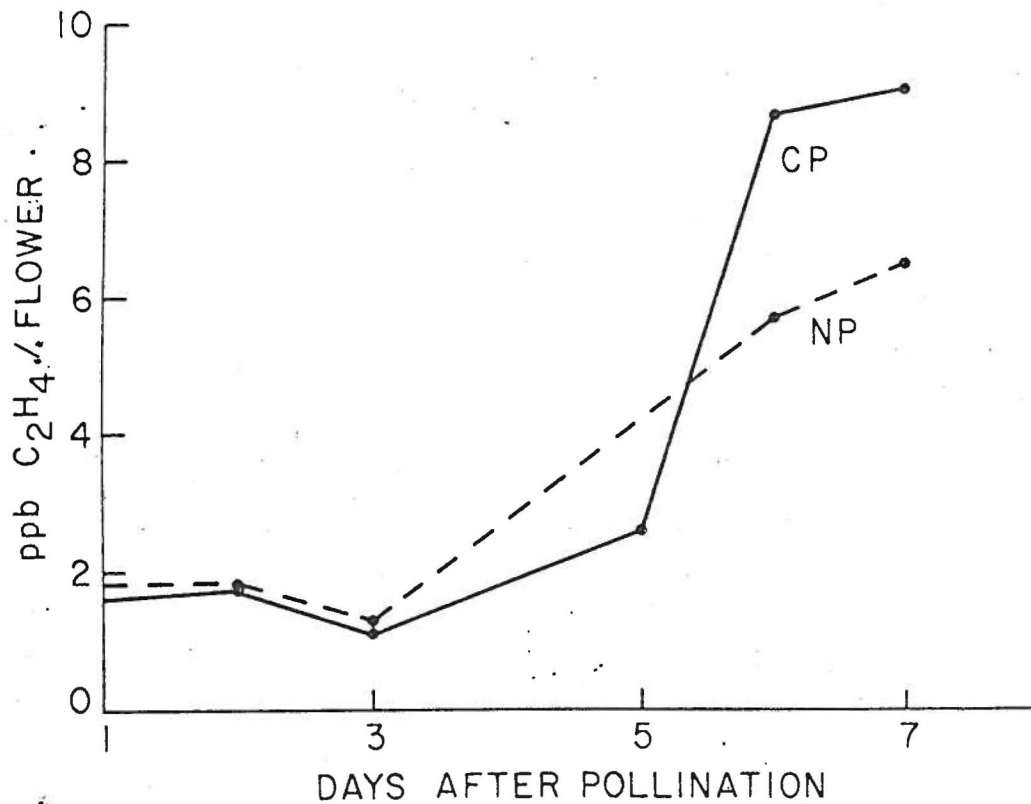


Fig. 4. Ethylene production by pistils from cross-pollinated ("CP") and non-pollinated ("NP") almond flowers.

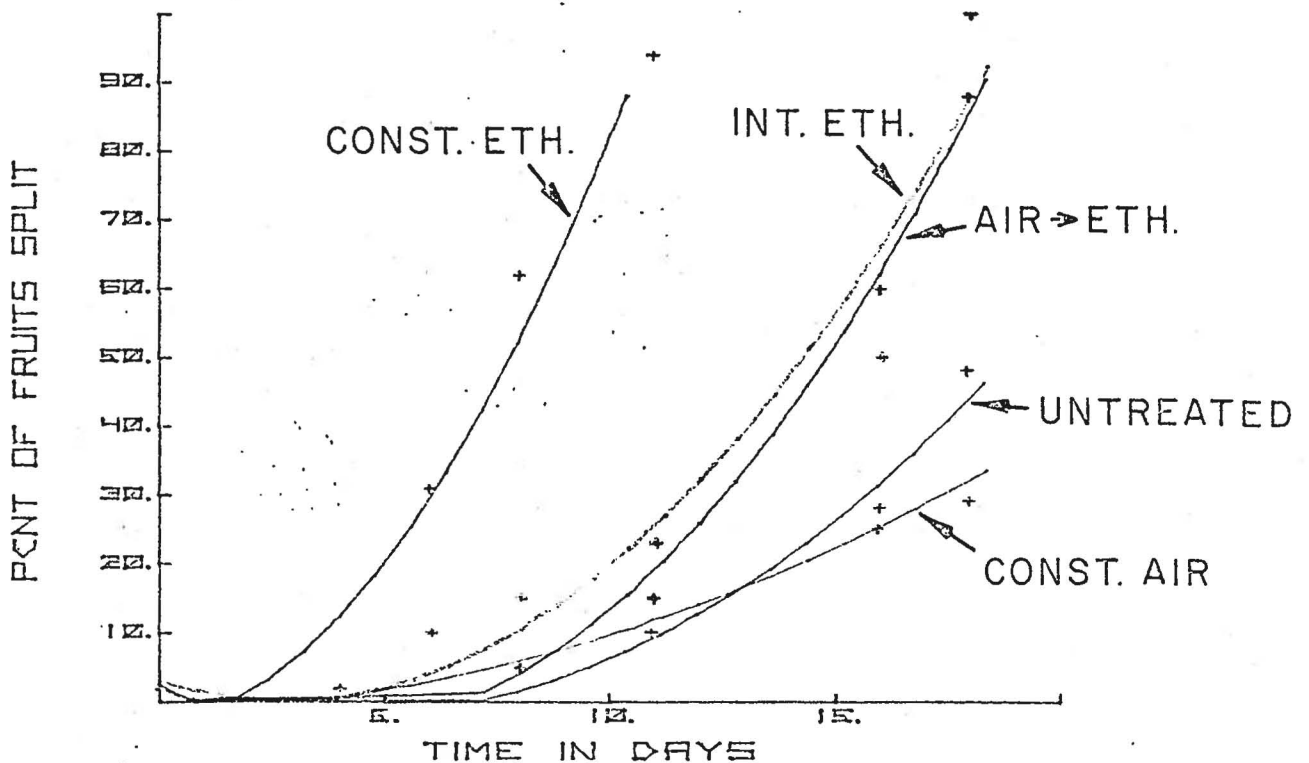


Fig. 5. Hull split in 'Nonpareil' almond fruits treated with 3 ppm ethylene ("CONST ETH"), ethylene for 1 day and air for 2 days, alternating ("INT ETH"), air only ("CONST AIR"), air for 1 week and then 3 ppm ethylene ("AIR ETH"), or fruits that were not treated.

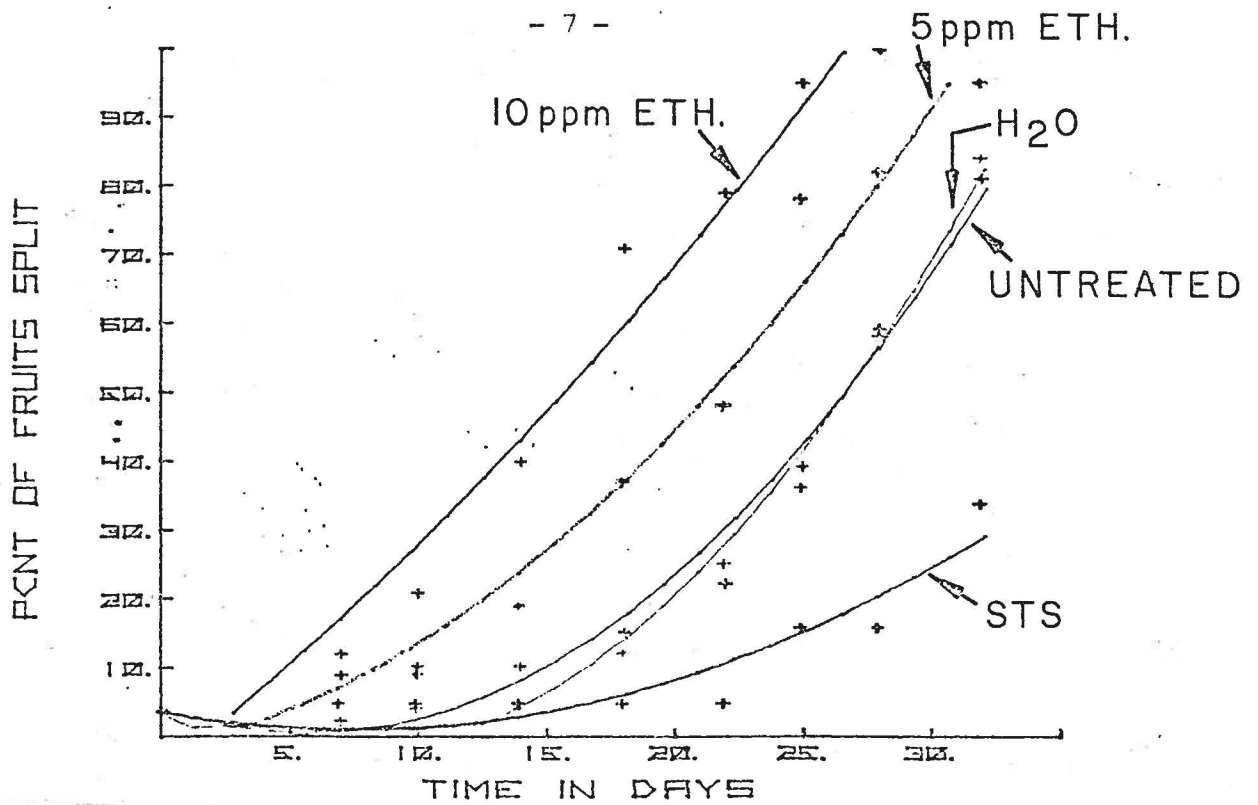


Fig. 6. Hull split in 'Ne Plus Ultra' almond fruits sprayed with 5 and 10 ppm ethephon ("ETH"), water, and 2 mM silver thiosulfate ("STS") at weekly intervals.

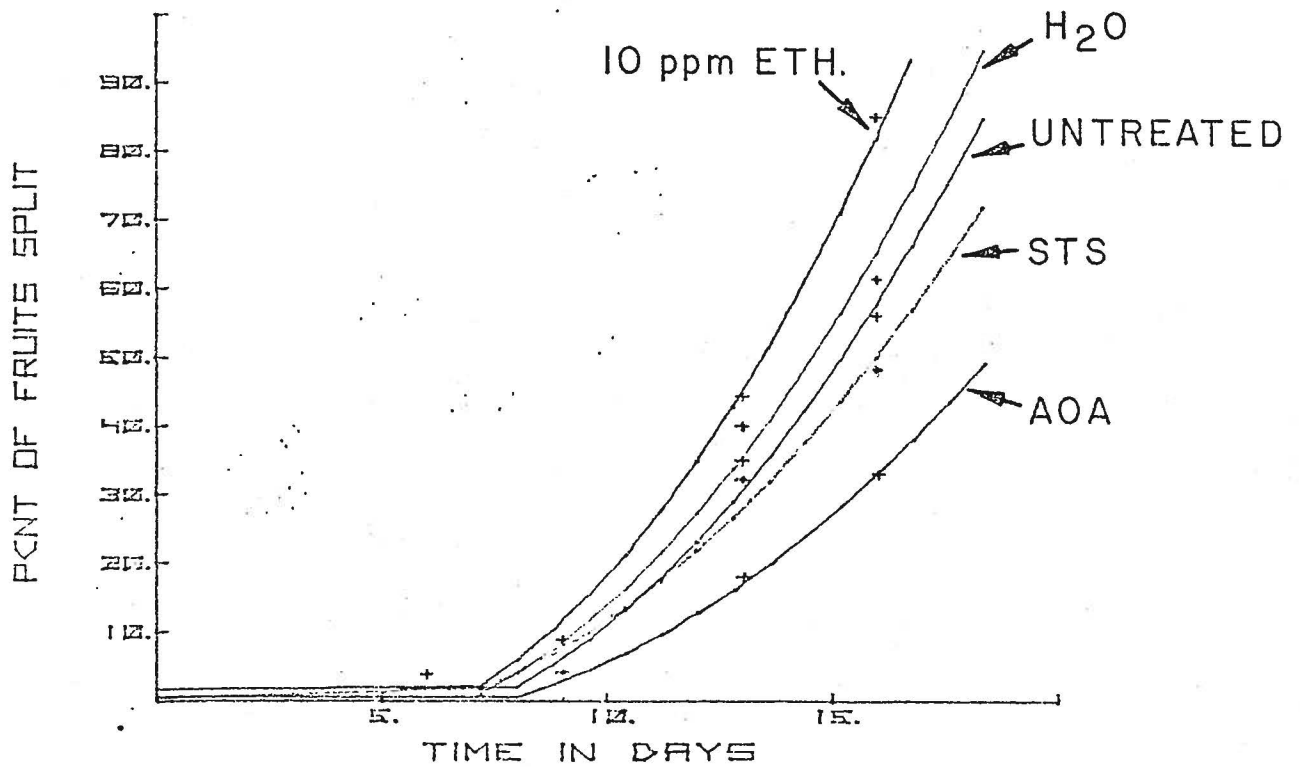


Fig. 7. Hull split in 'Mission' almond fruits sprayed with 10 ppm ethephon, water, STS, and 10 mM aminoxyacetic acid ("AOA") at weekly intervals.

Discussion

1. While we did not see an effect of orchard illumination on fruit set in our studies GTE personnel have shown beneficial effects in trials they have run for the past few years. We intend to repeat our tests in 1984 and have the promise of substantial cooperation from GTE.
2. It has been suggested that pollen from cultivars with a low-chilling requirement might be able to germinate more readily at low temperatures than pollen from cultivars with a higher requirement. If so fruit set might be enhanced under cold conditions if pollen from low-chilling cultivars were supplied. The information provided here (and in the appended manuscript) suggests that there is no clear correlation between chilling requirements and pollen germinability in almond. The 22% germination of 'Harriott' pollen at 1.5°C (Table 2 of appended paper) is rather remarkable.

The data in Figure 4 indicate that little pollen tube elongation will occur at temperatures below 10°C. This suggests that low temperature at bloom probably would not be a factor limiting fruit set except as it may affect bee activity.

3. Ethylene production by flowers has often been correlated with petal senescence and flower drop. ACC was found on almond pollen and could thus contribute to a rapid production of ethylene by floral tissues (as it does in carnations). The later production of ethylene by explanted pistils (Figure 4) may also contribute to the senescence of floral parts. However, because more ethylene is produced following cross-pollination, which leads to an enhancement of embryo sac growth and, presumably, fruit development, it seems more likely that this ethylene contributes to fruit set and development rather than degeneration.

Cross-pollination does not automatically provide a guarantee of fruit set (especially this past year) therefore the data for ethylene production (Fig. 4) and embryo sac growth (Fig. 3) of cross-pollinated flowers are likely to include measurements on flowers which experienced unsuccessful, as well as successful, fertilization. If there was a way to discriminate between these two "classes" of cross-pollinated flowers then the differences between cross- and non-pollinated flowers shown by Figures 3 and 4 might have been much greater.

4. The data shown in Figure 5 indicate that the almond fruit is quite responsive to ethylene gas at fairly low concentrations and the data in Figures 6 and 7 suggest that application of the ethylene-releasing compound ethephon (at concentrations much lower than are usually employed in orchards) can also promote nut maturation. These data do not necessarily implicate ethylene in endogenous control of dehiscence and abscission. However, AOA (which inhibits normal ethylene

production) slows maturation as does STS (which is known to prevent ethylene influences on many plant tissues). Thus, it appears as though ethylene production by almond fruits does play a part in regulating dehiscence and abscission. Therefore it may be possible to influence these developmentally-important events by controlling the production of ethylene by almond fruits or manipulating ethylene levels in the orchard.

Publications

A paper on objective '2' is appended. It will be published in "Euphytica." We anticipate that another year's work on objectives '3' and '4' will result in publishable information.

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