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High Temperature Induced Bud-failure Symptoms in Vegetative Buds of Almond Plants in Growth Chambers[1]

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Abstract. **Plants of non-bud-failure (susceptible but without symptoms) 'Nonpareil' almond [*Prunus amygdalus* Batsch] produced severe bud failure (BF) symptoms internally in the growing point after 5 and 10 weeks exposure to high temperature (43°C ± 2) in a growth chamber, but not in comparable plants grown in a greenhouse at a normal temperature (about 27°C). High levels of abscisic acid (ABA) were detected in normal plants exposed to the high temperature. Buds on non-BF plants showed much lower ABA under the same conditions although there was more prior to the beginning of high temperature exposure. Little significant effects on gibberellic acid (GA)-like levels were detected.**

Exposure to high temperature as determined by orchard location has been associated with the degree of severity of noninfectious BF symptoms in almond trees (6). Similarly exposure to high temperatures in a greenhouse has resulted in BF expression in trees carrying BF potential (3).

Elsewhere, we have described the development of internal morphological symptoms in buds of BF plants (5). These have been related to the seasonal pattern of vegetative bud development (4). During symptom development (beginning mid-August) and just prior to this, a critical period of high temperature stress occurs. Buds on both normal and BF plants showed comparable GA-like levels, but buds on normal plants showed higher ABA levels than buds on BF plants.

However, under orchard conditions the causal effect of high temperature on bud development, symptom expression, ABA and GA-like levels could not be established.

The work reported in this paper was conducted to duplicate the field study in growth chambers and to determine the effects of high temperature on internal BF symptom development and

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on ABA and GA production in buds from normal and BF plants. Since BF plants failed to grow (5), plants from non-BF (symptomless but with BF-potential) plants were used to provide BF plants.

Materials and Methods

Plant materials and growing conditions. Almond buds were collected from trees of the 'Wells' clone (normal) in Davis, and clone 3-8-1-63 of 'Nonpareil' in Winters, as described elsewhere (5), and grafted onto 'Lovell' peach [*Prunus persica* (L.) Batsch] seedling rootstocks growing in 4-liter containers. The potted plants were kept in a greenhouse until mid-December, transferred to a lathhouse for about 2 months chilling, returned to the greenhouse and then to the lathhouse again until July. After shoots grew 30 to 70 cm, some containers were taken to the growth chamber for high temperature treatment, and the others taken to the greenhouse to serve as controls. Only plants from non-BF (symptomless) plants were used because high percentages of buds from BF trees failed during propagation. The greenhouse plants were maintained at about 27°C. The growth chamber plants were kept under a day-night cycle of 16 and 8 hr, respectively. The light was 53 klux at shelf-level sustaining the containers. The day and night temperature were maintained at 43° ± 1 and 18° ± 1, respectively. Plants were watered twice daily (8 AM and 5 PM), to avoid desiccation due to heat.

Fifteen to 18 young plants from each source and each treatment were harvested at 0, 5, and 10 weeks of treatment. Buds of 3 to 4 plants of each clone and collection were excised and prepared as described elsewhere (4, 5) for fresh weight, dry weight, water content, anatomical investigations, ABA and GA-like analysis.

Results

Anatomy of shoot apices. At the start of the experiment, buds appeared dormant with well formed bud scales, similar to buds on orchard trees at this time of year.

None of the buds from the normal plants showed any abnormality at the shoot apex, but exhibited the typical 3-4 layered tunica overlaying the thick corpus. Sections of the apices stained more or less uniformly with Azure B indicating low physiological activity. This situation held with buds collected prior to the beginning of the experiment and after 5 and 10 weeks. It also held with plants kept at lower temperatures in the greenhouse and with those at higher temperatures in the growth chambers.

Similarly, buds from the non-BF plants were normal internally prior to the start of the experiment. These plants had been exposed to outdoor shaded conditions at Davis, California in spring up to July. Under these conditions BF symptoms have not previously been encountered. Also no buds from non-BF plants were affected in the lower temperatures of the greenhouse at 5 and 10 weeks.

However, 75% of the buds from non-BF plants grown in the high temperatures (43°/18°) of the growth chamber became severely BF at 5 weeks and 90% at 10 weeks. These apices showed all the degenerative stages of BF symptom development previously described (6). The most predominant symptom exhibited at both sampling periods was the complete separation of distorted and compressed cells from the surrounding tissue by a periderm-like layer.

Fresh weight, dry weight, and water content of the buds. Fresh weight, dry weight and water content of buds from non-BF plants were less than those from normal plants and the difference persisted throughout the test (Table 1). This difference also had occurred in orchard trees (4). Buds on normal plants increased in fresh weight at both lower and higher temperatures at 5 weeks, but only with fresh weight at 10 weeks. This increase was less at the higher temperature.

BF buds showed very low % moisture at the start of the experiment. At lower temperatures bud size increased slightly, then decreased, but moisture percentage increased. At higher temperatures fresh weight increased by 5 weeks and % moisture was high. By 10 weeks, when severe necrosis was evident, moisture percentage dropped. Dry weight showed an increasing trend during this period. This pattern is similar to that shown in the orchard.

ABA content of buds. Buds on normal plants showed low c,t- and t,t-ABA levels prior to the start of the experiment (Fig. 1). At the cooler temperature, there was very little found at 5 weeks, but the amount increased somewhat at 10 weeks. At high temperature, on the other hand, there was a very large increase in both c,t-ABA and t,t-ABA at 5 weeks, amounting to about a 24× increase in c,t-ABA and 35× for t,t-ABA (Fig. 1) compared to the start of the experiment. By week 10, however, the ABA content at these high temperatures had decreased to low levels.

Buds from the non-BF plants showed significant amounts of ABA of both types prior to the start of the experiment (Fig. 1). However, at both the 5th week and 10th week sampling the amount of c,t-ABA in buds of the lower temperature plants was low, essentially the same as the normal plants. At higher temperature, c,t-ABA at 5 weeks was higher that those at lower temperatures, but the amount was only half of that of the normal plants while t,t-ABA was only 1/10 of the normal. At 10 weeks the amount was low and at a par with the normal.

GA-like substances. At the start of the experiment the promoting activity in the alkaline fraction (biologically active) was greater in extracts from buds on normal plants than those on non-BF plants (Fig. 2).

In buds from the normal plants, GA-like activity remained high after 5 weeks at the lower temperature, but decreased by half at 10 weeks (Fig. 2). At high temperature, however, the GA-like activity decreased substantially at 5 weeks and to a very low level at 10 weeks.

GA-like activity was relatively high in buds from the non-BF plants at the beginning of the experiment. When grown at lower temperature very little could be detected at 5 weeks; more was detected at 10 weeks but this was still less than a third of that found at the beginning.

At the higher temperature, GA-like activity remained more or less the same at both 5 and 10 weeks with some fluctuations.

The GA-like activity in the acid fraction (biologically non-active) of buds from normal plants increased slightly at 5 weeks and decreased slightly at 10 weeks at cool temperature, compared to the starting material (Fig. 2). At high temperature, GA-like activity decreased.

In buds from non-BF trees, acid fraction GA-like activity was lower than the normal and remained about the same at cool temperature. At high temperature, there was less GA-like activity at 5 weeks but substantially more at 10 weeks.

Discussion

Plants from non-BF sources exposed to high temperature developed typical BF symptoms, whereas similar plants growing at low temperature produced none of these symptoms. Internal symptoms of affected meristems are identical to those exhibited by BF buds in the field (6). Thus, we must conclude that high temperature affected the viability of buds of the non-BF materials which are either converted to BF or required higher/longer temperature stress to produce symptoms. The normal buds were equally subjected to high temperature stress as non-BF buds and either were not senitive or represent a much lower level of BF-potential. Whether the normal plants would have eventually produced injury at these high temperatures is beyond the scope of this experiment.

Thus, the results reported in this paper provide further confirmation that high temperature stress is a stimulating factor

Table 1. Weight, moisture, and damage of buds from normal and BF plants exposed to different temperatures.

Temperature treatment	No. weeks	Normal Fresh wt (mg)	Normal Dry wt (mg)	Normal H_2O (%)	Normal Buds damaged (%)	BF Fresh wt (mg)	BF Dry wt (mg)	BF H_2O (%)	BF Buds damaged (%)
Greenhouse at 27°C	0	2.0	1.1	45	0	1.4	1.1	21	0
	5	2.8	1.8	38	0	1.9	1.3	34	0
	10	3.1	1.5	52	0	1.4	0.8	43	0
Growth Chamber 43°/19°	0	2.0	1.1	45	0	1.4	1.1	21	0
	5	2.4	1.5	38	0	1.8	1.2	44	75
	10	2.5	1.4	44	0	1.6	1.3	19	90

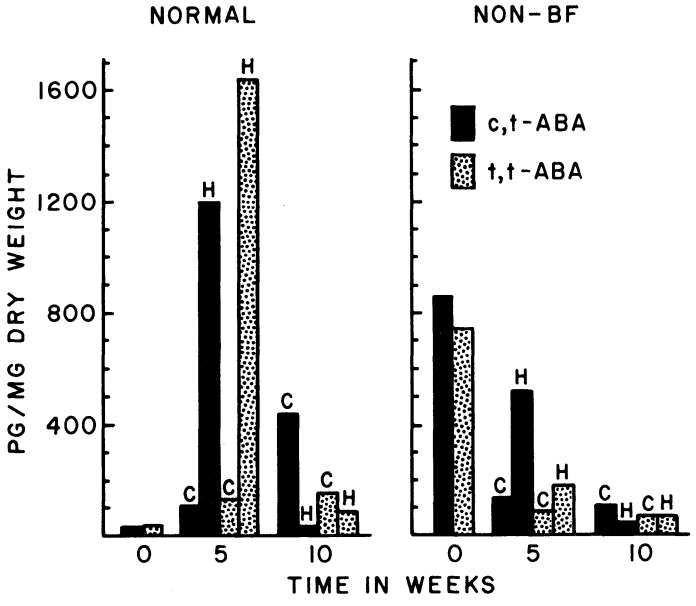

Fig. 1. Effects of temperature on cis, trans-ABA (c,t-ABA) and on trans, trans-ABA (t,t-ABA) in extracts of vegetative buds from normal and non-BF 'Nonpareil' almond plants growing in containers at 27°C (C) or 43°(H).

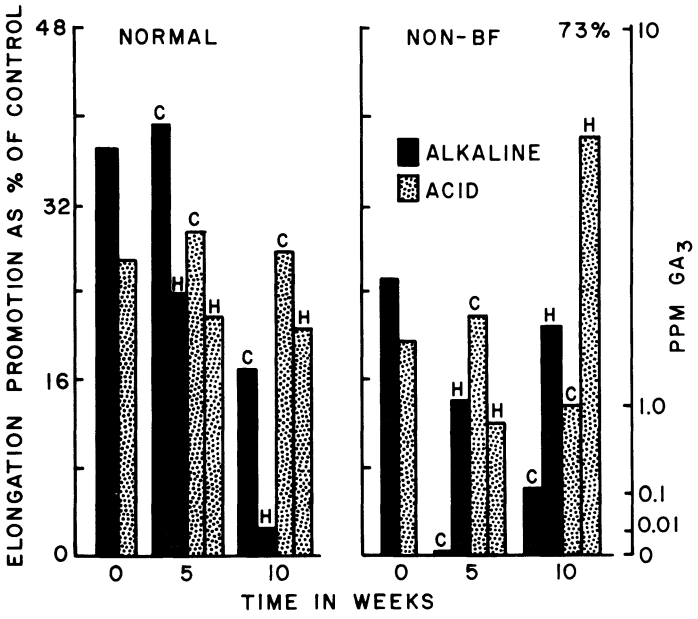

Fig. 2. Effect of temperature on GA-like substances in alkaline-ethyl acetate and acid-ethyl acetate fractions of extracts of vegetative buds (250 mg dry weight each) of normal and non-BF 'Nonpareil' plants growing in containers at 27°C (C) or 43°C (H). Extracts chromatographed on ITLC with isopropanol:ammonia:water (10:1:1, v/v) and bioassayed with lettuce hypocotyl test.

of BF symptom expression. Further, the evidence indicates that induction of ABA may play a key role in conditioning the normal plant to resist high temperature effects.

The large increase in both c,t- and t,t-ABA in normal buds suggests de novo synthesis of ABA rather than a release of a bound form. Milborrow and Noddle (8) reported de novo synthesis of ABA during water stress. ABA levels are increased by low temperature as well as by short photoperiods (1).

Many reports show ABA accumulation in growing tissue during water stress (8, 9, 10, 11, 12, 13, 14) to modulate stomatal behavior, which controls water and gas exchange, and retards growth to avoid stress injury. Such a mechanism may have operated under the high temperature stress conditions of these experiments. However, one cannot be sure whether the rise of ABA was due to the direct effect of high temperature, or to dehydration of the buds induced by the temperature.

Affected buds showed a water deficit (below 50% of dry weight), but this deficit existed before the start of the experiment, when symptoms were absent. This deficit could be a consequence of lack of enough ABA to control adequately the evapotranspirational system of plants (stomata). This further suggests that disruption in ABA metabolism precedes development of a water deficit in buds.

A more plausible explanation for our results can provide a working hypothesis for future research. Both normal and BF plants produce gibberellins in conjunction with growth activity. Plants exposed to stress, such as, high temperature or low moisture, react by producing ABA which functions to slow down/stop growth and induce "resistance." In BF plants the capacity to produce ABA is repressed such that the plant becomes increasingly sensitive to high temperature stress. Thus, the induction of high temperature resistance may be analogous to the induction of rest (2), induction of cold resistance, i.e., hardening-off, induction of drought resistance (7, 10, 13, 14) and others (8, 9, 11, 12). All involve the repression of growth and cellular activity which may be mediated through ABA.

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Antitranspirant Effects on Water Relations and Fruit Growth of Rabbiteye Blueberry[1]

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Abstract. **Vapor Gard (VG), a polymer of β-pinene applied at concentrations of 1.5 or 2.5% (by volume to rabbiteye blueberries (*Vaccinium ashei* Reade) influenced leaf temperatures, water balances, berry weight, and time of fruit harvest of plants with and without drip irrigation. A 2.5% VG spray uniformly covered entire leaves and increased midday xylem pressure potentials (Ψ_x) by 50% and leaf resistances (r_l) by 400%, decreased transpiration (T) by 80%, raised average leaf temperature by 2.2°C and resulted in phytotoxicity and leaf drop. A 1.5% spray did not significantly increase Ψ_x but doubled r_l and decreased T by 60% with no toxicity symptoms. When all cultivars are combined, the 1.5% spray applied to plants with and without irrigation increased berry weight by 31 and 17% but delayed berry maturation and decreased percentage total soluble solids of mature berries by 26 and 24%, respectively. Vapor Gard did not significantly change yields when used alone.**

Rabbiteye blueberry bushes are a shallow fibrous rooted plant tolerant to high temperature and capable of surviving extended periods of drought (11). They flower in March and are harvested in June, which is normally Florida's driest season. There have been a number of reports of antitranspirants (AT) improving the water balance of both irrigated and nonirrigated crops (5, 6, 7, 15, 16, 21, 22, 23, 26, 28, 29, 30). Enhanced fruit growth appears to depend upon the degree of film on the leaf surface (6). Larger fruit and a reduction in shrinkage resulted from increases in leaf water potential produced by greater resistance in the leaf-air pathway (6). There are also reports that AT will result in reduced dry weight of fruit and a delay in ripening if applied too early (15, 23).

This report was part of an overall water relation study of rabbiteye blueberry to determine if AT, with and without irrigation, could increase yields and fruit size without causing serious side effects such as later ripening or reduced soluble solids.

Materials and Methods

The experimental site near Gainesville Florida was a 0.8 ha planting of 5-year old 'Woodard', 'Bluegem', and 'Tifblue' rabbiteye blueberries spaced at 2 m within and 4 m between rows. Three plants of each cultivar were sprayed with 2.5% VG (Miller Chem. & Fert. Co., Hanover, Pa) on April 15 to provide preliminary information on rate for future applications.

The experimental design consisted of randomized blocks of 4 treatments and 3 replications with 4 plants per replication. Treatments were 1.5% (by volume) VG applied May 2 to bushes without irrigation, 1.5% VG applied May 2 to bushes with drip irrigation (21.6 liters per day) irrigation alone, and a non-irrigated control.

Soil moisture was monitored weekly with a Troxler neutron probe at a mean depth of 20 cm from March until July (4). Access tubes were placed under the plant canopy near 6 bushes with and 6 without irrigation.

Vapor Gard was applied with a small power sprayer. Leaf temperatures of 9 typical sun-exposed leaves each of treated and nontreated plants were sensed using 18 thermocouples appressed to leaf undersides with white paperclips.

Both Ψ_x and r_l were measured on sun exposed leaves 6 to 10 cm from the terminal respectively, with a Scholander pressure chamber (18, 24, 25) and a diffusion porometer (20, 31), during midday periods of maximum water stress. Data for all cultivars were combined, based on results from an earlier

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