

TuMV-C2 and TuMV-C3, but 'Crusader' and 'W-R 65 Days' are resistant only to TuMV-C2. The same reasoning can be applied to other cases (Table 1) and to TuMV-C4, strongly indicating that several genetic factors for immunity or resistance are involved. These appear to be inherited independently of each other.

Viral strain specificity is an important factor that must be considered in developing TuMV-resistant cultivars, since their performance will depend upon the presence and distribution of specific strains of this virus in a given locality. The Chinese lines PI 391560(1), PI 418957, PI 418959, and PI 419069, which are multi-resistant, represent valuable germplasm material that can be used for intra- and interspecific gene transfer. Immunity or resistance to TuMV in a number of other crucifers, such as broccoli, cauliflower, collards, cabbage, mustard, and turnip is greatly needed. A few amphidiploid hybrids of *B. campestris* ssp. *pekinensis* × *B. oleracea* var. *capitata* are available commercially (Hiratsuka N^o1, Hakuran, etc.). Thus, it may not prove difficult to transfer TuMV resistance factors from Chinese cabbage to cabbage, particularly those cultivars that are placed in cold storage.

Most of the cultivars tested were homogeneous populations, but some TuMV-susceptible lines included a few resistant individuals, and some TuMV-resistant lines included susceptible plants. Also there was some variation in seed lots of certain cultivars. Lim et al (5) also noted varying degrees of heterogeneity in 486 Chinese cabbage lines that were screened for resistance to an isolate of TuMV from Taiwan.

Although TuMV-C1, TuMV-C2, and TuMV-C3 are representative of a large number of isolates of the virus occurring in New York State, the isolation of TuMV-C4 from a multi-resistant plant indicates the complexity of the problem and the difficulties that might be encountered in obtaining durable sources of resistance.

Resistance in TuMV in *B. campestris* ssp. *pekinensis* has been very useful in identifying and categorizing strains of this virus. Likewise, by using these strains it has been possible to uncover additional immune or resistant cultivars of this species.

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Abscisic Acid in Pistachio as Related to Inflorescence Bud Abscission¹

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Abstract. Neither abscisic acid (ABA) levels in developing kernels nor in the developing inflorescence buds themselves were found to be related to abscission of inflorescence buds and consequent alternate bearing in pistachio (*Pistacia vera* L.).

Alternate bearing in pistachio results from abscission of inflorescence buds during heavy crop years and retention of buds during light crop years (3). Bud abscission occurs primarily in July and August. Crane et al. (4, 5) showed that the degree and rate of bud abscission were closely related with number of nuts on the branch. They increased as leaf area was decreased. Also, rapid seed development was found to occur at the same time as inflorescence bud abscission. It appeared, therefore, that competition for metabolites by the

seed was responsible for the abscission phenomenon. During that critical period, however, neither carbohydrate nor nitrogen levels in bark and wood of nut bearing branches were found to be appreciably different from those in nonbearing branches (6, 7). Levels of nitrogen in buds of bearing trees were the same as those in buds of nonbearing trees (14). The information at hand, therefore indicates that unfavorable nutrition is not the primary cause of bud abscission.

Bud drop was found to increase progressively as leaf area decreased or number of nuts per branch increased. Crane et al. (5), therefore, suggested that an abscission-inhibiting hormone from the leaves may be the limiting factor in bud abscission. Alternatively, it is conceivable that the developing nuts might control bud abscission by producing an abscission-promoting substance(s). Seeds are rich sources of hormones (2), including ABA (9, 13). Growth inhibitors, such as ABA, are potential

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abscission-promoting compounds (1), and the synthesis of ABA in developing kernels may trigger inflorescence bud abscission. High levels of ABA in the fruit have been shown to be closely related to flower and young fruit abscission in lupine (18) and in cotton (8). As with pistachio (4, 5, 14), removal of developing fruits reduced abscission of flower buds in pea (12) and in soybean (11). It was shown recently that developing pea fruits brought about increases in ABA concentration in axillary buds and also induced abortion of young fruits (16, 17). ABA levels in developing pistachio kernels and inflorescence buds were measured to determine if a relationship existed similar to that found in pea.

Materials and Methods

Plant materials and sampling. Nine-year-old 'Kerman' pistachio trees growing in a large block at the Wolfskill Experimental Orchard, Winters, Calif., were used as a source of samples. On May 22, 1978, all nuts were removed by hand from 5 trees. Five adjacent trees were left intact as controls. Five branches with about average current growth in length were tagged on each of the 10 trees for periodic determination of inflorescence bud drop. At least 5 branches each from bearing and defruited trees were sampled periodically from June to August, placed in crushed ice and brought to the laboratory. Inflorescence buds were excised from the leaf axils on current wood and weighed. A random sample of at least 100 fruits from the 1-year-old wood was weighed, after which the seeds were excised and weighed. The samples were stored at -20°C and later freeze-dried.

Extraction. The freeze-dried seed samples were ground to a fine powder in a Moulinex coffee grinder. The buds were finely chopped with a razor blade. Buds (150-200 mg) and seeds (250 mg) were homogenized separately in 10 ml of 80% methanol (MeOH) for 1 hr. Extraction with fresh MeOH for 30 min was repeated twice. The extracts were combined and concentrated to the aqueous (aq) phase in a rotating evaporator at 40°C ; the volume was adjusted to 9 ml with water.

Fractionation. The concentrated extract was adjusted to pH 8.0 with 1 N NaOH, frozen, thawed, then partitioned 3 times with 1/3 volume petroleum ether (PE) and with 1/3 volume ethyl acetate (EtoAc). The bulked EtoAc fraction was back-washed with 1/4 volume water. The water was combined with the aq phase. The aq phase was adjusted to pH 3 with 1 N HCl and the solution partitioned 3 times with 1/3 volume EtoAc. The bulked EtoAc was back-washed with 1/4 volume H_2O and H_2O was combined with the pH 3 aq phase. The acidic EtoAc was evaporated under N_2 gas and the residue dissolved in 0.1 M pH 8 phosphate buffer. Polyvinylpyrrolidone (pre-washed in MeOH, EtoAc, and acetone and airdried) (PVP) (100 mg) was added to the solution, stirred gently for 15 min, centrifuged, and PVP discarded. Samples were purified further by fractionating with PE at pH 8 and with EtoAc at pH 8 and 3. The acidic EtoAc fraction was evaporated under N_2 to dryness.

The pH 3 aq phase containing the bound form of ABA was treated with 12 N HCl for 1 hr at 60°C . After adjusting the pH to 3, the solution was partitioned 3 times with EtoAc; the bulked organic phase was dried under N_2 .

Gas-liquid chromatography. The dried residues were suspended in MeOH and methylated (Me) with diazomethane. The methylated samples were dried under N_2 and the residue was dissolved in EtoAc. A 1- μl aliquot was injected into a Hewlett-Packard Model 5730A gas chromatograph equipped with a ^{63}Ni electron capture detector and a spiral glass column (1.22 m \times 6 mm) packed with 3% OV 101 (methyl silicon gum rubber) over 100-120 mesh Chromosorb Q. The injection port, column, and detector temperatures were 250° , 190° , and 250°C , respectively. The carrier gas, CH_4 -Argon, had a flow rate of 30 ml/min. The Me-ABA was tentatively identified by comparison of R_T with authentic samples of Me-ABA

and Me-racemic ABA. The detector signal was interfaced with a Perkin-Elmer Sigma 10 peak area integrator with the following parameters: baseline was drawn by dropping a vertical to a baseline from the valley between peaks and the area, base and skim sensitivity were preset on the data system. Both peak height and integration of peak area were used for quantification. The percent recovery of ABA by the extraction procedure outlined above was about 80%.

Results

Morphological development of the pistachio fruit is like that of other drupes, except that the nucellus and integuments do not undergo extensive growth until about a month after the endocarp reaches its ultimate size. The endocarp attained its ultimate size by May 15, 1978 and embryonic development was evident on June 15 (Fig. 1). Once embryo development began, its growth was sigmoidal and its ultimate size was attained in about 8 weeks. The weight gained by the fruit after June 15 was practically all due to seed development. The seed comprised about 40% of the final fruit dry weight.

Initially both *trans-trans* ABA (t-ABA) and *cis-trans* ABA (c-ABA) were found in very low amounts, each less than 0.01 ng/seed (Fig. 2A). Both forms increased gradually during June. Rapid increases began in early July with the onset of seed enlargement. C-ABA rose dramatically to a high of about 0.74 ng/seed on Aug. 30 while t-ABA increased slightly to 0.06 ng/seed in mid-August and declined (Fig. 2A). There was more than a 5-fold increase in c-ABA per fresh weight of seed during the period of seed development (Fig. 2B, 2C). The concentration increase occurred, however, well after bud abscission had begun and reached maximum about the time bud abscission ceased on nut bearing trees. No bound ABA was detected in the seed.

The axillary inflorescence buds on current growth grew rapidly until the end of June, after which growth was slight (Fig. 3C). We had shown previously that development of the inflorescence structure is complete by June 30 and no differentiation of floral parts occurs during the summer months (15). The fresh weight of buds on fruiting trees was about 25% less than that on nonfruiting trees during the period of bud drop (Fig. 3C).

Abscission of the smaller inflorescence buds at the basal ends of the shoots began about mid-June and progressed distally. On both fruiting and defruited branches, the basal 2 or 3 buds were poorly developed and were subtended by small trifoliate

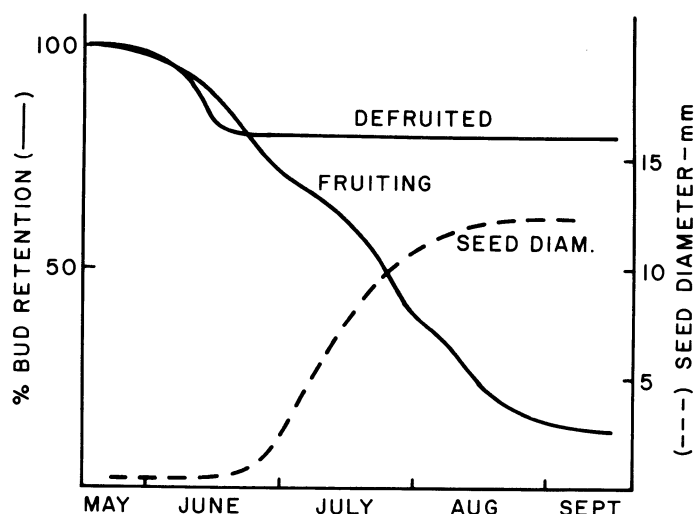


Fig. 1. The relationship of crop load and period of seed growth to inflorescence bud abscission in the 'Kerman' pistachio.

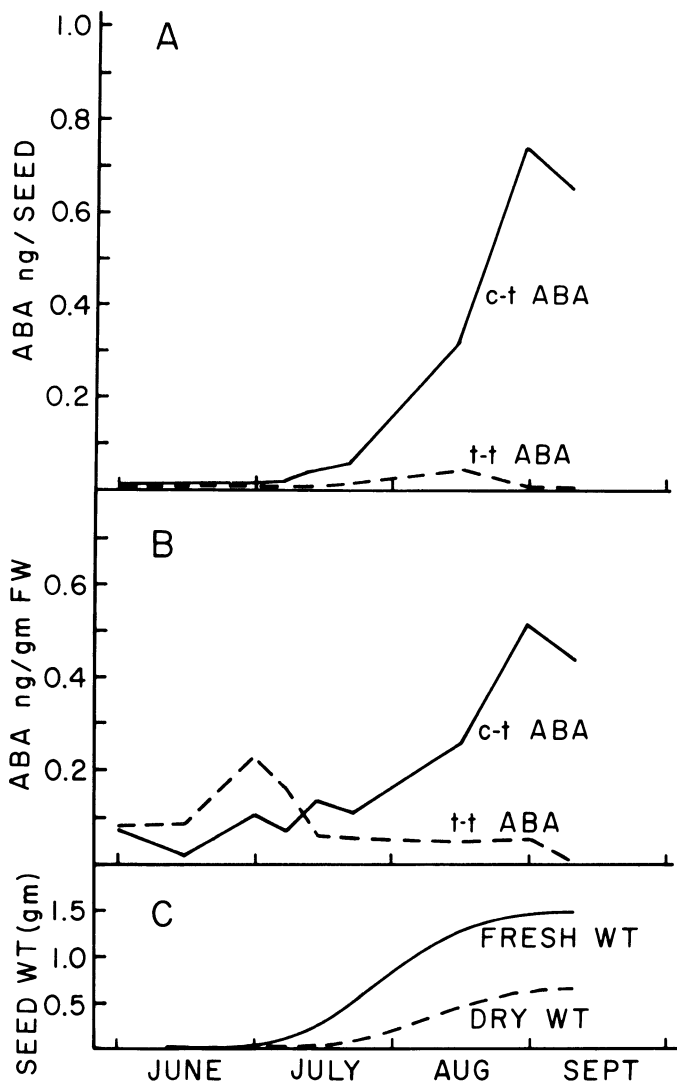


Fig. 2. ABA content of developing 'Kerman' pistachio seeds.

leaves. These basal buds abscised from both fruiting and defruited branches. As in previous years, bud drop began when embryo development became macroscopic and continued at about a constant rate until the end of August (Fig. 1). A correlative relationship existed between seed enlargement and inflorescence bud abscission. The presence of developing nuts to maturity ultimately caused 85% of the buds to abscise, while nut removal in May resulted in only 20% bud abscission (Fig. 1).

C-ABA was the only form detected in inflorescence buds. Changes in amounts of c-ABA in developing buds of fruiting and defruited trees followed similar patterns (Fig. 3A). Dramatic increases in c-ABA occurred in both types of buds just prior to onset of bud abscission, maximum amounts were maintained for relatively short periods which, in turn, were followed by gradual declines. Changes in concentration of c-ABA in the buds were somewhat different from changes in quantities (Fig. 3B), the difference being a reflection of difference in bud weight (Fig. 3C). The concentration of c-ABA in buds of fruiting trees declined to that of buds on defruited trees by the end of July but the buds in the former continued to abscise.

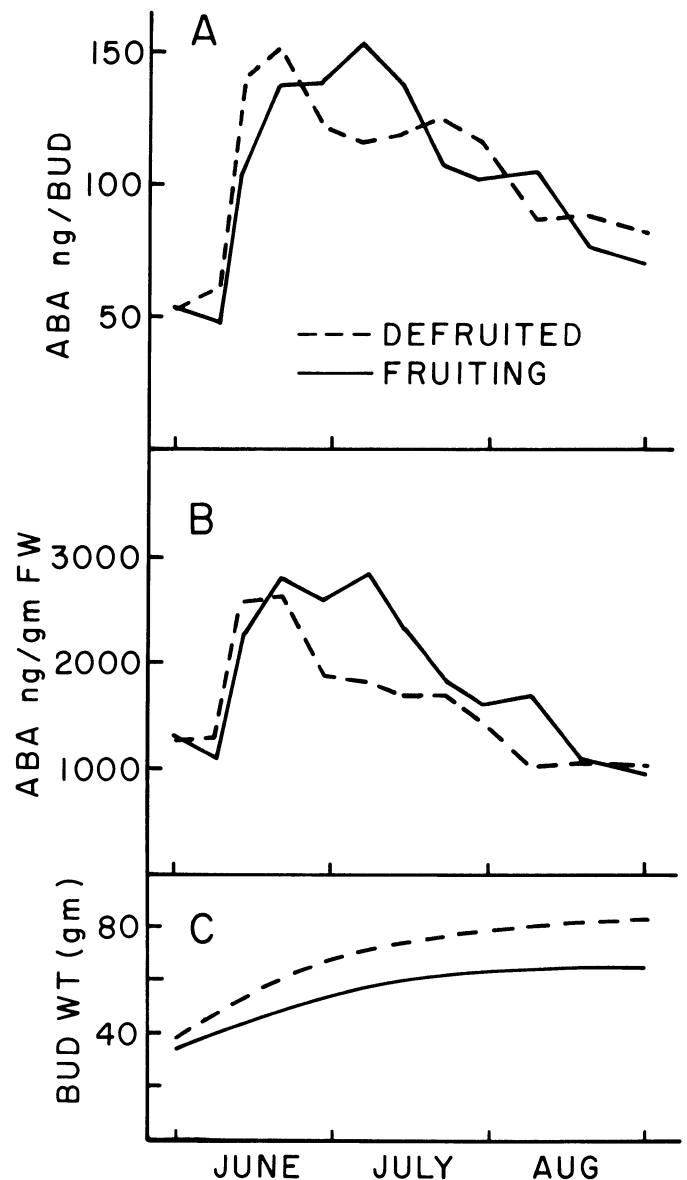


Fig. 3. Changes in ABA content of developing inflorescence buds on fruiting and defruited 'Kerman' pistachio trees.

Discussion

It is clear that abscission of pistachio inflorescence buds is under the influence of developing seeds and is a correlative phenomenon. As ABA in the seed increases during the period of bud abscission, it is tempting to associate that increase with bud abscission. The data presented indicate, however, that there is no relationship between c-ABA in the bud and crop load and between c-ABA in the bud and the process of bud abscission. The inflorescence buds began abscising in mid-June from defruited as well as fruiting trees, which indicates that something in addition to seed development is also involved in abscission. Developing seeds markedly affected bud abscission later on. ABA in the seed apparently is not involved in regulating bud abscission; since 1) abscission began prior to the rise in ABA level in the seed, 2) rate of abscission was constant while ABA in the seed increased and then decreased, and 3) the ABA concentration in the seed was about 10×10^3 times less than in buds.

The increase in seed ABA in late June was probably a reflection of increased metabolic activity associated with rapid

seed development, as has been demonstrated with periods of rapid growth in other crops (9, 10, 13). The continual rise in ABA beginning in July paralleled dry matter accumulation. Similar changes in ABA have been shown to occur in 'Winter Nelis' pear seed (13).

Levels of ABA in the buds also do not appear to be related to bud abscission for 3 reasons. Firstly, changes in ABA levels were similar in buds from both fruiting and defruited trees during the period of bud abscission. Secondly, distal inflorescence buds on fruiting trees continued to abscise for a period of 4 weeks after ABA level in the buds fell to that of buds on defruited trees. Thirdly, bud abscission on fruiting trees progressed at a constant rate although ABA levels fluctuated in the buds.

Other growth substances must be involved in abscission of pistachio inflorescence buds. Foliar application of auxin in June retarded bud drop (4) but, as determined later, did not prevent bud necrosis and eventual abscission (F. Takeda, unpublished). Application of 6-benzylamino purine to buds in June retarded their abscission by promoting growth and differentiation but when growth and development ceased in September abscission occurred (F. Takeda, unpublished). Thus, the evidence accumulated to date does not contradict the concept of an abscission-inhibiting hormone(s) produced in the leaves as controlling bud drop (5).

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Root Distribution of 'Coville' and 'Lateblue' Highbush Blueberry under Sawdust Mulch¹

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Abstract. The root system of the cultivated highbush blueberry (*Vaccinium corymbosum* L.) is primarily composed of fine, thread-like roots less than 1 mm in diameter. Essentially all of the roots of 7-year-old 'Lateblue' bushes were located within the area between the crown and the dripline. Most of the roots on 13-year-old 'Coville' bushes were also within this area. Roots were found 180 cm from the crown and at depths of 81 cm. Roots were in the decomposing low layers of the sawdust mulch but not within the upper, non-decomposing layers.

The root systems of many fruit-bearing plants have been studied in an effort to correlate the nature and extent of the root system with the ability to tolerate drought or pathogens (1, 2, 3, 9). Extensiveness of apple tree root systems was re-

ported by Wiggans (12), who noted that mature trees developed roots to depths and breadths of 9 m or more. Proebsting (10) studying the root systems of certain stone fruits and pear, reported that the major concentration of roots occurred at depths of about 0.6-1.5 m, though some roots penetrated to a depth of about 4.5 m. Green and Ballou (6), working with apples, reported a dense system of roots in the surface 15 cm of soil. This occurrence appeared independent of soil management systems such as mulching or clean cultivation.

There has been little research on the nature of root systems of bush fruits. This is especially true of the highbush blueberry.

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