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Effect of Kinins on Fruit Set and Development in *Vitis Vinifera*

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Experiments were performed in 1963 and in 1964 on four varieties of *Vitis vinifera* to determine the responses to the kinins, BTP (6-(benzylamino)-9-(2-tetrahydropyranyl)-9H-purine) and BA (benzyladenine). Both kinins were effective in increasing fruit numbers, and in some instances fruit growth. BTP was more effective than BA. BTP applied to Black Corinth clusters at bloom produced berries three to four times larger than the controls, and the pedicels were much thickened. Black Corinth was treated at bloom with BTP, gibberellin, and/or an auxin, 4-CPA. Gibberellin alone and a mixture of all three compounds produced the largest berries. Berries were elongated as a result of the gibberellin. 4-CPA and BTP produced about equally sized, almost round berries. Treatment of the apical half of Black Corinth clusters at bloom with BTP at 1,000 ppm enlarged only the treated berries. Treatment of the basal portion resulted in some movement towards the apical portion.

When Thompson Seedless clusters were treated with BTP and the mixture of BTP and gibberellin, the number of berries was increased. Berry size was increased only by gibberellin or the mixture of the two compounds. At the shatter stage BTP at 1,000 ppm increased berry size although not nearly so much as gibberellin alone or the gibberellin-BTP mixture. BTP or BA applied to Thompson Seedless at shatter stage increased numbers of berries.

BTP in range from 125 to 1,000 ppm increased berry numbers in Muscat of Alexandria, as did dipping of clusters on partially defoliated shoots. The set of berries on emasculated flowers of Black Corinth, Thompson Seedless, and Tokay was increased by BTP.

Clusters of Black Corinth, Thompson Seedless, Tokay, and Almeria were dipped in BTP at 1,000 ppm at bloom stage. Far more ovaries or berries fell from the treated than from the control clusters at shatter, except for Tokay. There was a tremendous increase in the number of seedless berries on the treated clusters of Almeria.

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Robert J. Weaver, J. van Overbeek, and Robert M. Pool

Effect of Kinins on Fruit Set and Development in *Vitis vinifera*[1,2]

INTRODUCTION

THE WORK of Gustafson (1936)[3] and subsequent experiments led to the general conclusion that auxin is the controlling factor in fruit set and growth. Data collected over the past six or seven years show that gibberellins, in addition to auxins, are also concerned with fruit set and development (Crane *et al.*, 1961; Weaver and McCune, 1960). It is now accepted that the third major group of plant hormones, the kinins, occurs in many fruits (Miller, 1961; Steward and Shantz, 1959, van Overbeek, 1962). Many experiments on tree fruits and other plants have shown that exogenous auxins and gibberellins are very effective in promoting fruit set and development. In 1962, the authors were the first to demonstrate that an applied kinin caused an increase in berry size of Black Corinth grape (Weaver and van Overbeek, 1963), and in subsequent experiments they demonstrated that a kinin could induce setting of fruit in grapes (Weaver *et al.*, 1965). Crane and van Overbeek (1965) demonstrated that a kinin would induce parthenocarpic fruit set and growth in the fig.

Our experiment on Black Corinth in 1962 stimulated us to perform further experiments in 1963 and 1964 to determine the importance of kinins in fruit set and development in several varieties of grapes. The combined effects of kinin, auxin, and gibberellin were studied, followed by a study of the effect of treating portions of clusters at bloom stage. Experiments were also performed on emasculated clusters and on clusters in which set was reduced by partial defoliation. Varieties which normally set poorly were included in these experiments.

MATERIALS AND METHODS

The experiments were conducted in an irrigated vineyard at the University of California, Davis. Used were mature vines of Black Corinth, Thompson Seedless, Muscat of Alexandria, Tokay, and Almeria (Ohanez). Black Corinth and Thompson Seedless were usually pruned to four canes, bearing from 8 to 12 buds; Almeria and Tokay were cordon-trained and spur-pruned, and Muscat of Alexandria was head-trained and spur-pruned.

Thompson Seedless was cluster-thinned to about 20 clusters per vine.

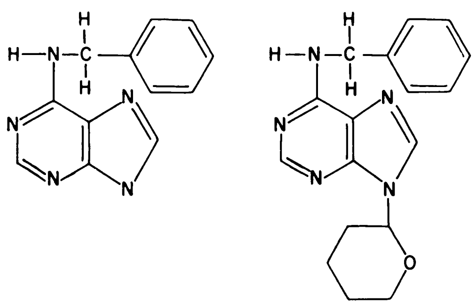
[1] Submitted for publication January 22, 1965.

[2] This work was supported in part by a grant from Shell Development Laboratory, Modesto, California.

[3] See "Literature Cited" for citations referred to in the text by author and date.

Where possible, an equal number of clusters was left on each cane, with all clusters being removed from the head of the vine. Berry thinning was accomplished by removing the apical half of the cluster unless otherwise stated.

The two kinins used were benzyladenine (BA) and 6-(benzylamino)-9-(2-tetrahydropyranyl)-9H-purine, a benzyladenine in which the hydrogen on nitrogen nine is substituted with a nonpolar ring structure of tetrahydropyran. The latter compound is hereafter referred to as BTP.



BA BTP

Both compounds were supplied by the Shell Development Company as a 1 per cent solution in isopropyl alcohol. Dilutions of the 1 per cent solution were made with distilled water so that a 1,000 ppm solution would contain 10 per cent isopropyl alcohol, and a 100 ppm solution of kinin would contain 1 per cent isopropyl, etc. To the final solution 0.1 per cent Tween 20 was added as a wetting agent. Potassium gibberellate was used for the gibberellin.

In dipping experiments clusters were momentarily immersed in a hormone solution. Sprays were applied with a three-gallon hand sprayer unless otherwise stated. The clusters and foliage were heavily sprayed until runoff. When necessary, cardboard shields were used to confine the spray to the intended area.

In an experiment in which shoots were partially defoliated, all foliage was removed except a one-square-inch stub on the leaf above the cluster (fig. 1). The purpose of partial defoliation was to reduce the percentage of set on untreated clusters (Coombe, 1962). These shoots were ringed with a knife, then the cluster was dipped.

Emasculation was performed as soon as five to ten calyptras had fallen from the flower cluster. The open flowers were removed, and the remaining ones were emasculated by pulling off the calyptra and the stamens with forceps. The purpose of emasculation was to remove the pollen, a rich source of plant hormones. To eliminate or minimize cross-pollination, flowers were emasculated at a time of little or no wind. Clusters were dipped immediately after emasculation. The clusters were then bagged for about ten days to prevent pollination while the stigmas were yet receptive. When the bags were removed counts were made of the ovaries that had fallen from the clusters, and at harvest time data was obtained on number of berries per clus-

Fig. 1. Diagram of partially defoliated and ringed shoot of Muscat of Alexandria for study of fruit set. Arrow denotes ring.

ter, berry size, and sometimes degree of maturation and, in one experiment, on percentage of hard seeds. Sometimes observations were also made on pedicel size.

Average berry size was determined either by weighing 100 or 200 berries in duplicate. A Balling hydrometer and hand refractometer was used to determine the percentage of total soluble solids in the juice. Total acidity was determined by diluting 10 ml of the juice to 50 ml with distilled water, and titrating with 0.133 N/NaOH, using phenolphthalein as an indicator. Results are expressed as gm of tartaric acid per 100 ml of juice—approximately the percentage of acid.

EXPERIMENTATION AND RESULTS

RESPONSE OF THREE VARIETIES TO KININS

The varieties Black Corinth, Thompson Seedless, and Muscat of Alexandria were tested. The purpose of this experiment was to determine whether grape flowers and berries respond to the third class of plant hormones, the kinins, in a manner similar to that produced by auxins and gibberellins.

Black Corinth

In these experiments the effect of BTP was compared to that of BA. Combined effects of BTP with gibberellic acid and/or an auxin, 4-CPA (4-chlorophenoxyacetic acid) were studied, and responses of Black Corinth to treatment of various portions of the clusters with BTP were followed.

Effect of BTP and BA. On June 4, 1963, ten uniform clusters at the full bloom stage were dipped in BTP or benzyladenine at 0, 15, 31, 62, 125, 250, or 1,000 ppm. Full bloom was considered to be that stage of development at which approximately 70 per cent of the calyptras had fallen. Another set of clusters served as untreated control. Ten clusters were used per treatment.

Ten days after treatment the pedicels of clusters treated with either of the kinins at concentrations of 250 ppm or higher were thicker. By July 1, BTP in range from 250 to 1,000 ppm had greatly increased berry size. BTP at 1,000 ppm resulted in berries that were almost all three to four times larger than the controls. The pedicels were five times the thickness of the controls. Some increase in berry size had resulted from the BA, but not nearly so much as from BTP.

The grapes were harvested on August 19 (table 1, fig. 2). The data showed that berry size had been increased by BTP in range from 125 to 1,000 ppm. An increase also resulted from BA but to a lesser degree. Even though the control data on the numbers of berries per cluster were lost, it is clear that there was an increasing number of berries with higher concentrations of kinins. Degrees Balling was least at the highest concentration of BTP. Around 50 per cent of the total number of berries remained green in clusters treated with BA in range from 62 to 1,000 ppm.

In another experiment entire vines were sprayed with BTP or BA at 0, 62, 250 and 1,000 ppm. Results were similar to those of the dipping treatment, and therefore are not given separately.

Effect of BTP, gibberellin and 4-CPA. On May 25, 1964, three or four days after full bloom, ten clusters were dipped in BTP at 1,000 ppm. Other series of clusters were dipped in 4-CPA at 60 ppm, gibberellin at 100 ppm, or a mixture of the three regulators consisting of BTP at 333 ppm, gibberellin at 33 ppm, and 4-CPA at 20 ppm. High concentrations were used so that very large berries could be obtained.

The fruit was harvested on August 13. The largest berries were produced by gibberellin alone and by the mixture of compounds (table 2). These berries were elongated, probably as a result of

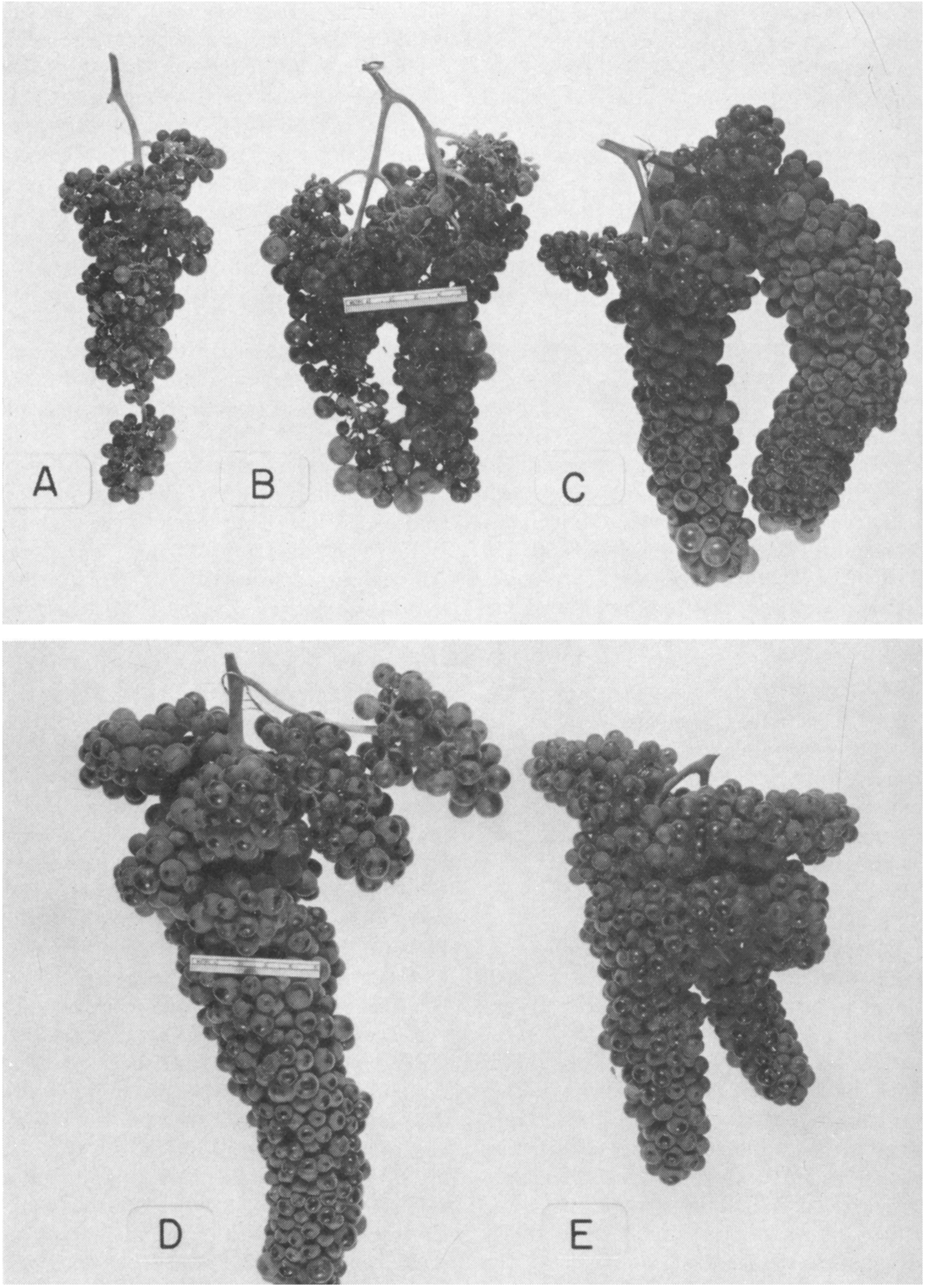


Fig. 2. Black Corinth clusters at harvest, 76 days after dipping in BTP at 0 ppm (A); 62 ppm (B); 500 ppm (C); and 1,000 ppm (D). (E), girdled but not dipped. With higher concentrations of kinin the berries become progressively larger. (Photographed August 19, 1963.)

TABLE 1

DATA AT HARVEST (AUGUST 19, 1963) FOR BLACK CORINTH GRAPES DIPPED IN BTP OR BA AT FULL BLOOM ON JUNE 4

(Figures are averages of ten replicate clusters)

Kinin	Concentration of kinin	Number of berries per cluster	Weight per berry	Degrees Balling	Total acid, gm tartaric per 100 ml
	ppm		*gm*		
BTP	1,000	728	0.53	16.1	1.24
	500	739	0.52	18.0	1.26
	250	261	0.41	18.0	1.20
	125	468	0.31	18.2	1.26
	62	449	0.19	20.4	1.22
	31	95	0.16	....	....
	15	172	0.17	20.3	1.36
BA	1,000	406	0.37	17.9	1.32
	500	338	0.29	18.5	1.26
	250	297	0.30	19.8	1.31
	125	311	0.30	17.5	1.32
	62	263	0.28	19.1	1.30
	31	83	0.18	19.7	1.21
	15	111	0.17	19.1	1.25
Control	0	*	0.19	*	*
L. D. S. at 5 per cent	...	...	0.05	2.6	....

* Control data for set were lost.

gibberellin (fig. 3). Auxin (4-CPA) and BTP produced about equally sized, almost round berries. The greatest number of berries was produced by 4-CPA and the smallest number by gibberellin, with BTP holding a middle position. About 15 per cent of the berries treated with BTP contained hard seeds. The degree of maturation as shown by degrees Balling and total acid showed little difference among treatments.

Treatment of portions of clusters. Three or four days after full bloom on May 25, 1964, the apical halves of ten clusters were dipped in BTP at 1,000 ppm. The basal halves of another series of clusters were sprayed with a De-Vilbiss No. 15 atomizer sprayer, and one series of clusters served as the untreated control. During spraying, cluster parts not intended to be sprayed were wrapped in waterproof plastic

TABLE 2

DATA AT HARVEST (AUGUST 13, 1964) FOR BLACK CORINTH GRAPES DIPPED IN BTP, 4-CPA, AND/OR GIBBERELLIN

(Figures are averages of ten replicate clusters)

Treatment			Number of berries per cluster	Weight per berry	Number of seeded berries per cluster	Degrees Balling	Total acid, gm tartaric per 100 ml
Concentration of 4-CPA	Concentration of gibberellin	Concentration of BTP					
ppm	*ppm*	*ppm*		*gm*			
0	0	0	413	0.28	2	21.6	1.04
60	0	0	700	0.56	..	21.0	0.87
0	100	0	320	1.52	..	20.8	0.89
0	0	1,000	505	0.61	75	19.9	1.15
20	33	333	...	1.47	..	20.2	0.97
L. S. D. at 5 per cent...	..	...	126	0.14	..	....	....

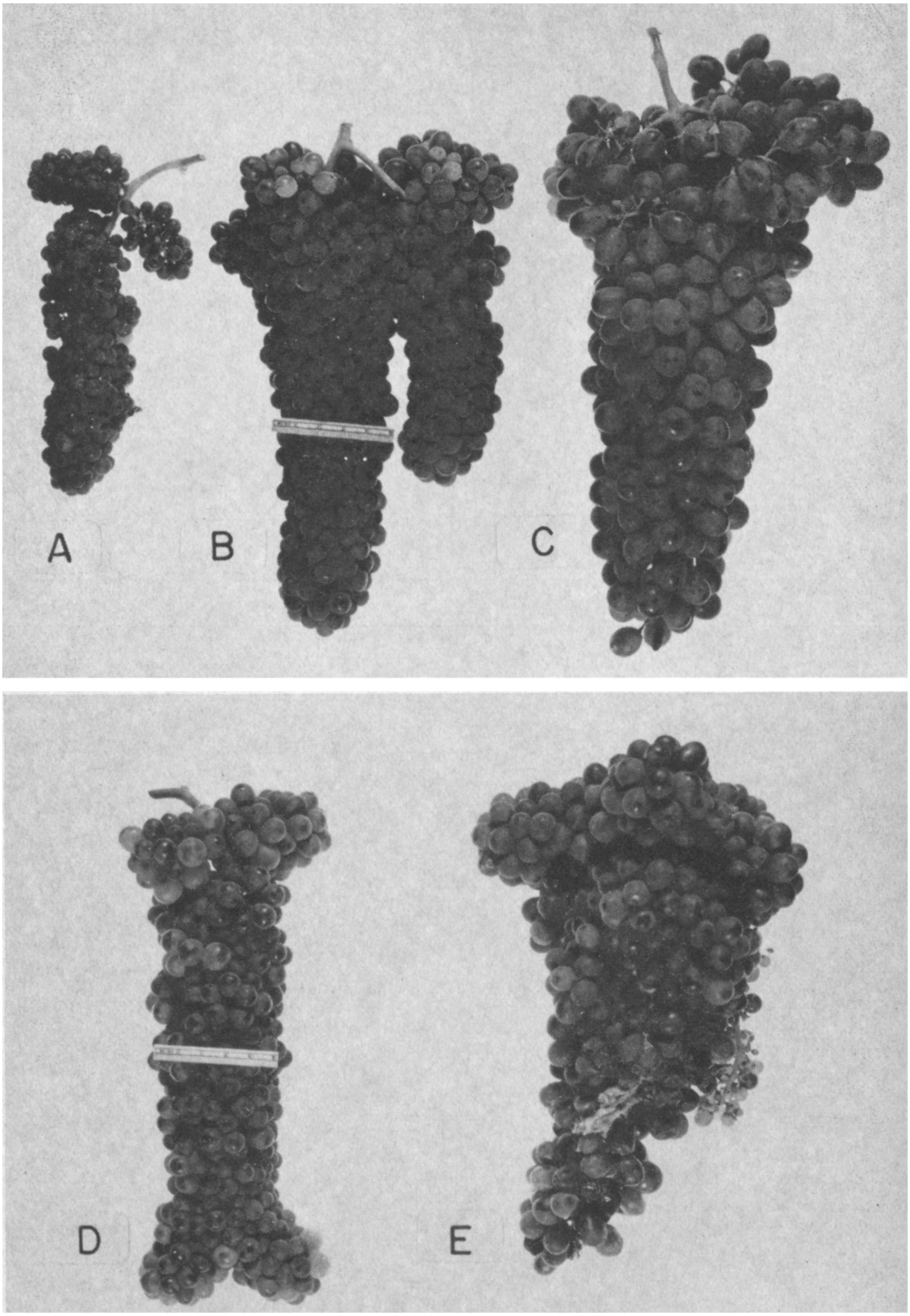


Fig. 3. Ungirdled Black Corinth clusters at harvest 80 days after being dipped in regulator solutions. (A) control; (B) 4-CPA at 60 ppm; (C) gibberellin at 100 ppm; (D) BTP at 1,000 ppm; and (E), 4-CPA at 20 ppm, gibberellin at 33 ppm, and BTP at 333 ppm. Large elongated berries resulted from gibberellin (C) and from the mixture (E); smaller round berries resulted from the 4-CPA (B) and the BTP (D). (Photographed August 13, 1964.)

sheets. The clusters were harvested August 13 (fig. 4). Control clusters had a large number of small berries. When the apical portion had been dipped, treated berries were greatly enlarged. However, the undipped berries on the basal portion were small like those of the controls. The branches were usually dead on the three or four lateral branches just basal to the dipped area (fig. 4). When the basal portion of the cluster was treated, treated berries were enlarged, but those in the untreated apical portion were also larger than the controls.

Thompson Seedless

For many years in California Thompson Seedless vines were girdled to produce larger berries for table use (Jacob, 1931). 4-CPA was used on a limited scale for the same purpose (Weaver, 1956). At present practically all Thompson Seedless for table use are commercially sprayed with gibberellin to produce large berries. Usually this practice is combined with girdling to obtain large berries. Our experiments were to determine the effect of the kinins, BTP and BA, on berry growth. The effects of applications at bloom and the berry shatter stage following bloom were studied. Also followed were the effects of using kinin with gibberellin.

Effect of BTP and gibberellin at full bloom. On May 22, 1964 at full bloom, ten clusters were dipped in BTP at 1,000 ppm, ten in gibberellin at 100 ppm, and ten in BTP at 500 ppm plus gibberellin at 50 ppm. One series of clusters served as untreated control. The clusters were not berry-thinned. Clusters were harvested on September 9 (table 3, fig. 5). The control clusters were loose with small berries. The number of berries per cluster was significantly increased by the BTP and by the mixture of BTP and gibberellin. These clusters were very compact. BTP had no effect on berry size. The gibberellin alone and the mixture of compounds produced the largest berries—elongated and with pedicels three times the diameter of the controls. Differences in maturation, as judged by degrees Balling and total acid, were not significant.

Effect of BTP and gibberellin at shatter stage. On June 9, 1964, ten clusters at the shatter stage were dipped in BTP at 1,000 ppm. Another series was dipped in gibberellin at 100 ppm, and a third series in BTP at 500 ppm plus gibberellin at 50 ppm. A fourth series was left as an untreated control. The clusters were harvested on September 9 (table 4, fig. 6). BTP at 1,000 ppm significantly increased berry size, although not nearly as much as the gibberellin alone or as the gibberellin-BTP mixture. Berries dipped in BTP had the same oval shape as the control berries, but their pedicels were about 50 per cent

TABLE 3

DATA AT HARVEST (SEPTEMBER 9, 1964) FOR THOMPSON SEEDLESS DIPPED IN BTP AND/OR GIBBERELLIN ON MAY 22, 1964

(Figures are averages of ten replicate clusters)

Treatment		Number of berries per cluster	Weight per berry	Degrees Balling	Total acid, gm tartaric per 100 ml
Concentration of BTP	Concentration of gibberellin				
ppm	*ppm*		*gm*		
0	0	292	1.19	17.4	0.71
1,000	0	838	1.15	16.6	0.79
0	100	357	2.47	17.0	0.80
500	50	805	2.18	16.8	0.78
L. S. D. at 5 per cent...........	...	270	0.59	...	....

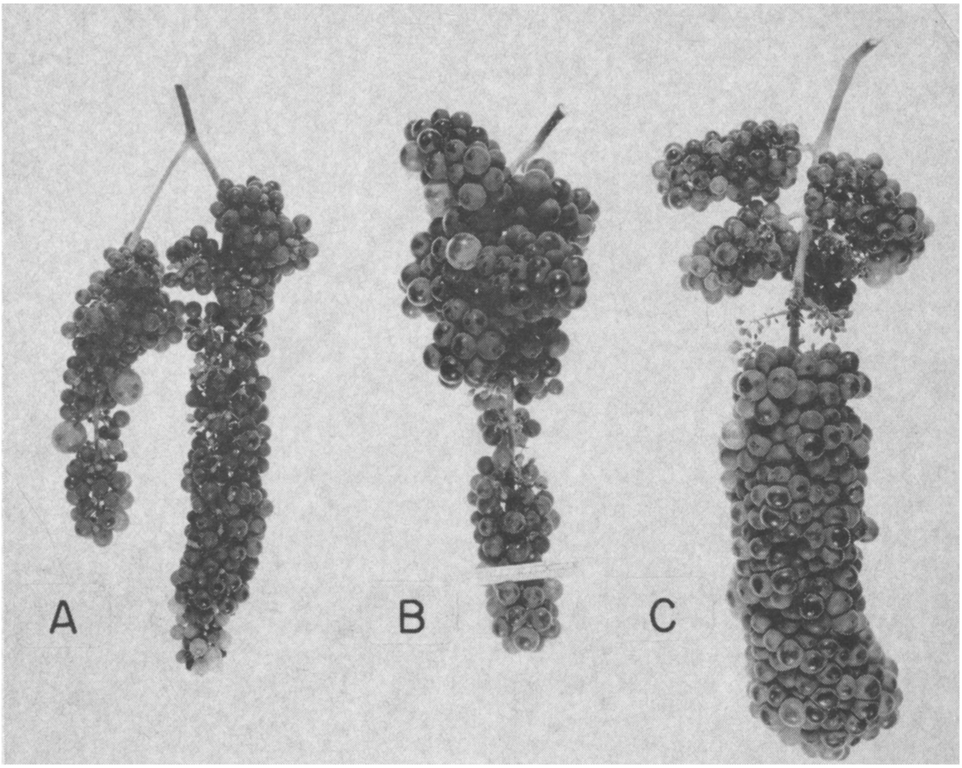


Fig. 4. Ungirdled Black Corinth clusters 80 days after treatment with BTP. (A) control; (B) basal half of cluster sprayed with compound at 1,000 ppm; and (C) apical half dipped in BTP at 1,000 ppm. In cluster in which basal half was treated (B), berries on apical half are enlarged; but when apical half was dipped (C) basal berries were not enlarged. On both treated clusters (B, C) some lateral branches between treated and untreated portion of cluster are very small. (Photographed August 13, 1964.)

TABLE 4

DATA AT HARVEST (SEPTEMBER 9, 1964) FOR CLUSTERS OF THOMPSON SEEDLESS DIPPED AT SHATTER STAGE IN BTP AND/OR GIBBERELLIN

(Figures are averages of ten replicate clusters)

Treatment		Weight per berry	Degrees Balling	Total acid, gm tartaric per 100 ml
Concentration of BTP	Concentration of gibberellin			
ppm	*ppm*	*gm*		
0	0	1.73	23.6	0.60
1,000	0	2.38	18.8	0.68
0	100	3.85	22.8	0.61
500	50	3.45	21.0	0.60
L. S. D. at 5 per cent.....	...	0.15	....	....

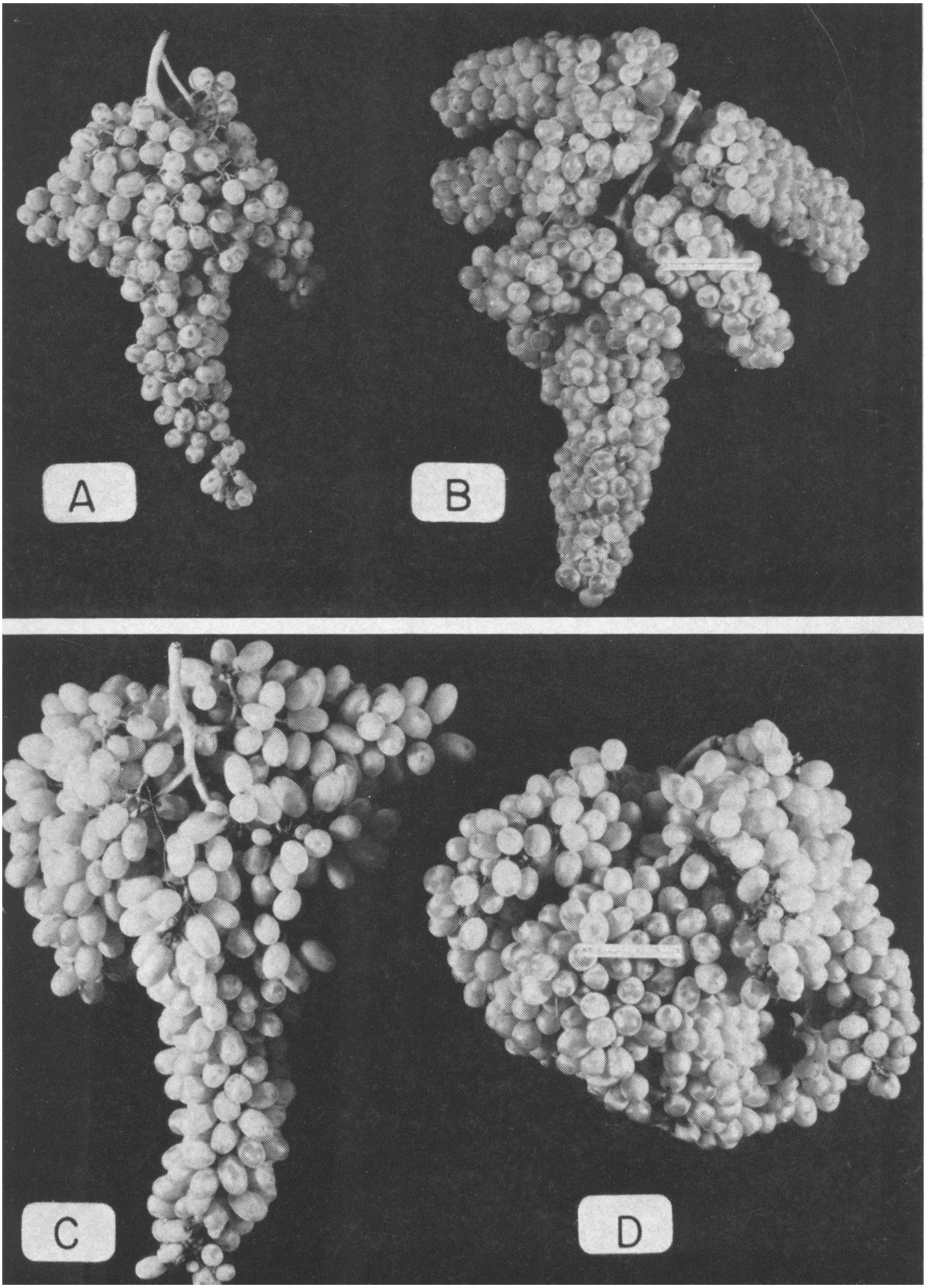


Fig. 5. Ungirdled Thompson Seedless cluster 100 days after dipping at full bloom into regulator solutions. (A) undipped control; (B) BTP at 1,000 ppm; (C) gibberellin at 100 ppm; and (D) BTP at 500 ppm plus gibberellin at 50 ppm. Clusters dipped into BTP (B) or BTP plus gibberellin (D) have a large number of berries and are very compact. The largest berries were produced by gibberellin alone (C). The shape of cluster (D) is of no special significance. (Photographed September 9, 1964.)

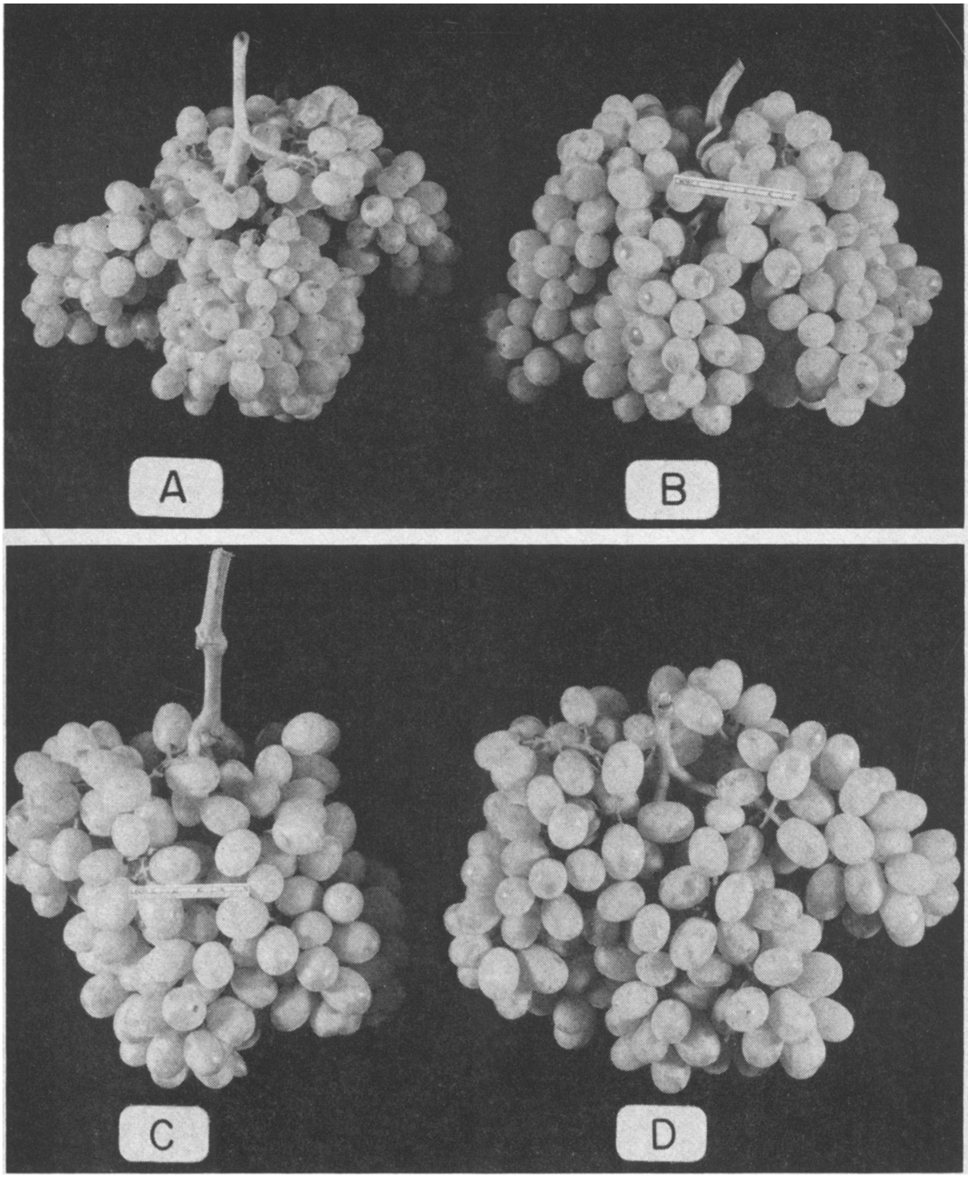


Fig. 6. Ungirdled Thompson Seedless clusters 92 days after dipping in regulators at shatter stage (June 9, 1964). (A) undipped control; (B) BTP at 1,000 ppm; (C) gibberellin at 100 ppm; and (D) BTP at 500 ppm plus gibberellin at 50 ppm. BTP resulted in some berry enlargement, but the largest berries resulted from the gibberellin (C) or BTP plus gibberellin (D). (Photographed September 9, 1964.)

thicker than the controls. Clusters dipped in gibberellin had slightly elongated berries and pedicels twice as thick as the controls. Clusters dipped in gibberellin-BTP mixture were similar to those dipped in gibberellin alone. BTP alone probably decreased the degrees Balling reading and increased the percentage of total acid.

Effect of BTP or BA at berry shatter stage. On June 14, 1963 when the berries were at the early shatter stage, clusters were dipped, ten per treatment, in BTP at 0, 31, 62, 125, 250, 500, or 1,000 ppm. A similar series was dipped in BA, and ten clusters served as the untreated controls. The clusters were harvested on September 17 (table 5). All treatments with BTP resulted in an increased number of berries because shatter was not completed at time of treatment, but only at 1,000 ppm did the compound cause an increase in berry size. BTP at 500 and 1,000 ppm increased pedicel thickness and size, and these clusters were stiff and woody. BTP at 1,000 ppm probably reduced degrees Balling and increased percentage of acid. Results with BA were similar but of a lesser magnitude.

The dipping treatments with BTP and BA were repeated by spraying the entire vines, one vine per treatment. The results again were similar to those of the dipping.

Muscat of Alexandria

Muscat of Alexandria is a poorly setting variety and as a consequence the clusters are usually too loose. If the numbers of berries could be increased in this variety, it would be of much greater commercial value.

Dipping of clusters in BTP at bloom. Ten clusters per treatment were dipped in BTP at 0, 62, 125, 250, 500 or 1,000 ppm on June 14, 1963, about three days after full bloom. At harvest on October 22, clusters dipped in BTP at 0, 62, 125, 250, 500 or 1,000 ppm, averaged respectively, 66, 63, 179, 137, 135, and 165 berries per cluster. Since the difference required for significance at the 5 per cent level among treatments was 47, berry numbers were significantly increased by concentrations of compounds ranging from 125 to 1,000 ppm. Many berries on the treated clusters were seedless and smaller than those of the untreated controls.

An experiment performed with BA gave similar results, with responses slightly lower than those in the BTP experiment.

Dipping of clusters on partially defoliated shoots in BTP. On June 14, 1963, 20 shoots were ringed, at the base and all foliage was removed except a one-square-inch stub which was retained on the leaf above the cluster to reduce set (fig. 1); then ten clusters were

TABLE 5

DATA AT HARVEST (SEPTEMBER 17) FOR THOMPSON SEEDLESS DIPPED IN BTP AT SHATTER STAGE ON JUNE 14, 1963

(Figures are averages of ten replicate clusters)

Concentration of BTP	Number of berries per cluster	Weight per berry	Degrees Balling	Total acid, gm tartaric per 100 ml
ppm		*gm*		
0	268	1.61	21.8	0.67
1,000	504	2.22	17.8	0.80
500	498	1.68	20.8	0.64
250	602	1.67	20.4	0.64
125	570	1.50	20.8	0.59
62	410	1 75	21.3	0.62
31	407	1.75	19.0	0.70
L. S. D. at 5 per cent.....	105	0.23	....	....

dipped in BTP at 500 ppm. Ten clusters served as untreated controls. Eight days later when set was complete, berry counts showed an average of 809 berries per cluster on treated clusters and 27 berries on untreated controls (fig. 7). The difference required for significance at the 5 per cent level was 341. BTP produced a striking increase in set.

RESPONSE OF EMASCULATED FLOWERS TO BTP

The varieties Black Corinth, Thompson Seedless, and Tokay were used. It has been shown previously that both auxin and gibberellin can induce set in emasculated clusters of these varieties (Weaver and McCune, 1960). The purpose of our experiments was to determine whether a kinin, BTP, is also effective in inducing set under these conditions. The purpose of emasculation was to remove all pollen as a source of plant hormone and to prevent fertilization. After the clusters had dried following dipping they were bagged to prevent cross-pollination until the stigmas were no longer receptive to fertilization.

Black Corinth

Black Corinth is parthenocarpic, but is is generally believed that the stimulus of pollination on ungirdled canes is required to induce set (Stout, 1936). This process is referred to as "stimulative parthenocarpy." Six clusters were emasculated and three were dipped in BTP at 500 ppm on May 20, 1964. Three clusters served as controls. All bags were removed on June 1 and the numbers of flowers and berries that had fallen from the clusters into the bags were counted. The data (table 6) reveal a far greater shatter from the control clusters. Clusters were harvested on August 11. All control clusters were dead and dried (fig. 8, top). There was a large number of berries on treated clusters although berry size was quite variable. More than one-third of the berries had hard seed coats. The treated berries were larger than those on unemasculated fruit on an adjacent vine, although their degrees Balling was lower.

Thompson Seedless

In Thompson Seedless pollination and fertilization occur but the embryo soon

TABLE 6

DATA AT TIMES OF FLOWER SHATTER AND HARVEST FOR GRAPES EMASCULATED AND DIPPED AT EARLY BLOOM IN BTP AT 500 PPM

(Figures are averages oı three replicate clusters)

Variety	Concentration of BTP	Number of flowers and berries fallen at shatter per cluster	Number of berries on cluster at harvest	Weight per berry	Berries with hard seeds	Degrees Balling	Total acid, gm tartaric per 100 ml
	ppm			*gm*	*per cent*		
Black Corinth	0	176	0	....	....	....	....
	500	15	245	0.30	37	17.9	1.53
L. S. D. at 5 per cent		3					
Thompson Seedless	0	392	0	....	....	....	....
	500	56	30	0.45	....	24.4	....
L. S. D. at 5 per cent		3					
Tokay	0	204	2	....	....	....	....
	500	46	50	....	....	....	....
L. S. D. at 5 per cent		3					

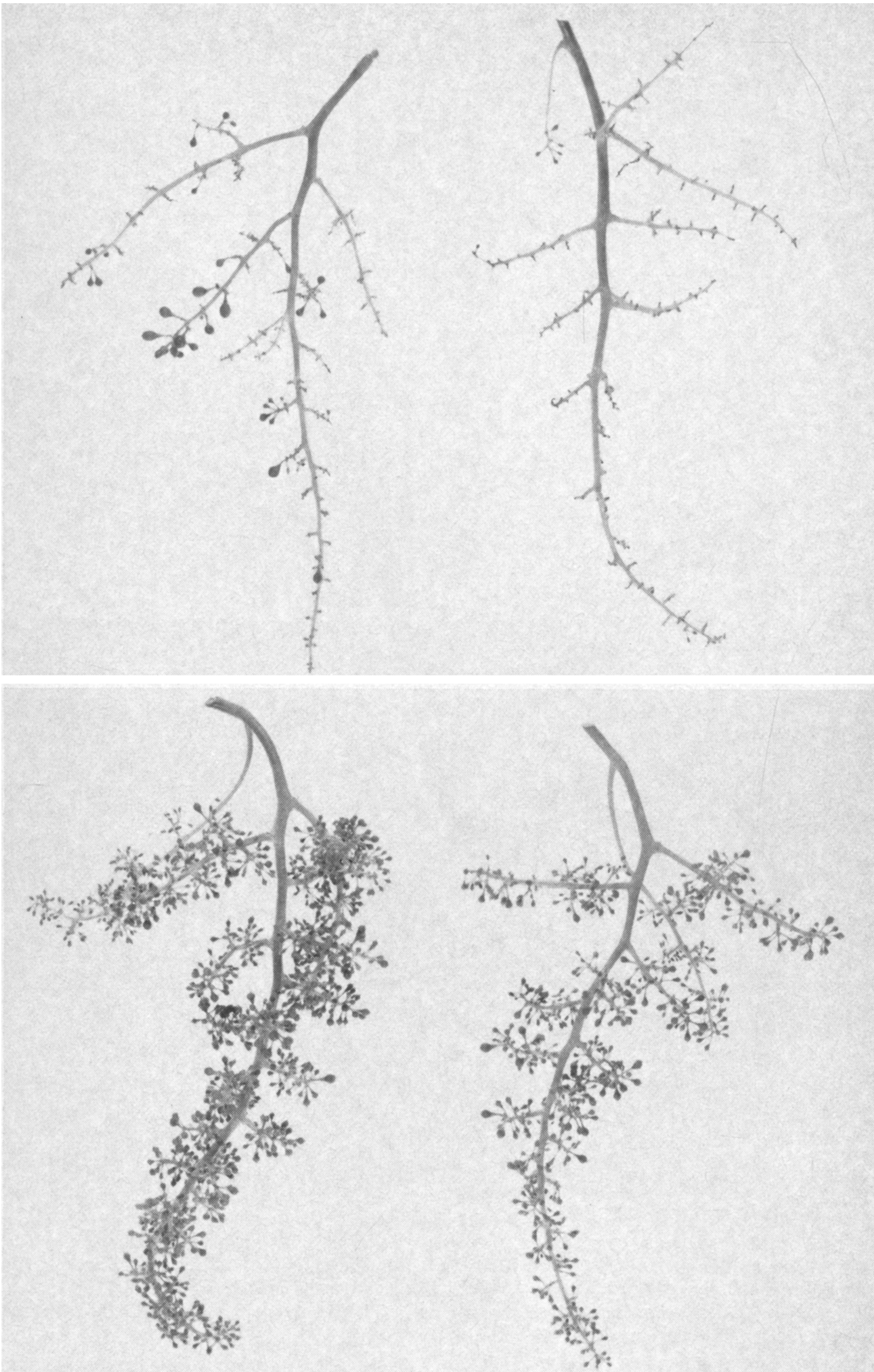


Fig. 7. Clusters of Muscat of Alexandria grapes on partially defoliated shoots eight days after dipping in BTP at late bloom stage (June 14, 1963). *Top*, control; *bottom*, dipped in BTP at 500 ppm. Dipping resulted in a large number of berries. (Photographed June 14, 1963.)

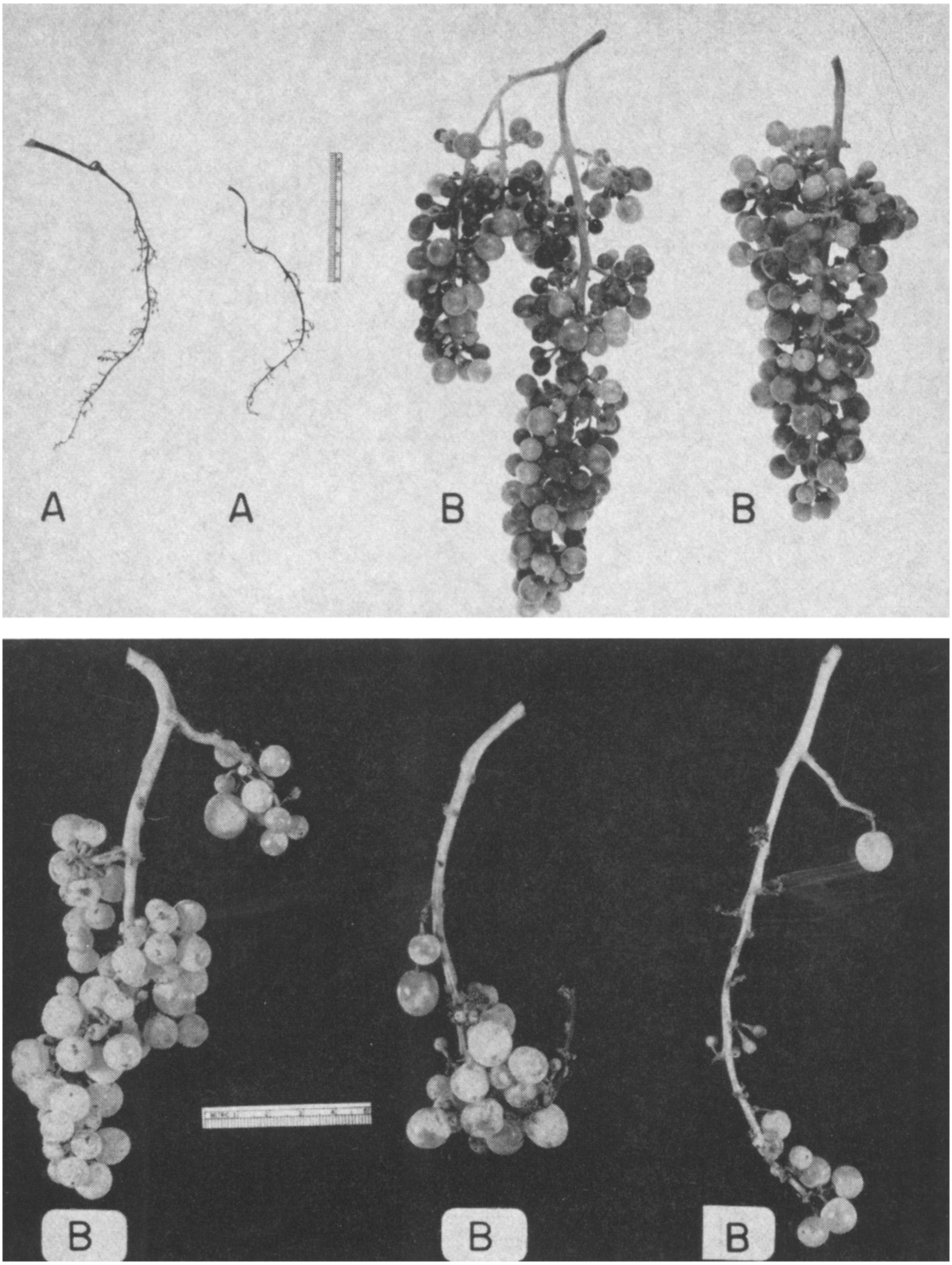
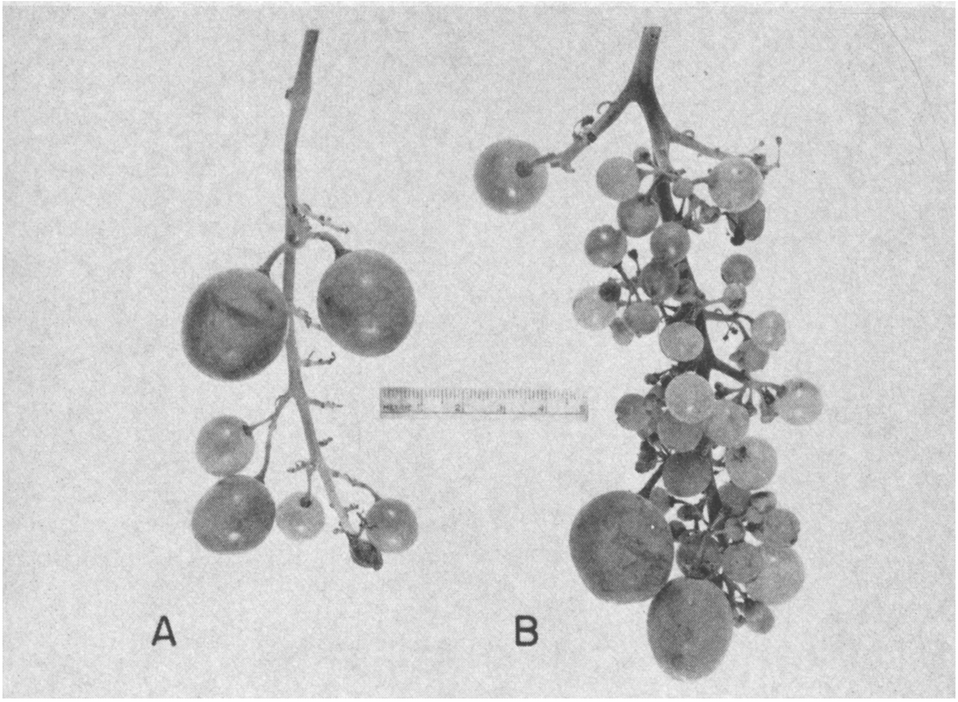


Fig. 8. Clusters of Black Corinth (*top*), Thompson Seedless (*bottom*) and Tokay (*right*) at harvests after emasculation and dipping at early bloom in BTP at 500 ppm. Black Corinth: (A) emasculated, untreated; (B) emasculated and dipped in BTP. All Thompson Seedless clusters shown were dipped in kinin; controls which had produced no berries were dried and lost. Two untreated Tokay clusters, not shown, produced no berries. Treatment with BTP (B) in all cases increased number of berries. (Photographed in fall of 1964).



aborts. Stout (1936) called this condition "stenospermy." Clusters were emasculated on May 26, 1964. Three clusters were dipped in BTP at 500 ppm, and three others served as untreated controls. On June 9 the bags were removed. More than five times more flowers and berries had fallen from the control clusters than from the treated (table 6).

Clusters were harvested on September 9. All control clusters were dead and dried, and no berries had set. The clusters dipped in kinin averaged 30 small berries per cluster (fig. 8, middle).

Tokay

Seed development in Tokay is normally complete in a high percentage of ovules. Clusters were treated on May 26. Three were dipped in BTP solution at 500 ppm and three served as untreated control. On June 9, when the bags were removed, more than four times as many flowers and berries had fallen from the control clusters as from the dipped cluster (table 6). At harvest two control clusters had set no berries, and the third six berries (averaging two berries), but about 50 berries had set on the treated clusters (fig. 8, right). The set on control clusters may have been a result of pollen contamination or failure to emasculate all of the flowers sufficiently early.

FOUR VARIETIES DIPPED IN BTP

The purpose of this test was to study the effect of kinin on varieties that have defective female parts (Black Corinth, Thompson Seedless), on a normal variety (Tokay), and on a variety with defective male parts (Almeria). Ten clusters each of Black Corinth, Thompson Seedless, Tokay, and Almeria (Ohanez) were dipped in BTP at 1,000 ppm at bloom stage. When dry, the clusters were bagged to prevent cross-pollination. At the time the bags were removed, about ten days later, the ovaries that had fallen were counted.

Black Corinth

Black Corinth was dipped on May 20, 1964, when about 5 per cent of the calyptras had fallen. On June 10 the bags were removed. More than ten times as many ovaries had fallen from the control clusters as from treated clusters (table 7). At harvest, on August 13, significantly more berries were on the treated than on the control clusters (table 7). The average weight of the BTP-treated berries was almost twice that of the control. About 5 per cent of the total number of berries on the treated clusters were very large and contained hard seed coats. The degrees Balling was lower on the treated cluster, probably because of the larger crop.

Thompson Seedless

Thompson Seedless clusters were treated and bagged on May 25 at an early bloom stage. On June 11 when the bags were removed, eight times more ovaries had fallen from the control clusters than from the treated clusters (table 7). At harvest on September 9 the number of berries on the treated clusters was larger, but the weight per berry was less (table 7, fig. 9).

Tokay

Tokay was treated at full bloom on May 26. On June 11 when the bags were removed, the treated clusters appeared to have a larger number of berries. Clusters were harvested on September 22, when about 10 per cent of the total surface of both untreated and treated clusters were red. The number of berries on the treated clusters was not significantly greater than on the untreated (table 7). Berry size was smaller on the treated clusters (table 7, fig. 10).

TABLE 7

DATA AT TIMES OF SHATTER AND HARVEST FOR FOUR VARIETIES DIPPED AT BLOOM TIME WITH BTP AT 1,000 PPM

(Figures are averages of ten replicate clusters)

Variety	Concentration of BTP	Number of berries and flowers fallen at shatter per cluster	Number of berries per cluster at harvest	Weight per berry	Degrees Balling	Total acid gm tartaric per 100 ml,
	ppm			*gm*		
Black Corinth	0	65	465	0.31	19.2	1.27
	1 000	6	708	0.56	17.4	1.29
L. S. D. at 5 per cent		2	71	0.20	....	....
Thompson Seedless	0	321	441	1.52	18.4	0.69
	1 000	40	720	1.20	16.0	0.78
L. S. D. at 5 per cent		2	172	0.20	....	....
Tokay	0	...	217	5.93	19.0	
	1 000	20	299	3.68	18.6	
L. S. D. at 5 per cent		...	138	1.76	....	
Almeria Seeded berries	0	547*	41	2.58	20.8	0.65
	1 000	147*	51	3.79	21.0	0.64
L. S. D. at 5 per cent		249*	N.S.	N.S.	....	....
Seedless berries	0	547*	2	1.18	....	....
	1 000	147*	280	0.44	23.6	0.57
L. S. D. at 5 per cent		249*	144	N.S.	....	....

* Includes flowers and berries from both seeded and seedless berries (see text).

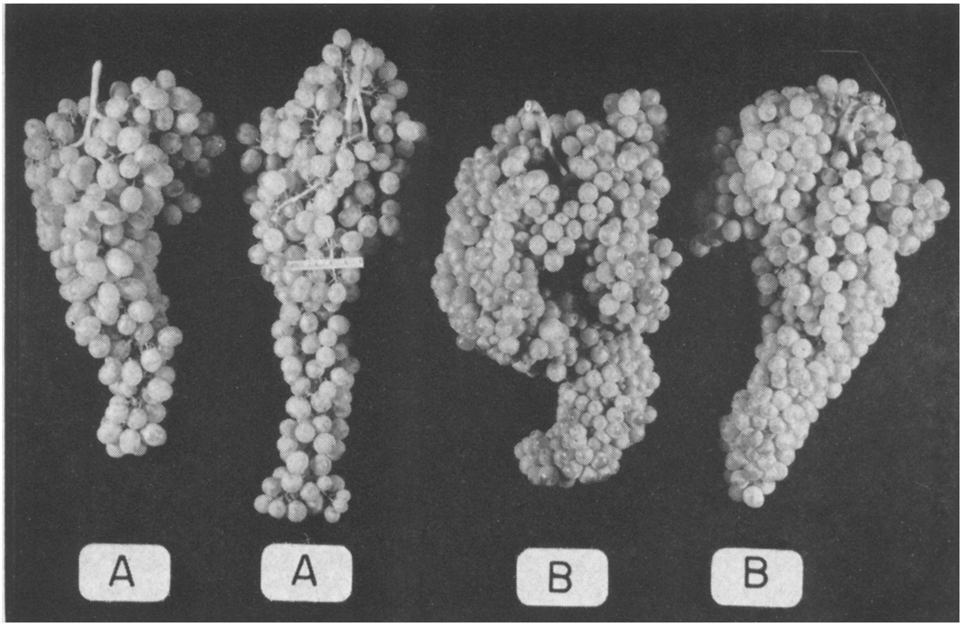


Fig. 9. Ungirdled clusters of Thompson Seedless 106 days after dipping in BTP at 1,000 ppm. (A) control; (B) dipped in kinin. Clusters dipped in BTP (B) have more but smaller berries than the control (A). (Photographed September 9, 1964.)

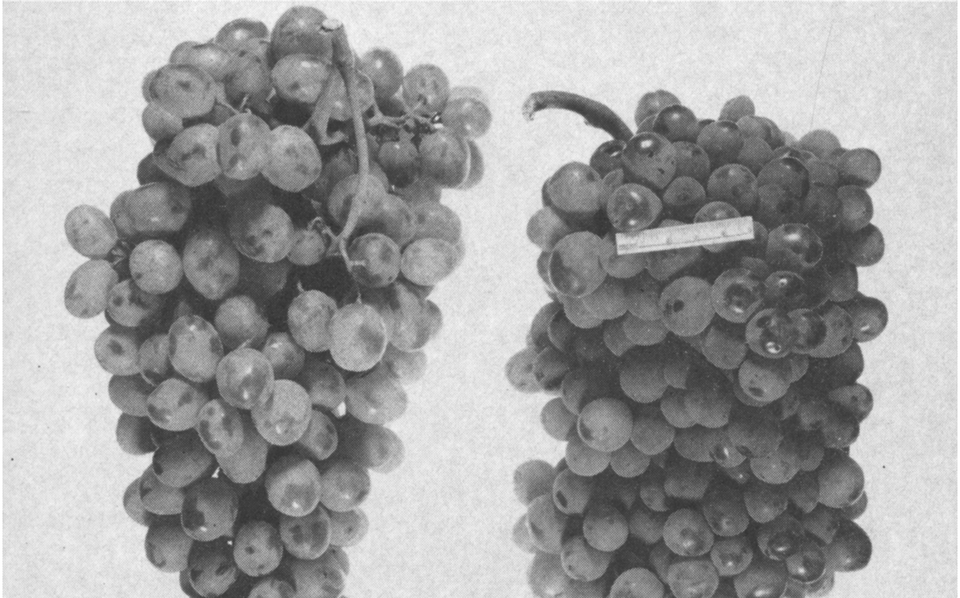


Fig. 10. Tokay cluster 119 days after dipping in BTP at 1,000 ppm. *Left,* control; *right,* dipped in kinin. Berries on cluster treated with BTP are smaller and cluster is more compact. (Photographed September 22, 1964.)

Almeria

Almeria clusters were treated on May 25, when about half of the calyptras had fallen. When the bags were removed 17 days later, 547 flowers per cluster had fallen from the untreated and 147 from the treated (table 7). At this point it was not yet possible to distinguish what would develop into seeded and what into seedless berries. Clusters were harvested on September 30. Since the kinin had resulted in a large increase in the number of seedless berries, data for berries with and without seeds were separately determined. Two control clusters had a large number of seeded berries, but the other eight had a very small number. Treated clusters had about the same number of seeded berries as the control, but there were far more seedless berries in the treated clusters (table 7, fig. 11). These seedless berries were very small—about the size of girdled Black Corinth berries.

DISCUSSION

In our early work with the effect of kinetin, the first kinin identified, on grapes we obtained no positive responses on either fruit set or fruit enlargement. With BA the results were either negligible or rather small. However, a new, more mobile kinin, BTP, has been very effective. Its greater solubility no doubt partly explains its effectiveness. At 25°C BA has a water solubility of 80 ppm while that of BTP is 200 ppm. In our experiments with BTP and BA the more striking responses were always obtained with BTP. BA often gave a response but to a lesser degree.

It is now apparent that the role of the kinins, in addition to those of the auxins and gibberellins, must be assessed in any study of fruit set and development. Hopefully, such studies may lead to an explanation of the factors involved.

In Black Corinth where portions of clusters were treated with BTP, the branches were usually dead or stunted on the three or four lateral branches adjacent to the treated area of the cluster. This stunting may occur because the treated portion acts so strongly as a sink that the adjacent branches do not receive enough nutrients or elaborated food materials for normal development. It is well known that kinins are a strong mobilizing agent for several compounds (Mothes *et al.*, 1959; Gunning and Barkley, 1963; Leopold and Kawase, 1964).

Experiments with Muscat of Alexandria and other varieties usually showed that BTP is effective in set of fruit. In Muscat of Alexandria many seedless berries set, but they remained small. Auxin also sets small seedless berries, but gibberellins often cause some reduction in set. This might indicate that kinin and auxin affect more the processes of fruit set and gibberellin more fruit growth and development. All three classes of growth regulators can induce set in emasculated clusters. However, gibberellin usually induces set of a relatively small number of larger, elongated berries while the kinin and auxin produce many small, round berries.

In one experiment about 5 per cent of the total number of berries on clusters of Black Corinth dipped in BTP had hard seeds. It has also been demonstrated that hard seeds occur in Black Corinth that have been treated at bloom with the auxin 4-CPA (Weaver, 1952). We have never observed a Black Corinth grape treated with gibberellin that has had a hard seed. This may be further evidence that gibberellin mainly affects the development of the berry and not the set, at least in the Black Corinth variety.

When the basal portion of a Black Corinth cluster was treated with BTP, some of the compound moved to the apical portion as was indicated by berry size. However, the movement might have

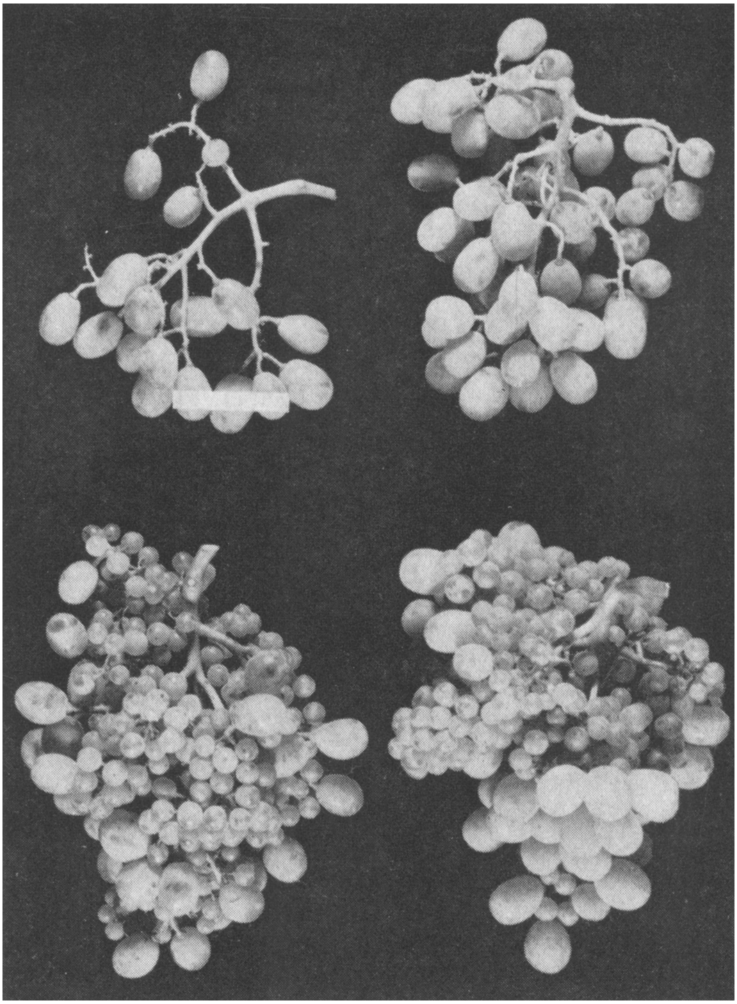


Fig. 11. Cluster of Almeria (Ohanez) 128 days after dipping in BTP at 1,000 ppm. *Top,* untreated; *bottom,* dipped in kinin. Control clusters are very loose (*top*) and many small seedless berries occur on the treated clusters (*bottom*). (Photographed September 30, 1964.)

been caused by a creeping of the liquid along the stem surfaces.

In Black Corinth and Thompson Seedless the effects of gibberellin on berry enlargement, and of BTP on increase of set, persisted even when the compounds were applied as a mixture. In Almeria BTP produced a good set of small berries. Perhaps if gibberellin were applied at the proper time, berry size might be increased two or three fold.

Kinins have already been demonstrated in the pruning wound sap of the grape vine (Loeffler and van Overbeek, 1964). A study of the naturally occurring kinins in grape flowers and berries would be very valuable in assessing the role of kinins in fruit set and development.

SUMMARY

The responses of Black Corinth, Thompson Seedless, Muscat of Alexandria, Tokay, and Almeria (Ohanez) varieties of *Vitis vinifera* to the kinins BTP (6-(benzylamino)-9-(2-tetrahydropyranyl)-9H-purine) and BA (benzyladenine) were studied in 1963 and 1964 at Davis, California.

The BTP and BA were both effective in increasing fruit numbers, and in some instances fruit growth. However, the more soluble and mobile BTP was always far more effective than the BA. Dipping experiments and spraying of entire vines were both effective.

BTP applied to Black Corinth clusters at bloom resulted in berries that were almost three to four times larger than the controls. The pedicels were five times thicker than those of the controls.

Black Corinth was treated at bloom with BTP, gibberellin, and/or the auxin 4-CPA. The largest berries were produced by gibberellin alone and by the mixture of all three compounds. These berries were elongated as a result of the gibberellin. 4-CPA and BTP produced about equally sized, almost round berries.

When the apical half of the Black Corinth clusters were treated at bloom with BTP at 1,000 ppm, only the treated portion had greatly enlarged berries. Branches of the untreated portion near the treated portion were usually dead or weak. When the basal portion was treated with BTP, the kinin appeared to move toward the apical portion of the cluster as judged by the enlargement of berries. Also, weakened branches occurred near the treated area.

BTP and gibberellin or a mixture of both were applied to Thompson Seedless clusters at full bloom. The percentage set was significantly increased by the BTP and the mixture. Berry size was increased only by the gibberellin or the mixture of the two compounds.

BTP and gibberellin or a mixture of the two were applied at shatter stage. BTP at 1,000 ppm significantly increased berry size, although not nearly so much as the gibberellin alone, or the gibberellin-BTP mixture. Both compounds used alone caused thickening of pedicels, but the gibberellin caused greater thickening. BTP produced normally shaped oval berries, but gibberellin resulted in elongated berries.

BTP or BA was applied at the shatter stage to Thompson Seedless. Only the compounds at 1,000 ppm caused an increase in berry size, although all treatments resulted in increased numbers of berries.

Dipping of clusters of Muscat of Alexandria at bloom in BTP at various concentrations greatly increased berry numbers in range from 125 to 1,000 ppm. Dipping of clusters on partially defoliated shoots in BTP also resulted in a great increase of berry numbers.

BTP resulted in a set of berries on emasculated flowers with Black Corinth, Thompson Seedless, and Tokay.

Clusters of Black Corinth, Thompson Seedless, Tokay, and Almeria (Ohanez) were dipped in BTP at 1,000 ppm at bloom stage. At shatter far more ovaries or berries fell from the treated than on the control clusters, except for Tokay. The Almeria tremendously increased the number of seedless berries on the treated clusters.

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