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Effect of gibberellin on seeded Vitis vinifera with special reference to induction of seedlessness*)

by

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Introduction

Early investigations indicated that seeded varieties of *Vitis vinifera*, unlike seedless varieties, not only do not increase berry size in response to gibberellin application but exhibit rather toxic effects and reduced crop weights (WEAVER and McCUNE 1959 a, b, BLAHA 1963 and DAVIDIS 1963). Recently, however, increased crop weights and seedlessness have also been reported in seeded *Vitis vinifera* (OOHATA and YOSHIDA 1960, AKABANE and YAMAZAKI 1961, KISHI and TASAKI 1960, KAJIURA 1962, ANON. 1962, RAO *et al.* 1962, HIDALGO and CANDELA 1963, KOLOLGI *et al.* 1963, VENKATARATNAM 1964 and CLORE 1965).

In view of these varied results, detailed studies were undertaken during the years 1961—64 at the Horticulture Division, Indian Agricultural Research Institute, New Delhi, India in the hope of clarifying the problem. The main object was to see the effects of gibberellin applied at various stages of cluster development on different seeded cultivars of *Vitis vinifera* varying in berry size, seed number per berry and vine vigour. Apart from obtaining basic understanding of this problem, the idea was to improve berry set in certain varieties and to explore the possibilities of inducing seedlessness.

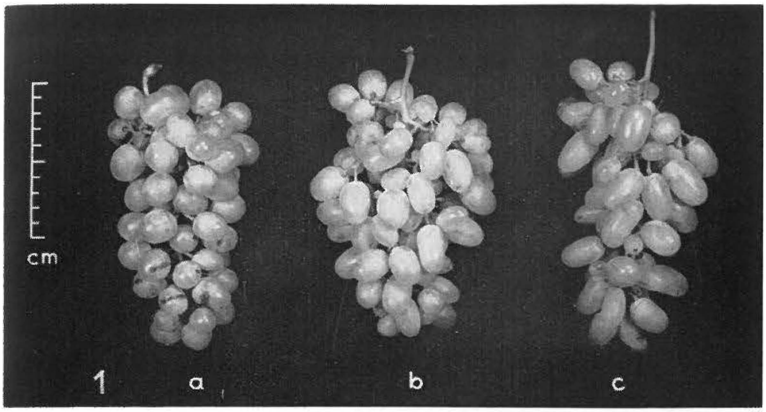
Material and Methods

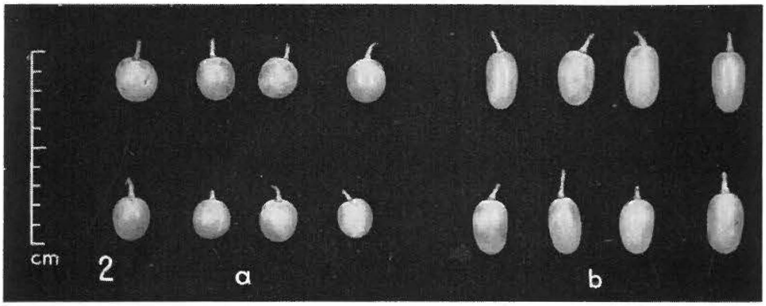
Randomized block and split plot designs were followed in the experiments reported here. Ten year old vines of Bhokri, Gros Colman (Pusa), Anab-e-Shahi, Alamwick, Bharat Early and Black Hamburg varieties trained on kniffin system were used in this investigation. Gibberellic acid (GA) was sprayed with a hand sprayer to clusters only of these varieties protecting other clusters with an alkathene sheet. Triton was used as a wetting agent. Data reported here were recorded at harvest time.

Tests of pollen germination were performed at the time of flowering in 20 per cent sucrose medium at a controlled temperature of 26—28° C. A drop of medium was placed over a clean cover glass and freshly dehisced pollen grains were dusted on the medium and then stirred for even distribution. A cavity slide smeared with grease around the cavity was then inverted over the cover glass. Cavity slides were then again carefully inverted along with the cover slip and placed in a petri dish lined with moist filter paper. Data on pollen germination were recorded after 24 hours.

Weight per berry was calculated from the weight of fifty or twenty five randomly taken berries. In berry number per cluster, shot berries were not included in the count. Seed number per berry was recorded after cutting open at least twenty

*) This study formed part of the Ph. D. thesis submitted by the senior author to the Post Graduate School, IARI, New Delhi, India.





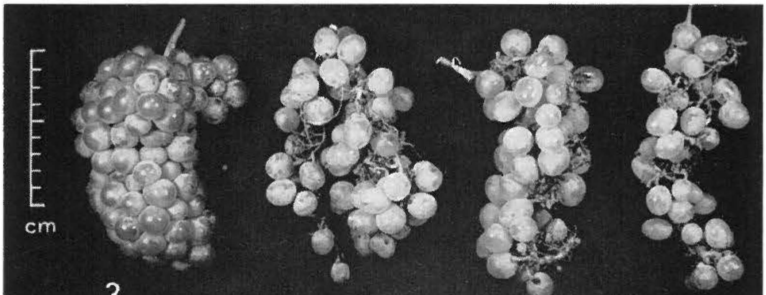


Fig. 1: Effect of GA on Anab-e-Shahi grape clusters sprayed at 2—3 days after full bloom at 0 ppm (a), 25 ppm (b) and 50 ppm (c).
Fig. 2: Effect of GA on the berry size of Anab-e-Shahi grape sprayed at 2—3 days after full bloom at 0 ppm (a) and 50 ppm (b).
Fig. 3: Effect of GA on Bhokri grape sprayed twice at 10—11 days before full bloom and 7—8 days after full bloom — (a) control, (b) 50 ppm, (c) 100 ppm and (d) 150 ppm.

five berries taken at random. From these counts, per cent seedless berries and percentage of berries in each seed class were calculated. Total soluble solids (T. S. S.) were measured by Zeiss hand refractometer and total acidity was determined by titrating 5 ml of juice with N/10 NaOH using phenolphthalein as an indicator in samples taken at random. The results were expressed in grams of tartaric acid per 100 ml of juice.

Results

(I) Effect of GA on Anab-e-Shahi sprayed at prebloom and postbloom stages

GA at 25 and 50 ppm was sprayed on clusters of Anab-e-Shahi at 7—8 days before full bloom and 2—3 days after full bloom. The results recorded at harvest time are summarized in Table 1.

In general no toxic effects were observed in this variety at these concentrations of GA, but the clusters were loosened. Table 1 shows that the bunch length was increased both at prebloom and postbloom sprays.

Table 1

Data at harvest (July 1, 1963) showing the effect of GA on Anab-e-Shahi grape sprayed at prebloom and postbloom stages

GA Concentrations (ppm)	Bunch length (cm)	Bunch weight (gg)	Berry weight (gg)	Berry number per cluster	Seedless berries (%)	T. S. S. (%)	Total acidity (%)
0 (control)	17.50	245.0	4.31	49.66	0	16.0	0.712
25 prebloom	20.66	220.3	4.66	45.66	26.3	15.5	0.675
50 prebloom	23.00	158.0	4.55	32.66	29.6	16.0	0.675
25 postbloom	17.83	269.0	5.04	48.33	8.4	16.5	0.712
50 postbloom	21.33	267.0	5.35	50.33	9.7	15.0	0.675
L.S.D. 5%	1.90	68.14	0.59	8.45	—	—	—
Significance	**	*	**	*			

* significant at 5% level; ** significant at 1% level.
Prebloom: 7—8 days before full bloom; Postbloom: 2—3 days after full bloom

Further perusal of Table 1 shows that the bunch weight and berry number per cluster were significantly reduced at 50 ppm GA applied at prebloom stage. However, even at prebloom application of GA, there was some increase in berry weight and appreciable number of seedless berries were induced (Fig. 1). With postbloom application of GA, the bunch and berry weights were increased but there were fewer seedless berries compared to prebloom spray (Fig. 1, 2). Berries were not thinned by application of GA at postbloom stage. Berries in clusters treated at both stages of development were quite elongated without there being very much effect on width. There was no marked effect on quality of the berries.

Table 2

Effect of GA on pollen germination of Bhokri clusters sprayed at prebloom stages

GA concentrations (ppm)	Percentage pollen germination 0	50	100	150
10—11 days before full bloom	86.4	2.46	0	0
4—5 days before full bloom	81.2	3.59	0.9	0

Table 3

Effect of GA on Bhokri sprayed at different stages of cluster development

GA Concentrations (ppm)	Bunch weight (gg)	Berry number per cluster	Berry weight (gg)	Seedless berries (%)	T. S. S. (%)	Total acidity (%)
Sprayed at 10—11 days before full bloom (S_1)						
0	206.2	97.6	2.07	0	13.5	0.66
50	121.8	85.5	1.38	90.1	15.0	0.78
100	111.0	76.0	1.41	97.5	16.5	0.82
150	81.8	65.7	1.24	98.1	18.0	0.91
Sprayed at 10—11 days before full bloom + 7—8 days after full bloom (S_2)						
0	221.2	104.7	2.11	0	13.0	0.67
50	165.5	75.7	2.09	87.0	14.5	0.79
100	145.2	63.0	2.31	96.6	17.0	0.84
150	129.0	56.8	2.17	97.3	18.0	0.88
Sprayed at 4—5 days before full bloom (S_3)						
0	230.0	108.0	2.04	0	14.0	0.69
50	215.0	117.2	1.82	37.8	15.0	0.72
100	177.2	100.5	1.77	41.4	15.0	0.76
150	168.0	109.7	1.74	40.3	16.0	0.78
Sprayed at 4—5 days before full bloom + 7—8 days after full bloom (S_4)						
0	233.2	110.7	2.10	0	14.0	0.69
50	203.5	92.3	2.15	37.3	14.0	0.70
100	205.8	95.0	2.12	38.3	14.5	0.73
150	211.0	101.0	2.04	45.4	15.0	0.73
L.S.D. 1[1])	30.92	10.96	0.261	—	—	—
L.S.D. 2[2])	30.01	11.29	0.268	—	—	—

[1]) L.S.D. 1 at 5% for GA concentrations at a given cluster stage.
[2]) L.S.D. 2 at 5% for cluster stages at a given GA concentration.

(II) Effect of GA on Bhokri clusters applied at different stages of cluster development

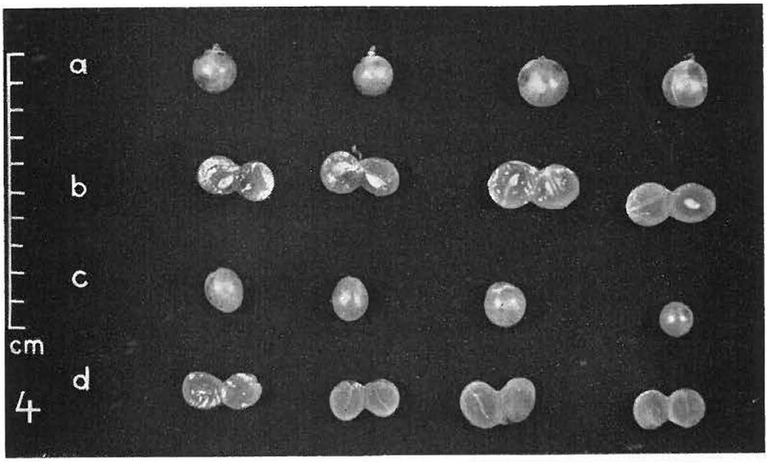
GA (50, 100 and 150 ppm) application was made at 10—11 days before full bloom alone (S_1), 10—11 days before full bloom combined with a spray at 7—8 days after full bloom (S_2), 4—5 days before full bloom alone (S_3) and 4—5 days before full bloom combined with a spray at 7—8 days after full bloom (S_4). The main object was to see the effect of high concentrations of GA alone and double sprays on the toxicity in general and seedlessness. The results are presented in Tables 2 and 3.

Table 2 clearly shows that GA application at 10—11 days before full bloom and 4—5 days before full bloom alone, adversely affected the pollen viability.

In general, GA treatments at any of the cluster stages did not produce shot berries. Bunches were extremely loosened and curved with S_1 and S_2 treatments. Cluster frame work with S_2 treatment was very tough and callused due to double sprays (Fig. 3).

It is evident from Table 3 that GA sprayed at 10—11 days before full bloom (S_1), induced over 90% seedless berries at all three levels of application. At the same time, however, bunch weight, berry number and berry weight were significantly reduced. With a second spray at 7—8 days after full bloom (S_2), berry weight and size were brought a little above control but the cluster weights were still markedly reduced (Fig. 3 and 4).

When GA application was made at 4—5 days before full bloom only (S_3), the seedless berries were induced appreciably but not to the same extent as with S_1 and S_2 treatments (Table 3). Bunch and berry weights were decreased but berry number per cluster remained constant showing that the decrease in bunch and berry weights



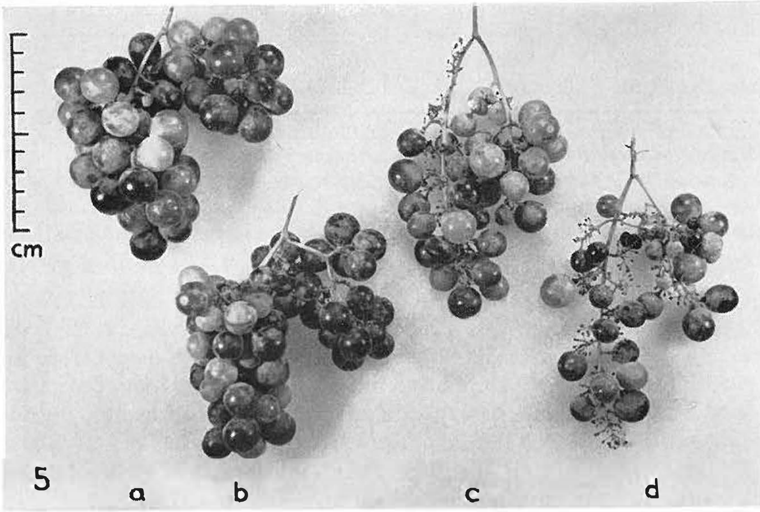
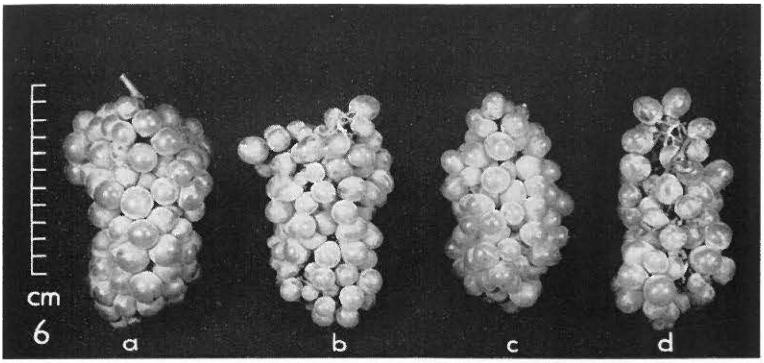


Fig. 4: Effect of GA sprays on berry seedlessness and size of Bhokri grape — a and b control, c and d berries from cluster treated with GA (100 ppm) twice at 10—11 days before full bloom and 7—8 days after full bloom.

Fig. 5: Clusters of Black Hamburg grape sprayed at full bloom stage with GA at 0 ppm (a), 5 ppm (b), 10 ppm (c) and 25 ppm (d).



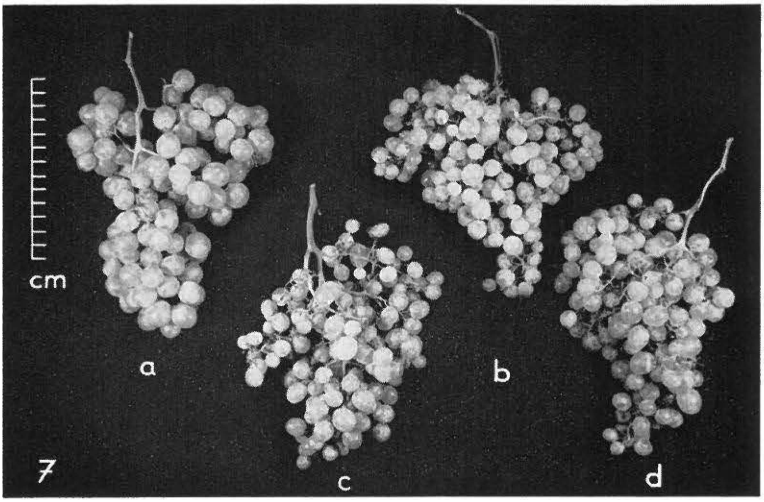


Fig. 6: Clusters of Bhokri grape after spraying at 4—5 days before full bloom alone with GA at 0 ppm (a), 50 ppm (b), 100 ppm (c) and 150 ppm (d).

Fig. 7: Clusters of Gros Colman (Pusa) grape sprayed at 5—6 days before full bloom with GA at 0 ppm (a), 50 ppm (b), 75 ppm (c) and 100 ppm (d).

was caused by seedlessness induced by GA. Notably no toxic effects were observed in this variety at such high concentrations of GA applied at 4—5 days before full bloom (Fig. 6). With the second spray of GA at 7—8 days after full bloom (S_4) the bunch and berry weights were increased to the level of control.

Both T. S. S. and total acidity were increased to a greater degree with S_1 and S_2 treatments at 100 and 150 ppm GA compared to last two S_3 and S_4 treatments (Table 3).

(III) Effect of GA on Gros Colman (Pusa) clusters applied at different stages of cluster development

In this variety, GA (50, 75 and 100 ppm) was sprayed at 5—6 days before full bloom (S'_1) and 2—3 days after full bloom (S'_2). Data collected at harvest time are given in Table 4.

It is clear from Table 4 that by spraying at 5—6 days before full bloom, bunch and berry weights were significantly reduced. However, there was some increase in fruit

Table 4

Effect of GA on Gros Colman (Pusa) grape sprayed at different stages of cluster development

GA Concentrations (ppm)	Bunch weight (gg)	Berry number per cluster	Berry weight (gg)	Seedless berries (%)	T. S. S. (%)	Total acidity (%)
		Sprayed at 5—6 days before full bloom (S'_1)				
0	365.1	152.5	2.35	0	19.0	0.45
50	192.0	144.5	1.25	77.7	19.5	0.40
75	210.0	160.8	1.30	80.0	18.6	0.37
100	201.8	168.5	1.20	74.1	18.2	0.37
		Sprayed at 2—3 days after full bloom (S'_2)				
0	353.3	141.6	2.43	0	19.5	0.46
50	384.6	204.3	1.86	63.8	19.0	0.51
75	348.6	195.1	1.75	71.9	18.0	0.61
100	320.9	189.1	1.66	67.3	18.0	0.57
L.S.D. 1[1])	18.81	10.00	0.187	—	—	—
L.S.D. 2[2])	18.55	9.84	0.165	—	—	—

[1]) L.S.D. 1 at 5% for GA concentrations at a given cluster stage.
[2]) L.S.D. 2 at 5% for cluster stages at a given GA concentration.

set and seedless berries were formed to quite a large extent (Fig. 7). Evidently the decrease in cluster and berry weights was due to formation of seedless berries.

Spraying at 2—3 days after full bloom (S'_2) the bunch and berry weights were still decreased but berry set was increased with an appreciable number of seedless berries (Table 4).

T. S. S. was decreased with both S'_1 and S'_2 treatments but the acidity was decreased with the first treatment and it increased with the last treatment. Thus quality of the fruits was better with spray applied 5—6 days before full bloom.

Table 5

Data at harvest (June 15, 1964) showing the effect of GA on Alamwick grape sprayed at prebloom stage

GA Concentrations (ppm)	Bunch length (cm)	Bunch weight (gg)	Berry weight (gg)	Berry number per cluster	Seedless berries (%)	T. S. S. (%)	Total acidity (%)
0	15.32	420.87	4.415	93.12	0	20.5	0.750
50	14.55	352.70	4.162	84.62	28.17	21.0	0.720
100	15.50	357.10	4.047	87.62	32.60	22.0	0.660
150	15.07	353.87	4.105	86.00	38.29	23.0	0.675
L.S.D. 5%	N. S.	N. S.	0.128	N. S.	—	—	—
Significance	—	—	**	—	—	—	—

N.S. = not significant; ** significant at 1% level.

(IV) Effect of GA on Alamwick grape applied at prebloom stage

Application of GA (50–150 ppm) to Alamwick grape at 4–5 days before full bloom induced 38.29% seedless berries (Table 5). However, bunch and berry weights were appreciably decreased and there was no notable decrease in number of berries per cluster suggesting that decrease in bunch and berry weights was mainly due to induced seedlessness. No toxic effects were observed on clusters. Quality of the berries was improved.

(V) Effect of GA on Bharat Early and Black Hamburg grapes sprayed at prebloom and postbloom stages

GA at 25, 50 and 75 ppm was applied to these varieties at (1) 13–15 days before full bloom plus another application at 7–8 days after full bloom (S''_1), (2) 7–8 days after full bloom only (S''_2). In general, in both these varieties, looseness of clusters and shot berry formation increased with the increase in GA concentration. Toxic effects were more apparent with double sprays. Higher concentrations of GA caused cracking and callusing of rachis. Bunch and berry weights and berry number were significantly reduced. Quality was improved to some extent with double sprays of GA.

Average seed content was reduced by double sprays of GA. Table 6 shows that GA application (25–75 ppm) at prebloom + postbloom resulted in an increased number of berries with one or two seeds coupled with a decrease in number of berries with three or four seeds. This implies that in these varieties the presence of at least 1 or 2 seeds is necessary for the growth of the berries.

Application of GA, even at very low concentrations (5–25 ppm) to Bharat Early and Black Hamburg at prebloom, full bloom and postbloom stages resulted in loose clusters and shot berry formation (Fig. 5). Bunch weight and number of berries were also decreased (Fig. 8).

Table 6

Effect of GA on the seed number of berries in Bharat Early and Black Hamburg

GA Concentrations (ppm)	Percentage of berries in each seed class											
	Bharat Early						Black Hamburg					
	5	4	3	2	1	0	5	4	3	2	1	0
(control)												
0	0	18.4	45.6	24.2	11.8	0	2.1	11.4	38.2	34.5	13.8	0
Prebloom + postbloom (S''_1)												
25	0	11.7	26.3	23.2	38.8	0	2.2	4.8	34.3	40.7	18.0	0
50	0	3.2	4.4	31.2	60.3	0.9	0	0	16.2	35.4	48.4	0
75	0	3.0	9.2	33.4	51.3	2.0	0	0	5.0	36.2	55.6	3.2
Postbloom alone (S''_2)												
25	0	27.4	39.4	23.1	10.2	0	2.4	16.2	39.2	24.4	17.8	0
50	0	17.3	41.2	22.1	18.4	1.0	0	16.2	32.7	30.2	20.7	0
75	0	19.5	43.1	22.2	14.2	1.0	2.3	9.9	35.4	25.3	25.0	2.1

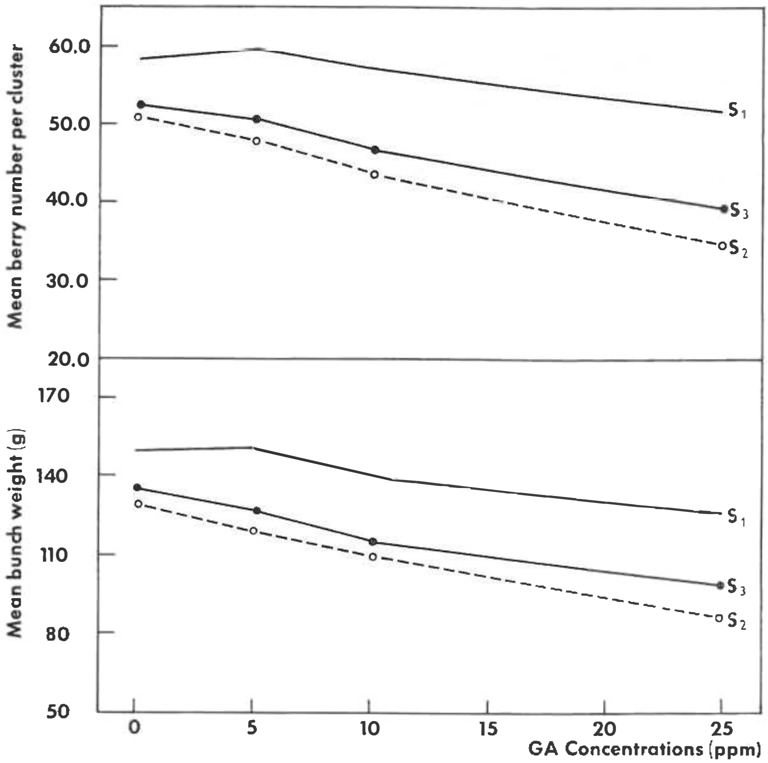


Fig. 8: Effect of GA on bunch weight and berry number in Bharat Early grape (1963 S_1 = 8—9 days before full bloom spray, S_2 = full bloom spray, S_3 = 2—3 days after full bloom spray.

Discussion

It has been shown that prebloom, full bloom and near full bloom applications of GA to Bharat Early and Black Hamburg cultivars reduced the berry number in a cluster. Similar results were obtained by Weaver and McCune (1959 a, b). In other seeded varieties, variation in sensitivity to exogenous GA was observed. In Bhokri, berry number was reduced by spraying at 10—11 days before full bloom but there was no thinning with sprays at 4—5 days before full bloom. In Anab-e-Shahi again, berry number was reduced by sprays at 7—8 days before full bloom but no thinning was recorded in Alamwick with sprays at 4—5 days before full bloom.

In Gros Colman (Pusa), application of GA at 5—6 days before full bloom or 2—3 days after full bloom increased the fruit set. Likewise, Gopalkrishna and Kerawala (1962) obtained increased fruit set in the shy-setting Gulabi grape and Manankov (1960) in Chaush variety. Randhawa and Sharma (1960) reported that Gros Colman (Pusa) has stamens shorter than pistils. It seems likely that GA application supplemented the pollination in this variety thereby increasing the fruit set. This dual effect of GA, i. e., increase in fruit set in some varieties and decrease in others, is probably due to different levels of endogenous growth factors.

A large number of seedless berries were formed in response to prebloom application of GA to Bhokri, Gros Colman (Pusa), Anab-e-Shahi and Alamwick cultivars. These results are in conformity with the findings of Rao *et al.* (1962), Kishi and Tasaki (1960) and Kajiura (1962). From the foregoing results it is evident that apart from varietal ensitivity, the stage at which GA is applied and the concentration of GA are the two most important factors in production of seedlessness. Thus by manipulation of these factors, in these varieties, quite a large number of seedless berries can be induced by GA application.

In Bharat Early and Black Hamburg, no seedless berries were formed in response to application of GA at prebloom + postbloom stages nor at the postbloom stage alone. Similarly, no seedless berries were recorded in Black Muscat grape following GA application at prebloom and immediately after full bloom stages (Dass 1965). GA application at prebloom + postbloom to Bharat Early and Black Hamburg grapes resulted in more berries with 1 or 2 seeds at the expense of berrie with 3 or 4 seeds. This suggests that at least 1 or 2 seeds are necessary in these varieties for berry growth. On the other hand, a large number of seedless berries were formed in Bhokri, Gros Colman (Pusa), Anab-e-Shahi and Alamwick grapes following GA application at prebloom stage.

Prebloom application of GA markedly affected the germination of pollen of seeded varieties and the extent of reduction in pollen germination was almost the same irrespective of the stage at which GA was applied. However, in Bhokri grape 41.4 and 97.5 per cent seedless berries were obtained following sprays at 4−5 days and 10−11 days before full bloom respectively. Itakura and Kozaki (1962) confirmed that apart from the effect on pollen germination, GA also affects ovule growth and development. Then the variation in percentage seedless berries induced at 4−5 days and 10−11 days before full bloom GA application to Bhokri grape, is probably due to extent of damage caused to ovules at these stages of GA application. This also implies that formation of seedless berries even with defective ovules in Bhokri, Anab-e-Shahi, Gros Colman (Pusa) and Alamwick but not in Bharat Early, Black Hamburg and Black Muscat grapes might be due to differential amounts of endogenous growth factors in these varieties. Apart from this, different amounts of anti-gibberellin factors might be present in them (Dass and Randhawa 1967). Probably a number of growth factors are interacting and detailed studies on this aspect would be well worth undertaking. Because of this differential ability to form seedless berries with exogenous GA, Dass and Randhawa (1965) suggested that varieties like Bhokri, Anab-e-Shahi, Gros Colman (Pusa) and Alamwick should be used as female parents in a programme of breeding for seedlessness.

Summary

Application of GA (25−50 ppm) to Anab-e-Shahi at prebloom stage decreased the bunch weight and berry number but weight of the berries was increased and up to 29.6% seedless berries were induced. GA sprays (25−50 ppm) at 2−3 days after full bloom increased the bunch and berry weights but few seedless berries were induced.

GA application (50−150 ppm) to Bhokri grape at 10−11 days before full bloom formed more than 90% seedless berries but bunch weights and berry number were reduced. With an additional second spray at 7−8 days after full bloom, berry size was improved but bunch weights were still reduced. At 4−5 days before full bloom application of GA, up to 41.4% seedless berries were induced but bunch and berry weights were again reduced. With another spray at postbloom stage bunch and berry weights were brought to the level of control.

In Gros Colman (Pusa) grape, GA application (50—100 ppm) at 5—6 days before full bloom and 2—3 days after full bloom also formed seedless berries but the bunch and berry weights were reduced. Fruit set was also improved at both stages of spray.

Prebloom GA application (50—150 ppm) to Alamwick grape, produced seedless berries up to 38.29% but bunch and berry weights were appreciably decreased. No effect on berry number was noticed.

Application of GA (25—75 ppm) at prebloom + postbloom and postbloom alone, decreased the bunch and berry weights and berry number markedly of Bharat Early and Black Hamburg varieties. Toxic effects like shot berry formation and cracking of rachis were noticed. No seedless berries were observed, however, more berries with 1 or 2 seeds were formed at the expense of berries with 3 or 4 seeds. Even GA application at low concentrations to these varieties (5—25 ppm) reduced bunch weights and berry number.

The differential response of these seeded varieties of *Vitis vinifera* is attributed to different amounts of growth factors and inhibitors present in them.

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