INFLUENCE OF GROWTH REGULATING CHEMICALS ON FRUIT SET, FRUIT QUALITY AND SEED VIABILITY OF MUSKMELON (*CUCUMIS MELO* L.)

by 28

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INTRODUCTION

Breeding programs with the muskmelon, Cucumis melo L. are often limited by poor set of hand-pollinated blossoms. Several reasons have been proposed for poor fruit set (1, 21, 22, 38).

Muskmelons are normally pollinated by honey bees, Apis mellifera L. (18, 21, 22, 23, 31). Therefore, it is necessary to control pollination in order to maintain pure breeding lines and to have selfed seed for selection purposes.

Many cultivars of Cucumis melo L. show an andromonecious sex expression (37). Hand-pollination of the perfect flowers cannot be easily accomplished unless the anthers are removed, and anther removal is a prerequisite for crossing different lines. After anther removal, the pistillate flower is pollinated using a staminate flower from the same plant for selfing, or from a different plant in crossing.

Chemical treatment of hand-pollinated flowers has been tried by various workers (2, 11, 27, 38). No treatment has proven entirely successful, since there is no widely used pollination technique except that of emasculation of the perfect flowers followed by hand-pollination. Jones' method with benzyladenine (BA) may prove to be an acceptable aid to increasing fruit set (11).

If chemical treatment can be used to increase fruit set it is important to know if the chemical affects melon quality. Important factors include melon weight, soluble solid content, flavor, melon size and external appearance. Seed number and viability are also important.

Environmental conditions influence fruit set in the muskmelon. Whitaker and Pryor (38) indicated that high temperature was involved in fruit set.

Experience of many workers indicate that high temperature, linked with low relative humidity, are important factors that can reduce fertilization success. Problems encountered in fruit set have been summarized by Mann (20).

The 1966 muskmelon breeding program at Kansas State University was severely limited due to very poor fruit set of hand-pollinated flowers. Weather conditions during the entire growing season were very hot and dry. This study was undertaken in an attempt to find a pollination technique that is suited to Kansas conditions and prevent a repeat of the 1966 failure.

The objectives of this study were (1) to evaluate effect(s) of the treatments of hand-pollinated flowers on fruit setting, (2) to evaluate effect(s) of the treatments on melon quality, seed number and viability and (3) to determine the influence of temperature and humidity on fruit set.

REVIEW OF LITERATURE

The problem of poor fruit set in muskmelon breeding programs is largely unsolved despite the efforts of various workers.

Wolf and Hartman (40) found value in removing the growing points and blossoms near the pollinated flower. On completely pruned plants fruit set was almost 80% of all pollinated flowers. Mann and Robinson (22) stated that fruit pruned vines rarely set more than 40% of the hand-pollinated flowers. Insect pollination of fruit pruned vines was about 70%. They felt that growing fruits have a marked effect in preventing further set. Mann (19) reported that set from open-pollinated flowers was much more uniform from day to day than from hand-pollinated flowers. Flowers which were hand-pollinated three times set no more fruit than flowers pollinated once. McGlasson and Pratt (16) thought it probable that a cycle of fruit set can be initiated at any desired

time by removing all fruits or perfect flowers for a few days. Fujishita (6) reported that there was no difference in rate of fruit set by pollen from a hermaphroditic flower or from a neighboring staminate flower.

Auxins were the first chemicals to be used to increase fruit set in the muskmelon. Burrell and Whitaker (2) thought that abscission of the young fruits was in some way related to the supply of a hormone-like or growth-promoting substance(s) and obtained 59% fruit set by applying indoleacetic acid to the stigma. Leopold (12) reported that in addition to the stimulation of fruit development, auxins applied at fruit set act to retard or delay abscission of the flower. Murray (26) reported that hormone sprays of alpha-naphthalene acetic acid and 2, 4, 5-tricholorophenoxyacetic acid resulted in better fruit set in cacao by increasing the length of time flowers remained on the tree. Whitaker and Pryor (38), using growth regulators separately and in combination, found that 4-chlorophenoxyacetic acid was the most effective in increasing fruit set in muskmelon. Alpha-naphthalene acetic acid alone did not increase fruit set in their experiment. Gustafson (9) applied indole butyric acid to several cucurbit species and although fruit growth was initiated, no fruits reached maturity. Norton (27) reported obtaining good crown set of melons by pollinating the afternoon before flowers normally open and using a saturated solution of indole butryic acid and alpha-naphthalene acetic acid in a sugar solution.

Knowledge of flower opening, temperature effects on fruit set, and pollen studies may be utilized by the plant breeder. Seaton and Kremer (28) found that the optimum temperature for anther and pollen dehiscence in muskmelon was 68-70° F. Griggs et al. (8) reported that muskmelon pollen stored for 30 days at a temperature of -18° C was capable of 98% germination.

Tarbaeva (30) reported that the best germination of muskmelon pollen was that collected from flowers between 7:00 and 11:00 AM. During storage at room temperature, (18-20° C), pollen viability was retained for 32-48 hours. Vasil (32) thought that the failure of seed setting could be the result of slow growth of the pollen tube or its early degeneration in the style. He found that pollen germination of Cucumis melo L. was best on a medium of 0.01% boric acid in 20% sucrose. Muir (24) suggested that pollen might contribute a co-enzyme or enzyme activator which, in conjunction with enzyme systems present in the ovary, might be capable of liberating active auxin from inactive precursors.

Discovery of the cytokinins has led to their being utilized to increase fruit set in several crops. Weaver et al. (34, 36) found that fruit set of the grape, Vitis vinifera, could be increased by the use of a cytokinin. However, Weaver et al. (35) reported that BA did not increase berry set or size in the Muscat grape. Crane (3) and Crane and van Overbeek (4) were able to induce parthenocarpy in the fig, Ficus carica, with cytokinins. Crane and van Overbeek (4) got a high initial percentage of fruit set but growth of a number of fruits ceased and abscised before maturity.

Jones (11) using BA on open-pollinated muskmelon flowers obtained increased fruit set and retarded senescence of fruits that failed to grow. He expressed little faith in auxins and gibberellins as a means of increasing fruit set.

MATERIALS AND METHODS

This experiment was conducted during the summer of 1967 at the Ashland Horticulture farm, Kansas State University. Soil type was a sandy loam with less than 1% organic matter. Two plantings were included in the experiment to

encompass a greater range of temperature differences. Each planting was of randomized complete block design, with four replicates per planting. Each replication contained five random treatments with five plants per treatment. Plants were spaced two and one-half feet apart in the row and five feet between rows. Replications, surrounded by guard rows, were twenty feet apart. Figure 1 shows the plots and position of the weather station. Analysis of variance for the data was according to Fryer (5). All data were analyzed with a least significant difference of 5%.

Foundation seed of the cultivar Gold Cup 55 was used for both plantings. Seeds for the first planting were planted in the greenhouse on May 17 and transplanted to the field on May 25. Plants were irrigated as needed and fertilized at a rate of 500 lb. of 10-10-10/acre. Insecticides and fungicides were applied as needed to maintain healthy growth. An attempt was made to obtain one self-pollinated melon per plant.

Pollination treatments were:

1) Control (OP): open-pollination of flowers by honey bees.
2) Standard treatment (HP): emasculation of perfect flowers followed by pollination with a staminate flower from the same plant.
3) Benzyladenine (BA): standard treatment followed by application of 2% benzyladenine in a lanolin paste to the base of the ovary (11). A glass stirring rod was used as the applicator.
4) Sugar-boron solution (SB): emasculation of perfect flowers followed by placement of a small drop of 0.01% boric acid, 20% sucrose solution on the stigma, and pollination with a staminate flower.
5) Indole butyric acid-naphthalene acetic acid (IBA-NAA): bud pollination the afternoon prior to normal opening of the flower. The perfect flower

was emasculated and pollinated with staminate flowers that were harvested the morning of pollination. The flowers were stored in a conventional refrigerator at 40° F and 90% relative humidity until 3:00 PM when pollination was conducted. After pollination a saturated solution of indole butyric acid and naphthalene acetic acid in one-half glycerine and one-half Karo syrup was applied to the sepals (27).

Pollinations were made on July 20, 21, and 22 with all open flowers on each plant being pollinated. Open-pollinated fruits were pruned from all plants before and after hand-pollination to eliminate competition between hand and open-pollinated fruits.

Flowers that were hand-pollinated were located from 3:00 to 4:00 PM the afternoon before pollination, and were clipped with a metal clip to prevent bees from visiting the flowers. All flowers were clipped again following hand-pollination, except for OP treatment.

Pollination techniques were rotated between the three workers doing the pollination except for the IBA-NAA treatment which was done by the author. The planting was irrigated June 18 to insure an adequate soil moisture level during pollination.

Seeds for the second planting were started on July 6 and set in the field July 11. Insecticides, fungicides, and irrigation were applied as needed to maintain healthy growth. Fertilization was the same as for the first planting. Pollination techniques, fruit pruning and irrigation prior to pollination were the same as the first planting. Dates of pollination were August 27, 28, and 29.

During pollination of both plantings continuous recordings of temperature and relative humidity were made with a Friez Hygro-Thermograph. A

standard United States Weather Bureau weather station located four inches above ground level was used to house the recording instruments.

After pollination, daily observations were made of each flower pollinated. Set and aborted flowers were recorded as soon as it was possible to determine their status.

Fruits were harvested at the full slip stage of maturity and evaluated for quality factors the same day. Each melon was:

1) weighed,
2) noted for external appearance,
3) sliced in cross section and total diameter and seed cavity measured,
4) tasted for flavor, and
5) tested for per cent soluble solids with a Bausch-Lomb hand refractometer.

All seeds and empty seed coats were cleaned from the melons, washed and allowed to dry.

In the first planting all melons that set were harvested. Many melons in the second planting were lost to diseases. Melon quality was very poor due to an extremely wet and cool ripening period. Melons of the second planting were not evaluated for quality factors. All melons that survived were harvested and the seeds were saved. Empty seed coats were separated from filled seed with a hand-operated seed cleaner and both were hand-counted.

Seeds were germinated in a controlled environment growth chamber. Two hundred seeds from each melon were selected at random and used in the test. If there was less than 200 seeds the total seed number was used. Seeds were sown in flats filled with vermiculite and covered one-half inch deep.

Due to limited space in the growth chamber it was necessary to test the seeds in two lots. The first received 12 hours of light and a day temperature

of 70-72° F. Slow germination was encountered, due to low night temperature, and adjustments were made for the second germination test. The day length for the second seed lot was 24 hours in order to maintain an air temperature of 80-82° F.

EXPERIMENTAL RESULTS

Tables 1 and 2 give the number of flowers pollinated for each treatment, and the per cent set for both plantings.

Table 1. Total number of attempted pollinations and per cent set for the first planting.

Treatment	Set/total pollinations	Per cent set
Control (OP)	8-86	9
Standard (HP)	2-62	3
Benzyladenine (BA)	31-68	46

Table 2. Total number of attempted pollinations and per cent set for the second planting.

Treatment	Set/total pollinations	Per cent set
Control (OP)	92-121	76
Standard (HP)	65-114	57
Benzyladenine (BA)	86-96	90

EXPLANATION OF PLATE I

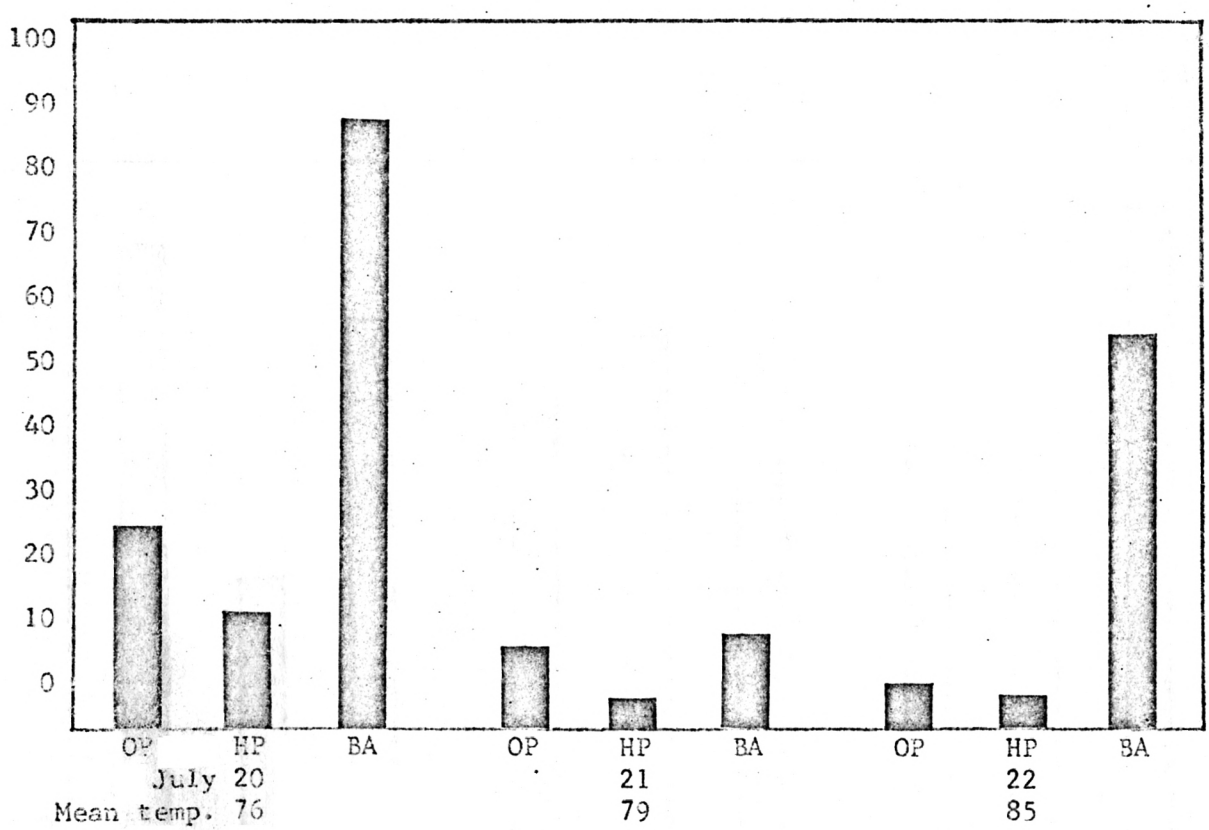
Fig. 1. Overall view of the first planting, with weather station in the center.

Fig. 2. Portion of a vine taken from the first planting showing injury due to BA.

The experiment did not allow for the same number of attempted pollinations for each treatment and it is suggested that in similar experiments serious consideration be given to this problem. Despite the uneven number of pollinations per treatment it can be seen from Tables 1 and 2 that BA increased fruit set enough on a percentage basis to be of practical value in breeding muskmelons. There was no fruit set with IBA-NAA and SB treatments.

Fig. 3 shows the per cent set of each treatment for each day of pollination in the first planting. The mean temperature for each day is also given. The mean temperature was determined by averaging the temperature at 8:00 AM and the highest for each day. Each day the relative humidity was 100% at 8:00 AM and dropped below 40% by 4:00 PM.

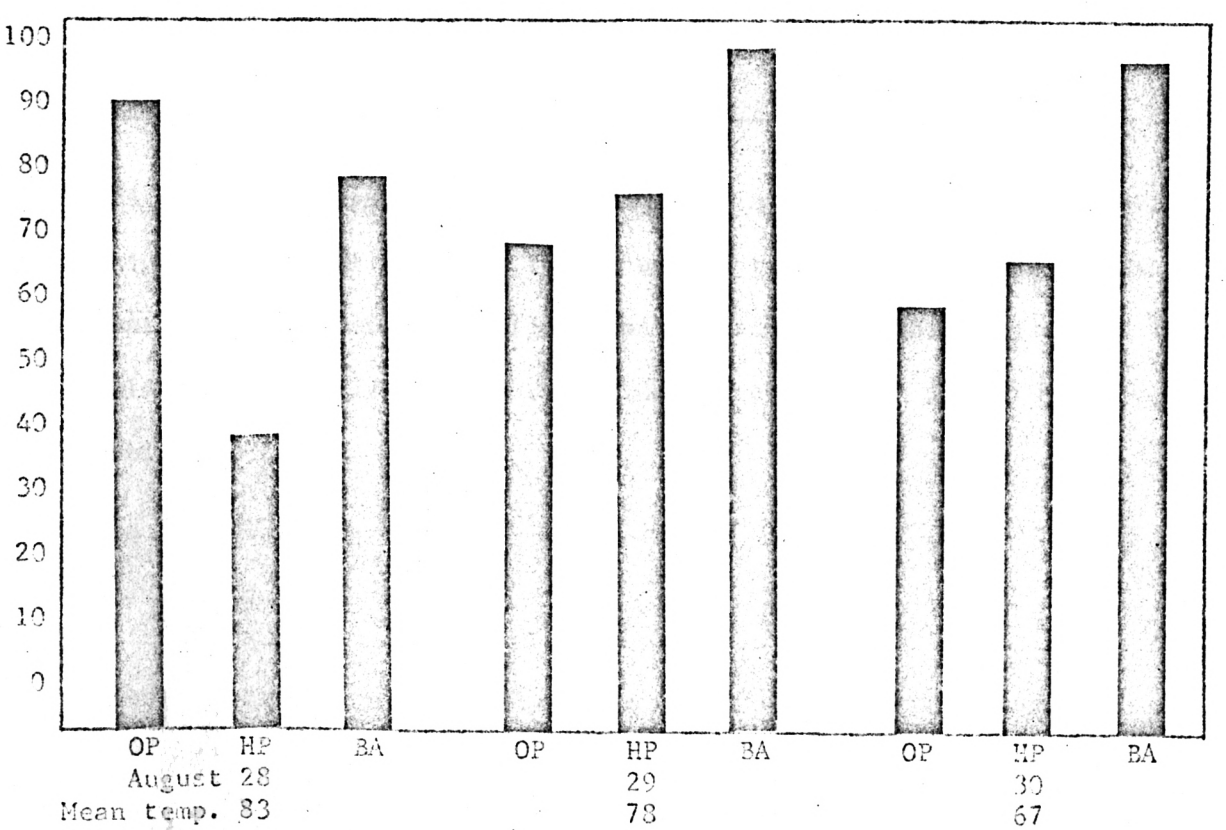
Fig. 3. Per cent set for each treatment per day and mean daily temperature for the first planting.



Best fruit set for all treatments was obtained on the first day, when the temperature was the lowest. Fruit set with BA was 90%. Set for the HP treatment was 13%, which is too low to be of value. Fruit set for open-pollinated melons was twice that of the HP treatment. The second day fruit set for the OP treatment was 8%, that of the HP treatment 0% and 9% for BA treatment. The third day fruit set of the OP treatment was 2%, HP treatment 0% and 56% for BA treatment. The HP treatment, for all three days would not have been satisfactory.

The same data for the second planting is shown in Fig. 4. Relative humidity was basically the same as in the first planting, but it is recognized that minor differences might affect fruit set. Fruit set on the first day was 90% for the OP treatment, 80% with the BA treatment, and 40% with the HP treatment. The highest mean temperature, 83° F, was recorded the first day.

Fig. 4. Per cent set for each treatment per day and mean daily temperature for the second planting.



Fruit set for the second day of pollination was 100% with the BA treatment. Set with the HP treatment was greater than that of the OP treatment, with values of 78% and 70% respectively. Set for the third day was similar to that of the second, with the HP treatment giving slightly better set than OP treatment. The BA treatment gave 98% set.

Table 3 shows that the number of seeds per melon were significantly reduced over the OP treatment by both the HP and BA treatments in the second planting.

Table 3. Mean seed number per melon for each treatment for the second planting.

Treatment	Mean seed number
OP	564
HP	453
BA	383
$LSD_{5\%}$ = 69	

BA treatment significantly reduced seed number over that of the HP treatment. Analysis of variance for OP and BA treatment for both plantings shows that the seed number was significantly reduced in the same manner in the second planting but there was no reduction in seed number due to planting date (Table 4).

Table 4. Mean seed number per melon for OP and BA treatments for both plantings.

Treatment	Mean seed number per melon		Mean
	Planting 1	Planting 2	
OP	570	564	567
BA	261	383	322
Mean	451	473	
$LSD_{5\%}$ treatment = 78			$LSD_{5\%}$ planting = NS

A reduction in seed number was paralleled with an increase in empty seed coats. Analysis of variance of data for the three treatments for the second planting (Table 5) shows that there was a significant increase in empty seed coats with the BA treatment over OP and HP treatments. The HP treatment gave more empty seed coats than that of the control (OP).

Table 5. Mean empty seed coat number for the three treatments for the second planting.

Treatment	Mean empty seed coat number
OP	85
HP	148
BA	216
$LSD_{5\%}$ = 16	

Analysis of variance of data for the OP and BA treatments for both plantings shows that there was a difference in the number of empty seed coats due to planting dates and treatments (Table 6).

Table 6. Mean empty seed coat number for OP and BA treatments for both plantings.

Treatment	Mean empty seed coat number per melon		Mean
	Planting 1	Planting 2	
OP	105	85	95
BA	419	216	318
Mean	262	150	
$LSD_{5\%}$ treatment = 85			$LSD_{5\%}$ planting = 85

There was no difference in seed germination due to treatment in the second planting (Table 7) and in the OP and BA treatments for both plantings (Table 8). All seedlings were grown until they had three true leaves and all appeared normal.

Table 7. Percentage seed germination for the three treatments for the second planting.

Treatment	Per cent germination
OP	89
HP	91
BA	87
$LSD_{5\%}$ treatment = NS	

Table 8. Percentage seed germination for OP and BA treatments for both plantings.

Treatment	Mean per cent seed germination		Mean
	Planting 1	Planting 2	
OP	95	89	92
BA	93	87	90
Mean	94	88	
$LSD_{5\%}$ treatment = NS			$LSD_{5\%}$ planting = NS

Melons from the three treatments could not be readily separated by appearance. BA treated melons showed slightly more cracking and slightly less netting than the OP and HP treated melons, but not enough to be of importance.

Internal appearance of all melons was the same, and there was no discernible difference in flavor. Only melons from the first planting were suitable for testing and a larger sample might have yielded some differences.

Some of the BA treated melons had very few seeds. One melon matured with two seeds. Two other melons had less than 50 seeds.

Fig. 2 shows a vine from the first planting on which a flower was treated with BA but did not set. This phytotoxic effect was present on two more plants in the same planting. During treatment of the flowers care was taken to limit BA to the flower but it is possible that BA was accidentally applied to the stem. The treated flower was directly above the tag.

DISCUSSION

Whitaker and Pryor (38) stated that to assure success in pollination it is necessary for the material (growth substance) to function only in assisting the young ovary in its development and preventing its separation from the plant. Benzyladenine may serve these two functions, but several points deserve further consideration.

Research findings have raised several questions that deserve attention. Luckwill (15) postulated that the initial stimulus to fruit growth in the majority of plants comes not from pollination but from the act of fertilization. He also stated that prevention of fruit abscission requires a steady supply of auxin to the abscission layer, which must originate in either the tissues of the seed or fruit.

Gustafson (10) found in female flowers of Cucumis sativus that there was a drop in auxin content at the time of flower opening. Mann and Robinson (22) found that fruit drop could not be blamed on embryo sac development, pollen tube growth, or early stages of seed development.

Sinnot (29) reported that in cucurbits increase in ovary size is accompanied by active cell multiplication before the opening of the flower, but after pollination growth of the ovary proceeds without appreciable cell division.

Goldacre and Bottomley (7) stated that cytokinin was responsible for cell division occurring in the receptacle of an apple fruitlet. Letham (14) found that the presence of stimulants of cell division in the developing apple receptacle is not entirely dependent on fertilization.

Crane (3) reported fruit growth is controlled by the ability of hormones to attract metabolites from other regions of the plant with the fruit

tissues surrounding the seeds tapping the metabolite supply and acting as storage organs.

Jones (11) stated that BA may have a mobilizing effect in developing muskmelon fruits. He thought that the mode of action of BA was to increase the competitive ability of treated fruits. In this experiment treated fruits were not in competition with developing fruits so other factors must be considered.

If after pollination, cell enlargement is chiefly responsible for increase in fruit size, one would wonder if application of a cytokinin would be important in this respect. It would be desirable to know if BA actually brings about cell division in the young fruit. Jones (11) reported that flowers which had failed to develop to maturity remained on the plant for several days and increased slightly in size.

It is possible that the cytokinin had a mobilizing effect on metabolites to the treated fruit and the fruit received a head start in growth compared to an insect-pollinated flower. The fact that a BA treated fruit reached maturity with only two viable seeds suggests that, if auxin is necessary to prevent abscission, BA caused a mobilization of auxin(s) in sufficient quantity to meet the demands of the developing fruits.

The reduction in seed number is in contrast to the findings of Jones (11) that the average number of seeds per melon remained unchanged by treatments. It would be interesting to know if empty seed coats could supply auxin without an embryo. No reason is proposed for the difference in the number of empty seed coats due to planting date.

Jones (11) reported the delay of abscission of BA treated muskmelon flowers and Crane (3) reported similar findings for the fig. In this

experiment no marked delay in fruit abscission was recorded. In many instances open-pollinated fruits remained on the plant several days before falling off.

Detailed studies of the fruit itself are needed before a satisfactory explanation of the effects of an exogenously applied cytokinin can be developed. Miller (23) aptly stated that attempts to explain kinetin (cytokinin) action in terms of its interaction with auxin, light, or other factors are fine except that they do not go far enough. Leopold (13) stated that the interaction of auxins, gibberellins, kinins, and growth inhibitors must be considered.

Fig. 3 shows that for the second day per cent set with BA treatment was lowered more in proportion than the other two treatments. This may have been due to poor pollinating technique. Some unknown factors may have been involved. The differences in set between the two plantings may have been due to differences in plant vigor and differences in temperature and relative humidity. The lowest night temperature during the pollination of the first planting was 59° F July 20. All daytime temperatures were above 87° F, 96° F was recorded for July 22. Relative humidity on July 21 dropped sharply after 8:00 AM. During pollination of the second planting the days remained below 86° F and the coolest night temperature was 60° F. The low temperature for the two days following the pollination period was 48° F. Relative humidity for the second day dropped off sharply after 9:00 AM.

Although plants from BA treated fruits were not grown to maturity it is unlikely that there was any interference in the genetic material of the seed. Wittwer and Dedolph (39) grew progeny of kinetin-treated Pisium sativus plants and found that no residual effects of the kinetin were transmitted through the seed.

Circumstances did not allow for proper evaluation of the effects of BA on melon maturity but it is possible that there might be some effect. Weaver and van Overbeek (33) working with cytokinin-treated grapes reported that 10% of the total number of grapes were still green at harvest. Mullins (25) reported that BA applied to the base of dormant grape cuttings stimulated growth of inflorescences but depressed the growth of leaves and shoots. He thought other growth substances might be involved. In the fig Crane and van Overbeek (4) found that cytokinin-induced parthenocarpic fruits matured about a week earlier than those of pollinated fruits.

The phytotoxic effect of BA was unusual in appearance and that it only occurred on three treated plants. The nature of the injury suggests that cell division and differentiation were adversely affected. Crane and van Overbeek (4) reported that the only vegetative response of figs sprayed with BA was abscission of the youngest leaf on treated shoots.

The use of BA should give good fruit set in muskmelons over a wide range of conditions. A reduction in seed number should be expected and melon selection should not be based on treated melons alone until more data on the effects of BA on melon quality is available.

No fruit set was obtained with the sugar-boron and IBA-NAA treatments. It was hoped that the sugar-boron solution would increase pollen germination with a resultant decrease in time from pollination to fertilization. Evidently this treatment was detrimental to pollen and fertilization was not accomplished.

Failure of the IBA-NAA treatment was probably due to several factors. Many times it is difficult to tell with certainty if a flower will open the following day. Because the flower was pollinated the day before it would normally open it was difficult to keep from damaging the flower when

pollinating. Also pollination was conducted at 3:00 PM during the warmest portion of the day when high temperature and low relative humidity could adversely affect pollen germination and growth. Best pollination success is to be expected when pollination is as close to natural as possible.

Results of this experiment indicate that the use of BA will increase fruit set of hand-pollinated muskmelon flowers over non-treated flowers.

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INFLUENCE OF GROWTH REGULATING CHEMICALS ON FRUIT SET, FRUIT QUALITY AND SEED VIABILITY OF MUSKMELON (*CUCUMIS MELO* L.)

by

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Muskmelon breeding programs are severely hindered by poor set of hand-pollinated flowers. Despite the efforts of many researchers this problem has remained largely unsolved. A successful aid to increasing fruit set would save time and labor in most breeding programs.

Auxins have been largely unsuccessful in increasing fruit set in the muskmelon (2, 27, 38). The cytokinin, benzyladenine (BA), has increased fruit set of open-pollinated muskmelon flowers (11). Jones (11) stated that BA may have a mobilizing effect in developing muskmelon fruits. He thought that increased fruit set was due to BA increasing the competative ability of treated fruits.

In the summer of 1967 benzyladenine (BA), indole butyric acid-naphthalene acetic acid (IBA-NAA), and a sugar-boron solution (SB) were applied to hand-pollinated flowers to determine their effects on fruit set, fruit quality, and seed viability. Insect pollinated (OP) and untreated hand-pollinated flowers (HP) were used for comparison. Two plantings were included to encompass a greater range of environmental differences.

Results of both plantings showed that BA increased fruit set, reduced seed number, increased number of empty seed coats and had no effect on seed germination. There was no fruit set with IBA-NAA and SB treatments. Open-pollinated flowers set better than untreated hand-pollinated flowers. Fruits could not be evaluated for quality due to diseases present during the growing season.