

**POLLINATION AND FRUIT SET
OF ACEROLA (MALPIGHIA GLABRA L.)**

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INTRODUCTION

The discovery by Asenjo and Freire de Gusman in 1946 (5) that Acerola, Malpighia glabra L., is one of the richest known sources of natural ascorbic acid has aroused much interest in this plant. The high natural ascorbic acid content of the fruit, a desirable source of vitamin C for food supplements and additives, has resulted in increased planting of this crop in Puerto Rico, Florida and Hawaii.

As with most new crops possessing potentialities, various cultural problems were encountered. One of these problems is that of fruit set. Ledin (27) in Florida reported that large numbers of bees were attracted to the flowers of the plant. The natural pollination resulted in heavy yields. In Hawaii, although flowers are produced in abundance, the natural fruit set is low. This study was initiated to determine factors contributing to low yields under natural conditions and the possible use of growth regulators to increase yields.

BOTANICAL DESCRIPTION

The Acerola belongs to the family Malpighiaceae, under the genus Malpighia, which consists of 30 to 40 species and is considered native to Tropical America. To the present time, there has been much controversy relative to the correct specific name for the cultivated varieties. Publications from Puerto Rico (1,3,4,5,9,33) refer to the Acerola as M. puniceifolia L., whereas those of Florida (25, 27) as M. glabra L. Ledin (27) stated that M. glabra L. was used in 1753 and M. puniceifolia L. in 1762. Therefore, based on the taxonomic principle of priority, M. glabra L. appears to be the acceptable name for Acerola.

The botanical relationships and plant characteristics have been described (4,27,33). The plant is a small tree with either upright or

droopy branching habits (Fig. 1). The entire, simple leaves with short petioles are borne on two types of woody branches--new terminals and short shoots or lateral spurs. The former have longer internodes and larger leaves. The $\frac{3}{4}$ to 1 inch in diameter flowers are produced in inflorescences in the axils of the leaves and are characterized by 5 petals, 4 of which are clawed. Petal color ranges from a pink tinge to dark pink. There are 10 stamens, 3 styles and a superior ovary. In Hawaii, flowering cycles occur approximately every 20 to 28 days, commencing in April and continuing into the latter part of November with frequent scattered flowering until January or even later. Three to 6 days after initial flowering, peak flowering occurs and it is not unusual for ripe fruits and profusion of flowers to be present on the plant at the same time.

The shallowly lobes, berry-like drupaceous fruits in Hawaii mature approximately 21 days after floral anthesis. The thin-skinned fruits are light orange to dark red when ripe. The 3-stoned pyrenous fruit ranges in weight from 2 to 10 grams and from $\frac{1}{2}$ to 1 inch in diameter.

Prior to 1954, a few isolated *Acerola* plants were in Hawaii. In the spring of that year, Mr. Frederick E. Haley^{1/} introduced seeds and rooted plant material from Puerto Rico and later from Florida for the first commercial orchard planting at Pupukea, Oahu. In 1955, 10 seedlings (PI 209269, 209270) were introduced by the Hawaii Agricultural Experiment Station from the United States Plant Introduction Garden, Glendale, Maryland.

^{1/} Personal communication from Mr. Raymond Haley, son of the late Mr. Frederick E. Haley.



FIGURE 1. TYPES OF GROWTH OF ACEROLA.

LEFT: UPRIGHT. RIGHT: DROOPY.

REVIEW OF LITERATURE

The low natural fruit set of horticultural fruit plants may be attributed to many factors. Dichogamy (2,46,57), production of inviable pollen (8,10), incompatibility (23,30,41,49,53) and structure of flowers that prevents insect pollination (44) are some of the causes of low fruit set.

Prevention of floral abscission and production of ovary swelling by use of water extracts of pollen by Fitting (17) was the first indication that specific substances were involved in parthenocarpic fruit set. Yasuda (58) produced parthenocarpic eggplant fruits by the use of pollen extracts. Thimann (52) found that auxins were involved and Gustafson (19,20) induced parthenocarpic fruit set and development in tomato, pepper, *Petunia* and *Salpiglossus*, by the application of synthetic growth regulators.

Gustafson (21) reported that varieties of several fruits which naturally set fruits parthenocarpically have higher auxin content than those closely related varieties which do not. The great increase in auxin content in the ovaries of *Nicotiana tabacum* immediately after pollination as reported by Muir (34) and the findings of Gustafson have shown that it is possible to artificially induce fruit set by application of additional auxins.

One of the first crops in which growth regulators were used to set fruits when conditions of pollination and fertilization were unfavorable was tomatoes. Under certain environmental conditions, such as weak light intensity (43,54,56) and high day temperatures (47), elongation of the pistils takes place and extends beyond the stamens. In these

cases, growth regulators have been used to set fruits which resulted in increased yield and increased fruit size. Others (22,24,36,37,45) have also reported these beneficial results when growth regulators were used in fruit setting of tomatoes.

In addition to increase of fruit set and production of seedless tomatoes, plant growth regulator sprays have been used on vegetable crops such as snapbeans (38,55), on fruit crops such as certain varieties of pears (7,18,40), Calmyra figs (13) and avocado (50), and on ornamental crops such as American holly (11).

MATERIALS AND METHODS

Materials

Acerola plants used in this study were from various sources. Seedlings 6663 (sour type) were derived from seeds acquired from the University of Florida in June 1955. Clone 269-2 (sour type) is one of the selections from the original 10 seedlings received from the United States Plant Introduction Garden, Glendale, Maryland. Clones Puerto Rico (sour type) and B-17 (sweet type) were received from Puerto Rico in 1955 and 1956, respectively. Florida Sweet is a selection received in 1957 from the Sub-Tropical Experiment Station at Homestead, Florida. Clones HSPA (sour type) and Maunawili (sour type) were received from the Hawaiian Sugar Planters' Association (HSPA). The Maunawili was obtained from a single plant growing in the HSPA sub-station at Maunawili, Oahu. Clones H-5 (sweet type) and 13½ (sour type) are designations of the Hawaiian Acerola Company and are of unknown parentage.

Growth regulators utilized were in crystalline forms. Where the acid forms were used, 95 percent ethyl alcohol was used as the bridging

solvent. In addition, a small amount of commercial wetting agent (Tween 20) was added to the distilled water solution.

Methods

All preliminary field and laboratory experiments were conducted on the Manoa campus of the Hawaii Agricultural Experiment Station, University of Hawaii. Experiments requiring large numbers of clonal plants were conducted at the Puna orchard of the Hawaiian Acerola Company at Puna, Hawaii.

Branch treatments were used in pollination studies conducted in the field. The peduncles of flowers and flower buds were marked with India ink. Bagging and emasculation were not practiced except in one phase of the study because of the slowness of the processes, danger of damage to the flowers and the normally low percent set.

Artificial pollination was carried out with bristle brushes as the pollen transferring agent. In self pollination, pollen from the same flower or from flowers of the same plant was used. In cross pollination, sources of pollen were from flowers of different clonal or seedling plants.

Fruit set was determined 6 days after application of treatments. Fruits at this age were approximately 3/16 inch in diameter and only those fruits which had peduncles marked were considered in evaluation of data.

The aqueous growth regulator solution was applied to branch treatments by a small hand atomizer. A 4½-gallon knapsack sprayer was used for whole tree treatments. Where whole trees were involved, a plastic screen 6 x 12 feet was employed to minimize drift to trees with

different treatments.

All laboratory experiments were carried out at room temperatures, unless otherwise specified.

After it was ascertained chromatographically following methods outlined by Strohecker, et al (51), that there were no interfering substances, the iodate titration method after Ballentine (6) was used for ascorbic acid determination.

Pollen was germinated in vitro on 1 percent agar. Media containing 8 to 12 percent sucrose (by weight) gave the best result. Pollen was sown on the agar-sucrose media and after 24 hours the percent germination was determined. For each sucrose level, random samples of 10 to 15 microscopic fields were observed.

In the study of dehiscence of anthers, flowers were excised from the plants at prescribed intervals and the anthers were checked with a dissecting microscope.

Two methods were used to derive the approximate date of flowering and peak flowering. Since fruits 1/3 ripe were considered mature for harvest under plantation practices at the Puna orchard, the approximate date of flowering for untreated trees was determined by deducting 20 days from the harvest date. The treated trees were considered to be at their peak flowering when at least 50 percent of the flowers on randomly sampled inflorescences had anthesised.

For analysis of data following a binomial distribution, confidence interval tables from Snedecor (48) were used. Yield data were summarized by analysis of variance as outlined by Snedecor (48). The Duncan's multiple range test (14) was used to delimit differences between means.

To determine the mean weight of fruits, estimates from random samples drawn from each treatment at each harvest were used.

EXPERIMENTAL RESULTS

Floral Anthesis, Stigma Receptivity and Anther Dehiscence

Ten to 14 days before anthesis of the flowers, light green flower buds become visible in the axils of the leaves of the previous vegetative flushes and frequently on recent flushes. As the peduncle increases in length and the flower buds grow, the pink colored corolla becomes exposed through the sutures of the sepals.

The opening of the flowers seems to be related to temperature. It was found that when excised flower buds which normally opened within 24 hours were placed in water at 58 degrees F., anthesis did not occur. When placed in water at 62 to 64 degrees F., anthesis commenced within a relatively short time. Illuminating the flowers at 58 degrees F. had no effect on the anthesis of the flowers.

Under summer field conditions in Hawaii, anthesis of flowers was usually completed by 5 or 6 o'clock in the morning, with the stigma becoming receptive almost immediately. When occasional low night temperatures (less than 66 degrees F.) were experienced during the summer, it was observed that complete opening of the flowers was delayed from 2 to 3 hours.

Dehiscence of the anthers was closely related to anthesis of the flowers except in the case of two clones (Table I). These two sweet types, B-17 and Florida Sweet, commenced dehiscence of the anthers approximately at noon and increased as the day progressed. However, this increase was not as pronounced as that of the sour types, over a

similar length of time after initial dehiscence.

TABLE I. PERCENT DEHISCENCE OF ANTHERS OF 6 CLONES OF AGEROLA (OCT. 18, 1959) GROWN AT THE UNIVERSITY EXPERIMENTAL FARM.

Clones	Temperatures (degrees F.) and Time (hour)						
	5 AM 73	6 AM 73	7 AM 76	8 AM 79	9 AM 80	12 Noon 88	3 PM 86
Maunswili	12.00	20.00	17.00	46.00	72.00	78.00	--
HSPA	18.00	28.00	62.00	82.00	86.00	88.00	--
269-2	22.00	58.00	76.00	88.00	100.00	96.00	--
Puerto Rico	6.00	28.00	46.00	74.00	86.00	82.00	--
B-17	0.00	0.00	0.00	0.00	0.00	2.00	9.00
Florida Sweet	0.00	0.00	0.00	0.00	0.00	0.00	6.00

Open Pollination

The percent fruit set resulting from open or natural pollination was found to be much lower than that induced by hand pollination. The mean natural set ranged from 1.31 to 11.53 percent (Table II).

Self Pollination

In the hand self pollination study, a total of 983 flowers from three clones and four seedlings was used. Data in Table III show that fruit set increased significantly over open or natural pollination, except with clone B-17 and seedling 6663, 4BT-43. As shown in Table I, clone B-17 commenced dehiscence of anthers at approximately noon. This retarded process and the low percent of anthers dehiscid could have

TABLE II. PERCENT FRUIT SET AND CONFIDENCE INTERVALS FOR FRUIT SET OF ACEROLA BY NATURAL POLLINATION.

Plant or clone	Number of flowers	Number of fruits set	Percent fruit set	Confidence intervals 95 percent
Maunawili	458	14	3.05	1.27-5.45
269-2	75	3	4.00	1.00-12.50
B-17	75	4	5.33	1.50-14.50
Maunawili*	53	1	1.88	0.00-10.74
6663*, 4BT-43	76	1	1.31	0.00-7.88
SAT-28	52	6	11.53	4.88-23.52
SAT-34	41	2	4.87	0.00-18.00

* Seedlings.

resulted in less time for pollen or a smaller amount of pollen to fall on the stigmatic surfaces and induce fruit set. Unfortunately, enther dehiscence for seedling 6663, 4BT-43 was not checked before the plant was rogued in the selection program.

The difference in percent fruit set between the seedling and clone of Maunawili was considerable and may be due to genetic variability. This is in agreement with Ledin's report (26) that Acerola grown from seeds are highly heterozygous and show considerable variation in flowering and fruiting.

Cross Pollination

Although cross pollination was carried out without emasculation, the increase in fruit set over self pollination shows the influence of cross pollination. Of a total of 1150 flowers involved in this study,

TABLE III. PERCENT FRUIT SET AND CONFIDENCE INTERVALS FOR FRUIT SET OF ACEROLA BY HAND SELF POLLINATION.

Plant or Clone	Number of Flowers	Number of Fruits Set	Percent Set Observed	Confidence Intervals 95 Percent
Maunavili	419	121	28.87	23.66-34.34
269-2	75	28	37.33	30.00-54.00
B-17	75	5	6.66	2.50-16.50
Maunavili*	105	17	16.19	10.06-25.87
6663*, 4BT-43	142	5	4.22	2.27-10.44
5AT-28	18	9	50.22	26.60-74.40
5AT-34	49	27	55.10	40.70-69.50

* Seedlings.

the percent fruit set ranged from 6.66 to 74.12 percent. In all cases, except where clone B-17 was used as the pollen source, increases in fruit set resulted from cross pollination (Table IV).

Comparisons of fruit set from cross pollination using the Maunavili clone and seedling as the female and 269-2 as the common pollen source, showed that the clone gave significantly higher fruit set. Thus the fruit set of the seedling has been shown to be lower, both when selfed and when crossed, using a common pollen source.

The percent fruit set from open, self and reciprocal cross pollinations of three clones was analyzed using confidence intervals. Significant differences were obtained among clones (Table V). Reciprocal crosses of 269-2 x Maunavili resulted in higher fruit set than

TABLE IV. PERCENT FRUIT SET AND CONFIDENCE INTERVALS FOR FRUIT SET OF ACEROLA BY HAND CROSS POLLINATION.

Plants	Number of Flowers Crossed	Number of Fruits Set	Percent Set Observed	Confidence Intervals 95 Percent
Maunswili x 269-2	399	299	72.43	66.59-77.41
269-2 x Maunswili	75	52	69.33	62.00-79.00
Maunswili x B-17	75	5	6.66	2.50-16.50
B-17 x Maunswili	75	40	53.33	48.00-70.00
Maunswili x 6663, SAT-28	201	149	74.12	66.69-80.66
Maunswili x 6663, SAT-34	64	37	57.81	50.32-74.36
Maunswili* x 269-2	90	29	32.22	24.60-45.60
6663, SAT-34* x 269-2	55	36	65.45	54.00-80.20
6663, SAT-28* x 269-2	40	28	70.00	59.50-80.24
6663, 4BT-43* x B-17	76	7	9.34	4.44-20.24

* Seedlings.

reciprocal crosses of B-17 x Maunswili. Differences were also observed for reciprocal crosses of B-17 x Maunswili. Lower fruit set resulted when B-17 was used as the pollen source. In addition, when the total set for all three methods (open, self, and cross) was analyzed, significant differences were also present. Hand pollination was superior to natural means and cross pollination was better than self pollination.

Pollination, whether carried out by insects, wind or by man, is essentially the transfer of pollen from the exposed microsporangial surfaces to the stigmatic surfaces. To ascertain whether sufficient pollen was transferred naturally to the stigmatic surfaces, stigmas of

TABLE V. TOTAL FRUIT SET AND CONFIDENCE INTERVALS FOR CLONES OF ACEROLA SET BY OPEN, SELFED AND CROSSED POLLINATION

Plants			Treatments			Total Fruits Set By Clones	Percent Set Observed	Confidence Intervals 95 Percent
			Open	Selfed	Crossed			
Maunawili	x	269-2	0	26	60	86	38.22	32-44
269-2	x	Maunawili	3	23	49	80	35.55	30-42
B-17	x	Maunawili	4	5	40	49	21.77	17-28
Maunawili	x	B-17	0	20	6	26	11.55	8-17
Total Set by Treatments			7	79	155			
Percent set Observed			2.33	26.33	51.66			
Confidence Intervals 95 Percent			1-5	20-32	46-58			

flowers left on the plants until mid-afternoon were examined. It was found that 1.24 pollen grains per stigma were present or 3.72 pollen grains per flower. The stigmas of many flowers were entirely void of pollen. The few pollen grains deposited on stigmas indicate the lack of pollinating agents.

The whole stigmatic surfaces of hand pollinated flowers were covered with pollen making accurate counting impossible.

When carpels of fruits from open, self and cross pollinated flowers were examined (Table VI), results showed that a positive relationship existed between fruit set and the number of fully formed endosperms. This suggests that low fruit set in *Acerola* under natural conditions is due to the lack of pollen transfer to the stigmatic surfaces.

The increase in fruit set by cross pollination over selfing, except when clone B-17 was used as the pollen source, indicates some degree of self incompatibility. Clone B-17 for all practical purpose is an undesirable source of pollen.

Pollination Requirement

Twenty fruits each from 15 seedlings and 2 clones were examined. Results show that under natural conditions, the number of fully formed seed in mature fruits varied from plant to plant and ranged from 0 to 38.33 percent per fruit (Table VII.).

Since fruit set and development were observed without seed formation, it was necessary to determine whether or not pollination was essential for fruit set. A total of 437 flowers from three clones was emasculated and bagged. Data, recorded in Table VIII, for the three clones indicate that pollination is not obligatory and that fruit set may occur through

TABLE VI. NUMBER OF FULLY FORMED ENDOSPERMS IN FRUITS OF ACEROLA RESULTING FROM OPEN, SELF AND CROSS POLLINATION.

Number of Endosperms	Treatments														
	Open					Self					Cross				
	0	1	2	3	Total Fruits	0	1	2	3	Total Fruits	0	1	2	3	Total Fruits
Clones or Plants															
6663, SAT-28	2	14	0	0	16	2	10	4	0	16	2	12	4	1	19*
6663, 4BT-43	18	4	0	0	22	12	8	0	0	20	2	5	0	0	7**
B-17	24	0	0	0	24	4	1	0	0	5	3	14	7	2	26***
269-2	22	2	0	0	24	9	8	3	2	22	7	9	5	6	27***
Mamawili	31	11	0	0	42	6	29	10	1	46 ⁶	15	13	33	13	74*

* Crossed with 269-2
 ** Crossed with B-17
 *** Crossed with Mamawili

TABLE VII. PERCENT FULLY FORMED SEEDS IN FRUITS OF OPEN POLLINATED SEEDLINGS AND CLONES OF ACEROLA.

Plant	Number of Fruits with the Following Number of Fully Developed Seeds				Number of Fully Formed Seeds from 60 Carpels	Percent Carpels with Fully Formed Seeds
	0	1	2	3		
269-2	18	2	0	0	2	3.03
Maunawili	14	6	0	0	6	10.00
6663, 4AT28	2	14	3	1	23	38.33
✓ 3BT3	20	0	0	0	0	0.00
3BT5	20	0	0	0	0	0.00
6BT17	20	0	0	0	0	0.00
6AT22	3	15	2	0	19	31.66
✓ 3BT7	4	11	5	0	21	35.00
3AT4	20	0	0	0	0	0.00
4AT14	19	1	0	0	1	1.66
5BT45	18	2	0	0	2	3.32
6AT26	6	11	3	0	17	28.33
6AT36	19	1	0	0	1	1.66
6AT38	19	1	0	0	1	1.66
5BT33	6	9	5	0	19	31.66
4BT17	5	12	3	0	18	30.00
5AT6	19	1	0	0	1	1.66

vegetative parthenocarpy.

TABLE VIII. PERCENT FRUIT SET OF ACEROLA AFTER EMASCULATION.

Clones	Number of Flowers Emasculated	Number of Fruits Set	Number of Fully Formed Endosperms	Percent Fruit Set
B-17	130	0	-	0.00
Maunawili	209	3	0	1.43
269-2	98	2	0	2.04

Muir (34) reported that parthenocarpic (stimulative) fruit set may take place as a result of the germ tube contributing an enzyme which releases a growth hormone from an inactive combination present in the style and ovary. The presence of ovules in many of the mature fruits and the extremely short germ tubes of pollen when germinated in vitro (to be discussed later) in relation to the stylar length, suggest that both vegetative and stimulative parthenocarpy may be involved.

Pollen Distribution

Wind. The pollen of Acerola is sticky and therefore does not readily become windborne. To determine the effect of wind as a pollen transferring agent, glass microscope slides 1 x 3 inch covered with thin layers of vaseline and/or agar, were placed on the leeward side of the plants at distances of 3 and 6 feet facing the prevailing wind. All slides were exposed for a period of 6 hours and removed to the laboratory for a count of the number of pollen grains trapped on the slides.

At 3 feet, the mean number of pollen grains trapped from 14 trials was 4.8 per slide. At 6 feet, the mean number from 11 trials was 2.2 per slide, indicating that wind is a poor pollinating agent.

Insects. A Florida report (27) states that bees are attracted in large numbers to the flowers of Acerola. Observations of experimental plantings at the Mid-Pacific and the Waimanalo Experiment Farms and at the Puna orchards did not conform to this report. Honey bees, Aphis mellifera and Syrphid flies, Eristalis arorum were the only insects readily visible but plantings of 30 to 40 trees in full bloom attracted fewer than a dozen of each of the two insects and occasionally were these insects seen working on the flowers. On the assumption that insect population was not abundant in the immediate vicinity of the trees, honey bee hives were moved to within 50 feet of the edge of the field and finally adjacent to flowering plants. Even these measures did not greatly increase the number of bees working on the flowers.

Another insect which could be detected with the aid of magnifying glasses and was almost always present on the flowers was a species of thrips.

Pollen Germination and Pollen Viability

Preliminary studies indicated that artificial germination of pollen grains of Acerola was successful with 1 percent agar and 4 to 14 percent sucrose (by weight). The 8 to 12 percent sucrose levels were optimum.

When pollen from three clones was germinated in petri dishes at three sucrose levels (8, 10, and 12 percent), there were differences between clones at each level. Table IX shows that at all levels, clone 269-2 had a higher germination rate than clones Maunawili and HSPA and

that there were no differences at each level between Maunswili and HSPA. For all three clones, maximum germination was obtained at 10 and 12 percent sucrose levels.

Initial germination of pollen on artificial medium was quite rapid. One-half hour after pollen was sown on the medium, germ tube growth was observed, with maximum germination reached within 4 to 6 hours.

TABLE IX. PERCENT GERMINATION OF POLLEN FROM THREE CLONES OF ACEROLA WITH 1 PERCENT AGAR AND AT THREE SUCROSE LEVELS

Percent Sucrose	Clones	Percent Germination Observed	Confidence Intervals 95 Percent
8	Maunswili	21.51	17.46-27.31
	HSPA	18.26	13.38-22.75
	269-2	49.65	44.17-55.83
10	Maunswili	36.03	30.33-41.67
	HSPA	28.01	22.42-33.72
	269-2	64.01	58.05-69.95
12	Maunswili	32.81	27.40-38.60
	HSPA	27.30	21.13-32.87
	269-2	66.00	60.20-71.80

Although germination commenced within a relatively short time, the germ tube lengths were comparatively short (mean length 0.278 mm). Upon termination of growth viscous exudates, secreted by the growing germ tubes, were present on the ends of the tubes (Fig. 2). Sucrose



**FIGURE 2. GERMINATED POLLEN GRAIN OF ACEROLA
WITH VISCOUS EXUDATE ON TERMINAL
OF GERM TUBE (170X).**

may not be the only growth factor needed for pollen germ tube growth. The stigma may provide other factors. When macerated stigmas and styles or extracts from stigmas and styles were placed on the agar, some increase in germ tube lengths resulted.

When pollen from flowers 1 day after anthesis was sown on artificial media, germination rates were less than 0.02 percent. For all practical purposes the day-old pollen can be considered non-viable.

Selection of Growth Regulators

The low natural fruit set, attributed essentially to lack of pollen transferring agents, indicated the feasibility of the study of growth regulators for induction of fruit set. Lewis (29) and Mitsch (39) reported that, with few exceptions, parthenocarpic fruit set was most easily induced in fruits which under open pollination contained large number of seeds. However, Griggs, et al (18), Batjer and Uota (7) using 2,4,5-Trichlorophenoxypropionic acid and Osborne and Main (40) using Naphthoxypropionic acid on pears and Connors (10) using 2,4-Dichlorophenoxyacetic acid on holly, have increased fruit set of these crops which contain relatively few seeds. On the basis of these data and the production of large numbers of seedless fruits in Acerola under natural conditions, the following organic compounds were utilized to determine their efficacy in promoting fruit set: Naphthaleneacetic acid (NAA) and its sodium salt (NaNAA), Naphthoxyacetic acid (NOAA) and its sodium salt (NaNAAA), Indoleacetic acid (IAA), Indolebutyric acid (IBA) and para-Chlorophenoxyacetic acid (PCA).

In this study, seedling and clonal plants were employed. For seedlings, all treatments of each compound tested were confined to an

individual tree. In cases where clones were used, all treatments were applied to trees within a clone, and the data for each compound compiled and presented as percent fruit set.

Table X shows that of the 5 acid and 2 sodium salt forms tested, PCA and IBA were most effective, the former over a wider range of concentrations. Although increase in fruit set with PCA was obtained up to 1000 ppm, fruits set with 500 ppm and 1000 ppm were deformed and very small. At lower concentrations, the fruits were less deeply lobed and much larger in size, though smaller than those of the control.

TABLE X. MEAN PERCENT FRUIT SET OF ACHROLA TREATED WITH FIVE ACID AND TWO SODIUM SALT FORMS OF GROWTH REGULATORS.

Treatments (ppm)	Growth Regulators						
	MAA	IBA	IAA	MeNAA	PCA	NaNOAA	NOAA
0	0	.46	19.47	4.77	4.83	0	4.34
10	2.38	.56	16.02	7.29	33.82	3.84	0
50	2.12	2.73	15.75	6.18	88.17	0	0
100	0	61.46	15.15	13.06	89.04	0	0
500	-	-	-	8.95	86.66	-	-
1000	0	-	-	-	82.40	-	-
5000	0	3.96	12.84	22.00	-	13.72	-

Although tremendous increase in fruit set was realized with PCA, undesirable effects in the form of leaf curling and yellowing of vegetative terminals resulted. Mature leaves present at the time PCA was applied showed no effects. Only young expanding leaves and leaves

which appeared after application of the chemical were affected. In severe cases, the terminals turned yellow and ultimately died. The severity of these conditions was related to the concentrations used. At 10 ppm and 50 ppm, leaf curling was less pronounced and less time was required for leaves to return to normal.

Although IBA sprays resulted in less fruit set than PCA, the undesirable leaf effects were not present. Fruits on the other hand, were similarly affected, though to a lesser degree.

Besides the undesirable effects, handling qualities of the fruits were improved. Fruits were more solid and did not bruise as easily as those from natural set. This is an important consideration from the standpoint of post harvest physiology. Ponting and Joslyn (42) working with apples have reported that ascorbic acid is more readily oxidized when the exocarp is bruised or broken. As has been shown with tomatoes (43) the use of growth regulator sprays did not alter the ascorbic acid content of Acerola (Table XI).

Since PCA was most effective for fruit set, further studies with derivatives of the phenoxyacetic acid series and the sodium salt form of PCA were carried out. Muir, et al (35) using growth in the AVENA straight-growth test as an index of relative activity of some of the chlorine substituted phenoxyacetic acids, reported that by virtue of change in the number and the positions of the chlorine atoms on the phenoxy ring, activity of the compound was drastically changed. When 6 compounds of the phenoxyacetic series were used at 100 ppm in inducing fruit set and on leaves of recent flushes, the relative trend of activity was related to the position of and/or number of chlorine

TABLE XI. ASCORBIC ACID CONTENT (MG PER 100 GMS) OF FRUITS OF ACEROLA FROM OPEN POLLINATION AND INDUCED WITH PARACHLOROPHENOXYACETIC ACID (100 PPM)

Plant	Open Pollination (Mgs. per 100 gms)	Parachlorophenoxyacetic acid (Mgs. per 100 gms)
R3T-1	1998	1795
R4T-2	2038	1961
R4T-4	1989	2073
R4T-6	1936	1980
R4T-8	2193	2138
4-14	2156	2046
8-11	1894	1771
12B-15	1510	1610
13-26	2195	2210
14-22	2276	2028
20-46	1635	1710
21-26	1730	1672
23-33	1908	2064
25-14	1512	1725

atoms on the chlorinated phenoxy ring (Table XII and Fig. 3). Phenoxyacetic acid and 2,6-Dichlorophenoxyacetic acid were ineffective in inducing fruit set or leaf curling. On the other hand, although 0-Chlorophenoxyacetic acid was ineffective in inducing fruit set, slight leaf curling resulted. With changes in the position of, and the addition of 1 or 2 chlorine atoms to the ring structure, a tremendous increase in activity, both in fruit set and leaf curling resulted. Of the three remaining compounds, PCA, 2,4-Dichlorophenoxyacetic acid and 2,4,5-Trichlorophenoxyacetic acid, the latter compound was least active. There were no noticeable differences in fruit set and in leaf curling between PCA and 2,4-Dichlorophenoxyacetic acid.

In a second study considering the acid and the sodium salt forms of PCA, opened flowers were treated with the following concentrations: 10, 50, 100, 500, 1000, and 5000 ppm.

Results (Table XIII) showed that the acid form gave consistently higher fruit set than did the salt form at like concentrations. At 100 ppm, maximum fruit set was obtained for both forms. The acid was significantly superior to the sodium salt. These results are in agreement with those reported by Grigg, et al (18); using 2,4,5-Trichlorophenoxypropionic acid and the alkaline amine salt on the fruit set of pears.

Visual observations of vegetative plant parts showed that leaf curling was present for both acid and the salt forms.

Studies with tomatoes (47, 56), fig (13) and pears (18) have shown that immature leaves appearing after application of growth regulators (PCA, 2,4-Dichlorophenoxyacetic acid and 2,4,5-Trichlorophenoxyacetic

TABLE XII. PERCENT FRUIT SET OF ACEROLA TREATED WITH GROWTH REGULATORS OF THE PHENOXYACETIC ACID SERIES (100 ppm)

Treatments	Number of Flowers	Number of Fruits Set	Percent Fruit Set	Confidence Intervals 95 Percent	Phyto-toxic Effects
Control	158	7	4.43	1.0-9.0	-
Phenoxy-acetic	162	5	3.08	1.0-8.0	-
O-Chlorophenoxy-acetic	153	3	1.96	0.0-7.0	+
P-Chlorophenoxy-acetic	147	106	72.10	63.0-80.0	++
2,4-Dichloro-phenoxyacetic	162	107	66.04	57.0-74.0	++
2,4,5-Trichloro-phenoxyacetic	179	83	46.36	37.0-55.0	+
2,6-Dichloro-phenoxyacetic	189	11	5.82	2.0-12.0	-

- Normal
 + Slight leaf curl
 ++ Heavy leaf curl



FIGURE 3. DEFORMATION OF LEAVES OF ACEROLA CAUSED BY APPLICATION OF GROWTH REGULATORS OF THE PHENOXYACETIC ACID SERIES. A. CONTROL, B. PHENOXYACETIC ACID, C. O-CHLOROPHENOXYACETIC ACID, D. P-CHLOROPHENOXYACETIC ACID, E. 2,4-DICHLOROPHENOXYACETIC ACID, F. 2,4,5-TRICHLOROPHENOXYACETIC ACID, AND G. 2,6-DICHLOROPHENOXYACETIC ACID.

TABLE XIII. PERCENT FRUIT SET OF ACHROLA TREATED WITH ACID AND SODIUM SALT FORMS OF PARACHLOROPHOXYACETIC ACID (MAUNAWILI CLONE).

Treatments (ppm)	Acid			Sodium Salt		
	pH	Percent Fruit Set	Confidence Intervals 95 Percent	pH	Percent Fruit Set	Confidence Intervals 95 Percent
0	5.6	3.33	0-13	5.6	3.33	0-13
10	5.1	35.00	25-52	5.7	50.00	40-67
50	4.5	58.00	50-76	5.8	53.33	45-70
100	4.0	91.66	81-97	5.8	63.33	55-79
500	3.4	85.00	72-92	5.9	60.00	51-74
1000	3.2	68.00	62-83	6.1	61.66	53-78
5000	2.8	35.00	25-52	6.2	13.33	7-27

acid) resulted in abnormalities with no effect on mature, fully expanded leaves. In Acerola, flowering and flushing occur at approximately the same time and application of PCA for fruit set has resulted in abnormal leaf conditions. Therefore, comparisons of IBA and PCA for induction of fruit set were made at the Puna orchard.

In this study PCA and IBA were both used at 100 ppm. Treatments consisted of control, PCA and IBA, in randomized blocks with three replicates. Each treatment plot consisted of 3, three-year-old trees of clone H-5.

When yields for all treatments were compiled and summarized, results showed that highly significant differences existed among all treatments (mean yields: control, 45 gm; IBA, 1402 gm and PCA, 2840 gm). In addition, differences in the mean weight of fruits were obtained (Table XIV). The inverse relationship between mean fruit weight and total yield was apparent in all treatments. This inverse relationship could be attributed to competition for plant nutrients among the developing fruits.

Receptivity of Different Stages of Blossom Development to Growth Regulator Sprays

The receptivity of the flowers over a wide range of blossom development determines to a great extent, the yield and the time interval which should be used between spray applications, if more than one is required. Roberts and Struckmeyer (45) and Leopold and Scott (28) in their work with tomatoes reported that auxin sprays were effective in inducing fruit set over a wide range of stages in blossom development: from small unopened buds up to the time that abscission of

TABLE XIV. ANALYSIS OF VARIANCE OF MEAN WEIGHT (GM) OF FRUITS OF ACEROLA (CLONE H-5) SET BY OPEN POLLINATION, INDOLE-BUTYRIC ACID (IBA) AND PARACHLOROPHENOXYACETIC ACID (PCA)

a) Analysis of Variance

<u>Source</u>	<u>df</u>	<u>ms</u>
Total	n 8	-
Replicates	2	0.06
Treatments	2	7.055**
Error	4	0.19

b) Results

Treatments	PCA	IBA	Control
Means	<u>2.30</u>	<u>3.62</u>	<u>5.46</u>

** Significant at 1 percent level.

The difference between any two means not underscored by the same line is significant.

flowers took place.

In preliminary spray studies of whole tree treatments with PCA, it was noticed that greater numbers of fruits were reaching maturity over longer periods than from open pollinated trees. When fruits from the sprayed trees were harvested over a period of several harvests and the carpels examined, it became apparent that fully formed endosperms were almost entirely lacking.

To establish definitely whether Acerola reacts to growth regulator sprays as do tomatoes, PCA at 100 ppm and 500 ppm was applied to different stages of blossom development. Flower buds 2 days and 1 day

before anthesis, flowers at anthesis and flowers 1 and 2 days after anthesis were used in this study. For treatments of flowers after anthesis, the peduncles of the flowers at the time of anthesis were marked, and the treatments applied as required.

Results (Table XV) when analyzed by use of confidence intervals showed that significant differences in fruit set were obtained with the different stages of blossom development and with the concentrations of the growth regulator.

Flowers at anthesis were significantly more receptive to PCA sprays than flower buds and flowers after anthesis, with no differences in response to 100 and 500 ppm treatments. Flower buds one day before anthesis and flowers 1 day after anthesis were receptive for both concentrations, with 500 ppm being more effective than 100 ppm. For flower buds 2 days before anthesis, 500 ppm treatment resulted in higher fruit set than at 100 ppm. These results indicate that flower buds and flowers before and after anthesis require greater stimulation than flowers at anthesis.

For flowers which received treatments 2 days after anthesis, it was observed that some of the flowers had already abscised. In the process of application of the treatments, abscission of additional flowers took place and no fruit set was achieved.

When fruit set by spray concentrations was analyzed, 500 ppm was superior to either 100 ppm or the control. However, under field practices, the increased fruit set resulting from use of the higher concentration would not warrant its use because of increased detrimental effects and greater possibilities of death to the vegetative

TABLE XV. RELATIVE EFFECTIVENESS OF GROWTH REGULATOR SPRAY (PCA) ON FRUIT SETTING OF ACEROLA AT DIFFERENT STAGES OF BLOSSOM DEVELOPMENT (MAURAWILI CLONE).

Treatments	Number of Fruits Set at the Following Conc. of Growth Regulator			Total Number of Flowers Treated	Total Fruits Set	Percent Fruit Set	Confidence Intervals 95 Percent
	Control	100 ppm	500 ppm				
-2 days	1	6	28	180	35	19.44	13-26
-1 day	2	25	34	180	61	33.88	27-42
Anthesis	4	52	51	180	107	51.66	44-60
1 day	0	24	31	180	55	30.55	22-39
2 days	0	0	0	180	0	0.00	0-3
<hr/>							
Total Number of Flowers Tested	300	300	300				
Total Number Fruits Set	7	107	144				
Percent Set Observed	2.33	35.66	48.00				
Confidence Intervals 95 Percent	1-5	30-42	42-54				

portions of the plants.

Results of this study indicate that maximum yields from each flowering cycle can be obtained with one application of growth regulator spray, provided the spray is applied during peak flowering.

Translocation of Growth Regulator

The possible use of growth regulators to induce fruit set in commercial enterprises has posed the question of relative rates of application per unit area. In addition to consideration of age of trees and number of trees per unit area, consideration must be made as to whether or not each and every flower must receive the stimulus from the growth regulator to induce fruit set.

To determine whether growth regulators applied to a flower was absorbed and translocated to another, two flowers from one inflorescence were designated as treated and untreated. To differentiate these two treatments, the peduncle of the treated flower was marked with 2 lines using India ink and the untreated marked with one. A third flower located on another branch was designated as the control.

The pronounced differences in fruit set between treated (76.30 percent) and untreated (6.25 percent) indicate that the growth regulator was not translocated to the untreated flower to effect fruit set. This was further substantiated by the daily rate of abscission of flowers between the control and the untreated flowers. For induction of fruit set, each flower must receive the stimulus from the growth regulator.

To determine the influence of absorption of growth regulator from the leaves and its subsequent translocation to effect fruit set,

all expanding and mature leaves on several tagged branches were swabbed with PCA (100 ppm) at the onset of flowering. Control was designated on a second series of flowering branches on the same tree. Then for the next 7 days, the peduncles of flowers that anthesised daily were marked.

Although swabbing of the leaves resulted in leaf curling to only expanding leaves, an insignificant decrease in percent fruit set (treated, 1.89 percent; control, 7.43 percent) was observed. These results indicate that the growth regulator when applied to the leaves may have been absorbed but did not effect fruit set.

Effect of Growth Regulator Spray on Flowering Habits

In many horticultural crops such as apples and pears, growers have been plagued with the phenomenon of alternate bearing caused by the depletion of carbohydrates in the plants in one year when yields are high, followed by low or no yield the next year or two. To alleviate this undesirable fruiting habit, growth regulators have been used with some degree of success to thin flowers and fruits during years of expected high yields. As has been shown, heavy fruit yields have been realized from trees sprayed with PCA, but its effect on subsequent flowering and fruiting habits is as yet unknown.

In this study, approximately 3-year-old trees of clone 13½ at the Puma orchard were used. Each treatment plot consisted of 3 trees, with 5 replicates. Treatments consisted of spraying different trees with PCA (100 ppm) at one flowering cycle and also at two successive flowering cycles.

Since it was impossible to establish to any degree of accuracy

the number of flowers that would appear on flowering cycles after treatment with growth regulator sprays, yields were used as an indication. Therefore, all fruits harvested during the period of this study (June 26 to Sept. 25, 1959) were recorded on the basis of individual trees and in the analysis of data, a single mean from each treatment was derived for each harvest period.

Results in Table XVI indicate that highest yields were obtained from the treatment in which two successive flowering cycles were sprayed, followed by those sprayed at one flowering cycle. In addition it was found that the subsequent flowering cycles of those treated with growth regulator sprays were delayed considerably.

Normally, in Hawaii, Acerola flowering cycles occur approximately every 20 to 28 days, with peak flowering occurring 3 to 6 days after the initial flowering. With the high yields realized from sprays, a tremendous amount of carbohydrate is removed from the plant. This normally would not occur under open pollination, thus, a longer period for replenishment may be required to reach a level conducive to flowering. As a result, only flowering cycles immediately after those in which growth regulators were used to induce fruit set were delayed, the first by approximately 18 days and the second by approximately 25 days.

In the case where trees were sprayed once, the yield obtained from the second flowering cycle which was delayed, was comparable to the open pollinated trees. This may be an indication that for all practical purpose, the residual effects of the growth regulator sprays were no longer present. In addition to the low yield, trees in this treatment

TABLE XVI. FLOWERING HABITS AND YIELD OF ACEROLA (CLONE 13 $\frac{1}{2}$) AS INFLUENCED BY APPLICATIONS OF GROWTH REGULATOR SPRAYS (PCA AT 100 PPM) (JUNE 26 TO SEPT. 25, 1959).

Treat-ments	1st Flowering (Initiation of Study)	Mean Yield (gm)	2nd Flowering (Date)	No. of Days Flowering Delayed	Mean Yield (gm)	3rd Flowering (Date)	No. of Days Flowering Delayed	Mean Yield (gm)	4th Flowering (Date)	No. of Days Flowering Delayed	Mean Yield (gm)	5th Flowering (Date)	Total Mean Yield (gm)
Control	June 26	114.0	July 20	0	82.76	Aug. 11	0	96.92	Sept. 2	0	68.70	Sept. 25	362.38
Sprayed Once	June 26	2,617.92	Aug. 7	18	103.43	Sept. 2	0	94.98	Sept. 25				2,816.33
Sprayed Twice	June 26	2,786.96	Aug. 7	18	1,434.66	Sept. 27*	25						4,221.62

* Estimated.

reverted to normal flowering in the third cycle, with harvest of fruits during the period September 19 to 22, followed immediately by the fourth flowering cycle.

The additional spray given on August 7 at the second flowering cycle to some trees resulted in yields which were less than those of the first spray. This is an indication of reduced flowering during the second cycle. In addition, the third flowering cycle appeared approximately 49 days after the second, a delay of 25 days from normal flowering.

These results indicate that utilisation of growth regulator sprays have definite benefits. The larger yields from fewer harvests result in less expenditure of time with an increase in efficiency. On the other hand, modification of horticultural practices such as application of additional fertilizer and study of the effects of prolonged application of sprays to the plants appear necessary.

Effect of PCA Concentrations on Flowering and Fruiting Behavior

Spraying for fruit set with growth regulator has resulted in high yields and altered the normal flowering cycles. Although spraying resulted in high yields, the mean weight of fruits decreased. Using the present method of hand picking, more time is required to harvest a given volume of the smaller fruits. Furthermore, use of PCA resulted in pronounced leaf deformation and in severe cases cessation of growth and yellowing of the terminals. These abnormal conditions would undoubtedly have adverse effects on future yields since flowering takes place on young stems. To study the effects of various concentrations of growth regulator sprays on these phenomena, PCA was used at concen-

treatments of 25, 50, 75 and 100 ppm for the period of one flowering cycle. Treatment plots consisted of 3 trees per plot in randomized blocks and replicated 3 times, using trees of clone 13 $\frac{1}{2}$ at the Puna orchard.

Summary of the data by analysis of variance showed that there were significant differences at the 1 percent level for yields (Table XVII) and mean weight of fruits (Table XVIII). Yields from 100 ppm treatment were significantly higher than those of any other treatment. In addition, yields from treatments with 50 and 75 ppm were significantly higher than those of the control and 25 ppm treatment. There were no differences between 50 and 75 ppm treatments and between the control and 25 ppm treatment.

TABLE XVII. VARIANCE ANALYSIS OF YIELD (GRAMS) OF ACEROLA SPRAYED WITH FIVE CONCENTRATIONS OF PARACHLOROPHOENOXY-ACETIC ACID (PCA).

a) Analysis of Variance

<u>Source</u>	<u>df</u>	<u>ms</u>
Total	14	-
Treatments	4	8,012,408.13**
Replicates	2	773,726.58
Error	8	245,569.36

b) Results

Treatments	Control	25 ppm	50 ppm	75 ppm	100 ppm
Means	<u>141.0</u>	<u>751.2</u>	<u>2910.0</u>	<u>2845.8</u>	<u>4055.1</u>

** Significant at 1 percent level.

The differences between any means not underscored by the same lines are significant.

TABLE XVIII. VARIANCE ANALYSIS OF MEAN FRUIT WEIGHT (GRAMS) OF ACKROLA (CLONE 134) SPRAYED WITH FIVE CONCENTRATIONS OF PARACHLOROPHENOXYACETIC ACID (PCA)

a) Analysis of Variance

<u>Source</u>	<u>df</u>	<u>ms</u>
Total	14	-
Replicates	2	0.37
Treatments	4	2.37**
Error	8	0.06

b) Results

Treatments	75 ppm	100 ppm	50 ppm	25 ppm	Control
Means	<u>2.33</u>	<u>2.60</u>	<u>2.70</u>	3.17	<u>4.57</u>

** Significant at 1 percent level.

Differences between any means not underscored by the same lines are significant.

With increased yield through the use of higher concentrations of growth regulator, decrease in the mean weight of fruits was observed (Fig. 4). However the highly significant differences in yield at 100 ppm treatment when compared with those set at 50 and 75 ppm treatments, did not exhibit an inverse relationship between yield and fruit size. Among these three levels (50, 75 and 100 ppm) there were no significant differences in the mean weight of fruits. These results suggest that the ability of PCA to induce fruit set is quantitative.

In addition to the differences in yield and fruit size, it was

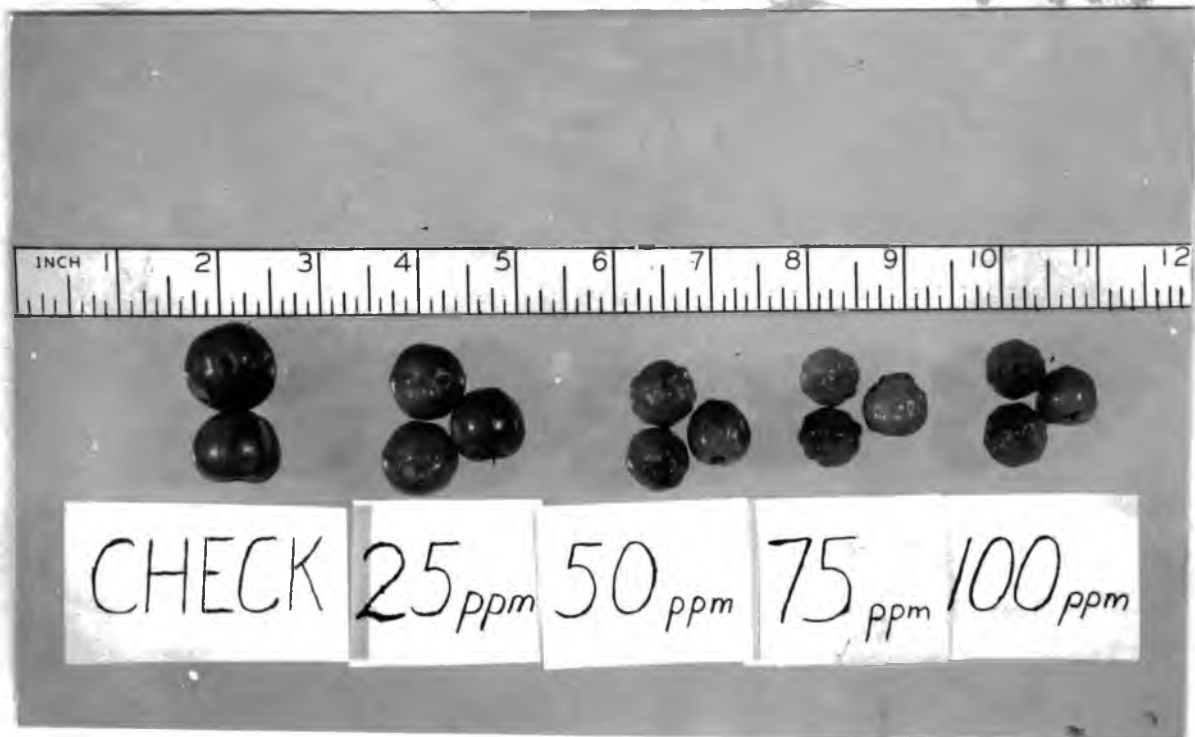


FIGURE 4. FRUITS OF ACEROLA (GLOBE 13½) SET WITH
PCA AT FIVE CONCENTRATIONS.

observed that the flowering habits and the period required for leaves to return to normal were related to the concentration of the growth regulator used. At the lower concentrations, the degree of leaf curling was less pronounced than at 75 and 100 ppm. Twenty-four days after application of sprays, only a few leaves from trees sprayed with 25 and 50 ppm were slightly curled, whereas those sprayed with 75 and 100 ppm were severely curled. Further observations made 17 days later showed that leaves of all sprayed trees had recovered, except those sprayed with 100 ppm which showed slight leaf curling.

The flowering cycles which appeared subsequent to application of sprays were also affected. Trees of the control and those treated with 25 ppm reverted to the normal flowering cycle, with moderate to heavy flowering 4 to 6 days after harvest. During this period, trees treated with 50 ppm produced scattered flowers only on branches that had not flowered during the previous cycle when sprays were applied. In addition, trees sprayed with 75 and 100 ppm were void of flowers for this period and heavy flowering did not occur until 28 days after harvest when all treatments were again in peak flowering.

DISCUSSION

The profound effects of environment on the behavior of plant life are well known. In Hawaii the yield of Acerola obtained from open pollination is one of these behaviors so affected. The disparity in yields of selected clones grown in Puerto Rico and Florida when compared to those grown in Hawaii has been so great that serious economic consequences to prospective growers in Hawaii may result.

The absence of insect activity among trees in full bloom in Hawaii

and Ladin's (23) report that in Florida bees were attracted in large numbers to the flowers indicated that low yields in Hawaii are attributable to lack of pollen transferring agents. This has been substantiated by the results of this study.

The ability of Acerola to set fruits after emasculation (Table VIII) and the preponderance of unfertilized ovules present in mature fruits from open pollination (Table VII) indicated that setting of fruits parthenocarpically is an inherent character. Increased set obtained by hand pollination and the subsequent increase in fully formed endosperms were related to the dehiscence of anthers and to the methods of pollination. These results indicate (increase in fruit set by crossing over selfing) that Acerola is to a degree self incompatible.

The receptivity of the stigma and the dehiscence of anthers immediately after anthesis of flowers indicated that dichogamy was not present in Acerola. In the sweet types, Clone B-17 and Florida Sweet, although anther dehiscence was delayed until noon, the stigma was receptive as shown by the increased fruit set obtained from crossing with clone Maunswili.

All these factors, however favorable, would not increase fruit set unless stimulation of the ovary through pollination was achieved. The reports of Fitting (17), Gustafson (19,20,21), Thiman (52) and Muir (34) have shown that pollen contributed auxins directly and/or stimulation indirectly to induce fruit set. Observations of stigma of flowers left under conditions of open pollination showed that an average of 1.24 pollen grains per stigma were present. The small amount of pollen on the stigma, due to ineffectiveness of wind and

insects as pollen transferring agents suggests that the auxin may not have been supplied in sufficient quantity.

In tomatoes, when environmental conditions resulted in deficiency of good pollen and modification of the floral structure, fruit setting difficulties were encountered. Growth regulators in these cases applied either to flower clusters or to the whole plants have been used to good advantage. In addition to overcoming the fruit setting problem, additional fruit set, increased fruit size and hastened maturity (55) have resulted.

The inherent characteristic of Acerola to set fruits parthenocarpically and Gustafson's suggestion that induction of parthenocarpic fruit set was accomplished most easily in plants whose ovary contained high natural auxin content suggested the use of growth regulators to overcome the fruit set problem. Of the growth regulators tested on Acerola in this study, Indolebutyric acid (IBA) and Parachlorophenoxyacetic acid (PCA) were found effective. The latter gave superior results and was effective over a wider range of concentrations. Although PCA at 100 ppm produced maximum yield, it resulted in small, highly lobed fruits, pronounced leaf curling, yellowing and in some cases death of vegetative terminals. At lower concentrations, 50 and 75 ppm, yields (Table XVII) and abnormalities were correspondingly less than at 100 ppm with less time required for leaves to return to normal. Use of IBA did not result in these abnormal conditions; however yields were not as large. This indicates that if maximum yields are to be obtained with the minimum of abnormalities, use of

PCA should be confined to the lower concentrations, preferably to 50 ppm, since no difference in yield between 50 and 75 ppm was evident.

In addition to the high yields obtained by the use of growth regulator sprays, other beneficial effects resulted. Fruits were more solid and did not bruise as easily, presumably minimising the loss of ascorbic acid (42). The effectiveness of growth regulator over a wide range of stages in blossom development is also an important consideration. Maximum yields from one application can be obtained provided the spray is applied at peak flowering.

In field practices, use of growth regulators for fruit set would result in a large portion of the spray solution being applied to the leaves. Edgerton and Haessler (15) in their work with apples reported that absorption of Naphthaleneacetic acid and Naphthaleneacetamide was influenced by the addition of "surfactant," and increased with an increase in temperature. Crafts (12) in addition, has proposed the theory that the different ages of leaves react to different levels of acidity, with the younger leaves being more receptive to absorption. To be effective, once growth regulator has been absorbed by the leaves, translocation from the leaves to the stem and within the stem must take place in order for fruit set to take place. Mitchell and Brown (31) using 2,4-Dichlorophenoxyacetic acid on beans, reported that this compound was readily translocated only under certain conditions. Leaves in which the sugar content was low as a result of being exposed to low light intensity for prolonged periods did not translocate the growth regulator readily. In addition, CO₂ free air and very young

leaves were found not conducive to translocation.

Upon being translocated into the stem, growth regulators in order to be effective must be further translocated from within the stem to the flowers. Mitchell and Brown (31) and Mitchell and Hamner (32) have reported that only downward movement in the stem took place, whereas, Ennis and Boyd (16) stated that translocation, both upward and downward in the stem of bean was influenced by the concentration of growth regulator and by the addition of Carbowax (polyethylene glycols).

Swabbing of both young expanding leaves and mature leaves with PCA did not influence fruit set. Curling of young expanding leaves may have been the result of absorption but translocation of the growth regulator may not have taken place to influence fruit set as shown by the lower fruit set (1.89 percent) below the control (7.43 percent).

The altered flowering habits may also be considered as another beneficial effect. When growth regulator sprays are used, large yields are obtained and tremendous amount of carbohydrate may have been removed from the plant, which normally would not occur. Where two flowering cycles occur under open pollination, use of growth regulators have resulted in the elimination of one of these cycles. The progressive increase in delay of flowering when treatment was applied to two successive flowering cycles indicates that additional work should be initiated to study interrelationships of growth regulator-fertilizer-flowering and the effect of prolonged use of growth regulator on the plant. Until more information from these studies is obtained, caution should be exercised in the use of these compounds.

SUMMARY

1. Problems concerned with fruit set and the use of growth regulator compounds to overcome this limitation in Acerola are presented.
2. Low natural fruit set in Acerola is attributed to lack of pollen transferring agents.
3. Some degree of self incompatibility is indicated in Acerola.
4. Clonal differences in germination (in vitro) of pollen grains present.
5. Indolebutyric acid and Parachlorophenoxyacetic acid were affective in promoting fruit set.
6. Parachlorophenoxyacetic acid was superior to Indolebutyric acid in fruit setting of Acerola.
7. Use of growth regulators for fruit set resulted in an inverse relationship between yield and mean fruit weight.
8. Ascorbic acid content was not altered in fruits induced by the use of growth regulators.
9. Acid form of Parachlorophenoxyacetic acid was superior to the sodium salt of this acid for fruit setting.
10. Use of Parachlorophenoxyacetic acid for fruit setting of Acerola resulted in no clonal differences, when based on yield.
11. Normal flowering habits were altered with the use of growth regulator (Parachlorophenoxyacetic acid).

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