

CHEMICAL INDUCTION OF STAMINATE FLOWERS ON GYNOECIOUS CUCUMBER (CUCUMIS SATIVUS)

Thesis for the Degree of M. S.
MICHIGAN STATE UNIVERSITY

LaMoine Devon Anhder

1959

3 1293 01097 275

175.014

LIBRARY
Michigan State
University

PLACE IN RETURN BOX to remove this checkout from your record.

TO AVOID FINES return on or before date due.

MAY BE RECALLED with earlier due date if requested.

DATE DUE	DATE DUE	DATE DUE
L = 3 1 5000		

1/98 c:/CIRC/DateDue.p65-p.14

CHEMICAL INDUCTION OF STAMINATE FLOWERS ON GYNOECIOUS

CUCUMBER (Cucumis sativus)

Ву

LaMoine Devon Anhder

AN ABSTRACT

Submitted to the College of Agriculture, Michigan State University of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Department of Horticulture

1959

Suppression of staminate flowers on lines segregating gynoecious plants was not achieved with Maleic Hydrazide, Alpha-naphthaleneacetic acid or 2-3-5 rri-iodobenzoic acid.

Application of N-para-chlorophenylphthalamic acid to gynoecious plants did not alter their sex expression. Staminate flowers were induced on gynoecious plants with foliar applications of gibberellic acid (A-3). Concentrations varying from 100 ppm to 5,000 ppm were effective. Induced staminate flowers produced viable pollen and self and sib pollinations using this pollen resulted in gynoecious progenies.

CHEMICAL INDUCTION OF STAMINATE FLOWERS ON GYNOECIOUS

CUCUMBER (Cucumis sativus)

Ву

LaMoine Devon Anhder

A THESIS

Submitted to the College of Agriculture, Michigan State University of Agriculture and Applied Science in partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

Department of Horticulture

1959



ACKNOWLEDGEMENTS

The author wishes to express his sincere appreciation and thanks to Dr. C. E. Peterson for his advice and supervision in planning the investigation and in the preparation of this thesis.

The writer is grateful to Drs. S. H. Wittwer and J. C. Ballard for their advice and suggestions on the experiments.

Acknowledgement is expressed to Drs. S. Honma, F. C. Elliot, and D. J. DeZeeuw for their helpful review of the manuscript.

The writer is indebted to his wife for her encouragement and sacrifices made throughout these investigations.

TABLE OF CONTENTS

	Page
INTRODUCTION	1
REVIEW OF LITERATURE	2
MATERIALS AND METHODS	6
A. Breeding Lines	6
B. Chemical Treatments	7
C. Data Recorded	12
RESULTS	13
A. Staminate Flower Suppression	13
B. Staminate Flower Induction	13
DISCUSSION AND CONCLUSIONS	23
A. Staminate Flower Suppression	23
B. Staminate Flower Induction	23
SUMMARY,	27
LITERATURE CITED	28

INTRODUCTION

Standard pickling cucumber varieties are monoecious and the ratio of staminate to pistillate flowers is approximately 25 to 1. A recent cucumber introduction from Korea, PI 220860, produced a number of completely pistillate (gynoecious) plants as well as monoecious types. The incorporation of this gynoecious flowering habit into pickling cucumbers resulted in the development of lines with varying degrees of "femaleness", ranging from gynoecious to normal monoecious. The plant breeder faced two problems in using this new introduction in developing methods for the economical production of hybrid cucumber seed; 1) Staminate flower removal would still be necessary if lines with low staminate-pistillate flower ratio were used as female parents.

2) Gynoecious lines would eliminate the necessity for staminate flower removal, but these lines could not be maintained.

The principal objectives of this work were first, to determine if the relatively few staminate flowers produced on certain lines not homozygous for the gynoecious character could be completely suppressed by the use of growth regulating chemicals, and second, to determine if staminate flowers could be induced on gynoecious lines by the use of growth regulating chemicals.

REVIEW OF LITERATURE

The available literature on the flowering habit of plants in response to environment and growth regulating chemicals is so extensive that generally only material pertaining to <u>Cucurbitaceae</u> and more specifically <u>Cucumis</u> sativus has been considered in this review.

Studies of the nodal sequence of flower types in the normal monoecious cucumber by Currence (1) and Nitsch et al. (21) indicate that the type of flower which develops in a given leaf axil varies with the flower's position on the plant. A gradual shifting from staminate to pistillate flowers occurring as the plant ages.

Tiedjens (24), Edmond (2), Nitsch et al. (21), Ito and Saito (9, 10) found that the length of photoperiod exerts a modifying influence on flower type. Long days increased the number of staminate flowers, while a reduction of light increased pistillate flower formation and decreased the number of staminate flowers. Ito and Saito (9, 10) also observed that warm nights apparently intensified the effect of long days. The effect of long days and warm nights was proportional to the duration of the treatment. In the same series of experiments Ito et al. (5) state that by pinching the main stem, staminate flowers of the Japanese cucumber were transformed into bisexual flowers, pistillate flowers and into somewhat mal-

formed lateral branches. They concluded that since all cucumber flowers in the primordial stages bear both sets of sex organs, the sex expression shifts according to the physiological condition of the node at which the flowers are borne, responding variably to environmental conditions and the age of the plant.

Other factors influencing sex expression have also been reported. Tiedjens (24) found that maturing fruit exert an inhibitory effect on the production of pistillate flowers as well as on the development of the plant. Varietal differences in sex expression have been described by Edmond (2) and Kumazawa et al. (13). High nitrogen and low water favor staminate flower induction under long days and high night temperatures and low nitrogen resulted in earlier pistillate flower production according to Ito and Saito (12). They found the effects of nitrogen and water, however, were masked when the plants were maintained under short days and at low night temperatures.

Numerous growth regulating chemicals have been employed in the study of cucumber sex expression. Laibach and Kribben (14, 15, 16, 17, 18) applied alpha-naphthaleneacetic acid (NAA) in a lanolin paste to the petiole stub of the second leaf of cucumbers and obtained an increase in the number of pistillate flowers. Spraying of terminal growth, although less effective, also increased the number of pistillate flowers. NAA has been

reported by Ito and Kato (4) to produce pistillate flowers even under long day light conditions.

The effect of NAA on the sexuality of a dioecious plant, <u>Cannibus</u>

<u>sativa</u> (hemp) was determined by Heslop-Harrison (3). In genetically

"male" plants, applications of 0.5% NAA in a lanolin paste to leaves arising
at the third node, induced pistillate flowers at sites which would normally
be occupied by staminate flowers.

The use of indole-3-acetic acid by Laibach and Kribben (16, 17) induced pistillate flowering in cucumbers at concentrations higher than required for NAA. Applications of 2-3-5 tri-iodobenzoic acid (TIBA) alone or in combination with other growth regulators did not influence sex expression. Rehm (23) used TIBA on watermelon and observed that more pistillate flowers were produced, but staminate flowers were never fully suppressed. Ito and Saito (6) report that TIBA applied to Japanese cucumber slightly retarded pistillate flower formation and that 2-4-dichlorophenoxyacetic acid accelerates pistillate flower formation.

Wittwer and Hillyer (26) reported the suppression of staminate flower buds in acorn squash for three to four weeks in the greenhouse following treatment with maleic hydrazide. The most effective treatment was 250 ppm applied when the first true leaf was fully expanded and again when the fourth or fifth leaf had expanded.

Yabuta and Hayashi (27) noted that gibberellin increased the number of staminate flowers preceding the first pistillate flower in monoecious plants. Wittwer and Bukovac (25) found the effects of gibberellin on cucumbers paralleled those of long days and low nitrogen levels in delaying the formation of pistillate flowers. The effects of various concentrations of gibberellin upon root elongation of germinating seedlings of several plants, including the cucumber, have been reported by Wittwer and Bukovac (25).

Tiedjens (24) indicated that the control of sex expression is by some chromosome mechanism. A predominately pistillate cucumber developed by Kumazawa et al. (13) is not affected by day length and temperature and is additional evidence of genetic control of sex expression. Peterson and Ballard (22) recently developed completely gynoecious lines that are not influenced by day length and temperature. The inheritance of sex expression has not been fully determined.

MATERIALS AND METHODS

A. Breeding Lines

Seed of the various lines used in this experiment was obtained from the cucumber breeding project at Michigan State University. The following breeding lines with known flowering habits were used:

- 1) M. S. U. 131-2, an inbred line known to frequently produce gynoecious plants.
- 2) M. S. U. 129, a gynoecious inbred line which produces an occasional staminate flower.
- 3) M. S. U. 1123, a completely gynoecious F_1 line resulting from a cross between M. S. U. 129 and a predominantly pistillate Japanese pollen parent.
- 4) M.S.U. 713-1, 713-2, 713-2, and 713-21, homozygous gynoecious sister lines in which chemical induction of staminate flowers on otherwise completely gynoecious plants had been accomplished.
- 5) SMR-12 and SMR-18, standard monoecious varieties.

 All cucumbers used with the exception of the Japanese cucumber were black spined pickling types.

B. Chemical Treatments

1) Staminate flower suppression in 1958.

Suppression of staminate flower productions on MSU line 131-2 by foliar applications of growth regulating chemicals in aqueous solutions was investigated.

MSU 131-2 was planted on the Horticultural Farm June 24, 1958 in plots consisting of three replications of ten plants each. Maleic hydrazide was used at concentrations of 250, 500 and 750 ppm. Alpha-naph-thaleneacetic acid was used at concentrations of 100, 200 and 400 ppm with 0.1 percent wetting agent (Tween 20) added. 2-3-5-tri-iodobenzoic acid was dissolved in acetone and used at concentrations of 25, 50 and 200 ppm. Each concentration of the above chemicals was sprayed on the entire plant at various stages of growth. Thirty non-treated plants were rogued for staminate flowers at the ten node stage. Forty additional plants were used as controls.

2) Staminate flower induction 1958.

To induce staminate flowers on MSU 129, a gynoecious inbred line, the potassium salt of gibberellic acid in powdered form was used in aqueous solution at concentrations of 100 and 250 ppm with 0.1 percent wetting

Supplied by Merck and Company, Rahway, New Jersey as the potassium salt of gibberellin A₃ (gibberellic acid) commercially available under the trade name, Gibrel.

agent (Tween 20) added. N-para-chlorophenylphthalamic acid was dissolved in acetone and used in aqueous solution at concentrations of 10, 20 and 100 ppm. Each concentration was applied to plants at various stages of development in plots of five plants each replicated three times. Twenty-five plants were maintained as controls.

3) Staminate flower induction, greenhouse 1958.

Observations in the field in 1958 indicated the need for further investigations with gibberellic acid. A commercial emulsion of gibberellic acid was used. Plantings were made August 20, in 8-inch pots in the greenhouse. Potted plants of MSU line 1123 were randomized into six plots of three plants each, and replicated three times. In one treatment 250 ppm of gibberellic acid was applied at the unfolding of the first true leaf and again at the tenth true leaf. Additional treatments were four applications at each of four concentrations - 250, 500, 1000 and 1500 ppm made at weekly intervals beginning at the unfolding of the first true leaf. Three plants were left untreated until approximately 25 nodes could be identified for sex expression. After it had been determined that no staminate flowers had formed, these plants were sprayed three times at weekly intervals with 1500 ppm of gibberellin. Six plants of pickling cucumber variety SMR-18 and six plants of MSU 129 were sprayed four times with 250 ppm at weekly intervals beginning with the unfolding of the first true leaf.

4) Staminate flower induction 1959.

Gynoecious plants in breeding lines grown in the greenhouse during the winter of 1958-59 were chemically treated to induce staminate flowers. Self pollination of these plants resulted in the availability of homozygous gynoecious lines for field work in 1959. Four of these gynoecious lines and the monoecious varieties SMR-12 and SMR-18 were used in a series of field experiments conducted in 1959. The plants for these experiments were started in the greenhouse and transplanted to the field while in the cotyledon leaf stage. The aerial parts of plants to be treated were sprayed with aqueous solutions of either the emulsified or powdered form of the potassium salt of gibberellic acid (A-3). No wetting agents were used. Concentration of 1,000, 1,500, 2,000 and 5,000 ppm were used. The 5,000 ppm concentration was applied in one treatment after complete unfolding of the first true leaf. Each of the remaining concentrations was applied in two treatments. One treatment when the first true leaf unfolded and again seven days later, the other when the first true leaf unfolded and again fourteen days later.

The commercial emulsion was used in the treatment of MSU line 713-1 and variety SMR-12 to determine if gynoecious and monoecious cucumbers vary in their response to gibberellic acid. Solutions of 100 ppm were applied at weekly intervals for a total of three applications beginning with the unfolding

of the first true leaf. Solutions of 250, 500, 1000 ppm were applied at the first true leaf stage and again seven days later. A control plot was maintained for each of the two lines. Each plot contained five plants, and treatments were replicated three times.

A method for increasing a gynoecious line was investigated by growing MSU 713-5 in a 12 by 80 foot screen isolation cage provided with a small hive of bees. The plants were grown in two rows four feet apart with 12 inches between plants in each row. One row was maintained as a control and the other was divided into four duplicated plots of six plants each. A standard concentration of gibberellic acid at 1,500 ppm was used. The plants were sprayed at weekly intervals beginning just before the unfolding of the second true leaf. The plants in one plot received a single application, in another, two applications, in another three applications, and the final plot received a total of four applications. The bees were placed with the cage after the final spray application.

The possibility of producing staminate flowers on gynoecious line MSU 713-2 by germinating seeds in an aqueous solution containing the potassium salt of gibberellic acid was investigated. Thirty-five seeds were placed in each of five Petri dishes June 22, 1959 on four thicknesses of paper toweling. The toweling in four Petri dishes was soaked with 7 milliliters of

gibberellic acid at one of the following concentrations: 10 ppm, 100 ppm, 1,000 ppm or 10,000 ppm. The fifth Petri dish contained distilled water. The dishes were placed in a seed germinator at a temperature of 82°F.

Twenty-four hours later 15 germinating seedlings from each dish were transplanted into peat pots filled with soil. After 48 hours in the germinator another 15 seedlings from each dish were transplanted into peat pots.

Fifteen seeds of SMR-18 were placed in each of two Petri dishes, one a control, the other treated with 7 milliliters of gibberellic acid at 10,000 ppm. The seedlings were removed after 48 hours and transplanted into peat pots. On June 29, all transplanted seedlings were placed in the field in plots of five plants each, replicated three times.

Soil applications of gibberellic acid in an inert clay carrier were used on remnant plants of the gynoecious lines. Treatments were 44.0 and 88.0 micrograms of gibberellic acid per plant. To insure uniform availability, 35 pounds of soil were removed from an area approximately 12 inches square and 6 inches deep. The soil was thoroughly mixed with the gibberellin-clay formulation and replaced. A single transplant was then planted in the soil. Five plants were used for each concentration. Plants used as control were transplanted in undisturbed soil.

The final experiment consisted of single foliar applications of gibberellic acid on gynoecious plants at the ten node stage at concentrations of 2, 500,

5, 000, 10, 000 and 20, 000 ppm in an effort to establish the concentration of the commercial emulsion of gibberellic acid necessary to cause injury.

C. Data Recorded

The nodal sequence of staminate and pistillate flower development on the main stem of each individual plant was mapped in each experiment.

Nodes were classified as having no visible flowers present, staminate flowers present, pistillate flowers present, or in some cases as having both staminate and pistillate flowers, regardless of whether or not all of the flowers reached maturity. A record was kept of the number of staminate flowers opening each day in the isolation cage. This record began July 20 when the first staminate flower opened and continued through August 6.

RESULTS

A. Staminate Flower Suppression

MSU line 131-2 exhibited considerable heterozygosity as evidenced by the amount of segregation in the control plots. There was no observable difference in the sex expression of treated and untreated plants. TIBA at 50 and 100 ppm and more than one application of 25 ppm resulted in death of all terminal growth. The occurrence of several completely gynoecious lines in breeding plots, and the discovery that staminate flowers could be produced on genetically gynoecious plants, eliminated the need for artifically suppressing staminate flowers. This newly available genetic control of staminate flower production turned attention to improving methods of stimulating staminate flower production on gynoecious plants.

B. Staminate Flower Induction

1) Summer 1958

Plants of MSU 129 treated with N-para-chlorophenylphthalamic acid at the 100 ppm concentration gave evidence of mild burning of the leaves within two days after each application. No other differences in appearance or sex expression between any of the N-para-chlorophenylphthalamic acid treated plants and non-treated plants were observed.

Plants of MSU 129 treated with gibberellic acid responded for short

intervals after each application with rapid and erect terminal growth. A slight yellowing of the young leaves was observed. A more noticeable effect was an increase in the number of staminate flowers formed. Plants given three applications at 250 ppm produced almost ten times as many staminate flowers as the untreated plants. The observed results and a schedule of treatments for gibberellic acid are found in Table I.

2) Greenhouse 1958

The effects of gibberellic acid at 250, 500, 1,000 and 1,500 ppm on MSU line 1123 grown in the greenhouse during the fall of 1958 are shown in Table II. The number of nodes at which staminate flowers appeared increased with each increase in concentration of gibberellic acid. No staminate flowers were found on the control plants. Successful hand pollinations were made on plants in each of these treatments. Three plants treated at 1,500 ppm after identification of approximately 25 pistillate nodes, produced several staminate flowers at approximately the 45th node. Four applications at 25 ppm at weekly intervals on six plants of MSU line 129 produced staminate flowers on five plants. The same treatment on six plants of variety SMR 18 resulted in the earliest pistillate flower appearing at node 24. No control plants of SMR 18 or MSU 129 were maintained.

TABLE I

Staminate Flower Production on MSU Line 129 as Influenced by Foliar Applications of N- \underline{p} -chlorophenylphthalamic Acid and Gibberellic Acid in 1958 Field Planting

Concentrations	Total plants	Plants with one or more staminate nodes	Total nodes	Total staminate nodes	Total staminate flowers	Staminate flowers per 100 nodes
N-p-chlorophenyl-	-1-					
phthalamic acid	7	-		-	-	ć
A 01	19	7	530	T 4	1 C	7.0
O <u>mdd or</u>	20 18	0 4	330 467	0 9		1.5
	•	•	Ç L	ı	ι	
A	19	4	000	ი	n	
20 ppm B	21	6	541	10	14	2.6
O	17	7	482	7	6	1.9
¥	17	က	437	4	ıv	1.1
100 ppm B	16	9	453	7	8	1.8
O	18	9	527	9	7	1.3
Gibberellic acid						
A	16	4	438	4	2	1.1
100 ppm B	17	5	480	6	12	2.5
Ö	17	7	499	6	16	3.2
A	17	10	496	12	15	3.0
250 ppm B	16	12	426	20	23	5, 4*
S	17	13	518	31	64	12.4*
Control	83	17	2, 249	17	30	1.3

A - Treatments were applied at 1-2 true leaf stage; B - at 2-3 and 3-5 leaf stage; and C - 2-3, 3-5 and 11-13 leaf stage. *Significant at the .01 level when compared with control.

TABLE II

The Effect of Foliar Applications of Gibberellic Acid on Gynoecious Inbred Line MSU 1123 Grown in the Greenhouse

Treatment	Total plants	Plants with one or more stamin- ate nodes	Total nodes classified	Total nodes at which staminate flowers occurred	Nodes at which staminate flowers occurred per 100 nodes
Control	6	0	250	0	0.0
1 250 ppm	6	7	275	2	0.7
$250~\mathrm{ppm}^2$	6	4	282	8	2.8
500 ppm	6	7	290	16	5.5
1000 ppm	6	&	280	52	18.6
1500 ppm	6	6	280	62	22. 1

Applications were made at the 1-2 true leaf stage and at the 10-12 leaf stage.

 $^{^2}$ Applications of this and remaining higher concentrations were made beginning at the 1-2 true leaf stage and repeated at weekly intervals for a total of four applications.

3) Summer 1959

The results of the potassium salt of gibberellic acid at concentrations of 1,000, 1,500, 2,000 and 5,000 ppm on MSU line 713-21 in 1959 are recorded in Table III. One application at 5,000 ppm resulted in 9.5 staminate flowers per 100 nodes. Two applications of each of the other concentrations at a 14 day interval resulted in a higher number of staminate flowers per 100 nodes than two applications at a 7 day interval. Each increase in concentration resulted in a higher number of staminate flowers than lower concentrations applied at the same frequency. The effect ranged from 2.9 to 10.0 staminate flowers per 100 nodes. Plants showing malformed terminal growth of the main stem were observed in all treatments. Clusters of pistillate flowers and tendrils, large forked tendrils, or several long petioled leaves at the stem terminal resulted in a determinate type growth. These distortions were found only on the main stem and usually seven or eight nodes beyond the last staminate flower. Control plants showed very little distortion of this type.

Three applications of gibberellic acid applied to variety SMR-12 at a concentration of 100 ppm resulted in 5.5 more staminate flowers per 100 nodes than were observed on untreated plants (Table IV). The same treatment applied to MSU line 713-1 resulted in 0.7 more staminate flowers per 100 nodes than was observed on non-treated plants. Concen-

TABLE III

The Effect of Foliar Applications of Gibberellic Acid on Gynoecious Inbred Line MSU 713-21 Grown in the Field 1959

1 Treatment	Total plants	Plants with one or more stamin- ate nodes	Total nodes classified	Total nodes at which staminate flowers occurred	Total staminate flowers	Staminate flowers per 100 nodes
Control	15	0	417	0	0	0.0
1000 ppm A B	14 16	4 8	308 425	7 16	9 20	2.9
1500 ppm A B	13 16	7 13	283 406	10 27	15 34	5.3 4.
2000 ppm A B	12 15	6 12	· 267 359	13	15 39	5.6 10.9
5000 ppm A A B	15	10	337	23	32	9.5

B treatments were applied at the first true leaf stage and again 14 days later. A treatments were applied at the first true leaf stage and again 7 days later.

 $^{^2}_{
m A}$ single treatment was applied at the first true leaf stage.

TABLE IV

The Effect of Foliar Applications of Gibberellic Acid on Monoecious Variety SMR-12 and Gynoecious Line MSU 713-21 Grown in the Field 1959

Treatment	Variety	Total nodes classified	Nodes at which staminate flowers occurred per 100 nodes	Average increase in number of staminate nodes per 100 nodes
	A ¹ .	295	83.1	
Control	$^{\mathrm{B}^2}$	294	0.0	•
3	А	324	88.6	5,5
roo ppin	В	276	0.7	0.7
250 npm	А	300	89.7	9.9
	В	294	0.0	0.0
500 ppm	A	306	89.9	6. 8
	В	301	0.3	0.3
1000 ppm	V	331	93. 1	10.0
;	В	303	3,6	3,6

¹SMR₋12

²MSU 713-1

 3 Three applications of 100 ppm; two applications of each of the other three concentrations.

trations of 250, 500 and 1,000 ppm each applied twice resulted in a greater number of induced staminate flowers on the monoecious plants than on the gynoecious plants. A schedule of treatments and results are in Table IV.

Applications of emulsified gibberellic acid at 1,500 ppm to gynoecious line MSU 713-5 in an isolation cage resulted in production of sufficient pollen for seed increase. The total number of staminate flowers induced increased with each successive spray application. The sequence of nodes at which staminate flowers appeared followed a definite pattern. A single application resulted in staminate flowers at two or three consecutive nodes beginning at approximately node seven. Two single applications at a seven day interval produced two or three consecutive staminate nodes beginning at approximately node seven, and two or three more beginning at approximately node 13. Three and four single applications at a seven day interval produced a short series of staminate nodes beginning at approximately node seven, another beginning at approximately node 13 and one or two staminate nodes beginning at approximately node 22. Normal pistillate flowers occurred before and after each series.

In the isolation cage some of the fruit in the row of gynoecious control plants contained less than a full set of seed. This was not observed in the row of treated plants. It is estimated that 3 to 4 pounds of viable seed will be harvested from the cucumbers in this 12 by 80 foot cage. A schedule of treatments and results are shown in Table V.

TABLE V

The Effect of Foliar Applications of Gibberellic Acid on Gynoecious Inbred Line MSU 713-5 Grown in an Isolation Cage 1959

Treatment	Dates of application	Total plants	Average number of staminate flowers per plant 2	Average number of flowers per 100 nodes of main stem
Control	1	74	0.0	0.0
1500 ppm	June 27	12	2, 3	9.0
1500 ppm	June 27 July 4	11	11.7	45. 1
1500 ppm	June 27 July 4 July 11	6	16.1	66.2
1500 ppm	June 27 July 4 July 11 July 18	12	18.1	70.9

I On date of first application the second true leaf was beginning to unfold.

 $^{^2}$ Flowers were counted as they opened from June 20 through August 6 on laterals and the main stem.

Sex expression of gynoecious line MSU 713-2 was not altered in plants grown from seeds germinated in gibberellic acid at concentrations up to 10,000 ppm. All plants in each treatment remained gynoecious in the field up to August 6. No observable effect on sex expression of the normal monoecious variety of SMR-18 resulted from the germination of seed in gibberellic acid at 10,000 ppm. Lateral root and root hair development was suppressed and the hypocotyl was noticeably elongated in seedlings of both varieties at all concentrations. Mild distortion of early leaves on plants grown from seeds germinated for 48 hours in concentrations of 10,000 ppm were observed in the field. This distortion persisted in the affected leaves but subsequent growth was normal throughout the season.

Gynoecious plants transplanted into soil containing gibberellic acid produced no staminate flowers. Growth habit of these plants was not noticeably different from the controls.

A single foliar application of gibberellic acid emulsion to gynoecious plants at 5,000, 10,000 and 20,000 ppm resulted in burning of leaves and death of terminal growth. Death of the plant did not occur and new lateral growth appeared a short time later. No apparent injury resulted from the 2,500 ppm application.

DISCUSSION AND CONCLUSIONS

A. Staminate Flower Suppression

Suppression of staminate flowers on segregating line MSU 131-2 was not achieved by applications of maleic hydrazide, alpha-naphthaleneacetic acid or tri-iodobenzoic acid. Death of terminal growth resulted after two applications of TIBA at 25 ppm and a single application at 50 or 200 ppm. Growth habit and sex expression of plants treated with NAA and M. H. were not visibly different from non-treated plants.

Further investigations of the chemical suppression of staminate flowers were not conducted because of the discovery of completely gynoecious cucumber lines and the successful induction of staminate flowers on genetically gynoecious plants.

B. Staminate Flower Induction

Plants of MSU 129, a line known to produce an occasional staminate flower, were treated with N-para-chlorophenylphthalamic acid. The sex expression of treated plants was not observed to be different from the controls. Slight burning of the leaves resulted at a concentration of 100 ppm but the plants soon recovered.

Staminate flowers were induced on gynoecious cucumber plants by foliar applications of gibberellic acid at concentrations varying from 100 ppm

to 5,000 ppm. Pollen smears indicated that pollen from induced staminate flowers was fertile. Fruit resulting from sib and self pollinations matured normally, produced viable seed, and plants grown from this seed were gynoecious.

A wide range of environmental conditions has not altered the sex expression of gynoecious plants. This indicated that complete genetic control of gynoecious sex expression is possible and can be utilized in cucumber breeding and hybrid seed production.

Gynoecious plants crossed with normal monoecious types produce gynoecious F_1 progenies. The induction of staminate flowers on these gynoecious F_1 plants by the use of gibberellic acid enables production of F_2 populations and provides the means for a more complete study of inheritance of sex expression.

Hybrid cucumber seed using monoecious lines has not been feasible due to the amount of hand labor required to remove staminate flowers or to make hand pollinations. The use of gibberellic acid to induce staminate flowers and produce female parent inbred lines will have wide application in the future development of \mathbf{F}_1 hybrids. This technique can also be expected to find use in commercial hybrid seed production.

Gibberellic acid applied at the unfolding of the first true leaf in 1959 induced the first staminate flower at approximately the fifth node.

Germination of seeds in gibberellic acid for 48 hours at concentrations as high as 10,000 ppm produced no staminate flowers. It appears that the flower primordia are not sufficiently developed 48 hours after seed germination to be affected by gibberellic acid, or that root applications are less effective than foliar applications in altering sex expression of gynoecious plants.

The appearance of pistillate flowers at nodes between series of staminate nodes on plants sprayed at seven day intervals indicates the possibility of increasing the number of staminate flowers produced in a given length of time by spraying at shorter intervals. A more uniform continuous staminate flower production would produce a more abundant supply of pollen for the increase of gynoecious breeding lines.

The results obtained by spraying gibberellic acid on monoecious SMR-12 and gynoecious MSU 713-1 at the same concentrations, suggests that the amount of gibberellic acid required to produce a staminate flower at a node normally producing only pistillate flowers may be greater for gynoecious than for monoecious plants.

Staminate flower induction became increasingly difficult on gynoecious plants as they matured. Apparently once the plant has developed a number of pistillate flowers, clearly establishing the gynoecious habit, it is difficult to interrupt this pattern and induce staminate flowers. The observed results support the conclusions of Laibach and Kribben (18) who stated that

the formation of pistillate flowers requires a higher concentration of growth substances than is required for staminate flower formation. They indicated that growth substances must reach a certain minimum concentration for a pistillate flower to be formed. Formation of a pistillate flower depletes the accumulated supply of the "pistillate producing" growth substances and the plant reverts to staminate flower production. The cycle is repeated throughout the life of the plant. This would account for the normal monoecious flowering habit of cucumber plants. It would also support the theory that gibberellic acid has an inhibiting effect on the synthesis of a growth regulating substance produced in gynoecious plants, changing the natural balance in the plant rather than directly influencing flower differentiation. The delayed effect of gibberellic acid, its non-persistence and its apparent lesser influence on older plants substantiate this theory.

SUMMARY

The objectives of this work were to determine if staminate flowers produced on certain lines not completely gynoecious could be suppressed by growth regulating chemicals and to determine if staminate flowers could be induced on gynoecious plants by the use of growth regulating chemicals.

Complete suppression of staminate flowers by foliar applications of Maleic Hydrazide at 250, 500 and 750 ppm, Alpha-naphthaleneacetic acid at 100, 200 and 400 ppm, or 2-3-5-tri-iodobenzoic acid at 25, 50 and 200 ppm was not achieved.

The sex expression of gynoecious plants was not observed to be affected by N-para-chlorophenylphthalamic acid at concentrations of 10, 20 or 200 ppm.

Staminate flowers were induced on completely gynoecious plants by foliar applications of gibberellic acid at concentrations varying from 100 ppm to 5, 000 ppm. Fruit resulting from the use of pollen from induced staminate flowers produced normal viable seed. Plants grown from this seed were completely gynoecious.

Through the use of gibberellic acid on gynoecious plants in an isolation cage provided with bees, seed increase was accomplished.

The incorporation of gynoecious flowering habit into pickling cucumber lines and the use of gibberellic acid to induce staminate flowers on gynoecious plants will have important application in the production of hybrid cucumber seed.

LITERATURE CITED

- 1. Currence, T. M. 1932. Nodal sequence of flower type in cucumber. Proc. Amer. Soc. Hort. Sci. 29: 477-479.
- 2. Edmond, J. B. 1930. Seasonal variation in sex expression of certain cucumber varieties. Proc. Amer. Soc. Hort. Sci. 27: 329-332.
- 3. Heslop-Harrison, J. 1956. Auxin and sexuality in Cannibus sativa. Physiologia Plantarum 9: 588-597.
- 4. Ito, H., and T. Kato. 1953. Factors responsible for the sex expression of Japanese cucumber. Jour. Hort. Assoc. Japan 22: 138-144.
- 7. K. Hashimoto and T. Saito. 1954. Factors

 responsible for the sex expression of Japanese cucumber. II. Anatomical studies on the sex expression and the transformation of the cucumber flower induced by pinching. Jour. Hort. Assoc. Japan 23: 65-70.
- 6. Ito, H., and T. Saito. 1956. Factors responsible for the sex expression of Japanese cucumber. III. The role of auxin on the plant growth and sex expression (1). Jour. Hort. Assoc. Japan 25: 101-110.
- of Japanese cucumber. IV. The role of auxin on the plant growth and sex expression (2). Jour. Hort. Assoc. Japan 25: 141-151.
- of Japanese cucumber. V. Causal interpretation of the effects of pinching and growth substances on the transformation of the primordia of the staminate flower nodes. Jour. Hort. Assoc. Japan 25: 213-220.
- Japanese cucumber. VI. Effects of the day length and night temperature, unsuitable for the pistillate flower formation, artificially controlled during the various stages of the seedling development in the nursery bed. Jour. Hort. Assoc. Japan 26: 1-8.
- Japanese cucumber. VIL Effects of long day and high night temperature treatment applied for short duration at the various stages of seedling development on the sex expression of flowers. Jour. Hort. Assoc. Japan 26: 149-153.

11. Ito, H., and T. Saito. 1957. Factors responsible for the sex expression of Japanese cucumber. VIII. Effects of long day and high night temperature treatment of short duration, accompanied by the growth substance spray application, on the sex expression of cucumber. Jour. Hort. Assoc. Japan 26: 209-214. . 1958. Factors responsible for the sex expression 12. of Japanese cucumber. IX. Effects of nitrogen application and watering under the controlled day length and night temperature in the nursery bed. Jour. Hort. Assoc. Japan 27: 11-20. 13. Kumazawa, S., M. Matumoto, and K. Fujieda. 1955. Cucumber breeding. (1) Breeding of the cucumber bearing more fruits under the long day and high temperature condition. Jour. Hort. Assoc. Japan 24: 1-5. 14. Laibach, F. 1952. Wuchsstoff und Blutenbildung. Beitrage Biol. Pflanzen 29: 129-141. 15. _____, and F. S. Kribben. 1949. Der Einfluss von Wuchsstoff auf die Bildung Mannlicher und Weiflicher Bluten bei einer Monozischen Pflange (Cucumis sativus L.). Ber. Deutsch Bot. Ges. 62: 53-55. . 1950 a. Der Einfluss von Wuchsstoff auf die Blutenbildung der Gurke. Naturwissenschaften 37: 114-115. . 1950 b. Uber die Bedeutung der Bindolylessigsaure fur die Blutenbildung. Ber. Deutsch Bot. Ges. 63: 119-120. . 1951 c. Der Einfloss von Wuchsstoff auf das Geschlect der Bluten bei einer Monozischen Pflanze. Beitrage Biol. Pflanzen 28: 64-67. . 1951 d. Die Bedeutung des Wuchsstoff fur die Bildung und Geschlechtsbestimmung der Bluten. Beitrage Biol. 19. Pflanzen 28: 131-144.

20. Lang, Anton, James A. Sandoval, and Ahmed Bedri. 1957. Induction

960-964.

of bolting and flowering in <u>Hyoscyamus</u> and <u>Samolus</u> by a gibberellinlike material from a seed plant. Proc. Nat. Acad. Sci. (U.S.) 43:

- 21. Nitsch, J. P., E. B. Kurtz Jr., J. L. Liverman and F. W. Went. 1952. The development of sex expression in cucurbit flowers. Amer. Jour. Bot. 39: 32-43.
- 22. Peterson, C. E., and J. C. Ballard. 1958. Development of gynomonoecious cucumber lines and their use in hybrid seed production. Abst. of paper presented before the 54th Ann. Meeting of Amer. Soc. Hort. Sci., Univ. of Indiana. 1958.
- 23. Rehm, S. 1952. Male sterile plants by chemical treatment. Nature 170: 38-39.
- 24. Tiedjens, V. A. 1928. Sex ratios in cucumber flowers as affected by different conditions of soil and light. Jour. Agr. Res. 36: 720-746.
- 25. Wittwer, S. H., and M. J. Bukovac. 1957. Gibberellins and higher plants: X. Field observations with certain vegetable crops. Mich. State Univ. Agr. Exp. Sta. Quart. Bul. 46: 352-364.
- 26. _____, and I. G. Hillyer. 1954. Chemical induction of male sterility in cucurbits. Science 120: 893-894.
- 27. Yabuta, T., and T. Hayashi. 1938. Biochemistry of bakanae disease. (In Japanese). Jour. Agr. and Hort. 13: 21-25.

ROOM USE ONLY

200 6 1963 W ROUN 612 COM