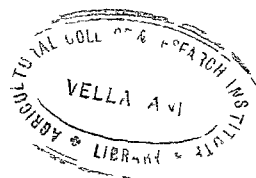


**STUDIES ON THE EFFECT
OF F. W-450 AND 2, 4-D AS MALE
GAMETOCIDES IN SESAME (*Sesamum indicum* L.)**



By

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THESIS

Submitted in partial fulfilment of the requirements for the award of the Degree of Master of Science in Agriculture (Agricultural Botany — Cytogenetics & Plant Breeding) of the University of Kerala

**DIVISION OF AGRICULTURAL BOTANY
AGRICULTURAL COLLEGE AND RESEARCH INSTITUTE
VELLAYANI, TRIVANDRUM**

1969



CERTIFICATE

This is to certify that the thesis herewith submitted contains the results of bonafide research work carried out by Shri N.P. Hariharan, under my supervision. No part of the work embodied in this thesis has been submitted earlier for the award of any degree.

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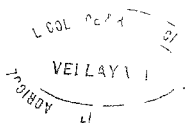
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N.P. HARIHARAN

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INTRODUCTION



INTRODUCTION

Sesamum indicum L. (Syn. S. Orientale L.) known variously as sesame, til, Sinsin, gingelly, gergelim, etc. belongs to the family Pedaliaceae. The generic name sesamum was taken from the Arabic "Samsin" by Hippocrates.

It is an important and very ancient oil yielding crop cultivated in India. Sesamum oil was one of the first, if not the first oil extracted from an oil seed by the ancient Hindus.

Intensive research is carried out in India for the improvement of this crop. One of the methods adopted is hybridization.

Manifestation of hybrid vigour or heterosis in development, earliness and yield of sesame has been reported by Riccoli and Mazani (1967) after their extensive studies with 32 varieties of sesame from different parts of the world.

Male-sterility is receiving increased attention as an aid to the production of large amount of hybrid seeds.

Gabelman (1956) used the term male-sterility to embrace a wide variety of abnormalities. He classified this term in to three categories:

- (1) Pollen sterility
- (2) Staminal sterility
- (3) Functional pollen sterility

A fairly large number of bisexual or monoecious plants show a specific hereditary constitution by which they produce little or no functional pollen whereas their ovular fertility remain completely or almost completely unimpaired.

Such natural male-sterile plants are effective females for a crossing programme. The employment of male-sterile lines makes the laborious procedure of hand emasculation superfluous. But the spontaneous occurrence of male-sterile lines is a factor beyond human control. Hence a search for methods of artificial induction of male-sterility was necessary for a more extensive utilisation of this phenomenon in breeding programme.

Many experiments have been conducted and it has been shown that certain chemicals can be used as male-gametocides. A male-gametocide is a substance which kills or make the male gametes functionless. There are

chemicals which affect both male and female gametes. Chemicals which show selective gametocidal action can only be used as a successful gametocide.

Stray reports are there on the occurrence of natural male-sterile lines in sesame (Beadle 1932, Sikka and Gupta (1947). But the occurrence was very rare and their isolation was unpredictable. Hence the experiment was undertaken to find out the methods of inducing male-sterility in sesame with F,W-450 and 2, 4-D. An account of the work is given in the following pages.

REVIEW OF LITERATURE

REVIEW OF LITERATURE



I. Chemical induction of male-sterility

Frank Eaton (1957) was the first to try a chemical as a gametocide. The experiment was carried out, with the chemical F,W-450, on cotton. Later several attempts have been made to induce male-sterility by treating plants with chemicals. This is a recent development in India but gaining momentum. The first report on the use of a chemical gametocide in India is in 1960, from the Agricultural Department, Gujarat, F,W-450 was the chemical and it was tried on cotton.

Some of the chemicals used for the induction of male-sterility and their application in the crop plants are reviewed.

(a) Sodium 2,3-dichloroisobutyrate (F,W-450 or Mendok)

F,W-450 is a chemical gametocide which when applied in optimum doses inhibit the normal development of gametes without imparting any deleterious effects to the plants as a whole. The chemical induces functional male-sterility in crop plants without adversely affecting female fertility.

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The chemical is readily soluble in water and is cent per cent active. Usually it is applied as a foliar spray and is easily absorbed by the plants. Studies with C¹⁴ labelled F,W-450 by Hilton (1958) showed that this chemical accumulates to a greater extent in anthers than in ovules. The flower characteristics like pubescence, thickness of cuticle and the stage of growth of the plant and other environmental factors are considered to be the entities which govern the absorption and translocation of the chemicals.

Different explanations are given for the mechanism of the chemicals in inducing male-sterility, but a satisfactory explanation is still to be sought in the fertile field of modern science.

Hilton (1958) gives a biochemical explanation for the effect of the chemical F,W-450. His experimental studies on cotton showed that F,W-450 is competent with pantoate an enzyme which synthesise pantothonate the absence of which causes sterility in plants. He states that the chemical accumulates to a greater extent in the anthers than in the ovules.

Kaul and Singh (1967), by their cytological and histological studies in Trigonella foenum-graecum

treated with F,W-450, concluded that upto the production of microspores the process was same in both treated and untreated plants. Thereafter the difference between treated and untreated plants became apparent soon after the young microspores were released from the mother cell wall.

In contrast to those from the untreated plants the young microspores from treated plants stopped growing. Instead they appeared to be subjected to some sort of mechanical pressure created by the enlarging tapetal cells as a result of which the contents of the microspores including the nuclei disintegrated. The importance of tapetal layer for the proper development of microspores has already been emphasised by a number of workers (Maheswari 1950). Evidence for this has accumulated from the histological and the cytological mechanisms of pollen degeneration in genic and cytoplasmic genic male-sterile lines of a number of crop plants (Zenkteler 1962, Dubey and Singh 1965, Joppa, McNeal and Welsh 1966). In all the cases the abnormal behaviour of the tapetal cells in one way or another was found to be associated with pollen abortion. The similarity in the mode of pollen abortion in chemically induced and genic male-sterile plants suggests that the

effects of the chemical may be same as those genetically controlled.

Dicotyledonous plants show greater response to F,W-450 than the monocotyledonous plants (Rhon and Hass Company 1960).

(1) Cotton

Cotton is the crop first received treatment with a chemical gametocide - F,W-450. The experiment has been carried out by Frank Eaton (1957) and he reported that there was delayed flowering due to the application of this chemical. The plants failed to produce pollen grains when they were sprayed with 1.2 per cent aqueous solution of F,W-450 but when hand-pollinated with pollen from control plants normal bolls with viable seeds developed.

Meyer et al (1958) reported that plants sprayed with F,W-450 produced negatively significant increase of growth over the checks owing to the death of apical meristem.

Bocanegra et al (1958) state that after treating the cotton variety Tanguis with one per cent solution, a reduction in size of the flowers, capsules and in the

number of viable seeds was observed. Four to six weeks after the treatment 90-100 per cent of the flowers contained a high proportion of sterile pollen. During the 3rd and 4th weeks some degree of female sterility was also observed. Hilton (1958) and Roux and Chirinian (1959) studied the effect of the chemical on cotton and reported that the chemical inhibited the enzymatic synthesis of pantothenate and accumulated to a greater extent in the anthers than there in the ovules. Investigations of Pate and Duncan (1960) on cotton revealed that all the concentrations used in their studies showed male and female sterility except 0.20 per cent which gave a selective sterility for male gametes.

Bhardwaj and Santhanam (1961) conducted an experiment at PILRCOM Centre, Coimbatore to study the effect of P,W-450 on cotton. A trial was laid out with K-6 (G. arboreum). The treatments comprised of 0.0, 0.1 and 0.2% concentrations of the gametocide spray at weekly and fortnightly intervals. The results indicated that 0.2% concentration of gametocide sprayed six times at weekly intervals, from the eleventh week after sowing induced cent per cent pollen sterility during the fifth and seventh weeks after the initial spray. However the gametocide did not appear to be highly selective and

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damage to female gametes was also encountered resulting in reduced seed set per boll. Seed sterility was normal in the control.

A preliminary study on the effect of F,W-450 as a selective gametocide was carried out by Avtar Singh and S.L. Soghal (1961) at the Regional PIRRCOM Centre, Sirsa. The treatment of Ph.Anc. cotton 320-F by the chemical resulted in a high degree of male-sterility which was at its peak in the third and fourth weeks from the date of application of the chemical. Marked female sterility was also induced by the chemical and the effect was more pronounced with frequent applications and higher concentration of the chemical.

Singh (1964) reported that this chemical is very toxic to cotton causing acute burning of leaves and shoot apices and reduction in size of flowers. He reported induction of male-sterility in cotton by the use of F,W-450.

Guru Swamy Raja (1967) conducted an experiment at the Reg. Res. Sta. Kovilpatti to study the phytogametocidal action of F,W-450 on the characters of M.C.U. 1 cotton. From this study a spraying schedule was proposed which induced and maintained a high level

of male-sterility with minimum adverse effects on other characters.

2. Tomato

To study the effect of F,W-450 on tomato Moore (1959) conducted trials under green house and field conditions. Green house studies on 146 varieties showed complete male-sterility for ten to thirteen days without causing female-sterility at concentrations of 0.075 and 0.15 per cent.

Field trials were conducted on eleven early varieties, with four concentrations (0.075, 0.15, 0.3 and 0.6 per cent) having three intervals each. The lower concentration 0.075 did not show any kind of pollen sterility. There was an increase in the percentage of pollen sterility as the concentration of the chemical increased. The concentration 0.15 per cent gave a high percentage of sterility for thirteen days with a slight reduction in female fertility. The high concentration 0.3 and 0.6 per cent almost prevented fruit set, when selfed. The highest concentration 0.6 per cent gave complete male-sterility for nineteen days beginning from twelve days after treatment. Pollen production was

normal, thirtyseven days after application.

Balakrishnar (1963) studied the effect of F,N-450 on tomato. Three varieties with three concentrations (0.15, 0.3 and 0.6 per cent) were tried. Spraying was done when flowers of the first truss were undergoing anthesis. Cent per cent sterility was noted with certain phytotoxic effect. Ovule fertility was normal as in the control.

(3) Watermelon

Henz and Mohr (1959) tested the effect of F,W-450 on watermelon. The initial spraying was done as the first pistillate blossoms appeared and spraying was repeated after six days. The result was that the mature female flower buds were seen unopened. The effect of F,W-450 in preventing the opening of the staminate buds appeared to be localised in the tip of the bud. Pinching of the tip permitted the immediate unfurling of petals. Anthers dehisced with production of apparently normal pollen inside the mature unopened staminal buds. No deleterious effects on female fertility were seen associated with the application of F,W-450.

Boswel (1960) conducted the same experiment on watermelon, with concentration of 0.12, 0.6, 2.0 and

2.5 per cent. All the concentrations caused phytotoxicity with male flowers being prevented from opening for a period of seven to ten days.

(4) Wheat

Chopra et al (1960) tried the chemical F,W-450 on wheat at I.A.R.I. But they failed to get any positive result. Wheat plants were sprayed once or twice with 0.25, 0.5, 0.75 and 1.0 per cent aqueous solutions of F,W-450. Plants sprayed with lower concentration were stunted and grass-like in appearance. Those plants which survived showed a delay in flowering. Small ear heads with shorter awns, tougher glumes and normal pollen grains were the result.

K.B. Porter and A.F. Wiese (1961) also tried this chemical on wheat.

The results obtained showed that this chemical is not well suited as a selective gametocide for wheat.

(5) Brinjal

Leelamma (1965) carried out an experiment to induce male-sterility in Brinjal. She reported complete absence of dehiscent anthers in the treated plants and such plants showed complete sterility. This complete

sterility lasted for a period of two weeks beginning from two weeks after treatment. There was a marked reduction in the percentage of fruit set on selfing and it was directly proportional to the concentration of the chemical. The size and weight of the selfed fruits were also found to decrease with increasing concentration. The seeds from selfed fruits showed a marked reduction in germination while the seeds from crossed fruits were found unaffected.

(6) Musk melon

Pundir, Nripendra and Singh (1935) recommended a multidirection foliar spray of aqueous F,W-450 for induction of male-sterility in the crop. The concentrations tried were 0.3%, 0.4%, 0.5% and 0.6%. The plants were severely damaged in case of high concentration and more number of sprayings. A concentration of 0.3 per cent as second spray showed functional male-sterility while 0.4 per cent concentration with two sprays showed the desired effect by inducing pollen sterility. Concentration of F,W-450 higher than 0.4 per cent showed deleterious effect.

(7) Onion

C.L. Kaul and S.P. Singh (1967) tried F,W-450 as a gametocide in Onion. The concentrations used were 0.01, 0.025, 0.05 and 0.1 per cent. The method of application was different from the usual. Equal volumes of the solutions of the chemical were injected at the base of the inflorescence bearing stalk when the inflorescence was a small protuberance.

The effect of the chemical on general growth of the plants was practically nil. The chemical also proved ineffective as a male gametocide as only a maximum of 30 to 40% sterility was obtained.

(8) Sunn hemp

Kaul and Singh (1967) conducted an experiment with F,W-450 on sunnhemp. The concentrations tried were 0.1%, 0.25% and 0.5%. Foliar sprays of the chemicals were made to run off.

Plants sprayed twice with 0.5% F,W-450 produced flowers with complete pollen sterility which lasted only for eleven days. The height of the treated plants were reduced. Flowers were smaller in size with the keel petals split partially or completely and the stigma exposed. Anthers of treated plants remained clumped

around the style in the form of a ring and did not dehisce. Fruit set was 35 to 50% of the control.

(9) Tobacco

Jose and Singh (1967) tried F,W-450 as a gametocide on Nicotiana rustica. The concentrations tried were 0.05%, 0.1% and 0.5%. The chemical was sprayed only once at the time of initiation of the main inflorescence. The experiments were carried out for two consecutive seasons. The concentration was later altered to 0.1%, 0.5% and 1%. Plants were sprayed twice at five day intervals. The treated plants did not show any effect at lower concentrations but the leaves became leathery. Flowering was simultaneous in almost all the treatments.

Two applications of the chemical at 1.0% made the pollen completely sterile in the apical region of the inflorescence for about one week. Three applications of the same concentration prolonged male-sterility at the apical region for two weeks and induced it totally in the middle region.

Of the various concentrations of F,W-450 only 1% had the significant effect in inducing high pollen sterility in different regions of the inflorescence.

(10) Bhindi

Duboy and Singh conducted an experiment on bhindi to study the gametocidal action of F,W-450. Pusa Sawani was the variety selected. The chemical was used at three concentrations 0.2%, 0.4% and 0.5%. Some plants were sprayed about a week prior to the initiation of the first floral buds and others were sprayed twice, the first prior to floral bud initiation and the second at the time of floral bud initiation.

Plant heights, time taken to flowering, total number of fruits per plant and number of seeds per fruit were recorded. Pollen sterility was checked by the acetocarmine method and ovular sterility judged by the percentage of fruit set and the number of seeds produced per fruit when the emasculated flowers of the treated plants were pollinated by the controls.

F,W-450 produced marked effects on the vegetative and floral parts of bhindi and is found to inhibit the height, number of leaves and the final yield. It was observed that any appreciable amount of male-sterility accomplished by drastic reduction in the percentage of bolls and the number of seeds formed. Complete male-sterility induced by 0.4 per cent F,W-450 was accompanied with 13% ovular sterility.

2, 4-D (2,4 - Dichlorophenoxy acetic acid)

2, 4-D was discovered independently in England and in the U.S.A. during the Second World War (Blackman 1945, Krans and Mitchel 1947, Nutman et al 1945, Slade et al 1945). It is mainly used as a weedicide. It can be used as a gametocide and more effectively for inducing parthenocarp and preventing fruit shedding in certain crops. But reports of its use as a successful gametocide are very few.

Rehm (1952) conducted an experiment to study the effect of 2, 4-D as a gametocide on plants of Hawksbury and Cape mountain Sweet water melons. The chemical was applied about a week prior to the opening of the first flowers. Full male-sterility was observed after one week when a dose of 5 ppm was applied. The pollen of the male-sterile plants showed various abnormalities. Pollen grains with vacuolated plasma and small size were observed. A high percentage of giant unreduced grains was seen. In sterile pollen these abnormalities amounted always to twenty per cent or more.

The female flowers were partly fertile and small sized fruits were produced. Fruit set was low. Lower concentrations did not produce male-sterility.

At concentrations of 50 and 100 ppm. the chemical was highly injurious to the plants and the effect lasted for weeks.

B. Choudhary et al (1960) sprayed cucumber plants with 5 ppm. and 10 ppm. of 2, 4-D. Female flowers were more in the treated plants. Crinkled leaves with etiolated veins and dark green intervienal areas appeared on the plants sprayed with 10 ppm. till eight to ten days after the second spray. The average final growth of the main axis was less than 2/3 of the average growth of the main axis in control plants.

Robert et al (1961) reported 2, 4-D as a pollenicide on grape. Dipping the flowering clusters in solutions of 2, 4-D increased the number of seedless berries. This was supposed to be due to the injurious effect of 2, 4-D to pollen germination or in other words due to its action as a pollenicide.

Choudhary and George (1964) obtained pollen sterility of 90-100%. lasting for 5-12 days in two varieties of brinjal by spraying the whole plant with 2, 4-D. Concentrations above 10 ppm. caused elongation, curling and cracking of tender parts of the stem.

Jose and S.P. Singh (1967) tried 2, 4-D as a gametocide on tobacco. It proved toxic at 0.5% concentration. All plants died after about ten days while at 0.05% linear growth was much retarded and leaves became smaller in size. Flowering was delayed. Male-sterility obtained was 59%. The ovaries contained less number of ovules in plants treated with 0.1% 2, 4-D. The results with Nicotiana rustica confirms that 2, 4-D is not a suitable gametocide.

Other chemicals

Ehrenberg et al (1956) studied the effect of ethoxy caffeine for inducing mutation and sterility in barley. These studies revealed that the chemical induced sterilities which corresponded to the effect of X-rays at 6000^r.

Nelson and Roseman (1958) tried the possibilities of chemical induction of male-sterility in maize by means of gibberellic acid (G.A) at concentrations of 500 ppm. and 1000 ppm. Foliar spray of the above concentrations, when the inflorescence was one inch in length, showed sterile or partially sterile tassels. Yermanos and Knowles (1960) produced male-sterile safflower plants with a foliar spray of G.A. at 100 ppm., in all the five varieties used.

Chopra et al (1961) studied the effect of uracil, thymine, yeast nucleic acid and TIBA on tomato and the above chemicals and Sodium nucleate on onion. Aqueous foliar sprays were given about a week prior to the opening of the first flower. All the chemicals failed to produce any kind of male-sterility in tomatoes. While in the case of onion cent per cent pollen sterility was noted in plants sprayed with TIBA at 10 to 100 ppm. Sodium nucleate at 4 per cent and thymine and uracil at 250 and 500 ppm. respectively.

Kaul and Singh (1967) conducted an experiment to study the effect of Maleic hydrazide (M.H) on cajanus cajan, using the concentrations of 0.1%, 0.5% and 1.0%. Burning of the shoot and leaf was noticed. All the treatments showed a significant decrease in plant height. A significant increase was observed in the number of branches and flowers. The size of the flowers was reduced in all treatments. But they failed to get complete male-sterility. All the treatments caused a significant decrease in the number of fruits per plant, number of seeds per fruit and total yield of grains per plant.

There are so many reports in the successful use of Maleic hydrazide as a male gametocide in various crops.

II. Pollen studies

(i) Pollen morphology

Pollen grains serve best in distinguishing between and showing relationship among the higher groups of plants of different families, genera and some times species.

Pollen grains are studied by several investigators. It consists of an intine and an exine.

The surface of the exine may be sculptured, smooth reticulate or smooth and with or without spines and it is a criterion by which plants of various classes and families can be identified. Wode House (1933), Lang (1937) and Erdtman (1954) studied the pollen morphology of Solanaceae.

(ii) Cytological studies on the sterility of pollen grains were carried out by two methods:

- (a) By staining
- (b) By germination in artificial media.

(a) Staining methods

Acetocarmine staining method was adopted for the study of pollen sterility, from a very early time.

Zirkle (1937) had described a method of testing the viability of pollen grains by aceto carmine staining method. Fresh unopened mature anthers were crushed in a drop of acetocarmine taken on a slide and the pollen grains were allowed to remain in the stain for fifteen minutes. By this time the grains got stained clearly. The pollen grains stained deeply and which looked plump and normal were counted as viable. Unstained and shrivelled were considered as non-viable or sterile.

Vietew (1952) used 2, 3, 5 - triphenyl tetra sodium chloride for testing the pollen viability of Zea mays. The best results were obtained when the test was carried out at 50°C using a two per cent solution. Oberle and Watson (1953) while utilising the above method on peach, apple, pear and grape pollens remarked that this method was ineffective for the determination of pollen viability in these crops. Jacopini (1954) recommended the treatment of pollen grains with two per cent of sodium biselenite for periods ranging from 1/2 to 2 hours, depending on the species for a rapid reliable means of determining pollen viability in stone and pome fruit trees. Grains with full germinative power turned pale yellow while non-viable grains did not change. King (1959) recommended a peroxidase-agar medium. Here the sterile pollen grains showed a blue colouration

while the viable pollen grains were colourless and swollen in appearance.

Ostapenko (1959) showed that the above mentioned staining methods would not give evidence for the sterility or viability testing of pollen grains. According to him these methods gave only a relative value in determining the percentage of pollen viability.

III. Germination in artificial media

A direct method of evaluating pollen viability is to find its actual germination in an artificial medium.

Kobel (1926) and Singh (1956) reported that Sucrose solution can be used as an artificial medium.

Schumacker (1935) and Thompson et al (1950) used boron to stimulate pollen germination in artificial media.

The medium used in this experiment was a combination of sucrose and boric acid, as an aqueous mixture.

MATERIALS AND METHODS

MATERIALS AND METHODS

The experiment was carried out in the Division of Agricultural Botany, Agricultural College and Research Institute, Vellayani, during the year, 1968-69. The object was to study the effects of the chemicals F,W-450 and 2, 4-D as male gametocides on gingelly (Sesamum indicum) Syn. Sesamum Orientale.

A. Materials

1. Seed material

Local variety "Onattukara black" tested for homogeneity for two generations was used as the seed material. Germination test was conducted and 98% germination could be obtained.

2. Chemicals

The chemicals used were Sodium 2,3-Dichloro isobutyrate (F,W-450 or Mondok) and 2, 4-Dichlorophenoxy acetic acid (2, 4-D). They were obtained from the Division of Agricultural Botany, Agricultural College and Research Institute, Vellayani.

Pilot studies were conducted to fix the dose of the chemicals. A maximum dose of 3100 ppm. was tried in case of F,W-450 and 200 ppm. in case of 2, 4-D. Treatment shock was observed in the plants but they revived within two days. Both the chemicals were applied ten days prior to the flowering of the plants. F,W-450 induced cent per cent pollen sterility but the shock

received by the plants was severe and so the dose was fixed to a lower concentration.

3. Treatment

(i) Spray application

- (a) F,W-450 at four concentrations
1500 ppm., 2000 ppm., 2500 ppm., and
3000 ppm.
- (b) 2, 4-D at four concentrations
50 ppm., 100 ppm., 150 ppm. and 200 ppm.
- (c) Distilled water

(ii) Stage of application

Three stages:

- (1) Ten days prior to flowering
- (2) At the time of flowering
- (3) Ten days after flowering

The following symbols were given for the treatments:

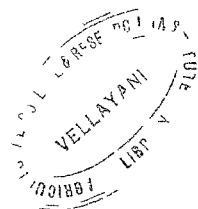
N	-	F,W-450
D	-	2, 4-D
L	-	Distilled water
C ₁	-	Control one
C ₂	-	Control two
N ₁	-	1500 ppm. F,W-450
N ₂	-	2000 ppm. F,W-450

- N₃ - 2500 ppm. F,W-450
- N₄ - 3000 ppm. F,W-450
- D₁ - 50 ppm. 2, 4-D
- D₂ - 100 ppm. 2, 4-D
- D₃ - 150 ppm. 2, 4-D
- D₄ - 200 ppm. 2, 4-D
- T₁ - First stage of application (Ten days prior to flowering)
- T₂ - Second stage of application (At the time of flowering)
- T₃ - Third stage of application (Ten days after flowering)

The different treatment combinations were the following:

- | | |
|------------------------------------|------------------------------------|
| (1) C ₁ | (12) N ₃ T ₂ |
| (2) C ₂ | (13) N ₄ T ₂ |
| (3) LT ₁ | (14) N ₁ T ₃ |
| (4) LT ₂ | (15) N ₂ T ₃ |
| (5) LT ₃ | (16) N ₃ T ₃ |
| (6) N ₁ T ₁ | (17) N ₄ T ₃ |
| (7) N ₂ T ₁ | (18) D ₁ T ₁ |
| (8) N ₃ T ₁ | (19) D ₂ T ₁ |
| (9) N ₄ T ₁ | (20) D ₃ T ₁ |
| (10) N ₁ T ₂ | (21) D ₄ T ₁ |
| (11) N ₂ T ₂ | (22) D ₁ T ₂ |

- (23) D_2T_2
 (24) D_3T_2
 (25) D_4T_2
 (26) D_1T_3
 (27) D_2T_3
 (28) D_3T_3
 (29) D_4T_3



B. Methods

1. Lay out

It was a pot culture experiment and the lay out was Completely Randomised Design. Total number of treatment was 29 and each one was replicated thrice. There were three plants under each treatment and the plants were equally spaced in a single pot. Of the three plants one was used for general observation and yield, one for floral and pollen studies and the remaining one for breeding work.

2. Preparation of the pot

Eighty seven pots of 9" x 12" size were filled with equal quantity of red earth, sand and dried cowdung (1:1:1). The pots were arranged with a spacing of 1 mt. x 1 mt. All the pots for raising plants for F₁W-450 treatment and 2, 4-D treatment were arranged separately in two groups. The pots for raising control plants and plants for treatment with distilled water were cautiously arranged

at a respectable distance from the other pots. The pots were marked with treatment symbols and then arranged.

3. Three shallow holes were taken at distances of 10 cm. within the pot and three healthy seeds were dibbled in each hole and were covered with a thin layer of soil. Thinning was done after ten days and three healthy seedlings were retained in each pot.

4. Spray treatment

Weighed quantities of the chemicals were first dissolved in one cc. of absolute alcohol each and the solution was prepared in distilled water. Spraying was done with a spray gun. The first spray was given 22 days after sowing and the same agreed with ten days prior to flowering - judged from the pilot studies. In all the cases spraying was done between 9 A.M. and 10 A.M; only after the desiccation of dew from the leaves.

5. Characters studied

I. Growth and morphological characteristics

(1) Visual observation

(2) Height of plant

(3) Number of flowers

(4) Pollen studies

(a) Pollen morphology

(i) Pollen shape

(ii) Pollen size

(b) Pollen sterility

(i) Acetocarmine staining method

(ii) Germinating pollen grains in artificial media

(iii) By crossing control plants with pollen from the treated plants.

(5) Ovular sterility

II. Mechanism of pollen sterility

Histological studies

III. Fruit set and fruit characters

Number of fruits per plant per treatment.

IV. Seed viability

I. Growth and morphological characters

1. Visual observations of the growth and development of plants under each treatment were made.

2. Height of plant

Only the final height of the plants were taken. Measurements were taken from the base to the tip of the plants and the data were analysed statistically.

3. Number of flowers

Opened flowers were daily counted and recorded. Flowering lasted for about one month. Total

number of the flowers was estimated and statistically analysed.

4. Pollen studies

(a) Pollen morphology

Studied ten days after treatment. Pollen grains were stained in acetocarmine and their shape and size were studied.

(i) Pollen shape

Pollen grains from freshly collected flowers were dusted in a drop of acetocarmine taken on a clean slide. Undehisced anthers were crushed with the blunt end of a mounted needle. The drop of acetocarmine with the pollen grains was covered with a cover slip and then observed under the low power of the microscope.

(ii) Pollen size

The diameter of the pollen grains was measured with an ocular micrometer. The ocular micrometer was standardised with the stage micrometer under the low power of the microscope. The size of 100 pollen grains taken at random from each treatment was measured and the mean diameter was calculated.

(b) Pollen sterility

(i) Acetocarmine staining method

Flowers were collected at intervals of two days, commencing from the fifth day of application of the

chemicals. This was continued till the restoration of male fertility as in the control. Fresh flowers were collected and kept in a desiccator for one hour and then the pollen grains were studied.

Staining and counting

Pollen grains were mounted in an acetocarmine glycerine medium. All the three replications for each treatment were studied. Pollens from each treatment were dusted in drops of acetocarmine taken on clean slides, separately. After 20 minutes the slides were observed under the low power of the microscope and sterile and fertile pollen grains were counted.

Well stained plumpy pollen grains were taken as fertile and unstained and shrivelled ones as sterile. Counts were taken for all the three replications at the rate of 15 microscopic fields for each replication. From these studies the mean percentage of pollen sterility for each treatment was calculated. The maximum sterility observed was on the 9th day after treatment and these data were analysed statistically.

(ii) Germination of pollen grains in artificial media

From the preliminary studies it was observed that medium containing sucrose and boric acid gave maximum

germination. Sucrose solutions of 15%, 20%, 30% and 40% were used in combination with 30 ppm., 50 ppm., 100 ppm. and 150 ppm. boric acid respectively. Tube elongations were also recorded. Small drops of sucrose boric acid solution were taken on clean sterile slides and pollen grains dusted in it. After a few minutes the slides were inverted and placed on glass rods in petri dishes. A piece of wet filter paper was placed at the bottom of each petri dish to provide a humid atmosphere and also an indirect stimulation to the germination of pollen grains. After 24 hours germinated and ungerminated pollen grains per treatment were counted.

A combination of 20% sucrose and 50 ppm. boric acid gave the best results.

(iii) By crossing the control plants with pollen from the treated plants

Five flowers were crossed in case of each treatment. Mature flower buds were emasculated on the previous evening of the opening day. Emasculation was done by gently pulling out the corolla along with the epipetalous stamens and then the flower was covered with a pollen proof paper bag. Pollination was done in the morning, before 7 A.M. Fresh flowers were collected in petri dish and the pollen grains were dusted on the stigma of the emasculated flowers. After pollination the emasculated flowers were covered and tagged.

The number of fruits per treatment was analysed statistically.

In all the cases maximum sterility was obtained on the 9th day after treatment. But in this case of crossing programme the availability of sufficient number of flowers within this limited time was not adequate. So the breeding programme was spread over a period of 14 days, after treatment.

(5) Ovular sterility

Tested by crossing treated plants with pollen from control. Five flowers were crossed in each case.

II. Mechanism of pollen abortion

Both L.S. of vegetative buds and C.S. of young anthers from control and treated plants were taken and studied under the microscope. Sections were taken with hand.

III. Fruit set and fruit characters

Numbers of fruits per plant in all the treatments and all the replications were taken and analysed.

IV. Seed viability

Viability was tested by conducting germination test.

100 seeds from each treatment were taken and sown in moist filter paper kept in a petri dish. Germination count was taken on the 4th day.

EXPERIMENTAL RESULTS

EXPERIMENTAL RESULTS

The various effects of F,W-450 and 2, 4-D on sesame were studied with special emphasis to their gametocidal action. The results are given below:

I. Growth and morphological characters

(1) Visual observations

Significant results were obtained with the application of the chemicals. All the doses except 3000 ppm. of F,W-450 showed gametocidal effects without any injury to the plants. Plants treated with the highest concentration showed treatment shock by marginal scorching of the leaves and general drooping of the plants, but they fully recovered in two days. The leaves and flowers of the treated plants were smaller in size when compared with those of the control. There was profuse and prolonged flowering in the case of all the F,W-450 treated plants. The effect of the chemical was pronounced at the first stage of application. The general growth and maturing of the F,W-450 treated plants were seen prolonged when compared with control.

All the concentrations of 2, 4-D were highly toxic to the plants. The damage was maximum at the first

stage of application and it was directly proportional to the increase in the concentration of the chemical. The plants were seen curled up and the leaves were thick and leathery. Gall formation was a characteristic feature in all the treated plants. All the plants were seen swollen considerably at the base immediately above the soil level. Shoot and leaves were hard and brittle and were dark green in colour particularly along the veins. Flowers and fruits were bigger in size and there was a tendency for conversion of the floral parts to vegetative parts. Style was persistent.

(2) Height of plants

The final heights of plants in each treatment are presented in Tables I and II.

The data obtained show that the chemicals as a whole were effective in regulating the linear growth of the plants. F,W-450 was very effective at all concentrations in increasing the height irrespective of the stages of application. The maximum height of 113 cm. was observed in plants treated with 2500 ppm. of F,W-450 at the second stage of application. The average height of the plants at the second stage of application was greater than those of the plants at the 3rd and 1st stages of application. Fig. I shows the graphical representation of the heights of plants in each treatment. All the plants except a few

TABLE I
Final height of plants (cm.) in each treatment*

Treatments	Rep.I	Rep.II	Rep.III	Mean	Stage III	Rep.I	Rep.II	Rep.III	Mean
Control-1	108	88	107	101.00	1500 ppm	99	103	101	101.00
Control-2	93	101	102	98.67	2000 ppm	91	111	99	100.30
Mean	<u>100.5</u>	<u>94.5</u>	<u>104.5</u>	<u>99.99</u>	2500 ppm	123	99	100	107.30
<u>Distilled water</u>					3000 ppm	102	115	113	110.00
Stage I	77	85	97	86.34	Mean	<u>103.75</u>	<u>107.00</u>	<u>103.25</u>	<u>104.66</u>
Stage II	91	100	96	95.67	<u>2, 4-D Stage I</u>				
Stage III	87	98	111	98.67	50 ppm	80	85	85	83.33
Mean	<u>85</u>	<u>94.34</u>	<u>101.34</u>	<u>93.56</u>	100 ppm	46	94	65	68.33
<u>F.W-450 Stage I</u>					150 ppm	46	32	65	47.66
1500 ppm	78	107	120	101.67	200 ppm	73	47	33	51.00
2000 ppm	104	103	102	103.00	Mean	<u>61.25</u>	<u>64.50</u>	<u>62.00</u>	<u>62.58</u>
2500 ppm	97	94	102	97.67	<u>Stage II</u>				
3000 ppm	88	95	108	97.00	50 ppm	58	64	70	64.00
Mean	<u>91.75</u>	<u>99.75</u>	<u>108.00</u>	<u>99.83</u>	100 ppm	91	91	97	93.00
<u>Stage II</u>					150 ppm	87	62	52	67.00
1500 ppm	86	93	104	94.33	200 ppm	51	79	49	59.66
2000 ppm	106	107	111	108.00	Mean	<u>71.75</u>	<u>74.00</u>	<u>67.00</u>	<u>70.91</u>
2500 ppm	117	110	112	113.00	<u>Stage III</u>				
3000 ppm	91	112	109	104.00	50 ppm	97	90	104	97.00
Mean	<u>100.00</u>	<u>105.50</u>	<u>109.00</u>	<u>104.83</u>	100 ppm	96	90	96	94.00
					150 ppm	84	91	93	89.33
					200 ppm	85	99	85	89.66
					Mean	<u>90.5</u>	<u>92.5</u>	<u>94.5</u>	<u>92.50</u>

* Fractions omitted

TABLE II
Analysis of variance for plant height - Final (Table I)

Source	SS	DF	Variance	F ratio
Total	33590	86	390	
Treatment	26286	28	938	7.50**
Between chemicals	13722	1	13722	109.77**
Between concentrations of F,W-450	234	3	78	0.62
Between concentrations of 2, 4-D	2459	3	819	6.55**
Between stages for F,W-450	194	2	97	0.77
Between stages for 2, 4-D	8268	2	4134	33.07**
Chemical vs. control	613	1	613	4.9*
Chemical vs. D. water	143	1	143	1.14
Error	7304	58	125	

C.D. for comparison between concentrations
of F,W-450 or of 2, 4-D = 10.52

C.D. for comparison between stages of
F,W-450 or of 2, 4-D = 9.00

** Significant at 5% and 1% levels.

* Significant at 5% level only.

at the first stage of application were, on an average higher than the control and distilled water treated plants. The difference between stages of application and between concentrations of F,W-450 were not significant.

2, 4-D was highly toxic to the plants. All the concentrations at all stages of application crippled the plants considerably. Table I shows that the mean height of plants in each treatment was less than that of the control plants. The injurious effect was maximum at the first stage of application and minimum at the 3rd stage.

(3) Number of flowers

The total number of flowers per plant in each treatment is furnished in Table III and its analysis of variance in Table IV.

The results show that the treatments were significant. The effect was highly significant between the chemicals.

F,W-450 was significant in increasing the number of flowers per plant. The mean number of flowers in all the stages of application and at all concentrations of the chemical was greater than that of control and distilled water treatments. There was significant difference between concentrations of F,W-450. Flower production was maximum in plants treated with 3000 ppm. followed by 2500 ppm.,

TABLE III

Total number of flowers per plant in each treatment

Treatments	Rep.I	Rep.II	Rep.III	Mean	Stage III	Rep.I	Rep.II	Rep.III	Mean
Control-1	97	49	91	79.00	1500 ppm	50	56	59	55.00
Control-2	108	65	72	81.66	2000 ppm	51	69	59	59.67
Mean	102.5	57.0	81.5	80.33	2500 ppm	75	65	176	105.34
<u>Distilled water</u>					3000 ppm	117	100	80	99.00
Stage I	96	95	86	92.33	Mean	73.25	72.25	93.50	79.67
Stage II	108	72	63	81.50	<u>2. 4-D Stage I</u>				
Stage III	82	47	63	64.00	50 ppm	65	42	45	50.67
Mean	95.33	71.33	70.66	79.11	100 ppm	30	37	31	32.67
<u>F.W-450 Stage I</u>					150 ppm	26	28	20	24.67
1500 ppm	103	128	65	90.66	200 ppm	5	6	7	6.00
2000 ppm	118	76	63	85.66	Mean	31.50	28.25	25.75	28.50
2500 ppm	92	106	66	88.00	<u>Stage II</u>				
3000 ppm	133	107	124	121.33	50 ppm	40	47	50	45.67
Mean	111.50	104.25	79.50	98.41	100 ppm	36	48	39	41.00
<u>Stage II</u>					150 ppm	22	20	27	23.00
1500 ppm	168	101	82	117.00	200 ppm	96	90	30	72.00
2000 ppm	55	70	82	69.00	Mean	48.50	51.25	36.50	45.42
2500 ppm	98	135	123	118.67	<u>Stage III</u>				
3000 ppm	186	105	138	143.00	50 ppm	58	57	60	58.34
Mean	127.00	103.00	106.00	112.00	100 ppm	50	59	40	49.67
					150 ppm	40	31	32	34.34
					200 ppm	15	5	6	8.67
					Mean	40.75	38.00	34.50	37.75

TABLE IV

Analysis of variance for total number of flowers per plant

Source	SS	DF	Variance	F ratio
Total	132575	86		
Treatment	103018	28	3679	7.22**
Between chemical	63665	1	63665	125.07**
Between concentrations of F,W-450	11961	3	3987	7.83**
Between concentrations of 2, 4-D	3491	3	1163	2.28
Between stages for F,W-450	6261	2	3130	6.14**
Between stages for 2, 4-D	722	2	361	0.70
Chemical vs. control	990	1	990	1.94
Chemical vs. D. water	1139	1	1139	2.23
Error	29557	58	509	

C.D. for comparison between concentrations
of F,W-450 and concentrations of 2, 4-D = 21.26

C.D. for comparison between stages of
F,W-450 and stages of 2, 4-D 18.42

** Significant at 5% level

1500 ppm. and 2000 ppm. respectively. Maximum number of flowers observed was 143 in plants treated with 3000 ppm. at the second stage. There was significant difference between stages of application of the chemical. The second stage of application averaged 112 flowers followed by 98 in the first stage and 80 in the third stage of application. Flowering was hastened by the application of F,W-450.

2, 4-D was not significant in increasing the number of flowers. The general average and the individual average of the 2, 4-D treatments were very low, when compared with F,W-450, control and distilled water treatments.

Treatment with F,W-450 was superior to all other treatments including control.

(4) Pollen studies

(a) Pollen morphology

(i) Pollen shape

In general the shape of the pollen grains was spherical, in control plants as well as in treated plants. But there were shrivelled or proliferated pollen grains in case of 2, 4-D treatments.

(ii) Pollen size

Table V gives the average of the pollen grains per treatment. The maximum diameter observed was 67.2 μ and the minimum 54.5 μ . The average size in control was

TABLE V
Size of pollen grains in μ

Treatments	Stages of application			Mean
	I	II	III	
Control-1	67.2	67.2	67.2	62.40
Control-2	57.6	57.6	57.6	
Distilled water	67.2	67.2	57.6	64.00
<u>F.N-450</u>				
1500 ppm	57.6	67.2	57.6	60.80
2000 ppm	57.6	57.6	57.6	57.60
2500 ppm	57.6	57.6	57.6	57.60
3000 ppm	57.6	57.6	57.6	57.60
Mean	57.6	60.0	57.6	56.72
<u>2. 4-D</u>				
50 ppm	54.5	54.5	67.2	58.74
100 ppm	67.2	57.6	57.6	60.80
150 ppm	54.5	56.5	57.6	56.20
200 ppm	54.5	54.5	54.5	54.50
Mean	57.92	55.77	59.22	57.55
General mean	61.50	50.74	59.84	60.15

62.4^μ and 64^μ in distilled water treated plants. Average size of F,W-450 treated pollen was 56.72^μ and that of 2, 4-D treated plants 57.55^μ. The general mean was 60.15^μ.

(b) Pollen sterility

(i) Acetocarmine staining method

Maximum sterility was observed on the 9th day after treatment in all the cases. The percentage of sterility on the 9th day alone was analysed statistically.

The data obtained are furnished in Table VI and its analysis of variance in Table VII.

The treatments were highly significant in inducing male-sterility. There was significant difference between the effects of the chemicals F,W-450 was superior to 2, 4-D for the concentrations selected.

The intensity of sterility was correlated with the concentration of the chemical and the stage of application. Significant difference was noticed between concentration of F,W-450. Maximum sterility was observed with 3000 ppm. and minimum with 1500 ppm. Thus the percentage of pollen sterility was directly proportional to the concentration. The first stage of application induced maximum sterility of 73% followed by 2nd with 67% and 3rd

TABLE VI

Percentage of pollen sterility by acetocarmine method - nine days after treatment

Treatments	Rep.I	Rep.II	Rep.III	Mean	Stage III	Rep.I	Rep.II	Rep.III	Mean
Control-1	12	18	20	16.67	1500 ppm	20	21	26	22.34
Control-2	11	15	18	14.67	2000 ppm	27	26	30	27.67
Mean	11.5	16.5	19.0	15.67	2500 ppm	31	35	36	34.00
<u>Distilled water</u>					3000 ppm	46	47	52	48.34
Stage I	12	15	8	11.67	Mean	31.00	32.25	36.00	33.00
Stage II	20	10	9	13.00	<u>2. 4-D Stage I</u>				
Stage III	9	10	8	9.00	50 ppm	38	40	41	39.67
Mean	13.67	11.67	8.34	11.22	100 ppm	50	57	60	55.67
<u>F.W-450 Stage I</u>					150 ppm	70	76	80	75.34
1500 ppm	60	65	56	60.34	200 ppm	100	100	100	100.00
2000 ppm	67	75	75	72.34	Mean	64.50	68.25	70.25	67.67
2500 ppm	70	75	79	71.34	<u>Stage II</u>				
3000 ppm	90	87	87	88.00	50 ppm	37	40	46	41.00
Mean	71.75	73.00	74.25	73.00	100 ppm	40	49	41	43.34
<u>Stage II</u>					150 ppm	50	57	60	55.67
1500 ppm	60	58	62	60.00	200 ppm	80	85	70	78.34
2000 ppm	61	66	70	65.67	Mean	51.75	57.75	54.25	54.58
2500 ppm	68	67	69	68.00	<u>Stage III</u>				
3000 ppm	70	76	73	73.00	50 ppm	30	29	25	28.00
Mean	64.75	66.75	68.50	66.67	100 ppm	31	30	36	32.34
					150 ppm	40	41	39	40.00
					200 ppm	57	31	36	41.34

TABLE VII

Analysis of variance for pollen sterility by acetocarrine
method nine days after treatment

Source	SS	DF	Variance	F ratio
Total	52210	86		
Treatment	51542	28	1840.7	84.43**
Between chemicals	455	1	455.0	20.87**
Between concentrations of F,W-450	2294	3	764.6	35.07**
Between concentrations of 2, 4-D	7116	3	2372.0	108.20**
Between stages for F,W-450	11045	2	5522.5	253.32**
Between stages for 2, 4-D	6314	2	3157.0	144.81**
Chemical vs. control	8593	1	8593.0	394.4**
Chemical vs. D. water	15380	1	15380.0	705.5
Error	1268	58	21.8	

C.D. for comparison between concentrations
of F,W-450 and between concentrations of
2, 4-D = 4.4

C.D. for comparison between stages for
F,W-450 and between stages for 2, 4-D = 3.8

** Significant at 5% and 1% levels.

stage with 83%. In inducing sterility all the concentrations of F,W-450 were superior to control and distilled water treatments.

With all its deleterious effects 2, 4-D was also significant in inducing pollen sterility. The percentage of sterility was directly proportional to the concentrations of the chemical. The range of sterility was from the degeneration of pollen grain to the complete inhibition of flower production. There was significant difference between stages of application. The first stage of application was effective in inducing the maximum sterility.

The flowers of the treated plants had half filled and completely empty anthers. In some total inhibition of anther production was also observed.

The effects of F,W-450 lasted upto 18 days after treatment and those of 2, 4-D were long lasting.

(ii) Sterility of pollen grains in artificial medium

The data are furnished in Tables VIII and IX.

The chemicals were highly significant in inducing pollen sterility. Significant difference was observed between chemicals. The general average of the 2, 4-D treatments was greater than that of the F,W-450 treatments.

TABLE VIII

Percentage of pollen sterility in artificial medium (20% sucrose + 50 ppm boric acid)

Treatments	Rep.I	Rep.II	Rep.III	Mean	Stage III	Rep.I	Rep.II	Rep III	Mean
Control-1	10	5	0	5.00	1500 ppm	30	31	35	32.00
Control-2	8	7	2	5.67	2000 ppm	24	32	27	27.67
Mean	9	6	1	5.34	2500 ppm	40	42	60	47.34
<u>Distilled water</u>					3000 ppm	54	52	55	53.67
Stage I	2	3	0	1.67	Mean	37.00	39.25	44.25	40.17
Stage II	12	13	20	15.00	<u>2. 4-D Stage I</u>				
Stage III	11	9	4	8.00	50 ppm	40	41	68	49.67
Mean	8.34	8.34	8.00	8.22	100 ppm	60	53	50	54.34
<u>F.W-450 Stage I</u>					150 ppm	72	64	59	65.00
1500 ppm	30	39	38	35.67	200 ppm	85	90	94	89.67
2000 ppm	50	48	54	50.67	Mean	64.25	62.00	67.75	63.92
2500 ppm	60	59	62	60.34	<u>Stage II</u>				
3000 ppm	80	82	79	80.34	50 ppm	40	38	39	39.00
Mean	55	57	58.25	56.75	100 ppm	54	42	51	49.00
<u>Stage II</u>					150 ppm	68	70	75	71.00
1500 ppm	24	25	30	26.34	200 ppm	99	97	96	97.34
2000 ppm	31	50	42	41.00	Mean	65.25	61.65	65.25	64.08
2500 ppm	44	52	50	48.67	<u>Stage III</u>				
3000 ppm	68	72	70	70.00	50 ppm	41	40	42	41.00
Mean	41.75	49.75	48.00	46.50	100 ppm	59	50	48	52.34
					150 ppm	70	64	62	65.34
					200 ppm	95	93	98	95.34
					Mean	66.25	64.25	62.50	63.50

TABLE IX
Analysis of variance for pollen sterility in
artificial medium

Source	SS	DF	Variance	F ratio
Total	59428	86		
Treatment	57760	28	2062.0	71.84**
Between chemicals	4709	1	4769.0	166.10**
Between concentrations of F,W-450	6859	3	2286.0	79.65**
Between concentrations of 2, 4-D	13452	3	4484.0	156.23**
Between stages for F,W-450	1681	2	840.0	29.26**
Between stages for 2, 4-D	8	2	4.0	0.14
Chemical vs. control	14186	1	14186.0	494.28**
Chemical vs. D. water	18219	1	18219.0	634.80**
Error	1668	58	28.7	

C.D. for comparison between concentrations
of F,W-450 or of 2, 4-D = 5.04

C.D. for comparison between stages for
F,W-450 or for 2, 4-D = 4.36

** Significant at 5% and 1% levels

Here also the intensity of sterility was directly proportional to the concentrations of the chemicals.

F,W-450 at concentration of 3000 ppm. gave an average of 68% sterility. Maximum sterility observed at this concentration was 80.34% at the first stage of application. The general average was 48%. Difference between stages of application was large.

Maximum sterility of 97.3% was observed with 2, 4-D at the highest concentration of 200 ppm. at the second stage of application but the effect of stages of application was not significant statistically.

In general the sterility observed in artificial medium was less than that obtained by acetocarmine staining method.

The length of pollen tubes, 24 hours after sowing in artificial medium is furnished in Table X. Maximum pollen tube growth was observed in treatment with distilled water (150.6^μ) and minimum (28.6^μ) in treatment with 2, 4-D. Tube growth in F,W-450 treated pollens was greater at lower concentration of 1500 ppm. The mean tube length being 145.94^μ and it was slightly greater than 145.5^μ the general average of control.

2, 4-D showed a retarding effect on pollen tube growth.

TABLE I
Length of pollen tubes in μ (after 24 hours)*

Treatments	Stages of application			Mean
	I	II	III	
Control-1	142.2	142.2	142.2	145.5
Control-2	148.8	148.8	148.8	
Distilled water	150.6	150.6	150.6	150.6
F, N-450				
1500 ppm	148.5	142.6	146.7	145.94
2000 ppm	140.5	138.6	100.7	126.6
2500 ppm	101.6	92.8	87.6	94.0
3000 ppm	72.6	73.8	70.9	74.1
Mean	115.8	113.2	101.5	110.16
2, 4-D				
50 ppm	65.6	70.2	58.9	64.9
100 ppm	60.3	63.7	56.8	60.3
150 ppm	43.2	47.8	40.4	43.8
200 ppm	38.5	28.6	30.7	32.6
Mean	51.9	52.6	46.7	50.4
General mean	121.86	121.5	117.96	114.16

*Pollen grains having tube length above 25 μ were counted as germinated.

In general the tube growth was inhibited with increased concentration of the chemicals. The graphical representation of pollen tube growth is given in Fig. V.

(iii) By crossing the control plants with pollen from treated plants

Tables VI, XII and XIII furnish the data. For comparison the control plants were selfed and the data were analysed along with those obtained in the case of the treated plants.

Fruit set was high in case of control and distilled water treatments thus showing the fertility of the pollen. On an average 96.5% and 95% of fruit set were obtained in control and distilled water treatments or the sterility was 3.5% and 5% respectively.

In case of F,W-450 maximum fruit set of 80% was observed at the lowest concentration of 1500 ppm. or the sterility in this case was only 20%. The minimum fruit set of 7% was observed in plants treated with 3000 ppm. at the first stage of application or the sterility obtained in this case was 93%. The general average of fruit set in F,W-450 treatments ranged from 22% upto 76% or the sterility observed was between 78% and 24%. The percentage of fruit set decreased with increase in concentration. Between stages the first stage of appli-

TABLE XI

FRUIT SET ON CROSSING CONTROL PLANTS WITH POLLEN FROM TREATED PLANTS*

Treatments	Rep. I	Rep. II	Rep. III	Mean	Stage I	Rep. I	Rep. II	Rep. III	Mean
Control-1	5	5	5	5.00	1500 ppm	4	5	3	4.00
Control-2	5	5	4	4.67	2000 ppm	3	4	4	3.67
Mean	5.00	5.00	4.50	4.84	2500 ppm	3	2	2	2.34
<u>Distilled water</u>					3000 ppm	2	1	2	1.67
Stage I	5	5	5	5.00	Mean	3.00	3.00	2.75	2.92
Stage II	5	5	4	4.67	<u>2, 4-D Stage I</u>				
Stage III	5	5	4	4.67	50 ppm	2	2	1	1.67
Mean	5.00	5.00	4.34	4.78	100 ppm	1	0	2	1.00
<u>F.W-450 Stage I</u>					150 ppm	0	0	1	0.34
1500 ppm	4	4	3	3.67	200 ppm	0	0	0	0.00
2000 ppm	4	3	3	3.34	Mean	0.75	0.50	1.00	0.75
2500 ppm	2	3	2	2.34	<u>Stage II</u>				
3000 ppm	1	0	0	0.34	50 ppm	2	2	1	1.34
Mean	2.75	2.50	2.00	2.42	100 ppm	1	1	2	1.34
<u>Stage II</u>					150 ppm	0	0	1	0.34
1500 ppm	4	3	4	3.67	200 ppm	0	0	0	0.00
2000 ppm	3	3	4	3.34	Mean	0.75	0.50	1.00	0.75
2500 ppm	2	3	2	2.34	<u>Stage III</u>				
3000 ppm	1	2	1	1.34	50 ppm	2	1	1	1.34
Mean	2.50	2.75	2.75	2.67	100 ppm	1	0	1	0.67
					150 ppm	0	0	2	0.67
					200 ppm	0	0	0	0.00
					Mean	0.75	0.25	1.00	0.67

* Five flowers were crossed in each treatment

TABLE XII

Analysis of variance for fruit set on crossing control plants with pollen from treated plants

Source	SS	DF	Variance	F ratio
Total	264	86		
Treatment	243	28	44	122.2**
Between chemicals	68	1	68	183.80**
Between concentrations of F,W-450	39	3	13	36.11**
Between concentrations of 2, 4-D	11	3	3.66	10.1**
Between stages for F,W-450	1	2	0.5	1.38
Between stages for 2, 4-D	1	2	0.5	1.38
Chemical vs. control	54	1	54	150.0**
Chemical vs. D. water	75	1	75	208.3**
Error	21	58	0.36	

C.D. for comparison between concentrations of F,W-450 or of 2, 4-D == 0.56

C.D. for comparison between stages for F,W-450 or for 2, 4-D == 0.48

** Significant at 5% and 1% levels.

TABLE XIII

Percentage of fruit set on crossing control plants
with pollen from treated plants

Treatments	Stages of application			Mean
	I	II	III	
Control-1	100	100	100	96.5
Control-2	100	100	80	
Distilled water	100	93	93	95
<u>F, W-450</u>				
1500 ppm	73	73	80	75.34
2000 ppm	67	67	73	69.00
2500 ppm	47	47	47	47.00
3000 ppm	7	27	33	22.34
Mean	48.5	53.5	58.25	53.42
<u>2, 4-D</u>				
50 ppm	33	27	27	29.00
100 ppm	20	27	14	20.34
150 ppm	7	7	14	9.34
200 ppm	0	0	0	0.00
Mean	15	15.25	13.75	14.67
General mean	72.7	72.35	71.8	64.89



cation gave an average of 48.5% fruit set or 51.5% sterility and it was the maximum between stages.

Percentage of fruit set was comparatively low in case of 2, 4-D treatments. The fruit set decreased with increased concentration of the chemical and it was zero in plants treated with 200 ppm. or cent per cent sterility as obtained at this concentration. Here also the first stage of application showed the least amount of fruit set claiming a high degree of pollen sterility.

In Fig. VI the percentage of fruit set is represented graphically.

(5) Ovular sterility

Ovular sterility was tested by crossing the emasculated flowers of the treated plants with pollen from control.

Tables XIV, XV and XVI furnish the data.

The fruit set, when compared with control and distilled water treatments was low in plants treated with the chemicals. Maximum fruit set was observed in case of F,W-450 or ovular sterility was less in this case. The average ovular sterility obtained in F,W-450 treatment was 38.81%. Maximum of 73% was obtained at concentration of 3000 ppm. At lower concentration of 1500 ppm. the sterility

TABLE XIV

Fruit set on crossing treated plants with pollen from control plants (Ovular sterility)*

Treatments	Rep. I	Rep. II	Rep. III	Mean	Stage III	Rep. I	Rep. II	Rep. III	Mean
Control-1	5	5	5	5.00	1500 ppm	5	4	4	4.34
Control-2	5	5	4	4.67	2000 ppm	4	4	3	3.67
Mean	5.00	5.00	4.50	4.84	2500 ppm	2	2	2	2.00
<u>Distilled water</u>					3000 ppm	1	2	1	1.34
Stage I	4	5	5	4.67	Mean	3.00	3.00	2.50	2.83
Stage II	5	4	5	4.67	<u>2. 4-D Stage I</u>				
Stage III	5	5	5	5.00	50 ppm	2	3	2	2.34
Mean	4.67	4.67	5.00	4.78	100 ppm	1	1	2	1.34
<u>F.W-450 Stage I</u>					150 ppm	0	0	0	0.00
1500 ppm	5	5	4	4.67	200 ppm	0	0	0	0.00
2000 ppm	4	4	5	4.34	Mean	0.75	1.00	1.00	0.92
2500 ppm	3	2	3	2.67	<u>Stage II</u>				
3000 ppm	2	1	1	1.34	50 ppm	1	2	3	2.00
Mean	3.50	3.00	3.25	3.25	100 ppm	2	0	1	1.00
<u>Stage II</u>					150 ppm	0	0	0	0.00
1500 ppm	5	4	4	4.34	200 ppm	0	0	0	0.00
2000 ppm	4	4	3	3.67	Mean	0.75	0.50	1.00	0.75
2500 ppm	3	3	3	3.00	<u>Stage III</u>				
3000 ppm	1	2	1	1.34	50 ppm	1	1	2	1.34
Mean	3.25	3.25	2.75	3.08	100 ppm	1	0	0	0.34
					150 ppm	0	0	0	0.00
					200 ppm	0	0	0	0.00
					Mean	0.50	0.25	0.50	0.42

* Five flowers were crossed in each treatment

TABLE XV

Analysis of variance for fruit set on crossing treated plants with pollen from control plants

Source	SS	DF	Variance	F ratio
Total	303	86		
Treatment	287	28	10.2	37.7**
Between chemicals	100	1	100	370.3**
Between concentrations of F,W-450	52	3	17.3	64.07**
Between concentrations of 2, 4-D	22	3	7.3	27.03**
Between stages for F,W-450	1	2	0.5	1.85
Between stages for 2, 4-D	2	2	1	3.70*
Chemical vs. control	48	1	48	177.77**
Chemicals vs. D. water	37	1	37	137.03
Error	16	58	0.27	

C.D. for comparison between concentrations of F,W-450 or of 2, 4-D = 0.48

C.D. for comparison between stages for F,W-450 or for 2, 4-D = 0.42

** Significant at 5% and 1% levels

* Significant at 5% level only

TABLE XVI

Ovular sterility

Percentage of fruit set on crossing treated plants
with pollen from control plants*

Treatments	<u>Stages of application</u>			Mean
	I	II	III	
Control-1	100	100	100	96.5
Control-2	93	93	93	
Distilled water	93	93	100	95.0
<u>E, W-450</u>				
1500 ppm	93	87	37	89.00
2000 ppm	27	73	73	77.76
2500 ppm	53	60	40	51.00
3000 ppm	27	27	27	27.00
Mean	86.75	61.75	56.75	61.19
<u>2, 4-D</u>				
50 ppm	47	40	27	38.00
100 ppm	27	20	7	18.00
150 ppm	0	0	0	0.00
200 ppm	0	0	0	0.00
Mean	18.5	15.0	3.5	14.00

*Fractions omitted

observed was 11% on an average. The sterility in control and distilled water treatments were 3.5% and 5% respectively. Ovular fertility was affected by chemical treatment and the intensity was directly proportional to the concentration of the chemicals.

Ovular sterility was maximum in 2, 4-D treatments. At higher concentration of 150 ppm. and 200 ppm. there was no fruit set at all or the sterility was complete. Fruit set was maximum at lower concentration of 50 ppm. indicating lower percentage of sterility. Ovular sterility was less at the first stage of application.

In Fig. VIII the percentage of fruit set is represented graphically.

II. Mechanism of pollen abortion

The cross section of young anthers from treated plants showed collapsed as well as hypertrophied tapetal cells and the pollen grains were disintegrated in the middle of the anther cavity. Aborted pollen grains were seen in certain cases. The abortion was noted at the later stages of pollen grain formation.

Longitudinal sections of vegetative buds showed no symptom of damage in both treated and control plants.

III. Fruit set and fruit characters

The data showing the number of capsules in each treatment are furnished in Table XVII and its analysis of variance in Table XVIII.

In case of F,W-450 the treatments were highly significant in increasing the number of capsules per plant. The general average of capsules for the F,W-450 treatment was 82. There was significant variation between concentrations of the chemical. Maximum number of 132 capsules was in plants treated with 3000 ppm. at the second stage of application. The stages of application were also significant for F,W-450. On an average the maximum number of capsules was at the second stage of application (89) and it was followed by the 1st and 3rd stages. Treatment with F,W-450 was superior to control and distilled water treatments.

2, 4-D showed a depressing effect on the number of capsules per treatment and the general average was only 43. The range was between zero and 31. Treatment with 2, 4-D was insignificant.

The percentage of total fruit set per plant is furnished in Table XIX. Maximum fruit set was in plants treated with 3000 ppm. of F,W-450, with an average of 90-34% greater than 85% in control and 80% in distilled

TABLE XVII

Final number of pods per plant in each treatment

Treatments	Rep.I	Rep.II	Rep.III	Mean	Stage III	Rep.I	Rep.II	Rep.III	Mean
Control-1	95	48	41	61.34	1500 ppm	40	44	54	46.00
Control-2	102	64	62	76.00	2000 ppm	47	65	54	55.34
Mean	98.50	56.00	51.50	68.67	2500 ppm	70	59	40	56.34
<u>Distilled water</u>					3000 ppm	101	92	73	88.67
Stage I	84	63	78	75.00	Mean	64.50	65.00	55.25	61.59
Stage II	64	94	53	70.34	<u>2, 4-D Stage I</u>				
Stage III	63	39	53	51.67	50 ppm	21	18	24	21.00
Mean	70.34	65.34	61.34	65.67	100 ppm	10	12	14	12.00
<u>F.W-450 Stage I</u>					150 ppm	15	12	10	12.34
1500 ppm	52	93	120	88.34	200 ppm	0	0	0	0.00
2000 ppm	58	108	70	78.67	Mean	11.50	10.50	12.00	11.33
2500 ppm	78	54	91	74.34	<u>Stage II</u>				
3000 ppm	121	97	110	109.34	50 ppm	20	28	19	22.34
Mean	77.25	88.00	97.75	87.67	100 ppm	15	16	20	17.00
<u>Stage II</u>					150 ppm	15	13	12	13.34
1500 ppm	42	156	80	92.67	200 ppm	0	0	0	0.00
2000 ppm	50	64	50	54.67	Mean	12.50	14.25	12.75	13.17
2500 ppm	92	128	113	111.00	<u>Stage III</u>				
3000 ppm	172	99	125	132.00	50 ppm	40	26	25	30.34
Mean	89.00	111.75	92.00	97.56	100 ppm	21	26	31	26.00
					150 ppm	18	15	12	15.00
					200 ppm	2	3	2	2.34
					Mean	20.25	17.50	17.50	18.42

TABLE XVIII

Analysis of variance for number of pods per plant
in each treatment

Source	SS	DF	Variance	F ratio
Total	132908	86		
Treatment	115242	28	4116	13.51**
Between chemicals	83164	1	83164	273.11**
Between concentrations of F,W-450	10720	3	3573	11.73**
Between concentrations of 2, 4-D	2744	3	915	3.00*
Between stages for F,W-450	8299	2	4149.5	13.62**
Between stages for 2, 4-D	324	2	162	0.53
Chemical vs. control	2299	1	2299	7.55**
Chemical vs. D. water	2415	1	2415	7.93**
Error	17666	58	304.5	

C.D. for comparison between concentrations
of F,W-450 or of 2, 4-D = 16.44

C.D. for comparison between stages for
F,W-450 or for 2, 4-D = 14.24

** Significant at 5% and 1% levels.

* Significant at 5% level only.

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water treatments. Average percentage of fruit set in F,W-450 treatment was 84.83 and it was very close of 85% in control. Percentage of fruit set was maximum at the first stage of application, 88.75 and it was closely followed by the second stage of application with an average of 86 and 3rd stage with an average of 79.75.

The percentage of fruit set in 2, 4-D treatments was very low. Maximum of 58% was observed in treatment with 150 ppm. and a minimum of zero in treatment with 200 ppm. Usually there is only one flower in each axil and this is between two aborted flower buds called nectarial glands. But in case of the F,W-450 treated plants all the three buds developed. Thus three capsules were seen in each axil of the F,W-450 treated plants. The capsules were smaller in size when compared with those in control plants.

Capsules of the 2, 4-D treated plants were bigger in size. But in treatments with both the chemicals there were empty and half filled capsules and seeds. Seeds of plants treated with 200 ppm. of 2, 4-D were smaller in size.

TABLE XIX
 Percentage of total fruit set per treatment*

Treatments	<u>Stages of application</u>			Mean
	I	II	III	
Control-1	77	77	77	85.00
Control-2	93	93	93	
Distilled water	81	87	81	83.00
<u>F.W-450</u>				
1500 ppm	89	79	84	84.00
2000 ppm	92	79	93	88.00
2500 ppm	84	94	53	77.00
3000 ppm	90	92	89	90.34
Mean	88.75	86.00	79.75	84.83
<u>2, 4-D</u>				
50 ppm	41	49	52	47.34
100 ppm	37	41	52	43.34
150 ppm	50	58	44	50.67
200 ppm	00	00	27	9.00
Mean	32.00	37.00	43.75	37.59
General mean	74.35	76.00	74.90	73.56

* Fractions are omitted.

IV. Seed viability

The general germination percentage of seeds is furnished in Table XX.

Percentages of germination in control and distilled water treatments were 90.5 and 79 respectively. The average percentage of F,W-450 was 71.84 showing that the chemical affected the viability of the seeds. Maximum germination percentage of 78.25 on an average was observed at the second stage of application.

Percentage of germination in 2, 4-D treatment was very low.

TABLE XX
General germination percentage

Treatments	Stages of application			Mean
	I	II	III	
Control-1	96	96	96	90.50
Control-2	85	85	85	
Distilled water	90	73	74	79.00
<u>E.W-450</u>				
1500 ppm	73	82	87	80.67
2000 ppm	53	78	72	67.67
2500 ppm	69	73	79	73.67
3000 ppm	46	80	70	65.34
Mean	60.25	78.25	77.00	71.84
<u>Z. d-D</u>				
50 ppm	64	59	52	58.34
100 ppm	84	70	42	65.34
150 ppm	74	47	22	47.67
200 ppm	00	00	20	6.67
Mean	55.50	44.00	34.00	44.50
General mean	77.35	75.25	73.20	72.33

DISCUSSION

DISCUSSION

The main aim of the experiment was to induce male-sterility in sesame. Numerous desirable and undesirable effects were observed.

At higher concentrations of F,W-450, treatment shock was seen accompanied by marginal scorching of the leaves. The chemical spray was seen accumulated along the margin of the leaves, and the high concentration of the chemical brought about the scorching of the leaves along their margins. Effects of this kind were reported by Pederson (1959) in alfalfa, Chopra et al (1960) in wheat, Nripendra et al (1963) in muskmelons and Singh (1964) in cotton.

Leaves and flowers of the plants treated with F,W-450 were bigger at the base and comparatively small at the tip of the shoot as in normal plants. But there was an overall increase in the number of leaves per treated plant and must be due to the morphogenic effects of the chemicals. Bocanegra et al (1968) in cotton and Kaul and Singh (1967) in sunnhemp also observed small sized leaves and flowers in plants treated with F,W-450. Kumar and Singh (1963) reported that the size of the

flowers was unaffected in sesame treated with F,W-450.

All the 2, 4-D treatments were highly toxic to the plants. The plants were seen curled up. The leaves were thick, leathery and dark green in colour. These changes were caused entirely due to the deleterious effect of the chemical. The change in texture of the leaves clearly indicates the abnormal development of the vegetative cells. Results of this type were observed by Choudhari and George (1964) in brinjal.

The flowers and fruits of plants treated with 2, 4-D were bigger in size. This can be due to the enhanced growth of the cells of the fruits and flowers stimulated by the chemical translocated selectively to them. This result is contrary to the results obtained by Fehm (1952) in watermelons. He observed small fruits in plants treated with 2, 4-D at a concentration of 5 ppm.

All the treatments with F,W-450 increased the height of plants. It means that the chemical was effective in stimulating shoot growth; a typical hormonal effect. The internodes were normal in length but the number was increased when compared with that of the control plants. This result is entirely different from those reported by Kumar and Singh (1963) in sesame and Dubey and Singh (1968) in bhindi. Meyer et al (1958)

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reported that in cotton F,W-450 caused negatively significant growth over the control owing to the death of the apical meristem. Such deleterious effects were not observed in this work.

2, 4-D in all treatments reduced the height of the plants considerably and it was one of the harmful effects of the chemical. All the growing points were seen destroyed and thus apical growth was suppressed. Reduction in height was directly proportional to the concentration of the chemical. Choudhary et al (1960) observed such effects in cucumber. Choudary and George (1964) reported elongation of the stem in brinjal treated with 2, 4-D but the branches were brittle and slender.

The total number of flowers in plants treated with F,W-450 was greater than that of the control or other treatments. There was a peculiarity noted with respect to the development of the flowers. Normally in an axil only one flower is seen with two nectarial glands on either side which are reduced flower buds. But in the case of all plants treated with F,W-450 there were three flowers in each axil and it was due to the development of the flower buds which in normal case abort to form the nectarial glands. Thus F,W-450 shows a stimulating effect on flower production and growth. This could be in the nature of a hormonal action. Further,

flowering was also hastened.

But these results are contrary to the findings of Chopra et al (1960), Kumar (1963) and Kaul and Singh (1967).

Flowering was seen delayed with the application of 2, 4-D. Jose and Singh (1967) reported effects of this kind in tobacco plants treated with 2, 4-D at 0.05% concentration.

In general the shape of the pollen grains in treated plants was spherical and seen unaltered. Eaton (1957) reported that the shape of pollen grains in cotton treated with F,W-450 showed great variation. Such results were also obtained by Narayana Swamy (1960) and Chopra et al (1960) in cotton. The inviable pollen grains in both treatments were shrivelled or ruptured along their periphery or were with vacuolated plasma. Rehm (1952) observed such effects in watermelons treated with 2, 4-D.

There was no considerable variation in size of pollen grains from F,W-450 treated and control plants. Pollen grains of 2, 4-D treated plants were small when compared with other treatments.

Nair (1964) and Leclamma (1965) reported the presence of small pollen grains in plants treated with

F,W-450. Rehm (1952) observed small pollen grains in plants treated with 2, 4-D.

Half filled and empty anthers were present in treatments with both chemicals. In some cases anthers were completely absent and in still others only part of the anthers were seen. Nondehiscent anthers were also frequent. These effects were more pronounced with 2, 4-D. These findings show clearly that 2, 4-D is an effective male gametocide.

Instances of the failure of anther dehiscence due to the application of F,W-450 were reported by Henz and Mohr (1959) in watermelon. Pate and Duncan (1960), Santhanam (1961) and Kaul and Singh (1967) reported such cases with the application of F,W-450 in cotton.

Method of judging pollen sterility by aceto-carmin staining has shown that the maximum pollen sterility was on the 9th day after treatment in all the cases. This general observation may be due to the uniform absorption and translocation of the chemicals, in all cases, irrespective of the stages of growth.

In treatment with both chemicals the percentage of pollen sterility obtained was directly proportional to

the concentration tried. F,W-450 at the highest concentration of 3000 ppm. induced 70% sterility on an average and it was upto 88% at the first stage of application. There were no deleterious effects other than a mild treatment shock which disappeared in two days. Eaton (1957), Singh (1964) and Guruswamy Raja (1967) reported that they could induce successful male-sterility with less side effects in cotton when they applied F,W-450. But Pederson (1959) in alfalfa, Starness and Hadley (1962) in soybeans, Kumar (1963) in sesame and Kaul and Singh (1967) in onion could get partial male-sterility with F,W-450 treatment.

2, 4-D could induce cent per cent pollen sterility with 150 ppm. and 200 ppm. but the plants were seriously injured so that it cannot be used as a successful male-gametocide.

Application prior to flowering was the most effective one. Results obtained by Moore (1959) and Balakrishnan (1963) in tomato agree with this. Dubey and Singh (1968) reported that the application of F,W-450 prior to flowering was more effective in inducing male-sterility. Reports show that the period of the effect of the chemicals varied with different crops.

But the data may not prove anything in the light of the recent reports from the Division of Botany, Government Agricultural College, Kanpur that the acetocarmine staining method for evaluating pollen sterility is unreliable in the case of sesame.

Pollen sterility assessed by germinating the pollen grains in artificial medium was less than that obtained by the acetocarmin staining method.

The medium contained 20% sucrose and 50 ppm. boric acid gave the maximum germination of pollen grains and the same was used as the standard medium.

F,W-450 at concentration of 3000 ppm. gave an average of 68% sterility. Maximum sterility observed at this concentration was 80.34% at the first stage of application. The general average of pollen sterility induced by this method was 48%. Maximum sterility of 97.3% was observed with 2, 4-D at concentration of 200 ppm in the second stage of application. Sterility was directly proportional to the concentration of the chemical.

Length of pollen tubes in treatment with F,W-450 and 2, 4-D was less than that of distilled water treated and control plants. But pollen tube length in treatment with 1500 ppm. of F,W-450 exceeded that of the control.

All the 2, 4-D treated pollen grains showed far less growth of pollen tubes when compared with all other treatments. In general the chemicals were having effect in inhibiting pollen tube growth. Thus the retarded growth of the pollen tubes can be due to the presence of the chemical in the mature pollen grains.

Mehrota and Sanghi (1966) reported that germination of pollen grains in artificial medium was the most reliable method for assessing pollen sterility in sesame.

Pollen sterility was also judged by crossing the control plants with pollen from treated plants. Result showed that the percentage of fruit set was very low at higher concentration of the chemicals. On the other hand fruit set was normal with pollen grains of untreated or control plants. From this it can be inferred that the pollen grains were sterile. The general average of fruit set in case of plants crossed with pollen from plants treated with 3000 ppm. of 2,4-D was 22.34% showing that the sterile pollen grains were maximum at this dose. 2, 4-D showed cent per cent failure of fruit set, indirect showing complete pollen sterility.

Ovular sterility was tested by crossing emasculated treated flowers with pollen from untreated plants or control. Ovular sterility was observed at higher and

lower concentrations of treatment with the chemicals and the intensity was directly proportional to the concentration. Ovular sterility was high in plants treated with 2, 4-D. The average maximum and minimum sterilities observed with F,W-450 treatments were 73% and 38.81% respectively. The sterility in control and distilled water treatments were 3.5% and 5% respectively.

Treatments with 2, 4-D was highly toxic to the ovary. Complete sterility was observed at high concentrations of 150 ppm. and 200 ppm.

Comparing the results obtained in control and distilled water treatments it is clear that the fertility of the ovary was affected by the chemicals. Presence of empty and half filled capsules and seeds also confirm that ovular sterility was produced with the application of the chemicals.

Moore (1959) reported that an application of 0.15% F,W-450 in tomato affected ovular fertility. Duboy and Singh (1966) reported that they observed 13% ovular sterility with 0.4% of F,W-450, in bhindi.

Mechanism of pollen abortion was studied. The cross section of anthers from the plants treated with the chemicals showed hypertrophied and collapsed tapetal cells.

This may be due to the action of the chemicals. The function of the tapetum is to nourish the developing microspores. When the tapetal cells were destroyed the developing sporogenous cells were starved and thus normal development was blocked. With continued starvation they disintegrated. Empty and partially developed microspores were shrivelled up. Empty microspores will not be able to maintain their shape and naturally they will get shrivelled up. Maheswari (1950) reported the importance of tapetal cells in growth and development of the microspores.

Kaul and Singh (1967) reported, after their studies in Trigonella foenum-graecum that the developing pollen grains were seen as if subjected to high pressure and the pressure was exerted by the hypertrophied tapetal cells. Singh and Hadley (1961) and Kumar (1963) reported the similar nature of tapetum in sorghum and sesame treated with chemicals. Hypertrophy of the tapetal cells was reported by Artsch Wager (1947) in semi-sterile Beta vulgaris L.

Longitudinal sections of vegetative buds showed no symptoms of damage in both treated and control plants indicating that the food supply was uninterrupted and it did not contribute to the disintegration of the pollen grains.

Number of fruits was more in plants treated with F,W-450 than that of control and all other treatments. This result is contrary to those obtained by Pate and Duncan (1960) and Richmond (1961) in cotton and Dubey and Singh (1968) in bhindi. Previously it was mentioned that the number of flowers in F,W-450 treatment was high. Therefore it is natural that the number of fruits will also be high.

Fruits of F,W-450 treated plants were small and there were empty and half filled capsules and seeds. It clearly shows that the reduction in the size of the fruits was due to the effect of the chemicals.

In all plants treated with F,W-450 there were three capsules per axil and were observed only after the treatment and the effect was distinct at the second and third stages of application because the presence of single capsules per axil produced prior to the treatment. Langham (1945) reported the presence of three capsules per axil as a hereditary character in sesame. But in this experiment all the plants of control, distilled water and 2, 4-D treatments produced only a single capsule per axil thus confirming that the presence of three capsules per axil in F,W-450 treated plants was entirely due to the effect of the chemical.

Frequency of empty capsules and seeds was high in 2, 4-D treatments.

The average yield of plants treated with F,W-450 was high as against the reports of Kumar (1963), in sesame and of Dubey and Singh (1968), in bhindi. Increase in yield is directly proportional to the number of capsules which in turn is correlated with the increase in the number of flowers.

It was also observed that the chemical affected the viability of the seeds. Percentage of germination was low in seeds obtained from F,W-450 treated plants and still low in those obtained from 2, 4-D treated plants.

SUMMARY

SUMMARY

The experiment was conducted to study the effects of F,W-450 and 2, 4-D as male gametocides in sesame, at three stages of growth.

The concentrations tried were 1500 ppm., 2000 ppm., 2500 ppm. and 3000 ppm. in case of F,W-450 and 50 ppm., 100 ppm., 150 ppm. and 200 ppm. in case of 2, 4-D.

2, 4-D was highly toxic to the plants at all concentrations. F,W-450 at 3000 ppm. caused treatment shock expressed as marginal scorching of the leaves.

Pollen sterility was tested by acetocarmine staining method, by germination of pollen grains in artificial medium and by crossing the control plants with pollen from treated plants.

The first stage of application was more effective in inducing pollen sterility and F,W-450 could induce 73% sterility without too much deleterious side effects. The plants treated with F,W-450 were vigorous in growth. Maximum pollen sterility was observed on the 9th day after treatment in all the cases.

Ovular sterility was judged by crossing the treated plants with pollen from control plants. Sterility was observed with the application of both the chemicals but the intensity was directly proportional to the concentration of the chemicals.

Viability of the seeds was also tested and it was found that the chemicals reduced the percentage of germination. Viability was too much reduced in seeds from plants treated with 2, 4-D.

In short resplendent results were obtained with F,W-450 and it was comparatively a better male gametocide for sesame. Results with 2, 4-D showed that this chemical will not be a successful male gametocide in sesame.

The never ending enthusiasm of man and the highly developed nature of his grey matter are mastering the nature in multidirectional fashion. I am optimistic to believe that this experiment will add a speck of information to the heap of modern agriculture.

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ILLUSTRATIONS

FIGURE I

Bar diagram showing final height of plants per
treatment (in cm.)

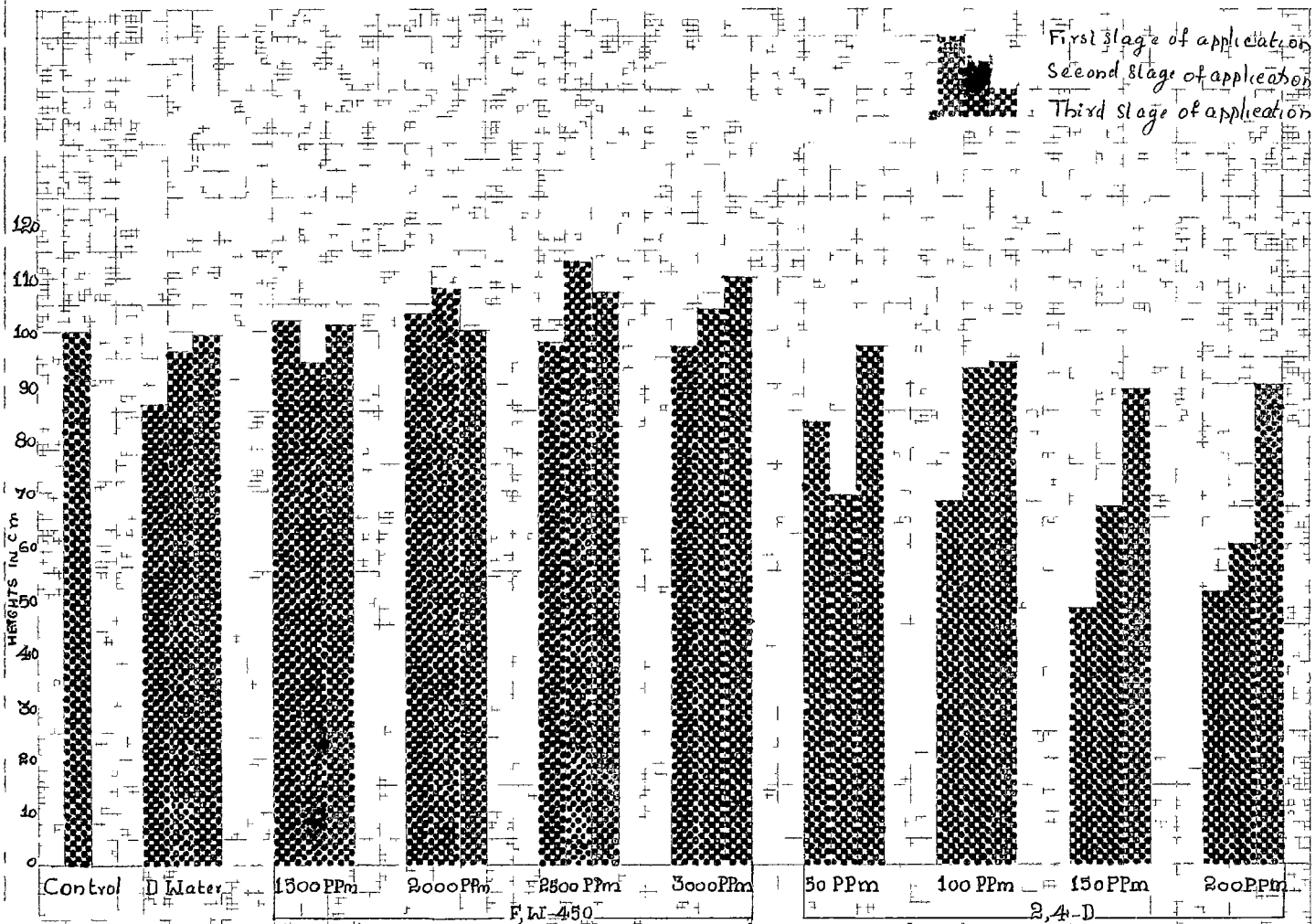
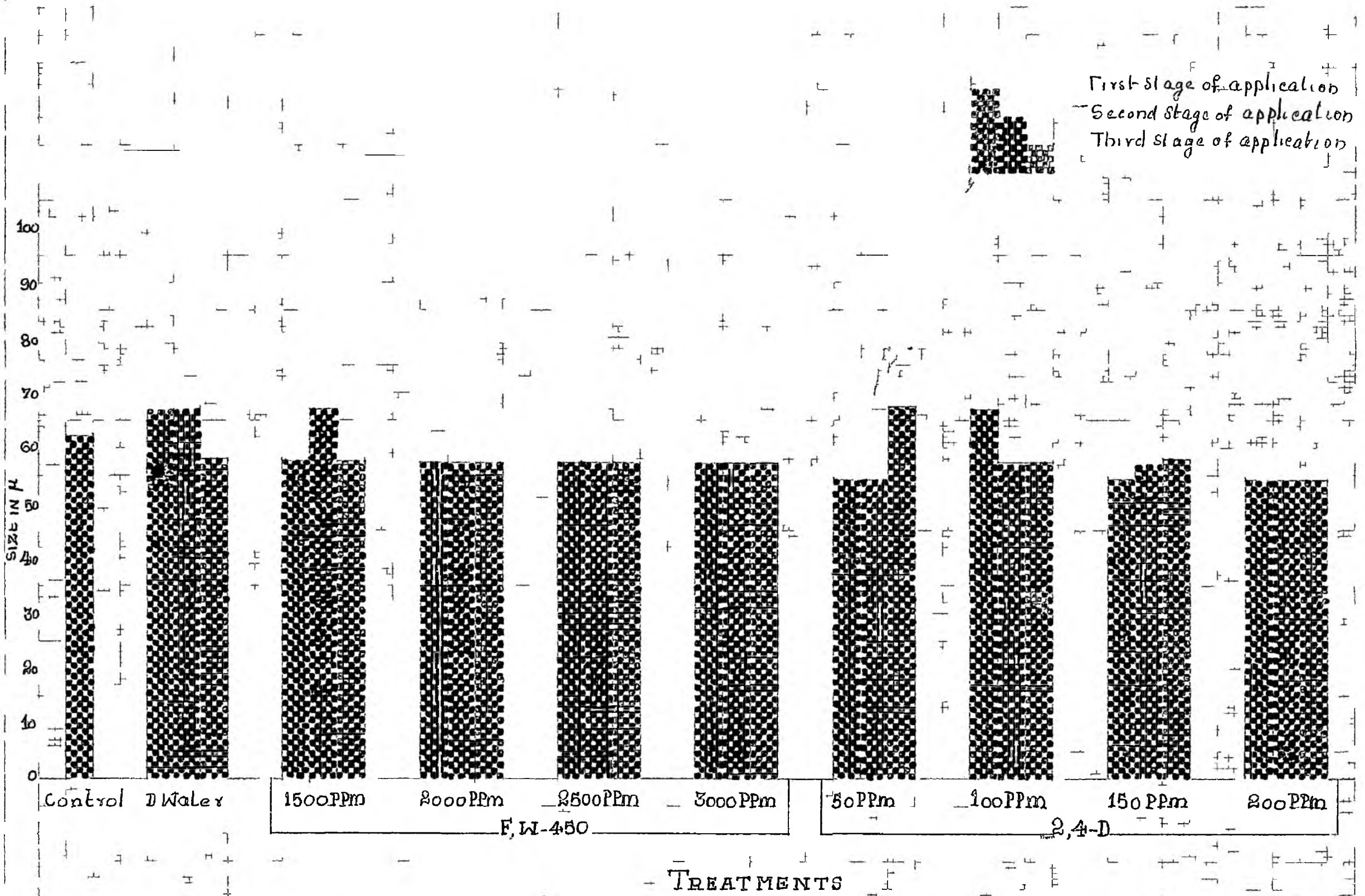


FIG 1

FIGURE II

Bar diagram showing size of pollen grains (in μ)
in each treatment



TREATMENTS

FIG 2

FIGURE III

Bar diagram showing percentage of pollen sterility
by acetocarmine staining method (9th day after
treatment)

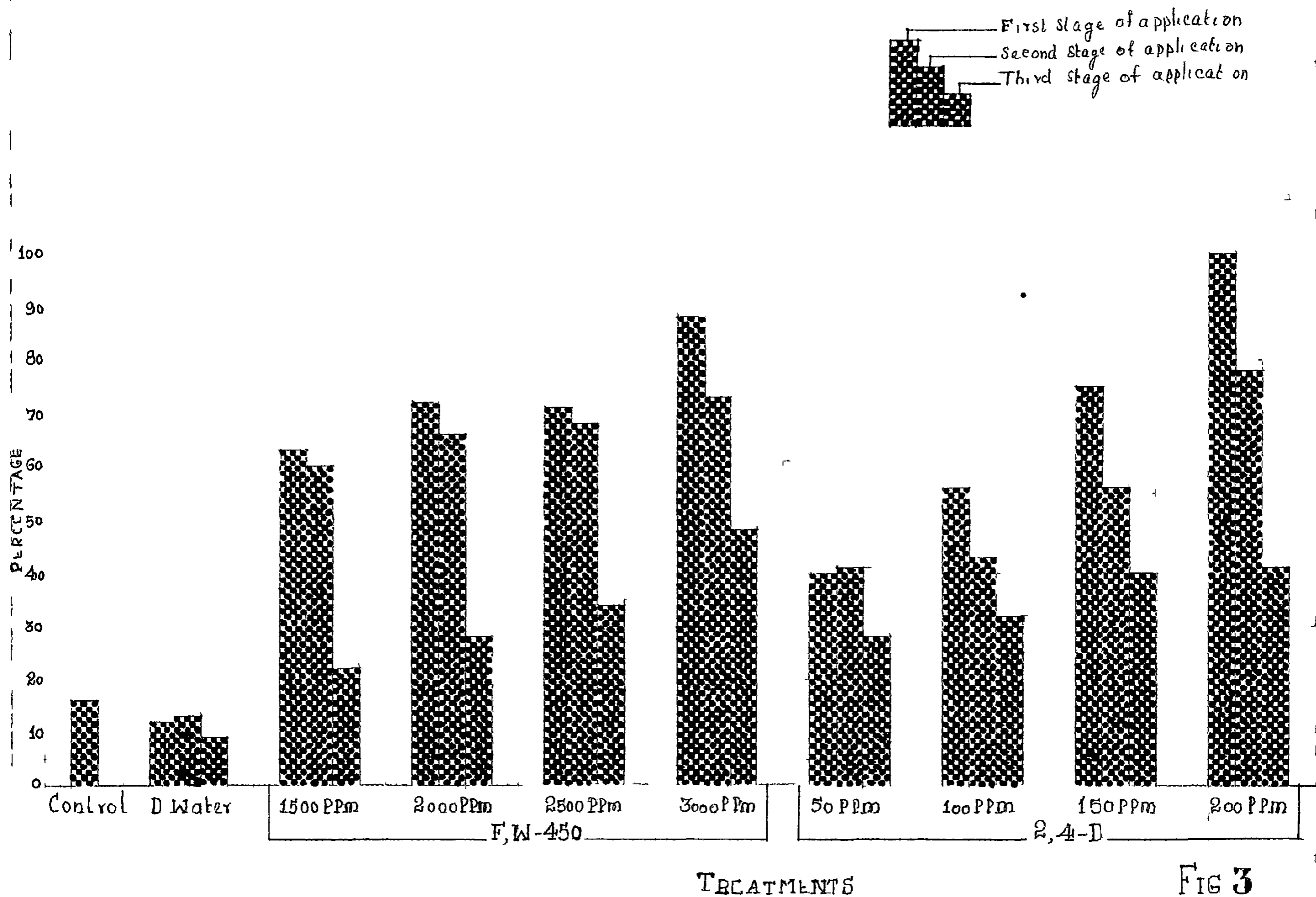


FIG 3

5/24

FIGURE IV

**Bar diagram showing percentage of pollen sterility
in artificial medium (9th day after treatment)**

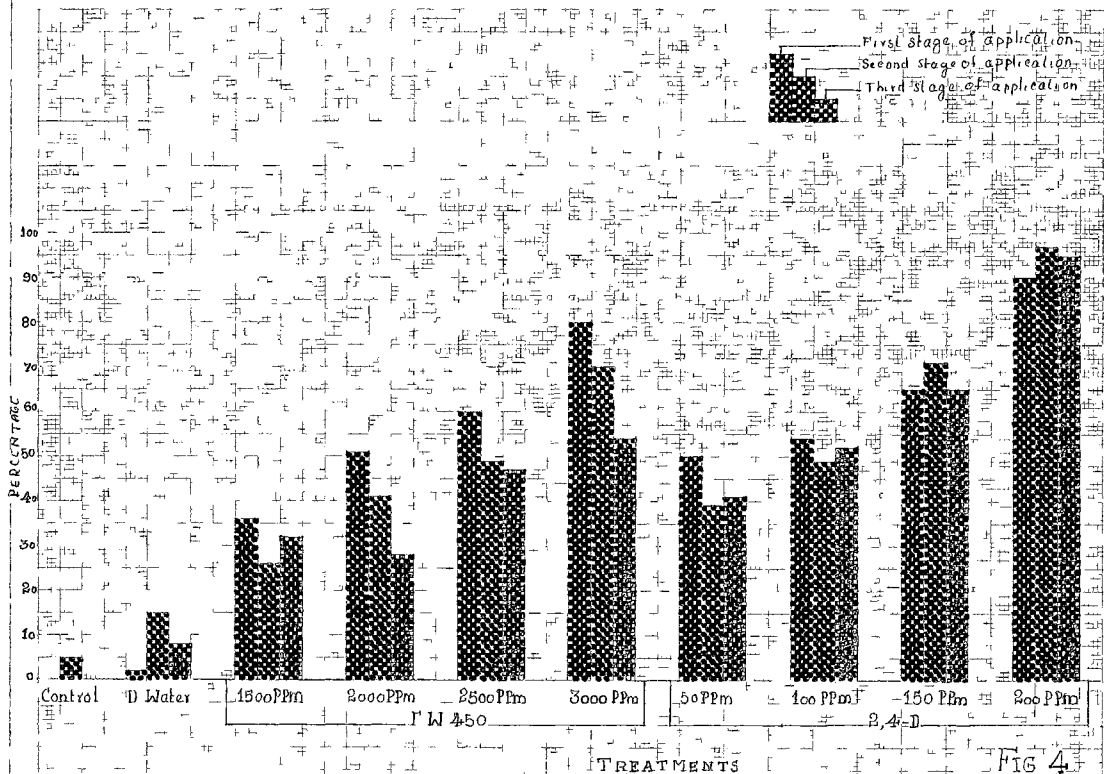


FIG 4

FIGURE V

Bar diagram showing length of pollen tubes in
artificial medium (after 24 hours)

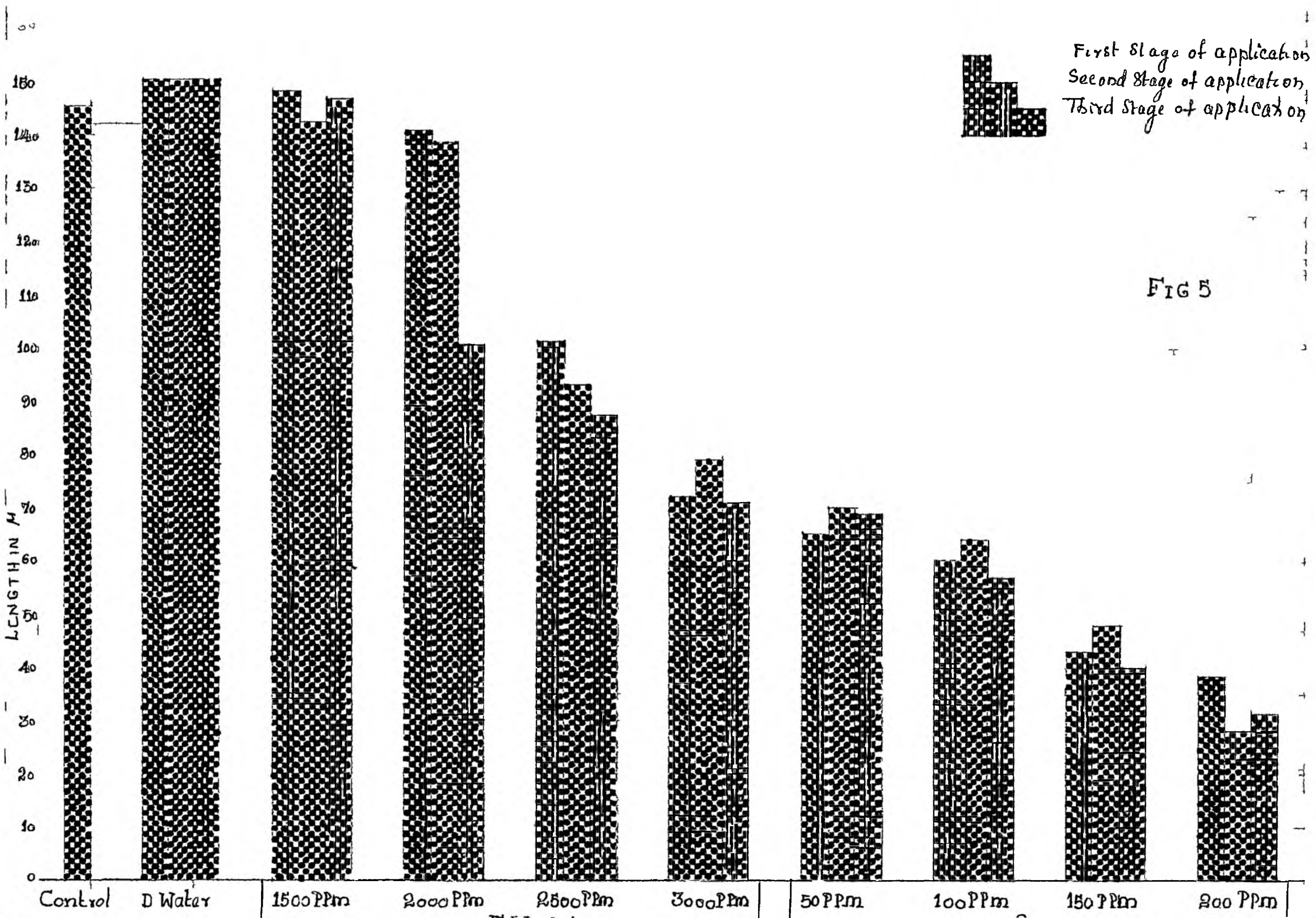


FIG 5

FIGURE VI

Bar diagram showing percentage of fruit set on
crossing control plants with pollen from treated
plants

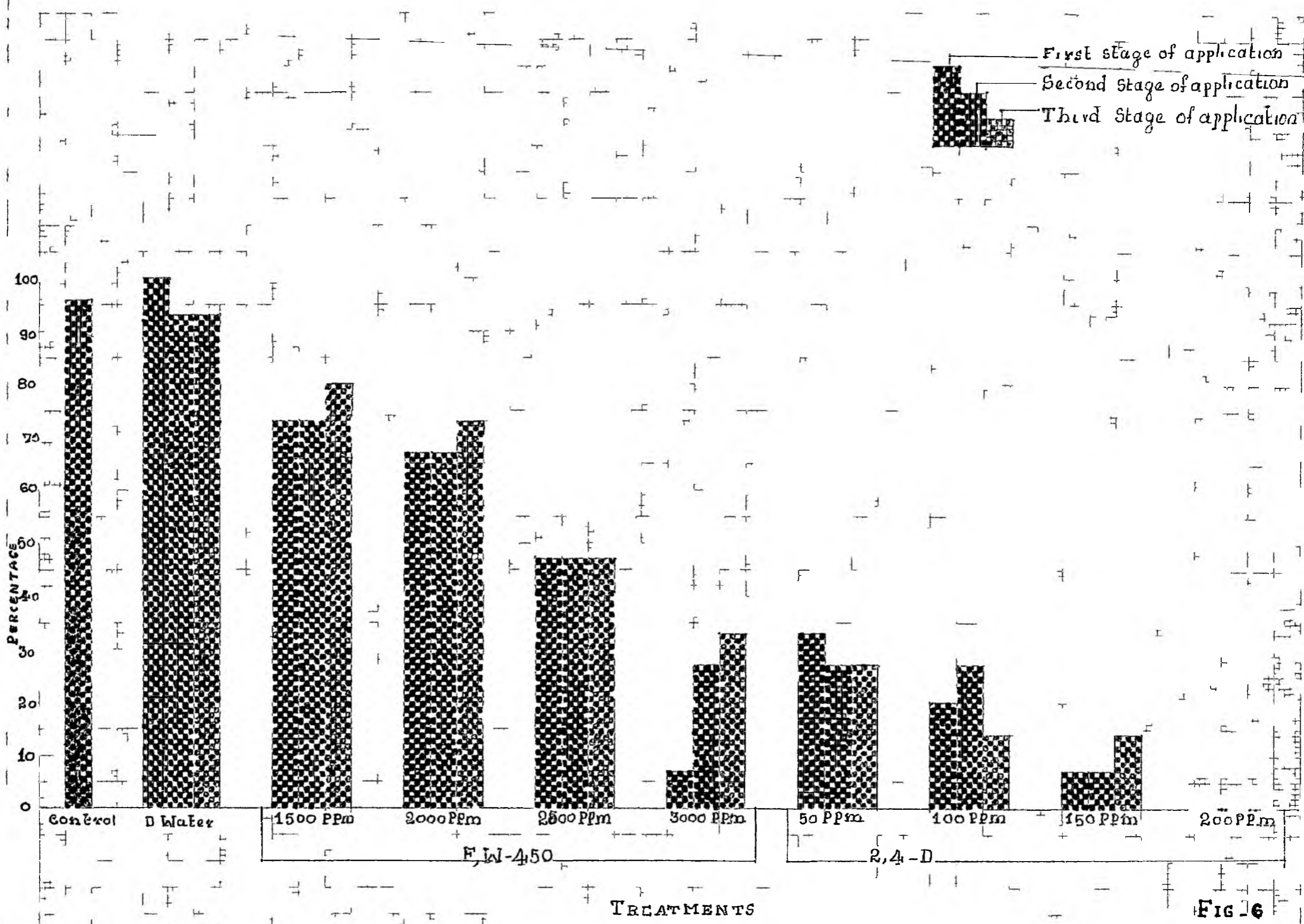


FIG. 6

FIGURE VII

Bar diagram showing percentage of fruit set on
crossing treated plants with pollen from control
plants (ovular sterility)

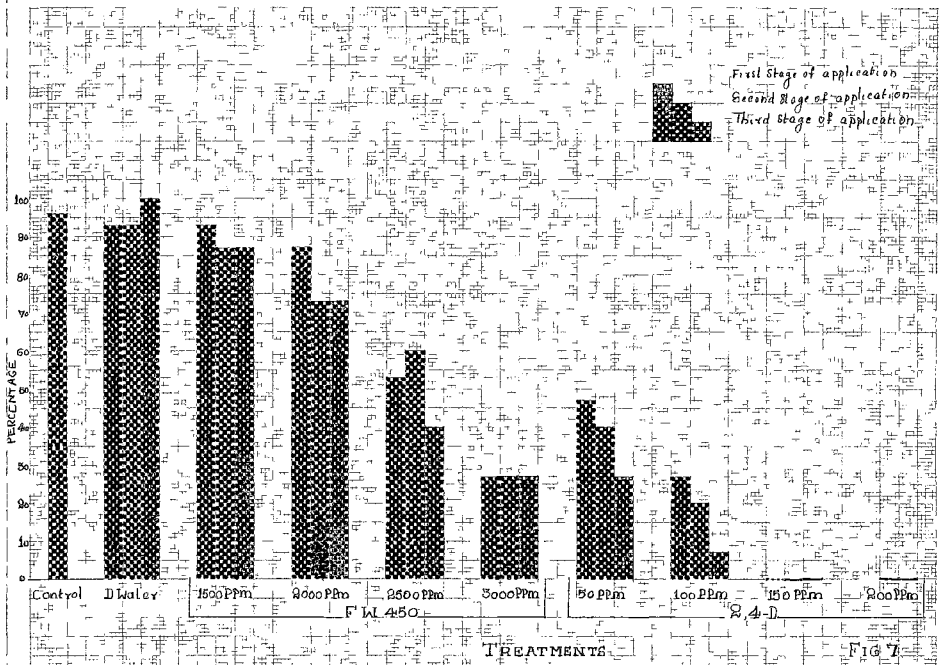


FIG 7

FIGURE VIII

Bar diagram showing percentage of fruit set per
treatment

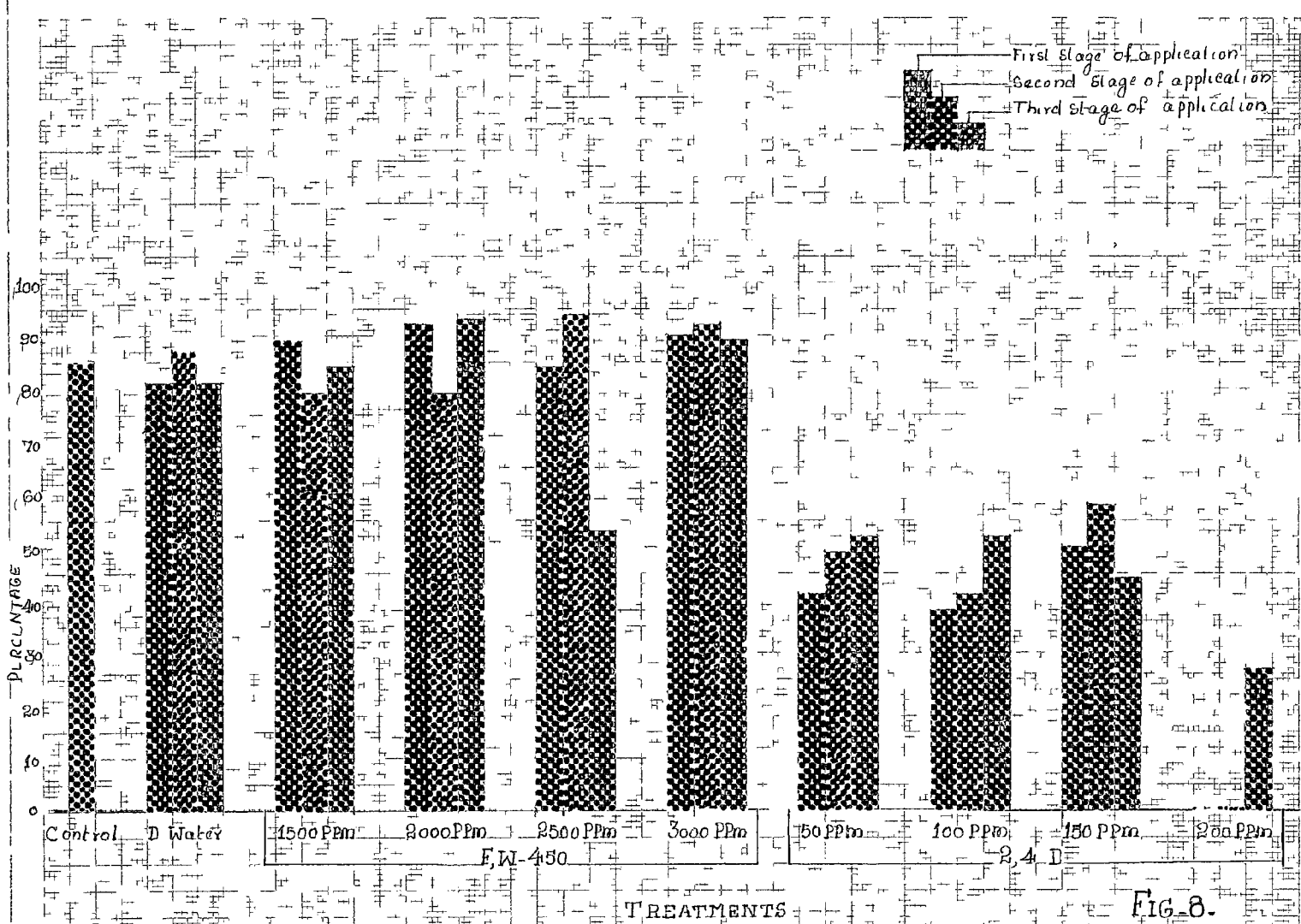


FIG. 8.

FIGURE IX

Bar diagram showing general germination percentage
per treatment

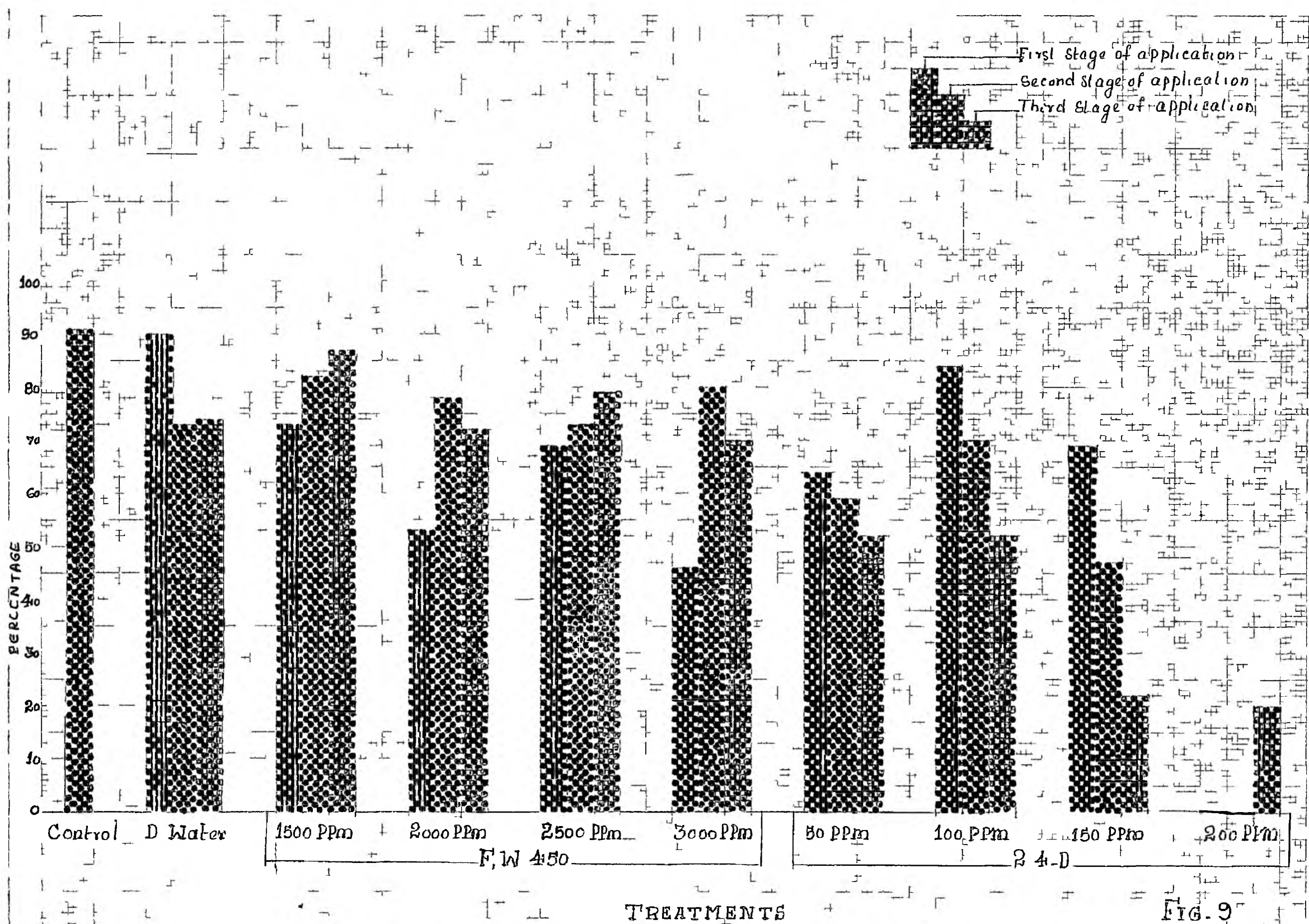


FIG. 9

PLATE I

Comparison between control plants and distilled
water sprayed plants

Left Control plants

Right Distilled water treated plants
(first stage)



PLATE II

**Comparison between control plants and distilled
water sprayed plants**

Left

Control plants

Right

**Distilled water treated plants
(second stage)**



PLATE III

Left Plants treated with 1500 ppm of F,W-450 at
the third stage of application.

Middle Control plants.

Right Plants treated with 3000 ppm of F,W-450 at
the third stage of application.



PLATE IV

Left Plants treated with 1500 ppm of F,W-450 at
the first stage of application.

Middle Control plants.

Right Plants treated with 3000 ppm of F,W-450 at
the first stage of application.



PLATE V

Left **Plants treated with 1500 ppm F,W-450 at
the second stage of application.**

Middle **Control plants.**

Right **Plants treated with 3000 ppm of F,W-450 at
the second stage of application.**



PLATE VI

Left Plants treated with 3000 ppm of F,W-450
at the third stage.

Middle Control plants.

Right Planted treated with 2000 ppm at the
third stage.



PLATE VII

- N_4T_1 - Plants treated with 3000 ppm of F,W-450
(first stage)
- N_3T_1 - Plants treated with 2500 ppm of F,W-450
(first stage)
- N_2T_1 - Plants treated with 2000 ppm of F,W-450
(first stage)
- N_1T_1 - Plants treated with 1500 ppm of F,W-450
(first stage)



PLATE VIII

Left

Control plants

Right

Treated plants (3000 ppm F,W-450 at the
third stage)



PLATE IX

<u>Left</u>	Control plants
<u>Middle</u>	Plants treated with 200 ppm of 2, 4-D (first stage)
<u>Right</u>	Plants treated with 50 ppm of 2, 4-D (first stage)

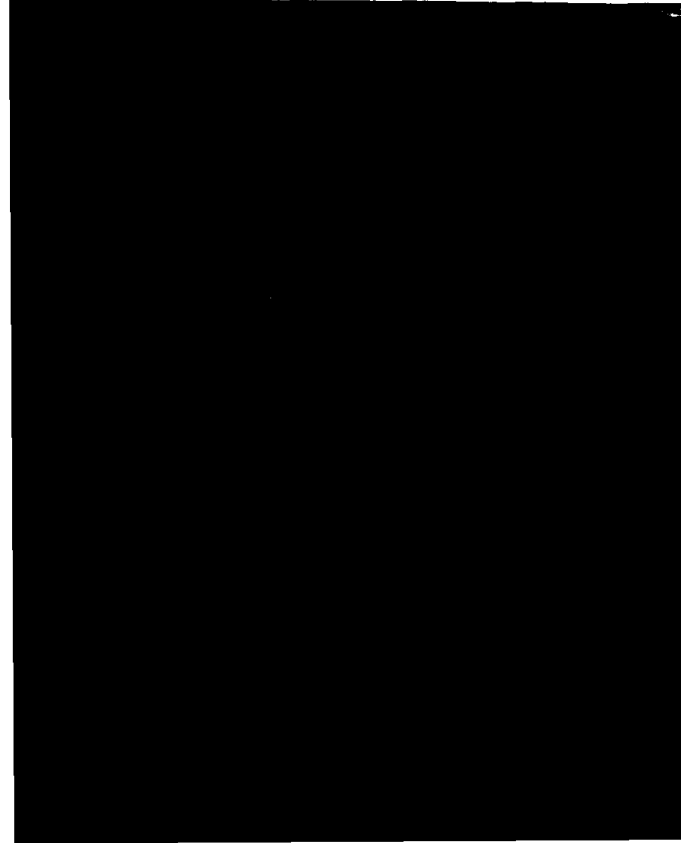
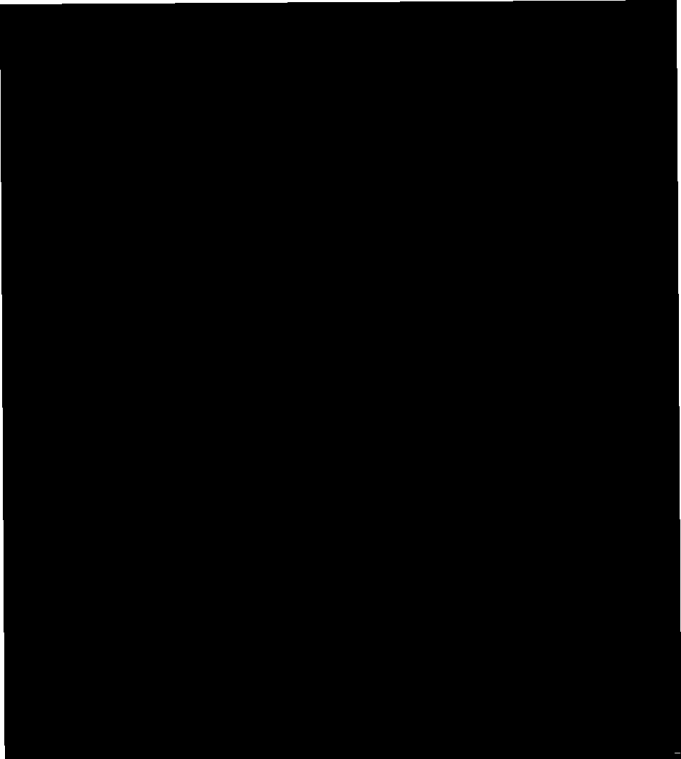


PLATE I

Capsules from different treatments

- | | |
|----------------|---|
| C | Control |
| L | Distilled water treated |
| 2, 4-D | Note the curled nature of the stem |
| F,W-450 | Note the presence of three capsules per axil |

FW450