

THE TOXIC EFFECTS OF DDT AND BHC DUSTS TO THE LEAVES OF WATERMELON

by

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INTRODUCTION

Since the prehistoric times, insect pest has been one of those very serious problems of human-beings. But as our scientific knowledge is increasing day by day, changes have been brought about in our living mode. The cultivated plants which supply us with food, clothing and other daily needs, are generally damaged by various insect pests. In order to have a better harvest of our cultivated economic plants, we should get rid of those weedy vegetations, diseases, and insect pests as clear as possible. As human knowledge is increasing, the methods of controlling the insect pests are also improved.

Of all the methods used to control pests, the insecticides are more frequently applied. The discussion of the insecticides in this paper is, however, limited only to P-P'DDT (2,2-bis) (P-Chlorophenyl-1. 1. 1.-trichloroethane) and γ -isomer of BHC (Benzene Haxachloride or 1, 2, 3, 4, 5, 6-haxachlorocyclo haxane), although a wide variety of the chemical substances is used for this purpose.

The phytotoxic phenomenon, as reported by Brown⁽¹⁾, made by the DDT and BHC on the leaves of cucumbitaceae, especially in watermelon (*Citullus vulgaris* schrad) is worthy of our special attention. And Crowell⁽²⁾ has also found that all compounds of the chlorinated hydrocarbons are capable of inflicting injury upon cucumbitaceae in general under the proper condition of growth.

The properties of P-P'DDT are well known to be used in insect control. In using the solid form of the DDT, it is not acutely toxic in light dosage to the plant, for it would normally or accidentally be ingested (according to shepard's report.)⁽³⁾. Meanwhile the insects would be still alive. But if it is used in continuous dosage of smaller quantities or over dosage, both the plants and the insects are getting hurt.

The γ -isomer BHC is the other commonest insecticide, but with higher toxicity to the insects. It is, however, less toxic to those domestic animals and men, because it is non-toxic even absorbed by the skin. The BHC, therefore, is more effective and

essential in killing insects than DDT, when it is applied with about $\frac{1}{10}$ in quantity⁽³⁾.

This experiment was planning to study the histological structure of normal leaves of watermelon compared with the phototoxic leaves by these two insecticides of different concentrations.

MATERIALS AND METHODS

On April 5th the seeds of watermelon were germinated in soil. Plants of 3-4 inches in height were transplanted to 21 flower pods, one to each pod, in which contains sterilized sand soil with adequate fertilizer. And it was placed in the green house of the Dept. of Botany, Taiwan Univ. Taipei, Taiwan.

Two kinds of insecticides, γ -isomer BHC and P-P'DDT were prepared in impregnated dusts* and their concentrations were as the following.

- (1) γ -isomer BHC, at 0.5% and 1%.
- (2) Refined P-P'DDT Set. pt. 103°C, at 2.5%, 5% and 10%.

By May 21st, more than 10 leaves of each plant have grown out. The impregnated dusts were applied to both upper and lower surfaces of the insect-free leaves of the watermelon by hand duster. To stick the dusts on the leaves, water spray is necessary. Then dusts about 0.003 gr per cm² were applied on the leaves of the two plants with each concentration of the two insecticides formulated as above, with uniform dusting.

Then 2 or 3 leaves were picked up from after the experiment proceeded for two weeks and three weeks respectively. Then they were fixed in F. A. A. (Formaline, alcohol and acetic acid) fluid. After 7 days the materials were imbedded in paraffin in the usual way and sectioned to 15-20 μ in thickness, stained with safranin and Delafield's Haematoxylin, and mounted in Balsam, and then the slides were ready to be examined microscopically.

RESULTS AND DISCUSSIONS

I. External Symptoms of the Leaves.

Three weeks after the treatment of both the insecticides of DDT and BHC, the injury has been found on every plant. The most parts of the leaves have brought about chlorosis. Some of the leaves were shrunk and with light epinasty. The results are given in table I.

In table I. It showed that 10% P-P'DDT, and 1% γ -isomer BHC killed the plant leaves more rapidly than the other concentrations. They were dried after a few weeks. About three weeks after each treatment of DDT, the terminal bud of the plant began to grow again, for the growing point of the plant had not been damaged, yet, and sometimes the plant grew faster than the untreated one. The phenomenon is explained by Allen⁽¹⁾, that the DDT treated on the lower leaves was being utilized by the upper

* The preparation of impregnated dusts.—The insecticides are dissolved at first in a small amount of acetone, then the solution is poured into fine talcum powder and stirred very well to form a pasts. Then let it dry in desiccator and sift it about 200 meshes in size, and stock it for using.

Table I. Showing the external symptoms of leaves after treated insecticides 0.003 gr/cm²

insecti- cides	γ -isomer BHC		P-P'DDT. Set. pt. 103°C		
	0.5 %	1 %	2.5 %	5 %	10%
weeks after treatment					
1 week	—	—	—	—	—
2 weeks	—	light chlorosis	—	pale green	chlorosis with yellow spot
3 weeks	chlorosis	severe chlorosis	chlorosis	severe chlorosis	severe chlorosis
4 weeks	chlorosis	leaves wilt	chlorosis	most wilt	most dried

portion of the plant acting as something closely resembling plant hormone with which the plant grew more rapidly.

II. Internal Anatomy of the Leaves.

The Internal structure of a normal leaf. There are 3 main parts of tissues shown by a cross-section of the normal leaf. (Fig. 1.)

1. Epidermis. The epidermal cells of both upper and lower surfaces are the same but the stomata and the hairs of upper surface were less than that of the lower surface. Each cell possesses a large central vacuole, but no chloroplasts, except in the guard cells. The outer cell wall is much thicker than the inner one.

2. Mesophyll. The palisade and spongy parenchyma. There is only one layer of palisade tissue below the upper epidermis. It is elongated downward from the upper epidermis to the spongy parenchyma, and arranged more regularly with intercellular spaces. The spongy parenchyma below the palisade layer, there are 4-5 layers of spongy parenchyma cells and arranged more irregularly, occupying more than half of the mesophyll with large intercellular spaces. The cells of both palisade and spongy parenchyma all contain chloroplasts.

3. Vascular bundle. The large vein or the midrib contains xylem and phloem, all of them being surrounded by parenchyma, and collenchyma cells.

III. Histology of the Injured Leaves.

The internal structure and the changes of the form of the leaves treated with the insecticides are shown in a cross section of the photomicrograph as in Fig. 2-11.

The leaves are treated with P-P'DDT dusts in 3 different concentrations.

1. 2.5% dusts. After 2 weeks (Fig. 2), the palisade parenchyma cells are much shortened but still have elongated cells comparing with that of the control and without any distinction from the spongy parenchyma. The intercellular spaces in both palisade and spongy tissue decreased. After 3 weeks (Fig. 3), these symptoms became rather more severe than (Fig. 2). The epidermis formed an undulate surface because of the shrinkage of the individual epidermal cell. The spongy tissue compressed together

and the amount of chloroplasts was reduced too.

2. 5% dusts. After two weeks (Fig. 4), the central vacuole of the palisade cells and the spongy cells were full with the chloroplasts and the intercellular spaces have reduced. After 3 weeks (Fig. 5), the injurious symptom was more prominently shown. The palisade and the spongy tissues were transformed into a mass and tightly packed together between the two epidermal layers. The discolouration and the decrease of the number of the chloroplasts were forming chlorosis.

3. 10% dusts. After two weeks (Fig. 6), the leaf blade have been flattened in some parts. The chloroplasts were less throughout the mesophyll but located on the walls of the cells. The palisade layer was becoming quite abnormal. There was practically no intercellular space remaining in the spongy tissues. After three weeks (Fig. 7), there were no other distinct characteristic changes but very seriously injured.

The above mentioned changes of the structures of the leaf of watermelon by DDT treatment, may be summarized in Table II.

structures	concentrations and weeks after DDT treatment					
	2.5 % 2 weeks	2.5 % 3 weeks	5 % 2 weeks	5 % 3 weeks	10% 2 weeks	10% 3 weeks
Epidermis	normal	slight shrinkage	normal	shrinkage	shrinkage	shrinkage
Palisade cell	normal with some shorten	normal with some shorten	normal with some shorten	abnormal	abnormal	abnormal
Spongy cell	intercellular space decreased	intercellular space decreased	intercellular space decreased	without inter-cellular spaces	without inter-cellular spaces	without inter-cellular spaces
Chloroplasts	normal	number decreased	condensed together	number decreased	very less in number	very less in number

The leaves treated with γ -isomer of BHC dusts.

1. 0.5% dusts (Fig. 8). After 2 weeks, the BHC dusts had been applied to the leaves, the structure of the leaves revealed no more distinct changes than the normal leaf but only the chloroplasts reduced in the mesophyll. After 3 weeks (Fig. 9), the symptoms appeared in the structure of the treated leaves were the same as that of 5% DDT dusts. The chloroplasts in the mesophyll cells were moved to the center. The only difference between the treatment of the 5% DDT dusts and 0.5% BHC dusts, in this case was that both the epidermis and the mesophyll were normal

2. 1% dusts (Fig. 10). After 2 weeks, the epidermis showed dryness in a certain spot (Fig. 10) indicated. The size of palisade tissue decreased and it was arranged irregularly. There was no intercellular space remaining in the spongy tissue. After 3 weeks (Fig. 11) on the leaves there appeared some dried spots. And the other structures brought about a severe injury.

The time factor must be taken into consideration in this experiment. The data listed in table I and II show that there was no marked difference in the structure of the leaves, under the concentration of 2.5% and 10% DDT dusts and 0.5% and 1% BHC dusts applied to the plant from the 2nd to the 3rd week, but the concentration

of 5% DDT dusts brought about a marked change in the structure of the treated leaves. This phenomenon is probably due to that the appropriate concentration of 5% DDT dusts is correlating to the time factor, below or above this concentration, it is either of no effect or too harmful to the plant life.

The above mentioned changes of structures of the leaf of watermelon after BHC treatment may be summarized in table III.

structures	concentrations and weeks after BHC treatment			
	0.5% 2 weeks	0.5% 3 weeks	1% 2 weeks	1% 3 weeks
Epidermis	normal	normal	scattered spots	scattered spots
Palisade cells	normal	normal	clapsed, began to disorder	clapsed, began to disorder
Spongy cells	normal	normal	without inter-cellular space	without inter-cellular space
Chloroplasts	decreased in number	moved to the center	number decreased	number decreased

There is another factor, the concentrations. We found that more than 5% DDT and 0.5% BHC dusts would be harmful to the plant even though they could control and protect against the insect pest. All those phenomena such as the reducing of chloroplasts, the decreasing of the rate of photosynthesis, the disappearing of the intercellular space, caused the leaves to stop their assimilative processes and weaken the living function or even bring them to death.

In controlling the insect pest on the watermelon therefore, it is better not to apply these two insecticides to the plant.

SUMMARY

(1) As the scientific knowledge is progressing day by day, the problem of phototoxicity might be solved with these well known insecticides.

(2) In controlling insect pest, both DDT and BHC are used widely and popularly, especially the impregnated dusts which are more effective to this purpose.

(3) When 5%, 10% DDT dusts and 0.5%, 1% BHC dusts are applied to the watermelon to control the insect pest, however, they have brought about chlorosis to the leaves which effect the internal structure. By examining the cross section of the treated leaves in 15-20 μ in thickness, it showed that the palisade and spongy parenchyma cells of the mesophyll become as a massive undifferentiated cells in which the chloroplasts are so reduced that their living functions are practically ceased and the plant soon wilted.

(4) The time factor is the length of time after applying the insecticides to the plant. The effect of chlorosis spots has been shown only to the concentration of 5% DDT dusts, at the time of the 2nd to the 3rd week. Therefore 5% DDT concentration at the period of the 2nd and the 3rd week after applying is the correlation factor of DDT dust to watermelon plants.

(5) To kill the insect pest of watermelon plants. The minimum concentration of insecticide required 5% for DDT and 0.5% for BHC dust while the maximum concentration of insecticide the watermelon plants capable to resist being 2.5% for DDT, and much less than 0.5% for BHC dust therefore these 2 insecticides are not recommended for controlling insect pests for plants of watermelon.

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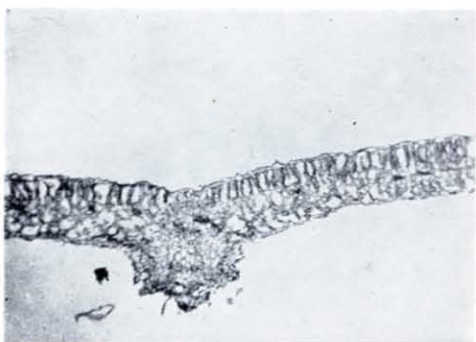


Fig. 1. X-section of Normal leaf of watermelon plant.



Fig. 2. X-section of leaf of watermelon. 2 weeks after 2.5% DDT treatment.



Fig. 3. X-section of leaf of watermelon. 3 weeks after 2.5% DDT treatment.

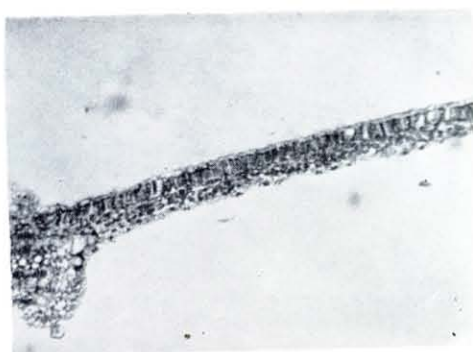


Fig. 4. X-section of leaf of watermelon. 2 weeks after 5% DDT treatment.

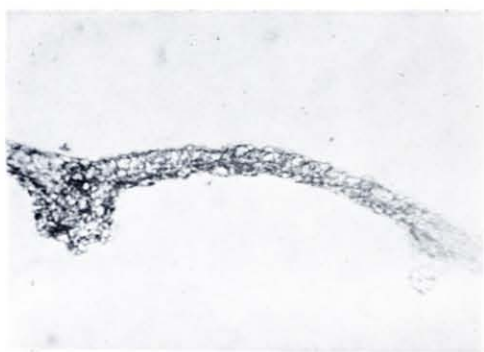


Fig. 5. X-section of leaf of watermelon 3 weeks after 5% DDT treatment.

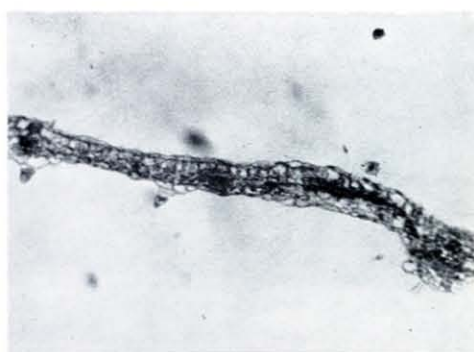


Fig. 6. X-section of leaf of watermelon 2 weeks after 10% DDT treatment.

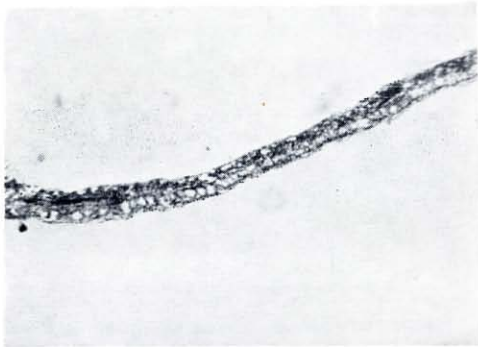


Fig. 7. X-section of leaf of watermelon. 3 weeks after 10% DDT treatment.

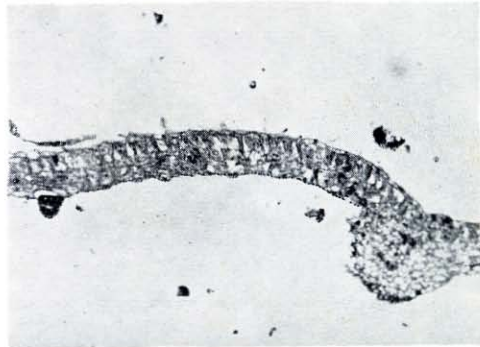


Fig. 8. X-section of leaf of watermelon. 2 weeks after 0.5% BHC treatment.

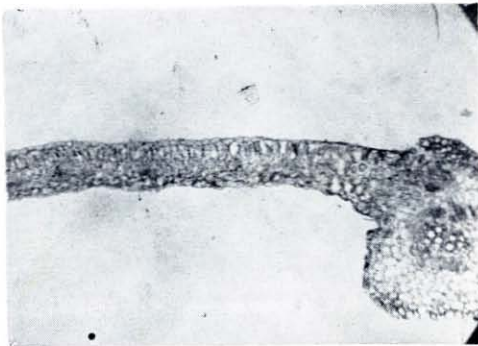


Fig. 9. X-section of leaf of watermelon. 3 weeks after 0.5% BHC treatment.

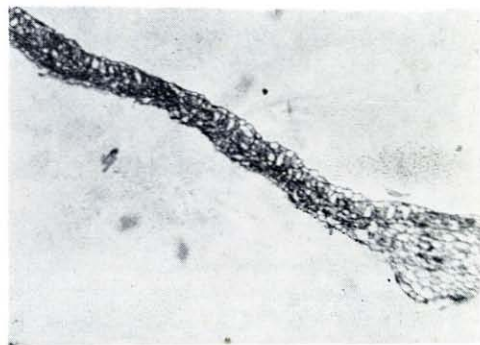


Fig. 10. X-section of leaf of watermelon. 2 weeks after 1% BHC treatment.

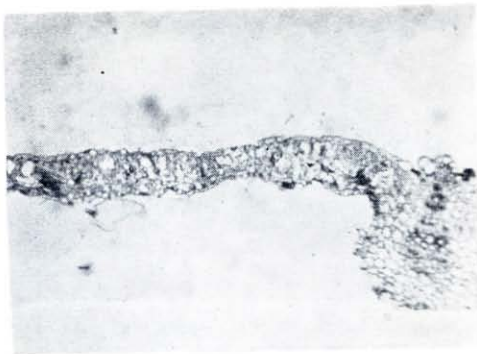


Fig. 11. X-section of leaf of watermelon. 3 weeks after 1% BHC treatment.