

PERSISTENCE OF DDT AND PARATHION
RESIDUES ON A PLANT SURFACE AS
INFLUENCED BY WEATHER FACTORS

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PERSISTENCE OF DDT AND PARATHION RESIDUES ON A PLANT SURFACE AS INFLUENCED BY WEATHER FACTORS

(MET EEN SAMENVATTING IN HET NEDERLANDS)

PROEFSCHRIFT

TER VERKRIJGING VAN DE GRAAD
VAN DOCTOR IN DE LANDBOUWKUNDE
OP GEZAG VAN DE RECTOR MAGNIFICUS IR. W. F. EIJSSVOOGEL,
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THEOREMS

I

The effect of sunshine upon insecticide residues should be taken into special account in formulating insecticides to be used in arid regions.

This thesis

II

A clear picture of the quantities and distribution of contact insecticides applied to a plant material can be obtained only by estimating the toxicants biologically as well as chemically.

This thesis

III

Plapp and Bigley's alternative explanation for the low Ali-esterase levels formed in many organophosphorous-resistant strains of the house-fly is insufficient.

PLAPP, W. and BIGLEY, W. S., (1961), J. econ. Ent., 54, 103-108

VAN ASPEREN, K. and OPPENOORTH, F. J., (1960), Ent. Exp. Appl., 3, 68-83

IV

The serious lack of experts in insect taxonomy in the Middle East requires more attention of the official authorities.

V

In Egypt, the present restrictions upon importation of seed potatoes as regards infection with the Black Scurf disease caused by the fungus *Rhizoctonia solani* are adequate.

STINO, M. N., (1960), A report presented to the Ministry of Agriculture, Egypt, U.A.R.

VI

In classifying elongated plant viruses it is better not to include aphid transmission as a characteristic of these viruses.

VII

When an active region of an antibody molecule consists of four amino acids, it is not probable that there might exist an essentially infinite number of different kinds of combining regions.

HAUROWITZ, F., Molecular structure and biological specificity (1957), p. 22
PAULING, L., ibidem, p. 188

VIII

"Hydroponics" is a practical proposition under Egyptian circumstances where farm acreage is in short supply.

IX

The aid and assistance offered to developing countries should include means for solving social problems concerned with technical and economic development.

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PERSISTENCE OF DDT AND PARATHION RESIDUES ON A PLANT SURFACE AS INFLUENCED BY WEATHER FACTORS

(met een samenvatting in het Nederlands)

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I. INTRODUCTION

Next to the acute toxicity of insecticides, the persistence of their residues on crop plants and vegetation is a very important feature determining their economic value.

This residual effect may be evaluated by chemical and biological means. Bioassays are as a rule directed towards the special effect to be studied, contact bioassays being used to evaluate the activity at the plant surface, while leaf-sandwich methods are used for stomach poisons. Chemical tests are usually applied to follow the total quantity of residue left.

In the literature, mostly either biological or chemical evaluation is mentioned in residue experiments. It occurred to us that a study in which both methods are combined, would be of great use for several reasons. It is known that many contact insecticides may penetrate into plant tissues. In such a case, the activity at the plant surface is much less than is suggested by chemical tests. On the other hand, contact bioassays merely give an impression of the insecticidal activity at the plant surface, and do not reveal the quantity of insecticide present within the plant. In the case of edible crops, the safety of the consumer demands a knowledge of the total quantity of insecticide present.

A comparison between contact bioassay and chemical determination may reveal the relation between the quantities inside and outside the plant. This would be of special value in studying the effect of climatological factors on insecticide residues, as a decrease in surface action could then be attributed to either a decrease in total residue (as revealed by the chemical test) or merely a decrease in surface residue (as revealed by both chemical and biological tests).

DDT and parathion were selected as test insecticides, representing the

chlorinated hydrocarbons and the organo-phosphorous insecticides, respectively. DDT is more stable and generally less toxic than parathion. Moreover, parathion is known to penetrate into plant tissues to an important degree which is true only to a lesser extent in the case of DDT.

The factors that affect the loss of insecticide deposits are certainly complex, but may be grouped in the following categories:

- A. The removal or breakdown of the toxicant brought about by atmospheric elements.
- B. The physical or chemical interactions of the insecticide with its solvent, diluent or any other constituents included in the formulation used as well as the interaction with the substrate to which the material is applied.
- C. Removal or dilution of the residue by plant growth, causing an increase in untreated plant surface.

It is clear that B and C are again subject to atmospheric elements.

Special attention was paid to standardize the effects of the factors of groups B and C. This was achieved by using the same formulation and plant surface. Standard methods were used for the determination of the residues and their effectiveness.

The possible causes of loss of insecticide residues applied to foliage in the open, that are included in group A, may be arranged under three headings: 1. mechanical losses, 2. losses due to other physical processes and 3. losses resulting from chemical changes of the insecticide.

Losses effected by mechanical factors include: removal by wind or by continuous brushing of leaves, washing of the plant surfaces by rain, masking the residue by dust or any foreign material.

The second group includes losses due to changes or alteration of the physical nature of the residue such as a change in the extent of adhesion of insecticide particles, liquefaction of a solid material, volatilisation and dissolution in rain-water.

Losses resulting from chemical changes include: photolysis of material by light and especially ultraviolet light, oxidation as a result of exposure to air, heat induced decomposition and degradation either occurring spontaneously or promoted by catalysts.

The final loss caused to an insecticide weathered for some time is undoubtedly the resultant of several smaller losses affected by some or many of the causes mentioned above. The degree of that loss may then be considered as the product of the stability of a given insecticide and the degree of "stress" produced by the environmental factors during a given time.

As temperature is an inevitable factor in all residue experiments exposed in the open, especially during summertime, it was advisable to include its effect in the present study. Moreover, temperature is certainly the major factor which limits the duration of insecticide deposits in many parts of the world, viz. the Middle and Far East where such materials are increasingly used in agriculture. In connection with temperature, sunshine is always mentioned as an important factor which affects toxicants either indirectly by raising the temperature or directly by its ultraviolet component. Evidence has been accumulating to show that even the most stable insecticides suffer considerable losses at exposure to direct sunshine. Rainfall, on the other hand, is generally considered as a weather component of special importance in both tropical and temperate zones of the

world that seems liable to bring about serious losses of insecticide deposits applied to crops in the field.

As the assessment of the effect of each of these weather factors under normal field conditions is usually very difficult, three groups of experiments were conducted in the laboratory to evaluate the exact effect of temperature, simulated rain and irradiation with an ultraviolet source on DDT and parathion deposits under controlled conditions.

II. MATERIALS AND METHODS

1. SELECTION OF PLANT SURFACE

Since the aim of the present work was to study the fate and persistence of DDT and parathion when applied to a plant surface, it was necessary first to find a plant surface, preferably leaves, possessing the following three characteristics: 1. A sufficient size to render easy both the bioassay and the preparation of samples for chemical analysis. 2. A smooth and flat surface to facilitate the formation of an evenly distributed film of insecticide. 3. Drought-resistant quality that would enable the exposure of the plant surface carrying the residue to harsh weather conditions without serious damage to the plant tissue.

Fully expanded ivy leaves (*Hedera helix*) were considered, after some preliminary tests, to be convenient for the purpose of this study. The leaves of this plant are of suitable size and have a smooth, flat and waxy surface. These leaves also proved to be highly drought-resistant and when kept in test tubes filled with water could withstand bright sunny weather with surface temperatures reaching sometimes 40°C for a week without significant damage to their tissues. When the weather was milder they could survive for much longer periods.

SLOAN *et al.* (1951) stated that plant growth causes apparent reduction in insecticide residues on any particular crop or foliage which would remain proportionally the same regardless of the insecticidal material applied. In order to avoid the effect of growth on the reduction of insecticide residues, cut fully grown leaves represented the best surface for our purpose.

Directly after being cut, the leaves to be used were gently cleaned in running tap water after which they were grouped in glass beakers with their stalks immersed in water and left at room temperature so that their surfaces became dry.

2. TEST INSECT

Calandra granaria L. was selected as the test insect. A continuous stock of these insects was started with a few hundred active young adults obtained from the standard stock at the laboratory. The insects were reared in glass jars on wheat. The whole stock comprising several jars, was maintained at 24°C and at a relative humidity of 50–60% inside a large round constant temperature cabinet throughout the experimental period. In each experiment a mixture of the adults from 3 or 4 cultures, started and renewed on the same date,

were used. Intact, active and normally sized adults were always selected for bioassay purposes.

All cultures were renewed every six weeks. The renewing process was carried out as follows: The weevils of all the cultures were separated from the consumed grain, debris and other remnants by means of a suitable sieve. All the insects of the different cultures were collected in one big jar. On a big smooth sheet of paper strongly illuminated from one side, the insects were poured out in batches, one after the other. Only the active, normal insects which moved quickly were collected in another clean jar. The remaining insects of each batch that failed to move phototactically were discarded. The active young weevils were then distributed equally in a number of clean jars provided with sufficient quantities of fresh wheat.

3. INSECTICIDES TESTED

Two insecticides were tested: 1. DDT (1,1,1-trichloro-2,2 bis (p-chlorophenyl) ethane). 25 % DDT (Gesarol) emulsifiable concentrate (e.c.) provided by Orgachemia Boxtel, Holland.

2. Parathion (0,0-diethyl 0-p-nitrophenyl thiophosphate). 25 % emulsifiable concentrate (e.c.) obtained from Philips-Duphar Amsterdam, Holland.

With either insecticide, the same e.c. stock was used in all experiments and was always kept in a cool dark place. The concentrations used were 0.0312 % for DDT and 0.025 % for parathion, unless otherwise stated. The required quantities of emulsions were always freshly prepared and thoroughly stirred for a few minutes just before spraying.

4. APPLICATION OF INSECTICIDES

Some details of the technique varied from one group of experiments to the other according to the point under investigation and will be mentioned at the appropriate place, but certain operations, technical details and equipment were common to all experiments and will be described here to avoid repetition.

In all experiments an accurately reproducible residue was required on one surface of the leaves. This necessitated a reliable and easy-to-use spraying apparatus. A water emulsion of either insecticide was used for spraying in all experiments. Water emulsions were found more convenient than other formulations because they are easier to prepare, more homogeneous and form excellent films on sprayed surfaces.

The TEN HOUTEN & KRAAK vertical spraying apparatus (figure 1) was used. It proved to be satisfactory, provided special attention was paid to constancy of air pressure, air flow and quantities of fluid sprayed. An electromagnetic piston pump (PP) of 40 Watt (Reciprotor-Denmark) was used to provide air flow since it proved much more satisfactory than compressed air cylinders. Air flow and pressure were regulated by means of a precision valve (PV) (Negretti & Zambra, England), and controlled by an adequate air flow-meter (FM) and an open manometer (M). A constant air flow of 18 litres per minute at a pressure of 16 cm mercury were applied in all sprayings. A Mariotte bottle (MB) was used as the liquid reservoir.

The insecticide emulsion reached the liquid feed tube of the nozzle through rubber tubes connected to the two arms of an inverted T-shaped burette tubing.

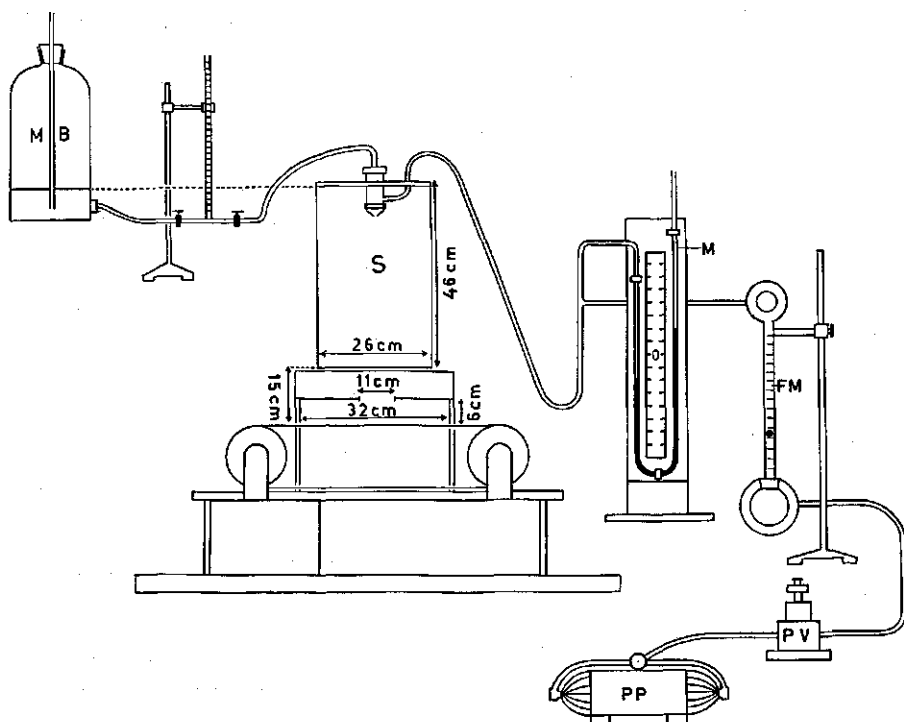


FIG. 1. Modified TEN HOUTEN & KRAAK vertical spraying apparatus (S), together with the piston pump (PP), precision valve (PV), flow-meter (FM), manometer (M) and the liquid reservoir (MB).

Both arms of the burette were provided with glass stopcocks by means of which the quantity of emulsion sprayed could be adjusted. An interchangeable tip of 1 mm inner diameter was used in all experiments. The leaves were sprayed one after the other without the use of a moving band or turning table. Where films of different densities were needed, the concentration of the insecticide in the water emulsion to be sprayed was changed, but the volume of the liquid, the area of the sprayed surface and the air pressure and flow were kept constant.

The leaves were placed on inverted Petri dishes with their upper surfaces exposed and were then stretched flat by means of leaden rings especially made for this purpose. These rings pressed the periphery of the leaves leaving the middle part flat and uncovered.

After pouring the water emulsion of the test insecticide into the reservoir of the spraying apparatus, the air compressor was started and the apparatus was adjusted and allowed to work for a while. The leaves were then sprayed one after another by placing them under a square 11×11 cm opening in the splash plate. Each leaf received 2 ml of emulsion. This was found to be the most efficient quantity giving complete coverage of the exposed part of the leaf without any excess. Sprayed leaves were then arranged on tables and left to dry at room temperature for six hours. At the end of the six hours, the

TABLE 1. Variability of fresh deposit of DDT emulsions during a representative spraying experiment

Disc no.	Weight of deposit in mg.	Disc no.	Weight of deposit in mg.	Disc no.	Weight of deposit in mg.
1	158	7	135	13	149
2	155	8	152	14	154
3	142	9	136	15	152
4	136	10	158	16	149
5	148	11	146	17	146
6	132	12	155	18	144

Average deposit per disc = 147 mg

S = 8.14

C.V. = 5.5%

lead rings were removed and each leaf was placed in a water-filled test tube with its stalk passing through a stopper.

In order to test the average quantity of fresh deposit per square unit area of sprayed surface in each experiment, a number (usually 20) of round filter paper discs, 7 cm in diameter, were also sprayed at the same time as, and alternating with, the test leaves. The filter paper discs were pinned on small wooden blocks, each in a Petri dish so that they did not touch the bottom or rim of the dishes. Before and immediately after being sprayed, each disc was weighed on a torsion balance. The difference between the two weights represented the weight of fresh deposit on the whole disc. Thus the fresh deposit per square centimeter could be found and from it the net residue of insecticide per unit area determined. It was possible in this way to measure the average residue of insecticide in each experiment.

The results of a representative experiment of this kind together with the standard deviation and coefficient of variation are given in table 1.

After each spraying the apparatus was thoroughly cleaned and rinsed with water and acetone.

5. BIOASSAY OF RESIDUES

The method applied by LOOSJES (1952) was used in the present work to determine the contact effect of residues on the test insect. The equipment and the different steps of this method are shown in plate 1. The procedure was as follows:

Special truncated porcelain cones with a diameter of 52 mm at the wider and 25 mm at the smaller opening were used to confine the test insects on the treated surfaces. The required number of these cones were prepared for use by slightly smearing the lower part of their inner walls with very small quantities of a high quality sewing machine oil. Preliminary tests had shown this oil to be harmless to the test insects. When this smearing was properly done, it was sufficient to prevent the test insects from crawling up the cones and escaping.

Ivy leaves carrying insecticide films to be tested were each placed on a separate, rectangular wooden plate bearing a suitable rectangular piece of rubber on its surface. A porcelain cone was then placed on each leaf with its smaller opening upwards. The cone could be fixed closely to the leaf surface by means of another rectangular wooden plate provided with a round hole in

its centre which fitted over the upper part of the cone and was held firmly against the lower plate by two rubber bands. When all the leaves had been fitted with cones, fifty *Calandra granaria* weevils were dropped on to each leaf through the upper opening of each cone. In DDT experiments the insects were exposed to residues for 24 hours, but in parathion experiments they were exposed to deposits for 3 hours only. At the end of the exposure period the weevils were transferred from the leaves to clean Petri dishes and provided with wheat for food. The weevils confined on each leaf were kept in one Petri dish.

The effect of insecticide residue was evaluated by means of mortality countings carried out twice for each batch of insects; on the fourth and seventh days in the case of DDT, and on the third and fifth days in the case of parathion. The time was counted from the end of the exposure period in both cases.

During the exposure period and later on when they were in Petri dishes the insects were always kept in constant temperature cabinets adjusted at 25°C and 50-60 % R.H.

Mortality countings were carried out by placing the insects on a warm plate (of constant temperature) and inspecting them individually. The weevils were assigned to one of the following three categories:

1. Normal (N)-Apparently normal and active insects in which no sign of abnormality whatsoever could be noticed.
2. Abnormal (A)-This group included all weevils which showed any sign of abnormality, regardless of the degree. Thus it included all the insects which could not be considered normal or dead.
3. Dead (D)-Those insects which failed to show any movement despite prodding.

After the first counting, the dead insects were discarded and only the insects of the first two categories were put back in the Petri dishes, together with food, and kept in the incubator till the second mortality counting.

A number of preliminary experiments showed that the figures provided by category 2 (abnormal insects) were not reliable since some insects of this group were able to recover while the rest either died or stayed abnormal for rather a long time. These experiments also showed that the values of the second mortality countings were more comparable and constant when separate experiments with the same concentration of either insecticide were compared. It is believed that the first mortality counting, being done after rather a short time from the end of treatment (exposure to residue), was not capable of giving reliable and representative figures for the total and final effect of any residue. This was especially true for residues in which the concentration of insecticide dropped to such a level that only low mortality ensued. As a result, all bioassay results mentioned in this work were based on the mortality figures of the second counting.

Each mortality figure in the bioassay was based upon a sample of eight treated leaves. As fifty weevils were exposed to the residue on each leaf, the number of weevils used in one sample amounted to 400. The size of this sample, with regard to both the number of leaves and also the number of test insects, was found to be reliable and adequately representative of the degree of effectiveness of any tested residue. The left half of table 2 shows the data of a second mortality counting, mean number of dead insects per leaf and the standard deviation for a sample of DDT-residue. The right half of the table presents the same data concerning a parathion sample.

TABLE 2. Variation in mortality within a sample of eight leaves treated with DDT or parathion

DDT sample				Parathion sample			
Leaf No.	N	A	D	Leaf No.	N	A	D
1	12	—	38	1	6	1	43
2	2	—	48	2	8	1	41
3	5	1	44	3	5	—	45
4	5	—	45	4	11	—	39
5	6	1	43	5	15	—	35
6	10	—	40	6	—	2	48
7	4	1	45	7	13	4	33
8	4	—	46	8	5	1	44
Total	48	3	349	Total	63	9	328

% mortality = 87.25%
Mean dead insects per leaf = 43.6
S = 3.25

% mortality = 82%
Mean dead insects per leaf = 41
S = 5.01

An equal number of untreated ivy leaves were tested simultaneously with each treated sample as a control. Such control samples were prepared and dealt with exactly in the same way as the treated samples, but were always kept in a separate incubator. The mortality figures of the control samples represented the natural mortality of the test insects together with the mortality caused by handling the insects. In all experiments, the mortality in the control samples was never more than 5 per cent and usually below 3 per cent. The percentages of mortality for the treated samples were corrected by means of the Abbott formula.

6. CHEMICAL ANALYSIS OF RESIDUES

In all experiments every sample included 16 leaves. Half this number was used in the bioassay and the other half provided the sample for the chemical analysis of the insecticide under investigation. Each sample for chemical analysis was prepared as follows:

By means of a special cutting device made of stainless steel a disc of 52 mm in diameter was cut from the central part of each of the 8 leaves. The discs of each sample were then stored in 75 ml of petroleum ether in a brown glass-stoppered bottle carrying date and code of the sample.

a. DDT analysis

The method of SCHECHTER *et al.* (1945) which was modified later by AMSDEN & WALBRIDGE (1954) was employed in the present work for the analysis of DDT residues.

The method of SCHECHTER *et al.* depends on the intensive nitration of DDT to polynitro derivatives and the production of intensive colours upon addition of methanolic sodium methylate to a benzene solution of the nitration products. The requirements and different steps of this method could be summarized as follows:

The petroleum ether extract of each sample was quantitatively transferred to

a 150 ml Erlenmeyer flask after being filtered through cotton wool and anhydrous sodium sulphate. All but 5 ml of the petroleum ether extract was then evaporated off on a gently warmed sandbath. The remainder being removed in an air stream. The residue was dissolved in 15 ml methanol and quantitatively decanted into a thick-walled centrifuge tube. The methanol was dried again on a waterbath and by means of an air stream. 5 ml of the nitrating acid were then added and vigorous nitration continued for 15 minutes at 100°C in a waterbath. The tube was then cooled to room temperature after which 30 ml of distilled water was first added and later 10 ml of 4 % potassium permanganate solution in water. The contents were transferred to a 125 ml separating funnel and peroxide solution carefully added until a clear colourless solution was obtained. Following this, 10 ml benzene was added and the funnel was then shaken vigorously for 1 minute. After the complete separation of layers the lower layer was drawn off and discarded. The remaining phase, the benzene, was washed once with water and as many times with 5 ml portions of sodium hydroxide solution as was required to obtain an alkaline reaction of the water-phase. The benzene phase was washed again with 5 ml water and twice with saturated sodium chloride solution (washing with sodium chloride solution was done only when there was no complete separation between the water and benzene phases). Finally the benzene layer was filtered through cotton wool and anhydrous sodium sulphate. The colour could then be developed by mixing 2 ml of the benzene solution with 4 ml of sodium methanolate solution in a 10 ml ground stoppered test tube and shaking vigorously. After 5 minutes the intensity of the blue colour was measured at 590 mμ in the spectrophotometer, using cuvettes with a 1 cm light path.

A standard calibration curve was first prepared by carrying out analyses of equal aliquots of plant extracts to which exactly measured quantities of the 25 % DDT e.c. had been added. The extinctions obtained were plotted against the quantities of DDT used. The resulting curve was a straight line. The range of concentrations used was 50–600 μg DDT per sample.

b. Parathion analysis

The widely used method described by AVERELL & NORRIS (1948) which was modified by GAGE (1950) and later by ZEUMER & FISCHER (1952) was used in this work for the estimation of parathion residues. This colorimetric method is based on the reduction of parathion with zinc to the amino compound. Diazotization of the amino compound and coupling with N-(1-naphthyl)-ethylene-diamine dihydrochloride produces an intense magenta colour.

The lack of stability of parathion and its quick decomposition, when exposed to weathering, into other compounds which might be more toxic has thrown suspicion on the ability of the method of AVERELL & NORRIS to detect all of the toxic residues of parathion. This point was investigated by FRAWLEY *et al.* (1958). After conducting a series of experiments, these authors concluded that when parathion was exposed to ultraviolet light for different periods the quantity of material reacting in the AVERELL-NORRIS chemical method decreased. They also found, however, that the toxicity of the material to flies appeared to decrease at about the same rate as the amount of material reacting in the AVERELL-NORRIS chemical method.

The *in vitro* anticholinesterase activity, on the other hand, was found to increase steadily. *In vivo* experiments revealed, however, that this increase was

less in ultraviolet-exposed samples of parathion than in unexposed samples and that the relative loss of anticholinesterase activity also closely paralleled the results obtained by the AVERELL-NORRIS chemical method. Thus the method proved to be reliable and comparable with biological tests on parathion residues.

The procedure is briefly as follows:

The petroleum ether extract of the treated sample of ivy leaves was quantitatively transferred to a 150 ml Erlenmeyer flask after filtration through cotton wool and anhydrous sodium sulphate. The bottle and filter were rinsed with 20 ml petroleum ether. The greater part of the solvent was evaporated off, the remainder being allowed to evaporate without heating. To the residue in the flask the following were added: 10 ml alcohol, 10 ml distilled water, 2 ml hydrochloric acid and 200 mg zinc dust. After fitting an air cooled condenser to the flask, the liquid was refluxed for 5 minutes. The air condenser and the walls of the flask were then rinsed with 10 ml water. After cooling, the liquid was filtered into a 50 ml graduated flask, the filter paper being rinsed with 10 ml water. 1 ml of sodium nitrite (p.a.) was then added to the filtrate and allowed to react for 10 minutes, after which 1 ml of ammonium sulphamate (p.a.) was similarly added and allowed to react for 10 minutes. On the addition of 2 ml of N-(1-naphthyl)-ethylenediamine dihydrochloride (p.a.), the colour was developed and measured at 550 m μ within 10 minutes from reaction.

A standard curve was prepared in a way identical to that described for DDT.

All chemicals and reagents used in the chemical analysis of residues of either insecticide were chemically pure unless otherwise stated. The colours were measured in a Beckmann spectrophotometer.

In the above procedures for determining DDT and parathion residues no attempt was made to free the petroleum ether extract from the plant pigments, oils, and waxes. This was due to the fact that the amount of impurities per sample was extremely small.

Owing to the fact that the plant samples carrying the DDT or parathion residues were left to stand in the solvent for some time, chemical analyses determined the total quantities of DDT or parathion both in and on the leaves.

A control analysis was always run on an untreated sample of ivy leaf discs side by side with each treated sample. The results of these control samples in terms of extinctions were applied, in experiments with either insecticide, as corrections to the extinction values of the corresponding treated samples.

III. FIELD EXPERIMENTS

1. LITERATURE

The data provided by the literature are arranged according to the following subjects:

- a. The persistence of DDT and parathion residues, particularly on plant surfaces, under field conditions, with a special consideration to the rates of loss.
- b-d. The effect of weathering upon insecticide deposit and the relative importance of temperature, rainfall and sunlight as regards the loss of residues in the open.

- e. Causes of loss of insecticidal activity other than weathering, e.g. penetration into plant tissue.

a. The rate of loss of residual effect

LINDGREN & BOYCE (1944) reported that in California DDT residues formed by spraying DDT-oilemulsion on citrus trees during winter time were effective for 60 days against the nymphs of California Red Scale, *Aonidiella aurantii* MASK. GUNTHER *et al.* (1946) stated that deposits varying between 12.3 and 37.2 μg per sq.cm on citrus leaves and fruits formed by spraying DDT dissolved in kerosene both with and without various auxiliary solvents had decreased steadily at an approximate rate of 0.25 μg per sq. cm. of leaf surface per day over an 86-day period. These tests were conducted in southern California in mid.-summer weather. The authors mentioned that heat and light induced decomposition of DDT were probably both involved. EDEN & ARANT (1948) reported that DDT deposits formed by dusting alfalfa plants decreased over 40 days of winter weathering and tended to become equal in all plots receiving DDT at the same rate, whatever the number of applications. The largest decrease occurred during the first ten days, probably as a result of shattering, rainfall or other mechanical processes. During the last 30 days the loss was probably due to decomposition. HADAWAY & BARLOW (1949) mentioned that a deposit formed by spraying DDT dissolved in equal parts of kerosene and cottonseed oil on avocado-pear foliage during summer time lost its effect almost completely after weathering for a week. SLOAN *et al.* (1951b) treated lettuce with DDT wettable powder or oil emulsion sprays at the rate of 1 and 0.75 pounds per acre. They also applied parathion wettable powder sprays at a rate of 0.075 pounds actual toxicant per acre. After 14 days weathering during July and August they found that DDT and parathion deposits had lost 94-95 % and 99 %, respectively, of their initial quantities. HOPKINS *et al.* (1952a) tested the persistence of aldrin, BHC, DDT, lindane and parathion applied as dusts on a crop of alfalfa during summer time. They found almost complete loss of all insecticides within thirty days from application. The growth of the crop was rapid and might have contributed to the loss. In a report from West Africa (1953) it is stated that 1 % emulsified solutions of DDT sprayed on the branch unions and along the twigs of cacao trees gave effective protection against *Sahlbergella singularis* HAGL. and *Distantiella theobromae* DIST. for 75 days. EBELING (1953) in Hawaii tested the ability of several insecticide residues applied on waxed cards to withstand weathering. The tests were made between November and March. He found that heavy parathion residues of 23.4 μg per sq. cm formed by wettable powder sprays, when protected from rain and sun, gave almost complete kill of the melon fly, *Dacus cucurbitae* COQ., as long as 126 days after application. When exposed to sun and rain during November and December parathion deposits lost much of their effectiveness within a week, whereas DDT deposits remained fairly effective for two weeks but lost all toxicity in three. After carrying out different series of experiments, EBELING *et al.* (1953) in a test conducted in Hawaii during early April showed that weathering for three days of DDT and parathion deposits formed on corn stalks by treatment with wettable powder sprays reduced the toxicity of both DDT and parathion deposits to the melon fly by 26 % and 84 %, respectively, as compared with their effectiveness after one day. In another experiment during late April, DDT was used at the rate of 2 and 4 lb per 100 gals, whereas parathion was applied at the rate of 0.125

and 0.25 lb per 100 gals. Treated corn foliage was weathered for 11 days. All treatments gave complete kill when tested one hour after application of the sprays. On the eleventh day, tests with DDT-treated leaves resulted in 61 % and 100 % kill, respectively, as compared with 0 % and 9 % mortality with the two parathion concentrations, respectively. BRUNSON & KOBLITSKY (1953) found in two separate experiments, that DDT sprayed on to peach foliage as wettable powder sprays applied at the same rate, was lost to the extent of 85 % and 53 % in three weeks of summer weathering. STOFBERG (1954) found in a test carried out in Eastern Transvaal in May 1953 in which young orange trees were treated with 2 % DDT as wettable powder and emulsion sprays, that protection against ants was afforded for 8 and 7 months, respectively. A 1 % DDT wettable powder or emulsion spray protected the trees for 5 months. DDT and some newer insecticides were tested against *Anthonomus eugenii* CANO. on caged pepper plants by ELMORE & CAMPBELL (1954). They found that the toxicity of 2 % DDT dust had decreased after ten days exposure to summer weather but not after five, whereas 1 % parathion dust was less toxic after five days and much less so after ten. WILSON (1955) reported that a technical DDT spray at the rate of 1 lb material in 2 U.S. gals diesel oil per acre during winter-time protected beet fields against *Circulifer tenellus* BAKER for 77 days after spraying. With faster growing varieties somewhat less protection was obtained. DI MARTINO (1956) mentioned that sprays with 0.025 % parathion or 0.25 % DDT in wettable powder or emulsified solutions applied during early winter in Sicily gave complete protection of citrus trees against *Empoasca decedens* PAOLI for 10 days in case of parathion and throughout the winter in case of DDT.

BOBB (1954), during summer, sprayed peach trees 2 or 3 times with parathion at the rate of 4.5 and 1.5 gals per tree. The sprays were formed of 1.5 and 4.5 lb of 15 % wettable powder parathion per 100 U.S. gals. Analyses of bark and leaf samples taken at weekly intervals showed that the initial residue was much greater on bark than on leaves and that residues sufficient to kill the crawlers of two different scale insects remained for a month on the bark as compared with less than a week on leaves. He also mentioned that rain had no significant effect on parathion residues. AYOUTANTIS *et al.* (1954) reported that emulsion sprays of 0.1 % parathion applied in early summer protected olive fruits against *Dacus oleae* GMEL. until picking time, and 0.03 % parathion provided protection for 65 days, while 0.015 % gave 40 % kill 45 days after spraying. CIAMPOLINI & TERROSI (1954) in experiments carried out on the coast of Tuscany during summer 1954, found that 0.1 % parathion emulsion spray applied to olive trees was still highly effective against *D. oleae* 10–15 days after treatment. BRAID & DUSTAN (1955) showed that sprays containing 1 and 2 lb wettable powder of 15 % parathion per 100 gals water on immature peaches applied during early summer resulted in initial residues of 30 and 70 µg toxicant per 100 sq. cm, respectively. The rates of decay of the residues were 2.2 and 4.2 µg per 100 sq. cm per day, respectively. These rates of loss were maintained for approximately 12–14 days, after which there was little further loss. WAITES & VAN MIDDELEN (1955) working under summer weather conditions in Florida found that parathion emulsion sprays at 4.4 oz. per acre applied to cabbage nine times, with or without DDT, gave residues of about 10–12 parts per million four hours after the last application. These deposits decreased by 85 % after 3 days and by 97 % after seven. On turnip tops treated three times with wettable powder sprays containing 1.76, 2.4 and 4.8 oz. parathion per acre, the residues

were 7.1, 15.1 and 31 p.p.m. four hours after the last treatment, but were reduced by 70–85 % after three days weathering and by 90 % after seven days.

The data reviewed above show a good deal of controversy as regards the duration of effectiveness and the rate of disappearance of either DDT or parathion. It is believed that most differences in results found by authors in different regions are due to the differences in the concentrations and doses used, the qualities of the many formulations applied, the nature and properties of the surfaces of the different crops on to which the toxicants were applied as well as the enormous variation in the intensity and severity of the different weather components prevailing at the location of the experiments. However, the long-lasting residual effect of DDT compares favourably with the short duration of parathion residues.

b. Effect of general weathering and temperature

Mechanical, physical and chemical losses are all included in the reduction of insecticides exposed to weather. The estimation of the separate effect of each weather factor is usually very difficult under field conditions, but the degree of confusion between these factors differs from one to the other. The action of rainfall and wind, which are generally considered as the most important mechanical factors in the disappearance of toxicant deposits may be confused, but it is not difficult to distinguish between their effects as they do not always coincide under normal field conditions. In contrast with this are the effects of heat and sunlight, which usually coincide. Exposing treated plants to field air temperature under shelters to avoid the effect of sunlight provides unreliable results. The elimination of sunlight in such experiments also results in a considerable decrease in plant surface temperature which is the actual temperature affecting the residues. Therefore, the exclusion of the effect of sunlight inevitably means a drop in the temperature examined in field experiments. Similar difficulties may be encountered in the study of other weather components.

Field experiments conducted by GAINES *et al.* (1948) showed that DDT dust was not effective in controlling thrips or harlequin bugs in certain years. They attributed this lowered effectiveness of DDT to the possible effect of high temperature and low humidity on the toxicant. The results of work conducted by GAINES & DEAN (1949 and 1950) and GAINES & MISTRIC (1951) showed that high temperature (32–38°C), relative humidity, rainfall, sunshine and dew are very important factors in reducing the toxicity of a number of organic insecticides. These authors also stated that emulsion sprays seemed to remain more toxic under certain field conditions than the dust formulations. TEOTIA & DAHM (1950) studied the loss of effectiveness of deposits of aldrin, chlordane, dieldrin, lindane and parathion on the basis of 50 mg toxicant per sq. ft. on unpainted and painted wooden panels. They found that weathering during the cooler months of the year when sunlight, precipitation and wind were the significant climatic factors, had accelerated the rate of degradation of the residual toxicity of all the insecticides tested. They concluded from laboratory experiments in which insecticide residues were applied to glass plates at the same dosage and exposed to certain combinations of temperature and relative humidity that high temperature combined with low humidity have a more deleterious effect on the residual toxicity against houseflies than low temperature with high humidity. DECKER *et al.* (1950) studied the rate of reduction of the toxicity of deposits from some organic insecticides including DDT and parathion. They

believed that, in the absence of rain, the rate of loss was correlated with the vapour pressure of the insecticide. Hence, the loss was activated by the increase of both temperature and wind. GAINES & MISTRIC (1952) reported that during a year when the daily temperatures were excessively high and the relative humidities low, high dosages of many organic insecticides were required to obtain high mortalities of the boll weevil, *Anthonomus grandis* BOH. It was necessary to use 4 or more times as much toxicant under field conditions as was needed under laboratory conditions to kill comparable percentages of weevils. They concluded that high temperature, sunshine, wide ranges of relative humidity, dew, or a combination of these factors greatly reduced the toxicity of the insecticides in the field. These workers also found that the toxicity of parathion residues was greatly reduced after 24 hours exposure to weather reaching a maximum temperature of 84–107°F (29–42°C). They considered that the loss was not only due to temperature but also to the other weather factors mentioned above. HOPKINS *et al.* (1952b) carried out a series of field experiments to estimate the relative importance of rain, sunlight and wind on the loss of DDT residue formed by treating a field of full-grown red clover with a 5 % DDT dust during two successive summers. Parallel tests to estimate the effects of the three possible combinations of any two of these factors as well as the effect of all the weather factors upon the insecticide residues were conducted at the same time. The initial deposit in all cases was 305 p.p.m. The average total quantity of rain was 1.64 inches, the period of sunshine amounted to 160.09 hours and the average wind speed was 6.5 m.p.h. at the end of the experiment. The plot exposed to all weather components resulted in a loss of 90 % of the initial residue after 7 days, 97 % after 14 days and 98 % after 24 days. They found that either rain or wind caused a rapid decrease of the deposit soon after application, but was much less effective during the later weeks. The interaction of rain and wind was slight, perhaps because either tends to remove mechanically the same less adherent material. They also showed that sunlight was not a speedy factor in the disappearance of DDT deposits, but that it was highly effective in eliminating the residues applied to red clover over long periods. This slow but continuous action of sunlight was explained by the ability of ultraviolet light to eliminate the loosely adhering as well as the persistent residues. The combined effects of rain and sun upon the deposits were approximately additive. The effect of all three weather factors together was somewhat larger than that of any two but not much larger than that of sunlight combined with either rain or wind. EBELING (1953) showed that rainfall was less important than sunlight and high temperature in the weathering of parathion wettable powders applied to waxed cards. He found that 5.23 inches of accumulated rain during a 5-day period with a mean maximum temperature of 24°C and a mean minimum temperature of 18.5°C did not affect the toxicity of parathion residues formed on cards by an 0.24 % wettable powder spray. However, the same period of rain resulted in the almost complete removal of effectiveness of parathion deposits on the upper sides of corn leaves in a neighbouring field. EBELING *et al.* (1953) mentioned that laboratory cage tests of foliage and fruits obtained from sprayed fields showed that the effectiveness of parathion was more impaired by rain than that of DDT, methoxychlor, lindane, chlordan, heptachlor, aldrin, dieldrin and EPN.

c. Effect of rainfall

GUNTHER *et al.* (1946) reported that two heavy rains during the latter part of Meded. Landbouwhogeschool, Wageningen 61 (6), 1-64 (1961)

a 3-month period of weathering did not have any effect on DDT deposits. GINSBURG *et al.* (1949) found that the amount of rainfall had a bearing on the harvest residue, but that the relationship was not always proportional. HADAWAY & BARLOW (1949) mentioned that 0.87 inches of rain during the first 24 hours after an application of DDT wettable powder spray washed off most of the deposit from exposed plants. KEISER & HENDERSON (1951) applied DDT as emulsified solutions and as suspensions to soy beans at the rate of 5 and 10 lb of actual toxicant per acre. They found that there was no difference in residue loss in the absence of rain, but the emulsions were more persistent after rains. SLOAN *et al.* (1951a) noticed that the effect of rain soon after application of toxicant was of great importance as regards the rate of disappearance of either DDT or parathion residues. They found that initial residues of 79.9 and 63 p.p.m. DDT on lettuce formed by emulsion sprays, dropped to 51 and 42.7 p.p.m., respectively, within 24 hours and after 0.22 inches rain. They showed that 0.57 inches of rain decreased parathion residues, applied as wettable powder sprays on lettuce from 1.85 to 0.31 p.p.m. within one day. However, this observation was less consistent with parathion than with DDT. They attributed this irregular consistency with parathion to the fact that parathion residues were so much reduced by other causes that the effect of rain could easily have been obscured. MISTRIC & MARTIN (1956a) showed that one-half inch of simulated rain applied immediately after treating cotton plants with parathion as an emulsion spray at the rate of 0.04 pound of actual toxicant per acre was slightly more detrimental to the effectiveness of residues than rain occurring 24 hours after treatment.

d. Effect of sunshine

GAHAN *et al.* (1945) found that deposits of DDT on wood at a density of 50 mg per sq. ft., whether applied as kerosene solutions, as emulsions or as suspensions, remained effective against mosquitos after exposing to sunshine for 24 weeks. GUNTHER & TOW (1946) mentioned that under field conditions DDT may disappear rapidly leaving a residue of almost non-insecticidal dehydrohalogenation products. LINDQUIST *et al.* (1946) carried out tests in which DDT deposits of different formulations including emulsions, suspensions and solutions in kerosene with various auxiliary solvents, applied at the rate of 200 mg per sq. ft. on different surfaces, were exposed to sunlight for 135 hours during 18 days. The results of these tests showed that DDT residues of all formulations suffered appreciable reduction in toxicity against houseflies. Deposits from suspensions and emulsions persisted longer than did deposits from solutions containing non-volatile solvents. The latter were also less persistent than residues from solutions containing volatile auxiliary solvents. These results indicate that DDT is more rapidly decomposed by sunlight when exposed in solution than as a solid deposit. CHISHOLM & KOBLITSKY (1947) reported that technical DDT dusted on glass slides, at the rate of about 3 mg per sq. cm., suffered a loss of 20.2–22.2 % as a result of exposure for 64 hours to mid-summer sunlight and temperature. VANDRAMINI (1947) found that exposure of DDT deposits to high intensities of ultraviolet radiation for one hour caused the same loss of effectiveness to *Drosophila melanogaster* as 30 days of natural light. CHISHOLM *et al.* (1949) showed that the toxicity of DDT deposits from emulsion formulations was reduced to a very low level after exposure for only 32 hours to sunlight. Deposits from wettable powder sus-

pensions exposed for 64 hours to sunlight showed high toxicity in the absence of alkaline or iron containing diluents, such as limonite, but suffered severe losses in the presence of these substances.

e. Penetration of insecticides into plant tissue

The attenuation of insecticide residues by growth becomes negligible when the toxicants are applied to full-grown plants. In such conditions, as in the case of the present tests, most of the loss is attributed to weathering, but a part of the formed residue may be lost by penetration through the plant tissues.

EBELING (1945) stated, "The addition of 1 % or 2 % of aluminium stearate to the DDT solution in kerosene largely reduced the penetration of the solvent and DDT into the lower surface of an orange leaf, while the DDT solution without the aluminium stearate completely penetrated into the leaf in less than ten minutes. Microphotographs showed a much heavier deposit of DDT crystals in the case of solutions containing aluminium stearate." GUNTHER *et al.* (1946) mentioned that as the kerosene-DDT spray was applied to citrus trees, much of it penetrated into the leaf tissues almost immediately carrying the dissolved DDT along with it. Within the 24 hours after spraying, the kerosene plus DDT slowly re-issued from the leaf to its surface whereupon evaporation of the solvent left a deposit of the DDT. The chemical analyses of residues from sprays containing only kerosene and DDT, collected by stripping citrus leaves with a solvent, showed an increase of DDT up to 40 % more than the original deposit which was determined on samples collected at cessation of drip of the spray. STAFFORD & HINKLEY (1946) reported that olive oil obtained from olives which had been sprayed with a mixture of a DDT wettable powder and a light soluble oil contained DDT. SYMES *et al.* (1948) found a slight penetration of DDT into leaves when the insecticide was applied as a solution in a heavy naphtha solvent and emulsified in water. However, when DDT was applied as 5 % solution in diesel oil, 30 % of the DDT could not be removed by washing with benzene, but was recovered by exhaustive extraction of the leaves with benzene. HADAWAY & BARLOW (1949) also observed penetration of DDT into leaves after spraying avocado-pear foliage with a 5 % DDT solution in equal parts of kerosene and cottonseed oil. MARTIN & BATT (1954) showed that apple foliage sprayed with DDT emulsion or suspension retains about 15 % of the DDT. This "internal" quantity was not removed by simple washing with carbon tetrachloride and persisted even when most of the external deposit had been lost by weathering for 70 days. BURT & WARD (1955b) using cabbage leaves, found similar results. Two weeks after treating the leaves with a crystalline suspension of DDT at a rate of 4 μ g per sq. cm., the "internal" DDT rose to a maximum value of about 10 % of the amount originally sprayed. Thereafter it remained constant while the external deposit declined. Leaves washed with benzene were found to contain DDT which could only be recovered by drying and extracting with solvent. They mentioned that an insecticide can penetrate into the tissue of a plant by two ways. When the medium containing the toxicant can enter the leaf it carries the insecticide with it, or in other cases, the toxicant after the evaporation of the medium diffuses through the cuticle or the stomata into the plant tissue. FOGG (1948a, b) showed that the aqueous media are unable to pass through the stomata, even in the presence of wetting agents, whereas oils readily pass through into the intercellular spaces of the foliage.

The penetrating capacity of parathion into plant tissues was studied by

LÜDICKE (1949a, b). He found that the application of diluted emulsions of 0.05 % E 605 f to spots on the leaf of the hostplants of partly developed larvae of various leaf mining *Diptera* caused almost complete mortality of larvae. The material was applied to spots directly above or below the larvae or round them in such a way as to isolate them from the rest of the leaf. Treating the leaves with the insecticide as spray also killed the larvae mining in them. D'AUSILIO (1951) conducted tests on the ability of parathion sprays to penetrate into olives and kill the larvae of *Dacus oleae* GMEL. within them. Infested olives were sprayed on two different sides with three different concentrations of parathion; 0.25 % and 0.1 %, prepared from two different wettable powders and 0.05 % diluted from an emulsified concentrate. These treatments gave 41 %, 44 % and 22 % mortality of the larvae inside the olives. Dissection of olives six days after treatment showed that the larvae were killed when they were moving towards the surface after being full-fed. He concluded from these results that the parathion did not penetrate very deep into the olives. CIAMPOLINI (1952) found high mortalities of different instars of *D. oleae* larvae in olive fruits picked from trees treated with 4 % parathion dust or 0.4–0.6 % parathion suspensions. He concluded that parathion had penetrated into the fruits and acted then as a stomach poison against the larvae. CIAMPOLINI & TERROSI (1954) mentioned that 0.1 % parathion emulsion spray caused 59.5 % egg mortality and 87 % kill of medium sized larvae within 3 days from application. The mortality of larvae reached 99 % within 7–8 days. The results indicated that parathion penetrates into olive fruits and acts as stomach poison.

The results of the work reviewed above indicate that either DDT or parathion are capable of penetrating into the plant tissue when applied to foliage, although it seems that the penetrating capacity of DDT when applied in water media is smaller than that of parathion.

2. EXPERIMENTAL

This group includes all experiments designed to expose the residues to weathering. The purpose was to follow the rate of disappearance of residual effect biologically as well as chemically by means of daily samples. The second aim was to try to evaluate the relative contributions of temperature, rainfall and sunshine towards the disappearance of either of the insecticide residues.

The sprayed leaves, provided with test tubes filled with water, were transferred to the field where they were fixed by means of their glass tubes to specially designed wooden trays placed at 80 cm above ground surface at an angle of 45° (plate 2). The leaves were pinned at their tips to the trays so that the least part of each leaf touched the wood. All the wooden trays were placed 60 cm from each other facing the south-western direction.

With the exception of the first DDT experiment, DDT and parathion experiments in 1960 were run simultaneously so that their residues were exposed, as much as possible, to the same weather conditions. Experiments with DDT always started two days in advance of those with parathion. A number of untreated leaves were also exposed in the same way to weathering to provide the control samples.

Each field experiment consisted of seven samples. In all the field and laboratory tests an initial sample was taken six to eight hours after spraying to determine the initial dose as indicated by chemical analysis and bioassay.

Except for the first field experiment with DDT, the concentrations of toxicants in the sprayed emulsions were chosen to give an initial mortality exceeding 90 % but never reaching 100 %. In this way it was hoped to obtain a significant decrease in mortality by the loss of comparatively small quantities of residue. This also avoided using concentrations too low to be comparable with practice.

Temperature and rainfall were continuously recorded during all field experiments. Recording of air temperature was made by means of the Speedomax automatic temperature recorder. Measurements of air temperature were made by exposing a thermocouple at a height of 80 centimeters above the ground surface. The surface temperature of leaves was determined by fixing two thermocouples closely to the surface of two different leaves. Measurements were made on a reference-point basis by keeping a special thermocouple inside a thermos bottle containing crushed ice and provided with a long-necked thermometer. The average maximum and minimum temperatures of both air and leaves were computed from the recording sheet of the apparatus.

Rainfall was measured daily by means of an appropriate recording rain-meter, built according to the system designed by the Royal Netherlands Meteorological Institute (K.N.M.I.).

Data concerning the periods of sunshine for each day were obtained from the weather station connected to the Laboratory of Physics, of the University of Wageningen, at about one kilometer from the experimental field. The records provided the number of minutes of sunshine per hour between 4 a.m. and 8 p.m. each day. This information has been rounded here to number of hours of sunshine per day.

In 1959, preliminary experiments were conducted in which maximum leaf surface temperature and rainfall were recorded daily.

3. RESULTS

a. Preliminary experiments in 1959

During the warm and dry summer of 1959, preliminary experiments were conducted with relatively high doses of insecticides averaging 48 $\mu\text{g}/\text{cm}^2$ for DDT and 4.2 $\mu\text{g}/\text{cm}^2$ for parathion.

The following results, which have been published briefly (FAHMY, 1960) may be mentioned.

In the DDT experiment rainfall was less than 9 mm, distributed over the first five days. Maximum leaf surface temperature did not exceed 29°C. Within this period, the DDT residue diminished for more than 80 % after which it remained stable for the next three rainless days. Mortality remained at the 98–99 % level, due to the high initial dose.

In the two parathion experiments, maximum leaf temperatures were up to 35°C. Rainfall occurred only during the first experiment, but only after the residue had decreased with more than 90 %. In both experiments, this decrease already occurred after 24 hours. After this, 4–5 % of the initial residue still remained chemically detectable, decreasing only slightly during the next 3–4 days. Mortality in the contact bioassay diminished to zero in 48 hours. It was concluded that the remaining parathion residue was located inside the leaf tissues.

The results of the two parathion experiments were almost identical, showing the reproducibility of the method (FAHMY, 1960, figures 4 and 5).

b. Experiments in 1960

During the spring and early summer of 1960, field experiments were carried out under recorded climatic conditions as described in chapter III, 2. The experimental period was characterized by rather limited rainfall.

TABLE 3. Experiment A. Effect of weathering for six days upon DDT residues as detected chemically and biologically by daily samples, together with the daily records of maximum and minimum air and leaf surface temperatures, rainfall and sunshine.

Number of days weathered	µg per leaf disc (21.25 cm ²)	µg per cm ²	% mortality	% decrease in residue	Daily % decrease in residue	% decrease in mortality	Max. air temperature °C	Min. air temperature °C	Max. leaf surface temperature °C	Min. leaf surface temperature °C	Rainfall in mm	Sunshine in hours
-	11	0.517	53	-	-	-	23.0	10.5	24.9	10.0	0.2	9.25
1	9.5	0.447	17	13.6	13.6	67.9	32.7	8.8	35.8	9.4	18.5	2.6
2	8	0.376	3.5	27.3	13.7	93.4	23.8	7.6	29.5	8.6	-	3.0
3	7	0.329	1.5	36.4	9.1	97.2	24.4	14.0	30.7	13.3	0.2	2.75
4	4.25	0.200	0	61.4	25.0	100	23.3	8.4	28.0	8.0	-	3.0
5	4.5	0.211	0	59.1	+2.3	100	15.2	7.0	16.8	7.6	1.4	0
6	4	0.188	0	63.6	4.5	100	-	-	-	-	-	-

TABLE 4. Experiment B. For explanation see table 3.

Number of days weathered	µg per leaf disc (21.25 cm ²)	µg per cm ²	% mortality	% decrease in residue	Daily % decrease in residue	% decrease in mortality	Max. air temperature °C	Min. air temperature °C	Max. leaf surface temperature °C	Min. leaf surface temperature °C	Rainfall in mm	Sunshine in hours
-	24.5	1.152	92	-	-	-	24.7	7.2	27.7	6.9	-	5.16
1	15.5	0.729	61	36.7	36.7	33.7	28.1	9.0	35.0	9.0	-	11.0
2	10.25	0.482	14	58.2	21.5	84.8	30.9	10.6	38.7	10.2	-	12.25
3	8.75	0.411	6.5	64.3	6.1	92.9	32.8	13.6	37.2	12.9	-	11.75
4	7.5	0.352	2.5	69.4	5.1	97.3	34.0	13.7	40.3	13.3	-	11.0
5	7	0.329	1	71.4	2.0	98.9	33.7	11.9	39.6	11.2	9.8	12.9
6	6.5	0.305	0	73.5	2.1	100	-	-	-	-	-	-

TABLE 5. Experiment C. For explanation see table 3.

Number of days weathered	µg per leaf disc (21.25 cm ²)	µg per cm ²	% mortality	% decrease in residue	Daily % decrease in residue	% decrease in mortality	Max. air temperature °C	Min. air temperature °C	Max. leaf surface temperature °C	Min. leaf surface temperature °C	Rainfall in mm	Sunshine in hours
-	23	1.082	91	-	-	-	28.3	11.1	31.4	10.2	-	6.0
1	13.75	0.647	53	40.2	40.2	41.8	34.5	15.4	39.5	14.4	-	12.0
2	9.25	0.432	8	59.8	19.6	91.2	28.5	15.8	32.4	14.9	-	3.65
3	8.25	0.388	3.5	64.1	4.3	96.2	36.3	16.2	41.7	15.5	-	12.8
4	7	0.329	1.5	69.6	5.5	98.4	34.9	14.2	40.5	14.0	-	13.75
5	6	0.282	0	73.9	4.3	100	23.5	10.1	26.8	9.8	-	2.8
6	6.25	0.294	0	72.8	+1.1	100	-	-	-	-	-	-

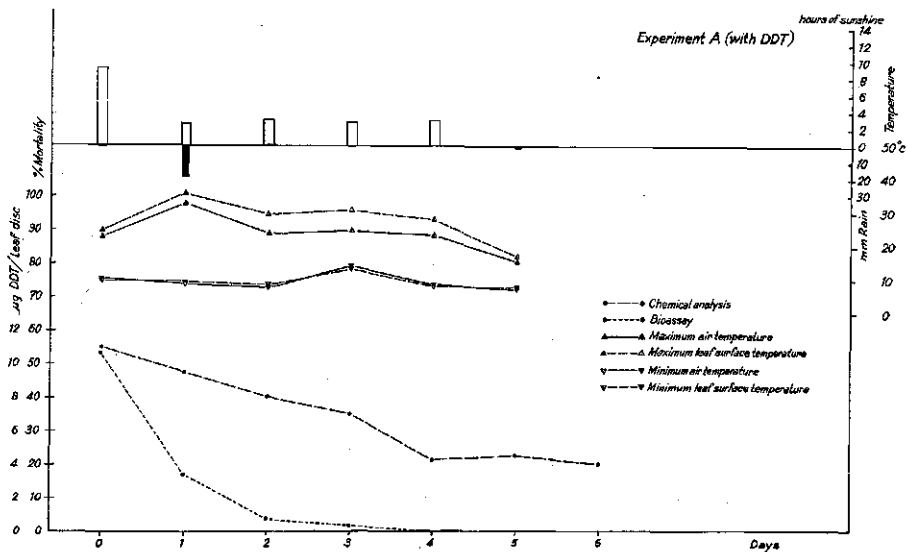


FIG. 2. DDT residues weathered for six days in experiment A as detected chemically and biologically by daily samples, together with the daily records of air and leaf surface temperature ($^{\circ}\text{C}$), rainfall (mm) and sunshine (hours).

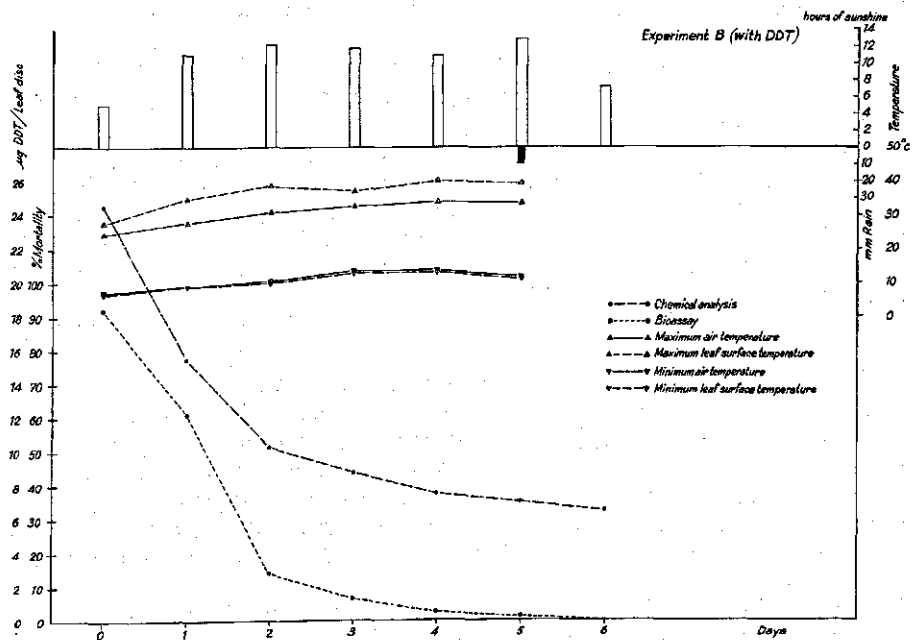


FIG. 3. Experiment B, for explanation see fig. 2.

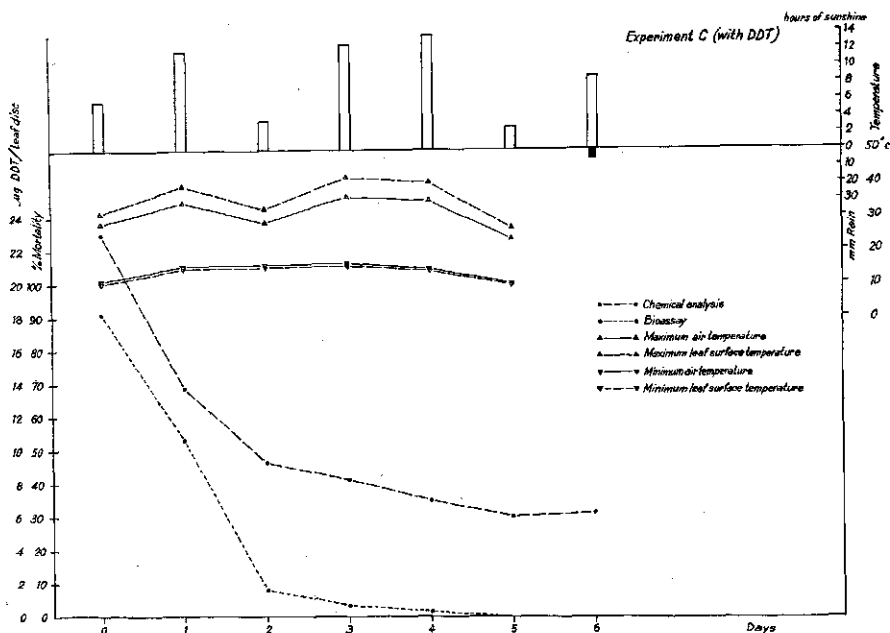


FIG. 4. Experiment C, for explanation see fig. 2.

During experiment B, C, D and E the sky was mostly clear, and temperature was relatively high for local conditions, reaching maxima up to 36.3°C.

α. DDT residues: The results of the three field experiments with DDT in 1960 are given in tables 3, 4 and 5 and figures 2, 3 and 4. These experiments will be referred to as tests A, B and C.

11.0 µg DDT per leaf disc (21.25 sq.cm), as initial residue, were sufficient to give 53 % mortality in test A, whereas the third sample of test B with 10.25 µg per leaf disc caused only 14 % kill. Again 9.25 µg DDT per leaf disc recovered from the sample taken after two days weathering in test C resulted in 8 % mortality. Apparently, the effectiveness of the residue per unit weight diminished quickly during weathering.

The sudden loss of 2.75 µg DDT per leaf disc on the fourth day of weathering, is remarkable.

The results of experiments B and C are almost identical. The initial residue of either test was reduced by 36.7 % and 40.2 %, respectively, after being weathered for one day. About 60 % of the deposits disappeared within 48 hours weathering. The rate of residue loss was reduced after two days weathering. In all DDT field tests the residues lost their effectiveness completely after being reduced to less than 7 µg per leaf disc which was reached by the loss of about 38 % in test A and about 70 % in both tests B and C of the initial deposits.

β. Parathion residues: Tables 6 and 7 and figures 5 and 6 present the results of the two field experiments with parathion residues. These experiments will be referred to as experiments D and E.

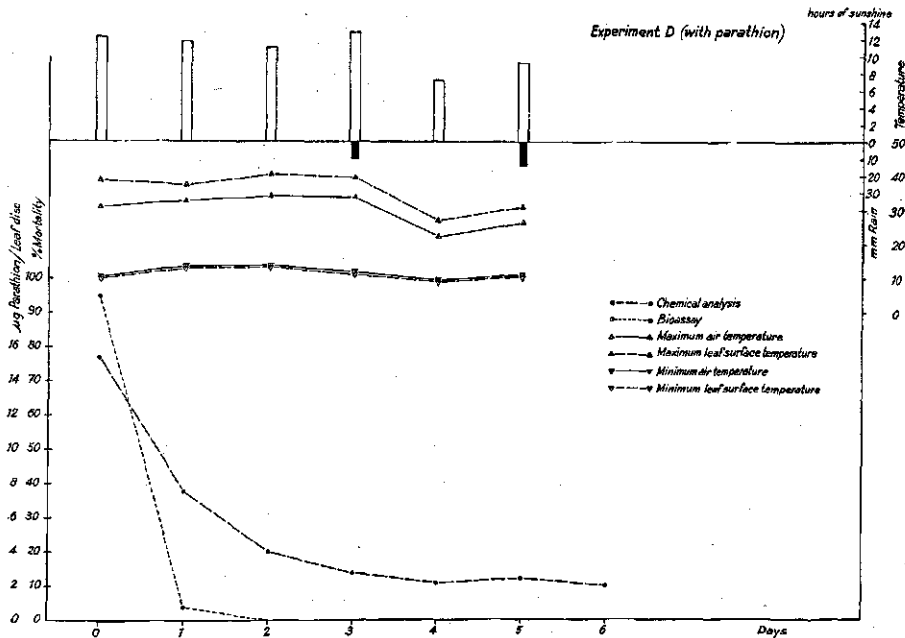


FIG. 5. Parathion residues weathered for six days in experiment D as detected chemically and biologically by daily samples, together with the daily records of air and leaf surface temperatures ($^{\circ}\text{C}$), rainfall (mm) and sunshine (hours).

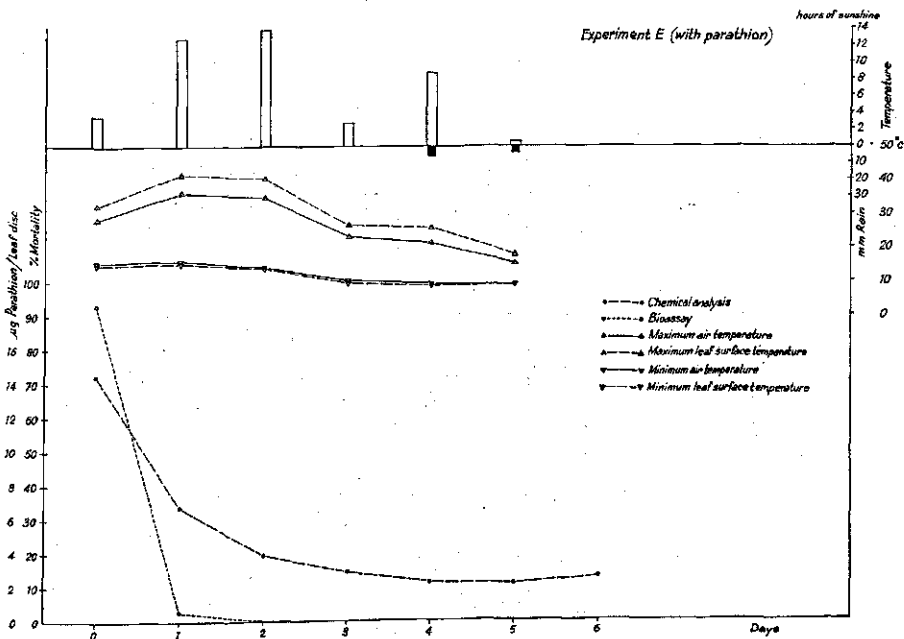


FIG. 6. Experiment E, for explanation see fig. 5.

After weathering for two days, the residues of either experiment failed to cause any mortality. The results of both tests indicate that a parathion deposit of less than 6 μg per leaf disc could effect no kill to the test insects.

The initial residues in test D and E suffered a reduction of 51.3 % and 53.5 %, respectively, after weathering for only 24 hours as compared with 87 % and 82.8 % loss after six days. The initial deposit of experiment D was reduced by 4.2 % more than in test E. The exposure to weather during the last five days of each experiment reduced the initial residues by only 35.7 % in experiment D and 29.3 % in experiment E. The relative effect of weathering upon the residue loss per day became very low when the residues were reduced to the level of about 3 μg parathion per leaf disc.

4. DISCUSSION

The results of the experiments reviewed under "literature" show a general agreement as regards the effects of weathering and the relative importance of the major weather components concerning the loss of insecticide deposits. All insecticides, which may largely differ in their stability, suffer considerable losses

TABLE 6. Experiment D. Effect of weathering for six days upon parathion residues as detected chemically and biologically by daily samples, together with the daily records of maximum and minimum air and leaf surface temperatures, rainfall and sunshine.

Number of days weathered	μg per leaf disc (21.25 cm^2)	μg per cm^2	% mortality	% decrease in residue	Daily % decrease in residue	% decrease in mortality	Max. air temperature $^{\circ}\text{C}$	Min. air temperature $^{\circ}\text{C}$	Max. leaf surface temperature $^{\circ}\text{C}$	Min. leaf surface temperature $^{\circ}\text{C}$	Rainfall in mm	Sunshine in hours
—	15.4	0.724	95	—	—	—	30.9	10.6	38.7	10.2	—	12.25
1	7.5	0.353	4	51.3	51.3	95.8	32.8	13.6	37.2	12.9	—	11.75
2	4	0.188	0	74.0	22.7	100	34.0	13.7	40.3	13.3	—	11.0
3	2.75	0.130	0	82.1	8.1	100	33.7	11.9	39.6	11.2	9.8	12.9
4	2.15	0.101	0	86.0	3.9	100	22.2	9.6	26.9	9.0	—	7.25
5	2.4	0.113	0	84.4	+1.6	100	26.3	10.9	30.7	10.5	14	9.3
6	2	0.094	0	87.0	2.6	100	—	—	—	—	—	—

TABLE 7. Experiment E. For explanation see table 6.

Number of days weathered	μg per leaf disc (21.25 cm^2)	μg per cm^2	% mortality	% decrease in residue	Daily % decrease in residue	% decrease in mortality	Max. air temperature $^{\circ}\text{C}$	Min. air temperature $^{\circ}\text{C}$	Max. leaf surface temperature $^{\circ}\text{C}$	Min. leaf surface temperature $^{\circ}\text{C}$	Rainfall in mm	Sunshine in hours
—	14.5	0.682	93.5	—	—	—	28.5	15.8	32.4	14.9	—	3.6
1	6.75	0.317	3	53.5	53.5	96.8	36.3	16.2	41.7	15.5	—	12.8
2	3.9	0.183	0	73.1	19.6	100	34.9	14.2	40.5	14.0	—	13.75
3	2.9	0.136	0	80.0	6.9	100	23.5	10.1	26.8	9.8	—	2.8
4	2.25	0.106	0	84.5	4.5	100	21.5	9.2	26.0	8.8	6	8.8
5	2.15	0.101	0	85.2	0.7	100	15.7	9.0	18.0	9.0	3.4	0.6
6	2.5	0.117	0	82.8	+2.4	100	—	—	—	—	—	—

by exposure to general weathering. This could be explained by the fact that "general weathering" means a complex of effects resulting from many factors which vary in the nature of their action. A stable synthetic toxicant may be resistant to many of these factors, but surely not to all. Thus by exposing this insecticide to general weathering, it will suffer from some or few adverse factors and the result will be a certain degree of loss. The extent of loss, within a certain span of time mainly depends on the degree of stability of the tested material together with the degree of severity of the adverse factors.

As stated before the separate effect of each weather factor when residues are exposed to general weathering cannot easily be ascertained, but in some cases these effects are clearly demonstrated.

a. Rates of loss

A comparison between the rate of loss of DDT and parathion deposits, as indicated by the results of experiments B, C, D and E (tables 4, 5, 6 and 7), shows that the parathion residues lost about 52 % of their amounts within 24 hours weathering as compared with about 38 % loss in DDT residues for the same period. The per cent final loss of DDT after six days weathering was almost equal to the loss of parathion after only 48 hours weathering, but the final loss of parathion after six days was only 10-14 % higher than that of DDT. This shows that the rate of loss of parathion was extremely high during the first two days of exposure to weather but diminished greatly after that. The rate of loss of DDT was also high in the first two days, but less than that of parathion and thereafter, the decrease in the rate of loss was rather gradual. DECKER *et al.* (1950) found considerable loss of parathion, lindane, aldrin, chlordan, dieldrin, toxaphene and DDT residues within the first 24 hours after application to various crops and trees. They also reported that sub-threshold quantities of DDT and toxaphene persist for very long periods which indicates that at some stage after application the rate of loss of most insecticides becomes extremely low. The decrease in the rate of loss with either toxicant during the later days of weathering may be due to several factors such as the presence of most of the remaining material inside the plant tissue where it is protected from weathering and the formation of a protective layer over the undecomposed insecticide by the already decomposed material.

b. Effect of temperature

TABLE 8. The general records of air temperature, rainfall, sunshine and leaf surface temperature for the six-day periods of each of the three field tests with DDT.

Code of test	Aver. max. air temp. °C	Aver. min. air temp. °C	Total rain in mm	Total sunshine period in hours	Aver. max. leaf surf. temp. °C	Aver. min. leaf surf. temp. °C
A	23.7	9.4	20.3	20.6	27.6	9.5
B	30.7	11.0	9.8	64.05	36.4	10.6
C	31.0	13.8	0	51.0	35.4	13.1

The average maximum and minimum air temperature, the total amount of rain, the total period of sunshine and the average maximum and minimum leaf surface temperature for the six-day weathering period of each of the three field experiments with DDT are given in table 8. The same data concerning the two parathion experiments are shown in table 9.

TABLE 9. The general records of air temperature, rainfall, sunshine and leaf surface temperature for the six-day periods of each of the two field tests with parathion.

Code of test	Aver. max. air temp. °C	Aver. min. air temp. °C	Total rain in mm	Total sunshine period in hours	Aver. max. leaf surf. temp. °C	Aver. min. leaf surf. temp. °C
D	30.0	11.7	23.8	64.45	35.5	11.2
E	26.7	12.4	9.4	42.35	30.9	12.0

It is shown in table 8 that the average maximum leaf surface temperature was 3.9–5.7°C higher than that of air during the DDT tests whereas table 9 shows that during the parathion experiments the average maximum leaf surface temperature was 4.2–5.5°C higher than that of air. In both cases the average minimum leaf surface temperature was slightly lower than that of air (less than 1°C). These records indicate that the residues applied to the leaves were actually affected by an average maximum temperature 4–5.5°C higher than that of the air and an average minimum temperature almost equal to that of the air.

The effect of temperature on the loss of insecticide deposits is shown by the difference in DDT residue loss between experiment A and either tests B or C. Table 8 shows that the average maximum air and leaf surface temperatures in test A were about 7°C and 8.5°C lower than the corresponding averages in either test B or C. The final per cent loss of residue after six days weathering was 63.64 in test A as compared with 73.5 and 72.8 for tests B and C, respectively. The decrease in residue loss in test A as compared with the latter two tests, amounting to about 10 %, is mainly attributed to the mild temperature and more cloudy periods during that test. Figure 2 shows that the initial residue in test A decreased gradually from 11 to 7 µg per leaf disc (a loss of 36.4 %) during three days weathering, but suddenly lost 2.75 µg (a loss of 25 %) during the fourth day only. This is very probably not due to rainfall, as is argued in chapter III, 4, c. Temperature records reveal that the maximum air temperature for that day was only 0.6°C higher than the previous day and even 8.3°C lower than two days before. On the other hand, the minimum air temperature of the fourth day was 14°C or 6.4°C higher than the previous day and on the whole higher than the minimum temperature of any other day during this experiment (the average minimum air temperature for the other five days of this test is 8.4°C or 5.6°C lower than that of the fourth day only). The loss of residue after the second day of this test amounted to 1.5 µg DDT per leaf disc. The maximum leaf surface temperature for that day was 35.8°C, whereas the maximum leaf surface temperature of the fourth day reached 30.7°C. Thus the difference in the actual maximum temperature to which the residues were exposed during these two days was only 5.1°C. On the other hand the minimum leaf surface temperature of the fourth day was about 4°C higher than that of the second day. This means again that the deposits were exposed during the fourth day of weathering to a higher mean temperature than they did during the second day. The periods of sunshine during the two days were comparable, 2.6 hours and 2.75 hours for the second and fourth days, respectively.

Other evidence of the importance of the duration of moderately high temperature was obtained in tests D and E. The per cent loss of parathion residue after one day weathering in both tests D and E were almost equal as shown in tables 6 and 7. Figures 5 and 6 show that the maximum air and leaf surface tempera-

ture for the first day in test D were 30.9°C and 38.7°C, respectively, or 2.4°C and 6.3°C higher than the corresponding temperatures of the first day in experiment E. In contrast, minimum air and leaf surface temperatures for the same day in test D were 10.6°C and 10.2°C, respectively, or 5.2°C and 4.7°C lower than the corresponding temperatures in test E. The two days were dry and the sun was shining for 12.25 hours that day in test D against 3.6 hours of sunshine during the first day of test E. The higher minimum temperature in test E as compared with test D also means that although the maximum air and leaf surface temperatures were lower in this day than in the first day of test D, yet the temperature remained high for a longer time during the first day of test E than it did in test D. Judging by the actual loss of parathion after the first day of weathering in both tests D and E, it could be concluded that the duration of a moderately high temperature for a longer time in test E had compensated for the increase in the maximum temperatures and for the longer period of sunshine in the first day of experiment D.

These findings may indicate that, under field conditions, when the temperature is high enough to cause a rapid loss of deposit (above 20°C) and within the normal temperature range (20–30°C), the destructive effect of temperature upon insecticide residues is determined by the product of temperature and exposure time and not only by temperature maxima.

c. Effect of rainfall

Rainfall failed to show any serious effect on residues through the present experiments with either insecticide. The quantities of rain in all the tests varied between 0.2 and 18.5 mm per day, but in all cases there was no special loss following rainfall that could demonstrate its effect. The only experiment in which rainfall occurred within the first 24 hours of weathering was test A (fig. 2) but the quantity, amounting to 0.2 mm, was too small to show any effect and within the second day of the same test there was 18.5 mm rain. Strikingly enough the residues suffered a loss of 1.5 µg per leaf disc per day in both cases which shows the extremely unimportant effect of rain on DDT residues tested in the present experiments. There was another 0.2 mm rain during the fourth day of test A, the same day characteristic of its high minimum temperature, but by no means the enormous loss of 2.75 µg per leaf disc detected in the sample of the following day could be attributed to this quantity of rain since 18.5 mm rain had a negligible effect two days before. During the other two field experiments with DDT the rainfall never occurred before the fourth day of weathering, and in all cases it had a negligible effect on the decrease of residues, the loss being equal or even less after rainy days than after dry days.

In parathion experiments rainfall seemed to have a more important effect, although it never occurred before the fourth day. In test D, (fig. 5) 9.8 mm rain during the third day was followed by a loss of 3.9 % of the initial residue. During the fifth day of the same experiment there was 14 mm rain which was followed by a reduction of 2.6 % in the original deposit. Since the temperature on both days was high, it is not possible to discern the exact effect of these quantities of rain on the parathion residues.

These results are generally in accordance with the results of the authors mentioned above who agreed about the detrimental effect of rainfall during the first 24 hours after treatment with insecticides and its relatively less injurious effect in later days. However, the extent of residue loss caused by rainfall depends

on the nature of deposits formed on the plant surfaces which in turn, is dependent on the formulations used. It seems that deposits formed by dusting or spraying suspensions are much more seriously affected by rainfall than those from emulsions.

d. Effect of sunshine

The effect of sunlight on the loss of either insecticide residues was always obscured in the experiments described here by the effect of temperature. Bright sunshine coincided in all tests with high temperature. There were several days with a temperature higher than 25°C and periods of sunshine less than 5 hours, but days with bright sun for 8 hours or more with a temperature less than 25°C did not occur except two times, viz. the first day of test A and the fifth day of test E. In the first day of test A (fig. 2) a combination of a maximum air temperature of 23°C and a minimum air temperature of 10.5°C together with 9.25 hours of sunshine and 0.2 mm rain resulted in a loss of 1.5 µg DDT per leaf disc which is equal to the loss resulted from the second day of weathering. The weather records of the second day indicate that the maximum air temperature was 32.7°C, the minimum air temperature was 8.8°C, the sun shone for 2.6 hours and there was 18.5 mm rain. As previously stated the effect of rainfall could be neglected. Thus, a moderate temperature (around 25°C) prevailing for several hours together with sunlight for a period more than 9 hours could be compared in their detrimental effect on DDT residue with a higher temperature (around 35°C) lasting for a short time and a period of sunshine less than 3 hours. This indicates that in certain cases an increase in intensity or duration of sunshine could compensate for a decrease in temperature as regards its destructive action upon insecticide residues. This assumption is supported by another observation on the general effect of weathering upon the final loss of DDT in tests B and C. The average maximum air and leaf surface temperatures in tests B and C (tables 4 and 5) were almost equal, but the average minimum air and leaf surface temperatures in experiment C were 2.8°C and 2.5°C, respectively, higher than those of test B which means that the air and the leaf surface temperatures remained high for a longer time in test C than they did in test B. On the other hand, the period of sunshine in test B was 13.05 hours longer than that of test C. In spite of the possible effect of these additional hours of sunshine upon DDT residue the final per cent losses of material in both tests were fairly similar. Since the 9.8 mm rainfall on the fifth day of test B had a negligible effect on DDT residues, the foregoing observations on the temperature and sunshine conditions of tests B and C indicate that additional periods of sunshine could compensate for slight differences in temperature level or duration.

During the fifth day of test E with parathion residue (fig. 6) the maximum air temperature was 21.5°C, the minimum air temperature was 9.2°C, the period of sunshine was 8.8 hours and there were 6 mm rain. This combination of weather factors resulted in a loss of 0.1 µg parathion per leaf disc. The loss is so small that it affords no chance to discern the effect of any of these weather components.

On the basis of the foregoing results the present author agrees with the statement of HOPKINS *et al.* (1952b) concerning the effect of sunlight upon DDT residues. Sunlight, within the normal intensities, seemed to act on residues of either DDT or parathion in a slow but continuous way which obviously re-

sembles the effect of mild temperatures. Its deleterious effect upon parathion, as well as the effect of temperature, was higher than upon DDT.

e. Penetration into plant tissue

Two preliminary tests, one with DDT and the other with parathion, were conducted to form an approximate idea about the duration of residues causing 70–80 % initial mortality when weathered. Table 10 shows the density of these residues and the resulting mortalities either on glass plates or on ivy leaves. The first row of the table represents the quantities of residues delivered on glass plates as determined by spraying intermittently, together with the plates, round filter paper discs which were weighed before and after spraying. The second row represents the quantities of residue delivered on the leaves as estimated in the same way, whereas the third row gives the densities of toxicants on ivy leaves as determined chemically on the initial samples.

TABLE 10. Mortalities caused by almost equal doses of DDT or parathion applied on glass plates and ivy leaves by spraying mentioned concentrations of either toxicant.

Treated surface	0.0156% DDT			0.00625% parathion		
	μg per leaf disc (21.25sq.cm)	μg per sq.cm	per cent mortality	μg per leaf disc (21.25sq.cm)	μg per sq.cm	per cent mortality
Glass plates, spr.	11.9	0.560	72.25	4.590	0.216	78.5
Ivy leaves, spr.	11.86	0.558	53	5.164	0.243	4.5
Ivy leaves, chem.	11.0	0.517		4.70	0.221	

These values indicate that almost the same residues that resulted in 72.25 % mortality in case of DDT and 78.5 % kill in case of parathion when applied to glass plates, gave only 53 % and 4.5 % mortality, respectively, when applied to ivy leaves. The slight decrease in DDT residue on leaves as compared with the residue on glass plates could not be responsible for the decrease of about 20 % in the resulting mortality. The assumption that parts of the deposits on leaves might have been lost through an unknown error is rejected by the results of the chemical analyses which show that the deposits in the tests with leaves were almost equal with the residues on glass plates. The only possible loss of residue then could occur by the penetration of certain quantities of the toxicant from over the surface to inside the foliage tissue where they could not contribute to contact action. It has been mentioned before that the values provided by the chemical analyses represent the quantities of insecticides on and in the foliage altogether. Therefore, the amount of toxicants that penetrated into the foliage and caused demonstrable reduction in the expected mortalities could only be revealed by comparing the results of chemical assay and bioassay.

The per cent mortalities of all samples in each of tests A, B and C with DDT and tests D and E with parathion were plotted on log dosage-probit mortality paper against the doses of either toxicant which were computed as quantities of insecticide per leaf disc. The obtained curves were, in all cases, straight lines (figs. 7 and 8).

The results of test A gave a steeper line than the lines formed by the results of tests B and C as shown in fig. 7. This indicates that either the DDT residue was more effective during test A or that the test insects, at that time, were more

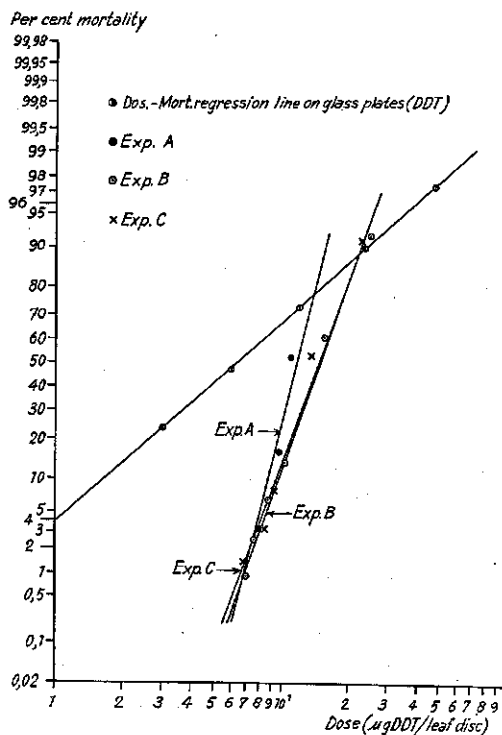


FIG. 7. Dosage-mortality regressions for field experiments A, B and C with DDT residues and the dosage-mortality regression for DDT deposits on glass plates.

susceptible than during tests B and C. The latter assumption seems to be more reasonable.

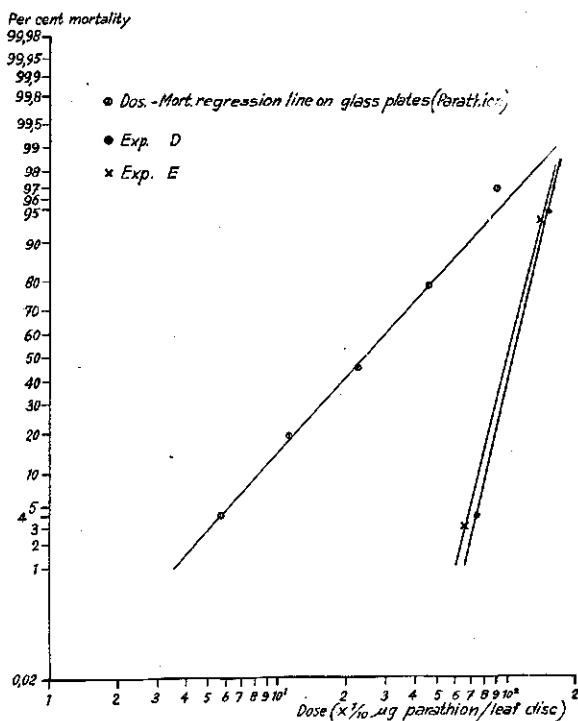
Residues weathered for more than one day, in parathion tests, failed to effect any mortality. Consequently, each parathion experiment was represented on the probit-log paper by two points only; those of the initial and one day weathered samples. However, the lines of both tests D and E (fig. 8) represent almost the same slope and trend. In general, they are somewhat steeper than the lines of both tests B and C with DDT.

Figures 7 and 8 also show the dosage-mortality regression lines for DDT and parathion. Each of these two lines was obtained by plotting the per cent mortalities that resulted from the residues formed on glass plates by a series of five graded concentrations on probit-log paper. The dosages were also computed as quantities of actual toxicant per each leaf disc.

The dosage-mortality line of parathion compared with the straight lines of both tests D and E indicates that the quantity of insecticide required to effect any mortality on glass plates is always smaller than the quantity needed to give the same mortality on leaves. These differences diminish gradually towards the higher doses.

The study of the dosage-mortality line of DDT on glass surfaces as compared with the straight lines of tests B and C reveals that, as in the case of parathion residues, smaller quantities of material are needed on glass surface to cause the same mortalities as on ivy leaves. This holds true for DDT residues that cause mortalities up to the level of about 90 %, after which the reverse occurs. This

FIG. 8. Dosage-mortality regressions for field experiments D and E with parathion residues and the dosage-mortality regression for parathion deposits on glass plates.



means that the amount of actual toxicant required to give mortalities higher than 90 % are smaller on leaves than on glass plates.

These observations are shown better in table 11 which presents the LD_{50} and LD_{92} for DDT residues on both glass plates and leaves and the LD_{50} and LD_{96} for parathion deposits on the same surfaces. These values were determined by interpolation with the dosage-mortality regression lines of either insecticide and with the straight lines obtained by plotting the results of experiments B and C, with DDT, and D and E with parathion, on the probit-log paper.

Table 11 shows that the LD_{50} for DDT on glass surface is smaller than that on leaves, whereas the LD_{92} of the same material on leaves is smaller than that on glass plates. On the other hand, the LD_{50} and LD_{96} for parathion on glass plates are smaller than the corresponding doses on leaves. The LD_{50} for para-

TABLE 11. Doses in μg per leaf disc and per sq.cm. required to cause 50% and 92% mortality with DDT residue and 50% and 96% kill with parathion residue on both glass plates and ivy leaves.

Surface	DDT residue				Parathion residue			
	LD_{50} in μg		LD_{92} in μg		LD_{50} in μg		LD_{96} in μg	
	per leaf disc	per sq. cm	per leaf disc	per sq. cm	per leaf disc	per sq. cm	per leaf disc	per sq. cm
Glass plates	6.40	0.301	27.0	1.270	2.4	0.113	10.0	0.470
Ivy leaves	14.75	0.694	23.5	1.105	10.5	0.494	15.5	0.729

thion on leaves is about 4.5 times higher than that on glass, while the LD₉₆ on leaves is only 1.5 times higher than that on glass plates.

The foregoing data lead to the following conclusions:

1. Under the present experimental conditions, parathion emulsion proved to have a greater penetrating capacity into ivy leaves than DDT emulsion.
2. Penetration into plant tissue starts soon after the application of material to the foliage and continues for some time. The amount that penetrates seems to be relatively constant and is almost independent of the density of residue applied.
3. The re-issuing of toxicants from within the tissue on to the leaf surface was not observed in all the field experiments with either insecticide, perhaps because the rate of loss by weathering was higher than the rate of re-issuance.
4. The density of DDT deposit required for 90 % or higher mortality is lower on ivy leaves than on glass plates, indicating that at such levels the material is more effective on leaves than on glass surfaces.

PRADHAN (1949) mentioned that DDT deposits were more toxic to adults of *Tribolium castaneum* HBST. at high relative humidities than at low ones. BURT & WARD (1955b) found that DDT residues on plant surfaces were more toxic to *T. castaneum* than on glass plates. They showed experimentally that the effectiveness of DDT deposits on glass plates was enhanced by the increase in the relative humidity at the tested surfaces.

Hence, the increased toxicity of DDT deposits that cause mortalities higher than 90 % on plant surface than on glass plates may be explained as follows: with low densities of residues, the effect of loss of DDT by penetration into the plant tissue is sufficiently large to obscure the increased toxicity of material on foliage as compared with glass surface. On the other hand, when high densities of DDT are used, any loss of toxicity due to penetration of the toxicant is more than compensated for by the increased toxicity of residues on plant surfaces as compared with their effectiveness on glass plates.

IV. LABORATORY EXPERIMENTS

Laboratory experiments comprised tests with DDT and parathion residues applied to ivy leaves. The purpose of these experiments was to determine the relative importance of temperature, simulated rain and ultraviolet light. This information was required as a basis for the interpretation of residue losses in the more complex field situation.

In addition, laboratory experiments were carried out to obtain dosage-mortality regression lines of both DDT and parathion residues on glass surface (figs. 7 and 8). This was done with the intention to be able to estimate the proportion of insecticide residue present at the leaf surface on the basis of contact bioassay. For this purpose, residues of varying densities were formed on glass plates by spraying a graded series of five concentrations (table 12). The doses are computed as quantities of actual toxicant per leaf disc (21.25 sq.cm) and per sq.cm. The table also shows the percentage of kill obtained when test insects were introduced on to these residues.

All five concentrations of each insecticide were sprayed on to the plates on the

TABLE 12. Per cent mortality resulting from DDT and parathion residues (computed as μg toxicant per leaf disc and per sq.cm) formed on glass plates by spraying five graded concentrations of each material.

No.	DDT				Parathion			
	concentration (%)	μg per leaf disc	μg per cm^2	% mortality	concentration (%)	μg per leaf disc	μg per cm^2	% mortality
1	0.0039	2.97	0.14	24	0.00078	0.57	0.027	4
2	0.0078	5.95	0.28	47	0.00156	1.14	0.054	19
3	0.0156	11.90	0.56	72.25	0.00312	2.29	0.108	45.75
4	0.0312	23.80	1.12	90	0.00625	4.59	0.216	78.5
5	0.0625	47.60	2.25	97.50	0.0125	9.18	0.432	97

same day and left to dry for six hours after which the effectiveness of each concentration was tested as described before. Eight glass plates 12×9 cm were sprayed with each concentration. The quantities of toxicants deposits were estimated by the method mentioned before in which round filter paper discs were sprayed alternatively with the glass plates.

A. Temperature Experiments

1. LITERATURE

There is a noticeable lack of literature pertaining to the influence of the different weather components on parathion deposits as studied by laboratory experiments. In contrast with this, DDT has been, and still is, the subject of several field and laboratory studies.

DOMENJOZ (1944) reported that little or no decomposition occurred on heating DDT at 150°C for 24 hours. FLECK (1944) studied the rate of evaporation from glass plates of DDT dusts deposited at 63.36 mg per 50 sq.cm . These were kept at 45°C with air passing over them and were weighed at 4-day intervals. After 37 days 4.22 mg DDT had evaporated. The material scraped from plates at the end of the test had a melting-point identical with that of the initial material. From this the author concluded that "loss of DDT from insecticidal spray deposits by volatilisation will occur too slowly to be of any importance". FLECK & HALLER (1944 and 1945) mentioned that small quantities of impurities found in insecticidal mixtures act as catalysts causing decomposition of DDT, which is accelerated at high temperatures. GUNTHER (1947) commented on the thermal decomposition of DDT in admixture with technical grade benzene hexachloride, presumably because of traces of iron in the BHC preparations. He also found that the chloride ion is not a catalyst for the thermal decomposition of DDT. CHISHOLM & KOBLITSKY (1947) reported that recrystallised and technical DDT applied to Petri dishes as dusts at the rate of about 3 mg per sq.cm . and kept at 112°F (44.5°C) for 72 hours decreased in weight by about 0.25% and 1.5% , respectively. About 75% of the loss of recrystallised DDT occurred within the first 24 hours, whereas only about half the loss of the technical DDT occurred in the same period. FLECK (1949) attributed the prolonged residual effect of DDT to its low vapour pressure and its stability to oxidation. DECKER *et al.* (1950), working with several insecticides including DDT and parathion, concluded that there is a positive correlation between the vapour

pressures of insecticides and their rates of loss from crops. BURT & WARD (1955a) studied the effect of two different temperatures upon the decrease of DDT deposits formed on plain and waxed glass plates by spraying a suspension of DDT crystals. After 26 days at 18°C there was no significant change in any of the deposits. At 43°C, the amount of DDT estimated chemically and biologically against *Triboleum castaneum* HBST. diminished rapidly and was negligible after 26 days. The loss of toxicity at 43°C was shown to be accompanied by loss of weight of the residues. They were able to reduce effectively the loss of deposit on plain glass plates by storing them in an atmosphere saturated with DDT vapour. They concluded from these results that loss of toxicity at the higher temperature was due to volatilisation of DDT and decomposition. The same authors (1955b) found that losses were proportionally greater for light deposits of DDT than for heavy ones. They attributed the reduced loss with heavy residues as being probably due to the fact that the rate of evaporation is limited by the saturation of the air near to the deposit. They commented that at similar temperatures but with free ventilation, the rate of evaporation may be much greater. BURGESS & SWEETMAN (1949) studied the duration of effectiveness of DDT deposits formed on galvanized and black enamelled wire screens by dipping them in a 5 % DDT solution in kerosene. The deposits kept at 37°C in moist conditions showed marked reduction in effectiveness against houseflies over a period of 39 months while similar screens held at 23°C in dry atmosphere remained highly effective throughout the same period. They concluded that the combination of a high temperature (37°C) with a high relative humidity (60 to 75 %) produced a more rapid reduction in toxicity of DDT than a low temperature (23°C) with a low relative humidity (25 to 40 %).

MARTIN (1950) observed a progressive and significant decrease in the toxicity of parathion towards *Calandra granaria* L. during 24 hours heating at 150°C. Similar results were obtained when methyl parathion was heated at the same temperature for only six hours. He found that the thiono sulfur content diminished to practically zero during the heating period and commented that this disappearance of contact toxicity would indicate that heat treatment results in changes more extensive than isomerization. WOODCOCK & STRINGER (1951) confirmed the isomerization of both parathion and its methyl homologue at temperatures above 140°C. They also showed that further heating of the S-ethyl isomer of parathion caused thermal decomposition and loss of toxicity to *C. granaria*. MCPHERSON & JOHNSON (1956) studied the thermal decomposition of five phosphorothioate insecticides including parathion. They attributed the absence of contact insecticidal activity of methyl parathion after being heated for six hours at 150°C, as reported by MARTIN (1950), to the possible formation of mixed polymetaphosphates. These materials have poor solubility in benzene or ether which indicates low permeability into the insect cuticle and, consequently, low contact insecticidal activity. MISTRIC & MARTIN (1956b) found that parathion deposits on cotton seedlings resulting from a water emulsion spray containing 0.24 pound actual toxicant per acre, caused an average daily mortality of 57 % to the cotton aphid, *Aphis gossypii* GLOV., when these were released on to plants immediately after treatment and kept exposed to residue for five days. The toxicity of the residue was reduced by 11 % if the seedlings were first kept for 24 hours after treatment at 105°F (40.5°C) in a shaded greenhouse before the test insects were released on to them.

2. EXPERIMENTAL

In these experiments, residues of DDT or parathion were subjected to two different temperatures in complete darkness. The decrease in residue was estimated by sampling at regular intervals.

Leaves sprayed and then left to dry for 6 hours were placed with their petioles immersed in water in 6 cm long flat-bottomed glass tubes. They were then arranged on suitably bored shelves inside a constant temperature cabinet adjusted to maintain the required temperature. Except for two small holes on both sides of each cabinet, the leaves were kept in darkness during the whole experiment. The two holes allowed for slow ventilation and escape of excessive moisture.

With DDT residues the effect of exposure to 25°C and 35°C was examined in two different experiments each of which included six samples in addition to the sample removed in the beginning to represent the initial residue. Subsequent samples were taken regularly every 48 hours.

Preliminary experiments showed that parathion might be more heat-labile than DDT. Hence, parathion residues were exposed to 20°C and 30°C. Each of the two parathion experiments consisted of six samples including the initial one. Sampling was done every 24 hours.

3. RESULTS

a. DDT residue

Table 13 and figure 9 show the results of the two experiments with DDT residues kept at 25°C and 35°C. These tests will be referred to as experiments F₁ and F₂.

The initial residue in experiment F₁ was reduced by 2.2 % after being kept at 25°C for two days. In spite of this loss of material the mortality resulting from this residue was 2.2 % greater than that caused by the original deposit. After four days at 25°C, 19.6 % of the initial DDT quantity was lost. The total loss of material at the end of this experiment amounted to 55.4 % which means that about 36 % of the residue was lost between the fourth and the twelfth days

TABLE 13. The effect of keeping DDT residues in darkness for 12 days at 25°C and 35°C as detected chemically and biologically by regular sampling every 48 hours. Residues are computed as µg DDT per sq.cm and per leaf disc (21.25 sq.cm).

Days kept at test temp.	Experiment F ₁ (at 25°C)						Experiment F ₂ (at 35°C)					
	µg per leaf disc	µg per cm ²	% mortality	% decrease in residue	% decrease in residue per 2 days	% decrease in mortality	µg per leaf disc	µg per cm ²	% mortality	% decrease in residue	% decrease in residue per 2 days	% decrease in mortality
-	23	1.082	92.5	-	-	-	23.75	1.117	93	-	-	-
2	22.5	1.058	94.5	2.2	2.2	+2.2	14.5	0.682	52	38.9	38.9	44.1
4	18.5	0.871	80	19.6	17.4	13.5	9.5	0.447	5.5	60	21.1	94.1
6	16	0.752	57	30.4	10.8	38.4	8.75	0.411	3	63.2	3.2	96.8
8	13.75	0.647	44.5	40.2	9.8	51.9	8	0.376	2	66.3	3.1	97.9
10	12.25	0.576	25	46.7	6.5	73	7.5	0.352	0.5	68.4	2.1	99.5
12	10.25	0.482	16	55.4	8.7	82.7	7	0.328	0	70.5	2.1	100

+ Mortality increased by this per cent in spite of a 2.2% decrease in residue.

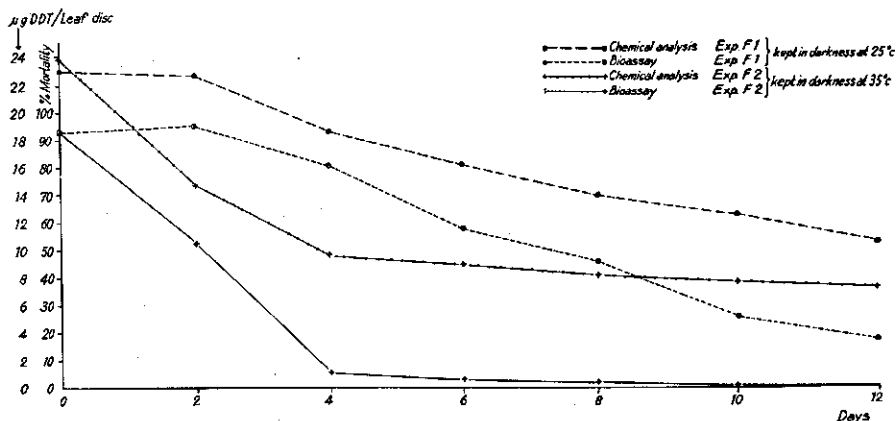


FIG. 9. DDT residues kept in darkness at 25°C and 35°C for 12 days as detected chemically and biologically by sampling every 48 hours (experiments F₁ and F₂).

of the experiment. Table 13 shows that the loss of residue during the last eight days was almost regularly distributed over this period at the rate of about 4.5 % per day.

The rate of residue reduction during experiment F₂ was much higher within the first four days than it was during the same period in experiment F₁. The loss of DDT after two and four days at 35°C was almost double what was lost when the residues were kept at 25°C for four and six days, respectively. After 48 hours at 35°C the loss was 36.7 % greater than for the same period at 25°C. At 12 days, however, the reduction of residues at the higher temperature was only 15.1 % greater than that suffered at 25°C. This indicates that there was a greater decrease in the rate of loss during the latter part of test F₂ than in experiment F₁. The slowing down in the rate of loss in the last period of experiment F₂ is also shown by the fact that during the last eight days of this experiment only 10.5 % of the initial deposit was lost as compared with 35.8 % loss for the same period in experiment F₁. Thus the DDT deposit kept at 35°C suffered an average daily loss of about 1.3 % of its initial quantity during the last eight days.

The results of experiment F₂ (table 13) show that a DDT deposit of less than 7.5 µg per leaf disc causes no mortality. Hence, it can be concluded that both the sublethal dose of DDT at the surface and the quantity of material that penetrated into the plant tissue amount to about 7 µg per leaf disc.

b. Parathion residue

The results of the two experiments in which parathion residues were held at 20°C and 30°C are shown in table 14 and figure 10. These two experiments will be referred to as experiments F₃ and F₄.

Whereas one day at 20°C reduced the initial deposit of parathion by 20 %, a similar period at 30°C caused 39.7 % loss or about double that at 20°C. In both experiments F₃ and F₄, however, more comparable losses were experienced during the succeeding four days amounting to 33.3 % and 27 %, respectively. After 5 days the loss at 30°C exceeded that at 20°C by 13.4 % which was nevertheless, 6.3 % less than the difference after one day. This means that during

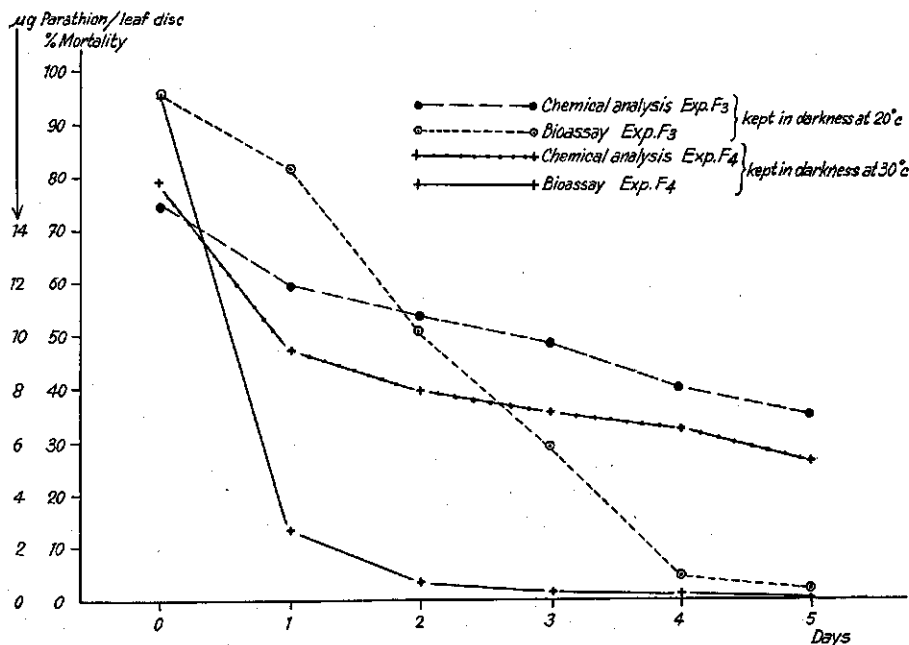


FIG. 10. Parathion residues kept in darkness at 20°C and 30°C for five days as detected chemically and biologically by sampling every 24 hours (experiments F₃ and F₄).

the last four days the average daily rate of residue loss at 30°C ($\pm 6.75\%$) was less than that at 20°C ($\pm 8.3\%$) during the same period.

A parathion residue of less than about 6.5 µg per leaf disc seemed unable to cause mortality to *Calandra* weevils. This non-effective remaining residue, therefore, represents both the sublethal dose at the surface and the quantity of material that penetrated into the leaves.

TABLE 14. The effect of keeping parathion deposits for 5 days in darkness at 20°C and 30°C as detected chemically and biologically by daily samples. Residues are computed as µg parathion per sq.cm and per leaf disc (21.25 sq.cm).

Days kept at test temp.	Experiment F ₃ (at 20°C)						Experiment F ₄ (at 30°C)					
	µg per leaf disc	µg per cm ²	% mortality	% decrease in residue	% decrease in residue per day	% decrease in mortality	µg per leaf disc	µg per cm ²	% mortality	% decrease in residue	% decrease in residue per day	% decrease in mortality
—	15	0.706	96	—	—	—	15.75	0.741	96.5	—	—	—
1	12	0.564	82	20	20	14.6	9.5	0.447	13.5	39.7	39.7	86
2	10.75	0.505	51	28.3	8.3	46.9	8	0.376	3.5	49.2	9.5	96.4
3	9.75	0.458	29	35	6.7	69.8	7.1	0.334	1.5	54.9	5.7	98.5
4	8	0.376	4.5	46.7	11.7	95.3	6.5	0.305	1	58.7	3.8	99
5	7	0.334	2	53.3	6.6	97.9	5.25	0.247	0	66.7	8	100

4. DISCUSSION

According to the data reviewed above, an increase in temperature might be expected to cause an increase in either the rate of chemical degradation of DDT, or its volatility, or both. It seems more possible that the loss of DDT deposits under conditions of high temperatures occurs through both decomposition and volatilisation together, but that the extent of decrease caused by each varies according to several factors among which the type of formulation is of great importance. The presence of alkaline or iron salts in the auxiliary constituents of the formulation, or the mixing of DDT with other toxicants, could enormously accelerate the rate of loss by chemical degradation. On the other hand, the use of very light densities of residues or the application of dusts of very fine particles leads to the increase of loss by volatilisation. The remarkably low vapour pressure of DDT indicates, however, that the disappearance of the material through volatilisation under normal field conditions at any rate could not be as important as the loss by chemical decomposition, especially when the effect of temperature is enhanced by other weather factors such as sunlight.

Tables 13 and 14 show that a parathion residue kept at 20°C for one day loses 20 % of its original quantity which is about ten times as much as the loss suffered by a residue of DDT held at 25°C for two days. The percentage loss caused to the DDT deposit at 35°C after 48 hours was similar to that found in the parathion residue kept at 30°C for only 24 hours. Again, the decrease in the original residues of DDT held at 25°C for 12 days and at 35°C for 8 days were 55.4 % and 66.3 %, respectively, which approximate closely to the losses of 53.3 % and 66.7 % suffered by parathion deposits kept at 20°C and 30°C, respectively, for only five days.

The above comparison between losses experienced by the two toxicants at the test temperatures shows that parathion is more heat-labile than DDT. The quick rate of disappearance of the parathion residue at even moderate temperatures, in comparison with the prolonged duration of DDT deposits at higher temperatures, is believed to be due to the chemical instability of parathion and its high vapour pressure. This latter was reported by BRIGHT *et al.* (1950) to be 3.78×10^{-5} mm Hg at 20°C. By comparison DDT is very stable and has a low vapour pressure estimated by BALSON (1947) to be 1.5×10^{-7} mm Hg at 20°C.

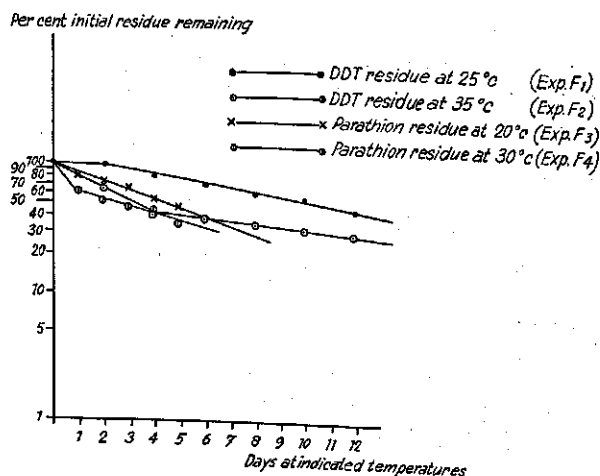


FIG. 11. Rates of loss of DDT and parathion residues as affected by indicated temperatures.

The rates of loss of DDT and parathion residues at the test temperatures are showed in figure 11. The line formed by DDT residues kept at 25°C is a straight line except for the first part which is flatter indicating a slower rate of loss within the first two days. The line representing DDT deposits kept at 35°C shows that the loss was higher during the first four days than during the latter eight days of the test. Parathion residues held at 20°C gave a steeper straight line than that of DDT deposits held at 25°C showing a higher rate of decrease. Parathion deposits kept at 30°C yielded a broken line steep to begin with but flattening directly after the first day. The rate of loss thus decreased appreciably after 24 hours.

Figure 11 indicates that, irrespective of the relative stability of each insecticide used, most of the loss of material suffered on exposure to high temperatures occurs within the initial period of exposure, after which the rate of loss decreases significantly. At a moderate temperature, on the other hand, the initial losses depend on the degree of stability of the material, whilst subsequent losses occur at a relatively higher level than are experienced at high temperatures. After a sufficient period of exposure at either high or moderate temperatures, the losses suffered by a certain toxicant will be almost equal. This may partly account for the frequent observation in field tests that weathering of insecticide deposits for several weeks at temperatures high enough to cause actual losses results in residues that are almost equal, irrespective of temperature.

In experiment F₁ with DDT (table 13), the sample taken after 2 days showed a 2.2 % increase in effectiveness in spite of a 2.2 % decrease in the deposit. This increase in mortality might be due to the re-emergence on to the surface of material from inside the leaf tissue, as was observed by GUNTHER *et al.* (1946) on citrus foliage. The reason why this re-issuing of toxicants was observed in this sample only could be that the detrimental effect of 25°C for 2 days on the DDT deposit was not sufficiently high to obscure the effect of the increased quantity of material on the surface.

B. Rain Experiments

1. LITERATURE

The effect of rainfall on DDT deposits was investigated by FENNAH (1945) by spraying microscope slides and exposing them to rain. The initial deposits consisted of crystalline aggregates. The residues were exposed to rainfall during two days at the end of which the intermittent rain amounted to 1.6 inches. Samples taken at intervals of 15, 30 and 60 minutes and two days showed a progressive erosion of the aggregates. In a deposit eroded soon after application of the spray, the crystals were small and irregular, indicating that some of the DDT had been washed away before it had crystallised out of solution. The removal of dry deposits of crystals appeared to be due to the mechanical action of rain-drops as they impinged on the slides, since the careful addition of a drop of rain-water to the sprayed surface of a slide caused no apparent loosening or removal of DDT crystals. The author comments that the degree of erosion from the exceptionally smooth surface used was probably maximal, and suggests that moderately heavy showers may seriously reduce a deposit of DDT on the upper surfaces of leaves. HOPKINS *et al.* (1952b) reported that KIRK (1951) had studied the effects of rain on DDT spray deposits on potatoes, by simulating

rainfall with overhead irrigation. He found that 0.5 inch of water applied just after drying of a spray of DDT wettable powder removed 86.4 % of the initial residue as detected chemically. When the water was not applied until 24 hours or more after spraying, the deposits were much less susceptible to erosion.

GAINES & MISTRIC (1952) mentioned that fresh parathion residues obtained by spraying a wettable powder on to potted cotton plants at a rate of half a pound of active ingredient per acre caused 82.22 % mortality to salt-marsh caterpillars. Similar residues but which had been exposed to half an inch of simulated rain caused only 22.4 % mortality, being 72.8 % less effective than unexposed residues. MISTRIC & MARTIN (1956b) found that parathion residues deposited on cotton plant seedlings from a water emulsion spray containing 0.24 pound actual toxicant per acre, caused an average mortality of 46 % to cotton aphids confined on the plants for five days at a constant temperature of about 20°C. When treated plants were exposed to half an inch of simulated rain 30 minutes after application of the spray, the average toxicity of the deposits was reduced by 23 %.

All the evidence points to an almost entirely mechanical effect of rain. However, the physical properties of an insecticide may affect the degree of loss by rainfall. DECKER *et al.* (1950) found that toxaphene and chlordane were much less affected by heavy rainfall amounting to 2.33 inches than aldrin, dieldrin and DDT. They attributed this stability of toxaphene and chlordane to the waxy and sticky nature of these two insecticides, in contrast to aldrin, endrin and DDT which are crystalline. In addition, in considering the losses suffered by DDT and parathion deposits exposed to rainfall, the solubility of these toxicants in water could be of great importance.

2. EXPERIMENTAL

The aim of these experiments was to wash the leaves carrying the insecticide residues with increasing quantities of water applied as a shower. This necessitated an apparatus which could spray evenly specific quantities of water on to the required number of leaves. For this purpose a simple apparatus was designed and constructed at the laboratory. In use it was surrounded with dark curtains to minimize the effect of light on the residues. Room temperature varied between 14.5°C and 22°C.

The main features of this rain apparatus (fig. 12) were:

1. Two circular turntables (T) standing side by side, each 1 m. in diameter. The tables were turned by means of an electric motor (M), provided with a gear-box which enabled the use of three different speeds. Each table carried two rows (R1, R2) of holders (S) in which the flat-bottomed glass tubes carrying the leaves were easily fitted. The two rows of each table were together capable of carrying 48 leaves; 27 and 21 leaves in the outer and inner rows, respectively. The outer row was 10 cm. and the inner row 20 cm from the periphery of the table. Water falling on each table during the "raining period" was collected between two walls each 12 cm high, and was later drained through a small tap fixed in the base of each table near the periphery.
2. Two identical swirling-jet spraying nozzles (N) (Duiker 1-The Hague), each with 0.6 mm opening, fixed at the two ends of a common inverted Y-shaped metal pipe, which was connected in turn with the water reservoir (RS) by

means of a thick-walled rubber tube. Water flow to the nozzles was regulated by a valve (V). Each nozzle was fixed 50 cm above and 15 cm from the periphery of its corresponding table. The cone of water droplets sprayed from each nozzle was sufficient to cover evenly, as the table rotated, a circular band about 35 cm broad. To reduce the turbulence caused by the rotation of the outer walls of the turning tables, two curved zinc plates (P) were fixed vertically between the two nozzles with their lower edges below the rim of the corresponding wall.

3. A water reservoir (RS), of the type common to most bucket sprayers, connected to the nozzles and a compressed air cylinder. The reservoir was provided with a manometer. Ordinary tap-water was used to fill the tank which had a capacity of about 30 liters.

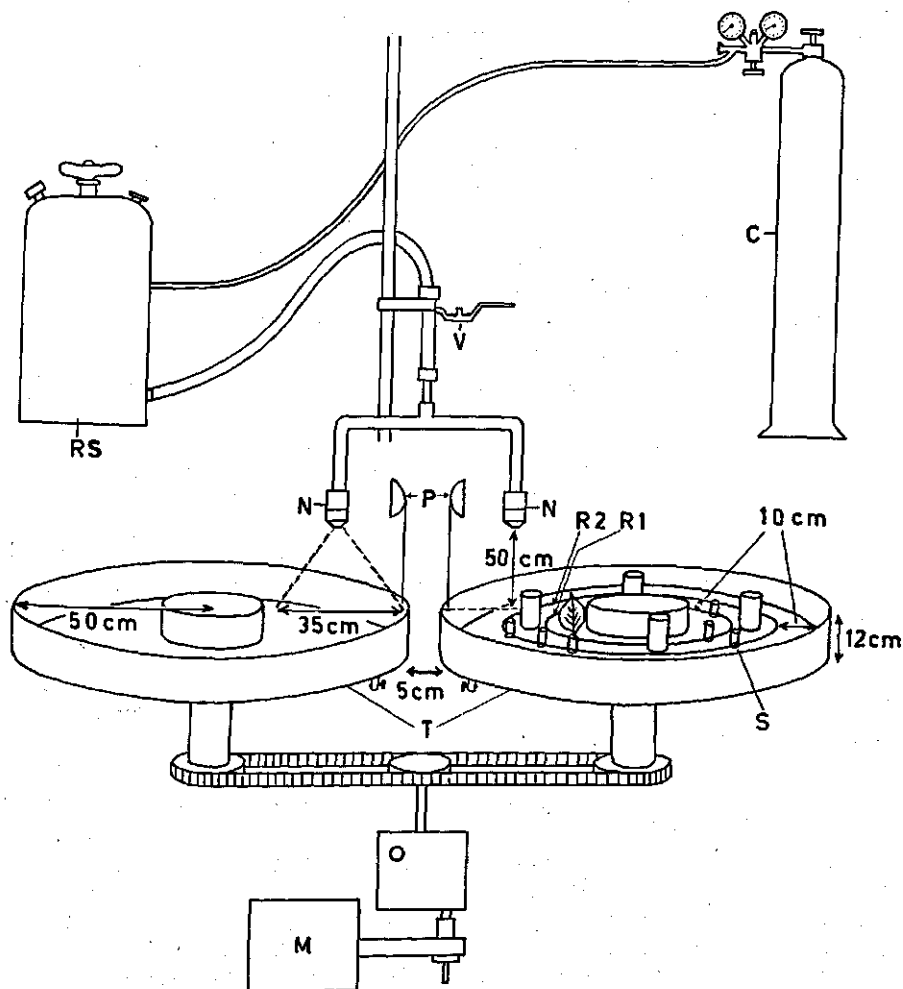


FIG. 12. Simulated rain apparatus; turntables (T), electric motor (M), rows of holders (R₁ and R₂), spraying nozzle (N), water reservoir (RS), valve (V), curved zinc plates (P) and compressed air cylinder (C).

4. A compressed air cylinder (C) to provide the required pressure in the water reservoir. Pressure was regulated and controlled by means of a reduction valve (Hornung-H 57) directly connected to the cylinder. The pressure applied in all experiments was 3 kg/sq. cm.

Calibration of the apparatus was carried out as follows:

The holders of each turntable were replaced by eight short glass cylinders arranged equidistantly on each table so that the cylinders stood exactly where the stands had been. The apparatus was allowed to work for a certain period after which the quantity of water in each glass cylinder was measured and the quantity per square centimeter calculated. The average difference between the quantities collected in the cylinders of the outer and inner rows of each table was always within the limits of 6 %, while the average difference between the total average of either table was usually within 8 %. Although these differences were small it was nevertheless considered advisable to minimize their effect by applying the following precaution: The leaves exposed to water on one turntable were transferred to the other table for the next exposure, with those leaves that had previously formed the inner row now occupying the outer row. Thus the differences were compensated except in the case of the first sample taken after only one simulated "rain period". At this stage, however, the difference would have been too small to take into consideration.

Two rain tests with either DDT or parathion residues were run concurrently, one starting a day before the other.

In the first experiment with each insecticide the residues were exposed to \pm 3 mm simulated rain daily and in the second experiment to \pm 6 mm daily. The first was achieved by showering the leaves for 8 minutes and the second for 16 minutes. Water droplets, though varying, had an average diameter of 1.1 mm.

Each rain experiment included seven samples in the case of DDT and six samples in the case of parathion, initial residue samples included.

In order to check the actual quantity of water delivered in each simulated rain period, 4 short glass cylinders were arranged equidistantly between the two rows of leaves on each turntable so that the cylinders were not covered by the leaves. The quantity of water per square centimeter was determined at the end of each "raining period" as described before. Special attention was paid to sampling so that each sample was a randomized one representing the leaves of the inner and outer rows of both tables.

3. RESULTS

a. DDT residue

Table 15 and figure 13 show the results of the two experiments in which DDT deposits were subjected to approximately 3 or 6 mm simulated rain daily for six days. These experiments are referred to here as experiments G_1 and G_2 .

Since the residues in these experiments were kept at room temperature, the resulting decrease in deposit could be due to the effect of both temperature and the simulated rain applied. The room temperature varied between 22°C and 16°C, with an average of about 19°C. The leaves wetted by the simulated rain were always left till the next day to dry before taking the daily samples.

There is evidence that almost all the loss detected in the residues was due to the simulated rain and not to the influence of temperature. A single application of 6.2 mm water in experiment G_2 caused 7.6 % loss of the initial residue. A

$\mu\text{g DDT/Leaf disc}$

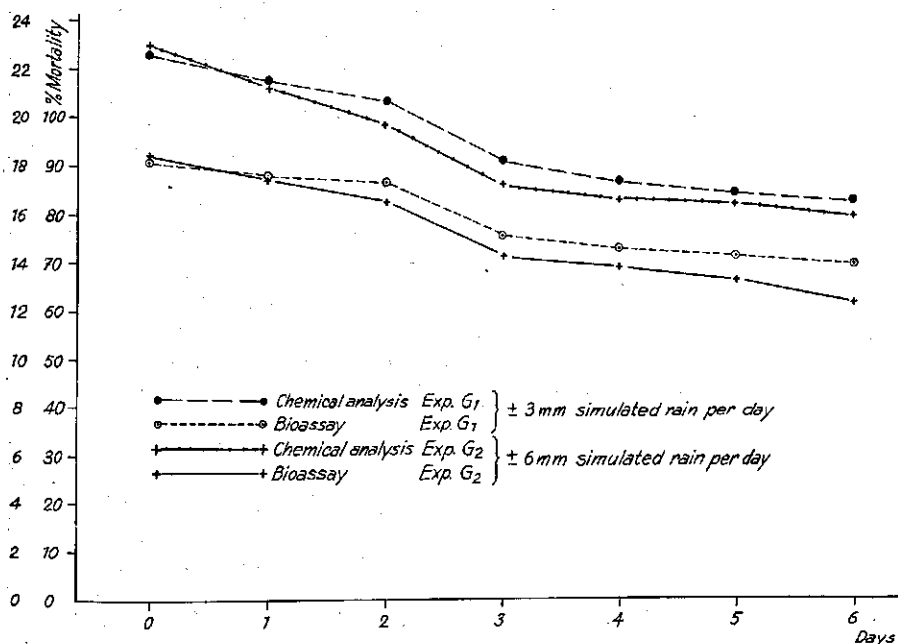


FIG. 13. DDT residues exposed to 6 applications of ± 3 mm or ± 6 mm simulated rain per day as detected chemically and biologically by daily samples (experiments G₁ and G₂).

similar loss of 7.8 % occurred in experiment G₁ after exposing the residues to 6.5 mm simulated rain delivered as 3.2 mm and 3.3 mm during the first two days. Moreover, DDT deposits in the last sample of experiment G₁ subjected to a total of 19.9 mm rain lost 25.6 % of their initial residue which agrees very closely with the loss suffered by the deposits in experiment G₂ that had been exposed to 18.3 mm simulated rain during three days only. These observations show that

TABLE 15. Effect of ± 3 and ± 6 mm simulated rain daily for 6 days upon DDT residue as detected chemically and biologically by daily samples. Residues are computed as $\mu\text{g DDT per sq.cm}$ and per leaf disc (21.25 sq.cm).

mm simulated rain per day	Experiment G ₁ (± 3 mm per day)						mm simulated rain per day	Experiment G ₂ (± 6 mm per day)					
	$\mu\text{g per leaf disc}$	$\mu\text{g per cm}^2$	% mortality	% decrease in residue	% decrease in residue per day	% decrease in mortality		$\mu\text{g per leaf disc}$	$\mu\text{g per cm}^2$	% mortality	% decrease in residue	% decrease in residue per day	% decrease in mortality
-	22.5	1.058	91	-	-	-	-	23	1.082	92	-	-	-
3.2	21.5	1.011	88.5	4.4	4.4	2.8	6.2	21.25	1.00	88	7.6	7.6	4.4
3.3	20.75	0.976	87	7.8	3.4	4.4	6.0	19.75	0.929	83	14.1	6.5	9.8
3.3	18.25	0.858	76	18.9	11.1	16.5	6.1	17.25	0.811	71.5	25.0	10.9	22.3
3.2	17.5	0.823	73.5	22.2	3.3	19.2	6.5	16.75	0.788	69.5	27.2	2.2	24.5
3.4	17	0.800	72	24.4	2.2	20.9	6.8	16.5	0.776	67	28.3	1.1	27.2
3.5	16.75	0.788	70	25.6	1.2	23.1	8.0	16	0.752	62	30.4	2.1	32.6

the losses of DDT were correlated with the quantities of simulated rain and not with the length of time during which the deposits were kept at room temperature.

Table 15 shows that one exposure to 3.2 mm simulated rain in test G_1 resulted in a loss of 4.4 % of the initial residue, which is 3.2 % less than the initial loss observed in experiment G_2 following one application of 6.2 mm simulated rain. Consequently, the 3.2 % increase in loss in experiment G_2 was due to the additional 3 mm water. The application of another 3.3 mm and 6.0 mm simulated rain in experiments G_1 and G_2 , respectively, increased the loss by almost exactly the same rate. Thus the decrease suffered by the initial deposits after the second application of simulated rain became 7.8 % in experiment G_1 and 14.1 % in experiment G_2 , or 3.4 % and 6.5 % more than the losses caused by one exposure to simulated rain in the respective experiments. Thereafter, this rate changed completely. The third application of simulated rain in both tests was more deleterious than the first two, causing far greater and almost equal losses of 11.1 % in experiment G_1 and 10.9 % in experiment G_2 .

The average daily rate of residue loss during the first three days of experiments G_1 and G_2 was 6.3 % and 8.3 %, respectively, compared with 2.25 % and 1.8 % for the last three days. The rate of loss for the whole period of experiment G_1 was about 4.3 % per day, whereas that for experiment G_2 was about 5.1 % per day. Of both experiments, therefore, it may be said that after the loss of about 20–25 % of the initial residue, the rate of loss diminished appreciably. Doubling the quantity of simulated rain clearly raised the rate of loss during the first three days of experiment G_2 , but the final loss of material was only slightly greater than in experiment G_1 .

In the sixth and the last samples of test G_2 , residues of 16.5 and 16.0 μg per leaf disc, respectively, caused 67 % and 62 % mortality. Table 13, however, shows that a density of 16.0 μg DDT per leaf disc in the fourth sample of temperature experiment F_1 gave only 57 % mortality. This result may indicate an increase in the effectiveness of the toxicant during rain experiments.

b. Parathion residue

The results of the two rain tests with parathion deposits are given in table 16 and figure 14. The residues were subjected for five days to approximately 3 or 6 mm simulated rain per day in the two experiments which are referred to here as experiments G_3 and G_4 , respectively.

Also in these experiments losses of material might be due to the effects of both room temperature and simulated rain. The maximum and minimum room temperatures were 21 °C and 14.5 °C, respectively, with a mean of about 17.5 °C.

Although in temperature experiments parathion proved to be a heat-labile insecticide, it is nevertheless thought that the temperature of the residues during these experiments was too low (around 15 °C) to cause serious losses. Moreover, simulated rain in both tests had such a deleterious effect that any loss caused by such low temperatures would by comparison be almost negligible. The per cent loss of initial residue in experiment G_3 resulting from two showerings of \pm 3 mm was equal to the loss caused by keeping dry parathion residues at 30 °C for five days as shown in table 14. Similarly, 6 mm simulated rain applied in one day in test G_4 was more detrimental than was 20 °C for five days. These examples show clearly the relatively slight effect of temperatures around 15 °C as compared with the considerable removal of material caused by simulated rain.

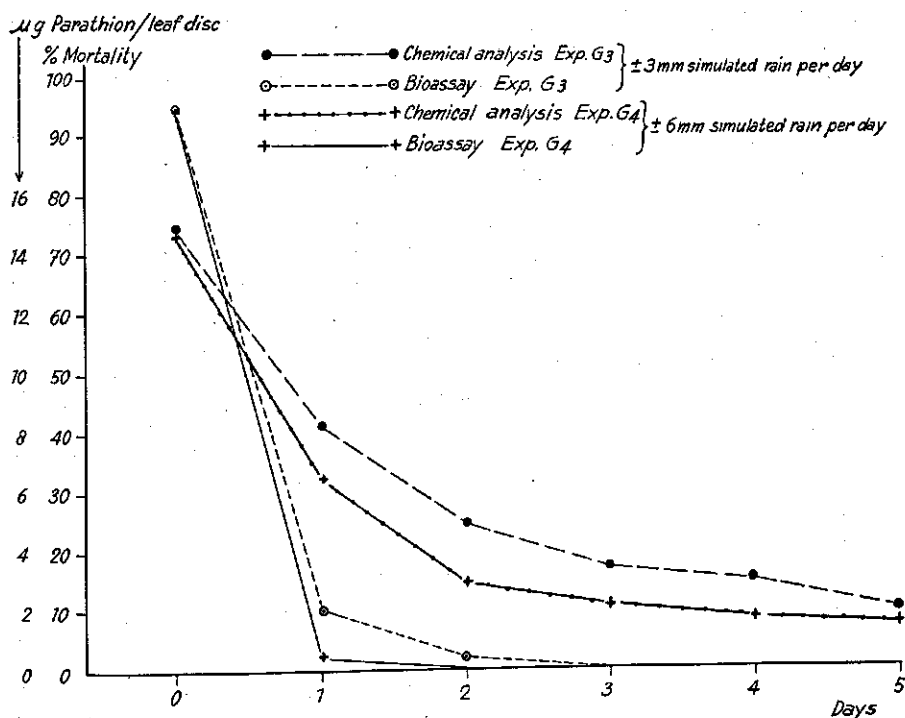


FIG. 14. Parathion residues exposed to 5 applications of ± 3 mm or ± 6 mm simulated rain per day as detected chemically and biologically by daily samples (experiments G_3 and G_4).

The results of parathion tests given in table 16 show that in experiment G_4 , the first 6 mm simulated rain caused 10.9 % greater loss than did the initial 3.1 mm in experiment G_3 . When simulated rain was applied a second time in each experiment, the difference in residue loss increased to 13 %. Three further applications in both tests reduced the differences to 8.1 %, 8.1 % and 3.1 %

TABLE 16. Effect of ± 3 mm and ± 6 mm simulated rain daily for 5 days upon parathion residues as detected chemically and biologically by daily samples. Residues are computed as μg parathion per sq.cm and per leaf disc (21.25 sq.cm).

mm simulated rain per day	Experiment G_3 (± 3 mm per day)						mm simulated rain per day	Experiment G_4 (± 6 mm per day)					
	μg per leaf disc	μg per cm^2	% mortality	% decrease in residue	% decrease in residue per day	% decrease in mortality		μg per leaf disc	μg per cm^2	% mortality	% decrease in residue	% decrease in residue per day	% decrease in mortality
-	15	0.706	95	-	-	-	-	14.75	0.694	95.5	-	-	-
3.1	8.25	0.388	10.5	45.0	45.0	88.9	6.0	6.5	0.305	2.5	55.9	55.9	97.4
3.2	5	0.235	2	66.7	21.7	97.9	6.1	3	0.141	0	79.7	23.8	100
3.0	3.5	0.165	0	76.7	10.0	100	5.9	2.25	0.106	0	84.8	5.1	100
2.9	3	0.141	0	80.0	3.3	100	6.0	1.75	0.082	0	88.1	3.3	100
2.9	2	0.094	0	86.7	6.7	100	5.8	1.5	0.070	0	89.8	1.7	100

during the third, fourth and fifth days, respectively. This indicates that towards the end of the experiments, total losses of material in tests G_3 and G_4 equaled gradually, regardless of differences in the quantities of simulated rain applied in each test.

The effectiveness of the deposits in experiment G_3 disappeared completely after 9.3 mm simulated rain, whereas in experiment G_4 no mortality was obtained after 12.1 mm.

The last application of simulated rain in experiment G_4 caused the loss of only 0.25 μg parathion per leaf disc, or about 1.7 % of the initial deposit. The quantity of material recovered from the last sample of this test amounted to only 1.5 μg per leaf disc making it the least dense residue achieved in any of the parathion tests. This shows the strong effect of rain on parathion deposits.

A density of 8.25 μg per leaf disc as found in the second sample of test G_3 caused 10.5 % mortality whereas comparable densities of 8 μg per leaf disc in the fifth sample of test F_3 and the third sample of test F_4 resulted in only 4.5 % and 3.5 % kills, respectively (table 14). From tables 14 and 16 other comparisons can be extracted all confirming the higher effectiveness of parathion deposits in the present tests over comparable deposits in the temperature experiments.

4. DISCUSSION

DDT has been frequently described as being "practically insoluble in water". Its true solubility was found by RICHARDS (1944) to be 0.0002 p.p.m. On the other hand, parathion was reported by MARTIN (1957) to be slightly soluble in water to the extent of 25 p.p.m. These values show that although parathion is not highly soluble in water it, nevertheless, has much greater solubility than DDT. This suggests that whereas the detrimental effect of simulated rain on DDT deposits in the present tests was mostly mechanical, the loss of parathion residue occurred through erosion as well as by dissolution of the insecticide itself.

It would appear that the serious losses suffered by parathion deposits (table 16), as compared with the slight decrease in DDT residues (table 15) during the present rain experiments may be attributed either to its greater solubility in water or perhaps to the properties of the other components included in its formulation. Unfortunately, the details of the formulations of these commercial insecticides have not been published.

Figure 15 shows the lines obtained when the logarithm of the per cent initial residues remaining in experiments G_1 , G_2 , G_3 and G_4 were plotted against time. DDT deposits exposed to six treatments of ± 3 mm simulated rain each gave a straight line representing a very low and almost regular rate of loss over the six days of the test. Residues of the same toxicant exposed to ± 6 mm simulated rain daily yielded a broken line, showing that the rate of decrease was higher during the first three days of the experiment than during the latter days. The two lines of the DDT tests approximate toward the end of the experiments indicating that equal losses of material would have resulted from both tests if further applications of simulated rain had been made. This may mean that the amount of residue removable by rain is limited and that the increase in quantity of rain results in the removal of these amount in a shorter time without seriously increasing the final loss of material.

Parathion deposits exposed to ± 3 mm or ± 6 mm simulated rain daily for

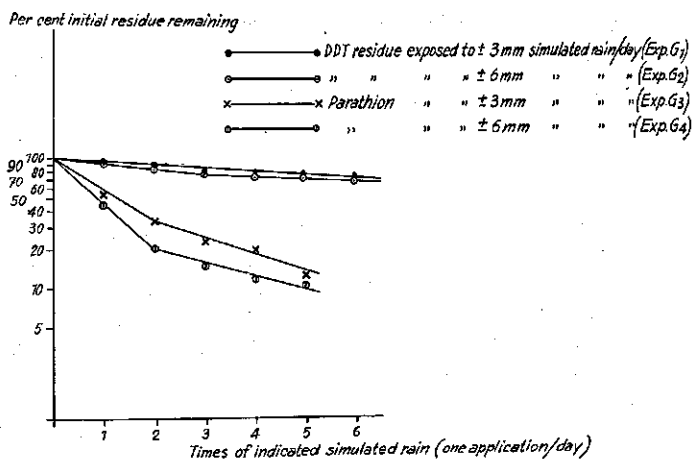


FIG. 15. Rates of loss of DDT and parathion residues caused by successive applications of ± 3 mm or ± 6 mm simulated rain.

five days both gave broken lines. Each line shows that the rate of loss caused by the first two applications of simulated rain was higher than that of the latter applications. The continued steepness of both lines indicates, nevertheless, that the rate of decrease remained high right to the end of both experiments.

Figure 15 also shows the enormous difference between the rates of loss of DDT and parathion residues under the present experimental conditions.

The increased effectiveness of several samples of both insecticides during rain experiments may be due to repeated exposure of fresh material as a result of washing.

C. Radiation Experiments

1. LITERATURE

GUNTHER *et al.* (1946) mentioned that far-ultraviolet radiation may induce photo-decomposition of DDT. LINDQUIST *et al.* (1946) studied the effect of ultraviolet light on DDT deposits formed on unpainted plywood boards, glass Petri dishes and plates by spraying them with solutions of the toxicant in kerosene and various auxiliary solvents to give a density of 200 mg per sq. ft. The residues were exposed at the distance of 55 inches to radiation from a 36 inch, G.E., 30-watt tube provided with a silvered reflector. It was believed that most of the energy was emitted in the range 2500–2600 Å. Irradiation for 88 or 132 hours reduced the toxicity of the deposits to houseflies in all cases, but it was found that the reduction in effectiveness was much less in the case of residues from solutions containing rather volatile auxiliary solvents that evaporate more quickly. DDT residues on glass plates were irradiated for several days until they became almost ineffective and were then treated with acetone. After evaporation of the acetone these deposits were as rapid in their knock down effect to exposed flies as they had been before the ultraviolet treatment. The authors commented that acetone had dissolved and redistributed the whole deposit and thus exposed to the surface some of the undecomposed DDT which was below the inactive

surface layer produced by the irradiation. FLECK (1949) studied the decomposition of DDT dissolved in γ -valerolactone and irradiated with ultraviolet light from a 100-watt mercury-vapour lamp. He showed that ultraviolet light catalyses the decomposition of DDT to 4,4' dichlorobenzophenone when air is present. The formation of the latter compound from DDT has been assumed to proceed first by elimination of hydrogen chloride from the trichloroethane group and then by oxidation of the resulting double bond. FLECK also found that 4,4' dichlorobenzophenone which accumulated outside irradiated crystals of DDT exert a marked protectant action on the intact insecticide. BLACKITH (1952*b*) mentioned that SAZENOV & ANDRÉEV (1949) have shown that decomposition of DDT by sunlight, measured by residual toxicity to houseflies, is almost entirely due to the ultraviolet component of sunlight. BLACKITH (1952*b*) used grain weevils in a bioassay to estimate the extent of decomposition of DDT exposed to ultraviolet radiation emitted by an Osira G.E.C. 125-watt, high-pressure mercury-vapour discharge lamp with the outer evacuated glass envelope removed. He found that DDT which had been dissolved in a mixture of volatile and non-volatile oils and applied to filter papers, decomposed at a rate of 25 % of the original quantity of material in 15 minutes. The author stated that the lamp yielded a substantially continuous spectrum ranging from about 2000 Å up to the visible region. BURT & WARD (1955*a*) reported that DDT applied with cabbage wax dissolved in toluene to glass plates at a density of 0.34 μg per sq. cm lost 95 % of its DDT content, as shown by chemical analyses, in 30 minutes when exposed at a distance of 6 inches from an ultraviolet lamp giving a high proportion of radiation at a wavelength of 2537 Å. Irradiation of crystalline DDT residues of about 1 μg per sq. cm on plain glass for the same period caused a loss of about 30 %. The authors commented that the difference in loss may be due either to a greater sensitivity of DDT when it is in solution, or to the greater area exposed to the action of radiation in the case of less dense deposits.

FRAWLEY *et al.* (1958) investigated the effect of ultraviolet light on parathion deposits formed in Petri dishes at the rate of about 5.1 mg per sq. in. The deposits were irradiated by a G.E. Uviark ultraviolet lamp placed 16 inches above the dishes. Regular sampling every two hours showed that the material suffered progressive losses which amounted to about 50 % of the initial quantity after 16 hours of irradiation as detected by chemical analyses and fly bioassay.

2. EXPERIMENTAL

Light-induced decomposition of insecticide residues has been mentioned in several papers. Most of these experiments are either field tests or laboratory experiments with filter paper or glass plates as a substratum. Studies on the effect of radiation upon insecticide residues applied to plant surfaces would be of great importance especially to sunny and arid regions where quick loss of insecticide residues is frequently observed.

The radiation tests in the present work were mostly concerned with the effect of a radiation spectrum resembling that of the sun upon DDT and parathion residues on a plant surface.

Three Philips "Biosol" lamps, two of Type A (each of 280 watt) and one of type B (510 watt), were used in these experiments. They were all provided with filters and reflectors. Leaves carrying the test residues were arranged on a table

TABLE 17. Radiation intensity at various wavelengths emitted by the three "Biosol" lamps together at a range of 75 cm.

Wavelength Å	Radiation intensity in 1000 ergs/cm ² /sec.	Wavelength Å	Radiation intensity in 1000 ergs/cm ² /sec.
3655	20.20	2804	0.41
3342	1.45	2753	0.086
3130	9.00	2699	0.067
3022	3.08	2652	0.200
2967	1.25	2537	0.064
2894	0.35	2483	0.007

around which the three "Biosol" lamps were fixed in such a way that the radiation reaching the surface of the table was as evenly distributed as possible. The lamps were placed at a distance of 75 cm above the table and remained at this position during all the radiation tests. The surface of the table was then divided into 64 equal squares and radiation intensity checked in each square by taking two readings with a Lux-meter for each square. Leaves were then exposed only in those squares giving a variation of less than 10 % as judged by L.m. readings.

An approximate estimation based on the data published by the manufacturers of the lamps (Philips, Holland) indicates that the radiation intensity at various wavelengths resulting from the three lamps together at 75 cm was as shown in table 17.

Two experiments were conducted with each of the two insecticides. In the first the residues were irradiated in a dry condition and in the second whilst wet. The latter was achieved by blowing a mist of fine water droplets from a small atomizer every 5-10 minutes over the exposed leaves so that their upper surfaces were kept slightly moist.

Room temperature was recorded every hour. Surface temperatures of the leaves were measured every 30 minutes by means of thermocouples and a galvanometer. These temperatures are shown in table 18.

By means of preliminary experiments, the most suitable periods of irradiation were determined for each insecticide. In both dry and wet experiments DDT residues were irradiated for 3, 6, 9 and 12 hours. Parathion residues, on the other hand, were irradiated for $\frac{1}{2}$, 1, 2 and 4 hours. Measurements of leaf surface temperature during the preliminary experiments indicated that 35°C was the highest temperature and that this temperature persisted for some time. Therefore, in order to extract the effect of radiation alone from the combined effects of radiation and temperature, two temperature experiments, one with DDT and another with parathion residues, were run simultaneously with the correspond-

TABLE 18. Records of average room and leaf surface temperatures during light tests with DDT and parathion.

Record	DDT experiments		Parathion experiments	
	dry	wet	dry	wet
Average room temperature	21.5°C	20°C	21°C	20.5°C
Average leaf surface temperature	34°C	30°C	32.5°C	29.5°C

ing dry radiation tests. In these temperature tests the residues of each insecticide were kept at 35°C in complete darkness inside a constant temperature cabinet. Two samples were taken in each case; with DDT the samples were taken after 6 and 12 hours, and with parathion after 2 and 4 hours.

3. RESULTS

a. DDT residue

The results of the two light experiments H₁ and H₂ are given in table 19 and figure 16. In experiment H₁ the residues were irradiated in a dry condition, whereas in experiment H₂, the deposits were kept moist during the whole irradiation period. The results of two further samples kept in darkness at 35°C for 6 and 12 hours are also presented.

The leaf surface temperatures recorded during these experiments indicate that the loss in material might result from both light radiation and high temperature. The samples held in darkness show that 27.7 % of the initial residue was lost in 12 hours. Since the average leaf surface temperature was only 34°C, and as this high temperature did not continue during all the period of irradiation, the loss attributable to high temperature may be estimated as 6 µg DDT per leaf disc.

During 12 hours at 35°C in darkness the initial residue lost twice as much as during the first 6 hours which means that the loss occurred regularly during the exposure period at a rate of 0.5 µg DDT per leaf disc per hour. The results of test H₁ are corrected for the effect of temperature by the application of this rate. The densities of deposits in test H₁, together with their corrected values, are given in table 20. The mortalities to be expected from these deposits were estimated by interpolation with the straight regression lines obtained by plotting, on probit mortality-log dosage paper, the per cent mortalities of tests H₁ and H₂ against the corresponding doses measured in µg DDT per leaf disc. The regression lines of the two experiments (figure 17) are practically identical.

TABLE 19. Effect of irradiating wet and dry DDT residues with "Biosol" lamps for 12 hours, as detected chemically and biologically. Residues are computed as µg DDT per sq.cm and per leaf disc (21.25 cm²).

Irradiation period in hours	Experiment H ₁						Experiment H ₂					
	µg per leaf disc	µg per cm ²	% mortality	% decrease in residue	% decrease in residue per 3 hours	% decrease in mortality	µg per leaf disc	µg per cm ²	% mortality	% decrease in residue	% decrease in residue per 3 hours	% decrease in mortality
—	23.5	1.105	92	—	—	—	24.25	1.141	93.5	—	—	—
3	15.5	0.729	61.5	34.0	34.0	33.2	17.75	0.835	72	—	—	22.9
6	13	0.611	34	44.7	10.7	63.0	14.25	0.670	46	26.8	26.8	50.8
9	10	0.470	10	57.5	12.8	89.1	13.25	0.623	36	41.2	14.4	61.5
12	7.5	0.353	2	68.1	10.6	97.8	12.5	0.588	25.5	45.4	4.2	72.7
										48.5	3.1	
Period in hours	Temperature test at 35°C in darkness											
6	20.25	0.952	83	13.8	—	9.8						
12	17	0.800	70.5	27.7	—	23.4						

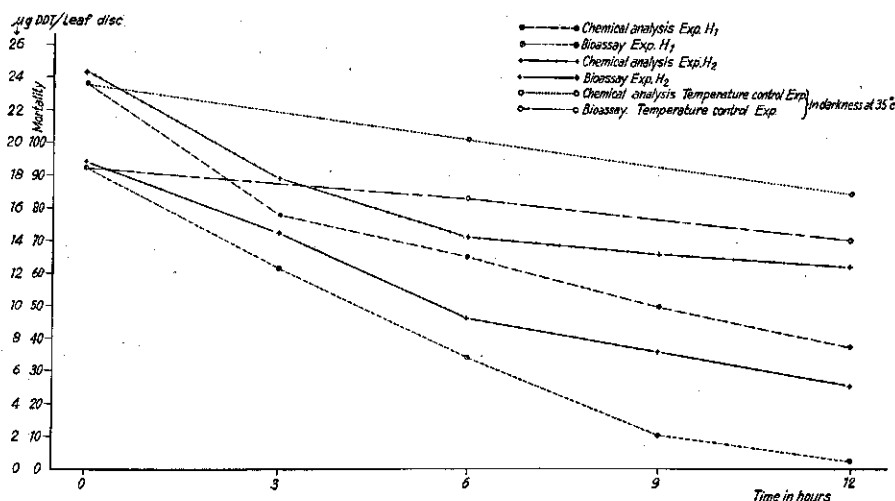


FIG. 16. DDT residues irradiated with "Biosol" lamps in a dry or a wet condition for 12 hours as detected chemically and biologically by sampling every 3 hours (experiments H₁ and H₂).

The per cent mortalities given in table 20 represent the average interpolated values obtained from the two regression lines.

The results of experiment H₂ cannot be presented in a similarly corrected form to show the effect of light alone owing to the absence of appropriate data. Since the average leaf surface temperature in test H₂ was, however, only 30°C, or 4°C lower than that of test H₁, it is believed that the detrimental effect of temperature was less in the former experiment.

Table 20 shows that the first 3 hours of irradiation reduced the initial residue of experiment H₁ by 27.7 %. Another 9 hours of irradiation resulted in a further 14.9 % loss of the original deposit. This means that the initial deposit was reduced at the rate of about 9.2 % per hour during the first three hours as compared with the much lower rate of about 1.65 % per hour during the next nine hours.

In experiment H₂, a reduction of 26.8 % in the initial residue took place during the first three hours of irradiation. This loss is very similar to that found in test H₁, after being corrected for the effect of temperature over the same period. The per cent loss of the initial deposit in test H₂ decreased to 14.4, 4.2

TABLE 20. Net radiation effect on DDT deposits during 12 hours exposure to "biosol" lamps.

Irradiation period in hours	µg per leaf disc (Exp. H ₁)	Loss caused by temperature per 3 hours µg p. leaf disc	Corrected residues µg per leaf disc	% mortality	% decrease in residue	% decrease in residue per 3 hours	% decrease in mortality
—	23.5	—	—	92	—	—	—
3	15.5	1.5	17	68	27.7	27.7	26.1
6	13	3	16	60	31.9	4.2	34.8
9	10	4.5	14.5	47	38.3	6.4	48.9
12	7.5	6	13.5	39	42.6	4.3	57.6

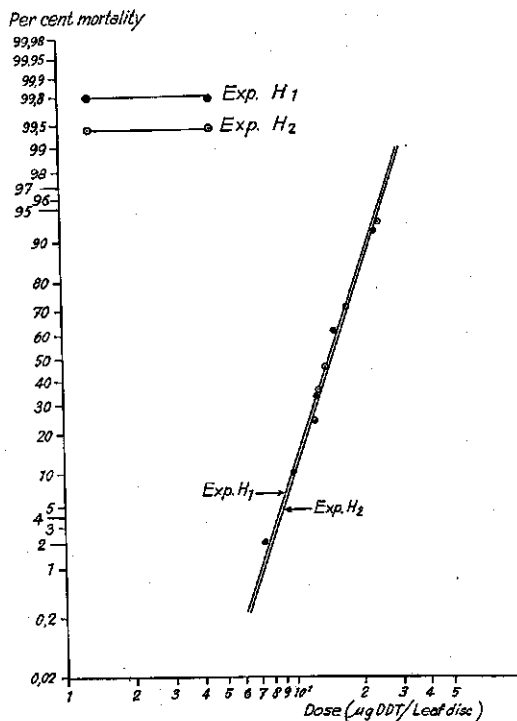


FIG. 17. Dosage-mortality regressions for light experiments H_1 and H_2 with DDT residues.

and 3.1 during the second, third and fourth 3-hour periods, respectively. Thus the mean rates of loss for the four 3-hour periods of this test are about 8.9 %, 4.8 %, 1.4 % and 1 % per hour.

TABLE 21. Effect of irradiating wet and dry parathion deposits with "Biosol" lamps for 4 hours, as detected chemically and biologically. Residues are computed as μg parathion per sq. cm and per leaf disc (21.25 cm^2).

Irradiation period in hours	Experiment H_1						Experiment H_2					
	μg per leaf disc	μg per cm^2	% mortality	% decrease in residue	% decrease in residue per interval	% decrease in mortality	μg per leaf disc	μg per cm^2	% mortality	% decrease in residue	% decrease in residue per interval	% decrease in mortality
—	16.5	0.776	96.5	—	—	—	16.75	0.788	97	—	—	—
0.5	14	0.658	96	15.2	12.1	0.5	15.5	0.729	98	7.5	7.5	+1.0
1	12	0.564	83	27.3	12.1	14.0	14.5	0.682	96.5	13.4	5.9	0.5
2	10	0.470	34.5	39.4	12.1	64.3	12.5	0.588	90	25.4	12.0	7.2
4	6.25	0.294	2	62.1	22.7	97.9	8.75	0.411	19	47.8	22.4	80.4
Period in hours	Temperature test at 35°C in darkness						+ The mortality increased by this per cent in spite of the decrease in residue					
2	13.25	0.623	93	19.7	—	3.6						
4	12.25	0.576	82.5	25.8	—	14.5						

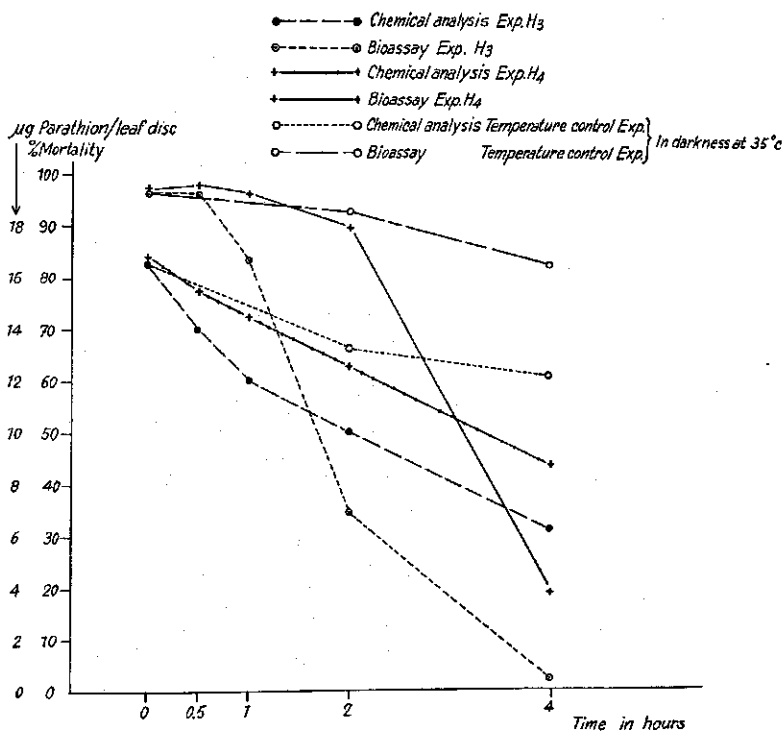


FIG. 18. Parathion residues irradiated with "Biosol" lamps in a dry or a wet condition for 4 hours as detected chemically and biologically at indicated intervals (experiments H₃ and H₄).

b. Parathion residue

Table 21 and figure 18 present the results of the radiation experiments with dry and wet parathion residues which are referred to as tests H₃ and H₄, respectively. The results of two further samples kept in darkness at 35°C for 2 and 4 hours are also given.

Losses detected in the material were again caused by the joint effect of high temperature and light radiation. This was especially true in the dry irradiation experiment. Parathion residues kept in darkness at 35°C lost 3.25 µg per leaf disc within the first 2 hours and an additional 1 µg per leaf disc after 2 hours

TABLE 22. Net radiation effect on parathion deposits during 4 hours exposure to "Biosol" lamps.

Irradiation period in hours	µg per leaf disc (Exp. H ₃)	Loss caused by temperature per interval µg p. leaf disc	Corrected residues µg per leaf disc	% mortality	% decrease in residue	% decrease in residue p. interval	% decrease in mortality
—	16.5	—	—	96.5	—	—	—
0.5	14	0.75	14.75	96	10.7	10.7	0.5
1	12	1.5	13.5	92	18.2	7.5	4.7
2	10	3	13	87.5	21.2	3.0	9.3
4	6.25	3.75	10	44	39.4	18.2	54.5

more. As the actual average temperature affecting the deposits during experiment H_3 was only 32.5°C , the loss attributable to temperature may be estimated at $3\text{ }\mu\text{g}$ per leaf disc for the first two hours and $0.75\text{ }\mu\text{g}$ per leaf disc for the next two hours. Owing to the fact that the initial loss occurred within rather a short time (2 hours) it will be assumed that the material was lost regularly during the two hours, viz. at the rate of $0.75\text{ }\mu\text{g}$ per leaf disc per half-hour. When the densities of the residues of test H_3 are corrected at this rate for the effect of high temperature, the samples taken after 30 minutes, one and two hours are increased by 0.75 , 1.5 and $3\text{ }\mu\text{g}$ per leaf disc, respectively. The residue sampled after 4 hours of irradiation is increased by $3.75\text{ }\mu\text{g}$ per leaf disc. These corrected densities represent the deposits remaining after experiencing only the effect of radiation. Mortality corrections were applied in the same way as for DDT. The regression lines of experiments H_3 and H_4 are shown in figure 19.

The results of the wet irradiation experiment were not corrected for the effect of temperature because appropriate correction data had not been determined. As the average leaf surface temperature was, however, 3°C lower in test H_4 than in test H_3 , it is fair to expect that the effect was less in the former experiment than in the latter.

Irradiation of deposits for the first 30 minutes in test H_4 reduced them by 7.5% which is 3.2% less than the loss corrected for temperature in experiment H_3 . This means that the effect of temperature during the first 30 minutes must have been very small. The reduced radiation effect may also be attributed to the presence of the thin water film over the deposits.

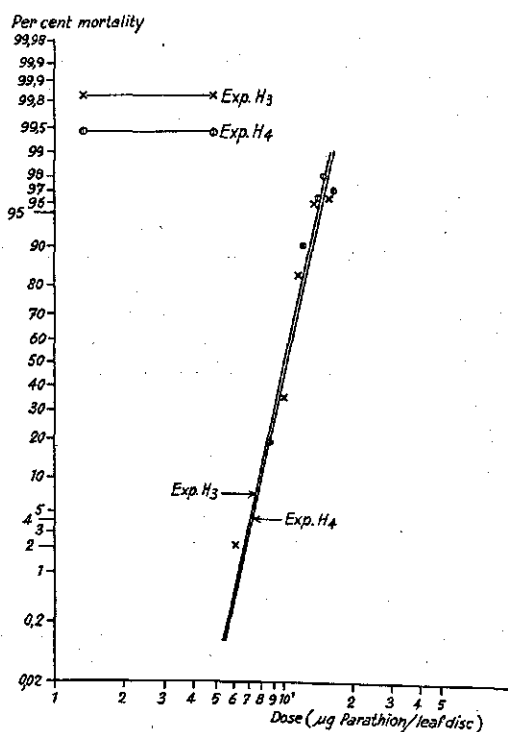


FIG. 19. Dosage-mortality regressions for light experiments H_3 and H_4 with parathion residues.

4. DISCUSSION

Evidence has been accumulating to show that many insecticide residues suffer losses on exposure to sunshine. DDT, which is known to have a prolonged residual action, has been found to be not so stable when exposed to direct sunlight. Since DDT loses nothing of its effectiveness during storage for long periods under indirect light conditions, the chemically active short ultraviolet light waves are suspected to be responsible for the detrimental effect of exposure to direct sunlight.

The importance of the shorter wavelengths to the decomposition of pyrethrum powder was mentioned by BLACKITH (1952a) who stated that blue light was slightly more active than red. Furthermore, when filter papers impregnated with pyrethrin solution in oil were irradiated with either an ultraviolet source or a normal 60-watt pearl tungsten filament lamp for 13 and 25 minutes, respectively, almost no pyrethrum breakdown occurred in case of irradiation with visible light although there was a significant decrease in the material exposed to ultraviolet radiation.

NASIR (1953) clearly showed the ultraviolet range to be most effective as regards the photolysis of DDT and pyrethrum films. He used BLACKITH's (1952a & b) technique to estimate the degree of decomposition in material. The exposure time was 20 minutes in all cases. This author found that decomposition of DDT occurs most quickly at wavelengths of 2200–2400 Å, although decomposition takes place at all wavelengths up to 2850 Å. Almost the same wavelengths were also found to be the most destructive towards pyrethrum films.

The rather long period of irradiation needed in the present tests to obtain serious decomposition with either DDT or parathion residues contrasts with the short time that was sufficient to cause considerable losses in the experiments conducted by BLACKITH (1952) and NASIR (1953). This may be due to the low radiation intensities at the effective wavelengths (less than 2850 Å) in the spectrum emitted by the lamps used in the work herein described. The shortest wavelength used here was 2484 Å which is longer than the most effective wavelengths mentioned by NASIR. On the other hand, the increased effectiveness of irradiation in the tests of the above-mentioned authors may be due to the fact that they irradiated DDT or pyrethrum dissolved in oils. It has been shown by LINDQUIST *et al.* (1946) and BURT & WARD (1955a) that DDT decomposes more rapidly when irradiated in solution than in a solid state. DDT was almost completely in a solid condition when irradiated in the present experiments. Relatively long irradiation periods were also needed to induce a serious loss in parathion which is liquid at ordinary temperatures and is known to be much less stable than DDT. This indicates again that in the present experiments it was not the conditions of the materials which was largely responsible for the low effectiveness of the irradiation but rather the low radiation intensity of the shorter, effective wavelengths and the complete absence of the very short wavelengths (lower than 2400 Å).

There was an excess of 19.6 % in the final loss of material in experiment H₁ over that which occurred in experiment H₂. Similarly, the final decrease in the initial deposits in experiment H₃ was 14.3 % more than that produced in experiment H₄. It does not seem reasonable that a difference of only 4°C and 3°C in the average leaf surface temperature in the wet irradiation tests as

compared with the dry ones could be the only cause of these considerable differences in the final losses. Most probably the water films in the wet irradiation tests with either toxicant reduced the final losses by lowering both the leaf surface temperature and the intensity of radiation that affected the deposits.

Sunlight may affect insecticide deposits by raising their temperature, or by the independent action of its ultraviolet component. In fact, the losses of toxicants exposed to direct sunlight are the result of both effects. As the present results have shown that moisture does not enhance the effect of radiation but instead rather reduces it, it seems clear that in arid and sunny regions, radiation and temperature are the most important weather factors limiting the residual persistence of insecticides.

The decrease in DDT and parathion deposits caused by temperature and simulated rain were shown to occur geometrically. It was found advantageous to plot the results of the light experiments with either toxicant on semi-log paper in the same way as the temperature and rain experiments. This method illustrates the rate or rates of loss within each experiment and facilitates a comparison of the rates of decrease in different tests. The curves of the light tests with DDT and parathion residues are shown in figures 20 and 21, respectively, together with the corresponding dry irradiation test after correction for the effect of temperature. With DDT residues (figure 20) the loss of material was in general at a lower rate in experiment H_2 than in experiment H_1 , but in both cases the decrease in material was highest within the first three hours of irradiation. The loss caused by light alone was only very slightly greater than that in test H_2 during the initial period of the experiment, and the difference decreased still more as irradiation was continued for the next nine hours.

The detrimental effect of irradiation during the first hour of experiment H_3 (figure 21) was greater than it was during the succeeding three hours. By contrast the rate of loss was almost regular during the whole period of irradiation in experiment H_4 and was in general lower than that in experiment H_3 . Exposure to light alone in the latter experiment caused greater loss within the first hour than it did in experiment H_4 during the same period.

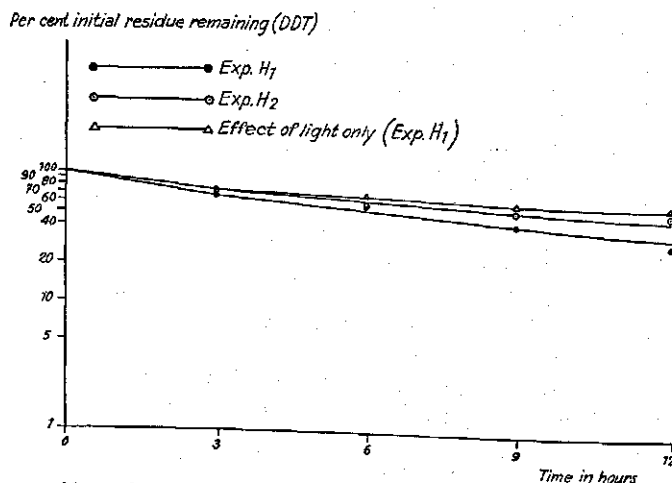
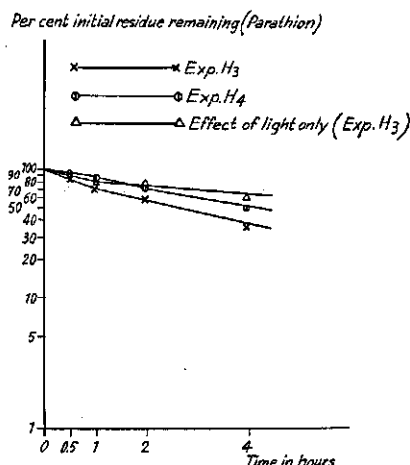


FIG. 20. Rates of loss of DDT residues irradiated with "Biosol" lamps for 12 hours.

FIG. 21. Rates of loss of parathion residues irradiated with "Biosol" lamps for 4 hours.



Twelve hours of exposure to ultraviolet light alone reduced the initial residue of DDT by 42.6 %, whereas only four hours of irradiation of parathion using the same light intensity caused 39.4 % loss. This shows that parathion is more seriously affected by light than DDT.

The increase of 1 % in the mortality caused by the residue of the second sample in experiment H₄ took place in spite of a decrease of 7.5 % in the quantity of material as compared with the initial deposit and could be explained by an increase in the density of material present at the surface of the leaves. This increase may have been achieved by the re-issuing of parathion from inside the plant tissue on to the surface. The enhanced mortality indicates that the amount of toxicant that returned to the surface was more than the quantity lost.

SUMMARY AND CONCLUSIONS

I. INTRODUCTION

In a study of weathering of DDT and parathion deposits on a plant surface, special attention was paid to temperature, rain, and ultraviolet radiation. Residues were evaluated by chemical analysis (total quantity present) and by contact bioassay (surface activity). In comparing the relation between the total quantity of insecticide residue evaluated chemically, and the mortality as shown by contact bioassay, with the dosage-mortality regression lines as obtained with sprayed glass plates, it proved possible to gain an impression of the penetration of the insecticides into the leaf tissues.

II. MATERIALS AND METHODS

The upper surface of full-expanded ivy leaves (*Hedera helix*) was used as the standard plant surface throughout the experiments. *Calandra granaria* L. was selected as the test insect. 25 % DDT (Gesarol) emulsifiable concentrate (Organchemia, Boxtel) and 25 % parathion e.c. (Philips-Duphar, Amsterdam) were used as standard formulated insecticides.

Spraying was done by means of the TEN HOUTEN & KRAAK spraying tower and nozzle. The coefficient of variation did not exceed 6 %.

For the contact bioassay, the method of LOOSJES (1952) was followed with some minor modifications.

For DDT analysis, the method of SCHECHTER *et al.* (1945) was followed, modified after AMSDEN & WALBRIDGE (1945).

Parathion analysis was carried out according to the method of AVERELL & NORRIS (1948) modified by GAGE (1950) and ZEUMER & FISCHER (1952).

III. FIELD EXPERIMENTS

During the warm, sunny and relatively dry spring and early summer of 1959 and 1960, residues on leaves were subjected to weathering in the open, on suitably arranged wooden trays. The experiments in 1959 were of a preliminary character, maximum leaf surface temperature and daily rainfall being read. In 1960, air and leaf surface temperatures and rainfall were recorded continuously.

Temperature and solar radiation most probably were the main factors limiting the residual effect of both parathion and DDT. Rainfall was scarce and played a minor role. High mean temperatures proved to be more important than occasionally high maxima.

DDT residues were much more persistent than parathion residues, the final loss of DDT after six days being almost equal to that of parathion after 48 hours only. However, a fraction of the parathion residue persisted for many days, decreasing very slowly. Apparently, this fraction was present inside the leaf tissues.

Comparison with dosage-mortality regression lines obtained on glass plates revealed that next to parathion, DDT penetrates through the plant surface, but only to a far less degree. Re-issuance of this "hidden" residue to the plant surface may result in an increase in residual effect, but is mostly masked by weathering.

IV. LABORATORY EXPERIMENTS

A. Temperature experiments

Use was made of slowly ventilated constant temperature cabinets. Treated leaves were kept in darkness at 25 and 35°C (DDT exps.), or at 20 and 30°C (parathion exps.). "Spontaneous" losses as accelerated by increase in temperature generally showed a geometrical decline.

At high temperatures, losses mainly occurred within the initial period of exposure, after which the rate of decrease diminished appreciably. At moderate temperatures, initial losses were much less but continued at a higher level over a longer period.

Parathion residues proved to be much more temperature-sensitive than DDT residues.

B. Rain experiments

A rain apparatus was constructed in which treated leaves were exposed to 3 and 6 mm of simulated rain daily during 5-6 days.

In case of DDT, the deposits showed a slow and almost linear decrease, being not proportional to the quantity of rain applied, the lines running almost parallel in the 3 and 6 mm experiments.

In case of parathion, the curves of the chemically determined residue showed a steep geometrical initial decline followed by a slow decrease at the sublethal level. After two applications, the curves at 3 and 6 mm ran almost parallel, indicating that losses are not proportional to rainfall also in the case of parathion residues.

C. Radiation experiments

Radiation tests were carried out using Philips "Biosol" lamps, two of type A (280 W) and one of type B (510 W), provided with filters and reflectors. The range of wavelengths was from 2483–3655 Å. Exposure times were 3, 6, 9 and 12 hrs for DDT, and $\frac{1}{2}$, 1, 2 and 4 hrs for parathion. Results were corrected for increase in temperature.

Exposure of DDT residue during twelve hours reduced the mortality in the contact bioassay from 90 % to practically zero. In parathion, a similar effect was obtained within 4 hours. In both cases, the remaining quantities of residue would have caused a considerable mortality percentage, if sprayed on glass plates of the same surface area.

In a similar series of experiments, leaves were kept moist during exposure to radiation. This moistening generally had a protective effect against radiation.

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SAMENVATTING

HOOFDSTUK I: INLEIDING

In dit onderzoek, dat betrekking heeft op de invloed van weersfactoren op residu's van DDT en parathion op een plantaardig oppervlak, is in het bijzonder aandacht geschonken aan temperatuur, regenval en ultraviolette straling. De residu's werden bepaald langs chemische weg (totale hoeveelheid insecticide) en door middel van biologische toetsing (residu-werking bij contact). Ter vergelijking werden dosering-mortaliteitscurven opgesteld, verkregen door het spuiten van de insecticiden op glazen platen. Door vergelijking van beide typen proeven kon worden nagegaan, in welke mate de insecticiden in het blad binnendringen.

HOOFDSTUK II: MATERIAAL EN METHODEN

De bovenzijde van volgroeide bladen van klimop (*Hedera helix*) werd als standaard-oppervlakte gebruikt. Proefinsect was de graanklander (*Calandra granaria* L.). Als standaard-insecticiden werden gebruikt 25 % emulgeerbaar DDT (Gesarol, Orgachemia, Boxtel) en 25 % emulgeerbaar parathion (Philips-Duphar, Amsterdam). Het toedienen der insecticiden geschiedde met behulp van het spuitapparaat volgens TEN HOUTEN & KRAAK. De variatie-coëfficiënt van de hoeveelheid spuitneerslag per oppervlakte-eenheid was niet hoger dan 6 %. De biologische toetsingen werden uitgevoerd volgens de methode van LOOSJES (1952) met enkele geringe modificaties. De chemische analyse van DDT geschiedde volgens SCHECHTER *et al.* (1945), de parathion-analyse volgens AVERELL & NORRIS (1948).

HOOFDSTUK III: VELDPROEVEN

Gedurende de warme, zonnige en vrij droge lente en voorzomer van 1959 en 1960 werden residu's op bladeren aan de weersomstandigheden blootgesteld op speciaal hiervoor vervaardigde rekken. De proeven van 1959 droegen een voorlopig karakter. Maximum bladtemperatuur en dagelijkse regenval werden afgelezen. In 1960 werden oppervlakte-temperatuur en regenval continu geregistreerd. Temperatuur en zonnestraling waren gedurende de proefperiode waarschijnlijk de voornaamste beperkende factoren voor de residu-werking van parathion en DDT. De regenval was gering en had geen belangrijk effect. Hoge gemiddelde temperaturen bleken van groter belang dan incidenteel hoge maxima. De bestendigheid van DDT-residu's was veel groter dan die van parathion. De afname van het DDT-residu na zes dagen was ongeveer gelijk met die van het parathion-residu na 48 uur. Een zekere fractie van het parathion-residu bleef echter aanwezig zonder mortaliteit te veroorzaken. Deze fractie bevond zich klaarblijkelijk in het inwendige van het blad. Vergelijking met proeven met glazen platen gaf tot resultaat, dat zowel DDT als parathion in de plant binnen-

dringt, hoewel DDT in veel geringer mate. Het aan de oppervlakte terugkeren van dit "verborgen" residu kan de residu-werking verhogen, maar wordt in de regel door verwerking te niet gedaan.

HOOFDSTUK IV: LABORATORIUMPROEVEN

A. Temperatuurproeven

In thermostaten met langzame ventilatie werden de planten blootgesteld aan temperaturen van 25 en 35°C (DDT), resp. 20 en 30°C (parathion). De "spontane" vermindering van het residu, die wordt versneld door stijging in temperatuur heeft een geometrisch verloop. Bij hoge temperaturen is de lijn steiler dan bij gemiddelde temperaturen. De temperatuurgevoeligheid van parathion residu's is groter dan die van DDT.

B. Beregeningsproeven

Met behulp van een speciaal geconstrueerd beregenings-apparaat werden dagelijkse hoeveelheden van 3 en 6 mm regenval nagebootst gedurende 5-6 dagen. In het geval van DDT vertoonde het residu een bijna lineaire afname, niet evenredig met de hoeveelheid toegediende beregening. In het geval van parathion vertoonde de residu-curve een steile geometrische afname, gevolgd door een langzame afname op sublethaal niveau. Ook hier was de afname niet evenredig met de hoeveelheid beregening.

C. Bestralingsproeven

De residu's werden bestraald door middel van Philips Biosollampen, twee van type A (280 W) en één van type B (500 W). De golflengte was 2483 tot 3655 Å. De expositietijd varieerde van 3-12 uur voor DDT en van $\frac{1}{2}$ -4 uur voor parathion. Correcties voor temperatuurstijgingen werden toegepast. Bestraling van een DDT-residu gedurende 12 uur reduceerde de sterfte in de biologische toetsingen van 90 % tot vrijwel nihil. Een dergelijk effect werd bereikt met parathion binnen 4 uur. In beide gevallen was het chemisch bepaalbare residu na afloop der bestraling voldoende om, op glazen platen gebracht, een belangrijke sterfte te veroorzaken. Dit zou kunnen wijzen hetzij op plaatselijke inactivering, hetzij op penetratie in het blad. Bevochtiging van het bladoppervlak gedurende de bestraling had een beschermende invloed tegen het stralingseffect.

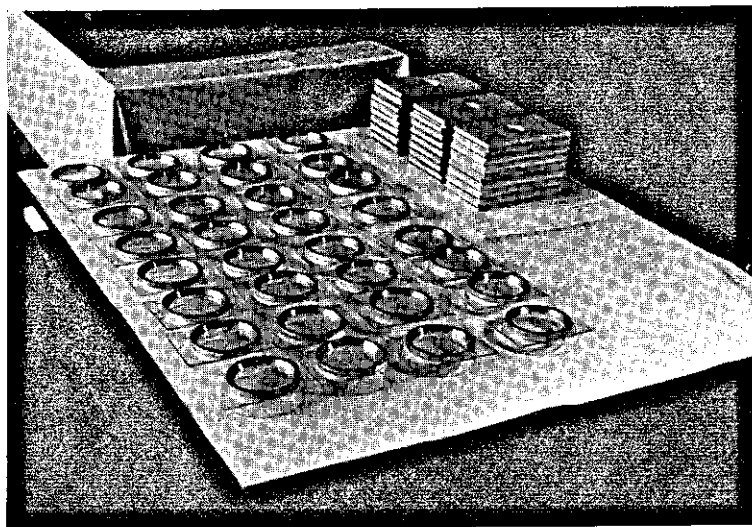
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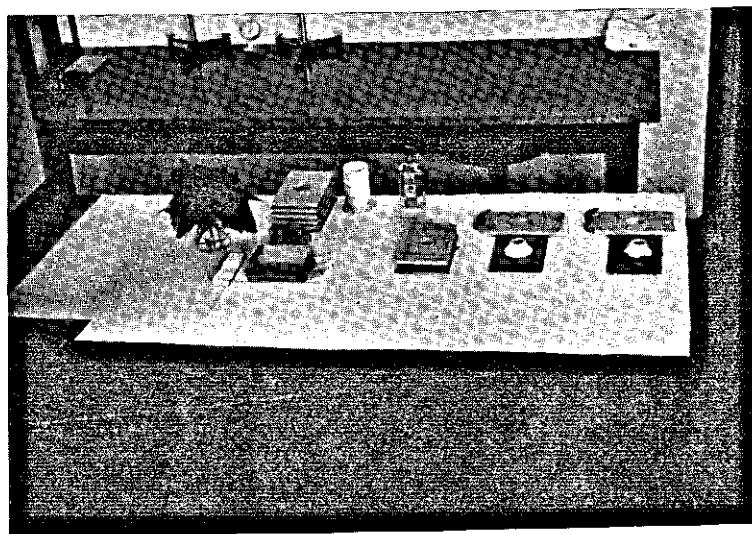
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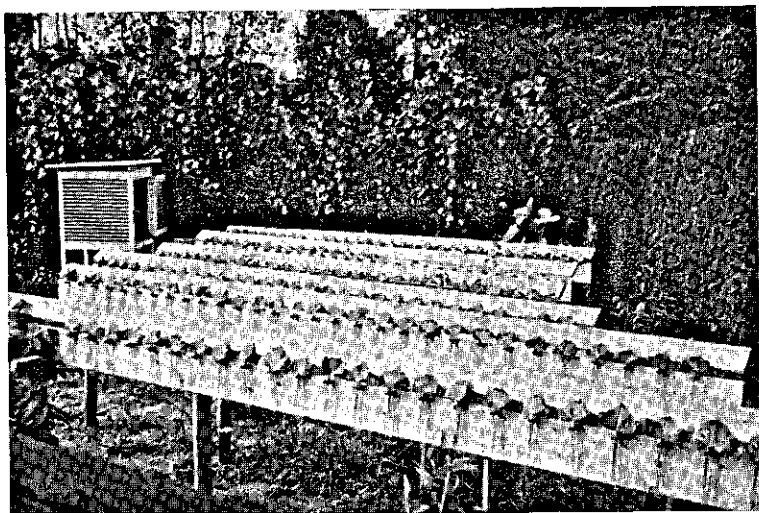


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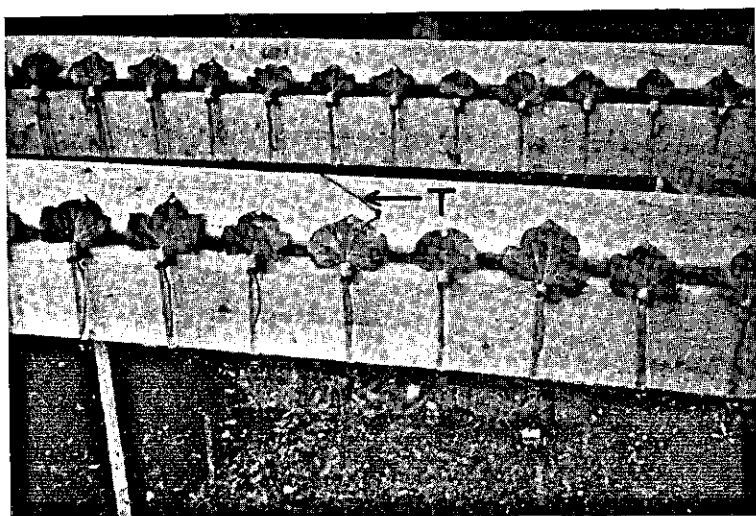


b

PLATE 1. a. Glass plates sprayed and left to dry at room temperature before testing the residues.
b. Preparing glass plates or ivy leaves for bioassay.



a



b

PLATE 2. a. Sprayed ivy leaves arranged on wooden trays for weathering.
b. Ivy leaves fixed on the wooden trays for weathering. Notice the thermocouple (T) pressed on a leaf.