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Insecticides and Fungicides Department

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DEPARTMENT OF INSECTICIDES AND FUNGICIDES

By F. TATTERSFIELD

During 1946 Dr. Potter was invited as a visiting professor for a year's stay in the United States by the Rhode Island Agricultural Research Station. He left England at the end of December and has spent nine months at Rhode Island and three months at the Agricultural Research Station, Connecticut. At the end of his stay he paid flying visits to other stations in the United States and in Canada.

For the purpose of this report the work has been divided into chemical and biological sections, but throughout it has been closely interlocked and mutual help has been given.

CHEMICAL WORK

The evaluation of pyrethrum flowers

This problem was raised in different ways during the year. In conjunction with the Imperial Institute we were called upon by the Kenya Pyrethrum Growers Association to recommend methods for the chemical evaluation of pyrethrum flowers.

In 1944-45 collaboration of this department with the Imperial Institute, Dr. S. H. Harper and Messrs. Stafford Allen had resulted in the re-determination of the factor used in determining pyrethrum I by the Wilcoxon method. This work has been drawn together and is ready for publication. The problem now is to draw up a method for applying either this or some other technique for use by analysts, which will give results of reasonable concordance. Discussions with the Imperial Institute and the Scott Laboratory at Nairobi have taken place and Mr. Lord is again cooperating in this difficult task. Preliminary work has already been carried out to ascertain the effect of variations in the technique employed. It is clear that the details will have to be laid down with great particularity if the past recurrent disputes with their resultant losses are to be avoided. During the year we had a conference with the Chairman of the Kenya Pyrethrum Board and with two representatives of the Imperial Institute about to visit the U.S.A. to discuss this matter.

Ryania speciosa

Samples of this plant were received some years ago from Trinidad and not found to have outstanding insecticidal properties. During 1945 fresh samples of root and stem were received *via* the Imperial Institute, Mr. Lord examined them and found that the toxicity of the plant to insects was not of a very high order and was specific in its effects; extracts, however, have a fairly quick knock-down from which insects tend to recover. The plant is worth further examination since it is reported to have specific effects and that the active principle is not easy to extract.

Analogues of D.D.T.

Further work was carried out by Mr. Lord in order to compare the toxicities of D.D.T. analogues. Tests with alkyloxy derivatives showed the ethoxy compound to be the most toxic; beyond this

toxicity decreased with increase in molecular weight, moreover, the slope of the probit line tended to become less steep. In this work the structure and purity of the various analogues were checked by degradation to the corresponding benzophenones and by estimating the amount of the appropriate halogen acid liberated by the action of caustic soda. Tests were made to ascertain the compatibility of D.D.T. with nicotine; the decomposition observed was studied and found to be catalysed by ferrous and ferric ions and by copper and aluminium ions. Similar tests were carried out with certain alkylamines. A start was made during the year on the effects of these compounds on the respiration of insects, thus obtaining a measure of their metabolic action. Microrespirometers of a modified Barcroft pattern were used, an initial strong stimulus was observed which declined as the series ascended from chlorine upwards. In order of speed of reaction they could be arranged: $F > Cl > Br > I$. Mr. Lord has also carried out some interesting experiments on the sorption of D.D.T. by chitin; it was found to be rapid, but so far no success has been achieved in measuring the rate of penetration.

Gammexane

Using *Oryzaephilus surinamensis* as test subject Mr. Lord compared the insecticidal values of the α , β , γ , and δ isomers of hexahydro-hexachlor-benzene. The γ -isomer was of the same order of toxicity as D.D.T., whereas the α and δ isomers were much less active. The β -derivative was too insoluble to allow of an estimate being made.

The effect of particle size and shape on the insecticidal properties of D.D.T.

Mr. McIntosh, after completing and testing out his dipping apparatus for determining insecticidal potency, has submitted an account of it for publication. Using this technique he has since carried out a long series of experiments in which he has compared the insecticidal values of various suspensions of D.D.T. in which both the particle size and shape were varied. This has involved much work in preparing D.D.T. suspensions of definite types, ranging from the colloidal to relatively large plates and long needles. Such preparations had to be replicable as several repetitions of the toxicity trials had to be made before confidence in the rather unlooked-for results could be felt. Using *Tribolium castaneum*, a relatively robust and easily handled insect test-subject, he showed that within the range of sizes tested toxicity varied directly with average particle size determined by microscopic measurement, but that the shape of the crystals might have a bearing on the results. Thus the order of potency could be expressed as follows: colloid < short plates < large plates < long needles. That the effect was not due to the technique employed was shown by some determinations in which the spraying tower was used and the results shown to hold, when the crystals were small enough to pass through the nozzle intact. The results found an explanation when analyses of the insects for D.D.T., retained on them after dipping, were carried out. This required a very large number of test subjects and the employment of a micro-analytical method. Using a colorimetric technique worked out by Schechter,

et al., Mr. McIntosh obtained evidence that when his insects were dipped in a colloidal preparation of D.D.T., less of the chemical was retained by the insects than when the long needles were used in the dipping fluid. The difference in amount retained was almost great enough to give a quantitative explanation of the above-mentioned toxicity difference. This finding is of considerable importance and McIntosh proposes to carry the work further with other crystalline insecticides.

BIOLOGICAL WORK

Work has been continued on the rearing of suitable plant insects as test-subjects. These have been needed in large numbers at all times of the year and some ingenuity has been required in securing them. Mrs. Gillham has been mainly concerned with the aphides, *Macrosiphoniella sanborni* and *Macrosiphum solanifolii* and others, while Miss Stoker has had charge of the mandibulate insects, such as the Mustard beetle (*Phaedon cochleariae*) and Diamond back moth (*Plutella maculipennis*) and others. Both have rendered great help in the carrying out of spray trials and in the examination of the results.

Daylight lamps have been in use for rearing purposes and after much delay the new batteries of lamps for comparing the effect on insect resistance, reproductive rate and plant growth due to illuminations of different quality, have been installed and a start made in their use.

Mrs. Gillham has continued the work in which she collaborated with Dr. Potter on the variation in resistance of aphides. The toxicity of various insecticides to aphid test-subjects has been studied over a period of years but a detailed analysis of the collected data has not yet been made.

In general there is a range of variation in resistance, but it seems difficult to correlate this with any particular set of conditions. There are some exceptions showing a greater degree of variation from the rest of the series but it is not known whether any direct environmental effect is involved.

Preliminary experiments with *Macrosiphum pisi* indicate that with this species resistance is influenced by the host plant, insects from clover appeared more resistant to rotenone than those from broad bean or pea.

A laboratory technique for determining the toxicity of stomach poisons

Mr. Way has been engaged for some time in elaborating methods for the valuation of these classes of insecticides. The first stage of the work has been carried to completion and an account is being prepared for publication. Many complexities have been encountered, some inherent in the type of insect used as test-subject or in the chemical compound. Some are of an operational nature. Thus, certain caterpillars fail to feed if confined in too close a space, and some compounds prove repellent or deterrent; in compounds of very considerable toxicity accurate measurement of the very fine deposits required is difficult.

Essentially, the technique should be capable of putting down on foliage very small but evenly distributed deposits, it should afford means of measuring the amount per unit area, and secure the administration of pre-determined doses to individual insects. The

latter entails keeping the foliage fresh and edible during and after the period of consumption. The assessment of the effects and the statistical analyses of the results follow.

The spraying apparatus consists of a tall cylinder of thin zinc sheet (5 ft. 6 in. tall and 1 ft. 3 in. diam.) which is earthed to prevent charging. The tower is held in a vertical position by adjustable wire braces and rests on a flange surrounding a circular aperture in the top of a stout table. Four tubular vents near the bottom allow for the escape of air displaced by the actual spraying operation, and the top tapers to a duct of 6 in. diam. which leads to the exterior.

Underneath the tower there is a circular spray platform in the table of the same diameter as the tower, with a plate-glass cover constituting a false bottom. Through a hole in the centre of both projects the nozzle of an atomiser. Both spray platform and glass plate can be independently withdrawn in separate slots. Foliage and a weighed sheet of aluminium foil of known area rest on the platform. With the false bottom in position the insecticide is sprayed into the tower and after allowing the larger droplets to fall on the false bottom the latter is removed to permit the mist of insecticide to settle on foliage and foil. By standardising the spraying procedure deposits can be predetermined to within 5 per cent. and the exact deposit per unit area determined by weighing the sheet of aluminium foil.

Known amounts of the sprayed foliage are then fed to insects by appropriate means which have to be adapted to the insect used and the insecticidal purpose under study; for example, if the toxicity to be determined is to be strictly limited to stomach action, then the food has to be administered so that only the mouth parts, at most, come into external contact with the poison. This can be secured by enclosing the insect in a confined channel across the top of which the strip of poisoned leaf is placed; but, as all caterpillars are not amenable to this treatment, a special feeding device is used in such cases; a strip of treated foliage of known area extruding from a small cork clamp, and with the treated surface protected by a celluloid cover, is enclosed with the insect in a small cage. If both external contact and internal effects combined are the subject of study, circles of the poisoned foliage can be enclosed with the insect upon it. The problem of preserving the foliage free from dessication has been a serious one, but devices for maintaining fresh foliage have been designed, and constant temperature and humidity chambers are used for after-treatment storage.

The technique for treating large insect species is highly satisfactory and has been used to determine the effect of insect body-weight on susceptibility to various insecticides. Detailed experiments were carried out using last instar larvae of *Diataraxia oleracea* and *Phlogophora meticulosa*. Results show that D.D.T. and γ -benzene hexachloride are highly toxic but that mature last instar larvae have a much greater resistance than young larvae of the same instar. This difference in resistance with size of larvae is not so noticeable when Lead Arsenate is used as the poison. The size factor in the test subject is shown to be one of great importance and this problem is being made the subject of further study.

The analysis of the results is complicated, the dosage per body weight of the insect has to be determined, entailing weighing large

numbers of individual insects. The Bliss and Gaddum probit technique is applicable to the statistical analysis of results but the probit plane conception of Finney has to be employed and this adds considerably to the length and complexity of the computations.

The effect of D.D.T. and benzene hexachloride on bees*

This work was undertaken at the instance of a sub-committee of an A.R.C. Conference on insecticides. A joint investigation between this department (M. J. Way) and the Bee department (Miss A. Synge) has involved a considerable amount of work, which was accentuated by the bad weather conditions ruling during the summer and early autumn of 1946. The results obtained were laid before a conference of advisory entomologists in December, 1946, and have been embodied in a report to the A.R.C. Although not yet in a final form the results can be summarised thus:

Laboratory tests under rather drastic conditions showed B.H.C. to be highly toxic to *A. mellifera*. Used as a contact insecticide, with the γ -isomer content well below field concentration, commercial preparations gave 100 per cent. kill. Field concentrations of D.D.T. proved only partially toxic. Tested in the laboratory as *stomach poisons*, using specially prepared suspensions, it was shown that D.D.T. in colloidal form is about four times as toxic as a crystalline preparation. Lead Arsenate *under these conditions* is somewhat less potent than D.D.T. and slow in its effects, but B.H.C. by comparison is very toxic.

Laboratory experiments by contact with D.D.T. and γ -B.H.C. on various bumblebee species showed that workers are similar in their susceptibility to workers of *A. mellifera*. Queens and Drones are more resistant, D.D.T. at high concentrations and under drastic conditions having little effect. This is a matter of some importance since in the spring *Bombus* queens are foraging and their loss means the potential loss of colonies of workers.

Field experiments showed that both *A. mellifera* and *Bombus* workers visiting open blossoms, treated with commercial D.D.T. sprays and dusts, are not appreciably affected. This finding, however, should not be interpreted as justifying the treatment of open blossom with D.D.T., since there is the risk that an activated form of this compound might have serious effects. Preliminary work, which owing to weather conditions and experimental difficulties gave only tentative results, did not indicate D.D.T. to be a serious danger to honeybee larvae. It would thus appear that, used with reasonable care, D.D.T. is safe to bees.

Field experiments with B.H.C. when applied to open blossom showed this insecticide to be highly lethal to bees and to be harmful to them after a period of at least four days. In view of this it presents a danger to visiting bees by chance contacts, even if sprayed before blossom opens. There was no repellency noticeable, and as speed of toxic action is not sufficiently quick to prevent contaminated workers returning to the hive it is considered that the use of B.H.C. might also be dangerous to nurse bees and larvae through contact with contaminated food brought to the hive by the workers.

* The letters B.H.C. are subsequently used for this material.

Toxicity of D.D.T. and B.H.C. to several parasite and predator insects (M. J. Way)

Foliage was treated with certain commercial preparations of D.D.T. and B.H.C. enclosed in cages with certain parasite or predator insects. Under these conditions it was shown that adults of several hymenopterous aphid parasites and adults of several Syrphid species are very highly susceptible to B.H.C. and are also killed by D.D.T. which, however, is not as toxic as γ -B.H.C. Syrphid larvae are not affected even after drastic treatment with D.D.T. but young larvae are killed by spraying and dusting with field concentrations of B.H.C. Fairly high concentrations of B.H.C. are required to kill mature syrphid larvae. Syrphid larvae are not affected by field concentrations of Derris spray and Nicotine dust and spray.

Batches of Coccinellid eggs were directly treated with certain commercial sprays and dusts of D.D.T., B.H.C., Derris and Nicotine. Results show that in these preparations D.D.T. and B.H.C. have apparently no ovicidal action but larvae are destroyed by B.H.C. after emergence. D.D.T. appears to cause little harm to emerging larvae. Derris has a powerful ovicidal action even at low concentrations while Nicotine spray shows no harmful effects.

Effect of D.D.T. and B.H.C. on vegetables and fruit crops (Miss R. I. Stoker)

The phytotoxicity of these insecticides has been studied and the possible tainting effect of B.H.C. examined. The decline in yield of tomato after treatment with D.D.T. has not been confirmed, nor have the distorting effects on cucumbers been repeated.

Small scale field experiments with several commercial preparations of B.H.C. were carried out on the following seedling vegetables: radishes, turnips, swedes, spinach, beetroot, peas, carrots, onions. The results of preliminary trials were:—

"Gammexane" Dispersible Powder	0.2%	isomer—	radishes, turnips, swedes, spinach, beetroot; scorched
"	"	"	—turnips and radishes slightly scorched
" General Purpose Dust	0.5%	"	—slight evanescent retarding on radishes
" Flea Beetle Dust	0.2%	"	—only applied to turnips and swedes with no marked effect

A field experiment on kale seedlings showed that sprays (Dispersible Powder) containing concentrations of less than 0.1 per cent. of the γ -isomer of B.H.C. could be used with safety.

It can be concluded from all these experiments that, following application of commercial sprays containing above 0.1 per cent. γ -isomer, brassica seedlings, beetroot and spinach are liable to damage and that there is some risk of retardation of growth and possibly germination by the use of both the General Purposes and the Flea Beetle B.H.C. dusts on brassicas.

Comparison of the effect of B.H.C. and D.D.T. on tomatoes (variety, Harbinger)

A carefully devised pot experiment in the open, in which was used Guesarol E spray (0.2 per cent. D.D.T.), B.H.C. Dispersible Powder at 0.01 and Liquid Agrocide at 0.015, 0.0075, 0.0037 per

cent. γ -isomer showed that these preparations had no direct toxic effects.

The effect of B.H.C. on the flavour of vegetable crops

Small rows of various vegetables were sprayed with a Mysto sprayer in such a way as to prevent much spray reaching the soil. The following are the results, concentrations being in terms of the γ -isomer:—

Peas.—Sprayed in flower (Dispersible Powder). Tainting at 0.1 and 0.05% particularly after cooking.

Carrot.—Young plants dusted (General Purpose Dust) and sprayed (Dispersible Powder). Tainting to young and fully matured carrots with 0.2% and 0.1% spray and 0.5% dust. Lower concentrations gave no taint.

Beetroot.—As for peas. 0.1% Dispersible Powder caused loss of flavour, lower concentrations did not.

Onion.—As for peas. No taint.

Marrow.—Liquid Agrocide 0.15–0.05%
Dispersible Powder 0.1–0.01% } All treatments imparted an earthy flavour.

Cauliflower.—Dispersible Powder 0.01–0.05%. Earthy flavour.

Lettuce.—As for peas. 0.1% caused a bitter taste. No taint at lower concentration.

Radishes.—Dispersible Powder 0.2 and 0.04%. No taint.

The insecticidal properties of film deposits of D.D.T. and B.H.C.

Dr. Pradhan, a Government of India State Scholar, has carried out investigations on these problems since his arrival in February, 1946. Since the potencies of D.D.T. and B.H.C. are relatively long-lasting, the effects of films laid down are likely to prove of practical importance. Dr. Pradhan has been engaged in developing appropriate laboratory techniques for studying them. Insect test subjects selected have been *Tribolium castaneum*, *Plutella maculipennis*, *Macrosiphoniella sanborni*, the first mentioned insect is being used as it is far easier to confine it on a film than most plant-feeding insects. Some difficulty has been experienced in the case of B.H.C. in separating the fumigant from the direct contact action, but this has been partially achieved.

Effect of type of surface on toxicity

Preliminary experiments have been carried out to test the effects of the following: wax, filter paper, bolting silk, leaves of water lily, marrow, cabbage and geum in order to cover a relatively wide range. It is clear that the effectiveness of D.D.T. will depend on the surface upon which it is deposited. The effect of temperature upon the toxicity and speed of reaction of D.D.T. films is being studied.

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