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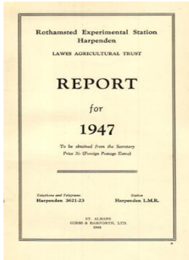
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Insecticides and Fungicides Department

C. Potter

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DEPARTMENT OF INSECTICIDES AND FUNGICIDES

By C. Potter

Dr. Tattersfield retired from the headship of the department in June, 1947, but fortunately was able to continue working in the department on a part-time basis. Dr. Tattersfield's outstanding contributions to the field of research on insecticides and fungicides were recognised by the award of the O.B.E. in the birthday honours list.

Dr. Potter, who had spent the previous year in the United States of America as a Visiting Research Professor of Entomology at the invitation of the Rhode Island State College and the Connecticut Agricultural Experiment Station, spent the first two and a half months of 1947 in the U.S.A., touring research stations on behalf of the Agricultural Research Council. He returned to the department at the end of March and was appointed head of the department at the beginning of June.

For convenience the work of the department may be set out under five headings:—

(1) General and chemical
(2) Chemical
(3) Physico-chemical
(4) Biochemical
(5) Biological

(1) General and chemical

Some of the newer organic insecticides have been examined. These include Velsicol 1068, Toxaphene and various samples of Hexaethyl tetraphosphate, which were primarily tested to investigate their direct contact effect. The samples of Hexaethyl tetraphosphate were examined at the request of the Agricultural Research Council and were worked on more extensively than the other two chemicals which gave rather disappointing results in preliminary trials.

Hexaethyl tetraphosphate has shown a high toxicity to the insect species under test and a small scale preliminary field test indicated that it was toxic to the active stages of the greenhouse red spider although it had little or no effect on the eggs of the mite. However, its very high toxicity to man is a factor militating against the use of this insecticide.

Some further work on the chemical and insecticidal properties of *Ryania speciosa* has been carried out. Extracts of this plant material have not shown any marked toxicity to the species of insect used, but as there is reason to believe that the toxic agents are rather specific in their effect the work is being continued. Mr. Lord has been responsible for a high proportion of this work.

Mr. Way made some preliminary trials on the stomach poison effect on the larvae of the tomato moth *Diataraxia oleracea* L. of four insecticides. He showed that the larvae of this species at the stage selected were completely unaffected by doses of Rotenone as high as 0·13 mg. per larva, thiodiphenylamine at 0·12 mg. per larva, the Triethanolamine salt of 3:5 dinitro-o-cresol at 0·044 mg. per larva and zinc fluoarsenate at 0·31 mg. per larva.

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As a result of work on the factors affecting the resistance of insects to insecticides, some of which is described later, it seems that the lepidopterous larvae used (*Diataraxia oleracea* L. and *Phlogophora meticulosa* L.) the stomach poison effect of DDT $>$ γ –BHC $>$ lead arsenate, and DDT is more effective than the gamma isomer of benzene hexachloride as a contact poison to these larvae.

Field experiments, statistically planned. A field experiment was carried out by Mr. M. J. Way to compare the efficiency of DDT and Benzene hexachloride for the control of pests of kale.

Two main treatments were carried out : (1) Drilling insecticidal dust with the seed and (2) Spraying and dusting kale seedlings after emergence. A study was made of seed germination, the emergence of seedlings and the growth of young plants.

No flea beetle infestation occurred and pest infestation generally was negligible in the two months following treatment. An analysis of the results showed that the insecticide treatments had conferred little benefit on the crop under these conditions.

Plots in which seeds were drilled with benzene hexachloride dust showed a slightly significantly higher proportion of emergence of seedlings than the DDT and control plots.

A further field experiment on the control of wireworms in Little Hoos field was started by Dr. Evans of the Entomology Department and Dr. Potter. The effect of DDT, Benzene hexachloride, Ethylene dibromide and the soil fumigant D.D. are being studied. The chemicals have been applied but the sampling has yet to be done.

(2) CHEMICAL

Collaborative work on the method of estimation of the pyrethrins has been continued and a number of slight variations in the mercury reduction method have been tested. Among the methods tried the use of n-hexane as a solvent instead of petroleum ether had been investigated and found to be satisfactory. The work is expected to continue with the department participating in a world collaborative scheme to compare the various methods and their modifications in use at the present time.

Mr. Lord has continued his study of DDT and 23 of its analogues and the alpha, beta, gamma and delta isomers of benzene hexachloride. Data on the toxicity of these compounds to the black chrysanthemum aphid *Macrosiphoniella sanborni* Gill. and *Oryzaephilus surinamensis* L. were obtained.

It was shown that the probit lines for the DDT analogues varied both in position and slope, those for the isomers of benzene hexachloride varied in position but not in slope. In general, neither the relative positions nor the relative slopes of the probit lines for the various substances were the same with the two species of insect.

The toxicity of the substances analogous to DDT appears to be related to molecular weight with a maximum occurring in the molecular weight range 300-450. Among the DDT analogues the slope of the probit line is apparently correlated with molecular volume as given by the parachor. It has been inferred that the variations in the slopes of the probit lines arises from differences in the interactions of the poisons with the test subject. It is also suggested that the evidence indicates that the action of DDT and benzene hexachloride have a reversible physico-chemical action rather than an irreversible chemical action.

(3) Physico-chemical work

Effect of particle size on the toxicity of suspensions of insecticide applied to the insect. Mr. McIntosh's previous work had shown that if the adult *Tribolium castaneum* Hbst. (flour beetle) is used as a test subject and dipping or spraying is used as the method of application, the toxicity of suspensions of DDT increases with increase of the crystal size of the DDT up to about 400μ. Crystals larger than this have not been tested. It has been found that the increase in toxicity is paralleled by an increased retention of the poison on the body of the insect. It is not suggested that this is the whole or even the major cause of the increase in toxicity.

Further tests were carried out using apterous viviparous females of the aphid *Macrosiphoniella sanborni* Gill. The results were unsatisfactory because considerable variation in both the absolute and relative toxicities was encountered. However, the evidence acquired indicated that a crystal size of $60 \times 15\mu$ was the least effective.

Rotenone. Using the dipping method and the adult saw-toothed grain beetle *Oryzaephilus surinamensis* L. as a test subject it was found that a decrease in the particle size resulted in an increase of toxicity and the magnitude of the particle size effect on toxicity far exceeds that obtained with DDT, over what is, if anything, a smaller range of sizes. A difference of 400 has been obtained between equal concentrations (w/v) of two suspensions of particles of two different sizes.

The results obtained to date indicate that crystal shape as apart from size have no demonstrable effect on toxicity of rotenone.

This work is being continued particularly with a view to explaining the results so far obtained.

By employing the methods he has developed here for preparing DDT suspensions of uniform crystal size Mr. McIntosh has prepared such crystals and mixed them with dust diluents, thus giving a series of dusts containing the toxic agent in a known and uniform shape and size. A number of these dust mixtures containing DDT crystals of various sizes in two diluents have been prepared for Dr. W. A. L. David of the Agricultural Research Council's Unit of Insect Physiology, Cambridge.

Sorption of DDT by chitin. Mr Lord has continued his studies on the sorption of DDT and its analogues by chitin. It has been shown that the amount sorbed is dependent on the surface area of the chitin and that sorption by the cuticle may be peculiar to the chitin since cellulose wool and silica powder do not possess this sorptive capacity for DDT and its analogues.

The rate of sorption and the amount sorbed is approximately the same for DDT as for its analogues.

(4) Biochemical work

Mr. Lord has been continuing his work on the effect of insecticides on insect respiration using respirometers of a modified Barcroft pattern. The results of the initial experiments on the effect of insecticides on metabolism and death rate have been examined. The inferences drawn from the data are as follows. Toxic concentrations of DDT and its analogues applied as dusts to adult saw-toothed grain beetle *Oryzaephilus surinamensis* L. increase the rate

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of oxygen uptake. Sub-lethal concentrations have no effect. With groups of starving insects the total oxygen uptake from the time of exposure till death is the same whether or not the insect is treated with DDT or its analogues, insects treated with a lethal dose of insecticide respire more rapidly and therefore die sooner than untreated insects. The stimulus of DDT to this species is apparently quantal in nature and the magnitude is independent of the concentration of DDT in the dust applied. The stimuli resulting from the action of DDT and its analogues are approximately equal, as are the rates at which the insects die.

The effect of Benzene hexachloride on adult *Oryzaephilus surinamensis* L. has been shown to be similar to that of DDT.

The qualitative effects of some other insecticides have been observed and it has been found that Rotenone and Lethane 371 depress the oxygen consumption while 3:5 dinitro-o-cresol causes the death of the insect far more rapidly than any other poison investigated.

This work is being continued. Modifications of the apparatus for the application of the poison *in situ* are being studied.

During the course of his work on the relationship between the weight of the insect and its susceptibility, Mr. Way carried out some work on the changes that occurred with larvae of tomato moth *Diataraxia oleracea* L. poisoned with DDT. Detailed studies were made of the total loss in weight, of the change in moisture content, and the change in dry weight during DDT poisoning. Over 60 per cent. loss of water occurred between the time of poisoning and death. The dry weight decreased by 3–4 per cent. and of this only 0·5 per cent. was loss in fat content.

Thus although 4–5 days may elapse before death occurs and during this period the larva is continuously active, fat does not appear to be utilised as a source of energy. Experiments showed that an untreated but starved larva utilised its stored fats.

(5) Biological

Factors affecting the susceptibility of insects to insecticides

(a) Effect of larval body weight on resistance.

During the latter part of 1946 and early 1947 Mr. M. J. Way made a detailed study of the relationship between larval body weight and the resistance of the larvae to various insecticides.

Preliminary studies with the last instar larvae of the tomato moth *Diataraxia oleracea* L. showed that as the larvae increased in weight so its resistance to stomach poisons increased. The extent of the increase varied with the insecticide used.

With lead arsenate, using larvae of weights from 0·15 gm. to 0·55 gm., there was a constant relationship between increase of weight and increase in resistance if the weight increase was x 2 the Median lethal dose had to be multiplied by 1·3.

With DDT the problem was not so simple and there is not a straight line relationship between increase in body weight and increase in resistance. There is a greater increase in resistance per unit increase in body weight at the higher larval weights than at the lower larval weights, e.g., if the larval weight is increased from 0·15 gm. to 0·30 gm. the median lethal dose increases from 0·00075 mg. per larva to 0·0030 mg. per larva, an increase of

0·015 mg. per gram; if, however, the larval weight increases from 0·45–0·55 gm. the m.l.d. increases from 0·0175–0·0440 mg. per larva, an increase of 0·265 mg. per gram.

The gamma isomer of benzene hexachloride showed a similar relationship to DDT but the changes were not so strongly marked.

These results were obtained using a technique for studying stomach poison action in which contact action was eliminated as far as possible.

A study was then made along the same lines on the effect of body weight on contact toxicity using DDT and the gamma isomer of benzene hexachloride. The dose per larva was determined by washing the insecticide from the larvae with benzene after spraying and estimating the amounts colorimetrically.

A similar relationship between body weight and resistance was found when the insecticides were applied as contact poisons as had been found when they were applied as stomach poisons although there were some differences in degree.

An interesting aspect of this comparison was that DDT proved to be more toxic as a contact poison than as a stomach poison if the amount retained on the body (producing a given percentage kill) is compared with the amount required to be ingested to produce the same kill. This can only be said to be true of this particular set of experiments.

This study with lepidopterous larvae on the relationship between body weight and resistance has proved very important in assessing the validity and significance of estimations and comparisons of toxicity. It seems probable that for stomach poisons at least, where it is only possible to use relatively small numbers of insects, estimates must be made by measuring the three variables, dose, body weight and mortality and the results analysed by the probit plane technique. It is hoped to continue this work later on.

Another item of interest brought out in the course of this work was that with the last instar larvae of tomato moth there was a sudden fall in resistance to the gamma isomer of benzene hexachloride just prior to pupation.

(b) Cuticular structure and the penetration of insecticides.

Toward the end of April, 1947, Mr. Way was seconded to the Entomology Department, Cambridge, under the direction of Dr. V. B. Wigglesworth where he commenced to work on the penetration of insecticides through the cuticle using larvae of the tomato moth as test subjects. This work is still in progress. Developmental changes in the structure of the cuticle have been studied and, as with other insects, complex structure is apparent. The cuticle consists in general of five layers of different composition which are all already present at the beginning of the last instar but which increase in thickness and change in composition during the development of the instar. In certain areas of the body wall the cuticle is specialised and may be thin with several of the layers missing. The cuticle overlying sense organs and certain areas of the prolegs and muscle insertions are examples of these areas. It would appear that the penetration of poison could occur most readily in these areas.

The difficulty of developing a technique whereby droplets of insecticide can be placed and confined in particular areas of cuticle resulted in the failure of many attempts to determine the rate of penetration in particular areas of the cuticle, all that can be said is that with the insect under test the penetration of DDT in certain parts of the body is very slow or does not occur at all. The head and tail regions seem more sensitive than the middle regions of the body.

(c) The effect of environment on toxicity.

Mr. McIntosh found in the course of his experiments that rotenone gave a higher kill of adult *Oryzaephilus surinamensis* L. when the insects were kept after treatment at 32° C. than at 20° C., while the reverse effect was found when DDT was used.

Dr. Pradhan has been studying the effect of changes in the environment on the toxicity of residual films of insecticide.

A study was first made of the relationship between the concentration of insecticide, the time of exposure and the mortality produced.

Various techniques were worked out to ensure continuous contact between the insect and the insecticide film.

At first when adult flour beetle *Tribolium castaneum* Hbst., which does not climb on glass surfaces were used as test subjects and DDT as insecticide, films were made on filter papers and the insects confined on the films by means of inverted whole filter funnels. When the gamma isomer of benzene hexachloride (γ–BHC) was used as insecticide there was evidence of a fumigant effect with this technique and the stems and part of the cone of the funnel was cut off so that free aeration could occur, rings of glass cut from glass tubes 4–5 cms. diam. were also used. When the larvae of the diamond back moth *Plutella maculipennis* Curt. were used as a test subjects and DDT as insecticide equal films were formed on two filter papers; then one of the papers was folded into a cone by a standard technique with the film on the inner surface of the cone, the cone was then inverted over the unfolded filter paper to form an enclosure, all surfaces of which were coated with insecticide, the insects were confined in this enclosure. The cone was kept in position by a circular iron ring. When γ–BHC was substituted for the DDT, at first the cone of filter paper was perforated to allow for aeration but this proved unsatisfactory and finally a similar arrangement was adopted to that used with DDT but bolting silk was used throughout instead of filter paper.

The variation in mortality with changes in concentration and exposure time was studied using adult *Tribolium castaneum* Hbst., larval *Plutella maculipennis* Curt. and nymphs of the black chrysanthemum aphid *Macrosiphoniella sanborni* Gill. DDT was first used and then γ–BHC. The adult *T. castaneum* proved the best test subject.

It was found that at a given temperature (80° F.), if the insects were given a short exposure time (24 hrs.) and examined immediately zero mortality was obtained at all the concentrations tested, thus producing a straight line curve parallel to the Y axis at zero mortality. At the other extreme with long exposures (36 days) 100 per cent. mortality was obtained at all the concentrations tested including controls, giving a straight line curve parallel to the

Y axis at 100 per cent. mortality. Intermediate exposures (all followed by immediate examination) gave curves of various shapes, for the most part sigmoid in character. This data also shows that as the strength of the film increases the average survival period on the film goes on gradually decreasing.

Some preliminary studies of the effect on toxicity of the surface on which the insecticidal film is formed were also made using both DDT and γ–BHC. The surfaces tested were glass, paraffin wax, filter paper, bolting silk, geum leaf, marrow leaf, cabbage leaf and leaves of water lily. It was found that films on waxy surfaces such as paraffin wax and leaves of water lily and cabbage were relatively non-toxic compared with glass and leaves of geum. An extensive series of experiments were then carried out to determine the effect of temperature on mortality of insects exposed to films of insecticide using adult *Tribolium castaneum* Hbst. and larval *Plutella maculipennis* Curt. From the results of these experiments together with a survey of the literature the following generalisations were inferred:

(a) That insect resistance to poisons changes with temperature as do its other vital activities, i.e., increasing up to a critical point and then decreasing.

(b) With insecticidal films the amount of poison reaching the site of action in unit time also varies with the temperature, generally, but not always, increasing with increased activity of the insect. The locomotor activity of the insect appears to play an important part in the pick-up of the insecticide. An instance where increased activity appears to result in decreased pick-up occurred with larvae of *Plutella maculipennis* Curt. where the web spun by the larvae as they moved formed a protective layer on the surface of the film.

(c) The overall effect of temperature on the insecticidal action of films is the result of the interaction between the two preceding factors, resistance and pick-up.

Finally some investigations were made on the effect of changes in relative humidity on the mortality of insects exposed to films of insecticide. In this series adult *Tribolium castaneum* Hbst. and the larvae of *Plutella maculipennis* Curt. were again used as test subjects. The two insecticides studied were DDT and 3:5-dinitro-o-cresol (DNOC).

With the adult *T. castaneum* the toxicity of both DDT and DNOC increases with increased relative humidity. This is true both during the course of exposure and after treatment.

However, when the larvae of *Plutella maculipennis* Curt. are exposed to the film for 24 hours and then inspected at once and the mortality assessed, it was found that increased relative humidity resulted in a decreased mortality with DDT films but increased mortality with films of DNOC.

No general principles could be deduced from the latter experiments and a survey of the literature. However, it seems likely that factors that must be taken into consideration besides that of the species and instar of insect and the nature of the insecticide, are the concentration of the insecticides used, the effect of humidity being discernible at some concentrations and not at others, and the range of humidity; differences may not show unless a wide range of humidities are tested.

Partly owing to the absence of Dr. Potter and partly owing to pressure of other work, only a little data was acquired on long term fluctuation of resistance, this work was done by Mrs. Gillham. Studies on the effect of environment on insect resistance apart from those carried out by Dr. Pradhan were also held up partly for the reasons given above but chiefly owing to lack of facilities for producing a controlled environment.

Insect rearing. In connection with this proposed work on rearing insects in an environment where temperature, humidity and light are controlled, Miss R. I. Stoker carried out some preliminary experiments on the effect of these conditions on plant growth. Three different illumination units were installed in a constant temperature room and controlled by means of time switches to give 14 hours light and 10 hours darkness. Unit (1) consisted of four 80 watt daylight fluorescent tubes. Unit (2) consisted of two 80 watt daylight fluorescent tubes and one 400 watt neon tube. Unit (3) consisted of two 400 watt neon tubes. The temperature of the room was approximately 27° C., the humidity was not strictly controlled.

Turnips were used as test plants. No difference was observed in germination rate but from then on there was a steady improvement in the growth and condition of the seedlings under the neon lights as compared with the fluorescent lights. The most noticeable difference was in leaf colour and size, the neon lights producing larger leaves of a darker green colour. The plants grown under the combined lights (Unit 2) gave plants rather less vigorous than the neon lights but similar in colour and texture. There were temperature differences between the units due to the large amount of heat produced by the neon lights, but this is not thought to account for the observed differences.

Many difficulties were encountered in this work owing to lack of facilities for temperature and humidity control and to frequent failures of the electricity supply but it is very much hoped that we shall be enabled to remedy these deficiencies and carry on with the work.

A considerable amount of work was carried out on the rearing of insects. The species now in regular use as test subjects were studied in order to improve the rearing techniques and additional species which might prove useful were investigated.

The ordered progress of this work also was handicapped by lack of facilities for illumination and controlled environment.

Phaedon cochleariae *F.* (*Mustard beetle*). Miss R. I. Stoker has made a detailed study of the laboratory rearing of mustard beetle. Besides being the only beetle of agricultural importance which has so far been found suitable for laboratory rearing, it has many good qualities as an experimental animal, among which are convenient size, ease of handling, absence of disease, and a host plant that can be made available throughout the year.

Detailed studies have been made on the fecundity of the adults, the viability of the eggs and the development of the larvae, pupae and adults under various conditions of lighting in the constant temperature room. It has been found that while the insect cannot be reared throughout the year in the glasshouse with natural illumination it is possible to do this in the constant temperature room with illumination from either neon or fluorescent lights. The evidence to date suggests that the diapause does not occur with insects reared continuously at 75° F. with illumination as previously described for 14 hours per day.

The major difficulty at present is the great variation in the fecundity of the adults. Experiments have been made to determine the factors affecting egg-laying, so far with no positive results. The work is being continued.

Myzus persicae *Sulz.* Mrs. E. M. Gillham has carried out some work to compare the rates of reproduction of this aphid under three different lighting conditions at approximately constant temperature with that occurring under greenhouse conditions where there were great variations in temperature and illumination. Considerable difficulties were again encountered due to deficiencies in the apparatus and failure of the electricity supply. In addition, the insects were difficult to handle. The general indications were that over the period during which data were collected there was no significant difference between the number of young produced under the various conditions although it was usually slightly lower under glasshouse conditions. This work is only preliminary and it is hoped to undertake more detailed work if the necessary staff is available. Stocks are kept going all the year round on cabbage and turnip.

Macrosiphoniella sanborni *Gill.* (*Black chrysanthemum aphid*). This aphid can now be reared under glasshouse conditions for most of the year but there are periods in midsummer and midwinter when it is difficult to rear in numbers. Facilities for added illumination might overcome this difficulty. It is our most useful test aphid.

Macrosiphum solanifolii *Ash.* (*Green potato aphid*). This is a most valuable aphid for test purposes because it is easily handled, it lives several days with food and is, in general, resistant.

So far, however, we have found it difficult to rear continuously in numbers in spite of its wide range of host plants. It is rather susceptible to fungus disease.

Work on this species is being continued.

Acrythosiphon onabrychia [Macrosiphum pisi] (*Pea aphid*). This insect is being reared because some evidence was obtained that its resistance varied with the host plant on which it fed. However, we do not know enough about its biology to rear it successfully. It was found in one season that cultures on broad bean remained viviparous throughout the winter in the cool glasshouse while in another season when it was reared on clover it produced oviparous females in October and remained in the egg stage throughout the winter. It is hoped to find a suitable technique for rearing this insect.

Aphis fabae *Scop.* (*Bean aphid*). Considerable difficulties are now recognised to exist in the identification of this insect. At present we rear a species obtained from nasturtiums which is continuously cultured on that host. It appears to be a distinct species, as yet unnamed, and attempts to establish it on broad bean have so far been unsuccessful although it will colonise dock.

Cultures can be maintained on nasturtiums throughout the year but during the winter months reproduction is very slow under glasshouse conditions, and again facilities for a controlled environment, including illumination, would be a great help. The insect is a very useful general test subject.

Myzus circumflexus *Buct.* This insect can be reared throughout the year on Arum lily (Richardia) and has proved a useful supplementary test subject.

Plutella maculipennis *Curt.* (*Diamond back moth*). This insect is reared all the year round in large numbers on cabbage and turnip. There is a complete absence of seasonal rhythm. The technique of rearing has been fully studied and this is probably the most easily reared in numbers of all the mandibulate species. It is used as a test subject for most phases of our work.

Diataraxia (Polia) oleracea *L.* (*Tomato moth*). This insect is used a great deal for stomach poison studies and other work. It is reared in large numbers on dock, cabbage and turnip. It normally has a diapause but this may be broken by (a) Rearing for the latter part of larval life under fluorescent lighting for 15 hours per diem, or (b) Keeping the pupae in a refrigerator for a minimum period of two months. So far 11–12 generations have been reared from the same stock at the rate of about 6 generations a year without serious trouble.

Xanthorhoe fluctuata *L.* (*Garden carpet moth*). This moth can be reared in large numbers on cabbage and turnip throughout the year. The normal diapause can be broken by refrigerating the pupae for a minimum of two months. It has not so far been widely used because it is not an economic pest.

Mamestra brassicae *L.* (*Cabbage moth*). This has been reared in large numbers on cabbage and turnip but shows more difficulty with diapause and disease than tomato moth. However, the pupae may be refrigerated for a year at least and brought out at any time to continue the life cycle.

Agrotis segetum *Schiff* (*Turnip moth*). The rearing of this species has been abandoned owing to its susceptibility to disease.

Phlogophora meticulosa *L.* (*Angle Shade° moth*) This species can be reared in large numbers using the foliage of most low plants as food. The data are not yet complete on methods of rearing throughout the year. The diapause can be broken, but all eggs laid by moths emerging in November proved to be sterile.

Abraxas grossulariata *L.* (*Magpie moth*). Work has been started on this species but no means has as yet been found for breaking the diapause.

Porthetria dispar *L.* (*Gypsy moth*). This insect has certain advantages as an experimental subject. The male and female larvae can be separated. There has been no trouble with disease and its polyphagous habits make it suitable for studies on the effect of host plant in resistance. However, since it feeds on deciduous plants it may not be reared all the year round. It has been found that by storing the eggs in a refrigerator and bringing them out at intervals during the late spring and summer, several generations of the moth can be reared in a year.

Sphinx ligustri *L.* (*Privet Hawk moth*). This insect is reared because its large size make it a suitable insect for special studies. It has been reared on privet, lilac and ash, but all attempts made to break the diapause so far have failed.

Cerura (Dicranura) vinula *L.* (*Puss moth*). This insect has been reared on willow and poplar but it is not thought to be a useful test animal.

Odonestis (Cosmotriche) potatoria *L.* (*Drinker moth*). This has been reared in small numbers on coarse grass but does not appear to be a likely subject. An attempt to break the diapause was unsuccessful.

Artia caja *L.* (*Garden tiger moth*). This insect has been reared on dock, cabbage and other plants. An unsuccessful attempt to produce a strain without a diapause was made by rearing at a constant temperature of 75° F. Further work may be done on this species.

Athalia colibri (*Turnip sawfly*). Since no hymenopterous pest was available as a test insect, rearing work was started on this species. Little data are as yet available but breeding under fluorescent lights broke the diapause on one occasion.

Effect of herbicides on bees

Mr. G. D. Glynne-Jones was seconded from the National Advisory Service in May, 1947, to work conjointly with the Insecticides Department and the Bee Department on the effect of herbicides on bees.

Preliminary experiments with proprietary herbicides showed that, at field strength, herbicides containing Dinitro-o-cresol were highly toxic both as stomach and as contact poisons to bees. Proprietary herbicides containing 2-Methyl-4-chloro-phenoxyacetic acid (MCPA) (hormone type) showed no evidence of contact effect at field strengths but some evidence of stomach poisoning effect.

Some work was then carried out to obtain a suitable laboratory technique for quantitative studies. By means of these techniques it was ascertained that the median lethal dose (m.l.d.) of sodium-dinitro-o-cresylate as a stomach poison to bees lay between 0·0020 and 0·0025 mg. per bee. It was further found that the m.l.d.'s as a stomach poison for 2–4 dichloro-phenoxy-acetic acid (DCPA) and its sodium salt were about the same and lay between 0·002 and 0·005 mg. per bee. A stable suspension of MCPA could not be prepared, but the toxicity of the sodium salt was similar to DCPA and its sodium salt.

Experiments on the contact poison effect showed the m.l.d. of DNOC to be 0·0025 mg./sq. cm. at 30° C. and of its sodium salt to be 0·0035 mg./sq. cm. at 30° C. The conditions of the test greatly affect the toxicity. MCPA and DCPA proved non-toxic at all the concentrations tested.

Experiments made by exposing bees to residual films of DNOC and its sodium salt showed that the conditions, particularly humidity, greatly affected the results, kills varying from zero to 100 per cent. mortality were recorded.

A series of experiments were made on the attractant, or repellent properties of DNOC and its sodium and ammonium salts. The general evidence was that these substances are repellent.

The overall evidence is that DNOC and its salts when applied as weedkillers are a potential danger to bees, particularly if applied to open blossom, the "hormone" type weedkillers are safer. Further work is required, especially in the field, before any very definite statements can be made.

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