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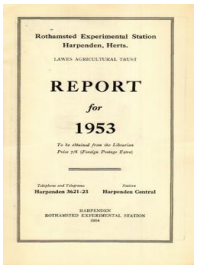

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PHYSICAL PROPERTIES AND CONTACT TOXICITY OF DDT AND SOME RELATED COMPOUNDS

By

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Research on biologically active compounds, including insecticides, is often done by making a group of similar chemicals and testing them by some standard method. This may lead to the discovery of new insecticides. But the aim is sometimes to correlate chemical structure with toxicity in the hope of finding some general rule by which the toxicity of any chemical in the group may be foretold.

The physical as well as the chemical properties of a compound can affect its toxicity, and when chemical structure is changed, physical properties are nearly always changed as well; so that it may sometimes be misleading to relate toxicity directly to chemical structure unless changes in the important physical properties are small.

Some of the work done in the past few years at Rothamsted has been aimed at finding out what effect the physical form can have on toxicity, and what physical properties are desirable in an insecticide when it is applied directly to the insects' bodies. This work is academic, but may in the end have some effect on the way insecticides are made up for field use.

All the compounds we used are chemically related to DDT. They are all crystalline solids which do not dissolve in water. They are all contact poisons. This means that the insects can be killed without having to eat the poisons; contact with the insects' bodies is enough. None of the poisons give off vapours that can kill the insects.

Two or more types of aqueous suspensions were made with each compound. One type (colloid) contained very small particles of supercooled liquid poison, probably about 0·0001 mm. in size. The others contained crystals, often about 0·05 mm. These crystals were uniform, but the size varied from one compound to another; in some cases several different types of suspension were made of a single compound, each containing uniform crystals of characteristic size.

The toxicity of each suspension was found by a method which involves dipping saw-toothed grain beetles (*Oryzaephilus surinamensis*) for a few moments in the suspension (McIntosh, 1947*a*). After this the suspension is drained off, and the beetles are left with a coating of poison sticking to them. The dipping does not drown the insects; they are kept for 24 hours or more after dipping, and then counts are made to see how many have died from the poison. It is important that the temperature of the insects is kept constant during this time, because changes in temperature nearly always affect the kill.

In this way the suspensions were compared in pairs; a suspension of crystals of each poison was compared with the same poison in

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colloidal form. This review discusses how the difference in toxicity between colloid and crystals may be related to the physical properties of the poison.

DDT was one of the compounds with which several different suspensions of crystals were made, each containing crystals of a different size. Crystals of DDT are needle-shaped or plate-shaped; the crystal size was varied from about 0·06 mm. to about 0·4 mm. When these suspensions were compared on grain beetles kept warm (27° C.) after dipping, the colloid was always the least toxic suspension; the longer the needles, the more toxic they seemed to be (McIntosh, 1946). The longest needles were about fifteen times more toxic than the colloid. This was unexpected, but the immediate cause was not hard to find. When the insects are taken from the suspension, poison sticks to them; it can be washed off, and the amount retained can be found by chemical analysis. This showed that the insects retain much more poison from a suspension of long needle-shaped crystals than from a suspension of colloidal particles. The extra dose received was in fact almost enough to account for the higher toxicity of the suspension of crystals (McIntosh, 1947*b*). Differences in toxicity amongst the other suspensions of DDT crystals can be explained in the same way; crystal size decides retention. Tests with other compounds besides DDT suggest that retention of this sort is purely mechanical. Retention by one insect species depends on crystal size only; different poisons with crystals of the same size are retained equally well. Plate-shaped crystals are not retained so well as needle-shaped crystals. Poorest retention was found with plate-shaped crystals of about 0·025 mm., and not with the very smallest particles. With some poisons there is no method for micro-analysis. In such a case the retention can be guessed by comparison with some other compound that gives analyzable crystals of the same size.

The results of all comparisons of toxicity by dipping must be corrected one way or the other to allow for differences in retention.

It may seem at first sight as if the results of the tests with DDT can be completely explained by differences in retention. This is not so. A very short description of what insect cuticle (skin) is like may make this clearer. Cuticles vary in structure from species to species, and even from one area of a single insect to another. But there is always a thin waxy layer on the very outside (Wigglesworth, 1948; Beament, 1948). The first thing a contact poison lying on the cuticle must do to get into the insect is to dissolve in this wax layer. Without this, nothing can happen to the insect. For this reason the need for a contact insecticide to be soluble in fat has often been stressed.

With DDT it is thought that there are certain spots on the cuticle surface which are more easily penetrated than the rest, or which form short-cuts to the site of action of DDT inside the insect (see, for instance, Schaerffenberg, 1949; Wiesmann, 1949; Fisher, 1952). Poison applied to them kills the insect more efficiently than the same amount of poison applied anywhere else. The wax covers the sensitive spots as well as the rest of the cuticle, and so the first step in penetration must be the same everywhere, whether the insect dies as a result or not.

The wax layer is very thin, and must soon become locally

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saturated beneath and round about particles of poison that are in contact with it. If saturation can be kept up long enough, especially on a sensitive spot, the insect will die.

After insects have been dipped, the film of poison sticking to them becomes dry. With colloidal poison the film is not even, but takes the form, to begin with anyway, of little blobs of supercooled liquid poison. The chances of hitting a sensitive spot with a blob or with a crystal must be about the same. One might naturally expect that the poison from the blobs would dissolve more quickly in the wax than the poison from the crystals; colloidal poison should be more toxic than crystals, or should act more quickly. However, the two forms of DDT are in fact almost equally toxic.

Counts of kill are usually made one or two days after treatment. The choice is largely one of convenience. But it did not seem to matter whether they were made after 1½ hours or 72 hours; the ratio of toxicities was always the same (McIntosh, 1949). So we have the unexpected fact that the speeds of solution of the two forms of DDT in wax are, as far as can be judged from the biological tests, nearly the same. Speed of solution does not seem to decide speed of kill.

What has been said so far applies to insects that are kept warm (27° C.) between dipping and counting. If the insects are treated with the same two forms of DDT and then kept cool (11° C.) instead of warm, the relative toxicity is reversed; the colloid is now more toxic than the crystals by about the same amount as it is less toxic to the warm insects. Tests by injection of suspensions into larger insects give similar results, and suggest that the difference in toxicity at 11° C. is largely a difference in speed of action; if the injected insects are kept cool for long enough, the kill from the crystals catches up on the kill from the colloid (McIntosh, 1951*a*). The process of dissolving is slowed down in cool insects, but it is slowed down more for the crystals than for the colloid. The physical theory of very small particles supports the idea that they should be relatively more toxic at lower temperatures (McIntosh, 1951*b*).

One effect of cooling the insects is to accentuate the difference in speeds of action between small and large particles, making it easier to measure. Other compounds related to DDT behave in somewhat the same way when tested as contact poisons on *O. surinamensis* kept cool after treatment. The colloidal form is always more toxic than crystals, but the size of the difference in toxicity varies from one analogue to another.

Two properties of dissolving materials might be expected to affect this difference in toxicity.

Firstly, the deposits left by the colloidal poisons are made up of globules to begin with, but often crystallize later. The speed at which this happens varies from compound to compound, and can be measured in *in vitro* tests. If the deposit crystallizes quickly, it is soon not very different from the deposit left by a suspension of crystals; the difference in toxicity between colloid and crystals is likely to be small.

Secondly, if it is in fact necessary for poison to saturate the wax layer, then the speed at which a poison can dissolve in the wax may be more important than the solubility itself. It is possible to measure

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the time it takes for crystals of a poison to bring about saturation of olive oil *in vitro*. This figure was taken as a guide to their speed of solution in insect wax. It was not possible to measure the speed of solution of deposits from colloidal poisons in olive oil; they dissolve quickly, and it was assumed that they all dissolve at the same speed. Different poisons with crystals of the same size do not necessarily bring about saturation at the same speed. If the crystals dissolve slowly, the difference in toxicity between colloid and crystals is likely to be large.

When allowance is made for differences in retention, the analogues fall into two groups. Each compound in the first group shows a difference in toxicity of about eight times; the colloid is about eight times more toxic than the crystals if counts are made one day after treatment. In the second group the differences in toxicity between colloid and crystals are very much bigger; the values found lie between thirty and eighty.

It was said that if a compound gives a slowly-crystallizing deposit from colloid *or* gives crystals that dissolve slowly, the difference in toxicity may be large. But the tests showed that each of the compounds giving a large difference in toxicity had *both* these qualities. One was not enough. The reason why both should be necessary is not clear. It may be that this is not a general rule; one quality or the other, if extreme enough, might produce a large difference in toxicity.

The lipoid-solubility, or solubility in fats, is often said to be important in deciding the toxicity of a contact insecticide. The implication, sometimes stated directly, is that in a group of very similar compounds like close analogues of DDT the most soluble compounds are the best contact insecticides (Martin & Wain, 1944; Browning *et al.*, 1948; Skerrett & Woodcock, 1952). It is certainly not true with this group of DDT analogues. They are all fat-soluble, but there is no relation at all between the toxicity (of colloid) and solubility in olive oil, which is often taken as a convenient measure of solubility in body fat. The difference in toxicity between colloid and crystals is not related to fat-solubility either.

The reactions of the insects to different sizes of particle seem to support the idea that the first step in penetration is solution of poison in some solvent, presumably the cuticle wax. It may seem rather obvious that it is better to use colloid than crystals, and that the qualities making for efficiency are slow crystallization of supercooled poison if it is applied as a colloid, and quick solution of crystals if the poison is applied as a solid. But the tests of DDT on warm and cool *O. surinamensis*, and of the other analogues, suggest that with this species the qualities that affect speed of solution do not decide speed of kill if the insects are kept warm after treatment; they are important only if the temperature is low. These qualities ought to apply to some extent to any stable contact insecticide and to almost any species of insect; this has still to be confirmed. The temperature at which they become important will not necessarily be the same for different species.

Crystallization of DDT can be prevented by mixing other compounds with it. This kind of non-crystalline DDT is more toxic than pure DDT in tests of dusts on sheep keds or vinegar flies (Riemschneider, 1950), and in tests of films on mosquitoes

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or DDT-resistant houseflies (Ascher, Reuter & Levinson, 1951; Ascher & Reuter, 1953). In the film tests the non-crystalline DDT may stick better to the insects than crystals do, and for this reason may seem to be more toxic. But Beran (1952) found that impure non-crystalline DDT is more toxic than pure crystalline DDT when equal amounts are applied directly to houseflies. In all these tests the insects were kept warm (24–28° C.) after the poison was applied. From this it seems more likely that a low crystallization tendency is in general a helpful property at all temperatures and not just at low ones.

In practice it will seldom be possible to apply solid contact poisons in colloidal form. They are often formulated in one crystalline form or another. If a poison is to be efficient it must be able to saturate the cuticle wax quickly. This may be the result of its own properties, or of formulation; but in either case attention should be given to speed of solution rather than fat-solubility, which has perhaps been over-emphasized in the past. Some degree of fat-solubility is certainly necessary, but it need not be very high.

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