The Occurrence of Insecticides in the Blood of Staff of a Locust Control Organization[1]

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The Desert Locust Control Organization for Eastern Africa (DLCO-EA) is an international organization financed by the countries of Eastern Africa and established for the control of the desert locust, Schistocerca gregaria (Forskal). This insect occurs intermittantly in vast numbers and can be the most devastating agricultural pest from West Africa through the Middle East to India. During a plague, swarms of adult locusts have been estimated to contain up to 50 million individuals per square kilometre with each swarm usually extending over several tens of square kilometres. Individually each adult locust weighs about 2g (Fig.1). The destruction of locusts on a large scale is most often carried out by applying concentrated insecticide solutions from small aircraft (Anti-Locust Research Centre, 1966). The insecticides used are crude BHC solutions containing 100-200 g/l of gamma isomer (lindane), dieldrin solutions containing 200 g/l of active ingredient and even technically pure fenitrothion or malathion. In addition to the DLCO-EA staff there are other personnel employed by the local Ministries of Agriculture who may be employed on locust control in addition to other pest control work. To facilitate operations in the different areas where spraying has to be carried out the Organization maintains bases at Asmara and Dire Dawa in Ethiopia, Nairobi in Kenya, Hargeisa and Mogadiscio in Somalia and Khartoum in the Sudan. The bases are equipped with vehicles, camp kit and large stocks of insecticides as necessary.

For many years the Organization has been concerned about the possible hazards presented by insecticides to the staff. In the mid-nineteen fifties when a 20% DNOC solution was used regular checks of DNOC levels in the blood of exposed staff were made (Rainey, 1958). The use of DNOC was stopped as soon as the safer BHC became readily available and, although occasional attempts were made, the regular monitoring of exposure to insecticides also had to stop due to lack of suitable equipment and techniques. At the end of 1971 however, owing to the wide-

[1] Any views and opinions expressed in this paper are those of the author and do not necessarily reflect the views of the FAO as a whole.

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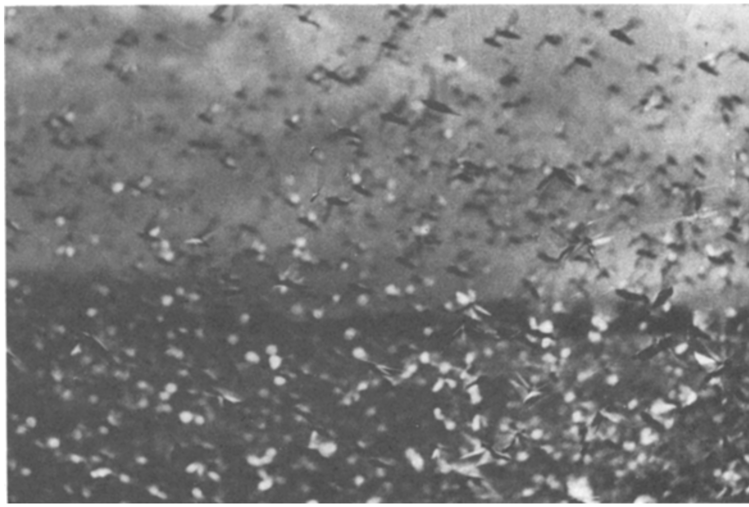

Fig. 1: A small section of a swarm of locusts.

spread concern about the use of organochlorine insecticides the Food & Agriculture Organization of the United Nations set up a project to study the implications of using alternative insecticides to the organochlorine compounds for locust control. The project is funded by the Swedish International Development Authority and the equipment now available enable measurements to be made of organochlorine insecticide levels and cholinesterase activity in blood. It therefore seemed appropriate to re-start the monitoring tests of blood of exposed staff.

Blood samples of 2.5 ml were taken from the arm of each person and stored in ice and later a deep freezer in a heparinised tube until the readings could be made. Because the numbers were large and the rate of working slow storage for over one month was needed for some samples. Each sample was analysed in two ways: firstly cholinesterase activity was measured by means of the radiometric method of Winteringham and Disney (1964). Secondly, a hexane extract of the blood was prepared for gas chromatography (GLC) in order to measure organochlorine insecticide concentrations.

METHOD FOR ORGANOCHLORINE INSECTICIDES

Several methods are available requiring little or no clean-up (see Zweig & Sherma, 1972) but centrifuging to break the emulsion formed using a non-aqueous solvent proved difficult and the following method was finally used. The blood was always frozen so was also haemolysed.

1 ml was pipetted into a glass test-tube together with 1 or 2 ml of redistilled hexane. The test-tube with stopper was then attached to a slowly rotating arm (8 rev/min) so that the tube rotated end-over-end for one hour. After standing, or ultra-sonic treatment, the hexane separated off and could be injected directly into the GLC. A Varian 1440 series GLC was used with Nickel electron capture detector, nitrogen gas flow 12 ml/min through a 200 x 3 mm stainless steel column containing 5% QF1 on Varaport 30. Temperatures, column 195, detector 290 (indicated), injector 200°C. At the moment of injection a stop clock was started and the time the top of each peak reached was noted. This proved to be a very sensitive confirmatory test as peaks were found which were consistently different from the reference standards by only a few seconds.

Checks were made for the presence of gamma-BHC, dieldrin (HEOD), p,p'-DDT and p,p'-DDE. When blood samples were spiked with the insecticides and left overnight recovery was complete except for DDE where it was estimated to be 80%. The GLC system used did not separate DDD from DDT but in practice this was not important because the amount of DDT present was normally small compared with the amount of DDE. Samples from some people gave several small peaks around the time of gamma-BHC. These might have been other BHC isomers, but when compared with alpha-, beta- and delta-BHC their retention times were not identical (see fig. 2).

The minimum quantities of each of the insecticides which could be detected by the procedures used are shown in Table I where they may be seen to be not more than 2% of the maximum permissible amounts (see below).

DISPLAY OF RESULTS

In expressing the results of the tests it was important to display them in a way that staff of all educational levels could readily understand. Many of the staff had been handling insecticides for years and as a result of frequent warnings as to the hazards involved, were concerned that they were being affected by the insecticides. A simple visual form was therefore used. The acetylcholinesterase activity values for the staff of each station were grouped together and the mean and standard deviation calculated. Individual values for each person were then expressed as fractions of a standard deviation above or below the mean value. Particularly low values were therefore readily visible.

A similar procedure was adopted for the organochlorine insecticides with the levels for each person expressed as a percentage of a level which should not be exceeded, the maximum permissible level. Although arbitrarily chosen, symptoms of poisoning are generally unlikely to occur below this level.

The advantages of using such an arbitrary number were thought to outweigh its possible disadvantages. Hunter (1968) in a review of the allowable concentrations of organochlorine pesticides in the body states that traces of these materials should be regarded as normal constituents of the body and proposed some acceptable levels which should not be exceeded. For dieldrin (HEOD) he proposed a level of 3.5 ng/ml for whole blood whilst Schafer (1969) observed that a blood level of less than 15 ng/ml is expected from populations not occupationally exposed to dieldrin. Following exposure to dieldrin it appears well established that with a concentration of 200 ng/ml of dieldrin in the blood a person will be liable to show symptoms of poisoning under suitable stress (Jager, 1970). Clinical symptoms of exposure have been observed within the range 20-530 ng/ml (Robinson & Hunter, 1966). From this wide range 200 ng/ml is clearly too high and 100 ng/ml was selected as a reasonable compromise.

Studies with DDT are not so precise. Both p,p'-DDT and its conversion product p,p'-DDE must be measured. Schafer (loc.cit.) suggested a value of not more than 150 ng/ml for the combined total of DDT plus DDE whilst Hunter (loc.cit.) proposed

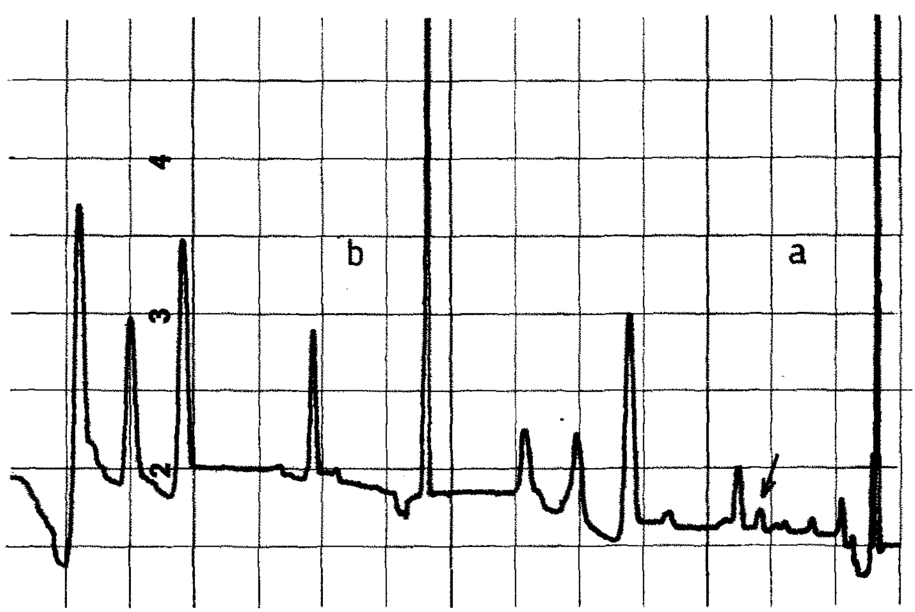

Fig. 2: GLC traces moving from right to left of (a) hexane extract from blood and (b) reference standards. Oven temperature programmed to rise from 150°C at time of injection at a rate of 4°C/minute. Reference standards: 20 pg gamma-BHC at 230, 80 pg of DDE at 475, 50 pg dieldrin at 574, and 250 pg DDT at 668 seconds. Peaks in blood sample occurred at 230 (arrowed), 475, 574, and 670 seconds corresponding to the insecticides.

TABLE I

Minimum detectable quantities of some organochlorine insecticides in blood compared with acceptable levels (ng insecticide per ml whole blood).

Insecticide	Minimum detectable concentration	Acceptable concentration for unexposed populations (1)	Upper acceptable limits used in this study
Lindane	0.4	N.A	20
Dieldrin (HEOD)	1	3.5	100
DDT (total equivalent	10	10	500

Notes (1) Hunter, 1968
N.A - not assessable on present information.

10 ng/ml as the threshold level for unexposed populations. In their study on blood and fat levels of DDT in unexposed people Robinson and Hunter (1966) found that the concentration of DDT in the fat was 306 times as great as in the blood. If the same factor holds for the studies made by Hayes et al. (1956) who fed volunteers 3.5 mg/kg DDT per day over a period of 18 months, then the level of 234 ppm DDT in the fat would correspond to a blood level of 764 ng/ml. Dale et al. (1967) found plasma levels of 395-737 ng/ml in occupationally exposed males with no clinical signs of intoxication. Single oral doses of 16 mg/kg and upwards are said often to produce convulsions whilst a dose of around 10 mg/kg may give rise to lesser toxic symptoms such as nausea, headache and sweating (Anonymous, 1951). Thus a daily dose of 3.5 mg/kg must be near the threshold where clinical symptoms of poisoning become manifest. A blood level for DDT plus DDE of 500 ng/ml was selected as the upper limit.

The rate of accumulation of gamma-BHC in the body tissues is much lower than for DDT although like DDT it is similarly stored in the fat. Three hours after the last convulsion in a boy who swallowed gamma-BHC the plasma was found to contain 290 ng/ml. Seven days later there were no symptoms and the plasma contained 20 ng/ml. Czegledi-Janko & Avar (1970) in a study of men occupationally exposed to lindane in a factory found a proportion of men with clinical symptoms and electroencephalogram (EEC) changes when the concentration of gamma-BHC in whole blood exceeded 20 ng/ml and this figure is selected as the upper limit (see Table I).

RESULTS

When the tests were started it was not anticipated that DDT would be the insecticide present in the blood of staff in the greatest quantity as almost all, when spraying, were using BHC and/or dieldrin. Extracts from the blood of some staff produced relatively enormous peaks which, at the time, were unknown. When however the times for reaching maximum peak height were compared with those for p,p'-DDE and p,p'-DDT the correspondence was exact (Fig. 2). A chart was drawn for each station as described so that every person could see the results from his own blood. Figure 3 shows the results from the Dire Dawa Base. It may be seen that the concentrations of DDE plus DDT are very high and this was presumably related to an exceptionally large outbreak of the African armyworm *Spodoptera exempta* (Walker) which had occurred earlier and against which DDT was used by many of the staff. In addition some DDT was no doubt used at home to kill mosquitoes which are abundant at times. Because of the high concentrations of DDE plus DDT a second set of blood samples were obtained and measured 7 months later from some of the Dire Dawa staff. These results are shown in Table II. Agreement between the cholinesterase values obtained on each occasion are reasonably good. Similarly the mean values for the concentrations of DDE and DDT show little change. This is surprising and suggests that there has been a continuing intake amongst at least some of the individuals concerned. Concentrations of the other insecticides were generally unimportant. The reality of the results became plain on talking to the staff with different from average results. Thus the person in Addis Ababa with the highest concentration of DDE plus DDT had transferred from Dire Dawa 18 months previously where he had knowingly been exposed to DDT and BHC (having complained of being unwell and seen by a doctor). The haematologist who did some of the tests had recently arrived from England and only DDE was detectable in her blood (30 ng/ml). The one man who had probably been most concerned with spraying operations throughout the past 10 years had the highest concentrations of gamma-BHC and dieldrin in his blood. In Hargeisa two people had noticeably higher concentrations of DDE plus DDT than average: one had recently transferred from Mogadiscio where DDT is used for mosquito control and the other had, some 18 months previously, been employed as a spray operator on a commercial cotton farm where DDT was much used. Thus the results themselves do appear to indicate the history of the person's absorbtion of insecticide whilst the method of display enabled the uneducated staff to see how the results from their bloods compared with those of their fellows.

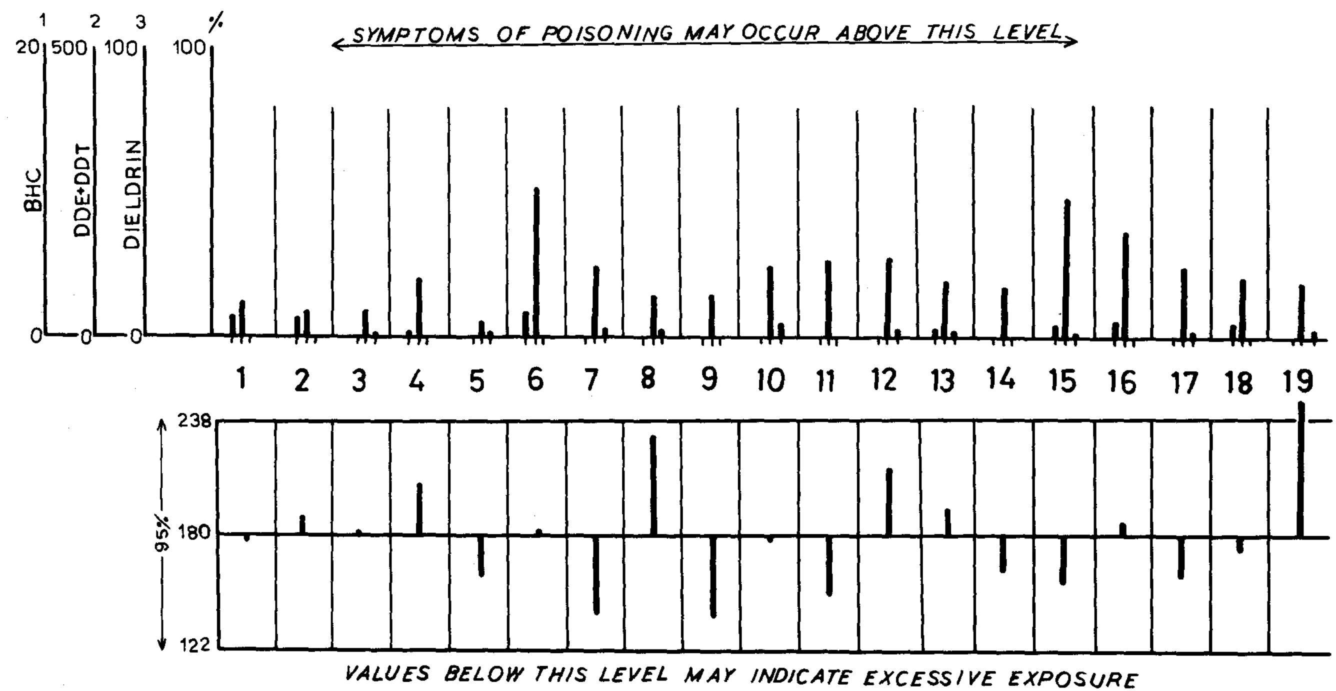

Fig. 3. Occurrence and effects of insecticides in blood. Nos. 1-19, different people. Upper trace: concentration of organochlorine insecticides expressed as percentages of the maximum permissible level. Lower trace; cholinesterase activity for each individual shown as a deviation above or below the mean, the ordinate showing the mean plus and minus two standard deviations (μM acetylcholine hydrolysed per ml whole blood per hour at 25°C).

TABLE II

Cholinesterase activities and concentrations of chlorinated hydrocarbon insecticides in bloods of individuals measured at 7 month interval. (Cholinesterase activity, μM acetylcholine hydrolysed /hour/ml whole blood at 25°C: concentrations in ng/ml)

Parameter measured and date of sample		Individual (number corresponds to that shown in fig. 3)							
		6	7	9	10	12	13	19	Mean Values
Cholinesterase value									
	August 1974	183	140	139	175	215	194	248	185
	March 1975	142	150	177	170	203	179	203	175
p,p'-DDE	August 1974	220	70	50	96	104	80	60	97
	March 1975	139	48	50	45	136	97	48	80
p,p'-DDT	August 1974	39	46	17	21	21	12	22	30
	March 1975	75	48	18	17	0	23	29	30
gamma-BHC	August 1974	1.7	0	0	0	0	0.4	0	0.3
	March 1975	0	0	0	0	0	0	0	0
Dieldrin	August 1974	0	2.4	0	5.5	2.3	1.3	0.8	1.8
	March 1975	10	1.0	4.1	8.8	0	4.4	3.6	4.5

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