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I hereby recommend that the thesis prepared under my supervision by Charles W. Kraul, M.D.

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Approved by:

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ENVIRONMENTAL HAZARDS OF THE

CHLORINATED HYDROCARBON INSECTICIDES

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by

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CHAPTER I

INTRODUCTION

With the introduction of DDT in 1942 as an effective insecticide, its subsequent development in the United States, and its eminently successful use during World War II by the armed forces of this country throughout the world, interest was aroused in the synthesis and development of new organic insecticides.

During this period there arose a deeper appreciation of the role insects play in the transmission of certain diseases to man and animals. Of equal importance was the growing realization of the necessity of protecting food crops more effectively from the ravages of plant destroying insects.

Among the chlorinated hydrocarbon insecticides which have been developed, the following compounds have had commercial success: DDT and its analogues, methoxychlor, DDD and DMC; benzene hexachloride and its gamma isomer, lindane; toxaphene; and the Diels-Alder condensation products, chlordane, heptachlor, aldrin and dieldrin. The newer products of this class, isodrin and endrin, are too new to have had extensive use.

Most of these compounds are now readily available to the general public in numerous formulations and in almost any combination.

The introduction of these newer toxic materials with such widespread application and free availability makes it important to determine the hazards which might exist for man. In this connection, it is of interest to discover, if possible, what does happen to man when he is exposed to these materials by accident or intention, either internally or externally, in greater or lesser quantity for a longer or shorter period of time.

In order to have a base-line from which to work, it is proposed to review briefly the chemical and physical properties of the important chlorinated hydrocarbon insecticides and the available toxicologic data derived from animal experimentation.

A review of the available reported human cases of intoxication due to apparent or real exposure to these insecticides will be undertaken for the purpose of trying to learn how these materials affect man. It will also be interesting to compare what has been described in animals with what has been reported to happen to man suffering from insecticide intoxication.

Furthermore, approximately 80 cases of alleged intoxication due to aldrin and dieldrin, and one case allegedly due to endrin, will be reported for the first time in this thesis. An opinion will be offered as to which of the cases are actual and which are apparent.

With this background, an attempt will be made to evaluate the environmental hazards associated with the manufacture and use of the chlorinated hydrocarbon insecticides.

From the information as outlined, it may be possible to reach some

tentative conclusions as to what protective measures should be instituted, how to make a diagnosis of chlorinated hydrocarbon insecticide intoxication, and what therapy has been found effective.

Lastly, some of the discussion regarding the concept of chronic chlorinated hydrocarbon insecticide intoxication will be examined.

No attempt has been made to consider the entomologic data except incidentally. In the consideration of aldrin and dieldrin some mention has been made regarding the effects upon insects. This was considered necessary in order to establish their ranking among the other chlorinated hydrocarbon insecticides as to effectiveness and persistence.

It should be noted that this thesis does not pretend to be an exhaustive review of the literature which deals with the toxicology of the chlorinated hydrocarbon insecticides. However, most of the important references have been consulted and, in most cases, direct reference to these sources has been made.

CHAPTER II

DDT

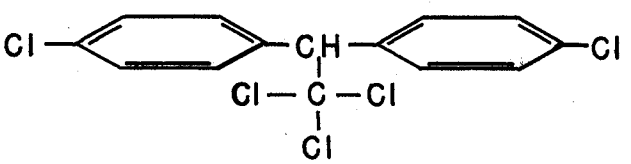
Historically, one can date the beginning of the chlorinated hydrocarbon insecticide industry with the development of DDT for field use in 1943 by the United States Army in cooperation with the various federal bureaus, scientific committees, universities, and private laboratories (1).

Although the synthesis of DDT had been accomplished by the German chemist, Othmar Zeidler (2) in 1874, it was Paul Mueller (3) who first demonstrated the outstanding insecticidal qualities of DDT in 1940, and for this contribution he was awarded the Nobel prize in 1948.

A. Physical and Chemical Data:

The compound commonly referred to as DDT has several names. Sometimes it is referred to as p,p'-dichlorodiphenyltrichloroethane. The official name in the United States Pharmacopoeia for this compound is Chlorophenothane. The correct chemical name is 2,2-bis(p-chlorophenyl)-1,1,1-trichloroethane.

The empirical formula is $C_{14}H_9Cl_5$. The structural formula may be represented as



It is convenient to consider DDT and its analogues as being built upon the structure of 1,1-diphenylethane, or it may be helpful to regard the DDT molecule as methane substituted with two chlorobenzene groups and a chloroform group.

This last concept of the DDT molecule has produced several theories which attempt to explain the effectiveness and the insecticidal property of DDT, with but little success (4).

When reference is made to pure DDT, the p,p' isomer is meant. It is a white crystalline substance without odor or taste. It is stable at ordinary temperatures and only slightly volatile, its vapor pressure being 1.5×10^{-7} mm. Hg at 20 degrees C. (5).

DDT exists in several isomeric forms depending upon the position of the substituted chlorine atoms, viz.: p,p'; o,p'; and o,o' isomers. However, the p,p' isomer is the insecticidally active component of technical DDT (6).

DDT is prepared by condensing one mol of chloral with two mols of chlorobenzene in 98 per cent sulfuric acid at 15 degrees C. The purity of DDT may be inhanced by using an excess of chloral (7).

Technical DDT is approximately 70 per cent pure, consisting of 70 per cent of the p,p' isomer, from 10 to 15 per cent of the o,p' isomer, and about 15 to 20 per cent of related compounds and impurities (DDD, o,p'-DDT sulfonate, PDB and excess chlorobenzene). It has a melting point of 89 degrees C. and a setting point of 88 degrees C. or higher (6).

DDT is almost insoluble in water (0.0002 ppm.) although colloidal solutions up to 0.2 ppm. in water may be formed. It is readily soluble in

most organic solvents. From 4 to 10 per cent solutions can be made in petroleum solvents and about 1.5 per cent solution in 95 per cent ethyl alcohol.

The solubility of DDT in a variety of solvents is presented in the following table:

Solubility of DDT at 27-30 degrees C.

Solvent	Grams/100 ml.
Acetone	58
Benzene	78
Chloroform	45
Cyclohexane	116
Ethyl ether	28
Ethyl alcohol 95%	2
Ethylene dichloride	59
Isopropyl alcohol	3
Kerosene, ordinary	8-10
Kerosene, refined	3.7-4.5
Xylene	57

Data from Jones et al (8).

Mixtures of methyl naphthalenes like Velsicol AR-50 are often added to maintain DDT in solution in petroleum distillates. By adding an emulsifier to appropriate DDT solutions, emulsion concentrates may be prepared.

Incompatibilities: DDT decomposes at 195 degrees C. Normally, sunlight does not decompose DDT. In oil solution, it is slightly decomposed by ultraviolet light. It has been reported that dusts lose their toxicity at a more rapid rate at higher relative humidities, which seems to be true also when they are used in tropical climates. Hot, dry weather seems to

hasten the loss of DDT from plants but this may be due to the increase in its volatility (7).

DDT is attacked by alkali to produce dehydrochlorination to dichloroethylene or the ethane analogue. However, dehydrochlorination may be delayed by such solvents as kerosene, fuel oil, or methylnaphthalenes. Because of the instability of DDT in alkaline media, it is incompatible with dolomite, fuller's earth and kaolin. It has been reported that there is slight decomposition when combined with Bentonite, Bordeaux mixtures, the fungicide Fermate, and some talcs and pyrophyllites. Oxides or chlorides of aluminum and iron catalytically decompose DDT (9).

DDT combines safely with lead and calcium arsenate, cryolite, fluosilicates, hydrated lime, lime-sulfur and Paris Green (9) (7).

Analytic Methods: The Schechter-Haller method (10) is considered the most satisfactory method available at present from the aspects of sensitivity (10 micrograms) and specificity. In principle, DDT is converted to its tetranitro derivative. Sodium methylate produces a blue color with p,p'-DDT and a violet color with o,p'-DDT. No color is produced by any of the other chlorinated hydrocarbon insecticides. This method is useful in detecting DDT residues in plants, foods and in animal tissues.

The Stiff-Castillo method (11) is also sensitive to 10 micrograms. In principle, DDT is heated in pyridine with xanthydrol and potassium hydroxide. A red color is produced. The amount of water in the pyridine is critical.

The Alessandrini modification of the Schechter-Haller method (12) is

simpler but is much less sensitive.

A spectrophotometric method is used which shows maximum absorption at 236 millimicrons (13).

Finally, methods based on the dehydrochlorination of DDT are available. Basically, they involve the addition of alkali and then the titration of chloride by Volhard's method (14). (a) Total chlorine method: The sample containing DDT is refluxed with metallic sodium and isopropyl alcohol. All of the chlorine atoms are removed from the molecule. The amount of DDT present is twice the weight of the chlorine determined. (b) Hydrolysable chlorine method: The sample is refluxed with potassium hydroxide in ethyl alcohol (or the sample may be treated with 4.5N ethanolic ammonium hydroxide at 45 degrees C.). Only one chlorine atom is removed (14).

It is possible to arrange conditions so that only the p,p'-DDT is dehydrochlorinated and not the o,p'-DDT or other impurities (7).

Formulations: There are several hundred formulations available that contain DDT (9). DDT may be obtained as solutions, (oil solutions in petroleum or other suitable solvents); emulsion concentrates which are diluted with water; powders for dusts; wettable powders for sprays; aerosols; aerosols for dispersions as smokes; paints; wall washes; wax; and 3-5 per cent of DDT in flat oil paint (7) (9).

B. Toxicity - Experimental Evaluation:

It would be pointless to attempt to review completely the literature dealing with the toxicity of DDT in warm-blooded animals since it has been done so thoroughly and well several times in the recent past. For the more

detailed reviews dealing with this subject, reference should be made to the articles by Stammers and Whitfield (15), Glassman and Buchan (16) and Heyroth (17).

However, statements about which there seems to be fairly general agreement regarding the toxicity of DDT will be included so that the background for discussing the reported human cases will be at hand.

Immediate Toxicity: The oral LD_{50} dosage ranges vary widely with the species of animal tested, among the individuals of a species, and with the vehicle used. The following table lists the oral LD_{50} ranges for DDT:

Oral LD_{50} Ranges for DDT

Test Animal	Dose DDT mg./kg.
Mice and Rats	150-250
Cats and Dogs	150-300
Guinea pigs and Rabbits	300-500
Monkeys	>200
Cows and Horses	>300
Sheep and Goats	about 1000

Data from Heyroth (17) and Council on Pharm. and Chem. A.M.A. (18)

The data presented by Konst and Plummer (19) appear to indicate that when DDT is given in dry form in capsules it is less toxic than when it is given in an oily solution. Rats, without exception, were able to tolerate doses of DDT up to 500 mg./kg., when administered in the dry state.

On the other hand, DDT in solution has a much greater tendency to be absorbed through the gastro-intestinal tract. The oral LD_{50} ranges quoted

in the table were obtained by using solutions of DDT in either olive oil or corn oil (20) (21) (22) (23).

When DDT is administered as an aqueous suspension by the oral route, data similar to those derived from the dry feeding experiments are obtained (19) (21) (24).

It appears that the absorption of DDT through the gastro-intestinal tract is enhanced by the presence of an oil. Even so, the absorption from the digestive tract is slow and incomplete.

There may be a latent period of several hours following the ingestion of a toxic dose of DDT before symptoms of toxicity are noted (21) (18).

Intravenous LD_{50} dose of DDT: The intravenous route of administration of a suitable preparation eliminates the uncertainties associated with gastro-intestinal absorption. By this method the most direct evidence concerning the immediate toxicity of this substance may be obtained.

The minimum intravenous lethal dose for most species of animals is approximately 50 mg. DDT/kg. body weight. The following table has been taken from National Research Council Report No. 149 (22):

The LD_{50} Dose of DDT Where Administered Intravenously as a Lecithin-peanut Oil Emulsion Containing 1% DDT

Animal	LD_{50}, mg./kg. of DDT	Hours until Death	Reference (22)
Rat (adult male)	40-50		(26) (25)
Rabbit	40-50	2-5	(26) (25)
Cat	30-40	2-5	(26) (25)
Cat	50	5½	(27)
Dog	Between 50 & 75		(28) (25)
Monkey	Between 60 & 75		(28) (25)

These data are in accord with those cited by Heyroth (17) in his review of the literature.

When DDT is administered intravenously as an emulsion in sufficient dosage, symptoms of intoxication appear within 3 to 5 minutes and are well developed within 30 minutes. Death or complete recovery of the animal from all symptoms within 18 hours following the intravenous administration of DDT seems to occur regularly (22) (25).

Intraperitoneal Injections of DDT: The experiments in which DDT was given by the intraperitoneal route indicate that DDT is not readily absorbed from the peritoneal cavity (29). The amount of DDT required to produce symptoms by this route is much greater than that required by the intravenous route (21) (30). In general, it is approximately 10 times the intravenous dose.

Intramuscular and Subcutaneous Injections of DDT: The variation in the dosage required to produce symptoms and death in animals, following intramuscular and subcutaneous injections of DDT, and the predominance of high values in the data of several investigators, suggest that DDT is poorly and irregularly absorbed following the injection of this insecticide by these routes (30) (19) (31).

Percutaneous Absorption: The conclusions of Draize, Nelson and Calvery (31) have been confirmed by other experimenters (32) (21). In the main, there is agreement that (a) powdered DDT is not absorbed through the skin, (b) absorption through the skin can occur when DDT is applied in a suitable solvent, and (c) DDT has no effect upon the skin of rabbits other

than the production of a mild erythema.

Cameron and Burgess (32) reported that powdered DDT applied to experimentally-produced wounds did not interfere with the normal rate of wound healing.

Inhalation of DDT: Neal and his co-workers (30) (33) have done experiments in which dogs, rats, guinea pigs and mice were subjected to only one exposure to DDT at the initial concentrations of 6.22, 12.44, or 54.4 mg./liter respectively without producing signs of intoxication in any of the animals except in mice which could tolerate only the lowest concentration. Other experiments indicated that young mice were more susceptible than mature mice (30).

From the consideration of the experimental data cited above it can be said that intoxication from absorption of dry DDT through the skin or the respiratory tract is unlikely.

Subacute and Chronic Toxicity of DDT: Glassman and Buchan (16) have summarized the data of several investigators which deal with the repeated oral administration of DDT. The following table is taken from their paper:

Reference	Dose DDT	Vehicle	Animals Species--Death		Duration of Experiment
Cameron & Burgess	50 mg./kg.	Liq. petrolatum & tragacanth	Rabbit	≠	30 days
			Rat	0	
	1000 ppm	Diet	Rat	≠	18 to 80 days
	500 ppm	Diet	Rat	0	3 months
	50 mg./kg.	Olive oil	Rabbit	≠	15 to 23 days
	50 mg./kg.	. . . . .	Cat	≠	12 to 15 days
Fitzhugh & Nelson	100-800 ppm	Diet	Rat	≠	2 years
Haymaker et al	150-350	Peanut oil	Dog	≠	90 days
Orr & Mott	100-200	. . . . .	Cow	0	3 weeks
	mg./kg.		Horse	0	3 weeks
			Sheep	0	3 weeks

Heyroth (17) stated that the data obtained by feeding experimentally contaminated diets to rats over long periods of time appear to indicate that when all of the food consumed by the rats contains DDT in concentrations less than 400 ppm., the length of their survival will not be lessened, and that when the dietary level is 200 to 400 ppm. their growth will be affected. However, a very few rats maintained on a diet of as low as 150 ppm. may exhibit signs of intoxication particularly when they lose weight rapidly for any reason. At high dietary levels, the reported results tend to be somewhat more variable, as might be expected from the fact that individual rats differ considerably in their resistance to DDT. The reproductive capacity of rats may be affected by the prolonged feeding of diets containing 50 ppm.

It has been pointed out several times (17) (34) that evidence of minimal tissue damage may not be uncovered by gross observations such as life-span of the animals, growth curve, or reproductive capacity. Biochemical or histological examinations of the tissues of the experimental animals may provide evidence of such damage.

Lehman (35) states that in the 2-year feeding experiments in which rats were fed different concentrations of DDT in the diet, it was observed that the lowest level of DDT fed which produced gross effects was 100 ppm.; the highest level fed which produced no gross effect was 50 ppm. The lowest level at which tissue damage occurred was 5 ppm.; the highest level which produced no tissue damage was 1 ppm.

The data which have been presented in the preceding paragraphs indicate

in a general way the extent of the chronic oral toxicity of DDT in experimental animals. The literature is voluminous on this point and it cannot be reviewed in detail in this paper.

Heyroth (17) stated in 1950 that data on at least 49 experimental groups of rats fed at various dietary levels of DDT have been published. The experimental data concerning dogs, guinea pigs, mice, chickens and turkeys were not included in that estimate.

Toxicity Following Percutaneous Absorption: There is good evidence that DDT powder applied to the skin in concentrations up to 5 per cent over long periods of time is not absorbed to any appreciable extent (31) (32) (30). However, in the presence of oil, solvents and greases, the absorption of DDT is facilitated, and enough DDT may be absorbed to produce symptoms and even death (32) (31) (21).

The early work of Draize, Nelson and Calvery (31) suggested that DDT has a mild but definite sensitizing property. The view is now held that usually DDT is not a skin irritant nor is it a cutaneous sensitizer (36) (37). However, Glassman and Buchan (16) quote Schwartz as stating that a small predictable incidence of cutaneous sensitivity to DDT powder may be expected.

It should be remembered that solvents and other materials used in the formulation of DDT preparations are capable of producing cutaneous irritation, sensitization and photosensitivity (1) (16).

Toxicity Following Inhalation: The large size of the particles and the relative insolubility of DDT powder make it unlikely that inhalation of DDT in this form would be a source of hazard (30). The use of the standard DDT

aerosol which contains 3 per cent of DDT will not produce symptoms of intoxication because of the rapid settling out of the DDT from the atmosphere (1). This phenomenon of rapid settling seems to be true of aerosol, smoke, or spray (30).

The "inert" diluents used in formulating DDT wettable powders may produce a persistent cough from bronchial irritation (18). Furthermore, solvents may also contribute occasionally to the production of irritation of the mucous membranes of the eyes and respiratory tract (30). In sufficient concentrations, solvents are capable of producing systemic effects such as nausea, vomiting, fatigue, headache, and other nervous system manifestations (1).

In contrast to the DDT powders, DDT emulsions and certain oil solutions are more likely to be absorbed by the lungs on inhalation (33) (37).

Exposure of Large Animals to DDT: Welch (38) administered aqueous suspensions of DDT by stomach tube to sheep. A dose of 2 gms. per kilogram produced extreme nervousness and muscular jerking within 24 hours, which lasted for five days. A dose of 1 gm./kg. produced muscular tremors and incoordination which disappeared within 48 hrs. A dose of 0.5 gm./kg. produced only slight nervous symptoms which subsided within 24 hours.

Cattle given 0.5 gm./kg. of DDT by stomach tube became highly nervous with symptoms of incoordination and muscular twitching. The animals were normal by the sixth day. A steer given a dose of DDT at the level of 0.125 gm./kg. by stomach tube showed no effect.

DDT administered to sheep in daily doses of 4.5 grams (approx. 0.1 gm./kg.)

produced grave symptoms of poisoning after ten days. The symptoms disappeared after a rest of four days. Following the resumption of the same dosage for twenty days, the symptoms recurred but disappeared after a rest of two days. For the remainder of the 60-day feeding period the sheep showed only mild symptoms. It was concluded that sheep acquired a tolerance toward DDT in that the symptoms became progressively milder toward the end of the experimental period (38).

Orr and Mott (23) administered powdered DDT at levels of 500, 1000, 1500 and 2000 mg./kg. to sheep, which survived this treatment.

Bushland, Wells and Radeleff (39) reported that cattle, sheep, goats, hogs and horses suffered no ill-effects from the application of sprays and dips made from wettable powders of DDT. A concentration of 1.5 per cent DDT was applied on eight occasions at intervals of four days.

Radeleff, Bushland and Claborn (40) state that all livestock can tolerate single applications of 8 per cent DDT. As many as ten applications of 2 per cent DDT at 2-week intervals have failed to produce clinical changes. Cattle also have tolerated 36 applications of 0.5 per cent DDT at the same intervals.

In general, DDT is a relatively safe insecticide for use on animals.

Experimental Observations On Man: A few experiments have been done in which man was the experimental subject. The subjects were exposed to DDT by the routes of ingestion, inhalation, and percutaneous absorption. Neal and his group (1) reported that a subject ingested 475 mg. of DDT in olive oil. At a later time, he ingested 770 mg. of DDT in olive oil (11 mg./kg.).

Thorough medical examinations both before and after the ingestion of DDT revealed no abnormalities which could be attributed to DDT. Examination of the urine showed that DDT was not present. However, DDA was found in the urine, the peak of concentration being present in the second 24-hour specimen. Small amounts of DDA were found to persist in the urine for a period of two weeks.

Domenjoz (24) reported that an experimental subject ingested 0.25 grams of pure DDT three times a day for three consecutive days. No ill effects which could be attributed to DDT were noted.

Perhaps the most interesting of the human experiments were done by Velbinger and his group (41) in Germany. Three subjects ingested DDT suspended in milk or cod-liver oil in doses of 250, 500, 750, 1000 and 1500 mg. The doses were usually taken after meals. The period of observation was twelve weeks.

The electrocardiogram, blood pressure, and sedimentation rate remained normal in the subjects. The urinalyses revealed no albumen or sugar. Urobilin and indican excretion were not altered. A questionable leucocytosis might have been present. A slight anemia was noted on occasion.

At 250 and 500 mg. dose levels, hyperesthesia about the lips was reported by some subjects. At 750 and 1000 mg. dose levels, sensation in the entire lower part of the face was affected. The gait was uncertain and movements unpleasant. Hypersensitivity to stimuli was noted, although the reflexes were normal. These effects began in two to three hours and were at the maximum at six hours after ingestion of DDT. This time period was variable

among the subjects.

Velbinger ingested the 1500 mg. dose of DDT. After 2½ hours a prickling sensation of the tip of the tongue, upper lip, and chin were reported. This sensation extended to involve the nostrils. Approximately four to five hours later, equilibrium was disturbed and a feeling of dizziness and numbness was noted. Nine hours after the ingestion of DDT, the disturbance of equilibrium still persisted. In addition, tremors of all the extremities were seen. Pupillary reactions and the nystagmogram were normal. The knee jerks were exaggerated and at times were clonic. At ten hours, standing on one leg became difficult. The finger-to-nose test indicated ataxia. The subject experienced headache and fatigue. At eleven hours, the subject vomited. He then slept until the next day when he reported complete recovery.

Lazar (42) reported that a man ingested about six pancakes made from DDT powder instead of flour without ill-effects.

Neal and his group (1) reported that two subjects were exposed to a dispersion of DDT in air, prepared by dispensing, every 15 minutes, 10.4 Gm. of an aerosol containing 5 per cent DDT, 10 per cent cyclohexanone, and 85 per cent "Freon" into a sealed chamber of 14,750 liters capacity, for one hour daily, on six consecutive days. These human subjects failed to show any subjective or objective manifestations of intoxication referable to DDT.

The same subjects were subjected to the dispersion of 10.4 Gm. of DDT aerosol of the same composition, every five minutes, for one hour daily, on five consecutive days. This exposure did not cause any subjective or objective

toxic effects referable to DDT.

Various investigators studied the effect of the application of DDT to human skin (32) (37) (43) (44) (45) (46). Patch testing revealed no evidence of sensitization or irritation due to DDT. The wearing of impregnated clothing and socks produced no effect which could be attributed to DDT. Experiments were reported by one investigator (48), in which the subjects kneaded DDT powder, acetone, and French chalk (47) and suffered no toxic effects.

Although human experimentation with DDT does not provide voluminous data, there is at least some indication of what might be expected to occur following exposure to DDT under known conditions.

For the most part, the human experiments involved short-term exposure to moderate dosages of DDT.

C. Metabolism. DDT may be absorbed into the animal body through the skin, respiratory tract, and through the gastro-intestinal tract under the proper conditions. Powdered DDT is not absorbed to any appreciable extent through the intact skin, nor is it readily absorbed through the lungs or the gastro-intestinal tract.

Absorption of DDT through the skin is facilitated by oily solutions and solvents. Absorption through the lungs is dependent upon the size of the DDT particle, the concentration of DDT in the air, the duration of exposure, and the solvent present. Lipoid solvents and the presence of fat in the intestines favor absorption (18).

Detoxification Mechanisms: DDT is partially detoxified in the liver to non-toxic metabolites which are excreted at irregular rates. The report

of Woodard, Davidow, and Lehman (49) seems to indicate that the metabolic breakdown of DDT occurs in the liver. That DDT is, in part, dechlorinated and oxidized to bis(p-chlorophenyl) acetic acid (DDA) has been demonstrated by several investigators, among whom may be mentioned White and Sweeney (50), Ofner, Woodard and Calvery (31), Stohlman and Smith (52) and Neal, Sweeney, Spicer and von Oettingen (53).

Degradation products may be found in the urine of man for several weeks following the administration of a single dose of DDT (1).

In experiments involving prolonged feeding of DDT to animals, the elimination of these degradation products increases until a plateau is reached and maintained (18).

Storage: The quantity of DDT stored depends on the level of intake and length of time over which the intake occurs (54) (49). DDT is found in the greatest concentrations in the fat depots, although it is present in all tissues (55) (56). It has been found in the blood and bile, and in liver, spleen, kidneys, adrenals and brain (21) (56). DDT has been demonstrated in the fat of almost all species of animals exposed to the insecticide. Reports are available which include analyses of the fat of cows, sheep, goats, monkeys, rabbits and rats (57) (28) (11) (35) (59).

Lehman presented data regarding the storage of DDT in the fat of rats. The following table has been adapted from the work done by him and his associates in the Food and Drug Administration (35):

Storage of DDT in the Fat of Rats as Determined by Chemical Methods

Feeding level ppm.	Duration of feeding	Average storage in ppm. Male	Female
1	15 weeks	13	18
5	15 weeks	53	98
10	15 weeks	65	137
50	15 weeks	284	588

From the above table, it will be seen that DDT is stored at a level which is 6 to 18 times that in the diet consumed. Females store larger quantities than males.

Another table is offered to show the rate of disappearance of stored insecticide from the fat of rats. The data are taken from Lehman's paper (60).

Rate of Disappearance of Stored DDT from the Fat of Rats
Values Expressed as Parts per Million

Initial Concentration Male	Female	Time on insecticide-free diet	Concentration at end of observation period Male	Female
26	32	4 weeks	19	30
63	104		43	81
82	160		46	71
284	588		157	267

It has been estimated that animals may accumulate amounts equivalent to several intravenous lethal doses of DDT without showing signs of intoxication (17). From the above tables it can be seen that the accumulation of DDT occurs at every level of intake. It appears that about 50 per cent of the

stored DDT is retained after three months.

Bushland, Claborn, Beckman, Radeleff and Wells (58) found that sheep and goats treated with dips containing 1.5 per cent of DDT made from either wettable powder or emulsion on eight occasions at 4-day intervals had stored DDT in the fat. The table summarizes the findings:

PPM. of DDT in Fat of Sheep and Goats After Eight Treatments at 4-day Intervals with Dips Containing 1.5% Insecticide

Formulation	After 1 month	After 7 months	
	Sheep	Sheep	Goats
DDT wettable powder	124	26	29
DDT emulsion	126	22	28

Laug and his co-workers (61) have reported the results of their analyses of 75 samples of human fat for their DDT content. The range of results extended from 0.0 ppm. in 15 samples to over 20.0 ppm. in 2 samples. The average value was equal to 5.3 ppm. The same report also dealt with the results of the determination of DDT in 32 samples of human milk obtained from patients with no known exposure to the insecticide. The range of concentration was between 0.00 ppm. in 2 samples to over 0.20 ppm. in 4 samples. The average value equaled 0.13 ppm. DDT.

Excretion: DDT is excreted slowly and incompletely from the animal body. Following oral administration of DDT, that part which appears in the feces is unchanged (63). Part of the DDT which is absorbed in the body appears within two to four days in the urine as DDA, to the extent of 75-80

per cent of the total urinary excretion, the rest being an ester of DDA (53). Some DDT is excreted in the bile in an undetermined form (62). That DDT is excreted in the milk of lactating animals and human beings has been established (64) (61). The remaining DDT has been believed to be stored in the fat depots of the body as unchanged DDT (11) (15).

Recently, there appeared a paper (65) which indicated that DDT may be stored as DDE (2,2-bis(p-chlorophenyl) 1,1-di-chloroethylene) in human fat. Sixty samples of human fat have been analyzed and it was found that the proportion of DDE found ranged from 33 to 90 per cent. Most samples contained more DDE than DDT. What the significance of this observation may be, remains to be determined.

D. Pathology: The pathologic changes that have been observed in animals poisoned with DDT vary with the dosage.

In general, the changes observed in the tissues of animals exposed to large doses of DDT are those of mild to moderate fatty degeneration. Liver changes are characterized by hyaline and fatty degeneration and central necrosis. The kidney damage is of a lesser degree and has been described as fatty degeneration of the epithelium of the convoluted tubules (67) (30). Others have described fatty degeneration of the myocardium and slight tissue damage in the adrenal glands (67) (23).

Animals given sufficient dosage of DDT over long periods of time have been reported to have developed centrolobular necrosis of the liver. This has been a consistant finding (68) (69) (30). Fatty degeneration of the tubular epithelium of the kidney has been present (30). Occasionally, there

may be the definite presence of focal necrosis of cardiac and voluntary muscle (30), but not all investigators have described these changes.

The liver findings are described as a characteristic complex of histological alteration typical of all the chlorinated hydrocarbon insecticides except methoxychlor (66). However, these changes do not occur in non-rodent animals.

Lehman (66) describes the complex as consisting of centrolobular hepatic cell enlargement, with increased oxyphilia, peripheral margination of basophilic granules, and (with formalin fixation) a tendency to hyalinization of the remainder of the cytoplasm. Qualitatively, it parallels an increase in liver weight. At levels such as 600 ppm. DDT the histologic changes are outstanding and unmistakable, but at low levels the minimal changes present require experience and careful comparison with controls for detection. At higher levels of feeding, non-characteristic changes such as focal necrosis, fatty degeneration, etc. may be mixed with characteristic ones.

Several English observers (70) (71) are not so certain that the characteristic liver changes are meaningful. Barnes questions the staining methods (70). Cameron and Chang (71) were not able to demonstrate these changes in the rats used in their experiments.

Various lesions involving the central nervous system have been reported. Vacuolization around large nerve cells in the spinal cord (69) (32), vacuolization around the cerebral motor nuclei (72), and lesions in the roof and dentate nuclei of the cerebellum have been described as oc-

curring in animals poisoned by DDT. The occurrence of the lesions in the roof and dentate nuclei could not be confirmed by other investigators (73). They were described as having been seen in dogs given from 50 to 360 mg./kg. DDT at irregular intervals over periods of time varying from 18 to 100 days. The findings in dogs that died from acute DDT poisoning within 24 to 48 hours following the administration of the compound did not vary from these in untreated control animals (73).

Non-specific effects from inanition may also occur. There may be atrophy of various organs and emaciation in animals chronically poisoned with DDT (18).

Symptoms of DDT Intoxication: In acute poisoning from DDT, the animal may show as a first symptom twitching of the eyelids (25). Generalized tremors may set in which are coarse and begin in the muscles of the head and neck. The tremors have an orderly caudad progression invading the entire musculature of the body with increasing intensity. The tremors are especially severe in the extremities. Convulsions, similar to those seen in strychnine poisoning, may be elicited by irritation and mechanical stimuli such as noise or jarring the animal. Death may ensue from respiratory failure. Ventricular fibrillation induced by the sensitization of the cardiac muscle by DDT to epinephrine may occur at times causing death from heart-failure (74) (75). Pulmonary edema, which is a common finding in poisoning due to solvents, may occur but is not characteristic of DDT intoxication.

Following prolonged absorption of DDT, animals may show a loss of body

weight, anorexia, mild anemia, muscular weakness, tremors that terminate in convulsive seizures, coma, and death (1) (76) (18).

It must be remembered that solvents, particularly kerosene, may produce symptoms which simulate those seen in DDT intoxication. Nervousness, loss of equilibrium, nausea, and vomiting may be caused by an overexposure to some solvents. Headache, fatigue, numbness, and tingling sensations of the lips are characteristic of poisoning due to solvents (1) (76).

Lehman (77) has interpreted the results of animal experiments in terms of human toxicology. He states that dry DDT is quite innocuous when applied upon the skin. However, he warns that solutions of DDT are absorbed and multiple contact constitutes a hazard. He estimated that daily contact of the skin with solutions representing 9 grams of DDT may be dangerous to man. The predominant symptoms of serious or fatal intoxication that one would expect to find in man are giddiness, nervous tension, involuntary muscular tremors, convulsions, depression, and respiratory failure. He estimated the lethal dose in man to be about 30 grams. He states that the onset of symptoms of poisoning after the ingestion of DDT may be delayed for several hours but may appear in one hour. Convulsive seizures are manifest in 30 to 40 minutes after the onset of tremors, and death may occur 2 to 24 hours after the onset of tremors.

The Committee on Pesticides of the Council on Pharmacy and Chemistry of the American Medical Association tabulated and reviewed 384 cases of DDT poisoning recorded in the available medical periodicals or on file with the health departments of the various states and large municipalities (18).

No case of dermatitis, conjunctivitis, hypersensitivity or fatality was reported in the industrial grouping. In the agricultural grouping, there were reported one case of dermatitis, one of conjunctivitis, one systemic intoxication, but no case of hypersensitivity or fatality. Under the miscellaneous groupings, there were 229 instances of accidental ingestion of powdered DDT that produced systemic intoxication. There was one case of intentional ingestion that resulted in a fatality and one case that caused systemic intoxication. The above cases refer only to those exposed to powdered DDT.

DDT in solution produced 38 cases of dermatitis, one case of hypersensitivity, 3 systemic intoxications but no case of hypersensitivity or fatality among the agricultural group. In the industrial grouping, one case each of conjunctivitis, hypersensitivity, systemic intoxication, and fatality was reported. Among the miscellaneous grouping, 7 cases of dermatitis, one case of hypersensitivity, 29 cases of systemic intoxication, of which 15 were due to accidental and 5 to intentional ingestion, and 6 fatalities, of which 5 were the result of accidental and one of intentional ingestion, were reported.

In the situations where the formulations were not specified the following cases were reported: Agricultural grouping; 5 cases of dermatitis, one of conjunctivitis, one systemic intoxication, and one fatality. The industrial grouping revealed that there were 6 cases of dermatitis, 2 of conjunctivitis, one systemic intoxication, but no case of hypersensitivity or fatality. In the miscellaneous grouping, there was reported

one case of hypersensitivity, 25 cases of systemic intoxication, of which 10 were due to accidental and 13 to intentional ingestion, and 5 fatalities, of which 4 were due to accidental and one to intentional ingestion.

Of the 384 cases summarized above, 66 cases occurred among the agricultural group, 13 cases in the industrial group, and the rest occurred in the miscellaneous group, the household and garden group accounting for 20 cases. Two hundred sixty-three cases were due to accidental ingestion, and 22 cases to intentional ingestion.

CHAPTER III

BENZENE HEXACHLORIDE AND ITS IMPORTANT ISOMERS

Benzene hexachloride has been known since 1825 when it was first synthesized by Michael Faraday (78). The first inkling that benzene hexachloride might have commercial possibilities came from an American chemist, Harry Bender (79), who mentioned in 1933 that it appeared to be a good insecticide. This thought was developed further by A.P.W. Dupire, who obtained a French patent in 1941 for the use of this compound as an insecticide (79). Working independently of the French chemists, F.D. Leicester in 1942 substituted benzene hexachloride in flea beetle powder in place of derris and found it effective (80). A year later, another English chemist, F.J.D. Thomas, discovered that the chief insecticidal principle of benzene hexachloride was its gamma isomer (80).

Four isomers of benzene hexachloride had been isolated long before the insecticidal properties of this compound had become known. Meunier (80) showed that benzene hexachloride had two isomers. F.E. Mathews (80) studied the alpha and beta isomers as far back as 1891. T. von der Linden (7), a German chemist, described the gamma and delta isomers of benzene hexachloride in 1912. Two more isomers have been isolated recently but of these only the epsilon isomer has had any study of its insecticidal properties (81). It should be mentioned that the various isomers were named by assigning to them the letters of the Greek alphabet in the order

of their isolation. These designations have nothing to do with the chemical descriptions of these compounds.

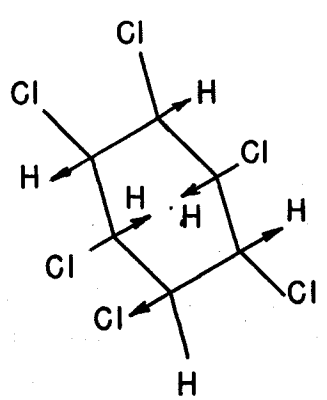
In 1949, it was decided to call the gamma isomer, lindane, in honor of von der Linden who first described it (7).

Benzene hexachloride has several aliases. It is referred to as BHC, gammexane, gamma isomer, 666, and 1, 2, 3, 4, 5, 6,-hexachlorocyclohexane. This last name is the correct chemical one, benzene hexachloride being a misnomer.

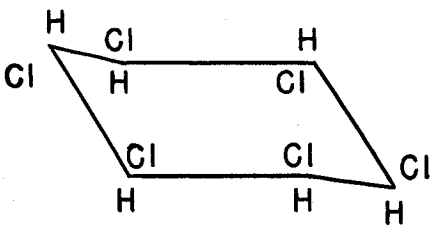
A. Physical and Chemical Properties: Technical benzene hexachloride is a buff-colored crystal which has a musty odor attributed to impurities and to breakdown products (80). This product is a mixture of optical isomers of which the gamma isomer is the compound of interest as an insecticide.

The empirical formula for benzene hexachloride is $C_6H_6Cl_6$. The correct chemical name is 1,2,3,4,5,6-hexachlorocyclohexane. (This compound should not be confused with hexachlorobenzene, C_6Cl_6, which is prepared by the chlorination of benzene in the absence of light.)

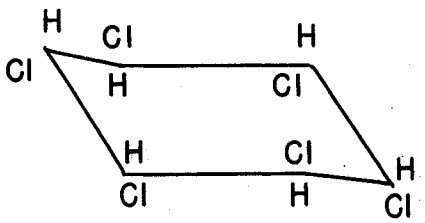
The structural formula for the gamma isomer has been written in several different ways. If the cyclohexane ring is considered to lay in a flat plane, then substitution could be either upwards or downwards from that plane. The structure can be shown as



Slade (80) states that the cyclohexane ring is centrosymmetrical and folded. One of the two sets of free valencies point straight outwards from the ring. The folding in the strainless form has been regarded as a chair with one carbon atom providing the back, another, opposite it, the foot, and the four remaining carbon atoms, the seat (7). The structure may be represented as



According to van Vloten, it is



Despite the fact that the crystal structure as determined by x-ray analyses shows three hydrogen atoms in the outer ring (85), the relative rates of dehydrochlorination seem to agree with Slade's concept of the structure (80).

Preparation: The chlorination of benzene in the presence of ultraviolet light produces "benzene hexachloride" (a mixture of optical isomers), heptachlorocyclohexane (4.0 per cent) and octachlorocyclohexane (0.6 per cent), and possibly some other products. The usual technical preparations contain from 10 to 12 per cent of the gamma isomer, but some contain as much as 42 per cent of this isomer.

To separate the gamma isomer, benzene hexachloride is dissolved in methyl alcohol. Only the gamma and delta isomers are taken up. Upon evaporation, the gamma isomer is the first to crystallize out. Further purification of the gamma isomer is obtained by dissolving it in chloroform and recrystallizing it (83).

The following table summarizes the more important properties of the better known isomers of benzene hexachloride:

Properties of Isomers of BHC (7)

Per cent in BHC	alpha 5	beta 70	gamma 12	delta 7	epsilon 3
M.P. Degrees C.	158	312	112.5	138	219
V.P. mm. Hg (40C) (Slade)	0.06	0.17	0.14	0.09	-
V.P. mm. Hg (20 C) (Slade)	0.02	0.005	0.03	0.02	-
V.P. mm. Hg (20 C) (Balson)	2.5×10^{-5}	2.8×10^{-7}	9.4×10^{-6}	1.7×10^{-5}	-

There is apparent disagreement between the data of Slade (80) and Balson (5) in regard to the vapor pressure of the various isomers. It would appear that the vapor pressures ought to be redetermined. Heyroth (86) has reported other data which would indicate that Balson's figures are too low while those of Slade are some thousand times too high.

Solubility: Benzene hexachloride is readily soluble in methyl-naphthalenes and other aromatic oils. It is only very slightly soluble in water. The following table presents the solubility of benzene hexachloride (the important isomers) in representative organic solvents and

in distilled water. The data are abridged from the table reported by Slade (80).

Solubilities of Isomers of Benzene Hexachloride at 20 Degrees C.

Solvent	Solubility g./100g solution			
	alpha	beta	gamma	delta
Acetone	13.9	10.3	43.5	71.1
Benzene	9.9	1.9	28.9	41.1
Carbon tetrachloride	1.8	0.3	6.7	3.6
Chloroform	6.3	0.3	24.0	13.7
Cyclohexane	17.3	12.1	36.7	49.9
Diethyl carbonate	10.2	4.1	28.4	46.3
Dimethyl acetyl	14.3	3.4	38.7	54.7
Diesel Oil	1.5	0.3	4.1	9.2
Dioxan	33.6	7.8	31.4	58.9
Ethyl alcohol	1.8	1.1	6.4	24.2
Naphtha; heavy (230-270)	5.8	1.5	18.1	30.4
Paraffin (138-212)	1.2	0.05	3.2	4.6
Toluene	9.0	2.1	27.6	41.6
Distilled Water (ppm.)	10.0	5.0	10.0	10.0
White Oil	0.7	0.02	1.9	1.1
Xylene	8.5	3.3	24.7	42.1

Compatibility: Benzene hexachloride is attacked by alkalies. All of the isomers except the beta isomer are destroyed by cold potassium hydroxide. All are destroyed by boiling potassium hydroxide (80). The dehydrochlorination process produces 1,2,4-trichlorobenzene and smaller amounts of 1,2,3, and 1,2,5-trichlorobenzene (7). Otherwise, benzene hexachloride is stable when exposed to light under ordinary conditions of temperature, when subjected to elevated temperatures, and when treated with hot water or concentrated nitric acid (80). In the formulation of insecticide benzene hexachloride may be used with sulfur, pyrophyllite and other inert carriers having a pH below seven (9). Technical benzene hexa-

chloride and lindane are available as dusts, wettable powders, in solutions of organic solvents, and in certain paints (9).

Analytical Methods: There are many methods available for the determination of benzene hexachloride and lindane. None is entirely satisfactory under all conditions. A few of the methods are listed below:

a. Total chlorine method (as for DDT) (87).

b. Spectroscopic method: This method is based on the presence of the absorption band in the ultraviolet range given by 1,2,4-trichlorobenzene obtained by the alkaline dehydrochlorination of BHC (88).

c. Hydrolysable chlorine method: The sample of BHC is treated with N alcoholic potassium hydroxide at 0 C for 50 minutes (dehydrochlorinates alpha, delta, and gamma isomers). A similar sample is treated for 15 minutes (dehydrochlorinates alpha and delta isomers only). By difference, the value for gamma isomer is obtained (89).

d. Cryoscopic methods: Based on the depression of the freezing point of the sample (90).

e. Methyl alcohol solution: Sample is dissolved in methyl alcohol and the solution is evaporated. The gamma isomer is the first to crystallize out (80).

f. Infra-red spectroscopy: (91).

g. Bioassay of insecticidal effectiveness of the sample (92)(103).

B. Toxicity - Experimental Determination

Acute Toxicity: Since technical benzene hexachloride is a mixture of at least six isomers and some by-products of manufacture, it is not surprising that the immediate toxicity of the technical material differs from that of lindane which contains 99 per cent of the gamma isomer. Furthermore, the important isomers of benzene hexachloride vary one from the other with respect to immediate toxicity.

Oral LD_{50} values for technical benzene hexachloride: Because of the variability of the techniques employed, with particular reference to the vehicles used, and probably because of differences in the composition of the commercial product investigated, the LD_{50} dosage for rats has varied considerably at the hands of investigators. Some of the data are tabulated below:

Acute Oral Toxicity of Commercial BHC for Rats

Author	Sample	LD_{50} mg./kg.
1. Slade (83); Taylor and Frodsham (84).	Solvent not stated. 13% gamma isomer.	1250
2. Letard & deSacy (93)	Aqueous suspension	3000
3. Guilhon (94).	Tech., 2 samples.	>5000
4. Heyroth (86).	10% solution in cotton seed oil	940

The oral LD_{50} dose of technical benzene hexachloride for white mice has been reported with only moderate variability, two of the investigators who employed similar techniques having obtained essentially the same results.

Acute Oral Toxicity of Commercial BHC for Mice

Author	Sample	LD_{50} mg./kg.
1. Heyroth (85).	5% suspension in peanut oil. 11.3% gamma isomer	364
2. Vashkov and Serebyakova (95).	apricot oil	approx. 400
3. Furman (97).	50% BHC (12% gamma isomer, 0.75% sod. lauryl sulfate and inert diluent)	> 600 ≅ 72 mg. gamma isomer

Oral LD_{50} of Lindane: Since the gamma isomer is the principle insecticidal component of BHC and is the compound of chief interest, it has been investigated perhaps more thoroughly than the technical BHC.

For ease of comparison, the following table presents the available data dealing with the immediate oral toxicity of the gamma isomer for rats and mice.

Oral LD_{50} Values for Gamma Isomer in Rats and Mice

Reference	LD_{50} in mg./kg.
Rats	
Lehman (100)	125
Woodard and Hagan (99)	177
Tareeva (101)	2000*
Slade (83)	190
Cameron (102)	200
Riemschneider (81)	225

* Used commercial gammexane contaminated with alpha, beta and delta isomers and by-products. (This would amount to a modified technical BHC and these results seem to fit better with those obtained for the technical BHC).

Oral LD_{50} Values for Gamma Isomer in Rats and Mice (continued)

Reference	LD_{50} in mg./kg.
Mice	
Heyroth (86)	84
Woodard and Hagan (99)	86
Furman (97)	--

An opinion has been offered that the other isomers are from 1/6 to 1/60 as toxic as the gamma isomer (99). Several authors have determined the LD_{50} values for the various isomers of BHC administered orally to rats. Their data are tabulated as follows:

Oral LD_{50} Values for Isomers of BHC for Rats
mg./kg. body weight

Isomer	Author		
	Slade (80)	Lehman (100)	Riemschneider (81)
Alpha	1700	500	1500
Beta	--	>6000	--
Gamma	190	125	225
Delta	1000	1000	750
Epsilon	--	--	*

* No harm resulted from feeding 100 mg. epsilon isomer to 150-gram rat.

From the above table it will be seen that Lehman does not agree with Slade or Riemschneider who believe the alpha isomer to be about one-half as toxic as the delta isomer.

There is complete agreement that the gamma isomer is by far the most toxic of the isomers, from the aspect of the immediate response to a single oral dose. Thus, one may suppose that the oral toxicity of a sample of benzene

hexachloride may be determined for the most part by its gamma isomer content. However, when technical benzene hexachloride is administered orally its toxicity is greater than that of the sum of the individual isomers given individually. Heyroth (86) calculated the gamma isomer content of the commercial sample which was being tested and found that the toxic dose was only one-half of what one might expect from its gamma isomer content. He states that the data of Slade (80) and Furman (97) also bear this out. Others (99) found that mixtures of alpha and gamma isomers and delta and gamma isomers were more toxic than one might predict from the individual median lethal doses.

Intravenous Administration of BHC: McNamara and Krop (103) found commercial BHC to be less toxic when administered intravenously to rabbits than one would have estimated from the gamma isomer content. These authors prepared a synthetic mixture of alpha, beta, gamma and delta isomers in the same proportion in which they were present in the commercial BHC. A comparison of the toxicity of the artificial mixture was made with that of each of the isomers injected separately. It was concluded that the synthetic mixture of isomers had an LD_{50} value for rabbits of 150 mg./kg. (approximately 20 mg. gamma isomer).

When the gamma isomer was given alone, 4.5 mg./kg. killed 9 of 11 rabbits. A dose of 6 mg./kg. of gamma isomer was uniformly fatal. The injection of the mixture of isomers which gave 12 mg. of gamma isomer/kg. produced a mortality of 14 per cent. It was also demonstrated that the delta isomer when injected alone 10 to 15 minutes before administering

the gamma isomer reduced the toxicity of the gamma isomer. These results apparently indicate that the toxic action of the gamma isomer is antagonized by some constituent of the mixture.

Furthermore, when either the beta or delta isomer was injected in the amounts ranging from 10 to 20 mg./kg. paralysis developed quickly. This effect is different from that observed when the gamma isomer is administered alone. Dallemagne and Philippot (104) were able to produce convulsions in unanesthetized rabbits with injections of gamma isomer using as little as 1.75 mg. per kilogram.

Dermal Application: Lehman (100) stated that commercial BHC in the dry form produced severe systemic poisoning in rabbits when applied upon the intact skin in a single dose of 780 mg./kg. Furman (97) reported that if the mice in his experiments were prevented from licking the fur after being dipped in solutions containing 5 and 10 per cent of insecticide prepared from commercial BHC 50 per cent wettable powder, the mice exposed to the 5 per cent dip were not harmed while those subjected to the 10 per cent dip died. Only one out of 13 mice survived the higher concentration. On the other hand, if the mice were not restrained from licking the fur, half of the group of mice exposed to a 5 per cent dip died, while the remainder of the group showed toxic symptoms. Other mice were subjected to 2 per cent spray and 1 per cent dip respectively, the mortality in the 2 per cent spray group was 3 deaths in 14 while only one death occurred in the group containing 10 mice that were exposed to the one per cent dip.

Lehman (100) reported that a single application of the gamma isomer

in the amount of 20 to 50 mg./kg. in an unspecified solvent to the skin of rabbits was lethal.

The alpha and beta isomers of BHC produced no signs of local irritation or of toxicity when single doses were applied upon the skin of rabbits. On the other hand, single applications of the gamma and delta isomers were irritating to the skin of rabbits (100).

Horton, Karel and Chadwick (105) applied a 40 per cent solution of lindane in acetone upon the skin of rabbits, guinea pigs, rats, and goats. The following table summarizes their findings.

Mortality Among Various Animals
Following Dermal Application of Lindane

Animal	Dose in mg./kg.	Mortality Ratio
Rabbit	200	0/1
	400	2/2
	1000	2/2
Guinea Pigs and Rats	250	No deaths
	500	No deaths
	1000	25% deaths
Goats	500	0/3
	1000	0/2

Cameron (102) found the LD_{50} range for the cutaneous application of gamma isomer in acetone to be 300 mg./kg. for mice and rabbits; 400 mg./kg. for guinea pigs; and 500 mg./kg. for rats.

Inhalation: There does not seem to be any information regarding the concentration of BHC dusts required to produce acute intoxication by inhalation.

Heyroth (86) reported experiments in which animals were exposed for 7 hours to air saturated with the vapor of the gamma isomer without producing ill-effects. Others (105) were not able to poison rabbits exposed to lindane vapors from impregnated cloth which lined the walls of the cage. The period of exposure was one week.

It appears unlikely that the other isomers of BHC which are less toxic under conditions of acute exposure and which have even lower vapor pressures than the gamma isomer could produce acute intoxication through the inhalation of vapors (86).

Chronic Toxicity: Various authors have reported their findings as to what happens to animals given oral doses of benzene hexachloride spaced over variable periods of time. To simplify comparisons, the data are best presented in tabular form.

Results Reported by Various Authors Following the Administration of Oral BHC

Author	Animal	Preparation	Duration	Results
Slade (80)	Rat	100 mg. mixture of isomers fed daily in diet.	2 mos.	No effect noted.
Taylor and Frodsham (84)	Rat	500 mg. mixture of isomers fed in daily diet.	57 days	Normal growth in immature rats.
Furman (97)	Mice	Pellets of diet supplying 1.5 mg. BHC/day per mouse.	30 days	No harm to 8 or 10 mice. Two died on 21st and 22nd day respectively. Mortality occurred after over 3 times the single dose required

Author	Animal	Preparation	Duration	Results
				to kill 37% of mice was administered in small doses. No evidence of any marked accumulative effect.
Penrod (106)	Rat	1500 ppm. BHC added to daily diet.		Tolerated. Ability to conceive and/or care for young may be impaired but data for conclusion not available.
Dallemagne & Philippot (104)	Dog	100 mg. BHC per kg. in oily solution daily.		Treatment stopped after 19 doses; posterior paresis developed progressing rapidly to paralysis. Killed 3 days later. Slight hepatic degeneration seen at autopsy. After 3rd treatment, convulsive seizures set off by external stimuli; posterior paralysis; death 5 days later.
Woodard & Hagan (99)	Dog	50 mg. BHC per kg. (15.3% gamma isomer) in corn oil given as capsule daily 5 days a week.		The 3 dogs tolerated treatment for 36,46 and 49 days respectively. Some liver damage noted at autopsy.

Fitzhugh, Nelson and Frawley added different amounts of technical BHC, as a 10 per cent solution in corn oil, to the diets of groups of male and female rats for their natural life-span (control group: 58.3 ♂ or - 7.1 weeks). The various groups of rats were fed technical BHC at the levels of 10,50,100, and 800 ppm. respectively. The group fed technical BHC at 800 ppm. lived only 40 weeks. A significant increase in the weight of the liver was

found in rats that were fed at the level of 100 ppm.

Lehman (35) on feeding groups of rats fed technical BHC for 104 weeks at different levels in their diets, found that the highest level tolerated without gross effects was 50 ppm., while 100 ppm. was the lowest level which produced gross effects. The highest level which produced no tissue damage was 10 ppm., but 50 ppm. was the lowest level at which tissue damage was noted.

Apparently this information was cited by Lehman from the experiments of Fitzhugh, Nelson and Frawley (107).

Chronic oral toxicity of lindane: The experiments in which comparisons of the toxicity of the several isomers of BHC are made will be reported later. Only those which deal with the gamma isomer will be presented here. A table comparable to that made for the chronic oral toxicity of BHC appears below.

Chronic Oral Toxicity of Lindane Reported by Various Investigators

Reference	Animal	Preparation	Duration	Results
Slade (80)	Rat	Fed 10,20 or 30 mg. gammexane per day.	5 wks.	No effect
Taylor and Fordsham (84)	Rat	Fed 10,20 or 30 mg. powdered gamma isomer per day.	27 days	No effect
Woodard & Hagan (99)	Dog	10 mg. lindane per kg. given daily except Sundays.		Two of 3 dogs tolerated this amount for 18 and 49 days respectively.

Reference	Animal	Preparation	Duration	Results
Lehman (35)	Dog	10 mg. lindane in corn oil/kg. given daily 5 days a week		Six dogs of 6 died. Earliest death in 14 days, the last in 221 days.
	Dog	15 mg./kg. as above		Seven animals of 7 died. Earliest death in 2 days, the last in 56 days.
Dallemagne & Philippot (104)	Dog	100 mg. lindane aqueous suspension daily		No excitation or paralysis. Refused food on 4th day.

Lehman (35) reported that groups of rats fed lindane at the level of 50 ppm. for 104 weeks showed no tissue damage, while those fed at 100 ppm. had tissue damage. Gross effects were noted at the 100 ppm. level, but not at 50 ppm.

Fitzhugh, Nelson and Frawley (107) observed groups of rats over their natural life-span, while they were being fed on diets containing the gamma isomer as a 10 per cent solution in corn oil at the levels of 5, 10, 50, 100, 400, 800 and 1600 ppm. and as the dry compound at levels of 10, 100, and 800 ppm.

The rats fed on diets containing the gamma isomer in corn oil at the level of 800 ppm. survived only 40 weeks. Symptoms of intoxication, including convulsions, were noted. Those which took the dry gamma isomer in the same manner at the level of 800 ppm. lived as long as the control group. The weight of the livers of this group was significantly increased, while the corresponding change occurred in rats fed gamma isomer in oil

solution at the level of only 100 ppm. The rats given 1000 ppm in corn oil showed symptoms of intoxication, including convulsions, but survived as long as those fed in the corresponding manner at the level of 800 ppm.

Lehman (35) stated that the highest dietary level of gamma isomer which is tolerated by rats for 9 months is probably 800 ppm. Penrod's (106) value of 0.075 per cent agrees closely with Lehman's.

Doisy and Bocklage (108) noted no toxic effect in rats fed on a diet containing the gamma isomer at the level of 200 ppm.

Chronic oral toxicities of the other isomers of BHC: Fitzhugh, Nelson and Holland (109) examined their data from an 80-weeks comparative feeding experiment and concluded that the alpha and gamma isomers have about the same order of toxicity. Lehman (35) has listed some data regarding the chronic toxicity of some of the isomers of BHC for the rat.

Chronic Toxicity for the Rat

Pesticide	Time of feeding in weeks	Lowest level with gross effects	Highest level without gross effects	Lowest level with tissue damage
Alpha	104	100	50	50
Beta	104	10	--	10*
Delta	104	800*	--	800*

* Lowest level fed

The highest level of alpha isomer fed that produced no tissue damage was 10 ppm. There were no other relevant data for beta or delta isomer.

Fitzhugh, Nelson and Frawley (107) added alpha and beta isomers in

10 per cent corn oil at levels of 10, 50, 100, 800 ppm. and 10, 100, 800 ppm. respectively to the diets of groups of rats and observed the rats over the course of their natural life-span. The animals fed on the alpha isomer at the level of 800 ppm. survived 40 weeks. The animals fed on the beta isomer in the same concentration in the diet lived only 4 weeks. Significant increases in the weights of the liver were found in the rats fed on beta isomer at the level of 10 ppm. and on alpha isomer at the level of 50 ppm. respectively. The growth of the rats began to be retarded at the level of 100 ppm. beta and 800 ppm. alpha isomer.

Lehman (35) believes that the highest tolerable level of alpha isomer in the daily diet of rats is 800 ppm. If the beta isomer is fed regularly to rats at the level of 500 ppm., severe poisoning is produced in two weeks.

Percutaneous absorption of BHC: Letard and deSacy (93) painted the shaved backs of 10 mice daily with a 1 per cent solution of BHC for 2 months without inducing any signs of skin irritation. Kirby (110) painted the nose, ears, paws and tail of a group of rats with a 5 per cent solution of BHC in acetone daily except Sundays for 8 weeks. The total amount of insecticide used was 1.7 g. per rat. No cutaneous damage was observed.

Lehman (35) reported fatalities among rabbits given dermal applications of BHC at the dose level of 200 mg./kg./day.

Exposure to Lindane: Heyroth (86) reported the observations made at

Kettering Laboratory on the effect of different concentrations of lindane in different forms on the skin of the rabbit. The findings are tabulated below:

Effects of Chronic Percutaneous Absorption of Lindane in Rabbits

Preparation	Dose	Result
1% solution in anhydrous isopropyl alcohol.	2 ml. portions applied to abdomen at 30 min. intervals over 3 hr. period. Done 5 days a week.	Death on 4th, 7th, 10th days of treatment. No evidence of cutaneous irritation.
1% solution in 91% aqueous isopropyl alcohol.	As above	Death on 5th and 10th days of treatment. No cutaneous irritation.
1% solution in anhydrous isopropyl alcohol.	2 ml./kg. applied at one time and allowed to remain 4 hrs. Repeated on successive days.	No deaths in 39 periods of exposure. No cutaneous irritation.
1% solution in 91% aqueous isopropyl alcohol.	As above	As above
1% gamma isomer dry powder.	2 g./kg. applied to abdominal skin for period of 4 hrs. a day.	1 rabbit died after 15 periods of exposure. Others survived 39 periods. No cutaneous irritation.
10% gamma isomer dusting powder.	2 g./kg. applied to abdominal skin for period of 4 hrs. daily.	Severe clonic and tonic convulsions in 1 rabbit. Tremors and weakness in other 2 rabbits after 2 periods of exposure. Symptoms abated as treatment continued for 30 days. No cutaneous damage.
Ointment - 1% gamma isomer	2 g./kg. for 4 hrs. daily.	Applied 31 days. No signs or symptoms of intoxication or local damage.
Ointment - 10% gamma isomer	0.2g./kg. for 4 hrs. a day.	Applied 28 days. Only gross observation was retardation of gain in body weight as compared to normal controls.

Histopathologic examination of the tissues of the rabbits exposed to the solutions made up with isopropyl alcohols revealed a moderately severe parenchymatous degeneration of the liver in all animals and moderate damage to the distal convoluted tubules of the kidneys of some.

When the dose was reduced to 2 ml./kg. the histopathologic changes were those of slight parenchymatous degeneration in the liver and focal subendocardial hemorrhages.

When the dry powder containing 1 per cent of the gamma isomer was applied, the microscopic tissue changes were reported as showing slight damage to the parenchyma in the central zone of the liver lobules. When the 10 per cent dusting powder was used, moderate parenchymatous degeneration of the liver was noted.

In animals subjected to applications of the 1 per cent ointment, the histologic changes, when present, occurred only in the liver and consisted of parenchymal degeneration varying from slight to severe.

Some of the rabbits subjected to contact with the 10 per cent ointment gave evidence of toxic degenerative changes in the liver.

Chronic Inhalation of Lindane: Heyroth (86) reported that one cat, 2 guinea pigs, 3 rabbits and 3 rats were exposed to air saturated with lindane vapor (0.6 mg./cu. meter of air) for 7 hours on each of 5 days per week for 171 days. No sign of illness was noted. Heyroth made the rough estimate that a full-grown rat breathes 150 cc air/min., equivalent in 7 hours to 63 liters of air. This amount of air saturated with lindane vapor contains only 0.04 mg. lindane. In a 24-hour period, the value would

be about 0.14 mg. lindane. This is approximately the amount that might be absorbed from 15 grams of food containing 10 ppm. (0.15 milligram).

Experiments with large animals: Bushland, Wells and Radeleff (39) applied 1.5 per cent (by weight) technical BHC prepared from the wettable powder containing 10 per cent of gamma isomer to cattle and hogs as sprays, to sheep and goats as dips, and to horses by sponge. The applications were repeated at 4-day intervals until 8 had been given. The animals were observed for 30 days following the 8th application. All of the animals survived the course of treatment and appeared to remain in good health.

A second experiment was performed applying BHC (12% gamma isomer) emulsion concentrate to the animals in the manner described above. All of the animals survived without apparent ill-effect.

W.C. McDuffie (39) treated cows with different concentrations of a wettable powder composed of the gamma isomer. A 1.5 per cent preparation killed 3 cows of a group of 3 in 24 hours. The concentration of 0.75 per cent of the same material affected 2 of 3 cows, one of which died but the other animal recovered.

Eddy and Graham (39) used a concentration of 0.5 per cent of the gamma isomer to prepare sheep dips from a suspension of wettable powder and to prepare an emulsion. The animals were unaffected by these dips.

BHC (10-12% gamma isomer) has been tested on a wide scale, the heaviest concentration used being 2.5 per cent. Sheep dipped in this formulation were not harmed.

Welch (38) administered different concentrations of BHC, prepared from the 50 per cent wettable powder, to cattle and sheep by stomach tube. He found that a single 2-gram dose administered to sheep produced severe symptoms of muscular spasm and blindness in 6 hours, but the sheep returned to normal by the 5th day. Single doses of one gram and 0.75 gram respectively caused only slight nervous symptoms at 24 hours, but the animals soon returned to normal.

A steer survived a dose of BHC suspension given by stomach tube in the amount of 0.125 gm./kg. without showing any effect.

Each of a number of sheep was given a capsule containing 2.25 grams BHC each morning until 60 doses had been given. After the 20th day, the sheep showed slight nervousness and incoordination, which disappeared and recurred from time to time during the experimental period (39).

Wilson (114) prepared doses of 0.3 g./kg. (39 mg. gamma isomer /kg.) from BHC powder containing 13 per cent of the gamma isomer, which was administered to calves. He found that this dose was toxic to calves but that older animals were more resistant.

Furman (97) administered doses of 50 per cent wettable BHC in capsules at rates of 0.05 gm./kg. and 0.074 gm./kg. respectively to cows. No ill effects were noted over a period of observation lasting 10 days.

Seven range heifers and one bull were sprayed with 0.32 per cent BHC aqueous suspension and showed no ill effects (40).

A total of 442 yearling range cattle were dipped in aqueous suspensions of 0.05 per cent BHC. No ill effects were noted over a 3-week period of

observation (40).

Most farm animals are resistant to poisoning by gamma BHC. Adult cattle and horses have withstood sprays and dips containing 0.2 per cent of the gamma isomer. Sheep, goats and hogs have withstood 0.5 per cent. Calves are not endangered, apparently, when treated with dips and sprays containing only 0.03 per cent.

Symptoms of benzene hexachloride intoxication: Furman (97) described the signs and symptoms of BHC intoxication in mice. He stated that the symptoms of poisoning were similar to those reported by Cameron and Burgess (32) and Guilhon (94). The body trembles and the mouse sneezes frequently, pawing at its nose. Breathing may become labored. The animal becomes excitable, jumping with convulsive spasms. Staggering becomes pronounced, gross incoordination becoming increasingly evident. A partial paralysis may be observed, particularly in the front legs. In the terminal stages of fatal poisoning the mouse is unable to stand, but may lie prostrate for several hours in convulsive spasms.

Radeleff and Bushland (21) state that the symptoms and lesions produced in farm animals by all the chlorinated hydrocarbon insecticides which they tested are the same for all practical purposes. They described two distinct phases. They may occur singly or follow one another. One phase is that of nervous stimulation. There is excessive salivation, grinding of the teeth, rolling of the eyes, muscular twitching, hyperexcitability and finally, convulsions. The other phase is one of nerve depression. The animals are dull, listless, fail to eat or drink, and appear blind.

In all instances of poisoning by BHC, symptoms occurred in less than 48 hours after exposure. Some animals were affected in as little as 2 hours, but most animals become affected after about 6 hours.

Death, regardless of the symptoms preceding it, is one of agony. The body assumes a position of opisthotonos. At time of death the body temperature usually is in excess of 110 degrees F.

Dallemagne and Philippot (104) describe the development of posterior paresis in the dog which rapidly progresses to paralysis. There may be convulsive crises induced by external stimuli.

In summary, BHC and lindane intoxication are characterized by nervous irritability, weakness, tremors, incoordination and convulsions.

Pathologic findings: Dallemagne and Philippot (104) (17), described a mild form of hepatic degeneration in dogs following the oral administration of 100 mg./kg. BHC in oily solution over a period of 19 days. Woodard and Hagan (99) also noted marked liver damage, moderate kidney damage, atrophy of various tissues, and lesser injury to other viscera in dogs following the daily oral administration of 50 mg. BHC (15.3% gamma isomer)/kg. over periods of from 36 to 49 days. Furman (97) noted hemorrhagic gastritis and duodenitis after the oral administration of BHC to mice. Woodard and Hagan (99) noted mild changes in the liver, bone marrow, lymphoid tissue, adrenal cortex and cerebrum in dogs fed 10 mg. gamma isomer/kg. daily except Sundays for from 18 to 49 days.

Kirby (110) reported cloudy swelling of the liver and kidney, some animals showing inflammation of the glandular stomach and degeneration of

the adrenals in 2 instances following the dermal application of 5% solution "666" in acetone to rats for three weeks.

Heyroth (86) reported that the microscopic examination of the tissues from mice given BHC orally revealed toxic degeneration in the viscera. The liver showed diffuse parenchymatous degeneration and similar changes were noted in the renal convoluted tubules.

Radeleff and Bushland (115) reported that the carcasses of poisoned calves appeared cyanotic, and hemorrhages were commonly observed along the gastro-intestinal tract and in the heart and lungs. The brain and spinal cord were congested and often edematous.

Fitzhugh, Nelson and Holland (109) noted that at 100 ppm. in the diet the isomers of BHC produced slight histologic changes in the liver. The alpha and gamma isomers also damaged the kidney at this level of feeding. The beta isomer caused the most damage to the liver, and even when fed at 10 ppm. it produced a questionable degree of liver damage. The alpha and gamma isomers, when fed at 10 ppm., failed to injure the liver or kidney.

Lehman (66) states that the livers of rats show more or less of the characteristic complex of histological alterations, which are described as consisting of centrolobular hepatic cell enlargement, with increased oxyphilia, peripheral margination of basophilic granules and (with formalin fixation) a tendency to hyalinization of the remainder of the cytoplasm. These changes are found in the livers of rodents and apparently not in those of rabbits or dogs.

The other organs are relatively uninvolved. High levels of alpha

isomer, lindane, and technical BHC caused slight or moderate increases in the amount of focal nephritis, and technical BHC caused a moderate degree of testicular atrophy.

Metabolism, Storage, Detoxification and Excretion: Wilson (114) found that the blood of cattle becomes insecticidally active and capable of killing tsetse flies and ticks when 0.25 g. BHC/kg. was administered to the cattle on each of 2 days and 0.125 g. was given on each of the succeeding two days.

Woodard, Davidow and Lehman (49) studied the urine of rabbits following the oral administration or the intravenous injection of the alpha isomer. The urinary extracts were separated into the neutral, phenolic and acidic fractions and studied. The findings are tabulated below:

	Fraction	Results
1.	Neutral	No unchanged material was found but in 2 of 8 rabbits an ultraviolet absorption curve like that of 1,2,4-trichlorobenzene was obtained.
2.	Phenolic	No 2,4,5-trichlorobenzene was found (might be expected to be formed by oxidation of 1,2,4-trichlorobenzene.)
3.	Acidic	Characteristic curves were obtained in ultraviolet but variable ones in infrared.

They concluded that these results suggest that the metabolism of the alpha isomer is highly variable.

Laug (118) found no gamma isomer in the tissues of rats given any one of the other isomers alone. This suggested that no stereochemical change to the gamma isomer occurs in vivo.

The experiments of Doisy and Bocklage (108) indicate that the addition of inositol to the diets of rats does not alleviate the toxic symptoms due to the gamma isomer.

Lehman (35) has stated that the gamma isomer is stored in the tissues at a lower level than that of DDT when these are fed at the same levels, and the storage of this isomer does not appear to be progressively cumulative with time, and that which has been stored disappears soon after the isomer is removed from the diet. On the other hand, the gamma isomer is found consistently in the brain of the animal while it is being fed on the isomer. In contrast to this finding, DDT does not appear in the brains of animals maintained on diets contaminated with DDT.

The storage of the various isomers in the fat of rats is indicated in the following table (100):

Storage of Isomers of BHC in Fat of the Rat

Isomer	Dietary Level 500 ppm. 12 weeks' feeding	100 ppm. 2 weeks' feeding
	ppm. in fat	ppm. in fat
Alpha	2400	40-100
Beta	7500	300-500
Gamma	400	--
Delta	500	--

Lehman (100) found that beta BHC is stored in the fat in concentrations greater than that in the diet and that the storage increases with time until a maximum is reached. The following table presents data obtained in long term experiments.

Storage of Lindane and Beta Isomer in
Fat of Rats and Dogs as Determined by Chemical Methods

Insecticide	Feeding level ppm.	Duration	Average Storage in ppm. Male	Female
		RAT		
Beta BHC	100	2 years	---	2000
	100	2 weeks	570	360
	100	6 weeks	1120	1460
	100	12 weeks	1300	2770
Lindane	100	2 years	90	120
	100	2 weeks	10	10
	100	6 weeks	90	70
	100	12 weeks	50	100
		DOG		
Beta BHC	about 1000	19-20 wks.		8825
Lindane	about 400	27 weeks		466

It is of interest to know the rate of disappearance of stored insecticide from the fat of experimental animals. The following data are derived from the Food and Drug Administration experiments (60).

Rate of Disappearance of Stored Insecticide from Fat of Rats

Insecticide	Initial Conc. ppm. Male	Female	Time on Insecticide-free diet weeks	Conc. in ppm. at end of observation period Male	Female
Beta BHC	675	1010	2	337	790
			4	135	450
			6	120	260
			8	13	120
Lindane	102	102	1	0	0

Laug (118) (119) collected the urine and feces of rats each 24 hours during the period of one month while they were being fed on diets containing 20 or 500 ppm. of gamma isomer. He found that from 0 to 4 per cent of the amount ingested appeared in the urine of the males but not in that of the castrated female rats. Only a very small amount of the gamma isomer was ever found in the feces. This finding seemed to indicate either that complete absorption occurred or that the isomer was destroyed in the gastro-intestinal tract.

The gamma isomer was found in the blood, liver, spleen, adrenal, muscle, brain, kidney and fat of rats fed 500 ppm. gamma isomer (120). Laug further states that a preliminary investigation showed that approximately 25 per cent of the gamma isomer disappeared when it was incubated at 38 degrees for two hours in contact with minced liver. This finding was interpreted to mean that the liver is particularly efficient in metabolizing the gamma isomer.

Other experiments have been done on large animals which indicate that storage of BHC does occur in the fat. Bushland, Claborn, Beckman, Radeleff, and Wells (121) reported that when sheep were dipped in 1.5% benzene hexachloride (12% gamma) emulsion on 8 occasions at 4-day intervals, the insecticide found in the fat after 1 month and 7 months respectively was 352 ppm. and 32 ppm.

In another experiment, Hereford cattle were sprayed 12 times at biweekly intervals with emulsions containing 0.025% gamma BHC (mixed isomers containing 12% gamma). The fat of these animals showed 26 ppm. of organic chlorine

2 weeks after the 12th spraying. Four weeks later the average was 11, and 10 weeks after the sprayings were completed the average was 4 ppm.

Cattle were fed lindane mixed with the feed at levels of 1, 10, and 100 ppm. respectively. The following results were obtained: (121)

Length of feeding period	Dietary level		
	1 ppm.	10 ppm.	100 ppm.
	ppm. in the fat		
2 weeks			65
10 weeks	2	8	100
2 weeks after feeding stopped	2	4	50

Barns were sprayed with lindane to give a calculated deposit of 25 mg./sq.ft. of surface. The spray material was made up from 25% lindane wettable powder (8 lbs. per 100 gal. water) and 20% lindane emulsion concentrate (5 qts. per 100 gals. water) and sprayed at a pressure of 100 lbs./sq. inch. Composite milk samples were taken at 1,3,7 and 14 days after the barns were sprayed. None of the analyses of the milk showed the presence of lindane (analytic method of Frawley and Davidow (122) (123)).

Dairy cows sprayed thoroughly on one occasion with 0.046% lindane had lindane in the milk to the extent of 1.6 ppm. on the 1st day, 0.6 ppm. on the 2nd, 0.3 ppm. on the 3rd day. On the 5th and 7th days the milk contained less than 0.2 ppm., which is the limit of accuracy of the test method. Other animals were sprayed with 0.03% lindane and in addition were given 100 mg. of lindane in their feed. Analyses of the milk on the first day gave a result of 0.6 ppm. and by the 3rd day no insecticide was found. Cows sprayed with 0.05% lindane had 1.0 ppm. in the milk on the first day,

0.3 ppm. on the 2nd, 0.4 ppm. on the 3rd day but by the 5th and 7th days the amount of insecticide determined was within the limit of accuracy of the method. Still other animals were treated with 0.1% lindane spray. On the first day it was reported that the milk contained 2.0 ppm., on the 2nd day, 0.6 ppm., on the 3rd day, 0.4 ppm., on the 5th day, 0.2 ppm., and on the 7th day, less than 0.1 ppm. No unpleasant odor or flavor was detected which could be attributed to lindane. Over 100,000 cows have been treated in this way in New York State. The treatment produced no harmful effects on the animals or in the quality of the milk (123).

Furman (98) reported that lactating cows treated with a drenching spray of 0.5 per cent of benzene hexachloride in aqueous suspension, or fed 40 mg. technical BHC/kg., yielded a maximum of 5.5 ppm. of gamma isomer in the cream on the day following the treatment. Smaller amounts were recovered in the cream for as long as 9 days thereafter in one instance (fly assay method). He also stated that the cream had a musty odor.

Davidow and Frawley (120) found that alpha, beta, gamma and delta isomers accumulate mainly in the adipose tissue of both dogs and rats. Some quantity of all of the isomers occurs in the kidneys, brain, liver and muscle tissue of rats. In dogs the alpha, beta and gamma isomers are found in the adrenals. The chronic toxicity of each isomer appears to have a direct relationship to storage in adipose tissue. The beta isomer persisted in the fat depots of rats for 14 weeks after the administration of BHC had been discontinued in the diet whereas the alpha, gamma and delta isomers had disappeared in three weeks.

Copor, Kerken and Klampau (124) studied the antagonistic and synergistic reactions of the beta and gamma isomers. They administered single doses of BHC and found that the irritation of the central nervous system, which would result following the administration of Cardiozol (pentamethylenetetrazol), was delayed or prevented. Other experiments were done in which it was determined that the intraperitoneal LD_{50} dose of the gamma isomer dissolved in rape oil was 75 mg./kg. for the rat. To test the reciprocal action by the isomers, the beta isomer was given intraperitoneally and 7 days later a dose of "gammexane" at least equal to that which would produce 50% mortality was injected. All of the animals remained alive and without symptoms. Using Cardiozol as an indicator, the inhibitory effect of "gammexane" on the irritation of the central nervous system was tested. Rats were given 80 mg./kg. of Cardiozol subcutaneously, a dose which normally causes convulsions. A dose of beta isomer in rape oil was given from 5-7 days before the gamma isomer. All of the animals survived, and only a few of them had convulsions. The results showed that "gammexane" was effective also after the previous administration of the beta isomer. The administration of the beta and gamma components simultaneously by mouth, showed synergism in preventing the Cardiozol convulsions in doses which of either alone could not have had that effect. These authors comment that almost nothing is known of the mechanism of action. No doubt the chlorinated cyclohexanes are altered in the organism.

Several authors believe that the convulsions induced by lindane intoxication are primarily of high central origin. Dallemagne and Philippot (125)(104)

found that dogs, in which the spinal cord had been cut, exhibited spasms in muscles innervated from above the cut, but not in those supplied by nerves that emerge caudad to the cut. The spasms are limited to the face in animals with the cord severed in the superior cervical region. In intact animals, they give place to flaccid paralysis in the posterior limbs after the lumbar injection of a spinal anesthetic. These same investigators (126) found that the content of potassium, but not that of calcium (104), was increased promptly with the onset of convulsions to 20-25 mg./100 ml. This was attributed to the effect of oxygen-deficiency upon the liver. In later work, with hepatectomized and eviscerated dogs, they found more potassium in these animals than in others with the liver intact. It is now believed that the source of the potassium is in the muscles.

McNamara and Krop (16) studied the electroencephalographic tracing from currarized dogs given doses of 4 mg. of gamma isomer/kg. intravenously. Within 30 seconds there appeared a marked increase in the potential and frequency of the brain-waves as in an epileptic attack. Although the heart-rate was greatly reduced, an increase in the blood pressure to well above 200 mm. Hg. occurred. The convulsions ended abruptly after 30 seconds, the blood pressure began to fall, and a normal heart rate was re-established. The blood pressure did not reach the preconvulsive level before the onset of another episode. In some dogs, the convulsive seizures were repeated more than 10 times. The characteristic changes in the EEG tracings elicited by lindane could be antagonized by the prior administration of a dose of the delta isomer, which given alone, produced central depression.

The uptake of oxygen by slices of the brains of rats given 100 mg. gamma isomer/kg. intraperitoneally was found to occur at a normal rate (127). When the entire brain of normal rats was homogenized with 0.05 ml. of 10 per cent solution of gamma isomer for gram of brain, the rate of uptake of oxygen was also unaffected.

Observations of the effects of BHC and its isomers in Man: No cases of serious or fatal human poisoning resulting from contact with benzene hexachloride have been reported. However, complaints of headache, irritation of the eyes and nasal passages, and a transient smarting of the hands and neck have been reported by men working in the field using the dusts (128) (129). Haller (129) states that there are reports that benzene hexachloride, when used as a dust in field applications, has caused nausea, headache, and irritation of the skin, eyes, nose and throat of some people exposed to it. An instance has been cited where workmen refused to apply dusts containing benzene hexachloride.

Tareeva (101) is said to have made 90-minute patch tests with doses of 200 and 500 mg. of impure gammexane in acetone. The only reaction reported was a slight transitory hyperemia.

Cannon and McRae (130) stated that L.W. Smith informed them that in 500 human patch tests lindane produced no instance of sensitivity. Several of these were said to have been repeated tests.

Heyroth (86) quoted Doctor J.G. Sanders of Commercial Solvents Corporation, as stating that in one manufacturing plant 3 or 4 men worked for 2 years or more without illness in filling drums by shovel with the finished

flaked product which contained 13 to 15 per cent of the gamma isomer. They worked in a fairly well ventilated medium sized room with a 15 foot ceiling and got considerably quantities of the dust in their hair, clothing and shoes. They were examined monthly.

Francone and Chena (131) reported typical contact dermatitis among workers manufacturing BHC. The dermatitis may appear within 10 days of starting work. In the majority of cases, the attack is mild, clears up rapidly, and does not recur. These authors reported 39 cases.

BHC has been used repeatedly on human subjects as an insecticide with no injurious effect (133). The armed forces of the United States have used lindane in Korea to destroy the body louse, which has become resistent to DDT (132). No report of any adverse effects has been published.

BHC and the gamma isomer have been used repeatedly as scabicides in the practice of dermatology. Wooldridge (133) used a 1% ointment in treating 33 patients with scabies. No irritation was noted. Only one patient developed a severe eczematoid reaction which was attributed to the perfume used in the ointment.

Petry (134) reported a case of poisoning believed to have been due to the inhalation of gamma BHC by a 44 year old healthy male baker who used 200-250 grams of a proprietary powder containing about 3% of the gamma isomer for dusting the bakery. Approximately 15 minutes after dusting, the patient had severe difficulty in breathing. The physician who saw him reported that the patient had severe cardiac and cerebral disturbances. The odor of the insect powder was detected on the breath of the patient. A day

later, he developed severe and frequent vomiting and motor weakness of the entire left side of the body with apesthesia of left arm and both legs. The symptoms gradually receded but even after three weeks there was involvement of the nervous system and the myocardium.

It was suggested that gamma BHC might prove to be much more toxic than has been shown to be the case hitherto if it were administered by inhalation.

Dallemagne and Philippot (117) stated that Dr. Malter would report the clinical observations on workers engaged in the manufacture of hexachlorocyclohexane who had developed organic nervous and mental symptoms, accompanied by anaphylactic manifestations. They commented, "Of course, it is possible that these pathologic disturbances are not due to hexachlorocyclohexane itself, but to intermediate products of its manufacture."

BHC has been administered as a vermifuge in the treatment of oxyuriasis. Barnes quoted Lendle and Schneider (135) (170) as recommending a dose of 45 mg. for 2 to 3 successive days. Klosa (136) stated that single doses of 800 to 1200 mg. have been taken without ill effect, but daily doses of 70 to 90 mg. taken for 10 to 14 days may produce diarrhea and dizziness. Graeve and Herrnring (137) believe that some individuals may be very susceptible in that two patients receiving treatment of oxyuriasis developed convulsions. Dr. C.C. Chesterman stated, in discussing the paper of J.M. Barnes (70), that he recently treated two adults for oxyuriasis with 50 mg. doses of BHC in gelatine capsules three times daily for 2 days without any

dietetic restrictions except that alcohol was proscribed. One of the patients was not cured by the one course. The other female patient was given 100 mg. thrice daily for 2 days, but shortly after taking the final dose had an epileptiform convulsion. She recovered completely. He commented that there is a very small margin, if any, between the safe and the effective dose of BHC when administered orally under these conditions.

CHAPTER IV

TOXAPHENE

Physical and Chemical Properties: Toxaphene is the common name for the chlorinated camphene compound which was formerly called Hercules 3956, an insecticide produced by The Hercules Powder Company.

Chemically, this insecticide is a polychlor bicyclic terpene containing 67-69 per cent of chlorine, with the average empirical formula of $C_{10}H_{10}Cl_8$. The structural formula has not been established (138).

The technical grade of toxaphene may be described as a cream-colored, waxy solid with a mild pine odor. Its specific gravity is 1.66 at 27 degrees C. Its melting range lies between 65-95 degrees C. (138). Toxaphene may be considered almost nonvolatile, losing about 0.1 per cent in weight when it is heated to 100 degrees C. for twenty hours (138).

Toxaphene is insoluble in water but it dissolves readily in organic solvents and oils. The following table cited by Parker and Beacher (139) gives the solubility of toxaphene in representative solvents.

SOLVENT	Grams/100 ml. at 27 degrees C.
Acetone	over 450
Benzene	over 450
Carbon tetrachloride	over 450
Ethylene dichloride	over 450
Toluene	over 450
Xylene	over 450
Hexane	over 450

SOLVENT	Grams/100 ml. at 27 degrees C. - continued
Turpentine	350-400
Kerosene	over 280
Fuel Oil	250-275
Deodorized kerosene	over 280
Thanite	100-120
Lube Oil	70- 80
White Oil 50	55- 60
Isopropanol	15- 18
Ethanol - 95%	10- 13
Water	Insoluble

Toxaphene is stable either alone or in solution, under ordinary conditions. When the material is heated or exposed to ultraviolet light, it is very slowly dehydrochlorinated, yielding about ten parts per million of hydrochloric acid an hour at 300 degrees C. (138). The presence of strong alkali or iron compounds hastens the dehydrochlorination process. The oil solutions may be stored safely at room temperature in clear glass bottles or in iron containers without appreciable deterioration (7). However, special precautions are needed to store concentrated solutions of toxaphene (138).

Compatibility: Toxaphene is compatible with many of the common insecticides and agricultural chemicals which include Thanite, pyrethrum, rotenone, DDT, lead and calcium arsenates, nicotine sulfate, Copper Compound A, Yellow Cuprocide, Bordeaux mixtures, sulfur, and neutral emulsifying and wetting agents (9). Because of the presence of a very labile chlorine atom in toxaphene, it is attacked by bases. Strongly alkaline materials should not be used in formulations of toxaphene (9).

Analytical Methods: At present, there is no suitable microanalytic

chemical method available for the specific identification of minute quantities of toxaphene in tissues or body fluids. However, toxaphene residues may be determined by the total chlorine method using the nitrobenzene modification of the Volhard titration technic (7) (140). It has been suggested that since there is one labile chlorine per molecule, the hydrolysable chlorine method might also be useful for analytical purposes (7). A fly bioassay method has been used (141).

The Toxicity of Toxaphene.

Oral LD_{50} Values for Rats, Guinea Pigs, and Dogs: Shelanski and Gellhorn, as quoted by McGee et al (142), found the oral LD_{50} doses of toxaphene in various vehicles and various concentrations of the insecticide for white rats and guinea pigs to be as follows:

Oral LD_{50} Dose Toxaphene

Animal	Preparation of Toxaphene Used	Dose in mg./kg. body weight
White Rat	1% emulsion in peanut oil and acacia	40
	20% kerosene solution	100-150
	5% kerosene solution	300-350
	5% corn oil solution	120-125
Guinea Pig	5% kerosene solution	350-375
	5% corn oil solution	250-280

Parker and Beacher (139) give oral LD_{50} dosages for toxaphene in

deodorized kerosene and in corn oil as 280 mg./kg. and 120 mg./kg., respectively, for white rats. Lehman (100) places the oral LD_{50} dose of toxaphene for the white rat at 60 mg./kg. body weight.

For the dog, the oral LD_{50} dose of toxaphene in corn oil is approximately 20-30 mg./kg. body weight (143). The same author states that it was found that as little as 250 mg./kg. of toxaphene in deodorized kerosene could kill while the majority of the dogs would survive 400 mg./kg. body weight. It is apparent that toxaphene is more toxic when it is fed in digestible oil than when it is fed in deodorized kerosene. The speed and extent of intestinal absorption appear to be factors in determining the oral acute toxicity for the dog. (144).

Lackey (143) has estimated the oral LD_{50} dose of toxaphene for dogs to be approximately 25 mg./kg. body weight when it is administered by stomach tube as a corn oil solution containing either 5 grams or 10 grams per 100 cc. solution. However, when kerosene solutions containing either 5 grams or 25 grams toxaphene per 100 cc. solution were administered by stomach tube, the absorption of the poison was greatly reduced due to the poorly absorbed solvent.

Convulsions and death commonly occur in dogs when a dose of 15 mg./kg. body weight of toxaphene is administered by stomach tube as a solution in corn oil. A dose of 50 mg./kg. is uniformly fatal (145).

Intravenous LD_{50} Values for White Rats: The intravenous LD_{50} dose of toxaphene for white rats was determined to be 13 mg./kg. body weight when the toxicant was administered as a one per cent emulsion in peanut oil and acacia (142).

Percutaneous Absorption in Rabbits and Dogs: The percutaneous absorption of toxaphene has been studied in the rabbit and the dog (146). Various preparations were used, such as toxaphene in dimethyl phthalate and in mineral oil, dusts containing various proportions of toxaphene and water dispersions of toxaphene.

In rabbits, the application of a sufficient dose of any one of the liquid preparations of toxaphene to the skin was lethal. None survived more than five days. A dose of 250 mg./kg. toxaphene in peanut oil may kill the rabbit within 24 hours. Kerosene solutions of toxaphene in doses of 100 mg./kg. body weight applied to the skin daily for six days gave "high" mortality rates in the rabbit and guinea pig (142). When toxaphene was applied to the skin of the rabbit as a 40 per cent dust, a dose of 500-1000 mg./kg. was required to produce symptoms of poisoning or death within 24 hours. Rabbits were not affected by the daily application of 5 per cent toxaphene dust in the dose of 125 mg./kg. (toxaphene) upon the skin for six weeks.

Lehman (100) states that a single dose of 780 mg./kg. toxaphene in solution, when applied to the skin of a rabbit, is lethal. Toxaphene induced fatalities in dosages of 200 mg./kg./day applied repetitively.

In dogs, no fatalities were produced by the application of a number of preparations of toxaphene upon the skin (146).

Toxaphene in concentrated form has been reported to cause moderate irritation of the skin of laboratory animals (100).

The administration of single doses of toxaphene to experimental animals

may be associated with wide variability in the responses of animals of the same species as well as of those of different species. This variability is due in considerable degree to the physical form in which the insecticide is administered to the concentration of toxaphene and the vehicle used, and to the route of administration, and in part to the "vagaries of the absorptive mechanisms in the various animal species" (144).

Subacute and Chronic Toxicity.

Rat and Guinea Pig Experiments: Toxaphene at 1200 ppm. in the diet of rats can be tolerated for two months (138). Other experiments (142) indicate that 100 ppm. and 800 ppm. of toxaphene in the diet of white rats and guinea pigs are tolerated for six months without causing any significant effect on growth, form and numbers of blood cells, urinary abnormalities, reproduction, mortality, or tissue changes. However, the repetitive feeding of toxaphene at 100 ppm. for two years to rats produces gross injurious effects such as retardation of growth, greater incidence of mortality, increased weight of certain vital organs, loss of appetite, and occasionally, other signs of chronic poisoning (35).

Lehman reports (35) that toxaphene was fed to rats for 104 weeks (two years). The lowest level in the diet which produced gross effects was 100 parts per million. Tissue damage was observed in the rats fed at the level of 100 ppm. in the diet. Apparently, the highest level in the diet which produced no tissue damage was 20 parts per million.

Dog Experiments: Dogs were given capsules containing toxaphene in corn oil on five days of each week. The animals given the dose of 5 mg./kg.

survived 1360 days and were then killed for examination. Of the two animals that were given 10 mg./kg. in corn oil, one animal died at 33 days and the other animal was killed at 1260 days for study (35).

Exposure of Large Animals to Toxaphene: The effect of toxaphene on dairy cattle, calves, sheep and goats has been the subject of several field studies. Leighton, Kuipen, and Smith (147) divided their study of the toxicologic effects of toxaphene on dairy cattle into three phases. Since toxaphene is used for the control of grasshoppers in salvaging forage and pasture crops, and in the control of flies, lice and ticks on cattle, it is of interest to know what effect toxaphene might have on the cattle subjected to the exposure associated with these practices. These authors determined the toxic levels of toxaphene by feeding fairly large amounts of the insecticide in the form of gelatin capsules containing wettable powder. Cattle that received more than five grams of the wettable powder daily developed toxic symptoms and finally died.

The second phase of this investigation involved feeding two Jersey cows while in milk production, 0.5 grams of "pure" toxaphene per day. This is the amount of insecticide that dairy cattle might consume in forage which has been sprayed with the recommended quantities of toxaphene required for the control of grasshoppers. The milk from these two cows was fed to eight calves from birth to thirty days. No abnormalities were observed in tissues of the calves when they were killed for gross and microscopic examinations.

The last phase of this work consisted of spraying eight Jersey cows every two weeks over a period of 3.5 months with two to three quarts of

spray containing 0.5 per cent of toxaphene made from wettable powder concentrate. Four cows were sprayed every two weeks for two months with 0.5 per cent toxaphene prepared from emulsifiable kerosene concentrate. In both cases toxaphene appeared in the milk for five to seven days after each spraying. The organic chloride content of the milk rose sharply to about five parts per million.

Radeleff and Bushland (148) have shown that 0.5 per cent emulsion dips and sprays of toxaphene produce no injury to cattle and calves. However, two applications of 1.5 per cent emulsion produced symptoms in calves. The 8.0 per cent emulsion is lethal to calves, whereas the 8.0 per cent suspension causes no irreversible illness. Cattle can withstand treatment with 4.0 per cent emulsion, although the majority of sheep so treated are affected adversely.

Bushland, Wells and Radeleff (39) have shown that sprays and dips of 1.5 per cent emulsions of toxaphene are safe for cattle, sheep, horses and goats.

Marsh (149) fed toxaphene-treated alfalfa to cattle and sheep. Fourteen yearling steers and fourteen ewe lambs were divided into seven lots of two steers and two lambs each. The individuals in six of these lots were fed on alfalfa hay which had been treated with toxaphene at different rates, varying from one pound per acre to eight pounds per acre. One lot which was used as a control group was given untreated alfalfa hay.

None of the animals showed any toxic effect except the two steers given hay treated with eight pounds of toxaphene per acre, which developed temporary nervous symptoms but recovered promptly and completely.

Metabolism of Toxaphene: Very little information is available regarding

the distribution, storage, fat in the body and excretion of toxaphene. This lack of knowledge arises from the fact that there are no suitable microanalytic chemical methods which are specific for toxaphene. At present, under certain conditions, it is possible to estimate the quantity of the insecticide by using methods which measure the total organic chloride content of the sample. It is also possible to assay the amount of toxaphene present in fat and muscle by using the fly bioassay method.

Detoxification Mechanisms: Lehman (111) assumes that toxaphene, like camphor, is slowly metabolized in the liver. This assumption is based on the chemical similarity of toxaphene to camphor and on the isolation of ethereal sulfate and glucuronic acid conjugates of toxaphene from the urine of animals exposed to toxaphene (111).

Storage: Toxaphene has been placed among the chlorinated hydrocarbon insecticides which appear to be metabolized and not stored in the fat in significant amounts (35). The extent of storage of toxaphene in the fat has been determined by the fly assay method. Rats were fed toxaphene at levels of 100, 400 and 1600 ppm. in their diet for two years. The average level of storage of toxaphene in the fat of the female rats was 0, 180, and 270 ppm., respectively.

In dogs that have been exposed to toxaphene for periods as long as 106 days, there was evidence of cumulative effects from single daily doses (35). This was taken to mean that toxaphene is stored in the body and is only destroyed or excreted with difficulty. Toxaphene appeared to be stored in the brain of the dog but none was found in the body fat.

Radeleff, Claborn, Beckman, Wells and Bushland (150) showed that young Hereford steers held on a moderately fattening diet could be sprayed repeatedly with 0.5 per cent toxaphene without accumulating large amounts of toxaphene in the fat and without damage to internal cellular structures of the body. The conclusion, with reference to the storage, is based on the results of organic chloride analyses.

Single applications of 0.5 per cent sprays produced no residue which was detectable two days, two weeks, or four weeks later. Some increase in organic chlorides was noted following two to six applications at two-week intervals. These increases disappeared in from 6 to 18 weeks following suspension of the sprays. No significant increases in organic chlorides occurred in the fat of cattle sprayed eight, ten or twelve times at bi-weekly intervals.

Fat from animals fed for four months on alfalfa hay which had been sprayed twice with toxaphene at the rate of one and two pounds per acre showed toxaphene concentrations of about 25 and 300 ppm., respectively. The concentrations of toxaphene in the muscle from these same animals were relatively low, the rib roasts giving results less than 1 ppm. and 7 ppm., respectively. The fat of the steer fed on hay, on which there had been two applications of four pounds per acre, contained about 700 ppm. while the lean meat from the same steer yielded 35 ppm. (151).

Fat samples taken by biopsy from steers at eleven, nineteen and twenty-three weeks after they had discontinued the consumption of treated hay showed that most of the toxaphene had been eliminated by the eleventh week.

By the nineteenth week there was no significant difference in the organic chloride content of biopsy samples from control animals and those fed treated hay.

The results with sheep were generally similar to those with steers, but the distribution of toxaphene within the animal tissues was somewhat different. The quantity of toxaphene in the muscle of sheep was higher than in the corresponding cuts of steers and was lower in the abdominal and subcutaneous fat. No measurable quantities of toxaphene were found in sheep slaughtered seven months after they had ceased to eat toxaphene-treated alfalfa hay (151).

Others (147) have reported their finding of toxaphene in concentrations ranging from 6.3 mg./kg. to 93.8 mg./kg. in the omental fat of cattle fed varying amounts of toxaphene in their diets. The amount of organic chloride found ranged from 67 ppm. to 160 ppm., respectively.

Excretion of toxaphene in milk: Cattle sprayed with 0.5 per cent toxaphene prepared from either emulsifiable kerosene concentrate or wettable powder concentrate, produced milk containing concentrations of organic chloride of about 5 ppm. (147).

It is fair to draw the conclusion from the data presented above that toxaphene is stored in the fat of animals following its absorption. However, it appears that toxaphene is stored in insignificant amounts and that it does not remain long in the fat depots.

One may question whether toxaphene is excreted in milk in any amount which can be considered significant.

The entire subject of the metabolism, storage and excretion of toxaphene from the animal body awaits the development of specific chemical methods of quantitative analysis which can be applied to tissues and body fluids.

Symptoms of Toxaphene Poisoning in Experimental Animals: McGee et al (142) cite unpublished observations on ten species of animals poisoned with toxaphene. The manifestations of poisoning by the insecticide consistently consist of a diffuse stimulation of the cerebrospinal axis resulting in generalized convulsions. Sublethal doses of toxaphene cause shorter and fewer convulsions. If the dose of toxaphene is lethal, death comes with respiratory failure after a series of convulsions of increasing severity.

It has been suspected that toxaphene has the medullary excitant effects of camphor because of the occurrence of salivation and vomiting followed by a reflex hyperexcitability to sudden noise (142).

Cattle exposed to toxic levels of toxaphene show extreme nervous irritability, evidenced by licking the ground and feet, refusal to lie down, frequent defecation, loss of appetite, fits of running into fences, buildings and other cattle, and the occurrence of convulsions at varying intervals of 24 hours or more (152).

Observations on goats, sheep and calves (152) indicate that two phenomena characterize their intoxication. Some animals appear to be stunned and blind, although they are capable of vision; others pass through a period of violent hyperexcitability.

Other investigators (153) have described the clinical manifestations of fatal poisoning in goats as increased reflexes, convulsions, muscular tremors,

oliguria, anorexia and anoxia.

It is clear that all observers have been impressed by the central nervous system involvement in the poisoned animals.

Abrupt rise in the arterial blood pressure in curarized animals following the intravenous injection of toxaphene has been reported (140).

The oral administration of toxaphene to unanesthetized dogs produces a slight increase in the arterial blood pressure. This was not observed in anesthetized animals. The change in the mean arterial blood pressure was taken to mean that stimulation of the vasomotor center occurs in the poisoned animals (142).

Increase in the cerebrospinal fluid pressure in animals has been noted (123).

Pathology: It may be of some importance to note that while two animals may receive identical doses of toxaphene, react in the same manner, and die in the same length of time, the gross and microscopic pathology of the two animals may, and often does, vary widely (152).

The major histopathologic changes in the animal body which are attributed to toxic doses of toxaphene are found in the liver and kidney (152)(153). The only pathologic effects observed histologically in the dog were degenerative changes in the liver parenchyma and in the renal tubules. The hepatic changes have been described as not differing to any great degree from those observed in animals poisoned with DDT. These degenerative changes involve an increase in the size of the centrolobular hepatic cells, which may go on to central necrosis and regeneration (153).

Rabbits subjected to applications of toxaphene upon their skin also showed degenerative changes in the liver parenchyma and renal tubules (146).

In cattle, the toxic doses of toxaphene produce severe fatty degeneration in the liver, injury to the renal tubules with moderate glomerular damage and early albuminurea (152). Cerebral degeneration and hemorrhages have also been described in cattle (152).

The lungs of many calves subjected to toxaphene dips were described as having a brilliant purple color when examined at autopsy (152).

Still other articles (153) (152) have given detailed descriptions of the organs removed from goats fatally poisoned by toxaphene. The lungs, grossly, were edematous and hyperemic. The heart was enlarged and there were petechiae on the epicardium along the course of the coronary vessels. The liver was enlarged, hyperemic and glossy. The kidneys showed irregular large and small areas of congestion on the surfaces, which extended into the cortex and medulla on the cut surface. Examination of the meninges, brain and spinal cord revealed markedly injected vessels distended with blood. There was some evidence of hemorrhage in the brain.

Microscopic examination revealed abnormalities in almost all of the organs. Only the pertinent observations are listed here: (153).

Brain and Spinal Cord: Congestion, edema. Spotted softening and degeneration of the neurons in the brain and spinal cord. Obvious neuronophagia.

Heart: Congestion, edema, hemorrhage. Round cell and neutrophilic infiltration. Cloudy swelling and fatty metamorphosis were con-

sistent findings.

Lungs: Congestion, edema, hemorrhage.

Kidneys: Marked cortical and medullary congestion, hemorrhage, thrombosis of small vessels. Distention of proximal end of some tubules and Bowman's space. Tubular epithelium undergoing cloudy swelling, fatty metamorphosis, coagulation necrosis, and degeneration.

Liver: Congestion, hemosiderosis, cloudy swelling, fatty metamorphosis, and focal necrosis around the central vein were pronounced. Sinusoids were distended with shreds of coagulated protein, chromatin, and cell debris. Some of the hepatic cells were shrunken, causing narrowing of the cords and concomitant enlargement of the adjacent sinusoids.

Human Cases of Toxaphene Poisoning: McGee et al (142) have collected eleven cases which appear to have been acute poisoning due to toxaphene. Three of the eleven patients died. The eight survivors apparently recovered promptly and without sequelae.

The case histories are tabulated in the next section. It is interesting to note that the autopsy findings reported in Case No. 1 are very similar to those observed in goats.

The clinical description of Case No. 4 fits in very well with what has been observed in animal experiments.

It is apparent from these case histories that there is an absence of warning symptoms such as nausea and cramps. Only in Case No. 5 and Case No. 11 was there any suggestion of nausea.

Furthermore, there are no manifestations of acute poisoning until acute overstimulation of the nervous system occurs. There are no abdominal cramps or diarrhea to serve as evidence of poisoning. Except for Case No. 5 vomiting had to be induced in these patients. In this respect human cases of poisoning differ from the intoxication seen in animals.

There does not appear to be anything which is recognizably pathognomic either in the signs or symptoms or pathologic findings indicative of toxaphene poisoning. A history of sufficient exposure with the findings of central nervous system excitability may give the clue to the diagnosis of acute toxaphene poisoning.

Tabulation of Human Cases of Toxaphene Poisoning - data from McGee et al (142).

Case No. 1

Age: 2 years 8 months, Negro male.

Exposure: Playing around metal strips contaminated with toxaphene. Possible ingestion of amber colored beeswax-like material.

Onset of Convulsions after exposure: 8 hours (?)

Outcome: Death 9 hours after exposure. (?)

Remarks: Died in convulsions. Autopsy report stated that there was pale froth from the mouth, heart greatly dilated and lungs markedly congested and edematous. Bronchial tree filled with faintly blood tinged froth. Stomach "dilated with air". Liver, gall bladder, spleen grossly normal. Kidneys rather swollen and hilus of right kidney showed purulent-like material, possible

a pyelitis. "Few petechial hemorrhages found in white substance of brain". Pathologist's report: "Symptoms and pathologic findings consistent with death due to CNS poison of convulsant type. Actual death due to suffocation during a convulsion."

Case No. 2

Age: 4 years, white male.

Exposure: Probable ingestion of unknown quantity of toxaphene emulsion in water from a gallon jug.

Onset of Convulsions After Exposure: 2 hours.

Outcome: Death 6 hours after exposure.

Remarks: Treated with intra-muscular magnesium sulfate; phenobarbital sodium, curare; cyclopropane anesthesia shortly before death. No autopsy. Convulsions continued at 2-5 minute intervals until death.

Case No. 3

Age: 17 months, white male.

Exposure: Drank half teaspoonful full strength toxaphene (60% concentrate in special solvents).

Onset of Convulsions After Exposure: Not stated.

Outcome: Death 11 hours after exposure.

Remarks: Convulsions developed; recurring with increasing frequency until death. No medical details. No autopsy.

Case No. 4

Age: 2 years, Negro male.

Exposure: Ingested unknown amount of chemical containing 20% toxaphene.

Onset of Convulsions After Exposure: Not stated.

Outcome: Recovery without sequellae.

Remarks: Hospitalized half-hour after onset of convulsions. Symptoms of mild cerebral excitement with hypertonicity of all extremities proceeding to generalized convulsions associated with marked laryngeal and pharyngeal spasm; respiratory embarrassment and cyanosis. Pupils fixed and dilated. Eyes had wild, staring expression. Neck stiff and back bowed. Treatment: large doses of sodium luminal and curare intramuscularly. Artificial respiration and airway during convulsive episodes. Twelve hours after hospital admission, relatively good condition. Recovered with no symptoms. Discharged from hospital two days later.

The next four cases occurred in a family of seven people who had eaten collards from a patch that had been sprayed twice with 9% toxaphene in aqueous emulsion. The collard greens were washed in three different waters before they were cooked with salt pork. Analysis of the washed collards for organic chloride calculated as toxaphene gave a result of 3.126 parts per million. Three other members of this family had eaten the collards but showed no evidence of illness.

Case No. 5

Age: 49 years, Negro female.

Exposure: Ate collards from a patch sprayed twice with 9% toxaphene in aqueous emulsion.

Onset of Convulsions After Exposure: None.

Outcome: Complete recovery.

Remarks: One and one-half hours after ingestion became sick at stomach and vomited. Took 2 ounces of castor oil in vinegar. No further symptoms.

Case No. 6

Age: 20 years, Negro female.

Exposure: Same as in Case No. 5.

Onset of Convulsions After Exposure: 2½ hours.

Outcome: Complete recovery.

Remarks: Convulsion of 10 minutes duration. Lucid interval with no recollection of convulsions. One and one-quarter hours later another convulsion occurred.

Case No. 7

Age: 12 years, Negro female.

Exposure: Same as in Case No. 5.

Onset of Convulsions After Exposure: 3 hours.

Outcome: Complete recovery.

Remarks: Lucid period of 30 minutes before the onset of the second convulsive episode.

Case No. 8

Age: 16 years, Negro female.

Exposure: Same as in Case No. 5.

Onset of Convulsions After Exposure: $3\frac{1}{2}$ hours (?).

Outcome: Complete recovery.

Remarks: Convulsion lasting 3-4 minutes.

The next three cases occurred in a family that had eaten chard from a garden that had been sprayed with toxaphene on the morning the chard was harvested.

Case No. 9

Age: (?) Mature white male.

Exposure: As stated above.

Onset of Convulsions After Exposure: 6 hours (?).

Outcome: Complete recovery.

Remarks: "Mind went blank. Unconscious for 1 hour. Periods of amnesia." No medical attention.

Case No. 10

Age: (?) Child, white male.

Exposure: As stated above.

Onset of Convulsions After Exposure: 5 hours (?).

Outcome: Complete recovery.

Remarks: "Wakened from sleep by jerking and twisting of muscles".

Case No. 11

Age: Adult, white female.

Exposure: As above. No convulsion.

"Felt light-headed and a feeling of mild nausea".

Outcome: Complete recovery.

So far, there has been no published case of chronic intoxication due to toxaphene.

McGee et al (142) also make a point of the fact that during the 5 years in which chemists have done research on toxaphene and in which scores of workers have been potentially exposed in the manufacture of toxaphene, there have been no instances of poisoning in man. Periodic examinations of these men have shown no alterations in the reflexes. The urine examinations and blood cell counts have been within normal limits. There have been no subjective manifestations indicating disorders of the nervous system, or any other signs of impairment to health due to exposure to toxaphene. It is further noted that other materials such as solvents, inert dusts, and chlorine have been more troublesome than toxaphene.

Treatment: Lackey and Weed (145) tested drugs recognized as having a depressant action upon the central nervous system. These drugs included morphine, methodon, paraldehyde, dilantin, tridione with urethane, and the sodium salts of amytal, pentobarbital and phenobarbital. All except paraldehyde were administered intravenously and in divided doses. These investigators poisoned 64 dogs with toxaphene using a dose of 50 mg./kg., which was administered after a fasting period of 18 hours. This dosage is

always lethal to dogs. It was found that convulsions were controlled and the life of the dogs preserved if adequate dosages of sodium salts of pentobarbital, phenobarbital or amytal were given intravenously. Treatment was more effective if started before the onset of convulsions. When convulsions have already developed, sodium pentobarbital was found to be the drug of choice.

Treatment of toxaphene poisoning should include the evacuation of the gastro-intestinal tract. Therefore, gastric lavage and saline cathartics are the accepted measures for the removal of ingested insecticides.

CHAPTER VI

THE DIELS-ALDER CONDENSATION PRODUCTS

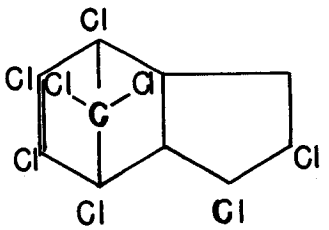
In 1950 Otto Diels and Kurt Alder shared the Nobel prize in Chemistry for their work on the diene synthesis. The reaction which has come to be known as the Diels-Alder condensation is the basis for the production of certain of the chlorinated hydrocarbon insecticides, among which are chlordan (1945), heptachlor (1945), aldrin (1949), dieldrin (1949), endrin (1951) and isodrin (1951). Since the details for manufacturing these products are still trade secrets, an illustration is given as to how a reaction might go. Hexachlorocyclopentadiene and cyclopentadiene are subjected to the Diels-Alder condensation reaction. The resulting product is treated with chlorine to produce chlordan (79).

CHLORDAN

The compound which is now called chlordan was described for the first time in 1945 (155). Chlordan has had several aliases among which are Velsicol 1068 and Octa-Klor. At present there is only one manufacturer of this insecticide, The Velsicol Corporation of Chicago.

Chemically, chlordan is 1,2,4,5,6,7,8,8-octachloro-2,3,3a,4,7,7a-hexahydro-4,7-methanoindene, or it is sometimes referred to as 1,2,4,5,6,7,8,8-octochloro-4,5 tethano-3a,4,7,7a tetrahydroindane; occasionally, as octachloromethano-hexahydro-indene.

The structural formula is given as:



Technical chlordan should contain not less than 60 per cent of chlordan, together with 25 to 40 per cent of related compounds (dicyclopentadienes) which are products of the normal manufacturing processes and are toxic for insects(156) (157).

Chlordan occurs as two structural isomers which are the cis and trans forms, or alpha and beta chlordan. These isomers cannot be separated from each other by distillation because the liquid superheats. However, by chromatographic adsorption on aluminum hydroxide, it is possible to separate the two isomers and also to obtain two further derivatives of tetrahydroindene. All of these are white crystalline solids. The known constituents of technical chlordan are as follows:

1. cis - 2,3,4,5,6,7,8,8-octachloro-2,3,3a,4,7,7a-hexahydro-4,7-methanoindene (alpha chlordan - M.P. 102-104 degrees C.)
2. trans-2,3,4,5,6,7,8,8-octachloro-2,3,3a,4,7,7a-hexahydro-4,5-methanoindene (beta chlordan M.P. 104-106 degrees C.)
3. 1 or 3a,4,5,6,7,8,8-heptachloro-3a,4,7,7a-tetrahydro-4,7-methanoindene (heptachlor M.P. 92-94 degrees C.)

4. 4,5,6,7,8,8-hexachloro-3a,4,7,7a-tetrahydro-4,7-methanoindene (hexachlor or Compound 237 M.P. 154 degrees C. with decomposition.)

5. 1 or 3a, 2,3,4,5,6,7,8,8-enneachloro-2,3,3a,4,7,7a-hexahydro-4,7,methanoindene (trichlor 237 M.P. 122-123 degrees C.)

Physical and Chemical Properties: Technical chlordan is a viscous, dark amber liquid with a faint terpene-like odor. It has a molecular weight of 409.8. The calculated chlorine content is 69.22 per cent. Chlordan boils at reduced pressure (2 mm. Hg) at 175 degrees C. The refined chlordan is pale amber with a faint aromatic odor. The vapor pressure is 1×10^{-5} mm. Hg at 25 degrees C. (156) (157).

Chlordan is insoluble in water but it is completely miscible with all apolar solvents. It is soluble in aromatic, aliphatic and chlorinated hydrocarbons as well as in ketones, esters and ethers. It is completely miscible with deodorized kerosene.

Chlordan is stable to acids, but, in the presence of alkali, it is readily dehydrochlorinated with the concomitant loss of its toxicity for insects.

It has been reported that chlordan has doubtful compatibility with basic lead arsenate, calcium arsenate, nicotine, Bordeaux mixtures, lime sulfur, zinc sulfate plus lime, and lime (7) (9) (156). Chlordan is fully compatible with the dinitro compounds, fluosilicates, other chlorinated

hydrocarbons, dithiocarbamate, sulfur and all other insecticides and fungicides (9) (156).

Analytical Methods: There are no specific chemical or physical methods for the determination of chlordan in body tissues or fluids.

A method for the qualitative test for chlordan in insecticide spray oils has been devised (158). A one milliliter sample is added to 2 milliliters of cellosolve-pyridine (40:10) solution and one milliliter of 1 N solution of alcoholic potassium hydroxide. This combination is heated in boiling water for 5 minutes. The development of a red color indicates the presence of chlordan. Very weak colors are disregarded.

A confirmatory test makes use of the odor of crude methyl naphthalene upon dehydration with sodium in boiling isopropyl alcohol. Toxaphene is the only other insecticide which will give this odor.

The total chlorine in chlordan may be determined. The sample is decomposed with sodium in refluxing anhydrous isopropyl alcohol. The sodium chloride which is formed is titrated with N/10 silver nitrate solution, using the electrometric titrimeter. Chlordan is calculated as follows: Total organic chlorine X 1.44 (the factor is based on the fact that pure chlordan contains 69.22 per cent of chlorine) (159). This is the method used in the Division of Insecticide Investigations, Bureau of Entomology and Plant Quarantine.

Davidow has devised a spectrophotometric method for the quantitative estimation of technical chlordan. This method is useful for alpha chlordan, beta chlordan, heptachlor and trichloro 237 when only one of these compounds

is present. The sample is added to n-hexane and diethanolamine-potassium hydroxide reagent and heated over boiling water for 30 minutes. Absorbency is noted at 521 millimicrons using the Beckman spectrophotometer (160).

The fly bioassay method has been useful in estimating small quantities of chlordan (161) (162).

Modifications of these methods have been described, which according to various authors have met specific needs (156) (163) (7).

The Toxicity of Chlordan: Oral LD_{50} Values for White Rats. Ingle (164) obtained the LD_{50} value of 225-250 mg./kg. However, Lehman (100) places the LD_{50} value at 500 mg./kg. Stohlman, Thorp and Smith (165) administered chlordan in olive oil by stomach tube to rats. The LD_{50} value which they obtained is 200-250 mg./kg. Ambrose et al (166) obtained an LD_{50} value of 590 mg./kg. when chlordan in cottonseed oil was given by gastric intubation.

Intravenous LD_{50} for Rabbits. Stohlman and Smith (167) found the LD_{50} value to be 20 mg./kg. They discovered that by the use of barbiturates this value could be increased to 60 mg./kg.

Percutaneous Absorption of Chlordan: Frings and O'Tousa (168) studied the toxicity of chlordan vapor and of solutions applied upon the skin of mice. After treating the mice for 20 weeks (5 applications per week) with 0.01 ml. of 2 per cent chlordan in kerosene and with large quantities of an aqueous suspension of the wettable powder until the total dose of 20 mg. of chlordan (740 mg./kg.) in kerosene solution and four times as much in

aqueous suspension had been given, the skin showed little or no dermatitis from the wettable powder but some roughening with local areas of bleeding and ulceration from the chlordan in kerosene. The vapor was employed in two experiments in which the skin, but not the respiratory apparatus was subjected to contact with the vapor; one, in which the compound was painted on the walls of the ventilated chamber every three weeks, and the other, by exposure of the skin of the animals to the saturated vapor from a bubbler. These experiments demonstrated that toxic symptoms could be produced, and death occurred in 1 to 10 days following the exposure for 9 - 15 hours daily to saturated chlordan vapor. It was concluded that chlordan can be absorbed from the skin either as a vapor or solution with relative ease.

Ambrose et al (166) applied chlordan in cottonseed oil to the skin of rats in amounts of 50 mg. per rat per day for three or four days, thereby killing all of the rats tested. However, chlordan in ethyl alcohol, when applied upon the skin of the rats, in equivalent amounts, did not produce toxic symptoms.

Lehman (100) states that chlordan produced severe systemic effects when applied upon the skin of the rabbit in a single dose of 780 mg./kg. In the experiments involving multiple applications, 200 mg./kg./day of chlordan produced fatalities.

The toxicity of chlordan vapors: The experimental evidence which deals with the toxicity of chlordan vapors is confusing. Lehman (100) was unable to maintain pigeons in a room which had been treated previously with chlordan

and then thoroughly scrubbed and aired. Frings and O'Tousa (168) stated that mice could not survive in air which had first passed through chlordan. Mice confined in a chamber the walls of which had been treated with chlordan also developed toxic symptoms. However, other investigators (169) (190) have failed to observe any harmful effects of chlordan vapor on pigeons or chickens confined from 30 to 60 days in a box the interior surface of which had been treated with chlordan. Heyroth and Witherup (171) subjected rats, mice and guinea pigs to aerosols of chlordan in methylene dichloride or dimethyl phthalate for 45 minutes on each of 38 days. The average initial concentration of chlordan was approximately 1.9 mg. per liter of air. No harmful effects were noted among the animals following such exposure. Ambrose, Christensen, Robbins and Rather (166) state that rats exposed for 120 hours to air passing through chlordan showed no evidence of intoxication.

Ingle (172) exposed female Swiss albino mice continuously for 14 days to a current of air (18 ml./sec.) which had first passed through 105 ml. of chlordan in a saturation train, without harm to the animals. However, when hexachlorocyclopentadiene was added to chlordan in various proportions, cessation of feeding and drinking, huddling together, lethargy, apparent blindness, and loss of coordination were noted among the animals. The onset and severity of the symptoms were directly proportional to the volume of added hexachlorocyclopentadiene. On this evidence, Ingle came to the conclusion that the reported toxicity of the vapors of the insecticide to mice should not have been attributed to chlordan but rather to an unreacted

intermediate. It would appear that chlordan as it is presently manufactured does not contain sufficient quantities of unreacted hexachlorocyclopentadiene or other intermediates to contribute significantly to its toxicity. This experiment helps to explain the conflicting results obtained by various investigators who reported on the toxicity of the vapors of chlordan at various times.

Heyroth and Witherup (171) showed that when aerosols of chlordan with either kerosene, methylene dichloride, or dimethyl phthalate were introduced into a chamber at 10 minute intervals over the period of an hour on three different occasions on each of four successive days in the initial concentration of 10 mg. of chlordane per liter of air, many of the animals showed typical signs of poisoning due to chlordan.

Subacute and Chronic Toxicity: Ambrose et al (166) administered chlordan in cottonseed oil to animals by gastric intubation in amounts of 25 mg./kg. or less per day for 5 days without provoking toxic symptoms. The administration of oral doses equivalent to 50 mg./kg. or more in a corresponding manner, resulted in toxic symptoms and death. These authors concluded that chlordan is a cumulative poison and that its rate of elimination or detoxication is slower than its absorption.

These authors also fed male and female rats on diets containing 0.001 to 0.128 per cent of chlordan for as long as 410 days. The data obtained indicate that chlordan in concentrations of 0.064 and 0.128 per cent in the diet increased the mortality rate; in the concentration of 0.032 per cent it produced definite retardation of growth; in concentrations of 0.002, 0.004 and 0.016

per cent it produced some suggestion of retardation in the growth of male, but not female, rats.

Lehman (35) reported that various levels of chlordan were fed to rats in their diet for 104 weeks. The lowest level at which gross effects were noted was 75 ppm. The highest level at which no gross effects were seen was 25 ppm. The lowest level fed to the rats was 2.5 ppm. and even at this level some evidence of tissue damage was said to have been found.

It appears that chlordan is stored as a metabolite and not as chlordan in the fat of the rat (35). Male rats fed on 2.5, 5.0, 10.0, 25.0, 75.0, and 200 ppm. in the diet for a period of two years showed an average storage of 75, 65, 50, 300, 500, and 800 ppm. respectively, while female rats, under the same conditions stored 90, 100, 115, 300, 750 and 4500 ppm. respectively. These results were obtained by the fly bioassay method.

It is apparent from the above figures that the metabolites of chlordan are stored in the fat in concentrations greater than those in the diet consumed, and that the storage progresses with time until a maximum is reached.

If chlordan is compared to DDT, with respect to its chronic toxicity, then the dietary concentration of 0.12 per cent of DDT is required to give a mortality rate approximating that of 0.05 per cent chlordan (35).

A level of concentration of chlordan of 10 ppm. in the diet of rats over a life time is sufficient to cause an increase in the weight of the liver. At 2.5 ppm. chlordan in the diet, minimal effects were observed (35). The significance of these effects, as expressions of toxicity, is not appar-

ent in either instance, however.

Rats were fed chlordan in the diet at levels of 5, 10, 30, 150, and 300 ppm. for a period of two years (173). Chlordan in the diet to the extent of 300 ppm. produced marked toxic effects, which began to appear by the end of the 12th week. At 150 ppm. it produced less marked effects, but the first observable symptoms occurred by the end of the 26th week. In the lower dosages, the effects were insignificant.

Chlordan, when fed at the level of 150 ppm. for 80 weeks, is apparently stored in the tissues of rats in quantities sufficient to be released with lethal effects when food is withdrawn for 48 hours. Rats given chlordan in the ration at 150 ppm. for 80 weeks, and then placed on chlordan-free diets lost the stored metabolite at such a rate that after 4 weeks the stored compound had decreased to below toxic levels.

The death rates were significantly higher among the 150 and 300 ppm. groups than among the controls. The mortality in the lower dosage groups did not differ significantly from the control group.

The young of litters from rats at all levels were normal with respect to size, growth, and development when allowed to nurse from non-treated foster mothers. If the young from rats at 150 and 300 ppm. groups remained with their lactating mothers, they showed definite symptoms of chlordan intoxication.

Ambrose et al (166) studied the storage of chlordan in the perirenal fat of rats on a diet containing 0.032 per cent of chlordan. After 5 days on the diet, there was appreciable storage of chlordan. Storage was no

greater after 148 days on the diet than at 5 days. Withdrawal of chlordan from the diet for 5, 10, or 20 days was accompanied by progressive decrease in the amount of residual storage to almost complete disappearance after 20 days. Rats on the diet for 405 or 410 days showed greater tissue storage than was observed after 5 or 148 days. The amount stored was greater in the perirenal fat of female than of male rats.

The symptomatology and pathologic findings will be discussed under appropriate headings in subsequent sections.

Dogs were fed various doses of chlordan in corn oil in capsules on five days of each week. The following table was adapted from Lehman (35) to indicate the survival time of the animals:

Daily Dose mg./kg.	Number of Animals	Number of Deaths	Earliest Death days	Last Death days
80	2	2	32	37
40	2	2	25	26
20	2	2	30	68
5	4	4	39	650

Dogs vary in susceptibility to poisoning with chlordan (174). Dogs were starved for 24 hours and then fed 50 per cent chlordan wettable powder in capsules with oil. A dose of 200 mg./kg. produced convulsions in one dog, but one of 700 mg./kg. had little effect on another dog.

Effect of Chlordan on Large Animals: Cattle, sheep, goats, hogs, and horses were treated with chlordan formulated as emulsions and as suspensions of wettable powder at the concentration of 1.5 per cent of the technical material. The insecticide was applied as sprays and dips, which were spaced

at 4-day intervals until they numbered eight. The sheep dipped in the chlordan emulsion died, as did also yearling lambs and yearling goats dipped in the suspension of chlordan wettable powder. Two goats dipped in chlordan emulsion died. The other species of animals survived. None of the animals that developed symptoms of chlordan intoxication recovered (149).

Sheep grazing on pastures sprayed with technical chlordan at rates up to 4 pounds per acre showed no apparent ill effects (175). In another experiment, six sheep were permitted to graze on pastures for 21 days immediately following the spraying of the plots with chlordan at rates from one pound to four pounds per acre. None of the animals showed any indication of having been poisoned by chlordan (38).

Welch (38) administered oral doses of chlordan in capsules on each of 60 days to sheep and steers. It was observed that 4.5 grams of a mixture of chlordan and xylene (3.5 grams chlordan) were extremely toxic to sheep. Even half of this dose was just about as toxic to sheep. When the dose was lowered to one gram of the chlordan-xylene mixture (0.77 grams chlordan), only mild symptoms were observed. This observer concluded that the maximum safe, single dose of chlordan for sheep was considerably less than 0.5 gram/kilogram. The maximum safe, single dose for cattle was not determined but no toxic effect was produced by 0.05 gram of chlordan per kilogram of body weight.

Metabolism of Chlordan: Woodward, Davidow and Lehman (49) stated in 1948 that chlordan is absorbed by the animal body but that its fate and urinary excretion was unknown. Some progress has been made since this statement

was made. However, observations dealing with the metabolic fate of this insecticide have been hampered by the lack of specific chemical methods for the detection of chlordan in minute quantities in the animal tissues and body fluids. To make the situation even more complex, chlordan is not a definite chemical entity but is, rather a mixture of substances.

Davidow, Hegan and Radomski (176) analyzed the liver, kidney, and fat of rats on diets containing technical chlordan in the concentration of 500 ppm. Using Davidow's spectrophotometric method, it was found that the fat produced the most intense color; however, the absorption spectrum was different from technical chlordan. Rats were fed on the individual components of technical chlordan. The following schedule was used: heptachlor, 62.5 ppm.; trichloro 237, 250 ppm.; and alpha and beta chlordan, 250 ppm. The absorption spectra obtained from the colored reaction products of the individual components and the material obtained from the fat were compared. It was concluded that the components of technical chlordan were stored as a metabolite.

The experiment was carried further. Dogs were fed heptachlor daily at the rate of 1,3, and 5 milligrams per kilogram of body weight. Rabbits were fed 5 and 20 milligrams of heptachlor per kilogram. The absorption spectra of the colored reaction product in the fat of the dog, rabbit, and rat were found to be identical. The lower doses permitted only the metabolite to be found in the fat. At the higher dose levels, both the metabolite and heptachlor were present. However, only the metabolite was found in the liver.

Ambrose et al (166) used the colorimetric method of Davidow (160) for the

estimation of the amount of chlordan stored in the tissues of the experimental rats. These animals had been fed on diets containing 0.032 per cent of chlordan for varying periods of time. From these observations they concluded that chlordan is either rapidly eliminated or is converted into some other substance which does not respond to the colorimetric method of Davidow. They also believe that complexes, other than chlordan - possibly degradation productions of chlordan - are present or are being formed at variable rates.

Stohlman and Smith (167) report that an unidentified chlorine-containing degradation product with acidic properties was recovered from the urine of rabbits treated with chlordan. Approximately one-third of its chlorine content was set free on hydrolysis at 100 degrees C. with sodium hydroxide in either absolute alcohol or in water. The meaning of this is not clear. However, the authors state that the conversion in the animal body of at least some water-insoluble chlordan to a water-soluble degradation product must facilitate the elimination of the poison through its excretion into the urine by the kidneys. It was thought that perhaps this may be a mechanism for its detoxification.

Storage: The fat of sheep and goats was analyzed for chlordan after the animals had been dipped at 4-day intervals in formulations containing 1.5 per cent of chlordan. A suspension of wettable powder of chlordan applied to sheep gave rise to 254 ppm. insecticide in the fat one month after the last dip. After 7 months only 19 ppm. chlordan remained in the fat of sheep. The goats showed no chlordan after 7 months. An emulsion preparation of

chlordan produced 112 ppm. in the fat of sheep one month after the last dip. After a lapse of 7 months there remained 25 ppm. of chlordan in the fat of the sheep. No chlordan was found in the fat of goats at the end of 7 months (121).

The fat of steers sprayed 12 times at bi-weekly intervals with an 0.5 per cent emulsion of chlordan contained organic chlorine in the average concentration of 17 ppm. 2 weeks after the last spraying. Four weeks later, the average had dropped to 6 ppm. and after 10 weeks, the organic chlorine dropped to the apparently normal level of 5 ppm. (121).

Claborn, Beckman and Wells (121) used 0.5 per cent of chlordan in a xylene emulsion and a wettable powder suspension. The spraying was repeated 3 weeks later. Analyses of the fat of the animals by the organic-chlorine method gave negative results.

Yearling Hereford cattle were fed chlordan in the diet at the level of 25 ppm. for 56 days. Chlordan in the concentration of 12 ppm. was found in the fat on the 28th day and in that of 19 ppm. on the 56th day of feeding. Twelve weeks after the feeding was discontinued, only 5 ppm. of the insecticide remained in the fat. It disappeared completely after 20 weeks. Delaine sheep fed on the same diet showed only 7 ppm. at the 28th day and 12 ppm. on the 56th day. No insecticide was found in the fat 4 weeks after the feeding was stopped. Yearling Hereford cattle fed for 112 days on a diet containing chlordan at the level of 10 ppm. had 11 ppm. in the fat at the end of the feeding period. Fat of Delaine sheep fed on the same diet contained 9 ppm. after the same feeding period (40).

Excretion in Milk: One cannot be sure of the significance of the small amounts of organic chlorine which have been found in cow's milk. However, analyses for organic chlorine indicate that chlordan is secreted in the milk in small amounts. Other analyses of the milk from cattle sprayed twice with 0.5 per cent of chlordan in a xylene emulsion and a wettable powder suspension were reported as negative when analyzed by the organic chlorine method (177).

Symptoms of Chlordan Poisoning in Animals: Radeleff (178) described the symptoms and signs of chlordan poisoning in goats and sheep subjected to sprays and dips of chlordan either as 1.5 per cent emulsion or 1.5 per cent wettable powder suspension. In the cases of acute poisoning, the animals experienced a sudden onset of symptoms with bleating, groaning, grinding of teeth, blindness, violent struggling, and terminal cyanosis. In cases of subacute and chronic poisoning, the symptoms had a gradual onset. There was partial to complete blindness, locomotor ataxia, circling, staggering, other actions which seem to indicate that the animals were avoiding imaginary objects, and periodic convulsions.

Others (153) reported that goats fed 750 mg./kg. of 40 per cent chlordan in a nontoxic diluent (attaclay) daily for seven days developed the following signs. Twenty-four hours after the first dose, dullness, partial loss of appetite, hyperesthesia to touch or loud noises, slightly dilated pupils, and occasional groaning and bleating were noted. After 48 hours, the animals had complete anorexia, slight loss of weight, widely dilated pupils and a hunched appearance with the flanks tucked up. As the symptoms developed, the animals

showed increased nervous reflexes, restlessness, frequent attempts at micturation, a staggering circling gait, and muscular stiffness of the hind legs. When attempts were made to examine or treat the animals, they developed paroxysms of convulsions lasting several minutes. Along with the convulsions, the legs were held either rigidly or made paddling motions - the respirations became shallow and rapid, and finally, the animals became cyanotic and died.

In the rat (164), the predominant signs produced by chlordan were anorexia, loss of weight, hyperexcitability, tremors and tonic and clonic convulsions.

Mice exposed to saturated chlordan vapor for 9 to 15 hours daily stopped eating and drinking and huddled together. Within a few hours, they became blind and lost coordination. Death occurred in one to ten days (168).

In chronically poisoned rats, the first noticeable signs were hyperexcitability and increased sensitivity, which sometimes progressed to violent convulsions lasting from 30 seconds to a minute. The convulsions were followed for several minutes by deep depression and prostration, after which the animals appeared normal until the next seizure. Convulsions could be induced by such stimuli as a jet of air, metallic sounds, or occasionally, by merely opening the cage. The tremors may be mild (173).

Pathology: Chlordan is considered to be a hepatic poison (112) (178). It seems to produce less liver damage but greater pulmonary damage than DDT. In rabbits, focal necrosis in the liver, congestion, edema, and exudates in the lungs were the outstanding lesions. In addition, degenerative changes

in the intestinal submucosa and in the convoluted tubules of the kidneys were noted (165).

In mice that died within one to ten days following exposure to saturated chlordan vapor, autopsies showed no definite changes except emaciation in those which survived longest (168).

Ambrose et al (166) could not demonstrate any abnormal hematologic changes among the experimental rats fed on diets containing from 0.001 to 0.128 per cent (by weight) of chlordan for as long as 410 days. The histopathologic alterations seen in the tissues of these animals were chiefly confined to the liver. Liver cells and liver cell nuclei and nucleoli were significantly enlarged in both male and female rats fed on diets containing 0.008, 0.016 and 0.032 per cent of chlordan. Inclusion bodies in the liver cells were not seen in male rats fed on diets containing 0.001 and 0.002 per cent of chlordan and in female rats fed on diets containing 0.008 per cent or less of chlordan. However, in male rats occasional liver cell inclusion bodies were seen in those fed at the level of 0.004 per cent of chlordan. These inclusion bodies were more numerous in animals given 0.008 per cent or more of chlordan in their diets. A few inclusion bodies were found in the liver cells of but one of a group of female rats fed on a diet containing 0.016 per cent of chlordan, while they were readily found in the livers of all of a group of females fed on diets containing 0.032 per cent of chlordan.

These authors noted that no necrotic or degenerating cells were present. Other organs and tissues showed no changes that could be ascribed to chlordan.

Autopsies on rats maintained for two years on diets containing 150 and 300 mg./kg. showed significant enlargement of the liver and kidneys in addition to pulmonary edema, hemorrhage and congestion. Histopathologic changes were noted in the liver marked by liver cell enlargement in the centrolobular areas, with increased oxyphilia and hyalinization of the cytoplasm, and bile duct proliferation. Centrolobular necrosis was extensive. Glycogen was almost entirely absent from the midzonal and periportal areas and was greatly reduced in the centrolobular region. The general picture was that of inanition. Myocardial changes consisted primarily of necrosis of the myofibrils. Hypertrophy of the medulla with some atrophy of the cortex were noted in the adrenal glands. Necrosis of some cells in the central area of the spleen was also noted. The duodenum showed localized areas of mucosal and submucosal degeneration (173).

In goats and sheep, the autopsies revealed pulmonary edema and focal areas of pulmonary emphysema. The right side of the heart was distended. The small intestine was congested and hemorrhagic with mucosal inflammation. The liver was hyperemic and swollen. The kidneys showed irregular hemorrhagic areas on the cut surface continuous with the cortex and medulla. The blood vessels of the meninges and brain were distended. An excess of cerebrospinal fluid was noted. The brain and spinal cord were slightly softened when sectioned (178) (153).

Histologically, endomyocarditis was noted. Fatty changes in the liver were described. Congestion of the meninges, brain and spinal cord was usually present. Focal miliary softening, necrosis of neurons, perivascular edema,

infiltration of round cells, and neuronophagia were constant findings (153).

Investigation of the Effects of Human Exposure to Chlordan: The published data regarding the effects of chlordan on human beings are scanty. Princi and Spurbeck (179) examined 34 persons engaged in the manufacture of chlordan, aldrin, and dieldrin who had been so employed from 11 to 37 months and had every opportunity to be exposed to these insecticides. These authors concluded that, under the conditions of their study, none of the organ systems of these men seem to have been affected by the concentration of these substances encountered in the air of a manufacturing and formulation plant (more than 5 mg. per cubic meter of air) for periods from one to three years.

No other published work could be found which deals with the examination of workers exposed to chlordan.

Poisoning Attributed to Chlordan in Human Beings: Only three cases of poisoning in human beings attributed to chlordan could be found in the literature to date.

Case No. 1: (180). Brief mention is made of one person who was subjected to contact of the skin with a 25 per cent solution of chlordan (equivalent to 30 grams of technical chlordan), who developed symptoms within 40 minutes and died before medical aid could be obtained.

Case No. 2: (181). A 15 months old female child drank perhaps a mouthful of chlordan suspension (one tablespoonful wettable powder in one quart water) some of which the child spat out. Within 2-3 minutes of ingestion the mother had induced vomiting with sodium bicarbonate and gagging. Three

hours later, the child developed generalized tremor, incoordination and ataxia. This was followed by convulsions which were described as having a tonic phase which alternated with short clonic phases. The convulsions were controlled by ethyl chloride inhalation 40 minutes later. Examination revealed dilated pupils which reacted to light, flushed face and irregular respirations. The child developed risus sardonicus, opisthotonus, bilateral ankle clonus, generalized hyperactive reflexes, Convulsions recurred about 5 hours after ingestion of the chlordan solution, and were only controlled after the administration of 0.345 grams of sodium amobarbital intravenously and 0.06 grams subcutaneously. Oxygen and parenteral fluids were given. After 16 hours, the child roused. Examination at that time revealed irritability, dilated pupils and ataxia. On the second day there was moderate ataxia and irritability. These finally disappeared after 2-3 weeks. The laboratory findings were within normal limits. It was estimated that the dose of chlordan that was ingested did not exceed 100 milligrams.

Case No. 3: (182). A 33 year old white woman complained of nausea, vomiting and cough. She had had a mildly acute upper respiratory infection 2-3 days before the onset of the presenting complaints. She entered her apartment, which had been sprayed with 1 or 2 per cent chlordane in oil base several hours before. She went to bed without airing the rooms and arose approximately 5½ hours later with a severe cough and a bout of vomiting. She had a history of "binge" drinking. The only positive physical finding was a smooth, firm, slightly tender liver three finger-breadths below the costal margin. After a week of hospitalization and symptomatic treatment she recover-

ed. The observers thought that this patient showed manifestations of poisoning due to the inhalation of an insecticide spray containing chlordan. The authors believed that the described involvement of the lungs and liver was consistent with the diagnosis of chlordan intoxication as demonstrated in animals.

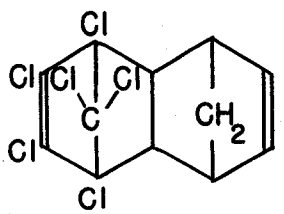
Heyroth (183) found it necessary to criticize the last case as not being a true case of chlordan intoxication. He is quite willing to accept the second case as being a bone fide case of chlordan intoxication.

One would not readily accept case No. 3 as being an example of poisoning due to chlordan. One could possibly consider very seriously the diagnosis of liver disease as a complication of alcoholism with greater justification than the stated diagnosis. It would appear that the authors made a poor choice of a title for this paper.

ALDRIN:

A - Physical and Chemical Data

This compound was developed by The Julius Hyman Company in 1949 under the code name of Compound 118. It was also known as octalene. Chemically, it is 1,2,3,4,10,10-hexachloro-1,4,4a,8,8a-hexahydro-1,4-endo,exo-5,8-dimethanoaphthalene with the empirical formula of $C_{12}H_8Cl_6$. The structural formula has been written as



From the planar representation there appear to be four sterioisomers (184). The recrystallized compound contains not less than 99 per cent of the above described compound which is available as the reference standard. The commercial form should contain not less than 95 per cent of the compound and not more than 5 per cent of related chlorinated hydrocarbons with insecticidal activity. The melting point of the commercial form should not be less than 90 degrees centigrade (185).

Physically pure aldrin is a white crystalline solid which is substantially odorless at room temperature but which has a mild pine-like odor when warmed. The melting point has been given variously as 104-104.5 degrees C., 100-102 degrees C., and 100-103 degrees C. It is practically insoluble in water. The solubility of aldrin in various organic solvents is given in the following table:

Solvent	Solubility at 30 degrees C. grams per 100 ml.
Methanol	9
Benzene	350
Acetone	159
Hexane	98
Base Oil (deobase)	89
Water	Insoluble

Data from Lidov et al (184)

Aldrin is readily compatible with most agricultural chemicals such as fertilizers, herbicides, fungicides and insecticides since it is stable in organic and inorganic alkali and hydrated metallic chlorides. Furthermore, under conditions of practical use aldrin appears to be unaffected by acidic reagents.

Aldrin may be formulated readily as a wettable powder, as an emulsifiable concentrate or an oil solution, and as low percentage dusts. Frear (9) lists the types of formulations offered commercially.

Analytical methods are avilable which lend themselves to the detection of aldrin in small quantities. Most of the methods are not specific for aldrin but are very useful if it is known that aldrin alone is present in the sample.

1. Colorimetric Method of Danish and Lidov (166).

If aldrin is treated with phenyl azide, aldrin-phenyldihydrothriazole is formed. Then, if this compound is treated with a dinitrophenyl diazonium salt in a strongly acid medium (hydrochloric acid), a compound of an intense red color is produced. In the final 10 ml. aliquot, 10-40 micrograms may be determined readily with the spectrophotometer. Chlordan, DDT, and methoxychlor, BHC, toxaphene and dieldrin do not interfere. Dimalone, an insect repellant, and Octacide 264, a pyrethrum synergist, do interfere. The waxy residues extracted from crop samples also interfere with the analysis.

2. Total Chlorine Method (187).

This is not a specific method for aldrin. However, it is useful when aldrin is known to be present alone. The oily residue is saponified and freed of resulting alcohol and salts of organic acids by dissolving the whole in Skellysolve B (hexane) and extracting with water before a chromatographic clean-up.

a. The sample is burned in a stream of air or oxygen in a quartz tube or lamp.

b. Hydrochloric acid is removed from exhaust gases by scrubbing with a dilute alkaline solution.

c. Microgram quantities of chloride ion found in the scrubbing solution may be determined by ampometric titration with a very dilute solution of silver nitrate.

This method is sensitive to 1 microgram if reduced-scale apparatus is used.

3. Bioassay Method of Sun and Sun (163).

This method is not specific. If it is known that the sample contains only aldrin, concentrations as low as 0.1 ppm. can be determined.

4. Microdetermination of aldrin and dieldrin by infrared spectroscopy (188).

Infrared spectrophotometric methods have been developed which utilize the 8.48 micron absorption peak of aldrin and 10.98 micron absorption peak of dieldrin. It is thought that DDT, lindane, toxaphene and chlordan would not interfere if these substances were not present in greater concentration than either aldrin or dieldrin.

5. Improved Danish-Lidov Method (187).

b. Comparison of Insecticidal Activity of Aldrin and Dieldrin and Certain Other Insecticides.

Relative Toxicity of Aldrin and Dieldrin to Insects

Insect	Chlordan	Aldrin	Dieldrin	DDT	Toxaphene	Gamma-BHC
Housefly	10	40	120	3	2	40
German Roach	10	60	300	.	.	30
American Roach	10	60	60	.	.	10

Relative Toxicity of Aldrin and Dieldrin to Insects - continued

Insect	Chlordan	Aldrin	Dieldrin	DDT	Toxaphene	gamma-BHC
Black Carpet Beetle	10	70	..	.	.	..
Milkweed Bug	10	140	96	.	.	45
Squash Bug	10	160	..	.	.	..
Confused Flour Beetle	10	68	200	.	.	30
Differential Grasshopper	10	40-50	70	.	.	..
Fall Webworm	10	100	..	12.5	.	..
Imported Cabbage Worm	10	40	..	6.7	3-4	..
Chinch Bug	10	80	80	.	.	40
Plum Curculio	10	60	..	.	.	..
Red Spider Mite	-	-	≠	-	-	-
Mexican Bean Beetle	-	-	≠	-	-	≠

(Using technical chlordan as standard of comparison - base value 10)

LD_{50} Values on Houseflies (Kearns, Weinman, & Decker)

(Based on topical application of some chlorinated compounds dissolved in 95 per cent ethyl alcohol. Values expressed as micrograms of toxicant per gram of fly weight. Tests done by W.N. Bruce, Ass't. Entomologist, Illinois Natural History Survey.)

Compound	LD_{50} gamma
DDT	20.5
Chlordan	4.0
gamma BHC	2.9
Aldrin	1.6
Dieldrin	1.1

LD_{50} Values for Single Compounds Tested for Contact and Stomach-Poison Effect Against M. differentialis Adults. (Weinman and Decker).

(Expressed as micrograms of toxicant per gram of weight of grasshopper)

Compound	Contact	Stomach poisons
DDT	9380	2579
Toxaphene	61	91.5

Compound	Contact	Stomach Poisons - continued
Chlordan	9.8	12.0
gamma BHC	3.4	6.7
Aldrin	1.8	2.3
Dieldrin	1.4	3.7

Kearns, Weinman, and Decker (189) state that their tests indicate the following order of decreasing toxicity: dieldrin, aldrin, heptachlor, gamma hexachlorocyclohexane, chlordan, toxaphene and DDT. This belief is based on observations of ten species of insects and they believe this represents, in general, the order of their relative toxicities.

Aldrin, like chlordan, exhibits residual effectiveness under field conditions for somewhat less than 3 weeks (184). Even when applied at the rate of five pounds per acre, leafy material so treated exhibits only a slight insecticidal effect after 3 weeks. Aldrin, therefore, is in the class of materials which exhibits pronounced initial toxicity but relatively short residual action. On the other hand, dieldrin shows pronounced initial-toxicity with a persistence comparable to DDT.

Per cent Mortality of Houseflies

(24-hours after a 30-minute exposure to a deposit of 50 mg. per sq. foot of some chlorinated insecticides at various intervals after application. Each figure based on 3 replicates of 25 flies each. Compounds deposited on glass plates - data from results of tests made by W.N. Bruce and G.F. Ludvik.)

	Age of Residues , Days					
	5	14	21	28	35	51
Compound	Per cent Mortality					
Chlordan	93.4	58.9	1.8	-	-	-
Aldrin	83.8	30.0	17.9	4.6	-	-
DDT	73.7	41.1	19.1	11.5	-	4.3
Dieldrin	100.0	100.0	-	95.7	71.1	64.3

Residual Toxicity of Chlorinated Insecticides to Adult Male German Roaches (1 mg/1000 sq. cm. deposits at various intervals after application. Percentage dead and moribund after periods of 24 and 48 hours of exposure.)

	Age of Deposit, Days											
	1		7		21		28		42		49	
	Period of Exposure, Hours											
	24	48	24	48	24	48	24	48	24	48	24	48
Compound	Percent Dead and Moribund											
Chlordan	100	100	24	92	0	0	0	0				
Gamma BHC	100	100	14	18	0	0	0	0				
Aldrin	100	100	22	100	0	0	0	0				
Dieldrin	100	100	100	100	100	100	100	100	46	100	24	90

(DDT not included because inactive at dosage level tested)

C - Toxicity of Aldrin - Experimental

Published data regarding the toxicity of aldrin for animals are not plentiful.

Immediate Toxicity: Treon, Gahegan and Coomer (190) determined the LD_{50} oral dosage of aldrin for white rats, using samples of aldrin manufactured by different processes; samples of different grades of purity; and samples of aldrin given in various solvents. Their data are tabulated below:

Immediate Oral Toxicity of Aldrin for White Rats

Compound	Solvent	LD_{50} mg./kg. (expressed in terms of 100 % Aldrin)
Thermal Aldrin - 60	Ultrasene	78.8
Straus Aldrin - 60	Ultrasene	26.3
Technical Crystalline Thermal Aldrin	TS-28-R (a toxic solvent)	19.8
Technical Crystalline Thermal Aldrin	Kerosene	71.3
Recrystallized Aldrin	Kerosene	66.3
Recrystallized Aldrin	Peanut Oil	48.3
Thermal Aldrin as Wettable Powder	Peanut Oil	41.8

Borgmann, Kitselman, Dahm and Pankaskie and Dutra (191) gave a solution of aldrin in corn oil in one oral dose to each of several animals of several species. Their findings are summarized as follows:

Immediate Oral Toxicity of Aldrin in Corn Oil

Species	LD_{50} mg./kg.
Mice	44
Male Rats	49
Female Rats	38
Guinea Pigs	33
Rabbits	50-80
Dogs	65-95

From the foregoing data it is apparent that the median lethal oral dose of aldrin is approximately 45-50 mg./kg.

Intravenous LD_{50} dose of Aldrin: Treon et al (190) found that the

minimum lethal dose of recrystallized aldrin, when given intravenously as an olive oil-saline emulsion to female rabbits, lay between 5.0-10.0 mg./kg.

Dermal Application of Aldrin: Lehman (100) stated that aldrin is approximately10 times more toxic when applied upon the skin than when ingested. Originally Barnes (70) was in apparent agreement with this statement, which was based on the observation that the daily application of aldrin in the dose of 5 mg./kg. upon the skin of the rabbit will cause death in seven to ten days. However, oral doses smaller than this given daily to rabbits will also produce death. Barnes (192) carefully points out that there is no satisfactory evidence for the generalization that aldrin is more toxic when applied upon the skin than when given orally.

Subacute and Chronic Toxicity: Borgmann, Kitselmann, Dahm and Pankaskie and Dutra (191) studied the subacute and chronic effects of aldrin in rabbits and rats. Five groups of rabbits were observed over the period of 90 days during which they were fed on diets containing 0.625, 1.25, 2.5, 5.0 and 10.0 mg. of aldrin per kg. of body weight, respectively. The groups given aldrin in dosages of 0.625 and 1.25 mg./kg. daily, respectively, tolerated these amounts without observable effects. All of the rats given 2.5 mg./kg. or more died except one fed at the 2.5 mg./kg. level.

The effects of feeding various amounts of aldrin to white rats over periods of time ranging from 90 days to 2 years, were also studied. More deaths occurred among the rats fed on diets containing 150 ppm. for 90 days than among the control animals. All rats maintained for 90 days on diets

containing 75, 5, 2, or 0.5 ppm. survived.

In the nine-months' experiments, the mortality among rats fed at levels of 45, 25, 15, or 5 ppm. did not differ significantly from that in the control group.

In the long-term study, it was noted at 16 months that the mortality was greater among rats fed 150 or 100 ppm. than among the controls. There was no difference in the mortality between the control group and the rats fed diets containing aldrin at levels of 50, 10, or 5 ppm.

Lehman's report (35) on the experiments on rats indicates that when aldrin was fed for 16 weeks in different concentrations, the lowest level which produced gross effects was 50 ppm.; the highest level which did not produce gross effects was 25 ppm.; tissue damage was noted at this level also. Dieldrin gave comparable results.

The length of survival of dogs given insecticides dissolved in corn oil in capsules daily on each of 5 days per week is shown below:

	Daily Dose mg./kg.	Number of Animals	Number of Deaths	Earliest Death in Days	Last Death Days
Aldrin	5	2	2	21	22
	2	2	2	24	178
	1	2	2	109	344
	0.5	3	0	1 animal living at 277 days; 2 at 180 days	

Treon, Dutra, Shaffer, Cleveland, Wagner and Gahegan (193) studied the effect of feeding aldrin to dogs over periods ranging from a few days to 9 months. They concluded that aldrin uniformly killed puppies when fed at 10 ppm., 25 ppm., or 50 ppm. in the diet.

Treon and Borgmann (194) reported that rats which had been fed aldrin or dieldrin to the extent of 5 ppm. in the diet for 7 to 18 months and were then offered water but no food, showed a slightly higher incidence of central nervous system reactions than that observed among the control animals. They believe that the effect of aldrin was slight, since these reactions appeared only shortly before the animals approached the end of their lives. The mortality rate among the test group and control animals was not significantly different.

Fitzhugh and Nelson (195) state that, from their limited data regarding the chronic toxicity of aldrin and dieldrin in the rat, these insecticides are as toxic as chlordan. In this connection, these authors have concluded that chlordan and beta-BHC induced the greatest damage to the rat in the long-term experiments.

Brown (7) also states that aldrin is reported to have a high level of chronic toxicity, producing liver damage.

Kitselmann, Dahm, and Borgmann (196) studied the acute, subacute, and chronic toxicity of aldrin (recrystallized, 99%) in larger animals. These included steers, heifers, dairy cows, and sheep. For the most part, these were feeding experiments in which three methods of introducing aldrin in the food were used. These investigators prepared the dosages as follows: (1) alfalfa was treated in the field with 0.5 pound of aldrin per acre. The alfalfa was cut 8 days after the application of aldrin, dried and baled, and then stored in a barn. The estimated residue was determined by biologic assay with houseflies. Not more than 8 parts aldrin per million were found

on the treated alfalfa at any time. (2) Known amounts of aldrin dissolved in corn oil were added to the daily ration. (3) Animals were given solutions of aldrin in corn oil by direct oral administration. The results are tabulated below:

Toxicity of Aldrin Administered to Large Animals

Animal	Method of Administration	Number of Doses	Symptoms	Subsequent Doses	Death
1. Ewe	Single daily dose of aldrin in corn oil mixed with forage 6 mg./kg. body wt.	15	Refused to eat on 15th day.	Given as drench-13 days.	30 days after first dose.
2. Steer	Fed aldrin at 7.5 mg./kg./day (approximately 19 gms. consumed).	8	Hyperirritability; convulsions 8th day.	2 days	10 days
3. Heifer	Fed 4 mg./kg./day (approx. 17.7 gms. consumed).	7	Hyperirritability (?)	14 days	21st day convulsions.
4. Jersey Cow	Fed 2 mg./kg./day (approx. 22 gms. consumed.)	18	Emaciation and decreased milk production.	23 days	41 days

Analyses of the tissues from the animals listed in the above table showed aldrin to be present in concentrations ranging from 50 ppm. down to less than 1 ppm. The body and peritoneal fat contained the largest amounts of aldrin. Histologic examination of the tissues of these animals revealed degenerative changes in the kidneys and liver.

There seems to be confusion as to what is meant by acute toxicity. Usually, the term acute toxicity as used carelessly by experimental workers,

relates to the effect that a compound produces when given in a single dose or in multiple doses over periods of 24 hours or less (34). A convenient expression of this acute, or immediate toxicity of a substance is the LD_{50}. It would appear, then, by this definition that the data just presented belong in another category. (It would be wiser and more in keeping with good linguistic practice if the terms acute, subacute and chronic, as categories of toxicity, were abandoned completely.)

These authors further concluded that neither sheep nor cattle fed on treated hay showed any toxic effects during 169 and 213 days, respectively. Furthermore, less aldrin than 4 ppm. was found in the liver and 2 ppm. in the body fat of the ewe which had eaten the treated hay for 169 days. In the examination of the cattle fed on the treated hay for 213 days, less than 1 ppm. was found in the fat biopsies. Less than 1 ppm. was found in the liver, the kidney, brain and the body fat of a heifer fed on the treated hay for 169 days. The omental fat of 3 heifers fed at levels of 2.0, 1.0, 0.5 mg./kg. per day for 64 days showed less than 1 ppm. after 100 days of grazing in aldrin-free pastures. Cattle in the sprayed hay group showed the same average gain in body weight as the control group. Milk from a Jersey cow fed on treated hay for 91 days showed no aldrin.

D - Tissue Storage and Dissipation

Borgmann et al (191) found small amounts of aldrin stored in liver, kidney, muscle and brain tissue. The greatest concentration of aldrin was found in the combined omental and perirenal fat of rats. Storage in the fat occurred among all rats fed diets containing 0.5 ppm. up to 50 ppm daily.

In the group given 0.5 ppm. in the diet for 3 months, 2 ppm. was found in the fat. The fat of an animal kept on a 10 ppm. diet for 23 months contained 15 ppm. Approximately 100 ppm. was found in the fat of rats given a diet containing 50 ppm. over the period of 6 months. Aldrin disappeared from the fat of rats in 3 to 9 weeks following the initiation of an aldrin-free diet.

E - Pathology

There seems to be fairly general agreement that the chief structural changes that can be attributed to prolonged ingestion of aldrin occur in the liver (195) (191) (66). These changes are similar to those seen among rodents exposed to other chlorinated hydrocarbon insecticides (66). Slight degenerative changes of the cells of layer IV of the cerebral cortex have also been described. Degenerative changes were found in the basal ganglia and cerebellum as well (191). Kidney damage is also reported (196).

F - Signs and Symptoms of Intoxication

Massive doses of aldrin administered to rats produce tremors, convulsions and death (191). After lesser but still lethal dosages, the animals become lethargic, vomit, develop anorexia and become emaciated. Central nervous system reactions such as hypersensitivity to external tactile and auditory stimuli, tremors, twitching, ataxia and convulsions are prominent (191) (190). Finally coma and death occur.

G - Intoxication in Human Beings

There is only one published case of poisoning due to aldrin in the literature. Spiotta (197) describes the case of a 23 year old white farmer

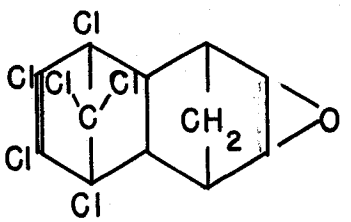
who drank an aldrin mixture equivalent to 25.6 of aldrin per kg. of body weight. Despite gastric lavage, convulsions began within 20 minutes and were finally controlled with large doses of barbiturates. On the second day, the patient developed hematuria and albuminuria, which persisted for 18 days. An electroencephalogram revealed generalized cerebral dysrhythmia which disappeared within 5 months. Liver damage as such, was not detected in this patient. The only abnormality was an elevated icteric index of 15. Symptomatic treatment consisted of removing the poison from the portal of entry immediately, control of convulsions with barbiturates, and supportive treatment to anticipate renal and hepatic injury. This patient was observed carefully. Recovery was reported as being complete.

Princi and Spurbeck (179) studied 34 persons engaged in the manufacture of chlordan, aldrin and dieldrin who had been working in this process from 11 months to 37 months. These investigators could find no evidence of any deleterious effects on the central nervous system, the liver, the kidneys, or the hemapoietic system.

Dieldrin

A - Chemical and Physical Properties

Dieldrin is produced by the oxidation of aldrin with per acids. This compound was developed in 1949 as an insecticide by The Julius Hyman Company under the code name of Compound 497. It has also been known as Octalox. Chemically, dieldrin is 1,2,3,4,10,10-hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,8a-octahydro-1,4,5,8-dimethano-naphthalene. The empirical formula is $C_{12}H_8Cl_6O$. The structural formula is written as



In its pure state, dieldrin is a white crystalline solid which is odorless. The melting point has been reported as 173 degrees C., and also as 175-176 degrees C. Its solubility in various solvents is as follows:

Solvent	Solubility of Dieldrin Grams/100 Gms.	
	26 degrees C.	0 degrees C.
Methanol	4.9	3.4
Acetone	54.0	35.4
Benzene	75.0	36.9
Hexane	7.7	2.5
Base Oil (Std. oil 10)	4.3	1.3
Water	Insoluble	

Data from Lidov et al (184)

Dieldrin is a quite stable oxide. Although it is decomposed by strong acids, it is stable to weak acids and alkalis. Under practical conditions of use, dieldrin remains insecticidally active under both acidic and alkaline conditions. It is barely volatile, as is indicated by its residual toxicity. Its persistence is comparable to that of DDT.

Specific analytical methods which will determine micro-quantities of dieldrin satisfactorily in all samples are not available as yet. Under certain conditions it may be possible to make determinations of micro-quantities of dieldrin by infrared spectroscopy (188). How practical this is remains to

be determined.

The relative toxicity of dieldrin to insects, the LD_{50} values for houseflies, the LD_{50} values for contact and stomach poison effect, and its residual effectiveness are shown in the tables in the section on the chemistry and activity of aldrin.

B - Toxicity of Dieldrin - Experimental

Immediate Toxicity of Dieldrin: Treon, Gahegan and Coomer (190) report the oral LD_{50} of dieldrin in olive oil administered to white rats by gavage to be 38.3 mg./kg.

Borgmann, Kitselmann, Dahm and Pankaskie and Dutra (198) found the oral LD_{50} of dieldrin in corn oil in several species of animals to be as follows:

Species	LD_{50} mg./kg.
Mice	38
Female rats	43
Male rats	47
Guinea pigs	49
Rabbits	45-50
Dogs	65-80

Others (100) report that the oral LD_{50} dose of dieldrin administered to white rats is approximately 65 mg./kg. of body weight.

Intravenous LD_{50} Dose: Treon et al (190) report that the LD_{50} dose of recrystallized dieldrin, when given intravenously as an olive oil-saline emulsion to female rabbits, lay between 2.5 and 5.0 mg./kg.

Dermal Application of Dieldrin: Hayes, Ferguson and Cass (199) applied various formulations of dieldrin to the skin of rats, mice, hamsters, guinea pigs, rabbits, dogs, cats and monkeys. They report the data from the experi-

ments on rats. It was found that a single dose of 400 mg. of the technical powder of dieldrin/kg., when applied to the skin of white rats, was always lethal. The 25 per cent concentrate in the same dosage gave 97 per cent mortality. A dose of 10 mg./kg. of a 0.62 per cent emulsion of dieldrin applied on somewhat more than 100 days gave a mortality of 30 per cent. During the same period 20 per cent of the control group died. In likening the conditions of the experiment involving prolonged applications on the skin to those which may obtain in the field use of the insecticide, these authors concluded that a single contact of the skin of a man with 100 cc or more of a 25 per cent formulation of dieldrin may be dangerous if it were not washed off promptly.

Lehman (100) has stated that dieldrin has the rare property of being ten times more toxic when applied on the skin than when ingested. It is now recognized that this generalization is unjustified and that there is no convincing evidence that such is the case.

Subacute and Chronic Toxicity of Dieldrin: Lehman (35) reported that rats on fed diets containing various amounts of dieldrin for 16 weeks suffered gross effects from the use of that containing 50 ppm., while no gross effects were noted among rats fed at the level of 25 ppm. in the diet. However, tissue damage was noted. This last amount was the lowest level fed to the rats.

Lehman (35) reported dieldrin was dissolved in corn oil and administered to dogs in capsules on 5 days per week, with the following results:

Survival Time of Dogs Fed Various Daily Doses of Dieldrin in Corn Oil

Daily Dose mg./kg.	Number of Dogs	Number of Deaths	Earliest Death Days	Last Death Days	Fate of Surviving Animals
10	2	2	16	17	
5	2	2	28	35	
2	2	2	22	35	
1	2	2	83	300	
0.5	4	2	14	201	2 animals living at 275 and 180 days, respectively.

Borgmann et al (198) found that after 90 days, no significant difference in mortality occurred among groups of rats fed on diets containing 125, 50 or 25 ppm. from that noted among the control rats. They state that rabbits appeared to be more susceptible than rats to diets containing dieldrin.

These authors found that a greater incidence of mortality occurred among female rats fed for 12 months on diets containing 150 ppm. than among controls. However, rats maintained on diets containing dieldrin at the level of 100 ppm. or less survived as long as the control group. It was also noted that the mortality was not greater among males fed at the level of 150 ppm. or less than among the corresponding controls.

Bundren, Howell, and Heller (200) found that dieldrin produced symptoms of intoxication in rabbits when applied on their skin once per week in the dose of 70 mg./kg. If a dose of less than 30 mg./kg. was applied upon the skin, no symptoms occurred. They also noted adverse effects when mature rabbits were fed dieldrin repetitively at the dosage level of 60 mg./kg. Similarly, rats died when fed at dosage levels of approximately 150 mg./kg. On the other hand only 2 of 11 rats dipped in 0.125, 0.187 and 0.250 per cent dieldrin spray solutions died.

Treon et al (193) noted that dieldrin incorporated in diets killed all dogs fed at levels of 25 ppm. or 50 ppm. However, dogs survived for 9 months when fed at the level of 10 ppm.

Treon and Borgmann (194) fed rats dieldrin to the extent of 5 ppm. in the diet for from 7 to 18 months. Food was then withdrawn and only water was offered to the rats. These animals exhibited a slightly higher incidence of central nervous system reactions than that observed among the controls. No significant difference in mortality rates was noted between the test animals and the controls. These investigators believe that the effect of dieldrin to be minor because the reaction appeared only shortly before the animals came to the end of their lives.

Toxicity Experiments with Large Animals: Radeleff, Bushland and Claborn (40) state that a dieldrin spray in concentrations of 0.5 per cent, when applied to cattle 3 times at intervals of 2 weeks produces symptoms of poisoning. A single spray in which the concentrations of dieldrin were 0.25, 2.00, 3.00 and 4.00 per cent, as applied to week old calves, cattle, sheep, hogs and goats, respectively, was not tolerated.

C - Storage and Excretion of Dieldrin

Borgmann et al (198) found small amounts of dieldrin stored in the liver, kidneys, muscle and brain tissue of animals. Like aldrin, the greatest concentration of dieldrin was found in the combined omental and perirenal fat of rats.

Storage of Dieldrin in Fat of Rats

Dieldrin in Diet ppm.	Duration of Feeding months	Dieldrin in Fat ppm.
5	7-11	15-19
10	6.5	43
15	7-11	30-50
20	6.5	68
50	11	35
50	6.5	56
75	7	18.5

It is apparent that the storage rate of dieldrin is somewhat greater than that of aldrin.

Radeleff et al (40) fed dieldrin to large animals at levels of concentration of 25 ppm. in each item of food for 28 to 56 days. Their findings are tabulated as follows:

Storage of Dieldrin in Fat of Animals Fed 25 ppm. Dieldrin in Each Item of Food

Animal	28 days feeding ppm. in fat	56 days feeding ppm. in fat	32 weeks after feedings discontinued
Yearling Hereford	75	74	Detectable
Delaine Sheep	43	69	Detectable

Bundren et al (200) fed rabbits dieldrin in the amount of 60 mg./kg. They found that the liver and kidneys of all animals so fed contained dieldrin. The amount recovered in the organs was usually in proportion to the amount fed per week and not to the duration of feeding.

These investigators analyzed milk obtained from cows sprayed with dieldrin for external parasite control and found dieldrin in the maximum concentration of 5.9 ppm. and subject to a gradual decrease even when the spray-

ing was continued.

Radeleff et al (40) found that cattle which were sprayed twice at 3-week intervals with 0.25 per cent of dieldrin had 17 ppm. in the fat 3 weeks after the second application. After 8 applications, 14 ppm. of dieldrin were found in the fat. It required 13 weeks for the residue of 14 ppm. to disappear.

Dairy cattle sprayed once with 0.5 per cent of dieldrin showed a maximum of 7 ppm. in the milk on the third day following that on which they were sprayed. The milk was practically free of dieldrin 21 days after the day of spraying (196).

D - Pathology

The only structural changes that can be attributed to prolonged ingestion of dieldrin occur in the liver (198). These changes were noted in rats given a diet containing the insecticide at levels of 10, 20 and 50 ppm. Strangely, none of 5 animals given dieldrin in the diet at 25 ppm. showed these changes, which were similar to those noted and reported as being characteristic of chlorinated hydrocarbon insecticide poisoning (66).

Fitzhugh and Nelson (195) noted, from their limited data, that they considered dieldrin as toxic to rats as chlordan. They observed varying degrees of hepatic cell enlargement with peripheral migration of granules.

Hayes, Ferguson and Cass (199) found no specific lesion or lesions at autopsy to account for the death of the animals poisoned with dieldrin in their experiments.

E - Symptoms

Among rats, the signs of intoxication which were observed regularly included anorexia, loss of body weight, hyperactivity, hypersensitivity to auditory and tactile stimuli, salivation, hyperpnea, nasal discharge, convulsions and coma (190) (198) (199). It has also been noted that of the various signs of poisoning, only long continued coma and excessive weight loss are incompatible with life (199).

The use of barbiturates in treating severe convulsions in dogs and monkeys gave encouraging results. Sedation reduced, but did not at once eliminate, hyperexcitability, incoordination and convulsions. The animals were able to eat and gradually improved their nutritional state. The effective procedure is to use sufficiently large doses to calm the individual animal and even to make it sleep the normal amount (199).

F - Toxicity of Dieldrin to Man

No reports of the actual poisoning of man by dieldrin have appeared in the medical literature to date. Princi and Spurbeck (179), mentioned previously in this paper, carried out a survey on employees of a plant manufacturing chlordan, aldrin and dieldrin. No abnormalities which could be attributed to exposure to these insecticides were found among these men.

One unpublished clear cut instance of intentional ingestion of a compound containing dieldrin is summarized (201).

A 44 year old white male drank approximately 50 ml. (probably less) of "Cupridor" which is reported to contain 75 per cent of copper carbonate (33.75 per cent Cu.), 3.75 per cent of dieldrin and an inert carrier. Approximately 2 hours after drinking this material, the patient began to vomit

CHAPTER VI

A review of part of the evidence for the concept of the delayed or cumulative toxicity, for man, of the chlorinated hydrocarbon insecticides seems to be warranted, since it appears that it is not known well enough to be common knowledge among many physicians.

At the outset, it would be well to recall some of the insecticides included in the group that is under consideration. First, there is DDT (1944), and its analogues, methoxychlor and DDD (TDE); then benzene hexachloride (1945), and its gamma isomer, lindane; toxaphene (chlorinated camphene); and finally, the Diels-Alder condensation products such as chlordane (1945), heptachlor, aldrin (1949) and dieldrin (1949). There are other compounds in these classes but the ones listed are those most commonly used in agriculture, in public health work, and around the household.

It should be stressed that these compounds are poisons, and, as has been indicated, are capable of producing, in experimental animals exposed to sufficient dosage, either physiologic effects or anatomic damage, or both. The LD_{50} ranges have been established for these compounds for various species of animals.

The median lethal oral dosages of the various compounds for the rat are listed in the following table, which has been adapted from J.R. Busvine (70). Thus, it is possible to have some concept as to how toxic one substance is as compared with another, providing the same species of animals is used throughout

and continued to do so for several hours. It was reported that he had eaten 2 meals of very fat pork, bread, beer and cidar within a 5 hour period prior to the attempt at suicide. He was not drunk.

The physician who attended this man 3 hours after the attempted suicide reported that the patient had severe abdominal cramps, pains in the face, arms and legs, and chest pain which was accompanied by cardiac arrhythmia (type not stated). Urobilinogen was found in the urine. Examination of the vomitus made clear the cause of illness. The patient was given morphine subcutaneously and confined to bed. Within 4 days all symptoms had subsided and the patient felt well. He refused further medical care and follow-up.

It is interesting to note in this case that, even after retention of the insecticide in the stomach for 2 hours, not enough was absorbed to produce the convulsions that might have been expected. Subjectively, at least, the patient felt well enough after 4 days to discontinue medical observation.

One might speculate regarding the effect of the copper content of this material on the absorption of dieldrin.

It appears unlikely that residual organ damage, with specific reference to the liver, would have occurred in this instance. It should be recalled that Spiotta (197) reported the complete recovery of a 23 year old white male who developed convulsions following the ingestion of aldrin. At no time could liver damage be demonstrated in this authenticated case of human intoxication.

A number of instances of human beings suspected of being poisoned with aldrin or dieldrin will be presented and discussed in the next section.

and the route of administration is the same.

Immediate Toxicity for Rats. Estimates of Median Lethal Oral Dosages (in mg./kg.) from Various Sources.

Aldrin	45 (Julius Hyman and Company)
Dieldrin	50-55 (Julius Hyman and Company)
Toxaphene	120-280(Parker and Beacher - 1947)
	60 (Lehman - 1948)
BHC	200 (Cameron - 1945)
	125 (Lehman - 1948)
	225 (Riemschneider - 1949)
Chlordan	225-250 (Ingle - 1947)
	500 (Lehman - 1948)
	200 (Riemschneider - 1949)
DDT	150 (Smith and Stohlman - 1944)
	800 (Cameron and Burgess - 1945)
	250 (Lehman - 1948)
	225 (Riemschneider - 1949)
DDD	2500 (Lehman - 1948)
Methoxychlor	greater than 6000 (Lehman - 1948)

As necessary as it is to have such data as the median lethal dose for various compounds, in this instance it is of greater practical importance to discover what happens when there is frequent exposure to small doses over a long period of time. The reasons for this are obvious. The widespread practice of using insecticides in agriculture is indicated by the figure cited by the Assistant Secretary of Agriculture, who stated that in 1948 farmers spent about 60 million dollars for these products. Today, the insecticide industry sells approximately 200 million dollars worth of their products, a considerable proportion of which are accounted for by the chlorinated hydrocarbon insecticides. Since many millions of pounds of the chlorinated hydrocarbon insecticides are produced and used each year, there is occasion for some concern as to what effects they might have on the workers engaged in

manufacturing and formulating these products, on the farmers who apply them to their crops, and on the consumers who eat the foods produced by these farmers.

Since the problem of frequent exposure to small doses of the chlorinated hydrocarbon insecticides over a long period of time is of immediate interest, we must look for a situation where these conditions might be met. DDT has had widespread use. There has accumulated a mass of information regarding its use, its occurrence in food, and its storage in tissues. Therefore, it would be convenient to use DDT as a prototype of the other chlorinated hydrocarbon insecticides.

The Food and Drug Administration examined a number of samples of fluid milk and certain of the other staple foods (202). The results obtained in 1949 are tabulated below.

Product	No. of Samples	Range of DDT in ppm.
Fluid Milk	120	Nil - 0.32
Evaporated Milk	37	Nil - 0.70
Wheat Flour	6	0.02 - 0.11
Corn Meal	3	0.04 - 0.07
White Bread	7	Nil - 0.11
Corn Grits	2	Nil
Butter	4	0.6 - 2.0
Lard	4	Nil - 0.8
Oleomargarine	8	Nil

In June 1951, another group of foods was examined for DDT. The following results were obtained:

Product	Number of Samples	Range of DDT in ppm.
Fluid Milk	50	Trace - 0.36
Evaporated Milk	6	Trace - 0.28

Product	Number of Samples	Range of DDT in ppm.
Meat - Beef	6	Nil - 2.4
Corn	6	Nil
Corn Oil	4	Nil
Butter	6	Nil - Trace
Lard	6	Nil - 1.4

In 1951, R.H. Robinson (203) chemist at the Oregon Experiment Station, Oregon State College, Corvallis, Oregon, presented the following data to the Delaney Committee during the hearings on the use of chemicals in food and cosmetics. The chart which Robinson presented shows the results of analyses of spray residues on some food crops in the condition in which they are eaten ordinarily.

Food Crop	Treatment	Spray Residue in ppm.
Apples (Hood River)	2 DDT sprays - 90% 2 DDT sprays - 40%	3.0 or less DDT 1.0 or less DDT
Pears (Medford)	2 DDT sprays - 85% 2 DDT sprays - 50%	3.0 or less DDT 1.0 or less DDT
Cherries (washed,canned)	Methoxychlor - 3 sprays or dusts	0.1-1.1 Methoxychlor
Prunes (fresh,harvest)	Methoxychlor - 50% wettable, 2 lbs./100 gallon.	0.34 Methoxychlor
Beans, string (fresh,frozen, canned)	DDT- 5% dust by ground duster and airplane Methoxychlor-5% as above	0-0.5 DDT 0-0.3 Methoxychlor
Corn (fresh)	DDT-5% dust	Negative for DDT
Peas (frozen)	DDT-3 and 5% dust	Negative for DDT
Head Lettuce - 4 in. size (fresh,harvest)	DDT-5% Methoxychlor-5%	0.07 DDT 0.05 Methoxychlor
Tomatoes (fresh,harvest)	DDT-3% dust	Negative for DDT

Robinson also presented data which show the approximate percentages of various foods eaten per person in the United States. These are based upon statistics obtained from the Western Canner and Packer (204) and the U.S. Department of Agriculture (205).

Approximate Percentages of Various Foods Eaten per Person in the United States.

Flour, cereal, bread, etc.	19.5	Beans	2.5
Meats, fish, poultry	11.9	Tomatoes	5.8
Fats, butter, oleo, etc.	5.6	Cabbage	2.4
Sugars, candy, jelly, etc.	5.7	Asparagus	0.3
Potatoes	20.5	Peppers	0.2
Potatoes, sweet	2.7	Cauliflower)	0.3
Peas	1.4	Broccoli)	
Corn	3.9	Spinach, Lettuce	0.3
Carrots	0.9	Apples	2.9
Beets	0.3	Pears	1.1
Pumpkin	0.3	Oranges	4.9
Onions	1.8	Grapefruit	2.5
Cucumber	0.6	Other fruits	1.7
	75.1 %		24.9 %
	Foods not contaminated with spray		Foods usually treated with pesticides

It can be seen that there are some discrepancies in the above table, for it has been shown in a previous table that flour, meat, butter and bread, may, at times, contain residual amounts of insecticide. However, it still serves as a rough approximation.

These data would seem to indicate that all people ingest some quantity of chlorinated hydrocarbon insecticide. One might inquire as to what happens to the insecticides which are ingested. There is no clear and complete answer to this question at this time. However, several bits of information are available.

Laug and his group (61) present data on the occurrence of DDT in human fat and milk. They refer to the storage of DDT in adipose tissue as an extremely

sensitive "biological magnifier" of trace amounts of ingested DDT. They arrived at this conclusion after it was demonstrated that 0.1 ppm. in the diet of rats leads to the storage of about 10 to 15 ppm. in their body fat. They then assumed that human beings also store DDT consumed in trace amounts, and thus, they used this "sensitive indicator" as a means of measuring the DDT exposure in the general population. They analyzed 75 samples of human abdominal fat which were obtained at autopsy, biopsy, or abdominal surgery.

The results of the analyses of human fat are given below.

Range of Concentration of DDT in 75 Samples of Human Fat

Range ppm.	Cases	Range ppm.	Cases
0.0	15	5.1 - 10.0	21
0.1 - 1.0	7	10.1 - 20.0	9
1.1 - 5.0	21	*Over 20.0	2

Average value equals 5.3 ppm.
* Actual values - 26 and 34 ppm.

Male and Female were equally represented. There was no significant difference between the sexes (p equals 0.05)

In addition, 32 samples of human milk were obtained from 32 different Negro women out-patients of a Washington, D.C. hospital. These samples were analyzed for DDT also.

Range of Concentration of DDT in 32 Samples of Human Milk

Range ppm.	Cases	Range ppm.	Cases
0.00	2	0.11 - 0.15	13
0.01 - 0.05	3	0.16 - 0.20	2
0.06 - 0.10	8	*Over 0.20	4

Average value equals 0.13 ppm. DDT
*Actual values - 0.25, 0.28, 0.42, 0.77 ppm.

If the results of the analyses of market milk as stated by the Food and Drug Administration are compared with the results of the analyses of human milk, they would be in the same range.

The results of the analyses obtained by Laug et al were interpreted to mean that traces of DDT in the average human dietary are of the order of 0.05 ppm. This is based on the assumption that man stores DDT in the body fat at the same rate as the rat. Their conclusion was that so far there is no evidence that this amount of DDT is harmful to human beings.

So far, data have been presented, some very rough and approximate and some quantitative, which are intended to show that (1) the chlorinated hydrocarbon insecticides are widely used; (2) usually, only trace amounts of these insecticides may be found as residues on certain fruits and vegetables, in dairy products, and in meat; (3) DDT, at least, may be found in human fat and milk in amounts which may bear some relationship to the amount of DDT absorbed and stored by whatever route.

In a sense, the situation indicated above is comparable to that involved in a prolonged feeding experiment. How can it be interpreted?

One medical author, Dr. Biskind, stated at a public hearing of a congressional committee that the observations dealing with the cumulative storage of the chlorinated hydrocarbon insecticides in body fat and their appearance in milk have been ignored or completely misinterpreted. He further charged that "the evidence regarding the toxicity of DDT and related compounds has been treated with disbelief, ignored, misinterpreted, distorted, suppressed, or subjected to some of the fanciest double-talk ever perpet-

uated".

This author, in at least two publications (206) (207) described the illnesses of several hundred patients which were allegedly due to chronic DDT poisoning. The symptoms which he listed included gastro-enteritis, nausea, vomiting, abdominal pain, coryza, cough, sore-throat, a feeling of constriction in the pharynx and substernal region, muscular weakness, extreme fatigue, stiff neck, intractable headache, herpes zoster and a variety of sensory disturbances.

The neurologic complications are interesting also. There may be unbearable emotional turbulence, excitement, irritability, anxiety, confusion, inability to concentrate, depression, apprehension, and forgetfulness. There may be feelings of tension in the extremities, and complaints of inability to keep the limbs immobile and free of fibrillary twitching of the muscles. There may be autonomic disturbances as sweating of the palms and tachycardia. The symptoms occur in waves which seem more likely to appear during periods of low blood sugar.

This catalogue of symptoms is impressive. However, the question remains, "Are these symptoms of DDT intoxication?" None of the diagnoses discussed in the Biskind papers can be said to be based on scientific evidence. The degree of exposure cannot be stated in most of the cases. There is no report of any careful and thorough investigation of any of the cases. The attempt to verify the causal relationship between DDT or any other substance and the symptoms noted was not made. The solvents have not been considered as possible etiologic agents in any of the cases. Finally, the symptoms are

so all-inclusive as to be meaningless in relation to any single syndrome.

For symptoms characteristic of chronic DDT intoxication in man, Biskind refers to a paper by Wigglesworth (48), who reported one case of what he chose to call DDT intoxication. The experience reported by Wigglesworth involved the evaporation of small quantities of DDT in acetone on the back of the hand, and the swabbing of the residual deposit with cotton wool soaked in acetone. There was another exposure to an acetone solution containing about 25 grams of DDT which was added to an inert dust and the mixture was kneaded with the hands for some minutes. It was reported that the subject experienced no reaction for the first day. "From 1 to 10 days later, a feeling of heaviness and aching developed in all the limbs, with weakness in the legs. Spasms of extreme nervous tension were described by the subject. There was some improvement during a holiday taken at this time, but on returning to work the condition deteriorated and three weeks later the perpetual aching in the limbs confined the patient to bed". The patient had not recovered fully one year later.

One cannot accept this observation as describing the picture of DDT intoxication in man. There is nothing clear-cut about this observation involving one subject to show a causal relationship between DDT and the symptoms.

Dangerfield (47) in an attempt to duplicate the above experiment, exposed three volunteers to a similar but considerably greater exposure to DDT. His conclusion stated that none of the volunteers suffered any irritant or toxic effect at all. In view of the negative results obtained and the

fact that Wigglesworth's case was one of symptoms and no signs, apart from muscular tremors, it seemed likely that the patient was suffering from an anxiety state and that DDT in acetone is not a particularly toxic preparation.

Another reference given by Biskind to substantiate his description of the characteristic symptoms of DDT intoxication is the experiment done by R.A.M. Case (209). In this experiment two men were kept in a very small chamber on two occasions to investigate the possibility of absorbing DDT through the skin. On the first occasion, the men sat on hard benches for 48 hours resting their bare backs against the steel walls treated with ordinary paint. The next occasion was two days later when the men re-entered the chamber, which had now been painted with a paint containing 2 per cent of DDT, and sat another 48 hours. During the second period of confinement, the subjects complained of aching limbs, extreme irritability, distaste for work, apparent mental incompetence, and violent joint pains on occasion. On the basis of Wigglesworth's report, Case attributed these symptoms to DDT intoxication. This was not a flawless experiment, since the DDT paint had not dried completely and the men complained of severe smarting of the eyes. Case commented that "This lacrimatory substance was more likely to have been the trace of chlorinated phenol added as a preservative than a product of DDT".

It seems clear that the symptomatology which Biskind attributes to chronic DDT intoxication has its support in one unproved case of DDT intoxication as reported by Wigglesworth.

To further confuse the picture, a report by Pottenger and Krohn (208) appeared recently which included brief summaries of the illnesses of ten patients taken from a file of over one hundred cases which these authors have treated in a period of one year as cases of chlorinated hydrocarbon insecticide poisoning. Their diagnosis is based on three findings: (1) characteristic symptoms; (2) evidence of liver pathology such as high blood cholesterol and high icteric index; and (3) presence of chlorinated hydrocarbon insecticides in fat obtained by biopsy. These are good criteria. However, the symptomatology is taken from Biskind's unsubstantiated reports, which are based on Wigglesworth's single observation, supported by Case's experiments which derive their diagnostic criteria from Wigglesworth's article. It is difficult to accept the presence of high blood cholesterol and high icteric index as indicating liver damage due to the absorption of chlorinated hydrocarbon insecticide. Lastly, the presence of chlorinated hydrocarbon insecticide in the body fat in itself merely means that there has been absorption and storage of these compounds in the body. It does not justify the diagnosis of intoxication.

So far, there are no published statistical studies which relate the concentration of chlorinated hydrocarbon insecticides in human fat to any histologic change in the liver which is characteristic of chlorinated hydrocarbon insecticide poisoning. Furthermore, there has not been reported any definite pattern of pathologic changes in the liver of human beings which can be correlated with chlorinated hydrocarbon insecticide absorption.

There is a lack of agreement among investigators regarding the exist-

ence of significant histologic alterations of the liver, much less the significance of it, in rats chronically exposed to low levels of DDT. Laug and his group (55) in 1950 reported the occurrence of minimal liver damage in response to the consumption of diet containing as little DDT as 5 ppm. On the other hand, Cameron and Chang (71) in 1951 failed to find liver damage at any level of dietary intake of DDT between 3.5 and 350 ppm.

So far, there are no published data to indicate that the amount of DDT stored in the fat of human beings is proportional to the degree of known exposure. However, in the rat there is evidence that this is the case (55).

It would appear that Pottenger's publication is open to the criticism that its author has taken for granted some principle which is equivalent to the result he seeks. In particular, this would apply to the list of human symptoms which it is assumed would be caused by prolonged exposure to small and unspecified amounts of the chlorinated hydrocarbon insecticides.

The human experience accumulated during the last war cannot be ignored. Several million people have been exposed to various amounts of DDT during delousing operations. The exposures in Naples and North Africa should be noted. In 1944 and 1945, the 820th Quartermaster Sterilization Company, U.S. Army, used over 37 tons of DDT in dusting over 2 million people for control of body lice (192). Until very recently, DDT was used extensively in Korea for the extermination of the body louse. No reports of DDT intoxication affecting these populations have appeared in the medical literature.

The criticism has been offered that DDT intoxication has not been looked for (208), and furthermore, that among those diagnosed as suffering from infectious hepatitis and homologous serum jaundice, there were those who were actually suffering from DDT poisoning. This is not easy to prove one way or the other. It is wellknown that infectious hepatitis has been a scourge of armies prior to the use of chlorinated hydrocarbon insecticides. The diagnostic criteria for infectious hepatitis rests on more solid ground than those for chronic DDT poisoning.

Workers engaged in manufacturing DDT have been studied (21) (192). No case of chronic DDT poisoning has been uncovered (192). Teams of people engaged in applying DDT for months to years have been studied in various parts of the world (43) (210). No case of proved chronic DDT poisoning has been reported. Large groups of people have been examined in the State of Washington, particularly in the Wenatchee district, where millions of pounds of DDT have been used over the past 8 years. Despite the fact that DDT intoxication has been looked for specifically, no case has been found. It is true that fat biopsies taken at elective surgery show DDT in the fat in many instances (192).

Deichmann, Witherup, and Kitzmiller (21) reported that two groups of workmen engaged in manufacturing DDT and exposed for periods up to 13 months showed no signs nor developed symptoms referrable to DDT absorption. The concentration of DDT in the general atmosphere breathed by the workmen ranged from 6 to 10 mgm of DDT per cu. meter of air, and in specific locations ran up to 26.6 mgm of DDT per cu. meter of air.

The only study dealing with workmen exposed to chlordan, aldrin, and dieldrin was done by Princi and Spurbeck (179). They examined 34 individuals carefully and repeatedly, 22 of whom had been exposed for periods ranging from 1 to 3 years in a manufacturing and formulation plant. The approximate concentration of these substances in the air of the working environment was determined at more than 5 mgm per cu. meter of air. These authors could find no evidence of any deleterious effects on the central nervous system, the liver, the kidneys, or the hematopoietic system.

There are other experiments involving human subjects which have been reported by competent observers that have not been mentioned. Neal and his group at the National Institutes of Health (1) and Velbinger and his co-workers (41) in Germany have contributed many observations regarding the effects of moderate amounts of DDT by ingestion, inhalation, and skin contact on man. They were not able to demonstrate any harmful effects on themselves or on other subjects in the experimental groups.

The point is worth emphasizing that there are no proved cases of chronic chlorinated hydrocarbon insecticide poisoning in the literature (18). Most of the reports which deal with chronic toxicity in man fall back on Biskind's publications or on animal experiments for substantiating evidence.

In conclusion, it can be stated that the clinical picture of chronic chlorinated hydrocarbon insecticide poisoning has not been fully elaborated in man. Work has been started on autopsy material in an attempt to find correlations between the chlorinated hydrocarbon insecticide concentration

in the body fat and the presence of characteristic liver lesions of whatever nature. In certain localities fat biopsies are being taken routinely at surgery in an attempt to correlate specific symptoms or syndromes with the presence of these insecticides in the body fat.

At present, a diagnosis of chronic chlorinated hydrocarbon insecticide intoxication can be attempted only if one has a definite history of continued exposure to these compounds. It is reasonable to assume that there will be a history of anorexia and weight loss. Headache and nervousness may be present. It has been emphasized by Princi (179) that these symptoms are not specific for chronic intoxication and that they may have other etiologies. Furthermore, there are no laboratory tests which are indicative of increased absorption or of true intoxication (18).

Further discussion of diagnosis and treatment will be undertaken in the discussion of cases of aldrin and dieldrin intoxication which have not been reported in the literature.

CHAPTER VII

THE TOXICITY AND HAZARDS OF ALDRIN AND DIELDRIN TO MAN

There is no disagreement among toxicologists that aldrin and dieldrin are toxic materials capable of producing symptoms of poisoning, tissue damage, and even death in human beings if the exposure to these insecticides is sufficiently great in terms of quantity and duration of exposure.

What is most interesting is the fact that after five years of use-experience involving millions of pounds of these insecticides, published reports of intoxication in human beings are limited to one case (197), an intentional ingestion of aldrin which was not fatal and in which recovery was apparently complete.

In the period from 1950 to 1952, eighty-seven cases of alleged intoxication due to aldrin, dieldrin and endrin have been collected and are presented for the first time in this paper. Although even cursory examination of some of these reported instances of intoxication indicate that they are something else, they will be considered because someone had suggested that a diagnosis of insecticide intoxication was in order. Spiotta's single case of aldrin intoxication appears in this tabulation.

For the purpose of comparison, and in the interest of simplicity, a table has been constructed listing each reported instance of alleged intoxication and an abstract of all available information pertaining to the

circumstances of exposure, symptoms, physical findings, laboratory findings, treatment given, and the duration of illness in each instance where that is known.

In many cases, it was not possible to obtain all of the information one might like to have for purposes of differential diagnosis. As a matter of fact, in most cases one can only hazard a guess as to the real nature of the illness. From the information at hand, it could be concluded that diagnoses were arrived at on the basis of vague suspicion. On other occasions, the patient made his own diagnosis on the basis of rumor. Sometimes groups of people were involved in reporting that they had been poisoned by either aldrin or dieldrin. Whatever the source of the reported cases of insecticide intoxication, investigations were undertaken by industrial hygienists, chemists and physicians by visiting the people involved, obtaining information from the attending physicians, or by consulting with the health officers in the region involved with the view of obtaining all the known facts. Sometimes these investigations were not very fruitful. Other times definite information was obtained which would help materially in evaluating the case.

It should be mentioned that Dr. Frank Princi, Associate Professor of Industrial Medicine, Kettering Laboratory, College of Medicine, University of Cincinnati, has given his files to the writer from which the table on the succeeding pages was derived.

ALLEGED CASES OF EXPOSURE TO ALDRIN, DIELDRIN, ENDRIN

	Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
1.	30	Preparing grasshopper bait with Aldrin.	Headache, "upset stomache", weakness, anorexia.	Tremor- no convulsions; Neurologic negative	Bed rest, laxatives Phenobarb.	Hospitalized 5 days - discharged recovered.	
2.	60	Mixing Aldrin with flour without protection or ventilation for 29 days.	Headache, dizziness, epigastric pain and discomfort, weakness, general malaise.	No tremors or convulsions; Neurologic negative.	Codeine gr ss tid.	Uneventful recovery in 24 hours.	
3.	40	Placed large strainer from Aldrin spray container in his mouth and blew it out. Also blew out 6 nozzles in same way. Also had Aldrin on his hands for 1 1/2 hrs. while he worked with the machine.	Violent nausea 1 1/2 hrs. after beginning exposure. Weakness for 24 hours. No other symptoms.	Not attended by a physician.	None	Completely recovered one week after original exposure.	
4.	49	Spread mixture of Aldrin and Chlordan on farm over unknown period of time.	Headache, vertigo, nausea, general malaise.	No tremors or convulsions; Neurologic negative.	Elix. Phenobarb. 2 oz. tid p.c. Bed rest	Returned to work in 3 days.	
5.	3 1/2	Sucked Aldrin emulsion from garden hose.	Nausea and weakness for 24 hours.	Not attended by physician.	None	Completely recovered in 3 days.	
6.	23	Swallowed six ounces of Aldrin in a suicide attempt.	Tonic and clonic convulsions in 20 min. Comatose.	N.P.N.-81. Icterus index - 15. Urine 3+ albumin with gross rbc. B.P. 170/88. No motor irritability. No pulmonary abnormalities. Abnormal EEG.	Emetic. Morphine Sulfate Barbiturates. Restraints.	No evidence of residual damage after discharge one month after admission.	
7.	?	Following airplane crash, contents of insecticide hopper (containing 2.5% Aldrin and 5% DDT) were dumped onto the pilot. He got an undeterminate amt. on his eyes, nose & mouth.	None at any time.	Hyperemia of conjunctivae.	None	No evidence of systemic intoxication.	

	Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
8.	30	Pilot, Adrin spray was sucked into the cockpit continuously while plane was in air. Boots, shoes and feet were soaked with solution.	Nausea and headache. Pinpoint pupils (?)	Pinpoint pupils or other clinical signs not observed by physician, L.M.D. considered symptoms to be functional.	None	Complete recovery in 1 week.	
9.	36	Pt. was opening 4 lb. bags of DDT and emptying them into large containers. Aldrin was being used in another part of building.	Delirium	Convulsions. Coma. Cheyne - stokes respiration.	Oxygen, i.v. glucose, barbiturates.	Complete recovery in 3 days.	Although reported as case of Aldrin poisoning, the person was admittedly never in contact with the material.
10.	25	Mixed Aldrin "dust" with hands in formulating plant for about 6 weeks. Conditions of exposure excessive.	Vomiting - no nausea. Headache. Dizziness.	Clonic and tonic convulsions. Pulse 118, resp. 24, B.P. 102/70. Temp. 98. Loss of weight.	Coramine, calscorbate, thiamine, activated charcoal, castor oil.	Complete recovery in 1 week.	
11.	19	Packaging Aldrin in formulating plant. Length of time unknown.	Nausea. General malaise.	Irritability & convulsions. Bronchitis, Neurologic negative.	Sedative, vitamines. Cough syrup.	Complete recovery in 3 weeks.	
12.	28	Mixing Aldrin by hand in formulating plant. Length of exposure unknown.	Headache, vertigo, vomiting.	Convulsions, Dyspnea, "slight anemia".	Oxygen, sedatives, bed rest.	Recovery in 1 week.	
13.	40	Mixing Aldrin by hand in formulating plant. Length of exposure unknown.	Headache, vertigo.	Lacerated wound of scalp. Neurologic negative.	Suture of wound.	No sequelae.	
14.	21	Mixing Aldrin by hand in formulating plant. Ingested unknown quantity of material.	Headache, vomiting, nausea.	Convulsions. Question of epileptiform convulsions. EEG showed cortical disturbance.	Supportives.	Diagnosis still undetermined. No sequelae to date.	
15.	18	Mixing Aldrin by hand in formulating plant. Length of exposure unknown.	Headache, nausea, general malaise.	Convulsions. Neurologic examination negative.	Sedatives and vitamines.	Complete recovery in 1 week.	

Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
16-24 (9 cases of alleged Aldrin poisoning)	Farmers and helpers using 25% emulsifiable oil diluted in 60-80 volumes of water. Also exposure to 2 1/2% Aldrin dust. Exposure from few hours to one month.	Headache, nausea, dizziness, epigastric pain, dryness in throat, pains in joints, tremors. One person complained only of "terrible dreams".	No laboratory findings, very few tests made.	?	All recovered from a few hrs. to some days. Most changed employment.	Information from Dept. Natural Health & Welfare. D.L. Henderson. M.D. Admittedly cases not studied very well if at all.
25. ?	Pilot. Allegedly crop dusting with Aldrin. Exposure unknown.	Vomiting, headache, numbness in rt. arm and leg. Loss of equilibrium, slurred speech. Difficulty in staying awake. Vision diminished. Double and triple vision. Nystagmus. Depression and irritability.	Incomplete information. Wife stated that spinal tap and x-ray were done at hospital. Diagnosis of brain cell atrophy made. This opinion disputed by another physician, who thought this was an acute toxicity.	?	Patient improving and back to work. Acute twitching has subsided.	Unable to evaluate from information at hand. Probably not Aldrin intoxication.
26. 33	Aldrin operator at manufacturing plant. Duration of exposure unknown.	Several grand mal type seizures.	EEG-mild showing of cerebral rhythm. Repeat EEG several weeks later showed improvement over above record and interpreted to be within normal limits. Physical exam., neurologic and all lab work negative except for slight elevation of urea nitrogen of 16.8 mgm%.	?	Improvement. No further seizures.	Question of epilepsy in family.
27. 60	Plantation worker. Spilled Aldrin 2 1/2% and DDT - 5% on right shoe.	Blister on dorsum of right foot appeared day following exposure.	No systemic complaints recorded.	Home treatment. L.M.D. gave treatment. X-ray.	Subsided. Told not to come in contact with insecticides, fertilizers, dusts, etc.	Pt. states his skin is very sensitive. He gets the same type of dermatitis from fuel oil and kerosene. farm

	Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
28.	42	Pesticide sprayer since 1947. Used 700-1000 gals. a day. Used DDT-50% wettable powder. Chlordan-50% wettable powder. Toxaphene-1 pt. to 100 gals. water. Lindane. Lead arsenate. Had used nicatine and paratarion in previous years.	Loss of energy; easy fatigability. Severe neck and head pains, marked night sweats. No complaints re g.i. or respiratory tracts.	Physical findings negative. Lab. findings negative. Chronic prostatitis.	?	No marked improvement	This is a diagnosis by exclusions because no findings were present to explain symptoms. A diagnosis made of chlorinated hydrocarbon insecticide poisoning.
29.	?	Pilot. Flew 3726 gals. 465.75 lbs of oil solution containing 3.3 oz. of 60% AES per gal. fuel oil. Flying hrs. 9 hrs. 25 min. 34 sec. Never wore respirator. Used also 2 oz of actual Aldrin per gal of oil solution. Got Aldrin on clothes. Never cleaned up and was negligent in his habits regarding "spills".	No symptoms. Died in fatal plane crash. No explanation of why or what caused accident.	Bioassay for Aldrin in tissues revealed nothing except apparent 0.7 ppm. of Aldrin found in fat.	None		Very difficult to ascribe accident to Aldrin intoxication.
30.	?	Pilot. Sprayed Dieldrin for 20 days. Exposure unknown.	Single fainting spell after spraying 1 week. After 20 days spraying developed asthesias of both lower limbs and back pain. Symptoms lasted about 2 hours.	?	?		
31.	?	Pilot. Supposed to have sprayed Dieldrin.	?	?	?		
32.	?	Spray pilot.	Became ill one day and fainted.	?	?		

	Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
33.	?	Pilot. Sprayed 2000 acres. Flew Dieldrin 17 days before wrecking plane. Used 2 gal. diesel oil and 1/6 gal. of the 1-1/2 lb. emulsifiable concentrate Dieldrin to acre. Used bare hands to transfer booms and nozzles to another plane.	Vesicles on lips.	Supposed to have increased WBC - 12,800.	?	?	Refused to fly Dieldrin after elevated WBC
34.	?	Ground worker for above. Helped to transfer booms and nozzles to another plane.	?	Supposed to have increased WBC - 16,800.			
35.	?	Pilot flying Dieldrin; exposure unknown.	Acute toxic state, severe headache, muscle aches and pains in back of legs.	Temp. 103°F. Liver function test (Hanger - +3). WBC 20,000 2nd day - 8,000 following day.	None	Rapid recovery.	
36.	43	Foreman of C. Chem. Co. Exposed to clouds of vapor when flake Dieldrin was sprayed.	Respiratory distress. g.i. upset.	?	Hospitalized 2 days; given oxygen therapy.	Recovered.	
37.	5	Child drank kerosene from can in which Dieldrin spray nozzle had been washed.	?	?	?	Recovered	Bioassay of material gave an approximate 0.05% Dieldrin.
38.	?	Dieldrin operator.	Few slight fainting spells. Occasional muscular twitches of neck. General feeling of mental unrest.	Abnormal EEG. Gradual but consistent increase in RBC in 3 counts - 4th count showed decrease.		Recovered	Question of epilepsy.
39-46 (8 stevedores refused to load a cargo of 2-1/2 Aldrin dust)		Apparently were working in hold when a bag broke.	Complained of burning eyes, itchy skin, nausea.	None	None	Returned to work.	Apparently this was an attempt to get more money for doing "dangerous work."

	Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
47.	?	Chemist and plant manager exposed to Dieldrin through skin of hands and respiratory tract.	Extreme tiredness and irritability - mild intoxication - extreme chills - sudden sweating and feverish feeling. Normal appetite.	None - not attended by a physician.	None	All symptoms gone 9 hrs. after initial exposure.	
48.	34	Operator of tractor spray rig 10 hrs. day for 14 days. No precautions - didn't wash arms or hands.	Nausea, abdominal pain, profuse watery diarrhea. Headache, irritability, vertigo.	Negative.	?	Progressive weakness and shortness of breath. Loss of appetite and progressive weight loss. Low grade generalized abdominal pain, occasional nausea and mild diarrhea.	
49.	?	Nozzle stopped up while spraying cotton. Nozzle came off and patient was sprayed with Aldrin. Didn't wash or change clothes.	Severe pain in back between shoulders. Nausea, vomiting, and "pretty well delirious". Smell of Aldrin makes him violently ill again.		?	Still "weak".	
50.	?	Exposure unknown except that he was spraying cotton.	Severe nose bleeds and "kind of ulcer in nose". Severe headache everytime he smells Aldrin.	?	?	Weak - unable to do hard work.	

Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
51-76 (26 men involved)	Employees of a formulating plant H-S Co.	"Contact" dermatitis. No other symptoms.	Described as "much like poison ivy". Onset at flexure crease at elbows, spreading over lower arms. Then neck is involved, with spread to chest and abdomen.	Cortisone (?) Antihistaminics (?)	Recovery	Materials handled are BHC, Toxaphene, Parathion, Calcium Arsenate, tetroethyl pyrophosphate, DDT, pentachlorophenol, ammonium sulfate and following Dieldrin formulations: Dieldrin 2.5% " " " 5% DDT - 40% S " " " 40% S " " " 10% S " " " 5% S Dieldrin 15% Concentrate Dieldrin 25% Concentrate Dieldrin 1-1/2% "emulsifiable"
77-78	"	"	"			
79-82	Employees of H-P dusting service. Using 2-1/2% Dieldrin 40% S comps. Some calcium arsenate dusting.	Dermatitis				
83. ?	Spread 100,000 lbs of dust. Dieldrin	Dermatitis. Pain in neck and shoulders - headache.	Dermatitis started at flexure crease at antecubital space, then under wrist watch strap, with spread to chest, back, arms, face and legs.	Cortisone	Recovery	Has been using insecticides since 1945.

	Age	Exposure	Symptoms	Physical Findings	Treatment	Outcome	Comment
84.	46	Pilot. Flew daily 2-3 weeks prior to onset. Mixed tanks and did spraying. Thought to have fumes and vapor. Skin absorption hands and face. 30 minute contact, removed from skin with soap and water. Dieldrin was used.	Headache, vertigo, nausea, general malaise, irritability. Feeling of heaviness of legs. No dermatitis.	Negative.	None	Apparently no progress.	Pt. now thinks his trouble is not due to Dieldrin.
85.	?	Pilot. Crop duster using Dieldrin.	Sometimes feels dizzy after dusting with Dieldrin. No dermatitis.	Negative.	None	?	?
86.	?	Spray operator driving rig downhill when sudden stop caused water solution of Dieldrin to drench operator. Did not wash.	Developed a dermatitis.	Dermatitis appeared 48 hrs. after exposure.	Cortisone.	Recovered.	
87.	?	1% Endrin (comp 269) applied to 4.1 acres with 2 row hand duster without special precautions.	Vomited in 1 hr. after work over. 8 hrs. later violent attack of dysentery. Rash resembling heat rash persisting 9 days after dust was used.	?	?	?	This man used "other materials" also was "doing hard work in extremely hot weather".

Nothing would be gained by attempting to discuss each entry in the preceding table. It seems simplest to consider the patients by groups. The following table summarizes the reported cases according to the circumstances of the alleged exposure to aldrin, dieldrin or endrin:

Number of Persons Allegedly Exposed to Aldrin, Dieldrin, Endrin

Type of Activity	Number of Persons Exposed to		
	Aldrin	Dieldrin	Endrin
Agriculture (total cases: 37)			
Farming	17	0	1
Professional Crop Dusting	4	13	-
Exterminators (spray operators)	1	1	-
Manufacture (total cases: 39)			
Manufacturing	1	1	
Laboratory	0	1	
Formulating	7	29	
Miscellaneous (total cases: 12)			
Accidental exposure	1	1	
Intentional ingestion	1	1	
Handling in transit	8	0	

In some of the instances included in the above table, it is difficult to determine the source as well as the degree of exposure to these insecticides.

In the agricultural group it is noted that 37 cases had been reported of which 22 were allegedly due to aldrin, 14 to dieldrin, and 1 to endrin.

Under the sub-group of farming, 17 cases of aldrin intoxication are noted, and 1 case of endrin poisoning. Of the 17 cases of exposure to aldrin, 9 cases are said to have occurred among farm laborers and helpers using aldrin in the form of 25 per cent emulsifiable oil diluted in 60 to 80 volumes of water. Aldrin dust (2.5%) was also used. The duration of exposure varied

from a few hours to one month. Headache, nausea, dizziness, epigastric pain, dryness of the throat, pains in the joints and tremors were the symptoms noted by this group. Not all of the listed symptoms appeared in all patients. One person complained only of "terrible dreams". The physical findings revealed little. No laboratory findings were reported. It should be noted that all patients recovered after periods ranging from a few hours to some days. Most of these men changed their employment. It seems that 8 of these cases are sufficiently typical to be called chlorinated hydrocarbon insecticide poisoning. The single patient who complained only of "terror dreams" cannot be considered as suffering from insecticide intoxication.

Another patient was engaged in preparing grasshopper bait with aldrin. His complaints included headache, anorexia, nausea and weakness. He had a tremor but convulsions did not develop. The physical findings were otherwise within normal limits. The patient was hospitalized for 5 days and was given symptomatic treatment consisting of bed rest, laxatives and phenobarbital. He made a complete recovery. This is considered to be a case of mild intoxication due to aldrin.

The 49 year old farmer who had been engaged in spreading a mixture of aldrin and chlordan complained of headache, vertigo, nausea and malaise. He did not develop tremors. Phenobarbital and bed rest were prescribed for 3 days after which time the patient returned to work.

Another farmer placed a large strainer from an aldrin spray container between his lips and blew it out. He also blew out 6 nozzles in the same

way. In addition, his hands had been kept contaminated with aldrin for approximately 1½ hours while he adjusted the machine. He developed a violent nausea about 2 hours after this exposure. Other than a feeling of weakness which lasted for 24 hours, no other symptoms developed. He did not seek medical help. Recovery was complete within one week after exposure. This, too, must be considered as a case of aldrin intoxication.

A 60 year old man was engaged in mixing aldrin with flour without protection or ventilation for 29 days at a plantation. He developed headache, dizziness, epigastric pain and discomfort, weakness and malaise. No neurologic abnormalities were noted. The patient was treated with codeine and he made an uneventful recovery within 24 hours. The exposure and the symptoms are sufficient to accept this as a case of insecticide poisoning.

A farm laborer applied a 1 per cent formulation of endrin to approximately 4 acres with a 2-row hand duster. About an hour after he completed his 8-hour's work, he vomited following which he developed a violent diarrhea. A skin rash appeared, which resembled a heat rash and persisted for about 9 days. Upon inquiry, it was learned this man had been using "other materials" and had been doing hard physical work in extremely hot weather. This particular situation is not considered to be typical of insecticide intoxication.

Another plantation employee was spraying cotton with aldrin when the nozzle became clogged and blew off. The man was sprayed with aldrin but he did not wash or change clothing. He complained of severe pain in the back between the scapulae. He became nauseated and vomited and was "pretty well

delirious". It was stated that the odor of aldrin made him violently ill. He complained of feeling weak some weeks after this episode.

His brother stated that he, too, becomes nauseated and develops a severe headache every time he smells aldrin. He stated that he has severe nose bleeds and a "kind of ulcer" in the nose. He thinks he is weak and unable to do hard work. This man had been spraying cotton also.

It appears that the former case is probably an intoxication due to aldrin with some exaggeration of symptoms during convalescence. The latter case does not fit into a diagnosis of insecticide poisoning.

One plantation worker spilled a quantity of insecticide containing 2.5 per cent of aldrin and 5 per cent of DDT on his shoe. A blister appeared on the dorsum of the right foot the day following this incident. No systemic complaints were reported. The patient treated himself and then received some X-ray therapy which produced complications, so that he sought treatment elsewhere. The patient has said that his skin is very sensitive and that he gets the same type of dermatitis from fuel oil and kerosene. This does not appear to be a lesion produced by aldrin or DDT. Over-treatment very likely contributed to the dermatologic disorder.

An operator of a tractor spray rig was exposed for 10 hours per day over a period of 14 days. No precautions against aldrin were taken. At the end of this time, he complained of nausea, abdominal pain, profuse watery diarrhea, headache, irritability and vertigo. No neurologic abnormalities were noted. The patient did not make a rapid recovery. This patient also complained of progressive weakness, anorexia, loss of weight

and shortness of breath. This must be considered a case of insecticide intoxication. However, some other disorder may explain some of the findings.

Seventeen cases of reported intoxication occurred among professional crop dusters. Exposure to dieldrin accounted for 13 cases; the remaining 4 persons had been applying aldrin. Of this sub-group 5 pilots working with dieldrin developed dermatitis but no systemic complaints. In one instance, cortisone was used therapeutically with success. In 4 of the cases there was exposure to sulfur and calcium arsenate as well as to dieldrin. It is not clear that these are at all typical of the effects of dieldrin on the skin.

Two pilots dispersing dieldrin had requested their physician to notify them if their white blood cell counts should ever increase to abnormal values. Because the white cell counts were reported, in one instance, to be 12,800, and in the other 16,800, these men refused to handle dieldrin. The rationale of basing an estimate of the degrees of exposure to chlorinated hydrocarbon insecticides upon an increasing white blood cell count has not been established.

Still another pilot, who had been exposed to dieldrin, developed a severe headache, muscle aches and pain in the legs, and a fever of 103 degrees F. The white blood cell count was 20,000 on the second day of the illness but dropped to 8,000 the following day. The patient made a rapid recovery without treatment. This is not a typical picture of intoxication due to insecticide.

Another pilot was rumored to have complained of illness, but when he was questioned he had no complaints.

Two spray pilots were said to have become ill and fainted. Further examination revealed nothing indicative of chemical intoxication. This could have been a fatigue phenomenon in that these men work hard during the crop dusting and spraying season. The demand for such service is great.

One pilot had no complaint other than dizziness.

The last pilot of the aldrin group complained of headache, vertigo, nausea, malaise and irritability. The physical findings were not abnormal. However, he made no progress towards recovery. He did not ascribe his complaints to excessive exposure to dieldrin or to aldrin.

Of the four pilots in this series that dispersed aldrin, one died in a plane crash. It was known that the pilot was negligent in his habits with regards to "spills" of insecticide on his clothes. Examination of the fat showed a concentration of aldrin approximating 0.7 ppm. This cannot be considered to be evidence of insecticide intoxication. The cause of the accident could not be determined. In view of the fact that nothing is known about any subtle effect that the chlorinated hydrocarbon insecticides might have upon flying skill, one cannot attribute the accident to the effect of the insecticide upon the pilot.

Another pilot engaged in distributing aldrin developed symptoms including headache, nausea, visual disturbances, disturbed equilibrium, numbness of the right arm and leg, depression and irritability. There is some question

about the diagnosis in this case since two widely varying opinions have been offered.

The other two pilots in the aldrin group had definite exposure to aldrin. One had no systemic complaints. The other pilot complained of nausea and headache and was reported to have had pin-point pupils. His physician was of the opinion that these complaints were functional in origin and not due to insecticide intoxication.

Of the entire sub-group of persons engaged in professional crop dusting, no case which is typical of chlorinated hydrocarbon insecticide poisoning is recognized. One might say that the 5 cases of dermatitis might be due to exposure to chlorinated hydrocarbon insecticide. The remaining cases, excluding the pilot that died in the airplane crash, are not typical of insecticide intoxication.

Under the sub-grouping of exterminators there is noted one case each of aldrin and dieldrin exposure. In one instance, dermatitis was the leading complaint, which responded well to cortisone therapy. The other patient had vague complaints. Because of his occupational history as an exterminator and in the absence of any other explanation for his symptoms, it appears that the tag of insecticide poisoning is attached to this patient.

Manufacture accounts for 39 alleged cases of intoxication from exposure to aldrin and dieldrin. One of the two cases under the sub-group of manufacture has been diagnosed as epilepsy, and the other as a brain tumor.

The single case reported in the laboratory sub-group, however definite

the exposure, does not appear to be typical of chlorinated hydrocarbon insecticide intoxication.

Among the formulating sub-group, six cases of alleged intoxication are typical of chlorinated hydrocarbon insecticide poisoning. Convulsions developed in these patients following excessive exposure to the insecticide. The last case had no contact with aldrin, and this cannot be attributed to either aldrin or dieldrin.

The 28 cases of dermatitis seen at a dieldrin formulating plant could be attributed to any of the compounds handled in that plant. The last patient was exposed to clouds of vapor when flaked dieldrin was sprayed. He developed a respiratory distress requiring oxygen inhalation but he recovered within two days.

In the miscellaneous group, four cases are unquestionably the result of exposure to either aldrin or dieldrin. One case of dieldrin intoxication is reported in another section of this paper.

The 8 cases reported under the sub-group handling in transit are not medical problems. Further investigation seemed to show that these men were looking for more money for doing "dangerous work".

This section can be summarized very briefly. Of the total cases reported, only 29 cases are considered to be actual cases of either aldrin or dieldrin intoxication. Thirty-three cases were called doubtful but have not been excluded. Twenty-seven cases were not considered to be insecticide intoxication.

It is apparent that the limited number of suspected cases seems to fall

in line with the estimate of the hazards of the various circumstances of exposure. The formulating plants seem to have the greatest hazards. The least hazardous place seems to be in the manufacturing of the active ingredients of a formulation. The crop-dusters and the professional exterminators have hazards but they seem fairly well controlled. The farmer has the dubious honor of leading in the number of cases considered to be true cases of poisoning. While realizing that it would be dangerous to draw sweeping conclusions from such limited data, which are unsatisfactory and incomplete in many ways, it is interesting that the trend is what one might predict from the very nature of the operations under each group.

This thought will be developed further in the chapter on the environmental hazards of the chlorinated hydrocarbon insecticides.

CHAPTER VIII

ENVIRONMENTAL HAZARDS OF CHLORINATED HYDROCARBON INSECTICIDES

There is no question about the fact that the chlorinated hydrocarbon insecticides are toxic materials which, if mishandled, are capable of producing injurious effects in human beings. It is worth reviewing the circumstances under which man may come in contact with these insecticides in sufficient quantity to cause illness.

Barnes ( ) discusses the possibilities under three headings: 1. Hazards during manufacture; 2. hazards during application; 3. and hazards to persons not directly concerned with the manufacture or application of the insecticides. This classification seems to be a logical one and will be followed in this paper.

The opportunity for exposure to toxic amounts of any of the chlorinated hydrocarbon insecticides would seem greatest during the development, pilot plant operation and large-scale production of the active ingredients of these materials. However, the well established chemical companies usually have some knowledge of the comparative toxicity of the material in question in relation to a known compound and plan the operation accordingly. These plans may have to provide for specially designed equipment or buildings in order to provide adequate safeguards to protect the employees from excessive exposure while handling the toxic materials.

In many cases, if the basic principles of good industrial hygiene

practice are followed and medical supervision of the employees established the degree of exposure to toxic materials may be controlled sufficiently to prevent any serious accident or ill-effects to the employees. At least, it has been the American experience that the hazard of excessive exposure to the chlorinated hydrocarbon insecticides during manufacture has been practically non-existent. (See papers by Princi and Spurbeck (179), Heyroth (17), and McGee et al (142).)

During manufacture, the greatest hazard of over-exposure to these insecticides occurs among the formulating plants. It is in these plants that the active ingredients are prepared in forms suitable for sale to the consumer.

It has been pointed out that this aspect of manufacture is highly competitive, the margin of profit comparatively small, and production seasonal. It is for these reasons that many of the formulating plant operators came to the conclusion that they cannot afford to buy special equipment for the purpose of protecting the health of employees, especially when the hazards seem not to be serious.

It should be mentioned that for the very reasons cited above, the operators of formulating plants should insist upon good plant housekeeping and high standards of personal hygiene from their employees. It must be through ignorance or indifference, that these inexpensive protective measures are often overlooked.

Despite the deficiencies in the protective health measures in some of the formulating plants, accidents of over-exposure and illness due to

chlorinated hydrocarbon insecticides have been relatively few. The literature on this subject is remarkably thin. One is surprised that so little has been written about workmen made ill by insecticides. Furthermore the proved instances of poisoning due to chlorinated hydrocarbon insecticides are so few that it might be suspected that the fears of those who anticipated many cases of chronic poisoning have not materialized.

The record is fairly good, and some acknowledgment should be made of the efforts of the manufacturers of the active ingredients to prevent excessive exposure to these materials among the employees of the formulating plants. Some of the chemical companies provide detailed information and exact specifications for the equipment to be used for producing specific formulations safely. Technical bulletins and manuals have been provided which describe the toxicity of the compounds and the possible health hazards involved in preparing the formulations. Some manufacturers have gone so far as to sell their products only to those formulators who are equipped to handle the ingredients in a safe manner and who provide adequate protective measures to prevent undue exposure of their employees to the materials.

It must be remembered that some formulators have very limited capital and cannot afford to install the special equipment which will provide even minimum protection to the employees. It is this kind of manufacturing operation that requires the most guidance.

It is not at all impossible to improve the controls over the health hazards in the production of the chlorinated hydrocarbon insecticides. State health departments who maintain occupational health divisions and

state labor departments charged with the responsibility of seeing to it that the laws regarding safe working environments are being complied with, may both contribute a great deal through inspections with the view of pointing out specific deficiencies and making specific recommendations for correction to plant operators who cannot afford their own industrial medical program or industrial hygienists.

Another "pressure" source can come from unions whose members are involved in the manufacture or formulation of insecticides. Frequently, a safe work environment is made a bargaining point in negotiating labor contracts.

The employee should participate in a preventive medical program by maintaining high standards of personal hygiene and using common sense in avoiding excessive exposure to the toxic materials.

The problem of insecticide poisoning in the group discussed above is not one of any great magnitude. The workmen are in an environment that can be controlled. Furthermore, the activities of the workers can be supervised and even restricted if that is indicated.

The above considerations may be summarized by stating that the hazards associated with the manufacture of the active ingredients of the chlorinated hydrocarbon insecticides are quite real. However, in actual practice, these hazards have been very well controlled. The hazards associated with the formulation of these materials are greater; but they have been less well controlled. The solution to these problems is not particularly difficult in as much as the principal control measure utilizes the fundamentals

of good industrial hygiene practice coupled with a medical control program. This kind of an approach will take care of the most urgent situations satisfactorily.

The greatest hazard of poisoning from the chlorinated hydrocarbon insecticides is found among the farmers and exterminators who must, in many instances, dilute the formulations to a suitable concentration for use on crops, livestock and buildings. The problem is one of the handling of concentrated materials in a careless and thoughtless manner. These people are less likely to have supervision, and very often, have no guidance except from their own understanding and appreciation of the need for careful handling of toxic materials. The incidence of insecticide intoxication may be reduced in this group through an educational program aimed at stressing the common sense approach of avoiding unnecessary exposure to these materials.

In this connection, the passage of the Federal Insecticide, Fungicide, and Rodenticide Act, on June 25, 1947, which is administered by the United States Department of Agriculture, provides for a system of labelling of insecticides. The manufacturer is required to tell the user what he must do to avoid trouble. In this way, the user would gain some knowledge regarding the safe handling of these materials. However, this device presupposes that the user is literate and is capable of understanding what he is to do.

Any person who plans to use an insecticide should inform himself of its characteristics and the conditions of safe use. Information is available from the Department of Agriculture, County agricultural agents, State

agricultural colleges, Agricultural experimental stations, Extension services and from the manufacturers.

Ignorance accounts for some accidents of over-exposure to the insecticides but the real danger lies in the person who exercises great care when he first uses the product, but who, experiencing no ill effects, becomes more and more careless until intoxication occurs.

Other schemes have considered the size and type of containers for packaging insecticides. It was thought that if the concentrated materials were used up in preparing a single batch of finished material, the weighing out and measuring operations would be minimized and the storage of half-used packages and drums of toxic materials would be made unnecessary. Some states, notably California, have used this approach with some success.

A generalization can be made that all of the materials under consideration may be in the form of dusts, liquids or pastes. Each of these present their own peculiar hazards in handling. There is agreement that the dusts are the most difficult to handle. On the other hand, the liquids have the disadvantage of having to be poured from drums which presents the possibility of splashing if handled carelessly. If the drums are large, they become clumsy to handle and give rise to the possibility of accidental dropping. The pastes have not had extensive use.

The hazards of applying insecticides in an area depends, in some measure, upon the methods of dispersal used. The simplest way of dispersing an insecticide is to apply it by hand or by manually operated equipment. This can be an untidy operation in which the operators become saturated or covered

with the material being applied. Despite the widespread use and the crudeness of the method, very few accidents resulting in insecticide poisoning have been reported in the medical literature.

The application of spray materials with hand sprayers is a common practice among orchardists. In order to cover an entire tree, the spray must be applied from all sides and it is almost impossible for the men to avoid direct contact with the insecticide. The contamination of the skin and the inhalation of dusts and sprays being used are the chief sources of exposure. Under these circumstances, it might be expected that serious illness would result. Again, the reports dealing with this subject are very few.

There are hazards involved in the use of mechanically operated ground equipment. More insecticide can be applied over a greater area in a given time than by the use of hand sprayers. However, the risks of using mechanically operated ground equipment are not very significant. If the pattern of application of the insecticide is planned, even with equipment which does not have gas tight cabins which are artificially ventilated, the hazard of excessive exposure can be fairly well controlled.

Aerial application of the chlorinated hydrocarbon insecticides has gained in popularity and is now used fairly generally in treating fields of all sizes. The people who load the insecticide hoppers of the aircraft are subjected to a greater hazard of exposure to insecticides than the pilots. This is not to say that the pilots are not subjected to drifts of clouds of insecticide in poorly planned operations. Pilots, too, must be protected from excessive exposure, just as those who are not engaged in

crop dusting. It has been noted that the United States Civil Aeronautics Administration reported 1,383 agricultural flying accidents in the period from 1948 to 1951. Only in eleven instances could the cause of the accidents be ascribed definitely to the effect of a chemical or other noxious agents. It would be difficult to demonstrate possible subtle effects of the chlorinated hydrocarbon insecticides upon pilots which could lead to some physiologic alteration affecting their flying skill. In order to protect the pilots engaged in crop dusting it has been suggested that particular attention be given to designing aircraft most suitable for this type of operation.

One cannot overlook the mechanics and maintenance men who must repair and service the mechanical equipment used in the application of the chlorinated hydrocarbon insecticides. These workers have had not reason to become familiar with the problems of handling poisonous materials. It may well be that most mechanics may service this type of equipment only infrequently and not be aware of the hazards involved. It would seem most practical to have people who know how to protect themselves from these materials clean up the equipment before it is sent in for service.

There is a rather large group of professional exterminators who work by contract with farmers. The employees of these firms of contract sprayers usually have more knowledge of these products regarding toxicity, the hazards involved, and how to protect themselves than the individual farmer who only takes care of his own requirements. Nevertheless, there are special risks attached to the job of professional exterminator. These men are

exposed continuously to a variety of insecticides depending upon the special need for a given insecticide. Despite the usual precautions, accidents have happened among this group. Even so, there is a decided lack of published information regarding the incidence of insecticide poisoning among this group. One comes to the conclusion that the potential hazards are realized only infrequently. Perhaps the answer lies in the fact that these men are better informed, and the opportunities are better for observing and protecting these men. Even in situations where rotation of the employees from a greater to lesser exposure is not possible, there does not seem to have been any epidemic of insecticide poisoning.

In the evaluation of the hazards of chlorinated hydrocarbon insecticides to individuals who are not directly concerned with the handling of insecticides, a variety of opinions have been expressed ranging from statements calculated to create panic to almost complete indifference. No proved cases of poisoning have occurred among residents of areas subjected to aerial application of the chlorinated insecticides. Furthermore, at present, the evidence indicates that the residents in these areas have no more DDT in their fat than those living in cities not subjected to aerial treatment with insecticides.

There is no special risk to those workers who handle harvested crops treated with chlorinated hydrocarbon insecticides, except that they may possibly absorb small quantities in the same manner as does the consumer.

The most dramatic evaluation of the hazards of the chlorinated hydrocarbon insecticides concerns the general public who may consume food contain-

ing traces of insecticide. The assessment of the hazards to the general population is difficult. The evidence, at the present time, is that the ingestion of trace amounts of insecticides in common use has not produced symptoms or clinical syndromes of poisoning in man. This has been discussed in a previous chapter of this report. The data reporting the presence of DDT in the fat of human beings and in animals have been given. The significance of these findings has not been assessed on the basis of lifetime studies in the human being. It might seem reasonable to some authorities to think that because the chlorinated hydrocarbon insecticides are stored rather readily in the fat, they are more likely to produce chronic effects. There is no evidence to substantiate this belief.

From the facts gathered to date, it appears that the hypothetical risks to the general population from exposure to trace amounts of chlorinated hydrocarbon insecticides are small.

This is not to say that the residue problem should be ignored. It would be desirable not to have to deal with such a problem at all but it exists.

Several approaches have been taken to the problem of insecticide residues in food. It has been found that in many instances if the material is applied correctly, in the proper amount and at the proper time in relation to the harvest date, the chlorinated hydrocarbon insecticide residue problem does not exist. In those instances where trace amounts of insecticide were present, efforts have been made to find ways of removing the residue. Where it has been impractical for one reason or another to ensure that

the food is free from residue, the "pure-food" approach has been taken to control the amount of residual insecticide which is allowed. In some cases, tentative tolerance limits have been set. This kind of control requires that there be available reliable, specific, chemical methods of sufficient sensitivity for the quantitative determination of chlorinated hydrocarbon insecticides. Such methods are not available for all compounds. Where the methods do not exist, it is useless to try to set legal or even tentative limits for levels of insecticides for foods.

Barnes (192) has suggested that more use be made of the nonspecific organic-chloride method. One would have to agree that this method is loaded on the side of safety. He reasons that if no organic compound containing chlorine is present, then there cannot be any of the original insecticide left. However, if some organic compound containing chlorine is found, it may or may not be the original compound, but for the purpose of computing the residue it will be taken as being the original material.

If the organic-chloride method can be adapted for use with all of the existing chlorinated hydrocarbon insecticides, it is Barnes' recommendation that it be adopted by all but the largest and best equipped laboratories.

The final group for consideration is concerned with the non-occupational poisonings which have followed the accidental ingestion, purposeful ingestion, or purposeful poisonings of other individuals. These circumstances present no special considerations with respect to the chlorinated hydrocarbon insecticides than those involving any other poisonous material found within the home or at drug stores. The hazards are obvious. How to cope with this

problem is not so obvious.

In summary, a few common sense rules regarding the safe use of the chlorinated hydrocarbon insecticides are offered:

1. Be informed of the characteristics of the insecticide used.
2. Follow the directions for safe use as stated on the label.
3. Avoid breathing dust or spray mists.
4. If there is danger of absorption through the skin, wear impervious gloves.
5. Change clothing after spraying or dusting and bathe.
6. Clothing that is contaminated with insecticide should be laundered before being re-used.
7. Plan the operation with due regard to wind direction.
8. Check the mechanical condition of the equipment before using.
9. Sprays are less likely to drift than dust.
10. Store the insecticide in a safe place so as to be out of reach of children and animals. Do not store near food.
11. Destroy empty containers immediately or bury them.
12. Keep yourself and your place of work clean.

Recommendations regarding the wearing of protective clothing are theoretically correct. However, it simply will not be followed where the requirement is greatest. Personal hygiene measures may be used.

CHAPTER IX

DIAGNOSIS AND TREATMENT OF CHLORINATED HYDROCARBON INSECTICIDE INTOXICATION

The diagnosis of intoxication due to excessive exposure to the chlorinated hydrocarbon insecticides should be based upon the history of unusual exposure to these materials. The symptoms of intoxication are those of stimulation of the central nervous system. One can expect to encounter hyper-irritability, tremors, convulsions and possibly coma. Nausea and vomiting may be present. However, if extremely large doses have been absorbed rapidly then these last two symptoms and signs will not occur. The reason for this is that vomiting usually stops with the onset of central nervous system symptoms. Convulsions usually occur if prompt treatment is not obtained. They may recur at intervals of half an hour more or less. It should be noted that, as yet, there are no laboratory tests or examinations which will offer any help in making the diagnosis.

The treatment is symptomatic. It is imperative to remove all ingested insecticide as soon as possible by stomach lavage. Saline cathartics may be given but the oily cathartics should be avoided since their presence in the alimentary tract promote absorption of these toxic materials.

Barbiturates in sufficient dosage to control convulsions are the

drugs of choice. The drug may be continued until hyper-irritability is no longer present. Supportive treatment may be necessary.

The symptoms of chronic intoxications following continued exposure to these insecticides may be anorexia, loss of body weight, headache and nervousness. The diagnosis is based upon a definite history of exposure to the chlorinated hydrocarbon insecticides. None of these symptoms is specific for insecticide intoxication. The diagnosis is not made without the history. Again, under these circumstances, there are no laboratory aids to guide the physician in making the diagnosis.

The treatment involves removing the patient from further exposure to those materials. Symptoms will frequently remit within a few days. It has been suggested that if the signs and symptoms persist for more than a week after exposure has stopped, then one can assume that the etiologic agent was not a chlorinated hydrocarbon insecticide.

In most cases the prognosis for complete recovery is good. So far, there is no good evidence to indicate that chronic illness will result.

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