Translocation of Some Chlorinated Hydrocarbon Insecticides into the Aerial Parts of Pea Plants

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The increased use of chlorinated hydrocarbon insecticides in soil has often raised the question of whether or not these insecticides might translocate into the aerial parts of plants from the soil in spite of their extremely low solubility in water. This was tested by resorting to very high concentrations of insecticide and using a soil of minimum sorptivity and complexity. Plants were also grown in insecticide-free sand within a glass container surrounded by lindane-treated sand. Lindane is translocated through the root system into the aerial parts of the plant. No evidence of absorption through the leaf cuticle of lindane vapors released from the soil surface was obtained. Plants grown in covered pots containing aldrin- or heptachlor-treated sand contained only the epoxides within the leaves and stems. Epoxidation of aldrin to dieldrin and of heptachlor to heptachlor epoxide apparently took place within the plant tissue, after aldrin or heptachlor had been absorbed through the root system. Plants grown in dieldrin- or heptachlor epoxide–treated sand contained those insecticides within their leaves and stems. No evidence of DDT translocation was obtained. Under field conditions, seeds from pea plants grown in a Carrington silt loam soil treated with 5 and 25 pounds per acre of aldrin or heptachlor contained no measurable insecticidal residues.

During recent years, increased attention has been given to the insecticide residue problem. Positive evidence of the translocation of benzene hexachloride from treated soils into the aerial portions of plants was reported by Starnes (*18*), who found that both aqueous suspensions and benzene extracts of potato foliage from plants grown in soil treated with 10 pounds of γ-benzene hexachloride per acre were highly toxic to *Aedes* mosquito larvae. Haines (*3*) reported "the presence of a toxic material in both guttated liquid and tissues of corn plants grown in lindane-treated nutrient solution." Paraoxon was found (*2*) in the fluid guttated from leaves of cabbage and wheat plants, grown in parathion-treated soil. Crops grown on lindane-, DDT-, and aldrin-treated soils contained from zero to various amounts of insecticides (*5*), depending on the insecticide, its concentration in the soil, the crop itself, and the soil type in which the crops were grown.

Mechanisms by which an insecticide applied to a soil might contaminate the aerial parts of plants grown in that soil are: rain or other physical factors resulting in the direct deposition of small amounts of treated soil on the foliage; volatilization of the insecticide from the soil and subsequent penetration through the leaf cuticle; and potential penetration of the compound through the roots and then translocation into the aerial parts of the plant.

To obtain more data on insecticidal residues in the aerial parts of the crops, peas were grown in an insecticide-treated quartz sand under greenhouse conditions, using extremely high concentrations of insecticide, and in a treated loam soil under field conditions. The object was to find out if insecticidal residues could be detected in stems and leaves at all, and if so, by what pathways they would find their way into the plant tissue. In addition, an attempt was made to determine the site of epoxidation of aldrin and heptachlor under such conditions.

Procedure

Laboratory Experiments. Peas (Wilt Resistant Alaska) were used throughout. They were grown in quartz sand treated at 30 p.p.m. with analytical grade lindane, aldrin, dieldrin, heptachlor, heptachlor epoxide, or *p,p'*-DDT. These conditions were extreme ones and not representative of agricultural practice. The quartz sand possesses neither the insecticide-retention qualities of any agricultural soil nor its microbial contents. Moreover, 30 p.p.m. (approximately 60 pounds per 6-inch acre) is far above what normally might be expected as an insecticidal residue content in agricultural soils. These extreme experimental conditions were selected to determine whether insecticides of extremely low water solubility could be translocated into plants through the root system.

The quartz sand was treated by pipetting measured amounts of an insecticide in acetone solution onto the sand contained in a wide-mouthed 1-gallon jar. The sand was mixed by rolling horizontally for 20 minutes on a specially designed apparatus (*8*). The acetone evaporation was aided by a very mild air stream.

Lindane was used for the first three sets of experiments because of its slight water solubility (to 10 p.p.m. at 20° C.) (*12*) and its relatively high vapor pressure, factors presumably favoring absorption by either roots or possibly the leaf cuticle.

1. Seventeen nonglazed clay pots (6 inches wide) were filled with lindane-treated sand. Twenty-four germinated pea seeds with a root length of $^3/_4$ to 1 inch were planted in each pot. In addition, 17 pots containing untreated sand were prepared in the same way.
2. Seventeen clay pots filled with lindane-treated sand were covered with extra-heavy aluminum foil. Twenty-four holes ($^1/_{16}$ to $^1/_8$ inch wide) were made in the aluminum foil of each pot. Peas were planted through those holes, the seed itself being left on top of the aluminum foil. Seventeen aluminum foil–covered pots containing untreated sand were prepared in the same way.

All pots stood in saucers and nutrient solution was added to the saucers as necessary (Figure 1). In this way water splashing was prevented. The aluminum foil also prevented excessive water loss and the possibility of codistillation of insecticidal vapors.

3. Ten peas were planted in untreated quartz sand within glass containers 2.5 inches wide. These jars were placed within clay pots 9 inches wide and surrounded by lindane-treated (30 p.p.m.) quartz sand (Figure 2). Nutrient solution was added as necessary to the surface of the untreated sand and distilled water to the surface of the treated sand.

4. Tests were conducted with aldrin, dieldrin, heptachlor, and heptachlor epoxide. For each insecticide used, 17 clay pots were filled with treated sand and covered with aluminum foil. Planting and watering were done as above. Seventeen control pots were similarly prepared.

5. Carbon-14–labeled *p,p'*-DDT was used, which had an activity of 79 counts per μg. per minute after subtraction of background. Because a relatively small amount of DDT was available, only one aluminum foil–covered pot containing DDT-treated quartz sand (30 p.p.m.) was planted with peas.

Twenty-one days after planting, when flowers started to develop, the plants were cut 1 inch above the sand surface. All plants from similarly treated sand were pooled and used for analyses. In addition, the sand was thoroughly mixed and aliquots were taken for analyses.

Field Experiments. In May 1958, duplicate Carrington silt loam plots (30 × 40 feet) were treated with aldrin and heptachlor at 5 and 25 pounds per acre and then rototilled to a depth of 4 to 5 inches. Treatment was done as described by Lichtenstein and Schulz (*9*). One year later the plots previously treated at the lower application rate were retreated with the same insecticides at 5 pounds per 5-inch acre. Two days after application in 1959, one 30-foot row was seeded with peas (Wilt Resistant Alaska) on both treated and control plots. At the end of July pea pods were harvested from the plants of each experimental plot. The seeds were separated from the pods. All seeds or all pods obtained from one particular plot were pooled for extraction and analyses.

Soils were sampled in May and September of 1959 as described by Lichtenstein (*5*).

Analytical Methods

Bioassays. Preliminary screening tests were conducted with two bioassay procedures.

Though chlorinated hydrocarbon insecticides have been reported (*10*) as inefficient aphicides, the pea aphid (*Macrosiphum pisi* Harris) was used. The bottom of a Petri dish (3.5 inches in diameter) was covered with moistened filter paper. Eight leaves taken at random from plants of one pot were placed at the periphery of the paper. In addition four stem pieces each 1 inch long were used. Twenty-five adult aphids were then introduced onto the center of the filter paper. Those aphids which did not start feeding after 15 minutes were replaced until all aphids had moved onto the plants. Four replicated Petri dishes containing plant material from one pot were set up. All together eight (2 × 4) tests were conducted with plants grown in two pots which contained sand treated with one particular insecticide. Mortality counts were made 29 hours after the pea aphids had been introduced. Corrections for check mortalities were made according to Abbott's formula (*1*).

In the second bioassay procedure *Drosophila melanogaster* Meig. was used as a test insect. Plants from two replicated pots were finely ground, dried with anhydrous sodium sulfate, and subsequently extracted with a mixture of hexane–isopropyl alcohol (2 to 1 by volume) by tumbling. After that the isopropyl alcohol was removed by washing with a saturated solution of sodium chloride. The hexane extract containing all the plant extractives ("crude") was then added quantitatively to small jars ($2^3/_4$ inches in diameter and 3 inches deep). After the solvent had been evaporated in a hood, 50 three-day-old flies were introduced to each jar. Mortality counts were made after 4 and 24 hours.

To minimize any possible masking effect from plant extractives, the residue in the jar was re-extracted after the first test with *Drosophila*. Hexane was used for re-extracting residues from plants grown in lindane-, aldrin-, or dieldrin-treated sand and pentane for the others. Waxes were removed by using acetonitrile (*4*). Hexane extracts were then added to a column (8 × 1 inch) of magnesium oxide–Celite 503 (1 to 1) and eluted with 600 ml. of redistilled hexane. The pentane extracts were cleaned up by using carbon as an absorbent, followed by a Florisil column (*7*) from which the residues were eluted with 200 ml. of a mixture of pentane-ether (94 to 6 by volume).

After the purified extracts had been concentrated, they were transferred quantitatively to the bioassay jars and the solvents were evaporated in a hood. Fifty 3-day-old flies were then introduced into each jar. Mortality counts were made at 24 and 48 hours.

Colorimetric Analyses. For extraction of pea vines the total amount of fresh plant material was passed through a food grinder. The macerated material weighing from 226 to 372 grams was then mixed with a double amount of anhydrous sodium sulfate and kept overnight in a refrigerator. The crop–sodium sulfate mixture was placed in 2-quart wide-mouthed Mason jars for extraction.

When pea pods or pea seeds were extracted, the total amount obtained from one field plot was ground and mixed and 300-gram aliquots were prepared for extraction as described above.

Sand and loam soils were screened and mixed prior to extraction. Four hundred grams of moist soil and 200 grams of anhydrous sodium sulfate were then placed in 2-quart wide-mouthed Mason jars. An additional portion of 100 grams of soil was dried for 24 hours at 50° C. to determine the dry weight of the soil.

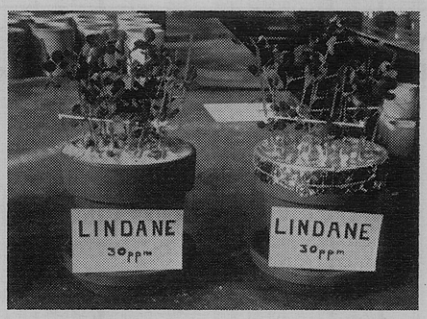

Figure 1. Peas growing in noncovered and covered pots containing lindane-treated sand

Figure 2. Peas growing in glass jars containing untreated sand

Surrounded by lindane-treated (30 p.p.m.) sand within clay pot

The extraction solvents used were a mixture of redistilled hexane and isopropyl alcohol (2 to 1 by volume) for aldrin, dieldrin, and DDT and a mixture of colorimetric pentane and acetone (4 to 1 by volume) for heptachlor and heptachlor epoxide. Two milliliters of solvent were used per gram of wet soil and 3 ml. of solvent per gram of fresh plant material.

After 1 hour of head-to-end tumbling, the jars and their contents were chilled to minimize evaporation of solvents during filtering. The supernatant liquid was then decanted through filter paper and the recovery volume was recorded to be used for calculating the results.

The isopropyl alcohol or the acetone was removed from the extracts by washing once with water and three times with a saturated solution of sodium

Table I. Mortalities of *Drosophila melanogaster* Meig. after Exposure to Extracts of Pea Vines Grown in Insecticide-Treated Quartz Sand (30 P.M.)

Insecticide Used	Plant Material Extracted, Grams	Hours of Exposure: 4	24	24	48
		Mortality from Plant Extract, %			
		Crude[a]		Purified[b]	
None	16.3	0	0	0	6
Lindane[c]	14.9	100	..	..	..
Lindane surrounded[d]	5.0	0	0	0	0
Aldrin	9.9	0	0	2	28
Dieldrin	10.4	0	0	0	36
Aldrin + dieldrin[e]	17.5	0	0	0	52
Heptachlor	17.5	0	0	80	..
Heptachlor epoxide	16.2	0	0	88	..

[a] Containing hexane-isopropyl alcohol–soluble materials.
[b] Purified by acetonitrile treatment and column chromatography.
[c] From covered and noncovered pots.
[d] Plants grown in untreated sand, but surrounded by lindane-treated sand.
[e] Plant material obtained from peas grown in two different pots, one containing aldrin-treated sand, the other dieldrin-treated sand.

Table II. Insecticidal Residues Recovered, P.P.M., from Sand and Pea Vines Grown in Treated Quartz Sand (30 P.P.M.)

Insecticides: Treated	Insecticides: Recovered	Sand, Chem. Anal.	Pea Vines: Chem. anal.	Pea Vines: Bioassay
Lindane	Lindane			
Open[a]		20.2 ± 0.2	18.0 ± 0.7	
Covered[b]		23.8 ± 0.3	21.9 ± 5.8	
Surrounded[c]		0.0	0.0	0.0
Aldrin[b]	Aldrin	24.85 ± 1.25	0.0	Traces
	Dieldrin	0.11 ± 0.07	0.34 ± 0.01	0.34 ± 0.02
Heptachlor[b]	Heptachlor	20.60 ± 1.30	Traces	Traces
	Heptachlor epoxide	Traces	0.17[d]	0.18 ± 0.06
Dieldrin[b]	Aldrin	0.0	0.0	
	Dieldrin	24.45 ± 0.9	1.08[e]	0.87 ± 0.09
Heptachlor epoxide[b]	Heptachlor	0.0	0.0	
	Heptachlor epoxide	29.30 ± 0.4	0.19[d]	0.22 ± 0.05
		Radioassay	Radioassay	
DDT (C^{-14})[b]	DDT (C^{-14})	26.70 ± 0.6	0.01	0.0

[a] Grown in noncovered pots.
[b] Grown in aluminum foil–covered pots.
[c] Grown in untreated sand but surrounded by lindane-treated sand.
[d] Absorption peak at 415 mμ. R_f value same as obtained from reference grade heptachlor epoxide.
[e] *Rf* value same as obtained from reference grade dieldrin.

chloride. The alcohol- or acetone-free phase was then dried over anhydrous sodium sulfate.

Extracts containing aldrin and dieldrin were cleaned up by using a column (8 × 1 inch) of magnesium oxide–Celite 503 (1 to 1). After the extract had been added to the column, aldrin was eluted with 100 ml. of redistilled hexane. Dieldrin could then be washed from the column with an additional 600 ml. of the same solvent. Aldrin and dieldrin fractions were analyzed according to the method of O'Donnell *et al.* (*13*, *14*). Known amounts of aldrin added to soils were recovered to an extent of 93 to 97%, and known amounts of dieldrin to an extent of 90 to 94%. The respective figures obtained when aldrin or dieldrin was added to crop material were 88 to 92%.

Extracts containing heptachlor or heptachlor epoxide were cleaned up as described by Lichtenstein and Polivka (*7*). Waxes contained in extracts of pea seeds or pea pods were removed by using acetonitrile (*4*). The two insecticides were analyzed according to the Polen-Silverman method (*15*, *16*). Known amounts of heptachlor or heptachlor epoxide added to soil were separately recovered to an extent of 90 to 96%. The figures obtained when crop material was used were 85 to 91% for heptachlor and 90 to 92% for heptachlor epoxide.

DDT extracts were cleaned up by passing through a column (6 × 1 inch) of aluminum oxide (reagent, Baker and Adamson). Analysis was done by radioassay using a Geiger-Muller counter, after the extract had been evaporated to dryness in planchettes. Results were corrected for self-absorption.

Lindane was analyzed according to the Schechter-Hornstein method (*17*). A special extraction procedure was eliminated (*6*), which permitted the determination of lindane directly in soils and crops. Known amounts of lindane added to soils and crops were recovered at approximately 100%.

In some cases colorimetric absorption curves were established for the recovered residue to check if the peak obtained from the unknown sample coincided with the peak obtained from a sample to which a known amount of an insecticide had been added. Where the amount of available material permitted, analyses were run in duplicate, using a soil or a crop blank for the determination of apparent insecticide content. In all cases known amounts of insecticides were added to insecticide-free samples. This permitted a check of the analytical procedure for each analysis done. The unknowns, after the value for apparent insecticide had been subtracted, were calculated on the basis of the values obtained from the known amounts. Results were expressed in parts per million, based on the dry weight for soils and the fresh weight for plants.

Fractions of extracts containing aldrin, dieldrin, heptachlor, or heptachlor epoxide were used for ascending paper chromatography as described by Mitchell (*11*). R_f values for the unknowns were established and compared to R_f values obtained from reference grade insecticides. In addition, aliquots of the extracts were used for a *Drosophila* bioassay. Mortalities obtained with unknown samples were compared to dosage-mortality standard curves obtained from serial dilutions of reference grade insecticide.

Data were reported as zero when results obtained by colorimetric analyses as well as paper chromatography were negative. Where the colorimetric analysis indicated the absence of insecticidal residues but R_f values obtained from the unknowns were the same as those obtained from reference grade insecticide, data were reported as "traces." When bioassay procedures were employed, results were reported as traces when mortalities obtained were below 15% (corrected for check mortalities).

Results and Discussion

Laboratory Experiments. Screening Tests by Bioassay. None of the plant material from pea plants grown in insecticide-treated quartz sand was toxic to the pea aphids, except that derived from the lindane experiment. Exposure to plant material obtained from plants grown in noncovered pots containing lindane-treated sand yielded 26± 9% mortality of the pea aphids. When plant material from covered pots was used, 63.5± 15% of the pea aphids died 29 hours after exposure. No insect mortality was observed on leaves

and stems obtained from plants grown in untreated sand surrounded by lindane-contaminated sand.

These results indicate that toxic material was present in the aerial parts of pea plants grown in lindane-treated sand. Moreover, more toxic material was present in plants grown in lindane-treated sand covered with aluminum foil.

The screening results with *Drosophila* are summarized in Table I. Crude extracts from plant material grown in lindane-treated sand were the only ones that caused any fly mortality. Purified plant extracts from peas grown in heptachlor- or heptachlor epoxide–treated sand killed nearly all the flies within 24 hours. Forty-eight hours' exposure of the flies yielded 24 to 49% mortality [corrected for check mortalities according to Abbott's formula (*1*)] with purified extracts of plants grown in aldrin- or dieldrin-treated sand. However, in the latter case, the amount of plant material available was only half of that used when flies were exposed to plant extracts obtained from peas grown in heptachlor- or heptachlor-epoxide–treated sand.

Chemical Analyses and Quantitative Bioassays. Insecticidal residues recovered from the aerial parts of pea plants grown in insecticide-treated sand are summarized in Table II.

If lindane vapors had been absorbed through the leaf cuticle, most of the insecticide would have been found within the plants grown in uncovered pots and possibly some in those grown in untreated sand surrounded by lindane-contaminated sand. The amount of lindane residues in leaves and stems of peas was greater than that recovered when the other insecticides were used. Plants grown in covered pots contained 22 p.p.m. of lindane, which was similar to that recovered from the sand. Volatilization of lindane and subsequent absorption of the insecticide through the leaves were considerably reduced by the aluminum foil cover. Thus the insecticide apparently entered the leaves and stems through the roots.

More lindane was lost by the sand in the noncovered than in the covered pots, probably because of a greater volatilization of the lindane from the sand surface. Plants grown in noncovered and covered pots contained similar amounts of lindane.

Under the described conditions no lindane residue was found in the plants growing in untreated sand surrounded by lindane-treated sand (Figure 2). Therefore, it seems rather unlikely that an aerial translocation of lindane took place.

Plants grown in aldrin- or heptachlor-treated sand contained only the respective epoxides: dieldrin (1.4% of aldrin residue in the sand) or heptachlor epoxide (0.83% of heptachlor residue in the sand). However, the amount of dieldrin in the sand was only 0.5% of the amount of aldrin recovered, and only traces of heptachlor epoxide were detected in the heptachlor-treated sand.

The epoxidation of heptachlor and especially aldrin is dependent on biological factors (*9*). Since quartz sand exhibits negligible biological activities, it was not surprising that the epoxidation of aldrin to dieldrin in this substrate was very small as compared to various soil types. It seems, therefore, that aldrin and heptachlor penetrated the root system and epoxidation took place within the plant tissue.

When plants were grown in dieldrin- or heptachlor epoxide–treated sand, only dieldrin (4% of dieldrin residue recovered from the sand) or heptachlor epoxide (0.7% of heptachlor epoxide residue recovered from the sand) was found in the aerial parts of the plants. Aldrin or heptachlor residues could not be detected in either the sand or the plant tissue.

Pea plants grown in sand treated with carbon-14–labeled DDT did not show any translocation of this insecticide. On the basis of extraction of plant material and radioassay, less than 0.01 p.p.m. of carbon-14 derived from DDT was found.

The availability of insecticides in soils to plants decreases with the increase of organic matter in soils (*5*). Therefore, repetition of the described experiments with agricultural soils would probably result in much lower residues or none at all within the plant tissue. Using economic dosages of insecticide would undoubtedly further reduce the residue content.

Table III. Recoveries of Aldrin (A), Dieldrin (D), Heptachlor (H), and Heptachlor Epoxide (HO) Residues

(From a Carrington silt loam and peas grown in 1959 on aldrin- or heptachlor-treated soils)

	Insecticides Applied to Soil, Lb. per 5-Inch Acre							
	Aldrin				Heptachlor			
	5[a]		25[b]		5[a]		25[b]	
	Recovered from Soils,[c] P.P.M.							
A	1.78		3.10		H	2.00		4.22
D	0.66		1.83		HO	0.39		0.78
T[d]	2.44		4.93		T	2.39		5.00
	Recovered from Seeds, P.P.M.							
	Chem. anal.	Bio-assay	Chem. anal.	Bio-assay		Chem. anal.	Bio-assay	Chem. anal.	Bio-assay
A	0.0	0.0	0.0	0.0	H	0.0	0.0	0.0	0.0
D	0.0	0.0	0.0	0.0	HO	0.0	0.0	0.0	<0.01
T	0.0	0.0	0.0	0.0	T	0.0	0.0	0.0	<0.01
	Recovered from Pods, P.P.M.								
A	0.0	0.0	0.0	0.0	H	0.0	0.0	0.0	0.0
D	0.0	<0.02	0.0	<0.02	HO	0.03[e]	0.03	0.04[e]	0.05
T	0.0	<0.02	0.0	<0.02	T	0.03	0.03	0.04	0.05

[a] Treated at 5 lb. per 5-inch acre in May 1958 and May 1959.
[b] Treated at 25 lb. per 5-inch acre in May 1958 only.
[c] Average of spring and fall analyses (chemical).
[d] Sum of aldrin and dieldrin or heptachlor and heptachlor epoxide, respectively.
[e] R_f value same as obtained from reference grade heptachlor epoxide.

Field Experiments

Results obtained from seeds and pods from pea plants grown in either aldrin- or heptachlor-treated Carrington silt loam are summarized in Table III. The total amounts of aldrin and dieldrin or heptachlor and heptachlor epoxide recovered from the soil during the summer of 1959 were considerably larger than would be present normally. Seeds from pea plants grown in these soils did not contain measurable amounts of insecticidal residues; neither did pods from plants grown on aldrin-treated soil. However, in pods from plants grown on heptachlor-treated soils, 0.03 to 0.04 p.p.m. of heptachlor epoxide was found as determined by chemical analysis, bioassay, and paper chromatography.

Therefore, under normal conditions no aldrin or heptachlor or their epoxides would be expected in pea seeds or pods from aldrin- or heptachlor-treated loam soils.

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ISOTOPE-LABELED INSECTICIDES

Synthesis of Carbon-14-Labeled Aldrin and Dieldrin

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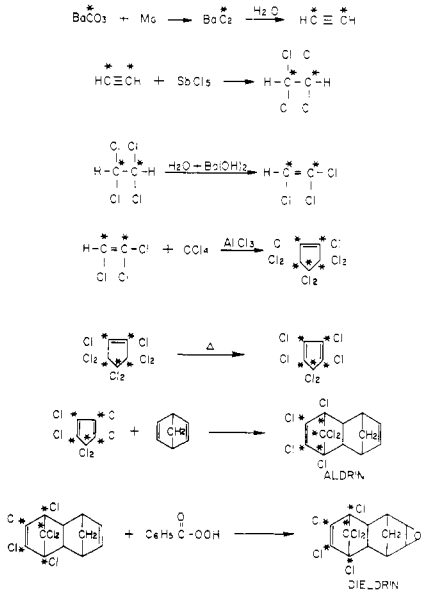
The insecticides 1,2,3,4,10,10-hexachloro-1,4,4a,5,8,8a-hexahydro-1,4-*endo*-exo-5,8-dimethanonaphthalene (aldrin) and 1,2,3,4,10,10-hexachloro-6,7-epoxy-1,4,4a,5,6,7,8,-8a-octahydro-1,4-*endo*-exo-5,8-dimethanonaphthalene (dieldrin) were prepared labeled with carbon-14. They were synthesized by labeling hexachlorocyclopentadiene and subsequent reaction with 2,5-norbornadiene. Starting with $BaC^{14}O_3$, a 28% yield of aldrin (melting point 102.5° C.) and a 22% yield of dieldrin (melting point 181° C.) were obtained. The specific activities were 3.6 and 3.5 ± 0.1 mc. per gram for aldrin and dieldrin, respectively.

DURING STUDIES at this laboratory on the fate of aldrin and dieldrin in susceptible and resistant strains of insects and in higher animals it became evident that the chemical methods now available for these compounds are not adequate, especially when an attempt is made to account fully for the dosage applied. It was obvious that C^{14}-labeled aldrin and dieldrin would be a great aid in our studies and, accordingly, their synthesis was undertaken. The present paper is a report on this work.

Aldrin is ordinarily prepared by the Diels-Alder reaction of hexachlorocyclopentadiene with 2,5-norbornadiene (bicyclo[2,2,1]hepta - 2,5 - diene), and dieldrin is prepared by the peracid oxidation of aldrin. The problem was to prepare either hexachlorocyclopentadiene or 2,5-norbornadiene with a C^{14} label. Synthesis of C^{14}-labeled hexachlorocyclopentadiene appeared to be the most promising approach.

The following six- and seven-step reaction scheme was used in the synthesis of C^{14}-labeled aldrin and dieldrin:



Carbon-14 labeled acetylene was prepared by a modification of the method of Cramer and Kistiakowski (*2*). The yields were approximately 70%, as determined by gas volumetric measurements with inactive runs.

The labeled acetylene was converted to tetrachloroethane by treatment with a mixture of antimony pentachloride and trichloride. This reaction as given by Krall (*3*) was found to give very low yields. After the acetylene had been complexed with the antimony pentachloride, it was necessary to decompose the complex by heating for a much longer period of time than that used by Krall to obtain satisfactory yields. With inactive acetylene, 85% yields were consistently obtained in this reaction.

Trichloroethylene was prepared by refluxing the tetrachloroethane with barium hydroxide and steam-distilling the product. Yields of 85 to 90% were obtained with inactive trichloroethylene.

Octachlorocyclopentene was prepared from the trichloroethylene by the method of Prins (*4*). This reaction gave a 70% yield.

The dechlorination of octachlorocyclopentene to hexachlorocyclopentadiene and the subsequent conversion to aldrin and dieldrin were based on