

CHROM. 8827

GAS CHROMATOGRAPHIC IDENTIFICATION OF SOME ORGANOCHLORINE PESTICIDES AND THEIR PHOTOALTERATION PRODUCTS BY MEANS OF KOVATS' RETENTION INDICES

F. I. ONUSKA and M. E. COMBA

Canada Centre for Inland Waters, Analytical Methods Research Section, P.O. Box 5050, 867 Lakeshore Road, Burlington, Ontario L7R 4A6 (Canada)

(Received August 22nd, 1975)

SUMMARY

Kovats' retention indices and temperature coefficients for some chlorinated methanonaphthalene and methanoindene pesticides and their photoproducts were determined on an OV-17 silicone liquid phase at three different temperatures. A detailed procedure for the off-line calculation of the retention indices, $\partial I/\partial T$ and I values are described. The program, written in BASIC, calculates the indices at three different temperatures and provides a computer-written report.

INTRODUCTION

Occurrence of photoaltered species of bridged polycyclic chlorinated hydrocarbons in the environment is of considerable importance and often goes undetected due to a lack of structural information. Several methods are available for their determination but these are either time-consuming, or highly specialized, and not normally performed in routine monitoring laboratories. In view of this, we focused our attention on the determination of Kovats' retention indices of some previously identified metabolites and organochlorine pesticides¹⁻³. Standardization of retention data in the form of a retention index system was proposed by Kovats⁴ and Ettre⁵. The retention index (I) of the substance eluted from a column relates its retention time to the retention times of a series of n -alkanes on the same stationary phase and thus becomes independent of other experimental parameters. Furthermore, it has been shown that identical structural differences between pairs of compounds lead to practically identical retention index differences⁶. Takacs and coworkers⁷⁻¹⁰ have shown that Kovats' retention indices may be predicted and are directly proportional to the free energy of solution of a compound in a stationary phase, which, in turn, can be related to the functional groups constituting the molecule.

Thus, retention indices are not only useful as compound-specific data, but may provide structural information also.

EXPERIMENTAL

Apparatus

The work was performed using a Perkin-Elmer Model 990 gas chromatograph with a flame ionization detector. As the temperature scale on this instrument could not be read to better than $\pm 0.5^\circ$, this temperature error was taken into account. A 20-min waiting period for column equilibration was considered to be sufficient for accurate retention times within the error limit.

A glass column (184 cm \times 2.1 mm I.D.) packed with 3% OV-17 silicone oil on Chromosorb W-HP was used. The carrier gas was helium at a flow-rate of about 30 ml/min.

Determination of retention indices

Retention indices were calculated using a Varian 620L-SS 100 MS computer or Wang 600 calculator. Retention times were measured by means of a stopwatch. A program was written in BASIC and is detailed in Appendix I.

The particular photoaltered product was mixed with an appropriate number of *n*-alkanes, or when overlapping prevented this method, the sample and mixture of *n*-alkanes were run alternately. The gas hold-up time was evaluated by injecting methane. The difference between the value thus obtained and the true gas hold-up time was insignificant in the present instance as the lowest retention indices determined were as high as 2008. The three standard temperatures used for measurement of retention indices were 190, 200, and 210°. In addition, the retention indices of the photoendrin were measured at 210, 220 and 230° and retention indices were obtained at lower temperatures by extrapolation.

RESULTS AND DISCUSSION

The photoisomerization of heptachlor, chlordene and 1-hydroxychlordene corresponded to the photo-conversion of *endo*-dicyclopentadiene derivatives to pentacyclodecanes and resultant caged structures. The chromatograms of irradiated heptachlor and chlordene (Figs. 1 and 2) showed both completely converted to their respective full-caged isomers. 1-Hydroxychlordene formed two photoproducts, which are well separated by gas chromatography (Fig. 3). Peak 1 represented a dehydrochlorinated product of 1-hydroxychlordene with subsequent formation to a half-caged ketone structure. The lower value obtained for its retention index can be attributed to the presence of the carbonyl functional group and dehydrochlorination as compared to the caged photoisomer (peak 2), where the hydroxyl group was retained and the index much higher. These values are summarized in Table I, with the retention data obtained for heptachlor and chlordene.

Dieldrin formed one photoisomer, as demonstrated in Fig. 4 and exhibited a large ΔI in comparison to the other photoproducts observed. This may be attributed to the formation of the caged structure with retention of the epoxide functional group.

In contrast, six photoproducts were observed on the chromatogram of irradiated aldrin (Fig. 5). The isomer of aldrin formed, represented by peak 5, occurred in a very low yield and displayed a difference of $\Delta I^{210} = 15$ in Kovats' retention index compared to a standard of photoaldrin. The major product observed was a hydro-

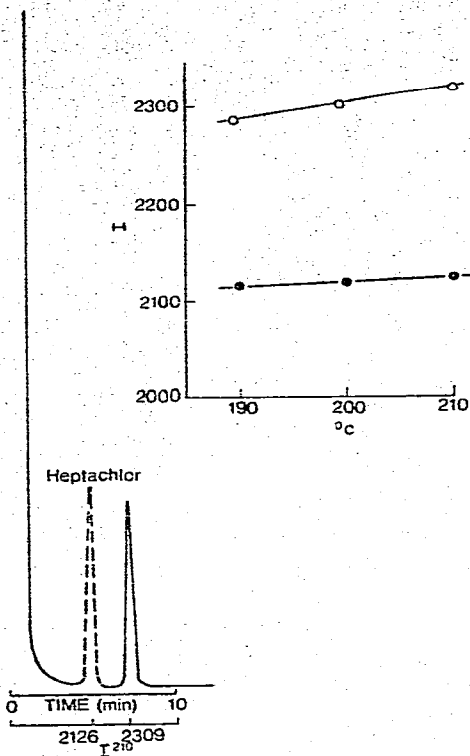


Fig. 1. Gas chromatogram of irradiated heptachlor.

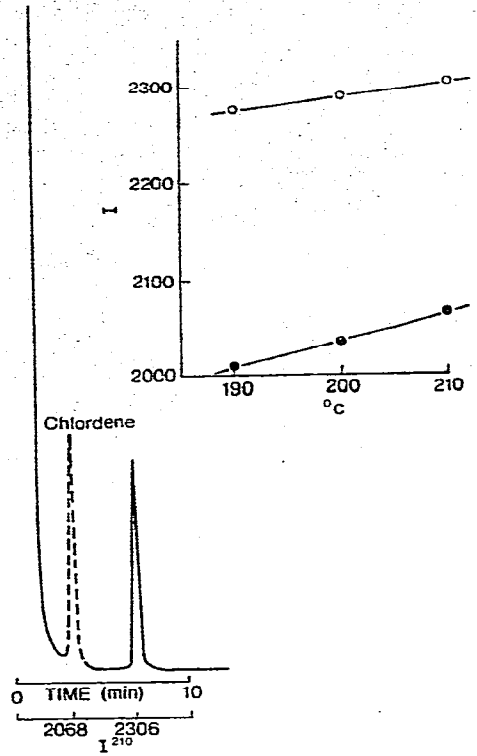


Fig. 2. Gas chromatogram of irradiated chlordene.

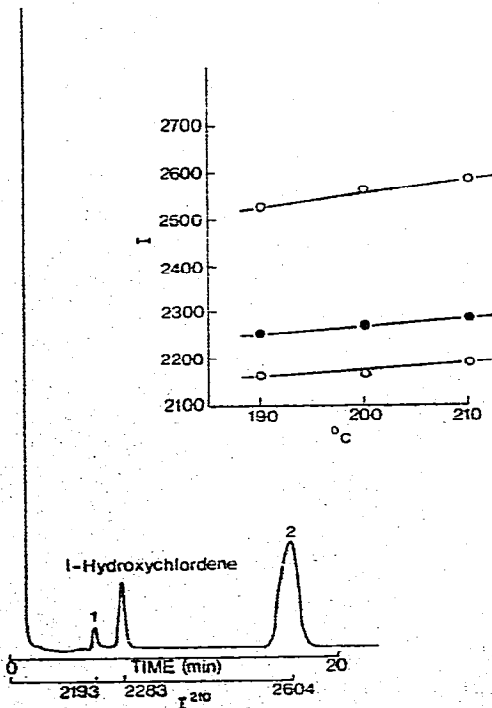
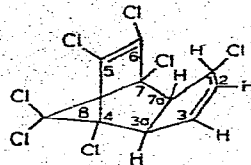
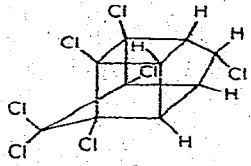
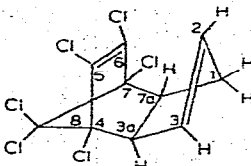
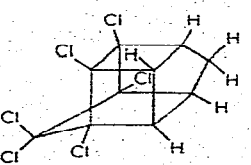
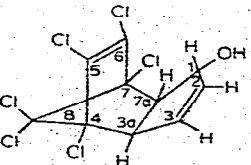
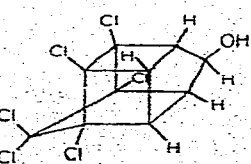
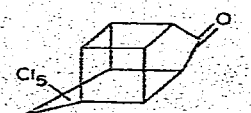


Fig. 3. Gas chromatogram of irradiated 1-hydroxychlordene.

TABLE I

KOVATS' RETENTION DATA FOR HEPTACHLOR, CHLORDENE, 1-HYDROXYCHLORDENE AND THEIR PHOTOPRODUCTS

 V_n = Net retention volume.

Compound	Peak No.	t^{210}	$\theta I (10^\circ C)$	ΔI	V_n	Structure
Heptachlor		2125.8	3.2	0.0	70.0	
Photoheptachlor		2308.6	17.7	182.8	94.3	
Chlordene		2067.8	29.9	0.0	31.9	
Photochlordene		2305.8	15.2	238.0	93.4	
1-Hydroxychlordene		2283.2	13.3	0.0	96.5	
Photo-1-hydroxy-chlordene	2	2604.0	38.8	320.8	269.8	
1-Hydroxychlordene photoproduct	1	2192.7	63.9	-90.5	70.4	

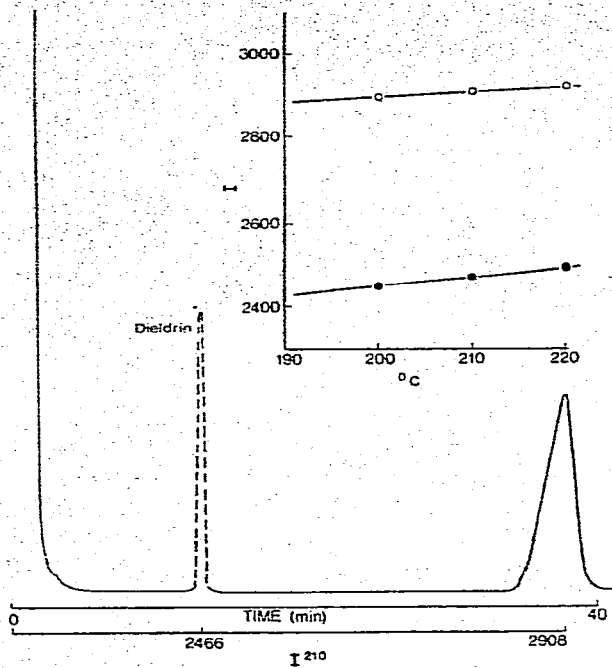


Fig. 4. Gas chromatogram of irradiated dieldrin.

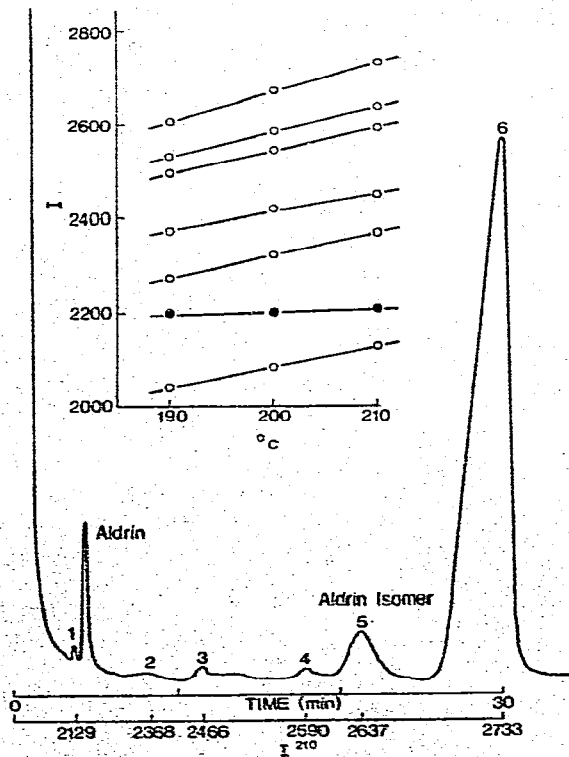
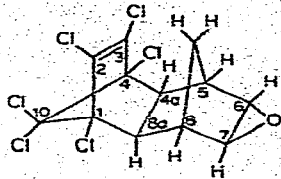
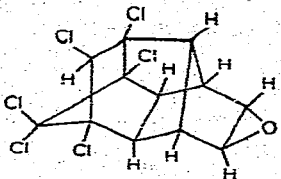
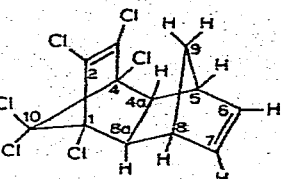
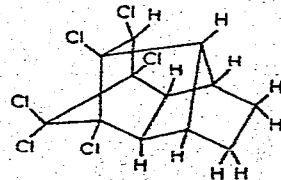


Fig. 5. Gas chromatogram of irradiated aldrin.

TABLE II
KOVATS' RETENTION DATA FOR ALDRIN, DIELDRIN AND THEIR PHOTOPRODUCTS

Compound	Peak No.	I^{210}	$\Delta I (10^\circ\text{C})$	ΔI	V_r	Structure
Dieldrin		2466.2	16.0	0.0	1195.2	
Photodieldrin		2908.0	9.5	441.8	179.2	
Aldrin		2206.2	5.6	0.0	55.0	
Aldrin photoproduct	1	2129.3	44.5	-76.9	47.3	unidentified
Aldrin photoproduct	2	2367.9	47.4	161.7	120.9	unidentified
Aldrin photoproduct	3	2465.9	23.9	259.7	176.4	unidentified
Aldrin photoproduct	4	2589.7	46.2	383.5	281.0	unidentified
Aldrin photoproduct	5	2636.9	52.4	430.7	335.4	$\text{C}_{10}\text{H}_8\text{Cl}_6$ (caged)
Aldrin photoproduct	6	2732.8	63.4	526.6	475.4	

generated, half-caged isomer of aldrin. The retention data for the remaining unidentified aldrin photoproducts are given in Table II with the data found for dieldrin.

cis-Chlordane formed one photoisomer after UV irradiation while *trans*-chlordane formed two photoproducts, as shown in Figs. 6 and 7. From the data in Table III, *cis*-chlordane had an increase in retention index of $\Delta I^{210} = 28.5$ compared to *trans*-chlordane, for the *endo-exo* chlorine-hydrogen atoms on the C_2 position. Whereas *cis*-chlordane formed the C_2 to C_5 or C_6 caged photoisomer, *trans*-chlordane

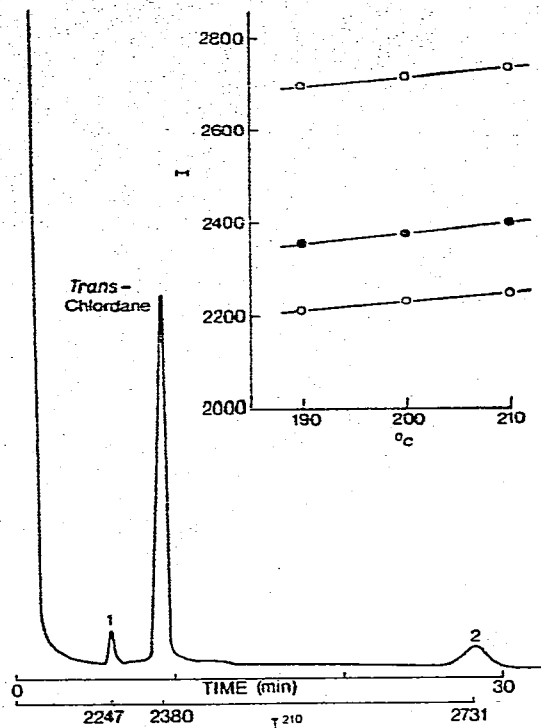
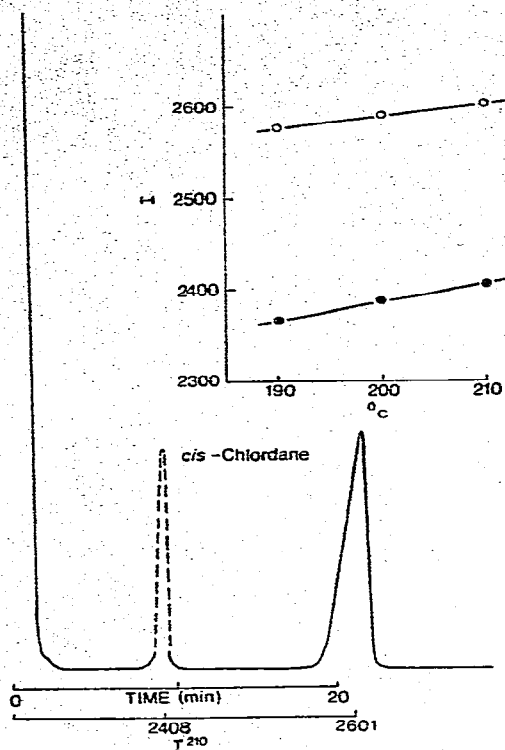
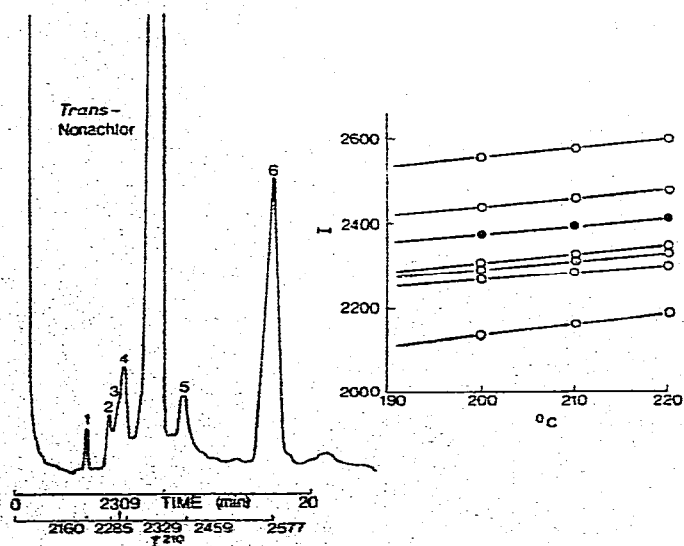
Fig. 6. Gas chromatogram of irradiated *cis*-chlordane.Fig. 7. Gas chromatogram of irradiated *trans*-chlordane.Fig. 8. Gas chromatogram of irradiated *trans*-nonachlor.

TABLE III
KOVATS' RETENTION DATA FOR *cis*- AND *trans*-CHLORDANE AND THEIR PHOTO-PRODUCTS

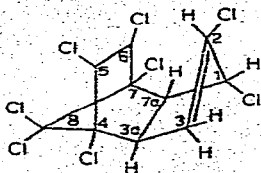
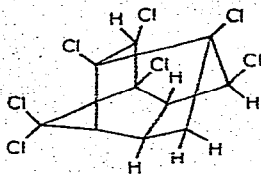
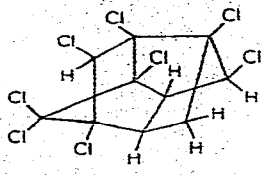
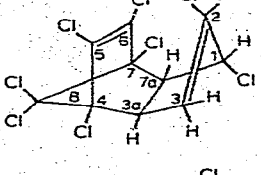
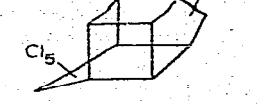
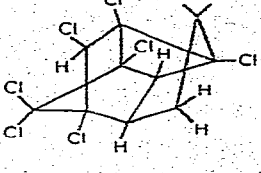
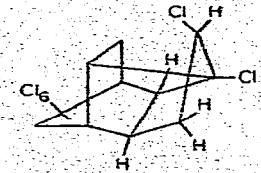
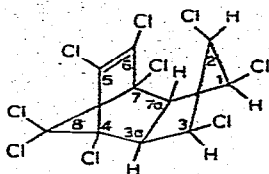
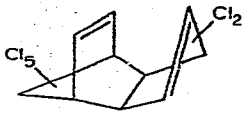

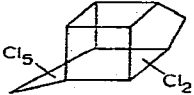
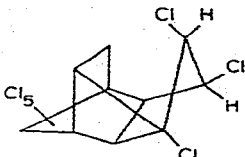
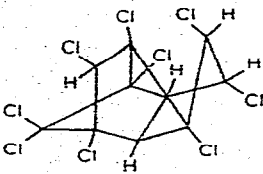
Compound	Peak No.	I^{210}	$\partial I (10^\circ C)$	ΔI	V_g	Structure
<i>cis</i> -Chlordane		2408.5	19.9	0.0	142.4	
Photo- <i>cis</i> -chlordane		2600.9	14.1	192.4	340.7	 or 
<i>trans</i> -Chlordane		2380.0	24.9	0.0	135.2	
<i>trans</i> -Chlordane photoproduct	1	2247.2	38.5	-138.8	83.1	
Photo- <i>trans</i> -chlordane	2	2731.5	18.5	345.5	452.0	 or 

TABLE IV

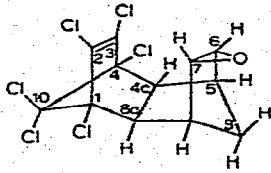
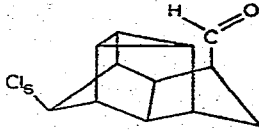
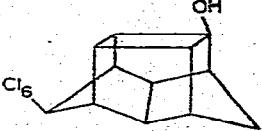
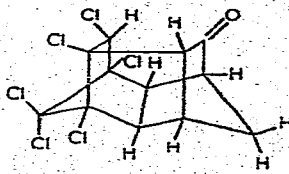
KOVATS' RETENTION DATA FOR *trans*-NONACHLOR AND ITS PHOTOPRODUCTS

Compound	Peak No.	I^{210}	$\partial I (10^\circ C)$	ΔI	V_x	Structure
<i>trans</i> -Nonachlor		2397.1	25.4	0.0	136.6	
<i>trans</i> -Nonachlor photoproduct	1	2159.5	24.2	-238.6	70.0	
<i>trans</i> -Nonachlor photoproduct	2	2285.2	18.0	-111.9	98.3	
<i>trans</i> -Nonachlor photoproduct	3	2309.0	17.7	- 88.1	110.0	photoheptachlor
<i>trans</i> -Nonachlor photoproduct	4	2329.0	21.0	- 68.1	115.8	
<i>trans</i> -Nonachlor photoproduct	5	2459.1	16.0	62.0	179.0	
Photo- <i>trans</i> -nonachlor	6	2576.6	21.5	179.5	276.4	

formed the C_1 to C_5 or C_6 caged structure with a difference of almost 50% in retention units, indicating the significant change in caging positions for the chlordane stereoisomers. The second *trans*-chlordane photoproduct (peak 1) was a dechlorinated, full-caged product of *trans*-chlordane, having a molecular composition of $C_{10}H_6Cl_6$.

Peaks 1 and 2 in the gas chromatogram of irradiated *trans*-nonachlor (Fig. 8) were dechlorinated products with respective molecular compositions of $C_{10}H_5Cl_7$ and $C_{10}H_6Cl_8$, with no apparent intramolecular bridging in their structures. Peak 3 was confirmed to be photoheptachlor, while peaks 4 and 5 were identified as dechlorinated $C_{10}H_5Cl_7$ and $C_{10}H_6Cl_8$ caged photoaltered products. Peak 6 was a *trans*-nonachlor analog with intramolecular bridging similar to that found for *trans*-chlordane and one of the four C_1 or C_3 to C_5 or C_6 isomeric possibilities. Table IV

TABLE V
KOVATS' RETENTION DATA FOR ENDRIN AND ITS PHOTOPRODUCTS

Compound	Peak No.	I^{210}	$\delta I (10^\circ C)$	ΔI	V_n	Structure
Endrin		2527.9	24.6	0.0	222.3	
Endrin photoproduct 1	1	2247.0	4.2	-280.9	-	unidentified
Endrin photoproduct 2	2	2436.0	8.0	-91.9	-	$C_{11}H_5Cl_6$
Endrin photoproduct 3	3	2470.0	16.0	-57.9	182.7	dieldrin
Endrin photoproduct 4	4	2619.5	25.0	91.6	-	caged isomer
Endrin (aldehyde)	5	2668.3	14.5	140.4	388.0	
Endrin (alcohol)	6	2704.6	17.4	176.4	-	
Endrin (ketone)	7	2822.5	23.5	294.6	-	
Endrin photoproduct 8	8	2844.8	24.7	316.9	730.0	unidentified
Endrin photoproduct 9	9	2877.2	25.6	349.3	-	unidentified

summarizes the Kovats' retention data and the structure found for the photoproducts of *trans*-nonachlor. The retention data obtained for endrin and its photoproducts (Table V) was quite reproducible, although structural changes were observed to occur by mass spectrometry. This was probably due to the metal connectors in the gas chromatographic manifold although endrin itself was not found to thermally decompose on the gas chromatograph. Peak 2 in the gas chromatogram of irradiated endrin (Fig. 9) was an uncaged $C_{11}H_8Cl_6$ product.

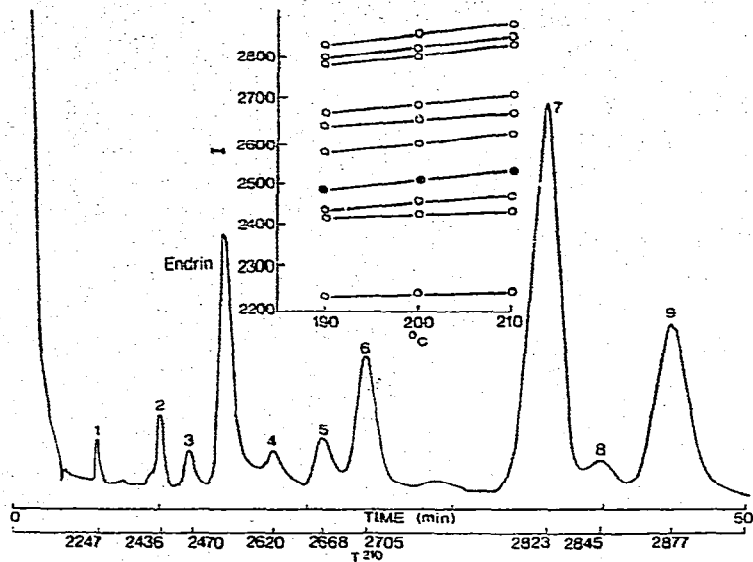


Fig. 9. Gas chromatogram of irradiated endrin.

Peaks 3, 4, 5, 6 and 7 were isomeric forms of endrin. Peak 3 was established to be dieldrin; peak 4, although not confirmed, was highly suspected to be a form of endrin aldehyde; peak 5 was in agreement with data obtained by Harrison *et al.*¹¹ for endrin aldehyde; peak 6 was identified as the endrin alcohol, previously reported by Nash *et al.*¹²; the major product (peak 7) was identified as endrin ketone.

APPENDIX I

Description of program used to calculate Kovats' retention indices (Figs. 10 and 11)

This program has been written so that the user may enter his data by using the conversational mode of the program.

The program will calculate the retention index number of any compound separated by gas chromatography by using the following formula:

$$I^{\circ} = 100 \cdot \frac{(\log t_x - \log t_{cn})}{(\log t_{cn+1} - \log t_{cn})} + 100 Z$$

where t_x is the retention time of a particular compound and t_{cn} and t_{cn+1} are the retention times for the hydrocarbons of two consecutive n -alkanes with carbon numbers n and $n+1$, respectively, and which bracket the compound x .

```
10 DIM D(10,10),C(20,6),A(10),I(10,10)
20 REM TO CALCULATE KOVATS INDICES
30 PRINT
32 PRINT
34 PRINT "PRINT # OF SAMPLES TO BE RUN"
35 INPUT Y1
36 FOR L= 1 TO Y1
37 PRINT "PRINT # OF TEMPERATURES USED"
60 INPUT T
70 PRINT "PRINT TEMPERATURES"
80 FOR I= 1 TO T
90 INPUT D( 1,I)
100 NEXT I
110 PRINT "PRINT # OF RETENTION TIMES & # OF COMPOUNDS"
120 PRINT "RETENTION TIMES FOUND AT ONE TEMPERATURE"
130 INPUT N1,N2
140 PRINT "PRINT # OF C ATOMS & RETENTION TIMES IN SECONDS"
150 PRINT "FOUND AT EACH TEMPERATURE"
155 FOR I= 1 TO N1
157 FOR J= 1 TO T+ 1
160 INPUT C(I,J)
163 NEXT J
165 NEXT I
200 PRINT "PRINT RETENTION TIME IN SEC FOR EACH COMP AT EACH TEMP"
210 FOR I= 2 TO N2+ 1
220 FOR J= 1 TO T
230 INPUT D(I,J)
240 NEXT J
250 NEXT I
260 FOR J= 1 TO T
270 FOR I= 2 TO N2+ 1
280 LET K= 1
```

Fig. 10.

```

300 IF D(I,J)>C(K,J+ 1) THEN 335
310 LET K=K+ 1
330 GOTO 300
335 IF D(I,J)>=(C(K+ 1,J+ 1)) THEN 310
340 LET B1= 1/ 2.3025?
342 LET B2=LOG(D(I,J))*B1
344 LET B3=LOG(C(K,J+ 1))*B1
346 LET B4=LOG(C(K+ 1,J+ 1))*B1
350 LET I(I- 1,J)= 100*((B2-B3)/(B4-B3))+ 100*C(K, 1)
360 NEXT I
370 NEXT J
380 FOR I= 1 TO N2
390 FOR J= 1 TO T- 1
400 LET I(I,J+T)=I(I,J+ 1)-I(I,J)
410 NEXT J
420 NEXT I
430 PRINT "          KOVATS' RETENTION INDICES"
440 PRINT "          ====="
450 PRINT
460 PRINT "COLUMN USED :"
470 PRINT "SAMPLE ANALYZED :"
480 PRINT
490 PRINT
500 PRINT "          RETENTION INDICES"
510 PRINT "          ====="
520 PRINT
530 PRINT TAB( 14);"I1";TAB( 27);"I2";TAB( 40);"I3"
540 PRINT "COMPOUND";TAB( 14);"@";TAB( 27);"@";TAB( 40);"@"
550 PRINT TAB( 12);D( 1, 1);TAB( 25);D( 1, 2);TAB( 38);D( 1, 3)
600 PRINT
610 FOR I= 1 TO N2
640 PRINT TAB( 13);I(I, 1);TAB( 26);I(I, 2);TAB( 39);I(I, 3);TAB( 52);
641 PRINT I(I, 4);TAB( 63);I(I, 5)
660 NEXT I
670 NEXT L
680 END

```

Fig. 10. BASIC program calculating Kovats' retention indices. For variables, see the list on p. 398. L and J are index counters for the variables Y and T for values of (1 to Y1) and (1 to T+1). B1 = Conversion factor to natural logarithms; B2 = t_x ; B3 = t_{cn} ; B4 = t_{cn+1} .

These indices will be calculated for each compound at one particular temperature. A maximum of ten compounds, and twenty hydrocarbon retention times can be entered. As well as calculating the index numbers, the program calculates and prints the differences between the particular compounds of interest at the different temperatures.

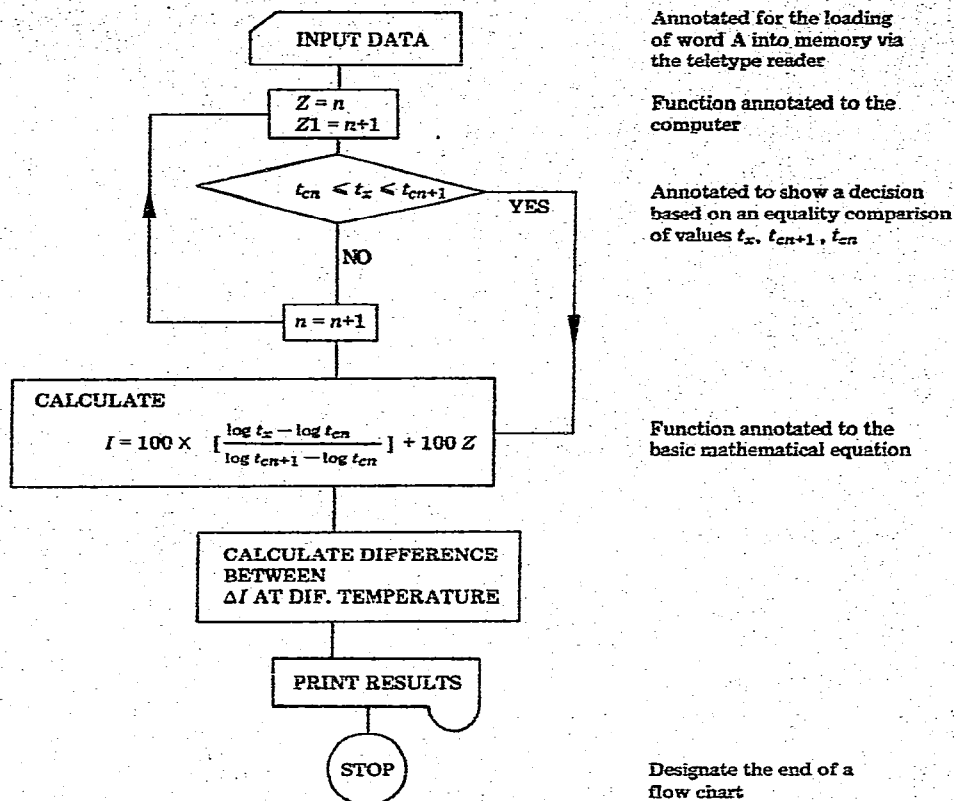


Fig. 11. Flow chart.

VARIABLE LIST

- A () contains the names
- C () contains the number of carbon atoms and the retention times of these hydrocarbons
- D () contains the values of the temperatures and the retention times for the compound under study
- I () contains the index numbers and differences
- K counter
- N counter
- N_1 number of carbons in a hydrocarbon retention times
- N_2 number of compounds under study retention times
- T number of temperatures
- Y_1 number of samples.

REFERENCES

- 1 F. I. Onuska and M. E. Comba, *J. Ass. Offic. Anal. Chem.*, 58 (1975) 6.
- 2 F. I. Onuska and M. E. Comba, *Biomed. Mass Spectrom.*, 2 (1975) 169.
- 3 F. I. Onuska and M. E. Comba, *Biomed. Mass Spectrom.*, 2 (1975) 176.
- 4 E. Kovats, *Helv. Chim. Acta*, 41 (1958) 1915.
- 5 L. S. Ettre, *Anal. Chem.*, 36 (1964) 31A.
- 6 D. Sissons and D. Welti, *J. Chromatogr.*, 60 (1971) 15.
- 7 J. Takács, C. Szita and G. Tarján, *J. Chromatogr.*, 56 (1971) 1.
- 8 J. Takács, Zs. Tálás, I. Bernáth, Gy. Czako and A. Fischer, *J. Chromatogr.*, 67 (1972) 203.
- 9 Z. Szentirmay, G. Tarján and J. Takács, *J. Chromatogr.*, 73 (1972) 11.
- 10 J. M. Takács, E. Kocsi, E. Garamvölgyi, E. Eckhart, T. Lombosi, Sz. Nyiredy, Jr., I. Borbély and Gy. Krasznai, *J. Chromatogr.*, 81 (1973) 1.
- 11 R. B. Harrison, D. C. Holmes, J. Roburn and J. O'G. Tatton, *J. Sci. Food Agr.*, 18 (1967) 10.
- 12 R. G. Nash and M. L. Beall, Jr., *J. Ass. Offic. Anal. Chem.*, 54 (1971) 959.