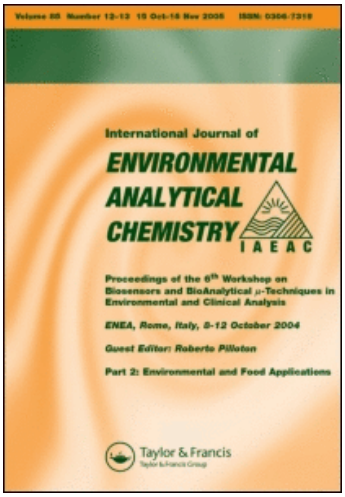

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Analysis of Toxaphene (PCC) and Chlordane in Biological Samples by NCI Mass Spectrometry†

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Several recent reports indicate that both toxaphene (polychlorocamphene, PCC) and chlordane are widespread pollutants in our environment. These insecticides, both complex mixtures of chlorinated hydrocarbons, are difficult to separate from each other. Thus the analyses of them in environmental samples are difficult and furthermore complicated by interferences from other chlorinated hydrocarbons such as PCB and DDT compounds. Mass fragmentography using the negative ions formed by chemical ionization proved to be useful to selectively detect the toxaphenes and the chlordanes without interferences from DDT. High concentrations of PCB still influence on the results and have to be removed in the clean-up procedure.

KEY WORDS: Toxaphene, chlordane, environment, analysis, mass spectrometry.

INTRODUCTION

Toxaphene (polychlorinated camphene, PCC) is an insecticide based on a complex mixture of chlorinated terpenes.[1–4] The commercial product Toxaphene (Boots Hercules Agrochemicals Co.) has been separated into more than 670 individual components[5] and the properties of these compounds vary over a wide range. The analysis of toxaphene in environmental samples by gas chromatography using electron capture detector (ECD) is complicated by interferences from other halogenated hydrocarbons such as the PCB, DDT and the chlordane families. We have investigated different clean-up methods for the separation of these

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compounds and chosen one, including sulphuric acid treatment and silica gel separation.[6,7] The toxaphene fraction from this treatment still contains some DDT compounds and the chlordanes and this makes the determination of toxaphene with ECD difficult.

In a previous investigation[6] we tried to use mass fragmentography for a more specific detection of toxaphene. The positive ions from electron impact (EI) and chemical ionization (CI) were studied. The sensitivity was in both cases low due to a pronounced fragmentation resulting in many different ions. The complex situation using positive ions from CI is also demonstrated by Cairus and coworkers.[8]

In order to detect toxaphene and chlordane without any cross-interferences we have used the negative ions formed by chemical ionization (NCI) in the mass spectrometer. This method has been used for selective detection of chlorinated pollutants in the environment by Dougherty and colleagues.[9] Dubay and coworkers have also applied NCI techniques for toxaphene detection,[10] and found very simple spectra representing almost only $(M-35)^-$ ions.

EXPERIMENTAL

Materials

1. Toxaphene (EPA, Code 6740).
2. Toxicant B (J. Casida, University of California, Berkeley, U.S.A.).
3. Chlordane (techn., Fluka).
4. α-Chlordane (EPA, Code 1220).
5. γ-Chlordane (EPA, Code 1240).
6. *trans*-Nonachlor (EPA, Code 5080).
7. *cis*-Nonachlor (D. Stalling, Fish and Wildlife Service, Columbia, U.S.A.).
8. Dechlorane® 603 (Hooker Chemical Corp.).
9. Aroclor 1260 (Monsanto Chemical Company).
10. Kieselgel 60 (0.040–0.063 mm, Merck).
11. Sulphuric acid (98%, BDH).
12. *n*-Hexane (redistilled until free from interferences).
13. Acetone (redistilled until free from interferences).
14. Diethyl ether (M & B).
15. Methane (99.999%, AGA, Stockholm, Sweden).

Gas chromatography

Instrument: Hewlett-Packard 5840.
Detector: ECD.

Column: OV-101, fused silica, 12 m, 0.21 mm i.d.
Carrier gas: 10% methane in argon, 0.3 ml/min.
Injector temperature: 250°C.
Detector temperature: 300°C.
Column temperatures: 50°C (5 min), 30°C/min to 190°C, 1°C/min to 220°C, 10°C/min to 260°C (20 min).
Injections: 3 μl splitless (glass wool in injector).

Mass fragmentography

Instrument: Finnigan 4021.
Column: OV-101, fused silica (direct into ion source).
12 m, 0.21 mm i.d.
Carrier gas: helium.
Injector temperature: 250°C.
Transfer line temperature: 250°C.
Column temperatures: 70°C (1 min), 20°C/min to 150°C, 5°C/min to 250°C (10 min).
Injection: 2 μl splitless.
Reagent gas: methane, 0.30 torr.
Ionization chamber temperature: 250°C.

In order to establish suitable ions for the mass fragmentographic detection complete spectra ($-500 \leqq m/z \leqq -160$) of the available reference compounds were scanned.

Investigation of interferences

Toxaphene and chlordane were determined in the following herring oil samples.

1. Unspiked.
2. Spiked with toxaphene (200 μg/g).
3. Spiked with α-chlordane (8 μg/g),
   γ-chlordane (8 μg/g),
   trans-nonachlor (8 μg/g),
   cis-nonachlor (2 μg/g).
4. Spiked with Arochlor 1260 (200 μg/g).

To study possible interferences, samples 1, 3, and 4 were analyzed for toxaphene and samples 1, 2 and 4 for the chlordanes.

Since PCB seem to influence the toxaphene detection, the preseparation can be critical especially for samples with high concentrations of PCB. From a Baltic seal sample, small fractions were collected from the silica

gel column and the latest eluting PCB isomeride was analyzed by gas chromatography-mass spectrometry.

Sample preparation

The extraction and clean-up was performed as described earlier.[6,7] In short this procedure includes extraction of tissues with a mixture of *n*-hexane and acetone followed by a mixture of *n*-hexane and diethyl ether, evaporation of the solvents and determination of the amount of the extracted lipid. One gram of the lipid was dissolved in *n*-hexane containing 200 ng of Dechlorane® 603 used as internal standard,[11] and this solution was treated with sulphuric acid for lipid removal.

Samples containing small amounts of PCB can be analyzed without further treatment, while samples with high PCB concentrations require further purification before analysis. This is accomplished by a silica gel column where a first fraction, eluted with *n*-hexane, contains the polychlorinated biphenyls, benzenes and naphthalenes. The second fraction is eluted with a mixture of diethyl ether and *n*-hexane (1:3) and contains most of the DDT compounds, the hexachlorocyclohexanes(HCH), the chlordane and toxaphene compounds.

RESULTS AND DISCUSSION

The gas chromatogram (ECD) in Figure 1 corresponds to the second silica gel fraction from herring oil. It is obvious that the chlordane, DDT and toxaphene compounds overlap each other and obstruct quantitative determinations.

In Figure 2 is shown a mass spectrum (NCI) of Toxicant B, a major component isolated from toxaphene.[12] The above mentioned loss of 35 mu is totally dominant and the ion formed can be used for a sensitive detection of this compound. To cover all chlorination degrees of the compounds present in the toxaphene standard at least six different mass numbers have to be monitored. To improve the selectivity, two ions in each cluster can be monitored, i.e. $m/z = 343$ and 345 for Toxicant B. If, in a sample, the ratio between these ions is not the same as in the reference spectrum, another compound is interfering.

In the same way suitable mass numbers for chlordane detection were found from scanned spectra of the available reference compounds (examples given in Figure 3) and the technical product. These spectra are more complex than PCC spectra but still four groups of "double" ions are enough for the quantitative determination. One of the groups (237 and 239) also proved to be ideal for the determination of the internal standard,

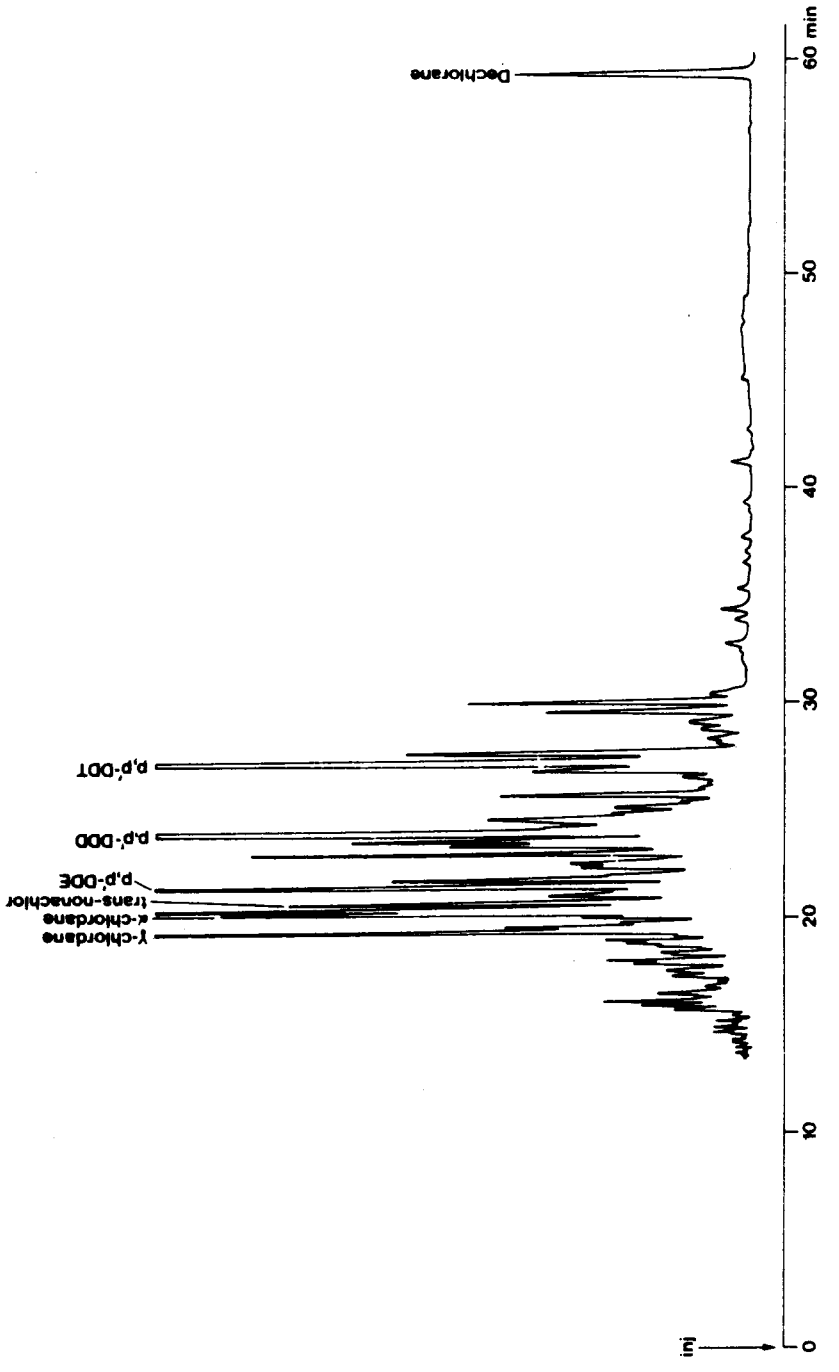

FIGURE 1 Gas chromatogram (ECD) of a Baltic herring sample after sulphuric acid treatment and preseparation on a silica gel column.

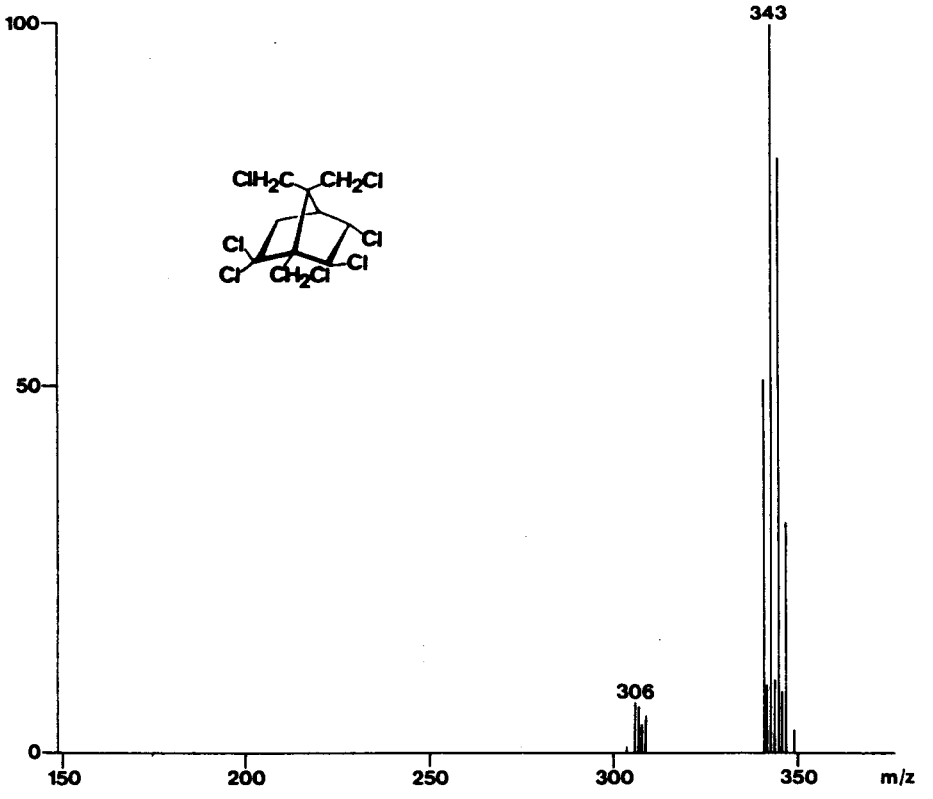

FIGURE 2 Mass spectrum (NCI) of "Toxicant B", a toxaphene component.[12]

Dechlorane® 603, used in the procedure. The long retention time of this compound (see Figure 1) excludes any risk for interferences with the chlordane components.

The ions monitored are given in Table I, which also includes $m/z = 360$ for PCB. The interferences from PCB on the detection of toxaphene and chlordane are demonstrated by the analyses of spiked samples (Table II). Both toxaphene and chlordane levels are elevated when PCB are added to the sample. The explanation to the strong influence on the toxaphene result can be observed in an NCI mass spectrum of a hexachlorobiphenyl compound in Figure 4. The $(M\text{-}19)^-$ ion gives signals at $m/z = 343$ and 345 used for toxaphene (see i.e., Figure 2). This fragmentation has been explained as the product of Cl loss from the oxidized molecular ion.[13]

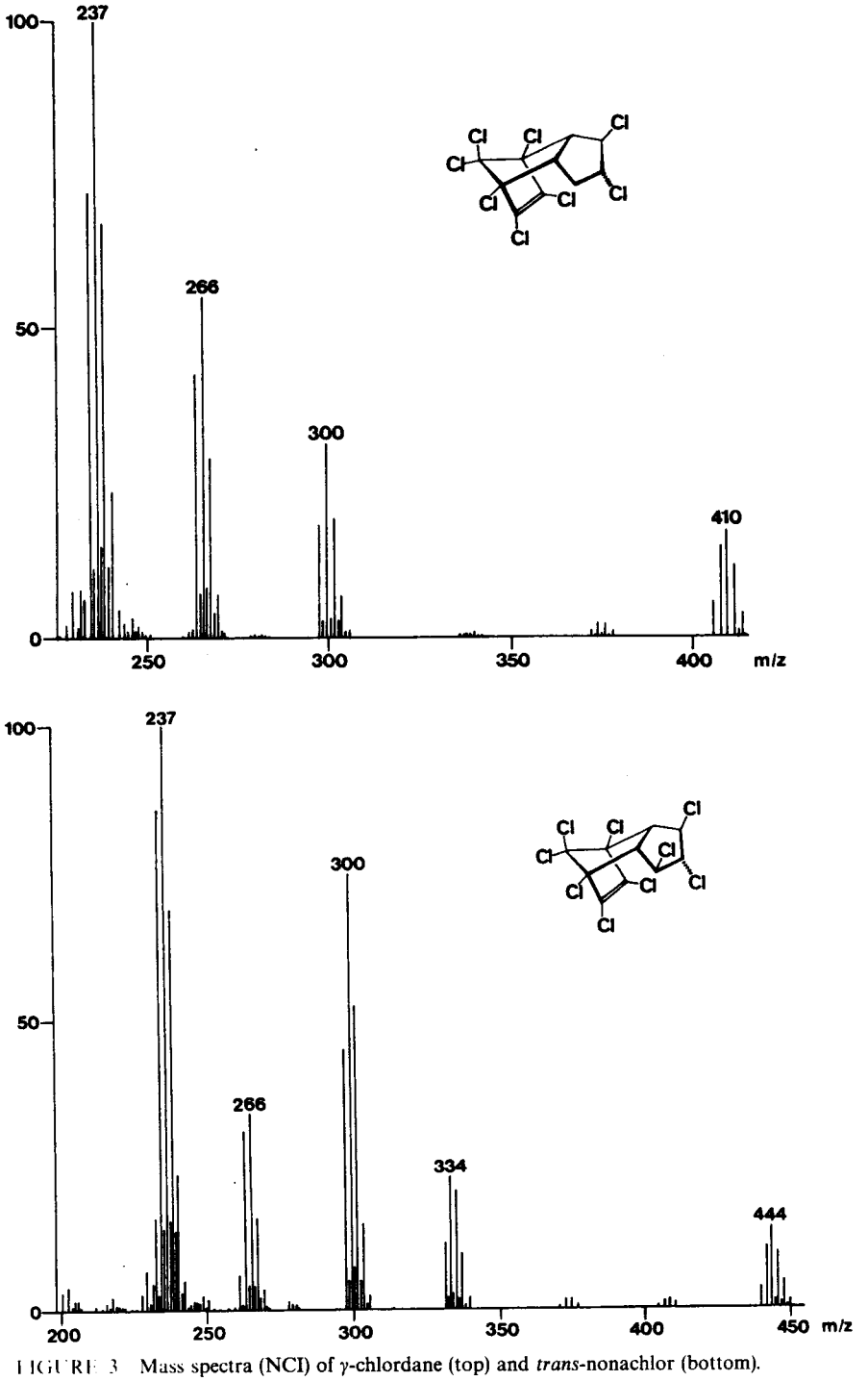

FIGURE 3 Mass spectra (NCI) of γ-chlordane (top) and *trans*-nonachlor (bottom).

TABLE I

Mass numbers used for simultaneous determination of toxaphene and chlordane by mass fragmentography (NCI).

m/z	Origin
237	Chlordane + internal standard
239	Chlordane + internal standard
264	Chlordane
266	Chlordane
300	Chlordane
302	Chlordane
309	Toxaphene
311	Toxaphene
334	Chlordane
336	Chlordane
343	Toxaphene
345	Toxaphene
360	PCB
379	Toxaphene
381	Toxaphene
413	Toxaphene
415	Toxaphene
447	Toxaphene
449	Toxaphene
481	Toxaphene
483	Toxaphene

TABLE II

Some interferences observed in quantitative analyses of toxaphene and chlordane in a herring sample.

Sample No.	Added compound	Amount μg/g lipid	Found (μg/g lipid)				
			toxaphene	α-chlordane	γ-chlordane	*trans*-nonachlor	*cis*-nonachlor
1	None	—	9.8	0.22	0.036	0.16	0.083
2	Toxaphene	200	n.a.	0.22	0.034	0.16	0.079
3	α-Chlordane	8	9.2	n.a.	n.a.	n.a.	n.a.
	γ-Chlordane	8					
	trans-nonachlor	8					
	cis-nonachlor	2					
4	Arochlor 1260	200	145	0.33	0.058	0.25	0.18

n.a. = not analysed.

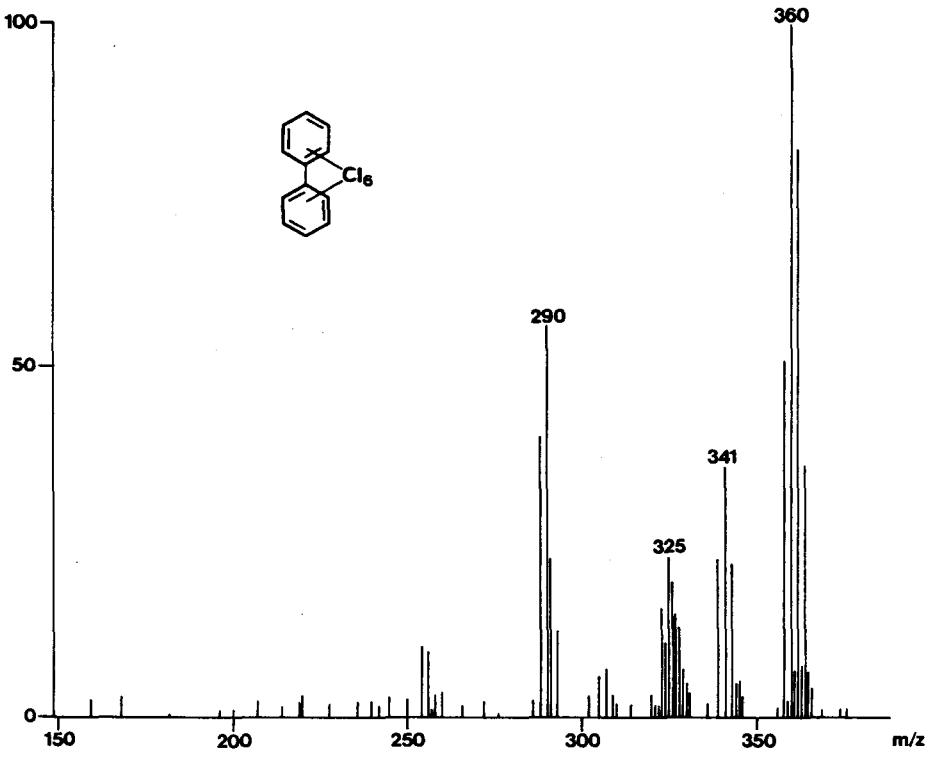

FIGURE 4 Mass spectrum (NCI) of a hexahclorobiphenyl. This compound, from a Baltic seal sample, was the last eluting PCB isomeride from the silica gel column used in the clean-up procedure.

The compound corresponding to the spectrum in Figure 4 was the last eluting PCB from the silica gel column and thus the $m/z=360$ can serve as an indicator for any PCB present in the second fraction. If a significant area appears in this mass chromatogram the preseparation has to be repeated.

There are also some influence from chlordane compounds at the $m/z=413$, 415, 447 and 449 used for toxaphene detection. This can, however, easily be overcome by selecting proper retention time windows for these signals during the area integration process.

Although present in high concentrations, none of the DDT compounds seems to influence the toxaphene or chlordane determinations.

Before the area determinations the results are normally controlled on the screen to avoid interferences, Figure 5 shows the chlordane mass

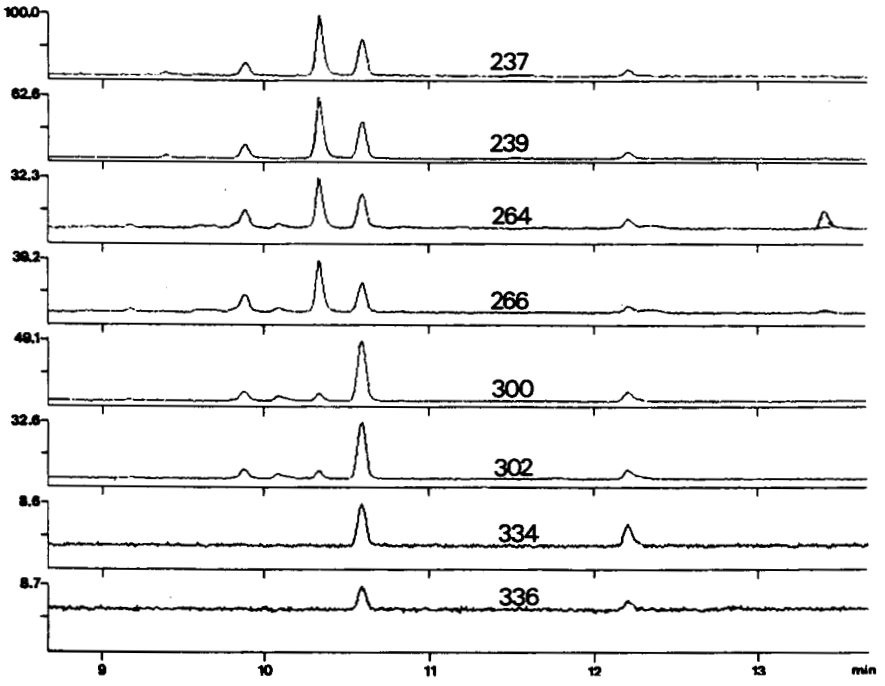

FIGURE 5 Individual mass chromatograms (NCI) for the mass numbers used for chlordane detection. From this figure it is rather easy to trace any compound with the wrong isotope ratio, such as the peak with a retention time of about 13.5 minutes.

chromatograms from an unspiked herring sample. At a retention time around 13.5 minutes a peak appears with wrong area ratio between $m/z = 264$ and 266. This peak must not be included in the total chlordane area.

The intensities from all mass numbers used for chlordane detection are added in one chromatogram (Figure 6). This can be used for detection of total chlordane with the technical product as reference. However, since the compounds we have used as references seem to constitute the major chlordane components in environmental samples, more reliable results are obtained if these are quantitated separately.

For the quantitative analysis of toxaphene we have to use the complex standard as reference material. This means that we will produce erroneous results if the composition of the chlorinated terpene mixture is different in the sample. We know that this is the case in most biological samples and therefore we can today only estimate the toxaphene concentrations.

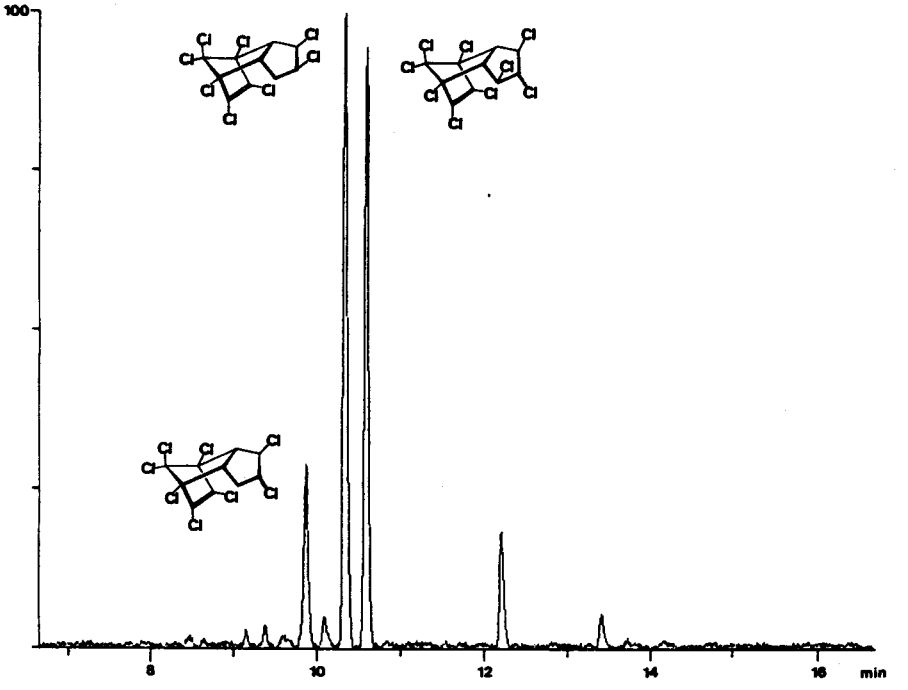

FIGURE 6 Mass fragmentogram based on the sum of intensities in the mass numbers used for chlordane detection. The sample is Baltic herring.

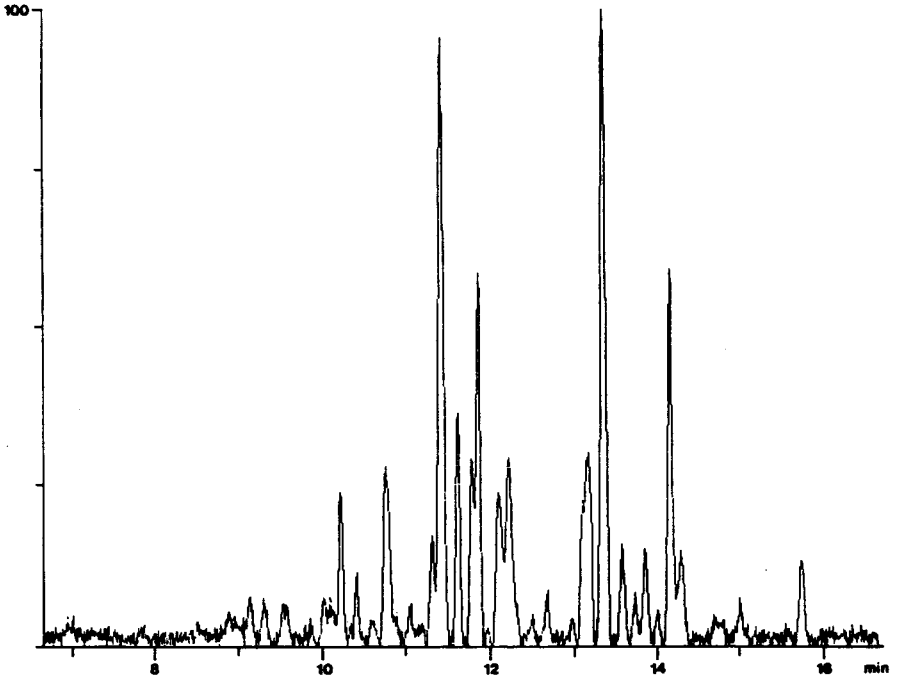

FIGURE 7 Mass fragmentogram based on the sum of intensities in the mass numbers used for toxaphene detection. The sample is Baltic herring.

These calculations can be made using the sum of toxaphene ions as shown in Figure 7. However, the mass fragmentographic method gives much more information as the chlorination degrees can be determined. Furthermore, the degree of unsaturations can be studied using the "double ion technique" described above. These factors are essential in studies of the degradation of toxaphene compounds. Such studies are in progress in our laboratory.

Acknowledgment

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References

1. S. A. Mashburn, Health and environmental effects of toxaphene. A literature compilation 1962–1978. National Technical Information Service (USA) ORNL/TIRC-78/5 (1978).
2. G. A. Pollock and W.W. Kilgore, *Residue Reviews* **69,** 87 (1978).
3. IARC, IARC Monographs on the Evaluation of the Carcinogenic Risk of Chemicals to Humans. Vol. 20. Some Halogenated Hydrocarbons. International Agency for the Research on Cancer. Lyon 1979, p. 327.
4. P. R. Durkin, P. M. Howard, J. Saxena, S. S. Lande, J. Santodonato, J. R. Strangl and D. H. Christopher, Reviews of the environmental effects of pollutants. X. Toxaphene. National Technical Information Service (USA). ORNL/EIS-130 (1980).
5. B. Jansson, Separation of toxaphene components, National Swedish Environment Protection Board, Internal Report, manuscript.
6. B. Jansson, R. Vaz, G. Blomkvist, S. Jensen and M. Olsson, *Chemosphere* **8,** 181 (1979).
7. U. Wideqvist, B. Jansson, L. Reutergård, G. Sundström and U.-B. Uvemo, The evaluation of a gas chromatographic method for the analysis of toxaphene, National Swedish Environment Protection Board, Internal Report NSL 82-01 (1982).
8. T. Cairus, E. G. Siegmund and J. E. Froberg, *Biomed. Mass Spectr.* **8,** 569 (1981).
9. R. C. Dougherty, *Anal. Chem.* **53,** 625A (1981).
10. M. A. Ribick, G. R. Dubay, J. D. Petty, D. L. Stalling and C. J. Schmitt, *Environ. Sci. Technol.* **16,** 310 (1982).
11. L. J. Miller and B. J. Puma, *J. Ass. Offic. Anal. Chemists* **62,** 1319 (1979).
12. J. E. Casida, R. L. Holmstead, S. Khalifa, J. R. Knox, T. Oshawa, K. J. Palmer and R. Y. Wong, *Science* **183,** 520 (1974).
13. R. C. Dougherty, J. Dalton and F. J. Biros, *Org. Mass Spectrom.* **6,** 1319 (1979).