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APPLICATION OF CAPILLARY GAS CHROMATOGRAPHY WITH MASS SELECTIVE DETECTION TO PESTICIDE RESIDUE ANALYSIS

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SUMMARY

A random sample of 76 pesticides with good gas chromatographic properties has been collected in order to test the applicability of the mass selective detector HP MSD for pesticide residue analysis. Using a capillary column, mass spectra of good quality were produced with 10 or 20 ng of each pesticide. Seventy two compounds were not only identified at this concentration with the PBM (probability based matching) search routine in the NBS library, but also remarkably with the best fit in the corresponding hit list. Lower concentrations can be detected only with selected ion monitoring (SIM). A mixture of 18 chlorinated pesticides was added to green pepper at the 10 ppb level. All compounds were detected with SIM using special time programming in one gas chromatographic analysis. The detection sensitivity of the mass selective detector in SIM mode approaches that of established selective detectors. The reliability of the results with respect to the identity of a pesticide residue, however, is orders of magnitude better. Therefore, this detector is a valuable tool for performing confirmatory analysis.

INTRODUCTION

Multiresidue analysis of pesticides in food and environmental samples must provide reliable identification and quantitation of a large number of compounds at very low concentrations. The analysis starts with an extraction step. Coextractants from the sample matrix are eliminated or reduced using a variety of clean-up procedures[1–5].

Gas chromatography (GC) with the selective electron-capture (ECD) and nitrogen–phosphorus (NPD) detection allows the detection of contaminants at trace levels in the lower ppb[a] range in the presence of a multitude of compounds extracted from the matrix to which these detectors do not respond. The number of compounds used in agriculture for plant protection has now surpassed 400. Additionally, the input of pollutants into the environment has increased and so it is impossible to separate all

[a] Throughout this article the American billion (10^9) is meant.

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these compounds in a single analysis, even with the application of high-performance capillary columns.

Reliability of multiresidue analysis can be achieved by different approaches. One technique that I have used for a decade is effluent splitting after the capillary column to two selective detectors. Each peak is characterized by its retention time and the response factors in the different detectors. Confirmation is accomplished by applying the same technique to a second capillary column coated with a separation phase of different polarity[6]. A second approach developed in our laboratory is two-dimensional capillary GC with pneumatic switching between two columns of differing polarities. Again, it is possible to increase the information content of the chromatographic data enormously by applying effluent splitting after the first or after both columns to several detectors[7]. A third approach is the application of a more extensive clean-up. One procedure widely used in Germany divides the sample into up to six fractions, applying chromatography on a small silica gel column[8]. Many of the pesticides are separated from overlapping matrix compounds by means of this method. At the same time, the classification of pesticides according to their partition coefficients in a system of water and organic solvents of increasing polarity is a valuable independent method.

With these methods, data processing on-line by means of a minicomputer may be used as a support in identifying suspected pesticide residues by checking the data against a data base of calibrated compounds[9].

The fourth and most sophisticated approach combines high-resolution GC with mass spectrometry (GC–MS). The mass spectrometer is without doubt the most specific detector available in multiresidue analysis. The specificity is based on the fact that molecules when bombarded with electrons of particular energy under vacuum conditions fragment following strict rules. The resulting fragmentation pattern reflects the individual molecular structure in a mass spectrum that is often considered as the fingerprint of the substance. These mass spectra show such specific characteristics that it is possible to differentiate many tens of thousands of compounds. It is of great importance that these mass spectra do not depend on the instrument used for measuring them but only on the ionization conditions applied. Standardized ionization conditions can easily be reproduced. Therefore, it is possible to compile all the mass spectra recorded all over the world in libraries. Such documented spectra are used to identify unknown compounds by comparing their spectra with those well established. The formerly laborious task of comparing mass spectra is now performed with the help of computers and sophisticated software programs. Within a few seconds such a computer program can perform a search of more than 40 000 documented mass spectra and draw up a list of a few mass spectra ranked according to their strongest resemblance to the one just recorded. In this way it is possible to identify an unknown peak in a gas chromatogram without having the corresponding test substance available.

Until recent years, the mass spectrometer appeared to be an instrument of great complexity that required a lot of maintenance and technical skill when used as an analytical tool. Nevertheless, from the beginning of reliable environmental studies it was recognized as the ultimate GC detector for confirming results obtained by means of the very sensitive selective detectors. Today, with mass selective detection (MSD), a small mass spectrometer is commercially available designed as a sensitive and most specific detector for high-resolution GC. MSD offers the full capacity of any

quadrupole mass spectrometer when coupled with a gas chromatograph. However, this detector is as easy to maintain and to handle as any other GC detector.

MSD can be used in two different modes that are very useful in pesticide multiresidue analysis: cyclic scanning or selected ion monitoring (SIM). In cyclic scanning the mass spectrometer acquires a series of mass spectra continuously over the whole time the GC separation process is underway. The summation of all the ions from the ionization process results in the total ion current (TIC) trace which looks like a signal recording from any GC detector. However, each point in the gas chromatogram represents a full mass spectrum stored in a data file on the disc which can be inspected at any time on the screen. These mass spectra can be manipulated by the computer software to obtain background subtraction or averaged spectra. Mass spectra corrected in this way can now be used for library search in order to identify the substance.

In SIM the sensitivity is very much increased at the expense of information. Instead of scanning the entire spectrum over the whole mass range, only a few ions are recorded which are indicative for the compound to be searched for. The gain in sensitivity is the result of longer specific sampling times for each of the ions selected. The highest detection sensitivity achievable with a mass spectrometric detector is single ion monitoring where the instrument is adjusted to collect only ions of one defined mass.

The application of SIM requires the knowledge of the mass spectrum of the compound to be analyzed and its retention time on the capillary column used. The combined data from SIM and the exact retention time provide the optimum in detection sensitivity and reliability with respect to the identity of the analyzed pesticide residue that can be achieved today.

In this paper a study of the analysis of 76 pesticides representing a variety of chemical structures by means of GC with MSD is reported.

EXPERIMENTAL

Instrumentation

Gas chromatograph HP 5890 with a mass selective detector MSD HP 5970 and MS ChemStation HP 59970 including an HP 59973 NBS mass spectral library (NBS REVE) and a split/splitless injector for capillary columns was employed.

Gas chromatography

A fused-silica column, 25 m × 0.20 mm HP-1 (cross-linked methyl silicone gum) with film thickness 0.33 μm was used with helium as the carrier gas. Temperature settings (°C): injection port, 250; transfer line, 260. Temperature programme (°C): 1 min at 100, 30°/min to 150, held for 2 min, 3°/min to 205, 10°/min to 240, 2°/min to 260, held for 10 min. A 1-μl volume of sample was injected manually applying the hot splitless injection technique with the split closed for 1 min.

Mass spectrometric acquisition parameters

Temperature settings (°C); transfer line, 180; ion source, 175; mass analyzer, 180. Scan parameters: scanned mass range, 50–500 daltons; scan rate, 1.22 scans/s; threshold, 500. Solvent delay: 2.5 min. The voltages of the repeller, draw out, ion focus,

TABLE I

COMPILATION OF PESTICIDES USED

Retention times (t_R in min) recorded under the experimental conditions. A = Acaricide; F = fungicide; H = herbicide; I = insecticide; N = nematicide; I* = insecticide used as the internal standard. HCH = BHC = isomers of 1,2,3,4,5,6-hexachlorocyclohexane. HCB = hexachlorobenzene.

No.	*Common name*	*CAS No.*	*Mol.wt.*	*Formula*	*Use*	t_R
1	Aldrin	309-00-2	364.92	$C_{12}H_8Cl_6$	I*	23.82
2	Amidithion	919-76-6	273.31	$C_7H_{16}NO_4PS_2$	I	23.62
3	Azinphos-ethyl	2642-71-9	345.38	$C_{12}H_{16}N_3O_3PS_2$	I	38.36
4	Bromophos	2104-96-3	366.00	$C_8H_8BrCl_2O_3PS$	I	25.11
5	Bromophos-ethyl	4824-78-6	394.05	$C_{10}H_{12}BrCl_2O_3PS$	I	26.87
6	Bromopropylate	18181-80-1	428.12	$C_{17}H_{16}Br_2O_3$	I	34.19
7	Carbophenothion	786-19-6	342.87	$C_{11}H_{16}ClO_2PS_3$	I	31.14
8	Chlorfenprop-methyl	14437-17-3	233.10	$C_{10}H_{10}Cl_2O_2$	I	12.31
9	Chlorfenvinphos	470-90-6	359.58	$C_{12}H_{14}Cl_3O_4P$	I	26.15
10	Chlormephos	24934-91-6	234.70	$C_5H_{12}ClO_2PS_2$	I	8.58
11	Chloroneb	2675-77-6	207.06	$C_8H_8Cl_2O_2$	F	10.17
12	Chloropropylate	5836-10-2	339.22	$C_{17}H_{16}Cl_2O_3$	A	29.53
13	Chlorothalonil	1897-45-6	265.91	$C_8Cl_4N_2$	F	19.16
14	Chlorthal-dimethyl	1861-32-1	331.97	$C_{10}H_6Cl_4O_4$	H	24.53
15	Chlorthion	500-28-7	297.66	$C_8H_9ClNO_5PS$	I*	24.79
16	Coumaphos	56-72-4	362.77	$C_{14}H_{16}ClO_5PS$	F	41.07
34	DDD-o,p'	53-19-0	320.05	$C_{14}H_{10}Cl_4$	I	28.53
18	DDD-p,p'	72-54-8	320.05	$C_{14}H_{10}Cl_4$	I	29.96
19	DDE-o,p'	3424-82-6	318.03	$C_{14}H_8Cl_4$	I	26.92
20	DDE-p,p'	72-55-9	318.03	$C_{14}H_8Cl_4$	I	28.17
21	DDT-o,p'	789-02-6	354.49	$C_{14}H_9Cl_5$	I	30.11
22	DDT-p,p'	50-29-3	354.49	$C_{14}H_9Cl_5$	I	31.67
23	Dialifos	10311-84-9	393.85	$C_{14}H_{17}ClNO_4PS_2$	I	38.77
24	Diazinon	333-41-5	304.35	$C_{12}H_{21}N_2O_3PS$	I	18.55
25	Dichlobenil	1194-65-6	172.01	$C_7H_3Cl_2N$	H	6.93
26	Dichlofenthion	97-17-6	315.16	$C_{10}H_{13}Cl_2O_3PS$	N	20.79
27	Dichlofluanid	1085-98-9	333.23	$C_9H_{11}Cl_2FN_2O_2S_2$	F	23.54
28	Dichloran	99-30-9	207.02	$C_6H_4Cl_2N_2O_2$	F	16.17
29	Dieldrin	60-57-1	380.91	$C_{12}H_8Cl_6O$	I	28.24
30	Dimethoate	60-51-5	229.26	$C_5H_{12}NO_3PS_2$	I	16.31
31	Disulfoton	298-04-4	274.41	$C_8H_{19}O_2PS_3$	IA	18.75
32	Ditalimfos	5131-24-8	299.29	$C_{12}H_{14}NO_4PS$	F	27.42
33	Endosulfan I (alpha)	115-29-7	406.93	$C_9H_6Cl_6O_3S$	I	27.18
	Endosulfan II (beta)	115-29-7	406.93	$C_9H_6Cl_6O_3S$	I	29.74
34	Ethion	563-12-2	384.48	$C_9H_{22}O_4P_2S_4$	I	30.18
35	Ethoprophos	13194-48-4	242.34	$C_8H_{19}O_2PS_2$	IN	13.48
36	Fenchlorphos	299-84-3	321.55	$C_8H_8Cl_3O_3PS$	I	22.25
37	Fenitrothion	122-14-5	277.24	$C_9H_{12}NO_5PS$	I	23.10
38	Fensulfothion	115-90-2	308.36	$C_{11}H_{17}O_4PS_2$	IN	29.74
39	Fenthion	55-38-9	278.33	$C_{10}H_{15}O_3PS_2$	I	24.17
40	Folpet	133-07-3	296.62	$C_9H_4C_{13}NO_2S_2$	F	26.34
41	Fonofos	944-22-9	246.33	$C_{10}H_{15}OPS_2$	I	17.94
42	Formothion	2540-82-1	257.27	$C_6H_{12}NO_4PS_2$	I	20.04
43	HCB	118-74-1	284.78	C_6Cl_6	F	15.98
44	HCH-alpha	319-84-6	290.83	$C_6H_6Cl_6$	I	15.54
45	HCH-beta	319-85-7	290.83	$C_6H_6Cl_6$	I	17.06
46	HCH-delta	319-86-8	290.83	$C_6H_6Cl_6$	I	18.85
47	Heptachlor	76-44-8	373.32	$C_{10}H_5Cl_7$	I	21.76

TABLE I *(continued)*

No.	*Common name*	*CAS No.*	*Mol.wt.*	*Formula*	*Use*	t_R
48	Heptachlorepoxide-c	1024-57-3	389.32	$C_{10}H_5Cl_7O$	I	25.76
49	Heptachlorepoxide-t	1024-57-3	389.32	$C_{10}H_5Cl_7O$	I	25.94
50	Heptenophos	34783-40-9	250.62	$C_9H_{12}ClO_4P$	I	11.94
51	Lindane	58-89-9	290.83	$C_6H_6Cl_6$	I	17.45
52	Methoxychlor	72-43-5	345.66	$C_{16}H_{15}Cl_3O_2$	I	34.58
53	Mevinphos	7786-34-7	224.15	$C_7H_{13}O_6P$	I	8.28
54	Mirex	2385-85-5	545.55	$C_{10}Cl_{12}$	I*	36.90
55	Parathion	56-38-2	291.26	$C_{10}H_{14}NO_5PS$	IA	24.33
56	Parathion-methyl	298-00-0	263.21	$C_8H_{10}NO_5PS$	IA	21.36
57	Phenkapton	2275-14-1	377.31	$C_{11}H_{15}Cl_2O_2PS_3$	A	34.68
58	Phosalone	2310-17-0	367.81	$C_{12}H_{15}ClNO_4PS_2$	IA	36.23
59	Phosmet	732-11-6	317.32	$C_{11}H_{12}NO_4PS_2$	I	34.09
60	Phosphamidon	13171-21-6	299.69	$C_{10}H_{19}ClNO_5P$	IA	20.87
61	Pirimiphos-ethyl	23505-41-1	333.39	$C_{13}H_{24}N_3O_3PS$	I	25.37
62	Pirimiphos-methyl	29232-93-7	305.34	$C_{11}H_{20}N_3O_3PS$	I	23.26
63	Propachlor	1918-16-7	211.69	$C_{11}H_{14}ClNO$	H	12.92
64	Propyzamide	23950-58-5	256.13	$C_{12}H_{11}Cl_2NO$	H	17.94
65	Prothiofos	34643-46-4	345.25	$C_{11}H_{15}Cl_2O_2PS_2$	I	27.91
66	Pyrazophos	13457-18-6	373.37	$C_{14}H_{20}N_3O_5PS$	FAI	38.26
67	Quintozene	82-68-8	295.34	$C_6Cl_5NO_2$	F	17.75
68	Sulfotep	3689-24-5	322.32	$C_8H_{20}O_5P_2S_2$	I	15.01
69	Tecnazene	117-18-0	260.89	$C_6HCl_4NO_2$	F	12.79
70	Tetrachlorvinphos	22248-79-9	365.97	$C_{10}H_9Cl_4O_4P$	I	27.17
71	Tetradifon	116-29-0	356.06	$C_{12}H_6Cl_4O_2S$	A	35.68
72	Tolylfluanid	731-27-1	347.26	$C_{10}H_{13}Cl_2FN_2O_2S_2$	F	25.98
73	Triadimefon	43121-43-3	293.76	$C_{14}H_{16}ClN_3O_2$	F	24.46
74	Triazophos	24017-47-8	313.32	$C_{12}H_{16}N_3O_3PS$	IA	30.75
75	Trifluralin	1582-09-8	335.29	$C_{13}H_{16}F_3N_3O_4$	H	14.69
76	Vinclozolin	50471-44-8	286.12	$C_{12}H_9Cl_2NO_3$	F	21.37

entrance lens and X-ray and the parameters for the quadrupole mass filter were set according to the values proposed by the program AUTOTUNE, which automatically optimizes these parameters using perfluorotributylamine (PFTBA) as a calibration standard.

Materials

Pesticides were obtained as test substances from Dr. Ehrenstorfer, Augsburg, F.R.G. and Promochem, Wesel, F.R.G., with 97–99% purity. All substances were dissolved in toluene (Promochem, Nanograde).

Clean up

Food samples were analyzed using method S 19 in ref. 1. Plant material with high water content was extracted with acetone. To plant material with low water content water was added to adjust the acetone: water ratio during extraction to 2:1 (v/v). The extract was saturated with sodium chloride and diluted in dichloromethane in order to separate excess of water. The evaporation residue of the organic phase or a fat solution was cleaned up by gel permeation chromatography on Bio-Beads SX-3 polystyrene

gel, using a mixture of cyclohexane and ethyl acetate as the eluent and an automated gel permeation chromatograph. The residue-containing fraction was concentrated and fractionated on a small silica gel column into six fractions. The eluents were mixtures of toluene with hexane or increasing amounts of acetone. The fractions were evaporated, brought to volume with toluene and analyzed by capillary GC with MSD. In our laboratory, fractions 1 and 2 are collected together as one fraction containing all chlorinated pesticides.

RESULTS

In Table I the 76 pesticides included in this study are compiled. Only compounds that are known to be analyzed easily by GC are listed. Most of them show good responses to the very sensitive ECD, NPD and flame photometric detection (FPD). These pesticides represent a variety in chemical structure as well as the different applications in agricultural production.

In Fig. 1 the TIC of a mixture of chlorinated hydrocarbon insecticides is shown. The mixture contains pesticides that have been extensively used all over the world from the early days of introduction of these chemicals for plant protection around 1950. They are now banned in most western industrialized countries but, however, are still in use in many other countries. Because of their great chemical stability they persist for decades in the environment. These substances can be analyzed at trace level by means of GC using ECD but this detection method is prone to many interferences and consequently the results are not always reliable. Fig. 1 represents the separation of 10 ng of each pesticide. By means of background subtraction, mass spectra of good quality were obtained that were added to a special sub-library for pesticides. All these spectra are in good agreement with those already stored in the NBS library (NBS REVE with HP 59973B Library software) or measured in our laboratory as well as others using GC–MS instruments of both the magnetic field and the quadrupole filter type[10–16].

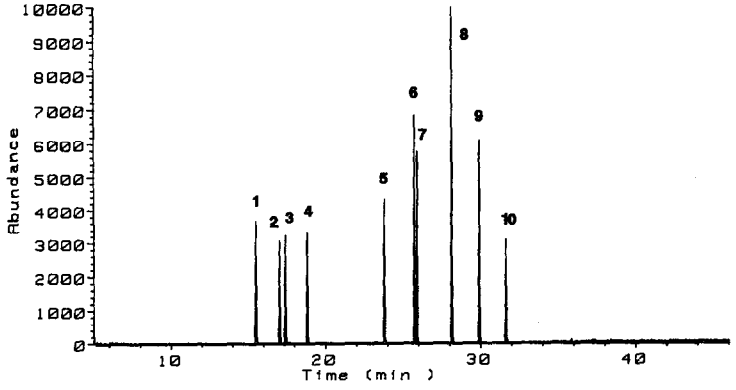

Fig. 1. TIC of chlorinated pesticides. A 10-ng amount of each of the following compounds was injected: 1 = α-HCH; 2 = β-HCH; 3 = lindane; 4 = δ-HCH; 5 = aldrin; 6 = *cis*-heptachlorepoxide; 7 = *trans*-heptachlorepoxide; 8 = p,p'-DDE; 9 = p,p'-DDD; 10 = p,p'-DDT.

In Fig. 2 the TIC of a mixture of organophosphorus pesticides is shown demonstrating the very good GC separation of all 29 compounds. Each of these peaks represents 20 ng of substance. The TIC trace gives an idea of the different yields in ionization intensities depending on the chemical structure. However, at this level of concentration, from each organophosphorus pesticide representative mass spectra were produced. In the same way, mixtures containing all the other pesticides were analyzed with the described GC–MSD method at concentration levels of 10 or 20 ng of each substance. Mass spectra of all compounds were directly (or after background subtraction) applied to a library search in the NBS library. Seventy two of the pesticides were identified correctly by means of the PBM (propability based matching) searching routine developed by McLafferty *et al.*[17–19]. The remaining four pesticides were not found because their spectra were not included in the NBS library. A compilation of the results of the library search is given in Table II. It is in fact most remarkable that all 72 compounds were not only found at concentrations of 10 or 20 ng per peak but also with the best fit. As is seen from the last row in Table II, several compounds were not indicated with their common names but with one applied by other organizations or with the chemical name according to IUPAC.

In Fig. 3 the mass spectra of the four pesticides not found in the NBS library are reported. This gives the opportunity to demonstrate the quality of spectra obtained with 10 or 20 ng of substance under the described experimental conditions that are exactly the same as applied to pesticide residue analysis.

After having entered all the mass spectra into the pesticide sub-library, preliminary measurements were performed with diluted solutions of the described pesticide mixtures. Applying the cyclic scan mode, a mixture of chlorinated pesticides containing 2 ng of each compound was analyzed as shown in Fig. 4. All 20 substances were correctly identified by means of a library search. The signal-to-noise ratio of the TIC trace indicates that a concentration of 2 ng per peak approaches the limit of detection of many pesticides with cyclic scanning.

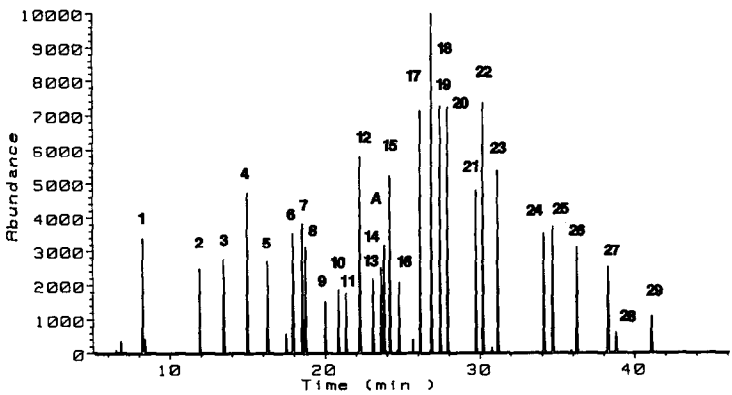

Fig. 2. TIC of organophosphate pesticides. A 20-ng amount of each of the following compounds was injected: 1 = mevinphos; 2 = heptenophos; 3 = ethoprophos; 4 = sulfotep; 5 = dimethoate; 6 = fonofos; 7 = diazinon; 8 = disulfoton; 9 = formothion; 10 = phosphamidon: 11 = parathion-methyl; 12 = fenchlorphos; 13 = fenitrothion; 14 = amidithion; 15 = fenthion; 16 = chlorthion; 17 = chlorfenvinphos; 18 = bromophos-ethyl; 19 = ditalimfos; 20 = prothiofos; 21 = fensulfothion; 22 = ethion; 23 = carbophenothion; 24 = phosmet; 25 = phenkapton; 26 = phosalone; 27 = pyrazophos; 28 = dialifos; 29 = coumaphos; A = aldrin (internal standard).

TABLE II

RESULT OF LIBRARY SEARCH WITH 76 PESTICIDES

No.	*Common name*	*CAS No.*	*Mol.wt.*	*NBS No.*	*Best fit*
1	Aldrin	309-00-2	361.875	34152	
2	Amidithion	919-76-6	273.025	26449	
3	Azinphos-ethyl	2642-71-9	345.036	33020	
4	Bromophos	2104-96-3	363.848	34387	
5	Bromophos-ethyl	4824-78-6	391.880	35899	
6	Bromopropylate	18181-80-1	425.946	37527	
7	Carbophenothion	786-19-6	341.973	32781	
8	Chlorfenprop-methyl	14437-17-3	232.005	21311	
9	Chlorfenvinphos	470-90-6	357.969	33865	
10	Chlormephos	24934-91-6	233.970	21673	
11	Chloroneb	2675-77-6	205.989	17624	Benzene, ...[b]
12	Chloropropylate	5836-10-2	338.047	32520	
13	Chlorothalonil	1897-45-6	263.881	25431	Tetrachloro[c]
14	Chlorthal-dimethyl	1861-32-1	329.901	31821	DCPA[a]
15	Chlorthion	500-28-7	296.962	28934	
16	Coumaphos	56-72-4	362.014	34155	
17	DDD-o,p'	53-19-0	317.953	30874	
18	DDD-p,p'	72-54-8	317.953	30875	1,1-Dichloro[d]
19	DDE-o,p'	3424-82-6	315.937	30697	
20	DDE-p,p'	72-55-9	315.937	30696	
21	DDT-o,p'	789-02-6	351.914	33482	
22	DDT-p,p'	50-29-3	351.914	33481	
23	Dialifos	10311-84-9	393.002	35976	Dialifor[a]
24	Diazinon	333-41-5	304.100	29598	Dimpylate[a]
25	Dichlobenil	1194-65-6	170.964	11664	Benzonitrile[e]
26	Dichlofenthion	97-17-6	313.969	30462	
27	Dichlofluanid	1085-98-9	331.961	32138	
28	Dichloran	99-30-9	205.964	17600	2,6-Dichloro[f]
29	Dieldrin	60-57-1	377.870	35144	
30	Dimethoate	60-51-5	228.999	21040	
31	Disulfoton	298-04-4	274.028	26595	
32	Ditalimfos	5131-24-8	299.037	29101	O,O-Diethyl[g]
33	Endosulfan	115-29-7	403.816	36627	
34	Ethion	563-12-2	383.987	35518	
35	Ethoprophos	13194-48-4	242.056	22777	
36	Fenchlorphos	299-84-3	319.899	31163	Ronnel[a]
37	Fenitrothion	122-14-4	277.017	26839	
38	Fensulfothion	115-90-2	308.030	29926	
39	Fenthion	55-38-9	278.019	26843	
40	Folpet	133-07-3	294.902	28695	
41	Fonofos	944-22-9	246.029	23060	
42	Formothion	2540-82-1	256.994	24582	
43	HCB	118-74-1	281.812	27405	
44	HCH-alpha	319-84-6	287.859	28091	alpha-Lindane
45	HCH-beta	319-85-7	287.859	28092	Cyclohexane[h]
46	HCH-delta	319-86-8	287.859	28093	delta-Lindane
47	Heptachlor	76-44-8	369.820	34677	
48	Heptachlorepoxide-c	1024-57-3	385.815	35535	
49	Heptachlorepoxide-t	1024-57-3	385.815	35535	
50	Heptenophos	34783-40-9			No match
51	Lindane	58-89-9	287.859	28090	

TABLE II (*continued*)

No.	*Common name*	*CAS No.*	*Mol.wt.*	*NBS No.*	*Best fit*
52	Methoxychlor	72-43-5	344.013	32941	
53	Mevinphos	7786-34-7	224.044	20347	
54	Mirex	2385-85-5	539.625	40520	
55	Parathion	56-38-2	291.032	28324	
56	Parathion-methyl	298-00-0	263.001	25275	
57	Phenkapton	2275-14-1	375.934	35055	
58	Phosalone	2310-17-0	366.986	34511	
59	Phosmet	732-11-6	316.994	30824	
60	Phosphamidon	13171-21-6	299.068	29096	
61	Pirimiphos-ethyl	23505-41-1	333.127	32142	
62	Pirimiphos-methyl	29232-93-7	305.095	29718	
63	Propachlor	1918-16-7	211.075	18300	Acetamide[i]
64	Propyzamide	23950-58-5	255.021	24288	
65	Prothiofos	34643-46-4			No match
66	Pyrazophos	13457-18-6	373.085	24935	
67	Quintozene	82-68-8	292.836	28520	Benzene ...[j]
68	Sulfotep	3689-24-5	322.022	31305	
69	Tecnazene	117-18-0	258.875	24834	Benzene ...[k]
70	Tetrachlorvinphos	22248-79-9	363.898	34285	
71	Tetradifon	116-29-0	353.883	33616	
72	Tolylfluanid	731-27-1			No match
73	Triadimefon	43121-43-3	293.092	28489	Butanone, ...[l]
74	Triazophos	24017-47-8	313.064	30419	
75	Trifluralin	1582-09-8	335.108	32328	
76	Vinclozolin	50471-44-8			No match

[a] Common name approved by organizations other than ISO or BSI.
[b] Benzene, 1,4-dichloro-2,5-methoxy.
[c] Tetrachloroisophthalonitrile.
[d] 1,1-Dichloro-2,2-bis(*p*-chlorophenyl)ethane.
[e] Benzonitrile, 2,6-dichloro.
[f] 2,6-Dichloro-4-nitroaniline.
[g] O,O-Diethylphthalimidophosphonothioate.
[h] Cyclohexane, 1,2,3,4,5,6-hexachloro.
[i] Acetamide, 2-chloro-N-(1-methylethyl)-N-phenyl.
[j] Benzene, pentachloronitro.
[k] Benzene, 1,2,4,5-tetrachloro-3-nitro.
[l] 2-Butanone, 1-(4-chlorophenoxy)-3,3-dimethyl-1-(1H-1,2,4-triazol-1-yl).

A survey of all compounds at this concentration level resulted in a correct recognition of all chlorinated pesticides, but pesticides of other structural classes were less reliably identified. Most of them would certainly not have been recognized in the presence of biological matrix compounds. Frequently, however, the presence of the individual pesticide was detected at the expected retention time in the chromatogram by extracting the indicative ions from the cyclic scan data.

The application of SIM enhances the detection sensitivity by orders of magnitude and at the same time the overlapping signals from the matrix compounds are considerably reduced. In Fig. 5 the SIM records of an analytical fraction of green pepper spiked with a mixture of chlorinated pesticides at the low concentration of 10

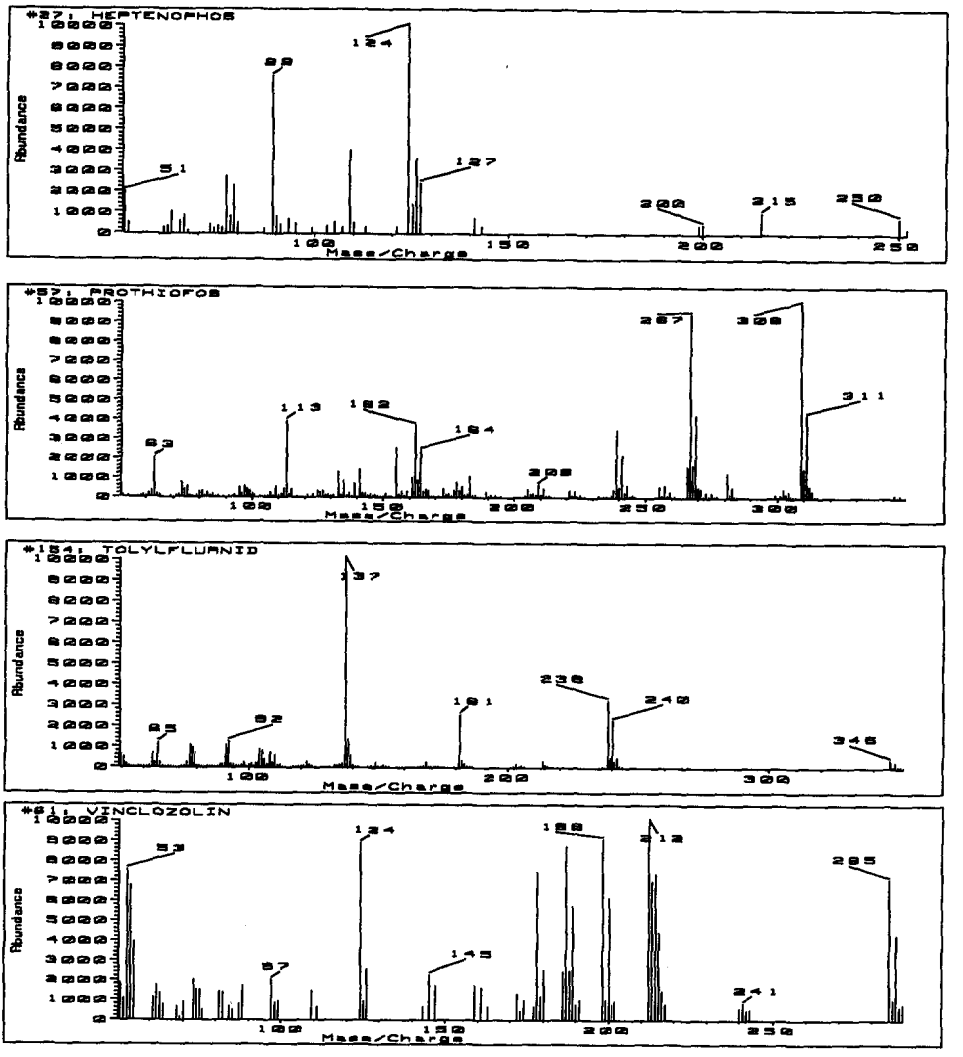

Fig. 3. Mass spectra of four pesticides not found in the NBS library. A 20-ng amount of each of heptenophos, prothiofos, tolylfluanid and vinclozolin was analyzed with GC–MS. Mass spectra after background subtraction.

ppb are presented. The injected extract volume of 1 μl contained about 50 pg of each pesticide together with the coextracted matrix compounds equivalent to 5 mg of vegetable. Highest sensitivity was achieved by selecting only a few masses in one time interval. This rule, however, was not applicable to all the time intervals. Seven masses had to be recorded in the first time interval between 15 and 20 min in order to screen simultaneously for HCB, quintozene and the HCH isomers. The SIM traces presented in Fig. 5 demonstrate that all the added chlorinated pesticides were detected in the spiked pepper sample at trace levels. At the same time the confirmation of 50 ppb endosulfan, first detected with GC–ECD in the green pepper sample, is presented.

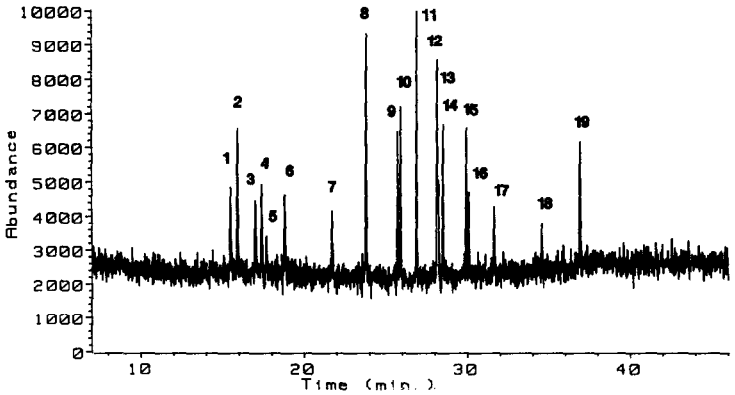

Fig. 4. Total ion chromatogram of a diluted mixture of chlorinated pesticides. A 2-ng amount of each of the following compounds was injected: 1 = α-HCH; 2 = HCB; 3 = β-HCH; 4 = lindane; 5 = quintozene; 6 = δ-HCH; 7 = heptachlor; 8 = aldrin; 9 = *cis*-heptachlorepoxide; 10 = *trans*-heptachlorepoxide; 11 = o,p'-DDE; 12 = p,p'-DDE; 13 = dieldrin; 14 = o,p'-DDD; 15 = p,p'-DDD; 16 = o,p'-DDT; 17 = p,p'-DDT; 18 = methoxychlor; 19 = mirex.

Although with GC–ECD two peaks representing endosulfan-I and endosulfan-II were detected, only endosulfan-I can be monitored with SIM at this low concentration, owing to the extensive fragmentation of endosulfan-II yielding only ions of low intensity.

DISCUSSION

The study confirmed that the combination of capillary GC with MSD can be applied to pesticide residue analysis in food samples. The MSD detector is really a mass selective GC detector that does not need the skill of maintainance that is necessary to operate a mass spectrometer. In our laboratory, MSD was much easier to handle than NPD, for instance. One of the spectacular features of GC–MS is the cyclic scan mode. After having performed a GC analysis a computer program can identify the peaks and automatically carry out a library search. The result of such a GC analysis is a list of compounds identified in an unknown sample.

The reliable recognition of 72 pesticides in the various test mixtures at amounts of 10 or 20 ng (Table II) was an impressive demonstration of the effectiveness of the PBM search routine developed by McLafferty *et al.*[17–19]. The library search was executed by an operator who did not know the composition of the pesticide mixtures. In all library searches for the 72 gas chromatographic peaks the pesticide was set into the first place (best fit) of a list of 10 compounds ranked for similarity of the sample spectrum to the over 42 000 spectra in the NBS library. This tempts one to believe that the fully automated qualitative analysis has been established at a very low concentration.

In pesticide residue analysis in food samples, however, this beautiful perspective is seldom reality. In Fig. 4 the analysis of a mixture of 2 ng of each pesticide is shown. These chlorinated pesticides were identified by an experienced analyst by means of the mass spectra after background subtraction and the retention time. Background

subtraction, however, did not yield mass spectra free of additional masses which decreases considerably the reliability of an automated library search.

When following established clean-up procedures[1,8], an amount of 2 ng of a pesticide means a concentration of 400 ppb in the food sample. Although all chlorinated pesticides were recognized at this concentration in test mixtures without interfering matrix compounds, about half of the pesticides under study did not produce mass spectra of sufficient quality. The problem mostly cannot be overcome by concentrating the sample extracts because the coextractants are enriched in the same proportion. On the other hand, for many pesticides, maximum tolerance limits are established at 10 ppb. At this concentration the identification of a pesticide by means of a full mass spectrum is expected only in a few very favourable situations with respect to the chemical structure of the compound and the coextractants from the food matrix. Therefore, pesticide residue analysis at this relevant low concentration has to rely on SIM. A good example to demonstrate the potential of this method is given in Fig. 5

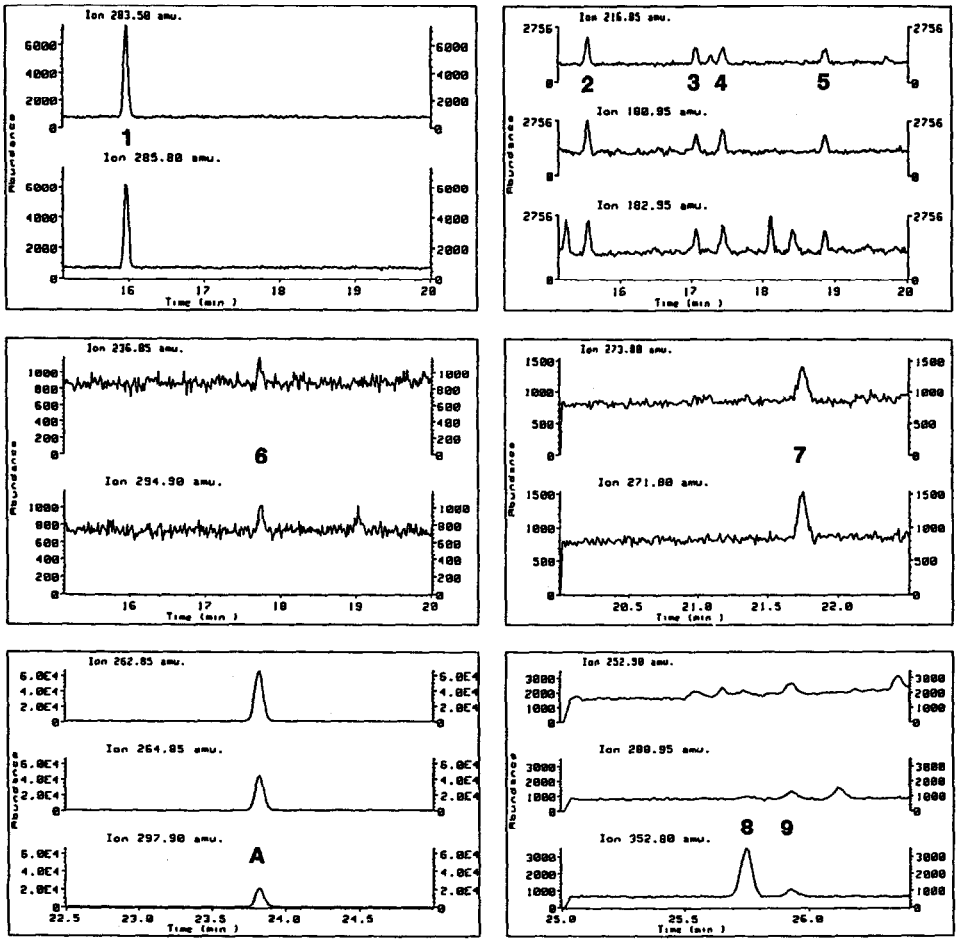

Fig. 5.

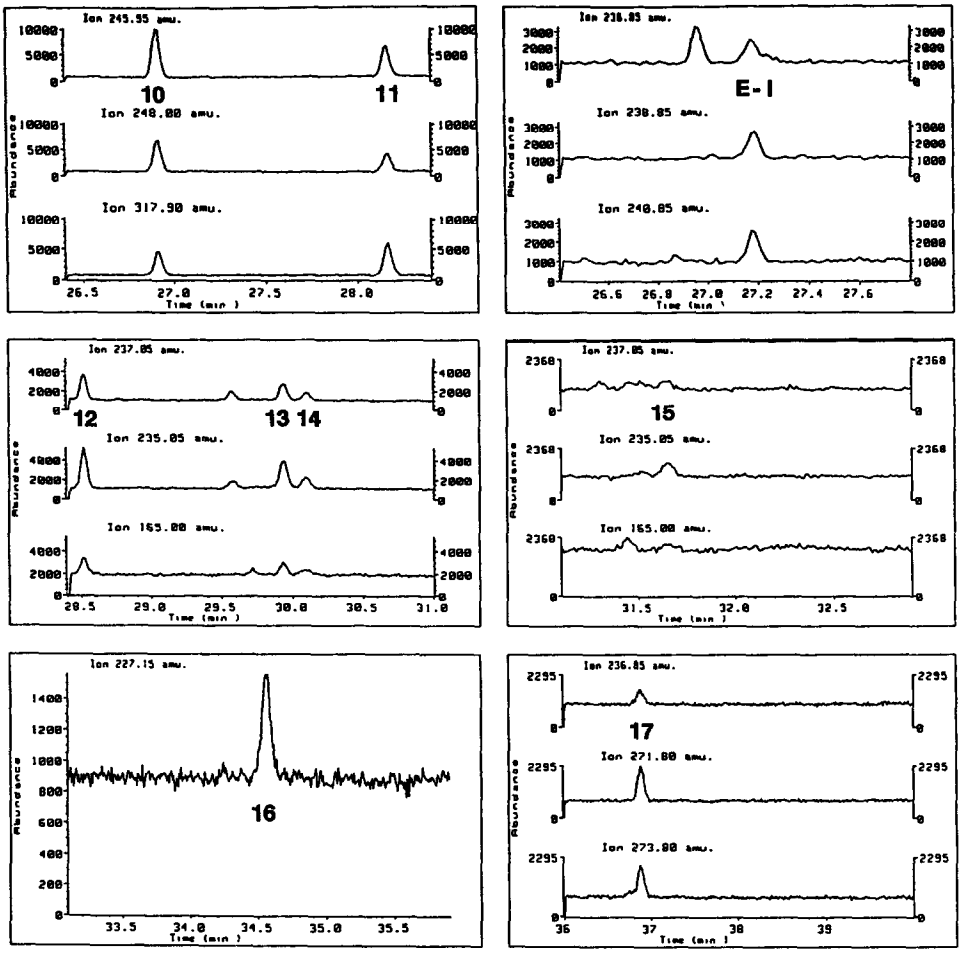

Fig. 5. SIM of chlorinated pesticides in green pepper. The food sample was spiked with 10 ppb of the following pesticides: 1 = HCB; 2 = α-HCH; 3 = β-HCH; 4 = lindane; 5 = δ-HCH; 6 = quintozene; 7 = heptachlor; 8 = *cis*-heptachlorepoxide; 9 = *trans*-heptachlorepoxide; 10 = o,p'-DDE; 11 = p,p'-DDE; 12 = o,p'-DDD; 13 = p,p'-DDD; 14 = o,p'-DDT; 15 = p,p'-DDT; 16 = methoxychlor; 17 = mirex. E-I = 50 ppb endosulfan I detected in the food sample. A = Aldrin (500 ppb) as internal standard.

where 18 chlorinated pesticides added to a vegetable sample at a concentration of 10 ppb were correctly detected. The knowledge of the retention times of the pesticides allowed the programming of time intervals with the appropriate selective ions. So it was possible to screen for all the important chlorinated pesticides in one GC analysis. The baseline of most of the ion traces was remarkably smooth. HCB forms a very stable molecular ion that produced the most intense signal of all compounds. Quintozene with only one of the six chlorine atoms substituted by the nitro group produces two ions in the higher mass region that only just formed a visible signal. It must be pointed out that the detection sensitivity for chlorinated pesticides approaches that of ECD, which has the reputation of being the most sensitive detection mode for

this compound group. One pesticide, however, can be detected far more sensitively with SIM than with ECD, namely methoxychlor which has never been traced at the 10 ppb level by ECD in our laboratory.

CONCLUSIONS

The results presented here demonstrate that capillary GC in connection with MSD allows one to detect and identify pesticide residues at concentrations which can be compared with those achieved by the other selective detectors in common use. Therefore, MSD can be highly recommended in pesticide residue analysis and in the control of other environmental pollutants for confirmatory analysis. This detector also has the potential for application to routine screening analyses.

A compilation of the mass spectra of all pesticides that can be determined with GC is considered as a prerequisite to achieve reliable results. It is recommended that they be filed in a separate sub-library with their common names. Otherwise, in routine analysis it may happen that a detected pesticide is not recognized because of its strange name when the pesticide is included in a hit list of 10 or 20 compounds in the PBM report. A few examples can be found in Table II. More important, however, is the creation of a list of all pesticides with their retention times and most suitable masses for SIM analysis.

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