Residues of Pentachloronitrobenzene and Related Compounds in Greenhouse Soils

by

R. H. DE VOS, M. C. TEN NOEVER DE BRAUW, and P. D. A. OLTHOF

Central Institute for Nutrition and Food Research TNO
Zeist, The Netherlands

Pentachloronitrobenzene (PCNB, also known as Quintozene, Terraclor) is used as a soil fungicide in greenhouses, mainly for the control of Rhizoctonia and Botrytis in lettuce.

During an examination (by gas liquid chromatography) of a number of soil samples from greenhouses not only a PCNB peak was found in the chromatograms, but also several other peaks, which were thought to be related to that compound. The identity of these peaks was investigated by mass spectrometry. The results of this study are described below.

EXPERIMENTAL

The soil samples were collected at random from greenhouses in several parts of The Netherlands, but mainly in the western part. In most cases PCNB had been used regularly as a soil fungicide.

The samples were analysed as received (without drying). The following extraction procedure was used.

To 50 g of a homogenized sample 10 ml water was added. After mixing, the sample was shaken for 30 minutes with 50 ml toluene plus 25 ml isopropylic alcohol. The solvent was decanted, and diluted with 150 ml of a 2 % aqueous sodium sulphate solution. After shaking the phases were allowed to separate. The aqueous layer was discarded. The toluene was washed with another 150 ml portion of aqueous sodium sulphate solution. After removing most of the aqueous layer, the toluene phase was centrifuged. A quantity of 10 ml of the clear toluene extract was transferred to a reagent tube with glass stopper. The liquid was shaken for 1 minute with 1 g Nuchar-Attaclay (Varian-Aerograph Cat. No. 2-003070-00). The mixture was then filtered. The filtrate obtained was analysed by gas-liquid chromatography with electron-capture detection (EC-GLC). The samples were diluted when necessary with hexane in order to operate within the linear range of the EC-detector.

Bulletin of Environmental Contamination & Toxicology,
Vol. 11, No. 6 © 1974 by Springer-Verlag New York Inc.

The method described above is used in this laboratory on a routine basis for soil samples. It gives good recoveries for a number of chlorinated pesticides, including PCNB.

Ten samples containing PCNB and the additional peaks mentioned under 'Introduction' were taken for an examination by a computer coupled mass-spectrometer/gas chromatography system (MS-GLC). In this case the purified toluene solutions were concentrated about 50 fold, in order to improve sensitivity. The instrumental conditions applied were as follows.

EC-GLC

Instrument: Microtek MT220 with two columns and two Ni-63 detectors (operating with pulsed voltage). Temperatures: - columns 185 °C, - injector 220 °C, - detectors 275 °C. Carrier gas: Argon with 10 % methane. The following two columns were used for each analysis:

a. Pyrex, 1.9 m x 4 mm (i.d.), containing 3 %OV-1 on Gas Chrom Q, 80-100 mesh. Carrier gas flow 55 ml/min.
b. Pyrex, 1.9 m x 4 mm (i.d.), containing 1.5 % OV-17 + 1.95 %OV-210 on Gas Chrom Q, 80-100 mesh. Carrier gas flow 85 ml/min.

MS-GLC

Instrument: Single focussing mass spectrometer CH4 Varian Mat connected through a silicon membrane separator with an Aerograph 1700 chromatograph. This combination is connected with a computer system SS 100 Varian Mat. The system comprises an A/D converter, a 620 i computer with 12 K core memory 16 bits words, two magnetic tape units, a scope monitor, an electrostatic plotter and teletype. The GLC analyses were performed on a 5 ft x 1/8 in. pyrex glas column packed with 10 % DC 200 on Gas Chrom Q 80-100 mesh. Temperature: 180 °C.Carrier gas: Helium, flow rate 20 ml/min. The mass spectrometer operated in the cyclic scan mode, taking every 3 sec a mass spectrum from the GLC effluent entering the mass spectrometer.

Several hundreds of mass spectra are obtained and stored on magnetic tape. When the MS-GLC analyses are completed the spectro system can supply the total ion current chromatogram, selected mass plots or mass chromatograms, and the mass spectra at any spot of interest in the chromatogram.

Further details are described by TEN NOEVER DE BRAUW and KOEMAN (1973).

RESULTS AND DISCUSSION

The compounds that could be identified in the soil extracts by MS-GLC are listed in Table 1.

TABLE 1

Compounds identified by MS-GLC in soil extracts

Identity	relative retention (aldrin=1.00) determined by EC-GLC	
	OV-1[1]	OV-17/OV-210[1]
Pentachloronitrobenzene (PCNB)	0.48	0.67
Pentachloroaniline (PCA)	0.63	0.87
Pentachlorothioanisole (PCTA)	0.88	1.12
Tetrachloronitrobenzene (TCNB)	0.27	0.35
Tetrachloroaniline (TCA)	n.d.[2]	n.d.
Tetrachlorothioanisole (TCTA)	n.d.	n.d.
Hexachlorobenzene (HCB)	0.42	0.43
Pentachlorobenzene (QCB)[3]	0.22	0.21

[1] see "Experimental"

[2] n.d. = not determined (no standard being available)

[3] QCB = quintochlorobenzene, this abbreviation is used rather than PCB in order to avoid confusion with polychlorobiphenyls

Their relative retentions (aldrin = 1.00) on the columns used in the EC-GLC system are included in this table. A typical chromatogram obtained by MS-GLC is shown in Fig. 1.

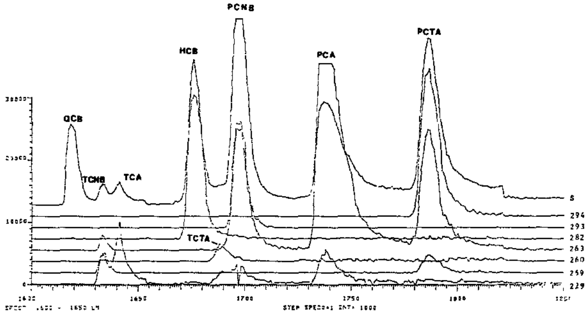

Fig. 1. Computer plot of a MS-GLC run (total ion current S and selected mass plots for the characteristic masses).

Among the compounds listed in Table 1 special attention was paid to pentachlorothioanisole (PCTA or methyl pentachlorophenyl sulfide). This compound has been identified as a metabolite of PCNB in dogs, rats and plants (KUCHAR et al., 1969). To our knowledge the presence of PCTA in PCNB treated soils has not been reported so far. As a further check the mass spectrum of synthesized PCTA, and of the peak in the sample chromatograms were compared. They were found identical. The mass spectrum is shown in Fig. 2.

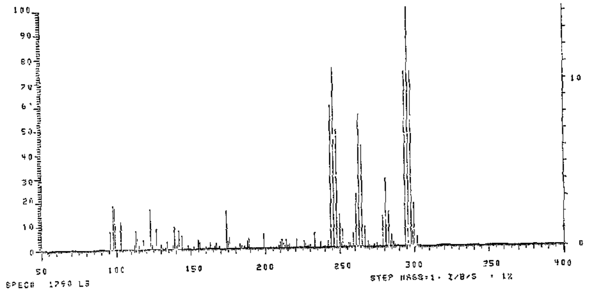

Fig. 2. Mass spectrum No. 1790LS of pentachlorothioanisole

The other compounds that were identified in the soil extracts can all be attributed to the application of PCNB to the soil. Pentachloroaniline (PCA) is a well known degradation product of PCNB. PCA is formed by micro-organisms in moist soils (KO and FARLEY, 1969).

Hexachlorobenzene (HCB) and pentachlorobenzene (QCB) are impurities in technical PCNB (FAO/WHO, 1969). The same applies to tetrachloronitrobenzene (TCNB). According to KUCHAR et al. (1969) the impurity in PCNB is 2,3,4,5-TCNB. The peaks found in the soil extracts had the same retention time as 2,3,5,6-TCNB. The latter compound ("Tecnazene") is also applicated in smokes as a fungicide for lettuce. Since no pure 2,3,4,5-TCNB was available it could not be checked which isomer was present in the soil extracts.

As was mentioned already EC-GLC was used as a screening prior to MS-GLC. Quantitation was carried out in this stage for residues of PCNB, HCB and TCNB, as these compounds are determined on a routine basis by the method described under "Experimental". The results are given in Table 2.

All data were confirmed qualitatively with MS-GLC. The other compounds (PCA, PCTA, TCA, TCTA, QCB) were analyzed qualitatively only by MS-GLC. Positive findings are indicated with + in Table 2. Quantitation was not possible, because elaborate recovery studies have not yet been carried out. Limited experiments indicated that with the extraction and

TABLE 2

Results of the analysis of 10 soil samples by EC-GLC and MS-GLC. Values are expressed in ppm (on wet weight basis)

Sample no.	PCNB	PCA	PCTA	TCNB	TCA	TCTA	HCB	QCB
I	3.3	+	+	0.09			0.25	+
II	1.2	+	+	0.13	+	+	0.25	+
III	1.7	+	+		+	+	0.18	+
IV	0.03	+					0.01	+
V	0.26	+	+				0.05	+
VI	1.2	+	+	0.06	+		0.13	+
VII	0.66	+	+		+		0.19	+
VIII	3.9	+	+	0.17			0.54	+
IX	0.11	+	+				0.03	+
X	0.05	+	+				0.02	+

clean-up method used, the recovery is about 40 % for PCA, and 90 % for PCTA (both determined at a 0.1 ppm level).

From the EC chromatograms it could be estimated that the PCA residues in the 10 soil samples ranged from 15-150 % of the PCNB residue present. For PCTA this range was 10-90 %. For more accurate data a quantitative technique will have to be worked out which is suitable for the simultaneous analysis of all compounds listed in Table 1.

ACKNOWLEDGEMENT

The authors gladly acknowledge the co-operation of: L. BRAVENBOER (Research and Experiment Station for Fruit and Vegetables under Glass, Naaldwijk, The Netherlands) for providing the samples; R.J.C. KLEIPOOL for synthesizing pentachlorothioanisole; and J. BOUWMAN and A.E. BROUWER for their technical assistance.

REFERENCES

FAO/WHO: 1969 Evaluations of some pesticide residues in food, p. 199, Rome, 1970.

KO, W.H. and FARLEY, J.D.: Phytopathol. 59, 64 (1969).

KUCHAR, E.J., GEENTY, F.O., GRIFFITH, W.P. and THOMAS, R.J.: J. Agr. Food Chem. 17, 1237 (1969).

TEN NOEVER DE BRAUW, M.C. and KOEMAN, J.H.: Sci. Total Environment 1, 427 (1973).