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Determination of permethrine and tetramethrine by isotachophoretic analysis of hydrolytic products

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ABSTRACT

A method for the isotachophoresis determination of the pyrethroid insecticides permethrine and tetramethrine in water is described. After extracting the insecticides from samples and evaporating the solvent, alkaline hydrolysis was carried out. The degradation products, *cis*- and *trans*-dichlorochrysanthemic acid, *cis*- and *trans*-chrysanthemic acid and phthalimide, were identified and determined by means of capillary isotachophoresis. The detection limit is 0.01 mg/l^{-1} for both insecticides with recoveries of 80–89% and 91–99% for permethrine and tetramethrine, respectively.

INTRODUCTION

Permethrine and tetramethrine are pyrethroid insecticides, for the determination of which various methods have been used. Recently voltammetric [1], bioassay [2] and spectrophotometric [3] techniques have been used. However, separation methods are the most important requirement. Some gas chromatographic (GC) determinations of permethrine and tetramethrine have been reported, using packed [4] and capillary columns [5], with flame ionization [6], electron-capture [7–9], Coulson electrolytic conductivity [10] and microwave plasma detection [11].

High-performance liquid chromatographic (HPLC) methods have been used in the normal- and reversed-phase modes with UV [12] and IR [13,14] detection. Columns with chiral packings are a promising approach [15].

Capillary isotachophoresis (ITP) has not previously been used for determining permethrine and tetramethrine insecticides. The direct isotachophoretic determination of non-ionic pesticides such as pyrethroids is not possible, but they can be determined after hydrolysis to ionic products.

This work is based on previous studies on the alkaline hydrolysis of alphamethrine [16] and the determination of alphamethrine and cypermethrine by ITP [17].

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EXPERIMENTAL

Chemicals

The solvents used were of Pestanal or analytical-reagent grade: pentane, light petroleum (Riedel-de Haën, Seelze, Germany), *tert.*-butyl alcohol (Reanal, Budapest, Hungary) (redistilled), diethyl ether and ethanol (Lachema, Brno, Czechoslovakia).

Chemically pure or analytical-reagent grade chemicals were used for ITP analyses: creatinine (Riedel-de Haën), morpholinoethanesulphonic acid (MES) (Serva, Heidelberg, Germany), and poly (vinyl alcohol) (PVA) Gohsenol GM-14L (M_r = 62 800). Hydrochloric acid was prepared by isothermal distillation and deionized water was prepared with a specific conductivity up to 1.5 μS cm^{-1} (mixed ion-exchange bed). Potassium phthalimide (Riedel-de Haën), anhydrous sodium sulphate (Lachema), argon (M 380) and a solution of Kolthoff-Vleeschhouwer buffer (pH 12, I = 0.193) were used.

Standards were chrysanthemic acid (*cis*, *trans*) and dichlorochrysanthemic acid (*cis*, *trans*), synthesized and provides by the Polish Academy of Sciences.

Model samples of water were prepared by addition of the formulations Reslin 25 EC (Wellcome Foundation, Berkhamsted, U.K.) [97.5 g l^{-1} of permethrine (75% *trans* and 25% *cis* isomers)] and Neopynamine NPB 13 EC (Sumitomo, Osaka, Japan) (13% of tetramethrine) to potable tap water to ensure a content of active compounds within the range 0.01–5.0 mgl^{-1}.

Apparatus

Isotachophoretic analyses were performed on a ZKI 001 ITP analyser (URVJT, Spišská Nová Ves, Czechoslovakia) in one- and two-capillary arrays. With the two-capillary system the current in the preseparation capillary (150 × 0.3 mm I.D.) was 30 μA. When using one capillary, the current was decreased from 100 to 30 μA to detection. The signal of the conductivity detector was recorded on a two-channel recorder at chart drive speeds of 1 and 2.5 mm s^{-1}. Sampling was performed with 10-, 25- and 50-μl microsyringes (Hamilton, Reno, NV, U.S.A.).

Isolation of insecticides from water

The isolation of permethrine and tetramethrine from water was tested by extraction with organic solvents (diethyl ether, pentane, light petroleum). The content of active compounds in the organic phase was determined by ITP after alkaline hydrolysis.

Double extraction with diethyl ether was used for the determination of permethrine. The water samples (volume 800 ml with addition of 80 g of sodium sulphate) were shaken with 40 ml and 15 ml of diethyl ether; for 10 min each. Double extraction with light petroleum was used for isolating tetramethrine. The water samples (volume 800 ml with addition of 20 g of sodium sulphate) were shaken with 20 ml and 15 ml of light petroleum, for 20 min each. The combined organic phases were evaporated on a water-bath with argon after transfer into glass ampoules.

All experiments were repeated three times. The blank was measured in the same way.

Alkaline hydrolysis

Alkaline hydrolysis of permethrine and tetramethrine was carried out in sealed glass ampoules (10 ml) in an argon atmosphere. Ethanol and *tert.*-butyl alcohol solutions of both insecticides (2.0 ml; $2 \cdot 10^{-3}$ mol l^{-1}) were added to 2.0 ml of buffer solution. After removal of air by argon, the ampoules were sealed and thermostated at 50, 65 and 80°C. After 24 h the ampoules were cooled and the contents were diluted to 25 ml with deionized water in a volumetric flask. A 10-μl volume of this sample was injected into the ITP analyser. The same procedure was applied without the argon atmosphere.

The hydrolysis of tetramethrine (2.0 ml of *tert.*-butyl alcohol solution) was carried out gradually with 1.0, 1.5, 2.0, 2.5, 3.0 and 4.0 ml of buffer solution to establish the influence of the alcohol:buffer volume ratio. The same procedure was carried out in an argon atmosphere with an ethanol solution of permethrine.

The time dependence of the recovery of hydrolytic products was studied by hydrolysis of 2 ml of tetramethrine in *tert.*-butyl alcohol ($2 \cdot 10^{-3}$ mol l^{-1}) with 1.5 ml of buffer solution in an air atmosphere. Time intervals were in the range 30 min–24 h. The same procedure was applied with permethrine (2 ml of $2 \cdot 10^{-3}$ mol l^{-1} solution in ethanol with 2.5 ml of buffer).

The hydrolysis of extracts of real samples was carried out as follows. For tetramethrine, after adding 0.65 ml of *tert.*-butyl alcohol and 0.5 ml of a buffer to the dry residue the ampoules were sealed and placed into the thermostat. After 6 h the cooled contents of the ampoules were transferred with deionized water into a 10 ml volumetric flask, diluted to volume and analysed by ITP. For permethrine, 0.5 ml of ethanol and 0.63 ml of buffer were added to the dry residue and, after air had been removed with argon, the ampoules were sealed and thermostated for 24 h at 50°C. The contents of the ampoules were transferred into 10-ml volumetric flask, diluted to volume and analysed by ITP.

All experiments were repeated three times.

ITP determination of decomposition products

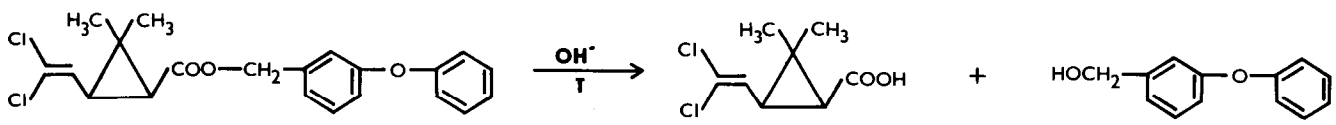
The samples prepared by hydrolytic procedures were analysed by ITP in the following operational system: leading electrolyte (L) = HCl (10^{-2} mol l^{-1}) + creatinine + PVA (0.05%), pH_L = 4.80; terminating electrolyte (T) = MES ($5 \cdot 10^{-3}$ mol l^{-1}).

The single- and double-capillary systems were used for the analyses. The contents of chrysanthemic and dichlorochrysanthemic acids and phthalimide in the hydrolysates were determined by the linear calibration method.

Determination of permethrine and tetramethrine in water

For the determination of permethrine in water samples, a double extraction with diethyl ether and hydrolysis in ethanol solution (alcohol:buffer = 1:1.25) were used. For tetramethrine double extraction with light petroleum and hydrolysis in *tert.*-butyl alcohol solution (alcohol:buffer = 1:0.75) were used.

The water samples (ten different concentrations the range 0.01–5.0 mg l^{-1}) were extracted and, after evaporation of the solvent, the total solids were hydrolysed. Degradation compounds (dichlorochrysanthemic acid and phthalimide) were determined by ITP of the hydrolysates.

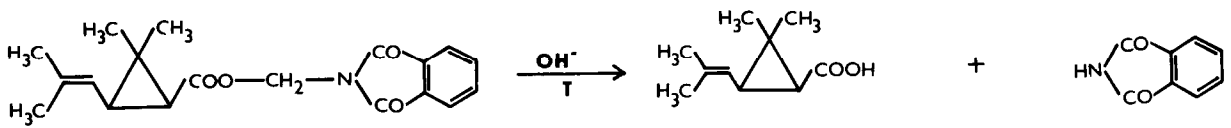


Permethrine

cis,trans-Dichlorochrysanthemic acid

(I)

Phenoxybenzyl alcohol



Tetramethrine

cis,trans-Chrysanthemic acid

(II)

Phthalimide

(III)

Fig. 1. Scheme of alkaline hydrolysis of permethrine and tetramethrine.

All experiments were repeated three times. The blank was measured in the same way.

RESULTS AND DISCUSSION

The reaction scheme of the alkaline hydrolysis of permethrine and tetramethrine is shown in Fig. 1. The degradation products, (I) dichlorochrysanthemic acid (from permethrine) and (II) chrysanthemic acid and (III) phthalimide (from tetramethrine), can be separated by ITP using one- and two-capillary arrays. Their ITP separation is shown in Fig. 2. The ITP determination of phenoxybenzyl alcohol is not possible as it does not migrate during the ITP separation.

The hydrolysis of permethrine in ethanol solution under an argon atmosphere gave the best recovery under the described conditions. With tetramethrine the best

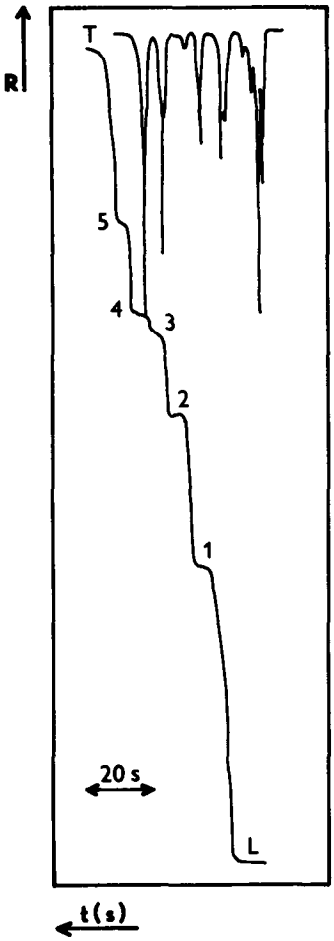


Fig. 2. Separation of degradation products. Injection of 40 μl of mixture ($1.5 \cdot 10^{-5}$ mol l^{-1}); one-capillary system; I = 30 μA. 1 = III; 2 = *trans*-I; 3 = *cis*-I; 4 = *trans*-II; 5 = *cis*-II. L = Cl^-; T = MES; t = time; R = resistance.

TABLE I

INFLUENCE OF TEMPERATURE ON THE RECOVERY OF HYDROLYSIS (AFTER 24 h)

Mean recoveries with standard deviations (n = 9).

Temperature (°C)	Dichlorochrysanthemic		Chrysanthemic		Phthalimide	
	Recovery (%)	S.D. (%)	Recovery (%)	S.D. (%)	Recovery (%)	S.D. (%)
50	92.0	2.32	57.2	1.82	90.6	2.21
65	92.5	2.28	57.8	1.91	90.9	2.25
80	92.9	2.31	57.9	1.78	91.2	2.19

results were attained *tert.*-butyl alcohol solution with an air atmosphere. Higher temperature of hydrolysis (65 and 80°C) did not significantly improve the recovery of hydrolysis (Table I). A temperature of 50°C was used in subsequent work.

The influence of alcohol:buffer volume ratio is important for the recovery of hydrolytic products (Table II). The best results were attained in solution with alcohol: buffer ratios of 1:1.25 for permethrine and 1:0.75 for tetramethrine.

The recovery of hydrolysis was calculated from the ITP-determined concentrations of compounds I (dichlorochrysanthemic acid), II (chrysanthemic acid) and III (phthalimide) in the hydrolysates. The time dependences were constructed as shown in Fig. 3.

Isolation of the insecticides from water by double extraction with diethyl ether (permethrine) or light petroleum (tetramethrine) gave the best results (Table III).

The ITP determination of permethrine and tetramethrine in water after extraction and hydrolysis yields reproducible and consistent results within a broad range of concentrations. The detection limits were of the order of 0.01 mg l^{-1} for both compounds. The method distinguished the *cis* and *trans* isomers of the insecticides. The recovery of the method is 80–89% for permethrine [relative standard deviation

TABLE II

INFLUENCE OF ALCOHOL:BUFFER VOLUME RATIO ON THE RECOVERY OF HYDROLYSIS (AFTER 24 h)

Mean recoveries with standard deviations (n = 9).

Alcohol:buffer volume ratio	Dichlorochrysanthemic		Chrysanthemic		Phthalimide	
	Recovery (%)	S.D. (%)	Recovery (%)	S.D. (%)	Recovery (%)	S.D. (%)
1:0.5	65.3	1.65	53.0	1.67	88.2	2.18
1:0.75	89.1	2.23	54.2	1.72	94.0	2.34
1:1	92.0	2.32	57.2	1.82	90.5	2.21
1:1.25	95.8	2.44	55.9	1.77	88.1	2.16
1:1.5	74.5	1.86	53.0	1.65	71.4	1.81
1:2	35.2	0.75	53.1	1.68	59.0	1.45

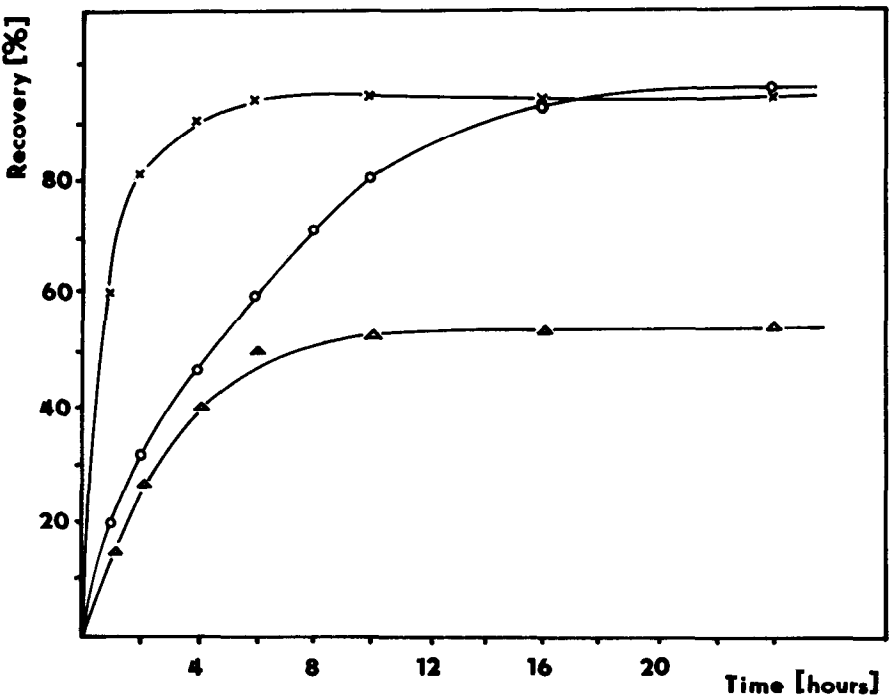


Fig. 3. Time dependences of recovery of hydrolysis (optimum conditions). △, Chrysanthemic acid (both isomers); ○, dichlorochrysanthemic acid (both isomers); X, phthalimide.

TABLE III

RECOVERY OF THE EXTRACTION OF PERMETHRINE AND TETRAMETHRINE FROM WATER (20 ml OF SOLVENT TO 400 ml OF WATER)

Mean recoveries with standard deviations (n = 9). Concentrations of insecticides present: 5.0 mg l^{-1}.

Solvent	Permethrine		Tetramethrine	
	Recovery (%)	S.D. (%)	Recovery (%)	S.D. (%)
Diethyl ether	88	1.31	44	1.68
Pentane	41	1.58	66	1.51
Light petroleum	44	1.21	98	2.03

TABLE IV

DETERMINATION OF PERMETHRINE AND TETRAMETHRINE IN WATER

Mean recoveries with standard deviations (n = 9).

Concentration of insecticides (mg l^{-1})	Permethrine		Tetramethrine	
	Recovery (%)	S.D. (%)	Recovery (%)	S.D. (%)
0.02	81.5	4.13	92.1	4.84
0.05	83.8	4.02	94.3	4.75
0.2	85.2	3.52	97.6	4.28
0.5	89.0	3.27	98.2	4.35
2.0	87.8	3.43	97.9	4.21

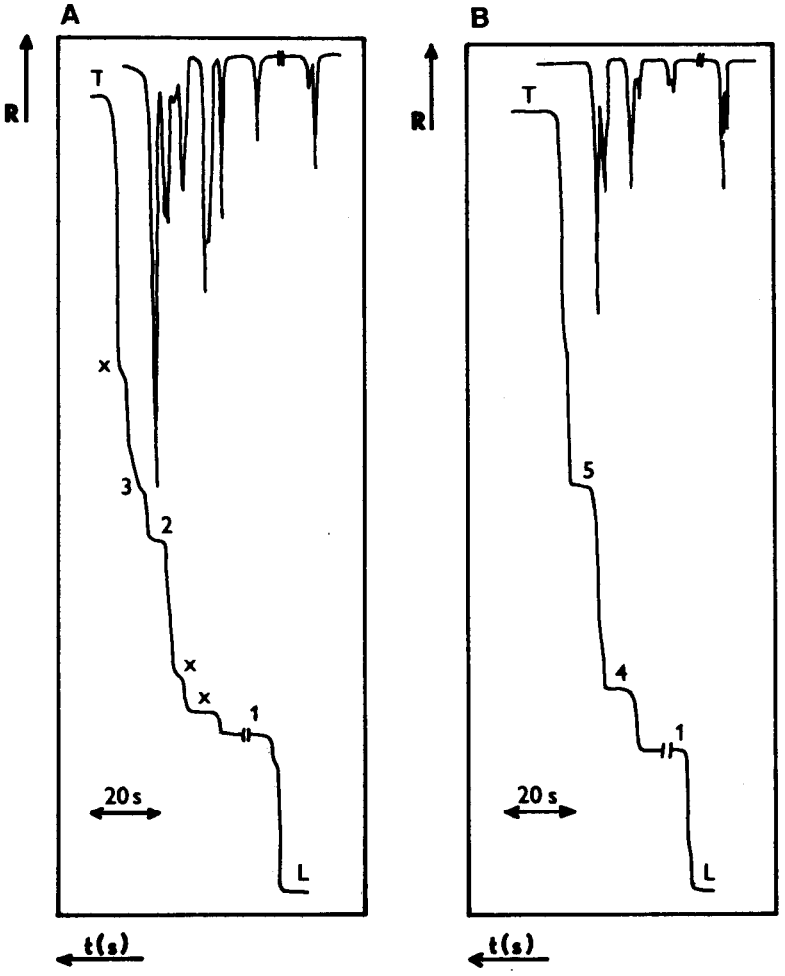


Fig. 4. Determination of (A) permethrine and (B) tetramethrine in water (0.2 mg l^{-1}). Injection of 20 μl of hydrolysate; one-capillary system; I = 30 μA. 1 = PO_4^{3-}; 2 = *trans*-I; 3 = *cis*-I; 4 = III; 5 = *trans*-II. L = Cl^-; T = MES, x = impurities; t = time; R = resistance.

(R.S.D.) = 4.15%] and 91–99% for tetramethrine (R.S.D. = 4.86%) in the range of concentrations of the insecticides in water of 0.01–5.0 mg l^{-1}.

Selected results of the ITP determination of permethrine and tetramethrine in water are summarized in Table IV. Typical isotachophoreograms are shown in Fig. 4. It is concluded that the proposed method is suitable for routine analyses in agriculture and ecology.

REFERENCES

1 P. Hernandez, J. Vicente and L. Hernandez, *Fresenius' Z. Anal. Chem.*, 334 (1989) 550.
2 L. H. Stanker, C. Bigbee, J. van Emon, E. Watkins, R. H. Jensen, Ch. Morris and M. Vanderlaan, *J. Agric. Food Chem.*, 37 (1989) 834.
3 J. M. Desmarchelier, *J. Stored Prod. Res.*, 12 (1976) 245.
4 N. Baba, A. Nagayasu and M. Ohno, *Agric. Biol. Chem.*, 34 (1970) 343.
5 A. Richard and G. Andermann, *J. Chromatogr. Sci.*, 22 (1984) 207.
6 R. A. Simonaitis and R.S. Cail, *Chromatographia*, 18 (1984) 556.
7 M. Chiba, *J. Environ. Sci. Health*, B13 (1978) 261.
8 A. E. S. M. Marei, L. O. Ruzo and J. E. Casida, *J. Agric. Food Chem.*, 30 (1982) 558.
9 W. Ebing, *Fresenius' Z. Anal. Chem.*, 327 (1987) 539.
10 G. H. Fujie and O. H. Fullmer, *J. Agric. Food Chem.*, 26 (1978) 395.
11 M. L. Bruce and J. A. Caruso, *Appl. Spectrosc.*, 39 (1985) 942.
12 D. Mourot, B. Delepine, J. Boisseau and G. Gayot, *J. Chromatogr.*, 168 (1979) 277.
13 E. Papadopoulou-Mourkidou, Y. Iwata and F. A. Gunter, *J. Agric. Food Chem.*, 29 (1981) 1105.
14 E. Papadopoulou-Mourkidou, Y. Iwata and F. A. Gunter, *J. Agric. Food Chem.*, 31 (1983) 629.
15 T. Doi, S. Sakane and M. Horiba, *J. Assoc. Off. Anal. Chem.*, 68 (1985) 911.
16 V. Dombek and Z. Stránský, *J. Chromatogr.*, 470 (1989) 235.
17 V. Dombek, *Thesis*, Palacký University, Olomouc, 1989.