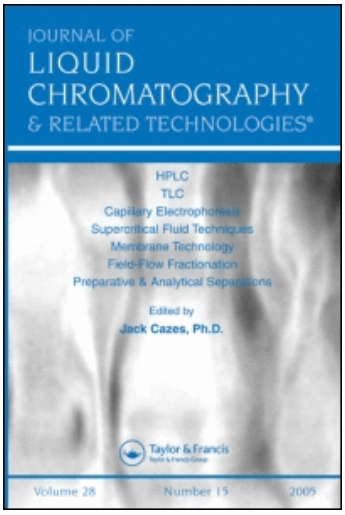

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RAPID AND SENSITIVE DETERMINATION OF CARBARYL, CARBOFURAN AND FENOBUCARB BY LIQUID CHROMATOGRAPHY WITH ELECTROCHEMICAL DETECTION

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ABSTRACT

A rapid and sensitive procedure is described for the determination of the N-methylcarbamates pesticides, carbaryl, carbofuran and fenobucarb, by liquid chromatography. The pesticides are previously hydrolyzed, in a separate step, to their phenolic derivatives, which, after the separation in a high speed column in a time lower than 2.5 min, are detected in a high sensitivity amperometric cell at a potential of +0.6 V. The method presents detection limits around 0.7 ppb and has been satisfactorily applied to the analysis of these compounds in spiked river water samples.

INTRODUCTION

Carbaryl, 1-naphtyl methylcarbamate, carbofuran, 2,3-dihydro-2,2-dimethylbenzo-furan-7-yl methylcarbamate, and fenobucarb, 2-sec-butylphenyl methylcarbamate, are pesticides of the N-methylcarbamates family, which have a wide range of applications and whose popularity has increased in the time, due i.e., to their short life. A number of papers appear in the bibliography,

concerning their determination in a variety of matrices, by high performance liquid chromatography. The separation between pesticides is generally carried out by reverse phase chromatography and with acetonitrile-water or methanol-water mobile phases. The detection is, in most cases, by UV spectrometry or by fluorimetry, and only in some instances are electrochemical methods used.[1-6] However, these can offer some advantages as greater sensitivity and selectivity without the need of various-step derivatization procedures as in fluorescence. Also, there are references about the coupling of HPLC systems with other techniques such as flow injection analysis, gas chromatography or mass spectrometry. Many of these methods make use of derivatization with different reagents after an alkaline hydrolysis which produces the phenolic derivatives of pesticides.

The hydrolysis reaction is also a good approach to carry out the electrochemical detection since phenols are molecules more easily oxidizable than carbamates, whose direct oxidation implies frequently to apply very high potentials (near +1.3 V). Kissinger et al[7] suggested this possibility, for the first time for carbofuran without going deeply into it, and, later, Olek et al[1] have studied the possibility of measuring several N-methylcarbamates after a pre-column hydrolysis technique. A potassium hydroxide solution in methanol is used to perform the hydrolysis at a temperature of 90 °C, and the procedure, which presents detection limits between 0.01 and 0.04 μg/mL (0.5 to 2 ng injected in 50 μL), is examined for the determination of carbamates in vegetables after liquid partitioning, solvent evaporation and purification on Florisil column steps. A subsequent paper by Krause et al.[2] describes a procedure in which carbamates are separated on a C_8 column using a gradient acetonitrile-water mobile phase and hydrolyzed in-line by post column addition of sodium hydroxide at 100 °C, and detected with a coulometric electrochemical detector. However, the separation takes near 25 min. and the method requires the removal of oxygen from the mobile phase solvents and sodium hydroxide solution. In this way detection limits of around 0.015 μg/mL (0.3 ng in an injected volume of 20 μl) are achieved.

In this paper, we describe a method to analyze three N-methylcarbamates, carbaryl, carbofuran and fenobucarb, in water, by HPLC. The latter is analyzed for the first time by LC with electrochemical detection. The pesticides are hydrolyzed in a prior separate step but the reaction is carried out by the simple addition of 0.02 M sodium hydroxide, at room temperature, according to the results recently reported about the hydrolysis of carbaryl.[8] The separation is effected on a high-speed column using an isocratic mobile phase similar to that used by Olek, and in a time shorter than 2.5 min., and the electrochemical detection is carried out with a high sensitivity amperometric cell. Thus, a simple and rapid procedure is proposed which presents lower detection limits than those mentioned for the other described procedures.

MATERIALS

Apparatus

The chromatographic equipment is composed of a 420 two piston HPLC Pump from Kontron Instruments, a 7125 Rheodyne sample injector equipped with a 20 μL loop, a Pecosphere 3x3 CR C_{18} column (3.3 cm x 0.46 cm, 3μm), a Coulochem II electrochemical detector equipped with a ESA model 5021 conditioning cell and a ESA model 5011 dual analytical cell protected by ESA filters containing 0.2 μm porous graphite filter elements. The high sensitivity analytical cell contains, in series, two porous graphite working electrodes, together with associated reference and counter electrodes.

The working electrodes are a large surface area coulometric electrode and a high efficiency amperometric electrode, more than seven times as efficient as conventional amperometric electrodes (70% vs. 5-10% efficiency). The conditioning cell contains a single porous graphite coulometric electrode.

The acquisition and treatment of data is controlled from an Olivetti 386 PC equipped with the PC Integration Pack software package from Kontron Instruments.

Reagents

Carbofuran, carbaryl and fenobucarb were obtained from Sigma Chemical Co. and used without further purification. Stock solutions of each pesticide in Milli Q grade water (2.5×10^{-4} M) were prepared by weighing. More diluted standards were prepared by suitable dilution. All other chemicals used were of HPLC grade.

HPLC Operating Parameters

The mobile phase was glacial acetic acid/water/acetonitrile, 0.5/49.5/50 made 0.01 M in sodium perchlorate. It was filtered through a 0.45 μm nylon membrane filter and degassed in ultrasonic bath before being used. The flow rate was adjusted to 1.0 mL/min and the system was equilibrated for at least 10 min. prior to injection of the prepared sample or standard.

The conditioning cell was set at 0 V and the detectors 1 and 2 of the analytical cell were set at +0.1 and +0.6 V respectively. The selected sensitivity in the PC Integration Pack was 500 nA full scale (1V).

METHODS

General Procedure for the Determination of Carbofuran, Carbaryl and Fenobucarb

To a 50 mL calibrated flask were added a suitable volume of the pesticide solution and 2 mL of 0.5 M NaOH solution. The solution was diluted with water to a volume near 40 mL and, after shaking for a few seconds, allowed to stand during 10 min. Then 1 mL of glacial acetic acid was added and the solution diluted to the mark with water. Samples were filtered through 0.45 μm nylon filter membranes and degassified in ultrasonic bath before their injection (20 μL) in the chromatographic system. Two chomatograms per sample were obtained and the mean of their peak heights was used as analytical signal.

Procedure for the Determination of Carbofuran, Carbaryl and Fenobucarb in River Water

Aliquots of 30 mL of river water, spiked with different amounts of the pesticides, were treated according to the general procedure and the obtained concentrations were calculated with the aid of the corresponding calibration plots.

RESULTS AND DISCUSSION

The alkaline hydrolysis of the pesticides carbofuran, carbaryl and fenobucarb gives rise to their phenolic derivatives, 2,3-dihydro-2,2-dimethyl-7-benzofuranol, 1-naphthol and 2-sec-butylphenol, respectively. These compounds are more easily oxidizable than the parents N-methylcarbamates and this can be made at a potential value lower than +1.0 V.

Recently,[8] it has been reported that quantitative hydrolysis of carbaryl can be obtained in only 30 s using sodium hydroxide solutions with concentrations ≥ 0.02 M. Also, the complete hydrolysis of carbofuran and fenobucarb is obtained in this medium in a short time and has been used as a previous step in their voltammetric determination.[9,10] If the hydrolyzed solutions are subsequently acidified they are stable at least during three hours.

At first, the hydrodynamic curves of the pesticides have been obtained to select the appropriate potential values to detect them in the used system. The mobile phase used was a solution 0.01 M of $NaClO_4$ in acetic

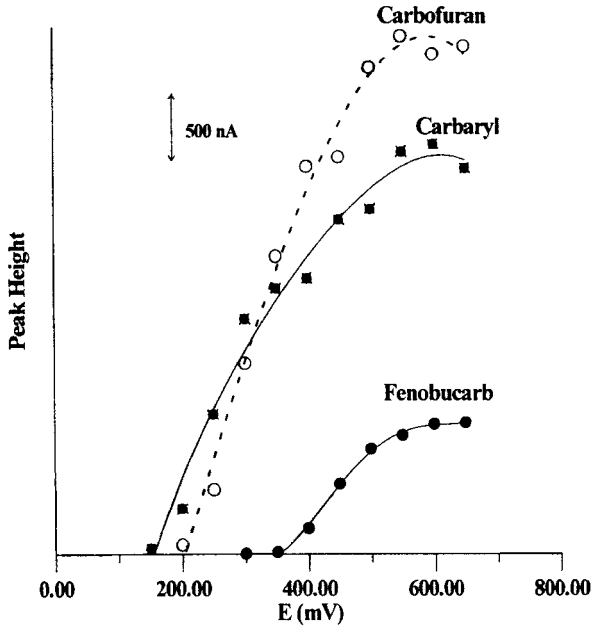

Figure 1. Hydrodynamic curves of carbofuran, carbaryl and fenobucarb in the porous graphite electrode of the coulometric detector.

acid:water:acetonitrile, 0.5:59.5:40, and the potential in the detector 1 was set at +0.1 V whereas the potential in the detector 2 is varied, taking different values between +0.15 and +0.75 V. In Figure 1 a plot of the results obtained is shown. According to these results, and in order to obtain the highest signal, we have selected to set the potential in the detector 2 at +0.6 V. The detector 1 was maintained at +0.1 V.

The influence of acetonitrile proportion in the mobile phase was studied varying it between 40% and 55% (v/v, acetonitrile/water), and the results can be found in Table 1. In all cases the resolution between the carbofuran and carbaryl peaks is satisfactory (Rs greater than 1), but we have selected a 50% proportion of acetonitrile due to the resolution value ($Rs_{1,2}$ greater than 1.5) and the capacity factor values (K' between 1 and 5) obtained. In these conditions the obtaining of the chromatogram takes only 2.5 min. The influence of the pH value of the mobile phase (between pH 2 and pH 5.5 with acetic acid or acetate buffer) on the resolution and capacity factor values, has also been studied. No differences have been observed but we have decided to prepare the mobile phase with acetic acid to avoid undesirable effects in base to changes in the pH values, with real samples, and variations in the oxidation potentials or in the

Table 1

Influence of the Acetonitrile Proportion in the Mobile Phase on the Retention of Pesticides Carbofuran, Carbaryl and Fenobucarb

	Carbofuran		Carbaryl			Febobucarb	
% AcCN (v/v)	t_R(min)	K′	t_R(min)	K′	$Rs_{1,2}$	t_R(min)	K′
40	1.35	2.97	1.86	4.17	3.64	4.31	11.68
45	1.17	2.08	1.51	2.96	2.29	3.02	6.95
50	1.00	1.63	1.21	2.18	1.58	2.17	4.71
55	0.86	1.32	1.00	1.70	1.06	1.64	3.43

detector response. The conductivity of mobile phase was increased with sodium perchlorate. Also, it is well known that the perchlorate are the less-adsorbable on the electrode ions, between the usually used. As result the selected mobile phase is that mentioned in the description of the chromatographic parameters: a solution 0.01 M of $NaClO_4$ in glacial acetic acid/water/acetonitrile 0.5/49.5/50.

In the study of the influence of the mobile phase flow rate we have found that the capacity factors only decrease slightly when the flow rate increases, and N (number of theoretical plates) changes in a similar way.

According to these results and the usefulness of obtaining the chromatogram in a short time we have selected a 1 mL/min flow rate. Different sensitivities have been assayed for the obtaining of the chromatograms and we have set it in 500 nA due to the good signal/noise ratio obtained. Figure 2 shows a chromatogram corresponding to a standard which is 3×10^{-8} M in the three analytes, obtained in the mentioned conditions

Determination Of Carbaryl, Carbofuran And Fenobucarb

Under the optimum experimental chromatographic conditions, a linear relationship between peak height or peak area and concentration of the pesticide was found for the three compounds in the range examined (4×10^{-9} M - 7×10^{-8} M for carbaryl and carbofuran and 4×10^{-9} M - 1×10^{-7} M for fenobucarb), showing the regression equation of Table 2. Due to the greater sensitivity (greater slope) and reproducibility (lower RSD) obtained by using peak height, this parameter has been selected as analytical signal to determine these analytes.

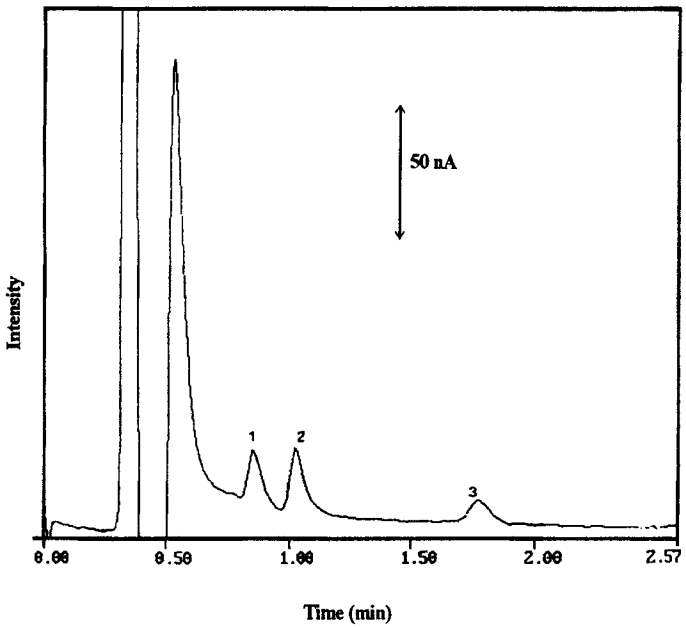

Figure 2. Chromatogram of a 3 x 10^{-8} M solution of carbofuran (1), carbaryl (2) and fenobucarb (3). Mobile phase: acetic acid/water/acetonitrile (0.5/49.5/50) 0.01 M in $NaClO_4$.

Table 2

Regression Parameters for the Pesticide Chromatographic Peaks

Compound	Signal	Slope	Intercept	Correlation Coefficient (r)	RSD (%) p=0.05, n=11
Carbofuran	Height	1.68×10^9	-7.82	0.9992	2.40
	Area	11.8×10^7	-0.37	0.9992	7.90
Carbaryl	Height	1.81×10^9	-2.85	0.9982	1.62
	Area	13.3×10^7	-0.12	0.9980	3.57
Fenobucarb	Height	6.16×10^8	+0.86	0.9997	2.95
	Area	6.65×10^7	+0.06	0.9930	5.88

Table 3

Detection Limit Values Calculated from the Chromatographic Peak Heights

Compound:	Carbofuran	Carbaryl	Fenobucarb
Method	**(M x 10^9)**	**(M x 10^9)**	**M x 10^9)**
3σ	1.00 (4.43)*	0.98 (3.94)	2.00 (8.29)
Winefordner and Long	2.11 (9.34)	3.04 (12.2)	1.60 (6.63)
Clayton ($\alpha = \beta = 0.05$)	2.60 (11.5)	3.76 (15.1)	2.14 (8.87)

* The quantities in brackets are the detection limit values in pg.

Table 4

Determination of Carbofuran, Carbaryl and Fenobucarb in Spiked River Water

Sample:	Concentration	1,2,3	4,5,6	7, 8, 9	10, 11, 12
Compound	**M X 10^8**				
Carbofuran	Added	3.0	5.0	4.0	6.0
	Found	2.86±0.09	4.75±0.03	3.82±0.06	5.76±0.16
Carbaryl	Added	3.0	5.0	4.0	5.0
	Found	2.96±0.22	4.76±0.32	3.91±0.23	4.83±0.26
Fenobucarb	Added	3.0	5.0	5.0	6.0
	Found	2.79±0.01	4.66±0.26	4.91±0.19	5.68±0.27

The detection limits have been calculated by means of different procedures. Hence, the Winefordner and Long[11] and the Clayton[12] methods have been applied as well as that based on the reproducibility of the analyte response at very low concentration.[13] The obtained values are in Table 3, and we can observe that those calculated by the Clayton method with specified assurance probabilities are slightly greater. The proposed procedure has been applied to river water samples, spiked with these compounds and the results are in Table 4.

CONCLUSIONS

In the analysis of N-methylcarbamates, methods of high performance liquid chromatography with electrochemical detection present advantages with respect to the UV or flourimetric detection, as high sensitivity, good reproducibility and easy chemical derivatization procedures. Hence, in the proposed method in which a coulometric detector is used, detection limits near 1 x 10^{-9} M are achieved, and only a previous step of alkaline hydrolysis is needed. This step is, however, simple and quick and does not require any non aqueous medium. Because of these characteristics, this procedure is recommended for controlling residues of these kinds of pesticides in natural waters.

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REFERENCES

1. M. Olek, F. Blanchard, G. Sudraud, J. Chromatogr., **325**, 239 (1985).

2. R. T. Krause, J. Chromatogr., **442**, 333 (1988).

3. W. J. Mayer, M. K. Greengerg, J. Chromatogr., **211**, 135 (1981).

4. M. B. Thomas, P. E. Sturrock, J. Chromatogr., **357**, 318 (1986).

5. S. Kaway, K. Goto, K. Kano, T. Kubota, J. Chromatogr., **442**, 451 (1988).

6. B. S. Seidel, W. Faubel, H. J. Ache, Kernforschungazent Karlsruhe KfK 5182, pp58, Berlin (1993).

7. P. T. Kissinger, K. Bratin, W. P.King, J. R. Rice, in **Pesticide Analytical Methodology**, J. Harvey Jr., G. Zweig eds, ACS Symposium Series n° 136, Washington, DC, pp 57-88 (1980).

8. K. D. Khalaf, A. Morales-Rubio, M. de la Guardia, Anal. Chim. Acta, **280**, 231 (1993).

9. A. Guiberteau, T. Galeano Díaz, F. Salinas, J .M. Ortiz, Anal. Chim. Acta, **305**, 219 (1995).

10. A. Guiberteau, T. Galeano Díaz, F. Salinas, J. M. Ortiz, J. M. Kauffmann, submitted to Electroanalysis.

11. J. D. Winefordner, G. L. Long, Anal. Chem., **55**, 712A (1983).

12. C. A. Clayton, J. W. Hines, P. D. Elkins, Anal. Chem., **59**, 2506 (1987).

13. J. A. Glaser, D. L. Foerst, G. D. Mckee, S. A. Quave, W. L. Budde, Environmental Science and Technology, **15**, 1427 (1981).

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