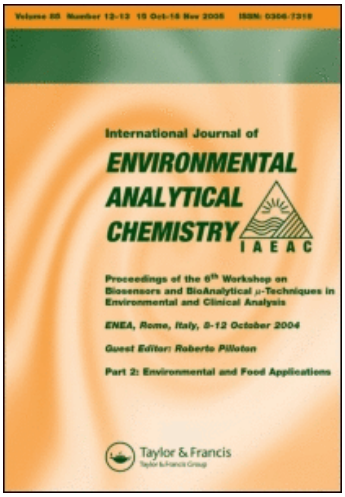

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Isodifferential Derivative Spectrophotometric Assay of the Carbamate Pesticide Carbaryl and its Metabolite in Biological Fluids and Commercial Formulations

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Isodifferential Derivative Spectrophotometric Assay of the Carbamate Pesticide Carbaryl and its Metabolite in Biological Fluids and Commercial Formulations

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Two types of applications (biological fluids and commercial formulations) have been used to demonstrate the advantages of UV derivative spectroscopy using the isodifferential assay for the quantitative simultaneous determination of the insecticide carbaryl and its metabolite 1-naphthol.

By a careful selection of the working wavelengths based on the graphical model to measure derivative amplitudes, it is possible to determine both compounds in the range of 0.2–40.0 μg/ml in presence of five and ten-fold concentration excess, respectively. Accurate and reproducible results are obtained with no preliminary separation step required.

KEY WORDS: Isodifferential derivative spectroscopy, carbaryl, 1-naphthol, metabolites, biological fluids.

1. INTRODUCTION

Carbaryl is a well-known carbamate insecticide widely used today to control a large number of insect pests. Because of its physical properties of volatility and slight solubility in water, there are two

main ways of contamination: (a) the environment (water supplies, rivers, lakes, well-waters, soils, etc...) and (b) animals and human beings exposed to it. The toxic effects are due to the inhibition caused in the cholinesterase enzyme and its teratogenic character.[1]

In basic media carbaryl is decomposed into 1-naphthol in water and soil. The metabolism of carbaryl *in vivo* consists of the same hydrolysis and it is excreted in the urine as 1-naphthol. Hence it is of great interest in agricultural and biomedical fields to quantify them at trace levels.

Among the numerous methods employed in the determination of carbaryl, spectrofluorimetry has become the method of choice. In most cases, due to the complexity of real samples or the lack of sensitivity, most of these methods require the coupling with a separation technique such as HPLC[2–5] or GC[6,7] or a derivatization step prior the determination.[8,9]

Direct analysis is always preferred to derivatization where possible because of the extra time and sample manipulation (as well as associated error) required to form a product.

Since strongly fluorescent derivatives are also strongly UV-absorbing compounds, the spectrophotometric detection have been widely used as an alternative to spectrofluorimetry in the analysis of carbaryl and its metabolite.[10–12]

As it has been recommended by the World Health Organization,[13] a simple and easy method, with applicability in any laboratory, is required for the quantitative determination of carbaryl and 1-naphthol as an alternative for the official colorimetric method actually in use.[14]

The work presented here competes favourably in simplicity, rapidity and detection limit with those proposed early (see Table I). The method was successfully applied to the simultaneous determination of carbaryl and 1-naphthol in commercial formulations and rat blood without prior separation by means of derivative spectroscopy, using a graphical model previously defined.[15] Its utility has been widely demonstrated.[16–19]

2. MATERIALS AND METHODS

Apparatus

Spectral measurements were made with a Shimadzu UV-240

Table I A review of methods for the quantitative determination of carbaryl

Method	Detection limit	Reference
Colorimetric	5.00 μg	20
TLC-colorimetric	0.02 μg/ml	21
Colorimetric	0.12 μg/ml	22
GC-fluorimetric	0.05 μg/ml	6
Fluorimetry	0.10 μg/ml	23
Fluorimetry	0.20 μg/ml	24
HPLC-photometry	1.00 ng	25
Colorimetry	0.20 μg/ml	26
GLC-flame ionization	50.00 μg/ml	27
GC-electron capture	0.05 μg/ml	28
GC-electron capture	25.00 ng/ml	29
HPLC-fluorimetric	26.00 ng	30
HPLC-fluorimetric	30.00 ng/ml	31
HPLC-fluorimetric	10.00 ng/ml	32
Fluorimetry	0.20 μg/ml	33
HPLC-photometric	0.10 μg/ml	34
HPLC-fluorimetric	0.05 μg/ml	35
GC-electron capture	0.50 μg/ml	36
HPLC-electrochemistry	3.80 ng/ml	37
Fluorimetry	4.00 μg/ml	38
HPLC-fluorimetry	40.00 ng/ml	2
Photometry	0.30 μg/ml	10
Fluorimetry	0.20 μg/ml	39
HPLC-photometric	0.10 mg/kg	11
HPLC-fluorimetric	0.02 μg/ml	4
GC-thermometric	5.00 ng/ml	7
LC-fluorimetric	—	9
Phosphorimetry	1.00 ng	40
LC-fluorimetric	0.05 μg/ml	41
Photometry	12.00 μg/ml	12
HPLC-fluorimetric	0.15 μg/ml	5
Photometry	0.06 μg/ml	this work

Graphicord recording spectrophotometer using quartz cells of 1 cm path length. The instrument parameters were: slit = 2 nm; scanning speed = 3 nm/s; and recording chart speed = 10 and 5 nm/cm.

Derivative spectra were recorded with a Shimadzu derivative spectrum attachment optional program/interface (OPI-2) model, with derivative order 1st to 4th and $\Delta\lambda$ values 1, 2 and 4 nm.

Reagents

Stock solutions of carbaryl and 1-naphthol (>99% purity, Riedel-de-Häen, AG Seelze, Hannover) were prepared in ethanol (Merck) at 2 g/l and 1.44 g/l respectively. A working standard solution of both compounds at 100 μg/ml were prepared by appropriate dilutions with ethanol.

Analytical procedures

Individual compounds Place an aliquot of the sample solution in ethanol containing 2–400 μg of carbaryl or 1-naphthol in a 10 ml volumetric flask. Add enough ethanol to obtain 5% v/v and dilute to volume with deionized water. Measure the absorbance at 279 nm or 292 nm respectively against a water blank. Record the 1st and 2nd derivative ($\Delta\lambda = 4$ nm) with 3 nm/s scanning speed. Measure the 1st derivative as the vertical distance in the dA scale from $\lambda_{max} = 287$ nm and $\lambda_{min} = 257$ nm and $\lambda_{max} = 320$ nm and $\lambda_{min} = 267$ nm for carbaryl and 1-naphthol, respectively and for 2nd derivative the vertical distance in the d^2A scale from $\lambda_{max} = 292$ nm and $\lambda_{min} = 282$ nm and $\lambda_{max} = 322$ nm and $\lambda_{min} = 315$ nm, for carbaryl/1-naphthol.

Binary mixtures Prepare a series (between 3–5 solutions) of each compound containing carbaryl and 1-naphthol between 0.2–40 μg/ml. Samples were prepared as described for individual compounds. Measure the 1st derivative value as the vertical distance in the dA scale from peak to base-line at 284 nm and 320 nm for carbaryl and 1-naphthol, respectively. Measure the 2nd derivative value as the vertical distance from trough to base-line at 293 nm and 321 nm for carbaryl and 1-naphthol, respectively.

Sample preparation

Commercial formulations Prepare vacuum filtration assembly consisting of 250 ml filter flask, funnel holder, and 30 ml medium porosity fritted glass Büchner funnel. Add accurately weighed sample containing 0.04 ± 0.01 g active carbaryl to Büchner funnel. Add 10 ml methanol to funnel. Do not apply vacuum. Let methanol stand in the funnel for 5 minutes. Apply vacuum until all liquid is in filter flask. Add another 10 ml to funnel and repeat the operation two

more times. Release vacuum and disassemble apparatus. Quantitatively transfer contents of filter flask to 50 ml volumetric flask. Dilute with additional methanol. Stopper and mix well by inverting several times. The final concentration was 800 μg/ml carbaryl. Aliquots of this solution were used for the analytical determination.

Blood samples The animal employed was a white rat Wistar of approximate weight 260 g which was anesthetized with sodium penthobarbital (50 mg). The blood extraction from the animal is done with heart punction in left ventricle with heparinized syringe (lithium heparine). The blood obtained was kept in the refrigerator until used (4°C).

To blood samples (0.25 ml) add different amounts of standard ethanolic solution of carbaryl (200 μg/ml) and/or 1-naphthol (200 μg/ml). The centrifuge tubes containing the blood samples and insecticide were placed in an ultrasonic bath (Ultrasons, Selecta®) for 20 seconds for the hemolisis to take place. Add 2.5 ml of ethyl acetate and shake them for 10 minutes. Centrifuge it at 3.500 r.p.m. and transfer 2 ml of supernatant liquid to a round bottom flask and take it to dryness in vacuum (40°C). Dilute the dry extract to 10 ml with deionized water. This solution was treated in the same manner as indicated under "analytical procedure".

3. RESULTS AND DISCUSSION

Spectrophotometric conditions and instrumental variables

The individual absorption bands of carbaryl and 1-naphthol in a 5% (v/v) ethanol:water mixture are compared in Figure 1 to the UV absorption spectrum of their mixture. Carbaryl and 1-naphthol could be readily determined individually by measuring their absorbance at 279 nm and 292 nm, respectively. The severe overlap of both absorption spectra due to the broad shape and close absorption maxima, precludes the simultaneous determination of both compounds using, exclusively, these zero order UV spectra.

The absorption spectra of carbaryl was studied in eleven different solvents to evaluate the solute-solvent interaction. No significant changes in absorbance or absorption maxima were observed, so we

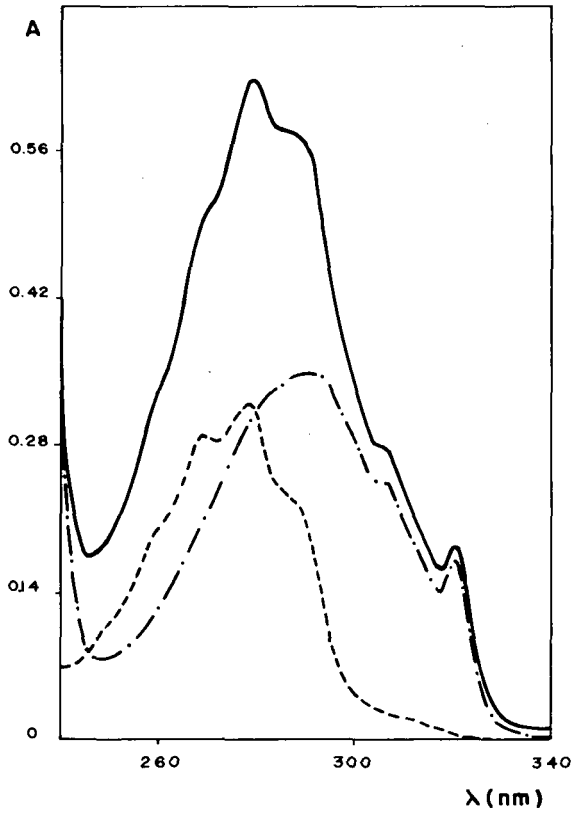

Figure 1 Normal absorption spectra of carbaryl (----), 1-naphthol (-.-.-) and their mixture (———) at 10 μg/ml in 5% (v/v) ethanol:water solution.

selected water as the more desirable solvent for carbaryl and 1-naphthol measurements.[41] The insolubility of the compounds in aqueous solution is avoided by dissolving the standards in ethanol and working with a fixed ethanol percentage of 5% throughout the experimental work.

The effect of day light and temperature onto the absorption maxima of both compounds was studied with no changes occurring during a minimum of 2 hours the samples exposed to light and measured at 25 ± 0.5°C.

The main instrumental parameters affecting the shape of the derivative spectra are the wavelength scanning speed (V_{scan}), wave-

length range over which the derivative is averaged ($\Delta\lambda$), response time (t_r), and the derivative order. In general, high V_{scan} and $\Delta\lambda$ give good amplitudes but small resolution, while t_r gives small amplitudes but good S/N ratios. However, if $\Delta\lambda$ is greater than the half-band width of the absorption spectra, the resolution of the derivative curves will be decreased.

In the present work, a medium V_{scan} (3 nm/s), $\Delta\lambda$ (4 nm) and 1st and 2nd derivative order are employed. However, using the graphical model described for the simultaneous determination of carbaryl and 1-naphthol, the parameter selection is not critical.

Quantitative analysis by UV and UV derivative spectroscopy

In principle, any binary mixture of compounds with different absorption maxima, such as this insecticide and its metabolite, may be directly analyzed by applying Beer's law to each component, independently of the other, and taking into account the possible cross-contributions of one absorption band to another.

As it is seen in Figure 1, due to the overlap between the two absorption bands of the compounds, the absorption spectrum of their mixture does not allow for useful measurements in that zone. In such a case, the application of derivatives gives additional possibilities to increase analytical selectivity, due to the fact that derivatives permit a division of the zero order UV spectra in different zones.

Figure 2 shows the 1st and 2nd derivative spectra of the compounds in a series of different concentrations, where the overlapping of zero order spectra seems to be resolved allowing a direct determination of carbaryl in presence of 1-naphthol and vice versa.

Various graphical measurements would be available for the amplitude of the 1st and 2nd derivative of the mixture. To select the proper wavelength maxima at which this quantitative determination could be carried out, the two series of spectra were minuciously analyzed. It is observed from Figure 2a that there is an isodifferential point in the carbaryl/1-naphthol series corresponding to the absorption maxima of the zero order spectra at $\lambda = 279$nm and $\lambda = 292$ nm, respectively.

These wavelengths are of analytical interest because carbaryl could

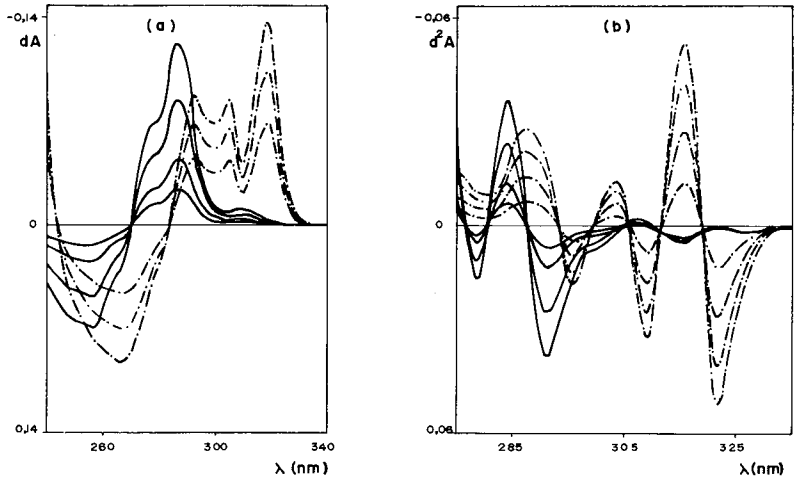

Figure 2 First (a) and second (b) derivative spectra of 5% (v/v) ethanol:water solutions of carbaryl (———) 15, 10, 5, 2.5 μg/ml and 1-naphthol (–.–.–) 20, 15, 10, 5 μg/ml.

be readily determined by measuring its absorbance at the isodifferential point of 1-naphthol, where its absorbance is negligible, while 1-naphthol could be determined by measuring the corresponding absorbance at the isodifferential point of carbaryl. Due to the fact that no contribution of carbaryl concentration is observed between 300–340 nm (see Figure 1), the naphthol would be measured at 320 nm instead of at its isodifferential point of 279 nm.

The graphical model system used for measuring these two compounds in a binary mixture is indicated in Figure 3a. The same reasoning was carried out with the 2nd derivative in Figure 2b, and the wavelength maxima for measuring carbaryl ($\lambda = 293$ nm) and 1-naphthol ($\lambda = 320$ nm), are indicated in Figure 3b.

In a broad sense, optimal conditions would be obtained when the 1st derivative amplitude of carbaryl is zero at the same wavelength in which the derivative of 1-naphthol is maximal. Assuming that Beer's law is obeyed in the concentration rate studied and that the bands of the mixture could be reduced to a Gaussian spectral shape, the following equations can be expressed for 1st (1) and 2nd (2)

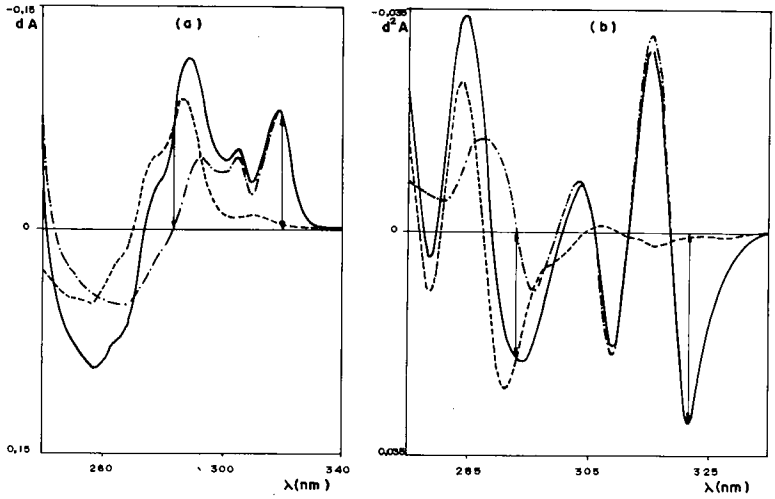

Figure 3 First (a) and second (b) derivative spectra of carbaryl (----), 1-naphthol (-.-.-) and their mixture (——) at 10 μg/ml in 5% (v/v) ethanol:water solutions. Arrows indicate graphical measurements based on isodifferential points.

derivative:

$$\frac{dI_1(x)}{dx} = \frac{-A_1(x-C_1)}{B_1^2} \exp\left|\frac{-(x-C_1)^2}{2B_1^2}\right| \tag{1}$$

$$\frac{d^2I_2(x)}{dx^2} = \frac{A_2}{B_2^2}\left|\frac{(x-C_2)^2}{B_2^2} - 1\right| \exp\left|\frac{-(x-C_2)^2}{2B_2^2}\right| \tag{2}$$

where A is the maximum amplitude, B is the band half-width and C is the abscissa value corresponding to the function maximum. The solutions for $d^2I_2(x)/dx^2 = 0$ and for $dI_1(x)/dx = 0$ are satisfied when $C_1 = x$ and $(x-C_2)/B_2 = 1$, e.g., when $C_1 - C_2 = B_2$. This implies that the best conditions for evaluation amplitude values of component 2 from the zero crossing of component 1 are obtained when band maximum separation $(C_1 - C_2)$ is equal to band half-width of compound 2 (B_2). Inversely, when $C_1 - C_2$ is equal to B_1, the evaluation of compound 1 present optimal conditions.

Based on this graphical model for quantitative individual and mixture analysis by derivative spectrophotometry, linear relation-

ships between absorbance A, dA and d^2A and concentration were established.

The calibration curves obtained with carbaryl and 1-naphthol solutions are linear between 0.2–40.0 μg/ml. From these analytical data, and applying linear regression analysis, the following equations would be applicable:

— CARBARYL: $A = 0.030|\text{CA}| - 0.005 \quad r = 0.9996$ (2)

$dA = 0.013|\text{CA}| + 0.001 \quad r = 0.9999$ (3)

$d^2A = 0.005|\text{CA}| + 0 \quad r = 0.9999$ (4)

— 1-NAPHTHOL: $A = 0.033|\text{NA}| + 0.004 \quad r = 0.9984$ (5)

$dA = 0.012|\text{NA}| + 0.003 \quad r = 0.9985$ (6)

$d^2A = 0.006|\text{NA}| + 0.001 \quad r = 0.9983$ (7)

The application of the graphical measurements indicated in Figure 3 for a general situation with the binary mixtures of carbaryl/1-naphthol with these specific wavelengths to the linear calibration graphs between 0.2–40.0 μg/ml give the following equations:

— CARBARYL: $dA = 0.007|\text{CA}| + 0.001 \quad r = 0.9999$ (8)

$d^2A = 0.002|\text{CA}| + 0.0003 \quad r = 0.9998$ (9)

— 1-NAPHTHOL: $dA = 0.007|\text{NA}| + 0.002 \quad r = 0.9980$ (10)

$d^2A = 0.003|\text{NA}| + 0.0002 \quad r = 0.9984$ (11)

It is observed that the slope of the derivatives for the mixture analysis is approximately half of the slope in the calibration curves with the individual compounds. This is due to the fact that in the last case, measurements are taken between a peak and a trough while in the first case, and to avoid the interference from one another, the measurements are taken between peak and base-line or trough and base-line.

The limit of detection, c_L, determination, c_Q (the lower limit of the calibration curve) defined by the IUPAC[42] as well as the sensitivity[43] of the individual analysis are presented in Table II.

The statistical analysis to evaluate the accuracy and precision of the different methods was studied using seven samples of carbaryl and 1-naphthol at two different points of the calibration curve. The mean concentration found, the standard error and the relative standard deviation R.S.D., are also given in Table II.

Application of the methods to synthetic mixtures

Among the equations given above, the ones defined for the individual analysis are a particular case of the ones for the mixture analysis and are only defined to characterize the method. So, for any real case, only Eqs. (8) to (11) would be applied.

To prove the applicability of the methods, these Eqs (8) to (11) have been applied to the determination of both compounds in synthetic binary mixtures with different concentration ratios. The results are indicated in Table III.

The tolerance criterion used has been a deviation of five times the standard deviation, s_s, of the mean value, $\bar{x}$, of seven measurements of carbaryl or 1-naphthol alone at 2 μg/ml. With this uniform criterion and looking at Table III, carbaryl can be quantitatively determined in presence of a five concentration excess of 1-naphthol using 1st derivative while 2nd derivative permits only a two-fold excess. In the manner, at the level tested of 2 μg/ml of 1-naphthol, carbaryl can be present in a ten-fold excess with the 1st derivative measurements and at seven-fold excess with the 2nd derivative.

These tolerance ratios are quite superior to those previously published by Larkin and Day[33] where it is possible to measure carbaryl in the range 0–1 μg/ml in presence of 1-naphthol up to 10 μg/ml and 1-naphthol in the same range up to 4 μg/ml of carbaryl. On the other hand, DeBerardinis and Wargin[2] by means of HPLC with fluorescence detection could determine carbaryl in a 5-fold excess of 1-naphthol and the latter with a 4-fold excess of carbaryl.

The zero order absorptiometric measurements were also applied to the synthetic mixtures shown in Table III but inadmissible absorbance values were obtained when measuring at $\lambda = 279$ nm and $\lambda = 292$ nm, for carbaryl and 1-naphthol, respectively. This indicated that the

Table II Analytical data of the different methods

Compound	Spectrum	s_A |μg/ml|	$c_L(k=3)$ |μg/ml|	Dynamic range |μg/ml|	Amt. taken |μg/ml|	Amt. found |μg/ml|	R.S.D. |%|	Error |%|
CARBARYL	Normal	1.5	0.2	0.7–40.0	1.0	1.3	3.7	3.3
	1st deriv.	2.2	0.07	0.2–40.0	1.0	0.9	3.2	2.8
	2nd deriv.	7.6	0.06	0.3–40.0	1.0	1.0	3.7	3.3
1-NAPHTHOL	Normal	6.4	0.09	0.3–40.0	1.0	1.0	19.8	17.7
	1st deriv.	2.7	0.02	0.2–40.0	1.0	0.7	4.5	4.0
	2nd deriv.	7.5	0.15	0.5–40.0	1.0	0.9	4.8	4.3

Table III Tolerance ratio in the quantitation of carbaryl/1-naphthol in binary mixtures

Mixtures[a]	Ratio CA:NA (w/w)	1st derivative		2nd derivative	
		Carbaryl[b]	1-naphthol[b]	Carbaryl[b]	1-naphthol
CA(2)+NA(2)	1:1	2.00±0.08	1.64±0.07	2.10±0.07	1.85±0.05
CA(2)+NA(4)	1:2	2.00±0.14		2.30±0.21	
CA(4)+NA(2)	2:1		1.58±0.06		1.82±0.04
CA(2)+NA(10)	1:5	2.30±0.11		2.70±0.19	
CA(10)+NA(2)	5:1		2.15±0.08		1.89±0.04
CA(2)+NA(14)	1:7	3.90±0.10		7.50±0.22	
CA(14)+NA(2)	7:1		1.90±0.10		2.24±0.08
CA(2)+NA(20)	1:10	4.40±0.17		10.90±0.21	
CA(20)+NA(2)	10:1		2.16±0.10		2.31±0.09
CA(40)+NA(2)	20:1		2.90±0.09		2.73±0.10

[a]In parenthesis, concentration in μg/ml.
[b]Concentration found ±S.D. in μg/ml.

direct technique could not be applied to the analysis of binary mixtures of these two compounds.

Accuracy and precision of the simultaneous quantitative determination of carbaryl and 1-naphthol by the proposed method were evaluated by analyzing a series of seven binary mixtures containing different concentrations of carbaryl/1-naphthol. The results are indicated in Table IV where it is evident that the 2nd derivative is more accurate and precise than the 1st derivative but less sensitive. This is also indicated in Table III where the 1st derivative permits higher tolerance ratios than the 2nd derivative.

Determination of carbaryl and/or 1-naphthol in commercial formulations and whole rat blood

The insecticide carbaryl (Sevin®) is widely used in Spain as a single component and as a component in mixed formulations, with percentages between 0.5–85.0%. The analysis of these formulations presents some problems due to the fact that all types of them cannot be extracted with chloroform. For such a reason, the applicability of the official method[14] is restricted because it uses chloroform as extracting solvent.

Table IV Accuracy and precision of the UV derivative mixture measurements

Mixture[a]		1st derivative		2nd derivative	
		Carbaryl	1-naphthol	Carbaryl	1-naphthol
CA(2)+NA(10)	$\bar{x}$	2.95	10.23	2.87	10.63
	s	0.11	0.40	0.08	0.20
	R.S.D.	3.87	3.93	2.33	1.77
	Error	3.46	3.51	2.08	1.58
CA(10)+NA(2)	$\bar{x}$	9.73	1.79	10.27	2.24
	s	0.18	0.08	0.21	0.04
	R.S.D.	1.89	4.38	2.04	1.81
	Error	1.69	3.92	1.82	1.62

[a]In parenthesis, concentration in μg/ml.
$\bar{x}$ and s, in μg/ml; R.S.D. and error, in %.

The extraction system used in this method is similar to that studied by McDermott[44] employing 100% methanol instead of chloroform, a more hazardous solvent. This extraction procedure is suitable for three wettable powders—commercial formulations in current use in Spain containing carbaryl. The label declarations and the results obtained when the 3 samples were analyzed for carbaryl and 1-naphthol by the proposed method are shown in Table V.

The method included a test to establish whether the observed absorbance is the result of unreacted 1-naphthol in the formulation or of 1-naphthol formed by decomposition of carbaryl during storage of the formulation.

From the data indicated in Table V it is deduced that normal absorptiometric measurements give positive recoveries while 1st and 2nd derivative result in percentages of carbaryl which are close to label declarations.

The accuracy and precision in terms of relative standard deviation after 2 extractions and 2 determinations compares favourably with other methods previously established.[6,27,44] The 1-naphthol analyzed can be only observed by the 1st derivative due to the low concentration detected, increasing as the carbaryl percentage increases.

Table V Direct analysis of carbaryl and 1-naphthol in commercial formulations

Formulation No.[a]	Spectrum	Carbaryl		1-naphthol
		%±R.S.D.[b]	Recovery, %	%±R.S.D.[b]
1 (5%)	Normal	6.14±0.44	122.8	—
	1st derivative	4.29±0.18	85.8	0.34±0.06
	2nd derivative	4.73±0.22	94.6	—
2 (7.5%)	Normal	9.07±0.75	120.9	—
	1st derivative	6.51±0.40	86.8	0.41±0.05
	2nd derivative	6.90±0.32	92.0	—
3 (85%)	Normal	109.89±7.93	129.3	—
	1st derivative	77.55±2.01	91.2	2.77±0.73
	2nd derivative	81.70±2.88	96.1	—

[a]Label declaration: Formulation 1-carbaryl 5% (Patatol AC); formulation 2-carbaryl 7.5% (Agres S-7.5); formulation 3-carbaryl 85% (Sevin 85).
[b]Mean value of 2 extractions and 2 determinations.

The anticholinesterase effect of carbaryl in human organism give importance to the establishment of agricultural and professional exposure limits to this insecticide in Spain, as they have been indicated for other countries.[4,5]

Since the extrahepatic metabolism of carbaryl *in vivo*, in large part, consists of the enzymatic and chemical hydrolysis of carbaryl to 1-naphthol, it is of great interest to quantitate the levels of each.

For such a reason, the applicability of the proposed method has been tested by the determination of carbaryl and 1-naphthol in spiked whole rat blood samples by recovery assays. The results obtained are indicated in Table VI together with the relative standard deviation of 3 individual extractions and 2 determinations, by 1st and 2nd derivative UV spectroscopy.

First derivative assays give better results for 1-naphthol and 2nd derivative for carbaryl, with recovery percentages between 70–135%. These values demonstrate the applicability of the proposed method for the determination of this insecticide and its metabolite 1-naphthol in two types of residue analysis at concentration levels relevant to this type of analysis.

Table VI Direct analysis of carbaryl and 1-naphthol in blood samples

Concentration added[a] g/ml	Spectrum	Concentration found, g/ml		Recovery, %	
		Carbaryl[b]	1-naphthol[b]	Carbaryl	1-naphthol
CARBARYL (2)	1st deriv.	2.35±0.31	—	117.5	—
	2nd deriv.	1.97±0.18	—	98.5	—
CARBARYL (1)	1st deriv.	0.98±0.18	—	98.0	—
	2nd deriv.	0.75±0.21	—	75.0	—
CARBARYL (2)+NAPHTHOL (2)	1st deriv.	2.70±0	1.71±0	135.0	85.5
	2nd deriv.	1.85±0	1.18±0.12	92.5	59.0
CARBARYL (2)+NAPHTHOL (1)	1st deriv.	2.70±0	0.85±0.20	135.0	85.0
	2nd deriv.	1.75±0.14	0.70±0.14	87.5	70.0

[a]In parenthesis, concentration of insecticides added to whole blood.
[b]Mean values of 3 extractions and 2 determinations.

4. CONCLUSIONS

The simultaneous determination and subsequent analysis of carbaryl insecticide and its metabolite 1-naphthol in whole blood and commercial formulations by UV derivative spectroscopy can be achieved. The described assay offers several advantages over other methods of analysis. Colorimetric and GLC-electron capture detection assays require conversion of all carbaryl to 1-naphthol prior to complexation or derivatization so that the relative contributions of each are unknown. Also, direct UV determination measures carbaryl plus 1-naphthol and, again, individual concentrations cannot be assessed.

By applying derivative spectroscopy with a correct wavelength selection (isodifferential model) for the determination of carbaryl and 1-naphthol it is shown that specific, rapid and direct measurements with at least the same detection limits and accuracy as in other methods are possible, without prior separation or ulterior complexation or derivatization step.

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