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Determination of the fungicides folpet, captan and captafol by cloud-point preconcentration and high-performance liquid chromatography with electrochemical detection

R. Carabias Martínez*, E. Rodríguez Gonzalo, M[a].G. García Jiménez[1], C. García Pinto, J.L. Pérez Pavón, J. Hernández Méndez

Departamento de Química Analítica, Nutrición y Bromatología, Universidad de Salamanca, 37008 Salamanca, Spain

Abstract

A method is described for the determination of residues of the fungicides Folpet, Captan and Captafol in river water using an isocratic high-performance liquid chromatographic system. Electrochemical detection with single and dual glassy-carbon electrodes was evaluated for possible amperometric detection of these fungicides; the reductive–oxidative detection mode with a dual electrode in the series configuration proved to be more appropriate than direct reductive detection with a single working electrode. To concentrate the fungicide residues, a cloud-point preconcentration step employing the non-ionic surfactant Triton X-114 was applied. Furthermore, the presence of the surfactant contributes to the stability of these analytes in aqueous solution, avoiding their hydrolysis. The method described shows good recoveries and good precision, with relative standard deviations of less than 10%. Detection limits of about 4 μg/l were achieved for all three fungicides studied for 15 ml of preconcentrated sample.

Keywords: Water analysis; Environmental analysis; Cloud point preconcentration; Sample preparation; Folpet; Captan; Captafol; Pesticides

1. Introduction

Folpet, Captan and Captafol are protective, non-systemic fungicides used for the treatment of foliar, soil-borne and seed-borne diseases. All three fungicides are widely used on fruit, vegetables and ornamental plants. As a result of their widespread use, fungicide residues can contaminate crops, wells and streams, due to spills, spraying or run-off.

The early colorimetric procedures [1,2] for the determination of Captan, Folpet and Captafol at the residue level have been largely replaced by methods involving the use of thin-layer chromatography [3,4] or gas–liquid chromatography [5,6]. Simultaneous determination by gas chromatography with electron-capture detection (GC–ECD) of Captan, Folpet and Captafol in various crops has been described by Pomerantz et al. [7] and Baker and Flaherty [8]. While all three fungicides are readily amenable to gas chromatographic–ECD, Captan and Folpet are difficult to resolve using methyl silicone stationary phases in conventional packed columns [9,10] routinely used for pesticide residue analysis. On-

*Corresponding author.

[1]Permanent address: Instituto de Investigaciones Científicas, Universidad de Guanajuato, Gto, L. Retana no 5, 36000 Mexico.

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column degradation of Captan and Folpet has been observed on OV-101 columns regularly used in the pesticide laboratory for multiresidue screening purposes. Gilvydis and Walters [11] reported that the phthalimide fungicides Captan, Folpet and Captafol are notoriously prone to adsorption and degradation in conventional packed columns because of the chemical activity of the support material; such adverse effects were less notable with wide-bore, open-tubular (WBOT) fused-silica columns. Degradation of Captan caused by column deterioration was earlier reported by Carlstrom [12].

Because of the possibility of decomposition of Captan and Folpet during GLC, several methods employing liquid chromatography have been developed for the analysis of these compounds in formulations [13–15], crops [16] and environmental waters [17]. Most of the reported HPLC methods use conventional or diode array UV detection, although separation and determination of the three fungicides by using photoconductivity [18] and mass spectrometry [17] detectors coupled to the liquid chromatographic system have also been reported.

Since the concentration levels of interest for environmental analysis are very low, a preconcentration step is needed. Usually, isolation and trace enrichment of pesticide samples are carried out by liquid–liquid extraction (LLE) [19,20] or solid-phase extraction (SPE) [21–23]. The latter technique is less laborious and time-consuming than LLE and eliminates large volumes of potentially hazardous solvents.

Liquid chromatographic methods combined with off-line [24–26] and on-line SPE [27–30] can be readily found for numerous pesticides, C_{18} bonded silica being the preferred sorbent. Occasionally, low recoveries of organic species have been described in the preconcentration of real environmental water samples [31,32]; these lower recoveries may be caused by humic substances present in real river waters that are also trapped by sorptive sites or to the formation of pesticide–humic complexes that could not be sufficiently extracted by the C_{18} bonded silica phase. Salau et al. [17] reported low recoveries for Captan, Folpet and Captafol when isolated from river water samples by using a C_{18} SPE disk; this may be attributed to hydrolysis of the fungicides in the disk due to the remaining water not being removed [33]. It has been described [34,35] that residual water surrounding the nonpolar matrix of the disks and cartridges after filtration of pesticides may cause hydrolysis of susceptible compounds.

The use of preconcentration steps based on phase separation by the cloud point technique [36,37] offers a convenient alternative to more conventional extraction systems. The small volume of the surfactant-rich phase obtained with this methodology permits the design of extraction schemes that are simple, cheap and of lower toxicity than extraction with organic solvents and that have results similar to those obtained by other separation techniques. This topic has been reviewed by Hinze and Pramauro [38]. The cloud point methodology can be used to separate and/or preconcentrate different analytes as a step prior to their determination in hydrodynamic analytical systems such as HPLC [39–41] or FIA [42].

Nevertheless, one of the limitations of the cloud point methodology is the high absorbance shown by many surfactants in the UV region, rendering this methodology useless when optical detection is used. Electrochemical detection overcomes this problem due to the electrodic inactivity of several commercially available surfactants such as Triton X-114.

The present work evaluates the efficacy of liquid chromatography with electrochemical detection using a dual glassy-carbon electrode for the determination of the fungicides Captan, Folpet and Captafol. Although direct amperometric detection of these fungicides can be performed due to the presence of a carbonyl group in their structure, the use of detection systems based on dual electrodes could be advantageous.

A procedure based on cloud point preconcentration with the non-ionic surfactant Triton X-114 has been developed for the isolation and trace enrichment of the fungicides in water samples. In addition to the extraction and preconcentration of the fungicides, the presence of Triton X-114 aids in the storage of aqueous environmental samples prior to the extraction step by preventing the loss of these fungicides because of its hydrolysis in aqueous medium. The proposed method was applied to the determination of the above-mentioned fungicides in water samples from the river Tormes (Salamanca, Spain).

2. Experimental

2.1. Apparatus

The HPLC system used was a modular component system consisting of a Spectra-Physics Model SP-8800 ternary pump, an SP 8450 UV–Vis detector, and an SP 4290 integrator. Electrochemical detection was carried out in an EG&G PARC 400 electrochemical detector. The electrodes were as follow: Ag/AgCl/0.1 *M* KCl reference electrode, a gold auxiliary electrode and a single MP 1305 glassy-carbon electrode or an MP 1304 dual glassy-carbon electrode. When both optical and electrochemical detection were applied, the two detectors were connected in series, the electrochemical being the last one in the series.

In all experiments, a Rheodyne 7125 injection valve with a 10 μl sample loop and a Spheri-5-RP 18 column, 250×4.6 mm, 5 μm (from Brownlee Labs) were used.

A Kokusan H-103 N centrifuge was used for the separation of the two phases obtained with the cloud point methodology.

2.2. Reagents

The fungicides Folpet, N-(trichloromethylthio)phthalimide; Captan, N-(trichloromethylthio)-4-cyclohexene-1,2-dicarboximide and Captafol, N-(1,1,2,2-tetrachloroethylthio)-4-cyclohexene-1,2-dicarboximide were obtained from Scharlau.

The non-ionic surfactant Triton X-114 was obtained from Fluka and used without further purification.

Standard solutions were prepared in HPLC-grade acetonitrile (Carlo Erba, Milan, Italy). All other chemicals were of analytical-reagent grade. Ultra-high-quality water obtained with an Elgastat UHQ water purification system was used.

All solvents and samples were filtered through 0.45 μm pore-size nylon membrane filters (Millipore).

2.3. Standards and samples

A stock standard solution containing a mixture of the three fungicides was prepared by dissolving 100 mg/l of each analyte in pure acetonitrile. This solution was stored at 4°C and further diluted to obtain working standard solutions.

River water samples were taken from the river Tormes at different points in the province of Salamanca (Spain). At the time of collection, Triton X-114 was added to the samples (up to 0.25%). Samples were kept under refrigeration at 4°C and analysed within 24 h of collection.

2.4. Procedure

2.4.1. HPLC operating conditions

The mobile phase was a mixture of acetonitrile–water (45:55, v/v) containing 0.01 *M* acetic acid–acetate buffer (pH 4.6) as the supporting electrolyte. The mobile phase was degassed by bubbling helium through it. The flow-rate was set at 1.35 ml min^{-1} and the sample volume injected for analysis was 10 μl. Quantification was carried out using the external standards method and taking the mean peak height value of three injections. UV detection was carried out at 220 nm.

2.4.2. Electrochemical detection and electrode pretreatment

Electrochemical detection of fungicides using a dual glassy-carbon electrode in the series configuration (reductive–oxidative mode) was as follows: Fungicides were reduced at the upstream electrode (W_1) set at a potential of $E_1 = -1400$ mV, the reduction products being detected in the oxidation mode at the downstream electrode (W_2) set at $E_2 =$ 800–900 mV, the intensity at W_2 being the analytical signal. Both electrodes were electrochemically pretreated every day as follows: W_1 was kept at −1500 mV for 10 min and for another 10 min at +1600 mV; after this, it was kept at −1500 mV for 10 min and, finally, it was set at the working potential (−1400 mV). Meanwhile, electrode W_2 was set at +1000 mV for 10 min and for another 10 min at −1000 mV; it was then kept at +1000 mV for 10 min and, finally, it was set at the measuring potential (800–900 mV)

2.4.3. Cloud point preconcentration

Samples containing 15.0 ml of cold solutions containing the analytes in 0.25% Triton X-114 were kept for 5 min in a thermostatic bath at 40°C.

Separation of the two phases was achieved by centrifugation for 5 min at 3500 rpm. From the surfactant-rich phase, 60 μl were collected in a Hamilton syringe and 10 μl aliquots of this solution were injected into the chromatographic system.

Between one injection and the next, the column was washed with 100% acetonitrile for 10 min. During this washing step, a potential of 0.0 mV was applied to the working electrode (W_1), while the measuring electrode (W_2) was kept at its potential; no electrode pretreatment was required between runs.

2.4.4. Determination of Folpet, Captan and Captafol in water samples

Before their analysis, river water samples were filtered through sintered glass filters (n° 5) to remove suspended particulate matter. Fungicides were added to water samples by placing 1 ml of standard solution in a volumetric flask and adjusting the volume to 100 ml with the water sample. Samples (15 ml) were cloud-point preconcentrated with 0.25% Triton X-114.

Quantification was carried out using the external standard method, employing river water samples spiked with known amounts of the fungicides and taking the mean peak height value of three injections.

3. Results and discussion

3.1. HPLC–UV detection

Using a reversed-phase C_{18} column with an acetonitrile–water (45:55, v/v) mobile phase containing 0.01 *M* acetic–acetate buffer at a flow-rate of 1.35 ml min^{-1} was found to give good resolution and a reasonable analysis time. The sensitivity obtained when using spectrophotometric detection was very different for the three fungicides, being useful for Folpet detection but insufficient for the detection of Captan and Captafol. Folpet showed maximum absorption at a wavelength of 220 nm, with a molar absorptivity coefficient of $5.4 \cdot 10^4$ l mol^{-1} cm^{-1}, whereas Captan and Captafol show absorption maxima at lower wavelengths (210 and 200 nm, respectively) with molar absorptivity coefficients that were ten-fold lower.

3.2. Single electrode detection

All three fungicides are reduced on a single glassy-carbon electrode, showing a cathodic wave due to the reduction of the carbonyl group (Fig. 1). Since the reduction process is due to the same electroactive group in all three compounds, the sensitivity of the method for all of them is similar.

The high negative potential (−1400 mV) of reduction of the fungicide Captafol requires exhaustive deoxygenation because of the high residual current due to the reduction of dissolved oxygen. Even so, the noise level at high negative potentials becomes very high, making this detection unsuitable for trace determinations.

3.3. Dual electrode detection

In order to overcome the limitations of direct reductive electrochemical detection on a single working electrode, a dual glassy-carbon electrode in the series configuration (reductive–oxidative mode) was also assayed. The upstream electrode operates as a "generator" electrode to convert the analyte into its reduced form which will be detected in the oxidative mode at the downstream "detector" electrode.

The hydrodynamic voltammograms of the three fungicidal compounds were obtained by modifying the potentials of the upstream (W_1) and downstream (W_2) electrodes. Optimum potential values of $E_1 = -1400$ mV and $E_2 = 800$–900 mV for the upstream (W_1) and downstream (W_2) electrodes, respectively, were therefore chosen for further experiments. Under these conditions, the sensitivity obtained for the three fungicides is suitable and is better than that obtained by spectrophotometric detection for the fungicides Captan and Captafol (Fig. 2).

As is well known, solid electrodes are subject to surface fouling, resulting in decreased sensitivity. Such fouling may be due to adsorption of the analyte itself or of the electrodic reaction products. To improve detector perfomance, different kinds of electrochemical pretreatments were tested, the most efficient being the application of several potential pulses to both electrodes (Fig. 3). Daily application of this pretreatment afforded reproducible behaviour in the detection system. However, since the loss of electrodic activity is intimately linked to the con-

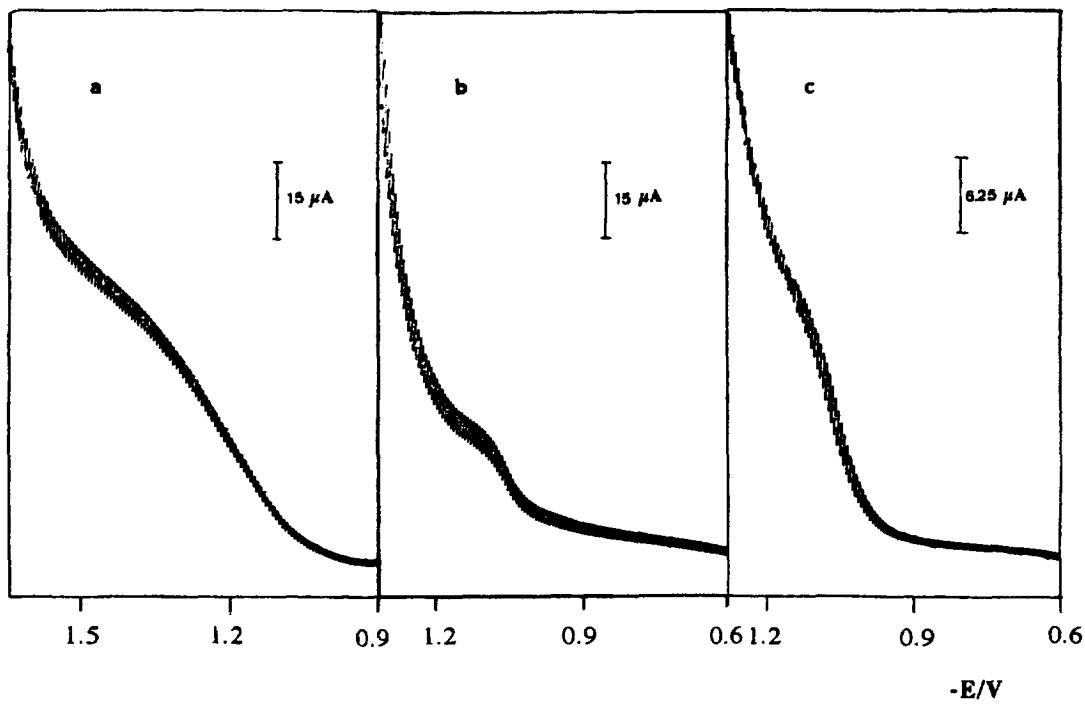

Fig. 1. Voltammograms obtained using acetonitrile–water (45:55, v/v) containing 0.1 *M* acetic-acid–acetate buffer for (a) Captafol, (b) Captan and (c) Folpet.

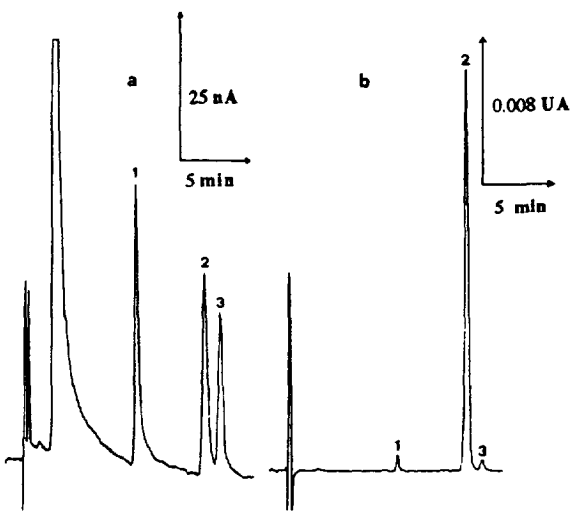

Fig. 2. Chromatograms obtained using (a) dual-electrode detection and (b) UV detection for (1) Captan, (2) Folpet and (3) Captafol. Chromatographic conditions are as described in Section 2.

centration injected, the chromatographic signal was observed to decrease gradually over time when the concentration of fungicide injected was of the order of 4 mg/l. No appreciable decrease in the chromatographic signal was seen for fungicide concentrations below 2 mg/l (Table 1).

3.3.1. Analytical data without preconcentration

Once the most suitable potential values for the detection of these fungicides had been determined, a study of the effect of the concentration of each fungicide on the peak height was carried out. The results obtained showed that for all three compounds the relationship between peak height and fungicide concentration was linear for concentrations below $2 \cdot 10^{-5}$ *M* (approximately 2.5 mg/l) (Table 1).

These findings show that electrochemical detection with a dual glassy-carbon electrode, in the reductive-oxidative mode, is much more favourable than spectrophotometric detection for the fungicides Captan and Captafol. The ratios between the slope obtained in the electrochemical calibration fits (s_{EC})

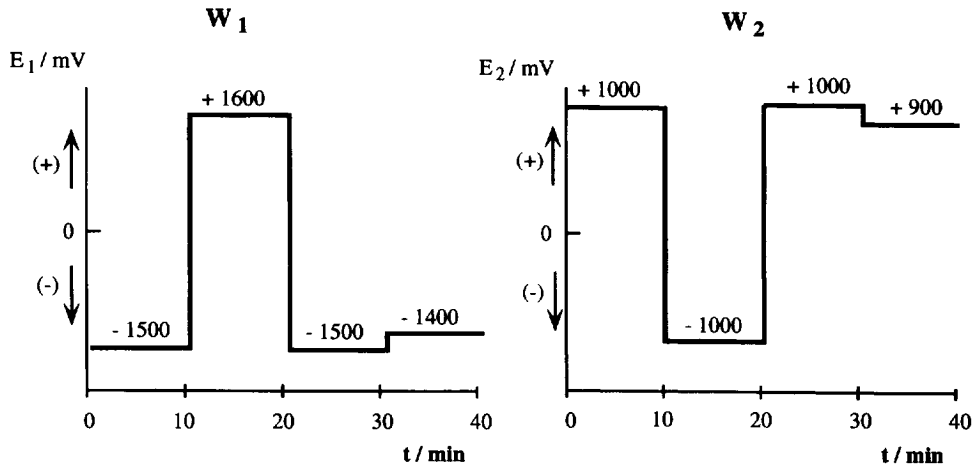

Fig. 3. Electrochemical pretreatment for dual-electrode detection in series. Applied potential versus time for both electrodes (W_1 and W_2).

and those obtained spectrophotometrically (s_{UV}) were 480 and 320 for Captan and Captafol, respectively. In the case of the compound Folpet, a (s_{EC}/s_{UV}) ratio of approximately 10 was found.

Additionally, electrochemical detection shows similar characteristics for all three fungicides, with the minimum concentration detectable being about 0.1 mg/l for Captan and Folpet and 0.2 mg/l for Captafol. On using spectrophotometric detection at 220 nm, the minimum concentration detectable for Folpet was 0.1 mg/l, but about 2–3 mg/l for the compounds Captan and Captafol.

The relative standard deviations, for a concentration level of 2 mg/l, varied between 1.3 and 3.6% for spectrophotometric detection, and between 3.1 and 6.9% for electrochemical detection (Table 1).

3.4. Cloud-point preconcentration

3.4.1. Chromatographic behaviour of surfactant-rich phases.

The cloud point temperature of the surfactant Triton X-114 varies between 18°C and 33°C for surfactant concentrations ranging between 0.05 and 12.5% respectively. These temperatures mean that Triton X-114 can be used to preconcentrate heat-

Table 1
Calibration fits for HPLC determination of fungicides without preconcentration

Fungicide	Intercept	Slope peak height units/mol l^{-1}	Correlation coefficient	R.S.D.[a] (%)
Dual electrochemical detection				
Captan[b]	$(2.0\pm2.4)\ 10^{3}$	$(3.32\pm0.54)\ 10^{10}$	0.9803	5.9
Folpet[b]	$(3.8\pm8.1)\ 10^{3}$	$(2.23\pm0.41)\ 10^{10}$	0.9861	3.1
Captafol[c]	$(1.9\pm0.5)\ 10^{4}$	$(1.26\pm0.06)\ 10^{10}$	0.9908	6.0
UV detection (220 nm)				
Captan	$(-1.3\pm4.5)\ 10^{3}$	$(6.92\pm0.37)\ 10^{7}$	0.9943	3.6
Folpet	$(-3.4\pm2.3)\ 10^{2}$	$(1.77\pm0.02)\ 10^{9}$	0.9993	1.3
Captafol	$(1.3\pm3.9)\ 10^{1}$	$(3.94\pm0.31)\ 10^{7}$	0.9881	2.9

[a]R.S.D. = relative standard deviation ($n=6$). Fortification level: $5.3\cdot10^{-6}$ *M* (1.62 mg/l) Captan; $4.5\cdot10^{-6}$ *M* (1.33 mg/l) Folpet; $7.7\cdot10^{-6}$ *M* (2.70 mg/l) Captafol.[b]$C<8\cdot10^{-6}$ *M*; [c]$C<1\cdot10^{-5}$ *M*; [d]$C<1.5\cdot10^{-5}$ *M*.

sensitive molecules. Additionally, rigorous control of the temperature in the centrifugation step is unnecessary.

The volume of the surfactant-rich phase obtained following phase separation depends on the volume to be preconcentrated, temperature and the percentage of surfactant. The phase ratio (volume of aqueous phase–volume of surfactant-rich phase) governs the preconcentration factor, which cannot be higher than the phase ratio, unless sensitization phenomena occur (in the detection step) due to the presence of the surfactant. Fig. 4 shows the variation in the maximum preconcentration factor and the percentage of Triton X-114 contained in the surfactant-rich phase as a function of the surfactant concentration, working with a sample volume to be preconcentrated of 15.0 ml.

The highest preconcentration factor (75) was found for a surfactant concentration of 0.25%, for which concentration a volume of surfactant-rich phase of 200 μl was obtained. This volume is easily handled and permits the injection of three 10 μl aliquots into the chromatographic system.

Fig. 5 shows the chromatograms obtained following injection of a surfactant-rich phase with spectrophotometric and dual electrochemical detection under the optimum conditions for fungicide separation. When spectrophotometric detection was employed, it was observed that elution of the surfactant shows two very broad peaks. Moreover, the surfactant coeluted with Captan, the detection of the fungicide therefore being impossible. Folpet and Captafol eluted between the two signals of Triton X-114; however, the low spectrophotometric sensitivity of Captafol prevented its detection. Accordingly, on injecting a surfactant-rich phase obtained from 15 ml of aqueous solution with approximately 30 μg/l of the fungicides, only the signal corresponding to Folpet was seen. When dual electrochemical detection was carried out, the surfactant gave rise to a signal at 11 min that did not interfere with those of the fungicides. This signal may be due to electroactive impurities in the surfactant or to species present in the distilled water that are electroactive and that are preconcentrated by the surfactant.

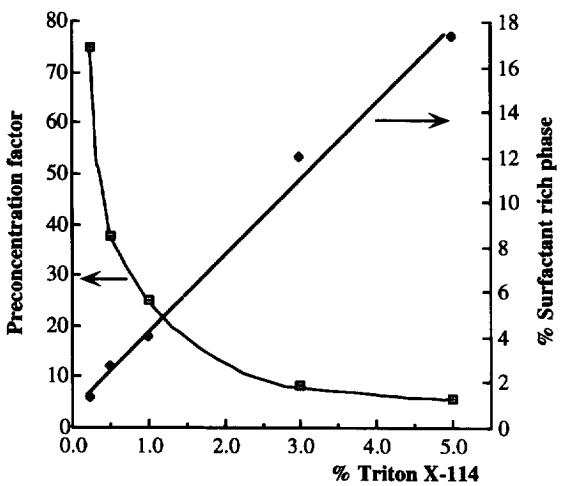

Fig. 4. Preconcentration factor and percentage of Triton X-114 in the surfactant-rich phase versus Triton X-114 concentration. A 15.0-ml volume of sample was preconcentrated.

The amount of organic modifier present in the mobile phase (45% (v/v) of acetonitrile) means that not all of the surfactant is eluted from the column after 30 min. To remove the Triton X-114 remaining in the stationary phase after the analytes have been eluted, a washing cycle with 100% acetonitrile over 10 min was performed after each injection. With this washing cycle it was then not necessary to perform new electrochemical pretreatments on the working electrodes, although the baseline did take some time to become stabilized. However, if during the washing cycle a potential of 0.0 mV was applied to the generating electrode (W_1) and this potential was later changed to -1400 mV the baseline stabilized rapidly.

The percentage of surfactant employed in the preconcentration process determines the volume of surfactant-rich phase and its choice affects the sensitivity of the method. The increase in the percentage of surfactant causes the analytical signal to decrease since the phase ratio becomes more unfavourable. In the cloud point methodology, work should be carried out using the lowest amount of surfactant possible and a sample volume to be preconcentrated that will give rise to a sufficient volume of surfactant-rich phase for repeated injection into the chromatograph. According to the results obtained, sample volumes of 15.0 ml with a surfactant percentage of 0.25% were preconcentrated.

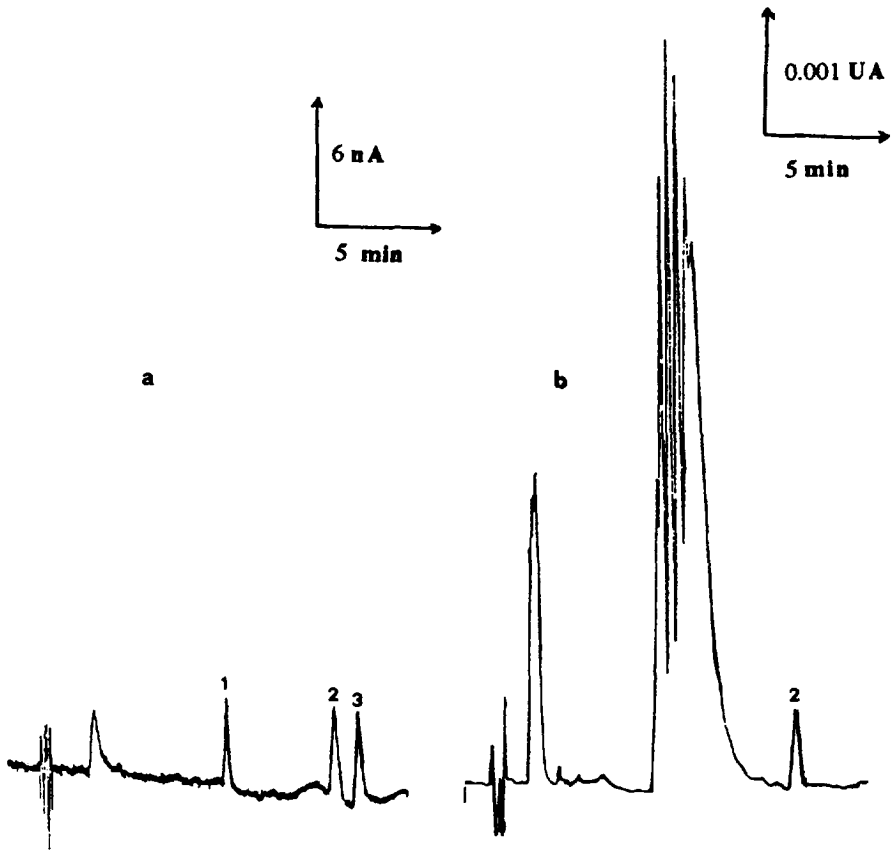

Fig. 5. Chromatograms of a surfactant-rich phase. Preconcentration of 15.0 ml of sample containing 0.25% Triton X-114. (a) Electrochemical detection and (b) UV detection. The concentrations of Captan, Folpet and Captafol were 30.5, 33.3 and 49.0 μg/l, respectively.

3.4.2. Stability of the analytical signal.

Injection of a surfactant-rich phase spiked with a suitable concentration of fungicides shows a lower electrochemical signal than that detected on injection of the same concentration dissolved in acetonitrile. By contrast, the spectrophotometric signal of Folpet did not undergo any variation. The decrease in the electrochemical signal was more pronounced for the fungicide Captan, which coelutes with the surfactant, than for Folpet and Captafol, which elute between the two maxima originated by Triton X-114. This loss in sensitivity is a well known phenomenon when working with solid electrodes and is due to processes of surfactant adsorption onto the electrode surface.

On preconcentrating sample containing fungicide concentrations close to 300 μg/l, the electrochemical signals decreased as the number of injections was increased. However, when ten surfactant-rich phases obtained by preconcentrating 30 μg/l of each fungicide were injected, no appreciable decrease in signal was observed (Table 2). These results indicate that although the presence of the surfactant leads to a loss of sensitivity, the response of the electrode is constant when concentrations lower than 100 μg/l are injected.

3.4.3. Analytical data with preconcentration by the cloud point method

Calibration graphs were constructed for 15.0 ml samples with 0.25% Triton X-114. In all cases, linear relationships were obtained between peak height and concentration (Table 2). Using electrochemical detection the calculated detection limits, for a signal-to-noise ratio of 2, were around 4 μg/l for Captan and

Table 2
Calibration fits for HPLC determination of fungicides after cloud-point preconcentration in the presence of 0.25% Triton X-114

Fungicide	Intercept	Slope peak height units/mol l^{-1}	Correlation coefficient	R.S.D.[a] (%)
Dual electrochemical detection				
Captan	$(8.0\pm2.1)\ 10^2$	$(7.41\pm0.24)\ 10^{11}$	0.9978	4.3
Folpet	$(3.3\pm0.3)\ 10^3$	$(5.83\pm0.04)\ 10^{11}$	0.9986	3.2
Captafol	$(-1.2\pm4.2)\ 10^3$	$(4.54\pm0.40)\ 10^{11}$	0.9923	3.4
UV detection (220 nm)				
Folpet	$(9.3\pm1.5)\ 10^2$	$(1.38\pm0.03)\ 10^{11}$	0.9986	5.0

[a]R.S.D. = relative standard deviation ($n=6$). Fortification level: $1.0\cdot10^{-7}$ *M* (30 μg/l) Captan; $1.0\cdot10^{-7}$ *M* (30 μg/l) Folpet; $1.3\cdot10^{-7}$ *M* (46 μg/l) Captafol.

Folpet and 6 μg/l for Captafol, values which are similar to the maximum levels admitted in the EEC for surface waters destined for the production of drinking water [43].

The relative standard deviations obtained at a concentration level of 30 μg/l were in the 3.2–5.2% range for electrochemical detection (Table 2).

The relationships between the slopes of the calibration fits obtained with and without preconcentration using the cloud point method are shown in Table 3. Using spectrophotometric determination the enhancement factor has a value close to that of the phase ratio, indicating that 100% of the the fungicide Folpet is extracted. Using electrochemical detection, the enhancement factors are lower than the phase ratio due to the decrease of the electrochemical signal in the presence of the surfactant.

3.4.4. Determination of fungicides in river water samples

Most methods described for the determination of fungicides in water samples make use of liquid–liquid and solid–liquid extraction. Another alternative is the cloud point methodology. In order to check the usefulness of this preconcentration technique for these fungicides, the proposed method was applied to the determination of fungicides in river water samples (River Tormes, Salamanca, Spain).

Table 3
Enhancement factor[a]

Fungicide	Enhancement factor
Dual electrochemical detection	
Captan	22
Folpet	26
Captafol	36
UV detection (220 nm)	
Folpet	78

[a]ratio of slopes of the calibration with and without the preconcentration step.

The fungicides studied in this work are not stable in aqueous solution and may undergo hydrolysis reactions during the time between sample collection and analysis. In the chromatograms obtained following injection (over 6 h) of spiked river water samples, a decrease was observed in the signals corresponding to Folpet and Captan together with the appearance of a new chromatographic signal due to the presence of phthalimide. This decrease was more marked in the case of Folpet, followed by Captan, whereas Captafol was more stable (Fig. 6a). Upon injecting an aqueous solution containing 0.25% Triton X-114, the signals were seen to be stable (Fig. 6b) and the signal corresponding to the phthalimide was not observed. Accordingly, it may be concluded that the fungicides are stable for at least 6 h and do not undergo hydrolysis reactions when in the presence of surfactant.

In order to prevent fungicide hydrolysis, surfactant (at a concentration of up to 0.25%) should be added to the samples at the time of collection, prior to the preconcentration step and to chromatographic analysis being carried out.

The river water samples analyzed here did not show any chromatographic signal corresponding to the pesticides studied. Samples were then fortified at two concentration levels between ca. 6 and 50 μg/l of each fungicide (Fig. 7). The analytical recoveries from this matrix ranged from 76 to 106% (Table 4).

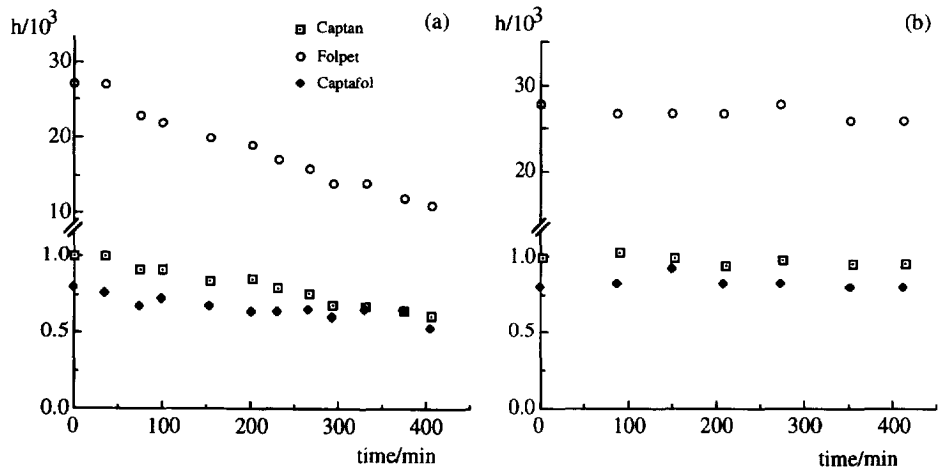

Fig. 6. Variation in the signals of the fungicides as a function of time. Chromatographic conditions were as described in Section 2. UV detection was used. The concentrations of Captan, Folpet and Captafol were 2.8, 3.1 and 4.5 mg/l, respectively. (a) In the absence of Triton X-114 and (b) in the presence of 0.25% Triton X-114.

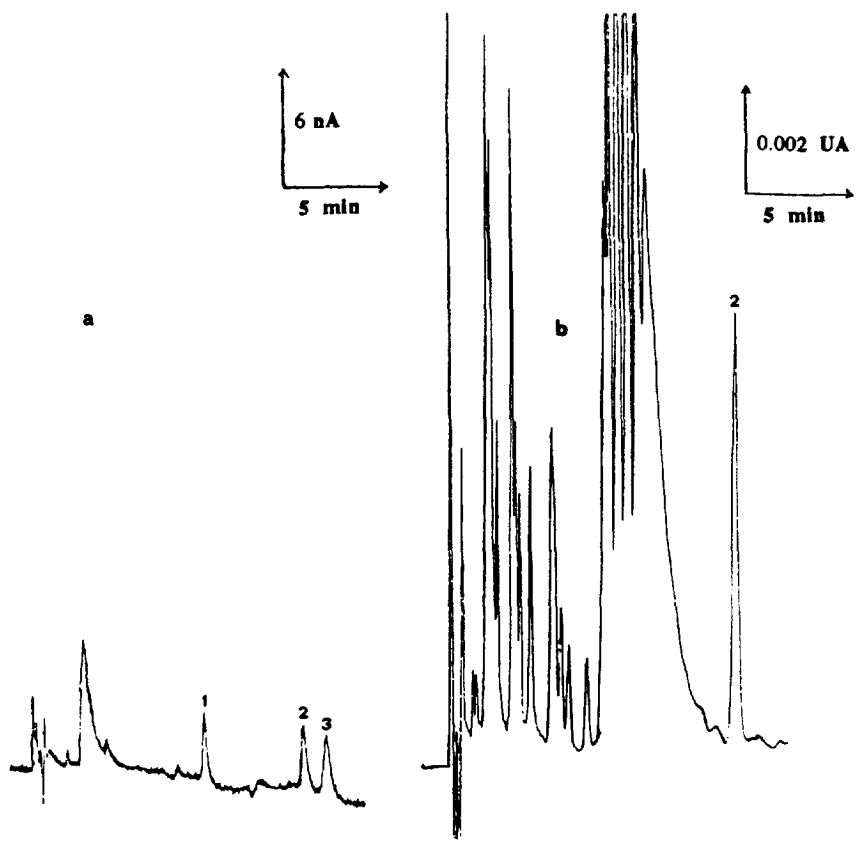

Fig. 7. Chromatograms obtained following the injection of a preconcentrated river water sample fortified at the 30 μg/l level. (a) Electrochemical detection and (b) UV detection.

Table 4
Fungicide recoveries from river water samples after cloud-point preconcentration in the presence of 0.25% Triton X-114

Fungicide	Added μg/l	Recovery (%)	R.S.D.[a] (%)
Captan	6.6 [b]	106	10
	17.4 [b]	96	8
Folpet	6.2 [b]	76	8
	16.0 [b]	93	8
	6.2 [c]	88	6
	16.0 [c]	98	6
Captafol	25.7 [b]	96	12
	51.0 [b]	86	8

[a]R.S.D. = relative standard deviation ($n = 4$)[b]Dual electrochemical detection[c]UV detection

4. Conclusions

Electrochemical detection with a dual glassy-carbon electrode, in the reductive–oxidative mode, coupled to HPLC affords greater sensitivity than spectrophotometric detection for Captan and Captafol, which have low molar absorptivity coefficients in the UV region.

The cloud point method has been applied to the preconcentration of the fungicides Captan, Folpet and Captafol. Electrochemical detection allowed the detection and simultaneous quantification of all three fungicides. By contrast, spectrophotometric detection only allowed the detection and quantification of Folpet.

The presence of Triton X-114 stabilizes the fungicides and prevents their hydrolysis in aqueous medium. Stabilization of the fungicides in the presence of surfactant offers a considerable advantage when attempting to perform reliable analyses in aqueous samples. The addition of surfactant at the time of sample collection is a simple way to avoid loss of fungicide during the period of sample storage. Thus, the determination will afford more precise data concerning the content of these fungicides in the aqueous samples.

Additional work should be carried out to shed further light on the possibilities of the cloud point methodology being used as a step prior to chromatographic separation and on the use of surfactants as stabilizers of pesticides in aqueous solutions.

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