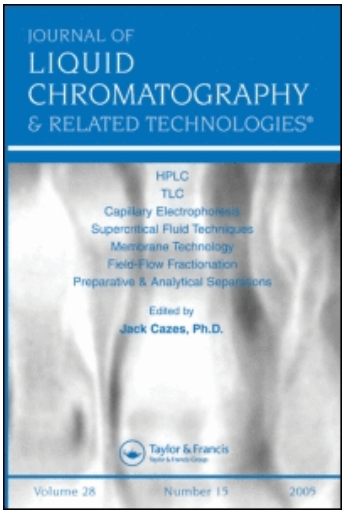

Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

ON-LINE SOLID PHASE EXTRACTION OF PESTICIDE RESIDUES IN NATURAL WATER, COUPLED WITH LIQUID CHROMATOGRAPHY AND UV DETECTION, USING VARIOUS SORBENTS

ZuLiang Chen; Mallavarapu Megharaj; Ravendra Naidu

Online publication date: 24 July 2002

To cite this Article Chen, ZuLiang , Megharaj, Mallavarapu and Naidu, Ravendra(2002) 'ON-LINE SOLID PHASE EXTRACTION OF PESTICIDE RESIDUES IN NATURAL WATER, COUPLED WITH LIQUID CHROMATOGRAPHY AND UV DETECTION, USING VARIOUS SORBENTS', Journal of Liquid Chromatography & Related Technologies, 25: 12, 1779 — 1790
To link to this Article: DOI: 10.1081/JLC-120005873
URL: http://dx.doi.org/10.1081/JLC-120005873

MARCEL DEKKER, INC. • 270 MADISON AVENUE • NEW YORK, NY 10016

JOURNAL OF LIQUID CHROMATOGRAPHY & RELATED TECHNOLOGIES
Vol. 25, No. 12, pp. 1779–1790, 2002

ON-LINE SOLID PHASE EXTRACTION OF PESTICIDE RESIDUES IN NATURAL WATER, COUPLED WITH LIQUID CHROMATOGRAPHY AND UV DETECTION, USING VARIOUS SORBENTS

ZuLiang Chen,* Mallavarapu Megharaj, and Ravendra Naidu

CSIRO Land and Water, Adelaide Laboratory, PMB 2,
Glen Osmond, SA 5064, Australia

ABSTRACT

A technique that couples on-line solid phase extraction with liquid chromatography has been developed for the determination of nine trace pesticide residues (simazine, atrazine, diuron, propazine, linuron, molinate, premetryne, mathalion, and fenitrothion) in natural water samples. Five commercially available sorbents (C_8, C_{18}, PRP-1, PLRPs, and Bond-Elut Env) were evaluated for their suitability for on-line solid phase extraction of the trace pesticides. High recoveries of all pesticides were obtained using C_{18}, PRP-1, PLRPs with good reproducibility and peak shape. C_{18} sorbent allowed the enrichment of up to 100 mL of water spiked with 2 μg/L of pesticides. Recoveries of the tested pesticides ranged from 60.4 to 98.3% ($n = 5$) with relative standard deviations better than 6.9%. Detection limits as

*Corresponding author. E-mail: zuliang.chen@adl.clw.csiro.au

DOI: 10.1081/JLC-120005873
1082-6076 (Print): 1520-572X (Online)

MARCEL DEKKER, INC. • 270 MADISON AVENUE • NEW YORK, NY 10016

low as 0.01 μg/L were achieved when 100 mL of sample was enriched. The method was used to determine pesticide residues in natural water with increased reliability and sensitivity, as well as shorted analysis time, when compared to off-line solid-phase extraction.

INTRODUCTION

Many pesticides are present in natural water at levels typically < 1 μg/L. Consequently, low detection limits are required to directly monitor pesticides in water and to study their fate and transport in the environment. Recently, there has been growing interest in trace enrichment using on-line solid phase extraction (SPE) to improve the detection of pesticides and other organic contaminants in water samples.[1–7] During on-line SPE/high performance liquid chromatography (HPLC), the sample is concentrated by SPE, and the solutes on the precolumn are directly desorbed into the analytical column. Therefore, on-line SPE/HPLC has the advantage of increased sensitivity, accuracy, and is less laborious than conventional off-line methods, because there is no sample manipulation between extraction and determination.[1]

Many different sorbents have been used for the trace enrichment of pesticides and their degradation products. Such sorbents include both *n*-alkyl bonded silicas and apolar copolymers.[2] *N*-alkyl bonded silicas such as C_8 and C_{18} have been used as universal extraction for many years.[1–7] In these sorbents, retention mechanism is primarily governed by hydrophobic interaction between solute and carbonaceous moieties of the alkyl chain, and therefore, these sorbents have been useful for the trace enrichment of non-polar or moderately polar pesticides.[3] Previous studies evaluating on-line cartridges for the enrichment of pesticides have shown that recoveries increase with alkyl group length ($C_2 < C_8 < C_{18}$).[1] The main drawbacks of these sorbents are their limited breakthrough volumes for polar solutes, and their narrow pH stability.[3] The most widely used apolar copolymer sorbents have styrene-divinybenzene (SDVB) backbones. Such copolymers include PRP-1, PLRPs, and Bond-Elut Env, and are frequently used for the on-line enrichment of organic contaminates in water.[5–7] The main advantage of these sorbents over *n*-alkyl bonded silica is their increased pH stability. These sorbents have also proved more suitable than C_{18} for the trace enrichment of polar pesticides from groundwater.[7] Since these sorbents have a higher degree of cross-linking and allow greater π–π interactions between aromatic compound and the sorbent, they are more suitable for polar compounds such as phenols and anilines.[8–10]

Many papers have reported the use of various sorbents for the enrichment of organic contaminates in water samples.[2–7] Only a few reports[11–13] have been

reported on the evaluated *n*-alkyl bonded silica or apolar copolymer sorbents for the enrichment of pesticides in water. In this paper, two *n*-alkyl bonded silica sorbents and three apolar copolymer sorbents commercially available, were systematically evaluated for their suitability for enrichment of nine pesticides commonly found in water samples. Criteria used in the evaluation included pesticide recovery, reproducibility, and peak shape. The parameters affecting the recoveries were investigated to establish the on-line SPE/HPLC method for the analysis of pesticides in trace level in natural water.

EXPERIMENTAL

Chemicals and Solutions

All pesticides were analytical grade reagents obtained from Aldrich and Sigma (Sydney, Australia), and were used without further purification. Pesticide standard solutions were prepared daily by dilution from a 100 mg/L stock solution in acetonitrile. Liquid chromatography grade acetonitrile was obtained from BDH (Pool, England) and filtered through a Millipore 0.45 μm membrane filter and degassed in an ultrasonic bath prior to use. Water samples were filtered through 0.45 μm disposable membranes prior to analysis. Real water samples were collected from different sampling sites in Griffith, NSW, Australia, where pesticide usage was intense. A 50 mL sample was loaded at a flow rate of 5.0 mL/L.

Equipment

All experiments were performed on a fully automated Varian on-line solid phase extraction HPLC system, which included an automatic sample processor Varian (9200 Prospekt), a solvent delivery system (9012), and a diode array detector (9065). The system was controlled by a Star Workstation. On-line SPE cartridges from Varian (5.8×4.6 mm) were used for the sample preconcentration and clean-up, and Waters C_{18} column (250×4 mm) was used for the separation of pesticides.

Procedure

Automation of on-line trace enrichment was performed using a Varian (9200 Prospekt) system using a 10×2.0 mm cartridge packed with the different sorbents (C_8, C_{18}, PLRs, PRP-1, and Bond-Elut Env). The conditioning process

MARCEL DEKKER, INC. • 270 MADISON AVENUE • NEW YORK, NY 10016

was performed sequentially with 10 mL of methanol, 10 mL of deionised water at 2.5 mL/min. The water sample was subsequently loaded at 5 mL/min, before the retained solutes were directly eluted by the mobile phase at 1 mL/min, to the analytical column in the backflush mode over 40 min. Analytical separation of pesticides was performed using a C_{18} analytical column at flow rate of 1 mL/min, using a binary gradient eluent composed of Mili-Q water and acetonitrile with the following protocol, 20% ACN to 30% ACN in 15 min, then up to 50% ACN in 5 min, and up to 70% ACN in 20 min using a flowrate of 1.0 mL/min. The mobile phase was returned to the initial conditions after 10 min of equilibration. The wavelength was set at 220 nm for the detection of all pesticides. Absorbance spectra were recorded from 200 to 300 nm.

RESULTS AND DISCUSSION

Evaluation of Sorbents

The selection of an appropriate SPE sorbent depends on the interaction between the sorbent and the solute of interest. Since the polarity of the tested pesticides varied, both *n*-alkyl bonded silica (C_8 and C_{18}) and three apolar copolymers (PRP-1, PLRPs, and Bond-Elut Env) were evaluated to determine the most appropriate sorbent for pre-concentration of all the pesticides. Percentage recovery (%) and relative standard deviation (RSD%) obtained from cartridges packed with the different sorbents are given in Table 1. Recoveries were determined using 50 mL water samples spiked with 2.0 μg/L of pesticides. For the *n*-alkyl bonded silica sorbent, lower recoveries were consistently obtained using C_8 cartridges. It has been observed previously, that C_8 usually exhibits a low breakthrough volume (V_b) for the moderately polar pesticides, which results in low recovery.[14–16] The recoveries using C_{18} were greater than 89.5% for all pesticides interested, except for fenitrothion (74.9%). Carbon-18 usually provides high recoveries for nonpolar and moderate solutes with octanol–water partition coefficients (log *Kow)* greater than 2.0.[17] It can be concluded, that high surface area of these sorbents lead to increase retention of polar pesticides by the hydrogen bonding between silanol and polar solute groups.[18,19] However, the reproducibilities in both sorbents were lower than 5.4% for all pesticides ($n = 5$), which satisfied the analytical requirement.

PLRPs and PRP-1 had similar recoveries for all pesticides with the exception of fenitrothion (Table 1). Bond-Elut Env also provided the generally good recoveries for most pesticides. However, lower recoveries for molinate (72.6%) and fentrothion (61.6%) were obtained when compared to PLRPs. The reproducibility for all three apolar copolymer sorbents ranged from 0.8–7.9%, which was considered acceptable. Since the breakthrough volumes of apolar

MARCEL DEKKER, INC. • 270 MADISON AVENUE • NEW YORK, NY 10016

Table 1. Recoveries and Relative Standard Deviations (RSD%, $n = 5$) for On-Line Solid Extraction of Pesticides Using Precolumns Packed with Different Sorbents

Solutes	C_8	C_{18}	PLPRs	PRP-1	Bond-Elut Env
Simazine	60.6 ± 3.9	102.6 ± 1.5	99.3 ± 1.1	100.3 ± 1.1	91.8 ± 1.5
Atrazine	79.9 ± 2.1	105.7 ± 1.0	104.5 ± 1.3	104.4 ± 0.8	101.6 ± 1.3
Diuron	85.2 ± 2.3	110.2 ± 2.2	108.9 ± 3.4	108.3 ± 3.15	101.9 ± 2.7
Propazine	86.6 ± 3.8	103.7 ± 0.8	99.3 ± 1.3	99.5 ± 1.0	100.7 ± 4.5
Linuron	80.7 ± 4.7	100.1 ± 1.2	99.4 ± 4.5	102.7 ± 4.3	109.2 ± 6.4
Molinate	85.6 ± 3.2	89.5 ± 3.3	87.9 ± 5.1	107.1 ± 4.6	72.7 ± 7.9
Premetryne	92.4 ± 2.3	105.1 ± 3.1	97.5 ± 5.8	104.3 ± 5.1	108.8 ± 3.2
Mathalion	91.8 ± 4.1	100.9 ± 2.0	93.8 ± 3.2	94.3 ± 2.8	86.9 ± 2.1
Fenitrothion	64.9 ± 3.5	74.9 ± 5.4	80.1 ± 5.3	68.3 ± 4.3	61.2 ± 4.9

copolymer sorbents are 25–40 times higher than that of C_{18} sorbent, they are frequently used for the extraction of very polar solutes.[20] The data in Table 1 shows that there was no significant difference in the recoveries of the pesticides using either C_{18} or PLRPs.

There was no difference in pesticide peak shapes when C_8, C_{18}, PLRPs, and PRP-1 sorbent were used, and the peak height differenced slightly due to the different sorbent breakthrough volumes. However, for the Bond-Elut Env sorbent, the band broadening of peak shapes was observed. This can be attributed to pesticides being retained more on the Bond-Elut Env sorbent than on the C_{18} analytical column, resulting in the pesticides being reconcentrated on the C_{18} analytical column on a broad band.[21–23] This indicates that the breakthrough volume of the pesticide obtained on Bond-Elut Env sorbent is higher than the other sorbents,[22] most likely due to higher cross-linking, which leads to significant peak broadening.[20] Therefore, while C_{18}, PLRPs, and PRP-1 were all suitable for the extraction of the pesticides with high recovery and good peak shape, C_{18} was chosen for all subsequent experiments because it is more frequently used for solid phase extraction and is a significantly cheaper alternative.

Optimising Recovery Parameters

The sample volume and the pesticide concentration can affect the detection limits. Three different sample volumes (50, 75, and 100 mL) spiked at the 2 μg/L of each pesticide were loaded onto the C_{18} sorbent. The resulting recoveries are shown in Table 2. The recovery of the tested pesticide decreased slightly as the sample volume increased. When a 100 mL sample volume was loaded onto C_{18} sorbent, recoveries, ranging from 86.2 to 98.3% were obtained with the exception of fenitrothion (60.4%). The extraction recoveries reduced with the increasing of volume may have resulted from the overload of the cartridge, because other interferences such as organic matter presented in water also absorbed on C_{18} sorbent, leading to reduction in the interaction between the pesticides and C_{18} sorbent.[2,3] Previous studies on SPE extraction by different sorbents have shown that recoveries varied with the water type (river, sea, and tap water) and organic matter content of the water.[23,24]

The effect of pesticides concentration on recovery was examined by passing 100 mL of water spiked with 0.1, 0.5, and 2.0 μg/L of each of the pesticides. The recovery and reproducibility for each pesticide were given in Table 3. Recovery first increased with increasing the pesticide concentration (0.1 to 0.5 μg/L). When the sample contained a 2 μg/L of each pesticide, recoveries decreased. However, the changes in both recovery and reproducibility were not significant overall. Decreased recovery indicates the absolute amount of pesticides falling

Table 2. Recoveries and Relative Standard Deviations (RSD%, $n = 5$) for On-Line SPE of Pesticides Using Three Volumes (mL) Loaded onto C_{18} Cartridge

Solutes	50	75	100
Simazine	102.6 ± 1.5	100.2 ± 3.2	94.3 ± 1.7
Atrazine	105.6 ± 1.0	101.5 ± 4.6	97.0 ± 2.2
Diuron	110.2 ± 2.2	105.7 ± 3.2	98.3 ± 6.9
Propazine	103.7 ± 0.8	101.1 ± 3.9	94.7 ± 5.2
Linuron	100.1 ± 1.1	99.7 ± 2.0	96.4 ± 2.5
Molinate	89.5 ± 3.4	93.9 ± 3.9	91.4 ± 4.9
Premetryne	105.1 ± 3.1	97.9 ± 3.9	84.3 ± 5.3
Mathalion	100.9 ± 2.0	92.3 ± 2.1	86.2 ± 4.6
Fenitrothion	74.8 ± 5.6	68.2 ± 5.7	60.4 ± 5.9

within the loading ability of the cartridge. The low recovery of fenitrothion may result from its low water solubility, possible adsorption onto glass containers, or transfer lines during sample enrichment. Improvement of recovery for this lipophillic solute on SPE cartridges was reported by the addition of organic solvent.[25] These results showed that the sample volume and the pesticide concentration affect the recovery. In summary, a 50 mL volume containing 0.5–2.0 μg/L of pesticide was suitable for enrichment on a C_{18} cartridge with high recovery, reproducibility, and peak shape.

Table 3. Recoveries and Relative Standard Deviations (RSD%, $n = 5$) for On-Line Solid Extraction of Pesticides Using Three Pesiticide Concentration (μg/L) and 100 mL Loaded onto C_{18} Cartridge

Solutes	0.1	0.5	2.0
Simazine	110.8 ± 3.2	100.6 ± 1.2	94.3 ± 1.7
Atrazine	113.2 ± 4.1	107.9 ± 2.6	97.0 ± 2.2
Diuron	115.3 ± 5.9	110.4 ± 5.2	98.3 ± 6.9
Propazine	118.2 ± 5.9	110.7 ± 4.3	94.7 ± 5.2
Linuron	98.7 ± 8.7	108.9 ± 6.8	96.4 ± 2.5
Molinate	80.2 ± 7.5	93.1 ± 7.6	91.4 ± 4.9
Premetryne	108.7 ± 6.8	105.3 ± 4.0	84.3 ± 5.3
Mathalion	99.6 ± 4.2	109.3 ± 4.5	84.3 ± 5.3
Fenitrothion	67.1 ± 4.2	75.6 ± 4.8	60.4 ± 5.9

MARCEL DEKKER, INC. • 270 MADISON AVENUE • NEW YORK, NY 10016

Table 4. The Calibration Data Obtained from On-Line SPE Using C_{18} Cartridge Loaded 50 mL Water Spiked with a Mixture of Pesticide at Level from 0.3 to 2.0 μg/L

Solutes	Regression	R^2	LOD (μg/L)
Simazine	$y = 3.9 \times 10^4 x + 9.6 \times 10^2$	1.0000	0.05
Atrazine	$y = 3.7 \times 10^4 x + 1.1 \times 10^3$	0.9999	0.05
Diuron	$y = 1.2 \times 10^4 x + 1.5 \times 10^2$	1.0000	0.05
Propazine	$y = 3.5 \times 10^4 x + 6.8 \times 10^2$	0.9999	0.05
Linuron	$y = 1.1 \times 10^4 x + 2.4 \times 10^2$	0.9991	0.07
Molinate	$y = 6.1 \times 10^3 x + 6.1 \times 10^3$	0.9991	0.08
Premetryne	$y = 3.5 \times 10^4 x + 1.1 \times 10^3$	0.9981	0.05
Mathalion	$y = 3.1 \times 10^3 x - 2.4 \times 10^2$	0.9982	0.1
Fenitrothion	$y = 8.4 \times 10^3 x + 1.4 \times 10^3$	0.9991	0.08

Analytical Characteristics and Real Sample Analysis

Studies of on-line SPE techniques have shown that calibration curves should be constructed with spiked solution using the whole on-line system, and identical to those employed during real sample analysis.[26] Therefore, calibration curves for each pesticide were constructed after loading 50 mL solution spiked

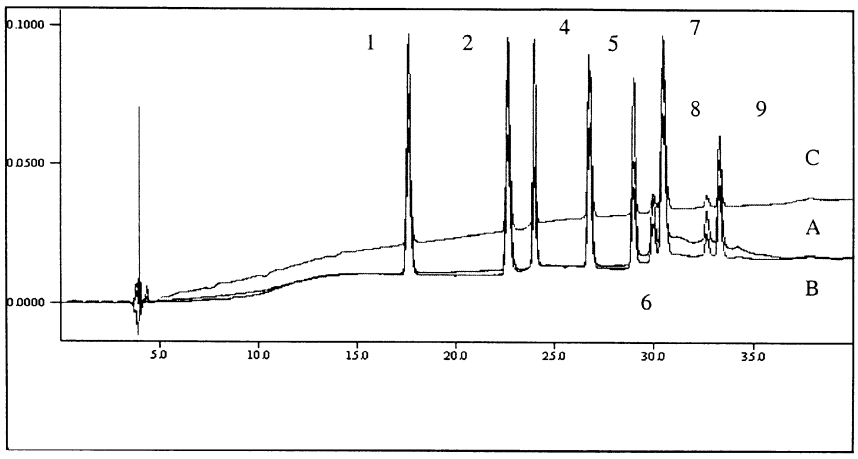

Figure 1. The UV scanning for the selection of the wavelength (A) 210nm; (B) 220nm; (C) 229 nm. (1) simazine; (2) atrazine; (3) diuron; (4) propazine; (5) linuron; (6) molinate; (7) premetryne; (8) mathalion; (9) fenitrothion. Conditions: 50 mL spiked with 2 μg/L pesticides passed onto the precolumn. C_{18} as the sorbent.

with 0.1–4.0 μg/L of each pesticide onto a C_{18} cartridge. Plots of peak area vs. pesticide concentration were linear for all pesticides tested in the range 0.5–3.0 μg/L with the correlation coefficients (r^2) greater than 0.998. The detection limits from passing 50 mL water were less than 0.05 μg/L. However, an increase in the volume (data not shown) may reduce the detection limit.

The detection sensitivities of the pesticides tested varied with wavelengths. Typical chromatograms obtained at 210, 220, and 229 nm are shown in Fig. 1. Lowest sensitivity is observed at 229 nm, while higher sensitivities for simazine, atrazine, propazine, and premetryne were observed at 220 nm. In contrast, high sensitivities for diuron, liuron, and fenitrothion were obtained at 210 nm. This variation can be attributed to the different molar absorptivities of the pesticides due to their different functions. Since detection at lower wavelengths often respond to interferences, a wavelength of 220 nm was chosen as the optimal wavelength for the detection of these pesticides in water.

The proposed method was tested using real water samples to determine if it could be used for monitoring the levels of pesticides in the samples. Figure 2 shows a comparison between (a) on-line solid phase extraction, (b) direct injection with 50 μL. These water samples were collected from sites with high agriculture activity where some pesticides had been applied. Most of the pesticides of interest were detected in the sample using on-line SPE. None were detected when only 50 μL of sample were directly injected onto an analytical column. Pesticides were identified by both spiking samples with known

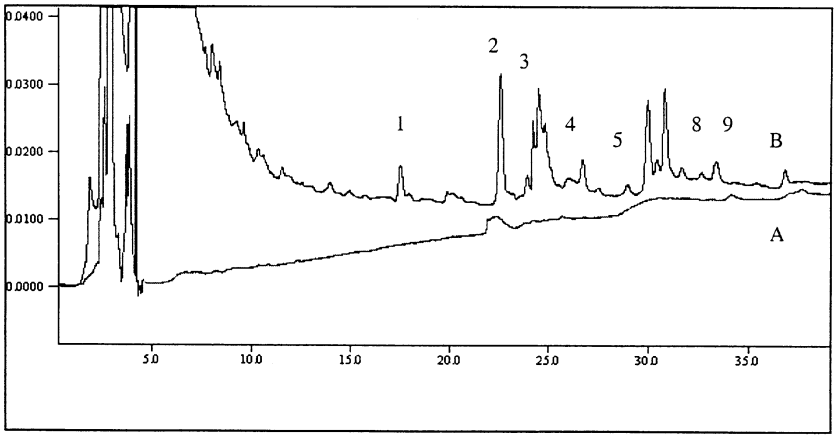

Figure 2. The chromatogram obtained from a real sample contained the pesticides (a) direct injection with 50 μL; (B) 50 mL water sample loaded onto C_{18} cartridge. Other conditions as in Fig. 1.

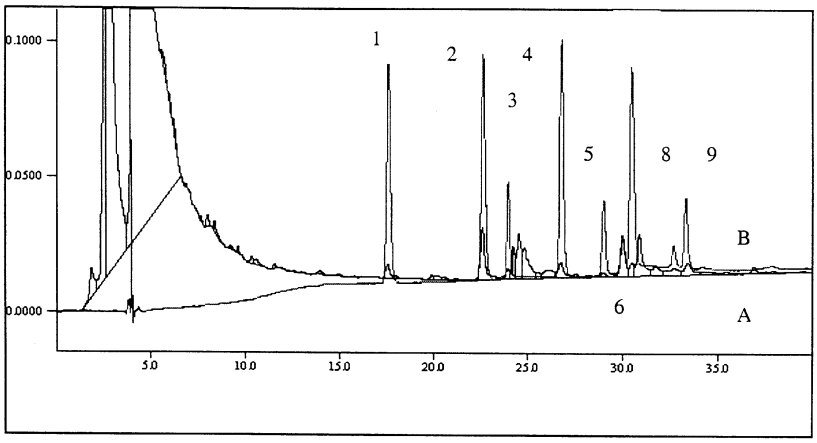

Figure 3. The chromatograms for the identification of the pesticides in water samples using standards. (A) standards; (B) samples containing the pesticides.

concentration of pesticides, and comparison of their retention times with known standards. Figure 3 shows the identification of the pesticides in water samples based on their retention time for a standard solution (a) and real sample (b). Pesticides (μg/L) were detected in these real samples, including simazine (0.26 ± 0.02), atrazine (1.01 ± 0.04), diuron (0.84 ± 0.04), propazine

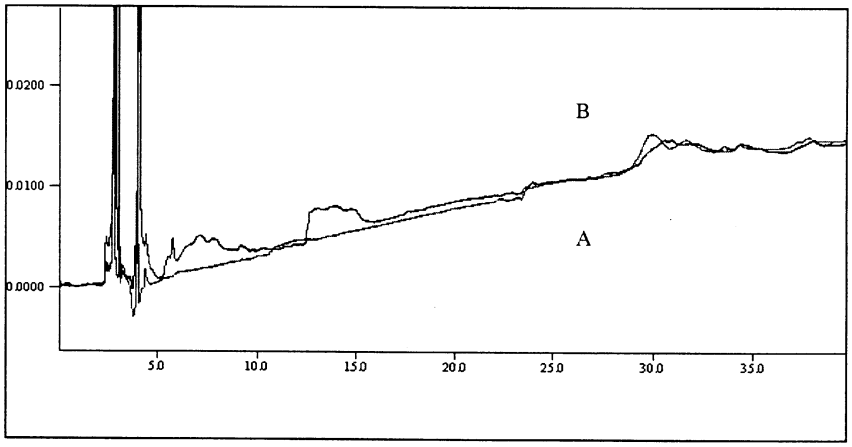

Figure 4. The chromatograms obtained from a sample (no pesticides). (A) Direct injection with 50 μL; (B) 50 mL water sample loaded onto a C_{18} cartridge.

(0.51 ± 0.03), linuron (0.57 ± 0.03), molinate (4.27 ± 0.08), mathalion (0.24 ± 0.02), and fenitrothion (0.44 ± 0.05). Figure 4 shows a river water sample that does not contain pesticides, processed using both on-line SPE (a) and direct injection (b). There are some peaks appearing on the chromatograms, but spiking with known pesticides confirmed these peaks were not the pesticides.

CONCLUSION

On-line solid phase extraction, coupled with reversed-phase liquid chromatography, allows detection of pesticides at trace levels (μg/L) and offers the advantages of high selectivity and sensitivity. Carbon-18, PRLP-s, and PRP-1 provide the best compromise between acceptable recoveries and reasonable peak shape. Recovery is not mainly dependent on the enrichment volume and pesticides concentrations, but possibly depends on the interferences in real samples. The enrichment of larger sample volumes (100 mL) generally resulted in increased in peak broadening for the on-line elution and separation. Analysis of real samples demonstrates that the proposed method could be suitable for the analysis of the pesticides in natural water.

REFERENCES

1. Barceló, D.; Henion, M.-C. *Trace Determination of Pesticides and Their Degradation Products in Water*; Elsevier: Amsterdam, 1997; 243–356.
2. Henion, M.-C. J. Chromatogr. A **1999**, *856*, 3.
3. Masqué, N.; Marcé, R.M.; Borrull, F. Trends Anal. Chem. **1998**, *17*, 384.
4. Henion, M.-C. J. Chromatogr. A **2000**, *885*, 73.
5. Huck, C.W.; Bonn, G.K. J. Chromatogr. A **2000**, *885*, 51.
6. León-González, M.E.; Pérez-Arribas, L.V. J. Chromatogr. A **2000**, *902*, 3.
7. Wells, M.J.M.; Yu, L.Z. J. Chromatogr. A **2000**, *885*, 237.
8. Masqué, N.; Galià, M.; Marcé, R.M.; Borrull, F. J. Chromatogr. A **1997**, *771*, 55.
9. Masqué, N.; Galià, M.; Marcé, R.M.; Borrull, F. Analyst **1997**, *122*, 55.
10. Masqué, N.; Pocurull, E.; Marcé, R.M.; Borrull, F. Chromatographia **1998**, *47*, 177.
11. Guenu, S.; Hennion, M.-C. J. Chromatogr. A **1996**, *737*, 15.
12. Pichon, V.; Chen, L.; Guenu, S.; Hennion, M.-C. J. Chromatogr. **1995**, *711*, 257.
13. Wissiack, R.; Rosenberg, E.; Grasserbauer, M. J. Chromatogr. A **2000**, *896*, 159.
14. Pichon, V.; Hennion, M.-C. J. Chromatogr. A **1994**, *665*, 269.

15. Pocurull, E.; Calull, M.; Marcé, R.M.; Borrull, F. Chromatographia **1994**, *38*, 579.
16. Huen, J.M.; Mayer, A.G.; Baltensperger, B.; Kern, H. Fresenius' J. Anal. Chem. **1994**, *348*, 606.
17. Hennion, M.-C.; Cau-Dit-Coumes, C.; Pichon, V. J. Chromatogr. A **1998**, *823*, 2352.
18. Puig, D.; Barceló, D. Chromatographia **1995**, *40*, 435.
19. Mclaughlin, R.A.; Johnson, B.S. J. Chromatogr. A **1997**, *790*, 161.
20. Pichon, V.; Cau-Dit-Coumes, C.; Chen, L.; Guenu, C.; Hennion, M.-C. J. Chromatogr. A **1996**, *737*, 25.
21. Fedeniuk, R.W.; Shand, P.J. J. Chromatogr. A **1998**, *812*, 3.
22. Pocurull, E.; Marcé, R.M.; Borrull, F.; Chromatographia **1998**, *41*, 521.
23. Lacorte, S.; Vreuls, J.J.; Salau, J.S.; Ventura, F.; Barceló, D. J. Chromatogr. A **1998**, *795*, 71.
24. Frenana-Alba, A.R.; Agüera, A.; Coutreras, M.; Peñuela, G.; Ferrer, I.; Barceló, D. J. Chromatogr. A **1998**, *823*, 35.
25. Brouwer, E.R.; Hermans, A.N.J.; Lingeman, H.; Brinkman, U.A.Th. J. Chromatogr. A **1998**, *48*, 201.
26. Bouzige, M.; Pichon, V.; Hennion, M.-C. J. Chromatogr. A **1998**, *823*, 197.

Received January 29, 2002
Accepted March 6, 2002
Manuscript 5754