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Semiautomated Determination of Pesticides in Water Using Solid Phase Extraction Disks and Gas Chromatography–Mass Spectrometry

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A method based on semiautomated solid phase extraction using octadecyl-bonded silica disks and gas chromatography−mass spectrometry, operated in selected ion monitoring mode, allows detection and quantification of approximately 100 pesticides and transformation products in drinking water. Samples (500 mL) were passed through the disk, and the retained pesticides were eluted with acetone and ethyl acetate. Typical recoveries for pesticides at 0.1 μg L^{-1} in water were in the range of 72−120% with relative standard deviations less than 20%. Calibration curves were linear over the range of 0.025−0.5 μg mL^{-1} (equivalent to a concentration range in drinking water of 0.05−1.0 μg L^{-1}).

KEYWORDS: SPEDEX; SPE disks; gas chromatography; mass spectrometry; analysis of drinking water; pesticide residues

INTRODUCTION

According to the European Union Directive (*1*), individual pesticides in drinking water must not exceed 0.1 μg L^{-1}. To achieve this, methods based on solid phase extraction (SPE) and solid phase microextraction (SPME) techniques have largely replaced liquid−liquid extraction (LLE) for the recovery of pesticides from aqueous samples. SPE is generally performed by passing a sample (0.1−1 L) through sorbent supported in a column, cartridge, or disk format. Of these, SPE disks make use of the smallest sized particles. The high surface area and uniform particle distribution result in extraction efficiencies comparable with those of conventional packed extraction columns or cartridges. The disk format provides a much greater cross-sectional area than the cartridge format. Consequently, high flow rates and large sample volumes can be used, eliminating the channelling effects reported to occur in packed cartridges (*2*, *3*). Octadecyl-bonded silica (C_{18}) is the most widely used sorbent in environmental and food analysis because of its nonselective trapping characteristics (*2*, *4*−*6*). Pesticides retained on the sorbent are eluted with a small volume of an appropriate organic solvent and can be concentrated by evaporation of the solvent prior to chromatographic analysis.

There are relatively few published methods for the determination of pesticides in water at 0.1 μg L^{-1} using SPE disks as compared with the number that employ SPE cartridges and/or SPME. Recently, Westbom et al. (*2*) reported the advantages of SPE using disks and developed a SPE/gas chromatography (GC)-electrochemical detection method for the determination of seven PCBs in water.

The aim of this work was to evaluate extraction using C_{18} SPE disks in a semiautomated system (SPE-DEX), combined with GC-mass spectrometry (MS) analysis, for the determination of approximately 100 pesticides at 0.1 μg L^{-1} in drinking water.

MATERIALS AND METHODS

Reagents, Standards, and Samples. Ethyl acetate [high-performance liquid chromatography (HPLC) grade], acetone (analytical grade), methanol (analytical grade), water (HPLC grade), and anhydrous sodium sulfate (analytical grade) were purchased from Fisher Scientific UK (Loughborough, United Kingdom). Pesticide reference standards (purity > 98.0%) were purchased from Qm$_x$ (Thaxted, United Kingdom) and LGC Promochem (Teddington, United Kingdom). Triphenyl phosphate (TPP) was purchased from Qm$_x$. Three working standard mixtures, containing 1 μg mL^{-1} of each compound, were prepared in ethyl acetate for use as spiking solutions. Matrix-matched calibration standards were prepared using blank sample extracts of water. Laboratory-spiked water samples (500 mL) were prepared for recovery experiments.

Materials. Octadecyl (C_{18})-bonded silica disks, (47 mm; BAKERBOND Speedisk, Cambridge, United Kingdom) were used for SPE. DryDisks (part no. 40-856-HT, Horizon Technology, Salem, United States) were evaluated for removal of water from solvent extracts. Nylon filters (0.45 μm) were purchased from Titan (Sun Sri, Wilmington, NC).

Instrumentation. The extraction was carried out using a SPE-DEX system (Horizon Technology) comprising a SPE-DEX controller and a SPE-DEX 4790 extractor. The system automatically delivers all solvents, conditions the disk, loads the sample directly onto the disk from the sample bottle, thoroughly rinses the sample bottle with solvents

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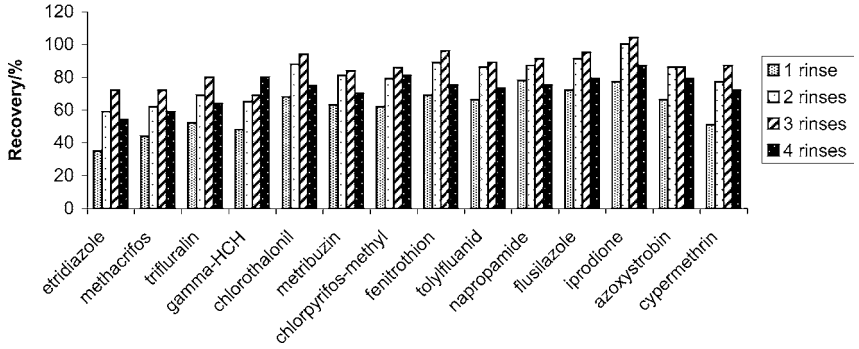

Figure 1. Recoveries from water for a representative group of early-, mid-, and late-eluting pesticides, including several different classes of pesticides. The C_{18} extraction disks were eluted with one constant rinse of acetone and 1–4 rinses of ethyl acetate ($n = 2$).

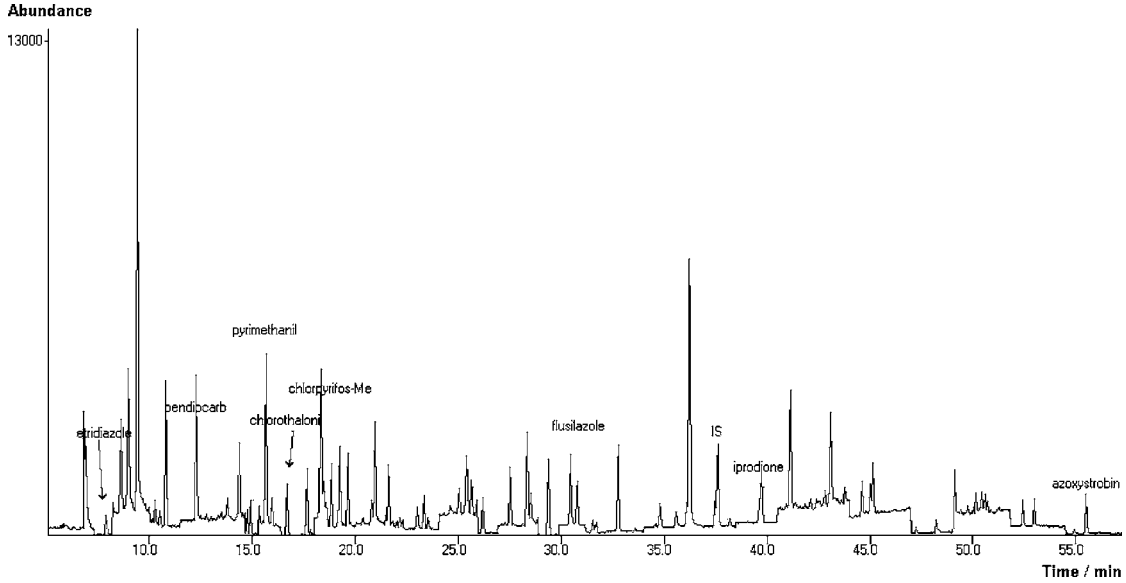

Figure 2. GC-MS RIC chromatogram of a fortified water sample, containing approximately 50 pesticides, at 0.1 μg L^{-1}.

to wash residual sample onto the disk, which is air-dried under vacuum, and elutes the pesticides retained on the SPE disk into a collection vessel. The cycle time for a 500 mL sample is approximately 24 min.

Quantification was performed using a 6890 Series Agilent gas chromatograph with a 5973 Agilent mass selective detector operated in selected ion monitoring (SIM) mode (three ions per analyte), controlled by MSD ChemStation software, version C.00.00 (G1701CA). A fused-silica capillary column (Rtx-5 MS phase, 30 m $\times$ 0.25 mm i.d., 0.25 μm film thickness) with helium carrier gas at 0.9 mL min^{-1} (constant flow) was used for all analyses. Injection (2 μL) was splitless at 250 °C, and the detector temperature was set at 280 °C. The oven temperature program was 100 °C for 4.2 min programmed to 150 °C at 12.5 °C min^{-1}, then to 230 °C at 2 °C min^{-1} (held for 1 min), and finally to 280 °C at 10 °C/min (held for 2.8 min). The total GC run time was 58 min.

Disk Extraction Procedure. A reversed phase sorbent material, C_{18}, was evaluated for a representative group of pesticides, using non-chlorinated elution solvents. A number of elution solvents were evaluated, but the following method offered most advantages. Each disk was conditioned with one rinse each of acetone, ethyl acetate, methanol, and HPLC water. Each rinse corresponds to approximately 5 mL of solvent. Before sample elution, the disk was allowed to dry, under vacuum, for 10 min. After the sample was loaded (500 mL), the disk was eluted with one rinse of acetone and three rinses of ethyl acetate giving a total cycle time of approximately 24 min. The final extract (20 mL) was dried over sodium sulfate and concentrated to less than 1 mL. The concentrated extract was applied to fresh sodium sulfate contained in a 5 mL syringe fitted with a 45 μm nylon filter, which was then rinsed with ethyl acetate until a volume of 5 mL was collected. The extract was concentrated to less than 1 mL, internal standard (TPP) was added, and the volume was made up to 1 mL with ethyl acetate.

Method Performance. To test the precision and the accuracy of the method, five replicate water samples spiked with pesticides at 0.1 μg L^{-1} and five water samples spiked at 0.01 μg L^{-1} were analyzed. Calibration curves generated from matrix-matched standards were used for quantification. All results were calculated using TPP as an internal standard to correct for volumetric errors.

RESULTS AND DISCUSSION

Extraction Procedure. The recovery of the pesticides using C_{18} disks was evaluated for a representative group of compounds, covering a wide range of polarity and volatility. A consideration in the design of the extraction method was to avoid the use of chlorinated solvents. Elution of pesticides from the C_{18} disk with one rinse of acetone followed by three rinses of ethyl acetate produced the best recoveries for most analytes, in the order of 70% at the 0.1 μg L^{-1} level (**Figure 1**). Eluting the disks with further volumes of ethyl acetate did not improve recovery significantly for any pesticide.

A water-miscible solvent (acetone) is employed to remove residual water from the disk. Subsequent elution employs a more hydrophobic solvent to improve recovery of the organic compounds from the disk. Thus, the final extract from SPE is a mixture of water, acetone, and ethyl acetate. Removal of residual water from the final extract and concentration prior to GC-MS analysis are critical steps because of the potential for analyte losses (*7*).

Table 1. Summary of Retention Times, Target Ions, and Average Recoveries for 96 Pesticides and Transformation Products in Water at 0.1 and 0.01 μg L^{-1} Levels, Using C_{18} Disks Followed by GC-MS Analysis in Positive EI, SIM Mode

				water			
				0.1 μg L^{-1} ($n = 5$)		0.01 μg L^{-1} ($n = 5$)	
	compound	t_R (min)	quant ion (target ions)[a] (*m*/*z*)	Rec (%)	RSD (%)	Rec (%)	RSD (%)
1	aldrin	15.41	263 (261, 265)	67	10	55	8
2	atrazine	10.33	200 (215, 202)	93	3	108[b]	8
3	azinphos-methyl	43.08	160 (77, 132)	82	7	IR	
4	azoxystrobin	55.53	344 (388, 372)	105	3	109	3
5	bendiocarb	12.28	151 (166, 223)	97	3	72	7
6	bifenthrin	40.68	181 (166, 182)	78	8	76	6
7	bromopropylate	40.04	341 (339, 343)	75	8	88	6
8	bupirimate	30.77	273 (208, 316)	102	4	97	6
9	captan	25.25	79 (80, 149)	78	15	112	13
10	carbaryl	12.71	144 (115, 116)	100	2	105	7
11	chlordane (cis)	19.93	373 (375, 377)	80	11	87	5
12	chlordane (trans)	18.99	373 (375, 377)	80	11	88	5
13	chlorothalonil	16.69	266 (264, 268)	110	3	106	6
14	chlorpropham	11.70	127 (213, 171)	77	10	75	6
15	chlorpyrifos	15.74	314 (197, 199)	77	9	78	6
16	chlorpyrifos-methyl	18.85	286 (125, 288)	98	3	87	2
17	chlorthal-dimethyl	14.11	301 (332, 330)	78	8	87	5
18	chlozolinate	25.19	259 (188, 331)	64	14	60	17
19	cyanazine	15.85	225 (212, 240)	82	7	108	6
20	cyfluthrin	49.34–49.63	226 (163, 165)	74	10	IR	
21	cyhalothrin-λ	45.15	181 (197, 208)	102	3	88	4
22	cypermethrin	39.72–40.18	163 (165, 181)	116	7	146	22
23	DDE-op	19.35	246 (248, 318)	77	9	NA	
24	DDE-pp	21.40	246 (248, 318)	92	4	85	5
25	DDT-pp	27.08	235 (237, 165)	82	9	80	6
26	deltamethrin	43.86	253 (181, 251)	70	7	IR	
27	dichlofluanid	21.28	226 (224, 167)	76	8	71	8
28	dicloran	13.80	206 (176, 208)	91	7	111[b]	25
29	dicofol	40.60	251 (253, 139)	65	7	IR	
30	dieldrin	21.41	263 (79, 277)	84	8	NA	
31	endosulfan (I)	19.71	241 (195, 197)	82	8	IR	
32	endosulfan (II)	23.61	195 (197, 159)	100	5	IR	14
33	endosulfan-sulfate	26.56	272 (274, 229)	84	7	75	10
34	ethofumesate	21.14	207 (161, 286)	78	10	91	5
35	etridiazole	7.88	183 (211, 248)	84	17	84	9
37	etrimfos	16.81	292 (181, 153)	78	9	76	6
39	fenarimol	45.06	139 (219, 330)	105	7	130	13
40	fenitrothion	14.79	277 (260, 109)	100	3	105[c]	12
41	fenvalerate	52.44–53.00	167 (225, 419)	116	6	82	12
42	flucythrinate	50.75–51.27	199 (157, 451)	78	8	71	14
43	flurochloridone	23.35–23.54	311 (187, 313)	98	3	98	3
44	flusilazole	30.45	233 (206, 315)	101	3	100	5
45	furalaxyl	25.95	242 (152, 95)	83	8	85	7
46	HCH-α	9.69	181 (183, 219)	76	14	81	6
47	HCH-β	10.69	181 (183, 219)	79	11	NA	
48	HCH-γ	10.79	181 (183, 219)	94	3	77	6
49	HCH-δ	11.81	181 (183, 219)	76	13	85	6
50	heptachlor	13.70	272 (274, 337)	70	13	88	3
51	heptachlor epoxide (trans)	17.79	353 (355, 351)	80	12	NA	
52	hexachlorobenzene	9.98	284 (286, 282)	68	13	60	5
53	iprodione	39.64	314 (316, 187)	104	3	74	2
54	isofenphos	25.39	213 (185, 255)	92	36	53	26
55	kresoxim-methyl	31.00	206 (131, 116)	81	8	85	7
56	malaoxon	13.30	127 (99, 109)	83	10	131	8
57	malathion	15.17	173 (127, 158)	99	4	111	7
58	mecarbam	25.61	131 (159, 329)	82	12	98	9
59	metalaxyl	19.76	206 (192, 220)	78	9	95	8
60	methacrifos	8.56	180 (208, 240)	99	6	76	5
61	methoxychlor	31.50	227 (228, 344)	91	10	108	4
62	metoxuron	7.11	183 (168, 140	71	11	74	10
63	metribuzin	18.48	198 (199, 214)	93	6	91	3
64	myclobutanil	30.22	179 (150, 181)	76	9	77	10
65	napropamide	28.36	271 (171, 128)	95	4	105	9
66	nitrothal-isopropyl	23.00	236 (212, 194)	95	3	113	22
67	ofurace	34.81	160 (232, 281)	99	4	94	7
68	oxadixyl	33.27	163 (132, 105)	80	8	84	6
69	oxychlordane	17.68	185 (115, 187)	94	3	NA	
70	paclobutrazol	27.18	236 (238, 167)	96	19	IR	
71	penconazole	24.81	248 (159, 161)	75	9	87	7
72	pendimethalin	17.58	252 (162, 253)	72	7	IR	
73	permethrin	37.86–38.09	183 (163, 165)	85	8	92	7
74	phenthoate	25.64	274 (93, 246)	98	4	94	6

Table 1 (Continued)

				water			
				0.1 μg L^{-1} ($n = 5$)		0.01 μg L^{-1} ($n = 5$)	
	compound	t_R (min)	quant ion (target ions)[a] (m/z)	Rec (%)	RSD (%)	Rec (%)	RSD (%)
75	phosalone	43.09	182 (121, 367)	115	4	122	5
76	phosmet	23.67	160 (161, 317)	106	4	106	3
77	pirimicarb	17.55	166 (238, 72)	75	9	75	6
78	pirimiphos-methyl	13.24	290 (276, 305)	92	5	96	4
79	procymidone	26.10	283 (96, 285)	78	7	71	8
80	profenofos	29.15	208 (139, 339)	81	8	85	8
81	propanil	12.94	161 (163, 217)	95	3	96	6
82	propargite	22.16	135 (173, 350)	98	4	105[c]	12
83	propiconazole	35.59–36.11	173 (259, 261)	106	4	IR	
84	propyzamide	15.37	173 (175, 255)	77	10	97	5
85	pyrimethanil	15.66	198 (199, 200)	95	3	87	3
86	quintozene	15.17	237 (295, 214)	75	10	69	6
87	simazine	10.17	201 (186, 173)	76	9	74	7
88	tebuconazole	36.92	250 (125, 252)	76	13	94	9
89	tecnazene	10.79	203 (215, 261)	73	11	70	5
90	terbuthylazine	10.84	214 (173, 216)	80	8	NA	
91	tetrachlorvinphos	27.53	329 (331, 333)	100	3	93	7
92	tetradifon	42.33	159 (111, 227)	79	8	68	10
93	tolylfluanid	25.04	137 (238, 240)	98	3	104	13
95	trifluralin	8.96	306 (264, 335)	94	3	89	5
96	vinclozolin	18.88	285 (213, 187)	78	10	77	7

[a] SIM windows were set to ensure dwell times between 25 and 100 ms for each ion. [b] Analyte detected and confirmed with two ions with a $S/N \geq 3{:}1$ at 0.01 μg L^{-1} but with low response ($S/N \leq 3{:}1$) for the lowest calibrant level 0.0025 μg mL^{-1}. [c] Analyte detected but not confirmed (S/N of a second ion is <3:1) at the spiking level. IR, insufficient response; NA, not analyzed. Note: All of the analytes that could not be confirmed at 0.01 μg L^{-1} were detected and confirmed at 0.1 μg L^{-1} with a S/N > 3:1.

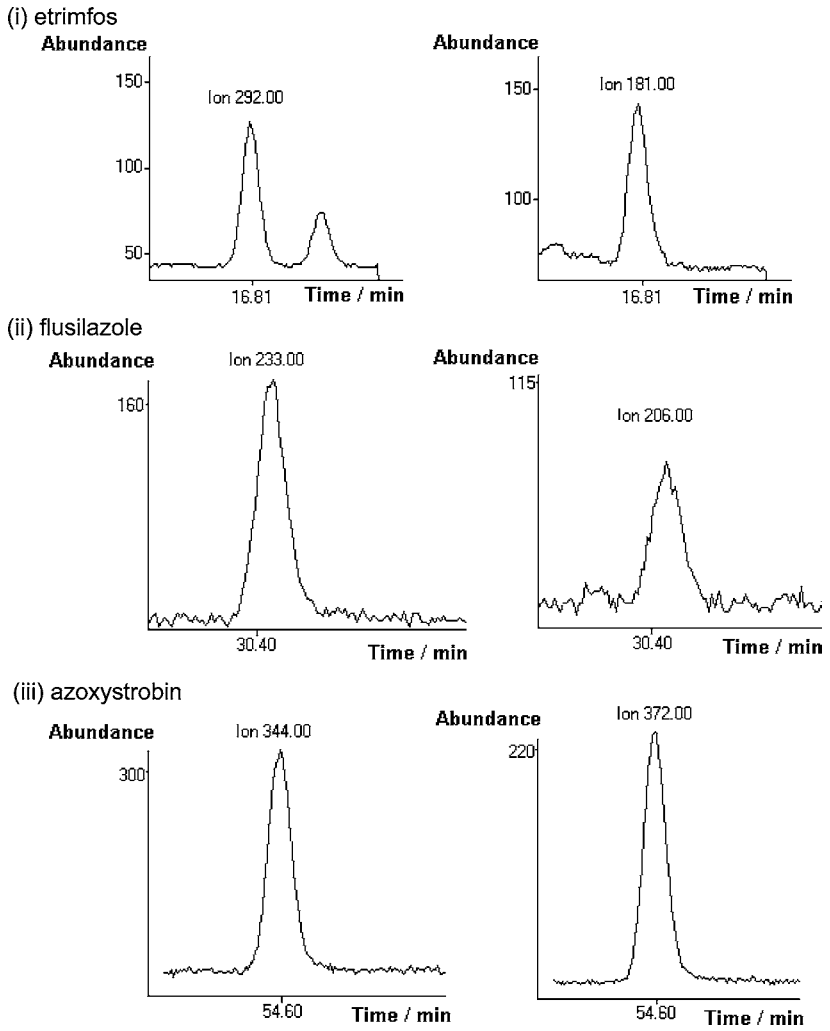

Figure 3. Partial SIM mass chromatograms from an extract of water spiked with pesticides at 0.01 μg L^{-1}: (i) etrimfos (m/z 292 and 181), early-eluting; (ii) flusilazole (m/z 233 and 206); and (iii) azoxystrobin (m/z 344 and 372), late-eluting.

Anhydrous sodium sulfate (Na_2SO_4) is commonly used to remove residual water from solvent extracts before analysis by GC-MS (*8*). An alternative technique, DryDisk Technology, was also evaluated (in syringe format) for removal of water from the extracts. DryDisk employs a PTFE membrane of the appropriate pore size and thickness to allow the solvent to pass through and residual water to be retained. The DryDisks did not remove the residual water from the extracts satisfactorily due to the high miscibility of water with ethyl acetate:acetone extracts. Furthermore, the absolute recoveries of the pesticides obtained after using DryDisks were reduced by approximately 30−50% as compared to Na_2SO_4 (e.g., recovery of simazine using Na_2SO_4 and DryDisks was 78 and 31%, respectively).

Method Performance. Calibration curves were linear over the ranges 0.025−0.5 and 0.0025−0.1 μg mL^{-1} (equivalent to concentration ranges in water of 0.05−1.0 and 0.005−0.2 μg L^{-1}, respectively) with correlation coefficients >0.98. The C_{18} disks gave recoveries in the range 60−116% for the 96 pesticides in laboratory-spiked water samples, with the majority of analytes having RSDs lower than 20% at the spiking levels of 0.1 and 0.01 μg L^{-1} (**Table 1**). A reconstructed ion chromatogram (RIC) of a fortified water sample with approximately 50 pesticides is shown in **Figure 2**.

Pichon (*9*) reported a linear relation between the average log k_w (retention factor of an analyte in water, k_w) values and the log $K_{o/w}$ ($K_{o/w}$ is the water−octanol partition coefficient) for closely related compounds and even for compounds having different polarities and chemical properties. For compounds with a log $K_{o/w}$ > 2.5−3, SPE C_{18} silicas are generally appropriate for multiresidue extraction. In the case of the pesticides successfully analyzed in this study, the range of log $K_{o/w}$ varies from 1.7 for metalaxyl to 7.43 for the very hydrophobic permethrin, with average recoveries of 87 and 88%, respectively. The most polar compounds, e.g., dimethoate (log $K_{o/w}$ 0.704), were not amenable to C_{18} sorbent extraction (data not shown) because the water solubility of these pesticides is too high. For these analytes, a graphitized carbon type sorbent (*10*) and/or liquid chromatography−tandem mass spectrometry (LC-MS/MS) analyses are more appropriate (*11*, *12*).

The majority of the pesticides was detected and confirmed (two or more ions) at 0.01 μg L^{-1} with a *S*/*N* 3:1 (**Figure 3**). Azinphos-methyl, cyfluthrin, deltamethrin, dicofol, endosulfan I, fenitrothion, paclobutrazol, pendimethalin, propargite, and propiconazole could only be screened at 0.01 μg L^{-1} using one selected ion, but all were easily detected and confirmed at 0.1 μg L^{-1}. Other studies using C_{18} disks for the multiresidue analysis of pesticides in water do not report comparable recovery and precision at 0.1 μg L^{-1} with those obtained in the present study (*13*, *14*).

Conclusions. A semiautomated SPE−GC-MS method was developed for the analysis of approximately 100 pesticides in drinking water. The method provides high concentration factors, giving low limits of quantification as required by EU legislation. SPE using disks provides a quick, simple, reproducible, and cost-effective method for the analysis of a large number of pesticides in drinking water. Satisfactory method precision and accuracy were achieved, due to good control of the sample and solvent manipulation in an automated environment. The SPE disk extraction method was less time-consuming and reduced the volume of solvent used by 90% in comparison with LLE. Another advantage of the method is the use of nonchlorinated solvents as elution solvents, reducing the "environmental burden". To fully enforce the EU Directive, the development of multiresidue LC-MS/MS methods for the determination of the more polar pesticides and transformation products is required.

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