

Analysis of Unknown Organic Pollutants in Sewage by Solid-Phase Extraction Combined with Gas Chromatography–Mass Spectrometry

Hong Gao, Ting Zhao, Qin Kong, Xingguo Chen*, and Zhide Hu

Department of Chemistry, Lanzhou University, Lanzhou 730000, P.R. China

Abstract

A rapid, simple, and reliable method of solid-phase extraction (SPE) combined with gas chromatography (GC)–mass spectrometry (MS) is developed for the analysis of a wide range of polarity of unknown organic pollutants in sewage. Wastewater samples are extracted by passing them through disposable C_{18} cartridges, and the extracts are then analyzed by GC–MS. Different SPE parameters for ten organic compounds in the list of priority pollutants suggested by the China Environmental Protection Agency (EPA) are studied, and their breakthrough volumes are determined. Extraction recoveries for the tested compounds are greater than 60%, except the recovery of 1,2-dichloroethane is 48%. The relative standard deviations are less than 7.8% ($n = 3$). The developed approach is successfully applied for the identification of organic components in a sewage sample. Over 220 organic pollutants are identified, with 5 of these present in the list of priority pollutants suggested by the U.S. EPA and 4 from the list by the China EPA.

Introduction

With rapid industry development, a large amount of untreated or inadequately treated industrial sewage has been discharged into rivers, resulting in the deterioration of the aqueous environment. Many unknown microcontaminants are present in environmental aqueous matrices. For treating industrial sewage and assessing and maintaining the quality of surface waters, it is not sufficient that only a target compound is monitored in environmental aqueous matrices. Therefore, the enrichment and identification of unknown organic pollutants in sewage and other environmental aqueous matrices have important environmental significance.

Gas chromatography (GC) and GC–mass spectrometry (MS) are the main analytical techniques employed in environmental organic analysis. A large amount of literature reports the deter-

mination of target organic compounds in various environmental matrices using GC or GC–MS (1–8). However, only a few papers have been devoted to the complete identification of a wider-range polarity of unknown organic contaminations in environmental aqueous matrices (9,10). GC–MS is a powerful technique to separate and identify organic pollutants in environmental samples because of its high sensitivity, selectivity, and separation properties (11–14). In China, however, the samples that contain a broad set of components that may be unknown have not been the subject of routine analytical schemes. Therefore, establishment of a rapid, economic, and reliable analytical procedure for the investigation of unknown organic pollutants in sewage is desirable.

Because of the hazardous nature of sewage, and because organic contaminants at high and low concentration levels exist simultaneously in industrial effluents, the clean up and enrichment step for water samples prior to instrumental analysis is necessary. In past decades, many advanced and important preconcentration techniques, such as purge-and-trap (12), headspace (15), supercritical fluid extraction (2,9), and solid-phase microextraction (16), have been applied for the enrichment of organic compounds in aqueous samples. However, special and expensive devices are required in the preconcentration process. Compared with these techniques, SPE is not only an effective technique for the clean-up and enrichment of water contaminants prior to their instrumental determination, but it is also economical and convenient. Disposable commercial cartridges are available. Compared with conventional liquid–liquid extraction (LLE), SPE offers some advantages that include a cleaner extract, less solvent handling, lower manual labor, and more rapid sample processing (17). In recent years, SPE has proved to be a convenient technique for the enrichment of various organic pollutants in environmental aqueous samples using various sorbents (18–22). Nowadays, SPE has been an alternative sample preparation method to LLE in many U.S. Environmental Protection Agency (EPA) methods for analysis of organic compounds in drinking water and wastewater (17). SPE still remains the most popular sample clean-up in the very active area of sample preparation in the field of separation science.

From a methodological point of view, two basic approaches

* Author to whom correspondence should be addressed.

can be performed: on-line and off-line SPE. In contrast to on-line SPE, one advantage of off-line SPE is the possibility of increasing the breakthrough volume by increasing the amount of the sorbent. Therefore, a higher enrichment factor can be obtained because of its ability to concentrate trace amounts of contaminants on a larger sorbent surface from a large volume of water sample. Another advantage is the ease of application to field sampling, which eliminates the problem of transportation and storage of voluminous samples. SPE uses most of the stationary phases (sorbent) and separation mechanisms available for high-performance liquid chromatography. During the enrichment step, the analytes should be well retained by the sorbent and not eluted by water. Therefore, reversed-phase materials such as sorbents are suitable to the preconcentration of organic contaminants from aqueous samples. The widely used stationary phases include octadecyl-bonded silica (C_{18}), divinyl-benzene-styrene copolymers, and carbon-based matrices. Of these, C_{18} is the most common and easily used because it displays a strong retention for most nonpolar and midpolar organic compounds because of its hydrophobic character. Therefore, disposable C_{18} cartridges were chosen in this study for the enrichment of organic contaminants in the sewage.

The aim of this work is to establish a simple off-line SPE extraction combined with a GC-MS procedure for the clean up, enrichment, and identification of a wider-range polarity of unknown organic pollutants in environmental aqueous samples. In this method, the factors affecting the extraction efficiency in SPE were studied to achieve the maximum recoveries. An untreated sewage sample from Yan-Er-Wan Wastewater Treatment Plant (Lanzhou, China) was analyzed with good results.

Experimental

Materials and reagents

The disposable cartridges containing 500 and 1000 mg of C_{18} adsorbent were purchased from Dalian Institute of Chemical Physics, Chinese Academy of Sciences (Dalian, China).

Methanol, dichloromethane, ethyl acetate, *n*-hexane, 1,2-dichloroethane, chlorobenzene, phenol, nitrobenzene, phenylamine, naphthalene (Second Tianjin Chemical Reagent Factory, Tianjin, China) were of analytical grade and were redistilled prior to use. Ethylbenzene, dimethyl phthalate, and di-*n*-butyl phthalate (First Shanghai Chemical Reagent Factory, Shanghai, China) were of chromatographic grade. Aliphatic standards from *n*- C_8 to *n*- C_{24} , and aromatic standards including benzene, *p*-xylene, *o*-xylene, 1,3,5-trimethyl-benzene, 1,2,4-trimethyl-benzene, propyl-benzene, (1-methylethyl)-benzene, tert-butyl-benzene, anthracene, and biphenyl (Second Tianjin Chemical Reagent Factory) were of chromatographic grade.

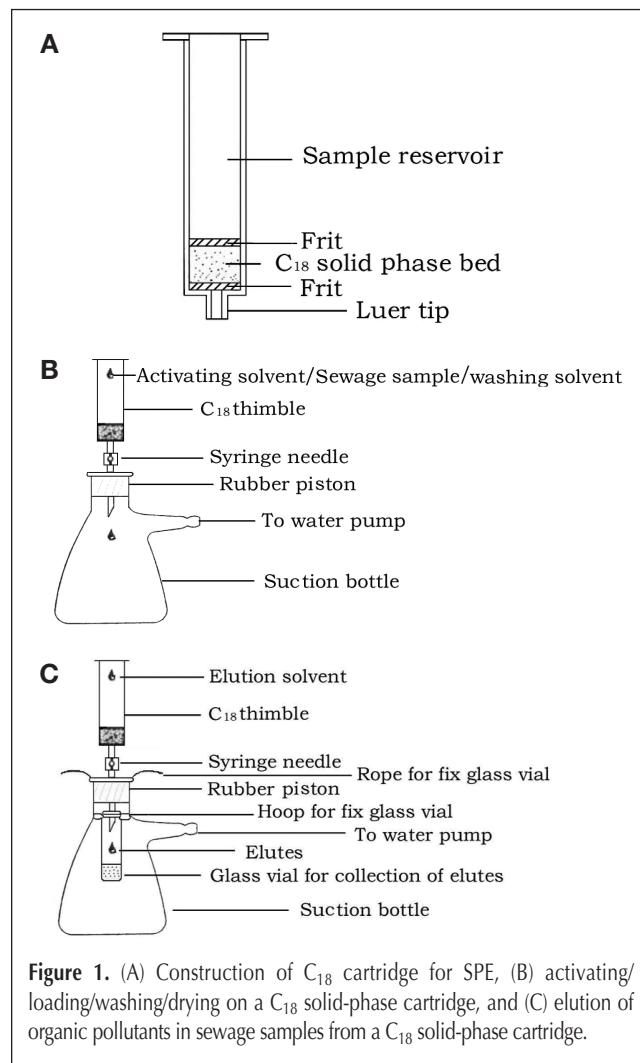
Stock standard solutions in the concentration range of 5000–10,000 mg/L were prepared with dichloromethane and stored at 4°C. Working standard solutions were prepared by diluting stock standard solutions with dichloromethane and stored in the same way. Ultrapure water was prepared with a Milli-Q water purification system (Millipore, Bedford, MA). The pH of aqueous samples was adjusted by using 0.1M of either

hydrochloric acid or sodium hydroxide solutions.

SPE procedure optimization

The SPE equipment, which is easily operated in any laboratory, consisted of a C_{18} cartridge and a suction bottle connected with a water pump. The cartridge was fixed on suction bottles by a syringe needle inserted into the piston (Figure 1).

Ten organic compounds (Table I) were chosen for SPE procedure optimization with disposable 500-mg cartridges. The cartridges were activated before use by passing 5 mL of the elution solvent through them, followed by 5 mL methanol and 5 mL ultrapure water. A volume of 10 mL of ultrapure water was spiked with a mixture containing 5–15 µg of each tested organic compound. The pH and methanol fraction of the spiked solution were adjusted according to the test condition and then loaded onto the cartridges at a flow rate of approximately 3–5 mL/min under reduced pressure using a simple water pump. After loading the water samples, the columns were washed with 5 mL of definite methanol-distilled water (v/v) according to the test condition, and they were then dried by centrifugation at 2000 rpm for 10 min. Finally, the analytes were eluted from the columns, according to the test conditions. The elutes were collected in glass vials and then evaporated under a gentle stream of



nitrogen to 50 μL for GC analysis.

All the experiments of SPE procedure optimization were carried out on a Varian CP3800 GC (Varian, Palo Alto, CA) coupled with a flame-ionization detector. A fused-silica capillary column (50-m \times 0.32-mm i.d., 0.33- μm film thickness, SE-54 cross-linked column) was used. The carrier gas was nitrogen with a flow rate of 2.5 mL/min. A split/splitless injector in the split mode was employed; the injector and detector temperatures were set at 260°C and 280°C, respectively. The temperature program was started at 40°C for 5 min, and a gradient of 4°C/min was used up to 255°C and held for 5 min. A 1.0- μL elute was injected into the injector, which was operated in the split mode at a ratio of 20:1.

Breakthrough experiment

Breakthrough volumes were estimated by measuring a series of peak areas (23) obtained after percolating 10, 25, 50, 100, 150, 200, 300, and 400 mL of ultrapurified water solutions containing 5 μg of each organic compound onto a disposable 500-mg C_{18} cartridge. These experiments were performed according to the optimized SPE procedure and same GC condition.

Sewage sample extraction

A volume of 250 mL of sewage sample was passed through the cartridge directly. A 1000-mg disposable C_{18} cartridge was employed for the extraction of sewage. The cartridge procedure was the same as the optimum SPE process described previously, but twice as much solvent volume was used. The elutes were collected in glass vials and then evaporated under a gentle stream of nitrogen to 50 μL for GC-MS analysis (enrichment factor, 5000) (19).

Qualitative analysis

The extracts of sewage sample were analyzed using a Model TRACE GC-MS (Thermo Finnigan, San Jose, CA) equipped with

an SE-54 fused-silica capillary column (30-m \times 0.25-mm i.d., 0.25- μm film thickness). The following temperature program was used: the initial column temperature was 40°C maintained for 5 min and programmed at 4°C/min to 230°C held for 20 min, then at 5°C/min to 280°C held for 5 min. A split/splitless injector in the split mode was used, and the injector and detector temperatures were set at 290°C and 300°C, respectively. Helium was the carrier gas at a flow rate of 1.0 mL/min. Sample extracts of 1.0 μL were injected in the split mode at a ratio of 20:1. The electron impact ionization conditions were: 70-eV ion energy and 41.00–461.00 m/z range in the full-scan mode. Solvent delay was set at 4.0 min. Working standard mixtures of 1.0 μL were solutions of aliphatic *n*-alkanes and aromatic hydrocarbons, which were injected at the same experiment condition to verify aliphatic *n*-alkanes and aromatic hydrocarbons compounds. Blank extracts of 1.0 μL were also analyzed.

Results and Discussion

SPE procedure optimization

In order to study the extraction efficiency for a wider range of organic pollutants, ten organic compounds with different boiling points (80–335°C) and different specific inductive constant (2.28–34.82) (Table I) in the list of priority pollutants of the China EPA were selected for SPE procedure optimization and breakthrough volumes determination. Among these ten compounds, all were present in the list of priority pollutants suggested by the U.S. EPA, except for phenylamine. The factors that could affect the SPE extraction efficiency were studied, including the elution solvent, elution volume, methanol fraction in aqueous samples (v/v), methanol fraction in washing solvent (v/v), and pH in aqueous samples.

Table I. The Boiling Point, Specific Inductive Constant, and Average Recoveries of 10 Tested Compounds in SPE at the Different Elution Solvent and Different Elution Volume ($n = 3$)

Compound				Average recovery (%)					
				Elution solvent			Elution volume (mL)		
No.	Name	b.p.*	ϵ^{\dagger}	A [‡]	B [§]	C ^{**}	3	5	7
1	1,2-Dichloroethane	84	10.4	48 (6.4) ^{††}	5 (5.3)	78 (2.1)	14 (3.6)	48 (6.4)	66 (4.8)
2	Benzene	80	2.28	60 (2.7)	13 (4.6)	6 (5.3)	15 (2.5)	60 (2.7)	55 (5.6)
3	Chlorobenzene	132	5.62	72 (2.3)	11 (3.4)	36 (3.2)	50 (4.1)	72 (2.3)	89 (1.6)
4	Ethylbenzene	136	2.41	80 (6.7)	17 (6.8)	52 (2.4)	58 (1.7)	80 (6.7)	98 (2.3)
5	Phenylamine	186	6.89	68 (5.6)	10 (7.2)	9 (3.1)	40 (4.3)	68 (5.6)	100 (1.9)
6	Phenol	182	9.78	69 (7.2)	18 (5.4)	5 (3.4)	14 (6.0)	69 (7.2)	81 (3.2)
7	Nitrobenzene	211	34.8	105 (2.3)	54 (1.5)	73 (2.2)	46 (3.8)	105 (2.3)	105 (6.2)
8	Naphthalene	218	2.54	95 (1.9)	50 (2.9)	88 (5.2)	68 (2.9)	95 (1.9)	110 (4.6)
9	Dimethyl phthalate	282	8.50	109 (3.7)	56 (1.7)	18 (4.3)	58 (3.0)	109 (3.7)	115 (6.1)
10	Di- <i>n</i> -butyl phthalate	335	6.44	110 (4.5)	39 (3.3)	26 (6.0)	49 (4.3)	110 (4.5)	102 (4.0)

* b.p, Boiling point of 10 tested compounds.

[†] ϵ , Specific inductive constant of 10 tested compounds.

[‡] A, Dichloromethane.

[§] B, Ethyl acetate.

^{**} C, *n*-Hexane.

^{††} Values in parentheses are RSDs based on three replicate analyses.

Influence of eluent solvent and elution volume

In the SPE procedure, the proper selection of the eluent solvent is the most important step. In order to choose an appropriate solvent for the elution of wider-range polarity organic pollutants from C₁₈ cartridges, dichloromethane, ethyl acetate, and *n*-hexane were tested, respectively. The effect of the different solvents on recovery is listed in Table I. The results indicate that good recoveries were obtained for most of the tested compounds using dichloromethane. Only naphthalene, 1,2-dichloroethane, and nitrobenzene gave better recoveries with *n*-hexane. Poor recoveries were obtained for nearly every compound when using ethyl acetate as the eluent solvent. Therefore, dichloromethane was chosen as the eluent solvent. Another reason for the choice of dichloromethane was that it has a better solubility for a wider range of more polar organic compounds.

Table I gives the recoveries obtained using different volumes

of dichloromethane as the eluent. The results indicate that an increase in the dichloromethane volume leads to an increase in the recoveries of almost all of the tested compounds. But, the operation with a larger volume of eluent in the SPE procedure was relatively difficult and brought a high blank, therefore 5 mL dichloromethane was adopted as the optimum volume.

Influence of methanol fraction in aqueous sample and washing solvent

The addition of an organic component to the aqueous sample can improve the adsorption efficiency of cartridges containing octadecyl-siloxane-bonded silica particles (17). A definite fraction of organic component in aqueous samples can ensure adequate wetting of the cartridge bed and improve extraction efficiency. Meanwhile, in order to wash some interferences for

obtaining a better chromatographic separation, an organic component fraction in washing solvent is necessary. Methanol is the most common and predominant organic modifier added to aqueous samples and washing solvent. The percent of methanol in aqueous samples and washing solvents reported in the literature varied from 1% to 70% (v/v) because compounds with different polarities were analyzed (17). Considering that our study focused on extracting a relatively wide range of polarity of unknown organic pollutants, the high percent of methanol in aqueous samples and washing solvent was not suitable. The higher polar organic compounds would lose because methanol fraction reached an amount for dissolving these compounds partially or completely. Therefore, in this study, the influence of fraction of methanol in the aqueous sample (v/v) and fraction of methanol in the washing solvent (v/v) was tested in the range of 3–10%. As can be observed from Table II, in the percent of methanol of the aqueous sample (v/v) tested, compared with the results of 5% and 3% methanol fraction, the recoveries of phenylamine and phenol in the 10% methanol fraction had a dramatic reduction such that 1,2-dichloroethane and benzene were lost completely and other compounds were lost to a various degree. The recovery of phenylamine with 3% fraction of methanol was better than in 5% of methanol, but the recoveries of 1,2-dichloroethane and benzene were the worst. Similar results were obtained in the 3% methanol fraction of the washing solvent (v/v) test, and although the recoveries of phenylamine and naphthalene were better than 5%, the recoveries of phenol, ethylbenzene, benzene, and 1,2-dichloroethane decreased particularly in benzene and 1,2-dichloroethane. In 10% methanol fraction of the washing solvent (v/v), compared with 3% and 5%, the recoveries of all tested compounds had a reduction at a different degree,

Table II. Average Recoveries of 10 Tested Compounds in SPE at Different Methanol Fraction in Aqueous Samples and Washing Solvent (*n* = 3)

Compound	No.	Name	Average recovery (%)						
			Methanol fraction in the						
			Aqueous sample (v/v)			Washing solvent (v/v)			
			3%	5%	10%	3%	5%	10%	
1	1,2-Dichloroethane	13 (2.1)*	48 (6.4)	–	12 (2.6)	48 (6.4)	14 (4.5)		
2	Benzene	17 (4.3)	60 (2.7)	–	13 (3.5)	60 (2.7)	15 (3.1)		
3	Chlorobenzene	42 (5.2)	72 (2.3)	19 (6.8)	58 (4.5)	72 (2.3)	53 (3.3)		
4	Ethylbenzene	40 (3.3)	80 (6.7)	25 (2.6)	68 (6.9)	80 (6.7)	62 (2.6)		
5	Phenylamine	102 (3.0)	68 (5.6)	7 (3.4)	107 (7.8)	68 (5.6)	72 (2.1)		
6	Phenol	60 (2.9)	69 (7.2)	4 (6.7)	32 (5.3)	69 (7.2)	26 (4.7)		
7	Nitrobenzene	101 (3.3)	105 (2.3)	27 (2.6)	112 (2.6)	105 (2.3)	64 (5.9)		
8	Naphthalene	108 (2.6)	95 (1.9)	26 (3.1)	99 (3.1)	95 (1.9)	74 (3.2)		
9	Dimethyl phthalate	110 (4.7)	109 (3.7)	28 (3.4)	103 (2.5)	109 (3.7)	105 (6.8)		
10	Di- <i>n</i> -butyl phthalate	104 (6.1)	110 (4.5)	34 (6.3)	105 (1.8)	110 (4.5)	109 (1.9)		

* Values in parentheses are RSDs based on three replicate analyses.

Table III. Average Recoveries of 10 Tested Compounds in SPE at Different pH (*n* = 3)

Compound	No.	Name	Average recovery (%)					
			pH					
			2.38	4.71	6.50	7.03	9.03	11.03
1	1,2-Dichloroethane	24 (4.9)*	38 (5.3)	48 (6.4)	43 (7.0)	32 (4.8)	22 (4.1)	
2	Benzene	36 (6.9)	42 (3.9)	60 (2.7)	53 (5.6)	35 (5.3)	36 (5.8)	
3	Chlorobenzene	59 (3.1)	68 (4.7)	72 (2.3)	50 (4.2)	59 (4.6)	56 (3.7)	
4	Ethylbenzene	60 (3.5)	78 (3.3)	80 (6.7)	58 (3.5)	61 (1.6)	53 (4.0)	
5	Phenylamine	14 (6.0)	40 (2.8)	68 (5.6)	72 (2.1)	92 (2.1)	92 (1.9)	
6	Phenol	88 (4.6)	73 (3.6)	69 (7.2)	53 (2.0)	22 (2.4)	18 (2.4)	
7	Nitrobenzene	76 (1.9)	83 (2.3)	105 (2.3)	101 (3.1)	113 (0.8)	115 (3.1)	
8	Naphthalene	68 (2.5)	76 (4.2)	95 (1.9)	97 (5.6)	97 (3.7)	94 (2.8)	
9	Dimethyl phthalate	89 (5.1)	98 (2.2)	109 (3.7)	110 (2.7)	103 (3.7)	111 (4.1)	
10	Di- <i>n</i> -butyl phthalate	84 (6.4)	89 (3.4)	110 (4.5)	108 (6.7)	107 (5.2)	107 (5.0)	

* Values in parentheses are RSDs based on three replicate analyses.

Table IV. Organic Pollutants Identified from Sewage

Peak no. identified	Compound	Retention time (min)	Peak area (%)	Pollutant mark	Peak no. identified	Compound	Retention time (min)	Peak area (%)	Pollutant mark
1	Pyridine	4.09	0.34		51	Iso-methyl-nonane	12.67	0.02	
2	Iso-hexanone	4.13	0.01		52	1-Ethyl-4-methyl-benzene	12.77	0.48	U* C*
3	2,2-Dimethoxybutane	4.31	0.02		53	3-Methyl-nonane	12.84	0.13	
4	2,5-Dimethylhexane	4.46	0.03		54	1,2,4-Trimethyl-benzene	13.13	0.23	U* C*
5	Spiro[2,4]hepta-4,6-diene	4.58	0.56		55	1-Methyl-2-propyl-cyclohexane	3.38	0.05	
6	2-Butyltetrahydro-furan	4.71	0.04		56	3-Dodecanol	13.55	0.02	
7	2-Methyl-butanoic acid, methyl ester	4.90	0.36		57	tert-Butyl-benzene	13.67	0.05	U* C*
8	Dimethoxy-methane	5.02	0.01		58	1-Decene	13.73	0.05	
9	2,3-Dimethyl-2-pentanol	5.12	0.06		59	1-Ethyl-3-methyl-benzene	13.82	1.20	U* C*
10	Iso-hexanone	5.36	0.01		60	<i>n</i> -Decane	14.11	4.18	
11	2-Propyltetra hydropyran	5.56	0.02		61	4-Decene	14.33	0.04	
12	<i>n</i> -Octane	5.64	0.07		62	(1-Methylpropyl)-benzene	14.43	0.02	U* C*
13	1-Hexanal	5.78	0.01		63	5-Butyl-4-nonene	14.65	0.05	
14	cis-1,2 -Dimethyl-cyclohexane	5.91	0.03		64	<i>n</i> -Undecane	14.82	0.03	
15	cis-4-Methyl-cyclohexanol	6.02	0.01		65	1,2,3-Trimethyl-benzene	14.93	0.57	U* C*
16	Acetic acid, butyl ester	6.32	0.01		66	1-Methyl-2-(1-methylethyl)-benzene	15.09	0.04	U* C*
17	2,4-Dimethyl-heptane	6.44	0.02		67	2,2-azobis[2-methyl]-propanenitrile (AZDH)	15.17	0.12	
18	1-Methoxy-hexane	6.74	0.04		68	3-Methyl-3-phenyl-azetidide	15.45	0.45	
19	2-Cyclohexyl-octane	7.00	0.04		69	3,7-Dimethyl-nonane	15.65	0.05	
20	Iso-trimethyl-cyclohexane	7.14	0.05		70	1-Ethyl-2,2,6-trimethylcyclohexane	15.83	0.07	
21	Iso-trimethyl-cyclohexane	7.28	0.01		71	1,2-Diethyl-benzene	16.01	0.05	U* C*
22	<i>N</i> -nitro-1-pentanamine	7.34	0.01	U*	72	1-Methyl-3-propyl-benzene	16.15	0.13	U* C*
23	2-Methyl-cyclopentanane	7.41	0.01		73	2-Methyl-undecane	16.28	0.03	
24	cis-4-Nonene	7.59	0.01		74	5,6-Dimethyl-nonane	16.39	0.09	
25	1-Isopropyl-5-methyl-2-pyrazoline	7.78	0.01		75	1-Ethyl-2,3-dimethyl-benzene	16.43	0.08	U* C*
26	5-Methyl-nonane	7.85	0.02		76	2,3,6-Trimethyl-octane	16.54	0.18	
27	1,2,4,4-Tetramethyl cyclopentene	7.98	0.02		77	Iso-methyl-decane	16.72	0.27	
28	Ethylbenzene	8.09	0.25	U C	78	Iso-methyl-phenol	16.78	0.14	U* C*
29	Iso-methyl-octane	8.18	0.02		79	3-Methyl-decane	16.96	0.26	
30	Iso-methyl-octane	8.26	0.04		80	<i>p</i> -Aminotoluene	17.04	0.01	C*
31	<i>p</i> -Xylene	8.51	0.18	U* C	81	1-Ethyl-2,3-dimethyl-benzene	17.16	0.11	U* C*
32	2,5-Dimethyl-3,4-hexanediol	8.69	0.05		82	1-Methyl-4-(1-methylethyl)-benzene	17.26	0.10	U* C*
33	Methyl-cyclodecane	8.77	0.01		83	1-Butenyl-benzene	17.36	0.05	U* C*
34	4-Ethyl-2-octanol	8.86	0.01		84	1-Methyl-3-(1-methylethyl)-benzene	17.51	0.29	U* C*
35	2,6-Dimethyl-2-octanol	9.06	0.03		85	Iso-methyl-phenol	17.66	0.28	U* C*
36	<i>O</i> -Xylene	9.43	0.65	U* C	86	Iso-methyl-phenol	17.74	0.04	U* C*
37	Cyclohexanone	9.65	0.02		87	Dibutyl acetal	17.88	0.56	
38	<i>n</i> -Nonane	9.78	0.42		88	<i>n</i> -Undecane	18.16	4.03	
39	4-Octen-3-one	10.08	0.01		89	2,5-Dimethyl-phenol	18.61	0.09	U* C*
40	3,3-Dimethyl-hexanal	10.45	0.01		90	4-Ethyl-1,2-dimethyl-benzene	18.76	0.17	U* C*
41	1,2-Dipropyl-cyclopropene	10.60	0.01		91	1,2,4,5-Tetramethyl-benzene	18.90	0.28	U* C*
42	(1-Methylethyl)-benzene	10.77	0.09	U* C*	92	3,7-Dimethyl-decane	19.12	0.07	
43	Propyl-cyclohexane	11.06	0.02		93	2-Methyl-decahydro-naphthalene	19.26	0.05	U* C*
44	3,6-Dimethyl-octane	11.22	0.10		94	Dimethyl carbamothioic acid, <i>o</i> -isopropyl ester	19.34	0.02	
45	3-Ethyl-2-methyl-heptane	11.45	0.07		95	Pentyl-cyclohexane	19.49	0.06	
46	Propyl-benzene	12.07	0.12	U* C*	96	Iso-methyl-indan	19.64	0.13	U* C*
47	1,1,2,3-Tetramethyl-cyclohexane	12.23	0.09						
48	3,4,5-Trimethyl-heptane	12.33	0.04						
49	1-Ethyl-2-methyl-benzene	12.41	0.57	U* C*					
50	1,3,5-Trimethyl-benzene	12.56	0.41	U* C*					

* Abbreviations: U*, substitutes or derivatives compounds of priority pollutants suggested by the U.S. EPA; C*, substitutes or derivatives compounds of priority pollutants suggested by the China EPA; U, priority pollutants suggested by the U.S. EPA; C, priority pollutants suggested by the China EPA; and B*, components were also contained in blank.

Table IV. (Continued) Organic Pollutants Identified from Sewage

Peak no. identified	Compound	Retention time (min)	Peak area (%)	Pollutant mark	Peak no. identified	Compound	Retention time (min)	Peak area (%)	Pollutant mark
97	Iso-methyl-indan	19.98	0.27	U* C*	140	5-Propyl-nonane	27.12	0.05	
98	3-Ethyl-2-methyl-heptane	20.16	0.04		141	Iso-methyl-tridecane	27.22	0.05	
99	5-Methyl-undecane	20.22	0.02		142	2-(1,1-Dimethylethyl)-4-methyl-phenol	27.43	0.15	U* C*
100	Iso-dimethyl-phenol	20.32	0.14	U* C*	143	Iso-methyl-tridecane	27.58	0.15	
101	Iso-dimethyl-phenol	20.42	0.28	U* C*	144	Iso-methyl-tridecane	27.81	0.08	
102	5-Propyl-decane	20.58	0.15		145	2,6,10-trimethyl-dodecane	27.95	0.32	
103	6-(4-Methylphenyl)-2,5-diphenyl-(1 α ,2 α ,5 α ,6 β)-3-cyclohexen-1-amine	20.70	0.04		146	Biphenyl	28.24	0.48	U* C*
104	3-Methyl-undecane	20.80	0.12		147	3,5-bis(1-methylethyl)-phenol	28.42	0.03	U* C*
105	1-Methyl-4-(1-methylpropyl)-benzene	20.94	0.05	U* C*	148	Ethaneperoxy acid, 1-cyano-1-(2-methylphenyl) ethyl ester	28.52	0.13	
106	2-Ethyl-phenol	21.09	0.06	U* C*	149	<i>n</i> -Tetradecane	28.80	1.10	
107	Bicyclo[2,2,1]heptan-2-ol, 7,7-dimethyl acetate	21.22	0.27		150	Iso-dimethyl-naphthalene	29.07	0.10	U* C*
108	Naphthalene	21.41	0.68	U C	151	Iso-dimethyl-naphthalene	29.13	0.08	U* C*
109	1-Methyl-4-(1-methyl-2-propenyl)-benzene	21.57	0.07	U* C*	152	Iso-dimethyl-naphthalene	29.51	0.25	U* C*
110	1,1-Dimethyl-2,3-dihydro- ¹ H-indene	21.77	0.09		153	Iso-dimethyl-naphthalene	29.65	0.23	U* C*
111	<i>n</i> -Dodecane	21.93	0.89		154	2-Phenyl-benzeneacetaldehyde	29.89	0.06	U* C*
112	1,2,4-Trimethyl-ethylbenzene	22.18	0.02	U* C*	155	Iso-ethyl-naphthalene	30.16	0.06	U* C*
113	2,3,6-Trimethyl-phenol	22.26	0.04	U* C*	156	1,4-Dimethyl-naphthalene	30.22	0.02	U* C*
114	2,6-Dimethyl-undecane	22.37	0.36		157	Octyl-cyclohexane	30.34	0.10	
115	5-Hydroxytryptamine- <i>n</i> -behenoyl	22.53	0.02		158	Iso-ethyl-naphthalene	30.61	0.04	U* C*
116	Dimethyltetrasulphide	22.58	0.02		159	Iso-trimethyl-dodecane	30.67	0.37	
117	1,1,3-Trimethyl-2-butyl-cyclohexane	22.63	0.14		160	2-Methyl-tetradecane	30.79	0.14	
118	Benzothiazole	22.98	0.11	B*	161	Iso-trimethyl-dodecanol	31.01	0.15	
119	3-Methyl-4-ethyl-phenol	23.23	0.07	U* C*	162	Decahydro-4,4,8,9,10-pentamethyl-naphthalene	31.15	0.08	
120	2-Cyclohexyl-dodecane	23.34	0.08		163	1-Iodo-dodecane	31.47	0.05	
121	Pentyl-cyclopentane	23.75	0.01		164	2-Methylbiphenyl	31.60	0.08	U* C*
122	2-Methyl-1-methylene-3-(1-methylethenyl)-cyclopentane	23.43	0.03		165	<i>n</i> -Pentadecane	31.94	1.13	
123	Iso-methyl-dodecane	23.52	0.13		166	Butylated hydroxytoluene	32.06	0.20	U* C* B*
124	2,3,4-Trimethyl-decane	24.00	0.08		167	Iso-trimethyl-naphthalene	32.79	0.05	U* C*
125	Iso-methyl-dodecane	24.19	0.07		168	Iso-trimethyl-naphthalene	32.99	0.10	U* C*
126	1,2,3,4-Tetrahydro-6-methyl-naphthalene	24.31	0.04	U* C*	169	Iso-trimethyl-naphthalene	33.44	0.10	U* C*
127	6,6-Dimethyl-undecane	24.45	0.31		170	Heptylcyclohexane	33.54	0.10	
128	7-Methyl-6-tridecene	24.58	0.15		171	3,3-Dimethyl-undecane	33.64	0.08	
129	1,5,6,7-Tetramethylbicyclo[3,2,0]hepta-2,6-diene	24.75	0.04		172	Octadecyliodide	33.82	0.14	
130	4,7-Dimethyl-indan	24.82	0.02	U* C*	173	3-Methyl-pentadecane	34.02	0.05	
131	3,7,11-Trimethyl-1-dodecanol	24.94	0.03		174	Diethyl phthalate	34.64	0.40	U C*
132	<i>n</i> -Tridecane	25.44	0.19		175	<i>n</i> -Hexadecane	34.92	1.08	
133	6-Ethyl-undecane	25.80	0.03		176	2-(Methylthio)benzothiazole	35.12	0.02	
134	1-Ethylidene-1H-indene	25.94	0.39	U* C*	177	2,4-Diphenyl-4H-1,3-benzodioxin	35.62	0.01	
135	Iso-methyl-tridecane	26.00	0.06		178	2,6,10-Trimethyl-pentadecane	36.20	0.51	
136	1,4,7,10-Cyclododecatetraene	26.20	0.09		179	6-Cyclohexyl-tridecane	36.54	0.17	
137	Durolquinone	26.30	0.03		180	3-Methyl-pentadecane	36.70	0.10	
138	1-Chloro-octadecane	26.58	0.04		181	3,7,11-Trimethyl-1-dodecanol	36.82	0.07	
139	Heptylcyclohexane	26.96	0.07		182	9-Hexyl-heptadecane	36.90	0.05	
					183	1-Methyl-7-isopropyl-naphthalene (eudalene)	36.99	0.01	U* C*
					184	6-Methyl-2,4-di-tert-butyl-phenol	37.07	0.05	U* C*
					185	Tert-butyl-2-(2-aimino-4-methyl-5-methoxyphenyl) acetate	37.14	0.06	C*

* Abbreviations: U*, substitutes or derivatives compounds of priority pollutants suggested by the U.S. EPA; C*, substitutes or derivatives compounds of priority pollutants suggested by the China EPA; U, priority pollutants suggested by the U.S. EPA; C, priority pollutants suggested by the China EPA; and B*, components were also contained in blank.

Table IV. (Continued) Organic Pollutants Identified from Sewage

Peak no. identified	Compound	Retention time (min)	Peak area (%)	Pollutant mark	Peak no. identified	Compound	Retention time (min)	Peak area (%)	Pollutant mark
186	<i>n</i> -Heptadecane	37.74	0.94		209	<i>n</i> -Henicosane	47.77	0.57	
187	1-Methyl-fluorene	38.00	0.05	U*	210	1-Dimethyl(prop-2-enyl) silyloxybutane	48.10	0.45	
188	Pentadecanal	38.20	0.09		211	4-Methyl-1-dimethyl (prop-2-enyl) silyloxyptentane	49.19	1.64	
189	Hexathiepane	38.74	0.04		212	<i>n</i> -docosane	50.00	0.53	
190	Tetradecanoic acid	39.63	0.13		213	<i>n</i> -tricosane	52.21	0.47	
191	Anthracene	40.12	0.04	U C*	214	<i>n</i> -Tetracosane	54.44	0.25	
192	1,2-Octadecanediol	40.24	0.12		215	<i>n</i> -pentacosane	57.32	0.17	
193	<i>n</i> -Octadecane	40.42	0.96		216	Bis(2-ethylhexyl)phthalate	58.36	0.13	U C* B*
194	2,6,10,14-Tetramethyl-hexadecane	40.56	0.92		217	Cholan-24-oic-acid,3-(acetyloxy)-7-oxo, methylester(3 α ,5 β)	58.97	0.07	
195	Decyl-cyclohexane	42.14	0.03		218	<i>n</i> -Hexacosane	61.08	0.10	
196	3-Methyl-octadecane	42.23	0.05		219	<i>n</i> -Heptacosane	66.09	0.03	
197	<i>n</i> -Nonadecane	42.98	0.71		220	Cholesta-3,5-diene	68.56	0.32	
198	2-Methyl-anthracene	43.19	0.06	U*	221	Cholest-4-ene	70.18	0.09	
199	1-Ethyl-iodide-quinolinium	44.19	0.17		222	Squalene	73.06	0.04	
200	Phthalic acid,butyl 8-methylnonyl ester	44.39	0.11		223	Cholesterol	74.38	0.13	
201	9-octyl-heptadecane	44.53	0.05		224	4,6-Cholestadien-3 β -ol	74.94	0.10	
202	<i>n</i> -Hexadecanoic acid	44.69	0.60		225	Cholesta-3,5-diene	75.92	0.47	
203	2-Mercaptobenzothiazole (2-MBT)	45.12	0.06		226	6-(4-Methylphenyl)-2,5-diphenyl-(1 α ,2 α ,5 α ,6 β)-3-cyclohexen-1-amine	76.42	0.05	
204	<i>n</i> -Eicosane	45.43	0.67		227	Dimethyl(octadecyloxy)propyl silane	79.28	0.02	
205	3,7-Dimethylphenothiazine	46.00	0.21		228	Cholestanol	81.83	0.82	
206	3,3-Diphenyl-5-methyl- ³ H-pyrazole	46.42	0.02						
207	Trimethylmethoxy silane	46.85	0.02						
208	Cyclic octaatomic sulfur	47.03	0.84						

* Abbreviations: U*, substitutes or derivatives compounds of priority pollutants suggested by the U.S. EPA; C*, substitutes or derivatives compounds of priority pollutants suggested by the China EPA; U, priority pollutants suggested by the U.S. EPA; C, priority pollutants suggested by the China EPA; and B*, components were also contained in blank.

except for dimethyl phthalate and di-*n*-butyl phthalate. Therefore, 5% was selected as the optimum methanol fraction in aqueous sample (v/v) and washing solvent (v/v).

Influence of pH

The influence of the pH in aqueous samples on the extraction efficiency was studied in the range of 2.38–11.03. The results are presented in Table III. It can be seen from the table that the best recoveries for most tested compounds were obtained at pH 6.50, except for phenylamine and phenol. The recoveries of phenylamine increased with the increasing of pH, up to pH 9.03. The recoveries of phenol decreased with an increasing pH; the best recovery was obtained at pH 2.38. This result can be explained from the structure of the two compounds. From this study, we can summarize that for most organic compounds in neutral aqueous solutions, not only alkali compounds but also acid compounds all have a certain extraction efficiency. Therefore, pH 6.50 was chosen as the optimum.

The optimum SPE condition for the pollutants may be summarized as follows: 5% methanol fraction and pH 6.50 in aqueous sample (v/v) was adopted, and 5% methanol fraction–water (v/v) as the washing solvent and 5.0 mL dichloromethane as eluent solvent. Extraction recoveries for 10

tested compounds were greater than 60%, except for 1,2-dichloroethane, and relative standard deviations were less than 7.8% ($n = 3$).

Breakthrough volumes

The breakthrough volume is a significant parameter in SPE. The determination of breakthrough volume may be determined by several methods, including experimental measurements and theoretical prediction (17,23,24). In this study, we chose a method (23) for estimating breakthrough volume, which consists of clean-up samples of increasing volumes, each containing the same amount of various analytes, and then measuring the peak areas of the pollutants eluted from the sorbent. When breakthrough occurs, the amount extracted decreases, as do the peak areas. The results showed that the first breakthrough occurred at 25 mL (10 μ g) for nitrobenzene and 50 mL (15 μ g) for 1,2-dichloroethane and phenol. Three compounds (chlorobenzene, ethylbenzene, and phenylamine) had breakthroughs at 150 mL (25 μ g). For the other compounds, breakthrough volumes were not reached in the range tested. In SPE, breakthrough volume of the solute was found to be dependent on the solute amount. Their concentration in wastewater was approximately at the parts-per-million (mg/L) level (25).

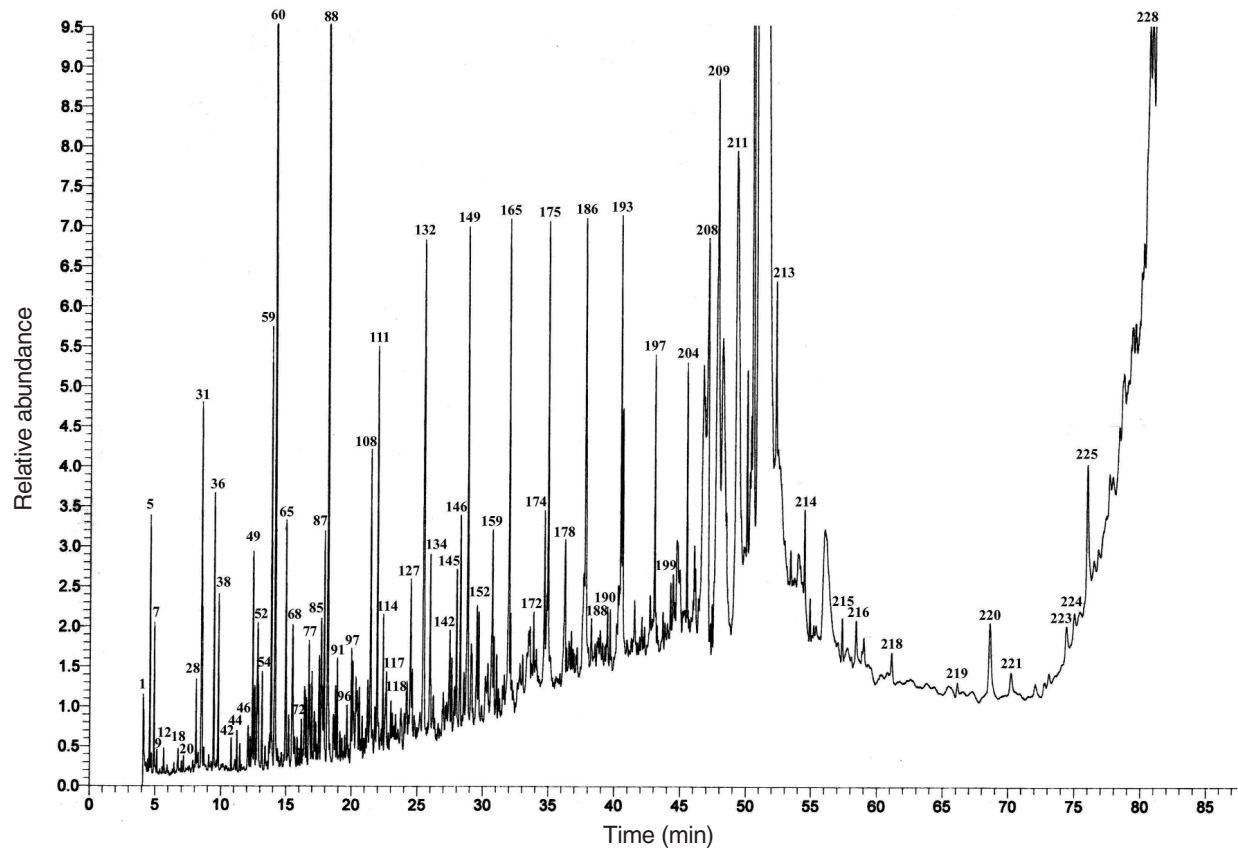


Figure 2. Total ion chromatogram of organic pollutants extracted from sewage. The numbers refer to the substances listed in Table IV.

Combining our test results (the smallest breakthrough amount was 10 μg for nitrobenzene on a 500-mg C_{18} cartridge), 250 mL of sample volume was percolated onto a 1000-mg C_{18} cartridge. Although this volume was too small for some nonpolar compounds to achieve a higher extraction amount, it could enrich higher polar compounds, thus avoiding their early breakthrough. This paper mainly emphasized enrichment and identification of mostly information about unknown organic contaminations in environmental aqueous sample, but also for accurate quantitation. Therefore, it is important to ensure the enrichment of organic pollutants in a wider polarity range.

Qualitative analysis of sewage sample

The identified results of the sewage sample at Yan-Er-Wan Wastewater Treatment Plant (Lanzhou, China) are summarized in Table IV and Figure 2. Table IV lists the names, retention time, and the percent of peak area of the major compounds found in sewage and marked priority pollutants. The majority of the 228 organic compounds identified were aliphatic and aromatic hydrocarbons. The aliphatic *n*-alkanes ranged from C_8 to C_{27} . Figure 2 indicates that many aromatic hydrocarbons and their substitutes and derivatives are detected in 8.00–40.00 min. Other groups mainly included ketone, aldehyde, amine, and phthalate ester. Five compounds appeared in the list of priority pollutants suggested by the U.S. EPA, four compounds in the list of the China EPA, and over 60 compounds were their substitutes and derivatives. The results also showed that the sewage mainly con-

tained petroleum and petrochemical pollutants and their substitutes and derivatives. All the major and minor components were identified by comparing their mass spectra with the NIST library. The identifications of aliphatic and aromatic hydrocarbons were partly confirmed by pure reference substances. The blank also contained bis(2-ethylhexyl) phthalate, butylated hydroxytoluene, and benzothiazole, which indicated that these contaminants already seriously polluted various environmental matrices. It was clear that many compounds with a high boiling point were present after 75.0 min and were not separated completely with GC–MS. The identification of organic pollutants with high boiling points in aqueous samples by LC–MS will be developed in a future study. This method of SPE extraction, combined with GC–MS, can also be used successfully for the analysis of organic pollutants in rivers, which will be discussed in another paper.

Conclusion

A simple method using off-line SPE combined with GC–MS has been proposed for the identification of a wide range of polarity of unknown organic pollutants in sewage. A high enrichment factor (5000) was obtained. The parameters affecting the SPE process were optimized. From this study we can anticipate that unknown organic pollutants can be easily identified if present in environmental aqueous samples using the established

method. The method is rapid and reduces analyst labor, turnaround time, and the risks to human health and the environment with a low solvent consumption.

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