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One step solvent bar microextraction and derivatization followed by gas chromatography–mass spectrometry for the determination of pharmaceutically active compounds in drain water samples

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ABSTRACT

For the first time, a simple and novel one-step combined solvent bar microextraction with derivatization with GC–MS analysis, was developed for the determination of pharmaceutically active compounds (PhACs) in water samples. In the procedure, the derivatization reagent was added in the extraction solvent (solvent bar), so that the analytes could be extracted from the aqueous sample and simultaneously derivatized in the solvent bar to enhance their volatility and improve chromatographic performance. After extraction, the derivatized analytes in the extract were directly injected into a GC–MS system for analysis. Six PhACs including naproxen, ibuprofen, ketoprofen, propranolol, diclofenac, and alprenolol were used here to develop and evaluate the method. The parameters affecting the derivatization and extraction efficiency including derivatization time and temperature, the proportion of derivatization reagent, the type of organic solvent, extraction time, extraction temperature, pH of sample solution, effect of ionic strength, and sample agitation speed, were investigated in detail. Under the most favorable conditions, the method provided good limits of detection ranging from 0.006 to 0.022 μg/L, linearity (from 0.1–50 to 0.2–50 μg/L, depending on analytes) and repeatability of extractions (RSDs below 9.5%, $n = 5$). The proposed method was compared to hollow fiber protected liquid-phase microextraction and solid-phase microextraction, and showed higher extraction efficiency and/or shorter extraction time. The proposed method was applied to the determination of six PhACs in drain water, and was demonstrated to be simple, fast and efficient.

1. Introduction

Pharmaceuticals are commonly and widely used to treat human illnesses. Subsequently, a large quantity of pharmaceutically active compounds (PhACs) and their metabolites have entered the aquatic environment mainly through human waste by excretion of metabolized and unmetabolized versions of these compounds, with some also being discharged during drug manufacturing processes [1–3].

In the past few years, these compounds have been found in various environmental water matrices including river water, waste water, groundwater, sewage water, etc. [4–6]. Even at the relatively low concentrations (ng/L to μg/L range) [2,7,8], PhACs may represent potential risks to aquatic life and human health. Hence, it is important and necessary to develop reliable and sensitive analytical methods for the determination of these compounds at trace levels in environmental aqueous matrices.

High performance liquid chromatography (HPLC) combined with mass spectrometry (MS) [2,3,9,10], diode array detection (DAD) [11,12] or ultraviolet detection [13,14], has been the primary method for the determination of PhACs in environmental aqueous samples.

However, LC–MS may suffer from matrix effects in the form of co-extractive components in the extract, leading to signal suppression and/or enhancement in ESI, and signal enhancement in APCI [7,14], reduced reproducibility, and relatively high limits of detection [15]. Moreover, LC-MS is still a relatively expensive instrument. In addition, if the extractant of the target analytes are not compatible with the mobile phase, an extra step of evaporation and reconstitution is needed [8], further complicating the analytical procedure.

Featuring high selectivity and sensitivity, as well as easy operation and low cost, gas chromatography (GC)–MS has also been widely used in the determination of PhACs in aqueous environment samples [1,7,8,16–18].

Due to their high polarity, PhACs are usually derivatized to reduce their polarity and improve their thermal stability, and volatility to obtain good GC performance.

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N-(*tert*-butyldimethylsilyl)-*N*-methyl-trifluoroacetamide (MTBSTFA) [4,5,7,18], *N*-methyl-*N*-(trimethylsilyl) trifluoroacetamide (MSTFA) [19], and bis(trimethylsilyl) trifluoroacetamide (BSTFA) [6,20,21] are the most commonly used derivatization reagents for PhACs containing hydroxyl or carboxyl functional groups [4,6,22]. MTBSTFA forms *tert*-butyldimethylsilyl (TBDMS) derivatives, which improve MS detection and chromatographic performance due to their high thermal and hydrolytic stability [5,23].

In the determination of PhACs in environmental matrices, a sample preconcentration step is usually required to obtain good selectivity and low limits of detection in the subsequent chromatographic analysis.

Solid-phase extraction (SPE) [6,7,9,24] and liquid–liquid extraction (LLE) [25] are conventionally employed as preconcentration methods for PhAC determination. However, both SPE and LLE are labor intensive and time consuming, and both require moderate to large amounts of organic solvents. Moreover, both may involve multiple steps. To address these disadvantages, solvent-minimized environmentally friendly sample preparation methods have attracted considerable efforts from many researches.

As a solvent free method, solid-phase microextraction (SPME) combines extraction and pre-concentration in a single step and has been widely used for various compounds [12,16]. However, SPME suffers from some shortcomings including analyte carry over and fragility and limited fiber lifetimes, especially if used in the direct immersion mode [26]. Furthermore, commercial SPME fiber coatings generally extract and/or desorb polar analytes with low efficiency.

Liquid-phase microextraction (LPME), a miniaturized analogue of LLE, overcomes some problems associated with SPME. Featuring simple, fast and efficient, LPME has been widely used in various modes, such as single-drop microextraction [27], hollow fiber protected LPME (HF-LPME) [8], dynamic HF-LPME [1,13,28], liquid–liquid–liquid microextraction [3,29], solvent bar microextraction (SBME) [26,30], electro membrane extraction [11,31], and semipermeable membrane devices [32–34].

Developed by Jiang and Lee [30], SBME was demonstrated to be a highly efficient extraction method based on the free and random movement of the solvent bar in a stirred sample solution during extraction, which greatly increases the mass transfer of analytes from the aqueous sample to the extraction solvent.

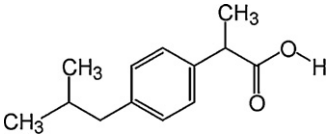
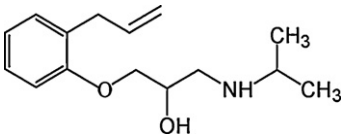
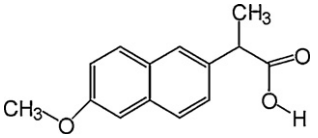
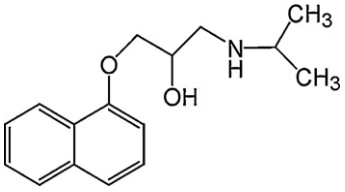
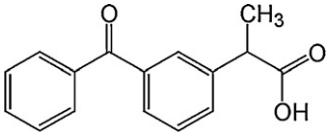
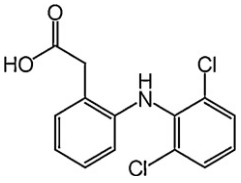
In this study, for the first time, a simple and fast method combining solvent bar microextraction and derivatization in one-step, with GC–MS analysis was developed for the determination of trace PhACs in drain water samples. In this procedure, derivatization reagent was directly added in the acceptor phase so that the analytes were derivatized simultaneously when they were extracted from the aqueous sample solution into the organic solvent (acceptor phase) in the lumen of the solvent bar, which avoids an extra, separate derivatization step and simplifies the extraction procedure. In order to evaluate the method, special attention was placed on the derivatization conditions (the proportion of derivatization reagent) and some factors affecting the extraction including the selection of acceptor phase, extraction time and temperature, pH of sample solution, salt addition, and agitation speed. Finally, the approach was applied to determine six PhACs in drain water samples.

2. Experimental

2.1. Chemicals and materials

Six PhACs, naproxen, ibuprofen, ketoprofen, propranolol, diclofenac, and alprenolol were purchased from Sigma–Aldrich (St. Louis, MO, USA). Their structures are shown in Table 1.

Table 1
Chemical structures of PhACs considered in this study.

Analyte	CAS number	Structure
Ibuprofen	15687-27-1	
Alprenolol	13655-52-2	
Naproxen	22204-53-1	
Propranolol	525-66-6	
Ketoprofen	22071-15-4	
Diclofenac	15307-86-5	

N-(*tert*-butyldimethylsilyl)-*N*-methyl-trifluoroacetamide (MTBSTFA) (97%) was bought from Sigma–Aldrich (Buchs, CH, Switzerland). HPLC-grade methanol, ethyl acetate, and *n*-hexane were purchased from Tedia Company (Fairfield, OH, USA). 1-Octanol and hydrochloric acid were bought from Merck (Darmstadt, Germany) while toluene was supplied by Fisher (Loughborough, UK). The *o*-xylene was obtained from Sigma–Aldrich (St. Louis, MO, USA). Sodium chloride (NaCl) was from Goodrich Chemical Enterprise (Singapore). Ultrapure water was produced on a Nanopure Water Purification System (Barnstead, Dubuque, IA, USA). A magnetic stirrer plate was purchased from Heidolph (Kelheim, Germany).

2.2. Apparatus and instrumentation

The Q 3/2 Accurel polypropylene hollow fiber (tubular type) was purchased from Membrana (Wuppertal, Germany). The inner diameter of the hollow fiber was 600 μm, the wall thickness was 200 μm, and the wall pore size was 0.2 μm.

The commercial SPME holder for manual use and polydimethylsiloxane (PDMS) fibers (100-μm film thickness) was obtained from Supelco (Bellefonte, PA, USA). Prior to use, the fibers were conditioned in the GC injector port at 250 °C for 30 min according to the instructions recommended by the supplier.

A microsyringe (10 μL) with a cone needle tip (SGE, Sydney, Australia) was used for filling the hollow fiber membrane with acceptor solution. A second microsyringe (10-μL) of the same type was used for drawing out analyte-enriched extractant from the hollow fiber membrane after extraction.

2.3. GC–MS analysis

Sample analyses were carried out on a Shimadzu (Kyoto, Japan) QP2010 GC–MS system equipped with a Shimadzu AOC-20i auto sampler and a DB-5 MS (J&W Scientific, Folsom, CA, USA) fused silica capillary column (30 m × 0.25 mm internal diameter (i.d.), 0.25 μm film thickness). Helium (purity 99.9999%) was employed as the carrier gas at a flow rate of 1.7 mL/min. samples were injected in splitless mode and sampling time was 2.0 min. The injector temperature was set at 300 °C and the interface temperature maintained at 280 °C. The GC oven was initially held at 80 °C for 0.5 min, and programmed to 250 °C at 10 °C/min and held for 1 min. Finally, it was programmed to 300 °C at 20 °C/min and held for 3 min. The solvent cut time was 6 min. The derivatives of PhACs were analyzed in selective ion monitoring (SIM) mode for quantitative determination. The monitored ions of the derivatives were selected based on the good selectivity and high sensitivity, and were set as follows: ibuprofen, *m*/*z* 263, 161; alprenolol, *m*/*z* 72, 205, 306; naproxen, *m*/*z* 287, 185; propranolol, *m*/*z* 72; ketoprofen, *m*/*z* 311, 295, 267; and diclofenac, *m*/*z* 352, 354, 214, and 409. All the experiments were performed in triplicate.

2.4. Sample preparation

A stock standard solution (1000 mg/L of each analyte) was prepared with methanol and stored in the refrigerator at 4 °C. External calibration was use for quantification of the analytes, where a series of standard solutions was prepared by diluting the stock solution and analyzing with GC–MS to obtain linear calibration plots for each analyte based on the chromatographic peak areas. Water samples were prepared by spiking ultrapure water with analytes at known concentrations to study extraction performance and evaluate the extraction conditions as indicated in the individual experiments.

Drain water samples were collected from a drain in the university campus into pre-cleaned glass bottles. All collected samples were transported to the laboratory immediately, and stored in the refrigerator at 4 °C until use. To avoid the possible loss of target analytes, the samples were extracted and analyzed without any prior treatment or filtration.

2.5. SBME with derivatization

The SBME procedure was carried out according to our previous work [26]. Briefly, to prepare the solvent bar, the hollow fiber was manually and carefully cut into 2.8-cm segments. These were ultrasonically cleaned in HPLC-grade acetone and dried in air before use. One end of the hollow fiber was heat-sealed. A suitable volume of acceptor phase (added with suitable ratio of derivatization reagent) was withdrawn into a 10-μL microsyringe with the cone needle tip. The needle tip was carefully inserted into the open end of the hollow fiber, and the mixture was introduced into the lumen of the fiber. Then the fiber was carefully removed from the needle. Its open end was carefully clamped with a pair of sharp-tipped pliers first, and was then heat-sealed by another pair of flat-tipped pliers which was pre-heated. The fiber formed a solvent bar with two sealed ends. No leakage was observed when heat-sealing the fiber.

The solvent bar was immersed in the organic solvent for about 25 s to impregnate the wall pores of the hollow fiber. The solvent bar was then placed in the sample solution for extraction. The aqueous solution was kept under stirring at 700 rpm during extraction

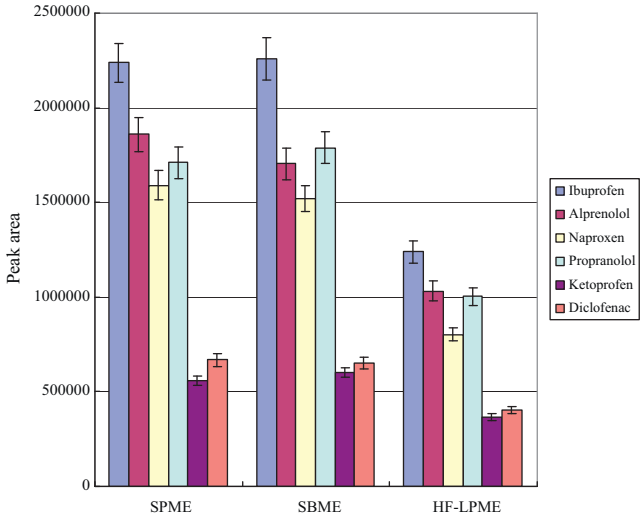


Fig. 1. Comparison of SPME, SBME, and HF-LPME.

procedure. After a prescribed time of extraction, the solvent bar was retrieved with a pair of tweezers. One end of the solvent bar was trimmed off with a sharp blade, and the analyte-enriched extractant was carefully withdrawn into a microsyringe. Finally, a 1-μL aliquot of the extractant was directly injected into the GC–MS system for analysis. The used fiber was discarded, and a fresh one was used for the next extraction.

2.6. Conventional HF-LPME with derivatization

Briefly, the hollow fiber was cut into 2.80 cm segments and cleaned ultrasonically in acetone and dried in air, and then one end was heat-sealed. A suitable volume of acceptor phase (together with a suitable amount of derivatization reagent) was introduced into the lumen of the hollow fiber using a 10-μL microsyringe with a cone tip needle. The fiber was immersed in 1-octanol for 25 s to impregnate its wall pores. Then, the fiber with the microsyringe was placed in a 10 mL of sample solution (in a 15-mL vial) for extraction for 40 min under a stirring speed of 700 rpm. After extraction, the hollow fiber-syringe assembly was removed from the sample solution. The extractant was carefully withdrawn into the syringe and subsequently, 1 μL of the extractant was directly injected into the GC–MS system for analysis.

2.7. SPME with derivatization

SPME was carried out using a manual SPME device with a PDMS coating (100 μm thickness). A 15 mL vial was filled with 10 mL sample solution. The fiber was immersed in the sample solution for 60 min extraction under magnetic stirring (700 rpm). After extraction, the SPME fiber was placed in the headspace of a 1.5 mL GC autosampler vial containing MTBSTFA for derivatization for 20 min. For GC–MS analysis, thermal desorption was carried out at the temperature of 280 °C for 3 min. Blank desorptions were carried out periodically to confirm that there was no contamination or carryover effect.

3. Results and discussion

3.1. Comparative studies

SBME was compared with HF-LPME and SPME in terms of the extraction performance. As shown in Fig. 1, the peak areas obtained by SBME and SPME were comparable, and much higher than that

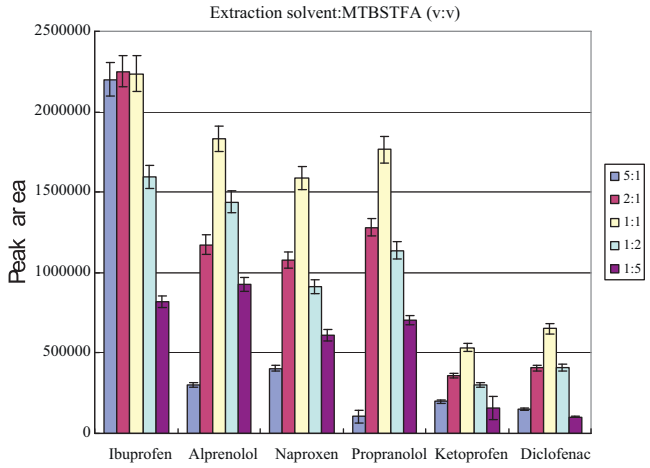


Fig. 2. Effect of organic solvent:MTBSTFA ratios on extraction.

of HF-LPME, although SPME extraction time was 60 min compared to 40 min for HF-LPME.

Compared to HF-LPME, not only was the extraction, but also the extraction efficiency of SBME was better. Both the movement of sample solution and solvent bar facilitate the contact of the solvent bar with the sample, thereby accelerating analyte transfer from the sample solution to the organic solvent.

Based on the comparable extraction efficiency with SPME, the extraction time for SBME with on-site derivatization was only 20 min, much less than that of SPME with derivatization (60 min extraction + 20 min derivatization). In addition, compared to the SPME fiber, which was much expensive and fragile, and could potentially suffer from carry-over effects if special precautions were not taken, the solvent bar was cost-effective and was not affected by carry-over since it was used only once, and then discarded.

3.2. Derivatization

3.2.1. Derivatization reagent

In the present work, PhACs were derivatized to enhance their volatility and improve chromatographic performance (preventing peak tailing) in the GC–MS analysis. The derivatization reaction with MTBSTFA forms the respective *tert*-butyldimethylsilyl (TBDMS) derivatives.

The molecular ions of TBDMS derivatives are relatively weak or absent; however, the parent compounds are characterized by having $[M-57]^+$ ions which are dominant with electron impact ionization mass spectrometry (EI-MS) [17,35]. In this study, except for the TBDMS derivatives of alprenolol and propranolol, the $[M-57]^+$ ions were the base peaks in the EI-MS for all other TBDMS derivatives, which favor the quantitative measurement of the PhACs under SIM mode. In addition, the TBDMS derivatives were thermally stable and resistant to hydrolysis [4,5].

3.2.2. Volume ratio of derivatization reagent

The volume of MTBSTFA added was the key factor affecting the derivatization. Different volume ratios of organic solvent:MTBSTFA (5:1, 2:1, 1:1, 1:2, and 1:5) were studied. The results are shown in Fig. 2. The peak areas of ibuprofen showed no significant increase with the increase of MTBSTFA ratios from 5:1 to 1:1. For the other five analytes, lower peak areas were observed at a lower proportion of MTBSTFA of 5:1, possibly indicating incomplete derivatization, especially for propranolol, of which the peak area was very low. With the organic solvent:MTBSTFA ratio increased from 5:1 to 2:1, the peak areas of these five analytes increased, and reached the maxima at an organic solvent:MTBSTFA ratio of 1:1. When the ratios were changed from 1:1 to 1:5, the peak areas for all analytes decreased, showing the reduced sensitivity for the analytes as well as poor GC resolution, as previously observed [35]. This could be explained by the fact that the GC stationary phase was affected negatively under a higher proportion of MTBSTFA due to the derivatization of the siloxane group [36]. Thus, the derivatization was carried out at an organic solvent:MTBSTFA ratio of 1:1 for subsequent experiments.

3.3. Optimization

The parameters that affect the extraction efficiency, including the type of organic solvent, extraction time and extraction temperature, the effect of ionic strength, sample pH, and agitation speed, were investigated to obtain the most favorable extraction conditions. The optimization was carried out with simultaneous derivatization (since an objective of the work was to reduce the number of separate processing steps), and was based on the extraction efficiency, in terms of the peak areas of analytes. All experiments were conducted in triplicate.

3.3.1. The type of organic solvent

The selection of organic solvent is critical in SBME. It was chosen based on the following considerations: (1) the analytes should have high partition coefficients in the organic solvent; (2) the solvent should be compatible with the polypropylene hollow fiber and then, also be easily and securely immobilized in its pores; (3) it should have very low water solubility to avoid dissolution in the sample solution; and (4) it should have low vapor pressure to prevent loss during extraction.

1-Octanol, toluene, hexane, ethyl acetate, and *o*-xylene, were studied in this work. The same solvent was, of course, used as extraction solvent and solvent impregnated in the wall pores of the hollow fiber. The results are shown in Fig. 3, which shows that the highest peak areas for all the analytes (except for ibuprofen) were obtained by 1-octanol, followed by toluene, then *o*-xylene, and finally hexane and ethyl acetate. 1-Octanol, toluene, and *o*-xylene give comparable peak areas for ibuprofen. Moreover, it was observed that 1-octanol was more easily immobilized in the pores of the hollow fiber.

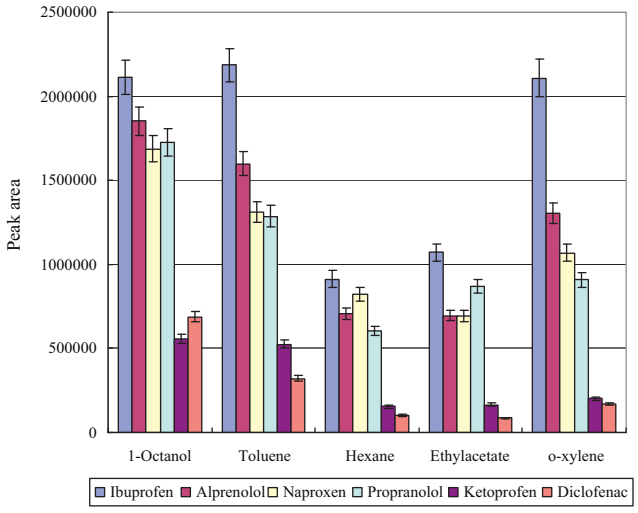


Fig. 3. Effect of the type of organic solvent on extraction.

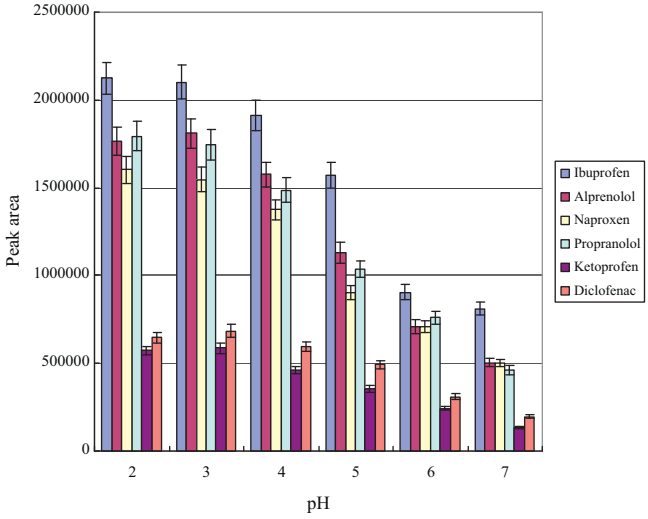


Fig. 4. Effect of sample pH on extraction.

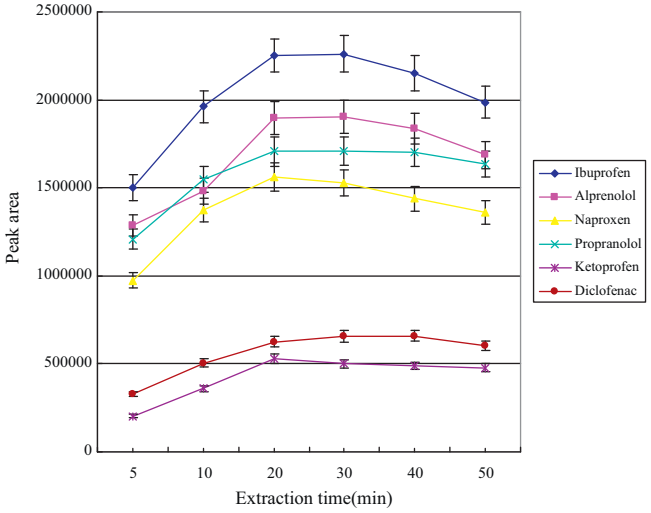


Fig. 6. Extraction time profiles.

3.3.2. *The pH of sample solution*

The pH of the sample solution plays an important role in SBME. The effect of pH on the extraction efficiency was investigated in the range of 2–7 by adding appropriate amount of HCl (0.1 M) in the sample solution. From Fig. 4, it can be seen that the peak areas for all analytes maintained constant when the pH values were 2–3, and further decreased with the increase of sample solution pH from 3 to 7.

The PhACs are weekly acidic, therefore, in order to obtain efficient extraction, the sample solution should be at a suitable pH to suppress their ionization, and keep them in their neutral states to be extracted into the organic solvent. The analytes could not be trapped and concentrated in the organic solvent in their ionized forms. Based on the above discussion, sample solutions were adjusted to a pH value of 3 in subsequent experiments.

3.3.3. *The effect of extraction temperature*

A series of experiments was carried out at 23 °C (room temperature), 30, 40, 50, 60, and 70 °C, respectively, to study the effect of temperature on extraction efficiency. Fig. 5 shows that the peak areas for all analytes were enhanced with the increase of temperature, up to ca. 60 °C and then, declined. Therefore, extraction was conducted at 60 °C.

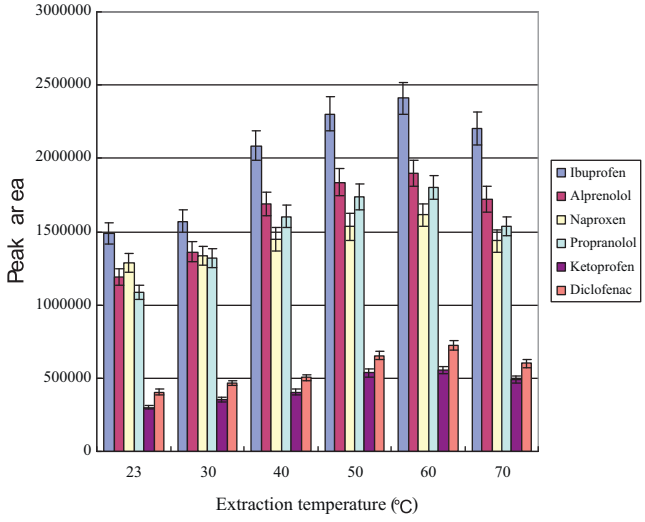


Fig. 5. Effect of temperature on extraction.

3.3.4. *Extraction time profiles*

A series of extraction times (5, 10, 20, 30, 40, and 50 min) was studied to evaluate their effect on extraction efficiency. Fig. 6 shows that the peak areas of all analytes increased quickly when the extraction time was increased from 5 to 20 min. Subsequently, the peak areas flattened out, indicating that equilibrium had been reached. The peak areas of most analytes decreased after 30 min, depending on different analytes. Such an observation with prolonged extraction time is common in LPME and SPME.

SBME is an equilibrium-based extraction process. Therefore, the extraction efficiency depends on analytes transferring from the sample solution to the organic solvent, which is time-dependent. The extraction efficiency could be enhanced by permitting extraction time to reach a point when equilibrium is attained, after which, any further increase would have no significant effect. Thus, in general, equilibrium time would be selected as the extraction time. On the other hand, due to possible solvent dissolution in the sample solution, the longer the extraction time, the greater the loss of organic solvent impregnated in the pores of hollow fiber, which may lead to a decrease in the extraction efficiency. In the present case, equilibrium was reached at 20 min extraction time.

3.3.5. *Effect of ionic strength*

Generally, in LLE, LPME and SPME, salt is added to the aqueous sample to improve the partition of analytes to the organic solvent (salting-out effect). In this study, various amounts of sodium chloride (NaCl) (ranging from 0 to 30%, w/v) were added to the sample solution to investigate this effect. Fig. 7 shows that the peak areas of all analytes increased slightly with the increase of the NaCl from 0 to 10%, and then remained almost constant in the range of 10–15%. However, the peak areas decreased for all analytes when the NaCl concentration was higher than 20%.

Therefore, NaCl concentration was limited to 10%, although 15% could have been used as well.

3.3.6. *Agitation speed*

As regards the effect of sample agitation on extraction efficiency, different stirring speeds from 300 to 1250 rpm were studied. As shown in Fig. 8, peak areas of all analytes were enhanced with the increase of the stirring speed from 300 to 700 rpm.

Under a higher stirring speed, the extraction was accelerated by the enhanced partitioning of the analytes into the organic solvent, as well as the continuous exposure of the solvent bar to fresh regions of the sample solution.

Table 2
Linear range, limits of detection, limits of quantification, recovery, and precision of SBME with derivatization of PhACs.

Analyte	Linear range (μg/L)	Correlation coefficient (*r*)	LOD (μg/L)	LOQ (μg/L)	RSD[a] (%, *n* = 5)
Ibuprofen	0.1–50	0.9931	0.006	0.030	4.7
Alprenolol	0.1–50	0.9929	0.008	0.030	7.1
Naproxen	0.1–50	0.9922	0.010	0.040	5.6
Propranolol	0.1–50	0.9913	0.012	0.040	7.0
Ketoprofen	0.2–50	0.9918	0.020	0.070	8.7
Diclofenac	0.2–50	0.9902	0.022	0.080	9.5

[a] Spiked at LOQ levels.

Table 3
Comparison of LODs with different methods.

Method	Analyte	Solvent and volume	LODs (μg/L)	Ref.
EME-HPLC–DAD	Non-steroidal anti-inflammatory drugs	pH 12 aqueous solution, 50 μL	0.08–3.36	[11]
SPME-GC–MS	Anti-inflammatory drugs		0.012–0.04[a]	[18]
Ultrasonic solvent Extraction-SPE-GC–MS	Pharmaceuticals	Acetone–ethyl acetate 18 mL and ethyl acetate, 8 mL	0.2–2.4 ng g^{-1} [a]	[24]
Pressurized hot water extraction-HF-LPME-LC–MS	Non-steroidal anti-inflammatory drugs		0.04–3.7 ng g^{-1} [a]	[3]
Dynamic LPME-GC–MS	Acidic pharmaceutically active compounds	1-Octanol, 2 μL	0.01–0.05	[1]
Dual SPME-LC–MS	Pharmaceutical compounds		0.005–0.05[a]	[2]
SPE-GC–MS	Pharmaceuticals	Methanol, 2 mL	0.025–0.28[a]	[7]
SPE-UPLC–MS	Pharmaceuticals	Methanol, 5 mL	0.0086–0.974	[9]
SPE-LC–MS	Pharmaceuticals	Methanol, 12 mL[b]	0.0002–0.281	[10]
Continuous-HF-LPME-GC–FID	Non-steroidal anti-inflammatory drugs	1-Octanol, 4 μL	0.001–0.002	[8]
Continuous SPE-GC–MS	Pharmaceuticals	Ethyl acetate, 0.4 mL	0.01–0.06 ng/L	[6]

[a] Limits of quantification (LOQ).
[b] 3 × 2 mL methanol, 2 mL methyltertbutylether–methanol (90:10), 2 mL (2% ammonium hydroxide in methanol), and 2 mL (0.2% sodium hydroxide in methanol).

On the other hand, under a higher agitation speed (>1000 rpm), air bubbles were produced and the loss of organic solvent impregnated in the wall of the hollow fiber might occur, leading to the decrease in peak areas for all the analytes.

Based on the above discussion, the most favorable SBME conditions were: 1-octanol:MTBSTFA (1:1) as acceptor phase, agitation speed of 700 rpm, addition of 10% (w/v) NaCl, sample solution at pH 3, extraction time of 20 min and extraction temperature of 60 °C. All the following experiments were carried out under these conditions.

3.4. Method validation

The performance and reliability of the developed one-step SBME with derivatization was studied by determining the repeatability, linear range, limits of detection (LODs), and limits of quantification (LOQs) for all the target analytes under the described conditions. Table 2 shows the results obtained. The current method exhibited good calibration plot linearity of 0.2–50 μg/L for ketoprofen and diclofenac, and 0.1–50 μg/L for other four analytes, with correlation coefficient (r) higher than 0.9902 for all analytes. The relative standard deviations (RSDs) were lower than 9.5%, indicating the method had good repeatability, which was investigated for five replicate analyses at the same operational parameters. The LODs, based on a signal-to-noise ratio (S/N) of 3, ranged from 0.006 to 0.022 μg/L. The LOQs, based on an S/N ratio of 10, ranged from 0.030 to 0.080 μg/L.

From Table 3, it can been seen that the LODs obtained were lower than those obtained by EME-HPLC–DAD [11], SPME-derivatization-GC–MS [18], ultrasonic solvent extraction–SPE-GC–MS [24], pressurized hot water extraction-HF-LPME-LC–MS [3], in the same range as those obtained by dynamic-HF-LPME-GC–MS [1], SPME-LC–MS [2], SPE-GC–MS [7], SPE-UPLC–MS [9], SPE-LC–MS [10], but higher than these achieved by continuous-HF-LPME-GC–FID [8]

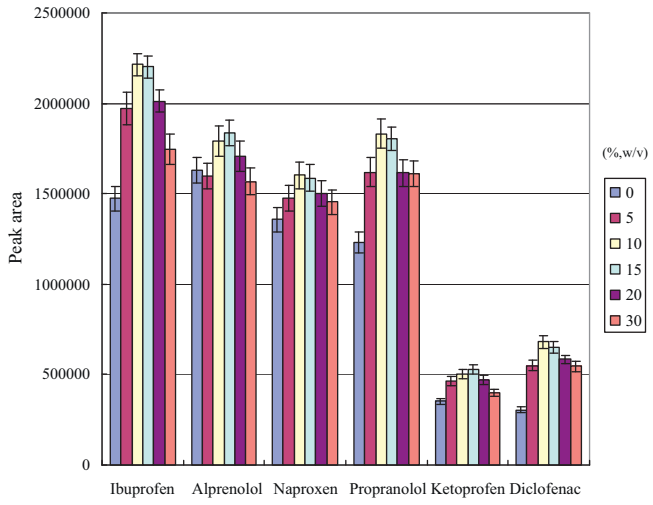


Fig. 7. Effect of ionic strength on the extraction.

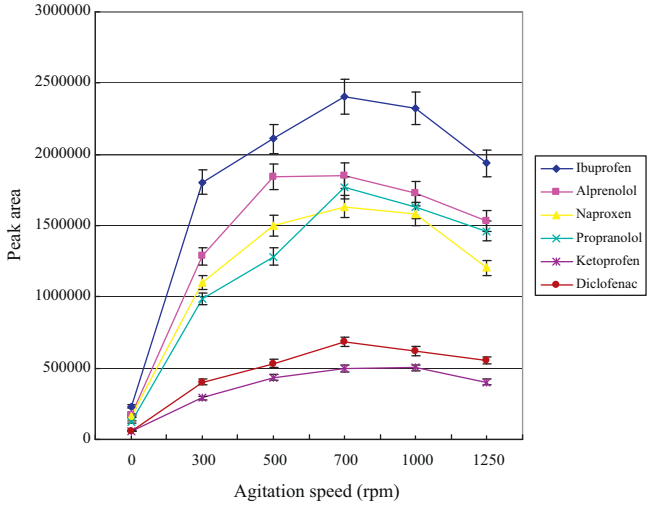


Fig. 8. Effect of agitation speed on extraction.

Table 4
Summary of results from analysis of PhACs in spiked genuine drain water samples by SBME with derivatization.

Analyte	Concentration of PhACs in drain water (μg/L)	Spiked drain water (10 μg/L of each analyte)	
		Relative recovery (%)	RSD (%)
Ibuprofen	0.15	99	7.6
Alprenolol	nd	92	7.9
Naproxen	0.21	102	6.7
Propranolol	0.26	101	6.2
Ketoprofen	0.42	105	9.3
Diclofenac	nd	88	9.0

nd: non-detected or below the limits of detection.

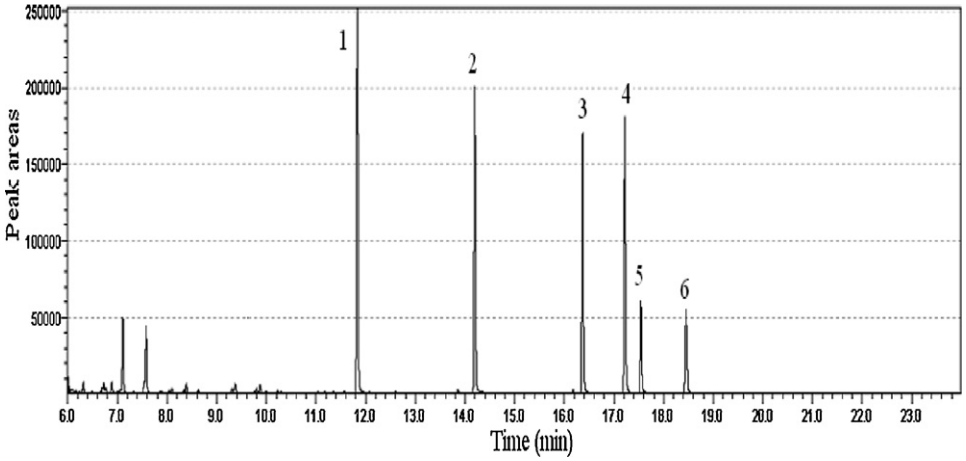


Fig. 9. Chromatogram of extractant of a spiked drain water sample under the most favorable extraction conditions, as given in the text. (1) Ibuprofen, (2) alprenolol, (3) naproxen, (4) propranolol, (5) ketoprofen, and (6) diclofenac.

and continuous SPE-GC–MS [6]. Compared to the last two mentioned techniques in which the sample solutions were delivered by pumps through the extraction devices, the proposed method was easier to perform.

3.5. Genuine water sample analysis

The method was applied to the analysis of drain water collected in the university campus. Samples were extracted as they were, without any pretreatment.

Ibuprofen, naproxen, propanolol, and ketoprofen were found in the samples (results listed in Table 4), while alprenolol and diclofenac were not detected, indicating in that either they were not present or their concentrations were below the LODs.

Furthermore, these genuine samples were spiked to a level of 10 μg/L of each compound and processed to assess matrix effects. Table 4 shows the relative recoveries, defined as the ratios of the peak areas of the analytes in the spiked genuine samples and the peak areas of the analytes in the spiked ultrapure water. It can be seen that the relative recoveries ranged from 88% to 105% for all analytes. This demonstrated that the drain water matrix had insignificant, if any, effect on the procedure. As an example, Fig. 9 shows a chromatogram of an extract of a spiked drain water sample, which was extracted using the present method under the most favorable conditions as described previously. The developed SBME with derivatization offers a suitable method for the determination of PhACs at trace level concentrations in genuine water samples.

4. Conclusion

A novel, simple, and fast method, combining simultaneous solvent bar microextraction and derivatization, was developed for the determination of pharmaceutically active compounds in water samples.

In this approach, the derivatization reagent (MTBSTFA) was added to the organic solvent (acceptor phase), so that the derivatization could occur simultaneously with the extraction. The extract could be directly injected into the GC–MS system for analysis. In the conventional way, the derivatization would be an extra step, applied after the extraction.

The random movement of the SBME in the sample solution facilitated the extraction, and conceivably also the derivatization reaction, aided by the temperature (60 °C) applied.

In comparing SBME with SPME, both of which gave comparable analytical results, the former overcame some shortcomings of SPME such as fiber fragility and carry-over, and prominently, the extraction time for SBME with derivatization (20 min) was much less than that of SPME (60 min extraction and 20 min derivatization). The present procedure is also cost-effective, relying only on affordable and easily accessible hollow fiber membranes.

With the proposed method, good LODs (as low as 0.006 μg/L) and linearity, and acceptable repeatability were achieved. SBME with simultaneous derivatization, in conjunction with GC–MS analysis, was demonstrated to be a fast and efficient method for the determination of pharmaceutically active compounds in drain water.

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