Article

Simultaneous Determination of Environmental Contaminants with Different Volatilities in Tap Water Samples Using a New Approach to Single-Drop Microextraction Procedure

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A new single-drop microextraction approach is proposed for the extraction of compounds with a wide range of volatilities from tap water samples with separation/detection by gas chromatography-mass spectrometry. This new microextraction mode is called direct immersion-headspace-single drop microextraction (DI-HS-SDME). Trihalomethanes (THMs), hydrocarbons with low molecular weight (benzene, ethyl benzene and xylenes) and polycyclic aromatic hydrocarbons (PAHs) were the model compounds. Ten milliliters of tap water samples and drop volume of 1.5 μL of 1-octanol were used. The optimal condition for DI-HS-SDME mode was 80 min of total extraction time (48 min at 40 °C in DI-SDME mode and 32 min at 12 °C in HS-SDME mode) and the addition of 1.2 g of NaCl. The analytical figures of merit were evaluated, the limits of detection ranged from 0.03 μg L^{-1} for *o*-xylene and ethyl benzene to 6 μg L^{-1} for THMs. The proposed method represents a promising alternative for the analysis of aqueous matrices which contain compounds with very different ranges of volatility.

Keywords: environmental contaminants, THM, PAH, microextraction

Introduction

Sample preparation is a crucial part of chemical/biological analysis and can be considered an important step of the analytical process. The main objectives associated with sample preparation are the removal of potential interferents, analyte preconcentration and the possibility of derivatization of the analytes into a more detectable form can also be achieved.[1] For organic trace analysis, this step mainly comprises extractions, which serve to isolate compounds of interest from the sample matrix. The concentration of target compounds is enhanced (sample enrichment) and the presence of matrix components is reduced (sample cleanup).[2] Several methods have been developed to perform efficient sample preparation prior to chromatographic analysis. Firstly, the so-called classical sample preparation methods were developed, notably liquid-liquid extraction (LLE) and solid-liquid extraction (SLE). These sample preparation methods have high extraction capacity; however, some disadvantages are associated with these techniques, such as the use of large amounts of toxic solvents, which is particularly negative

regarding environmental concerns in almost all activities. In some cases, using these classical techniques long extraction times are often required.

Due to these factors, sample preparation techniques which allow efficient extraction with low solvent consumption are welcome. In this regard, miniaturized sample preparation techniques have been developed. One of the most widespread of these techniques is the solid-phase microextraction (SPME), developed by Arthur and Pawliszyn,[3] and introduced in 1990. Years later, the liquid-phase microextraction (LPME) was introduced to the scientific community.[4-6] One variant of LPME consists of placing a very small amount (generally a drop) of an extracting solvent, on the tip of a microsyringe needle or a small polytetrafluoroethylene (PTFE) rod. This drop can be directly immersed (DI mode) into the sample or its headspace (HS mode); this approach is called single-drop microextraction (SDME). After a pre-determined extraction time, the solvent is withdrawn into the microsyringe and conducted to an analytical system. SDME is similar to solid-phase microextraction, where the preconcentration and extraction steps occur simultaneously. The SDME technique provides analyte extraction with a few microliters of an organic solvent,[7,8] and the major advantages associated

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with the SDME are the simplicity of application, low cost and high extraction and cleanup capacity.[9] SDME also has a wide range of applicability and has been used for determination of several types of analytes including pesticides,[10-16] organic acids,[17,18] phenols,[19-21] aldehydes,[22] haloacetic acids,[23] amines,[24] amino acids,[25] sulfonamides,[26] and metallic species such as Cd,[27] Sb,[28] Cr[VI],[29] and Se.[30] One of the disadvantages of using the SDME technique, mainly in the direct immersion mode, is the possibility of losses of solvent microdrop during the microextraction procedure. In addition, biological samples, such as plasma, may emulsify substantial amounts of organic solvents, and this may even enhance the instability problem.[31]

The extraction mode in both SPME and SDME techniques is selected due to characteristics of analyte/matrix. In general, for aqueous samples containing analytes with low volatility, such as polycyclic aromatic hydrocarbons (PAHs),[32] the DI mode presents higher efficiency. However, to analyze samples containing more volatile compounds, such as trihalomethanes (THMs)[33] or benzene, toluene, ethyl benzene, and xylene (BTEX),[6] the HS mode generally provides higher extraction efficiency.

The compounds known as PAHs and BTEX are among the most carcinogenic, mutagenic and toxic found in aquatic systems. There are several sources of these compounds in the environment, the main one being associated with spills involving the release of petroleum products like gasoline, diesel fuel, and lubricants, among others.[34,35] Trihalomethanes are compounds which can be formed by the reaction of chlorine with naturally occurring organic matters, principally humic acid and fulvic acid. This chlorine is applied to drinking water in order to deactivate microorganisms and/or to ensure the residual concentrations in drinking water distribution systems. THMs are all considered to be possible carcinogens and, therefore, human exposure to such compounds should be minimized.[36]

For samples containing different classes of compounds with a wide range of volatilities, the choice of only one microextraction mode (HS or DI) can decrease substantially the extraction efficiency for some compounds. Therefore, it is very complicated the achievement of efficient extractions for all compounds (volatiles and less volatiles) in samples containing analytes with huge differences in their volatilities/polarities.

In studies involving SPME, a new microextraction approach using both modes (DI-HS) in the same extraction procedure has been investigated. Using this approach, excellent results were reached for the extraction of compounds with a wide range of volatilities,[35,37] leading to the achievement of an extraction condition which allowed

good extraction capacity for all analytes (volatiles and less volatiles) studied. According to the best of our knowledge, the DI-HS mode used in the SDME procedure has not been previously reported. Therefore, the aim of this research was to propose an efficient single-drop microextraction approach for aqueous samples containing environmental contaminants with large differences in their volatilities, using both DI and HS modes (DI-HS-SDME) sequentially implemented in the same microextraction procedure. In this study, concentrations of THMs, benzene, ethyl benzene, *o-, m-, p*-xylene and PAHs in tap water samples (collected in Florianópolis, Santa Catarina State, Brazil) were determined. Multivariate optimizations were performed to determine the ideal extraction conditions for each mode. In addition, a comparison between the extraction efficiencies for the HS, DI and DI-HS modes was carried out.

Experimental

Instrumentation

A Shimadzu gas chromatograph (GC) equipped with a mass spectrometry (MS) detector (Kyoto, Japan) GC-MS QP-2010 Plus and chromatographic column Restek Rtx®-5MS (30 m × 0.25 mm × 0.25 μm) obtained from Restek Corporation (Bellefonte, PA, USA) were used in this study. The oven temperature program was: 40 °C (held for 10 min), 20 °C min⁻¹ to 80 °C and 6 °C min⁻¹ to 300 °C (held for 3 min). The injector temperature was 260 °C, the temperatures of the ion source and the interface were 270 and 260 °C, respectively. Ultra-pure helium was used as the carrier gas with a flow of 1 mL min⁻¹. The injection was performed in the split mode at a ratio of 1:10 and the mass spectrometer was operated in electron impact (EI) ion source mode at 70 eV. The analysis in selected ion monitoring (SIM) mode was carried out using the following *m/z* ratios for the quantification: chloroform (83), bromodichloromethane (83), dibromochloromethane (129), bromoform (173), benzene (78), ethyl benzene (91), *p-, m*-xylene (91), *o*-xylene (91), acenaphthylene (152), fluorene (166), phenanthrene (178), anthracene (178), pyrene (202), benzo(*a*)anthracene (228) and chrysene (228).

Materials

In this study, 15 mL vials (Supelco, Bellefonte, PA, USA) with PTFE/silicon septa, two thermostatic baths (New Technique, São Paulo, Brazil and Microquimica Ind. Com. Ltda., Palhoça, SC, Brazil), magnetic stirrers and an analytical balance (Mars Trade, Analytical Instrumentation, São Paulo, Brazil) were used. Also a 10 μL gastight®

microsyringe 1700 series (Hamilton, Reno, NV, USA) was employed to perform the single-drop microextractions.

Reagents and solutions

Sodium chloride P.A. (Vetec, Rio de Janeiro, Brazil), extraction solvents 1-octanol, toluene and *n*-hexane (Vetec), ultra-purified water (Mega Purity, Billerica, MA, USA), standard solutions of polycyclic aromatic hydrocarbons in acetone containing acenaphthylene, fluorene, phenanthrene, anthracene, benzo(*a*)anthracene, chrysene and pyrene (Supelco) at a concentration of 500 μg mL^{-1} were used in this study. Also, THM standard solutions containing chloroform, bromodichloromethane, dibromochloromethane and bromoform at a concentration of 200 μg mL^{-1} in methanol (Supelco) were employed. A standard solution containing benzene, ethyl benzene, *o-, p-, m*-xylene at a concentration of 2000 μg mL^{-1} in methanol (Supelco) was also used.

Sample collection

Tap water samples were collected directly from the laboratory at the Department of Chemistry, Federal University of Santa Catarina, located in the city of Florianópolis, Santa Catarina State, Brazil, and analyzed immediately after collection.

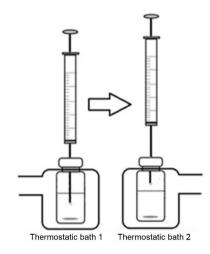## Selection of extraction solvent and drop volume for SDME procedure

In the procedure for the selection of the extraction solvent the following organic solvents were investigated: hexane, toluene and 1-octanol. Aqueous solutions containing 200 μg L^{-1} of all analytes were submitted to microextractions with 1.0 μL of each solvent. The HS and DI modes were used to verify the solvent which presented the best extraction performance. The stability of the microdrop using extraction time of 40 min and extraction temperature of 20, 30 and 45 °C was evaluated. Ten milliliters of ultra-pure water were used for all microextraction procedures.

After choosing the extraction solvent, the microdrop volume was optimized using 0.5, 1.0, 1.5 and 2.0 μL, also 10 mL of ultra-pure water were employed. In this optimization procedure, the extraction conditions for the HS mode were a temperature of 20 °C and extraction time of 20 min; for the DI mode the extraction conditions were 40 °C and 30 min of extraction. For this both optimizations, the geometric mean for all analytes was used as response and the microextractions were performed in triplicate.

Single-drop microextraction apparatus

SDME procedures were performed in three modes (HS, DI and DI-HS) using 10 mL of water sample. For the conventional HS and DI modes only one thermostatic bath was used with the temperature adjusted according to desired extraction conditions. In this procedure, after reaching the required extraction time, the 1-octanol microdrop was withdrawn into the needle of the syringe and transferred to the gas chromatograph injector. Magnetic stirrers were used to perform the microextractions; in the direct immersion mode a stirring rate of 250 rpm was employed; in regard to headspace extractions a rate of 700 rpm was used.

For the DI-HS approach, two thermostatic baths were placed side by side to facilitate the transfer of the vial containing the aqueous sample and the microsyringe needle. The scheme of the apparatus used for the DI-HS-SDME procedure is shown in Figure 1.

Figure 1. Scheme of the DI-HS-SDME procedure using a 10 μL microsyringe, firstly the direct-immersion microextraction was performed followed by headspace microextraction (adapted from Jain and Verma).[38]

Firstly, the microextraction in DI mode was performed in the thermostatic bath 1. After the extraction time, a 1.5 μL microdrop of 1-octanol was withdrawn and the vial containing the sample and the microsyringe needle was transferred to the thermostatic bath 2 (at lower temperature). After this, the organic solvent microdrop was exposed again, however at this time, in the sample headspace. After the extraction time, the 1-octanol microdrop was withdrawn into the needle and the solvent content was injected into the gas chromatograph injector.

Multivariate optimizations

To perform the optimizations, the geometric mean of the chromatographic peak areas obtained for the compounds

of interest was considered, the SDME procedure was performed using 1.5 µL of 1-octanol and ultra-pure water was used in all optimization steps. Aqueous solutions containing 20 µg L^{-1} of each analyte were employed. Three central composite designs were performed for the construction of the response surfaces in each optimization, using the software Statistica 8.0 (STATSOFT, Tulsa, OK, USA), 17 experiments were carried out in each case with triplicate in the central point. The following optimizations were performed: (*i*) optimization of extraction conditions for HS-SDME mode: firstly, the optimization to obtain the ideal extraction conditions for the HS-SDME mode was performed. The variables studied were the extraction time (10-60 min), extraction temperature (10-60 °C) and the mass of sodium chloride added (0-3.6 g); (*ii*) optimization of the extraction conditions for DI-SDME mode: optimization was also performed to achieve the ideal extraction conditions for the DI-SDME mode, with the following variables: extraction time (26-94 min), extraction temperature (28-62 °C) and the mass of sodium chloride added (0-3.6 g); (*iii*) optimization of extraction conditions for DI-HS-SDME mode: using the optimized temperatures obtained with the previous optimizations (DI-SDME and HS-SDME) a new central composite design was carried out to verify the ideal conditions for the extraction using the DI-HS-SDME mode. The variables studied were the total extraction time (26-94 min), percentage of the total time in which the extraction was performed in the HS-SDME mode (0-100%) and the mass of sodium chloride added (0-3.6 g).

Comparison among the different extraction modes

A comparison among the extraction efficiencies obtained for the three modes (DI-SDME, HS-SDME and DI-HS-SDME) was performed. For this purpose, extractions using an aqueous solution containing 20 µg L^{-1} of each analyte were carried out. The extractions were performed applying the following conditions: (*i*) DI-SDME: extraction time of 48 min at 40 °C with 0.2 g of NaCl; (*ii*) HS-SDME: extraction time of 32 min at 15 °C with 3.0 g of NaCl; (*iii*) DI-HS-SDME: extraction time of 48 min at 40 °C in the DI mode, followed by 32 min at 15 °C in the HS mode; NaCl mass of 1.2 g.

Analytical parameters of merit and analysis of real samples

The analytical parameters of merit, such as the linearity (R^2), linear range, precision (relative standard deviation, RSD), accuracy (recovery tests), limits of detection (LOD) and quantification (LOQ) were determined. To obtain the calibration curve, the method involving standard additions

into the tap water samples obtained from our laboratory was employed. The tap water samples were spiked with different concentrations of each analyte. For all trihalomethanes analyzed in this work, concentrations of 20.0, 30.0, 70.0, 100 and 200 µg L^{-1} were used. The following concentrations were used for the calibration curves regarding the other compounds; benzene: 0.3, 0.5, 1.0, 5.0 and 10 µg L^{-1}; ethyl benzene: 0.1, 0.5, 1.0, 5.0 and 10 µg L^{-1}; *p-, m*-xylene: 0.5, 1.0, 3.0, 5.0 and 10 µg L^{-1}; *o*-xylene: 0.1, 0.5, 1.0, 2.0 and 5.0 µg L^{-1}. Concerning PAHs the concentrations were as follows. For fluorene, chrysene and pyrene: 1.0, 2.0, 7.5, 10 and 15 µg L^{-1}; for phenanthrene and anthracene: 0.5, 1.0, 2.0, 3.0 and 10 µg L^{-1}; for acenaphthylene: 2.0, 4.0, 7.5, 10 and 15 µg L^{-1} and for benzo(*a*)anthracene: 0.5, 2.0, 7.5, 10 and 15 µg L^{-1}.

The first point of the calibration curve for each analyte was used as the LOQ; the value achieved for LOQ was divided by 3.3 and the LOD was obtained. Similar procedure for the calculation of LOD and LOQ was employed previously.[39,40]

To assess the accuracy, the recovery percentages of the analytes in samples previously spiked with known concentrations of the compounds were determined. In addition, the tap water samples without any spiking were analyzed and the content of the compounds studied was determined using the DI-HS-SDME approach.

Results and Discussion

Single-drop microextraction: choice of extraction solvent and drop volume

The extraction solvent and microdrop volume are considered very important factors to be optimized in a single-drop microextraction procedure. The solvent microdrop must not suffer dissolution or evaporation, in general, nonpolar and low volatility solvents are used.[41] Firstly, the solvent which provided the best extraction efficiency for all analytes was selected and in this regard *n*-hexane, toluene and 1-octanol were tested.

In this study, a new approach using DI and HS in the same single-drop microextraction procedure has been introduced and the solvent selection was based on the extraction efficiency and the stability of the solvent microdrop in both modes (HS and DI). Taking into account these factors, the microextractions using 1-octanol presented the best results and, in addition, the microdrop of 1-octanol showed better stability if compared with toluene and *n*-hexane. The stability and extraction efficiency obtained for 1-octanol is probably due to low water solubility, low vapor pressure, long hydrocarbon chains

which can easily accommodate analyte molecules of low polarity, and the presence of hydroxyl groups, which may be important for stabilizing polar functional groups present in the molecule.[41]

Regarding the 1-octanol microdrop volume employed in this study, the 1.5 μL drop showed better stability and extraction efficiency in comparison with the 0.5, 1.0 and 2.0 μL drops. The use of a 2.0 μL drop led to stability problems, mainly in direct-immersion mode; in this case, the drop became unstable and was often lost in some microextractions. The 0.5 and 1.0 μL drops did not show stability problems but lower extraction efficiencies were achieved if compared to the 1.5 and 2.0 μL drops. Therefore, a solvent drop volume of 1.5 μL of 1-octanol was used to fulfill this study.
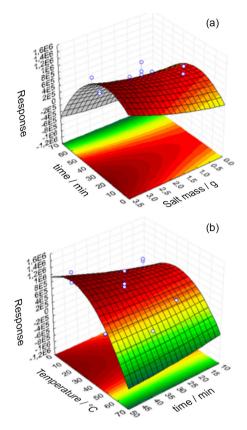
Optimization steps

Optimization of HS-SDME mode

The HS-SDME mode was optimized using a central composite design to determine the ideal extraction conditions in this mode. In this case, the extraction of the compounds with lower volatilities (PAHs with higher molecular mass) was greatly inhibited in the HS mode due to their characteristics being very different if compared to those of more volatile compounds (THMs, benzene, ethyl benzene and xylenes).

Therefore, because of this great difference of extraction efficiency among the different classes of compounds studied in this work, an alternative mode to SDME is welcome. For this aim, the analytes were divided in two groups and the geometric means of the chromatographic peak areas for the first group (THMs, benzene, ethyl benzene and xylenes), which has higher extraction efficiencies in the HS-SDME mode, were used to build response surfaces in order to determine the ideal extraction conditions for this extraction mode. Figure 2 shows the response surfaces obtained for the variables extraction time, extraction temperature and salt mass, considering only the geometric means of chromatographic peak areas obtained for more volatile compounds (THMs, benzene, ethyl benzene and xylenes) in the HS-SDME mode. The geometric mean was used to minimize the influence of large chromatographic peak areas corresponding to one or more compounds.

According to the response surfaces and considering only the more volatile compounds, higher chromatographic peak areas for the HS mode were found using extraction time of 35 min or higher, extraction temperature around 15 °C and NaCl mass of 3.2 g.

As discussed in previous sections, due to different behavior of the analytes, mainly regarding the volatility, the

Figure 2. Response surfaces obtained for HS-SDME, considering only the chromatographic peak areas obtained for the more volatile compounds (THMs and benzene, ethyl benzene and xylenes).

extraction temperature is an important factor to enhance the analytical response. Therefore, the optimized temperature achieved in the HS-SDME mode (15 °C) was also applied to DI-HS-SDME approach.

Optimization of DI-SDME mode

Using another central composite design, the experimental conditions for the DI-SDME mode were also optimized. The data used as the response were the geometric means of the chromatographic peak areas for the studied compounds.

Using the DI-SDME mode, the extraction of the compounds with lower volatilities (less volatile PAHs) was more efficient if compared to those obtained with HS-SDME mode. Based on these results, a similar optimization strategy performed in HS-SDME was used in DI-SDME mode, with the analytes divided into two groups. However, in this case, the geometric means for less volatile compounds (PAHs) were used to build response surfaces. Figure 3 shows the response surfaces obtained for the extraction time, extraction temperature and salt mass. As can be seen in Figure 3, higher chromatographic responses for DI-SDME were achieved using extraction time of 50 min or higher, extraction temperature of around 40 °C and 0.2 g of NaCl. As in the case of HS-SDME, the

variable extraction temperature (40 °C) was used for the
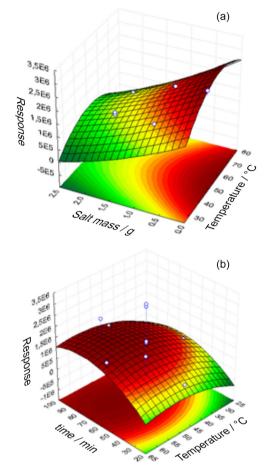DI-HS-SDME procedure.

Figure 3. Response surfaces obtained for DI-SDME mode, considering
only the chromatographic peak areas of the less volatile compounds
(PAHs).

Optimization of DI-HS-SDME mode

The extraction conditions to DI-HS-SDME mode were
also optimized. In the optimization step, the variables total
extraction time, percentage of time in headspace mode and
NaCl mass added into the aqueous samples were studied.
The temperatures used for the direct-immersion (40 °C)
and headspace (15 °C) modes were previously optimized
as described above.

To determine the ideal extraction conditions for
DI-HS-SDME the geometric means of the peak areas of
all studied compounds were selected. Therefore, in this
case, compromise conditions for all studied compounds
(THM, benzene, ethyl benzene, xylenes and PAHs) were
proposed. The response surfaces obtained for all analytes
are shown in Figure 4.

According to the response surfaces obtained in Figure 4,
it can be observed that 80 min of total extraction time with
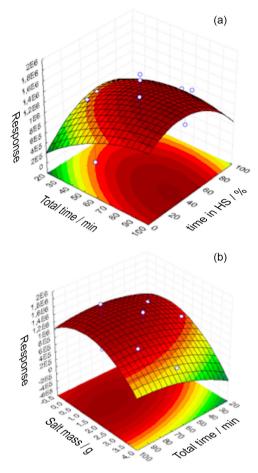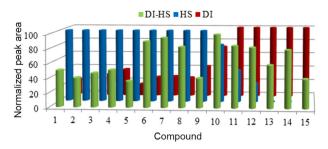40% of this total time (32 min) in the HS mode provided the

Figure 4. Response surfaces obtained for the DI-HS-SDME procedure,
considering the chromatographic peak areas of all compounds studied
(THMs, benzene, ethyl benzene, xylenes and PAHs).

highest extraction efficiency. Regarding the NaCl, a mass
of 1.2 g represented the ideal extraction conditions for all
analytes. These values were used as optimized extraction
conditions for DI-HS-SDME mode and for determination
of the analytical parameters of merit.

Comparison among the SDME modes

The extraction efficiencies of the three single-drop
microextraction modes (DI, HS and DI-HS-SDME) were
investigated in this study. The optimized experimental
conditions for each mode, as previously described, were
used. A bar graph showing the normalized chromatographic
peak areas corresponding to each analyte is shown in
Figure 5.

This bar graph shows the efficiencies of the single-
drop microextraction modes for each compound analyzed
in this study. As expected, the HS mode presented the
best extraction efficiency for more volatile analytes such
as THMs and benzene, ethyl benzene and xylenes; this
mode also presented higher extraction efficiency for

Figure 5. Extraction efficiencies for all compounds analyzed in HS, DI and DI-HS modes: (1) chloroform; (2) bromodichloromethane; (3) dibromochloromethane; (4) bromoform; (5) benzene; (6) ethyl benzene; (7) *p-, m*-xylene; (8) *o*-xylene; (9) acenaphthylene; (10) fluorene; (11) phenanthrene; (12) anthracene; (13) pyrene; (14) benzo(*a*)anthracene; and (15) chrysene.

acenaphthylene. However, for less volatile compounds the HS mode was not so efficient, and for PAH compounds heavier than pyrene (compound number 13 in Figure 5) no chromatographic peak area was achieved in this mode. Regarding the DI mode, it can be observed that this approach allowed higher values for the chromatographic peak areas for the heavier compounds (PAHs); however, for lighter compounds (THMs, benzene, ethyl benzene and xylenes) this mode presented lower extraction efficiencies. Considering the different extraction capacities of each mode, the DI-HS-SDME mode emerges as an interesting alternative for samples which contain classes of compounds with a wide range of volatilities. It can be observed in the bar graph (Figure 5) that the new approach can be applied

to all of the different classes of compounds with good extraction efficiency observed, particularly for benzene, ethyl benzene, xylenes and the lighter PAHs.

According to this comparison, the application of DI mode presented poor analytical responses for the extraction efficiency of volatile compounds; on the other hand, lower analytical responses were achieved for the heavier compounds with the application of only the HS mode. Therefore, this new approach, combining DI and HS sequentially in the same microextraction procedure (DI-HS-SDME), offers the important advantage of achieving "compromise conditions" for analytes with different behaviors. This factor emerges as a good alternative to samples containing several classes of analytes with different volatilities, and at same time, this approach allowed the determination of all studied compounds in a single microextraction procedure.

Analytical parameters of merit for the DI-HS-SDME mode

The analytical parameters of merit mentioned in the experimental section were evaluated. Also, recovery tests on tap water samples spiked with known concentrations of each analyte were performed, considering the lower and higher level of the calibration curve. All microextraction procedures were performed in triplicate and the arithmetic mean of the chromatographic peak area was used as response. The analytical parameters of merit obtained using the new DI-HS-SDME mode is shown in Table 1.

Table 1. Analytical parameters of merit obtained using the new DI-HS-SDME approach

Compound	Linear range / (μg L^{-1})	R^2	LOD / (μg L^{-1})	LOQ / (μg L^{-1})	RSD at lower level / %	RSD at higher level / %	Recovery at lower level / %	Recovery at higher level / %
Chloroform	20-200	0.9940	6	20	17	12	108	98
Bromodichloromethane	20-200	0.9979	6	20	18	6	97	99
Dibromochloromethane	20-200	0.9976	6	20	15	9	75	96
Bromoform	20-200	0.9911	6	20	10	2	125	101
Benzene	0.3-10	0.9898	0.09	0.3	19	13	97	99
Ethyl benzene	0.1-10	0.9934	0.03	0.1	5	9	71	101
p-, m-Xylene	0.5-10	0.9784	0.15	0.5	4	22	84	96
o-Xylene	0.1-5	0.9767	0.03	0.1	2	13	86	93
Acenaphthylene	2-15	0.9968	0.6	2	18	4	65	100
Fluorene	1-15	0.9699	0.30	1	25	17	118	94
Phenanthrene	0.5-10	0.9990	0.15	0.5	12	13	72	99
Anthracene	0.5-10	0.9998	0.15	0.5	12	12	97	99
Benzo(*a*)anthracene	0.5-15	0.9851	0.15	0.5	5	25	82	105
Chrysene	1-15	0.9725	0.30	1	2	15	84	108
Pyrene	1-15	0.9943	0.30	1	12	8	104	98

LOD: Limit of detection; LOQ: limit of quantification; RSD: relative standard deviation. Lower levels: THM, 20 μg L^{-1}; benzene, ethyl benzene and xylenes, 0.5 μg L^{-1}; and PAH, 2 μg L^{-1} (n = 3); higher levels: THM, 200 μg L^{-1}; benzene, ethyl benzene and xylenes, 5 μg L^{-1}; and PAH, 10 μg L^{-1} (n = 3).

As can be observed in Table 1, acceptable analytical parameters of merit for the extraction of THM, benzene, ethyl benzene, xylenes and PAHs from the tap water samples using the proposed DI-HS-SDME mode were obtained. The RSD varied from 2 to 25% and the recovery for spiked samples from 65 to 125%. The linear range obtained for the THMs varied from 20 to 200 µg L^{-1} and the limits of detection achieved were 6 µg L^{-1} for the four compounds of this class studied, with R^2 varying from 0.9911 to 0.9979. These results are similar to those obtained by Maia *et al.*,[42] who determined these compounds in swimming pool samples by SPME-GC with electron capture detection (ECD). With regard to the benzene, ethyl benzene and xylenes, the linear range obtained varied from 0.1 to 10 µg L^{-1} with R^2 values in the range of 0.9767 to 0.9972 and limits of detection of 0.03 to 0.15 µg L^{-1}. For the PAHs the linear range was between 0.5 and 15 µg L^{-1} with R^2 varying from 0.9699 to 0.9998 and the limit of detection from 0.30 to 0.60 µg L^{-1}. The results obtained with the proposed DI-HS-SDME procedure for these two classes of compounds are similar to those obtained by Bianchin *et al.*[35] for the determination of BTEX and PAHs in aqueous samples by SPME/GC-MS.

Both SPME and SDME sample preparation techniques are considered efficient and offer reduced environmental impact compared with classical sample preparation methods. However, the SPME technique, if compared to SDME, is associated with higher analysis costs. In addition to the extraction efficiency, low cost analysis is an important and desirable characteristic to make the analytical method suitable and widespread. Therefore, the proposed mode presented in this work (DI-HS-SDME) is highlighted because of its very low analysis costs and the good results obtained for the determination of environmental contaminants with wide range of volatilities in one single chromatographic determination.

Analysis of real samples

Tap water samples collected directly from our laboratory were analyzed applying the DI-HS-SDME approach and Table 2 shows the results obtained. A chromatogram obtained from tap water sample spiked with 20 µg L^{-1} for THM, 5 µg L^{-1} for benzene, ethyl benzene and xylenes and 10 µg L^{-1} for PAH is shown in Figure 6a. Also, a chromatogram obtained for real tap water samples (without spiking) is shown in Figure 6b.
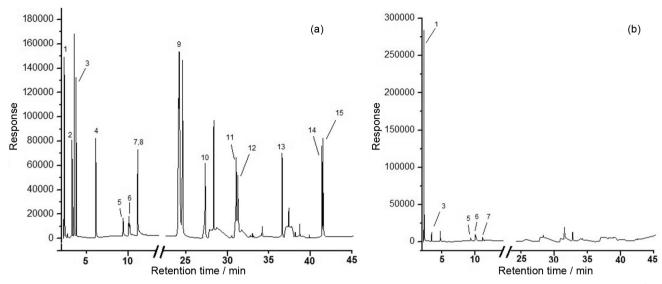
As can be seen in Table 2, some contaminants were found in tap water samples submitted to DI-HS-SDME procedure and chloroform presented the highest concentrations. In both samples, concentrations around 40 µg L^{-1} were obtained for

Table 2. Results obtained for the analysis of the tap water samples

Compound	Found concentration tap water sample 1 / (µg L^{-1})	Found concentration tap water sample 2 / (µg L^{-1})
Chloroform	47 ± 2.5	42 ± 2.2
Bromodichloromethane	DE	DE
Dibromochloromethane	ND	ND
Bromoform	ND	ND
Benzene	ND	ND
Ethyl benzene	DE	DE
p-, m-Xylene	DE	DE
o-Xylene	DE	DE
Acenaphthylene	ND	ND
Fluorene	ND	ND
Phenanthrene	ND	ND
Anthracene	ND	ND
Benzo(*a*)anthracene	ND	ND
Chrysene	ND	ND
Pyrene	ND	ND

DE: Detected, however, at concentration below the LOQ; ND: not detected.

this THM. The presence of this compound can be explained by the use of chemical treatment with free chlorine in order to inactivate some pathogenic microorganisms in water. Free chlorine can react with organic matter present in the water and generate compounds such as THMs. Another THM (bromodichloromethane) was also found in the analyzed samples, however, at concentrations below the LOQ. The United States Environmental Protection Agency (US EPA) and the European Union (EU) have set limits of 80 and 100 µg L^{-1}, respectively, for the sum of chloroform, bromodichloromethane, dibromochloromethane and bromoform compounds in drinking water.[43]

Ethyl benzene, *o*-, *m*-, *p*-xylene were also detected. These compounds are also characterized as environmental contaminants and thus the US EPA has established maximum contaminant levels (MCLs) for benzene (5 µg L^{-1}), ethyl benzene (700 µg L^{-1}) and xylenes (10,000 µg L^{-1}) in drinking water. An MCL value of 1.0 µg L^{-1} for benzene in drinking water has been established by European legislation.[44,45] In the studied tap water samples benzene, ethyl benzene and xylenes levels were below the LOQ. According to the regulatory agencies and to the contaminant levels found using the DI-HS-SDME approach, the levels of the THMs, benzene, ethyl benzene and xylenes in these tap water samples are below the established MCLs. For some PAH such as anthracene, the EU[46] established maximum allowed concentration of 0.1 µg L^{-1}. However, in the analysis of these real samples the studied PAH were not detected.

Figure 6. Chromatograms obtained by GC-MS-SIM from tap water samples. (a) Chromatogram obtained from tap water sample spiked with 20 μg L^{-1} of THM, 5 μg L^{-1} for benzene, ethyl benzene and xylenes; and 10 μg L^{-1} of PAH. (b) Chromatogram obtained from real tap water sample (without spiking). The elution order is as follows: (1) chloroform; (2) benzene; (3) bromodichloromethane; (4) dibromochloromethane; (5) ethyl benzene; (6) *p-, m*-xylene; (7) *o*-xylene; (8) bromoform; (9) acenaphthylene; (10) fluorene; (11) phenanthrene; (12) anthracene; (13) pyrene; (14) benzo(*a*)anthracene; and (15) chrysene.

Conclusions

The new approach to single-drop microextraction using direct-immersion and headspace extraction in the same microextraction procedure showed good performance for determination of environmental contaminants in water samples. This approach allowed the analysis of compounds with a wide range of volatilities in a single assay. According to the multivariate optimization, this procedure allowed to achieve a "compromise condition" for the extraction of all analytes. This extraction technique represents an excellent alternative to the traditional single-drop microextraction procedure, combining efficient extraction, low analysis costs and environmentally friendly characteristics. The comparison among the microextraction modes showed that DI-HS-SDME presented capacity of extraction of all analytes in a single assay with relatively good analytical responses if compared to DI and HS used separately. Despite the use of one additional experimental step, this proposed approach has an interesting extraction capacity and it can be explored on multiresidual analysis of different contaminants from aqueous samples, taking into account that several multiresidual methods explore analytes with completely different behavior regarding their volatility.

Acknowledgments

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