CHEMIJA
2024. Vol. 35. No. 1. P. 17–24

ISSN 0235-7216 eISSN 2424-4538
DOI: https://doi.org/10.6001/chemija.2024.35.1.3

Switchable hydrophilicity solvent-based hollow fibre liquid-liquid microextraction – headspace gas chromatography for determination of benzene and its derivatives in water samples

Vida Vičkačkaitė*,

Vasarė Viselgaitė,

Vilius Poškus

Department of Analytical and Environmental Chemistry, Vilnius University, 24 Naugarduko Street, 03225 Vilnius, Lithuania

A novel, cost-effective, simple and environmentally friendly method was developed for separating, preconcentrating and quantifying volatile analytes. That approach utilised switchable hydrophilicity solvent-based hollow fiber liquid-liquid microextraction in combination with headspace gas chromatography. The extraction was carried out from 1 l of water solution. 100 µl of nonanoic acid, immobilised within a polypropylene capillary, was selected as an extraction solvent. After the extraction, the capillary with the extract was transferred to a headspace vial, and headspace gas chromatographic analysis was performed. To facilitate the transition of volatile analytes to the headspace for subsequent analysis, nonanoic acid was converted to hydrophilic nonanoate by adding a sodium hydroxide solution.

The effectiveness of the suggested strategy was demonstrated in the determination of benzene and its derivatives in water samples. Various parameters affecting the extraction and headspace gas chromatographic determination were investigated and optimised. Under the optimal conditions, the analytical characteristics of the proposed technique were determined.

Using this approach, the limits of quantification for all analytes were found to be below the maximum acceptable concentration.

Keywords: headspace gas chromatography, hollow fiber liquid-liquid microextraction, switchable hydrophilicity solvents, benzene derivatives

INTRODUCTION

Headspace gas chromatography (HS-GC) has become one of the key techniques for analysing volatile compounds. This method simplifies sample preparation, allowing the selective analysis of volatile analytes and reducing the risk of matrix interference by introducing only the volatile compounds from the headspace into the GC system [1, 2]. However, to detect low concentrations, the pre-concentration of analytes is essential before HS-GC analysis. Hollow fiber liquid-liquid microextraction (HF-LLME) proposed by Pedersen–Bjergaard and Rasmussen in 1999 can be employed for this purpose [3]. A typical HF-LLME setup comprises a porous hollow fiber membrane, usually made of polypropylene, impregnated and filled with an organic solvent [4]. HF-LLME has several benefits, such as cost-effectiveness, an efficient cleaning and a high pre-concentration

* Corresponding author. Email: vida.vickackaite@chf.vu.lt

factor [5, 6]. Polypropylene hollow fibers are commercially available at a low cost, making them disposable after a single extraction, thereby minimising the risk of cross-contamination. The only attempt described in the literature to combine HF-LPME with HS-GC focuses on the determination of hexanal in a fat-rich food [7].

The organic solvent within the hollow fiber must exhibit a high extraction efficiency for the analytes. On the other hand, when performing a subsequent headspace analysis, it is crucial to transfer analytes from a solvent to the headspace as completely as possible. This becomes challenging when the analytes are highly soluble in the solvent.

Here, we propose resolving these two contradictory requirements for the solvent by employing switchable hydrophilicity solvents (SHSs).

SHSs can be converted from their hydrophobic into their hydrophilic forms, or vice versa [8]. The solvents have found various applications in different fields, and since 2015, their use in microextraction has been on the rise [9]. Several formats of SHS-LLME, such as air-, vortex- and ultrasound-assisted SHS-LLME, have been suggested [10]. However, to our knowledge, SHSs have not been combined with HF-LLME until now.

In our current research, we have developed a switchable hydrophilicity solvent-based HF-LLME strategy and hyphenated it to headspace gas chromatography. To illustrate the efficiency and practicality of this proposed HF-LLME-HS-GC strategy, we selected benzene and its derivatives as the model analytes.

EXPERIMENTAL

Materials and solutions

Octanoic acid (≥98%), nonanoic acid (≥97%) and bromobenzene (≥99.5%) were purchased from Sigma-Aldrich. The following analytes were purchased from Fluka: 1 – benzene (≥99%), 2 – toluene (≥99%), 3 – etylbenzene (≥99%), 4 – p-xylene (analytical standard), 5 – m-xylene (analytical standard), 6 – cumene (analytical standard), 7 – o-xylene (analytical standard), 8 – chlorobenzene (≥99.5%), 9 – 1,3,5-trimethylbenzene (analytical standard). Those analytes were purchased from Sigma-Aldrich: 10 – styrene (≥99%), 11 – 1,2,4-trimethylbenzene (≥98%), 12 – 1,2,3-trimethylbenzene (technical grade). Stock solutions of the analytes were prepared at a concentration of 100 mg/ml in acetone and stored at 4°C before use. Working solutions, encompassing a combination of analytes in acetone, were then derived from the stock solutions and further diluted with acetone or with nonanoic acid to achieve the desired concentration. Bromobenzene (2 mg/ml solution in acetone) was used as an internal standard (IS).

Instrumentation and conditions

Headspace gas chromatographic analysis was performed on a PerkinElmer Clarus 580 series gas chromatograph (PerkinElmer, USA) equipped with a flame ionisation detector (temperature 250°C, hydrogen flow 40 ml/min, air flow 400 ml/min). The GC system was equipped with the Zebron ZB-WAXPLUS capillary column (30 m × 0.32 mm id, 1 μm film thickness) (Phenomenex).

Headspace extraction and sample introduction was performed on a PerkinElmer Headspace Sampler Turbomatrix 16 (PerkinElmer, USA) equipped with a balanced pressure system. Twenty millilitre headspace vials were used in all experiments. A headspace vial was positioned in an HS autosampler and equilibrated at selected temperature. The needle temperature and the transition line temperature was 110°C. The settings of the headspace sampler were 1 min for pressurization and 0.06 min for injection. Helium was employed as carrier gas with 16.7 psi column head pressure. The injector temperature was held at 110°C. The oven temperature was programmed as follows: 60°C for 1 min, from 60 to 76°C at 2°C/min, from 76 to 140°C at 10°C/min, from 140 to 200°C at 45°C/min and held for 3 min.

Procedures

For HF-LLME the Accurel Q 3/2 polypropylene capillary with a 200 μm wall thickness, 0.2 μm pore size and 600 μm inner diametre (Membrana (Wuppertal, Germany)) was used. The capillary was cut into 27 cm long pieces and was filled with 100 μl of nonanoic acid using a 100 μl syringe. After extraction, the content of the capillary was rinsed into a headspace vial using 1 ml of 1 M NaOH.

RESULTS AND DISCUSSION

Solvent selection

When extracting from aqueous solutions, it is crucial to employ water-immiscible hydrophobic

solvents. In this study, we investigated two hydrophobic solvents, namely octanoic acid and nonanoic acid.

Initially, the efficiency of analytes transfer from the solvents to the headspace was investigated. For this, 10 µl of a solution containing benzene and its derivatives in acetone (20 mg/ml each), along with 100 µl of the respective solvent, were introduced into a headspace vial. The vial was subsequently heated for 10 min at 80°C, and the resulting gas phase was injected into a gas chromatograph with an injection time of 0.01 min. The migration of analytes from the solvents to the headspace showed considerable similarities, but noticeable differences were observed among the analytes, as ilustrated in Fig. 1. This variation can be attributed to the interplay between the volatility of the target compounds and their solubility within the matrix, affecting their transition into the headspace. The solvents employed in this study demonstrated significant hydrophobic characteristics (logP for octanoic acid and nonanoic acid is 3.05 and 3.42, respectively). Consequently, the transition of analytes from those solvents to the headspace was inversely related to the hydrophobicity of the analytes. Benzene, the most volatile (boiling point 80.1°C) and the least hydrophobic (logP 2.13) compound, exhibited the largest peak area. In contrast, the transition efficiency was notably diminished for the least volatile and the most hydrophobic trimethylbenzenes with boiling points ranging from 164 to 176°C and logP values ranging from 3.42 to 3.78.

Reducing the hydrofobicity of the matrix would diminish the affinity of hydrophobic analytes for the matrix thereby promoting their transfer to the headspace. Octanoic acid and nonanoic acid are switchable hydrophilicity solvents, hydrophobic under acidic conditions and hydrophilic under alcaline conditions [11]. In this study, those acids were transformed into their hydrophilic sodium salts through the addition of sodium hydroxide. Subsequently, the efficiency of analyte transfer into the headspace was reevaluated. For this, 10 µl of an analytes solution in acetone was combined with 100 µl of a solvent, 10 ml of a 0.1 M NaOH solution was added, and HS-GC analysis was conducted under the previously described conditions. Compared to the results obtained without the addition of NaOH (Fig. 1), all analytes exhibited increased peak areas. The benzene peak, however, showed the smallest increase, approximately 1.5 times, which can be attributed to its relatively high solubility in water (1790 mg/l). The impact of alkali addition was notably more pronounced for the more hydrophobic and less volatile analytes. For instance, the areas of the trimethylbenzene peaks increased by up to 10 times. The transfer of

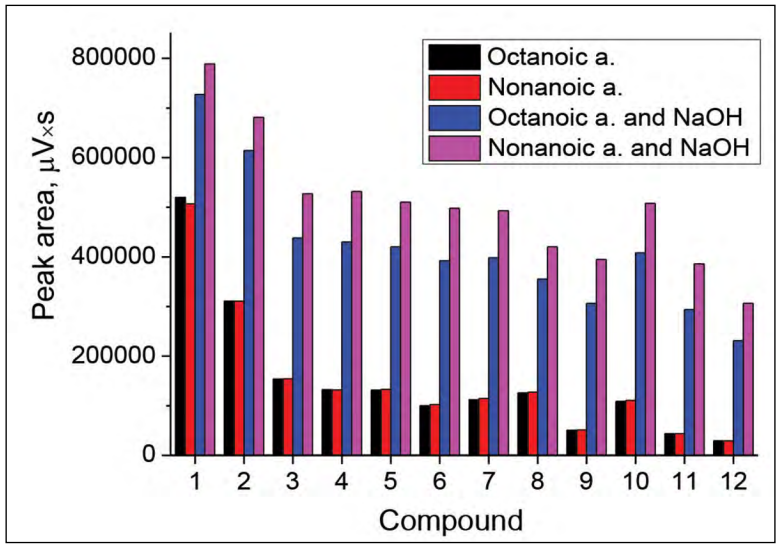

Fig. 1. Efficiency of analyte transfer from different solvents to the headspace without NaOH solution and after addition of 10 ml of 0.1 M NaOH solution. Solvent volume 100 µl, analytes volume 10 µl (20 µg/µl each) heated for 10 min at 80°C, injection time of 0.01 min. For compound identification see 'Materials and solutions'

analytes from nonanoic acid to the headspace was notably more efficient than from octanoic acid. Given this observation, coupled with the lower solubility in water when compared to octanoic acid, nonanoic acid was selected as the preferred solvent for the extraction of analytes from water samples.

For nonanoic acid the impact of a NaOH solution volume on the peak areas was examined. To 100 µl of a solution containing analytes in nonanoic acid (0.4 mg/ml each), 10 ml of a 0.1 M NaOH solution, 5 ml of a 0.2 M NaOH solution, or 1 ml of a 1 M NaOH solution was added. Subsequently, HS-GC analysis was carried out under the previously described conditions.

For all analytes, the largest peak areas were observed after adding 1 ml of a 1 M NaOH solution. Those areas were 2–3 times larger than those after adding 10 ml of the 0.1 M NaOH solution, and 1.2–1.8 times larger than those after adding 5 ml of the 0.2 M NaOH solution. This phenomenon can be attributed to the partial solubility of the analytes in the aqueous NaOH solution. As the solution volume decreased, the dissolved portion of the analytes decreased, leading to the increased concentrations of the analytes in the headspace. That effect was particularly notable for the most water-soluble benzene.

HS-GC conditions

Achieving a significant preconcentration necessitates a substantial sample volume and a minimal quantity of nonanoic acid. When a minute quantity of solvent is spread over the surface of the aqueous phase, its collection becomes challenging. To enhance the recovery of a small amount of nonanoic acid, we chose to implement HF-LLME, incorporating nonanoic acid within a polypropylene capillary.

To optimise the equilibration temperature, a 27 cm polypropylene capillary was filled with 100 µl of nonanoic acid containing 200 µg/ml of each analyte, using a 100 µl syringe. Following this, the capillary was placed into a headspace vial, and the content of the capillary was rinsed using 1 ml of 1 M NaOH. The vial was then heated at 60–95°C for 30 min. Higher temperatures were deliberately avoided to ensure that the equilibration temperature did not surpass the boiling point of the matrix medium. It was observed that the peak areas of the analytes consistently increased with rising temperatures (Fig. 2). Based on the results, the equilibration temperature of 95°C was selected as the optimal choice. At 95°C temperature, the influence of heating time on the peak areas was investigated. The results of HS-GC analysis indicated that the peak areas reached a stable state after heating for 10 to 12 min. Therefore,

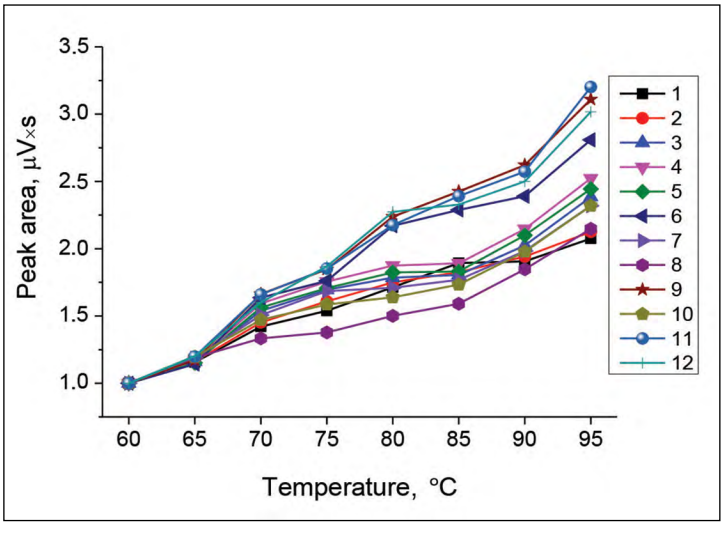

Fig. 2. Effect of the headspace vial heating temperature on the analytes peak areas. 27 cm length polypropylene capillary is felled with 100 µl of nonanoic acid containing 200 µg/ml of each analyte, then the solution washed with 1 ml of 1 M NaOH. Headspace vial (20 ml) heated for 30 min, the injection time 0.01 min. For chromatographic conditions see „Instrumentation and conditions', for compound identification see 'Materials and solutions'

a heating time of 12 min was chosen for further experiments.

The optimisation of the gas phase volume injected into a gas chromatograph was also conducted. The HS-GC equipment used in that work was equipped with pressure balanced sampling, enabling a direct control of the time width of the vapour plug entering the GC column. Injection time widths ranging from 0.01 to 0.12 min were examined. The results indicated (Fig. 3) that the peak areas gradually increased up to an injection time of 0.06 min. Based on that, an injection time of 0.06 min was selected.

HF-LLME from water

To reduce random and systematic sources of uncertainty arising from the sample extraction and HS-GC analysis, an internal standard was added to the sample. Bromobenzene was selected as an internal standard due to its properties closely resembling those of the analytes and its rarity in water samples.

The impact of extraction time on the extraction efficiency was investigated. For that, 100 µl of nonanoic acid was injected into a 27 cm long polypropylene capillary using a 100 µl microsyringe. The capillary was sealed with a clip and immersed in a 1 l water sample containing 100 µl of a bromobenzene solution in acetone (2 mg/ml) and 100 µl of a standard solution of analytes (2 mg/ml each). The extraction device is presented in Fig. 4. The extraction was conducted for 20 to 60 min. Subsequently, the content of the capillary was rinsed into a headspace vial with 1 ml of 1 M NaOH. The capillary was also placed into the headspace vial, and HS-GC analysis was performed under the optimised conditions. After 40 min of extraction, the relative peak areas stabilised, indicating the establishment of

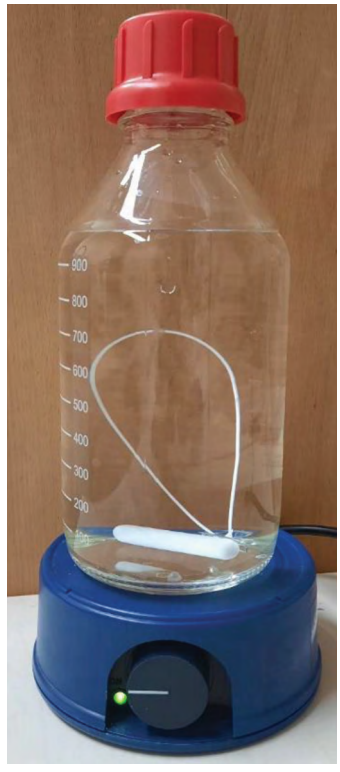

Fig. 4. Illustration of the HF-LLME process

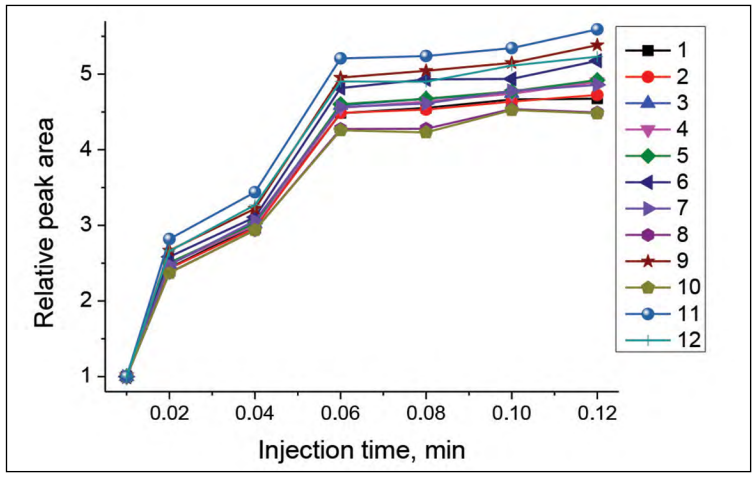

Fig. 3. Effect of the injection time on the analytes peak areas. 27 cm length polypropylene capillary is felled with 100 µl of nonanoic acid containing 200 µg/ml of each analyte, then the solution is washed with 1 ml of 1 M NaOH. Headspace vial (20 ml) is heated at 95°C for 30 min. For chromatographic conditions see 'Instrumentation and conditions', for compound identification see 'Materials and solutions'

equilibrium of the analytes and the internal standard between the aqueous and organic phases.

A HS-GC chromatogram of the extract obtained at the optimised conditions is presented in Fig. 5.

Analytical characteristics and application

The analytical characteristics of the proposed method were determined under the optimised extraction and HS-GC conditions. Calibration curves were constructed using 8 calibration points, each with three replicate injections. The limit of detection (LOD) was defined as the concentration yielding a signal three times the baseline noise. The limit of quantification (LOQ) was defined as the concentration yielding a signal ten times the baseline noise. Relative standard deviations (RSD) were determined by five replicate analyses of the sample at analyte concentrations of 10 and 100 µg/l. The linear concentration ranges, correlation coefficients, LOD, LOQ and RSD are presented in the Table.

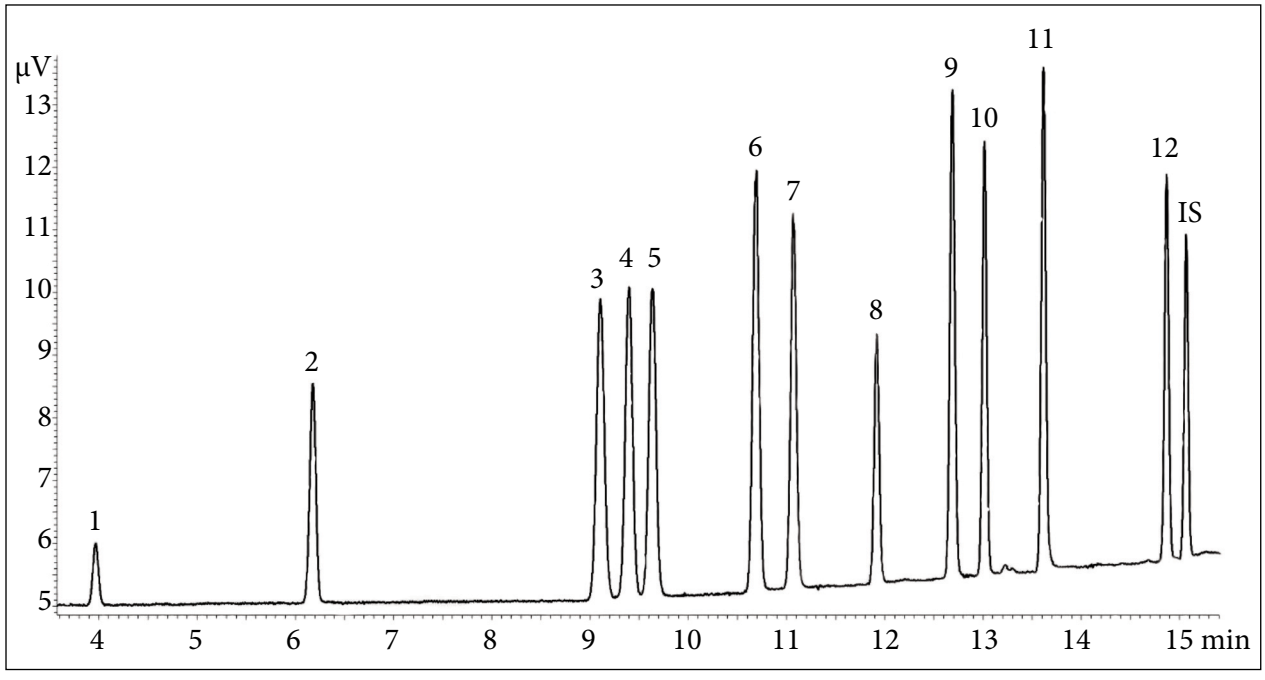

Fig. 5. HS-GC chromatogram of the extract obtained after HF-LLME of 1 L of the water sample containing 200 µg of analytes and bromobenzene (IS). For analytes identification see 'Materials and solutions'. For chromatographic conditions see 'Instrumentation and conditions'

Table. **Analytical performance of the developed procedure**

Analyte	LOD, µg/l	LOQ, µg/l	Linearity up to, µg/l	R^2	RSD, % ($n = 5$)	
					10 µg/l	100 µg/l
Benzene	0.78	2.6	200	0.9903	18.1	13.7
Toluene	0.33	1.1	200	0.9904	17.8	8.7
Etylbenzene	0.39	1.3	200	0.9925	11.8	4.5
p-Xylene	0.39	1.3	200	0.9917	10.4	4.2
m-Xylene	0.40	1.3	200	0.9922	9.9	6.7
Cumene	0.34	1.1	120	0.9915	12.8	3.6
o-Xylene	0.32	1.1	200	0.9904	12.6	3.1
Chlorobenzene	0.41	1.4	200	0.9911	11.9	6.1
1,3,5-Trimethylbenzene	0.29	1.0	120	0.9908	10.0	5.8
Styrene	0.32	1.1	200	0.9915	7.2	4.1
1,2,4-Trimethylbenzene	0.30	1.0	120	0.9912	11.0	6.4
1,2,3-Trimethylbenzene	0.29	1.0	120	0.9907	11.6	5.4

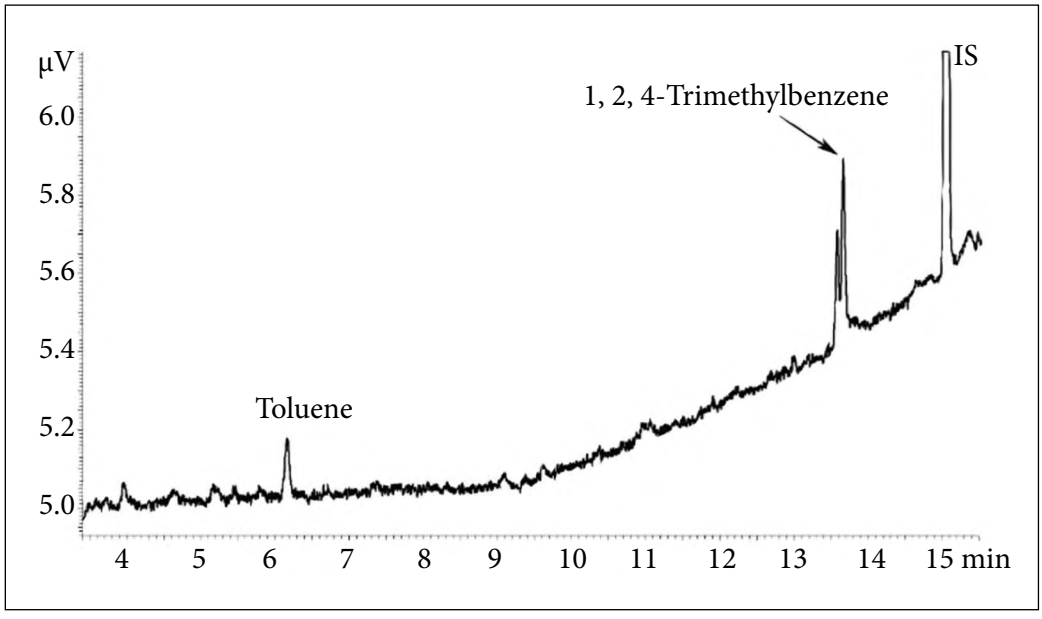

Fig. 6. HS-GC chromatogram of the extract obtained after HF-LLME of 1 L of the water sample from a puddle in the parking lot. For chromatographic conditions see 'Instrumentation and conditions'

For most of the examined analytes, the maximum acceptable concentrations in drinking water vary depending on the country and organisation but are not lower than 100 μg/l, except for benzene (5 μg/l) [12] and cumene (8 μg/l) [13].

Using the suggested strategy, the largest LOQ is observed for benzene (2.6 μg/l) but even in this case it is lower than the maximum acceptable concentration. For other analytes, the maximum acceptabal concentrations are much higher than the LOQs.

The proposed method was employed to analyse surface water. Samples were collected from a puddle in the parking lot and extracted without prior filtration. To a 1 liter aliquot of the water sample, 100 μl of the bromobenzene solution in acetone (2 mg/l) was added. The extraction and determination were carried out under the described conditions. A chromatogram of the extract is presented in Fig. 6. Toluene at a concentration of 2.8 μg/l and 1,2,4-trimethylbenzene at a concentration of 1.5 μg/l were detected in the sample.

CONCLUSIONS

A novel approach for the separation, preconcentration and quantification of volatile analytes was developed, utilising a switchable hydrophilicity solvent-based HF-LLME in conjunction with HS-GC. The strategy involves extracting analytes from water samples into nonanoic acid immobilised in a polypropylene capillary and subsequently converting the hydrophobic nonanoic acid into the hydrophilic nonanoate by adjusting the pH. This transformation allows hydrophobic volatile analytes to readily transfer from the hydrophilic solution into the headspace, where they are determined using HS-GC.

The proposed HF-LLME-HS-GC strategy is characterised by an excellent sample cleanup ability without the need for the pre-filtration of the sample. It is simple and cost-effective due to the utilisation of readily available equipment. The technique achieves an efficient enrichment of analytes from a large sample volume using a small volume of the extracting phase, making it environmentally friendly and economically advantageous. Additionally, the disposal of polypropylene hollow fibers after a single extraction eliminates the risk of cross-contamination.

The efficiency of the suggested strategy was demonstrated in the determination of benzene and its derivatives in water samples. However, the strategy has the potential to be applied to a variety of other volatile organic compounds.

Received 23 February 2024
Accepted 5 March 2024

References

1. C. Camarasu, C. Madichie, R. Williams, *TrAC*, **25**, 768 (2006).
2. N. H. Snow, G. P. Bullock, *J. Chromatogr. A*, **1217(16)**, 2726 (2010).
3. S. Pedersen-Bjergaard, K. E. Rasmussen, *Anal. Chem*., **71(14)**, 2650 (1999).
4. E. Carasek, J. Merib, *Anal. Chim. Acta*, **880**, 8 (2015).
5. M. Rutkowska, J. Plotka-Wasylka, M. Sajid, V. Andruch, *Microchem. J.*, **149**, 103989 (2019).
6. A. Prichodko, M. Mockūnaitė, V. Šmitienė, V. Vičkačkaitė, *Chemija*, **22(3)**, 155 (2011).
7. V. Vickackaite, I. Jurkute, V. Poskus, B. Bugelyte, *Sep. Sci. Plus*, **5(5)**, 163 (2022).
8. P. G. Jessop, D. J. Heldebrant, X. Li, C. A. Eckert, C. L. Liotta, *Nature*, **436(7054)**, 1102 (2005).
9. G. Lasarte-Aragones, R. Lucena, S. Cardenas, M. Valcarcel, *Talanta*, **131**, 645 (2015).
10. U. Alshana, M. Hassan, M. Al-Nidawi, E. Yilmaz, M. Soylak, *TrAC*, **131**, 116025 (2020).
11. H. K. Shih, T. Y. Shu, V. K. Ponnusamy, J. F. Jen, *Anal. Chim. Acta*, **854**, 70 (2015).
12. *National Primary Drinking Water Regulations* [https://www.epa.gov/sites/default/files/2016-06/documents/npwdr_complete_table.pdf].
13. J. P. Herman, L. Redfern, C. Teaf, D. Covert, P. R. Michael, T. M. Missimer, *Int. J. Environ. Res. Public Health*, **17(22)**, 8380 (2020).

Vida Vičkačkaitė, Vasarė Viselgaitė, Vilius Poškus

KEIČIAMO HIDROFILIŠKUMO TIRPIKLIAIS PAREMTA SKYSTAFAZĖ MIKROEKSTRAKCIJA KAPILIARE – VIRŠERDVĖS DUJŲ CHROMATOGRAFIJA BENZENUI IR JO DARINIAMS VANDENINIUOSE MĖGINIUOSE NUSTATYTI

S a n t r a u k a

Lakioms analitims atskirti, sukoncentruoti ir nustatyti sukurtas naujas, ekonomiškas, paprastas ir aplinkai draugiškas metodas. Šis metodas sujungia skystafazę mikroekstrakciją kapiliare, užpildytame keičiamo hidrofiliškumo tirpikliu, ir viršerdvės dujų chromatografiją. Ekstrakcija atliekama iš 1 l vandeninio tirpalo. Kaip ekstrahuojantis tirpiklis naudota nonano rūgštis, imobilizuota polipropileno kapiliare. Po ekstrakcijos kapiliaras su ekstraktu perkeliamas į viršerdvės analizės indelį ir atliekama ekstrakto viršerdvės dujų chromatografinė analizė. Siekiant palengvinti lakių analičių perėjimą į dujinę fazę, nonano rūgštis paverčiama hidrofiliniu nonanoatu, pridedant natrio hidroksido tirpalo. Pasiūlytos strategijos efektyvumas pademonstruotas nustatant benzeną ir jo darinius vandens mėginiuose. Ištirti ir optimizuoti ekstrakcijai ir viršerdvės dujų chromatografijai įtakos turintys parametrai. Optimaliomis sąlygomis nustatytos siūlomos metodikos analitinės charakteristikos. Nustatyta, kad visų analičių nustatymo ribos mažesnės už didžiausias leistinas koncentracijas geriamajame vandenyje.