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Development of a total analytical system by interfacing membrane extraction, pervaporation and high-performance liquid chromatography

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Abstract

This paper discusses the interfacing of continuous membrane extraction, pervaporation and on-line HPLC–UV detection into a total analytical system (TAS). Organics from a water sample were extracted into an organic solvent, and then concentrated via pervaporation prior to HPLC–UV detection. Factors affecting the system performance were studied. With optimized experimental parameters enrichment factors as high as 192 were obtained, the method detection limits were at low ng/mL levels, and the precisions were better than 5%.

Keywords: Membrane extraction; Pervaporation; HPLC–UV detection; Total analytical system; Continuous on-line analysis

1. Introduction

There has been much interest in integrating different analytical functions onto a single platform. The efforts have been mainly confined to the bio-analytical arena, where procedures, such as, cell lysis, extraction, PCR and electrophoresis have been integrated in a micro total analytical system (μ-TAS) [1,2]. These ideas are equally valid for conventional laboratory techniques. Typical approach for inorganic, organic, metals and biological analysis involves extraction and concentration followed by analytical detection. The development of total analytical system (TAS) requires the hyphenation of these steps so that continuous, on-line analysis can be carried out without manual intervention.

Let us take the example of the analysis of semi-volatile organics in water. Liquid–liquid extraction (LLE), solid-phase extraction (SPE) and solid-phase microextraction (SPME) [3–9] are the conventional extraction procedures. Although they have some excellent merits, there exist limitations when it comes to direct interfacing with instruments. Classical LLE is labor intensive, uses large amounts of solvents and is difficult to couple directly to an analytical instrument. LLE coupled with flow injection (FI) analysis was reported first time in the late 1970s [10] and since then it has been developed quickly [11–13]. It minimizes the reagent consumption, and can be carried out continuously followed by on-line or off-line detection. In SPE the analytes are extracted from an aqueous sample onto a solid sorbent, and then eluted with a suitable solvent. It has been automated on-line involving multiple batch processes such as conditioning, washing, and elution [14,15]. SPME, where the analytes are adsorbed onto a fused-silica coated fiber and then desorbed at a high temperature prior to analysis, it is simpler, but only suitable for high concentration analysis due to its low sensitivity. The extracts, especially those from SPE and LLE may need further concentration. Conventional methods for this include nitrogen blowing, rotary evaporator, or Kuderna–Danish concentrators. It is evident that both the extraction and concentration procedures involve several discreet batch operations, and are either time-consuming or labor intensive. Thus automated continuous sampling systems are needed.

In the realm of continuous, on-line extraction procedures, membrane extraction is one of the most promising techniques. It is simple, inexpensive, requires small solvent volumes and offers high enrichment. It allows on-line extraction, and has been coupled to gas chromatography (GC) [16–18], high performance liquid chromatography

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(HPLC) [19,20], mass spectrometry (MS) [21], GC–MS [22] and other analytical instruments [23]. Liquid phase membrane extraction can be classified into supported liquid membrane extraction (SLME) and liquid–liquid membrane extraction (LLME) [24]. SLME is a three-phase system in which the analytes are extracted from an aqueous sample into an acceptor phase via an organic extractant held in the pores of the membrane by capillary force. It is suitable for analyzing highly polar and ionizable compounds. LLME is a two-phase system where the analytes are extracted from an aqueous sample to an organic acceptor. The extraction occurs across a membrane, so that the two phases contact through the membrane pores without direct mixing. LLME can be used in any application as long as the compounds can be extracted into an organic solvent [20,25,26]. The driving force in LLME is the partition of analytes between the aqueous phase and the organic acceptor. The presence of membrane in LLME prevents emulsion formation, and other complex phenomena due to the physicochemical instability of the organic-aqueous interface, which occurs when the two phases are directly contacted, such as LLE.

In membrane pervaporation, a liquid mixture contacts a membrane, the volatile species selectively permeate through, and are removed by a vacuum or an inert stripping gas. It has been used in the analysis of volatile organics by selectively stripping from an aqueous medium [27], and for solvent removal in various industrial applications [28]. In this paper analytical-scale membrane pervaporation was carried out continuously for the removal of solvent from the membrane extraction step. The extract is passed through the lumens of hollow fiber membranes while a counter-current inert gas selectively removes the solvent, resulting in the enrichment of the analytes in the lumens. Temperature is one of the important variables in membrane pervaporation and the effect has been studied in previous research [29], thus it was not investigated in this study.

The objective of this study is to develop an automated and simple TAS by interfacing LLME, membrane pervaporation and HPLC–UV detection. These steps will perform extraction, concentration and detection respectively. The automated TAS will have the capability of continuous on-line monitoring of trace analytes in water.

2. Experimental

2.1. Experimental system

The experimental system is shown in Fig. 1. It included two hollow fiber membrane modules, two pumps (Hewlett-Packard 1050 HPLC pump) and a HPLC system (Hewlett-Packard 1050). The first pump was used for the delivery of the organic extractant, and the other for the water sample. An automatic six-port injection valve (Valco Instruments, Houston, TX, USA) was used to make repeat injections into the HPLC. The two membrane modules were structurally similar. Hollow fiber membranes were selected because they provided higher surface area per unit volume. The modules were made in the shell and tube format [16,30–37]. The first one served as the extraction module, and the latter as the pervaporation module. Water sample flowed through the shell side of the extraction module while the organic extractant flowed inside the hollow fiber lumens. The target analytes from the aqueous sample were extracted into the organic solvent in the membrane pores and then into the acceptor phase in the lumens. The extract continued to flow through the membrane lumen of the pervaporation module, where the nitrogen stripping gas flowed counter-current on the shell side. The evaporation of the solvent into the nitrogen flow concentrated the extract. The enriched extract was injected directly into the HPLC for analysis. The injection volume was 20 μL, and the injections were made automatically by a timer controlled six-port injection valve every 5 min.

2.2. Membrane module construction

The hollow fiber membrane modules were made with six pieces of composite membrane fibers packed in a Teflon tube. The length of the membrane used in the extraction and pervaporation modules were 128 and 144 cm, respectively. These

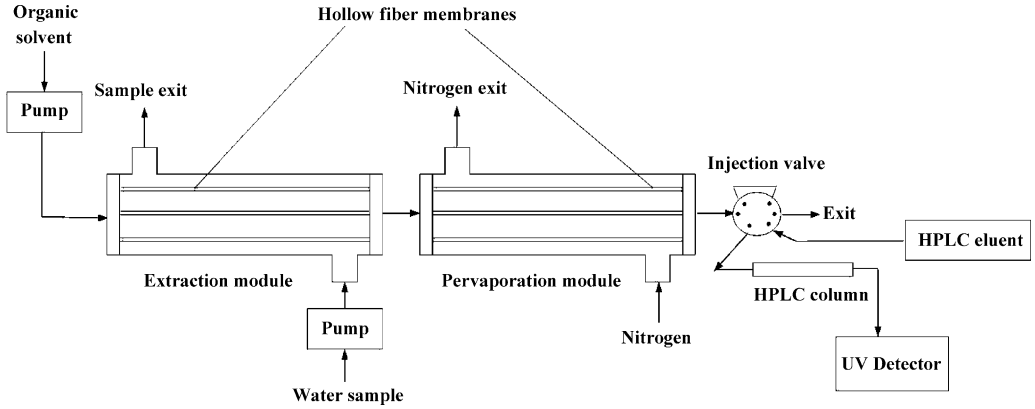

Fig. 1. On-line interfacing membrane extraction, pervaporation and HPLC–UV detection into a TAS.

could be rolled into 3–4 in. diameters spool. Each end of the PTFE tubing was connected to a tee union (Supelco, PA, USA). Epoxy (Resin Technology Group, LLC, S. Easton, MA, USA) was used to seal the space between the membranes and the tee, thus preventing the mixing of the water sample and the organic extractant. The hollow fiber membrane used in this study was a composite membrane with 0.260 mm O.D. and 0.206 mm I.D. (Applied Membrane Technology, Minnetonka, MN, USA). It consisted of a 1 μm thick homogeneous siloxane as the active layer deposited on micro-porous polypropylene tubing as the support.

2.3. Reagents

The model analytes used in this study were naphthalene and biphenyl, which were purchased from Aldrich (Milwaukee, WI, USA). HPLC-grade *n*-hexane (Fisher Scientific, NJ, USA) was selected as the organic extractant. Prior experiments in the laboratory have showed good pervaporation of *n*-hexane through non-polar membranes [29]. All other chemicals in this study were ACS grade reagents. Deionized water was obtained from a Milli-Q water purification system (Millipore, Bedford, MA, USA).

2.4. Chromatographic separation

Chromatographic separation was carried out by a Hewlett-Packard 1050 HPLC system with a Waters 486 Tunable Absorbance UV detector. The HPLC column was a 15 cm long Nova-Pack (Waters, Medford, MA, USA) C_{18} column with an I.D. of 3.9 mm. A mixture of acetonitrile–10 mM K_3PO_4 solution (45:55, v/v) was used as the mobile phase at a flow rate of 2 mL/min. The absorbance wavelength was set at 254 nm [37]. Minichrom V 1.62 software (VG Data System) was used for data acquisition.

2.5. Three experimental modes

The experiments were carried out in three different experimental modes, namely, 1–3. In Mode 1, only the membrane extraction module was used. In Mode 2, both extraction and pervaporation modules were used, but without the flow of stripping gas in the pervaporation step. This limited the solvent loss in the pervaporation step. In Mode 3, the membrane extraction was followed by membrane pervaporation with a flow of stripping gas, which resulted in a large solvent loss and led to a high degree of enrichment. Modes 2 and 3 are referred as the TAS modes. Any prior memory effect in the membranes was eliminated by flowing/washing the hollow fiber membranes with *n*-hexane for 2 min.

3. Results and discussion

Fig. 2 shows the concept of on-line interfacing of membrane extraction and pervaporation. The organic solvent, which is also referred to the acceptor, flowed inside of the lumens of hollow fiber membrane extractor, while the water sample (donor) flowed counter-current on the shell side. Analytes were extracted into the organic solvent. Some of the solvent could be lost by dissolving in water, thus concentrates the extract. It has been reported that this preconcentration approach was possible with polar, water-soluble solvents such as butyl acetate and isopropyl acetate, but not with non-polar solvents such as hexane [37]. In this study, pervaporation was used for analytes preconcentration. This approach is more universal and should work with all solvent with reasonable volatility. As the extract flowed in, the stripping gas selectively removed some of the solvent, resulting in a more concentrated extract for HPLC analysis. In general, the combination of the two membrane modules allowed both extraction and concentration to be carried out on-line, and continuously.

3.1. Extraction efficiency and enrichment factor

Enrichment factor (EF) is defined as the ratio of analyte concentration in the final extract to that in the original water

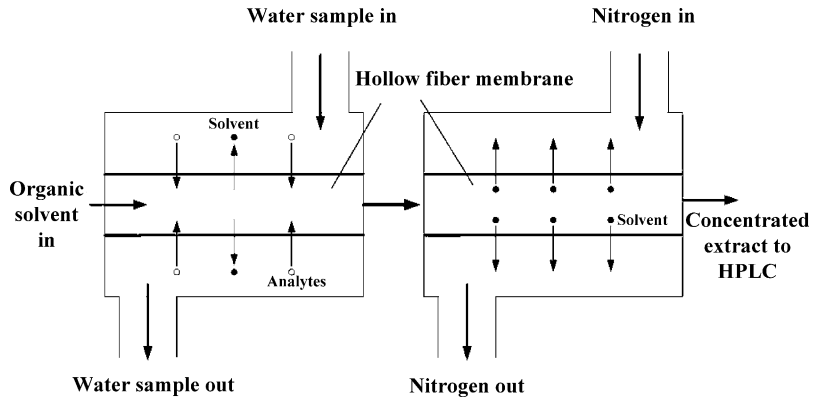

Fig. 2. The concept of membrane extraction and pervaporation.

sample:

$$\mathrm{EF} = \frac{C_s}{C_w} \quad (1)$$

C_s is the analyte concentration in the final extract, and C_w is the analyte concentration in the original water sample. A higher EF leads to a higher sensitivity and a lower detection limit. Extraction is usually quantified as extraction efficiency (EE), which is the fraction of analytes removed by the acceptor from the original water sample [24]. EE is computed as:

$$\mathrm{EE} = \frac{n_s}{n_w} = \frac{C_s V_s}{C_w V_w} = \mathrm{EF}\frac{V_s}{V_w} \quad (2)$$

n_s and n_w are the analyte mass in the final extract and in the original water sample, V_s and V_w are the volume of the concentrated extract and the original water sample, respectively.

In the continuous flowing system, the volume of the water entering in time t is expressed as:

$$V_w = F_w t \quad (3)$$

where F_w is the water flow rate. The volume of organic solvent is different throughout the system, as portion of the solvent is lost during the experiment. It is expressed as:

$$V_s = V_{si} - V_{ls} = V_{si} - V_{si}L_s = V_{si}(1 - L_s) \quad (4)$$

V_{si} is the initial volume of the organic solvent entering the extraction module while V_{ls} the solvent lost during the extraction and pervaporation, and L_s is the fraction of the solvent lost. The volume of the organic solvent entering the extraction module over time t is given as:

$$V_{si} = F_s t \quad (5)$$

where F_s is the flow rate of the entering organic solvent. According to Eqs. (4) and (5) the fraction of the solvent lost can be expressed as:

$$L_s = 1 - \frac{V_s}{F_s t} \quad (6)$$

Based on Eqs. (2)–(5) EE is expressed as:

$$\mathrm{EE} = \mathrm{EF}(1 - L_s)\frac{F_s}{F_w} \quad (7)$$

Thus, EE can be computed from the enrichment factor, flow rates of solvent and water sample, and the fraction of the solvent lost.

3.2. Comparison of the EF in the three experimental modes

Three different experimental modes (Modes 1–3, which were described in Section 2.5) were compared. The donor sample used was 0.5 μg/mL naphthalene and 0.1 μg/mL biphenyl in water. The water sample flow rate was 5 mL/min. The organic solvent (*n*-hexane) flowed at 0.1 mL/min. The flow rate of the stripping nitrogen in the pervaporation module in Mode 3 was 45 mL/min. The results are shown in Fig. 3. The EF somewhat increased (less than two times) when the pervaporation without nitrogen stripping was coupled to membrane extraction module. The enrichment factor increased significantly (4–9 times) in the presence of nitrogen stripping in the pervaporation module. Enrichment factors for naphthalene and biphenyl in Mode 3 were 93 and 188 respectively. The advantage of the TAS is clearly evident.

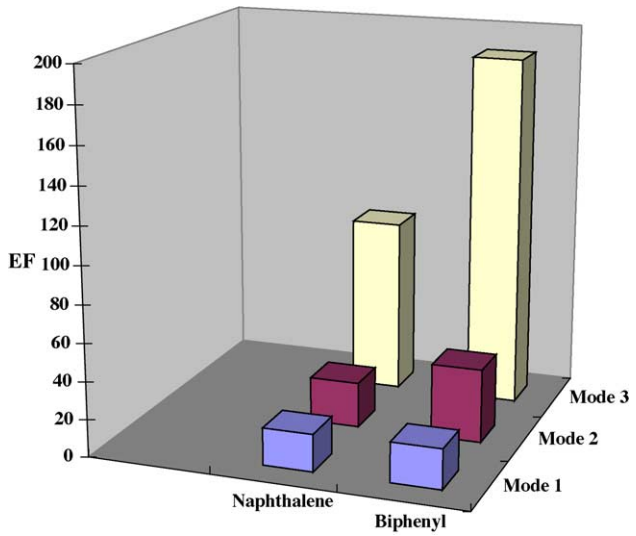

Fig. 3. Comparison of EF in the three experimental modes, Mode 1: membrane extraction only; Mode 2: membrane extraction and pervaporation without N_2 stripping; Mode 3: and membrane extraction and pervaporation with a N_2 flow rate of 45 mL/min. In each case the sample contained 0.5 μg/mL naphthalene (Nap) and 0.1 μg/mL biphenyl (Bph), the water flow rate was 5 mL/min, and the extractant (*n*-hexane) flow rate was 0.1 mL/min.

3.3. Influence of the water sample flow rate

The water sample flow rate played an important role in the determination of enrichment factor, extraction efficiency and solvent loss. The experiment was performed in Mode 2, the hexane flow rate was kept constant at 0.1 mL/min, while the water flow was changed from 1 to 5 mL/min. The concentration of naphthalene and biphenyl in the water sample was 0.1 μg/mL. The extract volume at the outlet of the pervaporation module was measured for time t, and the solvent loss was calculated using Eq. (6). The results are shown in Fig. 4.

The EF of naphthalene (Nap) and biphenyl (Bph) increased four times as the water flow increased from 1 to 5 mL/min. With an increased water flow rate, more analytes contacted the membrane, which led to a higher EF. The solvent loss increased slightly with the increase in water flow rate. The extraction efficiency decreased with the increase of water flow rate. Although more analytes were brought in, a larger fraction went unextracted. Higher flow rates were not employed as the high pressure across the membrane could decrease the lifetime of the extraction module. Donor flow rate of 4 mL/min was selected as a compromise between high enrichment and long lifetime.

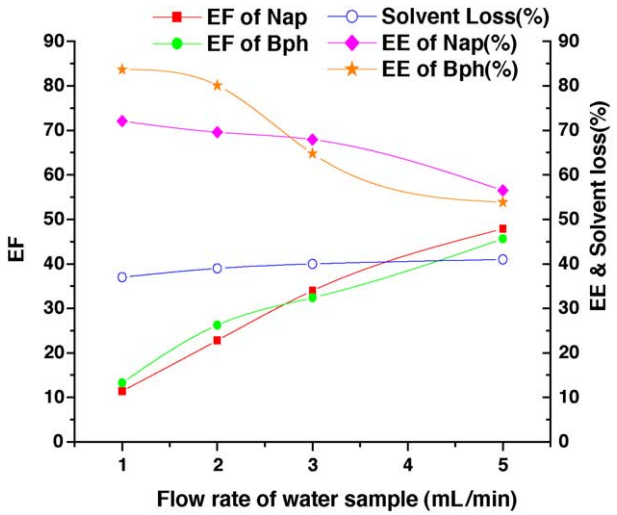

Fig. 4. EF, EE (%) and solvent loss (%) as a function of flow rate of the water sample (0.1 μg/mL, Nap and Bph). The extractant flowed at 0.1 mL/min. This was operated in Mode 2.

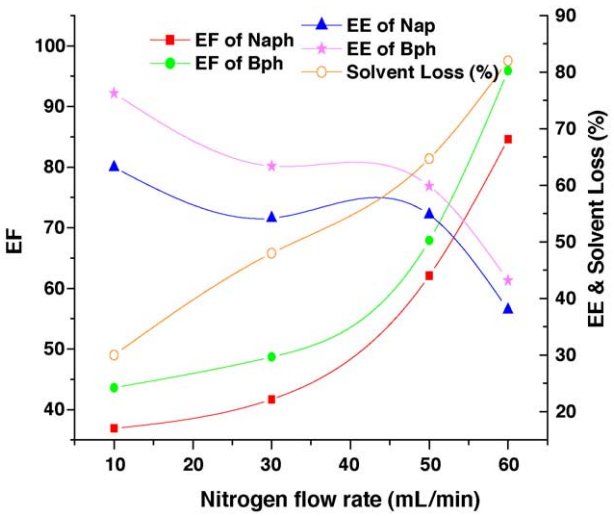

Fig. 5. EF, EE (%) and solvent loss (%) as a function of N_2 flow rate. Water sample containing 0.1 μg/mL Nap and Bph flowed at 4 mL/min. The extractant flow rate was 0.1 mL/min. This was operated in Mode 3.

The organic solvent in the extract could be lost either by dissolution into the aqueous phase or evaporation in the pervaporation module. The poor water solubility of *n*-hexane and no stripping gas in the pervaporation module resulted in relatively low solvent loss in Mode 2.

3.4. Influence of nitrogen flow rate in the pervaporation module

The effect of the nitrogen flow in the pervaporation module was tested in Mode 3, by varying the nitrogen flow rate from 10 to 60 mL/min. The inlet concentration of naphthalene and biphenyl was 0.1 μg/mL. The water and the extraction solvent flow rates were 4 and 0.1 mL/min, respectively. EF, EE and solvent loss as a function of N_2 flow rate are shown in Fig. 5. The EF increased with the N_2 flow rate, which was attributed to higher solvent removal. The EE decreased with the increase in N_2 flow rate as some analyte molecules were lost with the solvent in the pervaporation step. The goal of this study was to achieve higher enrichment, thus nitrogen flow rate of 60 mL/min was selected in the following study.

3.5. Influence of the acceptor flow rate

The effect of the organic solvent flow rate on EF was tested by operating Mode 3. The nitrogen flow rate was 60 mL/min. The sample contained 0.1 μg/mL each of naphthalene and biphenyl in water. The water flow rate was constant at 4 ml/min, while the extractant (*n*-hexane) flow rate was varied from 0.075 to 0.25 mL/min. The whole process was carried out on-line and the extract was injected into the HPLC every 5 min. The results are shown in Fig. 6.

EF decreased with the increase of the extractant flow rate. At lower flow rate, the contact time of the analytes with the solvent increased, thus more analytes could be extracted. In the pervaporation module also, the lower flow rate led to higher solvent removal, leading to higher EF. At flow rates lower than 0.075 mL/min, all the solvent was lost in the pervaporation module and no extract could be obtained for analysis. Therefore 0.075 mL/min was chosen as the acceptor flow rate. Enrichment factors as high as 192 were obtained under these conditions.

3.6. Analytical performance

Different analyte concentrations were assayed in Mode 3. The flow rates of the water sample, the organic solvent and the stripping nitrogen were 4, 0.075 and 60 mL/min, respectively. The linear dynamic ranges were found to be 25–100 and 5–100 ng/mL for naphthalene and biphenyl respectively, with correlation coefficient (R^2) above 0.998.

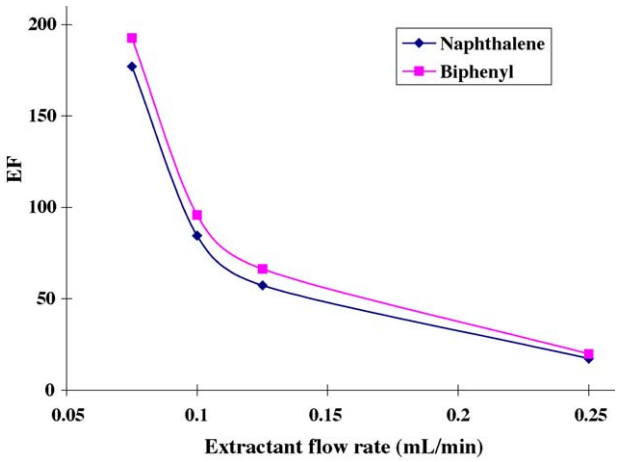

Fig. 6. EF as a function of the extractant flow rate. The sample containing 0.1 μg/mL Nap and Bph flowed at 4 ml/min. The N_2 flow rate was 60 mL/min. This was operated in Mode 3.

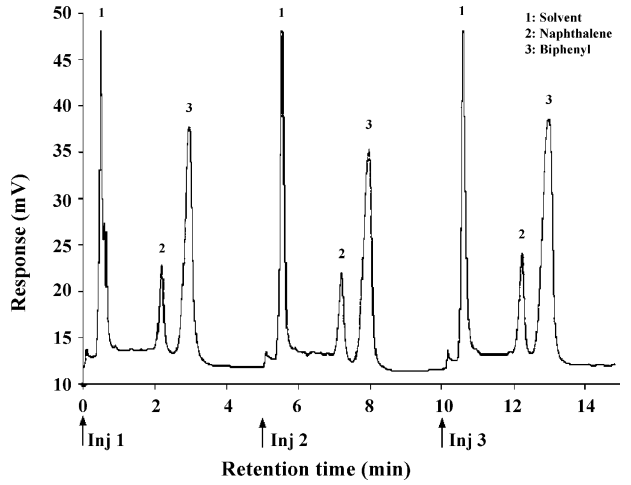

Fig. 7. Continuous monitoring of a water sample containing 50 ng/mL Nap and Bph. The flow rates of the water sample, the extractant and the N_2 were 4, 0.075 and 60 mL/min, respectively. Automatic injections were made every 5 min at Inj 1, Inj 2 and Inj 3.

Continuous automated monitoring was carried out at a relatively high frequency. The analytes were extracted into the organic solvent (*n*-hexane) and the solvent was continuously removed from the extract by pervaporation. The enriched extract was injected into the HPLC every 5 min by the automatic six-port injection valve. Sequence of chromatograms obtained during continuous on-line operation is shown in Fig. 7. The concentration of naphthalene and biphenyl in the water sample was 50 ng/mL. Good reproducibility in peak shape and retention time was observed. The relative standard deviations (RSDs) of naphthalene and biphenyl were 3.7 and 5.0% based on seven replicates, measured over three different days. The method detection limits (MDLs), which were obtained following a standard EPA procedure [38], were 5.8 and 7.9 ng/mL for naphthalene and biphenyl, respectively.

4. Conclusions

Interfacing on-line membrane extraction, pervaporation and HPLC–UV detection led to the development of a relatively simple and an effective TAS for the on-line monitoring of trace semi-volatile/non-volatile organic compounds. The membrane extraction served as the separation and the initial enrichment step, while the pervaporation as the final enrichment step. This system demonstrated enrichment factor as high as 192 and good analytical performance during the continuous monitoring. The important operational variables were the different flow rates, namely, the water sample, the solvent and the stripping gas.

Acknowledgements

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