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Journal of Chromatography A, 754 (1996) 197–205

JOURNAL OF CHROMATOGRAPHY A

Supported liquid membranes for sampling and sample preparation of pesticides in water

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Abstract

The use of supported liquid membranes for sampling and sample preparation of pesticides in water is described. A porous PTFE membrane is impregnated with a water-immiscible organic solvent, giving the supported liquid membrane. The optimisation of the extraction processes; involving the pH of the donor and the acceptor, choice of membrane liquid and donor flow-rate will be discussed. The technique has also been used for time integrating field sampling of acidic herbicides in natural water samples. Examples of sample preparation of phenoxy acids, sulfonylurea herbicides and triazines from natural water samples will be shown.

Keywords: Environmental analysis; Water analysis; Sample preparation; Membranes, supported liquid; Pesticides

1. Introduction

The sample handling step for the determination of different pollutants in natural water has lately become vital. The analytical instruments of today are well developed exhibiting good performances, therefore sample handling is now the critical step in the analytical procedure [1]. One class of pollutants that can be found at very low concentrations in water and still affect the environment are pesticides. Within the European Community the maximum permissible limits for pesticides are 0.1 and 1.0 μg/l for single pesticides in drinking and surface water respectively [2]. For determination of such low concentrations the sample handling step is essential.

The most commonly used methods for the sample preparation of pesticides in water samples are liquid–liquid extraction (LLE) and solid-phase extraction (SPE). The fact that LLE uses large volumes of organic solvents (often chlorinated) will make the technique less attractive in the future, since many countries today are planning for stricter regulations concerning the use of organic solvents.

During the last two decades the use of SPE for sample preparation has developed rapidly [3,4]. Both cartridges and precolumns packed with sorbents have been used and lately also solid-phase extraction disks have been developed, where the packing material is enmeshed in an inert PTFE matrix [5]. Today there are a large variety of SPE phases, cartridges and disks as well as automated instruments available on the commercial market. Many different sorbents have been used over the years and the most frequently used packing materials for the determination of organic pollutants have been summarised by Hennion [3]. Lately, very selective immunoaffinity sor-

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0021-9673/96/$15.00
PII S0021-9673(96)00218-X

bents have been developed for the extraction of triazine and phenylurea herbicides [6,7].

An alternative sample-handling technique was developed by Audunsson in the mid-1980s [8]: the supported liquid membrane (SLM) extraction. It utilises a porous PTFE membrane, which is impregnated with an organic solvent immiscible in water. The organic solvent in the membrane forms a barrier between the sample and the analytical instrument, which the analytes have to pass to be measured. The SLM technique can be described as a two-step LLE procedure in a flow system. It is typically applied to acidic or basic compounds, but recently also extraction of permanently charged compounds has been demonstrated [9].

In order to be extracted into the immobilised hydrophobic organic membrane liquid, the analytes must be uncharged. For acidic analytes, the pH in the aqueous donor therefore needs to be kept below the pK_a values of the analytes, rendering them uncharged. In this form, the analytes are then extracted into the membrane liquid. They diffuse across the membrane and are subsequently extracted into the aqueous acceptor, where the pH is kept above the pK_a. The acidic analytes will thus be charged and thereby non-extractable, and hence trapped in the acceptor. The transport through the membrane, as well as through the membrane–acceptor interface, will be driven by the concentration gradient of the uncharged species that arises across the membrane. By pumping the donor while keeping the acceptor stagnant, a selective enrichment of the analytes in the acceptor will be achieved. Basic compounds can be enriched with the opposite pH conditions.

The SLM technique has, during the last ten years, been actively developed and applied to biological as well as environmental sample types [10]. This paper will describe the use of SLM for the sampling and sample preparation for determination of some different pesticides in natural water samples.

2. Equipment and procedure

The core of the SLM technique is an organic liquid which is supported in the pores of a porous PTFE membrane. The most commonly used membrane type is Fluoropore FG (average pore size 0.2 μm, total thickness 175 μm of which 115 μm is polyethylene backing, porosity 0.70; Millipore, Bedford, MA, USA). Alternative membranes [PTFE and PVDF (polyvinylidene difluoride), with different characteristics and from different manufacturers] have been investigated [8,11]. The liquid membrane is formed by simply immersing the PTFE membrane in the organic solvent for at least 15 min. The membrane holder (see Fig. 1) consists of two blocks made of either PTFE or PVDF. The membrane is placed between the two blocks, forming two separate channels: the donor and the acceptor channel. The channels are arranged like Archimedes' spiral and were, in most investigations described here, 1.5 mm wide, 0.25 mm deep and 250 cm long. This gives a volume in each of the channels of ca. 950 μl. For other applications of the SLM technique (i.e. for biomedical analysis) other membrane configurations are used with channel volumes as small as ca. 1 μl [12].

The membrane device is placed in a flow system in which the flow-rates through the different channels are controlled by peristaltic pumps. For connection to a liquid chromatographic system pneumatic valves are used (see Fig. 2).

In all applications described here the final analysis has been performed with liquid chromatography, in most cases with a precolumn packed with either

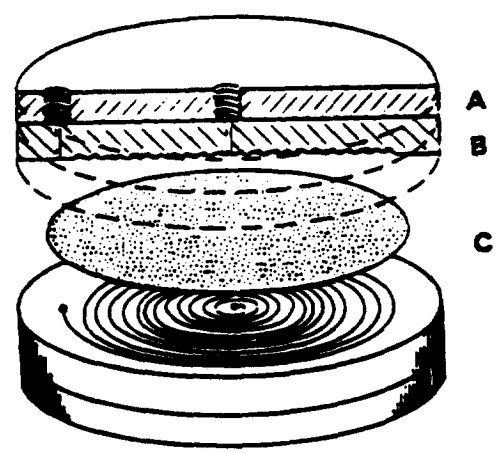

Fig. 1. Membrane separator: (A) aluminium backup; (B) PTFE block with grooves like Archimedes' spiral; (C) porous PTFE membrane with polyethylene backing. From Ref. [17] with permission; ©1992 American Chemical Society.

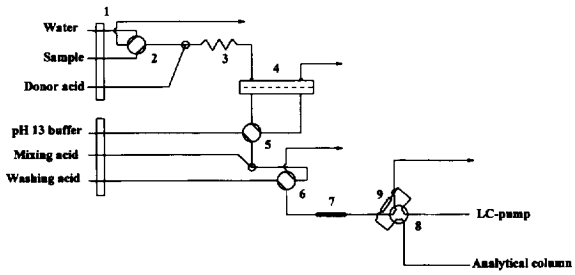

Fig. 2. Schematic flow system for SLM extraction. (1) peristaltic pump, (2, 5, 6) switching valves, (3, 7) mixing coils, (4) membrane separator, (8) precolumn and (9) Valco injector. From Ref. [24] with permission from Vieweg, Wiesbaden.

silica C_{18} or a polymeric material (e.g. PRP-1) to permit the injection of typically 1 ml from the acceptor phase. The precolumn has been used both in on-line connection to liquid chromatography (LC) as well as for focusing of the time-integrating collected samples.

3. Optimising the extraction process

3.1. Theory

The theory for the SLM extraction process has been thoroughly described by Jönsson et al. [13]. The extraction of analytes from the first aqueous phase (the donor) through the organic membrane to the second aqueous phase (the acceptor) is governed by several factors. Among them are the pH of the two phases, the polarity of the organic solvent and the flow-rate of the donor. The most important quantitative parameter for describing the extraction process is the extraction efficiency (E), defined as the fraction of analyte amount that is transported from the donor phase through the membrane to the acceptor phase.

The result of an extraction can also be viewed as an enrichment factor (E_e), i.e. the ratio of the concentration in the acceptor to that in the donor (i.e. in the original sample) or as an accumulation factor (E_a), the number of moles of analyte accumulated in the acceptor per time-unit. In Eqs. (1–3), the interrelationships of these factors are summarised. V_A, V_S, C_A and C_S are volumes and total concentrations in the acceptor and in the extracted sample, respectively, t is time and F_D is the donor flow-rate.

$$E = V_A C_A / V_S C_S \quad (1)$$

$$E_e = C_A / C_S = E V_S / V_A \quad (2)$$

$$E_a = V_A C_A / C_S t = E F_D \quad (3)$$

3.2. Influence of donor flow-rate

One of the limitations of the SLM technique is that the extraction efficiency will decrease with increasing donor flow-rate. However, from Eq. (3), it is seen that if the conditions are such that the decrease in E with flow-rate is small, then the accumulation factor E_a may increase with the donor flow-rate, however, on the expense of the requirement of a larger sample volume. From theory [13], it turns out that this is the case if the extraction of the analyte is limited by the mass transfer in the donor channel, i.e. if the partition coefficient K_p between the organic membrane liquid and the aqueous donor phase is relatively large ($K_p > 1$). On the other hand, if the rate of mass transfer through the membrane is the limiting factor (which is equivalent to the condition $K_p < 1$), E_a is independent of F_D.

Since all pesticides hitherto extracted with the SLM technique fulfill the condition $K_p > 1$, it might be favourable to use a higher donor flow-rate in these applications. With a higher donor flow-rate it is possible to either speed up the extraction time or, if the same extraction time is used, to lower the limit of detection. It is also practically possible to use higher donor flow-rate for environmental water samples, since the sample volumes are unlimited.

Typically, flow-rates around 1 ml/min are used with the SLM units described above. In a study of extraction of phenolic acids from circulating nutrient solutions for tomato cultivation [14], higher donor flow-rates were applied. As can be seen in Fig. 3, there is indeed an increase in E_a up to flow-rates as high as 6.5 ml/min. However, this advantage is offset by decreased membrane stability, while donor flow-rates around 2.5 ml/min are recommended to be used for extraction of this type of samples in the future.

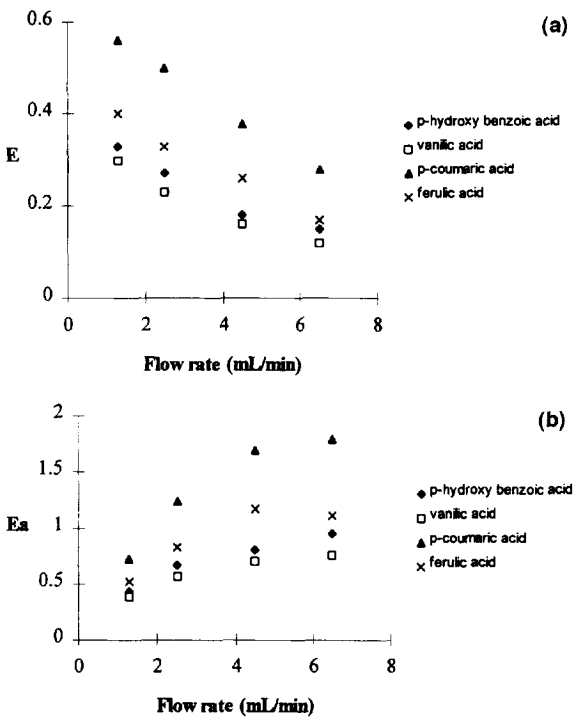

Fig. 3. (a) Extraction efficiencies, E, as function of donor flow-rate (ml/min) for four phenolic acids. Extraction of nutrient solutions spiked with 50 nM of each of the acids. (b) Accumulation factors, E_a, as function of donor flow-rate (ml/min) for four phenolic acids. Extraction of nutrient solutions spiked with 50 nM of each of the acids.

3.3. Influence of the donor and acceptor pH

When the SLM technique is applied to acidic or basic compounds, the extraction efficiency will partly be governed by the pH of the donor and acceptor phases. In the donor, the pH must be such that a sufficiently large fraction (α_D) of the analyte is uncharged and thus amenable to extraction into the membrane. It has turned out that this requirement in practice is not very severe: it is usually enough if pH is $<pK_a$ for acidic analytes (where $\alpha_D = 0.5$). In some cases, even somewhat higher pH values can be accepted, as the kinetics of the protonation equilibrium is fast.

The pH of the acceptor is more critical. The trapping capacity of the analytes in the acceptor is given by $C_A\alpha_A$, where α_A is the fraction of analyte that is in extractable form in the acceptor. The analytes are extracted as long as the gradient of uncharged analyte exists, i.e. when $C_A\alpha_A < C_D\alpha_D$. Thus, this relation gives the maximum enrichment factor possible to attain ($E_e < \alpha_D/\alpha_A$). However, the extraction efficiency is only constant as long as $C_A\alpha_A \ll C_D\alpha_D$

For relatively short extraction times, e.g. some hours, an α_A value below $5 \cdot 10^{-4}$ is sufficient [13]. This means that for extraction of acidic analytes, the pH in the acceptor solution should be 3.3 pH units above the highest pK_a value and correspondingly for basic analytes. Under typical circumstances, the extraction efficiency will then decrease by 1% per extracting hour [13]. For sampling periods of 24 h or longer, as practised in time integrating field sampling, a lower value of α_A is necessary, unless the extraction efficiencies are affected. For most compounds of interest these conditions are readily obtained. An exception is triazine herbicides, as will be discussed below. These compounds are weak bases with pK_a of about 1.5, so a complete trapping would require a pH of < -1.8, which is not easily arranged. This means that the trapping of triazines in the acceptor can never be complete.

3.4. Selectivity tuning by the membrane liquid

The selectivity of the extraction can be tuned by changing the composition of the organic solvent in the membrane. Generally, a more polar solvent will extract more interfering compounds, as seen in Fig. 4 [15]. These interferents, presumably humic substances, result in a hump in the beginning of the chromatogram. Therefore, the use of a solvent with as low polarity as possible is advantageous. However, with fairly polar analytes, their extraction efficiencies will also increase with increasing polarity of the membrane liquid. A compromise between good selectivity and high extraction efficiency is necessary and in the study referenced (involving sulfonyl urea herbicides), a membrane of 100% di-*n*-hexyl ether was chosen.

4. Time integrating sampling

One area where the SLM technique has been used successfully, apart from laboratory extraction before

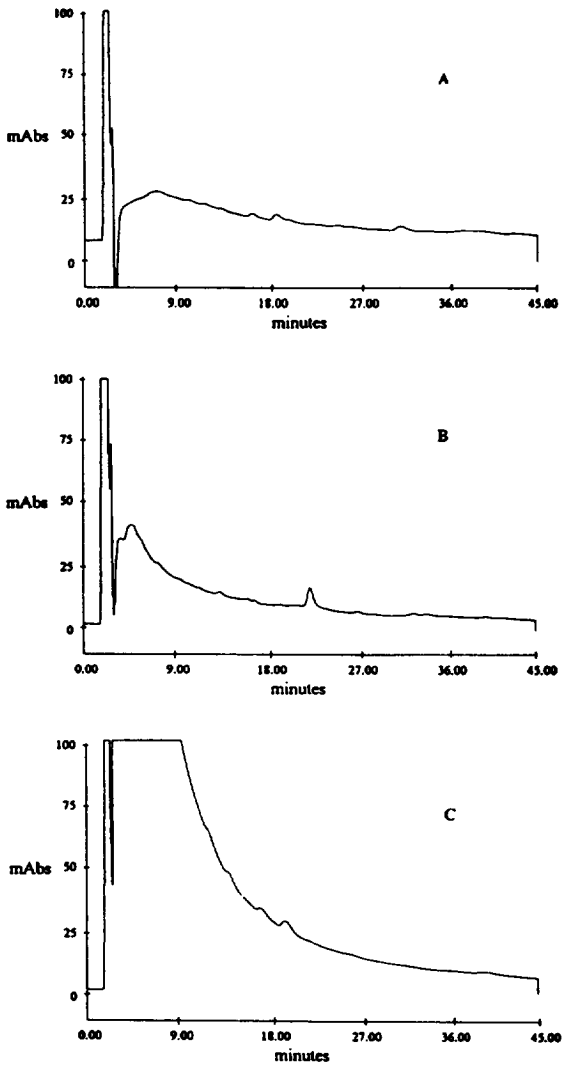

Fig. 4. Supported liquid membrane extraction of blank natural water samples. (A) *n*-Undecane–di-*n*-hexyl ether (1:1), (B) pure di-*n*-hexyl ether, (C) 5% TOPO in di-*n*-hexyl ether. From Ref. [15] with permission from Elsevier, Amsterdam.

liquid chromatography (LC) analysis, is for time-integrating sampling [16,17]. The whole set-up (see Fig. 5) of peristaltic pump, membrane holder and tubing etc., were used in field for continuous sampling of acidic herbicides. Typical sampling periods were 24 h. With a flow-rate of 1.0 ml/min, ca 1.4 l water is processed. After sampling, the acceptor plug (2 ml) is brought to the laboratory for final analysis. 1 ml of the acceptor plug is injected onto the

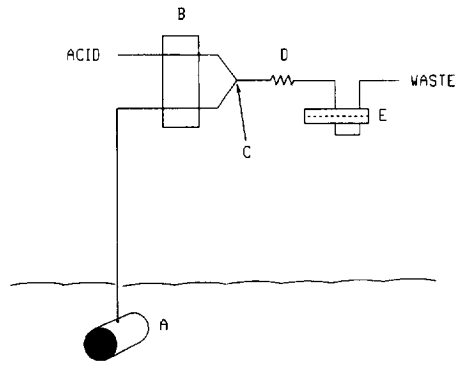

Fig. 5. Field sampling set-up for membrane enrichment of acidic herbicides in natural waters: (A) sampling point; (B) peristaltic pump; (C) confluence point of sample stream (0.8 ml/min) and a stream of 0.4 *M* H_2SO_4 (0.15 ml/min); (D) mixing coil; (E) membrane separator, with stopped flow in the acceptor. From Ref. [17] with permission; ©1992 American Chemical Society.

precolumn, which makes it possible to duplicate the analysis.

With the continuous sampling procedure using the SLM technique, a more reliable view of the leakage of herbicides out into a recipient is obtained than with a conventional grab-sampling procedure [16]. The concentration of 4-chloro-2-methylphenoxy acetic acid (MCPA) in a brook was determined using both continuous SLM sampling for 24 h sampling periods and grab sampling twice or three times per day. The samples were analysed by LC–UV; the grab samples were additionally extracted by LLE, derivatized and analysed by GC–MS.

Table 1 shows the concentration of MCPA obtained with both integrating sampling and grab sampling during a ten day period. The total leakage of MCPA to the recipient within the ten day period was 0.36 mg/ha with the integrating sampling procedure, while with grab sampling, it gave 0.17 and 0.18 mg/ha with either membrane work-up followed by LC–UV or LLE followed by GC–MS, respectively. The leakage is very variable in time and, therefore, the integrating sampling method is expected to give more reliable estimations of the total leakage if a large number of grab samples are not collected daily.

The found peaks of herbicide concentration corresponds to spraying the fields some days earlier in the run-off area. The concentrations in the brook are rather high within a short period of time. Thus, the

Table 1
Determination of MCPA in a Swedish brook for a period of 10 days[a]

Technique	Concentration of MCPA (ppb)									
	Day: 1	2	3	4	5	6	7	8	9	10
Integrating sampling	2.1	0.1	0.3	0.1	0.2	0.1	0.6	4.5	0.7	0.2
Grab sampling	0.2	0.3	0.1	0.1	0.1	0.1	1.5	1.2	1.2	0.1

[a] Values obtained either from integrating sampling using membrane technique for a period of one day, or from grab samples taken each morning. From Ref. [17] with permission.

grab sampling technique might give incorrect estimates of the mean concentration due to these rapid variations in concentration, if a high sampling frequency is not used. With the SLM time-integrating sampling procedure, the sampling is continuous and gives the average herbicide concentrations. It was also found in this investigation that changes in climate factors, like water and air temperature, did not affect the extraction efficiencies for the herbicides.

5. Applications

5.1. Phenoxy acids and similar acidic herbicides

Three phenoxy acids, 2,4-D (2,4-dichlorophenoxy acetic acid), MCPA and 2,4,5-T (2,4,5-trichlorophenoxy acetic acid), were extracted from spiked water containing 350 ppm artificial humic substances using a small membrane holder with an acceptor volume of 56 μl [18]. The membrane holder was connected on-line to HPLC (high-performance liquid chromatography) and the acceptor was transferred to a 100 μl injection loop after extraction. Several different membrane liquids were tested alone or in different mixtures, namely *n*-undecane, 1-decanol, 1-dodecanol, 1-tetradecanol and di-*n*-hexyl ether. When both the stability of the membrane and the extraction efficiencies of the analytes were taken into consideration, a 1:1 mixture of *n*-undecane and di-*n*-hexyl ether turned out to be the best membrane liquid. After 30 min enrichment (sample flow-rate of 0.25 ml/min), concentrations below 10 μg/l could be seen in the humic rich water (see Fig. 6). However, this limit of detection was too high and in all future works, a larger membrane holder was used in combination with a precolumn for focusing the acceptor. This, together with the use of sample flow-rates around 1 ml/min, permitted lower limits of detection.

The SLM technique was used for time-integrating field sampling, as described above, of six acidic herbicides (bentazon, 2,4-D, dicamba, dichlorprop, MCPA and mecoprop) [17]. A membrane composi-

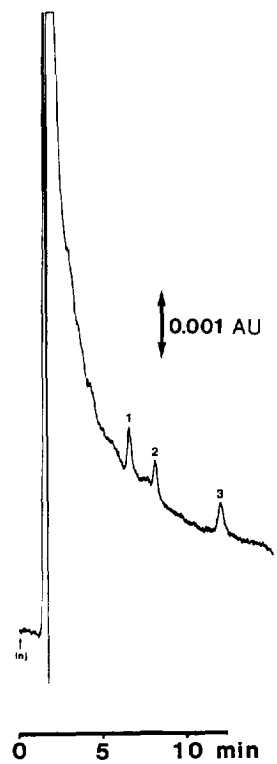

Fig. 6. Chromatogram showing the separation of (1) 2,4-D, (2) MCPA and (3) 2,4,5-T. Extraction of 7.5 ml of a humic acid solution spiked with 10 μg/l of each of the herbicides. From Ref. [18] with permission from Elsevier, Amsterdam.

tion of 1:1 *n*-undecane and di-*n*-hexyl ether was used also in this application. After a 24 h enrichment period, concentrations at sub ppb level could easily be seen in the collected samples.

For two other acidic herbicides, bromoxynil and ioxynil, successful preliminary studies have been made [19]. For this type of acidic herbicides, a membrane of pure *n*-undecane or a mixture of *n*-undecane–di-*n*-hexyl ether (1:1) gave the best extraction efficiencies. For a donor flow-rate of ca. 1 ml/min, extraction efficiencies of around 0.60 were obtained with these two membrane compositions, whereas a pure di-*n*-hexyl ether membrane gave extraction efficiencies of around 0.30.

5.2. Sulfonylurea herbicides

Sulfonylurea herbicides are a new type of very potent weed-killer which were introduced in the mid-1980s. Only some few grams of these herbicides are used per hectare, compared to several kilograms of conventional herbicides like phenoxy acids and triazines. This means that the concentrations of sulfonylurea herbicides that can be found in run-off water are extremely low. Four different sulfonylurea herbicides were investigated with the SLM approach [15,20], namely chlorsulfuron, metsulfuron methyl, tribenuron methyl and thifensulfuron methyl. The sulfonylurea herbicides were best extracted with 100% di-*n*-hexyl ether as membrane liquid. After 4 h of extraction, concentrations down to 0.1 μg/l of the herbicides could be found in spiked natural water samples, which is far below the limit of detection that is demanded within the European Community. However, with the low amounts used for spraying the fields, possibilities to determine even lower concentrations of the sulfonylurea herbicides would have been desirable. Therefore, post-column photolysis in combination with conductivity detection was tested as an alternative detection system to the normal UV absorption at 240 nm, since this has been reported by other authors to be successful for determining sulfonylurea herbicides [21]. Unfortunately, no improvement in lowering the limits of detection was found using the photoconductivity in combination with the SLM technique.

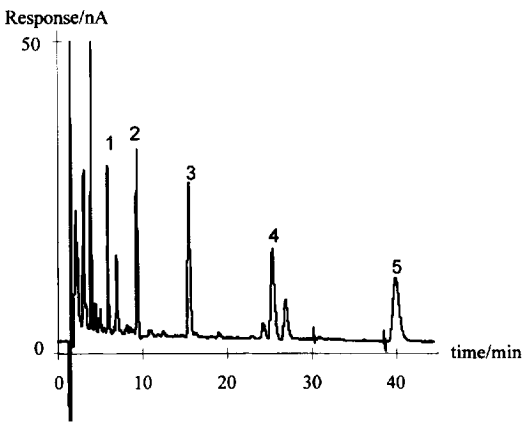

Fig. 7. Chromatogram after 30 min SLM enrichment of spiked Kävlinge river water. 1=4-chlorophenol (0.05 μg/l); 2=2,5-dichlorophenol (0.1 μg/l); 3=2,4,5-trichlorophenol (0.1 μg/l); 4=2,3,5,6-tetrachlorophenol (0.1 μg/l); 5=pentachlorophenol (0.1 μg/l). From Ref. [24] with permission from Vieweg, Wiesbaden.

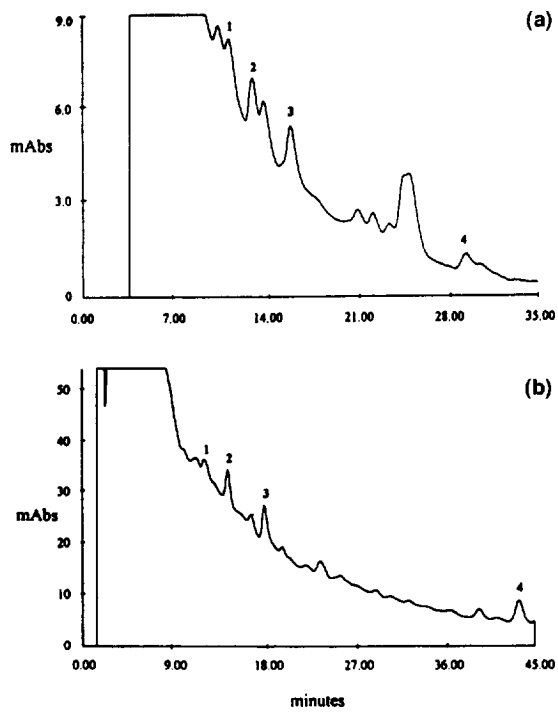

Fig. 8. (a) A 1 μg/l concentration of the sulfonylureas spiked in natural water enriched by SPE, 20 μl injection. (b) Chromatogram after supported liquid membrane enrichment of a natural water sample containing 0.2 μg/l of each of the four sulfonylureas. Peaks: 1=thifensulfuron methyl; 2=metsulfuron methyl; 3=chlorsulfuron; 4=tribenuron methyl. From Ref. [15] with permission from Elsevier, Amsterdam.

5.3. Triazines

Triazines constitute one of the most frequently used herbicide groups. Among the most common triazines are atrazine, propazine and simazine. They are basic compounds with a very low pK_a value (around 1.5). The possibilities of using SLM for the extraction of triazines from water samples have recently been investigated [22,23]. As mentioned above, the low pK_a values will give problems in trapping the triazines in the acceptor phase. An acidic solution of pH 0 or 0.8 was used as acceptor solution in these investigations. With this acceptor pH, only sample periods under 2 h were used in order to avoid back extraction of the analytes to the donor. It was possible to achieve extraction efficiencies as high as about 0.80 for propazine and simazine with the following extraction conditions: a pure di-*n*-hexyl ether membrane, donor flow-rate 0.2 ml/min, acceptor 0.5 *M* H_2SO_4 and donor 0.1 *M* NaOH [22]. However at this low acceptor pH, problems with the stability of the analytes have been encountered [23]. Further improvements on the extraction of triazines needs to be made, especially concerning the trapping of the analytes in the acceptor.

5.4. Related compounds

The SLM technique has also been used for sample preparation of chlorinated phenols from water samples [24]. Pentachlorophenol can be regarded as a pesticide, some of the other compounds are similar to herbicides and in some cases they can actually be degradation products of herbicides. A membrane system with a 100% *n*-undecane membrane was used for the extraction of 4-chlorophenol, 2,4-dichlorophenol, 2,4,5-trichlorophenol, 2,3,5,6-tetrachlorophenol and pentachlorophenol.

The combination of a very selective membrane solvent (100% *n*-undecane) and the connection of the membrane set-up on-line to a HPLC determination with electrochemical detection (1.0 V vs. Ag/AgCl), gave a very clean chromatogram after extraction of spiked natural water (see Fig. 7). With the sensitive electrochemical detection system, it was possible to determine concentrations below 25 ng/l of the phenols spiked to natural water samples after only 30 min SLM extraction time.

6. Discussion

The SLM technique is a good alternative to SPE for sample preparation of environmental water samples. A direct comparison between the SLM technique and SPE was made for the extraction of sulfonylurea herbicides [15]. Although the comparison was with an off-line SPE method, it was clearly seen that the SLM extraction gave a more selective extraction than with the SPE procedure (see Fig. 8 a and b). The SPE method used was basically performed according to Wells and Michael [25], using C_{18} packing material (0.5 g) and elution with methanol. The SLM technique is expected to give a more selective extraction than with conventional SPE packing materials like silica C_{18} and porous polymeric sorbents. With the new SPE packing materials based on immunoaffinity sorbents mentioned above [6,7], it is possible to achieve a selective extraction towards interfering substances. Furthermore, the SLM technique makes it also possible to extract very polar compounds, for which breakthrough might be a problem with SPE.

However, the SPE has some advantages over the SLM technique, especially the use of higher flow-rates (easily above 5 ml/min) and the applicability to a larger number of compound classes. With the SLM extraction technique, only compounds that can form ionised species can be extracted, while there are no such limitations for SPE. This means that some classes of pesticides, non-polar and non-ionisable compounds like organochlorine and organophosporus pesticides, cannot be extracted with the SLM technique.

Acknowledgments

The financial support from the Swedish Council for Forestry and Agricultural Research, the Swedish Natural Science Research Council and the Swedish Environmental Protection Agency are gratefully acknowledged.

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