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Determination of the semi-volatile compounds nitrobenzene, isophorone, 2,4-dinitrotoluene and 2,6-dinitrotoluene in water using solid-phase microextraction with a polydimethylsiloxane-coated fibre

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Abstract

Aqueous samples of the semi-volatile compounds nitrobenzene, isophorone, 2,4-dinitrotoluene and 2,6-dinitrotoluene, listed in the US Environmental Protection Agency (EPA) Method 609, were analysed by solid-phase microextraction (SPME) with a polydimethylsiloxane-coated fibre. SPME is a technique to extract organics from an aqueous matrix into a stationary phase immobilized on a fused-silica fibre. The analytes are thermally desorbed directly into the injector of a gas chromatograph. The sensitivity of this method decreased with increasing concentration of methanol and absorption temperature and increased with increasing concentration of Na_2SO_4. Samples of lake water were analysed using calibration graphs based on deionized water. The relative standard deviations of these analytes were 1–2% for analytes in deionized water and 2–4% for analytes in lake water. The detection limits were 9 ng/ml for nitrobenzene and 15 ng/ml for the other three analytes.

1. Introduction

Sample pretreatment is generally required for determination of trace organic pollutants in water. Liquid–liquid extraction and solid-phase extraction are the most commonly used techniques for semi-volatile and non-volatile compounds. The traditional method of liquid–liquid extraction requires large volumes of solvent for extraction and is protracted and tedious. Solid-phase extraction [1,2] requires less solvent, but for trace analysis a large volume of sample is generally required for preconcentration, and probably much time is required to load the sample solution. Contaminants may be eluted from the solid-phase packing [3].

Solid-phase microextraction (SPME) [4–16], recently developed by Pawliszyn and co-workers, eliminates most of the drawbacks in the preparation of an aqueous sample. SPME involves exposing a fused-silica fibre previously coated with a stationary phase [12] (or even a bare fused-silica fibre) to an aqueous solution containing organic compounds. The organic compounds partition between water and the stationary phase until equilibrium is reached. The fibre

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is then removed from the solution and the analytes are thermally desorbed into the injector of a gas chromatograph. The fibre is contained in a syringe to facilitate handling. The amounts of analytes absorbed on the fibre are linearly related to the concentrations of analytes in the aqueous sample. SPME, which takes only a few minutes for extraction and thermally desorbs analytes from the fibre coating, requires no solvents, is inexpensive, simple and readily adapted to automation [5].

The applications of SPME, studied by Pawliszyn and co-workers, include substituted benzenes in water [5,8,9] and headspace [11], caffeine in beverages [10], chlorinated compounds listed in the USA Environmental Protection Agency (EPA) Method 624 [7], polyaromatic hydrocarbons and selected polychlorinated biphenyls (PCBs) in water [6,16] and phenols listed in EPA Methods 604 and 625 [14,15]. Several fibre coatings, such as polydimethylsiloxane [8,11,16], polyacrylate [13,15] and polyimide [6], have been developed. The polydimethylsiloxane-coated fibre, the most successful, is the first kind of fibre available commercially (from Supelco) for an SPME fibre assembly [17].

It is of interest to establish whether SPME can also be used to determine other organic pollutants in water. In this work, we determined the semi-volatile priority pollutants listed in EPA Method 609 using the SPME fibre assembly. The desorption–time profile was explored. The effects of methanol, Na_2SO_4, duration of absorption and absorption temperature were investigated. The detection limits were 9 ng/ml for nitrobenzene and 15 ng/ml for isophorone, 2,4-dinitrotoluene and 2,6-dinitrotoluene. This method was applied to an environmental sample (lake water).

2. Experimental

The SPME fibre assembly was purchased from Supelco. The microextraction fibre was coated with polydimethylsiloxane (100 μm). The fibre was conditioned at 210°C before use.

SPME involves a few simple steps [4–7]. The fibre is withdrawn into the needle of the syringe (Hamilton Model 7005) and the needle is used to penetrate the septum of a sample vial (4 ml). The fibre is then inserted into the sample by depressing the plunger. The fibre coated with polydimethylsiloxane is completely immersed in the sample solution (3 ml). The sample solution in the vial is stirred with a stirring bar by the magnetic stirrer. The organic analytes become partitioned between the water and the stationary phase until equilibrium is reached (10 min). The plunger is withdrawn to retract the fibre into the needle and the syringe needle is then removed from the vial. For desorption, the needle is inserted into the GC injection port and then the fibre is exposed for 5 min. The injection position must be kept at a constant depth in all injections [5].

A gas chromatograph (Shimadzu GC 9-AM) with a split–splitless system, flame ionization detector and capillary column (Shimadzu CBP-10, 25 m × 0.2 mm I.D., film thickness 0.25 μm) were used. The fibre was desorbed in the GC injector at 210°C for 5 min. As the fibre began its desorption, the column temperature was kept isothermal at 70°C for 5 min, then increased at 7°C/min to 175°C and held isothermal at 175°C for 10 min. The detector temperature was 280°C. The carrier gas was nitrogen of purity 99.99%, further purified by passage through a gas purifier (Alltech) containing molecular sieve 5A and indicating Drierite and an oxygen-adsorbing gas purifier (OxiClear). The pressure of the carrier gas was 150 kPa and the flow-rate of make-up gas was 60 ml/min. The split flow-rate was 60 ml/min and the flow-rate for the septum purge was 2 ml/min. The GC temperature programme used for analysis the lake water was modified to avoid the interference peak that appeared near the 2,6-dinitrotoluene peak; the column was kept isothermal at 70°C for 5 min, then increased at 7°C/min to 120°C, at 1°C/min to 139°C and at 15°C/min to 175°C, and held isothermal at 175°C for 2 min. The same modified GC temperature programme was used to obtain the calibration graph for analytes in deionized water and lake water.

Stock standard solutions of nitrobenzene, iso-

phorone, 2,4-dinitrotoluene and 2,6-dinitrotoluene (TCI, Japan) were prepared in acetone (2000 μg/ml). Methanol (Optima grade; Fisher) and disodium sulfate (Osaca, Japan) were used to prepare the sample solution. Deionized water was prepared using a Millipore Milli-Q SP purification system. Lake water from National Tsing Hua University served as the environmental sample.

3. Results and discussion

A chromatogram of the mixed standard in water (1 μg/ml each) using SPME is shown in Fig. 1. Sharp peaks and good resolution were obtained under the analytical conditions used.

The absorption–time profile (Fig. 2) was obtained by monitoring the peak-area counts as a function of duration of exposure. The equilibrium period was 10 min for isophorone and 3 min for nitrobenzene, 2,4-dinitrotoluene and 2,6-dinitrotoluene. Factors that influence the equilibrium period were investigated by Pawliszyn and co-workers [4,5,9]. The equilibrium period is limited by mass transfer of the analytes through a thin, static aqueous layer at the fibre–solution interface. The equilibrium period increased with increasing distribution constant of the analyte and with increasing thickness of the fibre coating, but the equilibrium period decreased for the stirred solution [4,5,9]. As we analysed the sample under the constant conditions (the same thickness of fibre coating and stirring conditions), and the peak-area of isophorone was even smaller than that of nitrobenzene and 2,6-dinitrotoluene, we concluded that there must be other factors that affect the equilibrium period. The solubility [18] of isophorone in water (12 g/l) is much greater than that of nitrobenzene (1.9 g/l) and dinitrobenzene (about 0.3 g/l). These data imply that the extent of hydration of isophorone may be significantly greater than those of the others, such that diffusion of isophorone through the thin, static aqueous layer at the fibre–solution interface is slower than that of the others; hence the equilibrium period of isophorone is greater. In order to improve the sensitivity and precision, and extraction period of 10 min was chosen for subsequent experiments.

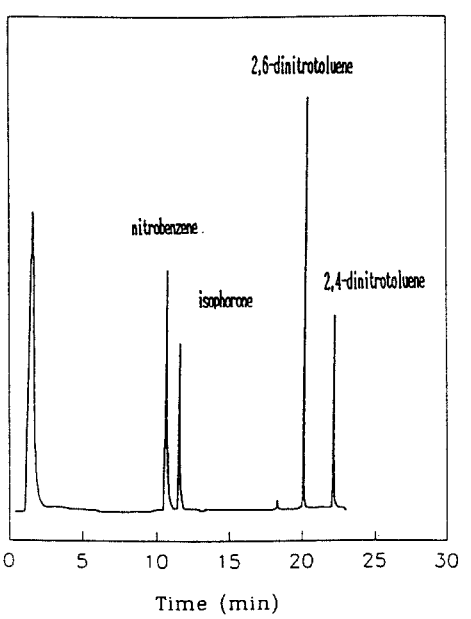

Fig. 1. Chromatogram of nitrobenzene, isophorone, 2,4-dinitrotoluene, 2,6-dinitrotoluene extracted from deionized water (1 μg/ml) by SPME for 10 min and desorbed in the GC injection port at 210°C for 5 min.

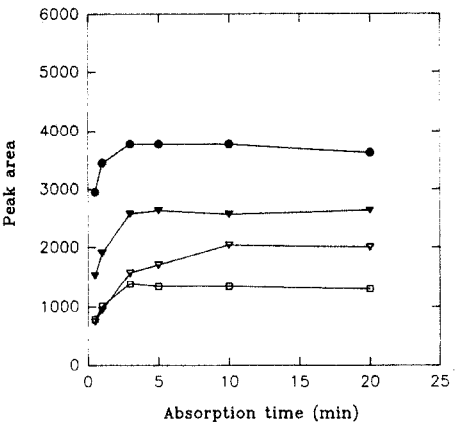

Fig. 2. Effect of absorption period on peak area of (●) nitrobenzene, (▽) isophorone, (□) 2,4-dinitrotoluene and (▼) 2,6-dinitrotoluene. Deionized water containing the analytes (0.4 μg/ml each) was extracted by SPME and desorbed in the GC injection port at 210°C for 3 min.

The desorption–time profile is shown in Fig. 3. Quantitative desorption of nitrobenzene and

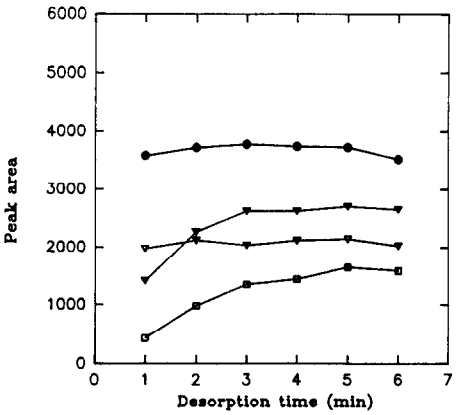

Fig. 3. Effect of desorption period on the peak area of the four analytes (symbols as in Fig. 2). Deionized water containing analytes (0.4 μg/ml each) was extracted by SPME for 10 min and desorbed in the GC injection port at 210°C.

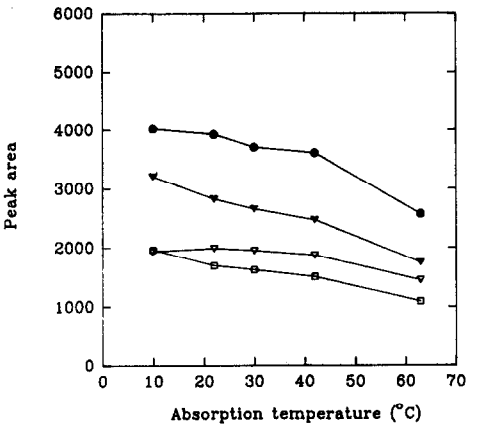

Fig. 4. Effect of temperature of absorption on the peak area of the four analytes (symbols as in Fig. 2). The sample solution (0.4 μg/ml each) was extracted by SPME for 10 min and desorbed in the GC injection port at 210°C for 5 min.

isophorone from the stationary phase of the fibre was achieved in 1 min or less, as the boiling points of the analytes [18] were near the desorption temperature. The period (5 min) required to desorb completely the less volatile analytes, 2,4- and 2,6-dinitrotoluene (b.p. 250 and 280°C), was much greater. Peak broadening of the analytes was avoided by cryofocusing of the analytes at the inlet of the GC column.

There was concern that the adsorbed semi-volatile analytes might not be efficiently removed from the fibre during thermal desorption for 5 min, and might thus contaminate the fibre for analysis of subsequent samples. To determine whether analyte retention was a significant source of error, the SPME fibre was inserted into the GC injection port a second time after analysis of the aqueous sample was completed. No carryover of analyte species was observed in the second GC analysis, demonstrating that 5 min were sufficient for thermal desorption.

The absorption–temperature profile is shown in Fig. 4. The amount of analytes absorbed decreased with increasing temperature of absorption. The distribution constant decreased with increasing temperature because absorption is generally an exothermic process; therefore, the amount of analytes adsorbed decreased with increasing temperature if equilibrium was reached. The rate of diffusion of the analyte species through the static aqueous layer at the fibre–solution interface decreases with decreasing temperature [5], such that less analyte is absorbed at a lower temperature if equilibrium was not reached. No effect of absorption temperature on the rate of diffusion was observed.

The effect of methanol on peak area is shown in Fig. 5. The amounts of 2,4- and 2,6-di-

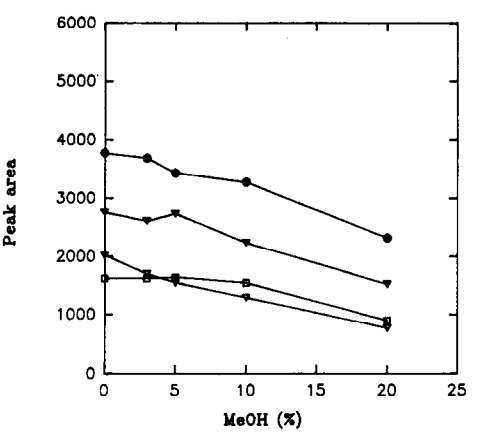

Fig. 5. Effect of methanol on the peak area of the four analytes (symbols as in Fig. 2). The sample solution (0.4 μg/ml each) was extracted by SPME for 10 min and desorbed in the GC injection port at 210°C for 5 min.

nitrotoluene adsorbed were little affected by methanol at concentrations less than 5% and 10%, respectively. The amount of nitrobenzene and isophorone absorbed on the fibre decreased with increasing concentration of methanol. An increased proportion of methanol in aqueous solution decreased the polarity of the aqueous sample so that the distribution constant decreased [5]. The more soluble analytes (isophorone and nitrobenzene) were more readily affected by methanol than the less soluble analytes (dinitrobenzene).

The effect on Na_2SO_4 on peak area is shown in Fig. 6. An increased Na_2SO_4 concentration in an aqueous sample increased the distribution constant of the analyte so that the amount of absorbed analyte increased [5]. Adding Na_2SO_4 to the sample solution may improve the sensitivity and the detection limit of SPME, but the amount of Na_2SO_4 added should be well controlled.

The precision of SPME for the determination of these analytes in deionized water and lake water was investigated. The relative standard deviations are given in Table 1. The values were about 1% for samples based on deionized water and 2% for samples of lake water, except for 2,4-dinitrotoluene, for which the values were 2% and 4%, respectively.

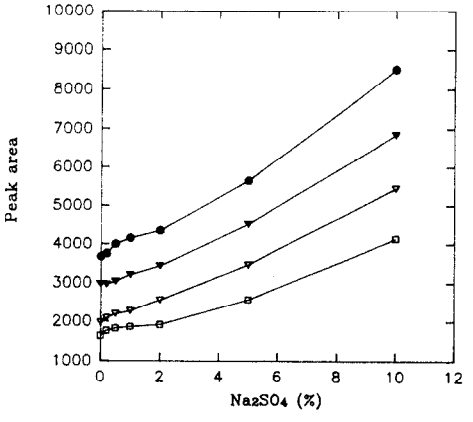

Fig. 6. Effect of Na_2SO_4 on the peak area of the four analytes (symbols as in Fig. 2). The sample solution (0.4 μg/ml each) was extracted by SPME for 10 min and desorbed in the GC injection port at 210°C for 5 min.

Table 1
Relative standard deviations (R.S.D.s) for the four analytes in lake water and deionized water

Compound	R.S.D. (%)[a]	
	Lake water	Deionized water
Nitrobenzene	0.8	1.7
Isophorone	0.5	0.5
2,4-Dinitrotoluene	0.9	2.1
2,6-Dinitrotoluene	2.0	4.4

[a] Data obtained for the concentrations of the four analytes from 0.4 to 10 μg/ml ($n = 15$).

The detection limits for the analytes in deionized water (0.05 μg/ml), calculated as three times the standard deviations of seven replicate runs, were 9 ng/ml for nitrobenzene and 15 ng/ml for the other three analytes.

Various amount of analytes (0.1, 0.4, 1, 2, 5 and 10 μg/ml) were added to deionized water and lake water to obtain the calibration graphs. The linearities were satisfactory. The slopes based on deionized water and lake water were almost identical, so that we were able to determine the analytes in lake water using calibration graphs based on deionized water.

4. Conclusions

Aqueous samples of the semi-volatile compounds nitrobenzene, isophorone, 2,4-dinitrotoluene and 2,6-dinitrotoluene were readily analysed using SPME with a polydimethylsiloxane-coated fibre. The sensitivity of this method decreased with increasing concentration of methanol and temperature of absorption and increased with increasing concentration of Na_2SO_4. Samples of lake water were analysed using calibration graphs based on deionized water. The relative standard deviations for these analytes were 1–2% in deionized water and 2–4% in lake water. The detection limits were 9 ng/ml for nitrobenzene and 15 ng/ml for the other three analytes.

Acknowledgements

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