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# Design of automated solid-phase microextraction for trace analysis of organic compounds in aqueous samples

Ralf Eisert, Janusz Pawliszyn*

*Department of Chemistry, University of Waterloo, Waterloo, Ont. N2L 3G1, Canada*

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**Abstract**

The evaluation of static absorption and three different agitation modes including magnetic stirring, fibre vibration method, and a self constructed flow-through extraction cell for solid-phase microextraction (SPME) of trace organics is presented in this study. A mixture of analytes which contained five pesticides: ametryn, parathion, prometryn, simetryn, and terbutryn was selected for direct absorption from aqueous samples using a 100 μm poly(dimethylsiloxane) fibre. The absorption–time profiles and precisions were studied under different conditions using spiked aqueous samples. A commercially available autosampler was investigated for static absorption and the fibre vibration method. The efficiency of the automated SPME analysis methods developed was compared to the classical extraction of magnetically stirred samples and on-line SPME using the recently developed flow-through cell concept. Besides the kinetic aspect of a fast absorption characterizing the efficiency of the absorption mode selected, the precision was studied for all systems investigated to estimate the analytical accuracy which could be achieved by using these absorption techniques. In general, all agitation modes showed a precision for most of the compounds below 5% R.S.D. The fibre vibration technique significantly increases the precision and sample output of the automated SPME–GC method for the analysis of semi-volatile compounds. © 1997 Elsevier Science B.V.

*Keywords:* Solid-phase microextraction; Extraction methods; Automation; Pesticides

## 1. Introduction

The number of applications for solid-phase microextraction (SPME) has been steadily increasing. SPME has been applied to the extraction of organic compounds from different matrices including air [1], water [2] and soil [3]. However, there is a need for the automation of the entire analysis. The reliability of SPME analysis for pesticides at trace concentration levels was demonstrated in a round robin test [4]. The results obtained indicate that SPME is a valid method for the determination of trace amounts of semi-volatile pesticides in water, even though the method was used 'as is', without any attempt to optimize the conditions especially the automation of the entire method. So far, SPME has been automated only for the static absorption mode using a commercial autosampler [5] which is a time consuming technique as far as equilibrium concentrations on the fibre should be achieved.

The direct extraction of the aqueous matrix, where the SPME fibre is directly exposed to the aqueous matrix, is usually used for semi-volatile compounds, e.g., polar pesticides such as triazines [6]. The diffusion coefficients in water are lower by several orders of magnitude compared to extraction from the

*Corresponding author.


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gas phase. Therefore, the mass transport in solution is slow compared to gas phase transport. In general, the time necessary to reach the equilibrium is higher by an order of magnitude under static absorption conditions when extracting aqueous solutions. There are other parameters, e.g., mixing the aqueous solution, which have a pronounced effect on the mass transport of the analyte into the fibre and therefore the total amount extracted after a certain time of exposure [7]. In general, there are two different agitation techniques under investigation. Firstly, the mixing of the aqueous solution surrounding the sample, e.g., magnetic stirring or a flow-through system. Secondly, moving the fibre itself in the aqueous sample, e.g., fibre insertion/retraction or vibration. The fibre insertion/retraction technique [8] represents the basic principle on which the new fibre vibration method is based. Sonication represents a mixture of both options which is considered very efficient. However, sample heating represents a major disadvantage of this technique [7].

Recently the new SPME III autosampler (Varian) was commercialized, including an option for agitation during the extraction (fibre vibration). This instrument is expected to increase the efficiency of the entire SPME method, similar to what has already been shown for magnetic stirring with manual SPME [7,8]. This will reduce the total absorption time for one sample and increase the precision of the entire method. Thus, the equilibrium will be achieved faster. One should keep in mind that under equilibrium conditions, the amount of analyte on the fibre is constant. Consequently, precision achieved under these conditions is expected to be the optimum.

In this study four different extraction processes are compared with respect to the efficiency (equilibrium time) and reproducibility (precision) of the SPME method. The potential of automated SPME using the fibre vibration method is evaluated in comparison to the manual sampling mode using either magnetic stirring or the new developed flow-through extraction cell. This cell is based on the concept that agitation is most efficient when the static layer surrounding the fibre is very small. Thus, a high linear flow should be sufficient for this purpose. The flow-through cell is expected to show similar characteristics (equilibrium time) to magnetic stirring.

## 2. Experimental

### 2.1. Materials

The pesticide standards of ametryn, parathion, prometryn, simetryn and terbutryn were purchased from Supelco (Bellefonte, PA, USA). They were of purity ≥96% and used as received. Methanol (HPLC grade quality) was from EM Science (Gibbstown, NJ, USA). Water was obtained from a Barnstead/Thermodyne NANO-pure ultrapure water system (Dubuque, IA, USA). Single standards with a concentration of 600 ng/μl were prepared for each compound using methanol (HPLC grade) as a solvent. A standard mixture using methanol (HPLC grade) was prepared containing the organic compounds at a concentration of 60 ng/μl. The aqueous samples were spiked with this solution to a final concentration of 300 μg/l.

### 2.2. Gas chromatography

Gas chromatographic investigations were carried out using a Varian 3400 CX gas chromatograph (Varian, Walnut Creek, CA, USA) equipped with a flame-ionization detection (FID) system. All injections with the SPME unit were performed automatically using a Varian 8200 autosampler controlled by the Varian Star version 4.5 software. A SPB-5 column (30 m×0.25 mm I.D., 0.25 μm film thickness) from Supelco was used for the GC separation. A septum programmable injector (SPI) held at 260°C was equipped with a SPME liner from Varian and 11.5 mm ID LB-2 septa from Supelco. The following temperature program was used: 50°C initial column temperature held constant for 3 min, then ramp at 10°C/min to 260°C, then hold at 260°C for 1 min. FID was operated at 300°C.

### 2.3. Solid-phase microextraction

A SPME fibre holder for automatic use from Supelco was used for our SPME investigations. The 100 μm poly(dimethylsiloxane) (PDMs) fibre from Supelco was used for all experiments. A VWR Scientific digital magnetic stirrer type 400S (San Francisco, CA, USA) was used for the SPME

experiments with stirred aqueous samples. The SPME fibre is placed off-center in the vial so the fluid flows past the fibre perpendicular to the fibre axis. For achieving optimum mixing of the liquid phase the speed of the magnetic stirrer was set to the following values. It was operated at 900 r.p.m. for the 16-ml vials using an octagonal 1/2 in. length×5/6 in. (1 in.=25.4 mm) diameter magnetic stirbar and at 2000 r.p.m. (representing the maximum setting) for the 2-ml vials using a 2 mm diameter×7 mm length magnetic stirbar (VWR Scientific).

Two types of vials were used in this study. The experiments with 2-ml vials were carried out with a clear screw cap 2-ml 12×32 mm vial (Supelco) sealed with red PTFE-faced silicone 8-mm septa and screw caps with 12-mm hole (Supelco). These vials are suitable for the 48-vial autosampler carrousel (Supelco). The experiments with 16-ml vials were carried out using serum type reaction 16-ml 25×50 mm vials from Supelco sealed with slit 20-mm septa made of silicone with PTFE lining and open center 20-mm crimp caps. These vials are suitable for the 12-vial autosampler carrousel (Supelco).

The 16-ml vials were filled with 12 ml of aqueous sample while the 2-ml vials were filled with 1.4 ml. For equilibration time experiments, water from a NANO pure water purification system was spiked with standard solutions containing 300 μg/l of each compound in methanolic solution. The overall methanolic concentration during experiments was held constant at 0.5% (v/v). Thus, no significant influence of methanol was observed. Normally the total amount of analyte extracted is reduced at high MeOH concentrations [2,5].

The procedure for SPME is very easy and can be summarized by the following steps. Firstly, the fibre is exposed to the sample for a period described below. Secondly, the fibre is withdrawn from the sample and introduced into the GC injector, where the thermal desorption occurs. All SPME steps were performed by the SPME 8200 autosampler except for the absorption using magnetic stirring and the flow-through extraction cell. An absorption time of 30 min was used in this study to determine the precision. This time does not necessarily represent the equilibration time for all compounds. However, most of the target analytes are at equilibrium or at least very close to it. A further increase of the total extraction time would not increase the precision for most analytes. In addition, 30 min absorption time was selected for practical reasons. The extraction time is similar to the total GC run time improving the sample output for routine analysis. Thermal desorption of the pesticides was carried out for 3 min. After this period no significant blank values were observed. The absorption–time profiles were studied for all compounds using all different absorption modes between 2 and 120 min total extraction time.

Most of the experiments were performed using the direct absorption from the aqueous sample either by autosampler control (static and fibre vibration method) or manual SPME absorption control (magnetic stirring). Using magnetic stirred aqueous samples the fibre is exposed close to the vortex formed in the vial. In this case only the SPME fibre dips into the aqueous sample. The syringe needle where the SPME fibre is drawn into when the plunger is retracted for piercing the septum of a vial or GC injector port is positioned outside the aqueous sample. This process can be easily controlled with manual injection. If the autosampler is used for the SPME absorption process the depth of the fibre and the needle in the vial are software controlled and cannot be varied by the usual software control panel. Therefore, part of the syringe needle may dip into the aqueous sample. During the vibration, when using the fibre vibration method, water may flush inside the needle. Thus, during the desorption a few droplets of water will be injected to the GC system causing peak broadening effects, which influences the precision. The distance between the end of the needle and the exposed fibre is only 1–3 mm using the autosampler control. It is nearly impossible to adjust the sample volume exactly that only the fibre will be exposed to the aqueous sample. By modifying the 8200.ini file of the software a change of the default parameters for fibre adjustments is possible. The vibration frequency of the fibre was measured using a stroboscope. The average frequency determined was $\nu = 1100\ s^{-1}$.

Calibration was performed by injecting liquid standards containing the organic compounds in an organic solvent such as methanol for calculating the

total amounts absorbed by the fibre. Therefore, the initial injector temperature was set to 50°C.

Fig. 1 shows an illustration of all four absorption processes used in this study and defined in Table 1.

### 2.4. Flow-through cell equipment

The newly developed flow-through extraction cell was connected to the aqueous sample as shown in Fig. 2. The sample is pumped by an IsoChrom LC pump from Spectra-Physics (San Jose, CA, USA) at 10 ml/min unless otherwise indicated. Total sample volume was 10 ml. The precision was checked using 30 min absorption time. Thus, the sample volume (10 ml) passed the extraction cell 30 times during this experiment. When using the HPLC pump a very constant flow can be achieved in the extraction cell. This is necessary to obtain a reproducible high linear flow around the fibre inside the extraction chamber. The measured precision of the flow is 0.2% R.S.D. when the pump was operated at 10 ml/min. Therefore, the time necessary to fill a 25-ml volumetric flask was determined six times ($n=6$). The fibre is mounted in the SPME device of the autosampler after the extraction from the Valco-tee unit.

Firstly, the fibre is exposed to the Valco three-way tee extraction unit by sealing with a 0.4 mm I.D. M-2B GC ferrule (Supelco) and a slipfree SFE connector from Keystone Scientific (Bellefonte, PA, USA). The inlet of the Valco-tee opposite to the SPME device inlet was connected to a 5-cm piece of PTFE tubing 0.3 mm I.D. ×1.58 mm O.D. (Supelco) which hosts the SPME fibre during the absorption step. The inlet PTFE tubing itself is connected via a zero volume 1/16 in. internal union to the 0.1 mm I.D. stainless-steel tubing which was connected to the HPLC pump outlet. The outlet of the Valco tee was connected by a two-piece 1/16 in. polyether ether ketone (PEEK) fingertight nut (Supelco) to a PTFE tubing 0.5 mm I.D. ×1.58 mm O.D. (Supelco). A 40-ml amber sample vial (Supelco) sealed with a PTFE/silicone septa (Supelco) containing the aque-

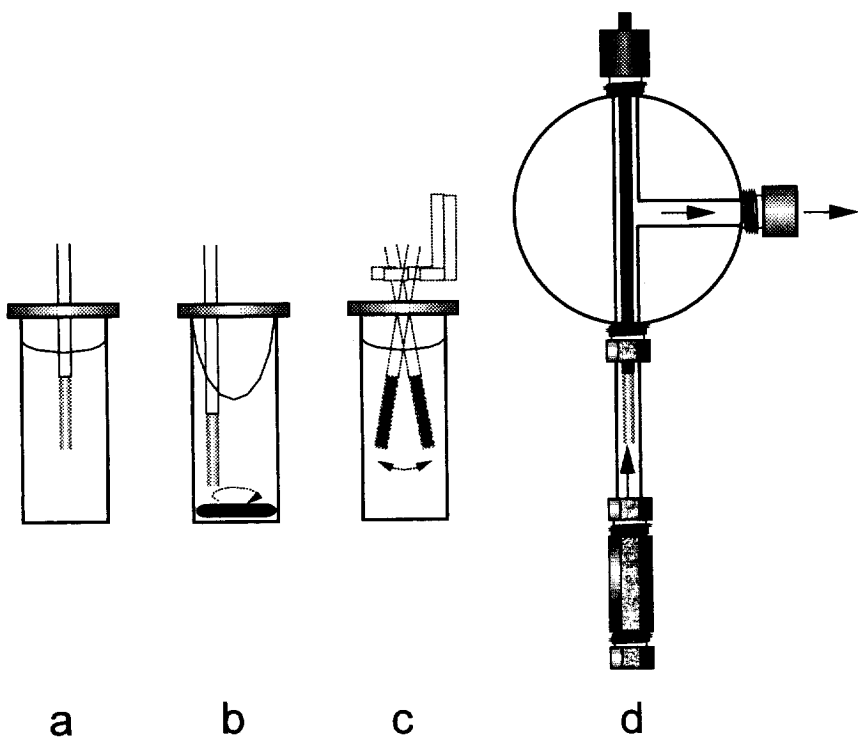


Fig. 1. Illustration of the four different absorption techniques used for SPME: (a) static absorption, (b) magnetic stirring of the sample, (c) fibre vibration method and (d) an on-line flow-through cell extraction.

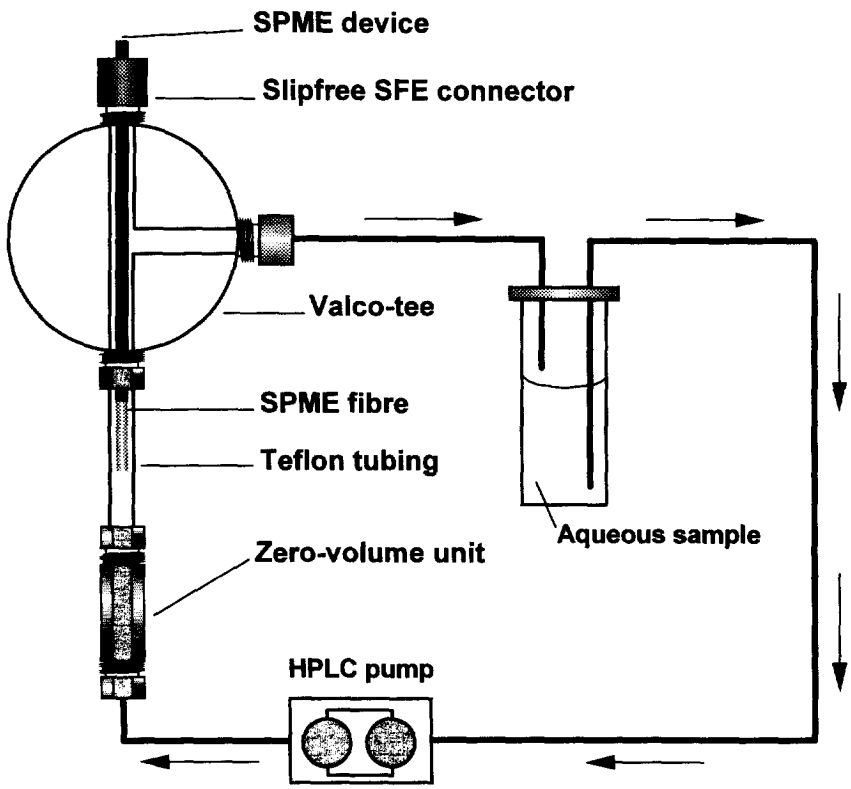


Fig. 2. Instrumental set-up for the on-line flow-through cell. The SPME fibre hosts in a Valco-tee unit sealed by a vespel ferrule. The aqueous sample is pumped by a HPLC pump from the sample vial through the extraction cell and back to the reservoir (closed loop).

ous sample was connected to the system as illustrated in Fig. 2. The HPLC pump was started immediately after exposing the fibre into the flow-through cell. The extraction was performed by simply pumping the sample in a closed loop. After the extraction period the fibre was withdrawn immediately from the sample and mounted to the autosampler unit, where it was introduced automatically into the GC injector, and analytes were thermally desorbed. An absorption time of 30 min was used in this study for evaluating the precision of the flow-through system. At this time equilibrium was nearly established for most of the organic compounds investigated.

## 3. Results and discussion

SPME is based on the equilibration of the analyte concentration between the sample and the stationary phase of the SPME fibre, based on the analyte distribution constants. Therefore, the diffusion of the analyte into the polymer represents the most important step for the mass transfer in the fibre. The accumulation of analyte in the fibre is controlled by diffusion in the (aqueous) matrix and the diffusion into the fibre. Depending on the extraction conditions, i.e., dynamic mode, the diffusion in the aqueous sample becomes nearly negligible. Without intensive mixing of the aqueous solution, the equilibration time increases considerably. In the static case, transport of the analyte is limited by diffusion in both the aqueous phase and the aqueous layer surrounding the fibre surface; during the absorption process, the concentration outside this layer steadily decreases, thus reducing the flux into the fibre. In the dynamic case using agitation, a thin layer of water still remains on the surface of the fibre

coating. Thus, the final equilibration time is determined by diffusion through this layer but no longer by the diffusion of the analytes in the aqueous sample. A typical example for the influence of intensive agitation of the aqueous solution on the extraction efficiency is shown in Fig. 3. When using static absorption conditions only for the analysis of compounds with low coating/sample distribution constants, $K$ values, such as simetryn, equilibrium is reached within 2 h (see Fig. 3a). The equilibrium is reached for all four $s$-triazines after 35 min when using agitation during the absorption (see Fig. 3b). Consequently, a high precision with R.S.D. values below 5% was achieved.

### 3.1. Equilibration times

Absorption–time profiles were studied for four different absorption modes (static, magnetic stirring, fibre vibration method and the new flow-through cell). Equilibrium time is defined as the time after which the amount of extracted analyte remains constant and corresponds within experimental error to the amount extracted at an infinite extraction time. Firstly, the extraction was performed under static conditions. Fig. 3a shows the results observed for four $s$-triazines and parathion under static absorption conditions. Within 120 min total extraction time only simetryn had reached equilibrium concentrations (see also Table 1). In a second experiment magnetic stirring was studied. This is the classical method for achieving faster mass transport into the fibre, thus reducing the equilibrium times (see Fig. 3b, Fig. 4b). The experiments were performed for both vial types (2 ml and 16 ml). In general, the velocity of the water sample was high when using large vials (16 ml) with an octagonal magnetic stirbar which facilitates a more effective agitation by forming a vortex. Thus, the equilibrium for all five compounds was reached very fast (within 50 min) using this absorption mode. In a third sequence the new fibre vibration technique using the autosampler was studied and compared to static absorption and the magnetic stirring extraction experiments. Figs. 3c and 4c

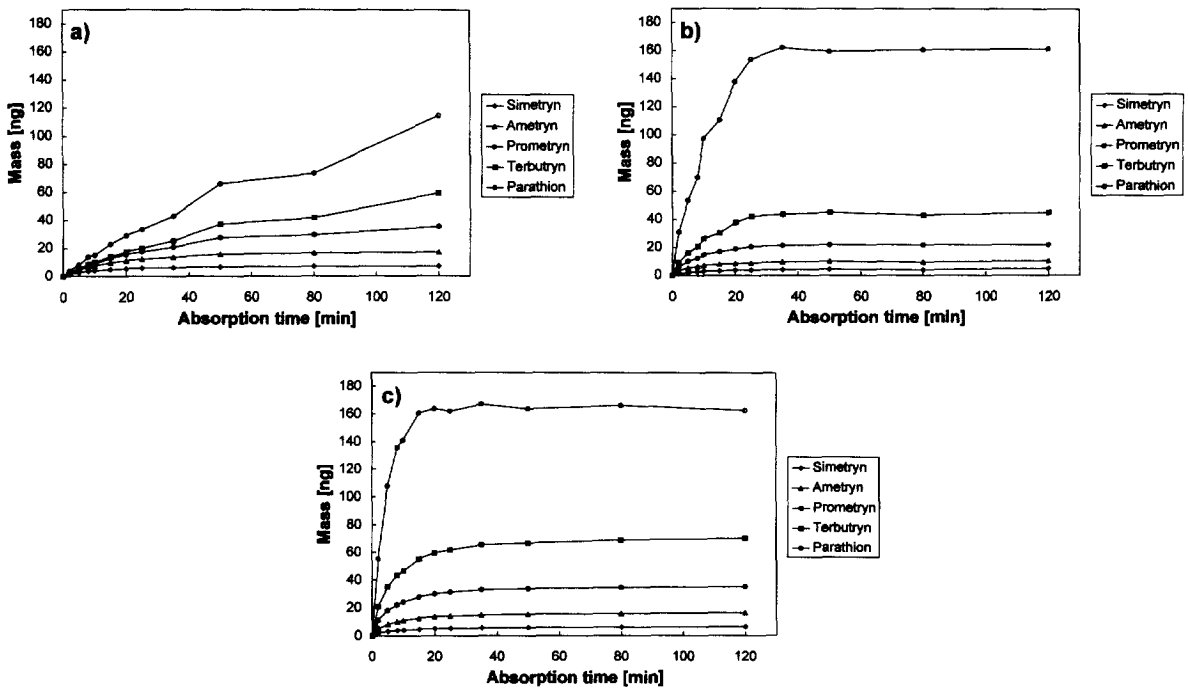


Fig. 3. Absorption–time profiles for four $s$-triazines and parathion using (a) static absorption conditions, (b) magnetic stirring and (c) fibre vibration method (for 2-ml vials). The equilibrium is reached substantially faster for agitation techniques.

Table 1
Equilibration times for all investigated compounds obtained under different absorption modes

| Number | Compound | Molecular formula | Molecular mass | $K_{ow}$[a] | Solubility in water (mg/l)[b] | Equilibration time (min) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | 2 ml sta[c] | 2 ml stir[c] | 2 ml vib[c] | 16 ml sta[d] | 16 ml stir[d] | 16 ml vib[d] | 40 ml flow[e] |
| 1 | Simetryn | $C_8H_{15}N_5S$ | 213.32 | 347 | 450 | 60 | 30 | 30 | 80 | 25 | 40 | 30 |
| 2 | Ametryn | $C_9H_{17}N_5S$ | 227.35 | 955 | 185 | >120 | 35 | 30 | >120 | 30 | 50 | 30 |
| 3 | Prometryn | $C_{10}H_{19}N_5S$ | 241.37 | 3236 | 33 | >120 | 30 | 35 | >120 | 30 | 80 | 35 |
| 4 | Terbutryn | $C_{10}H_{19}N_5S$ | 241.37 | 5495 | 25 | >120 | 30 | 35 | >120 | 50 | 80 | 35 |
| 5 | Parathion | $C_{10}H_{14}NO_5PS$ | 291.27 | 6761 | 11 | >120 | 30 | 20 | >120 | 35 | 35 | 25 |

[a] Data were obtained for compound Nos. 1–5 from Ref. [13].
[b] Data were obtained for compound Nos. 1–5 from Ref. [14] at 20°C.
[c] The results were obtained for 2-ml vials with 1.4 ml sample volume.
[d] The results were obtained for 16-ml vials with 12 ml sample volume.
[e] The results were obtained for 40-ml vials with 10 ml sample volume using the flow-through extraction cell.
Abbreviations: flow=SPME fibre is exposed to the flow-through extraction cell; sta=static absorption with no agitation of the aqueous sample; stir=mixing of the aqueous sample using magnetic stirring; vib=fibre vibration method using autosampler agitation.

summarize the results for both vials. The fibre vibration method works very efficiently when using the small 2-ml vials. The results show a faster equilibrium for the 2-ml vials than for the 16-ml vials. The vibration of the fibre is almost transferred to the SPME fibre in the sample. Equilibrium was achieved for all five compounds within 35 min when using the 2-ml vials. Even the 16-ml vials show an efficient extraction, but it takes more than 80 min for all compounds until the equilibrium was achieved (see also). So far, Fig. 3b,c and Fig. 4b,c show a cross-over relationship, i.e., the 2-ml vials are more

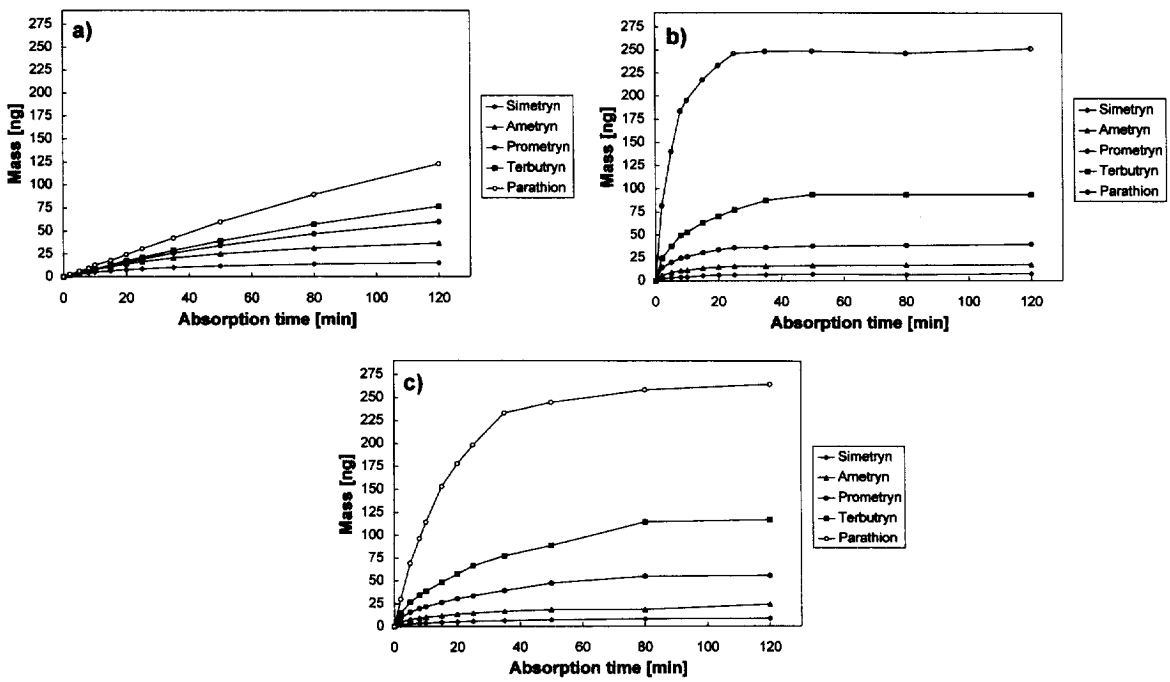


Fig. 4. Absorption–time profiles for four *s*-triazines and parathion using (a) static absorption conditions, (b) magnetic stirring and (c) fibre vibration method (for 16-ml vials). The equilibrium is reached substantially faster for agitation techniques.

efficiently extracted by the fibre vibration method while the 16-ml vials are more successful when using the classical magnetic stirring of the aqueous samples. This effect might be explained by the vortex of the water sample which is present when using the magnetic stirring. The dimension and design of the octagonal stirbar allows the establish of a vortex in the aqueous sample when a 16-ml vial is used. In general, there is only a very small vortex observed for the 2-ml vials. This is mainly a result of the stirbar design and the maximum rotation speed achieved by the magnetic stirrer. Therefore, the speed of agitating the sample in the vial was estimated to be lower, which effects the extraction efficiency of these vials. The agitation using the fibre vibration method is not as effective when performed in the 16-ml vials. The dampening of the fibre is different to the 2-ml vials and not very reproducible. Therefore, longer equilibration times and less accurate results will be observed, as is shown in Tables 1 and 2, respectively. However, the results are within the limits of other more traditional extraction techniques such as SPE (solid-phase extraction) or LLE (liquid–liquid extraction) [10]. The equilibration takes up to 80 min using the fibre vibration method in combination with 16-ml vials. However, the equilibrium is reached for the small vials within 35 min. We can conclude from these results, that the fibre vibration method works very efficiently. Furthermore, we have to keep in mind that for these experiments there was no operator necessary. The entire SPME analysis is automated completely for the fibre vibration method. This shows the great potential of this technique for routine analysis. The analysis of pesticides seems to be a promising field for the automated SPME–GC technique. The total amount which can be extracted by the fibre differs for the 2-ml and 16-ml vials. Thus, the influence of the sample volume has a pronounced effect on the total amount extracted by the fibre. Fig. 3b,c and Fig. 4b,c show a decrease by a factor of 1.6 when 2-ml vials are used for the extraction. There will be more analyte extracted using 16-ml vials. However, sensitivity is only slightly increased. One can conclude that for large sample volumes magnetic stirring is very effective. However, very precise results can be obtained using 2-ml vials and the fibre vibration technique which shows a higher efficiency when these vials were used. Furthermore, the efficiency of the fibre vibration method when using the 2-ml vials might be explained by the convection effect in the aqueous sample. Convection is more successfully established in the 2-ml vials than in the 16-ml vials which reduces the static layer of water remaining on the fibre. Thus, diffusion of the analyte in the fibre is substantially faster.

In general, the results obtained for the 2-ml and 16-ml vials show a similar trend when using different extraction conditions, except for the total amount extracted. Sonication is mentioned in addition to the extraction processes used in this study. As expected and shown in the past [7] it is a very efficient sample agitation technique for SPME. For example, very volatile organic compounds (VOCs) such as toluene, equilibrate in less than 1 min, which is very close to the theoretical prediction. Sonication was not investi-

Table 2
Precision achieved with autosampler SPME–GC–FID method of five selected pesticides using 2-ml and 16-ml vials and three different absorption modes

| No. | Compound | Precision (R.S.D. [%]) | | | | | |
|---|---|---|---|---|---|---|---|
| | | Static[a] | Magnetic stirring[a] | Fibre vibration[a] | Static[b] | Magnetic stirring[b] | Fibre vibration[b] |
| 1 | Simetryn | 0.9 | 2.6 | 1.4 | 1.2 | 4.2 | 3.2 |
| 2 | Ametryn | 3.0 | 1.3 | 1.1 | 2.6 | 0.7 | 3.4 |
| 3 | Prometryn | 7.0 | 1.1 | 1.1 | 4.1 | 0.7 | 3.9 |
| 4 | Terbutryn | 7.8 | 2.9 | 1.0 | 4.9 | 3.1 | 4.9 |
| 5 | Parathion | 11.3 | 2.3 | 0.8 | 5.4 | 0.8 | 4.1 |

[a] Precision achieved for 2-ml vials from five repetitive injections of 1.4 ml samples ($n=5$). The concentration was 300 μg/l for each compound. A 30 min absorption time was used.

[b] Precision achieved for 16-ml vials from five repetitive injections of 12 ml samples ($n=5$). The concentration was 300 μg/l for each compound. A 30 min absorption time was used.

gated in this comparison. The major disadvantage of the sonication technique is sample heating during the extraction and the loss of analytes caused by sonication induced decomposition, which has been reported in the literature [7]. To date, sonication is not available for automated SPME and the number of applications using sonication is negligible.

### 3.2. Precision

The results of checking the precision for five repetitive injections ($n=5$) when using the 2-ml vials are summarized in Table 2. During the 30 min absorption time, most of the investigated compounds reach their equilibrium under agitation conditions. The precision obtained for the 2-ml vials when using the fibre vibration method can be considered optimal. The R.S.D. was <3% for all compounds when agitation was used.

The results of checking the precision for five repetitive injections ($n=5$) when using the 16-ml vials are summarized in Table 2. The precision obtained for static absorption conditions is as poor as observed for the 2-ml vials. For semi-volatile compounds such as *s*-triazines the precision is increased by using magnetic stirring in comparison to static absorption.

Furthermore, even the peak shape and the poor resolution could have a significant influence on the precision of the direct absorption from the aqueous phase (injection of water when using the fibre vibration method). The loss of analytes into the headspace is more significant for volatile compounds which are not determined in this study. However, headspace sampling using the automated SPME and different septa is expected to show good results for VOCs. The precision for the semi-volatile compounds is still good (R.S.D. <5%). The method used for the GC determination works best for the semi-volatile compounds. The temperature of the samples might be increased if they are positioned for a longer time close to the hot GC injector (autosampler conditions) which is dependent on the absorption time used. The extraction yield of semi-volatiles is not significantly affected by this phenomena. However, VOCs and especially in combination with headspace sampling are expected to show a significant effect. Keeping in mind the fast accumulation of VOCs on the fibre, the temperature effect might be less significant than expected.

To evaluate, whether the vibration of the vial itself contributes to the efficiency of the fibre vibration method, the following experiments were investigated. The 16 ml vials exactly fit into the autosampler carrousel. Headspace vials with a smaller radius than the 16-ml vials were used to find out whether the vibration of the vial contributes to the amount extracted. The same experiments were investigated for the 2-ml vials which are usually show some vibration in their carrousel positions. Therefore, they were fixed by a piece of paper pressed between the vial and the carrousel wall. For both vials no significant effect was observed. The vibration of the vial which shakes the aqueous sample does not significantly contribute to the entire agitation process. There was no significant competition observed between moving the fibre and the vibration of the sample.

No significant absorption of the investigated analytes on the magnetic stirbar was obtained which might lower the amount extracted when using the magnetic stirring method.

### 3.3. Flow-through cell experiments

Fig. 5 shows the absorption–time profile for four *s*-triazines and parathion when using the flow-through cell. This absorption process shows a similar efficiency as the agitation techniques shown in Figs.

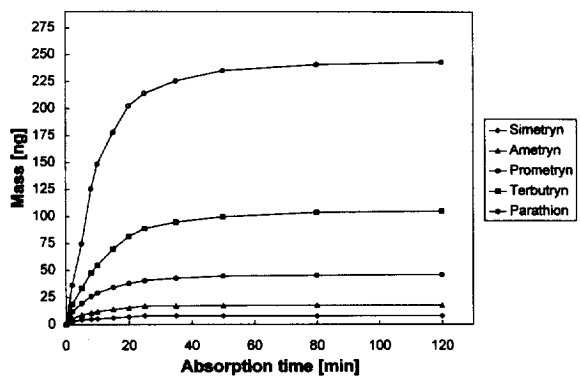


Fig. 5. Absorption–time profiles for four *s*-triazines and parathion using the flow-through cell for SPME absorption and 10 ml sample volumes. The sample was circulated at a flow-rate of 0.1 ml/min.

Table 3
Precision achieved with the autosampler SPME–GC–FID of five selected pesticides using the flow through cell and 10 ml samples

| No. | Compound | Precision (R.S.D. [%]) | |
|---|---|---|---|
| | | Flow 10 ml/min[a] | Flow 0.1 ml/min[b] |
| 1 | Simetryn | 5.3 | 3.4 |
| 2 | Ametryn | 4.4 | 3.9 |
| 3 | Prometryn | 3.1 | 4.9 |
| 4 | Terbutryn | 5.7 | 7.9 |
| 5 | Parathion | 4.6 | 5.5 |

[a] From five repetitive injections ($n=5$) and 30 min absorption time. The concentration was 300 μg/l for each compound. The flow-rate was 10 ml/min.
[b] From five repetitive injections ($n=5$) and 30 min absorption time. The concentration was 300 μg/l for each compound. The flow-rate was 0.1 ml/min.

3 and 4 for the same compounds. The instrumental set-up used for these experiments is shown in Fig. 2. In general, the samples were pumped at a flow-rate of 10 ml/min, achieving a high linear flow of the aqueous sample inside the PTFE tubing where the SPME fibre is positioned during the absorption. The precision ($n=5$) achieved for this technique is shown in Table 3. The results in Table 3 show a satisfactory precision for all compounds investigated (R.S.D. <8%). The flow-through cell was operated at different flow-rates to investigate the influence of laminar and turbulent flow characteristics on the absorbed amount.

Fig. 6 shows the results obtained under different

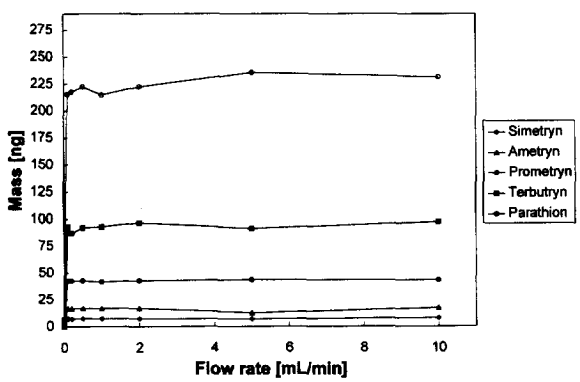


Fig. 6. Influence of varying the flow-rate from 0–10 ml/min on the absorbed mass of four *s*-triazines and parathion using the SPME flow-through cell and a sample volume of 10 ml. The absorption time was kept constant at 30 min during this experiment.

flow-rates varied from 0 to 10 ml/min. A flow of 0.1 ml/min was the minimum flow which could be achieved with a good precision (R.S.D. <1%). The five pesticides show no significant effect on the varied flow. The flow surrounding the fibre is still turbulent at 0.1 ml/min. Thus, no significant effect on the amount extracted was observed. This is a result of the small distance (only a few micrometer) between the surface of the fibre and the tubing. The laminar flow range is expected to show an influence on the amount extracted with increasing flow-rate, which was observed in a similar study on flow-through on-line SPME published recently [9]. The results obtained for the precision measured under different flow-rates (0.1 ml/min and 10 ml/min) are very similar to those demonstrated in Table 3.

A calibration including a concentration range from 3 to 300 ng/ml was studied for all five pesticides using the new flow-through cell. Five different concentrations were selected for this external calibration. Each standard was injected three times. There was no significant absorption effect observed on the tubing or elsewhere in the system. The calibration curves of all compounds were linear over at least two orders of magnitude. The square of correlation coefficient was $r^2>0.99$. Thus, the flow-through extraction cell shows good potential for on-line monitoring using external calibration methods.

Furthermore, two different operating techniques of the flow-through cell were evaluated. The circulation approach (for set-up see Fig. 2) was compared to an open system where the sample is only once pumped through the cell. Therefore, 30 ml samples with equal concentrations of 300 ng/ml were investigated in comparison to the circulated sample system which is described above. A flow-rate of 0.1 ml/min and 30 min absorption time were selected for this comparison. There was no significant effect observed on the extraction yield of the five pesticides. The variation of the results was within the limit of variation characterized by the precision. It is expected that compounds with high $K$ values would show a depletion of the sample concentration as a result of their high affinity to the fibre coating. This effect addresses the need for relatively high sample volumes to be analyzed. In this study a sample volume of 10 ml was sufficient. However, analytes

with high $K$ values (>50 000) can be completely released from the (aqueous) matrix when using small sample volumes (<10 ml).

The newly developed flow-through extraction cell shows a high efficiency for all compounds studied and a great potential for on-site monitoring of flowing aqueous samples, as has been recently published for on-line SPME systems based on a flow-through cell by Eisert [9].

## 4. Conclusions

The high precision achieved when using the fibre vibration method and the small (2 ml) vials may be attractive for many applications where the sample volume is limited, e.g., analysis of cloud, rain, or sediment water. However, the total amount which can be absorbed on the fibre is influenced by the sample volume, especially for compounds with high $K$ values [11] where exhaustive extraction might occur. Magnetic stirring still shows a slightly faster equilibration for compounds with higher $K$ values as compared to the fibre vibration method.

One can conclude from all the different absorption modes used in these experiments that the efficiency of SPME can be optimized by a high linear flow at the surface of the fibre. Thus, the stationary layer of water surrounding the fibre is kept to a very small radius, which decreases the time necessary for the diffusion of the analytes. Technically this can be achieved by either mixing the aqueous sample (magnetic stirring or flow-through) or moving the fibre itself relative to the water (fibre vibration method). The results obtained for all three different agitation methods show a similar trend as expected from this concept. The fibre vibration technique improves the efficiency for the automated SPME–GC for semi-volatile analytes. The sample output is increased while the precision obtained is still high. The long equilibrium times, usually necessary for semi-volatile pesticides, are significantly reduced which further improves the application of the SPME method for routine analysis.

SPME is mainly restricted to thermally stable compounds but additional derivatisation steps in the aqueous sample or directly on the fibre and recently developed SPME–HPLC coupling [12] extend the application range. The concept of the newly developed flow-through extraction cell where the extraction and desorption steps are combined seems to be an interesting principle which will be applied for thermally labile compounds using a new SPME–HPLC interface in the future. The flow-through cell technique could be useful for accommodating absorption and desorption in an automated SPME–HPLC system. Thus, the application range of automated SPME can be easily increased for thermally labile compound classes.

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