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Journal of Chromatography A, 999 (2003) 175–184

JOURNAL OF
CHROMATOGRAPHY A

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Rapid determination of polycyclic aromatic hydrocarbons in sewage sludges using microwave-assisted solvent extraction Comparison with other extraction methods

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Abstract

The presence of toxic organic micropollutants in municipal sewage sludges is a major problem on account of risks associated with the agricultural use of the sludges and therefore maximum tolerance limits are imposed. The aim of our study was to develop a reliable and fast analytical procedure for the determination of polycyclic aromatic hydrocarbons in sewage sludges, using focused microwave-assisted extraction. Optimization of the extraction conditions was performed on real matrices. The results of a 2^3 factorial design showed that extraction time was the only influential factor. The selected conditions (30 W, 10 min, 30 ml solvent) were used for real sludges and a certified marine sediment, leading to recoveries of between 56 and 75%. Results of an interlaboratory test confirmed these values. Finally, the technique was compared to traditional techniques (Soxhlet, sonication), and the more recent pressurized liquid extraction technique. Focused microwave-assisted extraction remains an attractive alternative technique due to its rapidity, the low solvent volume required and its moderate investment cost.

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Keywords: Factorial design; Microwave-assisted extraction; Extraction methods; Interlaboratory studies; Validation; Polynuclear aromatic hydrocarbons

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs) are well known environmental pollutants present at low concentrations; they are formed during combustion of carbonaceous materials at high temperature. Owing to their mutagenic and carcinogenic potential [1,2], they have been determined in several matrices, particularly waters, soils and sediments. Urban areas

are important sources of PAHs due to domestic fuel combustion, industrial emissions, car exhausts and natural background atmospheric deposition. As a consequence, rainwater and wastewater draining from urban areas into the sewerage system contain relatively large amounts of these pollutants. Due to their hydrophobicity, PAHs are primarily adsorbed on the biomass during activated sludge treatment of the wastewaters, and they remain partly undegraded due to their biological recalcitrance [3,4]. This results in the formation of sewage sludges that typically contain between 0.02 and 25 mg kg⁻¹ dry mass (dm) of each individual PAH [5–9]. The

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concern about their presence in sewage sludges is quite recent, and has resulted in the establishment of maximum acceptable concentrations for some PAHs in sludges to be utilized in agriculture. In France, the recent regulation of 8 January 1998 imposes a maximum acceptable limit for three PAHs: fluoranthene ($5.0 \text{ mg kg}^{-1} \text{ dm}$), benzo[*b*]fluoranthene ($2.5 \text{ mg kg}^{-1} \text{ dm}$), benzo[*a*]pyrene ($2.0 \text{ mg kg}^{-1} \text{ dm}$). A new draft directive of the European Union has regulated the total concentration of PAH allowed in sewage sludges for agriculture to $6.0 \text{ mg kg}^{-1} \text{ dm}$ [10].

Due to the large amounts of sewage sludges generated daily, there is a need for a fast analytical method to accurately determine PAHs. This study was undertaken to evaluate the use of microwave-assisted extraction (MAE) for that purpose. This technique has been successfully used for the rapid determination of PAHs from solid environmental matrices [11]. Thus, pressurized MAE (pMAE) using hexane–acetone (1:1, v/v) enabled the extraction of PAHs from spiked soils [12,13], marine sediments [14], certified soil and sediment samples [15], and real urban atmospheric particulate samples [16]. pMAE with dichloromethane was recommended for PAHs from coal [17]. In another study, acetone was found to give the best results as compared to various hexane–acetone mixtures for extracting PAHs from highly contaminated soils [18]. Other extracting media have also been reported for pMAE, such as a micellar medium for spiked PAHs from marine sediments [19], *N*-methyl-2-pyrrolidone for PAHs from residual sludge and sediment samples [20], as well as acetone–water (4:1, v/v). Focused MAE (fMAE) has also been used for extracting PAHs from environmental matrices, with solvents such as dichloromethane, hexane–acetone (1:1, v/v) or dichloromethane–acetone (1:1) [13,21–25]. Wetting of the matrix before extraction was sometimes found to enhance extraction [22,23].

It appears that most applications deal with the use of pMAE. In the few studies that report the use of atmospheric pressure MAE, the samples investigated were soils or sediments. The only study involving fMAE for sewage sludge reported the extraction of polychlorinated dibenzo-*p*-dioxins and furans using toluene (60 W, 10 min) with recoveries ranging from 30 to 70% [26]. This study aims to optimise fMAE

conditions for the determination of PAHs in real sewage sludge samples.

2. Experimental

All experiments were done in triplicate.

2.1. Reagents and chemicals

The reagents in this study were all used in the purchased form without additional purification or alteration. Individual standard solutions (10 mg l^{-1} in acetonitrile) of the following PAHs were obtained from CIL Cluzeau (Paris, France): fluoranthene (Fluo), benzo[*b*]fluoranthene (BbF), benzo[*a*]pyrene (BaP). A standard stock solution containing the 16 PAHs (10 mg l^{-1} in acetonitrile) was also used (Sigma–Aldrich, Supelco Division, Saint-Quentin-Fallavier, France).

Analytical-reagent grade copper metal and nitric acid solution (68%) were supplied by Prolabo (Briare, France), as well as HPLC-grade acetonitrile, *n*-hexane and acetone. PAH purities were guaranteed between 97 and 99.7%. Other purities were stated to be higher than 99%. Deionized water was produced with a Milli-Q system from Millipore (Saint-Quentin-en-Yvelines, France).

Copper bars (0.5 cm long) were cut and immersed in 30% nitric acid for 30 s for activation. The bars were then cleaned sequentially with acetone and hexane. After this treatment, they were ready to be added to the sludge samples before extraction in order to remove sulphur interferences by sulfide formation.

2.2. Sewage sludge samples

Sewage sludge samples were obtained from two municipal wastewater stations near Paris, Achères and Valenton. Sludge samples from both plants were received for two different periods, and were supposed to be composite samples over the sampled period. Upon reception in the laboratory, the sludges were kept frozen to avoid any modification during storage. Before analysis, large samples (around 100 g) were taken and oven dried (40°C) to stable mass (48 h). This 40°C temperature was reported to be the

highest recommended temperature that could be used without losses by volatilization for PAHs [27]. The absence of water should make the matrix more accessible to the organic extractant solvent [22,28]; in addition it greatly facilitated the homogenisation of the samples. Homogenisation was performed with a mortar and a pestle, and the fraction greater than 2 mm was removed through sieving. The obtained samples were bottled in a polypropylene box and stored in the dark at room temperature. The samples received for the interlaboratory study had been pre-treated before reception in our laboratory.

2.3. Microwave-assisted extraction

fMAE experiments were performed with a Soxhlet 100 system (Prolabo, Briare, France). During the extraction, the sample was mixed using a magnetic stirrer. A 1 g aliquot of dried sewage sludge was weighed. The hexane–acetone (1:1, v/v) mixture (30 ml unless specified in the text) was then added to the sample and the solution stirred. Activated copper bars (1 g) were added to each sample just before extraction to remove sulphur by sulfide formation. Extractions were performed at 30 W for 10 min unless specified otherwise in the text. The extracts were then filtered (using Whatman filter-papers, 0.45 μm) to remove the copper and the extracted matrix.

2.4. Pressurized liquid extraction

Pressurized liquid extraction (PLE) was performed using an ASE 200 Accelerated Solvent Extractor (Dionex, Voisins le Bretonneux, France). Samples (1 g) were accurately weighed into 11 ml cells. For some experiments, the sludge sample was mixed with 4 g of either alumina or silica prior to extraction. Hydromatrix (inert diatomaceous earth) was added to the sample to fill the cell. The sample cells were then sealed and placed into the carousel of the system. Extractions were done using hexane–acetone (1:1, v/v). The operating conditions were as follows: 100 °C, with a 5 min heat-up time, 140 bar and a static period of 5 min (flush volume: 70%). The extracted analytes were purged from the sample cell using pressurized nitrogen for 1 min. These conditions have been shown to give high efficiency [29].

2.5. Soxhlet extractions

Extractions of dried sludge (1 g) were performed with a mixture of hexane–acetone (1:1, v/v) (250 ml). Activated copper bars (1 g) were added to the sample before extraction. Extractions were performed for 6 h (4–5 cycles per h).

2.6. Ultrasonic extractions

The ultrasonic bath was a P Selecta® (50 W). The dried sludge sample (1 g) was placed in a vessel with 20 ml hexane–acetone (1:1, v/v). The vessel was immersed (above the level of the solvent) into an ultrasonic bath for 15 min. After that period, the sample was filtered, and the solid residue extracted two more times using the same procedure. The extracts obtained each time were then combined.

2.7. Clean-up of the extracts

In all cases, the extracts were concentrated to approximately 5 ml using a rotary evaporator, and finally to approximately 2 ml under a gentle stream of nitrogen before clean-up. The purification step was required for the removal of lipids and fats. It is usually achieved using an adsorbent, either silica or florisil [30]. In our study, purification was performed on disposable solid-phase extraction (SPE) silica cartridges (Supelclean LC-Si, 1 g, 6 ml, supplied by Supelco, Saint-Quentin-Fallavier, France). A Visiprep vacuum manifold system (Supelco) was used. Conditions for the clean-up procedure were selected to enable the quantitative recovery of PAHs. This was performed using standard solutions of the pollutants, as well as spiked sludge extracts to take into account the presence of matrix material. Cartridges were conditioned with 5 ml of *n*-hexane. PAHs were eluted using 4 ml *n*-hexane and 4 ml hexane–dichloromethane (1:1, v/v). After combining both fractions, the solvent was completely evaporated under a gentle stream of nitrogen and the residue redissolved in 2 ml acetonitrile.

2.8. Liquid chromatography–fluorescence detection

Extracts were analysed using HPLC coupled to fluorimetric detection (FLD). The HPLC system

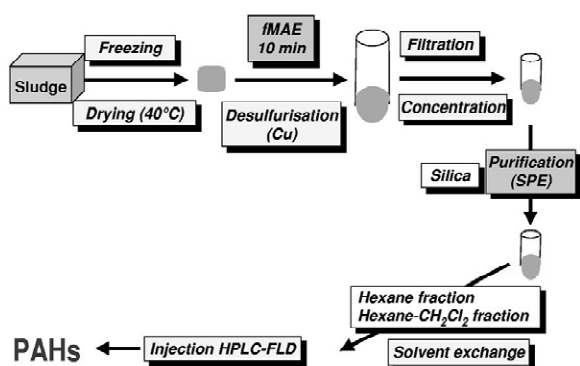


Fig. 1. Experimental procedure for the rapid determination of PAHs in sewage sludges.

consisted of a Varian 9010 high-pressure gradient pump, a Rheodyne Model 7125 injection valve equipped with a 20 μl loop, a Thermo Separation Science fluorimetric detector (FL3000), and a computer. Data analysis was performed using the TurboChrom TC4 Navigator. A Supelco LC-PAH column (250 \times 4.6 mm I.D., 5 μm particle size) was used, along with a pre-column (containing C_{18} -silica). Separation was performed using the following gradient: acetonitrile–water (60:40, v/v) for 5 min, followed by a 25 min ramp to 100% acetonitrile, this solvent being further maintained for 15 min. The total flow-rate was 1.5 ml min^{-1} . Detection was performed at selected excitation and emission wavelengths, respectively 230–410 nm for fluoranthene, 250–420 nm for benzo[*b*]fluoranthene and benzo[*a*]pyrene. Calibration was performed using standard solutions of PAHs in acetonitrile–water (60:40, v/v) in the range 25–200 $\mu\text{g l}^{-1}$. Standard solutions were analysed daily to check the fluorimeter sensitivity. The overall experimental procedure is depicted in Fig. 1.

3. Results and discussion

To allow comparison of the performances of the different techniques, dried sludge samples were extracted in all experiments. For the same reason, the same extraction solvent was used. A mixture of hexane–acetone (1:1, v/v) was chosen as it was

reported to be quite efficient for the extraction of pollutants such as PAHs from several environmental matrices [14–16]. Hexane allows solubilization of the PAHs, while acetone, due to its permanent dipole, is capable of creating dipole-induced dipole interactions with the π -electrons of the PAHs, thus facilitating their solubilization.

3.1. Optimisation of conditions for focused microwave-assisted extraction

fMAE recoveries may be influenced by the irradiation time, the applied microwave power, and the solvent volume. These effects were investigated and the results are presented in Table 1.

3.1.1. Influence of the irradiation time

Increasing the extraction time did not enhance the extraction. On the contrary, lower extraction efficiencies were sometimes observed, with worse overall repeatability. This decrease in recoveries was already observed during the fMAE of PAHs from sediments [23]. It may be due to losses caused by either volatilization or degradation during long microwave irradiation. Experiments performed on two different Achères sludge samples gave similar observations. Consequently, a 10 min irradiation time was adopted.

3.1.2. Influence of the microwave power

At high power the overall repeatability decreased, so that results were not significantly different from those at 30 W as already observed for sediments [23]. Consequently, mild heating at 30 W was selected to obtain acceptable repeatability.

3.1.3. Influence of the solvent volume

A solvent volume of 30 ml gave the best results for the investigated PAHs. A lower solvent volume led to lower recoveries, probably due to a saturation of the solvent by the extracted compounds (especially material from the sludge). Increasing the solvent volume did not increase the extraction efficiency. Instead, it led to a more diluted extract, so that a longer time was required for concentration before the clean-up step.

Table 1
Influence of fMAE conditions on PAH extraction from Achères sludges

Parameters	PAH concentration (mg kg ⁻¹ dm) in two sludge samples (Relative standard deviation RSD(%))							
	Achères 1 sludge			Achères 2 sludge				
	Fluo	BbF	BaP	Fluo	BbF	BaP		
Time (min)	10	0.652 (4.3)	0.103 (5.8)	0.062 (11.3)	10	1.005 (11.2)	0.262 (3.4)	0.151 (8.4)
	30	0.595 (35.8)	0.101 (65.3)	0.064 (57.8)	20	0.841 (14.5)	0.241 (11.8)	0.159 (2.5)
Power (W)	30	0.652 (4.3)	0.103 (5.8)	0.062 (11.3)	30	1.005 (11.2)	0.262 (3.4)	0.151 (8.4)
	90	0.830 (27.6)	0.110 (16.4)	0.089 (42.7)	45	0.836 (19.2)	0.257 (3.5)	0.152 (6.6)
		60				1.012 (3.0)	0.277 (10.2)	0.181 (8.2)
Solvent volume (ml)					20	0.837 (12.3)	0.206 (7.8)	0.151 (7.1)
					30	1.005 (11.2)	0.262 (3.4)	0.151 (8.4)
					40	0.906 (6.2)	0.287 (9.3)	0.167 (5.6)

3.1.4. Performance of an experimental factorial design

In order to highlight the most influential parameters and possible interactions, a two-level full factorial design 2³ was constructed using the above experiments. The design matrix is given in Table 2. The estimated effects (Table 3) show that time was the most influential factor for the three PAHs, with a negative effect in each case. On the other hand, the microwave power has only a slight negative effect in

Table 3
Estimated effects and interactions

Effects	Fluo	BbF	BaP
Time (min)	-3.101	-0.937	-0.527
Power (W)	-0.0609	-0.0007	-0.0079
Volume (ml)	-0.0113	0.0032	-0.0019
Time×power	0.0141	0.0089	-0.0002
Time×volume	0.0284	-0.0025	-0.0016
Power×volume	-0.0104	-0.0023	-0.0081
Time×power×volume	-0.0199	0.0048	0.0003

Table 2
Design matrix in the factorial design 2³ and response values (PAH concentrations in mg kg⁻¹ dm)

Run	Time (min)	Power (W)	Volume (ml)	Fluo	BbF	BaP
1	10	30	30	1.005	0.262	0.151
5	10	30	40	0.906	0.287	0.167
3	10	45	30	0.836	0.257	0.152
6	10	45	40	0.775	0.254	0.135
2	20	30	30	0.841	0.241	0.159
7	20	30	40	0.935	0.237	0.169
4	20	45	30	0.808	0.252	0.159
8	20	45	40	0.781	0.258	0.136

the case of fluoranthene and benzo[*a*]pyrene. Interactions between the parameters were not significant.

The final selected extraction conditions for fMAE of sludge samples were as follows: 30 ml hexane–acetone (1:1, v/v), 10 min under 30 W.

3.2. Validation of the experimental procedure

3.2.1. Estimation of PAH recoveries

As the extraction of spiked PAHs may overestimate recoveries of native PAHs, recoveries were estimated using real samples.

3.2.1.1. Analysis of real sludge samples

Sludge samples were regularly taken in the Valenton plant in order to make composite samples over 2 weeks which were then divided into parts. The first one was analysed by our laboratory, and the second one by the Institut Pasteur de Lille, an official laboratory. The method used by this laboratory was a 6 h Soxhlet extraction using hexane–acetone (1:1, v/v), followed by purification on silica SPE cartridges. Results are presented in Table 4. One can note a good agreement of our estimations with the values reported by the official laboratory. Quantitative recovery was achieved for fluoranthene and benzo[*b*]fluoranthene, while recoveries were near 65% for benzo[*a*]pyrene.

3.2.1.2. Analysis of certified marine sediment SRM 1941a

Recoveries were further estimated by extracting the certified marine sediment SRM 1941a. Results are presented in Table 4. They show acceptable repeatability, but with systematically lower mean concentrations than the certified values, due to non-quantitative extraction of the three PAHs. The estimated recoveries were $74.8 \pm 11.02\%$, $60.5 \pm 5.3\%$ and $56.1 \pm 5.1\%$ for fluoranthene, benzo[*b*]fluoranthene and benzo[*a*]pyrene, respectively. Previous experiments using pMAE with dichloromethane or hexane–acetone (1:1, v/v) (115 °C, 10 min) also reported non-quantitative extraction of PAHs from SRM 1941a [31]. Compared to the certified values, recoveries were found to be around 48% for benzo[*a*]pyrene, 68% for benzo[*b*]fluoranthene and 72% for fluoranthene, quite close to our estimated recoveries. Lopez-Avila et al. suspected adsorption of some extracted PAHs on co-extracted elemental sulphur that may have precipitated upon concentration of the extract. However, in our experiments, copper was added to the matrix in order to avoid the presence of sulphur in the extract. Therefore, it seems that incomplete extraction was observed using fMAE on dry sediment. Remoistening the dry matrix before extraction may have been of help in extracting PAHs.

Table 4
Comparison of estimated PAH concentrations with certified or expected values

Matrix	PAHs	Concentrations (mg kg ⁻¹ dm)			
		fMAE	Institut Pasteur	Certified	Interlaboratory test
Valenton 1 sludge	Fluo	0.77±0.17	0.78	–	–
	BbF	0.27±0.10	0.28	–	–
	BaP	0.20±0.05	0.29	–	–
Valenton 2 sludge	Fluo	0.74±0.03	0.82	–	–
	BbF	0.22±0.13	0.21	–	–
	BaP	0.16±0.15	0.25	–	–
SRM 1941a	Fluo	0.733±108	–	0.981±78	–
	BbF	0.449±39	–	0.740±110	–
	BaP	0.352±32	–	0.628±52	–
Achères sludge	Fluo	2.645±0.369 ^a	–	–	[1.692–2.366]
	BbF	0.883±0.082 ^a	–	–	[0.45–0.919]
	BaP	0.988±0.118 ^a	–	–	[0.34–0.99]

^a After correction by the recoveries estimated with the SRM 1941a.

3.2.2. Interlaboratory study

This study involved 9 laboratories, each applying its own experimental procedure for the determination of PAHs. Extraction methods were fMAE, pMAE, Soxhlet extraction, sonication, PLE, and further clean-up was performed on silica, alumina or florisil. Analysis was performed by HPLC–FLD or gas chromatography coupled to mass spectrometry. The sample received by each laboratory was a dried and homogenized Achères sludge sample. The recoveries estimated above for the certified marine sediment were taken into account to determine our final estimated sludge concentrations. Results are presented in Table 4. Our estimated concentrations always fall within the confidence interval of the interlaboratory study. However, we are just at the limit in the case of fluoranthene, with clearly higher estimated concentrations. This could be attributed to an overestimation of its peak area due to the presence in the extract of several interfering compounds with retention times very close to that of fluoranthene, despite the silica clean-up step. As already observed, saponification of the extract and further liquid–liquid partitioning might have improved fluorescence profiles, possibly due to the disruption of associations between minor PAHs and lipids during the basic treatment, followed by fatty acid removal during liquid–liquid partitioning [32].

However, this would have added two more steps to the experimental procedure, which increases the overall analysis time and the risks for losses or contaminations. For that reason, this treatment was not performed.

3.3. Comparison of fMAE with other extraction techniques

In order to compare the techniques under the same conditions, we used a dried matrix (Achères sludge) and the same solvent, hexane–acetone (1:1, v/v). Moreover, the concentration and silica clean-up steps were all performed after the extraction step. In that way, differences in PAH recoveries may be directly attributed to different extraction efficiencies.

3.3.1. Traditional techniques

Firstly the extraction efficiencies using fMAE were compared with results obtained using the classical Soxhlet extraction. As shown in Table 5, slightly lower extraction efficiencies were obtained by fMAE for most of the considered PAHs, except benzo[*a*]pyrene for which a higher value was recovered. The mean recovery of fMAE as compared to Soxhlet was $78.7 \pm 33.9\%$. A previous comparison of fMAE under similar conditions with Soxhlet extraction (2×24 h) also gave slightly lower re-

Table 5
Comparison of expected values with estimated concentrations for PAHs in an Achères sludge sample, using different extraction techniques

PAHs	Concentrations (mg kg ⁻¹ dm) (RSD (%))					
	Expected	fMAE	Soxhlet 1	Soxhlet 2	Sonication	PLE
Anth	nd	0.094 (12.7)	0.126 (8.7)	0.093 (9.7)	0.066 (11.0)	0.147 (0.6)
Fluo	1.20	1.005 (11.2)	1.600 (4.3)	1.410 (6.9)	0.525 (7.9)	1.277 (3.3)
Pyr	nd	0.868 (11.0)	1.334 (1.7)	1.177 (3.9)	0.757 (4.4)	1.207 (1.6)
BbF	0.43	0.262 (3.4)	0.340 (6.7)	0.350 (12.3)	0.161 (10.1)	0.443 (3.6)
BkF	nd	0.089 (10.4)	0.140 (3.5)	0.139 (7.3)	0.055 (15.9)	0.164 (2.5)
BaP	0.31	0.151 (8.4)	0.130 (24.3)	0.150 (20.3)	0.114 (13.5)	0.234 (4.7)
Sum	nd	2.469	3.670	3.319	1.678	3.472

nd: not determined.

coveries for the recent technique (mean around 87% as compared to Soxhlet) [23]. However, due to the rapidity of fMAE (10 min) and the low solvent volume required (30 ml) as compared to Soxhlet extraction (6 h, 250 ml), the benefits for using the recent technique are obvious. In addition, a higher repeatability was achieved by fMAE (RSDs: 3.4–12.7%) as compared to Soxhlet extraction (RSDs: 1.7–24.3%).

fMAE was also compared to sonication. As shown in Table 5, sonication, under our conditions, was much less efficient than fMAE, with recoveries ranging from 52.3 to 87.3% compared to fMAE. This is in agreement with a previous study reporting lower extraction efficiencies for this technique compared to Soxhlet extraction for PAHs in contaminated soils [27]. However, it must be pointed out that we used sonication without any optimisation of the experimental conditions. It is interesting to note that the lowest efficiency was obtained for fluoranthene, suggesting that this compound would require more time to be desorbed, as already observed in the case of acenaphthylene and acenaphthene from soils [33]. Conditions should be optimized to enable efficient extractions [34–37]. Yet, despite its simplicity, sonication requires manual steps, thus increasing the time devoted to the extraction step as well as possible losses. With regards to its repeatability, in our experiments we unexpectedly found RSDs similar to those of fMAE (4.4–15.9%).

3.3.2. Pressurized liquid extraction

Pressurized liquid extraction has been successfully applied to the determination of several organic pollutants, especially PAHs, in environmental matrices [27,29,38,39]. It was frequently reported to give comparable or even higher extraction efficiencies than other techniques, which could be explained by the use of a high pressure in the system, thought to allow a better penetration of the solvent into the matrix. Higher extraction efficiencies were achieved with PLE compared to fMAE (see Table 5). The total estimated concentration for the analysed PAHs is very close to those obtained using the two Soxhlet series, indicating similar recoveries for both techniques. Also, estimated concentrations for fluoranthene, benzo[*b*]fluoranthene and benzo[*a*]pyrene were in good agreement with the expected con-

centrations (i.e., estimated by an official laboratory that also used PLE for extraction). These results tend to prove the high reproducibility of this recent technique. fMAE extracted from 54.1 to 78.7% of the PAHs, with a mean value of $65.4 \pm 21.9\%$. In the particular case of fluoranthene, benzo[*b*]fluoranthene and benzo[*a*]pyrene we obtained respectively 78.7, 59.1 and 64.6%, values that are in good agreement with the extraction efficiencies estimated for extraction of the marine sediment SRM 1941a. The PLE technique was also superior in terms of repeatability with RSDs ranging from 0.6 to 4.7%, compared to 3.4–12.7% with fMAE.

3.3.3. Summary of the performances of the techniques used

With regards to the extraction efficiency, PLE and Soxhlet extraction gave significantly higher values. In the case of PLE, this could be explained by the temperature used (100 °C) as well as the application of pressure, both of which enable a better penetration of the solvent into the matrix. For the Soxhlet extraction, higher recoveries could be attributed to the extended time used. fMAE gave mean recoveries of 65 to 80%, while sonication yielded even lower values than fMAE. Once again, it must be underlined that sonication conditions were not optimized in these experiments. Considering the time devoted to the extraction step, fMAE was the fastest technique (10 min), ahead of PLE (around 20 min as the cell must be cooled down after the extraction), sonication (45 min) and Soxhlet (6 h). Additional steps may then be performed before analysis of the extract. Concentration and clean-up steps were common to all the extracts, but an additional filtration step was required for both fMAE and sonication, thus increasing the sample treatment time by about 20 min. Moreover, the solvent volume used is of importance, as it gives an indication of the dilution of the extract, which may be a serious drawback when considering trace compounds. The lowest volume was 20 ml (PLE), to be compared with 30 ml (fMAE), 60 ml (sonication) and 250 ml (Soxhlet). Finally, the selectivity of the technique could be a major advantage, especially with difficult matrices such as sludges. In our experiments, we found similar selectivities for all the techniques used, with numerous interfering compounds still remaining despite the

clean-up step. In the case of PLE, we also tested in situ clean-up using either silica or alumina. This was convenient as it avoided performing the manual silica clean-up step. However, the chromatograms showed numerous compounds still present, with more peaks at the beginning of the chromatogram. In some cases, it may be advisable to continue to perform the SPE clean-up after the extraction.

In summary, PLE appears to be a promising technique, giving high recoveries with moderate extraction times and solvent volumes. In addition, it allows some automation, requires no filtration step after the extraction, and purification may even be performed in situ. Yet, in the case of complex matrices such as sludges, an additional clean-up step should be used. The major drawback at the moment is the relatively high investment cost of commercial systems. fMAE on the other hand involves a much lower investment cost, as the systems operate under atmospheric pressure. The main limitations of these systems are the lack of automation and the need to filter or centrifuge after the extraction, which may induce losses or contaminations and which increases the overall sample treatment time. The Soxhlet extraction should not be omitted as it allows efficient extraction, using very cheap equipment. Its main drawbacks are, of course, the large solvent volumes used and the long extraction times required. Sonication may be an interesting and cheap alternative technique. However, our results clearly show that experimental conditions should be carefully optimized in order to ensure efficient extractions.

4. Conclusion

An experimental procedure involving fMAE, followed by concentration of the extract, silica clean-up using SPE, subsequent solvent exchange, and analysis using HPLC–FLD, was developed for the determination of PAHs in sewage sludges. Parameters were tested to find the best extraction conditions. Validation was performed by extracting a certified material as well as participating in an interlaboratory study. It appeared that, although non-quantitative extractions were obtained, extraction recoveries could be adequately estimated: $74.8 \pm 11.02\%$, $60.5 \pm 5.3\%$ and $56.1 \pm 5.1\%$ for fluoranthene, ben-

zo[*b*]fluoranthene and benzo[*a*]pyrene, respectively. The limits of detection ranged from 4 to $27 \mu\text{g kg}^{-1} \text{ dm}$ for the analysed PAHs, except for fluoranthene ($105 \mu\text{g kg}^{-1} \text{ dm}$) due to the presence of interfering compounds in the chromatogram. Improvements could probably be made by addition of water to the dried sludge before the extraction. In addition, as numerous interfering compounds were still present in the extract, the clean-up step should be modified to ensure cleaner extracts. For that purpose, a saponification of the extract may be used. An alternative would be to use a more selective sorbent than silica for the clean-up step. These improvements should lead to lower limits of detection.

Comparison of fMAE with other techniques (Soxhlet, sonication and PLE), under similar conditions (dried sludge, hexane–acetone as the solvent), showed the efficiency of PLE in terms of extraction recoveries as well as repeatability. However, due to its moderate investment cost, fMAE remains an attractive alternative technique. The Soxhlet apparatus also appeared to be efficient for extracting PAHs, but required much longer extraction times and higher solvent volumes. To be really competitive, sonication should be optimized, as low recoveries were obtained under the conditions used. It is interesting to point out that, with all these techniques, similar selectivities were obtained. Indeed, when organic solvents are used, the extraction step is not very selective, whatever the technique, so that a clean-up step is required for accurate quantification of trace organic compounds.

Acknowledgements

The authors thank the SIAAP (Syndicat Interdépartemental d'Assainissement de l'Agglomération Parisienne) and the SEDE company for their contribution to this study and their financial support. The authors also thank Dionex France for the loan of the Accelerated Solvent Extractor and for their precious advice in using this system.

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