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DETERMINATION OF POLYCYCLIC AROMATIC HYDROCARBONS IN WATER BY HPLC - FLUORIMETRIC DETECTION COUPLED WITH LIQUID–LIQUID AND SOLID-PHASE EXTRACTION

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Abstract. HPLC – fluorimetric detection coupled with liquid–liquid extraction (LLE) and solid-phase extraction (SPE) is applied for determination of 12 polynuclear aromatic hydrocarbons (PAHs). Three solvents – hexane, ethyl acetate and dichloromethane were tested in LLE. Ethyl acetate was the best solvent. The analytical parameters for different PAHs are as follows: recovery: 66–97%; reproducibility (V%): ± 0.87-14.29; sensitivity: 0.001-0.01 μg/l. SPE was performed by C^{18} (1000 mg) cartridges and two elution solvents – ethyl acetate and acetonitrile. Ethyl acetate was the better elution solvent. The analytical parameters for different PAHs are as follows: recovery: 0-75%; reproducibility (V%): ± 0.18-4.39; sensitivity: 0.003-0.01 μg/l. The method was applied for determination of PAHs in water samples from field studies. phenanthrene, fluoranthene, benzo(k)fluoranthene, benzo(b)fluoranthene, benzo(a)pyrene and indeno(1,2,3-cd)pyrene were found in trace amounts.

Keywords: polycyclic aromatic hydrocarbons, HPLC – fluorimetic detection coupled with liquid–liquid extraction, water quality.

AIMS AND BACKGROUND

Polycyclic aromatic hydrocarbons (PAH) are a group of over one hundred different chemicals that are formed during the incomplete burning of coal, oil and gas, garbage, or other organic substances like tobacco or char-broiled meat[1-4]. PAHs are produced by burning jet fuel and are also found in emissions from motor vehicles. They are detected in the air, water and soil. Animal studies have shown that polycyclic aromatic hydrocarbons can cause harmful effects on the skin, body fluids, and reactance to fight disease. Many of these compounds have been found to cause cancer in animal trials[5,6]. Therefore, they are considered to be toxic to the environment.

PAHs are poorly soluble in water, but because some of them are hazardous for health, the World Health Organisation and the European Union have set limits for their concentration in drinking water. They are:

– 0.01 μg/l for benzo(a)pyrene;

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– 0.1 μg/l for the sum of benzo(b)fluaranthene, benzo(k)fluoranthene, dibenzo(g,h,i)perylene and indeno[1,2,3-cd]pyrene.

PAHs have a natural fluorescence. Therefore, fluorimetric detection coupled with liquid–liquid and solid-phase extraction is widely applied for their determination[7-17].

EXPERIMENTAL

This method for determination of polycyclic aromatic hydrocarbons is based on HPLC – fluorimetric detection coupled with liquid–liquid extraction and solid-phase extraction.

Three solvents – hexane, ethyl acetate and dichloromethane were tested in liquid–liquid extraction of 7 PAH. Table 1 shows necessary quantities of extraction solvents per 1 l of a water sample. The time for every step of extraction is 10 min. Extracts are concentrated and chromatographically analysed.

Table 1. Necessary quantities of extraction solvent for extraction of 1 l of a water sample

Solvent	Quantity
Hexan	3 × 30 ml
Dichloromethane	3 × 30 ml
Ethyl acetate	1 × 100 ml + 1 × 60 ml

Ethyl acetate was the best solvent for most compounds. Ethyl acetate has low toxicity and the highest quantity is compensated with the low cost of the solvent.

CHROMATOGRAPHIC APPARATUSES

Technique:	HPLC Hewlett Packard 1050
Column:	Prosphere 300 PAH 5u (150 mm × 4.6mm)
Mobile phase:	A. Acetonitrile
	B. Water
Gradient:	0-3 min, 50% A
	3-25 min, 50% A to 100% A
	25-30 min, 100% A
Flow rate:	0.8 ml/min
Temperature:	25°C
Detector:	Fluorescence detector :
	0 min – λ_{exc} = 260 nm, λ_{em} = 350 nm
	11.5 min – λ_{exc} = 260 nm, λ_{em} = 420 nm
	28 min – λ_{exc} = 260 nm, λ_{em} = 500 nm
Sample size:	20 μl

RESULTS

ANALYTICAL PARAMETERS

Limit of detection for different PAH are given in Table 2.

Table 2. Limit of detection (LOD)

Compound	LOD (μg/l)
Phenanthrene	0.00036
Anthracene	0.00003
Fluoranthene	0.00105
Pyrene	0.00093
Benzo(a)anthracene	0.00051
Chrysene	0.00063
Benzo(b)fluoranthene	0.00054
Benzo(k)fluoranthene	0.00045
Benzo(a)pyrene	0.0087
Dibenz(a,h)anthracene	0.00066
Benzo(ghi)perylene	0.00045
Indeno (1,2,3-cd)pyrene	0.00051

Figure 1 shows a chromatogram of standard solution of PAH with the concentration 10 μg/l.

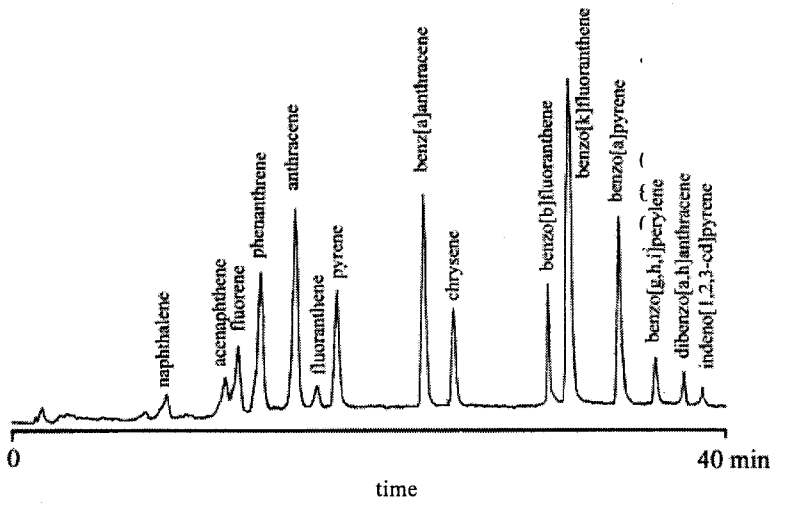

Fig. 1. Chromatogram of standard solution of PAH with the concentration 10 μg/l

The next figure shows comparison of recoveries for liquid–liquid extraction of water samples, containing 7 PAH with the concentration 0.01 μg/ l with solvents hexane, ethyl acetate and dichloromethane.

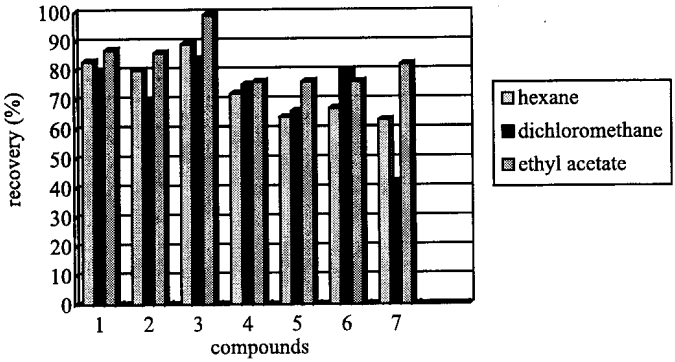

Fig. 2. Comparison of recoveries for liquid–liquid extraction of water samples, containing 7 PAH with the concentration 0.01 mg/l with solvents hexane, ethyl acetate, and dichloromethane
1 – chrysene; 2 – benzo(b)fluoranthene; 3 – benzo(k)fluoranthene; 4 – benzo(a)pyrene; 5 – dibenz(a,h)anthracene; 6 – benzo(ghi) perylene; 7 – indeno (1,2,3-cd) pyrene

Table 3 shows the analytical parameters of the method for liquid–liquid extraction of water samples, containing 7 PAH with the concentration 0.01 μg/ l with solvent ethyl acetate.

Table 3. Analytical parameters of the method for liquid–liquid extraction of water samples, containing 7 PAH with the concentration 0.01 mg/ l with solvent ethyl acetate

Compound	Sr n=6 (μg/ l)	VCr (%)
Chrysene	0.0010	14.3
Benzo(b)fluoranthene	0.0011	12.4
Benzo(k)fluoranthene	0.0009	11.8
Benzo(a)pyrene	0.0004	4.5
Dibenz(a,h)anthracene	0.0003	3.8
Benzo(ghi) perylene	0.0005	7.4
Indeno (1,2,3-cd) pyrene	0.0007	7.3

Note: n–number of samples; Sr – mean standard deviation; VÑr – variation ratio.

ANALYTICAL PARAMETERS FOR SOLID-PHASE EXTRACTION

Solid-phase extraction (SPE) was performed by C^{18} cartridges and two elution solvents – ethyl acetate and acetonitrile for 12 PAH. Acetonitrile was the better elution solvent for most of them. Figure 3 illustrates that.

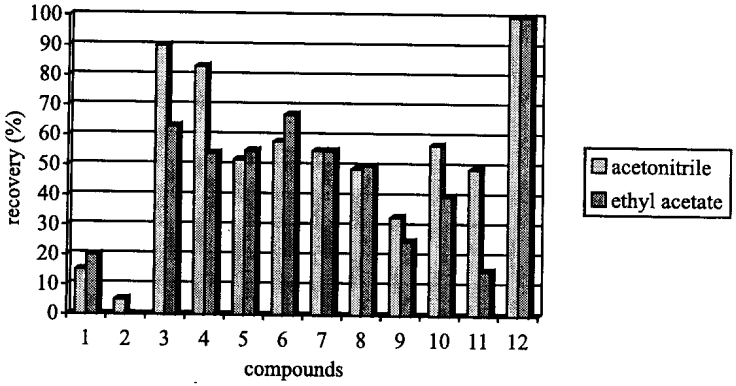

Fig. 3. Comparison of recoveries for solid-phase extraction of water samples, containing 12 PAH with the concentration 0.005 μg/ l with solvents ethyl acetate and acetonitrile
1 – phenanthrene; 2 – anthracene; 3 – fluoranthene; 4 – pyrene; 5 – benzo(a)anthracene; 6 – chrysene; 7 – benzo(b)fluoranthene; 8 – benzo(k)fluoranthene; 9 – benzo(a)pyrene; 10 – dibenz (a,h)anthracene; 11 – benzo(ghi)perylene; 12 – indeno (1,2,3-cd)pyrene

Table 4 shows the analytical parameters of the method for solid-phase extraction of water samples containing 12 PAH with the concentration 0.005 μg/ l with solvent acetonitrile.

Table 4. Analytical parameters of the method for solid- phase extraction of water samples containing 12 PAH with the concentration 0.005 μg/ l with solvent acetonitrile

Compound	Sr n=5 (μg/ l)	VCr (%)
Phenanthrene	0.00012	1.70
Anthracene	0.00001	0.18
Fluoranthene	0.00035	4.96
Pyrene	0.00031	4.39
Benzo(a)anthracene	0.00017	2.41
Chrysene	0.00021	2.97
Benzo(b)fluoranthene	0.00018	2.55
Benzo(k)fluoranthene	0.00015	2.12
Benzo(a)pyrene	0.00029	4.1
Dibenz(a,h)anthracene	0.00022	3.11
Benzo(ghi)perylene	0.00015	2.12
Indeno (1,2,3-cd)pyrene	0.00017	2.41

Note: n–number of samples; Sr–mean standard deviation; VCr–variation ratio.

The method was applied for determination of PAH in water samples from field studies. phenanthrene, fluoranthene, pyrene, benzo(k)fluoranthene, ben-

zo(b)fluoranthene, benzo(a)pyrene, dibenzo(a,h)anthracene and indeno(1,2,3-cd)pyrene were found in trace amounts.

Examples for the samples from field studies, pretreated with liquid–liquid extraction, are given in Table 5.

Table 5. Examples for samples from field studies, pretreated with liquid–liquid extraction

Drinking water (Sofia)	**Drinking water (Kostinbrod)**
Benzo(b)fluoranthene – 0.0004 μg/l	Pyrene – 0.001 μg/l
Dibenzo(a,h)anthracene – 0.0007 μg/l	Benzo(k)fluoranthene – 0.0005 μg/l
Benzo(g,h,i)perylene – 0.0009 μg/l	Benzo(a)anthracene – 0.0005 μg/l
	Dibenzo(a,h)anthracene – 0.0007 μg/l
Raw water (Sofia)	**Sewage water (Sofia)**
Phenanthrene – 0.02 μg/l	Phenanthrene – 0.09 μg/l
Anthracene – 0.0005 μg/l	Pyrene – 0.03 μg/l
Pyrene – 0.005 μg/l	Benzo(a)anthracene – 0.002 μg/l
Benzo(b)fluoranthene – 0.0004 μg/l	Chrysene – 0.006 μg/l
Benzo(a)anthracene – 0.0005 μg/l	Benzo(b)fluoranthene – 0.004 μg/l
Dibenzo(a,h)anthracene – 0.0007 μg/l	Benzo(k)fluoranthene – 0.001 μg/l
	Dibenzo(a,h)anthracene – 0.003 μg/l
Waste water (Vraza)	**Waste water (Mezdra)**
Chrysene – 0.004 μg/l	Fluoranthene – 0.02 μg/l
Benzo(b)fluoranthene – 0.004 μg/l	Pyrene – 0.01 μg/l
Benzo(k)fluoranthene – 0.002 μg/l	Benzo(a)pyrene – 0.001 μg/l
Benzo(a)pyrene – 0.009 μg/l	
Indeno(1,2,3-cd)pyrene – 0.002 μg/l	

Figure 4 shows a chromatogram of waste water sample from Mezdra.

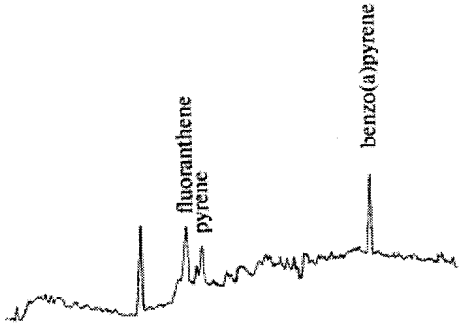

Fig. 4. Chromatogram of waste water sample from Mezdra

CONCLUSIONS

In conclusion, it should be outlined that the recovery in case of a liquid–liquid extraction for most compounds is significantly better than the recovery in case of a solid-phase extraction. Concentrations of PAH in drinking and surface water are usually low because they have low solubility in water. Therefore, liquid–liquid extraction is better for the similar kinds of water.

The solid-phase extraction is handier, fast and cheaper and can be used preferably for pretreatment of water samples with high concentrations of PAH.

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