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Comparison of four methods for the determination of polycyclic aromatic hydrocarbons in airborne particulates☆

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Abstract

High-performance liquid chromatography (HPLC) with ultraviolet and fluorescence detection and capillary gas chromatography (GC) with flame ionization and mass spectrometric (MS) detection were used to determine nineteen polycyclic aromatic hydrocarbons (PAHs) in airborne particulates. Sixteen of them are included in the prioritary pollutants list of the US Environmental Protection Agency. Five C_{18}-bonded silica HPLC columns and five GC capillary columns were checked to select the best conditions for the PAH mixtures. Samples were extracted by adding an organic solvent and sonication. The recoveries obtained were >75%. These results are compared with those obtained using Soxhlet extraction. The method was applied to the determination of PAHs at five sampling sites in the city of Valencia during 1 week. The results confirm that the best detection limits are obtained by HPLC–fluorescence, which is also the simplest, shortest and most economical method. In spite of its high maintenance cost, GC–MS in the single-ion monitoring mode is also suitable for the determination of PAHs in real samples using a low-volume system.

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs) are ubiquitous in atmospheric particulates, and with lead constitute the principal pollutants of urban areas. There are two kinds of systems to collect the particulates, high- and low-volume collection bubbler systems. Low-volume systems are more frequently used but the low volume of sample (*ca.* 2 m^3 during a sampling period of 24 h) requires a technique with very high sensitivity and makes the determination of PAHs in atmospheric particulate matter an important analytical problem [1,2].

The analysis of the purified extracts can be carried out using gas and liquid chromatographic methods with different detection devices to achieve simultaneously high resolution, sensitivity and selectivity [3,4].

High-performance liquid chromatography (HPLC) has been the selected technique for PAH determination. Ultraviolet (UV) absorption and fluorescence spectrometry provide sensitive and selective detection for PAHs in HPLC [5–8]. However, not all C_{18} stationary phases provide the same selectivity for PAHs owing to the influence of factors such as the bonded-phase type, silica substrate characteris-

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tics, alkyl chain length and C_{18} ligand density or selectivity [9,10].

Gas chromatography (GC) with capillary columns is also a method of choice for the separation and analysis of complex PAH mixtures with moderate to low molecular masses. Improvements in GC stationary phases have contributed to this [11,12], and a recent comparison of four high-temperature GC columns showed their usefulness in separating PAHs with a molecular mass of 328 [13]. GC with a capillary column has been used in combination with flame ionization detection (FID) [14,15]. However, the use of a quadrupole mass spectrometer operated in single-ion monitoring (SIM) mode is an innovation that allows selective and sensitive detection [16–19].

The analysis step is usually preceded by extraction of the PAHs from the air particulates retained in PTFE or glass-fibre filters. Soxhlet extraction and ultrasonication employing a variety of organic solvents including acetone, benzene, toluene and acetonitrile are the most commonly used processes [20–22].

The main purpose of this study was to develop a method for the routine determination of PAHs in air samples taken with low-volume collection systems. For this purpose, different analytical techniques, HPLC–UV, HPLC–fluorescence, GC–FID and GC–MS-SIM, were compared and different stationary phases for both the HPLC and GC systems were checked. Finally, to demonstrate the applicability of the method to real samples and establish whether the techniques are really suitable for monitoring of PAH contamination in urban areas, PAHs were determined in atmospheric particulate matter sampled at five sites in the city of Valencia.

2. Experimental

2.1. Materials

Organic solvents (acetonitrile, methanol, acetone, cyclohexane, chloroform, dichloromethane, methanol and toluene) were of HPLC grade (Romil, Leics., UK). Ultra-pure water was prepared by ultrafiltration of distilled water with a Milli-Q system (Millipore, Bedford, MA, USA).

Acenaphthene, acenaphthylene, anthracene, benz[*a*]anthracene, benzo[*b*]fluoranthene, 2,3-benzofluorene, benzo[*ghi*]perylene, benzo[*e*]-pyrene, chrysene, dibenzo[*a,h*]anthracene, phenanthrene, fluorene, naphthalene, perylene and pyrene were supplied by Aldrich Chemie (Steinheim, Germany), benzo[*a*]pyrene by Janssen Chimica (Geel, Belgium) and fluoranthene by Scharlau (Barcelona, Spain). Benzo[*k*]-fluoranthene and indeno[1,2,3-*cd*]pyrene were purchased from the Community Bureau of Reference (BCR). (Brussels, Belgium).

These standards were dissolved in acetonitrile at 500 μg/l, although anthracene and benzo[*a*]pyrene solutions contained toluene (20%) and dibenzo[*a,h*]anthracene solution chloroform (20%). Stock mixtures of PAH standards were made up from the individual solutions in acetonitrile.

2.2. Chromatographic determinations

HPLC analyses were carried out with a Shimadzu (Kyoto, Japan) SCL-6A controller equipped with two LC6A pumps, a Rheodyne Model 7125 injector (20-μl loop), an SPD 6A ultraviolet detector, a RF-53 fluorescence detector, a CTO-6AS camera for column thermostating and a C-R4A data processor. The following LC columns were used: two Supelcosil LC-PAH (Supelco, Bellefonte, PA, USA), two Spherisorb ODS-2 (Teknokroma, Middelburg, Netherlands), a Spherisorb C_8 and a Zorbax-CN (Shandon, Runcorn, UK) and a Green Hypersil-PAH (Delta Scientific, Madrid, Spain). The characteristics of the columns and the HPLC conditions are given in Table 1. The use of different mobile phase gradients and different wavelengths in UV or fluorescence detection were checked. The conditions reported in Table 1 were suitable for routine analysis.

GC–FID analyses were performed using a Konik (Sant Cugat del Valles, Spain) Model 3000 gas chromatograph equipped with a flame ionization detector, split–splitless injector and a Spec-

Table 1
Experimental HPLC conditions

Parameter	Column						
	S-PAH (1)	S-PAH (2)	ODS-2 (1)	ODS-2 (2)	C_8	CN	H-PAH
Column length (cm)	25	15	25	25	25	25	10
Column I.D. (mm)	4.6	4.6	4.0	4.0	4.0	3.6	4.6
Particle size (μm)	5	5	5	3	5	5	5
Column temperature (°C)	30	30	30	30	30	30	30
Mobile phase gradient	See below[a]						
UV detection:							
λ (nm)	250	250	250	250	250	250	250
Fluorescence detection:							
λ_{exc} (nm)	290	290	290	290	290	290	290
λ_{em} (nm)	385	385	385	385	385	285	385

[a] Mobile phase gradient in all instances: acetonitrile–water gradient changed from 50:50 through 60:40, 70:30, 80:20 and 90:10 to 100:0, with 5 min at each composition and a 5-min gradient between each.

tra-Physics (San Jose, CA, USA) integrator with double channels and a memory module. The following GC capillary columns were tested: two BP-5 (Scientific Glass Engineering, Sydney, Australia), a BP-10 (Scientific Glass Engineering), a BP-20 (Scientific Glass Engineering) and an RSL-400 (Alltech, Deerfield, IL, USA).

Analyses were also carried out with the same columns on an HP 5970 quadrupole mass spectrometer (Hewlett-Packard, Waldbronn, Germany) connected to an HP 5690A gas chromatograph equipped with a split–splitless injector. The chromatographic data were recorded on an HP 59970 MS Chem-Station.

The characteristics of the columns and the GC conditions are given in Table 2. Various initial temperatures, programming rates and upper isotherm lengths were also tested and the conditions reported in Table 2 were suitable for routine analyses.

2.3. Extraction procedures

The PAHs were extracted ultrasonically with 5 ml of acetonitrile from Whatman filter-papers in a borosilicate glass-stoppered tube. The extracts were filtered through Millipore FHLP 01300 filter-paper. The volume of the extract was reduced to about 0.3 ml by bubbling a gentle stream of nitrogen through the solution at room temperature. The extracts were transferred into a 500-μl volumetric flask and taken up with acetonitrile for direct analysis.

Soxhlet extraction of the PAHs was performed using 250 ml of dichloromethane for 12 h at 60°C. The solvent was then evaporated in an evaporator–concentrator at 40–50°C and the residue was dissolved in 0.5 ml of acetonitrile.

2.4. Sampling

Air samples were collected at five different locations in Valencia in a low-volume collection bubbler system (MCV, Barcelona, Spain), with an aspiration pump equipped with an electric motor and a 2–4 m^3 per 24 h aspiration capacity. Dust was collected in a Whatman filter No. 1 dry aspirater air counter (1.5–3 l/min ± 3%) to provide readings per litre volume of air. The different components were interconnected by glass tubes and plastic parts.

Total suspended particulate matter samples were collected daily in Valencia during 1 week in the spring of 1992. Each sample was collected over a period of 24 h and the air volume of each sample was about 2 m^3.

Table 2
Experimental GC conditions

Parameter	Column				
	BP-5 (1)	BP-5 (2)	BP-10	BP-20	RSL-400
Injector temperature (°C)	285	285	285	285	285
Injector splitless	Yes	Yes	Yes	Yes	Yes
Splitless time (min)	0.7	0.7	0.7	0.7	0.7
Injection volume (μl)	3	3	3	3	3
Column length (m)	25	50	50	25	25
Column I.D. (mm)	0.22	0.22	0.22	0.22	0.22
Film thickness (μm)	25	25	25	25	25
Oven temperature programme	See below[a]				
FID:					
Detector temperature (°C)	300	300	300	300	300
Hydrogen carrier gas flow-rate (ml/min)	1	1	1	1	1
Nitrogen make-up gas flow-rate (ml/min)	35	35	35	35	35
Air (ml/min)	250	250	250	250	250
Hydrogen (ml/min)	35	35	35	35	35
MS:					
Transfer line temperature (°C)	260	260	260	260	260
Source temperature (°C)	200	200	200	200	200
Electron energy (eV)	70	70	70	70	70
Helium carrier gas flow-rate (ml/min)	1	1	1	1	1

[a] Temperature programme in all instances: initial temperature 50°C, held for 0.8 min, then increased at 30°C/min to 100°C (held for 2 min) and at 5°C/min to 280°C (held for 10 min).

3. Results and discussion

3.1. HPLC determination

The selectivity of HPLC for the PAH mixtures is affected by the phase type, particle diameter, column length, mobile phase flow-rate and LC column temperature [13]. In this work, these factors have been studied in detail.

Table 3 shows the resolution values (R_s) for sixteen PAHs obtained with the four stationary phases studied [$R_s = 2\Delta t/(w_{b1} + w_{b2})$], where Δt is the distance between the maxima of the two peaks and w_b the width of the peak at half-height. Fluorimetric detection was used. The concentrations of the standard solutions used were the same as those used for the optimization of extraction procedure with each detector (see below).

The pairs of PAHs acenaphthene–fluorene, benz[a]anthracene–chrysene and benzo[*e*]pyrene–benzo[*b*]fluoranthene were not separated with normal commercial columns of octyl-, octadecyl- and cyanopropyl-bonded silica. This is due to the "monomeric" character of the stationary phase [13]. The retention time of benzo[*b*]fluoranthene is the same as that of its isomer benzo[*k*]fluoranthene using the most polar columns, octyl- and cyanopropylsilica.

Only by using special columns for PAH analyses can the separation of the sixteen PAHs be achieved. The special phases consist of "polymeric" C_{18}-bonded silica and improve the selectivity for the PAHs. With these special columns, values of $R_s > 1.5$ are obtained for the pairs of PAHs. The elution order of the sixteen PAHs studied did not vary with the different LC-columns tested.

The effect of the particle diameter on the resolution of the PAH mixture was studied using the ODS-2 columns. Fig. 1A shows the effect of the particle diameter on the resolution of the

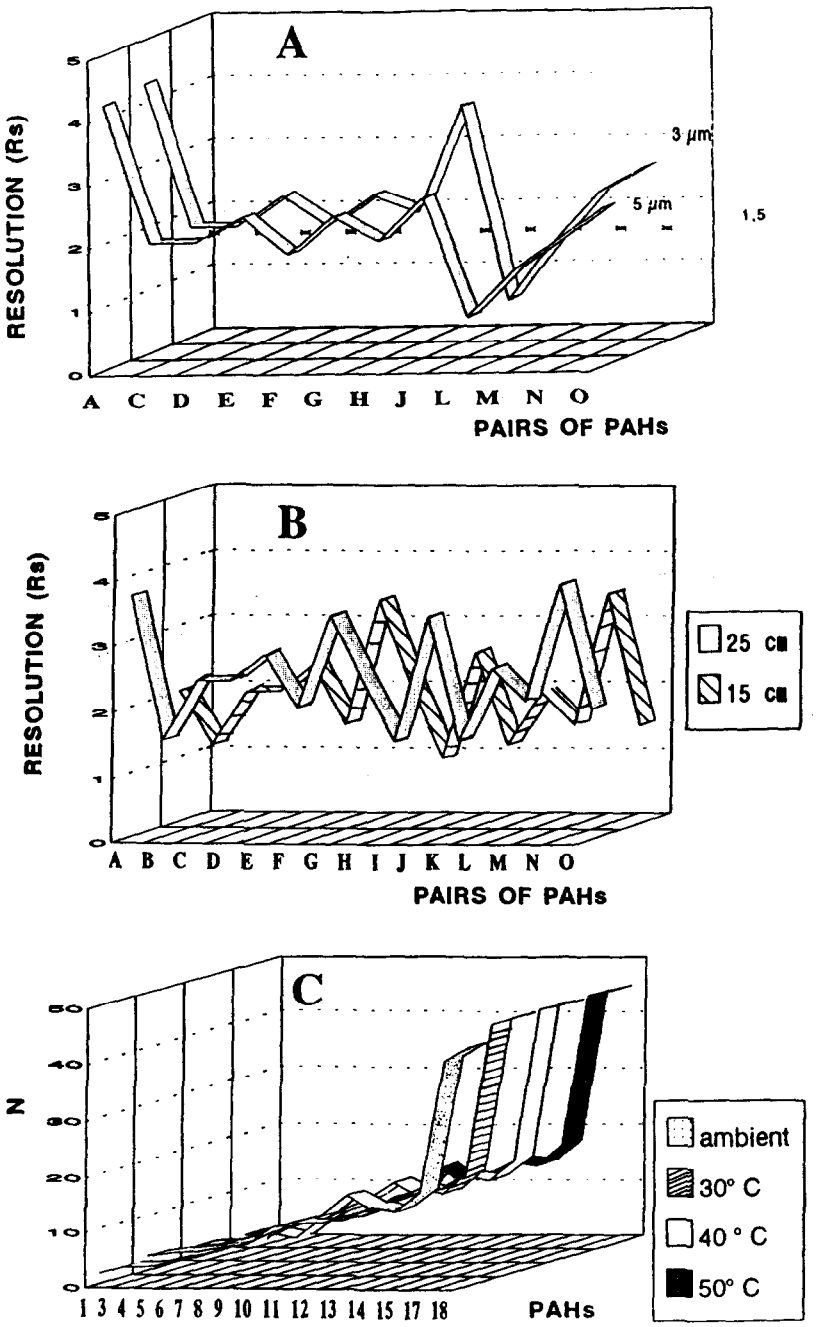

Fig. 1. Effect of instrumental parameters on the selectivity of PAHs. (**A**) Particle diameter vs. resolution; (**B**) column length vs. resolution. Pairs of PAHs: A = naphthalene–acenaphthene; B = acenaphthene–fluorene; C = fluorene–phenanthrene; D = phenanthrene–anthracene; E = anthracene–fluoranthene; F = fluoranthene–pyrene; G = pyrene–2,3-benzofluorene; H = 2,3-benzofluorene–benz[*a*]anthracene; I = benz[*a*]anthracene–chrysene; J = chrysene–benzo[*e*]pyrene; K = benzo[*e*]pyrene–benzo[*b*]fluoranthene; L = benzo[*b*]fluoranthene–benzo[*k*]fluoranthene; M = benzo[*k*]fluoranthene–benzo[*a*]pyrene; N = benzo[*a*]pyrene–dibenzo[*a*,*h*]anthracene; O = dibenzo[*a*,*h*]anthracene–benzo[*ghi*]perylene. (**C**) Column temperature vs. efficiency. For identification of PAHs, see Fig. 2.

Table 3
Resolution (R_s) obtained with the stationary phases tested

Pairs of PAHs	Stationary phase				
	S-PAH	H-PAH	C_8	C_{18}	CN
Naphthalene–acenaphthene	3.7	3.8	3.5	4.1	3.5
Acenaphthene–fluorene	1.5	1.3	0.0	0.0	0.0
Fluorene–phenanthrene	2.4	2.3	1.6	1.6	1.2
Phenanthrene–anthracene	2.4	2.1	0.8	0.8	0.0
Anthracene–fluoranthene	2.8	2.3	2.1	2.1	1.9
Fluoranthene-pyrene	2.0	1.5	0.2	0.2	0.3
Pyrene–2,3-benzofluorene	3.4	3.5	1.5	1.5	1.6
2,3-Benzofluorene–benz[*a*]anthracene	2.4	2.3	1.4	1.4	0.5
Benz[*a*]anthracene–chrysene	1.5	1.5	0.0	0.0	0.0
Chrysene–benzo[*e*]pyrene	3.4	3.5	2.7	2.7	2.8
Benzo[*e*]pyrene–benzo[*b*]fluoranthene	1.5	1.3	0.0	0.0	0.0
Benzo[*b*]fluoranthene–benzo[*k*]fluoranthene	2.6	2.4	0.0	0.0	0.0
Benzo[*k*]fluoranthene–benzo[*e*]pyrene	2.1	2.3	0.7	0.7	1.1
Benzo[*e*]pyrene–dibenzo[*a,h*]anthracene	3.9	2.3	2.4	2.4	0.0
Dibenzo[*a,h*]antracene–benzo[*ghi*]perylene	2.0	1.5	0.9	0.9	0.0

PAHs; acenaphthene–fluorene, benz[*a*]anthracene–chrysene and benzo[*e*]pyrene–benzo[*b*]-fluoranthene were not separated by the ODS-2 columns.

With the same stationary phase and column length, the resolution was improved in some instances (see Fig. 1A). However, the capacity factor was constant. This is due to a change in the peak form; the retention time is the same in both columns but the width of the base of the peak is narrower in the columns with a 3-μm particle diameter. Diminishing the particle size of the stationary phase does not improve the separation of the unresolved PAHs, but the accuracy and reproducibility are better.

In Fig. 1B the effect of the column length is illustrated using 15- and 25-cm Supelcosil-PAH columns. Although the increase in the column length leads to longer analysis times, it provides better resolution. The pairs of PAHs benz-[*a*]anthracene–chrysene and benzo[*e*]pyrene–benzo[*b*]fluoranthene were well separated in the 25-cm column whereas in the 15-cm column the separation was only partial.

The effect of the mobile phase flow-rate on the separation of the PAHs was also studied. Flow-rates of 0.8, 1.0, 1.2 and 1.4 ml/min were tested with the 25-cm Supelcosil-PAH column. A flow-rate between 0.8 and 1.2 ml/min provides a total resolution of the analyte compounds. With a flow-rate of 1.4 ml/min, the peaks corresponding

Table 4
HPLC detection limits (ng) with the fluorescence and UV detection

PAH	Fluorescence	UV
Naphthalene	2.00	16.60
Acenaphthylene	–	22.80
Acenaphthene	0.05	22.80
Fluorene	0.50	3.00
Phenanthrene	0.07	1.60
Anthracene	0.10	1.60
Fluoranthene	2.00	5.00
Pyrene	0.02	6.10
2,3-Benzofluorene	0.02	1.60
Benz[*a*]anthracene	0.01	6.10
Chrysene	0.02	2.20
Benzo[*e*]pyrene	0.03	4.00
Benzo[*b*]fluoranthene	0.05	3.00
Benzo[*k*]fluoranthene	0.05	3.20
Benzo[*a*]pyrene	0.03	6.10
Perylene	–	6.10
Dibenzo[*a,h*]anthracene	0.03	10.00
Benzo[*ghi*]perylene	0.01	8.00
Indenopyrene	–	3.00

Table 5
GC detection limits (DL, ng) with FID and MS-SIM detection and selected ions for SIM

PAH	FID DL	MS-SIM	
		Selected ion (m/z)	DL
Naphthalene	3.00	128	0.01
Acenaphthylene	1.00	152	0.02
Acenaphthene	0.90	154	0.02
Fluorene	0.90	166	0.01
Phenanthrene	0.80	178	0.01
Anthracene	0.90	202	0.01
Fluoranthene	0.80	202	0.01
Pyrene	0.80	216	0.01
2,3-Benzofluorene	0.70	228	0.02
Benz[*a*]anthracene	0.70	228	0.03
Chrysene	0.70	252	0.03
Benzo[*e*]pyrene	0.60	252	0.03
Benzo[*b*]fluoranthene	0.70	252	0.02
Benzo[*k*]fluoranthene	0.70	252	0.04
Benzo[*e*]pyrene	0.70	252	0.04
Perylene	0.70	252	0.04
Dibenzo[*a*,*h*]anthracene	0.50	276	0.04
Benzo[*ghi*]perylene	0.80	278	0.04
Indenopyrene	1.30	276	0.10

to acenaphthene and fluorene and to benz-[*a*]anthracene and chrysene were not separated. Small variations in the flow-rate usually affect the retention times but not the resolution.

The column temperature is an important parameter that can be used to modify selectivity for PAH separation. The Supelcosil-PAH column was thermostated at temperatures of 30, 40 and 50°C. Fig. 1C shows the variation in the column efficacy $[N = 5.545(t_r/W_h)]$ with temperature. The column temperature affected the efficiency of separation. The best results were obtained at 30°C. The temperature must be controlled in order to obtain reproducible retention times.

When these parameters were optimized, two detection systems (fluorescence and UV) were compared. Both detectors provided a linear response for a wide range of amounts injected, and their reproducibilities were similar and lower than 4.2%.

In Table 4, the detection limits with fluorescence and UV detection are compared. For

Table 6
Concentration of the working solution (mg/l) for fluorescence and UV detection

PAHs	Fluorescence	UV
Naphthalene	4.00	80
Acenaphthylene	–	160
Acenaphthene	0.20	80
Fluorene	2.00	16
Phenanthrene	0.10	8
Anthracene	0.40	8
Fluoranthene	4.00	16
Pyrene	0.15	8
2,3-Benzofluorene	0.05	8
Benz[*a*]anthracene	0.01	16
Chrysene	0.07	8
Benzo[*e*]pyrene	0.04	8
Benzo[*b*]fluoranthene	0.20	16
Benzo[*k*]fluoranthene	0.20	8
Benzo[*a*]pyrene	0.04	8
Perylene	–	16
Dibenzo[*a*,*h*]anthracene	0.02	16
Benzo[*ghi*]perylene	0.10	16
Indenopyrene	–	8

quantification in airborne particulates, fluorescence detection offers far greater sensitivity and, more important, is much more selective than UV detection. Only fluorene and fluoranthene present similar sensitivities with both detection techniques. However, there are three PAHs, acenaphthylene, indoperylene and perylene, that are not fluorescent at the excitation and emission wavelengths used, and they can be detected by UV spectrophotometry at 254 nm. Even the excitation and emission wavelengths can be changed in order to obtain optimum sensitivity and/or selectivity for these individual PAHs.

3.2. GC determination

A comparison of four GC columns with different stationary phases illustrates the usefulness of some of them for the high-resolution separation of PAHs.

The BP-20 and the RSL-400 columns gave low resolution for the pairs phenanthrene–anthracene and benz[*a*]anthracene–chrysene. Moreover, with the BP-20 column the peaks corresponding to the isomers perylene–dibenzo[*a*,*h*]anthracene and benzo[*b*]fluoranthene–benzo[*k*]fluoranthene were unresolved. The probable cause is the polarity of the stationary phase; these phases have polar characteristics and do not retain apolar compounds such as PAHs well.

With the BP-5 and BP-10 columns it was possible to resolve all the PAHs studied. A shorter analysis time was obtained with the BP-5 columns.

The influence of the column length on the

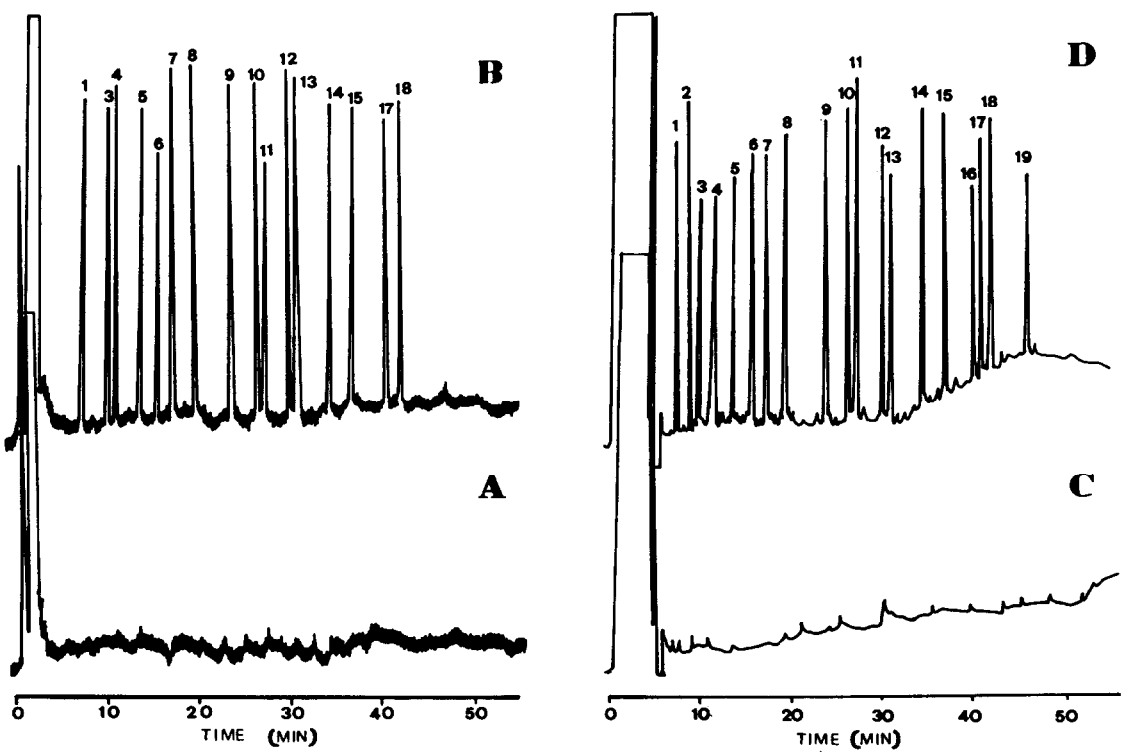

Fig. 2. Chromatograms for the HPLC analysis of 20 μl of an extract obtained by sonication of (A) untreated filter with fluorescence detection, (B) filter treated with sixteen PAHs with fluorescence detection, (C) untreated filter with UV detection and (D) filter treated with nineteen PAHs with UV detection. For concentrations, see Table 6. Peaks: 1 = naphthalene; 2 = acenaphthylene; 3 = acenaphthene; 4 = fluorene; 5 = phenanthrene; 6 = anthracene; 7 = fluoranthene; 8 = pyrene; 9 = 2,3-benzofluorene; 10 = benz[*a*]anthracene; 11 = chrysene; 12 = benzo[*e*]pyrene; 13 = benzo[*b*]fluoranthene; 14 = benzo[*k*]fluoranthene; 15 = benzo[*a*]pyrene; 16 = perylene; 17 = dibenzo[*a*,*h*]anthracene; 18 = benzo[*ghi*]perylene; 19 = indenopyrene.

separation of the PAH peaks was also studied. Increasing the length of the column (from 25 to 50 m) increased the analysis time but did not improve the resolution of the chromatographic peaks.

As a result of this study, the BP-5 column with a length of 25 m was selected for routine analyses because it provided the best separation between the PAHs with the shortest analysis time.

All these experiments were performed using FID; minimum detectable amounts ranging from 0.5 to 1.5 ng (see Table 5) were obtained. FID presented the same problem as UV detection: the sensitivity and selectivity were too low for application to real sample analyses.

To improve the sensitivity, detection with MS-SIM was checked. Table 5 shows the detection limits obtained using FID and MS-SIM for the nineteen PAHs studied. Table 5 also shows the m/z values for the selected ions in MS-SIM.

With GC–MS-SIM, the detection limits were suitable for the determination of PAHs in real samples when they are taken with low-volume systems.

3.3. Extraction procedure

To optimize the extraction procedure using sonication, the following parameters were studied: the organic solvent used in the extraction, evaporation conditions, shaking time and number of extractions.

Recovery experiments were carried out by placing on the Whatman filter 200 μl of the solution listed for each HPLC detector in Table 6. These should be equivalent at air concentrations between 0.4 and 0.002 $\mu g/m^3$ depending on the PAH for fluorescence detection and between 16 and 0.8 $\mu g/m^3$ for UV detection. For GC–FID and GC–MS-SIM, working solutions of 10 and 0.4 mg/l of each compound,

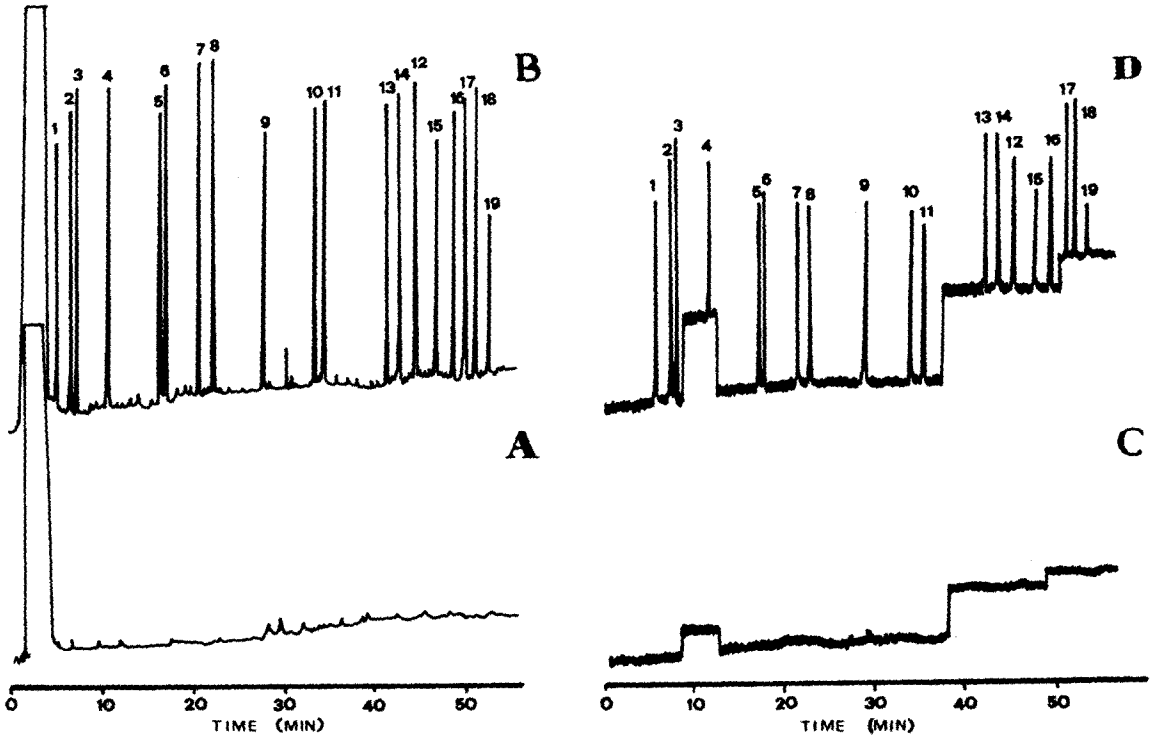

Fig. 3. Chromatograms obtained in the GC analysis of 1 μl of an exact of (A) untreated filter with FID, (B) filter treated with nineteen PAHs with FID, (C) untreated filter with MS-SIM and (D) filter treated with nineteen PAHs with MS-SIM. For concentrations, see text. Peak assignment as in Fig. 2.

respectively, were used. After the organic solvent had been evaporated, the filter was extracted.

First, four organic solvents were tested: dichloromethane, methanol, acetone, cyclohexane and acetonitrile. Methanol and cyclodhexane provided recoveries lower than 59%. Dichloromethane and acetone increased the recoveries to 66% and acetonitrile gave and best results with recoveries of about the 75%.

If the reproducibilities are compared the best results are also obtained with acetonitrile. Therefore, acetonitrile was chosen as the extraction solvent.

The evaporation conditions were checked. The recoveries diminished when the sample was dried. This phenomenon was more evident with the volatile PAHs; for example, naphthalene was not recovered when the organic solvent was evaporated. Evaporation with air and nitrogen streams was also tested; there were no significant differences in the recoveries.

Table 7
Mean recovery ±R.S.D. (%) ($n = 6$) from fortified filters using the sonication and Soxhlet extraction methods

PAH	Sonication	Soxhlet
Naphthalene	65 ± 14	30 ± 15
Acenaphthylene	76 ± 14	30 ± 16
Acenaphthene	80 ± 13	50 ± 10
Fluorene	79 ± 7	40 ± 8
Phenanthrene	99 ± 6	60 ± 9
Anthracene	76 ± 10	60 ± 10
Fluoranthene	84 ± 7	55 ± 12
Pyrene	85 ± 6	55 ± 9
2,3-Benzofluorene	79 ± 9	50 ± 7
Benz[*a*]anthracene	78 ± 5	55 ± 10
Chrysene	75 ± 4	55 ± 12
Benzo[*e*]pyrene	75 ± 4	57 ± 14
Benzo[*b*]fluoranthene	82 ± 4	58 ± 18
Benzo[*k*]fluoranthene	85 ± 4	59 ± 13
Benzo[*a*]pyrene	81 ± 6	50 ± 12
Perylene	82 ± 8	58 ± 11
Dibenzo[*a,h*]anthracene	83 ± 9	59 ± 9
Benzo[*ghi*]perylene	80 ± 6	40 ± 8
Indenopyrene	85 ± 9	48 ± 10

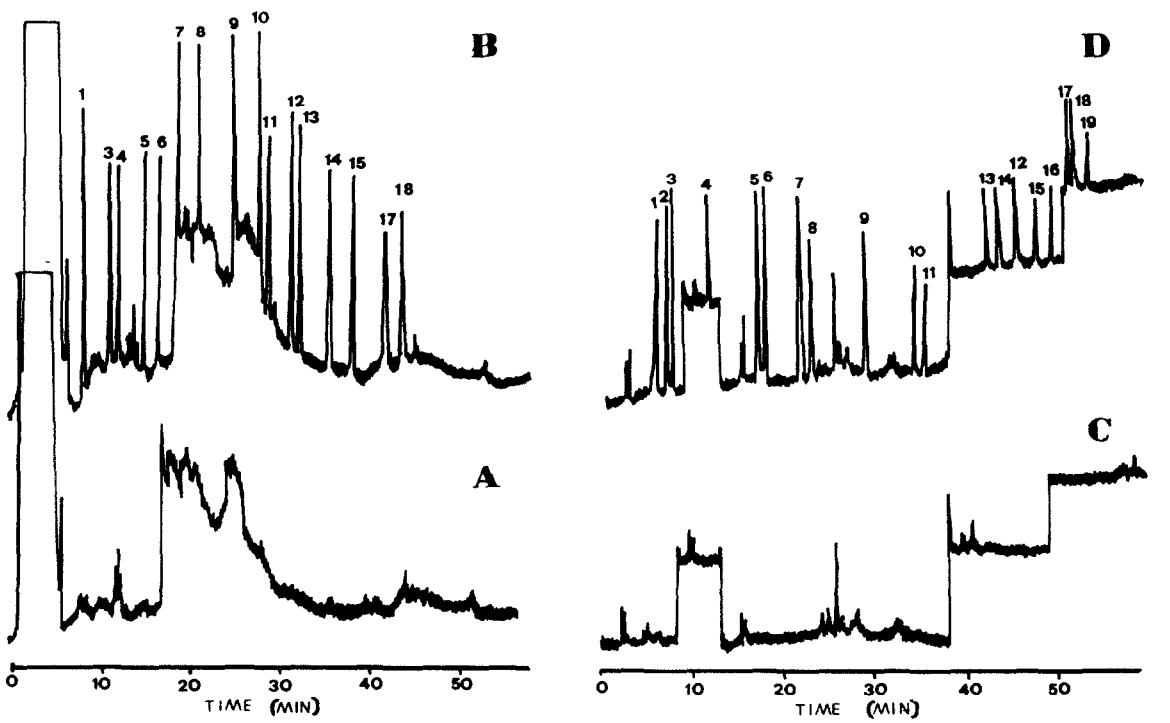

Fig. 4. Chromatograms obtained following Soxhlet extraction of (A) untreated filter by HPLC–fluorescence, (B) treated filter by HPLC–fluorescence, (C) untreated filter by GC–MS-SIM and (D) treated filter by GC–MS-SIM. For concentrations see Table 6 and text. Peak assignment as in Fig. 2.

The next step was to select the ultrasonication time. For this, Whatman No. 1 filter-paper treated with the nineteen PAHs was shaken for 5, 10, 20, 30 or 40 min with acetonitrile. The recoveries increased with shaking time up to 30 min, but shaking for more than 30 min gave lower recovery values.

The number of extractions was also studied. The filter-paper was extracted from one to five times. There was no improvement in the recoveries when the number of extractions was increased. However, the relative standard deviation increased with increase in the number of extractions.

Therefore, for maximum recoveries of PAHs, Whatman filter-papers treated with 5 ml of ethyl acetate in a borosilicate glass-stoppered tube were used. The tubes were placed in an ultrasonic bath for 30 min. The extracts were then filtered through Millipore FHLP 01300 filter-paper. The volume of the extract was reduced to about 0.3 ml by bubbling a gentle stream of nitrogen through the solution at room temperature. The extracts were transferred into a 500-μl volumetric flask and taken up with acetonitrile for direct determination.

Fig. 2A–D illustrate the chromatograms for unspiked filter extract and spiked filter extract analysed by HPLC–fluorescence (Fig. 2A and B) and by HPLC–UV (Fig. 2C and D). In both instances there were no interfering peaks in the blank chromatograms but a difference in sensitivity between the two detectors can be observed (see the concentrations in Table 6).

Fig. 3A–D show the same chromatograms but obtained using GC–FID and GC–MS-SIM. The FID blank (Fig. 3A) showed some interfering peaks, but the MS-SIM blank (Fig. 3C) showed none. Differences in sensitivity between the two detectors can also be observed.

To validate the method, the recoveries obtained were compared with those obtained using

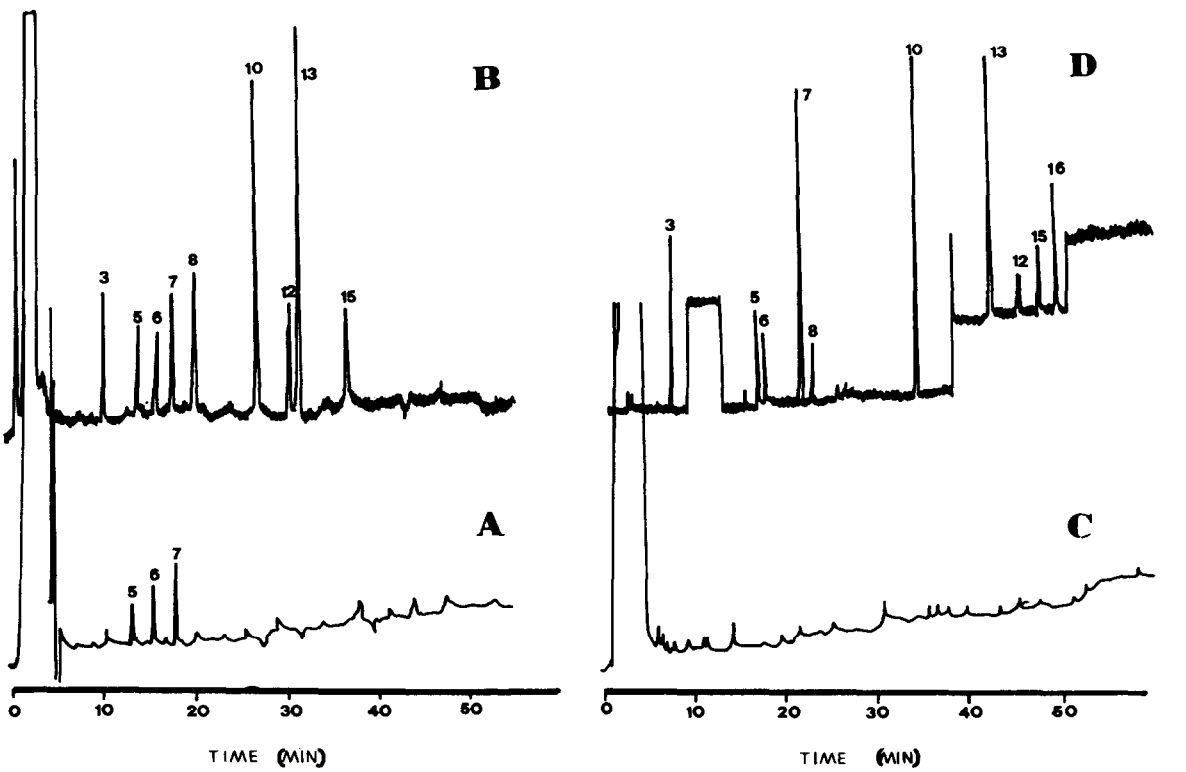

Fig. 5. Chromatogram obtained after sonication extraction of a real samples, (A) by HPLC–UV, (B) by HPLC–fluorescence, (C) by GC–FID and (D) by GC–MS-SIM. Peak assignment as in Fig. 2.

Table 8
Mean weekly values of PAHs (ng/m^3) in air samples in Valencia obtained by HPLC–fluorescence detection (F) and GC–MS (MS)

PAH	Sampling site									
	1		2		3		4		5	
	F	MS	F	MS	F	MS	F	MS	F	MS
Naphthalene	–	–	–	–	–	–	–	–	–	–
Acenaphthylene	–	–	–	–	–	–	–	–	–	–
Acenaphthene	5.7	5.9	7.6	7.5	–	–	9.0	9.0	8.8	8.9
Fluorene	–	–	–	–	–	–	–	–	–	–
Phenanthrene	2.0	1.9	3.8	3.8	10.7	11.0	8.9	9.0	–	–
Anthracene	–	–	–	–	20.3	20.0	–	–	–	–
Fluoranthene	87.8	87.8	70.8	71.2	189.8	191.9	64.2	64.4	–	–
Pyrene	3.3	3.4	1.2	1.3	–	–	–	–	–	–
2,3-Benzofluorene	–	–	–	–	–	–	–	–	–	–
Benz[*a*]anthracene	0.5	0.5	0.7	0.2	1.3	1.4	0.8	0.8	0.4	0.4
Chrysene	–	–	–	–	2.3	2.3	–	–	–	–
Benzo[*e*]pyrene	2.1	2.0	1.2	1.1	3.8	3.7	1.9	1.8	0.8	0.8
Benzo[*b*]fluoranthene	–	–	–	–	–	–	–	–	–	–
Benzo[*k*]fluoranthene	–	–	–	–	–	–	–	–	–	–
Benzo[*a*]pyrene	1.7	1.7	1.5	1.6	6.5	6.5	–	–	–	–
Perylene	–	1.1	–	–	–	–	–	–	–	–
Dibenzo[*a*,*h*]anthracene	1.2	1.1	–	–	–	–	1.3	1.4	–	–
Benzo[*ghi*]perylene	4.4	4.5	4.8	4.9	13.3	13.4	5.7	5.8	2.8	2.8
Indenopyrene	–	–	–	–	–	–	–	–	–	–

Soxhlet extraction. Table 7 shows the recoveries and relative standard deviations.

Continuous Soxhlet extraction used more organic solvent and the extraction time was longer. The recoveries obtained were lower than those obtained by ultrasonic extraction. The use of Soxhlet extraction provides blanks with many interfering peaks, some of which show the same retention times as the PAHs studied. Fig. 4A–D illustrate the chromatograms obtained by HPLC–fluorescence and GC–MS-SIM, which are the most selective detection methods studied. With the other methods the number of interfering peaks and the baseline noise increased because of the low selectivity. These chromatograms demonstrate the need for a clean-up procedure before PAH determination when Sohxlet extraction is used.

To verify the ultrasonication procedure, airborne particulate samples were taken over a period of 1 week at five locations in Valencia. The PAH concentrations were determined using HPLC–fluorescence and GC–MS-SIM. Table 8 shows the concentrations of the nineteen PAHs obtained using the two techniques. The differences were <3%. Although HPLC–fluorescence with fixed excitation and emission wavelengths does not permit the detection of acenaphthylene, indenopyrene and perylene, it should be the detection method of choice, because there were no peaks interfering with those of the PAH compounds present in the chromatogram obtained from environmental samples and the maintenance cost of the system is much lower than that of GC–MS-SIM for routine analysis.

Fig. 5 shows the chromatograms for a real sample obtained using the four systems. With HPLC–UV and GC–FID no peaks were detected.

The levels of PAHs found in the air samples taken at different locations showed a relationship between this concentration and the traffic intensity at the different locations.

4. Conclusions

Of the four techniques studied, HPLC with fluorescence detection is to be preferred for the measurement of PAHs in airborne particulate samples taken with low-volume systems. The results obtained using HPLC–fluorescence are comparable to those obtained with GC–MS-SIM, but the former has the advantage of being economical for use in routine analyses, which is not a characteristic of MS methods.

Using ultrasonic extraction with acetonitrile, the recoveries obtained are better than those found with Soxhlet extraction, the number of interfering peaks is smaller and the simplicity, speed and cost are much improved.

The results show that the level of PAH pollution in Valencia is not alarming, but a clear relationship between traffic intensity and the level of PAHs was found.

Future research will be devoted to the use of HPLC–wavelength-programmed fluorescence detection with the excitation and emission wavelengths changing during the chromatography to achieve optimum sensitivity and/or selectivity for the individuals PAHs. It will also focus on monitoring of PAH levels at different sites in Valencia periodically during the year to establish the pattern of pollution in the city.

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