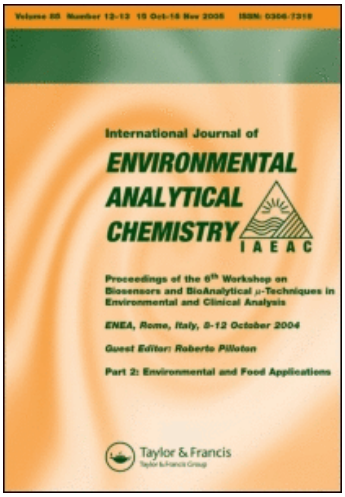

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Determination of Phenols from Automobile Exhaust by Means of High-Performance Liquid Chromatography (HPLC)†

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The paper describes a procedure to determine ten representative phenols (phenol, m-, p-, o-cresol, 2,4-, 2,6-, 3,4-, 3,5-xylenol, 2,3,6-, 2,4,6-trimethylphenol) in engine exhaust. Sampling was performed by a conventional Grimmer device. A sample of the water condensate collected was extracted with ether. After concentration of the ether extract phenols were isolated from other interfering compounds by stepwise elution on a Lobar column packed with LiChroprep Si 60 under controlled conditions. Relative recovery of the majority of reference phenols ranged from 90–100%. Separation of phenols into individual components was performed on polar packings as well as on Reverse Phase packings. Quantitative analysis was carried out on a prepacked Zorbax ODS column of 250 × 4.6 mm in acetonitrile/water of 30/70 (v/v) using a UV photometer of 254 nm wavelength. Phenols in the extract of the exhaust were identified on the basis of retention times and by spectroscopic means. Quantitative analysis was performed by peak height measurements using 2,4-dichlorophenol as internal standard. Concentration of phenols ranged from 1–10 ppm or 0.03–0.18 mg/m^3 of exhaust.

KEY WORDS: pollutants, automobile exhaust, HPLC of phenols, determination of phenols.

INTRODUCTION

Phenols and phenolic derivatives are considered potential pollutants. They arise from industrial waste, from the biological decay of certain pesticides and are also found in automobile exhaust and cigarette smoke. In the German Federal Republic the maximum allowed concentration (the so-called MAK-value) of phenol is limited to 5 ppm or 19 mg/m^3 due to its toxicity'[1] Phenol is determined by two acknowledged methods, the procedure according to Lahmann[1] and the 4-aminoantipyrine method.[2]

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The presence of phenols was first indicated in automobile exhaust by Smith *et al.*,[3] Hoffmann and Wynder[4] and Stanley *et al.*[5] The phenols o-, m-, p-cresol, 2,3-, 2,4-, 2,5-, 3,4- and 3,5-xylenol, 2-ethylphenol, tetrachlorophenol and pentachlorophenol were positively identified. The concentration of individual components measured was of the order of 0.2–2.3 mg/m^3 [6] and 0.5–100 ppm.[7] Gas Chromatography of native or silylated phenols,[6,7] Thin Layer[8–11] and Paper Chromatography[12–14] were the preferred methods for the estimation of phenols. The two latter chiefly employ dye reactions for improved detection. Karasek *et al.*[15] described the analysis of phenols extracted from diesel exhaust particulates by means of a GC-MS system.

The objective of the present work was to devise a procedure for estimating phenols from engine exhaust using High-Performance Liquid Chromatography (HPLC) techniques. HPLC offers advantages over other chromatographic methods in the analysis of such polar compounds, as no derivatisation is required and the separation is carried out at ambient temperature. The most critical and important steps of the investigation, the enrichment and isolation of the phenols from other constituents of the exhaust, are described in detail. Sampling was carried out using a Grimmer device.[16]

EXPERIMENTAL

Sampling

A single cylinder diesel engine and an Otto engine (a reciprocating petrol engine) were employed as test motors. Both engines were driven at two test levels: idling and full throttle. In each case the outlet of the exhaust pipe of the engine was connected to a conventional Grimmer device.[16] Sampling time was two hours. The determination of phenols was confined to the water condensate.

Methods for selective enrichment and isolation of phenols (see Table I)

Two different procedures were applied.

Procedure 1: (two consecutive extractions)

20 ml of the water condensate were added to 20 ml of 4 N sodium hydroxide solution, reagent grade. 10 ml of this alkaline solution were extracted with 5 × 10 ml of diethyl ether, reagent grade. The remaining alkaline solution was then saturated with CO_2 at room temperature until $NaHCO_3$ precipitated (pH = 8.0). The phenols were extracted from the sodium hydrogen carbonate solution with 5 × 10 ml of diethyl ether, reagent grade.

Procedure 2: (extraction followed by stepwise elution)

20 ml of the water condensate were extracted five times with diethyl ether, reagent grade. The solvent was evaporated from the combined ether extracts and the residue taken up in 1.5 ml n-pentane/diethyl ether 2:1 (v/v). This was then injected into a Lobar column size A, filled with LiChroprep Si 60 (dp = 40–63 μm) of E. Merck, Darmstadt. The column was pre-treated with n-pentane, reagent grade. The first five eluates (each 75 ml of: n-pentane, n-pentane/dichloromethane of 97/3 (v/v), n-pentane/dichloromethane of 93/7 (v/v), n-pentane/dichloromethane of 86/14 (v/v) and n-pentane/diethyl ether of 96/4 (v/v)) were discarded. The sixth and seventh fractions, which contain the phenols, were obtained by elution with 75 ml of n-pentane/diethyl ether of 89/11 (v/v) and 75 ml of n-pentane/diethyl ether of 77/23 (v/v).

Relative recovery of a series of reference phenols (listed in Table 2) was examined for both procedure 1 and 2 by two analytical techniques:

(i) UV photometric detection between 250–300 nm on a Leitz-Unicam photometer at a split width of 0.002 mm using the peak height method.[17].

(ii) potentiometric titration using a 0.1 N tetra-n-butylammonium hydroxide solution in benzene/methanol (10:1 v/v).[18] The measuring electrode was a conventional glass electrode; the reference electrode of type Ag/AgCl was filled with ethanol saturated with tetra-n-butylammonium iodide solution. Reproducibility was improved on substituting acetone for pyridine as solvent.[19] Titration was carried out at 285 K under dry nitrogen atmosphere.

Separation of phenols by HPLC

HPLC examinations were carried out

(a) on a DuPont Liquid Chromatograph 830 fitted with a 254 nm UV photometer and a flow controller.

(b) on a Hewlett-Packard Liquid Chromatograph 1084 A fitted with a 254 nm UV photometer and an automatic injection system. Columns were 250 × 4.6 mm. Several packings in combination with various eluent compositions were employed to determine the optimum phase system, e.g.

LiChrosorb Si 60, dp = 5 μm (E. Merck, Darmstadt)	n-heptane/acetonitrile of 98/2 (v/v) n-heptane saturated with acetonitrile n-heptane/methanol of 98/2 (v/v) n-heptane/acetic acid of 98/2 (v/v)
Nucleosil 5 NO_2, dp = 5 μm (Macherey & Nagel, Düren)	

LiChrosorb RP 8, dp = 5 μm	methanol/water of 40/60, 50/50, 60/40 (v/v)
LiChrosorb RP 18, dp = 5 μm (both of E. Merck, Darmstadt)	dimethylformamide/water of 50/50, 55/45, 60/40 (v/v)
ODS Hypersil, dp = 5 μm (Shandon, Frankfurt/M.)	acetonitrile/water of 20/80, 30/70, 35/65 (v/v)
Zorbax ODS, dp = 6 μm (as prepacked column, DuPont de Nemours Deutschland GmbH, Bad Nauheim)	dioxane/water of 35/65, 40/60, 50/50 (v/v) tetrahydrofuran/water of 40/60, 35/65, 30/70 (v/v)

Injection volume was 5 μl.

Qualitative analysis

Phenols were identified on the basis of retention time of reference substances. In addition, the resolved peaks of individual components were sampled and components were identified by UV-, NMR- and mass spectrometry.

Quantitative analysis

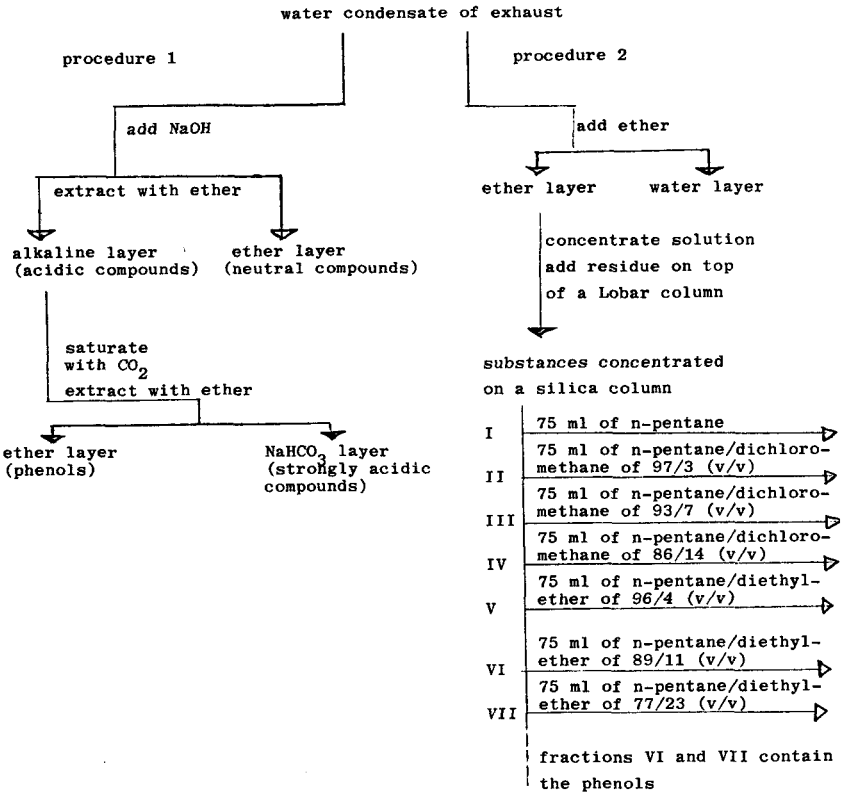
The internal standard was 2,4-dichlorophenol, reagent grade. Calibration factors f_i^h and f_i^a were derived from peak height and peak area measurements, respectively. The dependence of peak height, peak area and calibration factor on the chromatographic variables in quantitative analysis has been discussed in a previous paper.[20]

RESULTS AND DISCUSSION

Selective enrichment of phenols from automobile exhaust

The Grimmer sampling device,[16] originally developed for collecting the particulate matter of exhaust to measure the content of polynuclear aromatics, is suitable for use in this project. Most of the phenolic compounds emitted in the exhaust are water-soluble, and it is expected that they will accumulate in the condenser of the sampling device. Another possible method for selective enrichment would be the collection of phenols and other soluble compounds by an absorber system containing an absorbing liquid. This system can be arranged in such a way that

Table 1: Enrichment and isolation of phenols



only a part of the exhaust passes through the liquid. This is achieved by insertion of a probe in the centre of the cross-section of the exhaust pipe. The device was successfully applied by us in the enrichment of PNA from diesel engine exhaust,[21] but not yet used for phenols.

The enrichment of phenols is commonly accomplished by extraction with 2 N sodium hydroxide solution. This was first recommended by Hoffmann and Wynder.[22] Another procedure which is frequently applied is the carbon-chloroform extraction method.[23] The precision of this approach, however, strongly depends on the batch-to-batch reproducibility of the active carbon employed as support.

In our investigation we used first the traditional sodium hydroxi-

de/ether extraction and second a simple ether extraction of the native water condensate. In the first case an enrichment of acid compounds was achieved, whereas in the second case the ether extract contained acidic, neutral and basic compounds.

Separation into groups

Group separation is a widely accepted procedure in the resolution of complex mixtures, as in the analysis of pollutants emitted from engines. The standard procedures applied to phenols are those which are commonly in use in organic chemistry, such as steam-distillation, extraction from sodium hydrogen carbonate solution with ether and graduated elution on an adsorbent column. These procedures have to be carried out under strictly defined conditions and recovery studies must be made to check the efficiency of separation. We applied two different methods, as indicated in Table 1, for the separation of phenols from interfering compounds. Following procedure 1, after ether extraction the alkaline layer was adjusted to pH = 8.0 by introducing carbon dioxide, thus forming a sodium hydrogen carbonate solution.

On extraction with ether, phenols were separated at this stage from other more acidic compounds, the latter remaining as sodium salts in the $NaHCO_3$ layer. In procedure 2 the concentrated extract, taken up in n-pentane/diethyl ether of 2:1 (v/v) was injected on a standardized silica column. Separation of phenols was then accomplished by stepwise elution as indicated in table I. Separation from other polar compounds such as acids was tested by measuring the elution profiles of test mixtures containing reference phenols and carboxylic acids. Procedure 2 has the advantage that phenols can be resolved from less polar as well as from stronger polar compounds. The HPLC elution profile of the phenolic fraction obtained in procedure 2 shows that the solutes retained have a polarity ranging from phenol to trimethylated phenols, whereas the fraction isolated in procedure 1 contains a substantial amount of strongly polar compounds which renders the detection and quantitative determination of phenol more difficult.

The results of the recovery studies of a series of reference phenols following procedure 1 and 2 are collected in Table II. The data indicate recoveries in the range 90 to 100% for both procedures with the exception of o-cresol, 2,6- and 3,4-xylenol in procedure 1. Procedure 2 was adopted for quantitative work.

Choice of optimum phase system in HPLC

A test mixture of 10 representative phenols was employed to examine the

TABLE II

Relative recovery of phenols methods applied for quantitative analysis: (a) UV-spectroscopy (b) potentiometric titration

Compound	Relative recovery of phenols % enrichment and isolation according to			
	procedure 1		procedure 2	
	(a)	(b)	(a)	(b)
phenol	95,8 ± 1,7	94,1 ± 2,1	92,3 ± 1,0	97,4 ± 1,2
o-cresol	99,0 ± 1,1	85,5 ± 2,3	—	98,2 ± 2,0
m-cresol	100,0 ± 1,6	—	92,8 ± 1,6	100,0 ± 2,4
p-cresol	98,2 ± 2,1	95,6 ± 2,1	—	93,4 ± 1,3
2,4-xylenol	—	93,9 ± 2,0	—	100,9 ± 1,3
2,6-xylenol	98,2 ± 2,1	87,9 ± 5,7	—	101,0 ± 1,3
3,4-xylenol	—	88,4 ± 2,0	—	100,0 ± 2,0
3,5-xylenol	98,9 ± 1,2	91,8 ± 1,8	96,0 ± 1,8	99,0 ± 1,4
2,3,6-trimethylphenol	99,5 ± 1,0	100,0 ± 2,3	—	101,0 ± 1,4
2,4,6-trimethylphenol	—	90,9 ± 2,0	—	100,0 ± 2,0

resolution of more than 50 different phase systems listed in the experimental section. Both polar and unpolar, i.e. Reverse Phase, packings were included in the study. The nature of the support influences the elution sequence of the phenols, whereby the unpolar packing reverses the order produced for the polar one. Thus certain substances can be shifted to the beginning or the end of the chromatogram. This is useful in practice for the removal of an observed accumulation of unresolved peaks from one particular spot in a chromatogram to a more convenient location. The selectivity can then be enhanced by the choice of a suitable solvent composition. This approach appears to be a most practical one since the phenolic fraction still contains several dozen compounds in addition to the small number of reference substances and the composition as well as the relative amount can be expected to vary widely depending on the type of motor and its operating conditions. A selection of data obtained for some phase systems is shown in Table III.

Based on the observed selectivity coefficients r_{ji} of pairs of phenols, it is clear that no major differences exist between native and chemically modified silica packings. In some cases specific pairs such as p- and m-cresol, 3,5- and 2,6-xylenol remain difficult to resolve. Analysis time ranged from 20 to 50 minutes. It is also clear that for a given packing, selectivity can be improved to an appreciable extent by changing the solvent composition. A second optimization of the phase system must be carried out after separating the fractions from the extract. The phase

TABLE 3

Capacity factors k' and selectivity coefficients r_{ji} of phenols measured on various HPLC phase systems column dimensions: 250 × 4.6 mm

	phase system n-heptane/acetonitrile of 98/2 (v/v)				phase system n-heptane/acetic acid of 98/2 (v/v)			
	LiChrosorb Si 60		Nucleosil 5 NO_2		LiChrosorb Si 60		Nucleosil 5 NO_2	
phenolic compound	k'	r_{ji}	k'	r_{ji}	k'	r_{ji}	k'	r_{ji}
2,4,6-trimethylphenol	1,94	1,09	2,38	1,10	1,00	1,08	0,81	1,01
2,3,6-trimethylphenol	2,12	1,45	2,63	1,29	1,08	2,01	0,82	1,68
2,6-xylenol	2,43	3,16	3,38	4,51	2,17	3,00	1,38	3,25
2,4-xylenol	7,69	1,42	15,25	2,08	6,50	1,68	4,49	1,83
3,5-xylenol	10,92	1,00	31,75	0,50	10,92	1,07	8,22	0,88
o-cresol	10,93	1,25	16,00	1,38	11,67	1,11	7,23	1,36
3,4-xylenol	13,68	1,25	22,13	1,24	13,00	1,51	9,82	1,32
m-cresol	17,07	1,01	27,50	1,01	19,58	0,99	12,94	1,02
p-cresol	17,28	1,71	27,75	1,30	19,33	1,71	13,23	1,75
phenol	29,57		36,00		33,00		23,12	

Table III continued (1)

	phase system methanol/water of 50/50 (v/v)							
	LiChrosorb RP-8		LiChrosorb RP-18		ODS Hypersil		Zorbax ODS	
phenolic compound	k'	r_{ji}	k'	r_{ji}	k'	r_{ji}	k'	r_{ji}
phenol	1,14	1,83	1,30	2,02	1,00	2,03	1,38	2,01
p-cresol	2,09	1,02	2,63	0,98	2,03	1,04	2,77	1,00
m-cresol	2,13	1,06	2,58	1,11	2,12	1,06	2,78	1,12
o-cresol	2,26	1,49	2,86	1,59	2,24	1,67	3,11	1,56
3,4-xylenol	3,37	1,13	4,56	1,14	3,75	1,16	4,84	1,10
3,5-xylenol	3,81	1,01	5,19	1,00	4,34	0,92	5,33	1,02
2,6-xylenol	3,83	1,14	5,20	1,12	4,00	1,14	5,41	1,08
2,4-xylenol	4,38	1,42	5,81	1,66	4,54	1,60	5,82	1,62
2,3,6-trimethylphenol	6,23	1,12	9,67	1,10	7,26	1,13	9,43	1,13
2,4,6-trimethylphenol	7,00		10,67		8,19		10,69	

Table III continued (2)

	phase system acetonitrile/water of 30/70 (v/v)							
	LiChrosorb RP-8		LiChrosorb RP-18		ODS Hypersil		Zorbax ODS	
phenolic compound	k'	r_{ji}	k'	r_{ji}	k'	r_{ji}	k'	r_{ji}
phenol	0,85	2,11	2,50	2,08	1,48	1,97	1,68	1,96
p-cresol	1,79	0,94	5,19	1,00	2,91	1,00	3,29	0,98
m-cresol	1,69	1,20	5,19	1,14	2,91	1,13	3,24	1,15
o-cresol	2,03	1,47	5,91	1,60	3,30	1,47	3,73	1,47
3,4-xylenol	2,99	1,13	9,48	1,14	4,86	1,14	5,47	1,14
3,5-xylenol	3,38	1,22	10,76	1,14	5,56	1,12	6,26	1,20
2,6-xylenol	4,14	0,94	12,28	1,00	6,22	1,00	7,54	0.93
2,4-xylenol	3,89	1,94	12,28	1,79	6,21	1,81	7,02	1,93
2,3,6-trimethylphenol	7,55	1,08	21,99	1,07	11,22	1,06	13,57	1,07
2,4,6-trimethylphenol	8,16		23,53		11,92		14,55	

Table III continued (3)

	phase system dimethylformamide/water of 50/50 (v/v)		phase system DMSO/water of 50/50 (v/v)	
	Zorbax ODS		Zorbax ODS	
phenolic compound	k'	r_{ji}	k'	r_{ji}
phenol	1,82	1,78	4.06	1,97
p-cresol	3,24	1,02	8,00	1,05
m-cresol	3,29	1,18	8,40	1,10
o-cresol	3,88	1,29	9,28	1,70
3,4-xylenol	4,96	1,16	15,74	1,14
3,5-xylenol	5,76	1,05	18,00	0,79
2,6-xylenol	6,05	1,12	14,30	1,44
2,4-xylenol	6,80	1,48	20,57	—
2,3,6-trimethylphenol	10,09	1,10	—	—
2,4,6-trimethylphenol	11,07		—	—

Table III continued (4)

	phase system tetrahydrofuran/water of					
	30/70 (v/v)		35/65 (v/v)		40/60 (v/v)	
	Zorbax ODS		Zorbaz ODS		Zorbax ODS	
phenolic compounds	k'	r_{ji}	k'	r_{ji}	k'	r_{ji}
phenol	5,95	1,64	2,67	1,57	1,61	1,43
p-cresol	9,76	1,02	4,20	1,09	2,30	0,95
m-cresol	10,00	1,30	4,59	1,28	2,19	1,27
o-cresol	12,99	1,25	5,87	1,08	2,79	1,03
3,4-xylenol	16,22	1,15	6,33	1,07	2,88	1,13
3,5-xylenol	18,66	1,27	6,77	1,36	3,24	1,31
2,6-xylenol	23,62	0,91	9,22	0,89	4,26	0,96
2,4-xylenol	21,59	2,67	8,23	1,85	4,09	1,58
2,3,6-trimethylphenol	57,63	1,05	15,20	1,04	6,45	1,01
2,4,6-trimethylphenol	60,32		15,75		6,52	

systems offering the largest resolution and peak capacity were LiChrosorb RP 18 and Zorbax ODS using acetonitrile/water of 30/70 (v/v).

Detection of phenols is easily performed using a common UV-photometer operating at 254 nm wave length, phenols absorbing light sufficiently strongly in this range for the ppm concentration levels observed in practice. Fig. 1a and b show chromatograms of an experimental extract taken from one water condensate and subsequently separated on the identical HPLC column, the only difference being that the sample of chromatogram (a) was enriched and isolated according to procedure 2 whereas the sample of chromatogram (b) was purified according to procedure 1.

It is obvious that the extraction of water condensate with ether followed by stepwise elution provides a better resolution that procedure 1. The

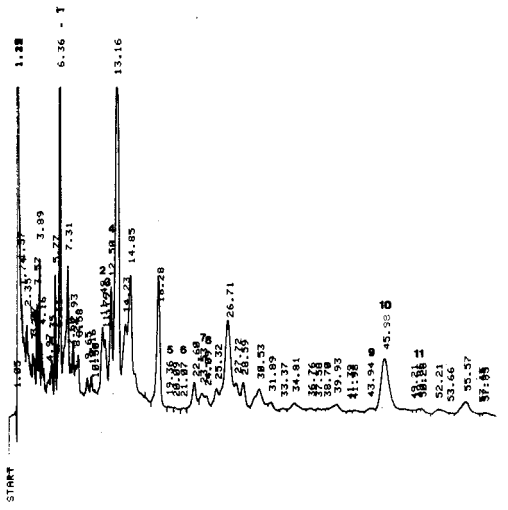


FIGURE 1a

FIGURE 1a Chromatogram of an extract of phenols enriched and isolated according to procedure 2.

conditions:	
column:	250 × 4.6 mm
packing:	Zorbax ODS, dp = 6 μm (prepacked column)
eluent:	acetonitrile/water of 30/70 (v/v)
flow rate:	0.8 ml/min
detector:	UV 254 nm
injection volume:	5 μl

The numbers indicate the retention times in minutes

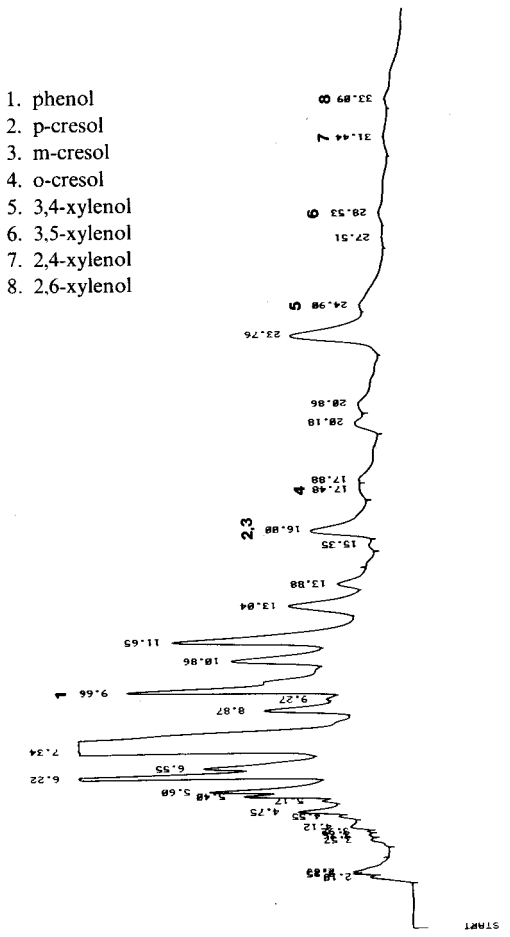


FIGURE 1b Chromatogram of an extract of phenols enriched and isolated according to procedure 1. Conditions as in Fig. 1a except the flow rate (1.6 ml/min).

positions of reference phenols in both chromatograms were indicated by retention times. In the case of chromatogram (b) fractions were collected and phenols were identified by spectroscopic methods. Inspection of chromatogram (b) shows that many compounds are eluted before the reference phenols, implying a more polar character, but which nevertheless cannot be carboxylic acids. These substances could possible comprise 2 or more aromatic ring systems carrying polar functional groups. Chromatogram (a) of the extract purified according to procedure 2 only includes substances ranging from phenol to trimethylated phenols, i.e. compounds composed of a single aromatic ring system.

Determination of phenols

Procedure 2 was chosen for enrichment and isolation. Separation of phenols into individual species was performed on a prepacked Zorbax ODS column of 250×4.6 mm in acetonitrile/water of 30/70 (v/v). The internal standard method was preferred to the external and absolute methods. The internal standard was 2,4-dichlorophenol which appeared between 2,3,6-trimethylphenol and 2,4,6-trimethylphenol in the chromatogram of Fig. 1a. Table IV contains the calibration factors of reference phenols measured from peak height and peak area, respectively, related to the standard. Some phenols of the extract could not be completely resolved and peak height was measured instead of peak area. Compounds following the cresols on the chromatogram required an enhancement of the sensitivity of the UV-detection system for a sufficiently high precision to be obtained.

The relative reproducibility of the determination was estimated at $\pm 5\,\%$. Table IV also presents some representative data of phenol content in diesel engine exhaust expressed in μg per ml condensate and mg per m^3 of exhaust. Phenol shows the highest concentration of all compounds but the content of the others is of the same order of magnitude. The concentrations found are in agreement with those of other published data.[6,7]

TABLE 4

Calibration factors and content of phenols in diesel engine exhaust

f_i^h calibration factor based on peak height measurements. f_i^a calibration factor based on peak area measurement internal standard: 2,4-dichlorophenol

			content	
	f_i^h	f_i^a	Q_i^h	Q_i^h
compound			*μg/ml of condensate*	*mg/m^3 of exhaust*
phenol	22,53	5,50	7,87	0,183
m-cresol	9,42	3,62	5,02	0,117
p-cresol	7,02	2,62	5,80	0,135
o-cresol	10,67	4,49	6,40	0,149
3,4-xylenol	3,94	2,18	1,33	0,031
3,5-xylenol	3,85	2,21	1,36	0,032
2,4-xylenol	2,88	1,09	4,56	0,106
2,6-xylenol	4,10	2,86	2,56	0,060
2,4,6-trimethylphenol	1,14	—	3,45	0,080
2,3,6-trimethylphenol	1,26	—	3,13	0,073

Conclusion

The results demonstrate that the method developed is easily adaptable to determine simple phenols in the ppm range in engine exhaust. By slight modification it can also be applied to the determination of phenols in industrial waste water and both surface and drinking water. In addition, it appears possible to identify and to estimate other phenolic compounds occuring in the exhaust as these are well-resolved by the HPLC-system. Moreover, applying procedure 2 to enrichment and isolation, other groups of constituents of the exhaust can be eluted by appropriate solvent compositions. The stepwise elution provides a highly reproducible method for the isolation of fractions which differ slightly in polarity. This furthermore facilitates the analysis of fractions into individual compounds.

It should be particularly emphasized that the application of HPLC techniques carries with it the potential to carry out semi-preparative or preparative separations for the collection of large amounts of substances for identification.

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