Phenols and Aromatic Alcohols (Phenol; 2- and 4-Methylphenol; DL-1- and 2-Phenylethanol; 3-Methylbenzyl alcohol; 2-Ethylphenol; 2,4-, 2,3- and 3,4-Dimethylphenol)

Application	Determination in urine
Analytical principle	Capillary gas chromatography
Completed in	September 1985

Summary

This method makes it possible to determine 10 phenols or aromatic alcohols sensitively and simultaneously in a single operation. The substances are either taken up as such or they are produced during the metabolic breakdown of aromatic hydrocarbons in the human body. The method is characterized by good analytical reliability, especially specificity, and resistance to interference. Thus it may be particularly recommended for routine use.

Acidified urine together with 3-ethylphenol which serves as an internal standard is subjected to steam distillation. This releases the phenols from their glucuronic or sulfuric acid conjugates and separates them simultaneously from the biological matrix. After extracting the distillate with an organic solvent, the phenols and alcohols are separated from each other and from other substances by capillary gas chromatography and determined using a flame ionization detector. Aqueous standards, which are processed as for the samples, are used for calibration.

Phenol

Within-series imprecision:	Standard deviation (rel.)	s_w = 16.8–7.3 %
	Prognostic range	u = 38.0–16.5 %
	At concentrations ranging from 12.0–26.8 mg phenol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 93–95 %

2-Methylphenol

Within-series imprecision:	Standard deviation (rel.)	s_w = 12.0 – 4.9 %
	Prognostic range	u = 27.1–11.1 %
	At concentrations ranging from 1.1–2.5 mg 2-methylphenol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 104–108 %

4-Methylphenol

Within-series imprecision:	Standard deviation (rel.)	s_w = 7.6–5.1 %
	Prognostic range	u = 17.2–11.5 %
	At concentrations ranging from 25.3 – 41.7 mg 4-methyl-phenol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 106–109 %

DL-1-Phenylethanol

Within-series imprecision:	Standard deviation (rel.)	s_w = 6.1– 4.6 %
	Prognostic range	u = 13.8–10.4 %
	At concentrations ranging from 1.8–4.6 mg DL-1-phenyl-ethanol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 95–96 %

2-Phenylethanol

Within-series imprecision:	Standard deviation (rel.)	s_w = 7.1–3.9 %
	Prognostic range	u = 16.1–8.8 %
	At concentrations ranging from 1.1–2.8 mg 2-phenyl-ethanol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r =100 %

3-Methylbenzyl alcohol

Within-series imprecision:	Standard deviation (rel.)	s_w = 6.5 – 4.7 %
	Prognostic range	u = 14.7–10.6 %
	At concentrations ranging from 0.9 –2.3 mg 3-methylbenzyl alcohol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 89 –90 %

2-Ethylphenol

Within-series imprecision:	Standard deviation (rel.)	s_w = 7.9–5.1 %
	Prognostic range	u = 17.9–11.5 %
	At concentrations ranging from 1.9–4.7 mg 2-ethylphenol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 89–90 %

2,4-Dimethylphenol

Within-series imprecision:	Standard deviation (rel.)	s_w = 6.7–2.3 %
	Prognostic range	u = 15.2–5.2 %
	At concentrations ranging from 1.9–4.7 mg 2,4-dimethyl-phenol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 92–95 %

2,3-Dimethylphenol

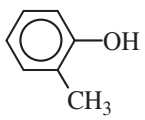
Within-series imprecision:	Standard deviation (rel.)	s_w = 5.9–4.7 %
	Prognostic range	u = 13.3–10.6 %
	At concentrations ranging from 1.0–2.3 mg 2,3-dimethyl-phenol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 92–100 %

3,4-Dimethylphenol

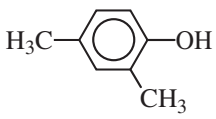
Within-series imprecision:	Standard deviation (rel.)	s_w = 5.8–4.1 %
	Prognostic range	u = 13.1–9.3 %
	At concentrations ranging from 1.1–2.7 mg 3,4-dimethyl-phenol per litre urine and where n = 10 determinations	
Inaccuracy:	Recovery rate	r = 97–109 %

Detection limit: approx. 0.3 mg Phenol or Aromatic Alcohol per litre urine

Phenols



2-Methylphenol
(o-Cresol)



2,4-Dimethylphenol
(2,4-Xylenol)

Determination of the above phenols in biological material, especially in urine, is suited for the screening of persons who are exposed to:
- benzene, toluene, the xylenes and ethylbenzene (BTX aromatics), or to
- these phenols

The BTX aromatics are employed in huge quantities for a wide variety of industrial purposes [1]. As constituents of petrol for motorized vehicles these aromatics also play an important role as pollutants of the atmosphere [2]. Since large numbers of people are therefore exposed to these highly volatile organics the evaluation of the health risk is of considerable importance.

In principle, biological monitoring of persons exposed to BTX aromatics may be carried out by measuring:

– the level of the substances in alveolar air or in blood, or
– the excretion of metabolites in urine

The excretion of the phenols seems to be of particular diagnostic value because of its close metabolic link with the proved or potential damaging principle of these aromatic hydrocarbons. The leukaemogenic properties of benzene, for example, are ascribed to its epoxide which is produced in intermediary metabolism in the human body and which is capable of reaction with the genetic material of human cells. Since it was recognized that the oxidation of the aromatic nucleus is a side branch of the human metabolism of alkylbenzenes [3, 4], the determination of alkylphenols in urine has been considered of greater significance. They, or their metabolic precursors could, in fact, be responsible for the blood count changes which have been observed in exposed spray-painters [5]. Phenol and 4-methylphenol are excreted physiologically but 2-methylphenol and the isomers of dimethylphenol are not normally present in urine.

The mean phenol level in urine of normal individuals is given as 4.6 to 9.4 mg/L [6]. The mean physiological urine level of 4-methylphenol is of the order of 50 mg/L [7].

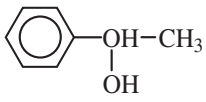
Average levels of 43 mg phenol, 2.7 mg 2-methylphenol and 19 mg 2,4-dimethylphenol per litre urine were excreted after occupational exposure to BTX aromatic concentrations of about 5 ml/m^3 (TRK value [Technical Guiding Concentration] for benzene) or 100 ml/m^3 (MAK value [1986] for toluene, xylene and ethylbenzene) [8, 9].

Phenol, cresols and dimethylphenols are employed in a large number of industrial processes such as productions of plastics, explosives, pharmaceuticals, etc. [10].

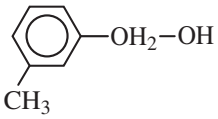
The central nervous system is the target organ during chronic assimilation of these phe-nolics which can result in headaches, nausea, disorientation, etc. Skin contact with these substances results in manifestations ranging from depigmentation of the skin to necrotic changes. The MAK value (1985) for both phenol and the isomers of cresol is given as 5 mL/m^3.

These substances have proved to be extremely problematical both in the technical aspects of worker protection and from the occupational medical viewpoint because they are assimilated at the work place not only via the lungs but also directly from the gas phase through the intact skin. Normal work clothes offer no protection against such substances. After exposure to phenol at a concentration in the region of the MAK value a urine concentration of about 285 mg/L may be expected [11].

Aromatic alcohols



DL-1-Phenylethanol



3-Methylbenzyl alcohol

The aromatic alcohols have only a limited use in industry. The phenylethanols, for example, are used in the production of flavourings, soaps, perfumes, etc., because of their pleasant odour. Limit values for tolerable concentrations of these substances in the air of such work places are currently not available.

More importance is assigned to these substances as intermediates in the breakdown of ethylbenzene and xylene in the human body. They are excreted in urine after the assimilation of these aromatics. The aromatic alcohols are relatively non-poisonous. For screening purposes, therefore, the determination of these alcohols offers no advantages over the assay of the aromatic carboxylic acids or the alkylphenols. It is, however, important to separate them from these other metabolites to obtain a specific result.

More detailed accounts of the BTX aromatics and their metabolites may be found elsewhere [12–17].

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Phenols and Aromatic Alcohols (Phenol; 2- and 4-Methylphenol; DL-1- and 2-Phenylethanol; 3-Methylbenzyl alcohol; 2-Ethylphenol; 2,4-, 2,3- and 3,4-Dimethylphenol)

Application	Determination in urine
Analytical principle	Capillary gas chromatography
Completed in	September 1985

Contents

1 General principles

The acidified urine together with the internal standard, 3-ethylphenol, is subjected to steam distillation. This releases the phenols from their glucuronic or sulfuric acid conjugates and separates them simultaneously from the biological matrix. After extracting the distillate with an organic solvent the phenols and alcohols are separated from each other and from other substances by capillary gas chromatography with a flame ionization detector. Aqueous standards which are processed as for the samples are used for calibration.

2 Equipment, chemicals and solutions

2.1 Equipment

Gas chromatograph with capillary injection system for split sampling (sintered glass liner), flame ionization detector and chart recorder

Quartz capillary: length: 30 m; inner diameter: 0.32 mm Stationary phase: SE 54, film thickness 0.25 µm (e.g. ict, Frankfurt)

5 µL Syringe for gas chromatography

Steam distillation apparatus (constructed in the laboratory, see Fig. 1 of the Method "Phenol" [photometric] in Vol. 1 of this series)

Shaker

100 mL Separating funnels with PTFE taps

50 mL Conical flasks

25 mL Tapered bottom flasks

Glass funnels

Fluted filter papers

10 mL Graduated test tubes with screw caps

2 mL Glass tubes with screw caps (with a 1 mL graduation, if necessary calibrated in the laboratory)

10 mL Graduated pipettes

Pasteur pipettes

50, 100 and 250 mL Volumetric flasks

1, 2.5, 5, 10, 15, 20 and 25 mL Transfer pipettes

2.2 Chemicals

The phenols and aromatic alcohols used were of the highest available grade of purity. Moreover they were gently sublimed or distilled under vacuum before their use for calibration.

Phenol, p.a. (e.g. Merck)
2-Methylphenol, p.a. (e.g. Fluka)
DL-1-Phenylethanol, > 99 % (e.g. EGA)
4-Methylphenol, for synthesis (e.g. Merck)
2-Phenylethanol, > 99 % (e.g. EGA)
3-Methylbenzyl alcohol, > 97 % (e.g. EGA)
2-Ethylphenol, for synthesis (e.g. Merck)
3-Ethylphenol, for synthesis (e.g. Merck)
2,4-Dimethylphenol, for synthesis (e.g. Merck)
2,3-Dimethylphenol, for synthesis (e.g. Merck)
3,4-Dimethylphenol, for synthesis (e.g. Merck)
Acetic acid, 96 % (e.g. Suprapur from Merck)
Sulfuric acid, 96 %, p.a.
Dichloromethane, for residue analysis
Sodium sulfate, anhydrous, p. a.
Acetone, for residue analysis
Ultrapure water (ASTM type 1) or double-distilled water
Nitrogen gas, purified (99.999 %)
Hydrogen (99.90 %)
Synthetic air (80 % purified nitrogen, 20 % oxygen)

2.3 Solutions

25% Sulfuric acid:
Approx. 50 mL ultrapure water is placed in a 100 mL volumetric flask. 12.7 mL 96 % sulfuric acid is added with a graduated pipette. After mixing, the flask is filled to the mark with ultrapure water.

Internal standard:
Stock solution:
250 mg 3-ethylphenol is dissolved in ultrapure water in a 250 mL volumetric flask which is then filled to the mark (1 g/L).

Internal standard solution:
10 mL of stock solution is diluted to the mark in a 100 mL volumetric flask with ultrapure water (100 mg/L).

The internal standard solution must be prepared freshly for each analytical series.

2.4 Calibration standards

Starting solution:
Approx. 250 mg phenol and 4-methylphenol, 50 mg DL-1-phenylethanol, 2-ethylphenol, 2,4-dimethylphenol together with 25 mg 2-methylphenol, 2-phenylethanol, 3-methylbenzyl alcohol, 2,3- and 3,4-dimethylphenol are weighed out and dissolved in acetone in

a 250 mL volumetric flask. Then the flask is filled to the mark with acetone (1 g/L, 0.2 g/L and 0.1 g/L, respectively).

Stock solution:
10 mL of the starting solution is diluted to the mark in a 100 mL volumetric flask with ultrapure water (100 mg/L, 20 mg/L and 10 mg/L, respectively). Calibration standards containing the individual phenols and aromatic alcohols in the concentration range 1–50 mg/L are prepared from the stock solution by diluting with ultrapure water as shown in the following table:

Volume of stock solution	Final volume of calibration standard	Concentration of calibration standards		
		Phenol 4-Methylphenol	DL-1-Phenylethanol 2-Ethylphenol 2,4-Dimethylphenol	2-Methylphenol 2-Phenylethanol 3-Methylbenzyl alcohol 2,3- and 3,4-Dimethylphenol
mL	mL	mg/L	mg/L	mg/L
2.5	50	5	1	0.5
5	50	10	2	1.0
10	50	20	4	2.0
15	50	30	6	3.0
20	50	40	8	4.0
25	50	50	10	5.0

The starting solution may be stored for about 2 weeks in the deep freeze but the calibration standards and stock solution must be prepared freshly for each analytical series.

3 Specimen collection and sample preparation

Urine is collected in polyethylene bottles and acidified (1 mL glacial acetic acid to 100 mL urine) as soon as it reaches the laboratory. It is stored in the refrigerator until further processing. Since the phenols in urine are relatively well protected from oxidation by their conjugation with glucuronic and sulfuric acids, the specimens may be stored in the refrigerator for up to a week, if necessary. Longer storage (up to half a year) should take place in the deep freeze.

The urine specimens are warmed to room temperature and shaken thoroughly before processing. 10 mL urine is then transferred to the steam distillation apparatus. 1 mL internal standard solution (see Section 2.3) and 3 mL 96 % sulfuric acid are added by pipette. This mixture is then subjected to steam distillation whereby 50 mL distillate is collected in a 50 mL volumetric flask in about 10 min.

The distillate is acidified with 2 mL 25 % sulfuric acid, transferred to a 100 mL separating funnel with a PTFE tap and mixed with 15 mL dichloromethane. The phenols

and aromatic alcohols are extracted by 15 min on a shaker. The lower, organic phase is run out into a 50 mL conical flask containing approx. 2 g anhydrous sodium sulfate. After at least 1 h drying time, the organic phase is filtered into a 25 mL tapered bottom flask through a fluted filter paper which is then rinsed with 2.5 mL dichloromethane.

Nitrogen gas is then passed over the filtrate until it has been reduced to about 5 mL. The filtrate is transferred to a 10 mL graduated test tube with a screw cap. The tapered bottom flask is rinsed with 1 mL dichloromethane and the organic phase further reduced in volume to 1 mL in the same manner. The concentrated extract is then transferred with a Pasteur pipette to a 2 mL glass tube with a screw cap. This is then stored in the deep freeze until analysis which should take place as soon as possible.

A reagent blank should always be processed in parallel. For this purpose 10 mL ultrapure water is employed instead of urine.

4 Operational parameters for gas chromatography

Capillary column:	Material:	Quartz
	Length:	30 m
	Inner diameter:	0.32 mm
Stationary phase:	SE 54, film thickness 0.25 µm	
Detector:	Flame ionization detector	
Temperatures:	Column:	5 min 60 ° C increase 5 °C per min up to 110 °C 10 min 200 °C 5 min cooling
	Injection block:	250 °C
	Detector:	300 °C
Carrier gas:	Purified nitrogen with column pressure 820 hPa (12 psi, flow rate 40 mL/min)	
Sample volume:	0.5 µL	

Under the conditions given above the following retention times were obtained. They serve as a guide only and the operator must establish for himself the resolving power of the capillary column used and the resulting retention behaviour of the substances being analysed.

Phenol	6.2 min
2-Methylphenol	8.6 min
DL-1-Phenylethanol	8.8 min
4-Methylphenol	9.4 min
2-Phenylethanol	10.6 min
3-Methylbenzyl alcohol	11.3 min
2-Ethylphenol	11.5 min
2,4-Dimethylphenol	11.8 min

3-Ethylphenol	12.5 min
2,3-Dimethylphenol	12.7 min
3,4-Dimethylphenol	13.2 min

Figs. 1 and 2 show examples of gas chromatograms of a processed aqueous standard and a processed urine sample.

5 Analytical determination

The operational parameters are set up as given above and 0.5 µL extract is injected into the gas chromatograph for each sample.

6 Calibration

10 mL of each aqueous calibration standard (see Section 2.4) is processed as for the samples and analysed by gas chromatography. The peak height or area of the signal at the appropriate retention time is determined for each phenol and aromatic alcohol. If necessary, reagent blank values are subtracted. The peak height or area is divided by that for the internal standard. The resulting quotients are plotted against the employed concentrations of the aqueous standards in mg/L to produce the calibration curve. Fig. 3 shows such a calibration curve for the determination of 2-methylphenol (o-cresol). In the concentration range given for the aqueous standards the relationship between the gas chromatographic signal and the phenol and aromatic alcohol concentration is linear.

7 Calculation of the analytical result

The peak heights or areas of the signals at the retention times characteristic for each of the phenols and aromatic alcohols are determined. If necessary, reagent blank values are subtracted. These peak heights or areas are divided by the values of the internal standard signal. Using the quotients so obtained, the corresponding concentrations in mg/L of the individual phenols and aromatic alcohols in the urine sample are read off the calibration curve.

8 Standardization and quality control

Materials for quality control are currently not commercially available and must therefore be prepared in the laboratory.

9 Reliability of the method

9.1 Precision

Pooled urine from unexposed persons was spiked with two defined concentrations of each of the phenols and aromatic alcohols analogous to the described preparation of the aqueous calibration standards (see Section 2.4). Each of the spiked urine samples was processed and analysed 10 times to determine the within-series imprecision. The details of concentrations, relative standard deviations (s_w) and prognostic ranges (u) for each phenol and aromatic alcohol are given in Tab. 1.

9.2 Accuracy

Recovery experiments were carried out to check the accuracy of the method. Urine was spiked with two known concentrations of each of the phenols and aromatic alcohols – as described in Section 9.1 – and analysed. The recovery rates were determined by comparison with processed aqueous standards. The values for the individual phenols and aromatic alcohols are given in Tab. 1.

A comparison of the processed spiked urine samples with standards which had not been subjected to the sample processing but, taking the concentration step into account, had been dissolved directly in the dichloromethane showed that losses of between 25 and 48 % occur during the processing procedure. Losses of about 60 % were demonstrated for phenol. As the recovery rates determined above demonstrate, these losses are largely compensated by the calibration procedure described.

9.3 Detection limit

Since reagent blank values were not recorded, the detection limit was determined as three times the signal to noise ratio, which is equivalent to a concentration of about 0.3 mg phenol or aromatic alcohol per litre urine.

If a further reduction of the detection limit is required, the urine volume to be processed may be increased.

9.4 Sources of error

Under the capillary gas chromatographic conditions described 3- and 4-methylphenol (m- and p-cresol) as well as 3- and 4-methylbenzyl alcohol and 2,4- and 2,5-dimethylphenol are not completely separated from each other. In addition, the retention times of 4-and 3-ethylphenol and of 2-methylbenzyl alcohol and 2-ethylphenol are almost identical so that a complete separation of these substances is not obtained. The following substances, which are also extractable by steam distillation and which must be

expected to be found in urine after exposure or metabolism, do not interfere in the gas chromatographic determination: 2-chlorophenol, nitrobenzene, 2-chloro-5-methylphenol, 2- and 3-nitrophenol, 3- and 4-chlorophenol, 3,4-dichlorophenol, 4-nitrophenol and 1,2-dinitrobenzene.

The evaporation of the dichloromethane extracts is a critical step in the sample processing. Evaporation to dryness must be avoided. The volume reduction must be brought about by evaporation under a stream of nitrogen gas and not on a rotary evaporator as this would cause marked losses of alkylphenols. The extracts must be dried carefully before the concentration step to avoid interference in the gas chromatogram. The processed samples should be analysed as soon as possible. Storage should take place in the deep freeze.

10 Discussion of the method

With the method described here [18] ten different phenols and aromatic alcohols, which either occur at the work place or are excreted as metabolites of alkylbenzenes, may be separated by capillary gas chromatography and determined sensitively. Concentration ranges of interest in occupational medicine and to some extent in environmental studies can thus be reliably assayed.

The described method separates the phenols and aromatic alcohols from the biological matrix by steam distillation. The urine is simultaneously subjected to an acid hydrolysis which releases the phenols from their conjugates. Investigation of specimens from persons exposed to alkylbenzenes demonstrated that the acid volume recommended here is sufficient to ensure complete hydrolysis of these compounds. The steam distillation and the subsequent extraction step result in a capillary gas chromatographic determination which is free of interference. The absence of matrix effects makes it possible to carry out the calibration with aqueous standards which have been subjected to the same processing procedure. 3-Ethylphenol is used as internal standard. Since it can be steam distilled, it can be added before the sample is processed so that variations in distillation and extraction yields as well as possible losses during the concentration step may be largely compensated by the calculation of peak ratios. This makes it possible to determine the majority of these phenols and aromatic alcohols with very good precision even at low concentrations. Most other gas chromatographic methods which are described in the literature [19–23] employ acid hydrolysis to release the phenols followed by an extraction step. Unlike the steam distillation procedure in which the chemically labile free phenols are continuously distilled out of the reaction mixture, in such procedures they remain in the hydrolysis solution for the whole incubation period. This increases the danger of chemical modification of the free phenols; in addition, as confirmed by *Engström* [23], the hydrolysis does not proceed to completion.

The steam distillation technique has also been successfully employed with a subsequent high pressure liquid chromatographic determination of a number of phenols.

Instruments used:
Gas chromatograph 3700 with FID and recorder 9176 from Varian

11 References

[1] *K. Weissermel* and *H.-J. Arpe:* Industrielle organische Chemie. Verlag Chemie, Weinheim 1978.

[2] *W. Dulson:* Die Zusammensetzung von Vergaserkraftstoffen und ihrer Verbrennungsprodukte. In: *K. Aurand, U. Hässelbarth, E. Lahmann, G. Müller,* and *W. Niemitz* (eds.): Organische Verunreinigungen in der Umwelt. Erich Schmidt Verlag, Berlin 1978, p. 350–355.

[3] *V.* Sĕdivek and *J. Flek:* The absorption, metabolism, and excretion of xylenes in man. Int. Arch. Occup. Environ. Health *37,* 205–217 (1976).

[4] *J. Angerer:* Occupational chronic exposure to organic solvents. VII. Metabolism of toluene in man. Int. Arch. Occup. Environ. Health *43,* 63–67 (1979).

[5] *J. Angerer* and *H. Wulf:* Occupational chronic exposure to organic solvents. XI. Alkylbenzene exposure of varnish workers: effects on hematopoetic system. Int. Arch. Occup. Environ. Health *56,* 307–321 (1985).

[6] *J. Lewalter, Th. Schucht, J. Angerer,* and *K. H. Schaller:* Benzene derivatives, suitable for steam distillation. Vol. 1. of this series, 1985, p. 31–56.

[7] *J. Angerer:* Chronische Lösungsmittelbelastung am Arbeitsplatz. V. Chromatographische Methoden zur Bestimmung von Phenolen im Harn. Int. Arch. Occup. Environ. Health *42,* 257–268 (1979).

[8] *J. Angerer:* Prävention beruflich bedingter Gesundheitsschäden durch Benzol, Toluol, Xylole und Ethylbenzol. Vol. 71 der Schriftenreihe Arbeitsmedizin, Sozialmedizin, Präventivmedizin. A. W. Gentner Verlag, Stuttgart 1983.

[9] *J. Angerer:* Occupational chronic exposure to organic solvents. XII. O-cresol excretion after toluene exposure. Int. Arch. Occup. Environ. Health *56,* 323–328 (1985).

[10] *G. D. Clayton* and *F. E. Clayton* (eds.): Patty's industrial hygiene and toxicology, Vol. II A–C. Toxicology. John Wiley & Sons, New York 1981–1982.

[11] *H. Ohtsuji* and *M. Ikeda:* Quantitative relationship betweeen atmospheric phenol vapour and phenol in the urine of workers in Bakelite factories. Brit. J. Industr. Med. *29,* 70–73 (1972).

[12] *J. Angerer:* Xylole. In: *D. Henschler* and *G. Lehnert* (eds.): Biologische Arbeitsstoff-Toleranz-Werte (BAT-Werte). Arbeitsmedizinisch-toxikologische Begründungen. Deutsche Forschungsgemeinschaft, Verlag Chemie, Weinheim, 2nd issue 1985.

[13] *D. Henschler* (ed.): Gesundheitsschädliche Arbeitsstoffe, toxikologisch-arbeitsmedizinische Begründung von MAK-Werten. Deutsche Forschungsgemeinschaft, Verlag Chemie, Weinheim, 9th issue 1983.

[14] *WHO:* Study group on internationally recommended health based occupational exposure limits to selected organic solvents. Genf 17.–23.6.1979.

[15] *NIOSH:* Criteria for a recommended standard. Occupational exposure to xylene. HEW Publication No. 75–168 (1975).

[16] *J. Angerer, R. P. Deutsch-Wenzel, H. Hermann, H. Jungen,* and *K. Täuber:* Wirkung von Toluol auf Mensch und Tier. Deutsche Gesellschaft fur Mineralölwissenschaft und Kohlechemie e.V. Projekt 174-7. Hamburg 1985.

[17] *J. Angerer, R. P. Deutsch-Wenzel, H. Hermann, H. Jungen,* and *K. Täuber:* Wirkung von Xylol auf Mensch und Tier. Deutsche Gesellschaft fur Mineralölwissenschaft und Kohlechemie e.V. Projekt 174-8. Hamburg 1984.

[18] *R. Heinrich* and *J. Angerer:* Capillar-gas-chromatographische Bestimmung von Alkylphenolen im Harn. Fresenius' Z. Anal. Chem. *322,* 766–771 (1985).

[19] *W. Woiwode, R. Wodarz, K. Drysch,* and *H. Weichardt:* Metabolism of toluene in man: Gas-chromatographic determination of o-, m- and p-cresol in urine. Arch. Toxicol. *43,* 93–98 (1979).

[20] *P. Apostoli, F. Brugnone, L. Perbellini, V. Cocheo, M. L. Bellomo,* and *R. Silvestri:* Biomonitoring of occupational toluene exposure. Int. Arch. Occup. Environ. Health *50,* 153–168 (1982).
[21] *K. Hasegawa, S. Shiojima, A. Koizumi,* and *M. Ikeda:* Hippuric acid and o-cresol in the urine of workers exposed to toluene. Int. Arch. Occup. Environ. Health *52,* 197–208 (1983).
[22] *P. Pfäffli, H. Savolainen, P. L. Kalliomäki,* and *P. Kalliokoski:* Urinary o-cresol in toluene exposure. Scand. J. Work Environ. Health *5,* 286–289 (1979).
[23] *K. M. Engström:* Urinalysis of minor metabolites of ethylbenzene and m-xylene. Scand. J. Work Environ. Health *10,* 75–81 (1984).

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Tab. 1. Within-series imprecision, recovery experiments and detection limits for the determination of phenols and aromatic alcohols in urine by gas chromatography

Phenol or Aromatic alcohol	*n*	Expected value (Blank + spiked concentration) mg/L	Determined value $\bar{x}$ mg/L	s_w %	*u* %	Recovery rate %	Detection limit mg/L
Phenol	10	2.7 + 10.2	12.0	16.8	38.0	93	
	10	2.7 + 25.4	26.8	7.3	16.5	95	0.3
2-Methylphenol	10	0^+ + 1.0	1.1	12.0	27.1	108	
	10	0^+ + 2.4	2.5	4.9	11.1	104	0.3
4-Methylphenol	10	12.6 + 10.6	25.3	7.6	17.2	109	
	10	12.6 + 26.6	41.7	5.1	11.5	106	0.3
DL-1-Phenylethanol	10	0^+ + 1.9	1.8	6.1	13.8	95	
	10	0^+ + 4.8	4.6	4.6	10.4	96	0.3
2-Phenylethanol	10	0^+ + 1.1	1.1	7.1	16.1	100	
	10	0^+ + 2.8	2.8	3.9	8.8	100	0.3
3-Methylbenzyl alcohol	10	0^+ + 1.0	0.9	6.5	14.7	90	
	10	0^+ + 2.6	2.3	4.7	10.6	89	0.3
2-Ethylphenol	10	0^+ + 2.1	1.9	7.9	17.9	90	
	10	0^+ + 5.3	4.7	5.1	11.5	89	0.3
2,4-Dimethylphenol	10	0^+ + 2.0	1.9	6.7	15.2	95	
	10	0^+ + 5.1	4.7	2.3	5.2	92	0.3
2,3-Dimethylphenol	10	0^+ + 1.0	1.0	5.9	13.3	100	
	10	0^+ + 2.5	2.3	4.7	10.6	92	0.3
3,4-Dimethylphenol	10	0^+ + 1.1	1.1	5.8	13.1	109	
	10	0^+ + 2.8	2.7	4.1	9.3	97	0.3

$^+$ In unspiked urine the concentrations of these phenols and aromatic alcohols were below the detection limit

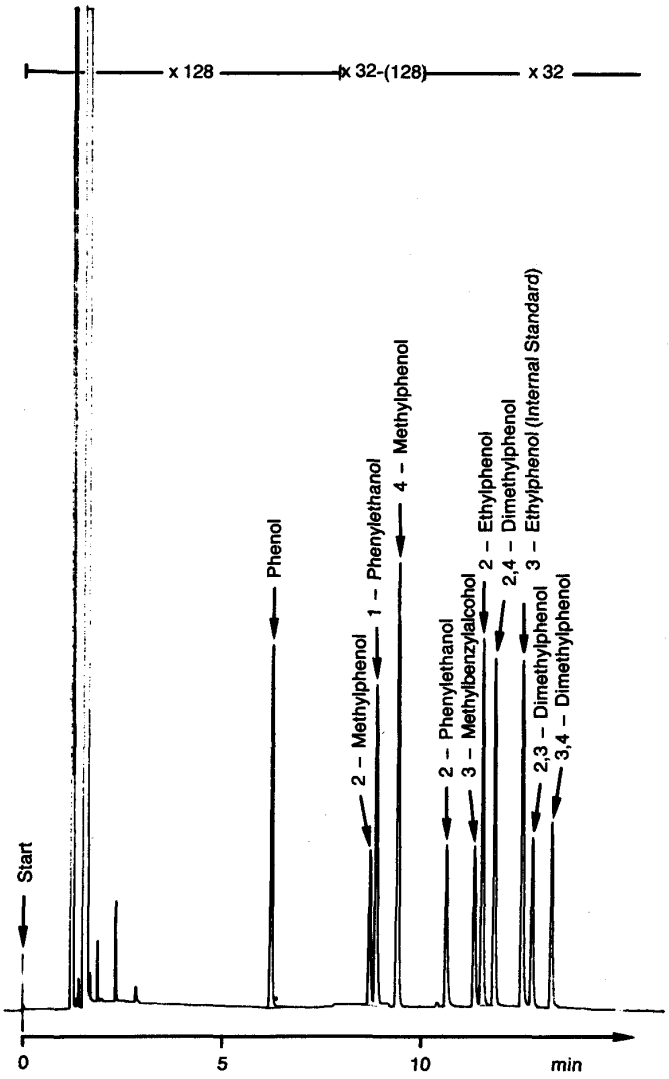

Fig. 1. Gas chromatogram of a processed aqueous standard containing 30 mg/L phenol and 4-methylphenol, 6 mg/L DL-1-phenylethanol, 2-ethylphenol and 2,4-dimethylphenol together with 3 mg/L 2-methylphenol, 2-phenylethanol, 3-methylbenzyl alcohol, 2,3- and 3,4-dimethylphenol. Operational parameters for capillary gas chromatography are given in Section 4.

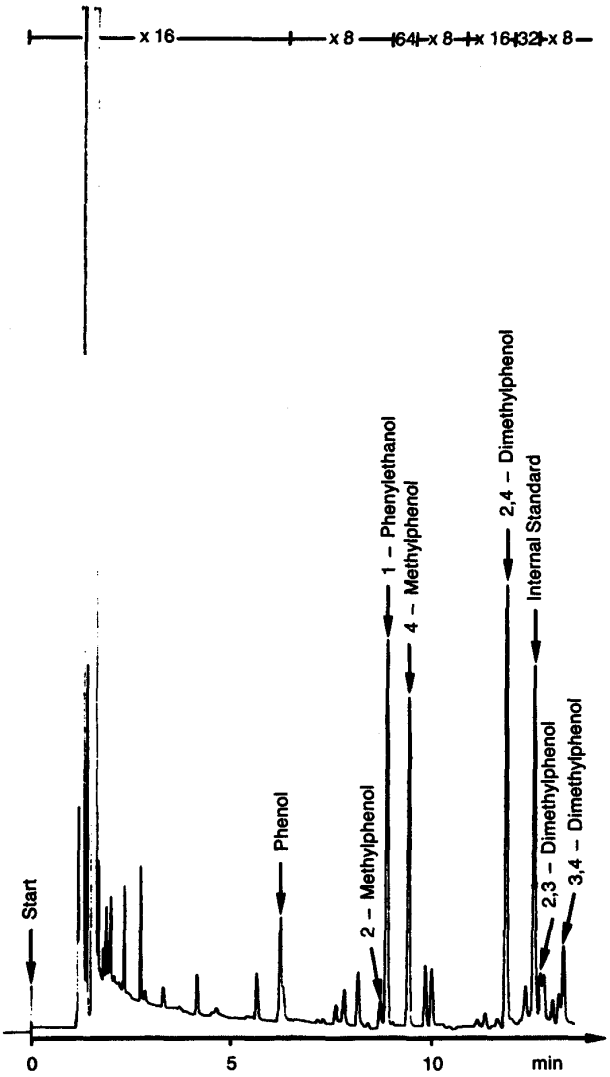

Fig. 2. Gas chromatogram of a processed urine specimen from an individual occupationally exposed to aromatic hydrocarbons.

Concentrations:		
	phenol	3.4 mg/L
	2-methylphenol	0.3 mg/L
	DL-1-phenylethanol	2.8 mg/L
	4-methylphenol	18.3 mg/L
	2,4-dimethylphenol	6.6 mg/L
	2,3-dimethylphenol	0.3 mg/L
	3,4-dimethylphenol	0.7 mg/L

Operational parameters for capillary gas chromatography are given in Section 4.

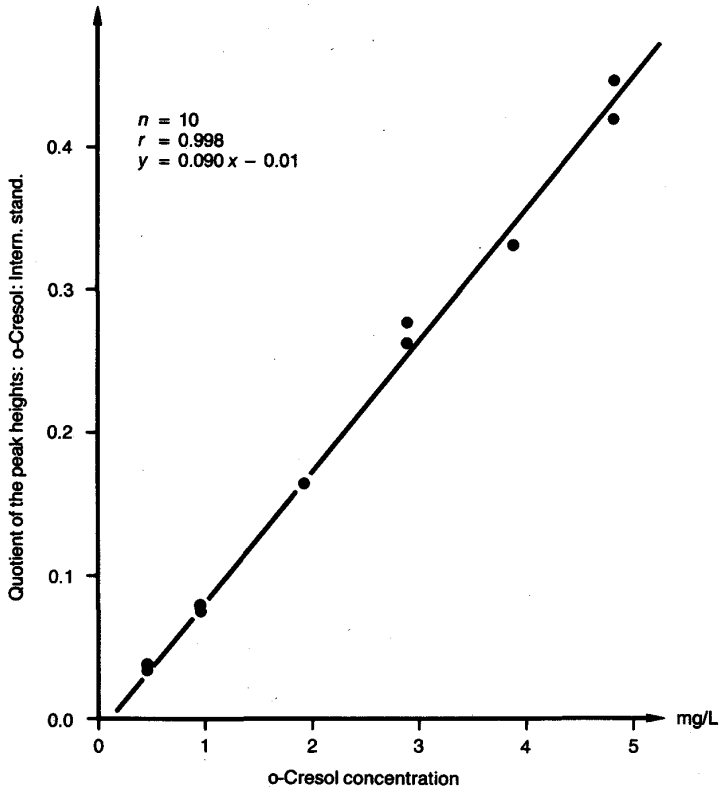

Fig. 3. Example of a calibration curve for the determination of 2-methylphenol in urine.