

B. Edler · C. Zwiener · F. H. Frimmel

Particle beam LC/MS screening of polar compounds of contaminated ground water samples from a former gas plant

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Abstract A commercial particle beam interface coupled to a single quadrupole mass spectrometer has been used for a nontarget screening of highly contaminated water samples from a former gas plant. A liquid chromatographic system has been used to separate hydrophilic compounds with a high percentage water gradient without any loss of efficiency in the MS interface. The ground water showed a high pollution burden in the total ion current chromatogram. Full scan EI mass spectra have been obtained for identification of unknown compounds which could be assigned to oxidation products of heterocyclic aromatic compounds, some known constituents of coal tar. The identification of 6(5H)-phenanthridinone has been confirmed by comparison with a corresponding standard. The difficulties of an unequivocal identification of isoquinolinone and methyl derivatives of quinolinone based on their EI mass spectra are discussed.

Introduction

At the beginning of this century, gasification processes were applied for the generation of city gas in nearly every town in Germany. These processes have received considerable attention because of the hazardous impact onto the environment during generation and purification of the gas. After the shut-down of almost all gas facilities in the 70th, highly contaminated sites have remained. Ground water samples taken from these sites often contain a great variety of aromatic and heterocyclic compounds [1] and their metabolites [2–4]. To estimate the potential environmental risk and the degree of contamination of these sites,

reliable analytical methods for the determination of both the contaminants and the possibly occurring polar metabolites are needed. The application of liquid chromatography (LC) coupled to mass spectrometry (MS) is a promising approach for a nontarget screening of contaminated water samples. This technique combines the advantages of the chromatographic separation power of LC for polar compounds and the confirmatory power available by EI mass spectra [5–7]. Furthermore, the mass spectra are amenable to computerized library searches. As a further advantage, the chemical ionization mode can be used to obtain additional information [8]. The aim of the work was to employ a commercially available particle beam interface for LC coupling to a single quadrupole mass spectrometer to estimate major contaminants in water samples obtained from a former gas plant.

Experimental

Sample collection

Sampling was done on the contaminated site of a former gas plant. Several wells of the ground water were sampled with a submersible pump (MP-1, Grundfos, Wahlstedt, Germany). The groundwater was pumped through a 3 m-well screen at the bottom of the well. The submersible pump was positioned 1.5 m below the water table. Before sampling, 7 well volumes of water were discarded to avoid the influence of stagnated water. The samples were then collected in 2 L stoppered glass bottles. The samples were cooled immediately and stored at 4°C in the dark.

Extraction and pre-concentration

Extraction and pre-concentration were carried out by solid phase extraction (SPE). Clear water samples were used without pretreatment. To avoid plugging of the SPE-column, cloudy samples were centrifuged for 20 min at 5000 g to remove suspended particles. For extraction, 1 L-samples were sucked through the preconditioned sorbent in glass columns at a flow rate of 10 mL/min. 400 mg of LiChrolut EN (Merck, Darmstadt) was used as the sorbent.

After sample extraction, the sorbent was washed with 3 mL deionized water to remove salts. Residual water was then pushed out with air by a syringe. The adsorbed compounds were immediately eluted with two 2.5 mL-portions of acetonitrile/methanol

Dedicated to Professor Dr. Karlheinz Ballschmiter on the occasion of his 60th birthday

B. Edler · C. Zwiener · F. H. Frimmel (✉)
Engler-Bunte-Institut der Universität Karlsruhe,
Lehrstuhl für Wasserchemie, Richard-Willstätter-Allee 5,
D-76131 Karlsruhe, Germany

(50:50 vol.). The combined organic extracts were brought to a final volume of 5 mL (preconcentration 1:200) and analyzed by LC/MS.

Gases and chemicals

Helium gas for the nebulizer of the particle beam interface (PBI) was of ultra-high purity grade (Helium 5.0, Messer Griesheim). Solvents (methanol, acetonitrile) were Baker HPLC grade. LC-water was prepared by ultrafiltration with a Milli-Q water purification system (Millipore, Germany).

All chemicals used for this work were of 99% purity and obtained from Aldrich (Steinheim, Germany). The following substances were used: 6(5H)-phenanthridinone (D1); 2-amino-9-fluorenone (D2); 4-amino-9-fluorenone (D3), and 9(10H)-acridinone (D4).

LC/MS-analysis

For HPLC-analysis, a binary pump (Merck-Hitachi L6200A, Darmstadt, Germany) and a UV/VIS detector (Merck-Hitachi L4000, Darmstadt, Germany) were used. A Purospher RP18 e column (length 10 cm, diameter 2.1 mm, particle size 5 μm) was purchased

from Muder & Wochele (Berlin, Germany). The following linear gradient of acetonitrile (MeCN) and LC-water was applied: at 0 min 5% MeCN; at 5 min 5% MeCN; at 25 min 98% MeCN; at 40 min 98% MeCN. The flow rate of the LC eluent was set at 0.2 mL/min and the column temperature at 30°C.

A Finnigan SSQ 710 (Single Stage Quadrupole) was coupled to the outlet of the UV-Detector via the Finnigan Particle Beam Interface.

The mass spectrometer was operated in the electron impact ionization mode (EI) with an ionizing energy of 70 eV, an emission current of 200 μA , and a multiplier voltage of 1400 V. The ion source block was set at 280°C, the manifold at 70°C.

FC 43 (perfluorotributylamine) was used for tuning procedures with the autotune software (Finnigan). The daily performance of the PBI was checked with several injections of caffeine (direct injection of 73 ng caffeine via the sample loop). The temperature of the desolvation chamber was set at 42°C. Typical operating pressures in the ion source chamber were 866.6 Pa, in the manifold 7.3×10^{-4} Pa.

Full-scan mode spectra were acquired in a mass range between m/z 60 and m/z 600 at a scan rate of 1 scan s^{-1} .

For computerized library searches, we used a NIST-library [9]. The criteria to check the plausibility of the library search results were:

- spectral fit > 90 out of 100 units
- check of the fragmentation pattern
- the chromatographic elution order on a reversed phase column
- inclusion of further information about the reliability of the occurrence of sample constituents in order to confirm the accuracy of the assigned compounds

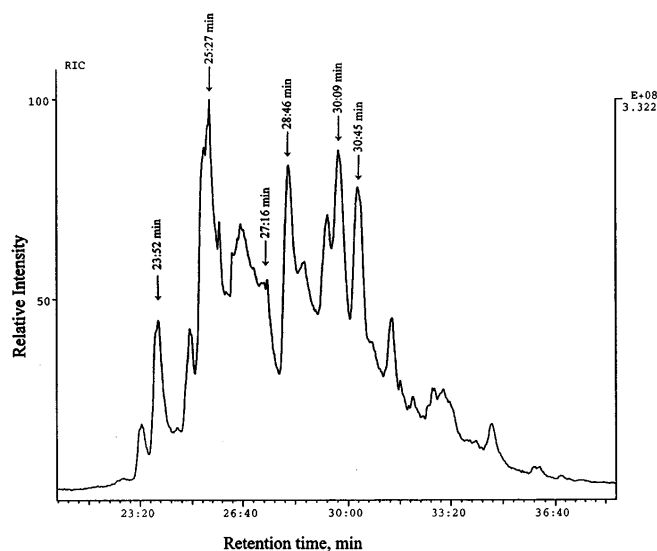


Fig. 1 Reconstructed ion current chromatogram (RIC) of the extract of a groundwater sample from the gas plant site

Results and discussion

Figure 1 shows the reconstructed ion current chromatogram (RIC) of the extract of a groundwater sample obtained by LC/EI-MS. It is remarkable that all compounds eluted in a close retention time window between 23 min and 33 min. The chromatographic separation was sufficient for reliable mass spectra evaluation of single peaks. Subtracted or underground corrected mass spectra were obtained for the peaks at the retention times (RT) 23:52, 25:27, 27:16, 28:46, 30:09, and 30:45 min (cf. Table 1). For the last two peaks, no reliable library results could be found. The other library matches were higher than 90%.

Table 1 lists the mass spectral data of the specified peaks and the corresponding library search results. Based on the results of the library search, the compounds be-

Table 1 Retention times and mass spectral data of the major peaks (cf. Fig. 1). Library search results represent the best fit and highest probability of the compound occurrence in the sample

Retention time (min)	MS data of sample peaks (m/z normalized abundance)	Library search results (fit > 90)
23:52	90 ₍₅₅₎ ; 118 ₍₇₂₎ ; 145 ₍₁₀₀₎	1(2H)-Isoquinolinone (A)
25:27	77 ₍₁₇₎ ; 103 ₍₁₅₎ ; 130 ₍₈₈₎ ; 159 ₍₁₀₀₎	2(1H)-Methylquinolinone (B)
27:16	130 ₍₁₁₎ ; 144 ₍₁₆₎ ; 158 ₍₃₆₎ ; 173 ₍₁₀₀₎	2(1H)-Dimethylquinolinone (C)
28:46	83 ₍₂₇₎ ; 99 ₍₁₅₎ ; 139 ₍₂₈₎ ; 167 ₍₆₇₎ ; 195 ₍₁₀₀₎	Positive matches for D ₁ ...D ₄ 6(5H)-Phenanthridinone (D ₁); 2-Amino-9-fluorenone (D ₂); 4-Amino-9-fluorenone (D ₃) 9(10H)-Acridinone (D ₄)
30:09	77 ₍₃₁₎ ; 103 ₍₃₉₎ ; 116 ₍₉₄₎ ; 132 ₍₁₀₀₎ ; 147 ₍₂₈₎ ; 280 ₍₁₉₎	No match
30:45	77 ₍₂₆₎ ; 91 ₍₂₆₎ ; 103 ₍₂₅₎ ; 116 ₍₁₀₀₎ ; 132 ₍₉₁₎ ; 184 ₍₂₈₎ ; 280 ₍₁₇₎	No match

Fig. 2 Chemical structures of the library search results A, B, C (cf. Table 1) – The exact positions of the methyl groups are not determined

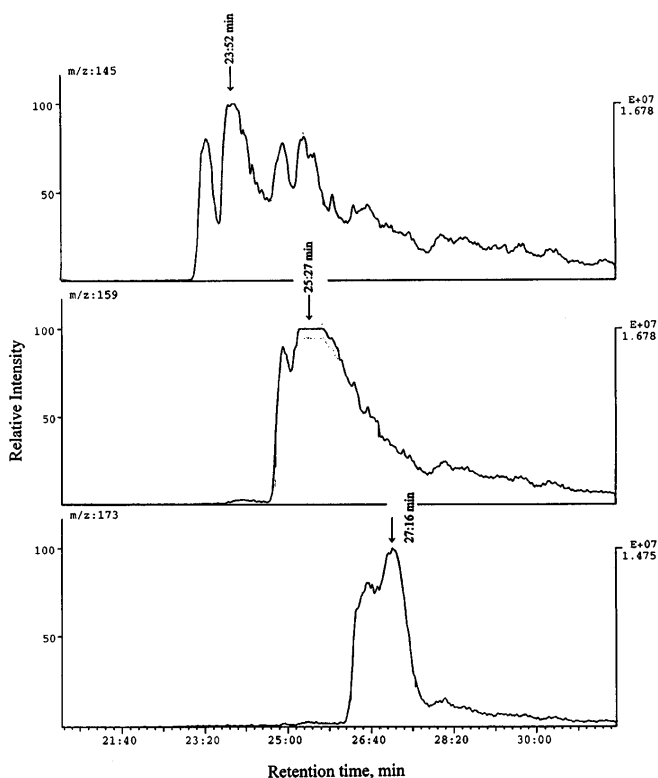
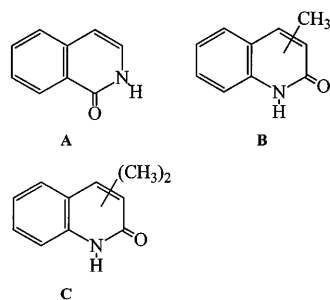


Fig. 3 Selected ion current chromatograms at specific *m/z* for quinolinone/isoquinolinone (*m/z* 145), methyl derivatives of quinolinone (*m/z* 159) and dimethyl derivatives of quinolinone (*m/z* 173)

tween RT 23 min and 28 min could be identified as derivatives of isoquinolinone, methyl-quinolinone, and dimethyl-quinolinone. The chemical structures are shown in Fig. 2. The selected ion current chromatograms (SIM) at *m/z* 145, 159 and 173 demonstrate the expected elution order of the compounds with the suggested structures (Fig. 3). The methyl groups of compounds B and C (cf.

Table 1) result in the corresponding extended retention times compared to the retention time of compound A. It should be mentioned that an unequivocal identification based only on MS is not possible in all cases.

Nevertheless, 1(2H)-isoquinolinone (compound A with the molecular formula C₉H₇NO and the relative molecular mass 145) shows a different mass spectra in comparison to other constitutional isomers of C₉H₇NO, as shown in Table 2. For instance, the mass spectral data of the suggested compound B (with the molecular formula C₁₀H₉NO and the relative molecular mass 159) and other constitutional isomers of C₁₀H₉NO are compared in Table 3. It is obvious that the position of the methyl group could not be assigned from the mass spectrum of 2(1H)-quinolinone, 3-methyl when compared to the mass spectrum of 2(1H)-quinolinone, 1-methyl.

For the peak at RT 28:46 min (cf. Table 1), four library search results with high matches (6(5H)-phenanthridinone (D₁); 2-amino-9-fluorenone (D₂); 4-amino-9-fluorenone (D₃), and 9(10H)-acridinone (D₄)) were obtained. All of them are constitutional isomers of C₁₃H₉NO. Figure 4 shows the corresponding structures and mass spectra. Figure 5 shows the mass spectrum for the peak at RT 28:46 min found in the RIC-chromatogram of the sample. Due to the similarity of the mass spectra of the compounds D₁, D₃, and D₄, a successful identification is only possible with the aid of chromatographic retention times of the corresponding standard compounds.

For this reason, the retention times of D₁, D₂, D₃, and D₄ were determined: 6(5H)-phenanthridinone (D₁; RT 28:46 min); 2-amino-9-fluorenone (D₂; RT 29:37 min); 4-amino-9-fluorenone (D₃; RT 29:07 min), and 9(10H)-acridinone (D₄; RT 26:08 min). As expected, the constitutional isomers elute in a narrow time window. Nevertheless, we could assign the retention time of 6(5H)-phenanthridinone (D₁) to the unknown compound at 28:46 min in the SIM- (*m/z* 195) and RIC-chromatogram. Verification was carried out by spiking the original sample with the standard compound at a concentration of 6 mg/L (data not shown). The area of the peak at RT 28:46 min was doubled in this way.

Further information about typical pollutants of a former gas plant could be obtained from the literature. Quinoline, isoquinoline, and phenanthridine have been identified after fractionating distillation of coal tar [1], a typical waste product of gasification or liquefaction processes [10]. Hence, the identified compounds in this work (isoquinolinone, methyl- and dimethyl-quinolinone, phenan-

Table 2 Mass spectral data of the constitutional isomers of C₉H₇NO (relative molecular mass 145). 1(2H)-isoquinolinone is the compound with the best match of the mass spectrum for the peak at 23:52 min and the library search

Compound	Normalized abundance							
	<i>m/z</i> 145	<i>m/z</i> 129	<i>m/z</i> 128	<i>m/z</i> 118	<i>m/z</i> 117	<i>m/z</i> 116	<i>m/z</i> 90	<i>m/z</i> 89
1(2H)-Isoquinolinone	100	–	–	20	8	6	23	19
5(2H)-Isoquinolinone	100	–	–	3	45	13	34	31
2(1H)-Quinolinone	100	–	–	8	70	11	30	27
5-Quinolinol	100	–	–	7	41	15	26	27
Isoquinoline, 2-oxide	100	11	7	34	7	6	54	39

Table 3 Mass spectral data of the constitutional isomers of C₁₀H₉NO (relative molecular mass 159). 2(1H)-quinolinone, 3-methyl is the compound with the best match of the mass spectrum for the peak at 25:27 min and the library search

Compound	Normalized abundance					
	m/z 159	m/z 141	m/z 131	m/z 130	m/z 117	m/z 116
2(1H)-Quinolinone, 3-methyl	100	15	31	98	9	2
2(1H)-Quinolinone, 1-methyl	100	12	47	69	15	8
2(1H)-Quinolinone, 4-methyl	75	–	24	100	–	–
1(2H)-Isoquinolinone, 2-methyl	100	7	13	20	3	21
4-Quinolinol, 1-methyl	100	–	84	53	2	12
4-Quinolinol, 2-methyl	100	–	51	72	13	4

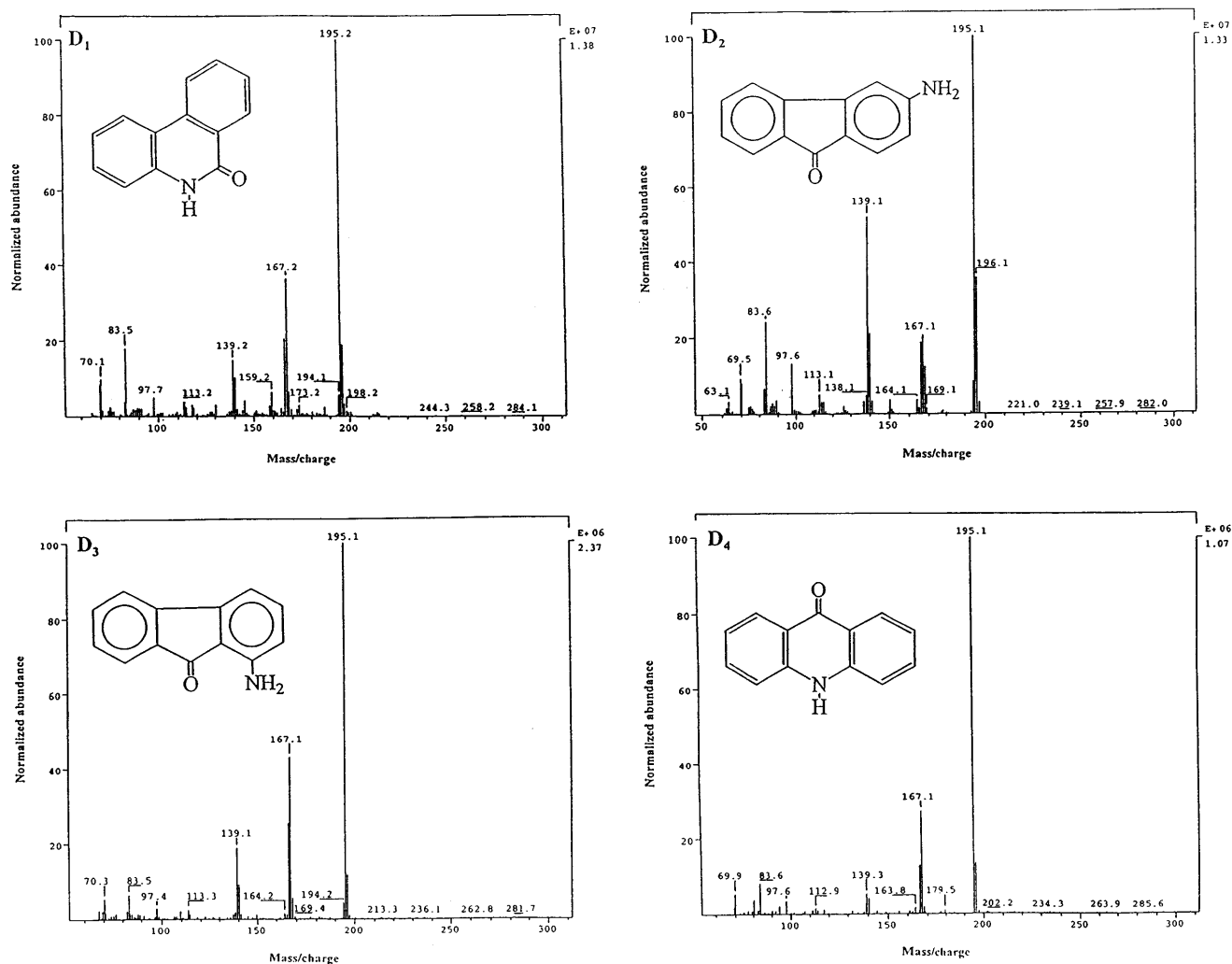


Fig. 4 Mass spectra and chemical structures of 6(5H)-phenanthridinone (D₁), 2-amino-9-fluorenone (D₂), 4-amino-9-fluorenone (D₃), and 9(10H)-acridinone (D₄)

thridinone) could be assigned to oxidation products of quinoline, isoquinoline, and phenanthridine formed during chemical or microbial conversion. For instance, similar oxidation products have been found during the microbial degradation of polynuclear aromatic hydrocarbons (PAH) [11–13].

Conclusions

Particle Beam LC/MS is well suited for a nontarget screening of highly contaminated water samples and could give some valuable hints to new pollutants. Based on the results of library searches and comparison with a pure standard, the oxidation products of typical constituents of coal tar could be identified. However, an unequivocal distinction of constitutional isomers based on their EI mass spectra is not possible in all cases and comparison with a standard is needed. The formation of the identified oxidation products could be a result of chemical or microbial degradation of the original pollutants. Fur-

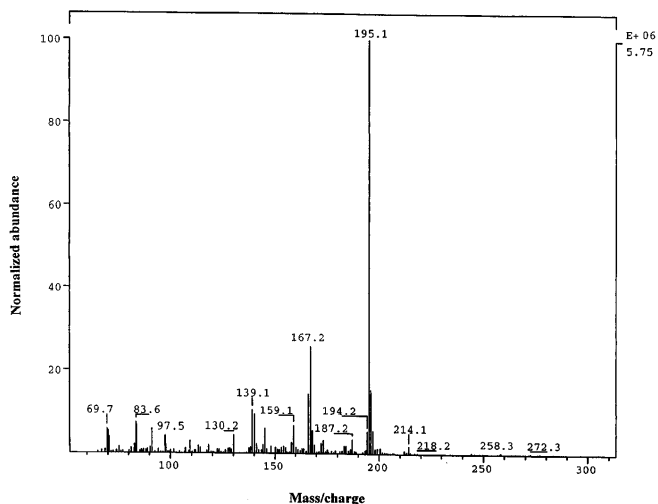


Fig. 5 Mass spectrum of the unknown compound at 28:46 min (cf. Fig. 1)

ther analyses at the hot spots of the contamination as well as along the ground water stream should give more information about origin and formation of the oxidized heterocyclic compounds.

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