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# Detectability enhancement by the use of large-volume injections in gas chromatography–cryotrapping–Fourier transform infrared spectrometry

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**Abstract**

The use of large-volume injections via a loop-type interface in GC–cryotrapping–FT-IR has been studied. *n*-Alkanes and polycyclic aromatic hydrocarbons (PAHs) were used as test compounds. When using 100-$\mu$l injections the analyte detectability was enhanced by two to three orders of magnitude compared with conventional split/splitless injections. For the alkanes, the absolute detection limits were nearly independent of the injected volume and the mode of injection. With the PAHs a water background (due to press-fit connections and six-port valves) somewhat disturbed the GC–FT-IR chromatograms and the spectra of the PAHs. This detracts from the system performance. Still, upon going from split/splitless injections to 100–200 $\mu$l injections, analyte detectability was increased by a factor of 30–500. The repeatability of the total 100-$\mu$l GC–cryotrapping–FT-IR procedure was satisfactory. The potential of the set-up was demonstrated by determining PAHs in river water at the 0.5 $\mu$g/l level by means of "micro" liquid–liquid extraction and 100-$\mu$l GC–cryotrapping–FT-IR. The FT-IR spectra obtained matched well with those of standard libraries.

*Keywords:* Large-volume injection; Cryogenic trapping; Sensitivity enhancement; Injection methods; Water analysis; Environmental analysis; Interfaces, GC–FT-IR; Alkanes; Polynuclear aromatic hydrocarbons

## 1. Introduction

The identification and quantitation of environmental pollutants becomes increasingly demanding as threshold values for microcontaminants still tend to be set lower for health and safety reasons. The current alarm level for individual pesticides in surface water and drinking water, for instance, is 1 $\mu$g/l and 0.1 $\mu$g/l, respectively [1]. Reaching such detection limits is a distinct challenge.

Gas chromatography (GC) is often used for trace analysis because of its high sensitivity, separation efficiency and speed of analysis. Detection is usually carried out by flame ionization detection (FID), the more or less element-selective nitrogen/phosphorus detection (NPD) and electron-capture detection (ECD), or by mass spectrometry (MS) if structural information is required. Unambiguous identification

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by MS, however, is not always possible, e.g. when one has to distinguish isomers. In such cases, Fourier transform infrared (FT-IR) spectrometric detection is the preferred alternative [2–5].

Coupling of GC and IR can be achieved by the "light-pipe" flow cell interface [6], and by low-temperature sample storage techniques such as matrix isolation [7] and direct deposition or cryotrapping [8]. The light-pipe is the most straightforward interface but a major drawback is the short residence time of the sample in the cell. The sensitivity is at best 10–100 ng of analyte injected on-column.

The principle of the sample storage techniques is immobilization of the GC chromatogram at cryogenic temperature, typically 4–80 K. The GC eluate is trapped as a small trace on a moving substrate. IR detection is carried out either immediately after deposition (on-the-fly) or after completion of the GC run (post-run scanning). This type of sample storage allows the use of IR microscopy and extended data acquisition of each spot on the substrate. As a consequence, the sensitivity of these techniques is two orders of magnitude better than GC–lightpipe–FT-IR resulting in detection limits of 0.1–1 ng on-column [9]. Admittedly, sample storage interfaces are more complex and require more operator involvement.

So far, sample volumes injected on sample storage GC–FT-IR systems have been limited to 1–2 $\mu$l, i.e., to $\mu$g/ml levels in terms of concentration in the extract solution. In principle, however, the analyte detectability (in concentration units) can be dramatically increased by injecting a larger volume of the sample solution. Several techniques to inject such volumes into a GC system have been developed, particularly for the on-line coupling of LC and GC. Examples are the on-column interface with partially concurrent solvent evaporation [10] and the loop-type interface using fully concurrent solvent evaporation [11]. Recently, the programmed temperature vaporizer (PTV) has been used for large-volume injection also [12]. Several applications of GC combined with these injection techniques have been reported [13–16].

Occasionally, large-volume injection has been applied to increase the detectability of GC–FT-IR, but only for instruments equipped with a light-pipe interface. Fehl and Marcott reported the development of a two-trap injection system to inject 100-$\mu$l volumes [17]. The system resembled a PTV injector with an additional cold trap between the PTV and the analytical column. Hu et al. used an on-column interface for 100-$\mu$l injections into a light-pipe GC–FT-IR instrument [18] using a 37-m long retention gap. An increase in analyte detectability by a factor of 100 was claimed but quantitative data were not shown. Full et al. demonstrated the use of an on-column interface for injections of up to 550 $\mu$l in an on-line LC–GC–lightpipe–FT-IR system [19]. Detection limits were not reported.

Obviously, further improvement of analyte detectability in GC–FT-IR can be achieved by applying large-volume injection on a more sensitive, but also more complex sample storage GC–FT-IR system. We therefore studied large-volume injection combined with GC–cryotrapping–FT-IR. A loop-type injection interface was chosen because of its rather simple optimization. The system was tested and optimized with standard solutions of alkanes and polycylic aromatic hydrocarbons (PAHs). The practicability of the system was tested by the determination of PAHs in river water extracts obtained by means of "micro" liquid–liquid extraction ($\mu$LLE).

## 2. Experimental

### 2.1. Chemicals

The standard mixture of PAHs in toluene was a Standard Reference Material (SRM 2260) from the National Institute of Standards and Technology (NIST), obtained from C.N. Schmidt (Amsterdam, Netherlands). The certified concentrations were around 60 $\mu$g/ml. The stock solution was used for the preparation of diluted standards in hexane and for spiking of river Rhine water samples.

A stock solution containing undecane ($C_{11}$), tridecane ($C_{13}$), pentadecane ($C_{15}$), heptadecane ($C_{17}$), nonadecane ($C_{19}$), uneicosane ($C_{21}$), trieicosane ($C_{23}$) and pentaeicosane ($C_{25}$) was prepared in *n*-hexane at a concentration of about 20 $\mu$g/ml.

For optimization of the large-volume injections the stock solution was diluted to a concentration of 50 pg/$\mu$l in *n*-pentane, *n*-hexane, ethyl acetate, acetonitrile and toluene. Ethyl acetate, *n*-pentane and *n*-

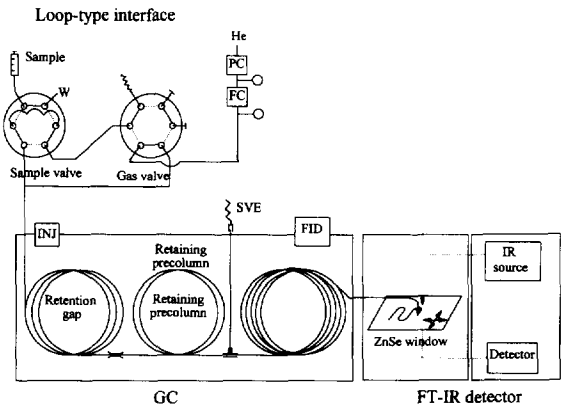


Fig. 1. Set-up of the large-volume injection–GC–cryotrapping–FT-IR system. Abbreviations: FC, flow controller; PC, pressure controller; INJ, injector; SVE, solvent vapour exit; W, waste.

hexane (all Nanograde) were from Mallinckrodt and were purchased from Promochem (Wesel, Germany). Toluene (glass distilled grade) and acetonitrile (HPLC grade) were obtained from Rathburn Chemicals (Walkerburn, UK). Anhydrous sodium sulfate was obtained from J.T. Baker (Deventer, Netherlands) and dried at 150°C before use. Helium 4.0 was purchased from Hoekloos (Schiedam, Netherlands).

### 2.2. GC equipment

A Carlo Erba MEGA 5160 gas chromatograph (Carlo Erba Strumentazione, Milan, Italy) equipped with a split/splitless injector and a flame ionization detector was used. The large-volume injection system (Fig. 1) consisted of a loop-type interface with two six-port rotating switching valves. The sample valve contained a sample syringe and a 100-$\mu$l loop.

The GC system contained a 5 m × 0.32 mm I.D. diphenyltetramethyldisilazane deactivated retention gap (BGB Analytik, Zürich, Switzerland), a 1 m × 0.25 mm I.D. CP-Sil-5 CB (Chrompack, Middelburg, Netherlands) retaining precolumn and a 25 m × 0.25 mm I.D. CP-Sil-5 CB analytical column with a film thickness of 0.25 $\mu$m. For the analysis of the real-life water samples a 1 m × 0.25 mm I.D. DB-17 (J & W Scientific, Folsom, CA, USA) retaining precolumn and a 15 m × 0.25 mm I.D. DB-17 analytical column with a film thickness of 0.15 $\mu$m were used. An early solvent vapour exit (SVE) was inserted between the retaining and the analytical column. The SVE was opened and closed manually by removing and attaching a press-fit connector to which a 0.60 m × 50 $\mu$m I.D. restriction capillary was connected (see Fig. 1). Large-volume injections of 100, 200 or 400 $\mu$l were performed by (i) filling the sample loop using a syringe, (ii) opening the solvent vapour exit and (iii) simultaneously switching the sample and gas valve so that the carrier gas pushes the sample plug via the retention gap to the GC system. After completion of the transfer, which is indicated by the pressure drop on the manometer inserted after the flow controller, both six-port valves were switched to the default position (shown in Fig. 1). The solvent vapour exit was closed after 30 s, and the GC temperature programme and the data acquisition were then started.

For the analysis of alkanes, the temperature was increased from the transfer temperature to 290°C (final hold time, 10 min) at 20°C/min. Transfer temperatures for 100-$\mu$l injections of various solvents are summarized in Table 1. The transfer temperature for the injection of 200- and 400-$\mu$l volumes were 3°C higher than for the 100-$\mu$l amounts. For the determination of the PAHs the

Table 1
Minimum transfer temperature and application range for various solvents

| Solvent | Boiling point (°C) | Optimized transfer temperature (°C) | Elution temperature of first *n*-alkane (°C) | First *n*-alkane[a] (C number) |
|---|---|---|---|---|
| *n*-Pentane | 36 | 60 | 200 | 15 |
| *n*-Hexane | 69 | 95 | 240 | 19 |
| Ethyl acetate | 77 | 100 | 240 | 19 |
| Acetonitrile | 81 | 112 | 260 | 21 |
| Toluene | 111 | 143 | 280 | 23 |

[a] First *n*-alkane of test mixture, which was quantitatively recovered.

temperature was increased to 180°C at 20°C/min, followed by an increase to 290°C at 5°C/min, with a final 10-min hold. The gas flow was set to 0.9 ml/min, resulting in a head pressure of around 80 kPa at 70°C for the 25 m and 55 kPa for the 15-m analytical column. To remove traces of water present in the helium, a water trap (Supelco, Bellefonte, PA, USA) was installed in front of the pressure and flow control of the GC system. For reference purposes, 1-$\mu$l split and splitless injections were carried out. The injector temperature then was 250°C and the head pressure 90 kPa.

### 2.3. FT-IR spectrometry

The infrared spectrometer used was a Digilab, FTS-40 Fourier transform instrument (Bio-Rad, Cambridge, MA, USA) equipped with a Digilab Tracer cryotrapping GC interface [2]. Data acquisition and processing was done with a SPC 3200 computer (Bio-Rad). The GC column was connected to a 1-m deactivated fused-silica transfer capillary of 150 $\mu$m I.D. by means of an aluminum ferrule connector (Bio-Rad). The transfer line was guided into the interface housing through a stainless-steel pipe. The interface housing was held at $1.3 \cdot 10^{-3}$ Pa to minimize condensation of compounds present in the ambient air. A fused-silica deposition tip of 60 $\mu$m I.D. was fixed to the end of the transfer line and located 30 $\mu$m above the surface of a moving IR transparent ZnSe window, which was cooled to 80 K with liquid nitrogen. The tip and the transfer line were kept at 250°C.

Eluting compounds were trapped as solids on the ZnSe slide, which was moved continuously by an X–Y stepping motor. A few seconds after deposition the trapped spots passed through the beam of an IR microscope. The immobilized chromatogram was thus scanned on-the-fly by averaging four spectra recorded at 2 scans/s for each step. After completion of the GC run, extended post-run scanning of certain retention times was performed by repositioning the corresponding coordinates of the ZnSe window into the IR beam. Post-run spectra were acquired by averaging 256 scans. The optical resolution of on-the-fly and post-run spectra was 8 $cm^{-1}$

GC–FT-IR chromatograms of the integrated IR absorption as a function of retention time were constructed by standard Gram-Schmidt vector orthogonalization. Functional-group GC–FT-IR chromatograms of preselected wavelength regions were applied to reduce interferences of compounds not showing absorption in that wavelength region [3]. The interval 2820–2980 $cm^{-1}$ was chosen for the alkanes, and 700–950 $cm^{-1}$ for the PAHs, as the strongest absorption bands of these compounds occur in these regions.

### 2.4. Liquid–liquid extraction

A 1-l water sample held in a 1-l Erlenmeyer flask was extracted twice with 0.6 ml of *n*-hexane. After shaking for 5 min, a laboratory-made micro-extraction adapter modified slightly from Ref. [20] was placed on top of the 1-l flask via an NS 24 connection. The flask was tilted and water added via the wider glass tubing so that the hexane layer was pushed into the glass capillary, and could then be removed with a pipette. After removing about 30 ml of water from the Erlenmeyer flask, another 0.6 ml of *n*-hexane was added and the extraction process repeated. A total of about 1 ml of hexane was recovered. The extract was dried over anhydrous sodium sulfate.

## 3. Results and discussion

### 3.1. Optimization of large-volume injections

Large-volume injections via a loop-type interface require optimization of two basic parameters: (i) the transfer temperature and (ii) the closing time of the early solvent vapour exit necessary to minimize the amount of solvent reaching the FT-IR detector.

#### 3.1.1. Optimization of transfer temperature for various solvents

When injecting large volumes of solvent into the GC system via a loop-type interface the solvent is evaporated under fully concurrent evaporation conditions. The only parameter to be optimized is the transfer temperature, which has to be above the boiling point of the solvent. At too low a transfer temperature the solvent evaporation is too slow and the solvent film will reach the retaining precolumn;

this will result in severe peak distortion. If the transfer temperature is too high, the more volatile analytes will be (partly) lost. The minimal transfer temperatures to avoid flooding the retaining precolumn for various solvents were optimized with a 50 pg/$\mu$l solution of the uneven ($C_{11}$–$C_{25}$) *n*-alkanes in *n*-pentane, *n*-hexane, ethyl acetate, acetonitrile and toluene. For reasons of simplicity, FID was used for optimization. The solvent vapour exit was closed 30 s after completion of the transfer which was indicated by the pressure drop as described above. The first *n*-alkane which was quantitatively recovered with the various solvents is listed in Table 1. The difference between the boiling point and the optimized transfer temperature is around 25°C for *n*-pentane, *n*-hexane and ethyl acetate, and around 31°C for acetonitrile and toluene. The difference between the transfer temperature and the elution temperature of the first alkane is about 140°C. *n*-Hexane was used as solvent in all further work for reasons of practicability. That is, $C_{19}$ can be determined without, and $C_{17}$ with some loss.

### *3.1.2. Optimization of closure of early solvent vapour exit*

Obviously, 1-$\mu$l splitless injections onto the GC system will cause a vapour cloud at the end of the deposition tip that may be partly spread over the deposition window of the FT-IR detector. It is known from previous GC–cryotrapping–FT-IR experiments that part of this solvent cloud may crystallize on the window, despite the high vacuum [3]. As a consequence, the chromatogram as well as the IR spectra may be obscured by solvent absorption. Similar problems can be expected when applying large-volume injections. In this set-up solvent vapour can reach the FT-IR detector in two ways:

1. during solvent transfer, via the T splitter; most of the vapour is led to waste through the solvent vapour exit, but a small part is directed to the FT-IR interface as a result of the vacuum at the end of the transfer line.
2. if the solvent vapour exit is closed too early; solvent that is left in the retention gap and retaining precolumn will reach the FT-IR detector.

In principle, the amount of solvent reaching the FT-IR detector during analyte transfer can be decreased by reducing the transfer time, i.e. by increasing the transfer temperature. However, this option was discarded, since it would result in a loss of more volatile compounds.

The optimal time delay between completion of the transfer and closure of the solvent vapour exit was established for 100-$\mu$l injections of the uneven *n*-alkanes in *n*-hexane. FID was used for this study, since the amount of solvent transferred cannot be monitored by the FT-IR detector. The solvent peak width was recorded as a function of the time delay to provide a measure of the amount of solvent transferred to the detector. The solvent peak width did not decrease any further when closing the solvent vapour exit later than 30 s after completion of the transfer. With a time delay longer than 30 s, loss of alkanes, e.g. $C_{17}$ and $C_{19}$ was observed. As no significant difference in the recovery of $C_{17}$ was observed when closing the solvent vapour exit either immediately after completion of the transfer or 30 s later, the solvent vapour exit was always closed with a time delay of 30 s.

## *3.2. Performance of large-volume injection–GC–cryotrapping–FT-IR*

The performance of large-volume injection–GC–cryotrapping–FT-IR in terms of analyte detectability and repeatability was studied for injection volumes of 100–400 $\mu$l. The *n*-alkanes and PAHs were used as test compounds.

### *3.2.1. Repeatability*

The repeatability was tested for 100-$\mu$l injections of an *n*-alkane standard solution in hexane at the 50 pg/$\mu$l level. Quantitation with GC–FT-IR can be accomplished by using the peak height of (1) the Gram–Schmidt chromatogram, (2) the functional group GC–FT-IR chromatogram and (3) the strongest band in the FT-IR spectra. The results for the latter two methods are shown in Table 2. The relative standard deviation (R.S.D.) was 15% or less ($n$=6) for all *n*-alkanes when using the functional-group chromatogram. Significantly higher R.S.D. values were obtained for $C_{17}$ and $C_{19}$ when using the strongest band in the spectrum. This effect can be attributed to variations in the background and back-

Table 2
Repeatability of 100-$\mu$l injections ($n=6$) of $n$-alkanes (50 pg/$\mu$l) in GC–cryotrapping–FT-IR

| Alkane | R.S.D. (%) for response obtained [a] | |
|---|---|---|
| | Functional group chromatogram | Highest peak in IR spectra |
| $C_{17}$ | 15 | 27 |
| $C_{19}$ | 13 | 29 |
| $C_{21}$ | 11 | 15 |
| $C_{23}$ | 9 | 15 |
| $C_{25}$ | 8 | 15 |

[a] Response obtained from functional-group GC–FT-IR (2820–2980 cm$^{-1}$) chromatogram and highest peak in IR spectra (2820–2980 cm$^{-1}$ range).

ground subtraction, and to a somewhat larger discrimination during the transfer.

### 3.2.2. Detection limit

The absolute detection limit of a GC system is determined by the amount of analyte passing through the detector, the sensitivity and noise of the detector and the peak shape of the analyte. In the ideal situation all material injected will reach the detector. Consequently, the detectability of an analyte in an extract can be enhanced with a factor that is directly proportional to the injected volume. It follows that the limit of detection of a total procedure, i.e. sample preparation and separation-cum-detection, can be dramatically improved by injecting a larger portion of the extract. In order to study the analyte detectability with large-volume injection–GC–cyrotrapping–FT-IR, injections of 100-, 200- and 400-$\mu$l amounts were compared with that of 1-$\mu$l (split 1:10 and splitless).

Using GC–FID, the theoretically expected increase in analyte detectability in concentration units was indeed observed, or, in other words, the absolute detection limits were independent of the injection volume and the injection method used (Table 3). It should be noted, that the solvent must be very pure to prevent reduced performance as a result of interfering solvent contaminants in cases of large-volume injection.

The situation was marginally less good when using the functional-group GC–FT-IR chromatogram. No problems were observed up to, and including, 100-$\mu$l injections, even though some baseline distortion due to solvent contamination was observed for the 100-$\mu$l injections (Fig. 2). A noticeable loss of performance, and, consequently, a modest increase of the absolute detection limits (from 250 to 300 pg) was found with the 200- and 400-$\mu$l injections. Actually, during the transfer, the formation of a distinct solvent spot on the deposition window was observed (cf. Section 3.1.2.). As the position of the deposition tip can not be changed during the transfer, 100-$\mu$l injections were considered to be most appropriate for routine work. It

Table 3
Detection limits expressed in concentration units (pg/$\mu$l) ($S/N=3$) of alkanes and PAHs in GC–cryotrapping–FT-IR and GC–FID using split, splitless and large-volume injection

| Analytes | Detector | Detection limit [pg/$\mu$l or (pg)] | | | | |
|---|---|---|---|---|---|---|
| | | Split | Splitless | Large-volume injection | | |
| | | 1 $\mu$l (1:10) | 1 $\mu$l | 100 $\mu$l | 200 $\mu$l | 400 $\mu$l |
| $n$-Alkanes | FID | 200 | 20 | 0.2 | 0.1 | 0.05 |
| | | (20) | (20) | (20) | (20) | (20) |
| | FT-IR | 2500 | 250 | 2.5 | 1.5 | 0.75 |
| | | (250) | (250) | (250) | (300) | (300) |
| PAHs | FID | 700 | 70 | 1 | 0.5 | |
| | | (70) | (70) | (100) | (100) | |
| | FT-IR | 30 000 | 3000 | 90 | 60 | |
| | | (3000) | (3000) | (9000) | (12 000) | |

The numbers given in brackets are absolute detection limits (pg).
Detection limits were obtained from GC–FID or functional-group GC–FT-IR chromatogram (700–950 cm$^{-1}$ for PAHs and 2820–2980 cm$^{-1}$ for alkanes). Detection limits are average values for all alkanes, and for the PAHs from fluoranthene to benzo[$k$]fluoranthene (peaks 4–9, see Fig. 3).

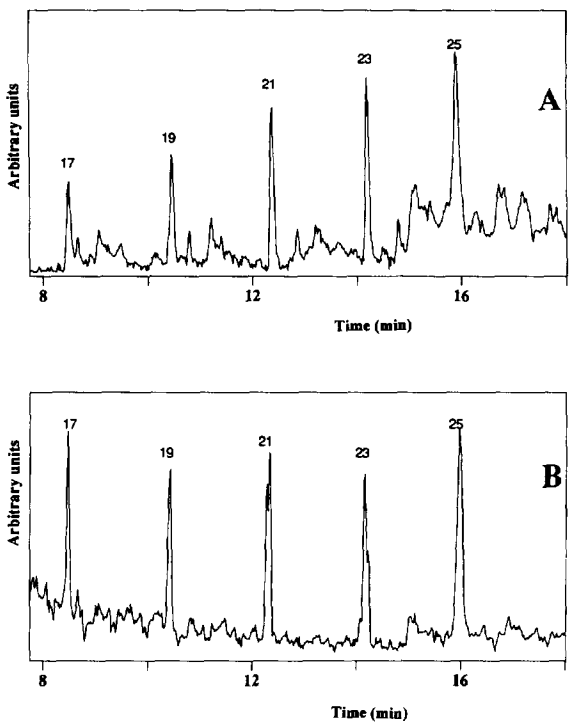


Fig. 2. Functional-group GC–FT-IR chromatogram (2820–2980 $cm^{-1}$) of uneven *n*-alkanes. (A) 100-$\mu$l injection of a 10 pg/$\mu$l per compound standard solution in hexane, (B) 1-$\mu$l splitless injection of a 1000 pg/$\mu$l standard solution in hexane.

should be noted that use of the cryotrapping interface might result in some loss of chromatographic resolution. To mention an example, the peak width at half height of nonadecane obtained with a 1-$\mu$l splitless injection of a 5 ng/$\mu$l standard solution was 2 s with GC–FID and 5 s with GC–cryotrapping–FT-IR.

As the data included in Table 3 show, somewhat larger problems were encountered with the PAHs. Still, the overall picture is highly encouraging. With GC–FID, the gain in analyte detectability expressed in concentration units effected by going from split to 200-$\mu$l injections was 1500-fold rather than the theoretical 2000-fold, or, in other words, the absolute detection limits increased some 50%. The reduced performance was attributed to somewhat broader peaks with large-volume injections than with split injection. After transfer the analytes are spread over the retention gap prior to refocusing by the phase-ratio effect [21]. If the retention gap displays some retention towards the PAHs due to the presence of active places on the inner wall or the surface of the deactivation layer, peaks will be somewhat broadened already at the start of the separation in the analytical column.

As was to be expected, detection of PAHs by means of GC–FT-IR was less straightforward than that of the *n*-alkanes. A larger wavenumber interval (700–950 $cm^{-1}$) had to be taken for the functional-group chromatogram while the detector noise in this region is relatively high. Besides, the extinction coefficients of the aromatic C–H out-of-plane absorption bands are smaller. Additionally, it should be noted that at the time of this study the obtained absolute detection limits for split/splitless injections of PAHs were somewhat worse than those reported earlier [3]. This has to be attributed to a decrease in performance of the FT-IR detector.

As regards GC–FT-IR of the PAHs, the justified concern about the ca. four-fold loss in absolute analyte detectability is distinctly outweighed by the impressive 500-fold gain effected by using a 200-$\mu$l rather than a split injection (Table 3). As regards the somewhat deteriorated performance, next to some decrease in retention gap performance, the higher water background in the large-volume injection system is a plausible explanation. As reported earlier, the performance of GC–cryotrapping–FT-IR is very sensitive to traces of water and this effect is particularly apparent if the analytes absorb in the same wavelength region as ice [3,22]. This is the case in the PAH-specific region of 700–950 $cm^{-1}$. In the present set-up, the press-fit connections and the six-port valves appeared to be the most critical items.

It should be added that the detection limits reported for the PAHs in Table 3 were calculated on the basis of the data for fluoranthene to benzo[*k*]fluoranthene (peaks 4–9 in Fig. 3). The detection limits for benzo[*e*]pyrene, benzo[*a*]pyrene and perylene are about 2–3-fold higher than for these PAHs irrespective of the injection mode used. This is due to some peak broadening at high retention times and the higher background caused by column bleeding. Consequently, the gain in analyte detectability achieved upon going from splitless to large-volume injection was the same as for the earlier eluting compounds.

The detection limits of most PAHs were found to

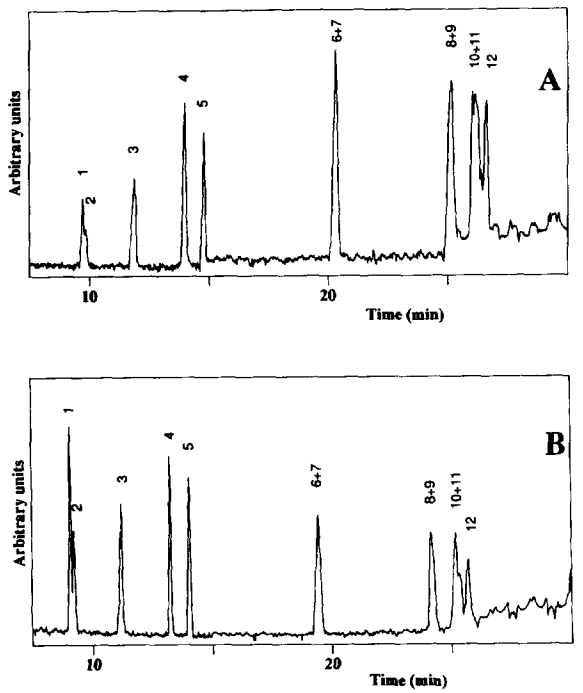


Fig. 3. Functional-group GC–FT-IR chromatogram (700–950 $cm^{-1}$) of PAHs. (A) 100-$\mu$l injection of a 600 pg/$\mu$l per compound standard solution in hexane; (B) 1-$\mu$l splitless injection of a 60 000 pg/$\mu$l standard solution. Peak assignment: 1=phenanthrene; 2=anthracene; 3=1-methylphenanthrene; 4=fluoranthene; 5=pyrene; 6=benz[*a*]anthracene; 7=chrysene; 8=benzo[*b*]fluoranthene; 9=benzo[*k*]fluoranthene; 10=benzo[*e*]pyrene; 11=benzo[*a*]pyrene; 12=perylene. Phenanthrene, anthracene and 1-methylphenanthrene were partly lost with the 100-$\mu$l injection (A).

be around ten-fold better when quantifying by means of a peak in the IR spectrum rather than by the peak in the functional group chromatogram. To quote an example, the detection limit of fluoranthene for a 100-$\mu$l injection was 10 pg/$\mu$l for the strongest peak in the spectrum (Fig. 4), while it was 100 pg/$\mu$l when using the functional-group chromatogram (Fig. 3A).

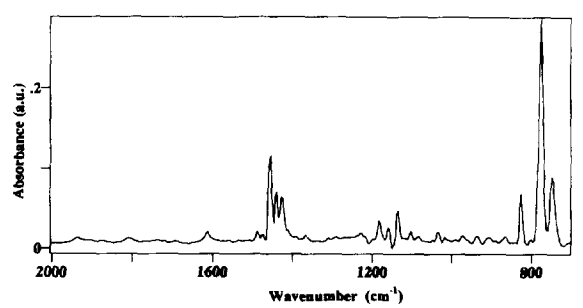


Fig. 4. GC–cryotrapping–FT-IR spectrum of fluoranthene obtained with a 100-$\mu$l injection at the 600 pg/$\mu$l level.

### 3.3. *Off line* $\mu$LLE/GC–cryotrapping–FT-IR

The practical usefulness of GC–cryotrapping–FT-IR with 100-$\mu$l injections was demonstrated by the offline combination with "micro" liquid–liquid extraction ($\mu$LLE). A 1-l sample of river Rhine water, sampled at Lobith, Netherlands, September 5, 1994, and spiked with PAHs at the 0.5 $\mu$g/l level, was extracted twice with 0.6 ml *n*-hexane which resulted in 1 ml of extract. The sample preparation was simple and rapid, since it only involved "micro" liquid–liquid extraction with no further need of evaporation of part of the solvent.

All PAHs showed up in the functional-group GC–FT-IR chromatogram (700–950 $cm^{-1}$) (Fig. 5A) with recoveries between 80 and 90% when using the absorbance of the strongest peak in the spectrum. Characteristic FT-IR spectra of all PAHs were obtained, as is illustrated for the isomers benzo[*a*]pyrene and benzo[*e*]pyrene in Fig. 6. Although these compounds were not completely separated in the functional-group GC–FT-IR chromatogram (Fig. 5A), interference-free FT-IR spectra were obtained from the wings of the GC peaks (Fig. 6A and C). The significant differences between the FT-IR spectra of both isomers clearly demonstrate the potential of GC–cryotrapping FT-IR in the identification of

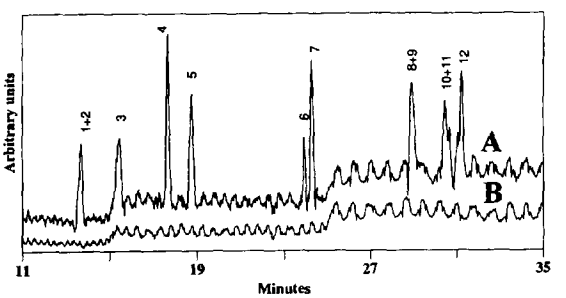


Fig. 5. Functional-group GC–FT-IR chromatogram (700–950 $cm^{-1}$) obtained after injection of a 100-$\mu$l aliquot (10%) of a hexane extract obtained with $\mu$LLE of 1 l of river Rhine water (A) spiked at the 0.5 $\mu$g/l level and (B) non-spiked. For peak assignment, see Fig. 3.

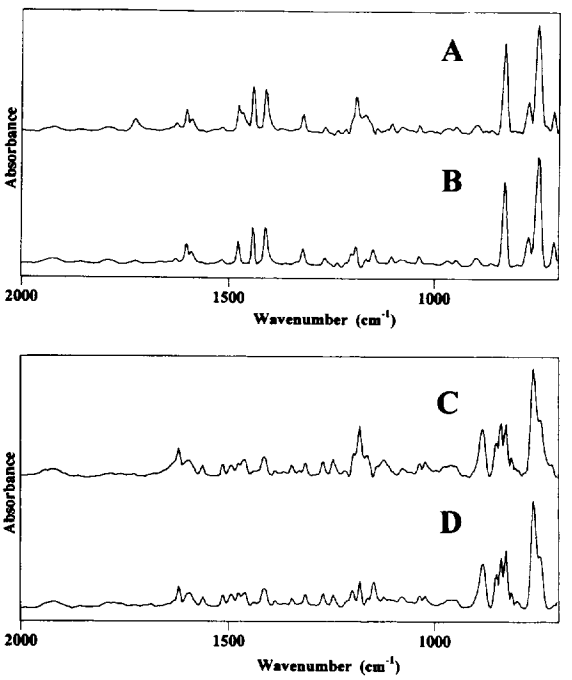


Fig. 6. FT-IR spectra of benz[*e*]pyrene (A) and benz[*a*]pyrene (C) from the spiked river Rhine water extract (Fig. 5A, peaks 10 and 11, respectively). For comparison, the library spectra are added (B, D).

molecules with closely related structures. Besides, the acquired GC–FT-IR spectra could easily be identified using a library with reference cryotrapped spectra (Fig. 6B and D). The GC–cryotrapping–FT-IR spectra were also found to closely resemble reference spectra obtained with the conventional KBr pelleting technique. This endorses earlier conclusions on the usefulness of standard libraries for the identification of cryotrapped spectra [3,22].

Detection limits for the PAHs in the river water sample were determined at 0.1–0.25 $\mu$g/l for fluoranthene, pyrene, benz[*a*]anthracene and chrysene (PAH peaks 4–7 in Fig. 3), and around 0.4 $\mu$g/l for benzo[*b*]fluoranthene, benzo[*k*]fluoranthene, benzo[*e*]pyrene, benzo[*a*]pyrene and perylene (PAH peaks 8–12 in Fig. 3), despite the fact that the baseline of the functional-group GC–FT-IR chromatogram showed some distortion due to a water background (Fig. 5). Detection limits were again about ten-fold better when using spectra instead of functional-group chromatograms. PAHs were not detected in the unspiked river water; neither in the functional-group chromatogram (Fig. 5A) nor in the spectra acquired by extended post-run scanning at the appropriate retention times. This example demonstrates that it is possible to detect microcontaminants with GC–cryotrapping–FT-IR not only below the alert level of 1 $\mu$g/l for river water but even down to the maximum allowed level of 0.1 $\mu$g/l in drinking water.

## 4. Conclusions

Large-volume injection by means of a loop-type interface can be carried out successfully in conjunction with GC–cryotrapping–FT-IR. The hyphenation permits enhanced detectability of analytes by about two orders of magnitude compared to conventional split/splitless GC–cryotrapping–FT-IR. Injected volumes of 100 $\mu$l were proved to be most appropriate. Injection of larger amounts causes interfering solvent crystallization, inherent to the principle of trapping the GC eluate at cryogenic temperatures. More effective solvent elimination techniques should reduce this drawback and are currently being studied.

Optimization of the experimental set-up is relatively simple but the system is sensitive to traces of water. Small leaks turned out to be highly detrimental in this respect, press-fit connections and six-port valves being the most critical parts.

For *n*-alkanes concentration levels of 2.5 pg/$\mu$l can be determined when using 100-$\mu$l injections and on-the-fly detection. The increase in analyte detectability for the PAHs was smaller than for the alkanes, due to reduced instrument performance and ice interference. Still, 30–500-fold improvements for 100–200-$\mu$l injections compared with 1-$\mu$l split (1:10) and splitless injections were readily achieved.

The repeatability for 100-$\mu$l injections was satisfactory with R.S.D. values of 10–20% ($n$=6) at the 50 pg/$\mu$l level.

The applicability of large-volume injection GC–cryotrapping–FT-IR to real-life analysis is promising. As demonstrated, the determination and identification of PAHs in river water is possible down to a level of 0.5 $\mu$g/l, even when using simple "micro" liquid–liquid extraction as a sample preparation technique. The FT-IR spectra obtained match well

with those of standard libraries, which facilitates identification and unambiguous discrimination of isomers. The present system may therefore be considered a viable approach to trace-level environmental analysis. Current research is directed at setting up a fully on-line SPE–GC–cryotrapping–FT-IR system and at applying the system to a number of real-life studies which should convincingly demonstrate this claim.

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