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GAS CHROMATOGRAPHIC–MASS SPECTROMETRIC DETERMINATION OF BISPHENOL-A IN WATER AT NANOGRAM PER LITER CONCENTRATIONS

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A GC-MS method for the determination of bisphenol-A (BPA) in water has been developed and validated for a concentration range of 5.0–514.8 ng/L. In contrast to hitherto published methods we report validation data in detail for the entire applicable concentration range of the developed method. The method employs liquid/liquid extraction for analyte and internal standard isolation and concentration. The D_8-BPA analog is used as surrogate standard to increase method accuracy and precision. The identity confirmation of BPA is performed using the ion intensity ratios of the molecular ion and the M-15 fragment. Trifluoroacetylation of BPA and D_8-BPA is employed in order to obtain less polar derivatives. As a result of the derivatization narrow and symmetrical gas chromatographic peaks are obtained which allow the quantification of very small amounts of BPA. The relative recoveries of BPA are greater than ninety percent over the entire validated concentration range. The method quantification limit is 5.0 ng/L BPA, which is the lowest validated concentration. The impact of suspended particulate matter frequently present in surface water samples on the quantification results could be demonstrated. It was shown that false positive results can be obtained due to losses of the surrogate standard by adsorption on the particulates present.

Keywords: Bisphenol-A; Mass Spectrometry; Determination; Internal Standard

INTRODUCTION

Bisphenol-A (BPA, 2,2-*bis*-(4-hydroxyphenyl)propane, CAS registry no. 80-05-7) is used in many industrial applications like epoxy resins and polycarbonates. The total worldwide production capacity of BPA for the year 1994 was 1.76 MM metric tons[1]. A release of 800 kg/a of BPA from industrial waste water treatment plants in Germany into the hydrosphere has been reported in 1995[1]. Recently the German Federal Environmental Agency (Umweltbundesamt, UBA) reported monitoring data for BPA in several matrices and in particular in surface waters[2]. These data show that surface waters contain BPA concentrations below 50 ng/L in most cases.

Hitherto published analytical methods for the determination of BPA in water at ng/L concentrations generally do not mention validation data to an extent which allows an

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accurate assessment of the reported data applying these methods[3–8]. The lack of validation data in published methods prompted us to develop and validate a method for BPA quantification at ng/L concentrations and to report the validation data in detail.

The direct determination of BPA at ng/L concentrations in aqueous samples, after appropriate extraction and enrichment, by gas chromatographic and gas chromatographic mass spectrometric methods is strongly hindered by the polar nature of this compound. As a result of this inherent property of BPA poor gas chromatographic peak shapes are frequently encountered in the respective chromatograms leading to a decreased or even complete loss of sensitivity. In particular samples containing traces of polar involatile impurities show the effect of a fast degradation of the gas chromatographic system. The removal of such impurities by sample clean-up steps is often laborious and can be even counter productive due to losses of the analyte during these steps. The recently described validated analytical method for BPA determination in river water at 1 μg/L concentrations could not be applied to ng/L concentrations due to the described problems[9].

The problems related to the direct analysis of BPA can be overcome by derivatizing the phenolic hydroxyl groups in order to yield less polar derivatives. The reported thermal degradation of BPA analogs, which generates free BPA, would require a derivatization at ambient temperature[9]. Silylation, in particular trimethyl silylation, and acetylation have been used as well as the acylation with perfluoro acyl groups[4, 6, 7, 8, 10, 11]. The perfluoro acyl derivatives allow sensitive GC-ECD detection as well as the application of positive EI and negative CI mass spectrometric methods. We describe in this paper the development and validation of a BPA GC-MS quantification method in water using the *bis*-trifluoracetylated derivative. The method was validated over the concentration range of 5–515 ng/L BPA. The impact of adsorption phenomena on the quantitative BPA results was also investigated with surface water and water from a biological treatment plant containing sediment and suspended biological material.

EXPERIMENTAL

Reagents and materials

Analytical grade dichloromethane was obtained from Merck, Darmstadt, Germany. Ring-D_8 Bisphenol-A (D_8-BPA) was purchased from Promochem GmbH, Postfach 10 09 55, D-46469 Wesel, Germany. Bisphenol-A and polycarbonate were obtained from Dow Deutschland Inc., Werk Stade. Gas chromatography grade trifluoroacetic anhydride was obtained from Merck, Darmstadt, Germany. 1,2,4-Tribromobenzene (97%) was obtained from Sigma-Aldrich, D-82039, Deisenhofen, Germany.

Glassware and filtration sevice cleaning

All glassware and the filter device used during the sample preparation were thoroughly cleaned by several washing steps prior to their use. The cleaning steps comprised washing with 1 N sodium hydroxide solution and subsequent rinse by deionized water, washing with a laboratory detergent (Mucasol, obtained from Merz, Frankfurt, Germany) and rinse by deionized water and a final wash step with dichloromethane.

Sample preparation

River water samples and fortified water samples were prepared as follows. Samples containing particulate matter were filtered through a 0.45 µm cellulose acetate filter prior to the extraction using an all Teflon^{TM} pressure filter device (Schleicher and Schuell, model MD 142/7/3, P.O. Box 4, D-37582 Dassel, Germany). The filtrates were acidified (pH = 1) with 4N hydrochloric acid and 15 g of sodium chloride were added. Five microliters of a surrogate standard solution of 21.8 ng/µL D_8-BPA in methanol were added to 500 mL of the filtrates. 500 mL of the ring D_8-BPA fortified samples were subsequently extracted with three 15 mL portions of dichloromethane in 1 L separation funnels for 15 min on a mechanical shaker at 175 strokes/min. The combined extracts were dried over anhydrous sodium, sulfate, concentrated to a volume of *ca.* 2 ml using a rotational evaporator at 25–30°C/100 mPa and then evaporated to dryness in a gentle stream of nitrogen. The residue was dissolved in 50 µL of dichloromethane and derivatized with 150 µL of trifluoro acetic anhydride at ambient temperature with gentle shaking for 30 min. The excess reagent was removed in a gentle stream of nitrogen, the residue was re-constituted in 300 µL of dichloromethane and 10 µL of the recovery internal standard 1,2,4-tribromobenzene in dichloromethane (35.8 ng/µL) was added. The sample was now ready for GC-MS quantification.

Samples A and B of Table III were prepared as described above with the exception that an aliquot of Sample B was diluted with water by a factor of one hundred.

GC-MS conditions

The samples were analyzed on a 5971 Hewlett-Packard MSD equipped with a 5890 Series II gas chromatograph and a 7673 autosampler. The separation was achieved on a Chrompack CP SIL 8 CB, 30 m, 0.25 mm inner diameter, 0.25 µm phase layer thickness capillary column operated with a helium head pressure of 0.7 bar. The gas chromatograph was programmed with 80°C (2 min)–10°C/min–285°C (10 min). The injector temperature was set to 285°C and the interface temperature was held at 295°C. A 1.0 µl aliquot of the samples was injected in the splitless mode for 30 s. The MSD was operated in the SIM mode and the following ions were monitored with dwell times of 100 ms: BPA: target $m/z = 405.0$, qualifier $m/z = 420.0$; D_8-BPA: target $m/z = 413.0$, qualifier $m/z = 428.0$; 1,2,4-tribromobenzene: target $m/z = 314.0$, qualifier $m/z = 316.0$. Full scan spectra of the *bis*-tifluoro acetates of BPA and D_8-BPA derivatives were acquired with the Hewlett-Packard MSD in the scan mode of appropriately concentrated samples with the above operating conditions.

Matrix calibration standards

Matrix calibration standards were prepared by fortifying 500 mL of deionized water with 3.9, 7.9, 19.8, 27.7, 39.6, 59.4, 79.2, 99.0, 237.6, 316.8, 396.0, 475.2, 594.0 ng/L of BPA (1–150 µL of a stock solution in methanol) and 218.0 ng/L D_8-BPA (5 µL of a stock solution in methanol). The standards were processed as described above. A calibration curve was established by plotting the peak area ratios of $m/z = 405$ and 413 *vs.* the BPA concentrations. All data were corrected for isotopic crossover of BPA and D_8-BPA according to the method of Barbalas and Garland[12].

Method validation samples

Method validation samples were prepared by fortifying 500 mL of deionized water with 5.0, 19.8, 51.5, 99.0, 257.4 and 514.8 ng/L BPA (10–260 µL of BPA stock solutions in methanol of 0.25 ng/µL and 1.98 ng/µL). They were processed and analyzed as described above.

BPA isotope crossover

500 mL of deionized water samples were fortified with 198.0 ng/L BPA or 218.0 ng/L D_8-BPA (5 µL of BPA or D_8-BPA stock solutions in methanol), processed and analyzed as described above. The contributions of BPA to $m/z = 413$ and D_8-BPA to $m/z = 405$ were determined.

Fortified river water samples

Three aliquots of River Elbe water were processed and analyzed as described above in order to determine the BPA background concentration. Aliquots of the filtered river water sample were then fortified in duplicate with 55.4, 118.8, and 236.6 ng/L of BPA. The samples were processed and analyzed as described and the recovery of the fortified BPA determined.

Polycarbonate fortified water sample

500 mL samples of deionized water were fortified with 1 µg/L of polycarbonate (5 µL of a polycarbonate stock solution in dichloromethane). The samples were processed and analyzed as described above.

RESULTS AND DISCUSSION

Method development

The choice for the BPA *bis*-trifluoroacetyl derivative (*bis*-TFA-BPA) is based on several factors. Trifluoroacetyl (TFA) groups can be easily and conveniently introduced using the very reactive trifluoro acetanhydride. The trifluoro acetylation reaction proceeds fast and is quantitative at room temperature after thirty minutes when using an appropriate excess of reagent. Due to its volatility the excess reagent can be easily removed in a gentle stream of nitrogen prior to the gas chromatographic–mass spectrometric analysis. The generation of false positives by thermal degradation of BPA analogs to BPA during splitless injection has been described in the literature[9]. However with this method a thermal degradation of BPA analogs to BPA during the employed splitless injection technique would not lead to false positives since the derivatizing agent is completely removed prior to the GC-MS analysis and consequently the formation of the *bis*-TFA-BPA derivative is not possible anymore. The generation of false positives during the derivatization step was checked by fortifying deionozed water with 1 µg/L of polycarbonate. Polycarbonate was chosen as BPA analog because it represents, as far as quantity is concerned, with 1.4 million metric tons in 1996 the largest portion of BPA containing products worldwide[13]. No BPA was observed in the polycarbonate fortified samples.

The quantitative method employs dichloromethane extraction and enrichment of BPA along with the D_8-BPA surrogate standard. BPA and its deuterated analog are determined after derivatization by positive EI GC-MS in the single ion monitoring (SIM) mode of operation. The full scan spectra of BPA and D_8-BPA are shown in Figs. 1 and 2. Quantification was performed using the fragments $m/z = 405$ for BPA and $m/z = 413$ for D_8-BPA respectively. Component identity confirmation was performed with the ion intensity ratios of the mentioned fragments and the respective molecular ions of the components i.e. 405/420 for BPA and 413/428 for D_8-BPA. Ion chromatograms of $m/z = 405$ and 420 reflecting the signal to noise ratios (S/N) of 5 ng/L BPA in deionized water are shown in Fig. 3.

The quantification of BPA was performed using matrix standard calibration. Deionized water was fortified with known BPA concentrations over the concentration range of 3.9–594.0 ng/L and a constant concentration of 218.0 ng/L of D_8-BPA. The BPA concentrations were plotted *vs.* the peak area ratios of $m/z = 405$ to $m/z = 413$ and fit to linear calibration curves of the equation

$$Q_A = C \times \frac{R_A}{R_s},$$

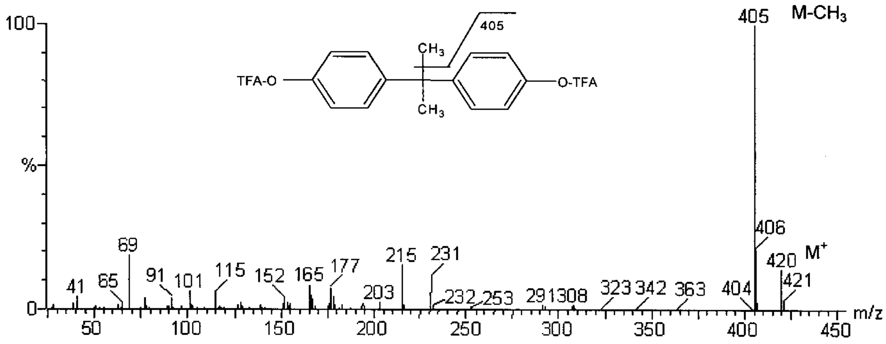

FIGURE 1 pos. EI mass spectrum of *bis*-TFA BPA.

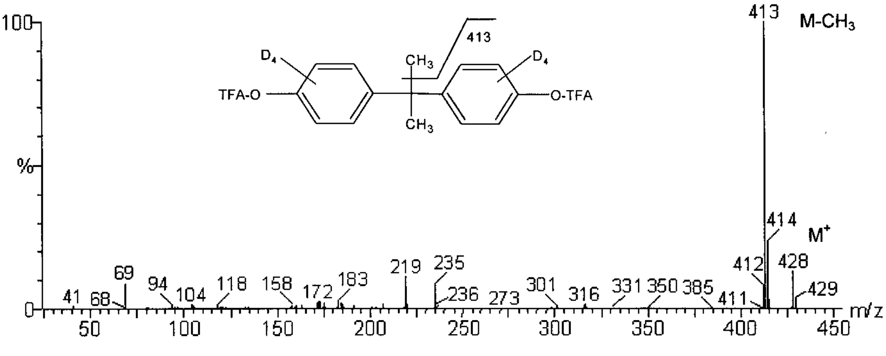

FIGURE 2 pos. EI mass spectrum of *bis*-TFA D_8-BPA.

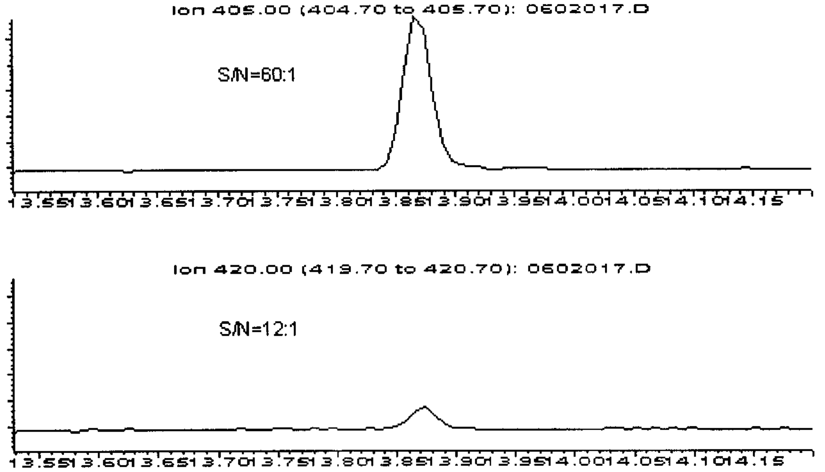

FIGURE 3 Ion traces of target and confirmation ions of 5 ng/L BPA in water.

with Q_A, concentration of BPA; R_A, peak area of BPA; R_s, peak area of D_8-BPA; C, slope of the curve.

The fit factors (r^2) of such curves were typically greater than 0.996 which demonstrates the linearity of the method. All peak areas were corrected for BPA and D_8-BPA isotope crossover. The isotope crossover was determined by fortifying deionized water samples with respective amounts of BPA or D_8-BPA and submitting these samples to the entire described sample preparation procedure. All data were corrected for the native BPA portion observed in D_8-BPA by considering the $m/z = 405$ response contribution of D_8-BPA[12].

Method validation

The method was validated over the concentration range of 5.0–514.8 ng/L BPA in deionized water. The method validation was based on relative and absolute recovery data obtained with pure reagent water samples. Absolute recoveries were determined with the recovery internal standard 1,2,4-tribromobenzene, which was added to the final sample ready for GC-MS quantification. The relative recoveries were between 91.1 and 101.3 percent at all concentration levels and the absolute recoveries were with few exceptions greater than eighty percent. Interday recovery data have been determined in order to document the robustness of the method. The validation data are reflected in Table I.

The applicability of the method to surface water was checked with River Elbe water. However a similar method validation with River Elbe water was not possible since river water samples free of BPA were not available. Instead the validity of the method for river water was verified by determining the BPA concentration in the river water and subsequent addition of approximately 50 ng/L, 100 ng/L and 250 ng/L of BPA. The recoveries of the added BPA amounts were determined. They were found to be between 96.7 and 108.3 percent. It is important to mention in this context that further clean-up steps of the river water extracts were not necessary. The recovery data are shown in Table II.

TABLE I Relative and absolute recoveries of BPA from deionized water fortified with 5.0–514.8 ng/L

Nominal concentration	*Day 1*			*Day 2*		
	Rel. recovery (%)	*Abs. recovery (%)*	*Confirmation ratio*	*Rel. recovery (%)*	*Abs. recovery (%)*	*Confirmation ratio*
5.0 ng/L	91.8 ± 9.7	81.6 ± 3.6	6.9 ± 0.4	91.1 ± 6.9	83.4 ± 3.8	6.9 ± 0.4
19.8 ng/L	92.8 ± 4.4	81.8 ±1.8	7.1 ±0.1	94.5 ±6.6	81.3 ± 2.5	7.4 ± 0.3
51.5 ng/L	102.3 ± 5.1	76.3 ± 5.3	7.3 ± 0.8	96.8 ± 3.3	82.5 ± 7.1	6.9 ± 0.4
99.0 ng/L	97.2 ± 3.3	86.0 ± 7.7	7.2 ± 0.3	97.8 ± 3.3	83.0 ± 3.7	7.3 ± 0.4
257.4 ng/L	97.8 ± 3.6	84.9 ± 3.1	7.1 ± 0.4	98.1 ± 4.0	82.6 ± 2.7	7.1 ± 0.5
514.8 ng/L	99.2 ± 3.0	79.0 ± 4.1	6.7 ± 0.5	96.8 ± 4.5	79.2 ± 1.4	7.1 ± 0.5

The recoveries were determined by five replicate analyses at each concentration level on each day. Rel. Recovery: the absolute recovery is the ratio of the observed BPA ng/L concentration determined with the D_8-BPA surrogate standard and the nominal ng/L concentration × 100%.

Abs. Recovery: the absolute recovery is the ratio of the ng BPA amount in the extract determined with the 1,2,4-tribromobenzene recovery internal standard and the nominal ng BPA amount in the sample × 100%.

Confirmation Ratio: ion $m/z = 405$ response/ion $m/z = 420$ response.

TABLE II Relative recoveries of BPA from fortified River Elbe water samples

Fortified BPA concentration	*Rel. Recovery (%)*	*Confirmation ratio*
55.4 ng/L	108.3 ± 11.5	7.7 ± 0.2
118.8 ng/L	96.7 ± 6.2	8.1 ± 0.2
237.6 ng/L	101.5 ± 2.6	7.5 ± 0.1

Adsorption of BPA on suspended particulate matter

Surface water samples frequently contain suspended particulate matter. It can be expected that such samples will contain BPA in solution and adsorbed on the solids. The distribution of BPA in solution and adsorbed on the solids will be strongly dependent on the nature of the particulates and the solubility of BPA in water, which is affected by the solution pH, salt content and other factors. An impact of the sample matrix on the analytical results can be expected in particular at very low concentration levels. Small changes of the nature of the sample matrix can impact the quantitative results generated. Therefore the analytical requirements for each new sample matrix have to be evaluated and optimized.

A feasible approach to assess the impact of the sample matrix and to optimize the analytical procedure is the analysis of filtered and unfiltered particulates containing samples as well as clean water reference samples. In particular the comparison of the recovery rates of the D_8-BPA surrogate standard of filtered and unfiltered samples will indicate losses due to adsorption on the solids present.

The investigation of filtered and unfiltered surface water samples containing sediments and respective samples containing biological sludge of a waste water treatment plant clearly showed that the D_8-BPA surrogate standard is adsorbed by the solids present leading to poor surrogate standard recoveries. As a result of this phenomenon false positive results are obtained when the unfiltered samples are quantitated using the D_8-BPA surrogate standard. However the quantitative evaluation of the same

TABLE III Comparison of BPA concentrations determined in filtered and unfiltered water samples containing particulate matter. Quantitative results were obtained with (a) the D_8-BPA surrogate standard and (b) the 1,2,4-tribromobenzene recovery standard; Sample A: Surface water containing *ca.* 30 mg/L of sediment; Sample B: water containing *ca.* 50 mg/L of waste water treatment plant sludge

Sample	*Filtered samples*		*Unfiltered samples*	
	Concentration	*D_8-BPA recovery*	*Concentration*	*D_8-BPA recovery*
a) Quantitation with the D_8-BPA internal standard				
Sample A	112.9 ng/L	83.7%	176.96 ng/L	52.5%
Sample B	244.0 µg/L	89.9%	418.2 µg/L	48.8%
b) Quantitation with the 1,2,4-tribromobenzene recovery standard				
Sample A	118.9 ng/L		121.0 ng/L	
Sample B	225.5 µg/L		229.0 µg/L	

data with the 1,2,4-tribromobenzene recovery standard, which is added to the extract for D_8-BPA absolute recovery determinations, showed that the extracts of filtered and unfiltered samples contained virtually identical BPA concentrations. The results of respective experiments are summarized in Table III.

The concentrations of the unfiltered samples determined with the D_8-BPA surrogate standard are overestimated by *ca.* 50% for Sample A and 74% for Sample B. The data reflected in Table III show that the applied sample preparation procedure is only capable to extract the dissolved portions of BPA and D_8-BPA. Adsorbed BPA is not isolated and thus the total BPA content in the sample, which would also include absorbed BPA, is not determined. The isolation of BPA from particulates would require more stringent conditions as described in the literature[14]. In order to be able to observe the loss by adsorption phenomena it is recommended to determine the recovery of the D_8-BPA surrogate standard by use of a recovery standard or by monitoring the absolute abundance of D_8-BPA in the sample and compare it with respective responses of clean water standards.

Sample filtration efficiency

The sample filtration efficiency was investigated by direct comparison of BPA fortified deionized water at 5 and 50 ng/L concentration levels. The results, which are reflected in Table IV, show that the relative recoveries were not impacted by the filtration step at both concentration levels. Although comparative experiments have not been performed in this context it is assumed that an all Teflon$^{\text{TM}}$ filtration device has to be used at these

TABLE IV Recoveries of BPA from fortified and unfiltered deionized water samples (duplicate determinations)

Fortified BPA concentration	*Relative recovery*	
	Filtered sample	*Unfiltered sample*
5.0 ng/L	91.5 ± 8.2%	92.8 ± 4.8%
53.5 ng/L	95.0 ± 1,2%	95.7 ± 1.4%

low concentrations. Telfon^{TM} is known to have a comparable low tendency to absorb organics on its surfaces.

Data integrity verification

Control samples, method and solvent blanks were run in order to demonstrate the integrity of the generated data. In this context it is important to have a procedure to clean efficiently all glassware and devices that have been in contact with BPA and the deuterated analog. The laboratory personnel should strictly follow the procedure.

CONCLUSIONS

A GC-MS method for the determination of BPA in water has been developed and validated for a concentration range of 5.0–514.8 ng/L. The method employs liquid/liquid extraction for analyte and internal D_8-BPA standard isolation and concentration. Trifluoroacetylation of BPA and D_8-BPA is employed in order to obtain less polar derivatives. The relative recoveries of BPA are greater than ninety percent over the entire validated concentration range. The impact of suspended particulate matter present in samples on the quantification results could be demonstrated. False positive results are obtained due to losses of the surrogate standard by adsorption on the particulates present.

References

[1] Bisphenol A (2,2-*bis*-(4-hydroxyphenyl) propane), BUA 203 (December 1995) Gesellschaft Deutscher Chemiker, S. Hirzel Wissenschaftliche Verlagsgesellschaft, Stuttgart 1997, English Edition: ISBN 3-7776-0798-3.

[2] Umweltbundesamt (UBA, German Federal Environmental Agency) BPA Monitoring Data, June 1999.

[3] A. Wenzel *et al*., Frauenhofer-Institut Umweltchemie and Ökologie – IUCT – and Abt. Biochemische Ökologie–EBÖ, Research Report 216 02 011/11, pp. 24–34 (1998).

[4] G. Matsumoto, R. Ishiwatari and T. Hanya, *Water Res*., **11**, 693–698 (1997).

[5] G. Sun and P.W. Geno, Determination of trace endocrine active phenolic compunds by GC/MS. *Meas. Toxic. Relat. Air Pollut., Proc. Spec. Conf.*, Vol. 2, pp. 477–485. Air and Waste Management Association, Pittsburg, PA (1997).

[6] R.A. Rudel, S.J. Melly, P.W. Geno, G. Sun and J.G. Brody, *Environ. Sci. Technol*., **32**, 861–869 (1998).

[7] A. Gonzalez-Casado, N. Navas, M. del Olmo and J.L. Vilchez, *J. Chromatogr. Sci.,* **36**, 565–569 (1998).

[8] K. Ozaki, R. Inomata, M. Nishiwaki and E. Kitajima, *Niigata-ken Kogai Kenkyusho Kenkyu Hokoku. Niigata*, **2**, 8–12 (1977).

[9] D.A. Markham, D.A. McNett, J.H. Birk, G.M. Klecka, M.J. Bartels and C.A. Staples, *Intern. J. Environm. Anal. Chem*., **69**, 83–98 (1998).

[10] D.R. Knapp, *Handbook of Analytical Derivatization Reactions*. John Wiley and Sons, New York, NY (1979).

[11] J. Hetper, M. Kiedik, J.B. Kyziol and Z. Daszkiewicz, *Chem. Anal*., **37**, 399–403 (1992).

[12] M.P. Barbalas and W.A. Garland, *J. Pharm. Sci*., **80**, 922–927 (1991).

[13] K.-L. Ring, H. Janshekar and N. Takei. *Chemical Economics Handbook*. SRI Consulting, 333 Ravenswood Avenue, Menlo Park, CA 94025-3477 (1997).

[14] O.P. Heemken, B. Stachel, N. Theobald and B.W. Wenclawiak, *Arch. Environ. Contam. Toxicol*., **38**, 11–31 (2000).