

Influence of the injection technique and the column system on gas chromatographic determination of polybrominated diphenyl ethers

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Abstract

In this paper, we present an investigation of the influence of the gas chromatographic separation system on the determination of polybrominated diphenyl ethers (PBDEs). Capillary columns, retention gaps and press-fit connectors, as well as different injection techniques have been evaluated with respect to yield and repeatability. The split/splitless injection has been optimized and compared to on-column injection, the septum equipped temperature programmable injector (SPI) and the programmable temperature vaporizing (PTV) injector. Furthermore, a comparison of the different operational modes of the PTV injector is presented. The results show that there are large variations in the yield of PBDEs depending on the column and the injection systems. Especially the high molecular weight BDE congeners can be subject to severe discrimination. Unfavorable conditions can lead to a complete loss of nona and deca substituted BDE congeners.

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1. Introduction

During the last years, there has been an increasing interest directed towards polybrominated diphenyl ethers (PBDEs) as environmental pollutants. PBDEs have been found in samples from various environmental compartments such as sediment [1,2] and fish [3,4], but also in human blood [5] and breast milk [6,7]. Increased concentrations of PBDEs have been found in the blood of occupationally exposed groups working at recycling plants for electronic equipment [8].

The main use of PBDEs is as flame-retarding additives to plastic materials in electronic appliances [9]. PBDEs are merchandised as technical mixtures of diphenyl ethers with a varying degree of bromination, containing mainly tetra to decabrominated diphenyl ether. Since the technical pentabrominated diphenyl ether mixture has been recognized as an environmental pollutant, the main PBDE product used today has shifted towards deca-BDE (BDE-209) [10].

However, most reports of PBDEs in environmental samples do not include analysis of the highly brominated products. Only a few determinations of BDE-209 has been reported, for instance from analysis of air [11,12] and human blood [8].

In 2002, the accounts of a round robin study of laboratory quality control regarding determination of PBDEs in marine samples were published. This study showed that there was a good agreement in the determination of brominated diphenyl ethers with up to six bromine substituents [13]. BDE-209 was the only BDE with more than six bromine substituents that was included in the study and the determination of this compound was evidently difficult. There were large discrepancies in the reported concentrations, and the analysis with respect to BDE-209 was not under control in most of the participating laboratories. A conclusion that can be drawn from that investigation is that conventional analytical methods for determination of halogenated pollutants, such as methods developed for PCBs, are not satisfactory for determination of PBDEs with a high degree of bromination.

High resolution between different BDE congeners and low detection limits has made GC the standard analytical method for PBDEs. Capillary columns with a length between 30 and

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60 m, an inner diameter of 0.25–0.32 mm and a non-polar stationary phase, like (5% phenyl-methylpolysiloxane) or 100% dimethylpolysiloxane, with a film thickness between 0.1 and 0.25 μm , are normally used for the determination of mono to heptaBDEs. In some cases shorter columns have been used for the determination of the high molecular weight BDE congeners such as BDE-209 [13]. The most commonly used GC injection technique for PBDEs is splitless injection [13]. However, the septum equipped temperature programmable injector (SPI) [12], as well as the programmable temperature vaporizing (PTV) injector [14] and on-column injector [15] have been used. Common detection principles in gas chromatographic analysis of PBDEs are mass spectrometry (MS) and electron capture detection (ECD) [1,16–18].

To inject and separate high molecular weight compounds such as the octa, nona and deca substituted BDE congeners, high temperatures are needed in some of the GC injection and column systems. However, the high molecular weight BDE congeners are thermally labile compounds, e.g. BDE-209 starts to degrade at temperatures around 300 °C [9]. This means that a critical view have to be put on the selection of the components of the gas chromatographic system, since discrimination and degradation of PBDEs may occur during injection, as well as during the separation. Initial studies at our laboratory indicated large variations in the yield of PBDEs on different GC-systems. In some cases, the BDE-209 peak completely vanished.

In this paper, we give an account of our study of the influence of column systems and injection techniques on the GC analysis of PBDEs. Sources to severe analytical errors and suggestions for optimization of the GC analysis of high molecular weight PBDEs are presented.

2. Experimental

2.1. Chemicals

The individual PBDE congeners are identified by the IUPAC numbering system for PCBs [19]. A stock solution was prepared by dissolving the individual congeners: BDE-2, 3, 7, 13, 17, 47, 49, 99, 100, 153, 154, 183, 184, 190, 209 (Department of Environmental Chemistry, Stockholm University, Sweden), BDE-191, 196, 197, 203, 206, 207 (Wellington Laboratories, Ontario, Canada) and 1,2-bis(2,4,6-tribromophenoxy) ethane (BTBPE) (Institute of Applied Environmental Research, ITM, Stockholm University, Sweden) in toluene. Test solutions were then made by dilution in *n*-hexane giving concentrations in the range of 75–200 pg/ μL . During the retention gap and press-fit study, a second test mixture was used. It was prepared by dissolving Bromkal 70-5DE and Bromkal 79-8 together with BDE-119, BDE-190 and Dechlorane[®] 603 (C₁₀Cl₁₂) in toluene (SupraSolv, Merck, Darmstadt, Germany) in the following concentrations: Bromkal

70-5DE 280 pg/ μL , Bromkal 79-8DE 460 pg/ μL , BDE-119, BDE-190 185 pg/ μL and Dechlorane[®] 250 pg/ μL .

Comparison of for example columns was made by normalizing the areas of the individual congeners for each column towards the corresponding congener area for the column that was considered to give the best results, i.e. the highest yield of PBDEs. Each experiment was repeated five times, and the average normalized area and relative standard deviation was calculated.

2.2. Air sampling

Air samples were collected at a recycling plant in Stockholm, Sweden, where electronic equipment is dismantled, using AirCheck 2000 personal air sampling pumps (SKC Inc., PA, USA). A glass fiber filter (25 mm binder free borosilicate glass fiber filter (type A/E, Gelman Sciences Inc., Ann Arbor, MI, USA) and two cylindrical polyurethane foam plugs (PUFs) (length 15 mm, diameter 15 mm, porosity 60 ppi, density 25 kg/m³, Specialplast AB, Gillinge, Sweden) were mounted in series in a sampling head of anodized aluminum. The second PUF served as a control of sampler break through. All adsorbents were washed according to a previously published procedure [20].

The sampling time was 12.0 h and the sampling rate 3.00 L/min, giving a sampled volume of 2.16 m³ for all samples. Filter and adsorbents were separately extracted by use of ultrasonic assisted solvent extraction in 2 × 5 mL of dichloromethane during 2 × 20 min, giving a total extract volume of 10 mL. The solvent was changed to *n*-hexane and the volume reduced to 1 mL. To remove polar and particulate material, the sample was applied on a 1 g NH₂ solid phase extraction (SPE) cartridge (Isolute, International Sorbent Technology, UK) and eluted with 10 mL hexane.

2.3. Columns, retention gaps and press-fit connectors

Ten columns were tested: DB-1, DB-5MS, DB-5MS ($d_f = 0.25 \mu\text{m}$) and DB-35MS ($d_f = 0.15 \mu\text{m}$), DB-200 ($d_f = 0.25 \mu\text{m}$), HP-1, HP-5MS and DB-XLB from Agilent Technologies (Palo Alto, CA, USA), RTX-500 ($d_f = 0.18 \mu\text{m}$) from Restek (Restek Corp., Bellefonte, PA, USA) and a VF-5MS, FactorFOUR column from Varian (Varian Inc., Palo Alto, CA, USA). All columns had a length of 15 m, an inner diameter of 0.25 mm and contained bonded and cross-linked stationary phases with a film thickness (d_f) of 0.10 μm if not otherwise stated.

Six fused silica retention gaps were included in the study. An untreated fused silica (untreated), a PEG-deactivated fused silica (polar), a phenyl/methyl-deactivated fused silica (intermediate-polarity) and a methyl-deactivated fused silica (non-polar) retention gap were obtained from Supelco Inc. (St. Louis, MO, USA). A phenyl/methyl-deactivated fused silica (intermediate-polarity) from J&W Scientific and a Siltek[®] deactivated retention gap from Restek Corp.

Table 1
The settings of the PTV injector for the four investigated operational modes

Injection mode	Start temperature (°C)	End temperature (°C)	Temperature programming (°C/min)	Pulse pressure, time (kPa, min)
Splitless	300	300	–	
Pulsed splitless	300	300	–	400, 2
Temperature programmed splitless	60	300	700	
Temperature programmed pulsed splitless	60	300	700	400, 2

(Bellefonte, PA, USA) were also included. All retention gaps had a length of 3 m and an inner diameter of 0.32 mm.

The following all-glass press-fit connectors were investigated: Siltek[®] deactivated universal (Restek Corp.), CP-Quick-Seal, 0.25 mm/0.32 mm (Varian Inc., Palo Alto, CA, USA), and Two-Way Fused Silica Universal Union (J&W Scientific).

2.4. Instrumentation

For the column evaluation and optimization of the programmed temperature vaporizer (PTV) as well as the splitless injection, an Agilent 6890 gas chromatograph equipped with a 7683 auto-sampler, an on-column injector, a PTV (CIS-4 Gerstel, Mülheim an der Ruhr, Germany) or a split/splitless injector and a micro electron capture detector (μ ECD, Agilent) or a 5973 mass selective detector (Agilent) was used. The on-column injector was operated in track-oven mode. The settings for PTV and splitless injector parameters are shown in Tables 1 and 2, respectively.

The oven was programmed from 50°C (2 min) up to 325°C (5–15 min) at a rate of 20°C/min. Nitrogen was used as carrier gas at a flow rate of 30 cm/s.

The μ ECD temperature was 350°C and nitrogen was used as make-up gas at a flow of 60 mL/min.

When using the MS detector, helium was used as carrier gas at a flow rate 30 cm/s, methane was used as reagent gas at a pressure of 2×10^{-4} Torr and the electron energy was set to -175 eV. The ion source had a temperature of 200°C and the quadrupole was kept at 150°C. The m/z 79 and 81 ions characteristic for the bromine ion, was monitored in electron capture negative ion mode.

For investigation of retention gaps, press-fit connectors and the septum equipped programmable injector (SPI) a Varian 3400 gas chromatograph (Varian Inc.) was used. It was equipped with a Varian 8100 auto-sampler, a Varian 1093 SPI, with a deactivated high performance insert and a Varian electron capture detector (ECD, ^{63}Ni) at a temperature of 330°C and a nitrogen make up gas flow of 30 mL/min.

Table 2
The investigated domain for optimization of the splitless injector

Parameter	Minimum	Maximum
Temperature (°C)	255	325
Splitless time (min)	0.15	4.35
Pressure pulse (bar)	0.1	4

Nitrogen was also used as the carrier gas at a flow rate of 30 cm/s. The GC oven temperature program was as follows: initial temperature 60°C during 4 min, a first ramp of 40°C/min up to 175°C, a second ramp of 10°C/min up to 305°C. After a hold time of 5 min, a final ramp of 40°C/min up to 325°C was applied with a hold time of 3 min. The SPI was programmed as follows: from 60°C the temperature was increased by 150°C/min up to 320°C which was held for 18 min. The SPI was then cooled using liquid CO₂.

3. Results

To optimize the gas chromatographic separation system with respect to PBDE, the influence of the column system, including retention gaps and press fit connectors as well as injection technique were investigated and evaluated.

3.1. GC injectors

The on-column, splitless, programmed temperature vaporizing and septum equipped temperature programmable injectors, were investigated with respect to discrimination and precision using both a PBDE standard solution and real PBDE air sample extracts.

3.1.1. The on-column injector

In the cold on-column injector, the sample is introduced directly into the column dissolved in a solvent. Thermal degradation and discrimination due to the sample injection system is therefore unlikely. The precision with regard to peak areas was high, with an average R.S.D. of 1.7% for all the investigated congeners and an R.S.D. of 1.2% for BDE-209. Two air samples were also analysed using the on-column injector, and the relative standard deviations were calculated as the average of three injections per sample. In this case, the R.S.D.s for the determined concentrations of BDE-47 and BDE-209 were 1.8 and 2.9%, respectively. These properties made us select the on-column as the “reference” injector for comparisons of injection techniques as well as for the further studies of the column system.

3.1.2. The septum equipped temperature programmable injector

The SPI is a cold, “at-column” injection technique where the sample is injected close to the entrance of the column. The column is tightened to the liner just as in a press-fit

connector, providing a high degree of analyte transfer. Still a solution, the sample is pushed into the column by the carrier gas in a process similar to that of an on-column injection. A temperature increase of the SPI thermally desorbs remaining high boiling compounds from the injector liner and into the column system where a retention gap is used to focus the chromatographic start band.

The SPI demonstrated a very good reproducibility, having R.S.D. values between 0.4 and 1.5% for the relative peak areas of the investigated PBDEs. Furthermore, discrimination of the PBDE analytes in the standard solution was low, i.e. comparable to the on-column injection. The SPI was not used for determination of PBDE in the air samples. An electron capture detector provides poor selectivity, compared to a mass spectrometer, and it was the only available detector in combination with the SPI during this investigation.

The SPI has successfully been used, by for example Sjödin et al [12], for determination of PBDEs, but since the production of this injector has ceased, the availability is limited. However, the manufacturing of tapered liners for splitless injectors and PTVs makes so-called “direct injection” possible. This is an alternative way to use the at-column injection technique.

3.1.3. The split/splitless (S/SL) injector

In the split/splitless injector the sample solution is vaporized in the injector liner insert and the analytes are subsequently transferred to the column in the form of vapor. This process can degrade thermally labile compounds and discriminate high molecular weight analytes. As the sample is not instantly vaporized, portions of the analytes may end up below the splitpoint, or even in the bottom of the injector, from where transfer of in particular high-boiling analytes is difficult [21–23]. The poor inertness of these parts of the injector could also catalytically induce thermal degradation. Furthermore, sample evaporation may occur inside the needle or on the needle tip, which lead will to discrimination of high molecular weight analytes.

However, the splitless injector is robust in terms of routine use and capacity to manage dirty samples. In addition, it is available in most laboratories equipped with gas chromatographs. To investigate this injector further, a full factorial design experiment was conducted to optimize the yield of PBDEs and in particular the decabrominated congener BDE-209. The experimental domain is shown in Table 2. The temperature and splitless time were the only parameters with significant effect on the response. The investigation indicated that both parameters should be kept as high as possible for maximum yield of all the BDE congeners and in particular BDE-209, Fig. 1. A conclusion that can be drawn from this is that the discrimination of heavy molecular weight PBDE congeners is a more important factor than thermal degradation in the splitless injector.

No significant increase in the response for any of the BDE congeners was observed when a pressure pulse was applied during the splitless time, i.e. during the transfer of the sample

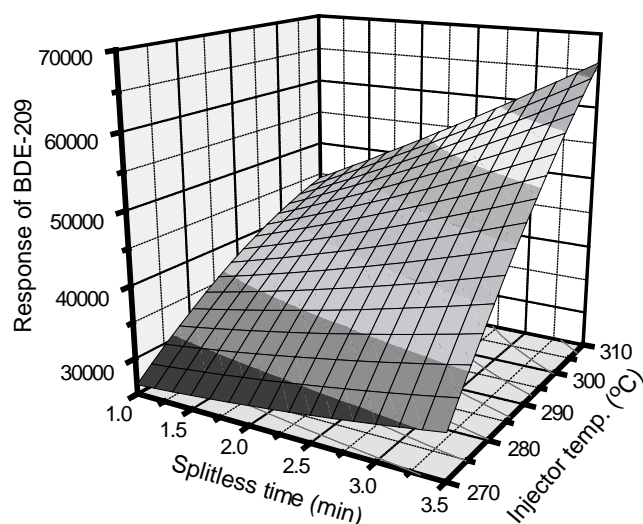


Fig. 1. Response surface of detector response for BDE-209 as a function of splitless time and injector temperature, with pressure held constant at 2.3 bar.

from the injector to the column. It should be stressed that the effect of the pressure pulse is also influenced by the geometry and the design of the liner and the injector. For other injector brands or types, for example the PTV (see below), this parameter may be of significance.

The optimized split/splitless injection, i.e. an injector temperature of 325 °C (the isothermal limit for many columns) and a splitless time of 4 min, was compared to a split/splitless injection using the mean settings taken from an interlaboratory study conducted by Jacob de Boer [13], i.e. an injection temperature of 275 °C and a split less time of 2 min.

The results obtained with the optimized and the mean settings of the splitless injector differed significantly with regard to both precision and discrimination in the GC analysis of PBDEs. The average R.S.D. for the relative peak areas was for the optimized method 1.1% and for the mean settings 9.3%. For BDE-209 the R.S.D. was 2.0 and 24.3%, respectively. Determination of BDEs in the air sample with the optimized method gave R.S.D.s for BDE-99 and BDE-209 of 5.0 and 13.7%, respectively. The corresponding values for the mean settings were 8.9 and 15.0%. However, even though the optimized method gives a 2.7 times higher yield of BDE-209 than the mean settings, it shows a severe discrimination in comparison to the on-column technique, Figs. 2 and 3. Furthermore, the calculated air concentrations of BDE-209 were in the range of 90–208% of those determined with the on-column injector. This illustrates the irreproducibility of the split/splitless injector with regard to determination of the high molecular weight BDE-congeners.

3.1.4. The PTV injector

The design of the PTV reassembles that of the traditional split/splitless injector, but it has a lower thermal mass and a narrower liner insert.

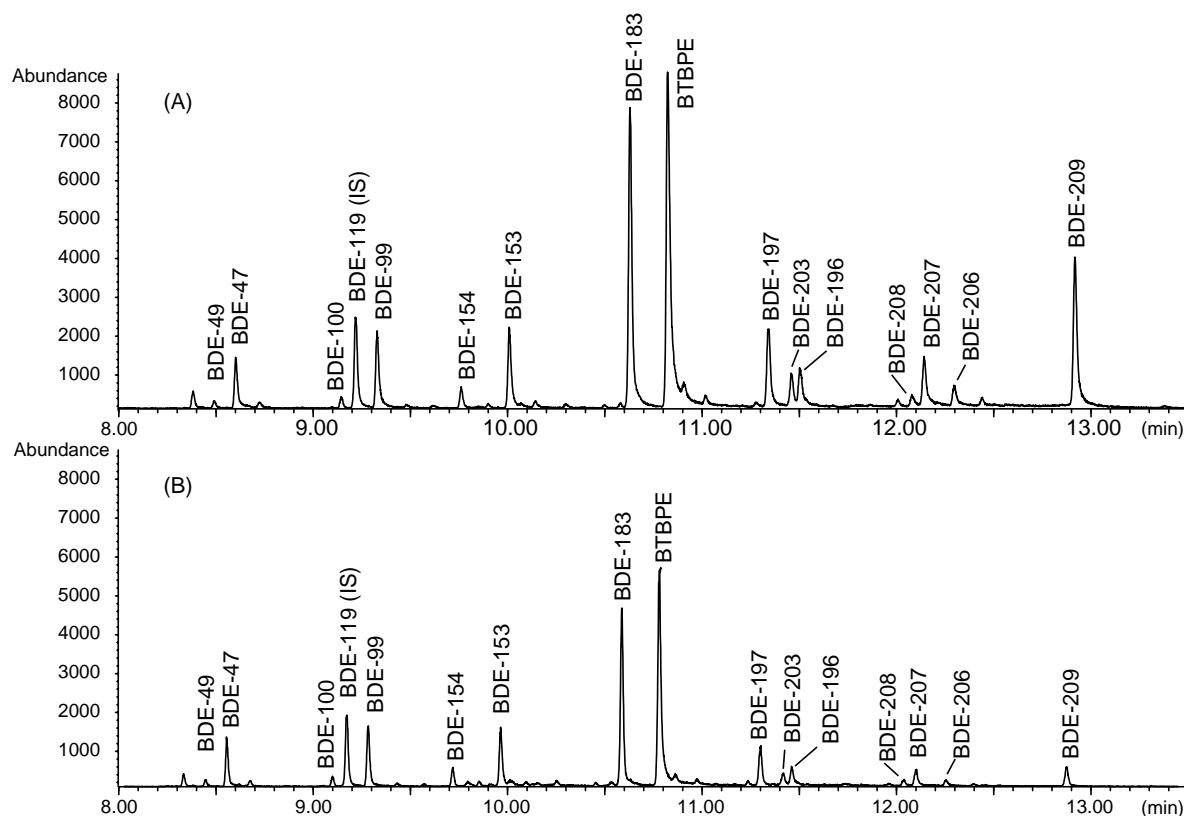


Fig. 2. GC-MS ECNI chromatograms of an air sample from an electronics dismantling facility, injected with the on-column (A) and splitless technique (B) respectively, on a 15 m DB-5MS (i.d. 0.25 mm, d_f 0.1 μ m) column. BTBPE = 1,2-bis(2,4,6-tribromophenoxy) ethane.

The evaporation of the sample in constant temperature mode is assumed to be ballistic due to the high injection temperature, as in the common splitless injector. This may result in sample components being disposed below the split-point in the injector and is thus not transferred quantitatively to the column. A more controlled evaporation of the sample is achieved in temperature programmed splitless mode when a steep temperature gradient is applied to transfer the sample on to the column. In this mode both the yield and the relative standard deviation is significantly improved.

Four different injection modes of the PTV were evaluated regarding repeatability and discrimination of the PBDEs.

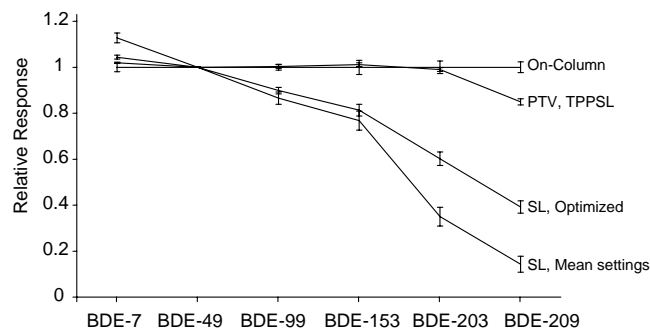


Fig. 3. The relative response for five selected BDE-congeners relative to BDE-49, using different injection techniques, where the response for on-column was set to one. Mean values are plotted ($n = 5$) and the error bars correspond to the standard deviation.

Fig. 4 illustrates the differences in performance of these different modes. If the PTV is operated in constant temperature splitless mode (SL), i.e. as a traditional splitless injector, there is a severe discrimination of the high molecular weight BDE congeners. The relative standard deviation is also higher compared to the other injection modes. By employing a pulsed pressure during the injection, pulsed pressure constant temperature splitless mode (PSL), the overall yield of the BDE congeners was increased. The increase in the yield was higher for the high molecular weight

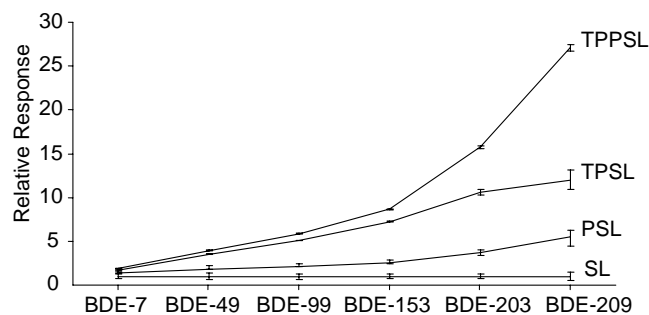


Fig. 4. The response for six selected BDE-congeners using different injection modes of the PTV injector, where the response for isothermal splitless was set to one. SL: isothermal splitless, PSL: pulsed isothermal splitless, TPSL: temperature programmed splitless and TPPSL: temperature programmed pulsed splitless. Mean values are plotted ($n = 5$) and the error bars correspond to the standard deviation.

BDE congeners such as BDE-209, with a fivefold increase in the peak areas, compared to a twofold increase in peak areas for BDE-47. The best results were obtained when both a temperature gradient and pulsed pressure (TPPSL) was applied. In this case, the peak area for BDE-209 was 28 times larger than the peak area obtained with the PTV in SL-mode. The yield of the high molecular weight BDE congeners was lower for the temperature programmed splitless (TPSL) compared to temperature programmed pulsed splitless (TPPSL) mode. The peak areas were 1.1 and two times larger for BDE-47 and BDE-209, respectively. This was to be expected since the pressure pulse is applied in order to increase the yield of low vapor pressure compounds such as the high molecular weight BDE congeners.

In comparison to on-column injection, temperature programmed pulsed splitless mode gave equally high yield of all the selected congeners, with the exception of BDE-209, for which a moderate, but significant, discrimination was observed, Fig. 3. This phenomenon was however reproducible, demonstrated by the low standard deviation (R.S.D. = 1.6). For the air sample the R.S.D.s for BDE-99 and BDE-209 were 1.2 and 4.3%, respectively.

These results shows that the PTV injector should be operated in temperature programmed pulsed pressure injection mode for optimal injection of PBDEs. It is not recommended to use the PTV as a splitless injector in constant temperature splitless mode for the determination of PBDE.

3.2. The GC column system

3.2.1. Retention gaps

Retention gaps are used to focus the start band of high molecular weight compounds to obtain narrow peaks often in connection with on-column and large volume injection techniques. They are also used as guard columns for protection of the analytical column. Since the retention gap may be several meters long, the analytes are in contact with a relatively large glass surface area. This surface can be either untreated fused silica with active silanol groups, or deactivated fused silica with non-polar, intermediate polar or polar deactivation.

We found that degradation of high molecular weight BDE congeners do occur in some of the investigated retention gaps. The best result, i.e. the least degradation, was obtained with Siltek® deactivated retention gaps. According to the manufacturer, these retention gaps have a highly inert glass surface, not susceptible to formation of active silanols to the same extent as other retention gap deactivation techniques. In Fig. 5 the normalized relative peak areas for the investigated PBDE congeners on a Siltek®, a non-polar and an untreated fused silica retention gap are shown. Corresponding curves for the polar and intermediate polar retention gaps are not shown in the figure, but falls between the curves for the non-polar and the untreated retention gaps. Compared to the Siltek® deactivated retention gap, the non-polar and untreated retention gaps exhibited a degradation of BDE-209 of

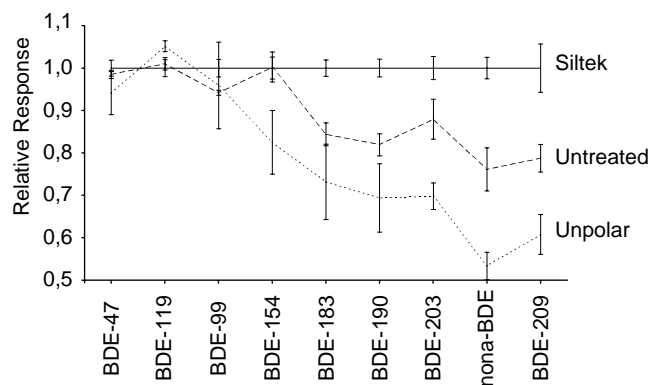


Fig. 5. The relative peak areas obtained using different retention gaps. The peak area ratios for the Siltek® deactivated retention gap have been set to one. Mean values are plotted ($n = 5$) and the error bars correspond to the standard deviation.

around 20 and 40%, respectively. Furthermore, the Siltek® deactivated retention gap gave the smallest relative standard deviation of the relative areas and thus better precision of the analytical method. A retention gap with intermediate polarity from J&W Scientific was also tested, but the performance with respect to PBDEs drastically deteriorated within a very short time of use.

3.2.2. Press-fit connectors

Press-fit connectors are used to connect the parts of the column system, such as retention gap and analytical column, in a simple way. Since they are made of glass, the surface has active silanol groups, which, if not deactivated, may have an influence on sensitive analytes. Press-fit connectors, from Varian, Agilent, and a Siltek® deactivated press-fit from Restek were investigated. This was made by adding two identical press-fit connectors to a 15 m DB-5 MS column. The first connector was added 20 cm from the injector end, and the second connector added 20 cm from the detector end of the column.

The Restek and Agilent connectors exhibited similar yield of PBDEs within the experimental error, but the Restek® connector gave lower standard deviations of the relative peak areas. When the press-fit connector at the detector end of the column was removed, similar results were obtained, but the relative standard deviations decreased. The Varian connector exhibited an increased discrimination of high molecular weight PBDEs. Compared to the Siltek® deactivated Restek connector it exhibited an increasing discrimination from BDE-190 up to BDE-209. For BDE-209 the decrease in yield was around 9%.

3.3. Capillary columns

We had earlier observed large variations in the yield of PBDEs from different kinds of capillary columns. The present study reveals a large variation between similar types of columns from different manufacturers. It also showed that a

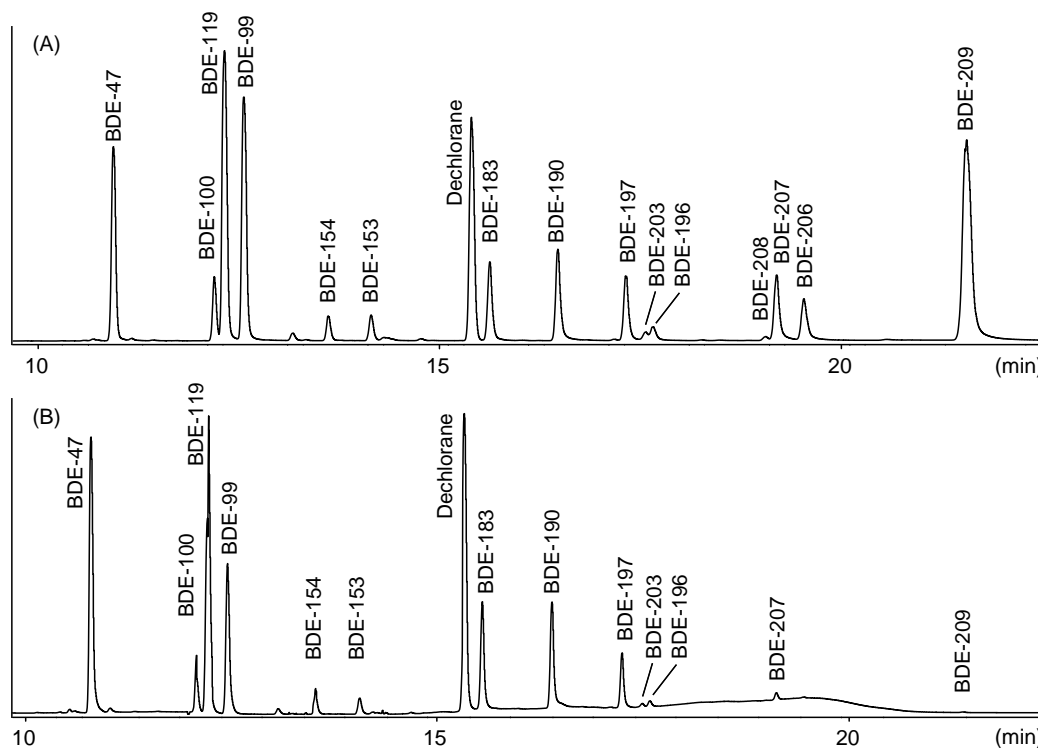


Fig. 6. Chromatograms from GC-ECD analyses of the PBDE test mixture on the DB-1 (A) and HP-1 (B) columns, respectively. Both columns had the dimensions $15\text{ m} \times 0.25\text{ mm}$, film thickness $0.1\text{ }\mu\text{m}$, and were connected to a $3\text{ m} \times 0.32\text{ mm}$ Siltek[®] deactivated retention gap; $1\text{ }\mu\text{l}$ injected with SPI.

number of column parameters have an influence on the yield of PBDEs. These parameters are the type and thickness of the stationary phase, and the length of the column.

3.3.1. Column brand

Columns with similar stationary phases from two manufacturers were evaluated; DB-1 and DB-5MS from J&W Scientific and HP-1 and HP-5MS from Agilent Technologies. There was no significant difference in yield of the low molecular weight PBDEs observed for any of these columns. However, for PBDEs with a higher degree of bromination, a significant difference between the column brands was observed. For the HP-1 column the degradation increased with the degree of bromination, and the nona-BDE as well as BDE-209 vanished almost completely. The chromatograms of PBDEs obtained from the two 100% dimethylpolysiloxane columns (DB-1 and HP-1) are shown in Fig. 6. Retention times are similar on both columns, but in the lower chromatogram representing the HP-1 column, the BDE-209 peak has disappeared. The hump of material present in the region from the heptabrominated BDE-183 to just prior to the expected elution time of BDE-209 was analyzed by mass spectrometry. It indicated that the material consisted of degradation products of nona-BDEs and BDE-209.

A comparable result was obtained for the two (5% phenyl-methylpolysiloxane) columns DB-5MS and HP-5MS. The DB-5MS showed similar results as the DB-1 column, while the HP-5MS exhibited a similar behavior as the HP-1 column, with an increasing degree of degradation of the

high molecular weight PBDEs. On this column, the yield of BDE-209 was 24%. Since the processes of commercial column manufacturing are confidential information, there is unfortunately no information about the difference in treatment of the two brands of columns, which could have explained the large difference in column performance.

3.3.2. Stationary phase

Four columns with different types of stationary phases were evaluated; DB-XLB (low polarity) and a DB-200 ((35% Trifluoropropyl)-methylpolysiloxane) from Agilent Technologies, VF-5MS (5% phenyl-methylpolysiloxane) from Varian and RTX-500 (Crossbond carborane/dimethyl polysiloxane) from Restek Corporation. The DB-XLB and

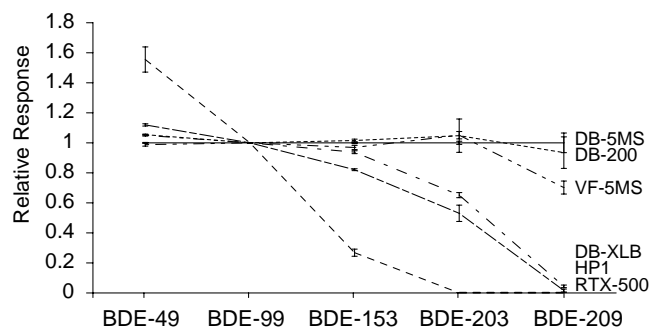


Fig. 7. The relative response of five selected BDE-congeners using different columns. The response for DB-5MS was set to one. Mean values are plotted ($n = 5$) and the error bars correspond to the standard deviation.

VF-5MS columns have well deactivated surfaces with a very low bleed, which may reduce the degradation of PBDEs in the column. The RTX-500 is a high temperature stable column allowing even the most high-boiling compounds to be eluted during the temperature ramp. The DB-200 is a column of intermediate polarity, which may be suitable for the analysis of PBDE. In addition, a HP-1 and a DB5-MS were included in the study as reference columns. An on-column injector was used to inject 1 μL of a PBDE standard solution (100–200 $\text{pg}/\mu\text{L}$) and the ratios between four selected BDE congeners (BDE-47, BDE-153, BDE-203 and BDE-209) and BDE-99 were calculated for all columns. These values were then compared to the DB-5MS column, for which the ratio of the selected congeners and BDE-99 was set to one.

The yield, in particular of BDE-209, differs significantly between the columns, Fig. 7. The best results, i.e. lowest discrimination and highest precision, were obtained with the DB-5MS column. The DB-200 and VF-5MS columns gave reasonable response of BDE-209, whereas the discrimination of BDE-209 on the DB-XLB, RTX-500 and the HP-1 columns was severe. When using the RXT-500 column BDE-209 could not be detected even when injecting as much as 1700 pg .

3.3.3. Stationary phase polarity

Three columns from the same manufacturer (J&W Scientific) with different stationary phase polarity were investigated; DB-1 with 100% dimethylpolysiloxane, DB-5MS

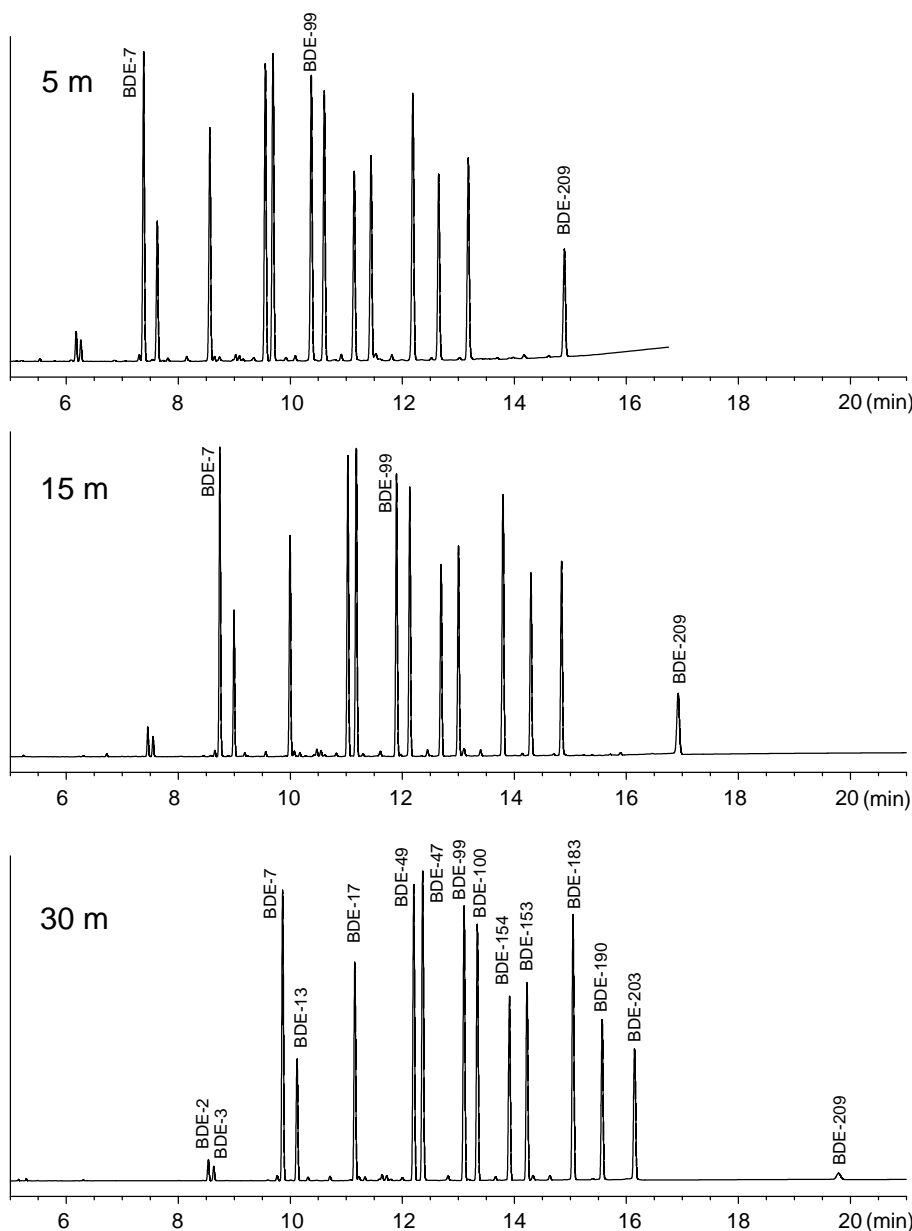


Fig. 8. GC-MS ECNI chromatograms obtained with 5, 15 and 30 m DB-5MS columns, respectively. The inner diameter was 0.25 mm and the film thickness 0.1 μm for all columns; 0.5 μL injected on-column.

with (5% phenyl-methylpolysiloxane) and DB-35 with (35% phenyl-methylpolysiloxane). The DB-1 and DB-5MS showed similar properties, while the DB-35 demonstrated an increasing degradation of high molecular weight BDE congeners starting with heptaBDE and continuing up to BDE-209. The yield of BDE-209 was reduced to around 20% on the DB-35 column compared to the DB-1 and the DB-5MS columns.

3.3.4. Stationary phase thickness

Two 15 m long DB-5MS columns with different stationary phase thickness, 0.1 and 0.25 μm , were investigated. The retention time of BDE-209 was 23.3 and 25.6 min, respectively. From BDE-153 and onwards, the yield of PBDEs was significantly lower on the column with the thicker stationary phase. For hepta- to nona-BDE the relative area decreased to around 70%. For BDE-209, the relative area was as low as 40%. The increased stationary phase thickness increases the elution temperature for an analyte and thus its time in the stationary phase at elevated temperatures. Here it is demonstrated that even such a small retention time difference as 2.3 min has a significant influence on temperature labile PBDEs such as BDE-209.

3.3.5. Column length

The time an analyte spends in the column is also a function of the length of the column. By shortening the column, thermally labile BDE congeners will spend less time at elevated temperatures in the column system. The difference in discrimination of high molecular weight BDE congeners was investigated by using columns (DB-5MS, 0.25 mm i.d.) of three lengths 30, 15 and 5 m. The effect of degradation was strongest for BDE-209 but still significant down to hepta-BDE, Fig. 8. This demonstrates the importance of designing the GC system in such a way that the shortest possible residence time in the column system is obtained for the PBDE analytes.

3.4. Column oven programming

The program of the GC oven influences, aside resolution, both the shape and area of the analyte peaks. Due to the relatively high boiling point of in particular BDE-209, a high final temperature is required to achieve a narrow peak. On the other hand, high temperatures will lead to degradation of the thermally labile BDE congeners. For this reason the influence of the final GC oven temperature was investigated between 275 and 350 $^{\circ}\text{C}$ on a 15 m DB-5MS column, with on-column injection.

As illustrated in Fig. 9, there was a significant decrease in the area for higher final temperatures, as an effect of thermal degradation. Due to band broadening at low temperatures and degradation at higher temperature the peak heights vary within the investigated domain and a maximum for the peak height is found around 325 $^{\circ}\text{C}$. The peak height is important

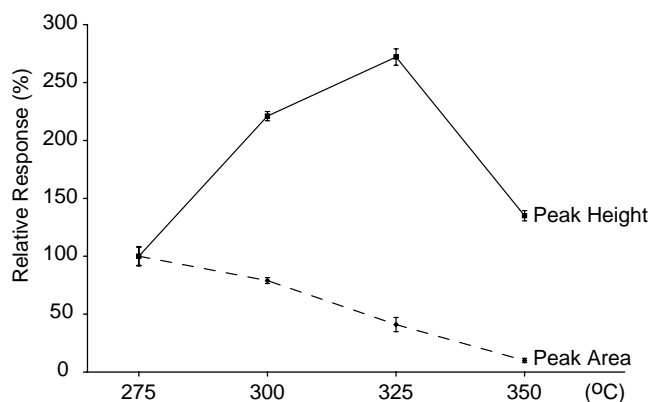


Fig. 9. The relative peak areas and peak heights of BDE-209 for four final GC oven temperatures. The plotted values are the mean percentages ($n = 3$) of the values obtained with 275 $^{\circ}\text{C}$ and the error bars correspond to the standard deviation.

in terms of LODs and LOQs, as well as for achieving a well-defined peak.

A final oven temperature of 300 $^{\circ}\text{C}$ may be a good compromise between degradation and band-broadening. With this final temperature large peak areas as well as low LODs and LOQs were obtained.

4. Conclusions

The main conclusion that can be drawn from the results presented above is that the composition and operation of the GC system has a significant influence on the gas chromatographic determination of PBDEs. The injection technique, type of retention gap, press-fit connector, column brand, stationary phase and column length significantly affect the yield of PBDEs from the chromatographic system, as well as the precision of the determination. By selecting a non-optimal GC system combination, the yield of nona- and deca-BDE can be decreased to zero, and the precision of the determination strongly decreased. This is true not only for the high molecular weight BDE congeners, as BDE congeners with down to five bromine substituents can be subject to discrimination and decreased precision.

The on-column injector is the most suitable injector for determination of PBDEs in clean samples. For a more complex sample matrix the PTV provides a good compromise between robustness and yield of high molecular weight congeners. In contrast to what have earlier been described in the literature, the PTV does not, correctly operated, degrade thermally labile BDE congeners to any large extent.

The recommendations for PBDE determination on GC are to use a short, non-polar DB column with a thin (0.1 μm) stationary phase. A PTV-injector, operated in temperature programmed pulsed splitless mode is recommended and, the retention gap should be Siltek[®] deactivated, as well as the press-fit connector used for connecting the retention gap and the analytical column.

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