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ANALYSIS OF POLYBROMINATED BIPHENYLS

J. J. DE KOK, A. DE KOK and U. A. Th. BRINKMAN*

Department of Analytical Chemistry, Free University, Amsterdam (The Netherlands)

and

R. M. KOK

Laboratory of Toxicology and Pharmacy, Academic Hospital of the Free University, Amsterdam (The Netherlands)

SUMMARY

High-performance liquid chromatography, gas chromatography-mass spectrometry, ultraviolet spectrometry and nuclear magnetic resonance spectrometry have been used to characterize the behaviour of a number of individual brominated biphenyls and to assess the composition of several mixtures of these compounds. The dependence of retention and spectral characteristics on the bromine content of the substituted biphenyls is discussed and the behaviour of polybrominated biphenyls is compared with that of polychlorinated biphenyls.

The applicability of high-performance liquid chromatography to photodegradation studies is demonstrated.

INTRODUCTION

Polybrominated biphenyls (PBBs) are a group of industrial compounds used as additive-type¹ flame retardants in synthetic polymers. Despite their structural similarity to the polychlorinated biphenyls (PCBs), which are well known environmental pollutants, the PBBs have received relatively little attention from chemists and biologists. Recently, an increased interest in the characteristics and use of PBBs has been shown, which is no doubt based on the ever-increasing knowledge of the potential hazards of polyhalogenated biphenyls and related types of compounds, and the Michigan PBB incident²⁻⁶, in which a large number of dairy cattle, pigs and chickens were accidentally poisoned. Moreover, in 298 persons exposed to contaminated products as a result of that incident concentrations of PBBs in blood, milk and adipose tissue of up to 2.26, 92.66 and 174.0 ppm, respectively, were found⁷. However, one should add that to date no acute or short-term illness has been clearly attributed to exposure to PBBs^{3,6}.

In the past 3 years, papers have been published on the determination of PBB residues and metabolites in dairy products^{8,9} and dry animal feeds¹⁰, the retention

* To whom correspondence should be addressed.

and excretion of PBBs by hens and cows^{11,12}, their excretion into the milk of cows^{13,14}, the distribution and clearance of PBBs in cows and calves¹⁵, the fate of PBBs in soil^{16,17}, their uptake by plants¹⁷, and the toxicology¹⁸⁻²⁴, metabolism²⁵ and photodegradation^{23,26-29} of PBBs. The identification of 2,4,5,2',4',5'-hexabromobiphenyl as the major component of the commercially available fireMaster BP-6 has been reported by two groups of workers^{28,30}.

Sundström *et al.*³¹ have written an extensive review on the synthesis of PBBs, which includes a section on the gas-chromatographic properties of these compounds. They pointed to the greater selectivity in PBB compared with PCB synthesis, which should be an advantage for industrial applications. Information concerning the availability, average composition, general characteristics and efficiency in flame retardancy is given in technical bulletins³²⁻³⁵ and in some papers^{1,36}. A survey of the literature on the analysis of PBB mixtures, which is almost invariably carried out by gas chromatography (GC) or combined GC-mass spectrometry (GC-MS), is included in the pertinent sections below.

In this paper, we discuss data on the analysis of PBBs by means of high-performance liquid chromatography (HPLC) and thin-layer chromatography (TLC) as well as by GC and spectrometric techniques.

EXPERIMENTAL

Materials

Small samples of a large number of individual PBBs and the industrial PBB mixtures "octabromobiphenyl" (FR 250 13A, Dow Chemical, Midland, Mich., U.S.A.) and "hexabromobiphenyl" (fireMaster BP-6, Michigan Chemical Corp.*, Chicago, Ill., U.S.A.) were gifts from RFR Corp. (Hope, R.I., U.S.A.). It should be noted that the samples were received prior to the issue of the RFR catalogue for 1977, which states that all individual PBB isomers have a purity of 99%. A sample of octabromobiphenyl XN-1902, stated to be similar to the material used in the toxicological studies quoted in refs. 22 and 23, was obtained through the courtesy of Dr. J. Norris (Dow Chemical), while Borg-Warner Chemicals (Amsterdam, The Netherlands) supplied a substantial amount of fireMaster BP-6. 4,4'-Dibromobiphenyl and 2,4,6,2',4',6'-hexabromobiphenyl were purchased from Fluka (Buchs, Switzerland) and Aldrich Chemical Co. (Milwaukee, Wisc., U.S.A.), respectively. A sample of the PBB mixture Bromkal 80-9D was a gift from Chemische Fabrik Kalk (Cologne, G.F.R.), while Péchiney Ugine Kuhlmann (Paris, France) and Hexcel (Basildon, Great Britain) supplied samples of their decabromobiphenyl preparations. Dr. Å. Norström (Umeå, Sweden) kindly sent us a small amount of the major component of fireMaster BP-6, while Dr. C. A. Wachtmeister (Stockholm, Sweden) provided us with a sample of 3,5,3',5'-tetrachlorobiphenyl. All samples were homogenized before use.

Solutions of individual PBBs were prepared in *n*-hexane. Because the solubility of brominated biphenyls in *n*-hexane and many other common organic solvents decreases rapidly with increasing bromine content, the PBB mixtures were dissolved

* Michigan Chemical Co. merged with Velsicol Chemical Corp. (Chicago, Ill., U.S.A.) on January 1st, 1977.

in warm carbon tetrachloride. All solutions were kept in bottles wrapped in aluminium foil in order to prevent photodegradation of the brominated biphenyls.

Methods

A Siemens S100 liquid chromatograph equipped with a 10- μ l automatic injection system and a Zeiss PM2 DLC UV detector was used for HPLC. The column was a stainless-steel tube, 25 cm \times 3 mm I.D., pre-packed with 5- μ m LiChrosorb SI 60 silica gel (Merck, Darmstadt, G.F.R.). *n*-Hexane (ChromAR, Mallinckrodt, St. Louis, Mo., U.S.A.), dried over molecular sieve 5A, was used as the mobile phase. The column was thermostated at $27 \pm 1^\circ$.

Reversed-phase TLC was carried out on Kieselguhr (Kieselguhr G, Merck) impregnated with paraffin oil. Development with acetonitrile-methanol-acetone-water (20:20:9:1) and acetonitrile-methanol-water (8:9:3) in a saturated chamber for a length of run of *ca.* 18 cm takes 1 h. Detection was effected by spraying with a solution of 0.5% toluidine in 80% ethanol containing 0.5% of glacial acetic acid, and subsequent irradiation under UV light (254 nm); blue or blue-green spots show up. A more detailed description of the TLC procedure is given elsewhere³⁷.

Gas chromatograms were run on an HP 5710A gas chromatograph (Hewlett Packard, Avondale, Pa., U.S.A.) using glass columns packed with 1% SE-30 on Chromosorb W (HP) (80-100 mesh) (1.95 m \times 2 mm I.D.) or 2% OV-17 on Chromosorb W (AW DMCS) (80-100 mesh) (1.25 m \times 2 mm I.D.), and a Pye Unicam (Cambridge, Great Britain) GCV chromatograph using glass columns packed with 4% OV-101 on Chromosorb W (HP) (80-100 mesh) (2.10 m \times 2 mm I.D.). Both chromatographs were equipped with a flame-ionization detector; quantitative analyses were performed by means of electronic integration. For GC-MS measurements, a Finnigan 9500 gas chromatograph (Finnigan, Hemel Hempstead, Great Britain) connected with a Finnigan 3200 quadrupole mass spectrometer with electron impact at 70 eV and a source temperature of 230 $^\circ$ was used.

¹H- and ¹³C-NMR spectra were recorded (by Dr. O. S. Akkerman, Department of Organic Chemistry) with a Bruker WH 90 F.T. spectrometer (Bruker-Physik, Karlsruhe, G.F.R.). ¹H chemical shifts were measured in CDCl₃ as a solvent.

Melting points were determined with a Kofler-Mikroheiztisch equipped with a RCH Mikroskop (C. Reichert Optische Werke, Vienna, Austria). UV spectra were recorded on a Beckman Acta CIII spectrometer using cells with quartz windows.

RESULTS AND DISCUSSION

Purity of the samples

In order to check the purity and/or identity of the various samples, high-performance liquid and gas chromatograms were run using UV and flame-ionization detection, respectively. Assuming equal responses for the main component and any impurities present, a purity of over 95%, and often even 99%, could be calculated for most individual PBBs. For these products, melting points were in good agreement with those reported by Sundström *et al.*³¹. The samples of 2,5,3'-tri- and 3,5,3',5'-tetrabromobiphenyl were found to have a purity of about 90%. The samples of 2,4,6,2',4',6'-hexa- (RFR and Aldrich) and 2,3,5,6,2',3',5',6'-octabromobiphenyl were found to be very impure preparations; their composition as well as that of the

TABLE I
 UV SPECTRAL DATA FOR INDIVIDUAL PBBs AND CORRESPONDING PCBs
 UV conditions: solutions in *n*-hexane; Beckman Acta CHH spectrometer. Data on PCBs taken from refs. 37 and 38.

PBB isomer	PBB			PCB		
	Main band λ_{max} (nm)	$\text{Log } \epsilon$ ($l \cdot \text{mole}^{-1} \cdot \text{cm}^{-1}$)	<i>k</i> band λ_{max} (nm)	Main band λ_{max} (nm)	$\text{Log } \epsilon$ ($l \cdot \text{mole}^{-1} \cdot \text{cm}^{-1}$)	<i>k</i> band λ_{max} (nm)
Biphenyl	201	4.66	246	201	4.66	246
2-	201	4.51	240	201	4.65	239
3-	205	4.60	248	204	4.61	248
4-	200	4.67	254	199	4.67	253
2,2'-	198	4.64	220-230	199	4.59	230-240
2,5-	203	4.49	226	203	4.61	240
4,4'-	201	4.64	261*	200	4.66	258
2,4,6-	213	4.70	220-230	204	4.72	240
2,5,2'-	200	4.66	235-245	199	4.67	240
2,5,3'-	213	4.57		213	4.64	248
2,5,4'-	205	4.60	245-255	204	4.63	240-245
2,4,2',5'-	207	4.66		205	4.76	
2,5,2',5'-	204	4.67	235-245	201	4.69	
3,5,3',5'-	220	4.76	235-240	216	4.78	
2,4,5,2',4',5',**	216	4.66	255	212	4.76	252
3,4,5,3',4',5'-	227	4.76	272	212	4.76	245-250
2,3,5,6,2',3',5',6',**	224*	4.85		222	4.74	265
Nona-***	225	5.18		211	4.99	
Deca-	227	5.11		215	5.06	

* Results agree fully with recent findings of Ruzo *et al.*³⁷.

** See section on PBB mixtures.

various decabromobiphenyl samples will be discussed in more detail in the section on PBB mixtures.

Individual PBBs

UV spectrometry. Table I summarizes data on the UV absorption of the PBBs in the 200–360-nm region and compares these with data previously published^{37,38} for PCBs. The main band in these spectra is caused by $\pi \rightarrow \pi^*$ electron transitions, while the *k* band is generally attributed to the conjugated biphenyl system with the contribution of both biphenyl rings. Several spectra are shown in Fig. 1.

As for the main band, substitution of chlorine by bromine leads to insignificant changes (0–2 nm) in λ_{\max} with all low-substituted polyhalogenated biphenyls. However, with more highly halogenated compounds, bathochromic shifts of about 5–13 nm occur. It is noteworthy that exceptionally large shifts occur with the non-*ortho*-substituted 3,5,3',5'-tetra- and 3,4,5,3',4',5'-hexabromobiphenyl as well as with 3,5,3',5'-tetrachlorobiphenyl. This confirms and extends our earlier observations³⁷ on the similar behaviour of 3,4,3',4'-tetra- and 3,4,5,3',4',5'-hexachlorobiphenyl. The intensities of the main bands are markedly similar for PBBs and PCBs.

With the *k* band, the introduction of bromine atoms in positions *meta* or *para* to the phenyl–phenyl bond induces a shift in λ_{\max} towards the visible region, as is strikingly illustrated by 3,5,3',5'-tetra- and 3,4,5,3',4',5'-hexabromobiphenyl. On the other hand, *ortho* substitution, which causes a considerable hindrance for free rotation of the rings and thus a loss in coplanarity, effects a sharp decrease in the extinction coefficient of the *k* band. Moreover, the band itself shifts towards the ultraviolet region and shows up as a shoulder on the main absorption band.

HPLC. High-performance liquid chromatograms of all individual PBBs were run in the system silica gel–dry *n*-hexane. The capacity factor, k_i , of component *i* was determined from its retention time, t_{Ri} , and the retention time, t_{R0} , of a non-retarded component (*n*-pentane):

$$k_i = (t_{Ri} - t_{R0})/t_{R0} \quad (1)$$

The relative retention, $\alpha_{\text{PBB/PCB}}$, of corresponding individual PBBs and PCBs was calculated from their capacity ratios:

$$\alpha_{\text{PBB/PCB}} = k_{\text{PBB}}/k_{\text{PCB}} \quad (2)$$

From the results summarized in Table II, several conclusions can be drawn, as follows.

(1) Replacement of a chlorine with a bromine atom invariably leads to a longer retention time. Limiting ourselves to the low-substituted biphenyls (mono- to tetra-), where three or four pairs of compounds are available for comparison, we can conclude that $\alpha_{\text{PBB/PCB}}$ is chiefly determined by the degree of substitution: mono, 1.05; di, 1.2; tri, 1.3; tetra, 1.5. The order of elution in the PBB series occasionally is slightly different from that encountered in the PCB series, as is evident from the “reversals” that occur with the 4,4'-di- and 2,4,6-tri-, and with the 2,5-di- and 2,5,2'-trihalogenobiphenyls. This effect may be due to the enhanced *ortho* effect observed with PBBs (see below).

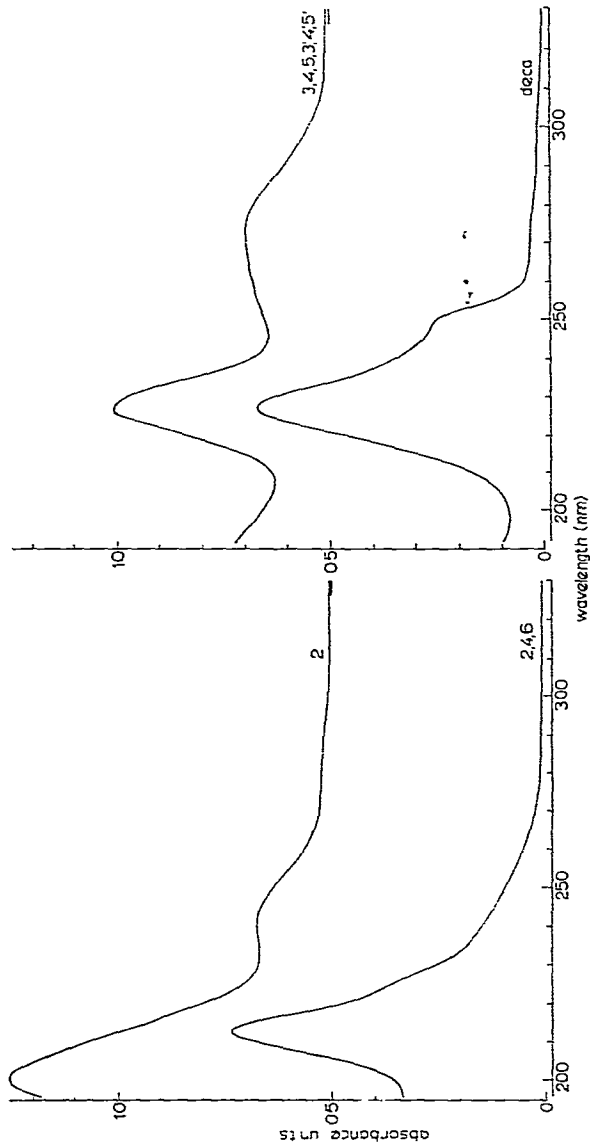


Fig. 1. UV absorption spectra of 2-mono-, 2,4,6-tri-, 3,4,5,3',4',5'-hexa- and decabromobiphenyl. Concentrations: 41, 43, 41 and 16 ppm, respectively, in *n*-hexane. Cell path length, 2 mm.

TABLE II

k VALUES OF INDIVIDUAL PBBs AND CORRESPONDING PCBs IN HPLC

HPLC conditions: silica gel (5- μ m LiChrosorb SI 60)-dry *n*-hexane; flow-rate, 1.4 ml/min; UV detection at λ_{max} ; temperature, $27 \pm 1^\circ$. Data on PCBs taken from refs. 37 and 38.

PBB isomer	<i>k</i>		$\alpha_{\text{PBB/PCB}}$
	PBB	PCB	
Biphenyl	9.26		—
2-	6.89	6.37	1.08
3-	4.86	4.60	1.06
4-	5.21	5.03	1.04
2,2'-	6.67	5.51	1.21
2,5-	4.17	3.57	1.17
4,4'-	3.31	2.88	1.15
2,4,6-	3.61	2.84	1.27
2,5,2'-	4.26	3.18	1.34
2,5,3'-	2.75	2.15	1.28
2,5,4'-	2.66	2.06	1.29
2,4,2',5'-	2.71	1.89	1.34
2,5,2',5'-	2.92	2.02	1.45
3,5,3',5'-	1.54	0.94	1.64
2,4,5,2',4',5'-	2.10	1.16	1.81
3,4,5,3',4',5'-	3.05	1.50	2.03
2,3,5,6,2',3',5',6'-	2.36*	0.98	2.41
Nona-	2.36		
Deca-	2.23	0.47	4.74

* Under the stated HPLC conditions, the 2,3,5,6,2',3',5',6'-octabromobiphenyl "mixture" (see section on PBB mixtures) yields a single peak.

(2) In general, the introduction of an increasing number of bromine (or chlorine³⁸) atoms into the biphenyl nucleus leads to a decrease in retention time. However, the introduction of bromine atoms *ortho* to the phenyl-phenyl bond distinctly promotes retention. This exception to the general rule is strikingly demonstrated in the series of mono-, di-, tri- and tetra-substituted bromobiphenyls. Combined *ortho*- and 2,3-substitution³⁸ effects probably cause the relatively high retention times of the octa-, nona- and deca-substituted PBBs, which are only slightly less than those of several tri- and tetra-substituted PBBs. Actually, whereas decachlorobiphenyl moves ahead of all PCBs studied (over 60 individual PCBs and a large number of commercial mixtures), two of the presently available PBBs, *viz.*, 3,5,3',5'-tetra- and 2,4,5,2',4',5'-hexabromobiphenyl, are eluted in front of decabromobiphenyl (see the next section). In view of the discussion given in ref. 37, we can conclude that the same will be true for 2,4,5,2',4',6'- and, even more so, 2,4,6,2',4',6'-hexabromobiphenyl. The increase in the *ortho*-effect noted with PBBs compared with PCBs is probably a consequence of the introduction of bromine instead of the less bulky chlorine atoms into the biphenyl nucleus.

(3) In a recent paper on the HPLC of PCBs³⁷, it was noted that 2,4,6-substitution effects an unexpectedly large decrease in retention time. Now, a similar effect is seen to show up with the purely *meta*-substituted halogenobiphenyls 3,5,3',5'-tetrabromo- and -tetrachlorobiphenyl. One might add that a *meta* effect is also

apparent in, *e.g.*, the series of mono-substituted PBBs and PCBs, where the isomers substituted in the 3-position move clearly ahead of the corresponding 2- and 4-monohalogenobiphenyls. No explanation can be forwarded here. However, it is interesting to note that the halogen atoms in the 2,4,6-substituted biphenyls are in a *meta* position relative to each other, although, admittedly, not to the central C-C bond.

GC. The commercially available PBB mixtures invariably have a high bromine content, which ranges between *ca.* 75 and 85% (w/w). As became apparent after several preliminary runs, HPLC in the system silica gel-dry *n*-hexane offers poor resolution for such mixtures. Therefore, the GC properties of the individual PBBs were studied in order to devise an alternative method of analysis. The results for various conventional GC systems are summarized in Table III; they are markedly similar to those reported by Sundström and co-workers^{27,31}. Two aspects are worth mentioning. (1) GC offers excellent resolution with the highly brominated biphenyls and, therefore, can be recommended for the analysis of the commercial PBB mixtures. In all further studies, 1% SE-30 and 2% OV-17 were used indiscriminately as stationary phases. (2) According to expectations based on GC studies of PCBs, the retention generally increases with increasing substitution of bromine atoms into the biphenyl nucleus. In this study, no chromatograms were run for PCBs and therefore it is not possible to calculate $\alpha_{\text{PBB/PCB}}$ values. Nevertheless, comparison of the data

TABLE III
RELATIVE RETENTION TIMES OF INDIVIDUAL PBBs IN GC

PBB isomer	Relative retention time			
	4% OV-101* (210°)	1% SE-30**		2% OV-17** (280°)
		235°	250°	
2-	0.21	—	—	—
3-	0.26	—	—	—
4-	0.26	—	—	—
2,2'-	0.33	—	—	—
2,5-	0.41	—	—	—
4,4'-	0.62	—	—	—
2,4,6-	0.70	—	—	—
2,5,2'-	0.70	—	—	—
2,5,3'-	0.92	0.15	—	—
2,5,4'-	0.98	0.16	—	—
2,4,2',5'-	1.57	0.23	—	—
2,5,2',5'-	1.48	0.22	—	—
3,5,3',5'-	2.68	0.40	—	—
2,4,5,2',4',5'-	—	1.13	1.08	1.48
3,4,5,3',4',5'-	—	2.67	2.37	3.39
Nona-	—	—	9.13	17.0
Deca-	—	—	13.2	27.2
<i>p,p'</i> -DDE	1.00	—	—	—
Decachlorobiphenyl	—	1.00	1.00	1.00

* GCV; carrier gas, N₂; flow-rate, 30 ml/min; injector temperature, 270°; detector temperature, 300°.

** HP 5710A; carrier gas, He; flow-rate, 18 (235°), 30 (250°) or 40 (280°) ml/min; injector temperature, 300°; detector temperature, 300°.

reported in Table III and ref. 31 with the GC retention-time data on PCBs collected by Sissons and Welti³⁹ clearly shows that $\alpha_{\text{PBB/PCB}}$ is determined primarily by the degree of substitution (as is also true in HPLC), its value increasing rapidly with an increasing number of substituents.

PBB mixtures

Literature data on the composition and analysis of commercially available PBB mixtures are reported in Table IV. In addition, we note that, according to the manufacturers, the production of fireMaster BP-6 ceased in late 1974, while Dow's and duPont's octabromobiphenyl have never been marketed^{40,41} as commercial prod-

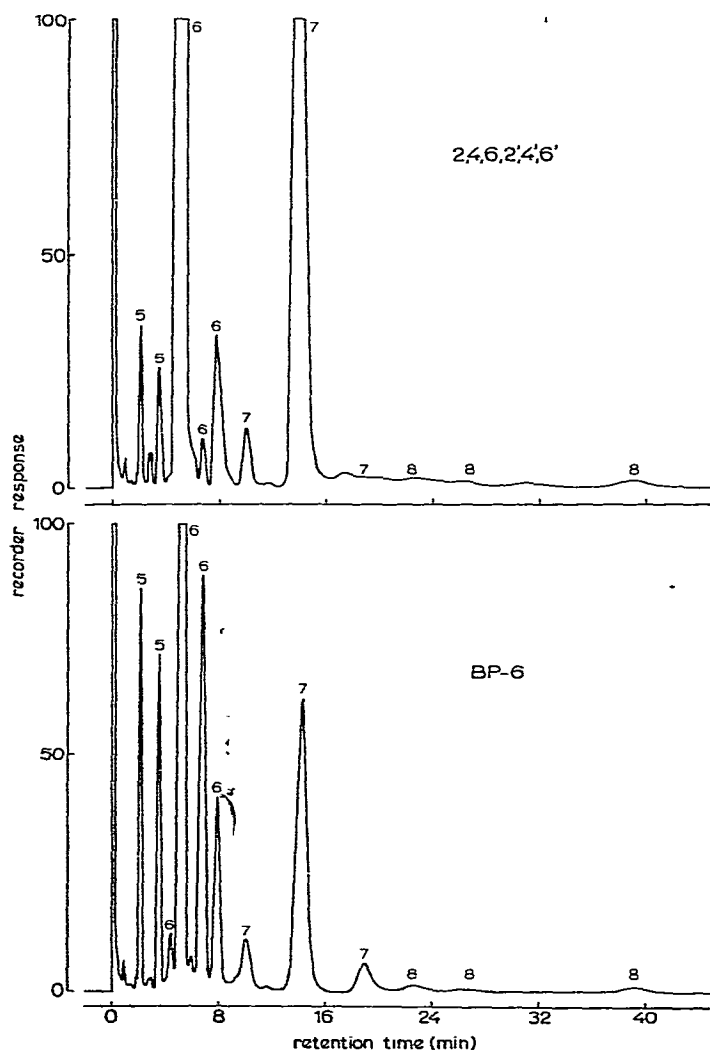


Fig. 2. GC of fireMaster BP-6 and 2,4,6,2',4',6'-hexabromobiphenyl (RFR) on 2% OV-17 on Chromosorb W (AW DMCS). Column temperature, 250°; flow-rate (helium), 40 ml/min; attenuation, 10×2 . Numbers above the peaks designate the number of bromine atoms per biphenyl nucleus.

TABLE IV

SURVEY OF LITERATURE DATA ON THE ANALYSIS OF PBB MIXTURES

Composition: m, major component; t, traces; [], unresolved. Analysis: 1, GC-MS; 2, GC; 3, FID; 4, EC; 5, ¹H-NMR; 6, ¹³C-NMR; 7, TLC.

PBB mixture (manufacturer)	Wt.-% of Br	Melting point range (°C)	Methods of analysis	Wt.-%**										Remarks	Reference
				Br ₁₀	Br ₉	Br ₈	Br ₇	Br ₆	Br ₅	Br ₄					
fireMaster BP-6* (Michigan Chem. Corp.)	75	72-?	1,4 2			[14]	13.8	62.8	10.6	2			11.4% other PBBs ≥ 6 minor components 5 peaks	45 13 14	
Lot 03271			1,4,7 1,3,5		[]	[]	m	[70]					m ≡ 2,4,5,2',4',5'- (m.p. 128-130°) Traces of Br ₄ -Br ₆ naphthalenes	8 30 46	
Lot RP-158 (1971)			1,4				12.5	72.5 (3)	9	4			Br ₆ :55, 10, 7.5%	15	
Lot 6244A (1974)			1,4				13	77.5 (3)	5	4.5			Br ₆ :62, 11, 4.5%	15	
Lot 6244A			1,6		(1?)	(2)	(4)	(2)					≥ 11 other components m ≡ 2,4,5,2',4',5'- 2Br ₅ + 3Br ₆ + 1Br ₇ = 98% 73% hexa- ≡ 2,4,5,2',4',5'- (m.p. 159-161°)	17	
Octabromobiphenyl XN-1902 (Dow Chemical)	82	200-250		5.7 6	47.4 60	45.2 33	1.8 1							22, 23 20	
Bromikal (Chem. Fabrik Kalk)	81-82.5	220-290												33	
80-9D 80			1,3	m t		72 (3)	27	1					Br ₆ :14, 16, 42%	42	
Decabromobiphenyl HFO 101 (Hexcel)	84	380												34	
Adine 0102 (Péchiney)	83-85	370-375											Technical quality	35	

* fireMaster FF-1 = fireMaster BP-6 plus added anti-caking agent¹⁵.

** No. of isomers in parentheses.

ucts. Scrutiny of Table IV reveals that, apart from information provided by the manufacturers, there is a conspicuous lack of information for all PBB mixtures except fireMaster BP-6. As for the widely diverging results of the analyses of this mixture, one should bear in mind that according to Ruzo (as quoted in ref. 30) the exact composition of the product seems to vary between batches, and also within each batch according to the sampling method. Because, moreover, the response factors for the individual PBBs, whether with flame-ionization, electron capture or UV detection, are usually not known accurately and only one batch of a product is usually at one's disposal, the data given in Table IV as well as those obtained by the present authors should be considered as rough values.

"Hexa" mixtures

Gas chromatograms for fireMaster BP-6 and a commercial sample of 2,4,6,2',4',6'-hexabromobiphenyl are compared in Fig. 2, while the relative merits of GC, HPLC and TLC as methods of analysis with fireMaster BP-6 are demonstrated in Fig. 3. UV absorption spectra of the various samples of 2,4,6,2',4',6'-hexabromobiphenyl, fireMaster BP-6 and its main component were found to be virtually identical with $\lambda_{\text{max.}} = 217\text{--}219\text{ nm}$ and $\log \epsilon = 4.7\text{--}4.8$, a k band at around 240–250 nm being conspicuously absent.

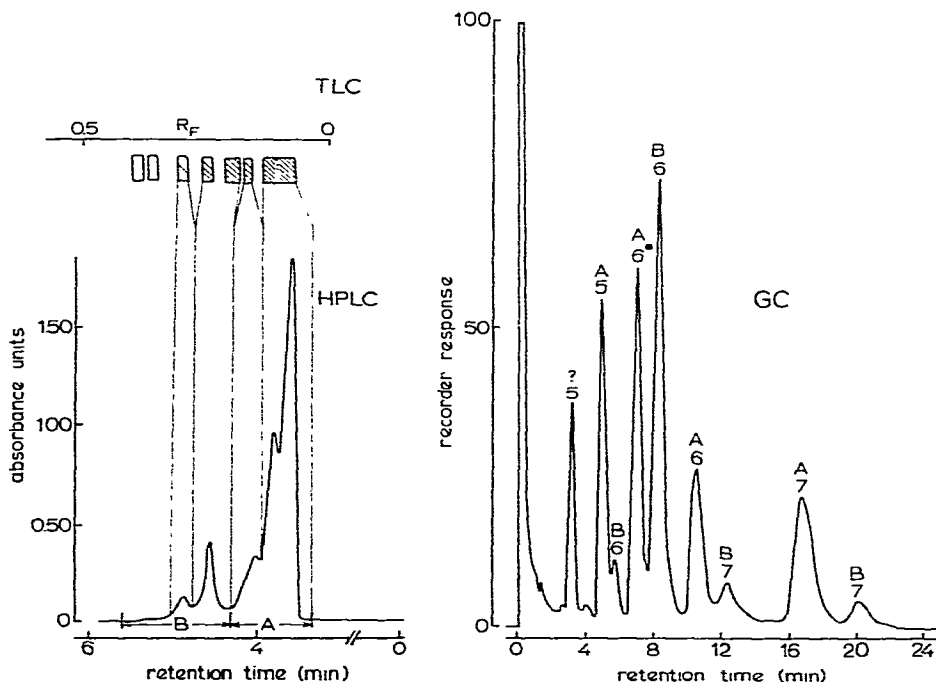


Fig. 3. Comparison of the behaviour of fireMaster BP-6 in reversed-phase TLC, HPLC and GC. TLC: Kieselguhr impregnated with paraffin oil/acetonitrile-methanol-water (8:9:3). HPLC: silica gel-dry *n*-hexane; flow-rate, 1.4 ml/min; temperature, $27 \pm 1^\circ$; detection at 217 nm. GC: 1% SE-30 on Chromosorb W (HP); column temperature, 235° ; flow-rate (helium), 18 ml/min; attenuation, 10×4 . A and B denote fractions collected in HPLC and analysed by GC. Chromatograms displayed in the figure are for crude fireMaster BP-6 (HPLC and TLC) and fraction enriched in minor components (GC). Asterisk denotes position of main fireMaster component in GC.

In order to facilitate peak assignment in GC and HPLC, the main component of fireMaster BP-6 was recrystallized⁴² from carbon tetrachloride, while a fraction enriched in the minor components was obtained by evaporation of the mother liquor. The latter fraction was subjected to GC-MS in order to determine the bromine number of minor peaks. It was also used in an HPLC fractionation experiment, in which two fractions (termed A and B in Fig. 3) were collected in amounts sufficient to allow their analysis by GC. In this experiment, development was carried out at *ca.* 0.5 instead of the normally used 1.4 ml·min⁻¹; this allowed us to discern 11 peaks in the high-performance liquid chromatogram. In TLC, much the same separation as that shown in Fig. 3 was obtained when the present mobile phase was replaced with methanol-acetonitrile-acetone-water (20:20:9:1), although the range of *R_F* values was noticeably higher in the latter instance.

In both HPLC and GC, the main component of fireMaster BP-6 was found to be identical with the sample of 2,4,5,2',4',5'-hexabromobiphenyl provided by Dr. Norström as well as with the main component of the two commercial samples of 2,4,6,2',4',6' hexabromobiphenyl at our disposal. The unequivocal and decisive proof of the identity of the 2,4,*x*,2',4',*x*'-hexabromobiphenyls was obtained through ¹H- and ¹³C-NMR measurements, our experimental values agreeing fully with those reported by Jacobs *et al.*¹⁷ and Sundström *et al.*³⁰, respectively. In conclusion, 2,4,5,2',4',5'-hexabromobiphenyl is the main component of fireMaster BP-6 as well as the 2,4,6,2',4',6'-hexabromobiphenyl samples, all of which, in addition, contain a large proportion of hepta-substituted bromobiphenyls. The combined data in Tables IV and V suggest the following average composition of fireMaster BP-6: penta-, 5–10%; hexa-, 65–80%; hepta-bromobiphenyl, 10–20%; remainder, 0–5%. Aldrich's 2,4,6,2',4',6'-

TABLE V

COMPOSITION OF PBB MIXTURES

For information on origin of samples and for experimental conditions, see text.

Sample	Main band		Wt.-%*					
	λ_{max} (nm)	Log ϵ (l·mole ⁻¹ ·cm ⁻¹)	Br ₁₀	Br ₉	Br ₈	Br ₇	Br ₆	Br ₅
2,4,6,2',4',6' (RFR)	217	4.74			(3)	12 (2)	84 (3)	1 (2)
2,4,6,2',4',6' (Aldrich)	217	—			2 (3)	24 (3)	73 (4)	4 (2)
"Hexabromobiphenyl" (RFR)	217	4.76			(3)	25 (3)	67 (4)	4 (2)
fireMaster BP-6 (Michigan)	219	4.70			1 (3)	18 (3)	73 (4)	8 (2)
2,3,5,6,2',3',5',6' (RFR)	224	4.85	1	28 (1)	46 (3)	23 (1)	2 (1)	
"Octabromobiphenyl" (RFR)	225	4.83	4	54 (1)	38 (3)	2 (1)	(1)	
Octabromobiphenyl XN-1902 (Dow)	225	—	2	34 (1)	57 (3)	7 (1)	(1)	
Bromkal 80-9D (Kalk)	224	4.99	9	65 (1)	25 (3)	1 (1)		
Decabromobiphenyl (RFR)	228	—	71	11 (1)	7 (2)	4 (1)	4 (3)	
Decabromobiphenyl HFO 101 (Hexcel)	227	5.11	96	2 (1)			(1)	
Decabromobiphenyl Adine 0102 (Péchiney)	227	—	96	4 (1)				

* No. of isomers in parentheses.

hexabromobiphenyl has virtually the same composition, whereas the sample obtained from RFR contains distinctly less penta- and hepta-substituted PBBs.

Four further conclusions merit attention. (1) The greater part of the gas chromatograms for firemaster BP-6 recorded in the literature reveal the presence of a limited number of peaks. The present data confirm the assignments made by Jacobs *et al.*¹⁷, and extend these by attributing bromine numbers to three then unknown peaks (a, b and c in Fig. 1 in ref. 17). Moreover, they demonstrate the presence of three more peaks due to octabromobiphenyls. (2) Non-*ortho*-substituted 3,4,5,3',4',5'-hexabromobiphenyl is absent from all samples under investigation, as became evident from both HPLC and GC-MS data. (3) Figs. 2 and 3 show that, at least for the present chromatographic systems, the dependence of retention on the number of bromine substituents is more strictly adhered to in GC than it is in HPLC. (4) Comparison of the straight-phase HPLC and reversed-phase TLC chromatograms in Fig. 3 reveals that a strict relationship exists between the R_F sequence and the order of elution times, as has also been found to apply for several highly chlorinated biphenyl mixtures³⁷. On the basis of the earlier discussion³⁷, it can be suggested that analysis of highly brominated biphenyl mixtures by reversed-phase HPLC will yield an improved resolution (also see refs. 43 and 44).

"Octa" to "deca" mixtures

Gas chromatograms of 2,3,5,6,2',3',5',6'-octabromobiphenyl, octabromobiphenyl XN-1902 and Bromkal 80-9D are shown in Fig. 4.

In HPLC on silica gel, elution with dry *n*-hexane at a flow-rate of *ca.* 1.4 ml/min yielded only a single peak in the chromatograms of all mixtures chiefly containing octa-substituted and/or higher substituted PBBs, the retention times varying between 3.75 and 3.85 min (*cf.*, Table II). The UV absorption characteristics of 2,3,5,6,2',3',5',6'-octabromobiphenyl, a nona-substituted PBB (see below) and decabromobiphenyl have already been recorded in Table I. Virtually identical results were obtained for the octabromobiphenyl mixtures, Bromkal 80-9D and the commercial decabromobiphenyl samples, respectively (*cf.*, Table V).

Comparison of the retention times of the various peaks of the sample of 2,3,5,6,2',3',5',6'-octabromobiphenyl and fireMaster BP-6 reveals that the only hexa-substituted PBB present in the former compound is 2,4,5,2',4',5'-hexabromobiphenyl. Moreover, it is probable that the hepta-substituted isomer identified in 2,3,5,6,2',3',5',6'-octabromobiphenyl is identical with the second largest component of fireMaster BP-6. As for the octa-substituted isomers, three separate peaks are present in each of the "octa" mixtures available, the same peak being the main one in all instances. ¹H-NMR spectra recorded in our laboratory for the sample of 2,3,5,6,2',3',5',6'-octabromobiphenyl as well as for octabromobiphenyl (RFR) and Bromkal 80-9D demonstrate that the resonance line at 7.37 δ (parts per million from tetramethylsilane) can be attributed to the main octa-substituted isomer (see below). Because this line has a singlet character, one can conclude that the major octa-substituted PBB is either 2,3,4,5,2',3',4',5'-, 2,3,4,6,2',3',4',6'- or 2,3,5,6,2',3',5',6'-octabromobiphenyl. Of these, the first and/or second are to be preferred if one assumes that the component was formed by further bromination of the major constituent of a mixture such as fireMaster BP-6, *i.e.*, 2,4,5,2',4',5'-hexabromobiphenyl. This is the more likely because an unambiguous synthesis of 2,3,5,6,2',3',5',6'-octabromobiphenyl

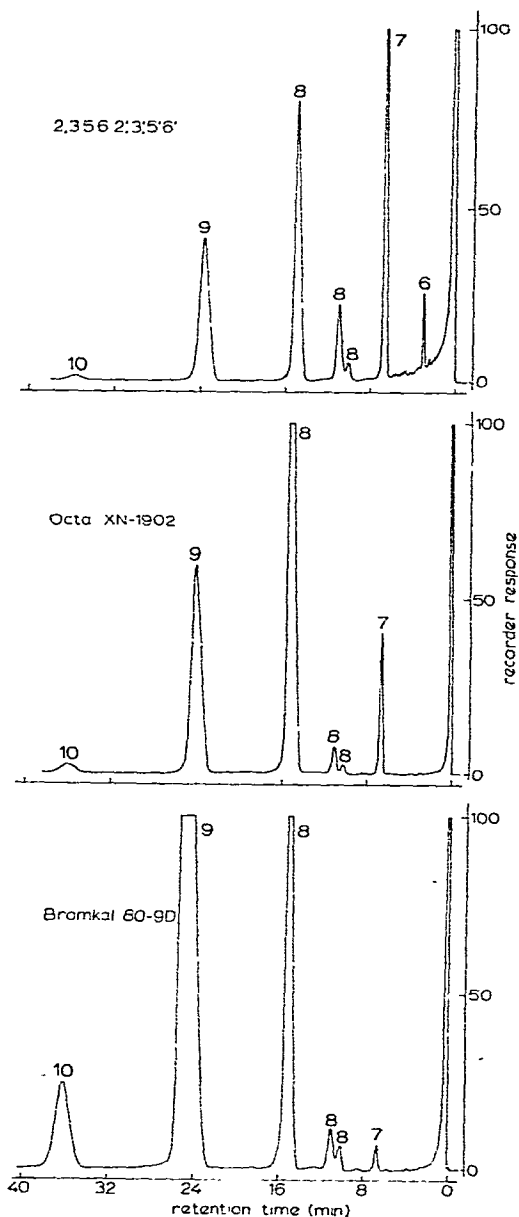


Fig. 4. GC of 2,3,5,6,2',3',5',6'-octabromobiphenyl, octabromobiphenyl XN-1902 and Bromkal 80-9D on 1% SE-30 on Chromosorb W (HP). Column temperature, 250°; flow-rate (helium), 30 ml/min; attenuation, 10 × 4. Numbers above the peaks designate the number of bromine atoms per biphenyl nucleus.

seems to be excluded in view of its overall composition. Since our suggestion contradicts the assignment of structure provided by the manufacturers, a definite conclusion cannot be drawn as yet.

From Fig. 4, it is evident that the major components of the "octa" mixtures are also the main constituents of Bromkal 80-9D. Repeated crystallization from dichloromethane allowed us to obtain a fraction enriched (88%) in nona-substituted PBB(s) and having a melting-point of 280–285°; its UV and HPLC characteristics are included in Tables I and II, respectively. The $^1\text{H-NMR}$ spectrum of the "nona" fraction, which current studies by $^{13}\text{C-NMR}$ spectrometry suggest to be the 2,3,4,5-, 6,2',3',4',5'-substituted isomer, displays a singlet resonance line at 7.45 δ . As crude Bromkal 80-9D yields main resonance lines at 7.37 and 7.45 δ (both singlets), one can conclude that the 7.37 δ line is due to the principal octa-substituted isomer, which also is a main component of the "octa" mixtures discussed above.

As for the overall composition of the various mixtures (*cf.*, Table V), one should note that three of the five "octa" samples analysed to-date contain a larger proportion of nona- than of octa-substituted PBBs. The present calculation of the composition of Bromkal 80-9D agrees well with the manufacturer's data as regards the nature of the main component, *i.e.*, a nona-substituted PBB. However, it is widely divergent from that recorded by Norström *et al.*⁴², who reported the presence of only trace amounts of nona-brominated biphenyls. However, these workers identified three octa-substituted isomers in a weight ratio which is analogous to that observed for all of the mixtures depicted in Fig. 4. A possible explanation may lie in the fact that Norström *et al.*⁴² used the term "Bromkal 80" rather than "Bromkal 80-9D"; *i.e.*, the composition of their PBB mixture may have been different from that used in our study. With the fully brominated products, the samples obtained from Hexcel and Péchiney were found to be very pure preparations, while the RFR sample contained considerable amounts of lower (nona- to hexa-) substituted isomers.

Photodegradation

In the preceding sections, it has been shown that HPLC in the system silica gel-dry *n*-hexane is not suitable for the analysis of mixtures of highly brominated biphenyls; however, with lower substituted PBBs, efficient resolution occurs. Therefore, several experiments were performed in order to test the applicability of HPLC in photodegradation studies. Because it has been reported^{26,27} that decomposition of PBBs dissolved in *n*-hexane and irradiated at 300 nm chiefly occurs through debromination, the major products arising from loss of bromine atoms in the position *ortho* to the phenyl-phenyl bond, as an additional advantage, multiple-wavelength detection will help to discriminate between *ortho*- and non-*ortho* substituted products (*k* band). Our data agree fully with those of Ruzo *et al.*²⁷: 4,4'-di- and 4- and 3-mono-bromobiphenyl are among the products invariably detected after irradiation of the highly brominated mixtures, whereas 2-mono- and 2,2'-dibromobiphenyl are notably absent. As an illustrative example, a series of chromatograms of a solution of fireMaster BP-6 in *n*-hexane, irradiated at 300 nm for 0, 10 and 60 min, and recorded at 217, 240 and/or 261 nm, is shown in Fig. 5. The group of peaks that have a retention time of *ca.* 2 min is probably due to the formation of polybrominated benzenes.

Lastly, comparison of gas chromatograms of irradiated fireMaster BP-6 published by Erney²⁶ and the present GC data strongly suggests that peak no. 5 in Fig. 1 in ref. 26 is identical with that of one of the two pentabromobiphenyls identified by us in crude fireMaster BP-6 (Fig. 2). Combining Erney's statement that this peak is the only one to display increased intensity in the initial stages of irradiation,

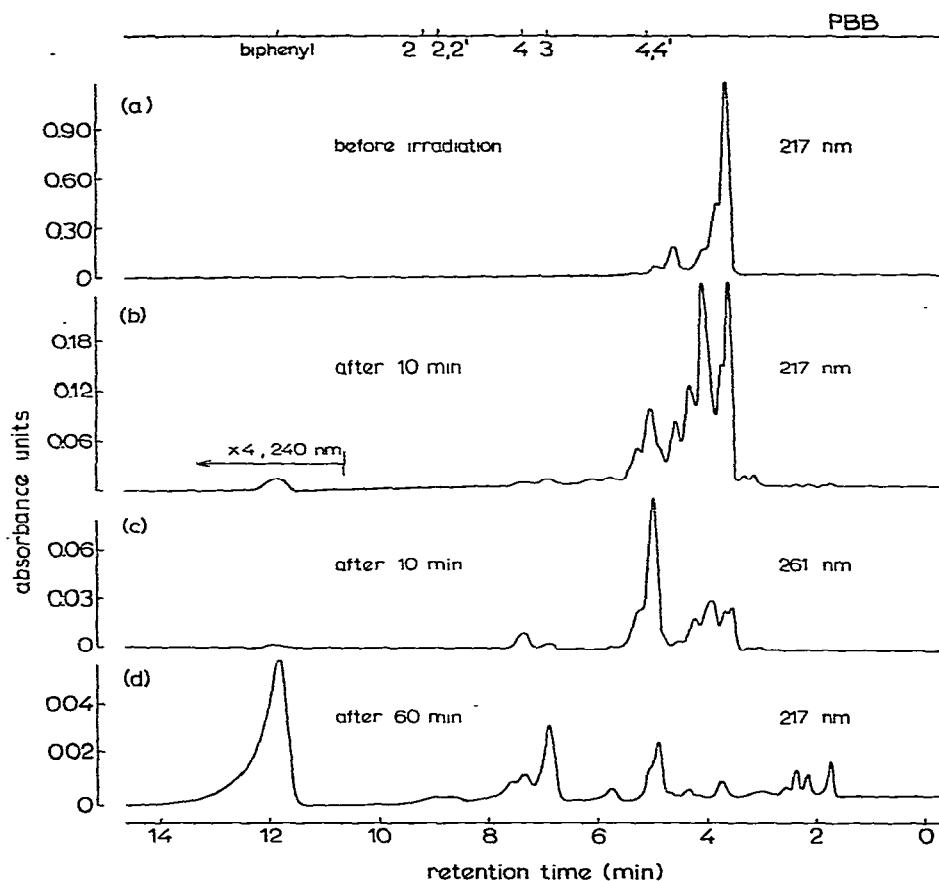


Fig. 5. HPLC in the system silica gel-dry *n*-hexane (flow-rate, 1.4 ml/min; temperature, $27 \pm 1^\circ$; UV detection as indicated) of fireMaster BP-6 before (a) and after irradiation with a 100-W mercury lamp for 10 (b, c) and 60 min (d). The solutions in *n*-hexane (1000 ppm) were irradiated at ambient temperature in cells with quartz windows placed at a distance of *ca.* 10 cm from the light source.

the by now well established phenomenon of preferential loss of *ortho*-bromine atoms in photodegradation experiments, and the fact that 2,4,5,2',4',5'-hexabromobiphenyl is the main constituent of fireMaster BP-6, we tentatively identify one of the penta-substituted isomers as 2,4,5,3',4'-pentabromobiphenyl.

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