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Elution order of the 209 polychlorinated biphenyls on a high-temperature capillary column

Walter Vetter[a,*], Bernd Luckas[a], Jan Buijten[b]

[a]*Friedrich-Schiller-Universität Jena, Institut für Ernährung und Umwelt, Dornburger Strasse 25, D-07743 Jena, Germany*
[b]*Chrompack International, Research and Development, P.O. Box 8033, NL-4330 E Middelburg, Netherlands*

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Abstract

Retention times of the 209 polychlorinated biphenyls (PCBs) were determined by gas chromatography in combination with electron ionization mass spectrometry on a newly developed stationary phase which was suitable to 380°C. The so-called Thermocap A phase consisted of 50% methyl–50% phenyl polysiloxane which was prepared by using an in situ process. On Thermocap A, the sum of the McReynolds constants was five times lower than on conventional equimolar methyl–phenyl phases distributed with the tradenames CP-Sil 24 or DB-17. The Thermocap A column showed some unique elution orders of PCB congeners. For example, PCB 133 eluted in front of PCB 136 which was not reported on any other stationary phase before. Most of the PCB congeners which are part of national regulations in Europe eluted without interference from the Thermocap A phase. The changed elution order and the high-temperature stability recommends the use of the Thermocap A phase for PCB analyses alone or as a confirmatory column. © 1998 Elsevier Science B.V.

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1. Introduction

Polychlorinated biphenyls (PCBs) are highly stable industrial products [1]. A variety of 209 PCB congeners is possible, and about 140 have been detected in different technical mixtures (popular tradenames are Aroclor, Clophen, and Kanechlor) [2]. Properties of PCB congeners are largely dependent on the number of *ortho*-substituents. Expressed in simple terms, non-*ortho* and mono-*ortho*-PCBs are the most toxic ones [3], di-*ortho*-PCBs are often highly abundant in biological samples, tri-*ortho*- and tetra-*ortho*-PCBs may exist as stable atropisomers [4].

Once introduced into the environment, PCBs resist biological degradation and accumulate in higher organisms of the food chains. In awareness of the toxicity of PCBs residue limits have been introduced in many countries. In Germany, the Netherlands and other countries, PCB levels in food are monitored by quantitation of selected PCB congeners [5,6]. For a better understanding of the impact of PCBs on the environment congener specific analyses are carried out using high-resolution gas chromatography (GC).

Due to the many PCB congeners in environmental samples and technical mixtures, coelutions cannot be excluded on any stationary phase. On the ''classical''

*Corresponding author. Tel.: +49 3641 949657; Fax: +49 3641 949652; E-mail: b5wave@rz.uni-jena.de

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GC phase for PCB separations, SE-54 (94% methyl–5% phenyl–1% vinyl), several major abundant and particularly toxic PCBs coeluted with other PCB congeners [7–9]. Therefore, alternative phases were suggested to substitute for the ''classical'' GC phase for PCB separations [10–14].

Stationary GC phases with high temperature stability also received attention during the last years. We have analyzed all 209 PCBs on the newly developed Thermocap A phase and investigated the suitability of this high temperature column for the separation of PCB congeners.

2. Experimental

2.1. Characterization of the stationary phase of Thermocap A

The Thermocap A column was a prototype of a phase developed by Chrompack (Middelburg, Netherlands). Thermocap A is an in situ prepared, high-temperature stable, siloxane-based stationary phase with 50% methyl and 50% phenyl substituents. Due to the nature of the in situ process, the Thermocap A is extremely stable from a mechanical and a temperature point of view [15].

A GC stationary phase is usually characterized with the McReynolds constants benzene (x'), 1-butanol (y'), 2-pentanone (z'), 1-nitropropane (u'), and pyridine (s') at 120°C [16]. The P-value which is the sum of the retention indices of the McReynolds probes of the respective phase relative to squalane ($P=x'+y'+z'+u'+s'$) was 179 for Thermocap A. The P-value of the Thermocap A ($P=222$) was significantly lower than the P-value of conventional 50% methyl–50% phenyl polysiloxane phases such as CP-Sil 24 ($P=1000$) or OV-17 ($P=884$). Occasionally, P-values are considered as the general polarity of a stationary phase [16]. Yet, this principle cannot be applied to the Thermocap A phase since its P-value of 179 is typical of nonpolar 100% methylpolysiloxane phases such as CP-Sil 5 or DB-1 ($P=217$). However, this example enlightens the peculiarity of the new Thermocap A phase. Expressed in terms of the Chrompack scale (relative to OV-275≡100.0), the CP-Sil value (Chrompack scale) was 4.3 [15].

The retention volume for dodecane (at 140°C) of the Thermocap A phase was more than 10 times higher than on CP-Sil 5 (100% dimethyl polysiloxane) [15] and therefore, a very thin film thickness (0.05 μm) was chosen for the present investigation. The column parameters were 60 m×0.25 mm I.D. with 0.05 μm film thickness.

2.2. GC–electron ionization mass spectrometry (GC–EI-MS)

GC–EI-MS was performed on an HP 5890 gas chromatograph in combination with an HP 5971 MSD mass spectrometer (Hewlett–Packard, Waldbronn, Germany). An HP 7673 autosampler (Hewlett–Packard) was used for splitless injections (1 min, injector temperature: 270°C). The carrier gas helium was used at a column head pressure of 1.1 bar (linear velocity at 120°C was 23.5 cm/s). The transfer line to the mass spectrometer was set at 280°C.

The GC oven temperature program followed the general suggestions for the analysis of PCBs with a fast heating rate to a temperature around 150–170°C, followed by a slow heating rate [2]. After injection at 80°C (1 min) the oven was heated at 15°C/min to 165°C, then by a rate of 1.5°C/min to 290°C, and a rate of 25°C/min to the final temperature 380°C which was held for 1.4 min. The total run time was 95 min.

In the present study we use the IUPAC nomenclature for single PCB congeners which applies the system introduced by Ballschmiter and Zell [17] in a slightly modified form [18] (see Table 1). For more information, we additionally present in brackets two numbers separated by an oblique as recently suggested [10]. The first number reflects the degree of chlorination, and the second number after the oblique stands for the number of *ortho*-chlorine atom of the respective PCB congener. For example, PCB 153 (6/2) means that PCB 153 is a hexachlorobiphenyl with two *ortho*-substituents [10].

In the full scan mode, mass chromatograms were recorded 20 min after injection within a mass range of m/z 150–440. The threshold was set at 100 and the electron multiplier voltage at 1894 V. Scan time was 2.4 scans/s. PCB 37 (3/0), PCB 77 (4/0), PCB 126 (5/0), PCB 169 (6/0), PCB 189 (7/1), and

Table 1
Structure and retention times of the PCB congeners on Thermocap A; for GC conditons see Section 2. European regulation relevant PCB indicator congeners [5,6] are printed in bold face

Congener	Cl substitution on ring a–ring b	t_R (min)	Congener	Cl substitution on ring a–ring b	t_R (min)
1 (1/1)	2–0	28.58	47 (4/2)	24–24	52.42
2 (1/0)	3–0	30.66	65 (4/2)	2356–0	52.66
3 (1/0)	4–0	30.66	43 (4/2)	235–2	52.74
10 (2/2)	26–0	36.05	62 (4/2)	2346–0	52.79
7 (2/1)	24–0	36.40	68 (4/1)	24–35	52.98
9 (2/1)	25–0	36.53	48 (4/2)	245–2	53.17
4 (2/2)	2–2	36.58	45 (4/3)	236–2	53.20
14 (2/0)	35–0	37.84	80 (4/0)	35–35	53.27
8 (2/1)	2–4	38.35	57 (4/1)	235–3	54.40
6 (2/1)	2–3	38.37	59 (4/2)	236–3	54.56
5 (2/1)	23–0	39.33	46 (4/3)	23–26	54.67
11 (2/0)	3–3	40.62	63 (4/1)	235–4	54.92
13 (2/0)	3–4	40.82	67 (4/1)	245–3	54.94
12 (2/0)	34–0	41.03	44 (4/2)	23–35	54.95
15 (2/0)	4–4	41.28	42 (4/2)	23–24	55.09
30 (3/2)	246–0	41.87	64 (4/2)	236–4	55.12
19 (3/3)	26–2	44.40	74 (4/1)	245–4	55.37
17 (3/2)	24–2	44.42	71 (4/2)	26–34	55.84
18 (3/2)	25–2	44.59	58 (4/1)	23–35	55.85
23 (3/1)	235–0	45.05	70 (4/1)	25–34	56.16
24 (3/2)	236–0	45.25	61 (4/1)	2345–0	56.18
34 (3/1)	35–2	45.34	41 (4/2)	234–2	56.26
29 (3/1)	245–0	45.45	104 (5/4)	246–26	56.61
27 (3/2)	26–3	45.79	103 (5/3)	246–25	56.75
32 (3/2)	26–4	45.95	66 (4/1)	24–34	56.76
26 (3/1)	25–3	46.18	121 (5/2)	246–35	57.07
25 (3/1)	24–3	46.35	76 (4/1)	345–2	57.20
31 (3/1)	25–4	46.45	100 (5/3)	246–24	57.25
28 (3/1)	**24–4**	**46.82**	79 (4/0)	34–35	57.44
16 (3/2)	23–2	47.31	40 (4/2)	23–23	58.02
36 (3/0)	35–3	47.46	55 (4/1)	234–3	58.11
39 (3/0)	35–4	47.91	60 (4/1)	234–4	58.78
21 (3/1)	234–0	48.52	94 (5/3)	235–26	59.20
33 (3/1)	34–2	48.78	78 (4/0)	345–3	59.21
20 (3/1)	23–3	49.35	92 (5/2)	235–25	59.27
22 (3/1)	23–4	49.61	56 (4/1)	23–34	59.47
50 (4/3)	246–2	49.74	98 (5/3)	2446–23	59.69
38 (3/0)	345–0	49.89	111 (5/1)	235–35	59.73
35 (3/0)	34–3	51.10	93 (5/3)	2356–2	59.75
69 (4/2)	246–3	51.26	96 (5/4)	236–26	59.78
54 (4/4)	26–26	51.67	95 (5/3)	236–25	59.86
37 (3/0)	34–4	51.71	81 (4/0)	345–4	59.87
52 (4/2)	**25–25**	**51.82**	90 (5/2)	235–24	59.88
75 (4/2)	246–4	51.86	102 (5/3)	245–26	59.94
53 (4/3)	25–26	51.92	**101 (5/2)**	**245–25**	**59.99**
51 (4/3)	24–26	51.97	88 (5/3)	2346–2	60.10
49 (4/2)	24–25	52.08	91 (5/3)	236–24	60.23
73 (4/2)	26–35	52.23	113 (5/2)	236–35	60.34
72 (4/1)	25–35	52.37	120 (5/1)	245–35	60.43

(Cont.)

Table 1. Continued

Congener	Cl substitution on ring a–ring b	t_R (min)	Congener	Cl substitution on ring a–ring b	t_R (min)
99 (5/2)	245–24	60.58	134 (6/3)	2356–23	68.83
119 (5/2)	246–34	61.09	143 (6/3)	2345–26	69.07
155 (6/4)	246–246	61.23	142 (6/3)	23456–2	69.09
112 (5/2)	2356–3	61.25	141 (6/2)	2345–25	69.12
77 (4/0)	34–34	61.42	131 (6/3)	2346–23	69.33
109 (5/2)	2346–3	61.62	188 (7/4)	2356–246	69.46
117 (5/2)	2356–4	62.00	159 (6/1)	2345–35	69.65
115 (5/2)	2346–4	62.42	137 (6/2)	2345–24	70.13
83 (5/2)	235–23	62.58	184 (7/4)	2346–246	70.20
116 (5/2)	23456–0	62.60	163 (6/2)	2356–34	70.30
84 (5/3)	236–23	62.85	130 (6/2)	234–235	70.41
89 (5/3)	234–26	62.88	162 (6/1)	235–245	70.45
87 (5/2)	234–25	63.11	132 (6/3)	234–236	70.46
97 (5/2)	245–23	63.24	160 (6/2)	23456–3	70.55
86 (5/2)	2345–2	63.25	158 (6/2)	2346–34	70.82
125 (5/2)	345–26	63.44	164 (6/2)	236–245	71.02
148 (6/3)	235–246	63.56	**138 (6/2)**	**234–245**	**71.11**
124 (5/1)	345–25	63.67	167 (6/1)	245–345	71.23
85 (5/2)	234–24	63.76	178 (7/3)	2356–235	71.65
107 (5/1)	235–34	63.90	166 (6/2)	23456–4	71.68
108 (5/1)	234–35	64.06	179 (7/4)	2356–236	72.18
150 (6/4)	236–246	64.11	175 (7/3)	2346–235	72.45
110 (5/2)	236–34	64.22	129 (6/2)	2345–23	72.52
82 (5/2)	234–23	64.46	187 (7/3)	2356–245	72.75
123 (5/1)	345–24	64.51	176 (7/4)	2346–236	72.96
118 (5/1)	**245–34**	**64.56**	182 (7/3)	2345–246	73.16
106 (5/1)	2345–3	64.88	183 (7/3)	2346–245	73.48
154 (6/3)	245–246	64.55	156 (6/1)	2345–34	74.03
152 (6/4)	2356–26	65.40	186 (7/4)	23456–26	74.16
151 (6/3)	2356–25	65.62	185 (7/3)	23456–25	74.21
114 (5/1)	2345–4	65.68	128 (6/2)	234–234	74.49
165 (6/2)	2356–35	65.94	192 (7/2)	23456–35	74.82
133 (6/2)	235–235	65.99	157 (6/1)	234–345	75.12
145 (6/4)	2346–26	66.05	169 (6/0)	345–345	75.19
144 (6/3)	2346–25	66.27	181 (7/3)	23456–24	75.34
127 (5/0)	345–35	66.43	172 (7/2)	2345–235	75.47
147 (6/3)	2356–24	66.53	174 (7/3)	2345–236	75.89
161 (6/2)	2346–35	66.57	177 (7/3)	2356–234	76.04
135 (6/3)	235–236	66.67	**180 (7/2)**	**2345–245**	**76.21**
146 (6/2)	235–245	66.81	193 (7/2)	2356–2356	76.24
139 (6/3)	2346–24	66.91	202 (8/4)	2356–2356	76.57
136 (6/4)	236–236	66.94	171 (7/3)	2346–234	76.87
122 (5/1)	345–23	67.21	191 (7/2)	2346–345	77.00
149 (6/3)	236–245	67.49	201 (8/4)	2346–2356	77.63
153 (6/2)	**245–245**	**67.64**	173 (7/3)	23456–23	77.66
140 (6/3)	234–246	67.65	204 (8/4)	23456–246	77.94
168 (6/2)	246–345	68.03	197 (8/4)	2346–2346	78.55
105 (5/1)	234–34	68.13	190 (7/2)	23456–34	79.25
126 (5/0)	345–34	68.76	198 (8/3)	23456–235	79.91

Table 1. Continued

Congener	Cl substitution on ring a–ring b	t_R (min)	Congener	Cl substitution on ring a–ring b	t_R (min)
189 (7/1)	2345–345	79.96	194 (8/2)	2345–2345	84.43
170 (7/2)	2345–234	79.97	195 (8/3)	23456–234	84.52
200 (8/4)	23456–236	80.27	205 (8/20	23456–345	84.54
199 (8/3)	2345–2356	80.57	207 (9/4)	23456–2346	85.33
103 (8/3)	23456–245	80.10	206 (9/3)	23456–2345	88.47
196 (8/3)	2345–2346	81.47	209 (10/4)	23456–23456	91.24
208 (9/4)	23456–2356	84.26			

further PCB congeners were used to determine the time windows later used in the selected ion monitoring (SIM) mode. From earlier data it was known that the PCB congeners mentioned above are the last eluted isomers within the respective chlorination group [2,7,19,20].

The investigations were performed by application of parameters suggested by Frame [2]. In the SIM mode, the most abundant ion was recorded for every degree of chlorination using m/z 172.0 for 2-fluorobiphenyl (internal standard 1=I.S. 1), m/z 188.0 for monochloro-PCBs, m/z 221.9 for dichloro-PCBs, m/z 255.9 for trichloro-PCBs, m/z 291.9 for tetrachloro-PCBs, m/z 325.8 for pentachloro-PCBs, m/z 359.8 for hexachloro-PCBs, m/z 395.8 for heptachloro-PCBs, m/z 429.7 for octachloro-PCBs, m/z 463.7 for nonachloro-PCBs, and m/z 497.7 for the decachloro-PCB 209 (10/4) which was used as internal standard 2 (I.S. 2).

Seven time windows with four ions respectively were recorded with a dwell time of 32 ms (5.05 cycles/s) and a multiplier voltage of 2250 V. After the solvent delay the first time window started at 21:00 min (I.S. 1, mono- to trichloro-PCBs), the second at 35.00 (di- to pentachloro-PCBs), the third at 50.00 (tri- to hexachloro-PCBs), the fourth at 60.00 (tetra- to heptachloro-PCBs), the fifth at 70.00 (penta- to octachloro-PCBs), the sixth at 80.00 (hexa- to nonachloro-PCBs), and the seventh at 86.00 (hepta- to decachloro-PCBs).

After the set up of the SIM method, 30 solutions containing 7 PCB congeners as well as 2-fluorobiphenyl (I.S. 1) and PCB 209 (I.S. 2), respectively, were injected. The standard solutions were assembled and distributed by Frame [2].

2.3. Gas chromatography–electron capture detection (GC–ECD)

Standards and samples were investigated with GC–ECD on an HP 5890 gas chromatograph (Hewlett–Packard). The samples were splitless injected at 250°C. The carrier gas was helium and the make-up gas was nitrogen. The linear velocity at 120°C was 25.7 cm/s. The detector temperature was set at 390°C. The GC oven program was the same as with GC–EI-MS except a starting temperature of 120°C was used.

3. Results and discussion

3.1. GC properties of the Thermocap A phase

As mentioned above, the Thermocap A phase consists of 50% methyl and 50% phenyl polysiloxane. Usually, the value of the McReynolds constant x' is proportional with the phenyl content of a phase [16] which was not the case for Thermocap A. So it was interesting to study if the new phase behaves as a typical ''phenyl phase'' with respect to the elution order of PCBs. Interaction of PCBs with stationary phases with phenyl substituents was already discussed by Larsen [21].

Despite the very thin film thickness, the PCB 28 (3/1) eluted at 222°C from the 60 m Thermocap A column. The high elution temperatures were responsible for the relatively low elution range of $<$63 min from PCB 1 (1/0) to PCB 209 (10/4) on Thermocap A, while CP-Sil 2 (a nonpolar phase chemically bonded and crosslinked 4% silicon-containing high-

molecular-mass hydrocarbon [22], $P=103$), CP-Sil 8/20% C_{18} [4/5 (95% methyl–5% phenyl) and 1/5 (methyl octadecyl) polysiloxane, P=ca. 300], and CP-Sil 19 (86% methyl–7% phenyl–7% cyanopropyl polysiloxane, $P=805$) had an elution range of ca. 70 min using comparable GC conditions [23].

Already first runs confirmed the stability of the new phase. In the full scan mode, the column bleed was very low even at 380°C (see Fig. 1). Although the PCBs eluted at much lower oven temperatures [the last eluted PCB 209 (10/4) left the Thermocap A phase at 320°C] the final temperature was set at 380°C. Even after ca. 100 injections the alteration of the retention times was negligible. PCB 209 (10/4) was used as test substance and 10 injections one after the other showed excellently reproducible retention times (average 91.24 min±0.05).

Table 1 lists structure and retention times of the PCB congeners on Thermocap A. The Thermocap A phase eluted PCBs largely in groups of isomers. The

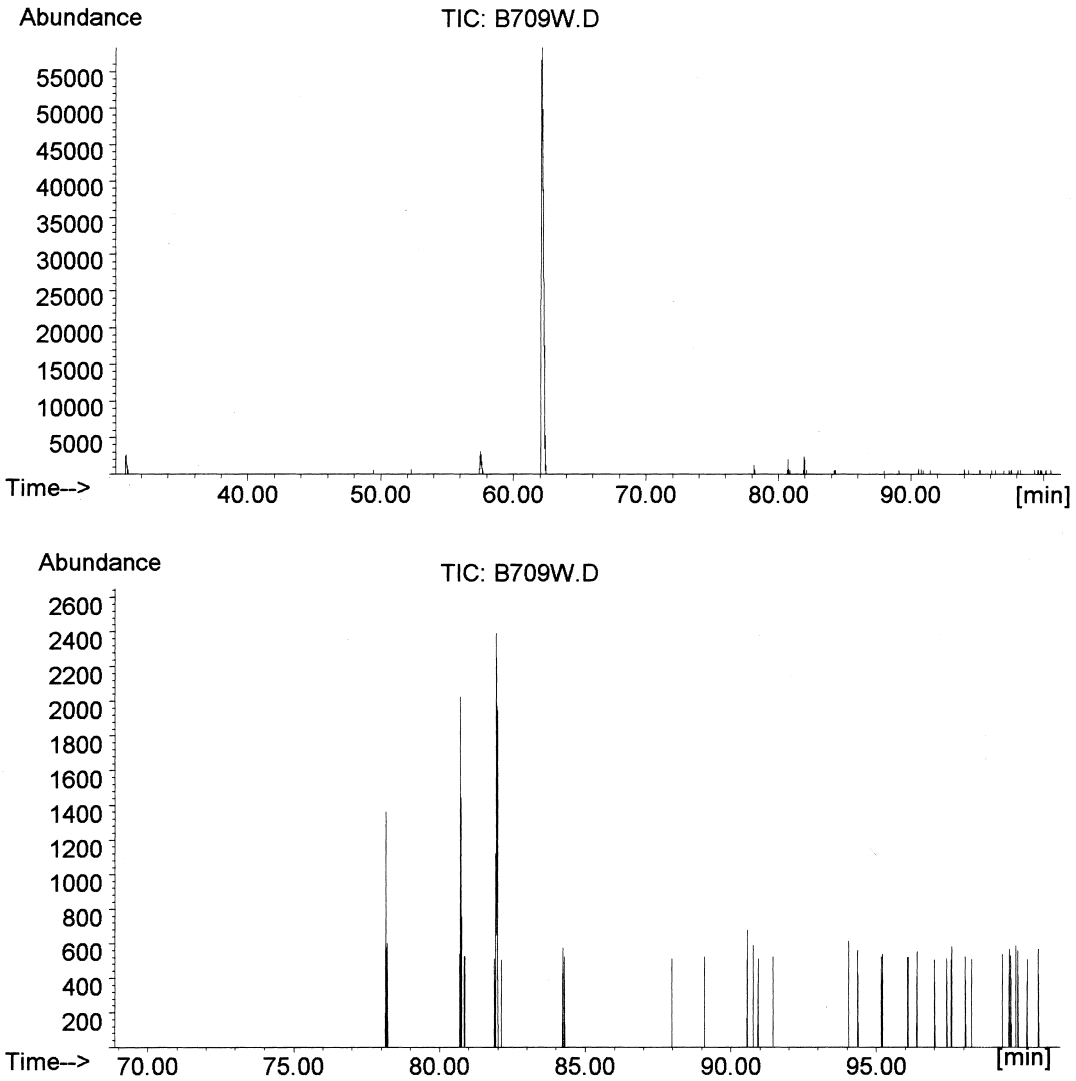

Fig. 1. GC–EI-MS full scan chromatogram (m/z 150–m/z 440) of 1,2,3,4,6,7-hexachloronaphthalene (10 ng/μl). (a) Full scan chromatogram; (b) zoomed time range from 70–95 min demonstrating the low bleed of Thermocap A (for conditions see Section 2).

retention range of isomers was lower than on other stationary phases and we observed fewer overlaps of PCBs with different degree of chlorination as compared with other stationary phases of similar or different *P*-value [24]. On the other hand, the Thermocap A phase did not elute PCB isomers in order with decreasing number of *ortho*-substituents which was observed on nonpolar stationary phases [8,19–21]. On the nonpolar CP-Sil 2 phase (this phase was earlier named CP-Select for PCBs [10,20,23,23]) no deviations from this rule were observed [20]. Recently, we investigated the influence of the PCB substitution patterns on the retention time [24]. Instead of evaluating data for all 209 PCBs, those with a 2,4,5-substitution pattern on one ring were chosen and considered as representative [23]. The PCBs with a 2,4,5-substitution pattern on one ring were selected and then, the influence of the pattern on the second ring on the retention time was studied. On the nonpolar CP-Sil 2 and CP-Sil 8/20% C_{18}, the semipolar CP-Sil 19, and the very polar CP-Sil 88 phases, PCBs with trichloro patterns on the second ring eluted in the following order [24]:

2,4,6 < 2,3,6 < 2,3,5 < 2,4,5 < 2,3,4 < 3,4,5.

This is in contrast to the Thermocap A phase which eluted 2,3,5-substituted congeners in front of 2,3,6-substituted PCBs. As a consequence, the di-*ortho*-2,3,5,2′,3′,5′-substituted PCB 133 (6/2) eluted 1 min in front of the tetra-*ortho*-2,3,6,2′,3′,6′-substituted PCB 136 (6/4). Elution of PCB 133 in front of PCB 136 is unique for GC stationary phases used for PCB separations [2,7,17,20,21,25]. Larsen reported that stationary phases with a high number of phenyl substituents particularly retain PCBs with vicinal, nonsubstituted carbons [21]. This partly explains the late elution of 2,3,6-substituted PCBs (vicinal hydrogens in the 4 and 5 position) on Thermocap A. However, in the recent collaborative study organized by Frame, none of the 16 tested stationary phases eluted PCB 133 in front of PCB 136. This multicolumn testing program also included the classical 50% methyl–50% phenyl polysiloxane (CP-Sil 24 or DB-17) phase which eluted PCB 136 in front of PCB 133. Obviously, the basic principle of phenyl-containing phases comes even more to fruition on Thermocap A.

Table 2 lists normalized retention times (*NRT*s) of the 2,4,5-substituted PCBs and compares Δ*NRT* values of neighboring PCB congeners. *NRT*s (see explanation in Table 2) were calculated on the basis of a *NRT* of 0.00 for PCB 29 (3/1) and a *NRT* of 100.00 for PCB 203 (8/3) [24]. Using *NRT*s,

Table 2
Normalized retention times of 2,4,5-substituted PCBs on Thermocap A

PCB congener	Pattern on 2nd ring	*NRT*[a]	Δ*NRT*[b]
PCB 29 (3/1)	–	0.00	
			21.65
PCB 48 (4/2)	2	21.65	
			4.97
PCB 67 (4/1)	3	26.62	
			1.21
PCB 74 (4/1)	4	27.83	
			12.82
PCB 102 (5/3)	26	40.65	
			0.13
PCB 101 (5/2)	25	40.78	
			1.24
PCB 120 (5/1)	35	42.02	
			0.42
PCB 99 (5/2)	24	42.44	
			7.46
PCB 97 (5/2)	23	49.90	
			3.68
PCB 154 (6/3)	246	53.58	
			0.02
PCB 118 (5/1)	34	53.60	
			6.32
PCB 146 (6/2)	235	59.92	
			1.90
PCB 149 (6/3)	236	61.82	
			0.42
PCB 153 (6/2)	245	62.24	
			9.74
PCB 138 (6/2)	234	71.98	
			0.33
PCB 167 (6/1)	345	72.31	
			4.27
PCB 187 (7/3)	2356	76.58	
			2.05
PCB 183 (7/3)	2346	78.63	
			7.65
PCB 180 (7/2)	2345	86.28	
			13.72
PCB 203 (8/3)	23456	100.00	

[a] *NRT* = normalized retention times calculation mode: $NRT_X = (t_R X - t_R \text{PCB } 29)/(t_R \text{PCB } 203 - t_R \text{PCB } 29) \times 100$ [23].
[b] Δ*NRT* = difference in *NRT* of two neighboring PCB congeners.

retention times on stationary phases with different polarity are more easily compared [24].

PCB isomers with 2,6- and 2,5-substitution, 3,5- and 2,4-substitution, 2,3,6- and 2,4,5-substitution, as well as 2,3,4- and 3,4,5-substitution had very similar *NRT*s ($\Delta NRT < 0.5$) on Thermocap A. On CP-Sil 2, CP-Sil 8/20% C_{18}, CP-Sil 19, and CP-Sil 88, these four pairs had $\Delta NRT > 4$ without exception [24]. Another interesting thing was that the last eluted dichloro-congener (3,4) and the first eluted trichloro-congener (2,4,6) had similar *NRT*s on Thermocap A, while ΔNRT ranged from 9 to 30 on the other stationary phases mentioned above. These examples clearly demonstrate the peculiarity of Thermocap A with regard to the elution order of PCB congeners.

3.2. Elution order of regulated and abundant PCB congeners in biota

In Europe, an important criterion for the suitability of a stationary phase for PCB analysis is the unambiguous elution of the so-called PCB indicator congeners PCB 28 (3/1), PCB 52 (4/2), PCB 101 (5/2), PCB 138 (6/2), PCB 153 (6/2), PCB 180 (7/2), and PCB 118 (5/1) which are part of national regulations [5,6]. For the PCB indicator congeners we detected the following interferences on Thermocap A: (i) PCB 52 (4/2) and PCB 75 (4/2); (ii) PCB 101 (5/2) and PCB 102 (5/3); (iii) PCB 153 (6/2) and PCB 140 (6/3); (iv) PCB 138 (6/2) and PCB 164 (6/2); (v) PCB 180 (7/2) and PCB 193 (7/2); as well as (vi) PCB 118 (5/1) and PCB 123 (5/1), PCB 82 (5/2), and PCB 154 (6/3). However, examples (i), (ii), and (v) play no role in real samples since the interfering PCB had only low abundance in technical mixtures, (iii) PCB 140 was absent in technical mixtures, and (iv) PCB 164 (6/2) is low in technical mixtures and fish [2,26,27] while it was not detected in seal blubber [10].

On the other hand, PCB 28 (3/1) and PCB 31 (3/1) were more than just baseline separated. PCB 90 (5/2) and PCB 101 (5/2) which coeluted on SE-54 [7], CP-Sil 8 [28,29], HT-8 [12], and CP-Sil 2 [10] were also separated on Thermocap A.

Fig. 2 shows the separation of PCB 163 (6/2), PCB 164 (6/2), and PCB 138 (6/2) on Thermocap A. These three PCB congeners coeluted on SE-54 [7]. PCB 163 (6/2) eluted more than 45 s in front of

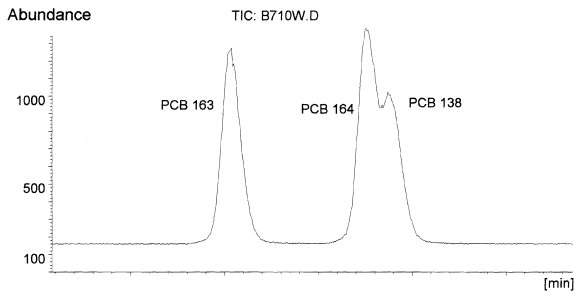

Fig. 2. GC–EI-MS separation of PCB 163 (6/2), PCB 164 (6/2), and PCB 138 (6/2) on Thermocap A (for conditions see Section 2).

PCB 138 (6/2). PCB 163 (6/2) is a major PCB congener in technical mixtures [26] and marine biota [10,30]. We did not even detect an interference of PCB 163 (6/2) on Thermocap A. PCB 99 (5/2), an abundant PCB in human milk [1] and polar bears [31], eluted without interference from the Thermocap A. A disadvantage was coelution of PCB 170 (7/2) with PCB 189 (7/1) and PCB 198 (8/3), since the first two are among the 13 PCB congeners the World Health Organization (WHO) has allocated dioxin-like toxic equivalency factors [3].

PCB 132 (6/3), an interferent on SE-54 [25], eluted long after PCB 153 (6/2) on Thermocap A (see Table 1). A higher t_R of PCB 132 (6/3) compared with PCB 153 (6/2) was also reported on other stationary phases with phenyl groups [7,21], cyanopropyl groups [24], mixtures of both [32], but not on nonpolar phases [19,24].

3.3. Toxic mono-ortho- and non-ortho-PCBs

Since Thermocap A did not separate PCB isomers according to their number of *ortho*-substituents, we expected more coelutions of these toxic PCB congeners with isomers as compared with nonpolar stationary phases. Non-*ortho*-PCBs 77 (4/0), 126 (5/0), and 169 (6/0) can barely be determined in environmental samples without preseparation from the *ortho*-substituted PCB congeners. Therefore, coelution of these three toxic PCBs with *ortho*-substituted PCB congeners is only a minor disadvantage. However, high toxic equivalency factors have also been assigned to several mono-*ortho*-PCBs

[3]. These PCB congeners (respective interferences are given in square brackets) are PCB 105 (5/1), PCB 114 (5/1) [interference: PCB 151 (6/3)], PCB 123 (5/1) [interferences: PCB 118 (5/1), PCB 82 (5/2), PCB 154 (6/3)], PCB 156 (6/1), PCB 157 (6/1) [interference: PCB 169 (6/0)], PCB 167 (6/1) and PCB 189 (7/1) [interferences: PCB 170 (7/2), PCB 198 (8/3)] [3]. Note that some of the interferences can be distinguished with GC–MS which is the common method for quantitation of toxic non-*ortho*- and mono-*ortho*-PCBs.

As already mentioned in Section 1, coelutions cannot be excluded on a single PCB column. In total we found 65 pairs of PCBs with $\Delta t_R < 0.1$ (see Table 1). For comparison, Δt_R of PCB 164 and PCB 138 was 0.09 (see Fig. 2). However, on Thermocap A these coelutions were mainly limited to less important PCB congeners.

Fig. 3 shows the GC–ECD chromatogram of the blubber extract of a female juvenile harbour seal (*Phoca vitulina*) from the North Sea (Wadden Sea, Germany). The chromatogram was dominated by the peak of PCB 153 (6/2). Note the interfering peak of PCB 149 (6/3) prior to PCB 153 (6/2). Another interesting point was the abundant peak of PCB 163 (6/2). As discussed above PCB 163 (6/2) interferes with PCB 138 (6/2) on SE-54/DB-5 (note that the ECD response of PCB 163 (6/2) was higher than the ECD response of PCB 138 (6/2) [10]). The two peaks in front of PCB 180 are caused by PCB 174 (7/3) and PCB 177 (7/3). No significant interference from PCBs was found for 1,1′-(2,2-dichloroethenylidene)-bis(4-chlorobenzene) (*p,p′*-DDE) and hexachlorocyclohexane isomers eluted prior to PCB 23 (3/1).

We used 380°C as the final oven temperature for GC–ECD investigations. The detector was set at 390°C which is just below the maximum temperature of the ECD. Such high detector temperatures support dissociate electron capture. For routine analysis with GC–ECD we suggest 350°C as the maximum temperature and a shorter Thermocap A column (50 m instead of 60 m) due to the high elution temperatures and the run times on the present 60 m column.

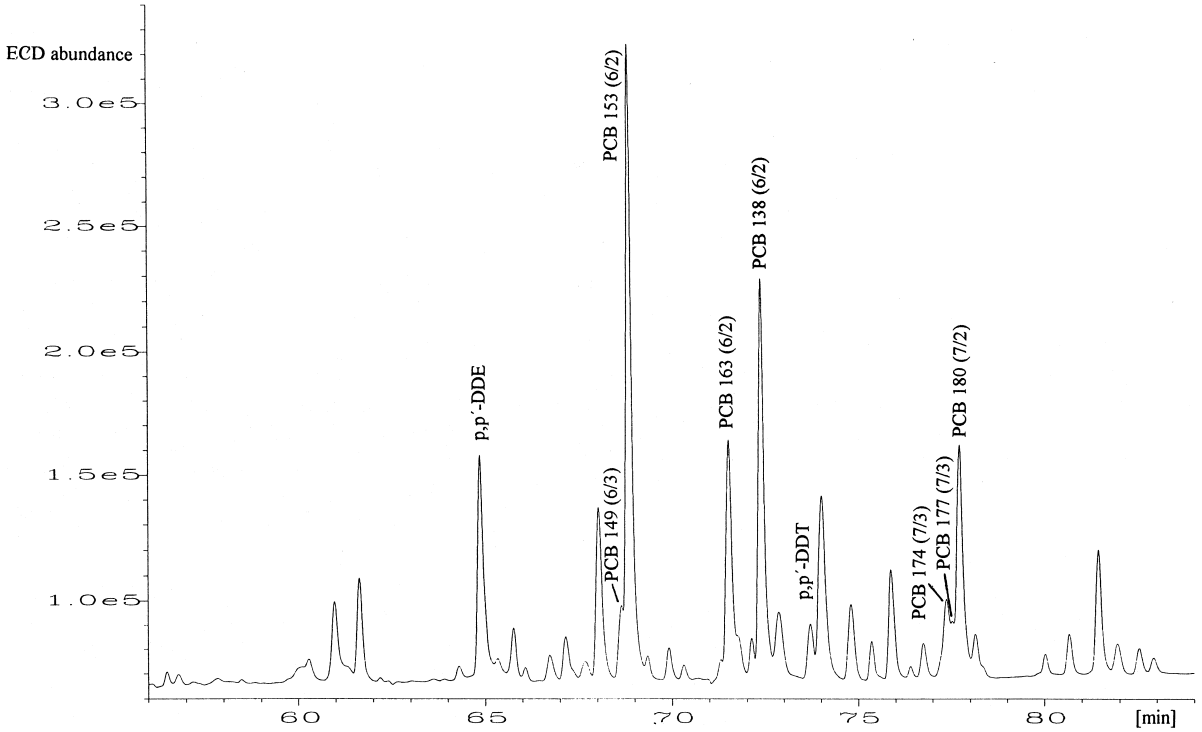

Fig. 3. GC–ECD chromatogram of a blubber extract of a harbour seal (*Phoca vitulina*) on Thermocap A (for conditions see Section 2).

Finally, it can be concluded that the Thermocap A column is an interesting supplement to the stationary phases which are recommended for the congener specific analysis of PCBs.

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