Calculation of the Real PCB Content in Environmental Samples

II.* Gas Chromatographic Determination of the PCB Concentration in Human Milk and Butter

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Berechnung der wahren PCB-Gehalte in Umweltproben
II. Ermittlung des PCB-Gehaltes von Frauenmilch und Butter

Zusammenfassung. Sämtliche in Frauenmilch und in Butter vorkommenden Komponenten des Gemisches der polychlorierten Biphenyle (PCB) wurden quantitativ bestimmt. Die Eichung erfolgte dabei mit Hilfe eines technischen PCB-Gemisches von bekannter Zusammensetzung. Der „wahre" Gesamtgehalt an PCB, berechnet durch Addition der Einzelwerte war bei Frauenmilch um 50%, bei Butter 40% niedriger als die Konzentrationen, die sich nach der sonst üblichen Berechnungsweise durch Bezug auf einige hohe Peaks technischer PCB-Gemische ergeben.

Außerdem wurden die annähernden Abbauraten der einzelnen PCB-Komponenten im Vergleich zu einem technischen PCB-Gemisch (Clophen A 60) berechnet.

Summary. All individual polychlorinated biphenyls (PCB's) present in human milk and in butter were quantified. For calibration, a technical PCB mixture of known composition was used. The "real" total PCB concentration was determined by the addition of concentrations of the individual components. In human milk the calculated content was 50% and in butter 40% lower than values obtained by the usual calculation based on evaluation of some higher peaks of technical PCB mixtures.

In addition, the approximate metabolization rates of the individual PCB components were calculated by comparison with the pattern of a technical PCB mixture (Clophen A 60).

Introduction

Polychlorinated biphenyls (PCB's) had first been detected in 1966 by Jensen et al. [2, 3] in the lipids of sea eagles and other wildlife, and later also by some other authors. In 1969/70 they were discovered, identified and quantified by Acker and Schulte [4, 5] in human fat tissue and mother's milk. After that, investigations were initiated worldwide to trace the compounds in the biosphere and PCB's were recognized as ubiquitous environmental contaminants. The quantification of PCB's, however, involved considerable difficulties. The reason was the quantity of PCB compounds with different numbers and different positions of chlorine atoms in the molecule, and the low resolution of the PCB components on packed GLC columns. Some years later, introduction of glass capillary columns [6] made a complete separation (with exception of a few critical pairs) and identification of most components possible (see ref. [5–8] in [1], [7]). Most of the pure individual PCB's, however, were not available for calibration. Therefore, simple calculation based on individual peak areas was not possible, because PCB isomers show different ECD responses.

A multitude of methods for the approximate quantification of PCB concentrations has been developed:

perchlorination with $SbCl_5$ to decachlorobiphenyl and its gas chromatographic determination [8, 9],

reduction with $LiAlH_4$ to biphenyl and its determination by HPLC [10, 11],

use of p,p'-DDE as a reference substance [12, 13], quantification of only a few individual PCB's with pure reference PCB's (see e. g. [14]),

use of technical PCB-mixtures as reference, and calculation by measuring some characteristic peaks (see e.g. [4, 5, 15–17]).

The last procedure, the calculation by technical PCB-mixtures similar in pattern to the samples, had been used most frequently up to now. For warm-blooded organisms, Aroclor 1254 or Clophen A 60 were used by most laboratories; and the relation of two or three high characteristic peaks to the corresponding peaks in the samples was measured.

It became clear very early that in this way only an approximate determination was possible. This is especially true for biological material of warm-blooded organisms, in which a series of PCB's is eliminated or drastically reduced because of biodegradation.

The results of different laboratories are only comparable if the same procedure for determination and calculation has been used. The main reason for the delay in issuing official PCB-limits for foods etc. apparently consisted in analytical difficulties.

After developing a method for the determination of all individual PCB's in technical mixtures, it was applied to Clophen A 60 and Clophen A 30 [1]. With the aid of these values it is now possible to calculate the real PCB contents in foods, for example. The method was applied to mother's milk and butter in this publication.

Dedicated to Prof. Dr. L. Acker on the occasion of his 70th birthday
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Offprint requests to: E. Schulte

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Experimental

Reagents

Petroleum ether, 40–60°C, Roth No. 9320
Dichlormethane, Roth No. 8424
n-Hexane, Roth No. 3907
} distilled over 150 cm Raschig column

Florisil 60–100 mesh, Roth No. 0101, heated at 550°C
Silica gel 60, Merck No. 7754, dried at 130°C, deactivated with 1.5% water

Acetonitrile, Merck No. 17
Sulfuric acid 95–97%, Ferak No. 01456
Demineralized water
} purified by extraction with petroleum ether

Equipment

Glass columns
a) for cleanup [18], 20 mm i. d., 50 cm long
b) for the separation of PCB's from pesticides [19], 7 mm i. d., 23 mm long
Separation funnel 250 ml
Round-bottom and pear-shape flasks
GC with Ni-63-ECD: Varian 3700
GC-MS combination: Hewlett Packard MS 5980 A coupled with a Hewlett Packard GC 5710

Preparation of Human Milk Extracts. 10 g of human milk are directly used for extraction and cleanup according to Stijve [18]. The solvent is evaporated, the extract dissolved in 1–2 ml n-hexane and fractionated according to Specht method [19], but with 2 g silica gel. PCB's are eluted with 15 ml n-hexane.

Preparation of Butter Fat Extracts. 20 g of butter are melted in a water bath at 50°C and centrifuged. 10 g of the upper phase are dissolved in 75 ml petroleum ether (saturated with acetonitrile) and extracted with three 40 ml portions of acetonitrile (saturated with petroleum ether) in a separation funnel according to [20]. The combined acetonitrile extracts are evaporated to dryness in a round-bottom flask, dissolved in 60 ml petroleum ether and shaken with 20 ml sulfuric acid in a separation funnel. After 2–3 h the lower phase is discarded. After shaking with another portion of 10 ml sulfuric acid and standing overnight, the lower phase is discarded again. Shaking with 10 ml sulfuric acid is repeated and the acid phase discarded immediately thereafter. The petroleum ether phase is washed neutral with several portions of water and evaporated to dryness. This extract is fractionated as described above for human milk.

Gas Chromatographic Separation Conditions

Glass capillary column: 50 m × 0.28 mm i.d.; coated with SE-30; film thickness about 0.1 µm.
Carrier gas: 1.5 bar nitrogen; split 1:10.
Temperature program: 140 → 260°C with 3°C/min.

PCB blank values were below 5% and the recovery was 95% for human milk. The recovery of PCB in butter fat was only about 50%, but the extracts were very pure. The discrimination during the partitioning step, caused by a little difference in recoveries of the individual PCB's, was compensated by a standard run.

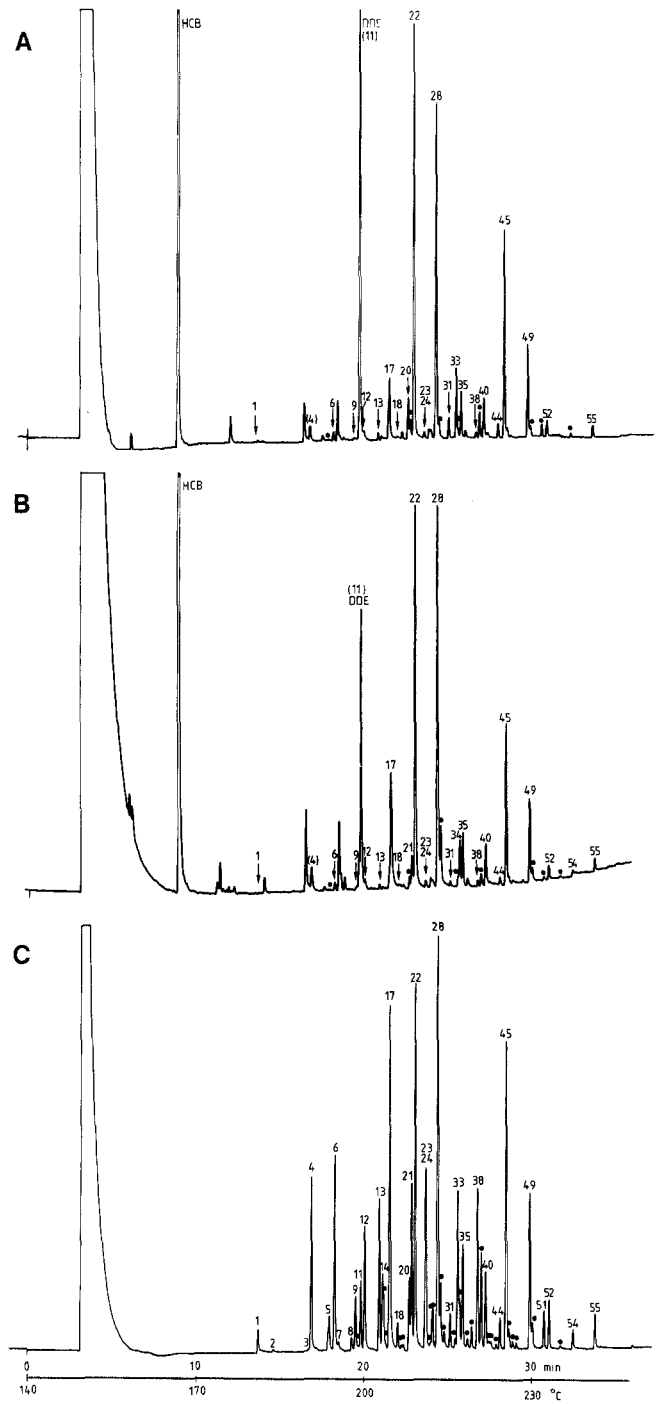

Fig. 1. Gas chromatogram of an extract from human milk (**A**), butter fat (**B**) and Clophen A 60 (**C**). Column: 50 m glass capillary, 0.28 mm i.d., coated with SE-30. For more details about sample preparation and separation conditions see text

The samples of human milk were collected from the region of Freiburg and the butter samples from different areas of the Federal Republic of Germany in 1983.

Results

GLC chromatograms of purified human milk and cow butter extracts, and of Clophen A 60 are shown in Fig. 1. The quantitative composition of Clophen A 60 had been

Table 1. Concentrations of the individual PCB components in human milk and in butter

Peak No.	Number of Cl atoms	Structure	Systematic numbering [7]	Percentage in Clophen A 60 [1]	*Human milk*					*Butter*				
					Concentrations in µg/kg fat (ppb), $n = 8$			Percentage in the pattern of the samples	Residual concentration in %[a]	Concentrations in ng/kg fat (ppt), $n = 6$			Percentage in the pattern of the samples	Residual concentration in %[a]
					$\bar{x}$	min	max			$\bar{x}$	min	max		
1	2	3	4	5	6	7	8	9	10	11	12	13	14	15
1	4	2,2′,5,5′	52	0.69	1.8	0.6	2.8	0.16	14	120	58	210	0.47	54
2	4	2,2′,3,5′	44	0.07	0.5	0.3	0.7	0.04	37	12	8	20	0.05	53
3	4	2,3′,4′,5	70	0.09	0.6	0.4	0.9	0.06	35	18	11	37	0.07	62
4	5	2,2′,3,5′,6	95	3.77	4.3	1.5	14	0.39	5.9	120	23	220	0.48	9.8
5	5	2,2′,3,4′,5[b]	90	1.11	2.5	0.9	7.4	0.22	12	110	21	220	0.45	31
6	5	2,2′,4,5,5′	101	5.20	3.9	2.5	10	0.35	3.9	200	140	270	0.79	12
7	5	2,2′,4,4′,5[b]	99	0.18	1.0[f]	interfering	peak	0.09	29[f]	interfering	peak[j]			
8	5	2,2′,3′,4,5	97	0.30	1.1	0.7	1.1	0.10	19	18	13	29	0.07	19
9	5	2,2′,3,4,5′ 2,3′,4,4′,6[b]	87 119	1.08	1.4	0.8	2.0	0.13	6.7	64	18	110	0.26	18
10	5	2,2′,3,4,4′[b]	85	0.05	0.9	0.2	2.0	0.08	93	interfering	peak[j]			
11	6	2,2′,3,3′,6,6′	136	1.59	2.0[f]	interfering	peak	0.18	6.5[f]	interfering	peak[j]			
12	5			3.0	2.0[f]	interfering	peak	0.18	3.4[f]	160	95	240	0.64	17
13	6	2,2′,3,5,5′,6	151	2.87	2.8	0.6	6.2	0.26	5.0	150	80	220	0.59	16
14	6	2,2′,3,3′,5,6′[b]	135	1.78	2.3	0.8	6.6	0.21	6.7	100	71	200	0.41	17
15	6	2,2′,3,4′,5,6[b]	147	0.70	0.9	0.4	2.2	0.09	6.6	51	43	91	0.21	23
16	6			0.05	0.2	0.1	0.4	0.02	21	5	4	6	0.02	31
17a, b	6	2,2′,3,4′,5′,6	149	9.39	75[g]	47	98	6,8	36	2880[g]	1940	4200	12	83
	6	2,2′,3,4,5′,6[b]	144											
17d	5	2,3′,4,4′,5[b]	118	1.31										
18	6	2,2′,3,3′,5,6[b]	134	0.59	3.3	1.4	7.6	0.30	29	64	29	120	0.26	34
19	6	2,2′,3,3′,4,6[b]	131	0.13	2.4	1.2	4.2	0.22	96	93	52	110	0.38	
20	6	2,2′,3,4′,5,5′[b]	146	1.64	26	17	37	2.4	82	720	260	1260	2.9	136
21	6	2,2′,3,3′,4,6′[b]	132	3.80	7.5	4.4	16	0.68	10	300	250	600	1.2	24
22	6	2,2′,4,4′,5,5′	153	9.88	308	176	579	28	161	5490	2880	9180	22	172
23	6	2,2′,3,4,5,5′	141	3.05	1.8[f]	1.9*	1.1	6.5	0.17	2.4	160	75	260	0.65	12
24	7	2,2′,3,3′,5,6,6′[b]	179	1.09	2.0[f]									
25	6	2,2′,3,4,4′,5[b]	137	0.07	2.8		1.6	4.0	0.26		61	36	85	0.25	
26	6	2,2′,3,3′,4,5′[b]	130	0.48	4.2[f]	8.4*	2.8	28	0.77	42	250	160	350	1.0	75
27	7	2,3,3′,5,5′,6[b,d]	165	0.55	16[f]										
28	6	2,2′,3,4,4′,5′	138	11.5	241	151	316	22	108	5640	2950	8170	23	152
29	6			1.29	6.0	2.4	9.2	0.55	24	550	290	790	2.2	132
30	6	2,2′,3,3′,4,5[b]	129	0.35	0.6	0.2	1.2	0.05	8.9	25	12	45	0.10	22
31	7	2,2′,3,3′,5,5′,6[b]	178	0.66	7.6	5.7	14	0.69	60	85	26	170	0.34	40
32	7	2,3,3′,4,5,6[b,d]	160	0.17	1.0	0.8	1.1	0.09	30	16	8	28	0.07	29
33	7	2,3,3′,4′,5,6[b,d]	163	3.26	34[h]	33.4*	30	58	3.1	41	1140[k]	860	1940	4.6	83
34	6	2,2′,3,3′,4,4′	128	0.96	2.0[h]										
35	7	2,2′,3,4′,5,5′,6	187	1.92	20	10	36	1.9	54	650	270	1170	2.6	105
36	6	2,3,3′,4,5,5′[b]	159	0.44	8.3	4.4	13	0.76	98	160	94	250	0.66	112
37	7	2,2′,3,4,5,5′,6	185	0.36	0.7	0.3	2.1	0.06	10	31	23	59	0.12	27
38	7	2,2′,3,3′,4,5,6′[c] 2,2′,3,3′,4,5,6[b]	174 173	3.64	3.7	1.1	12	0.33	5.3	130	86	230	0.53	11
39	7	2,2′,3,3′,4,5,6′[b]	174	1.74	10	5.9	19	0.93	30	130	73	210	0.53	23
40a	7	2,2′,3,3′,4′,5,6[b]	177	1.08	34[i]	25	52	3.1	88	700[i]	400	1090	2.8	109
40b	6	2,3,3′,4,4′,5[b]	156	0.91										
41	8	2,2′,3,3′,5,5′,6,6′	202	0.23	6.6	3.8	9.2	0.6	92	46	19	74	0.19	38
42	7			0.14										
43	8	2,2′,3,3′,4,5′,6,6′[b]	201[e]	0.11	0.7	0.4	1.6	0.07	33	110	22	150	0.44	
44	7	2,2′3,3′,4,5,5′[b]	172	0.66	6.3	3.6	12	0.57	49	86	35	160	0.35	40

45	7	2,2′,3,4,4′,5,5′	180	6.98	135	99	169	12.3	100	2260	1130	3690	9.1	100
46	7			0.45	6.5	4.1	8.2	0.59	75	97	45	150	0.39	67
47	7			0.21	2.8	2.0	3.8	0.25	69	48	26	83	0.20	71
48	8			0.13	0.4	0.2	0.8	0.04	16	16	7	22	0.06	38
49	7	2,2′,3,3′,4,4′,5[b]	170	3.70	63	45	78	5.8	88	930	510	1510	3.80	78
50	7			0.83	10	5.4	13	0.91	62	220	120	300	0.88	82
51	8	2,2′,3,3′,4,5,5′,6[b]	198	0.69	6.1	5.0	9.2	0.56	46	19	10	32	0.08	8.5
52	8	2,2′,3,3′,4,5,5′,6′	199[e]	1.06	11	7.8	12	0.97	54	210	86	370	0.85	61
53	7	2,2′,3,3′,4,4′,5,6′[b,d]	196	0.17	2.9	1.6	5.3	0.26	88	23	16	40	0.09	42
54	8	2,2′,3,3′,4,4′,5,6[b]	195	0.44	3.3	0.6	4.8	0.30	39	86	44	160	0.35	60
55	8	2,2′,3,3′,4,4′,5,5′	194	0.93	10	6.6	15	0.94	56	170	72	290	0.68	57
				99.52	1097 (= 1.1 µg/kg)			99.89		24719 (= 24.7 µg/kg)			100.16	

[a] Remainder of PCB's in the samples related to Clophen A 60 in % (see text for further details)
[b] Indicated only by [7]
[c] Indicated only by (ref. [5–6] in [1])
[d] By [7] indicated structure not corresponding to Cl-numbers found by us
[e] Systematic numbering corrected by us; see [1], (200 → 201 and 200 → 199)
Results of capillary-GC/MS-measurement (SID) with a selected sample:
[f] Determined under interference peak
[g] Pentachlorobiphenyl nearly completely remained, hexachlorobiphenyls completely absent
[h] Hexachlorobiphenyl (peak No. 34) nearly completely absent, heptachlorobiphenyl (peak No. 33) nearly completely remained
[i] Hexa- and heptachlorophenyls in nearly equal concentrations
[j] Chlorobiphenyl under it absent
[k] Hexachlorobiphenyl remained, heptachlorobiphenyl absent
[*] Mean value by ECD. Figures before the bracket are from selected samples by GC/MS
The values lower than about 2 µg/kg for human milk and 200 ng/kg for butter show a higher degree of inaccuracy

previously determined [1]. This mixture was now used as reference. Lower chlorinated mixtures as reference, e. g. Clophen A 30, are not necessary for samples from warm-blooded organisms. These PCB's, mainly used in closed systems, are not so widespread in the environment and have a better biodegradability.

The quantitative results of 8 human milk and 6 butter samples are summarized in Table 1.

In the first column the same continuous numbering of GC peaks appears as in [1]. Column 2 shows the number of chlorine atoms per molecule of the compounds, ascertained by us with a GC-MS combination. In the third column the structures of PCB's identified in our own laboratory with pure PCB standards (see ref. 5–8 in [1]), and identified by Ballschmiter and Zell [7] with retention indices and partly with pure PCB standards are listed. There are some discrepancies (cf. footnote d and peak 9 and 38 in Table 1). In column 4 the systematic numbers [7] corresponding to column 3 are listed. Column 5 shows the concentrations of the individual PCB's in Clophen A 60 [1] for comparison.

In columns 6–8 the concentrations of the individual PCB's in human milk in µg/kg (ppb) on fat base (6 = mean, 7 = lowest and 8 = highest value) are presented. In column 9 the percentages of the individual PCB's in the total PCB pattern of human milk are calculated. These values can be used for the calculation of the real total PCB content from the concentrations of single PCB's, e. g. measured by using pure standard substances, with a lower reliability, however. For this, the content of the individual PCB in the sample of human milk has to be divided by the corresponding percentage in column 9 and to be multiplied by 100. This procedure gives reasonable results if the PCB pattern is quite similar to our samples.

Column 10 shows the "residual percentages". These values represent the non-metabolized portion of individual PCB's by comparing the patterns of human milk and Clophen A 60. The concentrations of peak No. 45 in the samples (col. 6) and in Clophen A 60 (col. 5) are equated with 1 arbitrarily, and the concentrations of the other components are converted correspondingly. After this, the relations of the resulting figures of samples to those of Clophen A 60 are calculated in %:

$$\frac{\text{figure of sample}}{\text{figure of Clophen A 60}} \cdot 100.$$

Clophen A 60 is the most obvious mixture for this comparison because the patterns are very similar in the back part of the chromatograms. Some higher peaks of low biodegradability have values of about 100% (e. g. peak No. 28 and 49). The 2,2′, 4,4′, 5,5′-hexachlorobiphenyl (systematic number 153, peak number 22) was found in higher amounts in the samples than in Clophen A 60 in relation to PCB No. 180, so that values of about 150% result. This can be explained by the assumption that lower chlorinated PCB mixtures containing this compound may have contributed to the contamination too, but most of their other components are biodegraded. The values, being clearly below 50%, demonstrate a marked metabolization (e. g. peak number 4, 6, 12, 13, 21, 23, 38). But a number of values within the range of about 50–70% belongs to components of about 1% concentration and below in Clophen A 60, which are hardly biodegradable because of their structure (e. g. peak number 35, 44, 51, 52, 54, 55). This fact cannot

Table 2. Comparison of values calculated by different methods

		Human milk (in mg/kg)									Butter fat (in µg/kg)						
Sample No.		1	2	3	4	5	6	7	8	$\bar{x}$	1	2	3	4	5	6	$\bar{x}$
Sum of the individual concentrations[a]		1.45	1.08	0.90	1.13	0.71	1.23	0.99	1.34	1.10	16.6	36.9	31.1	18.9	27.2	16.6	24.7
Calculated by system No.	peak No.																
153	22[b]	3.53	2.63	2.57	2.97	1.54	3.46	2.60	3.05	2.79	37.4	93.1	60.0	41.6	57.4	41.0	55.1
138	28[b]	2.78	2.19	1.81	2.14	1.13	2.31	1.75	2.58	2.09	26.5	71.6	60.4	37.9	57.8	28.1	47.1
180	45[b]	1.81	1.71	1.62	2.33	1.21	2.34	1.83	2.42	1.91	24.3	52.7	48.7	20.5	40.6	17.8	34.1
$\bar{x}$[c]		2.71	2.18	2.00	2.48	1.29	2.70	2.06	2.68	2.26	29.4	72.5	56.4	33.3	52.0	29.0	45.4
Conversion factor[d]		0.54	0.49	0.45	0.46	0.55	0.47	0.48	0.50	0.49	0.57	0.51	0.55	0.57	0.52	0.57	0.54
Percentage of peaks No. 22, 28, 45[e]		70	58	64	63	60	62	59	55	61	50	57	52	52	55	51	53

[a] Sum of the individual concentrations determined according to the method described in this publication
[b] PCB content, calculated by single peaks of Clophen A 60
[c] Mean value of [b], which has been given as "PCB concentration" formerly ("three peak method")
[d] Factor for converting the values [c] to [a]
[e] Sum of the percentages of peaks No. 22, 28 and 45 in the pattern of the samples

be explained plausibly at the moment. On the whole, our hypothesis for the biodegradability of PCB's 10 years ago [21] has been confirmed. The rules were made at that time on the basis of only a few identified PCB's. Some peaks, thought to consist of one PCB, were in the meantime identified as mixed peaks, e. g. number 17, which consists of penta- and hexachlorobiphenyls [22].

In the following columns 11–15 the corresponding values for butter fat are listed. The concentrations in the columns 11–13 are given in ng/kg (ppt).

In the first communication [1] we described the separation of "critical pairs" of hardly resolvable PCB's in Clophen A 60. This was possible with two columns of different polarity coupled by a column switching system according to Deans and the Live-GC. These groups could not be resolved in this way, because a system with two ECD's was not at our disposal. The error caused by overlapping is low. Some components in mixed peaks of PCB's with a different number of chlorine atoms were quantified with the aid of a GC-MS combination (see foot-notes f–k in Table 1).

Discussion

The procedure described here implies the exact quantitative determination of (nearly) all individual PCB's which renders a differential toxicological evaluation of PCB mixtures possible. Furthermore, the sum of the single PCB's gives the real total PCB concentration.

On the other hand, the question arises, to which extent these results are comparable with those of other analyses obtained by other methods. The calculation method, by relating to 2–3 high peaks (systematic numbers 153, 138 and 180) of a technical mixture, which had been used in the past most frequently for the material analysed here, gives too high values, as mentioned above (see Table 2[e]). This method is customary in many laboratories even today, because it is simple and includes a pattern recognition. It can, however, be applied with an accuracy sufficient in most cases by converting these values to the real total PCB content by a "correction factor" (see Table 2[d]). This factor is obtained by dividing the real content (sum of individual PCB) and the value calculated by relating to 3 peaks of a technical mixture (Clophen A 60).

In recent years the calculation of only a few characteristic PCB's with pure reference PCB's has been described (cf. [14]) and is proposed for legislative purposes. The sum of the three most characteristic peaks (systematic number 153, 138 and 180) amounts to more than 50% of the pattern in the samples, for human milk between 55 and 70%, 61% in mean (see Table 2[e]). The real PCB content can be calculated with some reliability by multiplying the sum of the measured concentrations of these three PCB's by a factor of 1.64 (= 100/61).

In the second part of Table 2 the analogous values of butter fat are listed.

Of course, these factors are not absolutely exact for all samples of the type analysed here because of "biological fluctuations"; but they are applicable at least for most samples from Western Europe with a sufficient certainty in residue analysis. On the other hand, it is no problem for specialized laboratories to evaluate all peaks in the way described here, e. g. with a suitable integrator.

Indeed it has been published [7] that Aroclor 1260 and Clophen A 60 have the same chlorine content (60%) and the same average number of chlorine atoms per molecule (6.3), but the definition of the former manufacturers is that Aroclor 1260 has 60% chlorine resulting in 6.26 chlorine atoms per molecule in average, and that Clophen A 60 has an average number of 6 chlorine atoms per molecule result-

ing in 58.9% chlorine theoretically, whereas the manufactured product contained 58% [23]. It can be clearly seen by comparing the chromatograms of the two mixtures that Aroclor 1260 has another quantitative composition (higher peaks in the back part of the chromatograms) than Clophen A 60. Nevertheless, e.g. Aroclor 1254 can be used as a reference in the way described here if its quantitative composition is analysed as in [1]. Under these conditions the resulting sample data are independent of the used mixtures, provided that all components to be calculated are present.

The quantitative composition of technical PCB mixtures and – in a similar way here – of sample extracts has also been analysed by some other laboratories. But in these cases either the separation efficiency of the packed columns used by most authors was not sufficient for good resolution, or the calibration mode was not clearly, or not at all given, or appeared to be unsuitable [24–28].

We mostly use a mixture of SE-30 and SE-52 (1 + 1) instead of pure SE-30 in routine as stationary phase in GC, because the HCH-isomers and the HCB are better resolved [29], but the separation of PCB's remains virtually identical.

The results we published some years ago [30, 31] for human depot fat and human milk were calculated by the "three peak method". By multiplying by a factor of 0.5, the results can be corrected to the "real PCB content" with sufficient reliability.

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