

Levels of polychlorinated dibenzo-p-dioxins, dibenzofurans, biphenyls and pesticides in harp seals from the Greenland Sea

M. Oehme^a, M. Schlabach^a, K. Hummert^b, B. Luckas^b and E.S. Nordøy^c

^aNorwegian Institute for Air Research, P.O. Box 64, N-2001 Lillestrøm, Norway

^bInstitute of Food and Environment, Dornburgerstr. 25, D-07743 Jena, Germany

^cDepartment of Arctic Biology, University of Tromsø, Breivika, N-9037 Tromsø, Norway

1. Introduction

PCDD and PCDF are widely spread in the environment and can even be detected in marine mammals from the Canadian and European part of the Arctic¹⁻³). So far, a very limited number of animals have been analysed. In addition, in 2 studies only a few PCDD/PCDF congeners could be detected due to levels close to the detection limits of the applied methods^{2,3}). Furthermore, with one exception³) only results for PCDD/PCDF were reported and not for other organochlorines such as PCB and DDT. However, to study possible correlations between levels of PCDD/PCDF and other persistent polychlorinated compounds, such data have to be available from the same samples.

The objectives of this study were to extend the very limited data base about levels of PCDD/PCDF in Arctic marine mammals and to quantify simultaneously other persistent organochlorines (polychlorinated non-ortho, mono-ortho and di-ortho substituted PCB as well as polychlorinated pesticides). Concentrations of PCDD/PCDF in Arctic seals are only available for ringed seal blubber and one harp seal sample. Therefore, harp seals (*Phoca groenlandica*) were selected for this study to obtain information about levels in a different species. The animals were caught east of Greenland. From this area levels of organochlorines in pinnipeds have not been published before.

Mainly male harp seals with a large age distribution were selected since they do not have the possibility to eliminate accumulated organochlorines by lactation.

2. Methods

Seal samples

The tissues examined in this study came from animals which were caught in the West Ice region (72-75°N and 5-10°W) of the Greenland Sea in 1991. Two female and ten male individuals were sampled randomly and about 50 g of blubber from above the sternum and brain were collected, and wrapped in aluminium foil and stored at -20°C. Ages were obtained from the cementum growth layers in canine teeth. Details about origin, age and sex are given in Table 1.

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Determination of PCDD/PCDF and coplanar PCB

For quantification of PCDD/PCDF and coplanar PCB the same method was applied as recently described in detail for the determination of sub-ppt levels in Antarctic seals⁴⁾. Therefore, only a brief description is given. 5 g of seal blubber was homogenised with 100 g of waterfree sodium sulphate. After dehydration of the sample the salt-like mixture was homogenised once more, and a mixture of ¹³C-isotope-labelled compounds was added (totally 4-16 pg for each congener). The sample was transferred to the top of the first column of the multicolumn clean-up system. Final clean-up was carried out using a combination of two Pasteur pipettes, one filled with sulphuric acid coated silica and one with aluminium oxide. PCDD/PCDF and coplanar PCB congeners were separated on a fused silica capillary of 30 m length, 0,25 mm id. coated with 0,1 µm of a polymethyl-polyphenyl siloxane (e.g. DB5, J&W Scientific). Quantification was carried out by high resolution mass spectrometry at a resolution of 10'000. Quality assurance measures were included according to international standards⁵⁾.

PCB and other polychlorinated compounds

10 g of blubber or brain was digested using a mixture of 10 ml perchloric (70%) and 10 ml acetic acid at 80 °C for 4 hours. After extraction with 80 ml hexane the organic phase was treated with 50 ml of concentrated sulphuric acid. After evaporation of the solvent close to dryness, 2 ml iso-octane was added and the final extract was shaken out once more with 1 ml of sulphuric acid. Quantification was carried out by ECD on two capillary columns: 50 m x 0.32 mm i.d. coated with 0,17 µm HP5 or 0.25 µm OV 1701 (Macherey & Nagel). The separation conditions were as follows: Injector temperature 220 °C, splitless injection of 1 µl; detector temperature 300 °C; temperature program, 180 °C for 4 min, then 6 °C/min to 210 °C, 4 min, 4 °C/min to 270 °C, isothermal 25 min. Recently, the applied method was successfully controlled by an interlaboratory study where 14 laboratories participated⁶⁾.

3. Results and Discussion

To our knowledge this is the first study where levels of PCDD/PCDF, PCB including coplanar PCB and chlorinated pesticides were determined simultaneously in individual seal samples from the Arctic. Very little is known about levels of polychlorinated compounds in Arctic harp seals. Only data for PCB and DDT levels are available from one study carried out by Ronald et al.⁷⁾. Furthermore, the only reported PCDD/PCDF levels were from one pooled sample of 3 juvenile female harp seals from a not specifically described area²⁾.

PCDD and PCDF

Detection limits for blubber in the order of 0,05-0,1 pg/g at a signal-to-noise ratio of 3:1 allowed to detect all 2,3,7,8-chlorine substituted PCDD/PCDF congeners except 1,2,3,7,8,9-HxCDD and 1,2,3,4,7,8,9,-HpCDF in the blubber samples (see Table 1). Most of the blubber data fulfilled the criterion that blank values should be equal to the detection limits or at least 10 times lower than the measured concentrations. Blank levels for OCDD calculated with the same sample amount were in the order of 0,5 pg/g and, therefore, contribute significantly to the measured concentrations in blubber and brain. Only 2,3,7,8-TCDF was found in measurable quantities in brain.

Ringed seals and harp seals feed on both crustaceans and fish from the upper water layers. Compared to the average levels found in Arctic ringed seal blubber^{1-3,8)} the

TABLE 1

PCDD and PCDF concentrations (pg/g) in seal blubber (*Phoca groenlandica*). Recovery rates of added ^{13}C -marked 2,3,7,8-chlorine substituted isomers were within 83-103 %. All values are corrected for recoveries.

Sample no. Sampling site	1 75°10'N 10°06'W	2 72°43'N 09°16'W	3 72°49'N 09°01'W	4 72°52'N 09°10'W	5 74°00'N 13°30'W	6 74°00'N 13°30'W	7 75°00'N 09°29'W	8 75°33'N 04°59'W	9 75°33'N 04°59'W	10 75°33'N 04°59'W
Sex	female	male	male	male	male	male	male	male	male	male
Age	1	6	3	16	18	6	≈10	13	10	4
2,3,7,8-TCDD	0,75	0,78	0,71	0,73	0,69	0,55	1,26	1,20	0,52	0,90
1,2,3,7,8-PeCDD	1,76	3,25	2,14	2,00	2,93	1,75	5,20	4,01	1,67	2,38
1,2,3,4,7,8-HxCDD	0,31	0,78	0,52	0,53	0,55	0,57	1,24	0,76	0,40	0,42
1,2,3,6,7,8-HxCDD	1,42	3,73	1,84	2,13	3,11	2,30	9,34	4,29	2,06	2,20
1,2,3,7,8,9-HxCDD	0,35	0,45	0,46	0,38	0,51	0,43	0,81	0,47	0,30	0,34
1,2,3,4,6,7,8-HpCDD	0,44	0,64	0,66	0,96	0,93	0,79	1,02	0,94	0,66	0,91
OCDD	2,25	2,32	2,50	6,59	5,83	1,27	3,38	3,10	3,49	3,57
2,3,7,8-TCDF	5,56	4,48	10,4	3,66	4,82	5,38	4,03	8,72	3,65	8,04
1,2,3,7,8-PeCDF ^a	0,66	0,65	0,85	0,55	0,70	0,59	0,79	1,07	0,41	0,73
2,3,4,7,8-PeCDF	2,86	4,65	5,11	2,91	3,37	2,68	6,73	6,02	2,11	4,46
1,2,3,4,7,8-HxCDF ^a	0,74	1,03	1,38	1,47	0,83	1,38	2,87	0,87	0,56	1,98
1,2,3,6,7,8-HxCDF	0,52	1,07	0,85	0,83	0,68	0,73	1,79	0,77	0,50	0,92
1,2,3,7,8,9-HxCDF	<0,16	0,10	0,07	<0,15	0,09	0,10	<0,12	0,03	<0,11	<0,25
2,3,4,6,7,8-HxCDF	0,34	0,73	1,11	1,02	0,46	1,04	2,53	0,57	0,35	1,11
1,2,3,4,6,7,8-HpCDF	0,41	0,35	0,53	0,59	0,47	0,79	0,92	0,41	0,29	0,68
1,2,3,4,7,8,9-HpCDF	<0,17	<0,16	0,07	<0,13	0,13	0,13	<0,06	<0,04	<0,03	<0,22
OCDF	0,56	0,48	0,50	0,72	0,75	1,27	0,21	0,37	0,39	0,91
2,3,7,8-TE (PCDD/F) ^b	4,0	5,99	6,02	4,23	4,97	3,98	9,53	7,89	3,22	5,88
3,3',4,4'TeCB	158	91	211	141	111	184	152	410	129	199
3,3',4,4',5-PeCB	131	205	167	149	213	130	472	326	156	198
3,3',4,4',5,5'-HxCB	24,4	128	99	256	30,4	213	942	57	38,3	115
2,3,7,8-TE (PCB) ^c	15,9	27,8	23,8	29,1	23,9	25,5	95,9	39,6	18,8	27,5

^a Not separated from 1,2,3,4,8-PeCDF or 1,2,3,4,7,9-HxCDF, respectively; ^b Nordic model; ^c see¹¹⁾; <, individual detection limit at a signal-to-noise ratio 3:1.

TABLE 2

Concentrations of polychlorinated pesticides, hexachlorbenzene and polychlorinated biphenyls in seal blubber (*Phoca groenlandica*). All results are given in ng/g wet weight. The lipid concentration for all samples varied between 83-86%.

Sample no. Sampling site	1 75°10'N 10°06'W	3 72°49'N 09°01'W	4 72°52'N 09°10'W	5 74°00'N 13°30'W	6 74°00'N 13°30'W	7 75°00'N 09°29'W	8 75°33'N 04°59'W	9 75°33'N 04°59'W	11 75°33'N 04°59'W	Average x	Std. dev.
Sex	female	male	male	male	male	male	male	male	male		
Age	1	3	16	18	6	≈10	13	10	10		
HCB	210	87,4	277	61,0	176	179	47,6	33,0	83,3	128,3	84,7
α-HCH	21,1	12,5	33,3	42,3	28,6	24,7	26,4	19,7	22	25,6	8,6
α(+)/α(-) ratio	1,68	1,23	1,70	1,08	1,43	1,29	1,28	2,45	1,27	1,5	0,4
β-HCH	3,9	<2,4	10,6	7,5	6,3	4,4	8,7	5,3	2,8	6,2	2,6
γ-HCH	2,1	<1,9	3,4	6,3	5,5	2,8	7,8	4,2	2,0	4,3	2,1
PCB 28	2,4	3,4	<2	<2	4,9	<2	<2	5,8	3,6	3,1	1,4
PCB 31	1,1	1,3	n.a.	n.a.	3,6	n.a.	n.a.	3,4	2,8	2,4	1,2
PCB 52	13,2	18,8	22,7	40,9	22,9	35,7	16,7	26,3	20,0	24,1	9,0
PCB 101	25,8	29,1	60,5	83,3	29,6	90,0	21,1	41,7	39,1	46,7	25,5
PCB 105	5,8	7,6	n.a.	n.a.	8,2	n.a.	n.a.	11,1	9,8	8,5	2,0
PCB 118	16,2	18,4	n.a.	n.a.	32,0	n.a.	n.a.	35,6	29,2	26,3	8,5
PCB 153	48,2	58,1	484	1041	80,0	550	200	111	76,9	294,4	337,4
PCB 156	1,7	2,2	n.a.	n.a.	3,5	n.a.	n.a.	4,8	3,1	3,1	1,2
PCB 138+163 ^a	42,5	50,0	325	769	75,0	346	183	97,1	76,6	218,2	236,3
PCB 180	13,3	20,0	168	228	23,8	157	63,64	30,7	22,2	80,7	81,3
ΣPCB ^b	156,9	188,9	-	-	259,7	-	-	336,8	261,1	-	-
ΣPCB ^c	453	541	4045	9050	775	4480	1915	1041	768	2560	2865
p,p'-DDE	116	176	1047	2757	273	1041	736	479	230	761,7	829,4
p,p'-DDT	26,6	48,4	78,1	263	40,0	52,6	90,9	41,7	31,6	74,8	73,6
p,p'-DDD	8,6	27,4	14,9	54,5	21,9	21,7	17,4	11,1	8,7	20,7	14,2
ΣDDT	151	252	1140	3075	335	1115	844	539	270	857,9	911,7
ΣPCB/ΣDDT	3,0	2,2	3,6	2,9	2,3	4,0	2,3	1,9	2,8	2,8	0,7

^a Also co-elution with PCB 164; ^b calculated on basis of the 9 measured PCB, ^c calculated from PCB 138 and 153 according to Ballschmiter and Zell¹²⁾

PCDD/PCDF concentrations in harp seal blubber expressed as 2,3,7,8-TEQ (Nordic modele) are about a factor of 2-3 lower. In the only harp seal blubber sample (3 juvenile females, <1 year) analysed before²⁾, most PCDD/PCDF congener levels were below the detection limit of 1-2 pg/g. The concentrations for the four detected congeners 2,3,7,8-TCDF, 1,2,3,7,8-PeCDD/PeCDF and 2,3,4,7,8-PeCDF were comparable with the levels found in this study.

Beside OCDD, 2,3,7,8-TCDF is the most dominant congener in both Arctic species. Despite significant differences in the age and sex distribution between the analysed groups of Arctic ringed (about same number of males and females, 3-10 years old⁴⁾) and harp seals (mainly male seals, 3-18 years old), the congener profiles were quite similar.

Non-ortho substituted PCB

As can be seen from Table 1, in harp seal blubber the 2,3,7,8-TEQ levels for the 3 coplanar PCB CB 77, 126 and 169 are a factor of about 3-10 times higher than for PCDD/PCDF. The ratio between CB 77 and CB 126 in harp seal blubber varied between 0,3 to 1,4 which is similar to the results reported for Canadian ringed seal blubber (0,3 to 2,4)⁹⁾. In ringed seal blubber from the Canadian Arctic the level of CB 169 was always much lower than of CB 77 and CB 126⁹⁾. To our surprise the CB 169 concentrations were the highest of all 3 congeners in some harp seal blubber samples.

Other polychlorinated biphenyls

Σ PCB concentrations in harp seal blubber (average 2560 ± 2865 ng/g wet weight) estimated from PCB 138+163 and 153 (see Table 2) were comparable to those reported for male harp seals from the northern Baffin Bay (3760 ± 3650 ng/g wet weight)⁷⁾. Levels were in the same range in male ringed seals from the Canadian Arctic¹⁰⁾ and from Spitsbergen^{1,8)}.

Single PCB congeners were also determined in brain. PCB levels in brain from Arctic seals have not been reported before. The blubber/brain concentrations ratios (wet weight basis) differed considerably both between animals and different congeners. The ratio for PCB 52 in males covered a large range (2,5-14) and varied even more for PCB 153 (5-37). We feel that the PCB levels in brain are surprisingly high compared to blubber.

Chlorinated pesticides

Levels of p,p'-DDE in harp seal blubber from males from the Greenland Sea (Table 2) were lower than in ringed seals from Spitsbergen¹⁾ and in male harp seals from the Canadian Arctic⁷⁾. Concentrations of p,p'-DDT and p,p'-DDD were in average 10 respectively 40 times lower. No significant correlation ($r=0,3-0,4$, $p<0,1$) could be found between levels of p,p'-DDE, p,p'-DDT and p,p'-DDD in harp seal blubber.

α -HCH and total HCH concentrations in harp seal blubber were considerably lower than in ringed seal blubber from the Arctic while β - and γ -HCH levels were comparable^{1,10)}. Hexachlorobenzene levels were about one order of magnitude higher than in ringed seals.

Correlations between different compound groups as well as age

The different compound groups quantified in the seal blubber extracts are quite different in terms of sources, transport mechanisms, toxicological interactions, bioaccumulation and metabolism. The obtained data were analysed for possible correlations between single compounds and substance classes as well as between age and concentration

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levels. Though the number of individual was very limited, the following correlations at a significance level of $p < 0.05$ were found.

The levels of PCB congeners with 5 or more Cl atoms correlated very well with those of p,p'-DDE (e.g. for PCB 153, $r = 0.977$). However, no relationship was found between di-ortho substituted PCB and any of the PCDD/PCDF congeners or non-ortho PCB. Both non-ortho and other PCB are part of technical PCB mixtures, and one can expect that congeners with the same number of chlorines behave similar concerning long range transport to the Arctic. However, di-ortho and non-ortho substituted PCB behave completely different regarding distribution in the body, half life and enzyme inducing properties. Non-ortho substituted PCB have dioxin-like properties. A surprisingly good correlation between levels of 1,2,3,7,8-PeCDD and 3,3',4,4'-PeCB was found ($r=0.967$).

α -HCH levels correlated well with those of PCB 153 ($r=0.843$). Comparing α -HCH with γ -HCH a weaker relationship just above the significance level for $p < 0.1$ was found ($r=0.633$). This indicates that PCB 153 and α -HCH behave more similar in terms of bioaccumulation and persistency than α - and γ -HCH.

Despite the limited number of analysed samples, age was positively correlated with levels of Cl₅- and Cl₆-PCB such as PCB 153 ($r=0.875$) as well as p,p'-DDE ($r = 0.804$).

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