Concentration and congener pattern of polychlorinated biphenyls in blubber and liver of Hubbs' beaked whale (*Mesoplodon carlhubbsi*), using a sulfoxide and an Ag-ION solid phase extraction cartridge as a simplified cleanup technique for biological samples

Katsunori Anezaki*[a], Ayaka Matsuda[b], Takashi Matsuishi[c]

[a] Hokkaido Research Organization, Environmental and Geological Research Department, Institute of Environmental Sciences, Hokkaido, Japan

[b] Graduate School of Fisheries Sciences, Hokkaido University, Hokkaido, Japan

[c] Faculty of Fisheries Sciences, Hokkaido University, Hokkaido, Japan

Type of paper: Shorter Research Note

***Corresponding Author:**

Katsunori Anezaki

Hokkaido Research Organization, Environmental and Geological Research Department, Institute of Environmental Sciences

N19W12, Kita, Sapporo, Hokkaido 060-0819, Japan

Tel: +81-11-747-3521

Fax: +81-11-747-3254

Email: anezaki@hro.or.jp

Abstract

We performed the first known study of polychlorinated biphenyls (PCBs) concentrations and patterns in the blubber and liver of a Hubbs' beaked whale. Samples were pretreated with Supelclean™ sulfoxide and Discovery® Ag-ION solid phase extraction cartridges to remove whale oil. PCB concentrations in the blubber and liver were 13,000 and 7,300 ng/g lipid, respectively. Highly poisonous congeners such as dioxin-like (DL) PCBs tended to accumulate in the liver. The toxic equivalents (TEQ) of DL-PCBs in the liver (740 pg-TEQ/g lipid) were higher than those in the blubber (74 pg-TEQ/g lipid). The blubber and liver samples showed that hexachlorinated biphenyls were dominant among homologues, and PCB-153 was dominant among congeners. Several congeners accumulated disproportionately in the blubber and the liver (PCB-28, 52, 74, 99, and 118), while others did not persist (PCB-31, 70, and 110). This indicates that PCBs are metabolized differently according to their specific composition.

Keywords: congener specific analysis, Hubbs' beaked whale (*Mesoplodon carlhubbsi*), polychlorinated biphenyls, pretreatment method

1. Introduction

About 1.3 million tons of physically and chemically stable polychlorinated biphenyls (PCBs) have been produced and used worldwide for a variety of purposes (Breivik et al., 2002). They are primarily used as insulating oil for transformers and capacitors, but have also been used for carbonless paper and plasticizers in open systems. PCBs have been improperly disposed of and released into the environment as waste. Even though production of PCBs ended in the 1970s and their use is being abolished in countries in accordance with the Stockholm convention (Stockholm Convention, 2001), they are still found in the environment. In living organisms, PCBs tend to accumulate in species at the top of the food chain, especially in the ocean, and marine mammals such as whales and dolphins have highly concentrated PCBs. Therefore, monitoring and environmental dynamics research has been conducted around the world (Bachman et al., 2014; Corsolini et al., 1995; Dorneles et al., 2015; Fair et al., 2010; García-Álvarez et al., 2014; Hoguet et al., 2013; Kajiwara et al., 2006; Moon et al., 2010; Mössner and Ballschmiter, 1997; Norstrom and Muir, 1994; Ross et al., 2000; Tanabe et al., 1983; Yordy et al., 2010).

Analysis of whale tissue for PCBs, however, is problematic because the oil must be removed. Alkalinolysis with potassium hydroxide, treatment with sulfate or a florisil column, and gel permeation chromatography are commonly used. Supelclean™ sulfoxide and Discovery® Ag-ION solid phase extraction cartridges (sulfoxide SPE and Ag-ION SPE) can remove oil (Numata et al., 2008). These columns are primarily used to analyze PCBs in mineral oil and have been used to analyze PCBs in sea sediments and chemical products (Anezaki, 2011; Anezaki and Nakano, 2015).

In this study, we tested this method as part of the analysis of blubber and liver samples from Hubbs' beaked whale (*Mesoplodon carlhubbsi*, family Ziphidae, parvorder Odontoceti) for congener-specific analysis of PCBs. This species is distributed in the North Pacific. With only 65 stranding records available (Bachara, pers. comm., 2016), its biology is poorly understood, and contaminant data are lacking (Mead et al., 1982; Taylor et al., 2008). The aim of this study was to measure the level of each PCB congener in this little-studied species for comparison with other whale species and a Kanechlor mixture as a reference standard.

2. Experimental

2.1 Samples and reagents

Samples were collected from a stranded Hubbs' beaked whale (Stranding Network Hokkaido identification SNH15011; body length 493 cm (Fig. S1), found on 14 April 2015 at Samani, Hokkaido, Japan (42.12N, 142.95E, Fig. S2); it died within 4 h of discovery. The corpse was dissected in situ on 15 April 2015 and was identified as a mature male following Mead et al. (1982). Blubber and liver samples were collected in plastic bags, frozen, transported to the laboratory, and stored at -30°C until analysis. The details of PCB standards and other reagents are provided elsewhere (Anezaki and Nakano, 2014; Anezaki and Nakano, 2015).

2.2 Sample preparation and chemical analysis

Prior to analysis, the liver sample was homogenized using a GRINDOMIX (GM200, Retsch, Germany), and the blubber sample was mashed manually. Approximately 1–3 g of the sample was thoroughly with sodium sulfate and extracted for 6 h with a Soxhlet extractor (B-811, hot extraction mode, BÜCHI, Switzerland) with acetone and hexane (1:1). The extracted liquid was separated, and a surrogate was added. An aliquot was dissolved in hexane and treated several times with concentrated sulfuric acid, placed into a multilayered silica gel column (sulfuric acid and potassium hydroxide silica gel), and eluted with hexane. The eluate was placed in a silver nitrate silica gel column to remove the sulfur and eluted with 5% dichloromethane/hexane. The eluate was added to a sulfoxide SPE cartridge, and the impurities were removed using 9 mL of hexane. An Ag-ION SPE cartridge was then attached to the bottom of the sulfoxide SPE cartridge, and the sample was eluted with 50 mL of 5% dichloromethane/hexane. This eluate was subsequently concentrated and syringe spikes were added to make a 50 μL nonane solution. Congener-specific analysis for PCBs was conducted for all samples. PCBs were quantified using a high-resolution gas chromatograph (HP6890, Agilent Technologies, USA) in combination with a high-resolution mass spectrometer (JMS700D, JEOL, Japan). The samples were analyzed with an HT8-PCB capillary column (60 m, 0.25 mm inner diameter, Kanto Kagaku, Japan). The target PCBs were monitored in selected ion monitoring mode for each homologue group.

2.3 Lipid content analysis

Approximately 5 g of the sample was placed in a Teflon™ vessel with 20 mL of chloroform and 40 mL of methanol and were homogenized with a POLYTRON® homogenizer (PT3100, KINEMATICA AG, Switzerland). Another 20 mL of chloroform was added and the contents were homogenized again. After filtration, the residue was returned to the vessel with 80 mL of chloroform and methanol (1:1), and the contents were homogenized and filtered. The filtrate was placed in a separating funnel with 60 mL of pure water and lightly shaken. After settling for a few hours, the chloroform phase was dehydrated with sodium sulfate and concentrated. A constant mass was obtained with phosphorus (V) oxide desiccant in a desiccator, and the lipid content was calculated from the weight difference.

2.4 Quality control and quality assurance

Detection limits depended on congener and sample size, and each congener varied from 40 to 90 pg/g wet weight (blubber) and 10 to 20 pg/g wet weight (liver). Peak concentrations were determined by isotope dilution quantification using surrogates. The surrogate recovery rates of PCBs were within the range of 50–120%. Laboratory blanks were also checked regularly, and values were found to be extremely low. Two standard reference materials (SRM 1974c Mussel Tissue from the National Institute of Standards and Technology (Maryland, USA) and CRM 7906-a Kanechlor Mixture (Kanechlor300:400:500:600 = 1:1:1:1; KC-mix) from the National Institute of Advanced Industrial Science and Technology (Tsukuba, Japan) were used to test the accuracy of the method; SRM 1945 whale blubber was not available. On average, obtained values from SRM 1974c and CRM 7906-a were different from the certified values by +20% and -3.7%, respectively. The blubber and liver samples were analyzed three times, and the total PCB values were compared to determine precision (relative standard deviation: 12% [blubber], 4.0% [liver]).

3. Results and discussion

3.1 Effects of sulfoxide and Ag-ION SPE cartridges

The lock-mass calibration method uses known standard substances such as perfluorokerosene (PFK) to correct mass measurements, but PCB congeners cannot be analyzed accurately when oil is present because it interferes with the lock mass monitor channel; therefore, oil must be removed during sample pretreatment. Fig. 1 shows the lock-mass chromatogram for blubber not pretreated with sulfoxide and Ag-ION SPE cartridges. The PCB chromatograms did not show interference or baseline fluctuations for each homologue (data not shown), but fluctuations and a lock mass decline were observed for m/z 242.986 and m/z 330.979. Pretreatment with the sulfate and multilayer silica gel column chromatography did not remove all interference components. A blubber sample pretreated with a sulfoxide and an Ag-ION SPE cartridge showed reduced lock mass fluctuation and decline (Fig. 2). Presumably, the sulfoxide and Ag-ION SPE cartridges effectively removed a blubber-specific oil that interferes with the identification and quantification of the target substances and also causes the measuring apparatus to deteriorate, thereby improving the analysis quality of target substances.

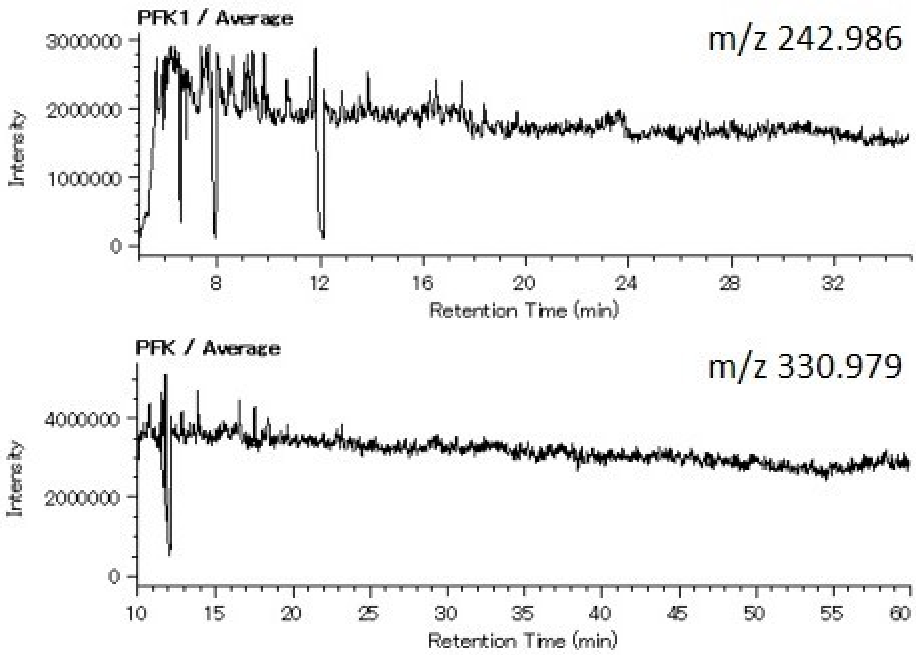

Fig. 1 Mass chromatograms of perfluorokerosene as lock mass (without treatment with Supelclean™ sulfoxide and Discovery™ Ag-ION solid phase extraction cartridges).

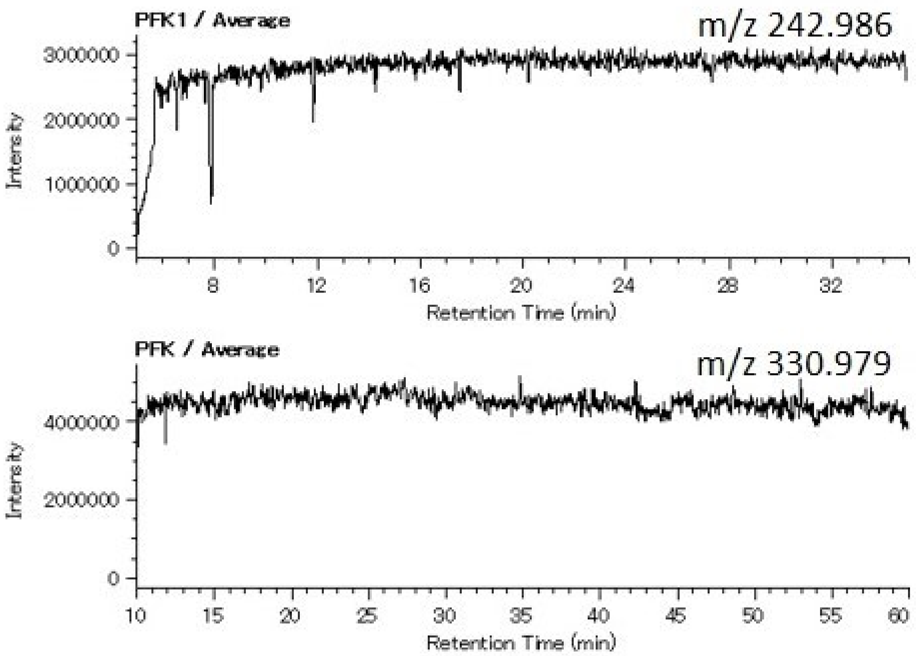

Fig. 2 Mass chromatograms of perfluorokerosene as lock mass (after treatment with Supelclean™ sulfoxide and Discovery™ Ag-ION Ion solid phase extraction cartridges).

3.2 Concentrations and patterns

Table 1 shows the concentrations of primary congeners and homologues in the blubber and liver of the Hubbs' beaked whale. Table S1 shows the concentrations of all congeners and homologues and the composition of a KC-mix. The total PCB concentrations of the blubber and liver were 13,000 and 7,300 ng/g lipid, respectively. The blubber to liver ratio (B/L) was 1.8. Among homologues, hexachlorinated biphenyls had the highest concentrations in both the blubber and liver (6,100 and 3,200 ng/g lipid, respectively), followed by pentachlorinated, heptachlorinated, and tetrachlorinated biphenyls. Monochlorinated and dichlorinated biphenyls were scarcely represented. This type of homologue composition is found in other whales (Corsolini et al., 1995; Fair et al., 2010; Storelli et al., 2012; Wafo et al., 2012). Seven indicator PCBs (PCB-28, 52, 101, 118, 138, 153, and 180) accounted for 41 and 42% of total PCBs in the blubber and liver, respectively. The B/L of tri- to nonachlorinated biphenyls was 1.3–2.0; the ratio of decachlorinated biphenyl (PCB-209) was 0.69 (Table 1), indicating a higher concentration in the liver. Among indicator PCBs, a hexachlorinated biphenyl, PCB-153, had the highest concentration of all congeners, with 1,900 and 990 ng/g lipid in the blubber and the liver, respectively. PCB-118 and 138 had the next highest concentrations. This type of congener composition is shared by other whales (Moon et al., 2010; Yordy et al., 2010; García-Álvarez et al., 2014). Although the B/L of indicator PCBs were 1.0–2.3 (mean 1.8), those of dioxin-like PCBs (DL-PCBs) were 1.0–1.9 (mean 1.0). The blubber to liver ratio of PCB-126 (highest toxic equivalency factor (TEQ), 0.1) was 0.062. DL-PCBs accumulated more in the liver than in the blubber, with 740 and 74 pg-TEQ/g lipid, respectively. PCB concentrations in the blubber of male odontocetes around the world are shown in Table 2. These vary with species, but East Asian odontocetes had 15,000–30,000 ng/g lipid (Kajiwara et al., 2006; Moon et al., 2010), while odontocetes from the Hawaiian Islands had 7,690–50,200 ng/g lipid (median: 17,050 ng/g lipid) (Bachman et al., 2014). Our results match these studies, indicating that the PCB concentrations in the blubber of whales in the northern Pacific Ocean are consistent. The PCB concentration in the blubber of odontocetes in Alaska and polar regions has been reported as 5.8–9,070.0 ng/g lipid (Dorneles et al., 2015; Hoguet et al., 2013; Norstrom and Muir, 1994), while in the Mediterranean Sea off Italy, it was 250–2,100 ng/g lipid (Corsolini et al., 1995). The value obtained in this study is larger than the values in those regions, but smaller than the values in Florida and South Carolina in North America (28,600–255,000 ng/g lipid (Fair et al., 2010; Yordy et al., 2010)), British Columbia (37,400–251,200 ng/g lipid (mean value) (Ross et al., 2000)), and the Canary Islands (13,199–141,796 ng/g lipid (García-Álvarez et al., 2014)).

3.3 Comparison with Kanechlor

The congeners with a high concentration level/ratio in the blubber and liver (PCB-93/95/98, 99, 139/149, and indicator PCBs) accounted for a few percent of all the PCBs in the KC-mix (Table S1), indicating that the most abundant PCBs found in the whale were derived from technical products. PCB-70 and 110 were 4.0% and 3.4% in the KC-mix, respectively, but had low concentrations in the blubber and liver. Abundance relative to the reference was calculated by setting the tri- to octachlorinated biphenyl homologue concentrations in the blubber, liver, and the KC-mix to 100 and comparing the ratio of each congener (Fig. 3). The more abundant congeners in the KC-mix were also present in the blubber and liver. However, the ratios of PCB-28 (2,4,4'), 52 (2,2,5',5'), 74 (2,4,4’,5), 99 (2,2',4,4',5), and 118 (2,3',4,4',5) in the blubber and liver were more than twice those of the KC-mix. In contrast, PCB-31 (2,4',5), 70 (2,3',4',5), and 110 (2,3,3',4',6) were abundant in the KC-mix but were very low in the blubber and liver. PCBs with vicinal nonchlorine substituted positions, such as positions 4 and 5 of PCB-110, are swiftly epoxidized and metabolized in living organisms, while PCBs with chlorine-substituted positions 2, 4 or 2, 4, 5 remain inside living organisms (Kato et al., 1980, Grimm et al., 2015). Hirai et al. (2005) reported that PCB-28, 74, 99, 118, and others remain in human blood; these congeners accumulated in the whale. Although PCB-52 has an easily metabolized chlorine-substituted structure, it accumulated in the whale, so further investigation is warranted.

In recent years, PCB-11 and 209 have received attention because they are used in pigments, and they are scarcely present in technical products such as Kanechlor and Aroclor (Anezaki and Nakano, 2014; Hu and Hornbuckle, 2010). PCB-11 was not identified in the whale tissue samples. Other dichlorinated biphenyls were scarcely detected and were presumably metabolized. The amount of PCB-209 in the blubber and liver was 6.9 and 10 ng/g lipid, respectively. However, the source of this congener could not be determined.

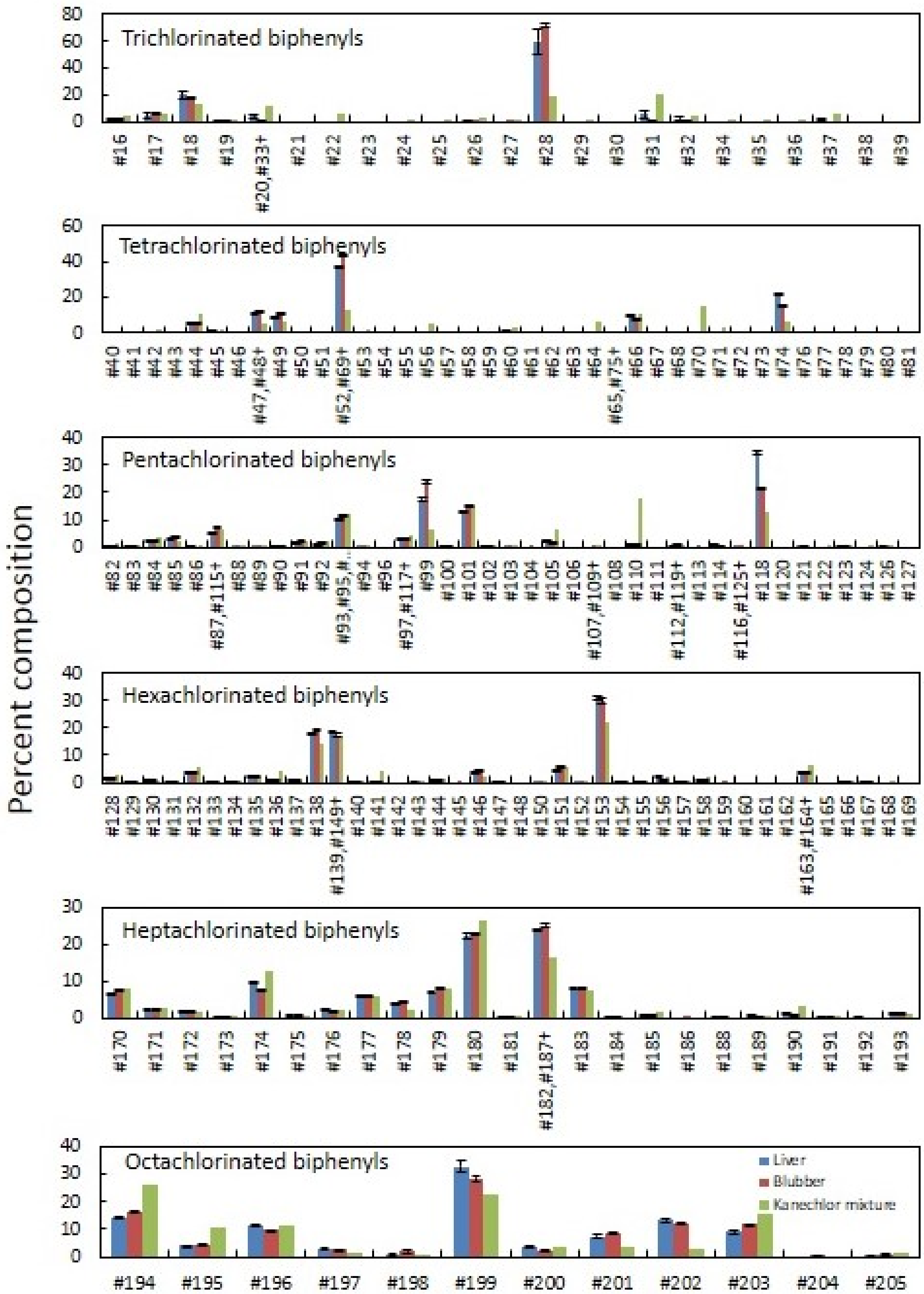

Fig. 3 Ratio of the amount of each congener in the blubber and liver of Hubbs' beaked whale and Kanechlor mix, under the assumption that the concentration of each homolog (tri- to octachlorinated biphenyls) is 100. Cluster congeners that coelute on HT8-PCB column are expressed by "†".

4. Conclusion

We provided the first known analysis of PCBs in the blubber and liver of Hubbs' beaked whale, a rare species. Whale oil was effectively removed from samples with sulfoxide and Ag-ION SPE cartridges. While the total concentration of PCBs in the blubber was higher than in the liver, the DL-PCB concentrations and toxic equivalents were higher in the liver. The PCB concentrations were consistent with those from whales in East Asia and the Hawaiian Islands. The congener composition generally had the same pattern as a Kanechlor mixture, but with some variations. These differences suggest that the fate of congeners in whales is dependent on their metabolism, providing insights into the environmental dynamics, fate, and toxicity evaluation of PCBs.

Acknowledgments

We thank Dr. Keiko Kubo and P.E. Atsushi Takahashi for assistance with the PCBs analysis and Mr. Wojtek Bachara for data on whale strandings.

Funding: This work was supported by the Japan Society for the Promotion of Science [grant number 24710016].

Conflicts of interest: None.

Author contributions:

K. Anezaki: Conception and design of the study.

K. Anezaki: Analysis and interpretation of data.

A. Matsuda, T. Matsuishi: Collection and assembly of sample data.

K. Anezaki, T. Matsuishi: Drafting of the article.

K. Anezaki: Critical revision of the article for important intellectual content.

K. Anezaki: Final approval of the article.

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Electronic supplementary material for:

Concentration and congener pattern of polychlorinated biphenyls in blubber and liver of Hubbs' beaked whale (*Mesoplodon carlhubbsi*), using a sulfoxide and an Ag-ION solid phase extraction cartridge as a simplified cleanup technique for biological samples

Katsunori Anezaki,[1]* Ataka Matsuda,[2] Takashi Matsuishi[3]

[1] Hokkaido Research Organization, Environmental and Geological Research Department, Institute of Environmental Sciences, Hokkaido, Japan

[2] Gradulate School of Fisheries Sciences, Hokkaido University, Hokkaido, Japan

[3] Faculty of Fisheries Sciences, Hokkaido University, Hokkaido, Japan

*Corresponding Author: E-mail: anezaki@hro.or.jp

Number of supplementary manuscript pages: 5

Number of supplementary figures: 2

Fig. S1 Hubbs' beaked whale stranded in Hokkaido, Japan in 2014.

Fig. S2 Sampling location (●) of Hubbs' beaked whale.

Number of supplementary tables: 1

Table S1 All congener concentrations of PCBs (ng/g lipid) for blubber and liver in Hubbs' beaked whale and PCBs weight percentage in Kanechlor (CRM 7906-a Kanechlor Mixture; KC-mix)

Figure S1. Hubbs' beaked whale stranded in Hokkaido, Japan, in 2014.

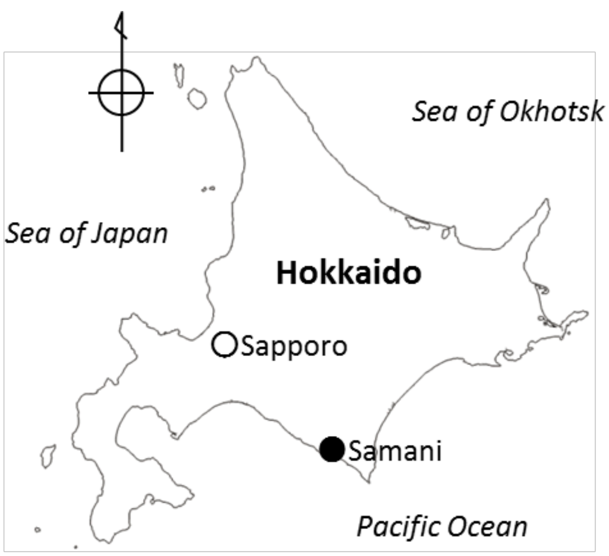

Figure S2. Sampling location (●) of Hubbs' beaked whale.

Table S1. All congener concentrations of polychlorinated biphenyls (ng/g lipid) for blubber and liver in Hubbs' beaked whale and PCBs weight percentage in Kanechlor (CRM 7906-a Kanechlor Mixture; KC-mix)

	Blubber	Liver	KC-mix
Lipid content (%)	89.8	2.0	
PCB-1	0.040	n.d.	0.0058
PCB-2	0.016	n.d.	n.d.
PCB-3	0.21	1.1	0.0029
Mono-CBs	0.27	1.1	0.0087
PCB-4	0.089	n.d.	0.33
PCB-5/8	0.17	0.69	1.2
PCB-6	n.d.	n.d.	0.16
PCB-7	n.d.	n.d.	0.017
PCB-9	n.d.	n.d.	0.036
PCB-10	n.d.	n.d.	0.0063
PCB-11	n.d.	n.d.	n.d.
PCB-12/13	n.d.	n.d.	0.022
PCB-14	n.d.	n.d.	n.d.
PCB-15	n.d.	0.18	0.40
Di-CBs	0.26	0.86	2.1
PCB-16	0.59	0.44	0.95
PCB-17	2.5	1.2	1.1
PCB-18	6.7	5.7	2.7
PCB-19	0.29	0.16	0.24
PCB-20/33	0.21	1.1	2.2
PCB-21	n.d.	n.d.	n.d.
PCB-22	n.d.	n.d.	1.3
PCB-23	n.d.	n.d.	n.d.
PCB-24	n.d.	n.d.	0.041
PCB-25	n.d.	n.d.	0.23
PCB-26	0.023	0.25	0.58
PCB-27	0.025	n.d.	0.19
PCB-28	26	17	3.8
PCB-29	n.d.	n.d.	0.031
PCB-30	n.d.	n.d.	n.d.
PCB-31	0.17	1.6	4.0
PCB-32	0.043	0.62	1.0
PCB-34	n.d.	n.d.	0.013
PCB-35	n.d.	n.d.	0.031
PCB-36	n.d.	n.d.	0.18
PCB-37	n.d.	0.37	1.1
PCB-38	n.d.	n.d.	n.d.
PCB-39	n.d.	n.d.	n.d.
Tri-CBs	37	28	20
PCB-40	1.8	2.2	0.36
PCB-41	1.4	1.4	0.25
PCB-42	5.4	3.3	0.69
PCB-43	n.d.	n.d.	0.058
PCB-44	58	33	2.9
PCB-45	7.0	5.1	0.45
PCB-46	1.8	1.1	0.17
PCB-47/48	120	60	1.4
PCB-49	110	50	1.9
PCB-50	0.34	n.d.	0.0079
PCB-51	2.0	n.d.	0.14
PCB-52/69	450	200	3.6
PCB-53	3.2	1.8	0.44
PCB-54	n.d.	n.d.	0.0095
PCB-55	0.77	1.4	0.063
PCB-56	n.d.	0.43	1.5
PCB-57	0.090	n.d.	n.d.
PCB-58	6.5	n.d.	n.d.
PCB-59	0.073	0.37	0.21
PCB-60	9.9	6.8	1.0
PCB-61	n.d.	n.d.	n.d.
PCB-62	n.d.	n.d.	n.d.
PCB-63	0.15	n.d.	0.15
PCB-64	0.41	n.d.	1.7
PCB-65/75	9.0	2.0	0.034
PCB-66	78	52	3.1
PCB-67	n.d.	n.d.	0.11
PCB-68	n.d.	n.d.	n.d.
PCB-70	0.18	0.37	4.0
PCB-71	8.6	n.d.	1.0
PCB-72	n.d.	n.d.	n.d.
PCB-73	0.28	0.26	n.d.
PCB-74	160	120	1.7
PCB-76	n.d.	n.d.	0.058
PCB-77	5.0	2.6	0.25
PCB-78	0.016	n.d.	n.d.
PCB-79	n.d.	n.d.	n.d.
PCB-80	n.d.	n.d.	n.d.
PCB-81	1.4	1.1	0.011
Tetra-CBs	1100	540	27
PCB-82	12	7.0	0.30
PCB-83	20	9.5	0.12
PCB-84	76	47	0.69
PCB-85	130	62	0.48
PCB-86	n.d.	0.97	0.063
PCB-87/115	250	110	1.3
PCB-88	0.16	0.18	0.0058
PCB-89	2.0	0.40	0.030
PCB-90	21	8.0	0.079
PCB-91	83	43	0.35
PCB-92	58	27	0.42
PCB-93/95/98	390	210	2.3

Cluster congeners that co-eluted on HT8-PCB column are expressed by "/". n.d. = not detected

Table S1. *Continued*

	Blubber	Liver	KC-mix		Blubber	Liver	KC-mix
PCB-94	2.3	1.0	0.014	PCB-150	2.7	1.4	0.0016
PCB-96	n.d.	n.d.	n.d.	PCB-151	340	150	0.90
PCB-97/117	110	59	0.85	PCB-152	1.6	0.530	0.0013
PCB-99	820	360	1.2	PCB-153	1900	990	3.5
PCB-100	8.4	3.8	0.014	PCB-154	27	11	0.013
PCB-101	510	270	2.9	PCB-155	6.7	3.4	n.d.
PCB-102	12	9.5	0.095	PCB-156	76	80	0.33
PCB-103	0.32	0.13	0.038	PCB-157	23	24	0.053
PCB-104	0.062	n.d.	n.d.	PCB-158	82	43	0.38
PCB-105	59	46	1.2	PCB-159	1.2	n.d.	n.d.
PCB-106	n.d.	n.d.	n.d.	PCB-160	n.d.	n.d.	n.d.
PCB-107/109	2.4	n.d.	0.17	PCB-161	n.d.	n.d.	n.d.
PCB-108	n.d.	n.d.	n.d.	PCB-162	n.d.	n.d.	n.d.
PCB-110	29	18	3.4	PCB-163/164	250	130	1.1
PCB-111	n.d.	n.d.	n.d.	PCB-165	n.d.	n.d.	n.d.
PCB-112/119	28	12	0.058	PCB-166	8.9	4.6	0.040
PCB-113	0.40	n.d.	n.d.	PCB-167	12	18	0.095
PCB-114	23	20	0.095	PCB-168	n.d.	n.d.	0.011
PCB-116/125	1.4	0.57	n.d.	PCB-169	n.d.	n.d.	n.d.
PCB-118	730	710	2.4	Hexa-CBs	6100	3200	16
PCB-120	n.d.	n.d.	n.d.				
PCB-121	2.1	0.530	n.d.	PCB-170	190	84	1.1
PCB-122	n.d.	n.d.	0.033	PCB-171	58	29	0.35
PCB-123	7.4	5.8	0.044	PCB-172	47	23	0.21
PCB-124	n.d.	n.d.	0.11	PCB-173	2.0	1.3	0.024
PCB-126	0.44	7.1	0.0063	PCB-174	200	120	1.7
PCB-127	n.d.	n.d.	n.d.	PCB-175	19	8.8	0.063
Penta-CBs	3400	2100	19	PCB-176	46	27	0.30
				PCB-177	160	72	0.79
PCB-128	110	61	0.44	PCB-178	110	51	0.29
PCB-129	11	6.7	0.10	PCB-179	210	91	1.1
PCB-130	75	35	0.12	PCB-180	590	280	3.5
PCB-131	13	8.5	0.033	PCB-181	3.2	2.1	0.0074
PCB-132	220	120	0.90	PCB-182/187	650	310	2.1
PCB-133	28	9.5	0.032	PCB-183	210	100	0.95
PCB-134	38	21	0.13	PCB-184	4.9	2.9	n.d.
PCB-135	140	80	0.36	PCB-185	21	9.4	0.22
PCB-136	79	40	0.63	PCB-186	0.087	n.d.	n.d.
PCB-137	76	36	0.11	PCB-188	4.2	2.3	n.d.
PCB-138	1200	570	2.3	PCB-189	8.9	11	0.039
PCB-139/149	1100	590	2.7	PCB-190	26	13	0.39
PCB-140	3.7	2.0	0.0053	PCB-191	7.8	4.0	0.058
PCB-141	25	14	0.69	PCB-192	n.d.	0.20	n.d.
PCB-142	n.d.	n.d.	n.d.	PCB-193	31	15	0.17
PCB-143	2.9	2.1	0.0053	Hepta-CBs	2600	1300	13
PCB-144	68	34	0.20				
PCB-145	0.16	n.d.	n.d.	PCB-194	43	19	0.79
PCB-146	300	130	0.36	PCB-195	11	5.3	0.32
PCB-147	15	6.2	0.027	PCB-196	25	15	0.33
PCB-148	n.d.	n.d.	n.d.	PCB-197	6.7	4.0	0.035

Cluster congeners that co-eluted on HT8-PCB column are expressed by "/". n.d. = not detected

Table S1. ***Continued***

	Blubber	Liver	KC-mix
PCB-198	5.0	1.0	0.029
PCB-199	74	44	0.69
PCB-200	6.4	4.6	0.11
PCB-201	23	9.7	0.12
PCB-202	32	18	0.090
PCB-203	31	12	0.46
PCB-204	0.43	n.d.	n.d.
PCB-205	1.9	0.65	0.040
Octa-CBs	260	130	3.0
PCB-206	12	5.9	0.21
PCB-207	7.9	6.1	0.032
PCB-208	8.7	5.1	0.035
Nona-CBs	29	17	0.28
Deca-CB (PCB-209)	6.9	10	0.0028
Total PCBs	13000	7300	(100)

n.d. = not detected

Table 1 PCB concentrations (ng/g lipid) and lipid content (%) for blubber and liver in Hubbs' beaked

	blubber	liver	blubber/liver (B/L)
Congener			
PCB-11	n.d.	n.d.	
PCB-28[a]	26	17	1.5
PCB-52[ac]	450	200	2.3
PCB-77[b]	5.0	2.6	1.9
PCB-81[b]	1.4	1.1	1.3
PCB-101[a]	510	270	1.9
PCB-105[b]	59	46	1.3
PCB-114[b]	23	20	1.2
PCB-118[ab]	730	710	1.0
PCB-123[b]	7.4	5.8	1.3
PCB-126[b]	0.44	7.1	0.062
PCB-138[a]	1,200	570	2.1
PCB-153[a]	1,900	990	1.9
PCB-156[b]	76	80	0.95
PCB-157[b]	23	24	0.96
PCB-167[b]	12	18	0.67
PCB-169[b]	n.d.	n.d.	
PCB-180[a]	590	280	2.1
PCB-189[b]	8.9	11	0.81
Homologue			
Mono-CBs	0.27	1.1	0.25
Di-CBs	0.26	0.86	0.30
Tri-CBs	37	28	1.3
Tetra-CBs	1,100	540	2.0
Penta-CBs	3,400	2,100	1.6
Hexa-CBs	6,100	3,200	1.9
Hepta-CBs	2,600	1,300	2.0
Octa-CBs	260	130	2.0
Nona-CBs	29	17	1.7
Deca-CBs(PCB-209)	6.9	10	0.69
Total PCBs[d]	13,000	7,300	1.8
Total indicator PCBs	5,400	3,000	1.8
WHO-TEQ[e]	74	740	0.10
lipid content	89.8	2.0	

n.d. not detected
a Indicator PCBs
b Dioxin-like PCBs
c PCB-52 include co-eluting PCB-69
d Sum of concentrations of all congeners
e WHO-TEQ is calculated for dioxin-like PCBs using WHO-TEF (2006). Unit: pg TEQ/g lipid

Table 2 Comparison of PCB concentrations (ng/g lipid) in the blubber of odontocetes from various locations in the world

Location	Cetacean species	Year	Sex, lifestage, & residency	Sample number	Total PCBs	References
Chukchi Sea-Baffin Bay	Beluga	1983–1989	Male	103	mean 4,670 (2,530–5,580)	Norstrom and Muir (1994)
Baffin Bay	Narwhal	1982–1983	Male	15	mean 5,180	Norstrom and Muir (1994)
Cook Inlet, Alaska	Beluga	1989–2006	Male	15	median 1,640 (441–4,530)	Hoguet et al. (2013)
Eastern Chukchi Sea, Alaska	Beluga	1989–2006	Male	26	median 4,860 (2,190–9,070)	Hoguet et al. (2013)
Italian coastal waters	Bottlenose dolphin Risso's dolphin	1992	Adult & sub adult mal	6	mean 1,170 (250–2,100)	Corsolini et al. (1995)
British Columbia coastal waters, Canac	Killer whale	1993–1996	Adult male		mean ± standard error	
			northern residents	8	37,400 ± 6,100	
			southern residents	4	146,300 ± 32,700	Ross et al. (2000)
			transients	5	251,200 ± 54,700	
Main Hawaiian Islands	Seven delphinid species	1997–2011	Adult male	8	median 17,050 (7,690–50,200)	Bachman et al. (2014)
Japan coastal waters	Stejneger's beaked whale	2000–2001	Male	5	mean 19,000 (17,000–20,000)	Kajiwara et al. (2006)
Japan coastal waters	Melon-head whale	2001	Male	5	mean 24,000 (15,000–30,000)	Kajiwara et al. (2006)
Western Antarctic Peninsular waters	Humpback whale	2000–2003	Male	24	median 40 (5.8–590.0)	Dorneles et al. (2015)
Sarasota Bay, FL, USA	Bottlenose dolphin	2002–2005	Adult male	9	geomean 61,300 (44,100–85,400)	Yordy et al. (2010)
Indian River Lagoon, FL, USA	Bottlenose dolphin	2003–2005	Adult male	33	geomean 79,800 (35,000–227,000)	Fair et al. (2010)
Charleston, SC, USA	Bottlenose dolphin	2003–2005	Adult male	36	geomean 94,000 (28,600–255,000)	Fair et al. (2010)
Korea coastal waters	Long-beaked common dolphi	2006	Adult male	12	mean 15,000 (1,200–25,000)	Moon et al. (2010)
Canary Islands	Bottlenose dolphin	2009–2011	Adult male	3	(13,199–141,796)	García-Álvarez et al. (2014)
Samani coast, Hokkaido, Japan	Hubbs' beaked whale	2014	Adult male	1	13,000	This study

Table S1. All congener concentrations of PCB (ng/g lipid) for blubber and liver in Hubbs' beaked whale and PCB weight percentage in Kanechlor (CRM 7906-a Kanechlor Mixture; KC-mix)

	Blubber	Liver	KC-mix		Blubber	Liver	KC-mix
				PCB-44	58	33	2.9
Lipid content (%)	89.8	2.0		PCB-45	7.0	5.1	0.45
				PCB-46	1.8	1.1	0.17
PCB-1	0.040	n.d.	0.0058	PCB-47/48	120	60	1.4
PCB-2	0.016	n.d.	n.d.	PCB-49	110	50	1.9
PCB-3	0.21	1.1	0.0029	PCB-50	0.34	n.d.	0.0079
Mono-CBs	0.27	1.1	0.0087	PCB-51	2.0	n.d.	0.14
				PCB-52/69	450	200	3.6
PCB-4	0.089	n.d.	0.33	PCB-53	3.2	1.8	0.44
PCB-5/8	0.17	0.69	1.2	PCB-54	n.d.	n.d.	0.0095
PCB-6	n.d.	n.d.	0.16	PCB-55	0.77	1.4	0.063
PCB-7	n.d.	n.d.	0.017	PCB-56	n.d.	0.43	1.5
PCB-9	n.d.	n.d.	0.036	PCB-57	0.090	n.d.	n.d.
PCB-10	n.d.	n.d.	0.0063	PCB-58	6.5	n.d.	n.d.
PCB-11	n.d.	n.d.	n.d.	PCB-59	0.073	0.37	0.21
PCB-12/13	n.d.	n.d.	0.022	PCB-60	9.9	6.8	1.0
PCB-14	n.d.	n.d.	n.d.	PCB-61	n.d.	n.d.	n.d.
PCB-15	n.d.	0.18	0.40	PCB-62	n.d.	n.d.	n.d.
Di-CBs	0.26	0.86	2.1	PCB-63	0.15	n.d.	0.15
				PCB-64	0.41	n.d.	1.7
PCB-16	0.59	0.44	0.95	PCB-65/75	9.0	2.0	0.034
PCB-17	2.5	1.2	1.1	PCB-66	78	52	3.1
PCB-18	6.7	5.7	2.7	PCB-67	n.d.	n.d.	0.11
PCB-19	0.29	0.16	0.24	PCB-68	n.d.	n.d.	n.d.
PCB-20/33	0.21	1.1	2.2	PCB-70	0.18	0.37	4.0
PCB-21	n.d.	n.d.	n.d.	PCB-71	8.6	n.d.	1.0
PCB-22	n.d.	n.d.	1.3	PCB-72	n.d.	n.d.	n.d.
PCB-23	n.d.	n.d.	n.d.	PCB-73	0.28	0.26	n.d.
PCB-24	n.d.	n.d.	0.041	PCB-74	160	120	1.7
PCB-25	n.d.	n.d.	0.23	PCB-76	n.d.	n.d.	0.058
PCB-26	0.023	0.25	0.58	PCB-77	5.0	2.6	0.25
PCB-27	0.025	n.d.	0.19	PCB-78	0.016	n.d.	n.d.
PCB-28	26	17	3.8	PCB-79	n.d.	n.d.	n.d.
PCB-29	n.d.	n.d.	0.031	PCB-80	n.d.	n.d.	n.d.
PCB-30	n.d.	n.d.	n.d.	PCB-81	1.4	1.1	0.011
PCB-31	0.17	1.6	4.0	Tetra-CBs	1100	540	27
PCB-32	0.043	0.62	1.0				
PCB-34	n.d.	n.d.	0.013	PCB-82	12	7.0	0.30
PCB-35	n.d.	n.d.	0.031	PCB-83	20	9.5	0.12
PCB-36	n.d.	n.d.	0.18	PCB-84	76	47	0.69
PCB-37	n.d.	0.37	1.1	PCB-85	130	62	0.48
PCB-38	n.d.	n.d.	n.d.	PCB-86	n.d.	0.97	0.063
PCB-39	n.d.	n.d.	n.d.	PCB-87/115	250	110	1.3
Tri-CBs	37	28	20	PCB-88	0.16	0.18	0.0058
				PCB-89	2.0	0.40	0.030
PCB-40	1.8	2.2	0.36	PCB-90	21	8.0	0.079
PCB-41	1.4	1.4	0.25	PCB-91	83	43	0.35
PCB-42	5.4	3.3	0.69	PCB-92	58	27	0.42
PCB-43	n.d.	n.d.	0.058	PCB-93/95/98	390	210	2.3

Cluster congeners that co-eluted on HT8-PCB column are expressed by "/". n.d. = not detected

Table S1 continued

	Blubber	Liver	KC-mix		Blubber	Liver	KC-mix
PCB-94	2.3	1.0	0.014	PCB-150	2.7	1.4	0.0016
PCB-96	n.d.	n.d.	n.d.	PCB-151	340	150	0.90

PCB-97/117	110	59	0.85
PCB-99	820	360	1.2
PCB-100	8.4	3.8	0.014
PCB-101	510	270	2.9
PCB-102	12	9.5	0.095
PCB-103	0.32	0.13	0.038
PCB-104	0.062	n.d.	n.d.
PCB-105	59	46	1.2
PCB-106	n.d.	n.d.	n.d.
PCB-107/109	2.4	n.d.	0.17
PCB-108	n.d.	n.d.	n.d.
PCB-110	29	18	3.4
PCB-111	n.d.	n.d.	n.d.
PCB-112/119	28	12	0.058
PCB-113	0.40	n.d.	n.d.
PCB-114	23	20	0.095
PCB-116/125	1.4	0.57	n.d.
PCB-118	730	710	2.4
PCB-120	n.d.	n.d.	n.d.
PCB-121	2.1	0.530	n.d.
PCB-122	n.d.	n.d.	0.033
PCB-123	7.4	5.8	0.044
PCB-124	n.d.	n.d.	0.11
PCB-126	0.44	7.1	0.0063
PCB-127	n.d.	n.d.	n.d.
Penta-CBs	3400	2100	19
PCB-128	110	61	0.44
PCB-129	11	6.7	0.10
PCB-130	75	35	0.12
PCB-131	13	8.5	0.033
PCB-132	220	120	0.90
PCB-133	28	9.5	0.032
PCB-134	38	21	0.13
PCB-135	140	80	0.36
PCB-136	79	40	0.63
PCB-137	76	36	0.11
PCB-138	1200	570	2.3
PCB-139/149	1100	590	2.7
PCB-140	3.7	2.0	0.0053
PCB-141	25	14	0.69
PCB-142	n.d.	n.d.	n.d.
PCB-143	2.9	2.1	0.0053
PCB-144	68	34	0.20
PCB-145	0.16	n.d.	n.d.
PCB-146	300	130	0.36
PCB-147	15	6.2	0.027
PCB-148	n.d.	n.d.	n.d.
PCB-152	1.6	0.530	0.0013
PCB-153	1900	990	3.5
PCB-154	27	11	0.013
PCB-155	6.7	3.4	n.d.
PCB-156	76	80	0.33
PCB-157	23	24	0.053
PCB-158	82	43	0.38
PCB-159	1.2	n.d.	n.d.
PCB-160	n.d.	n.d.	n.d.
PCB-161	n.d.	n.d.	n.d.
PCB-162	n.d.	n.d.	n.d.
PCB-163/164	250	130	1.1
PCB-165	n.d.	n.d.	n.d.
PCB-166	8.9	4.6	0.040
PCB-167	12	18	0.095
PCB-168	n.d.	n.d.	0.011
PCB-169	n.d.	n.d.	n.d.
Hexa-CBs	6100	3200	16
PCB-170	190	84	1.1
PCB-171	58	29	0.35
PCB-172	47	23	0.21
PCB-173	2.0	1.3	0.024
PCB-174	200	120	1.7
PCB-175	19	8.8	0.063
PCB-176	46	27	0.30
PCB-177	160	72	0.79
PCB-178	110	51	0.29
PCB-179	210	91	1.1
PCB-180	590	280	3.5
PCB-181	3.2	2.1	0.0074
PCB-182/187	650	310	2.1
PCB-183	210	100	0.95
PCB-184	4.9	2.9	n.d.
PCB-185	21	9.4	0.22
PCB-186	0.087	n.d.	n.d.
PCB-188	4.2	2.3	n.d.
PCB-189	8.9	11	0.039
PCB-190	26	13	0.39
PCB-191	7.8	4.0	0.058
PCB-192	n.d.	0.20	n.d.
PCB-193	31	15	0.17
Hepta-CBs	2600	1300	13
PCB-194	43	19	0.79
PCB-195	11	5.3	0.32
PCB-196	25	15	0.33
PCB-197	6.7	4.0	0.035

Cluster congeners that co-eluted on HT8-PCB column are expressed by "/". n.d. = not detected

Table S1 contunued

	Blubber	Liver	KC-mix
PCB-198	5.0	1.0	0.029
PCB-199	74	44	0.69
PCB-200	6.4	4.6	0.11

PCB-201	23	9.7	0.12
PCB-202	32	18	0.090
PCB-203	31	12	0.46
PCB-204	0.43	n.d.	n.d.
PCB-205	1.9	0.65	0.040
Octa-CBs	260	130	3.0
PCB-206	12	5.9	0.21
PCB-207	7.9	6.1	0.032
PCB-208	8.7	5.1	0.035
Nona-CBs	29	17	0.28
Deca-CB (PCB-209)	6.9	10	0.0028
Total PCBs	13000	7300	(100)

n.d. = not detected