

## Species-Specific Accumulation of Dioxin Related Compounds in Cetaceans Collected from Japanese Coastal Waters

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### Introduction

Polychlorinated dibenzo-*p*-dioxins (PCDDs) and dibenzofurans (PCDFs) are extremely hazardous and persistent chemicals identified as contaminants in chlorophenols, herbicides, fly ash and other incineration products. Dioxin-like PCBs including non- and mono-*ortho* coplanar PCBs are referred to as dioxin related compounds and are evaluated on par with PCDD/Fs in environmental risks since they have a high toxicity, similar to that of PCDD/Fs<sup>1</sup>. These congeners have a range of physicochemical characteristics, which profoundly affect their persistence, environmental distribution, and bioaccumulation in aquatic food chains.

Fish-eating wildlife such as marine mammals are particularly vulnerable to such contamination given their long lives, high trophic level, relative inability to metabolize many persistent organic pollutants (POPs), and the biomagnification of these contaminants in aquatic food chains. However, most studies dealing with PCDDs and PCDFs in marine mammals have been carried out on pinnipeds, and data on PCDD/Fs levels in cetaceans are scarce.

The present study is aimed at understanding the recent pattern of contamination by dioxin related compounds including non- and mono-*ortho* coplanar PCBs and PCDD/Fs in three cetacean species collected from Japanese coastal waters during 1998-2001, and also to discuss the factors determining the accumulation.

## Materials and Methods

**Samples.** Three cetacean species collected from Japanese coastal waters during 1998-2001 were used in this study, a coastal species (Finless porpoise, *Neophocaena phocaenoides*,  $n=5$ ) stranded along Seto-Inland Sea, and two offshore species collected at northern Japan (Dall's porpoise, *Phocoenoides dalli*,  $n=10$ ) and another with unknown distribution stranded along the coastal line of the Japan Sea (Stejneger's beaked whale, *Mesoplodon stejnegeri*,  $n=10$ ) (Figure 1). For Dall's porpoises, two populations, *dalli*- and *truei*-type, that have different migration routes were included in this study. Blubber samples were employed for chemical analysis.

**Chemical analysis.** Blubber samples were extracted in a Soxhlet apparatus with dichloromethane. An aliquot of the extract, after adding  $^{13}\text{C}_{12}$ -labeled internal standards, was added to a gel permeation chromatography (GPC) column for lipid removal. The GPC fraction containing organohalogenes was concentrated and passed through an activated silicagel S-1 column for clean-up. Separation of PCDD/Fs and coplanar PCBs fraction was performed by passing through activated alumina and carbon-dispersed silicagel packed in a glass column.  $^{13}\text{C}_{12}$ -labeled internal standards were added to final solution prior to GC-MSD analysis. Identification and quantification of 2,3,7,8-substituted PCDD/Fs and non-*ortho* coplanar PCBs was performed using HRGC (Agilent 6890)-HRMS (JEOL JMS-700D), and mono-*ortho* coplanar PCBs using HRGC (Agilent 6890)-DFMS (JEOL GCmate II). TEQs (toxic equivalencies) were calculated using WHO-TEFs for human and mammals <sup>1</sup>.

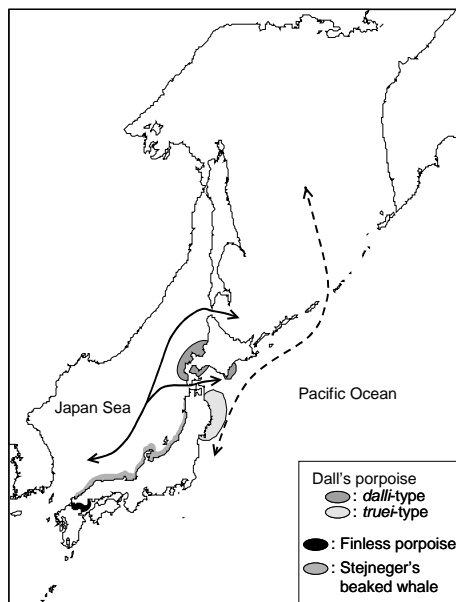


Figure 1. Sampling locations of cetaceans collected from Japanese coastal waters. Solid and dashed arrows indicate possible migration routes of two populations of Dall's porpoise around Japan, *dalli*- and *truei*-type, respectively.

## Results and Discussion

The residual concentrations of non- and mono-*ortho* coplanar PCBs and PCDD/Fs in the blubber of finless porpoises, Dall's porpoises and Stejneger's beaked whales are summarized in Table 1. Concentrations of 2,3,7,8-substituted PCDD/Fs and total coplanar PCBs in cetaceans from Japan varied from 8.9 to 290 pg/g lipid wt and from 150 to 1,000 ng/g lipid wt, respectively. It is noteworthy that coplanar PCBs were detected at much higher levels, from 4,000 to 15,000 times higher, than toxic PCDD/Fs in all the samples analyzed. This implies that these PCB congeners may have significant toxic impact on those forms in spite of their relative low toxic potency in comparison to PCDD/Fs.

*Japan Sea vs. Pacific Ocean*

Table 1. Concentrations and TEQs (pg/g lipid wt.) of PCDD/Fs and coplanar PCBs in the blubber of cetaceans collected from Japanese coastal waters

Species Collected year	Finless porpoise 1998-2000	Dall's porpoise ( <i>truei</i> -type) 2000	Dall's porpoise ( <i>dalli</i> -type) 2000	Stejneger's beaked whale 2000-2001	Stejneger's beaked whale 1998-2000
Sex	Male	Male	Male	Male	Female
<i>n</i>	5	5	5	5	5
Lipid (%)	69 ± 14	85 ± 1.0	80 ± 3.8	75 ± 2.7	81 ± 2.7
<b>Concentration</b>					
Total PCDDs	130 ± 69	3.7 ± 1.6	8.8 ± 5.5	12 ± 1.3	12 ± 4.4
Total PCDFs	39 ± 17	13 ± 4.1	22 ± 14	36 ± 4.7	36 ± 6.8
Total PCDD/Fs	170 ± 86	17 ± 5.7	31 ± 19	47 ± 5.9	49 ± 11
Total non-ortho Co-PCBs	640 ± 180	230 ± 60	490 ± 300	1,500 ± 310	1,200 ± 300
Total mono-ortho Co-PCBs	720,000 ± 210,000	180,000 ± 36,000	450,000 ± 74,000	730,000 ± 180,000	320,000 ± 67,000
Total Coplanar PCBs	720,000 ± 210,000	180,000 ± 36,000	450,000 ± 74,000	740,000 ± 180,000	320,000 ± 68,000
<b>TEQs</b>					
Total PCDDs	5.1 ± 1.6	1.2 ± 0.49	1.6 ± 1.0	1.9 ± 0.24	2.1 ± 0.41
Total PCDFs	6.1 ± 2.0	2.6 ± 0.90	3.6 ± 2.6	4.4 ± 0.70	4.4 ± 0.83
Total PCDD/Fs	11 ± 3.5	3.8 ± 1.4	5.2 ± 3.7	6.3 ± 0.89	6.4 ± 1.1
Total non-ortho Co-PCBs	11 ± 2.8	7.8 ± 2.3	19 ± 15	77 ± 17	60 ± 17
Total mono-ortho Co-PCBs	98 ± 30	24 ± 5.5	69 ± 13	150 ± 37	56 ± 12
Total Coplanar PCBs	110 ± 31	32 ± 7.4	88 ± 26	220 ± 53	120 ± 27
Total TEQs	120 ± 35	36 ± 8.8	93 ± 30	230 ± 54	120 ± 28

Values are means ± standard deviation

To elucidate the contamination status of dioxin related compounds, Dall's porpoise presents a unique opportunity because we could obtain samples for comparison from the Japan Sea and the Pacific Ocean on their migration routes (Figure 1). When residue levels in Dall's porpoises from different populations were compared, concentrations of all the contaminants detected in *truei*-type porpoise from the Pacific coast of northern Japan were significantly lower than *dalli*-type, which migrate inside the Japan Sea and the Sea of Okhotsk (Table 1, Figure 1). Among the congeners analyzed, *truei*-type porpoise from the Pacific coast of Japan showed less contribution of O<sub>8</sub>CDD and higher 23478-P<sub>5</sub>CDF (Figure 2). On the other hand, samples from the Japan Sea including *dalli*-type porpoise and Stejneger's beaked whale showed similar composition pattern with high O<sub>8</sub>CDD, 234678-H<sub>6</sub>CDF and 1234678-H<sub>7</sub>CDF proportions. These results indicate that the load of dioxin related compounds in the Japan Sea is higher than the Pacific, indicating the possible contribution of these compounds as contaminants in the effluents containing pentachlorophenol (PCP) used in the agriculture in the countries surrounding the Japan Sea. Uncontrolled and low-temperature burning of wastes in dumping sites is one of the ways generating PCDD/Fs in Asian developing countries<sup>2</sup>. Emissions from China and Southeast Asia are presumable contamination sources to the Japan Sea. Additionally, the closed ecosystem of the Japan Sea has resulted in the accumulation of relatively less volatile compounds. Our previous research revealed that HCHs and DDTs concentrations in *dalli*-type porpoises from the Japan Sea were also significantly higher than those in *truei*-type of the Pacific Ocean and the effluents from surrounding counties of the Japan Sea were suggested as a possible source<sup>3</sup>.

*Coastal vs. Offshore Species*

Contamination levels of PCDDs were highest in finless porpoise, which represents a coastal species in Japan, followed by Stejneger's beaked whale, *dalli*- and *truei*-type Dall's porpoises (Table 1). Congener profile in finless porpoise with highest contribution by O<sub>8</sub>CDD among

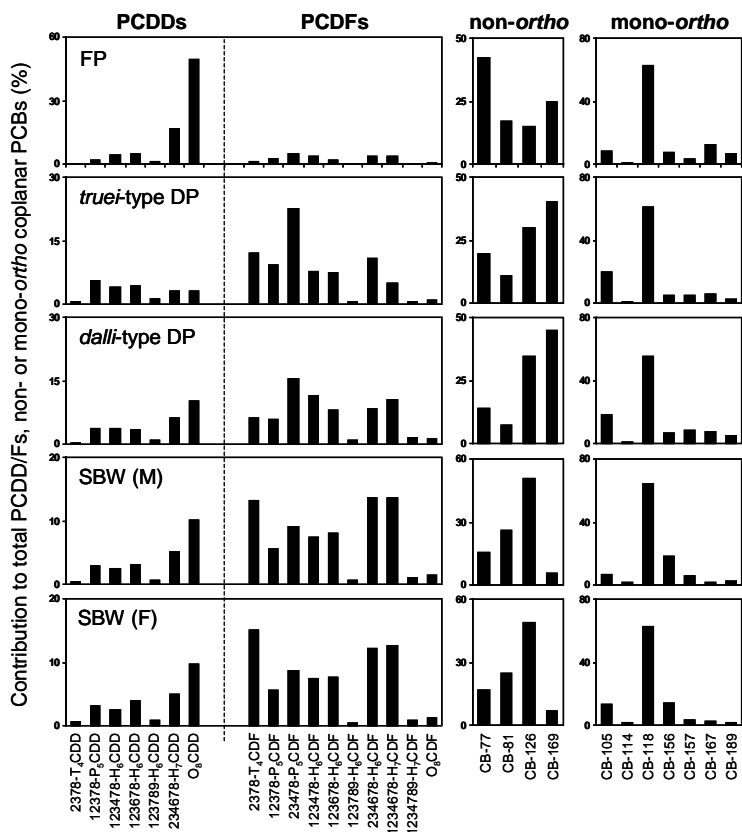


Figure 2. Congener-specific patterns of PCDD/Fs, non- or mono-ortho coplanar PCBs in the blubber of cetaceans collected from Japanese coastal waters. DP: Dall's porpoise; SBW: Stejneger's beaked whale; FP: Finless porpoise; F: female; M: male.

PCDD/Fs was apparently different from those observed in offshore species (Figure 2). Concentrations of PCDDs in coastal species (finless porpoise) were higher than PCDFs, while PCDFs were higher in offshore species. These results indicate that PCDFs were easily transported to offshore in contrast to PCDDs which remain locally. Due to the non-detection of PCDDs and the identification of PCDFs in killer whales from open ocean, Ono *et al.*<sup>4</sup> concluded that PCDFs have greater potentiality of being a global pollutant than PCDDs. Among the coplanar PCBs, Stejneger's beaked whale showed the highest coplanar PCBs concentration among the samples analyzed with higher contribution from non-ortho coplanar PCBs (Table 1). Interestingly, finless porpoises retained a high composition of CB-77 even though this congener is most easily metabolized in animal bodies (Figure 2), indicating continuous exposure by PCB formulations that contain CB-77 in high amount.

### Gender Differences

Generally, cetacean females off-load majority of their organochlorine burden to their calf during reproduction. Effects of age and sex of cetaceans on accumulation of dioxin related compounds are scarcely reported so far. In the present study, gender difference in accumulation pattern of dioxin related compounds were clarified using Stejneger's beaked whales. As shown in Table 1, in contrast to mono-*ortho* coplanar PCBs, there were no gender-related patterns in PCDDs, PCDFs and non-*ortho* coplanar PCBs in beaked whales. These results may partly reflect a limited food chain biomagnification, or low dietary concentrations of these compounds. However, these results more likely indicate minimal bioaccumulation in beaked whales, as a consequence of the metabolism and excretion of planar compounds.

### Toxic Equivalents (TEQs)

Total TEQs levels in Stejneger's beaked whale were highest, followed by finless porpoise and Dall's porpoise (Table 1). Figure 3 shows percentage contributions of PCDDs and PCDFs, non- and mono-*ortho* coplanar PCBs to total TEQs. In general, the percentage contribution to total TEQs by mono-*ortho* coplanar PCBs were approximately 40~80%, whereas PCDD/Fs accounted for only 10% (Figure 3). Interestingly, contribution by non-*ortho* coplanar PCBs in Stejneger's beaked whale was higher than other species and, in female whales the contribution by non-*ortho* coplanar PCBs exceeded that of the mono-*ortho* coplanar PCBs.

In Japanese cetaceans studied, it was found that the toxic load of TEQs by PCDD/Fs is much lower than that of coplanar PCBs, again indicating that toxic coplanar PCBs are at present the most hazardous known organochlorines for cetaceans around Japan.

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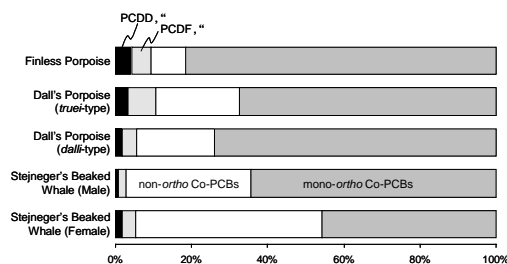


Figure 3. Comparison of TEQ compositions in cetaceans from Japanese coastal waters