Bioaccumulation of non 2,3,7,8-substituted PCDFs in Fishes of Environmental Samples

Nobuyasu Seike [A]. **Takanori Sakiyama** [B]. **Muneaki Matsuda** [A]. **Tadaaki Wakimoto** [A]

[A] Department of Environment Conservation, Ehime University
Tarumi 3-5-7, Matsuyama 790, Japan

[B] Osaka City Institute of Pablic Health and Environmental Sciences
8-34, Tojo-cho, Tennoji-ku, Osaka 543, Japan

INTRODUCTION

Polychlorinated dibenzo-*p*-dioxins and Polychlorinated dibenzofurans (PCDDs/DFs) have been emitted from various sources to the environment. They are mostly municipal waste incinerators, metal works (Iron, steel and nonferrous), pulp mills, polychlorinated biphenyls (PCBs) and organochlorine pesticides. Several studies reported that PCDDs/DFs from these sources were detected in biota of environmental samples [1),2)]. General reports had mentioned about only 2,3,7,8-sustituted isomers. But there are a few reports that non 2,3,7,8-substituted isomers were detected in the crustacean [3)]. On some dose experimental studies, when fishes and animals were exposed by PCDDs/DFs, not only 2,3,7,8-substituted isomers but also non 2,3,7,8-substituted isomers were accumulated in the tested fishes and animals [4),5)].

Fishes occupy an important position in Japanese food stuff. If the non 2,3,7,8-substituted isomers occured in fishes, humans may be exposed by these compounds via fishes.

In this study, we tried to extract the fat of edible parts with large quantities in order to achieve much lower detection limit. The objection of this study was to detect 2,3,7,8-substitutd isomers and non 2,3,7,8-substituted isomers from fishes.

EXPERIMENTAL

Samples

Fish samples were collected from some estuaries and ponds in Matsuyama in Japan. Fish samples are Mullet (*Mugil cephalus*), Redlip mullet (*Liza haematocheila*), Large-mouth bass (*Micropterus salmoides*) and Bluegill (*Lepomis macrochirus*). These were divided into the edible parts and internal organs (only the edible parts were used). The edible parts were homogenized by the mincer. Samples were preserved in the freezer at -30 ℃ until analysis were perfomed.

Extraction and Analysis

In order to ahieve much lower detection limit, we used large quantities (about 400g) for each samples. The edible parts were Soxhlet-extracted with dichloromethane for 24 hours.

After extraction, alkali digestion and H_2SO_4 treatment were carried out. The clean up were performed with silicagel, alumina and actived carbon column chromatography. We used HPLC (High Performance Liquid Chromatography) as a final stage clean up and determined by HRGC-HRMS (JEOL SX-102A) with capillary column SIL-PAK 88.

RESULTS AND DISCUSSION

Most components of PCDDs/DFs were detected in all fish samples.

Polychlorinated dibenzo-p-dioxins (PCDDs)

All 2,3,7,8-substituted isomers were detected (0.2 - 8.6 pg/g Fat bases) in fish samples excluding OCDD . Some non 2,3,7,8-substituted isomers were detected (Not detect - 3.0 pg/g Fat bases) in a few samples. Concentrations of these isomers were lower than 2,3,7,8-substituted isomers in each congeners. Sijm et al. reported that a large number of PCDDs/DFs isomers were taken up by the tested fishes at 11 hours of exposure. But at 120 hours exposure, only 2,3,7,8-substituted isomers were detected [4] . The reason of that might be because non 2,3,7,8-substituted PCDDs isomers were eliminated rapidly. Therefore, it was appeared that only 2,3,7,8-substituted PCDDs isomers were accumulated in fishes of this study.

Polychlorinated dibenzofurans (PCDFs)

Figure 1. shows the isomer compositions of PCDFs congeners in the Mullet and Redlip mullet. They graphed relatively when the most predominant isomer was 1.

TeCDFs Most of TeCDFs isomers (38 out of 34) were detected in all samples. The most predominant isomer was 2,3,7,8-TeCDF. Its concentration levels were 0.3 - 24.6 pg/g and other TeCDFs isomers were Not detect - 12.7 pg/g. 2,3,6,8-, 1,2,6,9-, 2,3,4,8- and 1,2,8,9-TeCDF were not detected in all samples.

PeCDFs Most of PeCDFs isomers (28 out of 21) were detected in all samples. The most predominant isomer was 2,3,4,7,8-PeCDF. The next predominant isomers were 1,2,4,7,8-, 1,2,4,6,8-, 1,2,3,7,8-/1,2,3,4,8-, 2,3,4,6,7-, 1,2,3,6,8-/1,3,4,7,8-, and 1,4,6,7,8-/1,2,3,4,7-PeCDFs. The ratio of non 2,3,7,8- substituted isomers to 2,3,7,8-substituted iomers were higher than that of TeCDFs. 1,3,4,7,9-, 1,3,4,6,9-, 1,2,3,7,9-, 1,2,6,7,9-, 1,2,3,6,9-, 1,2,3,4,9- and 1,2,3,8,9-PeCDF were not detected in all samples.

HxCDFs Most of (16 out of 15) HxCDFs isomers were detected in all samples and non 2,3,7,8-substituted isomer (1,2,4,6,8,9-HxCDF) had the ratio similar to 2,3,7,8-substituted isomers. The most predominant isomers were 1,2,3,6,7,8-, 2,3,4,6,7,8- and 1,2,4,6,8,9-HxCDF in HxCDFs. The next predominant isomers were 1,2,4,6,7,8-, 1,2,4,6,7,9-/1,2,3,4,7,8-, 1,2,3,4,6,9-/1,2,3,6,8,9-, 1,3,4,6,7,8-, 1,2,3,4,6,7- and 1,2,3,4,6,8-HxCDF. The ratio of non 2,3,7,8-substituted HxCDFs isomers were higher than that of TeCDFs and PeCDFs. But concentrations of HxCDFs isomers were lower than that of TeCDFs and PeCDFs. 1,2,3,6,7,9-HxCDF was not detected in all samples.

HpCDFs 1,2,3,4,6,7,8-HpCDF was the most predominat isomers in all samples (0.8 - 7.2 pg/g Fat bases). There were few non 2,3,7,8-substituted isomers in all samples.

OCDF OCDF was not detected in all samples.

For the PCDFs, various 2,3,7,8-subsituted and non 2,3,7,8-substituted isomers were detected in fishes. When the numbers of chlorine atoms increased, the predominance of 2,3,7,8-substituted isomers were vanished (TeCDFs > PeCDFs > HxCDFs). To say in other words, the ratio of non 2,3,7,8-substituted isomers in each congeners were increased (HxCDFs > PeCDFs > TeCDFs). This shows that non 2,3,7,8-substituted isomers tend to be accumulated with the number of chlorine atoms increased. We confirmed previously that TeCDFs isomers tended to be more taken up than PeCDFs and HxCDFs. But an elimination of TeCDFs isomers was more rapidly than PeCDFs and HxCDFs. So, high persistant 2,3,7,8-TeCDF was predominantly detected. For the HxCDFs, many non 2,3,7,8-substituted isomers deteced in fishes. We assumed that an elimination was lower than TeCDFs and

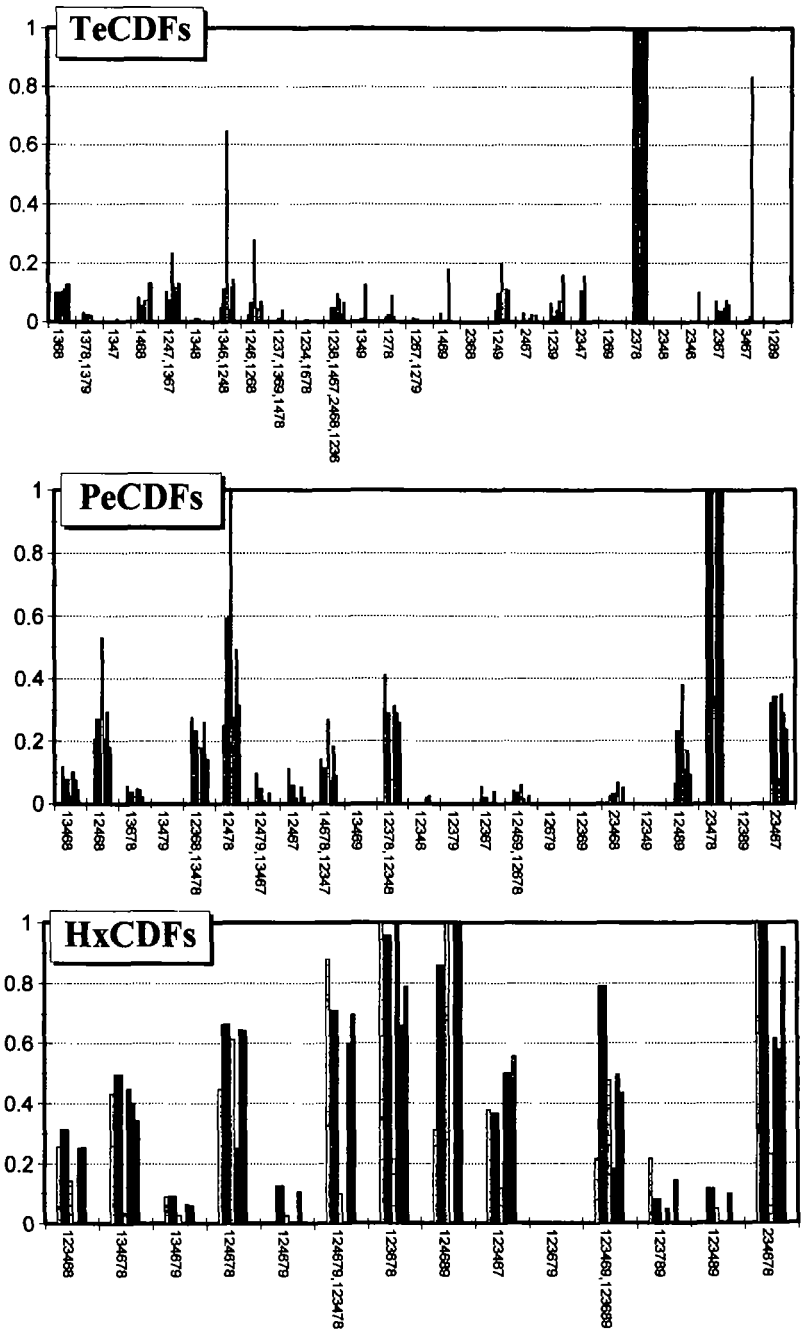

Figure 1. Isomer compositions of PCDFs congeners in Mullet and Ledlip Mullet

PeCDFs if HxCDFs isomers were taken up by fishes once. Therefore, a lot of HxCDFs isomers were persisted in fishes.

In this study, we confirmed that a lot of non 2,3,7,8-substituted isomers existed in the edible parts of fishes. If various non 2,3,7,8-substituted isomers exist in the fishes on the market, humans would been exposed to these compounds. Kuroki et al. confirmed that non 2,3,7,8-substituted isomers persisted in humans (Yusho patients) [6], what is more, Safe et al. reported that a large number of non 2,3,7,8-substituted PCDFs isomers were potentially toxic like 2,3,7,8-substituted isomers [7]. But International-TEFs is inapplicable for non 2,3,7,8-substituted isomers. We suggest that the risk assessment should be performed for not only 2,3,7,8-substituted isomers but also non 2,3,7,8-substituted isomers, in particularly next predominant isomers in this study.

CONCLUSIONS

- Most of PCDFs isomers (38 out of 34 TeCDFs, 28 out of 21 PeCDFs, 16 out of 15 HxCDFs and 4 out of 4 HpCDFs isomers) were detected in fishes of environmental samples.
- Most of 2,3,7,8-substituted isomers were persisted in fishes.
- The risk assessment for PCDDs/DFs should be performed including non 2,3,7,8-substituted isomers in environmental samples.

REFERENCES

1) M.Van den Berg, F.Blank, C.Heeremans, H.Wagenaar, K.Olie (1987) : Presence of Polychlorinated Dibenzo-*p*-dioxins and Polychlorinated Dibenzofurans in Fish-Eating Birds and Fish from The Netherlands. *Arch. Eiviron. Contam. Toxicol.* 16, 149-158

2) C.Rappe, Per-Anders Bergqvist, Lars-Owe Kjeller (1989) : Levels, Trends and Patterns of PCDDs and PCDFs in Scandinavian Environmental Samples. *Chemosphere*, Vol.18, pp.651-658

3) C.Rappe, Per-Anders Bergqvist, Lars-Owe Kjeller, S.Swanson (1991) : Levels and Patterns of PCDD and PCDF Contamination in Fish, Crabs and Lobsters from Newark Bay and The New York Bight. *Chemosphere*, Vol.22, pp.239-266

4) D.T.H.M.Sijm, H.Wever, A.Opperhuizen (1993) : Congener-Specific Biotransformation and Bioaccumlation of PCDDs and PCDFs from Fly Ash in Fish *Environmental Toxicology and Chemstry,* Vol.12, pp.1895-1907

5) U.G.Ahlborg, H.Håkansson, G.Lindström, C.Rappe (1990) : Studies on the Retention of Individual Polychlorinated Dibenzofurans (PCDFs) in the Liver of Different Species. *Chemosphere,* Vol.20, pp. 1235-1240

6) H.Kuroki, K.Haraguchi, Y.Masuda (1987) : Polychlorinated Dibenzofuran (PCDF) Congeners in the Tissues of Patients with Yusho and Normal Japanese *Chemosphere*, Vol.16, pp.2039-2046

7) S.Safe, T.Sawyer, G.Mason, S.Bandiera, B.Keys, M.Romkes, J.Piskorska-Pliszczynska, B.Zmudzuka, L.Safe (1985) : Polychlorinated Dibenzofurans; Quantitative Structure Activity Relationships *Chemosphere*, Vol.14, pp.675-683