PCDDS AND PCDFS CONTAMINATION IN THE NORTHERN PACIFIC AREA REFLECTED ON SQUID LIVER TISSUES

Shunji Hashimoto*, Yasuyuki Shibata*, Hiroyuki Tanaka†, Akihiko Yatsu§ and Masatoshi Morita*

*National Institute for Environmental Studies
16-2 Onogawa, Tsukuba, Ibaraki 305-0053, JAPAN
†National Research Institute of Far Seas Fisheries
5-7-1 Orido, Shimizu, Shizuoka 424-0902, JAPAN
§National Research Institute of Fisheries Science
2-12-4 Hukuura, Kanazawa-ku, Yokohama, Kanagawa 236-0004, JAPAN,

Introduction

Spreading environmental pollution by dioxins in the global scale has been at rated concern. In fact, those compounds have been detected from marine mammals in the North Pacific and the Arctic region (1-3). Understanding the distribution and the behavior of them in the ocean environment is apparently important for human and the members of the ecosystem if we consider their toxicity and persistency.

The use of bio-indicator is valuable because the concentration of PCDDs and PCDFs is so low that it is not easily determined in seawater. Squid can be such type of bio-indicator because its liver contains a lot of lipid, which PCDDs and PCDFs can be accumulated in. In particular, a family of neon flying squid, *Ommastrephidae*, inhabits worldwide in the sea and accumulates pollutants at a high rate within one or two years due to the habit as a carnivore and the short life span less than one or two years.

In this study, squid liver was examined as the monitoring index of dioxins. It was also investigated the distribution of dioxins in the ocean environment and considered the characteristic of pollution at each area by the isomer composition in samples.

Materials and Methods

Squid samples were collected by fishery vessels operating from the Pacific Ocean and the other sea during 1995 to 1998. Samples were immediately iced after catching and carried to our laboratory. 1-17 livers of squids on the same sampling site were combined and homogenized as the representative sample. They were packed into a glass container and stored at -20 °C until analysis.

After the sample defrosted, about 2 g of sample was spiked with ^{13}C labeled standard mixture (5 pg of tetra, penta, hexa, and hepta chlorinated isomer, 10pg of octachlorinated isomer). The sample was extracted by an extractor, ASE-200 (Dionex, US). The conditions were as follows; solvent: acetone/hexane (50/50 v/v), temperature: 150 °C, pressure: 2,000 psi, static time: 5 min, flush volume: 40 %, purge time: 60 sec, cycle: 3 times.

The extract was purified with sulfuric acid shaking and coupling column silica gel on to activated carbon.

The GC/MS analysis was performed on a JMS-700 high performance double focusing mass spectrometer (JEOL, Japan) coupled to a HP 6890 gas chromatograph (Hewlett Packard, US).

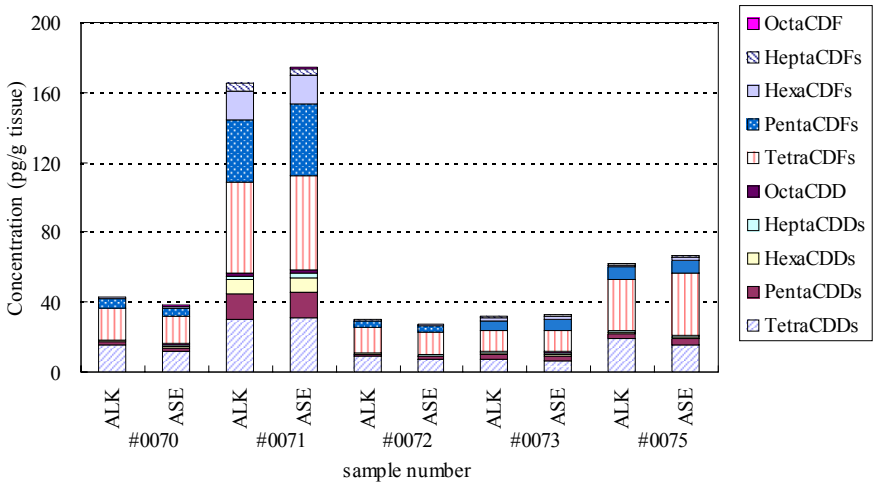

Figure 1 Comparison of the result in the different extraction method. Samples were extracted by alkaline digestion (ALK) and acute solvent extraction (ASE).

The sample solution was introduced into the GC/MS by a FOCUS and an OPTIC2 large volume sample injecting system (Atas-USA, US) equipped an At-Column (GL Science, Japan).
The mass profiles of the selected ions were obtained during GC elution. Identification was based on examination of isotopic ratio of the selected ions and retention time of the GC separation. The area of the mass profile peaks of the quantification ions was used for the quantitative analysis. Quantified values were calculated by the internal standard methods for PCDDs and PCDFs.
Data were processed by Excel 97 for Windows (Microsoft, US), and statistic analysis was performed by STATISTICA 5.1 for Windows (StatSoft, Inc., US).

Results and Discussion

The extraction efficacy of the acute solvent extraction (ASE) was confirmed by comparing with the method using a mild alkaline digestion (ALK) (1). The concentrations of PCDD and PCDF homologues by two methods are shown in figure 1. The concentration ratios of ASE to ALK were calculated on 43 major isomers. The results (average: 1.02, SD: 0.181, n=215) of calculation showed the good coincidence between the methods. Therefore, it was concluded that ASE was effective for the analysis of dioxins in the squid liver.

PCDDs and PCDFs were detected from all squid liver samples at the range of 6.6-59 pg/g and 4.9-120 pg/g on tissue weight basis, respectively, as shown in figure 2. Relatively high concentrations of dioxins were detected in the sea off Japan, while the lowest level of dioxins was found in the sample from the offshore of New Zealand. The regional difference may reflect the industrial activity in these areas.

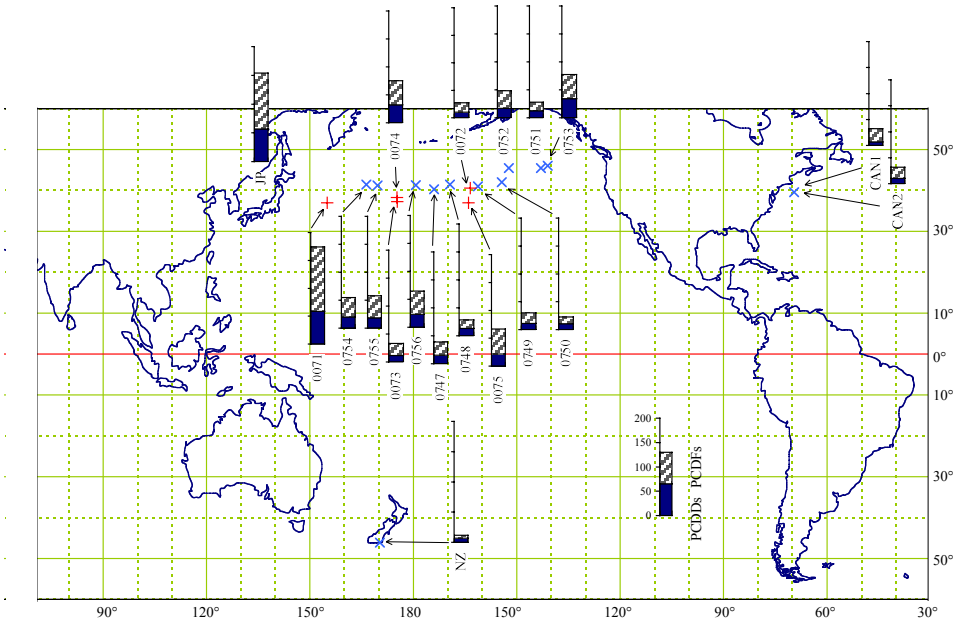

Figure 2 PCDD and PCDF levels (pg /g tissue) in the squid liver samples from the Pacific Ocean and offshore of eastern coast of Canada.

The principal component analysis (PCA) was carried out based on isomer composition using 104 peaks identified on SIM chromatograms. The result was plotted on figure 3. The sum of the first (86.9%) and the second (7.60%) principal component was 94.5%. 1368-TCDD and some TCDFs including 2,3,7,8-TCDF, 1,3,4,6/1,2,4,8-TCDF, 3,4,6,7-TCDF and 2,4,6,8/1,2,3,8/1,4,6,7/1,2,3,6-TCDF, marked at high score on the first and the second principal component, respectively. In figure 3, the samples can be classified into 3 groups. There is a possibility that each of those areas was polluted by dioxins from different sources.

Atmosphere is known as a major pathway of long-range transportation of pollutants such as PCBs, DDT and HCH. PCDDs and PCDFs have been detected from the remote North Atlantic atmosphere (4). It may be reasonable to assume that those compounds detected in the squid liver samples have been transported from land. In the case of the samples in the northern North Pacific Ocean, the origin of those compounds may be in the East and West Pacific rim countries. The homologue compositions in the squid liver samples are different from incineration samples, which may be caused by selective uptake (or clearance), water solubility, and/or the loss of higher chlorinated compounds in air during long-range transportation. It is known that higher chlorinated compounds associated with particulate have short atmospheric lifetime when dioxins emitted into the air from incinerators (5,6).

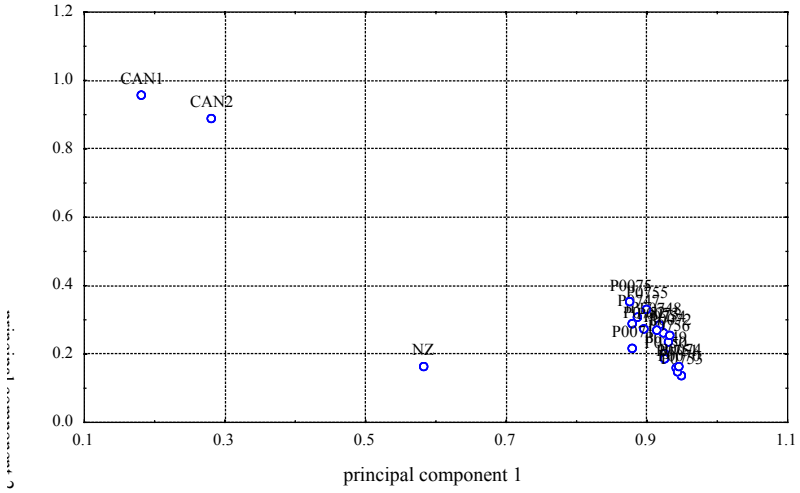

Figure 3 Standardized varimax-rotated factor loading plot for PCA based on the PCDD and PCDF isomer composition of the squid liver samples.

Squid distributes widely in the ocean and accumulates PCDDs and PCDFs into detectable concentrations. Although biological meaning of this accumulation is not clear, it is true that this animal is one of the most suitable bio-indicator for the monitoring of dioxin and related compounds in the marine environment.

Acknowledgement
We thank all of fisheries concerned to cooperate. And we thank to the Center for Global Environmental Research (CGER, Japan) for promoting this study.

References

1. Ono, M., Kannan, N., Wakimoto, T. and Tatsukawa, R.; *Marine Pollution Bulletin*, **1987**, 18, 640
2. Oehme, M., Fürst, P., Krüger, Chr., Meemken, H.A. and Groebel, W.; *Chemosphere*, **1988**, 17, 1291
3. Buckland, S.J., Hannah, D.J., Taucher, J.A., Slooten, E. and Dawson, S.; *Chemosphere*, **1990**, 20, 1035
4. Baker, J.I. and Hites, R.A.; *Environ. Sci. Technol.*, **1999**, 33, 14
5. Eitzer, E.T. and Hites, R.A.; *Environ. Sci. Technol.*, **1989**, 23, 1396
6. Jaarsveld, J.A. and Schutter, M.A.; *Chemosphere*, **1993**, 27, 131