BIOACCUMULATION OF PCDD/DFS AND DIOXIN-LIKE PCBS IN THE SOIL FOOD WEB OF FALLOW RICE FIELDS IN JAPAN

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Abstract

The objective of this study is to investigate the concentration of PCDD/DFs and dioxin-like PCBs in a fallow soil ecosystem and to reveal its bioaccumulation mechanism. Soil, earthworm and lesser Japanese mole (*Mogera imaizumi*) samples were collected in the two fallow rice fields in Kanagawa Prefecture, Japan. The TEQ concentrations of dioxins in soils (organic carbon base) and earthworms (fat base) were same level, but those in mole livers (fat base) were much higher than those in earthworms. The congener profiles of soil and earthworm are quite similar. However, there were major differences in the congener profiles between mole's muscle and liver, and earthworms. This might have been a result of mole's characteristic accumulation of dioxins in liver and also congener specific metabolism of dioxins in mole body. This study showed that dioxins do not accumulate from soil to earthworm but do accumulate from earthworms to mole liver, implying that this might be one of the major exposure routes to their predators.

Introduction

The "SATOYAMA", the characteristic rural landscape in Japan, is the valuable habitat for wildlife including rare species such as raptorial birds. Fallow rice fields are commonly found in these places due to the Japanese government policy of reducing rice production and provide precious feeding habitats for wildlife. Due to the widespread past use of agrichemicals (mostly chlornitrofen (CNP) and pentachlorophenol (PCP)) containing dioxins as impurities, rice paddy soil in Japan is reported to be contaminated with relatively high residue of dioxins [1]. Therefore, fallow rice fields can be a potential source of dioxin exposure for wildlife. There have been scarce studies, however, on these fallows. In this study, region-specific dioxin bioaccumulation through soil-earthworm-mole food chain in fallow rice field was investigated.

Materials and Methods

Sample Collection:

Soil, earthworm, and mole samples were collected from two fallows (Fallow A and B) in Kanagawa Prefecture, Japan. Five squares were randomly set in each 10 m ×10 m fallow. Each square was 50 cm ×50 cm size with depth of 20 cm. Soil and earthworm (Megascolecidae and Lumbricidae) samples were collected from each square and pooled as one sample. The lesser Japanese moles (*Mogera imaizumi*) (n=2) were collected by trap from the fallows. Earthworm samples were taken by digging and hand-sorting, starved on the wet filter paper for three days. All samples were kept -30˚C until analysis.

Analysis:

Whole body of earthworm and mole's liver and muscle of shoulder were used for analysis. All samples were freeze-dried prior to analysis. Details of the analytical procedures were based on the official method established by Ministry of the Environment (MOE) of Japan [2]. The ^{13}C-labeled internal standards were added to all samples and then extracted in a Soxhlet apparatus with distilled toluene for 16 hours. The extracts of earthworm and mole samples were concentrated to 10 ml. Then, 1 ml of the concentrate was used for measuring the lipid contents by gravimetric method. Multilayered silica gel chromatographic column and activated carbon column were used for cleanup procedure. Finally, ^{13}C-labeled recovery standard was spiked for HRGC/HRMS analysis. Tetra to octa-chlorinated PCDD/DFs and dioxin-like PCBs were quantified with DB-5 column (J&W Scientific) [3] and SP2331 column (Supelco) [4].

Statistical analysis:

Cluster analysis of congener profiles was executed with STATISTICA Version 6 (StatSoft, Inc., 2003).

Results and Discussion

The Observed Concentration:

The total concentrations of PCDD/DFs (tetra to octa-chlorinated congeners) and dioxin-like PCBs in soils from Fallow A and B were 44,000 (n=1) and 18,000 (n=1) pg/g, on dry weight basis, respectively. The concentration in earthworms from Fallow A and B were 900,000 (n=4), and 150,000 (n=1) pg/g, on fat weight basis, respectively. For mole samples, the liver samples for Fallow A, and B were 740,000 (n=1), and 470,000 (n=1) pg/g, and the muscle samples were 21,000 (n=1), and 30,000 (n=1) pg/g, on fat weight basis, respectively. In terms of TEQ concentration, soils were 57 and 16 pg-TEQ/g-dry; earthworms, 670 and 150; mole livers, 2500 and 6500; mole muscles, 89 and 150 pg-TEQ/g-fat for Fallow A and B, respectively.

The observed soil concentrations in this study were in the same level as those in rice paddy soil taken in 2002 in Japan as reported by MOE and MAFF (Ministry of Agriculture, Forestry and Fisheries of Japan), which ranged 0.68~170 pg-TEQ/g-dry [5]. The congener profiles for Fallow A and B indicated that chloronitrofen (CNP) was sprayed in the past. Kunisue et al. (2006) [6] reported that the concentrations for carcasses of lesser Japanese moles (n=5) were 27~190 pg-TEQ/g-fat. They were in the same levels observed for mole muscles in this study. The reported concentrations of other species of terrestrial animals were 820, 280, and 5.8 pg-TEQ/g-fat, for large Japanese field mouse (*Apodemus speciosus*) [7], Japanese raccoon [8], and wild boar [9], respectively. The observed concentrations of biota samples in this study were not elevated compared to other reports. The concentrations in earthworms, however, were relatively high.

Congener specific accumulation:

—The comparison of congener profiles

In order to compare the similarity of congener profiles in each samples, profile data set were implemented to cluster analysis. As a result, the congener profiles of the soils and the earthworms were very similar; however the moles were different (**Figure 1**). These results suggested that the congener profiles of the earthworms reflected the profiles of the habitat soil. The congener profile of moles, however, did not reflect those in earthworm, their prey, probably due to the metabolism of dioxins in the body.

—The accumulation characteristic of earthworm

Regression analyses showed significant relationship between log C_{soil} and log $C_{earthworm}$ (**Figure 2**). The high residue congeners in soil were found in high concentrations in earthworm. So earthworm reflected the concentration of habitat soil.

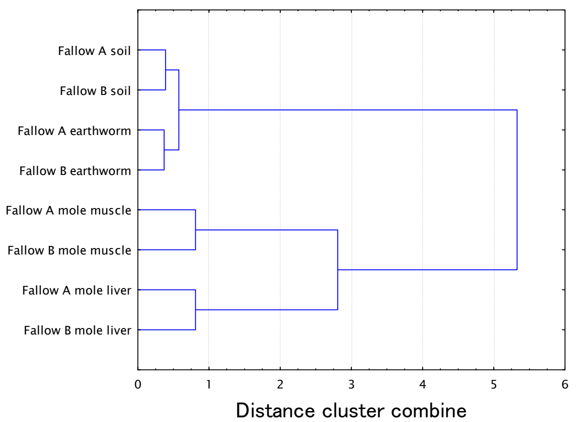

Figure 1 Dendrogram of cluster analysis based on congener profiles of PCDD/DFs and dioxin-like PCBs

(Congener profile is based on the proportion of a congener within each homologue for PCDD/DFs)

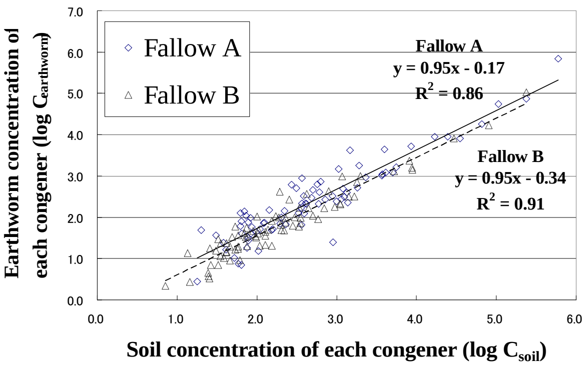

Figure 2 Relationship between the congener concentrations in soil and those in earthworm

—The accumulation characteristic of mole's tissue

The accumulation characteristic of mole's tissue differed from those of earthworm. This difference suggested that metabolism of dioxins in mole's body was significant. The ratio of 2,3,7,8-substituted PCDD/DFs and dioxin like-PCBs congeners in liver/muscle are shown in **Figure 3**. The ratio of liver and muscle concentrations tended to increase with the degree of chlorination of PCDD/DFs. For dioxin-like PCBs, the ratio for PCB #126 was dominant. These results suggested that the similar mechanism occurs in moles and raccoon dogs (Kunisue et al., 2006) [6].

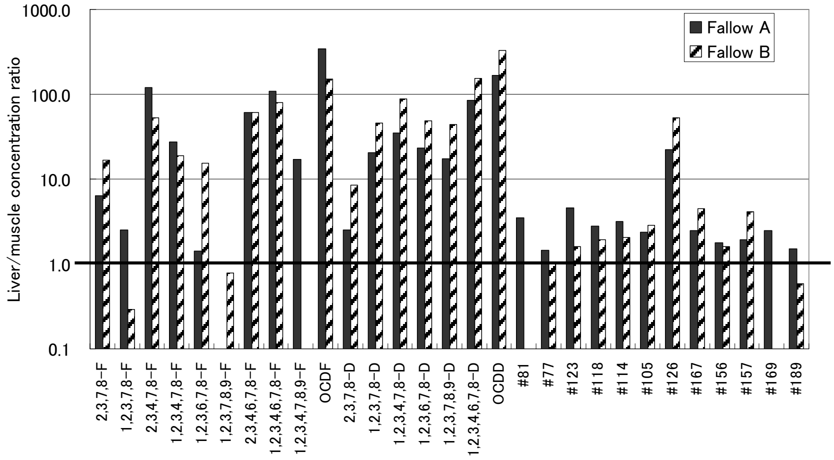

Figure 3 The liver/muscle concentration ratio for lesser Japanese mole

Bioaccumulation factor:

—**Bioaccumulation factor in TEQ**

Bioaccumulation factor was calculated from the observed concentrations. Biota-soil accumulation factors (BSAF) were obtained by dividing the concentrations in earthworms (pg-TEQ/g-fat) with those in soils (pg-TEQ/g-organic carbon) for each habitat. Biomagnification factors (BMF) were obtained by dividing concentrations in mole's tissues (pg-TEQ/g-fat) with those in earthworms (pg-TEQ/g-fat). As a result, BSAF for earthworms were 0.46 for Fallow A and 0.55 for Fallow B. On the other hands, BMF for mole liver were 3.8 for Fallow A and 42 for Fallow B. The BMF for mole muscle were 0.13 for Fallow A, and 0.94 for Fallow B. In this study, high bioaccumulation was found only for earthworm-mole liver relationship.

—**Bioaccumulation characteristic of 2,3,7,8-substituted PCDD/DF and dioxin-like PCB congeners**

The BSAF and BMF of each 2,3,7,8-substituted PCDD/DF and dioxin-like PCB congener were shown in **Figures 4 and 5**. For both fallows, 1,2,3,4,7,8-HxCDD had the highest BMF among PCDD/DFs in mole. In addition, BMF tended to increase with the degree of chlorination of 2,3,7,8-PCDD/DFs. When congener profile of BSAF and BMF (liver and muscle) are compared, resemblances are noticed between Fallow A and Fallow B. This may be due to the similar soil dioxin congener profiles in both sites and/or to the specific metabolism in moles.

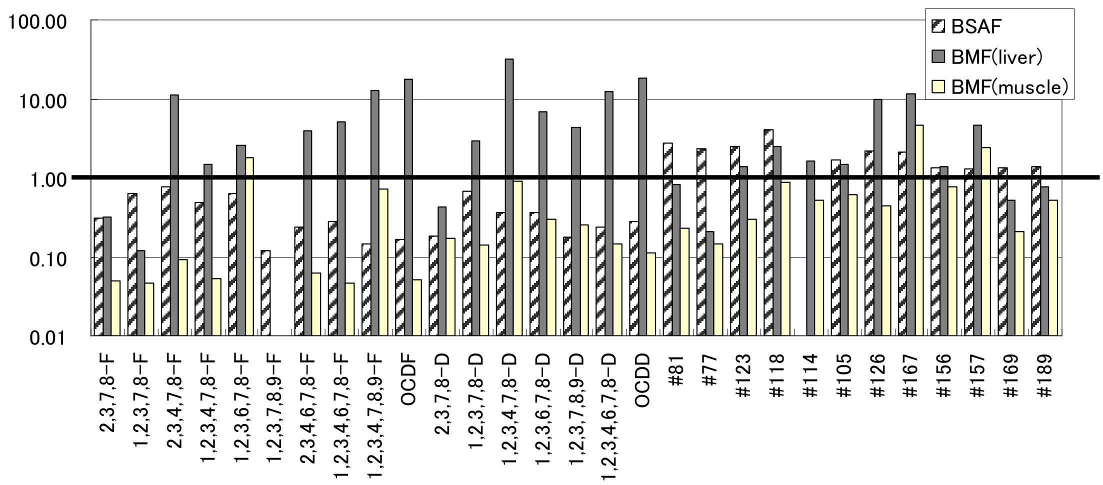

Figure 4 BSAF (earthworm/soil) and BMF (mole/earthworm) in Fallow A

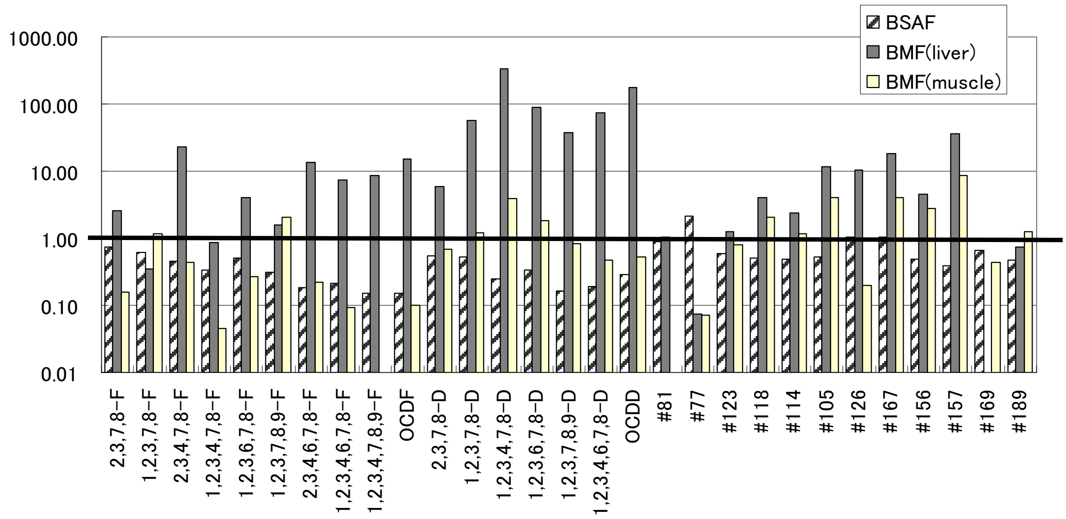

Figure 5 BSAF (earthworm/soil) and BMF (mole/earthworm) in Fallow B

Conclusions

The relatively high concentrations of dioxins were found for earthworms, moles from fallow rice fields. While the profiles of PCDD/DF and dioxin-like PCB congeners in earthworms were quite similar to those in the soils, there were major differences in the congener profiles between mole's muscle and liver, and earthworms. This might have been a result of mole's characteristic accumulation of dioxins in liver and also congener specific metabolism of dioxins in mole body. The results also suggested that dioxins do not accumulate from soil to earthworm, however do accumulate from earthworms to mole liver. Therefore, in fallow rice field, possible major dioxin and dioxin-like PCB transfer pathway is through liver of mole to their predators. It should be noted that the slightly high concentrations of these chemicals are found in earthworms.

Acknowledgements

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