# DIOXINS CONTAMINATION IN LEAFY VEGETABLES BASED ON THE SURFACE STUDY

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## Introduction

Human background exposure to PCDD/Fs and Co-PCBs through the vegetables consumption has been considered as negligible since the contamination levels in vegetables crops were found very low levels in Japan. Various studies in the levels on dairy products can be found in the literature, but instead of its widespread consumption, so far no much information has been published concerning the levels and the contamination mechanism of these pollutants in vegetables crops. Thereby, there has been an increasing demand for the systematical studies of contamination mechanism in vegetables.

According to McCrady et al.[1] and McLachlan[2]; they focused on the pathways of PCDD/Fs which is related to plant uptake: (1) root uptake and transport to the shoot, (2) contamination of shoots by soil particles and atmospheric deposition, (3) uptake of vapor phase by aerial plant parts. Since then, we found no reports concerning the detailed investigation of contamination pathways of PCDD/Fs and Co-PCBs in vegetables crops.

In this study, the effect of PCDD/Fs and Co-PCBs in leafy vegetables is investigated in the view of contamination pathway by which PCDD/Fs and Co-PCBs transport/adhesion to leafy vegetables.

## Methods and Materials

*Standards and reagents*

All PCDD/Fs and Co-PCBs used as internal, recovery and calibration standards were purchased from Wellington Laboratories (Guelph, Ontario, Canada). All solvents used were for organic trace analysis and were obtained from Kanto Kagaku (Tokyo, Japan).

*Sample cultivations*

Two types of leafy vegetables were selected for this study. The spinach and egoma (*perilla*) plants represented an edible leaf. Due to differences in growth rates, each species was planted, grown, and harvested independently of the others. The leafy vegetables were sown and cultured in open field at the experimental site of National Institute for Agro-Environmental Sciences.

*Analytical procedures of leafy vegetable samples*

The slightly modified method of analysis of foods in Japan (the Ministry of Health and Welfare, Japan) was followed for leafy vegetable sample preparation procedures, analytical techniques and quality control strategies. The vegetable samples were carefully washed with running tap water. All samples were mixed in a food processor (stems of spinach were included). Homogenized samples were extracted three times using a mixture of hexane/acetone (1:1). After treatment with concentrated sulfuric acid, the crude extracts were cleaned up using a multiplayer silica column (silver nitrate/acid/ base-modified silica gel) and activated carbon column chromatography. All samples were spiked with 17 kinds of $^{13}C$-labelled analogues of 2,3,7,8-Cl-substituted congeners and 12 kinds of $^{13}C$-labelled mono/non-ortho-PCBs (IUPAC no. #123, 118, 114, 105, 167, 156, 157, 189 and #81, 77, 126, 169) before extraction.

*Determination of PCDD/Fs and Co-PCBs*

Detection of PCDD/Fs and Co-PCBs has been described previously [3].

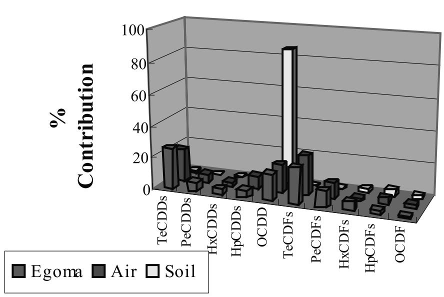
*Scanning Electron Microscopy (SEM)*

SEM (JEOL, JSM-5610LV) equipped with energy dispersive X-ray analysis (EDS) (JEOL, JED-2201) system was used to study the microstructure and in situ chemical composition of individual site on leafy vegetables surface. Qualitative analysis of the elements presented in the samples was acquired in the same operating conditions.

## Results and Discussion

In order to be able to compare and contrast the influence of cultural environment, PCDD/Fs and Co-PCBs concentrations of atmosphere and soil in the area of cultivation were measured. The percentage contribution of dioxins in the tested soils was found to be about 85 % of OCDD (total 11.06 pg-TEQ/g dry). In the case of the atmosphere, PCDD/Fs and Co-PCBs concentrations of the area assessed were 5.05 pg/m$^3$ (0.015 pg-TEQ/m$^3$).

The observed PCDD/Fs contribution patterns in spinach and egoma (*perilla*) were very similar with the patterns in atmosphere in the area of cultivation (Fig.1). This result was mainly attributed to contamination of shoots by atmospheric deposition. Total PCDD/Fs contributions were good agreement with those of Eun et al.[3].



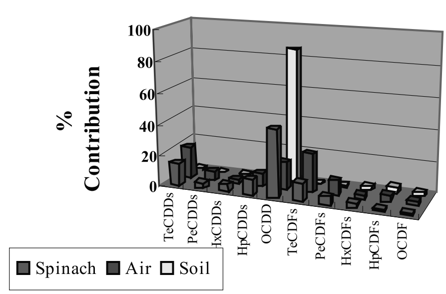




**Figure 1.** Patterns of the PCDD/Fs in egoma, spinach and their cultural environment

As in the OCDD, the percentage contributions of spinach and egoma were found to be 43 and 17 %, respectively. To explain this difference, it is necessary to investigate the trace for their surface conditions. Figure 2 shows typical SEM pictures of spinach and egoma surface. They were seen consistently for different regions in several of the scans. It can be seen that Fig. 2 (a) shows the micrograph of adhesion particles on spinach surface although the spinach samples were carefully washed with running water. This result is related to the strongly attached soil particles on spinach surface.

Evidence of attached soil particles on spinach surface was also supported by the EDS results (Fig. 3). From the element analysis results on spinach surface, strong Al and Si Ka peaks were detected. These elements were found to be the major components of cultivated soil. On the other hand, egoma (Fig. 2 (b)) did not seriously contaminate by soil particles.

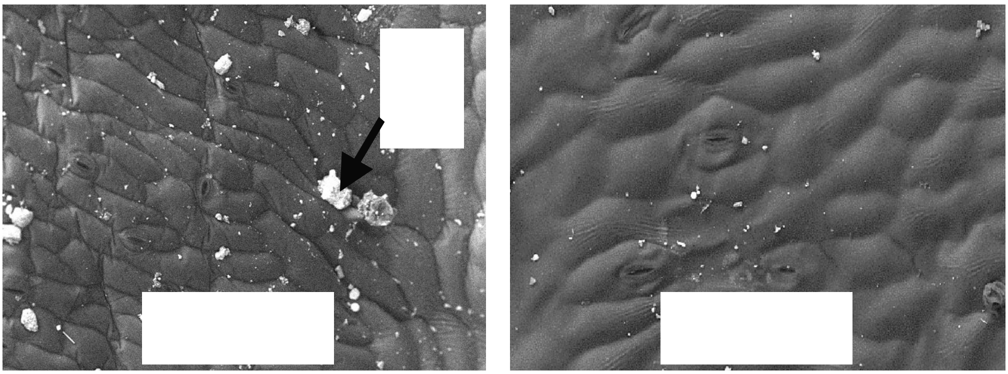


**Figure 2.** SEM micrographs (300X) of spinach and egoma (perilla) leaf. The SEM/EDS analysis for a showed in Fig. 3.

These differences could basically be explained as the top length of plant. The top length of spinach and egoma plant was about 30 and 100 cm, respectively. It is apparent that spinach is easier influenced by soil particles than that of egoma. Our findings suggest that the top length of the plant and the sampling position significantly influences the analysis. This study provides thus important information for the assessment of public health risks.

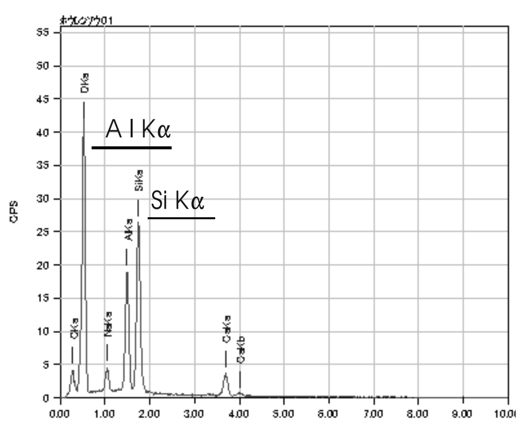



**Figure 3.** EDS element analysis for the point, a in Fig. 2.

**References**

1. McCrady, J.K., McFarlane C., Gander, L.K. (1990) Chemosphere 21, 359
2. McLachlan, M.S. (1997) Chemosphere 34, 1263
3. Eun H., Ishii Y., Kobara Y., Ishihara S., Uegaki R., Seike N., Kuwahara M., Nakamura K., Narita I and Ueji M (2001) Organohalogen Compounds 51, 321