Introduction

Cadmium (Cd) is one of the most widespread hazardous metals found in agricultural soils. In a market-basket study conducted in Japan, 7% of 381 eggplant samples were found to exceed the maximum Cd concentration (0.05 mg kg^{-1}) for fruiting vegetables recommended by codex Alimentarius Commissions. These results indicate the urgent requirement for cultivation systems that reduce the Cd concentration in eggplant. Grafting eggplant *Solanum melongena* onto *Solanum torvum* has been reported to be an effective method for reducing the Cd concentration in the fruits of eggplant by up to one-fourth[1)]. The Cd concentration in the shoot and xylem sap of *S. melongena* was higher than that of *S. torvum*; however, the concentration in the roots of both species was almost identical[2)]. It was likely that the limited translocation of Cd into the shoot of *S. torvum* was partly due to the presence of a barrier function in root tissues. In this study, we compared the microscale distribution pattern of Cd in root tissues of *S. melongena* and *S. torvum* by using the synchrotron micro-X-ray fluorescence (SR μ-XRF) technique and thus elucidated the role of the roots of *S. torvum* in restricting Cd transport to the shoot.

Materials and Methods

Hydroponically cultivated *S. melongena* cv. Senryo 2 and *S. torvum* cv. Torubamubiga were exposed to $CdCl_2$ (17 mmol L^{-1}) for 24 h with fresh nutrient solution at pH 5.5 in a controlled environment chamber. From 5-mm root segments obtained at a distance of 10 and 40 mm from the root apex, 200-μm thick transverse sections were prepared using a microtome. The freshly prepared sections were frozen immediately by contacting them on an aluminum block with a mirrored surface that was pre-cooled by liquid nitrogen; the sections were subsequently freeze-dried. The freeze-dried sections were then mounted on an acrylic plate for SR μ-XRF analysis. A few of the sections were stained with berberine hemisulfate/aniline blue[3)] to identify the presence of Casparian bands by fluorescence microscopy. The distribution of Cd, Zn, and Fe in the transverse sections of the root was mapped using SR μ-XRF at BL37XU, SPring-8, Japan Synchrotron Radiation Research Institute. The energy of the incident X-ray beam was adjusted at 30 keV to excite the K-edge of Cd. The beam spot size was 2.0 μm wide and 1.0 μm in height, and the scanning step size was 3 μm. The spectral acquisition time was set to 2 s.

Results and Discussion

Casparian bands were present both in the exodermis and endodermis at a distance of 40 mm from the root apex, but not in those at a distance of 10 mm from the root apex. Cd was localized both in the central cylinder as well as the epidermis in the transverse sections of *S. melongena,* regardless of the distance from the root apex. In the sections of *S. torvum* taken at 10 mm from the root apex, the intensity of X-ray fluorescence derived from Cd Kα was very weak, and Cd was evenly distributed. However, Cd was significantly localized in the

endodermis at 40 mm from the root apex. Those distribution pattern of Cd in root suggested that more Cd was transported to central cylinder for S. *melongena* than for S. *torvum*, in agreement with the higher Cd concentration in xylem sap for S. *molongena*..

Radial transport of Cd toward the central cylinder in the root of *S. torvum* was restricted by an endodermal apoplastic barrier—a Casparian band or suberin lamella. This barrier function in the roots of *S. torvum* is one of the mechanisms responsible for low Cd loading to the xylem compared with *S. melongena.*

Acknowledgement

This work was financially supported by the Program for Promotion of Basic Research Activities for Innovative Biosciences (PROBRAIN) and was carried out under the approval of the SPring-8 Program Advisory Committee (2008A1087).

References

1) Arao, T., Takeda H. and Nishihara E.: Reduction of cadmium translocation from roots to shoots in Eggplant (*Solanum melongena*) by grafting onto *Solanum torvum* rootstock, Soil Sci. Plant Nutr. 2008;54:555-559.
2) Mori, S., Uraguchi, S., Ishikawa, S., Arao, T. Difference of affinity for Cd in xylem loading is a critical factor for determining the Cd concentration in shoot of egg plant (*Solanum melongena*) and a related plant (*Solanum torvum*) in low Cd concentration medium. Env. Exp. Bot. 2009;*in review*
3) Brundrett MC, Enstone DE, Peterson CA. A berberine-aniline blue fluorescent staining procedure for suberin, lignin, and callose in plant-tissue. Protoplasma 1988;146:133–142.