

# Differences of cadmium uptake and accumulation among soybean (*Glycine max*) cultivars

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## Abstract

Soybean (*Glycine max*) was cultivated in the field with high Cd concentration of FM Honmachi, Tokyo University of Agriculture and Technology, to investigate the differences in the amount of Cd uptake and accumulation among soybean cultivars. Three cultivars of soybean; Enrei, Tsurunoko and Tsukui were used in this study. The order of Cd concentration in green beans (pea in a pod, usually served after boiling as it is) and soybean seeds was Enrei < Tsurunoko < Tsukui with the mean value of 0.21, 0.27 and 0.42 mg/kg DW for green beans and 0.27, 0.34 and 0.51 mg/kg DW for soybean seeds. The relationship between Cd concentration in soil and soybean seeds was different among cultivars; Enrei had a negative trend, but Tsukui had a positive trend. The Cd uptake rate by plant was also different among cultivars. Tsukui had a faster Cd uptake rate than the others. Before the green bean stage, Tsukui showed the fastest Cd uptake, of 0.10 Cd µg/d/plant. After green bean stage, Tsurunoko was the fastest with 0.85 Cd µg/d/plant.

## Key Words

Heavy metal, variety, upland, bean.

## Introduction

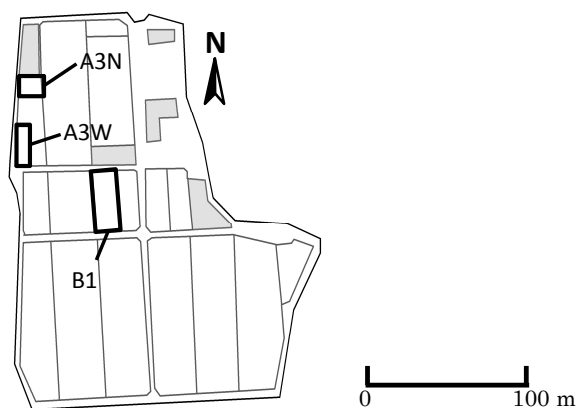
Cadmium (Cd) is a highly toxic element for human beings because of its extremely long biological half-life. Excessive Cd intake causes serious disease of renal and bone tissue in a human body. Therefore, the Codex Committee on Food Additives and Contaminants has announced an upper allowable limit for Cd concentration in staple crops, such as polished rice, wheat and vegetables (Codex Committee 2005). However, Codex Committee decided not to determine the allowable limit of Cd concentration in soybean, because soybean is not a main food product in the world (Codex Committee 2004). Meanwhile in Japan, soybean products are traditional major food like *nattou*, *tofu*, *soysource* and *miso*. The results of a large-scale survey of domestic agricultural products revealed that the Cd concentration of 16.7 % of soybean seeds exceeded intake of 0.2 mg/kg that is the international allowable limit proposed by the Codex Committee until 2004, and is much higher proportion than that of other upland crops (Ministry of Agriculture, Forestry, and Fisheries of Japan 2002). With respect to genetic characteristic for Cd uptake, Arao *et al.* (2002) reported genotypic differences in cadmium uptake and distribution in soybeans cultivated in pot and under low Cd concentrations in the field. Ministry of Agriculture, Forestry, and Fisheries of Japan decided to promote low Cd uptake soybean cultivars and encouraged farmers to cultivate them in high Cd concentration fields (2007). Sugiyama *et al.* (2007) found the cultivars with low Cd uptake accumulated much higher Cd in their roots than those of the cultivar with high Cd uptake. Using tracer Cd, Kawasaki and Oda (2004) and Kawasaki *et al.* (2005) reported that Cd transported to seeds was absorbed before full seed stage and Cd absorbed at the beginning of growing stage was accumulated in leaf. Oda *et al.* (2004) showed the growing stage where Cd concentration in seeds become the highest was from full pod to full seed stage. Decreasing soil Cd concentration reduced Cd concentration in soybean seeds (Maejima *et al.* 2007). In this study, the amount of Cd uptake and accumulation in green beans and soybean seeds among different cultivars and differences of Cd uptake rate among cultivars were investigated.

## Methods

### *Cultivation of soybean*

The cultivation experiment on soybeans was conducted at Honmachi Farm shown in Figure1 (A3N, A3w and B1), Field Science Center for Education and Research of Tokyo University of Agriculture and Technology on an alluvial lowland derived from Tama River sediment (Figure 1). The mean values of soil Cd concentration at A3N, A3W and B1 field (Gray Lowland Soils is comparable to an Aquic Fluvents (Soil Survey Staff 1990) were 1.05, 1.31 and 1.00 mg/kg, respectively. Three different soybean cultivars, Enrei,

Tsurunoko and Tsukui, were selected from many varieties in Japan. Enrei was regarded as a low Cd uptake cultivar and recommended by Ministry of Agriculture, Forestry, and Fisheries of Japan, 2007. Soybean was seeded in May-July of 2008 and harvested in October-November of 2008.



**Figure 1. The Honmachi Farm, Field Science Center for Education and Research of Tokyo University of Agriculture and Technology.**

#### *Soil analysis*

Soil samples ( $n=36$ ) at the point of soybean cultivation field were collected from just below the harvested soybean. After air-drying, soil samples were passed through a 2 mm nylon sieve. Twenty five mL of 0.1 mol/L HCl solution was added to 5 g soil samples. The mixture was shaken by a mechanical end-over-end shaker (Daiki, DIK 2102) at 25 °C. The extract was filtered with a No. 5 C filter paper (Advantec) and Cd concentration in the filtrate was determined by a flame atomic absorption spectrophotometer (Hitachi Z-5010).

#### *Plant analysis*

Green beans and soybean seeds were collected as whole body including root, which was removed from the stem. Soybean seeds were washed with tap water and then deionized water, dried at 70 °C for 36 hours in a ventilated oven and ground with a mixer mill (MM301, Restch) for subsequent Cd analysis. The ground samples were digested using extra-pure water, H<sub>2</sub>O<sub>2</sub> and HNO<sub>3</sub> (1:1:8 mL) in teflon containers in a microwave apparatus. The solution was filtered with a 0.45 µm Membrane filter (Millipore). The Cd concentration in the digests was determined using Inductively Coupled Plasma Mass Spectrometry (Thermo Fisher, X series II).

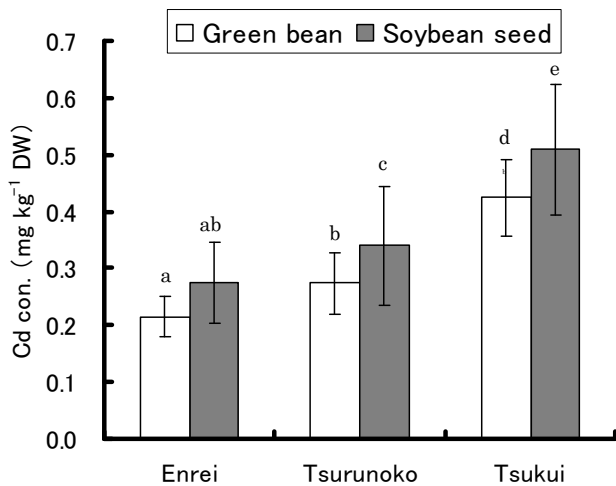
#### *Statistical analysis*

All statistical analyses were performed using JMP 8 (SAS Institute Inc., Cary, NC, USA). Tukey's HSD test was used to determine the differences of Cd concentration in seeds among cultivars.

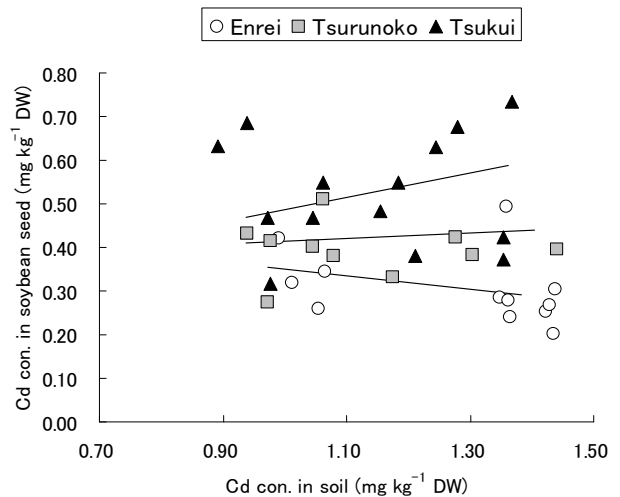
### **Results**

The mean value of Cd concentration in green beans and soybean seeds among cultivars are shown in Figure 2. The mean value of Cd concentration in green bean seeds, harvested after about 87~107 days from seeding, was 0.21, 0.27 and 0.42 mg/kg DW for Enrei, Tsurunoko and Tsukui, respectively. The mean value of Cd concentration in soybean seeds, harvested after about 140 days from seeding, was 0.27, 0.34 and 0.51 mg/kg DW for Enrei, Tsurunoko and Tsukui, respectively. There was a significant difference of Cd concentration in green beans and soybean seeds among cultivars ( $p<0.05$ ). Enrei had the lowest Cd concentration for soybean seeds and Tsukui gave the highest value. Tsurunoko and Tsukui showed significant difference in Cd concentration between green bean and soybean seeds ( $p<0.05$ ).

The relationships between Cd concentration in soil and soybean seeds are shown in Figure 3. This result revealed that the relationship was different among cultivars. When soil Cd concentration increased, Cd concentration in Enrei seeds, which provided low Cd concentration in seeds, slightly decreased but Cd concentration in Tsukui seeds, which exhibited high Cd concentration in seeds, increased. However, these trends are not highly significant.

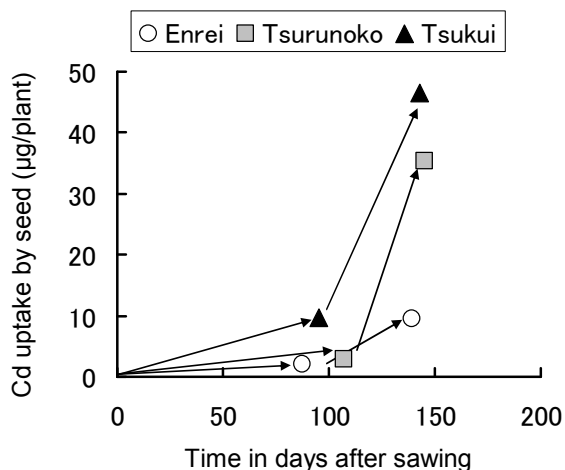


**Figure 2. Cd concentration in green bean and soybean seed of three cultivars. Error bar shows standard deviation. Means followed by the same letter are not significantly different at the 5% level (Tukey's HSD).**



**Figure 3. The relationship between Cd concentration in soil and soybean seeds for each cultivar.**

Figure 4 shows Cd uptake rate to soybean seed during cultivation period. This result showed Cd translocation and accumulation in seed from root. From this result, rapid Cd accumulation in seeds occurred when green bean changed to soybean, and the Cd uptake rate was different among cultivars. While, Oda *et al.* (2004) reported that the growing stage, where Cd concentration in seeds cultivated hydroponically the highest, was from full pod stage to full seed stage (peas in pods become bigger). This discrepancy depends on the cultivation methods and period of soybean. Before they had green beans, Tsukui had the fastest Cd uptake rate. After they had green beans, Tsurunoko had the fastest Cd uptake rate. This might be caused by the number of pods (Tsurunoko had the most pods among them: date not shown). In order to survey Cd in soybean seeds translocated from other organs or soil directly, Cd concentration in other organs should be analysed.



**Figure 4. Cd uptake by seeds during the cultivation period.**

## Conclusion

There were significant differences of Cd uptake among soybean cultivars cultivated in the same upland fields. The relationship between soil Cd concentration and soybean Cd concentration was different between cultivars. Cd uptake by soybean seeds happened extensively when green beans changed to soybeans.

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