Cesium uptake and translocation from tea cutting roots (Camellia sinensis L.)

Cesium uptake and translocation from tea cutting roots (*Camellia sinensis* L.)

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Abstract

To estimate the uptake of radiocesium (^{137}Cs) by tea plant roots, 1-year-old rooted tea cuttings (*Camellia sinensis* L. cv. Yabukita) at the time of bud opening were cultivated hydroponically for 27 days in pots containing nutrient solutions with or without $^{137}CsCl$ (600 Bq mL^{-1}). Total ^{137}Cs radioactivity of whole tea plants were 6.1 kBq g^{-1} dry weight. The plant/solution ^{137}Cs transfer factors of different tissues were in the range of 2.6 (in mature leaves) to 28.2 mL g^{-1} dry weight (in roots), which were lower than those reported in wheat and spinach. In total, 69% of ^{137}Cs remained in roots and 31% was transported from roots to shoots. The results indicated that ^{137}Cs was preferentially translocated to new shoots, which are used for manufacturing tea, over mature leaves.

Keywords

Tea plants (*Camellia sinensis* L.); Fukushima Dai-ichi Nuclear Power Plants; Radiocesium; Root uptake

Abbreviations

TFs, transfer factors; FDNPP, Japan's Fukushima Dai-ichi Nuclear Power Plant (FDNPP)

1. Introduction

Radioactive cesium (i.e., ^{134}Cs and ^{137}Cs) released by Tokyo Electric Power Company Holdings's (TEPCO's) Fukushima Dai-ichi Nuclear Power Plant (FDNPP) accident caused by the Great East Japan Earthquake and tsunami of March 11, 2011, has been detected at levels that exceed the provisional permitted value in tea leaves around the east area in Japan. In a report by the Ministry of Health, Labour and Welfare (2011), the first crops of tea harvested in Ibaraki, Chiba, Gunma, Kanagawa and Shizuoka Prefectures after the accident were contaminated with radiocesium at a higher level than the provisional regulation value for raw food materials set by the Food Safety Commission of Japan, that is 500 Bq kg^{-1} (sum of ^{134}Cs and ^{137}Cs activities). Therefore, as the shipment of tea leaves in these areas was stopped and workers suffered significant economic losses. During the 2 years following the TEPCO's FDNPP accident, Hirono and Nonaka (2016) reported a exponential decrease in the concentration of ^{134}Cs and ^{137}Cs in new shoots of tea plants in Shizuoka, Japan, approximately 400 km southwest from the FDNPP. In a previous study, we showed that radiocesium was primarily absorbed from the lower surface of tea leaves through the stomata, and that the greater part was transported to newly emerged tea tissues, especially new shoots, during the new shoot growth period (Ikka et al., 2018). Therefore, new shoots harvested in the first flush of 2011 were indirectly contaminated by FDNPP's radiocesium fallout. However, because tea plants are a perennial crop, there is concern about the long-term effects of indirect contamination from the soil or source organs other than the new shoots that are used to make tea products (Tagami et al., 2020, 2012). To reduce radiocesium amounts in tea plants, pruning was most effective measure (Hirono and Nonaka, 2016).

In the FDNPP accident, the soil of tea fields was contaminated by radionuclides (Takeda et al., 2013). In particular, long-lived ^{137}Cs in the soil will be a source of radiocesium contamination in tea plants for the long term through root uptake. In general, Cs is not an essential element for plant growth and therefore its absorption mechanism and use by plants has not been studied in depth. As Cs is an alkali metal like K, the two have similar chemical properties and K transport systems also function in Cs uptake by roots (Avery et al., 1993, 1991; Zhu and Smolders, 2000). Previous studies showed that ^{137}Cs activities in the primary and secondary roots of tea plants after the Chernobyl Nuclear Power Plant accident in 1986 greatly decreased in 1987, but then gradually increased up to 1993 (Topcuoğlu et al., 1997). Ertel and Ziegler (1991) reported that 20% of the ^{137}Cs translocated into new leaves of larch (*Larix decidua* Mill.) and about 50% of that into sycamore maple (*Acer pseudoplatanus* L.) resulted from root uptake during the 2.5 years after the Chernobyl accident. These reports suggest that tea plants might take up ^{137}Cs from contaminated soils over the long term. However, the rate of ^{137}Cs uptake by tea roots has not previously been estimated.

The ^{137}Cs uptake activities of plants is estimated as the soil-to-plant transfer factor (TF) (Ehlke and Kirchner, 2002). However, in the literature, for a number of long-lived radionuclides, reported soil-to-plant TFs showed variations that exceeded three orders of magnitude (Coughtrey and Thorne, 1983; Frissel, 1992). The soil-to-plant TF is a macroscopic parameter that integrates various soil chemical, soil biological, hydrological, physical, and plant physiological processes, each of which shows its own variability. Therefore, in several plant species, TFs have been estimated in hydroponic culture by determining the solution-to-plant TFs, enabling more precise measurement of the ^{137}Cs uptake activities of plant roots (Smolders et al., 1997; Smolders and Shaw, 1995).

In this study, we examined radiocesium uptake into tea roots by applying ^{137}Cs into hydroponic solution supplied to cultured tea plants, and determined the TFs of ^{137}Cs in several plant parts during the development of new shoots.

2. Materials and methods

2.1. Plant materials and hydroponic experiments

One-year-old rooted tea cuttings (*Camellia sinensis* L. cv. Yabukita) were carefully washed with tap water to remove soil and then transplanted to a/5,000 Wagner pots supplied with continuously aerated hydroponic solution (volume 3 L) in a greenhouse in Shizuoka University (Shizuoka, Shizuoka, Japan). The hydroponic solution was prepared according to Konishi et al., (1985), adjusted to pH 4.2 with 1 M H_2SO_4 and renewed every week until the start of treatment.

In 2014, we conducted the following hydroponic experiment to evaluate ^{137}Cs uptake from tea roots. After 9 weeks, to synchronize the growth stage of tea plants, one bud and three leaves of every new shoot were cut off. After 3 weeks, at the time of bud opening (Supplementary Fig. 1), three tea plants were transplanted into each one new pot containing the hydroponic solutions (volume 3 L) with (+Cs) or without (−Cs) using 3.7 MBq L^{-1} $^{137}CsCl$ solution (Eckert and Ziegler Isotope Products, Valencia, CA, USA). At the start of the treatment, the final ^{137}Cs concentration of the +Cs treatment solution was 600.0 Bq mL^{-1}. Each treatment was carried out with three replicates. During the treatment, tea plants were cultivated in draft chambers of the Radiochemistry Research Institute, Shizuoka University. In the draft chamber, light/dark periods were 12 h/12 h, and light intensity at the canopy was kept at around 200 μmol m^{-2} s^{-1} PPFD using two white LED lamps (Natural Spectrum light, VeriLux Inc., Vermont, USA). The +Cs and −Cs solutions were not changed during treatment, but the solutions were maintained at a volume of 3 L using −Cs hydroponic solution. After 27 d, at the opening of the third to fourth leaves, tea plants were washed with tap water at least three times then wiped dry. Each tea plant (one replicate) was separated into new shoots, mature leaves, upper trunk, lower trunk, and roots. After measuring fresh weights, each part was kept at −30 °C until ^{137}Cs analysis. Representative plants were used to perform ^{137}Cs image plate analysis.

2.2. Measurement of ^{137}Cs radiation

Collected samples were dried for 3 d at 60 °C, then weighed samples were placed in 10-mL polystyrene tubes for radioactivity analysis. The radioactivity of ^{137}Cs was evaluated by an auto-well gamma system (ARC-380CL, Aloka Inc.) calibrated using a standard ^{137}Cs source with each part. Radioactivity values of tea plant samples were obtained after subtraction of the background values from samples without applied ^{137}Cs.

2.3. Calculation of ^{137}Cs transfer factors from solution to tea plants

In the experiment to study ^{137}Cs uptake by tea roots, transfer factors (TFs) of ^{137}Cs from the solution to tea plants (plant/solution ^{137}Cs TF; mL g^{-1} dry weight, DW) were calculated as the ratio of the ^{137}Cs activity concentration in each plant part (Bq g^{-1} DW) to that of the treatment solution (600 Bq mL^{-1}). ^{137}Cs radioactivity (kBq plant^{-1}) of whole plants was calculated by summing the data of each plant part. And, ^{137}Cs radioactivity (kBq g^{-1} DW) of whole plants was calculated from the ^{137}Cs radioactivity (kBq plant^{-1}) and DW.

2.4. Imaging plate analysis

One set of shoots and roots of +Cs treatments was put into a polyethylene film bag, closely set on the imaging plate (BAS-MS2040, FUJIFILM, Tokyo, Japan), and exposed in darkness for 21 d at room temperature. Radioactivity distribution images were obtained by scanning the imaging plates with the molecular measurement program FX Pro Plus (BioRad Laboratories, Inc., California, USA).

2.5. Statistical analyses

Data were statistically analyzed using Tukey's honestly significant difference (HSD) test to determine significant differences among groups. P-values < 0.05 were considered significant. Statistical analyses were performed using R software ver. 4.0.2.

3. Results and Discussion

Each of five parts and total dry weights (DW) at the end of treatment are shown in Table 1. The DW of new shoots was 0.9 g plant^{-1}; two or three new shoots per plant emerged during 27 d of cultivation in the draft chamber with radiocesium application, meaning that it is possible to evaluate the translocation of the radiocesium from tea roots to new shoots.

^{137}Cs radioactivity and TFs from tea roots are shown in Table 1. Total ^{137}Cs radioactivity of whole tea plants was 6.1 kBq g^{-1} DW. Comparing the radioactivity among plant parts, it was highest in roots (16.9 kBq g^{-1} DW) and lowest in mature leaves and upper trunk (1.5 and 2.3 kBq g^{-1} DW, respectively). The radioactivity of new shoots (4.2 kBq g^{-1} DW) was between that of roots and mature leaves. These results were supported by the image plate analysis (Fig. 1). The results indicate that ^{137}Cs was preferentially translocated to new shoots, which are used for manufacturing tea, rather than mature leaves. The radioactivity of the lower trunk (3.2 kBq g^{-1} DW) was tended to be higher than that of the upper trunk because a part of the lower trunk was immersed in the treatment solution (Supplementary Fig. 1). Thus, the ^{137}Cs radioactivity of the lower trunk might include not only the ^{137}Cs transported from roots but also the ^{137}Cs absorbed directly from the treatment solution itself and remaining adventitious roots. This means a limitation of the hydroponic test using clonal tea cuttings, which have adventitious roots from the base of the lower trunk. Based on ^{137}Cs radioactivity of each plant part and the nutrient solution (600.0 Bq mL^{-1}), the plant/solution ^{137}Cs TFs were in the range from 2.6 (in mature leaves) to 28.2 mL g^{-1} (in roots) (Table 1). The ^{137}Cs TF of new shoots (6.9 mL g^{-1}) was around one-third of the roots' value and similar to that of the whole tea plant (10.2 mL g^{-1}). In wheat (*Triticum aestivum* cv. Tonic) cultivated for 21 d in nutrient solution containing 3 mM K^+, 0–4.24 mM NH_4^+ or 4.24–4.98 mM NO_3^-, 0.25–2.49 mM Ca^{2+} and 0.72–1.27 mM Mg^{2+} with 5 Bq mL^{-1} $^{137}Cs^+$, the plant/solution ^{137}Cs TF was 30–60 mL g^{-1} for the shoot and 60–140 mL g^{-1} for the roots (Smolders and Shaw, 1995). In spinach (*Spinacia oleracea* L. cv. Subito), the plant/solution ^{137}Cs TF was 41–117 mL g^{-1}, when cultivated in solution containing 5 Bq mL^{-1} ^{137}Cs with the following range of cationic concentrations: 0.53–10.4 mM K; 0–8.47 mM NH_4; 0.15–5.0 mM Ca; 0.08–2.0 mM Mg (Smolders et al., 1997). It has been shown that the concentrations of the cations, K^+, NH_4^+, Ca^{2+}, and Mg^{2+}, in the culture solution affected the ^{137}Cs uptake of plants (Zhu and Smolders, 2000). The concentrations of 1mM K^+, 1.8 mM NH_4^+, 0.5 mM Ca^{2+}, and 0.4 mM Mg^{2+} in the tea culture solution used in our experiments (from Konishi et al., 1985) were within the range of those in the above-mentioned wheat and spinach culture solutions. These results indicated that the ^{137}Cs uptake activity of tea plant roots from the solution was lower than those of wheat and spinach, although it is necessary to consider the difference in ^{137}Cs concentrations and its ratio to K of the culture solutions and the ^{137}Cs adhesion on surface.

It is known that Cs^+ enters into plant cells through potassium ion (K^+) transporters (Avery et al., 1993, 1991; Sacchi et al., 1997; Sheahan et al., 1993; Zhu and Smolders, 2000), therefore any factor that influenced K^+ uptake might also affect Cs^+ uptake. It was reported that pH affected the activity of K^+ transporters (Maathuis and Sanders, 1996). In *Riccia fluitans* cultivated in culture solution buffered at pH 6.5–9.0, Cs uptake rates under K^+ deficiency showed a maximum at pH 7.5 with declines under more acid or more alkaline conditions, but were not affected by pH under K^+ sufficiency (Heredia et al., 2002). In our experiment, the culture solution was adjusted to an acidic condition (pH 4.2), because the optimum pH for tea plant growth was 4.0–5.0 in the presence of 0.4 mM Al^{3+} (Konishi et al., 1985; Yamashita et al., 2020). Konishi et al. (1985) reported that 1 mM K^+ was adequate for tea growth in solution culture, and K uptake by tea plants was stimulated under this acidic condition in the presence of Al. These results might mean that the cation concentrations under the acidic conditions of our culture solutions-did not bring about the lower Cs^+ uptake activity of tea plants.

Among five parts of tea plants, the ^{137}Cs radioactivity was the highest in roots (31.6 Bq plant^{-1}) followed by new shoots (3.7 kBq plant^{-1}), lower trunk (3.5 kBq plant^{-1}), mature leaves (3.4 kBq $plant^{-1}$), upper trunk (3.1 kBq $plant^{-1}$) (Table 1). Thus, 69% of the total amount of ^{137}Cs per plant remained in roots, and 31% was transported from roots to shoots. In previous reports, tissues that contained low K concentration, such as ears, fruits, or wood, were also found to be low in Cs (Zhu and Smolders, 2000). Konishi et al. (1985) reported that K concentration was the highest in roots (30.6 mg g^{-1} DW), followed by shoot tips (20.3 mg g^{-1} DW), which were similar to mature leaves (18.3 mg g^{-1} DW) > stems (14.8 mg g^{-1} DW) in 1-year-old rooted tea cuttings cultivated in culture solution. Given this report (Konishi et al., 1985) of the K distribution in tea plants, it is possible that the concentration of radiocesium was also high in the roots. However, in our results, the Cs distribution among the shoot tips, mature leaves, and trunk of tea plants did not exactly reflect that of K (Table 1). Topcuoğlu et al. (1997) detected radiocesium only in the roots of tea plants in Turkey 8 years after the Chernobyl NPP accident. This finding suggested that the rate of Cs transport into the xylem in the tea plant roots was low although there is a difference between hydroponics and field cultivation. It was reported that an important property of plants in relation to ^{137}Cs uptake was high growth rate and high biomass production in a given environment (Soudek et al., 2004). Generally, woody plants showed a lower capacity for water uptake and a slower growth rate, compared with annual plants. In tea plants, such properties might contribute to the low ^{137}Cs uptake.–

4. Conclusion

In conclusion, we investigated the uptake of ^{137}Cs radioactivity and its transfer from tea roots to other plant parts in model hydroponic conditions. The plant/solution ^{137}Cs TFs among different tissues of tea plants were in the range 2.6 (in mature leaves) to 28.2 mL g^{-1} (in roots), which were lower than values previously reported in wheat (Smolders and Shaw, 1995) and spinach (Smolders et al., 1997). In total, 69% of ^{137}Cs remained in the roots, and 31% was transported from the roots to shoots. Our results indicated that ^{137}Cs was preferentially translocated to new shoots over mature leaves in tea plants. Our results will contribute to understanding the long-term effects of indirect radiocesium contamination from tea roots.

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Conflict of interests

The authors declare no conflict of interests associated with this manuscript.

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Supplementary information

Supplementary Fig. S1 Tea plant in hydroponic culture at the time of bud opening at the start of treatment

Table legends

Table 1 ^{137}Cs radioactivity and transfer factor from tea roots

Tissues	Dry weight (g plant^{-1})	^{137}Cs radioactivity (kBq g^{-1} DW)	Plant/solution ^{137}Cs transfer factor	^{137}Cs radioactivity (kBq plant^{-1})	Proportion of ^{137}Cs radioactivity (%)
New shoots	0.9 ± 0.5	4.2 ± 0.6 b	6.9 ± 1.1 b	3.7 ± 1.5 a	8.2 ± 3.4 a
Mature leaves	2.2 ± 0.8	1.5 ± 0.4 a	2.6 ± 0.6 a	3.4 ± 1.4 a	7.5 ± 3.1 a
Upper trunk	1.3 ± 0.2	2.3 ± 0.1 ab	3.9 ± 0.2 ab	3.1 ± 0.3 a	6.9 ± 0.7 a
Lower trunk	1.1 ± 0.3	3.2 ± 0.9 ab	5.3 ± 1.5 ab	3.5 ± 1.2 a	7.7 ± 2.5 a
Roots	1.9 ± 0.2	16.9 ± 1.5 c	28.2 ± 2.5 c	31.6 ± 5.5 b	69.6 ± 12.2 b
Whole plants	7.5 ± 1.8	6.1 ± 0.3	10.2 ± 0.5	45.4 ± 8.8	100.0 ± 19.4

Values are mean ± SD (n=3). Different letters indicate significant differences among tissues (Tukey's HSD test, $P < 0.05$).

Figure legends

Fig. 1 Distribution of ^{137}Cs radioactivity after uptake and translocation by tea roots

Imaging plate analysis of a 1-year-old rooted tea cutting grown for 21 d in hydroponic culture with ^{137}CsCl (600 Bq mL^{-1}). Left and right images show the plant appearance and distribution of radioactivity, respectively.