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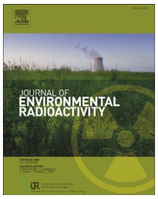

Effects of accompanying anions on cesium retention and translocation via droplets on soybean leaves

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ABSTRACT

Plant foliar uptake and translocation is an important pathway for the migration of radiocesium to the human diet. This study reports the effects of accompanying anions (NO_3^-, SO_4^{2-}, CO_3^{2-}, and I^-) on cesium retention and translocation. An experiment to simulate cesium retention and translocation was conducted in a greenhouse by applying droplets of stable cesium solutions to the upper surface of four soybean [*Glycine max* (L.) Merr.] trifoliate leaves. The average percentages of cesium retention with the accompanying anions NO_3^-, SO_4^{2-}, CO_3^{2-}, and I^- on the leaves were 7.2, 21.5, 49.3, and 10.2%, respectively. Retention values of the four treatments were stable during the 3-day exposure period, indicating that cesium could be absorbed and penetrate the cuticle quickly once it was dissolved. Scanning electron microscopy coupled with energy dispersive X-ray microanalysis showed that particles containing cesium remained on the leaf surfaces after washing. Also, nano-sized particles containing cesium were observed inside the leaf tissues. Cesium concentrations in the uncontaminated leaves, pods, stems, and roots increased during the study period indicating cesium redistribution from the contaminated leaves.

1. Introduction

^{134}Cs and ^{137}Cs are two important fission radioisotopes released from nuclear installations in accidental situations and during the explosion of nuclear weapons. These radioisotopes have attracted significant attention in environmental science because of their relatively high fission yield, long half-life, and high mobility in organisms. Consumption of contaminated food is one cause of internal radiation in humans and grazing animals. Traces of cesium isotopes migrating from the environment to humans have attracted global concern. Cesium radioisotopes are transferred into crops through immediate deposition from the atmosphere and/or absorption from soil through roots. The deposition of radioisotopes on vegetation and soil is the starting point of radioisotope transfer in the terrestrial environment and in food chains. Wet deposition onto plant organs is a potential major source of contamination in food chains (Kinnersley et al., 1997). Thus, understanding the processes that affect the behavior of these radionuclides in agricultural products can improve the assessment of risks in the ingestion of contaminated food by humans and it can encourage policymakers to take appropriate actions to protect the environment and safeguard human health.

Their aquatic ancestors have given terrestrial plants the ability to absorb and retain nutrients through their leaves (Mengel, 2001). Contaminants may also be transferred through the same process. Contaminants may cross several physical barriers such as cuticular membranes and epidermal hairs before entering the cytosol of epidermal cells. This penetration is strongly reliant on plant species, weather conditions, physiological status, and speciation of the element (Kinnersley et al., 1997; Pröhl, 2009; Schönherr and Luber, 2001; Thiessen et al., 1999). According to Chamel et al. (1991), contaminants may be adsorbed in the cuticle, diffuse through the cuticle, be desorbed in the apoplast, and absorbed by the subjacent mesophyll cells. Owing to the dual nature of cuticular constituents (lipids/hydrophilic molecules), the hydrophilic pathway can be accessed by ions and hydrophilic solutes through aqueous pores (Schönherr, 2000), which are located on anticlinal walls and on cuticular ledges of stomata guard cells (Eichert and Burkhardt, 2001; Schönherr, 2006). In addition, cesium is readily absorbed by leaves and redistributed into the whole plant through the xylem and phloem (Henner et al., 2005; Peuke, 2010). Also, compared with other potentially toxic elements deposited on leaves, cesium is classified in the group of mobile elements (Carini et al., 2003; Colle et al., 2009). Tissues that have low potassium concentrations (on a

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dry weight basis), such as ears, fruits, or wood, are also low in cesium. Cesium can be transported efficiently in the phloem. The higher fraction of cesium that is recirculated can be attributed to the selectivity of potassium uptake relative to cesium in leaf cells (Zhu and Smolders, 2000). Therefore, translocation redistributes a chemical substance from the time it is absorbed by the plant by transport to other parts of the plants that have not been contaminated directly. Several studies have revealed that the growth periods of plants influence cesium translocation, with redistribution from contaminated parts reaching a maximum when deposition occurs during the anthesis and fruiting period (Bengtsson et al., 2011; Carini et al., 2003; Colle et al., 2009; Macacini et al., 2002; Madoz-Escande et al., 2009). Many isotopic studies have reported that the distribution of radioactive (^{137}Cs and ^{134}Cs) and stable cesium (^{133}Cs) isotopes in natural plants is strongly correlated (Kamei-Ishikawa et al., 2011; Tsukada et al., 2002; Vinichuk et al., 2010). Moreover, no significant differences in bioavailability have been reported between the uptakes of these isotopes (Karadeniz and Yaprak, 2007; Rühm et al., 1999; Salt et al., 2004; Soudek et al., 2006).

Numerous soil and hydroponic experiments have focused on cesium translocation from plant roots to other plant organs (Kang et al., 2012; Moogouei et al., 2011; Singh et al., 2008, 2009; Soudek et al., 2004), as well as the absorption and uptake of cesium through leaves (Brambilla et al., 2002; Carini et al., 2003; Carini and Lombi, 1997; Henner et al., 2005; Macacini et al., 2002; Madoz-Escande et al., 2009; Shaw et al., 2007). However, data on the effect of accompanying anions on the retention and translocation of cesium in the leaves of contaminated plants are rare. Middleton and Sanderson (1965) revealed that cesium phosphate and cesium iodide have higher absorption rates than cesium sulfate. Hasegawa et al. (2009) reported a higher absorption rate for cesium as nitrate than as chloride in foliar uptake by radish (*Raphanus sativus* L. cv. Redchim). The chemical forms of wet deposition of radiocesium on plants are potentially diverse. Anions in precipitation that occurs as wet deposition typically include nitrate, sulfate, and carbonate. Iodide is another potential wet deposition anion that may also be present in the air, particularly in areas bordering oceans.

The current study aimed to determine the effect of four accompanying anions on the retention behavior of stable cesium (^{133}Cs) on soybean leaves by applying cesium solutions as droplets on the upper surface of soybean leaves. In addition, scanning electron microscopy coupled with energy dispersive X-ray (SEM-EDX) was used to determine the morphology and elemental composition of residual cesium in the leaves. The study also investigated the translocation of stable cesium within soybean plant organs following wet deposition on leaves.

2. Materials and methods

2.1. Cultivation of soybean

In April 2011, the experimental soil was collected at 0–30 cm depth from an agriculture area in a southeastern suburb of Beijing. The soil was air dried and then ground following the removal of twigs and stones, before being sieved through a 2-mm nylon screen. The physico–chemical characteristics of the soil were then analyzed. The particle size of the soil was determined using the pipette method, and the soil pH was measured in a 1:2.5 (w/v) mixture of soil and water using a glass pH electrode (Sartorius PB-10, Germany) (Lu, 2000). The $CaCO_3$ content of the soil was measured using the gas-volumetric method, and the organic carbon content of the soil was determined using the potassium dichromate oxidation spectrophotometric method (Lu, 2000). Total phosphorus was determined following H_2SO_4–$HClO_4$ digestion by the molybdenum blue spectrophotometric method, and total nitrogen was measured by the Kjeldahl method (Lu, 2000). The cation exchange capacity (CEC) was determined using the Ba–Mg exchange method (Lu, 2000). Soil cesium analyses used the USEPA-3050B method (USEPA, 1996), incorporating acid (HNO_3–HF–$HClO_4$) digestion in a Teflon pressure digest vessel at 165 °C for 4 h, with determination of the cesium content in solution by atomic absorption spectrometry (AAS) with a graphite tube equipped with an automatic sampler (ZEEnit 700, Analytik Jena, Germany) (Willey and Martin, 1995). The limit of detection for cesium was 0.02 μg L^{-1}. The main results of the experiment are shown in Table 1.

Table 1
Physico–chemical characteristics of the soil used for soybean plant culture.

Parameters	Value
pH in H_2O (1:2.5)	8.11
CEC (cmol kg^{-1})	9.87
$CaCO_3$ (g kg^{-1})	10.91
Organic matter (g kg^{-1})	18.60
Total nitrogen (g kg^{-1})	0.82
Total phosphorus (g kg^{-1})	0.88
Sand (2.0–0.05 mm) (%)	48.56
Silt (0.05–0.002 mm) (%)	50.41
Clay (<0.002 mm) (%)	1.03
Texture	Silty loam
Cesium (mg kg^{-1})	5.37

Soybean [*Glycine max* (L.) Merr.] was chosen as the experimental species because it is one of the most important and widely produced edible crops in China and globally. Also, the leaf cuticle structure of soybean is relatively homogeneous (Shaw et al., 2007). In this study, soybean plants were cultivated in polyvinyl chloride pots (120 mm inner diameter and 110 mm depth) filled with 1.5 kg of dry soil. Soluble base fertilizers at rates of 200 mg kg^{-1} of N, 150 mg kg^{-1} of P, and 200 mg kg^{-1} of K were applied as analytical grade NH_4NO_3 and KH_2PO_4 reagents to each pot and water was added to reach the maximum water holding capacity of the soil, which was then allowed to equilibrate for 2 weeks (Wu et al., 2009). Five seeds were sown in each pot on 13 July 2011 and thinned to four plants per pot on the third day after germination. Plants were grown in a greenhouse throughout the experiment, and watered with deionized water. The greenhouse did not have climate control but operated at an ambient temperature with natural illumination (cycle period of 11–14 h of light and 10–13 h of darkness). The maximum and minimum temperatures in the greenhouse were 28 °C and 19 °C, respectively. Cesium contamination of the leaves occurred at 57 d after sowing. On the BBCH scale (Von et al., 1997), the plants were then at the fruit and seed stage of development. The development stage is critical in relation to maximal development of foliage (maximal retention) and maximal internal fluxes to sustain seed and fruit filling (Madoz-Escande et al., 2004; Shinonaga et al., 1999a, b; Vandecasteele et al., 2001).

2.2. Droplets experiment

Solutions of cesium compounds, including $CsNO_3$, CsI, Cs_2SO_4, and Cs_2CO_3 were used as the four treatments in the experiment. Each solution contained 1.33 mg mL^{-1} of cesium. Each treatment was applied to 12 plants (three randomly selected3 pots of four plants). The pots were separated from one another to prevent cross-contamination. The four uppermost expanded trifoliate leaves (three leaflets) of each plant were contaminated by applying 10 drops of 2 μL droplets of Cs solution onto each leaflet, beginning with the uppermost leaf. Each plant in the four treatments finally received 240 μL of solution equivalent to 319 μg Cs, whatever the