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# Potassium applications reduced cesium uptake and altered strontium translocation in soybean plants

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## ABSTRACT

After the Tokyo Electric Power Company's Fukushima Dai-ichi Nuclear Power Plant accident in 2011, radioactive cesium (RCs) was released in greater concentrations than radioactive strontium (RSr) in the surrounding environment. Most of the countermeasures were developed to mitigate the RCs transfer from the soil to plants. However, to avoid what has happened after the Chernobyl and Mayak accidents, preventing the transfer of RSr from soil to plants should be a priority. Although the application of potassium (K) fertilizers is the most effective method for preventing agricultural crops from absorbing RCs in contaminated fields, this implementation increases the cost and labor requirements. Considering the preparedness for nuclear accidents, it remains unclear how this countermeasure will be affected if RCs and RSr are released simultaneously. We aimed to explore the effect of K applications on cesium (Cs) and strontium (Sr) uptake and their interaction with and correlation to other elements in the soybean plants and soil. The field experiments were conducted in Fukushima Prefecture, Japan, using different K applications (i.e., no, normal, and high K applications). The dry weight and mineral concentrations of K, Cs, Sr, calcium (Ca), magnesium (Mg), and nitrogen (N) concentration in plants and exchangeable K (ExK), exchangeable Cs (ExCs), exchangeable Sr (ExSr), exchangeable Ca (ExCa), exchangeable Mg (ExMg), NH<sub>4</sub><sup>+</sup> (ammonium), and NO<sub>3</sub><sup>-</sup> (nitrate) concentrations in the soils were evaluated. This study revealed that K application reduced Cs, Ca, and Mg uptake but did not affect the ExSr, ExCa, and ExMg concentrations in the soil and did not change the uptake of Sr. On the other hand, K concentration of the plant especially at later growth stage, which indicates re-translocation of Sr was negatively regulated by K concentration.

## ARTICLE HISTORY

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## KEYWORDS

Soybean; cesium; strontium; countermeasure; potassium

## 1. Introduction

The earthquake and subsequent tsunami that occurred at the Tokyo Electric Power Company's Fukushima Dai-ichi Nuclear Power Plant (FDNPP) in 2011 released several radionuclides including radioactive cesium (RCs) and radioactive strontium (RSr) to eastern Japan (Morino, Ohara, and Nishizawa 2011). RCs have relatively long half-lives (e.g., 30.2 years for <sup>137</sup>Cs and 2.06 years for <sup>134</sup>Cs) and high-energy emissions of β and γ radiation. <sup>90</sup>Sr is a β-ray emitting radionuclide with a high fission yield, relatively half-life (29 years), and high transferability to plants (Konno and Takagai 2018; Tsukada et al. 2005). RCs has been released in greater concentrations than RSr in the case of the FDNPP accident. However, the transfer of RSr from soil to plants also needs to be prevented to avoid what has happened in the previous Chernobyl and Mayak accidents (Chu et al. 2015).

After the FDNPP accident, the application of potassium (K) fertilizers was the most effective method and practical countermeasure for preventing agricultural crops from absorbing RCs in contaminated fields (Fujimura et al. 2014; Kato et al. 2015; Kubo et al. 2015; Matsunami et al. 2021). However, the result of this implementation may also influence strontium (Sr) behavior,

even though it is less of an issue in the case of the FDNPP accident. Nuclear accident preparedness must include countermeasures, especially if RCs and RSr will be handled simultaneously. How K application for RCs mitigation alters RSr uptake should be evaluated. This may help support and validate the application of K to reduce the absorption of RCs and RSr in the event of a future incident.

The addition of K application to the soil may also change the soil's nutrient balance, which impacts on plant growth. Nitrogen (N) is a determinant element in soybean growth (Osaki, Shinano, and Tadano 1992). Previous reports have shown that cesium (Cs) uptake can increase with increasing nitrogen (N) concentration and decreasing K concentration in plants (Evans and Dekker 1969; Belli et al. 1995). The uptake of elements such as Cs, Sr, K, and calcium (Ca) are positively correlated with each other (Chu et al. 2015) but their interaction with N remains poorly understood. Thus, a study of the role of these various nutrients in response to the effect of K application to reduce the rate of radionuclide transmission from soil to plants is necessary to ensure a harmonious balance between soil and plants. This nutrient balance is the key to increasing nutrient use efficiency to maintain soil fertility and plant productivity.

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In this study, we conducted field experiments on soybean plants treated with K applications in Date City, Fukushima Prefecture, for two growing seasons (2019–2020). Despite studies that have found soil K and Ca affect Cs and Sr uptake in lettuce (Roca and Vallejo 1995), their interrelationship has not been elucidated especially between K and Sr. The K applications have been shown to reduce Cs transport from soil to plants, but it has not been determined if K also reduces Sr transport. And if it occurs, K application for the remediation of agriculture after nuclear accidents can be used not only for radioactive Cs but also for Sr. The purpose of this study is to determine how K application affects the uptake of Cs and Sr simultaneously by soybean plants, one of Fukushima's most important agricultural products.

## 2. Material and methods

### 2.1. Experimental design

The experiment was conducted at Date City in Fukushima Prefecture, Japan in 2019 and 2020. The soil type is gray lowland soil, based on the classification of cultivated soils in Japan. Each field was fertilized before cultivation with the following nutrients: 20 kg N  $\text{ha}^{-1}$  ( $(\text{NH}_4)_2\text{SO}_4$ ) before the seedling stage, and 60 kg N  $\text{ha}^{-1}$  after the flowering stages, and 100 kg  $\text{P}_2\text{O}_5 \text{ ha}^{-1}$  ( $\text{Ca}_3(\text{PO}_4)_2$ ) with 1000 kg  $\text{ha}^{-1}$  magnesium lime applied on the same day. The experimental treatments comprised three application levels of K: no K application, normal K application (increasing the level of exchangeable K (ExK) up to 25 mg  $\text{K}_2\text{O} \text{ 100 g}^{-1}$  and applying 100 kg  $\text{K}_2\text{O} \text{ ha}^{-1}$ ), and high K application (increasing the level of ExK up to 45 mg  $\text{K}_2\text{O} \text{ 100 g}^{-1}$  and applying 100 kg  $\text{K}_2\text{O} \text{ ha}^{-1}$ ). The amount of K (100 kg  $\text{K}_2\text{O} \text{ ha}^{-1}$ ) was applied as potassium sulfate ( $\text{K}_2\text{SO}_4$ , 50.0%  $\text{K}_2\text{O}$ ). In 2019, 132 g/22.5 m<sup>2</sup> of K was applied to increase the level as normal K application and 551 g/22.5 m<sup>2</sup> of K was applied to increase the level as high K application. In 2020, 0 mg/22.5 m<sup>2</sup> of K was applied to increase the level as a normal K application, and 664 mg/22.5 m<sup>2</sup> to increase the level as a high K application. A randomized block design with three replications in each field was used. Soybeans (*Glycine max* (L.) Merr. var. Tachinagaha) were sown on May 27 and harvested on 28 October 2019; and sown on June 23 and harvested on 26 October 2020.

### 2.2. Sample collection

Plant and soil samples were collected from each field at each growth stage (vegetative, flowering, pod formation, maturity, and harvest). Plant samples were divided into the stems (including petiole), leaves, pods, and seeds. They were then dried at 80°C for two days, weighed, and homogenized for subsequent elemental analysis.

Soil samples were collected from a depth of 15 cm around the plant roots using a worm scoop (Fujiwara Scientific Co., Ltd.). Soil samples were air-dried at 40°C for 1 week and then passed through a 2.0 mm sieve, for chemical analysis.

### 2.3. Mineral element measurements

The elemental composition of the plants and soil was determined as previously described (Watanabe et al. 2021). Dried plant samples (stems, leaves, pods, and seeds) were incubated with 2 mL of 61% (w/v)  $\text{HNO}_3$  at 110°C in a DigiPREP apparatus (SCP Science, Quebec, Canada) for 2 h until the solution had almost disappeared. After cooling, 0.5 mL of hydrogen peroxide ( $\text{H}_2\text{O}_2$ ) was added, and the samples were incubated at 110°C for 20 min. Once digestion was complete, the tubes were cooled, and the volume was adjusted to 10 mL by adding 2% (w/v)  $\text{HNO}_3$  to ultrapure water. The concentrations of K, Cs, Sr, Ca and Mg in the digests were analyzed using an inductively coupled plasma mass spectrometry (ICP-MS: ELAN DRC-e; PerkinElmer, Waltham, MA, U.S.A.). To measure N concentrations in the plants using the Kjeldahl method (Watanabe et al. 2015), the dried plant samples were digested with  $\text{H}_2\text{SO}_4$  (98%)- $\text{H}_2\text{O}_2$ .

For elemental analysis of the soil, ExK, exchangeable Cs (ExCs), exchangeable Sr (ExSr), exchangeable Ca (ExCa), and exchangeable Mg (ExMg) were extracted from dried soil samples at a soil: solution ratio of 1:10 in 1 M ammonium acetate ( $\text{NH}_4\text{OAc}$ ) with shaking for 1 h. Soil ExK, ExCs, ExSr, ExCa, and ExMg concentrations were measured using ICP-MS. For the analysis of inorganic N, ammonium ( $\text{NH}_4^+$ ) and nitrate ( $\text{NO}_3^-$ ) in the soil, the dried soil samples (4 g) were shaken with 2 M KCl (40 mL) in 50 mL polycarbonate tubes on and end-to-end shaker (150 rpm, 60 min). The soil was filtered using filter paper, and the concentrations in the supernatant were determined by colorimetric assays at 630 and 538 nm with a microplate reader (BioTek EPOCH<sup>2</sup> Microplate Reader) for ammonium and nitrate, respectively.

For measurement of ammonium, we used the Indophenol method according to (Scheiner 1976), with minor modifications. The supernatant was prepared by adding 1 mL of the sample solution, 2 mL of distilled water, and 1 mL of a combination of A and B color-developing liquids (0.4 mL of A and 0.6 mL of B color-developing liquids). The A color-developing liquids are containing 15 g of phenol dissolved in 200 mL buffer solution and adjusted to 250 mL by adding 0.05 g of sodium nitroprusside. The B color-developing liquids are containing 40 mL of 1 M NaOH to 1 mL of sodium hypochlorite solution and adjust to 100 mL with distilled water. Before measuring the absorbance, the supernatant was well stirred and left for 60 minutes.

For the measurement of nitrate, we used the Ando and Ogata method (Ando and Ogata 1980). Prepare for 2.5 M ammonia solution by adjusting 16.7 mL of 28% special grade ammonia to 100 mL with 0.25 M KCl solution. Prepare for reduction auxiliary stock solution by dissolving 186.4 g of KCl in 0.8 L of distilled water, add 167 mL of special grade ammonia water (28%), then adjusts to 1 L with distilled water. The first color former solution was prepared by adding 100 mL of distilled water and 100 mL of concentrated HCl to 500 mg of sulfanilamide to dissolve and then adjust to 1 L with distilled water. The second color former solution was prepared by dissolving 50 mg of *N*-1-Naphthylethylenediamine dihydrochloride ( $\text{C}_{12}\text{H}_{16}\text{Cl}_2\text{N}_2$ ) with distilled water and adjusting to 1 L. The reduction auxiliary stock solution was diluted 10-fold and took 4 mL then placed in a test tube with adding 1 mL of sample solution,

0.75 g of metallic zinc, and plugged. The test tubes were immediately shaken for 15 minutes and took 2 mL of the supernatant into another test tube. The supernatant was left for 20 minutes and add 2 mL of each of the first and second color former solutions. The supernatant was well stirred and left for 10 minutes before measuring the absorbance.

#### 2.4. Statistical analyses

Data were statistically analyzed using SPSS Statistics 25 (SPSS Inc., Chicago, IL, U.S.A). One-way analysis of variance (ANOVA) was used to evaluate the results at  $p < 0.05$  probability level. Tukey's test was calculated only when the ANOVA F-test indicated significant treatment effects at the significant level ( $p < 0.05$ ). Values are reported as mean  $\pm$  SE of three replicates. Pearson's correlation analysis was conducted to evaluate the relationship between the mineral elements in the plant and soil. The figures were visualized using R Studio.

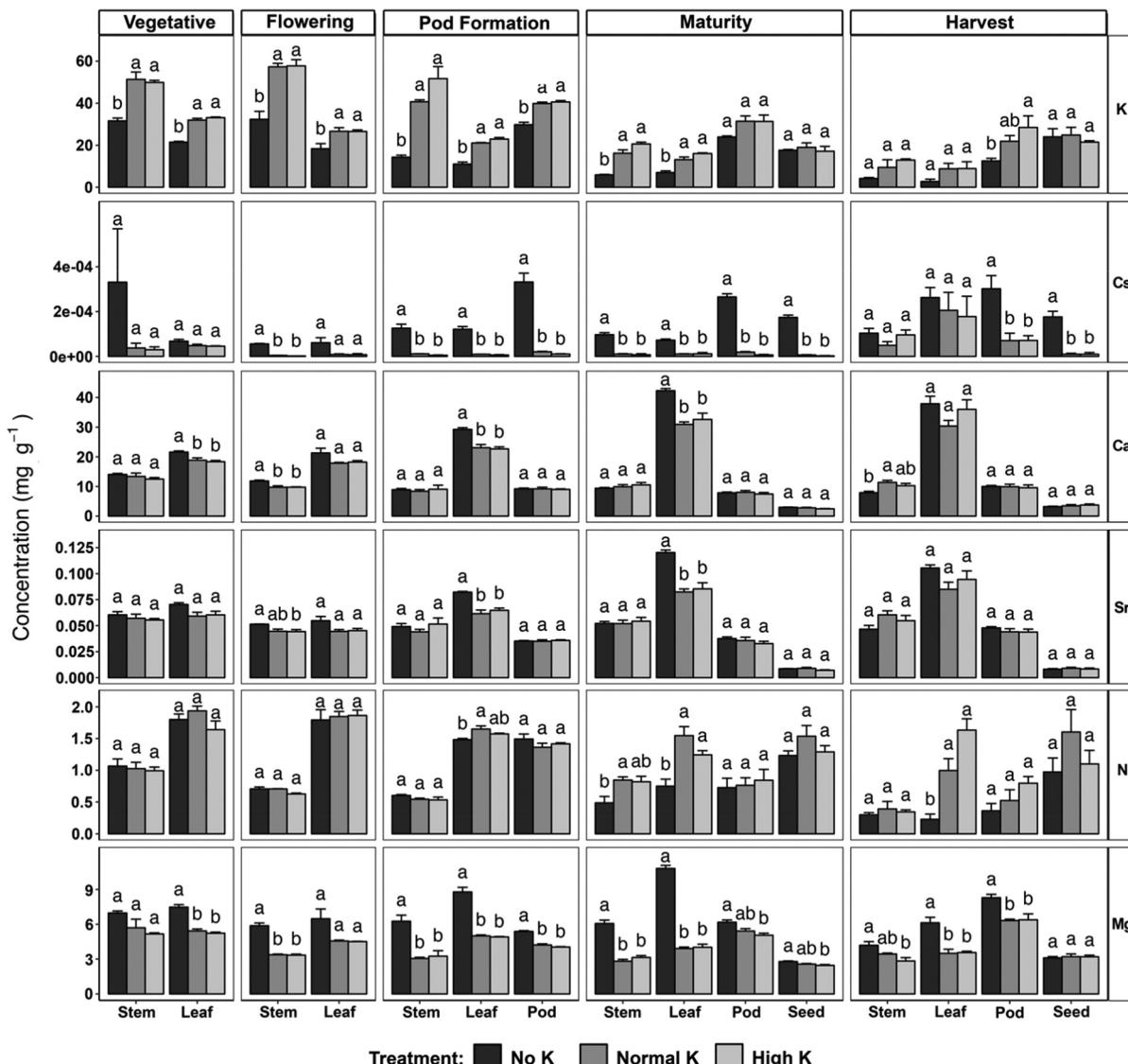
### 3. Results

#### 3.1. Dry weight

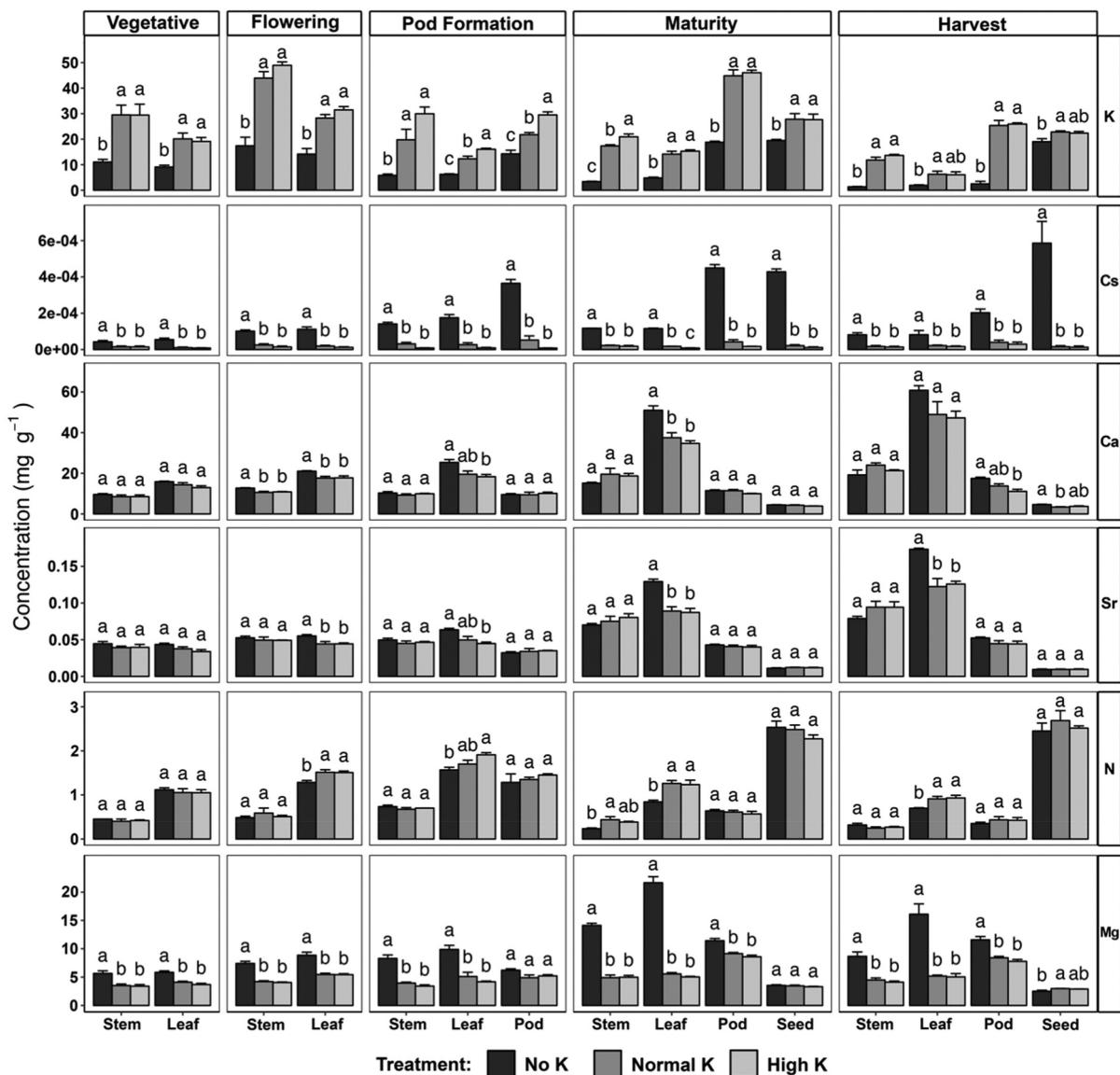
The effect of K applications on the dry weight of the stems, leaves, pods, and seeds of soybean plants is shown in Figure S1. The dry weight did not differ in all growth stages during the two years with increasing K application, except for the stems and leaves at the vegetative stage, stems, and pods at harvest in 2019, and stems and leaves at maturity and stems at harvest in 2020. The dry weight was relatively lower without the application of K.

#### 3.2. Effect of K application on mineral elements in plant

The concentrations of K, Cs, Sr, Ca, Mg, and N in various parts of the soybean plants in response to K applications are shown in Figures 1 and 2 for 2019 and 2020, respectively. The K concentration in all plant organs (stems, leaves, pods, and seeds) differed significantly among K applications in both years,



**Figure 1.** The effect of K applications on K, Cs, Sr, Ca, N and Mg concentrations in soybean plants at date city in 2019. The vertical bars indicate the standard error of three replicates. The different letters (a, b, and ab) represent significant differences ( $p < 0.05$ ) according to Tukey's test following a one-way ANOVA ( $n = 3$ ).



**Figure 2.** The effect of K applications on K, Cs, Sr, Ca, N and Mg concentrations in soybean plants at date city in 2020. The vertical bars indicate the standard error of three replicates. The different letters (a, b, ab, and c) represent significant differences ( $p < 0.05$ ) according to Tukey's test following a one-way ANOVA ( $n = 3$ ).

except for pods and seeds at maturity, and stems, leaves, and seeds at harvest in 2019. The K concentration in the plant organs was relatively higher in applications with higher K levels. For example, the K concentration in pods was ten times higher in the high K application than in the no K application at the harvest stage in 2020. However, the stable Cs in all plant parts differed significantly among K applications in both years, except for stems and leaves at vegetative and harvest stages in 2019. Increased K application has generally led to decreased stable Cs concentrations in the plant parts. For example, the stable Cs concentration of seeds at the harvest stage in 2020 was 46 times lower with high K application than without K application.

Except for the leaves at the pod formation and maturity stages in 2019 and the flowering to harvest stages in 2020, the Sr concentration did not differ among K applications. Increased K application has generally led to decreased Sr concentrations in the leaves. For example, the largest

decrease in the Sr concentration of leaves was observed in high-K applications at the maturation stage in 2020. However, the Ca concentration in leaves and stems relatively decreased with increasing K application levels at all growth stages in both years, except for the vegetative and harvest stages in 2019, and the vegetative stage in 2020. The largest decrease in Ca concentration was observed in pods at the harvest stage in 2020. In contrast, the Ca concentration of the stems increased with increasing K application level at the harvest stage in 2019. Moreover, Mg concentration in all plant organs differed significantly among K applications in both years, except for stems at vegetative, leaves at flowering, and seeds at harvest in 2019, and pods at pod formation and seeds at maturity in 2020. Increased K application has generally led to decreased Mg concentrations in the plant parts. For example, the Mg concentration of leaves at the maturation stage in 2020 was 76 times lower with high K application than without K application.

The N concentration of stems and leaves generally increased with increasing the K application levels from pod formation to harvest stages in 2019 and flowering to harvest stages in 2020. The largest differences in N concentration of soybean plants were between the high K application and the no K applications. For example, at the harvest stage in 2019, the leaf N concentration was seven times higher with the high K application than with no K application. Conversely, the differences were smaller at early stages in both years, such as during vegetative growth and flowering in 2019 and vegetative growth in 2020.

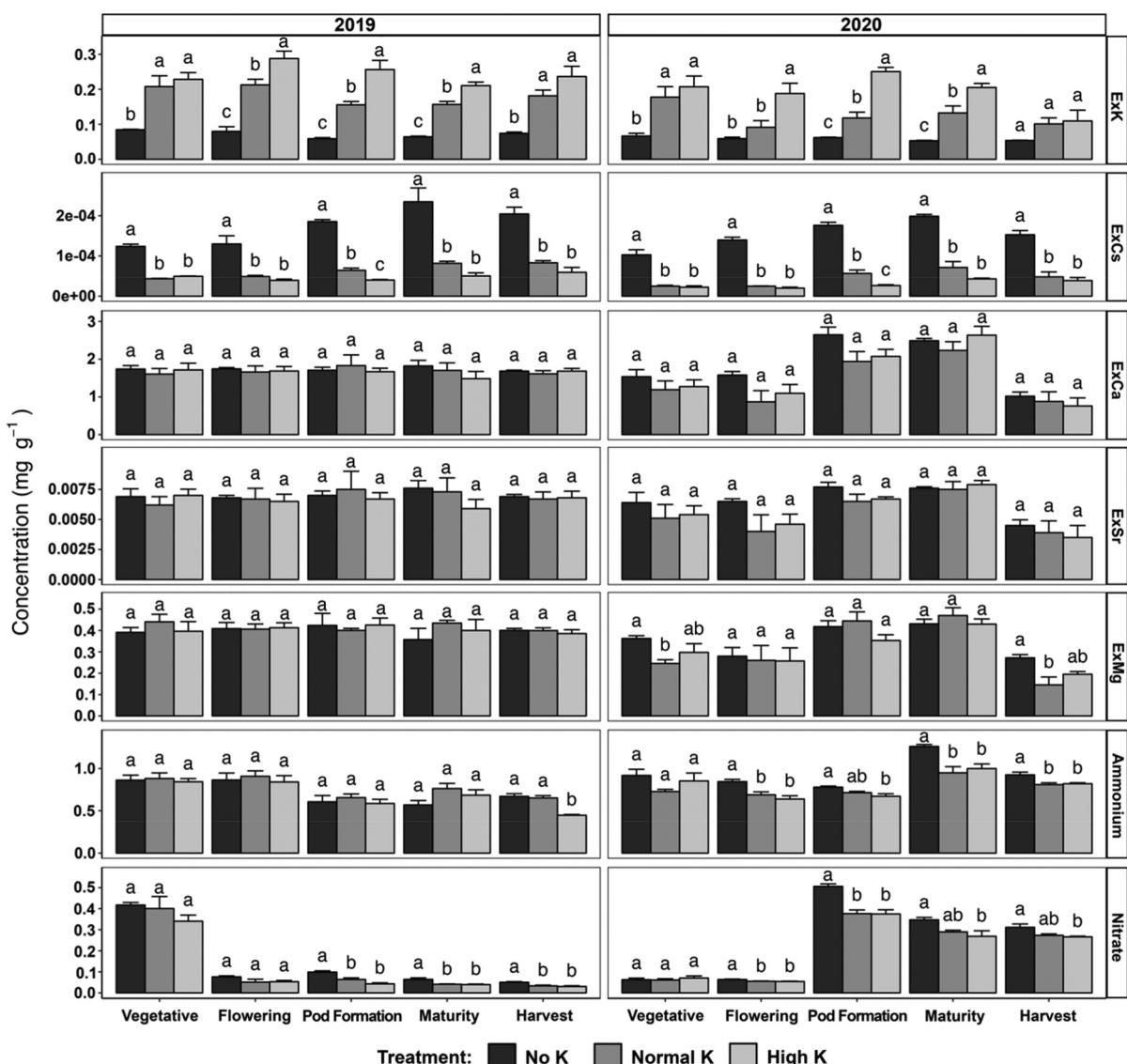
### 3.3. Effect of K application on mineral elements in soil

The ExK, ExCs, ExSr, ExCa, ExMg, ammonium, and nitrate concentrations of the soil in response to K applications are shown in Figure 3. The ExK concentration in the soil differed significantly among K applications in both years, except for the

harvest stage in 2020. The ExK concentrations in the soil was relatively higher in applications with higher K levels. For example, ExK concentrations at the pod formation stage in 2019 were four times higher with high K application than without K application.

However, the ExCs concentrations at all stages differed significantly among K applications in both years. Moreover, increased K application has generally led to decreased ExCs concentrations in the soil. For example, the highest reduction in ExCs concentrations in the soil was at the flowering stage in 2020, which decreased six-fold with high K application. However, the ExSr, ExCa and ExMg concentrations in all stages did not differ among the K applications at any growth stages in either year, except at vegetative and harvest in 2020 of ExMg concentrations.

The ammonium concentration in the soil generally decreased with increasing the K application levels at the harvest stage in 2019 and from flowering to harvest stages in 2020.



**Figure 3.** The effect of K applications on ExK, ExCs, ExSr, ExCa, ExMg, ammonium and nitrate concentrations in the soil at date city in 2019 and 2020. The vertical bars indicate the standard error of three replicates. Different letters (a, b, c, and ab) represent significant differences ( $p < 0.05$ ) according to Tukey's test following a one-way ANOVA ( $n = 3$ ).



The largest decrease in ammonium concentration in the soil was observed in the harvest stage with high K application in 2019. On the other hand, the difference was small during the vegetative to maturity stages in 2019 and the vegetative stage in 2020. However, the nitrate concentration in the soil was reduced by increasing the K application from pod formation to harvest in 2019 and from flowering to harvest in 2020. A large decrease in nitrate concentration in the soil was observed during the pod formation stage in 2019 with high K application. The reduction in nitrate in the soil was two times higher with K application than without K application at the pod formation stage in 2019. In contrast, the difference in nitrate concentration was relatively small during the vegetative and flowering stages in 2019 and the vegetative stage in 2020. Moreover, the nitrate concentration in 2020 increases from flowering to pod formation.

### 3.4. Correlations among mineral elements in plant and soil

The Pearson correlation analysis was performed to establish the correlation between the concentration of each pair of elements in plants (K-Cs, K-Sr, K-Mg, Sr-Ca, Sr-N, and Sr-Mg) and soil (ExK, ExCs, ExK-ExSr, ExK-ExMg, ExSr-ExCa, ExSr-ExMg, ExSr-ammonium, and ExSr-nitrate) under K application (Table 1). The Pearson correlation analysis of mineral elements in plants showed that K concentration was significantly negatively correlated with Cs and Sr concentrations in both years, and but only with Mg concentrations in 2020 (Figure S2). The Sr concentration was significantly positively correlated with Ca and Mg concentration in both years and significantly inversely correlated with N concentration in 2020 (Figure S3). In order to further elucidate the relationship between K and Sr, we explored its relationship in growth stages and plant organs. The K concentration was significantly negatively correlated with Sr concentration in all growth stages except vegetative and in all plant organs except seed (Figure 4).

The Pearson correlation analysis of mineral elements in soil showed that the ExK concentration in the soil was significantly negatively correlated with ExCs concentrations in the soil in both years but was not likely to be correlated with ExSr and ExMg concentrations in the soil (Figure S4). The ExSr concentration in the soil was significantly positively correlated with the

ExCa and ExMg concentrations in the soil in both years (Figure S5). The ExSr concentration in the soil was unlikely to be correlated with ammonium and nitrate concentrations in the soil in 2019 but significantly in 2020 (Figure S5).

## 4. Discussion

Since the nuclear incident in the FDNPP, the practice of applying fertilizer in cultivation activities has been recommended as a preventive measure to reduce RCs transmission to the edible parts of plants. The application of K in cultivation has successfully reduced the rate of RCs translocation (e.g., Kato et al. 2015). However, there is no explanation for how this counter-measure relates to Sr. It is necessary to consider the simultaneous release of RCs and RSr into the environment should another nuclear accident occur. This implementation may also affect the nutritional balance and the state of other elements such as K, Cs, Sr, Ca, Mg and N, and the interrelationships of these elements are poorly understood.

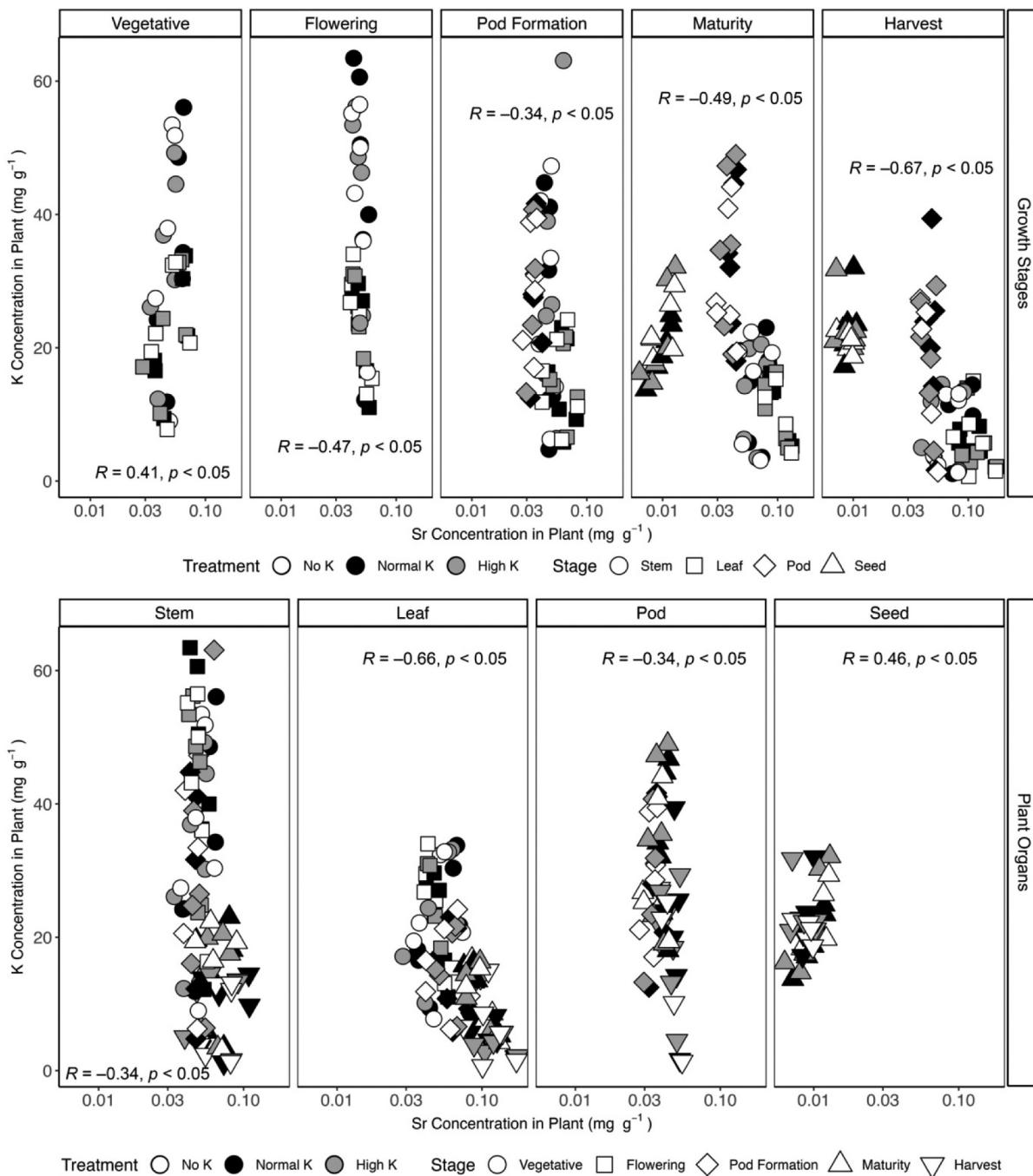
### 4.1. Effect of K application on K, Cs, Sr, Ca, Mg, and N concentration in plant

Except for the pods and seeds at the mature stage and stems, leaves, and seeds at the harvest stage in 2019, the K concentration in all plant parts differed significantly among K application in both years (Figures 1 and 2). The largest difference in K concentration in soybean plants was in the pods at the harvest stage in 2020, when high K application was ten times higher than no K application. A higher K concentration in plant organs can be attributed to a higher K concentration in the soil, although the dry weight does not generally vary among the K treatments (Figure S1), as confirmed by previous studies on buckwheat (Kubo et al. 2017) and soybean (Matsunami et al. 2021). However, increased K application has generally led to a decrease in stable Cs concentrations (Figures 1 and 2) in all plant organs. The largest decrease in stable Cs concentration was observed in the seeds at the harvest stage in 2020 with high K application. A similar tendency was also observed in the analysis of RCs uptake behavior using the same field (Suzuki et al. 2023), which indicate that a similar behavior of RCs and Cs was observed nine years after the accident. These

**Table 1.** Tabulated summary of Pearson correlation analysis among elements in plant and soil.

Parameters	2019		2020	
	Pearson's <i>r</i>	<i>p</i> -value	Pearson's <i>r</i>	<i>p</i> -value
K vs Cs (plant)	-0.29	0.05*	-0.22	0.05*
K vs Sr (Plant)	-0.28	0.05*	-0.49	0.05*
K vs Mg (plant)	-0.17	0.05	-0.39	0.05*
Sr vs Ca (plant)	0.92	0.05*	0.93	0.05*
Sr vs N (plant)	0	0.95	-0.47	0.05*
Sr vs Mg (plant)	0.5	0.05*	0.52	0.05*
ExK vs ExCs (soil)	-0.83	0.05*	-0.7	0.05*
ExK vs ExSr (soil)	-0.13	0.4	0.16	0.3
ExK vs ExMg (soil)	-0.07	0.7	0.17	0.3
ExSr vs ExCa (soil)	0.92	0.05*	0.93	0.05*
ExSr vs ExMg (soil)	0.88	0.05*	0.96	0.05*
ExSr vs ammonium (soil)	-0.11	0.5	0.4	0.05*
ExSr vs nitrate (soil)	-0.04	0.8	0.34	0.05*

Significant modules are denoted with \* (*p* < 0.05).



**Figure 4.** Pearson's correlation between K and Sr concentration in soybean plants after K applications at date city in the different growth stages and plant organs.

results are consistent with other findings that K application effectively reduces RCs (Kato et al. 2015) and stabilizes Cs (Tsukada et al. 2002) in rice. K application might be inhibiting the translocation of Cs from soil to plant regardless of radioactive or stable status due to competition between K and Cs at the root absorption site. Plant root cell membranes appear to be mainly involved in Cs uptake via two transport pathways, namely the K transporter and the K channel pathway (Zhu and Smolders 2000). Cs and K have similar chemical properties and compete for the binding sites in proteins (Avery 1995). According to (Fujimura et al. 2014), adequate external K levels inhibit the expression of K transporters with a high affinity for Cs.

The increased K application led to decreased Sr concentration in leaves, Ca concentration in the leaves and stems (Figures 1 and 2) as confirmed by previous studies (Neilsen and Edwards 1982; Melnitchouk and Hodson 2004; Tuma et al. 2004). Additionally, Mg concentrations in plant parts were reduced relatively by increased K application (Figures 1 and 2) as confirmed by previous studies (Lanyon, Heald, and Page 1983; Kang, Seo, and Ishii 2019). Our results showed that with a high K application, the leaves at maturation stages in 2020 displayed the largest drop in Sr and Mg concentrations. However, the largest decrease in Ca concentration appeared with high K application in the pods at the harvest stage in 2020. Sr and Ca or Mg are chemically similar and compete for the same receptor sites on biological membranes, but Sr cannot replace



Ca or Mg in plants. More than 50% of Sr is absorbed by plants and accumulates in cell walls, similar to Ca (Anupama, Ashok, and Naveena 2016; Gupta et al. 2018; Qiu et al. 2021). Ca is relatively immobile in plant cells and is not readily circulated to tissue parts despite the high Ca applied (Knez and Stangoulis 2021). Ca is involved in the response to external stimuli in major cellular processes, but the response depends on the movement of K across membranes (Sardans and Penuelas 2021) while Sr is transported from the roots via a K plasma membrane transporter (Burger and Lichtscheidl 2019). Moreover, the presence of high Ca concentrations can also lead to Mg deficiency (Hansen and Munns 1988; Plaut and Grieve 1988). In case of Mg, the physiological role of this divalent cation is different from Ca. There are many fundamental functions of Ca in cellular metabolism, while Mg plays a significant role in chlorophyll synthesis, ion transport, and cation balance regulation, in addition to activating more than 300 enzymes (He et al. 2012). As in this study, the decreased Ca concentrations were similar to the decreases in Sr concentrations due to the K application. The Sr and Ca concentrations and also Sr and Mg concentrations had a similar pattern in leaves, i.e., decreasing with increasing K application at the pod formation and maturity in 2019, and at the flowering and maturity stage in 2020 (Figures 1 and 2). Sr may use the same cell entry mechanisms as Ca and Mg, such as plasma membrane transporters, due to its physicochemical similarities (Burger and Lichtscheidl 2019). Ca can be replaced by Sr, thereby lowering Mg and Ca content in plants (Burger and Lichtscheidl 2019; Moyen and Roblin 2010). Increasing K in soil solution might influence the presence of other ions (including K, Ca, Mg and Sr) which is played a role in the adsorption and release of Sr and RSr due to competition for exchange sites, thus influencing Sr uptake (Burger and Lichtscheidl 2019). Also, the involvement of other elements, such as Sr, interferes with Ca uptake (Rato et al. 2010) and Mg uptake (Moyen and Roblin 2010). The decreased Sr and Ca levels in the leaves in this study as confirmed by previous reports (Moyen and Roblin 2010; Chen et al. 2012; Zhang et al. 2020) might be due to the indirect involvement of Mg after K was applied (Trankner, Tavakol, and Jakli 2018; Ding et al. 2016). While these findings suggest that K application in the soils suppresses Sr transport to the leaves but does not affect the soybean's dry weight it might be due to ExMg concentration in the soils (Figure 3) affected to Mg uptake then dry weights as confirmed by a previous study (Hailes, Aitken, and Menzies 1997). The Mg element is instrumental in forming dry matter and partitioning carbon to sink organs since carbohydrate accumulation in source leaves is reduced by Mg deficiency (Gransee and Fuhrs 2013) as confirmed by our results (Figures 1–2). There is a similar pattern between Sr and Mg concentrations in leaves that decreased with increasing K. Despite its small amount in this study, Sr is not detrimental to plant growth, and its uptake is a side effect of divalent cation absorption. Nevertheless, the K level in this study was adequate for seed production.

#### **4.2. Effect of K application on ExK, ExCs, ExSr, ExCa, ExMg, ammonium, and nitrate concentration in soil**

The ExK concentrations in the soil were relatively higher in application with higher K levels in both years, except for the harvest stage in 2020 (Figure 3). This is expected owing to the application of K in this study. However, there is no difference between normal and high K application to ExK concentrations in the soil at

beginning (vegetative) and at the end of growth stages (harvest) in both years. It might be due to the characteristics of K uptake which is slowed in the vegetative stage but increases rapidly during flowering maturity stages (Halevy 1976; Mullins and Burmester 1990). In addition, it could be caused by soil properties, even though they were excluded from this study. Uzoho and Ekeh (2014) reported that soil K status affects by sand, silt, clay, silt/clay ratio, organic matter, pH, total N, P, Ca, Mg, cation exchange capacity, base saturation, sodium (Na), and hydrogen (H).

K application generally led to decreased ExCs concentrations in the soil in both years (Figure 3). Increasing ExK in the soil tends to decrease ExCs, which has also been observed in various crops (Kato et al. 2015; Kubo et al. 2015, 2017, 2018; Matsunami et al. 2021). ExK concentrations in the soil increased following K fertilizer application and suppressed RCs uptake during the growing season (Komatsu et al. 2017), which is also expected for stable Cs. Generally, Cs ions exchange with K ions at frayed edge sites (Okumura, Nakamura, and Machida 2013). The reduction in ExCs might be due to formation of the frayed edge sites on the clay minerals after K applied (Kubo et al. 2017). In contrast, the ExSr and ExCa, and ExMg concentrations in the soil (Figure 3) did not differ among K applications in either year, except at vegetative and harvest in 2020 of ExMg concentrations. K, Ca and Mg (which have similar characteristics to Sr) share the same soil particle binding site (Bonomelli, Gil, and Schaffer 2019). Consequently, excess or deficiency of K can affect the availability of the other cations in the soil, including Sr and Ca or Sr and Mg. For example, K application to the soil might decrease the ease of replacing the clay fraction Mg, and consequently, less Mg is available for uptake (Hovland and Caldwell 1960). Tuma et al. (2004) revealed that K and Ca could not be handled separately from other nutrients because antagonistic relationships arise. Though we have found that Sr uptake was decreased in the shoot of soybean by the application of K, we did not analyze the root, so the possibility of root-to-shoot transfer was decreased by K application is remained. In order to determine whether K application has caused Sr uptake and/or distribution among plants, a confirmation test should be conducted.

The ammonium and nitrate concentrations in the soil (Figure 3) generally decreased with increasing K application, as confirmed by previous reports (Beauchamp 1982; Nguyen et al. 2001; Ajazi et al. 2013). Increased K application might result in saturation in the interlayer space in the soil, which decreases ammonium fixation or impairs ammonium release (Scherer 1982; Nieder, Benbi, and Scherer 2011) and decreases nitrate concentrations in the soil. If plants do not absorb nitrate in the soil solution, it can easily leach because nitrate and soil are negatively charged (Ito 2018). Moreover, nitrate levels in the soil increased from flowering to pod formation in 2020, might be due to the additional application of N fertilizer in this study that applied after flowering stages then influences nitrate concentration in the soil.

#### **4.3. The relationship of Sr with K with the reduction of Cs concentration**

Correlation analysis showed that the K concentration in the plant organs was significantly negatively correlated with stable Cs and Sr concentrations in the plant organs in both years, and but only with Mg concentrations in 2020 (Figure S2). However,

the effect of K application on Sr concentration was not observed unlike Cs or Mg (Figures 1 and 2). These results reveal that K application reduces the transfer of Cs and Mg from soil to plant organs. However, the different mechanisms should be considered in the case of K's relationship with Sr. Though it was also indicated that higher K concentrations in the plant organs were accompanied by lower Cs, Sr, and Mg concentrations in the plant organs after K application, as confirmed by previous studies (Kabata-Pendias and Szeke 2015; Myrvang et al. 2016), we have further analyzed the relationship between K and Sr relationship divided by the growth stages and plant organs (Figure 4). The relationship between K and Sr clearly changed with growth stages, and it was demonstrated that during the early growth stages, the Sr concentration was rather stable regardless of the K concentration level in the plant while there was a clear negative relationship was observed during the maturity and harvest stages. This is indicating that the K and Sr movement in the plant is different. Plants can rapidly transfer K throughout their entire organ system, but Ca becomes largely immobile, and Sr behaves similarly to Ca (Creger and Allen 1969; Mengel and Munson 1985). It is further confirmed by the differences between plant organs (Figure 4). Significant negative correlations were observed except for seeds, and the relationship was most obvious in the leaf, and it was also confirmed that the K and Sr decreased with the growth. Since our results only showed a reduction in Sr concentration in the leaves while reductions in Mg concentrations relatively occurred in all plant organs after K application (Figures 1–2), also the Sr concentration was significantly positively correlated with Ca and Mg concentration in both years (Figure S3), it was estimated that the absorption of Sr was not disturbed by the K application while the re-translocation of Sr was disturbed, while it is further requested to investigate how Sr was stored and distributed with the change of K levels in the organs. When Sr was absorbed into plants and their cells using K and Ca channels (Burger and Lichtscheidl 2018, 2019), the K effect may have altered the behavior of these channels.

Moreover, the ExK concentration was significantly negatively correlated with the ExCs concentration in both years (Figure S4). The ExK concentration in the soil was unlikely to be correlated with the ExSr concentration in the soil and ExMg concentrations in the soil (Figure S4). It can be explained that increasing the ExK concentration in the soil suppressed ExCs and not ExSr, ExCa and ExMg concentrations in the soil. Previously, Roca and Vallejo (1995) reported on the effects of soil K and Ca on RCs and RSr transfer to lettuce plants, but the relationship of K to Sr has not yet been clarified. Our results revealed that K application directly decreased Cs uptake, and indirectly decreased Sr and Mg uptake by the soybean plants. During root absorption, it may be not only the involvement and competition between K and Cs that influence Cs translocation from soil to plants, but there may also be the effect of related elements (such as Mg) on Sr translocation from soil to plants. The large amounts of K competitively inhibited Mg uptake and resulting in reduced protein synthesis (Guo et al. 2016). An indirect relationship between K suppress the uptake rate of Sr needs further investigation. It is particularly important to examine the mineral balance of the related elements that might support the application of K for reducing RCs and RSr

translocation from soil to plants in a preparedness scenario for nuclear accidents.

There was significantly positively correlated between Sr with Ca and Sr with Mg concentration occurred in this study, either in plant or soil. However, the role of K in the soil-plant interaction between Sr and Ca is not yet clear. The results of this recent study illustrate that (1) K application did not affect Sr transport, but it might affect re-translocation of Sr; (2) the link between K and Sr clearly changed with different growth phases, and it was shown that throughout the early growth stages, the Sr concentration was largely steady regardless of the quantity of K in the plant; (3) it is likely that K interacts with both Sr and Ca or Mg when entering the plant's cells, since K and Ca or Mg share channels during transport to plant organs with Sr (Burger and Lichtscheidl 2019); or (4) during the reduction of Cs uptake using K application to the soil, these cations (Ca, Mg and Sr) compete through competition in the apoplast of the root cortex (Smolders et al. 1997). The Sr concentration was small and there was no actual reduction was observed by K application, which indicate that K application does not support the idea to reduce RSr. However, as the re-translocation of Sr was reduced when K concentration is low, it is important to investigate whether this mechanism could occur in other species and/or in the combination of other minerals in the future.

## 5. Conclusions

Our results indicate that K application to soybean plants influenced the variables in this study. K application generally increased the K and N concentrations and decreased the Cs, Ca, and Mg concentrations in plant organs but did not affect dry weight. In addition, increased K application typically leads to decreased Sr concentrations in the leaves. K application increased ExK and decreased ExCs, ammonium, and nitrate concentrations in the soil but did not affect ExSr, ExCa, and ExMg. Our findings imply that increasing ExK concentration in the soil suppressed only ExCs and not ExSr concentration, but it might have affected Sr re-translocation into plant tissues. Though we have hypothesized that Sr uptake was also reduced by the application of K fertilizer, however the results did not support the idea, on the other hand, re-translocation of Sr in the plant was negatively affected by K concentration. Research on the impact of the exclusive use of K fertilizer on Sr translocation and the involvement of other related mineral elements is required.

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