




Research Article

Distribution of cesium and cationic mineral elements in napiergrass



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Abstract

Napiergrass is fast-growing perennial known for its high potential for accumulation of cesium (Cs). Cs is highly mobile within a plant and can be distributed to various plant organs. Here, we investigated the distribution of cesium-133 (¹³³Cs) and competitively translocated cationic minerals, such as potassium (K), calcium (Ca), and magnesium (Mg), in different organs of napiergrass. Treatments comprised four concentrations of ¹³³Cs applied to soil: 0 (as control); 300; 500; and 1000 μM. Leaf blades contained significantly higher concentrations of ¹³³Cs than stems under 300 and 500 μM ¹³³Cs treatments ($P < 0.01$). Specifically, significantly greater ¹³³Cs content was measured in younger parts of stems and leaf blades compared with mature or older plant parts. The ¹³³Cs content in younger parts was 5302, 13,059, and 51,678 mg kg⁻¹ in stems and 6961, 16,363, and 52,781 mg kg⁻¹ in leaf blades under 300, 500, and 1000 μM ¹³³Cs treatments, respectively. Distribution ratios of K were higher in stems than in leaf blades in all ¹³³Cs-treated conditions ($P < 0.05$). A significantly negative correlation was found between K and Ca or Mg in leaf blades, suggesting that ¹³³Cs and K are similarly competitive with Ca or Mg within napiergrass. We conclude that ¹³³Cs is distributed to younger plant parts, especially leaf blades, and that translocation of Ca and Mg is strongly inhibited by the presence of ¹³³Cs or K within organs. This suggests that ¹³³Cs or K can inhibit Mg translocation and could lead to Mg deficiency in younger plant parts.

Keywords Competition · ¹³³Cs · Macronutrients · *Pennisetum purpureum* Schum · Plant organ · Translocation

1 Introduction

Many areas surrounding the Fukushima Daiichi Nuclear Power Plant in Fukushima Prefecture, Japan, remain highly contaminated with long-lived radiocesium (Cs) since the 2011 tsunami, in particular ¹³⁷Cs, which has a half-life of 30 years [12]. ¹³⁷Cs is one of the most dangerous radionuclides; it is highly water soluble and has a marked tendency to accumulate in sediment and aquatic organisms [2, 27].

Cs and potassium (K) are both Group I alkali metals [9], but the higher atomic weight and ionic radius and lower hydration energy of Cs result in slightly different behaviors [2]. It has been suggested that competitive and inhibitory interactions between ¹³⁷Cs and K play an important role

in the uptake and translocation of alkaline and alkaline-earth metals by plant roots [19]. One possible reason for this inhibition could be that small amounts of ¹³⁷Cs block K⁺-channels [27]. Low concentrations of ¹³⁴Cs were shown to be easily taken up by plant roots and translocated to aboveground plant parts [17]. ¹³⁴Cs translocation and accumulation vary by plant species and plant organs [20]. Because Cs is mobile and can be easily distributed, its distribution within plants or within organs also varies among different plant species [2]. Examples include uptake via the vascular system of stems and leaves in *Arabidopsis thaliana* [9], seeds in *Vicia faba* [15], growing parts such as fruits, leaves, twigs, and bark in woody trees [1, 18], new leaves in cedar plants [16], leaves in Indian mustard [22], and leaves in *Chengiopanax sciadophylloides* [24].

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The uptake of nutrient elements by roots is also influenced by the presence of Cs. Cs is transported from the soil solution to the plant by various cation transporters located in the plasma membrane of root cells [25, 27]. The concentration of K in soil can influence Cs uptake, with greater uptake occurring as K concentrations increase, suggesting that K channels are blocked, potentially affecting mineral nutrition [21]. Therefore, it is important to consider the nutrient status of the soil [2], because interactions with the analogs K and calcium (Ca) affect ^{137}Cs transfer [6]. Additionally, it is well established that high concentrations of K in soil competitively inhibit magnesium (Mg) uptake, and that even within plants, excess K competes with Mg to reduce protein synthesis function [8].

Napiergrass (*Pennisetum purpureum* Schum) produces the greatest shoot-mass of all herbaceous plants [5], and it has a relatively high ^{137}Cs removal ratio (CR) among species studied for phytoremediation [10, 11]. In a previous field study, a maximum CR of 0.57% was achieved in napiergrass planted at high density on soils contaminated with high levels of ^{137}Cs (3404 kBq m^{-2}) [12]. We also previously confirmed that ^{133}Cs and ^{137}Cs became more localized in leaf blades of napiergrass compared with stems [10, 11]. However, the distribution of ^{133}Cs within napiergrass stems or leaf blades after translocation has not been fully elucidated. We anticipated that ^{133}Cs distribution would vary between leaf blades and stems and may in fact also be quite different depending on organ maturity. Therefore, in the present study, we hypothesized that ^{133}Cs content would be highest in the leaf blades of napiergrass following translocation, and that ^{133}Cs would be primarily distributed in the younger parts of stems and leaf blade organs. We further hypothesized that alkaline cationic minerals, such as K, Ca, and Mg, would be competitively distributed in the presence of ^{133}Cs within plant organs.

2 Materials and methods

We used a common variety of napiergrass (var. Merkeron), which showed high ^{137}Cs accumulation compared with other napiergrass varieties [11]. The experiment was conducted in a rainout greenhouse in Goshogawara, Aomori Prefecture (40.5494 N, 140.2743 E), northern Japan, between May 25 and August 21, 2017 (88 days). In this study, five pots (replicates) per each ^{133}Cs treatment concentration were used. Four-week-old nursery plants were transplanted into 1/2000a Wagner pots filled with 7 kg dried commercial soil on May 25, 2017. A compound fertilizer, N–P–K (15–15–15), was applied as a basal dressing at 4.0 g pot^{-1} , and the same amount was applied as a top dressing once per month (total two times) until harvest. The maximum and minimum temperatures in the greenhouse were measured using a data logger (Temperature and Humidity USB Datalogger DL171, AS ONE Co. Ltd., Osaka, Japan) throughout the experiment (Fig. 1).

Cs (atomic weight ^{133}Cs) at concentrations of 0 (control), 300, 500, and 1000 μM , as cesium chloride (CsCl) in 2 L water, was applied to the soil in each pot prior to transplanting. Pots were watered by applying 1050 mL of tap water when soil tension reached approximately -20 to -30 kPa . The total water applied equaled 15% of the total soil volume during the growth period. Soil tension in the pots was continuously assessed at a depth of 20 cm using a tension meter (DIK-8333, Daikirika Co. Ltd.) until harvest.

Before being harvested, five individual plants were measured for plant height, tiller number, and SPAD value (SPAD-502; Minolta Co., Ltd., Osaka, Japan); the SPAD value indicates chlorophyll content or color of a leaf. These plants were harvested, on the same day, from each ^{133}Cs -treatment group to determine the ^{133}Cs content in each part of the leaf blade and stem (including the leaf sheath).

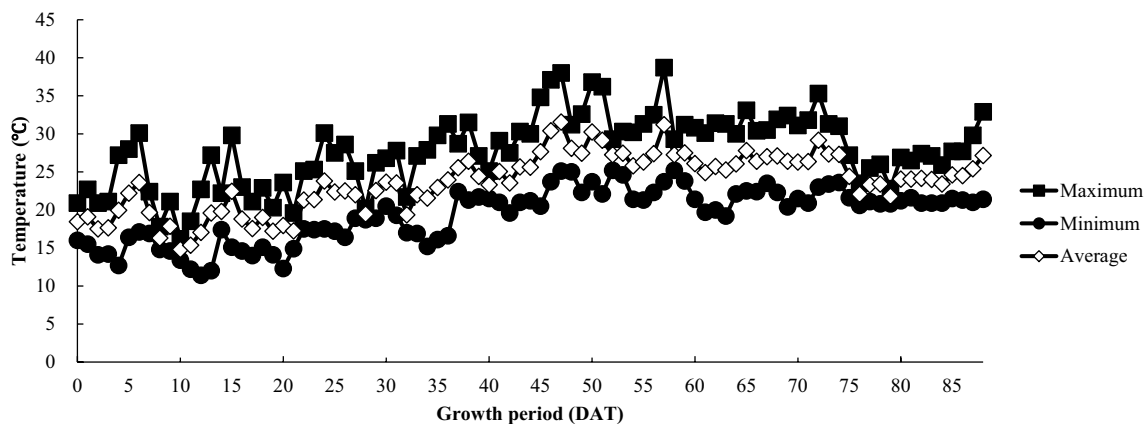


Fig. 1 Daytime air temperature during the plant growth period. DAT = days after transplanting

The harvested leaf blades and stems were further separated into young, mature, and old sections (Fig. 2), and then the plant materials were dried in an oven at 80 °C for 72 h. To determine ¹³³Cs concentration within each plant, 0.2 g each of dried leaf blades and stems were digested in 10 mL HNO₃ (nitric acid) using a Milestone microwave digestion system (ETHOS; Milestone Inc., Sorisole, Italy). After cooling, the samples were centrifuged and supernatants were passed through a 0.45-µm filter. ¹³³Cs concentrations were measured using an inductively coupled plasma-mass spectrophotometer (ICP-MS; PerkinElmer Elan, Co., Ltd., Fremont, CA, USA) according to a protocol by Kang et al. [10].

2.1 Statistical analysis

Five plant replicates in the four ¹³³Cs treatment groups were measured for plant height, tiller number, SPAD value, and ¹³³Cs content within both leaf blades and stems. The data obtained in the experiments were analyzed using Tukey’s multiple-range test to determine the significance of any differences between the mean values, using KaleidaGraph (ver. 4.1, Synergy Software) software. Significant differences in ¹³³Cs content between mean values in leaf blades and stems were determined using Fisher’s least significant difference test.

3 Results and discussion

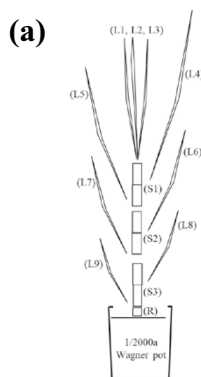
We investigated where ¹³³Cs is specifically distributed in the aboveground parts of napiergrass and also examined cationic mineral distribution within plant organs in the presence of ¹³³Cs under a range of soil treatment concentrations. To assess the toxic effect of Cs on napiergrass, we measured various morphological and physiological stress parameters, including plant height, SPAD values, and tiller number. In our study, no significant differences were observed in plant height, SPAD values, or tiller number of napiergrass with increasing ¹³³Cs concentration in soil (Table 1). For the other plants investigated, chlorophyll

Table 1 Effect of different levels of cesium-133 (¹³³Cs) treatment on plant height, SPAD value (SPAD), and tiller number

Treatment	Plant height (cm)	SPAD	Tiller number (plant ⁻¹)
0 µM ¹³³ Cs	119.2 ± 2.4ns	31.4 ± 0.8ns	19.4 ± 2.0ns
300 µM ¹³³ Cs	115.9 ± 2.7ns	32.9 ± 0.7ns	15.6 ± 0.7ns
500 µM ¹³³ Cs	111.0 ± 0.8ns	31.4 ± 0.4ns	19.6 ± 1.1ns
1000 µM ¹³³ Cs	111.7 ± 2.5ns	30.9 ± 0.3ns	19.2 ± 1.5ns

ns indicates no significant difference at the 5% level by Tukey’s multiple-range test

Values indicate the mean ± standard error (n=5 individual plants)



(b)

Label	Organ	Maturity	Corresponding Part	Description
ST1	Stem	Young	S1	Upper part (from top to 10 cm below, including leaf sheath)
ST2	Stem	Mature	S2	Middle part (from top between 10 cm and 20 cm, including leaf sheath)
ST3	Stem	Old	S3	Lower part (from top between 20 cm and 30 cm, including leaf sheath)
LB1	Leaf blade	Young	L1, L2, L3	Partly expanded uppermost leaves
LB2	Leaf blade	Mature	L4, L5, L6	Fully expanded mature leaves
LB3	Leaf blade	Old	L7, L8, L9	Old leaves, including dried leaves

(R); The lowest part of the stem was not collected.

Fig. 2 Schematic diagram showing the parts of organ material collected from napiergrass and used in the present study (a) and their descriptions (b)

content and several chlorophyll fluorescence parameters in *Plantago major* (2–20 mM ¹³³Cs) [4], *Arabidopsis halleri* (0.2–20 mM ¹³³Cs) [3], and *Brassica juncea* (25–100 mg L⁻¹ ¹³³Cs) [26] significantly decreased under Cs exposure during hydroponic growth; the dry weight of leaves, however, did not differ significantly compared with control plants [3, 4, 26]. Previously, we observed that napiergrass exhibited reduced plant height and SPAD value and increased tiller number at relatively high ¹³³Cs concentrations (1000–3000 μM) in hydroponic culture solution [10]. However, the results with regard to plant height, SPAD value, and tiller number at a concentration of 1000 μM ¹³³Cs differed in the present study compared with the previous study [10]. We considered that this difference might have arisen due to the different cultivation conditions between soil and hydroponic media, therefore further work is necessary to explore these growth differences.

The content of ¹³³Cs in the aboveground parts of napiergrass significantly increased with increasing ¹³³Cs concentrations in soil (Table 2). The mean ¹³³Cs content within leaf blades was significantly higher than in stems following treatment with 300 and 500 μM ¹³³Cs. As with our study, a trend of ¹³³Cs accumulation (leaf > stem) in a hydroponic experiment was also observed for leaves of *Brassica juncea* and *Vicia faba* under conditions of 25 and 50 mg kg⁻¹ Cs [7], *Plantago major* under 0.002 to 20 mM Cs [4], and *Calla palustris* under 0.5 and 1 mM Cs [14]. Similar Cs accumulation in leaf blades was also observed in napiergrass grown on ¹³⁷Cs-contaminated soil, where ¹³⁷Cs accumulation was approximately two to three times greater in leaf blades of napiergrass than in the stems [11]. On the other hand, in the present study, significant differences in ¹³³Cs content between leaf blades and stems disappeared at relatively high concentrations of ¹³³Cs

treatment (1000 μM). Similar results were observed in a hydroponic experiment, with differences between the ¹³³Cs content of leaf blades and sheaths (including stems) of napiergrass diminishing at 7 weeks after transplanting under 1000 and 3000 μM ¹³³Cs-treated hydroponic solution [10]. Here, we found a significantly greater distribution of ¹³³Cs within the younger parts of stems and leaf blades (ST1 and LB1) of napiergrass than within the older parts (ST3 and LB3) (*P* < 0.05). This trend of higher ¹³³Cs distribution in younger plant parts was observed across all ¹³³Cs treatments and represents a new finding in napiergrass. Concerning ¹³³Cs, however, small differences in the distribution of ¹³³Cs were observed between the stems (ST1) and leaf blades (LB1) of younger parts at 1000 μM ¹³³Cs compared with 500 μM ¹³³Cs treatment (Table 2). In particular, the ¹³³Cs content of leaf blades (LB2) in mature organs under 1000 μM ¹³³Cs was lower than in stems (ST2). Lai et al. [15] observed various growth stages of *Vicia faba* under soil conditions with high concentrations of ¹³³Cs (25, 50, and 100 mg kg⁻¹) and determined that the pattern of ¹³³Cs accumulation and redistribution in plants depends on the plant growth time and the ¹³³Cs concentration. Therefore, we considered that napiergrass, although it has a large capacity for Cs accumulation, may have exhibited limited translocation from stem to leaf blades under excess ¹³³Cs exposure in this study.

Table 3 shows the distribution ratio of cationic mineral elements within napiergrass in the presence of ¹³³Cs under different concentrations of ¹³³Cs treatment. The distribution ratio of cationic minerals was affected by plant organ as well as maturity. Potassium (K), which behaves similar to ¹³³Cs, was easily translocated and accumulated within plants when the ¹³³Cs treatment concentration was relatively low, at 300 and 500 μM. However, K translocation was

Table 2 Cesium-133 (¹³³Cs) content in the stems (ST) and leaf blades (LB) at different levels of ¹³³Cs-treated soil conditions

Label	300 μM ¹³³ Cs	500 μM ¹³³ Cs	1000 μM ¹³³ Cs
ST1	5301.7 ± 357.4a	13,059.1 ± 584.7a	51,677.6 ± 2214.2a
ST2	4951.5 ± 333.8ab	12,387.5 ± 980.1a	45,003.2 ± 2994.4a
ST3	3868.5 ± 220.6b	8997.0 ± 486.9b	28,513.5 ± 1686.9b
Mean (stem)	4707.2 ± 232.4	11,481.2 ± 610.2	41,731.5 ± 2891.7ns
LB1	6960.7 ± 623.0a	16,363.3 ± 1189.0a	52,780.6 ± 689.4a
LB2	5333.1 ± 172.0b	13,746.6 ± 716.4ab	38,844.7 ± 1193.3b
LB3	5048.4 ± 164.6b	12,562.7 ± 597.3b	33,780.4 ± 718.4c
Mean (leaf blade)	5780.7 ± 305.1**	14,224.2 ± 630.6**	41,801.9 ± 2200.0ns

ST1, ST2, ST3 and LB1, LB2, LB3 indicate young, mature, and old organs in stems and leaf blades, respectively

Values indicate the mean ± standard error (*n* = 5 individual plants)

The same letters indicate no significant difference within the stem or leaf blade at the 5% level according to Tukey’s multiple-range test

ns indicates no significant difference at the 5% level

** indicates significant correlations between the means of stems and leaf blades at the 1% level by Fisher’s least significant difference test

Table 3 Distribution ratio of cationic minerals compared with cesium-133 (^{133}Cs) in each part of the stems (ST) and leaf blades (LB) grown in different ^{133}Cs treatment concentrations

Treatment	Label	K/ ^{133}Cs	Ca/ ^{133}Cs	Mg/ ^{133}Cs
300 μM ^{133}Cs	ST1	6.74 ± 0.30b	0.20 ± 0.01c	0.36 ± 0.02bc
	ST2	9.42 ± 0.32a	0.26 ± 0.02bc	0.85 ± 0.06a
	ST3	8.39 ± 0.20ab	0.22 ± 0.01bc	0.89 ± 0.06a
	LB1	3.48 ± 0.21c	0.17 ± 0.02c	0.15 ± 0.01c
	LB2	3.74 ± 0.09c	0.43 ± 0.02b	0.25 ± 0.02bc
	LB3	3.33 ± 0.09c	0.90 ± 0.06a	0.50 ± 0.04b
500 μM ^{133}Cs	ST1	2.80 ± 0.08b	0.08 ± 0.00c	0.14 ± 0.00bc
	ST2	3.51 ± 0.10a	0.11 ± 0.01bc	0.34 ± 0.02a
	ST3	3.12 ± 0.14ab	0.10 ± 0.00bc	0.34 ± 0.01a
	LB1	1.55 ± 0.06c	0.06 ± 0.00c	0.06 ± 0.00d
	LB2	1.48 ± 0.05c	0.15 ± 0.00b	0.08 ± 0.00cd
	LB3	1.44 ± 0.06c	0.31 ± 0.02a	0.16 ± 0.01b
1000 μM ^{133}Cs	ST1	0.73 ± 0.01b	0.02 ± 0.00c	0.04 ± 0.00d
	ST2	1.01 ± 0.02a	0.04 ± 0.00b	0.10 ± 0.00b
	ST3	0.85 ± 0.03b	0.04 ± 0.00b	0.13 ± 0.00a
	LB1	0.48 ± 0.00c	0.02 ± 0.00c	0.02 ± 0.00e
	LB2	0.57 ± 0.01c	0.05 ± 0.00b	0.02 ± 0.00de
	LB3	0.56 ± 0.01c	0.13 ± 0.00a	0.06 ± 0.00c

ST1, ST2, ST3 and LB1, LB2, LB3 indicate young, mature, and old organs in stems and leaf blades, respectively

Values indicate the mean ± standard error ($n=5$ individual plants)

The same letters indicate no significant difference within the stems or leaf blades at the 5% level according to Tukey's multiple-range test

Distribution ratio calculated as cationic mineral content divided by ^{133}Cs content in each part

K potassium; Ca calcium; Mg magnesium

heavily suppressed at ^{133}Cs concentrations of 1000 μM . It is well established that the transport of Cs from the soil solution into a plant usually happens via K and Ca transporters [25, 27]. Komínková et al. [14] reported a study in which *Calla palustris* plants were exposed to ^{133}Cs -treated hydroponic solutions (0.5 and 1 mM ^{133}Cs) and several concentrations of K (0.5, 1, 2, 5, and 10 mM K); they found that ^{133}Cs uptake and translocation were affected not only by the external concentration of K but also by the external concentration of ^{133}Cs . Similarly, Burger and Lichtscheidl [2] stated that a low concentration of K in plants increased their uptake of Cs and that higher concentrations led to reduced Cs uptake. On the other hand, K uptake is also influenced by ^{133}Cs content in the soil. In the present study, the K distribution ratio was consistently higher in stems than in leaf blades in all ^{133}Cs treatment groups. This is contrary to the ^{133}Cs distribution measured within plant organs (Table 2) and suggests that ^{133}Cs and K, which have chemically similar behavior, inhibit each other within napiergrass. It is well established that Cs (^{133}Cs and ^{137}Cs)

ions are chemically similar to K ions under several growth conditions [1, 9]. In the present study, Ca content in plant organs was greatly suppressed by the presence of ^{133}Cs at all treatment levels (Table 3). Ca distribution ratios (Ca/ ^{133}Cs) decreased with increasing ^{133}Cs concentrations, because the Cs uptake competitively into plants using Ca and K transporters in the plasma membrane [2, 25]. The distribution ratio of Ca in leaf blades was significantly higher in older leaf blades compared with younger ones (Table 3, Ca/ ^{133}Cs). A similar trend in distribution was also observed in the stems, because there is almost no redistribution of Ca within the plant following root uptake [13], resulting in lower Ca distribution ratios in newly emerged stems and leaf blades (younger plant parts). In contrast, Ca accumulation in the leaves of *Plantago major* significantly increased in the 2 mM ^{133}Cs -treated hydroponic condition [4]. Concerning the accumulation of Ca in the presence of ^{133}Cs in hydroponic culture medium, Burger et al. [4] concluded that the presence of ^{133}Cs in the medium did not decrease Ca uptake; decreased biomass may therefore possibly be related to a K deficiency. As with Ca, the distribution of Mg in the presence of ^{133}Cs was also greatly suppressed, but, in contrast, Mg was found to be significantly more likely to be distributed among stems rather than leaf blades (Table 3, Mg/ ^{133}Cs). Within the stems, Mg was localized primarily in the more mature or older parts than in younger parts. Similar to ^{133}Cs , Mg uptake is competitively inhibited by large amounts of K, and excess K competes with Mg resulting in reduced protein synthesis [8]. Considering that Cs uptake occurs via K and Ca channels [25], we suggest ^{133}Cs has similar behavior to K within plants, which also affects the translocation or distribution of cationic minerals.

Different levels of ^{133}Cs concentration in soil led to significant correlations between cationic minerals (Table 4). A significantly negative correlation between K and Ca ($P < 0.001$) and K and Mg ($P < 0.01$) was observed in the leaf blades compared with the stems. This could be explained by the chemical similarity between ^{133}Cs and K, and competition between K and Mg or Ca translocation within plant organs. From this, in the present study, we considered that ^{133}Cs or K, which are effectively redistributed within plants, have similar competitive behaviors toward Ca or Mg in napiergrass. Conversely, no competition was observed between Ca and Mg within napiergrass (Table 4, Ca vs. Mg). Karley and White [13] suggested that this lack of competition was due to phylogenetic constraints or the control of Ca and Mg concentrations in tissues. Similarly, Smolders et al. [23] observed the uptake of ^{137}Cs in spinach under 15 different nutrient solutions containing ^{137}Cs and showed ^{137}Cs levels were significantly reduced, approximately threefold, by increasing Ca and Mg concentrations. These divalent cations compete with Cs uptake through

Table 4 Effect of different concentrations of cesium-133 (^{133}Cs) treatment on the correlation coefficient between cations in stems (ST) and leaf blades (LB)

Treatment	K versus Ca		K versus Mg		Ca versus Mg	
	ST	LB	ST	LB	ST	LB
300 μM ^{133}Cs	0.7701***	-0.8185***	0.5290*	-0.7176**	0.4785	0.9786***
500 μM ^{133}Cs	0.9211***	-0.7998***	0.3762	-0.7267**	0.5369*	0.9768***
1000 μM ^{133}Cs	0.8193***	-0.8134***	0.1960	-0.6883**	0.6898**	0.9548***

K potassium; Ca calcium; Mg magnesium

*, **, and *** indicate significant correlations at the 5%, 1%, and 0.1% levels

competition in the apoplast of the root cortex [23]. With regard to the competition between cationic minerals within napiergrass, we suggest that high levels of ^{133}Cs or K translocation will inhibit not only Ca but also Mg translocation and lead to Mg deficiency.

4 Conclusion

We investigated the distribution of cesium-133 (^{133}Cs) and competitively translocated cation minerals, such as potassium (K), calcium (Ca), and magnesium (Mg), in different organs of napiergrass under ^{133}Cs -treated conditions. The results of our experiment showed that: (1) ^{133}Cs content was significantly higher in leaf blades than in stems, (2) ^{133}Cs was principally distributed throughout the younger parts of the stems or leaf blades, and (3) translocation of Ca and Mg, particularly Mg, was strongly inhibited by the presence of ^{133}Cs or K within plant organs.

Our results suggest that large amounts of ^{133}Cs or K translocation could lead to nutrient imbalance, especially Mg deficiency, in younger plant organs of napiergrass. Further studies are necessary to verify the competition between ^{133}Cs and cationic minerals, particularly Mg, under relatively low ^{133}Cs concentrations, such as 300 or 500 μM , applied with different levels of K fertilizer.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

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