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RESEARCH ARTICLE

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Influence of Ectomycorrhizal Colonization on Cesium Uptake by *Pinus densiflora* Seedlings

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ABSTRACT

Radionuclides were deposited at forest areas in eastern parts of Japan following the Fukushima Daiichi Nuclear Power Plant incident in March 2011. Ectomycorrhizal (EM) fungi have important effects on radiocaesium dynamics in forest ecosystems. We examined the effect of colonization by the EM fungus *Astraeus hygrometricus* on the uptake of cesium (Cs) and potassium (K) by *Pinus densiflora* seedlings. Pine seedlings exhibited enhanced growth after the EM formation due to the colonization by *A. hygrometricus*. Additionally, the shoot Cs concentration increased after the EM formation when Cs was not added to the medium. This suggests that *A. hygrometricus* might be able to solubilize Cs fixed to soil particles. Moreover, the shoot K concentration increased significantly after the EM formation when Cs was added. However, there were no significant differences in the root K concentration between EM and non-EM seedlings. These results suggest that different mechanisms control the transfer of Cs and K from the root to the shoot of pine seedlings.

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KEYWORDS
Astraeus; mycorrhizal synthesis; pine; potassium; stable Cs

1. Introduction

High concentrations of radionuclides were released into the environment because of the accident at the Fukushima Dai-ichi Nuclear Power Plant in Japan in 2011. The contaminated area is covered mainly by forests. Therefore, radiocesium was initially deposited on the forest canopies and then the forest floor because of rainfall and accumulation of plant litter [1]. Radiocesium was released from decomposing plant debris into the mineral soil layer, where cesium (Cs) may be absorbed by plant roots or fixed in soil particles. Because of the long half-life of ^{137}Cs, contamination by radiocesium is expected to persist for decades [1]. In Europe, forests were contaminated by radionuclides after the accident at the Chernobyl Nuclear Power Plant in 1986. However, migration of radionuclides into the mineral soil layer was faster in Fukushima than in Chernobyl because of faster litter decomposition due to higher temperatures and more precipitation [1].

The accident in Chernobyl increased ^{137}Cs and ^{134}Cs concentrations in a variety of mushrooms in Europe [2]. After the 2011 accident in Fukushima, the radiocesium content in fungal fruiting bodies was higher in samples collected near the nuclear power plant than in samples collected further away [3]. Following both accidents fruiting bodies of ectomycorrhizal (EM) species generally accumulated more ^{137}Cs than those of saprotrophic species, although the uptake of radiocesium varied among the examined fungal species [2,3].

Ectomycorrhizal fungal species form symbiotic associations with some woody plants and develop extensive nets of vegetative hyphae to collect minerals directly from soil [2]. Thus, EM fungi are considered to have important effects on radiocesium dynamics in forest ecosystems [4]. The ability of EM fungi to absorb Cs from the substrate has been investigated in studies [5,6] in which ^{137}Cs and stable Cs were used because of their similar chemical behaviors. This suggests that stable Cs may be useful for predicting radiocesium transport [7]. We previously observed *Suillus*, *Pisolithus*, and *Rhizopogon* absorbed more Cs than other EM and saprotrophic species when grown on NH_4-containing medium while *Astraeus* and *Scleroderma* species did more Cs than other species grown on NO_3-containing medium. These EM species occur in soils with a poorly developed humus [8] and exhibit a high potential for absorbing soil minerals because of the production of organic acids [9]. The ability of EM fungi to absorb radionuclides from soil may affect the radionuclide accumulation in their host plants. An earlier report indicated that EM pine seedlings may accumulate 3–5 times more ^{90}Sr from the

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contaminated soil than seedlings without ectomycorrhizae [10]. In contrast, another study revealed that EM colonization lowers the ^{134}Cs concentration in spruce seedlings [4]. Moreover, mycorrhizal heather plants were observed to translocate a relatively large proportion of Cs from a liquid medium into the shoot, although the total Cs content in the mycorrhizal plants was lower than that of non-mycorrhizal plants [11]. Consequently, it is unclear whether the radionuclides in the mycorrhizal fungal biomass is preferentially translocated into the host plants, and more research is necessary to clarify the effects of EM symbiosis on the accumulation of radionuclides in plants.

Astraeus hygrometricus, which forms an EM association and is common in Japan, Korea, and northeastern China [12], accumulates relatively large amounts of Cs, especially when NO_3 is provided as an N source in the medium [6]. In a preliminary study, we examined the accumulation of stable Cs in the shoot of *Pinus densiflora* colonized by *A. hygrometricus*, but the accumulation of potassium (K) in the plants was not examined [13]. Similar to Cs, K is a monovalent cation and is important for all organisms. Because of the similarity in the physical and chemical properties of K and Cs, it is likely that Cs accumulates in cells through the K-transport system [14]. An earlier study concluded that Cs accumulation is affected by K in environment [14]. In the present study, we examined the effect of an EM fungus on the uptake of Cs by its host plant in a pot experiment using *P. densiflora* seedlings and an *A. hygrometricus* isolate. Also, the K content in seedlings was measured, and Cs uptake by mycorrhizal plants was examined in terms of K uptake.

2. Materials and methods

2.1. Plant preparation

Pinus densiflora seeds (lot number: 03-124) were obtained from a seed stock of the Forestry and Forest Products Research Institute (FFPRI), Tsukuba, Japan. Seeds were immersed in running water for one night and then surface sterilized in 30% H_2O_2 for 15 min. They were then placed on 0.85% agar and incubated at ~23 °C under light (photon flux density of about 65 µmol m^{-2} s^{-1}) provided by cool white fluorescent lamps for a 16-h light/8-h dark cycle. Germinated seeds were transferred to plastic bottles prepared as follows. A substrate comprising 400 mL pumice (Hyuga-tsuchi; particle diameter: 3–6 mm) and 160 mL nutrient solution was added to plastic bottles, which were then autoclaved at 121 °C for 30 min [15]. The contents of total and exchangeable Cs and K in the pumice were measured using an inductively coupled plasma mass spectrometer (ICP-MS, 7700a, Agilent Technologies, Tokyo, Japan) after digestion with HNO_3 for the total content or after extraction with 1 N ammonium acetate for exchangeable contents (Table 1). We used four nutrient solutions in the media to investigate the effect of the culture medium on mycorrhizal synthesis. Three of the solutions have been used for culturing fungi [modified Melin-Norkrans liquid medium (MMN) [16], Kawai-Ogawa liquid medium (KO) [17], and Ohta liquid medium (OT) [18]]. The fourth nutrient solution was Hyponex liquid medium (HYPO; N:P:K = 6:10:5; Hyponex Japan Corp., Hyogo, Japan) (Table 2). Distilled water (DW) was used as a control. Sugars were removed from MMN, KO, and OT to eliminate the possibility of the synthetic medium might induce a host defense response against a normally compatible fungal symbiont [19]. The number of bottles prepared for each treatment is indicated in Table 2. After transferring pine seedlings into bottles, they were incubated in the laboratory at 23 °C under light (photon flux density of 60 µmol m^{-2} s^{-1} at the top of the bottles) for a 16-h light/8-h dark cycle.

Table 1. Total and exchangeable Cs and K concentrations in the pumice soil.

Total Cs (µg/g)	Total K (mg/g)	Exchangeable Cs cation (µg/g)	Exchangeable K cation (mg/g)
0.19 ± 0.01*	15.7 ± 0.1	0.009 ± 0.000	0.081 ± 0.003

*Values represent means ± standard errors calculated for three replicates.

Table 2. Number of replicates for treatments.

Nutrient solution	Mycorrhizal inoculation	Cs addition	Number of replicates
HYPO	+	+	3
	+	−	3
	−	+	3
	−	−	2
OT	+	+	3
	+	−	2
	−	+	2
	−	−	2
MMN	+	+	3
	+	−	3
	−	+	3
	−	−	3
KO	+	+	3
	+	−	3
	−	+	3
	−	−	2
DW	+	+	2
	+	−	3
	−	+	3
	−	−	3

HYPO: Hyponex liquid medium; OT: Ohta liquid medium; MMN: Modified Melin-Norkrans liquid medium; KO: Kawai-Ogawa liquid medium; DW: distilled water.

2.2. Mycorrhizal synthesis

An *A. hygrometricus* isolate (FFPRI 460507) [6] was obtained from a fruiting body collected from the

Table 3. *F* values and significance levels for the ANOVA of growth parameters.

		Total plant dry weight		Shoot dry weight		Root dry weight		Root/shoot		Mycorrhization rate*	
Source of variation	*df*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*
Nutrient	4	2.720	.046	4.837	.003	1.551	.210	19.455	.000	4.951	.007
Mycorrhizal inoculation	1	7.114	.012	15.953	.000	0.023	.882	24.871	.000	–	–
Cs addition	1	0.739	.396	0.002	.964	4.466	.042	2.913	.097	0.102	.753
Nutrient × Inoculation	4	0.699	.598	1.024	.409	0.697	.599	1.491	.227	–	–
Nutrient × Cs	4	0.283	.887	0.752	.563	0.536	.710	0.884	.484	1.269	.318
Inoculation × Cs	1	0.155	.697	0.395	0534	0.002	.968	1.694	.202	–	–
Nutrient × Inoculation × Cs	4	0.711	.590	0.745	.568	0.715	.588	2.237	.086	–	–

*Mycorrhization rate = number of mycorrhizal root tips/total number of root tips.

Table 4. *F* values and significance levels for the ANOVA of the plant Cs content.

		Total plant				Shoot				Root			
		+Cs		−Cs		+Cs		−Cs		+Cs		−Cs	
Source of variation	*df*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*
Nutrient	4	1.806	.172	1.064	.406	1.891	.156	3.785	.024	7.259	.001	5.647	.005
Mycorrhizal inoculation	1	0.090	.768	7.721	.013	2.268	.149	16.486	.001	5.054	.037	1.347	.263
Nutrient × Inoculation	4	1.129	.374	1.158	.366	0.958	.454	1.078	.400	2.062	.128	0.705	.600

Table 5. *F* values and significance levels for the ANOVA of the plant Cs concentration.

		Total plant				Shoot				Root			
		+Cs		−Cs		+Cs		−Cs		+Cs		−Cs	
Source of variation	*df*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*
Nutrient	4	1.133	.373	0.142	.964	1.619	.213	0.809	.537	6.806	.002	3.694	.026
Mycorrhizal inoculation	1	0.911	.353	0.573	.460	0.857	.367	5.134	.038	4.813	.042	1.376	.258
Nutrient × Inoculation	4	0.651	.633	0.532	.714	1.089	.391	0.749	.573	1.304	.306	0.305	.870

evergreen oak forests in Kuma, Kumamoto Prefecture, Japan. These species appears under a variety of tree species, suggesting a wide host range and the strain used for the present study can form its ectomycorrhizae not only on oaks (*Quercus serrata* and *Q. phillyraeoides*) but on pine (*P. densiflora*) well in pot culture (data not shown). The *A. hygrometricus* inocula were prepared as follows. The isolate was cultured in 5 mL OT [18] in a 6.5-mL multi-well plate incubated at 23 °C in darkness for 4 weeks. One month after transferring seedlings to the bottles, the inocula were washed with sterile DW and then added to the substrate in the bottle. The bottles were then incubated under the same conditions as described in 2.1. Plant preparation subsection.

After seedlings were colonized by *A. hygrometricus*, 10 mL sterilized CsCl solution was added to the pumice for a final Cs concentration of 10 µg/g dry soil. The control samples were treated with 10 mL sterilized DW instead of CsCl. Two to three replicates were prepared for each treatment (Table 2).

2.3. Harvesting

Two months after the Cs-treatment, the plants were harvested and washed with DW. The root system was collected under a dissecting microscope using fine forceps to remove the pumice soil, and EM formation was characterized by the presence of fungal hyphae networks around short roots that formed an external mantle or sheath [12]. The numbers of EM and non-EM root tips were counted under a dissecting microscope, and mycorrhization rate (the percentage of EM root tips = the number of EM root tips/(the number of EM root tips + non-EM root tip) × 100) was calculated [20]. Samples were oven-dried at 60 °C for 24 h before measuring seedling dry weights. The root-to-shoot (R/S) ratio was calculated based on the shoot and root dry weights. The amounts of Cs and K in samples were measured using the ICP-MS as shown in 2.1. Plant preparation subsection.

All data underwent one- and two-way analyses of variance, while the Tukey–Kramer test was used for mean separations. The results of statistical analysis were shown at Tables 3–7, where *dF*, *F* and *p* mean number of degrees of freedom, *F* ratio, and *p* values, respectively, and *p* values less than .05 were considered statistically significant.

3. Results

3.1. Plant growth

Pine seedling shoots grew significantly better after the EM formation due to the colonization by

Table 6. *F* values and significance levels for the ANOVA of the plant K content.

		Total plant				Shoot				Root			
		+Cs		−Cs		+Cs		−Cs		+Cs		−Cs	
Source of variation	*df*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*
Nutrient	4	1.594	.219	0.816	.533	2.546	.075	2.041	.137	1.035	.416	2.747	.065
Mycorrhizal inoculation	1	15.12	.001	6.407	.022	16.35	.001	7.344	.015	1.149	.298	0.687	.419
Nutrient × Inoculation	4	1.350	.290	0.466	.760	2.517	.078	0.606	.664	0.928	.470	0.556	.698

Table 7. *F* values and significance levels for the ANOVA of the plant K concentration.

		Total plant				Shoot				Root			
		+Cs		−Cs		+Cs		−Cs		+Cs		−Cs	
Source of variation	*df*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*	*F*	*p*
Nutrient	4	1.123	.376	1.864	.166	2.052	.130	2.076	.132	0.900	.485	9.659	.000
Mycorrhizal inoculation	1	9.01	.008	3.895	.066	7.17	.015	0.15	.699	1.088	.311	0.201	.660
Nutrient × Inoculation	4	0.957	.455	1.133	.376	1.666	.202	0.712	.596	0.380	.820	2.335	.100

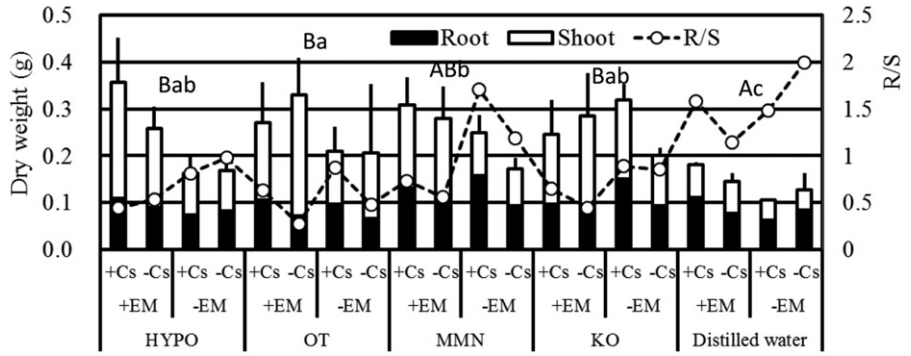

Figure 1. Growth of *Pinus densiflora* seedlings 10 months after being inoculated with *Astraeus hygrometricus*. Values are presented as the means with standard errors. The number of replicates is indicated in Table 2. +EM: seedlings inoculated with *A. hygrometricus*; −EM: seedlings not inoculated with *A. hygrometricus*; +Cs: cesium chloride (CsCl) solution was added to the pumice (10 μg Cs per gram of dry pumice); −Cs: distilled water (DW) was added to the pumice; HYPO: Hyponex liquid medium; OT: Ohta liquid medium; MMN: Modified Melin-Norkrans liquid medium; KO: Kawai-Ogawa liquid medium. Values with different letters (A–B for growth of the shoot; a–c for the R/S ratio) are significantly different at $p = .05$.

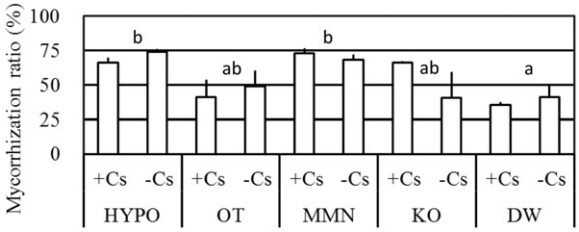

Figure 2. Mycorrhization rate of the *Pinus densiflora* seedlings 10 months after being inoculated with *Astraeus hygrometricus*. The mycorrhization rate (%) was calculated as the number of mycorrhizal root tips divided by the total number of root tips. Values are presented as the means with standard errors. Abbreviations are defined in Figure 1 legend. Values with different letters are significantly different.

A. hygrometricus ($p < .000$), while root growth did not improve (Table 3, Figure 1). Accordingly, the R/S ratio of EM seedlings was significantly lower than that of non-EM seedlings ($p < .000$; Table 3). Moreover, shoots of EM and non-EM seedlings supplemented with a nutrient solution grew significantly better than those supplemented with DW. Additionally, the mycorrhization rate of the seedlings treated with MMN or HYPO was significantly higher than that of the control seedlings treated with DW (Figure 2). Furthermore, addition of Cs at the concentration of 10 μg/g dry soil did not affect total seedling growth ($p = .396$) or the mycorrhization rate ($p = .753$) after the inoculation with *A. hygrometricus* (Table 3).

3.2. Accumulation of cesium

Seedling Cs contents (i.e., amount of Cs per plant) varied depending on the seedling part, EM formation, Cs addition, and nutrient solution type (Table 4, Figure 3(a,b)). The Cs content of EM seedlings was significantly higher than that of non-EM seedlings when Cs was not added ($p = .013$), while there was no significant difference in Cs content between EM seedlings and non-EM seedlings when Cs was added ($p = .768$). The shoot Cs content of EM seedlings was significantly higher than that of non-EM seedlings when Cs was not added ($p = .001$), while the root Cs content of EM seedlings was significantly lower than that of non-EM seedlings when Cs was added ($p = .037$). The shoot Cs content of seedlings treated with OT was higher

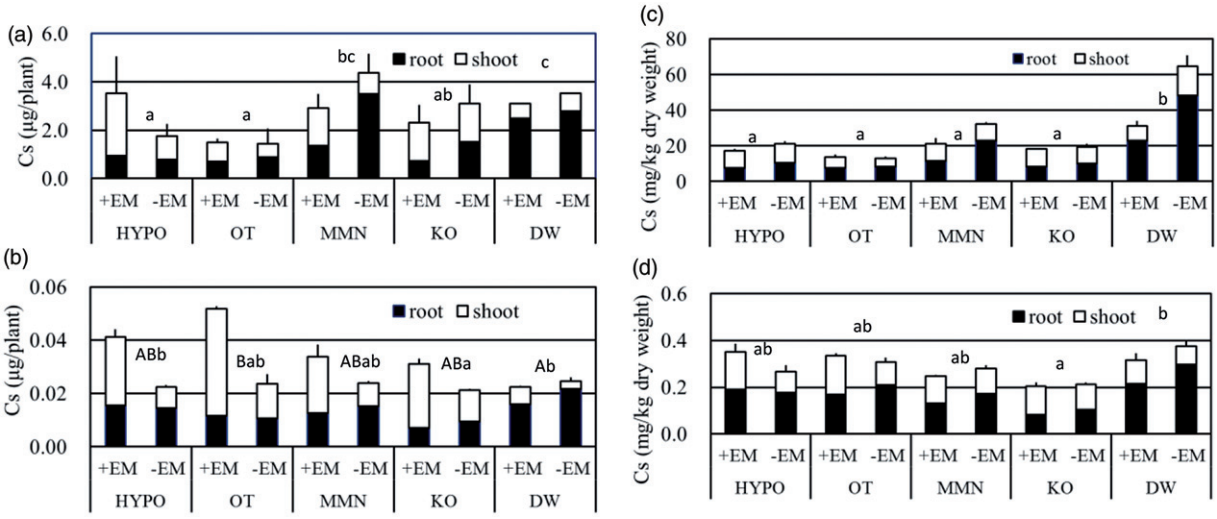

Figure 3. Cesium (Cs) content and concentration in the shoot and root of *Pinus densiflora* seedlings 10 months after being inoculated with *Astraeus hygrometricus*. (a) Cesium content in seedlings (amount of Cs per plant) treated with cesium chloride (CsCl) solution (10 μg Cs per gram of dry pumice). (b) Cesium content in seedlings treated with distilled water (DW). (c) Cesium concentration in seedlings (amount of Cs per gram of plant dry weight) treated with CsCl solution (10 μg Cs per gram of dry pumice). (d) Cesium concentration in seedlings treated with DW. Values are presented as the means with standard errors. Abbreviations are defined in Figure 1 legend. Values with different letters (A–B for the shoot; a–c for the root) are significantly different at $p = .05$.

than that of seedlings treated with DW when Cs was not added (Figure 3(b)). The root Cs content of seedlings supplemented with nutrient solutions was lower than that of the control seedlings with DW. The differences were significant between seedlings treated with KO and DW without the addition of Cs, and between seedlings treated with different nutrient solutions, except for MMN and DW with the addition of Cs.

The seedling Cs concentration (i.e., amount of Cs per gram of plant dry weight) also differed depending on the seedling part, EM formation, Cs addition, and nutrient solution type (Table 5, Figure 3(c,d)). The Cs concentration was higher in the root than in the shoot regardless of EM colonization and Cs addition. Additionally, the shoot Cs concentration of EM seedlings was significantly higher than that of non-EM seedlings when Cs was not added ($p = .038$). Meanwhile, the root Cs concentration of EM seedlings was significantly lower than that of non-EM seedlings when Cs was added ($p = .042$). The root Cs concentration of seedlings treated with a nutrient solution was significantly lower than that of control seedlings treated with DW when Cs was added. When Cs was not added, significant differences were observed only between seedlings treated with KO and DW.

3.3. Accumulation of potassium

The seedling K content (i.e., amount of K per plant) varied depending on the seedling part and EM formation (Figure 4(a,b)). The K content in the shoot or in the whole seedling increased after the EM formation (Table 6).

The seedling K concentration (i.e., amount of K per gram of plant dry weight) also differed depending on the seedling part, EM formation, Cs addition, and nutrient solution type (Table 7, Figure 4(c,d)). The K concentration in the shoot ($p = .015$) and in the whole seedling ($p = .008$) increased after the EM formation when Cs was added (Table 7). The root K concentration of seedlings supplemented with DW was significantly higher than that of seedlings treated with HYPO, MMN, or KO when Cs was not added (Figure 4(d)).

4. Discussion

4.1. Plant growth

The growth of the *P. densiflora* seedlings improved significantly after the EM formation due to the colonization by *A. hygrometricus* (Table 3, Figure 1). Additionally, the R/S ratio of the EM seedlings was significantly lower than that of the non-EM seedlings (Table 3, Figure 1). These results suggest that *A. hygrometricus* improved shoot growth, which is consistent with the results of a previous study [15]. Ectomycorrhizal fungi enhance the growth of colonized seedlings by increasing the uptake of nutrients and water [21]. Furthermore, some mycorrhizal fungi produce a hormonal substance affecting the growth of the associated plant [22].

Seedlings treated with a nutrient solution grew better and exhibited greater EM formation than control seedlings treated with DW, and the differences were significant for KO (regarding growth) and for HYPO and MMN (regarding EM formation)

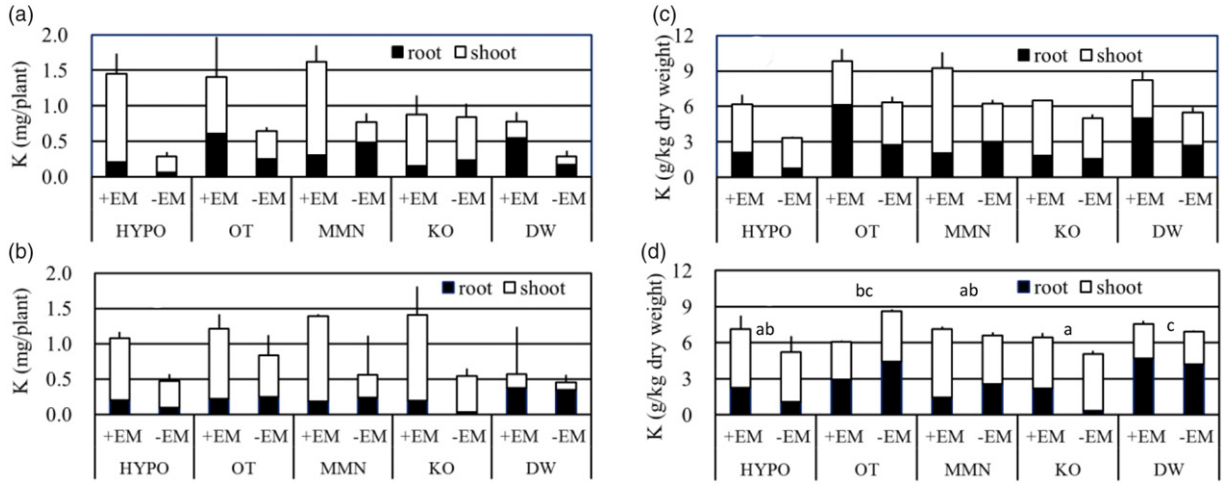

Figure 4. Potassium (K) content and concentration in the shoot and root of *Pinus densiflora* seedlings 10 months after being inoculated with *Astraeus hygrometricus*. (a) Potassium content in seedlings (amount of K per plant) treated with cesium chloride (CsCl) solution (10 μg Cs per gram of dry pumice). (b) Potassium content in seedlings treated with distilled water (DW). (c) Potassium concentration in seedlings (amount of K per gram of plant) treated with CsCl solution (10 μg Cs per gram of dry pumice). (d) Potassium concentration in seedlings treated with DW. Values are presented as the means with standard errors. The number of replicates is indicated in Table 2. Abbreviations are defined in Figure 1 legend. Values with different letters (a–b for the root) are significantly different at $p = .05$.

(Table 3, Figures 1 and 2). An earlier investigation concluded that EM formation with *Tricholoma matsutake* is influenced by the nutrient solution added to the soil [23]. Nutrient supplies, especially sugar, phosphate, and nitrogen, reportedly affect EM formation and the growth of EM plants. The addition of glucose to the synthesis medium was observed to induce a host-defense response against a normally compatible fungal symbiont [19]. In contrast, a glucose-containing substrate is necessary for the efficient colonization of *P. sylvestris* by the EM fungus *Lactarius deliciosus* under aseptic conditions [24]. Meanwhile, a previous study involving the screening of eucalyptus plants with 16 isolates of an EM fungus revealed that the phosphorus content of the substrate affects the growth and EM root length of the seedlings [21]. The mycorrhization of Norway spruce (*Picea abies*) reportedly decreases with increasing N concentrations [25]. In the present study, the EM formation in pine seedlings was not significantly different among the tested nutrient solutions (KO, HYPO, MMN, and OT). A previous report indicated that OT can induce the thickening of the Hartig net mycelium in the ectomycorrhizae formed on pine seedlings colonized by *T. matsutake* [26].

We also observed that the addition of Cs did not change the growth and EM formation of pine seedlings (Table 3). Similarly, the growth and mycorrhization of *Medicago truncatula* are not disrupted by the addition of stable Cs (0.15–15.6 μg g^{-1}) [27]. Moreover, Cs is not toxic to plants at natural environmental concentrations (i.e., below ~27 μg g^{-1}) [28]. In the present study, the 10 μg g^{-1} Cs added to the pumice is nontoxic to plants.

4.2. Accumulation of cesium

When Cs was not added, the Cs contents were significantly higher in the whole EM seedling and shoot than in the non-EM seedling and shoot (Table 4, Figure 3(b)). The pumice soil used in this experiment contained 0.2 μg g^{-1} Cs (Table 1), most of which was fixed in soil particles, making it difficult for the plant roots to absorb the Cs [14]. Therefore, the tested EM species may have released Cs from pumice, absorbed it, and transferred it to the colonized seedlings. Some EM fungal species solubilize minerals derived from soil particles [9]. Additionally, *A. hygrometricus* biomass can accumulate Cs [6]. The ability of EM fungi to solubilize minerals fixed in soil particles and accumulate the minerals may be important for the bioaccumulation of Cs by plants. *Rhizopogon roseolus*, which is an EM fungus, can increase the uptake of radiocesium by *P. pinaster* [29]. In contrast, the colonization of *Q. serrata* seedlings by the EM fungus *Cenococcum geophilum* reportedly decreases radiocesium uptake [30]. These results imply that the Cs uptake by an EM fungus and the transfer into the colonized plants depend on many factors, including environmental conditions during cultivation and the combination of fungi and host plants.

Nutrient conditions also influenced the accumulation of Cs in the seedling root (Tables 4 and 5, Figure 3). The root Cs level of seedlings supplemented with a nutrient solution was lower than that of control seedlings treated with DW. Cesium is a monovalent cation chemically similar to other elements such as Li^+, Na^+, K^+, and Rb^+. Sodium and K in the nutrient solutions used in this study should decrease the uptake of Cs from the soil by an EM

fungus and the colonized plant [28]. During the translocation of minerals absorbed by the root to the shoot, Cs is discriminated with other monovalent cations, especially K [14]. This tendency was more apparent when Cs concentration in the root was higher following the addition of Cs to the soil (Table 5, Figure 3(c)).

The Cs concentration was higher in the root than in the shoot (Figure 3(c,d)), which is consistent with the results of previous studies [29,30]. Plant roots are in direct contact with soil particles containing Cs, and a relatively small proportion of Cs is translocated to the shoots [30]. The distribution of Cs is altered by the colonization by an EM fungus. Significantly less ^{134}Cs accumulates in the needles of EM seedlings than in the needles of non-EM seedlings [4]. Moreover, the ^{137}Cs concentration tends to be lower in the leaves and stem than in the root, more so in EM plants than in non-EM plants [30]. In contrast, in the present study, the shoot Cs concentration increased in response to the colonization by the EM fungus when Cs was not added (Table 5, Figure 3(d)). However, the underlying mechanism has not been characterized.

We observed that the shoot K concentration increased significantly after the EM formation, but only when Cs was added (Table 7, Figure 4(c)). This may indicate that Cs improves the translocation of K from the EM root to the shoot. Cesium usually decreases the uptake of K from the soil because Cs and K share the same transport mechanisms [28]. In the present study, there were no significant differences in the root K concentration between the EM and non-EM seedlings (Table 7), which is similar to the root Cs concentration. These results suggest that the root K and Cs concentrations are mainly influenced by soil nutrient conditions.

The EM formation differentially affected the uptake of Cs and K from the soil and the subsequent transfer from the root to the shoot. The EM formation increased the K accumulation in the shoot when Cs was added, or had no effect when Cs was not added (Table 7). In contrast, the EM formation enhanced the Cs accumulation in the shoot when Cs was not added (Table 5), while it decreased the Cs accumulation in the root when Cs was added to the soil. These patterns may vary depending on the fungal species and the host plant. The ability of fungi to release Cs from the soil and absorb Cs is important for the translocation of Cs from the soil to the colonized plant. Additionally, EM fungal species should be screened regarding their ability to release, absorb, and translocate Cs to clarify the role of EM species during the transfer of Cs from the soil to colonized plants.

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Disclosure statement

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