



# Effects of NaCl Application on Cesium Accumulation in the Aboveground Parts of Quinoa (*Chenopodium quinoa* Willd.)

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Received: 7 July 2020 / Accepted: 4 November 2020 / Published online: 11 November 2020  
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**Abstract** In this study, we clarified the accumulation and concentration of Cs, Na, and K in each organ (leaves, stem, and panicle) of quinoa (*Chenopodium quinoa* Willd.) under NaCl application condition. Pot experiments using Wagner pots (1/5000a) were conducted in an experimental field at Nihon University in 2018 and 2019, using quinoa variety CICA-127. The growth of quinoa as well as Cs accumulation and concentration was promoted by increasing the amount of NaCl applied. Quinoa accumulated most of the Cs in the leaves, and it was not translocated from the leaves to panicle after the seed filling stage. Cs accumulation by the aboveground parts under NaCl application was at least four times higher than that in the control. Accumulation of Na in stem was highest among organs. The quinoa plants had the mechanism to accumulate Na in the stem. Quinoa has bladder cells on the leaf surface, and excess Na accumulates in these cells. It is unknown whether bladder cells are present on the surface of the stem. Since Cs and Na inhibited the growth of plants, it is necessary to clarify the suppression method of stunting by Cs and Na. Thus, we believe that quinoa can be used for phytoremediation of Cs. Quinoa varieties with high Cs absorption need to be selected for effective phytoremediation in the future experiment.

**Keywords** Cesium · NaCl · Phytoremediation · Quinoa (*Chenopodium quinoa* Willd.) · Salt tolerance

## 1 Introduction

A large amount of radioactive cesium ( $^{134}\text{Cs}$  and  $^{137}\text{Cs}$ ) was released into the environment after the accident at Fukushima Daiichi Nuclear Power Plant that occurred on March 11, 2011 (Chino et al. 2011; Fujii et al. 2014). Contamination of soil surface with  $^{137}\text{Cs}$  occurred in a vast land area in Fukushima Prefecture, Japan. Radioactive Cs deposited on the soil surface was transferred from soil to crops (Tanoi et al. 2013; Tsukada et al. 2002; Kubo et al. 2017), and the crops that accumulated radioactive Cs could not be shipped to the market (Miyahara 2014; Watanabe 2014; Sato 2014). In Cs-contaminated land, the soil surface was stripped to remove radioactive Cs (Ministry of Agriculture, Forestry and Fisheries of Japan 2013). However, this method adversely affected the physical properties and ecosystem of the soil. Phytoremediation is a useful technique for decontaminating soil, and it can help decontaminate large areas of agricultural land at low cost (Broadley et al. 1999; Hayakawa and Kurihara 2002; Sato 2014; Ogata et al. 2015; Kubo et al. 2017). For example, after the Chernobyl nuclear accident, amaranth (*Amaranthus* spp.) plants were used to remove radioactive Cs from contaminated soil (Dushenkov et al. 1999). As dicotyledonous (Magnoliopsida) plants have higher Cs accumulation ability than monocotyledonous (Liliopsida) plants, the use of dicotyledonous plants for

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phytoremediation was more effective than that of monocotyledonous plants. Many plant species of Dicotyledons, including sunflower (*Helianthus annuus* L.), kokia (*Bassia scoparia* L.), and quinoa (*Chenopodium quinoa* Willd.), have been used for phytoremediation (Hirayama et al. 2012; Ogata et al. 2015; Tamaoki et al. 2016). Quinoa is a plant species with high Cs accumulation ability (Broadley et al. 1999). Therefore, the use of quinoa is considered to be effective for the removal of radioactive Cs from soil. Moreover, the growth of quinoa was promoted by applying NaCl to the soil; Cs accumulation increased with an increase in Na accumulation (Isobe et al. 2019). However, as Cs is toxic to plants (Hampton et al. 2004; Adams et al. 2013), the absorbed Cs in plants should be detoxified or expelled. Most of the absorbed Cs from roots is translocated to leaves, flowers, and fruits (Menzel and Heald 1955; Zhu and Smolders 2000). For example, in *H. annuus* L., most of the absorbed Cs accumulated in young leaves (Soudek et al. 2006). Buckwheat (*Fagopyrum esculentum* M.) mainly absorbed Cs until the flowering stage and translocated Cs from the roots to shoots and from shoots to the reproductive organs (Kubo et al. 2017). In tomato (*Solanum lycopersicum* L.), the absorbed Cs is mainly accumulated in the leaves, but less than 10% of absorbed Cs is stored in fruits after the reproductive stage (Sabbarese et al. 2002). In rice (*Oryza sativa* L.), 73% of absorbed Cs accumulated in straw (Tsukada et al. 2002). The distribution of absorbed Cs varies among different plant species, and the accumulation rate in each organ varied with the growth stage (Kubo et al. 2017).

Quinoa is one of the plants with high cesium accumulation, and Cs accumulation of quinoa was promoted by applying NaCl to the soil (Isobe et al. 2019). However, the distribution of accumulated Cs in each organ and the translocation of Cs among different growth stages are not clear. In this study, we aimed to clarify the organs of quinoa in which Cs was accumulated. Furthermore, the Cs concentration in each organ was analyzed under conditions of NaCl application.

## 2 Materials and Method

The study was conducted in an experimental field at Nihon University (Fujisawa City, Kanagawa, Japan) in 2018 and 2019 using quinoa variety CICA-127. Wagner

pots (1/5000a; diameter around 16 cm, height around 25 cm) were used for the study.

For the experiment, 2.6 kg of field soil, 0.95 g of ammonium sulfate (TORAY Ind. Inc., Tokyo, Japan), 1.14 g superphosphate (Katakura & Co-op Agri Corporation), and 0.10 g of CsCl (Wako Pure Chemical Industries, Tokyo, Japan) were applied to all pots. Three different treatments were performed: NaCl-1 plot, NaCl-2 plot, and a control with no added NaCl. In NaCl-1 plot and NaCl-2 plot, 9.75 and 19.5 g of NaCl were applied to the soil, respectively. NaCl and fertilizer were equally mixed with 2.6 kg of soil, and CsCl was added to the top 5 cm of soil in all plots. Twenty quinoa seeds per pot were sown on August 16, 2018, and August 26, 2019. All pots (14 pots per plot) were placed randomly and independently in an unheated glass (roof) and net (side) house from sowing to sampling of aboveground parts. Weeds, diseases, and insects were controlled as necessary during the cultivation period. The seedlings were thinned to three plants per pot at the second or third leaf stage.

On October 9, 2018, and November 7, 2018, and on October 7, 2019, and November 11, 2019, the aboveground parts of plants from five (October 9, 2018, and October 7, 2019) or nine (November 7, 2018, and November 11, 2019) pots in each plot were cut at the soil surface. Fresh and dry weights of aboveground parts (leaf, stem, and panicle) and Cs, K, and Na concentration and accumulation of each organ (leaf, stem, and panicle) were measured. The leaf areas were measured with a leaf area meter (LO-310, LI-COR Inc., Lincoln, NE, USA). The fresh and dry weights of the aboveground parts of plants were measured using an electronic balance. The dry weight of aboveground parts of plants was determined after oven drying the fresh samples at 80 °C for 48 h. Each dried organ (leaf, stem, and panicle) was ground to a powder using a blender. To measure Cs, K, and Na concentrations, 0.5 g of ground material was digested in 20.0 mL HClO<sub>4</sub> (Kanto Chemical Co., Inc., Tokyo, Japan) for 3 h at 100 °C using an acid digestion system, and Cs, K, and Na concentrations were determined by atomic absorption spectrophotometry (iCE 3300 AAS Thermo Fisher Scientific, Waltham, MA, USA). Cs, K, and Na accumulation by each organ was measured by multiplying the dry weight of each organ by the Cs, K, and Na concentration.

All values were expressed as averages and standard errors. The data were analyzed statistically, and significant differences at a 5% level among the plots or organs

were determined by Tukey multiple means test using Kaleida Graph ver.4.0 software.

### 3 Results

#### 3.1 Quinoa Growth

The leaf area as well as fresh and dry weights of aboveground parts of plants increased with increasing NaCl application rates. Significant differences at the 5% level were observed in leaf area, fresh weight, and dry weight between plots, except for the dry weight of aboveground parts of plants at 54 days after sowing in 2018 (Table 1).

#### 3.2 Cs, K, and Na Accumulation

Accumulation of Cs and Na by the aboveground parts of plants increased with increasing NaCl application rates in both years. Significant differences at the 5% level were observed in Cs and Na accumulation between plots (Table 2). Accumulation of K by the aboveground parts of plants increased with increasing NaCl application rates in both years. However, significant differences at the 5% level were observed only in K accumulation between plots at 83 days after sowing in 2018 (Table 2).

Accumulated of Cs by each organ (leaf, stem, and panicle) increased with increasing NaCl application rates. Significant differences at the 5% level were observed in Cs accumulation by each organ between plots, except for stem at 77 days after sowing in 2019. Accumulation of Cs by leaves at 54 days after sowing in 2018 and 42 days after sowing in 2019 was the highest among all organs. However, at 83 days after sowing in 2018 and 77 days after sowing in 2019, the accumulation of Cs by panicle was the highest among all organs (Table 3).

The accumulation of K was not affected by NaCl application in different plots. Among all organs, accumulation of K by leaves or stems was the highest at 54 days after sowing in 2018 and 42 days after sowing in 2019. There were significant differences at the 5% level in K accumulation between organs. At 83 days after sowing in 2018 and 77 days after sowing in 2019, the accumulation of K by panicle was higher than that by the other organs at 5% level (Table 3).

Accumulation of Na by each organ (leaf, stem, and panicle) increased with increasing NaCl application rates. Significant differences (5% level) in Na

accumulation were observed among plots in all organs, except for leaves at 54 days after seeding in 2018. Accumulation of Na by stems was the highest among all organs. There were significant differences at 5% level in Na accumulation between organs, except for stems at 42 days after sowing in 2019 (Table 3).

#### 3.3 The concentration of Cs, K, and Na in Each Organ

The concentration of Cs in each organ (leaf, stem, and panicle) increased with increasing NaCl application rates. A significant difference (5% level) was observed between plots, except for stem and panicle at 83 days after sowing in 2018. Among the different organs, the concentration of Cs was the highest in leaves. There were significant differences in concentration of Cs at the 5% level between organs (Table 4).

Among different plots and organs, the concentration of K was not affected by NaCl application (Table 4).

Concentration of Na in each organ (leaf, stem, and panicle) increased with increasing NaCl application rates. There were significant differences at the 5% level between the plots, except for Na concentration in panicle 54 days after sowing in 2018. Among the different organs, concentration of Na in the stem was highest, except for 19.5 g NaCl at 77 days after sowing in 2019 (Table 4).

### 4 Discussion

#### 4.1 Quinoa Growth and Accumulation of Cs, K, and Na in Aboveground Parts

Quinoa (*C. quinoa* Willd.) is a halophyte with high salinity tolerance, and its growth is promoted under high NaCl conditions (Isobe et al. 2014; Saleem et al. 2017). In this study, the leaf area, aboveground fresh weight, and aboveground dry weight of quinoa increased with the increasing application rate of NaCl (Table 1). This result is almost similar to that of the study by Isobe et al. (2019). Cs accumulation by quinoa is promoted by low available K content in soil (Li et al. 2015) and application of NaCl (Isobe et al. 2019). One of the reasons for increased Cs accumulation with decreasing available K content in soil was that Cs and K belong to group I alkali metals (White and Broadley 2000; Nishioka et al. 2011). In addition, the influx of Cs into root cells is mediated by the same molecular mechanism of K influx (White

**Table 1** Effects of NaCl application on the quinoa growth

Year	Day after sowing	Plots	Leaf area (cm <sup>2</sup> /pot)	Fresh weight of aboveground parts of plants (g/pot)	Dry weight of aboveground parts of plants (g/pot)
2018	54 days	Control	374.5 ± 27.2 b	18.5 ± 0.82 b	3.70 ± 0.18 a
		9.75 g NaCl	585.2 ± 20.2 ab	27.9 ± 0.58 a	4.70 ± 0.13 a
		19.5 g NaCl	609.8 ± 101.0 a	27.9 ± 3.98 a	3.74 ± 0.4 2 a
	83 days	Control	153.8 ± 11.2 b	20.7 ± 1.07 b	5.74 ± 0.30 c
		9.75 g NaCl	145.4 ± 19.2 b	34.3 ± 1.35 a	8.27 ± 0.24 a
		19.5 g NaCl	271.6 ± 20.4 a	33.6 ± 1.66 a	6.81 ± 0.34 b
2019	42 days	Control	299.6 ± 25.1 b	14.1 ± 1.19 b	1.60 ± 0.31 b
		9.75 g NaCl	397.5 ± 59.7 b	21.0 ± 3.21 ab	2.86 ± 0.64 ab
		19.5 g NaCl	557.5 ± 33.0 a	27.9 ± 1.22 a	3.27 ± 0.22 a
	77 days	Control	141.8 ± 17.1 b	21.9 ± 2.83 b	6.75 ± 0.76 b
		9.75 g NaCl	215.7 ± 49.9 a	30.0 ± 3.76 a	8.77 ± 1.05 ab
		19.5 g NaCl	283.5 ± 52.5 a	39.4 ± 1.75 a	9.24 ± 0.44 a

Average ± standard error. Values followed by different letters are significantly different at  $P = 0.05$  by the Tukey's multiple test

and Broadley 2000; Adams et al. 2015; Furukawa 2014). On the other hand, the reason for increased Cs accumulation instead of K under high saline conditions (Table 2) is probably to increase salt tolerance by Na accumulation (Tuteja 2007). As K fertilizer was not applied in this study, it was considered that Cs accumulation was promoted by lower available K content of soil (Table 2). Because Cs is toxic to plants (Hampton et al. 2004; Adams et al. 2013), the accumulated Cs should be detoxified or expelled out of the plant. Therefore, the metabolic changes or excretion

mechanism of accumulated Cs in quinoa plants should be clarified in future studies.

#### 4.2 Accumulation and Concentration of Cs in Each Organ

The accumulation of Cs and Na by each organ increased with increasing NaCl application rates in both years. However, the accumulation of K by each organ did not increase with increasing NaCl application rates (Table 3). These results showed the same trend as the accumulation of each chemical element (Cs, K, and Na)

**Table 2** Effects of NaCl application on the Cs, Na, and K accumulation of aboveground parts of plant

Year	Plots	Cs (mg/pot)		K (mg/pot)		Na (mg/pot)	
		54 days after sowing	83 days after sowing	54 days after sowing	83 days after sowing	54 days after sowing	83 days after sowing
2018	Control	2.60 ± 0.16 b	3.31 ± 0.33 b	137.38 ± 9.31 a	131.39 ± 13.11 b	0.16 ± 0.01 b	0.22 ± 0.03 b
	9.75 g NaCl	7.65 ± 0.32 a	7.88 ± 0.43 a	160.94 ± 12.34 a	162.07 ± 5.67 ab	19.59 ± 0.90 ab	38.53 ± 3.31 a
	19.5 g NaCl	7.84 ± 1.45 a	7.49 ± 0.93 a	188.08 ± 28.97 a	223.85 ± 23.06 a	43.86 ± 12.62 a	50.86 ± 7.33 a
2019	Control	2.75 ± 0.34 c	8.23 ± 0.88 b	65.50 ± 7.58 a	76.54 ± 15.30 a	1.92 ± 0.21 b	4.31 ± 0.28 b
	9.75 g NaCl	9.63 ± 0.55 b	18.21 ± 4.88 ab	94.86 ± 32.89 a	107.39 ± 31.16 a	43.83 ± 6.05 a	43.13 ± 11.77 ab
	19.5 g NaCl	13.53 ± 1.77 a	27.31 ± 1.74 a	161.00 ± 22.41 a	93.15 ± 6.38 a	65.53 ± 8.44 a	92.34 ± 16.21 a

Average ± standard error. Values followed by different letters are significantly different at  $P = 0.05$  by the Tukey's multiple test

**Table 3** Effects of NaCl application on the Cs, Na, and K accumulation of each organ

	Year	Day after sowing	Plots	Leaf		Stem		Panicle				
Cs (mg/pot)	2018	54 days	Control	1.42±0.08	b	A	0.96±0.11	b	B	0.22±0.05	b	C
			9.75 g NaCl	4.26±0.24	ab	A	2.12±0.13	a	B	1.27±0.23	a	B
			19.5 g NaCl	5.74±1.23	a	A	1.50±0.31	ab	B	0.60±0.04	b	B
		83 days	Control	0.74±0.15	b	B	0.83±0.12	b	B	1.74±0.19	b	A
			9.75 g NaCl	1.52±0.02	ab	B	1.52±0.13	a	B	4.84±0.54	a	A
			19.5 g NaCl	2.67±0.45	a	AB	1.12±0.14	ab	B	3.69±0.50	a	A
	2019	42 days	Control	1.88±0.31	b	A	0.88±0.11	b	B	—		
			9.75 g NaCl	7.01±0.73	a	A	2.62±1.05	ab	B	—		
			19.5 g NaCl	9.68±1.31	a	A	3.85±0.47	a	B	—		
		77 days	Control	1.18±0.23	b	B	2.89±0.25	a	B	5.82±0.62	b	A
			9.75 g NaCl	4.98±1.61	ab	A	5.72±1.36	a	A	9.42±1.25	a	A
			19.5 g NaCl	6.86±1.82	a	B	6.85±1.27	a	B	12.23±0.70	a	A
K (mg/g)	2018	54 days	Control	50.74±2.46	a	B	74.75±7.04	a	A	11.90±4.05	b	C
			9.75 g NaCl	60.37±9.70	a	A	59.10±10.24	a	A	40.04±4.57	a	A
			19.5 g NaCl	106.12±21.39	a	A	59.85±14.23	a	AB	22.11±4.32	ab	B
		83 days	Control	11.63±1.33	b	B	26.03±3.11	ab	B	93.74±15.73	a	A
			9.75 g NaCl	8.65±0.69	a	B	13.32±1.34	b	B	140.10±4.81	a	A
			19.5 g NaCl	44.06±2.65	a	B	33.11±6.28	a	B	146.69±16.41	a	A
	2019	42 days	Control	35.33±6.03	a	A	30.17±2.26	b	A	—		
			9.75 g NaCl	41.24±14.12	a	A	53.62±21.58	ab	A	—		
			19.5 g NaCl	66.49±10.00	a	A	94.51±12.47	a	A	—		
		77 days	Control	10.85±1.97	a	B	18.84±2.20	a	B	54.77±5.44	b	A
			9.75 g NaCl	23.98±8.32	a	B	14.5±1.57	a	B	81.60±1.57	a	A
			19.5 g NaCl	22.5±4.53	a	B	6.46±0.56	b	B	56.05±0.56	ab	A
Na (mg/g)	2018	54 days	Control	0.04±0.01	a	B	0.12±0.01	b	A	0.01±0.00	b	C
			9.75 g NaCl	5.81±2.09	a	B	14.30±0.97	ab	A	0.45±0.07	a	B
			19.5 g NaCl	10.09±4.38	a	B	33.50±8.19	a	A	0.27±0.05	a	B
		83 days	Control	0.02±0.01	b	B	0.17±0.03	b	A	0.03±0.00	b	B
			9.75 g NaCl	3.86±0.31	a	B	31.15±3.85	a	A	3.52±0.31	a	B
			19.5 g NaCl	9.78±1.78	a	B	37.50±4.85	a	A	3.58±1.16	a	B
	2019	42 days	Control	0.79±0.13	b	A	1.13±0.14	b	A	—		
			9.75 g NaCl	17.02±2.99	a	A	26.81±6.37	a	A	—		
			19.5 g NaCl	24.69±3.00	a	A	40.84±5.46	a	A	—		
		77 days	Control	0.20±0.04	b	C	3.60±0.26	b	A	0.63±0.06	b	B
			9.75 g NaCl	5.53±1.83	b	B	36.02±9.79	ab	A	1.71±0.22	b	B
			19.5 g NaCl	18.24±4.57	a	B	64.22±12.75	a	A	8.87±0.54	a	B

Average ± standard error. Values followed by different letters are significantly different at  $P=0.05$  by the Tukey's multiple test. The lower case letters indicate between plots in each sowing date, and the capital letters indicate between organs in each plot

by the aboveground parts (Table 2) and were almost similar to the findings of Isobe et al. (2019).

The accumulation of Cs by leaves was higher than that by the other organs (stem and panicle) at 54 days after sowing in 2018 and 42 days after sowing in 2019.

However, at 83 and 77 days after sowing, the accumulation of Cs by panicle was higher than that by the other organs (leaf and stem) (Table 3). In this study, the flowering stage was observed at 54 days after sowing in 2018, vegetative stage was observed at 42 days after

sowing in 2019, and seed filling stages were observed at 83 and 77 days after sowing. Thus, it was considered that Cs accumulation by leaves was higher than that by the other organs from the vegetative stage to the flowering stage, and Cs accumulation by panicle was

higher than that by the other organs after seed filling stage. The reason for the higher Cs accumulation by panicle than by the other organs was not due to the high Cs concentration in panicle (Table 4) but by the high dry weight of panicle. In contrast, decreased Cs

**Table 4** Effects of NaCl application on the Cs, Na, and K concentration of each organ

	Year	Day after sowing	Plots	Leaf		Stem		Panicle					
Cs (mg/pot)	2018	54 days	Control	1.05 ± 0.11	b	A	0.50 ± 0.03	b	B	0.84 ± 0.04	b	A	
			9.75 g NaCl	2.44 ± 0.12	a	A	0.98 ± 0.05	a	B	1.97 ± 0.21	a	A	
			19.5 g NaCl	3.21 ± 0.41	a	A	0.89 ± 0.01	a	B	1.97 ± 0.35	a	AB	
		83 days	Control	1.10 ± 0.15	b	A	0.43 ± 0.07	a	B	0.78 ± 0.12	a	AB	
			9.75 g NaCl	2.44 ± 0.08	a	A	0.43 ± 0.06	a	C	1.06 ± 0.11	a	B	
			19.5 g NaCl	2.71 ± 0.31	a	A	0.47 ± 0.06	a	B	1.11 ± 0.01	a	B	
		2019	42 days	Control	2.59 ± 0.07	c	A	1.49 ± 0.03	c	B	—		
				9.75 g NaCl	4.27 ± 0.06	b	A	2.04 ± 0.02	b	B	—		
				19.5 g NaCl	5.60 ± 0.14	a	A	2.61 ± 0.03	a	B	—		
	77 days	Control	2.95 ± 0.01	c	A	1.22 ± 0.01	b	C	2.02 ± 0.01	c	B		
		9.75 g NaCl	5.36 ± 0.07	b	A	1.60 ± 0.03	a	C	2.69 ± 0.02	b	B		
		19.5 g NaCl	6.12 ± 0.07	a	A	1.64 ± 0.01	a	C	3.16 ± 0.03	a	B		
K (mg/g)	2018	54 days	Control	37.15 ± 0.29	b	A	39.49 ± 0.93	a	A	44.36 ± 4.14	b	A	
			9.75 g NaCl	33.76 ± 5.61	b	B	27.59 ± 5.38	a	B	62.72 ± 0.79	a	A	
			19.5 g NaCl	59.62 ± 2.56	a	A	35.66 ± 6.26	a	B	67.53 ± 1.53	a	A	
		83 days	Control	17.82 ± 1.73	b	B	13.54 ± 1.62	a	B	40.17 ± 1.06	a	A	
			9.75 g NaCl	13.84 ± 0.53	b	B	13.82 ± 0.63	a	C	30.68 ± 1.35	b	A	
			19.5 g NaCl	45.43 ± 3.77	a	A	13.38 ± 0.47	a	B	44.38 ± 2.83	a	A	
		2019	42 days	Control	48.67 ± 1.43	a	A	52.09 ± 3.30	b	A	—		
				9.75 g NaCl	48.59 ± 1.21	a	A	41.33 ± 3.05	b	A	—		
				19.5 g NaCl	38.39 ± 1.89	b	B	63.84 ± 0.13	a	A	—		
	77 days	Control	27.20 ± 0.26	a	A	7.90 ± 0.38	a	C	19.26 ± 1.08	b	B		
		9.75 g NaCl	25.33 ± 1.25	a	A	4.49 ± 1.13	b	B	23.07 ± 0.94	a	A		
		19.5 g NaCl	20.66 ± 1.01	b	A	1.70 ± 0.42	b	C	14.63 ± 1.03	c	B		
Na (mg/g)	2018	54 days	Control	0.03 ± 0.00	b	B	0.06 ± 0.00	b	A	0.03 ± 0.00	a	B	
			9.75 g NaCl	3.24 ± 1.15	ab	B	6.63 ± 0.61	b	A	0.71 ± 0.09	a	B	
			19.5 g NaCl	5.07 ± 1.72	a	B	19.81 ± 3.49	a	A	0.95 ± 0.37	a	B	
		83 days	Control	0.04 ± 0.01	c	B	0.09 ± 0.01	c	A	0.01 ± 0.00	b	B	
			9.75 g NaCl	6.18 ± 0.27	b	B	8.69 ± 0.31	b	A	0.77 ± 0.06	a	C	
			19.5 g NaCl	9.89 ± 1.20	a	B	15.49 ± 0.81	a	A	1.03 ± 0.19	a	C	
	2019	42 days	Control	1.09 ± 0.03	c	A	1.93 ± 0.02	c	A	—			
			9.75 g NaCl	13.38 ± 0.19	b	B	16.05 ± 0.18	b	A	—			
			19.5 g NaCl	14.32 ± 0.15	a	B	27.58 ± 0.16	a	A	—			
77 days	Control	0.51 ± 0.01	c	B	1.52 ± 0.02	c	A	0.22 ± 0.01	c	C			
	9.75 g NaCl	5.93 ± 0.21	b	B	9.93 ± 0.43	b	A	0.50 ± 0.02	b	C			
	19.5 g NaCl	16.38 ± 0.11	a	A	15.34 ± 0.28	a	A	2.29 ± 0.04	a	B			

Average ± standard error. Values followed by different letters are significantly different at  $P=0.05$  by the Tukey's multiple test. The lowercase letters indicate between plots in each sowing date, and the capital letters indicate between organs in each plot



accumulation by leaves after the seed filling stage was mainly due to the decrease in leaf dry weight. The phenomenon of accumulation of most of the Cs absorbed by roots in the leaves (Table 4) was also observed in konjac (*Amorphophallus konjac* K. Koch), rapeseed (*Brassica napus* L.), and amaranth (*Amaranthus* spp.) (Hirayama 2012; Hirayama et al. 2013; Ogata et al. 2015). In the present study, the Cs concentration in leaves was highest among all organs from the vegetative stage to the seed filling stage (Table 4). Isobe et al. (2019) clarified that the weight of the panicle increased from the flowering stage to seed filling stage, but the Cs concentration of the above-ground parts did not increase after flowering stage. Thus, it was hypothesized that quinoa accumulates most of the Cs in leaves, and after the seed filling stage, most of the Cs did not translocate from the leaves to the panicle. And it is considered that Cs is not accumulated to grains at seed filling stage. To clarify the above hypothesis, it is necessary to compare the Cs concentration in fresh leaves and defoliation and clarify the change in Cs concentration in leaves with growth progress. However, in this experiment, the Cs concentration of grain after seed filling stage was not analyzed and did not compare the Cs concentration in fresh leaves and defoliation. And, in the future, it is necessary to investigate the Cs concentration of grain and leaves at maturity stage. At present, it is considered that the removal of radioactive Cs from soil by plants (phytoremediation) is not realistic, because the Cs accumulation ability of plants is very low (Sato 2014; Ogata et al. 2015). However, the Cs accumulation ability of quinoa and growth of its aboveground parts were promoted by the application of NaCl (Tables 1, 3, and 4). Cs accumulation by the aboveground parts in NaCl application plots was at least four times higher than that in the control (Table 2). Thus, quinoa can be used for phytoremediation of Cs by selecting varieties with high Cs accumulation ability; the conditions that increase Cs accumulation by quinoa should be clarified in the future.

#### 4.3 Na Tolerance Mechanism of Quinoa

The accumulation of Na by stem was highest among all organs in both years (Table 3). A possible reason for this is the highest dry weight of stem among all organs (particularly, at 54 days after sowing in 2018). Another reason could be the highest Na concentration in stem among all organs (Table 4). Increased Na accumulation

in stem was also observed in watermelon (*Citrullus lanatus* (Thunb.) Matsum. & Nakai) and pumpkin (*Cucurbita* spp.) (Yamauchi et al. 1986). In rice (*O. sativa* L.), most of the Na accumulated in leaf sheaths to avoid excessive Na accumulation in leaf blade and panicles (Kobayashi et al. 2017). The expression of *OsHKT1;5* gene was responsible for the accumulation of Na in leaf sheaths in rice, and it prevented Na translocation to leaf blade and panicle (Kobayashi et al. 2017). In wheat, the high salt tolerance varieties showed low Na concentration in leaf blade (Poustini and Siosemardeh 2004). Taken together, the lower Na concentration in leaf blade than in stem (Table 4) and the higher dry matter weight and leaf area under the application of NaCl (Table 1) is evidence for the high salt tolerance of quinoa.

The growth of many plants is inhibited by increased Na accumulation, and these plants could die by excessive Na accumulation (Tuteja 2007). Na suppressed plant growth by suppressing water absorption via increasing osmotic pressure of root cells and suppressing the absorption of other essential elements (Flowers and Yeo 1995), Khan et al. 2000; Horie et al. 2011; Gupta and Huang 2014). In plants, there are various preventive mechanisms against growth suppression by Na. For example, excessive Na accumulated by mangroves is excluded in the root and hypocotyls (Krishnamurthy et al. 2014). In *Shichimensou* (*Suaeda japonica* Makino), a halophyte, the leaves fall to exclude excessive Na (Shimizu et al. 2008). Furthermore, in *Mesembryanthemum crystallinum* L., excessive salts are accumulated in bladder cells, which are present on the surface of aboveground parts (Adams et al. 1998). In quinoa, bladder cells are present on the surface of leaves, and excessive Na is accumulated in these cells (Adams et al. 1998; Agarie et al. 2007; Adolf et al. 2013). In the present study, most of the accumulated Na was accumulated in the stem, and Na concentration in the stem was higher than that in the other organs (leaves and panicle) (Tables 3 and 4). However, the presence of bladder cells on the surface of the stem has not yet been confirmed in quinoa. Therefore, in the future, it is necessary to clarify the preventive mechanisms against growth suppression by Na, such as the presence of bladder cells on the surface of stem and accumulation of Na in each cell organelle.

## 5 Conclusion

In summary, we clarified the accumulation and concentration of Cs, Na, and K in each organ (leaves, stem, and panicle) of quinoa under NaCl application condition. The growth of quinoa as well as Cs accumulation and concentration was promoted by increasing the amount of NaCl applied. Quinoa accumulated most of the absorbed Cs in the leaves, and Cs was not translocated from the leaves to panicle after the seed filling stage. Thus, it was considered that the most efficient Cs removing growth stage was the maximum leaf area stage of quinoa, was not seed filling or maturity stage. Cs accumulation by the aboveground parts under NaCl application was at least four times higher than that in the control. Thus, quinoa can be used for phytoremediation; effective phytoremediation can be achieved by selecting varieties with high Cs accumulation. Moreover, for the use quinoa in phytoremediation of Cs, the environmental conditions that increase Cs accumulation should be clarified in future studies.

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