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Growth Responses and Accumulation Characteristics of Three Ornamental Plants to Sn Contamination in Soil

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Abstract: Decorative ornamental plants have been applied as hyperaccumulators/phytoremediators to a wide spectrum of heavy metal contaminants. In this study, pot culture experiments were conducted to investigate the Sn tolerance and accumulation in *Impatiens balsamina* L., *Mirabilis jalapa* L. and *Tagetes erecta* L., in order to assess the possibility of these three ornamental plants to be used as phytoremediators of Sn-contaminated soil. Results show that all three plants exhibited strong tolerance to Sn contamination, and no significant visual toxicity was observed for all three plants grown under most of the Sn treatments. The amount of Sn accumulated in the three plants was positively correlated with the Sn concentration in the soil. The order of the Sn accumulative capacity was *Impatiens balsamina* > *Mirabilis jalapa* > *Tagetes erecta*. *Impatiens balsamina* and *Tagetes erecta* showed a low translocation ability (TF) (<1), and the roots accumulated the highest Sn concentration, but *Impatiens balsamina* showed a relatively high bioconcentration factor (BCF, Sn concentration in each part > 100 mg/kg after Sn treatment of 500 mg/kg). Meanwhile, the TF of *Mirabilis jalapa* was >1, and the fluorescence accumulated the most Sn. In combination with the adaptation to high concentrations of various heavy metals, these three ornamental plants are potential candidates for Sn mining tailings or contaminated soil.

Keywords: Sn; toxicity; accumulation; phytoremediation; soil

1. Introduction

Tin (Sn) generally exists as +IV and +II oxidation states [1]. Sn compounds have been widely applied for industrial purposes. For example, SnO₂ was used as a catalyst in certain industrial processes and a polishing powder for steel, SnCl₂ was used as a reducing agent in the manufacture of polymers and dyes, SnF₂ and Sn pyrophosphate were used in dentifrices and organic Sn compounds were used as stabilizers in plastics and preservatives, as well as biocides for bacteria, fungi and insecticides [1–3]. Though elemental Sn is nontoxic and a normal part present in the tissues of plants and animals [3], the above Sn compounds and organic Sn compounds are toxic to bio-organisms. It has been reported that excess uptake of SnCl₂ reduced the calcium content and the strength of bone and decreased hemoglobin and red blood cells, which led to anemia for the tested rats [1,3]. Excess SnCl₄ in human lymphocytes induced chromosome aberrations, micronuclei and sister chromatid exchanges (SCEs). Lastly, previous studies found SnF₂ caused DNA damage in cultures of human lymphocytes [1,3].

Currently, in the early 21st century, China has the largest Sn production worldwide, accounting for nearly half of the global production [4]. Improper exploration and refinement of Sn produced a substantial amount of abandoned mine wastes and led to serious



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Copyright: © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). environmental pollution. After well-known Sn mining activities, a mined legacy land composed of a complex mosaic of lagoons, overburden dumps and tailings usually had unfavorable conditions for vegetation and became a major pollution source of soil, air and surface and underground water [5–8]. For example, Ashraf et al. (2011) observed that Sn concentrations were 5200–5832, 194.80–817.55 and 109.56–658.12 mg/kg (dry weight) in the soil, plant roots and plant shoots, respectively, when investigated in a decommissioned Sn mining region [6]. Schreck et al. (2012) surveyed a peri-urban zone with metallurgic activities and found that Sn concentrations were 2.6 \pm 0.5 mg/cm² and 1.3 \pm 0.1, 7.9 \pm 1.9 and 16.4 \pm 0.7 mg/kg (dry biomass) in atmospheric fallouts, lettuce, parsley and ryegrass, respectively, after being exposed to such atmospheric fallouts for 4 weeks [7].

Studies have been conducted to investigate the behavior and dynamics of Sn in the environment. It is generally considered that Sn is highly immobile and shows a poor downward migration rate, and it tends to complex with organic matter at a horizontal level where organisms are abundant [9–11]. For example, Murata et al. (2018) found that Sn was retained in the uppermost (0–2 cm) soil layer even after being added for 8 years, and the proportion of mobile Sn (mostly existing as a Me-Org fraction) remained constantly at 12–17% [12]. Nakamaru and Uchida (2008) showed that the sorption behavior of Sn was highly correlated with the amount of active Al- (Al-(hydr)oxide and Al-humus complex) in the soil [13]. The immobile and insoluble property of Sn in the environment challenged the traditional washing, flushing and solidification remediation techniques. Meanwhile, the phytoremediation technique, referring to the phytoextraction, phytodegradation, rhizofiltration, phytostabilization and phytovolatilization of contaminants [14,15], and maintaining the biological activity and physical structure of contaminated sites in the meantime [16], was proposed as a potentially efficient remediation technique. Phytoremediation has been widely applied to the remediation of heavy metal contamination for a long time, and hyperaccumulators/phytoremediators have been found for a wide spectrum of heavy metal elements. However, phytoremediation of Sn has rarely been studied due to the lower toxicity of the Sn element compared with other heavy metals such as Cd, Cr and Cu [17,18].

The aim of the present study was to investigate the toxicity and accumulation of Sn in plants, thus trying to search for potential hyperaccumulators/phytoremediators of the Sn element. Three ornamental plants, *Impatiens balsamina* L., *Mirabilis jalapa* L. and *Tagetes erecta* L., were chosen as test plants, since they are widely used decorative plants in cities and separated from the food chain, and they were also characterized as hyperaccumulators/phytoremediators of heavy metals, including Cd, Pb, Cu, Zn and As. This was important since Sn was generally associated with these heavy metals in Sn mining tailings or contaminated sites. Accordingly, the ability of the three ornamental plants to tolerate and accumulate elevated levels of Sn in the soil was investigated by pot cultural experiments. The bioconcentration factor (BCF) and translocation factor (TF) were calculated to assess the potential of these three ornamental plant species as Sn hyperaccumulators/phytoremediators.

2. Materials and Methods

2.1. Substrates and Plants

The surface soil (0 to 20 cm depth) was collected from the Garden of Nankai University, Tianjin, China, without prehistory of Sn contamination. The soil samples were ground using pestles, sieved through a 4 mm mesh and thoroughly mixed to yield a homogenous soil composite. The soil was air dried and filled into plastic pots.

Ornamental plant species of *Impatiens balsamina*, *Mirabilis jalapa* and *Tagetes erecta* were used for the experiments. Plant seeds were purchased from the Xuefu Flower Market, Tianjin, China. These plants can be easily transplanted and cultivated and have strong adaptability to harsh environments. In addition, they have high biomass production and wide geographic distributions. Before planting, the plant seeds were pre-treated by

sterilizing in 3% hydrogen peroxide for 20 min to prevent fungal contamination, washing with water several times and then drying softly with tissue.

2.2. Plant Growth Experiment

A plant growth experiment was conducted in the greenhouse of Nankai University at Tianjin, China. An amount of 1200 g of air-dried soil was weighed into plastic pots $(\Phi = 20 \text{ cm}, \text{H} = 15 \text{ cm})$. The form of Sn spiked in the tested soils was SnCl₂·2H₂O and the concentrations of Sn spiked in the tested soils were 100, 500, 1000 and 2000 mg/kg, signed as T1, T2, T3 and T4, respectively. The concentrations of Sn were 2–3 times higher than the soil background levels and were in accordance with the current situations and levels of soils contaminated by Sn tailings from Gejiu, Yunnan [5]. A control experiment (CK) was performed without an external Sn addition. Replicates were conducted for all the treatments to minimize experimental errors and allowed to equilibrate completely for 28 days. After that, 2–3 seedlings of similar biomass and height which were grown for about 1 month were transplanted into the pots. The number of seedlings for each treatment was kept equal. The plants were naturally illuminated with a light/dark cycle of approximately 15/9 h, and the temperature was kept around 23–30 °C. Soil moisture was maintained at 65% of the field water holding capacity with tap water (no Sn detected). No additional fertilizer was added. The plants were harvested at the seed maturity stage after three months. The plant height was measured before harvesting.

2.3. Substrates and Plants

Harvested plants were carefully rinsed with tap water followed by distilled water to remove dirt and soil particles. The plant samples were then divided into roots, stems, leaves and inflorescences and dried at 105 °C for 30 min and then at 70 °C to constant weight (dry weight) in an oven (around 3 days). After the dry weight was recorded, the separated plant tissues were ground to a powder. Around 0.25 g of dry biomass was digested in a solution containing 6 mL of concentrated HNO₃ (65%) and 1 mL of H₂O₂ (30%) by a microwave digestion instrument. The digested solutions were filtered through 0.45 μ m membranes and Sn concentrations in digested solutions of plant tissues were determined using the flame atomic absorption spectrophotometer (AA240FS, VARIAN). The wavelength for the Sn determination was 286.3 nm. A certified standard reference material (bought from an authoritative company in Shijiazhuang, China) was used to ensure the quality control of measured Sn values.

The bioconcentration factor (BCF) and translocation factor (TF) were calculated to assess the phytoremediation potential of the different ornamental plants [19,20].

$$TF = (Concentration of metal in shoot)/(Concentration of metal in root)$$
 (2)

The initial concentration of metal in the substrate (in soil) was measured before plant harvest (both the control as well as the treatment) [19].

All data were statistically analyzed by SPSS 13.0, Origin 8.5 and Excel 2011. Oneway analysis of variance with the least significant difference (LSD) and Duncan test was conducted to explore the differences between Sn treatment samples and controls, taking p < 0.05 as significant. Results were expressed as mean \pm standard deviations (SD).

3. Results

3.1. Sn Tolerance under Soil Culture Conditions

All three tested ornament plants grew well in Sn-spiked soil and no death was recorded during the whole experimental period. The leaf color of the three tested ornamental plants did not show any appreciable changes (Figure S1). The plant height of Impatiens balsamina and Mirabilis jalapa grown in Sn-loaded soils did not show visible differences (Figure 1A). However, the growth of Tagetes erecta was reduced by 10% under the T4 treatment compared to the control experiment (Figure 1A). It seemed that the plant heights of Impatiens balsamina, Mirabilis Jalapa and Tagetes erecta were not sensitive to Sn contamination, although the growth of Tagetes erecta was restrained under the highest Sn treatment (T4) to some extent.

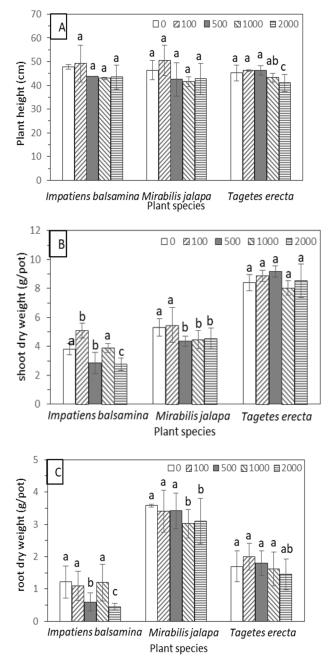


Figure 1. Sn tolerance under soil culture conditions for three ornamental plants: (**A**) stem length under different Sn treatments; (**B**) shoot biomass (dry weight) under different Sn treatments; (**C**) root biomass (dry weight) under different Sn treatments. Bar diagrams in the same column followed by the same letter are not significantly different, whereas those with different letters indicate significant differences (p < 0.05). Bars indicate standard errors (n = 3).

The dry biomass of plant tissues was also used to assess plant tolerance to Sn contamination. For Impatiens balsamina, the dry biomass of shoots increased significantly at the lowest Sn concentration (100 mg/kg), followed by a significant decrease under the higher Sn concentration treatment (Figure 1B). For Mirabilis jalapa, the dry biomass of shoots reduced under different Sn treatments except for T1, but the dry biomass remained constant with increasing Sn concentrations from the T2 to T4 treatments (Figure 1B). The dry biomass of Tagetes erecta increased to a different extent compared to the control (CK), except for T3 (Figure 1B).

In terms of root biomass, Impatiens balsamina and Mirabilis jalapa showed a reduced biomass compared with the controls (CK), especially under the T4 treatment, where the dry biomass was 60% and 14% less than that under the control, respectively (Figure 1C). Similarly, the root biomass of Tagetes erecta decreased with the increase in Sn treatments, except for T1 and T2 (Figure 1C).

The results of the plant height and dry biomass showed that appropriate Sn concentrations may facilitate plant growth, and all three ornamental plants had strong tolerance to Sn contamination. The intuitionistic Sn tolerance of the tested plants was in the order of Mirabilis jalapa > Tagetes erecta > Impatiens balsamina.

3.2. Sn Accumulation Characteristics under Soil Culture Conditions

A summary of plant tissue analyses on Sn concentrations is provided in Table 1. In general, the accumulated Sn concentrations for all three plants were positively correlated with the Sn concentration in the soil. For Impatiens balsamina, the order of Sn accumulation in different tissues was root > fluorescence > leaf > stem under different Sn treatments, except for T3, which accumulated more Sn in the stem than that in the inflorescence and leaf. The accumulated Sn concentrations in different tissues systematically increased with an increasing Sn concentration in the soil, except for the T4 treatment. The Sn concentration in the roots of Impatiens balsamina reached 340.7 mg/kg under the T3 treatment (Sn = 1000 mg/kg). For *Mirabilis jalapa*, the Sn concentration in plant tissues generally positively correlated with the Sn concentration in the soil, and florescence showed the highest Sn accumulation, followed by the root, leaf and stem for T1, T2 and T3 treatments. The Sn concentration in the florescence and stem under the T3 treatment was 131 and 55.5 mg/kg, respectively. For Tagetes erecta, the roots accumulated the most Sn and the concentration increased with increasing Sn concentration in the soil, and the Sn concentration in roots exceeded 95.8 mg/kg under the T4 treatment. Meanwhile, the stem, leaf and florescence did not show detectable Sn concentrations except under T1 and T4 treatments.

Plant Species	Treatment (mg/kg)	The Concentration of Sn (mg/kg)					
		Root	Stem	Leaf	Inflorescence	Shoot ¹	Soil
Impatiens balsamina	0	nd	nd	nd	nd	nd	nd
	100	86.2 ± 18.6	72.6 ± 4.9	79.1 ± 10.8	80.7 ± 1.7	77.4 ± 2.5	52.7 ± 2.5
	500	140.7 ± 1.8	95.3 ± 13.0	98.0 ± 9.3	111.6 ± 24.8	100.7 ± 14.4	265 ± 14.4
	1000	340.7 ± 23.8	229.9 ± 22.2	125.0 ± 29.9	122.5 ± 1.1	161.4 ± 19.9	520.7 ± 19.9
	2000	244.8 ± 12.9	95.5 ± 2.9	114.3 ± 23.8	133.5 ± 52.4	114.4 ± 26.4	1040 ± 26.4
Mirabilis jalapa	0	nd	nd	nd	nd	nd	nd
	100	22.2 ± 38.4	23.9 ± 22.3	23.2 ± 27.3	100.0 ± 58.5	25.5 ± 24.2	53.1 ± 24.2
	500	34.5 ± 23.8	12.4 ± 11.4	41.2 ± 16.8	110.8 ± 27.1	35.3 ± 10.4	271.6 ± 10.4
	1000	64.2 ± 20.7	55.5 ± 47.7	58.8 ± 14.0	131.0 ± 32.9	65.7 ± 40.8	547.5 ± 40.8
	2000	74.3 ± 28.0	76.6 ± 27.2	93.7 ± 23.3	91.0 ± 72.4	90.0 ± 16.6	1000 ± 16.6
Tagetes erecta	0	nd	nd	nd	nd	nd	nd
	100	18.0 ± 20.5	9.9 ± 17.1	5.4 ± 9.4	7.0 ± 10.4	7.4 ± 12.3	52.9 ± 12.3
	500	17.9 ± 28.2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	252 ± 22.4
	1000	31.2 ± 25.2	0.0 ± 0.0	5.3 ± 6.1	0.0 ± 0.0	1.8 ± 2.0	360 ± 2.0
	2000	95.8 ± 42.4	5.1 ± 7.7	14.7 ± 11.5	14.1 ± 6.9	11.3 ± 9.9	1130 ± 9.9

Table 1. Accumulation characteristics of plants grown in Sn-contaminated soil.

¹ The Sn concentration in shoots was calculated by the mean values of the Sn concentration in the roots, stem, leaf and inflorescence.

As shown in Table 1, the order of the Sn accumulative capacity was *Impatiens balsamina* > *Mirabilis jalapa* > *Tagetes erecta* for the three ornamental plants. Large differences between tissues are expected because of the variation in the abilities of individual plants to uptake Sn. *Impatiens balsamina* and *Mirabilis jalapa* show some characteristics as a hyperaccumulator of Sn.

3.3. Sn Accumulation Characteristics under Soil Culture Conditions

The measurements of BCF and TF values described the Sn accumulation potential of the three ornamental plants (Figure 2). The treatment of 100 mg/kg Sn in the soil exhibited the highest BCF and TF values for *Impatiens balsamina*, of which a BCF value > 1 was observed (Figure 2A). The highest Sn treatment (2000 mg/kg) in the soil showed the lowest BCF (Figure 2A) and TF (Figure 2B) values. It is worth noting that TF values lower than 1 were obtained for all treatments, though the concentrations of Sn in plant tissues were substantial for all treatments.

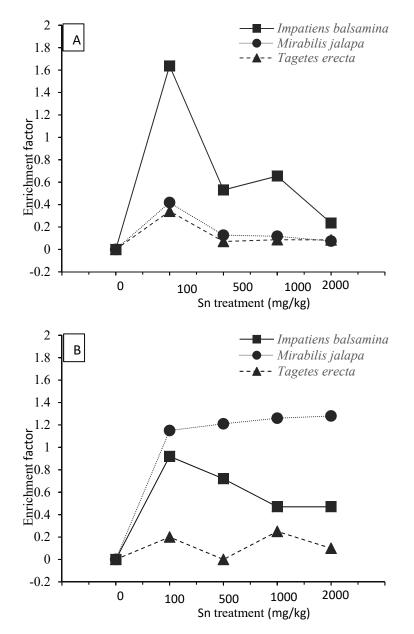


Figure 2. Sn accumulation potential of three ornamental plants: (**A**) bioconcentration factor (BCF) (concentration ratio of root to soil) of three ornamental plants; (**B**) translocation factor (TF) (concentration ratio of shoot to root) of three ornamental plants.

Mirabilis jalapa exhibited relatively lower BCF values, and all BCF values were <1 for all treatments of Sn levels in the soil (Figure 2A). The BCF values were negatively correlated with the Sn concentration in the soil. By contrast, the TF values were positively correlated with the Sn concentration in the soil, and TF values higher than 1 were obtained for all the treatments (Figure 2B).

For *Tagetes erecta*, neither the BCF value nor the TF value was more than 1 (Figure 2). The highest BCF value was exhibited at the treatment level of 100 mg/kg of Sn in the soil, and then the BCF values reduced significantly to negligible values when exposed to higher Sn concentrations in the soil. The TF values were significantly low for all the Sn treatments.

As shown in Figure 2A, the BCF values for the three ornamental plants were in the order of *Impatiens balsamina* > *Mirabilis jalapa* > *Tagetes erecta*, which is in accordance with the accumulative capacity. By contrast, the TF values for the three ornamental plants were in the order of *Mirabilis jalapa* > *Impatiens balsamina* > *Tagetes erecta*. Only *Mirabilis jalapa* obtained a TF value higher than 1, which means that the Sn concentration in shoots was higher than that in the roots, and the potential transferring ability of Sn from the roots to the shoots. For *Impatiens balsamina*, although the TF values were less than 1, the Sn concentration in shoots exceeded 100 mg/kg, which was one of the criteria of a hyperaccumulator.

4. Discussion

Despite the high concentration of Sn in the plant tissue of *Impatiens balsamina*, *Mirabilis jalapa* and *Tagetes erecta*, and the fact that the accumulation concentration generally positively correlated with the Sn concentration in the soil (Table 1), no visual symptoms (e.g., plant height decrease) of Sn toxicity were observed, indicating that these ornamental plants are Sn-tolerant species [21]. For *Impatiens balsamina*, the Sn concentration in each part exceeded 100 mg/kg from T2 to T4 treatments; however, the TF was lower than 1, which indicates the low delivery of Sn from the roots to the shoots. Meanwhile, for *Mirabilis jalapa*, the aerial tissue—florescence—accumulated 91.0 \pm 72.4 mg/kg of Sn, which was higher than that in the root tissue. *Mirabilis jalapa* showed a higher translocation ability (TF > 1), though the Sn concentration in each part was no more than 100 mg/kg. *Impatiens balsamina*, *Mirabilis jalapa* and *Tagetes erecta* displayed some characteristics of hyperaccumulators applied to Sn contamination. For example, the main properties for hyperaccumulators include: (1) the accumulative concentration of heavy metals in plant shoots within the threshold of 100–10,000 mg/kg dry weight depends upon toxic levels; (2) BCF > 1; (3) TF > 1; and (4) extremely high tolerance to heavy metals; no visual toxicity to heavy metals [15,16].

As shown in Figure 1, *Mirabilis jalapa* and *Tagetes erecta* were able to develop a profuse root system and all three ornamental plants produced a high aboveground biomass when growing in Sn-contaminated soils. Profuse root systems and biomass were important factors for these plants to explore a great soil volume and retain heavy metal contaminants in the roots and translocate large amounts of heavy metal(loid)s to the aboveground tissues such as the shoots and florescence [16,22]. Different Sn uptake and phytoremediation abilities are expected since individual plants have their own special characteristics such as growth rates, root systems, metabolism pathways and endurance environments. It seemed that *Impatiens balsamina* and *Mirabilis jalapa* remediated the Sn-contaminated soil mainly by the phytoextraction process, while the *Tagetes erecta* plant might remediate the Sn-contaminated soil through either the phytostabilization or rhizostabilization process [15,22].

These three ornamental plants are potential candidates as phytoremediators of Sncontaminated soil. They exhibited high tolerance characteristics and accumulated substantial Sn in tissues. When applying these three ornamental plants to Sn-contaminated sites, considerable amounts of Sn could be extracted when plants were harvested, the environment was esthetically pleasant and the ecosystem could be recovered at the same time. However, studies are required to explore the molecular mechanisms of bioaccumulation and translocation of Sn to plants and find appropriate biodegradable chelators or methods to enhance the Sn phytoremediation efficiency. It is widely reported that many different biodegradable chelators (e.g., EDTA, EDDA and NTA) [23–27], nanomaterials [28], earthworms [29], bacteria (e.g., *Pseudomonas* and *Enterobacter*) [30,31] or fungi [32,33] have been applied to enhance the metal accumulation in plant tissues and facilitate the translation of metals from the roots to the aboveground shoots. Sn released to the soil environment usually existed as stable tetravalent compounds (Figure S2), had no effect on the viability of either the intact cells or protoplasts of plants and was mainly uptaken by plants through cell wall binding [34].

The effectiveness of plant species in metal phytoremediation is determined by the bioavailability of metal cations, plant species, the growth medium and the environment [16,22]. Thus, it is important to explore the capability of the proposed phytoremediators to cope with various biotic (disease and pest resistance) stresses and adapt to abiotic (heat, drought and salt tolerance) stresses when surviving under high concentrations of heavy metal contaminants. In the future, the practical application of phytoremediation and the final rational disposal of ornamental remediation plants should be strengthened, as most of the current research focuses on laboratory results and lacks rational disposal techniques. Meanwhile, the identification and selection of ornamental plants as sustainable phytoremediators should be continued to broaden the phytoremediator database.

5. Conclusions

The objective of this study was to explore the tolerance and accumulative properties of three ornamental plants to Sn contamination, in order to further discuss the possibility of these three ornamental plant species to be used as phytoremediators or hyperaccumulators in Sn-contaminated sites. The relatively minimal visual symptoms (e.g., plant height) of Sn toxicity, profuse root systems and abundant biomass indicate that these three ornamental plants are Sn-tolerant species. Increasing the accumulative Sn concentration in these three ornamental plants provided the possibility of application of phytoremediation to Sn contamination. In particular, for *Impatiens balsamina* and *Mirabilis jalapa*, both accumulated more than 100 mg/kg of Sn in plant tissue or showed a high translocation ability (TF > 1) and have been characterized as hyperaccumulators/phytoremediators of Cd, Pb, Cu, Zn and As; therefore, they are potential phytoremediators of those Sn mining tailings or contaminated soils where Sn usually coexisted with multiple heavy metal contaminants. In the future, more work is also needed to investigate the molecular mechanisms of bioaccumulation and translocation of Sn to plants and conduct studies to boost the Sn accumulation capacity further.

Supplementary Materials: The following are available online at https://www.mdpi.com/2077-047 2/11/3/205/s1, Figure S1: Ornamental plant growth under Sn treatment: (A) *Impatiens balsamina*, (B) *Tagetes erecta*, (C) *Mirabilis jalapa*, (D) root of *Tagetes erecta*, Figure S2: Sn speciation in 0.01 M background electrolyte solutions before adsorption as a function of pH (Sn concentration of 2000 kg/kg in soil), Table S1: Basic properties and Sn concentration of the tested soil.

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