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The potential of *Sedum alfredii* Hance for the biosorption of some metals from synthetic wastewater

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ABSTRACT

The capacity of a terrestrial Zn/Cd hyperaccumulator *Sedum alfredii* Hance to purify water polluted by Zn, Cd, Cu and Pb along with their removal pathway was investigated. *S. alfredii* Hance was grown in synthetic wastewater in 2.5-L capacity containers contaminated with (mg L^{-1}) 19.20 Zn, 11.24 Cd, 3.27 Cu and 0.53 Pb, respectively. The supplied metal concentrations were comparable with those in plating industry wastewater. After a growth period of 7 days the average removal rates for Zn, Cd, Cu and Pb, were 94.07%, 82.33%, 96.03% and 69.61% respectively. Both root surface adsorption and absorption contributed to metal uptake. Root surface adsorption accounted for 67.74%, 24.03%, 66.53% and 70.77% of the total respective Zn, Cd, Cu and Pb, removal by *S. alfredii*. Addition of heavy metals stimulated the growth of *S. alfredii* over the control (without any addition of heavy metals) that might cause enhanced metal removal from wastewater. These results indicated promising potential of *S. alfredii* for remediation of a mixture of heavy metals containing wastewater. Rhizospheric microorganisms, root exudates and metal adsorption on the root surface of *S. alfredii* might be possible contributors in phytoremediation of Zn, Cu and Pb from wastewater.

1. Introduction

Electroplating industry is one of the major contributors of heavy metal-laden effluents into the environment. Conventional technologies for the removal and recovery of heavy metals from electroplating wastewater include chemical precipitation, ion exchange, membrane separation and adsorption by activated carbon. Most of these methods have some bottlenecks such as high operational costs and need for disposal of resulting sludge, particularly when the unwanted heavy metals are present at low concentrations i.e. <100 mg L^{-1} [1–3]. Phytoremediation is a recently developed biotechnology that offers many advantages like its low operational cost and environmental-friendly technique with minimal secondary pollution, and therefore is a hot spot of scientific research and environmental remediation application [4,5].

Aquatic macrophyte treatment systems (AMATS) have extensively been employed for water purification. Aquatic macrophytes like *Salvinia herzogii*, *Pistia stratiotes*, *Hydromistia stolonifera*, *Eichhornia crassipes*, *Lemna minor*, *Azolla filiculoides* and *Spirodela polyrhhiza* can uptake various heavy metals from the growth medium [6–10]. However, the metal removal efficiency by these plants is low due to their small size and slow growing roots. The higher water content of aquatic plants also complicates their drying, composting and incineration. In contrast, terrestrial plants develop much longer, fibrous root systems covered with root hairs offering an extremely high surface area [11]. These roots can be easily dried in open air. Dushenkov et al. [11] used *Brassica juncea* (Indian mustard) to remove Zn, Cd, Cr, Ni, Cu and Pb from aqueous streams. The roots of *Brassica juncea* accumulated these metals 131–563-fold (on a dry weight basis). Rhizo-filtration—the use of plant roots to remove heavy metals from aqueous medium is an emerging environment cleanup technology. However, most of the identified hyper-accumulators grow slowly on land, harvested only once per year and have a limited biomass yields. Therefore, their application for wastewater remediation is limited [12]. Literature review showed that few studies have reported the use of terrestrial hyper-accumulators to treat metal contaminated water.

S. alfredii has proven to be a Zn/Cd-hyper-accumulator [13,14] and Pb-accumulator [15] that efficiently propagates by asexual reproduction. It can be harvested three to four times around a year, with a biomass yield as high as 1800 kg hm^{-2} under favorable conditions in a growing season [13].

The objectives of the present study were to evaluate the potential of *S. alfredii* to treat synthetic wastewater containing a mixture of Zn, Cd, Pb and Cu and to investigate the mechanism of metal removal. Such an ability of *S. alfredii* may be employed to treat wastewaters like those from electroplating industry.

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2. Materials and methods

2.1. Plant material

Sedum alfredii was obtained from an old Pb/Zn mined area in Quzhou city of Zhejiang province, China. *S. alfredii* is a perennial and succulent herb with clumped population. Prior to use, the plant samples were clipped with shears to approximately 2–3 cm long from the tip leaves, with 3–4 pairs of leaves and 2–3 pairs of nodes, cleaned with tap water and grown in 3-L plastic containers having basic nutrient solution in a greenhouse.

2.2. Plant pre-culturing

The culture medium and growth conditions used to raise the nursery were according to Liu et al. [16] with slight modifications. Healthy and uniform plants were chosen and grown in 3-L black plastic containers in metal-free basic nutrient solution with the following composition: 2 mmol L^{-1} Ca $(NO_3)_2 \cdot 4H_2O$; 0.1 mmol L^{-1} KH_2PO_4; 0.5 mmol L^{-1} $MgSO_4 \cdot 7H_2O$; 0.1 mmol L^{-1} KCl; 0.7 mmol L^{-1} K_2SO4; 10 μmol L^{-1} H_3BO_3; 0.5 μmol L^{-1} $MnSO_4 \cdot H_2O$; 0.01 μmol L^{-1} $(NH_4)_6$ Mo_7O_{24}; 100 μmol L^{-1} Fe-EDTA. The pH of the nutrient solution was adjusted daily to 5.5 using 0.1 mol L^{-1} NaOH or 0.1 mol L^{-1} HCl. The nutrient solution was aerated continuously and renewed after every 4 days. The cultured plants were maintained in a greenhouse with controlled temperature (25 ± 5 °C) and day/night humidity of 70%/90%. After growth by 30 days, the new stems were clipped in a similar fashion as mentioned above and were cultured again for 45 days, and then used for the experiments.

2.3. Preparation of heavy metal loaded wastewater

The phytoremediation of four heavy metals (Zn, Cd, Cu and Pb) was investigated in the present study. They were added as cadmium sulfate ($CdSO_4 \cdot 8/3H_2O$), copper sulfate ($CuSO_4 \cdot 5H_2O$), zinc sulfate ($ZnSO_4 \cdot 7H_2O$) and lead sulfate ($PbSO_4$). The concentration of each metal was similar to those found in effluents of Zn-plating factories in China. The final nutrient and the heavy metal concentrations were listed in Table 1.

2.4. Hydroponics metal removal by *S. alfredii*

The raised plant samples from pre-cultures were grown in de-ionized water for 24 h and the roots were soaked in 20 mmol L^{-1} Na_2EDTA solution for 15 min to remove any adsorbed metals on root surface and then rinsed 3 to 4 times with de-ionized water to clean any adhered chemical substances.

Six plants were grown in each container receiving 2.5 L nutrient solution. Two controls were administered in the experiment along with a treatment. One of the controls was treated with heavy metals without any plants to observe heavy metal precipitation and the other control had plants without heavy metal addition to examine the effect of metal loading on the growth of *S. alfredii* in the aqueous medium. The treatment consisted of plants grown in heavy metal solution and each treatment was in triplicates. The containers were placed in a greenhouse under controlled temperature (25–30 °C) and day/night humidity of 70%/90%. The solution was aerated for 1 h at an interval of 4 h and the plants were harvested after growth of 7 days. At harvest, subsamples of plant roots from the treatment containers were soaked in 20 mmol L^{-1} Na_2EDTA (disodium ethylenediamine tetraacetate) solution for 15 min to remove metal ions adhering and adsorbing on the root surface (exclusion of adsorbed metals). The remaining samples of the plant roots from the treatment containers were rinsed 3 to 4 times with de-ionized water to remove metal ions adhering to the root surfaces keeping the adsorbed ones. The harvested plants were separated into leaves, stems and roots and oven dried at 70 °C for 72 h. Fresh and dry weights were recorded.

Table 1
The concentrations of metals in the treated water used in culture study.

Elements	Concentration in water	Added from reagents	Total
	mg L^{-1}		
Potassium	3.50	63.53	67.03
Nitrate	2.35	40.35	42.70
Calcium	28.45	28.57	57.02
SO_4^{2-}	30.34	134.40	164.74
Soluble phosphorus	0.25	4.00	4.25
Magnesium	3.22	6.00	9.22
Iron	0.03	0.11	0.14
Manganese	0.08	0.03	0.11
Copper	0.07	3.20	3.27
Zinc	0.04	19.20	19.24
Cadmium	0.00	11.24	11.24
Lead	0.01	0.52	0.53

2.5. Sampling and analysis

Water samples from growth medium (5 mL each) were collected from each container at intervals of 24 h and analyzed for Zn, Cd, Cu, and Pb concentrations using inductively coupled plasma-mass spectrometry (ICP-MS, Agilent, 7500a, Japan). Plant samples (0.2 g each) were also collected at the end of the experiment and analyzed for the heavy metal concentrations. The plant samples were dried in an oven at 70 °C for 72 h, and ground to <1 mm before analysis. About 0.1 g of each plant sample was digested with concentrated acid mixture (HNO_3:$HClO_4$, 5:1) at 200–220 °C [17]. The Zn, Cd, Cu and Pb concentrations in the digested solution were also analyzed by ICP-MS.

2.6. Data analysis

The weight of individual heavy metal in the plant was calculated as follows: each heavy metal weight in whole plant/dried whole plant weight = (heavy metal concentration in roots × weight of dried roots + heavy metal concentration in stems × weight of dried stems + heavy metal concentration in leaves × weight of dried leaves)/(dried roots + dried stems + dried leaves).

The weight of individual metal in a pot was calculated as: weight of heavy metal in whole plant/pot = (heavy metal concentration in roots × weight of dried roots + heavy metal concentration in stems × weight of dried stems + heavy metal concentration in leaves × weight of dried leaves) / pot.

The weight of metals adsorbed on root surface was calculated by difference of heavy metal concentration in root with and without soaking in 20 mmol L^{-1} Na_2EDTA solution for 15 min.

All data were presented as mean values of three replicates. A SPSS™ statistical software package (Version 11.0) was used for statistical analysis. One-way ANOVA was performed to determine whether the means were significantly different at $P<0.05$.

3. Results and discussion

3.1. Zinc

The initial Zn concentration in the control containers without plants was 19.24 mg L^{-1} that decreased to 17.12 ± 0.22 mg L^{-1} at the end of the experiment due to precipitation, with a removal rate of 11.02 ± 1.45%. Whereas, Zn concentration in the containers with plants decreased from 19.24 to 1.14 ± 0.16 mg L^{-1}, with a removal rate of 94.07 ± 0.83% (Table 2). Approximately, 91.84% of Zn reduction occurred within 3 days, while the negligible change in Zn concentration occurred in the remaining 4 days (Fig. 1a). The measured Zn concentrations in roots, stems and leaves of the plants from the