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Phytoremediation of Cd- and Pb-contaminated soil with biodegradable chelants by Brassica Juncea

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Abstract. Effect of the biodegradable chelants (Sophorolipid, Saponin, β -cyclodextrin and Polyaspartic acid) on phytoremediation of Cd- and Pb- contaminated soil by Brassica Juncea was investigated in this work. Especially, the effect of the kind and the adding dosage of the chelants was studied during the plant growth. Plants were grown in an environmental control system. The biomass of the whole plant was weighed, and the concentration of Cd and Pb in shoot and root were determined using ICP-MS. Consequently, the following matters have been obtained: (1) All the chelants had the excellent stabilization of Cd and Pb in root. (2) The chelants resulted in a sharply biomass loss. (3) From bioconcentration factor, the chelants were more beneficial for the translocation of Pb than Cd, especially Saponin.

1. Introduction

Soil contamination has been or is becoming serious in China. About 19.4 % arable soil has been polluted, and the soil contaminants mainly include Cadmium (Cd), Nickel (Ni), Copper (Cu), Arsenic (As), Mercury (Hg), Lead (Pb), DDT, PAHs and so on according to the report on the national general survey of soil contamination in April 2014. Among these soil contaminants, it is difficult and intractable to remediate the polluted soil by heavy metals because of their non-degradability and indefinite persistence in the environment. Cd and Pb are the most common heavy metals found at contaminated soils. Their accumulation in soils has been shown to cause a variety of environmental problems, including loss of vegetation, rice pollution, groundwater contamination, and Cd or Pb toxicity in food chain. Phytoremediation of heavy metals has the advantages of low-cost, environmental-friendly and minimal soil disturbance^[1-2]. An ideal plant for phytoremediation can be regarded as one with high biomass production and high tolerance for metal accumulation and translocation in the aboveground portion. In fact, most plants present either low biomass production, weak translocation, or lower metal accumulation capacity, which make them less suited for phytoremediation applications. Therefore, chemically enhanced agents, such as chelants, are applied to improve the phytoremediation efficiency of given plants as a viable strategy^[3-4]. The objective of this research is to investigate the ability of the four chelants including Saponin, Sophorolipid, β -cyclodextrin and Polyaspartic acid for remediation effect of Cd- and Pb-contaminated soil with Brassica juncea.

2. Experimental

2.1. Apparatus and Reagents. A plant environmental control system (RTOP-300D, TOP Instrument, China) was used to cultivate plant. An inductively coupled plasma mass spectrometer (ICP-MS) (NexIONTM 350X, PerkinElmer Co. Ltd., USA) was employed to determine the concentration of Pb



and Cd. Plant and soil samples were digested with a microwave digestion system (WX-6000, PreeKem Scientific Instruments Co. Ltd., China).

Metallic salts, $\text{Cd}(\text{NO}_3)_2$ and $\text{Pb}(\text{NO}_3)_2$, were purchased from Tianjin Guangfu Technology Development Co. Ltd. (China). Saponin and Sophorolipid were purchased commercially from Sigma-Aldrich, Inc. (Germany). β -cyclodextrin and polyaspartic acid were purchased from J&K Scientific Ltd. (China). All chemical reagents used were of analytical grade. Ultrapure water ($>18.2 \text{ M}\Omega$) was used throughout the work. The nutrient solution including 18.6 mg/L N, 5.6 mg/L P and 8.6 mg/L K was prepared by dissolving KH_2PO_4 , KNO_3 and $\text{CO}(\text{NH}_2)_2$ in ultrapure water, then used as fertilizer during the period of the plant cultivation.

2.2. Preparation of Cd- and Pb-contaminated Soil. The contaminated soil (50 mg/kg Cd and 500 mg/kg Pb) used in the present study was prepared by adding the solution containing two kinds of metallic salts to a soil “Expanded Vermiculite” from Liheng mineral processing plants in Lingshou (China), which has high moisture retention capability, air capacity and nutrient preserving capability. Physical and chemical characteristics of the Expanded Vermiculite are listed in Table 1. Their measure methods are the same as shown in our previous paper^[5].

Table 1 Physical and chemical characteristics of the Expanded Vermiculite

Parameters	pH (H_2O)	pH (KCl)	Moisture content %	TOC %	CEC cmol/kg	TN g/kg	TK g/kg	TP mg/kg	Cd mg/kg	Pb mg/kg
Value	6.78	5.18	1.64	2.2	49.64	0.38	22	1.2	0.47	Not detected

2.3. Uptake of metals (Cd and Pb) by Brassica Juncea. The contaminated soil was used to fill 500 mL plastic pots (280 g soil per pot) and moistened with ultrapure water to reach approximately 60 % water holding capacity. Each pot was planted with five Brassica Juncea seeds, and germinated in the plant environmental control system with a 12 h day (27 °C) under the illumination (7500 lx) and a 12 h night (20°C). The nutrient solution above-mentioned was used as basal fertilizers to supply nitrogen (N), phosphate (P) and potassium (K) for plants. Pots were watered every day with the nutrient solution and ultrapure water according to water loss by weight to maintain 60 % water holding capacity. Following seedling emergence, the pots were thinned to one plant per pot. After 10 days of growth, Sophorolipid, Saponin, β -cyclodextrin and Polyaspartic acid were added to the soil surface as solutions at 0.2, 0.5 and 1.2 g/kg soil, and the treatments were marked as H-1, H-2, H-3, Z-1, Z-2, Z-3, β -1, β -2, β -3, J-1, J-2, J-3, respectively. The pots with no chelants were kept as a blank, and each treatment was performed in duplicate with a random block design, thus, 26 pots were used.

2.4. Extraction of Plant Samples and Determination of Cd and Pb. Plants were harvested after 60 days growth by cutting stems 1 cm above the soil surface. The above parts were considered as shoot and the below parts were as root. Shoot and root samples were washed, dried, and then ground. Dried plant samples were digested in a microwave digestion system for metal analysis. Cd and Pb were determined by ICP-MS.

3. Results and Discussion

3.1. Concentrations of Cd, Pb in Plant. The concentrations of Cd, Pb in shoot and root are shown in Table 2. For the concentrations of Cd in shoot, only Saponin shows the enhancement among four chelants. Cd concentration is 125.8 mg/kg with the adding dosage of 0.5 g/kg Saponin, that is, the maximum enhanced rate reaches 19.2 %. The Cd concentrations in root are significantly enhanced by adding the chelants comparing with that in the blank. They are up to 232.1 and 328.8 mg/kg with 0.5 g/kg Sophorolipid and 1.2 g/kg Polyaspartic acid, and 1.8 and 2.5 folds of the blank, respectively. For each chelants, the highest is obtained with the middle adding amount (0.5 g/kg) except for polyaspartic acid. These suggests that the chelants have the excellent stabilization of Cd in root.

The concentrations of Pb in shoot increase with the increase of the adding dosage of the chelants except for Saponin, and most of them are between 15 ~ 24 mg/kg. In addition, the concentrations of Pb adding 1.2g /kg Sophorolipid and 0.5 g/kg Saponin are much higher than the others, more than 2 folds of that in the blank, 34.81 mg/kg and 36.93 mg/kg, respectively. The concentrations of Pb in root is the same enhancement with Cd in root, all the chelants have the ability to enhance Pb stabilization in root because the Pb concentrations in 7/9th treatment pots are higher than that of the blank. It can be found that the enhanced rate of Sophorolipid is the highest, and β -cyclodextrin is the lowest by comparing each other. The Pb concentrations are much lower than those reported regardless of in shoot and root, which may be attributed to the lower Pb concentration in soil [6].

Table 2 Contents of Cd,Pb in shoot and root of plants (mg/kg dry weight)

Treatment	C _{Cd} in shoot	C _{Cd} in root	C _{Pb} in shoot	C _{Pb} in root
t				
H-1	84.82	142.6	16.66	305.8
H-2	94.87	232.1	18.81	195.6
H-3	100.5	128.0	34.81	325.8
Z-1	109.8	153.3	15.07	232.3
Z-2	125.8	186.5	36.93	228.9
Z-3	123.2	160.4	23.50	132.6
β -1	81.95	102.8	16.37	169.5
β -2	89.48	155.5	16.83	130.9
β -3	86.48	133.6	18.31	173.6
J-1	68.96	136.3	14.56	164.2
J-2	76.38	153.3	17.40	175.4
J-3	94.35	328.8	18.70	212.5
B	105.5	132.6	15.88	166.8

3.2. Effect of Chelants on the Biomass. The shoot and root biomass (dry weight) of *Brassica juncea* in all treatments after 60 days growth are shown in Fig. 1 and Fig. 2. In the presence of chelants, the biomass of *Brassica juncea* exhibits reduction compared with that of the blank, and the shoot and root biomass of the treatment adding 1.2 mg/kg sophorolipid is only about 1/4 and 1/5 of that of the blank, respectively. The reduction amount of the shoot and root generally increases with the increase of the adding dosage for each chelant, which is caused by the increase of available heavy metal (Pb and/or Cd) inhibiting the plant growth. It is also found that the reduction difference is clear for the different chelants, it is the maximum for Saponin and the minimum for β -cyclodextrin. The biomass loss of the shoot is consistent with that of the root except for Saponin, and the biomass is proportional to the water content in *Brassica juncea* (Data are not shown here). From these results, the biomass depends on the kind and the adding dosage of the chelants, but the reduction of the biomass is undesirable for the phytoremediation of heavy metal polluted-soil, which can be solved by using plant regulators [7-8].

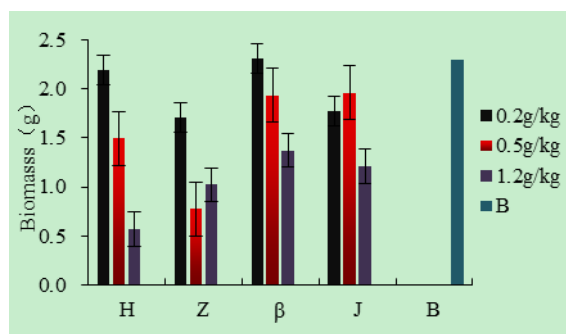


Fig. 1 Shoot biomass

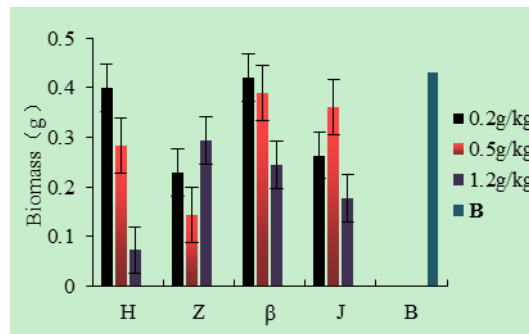


Fig. 2 Root biomass

3.3. Bioconcentration Factor (BCF) of Cd and Pb in Plant. To learn the translocation of heavy metals studied from root to shoot by *Brassica juncea*, BCF is calculated according to the ratio of the concentrations of Cd or Pb in shoot to those of Cd or Pb in root. BCF of Cd and Pb is shown in Fig. 3 and Fig. 4. Although BCF of Cd is much higher than that of Pb, BCF of Pb is obviously elevated under adding the chelants comparing with the blank. BCF of Cd treated by the chelants (except for 0.2 g/kg β -cyclodextrin) is lower than that in the blank. These show that no ideal Cd enhanced accumulation is obtained in *Brassica juncea* with the chelants. BCF of Pb treated with the four chelants is more than that in the blank to some extent. Especially BCF is 0.18 with 1.2 g/kg Saponin, and equal to 1.8 times of that of the blank. These results suggest that Saponin is more effective for the accumulation of Pb in shoot than other chelants, which is more beneficial for the translocation of Pb with adding Saponin.

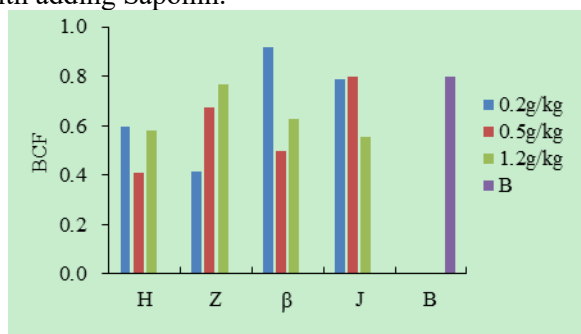


Fig. 3 BCF of Cd

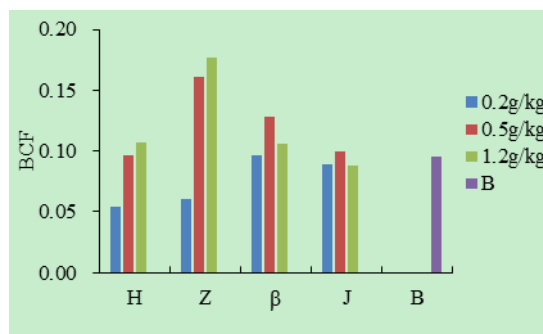


Fig. 4 BCF of Pb

4. Conclusions

Effect of Sophorolipid, Saponin, β -cyclodextrin and Polyaspartic acid on phytoremediation of Cd and Pb in soil by *Brassica Juncea* was investigated. The results show that the effect of the phytoremediation varies with the kind and the dosage of adding the chelants. Further study is necessary in order to evaluate whether the chelants may represent an effective tool for the removal of metals from a Pb- and Cd- polluted soil for practical use, such as applicable concentration of heavy metals in soil, adding method of the chelants and optional plants.

Acknowledgments

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