Phytoextraction potential of wetland plants for Copper in Water Bodies

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Abstract

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Copper is the most common heavy metal contaminant in the environment. Wetland construction engineering and technology have been used to control water pollution due to their low cost and efficiency and the hydrophytes have been the most important constituents of wetland construction. In this experiment, during April of 2014, Cu^{2+} accumulation content in different parts of *Acorus calamus* and *Phragmites australis* were investigated based on hydroponic experiments of different Cu^{2+} concentration solutions. Cu^{2+} concentrations in the water body were 0, 10, 25, 60, 100, 200 and 500 mg/l, respectively. The results showed that there were significant Cu^{2+} concentration differences between the above- and below-ground parts of *Acorus calamus* and *Phragmites australis*. Cu^{2+} content in the above- and below-ground parts of wetland plants increased with hydroponic solution Cu^{2+} concentrations, resulting in a significantly positive correlation between Cu^{2+} content and concentrations of hydroponic solutions. There was a significant difference in Cu^{2+} content in the wetland plants under all hydroponic solution Cu^{2+} concentrations. *Acorus calamus* exhibited the greatest Cu^{2+} accumulation in above- and below-ground parts. *Acorus calamus* and *Phragmites australis* can be selected for application on the phytoremediation of water polluted by heavy metals due to their excellent Cu^{2+} accumulation ability.

Keywords: *Acorus calamus*, *Phragmites australis*, Cu^{2+}, accumulation.

Resumen

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El cobre es el metal pesado contaminante más común en el medio ambiente. La ingeniería y tecnología de construcción de humedales se han utilizado para el control de la contaminación del agua debido a su economía y eficiencia, y las hidrófitas han sido los componentes más importantes de la construcción de humedales. En este estudio, llevado a cabo en abril de 2014, se investigó la acumulación de Cu^{2+} en diferentes partes de Acorus calamus y Phragmites australis mediante métodos hidropónicos de soluciones con diferentes concentraciones de Cu^{2+}. Las concentraciones de Cu^{2+} en el cuerpo de agua fueron de 0, 10, 25, 60, 100, 200 y 500 mg/l. Los resultados mostraron que existen diferencias significativas en la concentración de Cu^{2+} entre las partes de Acorus calamus y Phragmites australis que están por encima y las que están por debajo del suelo de. El contenido de Cu^{2+} en las partes de arriba y debajo del suelo de plantas de humedal aumentó con solución hidropónica de Cu^{2+}, dando por resultado una correlación significativamente positiva entre el contenido de Cu^{2+} y las concentraciones de soluciones hidropónicas. Hubo una diferencia significativa en el contenido de Cu^{2+} en las plantas de humedal con todas las concentraciones de Cu^{2+} en solución hidropónica. Acorus calamus exhibió la mayor acumulación de Cu^{2+} en las partes por encima y por debajo del suelo. Acorus calamus y Phragmites australis se pueden seleccionar para su aplicación en fitoremediación de aguas contaminadas por metales pesados, debido a su excelente capacidad de acumulación de cobre.

Palabras clave*: Acorus calamus, Phragmites australis, Cu^{2+}, acumulación.*

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Introduction

Heavy metals are harmful to aquatic ecosystems, and remain in the environment for a long time (Ameh & Akpah, 2011; Bissenbaev, Ishchenko, Taipakova, & Saparbaev, 2011; Tanhan, Kruatrachue, Pokethitiyook, & Chaiyarat, 2007). Copper is a toxic heavy metal pollutant that quickly accumulates in animals and humans through the food chain, which seriously affects the metabolism of the human body. The human body releases Cu^{2+} very slowly, causing damage to bodily organs that are irreversible. Therefore, Cu^{2+} pollution treatment has become an urgent subject. Traditional methods of treatment to heavy metal pollution in soil mainly use the mixing of soil, leaching method, chemical modifiers, and so on. Not only are these physical and chemical methods expensive, they cannot be applied into small areas and cause second pollution. Such methods cannot fundamentally solve the Cu2+ pollution in soil. (He, Huang-Xiao, & Chen, 2011; Visioli & Marmiroli, 2013; Mirza, Hossain, & Masayuki, 2010). In recent years, people have found the bioconcentration ability of some plants to heavy metals. It is a high efficiency, environmental protection and cost control measures to use such plants as phytoremediation of heavy metal pollution. This treatment measure has very broad application prospects (Engelen, Sharpe-Pedler, & Moorhead, 2007; Chaumont *et al*., 2012; Rajkumar, Sandhya, Prasad, & Freitas, 2012; Zhou *et al*., 2007).

The calamus (*Acorus calamus L.*) and reed (*Phragmites australis*) are common across China's natural wetlands. The high Cu^{2+} absorption features of these two plants species provide scientific basis for phytoremediation of heavy metal pollution in water bodies.

Materials and methods

Acorus calamus and *Phragmites australis* were collected from the Fuxi River basin of Sichuan Province, China.

In April of 2014, the experiment was carried out with hydroponic cultivation in a ventilated plastic shed. Foam boards were placed on the clean plastic barrels with three holes to serve as a plant carrier. Three plants of similar height were selected from each species and transplanted into the foam carriers, and the root of the plants were immersed in the nutrient solution with the modified Hoagland formula: 945 mg/l $Ca(NO_3)_2 \cdot 4H_2O$, 506 mg/l KNO_3, 80 mg/l NH_4NO_3, 136 mg/l KH_2PO_4, 493 mg/l $MgSO_4$, 2.5 ml Fe-EDTA, 5 ml trace element solution (0.83 mg/l KI, 6.2 mg/l H_3BO_4, 22.3 mg/l $MnSO_4$, 8.6 mg/l $ZnSO_4$, 0.25 mg/l Na_2MoO_4, 0.025 mg/l $CuSO_4$, 0.025 mg/l $CoCl_2$), pH = 6.0.

After two weeks of continued growth of the plant's, the $Cu(NO_3)_2$ was added to the aqueous solution according to the concentration gradient: 7 concentration process of 0, 10, 25, 60, 100, 200 and 500mg/l. Each of concentration process was repeated six times.

The well-growing plants were collected after three weeks and divided into above- and below-ground parts. They were washed with distilled water and deionized water twice, treated with deactivation enzymes at 105°C, and dried to a constant weight for eight hours at 70°C. After drying, weighing, grinding and digestion, the plant samples were determined Cu^{2+} concentration by atomic absorption spectrophotometer.

Copper retention rate was calculated by formula (1) (Xia & Shu, 2001).

$$Cu^{2+} \text{ retention rate}(\%) = (C1\text{-}C2)/C1 \times 100 \quad (1)$$

C1: Cu^{2+} content in underground part of plants, mg/kg.
C2: Cu^{2+} content in aboveground part of plants, mg/kg.

The experimental data was analyzed by variance analysis (ANOVA), LSD test, and correlation analysis between Cu^{2+} concentration of solution and enrichment concentration of plants by Origin 9.0 software.

Results and discussion

The Two-way ANOVA analysis of Cu^{2+} contents of the two emergent plants is as follows: The

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plant species and the Cu^{2+} concen tration process significantly affected the accumulation of copper in plants. The analysis results were showed in Table 1.

Calamus and reeds showed very significant differences at seven concentration processes for the Cu^{2+} accumulation capacity in different emergent plants. The Cu^{2+} accumulation content in different plant species (calamus and reeds) has significantly affected ($P < 0.01$). In addition, the interaction of different Cu^{2+} concentrations and plant species also significantly affects Cu^{2+} enrichment capacity of wetland plants ($P < 0.01$).

Table 2 shows Cu^{2+} accumulation in *Acorus calamus* and *Phragmites australis*.

Figure 1a and 1b indicated that the Cu^{2+} accumulation contents in calamus and reed showed significant differences. In general, Cu^{2+} accumulation amounts in calamus were significantly more than in the reed. The calamus has a stronger ability to absorb and transport Cu^{2+} than reed does.

The experiment results showed that the Cu^{2+} accumulation contents of calamus were significantly more than that of reed in both the above- and below-ground parts. The Cu^{2+} enrichment amounts in the underground parts of the two plant species were not significantly different at low Cu^{2+} concentrations (10 mg/l and 25 mg/l). Yet, the Cu^{2+} enrichment amounts in calamus were significantly more than that

Table 1. The ANOVA analysis of Cu^{2+} accumulation

	F	P
Concentration process	33.13**	<0.001
Plant species	5.99*	0.0028
Interaction	5.60*	0.0038

* Notes: ** indicates a significant difference at p = 0.001; *indicates a significant difference at p = 0.01

Table 2. Cu^{2+} accumulation in Acorus calamus and *Phragmites australis*.

Plant part	**Cu^{2+} concentration (mg/l)**	***Acorus calamus* (mg/kg)**	***Phragmites australis* (mg/kg)**
Aboveground	0	7.4 ^{a}	122.79 ^{b}
	10	115.09 ^{a}	159.92 ^{b}
	25	681.23 ^{a}	143.61 ^{b}
	60	899.17 ^{a}	223.69 ^{b}
	100	4 107.53 ^{a}	198.39 ^{b}
	200	6 208.5 ^{a}	264.75 ^{b}
	500	4 206.9 ^{a}	349.73 ^{b}
Underground	0	37.85 ^{a}	113.17 ^{b}
	10	282.73 ^{a}	236.54 ^{b}
	25	955.3 ^{a}	768.41 ^{b}
	60	2 980.97 ^{a}	373.75 ^{b}
	100	8 107.53 ^{a}	429.22 ^{b}
	200	4 271.03 ^{a}	379.52 ^{b}
	500	2 198.27 ^{a}	448.83 ^{b}

* Notes: Values with different letters in the same element of the same species and same column indicate a significant difference at $p = 0.05$ according to the LSD test.

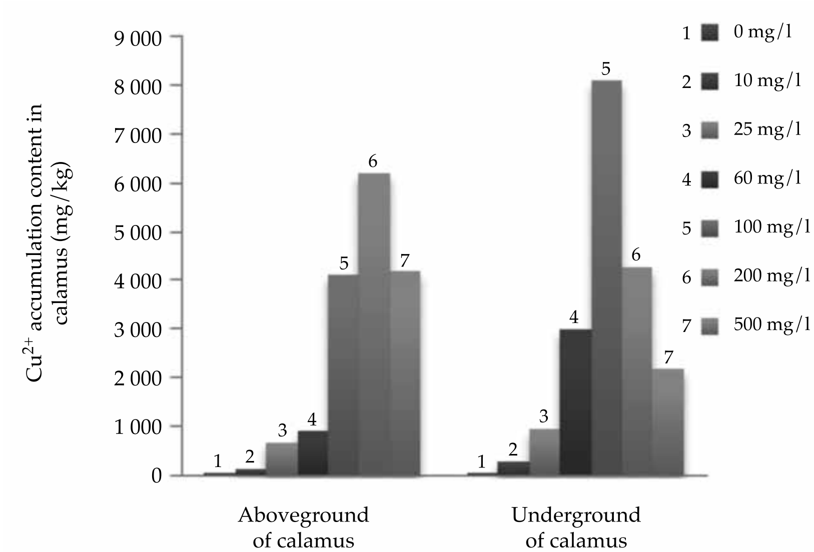

Figure 1a. The Cu^{2+} accumulation mean content in *calamus*.

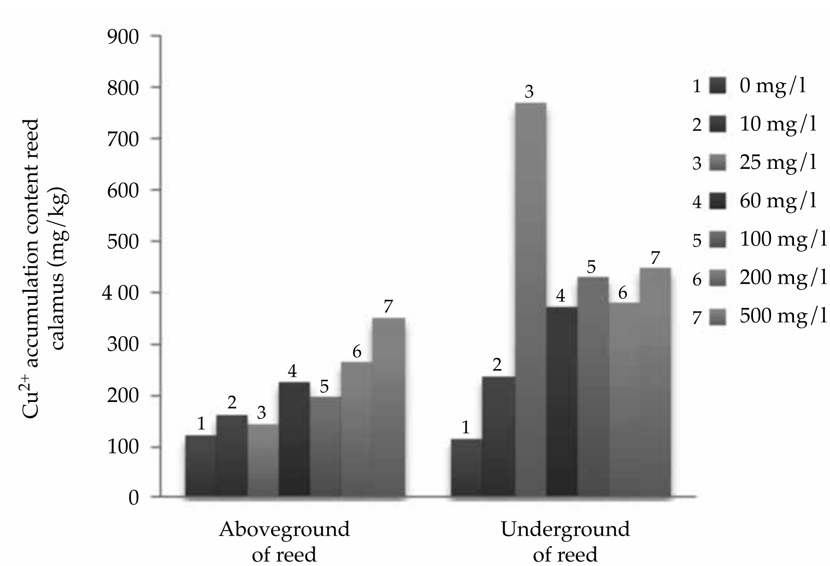

Figure 1b. The Cu^{2+} accumulation mean content in reeds.

in the reed at higher Cu^{2+} concentrations (60 mg/l, 100 mg/l, 200 mg/l and 500 mg/l). The aboveground parts of plants were similar to belowground parts

Overall, in lower concentrations (10 and 25 mg/l), Cu^{2+} accumulation quantity in Acorus calamus and Phragmites australis were similar at the 0.05 level. Cu^{2+} accumulation quantity in the above- and below-ground reed parts were significantly different at higher Cu^{2+} concentrations, but the same parts of the calamus were not significantly different. The two plant species showed different accumulation abilities at different Cu^{2+} concentrations; calamus accumulated more Cu^{2+} than the reeds did, as shown in Figure 1a and 1b.

The accumulation amount of Cu^{2+} in calamus and reed increased with the increase of pollutant concentration. At medium and low concentrations, the contents in the underground part of calamus were more than that in the aboveground part, and the average retention rate was more than 50%. However, it was the opposite at high concentrations (200 and 500 mg/l); the content in the underground parts was lower. The calamus has a strong ability to enrich and migrate Cu^{2+}. The content of underground part of reed was higher than that of aboveground parts at six concentration processes, indicating that the root system of reed had a strong retention effect on Cu^{2+}, and the average retention rate was more than 40% (Table 3). There was no significant difference in mean retention rate between the two species.

The results of the correlation analysis of Cu^{2+} contents in the aboveground parts of calamus and hydroponic solution Cu^{2+} concentration showed that a maximum Cu^{2+} content was achieved when the concentration was 200mg/l. The variation curve of Cu^{2+} content in the underground and aboveground parts were similar. The optimum enrichment concentration for copper was 100mg/l (Figure 2).

Table 3. Cu^{2+} retention rate of calamus and reed.

Cu^{2+} concentration(mg/l)	*Acorus calamus* (%)	*Phragmites australis* (%)
0	80.45	-8.50
10	59.29	32.39
25	28.69	81.31
60	69.84	40.15
100	49.34	53.78
200	-45.36	30.24
500	-91.37	22.08

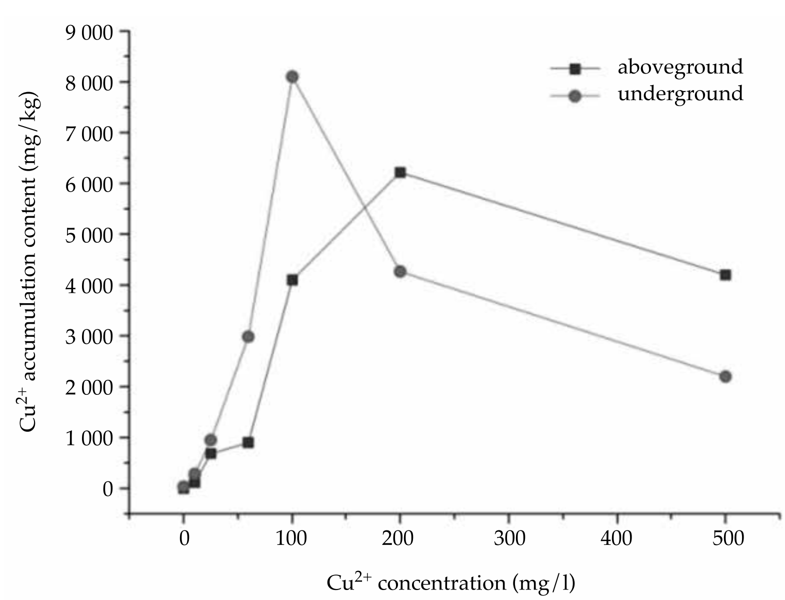

Figure 2. The correlation between Cu^{2+} accumulation content in *Acorus calamus* and Cu^{2+} concentration.

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Cu^{2+} accumulation contents in plants reaches a maximum with the increase of Cu^{2+} concentration in hydroponic solution. Cu^{2+} accumulation contents in plants decreased if the concentration exceeded the optimum enrichment concentration, indicating that the toxicity of heavy metals to plants has exceeded its tolerance, inhibiting plant growth and halting the accumulation of Cu^{2+}.

The Cu^{2+} enrichment amount in the aboveground of reed increased with the increase of Cu^{2+} concentration in the solution through the correlation analysis curve (Figure 3). Cu^{2+} accumulation content showed slight fluctuations at low concentration processes (10, 25, 60 and 100 mg/l), but Cu^{2+} accumulation significantly increase as the hydroponic solution of Cu^{2+} concentration increased. Therefore, *Phragmites australis* showed good Cu^{2+} tolerance. Cu^{2+} content in the underground parts of reed gently increased as hydroponic solution Cu^{2+} concentration increased, showing a positive correlation relationship. The enrichment amount of Cu^{2+} in the underground part was significantly more than that in the aboveground part. As can be seen from Figure 3, Cu^{2+} accumulation contents in the underground portion of reed fluctuated under low Cu^{2+} concentrations. As Cu^{2+} content increased, Cu^{2+} content stabilized and Cu^{2+} content reached its maximum in reeds.

The absorption of copper by calamus was higher than that of reed at seven concentration processes, but reed tolerance to high Cu^{2+} concentration was higher than calamus. Therefore, calamus is suitable for the ecological restoration of plants for medium and low concentration of heavy metal polluted water bodies.

According to the results of correlation analysis between Cu^{2+} content in plants and Cu^{2+} concentration in the hydroponic solution, the Cu^{2+} accumulation content(y) in aboveground and underground parts of *Acorus calamus* and *Phragmites australis* increased as Cu^{2+} concentration (x) increased; there was a significant positive correlation (Table 4). Thisindicated that the concentration of heavy metals in the growing medium plays an important role that impacts plant absorption and heavy metal accumulation.

Cu^{2+} primarily accumulates in plant roots (Stolts & Gregor, 2002). This is because Cu^{2+} in roots is the primarily forms from precipitation. Additionally, there is ionic and complex states

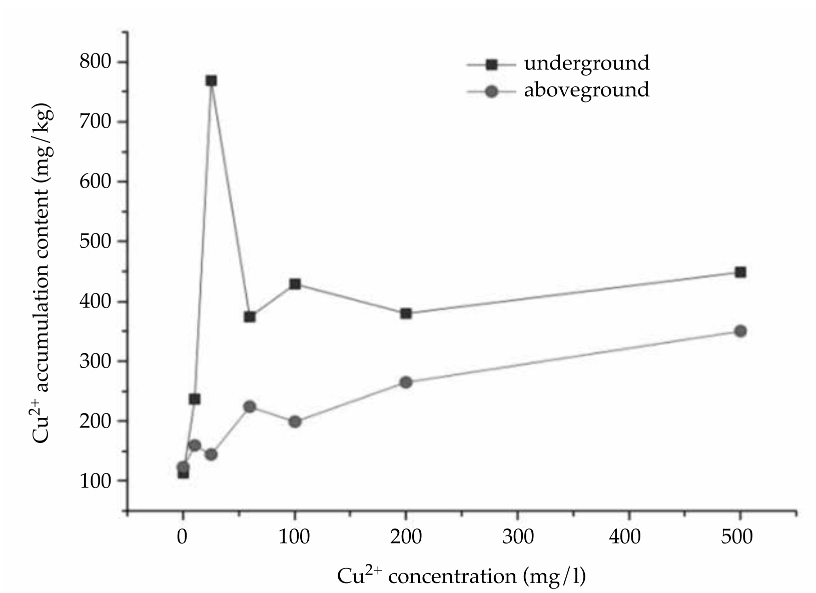

Figure 3. The correlation between Cu^{2+} accumulation content in *Phragmites australis* and Cu^{2+} concentration.

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Table 4. Correlation of Cu^{2+} accumulation content in copper-contaminated water to wetland plants.

Plant	Plant part	Correlation Function	R^2
Acorus calamus	Aboveground	$4.4082 \times 10\text{-}^{16} x^{22.1631x^{-0.18477}}$	0.97386
	Underground	$2.7194 \times 10^{-16} x^{25.4253x^{-0.20935}}$	0.93498
Phragmites australis	Aboveground	$125.1777 + 7.04417x^{0.55753}$	0.85224
	Underground	$463.1011 - 370.54424x^{0.88195}$	0.45869

of Cu in the plant saps. It is difficult to transport Cu^{2+} in plant roots due to retention, passivation, or precipitation. Cu^{2+} accumulates in the root surface mainly in the form of microcrystals on cell walls. Plants accumulated them in the roots, which prevents harmful ions from inhibiting photosynthesis. In this study, Cu^{2+} accumulation quantities in the underground parts of the two wetland plants were significantly greater than those in the aboveground parts (Ren, Tao, & Yang, 2009).

Conclusions

The two wetland plant species showed strong Cu^{2+} enrichment abilities at different Cu^{2+} concentration conditions, both in under-ground microcrystals on cell walls. ons from and aboveground parts. The contents of Cu^{2+} in calamus and reed were significantly different at the same Cu^{2+} concentration. Among same type of wetland plants, while below 200 mg/l, Cu^{2+} accumulation increased as the Cu^{2+} concentration in the solutions increased.

Because they have the ability to effectively-absorb and enrich copper, both Acorus calamus and *Phragmites australis* can be used as selective species for phytoremediation of contaminated water bodies with heavy metal. The *Phragmites australis* has better tolerance to heavy metals Cu^{2+}, and compared to other plants, the *Acorus calamus* has stronger absorption and accumulation Cu^{2+} abilities (Table 5). Overall, *Acorus calamus* is a better heavy metal contaminated water remediation plant than the *Phragmites australis*.

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Table 5. Possibilities accumulation ability of several plants for Cu^{2+} (Zhang, 2014).

Family	Plant	Possibilities of Cu^{2+} accumulation
Convolvulaceae	*Ipomoea alpina*	12 300 mg/kg
Scrophulariaceae	*Lindernia perennis*	9 322 mg/kg
Labiatae	*Gutenbergia katangense*	8 356 mg/kg
Cyperaceae	*Bulbostylis mucronata*	7 783 mg/kg
Amaranthaceae	*Pandiaka metallorum*	6 260 mg/kg
Leguminosae	*Vigna dolomitica*	3 000 mg/kg
Asteraceae	*Anisopappus davyi*	2 889 mg/kg
Pottiaceae	*Didymodon constricta*	1 962 mg/kg (Peng & Zhang, 2007)

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