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Heavy Metal Sorption by Aquatic Plants in Taiwan

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Toxic heavy metal contamination frequently occurs in rivers, estuaries, wetland, coastal areas and other ecosystems. For example, the Erh-jen Chi and its estuary in Taiwan were reported to have high metal contamination (Lee, 1996). Metal contamination sources are typically derived from industrial waste discharge, sewage effluent, and harbor activities. (Bervoets et al., 1994) Conventional re-mediation methods, such as precipitation, chemical oxidation or reduction, ion exchange, filtration, membrane technologies or evaporation process, are generally ineffective for removing some metals in an aquatic system. Incontrast, aquatic plants provide a viable alternative for metals remediation if proper disposal of the spent plants can be employed. (Jackson et al., 1994) Among the merits of this phyto-remediation process include its low energy, low cost, efficient in low level contaminants, metal recoverable, flexible on treating contaminants either on-site or in a phytoreactor. (Sajwam et al., 1994; Dani et al., 1996) Furthermore, the affinity for calcium and magnesium by these biosorbents are often quite low (Ramelow et al., 1992). This feature could make them highly competitive with ion-exchange resins in hard water.

Aquatic plants in relation to their ability to sequester heavy metals have received extensive interest (Gupta and Chandra, 1994; Muramoto and Oki, 1983; Sela et al., 1988; Seal et al., 1989). This interest has focused primarily on aquatic bryophytes (mosses and liverworts) as biomonitors of heavy metal pollution (Mouvet et al., 1993; Chawia et al., 1991; Raphael et al., 1990). Satake et al. (1990) found 2.4% lead was adsorbed in aquatic liverworts when it was exposed to 20 ppb metal solution. The uptake of lead ions by liverworts was reported as rapid as 300 µg/g biomass/hr in a laboratory experiment (Guillizoni, 1991). However, Satake et al. (1990) also reported Hg uptake rates by liverwart growing in an acid stream at medium concentrations of 0.6ppb. The adsorption capability of mercury was found only to be 10 to 30 µg/g/day.

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The over 30-fold differences in adsorption capability could be attributed to the type of plant or metal species, metal concentrations and specific experimental conditions (laboratory vs. field measurements).

This study screened twenty one strains of local aquatic plants to search for promising candidates in phyto-remediation process for metal removal. This studyalso examined kinetic and equilibrium adsorption characteristics of the promising aquatic species. Different ionic metal strength, size of the adsorbent and freshness and source of the plants, were studied to provide information for designing a phytoreactor or a constructed wetland to remove heavy metals of copper, zinc, lead, cadmium and nickel.

METHODS AND MATERIALS

Weighedamounts of blotted dry plant biomass (about 1 gram) were added to known metal solutions yielding 50 mL total volume in 120 mL Erlenmeyer flasks equipped with screw caps and, then, incubated in a shaker for 120 minutes. The pHs of the contact solution both before and after the experiments were measured. Initial pH for the metal solution was adjusted to 5 with NaOH. (Wang et al, 1998) Twenty one plant species shown in Table 1 were used for initial screening. The contacting experiments were performedby cutting plant biomass into small pieces (0.5 cm^2) . Experiments were also performed with uncutted biomass (whole plant without root) to determine the shape and size of biomass on metal adsorption. All experiments without a buffer solution were performed in triplicate, while those experiments with a buffer solution were performed in duplicate. At the end of the experiments, the contents of the flasks were strained to separate the biomass from the supernatant. The supernatant solution was measured with a Hitachi Model Z-8000 Polarized Zeeman Atomic Absorption Spectrophotometer. The biomass was then blotted dry before digestion with nitric acid in a microwave digester (MLS-1200 Mega Milestone). The protocols used were validated through exploration of the key variables: time, metal concentration (initial and final) and metal content in the biomass. Both inert (e.g. dried in oven at 60 °C), and living biomass samples were used in above experiments to establish adsorption isotherms under specified pH and ionic strength. The amounts of metal adsorbed and dried weight of biomass were used to calculate specific adsorption capability (metal adsorbed/ driedweight of biomass). Most of the biomass was collected from southern Taiwan, such as Tsen-Wen Dam, Tainan, Cheng-thing lake, Kaohsiung, Tong-Kung River, Sunfeng Aquatic Plants & Tropical Fish Farm, Tainan. The water used in all the experiments was purified using Milli RO Plus/Milli Q water purification devices. Metal standards were purchased from Merck. Next, the stock buffer solutions were prepared by

1020 µL glacial acetic acid and 5.745 g sodium acetate and diluting to 100mL with Milli Q water. Finally, standard material (aquatic plant, *Lagarosiphon major,* CEC Community Bureau of Reference B.C.R. Nr. 60) was used as a control sample for quality control.

Table 1. Aquatic plants screened for metal adsorption capability in this research.

Scientific Name	Code-name
Ammania gracilis	amm.
Cabomba carolinianqa	ca.
Ceratopteris thalictroides (L.) Brongn.	tha.
Coccoloba uvifera (L.) L.	uv.
Echionoborus grandifolius	gr.
Hydrocotyle leucocephala	le.
Hygrophila polysperma	pol.
Limmophila aquatica	aqu.
Ludwigia oalustris	pa.
Ludwigia ovalis Miq.	ov.
Ludwigia peruensis	per.
Myriophyllum elatinoides	ela.
Myriophyllum scabratum	sea.
Myriophyllum spectaum	spe.
Myriophyllum propiunum	pro.
Najas graminea Del.	gra.
Najas sp.	hyg.
Potamogeton crispus L.	cri.
Potamogeton malaianus Miq.	mal.
Rotala aquatica	in.
Vallisneria spiralis	sp.

RESULTS AND DISCUSSION

Experiments were designed to screen plants for their ability to remove metal from contaminated water. Thirty two contacting experiments were performed to screen twenty one aquatic plants for their adsorption capability. The time course [exposure concentration vs. contacting time] experiments was initially performed with plants *Najas graminea* Del. to determine the optimal contact time. The result showed that there were no significant increase in metal adsorption by the biomass after the first 60 minutes. Most screening experiments were conducted with lead and zinc with initial concentrations of 2 mg/L and 1 mg/L, respectively. Figure 1 and Figure 2 depict the screening results of each specific plant. Eachplant exhibiteddifferent adsorption capability. Three of those plants collected, i.e. *Najas graminea* Del. (GRA), *Myriophyllum scabratum* (SCA) and *Myriophyllum elatinaides* (ELA), had higher adsorption capacity than others. The adsorption isotherms of copper, zinc, lead, cadmium and nickel onto these three plants, GRA, SCA and ELA, were therefore performed with initial metal concentration ranged between 10 to 250 mg/L at pH5 and 60 min. contact time.

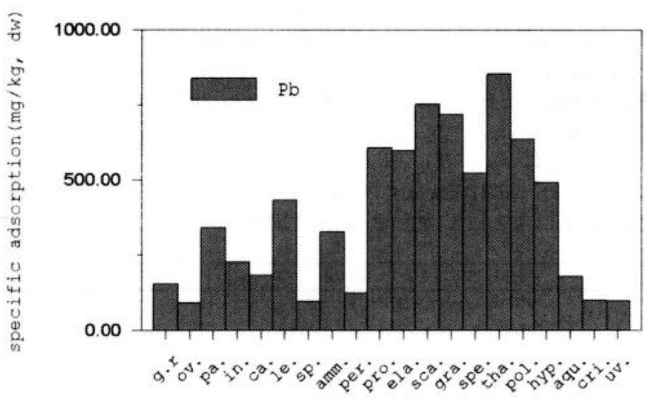

Figure 1. Specific adsorption of lead with 2 mg/L initial concentration.

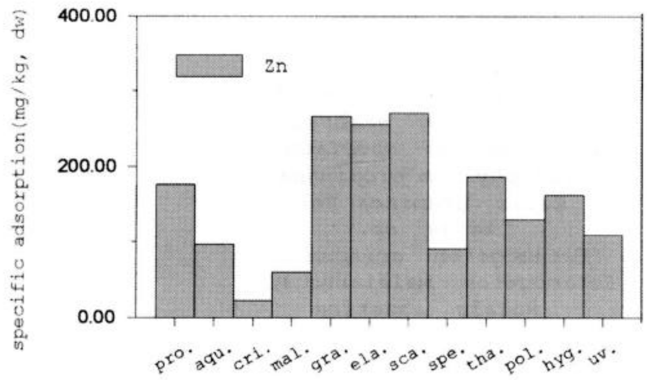

Figure 2. Specific adsorption of zinc with 1 mg/L initial concentration.

Langmuir plots were executed to obtain the maximumadsorption, Q_{max} and adsorption intensity, b, in the linearized form of isotherm:

$$\frac{C}{Q} = \frac{C}{Q_{\max}} + \frac{1}{bQ_{\max}}$$

Table 2 summarizes the results of all fivemetals for the three aquatic plants without buffer solution. In general, the adsorption capability of *Myriophyllum scabratum* and *Myriophyllum elatinaides* were slightly higher than that of *Najas graminea* Del. in both capacity and concentration. However, *Najas graminea* Del. was most easily cultured, among the three plants tested above and, therefore, was adopted for further studies discussedhereafter. Table 3 summarizes the adsorption results of five metals on *Najas graminea* Del. buffered at pH 5.0. For comparison, Figure 3 presents the isotherms of lead on *Najas graminea* Del. buffered at pH 5.0 and unbuffered (initial pH 5.0). The increase of ionic strength caused by the buffer solution reduced the adsorption capability of the biomass. In addition, the maximum adsorption capacity of buffered culture was decreasedby about 50% and the adsorption intensitywas reducedto about 5% of the un-buffered one. Figure 4 compares freshly sampled and three months lab-cultured samples. Notably, their adsorption characteristics were not significantly different. Also, the biomass after dried at 60 °C was used to test possibility of using dried biomass as sorbent. From Figure 5 and Table 4, we concluded: (i) while wholeplant (un-cut) was used, no significant difference in the maximum adsorption capacity was observed; the value of b, however, was reduced in an extent of ca. 50% of cutted sample; and (ii) roughly 30% decrease in the maximum adsorption capacity and adsorption intensity was observed, based on dried (mainly adsorption) versus fresh biomass (adsorption and uptake by aquatic plants) used as sorbent.

Table 2. Summary results of the adsorption characteristics of five metals of three aquatic plants without buffer (initial pH=5, final pH=6.3-7.0).

		Cu	Zn	Pb	Cd	Ni
gra.	Qmax(mg/g)	94.6	20.3	56.8	15.7	17.5
	b(L/g)	1.67	0.73	6.78	1.29	0.50
	blank(mg/kg)*	10	24	20	16.3	11
	BCF(max.)**	1.78E+03	2.18E+03	9.64E+03	1.16E+03	7.00E+02
sca.	Qmax(mg/g)	213	41.3	140	45.6	34.9
	b(L/g)	4.28	2.57	13.8	1.67	1.91
	blank(mg/kg)*	15.5	28	17	<15.9	10
	BCF(max.)**	4.68E+03	2.34E+03	2.28E+04	1.61E+03	1.42E+03
ela.	Qmax(mg/g)	194	44.0	105	36.3	32.9
	b(L/g)	3.27	1.76	13.3	1.84	1.43
	blank(mg/kg)*	45	24	11	<18.3	<18.3
	BCF(max.)**	3.61E+03	2.18E+03	2.03E+04	1.87E+03	1.22E+03

*The metal concentration in biomass before contacting experiments.
**Bioconcntration Factor=(specific adsorption/exposure concentration)

Table 3. Summary results of the adsorption characteristics of five metals in *Najas graminea* Del. buffered at pH=5.

	Cu	Zn	Pb	Cd	Ni
Qmax(mg/g)	*	*	33.8	*	*
b(L/kg)	31.6	38.2	301	31.6	18.7
blank(mg/kg)	9.9	<15.6	8.8	<15.6	<15.6
BCF(max.)	5.25E+01	1.68E+02	4.10E+02	5.58E+01	1.15E+02

* Not available, linear adsorption isotherm was not available

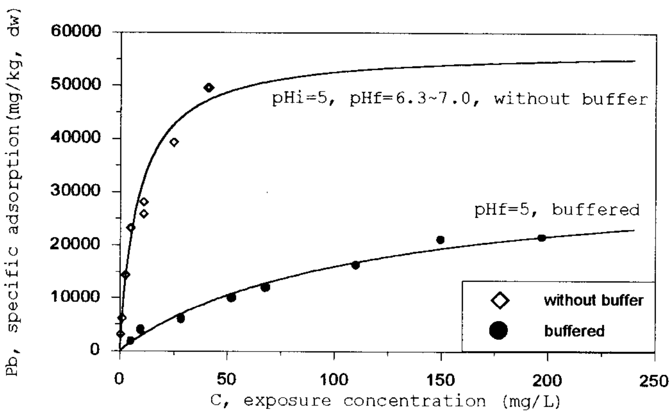

Figure 3. Comparison of plant adsorption isotherm for lead, *Najas gramina* Del. under different concentration of solution.

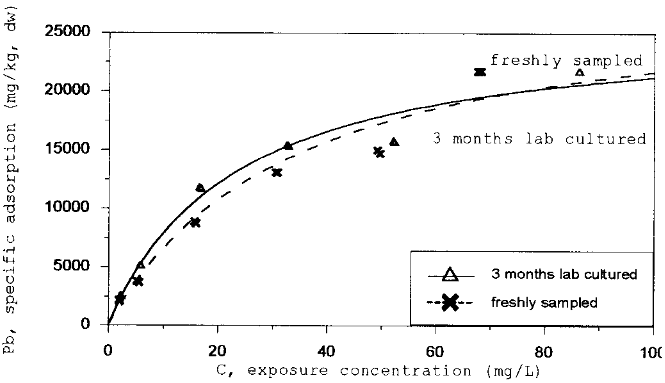

Figure 4. Effect of culture ofbiomass, *Najas graminea* Del. on the adsorption isotherm for lead and cadimum.

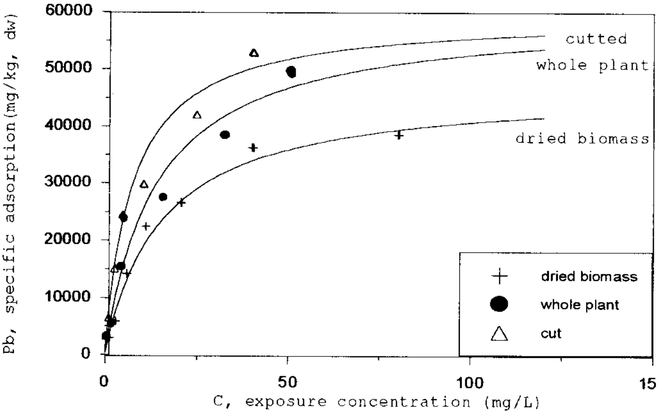

Figure 5. Comparison of plant adsorption isotherm for lead, *Najas graminea* Del. by different pretreatment of biomass.

Table 4. Summary results of the adsorption characteristics of lead and cadmiumonto Najas *graminea* Del. by different pretreatment of biomass.

		Pb	Cd
dried	Qmax(mg/g)	50.5	15.3
	b(L/g)	2.84	0.38
	blank(mg/kg)	19	<15.6
	BCF(max.)	2.73E+03	4.57E+02
uncut	Qmax(mg/g)	68.9	24.6
	b(L/g)	3.89	0.57
	blank(mg/kg)	19	<15.6
	BCF(max.)	1.05E+04	6.00E+02
cut	Qmax(mg/g)	69.0	26.1
	b(L/g)	6.96	1.13
	blank(mg/kg)	19	<15.6
	BCF(max.)	6.61E+03	1.18E+03

This study concluded that *Najas graminea* Del., *Myriophyllum scabratum* and *Myriophyllum elatinaides,* collected in Taiwan had promising potential for metal removal. In addition, aquatic chemistry (ionic strength) of contacting solution and preparation of biomass (inert or living) affected metal adsorption capability. However, uncutted whole plant and the cutted sample did not significantly differ with respect to metal adsorption characteristics. Also, biomass obtained from lab culture did not markedly differ from freshly sampled biomass (from field) in terms of metal adsorption capability. These three aquaticplants could be grown in river, stream, acid mining drainage or grown in clean water systems initially and then transfer to the polluted water to remove metals from aquatic systems.

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REFERENCES

Bervoets L, Panis L I, Verheyen R (1994) Trace metal levels in water, sediments and Chironomus GR. Thumni from different water courses on flanders (Belgium). Chemosphere 29:1591-1601.

Chawia G, Singh J, Viswanathan P N (1991) Effect of pH and temperature on the uptake of cadmium by *Lemna minor.* Bull Environ Toxicol 47:84-90.

Dani Vrhovsek, Vlasta Kukanja, Tjasa Bulc (1996) Constructed wetland (CW) for industrial waste water treatment. Wat Res 30:2287-2292.

Guilizzoni P (1991) The role of heavy metals and toxic material in the physiological ecology of submersed macrophytes. Aquatic Botany 41:87-109.

Gupta M, Chandra P (1994) Lead Accumulation and Toxicity in *Vallisneria Spirallis* (L.) and *Hydrilla verticillata* (F.). J Environ Sci Health A 29:503-516.

Jackson L, Kalkff J, Rsmussen JB (1994) Sediment pH and redox potential affect the bioavailability of Al, Cu, Fe, Mn, and Zn, to rooted aquatic macrophytes. Can J Fish Aquat Sci 50:143-148.

Lee C L, Chen H Y, Chuang M *Y* (1996) Use of oyster, *Crassostrea Gigas,* and ambient water to assess metal pollution status of the charting coastal area, Taiwan, after the 1986 green oyster incident. Chemosphere 33:2505-2532.

Mouvet C, Morhain E, Sutter C, Counturieux N (1993) Aquaticmosses for the detection and follow up of accidental discharges in surface water. Wat Air Soil Pollut 67:333-347.

Muramoto S, Oki Y (1983) Removal of some heavy metals from polluted water by water hyacinth (*Eichhornia crassipes*) . Bull Environm Contam Toxicol 30:170-177.

Ramelow G J, Fralic D, Zhao Y (1992) Factors affecting the uptake of aqueous metal ions by dried seaweed biomass. Microbios 72:81-93.

Raphael Nir, Avital Gasith , Albert S Perry (1990) Cadmiumuptake and toxicity to water hyacinth : effect of repeated exposures under controlled conditions. Bull Environ Toxicol 44:149-157.

Sajwan K S, Ornes W H (1994) Phytoavailability and bioaccumulation of cadmium in duckweed plants. J Environ Sci Health A 29:1035-1044.

Satake K, Shibata K , Bando Y (1990) Mercury sulphide (HgS) crystals in the cell walls of the aquatic bryophyte *Junerrgmannia vulcanicola Steph.* and *Sapania undulata* (L.) Dum. Aquat Bot 36:325-341.

Sela M, E Tel-Or, Fritz E, Huttermann A (1988) Localization and toxic effects of cadmium, copper and uranium in *Azolla.* Plant Physiol 88:30-36.

Sela M, J Garty, E Tel-Or (1989) The accumulation and the effect of heavy metals on the water fern *Azolla filiculoides.* New Phytol 112:7-12.

Wang T C, Weissman J C, Ramesh G, Varadarajan R, Bennemann J R (1998) Heavy metal binding and removal by Phormidium. Bull Environ Contam Toxicol 60:739-744.