

Removal of Lead Using Some Aquatic Macrophytes

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Lead is one of the most toxic heavy metals that accumulates in the earth's crust since its release from the industrial revolution era. The environmental problems about Pb accumulated in the natural resource have occurred around the world including Thailand. Aquatic plants are used widely to remove metals in the contaminated water resource. Based on earlier reports on Pb removal and bioaccumulation, aquatic plants such as *Ceratophyllum demersum*, *Hydrorhiza aristata*, *Eichhornia crassipes*, *Spirodela polyrhiza*, *Myriophyllum spicatum*, *Azolla pinnata*, showed the ability to accumulate Pb at high concentrations (10,000 – 25,000 mg/kg) and remove this metal at high percentage (Muramoto and Oki, 1983; Jain *et al.*, 1990; Rai *et al.*, 1995; Wang *et al.*, 1996; Axtell *et al.*, 2003). Heavy metal removal by aquatic plants can be greatly enhanced by the judicious selection of appropriate aquatic plant species. Selection is based on the type of metal to be remediated, the geographic location, environmental conditions, and the known accumulation capacities of the species (Zayed *et al.*, 1998). For this reason, it is important to develop knowledge about the abilities of aquatic plants to absorb and accumulate heavy metals. Since there was little information on removal and bioaccumulation Pb in aquatic plants, this study was conducted to compare the bioaccumulation and removal efficiency in four aquatic macrophytes, *C. demersum*, *Hygrophila difformis*, *Cabomba caroliniana*, and *Ludwigia hyssopifolia*. These aquatic plants can grow and propagate easily in contaminated water resources in Thailand.

MATERIALS AND METHODS

Aquatic macrophytes (*C. demersum*, *H. difformis*, *C. caroliniana* and *L. hyssopifolia*) were collected from natural ponds in Suphanburi province. They were acclimatized in 10% Hoagland's solution with aeration under laboratory conditions (temperature 27°C, light intensity 20,000 LUX, photoperiod 12 hours/day) at least for 7 days. Plants were weighed initially and then treated with various Pb(NO₃)₂ concentrations: 0 (as control group), 0.1, 1, and 10 mg/L in plastic bowls filled with 800 mL of 10% Hoagland's solution. Each experiment was done in triplicate. Plants were kept under

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similar laboratory conditions as described in acclimatization. The pH of all solutions was adjusted to 5.7 everyday. Plants were harvested after 0, 3, 6, 9, 12, and 15 days. The procedures for the analysis of Pb in the samples were performed according to the Standard Methods (APHA, 1998). Plants were rinsed twice with distilled water, blotted dry and whole plant fresh weight determined. They were oven dried at 100°C overnight, plant dry weight determined, then acid digested (APHA, 1998). Total Pb content in the entire plant was determined using a flame atomic absorption spectrophotometer (FAAS). In addition, the analysis of the remaining residual solution was performed. 100 mL each of solution after removal of plants were allotted to a glass tube, acid digested, and Pb content determined by FAAS. The relative growth of plants was calculated by dividing the final biomass (g) with the initial biomass (g) of treated plants and multiplied by one hundred. The bioconcentration factor (BCF) of each plant species was determined by dividing the metal concentration in plant tissue at harvest (mg/kg) by initial concentration of metal in the external solution (mg/L). The mean values of relative growth and metal content were calculated and subjected to the analysis of variance (ANOVA) using a randomized block design and Least Significance Difference Method (LSD) on the SPSS for Windows program.

RESULTS AND DISCUSSION

All tests were significant at $P=0.05$. The relative growth of plants after 15 days of contact in Pb solutions at various concentrations is shown in Figure 1. The relative growth of plants generally decreased with the increase in Pb concentration. *H. difformis* showed the highest relative growth at each Pb concentration, followed by *C. demersum* and *C. caroliniana*. The lowest relative growth was found is *L. hyssopifolia* exposed to 10 mg/L of Pb.

Pb accumulation by aquatic plants is shown in Figure 2. There was a significant increase of Pb in plant tissues when initial concentration was increased. Pb accumulation varied depending on plant species. At very low Pb concentration (0.1 mg/L), *H. difformis* showed the highest accumulation (348 mg/kg) after 9 days of exposure. At higher Pb concentration, *C. demersum* showed substantial accumulation, i.e., 1621 mg/kg at 1 mg/L Pb after 6 days, and 6982 mg/kg at 10 mg/L Pb after 9 days.

The percentage removal of Pb is shown in Figure 3. In general, the maximum removal (80-100%) was in the first 3-6 days for all plants at 0.1 mg/L. The percentage removal of Pb decreased with higher Pb concentration. *C. caroliniana* showed the best removal efficiency (80-90%) at higher Pb concentrations (1, 10 mg/L) after 12-15 days of exposure.

The BCFs for Pb in four aquatic plants at different concentrations and exposure times are shown in Figure 4. There was a gradual decrease in BCF with increasing Pb concentration. At 0.1 mg/L Pb, *H. difformis* showed the highest BCF value (3480) after 9 days of exposure. However, at higher Pb concentrations, *C. demersum* showed the highest BCF values after 6-9 days of exposure (1621 at 1 mg/L; 698 at 10 mg/L).

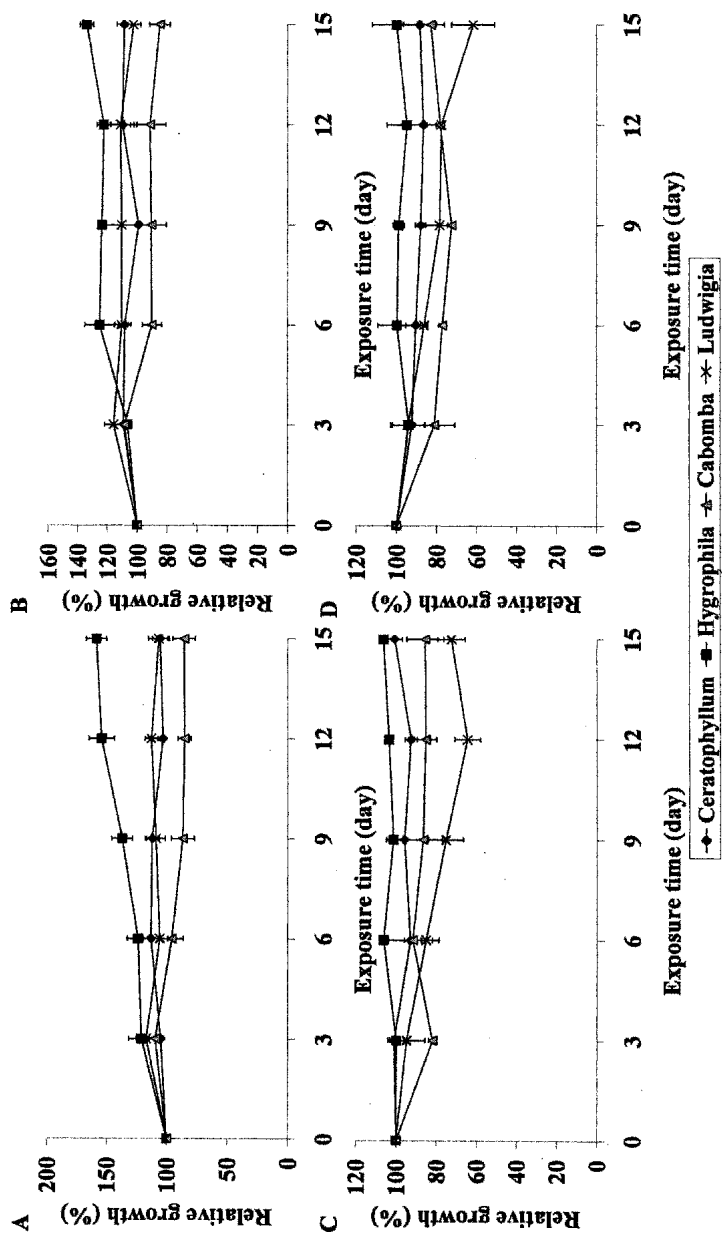


Figure 1. The effects of Pb on plant relative growth at various concentrations and exposure times. (A) control (B) 0.1mg/L (C) 1mg/L (D) 10 mg/L.

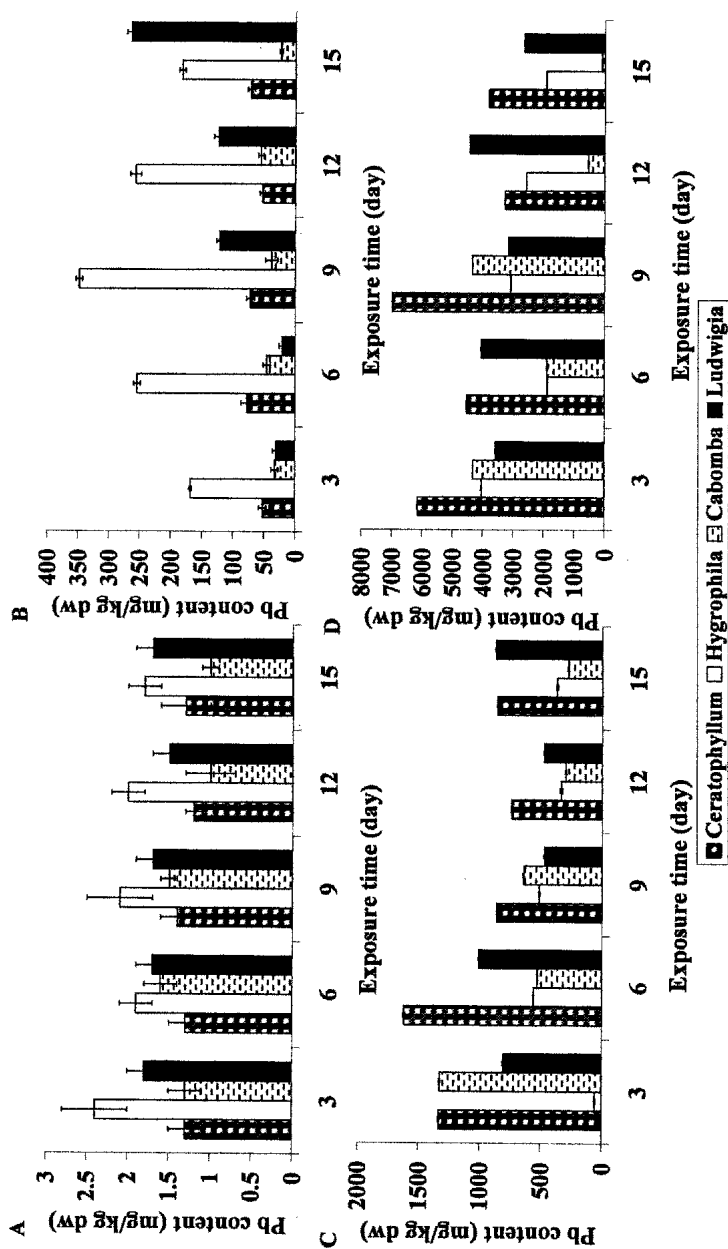


Figure 2. Pb accumulation by plants at various concentrations and exposure times. (A) Control (B) 0.1mg/L (C) 1mg/L (D) 10 mg/L.

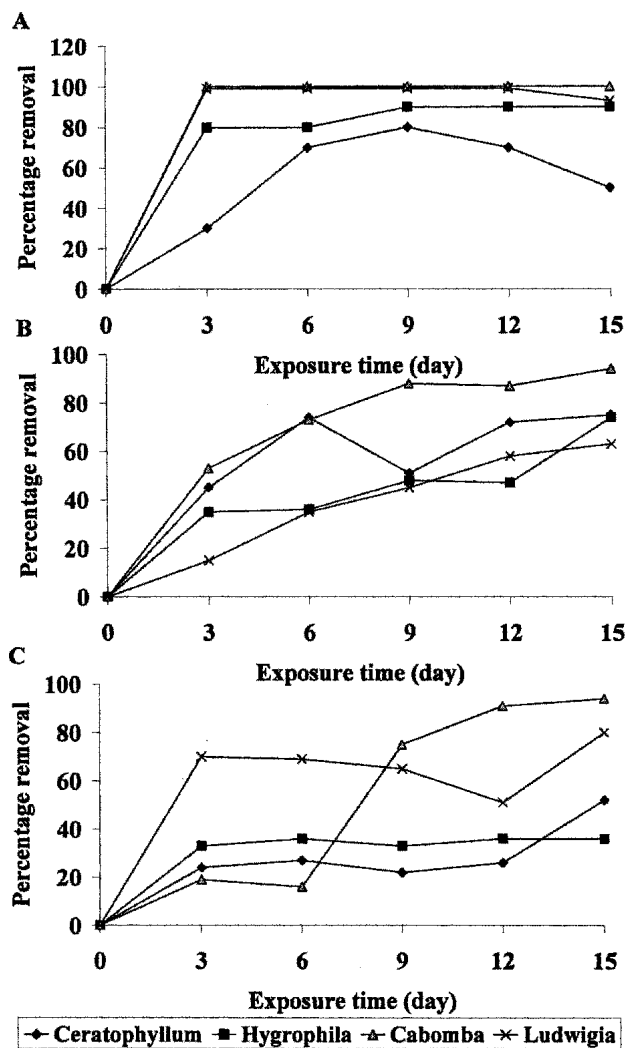


Figure 3. Percentage removal of Pb by plants at various concentrations and exposure times. (A) 0.1 mg/L (B) 1 mg/L (C) 10 mg/L.

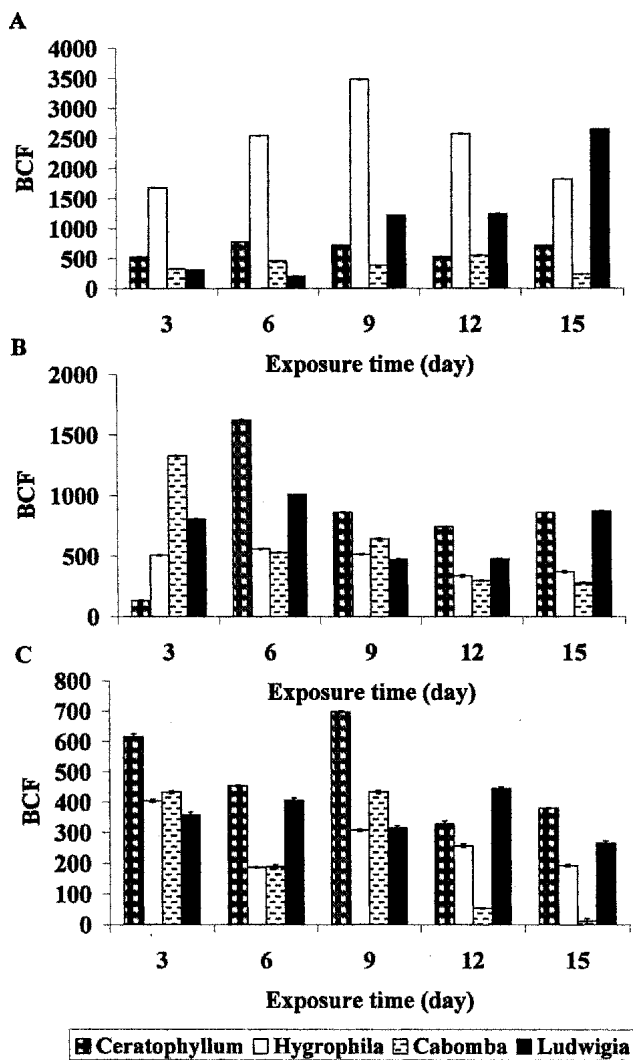


Figure 4. Bioconcentration factors of plants at various Pb concentrations and exposure times. (A) 0.1 mg/L (B) 1 mg/L (C) 10 mg/L.

The results show that, among the four species of aquatic macrophytes tested (*C. demersum*, *H. difformis*, *C. caroliniana* and *L. hyssopifolia*), *C. demersum* and *H. difformis* exhibited higher relative growth and were able to remove Pb at all experimental concentrations up to 15 days of treatment. *C. demersum* could remove and accumulate Pb at higher concentration than *H. difformis*, i.e. 6982 mg/kg Pb when supplied with 10 mg/l Pb after 9 days of exposure. Our result conforms with the earlier report that *C. demersum* could accumulate high concentration of Pb (Rai *et al.*, 1995). Several studies showed that *C. demersum* could accumulate other metals as well such as Cd, Cu, Fe, Cr and Mn (Garg and Chandra, 1991; Ornes and Sajwan, 1993; Gupta and Chandra, 1996; Bunluesin *et al.*, 2004), indicating that *C. demersum* are unspecific collectors of metals. However, an earlier study demonstrated that aquatic macrophytes showed both contracting characteristics of being either unspecific accumulators of metals or metal specific species (Rai *et al.*, 1995).

The maximum removal of Pb occurred after 3-6 days for all plants tested. The removal rates ranged from 80-100%. The percentage removal of Pb continuously decreased with time. This is indicative of a fast attainment of saturation state in the plants (Srivastava *et al.*, 1994). As soon as the saturation state was reached, it seemed a little difficult for plants to further absorb Pb, so the concentration was relatively constant with the passage of time. Among the four tested plant species, *C. caroliniana* showed the highest removal percentages (100 % at 0.1 mg/L after 15 days). The percentage decreased with the increase in Pb concentration (94% at 1 and 10 mg/L). Similar results have been reported for the removal of other metals by water hyacinth (Chigbo *et al.*, 1982; Muramoto and Oki, 1983), Cr and Ni by *Salvinia molesta* and *polyrhiza*, (Srivastav *et al.*, 1994), and Pb and Zn by *A. and Lemna minor* (Jain *et al.*, 1990).

Bioconcentration factor is a useful parameter to evaluate the efficiency of the plants accumulating metals and this value is calculated on a dry weight basis. Metal accumulations by macrophytes can be affected by metal concentrations in water and sediments (Lin and Zhang, 1990). In general when the metal concentration in water increases, the amount of metal accumulation in plants increases, whereas the BCF value decreases. The highest BCF value (3480) was found in *H. difformis* followed by *L. hyssopifolia* (2644) at 0.1 mg/L Pb. At higher Pb concentration (1 mg/L), *C. demersum* and *C. caroliniana* showed the BCF values of 1621 and 1327, respectively. Zayed *et al.* (1998) and Zhu *et al.* (1999) considered a plant with a BCF of over 1000 (a 100-fold compared on a fresh weight) as a hyperaccumulator. Based on this criterion, *C. demersum* and *C. caroliniana* were good hyperaccumulators at low Pb concentration (0.1 mg/L), while *H. difformis* and *L. hyssopifolia* were good hyperaccumulators at higher Pb concentration (1 mg/L). These four aquatic macrophytes could be useful in the removal of Pb from contaminated water.

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