

Efficiency of *Spirodela polyrhiza* for Removal and Uptake of Pb and Ni

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Abstract: *Chronic heavy metal toxicity is often caused to humans due to widespread industrial use of heavy metals and their discharge along with the wastewater into the local water bodies. To develop a cost effective and eco-friendly treatment technology, the potential of duckweed Spirodela polyrhiza for phytoremediation of Pb and Ni individually have been investigated in the present study. Phytoremediation is a plant based technology that is a viable alternative for the remediation of contaminated water.*

The experiment was carried out at different initial concentrations of 0.91, 1.97 and 2.88 mg/L for Pb and 2.92, 3.9 and 4.9 mg/L for Ni respectively along with a control set up for 22 days. The percentage of Pb and Ni removed from the solution was decreased when initial Pb and Ni concentrations were increased. The fresh biomass production of Spirodela polyrhiza was found to be affected and decrease more in case of Pb than Ni over the experimental period with increase in initial Pb and Ni concentrations individually and chlorosis of Spirodela polyrhiza was a toxicity symptom noted when concentration of Pb and Ni exposure increased. Spirodela polyrhiza was found to accumulate more than 1000 mg/kg of Pb and Ni in their dry biomass at all concentrations. With increase in Pb concentration in external solution, the accumulation of Pb in the Spirodela polyrhiza was found to increase with a maximum accumulation of 7051 mg/kg obtained at 2.88 mg/L Pb. Ni accumulation decreased with increase in initial concentration. Correlation between initial concentration of Pb in external solution and accumulation of Pb in Spirodela polyrhiza was high and positive. Results indicate that Spirodela polyrhiza is tolerant of Pb and Ni and is hence suitable for the removal of both at lower concentrations.

Keywords: *Lead, Nickel, Spirodela polyrhiza, phytoremediation, removal, accumulation*

1. INTRODUCTION

Rapid urbanization and industrialization has led to heavy metal pollution of both land and water resources. Aquatic ecosystems get polluted due to discharge of effluents from industries and

therefore their remediation is important. This paper presents the study of removal and uptake of toxic heavy metals Pb and Ni from aqueous solution by phytoremediation in batch study. Lead is one of the most common heavy metal [1] and its major sources include paints, pesticides (lead arsenate) [1], electroplating industry wastewater etc. Ni is another commonly available heavy metal used in industries and has been included as a probable human carcinogen [2].

Phytoremediation, a green technology has been found to be a cost effective and environment friendly process for wastewater treatment. This technology utilizes the ability of some plants to remove and uptake toxic metals in their biomass. In the present study, plants used are duckweeds *Spirodela polyrhiza* that are commonly available floating weeds that can remove as well as accumulate different toxic heavy metals in their biomass.

2. MATERIALS AND METHODS

Aquatic weeds *Spirodela polyrhiza* were collected from a local natural pond near Jadavpur University, Salt Lake Campus, Salt Lake. The collected plants were then washed with distilled water to remove any debris if present. Further they were acclimatized for a period of seven days in the pond water of the respective pond under laboratory conditions. In plastic water tubs of 25L capacity, an amount of 20L pond water was poured for each experimental set-up. The depth of the solution was found to be 17.5cm. *Spirodela polyrhiza* were incubated with different concentrations of PbCl₂ (0.91, 1.97 and 2.88) mg/L and NiCl₂ (2.92, 3.9 and 4.9) mg/L respectively. 40 gm of *Spirodela polyrhiza* was used per experimental tub. Each experiment was done in triplicate. All chemicals used were of analytical grade.

After completion of experimental period of 22 days, the plants were harvested, washed with distilled water, were blotted dry and their fresh weight were determined. Further to obtain the plant dry weight, the plants were oven-dried for 48 hours at 60°C [3], dry weight recorded and were ground to powder and further digested and metal was quantified in plant biomass as well as in treated water samples collected using Atomic Absorption Spectrophotometer (Perkin Elmer). The procedure followed for Pb analysis in samples was performed according to Standard Methods [4]. The metal concentrations in plants were expressed in mg per kg dried biomass. Metal concentrations in aqueous solutions were expressed in mg/L.

3. RESULTS AND DISCUSSION

Morphological Symptoms

The visual changes observed in case of duckweed *Spirodela polyrhiza* after being treated with different initial concentrations of Pb for 22 days were minor, such that plants were found to be healthy and green. Little effects were noted in case of higher initial Pb concentration, i.e 2.88

mg/L. There was partial wilting and gradual yellowing of leaves with increase in number of days of treatment at initial concentration of 2.88 mg/L Pb. Similar observations of decline of photosynthetic pigments at different concentrations of Pb was obtained in *Ceratophyllum demersum* [5]. Pb inhibits ALAD activity and overall chlorophyll biosynthesis by binding with SH group of enzymes [6]. At different initial Ni concentrations, biomass production of *Spirodela polyrhiza* was decreased as compared to that of control. Frond chlorosis appeared at 3.9 mg/L Ni exposure with more prominence at 4.9 mg/L Ni concentration obtained earlier (after 10th day). At concentrations of 2.92 mg/L Ni concentrations, no major visible toxicity symptoms were found.

Percentage Removal of metals

There was a decreasing trend of removal percentage with the increase in initial Pb concentration. At concentration of 0.91 mg/l Pb, a maximum of 93.19 % removal was achieved (Fig.1). Removal percentage was reduced to 82.23 % when initial Pb concentration was increased to 1.97 mg/L. Results indicate that with increase in initial Pb concentration, the removal rate decreased. This suggests saturation of finite number of binding sites present on the plant biomass. Similar results were obtained when removal of Pb by different aquatic macrophytes was studied [11]. Correlation between the initial concentration of Pb in the ambient solution and the concentration of Pb removed after 22 days was found to be positive ($r = 0.951$, $p < 0.001$). Pb removal of 50 % in aqueous phase was obtained after first 24 hours [7]. Researchers found similar observations of decrease in Pb concentration in water with time [8]. Research was conducted to study removal of Pb and Cd and it was reported that surface adsorption was the main mechanism of Cd removal [9] by *Spirodela polyrhiza*. They also reported that the predominant mechanism of Pb removal by *Salvinia minima* was surface adsorption.

Results presented in Fig.2 showed significant critical differences ($P=0.05$) in case of removal of Ni by *Spirodela polyrhiza* at different initial concentrations. Maximum Ni removal was obtained at initial Ni concentration of 2.92 mg/L at 22nd day of exposure and minimum removal obtained at initial concentration of 4.9 mg/L Ni [10]. Removal percentage of Ni decreased with increase in initial Ni concentration. The drop in % removal at higher Ni concentrations was due to increase in metal ions such that adsorption sites for Ni ions on the plant biomass remained the same. At lower initial metal concentrations, adsorption sites on plants were sufficient enough for the sorption of Ni. The rate of metal removal decreased eventually with increase in number of days of exposure. Positive correlation was obtained ($R^2 = 0.84$, $p < 0.005$) between the initial Ni concentration in the ambient solution and the concentration of Ni removed from the solution after 22 days.

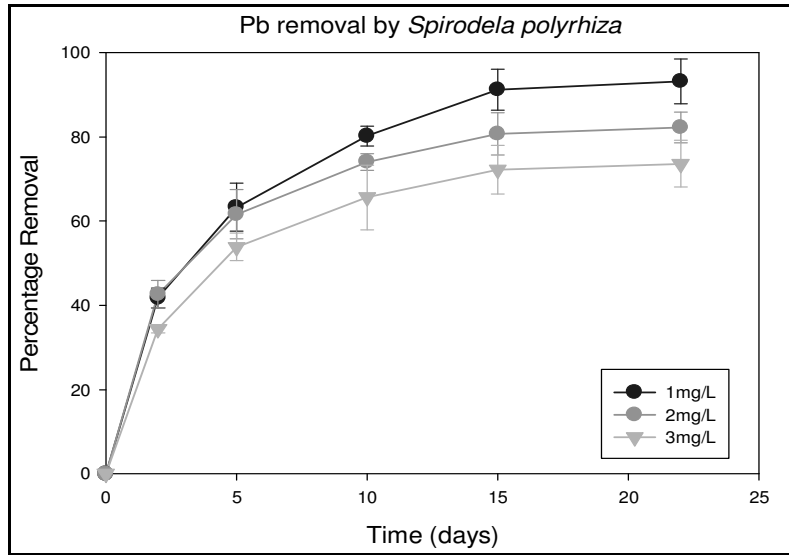


Fig.1 Percentage removal of lead by *Spirodela polyrhiza*. Bars denote \pm standard deviation from the mean of three replicates

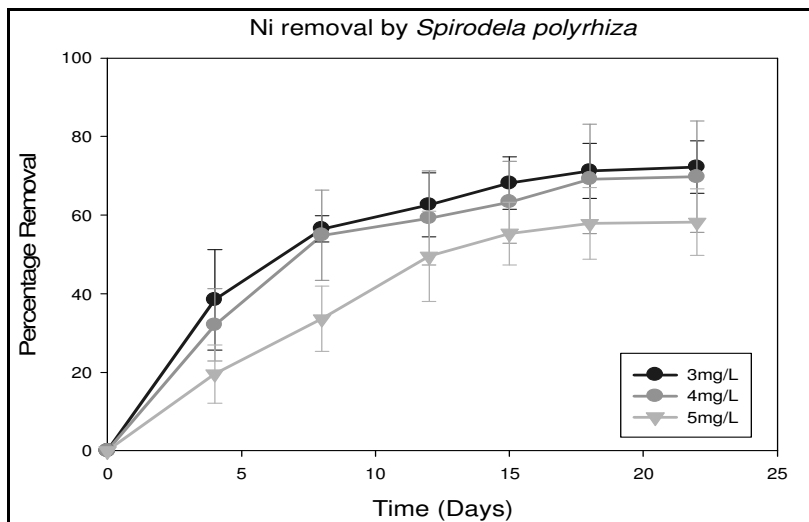


Fig.2 Percentage removal of Ni by *Spirodela polyrhiza* at different initial concentrations. Bars denote \pm standard deviation from the mean of three replicates. Data for initial concentrations of Ni (3.9 and 4.9) mg/L [10]

Accumulation of Pb and Ni in Spirodela polyrhiza

The metal accumulation in plant increased with the increase in initial lead concentrations (Table.1). Anova suggested that statistical significant differences existed in Pb accumulation by *Spirodela*

polyrhiza at different Pb initial concentrations ($p = 0.004$). Correlation ($r = 0.97$, $p < 0.005$) between initial concentration of Pb in external solution and accumulation of Pb in *Spirodela polyrhiza* was high and positive. Similar results were also obtained when aquatic plant *Phylidrum lanuginosum* was exposed to Pb [11]. It was found to accumulate more Pb with increase in initial concentration of Pb in solution. In another experiment, authors reported [12] that duckweed *Spirodela polyrhiza* has the ability to accumulate good amount of Pb from water. It has been reported by researchers [5] that aquatic macrophyte *Ceratophyllum demersum* accumulated high amount of Pb in their biomass and the accumulation increased with increase in concentration of Pb in aqueous solution. High accumulation of Pb by plants has been attributed to the binding capacity of Pb to the cell wall of plants and also to other ligands such as GSH, OCs etc. They also reported that Pb remains bound sometimes even to vacuoles. It was found that with increase in duration of treatment, Pb accumulation rate of *Ceratophyllum demersum* decreased due to saturation of Pb binding sites [5].

Maximum accumulation of Ni was obtained by *Spirodela polyrhiza* at 2.92 mg/L Ni. Over an exposure period of 22 days, 6960.98 ± 636.23 mg/kg Ni accumulated at 2.92 mg/L concentration. The duckweed accumulated more Ni at low concentration. With increase in concentration to 3.9 and 4.9 mg/L Ni, accumulation by *Spirodela polyrhiza* decreased to 4498.57 ± 67.41 and 3679.31 ± 350.72 mg/kg.

Table.1. Accumulation of Pb by *Spirodela polyrhiza* at different initial Pb concentrations

Desired Initial Pb concentration (mg/L)	Actual Initial Pb concentration (mg/L)	Pb in dry biomass (mg/kg)	Desired Initial Ni concentration (mg/L)	Actual Initial Ni concentration (mg/L)	Ni in dry biomass (mg/kg)
1	0.91 ± 0.03	1806 ± 24.68	3	2.92 ± 0.17	6960.98 ± 636.23
2	1.97 ± 0.09	4436 ± 283.58	4	3.9 ± 0.22	4498.57 ± 67.41
3	2.88 ± 0.03	7051 ± 631.54	5	4.9 ± 0.17	3679.31 ± 350.72

Statistical differences between accumulation of Ni at different concentrations was significant ($p < 0.001$). Correlation between initial Ni concentration in external solution and Ni accumulated in *Spirodela polyrhiza* was high and negative ($r = -0.89$, $p < 0.005$). Table.1 includes values of accumulated Ni in *Spirodela polyrhiza*. The residual Ni concentration was negatively correlated ($r = -0.73$, $p > 0.01$) with the amount of Ni accumulated in *Spirodela polyrhiza*.

4. CONCLUSION

It can be concluded that *Spirodela polyrhiza* has good capacity to remove heavy metals Pb and Ni from aqueous solutions. It has ability to accumulate Pb and Ni to a large extent. The toxic effect of Ni on *Spirodela polyrhiza* was less than that of Pb as obtained from their morphological changes and growth. Percentage removal of Pb by *Spirodela polyrhiza* was more than that obtained for Ni. From accumulation values, it could be inferred that *Spirodela polyrhiza* accumulates both Pb and Ni to a good extent. It hence acts as a promising macrophyte for remediation of Pb and Ni and could thus be used for removal of both the metals from aqueous solutions.

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