

# Assessment on Cr, Cd, As, Ni and Pb uptake and phyremediation potential of *Scirpus mucronatus*

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**Abstract:** A laboratory experiment was conducted to examine the heavy metals (Cr, Cd, As, Ni and Pb) phytoaccumulation potential of an aquatic macrophytes *Scirpus mucronatus*. The macrophyte were transferred to the laboratory containing nutrient solution enriched with 1.0, 2.0, 4.0, 8.0 and 16 mg L<sup>-1</sup> of Cr, Cd, As, Ni and Pb and were separately harvested after 2, 4, 6, 8 and 10 days. The bioaccumulation study showed a linear relationship for Cr accumulation but curvilinear in the case of the other metals with the exposure time (2–10 d). The accumulation of all the metals was found to be higher in the root than the shoot. The maximum bioconcentration factor (BCF) and translocation factor (TF) value were found in Pb (1387 and 0.95) and minimum BCF and TF values were found in Ni (386) and Cd (0.51) treated plants respectively. The experimental results demonstrated that the *S. mucronatus* can be used for removal of Cr, Cd and Pb from contaminated water.

**Key words:** *Scirpus mucronatus*, heavy metals, Bioconcentration factor (BCF), Translocation factor (TF)

## Introduction

Water though an indispensable resource for human life is yet one of the most badly abused resources. For centuries, especially in urban areas, water has been polluted and used as dumping places for all sorts of domestic and industrial waste as well as sewage. Over 75 to 90 percent of people in developing countries are exposed to unsafe drinking water [1], hence proper water treatment is inevitable in order to ensure healthy life. In recent times, the occurrence of excess metal contaminants especially the heavy metals has become a problem of increasing concern in natural loads [2]. This situation has arisen to global deterioration of environmental quality and especially due to rapid growth of population, increased urbanization and large-scale industrialization and production of variety of chemical compounds [3], lack of environmental regulations in exploration and exploitation of natural resources, extension of irrigation and modern agricultural practices [4]. Heavy metal pollution is a global problem and the various sources of heavy metals are from the industrial waste and fertilizers which causes serious concern in nature as they are non-biodegradable and accumulate at high levels when they are release in excess levels into the environment although [5].

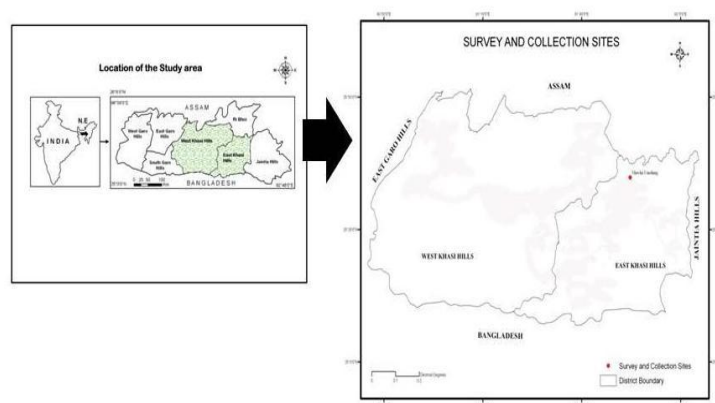
A variety of techniques which includes chemical, physical and biological technology have been used to remediate heavy metal contamination from soil or water. Toxic metals from industrial effluents have been remove by various other techniques such as precipitation, reduction, artificial membranes, and ion exchange, but however these techniques generate a huge amount of waste e.g., sludge, metal rich waste, etc which is difficult to dispose of and therefore, dangerous to the environment and they are also generally expensive, relatively inefficient [6]. Phytoaccumulation, one of the biological indicators which indicate the degree of absorption of heavy metals in plants has lately gained its applicability

because its cost-effectiveness, long-term and ecological aspect [7]. Aquatic macrophytes have received great attention and have shown to be one of the candidates in the aquatic system for pollutant uptake and biological indicators of heavy metal [8].

The objective of the present study was to assess the phytoaccumulation potential of *S. mucronatus* when exposed to different types of heavy metals and concentrations under laboratory conditions. The experiments were performed in a contained environmental set up in order to eliminate all external environmental factors.

## 2. Materials and Methods

*S. mucronatus* an emergent are one of the major natural constituent of wetland and riverside vegetation. They are sampled as shown in Fig 1 from water body of Mawlai Umshing, (Lat 25°36'36.76N Long 91°54'05.11E), Meghalaya, India in the month of April- September 2011 and collected in polyethylene bags and transferred to the laboratory.



**Figure 1:** Map showing location and collection sites of aquatic macrophytes

Plants were washed several times with tap and distilled water in order to remove any adhering soils and plants of similar size, shape and height were selected and kept separately in a 40L capacity tank which contained half strength Hoagland's solution of pH = 7 [9], and kept for 15 days prior to experimentation. The Hoagland solution was modified by omitting ferrous sulfate in order to prevent the As precipitation by iron. After 15 days the acclimatized plants were transferred and maintained in 5% Hoagland's solution containing working Cr, Cd, As, Ni and Pb standard solutions of different concentrations 1.0, 2.0, 4.0, 8.0 and 16.0 mg L<sup>-1</sup> and then they were exposed to Cr, Cd, As, Ni and Pb concentrations at a time interval of 2, 4, 6, 8 and 10 days. Heavy metals of analytical grade, were supplied as K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>, CdCl<sub>2</sub>·2H<sub>2</sub>O, As<sub>2</sub>O<sub>3</sub>, NiCl<sub>2</sub>·6H<sub>2</sub>O and Pb(NO<sub>3</sub>)<sub>2</sub> (Himedia) were used as the source of Cr, Cd, As, Ni and Pb. Experiments were carried out separately for Cr, Cd, As, Ni and Pb concentrations and under controlled temperature (24±1<sup>o</sup>C) and light (3500 Lux) conditions. After each time interval the plants were collected and washed with deionised water to remove any metal adhering to its surface. The washed plant samples were carefully dried the adherent water using absorbent paper and then they are separated to roots and shoots. Samples were dried for 48h in an oven at 70±5<sup>o</sup>C. The dried oven plant root and shoot was then chopped and finally powdered using a mortar and pestle to ensure homogeneity for facilitating organic matter digestion. One control plant groups were also set up where no Cr, Cd, As, Ni and Pb were added into the medium was not added.

For digestion, the plant samples were carried out according to [10]. Atomic Absorption Spectrophotometer (AAS 3110, Perkin-Elmer) and was used to determine the Cr, Cd, As, Ni and Pb contents in plant root and shoot parts. The bioconcentration factor (BCF) is a useful parameter and it provides the ability index of a plant to accumulate metals with respect to metal concentration in the medium and it was calculated on a dry weight basis [11].

$$BCF = \frac{\text{Trace elements concentration in plant tissue } (\mu\text{g g}^{-1})}{\text{Initial concentration of the element in the external nutrient solution } (\text{mg L}^{-1})}$$

Translocation Factor (TF) is generally the translocation of heavy metal from roots to aerial part and indicates the internal metal transportation of the plant. The translocation factor is determined as a ratio of metal accumulated in the shoot to metal accumulated in the root [12].

$$TF = \frac{[\text{Metal}]_{\text{Shoot}}}{[\text{Metal}]_{\text{root}}}$$

Wherein, TF>1 indicates that the plant translocate metals effectively from the root to the shoot.

### 3. Statistics analyses

ANOVA and multiple linear regressions were performed for all the data to confirm their validity using SPSS 17. The data were all presented as mean ± standard error of three replicates. Fisher least significant difference (LSD) test was performed at p < 0.05 to check the significant difference between the means for different uptake at different Cr, Cd, As, Ni and Pb concentrations.

## 4. Results and Discussion

### 4.1 Accumulation of Cr, Cd, As, Ni and Pb

Cr, Cd, As, Ni and Pb content in the roots and shoots of *S. mucronatus* showed increases in metal accumulation in the roots and shoots if metal concentrations and time period are enhanced. At Cr, Cd, As, Ni and Pb concentration of 1, 2, 4, 8 and 16mg/L, the metals content (Fig-2) in *S. mucronatus* increased to the maximum 3046, 3765, 4663, 4716 and 4384 µg/g dry weight for Cr, 2144, 3531, 5917, 7048 and 3513 µg/g

dry weight for Cd, 2152, 3539, 5922, 7056 and 3517 µg/g dry weight for As, 4297, 7071, 6856, 14105 and 7031 µg/g dry weight for Ni and 6449, 8726, 17759, 21162 and 19549 µg/g dry weight for Pb at 2<sup>th</sup>, 4<sup>th</sup>, 6<sup>th</sup>, 8<sup>th</sup> and 10<sup>th</sup> day of harvesting and accumulation ranges from 320-4716 µg/g dry weight for Cr, 225-7049 µg/g dry weight for Cd, 226-7057 µg/g dry weight for As, 451-14105 µg/g dry weight for Ni and 677-21162 µg/g dry weight for Pb. The maximum accumulation was found on the 8<sup>th</sup> day (16mg/L) for Cr and on the 8<sup>th</sup> day at 8mg/L in the case of Cd, As, Ni and Pb and minimum was on 2<sup>nd</sup> day (1mg/L) of exposure time for all the metals. Statistical analyses was conducted and it was found out that there was a significant increased (p<0.05) in metals (Cr, Cd, As, Ni and Pb) accumulation by *S. mucronatus* with the increased of metal concentration in the medium but however it was reverse in the case of the number of days where it shows no significant increased (p<0.05) in metal accumulation with the increased in the number of days.

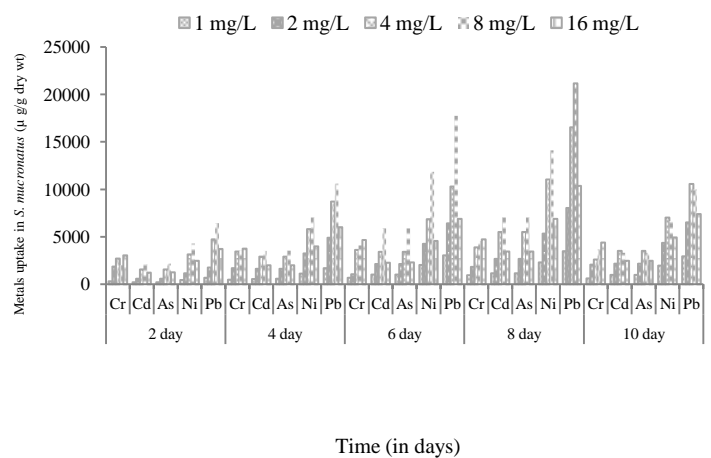


Fig 2. Metals accumulation in *S. mucronatus*

Results indicated that *S. mucronatus* have the ability to uptake Cr, Cd, As, Ni and Pb from the surrounding solution as in agreement with the reports of earlier studies that aquatic plant tend to adapt themselves to cope-up with metals toxicity [13]. The major factor influencing in metal uptake efficiency of plants depends on the metal concentration in the medium [14]. Generally it is suggested that the important uptake route in plants are the roots, and it is expected that roots will have a higher uptake as compared to the shoot [15]. It was found out that the roots of *S. mucronatus* accumulate higher Cr, Cd, As, Ni and Pb concentrations as compared to the shoots part. The accumulation of Cr, Cd, As, Ni and Pb in the shoots of an emergent plant is generally dependent on the roots as its primary source [16]. Root morphology plays an important role in the ability of plants to accumulate heavy metals, generally plants with long, fine roots formed a larger root system which in turn helps in efficient acquisition of nutrients or metal than those plants which have a short and thick roots [17] which is observed also in *S. mucronatus* with a long fine roots system and have a higher Cr, Cd, As, Ni and Pb concentration in the roots by increasing root water contact. The Cr, Cd, As, Ni and Pb accumulation in the roots and shoots of *S. mucronatus* exposed to Cr, Cd, As, Ni and Pb at 2 days was significantly lower than metals accumulation at 8<sup>th</sup> day, this finding corresponds to that of [18].

The absorption pattern in the present study corroborated with the findings of [19] where emergent species have high accumulates in roots and lowest accumulations in

shoots. Additionally [12] mentioned that metals accumulated by wetland plants were mostly distributed in root tissues, suggesting that an exclusion strategy for metal tolerance widely exists in them. In the roots of *S. mucronatus* the accumulation of Cr, Cd, As, Ni and Pb is directly proportional with the increase of metals concentration in the medium which corroborates with the findings by [20]. Thus, higher concentrations of Cr, Cd, As, Ni and Pb in the roots of *S. mucronatus* corroborates with earlier studies of [21], [22], [23].

Correlation and multiple regression analyses were conducted to examine the relationship between Cr, Cd, As, Ni and Pb uptake by *S. mucronatus* and potential predictors (concentrations of Cr, Cd, As, Ni and Pb in the medium and time). Table 1, 2, 3, 4 and 5 summarizes the descriptive statistics and analysis results for Cr, Cd, As, Ni and Pb. As can be seen each of the uptake is positively and significantly correlated with the Cr, As and Pb concentration in the medium for *S. mucronatus*, indicating that with the increase in concentration in the medium they tend to have higher uptake of Cr, As and Pb into the plant tissues. However, in case of Pb uptake is significantly correlated with times i.e., the no of days have a significant outcome on the uptake of Pb, but time is not significantly correlated with Cr and As uptake. However, in the case of Cd and Ni their uptake to the plant tissues is not significantly correlated with concentration and time i.e., the Cd and Ni concentration in the medium and number of days does not have any significant effect on the uptake of Cd and Ni to the plant tissues.

The multiple regression model with all two predictors produced  $R^2 = .666$ ,  $F(2, 27) = 26.97$ ,  $p < .001$ ,  $R^2 = .202$ ,  $F(2, 27) = 4.660$ ,  $p < .001$ ,  $R^2 = .542$ ,  $F(2, 27) = 15.95$ ,  $p < .001$ ,  $R^2 = .208$ ,  $F(2, 27) = 2.885$ ,  $p < .001$ ,  $R^2 = .564$ ,  $F(2, 27) = 17.49$ ,  $p < .001$  for Cr, Cd, As, Ni and Pb respectively. As can be seen in Table 1, 2, 3, 4 and 5 the concentration of Cr, As and Pb in the medium had significant positive regression weights, indicating with higher Cr, As and Pb concentration in the medium were expected to have higher Cr, As and Pb uptake in *S. mucronatus*. However, in case of Pb had significant positive regression weights with time i.e., the no of days have a significant outcome on the uptake of lead, but time does not had significant positive regression weights with Cr and As uptake.. However, in the case for Cd and Ni, concentration and time i.e., number of days did not contribute to the multiple regression model and it is does not have a significant regression weights, indicating that uptake of Cd and Ni in *S. mucronatus* does not fully depend on Cd and Ni concentration and time period.

#### 4.2 Bioconcentration factor (BCF) of Cr<sup>+6</sup>

Bioconcentration factor (BCF) value indicates the ability of the plant to accumulate metal in their tissue parts. The BCF values (Fig-3) at different Cr, Cd, As, Ni and Pb concentrations (1, 2, 4, 8 and 16mg/L) were evaluated at 2, 4, 6 8 and 10 day. The maximum BCF for Cr was 1034 when treated with 2mg/L (10<sup>th</sup> day), 1377 at 4mg/L (8<sup>th</sup> day) for Cd, 937 at 1mg/L (8<sup>th</sup> day) for As, 386 at 2mg/L (6<sup>th</sup> day) for Ni and 1387 at 4mg/L (10<sup>th</sup> day) for Pb respectively.

**Table 1:** Summary statistics, correlations and results from the regression analysis for Cr

Variable	mean	std	correlati on with uptake	multiple regression weights	
				B	β
Uptake	2221.90 00	1640.37 349			
Time (in days)	6.0000	2.87678	.144	81.875	.14 4
Concentrati ons (mg/L)	5.1667	5.58374	.804***	236.099 ***	.80 4

\* p < .05 \*\* p < .01 \*\*\*p<.001

**Table 2:** Summary statistics, correlations and results from the regression analysis for Cd

Variable	mean	std error	correlatio n with uptake	multiple regression weights	
				B	β
Uptake (µg/g dry wt)	2114.5 3	748.90 3			
Time (in days)	6.00	104.77 9	.298	188.33 4	.29 8
Concentratio ns (mg/L)	5.17	53.983	.409	133.21 9	.40 9

\* p < .05 \*\* p < .01 \*\*\*p<.001

**Table 3:** Summary statistics, correlations and results from the regression analysis for As

Variable	mean	std error	correlatio n with uptake	multiple regression weights	
				B	β
Uptake	2237. 4	897. 4			
Time (in days)	6.0	125. 5	.196	188.6	.19 6
Concentration s (mg/L)	5.1	64.6	.709***	352.2** *	.70 9

\* p < .05 \*\* p < .01 \*\*\*p<.001

**Table 4:** Summary statistics, correlations and results from the regression analysis for Ni

Variable	mean	std	correlati on with uptake	multiple regression weights	
				B	β
Uptake	5081.08 00	1642.3 15			
Time (in days)	3.0000	445.60 8	.385	903.94 0	.38 5
Concentratio ns (mg/L)	6.2000	115.51 9	.244	148.59 2	.24 4

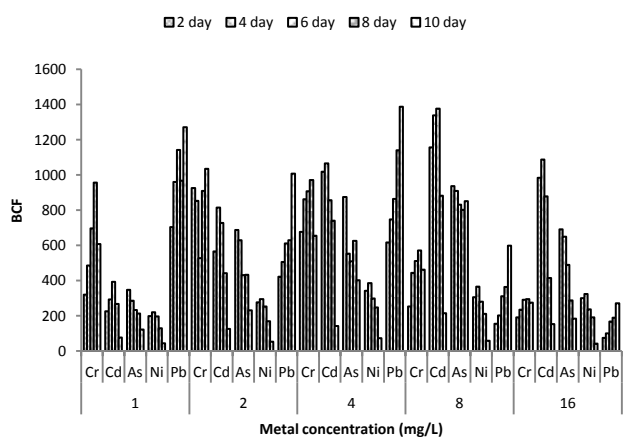
\* p < .05 \*\* p < .01 \*\*\*p<.001

**Table-5:** Summary statistics, correlations and results from the regression analysis in Pb

Variable	mean	std error	correlation with uptake	multiple regression weights	
				B	β
Uptake	1875.16	482.788			
Time (in days)	6.00	67.547	.348*	185.058*	.348
Concentrations (mg/L)	5.17	34.801	.666***	182.413**	.666

\* p < .05 \*\* p < .01 \*\*\* p < .001

Plants which have the ability to accumulate heavy metal in the tissues are generally classified as a good accumulator. Generally it is considered that a plant useful for phytoremediation should have a BCF value greater than 1000 [11]. In the present study, the BCF values for Cr (1034), Cd (1377), Pb (1387) was above 1000, which may be considered as a good accumulator of Cr, Cd and Pb as compared to As (937) and Ni (386).



**Fig 3.** Bioconcentration Factor for Cr, Cd, As, Ni and Pb in *S. mucronatus*

### 4.3 Translocation factor (TF) of Cr, Cd, As, Ni and Pb

Translocation Factor (TF) in plants is the ratio of heavy metal accumulation in the shoots parts to the roots. Translocation of heavy metal in plants are generally dependent on plant species, type of heavy metals and various environmental factors like pH, redox potential (Eh), temperature, salinity [24]. Yanqun *et al.* [25] reported that a TF value greater than 1, the plants are considered as an accumulator species, whereas TF lesser than 1 is an excluder species. The TF > 1 indicated that there is a transport of metal from root to leaf probably through an efficient metal transporter system [26], metals sequestration in the leaf vacuoles and apoplast [27]. According to Yoon *et al.* [28] TF value more than 1 of plant species indicates their hyperaccumulation potential and are known as hyperaccumulator plants.

The TF values for Cr, Cd, As Ni and Pb under different treatments are shown in Table-6, 7, 8, 9 and 10. From the tables it appeared that in most of the treatments the TF value (Table-6) were lesser than one indicating that Cr, Cd, As, Ni and Pb which indicates a lesser amount of Cr, Cd, As Ni and Pb is translocate from roots to shoots. The roots of the emergent aquatic plant (*S. mucronatus*) accumulate higher concentrations of Cr, Cd, As Ni and Pb than shoots, which

indicate that limited Cr, Cd, As Ni and Pb translocation occur from the roots to the shoots [12]. The decreased Cr, Cd, As Ni and Pb in the *S. mucronatus* leaves of the present study may be to casparian bands acting as effective barrier for the movement of cadmium into the stele [29]. In *S. mucronatus* Cr, Cd, As Ni and Pb are accumulated primarily in the root system which is the strategy developed to tolerate metals phytotoxicity by limiting upward transport of metals which corresponds to the findings of [18], [30]. Deng *et al.*, [12] reported that emergent species accumulated high concentrations of metals in their roots but much less in their shoots, and the accumulation increased further with increased external concentration which is in accordance with the present study for *S. mucronatus*.

**Table 6:** Translocation Factor for Cr, Cd, As, Ni and Pb in *S. mucronatus*

Time (in days)		Metals concentration (mg/L)				
		1	2	4	8	16
2	Cr	0.28	0.17	0.19	0.13	0.53
	Cd	0.65	0.49	0.37	0.35	0.14
	As	0.33	0.39	0.58	0.56	0.70
	Ni	0.48	0.49	0.42	0.57	0.59
	Pb	0.27	0.27	0.14	0.18	0.16
4	Cr	0.26	0.46	0.23	0.32	0.34
	Cd	0.12	0.16	0.37	0.51	0.15
	As	0.26	0.42	0.54	0.74	0.60
	Ni	0.52	0.53	0.32	0.46	0.58
	Pb	0.21	0.22	0.34	0.40	0.20
6	Cr	0.58	0.41	0.28	0.26	0.23
	Cd	0.24	0.36	0.44	0.30	0.23
	As	0.37	0.28	0.37	0.19	0.27
	Ni	0.41	0.42	0.36	0.31	0.39
	Pb	0.25	0.28	0.36	0.36	0.39
8	Cr	0.68	0.31	0.37	0.30	0.17
	Cd	0.21	0.28	0.24	0.42	0.30
	As	0.61	0.89	0.84	0.75	0.44
	Ni	0.38	0.44	0.33	0.36	0.42
	Pb	0.41	0.29	0.41	0.57	0.54
10	Cr	0.26	0.04	0.08	0.23	0.12
	Cd	0.14	0.23	0.17	0.21	0.14
	As	0.50	0.50	0.54	0.58	0.72
	Ni	0.36	0.46	0.21	0.37	0.54
	Pb	0.33	0.95	0.47	0.53	0.58

In this way, in *S. mucronatus* Cr, Cd, As Ni and Pb may be taken up from nutrient medium through the roots and the concentration in the roots is almost one order of magnitude higher than leaves which shows that Cr, Cd, As Ni and Pb does not significantly moved to above-ground parts which corroborates with the findings of Baldantoni *et al.*[31].

### 5. Conclusion

The study shows that *S. mucronatus* could efficiently reduce the Cr, Cd and Pb contents in wastewater and can readily uptake in their plant parts. Based on this study, *S. mucronatus* could be a candidate for phytoremediation of Cr, Cd and Pb contaminated water. Furthermore, studies are needed to evaluate the on-site application of these plants for phytoremediation.

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## Author Profile

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