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Remediation of Heavy Metals through Aquatic Macrophytes from Water Bodies of Bundelkhand Region of Uttar Pradesh

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Abstract: *The Bundelkhand region – approximately an area of 70,000 square kilometers with 21 million people, comprising 13 districts of Madhya Pradesh (MP) and Uttar Pradesh (UP) – is facing its worst ever drought spell in living memory . There are numbers of century old historical lakes in this region which are getting polluted day by day due to the growth of the small scale industrial corridor, nutrient loading and rapid anthropogenic activities. The increasing levels of metals in the aquatic ecosystem, their entry into food chain and the overall health effects are of major concern to researchers in the field of ecology. There are out of twelve existing plants in different water bodies, four aquatic macrophytes namely Eichorniacrassipes, Pistiastratiotes, Lemna minor and Vallisneriaspiralis were selected for phytoremediation study on the basis of their abundance in selected study areas. Concentration of six trace metals have been estimated in above mentioned macrophytes i.e., Copper (Cu), Chromium (Cr), Iron (Fe), Manganese (Mn), Lead (Pb), and Zinc (Zn). From a phytoremediation perspective, E. crassipes and Pistiastratiotes are promising plant species for remediation of polluted water bodies of Bundelkhand region.*

Keywords: Bundelkhand region, *Eichhornia*, *Pistia, Lemna, Vallisneria*, Heavy metals.

Introduction

Metals occur naturally in the earth's crust and are neither created nor destroyed by anthropogenic or biological process. However, their redistribution by the minerals and metals industry (mining and smelting), power generation, fossil fuel combustion, and many other industrial processes may be of concern since an increase in metal concentrations in our environment could pose a threat to human and ecosystem health (Nriagu, 1991). As a consequence of the industrial revolution there is an enormous and increasing demand for heavy metals that leads to high anthropogenic emission of heavy metals in the biosphere (Vangronsveld and Cunningham, 1998). Nonradioactive As, Cd, Cu, Hg, Pb and Zn and radioactive Sr, Cs, and U are the most important metallic pollutants (Raskin *et al.,* 1997). These metals become an environmental concern when their concentrations begin to affect human health and the environment. A common characteristic of heavy metals regardless of whether they are biologically essential or not, they may already exert toxic effects at low concentrations (Kabata-Pendias and Pendias, 2001). Unlike organic molecules, toxic metals cannot be degraded but only be remediated. It requires consequently the intervention of mankind. In societies like our India with developing economics, the optimum development, efficient utilization and effective management of their water resources should be the dominant strategy for economic growth. But in recent years unscientific management and use of this resources for various purpose almost invariably has created undesirable problems in its wake, water logging and salinity in the case of agriculture use and environment pollution of various limits as a result of mining, industries and municipal use (Rai and Pal, 2001; Kumar *et al*, 2008; Kumar and Pal, 2011). Phytoremediation, an emerging cleanup

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technology for contaminated groundwater and wastewater that is both low-tech and low-cost, is defined as the engineered use of green plants (including grasses, forbs, and woody species) to remove, contain, or render harmless such environmental contaminants as heavy metals, trace elements, organic compounds and radioactive compounds in any aquatic systems (Kumar and Pal, 2011; Sarma, 2011; Singh *et al*., 2012). Macrophytes are considered as important component of the aquatic ecosystem not only as food source for aquatic invertebrates, but also act as an efficient accumulator of heavy metals (Devlin, 1967; Chung and Jeng, 1974; Rahman and Hasegawa, 2011; Ndimele and Jimoh, 2011; Pant *et al*., 2011). They are unchangeable biological filters and play an important role in the maintenance of aquatic ecosystem. Now a day, for cleaning the water resources, various techniques such as flocculation, sedimentation, carbon-absorption exchange etc. are available. However, the chemical and energy cost associated with these advanced techniques has been a serious constraint in adapting them in a developing country like India. In this context, a phytoremediation system which utilizes naturally occurring aquatic plants in waste water to absorb toxic metals could provide an inexpensive mean to indicate and remove toxic metals from polluted water bodies.

The present study has been conducted with fallowing objectives -

I. Analysis of physico-chemical properties of selected water bodies.
II. Observation of metal concentration in selected areas.
III. Macrophytic observations in different study areas
IV. Metal concentration of selected macrophytes in field condition.

Materials and Methods

Study Area: The Bundelkhand region lies between approximately 23º10' and 26º27' North latitude and 78º4' and 81º34' East longitudes, and comprise four districts of Chitrakut division, three districts of Jhansi division, five district of Sagar divisions and one district of Gwalior division. There are enormous historical water bodies in this region. Three aquatic bodies namely Laxmi Taal, Antiya Taal and Baruwa Sagar have been selected for present study which is shown in the map (Figure 1) and periodic water samples have been collected from these lakes of Jhansi district of Bundelkhand region of Uttar Pradesh, India which is located at 25º12' – 25º16' N and 78º18' – 79º23'E.

Laxhmi Taal (Lake): It is situated a few meters from the gate of historical Jhansi city in east. The area of lake is 0.162 km^2. It is polluted mainly anthropogenic activities as the sewage system of Jhansi directly input their organic load into this lake.

Antiya Taal (Lake): The lake is shallow with an area of 0.03 km^2 and is surrounded by residential house all side. The lake receive enormous quantity of organic matter from the nearest residential houses which causes them heavy polluted and which has accelerated the process of eutrophication.

Barua Sagar (Lake): Barua Sagar is a historical place located about 25 km from Jhansi in Uttar Pradesh, India. It is situated on the bank of the Betwa River; the place is named after the Barua Sagar Taal, a large lake created about 260 years ago when Raja Udit Singh of Orchha built the embankment. Area is 4.64 km^2; altitude is 210 mtrs above MSL.

Preservation and analysis of water samples were based on standard methods shown in Table 1 and by the method of American Public Health Association (APHA, 2005).

Analysis of Heavy Metals in Water: The water samples were taken in evaporating dishes and acidified to methyl orange with conc. HNO_3. Further 5 ml conc. HNO_3 was added and evaporated to 10 ml. Then it was transferred to a 125 ml conical flask. 6 ml of conc. HNO_3 and 2 mL $HClO_4$ (70%) were added. After that heated gently, till white dense fumes of $HClO_4$

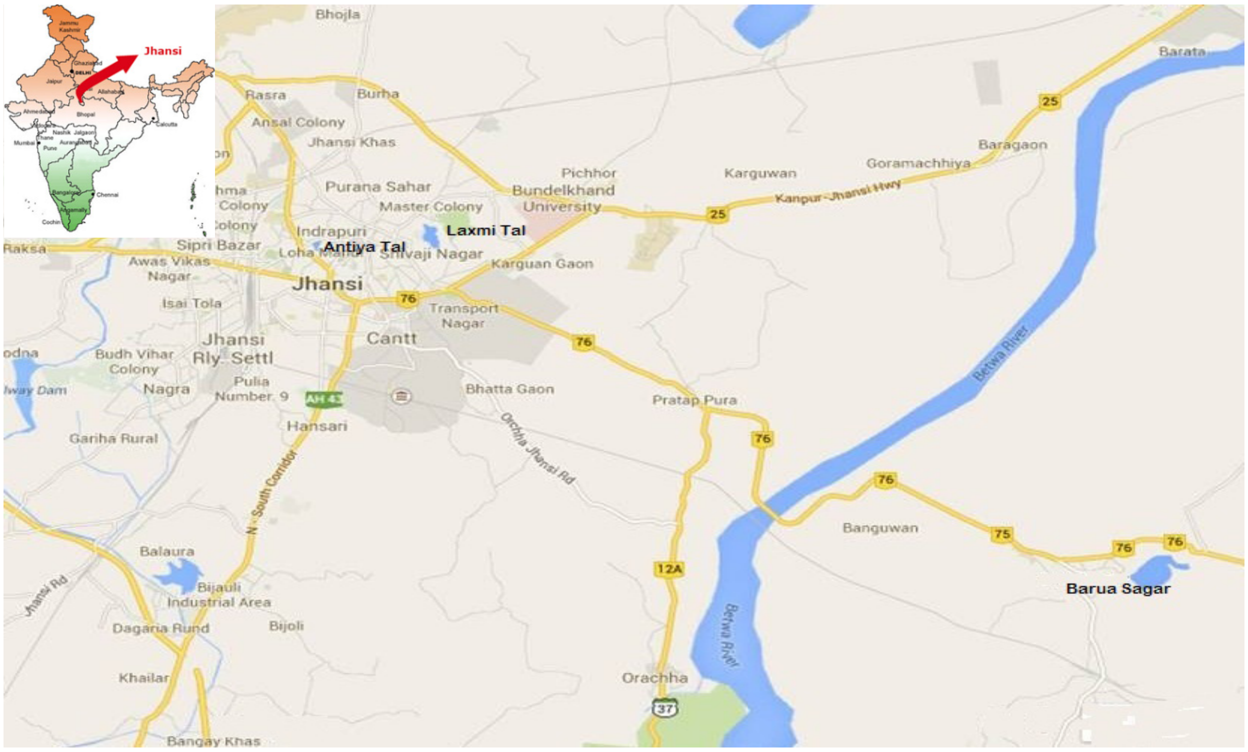

Fig. 1 Location of the selected water bodies for study.

appear. The digested samples were cooled at room temperature, filtered through Whatman No. 42 and finally the volume was made up to 100 ml with double distilled water. The solution was used for the determination of heavy metals. The metals were analyzed by Atomic Absorption Spectrophotometer (Perkin Elmer).

Plant Sampling: After the identification of tolerant and sensitive aquatic macrophytes from study areas, four existing aquatic macrophytes were selected on the basis of their abundance as well as passive biomonitors for estimating the toxicity status induced by heavy metals.

The plant species *Eichhornia crassipes* (Mart.) Solms, *Pistieastratiotes* L., *Lemna minor* L., *Vallisneria spiralis* L. have been selected for the study of heavy metal content. Healthy aquatic plants were collected by hand, washed with lake water then after distilled water to remove periphyton and sediment particles. The collected plant species were preserved in plastic bags, labeled carefully and brought to the laboratory.

Analysis of Heavy Metals in Aquatic Plant Species: Heavy metals were analyzed in harvested plants which were thoroughly washed with distilled water, and dried in an oven at 80ºC for 48 hr. Dried plant tissue (1 gm) were digested in HNO_3 (70%) and $HClO_4$ (70%) (3:1 v/v). Heavy metals in aquatic plants species was estimated by using Perkin – Elmer atomic absorption spectrophotometer.

Results and Discussion

Present investigation following water bodies e.g. Laxmi Lake, Antiya Lake, and Baruwa Sagar respectively have been selected in Jhansi district of Bundelkhand region. Observations and finding of present work have been tabulated and are as follows:

Physico-chemical Properties in Different Water Bodies: The data of physico-chemical properties of respective study areas are presented in the Table 2. On the basis of eighteen physico-chemical parameters it may

Table 1 Methodologies and instruments used for analytical work.

1	pH	Electronic	Microprocessor based Portable Soil & Water Analysis kit / Jyoti
2	Electrical Conductivity (EC)	Electronic	Microprocessor based Portable Soil & Water Analysis kit / Jyoti
3	Temperature	Electronic	Microprocessor based Portable Soil & Water Analysis kit / Jyoti
4	Total Hardness	Titration method	-------------------------
5	Alkalinity	Titration method	-------------------------
6	Total Dissolved Solid (TDS)	Electronic	Microprocessor based Portable Soil & Water Analysis kit / Jyoti
7	Dissolved Oxygen (DO)	Electronic	Microprocessor based Portable Soil & Water Analysis kit / Jyoti
8	Biochemical Oxygen Demand (BOD)	Winklers iodometric method	BOD Incubator
9	Chemical Oxygen Demand (COD)	Reflux digestion method	COD Incubator
10	Nitrate	Spectrophotometric method	UV Visible Spectrophotometer / Elico, SL 159
11	Phosphate	Spectrophotometric method	UV Visible Spectrophotometer / Elico, SL 160
12	Sulphate	Spectrophotometric method	UV Visible Spectrophotometer / Elico, SL 161
13	Sodium	Flame photometric method	Flame Photometer / Systronic 130
14	Potassium	Flame photometric method	Flame Photometer / Systronic 131
15	Calcium	Titration method	--------------------------
16	Magnesium	Titration method	--------------------------
17	% Na	Formulated	--------------------------
18	Sodium Absorption Ratio (SAR)	Formulated	--------------------------
19	Chlorophyll and Carotenoid estimation	Centrifugation & Spectrophotometric method	Remi R-4C DX & UV Visible Spectrophotometer / Elico, SL 161
19	Heavy Metals	AAS method	Atomic Absorption Spectrophotometer / Perkin Elmer,

be concluded that Laxmi Taal is more contaminated and followed by Antiya Taal and Baruwa Sagar respectively. This is because discharges of domestic and small scale industries of Jhansi city are come to the Laxmi Taal and Antiya Taal directly through the inlets. Baruwa Sagar is less polluted due to the far away of populated areas and only agricultural discharges are coming in to this lake through runoff from in and around adjacent agricultural lands and could be use for irrigation only after minor treatment (Maurya *et al.,* 2012).

Metal Concentration in Different Water Bodies: The data of heavy metal concentration in water of respective study areas are presented in the Table 3. Among nine metals, except Al and Cd all are found that the beyond the limit of Indian standard. Presence of Pb in water may be recognized more harmful for any organisms. The average concentration of lead was significantly higher (1.526 mg/l) in Laxmi Lake and followed by Antiya Taal. The high level of Pb in water of lake could be attributed to the small scale industrial and agricultural discharge and

Table 2 Physico-chemical properties in water of selected areas.

Sl. No.	Parameters	Laxmi Taal	Antiya Taal	Baruwa Sagar
01.	Temp	24.29 ± 0.045	24.18 ± 0.078	16.95 ± 0.016
02.	pH	7.46 ± 0.020	7.44 ± 0.025	7.525 ± 0.008
03.	EC	931 ± 0.523	629 ± 0.762	481.7 ± 1.23
04.	Al	240 ± 0.843	235 ± 0.432	60.975 ± 0.67
05.	TH	197 ± 0.432	242 ± 0.467	111.475 ± 0.92
06.	NO_3	1.58 ± 0.010	1.49 ± 0.03	1.975 ± 0.03
07.	DO	3.92 ± 0.02	5.85 ± 0.021	7.025 ± 0.001
08.	Mg	19.3 ± 0.03	22.24 ± 0.08	26.9 ± 1.12
09.	TDS	625 ± 0.657	437 ± 0.467	229 ± 2.32
10.	PO_4	0.54 ± 0.02	0.75 ± 0.02	0.717 ± 0.015
11.	SO_4	18.0 ± 0.02	21.7 ± 0.02	16.4 ± 1.73
12.	BOD	2.57 ± 0.035	4.42 ± 0.037	3.925 ± 0.08
13.	COD	185 ± 0.656	143 ± 0.873	35.25 ± 1.31
14.	Na	263 ± 0.63	246 ± 1.34	258.5 ± 1.43
15.	K	44.3 ± 0.04	38.7 ± 0.05	39.25 ± 1.14
16.	Ca	42.3 ± 0.10	56.0 ± 0.06	48.575 ± 0.08
17.	Na%	69.7 ± 0.02	67.73 ± 0.65	69.245 ± 0.92
18.	SAR	47.50 ± 0.168	39.30 ± 0.459	42.395 ± 0.06

Values are Mean ± SE (n = 3); Units: - Concentration in mg/l, except pH; Temperature (ºC); EC (µS/cm); SAR (meq/l)

dust which holds a huge amount of lead from the combustion of petrol in automobile cars. Because of Batteries, radiators for cars and trucks, and some colors of ink also contain lead which also had been available nearby this lake and flow through drainage in these lakes. Similar observation was found by Hoo (2004) in Labu river of Malayasia; Egborge (1994) in Warri river in Nigeria, Yadav and Kumar (2011) in Kosi river; Zaidi *et al*. (2011) in Betwa and Pahuj rivers of India.

Macrophytic Observations in Different Study Areas: The availability of macrophytic composition in selected aquatic bodies and their variation of abundance in respective study areas are depicted in the Table 4 and summarized as follows -

Aquatic macrophytic diversity and its role in understanding the fresh water ecosystem dynamics have tremendous significance. In present investigations an initiative have been taken to identify the tolerant and sensitive aquatic macrophytes grown in different selected study areas during the year 2013 to 2014 respectively. There are total twelve species of aquatic macrophytes have been found in selected study areas such as *Eichhornia crassipes* (Mart.) Solms*, Hydrilla verticillata* (L.F.) Royle*, Ipomoea aquatic* (Forssk.), *Vallisneria spiralis* (L.), *Najasgraminea* (Del.), *Pistiastratiotes* (L.), *Typhadomingensis* (Pers.), *Nymphaea lotus* (L.), *Potamogetoncrispus* (L.), *Chara spp.* (L.), *Cyperus spp.(*L.), and *Lemna spp.* (L.).

All the twelve above mentioned species have been found in Laxhmi Lake and except *Najas Species* remaing eleven species were also found in Antiya taal and Baruwa Sagar although they showed variation in abundance according to the seasonal variations. *Eichhornia crassipes* was found abundantly in almost every season throughout the experimental period of the year. *Hydrilla verticillata, Vallisneria spiralis, Chara spp.* and *Lemna Spp.* was found in winter and pre-monsoon seasons.

Table 3 Average Metals concentrations in water of selected areas.

Sl. No.	Metals	Laxmi Taal	Antiya Taal	Baruwa Sagar	Indian Standard
01.	Al	0.0075 ± 0.0003	0.006 ± 0.001	0.018 ± 0.001	NA
02.	Cd	0.0015 ± 0.0003	0.002 ± 0.0005	0.002 ± 0.0001	.01
03.	Cr	0.338 ± 0.007	0.161 ± 0.013	0.064 ± 0.002	.05
04.	Cu	0.070 ± 0.008	0.015 ± 0.005	0.016 ± 0.001	.05
05.	Fe	1.497 ± 0.011	1.304 ± 0.005	0.672 ± 0.012	.3
06.	Pb	1.526 ± 0.024	1.187 ± 0.001	1.174 ± 0.002	.05
07.	Mn	1.649 ± 0.023	1.846 ± 0.021	2.695 ± 0.003	.05
08.	Ni	1.429 ± 0.017	NA	NA	.02 *
09.	Zn	0.020 ± 0.006	0.007 ± 0.001	0.076 ± 0.004	5

Values are Mean ± SE (n = 3); Unit: - concentration in mg/l, NA = not available *WHO Standard

Table 4 Identification of tolerant and sensitive aquatic macrophytes grown in the study areas.

Sl. No.	Species name	LaxmiTaal	AntiyaTaal	BaruwaSagar
01.	*Eichhorniacrassipes*(Mart.) Solms	+++	+++	++
02.	*Hydrillaverticillata*(L.F.) Royle	++	++	+++
03.	*Ipomoea aquatic* (Forssk.),	+	+	++
04.	*Vallisneriaspiralis* (L.)	++	+	+++
05.	*Najasgraminea* (Del.)	+	NA	NA
06.	*Pistiastratiotes*(L.)	+	+	++
07.	*Typhadomingensis* (Pers.)	+	+	+
08.	*Nymphaea lotus* (L.)	+	+	++
09.	*Potamogetoncrispus* (L.)	++	+	+++
10.	*Chara spp.* (L.)	+	+	++
11.	*Cyperus spp.* (L.)	+	+	+
12.	*Lemna spp.* (L.)	++	++	+

+++ = High, ++ = Medium, + = Low; NA = Not Available

Metal Concentration in Plant Species (macrophytes) in Field Condition: The data of heavy metal concentrations in selected plant species in the field condition are presented in the Table 5. Metals accumulation by selected plant species have been compared with the average concentrations of metals in the water bodies and depicted in Figure 2. There are four aquatic macrophytes namely *Eichornia crassipes, Pistia stratiotes, Lemna minor* and *Vallisneria spiralis* were selected for phytoremediation study on the basis of their abundance in selected study areas. Concentration of six trace metals have been estimated in above mentioned macrophytes i.e., Copper (Cu), Chromium (Cr), Iron (Fe), Manganese (Mn), Lead (Pb), and Zinc (Zn) for their bioaccumulation abilities.

In recent years, it has been reported that some plants species; known as hyperaccumulators, which are usually present in heavy metal-contaminated areas, have the ability to accumulate unusually high concentration of heavy metals without dramatically being physiologically impacted (Hu *et al.*, 2010). The level of toxic metals (Pb, Cr, Hg etc) can be reduced from contaminated water by a number of aquatic plants taken up by the roots system and transported to the stems and leaves without showing toxicity syndrome has confirmed by many studies (Rai *et al.*, 1995; Cardwell *et al.*, 2002; Abidal and Harikrishna 2010; Sarma, 2011). Maximum accumulation of Cu, Fe, Cr, Pb and Zn was recorded in *Eichornia crassipes* where as Mn was observed maximum in *Pistia stratiotes*. Therefore as per Table 5 and Figure 2, among four mentioned species *Eichhornia crassipes* may be consider one of the promising species followed by *Pistia stratiotes*, *Vallisneria spiralis* and *Lemna Minor* respectively for phytoremediation of heavy metals from aquatic bodies of Bundelkhand region.

The lakes and reservoirs, all over the study area without exception, are in varying degrees

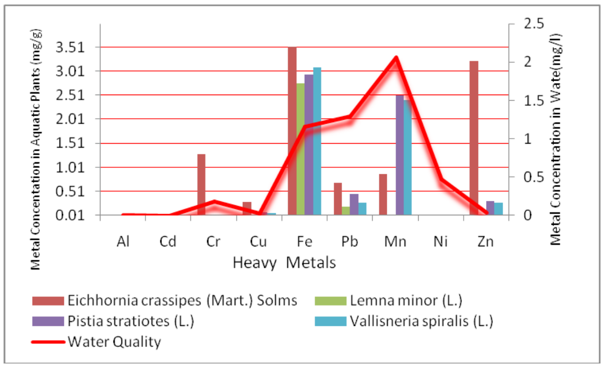

Fig. 2 Metal accumulation in various plants in response to metal content in water.

of environmental degradation. The degradation is due to encroachments eutrophication (from domestic/municipal and small scale industrial discharges). There has been a quantum jump in population during the last century without corresponding expansion of civic facilities resulting in lakes and reservoirs, especially the urban ones, becoming sinks for contaminants. Finally it may be concluded that, accelerated eutrophication is a widespread and significant threat to aquatic water bodies of Bundelkhand region. Among the most threatened Lakes are those located in or near urban settlements (Laxhmi and Antiya Lake) which can result in a rapidly increasing nutrient load in Lake because of uncontrolled point and non-point effluent as well as domestic discharges. Thus, although lake ecosystems constitute an essential resource for many ecosystem services and human activities, urban lakes often exhibit serious degradation that interferes with these services and benefits. Present work suggests that, phytoremediation has become an effective and affordable technological solution used to extract or remove inactive metals and metal pollutants from contaminated water bodies. This technology is environmental friendly and potentially cost-effective. Hence, native species of macrophytes like *Eichhornia crassipes, Pistia stratiotes, Vallisneria spiralis*and *Lemna spp.* were efficient and cost effective for accumulation of heavy metals from contaminated water bodies of this region and they may be recommended as phytoremediator species.

Table 5 Average metals concentration in selected aquatic macrophytes grown in the study areas.

Sl. No.	Metals	Eichhornia crassipes (Mart.) Solms	Lemna minor (L.)	Pistia stratiotes (L.)	Vallisneria spiralis (L.)
01.	Cu	0.283 ± 0.002	ND	0.073 ± 0.002	0.052 ± 0.003
02.	Fe	3.528 ± 0.01	2.758 ± 0.05	2.946 ± 0.05	3.083 ± 0.04
03.	Cr	1.285 ± 0.04	ND	ND	0.025 ± 0.001
04.	Mn	0.875 ± 0.006	0.001 ± 0.0001	2.512 ± 0.013	2.409 ± 0.07
05.	Pb	0.691 ± 0.02	0.190 ± 0.003	0.454 ± 0.006	0.273 ± 0.006
06.	Zn	3.223 ± 0.007	0.026 ± 0.001	0.302 ± 0.01	0.264 ± 0.002

All values are mg/L (ppm)

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