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Research paper

Phytoremediation of battery industry effluent through aquatic macrophytes

Mahrine Rashid [1] *, Iftilkhar Ahmad [1], Ashique Muhammad [1], Mahmood Ul Hassan [1]

[1] *Department of Plant and Environmental Protection Pakistan. PARC Institute of Advance Studies in Agriculture. National Agricultural Research Centre, Islamabad The University of Agriculture, Peshawar, Pakistan*
**Corresponding author E-mail: mahrine.rashid@gmail.com*

Abstract

Research was conducted to quantify the level of copper (Cu), chromium (Cr), cadmium (Cd) and lead (Pb) contamination in battery industry effluent and to assess the remediation potential of three invasive aquatic macrophytes *Eichhornia crassipes*, *Pistia stratiotes* and *Hydrocotyle umbellata* by growing on industrial effluent collected from Lead acid Battery industry.
The effluent was heavily contaminated with Pb (10mg/l) and sulphuric acid (pH 2- 2.1). Due to high Pb concentrations and low pH (2-2.2) the plants were unable to survive. Mortality rate of *E. crassipes* was 96 % while *P. stratiotes* and *H. umbellata* were 100% rotten. The experiment was repeated after adjusting the effluent pH to 7-7.5 to increase the plant life.
Plant parts and wastewater samples were analyzed after every 3 days interval uptil 21st day. The amount of Cr, Cd and Cu in the effluent was 0.076 mg /L, 0.036 mg /L and 0.097 mg /L, which was in permissible limits of NEQs (1.0 mg/l, 0.1 mg/l and 1.0 mg/l) respectively. Pb was found 10 times higher i.e. 10 mg/l than the permissible limit 0.5 mg/l. *E. crassipes* removed Pb>Cr>Cu>Cd while *P. stratiotes* and *H. umbellata* reduced Cd, Cr, Cu more than Pb from the effluent. *E. crassipes* was most efficient Pb removing plant in 21 days of experiment.

Keywords: *Battery Industry; Effluent; Invasive Aquatic Macrophytes; Pb; Phytoremediation; Water Hyacinth.*

1. Introduction

Heavy metals are among most hazardous environmental pollutants. Besides natural activities, almost all human activities also contribute to production of metals as a side effect. Migration of these pollutants to contaminated sites such as effluents, sewage, dust or leaching into the soil is a common examples of events contributing to infection (Bieby Voijant Tangahu et al., 2011). Contamination of soil, surface and groundwater is key environmental problem causing accumulation of heavy metals and many other pollutants resulting in the deterioration of biological ecosystems.
Heavy metals such as lead (Pb), chromium (Cr), zinc (Zn), cadmium (Cd), copper Cu), and nickel (Ni) are contaminating forms, which are produced in large quantities by improper disposal of batteries, household, industrial, agricultural and mining waste and eventually causing severe environmental impact. These components produce bioaccumulation effects, categorizing them as precarious elements that are necessary to be removed from ecosystem (Cleide Barbieri de Souza and Gabriel Rodrigues Silva, 2019). Toxic heavy metals which include Pb, Co, Cd may be differentiated from other pollutants, on the grounds that they can't be biodegraded but may be accrued in living organisms, therefore causing various diseases and issues even in extraordinarily decrease concentrations (Bieby Voijant Tangahu et al., 2011).
Main focus of the current study is phytoremediation of Battery industry effluent. Lead acid batteries have become a widespread power source for commercial, household and industrial applications but the use of non-toxic and non-hazardous substitute materials has not rapidly developed (Buchmann, 2011). According to a 2005 estimate, the worldwide battery industry generates US $ 48 billion in sales each year (MIT, 2009) with 6% once a year growth. In Pakistan, there are 39 battery manufacturing industries dealing with different types of batteries (http://www.pakbd.com/company).
Battery wastewater is distinguished by heavy metals chiefly, like lead (Pb), cadmium (Cd), chromium (Cr), Copper (Cu), mercury (Hg), arsenic (As) etc and, chlorine and sulphates, biological oxygen demand (BOD), chemical oxygen demand (COD) and total dissolve solids (TDS). Level of the pollutants in lead acid battery wastewater depends on process of battery manufacturing (Ramus and Hawkins, 1992). Batteries contain acerbic effects that can cause burns and are hazardous to eyes and skin (MIT, 2009).
Lead is an ancient metal recognized and used by humans; nevertheless it is severely lethal to humans and aquatic life. The high levels of lead (Pb) may result in toxic effects in humans which in turn cause problems in the synthesis of blood (Hb), defect kidneys, reproductive system, gastrointestinal tract (GIT), joints, and severe damage to nervous system (Govind and Madhuri, 2014). Workers exposed to lead were found to be impaired respiratory function and with elevated blood lead and zinc protoporphyrin concentration (Jurdziak et al., 2015). A study conducted on Uruguayan in first grade children showed oxidative stress by low level lead exposure, signifying its potentially adverse effects (Roy et al., 2015). Excess Pb toxicity may cause number of morphological and physiological impacts in plants e.g. stunted growth, chlorosis blackening of root system, it inhabits photosynthesis, imbalance nutrition and water balance etc in plant (Pallavi and Dubey, 2005). Chromium (Cr) is toxic and produces anaemia in human and change in hematological parameters in the L. rohita fish (Praveen et al., 2013). The cadmium (Cd) may cause increased blood pressure, kidney dysfunction, and respiratory problems. Similarly,

exposure to nickel (Ni) can cause decrease in body weight, heart and liver damage, and skin irritation. Mutagenicity, carcinogenicity, teratogenicity, immunosuppression, poor body condition and impaired reproduction are toxic effects of heavy metals (Govind and Madhuri, 2014).

Bioaccumulations of large quantities of heavy metals and its toxic effects on biota has led to various research studies and methods for management and treatment of heavy metal polluted water (Gupta, 2013).

Expensive conventional methods and environmental hazards have enforced scientists to explore cost effective ways to remove heavy metals from effluent. For similar reasons, biologists have focused on accumulation of heavy metals by aquatic macrophytes and reported bioremediation through plants is effective way of treating industrial effluent. (Licina et al., 2007). Several aquatic macrophytes such as *Eichhornia crassipes*, Lemna sp., Azolla sp., Spirodella sp. and Pistia sp. Are frequently and effectively have been used for removing heavy metals from different industrial effluent (Gupta etl. 2012; Saha et al 2015)

Plants utilized for phytoremediation must possess capability of rapid growth, high biomass, hairy and deep-root system, and high bioaccumulation coefficient. (Pooja and Kaushal , 2018) . The free floating/floating-leaved aquatic plants like water lettuce (Pistia stratiotes), common salvinia (Salvinia minima), water hyacinth (*Eichhornia crassipes*), duckweed (Lemna spp. and Spirodela polyrrhiza), pennywort (*Hydrocotyle umbellata*) are suitable candidates for heavy metal removal (Cleide Barbieri de Souza and Gabriel Rodrigues Silva. 2018, John et al., 2008; Maine et al., 2004; Mishra et al., 2008; Sanchez-Galvan et al., 2008).

These aquatic plants have ability to absorb high levels of contaminants concentrated either in their roots, shoots and/or leaves (Raksin et al., 1994; Cunningham and Ow 1996; Baker et al., 1994). Baker and Brooks (1998) studies revealed that metal accumulated in plants contain more than or up to 0.1% i.e. > 1000 mg/g of copper, cadmium, chromium, lead, nickel. For cadmium and other rare metals, it is > 0.01% by dry weight. Therefore, macrophytes have been frequently experienced for heavy metal removal (Mishra and Tripathi, 2008). Several aquatic species like water pennywort (Hydrocotyle umbellate L.), duckweed (Lemna minor L.) (Mo et al., 1989) and water hyacinth (*Eichhornia crassipes* Mart. Solms have shown ability to remove heavy metals from water, (Zhu et al., 1999).

In this study, the research objective are to quantify levels of heavy metals i.e. Lead (Pb), Copper (Cu), Chromium (Cr), Cadmium (Cd) in the wastewater of battery industry, evaluation of remediating efficacy of three aquatic plants i.e. Pistia stratiotes, *Hydrocotyle umbellata* and *Eichhornia crassipes* and to demonstrate their capability of removing heavy metals (Pb, Cu, Cr, Cd) from the wastewater and screen out best candidate specie for management of battery industry effluent. These metals were selected for their toxic properties, their capability to contaminate environment and their high indices in effluents. The selected plants were reported most abundant, fast growing and prominent in research studies.

2. Materials and methods

2.1. Study area

Hattar Industrial Estate is an area of 1,063 acres (containing 215 houses operational and 378 closed, 162 under construction and 98 sick industrial units); this industrial estate encloses chemical, vegetable oil manufacturing, steel, paper, cement, marble, pharmaceutical, textile, Lead acid battery, poultry feed and beverages industries. According Environmental Protection Agency (EPA) report these factories are key source of pollution in area (Sial et al., 2006). The area is surrounded by natural surface drains and the effluents of all the factories are disposed in these aquifers and ultimately fall into the Haro River.

Fig. 1: Google Maps Image

2.2. Material and method

2.2.1. Water samples collection and analysis

Untreated industrial effluents of Lead acid Battery industry, Taxila were collected from the study site and stored in polythene bottles for initial characterization and phytoremediation experiments. The samples were subjected to nitric acid digestion using the microwave-assisted technique, setting pressure at 30 bars and power at 700 Watts (Lokhande et al., 2011). Standard methods and procedures (APHA 2001) were used to analyze physiochemical properties, pH and temperature. Copper (Cu),Chromium (Cr),Cadmium (Cd) and Lead (Pb) was analyzed by using Atomic Absorption Spectrometry Perklin Elmer- AAnalyst 700 at Water and Soil Analysis Lab according to methods (Clesceri et al., 1998).

i) Plant analysis:

Plants (Water Lettuce, Pennywort and water hyacinth) were acclimatized in plastic tubs with fresh water in 45 days. After 45 days twenty five plants of each species (Water Lettuce, Pennywort and water hyacinth) were set in effluent (at least 20 lit/ tub) and allowed to stay in effluent for 21 days (Ganjo and Khwakaram, 2010). Plant samples from each experimental tub were harvested and analyzed according to standard methods of APHA (1998) for heavy metal Copper (Cu), Chromium (Cr), Cadmium (Cd) and Lead (Pb) at Land Resource Research Institute (LRRI) PARC, Islamabad by Atomic Absorption Spectrometry (AAS).

Plant species were for heavy metals For heavy metal analysis the plant samples were washed with distilled water, air dried and placed in oven at 70-80˚C for 48 hours (Rathor et al, 2014). The powder form of different parts of plants was digested using double acids mixture (nitric and perchloric acid) and filtered through Whatman 42 filter paper. After nitric and perchloric acid digestion, the solution was transferred to 50 ml volumetric balloon and reading under taken by Atomic Absorption Spectrometer for heavy metals. Analysis of heavy metals was done after every 3 days for at least 21days as performed by Upadhyay et al., 2007.
The pH of effluent was 2.1 which was adjusted to 7- 7.5 for plant survival. The heavy metal uptake from contaminated battery effluent by Water Lettuce (Pistia stratiotes), Pennywort (*Hydrocotyle umbellata*.), and water hyacinth (*Eichhornia crassipes*) was recorded. Experiment was performed in triplicate under natural conditions for 21 days Hydraulic Retention Time (A. R. Upadhyay et al., 2007). The effluent and plants were sampled periodically after every 3 days for a period of 21 days and analyzed for physicochemical parameters (Tangahu et al., 2011).

ii). Translocation Factor:

Translocation factor (movement ability of plants to accumulate heavy metal in shoots and roots) was calculated by equation TF= $\left(\frac{Cs}{Cr}\right)$i, where i denotes the heavy metal. Cr represents the amount of trace element accumulated in the roots (mg/ kg) and Cs represents the amount of trace element accumulated in the shoots (mg/ kg) (Sivalingam et al., 2016).

iii). Bioconcentration Factor :

Bioconcentration (amount of heavy metal in plant tissue and in surrounding environment) was calculated by equation BCF= $\left(\frac{P}{E}\right)$i, Where i denote the heavy metal, P represents trace element concentration in plant tissues (mg/ kg dry wt.); E represents the trace element concentration in the water (mg/L) or in the sediment (mg/ kg) (Sivalingam et al., 2016).

2.2.2. Statistical Analysis

Three aquatic plant species with three replicates were arranged in completely randomized design (CRD). The data was statistically analyzed using two factor (Plant species and time) factorial experiment under CRD. Analysis of variance (ANOVA) of the measured parameters was performed using Statistix 8.1 or Minitab 15 software. Results were represented in graphs and tables with the help of Microsoft Excel 2010.

3. Results

Analysis of effluents for the baseline data showed Pb (lead) to be ten times higher than permissible limits (0.5 mg/l) either for effluent disposal to environment or for agricultural usage. Table 1 showed the pH value range from 2.1 -2.3 which is under the permissible limits (highly acidic) described by National Environmental Quality Standards (NEQS) 6-10 at constant temperature value 30-35^0 C. Cr, Cd and Cu in effluent was 0.076 mg /L, 0.036 mg /L and 0.097 mg /L respectively which is in the permissible limits of NEQs (1.0 mg/l, 0.1 mg/l and 1.0 mg/l) respectively. Waste water characteristics are listed in Table 1.
Experiment was started by adjusting pH to 7.0 -7.5 using NaOH (1 liter effluent + 7ml of 1M NaOH). 25 plants of *E. crassipes, P. stratiotes* and *H. umbellata* were placed in effluent. Plants and water samples were taken for the analysis of heavy metal after every third day of the experiment (0, 3, 6,9,13, 15, 18 and 21). Positive control was set with tap water and experimental plants.

Table 1: Physico-chemical analysis of the effluent of battery industry

Parameter	Effluent range	NEQs for industrial effluent *	Agriculture standard values (WHO)**
Temperature	30-35^{0}C	40 ^{0}C	40 ^{0}C
pH	2.1	6-10	6.5-8.5
TDS	9930	3500 mg/l.	450-2000 mg/l
Pb	10 mg /L	0.5 mg/l	> 0.5 mg/l
Cd	0.036 mg /L	0.1 mg/l.	0.01 mg/l
Cr	0.076 mg /L	1.0 mg/l.	0.1 mg/l
Cu	0.097 mg /L	1.0 mg/l.	0.01 mg/l

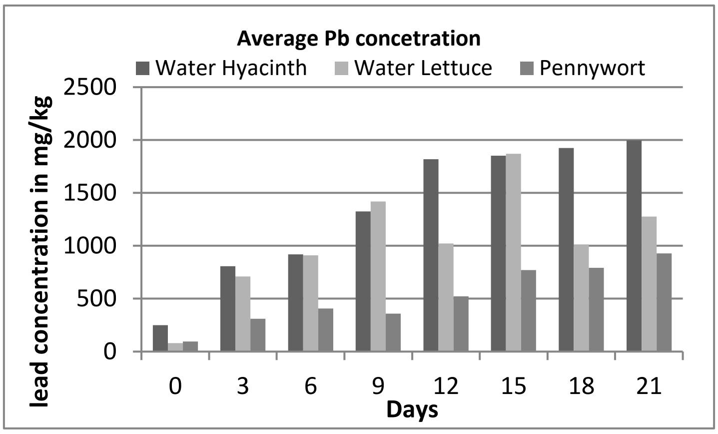
* National environmental quality standards (NEQs) for municipal and liquid industrial effluents (mg/l; the Gazette of Pakistan, Extra, August 10, 2000.

**World Health organization, Guidelines for drinking water quality. 1993.

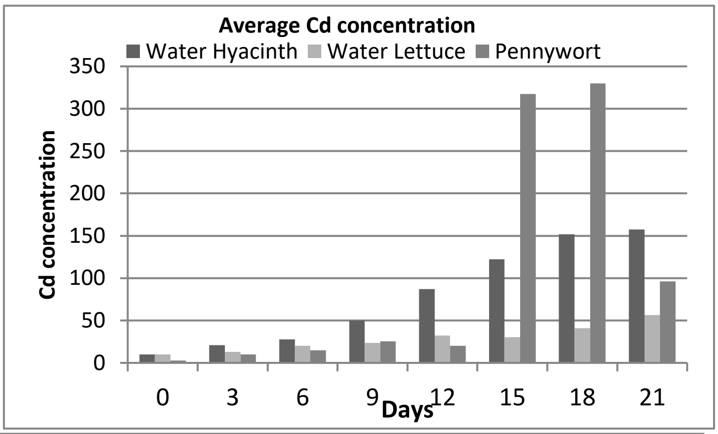
3.1. Average concentration of heavy metals in plants

The average amount of lead in *E. crassipes* and *P. stratiotes* (1999 mg/kg, 1276 mg/kg) respectively were higher than pennywort (928 mg/kg). The average amount of Cd was higher in *E. crassipes* (157.5 mg/kg) than *H. umbellata* and *P. stratiotes* (96 mg/kg, 56.4 mg/kg). Cr was found higher in *P. stratiotes* (210 mg/kg) than *E. crassipes* and *H. umbellata* (182.6 mg/kg, 149 mg/kg). Cu was found higher in *H. umbellata* (52 mg/kg) than *P. stratiotes* and *E. crassipes* (40.3 mg/kg, 30.2 mg/kg) (Fig: 1; A, B, C, D)

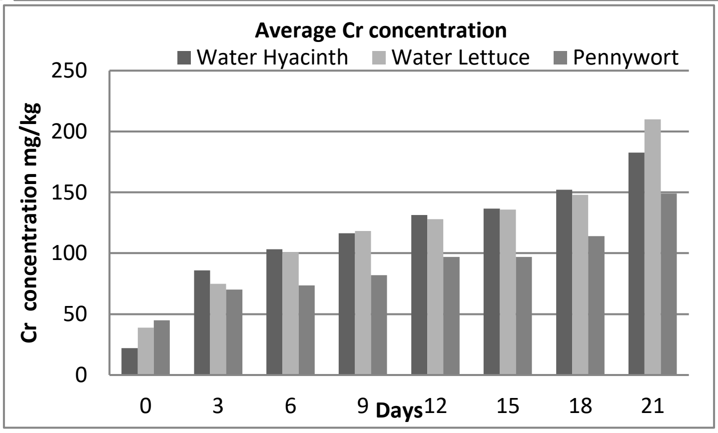
Figure 1: Concentration of heavy metals in plants



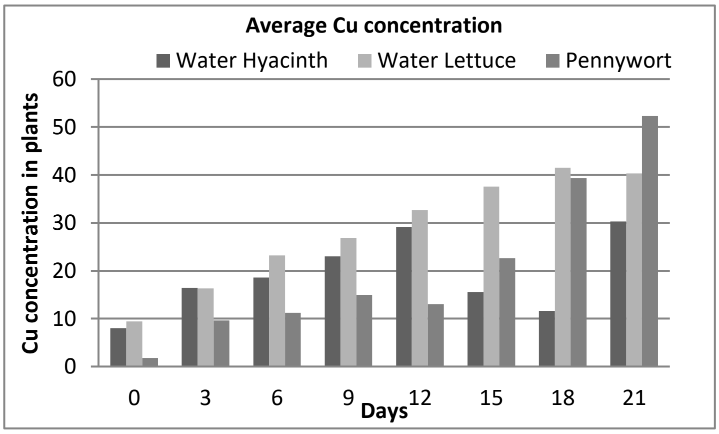
A



B



C



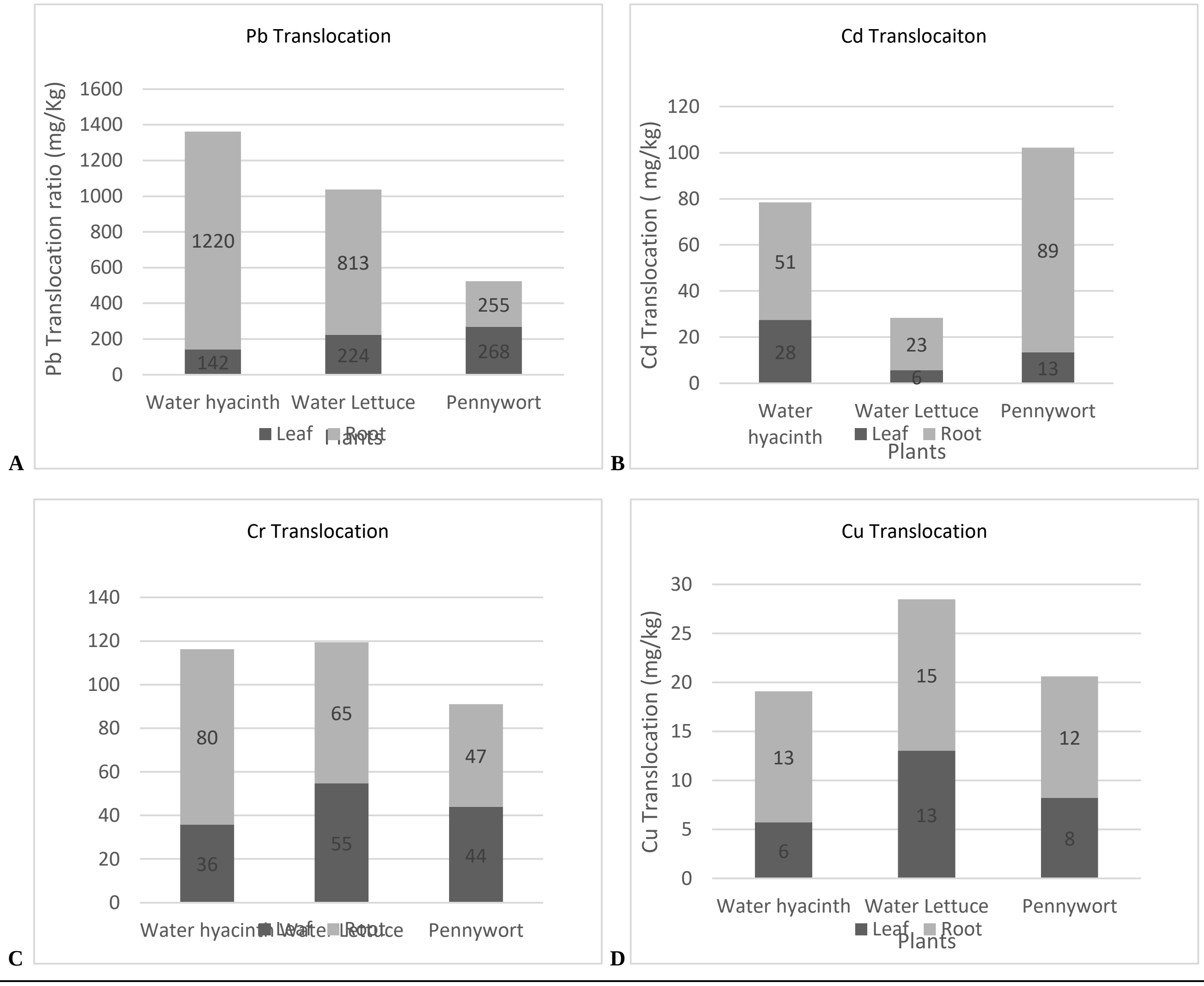
D

3.2. Translocation Factor (TA)

Figure 2 (A, B, C, D) explains translocation ability of plants to accumulate heavy metal in shoots and roots.

Pb was highly accumulated in roots of *E. crassipes* (1220 mg/kg) than *P. stratiotes* and *H. umbellata* roots (813 mg/kg and 255 mg/kg respectively), while shoots of *H. umbellata* were highest accumulators of Pb (268 mg/kg) than shoots of *E. crassipes* and *P. stratiotes* (142 mg/kg and 224 mg/kg respectively). Cd was highly accumulated in roots of *H. umbellata* (89 mg/kg) than roots of *E. crassipes* and *P. stratiotes* (51 mg/kg, 23 mg/kg) while 28 mg/kg was accumulated in *E. crassipes* shoots and 6 mg/kg and 13 mg/kg in shoots of *P. stratiotes* and *H. umbellata*. Cr was more accumulated in roots of *E. crassipes* (80 mg/kg) than shoots (36 mg/kg) while 65 mg/kg and 47 mg/kg were recorded in roots of *P. stratiotes* and *H. umbellata* and in shoots 55 mg/kg and 44 mg/kg respectively. The accumulation and translocation of Cu was found to be relatively higher in roots (13 mg/kg , 15 mg/kg , 12 mg/kg) than shoots of *E. crassipes, P. stratiotes* and *H. umbellata* (6 mg/kg, 13 mg/kg, 8 mg/kg respectively).

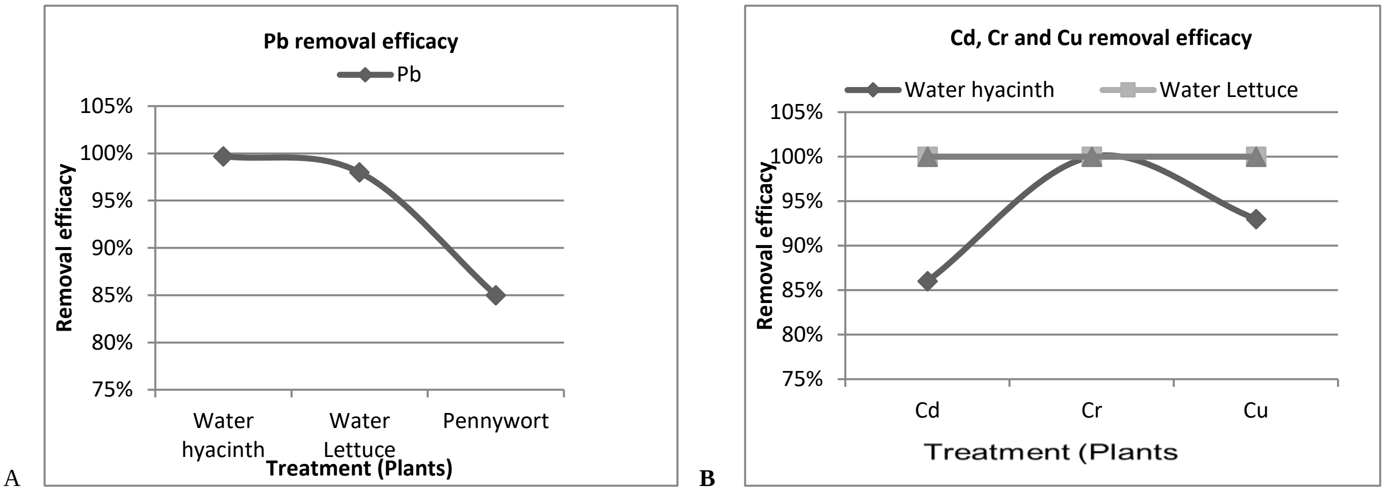
Figure 2: Translocation of heavy metals in Shoot vs. Root



3.3. Heavy metal removal efficacy

All three plants were found to be hyperaccumulator of Pb, Cd, Cr and Cu. *E. crassipes* accumulated 100% Pb and Cr , 86% Cd, 93% Cu, *P. stratiotes* accumulated 98% Pb, and 100% Cd, Cr and Cu while *H. umbellata* 85% removed Pb and 100% Cd, Cr and Cu (Fig 3 A, B). Removal of Pb from effluent was highly recorded in E crassipes with constant and gradual uptake from day 1 to day 21 (250 mg/l, 3911 mg/l) as compared to *P. stratiotes* (80 mg/l, 1276 mg/l) and *H. umbellata* (97 mg/l, 928 mg/l) respectively (Fig 4 A, B, C) in which fluctuation from day 9 to day 21 in absorption was observed.

Figure 3: Percentage removal efficacy of plants



E. crassipes absorbed 100% Pb, 86% Cd, 100% Cr and 93% Cu, *P. stratiotes* reduced 98% Pb, 100% Cd, 100% Cr and 100% Cu and *H. umbellata* reduced 85% Pb, 100% Cd, 100% Cr and 100% Cu from the effluent. *E. crassipes* removed Pb>Cr>Cu>Cd while *P. stratiotes* and *H. umbellata* reduced Cd, Cr, Cu more than Pb from the effluent (Table 2). *E. crassipes* was most efficient Pb removing plant in 21 days of experiment.

Table 2: Removal of Heavy metal from effluent

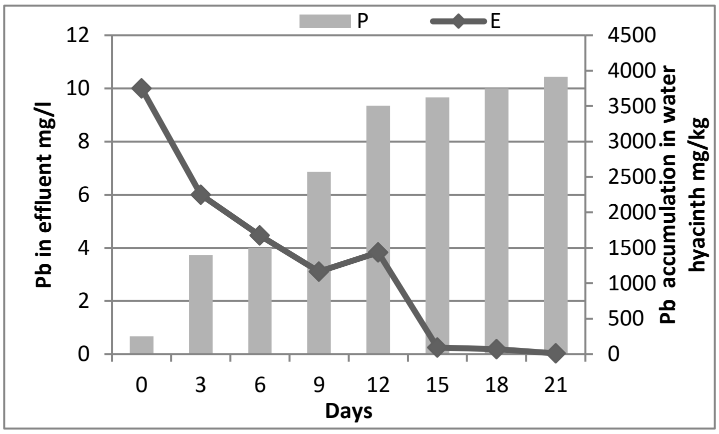
Heavy metals (HMs)	HMs in effluent 1st day (mg/l)	Plants in effluent	HMs in effluent 21st day (mg/l)	% age decrease of HMs from effluent	NEQs for industrial effluent *	Agriculture standard values (WHO)**
Pb	10 (mg/l)	*E. crassipes*	0.027	100%	0.5 mg/l	> 0.5 mg/l
		P. stratiotes	0.2	98%		
		H. umbellata	1.5	85%		
Cd	0.036 (mg/l)	*E. crassipes*	0.005	86%	0.1 mg/l.	0.01 mg/l
		P. stratiotes	0.000	100%		
		H. umbellata	0.000	100%		
Cr	0.076 (mg/l)	*E. crassipes*	0.000	100%	1.0 mg/l.	0.1 mg/l
		P. stratiotes	0.000	100%		
		H. umbellata	0.000	100%		
Cu	0.097 (mg/l)	*E. crassipes*	0.007	93%	1.0 mg/l.	0.01 mg/l
		P. stratiotes	0.000	100%		
		H. umbellata	0.000	100%		

* National environmental quality standards (NEQs) for municipal and liquid industrial effluents (mg/l; the Gazette of Pakistan, Extra, August 10, 2000.

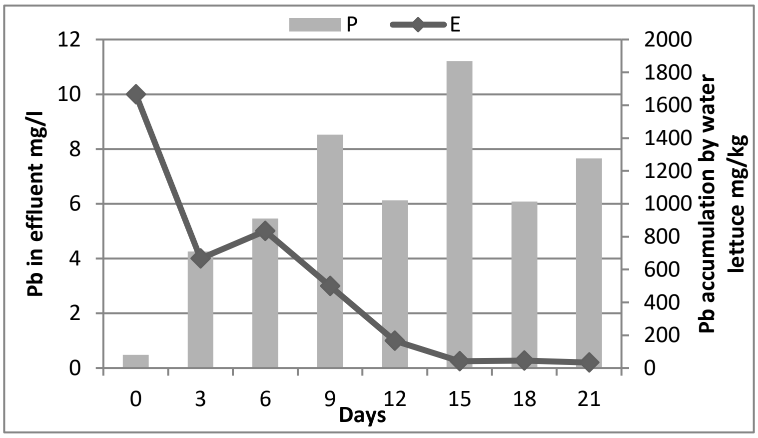
**World Health organization, Guidelines for drinking water quality. 1993

Figure 4: Pb removal from effluent and uptake by plants

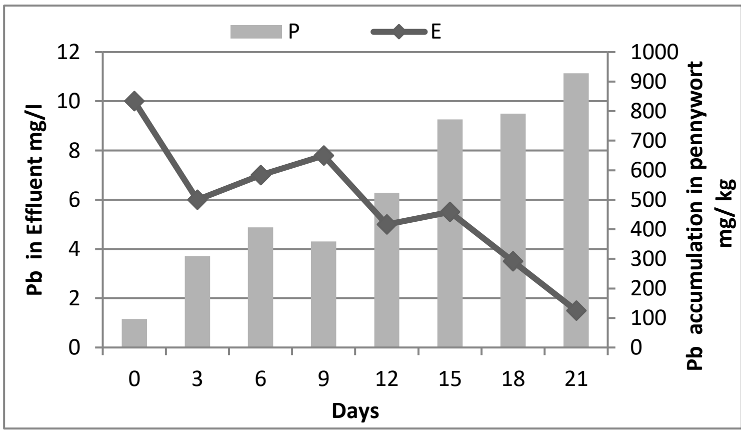
A. Water hyacinth



B. Water lettuce



C. Pennywort



Analysis of Variance (ANOVA) of the measured parameters was performed using software Statistix 8.1 and variable means were compared using Least Significant Difference (LSD). All the means were significantly different from one another. The mean maximum and minimum contents of heavy metals in aquatic plants are listed in table 3. According to data presented (Tab. 3), higher mean content of Pb was found in *E. crassipes*(1412.9) at 5% and least was absorbed by *H. umbellata*. Higher mean contents of Cd were found in *H. umbellata* (103.49) and least in *P. stratiotes* (28.35). Similarly, Cr and Cu was significantly absorbed by *P. stratiotes* (119.41, 28.47) and least absorbed by *H. umbellata* and *E. crassipes* (91.00, 19.068) respectively.

Table 3: Means showing heavy metals (Pb, Cd, Cr and Cu) accumulation values.

Treatment	Pb	Cd	Cr	Cu
E. crassipes	1412.9**A**	75.97**B**	116.32**B**	19.068**C**
P. stratiotes	958.9**B**	28.35**C**	119.41**A**	28.47**A**
H. umbellata	517.4**C**	103.49**A**	91.00**C**	20.603**B**
LSD (5%)	3.4298	1.0470	1.6813	0.9616

At the end of experiment, physical condition was recorded. Necrosis, stunted growth and lack of chlorophyll were observed in plant leaves of all plants. Symptoms of water deficiency in plants were also recorded. No new plantlets were produced neither flowering occurred while reverse conditions like new plantlets, flowering and fresh green leaves were observed in the control condition (tap water). *P. stratiotes* showed lack of new plantlet production, necrosis and rolling of leaf tips. *H. umbellata* exhibited chlorosis, stunted growth and no production of new plantlets (Plate 4). Water deficiency, no new plantlets and flowering was observed in E.crassipes leaves at the end of experiment. In the control condition all plants produced new plantlets, flowering and leaves were found fresh green indicating maximum chlorophyll contents in plants.

4. Discussion

The research carried out to decontaminate the industrial effluent by using aquatic plants *E. crassipes*, *P. stratiotes* and *H. umbellata*, making it under the permissible limits for disposal. The objective of study was to analyze the chemical composition of battery industry effluent and reduce its hazardous impacts (Heavy metals). Lead (Pb) and sulfuric acid are main component of Battery manufacturing industry. The chemical composition of effluent was dominated by highly acidic pH (2.1) and Pb (10 mg/l). Other metals (Cd, Cr, Cu) were under the permissible limits (0.036 mg/l, 0.076 mg/l, 0.097 mg/l,) outline by NEQs of industrial and WHO. The study was focused on the Lead and acidic waste water of industry. Once soil/water become contaminated, the heavy metals remain in ecosystem. Since Pb is not biodegradable, it has capability to remain in environment (Pehlivan, 2009).

E. crassipes, *P. stratiotes* and *H. umbellata* were selected for experiment because they efficiently remove heavy metals and other pollutants with high reproduction rate, efficiency and tolerance of ecological factors.

The advantage of employing NaOH solution for adjusting pH of effluent to 7-7.5 is minimum sludge formation during neutralization reaction and product form is water-soluble Na_2SO_4 (sodium sulfate). Macchi et al.(1993) observe that in lead acid battery effluent sodium hydroxide solution precipitate (as hydroxides) resident ferric ions in the battery wastewater and scavenge the lead ions from the wastewater. Based on the minimal mass of moist sludge produced, acceptable residual lead concentration and lowest (among NaOH solutions) TDS of treated acidic waste water is suggested as an alternative agent to remediate the acidic wastewater from lead-acid battery manufacturing industry (Rao et al., 2010).

Pb uptake behavior of plants differ in plants (Fig 4 A, B,C). *E. crassipes* was the most successful candidate for removing Pb from the effluent and least accumulated by *H. umbellata* ($P<0.05$). The *P. stratiotes* showed fluctuation during the time period. At initial phase 0-9days the uptake was slow in *E. crassipes* than it was raised high on the 9th day after wards it was rapid but steady. These results followed Leboudi et al., 2008 experiment in which the duration of slow initial phase of removal for Pb was during the period 2 to 6 days; thereafter, a much rapid rate of removal continued for remaining duration. Zhu et al. (1999) and Vesk et al. (1999) showed similar results of removing metals from industrial effluent treated with *E. crassipes* . Majeed et al., 2014 studies showed high removal of Pb (63.2 g^{-20L}) than Water Lettuce from the effluent of Fuse Shop C-12, Pakistan Ordinance Factory. In *P. stratiotes* fluctuation of heavy metal in roots and shoots was observed from 9th to 21st day of experiment as reported by Vardanyan and Ingole, 2006, high metal concentrations in the roots of *P. stratiotes* . *H. umbellata* also showed fluctuation in absorption from day 0 to day 21.

The physical condition was recorded. *E. crassipes* accumulated most efficiently and steadily until the end of experiment, but necrosis and lack of chlorophyll was observed in most of plant leaves. Water deficiency was also recorded. No new plantlets were produced neither flowering occurred while reverse conditions were observed in the control condition; new plantlets, flowering and fresh green leaves were observed. *Pistia stratiotes* also showed lack of new plantlet production, necrosis and rolling of leaf tips. *H. umbellata* exhibited chlorosis, stunted growth and no production of new plantlets.

The conditions are as described by different scientists that higher concentrations of Pb significantly impact physiological activities of plant including water status and causing water deficit (Patra et al. 2004; Sharma and Dubey, 2005). Plants exposed to Pb ions showed a considerable decrease in the dry weight of different plant parts (Kosobrukhov et al. 2004; Sharma and Dubey, 2005), root elongation and new root development (Odjegba and Fasidi 2004, Sivalingam, 2016), and a decline in the total chlorophyll content , photosynthetic activity decline in the photosynthetic rate (Kambhampati et al. 2005, Tangahu et al, 2011). It has been shown that plants exposed to Pb ions showed a inhibited activities of Calvin cycle enzymes, as well as a deficiency of Carbon mono oxide due to stomatal closing (Sharma and Dubey, 2005). Mane et al. (2011) indicated that at lower concentrations (5 mg/L) of heavy metals, the plant growth was normal and removal efficiency was greater. Concentrations greater than 10 mg/L, the plant started wilting and removal efficiency was reduced due to toxicity at higher metal concentrations.

Aquatic plants growth gets double and triple within 20 - 30 days and because of fast reproduction, decaying of old plant parts initiate (Vardanyan and Ingole. 2006). At the end of experiment, the plants were deteriorating and it was observed it is best to harvest the plants after 21 days. This evolution indicates that maximum period to allow the plants in the system is 21days, Because plant reproduces rapidly

and decays, the efficacy of the system is intimately linked to its careful management trough periodic harvesting of part of the biomass produced (Gupta et al., 2012). Decomposing of plants occur by consuming oxygen the process release the nutrients absorbed by the plants. (Cooke et al, 2005). Our results follow Fonkou et al., (2002) study who indicated that number of leaves per plant decreases, as a result of decay of the basal leaves that fall back into water, then releasing the substances that were absorbed after 15 days in all treatment ponds.
According to the table 3 presenting the mean maximum and minimum contents of heavy metals in aquatic plants, higher mean content of Pb was found in *E. crassipes* (1412.9) than *P. stratiotes* and *H. umbellata*. *E. crassipes* is best accumulator of Pb from heavy metal contaminated effluents (Majeed et al., 2014 and Carvalho and Lenzi, 2000). Higher mean contents of Cd were found in *H. umbellata* (103.49) and least in *P. stratiotes* (28.35). The high values of bioconcentration factor of Cd (7173, at 0.2 mg/L) on day 9 of exposure was reported by Panyakhan et al., 2006 indicating *H. umbellata* better candidate for removal of Cd and Zn from contaminated water. Similarly, Cr and Cu was significantly (P> 0.05%) absorbed by *P. stratiotes* (119.41, 28.47) and least absorbed by *H. umbellata* and *E. crassipes* (91.00, 19.068) respectively. Similar results were reported by Majeed et al., 2014, presenting *P. stratiotes* to be hyeraccumulator of Cr and Cu.

5. Conclusion and recommendations

The content of heavy metals Pb, Cr, Cu, and Cd in the phytomass of three aquatic plants growing in effluent coming from battery industry with the target of applying the findings to applications for control of heavy metal discharges in industrial effluents was studied. The main constituents of battery industry effluent include Lead (Pb) and high acidity (2.1-2.3). The highest value of lead (Pb) was observed in *Eichhornia crassipes*, Cr and Cu in *P. stratiotes* and Cd in *H. umbellata*. *E. crassipes* was found to be most successful in this experiment and recommended to be used to remediate battery industry effluent after adjusting the pH to 6-7, as it absorbed high quantities of Pb. The study reveals the potential and effectiveness of aquatic plant especially *E. crassipes* for remediation battery industry effluent. Removal after 21 to 25 day is highly suggested as with the passage of time plants will die and nutrients absorbed will be added to effluent.
Heavy metals uptake, by plants using phytoremediation technology, seems to be a successful way to remediate heavy metals-contaminated environment. Several factors must be considered in order to accomplish a high performance of remediation result. It can be recommended that

- Physicochemical properties of the battery industry effluent indicated emission of highly acidic water containing loads of heavy metal specifically Lead (Pb).
- Industrial effluents should be treated before discharge from industry to reduce perilous impacts of acidity and heavy metals to surrounding biota environment.
- The most important factor is to select appropriate plant species which can be used to uptake the excessive contaminant. Phytoremediation potential of three aquatic macrophyte species (*P. stratiotes*, *E. crassipes*, and *H. umbellata*) cited in this study, *Eichhornia crassipes* was found most effective due to data achieved and available, research studies, fast growth, ability of biosorption and resistance to contaminants.
- Government should rigorously enforce environmental laws to avoid water pollution.

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