Phytoremediation of heavy metals from industrial wastewater by using aquatic macrophytes

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2018

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TO MY

PARENTS

CERTIFICATES

This is to certify that the thesis titled "**Phytoremediation of heavy metals from industrial wastewater by using aquatic macrophytes**" submitted by Ms. Sangeeta Anand is an original work and has not been previously submitted in part(s) or full for the award of any other degree to this any other university.

The thesis submitted to Babasaheb Bhimrao Ambedkar University, Lucknow satisfies all the requirements as stipulated in the Doctor of Philosophy (Ph.D.) regulations-1999 as amended in 2008/2010 and it is fit for submission and evaluation for the award of the Doctor of Philosophy of the University.

Supervisor

Head of the Department

DECLARATION

I hereby declare that the thesis entitled **“Phytoremediation of heavy metals from industrial wastewater by using aquatic macrophytes”** is my own work conducted under the supervision of Dr. Narendra Kumar in the department of Environmental Science at Babasaheb Bhimrao Ambedkar University, Vidya Vihar, Raebareli Road, Lucknow and also approved by Departmental Research Committee (DRC).

I further declare to my best Knowledge, the thesis does not contain any part of work, which has been earlier submitted for the award of any other degree either in this University or in any other University/Deemed University.

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PREFACE

Water is precious natural resource that supports life on earth. Unfortunately, it has been continuously subjected to over-exploitation due to ever increasing industrialization. Various water bodies which serve as an essential source of water supplies to public are getting contaminated due to discharge of industrial wastewater contaminated with heavy metals and other inorganic pollutants. The minimization of the health and environmental impacts of the heavy metals in water needed economically viable and effective technologies in order to supply safe water for drinking and agriculture and preserves precious natural resources and biological lives.

Phytoremediation is an innovative technique to handle the water pollution problem with the help of plants because plants are solar-driven, their relative growth rate is high and they are also natural accumulators of elements thus they are cost effective, less destructive, and environmentally friendly method for wastewater treatment. Though, it is a cheaper method, there yet is an urgent need of expert project designers with field experiences who choose the proper species and cultivators for heavy metal removal.

Regular monitoring of industrial wastewater discharge, plays an important role in minimizing the effect of water pollution. This study shows the phytotoxic effect of industrial effluents on seed germination. As well as results of this study indicates that the application of aquatic macrophytes i.e. *Eichhornia crassipes, Pistia stratiotes* and *Lemna gibba* have excellent efficiency to accumulate higher metals concnetration from industrial wastewater.

Sangeeta Anand

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LIST OF ABBREVIATION

ANOVA	Analysis of Variance
cm	Centimeter
ºC	Degree Centigrade
DW	Dry Weight
%	Percentage
mg g^{-1}	Milligram per gram
µg g^{-1}	Microgram per gram
$mgkg^{-1}$	Milligram per kilogram
µ mol/dm^3	Micromole per meter cube
DO	Dissolved oxygen
Cu	Copper
Se	Selenium
Zn	Zinc
Hg	Mercury
Pb	Lead
Mn	Manganese
BOD	Biological oxygen demand
COD	Chemical oxygen demand
TDS	Total dissolved solid
TSS	Total suspended solid
GI	Germination index
PI	Phytotoxicity index
SVI	Seedling vigor index

RE	Removal efficiency
TF	Translocation factor
ECR	Enrichment coefficient (root)
ECS	Enrichment coefficient (Shoot)
ANOVA	Analysis of variance
NTU	Nephelometric Turbidity Unit
Fig.	Figure
OD	Optical Density
O&G	Oil and Grease

Chapter 1
Introduction

INTRODUCTION

Water is a valuable natural resource for agriculture, human being and sustaining life on planet earth. Unfortunately, during the past few decades over exploitation of water by various human activities such as rapid industrialization, urbanization and unplanned agricultural practices added an enormous amount of contaminant loads in water (CPCB, 2008; Giripunje et al., 2015). A large number of industries including textile, tannery, paper and pulp, battery, printing, electroplating, iron–steel, pesticide, paint and pharmaceutical etc. consume large volumes of water and chemicals whose composition and toxicity differs, releases as wastewater to various water systems which is leading concern, specifically for developing countries like India, where the lack of technical knowledge, weak implementation of environmental strategies, and limited fund has given rise to serious trials. The discharged wastewater contaminants are directly related to nature of industries. However, this wastewater has serious negative impact not only on land, groundwater, and surface water bodies, but also on the aquatic and the terrestrial ecological system (Zinabu and Zerihun, 2002). Truong and Smeal (2003) explained that industrial wastewater contains high level organic (hydrocarbons, phenol, biphenyl, protein, carbohydrate, oil, grease, etc.) and inorganic (dissolve solids, nutrient like nitrate, phosphate and heavy metals) pollutants. In the process of decomposition of organic pollutant, dissolved oxygen (DO) of water consumed at a higher level by microorganism, which causing oxygen depletion and several consequences to aquatic biota (Rashed, 2010).

Among these water pollutants, heavy metals are of major concern because of their persistent and bio-accumulative nature (Lokeshwari and Chandrappa, 2007; Chang et al., 2009; Yadav et al., 2009; Singh et al., 2012a). The term heavy metal refers to a metallic constituent that relative high density is between 5.306 to 22.00 g/cm^2 (Hawkes

1997; Sood et al., 2012) and toxic even very low concentration (Lenntech water treatment and air purification, 2004). Swaminathan (2003) reported that since the beginning of the industrial revolution, environmental pollution with toxic heavy metal pollution increase dramatically. They are generally classified into 3 categories- 1) -Essential metals which are required for biochemical and physiological functions, for example- In formation of blood, teeth, bones and other tissues, osmoregulation of body fluids, participate as enzymatic catalytic in biological mechanism, etc. 2) – Non-essential these not required for any function and 3)- Trace metals are useful in the growth and function of the brain, homeostasis of the lungs etc.

During the last decade, the toxicity of heavy metals becomes specific concern, as its concentration in cultivated plants, increases continuously thus risk induced by the consumption of contaminated food because plants represent the main route of heavy metal entry into the food-chain, presenting a danger to human health (Tatar et al., 1999). In plants, metal toxicity affects various physiological processes such as photosynthesis, nutrient uptake, cell elongation, nitrogen metabolism, water balance and transpiration (Demchenko et al., 2005; Alam et al., 2007; Chen et al., 2009a; Amari et al., 2014). Additional metals have an injurious effect on membrane function and inhibit enzymes activities at biochemical level (Gajewska et al., 2009). Romero-Puertas et al., (2007) and Yan et al., (2015) provide evidence that metal toxicity is associated with oxidative stress as imitated by the increase concentration of hydroxyl radicals, superoxide anions, nitric oxide and hydrogen peroxide. All of these alter physiological processes concluding finally in reduced crop yield and quality (Gajewska et al., 2006). Metals decrease root elongation, vessel diameter, tip damage, root hair are collapse, decrease lateral root formation, enhance lignification, alter the structure of endodermis and hypodermis also affect shoot growth and height (Shah et al., 2010). Metal directly transport to the shoot and impact on cellular metabolism. Morphology of the leave is also change due to metal toxicity. Drying of the leaves, necrosis and chlorosis are the main symptoms of metals toxicity in leaves. Saha et al., (2016) reported that metals toxicity also causing burning of leaf tips, slowing, and leaf growth basically young leaf in plants.

Heavy metals generally enter in human body may be food chains, ingestion and inhalation. Though, when these metals enter into the body it stimulates the immune system and may cause nausea, anorexia, vomiting, gastrointestinal abnormalities, and dermatitis (Tchounwou et al., 2012). It also damages the mental and central nervous systems, kidney, lungs, livers, other important organs and change blood composition (Guilarte et al., 2008; Rathnayake et al., 2013; Farhan and Khadom, 2015). The long-term exposures of heavy metals have also shown physical, muscular, and neurological impairments. The deteriorating processes heavy metals are similar to Alzheimer's disease, Parkinson's disease, muscular dystrophy and multiple sclerosis (Guilarte et al., 2008). Another disease such as lung cancer and damage to human's respiratory systems has also been found to develop following high rate exposure to metals (Pandey and Madhuri, 2014). Apart from the toxic effect, some metals, i.e., Cu, Se and Zn, found important and beneficial in human metabolism for example, Cu acts as co-factors for various enzymes of redox cycling at low concentration (Sedlak and Hoigne, 1993; Rathnayake et al., 2013). However, at higher concentration disrupts the human metabolism leading to anaemia, liver and kidney damage, stomach and intestinal irritation.

Due to high solubility of metals in the aquatic environments, they can be easily absorbed by living organisms and after entering into food chain, large concentrations of may accumulate in the human body. If the metals are consumed beyond the acceptable concentration, they can cause serious health disorders (Babel and Kurniawan, 2004; Sood et al., 2012).

The problem of heavy metal pollution needs continuous monitoring and investigation. These metals do not degrade and have a tendency to biomagnified in human through food chain; hence there is an essential to remove heavy metals from contaminated sites. Therefore, many researchers have focused on specific control methods to remove heavy metals from different kinds of waste water system (Adhoum et al., 2004; Heidmann and Calmano, 2008; Kabdaslı et al., 2012; Ahmad et al., 2016a). Various Artificial methods include chemical precipitation, ion-exchange, adsorption, membrane filtration; electrochemical treatment, are used to clean heavy metal pollutants (Fenglian and Wang, 2011; Radjenovic and Sedlak, 2015; Ahmad et al., 2016a). Each of the methods has specific benefits and limitations but in general none of them is cost-effective and eco-friendly and also depend on power source and skilled personnel (Rai, 2009; Rahman and Hasegawa, 2011; Bokhari et al., 2016).

Phytoremediation is a direct used of macrophytes for in-situ and ex-situ removal of heavy metals. It is found very effective for heavy metal treatment with shallow and low level of contamination (Rezania et al., 2016). Process causes plants to remove, detoxify, or immobilize heavy metals in water, through biological, chemical, or physical activities with plants (Rahman and Hasegawa, 2011; Ali et al., 2013). Macrophytes have some unique organisms with extensive metabolic and absorption capabilities as well as transport systems that can take up heavy metals from wastewater (Rai, 2008; 2009; Dixit and Dhote, 2010; Fazal et al., 2015). The process removed heavy metals from water through binding or degradation and detoxification and then macrophytes can be subsequently harvested, processed, and disposed.

Phytoremediation is emerging technique and against to drawbacks of artificial methods, developed in Queensland, Australia by the Department of Natural Resources and Mines (Koelmel et al., 2015). Seidal, (1976) and Wolverton and McDonald, (1976) experimentally proved the importance of macrophytes in removing contaminants from water bodies. Many studies have been conducted to improve the wastewater quality by using phytoremediation process problem (Arora et al., 2004; Chen et al., 2009b; Jafari and Akhavan, 2011; Sasmaz et al., 2015; Rezania et al., 2016). It is a green and environmental friendly wastewater treatment technology with natural recycling, as well as an innovative tool for removing heavy metals from water. Macrophytes are powered by solar energy and thus make this technology cost-effectively, with great potential for achieving environmental sustainable.

Several classification schemes were found relating to the types of phytoremediation, the most common of which are presented below.

1. Rhizofiltration is the technique of utilizing plant roots to absorb toxic metals from contaminated ground water or polluted effluents. It was found that it is potential technique for removal of wide range of organic and inorganic contaminants. Rhizofiltration reduces the movability of contaminant and prevents movement to the ground water thus reduces the bioavailability for entry into the food chain. So, in condensed form we can state this technique of utilizing the potential plants in uptake the pollutants from the contaminated site. It presents great opportunity of using suitable plant species to clean up the environment (Rawat et al., 2012).

2. Phytoextraction is the uptake of contaminants from water and soil by plant roots and their translocation to and accumulation in above ground biomass i.e., shoots (Sekara et al., 2005; Yoon et al., 2006; Rafati et al., 2011). Metals translocation to shoots is a crucial biochemical process and is desirable in effective phytoextraction because, the harvest of root biomass is generally not feasible (Zacchini et al., 2009; Tangahu et al., 2011; Ali et al., 2013).

3. Phytostabilization is the use of the certain plants for stabilization of pollutant in soil/water (Singh and Prasad, 2015). This technique is used to reduce the movability and bioavailabilty of pollutant in environment, thus preventing their movement to ground. Plant can immobilize heavy metals in soil through sorption by root, precipitation and complexation etc. (Yoon et al., 2006; Wuana and Okieimen, 2011). Phytostabilization limits the accumulation of heavy metals in the plant and minimize their leaching into underground water.

4. Phytovolatilization pollutants were uptake from water by plants and their subsequent conversion to the volatile form and releases into atmosphere. This technique is used for organic pollutant and some heavy metals like Se and Hg. Phytovolatilization is limited by that it does not remove the contaminant completely but it transfers pollutant to one segment to another segment (soil to water). Consequently, this technique is controversial in phytoremediation techniques. (Padmavathiamma and Li, 2007).

5. Phytodegradation is a technique for degradation of organic pollutant by plant in the presence of enzymes like oxygenase and dehalogenase, however, it is not dependent on rhizospheric microorganisms (Vishnoi and Srivastava, 2008).

Plants can accumulate organic pollutants and detoxify them through their metabolic activities. Phytodegradation is limited to removal of organic pollutant only because heavy metals are non- biodegradable. Recently, scientists have been shown their interest in studying Phytodegradation of various organic pollutants including synthetic herbicides and insecticides (Doty et. al., 2007; Ali et al., 2013).

Macrophytes are aquatic plant growing in or near water bodies. The term aquatic macrophytes refer to microscopic form of aquatic vegetation. Arber, (1920) and Sculthorpe, (1967) classified macrophytes as following categories:- 1)- **Emergent plants** which are having roots in soil and the plant growth is rising to considerable heights above the water, (*Typha, Phragmites, Sagittaria, Eleocharis, Polygonum etc.*) 2)- **Submerged plants-** These are grown mostly below the water surface (*Hydrilla, Marcillium, Chara, Nitella etc.*) 3)- **Free floating plants** floating on the surface, (*Pistia, Lemna, Azolla, Salvinia, Trapa, Eichhornia* etc.), 4)-**Floating leaves**- they are found in waterlogged sediment and found at depths of about 0.5–3.0 m (*Nymphea, Potamogeton, Nelumbo, Hydroryza etc.*).

Aquatic macrophytes are more appropriate for treatment of waste water as compare to terrestrial. They have many different physiognomies such as growing rate, huge production, contaminants uptake ability, and better extraction due to direct interaction with wastewater. Macrophytes perform imperative roles at functional and structural levels of aquatic water bodies, such as changes in water movement, shelter to aquatic habitat, and a good food source and at functional level they alter the water quality by balancing oxygen, mineral cycle and heavy metal accumulation (Dhote and Dixit, 2009). Macrophytes have the ability to accumulate heavy metals and this makes them attractive for research (Mishra et al., 2008; Rai and Tripathi, 2009). The potential of macrophytes for phytoremediation is chiefly dependent on plant species and difference storage potential for heavy metal. Some environmental factors should be sustained like chemical species, initial concentration of the metal, interface of different heavy metals, temperature, pH, redox potential, and salinity.

The macrophytes *Lemna gibba, Pistia stratiotes and Eichhornia crassipes* are major part of aquatic ecosystem and show high removal efficiency for Hg, Pb, Zn and Mn. They absorb the contaminants and translocate their roots and shoots from polluted waste water (Zhu et al., 1999; Rai, 2009).

Objective

1- To analyse the physico-chemical characteristics including heavy metals of different industrial wastewater.

2- Evaluation of phytotoxicity of different industrial wastewater through seed germination test on different varieties of crop seeds.

3- To study the partitioning of metals within the different aquatic weeds exposed to different industrial wastewater.

4- Comparative study on the phytoremediation potential of various aquatic weeds to remediates heavy metals from different wastewater.

5- Identification and application of aquatic weeds for the removal of heavy metals from different industrial wastewater.

Chapter 2

Review of literature

REVIEW OF LITERATURE

Water is a most important natural resource available on earth. All living biota requires water for their survival hence considered as a most essential natural resource. It covers about 71% of total earth's surface, but only 2.5% of water is available as fresh water. World population is increasing day by day, causes pressure on water security. Further, the demand and supply of water for human needs is increasing, causing a great pressure on available limited sources of freshwater (Wichelns et al., 2015; Rashidi et al., 2015). Quality of water is affected due to rapid industrial proliferation, urbanization, and releases huge amount of wastewater contaminated with various inorganic and organic pollutants which substantially changes the water quality of natural water bodies (Marshall et al., 2007; Rajasulochana and Preethy, 2016; Victor et al., 2016b).

Organic and inorganic contaminants such as heavy metals, nitrate, phosphorous, suspended solids, may be present in water which commonly discharged from residential and industrial sources. These pollutants cause adverse effects on water and soil quality which directly or indirectly affect animal and plant nutrition, as well as human health (Jomova et al., 2011; Chon et al., 2012). Heavy metals are inorganic pollutants, with metallic properties i.e. conductivity, ductility, stability as cations, ligand specificity etc. and their atomic number >20, which contaminate water, soil and air (Ali et al., 2013).

These metals are leading contamination for environment because these are not naturally biodegradable and can be transferred through trophic levels and accumulate in the biota insistently (Gall et al., 2015; Nancharaiah et al., 2016). They present at different valance states, so one element is less or more toxic in different valence states. Normally heavy metals occur in low concentration but due to human activities (mining, smelting, fuel combustion, agrochemical applications, and waste disposal etc.) increased its concentration in ecosystem (Prasad, 2011).

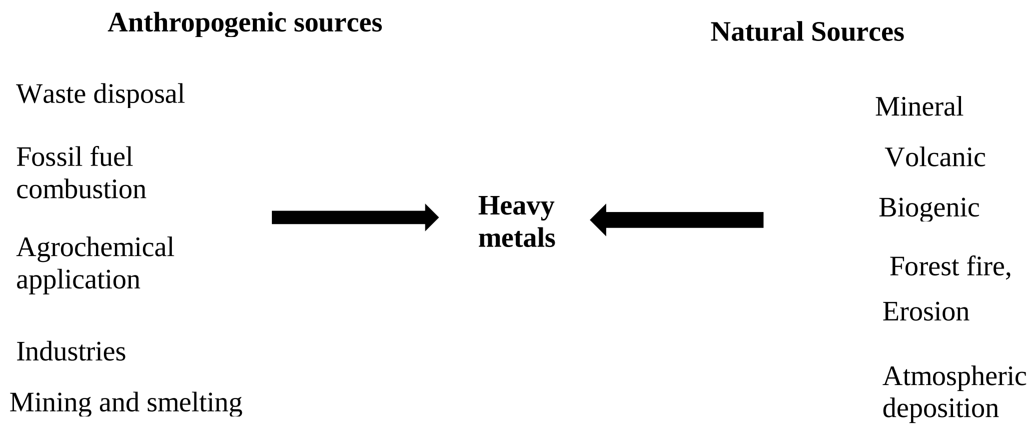

Fig. 2.1: Anthropogenic and natural sources of heavy metals (Nagajyoti et al., 2010; Yadav, 2010; Prasad, 2011; Chibuike and Obiora, 2014; Oves et al., 2016).

Some metals are essential at low concentration for healthy function of biota like Mn, Zn, Cr, Mo, Fe, Ni etc. but toxic at higher concentration and some are non-essential and extremely toxic even very low concentration like Pb, Hg, Cd etc. (Nagajyoti et al., 2010; Prasad, 2011; Chibuike and Obiora, 2014; Rezania et al., 2016). Heavy metals are easily absorbed in living cells. Heavy metal such as Cd, Pb, Zn, Hg, Mn, Cu, Cr, Ni and Fe are hazardous among the chemical concentrated industries. They are entering into food chain and if they are beyond limits they accumulates in human bodies and cause serious health hazards (Babel and Kurniawan, 2004; Barakat, 2011; Sood et al., 2012).

Table 2.1: Heavy metal and their impact on human and plant health

Metals	Impact on human health	Impact on plants	References
Pb	Osteoporosis, inhibits formation of hemoglobin, loss of IQ (intelligence quotient), high blood pressure, anemia, gastrointestinal effects	Inhibits activities of enzymes, Chlorosis, decrease photosynthesis and transpiration rate, imbalance of minerals.	Sharma and Dubey, (2005); Singh et al., (2011); Singh and Kalamdhad, (2011)
Mn	Nerve damage, hallucination, lung embolism, bronchitis, headache, dullness, insomnia, paralysis, emotional disturbance, laziness etc.	Reduction of photosynthesis, necrosis, retarded germination, slow growth rate, Leaf crinkling	Millaleo et al., (2010); Zhao et al., (2012)
Zn	Vomiting, fever, nausea, stomach cramp, diarrhoea, coeliac disease, sickle cell animea etc.	Necrosis on leaves, reduce growth of root, decline lipid peroxidation and respiration	Singh and Kalamdhad, (2011) Gutierrez-Carbonell et al., (2013);
Hg	Damage brain, neurological and renal disturbances, tremor, memory problem, lung damage	Blocking of important enzymes, distribution in cell membrane, reducing seed feasibility, reduce photosynthesis, transpiration rate and chlorophyll synthesis	Mukherjee and Fryar, (2008); Martin and Griswold, (2009)
Cu	Fever, dark urine, Diarrhoea, weakness, difficulty breathing	Inhibits growth, interference in photosynthesis and respiration, reduce	Yruela, (2005)

		biomass, affect seed germination.	
Cr	Cuase lung cancer, damage hereditary material of cell, lead cell mutation.	Affect photosynthesis, growth reduction, chlorosis, wilting of plants, root become thin brittle and brown and leave turns pales and frailer under Cr stress	Nriagu and Nieboer, (1988); Shanker et al., (2005); Singh et al., (2013)
Cd	Interfere cell metabolism, lead hepatotoxicity, and reduce weight and growth, and cause anemia, renal changes, less development of testis, difficulties in pregnancy.	Disturb in chloroplast, metabolism, decrease enzymatic and non-enzymatic antioxidants, affect the availability of mineral from soil, chlorosis, leaf curl and stunting, dercease N_2 fixation	Benavidas et al., (2005); Swarup et al., (2006); Reis et al., (2010)
Ni	Produce headache, nausea, respiratory disorder, damage reproductive health, mimic the action of estrogen	Disturb cell membrane function, decrease chlorophyll content, reduce plant nutrients, decrease growth affect Calvin cycle and CO2 fixation.	Sreekanth et al., (2013); Asati et al., (2016)
Fe	Reduce growth, diarrhoea, anorexia, reduce feed intake, metabolic acidosis and death, vomiting, mild abdormal pain	Damage membrane, protein, lipid and DNA	Gupta and Gupta, (1998); Asati et al., (2016)

Effect of various heavy metals on seed germination is carefully assessed because this provide meaningful behaviour of seed species in metals contaminated environment and provide suitability of species for future survival (Singh, 2005). Seed germination is defined as sequential morphological process which results transformation of embryo into a seedling (Penfield, 2017). Koeppe (1977) studied on aspect of germination, enzyme activities and respiration in plants and found that Cd as an inhibitor which effect the enzyme activities and ATP synthesis. Singh et al. (1982) examined the germination in wheat seed and found decrease growth in radicles.

These growing problems of heavy metal contamination have significant negative effect on human livelihoods, economic development, and environmental quality throughout the world. Hence it has become an essential need for today's environment to protect water from getting polluted or to develop cost effective remedial method for its protection. The researchers tried to develop effective techniques due to main issue of heavy metals existence in wastewaters and various approaches have been done for the enhancement of cheaper and more effective technologies (Adhoum et al., 2004; Dhir et al., 2009; Heidmann and Calmano, 2008; Ahmad et al., 2016a). Two types of control methods being used for remove heavy metal, 1) artificial or conventional methods include chemical precipitation, ion-exchange, adsorption, membrane filtration; electrochemical treatment technologies, etc. and 2) natural methods and non-conventional i.e. biosorption, bioremediation, phytoremediation (Singh et al., 2012a).

Table 2.2: Artificial or conventional method, its mechanism and disadvantages for removal to heavy metals from waste water

Techniques	Mechanism	Advantages	Disadvantages	References
Chemical Precipitation	Chemicals like lime, pyrite, tri-mercapto-triazine, potassium and sodium thiosulphates, potassium ethyl xanthate etc. reacts with heavy metal ions and form insoluble precipitates.	Simple and Cheap	Produce large sludge containing metals, High maintenance cost due to sludge disposal, Start-up and shutdown time is longer, Toxic gases generation, Cr is not removing by this process.	Fenglian and Wang, (2011); Ahmad et al., (2016b)
Ion exchange	Ion exchange resin exchange their cation with metal ions in waste water. Commonly used ion exchanger is sulfonic acid group (-SO_3H) and carboxylic acid group (-COOH) in which metal ions exchange with H^+ ions with following process- $R\text{-}SO_3H + M^+ \longrightarrow (R\text{-}SO_3)\,M + H^+$ $R\text{-}COOH + M^+ \longrightarrow (R\text{-}COO)\,M + H^+$	Effective, Pure effluent metals recovery are possible	High operational cost, Ion exchanger quickly reduce its exchange capacity	Parmar and Thakur, (2013); Ahmad et al., (2016a)
Adsorption	Absorbent such as agricultural waste, clay, saw dust, peat, gram husk; activated carbon, zeolites, fly ash, lignin etc. absorbed heavy metal from aqueous solution.		Relatively high cost, Adsorbent progressively deteriorate its capacity, its performance depends on adsorbent types	Kurniawan et al., (2006); Fenglian and Wang, (2011); Ahmad et al., (2016b)

Membrane filtration	Membrane pore size are smaller than size of metals where metals trapped. Removal efficiency depends on metal ion concentration, membrane characteristics, pH and ionic strength of solution.	Microfiltration and ultrafiltration highly efficient which fracturing particle according to size, No phase change involve, function at low temperature	Limited flow rates, Complex process, Higher maintenance and operational cost	Qin et al., (2007); Sampera et al., (2009); Zhao et al., (2016)
Reverse osmosis	Process uses semi-permeable membrane, allowing fluid to pass through it and purified, while rejecting the contaminants.	No harmful Chemicals production,	Membranes sensitive to hard water, Ineffective in removing As^{3+}, Power consumption	Fenglian and Wang, (2011); Zhao et al., (2016); Ahmad et al., (2016a)
Electrodialysis	Separate heavy metals through charged membranes from solution using an electric field as the driving force.	Low energy consumption	Complex process, High cost, Low permeable flux	Mohammadi et al., (2005); Barakat, (2011); Ahmad et al., (2016a)

Coagulation and flocculation	Coagulants such as aluminium, ferrous sulfate and ferric chloride, are used in the wastewater treatment by charge neutralization of particles and by enmeshment of the contaminants which formed amorphous precipitates. Flocculation used polymers in solid-liquid separation process for removal of suspended and dissolved solid, organic matter and metals from water. flocculants, all dispersed particles are aggregated together and form floac (large particles) purified water	Simple and cost effective, enhance filtration process, separate many kings of pollutants from water	Large volume of sludge produces, High chemical consumption, time consuming	Sharma et al., (2006); El-Samrani, et al., (2008); Fenglian and Wang, (2011); Ahmad et al., (2016a)
Electrochemical treatment	Several technologies are involved electrocoagulation, electro- flotation, and electrodeposition etc. Electrocoagulation contains the generation of coagulants by dissolving electrically with using electrodes. The metal ion generates at the anode, and hydrogen gas out from the cathode which can help to float the flocculated particles on water).	Fast treatment, remove less colloidal particle, less maintenance	High chemical and energy consumption, Huge sludge production, Poor settling	Chen et al., (2004); Aziz et al., (2004); Sires, et al., (2014); Radjenovic and Sedlak, (2015); Gunatilake et al., (2015)
Flotation	Dissolved air flotation (DAF) permit air bubbles attach to the suspended particles, developing lower density agglomerate, causing flocs to rise through the water and collecting in the form of sludge at the surface		High cost, increasing ion strength decrease floatation efficiency	Lundh et al., (2000); Fenglian and Wang, (2011); Ahmad et al., (2016a)

Although these artificial techniques are effective for remediation purpose, but their operational and maintenance cost is very high and their by-product are highly toxic in nature. Hence, restoration of polluted aquatic ecosystem needs eco-friendly, cost effective and natural remediation technologies.

2. Natural or Non-conventional methods

2.1 Biosorption

Biosorption is natural method, which utilizes various natural materials of biological origin, including bacteria, fungi, yeast, algae, barks, dry leaves, fruits etc. These biosorbents have metals sequestering properties and effectively sequester dissolved metal ions from complex solutions with high efficiency and quickly. (Veglio and Beolchini, 1997; Chen et al., 2014; Salman et al., 2015; Pokethitiyook and Poolpak, 2016). *Saccharomyces cerevisiae* was the first eukaryote to have its complete genome sequenced and this will undoubtedly lead to a new application (Kapoor and Viraraghavan, 1995). Nasir et al. (2007) studied that the removal of lead and zinc from aqueous solutions using chemically modified distillation sludge of *Rosa centifolia* (rose) petals. Maximum adsorption of both metal ions was observed at pH 5. Arshad et al. (2008) utilized leaves and stem bark of the *Azadirachta indica* (neem) tree for removing zinc from water in different pH. They explained that the uptake of Zn was initially fast, but progressively slowed down which indicating penetration inside the adsorbent. The maximum adsorption capacity for zinc is 137.57 mgg^{-1} for bark and 147.19 mgg^{-1} for leaves. They also explained that pH, particle size of adsorbent, dose of adsorbent, initial concentration of metal and time duration highly affect process. Other benefits include low cost with high efficiency, and the reduction of chemical or biological sludge, no any additional food requirement, and the possibility of renewing biosorbent and extraction of metals (Rajoriya and Kaur, 2014).

2.2 Bioremediation

It is also natural process which use of living microorganisms to degrade environmental pollutants into less toxic forms (Ma et al., 2015). It includes bacteria and fungi to detoxify hazardous substances to human health as well as for environment. Bioremediation involved those microorganisms are capable to degrade pollutant from polluted site (Yu et al., 2011; Zeraatkar et al., 2016; Tiwary and Dubey, 2016; El-Gendy and El-bondkly, 2016; Kang et al., 2016). Heavy metals are transformed by microorganisms through their metabolic processes. It can be effective only where environmental conditions permit microbial growth activity and its application involves the manipulation of ecological parameters to permit microbial growth and degradation to precede faster rate (Mani and Kumar, 2004; Dixit et al., 2015; Ghosh et al., 2015). This technique is more economical and effective compare to artificial methods, contaminants can be treated on site and reducing exposure risks and transportation accidents (Zeraatkar et al., 2016).

2.3 Phytoremediation

Phytoremediation term originate from Greek word “Phyto” means plants and latin word “remedium” means removal or restoration balance, consist of mitigating heavy metals from water with the help of living plants that can have ability to accumulate metals (Prasad and Freitas, 2003; Sasmaz et al., 2008; Kalve et al., 2011; Dixit et al., 2011; Sarma, 2011; Singh and Prasad, 2011; Singh et al., 2012b; Vithanage et al., 2012; Sood et al., 2012). It is an energy-efficient, cost efficient and aesthetically pleasing method for moderate to low levels concentration of metals (Rupassara et al., 2002; Banjoko and Eslamian, 2016). The potential for this technology is high due to the prevailing climatic conditions which favours plant growth and stimulates microbial activity (Zhang et al., 2010b). In natural ecosystems, plants act as sieves for metabolize substances produced by nature (Rahman et al., 2016).

Plants has specific role in phytoremediation and could be different in metal uptake mechanism. Basically, heavy metals accumulate, translocate and concentrate in the green parts of plants (Bhargava et al., 2012). As Nyquist and Greger (2007) indicated uptake mechanism of aquatic plant i.e. depends on the type of the plant and level of wastewater occurs by direct absorption from the water to plant. This process was followed by passive or active transport of metals across membranes and by roots. Jha et al. (2010) explained their mechanism especially in submerged species as well as free floating plants. The growth rate of plant and the metal concentration in tissues have directly specially effects on removing capacity of plants (Rahman, and Hasegawa, 2011). Accumulation firstly occurred in roots from where they translocate into whole body of plants (Giripunje et al., 2015). Vymazal (2016) showed that the significance of aquatic plants with high shoot biomass have high ability to accumulate heavy metals. As expected, the heavy metals concentrations usually in root is much higher than in shoot of biomass. Indeed, this limitation of metals in the roots acted as an initial protection of the photosynthetic apparatus in their aerial parts (Drazkiewicz and Baszynski, 2005).

Table 2.3: Average annual percent increase from 1999 to 2011 in articles and patents containing the given processes (Koelmel et al., 2015).

Processes	Patent	Articles
Phytoremediation	12%	24%
Bioremediation	4%	12%
Remediation (with conventional methods)	6%	12%
Constructed wetland	14%	16%

Using plants for metal removal has attracted more attention in last two decades (Jha et al., 2010). When number of articles containing phytoremediation as a percentage of total articles per year calculated then found that the articles with phytoremediation is increasing at a linear rate in last 16 years (Figure 1).

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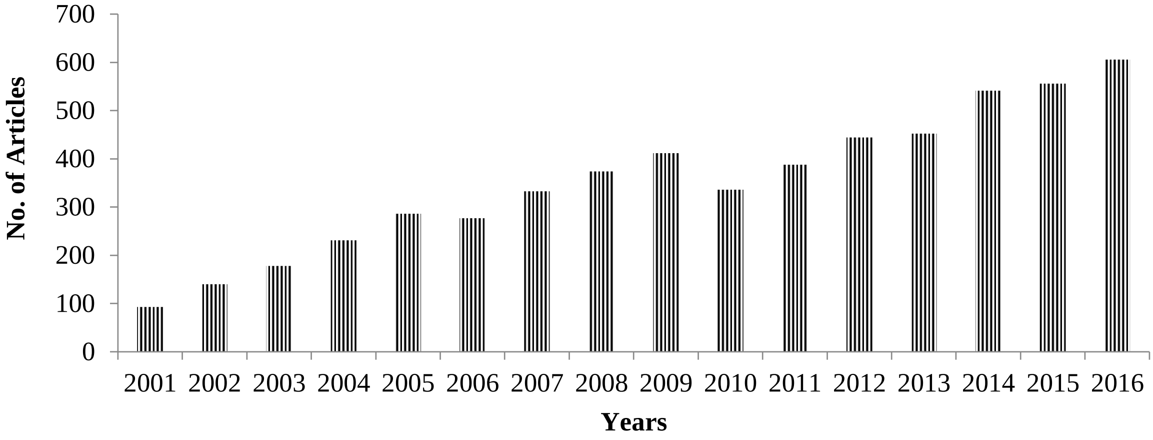


Fig. 2.2: Publications in field of phytoremediation from last 16 years (Sciencedirect.com.)

Table 2.4: Historical advances in era of phytoremediation using macrophytes

Decades	Summary	Scope of works	Highlight/focal point	References
70s	• Due to massive capacity of nutrient uptake from water, macrophytes used for remediation. • Submerged and floating quickly uptake pollutant from water • Levels of toxic elements in the plants were at least an order of amount higher than in water.	Basically, research on remediation potential of aquatic plants specially submerged plants	Potential of uptake	Boyd, (1970); Cowgill, (1974); Hutchinson, (1975)
80s	• Floating and Emergent plants can uptake contaminants through roots while submerged plants by leaves both.	Study on emergent and floating plants capability	Species determination	Denny, (1980,1987)
90s	• Rates of toxic metals uptake and removal of plants is greater than 1000µgg^{-1} is called as hyperaccumulator.	Research on metal accumulation through root and foliar parts of aquatic plants.	Importance of various mechanisms	Outridge and Noller, (1991); Sharma and Gaur, (1995)
00-10	• Aquatic plants can efficiently remove heavy metals which is	Monitoring the effective role of	Effectiveness of species	Hu et al., (2003), Kamal et al.,

	largest categories of pollutants. • Aquatic macrophytes also used as non-living, for removal and monitoring of heavy metals.	macrophytes and Developing of hyper-accumulator plants.		(2004); Rai, (2009)
10-16	• Uptake mechanisms of green plants which can accumulate pollutants with high ability. • Efficacy of phytoremediation process (low cost, low energy consumption) in contrast with the conventional methods and no special care is required. • Chemical like chelating agents are used to enhance the remediation potential of hyperaccumulating plants.	Focus on mechanism and improvement of techniques efficiency	Optimization towards Implication	Sharma et al., (2012); Ali et al., (2013b); Samsaz et al., (2015)

Various techniques were used as phytoremediation process which are phytoextraction, phytodegradation, phytostabilization, phytovolatilization and rhizofiltration (Rahman and Hasegawa, 2011). As specified by Thakur et al. (2016) and according to Table 2.5, rhizofilteration are commercially important and specific technique for wastewater remediation among other techniques.

2.1. Rhizofiltration

Rhizofiltration is defined as using root system of aquatic macrophytes for removal of heavy metals (Favas et al., 2014). Various studies were accepted the high ability of rhizofiltration of heavy metals from the aqueous streams by aquatic macrophytes (Zhang et al., 2010a; Rawat et al., 2012). Rhizofiltration also konow as blastofiltration (use of seedlings) and caulofiltration (caulis are Latin word means shoot, use of excised plant shoots) (Mesjasz-Przybylowicz et al., 2004). Which plants have dense root system and grown hydroponically have maximum removal efficient of heavy metals by this process (Xie et al., 2009; Roy et al., 2015). The efficiency process highly depends on the physicochemical characteristics of the plants or photosynthetic surface microorganisms (Olguin and Sanchez-Galvan, 2012). Absorption through plants, roots play vital role in this system, so large root surface areas are usually required. Rhizofiltration reduces the movability of contaminant and prevents movement to the water thus reduces the bioavailability and entry into the food chain. It offers great opportunity of using suitable aquatic plant species to clean up the water bodies (Rawat et al., 2012). Abhilash et al. (2009) stated that rhizofiltration process not interfered plant growth rate in heavy metal removal, while it is not proper method for onsite treatment of plants with small root and slow growth rate. They also described that an ideal plant should have great production rate with high tolerance potential to metal accumulation and also should harvestable in feasible ways. Pratas et al. (2014) reported that some environmental factors impact on growth of pants i.e. temperature, nutrient balance and reproduction maintenance in laboratory are limitation of rhizofiltration. Elless et al. (2003) found some advantages of rhizofiltration methods in arsenic removal such as specificity of biological mechanisms, efficient uptake of arsenic and arsenite, lower cost of operation, free disposal waste and performable in low light condition. Hence, the operational design of treatment system can be influenced by some on site condition like changes in water flow rate which can be solved in an economical way.

Table 2.5: Techniques of phytoremediation

Techniques	Application	Contaminants	Accumulation	Description	References
Rhizofiltration	Water	Inorganic/organic	Root	Pollutants absorbed from contaminated waters by aquatic plants	Rahman and Hasegawa, (2011); Rawat et al., (2012); Ali et al., (2013a); Rezania et al., (2016)
Phytoextraction	Soil/water	Inorganic/organic	Shoots	Accumulation of pollutants in by root and translocate them to upper parts	Tangahu et al., (2011); Ali et al., (2013a); Oosten and Maggio, (2015); Sharma et al., (2015); Thakur et al., (2016)
Phytovolatilization	Soil/water	Organic	Release into atmosphere	Volatilisation occurs in leaves and pollutants convert to volatile form	Padmavathiamma and Li, (2007); Oosten and Maggio, (2015); Sharma et al., (2015); Thakur et al., (2016),
Phytostabilization	Soil/water	Inorganic/organic	Rhizoshpere	Plant can immobilize heavy metals in soil through sorption by root, precipitation, complexation.	Wuana and Okieimen, (2011), Ali et al., (2013a); Oosten and Maggio (2015); Sharma et al., (2015); Thakur et al., (2016)

Phytodegradation	Soil/water	Inorganic/organic	Within the plant tissues	Degradation of organic pollutant by plant in the presence of enzyme like oxygenase and dehalogenase it is not dependent on rhizospheric microorganisms.	Oosten and Maggio, (2015); Sharma et al., (2015); Cacadore and Durate, (2015)
Phytodesalination	Soil	organic	Within the plant tissues	Removal of salts from soils by conversion	Mackova et al., (2007)
Rhizodegradation	Soil	Inorganic/organic	Rhizoshpere	Degradation of organic by Rhizospheric microorganisms	Zorrig et al., (2012); Bano et al., (2013); Li et al., (2016); Jia et al., (2016)

Mechanism of phytoremediation

Phytoremediation process is an ability of plants to bioaccumulate, degrade, or reduce contaminants from soils or water. Macrophytes possess specific mechanism to accumulate and remove heavy metals. Four processes are generally involved 1) Uptake of heavy metal, 2) translocation and complexation with chelating agents, 3) Detoxification and 4) compartmentalization (McGrath and Zhao, 2003; Bhargava et al., 2012; Revathi and Venugopal, 2013).

Uptake of metal ions

The uptake of heavy metals into roots occurs either by passive diffusion through the cell membrane or by active transport through electrochemical potential gradients mediated by carriers. These carriers can be chelating agents, such as organic acids or proteins that bind to the metal ions species (Fergusson, 1990; Hossain et al., 2012). There are two pathways available for metal ions to enter into the roots: apoplastic and symplastic. The apoplastic movement of the metal ions is possible only as non-cationic metal chelates because cell walls have comparatively high exchange capacity for cations (Raskin et al., 1997; Chojnackua et al., 2005). For example, Krems et al. (2013) showed that *Elodea canadensis* binds in its cell membranes approximately 70% Cd from the solution with concentration of 0.6 µmol/dm^3, 30% Zn from the solution with concentration of 30 µmol/dm^3 and 50% Cu from the solution with a concentration of 1.0 µmol/dm^3. It was confirmed that most of the accumulated zinc and copper is transported through a cell membrane inside a cell, whereas cadmium remains in the cell membrane of the plant. Thus, as the most of the metal ions are insoluble and unable to move on their own in vascular system, they are immobilized in apoplastic and symplastic compartments after forming carbonate, sulphate, or phosphate precipitates (Raskin et al., 1997; Garbisu and Alkorta, 2001; Sheoran and Sheoran, 2006).

Efficiency of phytoremediation lays in the efficiency of roots to synthesis certain chemicals which cause heavy metals to rise in plant body. Root exudates and changes in pH of rhizosphere, may cause metals to precipitate onto root surfaces. Plant have some avoidance strategy for lessing metals by preventing excess heavy metals entering into root either by complexing or by precipitating. Root environment produce biogeochemical conditions that result in precipitation of contaminants onto the roots or into water.

Translocation and complexation of metal ions

After uptake translocation process occur, in which hyperaccumulator plants translocate a high metal ion into the shoot via symplast through xylem where they are mostly deposited in vacuoles (Tester and Leigh, 2001; Prasad, 2004; Jabeen et al., 2009). Vacuoles are the cellular organelles with low metabolic activities (Denton, 2007). The high negative latent potential of plasma membrane helps in movement of metal ions due to electrochemical gradient (Raskin et al., 1994). Heavy metal sequestration in the vacuole is one of the ways to remove excess metal ions from the cytosol and may reduce their interactions with cellular metabolic processes (Assunção et al., 2003; Sheoran et al., 2011). Specialized protein (ATPase, natural resistance-associated macrophage proteins (Nramps), Zinc ion permease (ZIP), cation diffusion facilitator (CDFs), zinc regulated transporter (ZRTs,), cation antiporters) embedded in membrane system which play important role in translocation process (Guerinot, 2000; Williams et al., 2000; Gaxiola et al., 2002, Rahman and Hasegawa, 2011).

The superfamily of P-type ATPases performs the function of transport of a wide range of cations across the cell membranes. For example, there are 8 heavy metal ATPases, 6 Nramps, and 15 ZIPs present in *Arabidopsis* (Williams et al., 2000; Maiser et al., 2001; Hall and Williams, 2003; Mills et al., 2003). The ATP-binding cassette

(ABC) superfamily is another important and diverse family of transmembrane proteins involved in a wide range of transport functions by utilizing energy from ATP hydrolysis (Rea 1999; Martinoia et al., 2002; Barcelo and Poschenriedes, 2003; Chen et al., 2015). They have a role in Mg-ATPhydrolysis- driven vacuolar sequestration of glutathione S-conjugates (GS-conjugates) (Martinoia et al., 1993; Pinho and Ladeiro, 2012). The natural resistance-associated macrophage proteins (Nramps) is also an important integral membrane protein family involved in transport of metal ions in a wide range of organisms, including both prokaryotes and eukaryotes. They facilitate the uptake of many heavy metal ions (Cu^{2+}, Mn^{2+}, Co^{2+}, Fe^{2+}, and Cd^{2+}) (Chen et al., 2003; Jan and Parray, 2016). The ZIP (ZRT, IRT-like proteins) family of genes is involved in the transport of many cations (e.g., Fe, Mn, and Zn) (Guerinot, 2000). Different subfamilies of ZIP gene superfamily exhibit variations in terms of substrate and specificity (Guerinot, 2000; Mäiser et al., 2001). The ZIP family members have been identified from both prokaryotes and eukaryotes. On the basis of alignment of the predicted amino acid sequences, ZIPs could be grouped into four subfamilies (Gaither and Eide, 2001). However, ZIP higher plant genes could be grouped together (Mäiser et al., 2001). The cloning of various ZIP/NRAMP transporter genes from plant species and other organisms has shown a wide range of metal specificity and sequences (Guerinot 2000; Williams et al., 2000). These investigations of ZIP and NRAMP gene families suggested the existence of consensus regions on the amino acid sequences which could be responsible for the determination of the metal transport. For example, the histidine rich region found between transmembrane domains III and IV of AtIRT1 is a cytoplasmic metal ion-binding site (Zhao and Eide, 1996). Available literature clearly shows that there are a number of carriers / routes for the cellular uptake of various HMS including both essential and non-essential ones. They do show significant non-specificity.

Heavy metal ion chelation and compartmentalization

Once the metal ions get entry into cytosol, they can be removed by chelation (Clemens 2001; Jabeen et al., 2009). The chelation and compartmentalization are defence mechanisms implied by plants to resist the harmful effects of metals (Cunningham et al., 1995; Sheoran et al., 2011). There are many organic and inorganic ligands present in the cytoplasm that are capable of heavy metal chelation. The main organic compounds involved in metal ion chelation are organic acids, amino acids, phytochelatins (PCs), metallothioneins (MTs), and cell wall proteins (Hall 2002; Sharma and Dietz, 2006; Fulekar et al., 2009; Seth, 2012). The main inorganic compounds are phosphates and silicates and organic compounds are malate and citrate (Bourg and Loch, 1995; Bhargava et al., 2012). These metal-chelating compounds reduce heavy-metal induced phytotoxicity by reducing the free metal ion concentrations through chelation (Salt et al., 1998). For example, Zn is converted to Znphytate Al to Aloxalate, Pb to Pb-carbonate, Pb-sulphate, and Pb-phosphate (Salt et al., 1998, Hall, 2002). The resulting complex is transported actively from the cytosol via tonoplast into the vacuoles. Vögeli-Lange and Wagner (1990) studied the protoplasts of tobacco plants (*Nicotiana rustica var. pavonii*) which exposed to Cd and reported that Cd was chelated by phytochelatins (PCs) and the Cd-PC complex was transported actively into the vacuole through tonoplast.

The role of other organic metal chelators in HM ions detoxification from cytoplasm is also important. Boominathan and Doran (2003) studied the Cd and Ni uptake and organic acid response in the hairy roots of *Thlaspi caerulescens and Alyssum bertolonii*, respectively and observed that the levels of citric, malic, and malonic acids were constitutively high in the hairy roots of both species and reported only 13 % of the total Cd and 28 % of the total Ni in *T. caerulescens* and *Alyssum bertolonii* hairy roots, associated with organic acids. Nicotinamine (NA) is another important metal chelator found in plants that forms strong complexes with most of the transition metal ions. The role for NA was projected in Ni-exposed roots of *T. caerulescens* when Ni- NA complex is identified after Ni-hyperaccumulation (Vacchina et al., 2003; Mari et al., 2006). The vacuoles are the last stop for these metal–chelator complexes as these complexes are removed from the cytoplasm by efflux of ions into vacuoles (Hall, 2002). This is achieved by the increased ability to transport metals into the vacuoles. For example, overexpression of a Zn transporter (ZAT) has been shown to increase the Zn tolerance by its sequestration into the metal ion sequestration in the apoplast or specialized cell types, e.g., epidermal cells, mesophyll cells, and trichomes (Eapen and D'souza, 2005). For example, Cd, Mn, and Pb were found to be sequestered in trichomes (Salt et al., 1998). Another effective way to get rid of excess ions is the translocation of metals into old leaves, which are then removed as a result of natural leaf shedding. For example, Zn was reported to accumulate in leaves during the last week prior to leaf shedding; thereby, plants use vacuoles (Hall, 2002). A few other compartmentalization strategies are also employed by plants other than metal ion sequestration into vacuoles. These include the heavy the leaf falls as a means of reducing the level of toxic metals inside the plant body (Ernst et al., 1992).

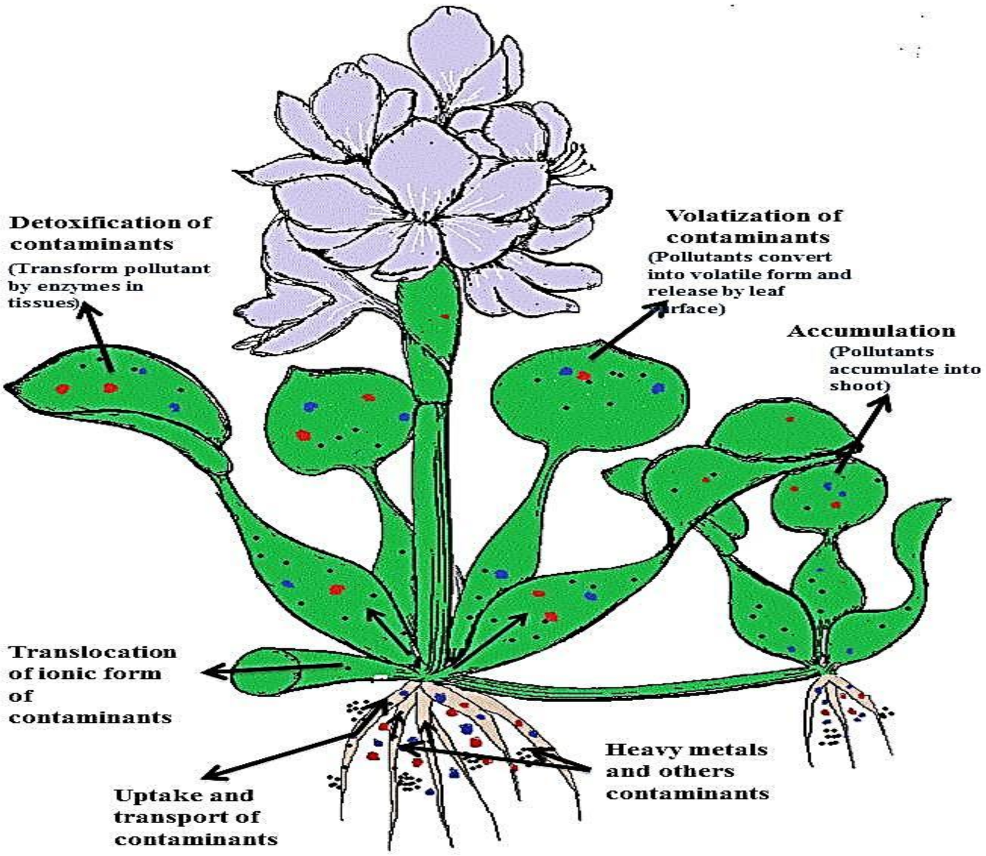


Fig 2.3: Phytoremediation mechanism and its techniques in plants

Factors affecting the phytoremediation

There are some factors which can affect the phytoremediation technique of heavy metals i.e. pH, temperature, light, salinity, organic acids, cations and anions present in growing medium etc. (Fritioff et al., 2005; Picard et al., 2005; Herb and Stefan, 2006; Chen et al., 2010;). It has been well established that the pH in a range of 2-5 increase the metal uptake while decreases at higher pH. Fritioff et al. (2006) examined the effect of temperature and salinity on uptake of metals in *Elodea Canadensis* and *Potamogeton natans* and reported that the metal uptake increases with increasing in temperature and decrease in salinity.

Plant species

The uptake of compounds is affected by plant species characteristic. The success of the phytoremediation techniques depends upon the identification of suitable plant species can accumulate heavy metal and produce huge amounts of biomass using traditional crop production and environment management practices (Ramirez-Rodriguez et al., 2005). Nowadays, the selection of native plants for phytoremediation for heavy metals is under investigation which also can help to determine the effect of different parameters on phytoremediation efficiency (Roy et al., 2015). The removal efficiency is highly related to growth rate, tolerance of plants to high level of metals and great adaptively to the different environments (Rezania et al., 2016).

Root zone

The Root zone is a special interest in phytoremediation. It can absorb metals and metabolizing inside the plant tissue. Degradation of contaminants in the soil and water by plant enzymes exuded from the roots is phytoremediation mechanism (Rawat et al., 2012; Rezania et al., 2016).

Properties of medium

Agronomical practices like addition of cheaters, fertilizers and pH adjustment influence the capacity of phytoremediation (Prasad et al., 2003; Rai, 2009). The amount of Pb uptake by plants is affected by the pH, salinity and the temperature (Tangahu et al., 2011). To reduce the lead by plants the pH of soil is adjusted with lime to a level of 6.5 to 7.0 (Traunfeld and Clement, 2001). Sanyahumbi et al. (1998) reported that lead removal remained at approximately 90% between 10˚C and 50˚C and varied from 30% of the initial lead concentration at pH 1.5 to approximately 95% at pH values of 3.5 and 4.5. The impact of salinity of heavy metal uptake was investigated through *Potamogeton natans* and *Elodea canadensis* and found that metal removal efficiency increased with decreasing salinity and increasing temperature (Fritioff et al., 2005).

Chelating agent

Uptake of heavy metals may be through addition of chelating agents i.e. EDTA, DTPA (diethylene tri amine penta acetic acid), citric acid, oxalate, phthalate, and Phosphonates (Tangahu et al., 2011). Chelating agents are commonly used to raise the bioavailability of heavy metals, thus enhancing their uptake capacity of plants. Chen et al. (2010) studied the phytoremediation of Cr (III) by *Ipomonea aquatica* from water contaminated with Cr (III) in presence of EDTA and chloride ions and reported that the accumulation of Cr (III) in root was increased with an increase in Cl. Further, it was also statistically postulated that the Cr concentration in root was significantly correlated with Cr-EDTA speciation and Cr accumulated in shoot was also significantly correlated with Cr-Cl speciation.

Vegetative uptake

It is affected by the ecological situations (Burken and Schoon, 1998). The temperature affects growth substances and consequently roots length. Root structure under field conditions differs from that under greenhouse condition (Merkl et al., 2005). Understanding mass balance analyses and the metabolic fate of pollutants in plants are the keys to sustaining the applicability of phytoremediation (Mwegoha, 2008).

Chemical properties of contaminants

Metal uptake by macrophytes depends on the bioavailability of the metal in the water, which in turn depends on the retention time of the metal, and interaction with other elements as well as substances in the water (Tangahu et al., 2011). Weekly soluble metal (Cr, Ag) absorbed by macrophyte in traces but soluble element (Zn, Cu, Ni) easily absorbed and transport into leaves (Siebielec et al., 2006; Laghlimi et al., 2015).

Macrophytes

Macrophytes are a diverse group of photosynthetic organisms found in water bodies. They include Bryophytes (mosses, liverwort etc.), Pteridophytes (ferns) and Spermatophytes (flowering plants). Chamber et al. (2008) reported that macrophytes divided into seven different plant divisions: Spermatophyta, Pteridophyta, Bryophyta, Xanthophyta, Rhodophyta, Chlorophyta, and Cyanobacteria. Arber (1920) and Sculthorpe, (1967) categorized macrophytes into four different categories depending on their growth forms: -

1) Emergent macrophytes- plants rooted in soil and also emerging to a significant height above water (e.g., ***Typha latifolia, Phragmites australis, Sagittaria trifolia, Eleocharis, Cabomba aquatica, Polygonum hydropiper, Eleocharis plantagenera, Scirpus mucronatus, Alternanthera philoxeroides***).

2) Submerged macrophytes- plants grow below the surface water include few ferns, numerous mosses and some angiosperm (e.g., ***Hydrilla verticillata, Ceratophyllum demersum, C. submersum Myriophyllum aquaticum, Elodea canadensis, Vallisneria americana, Utricularia vulgaria, Najas graminea etc.***)

3) Free floating macrophytes- plants that non-rooted to the substratum and floats on the surface of the water (e.g., ***Pistia stratiotes, Lemna gibba, Azolla pinnata, Salvinia molesta, Trapa natans, Eichhornia crassipes, Ipomoea aquatica,*** **etc.**)

4) Floating leaves macrophytes- plants occur submerged sediment and leaves are floating with long flexible petiole on the surface; mainly include angiosperm (e.g., ***Nymphea alba, Potamogeton cripus, P. natans, P. pectinatus, Nelumbo nucifera, Hydroryza aristata*** **etc**). Boyd (1970); Stewart (1970); Wooten and Dodd (1976); and Conwell et al. (1977) were inventers to demonstrate the pollutants removal potential of aquatic macrophytes. They considered as important components of the aquatic system,

not only as a food source, but because they also act as an effectual accumulator of heavy metals (Devlin, 1967; Rai, 2009; Deval et al., 2012; Sood et al., 2012).

Phytoremediation potential of macrophytes

Today, several researchers are vigorously involved in this attractive area i.e. phytoremediation. Various scientist studies are proved that floating plants i.e. *Lemna minor, Eichhornia crassipes, Pistia stratiotes* and *Salvinia herzogii* offer promising results for metal removal from industrial (Mahmood et al., 2005; Verma et al., 2005; Khan et al., 2009; Yasar et al., 2013; Akter et al., 2014; Sasmaz et al., 2015). Fritioff et al. (2005) suggested that submerged plants are completely flooded and have the capability to take up metals straight from the water and reducing metal concentrations in secondarily treated wastewaters. *Eichhornia crassipes* has high removal rates for various heavy metals like iron (Fe), zinc (Zn), copper (Cu), chromium (Cr), manganese (Mn), mercury (Hg) cadmium (Cd), and arsenic (As) from aqueous solutions and store metals in their bladders, followed by their translocation to stems, leaves and roots (Jadia and Fulekar, 2009; Muhammad et al., 2010; Rizwana et al., 2014). Rahman et al. (2008b) investigated *Salvinia natans* and *Salvinia minima*, for arsenic remediation reveal that As uptake increased with increasing exposure time and As concentration in the solution. Jafari et al. (2010) observed the accumulation potential of *Azolla microphylla* (94%), *A. filiculoides* (96%), *A. pinnata* (71%), and *A. microphylla* (98%), for Pb, Cu, Mn and Zn respectively, and *A. caroliniana* also has potential to accumulate Hg and Cr. The capacity of duckweed (*Lemna* sp.) to remove toxic heavy metals from water plays an important role in removal and accumulation of metals from contaminated water. Sasmaz and Obek (2009) reported that the aquatic plant *L. gibba* was used for the accumulation of As from secondary effluents as an alternative method for treatment. Miretzky et al. (2004) mentioned that the percentages removed by *P. stratiotes* were very high (>85% Pb, Cr, Mn and Zn) and also explained that *P. stratiotes* was able to eliminate the metals almost completely in the first 24 h of exposure. Chen et al. (2009b) investigated that *Ipomonea aquatic* remove Cr (III) from aqueous solution in the presence of chelating agent's chloride and EDTA and chloride can increase the solubility of Cr and enhance the bioaccumulation in shoots and roots of the plant.

Rai (2008) studied the phytoremediation capacity of *Azolla pinnata* for heavy metals removal and revealed an inhibition of its growth by 27.0-33.9% highest in the presence of Hg at 0.5 mg L^{-1} in comparison with control. After 2 weeks of the experiment, metal concentration was decreased up to 70-94%. Adhikari et al. (2010) evaluated the phytoremediation potential of two wetland plant species i.e., *Typha angustifolia* and *Ipomoea carnea* and reported that both plants show capacity for the removal of Pb from contaminated wastewater. Chandra and Yadav (2010) also estimated *Typha angustifolia* for remediation potential of various heavy metals (Cu, Pb, Ni, Fe, Mn, and Zn) and resolved that the Typha could be a possible phytoremediator for heavy metals from industrial wastewater at optimized condition. Unnikannan et al. (2013) evaluated the toxicity of Cr in *Salvinia natans, Pistia stratiotes* and *Eichhornia crassipes* and found that these plants accumulated higher concentrations of Cr in their tissues and classified as hyperaccumulators for Cr. Kumar et al. (2012) assessed the phytoremediation of heavy metal contaminated soil using five macrophytes viz., *Hydrilla verticillata, Eichhornia crassipes, Ipomoea aquatic, Bacopa monnieri* and *Marsilea minuta* and recommended that *E. crassipes* can be used for phytoremediation of Ni and Cu while *M. minuta* and *H. verticillata* can be applied for removal of Pb and Cr respectively from the contaminated water bodies. Susselan et al. (2006) acknowledged the effectiveness of *Mimosa pudica* for rhizofilteration of Hg, Cd, U and

Zn. Upadhyay et al. (2007) reported the sequence some macrophytes in order of removal efficiency of heavy metals by *Eichhornia crassipes*, *Lemna minor, Pistia stratiotes, Azolla pinnata, and Spirodela polyrhiza* was Fe<Cu <Cr <Cd<Zn<Ni which showed removal capacity of *E. crassipes* is highest.

Heavy metals uptake efficiency was calculated for *Salvinia* and *Spirodela*, and between two *Spirodela* was found to be more effective than *Salvinia* for the removal of lead and zinc (Srivastav et al., 1993; Chen et al., 2016). The accumulation and impact of Zn and Cu was studied in *Lemna minor* showed that Cu was comparatively more toxic than Zn, and both metals showed a synergistic effect regarding accumulation pattern (Dirilgen and Inel, 1994). Bioaccumulation and toxicity executed biochemical changes have been observed in *Vallisneria spiralis*, a rooted submerged macrophyte (Gupta and Chandra, 1998; Vajpayee et al., 2001; Rai, 2009; Zhu et al., 2016).

Table 2.6: Macrophytes and their phytoremediation potential for various metals

Water sources	Macrophytes	Common name	Metals	Accumulation in macrophytes (µg g^{-1})	Duration	References
Municipal waste water	*Phragmites australis*	common reed	Zn Mn Cu	20.5 52 6.57		Vymazal et al., (2010)
Leather industry waste water	*Salvinia Molesta*	Giant salvinia Or Kariba weed	Cu Cr Pb Cd	2.035 1.052 1.924 0.018	15 days	Ranjitha et al., (2016)
Synthetic solution	*Hydrilla verticillata*	Esthwaite Waterweed or Waterthyme	Cd	1.21	10 days	He et al., (2016)
Constructed wetland	*Cyperus alternifoltus*	Umbrella papyrus, Umbrella sedge or Umbrella palm	Al Cd Cu Mn Pb Zn	82.8 1.3 363 16.8 0.8 347	-	Cheng et al., (2002); Sun et al (2013)
Coal mining waste water	*Spirodella polyrrhiza*	Greater duckweed	Fe Cr Cu Cd Zn Ni	0.35 0.36 0.12 0.22 0.26 0.14	21 dyas	Mishra et al., (2008)
Synthetic solution	*Penisetum purpureum*	Napier grass or Elephant	Cr	925.0	8 weeks	Mant et al., (2006)

		grass or Uganda grass				
Synthetic solution	*Brachiaria decumbens*	Signalgrass	Cr	1694.5	8 weeks	Mant et al., (2006)
Synthetic solution	*Phargmites australis*	Common reed	Cr	406.2	8 weeks	Mant et al., (2006)
River water	*Eleocharis acicularis*	Needle spikerush and Least spikerush	Cu	8650	1 year	Sakakibara et al., (2011)
			Zn	6780		
			As	387		
			Cd	41.3		
			Pb	26.9		
Synthetic solution	*Ipomoea aquatica*	Water spinach	Cr	13218	14 days	Chen et al., (2010)
Industrial waste water	*Azolla pinnata*	Water velvet	Hg	310	13 days	Rai, (2008)
			Pb	740		
Synthetic solution	*Lemna minor* *Spirodella polyrrhiza*	Duckweed	Pb	561330	7 days	Leblebici and Aksoy, (2011)
Synthetic solution	*Azolla pinnata*	Water velvet	Cr	9125.3	7 days	Arora et al., (2006)
	A. microphyllum			14931.7		
	A. filiculoides			12383.6		
River water	*Phragmites australis*	Common reed	Cu	11.63	30 days	Salman et al., (2015)
Synthetic solution	*Phragmites cummunis*	Common reed	Fe	2813	28 days	Chandra and Yadav, (2011)
			Mn	814.40		
			Zn	265.80		
			Pb	92.80		
Waste water	*Egeria densa*	waterweed	As	195.95	14 days	Baker et al., (2013)
			Zn	441.38		

Synthetic solution	*Elodea canadensis*	Canadian Waterweed	Cu Zn, Cd	0.172 0.263 0.807	7 days	Torok et al., (2015)
Industrial waste water	*Myriophyllum spicatum*	*Eurasian watermilfoil*	Ni Cu Zn Pb Cd	2.4 8.2 3.6 1.3 2.8	60 min.	Lesage et al., (2007)
Syththetic solution*	*Lemna trisulca* *L. Minuta* *L. minor*	*Duckweed*	Zn	14931.7 12397.8 2397.8	10 days	Jafari and Akhavan, (2011)
Textile waste water	*Digitaria longiflora*	Crabgrass	Cu Cd Cr Pb Ni	0.15 0.12 0.11 2.34 0.05	30days	Yasar et al., (2013)
Tannery sludge	*Hydrocotyle umbellate*		Cr Cu Zn	18200 6660 15560	90 days	Khilji and Bareen, (2008)
Coal mine effluent	*Azolla pinnata*		Hg	0.44		Mishra et al., (2009)
Synthetic solution	*Callitriche cophocarpa*	Water starwort	Cr	470	10 days	Augustynowicz et al., (2010)
Lake water	*Trapa natans*		Cd Pb Cr	1.18 17.4 180	2 months	Petrovic et al., (2016)

Synthetic solution* prepared solution in known concentration

Lemna gibba, *Pistia stratiotes and Eichhornia crassipes* are major part of aquatic ecosystem and show high efficiency heavy metal removal. They absorb the contaminants and translocate their roots and shoots from polluted waste water (Zhu et al., 1999; Rai, 2009; Vesely et al., 2011; Prajapati, et al., 2012; Bokhari et al., 2016).

Duckweeds (*Lemna gibba*)

Duckweed, a common name of *Lemna*, *Spirodela*, *Wolffia and Wolffiella* belonging to Lemnaceae family, are the smallest and fastest growing angiosperms. The whole plant body is reduced to form a flat small leaf-like structure called fronds. They can survive in a wide range of pH and temperature (3.5–10.5 and 7–35°C), respectively. Due to the fact of high sensitivity to different pollutants, duckweed known as the aquatic plant which the special environmental concern needed (Radic, et al., 2010). They are dispersed globally in aquatic ecosystems and have been tested for wastewater treatment (Zirchy and Reed, 1988; Korner et al., 2003; Khellaf and Zerdaoui, 2009; Bokhari et al., 2016). Recently, Newete and Byrne (2016) showed that the *Lemna* species are the most usable plant for phytoremediation in comparison with the other aquatic macrophytes, this plant for nutrient recovery of nitrogen, phosphorus and toxic metals from domestic and agricultural wastewater used in many studies (Mohedano et al., 2012; Zhang et al., 2014). It has been used to recover heavy metals for over 30 years which the most of studies conducted with representatives *of Lemna genus, Lemna minor* and *Lemna gibba* (Guimaraes et al., 2012).

Within family, *Lemna gibba* represents the species most widely used in toxicity analysis, and in bioassays of water quality (Brain and Solomon, 2007; Radic et al., 2010). Duckweeds have been studied for treating industrial and municipal waste water, and for the removal of nutrients from lake and pond water (Forni et al., 2001; Korner et al., 2003). The results of several laboratory experiments accomplished with small-scale duckweed-covered group systems for nutrient removal and degradation of organic substances (Verma and Suther, 2015). The capacity of *Lemna gibba* to accumulate metals, and their defence mechanisms against the toxicity, have been reported by several authors (Teisseire and Guy, 2000; Axtell et al., 2003; Goulet et al., 2005; Oporto et al., 2006; Naumann et al., 2007; Khellaf and Zerdaoui, 2009; Megateli et al., 2009; Malec et al., 2010; Verma and Suther, 2015a,b). Rakhshaee et al. (2009) used potentiometric titration as analytical method to describe the portion of –COOH/–COO– on the heavy metals accumulation, and they found an increasing removal efficiency of metal ions by increasing the carboxylate from 0.92 to 2.42 mmolg^{-1} *Lemna*. Table 2.7 shows of capacity *Lemna* species for heavy metal removal.

Table 2.7: Phytoremediation capacity of *Lemna gibba*

Waste water sources (mgl^{-1})	**Metals**	**Removal efficiency (%)**	**Duration**	**References**
Sewage water	Cu Pb Zn Cd	100 100 93.6 66.7	8 days	El-Kheir et al., (2007)
Synthetic solution (10 mgl^{-1})	Zn	61.3	7 days	Khellaf and Zerdaoui (2009)
Synthetic solution (6 mgl^{-1})	Ni Cd	50 86.4	4 months	Parnian et al (2016)
Industrial waste water	Cr	89	7 days	Khan et al., (2009)
Synthetic solution (20 mgl^{-1})	Pb	89	10 days	Singh et al., (2012b)
Municipal waste water	Cu	55	15 days	Apelt (2010)

Synthetic solution (10 mgl^{-1})	Pb	96.7	7 days	Verma and Suthar (2015a)
	Cd	52.1		
Coal mining waste water	Cu	101.5	3days	Sasmaz et al., (2015)
	Pb	382		
	Zn	119		
Municipal and industrial waste water (Mixed)	Ni	99	7days	Bokhari et al., (2016)
Synthetic solution (10 mgl^{-1})	Pb	96.4	12 days	Abdallah, (2012)
	Cr	94.8		
Synthetic solution (20 mgl^{-1})	Zn	83	10days	Jafari and Akhavan, (2011)

Water lettuce (*Pistia stratiotes*)

Pistia stratiotes is a free-floating aquatic plant in the Araceae family with grey-green leaves, plant occur singly or connected to others by short stolons. Plants have numerous, feathery, roots. Leaves are often spongy near the base, densely soft pubescent with obvious parallel veins, slightly broader than long (Langeland et al., 2008). Flowers are inconspicuous, clustered on small, fleshy stalks nearly hidden in leaf axils, with single female flowers below and a whorl of male flowers above. Fruit grows from female flowers as a many-seeded green berry (Langeland et al., 2008). Water lettuce reproduces rapidly by vegetative off shoots formed on short, brittle stolons. It was found to be an effective removal tool for Cr, Cu, Cd, Se and Hg as reported by Qian et al. (1999) and Odjegba and Fasidi, (2004). *Pistia stratiotes* was also exposed to absorbed bromate and chromate ions from industrial solutions (Tel-or and Forni, 2011). As demonstrated by Gupta et al. (2012) that's large proportion of Fe, K, Mg, Mn, Ca, Cd, and Co attached to the external root surfaces of water lettuce by adsorption. Hua et al. (2012) found in Mn polluted wastewater, *Pistia stratiotes* can be effective phytoremediator plant. Great growth rate in wetlands, high coverage of water surface and easy harvestable is the key of water lettuce effectiveness. According to Lu et al. (2011) higher proportion of the metals plant remained in the roots rather than being transported to the shoots. Mishra et al. (2009) found that the water lettuce removed 80% of mercury (in concentration of 2 µg/L to10 µg/L) from the coal mining effluent in 21 days and the accumulation of Hg at low concentration was four times more into the roots in compare to the shoot. They also explained that higher accumulation of Hg in plants proportionally decreases the NPK, Chlorophyll and protein contents in plants. Table 2.8 shows capacity of *Pistia stratiotes* for heavy metal uptake from wastewater.

Table 2.8: Phytoremediation capacity of *Pistia stratiotes*

Water sources	Metals	Removal efficiency (%)	Duration	References
Coal mining waste water	Hg	97	21 days	Mishra et al., (2009)
Wetland water	Cr Co	100 86	4 days	Prajapati et al., (2012)
Pond water	Cd	60	21 days	Das et al., (2014)
Tannery waste water	Cr	58.27	15 days	Akter et al., (2014)
Textile waste water	Cd Cr Pb	88.58 95.11 112.04	-	Yasar et al., (2013)
Synthetic solution	As	53.63	10 days	Singh et al., (2015)
Steel foundry waste water	Pb Cu	70.7 66.55	7days	Aurangzeb et al., (2014)
Synthetic solution	Zn Fe	84.3 80.0	15 days	Miretzky et al., (2004)

	Mn	86.81		
	Cu	73.5		
	Cr	99.63		
	Pb	99.74		
Industrial waste water	Pb	31.5	15 days	Sukumaran, (2013)
	As	75		
	Cu	69.38		
	Cd	27.27		

Water hyacinth (*Eichhornia crassipes*)

Eichhornia crassipes commonly known water hyacinth is rapidly growing aquatic macrophytes which can double its biomass in a few days and one of the world's most troublesome weeds, this quality has also made it an applicant for use in phytoremediation (Dhote & Dixit, 2009). Many scientists proved that water hyacinth can accumulate heavy metals (Shaban et al., 2005; Sanita di Toppi et al., 2007; Skinner et al., 2007; Alvarado et al., 2008; Li et al., 2016). According to Odjegba and Fasidi (2007) *E. crassipes* is a promising macrophyte for remediation contaminated water, polluted with of Zn, Cr, Cu, Cd, Pb, Ag and Ni. It reduces of the very toxic Cr (VI) to the less toxic Cr (III) by the fine lateral roots, and translocate to leaf tissues (Lytle et al., 1998; Saha et al., 2016). This macrophyte induced transformation can be used in detoxification of contaminated wastewater. The phytoremediation efficiency of *Eichhornia* developed under diverse nutrient conditions has been tested for Fe-rich constructed wetlands (Jayaweera et al., 2004; 2007; 2008). Fe eradication happened largely by the process of Rhizofiltration. In addition to heavy metals, the capacity of *Eichhornia* to accumulate nitrogen about 60–85% from solutions containing different nitrogen loads (Fox et al., 2008; Xiang et al., 2009). The results of a field study undertaken in Hua-jia-chi pond on *Eichhornia* confirmed the potential of the species to decrease the concentration of N and P in water as well as in sediment (Xiang et al., 2009). The efficacy of sewage purification by water hyacinth has been exposed by Zimmels et al. (2006) in the laboratory also explain that cascade and semi-continuous pilot experiments verified that the plants are proficient of decreasing all tested parameter of water quality to levels that permit the use of the purified water for irrigation of tree crops.

Table 2.9: Phytoremediation capacity of *Eichhornia crassipes*

Water sources	Metals	Removal efficiency (%)	Duration	References
Synthetic solution	As	53.63	10 days	Singh et al., (2015)
River water	Cd Fe Ni	84.59 64.28 67.78	3 months	Bais et al., (2015)
Synthetic solution	Cd	90.8	21 days	Das et al., (2014)
Pond water	Pb Zn Cu Cd	75 43 29 61	-	Li et al., (2016)
Textile waste water	Cd Cu	69.93 102.35	7days	Yasar et al., (2013)
Textile waste water	Cu Zn Cr	94.44 96.88 94.78	96%	Mahmood et al., (2005)
Pulp and paper industry waste water	Pb Zn	80.3 73.4	20 days	Verma et al., 2005

Synthetic solution	Cr	81	30 days	Hadad et al., (2011)
	Ni	95		
	Zn	70		
Synthetic solution	Cr	89	28 days	Dixit and Dhote, (2010)
Steel industry waste water	Cd	82.8	30 days	Aurangzeb et al., (2014)
	Cu	78.6		
	As	74		
	Al	73		
	Pb	73		
	Zn	65.2		
	Cr	62.8		
	Fe	61		
Industrial waste water	Pb, Hg,	83.4	2 days	Fazal et al., (2015)
	Ni,	99.9		
	Cd	95.1		
		97.5		

It can be concluded that the *Pistia stratiotes, Eichhornia crassipes* and *Lemna gibba* are the effective free-floating aquatic plant species in phytoremediation process. Hence, wide range of submerged and emergent aquatic plants also has ability to uptake many types of heavy metals from different types of wastewater resources but these free-floating aquatic plants were more effective. Among phytoremediation techniques rhizofiltration was the method to uptake heavy metals from water bodies. Nowadays, the selection of native plants for phytoremediation of target heavy metals is under investigation which also can help to determine the effect of different parameters on phytoremediation efficiency. The removal efficiency is highly related to growth rate, tolerance to high level of metals and great adaptively to the different environments.

Chapter 3

Materials and Methods

MATERIALS AND METHODS

Study Location

1. Lucknow (Flashlight and battery manufacturing industry, Eveready, situated at 26. 89°N, 80.89°E).
2. Kanpur (Panki, thermal power plant, situated at 26.48°N and 80.24° E)
3. Kanpur (Cawnpore monochemi industry, situated at 26.27°N and 80.20°E)
4. Auraiya (Petrochemical industry, situated at 26.63°N and 78.53°E)

Water sampling

The wastewater sample for this study was collected from outlet pipes of treatment plant of the industries in 20l plastic containers.

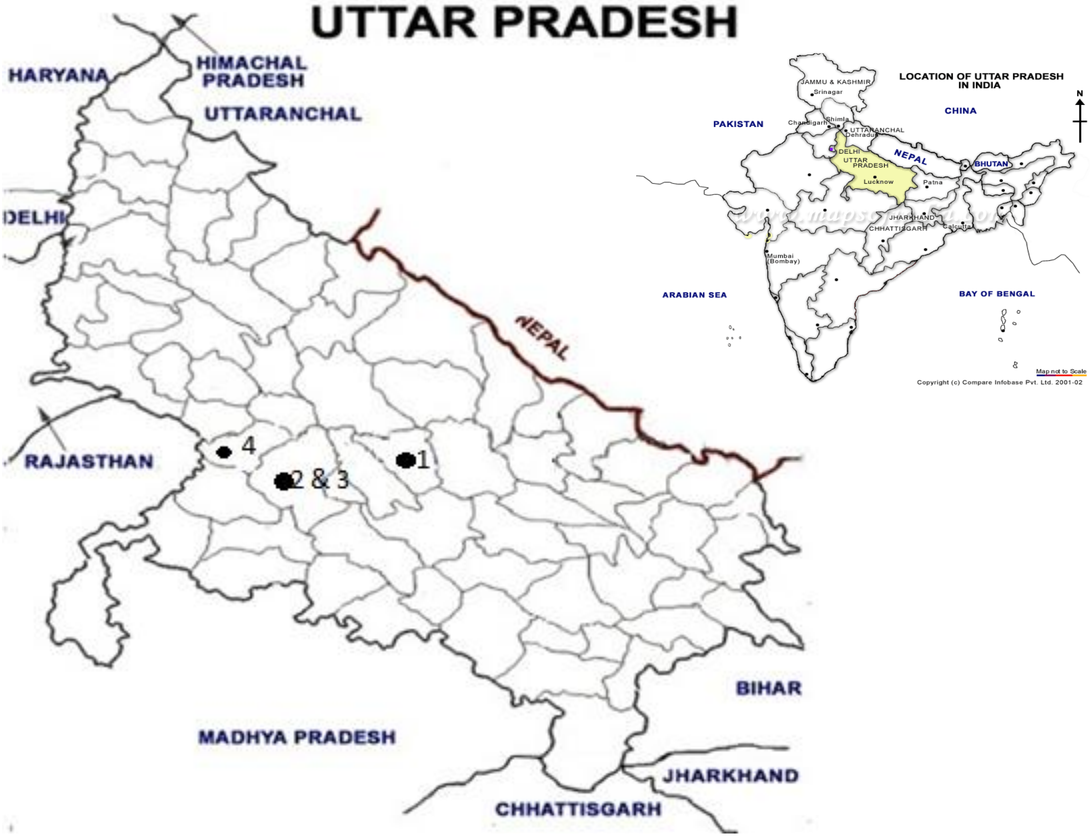


Fig. 3.1 Experimental location

Plant sampling

Hygroryza aristata, Azolla pinnata, Lemna gibba, Enhydra fluctuans, Trapa natans, Salvivnia molesta, Pistia stratiotes, Ipomoea aqauatica, Hydrilla verticillata, ceratophyllum submerus and Eichhornia crassipes with uniform length and weight was collected from river Gomti, Lucknow. All the collected plants were put in hydroponic system containing tap water and Hoagland solution for 2-week acclimatization before being exposed to heavy metals (Shah et al. 2016). *Pistia stratiotes, Eichhornia crassipes* and *Lemna gibba* are major part of aquatic ecosystem and absorb large contaminants and translocate their roots and shoots from industrial wastewater

Experimental setup for seed germination

Three varieties of rice-(Basmati- 198, Pusa sugandh and Sambhamsoori), pulses (Chick pea K-3256, Moong N-1, and Massor VL-1) and oilseeds (Mustard BR-40, Alsi KL-43 and Sesame G. til-3) seeds were used as test material. 40 seeds of each crop plant of each variety were placed in sanitized glass petridishes in triplicates and allowed to germinate on filter paper moisten with different dilution of various industrial wastewater at room temperature. For control, same procedure is followed with tap water for each experiment. After 48 hours germination measured, the root length in cm was measured and the emergence of the radicle was taken as the criterion for germination.

Experimental setup for phytoremediation

Macrophytes were allowed to grow in different dilution (100%, 75%, 50%, 25% and 0%) of industrial waste water in plastic tubs measuring 25 . Plants were harvested at the interval of 1 week. The experiment was continued till five weeks i.e. 3 samples of plants along with wastewater were examined for changes in heavy metals concentration. After

35 days plants were uprooted to determine the metal translocation within the plant and the efficiency were calculated with respect to the initial concentration of wastewater.

Fig. 3.2 Experimental setup for phytoremediation.

Analysis of Waste water sample

1- pH

Reagent used-

- Pthalate buffer (4 pH),
- Phosphate buffer (7 pH),
- borax buffer (9 pH)

Procedure-

First calibrate the pH meter with the help of buffer 4 pH, 7 pH and 9 pH. After calibration, rinse the glass electrode with distilled water, dry with a tissue paper then place in wastewater sample and record pH value.

2- **Conductivity**

Reagent used-

- **KCl**

Procedure-

First calculate cell constant by using standard KCl with the help of conductivity electrode. Then wastewater conductivity was recorded.

Calculation-

$$\text{Cell constant} = \frac{1412}{\text{C}} \text{x} \, [1 + 0.019(\text{t} - 25)]$$

$$\text{Concuctivity} = \text{Cm} \times \text{Kc} / [1+0.019(\text{t}-25)]$$

Where,

C= Measured conductivity of KCl

Kc= Cell constant (cm^{-1})

t= Observed temperature

Cm= Measured conductivity of sample

3- **Acidity**

Reagent used-

- **Standard sodium hydroxide** (0.05N)-0.8g NaOH dissolved in 1000ml distilled water. Stored in air tight glass bottle.

- **Phenolphthalein indicator-** 0.1 g Phenolphthalein indicator in 100ml ethyl alcohol and 100ml distilled water.
- **Methyl orange indicator-** 0.1 g methyl orange in 200ml.

Procedure-

50 ml of waste water sample taken, add 2 -3 drop of methyl indicator. No colour appears, add 2 drop of phenolphthalein indicator and titrate with 0.02N NaOH standard. Faint pink colour appears in last.

Calculation-

$$\text{Acidity (mg/l)} = \frac{\text{ml of titrant used}}{\text{ml sample is taken}} \times 1000$$

4- Alkalinity

Reagent used-

- **Standard H_2SO_4 (0.02N)-** 3ml conc. H_2SO_4 in 1000 ml (0.1N) and then dilute up to 1000ml (0.02N).
- **Phenolphthalein indicator-** 0.1 g Phenolphthalein indicator in 100ml ethyl alcohol and 100ml distilled water.
- **Methyl orange indicator-** 0.1 g methyl orange in 200ml.

Procedure-

Alkalinity of the sample is measured titrating with 0.02 N H_2SO_4 (3ml of concentrated H_2SO_4 in 1000ml distilled water and further diluted 1000 times with distilled water) and by adding 2-3 drop of methyl and phenolphthalein indicator one by one.

Calculation-

$$\text{Alkalinity(mg/l)} = \frac{A}{V} \times N \times 50 \times 1000$$

Where, N= normality of H_2SO_4 and

A = H_2SO_4 used in titration.

V= Volume of sample taken

5- **Dissolved oxygen (DO)**

Reagent used-

- **Manganous sulphate-** 36.4g in 100ml distilled water.
- **Alkaline iodide azide solution-** 50g NaOH, 15g KI, and 1g sodium azide in 100ml distilled water.
- **Starch indicator-** 0.5 g in 100ml distilled water.
- **Standard sodium thiosulfate (0.025N)-** 3.015 sodium thiosulfate in 500ml distilled water.

Procedure-

DO of sample is measured by azide or winkler's method. Wastewater collected in 300ml BOD bottles and fixed with 1ml $MnSO_4$ and 1ml alkaline azide solution and then add 1ml H_2SO_4 and 1ml starch indictor after that titrate with 0.025N sodium thiosulfate, blue colour disappears.

Calculation-

$$\mathrm{DO\ (mg/l)} = \frac{\mathrm{ml\ of\ titrant} \times N \times 8}{V2(V1 - v)V1} \times 1000$$

Where,

N = Normality of titrant (sodium thiosulphate)

V1 = Volume of BOD bottles,

V2= Volume of content titrated, and

v = Volume of $MnSO_4$ and Alkaline azide used.

6- Biological oxygen demand (BOD)

Reagent used-

- **Manganous sulphate-** 36.4 g in 100ml distilled water.
- **Alkaline iodide azide solution-** 50g NaOH, 15g KI, and 1g sodium azide in 100ml distilled water.
- **Starch indicator-** 0.5 g in 100ml distilled water.
- **Standard sodium thiosulfate (0.025N)-** 3.015 sodium thiosulfate in 500ml distilled water.

Procedure-

First diluted the sample with aerated distilled water and transferred into BOD bottles. K one bottle kept for determination of the initial DO and others bottles incubated at 20°C for 5 days, bottles have air tight. Prepare a blank by using aerated distilled water. After 5th day final DO was measured and BOD was calculated.

Calculation-

$$\mathrm{BOD\ (mg/l)} = \frac{(\mathrm{D1} - \mathrm{D2}) - (\mathrm{B1} - \mathrm{B2})}{\mathrm{P}} \times \mathrm{F}$$

Where,

D1= DO of the sample bottle on 0-day (before incubation).

D2= DO of the sample bottle after 5th day.

B1= DO of the blank bottle on 0-day.

B2= DO of the blank bottle after 5th day.

F= ratio of seed in diluted sample to seed in seed control

P= decimal fraction of the dilution water used.

7- Hardness

Reagent used-

- **Buffer solition-** Dissovle 8.45g NH_4Cl in 71.5 ml NH_4OH, add 0.75g EDTA Mg salt and dilute 250ml with distilled water.
- **Eriochrome black T indicator-** 1 pinch per sample.
- **Standard EDTA-** Dissolve 3.72 g EDTA disodium salt in 1000ml distilled water.
- **Inhibitor-** Disslve 0.75 g hydroxylamine hydrochloride in 50ml alcohol.

Procedure-

50 ml of water sample is taken in conical flask and add 2 ml of buffer solution, 1 ml inhibitor and pinch of Erichrome black T indicator then titrate with standard 0.01 M EDTA solution till red wine colour turn blue. Same procedure is done of blank.

Calculation-

$$\text{Total hardness (mg/l)} = \frac{\text{ml of titrant used}}{\text{ml of sample taken}} \times 1000$$

8- Total solid

Procedure-

Clean 100 ml beaker was taken and dried at 103 to 105°C for 1h. Store and cool the dish in desiccator until needed. Put 50 ml unfiltered well-mixed sample and dried in hot air oven at 80°C to 100°C up to dryness then cool in desiccator and take the final weight. Cycle of drying, cooling, desiccating and weighing repeated until a constant weight is obtained.

Calculation-

$$\text{Total solid(mg/l)} = \frac{(\text{Wf} - \text{Wi})}{\text{ml of sample taken}} \times 1000$$

Where,

Wi= Initial weight of the evaporating dish (g)

Wf= Final weight of evaporating dish (g)

9- Total suspended solid (TSS)

Firstly, initial weight of filter paper (0.45µm) was taken, and then 50 ml of waste water sample filter through it and then filter paper dried in hot air oven at 80°C to 100°C up to dryness then cool in desiccator and take the final weight. Cycle of drying, cooling, desiccating and weighing repeated until a constant weight is obtained.

Calculation-

$$\text{Total suspended solid(mg/l)} = \frac{(\text{Wf} - \text{Wi})}{\text{V}} \times 1000$$

Where,

Wf= Initial weight of the evaporating dish (g)

W2= Final weight of evaporating dish (g)

V= Volume of the sample taken (ml)

10- Total dissolved solid (TDS)

50 ml of filtered sample was taken and dried in hot air oven at 80 °C to 100°C up to dryness then cool in desiccator and take the final weight. Cycle of drying, cooling, desiccating and weighing repeated until a constant weight is obtained.

Calculation-

$$\text{Total dissolved solid(mg/l)} = \frac{(\text{Wf} - \text{Wi})}{\text{V}} \times 1000 \times 1000$$

Where,

Wi= Initial weight of the evaporating dish (g)

Wf= Final weight of evaporating dish (g)

V= Volume of the sample taken (ml)

11- Chemical oxygen demand (COD)

Reagent used-

- **Standard potassium dichromate (0.25N)-** Dissolve 12.25 g in 1000ml distilled water.
- **Sulphuric acid-silver sulpfate-** 5g silver sulphate in 500ml conc. H_2SO_4.
- **Standard ferrous ammonium sulphate (0.25N)-** Dissolve 98g ferrous ammonium sulphate in 400ml distilled water with 20ml H_2SO_4 and dilute upto 1000ml**.**
- **Ferroin indiactor-** 8 to 10 drops per sample.
- **Mercuric sulphate-** 0.02 g per sample used.

Procedure-

COD of the waste water sample is measured by using reflux digestion method. 10 ml of sample (1ml waste water + 9ml distilled water) was taken in digesting tubes, in this 5ml of $K_2Cr_2O_7$, 15ml of H_2SO_4+ $AgSO_4$ and 0.02g $HgSO_4$. Sample are reflux for 2h, then cool and makeup volume up to 70ml. 8 drop of ferroin indicator was added and then titrate with standard ferrous ammonium sulphate solution, blue green colour change into red wine colour. Same procedure repeated with blank.

Calculation-

$$\mathrm{COD\ (mg/l)} = \frac{(\mathrm{A} - \mathrm{B})}{\mathrm{V}} \times \mathrm{N} \times 8 \times 1000$$

Where,

A = ml of ferrous ammonium sulfate used for blank

B = ml of ferrous ammonium sulfate used for sample

N = Normality of ferrous ammonium sulfate

V= Volume of the sample taken (ml)

12- Phosphate

Reagent used-

- **Ammonium molybdate-** Dissolves 2.5 g ammonium molybdate in 17.5 ml, add 28 ml H_2SO_4 in 40 ml distilled water then add in to molybdate solution and dilute upto 100ml with distilled water.
- **Stannous chloride-** 2.5 g stannous chloride in 100 ml glycerol, heat in water bath with continuous stirring.

Procedure-

For analysis of phosphate 100 ml sample taken and Add, 4 ml of molybdate reagent and 0.5 ml of stannous chloride reagent. After 10 min, but before 12 min, absorbance measured by using photo spectrometer at 690 nm and compare with a calibration curve using distilled water blank.

Calculation-

K-Factor = Absorbance/ Concentration

Phosphate (µg ml^{-1}) = K- factor × Absorbance

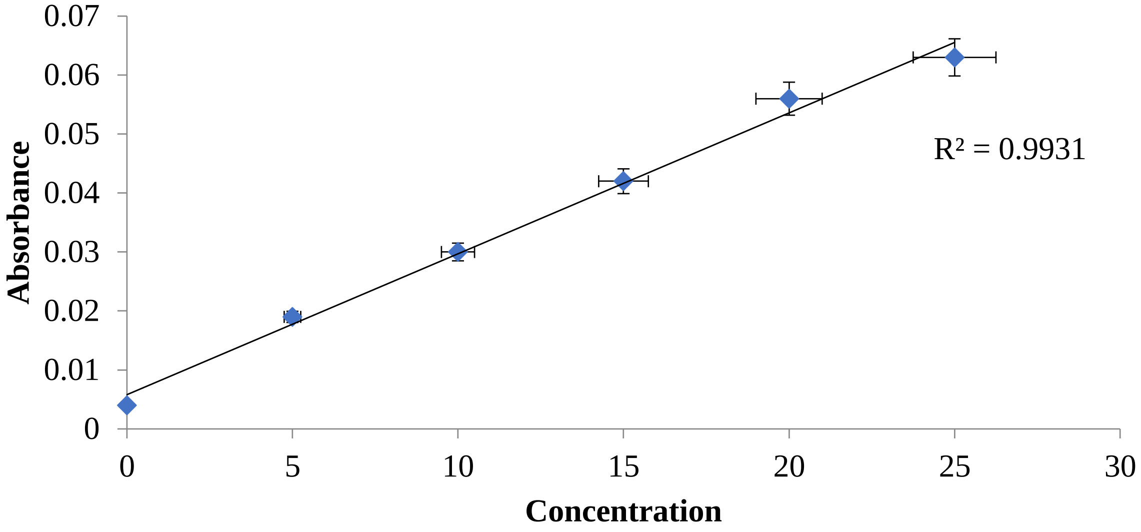


Fig. 3.3: Standard Curve of Phosphate

13- Nitrate

Reagent used-

- **Salicylic acid-**2.5 gm salicylic acid in 50 ml conc. H_2SO_4.
- **Sodium hydroxide (2N)-** 10g NaOH in 125 ml distilled water.

Procedure-

Nitrate is measured by Catalado method (Catalado et al., 1975) with the help of 5% Salicylic Acid and 2N NaOH. Add 0.5 ml of 5% salicylic acid and 9.0 ml of 2N NaOH in 0.5 ml of water sample in a test tube, orange- yellowish color. Take O.D. at 410 nm.

Calculation-

Nitrate = K- factor × concentration

K- Factor = Absorbance/ Concentration

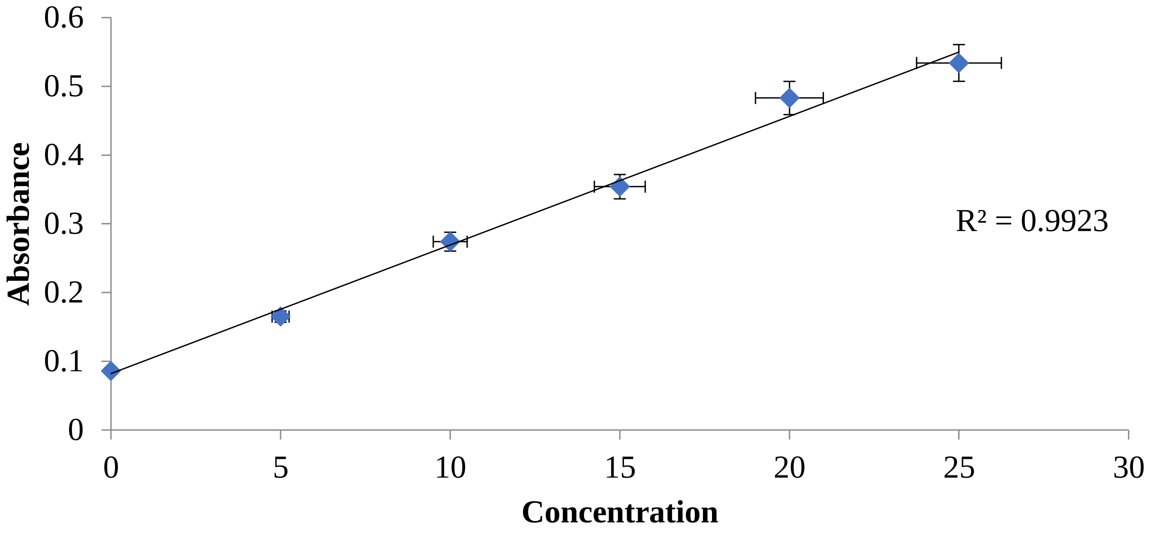


Fig. 3.4: Standard Curve of Nitrate

14- Total kjeldhal nitrogen

Reagent used-

- **Digestion reagent-** Dissolve 67g K2SO4 and 3.65g $CuSO_4$ in 400ml distilled water, 67 ml H_2SO_4 added carefully in solution and then diluted upto 1000ml.
- **Phenolphthalein indicator-** 1g phenolphthalein indicator in 100ml and 100ml distilled water.
- **Sodium hydroxide-sodium thiosulfate reagent-** Dissolved 100g NaOH and 5g in 200ml distilled water.
- **Mixed indicator-** 200mg methyl red in 100 ml alcohol, 100mg methyl blue in 50 ml alcohol and mixed.
- **Boric acid and mixed indicator solution-**10g boric acid, 5ml mixed indicator and 500ml distilled water.
- **Standard H_2SO_4 (0.02N)-** Add 3ml H_2SO_4 in 1000ml (0.1N) distilled water and then dilute 20ml solution with H_2SO_4 to obtained 0.02 N H_2SO_4.

Procedure-

50 ml sample taken in Kjeldahl flask, carefully add 50 ml of digestion reagent and digest sample until coloured - turbid samples will become transparent and pale green, cool, dilute to 300 ml with water and mix well and carefully add 50 ml of sodium hydroxide-thiosulfate reagent allow to distillation. Take 50 ml of boric acid indicator solution in 500 ml and place it below the condenser of the distillation assembly, 200 ml of distillate is collected in flask (Pink colour of sample turn green due to ammonia absorption). Titrate the distillate with O.2N H2S04 untilled pink colour appears. Run distilled water blank in the same manner.

Calculation-

$$\text{Total kjeldahl nitrogen (mg/l)} = \frac{(\text{A} - \text{B})}{\text{V}} \times \text{N} \times 14 \times 1000$$

Where,

A= Volume of H_2S0_4 required for sample, ml

B= Volume of H_2S0_4 required for blank, ml

N= Normality of H_2S0_4 use as titrant.

V= ml of sample taken

15- Chloride

Reagent used-

- **Potassium chromate-** Dissolving 2.5g in 250 ml distilled water.
- **Silver nitrate (0.02N)-** Dissolving 0.958g Silver nitrate in 200 ml distilled water.

Procedure-

For measurement of chloride, 20 ml of waste water sample taken and 1 ml of potassium chromate indicator was added and then titrate against silver nitrate untilled colour change yellow to brick red.

Calculation-

$$\text{Total kjeldahl nitrogen (mg/l)} = \frac{\text{V1}}{\text{V}}\text{N} \times 35.15 \times 1000$$

Where,

V1= Volume of $AgNO_3$ used

N= Normality of $AgNO_3$

V= ml of sample taken

16-Oil and Grease

Reagent used-

- **Concentrated HCl**
- **Petroleum ether**

Procedure-

250 ml sample taken in 500ml beaker and acidify with adding 5 ml HCl in them. Then transfer sample into separating funnel by adding 30 ml of petroleum ether and vigorously shaken for 5 min. Allowing sample to layer formation. Upper layer is petroleum ether and lower is of water become distinct. Lower one is discarded from separating funnel. Then filtered sample in weighted beaker and evaporate them in water bath, then final weight is taken.

Calculation-

$$\text{Oil and grease (mg/l)} = \frac{\text{W2} - \text{W1}}{\text{V}} \times 1000$$

Where

W1= Initial weight of beaker,

W2= Final weight of beaker, and

V= Volume of sample taken

17- Sodium

Reagent used-

- **Stock solution (1000ppm)-** 2.54 g NaCl in 1000ml distilled water.
- **Working solution (100ppm)-** 10 ml of stock solution in 90 ml distilled water.
- **Sodium standard-**
    - 10ppm- 10 ml working solution in 90 ml distilled water.
    - 20ppm-20 ml working solution in 80 ml distilled water.
    - 30ppm-30 ml working solution in 70 ml distilled water.
    - 40ppm-40 ml working solution in 60 ml distilled water.
    - 50ppm-50 ml working solution in 50 ml distilled water.

Procedure- Standardized flamephotometer using different standards and plot standard graph between concentration and emission of standard sodium concentration. Samples reading also taken in filtered wastewater through flamephotometer.

18- Potassium

- **Stock solution (1000ppm)-** 1.90 g KCl in 1000ml distilled water.
- **Working solution (100ppm)-** 10 ml of stock solution in 90 ml distilled water.
- **Sodium standard-**
    - 10ppm- 10 ml working solution in 90 ml distilled water.

  - 20ppm-20 ml working solution in 80 ml distilled water.
  - 30ppm-30 ml working solution in 70 ml distilled water.
  - 40ppm-40 ml working solution in 60 ml distilled water.
  - 50ppm-50 ml working solution in 50 ml distilled water.

Procedure- Reading taken on flamephotometer using potassium filter (Same as sodium).

19- Calcium

- **Stock solution (1000ppm)-** 2.76g in 1000ml distilled water.
- **Working solution (100ppm)-** 10 ml of stock solution in 90 ml distilled water.
- **Calcium standard-**
  - 10ppm- 10 ml working solution in 90 ml distilled water.
  - 20ppm-20 ml working solution in 80 ml distilled water.
  - 30ppm-30 ml working solution in 70 ml distilled water.
  - 40ppm-40 ml working solution in 60 ml distilled water.
  - 50ppm-50 ml working solution in 50 ml distilled water.

Procedure- Reading taken on flamephotometer using calcium filter (Same as sodium).

20- Sulfate

Reagent used

- **Buffer solution**- Dissolve 30 g magnesium chloride, 5g sodium acetate, 1g potassium nitrate and 20 ml acetic acid in 1000ml distilled water.
- **Barium chloride-** one pinch per sample.
- **Standard sulphate-** Dissolve 0.147g in 1000ml distilled water.
- **Working solution** – 10 ml stock solution in 90 ml distilled water.
  - 10ppm- 10 ml working solution in 90 ml distilled water.
  - 20ppm-20 ml working solution in 80 ml distilled water.

- 30ppm-30 ml working solution in 70 ml distilled water.
- 40ppm-40 ml working solution in 60 ml distilled water.
- 50ppm-50 ml working solution in 50 ml distilled water.

Procedure- Take 100ml sample; add 20 ml buffer solution and 0.3 g $BaCl_2$ mixed well in magnetic stir for 1 min. Take absorbance at 420nm at spectrophotometer.

Calculation-

From the calibration curve,

$$y= mx+c$$

Where,

y= absorbance of the sample,

m=slope of the straight line,

x= concentration of sulphate (mg)

$$\text{Sulphate (mg/l)} = \frac{\text{Conc. of suphate}}{\text{Sample taken}} \times 1000$$

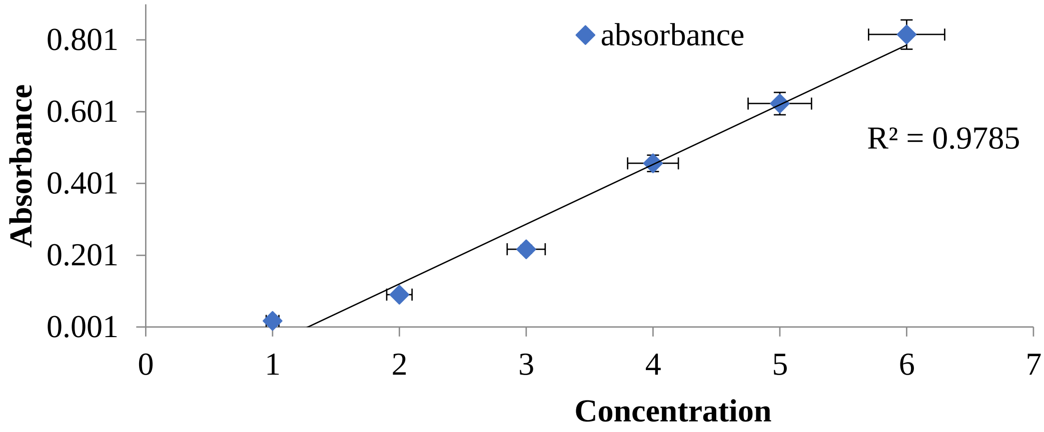


Fig 3.5 Calibration curve of sulphate

21- Heavy metal digestion

Reagent used-

- **Nitric Acid-**
- **Perchloric acid-**

Procedure-

50 ml of a well-mixed sample into 250 ml conical flask has been taken and digested with aquaregia (HNO_3 and $HClO_4$ acid in 3: 1 ratio v/v) on hot plate untilled sample volume reduced to 10 ml. then transfer sample in 50ml volumetric flask diluted with 0.1N HNO. Plant sample also digested with same method All the plants samples washed and cut into small pieces and dried in oven at 60ºC temperature and then homongeniged using mortal pestle. One gram of sample digested with aquaregia (HNO_3 and $HClO_4$ acid in 3:1 ratio v/v) when sample became clear and white, Fumes of $HClO_4$ appeared (AOAC 1984) and then diluted with 0.1N HNO3 and filter with 0.45 µm filter paper. Then metals were analysed by atomic absorption spectrophotometer.

Calculation-

$$\text{Heavy metals (mg/l)} = \frac{(A - B)}{V} \times V1 \times F$$

Where,

A= Heavy metal conc. in sample

B= Heavy metal conc.in blank

V_1= Volume makeup and

F= Dilution factor

V= Sample taken in ml

Effluent Tolerance Index (ETI) was also calculated from the radicle length data using the following formula (Turner and Marshal, 1972).

$$\mathrm{ETI} = \frac{\text{Radicle length of seed test}}{\text{radicle length of control}}$$

Phytotoxicity percentage was deduced from the effect of wastewater on radicle growth using the following formula developed by Chou et al., (1976; 1978) and modified by Ray and Banerjee, (1994):

$$\text{Phytotoxicity (\%)} = \frac{\text{Radicle length of control} - \text{radicle length of test}}{\text{radicle length of control}} \times 100$$

Germination percentage estimate the viability of seeds calculated using following formula- (Ellis and Roberts, 1980).

$$\text{Germination (\%)} = \frac{\text{No. of seed germinated}}{\text{No. of seeds}} \times 100$$

Germination index (GI) provides a comprehensive interpretation, seed germination and root elongation, according to the equation (IRSA, 1983):

$$\text{Germination Index (GI)} = (G_S L_S) / (G_C L_C)$$

Where, G_S and L_S are seed germination (%) and root elongation (cm) for the sample G_c and L_c the corresponding control values. To facilitate the comparison between different tests, the GI is here expressed as a percentage of the GI for controls, which is equal to 100.

Phytotoxicity index (PI) was evaluated based on germination percentage and radicle length of seeds according to Mekki et al. (2007) it was calculated by following equation-

$$\mathrm{PI} = (1 - \frac{\text{Radicle length in treated effluent}}{\text{Radicle length in control}})$$

Seedling vigour index evaluated the supplement germination and viability and understand performance of a seed (Abdul-Baki and Anderson, 1973).

SVI= Germination % × radicle length

Removal Efficiency (RE) After five weeks of treatment metals concentration in the effluent was analysed and the removal efficiency was calculated following the formula giving by Tanhan et al. (2007).

$$\text{Removal efficiency} = \frac{\text{Ci} - \text{Cf}}{\text{Ci}} \times 100$$

Where Ci is initial concentration of metals in sample and Cf is the final concentration of metals in the water sample after treatment and is expressed as percentage (%).

Translocation factor (TF) is the ratio of heavy metal in root to shoot and is used to determine the plants potential for heavy metals accumulation (Barman et al., 2000; Gupta et al. 2008).

$$TF = \frac{\text{Concentration of metal in plants in shoots}}{\text{Concentration of metal in corresponding plant roots}}$$

Enrichment coefficients (EC) are a very important factor, indicate the relationship of metals in macrophytes and water also named as bioconcentration factor (Sasmaz et al., 2008). The Enrichment coefficient (EC) was calculated as follows (Rahmani and Sternberg, 1999).

$$\text{Enrichment coefficient} = \frac{\text{Metal accumulated in plants parts}}{\text{Metal concentration in water}}$$

Statistical Analysis

The data (n=3) were analyzed statistically by one-way ANOVA (SPSS 16.0, Statistical Package and MS Excel) using tukey test determine the significance of difference among treatments at probability (*p*) 0.05 and 0.001.

Chapter 4

Results and Discussion

Chapter 4.1

To analyse the physico-chemical characteristics including heavy metals of different industrial wastewater.

To analyse the physico-chemical characteristics including heavy metals of different industrial wastewater.

With the industrialization of country large volume of wastewater or effluent generated daily a principal factor of water pollution which contributing to oxygen demand and nutrient loading of the water bodies, promoting algal blooms and damaged aquatic ecosystem (Okareh and Adeolu, 2015). Characteristics of the effluent depends on the nature of the industries and their treatment. In Uttar Pradesh various polluted industries are categories by CPCB, in which battery industry, Petrochemical industry, chemical industry and thermal power plants were selected for my research experiments. The aim of this study to check the quality level of these industries which is daily discharged into municipal drain which eventually flows into rives, a primary source of agriculture and fishing activities.

Physico-chemical properties

The all industrial effluent was colorless except flashlight manufacturing industry effluent which is greenish brown may be due to color producing compound, or may be excess algal growth, sometimes temperature and pH also responsible for water colored. Excess color reduces the photosynthesis activity of macrophytes and also affects other important parameters such as DO, BOD. Temperature is one of the most important ecological features for life function which was the set of biochemical reactions. It was varying between 25-32ºC in all industrial effluent. The main reason of temperature is cooling system of manufacturing and thermal plants industries. It can control behavioral characteristics of microorganisms, gases and salts solubility in water.

Turbidity is the cloudiness of water that’s caused due to the presence of excessive amounts of suspended matter, such as mud, silt, clay and calcium. Turbidity was found 65.5, 27.3, 43.2, 20.17 and 0.03 NTU in flashlight manufacturing, petrochemical, and chemical industry, Panki thermal power plants, and control respectively. Water appearance with <10 NTU turbidity is usually acceptable (BIS, 2003). Excessive turbidity, in water is not only aesthetically disagreeable; it may also cause health distress. Effluent from flashlight and battery manufacturing and chemical industry are highly turbid as compare to petrochemical and thermal power plants.

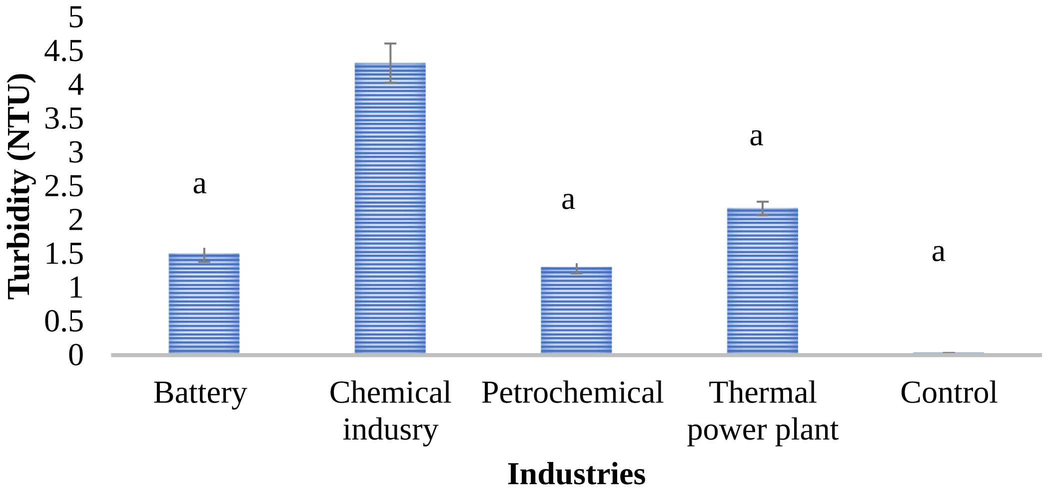


Fig 4.1.1: Turbidity found in industries as well as control. (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values).

pH is the simple parameter but extremely important, is a measure of the acidity or alkalinity of water (Lokhande et al., 2011). Its affect aquatic system and change toxicity of other pollutants in one form or the other, for example heavy metal toxicity get enhanced at particular pH value which makes them an important water parameter to decide the quality of the wastewater effluent (Tafesse et al., 2015). pH of the effluent was found 7.2, 7.1, 8.4, 7.7 and 6.9 of flashlight manufacturing industry, petrochemical industry, chemical industry and Panki thermal power plant and control respectively.

Results indicates that nature of the industrial wastewater are slightly alkaline and under permissible level of CPCB (6.5-8.5).

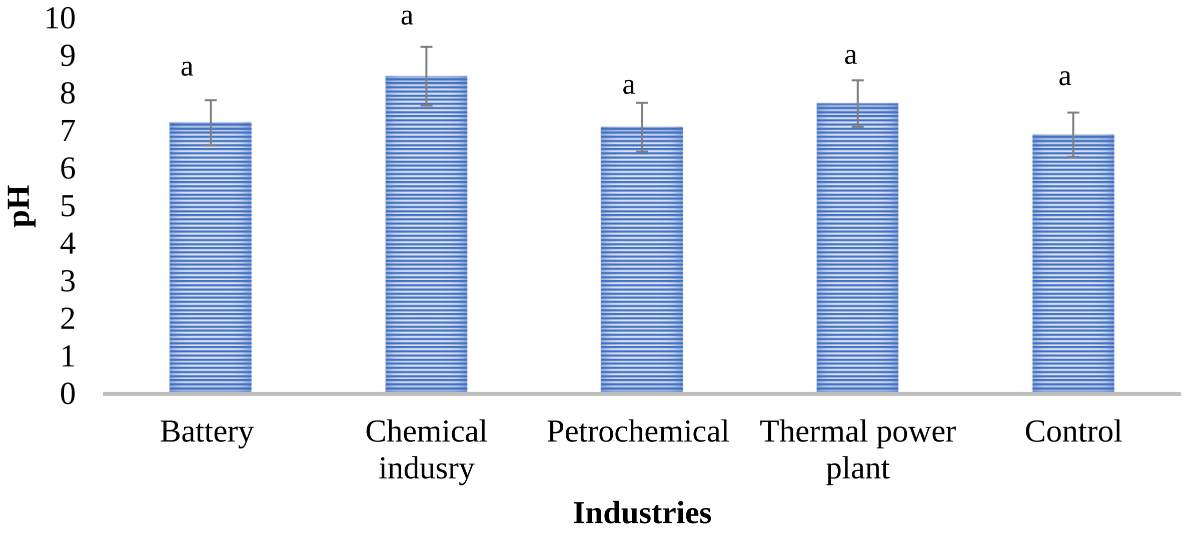


Fig. 4.1.2: pH recorded in industries as well as control (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Electrical conductivity (EC) of samples was range from 968-1279 S/cm, indicate the reasonable level of pollution (permissible level 1000µS/cm, given by WHO, (2002). EC the combination of dissolved and dissociated substance depends on temperature, dissociation, concentration and migration of ions in electric field, but it doesn't give ideas which types of ions present. The high conductivity value in the effluents can increase the salinity of the receiving river, which may result in adverse ecological effects on the aquatic biota and such high salt concentrations hold potential health hazard (Okareh and Adeolu, 2015).

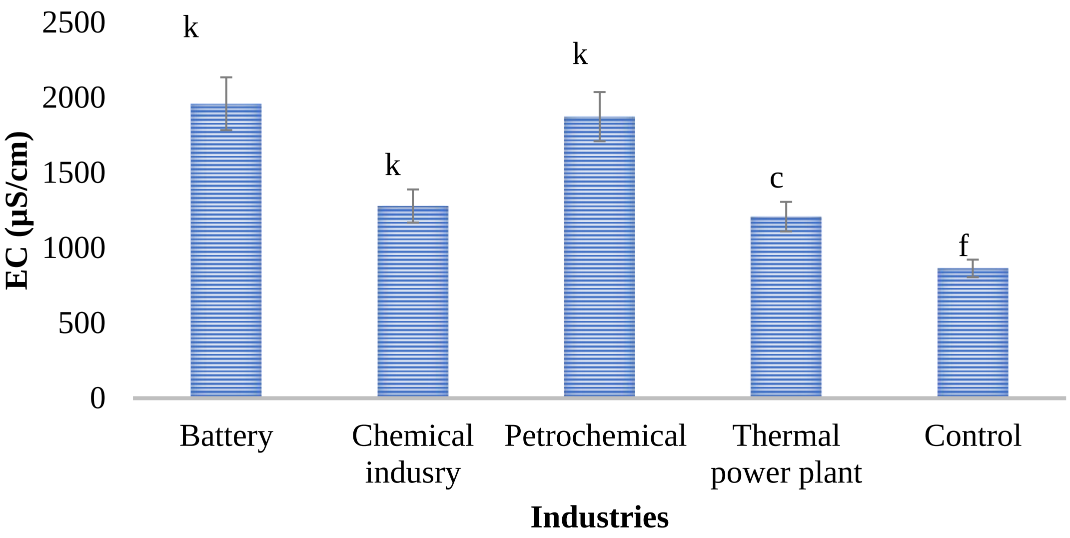


Fig. 4.1.3: Electric conductivity recorded in industries as well as control (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Solids refer to total suspended solids (TSS) and total dissolved solid (TDS) present in wastewater sample. TSS composed of silt, decaying matters, suspended industrial waste. TSS was found to be 123.22, 172, 121.05, 54.02 and 1.58 mgl^{-1} of flashlight manufacturing industry, petrochemical industry, chemical industry and Panki thermal power plant and control respectively which was found higher than prescribed industrial wastewater discharge limits of CPCB (30-50mgl^{-1}). TDS composed mainly of carbonates, bicarbonates, chlorides, phosphates, nitrates calcium, magnesium, potassium and organic substances. In the analysis TDS lies in the range of 1455.66 mgl^{-1} in the effluent from thermal power plant and 3230.90 mgl^{-1} in effluent collected from battery manufacturing industry and the average value of TDS in the effluent from petrochemical, chemical industries and control were obtained to be 2785.2, 2893.01 and 293.4 mgl^{-1} respectively. According to Indian Council of Medical Research (ICMR), (1975) water on the basis of its TDS was classified as, TDS$\leq$ 500mgl^{-1}, suitable for drinking purposes, ≤1000 mgl^{-1} pemissible for drinking water, ≤ 2000 mgl^{-1} used for irrigation, above 3000 mgl^{-1} not used of irrigation. On the basis of classification, it was observed that other than effluent from battery manufacturing industry not safe for irrigation purpose. Plants weaken in high TDS, so irrigation with high TDS water will decrease crop production.

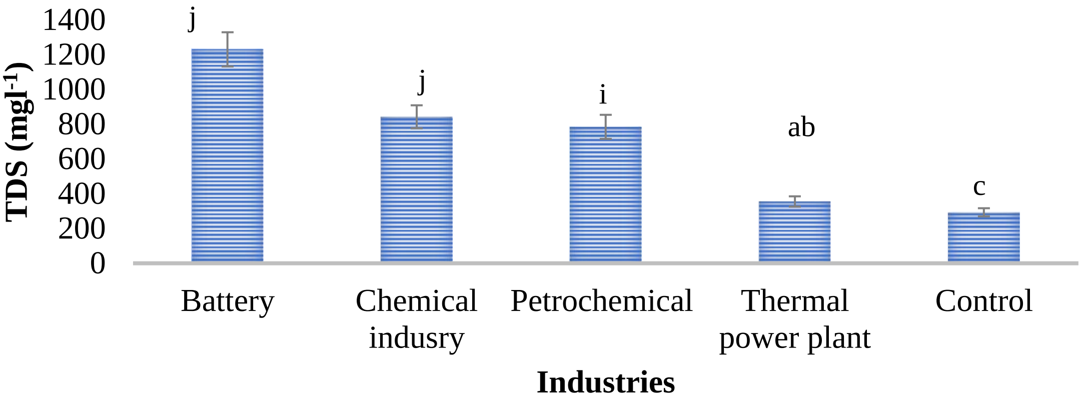


Fig. 4.1.4: TDS analysed in industries as well as control (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

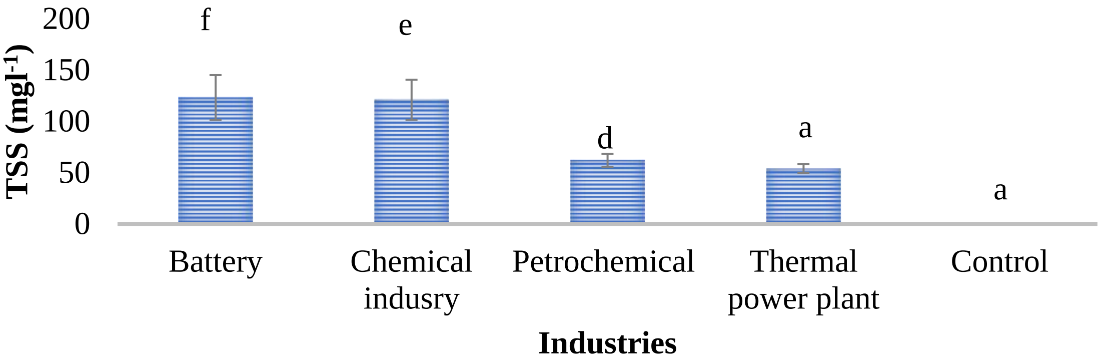


Fig.4.1.5: TSS found in industries as well as control (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Presence of calcium and magnesium cations contribute to water hardness (Chaudhary et al., 2005; Acharya et al., 2008). Total hardness of effluents was found in the range of 266.31 to 458.07 mgl^{-1} in which chemical industry effluent has maximum hardness followed by petrochemical, flashlight and battery manufacturing and thermal power plants. Irrigation with high hardness effluent will adversely affect soil, convert into alkaline and decrease productivity.

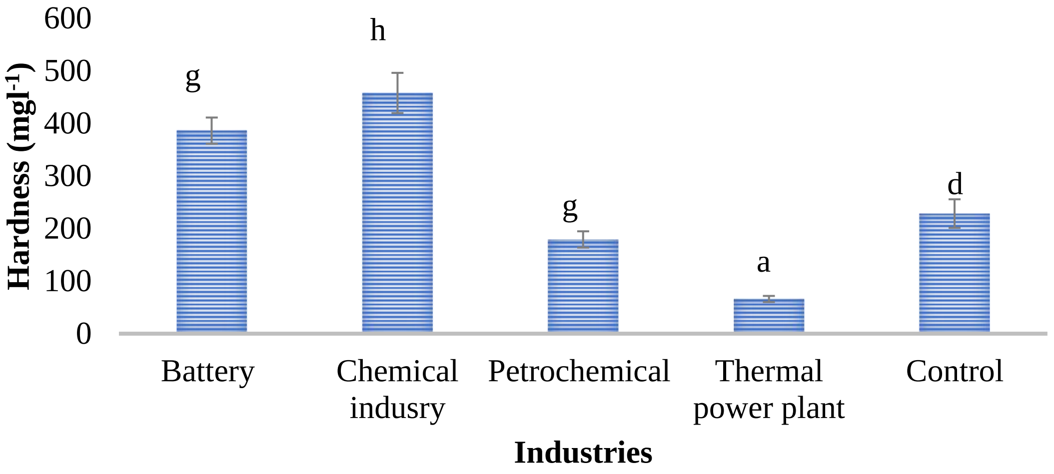


Fig. 4.1.6: Hardness analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Alkalinity is measure strength of water which shows the capacity of effluent to neutralize acid. It is the quantity of negative ions i.e. CO_3^-, HCO_3^-, OH^- etc. It's also estimate the ability of effluent to resist changes in the pH upon additional acids. Alkalinity of the effluent in battery manufacturing, petrochemical, chemical industries and thermal power plant was found to be 441.69, 276.5, 351.24, 159.33 mgl^{-1} respectively. Water with higher alkalinity is corrosive and cannot used for many purpose.

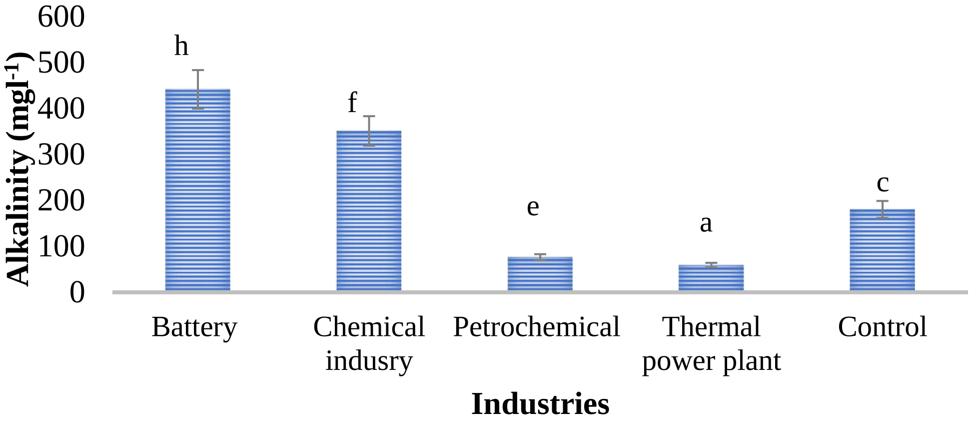


Fig. 4.1.7: Alkalinity analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Sulphates usually occurs naturally and cause water hardening and, therefore, higher levels are not suggested. It's may undergo transformations to H_2S depending largely upon the redox potential of water. It's was found to be 5.30, 13.32, 22.30, 6.70 and 2.1 mgl^{-1} respectively in the effluent of flashlight and battery manufacturing, petrochemical, chemical industries, panki thermal power plant and control.

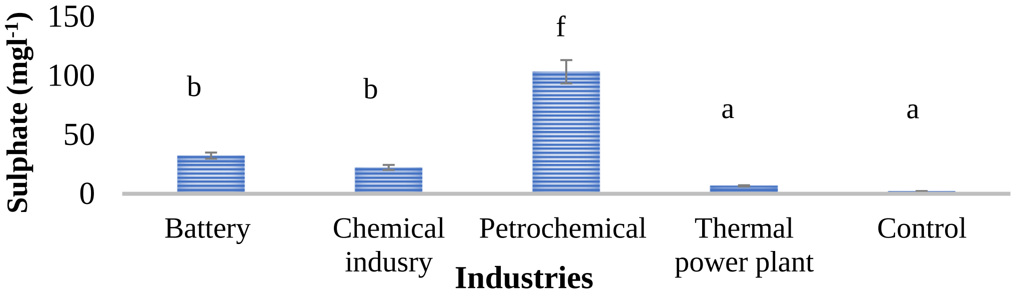


Fig. 4.1.8: Sulphate analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values).

Chloride is natural content of water with widely varying concentration and excessive concentration is based on deliciousness and its potentially corrosiveness. If the chloride is > 250 mgl^{-1} (WHO limit), the water is salty and people who are not used to high chlorides may be subject to laxative effects. Chloride is necessary for aquatic life, but huge concentration negatively affects reproduction rate, species mortality, changing characteristics of ecosystem. Chloride in effluent of flashlight and battery manufacturing, petrochemical, chemical industries, Panki thermal power plant and control was found to be 696.37, 221, 420.4, 120.9 and 32.8 mgl^{-1} respectively. Results indicates maximum chloride contents of 696.37 and 420.4 mgl ^{1} collected from battery manufacturing and chemical industry, reason may due to use chlorine compounds, such as acids (Hydrochloric, Hypochloric acid, $AlCl_2$) and chlorine gas are used as raw materials in various processes in manufacturing industries.

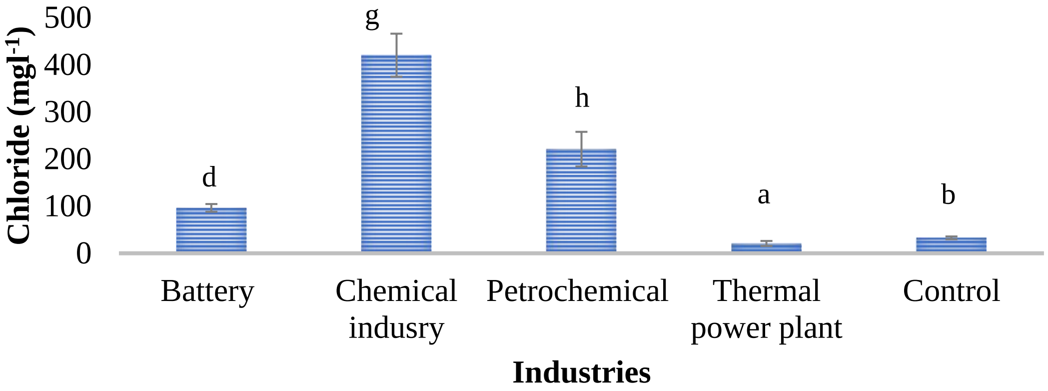


Fig. 4.1.9: Chloride analysed in industries effluent as well as control (tap water). **(**One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values).

Dissolved oxygen (DO) in water indicate that's water to support biological life. Dissolved oxygen levels are found to be very low which also shows excess organic matter concentration. More than 4 mgl^{-1} is desirable (WHO, 2002) but all the samples show low amount of DO. DO was found to be 1.38, 1.48, 1.62, 2.03 and 6.67 mgl^{-1} in flashlight manufacturing, petrochemical, chemical, panki thermal power plant and control respectively. All industries show low value of DO may be due to increased metabolic activity of microorganisms with high temperature of water which consumes a lot of oxygen for decomposition of organic matters.

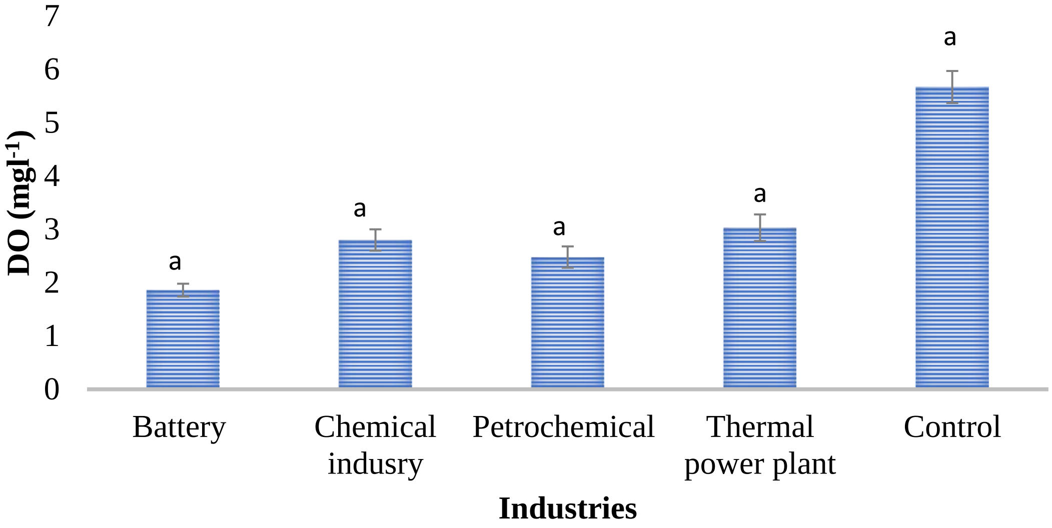


Fig. 4.1.10: Dissolve oxygen analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at $p<0.05$. Values followed by different alphabets are significantly differences between values**).**

Biological oxygen demand (BOD) is a measure of the quantity of oxygen used by bacteria for oxidation of organic matter and one of the important characteristics of measuring organic matter pollutant present in water. BOD in the effluent of flashlight manufacturing, petrochemical, chemical, panki thermal power plant and control was found to be 263.61, 135.5, 183.02, 67.30 and 4.06 mgl^{-1} respectively which is higher than guide line limit of WHO i.e. 30 mgl^{-1}. Higher BOD also means low oxygen available for microorganism in water bodies.

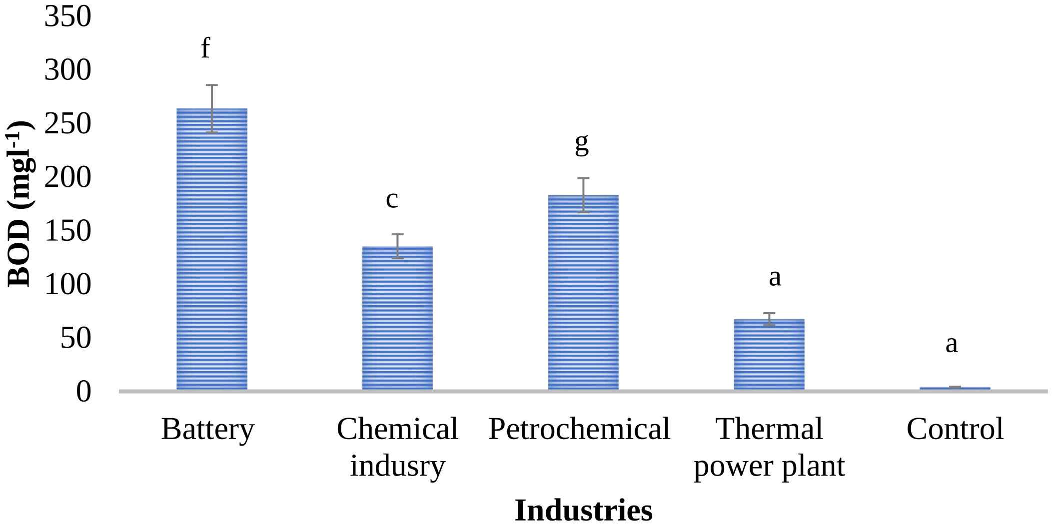


Fig. 4.1.11: BOD analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Chemical oxygen demand (COD) an important, parameter for industrial wastewater studies, it's a quantity of dissolved oxidizable organic matter including biodegradable and non-biodegradable matter present in wastewater. COD of effluent was recorded as 654.44, 542.2, 849.06, 284.42 and 25.97 mgl^{-1} respectively in all four industries. According to WHO (2002) prescribed limit for COD is 250 mgl^{-1}) observed COD value in all effluent is much higher than permissible level. High COD levels show toxic condition and the presence of biologically resistant organic substances (Poddar et. al., 2017).

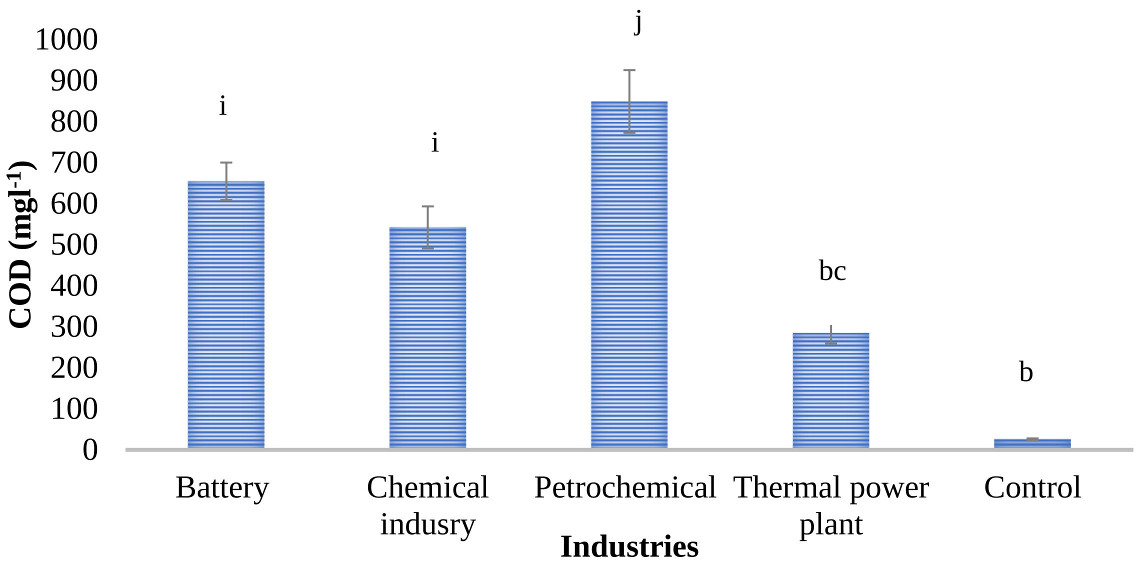


Fig. 4.1.12: COD analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Dispersed oil and grease is an important parameter for wastewater quality and safety. Oil and grease (O&G) forms a film on the surface of water and coat plant and animal which causes injuries to aquatic microorganism's animal, plant, and correspondingly mutagenic and carcinogenic for human being (Lan et. al. 2009; Islam et al. 2013; Abd El-Gawad et al., 2014). O&G was found to be in the range of 3.5-16.5 mgl^{-1} in industries while in control it is below detectable level. Petrochemical and chemical industry shows significant level of O&G contribution. Discharge effluent contaminated with oil and grease forms layer on water surface which decreases DO, reduce biological activity because oil film was form around the microbes in suspended matter of water. Then oxygen molecules are difficulty to be oxidative for microbial on hydrocarbon molecules and cause ecology damages to water bodies (Alade et al., 2011; Facchin et. al., 2013). Moreover, petroleum or grease spilled over water also produces chemicals that are extremely harmful for marine animals.

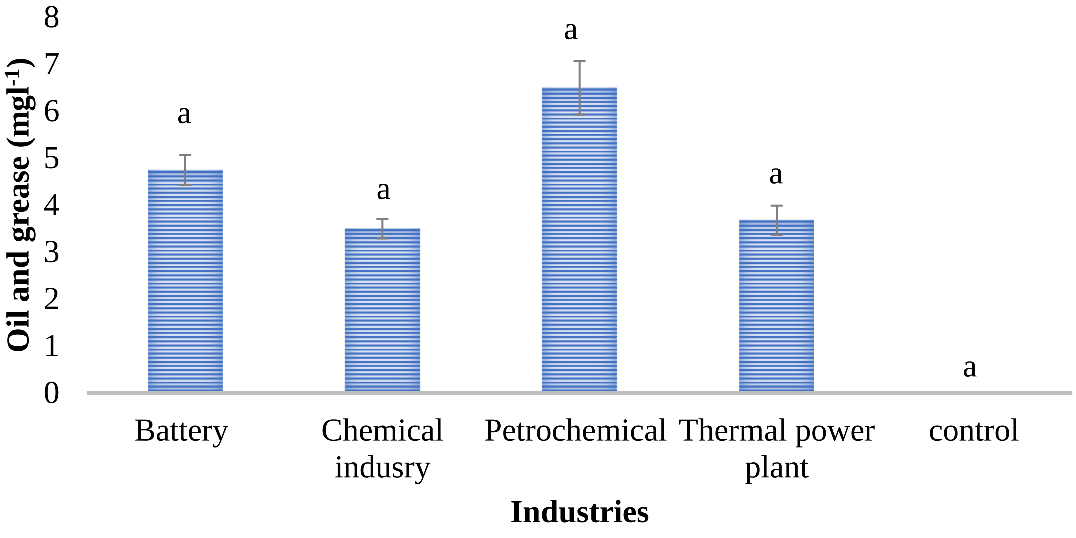


Fig. 4.1.13: Oil and grease analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at p<0.05. Values followed by different alphabets are significantly differences between values**).**

Nitrate is the most common form of nitrogen present in nature, produce after decomposition of organic matter (Rai, 2010; Agca et al., 2014). Nitrate is reasons of eutrophication which affects aesthetics on fresh water lakes, rivers and results in odour and appearance problems. It is also known factor for algae growth, can be toxic to animals such as cattle, affects taste of water, plug filtration units and raise the costs of water treatment. Nitrate concentration in effluents was reported 0.78, 0.13, 4.06, 0.95 and 7 mgl^{-1} in flashlight manufacturing, petrochemical, and chemical industry, panki thermal power plant and control respectively which is very low.

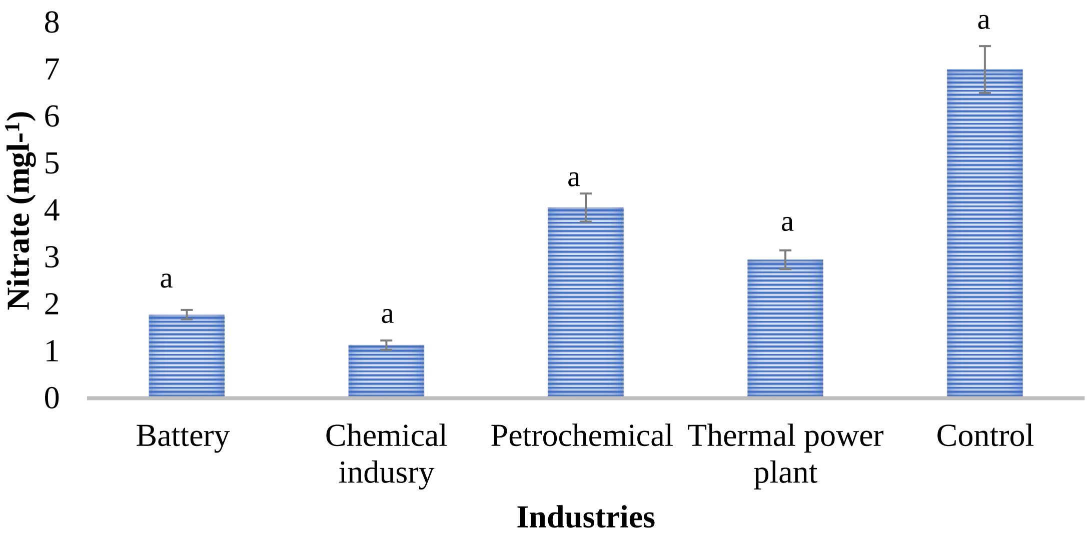


Fig. 4.1.14: Nitrate analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values**).**

Phosphate is essential element for growth of aquatic organism but serious problem is arising when excess phosphate in treated effluent is discharged to aquatic bodies. Phosphate varies in the range of 1.23 mgl^{-1} in effluent of thermal power plant to 6.7 mgl^{-1} effluent from chemical industry. In the battery manufacturing industry, it was reported to be 3.13 mgl^{-1} and 2.45 mgl $^{-1}$ in effluent of petrochemical industry. Synthetic detergent is the main source of phosphate, detergents create frothing, and can harm invertebrates and fish.

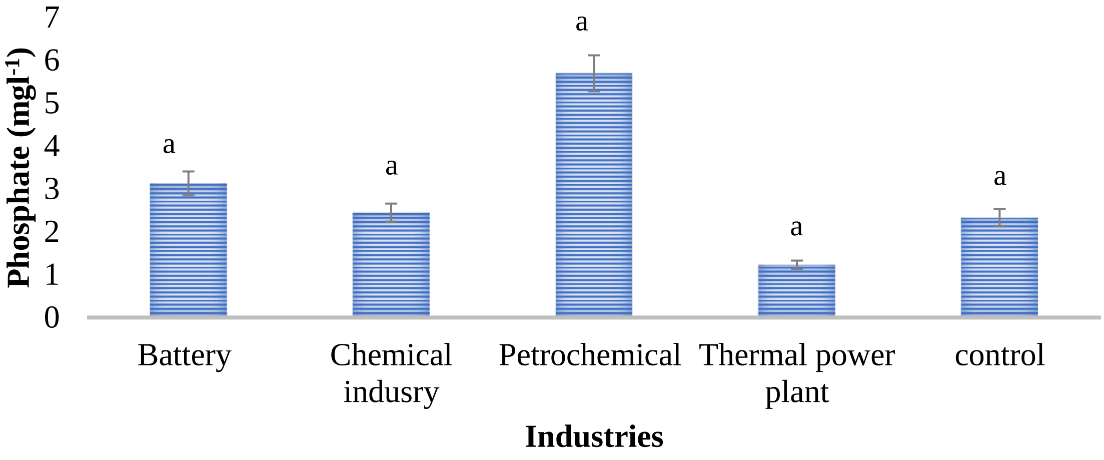


Fig 4.1.15: Phosphate analyzed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values)**.**

Sodium is natural cation found in all fresh water, but excess concentration could be credited to by sodium salts and soaps is identified as a risk factor for high blood pressure. According to Tekade et al. (2011) 20 mgl^{-1} sodium suggested as safe. Effluent with higher Na concentration is not suitable for irrigation purpose and deteriorate the soil fertility. In the effluent of battery manufacturing, chemical, petrochemical industries, thermal power plant and control sodium was found to be 53.42, 48.11, 25.12, 2 and 0.01 mgl^{-1} respectively.

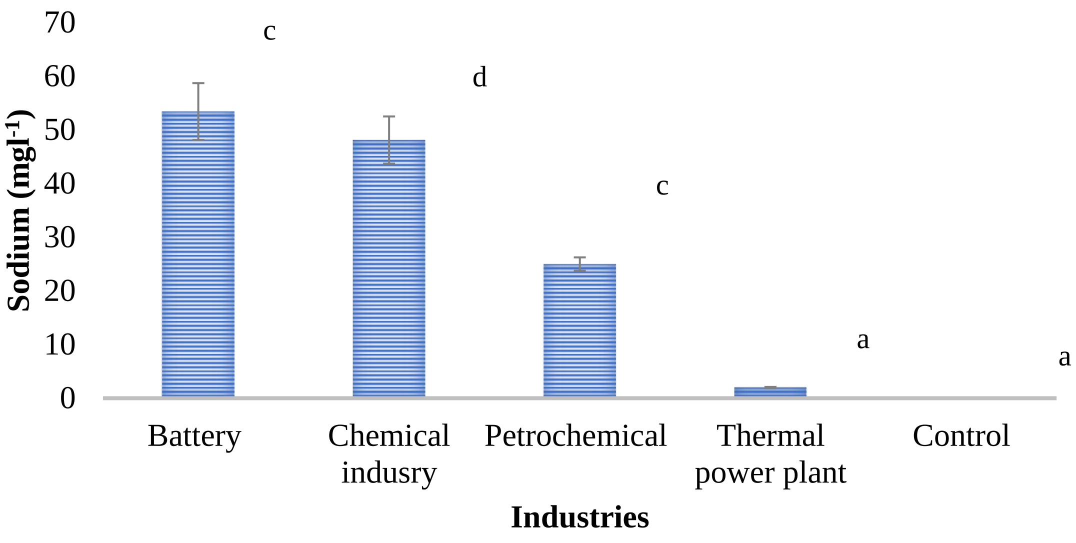


Fig. 4.1.16: Sodium analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at $p<0.05$. Values followed by different alphabets are significantly differences between values).

Potassium is water soluble and remains in solution without undergoing any precipitation. Similarly, it is not very much important from the health point of view but in large quantities may be laxative. Potassium in the effluent of battery manufacturing, chemical, petrochemical industries, thermal power plant and control was found to be 10.78, 3.53, 17.6, 2.1 and 2 mgl^{-1} respectively. The concentration of potassium in wastewater is very low but high value being an indication of pollution.

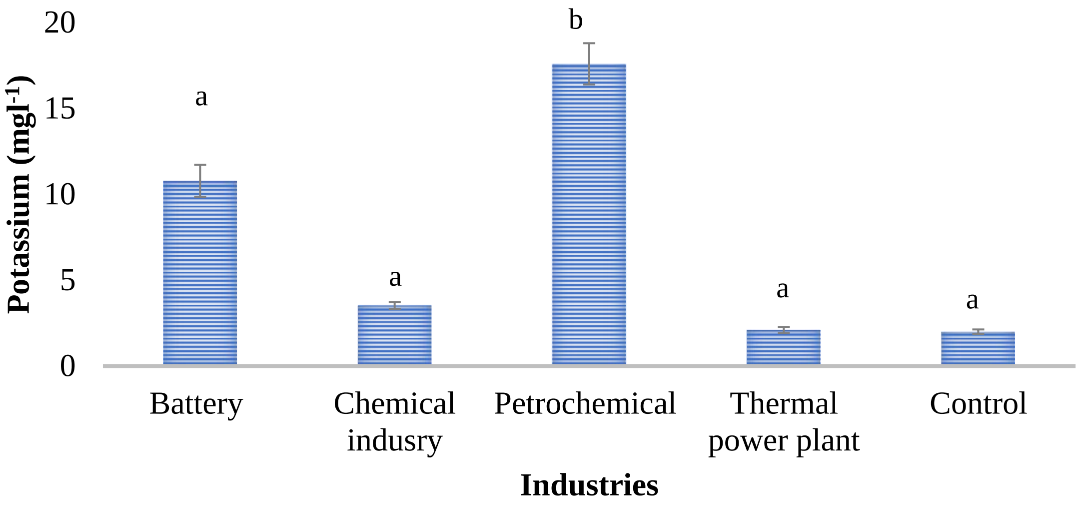


Fig. 4.1.17: Potassium analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values).

Calcium is a natural component, used as a protective coating inside steel pipes and boilers. In cell its activates several enzymes, and initiates chemotropic responses in pollen, large amounts of Ca also applied in agricultural as fertilizers. Calcium was found in the range of 1 mgl^{-1} in thermal power plant to 33.46 mgl^{-1} in chemical industry. In the effluent of battery manufacturing industry, it was found to be 28.24 and in petrochemical effluent it was 17.23 mgl^{-1}.

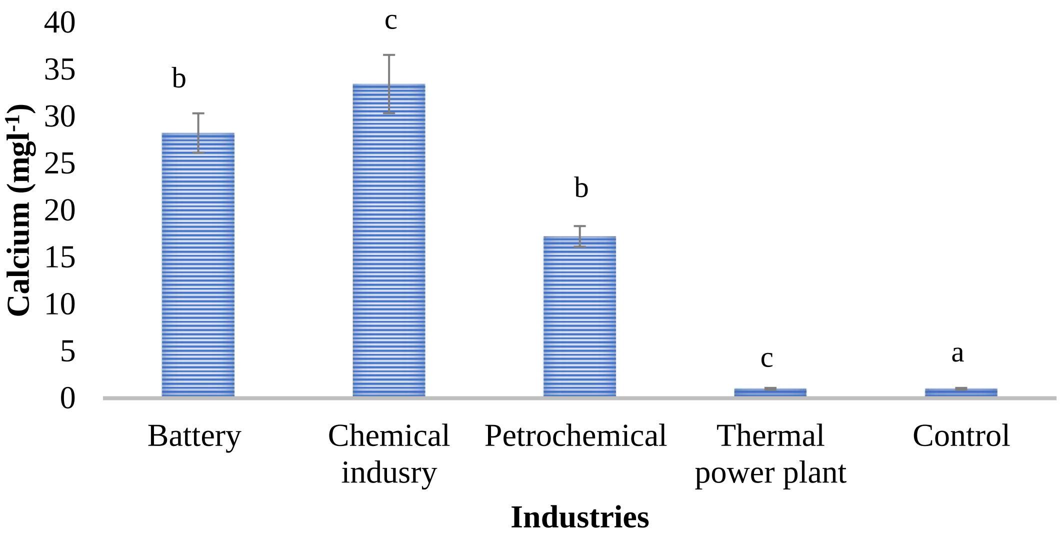


Fig. 4.1.18: Calcium analysed in industries effluent as well as control (tap water) (One-way ANOVA was performed to compare the means of different treatments at *p*<0.05. Values followed by different alphabets are significantly differences between values).

Heavy metals

Heavy metal i.e. Hg, Fe, Mn, Zn, Cu, Pb, Ni, Cr, and Cd analysis of various wastewater samples from four different industries at Uttar Parades are reported. Concentration of Hg is very higher i.e. 2.65 mgl^{-1} in the battery manufacturing industry for the making of mercury oxide batteries which is useful in the application requiring high energy. In thermal power plants it is below detectable level (BDL) but in the effluent of chemical and petrochemical industry it was found to be 0.03 and 0.39 mgl^{-1} respectively, which is also higher than permissible limit of CPCB i.e. 0.01 mgl^{-1}. This is non-essential element for human as well as plant and causing neurological disorder humans.

Fe was recorded in effluent of battery manufacturing, chemical, petrochemical industries and thermal power plants as 2.59, 0.92, 8.82 and 0.65 mgl^{-1} respectively.

According to result petrochemical industry found maximum concentration of Fe which is higher than permissible limits of CPCB i.e. 3mgl^{-1}. In comparison with CPCB standard for Fe, other industry was in normal range and not exceeded. Iron is not considered to be hazardous to health, in drinking water it causes metallic taste and an offensive odour and pipes become clogged if Fe in higher concentration (Ramola and Singh, 2013).

Mn is essential metal for plants because they involved in photosynthesis and enzyme activity (Bonanno and Giudice, 2010). It was found to be in the range of 0.02 mgl^{-1} in thermal power plant to 1.94 mgl^{-1} in battery manufacturing industry. Permissible limit of Mn is 2 mgl^{-1}, so all industries found under limit.

Concentration of zinc in effluent of battery manufacturing, chemical 1.75, 2.86, 13.13 and 0.79 mgl^{-1}. The permissible limit of zinc in water according to WHO standards is 5mgl $^{-1}$. according to the CPCB standards petrochemical effluent found beyond the permissible limit. Zinc is important trace elements that play a vital role in the physiological and metabolic process of many organisms. Nevertheless, higher concentrations of zinc can be toxic to the organism.

Cu is maximum found in chemical industry effluent i.e. 3.12 mgl^{-1} which is slightly higher than prescribed limit (3 mgl^{-1}) by CPCB. Others industrial effluent found in the range of 0.89 to 0.02 mgl $^{-1}$ that's under permissible level. Highly presence of Cu in the water is accredited to corrosion of alloys in the pipe fittings, (Callatay and Francois, 2005).

Pb recorded in battery manufacturing and petrochemical industries i.e. 2.82 and 0.36 mgl^{-1} respectively, in other Pb below detectable level. Lead is other important metal in a battery effluent because it is a major raw material in the manufacture of lead acid batteries. Lead at very low concentration is toxic and hazardous to most forms of life (permissible limit 0.1 mgl^{-1}). Lead has a negative influence on both children and adults. In children, Pb reduces the physical growth and mental growth. The intelligent quotient of children is diminished, and symptoms of irritability and fatigue could be observed. Pregnant women exposed to Pb have higher rates of infertility, miscarriage and still births (Ediin et al., 2000). The chronic effect of Pb on man includes neurological disorders, especially in the fetus and in children. This can lead to behavioral changes and impaired performance in IQ tests (Singh et al., 2011).

Ni was found to be 0.32 mgl^{-1} in petrochemical industry and in other industries it is below detectable level. CPCB standard for Ni is 3 mgl^{-1} which means it concentration is acceptable in petrochemical industry.

Cr was found in effluent of battery manufacturing, chemical, petrochemical industry and thermal power plant to be 0.12, 0.01, 0.43 and 0.01 mgl^{-1} respectively. According to the result petrochemical and battery manufacturing industries found higher than discharge standard of CPCB i.e. 0.1 mgl^{-1}. Chromium is important for human and animal diet but excess it will be carcinogenic (Venugopal and Mohanty, 2011). It accumulates in skin, lungs, fat muscles, nails, hair and placenta where it is traceable to various health condition (Adeleken and Abegunde, 2011). Its exposure can cause lung cancer, tumor, dermatitis and ulcers (Sarkar, 2005).

Cd was found only in battery manufacturing and chemical effluent which was recorded as 2.18 and 0.55 mgl^{-1}. higher exposure of Cd can cause renal disfunction, metabolism disorder and cancer and low concentration adverse changes in arteries of kidney, blood pressure, kidney (Alloway et al., 2005; Mehbrahtu and Zerabruk, 2011). In plants, its inactivates some enzymes and induces oxidative stress (Wieczorek et al., 2005). The maximum contaminant level of Cd allowed in water by CPCB is 2 mgl^{-1}

according to this standard effluent of battery industry found risky to irrigation purposes (Monudu and Anyakora, 2010).

Table 4.1.1: Heavy metal concentration (mgl^{-1}) in different industrial wastewater and control water

Heavy metals	Battery	Chemical	Petrochemical	Thermal power plant	Control	CPCB standard
Hg	2.65 ± 0.43^{f}	0.03 ± 0.002^{a}	0.39 ± 0.021^{a}	BDL	0.0015 ± 0.0001^{a}	0.01
Fe	2.59 ± 0.23^{f}	0.921 ± 0.08^{d}	8.82 ± 0.78^{b}	0.65 ± 0.03^{b}	0.001 ± 0.0001^{a}	3
Mn	1.94 ± 0.13^{d}	0.76 ± 0.02^{c}	0.23 ± 0.02^{a}	0.022 ± 0.008^{a}	0.002 ± 0.0001^{a}	2
Zn	1.75 ± 0.11^{c}	2.86 ± 0.059^{e}	13.13 ± 0.98^{c}	0.79 ± 0.096^{c}	0.004 ± 0.0002^{a}	5
Cu	0.68 ± 0.03^{b}	3.12 ± 0.003^{f}	0.89 ± 0.073^{a}	0.024 ± 0.007^{a}	0.001 ± 0.0001^{a}	3
Pb	2.82 ± 0.01^{f}	BDL	0.36 ± 0.023^{a}	BDL	BDL	0.1
Ni	BDL	BDL	0.32 ± 0.021^{a}	BDL	0.03 ± 0.0001^{a}	3
Cr	0.12 ± 0.003^{a}	0.018 ± 0.005^{a}	0.43 ± 0.027^{a}	0.014 ± 0.005^{a}	0.006 ± 0.0003^{a}	0.1
Cd	2.18 ± 0.02^{e}	0.55 ± 0.022^{b}	BDL	BDL	BDL	2

- All values reported in mgl^{-1}. Data analysed by one-way ANOVA at $p<0.05$. All the values are means of three replicates (n=3) ± S.D. Values followed by different alphabets are significantly differences between values

Physico-chemical study of industrial wastewater showed occurrence of heavy metals and other inorganic pollutants like TSS, TDS, chloride, oil and grease, cause of concern for the public health. Present study concluded that existing situation if mishandled can cause irreparable ecological and environmental harm in the long-term well masked by short term economic prosperity. Therefore, the water supplied by the all industries should be treated prior to supply, for optimal removal of hazardous pollutants, so that public health is protected.

Chapter 4.2

Evaluation of phytotoxicity of different industrial wastewater through seed germination test on different varieties of crop seeds

Evaluation of phytotoxicity of different industrial wastewater through seed germination test on different varieties of crop seeds.

The industries of chemical, petrochemical, battery manufacturing and thermal power plants is significant for economy of Uttar Pradesh, India, but produce large amount of wastewater which can dispose of either through discharge into surface water or by spreading on land. This effluent characterized by dark color, high pH, TDS, TSS, oil grease and heavy metals (Mekki et al., 2013). All these characteristics have potential to reduce growth of microorganism, seed germination and cause other adverse environmental problem. The main problem regarding to disposal of wastewater, is to find an environment friendly and economically feasible disposal method, because direct disposal is not good for aquatic water bodies.

Various researchers also have reported potential phytotoxic effects on various crop seeds under certain conditions (Hanifi and El-Hadrami, 2007; Saadi et al., 2013; Madhvi et al., 2014a). A significant degree of this phytotoxicity is attributed to the higher heavy metals and other inorganic content present in effluent (Lanciotti et al., 2005). The application of these effluent can be toxic to crop and inhibits the seed germination. Therefore, objective of this study to evaluate the phytotoxicity of different industrial wastewater through seed germination on rice, pulse and oil seeds.

The radicle length of crop seeds i.e. rice, pulses and oil seeds show different results in different industrial effluents presented Table 4.2.1. Radicle length of Basmati-198 rice was found to be 2.45, 2.40, 2.32 and 2.39 cm at 0% (control), 2.06, 1.69, 1.92 cm and 1.43 at 50% effluent and 1.43, 1.20, 1.54 and 1.43 at 100% concentration, In Pusa Sugandh it was found to be 2.17, 2.19, 2.25 and 2.16 cm; 1.70, 1.46, 1.94 and 1.63 cm; and 1.20, 0.87, 1.46, 0.92 cm at 0, 50 and 100% concentrated effluent respectively and in third variety of rice i.e. sambhamasuri it was found to be 2.22, 2.13, 2.23 and 1.91 cm; 1.50, 1.64, 1.94 and 1.63 cm; and 1.06, 0.95, 1.49, 1.04 cm at 0, 50 and 100% concentration respectively. Radicle length was highest in basmati seeds as compare to two other varieties of rice except in battery manufacturing industry effluent. In the case of battery manufacturing effluent Sambha masuri show high radicle length in absolute effluent but in half dilution, Pusa sugandh shows maximum radicle length. In comparison with 0% (control) water radicle length of seed grown in 50% and 100% wastewater was least which mean pollutants water decrease the germination.

In the pulses, radicle length of chick pea was found to be 2.49, 2.37, 2.28 and 2.53 cm at 0%, 1.83, 1.71, 1.82 and 1.84 cm at 50%, and 0.86, 1.31, 1.52 and 1.25 cm at 100% respectively. In Moong N-1 length was found to be 2.32, 2.36, 2.24 and 2.38 cm at 0%, 1.89, 1.81, 1.92 and 1.74 cm at 50% and 1.20, 1.20, 1.65 and 1.07 cm at 100% concentration and other variety of pulse i.e. Massor VL-1 was found to be 2.05, 1.40, 1.33 and 1.93cm at 0%, 1.63, 1.15, 1.12 and 1.45 cm at 50% and 1.08, 0.94, 0.89, 1.11 cm at 100% concentration respectively. In the case pulses, moong shows maximum radicle length in both effluent concentration as well as in control in all industries.

Radicle length of oil seed of Mustard BR-40 was found to be 2.27, 1.84, 1.91 and 2.25 cm at 0%, 1.83, 1.56, 1.75 and 1.91 cm at 50% and 1.23, 1.17, 1.46 and 1.33 cm at 100%, in Alsi KL-43 radicle length was found to be 2.18, 1.86, 2.17 and 2.18 cm at 0%, 1.71, 1.68, 1.75 and 1.76 cm at 50% and at 100% it was 1.22, 1.35, 1.24 and 1.13 cm, respectively. In Sesame G. til-3 radicle was found to be 1.53, 1.27, 1.34 and 1.45 cm at 0%, 1.18, 1.15, 1.13 and 1.07 cm at 50% and 0.90, 0.86, 0.92 and 0.84 cm at 100% concentrated effluent respectively. Comparison of radicle length in oil seeds, mustard seeds shows maximum length in all effluent concentration except in chemical industry effluent. In chemical industry Alsi has found higher length in both dilution as well as in control.

The effect of industrial effluents on radicle growth depends on the nature of toxic substances, which alter the seed water interaction necessary for triggering enzyme activity (Baruah and Das, 1998; Malla and Mohanty, 2005). It is commonly believed that initial effects of effluents occur at biochemical level, which finally results in delayed germination and retardation in seedling growth spontaneously. When the concentration of toxicants in the effluents exceeds the detoxifying capacity of the tissue through their normal metabolism, there is decrease in the biochemical parameters (Chlorophyll, protein, amino acids, sugar, nucleic acids), which are directly proportional to the concentration of effluents.

The effects of effluent on radicle length and in germination depends on effluent concentration and exposure time. Maximum length was observed in control seed which are moisten by tap water. In the case of effluent 50% concentration shows better performance as compare to 100% concentration. We can say that if industrial effluent can be diluted with water it performs better and also used in agricultural field for irrigation.

Table 4.2.1: Radicle length of seed germinated in different industrial effluent with different concentration.

Test Material		**Growth media**	**Length of radical (cm) ±SD**			
			Petrochemical	**Chemical**	**Thermal power plant**	**Battery**
Rice	Basmati-198	Control	2.45±0.08	2.40±0.02	2.32±0.04	2.39±0.03
		Effluent	1.43±0.07	1.20±0.02	1.54±0.02	0.87±0.02
		Effluent + tap water (1:1)	2.06±0.6	1.69±0.04	1.92±0.04	1.43±0.03
	Pusa sugandh	Control	2.17±0.2	2.19±0.06	2.25±0.02	2.16±0.05
		Effluent	1.20±0.03	0.87±0.01	1.46±0.03	0.92±0.01
		Effluent + tap water (1:1)	1.70±0.02	1.46±0.02	1.94±0.03	1.63±0.02
	Sambha masuri	Control	2.22±0.03	2.13±0.02	2.23±0.03	1.91±0.02
		Effluent	1.06±0.05	0.95±0.02	1.49±0.06	1.04±0.05
		Effluent + tap water (1:1)	1.50±0.05	1.64±0.02	1.81±0.02	1.37±0.01
Pulse	Chick pea	Control	2.49±0.05	2.37±0.02	2.28±0.02	2.53±0.03
		Effluent	0.86±0.03	1.31±0.03	1.52±0.02	1.25±0.02
		Effluent + tap water (1:1)	1.83±0.05	1.71±0.03	1.82±0.05	1.84±0.02
	Moong N-1	Control	2.32±0.04	2.36±0.02	2.24±0.02	2.38±0.03

		Effluent	1.20±0.03	1.20±0.07	1.65±0.03	1.07±0.05
		Effluent + tap water (1:1)	1.89±0.05	1.81±0.05	1.92±0.03	1.74±0.02
	Massor VL-1	Control	2.05±0.05	1.40±0.03	1.33±0.02	1.93±0.02
		Effluent	1.08±0.06	0.94±0.02	0.89±0.03	1.11±0.02
		Effluent + tap water (1:1)	1.63±0.03	1.15±0.02	1.12±0.01	1.45±0.03
	Mustard BR-40	Control	2.27±0.05	1.84±0.01	1.91±0.08	2.25±0.02
		Effluent	1.23±0.05	1.17±0.05	1.46±0.02	1.33±0.02
		Effluent + tap water (1:1)	1.83±0.01	1.56±0.02	1.75±0.02	1.91±0.03
	Alsi KL-43	Control	2.18±0.03	1.86±0.03	2.17±0.03	2.18±0.07
Oil seed		Effluent	1.22±0.03	1.35±0.01	1.24±0.02	1.13±0.08
		Effluent + tap water (1:1)	1.71±0.03	1.68±0.04	1.75±0.05	1.76±0.02
	Sesame G Til-3	Control	1.53±0.01	1.27±0.04	1.34±0.03	1.45±0.03
		Effluent	0.9±0.02	0.86±0.03	0.92±0.05	0.84±0.01
		Effluent + tap water (1:1)	1.18±0.02	1.15±0.02	1.13±0.02	1.07±0.05

Effluent tolerance index (ETI)

Effluent tolerance index in rice its varies between 0.36 to 0.66, 0.40 to 0.65, 0.45 to 0.67 and 0.60 to 0.88, 0.67 to 0.86, 0.68 to 0.85 in Basmati, Pusa sugandh, and Sambha masuri respectively, in pulses its varies between 0.35 to 0.67, 0.45 to 0.73, 0.53 to 0.63 and 0.72 to 0.80, 0.73 to 0.85, 0.75 to 0.84 in Chick pea, Moong and Massor respectively and in oil seed its recorded in the range of 0.54 to 0.76, 0.52 to 0.73, 0.58 to 0.59 and 0.81 to 0.91, 0.78 to 0.90, 0.77 to 0.91 in Mustard, Alsi and Sesame at 100 and 50% concentrated industrial effluent respectively.

The order of toxicity tolerance in different crop seeds varies depending upon the nature of effluent. Result conclude that rice is the susceptible crop and oilseeds are more tolerant crops towards toxicity of effluents. Among the crops seeds, the overall results revealed the following sensitivity in decreasing order i.e. Rice>Pulse >Oil seeds, in both effluent concentration. Oil seeds and pulse shows higher tolerance as compare to rice. Huma et al. (2012) determine the effect of pharmaceutical industrial wastewater on the germination of mustard, rapeseeds, coriander, fennel, fenugreek and barley. Result shows that fenugreek and barley are metal tolerant plants and they can maintain their germination at concentration of wastewater. Mosse et al. (2010) studied the seed germination and vegetative growth of common crop seeds Barley, Lucerne, Phalaris and Millet in whiney wastewater and result shows that wastewater increases the germination time and restrict early growth. They suggested that barley seeds are potential useful tool for winery wastewater. Rehman et al. (2009) evaluated the effect of textile effluent on early growth and seed germination of some winter vegetable i.e. Radish, Turnip and Mustard. Effluent reduce the growth and germination in all the seeds and turnip were observed as susceptible and radish was tolerant species.

Table 4.2.2: Effluent tolerance index (ETI) of seed exposed to different industrial effluent.

Test Material		Growth media	ETI			
			Petrochemical	Chemical	Thermal power plant	Battery
Rice	Basmati-198	Control	1.00	1.00	1.00	1.00
		Effluent	0.58	0.50	0.66	0.36
		Effluent + tap water (1:1)	0.88	0.67	0.78	0.60
	Pusa sugandh	Control	1.00	1.00	1.00	1.00
		Effluent	0.55	0.40	0.65	0.42
		Effluent + tap water (1:1)	0.78	0.67	0.86	0.76
	Sambha masuri	Control	1.00	1.00	1.00	1.00
		Effluent	0.48	0.45	0.67	0.54
		Effluent + tap water (1:1)	0.68	0.77	0.85	0.71
Pulse	Chick pea	Control	1.00	1.00	1.00	1.00
		Effluent	0.35	0.55	0.67	0.49
		Effluent + tap water (1:1)	0.73	0.72	0.80	0.73
	Moong N-1	Control	1.00	1.00	1.00	1.00
		Effluent	0.52	0.51	0.73	0.45
		Effluent + tap water (1:1)	0.81	0.77	0.85	0.73
	Massor VL-1	Control	1.00	1.00	1.00	1.00
		Effluent	0.53	0.67	0.67	0.57
		Effluent + tap water (1:1)	0.79	0.82	0.84	0.75
Oil seed	Mustard BR-40	Control	1.00	1.00	1.00	1.00
		Effluent	0.54	0.64	0.76	0.59
		Effluent + tap water (1:1)	0.81	0.85	0.91	0.85
	Alsi KL-43	Control	1.00	1.00	1.00	1.00
		Effluent	0.56	0.73	0.57	0.52
		Effluent + tap water (1:1)	0.78	0.90	0.81	0.80
	Sesame G Til-3	Control	1.00	1.00	1.00	1.00
		Effluent	0.59	0.68	0.69	0.58
		Effluent + tap water (1:1)	0.77	0.91	0.84	0.74

Phytotoxicity in all variety of rice, pulse and oil seed is shown by different types of effluents presented in Table 4.2.3. In Basmati-198 effluent from battery industry cause maximum toxicity i.e. 63.55% followed by chemical, petrochemical and thermal power plant i.e. 49.72, 41.71 and 33.57% respectively. In both Pusa sugandh and

Sambha masuri, absolute effluent chemical industry causes maximum toxicity i.e. 60.03 and 55.47% respectively. In 50% diluted effluent concentration battery, chemical and petrochemical effluent found toxic to Basmati-198 (39.8%), Pusa sugandh (33.28%) and Sambha masuri (32.38%) respectively. Effluent of thermal power plant is least toxic to all variety of rice in comparison.

In pulses, chick pea shows maximum phytotoxicity against petrochemical effluent i.e. 65.33% followed by battery, chemical and thermal power plant effluent i.e. 50.59, 44.59 and 33.23%, Moong N-1 maximally shows toxicity i.e. 54.90% through battery effluent followed by chemical, petrochemical and thermal power plants 49.01, 48.28 and 26.56% and in Massor VL-1 petrochemical effluent shows maximum toxicity which is 47.4 % followed by battery, chemical and thermal power plant 42.69, 33.08 and 32.78% at 100% concentration. At 50% concentration of effluent phytotoxicity range varies from 14.54 to 27.14%. battery effluent shows maximum toxicity for Chick pea, Moong and Massor respectively.

At 100% concentration of effluent shows maximum toxicity in Mustard BR-40 by petrochemical effluent i.e. 45.59% followed by, battery, chemical and thermal power plant respectively. In Alsi KL-43 and Sesame G til-3 battery effluent cause higher toxicity as compare to others. Same in the case of 50% concentration petrochemical and battery manufacturing industry effluent shows higher toxicity in mustard (18.97%), Alsi (21.56%) and Sesame (26.21%) respectively. The trends of phytotoxicity of different indusrial effluent and as follows-

- **Petrochemical industry effluent:** Chick pea>Sambha masuri> Moong> Massor> Mustard> Pusa Sugandh>Alsi>Basmati>Sesame.
- **Chemical industry effluent:** Pusa Sugandh> Sambha masuri>Basmati>Moong>Chick pea>Mustard>Massor> Sesame>Alsi.

- **Thermal power plant effluent:** Alsi> Pusa Sugandh>Basmati>Chick pea>Sambha masuri>Massor>Sesame>Mustard.
- **Battery manufacturing industry effluent**: Basmati>Pusa Sugandh>Moong>Chickpea>Alsi>Sambhamasuri>Massor>Sesame>Mustrad.

Different crop varieties show different sensitivities to the toxic nature of effluents. Oil species shows minimum toxicity whereas rice species shows maximum toxicity. The degree of phytotoxicity of industrial effluents depends not only on the chemical composition of the effluents but also on the type of crops cultivated. Nature of the different types of crop plants exposed to industrial effluents found in decreasing order i.e. proteinaceous seeds> oil seeds> starchy seed (Singh et al., 2010). In overall three specie of rice, pulse and oil, Basmati, Chick pea and Alsi found susceptible to battery manufacturing, petrochemical, chemical industry effluent and thermal power plant effluents.

Ray and Barman (1988) studied the phytotoxic nature of the different types of crop plants exposed to steel & tar, cycle and distillery effluents and found the following sensitivity in decreasing order of proteinaceous seeds> oil seeds> starchy seed. Nath et al. (2008) examined the combined effect of sugar and distillery effluent on wheat, black gram, barley, garden pea and mustard germination. All seeds show inhibition in fresh weight with increasing effluent concentration. The level of phytotoxicity found as Barley> garden pea>wheat>black gram>mustard and also explain that mixed effluent is not advisable for irrigation. Mami et al. (2011) reported that effect of different concentration of Fe, Pb and Cu (0.0001, 0.001, 0.01, 0.1 and 1%) on seed germination on two different varieties of tomatoes. They found that heavy metals with higher concentration cause metabolic disorder and inhibits growth of seeds. Comparing with Fe and Pb, Cu had less inhibitive effect with increasing concentration.

Raia and Khan (2010) studied the different industrial effluent containing heavy metals Fe, Cu, Mn and Zn and its effect on the germination of Barley and observed that heavy metals accumulates in edible part of plants and through food chain its reached to consumers and cause adverse effects. Shaikh et al. (2013) assess the phytotoxicity of Mn, Cd, Cr and Zn on wheat in concentration of 0, 2, 4, 6, 8 and 10mgl^{-1}. In comparison with control result shows gradual reduction of root and shoot growth with increasing concentration.

Table 4.2.3: Phytotoxicity in various seed caused by various industrial effluent with different concentration.

Test Material		Growth media	Phytotoxicity (%)			
			Petrochemical	Chemical	Thermal power plant	Battery
Rice	Basmati-198	Control				
		Effluent	41.71	49.72	33.57	63.55
		Effluent + tap water (1:1)	11.82	33.47	21.66	39.80
	Pusa sugandh	Control				
		Effluent	44.72	60.03	35.16	57.54
		Effluent + tap water (1:1)	21.59	33.28	13.74	24.46
	Sambha masuri	Control				
		Effluent	52.32	55.47	33.23	45.57
		Effluent + tap water (1:1)	32.38	22.97	18.78	28.52
Pulse	Chick pea	Control				
		Effluent	65.33	44.59	33.24	50.59
		Effluent + tap water (1:1)	26.51	27.57	20.41	27.14
	Moong N-1	Control				
		Effluent	48.28	49.01	26.56	54.90
		Effluent + tap water (1:1)	18.62	23.38	14.54	26.75
	Massor VL-1	Control				
		Effluent	47.40	32.78	33.08	42.69
		Effluent + tap water (1:1)	20.62	17.58	15.79	25.13
Oil seed	Mustard BR-40	Control				
		Effluent	45.59	36.46	23.52	40.68
		Effluent + tap water (1:1)	18.97	15.16	8.54	15.24
	Alsi KL-43	Control				
		Effluent	43.88	27.37	43.10	48.32
		Effluent + tap water (1:1)	21.56	9.66	19.33	19.66
	Sesame G Til-3	Control				
		Effluent	41.30	32.28	30.85	41.84
		Effluent + tap water (1:1)	22.61	9.19	15.67	26.21

Germination

Germination is believed to be the most sensitive and vulnerable to water availability and to the solids and other toxic compounds of the growth medium (Khan et al., 2002). The decrease in water intake of the seed in toxic condition, osmotic, and ion toxic effect prevents the seed germination (Rahman et al., 2008). Moreover, salt concentration reduces the germination rate (Sabir and Ashraf, 2005; Akbari et al., 2007). This is an important phenomenon as the seeds that germinate faster can make use of the limited soil and water and establish seedlings, which is less sensitive to stress conditions, before stress conditions prevail (Akbari et al., 2007). Other researchers have reported that salt stress resulted in a reduced cell turgor and depressed rates of root and leaf elongation (Fricke et al., 2006). It has been reported that high concentrations of industrial effluent result on strong prohibition of seeds and seedling growth (Casa et al. 2003; Komilis et al. 2005).

Seed germination was calculated in the percentage shown in the Table 4.2.4. In Basmati range was 76.67 to 90.00%, 70.00 to 86.67% and 56.67 to 80.00%, in Pusa Sugandh germination varies between 80.00 to 86.67, 70.00 to 83.33% and 60.00 to 76.67% and in Sambha masuri it was range between 83.33 to 90.00%, 76.67 to 80.00% and 46.67 to 73.33% in 0%, 50% and 100% concentration in all industrial effluent respectively. Maximum variation shows in 100% effluent concentration of all the industries. All three rice varieties show maximum germination in petrochemical effluent while battery manufacturing industry show minimum germination. In the germination of pulses, chick pea germination varies between 76.67 to 93.33%, 70.00 to 83.33% and 56.67 to 76.67%, in Moong its range between N-1, 86.67 to 100%, 80.00 to 90.00% and 60.00 to 83.33% and in case of its range between Massor VL-1, 80.00 to 86.67%, 70.00 to 80.00% and 56.57 to 66.67% in 0, 50 and 100% concentrated effluent. Like rice, pulses are also maximally generated in petrochemical industry effluent except Moong-N-1 which is generated in thermal power plant effluent. Minimum gemination was shown by battery manufacturing industry. In germination of oil seed, Mustard BR-40 germination found in the range of 80.00 to 90.00%, 70.00 to 80.00% and 60.00 to 70.00%, in Alsi KL-43 it was found in the range of 76.67 to 90.00, 63.33 to 80.00% and 53.33 to 70.00% and in the Sesame G til-3 germination range found in between 86.67 to 93.33%, 76.67 to 83.33 and 63.33 to 73.33 % in 0, 50 and 100% concentrated effluent. Mustard and Sesame are show maximum germination in petrochemical effluent but Alsi shows in thermal power plant. Like rice and pulse oil seeds are also minimally germinated in battery manufacturing effluent.

Ramanna et al. (2002) conducted the experiment on seed of Chilli, Onion, Cucumber, Bottle guard and Tomato, to see the effect of different concentration (0, 5, 10, 15, 25,50, 75 and 100%) of raw distillery effluent on seed germination, speed of germination and peak value of germination and found complete failure of germination was observed in 75% and 100% concentration. Narain et al. (2012) reported the effect of distillery effluent (different concentration) on seed germination in *Cicer arietinum* and found percentage germination, radicle length and chlorophyll contents decline with increasing effluent concentration. 25% effluent did not show any inhibitory effect but seeds in 100% effluent did not survive long. They clarify that if concentration of pollutant is low in effluent, its promoting sustainable agriculture and also conservation of water resource.

Reduction in germination in absolute wastewater may be due to the higher amounts of pollutants like solids etc. present in the water, which is reason of fluctuations in the osmotic relationship of the seed and water. Thus, less water absorption takes place which delay germination due to, higher salinity. The solids present in medium act as a limiting factor and it can be responsible for delay in the germination (Adraino et al., 1982). The other possibility of least germination may be due to the presence of higher concentration of heavy metals in the effluent, causing depletion of the tricarboxylic acid cycle which reduces the respiration rate and subsequent germination (Kirkby, 1968; Ashwini et al., 2014).

Tomulescu et al. (2004) studied the toxicity of heavy metals (Cu, Zn and Pb) and observed that Cu and Zn is significantly increase the germination if individually used but germination decrease when used together. Pb was found toxic even very low concentration but its toxicity also increases when applied with Cu. Ali et al. (2004) also studied the phytotoxicity test of Cu and Cr on barley seeds and they found that toxicity of Cu and Cr to root is generally higher but Cr toxicity was higher than Cu. As we know that Cu is essential metal for plant growth but in combination with other metals its cause toxicity to plants. Several researchers reported heavy metal tolerance in plant they found radicle growth towards heavy metals (Punz and Siegardt, 1993; Ali et al., 2004; Farooqi et al., 2009). Cu is toxic to Mustards, Cd was toxic to Cucumber and Mustard (Moreno-casellles et al., 2000), Cu and Cd was toxic to Brinjal (Neelima and Beardsell, 1981), heavy metal also found toxic to wheat seedlings (Oncel et al., 2000). According to Saxena et al. (1986) the low amount of oxygen in dissolved form due to the presence of higher concentration of solids and oil and grease in effluent, reduces the energy supply through anaerobic respiration resulting in restriction of the growth and development of seedlings.

Table 4.2.4: Germination of various variety of rice, pulse and oil seed in different concentration of effluent from various industry

Test Material		Growth media	Germination (%)			
			Petrochemical	Chemical	Thermal power plant	Battery
Rice	Basmati-198	Control	90.00	86.67	83.33	76.67
		Effluent	80.00	70.00	70.00	56.67
		Effluent + tap water (1:1)	86.67	80.00	76.67	70.00
	Pusa sugandh	Control	86.67	86.67	83.33	80.00
		Effluent	76.67	63.33	63.33	60.00
		Effluent + tap water (1:1)	83.33	76.67	73.33	70.00
	Sambha masuri	Control	90.00	83.33	86.67	83.33
		Effluent	73.33	70.00	66.67	46.67
		Effluent + tap water (1:1)	80.00	76.67	76.67	76.67
Pulse	Chick pea	Control	93.33	86.67	83.33	76.67
		Effluent	76.67	66.67	70.00	56.67
		Effluent + tap water (1:1)	83.33	80.00	76.67	70.00
	Moong N-1	Control	96.67	96.67	100.00	86.67
		Effluent	76.67	80.00	83.33	60.00
		Effluent + tap water (1:1)	90.00	86.67	86.67	80.00
	Massor VL-1	Control	83.33	80.00	80.00	86.67
		Effluent	66.67	63.33	63.33	56.67
		Effluent + tap water (1:1)	80.00	73.33	73.33	70.00
Oil seed	Mustard BR-40	Control	90.00	83.33	86.67	80.00
		Effluent	70.00	66.67	70.00	60.00
		Effluent + tap water (1:1)	80.00	73.33	76.67	70.00
	Alsi KL-43	Control	83.33	86.67	90.00	76.67
		Effluent	63.33	70.00	66.67	53.33
		Effluent + tap water (1:1)	73.33	76.67	80.00	63.33
	Sesame G Til-3	Control	90.00	93.33	86.67	86.67
		Effluent	73.33	73.33	70.00	63.33
		Effluent + tap water (1:1)	83.33	80.00	76.67	80.00

Germination index

Seed germination index is the promising tool which provides comprehensive interpretation for root elongation and seed germination (IRSA, 1983). They offering no. of advantage in commonly used bioassays including easy to obtaining plant and seeds, higher no. of test plant, possibility of test, detection of two different point at same time (root elongation and seed germination), low cost, easy and rapidity in use. Ecological relevance is satisfied the fact that this test provides information on the response of higher plants.

Germination index calculated in rice, pulse and oil seed at 100% concentrated effluent was range between 26.94 to 55.80, 29.21 to 49.28 and 30.48 to 51.36 in Basmati, Pusa sugandh and Sambha masuri, 31.22 to 61.20, 37.48 to 53.22 and 42.32 to 61.77 in chick pea, moong and Massor and 42.32 to 61.77, 35.95 to 58.66 and 42.5 to 55.86 in Mustard, Alsi and Sesame. At 50% dilution of the effluent it was range between 54.96 to 84.91, 59.02 to 75.91 and 60.10 to 75.19 in Chick pea, Moong and Massor and 72.03 to 80.91, 66.36 to 79.92 and 68.12 to 77.84 in Mustard, Alsi and Sesame.

Kumar et al. (2014) reported that battery manufacturing industry wastewater reduced seed germination and inhibits radicle length in *Trigonella fornumgracum* and *Raphanus sativus.* They revealed that effluent discharged by the industry concerned is highly variable in its phytotoxic nature, and phytotoxicity also varied with plants. According to Kumar et al. (2014), germination percentage and radicle length is combined interpretation of industrial effluent for germination index which varies in ranged between 0.11 to 0.67 & 0.36 to 0.93 for Radish and Methi respectively. The germination index in both the crops decreased with increase in concentration. However, the extent of reduction was more in Radish as compared to the Methi. The results showed that the Battery manufacturing industry effluent has reduced the seed germination and inhibited the radicle growth in both the plants. The present investigation revealed that the effluent discharged by the factory concerned is highly variable in its phytotoxic nature, further the phytotoxicity also varied with plant varieties means Methi relatively tolerant as compare to Radish.

Table 4.2.5: Germination index of rice, pulse and oil seed variety grow in different concentration of effluent.

Test Material		Growth media	Germination index (%)			
			Petrochemical	Chemical	Thermal power plant	Battery
Rice	Basmati-198	Control	100.00	100.00	100.00	100.00
		Effluent	51.81	40.61	55.80	26.94
		Effluent + tap water (1:1)	84.91	61.41	72.07	54.96
	Pusa sugandh	Control				
		Effluent	48.90	29.21	49.28	31.85
		Effluent + tap water (1:1)	75.39	59.02	75.91	66.10
	Sambha masuri	Control				
		Effluent	38.85	37.41	51.36	30.48
		Effluent + tap water (1:1)	60.10	70.87	75.19	65.76
Pulse	Chick pea	Control				
		Effluent	28.48	42.63	56.08	36.52
		Effluent + tap water (1:1)	65.62	66.86	73.22	66.52
	Moong N-1	Control				
		Effluent	41.02	42.20	61.20	31.22
		Effluent + tap water (1:1)	75.76	68.69	74.07	67.61
	Massor VL-1	Control				
		Effluent	42.08	53.22	52.98	37.48
		Effluent + tap water (1:1)	76.21	75.55	77.19	60.47
Oil seed	Mustard BR-40	Control				
		Effluent	42.32	50.83	61.77	44.49
		Effluent + tap water (1:1)	72.03	74.66	80.91	74.17
	Alsi KL-43	Control				
		Effluent	42.65	58.66	42.15	35.95
		Effluent + tap water (1:1)	69.03	79.92	71.71	66.36
	Sesame G Til-3	Control				
		Effluent	47.83	53.21	55.86	42.50
		Effluent + tap water (1:1)	77.39	77.84	74.60	68.12

Germination rate index was analyzes according to Rusan et al. (2015) which indicate the toxicity, when the value is higher means more rapid rate of germination and shows better performance (Wang, 2004). Lowest germination rate index shows in Sesame (0.42) grown in battery effluent while highest shows in moong (0.93) in thermal power plant at 100% concentrated effluent. In case of 50% dilution same species shows least (0.54) and higher (1.20) values with battery and petrochemical effluent.

Table 4.2.6: Germination rate index in three different variety of rice, pulse and different industrial wastewater.

Test Material		Growth media	Germination rate index (GRI)			
			Petrochemical	Chemical	Thermal power plant	Battery
Rice	Basmati-198	Control	1.23	1.20	1.16	1.19
		Effluent	0.72	0.60	0.77	0.44
		Effluent + tap water (1:1)	1.08	0.80	0.91	0.72
	Pusa sugandh	Control	1.09	1.10	1.13	1.08
		Effluent	0.60	0.44	0.73	0.46
		Effluent + tap water (1:1)	0.85	0.73	0.97	0.82
	Sambha masuri	Control	1.11	1.07	1.12	0.96
		Effluent	0.53	0.48	0.75	0.52
		Effluent + tap water (1:1)	0.75	0.82	0.91	0.69
Pulse	Chick pea	Control	1.25	1.19	1.14	1.27
		Effluent	0.43	0.66	0.76	0.63
		Effluent + tap water (1:1)	0.92	0.86	0.91	0.92
	Moong N-1	Control	1.38	1.31	1.27	1.34
		Effluent	0.77	0.77	0.93	0.70
		Effluent + tap water (1:1)	1.20	1.17	1.13	1.12
	Massor VL-1	Control	1.03	0.70	0.67	0.97
		Effluent	0.54	0.47	0.45	0.56

		Effluent + tap water (1:1)	0.82	0.58	0.56	0.73
Oil seed	Mustard BR-40	Control	1.13	0.92	0.96	1.13
		Effluent	0.62	0.59	0.73	0.67
		Effluent + tap water (1:1)	0.92	0.78	0.88	0.96
	Alsi KL-43	Control	1.09	0.93	1.09	1.09
		Effluent	0.61	0.68	0.62	0.57
		Effluent + tap water (1:1)	0.86	0.84	0.88	0.88
	Sesame G Til-3	Control	0.77	0.64	0.67	0.73
		Effluent	0.45	0.43	0.46	0.42
		Effluent + tap water (1:1)	0.59	0.58	0.57	0.54

Phytotoxicity index (PI) in the measurement of germination and radicle length of the seeds. Its values range between 0 to 1 in which higher value indicate higher toxicity or negative effect and low values indicate lower toxicity or positive effect. Table 4.2.7 shows the PI of the effluents of all four industry. Calculated PI shows that 50% dilution reducing the toxicity of effluent. Sesame shows the least PI (0.09) at 50% effluent concentration in chemical industry effluent while chick pea shows higher PI (0.65) in petrochemical effluent at 100% concentration. Relatively good performance by 50% indicates the possibility of decrease the phytotoxicity with increasing the dilution with water also considered least expansive. Many techniques are used to treatment of industrial effluent but usually expansive and economically not feasible.

Table 4.2.7: Phytotoxicity index (PI) in different variety of seeds grown in different industrial effluent.

Test Material		Growth media	Phytotoxicity index (PI)			
			Petrochemical	Chemical	Thermal power plant	Battery
Rice	Basmati-198	Control				
		Effluent	0.42	0.50	0.34	0.64
		Effluent + tap water (1:1)	0.12	0.33	0.22	0.40
	Pusa sugandh	Control				
		Effluent	0.45	0.60	0.35	0.58
		Effluent + tap water (1:1)	0.22	0.33	0.14	0.24
	Sambha masuri	Control				
		Effluent	0.52	0.55	0.33	0.46
		Effluent + tap water (1:1)	0.32	0.23	0.19	0.29
Pulse	Chick pea	Control				
		Effluent	0.65	0.45	0.33	0.51
		Effluent + tap water (1:1)	0.27	0.28	0.20	0.27
	Moong N-1	Control				
		Effluent	0.44	0.41	0.27	0.48
		Effluent + tap water (1:1)	0.13	0.11	0.11	0.16
	Massor VL-1	Control				
		Effluent	0.47	0.33	0.33	0.43
		Effluent + tap water (1:1)	0.21	0.18	0.16	0.25
Oil seed	Mustard BR-40	Control				
		Effluent	0.46	0.36	0.24	0.41
		Effluent + tap water (1:1)	0.19	0.15	0.09	0.15
	Alsi KL-43	Control				
		Effluent	0.44	0.27	0.43	0.48
		Effluent + tap water (1:1)	0.22	0.10	0.19	0.20

	Sesame G Til-3	Control				
		Effluent	0.41	0.32	0.31	0.42
		Effluent + tap water (1:1)	0.23	0.09	0.16	0.26

Seedling vigor is another important parameter which evaluated the supplement germination and viability and understand performance of a seed. According to the ISTA (1966) seed vigor is the "sum of those properties of the seed which control the level of activity of the seeds during seedling emergence".

Seedling vigor index estimated in basmati rice found to be 220.80, 208.00, 193.61 and 182.98 at 0%, 187.49, 127.73, 139.53, 100.5 at 50% and 114.40, 84.47, 108.03, 49.30 at 100% concentrated effluent in petrochemical, chemical, thermal power plant and battery industry.

Table 4.2.8: Seedling vigor index of different variety of rice, pulse and oil seed in response to 50% and 100% concentration of industrial effluent.

Test Material		Growth media	Seedling vigour index (%)			
			Petrochemical	Chemical	Thermal power plant	Battery
Rice	Basmati-198	Control	220.80	208.00	193.61	182.98
		Effluent	114.40	84.47	108.03	49.30
		Effluent + tap water (1:1)	187.49	127.73	139.53	100.57
	Pusa sugandh	Control	188.64	190.09	188.06	173.33
		Effluent	92.26	55.52	92.68	55.20
		Effluent + tap water (1:1)	142.22	112.19	142.76	114.57
	Sambha masuri	Control	200.10	177.78	193.84	159.72
		Effluent	77.73	66.50	99.56	48.69

		Effluent + tap water (1:1)	120.27	125.99	139.28	105.03
Pulse	Chick pea	Control	232.40	205.40	190.56	193.97
		Effluent	66.19	87.56	106.87	70.83
		Effluent + tap water (1:1)	152.50	137.33	139.53	129.03
	Moong N-1	Control	266.80	253.91	253.33	232.27
		Effluent	117.81	123.20	154.17	84.40
		Effluent + tap water (1:1)	215.40	203.38	195.29	179.47
	Massor VL-1	Control	171.11	112.27	106.40	167.84
		Effluent	72.00	59.74	56.37	62.90
		Effluent + tap water (1:1)	130.40	84.82	82.13	101.50
Oil seed	Mustard BR-40	Control	204.00	153.89	165.82	180.27
		Effluent	86.33	78.22	102.43	80.20
		Effluent + tap water (1:1)	146.93	114.89	134.17	133.70
	Alsi KL-43	Control	181.67	161.49	195.60	167.64
		Effluent	77.48	94.73	82.44	60.27
		Effluent + tap water (1:1)	125.40	129.06	140.27	111.26
	Sesame G Til-3	Control	138.00	118.53	116.13	125.67
		Effluent	66.00	63.07	64.87	53.41
		Effluent + tap water (1:1)	98.89	92.27	86.63	85.60

The toxicity cause by different industrial wastewater to three varieties of rice, pulse and oil seeds depends on the nature of seed and effluent. Several studies have been done on the chemical nature of industrial effluent and results shows that every industry has its own waste product that's pollute water bodies. The degree of phytotoxicity of industrial effluents depends not only on the chemical composition of the effluents but also on the type of crops cultivated. Different crop varieties show different sensitivities to the toxic nature of effluents.

The germination of seeds as well as its radicle growth depends not only on chemical nature of the effluent and the plant species, but also on the interaction of the solution with the seed coat as well as membrane of the seeds. At germination level, seeds use their reserved food as a source of energy to initiate metabolic activity. In this study, results revealed that the dilution of wastewater increased the radicle growth and reduced the percent phytotoxicity, which means, that the pollutants responsible for the retardation of the radicle growth were also diluted and had become less toxic.

From the comparison of results, it is evident that the effluent with dilution can be used for irrigation of crops. The oil seed varieties followed by pulses and rice show tolerance to 50% effluent. It clearly suggests that carbohydrate rich seeds are less susceptible to industrial effluent. The order of tolerance in different crop seeds varies depending upon the nature of effluent. In general rice is the most susceptible crop and oilseeds are more tolerant crops towards toxicity of effluents of chemical and battery manufacturing effluent, which are rich in heavy metal toxicants. Pulses are most susceptible to toxicity of petrochemical industry and chemical industry effluents.

The present investigation reveals that the effluents discharged by the industries concerned are highly variable in their phytotoxic nature. The varietal differences in the test materials also caused due to differences in the response of crops to industrial effluents, signifying that all plants, even when closely related, are not equally sensitive to similar changes in environmental conditions. However, plants having seeds with the same type of food reserve, such as starch, protein or oil, tend to behave more or less in a similar way, though to different extent depending upon the variety, while there appears to be some difference among the three classes of the test materials used. The information on the varietal differences among the different types of plants species will be helpful for the screening of variety for cultivation in fields polluted with the industrial effluents. However, growth of plants in the field studies can be improved by the degradation of pollutants by other physical & chemical factors and also by the physiological development after certain stage of plant life, which is controlled by the specific gene activity.

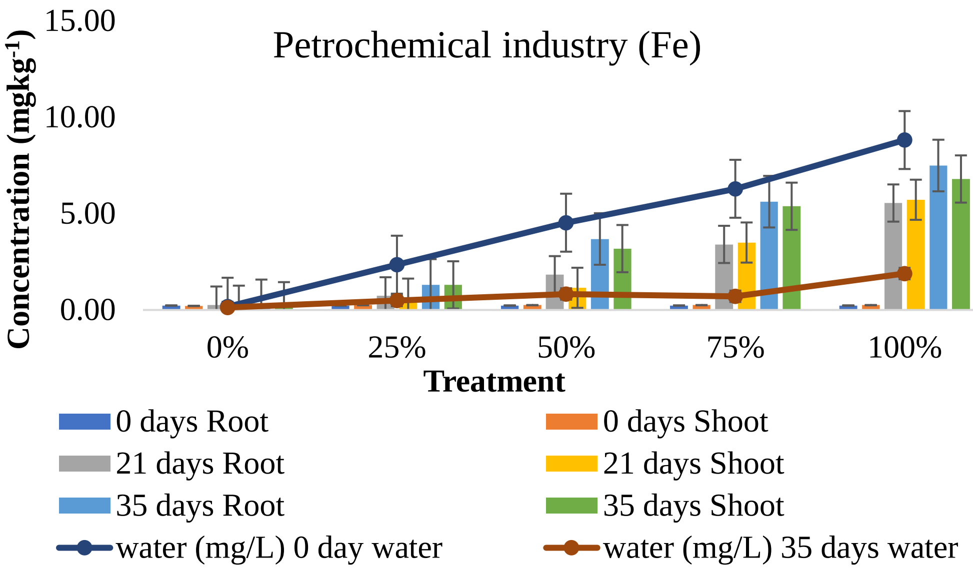
Chapter 4.3

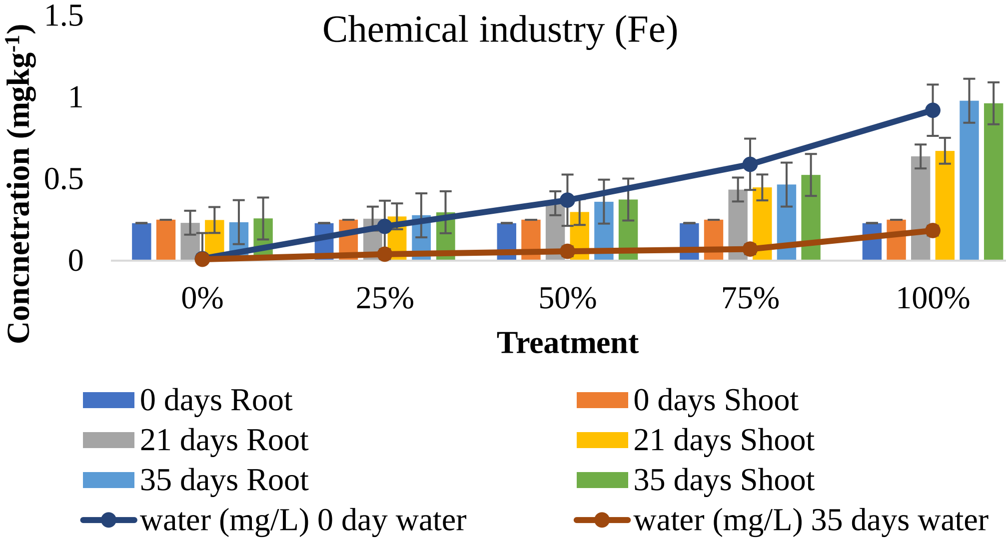
To study the partitioning of metals within different aquatic weeds exposed to different industrial wastewater.

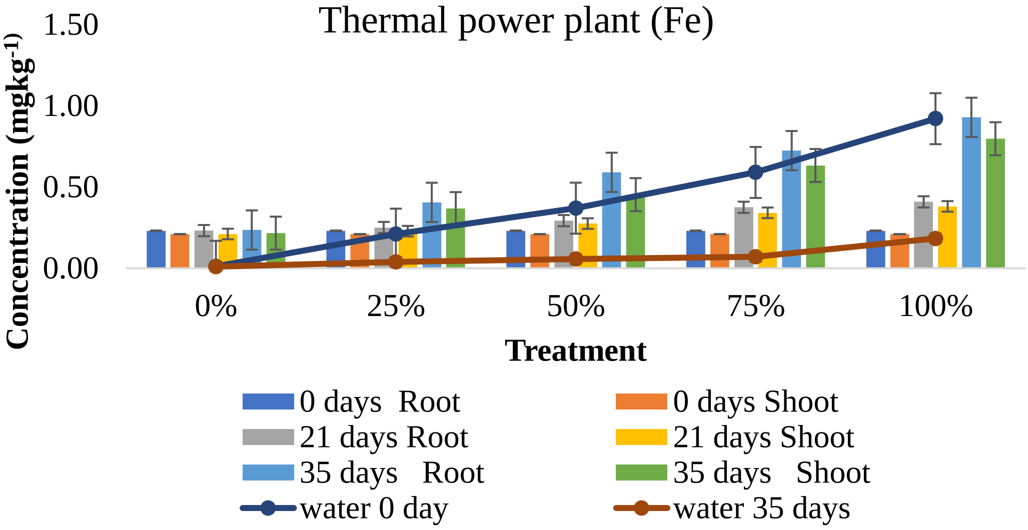
To study the partitioning of metals within different aquatic weeds exposed to different industrial wastewater.

Industrial wastewater pollution is most important and discussed concern throughout the world (Vessely et al., 2011). Many conventional approaches are used to control effect of wastewater on aquatic environment. Phytoremediation is efficient technology to remove heavy metals and other inorganic substance from effluent and improve its quality. However, the country has a rich diversity of aquatic macrophytes with potential to hyperaccumulate many heavy metals and can be used as an alternative way of remediating aquatic metal burden (Rai, 2008). *Pistia stratiotes* is extensively used for heavy metal removal. *Pistia* is a genus of aquatic macrophytes in the family Araceae, commonly called as water lettuce. It floats on the surface of the water and root are hanging beneath floating leaves. They are natural hyperaccumulators of many toxic heavy metals. The ability of *Pistia stratiotes* to clean wastewaters contaminated with heavy metals such as Hg, Mn, Zn, Pb, solid, Chlorides nitrate and phosphate was observed to find its relevance and as an application in phytoremediation technique. Phytoremediation using aquatic plants with high metal removal capacity provides an energy-efficient (use of solar energy) and cost-effective approach for reducing nonpoint source pollution in surface waters at a large scale. *Eichhornia crassipes* is naturally occurring worst aquatic weed widely used for wastewater treatment by uptake of pollutant.

Iron (Fe) is an essential metal for plants which is require in chlorophyll formation, enzyme activity and anions acceptance from substrate (Pavlovic et al., 2009). Fe accumulation in root and shoot in *E. crassipes* has been shown in field studies, for this its use as biological monitor for metals (Zaranyika and Ndapwadza, 1995). Fe concentration in root, shoot and water (before and after treatment) at 0 (control), 25, 50, 75 and 100% industrial effluent concentration are presented in fig 3.1. After 21 days of exposure to effluent of petrochemical industry, *E. crassipes* accumulates 0.25, 0.73, 1.83, 3.40 and 5.55 mgkg^{-1} Fe in root and 0.22, 0.59, 1.15, 3.50 and 5.70 mgkg^{-1} Fe in shoot respectively. After 35 days of duration *E. crassipes* accumulates 0.24, 1.30, 3.69, 5.62 and 7.50 mgkg^{-1} in roots and 0.22, 1.30, 3.19, 5.38 and 6.80 mgkg^{-1} in shoots respectively. Result shows that at 0%, Fe concentration low bioaccumulation occurred as compared with other concentration which is higher. However, lowest concentration found in shoot, while higher in roots. In exposure of chemical industry effluent for three weeks *E. crassipes* accumulate 0.23, 0.26, 0.35, 0.45, 0.64 mgkg^{-1} and 0.24, 0.27, 0.30, 0.45 and 0.64 mgkg^{-1} and After 35 days 0.24, 0.28, 0.36, 0.45, 0.98 mgkg^{-1} and 0.26, 0.30, 0.37, 0.52, 0.96 mgkg-1 Fe at 0, 25, 50, 75 and 100% concentrated effluent. Solton and Rashed (2003) explained that *E. crassipes* highly efficient in accumulation due to having their higher biomass, fibrous root and broad leaves which absorbed higher Fe from wastewater medium. When *E. crassipes* exposed to thermal power plant effluent it accumulates 0.23, 0.25, 0.29, 0.38, 0.41 and 0.21, 0.23, 0.28, 0.34 and 0.38 mgkg^{-1} Fe after 21 days and 0.24, 0.41, 0.59, 0.72, 0.93 and 0.22, 0.37, 0.45, 0.63, 0.80 mgkg^{-1} Fe after 35 days from 0, 25, 50, 75 and 100% concentrated effluent. In exposure of battery manufacturing industry effluent plants accumulates 0.24, 0.58, 0.77, 1.13, 1.21 and 0.22, 0.42, 0.65, 0.97, 1.11 mgkg^{-1} Fe after 21 days in root and shoot and 0.25, 0.59, 0.89, 1.26, 1.41 and 0.23, 0.43, 0.67, 1.02, 1.29 mgkg^{-1} Fe in root and shoot from 0, 25, 50, 75 and 100% effluent respectively. Accumulation was increase as increase effluent concentration in all the industries. Fe concentration was being previously higher in plant used in experiment.


Petrochemical industry (Fe)
Concentration (mgkg^{-1})
15.00
10.00
5.00
0.00
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Chemical industry (Fe)
Concnetration (mgkg^{-1})
1.5
1
0.5
0
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Thermal power plant (Fe)
Concentration (mgkg^{-1})
1.50
1.00
0.50
0.00
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water 0 day
water 35 days

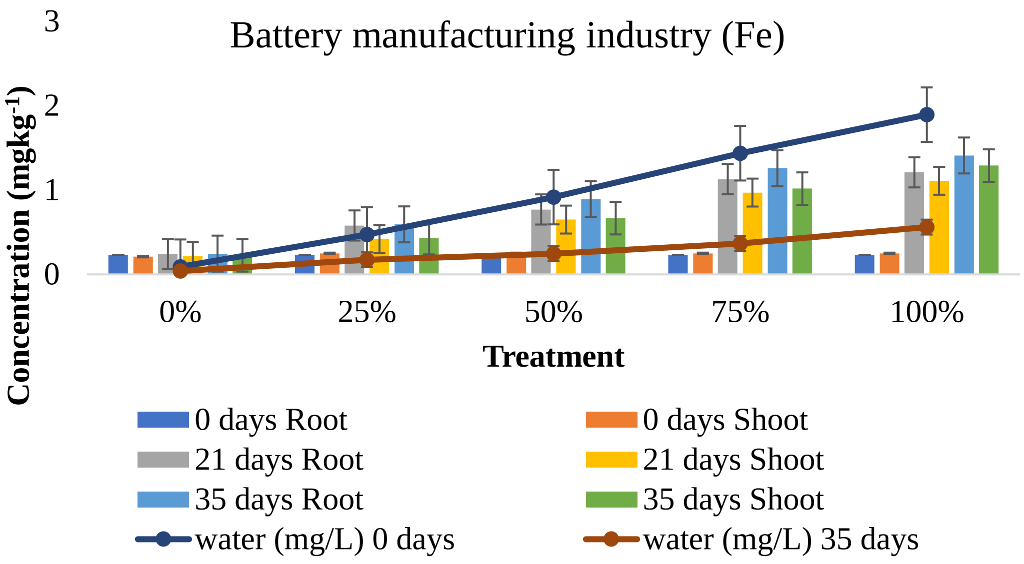
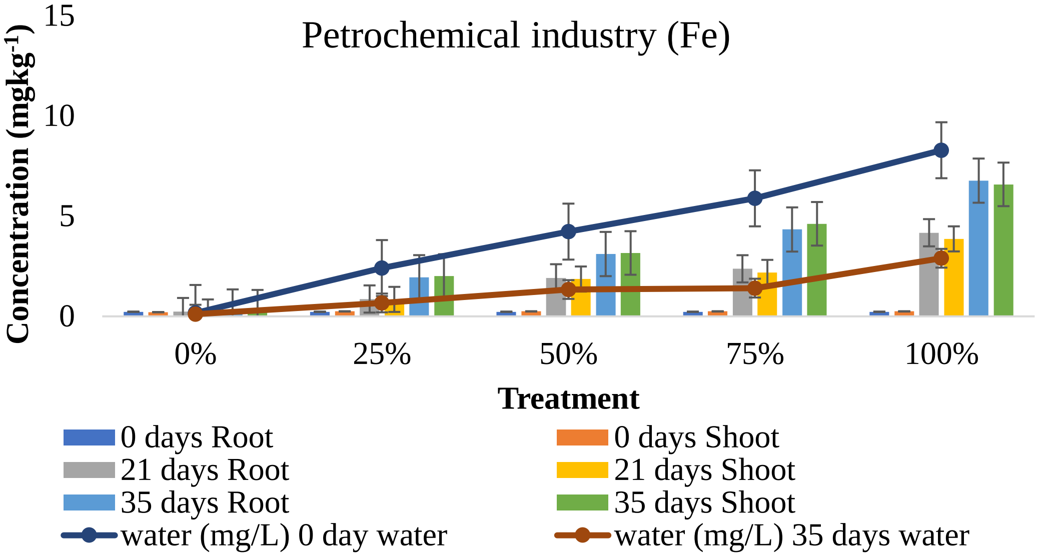
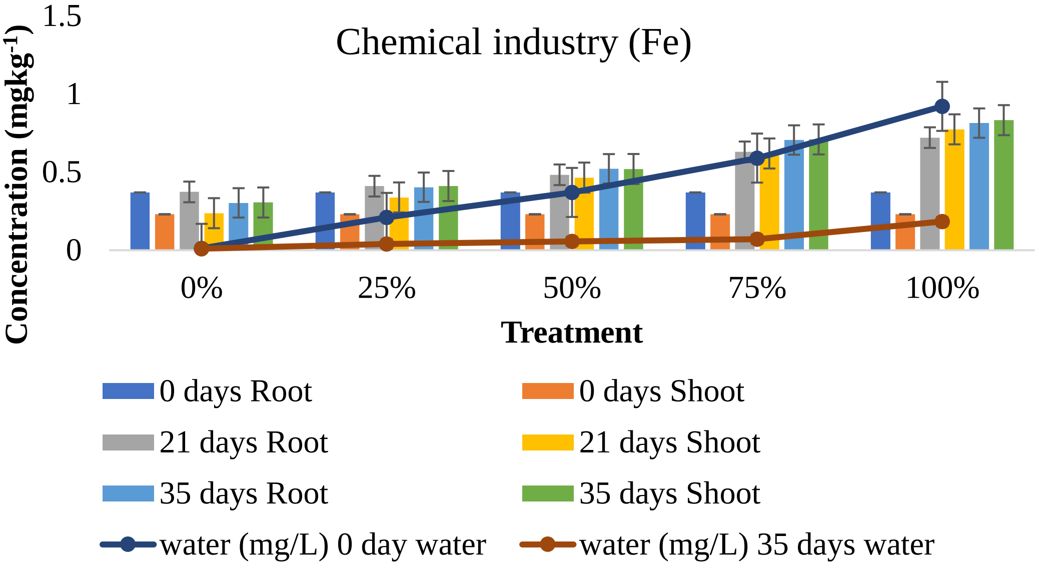
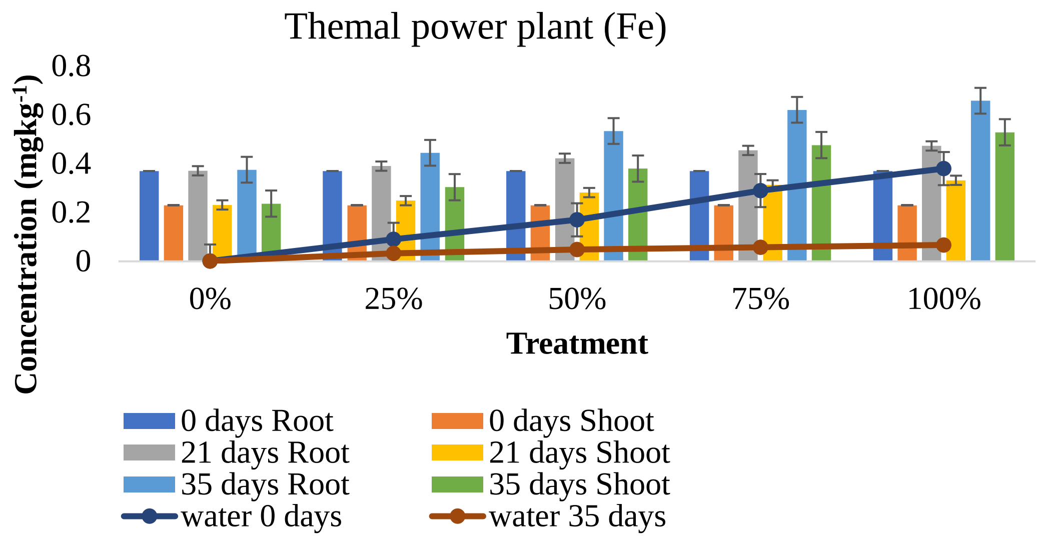


Fig. 4.3.1 Fe concentration in *E. crassipes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration.

Fe concentration initially found in *L. gibba* was 0.37 and 0.23 mgkg^{-1} in their root and shoot respectively. After 21 days of exposure it increases i.e. 0.34, 0.87, 1.92, 2.37, 4.17 mgkg^{-1} and 0.23, 0.85, 1.86, 2.19, 3.87 mgkg^{-1} in their root and shoot parts with 0, 25, 50, 75 and 100% concentrated effluent exposure. Subsequently, 0.37, 1.95, 3.11, 4.34, 6.77 mgkg^{-1} in root and 0.23, 2.02, 3.16, 4.62, 6.59 mgkg^{-1} in shoot accumulate after 35 days of exposure. From chemical industry effluent its accumulate 0.37, 0.41, 0.48, 0.63, 0.73 mgkg^{-1} and 0.24, 0.34, 0.47, 0.62, 0.77 mgkg^{-1} after 21 days and after 35 days its accumulates 0.38, 0.43, 0.54, 0.71, 0.81 mgkg^{-1} and 0.24, 0.41, 0.52, 0.71, 0.83 mgkg^{-1} in their root and shoot respectively. In thermal power plant effluent exposure, its accumulate 0.37, 0.39, 0.42, 0.45, 0.47 mgkg^{-1} and 0.23, 0.25, 0.28, 0.31, 0.33 mgkg^{-1} after 21 days and 0.38, 0.44, 0.53, 0.62, 0.66 mgkg^{-1} and 0.24, 0.30, 0.38, 0.48, 0.53 mgkg^{-1} after 35 days. In exposure of battery manufacturing industry effluent *L. gibba* accumulates 0.37, 0.45, 0.52, 0.64, 0.91 mgkg^{-1} and 0.23, 0.26, 0.32, 0.34, 0.67 mgkg^{-1} after 21 days and after 35 days its accumulate 0.37, 0.55, 0.62, 0.78, 1.09 and 0.23, 0.27, 0.41, 0.48, 0.77 mgkg^{-1} after 35 days.


Petrochemical industry (Fe)
Concentration (mgkg-1)
15
10
5
0
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Chemical industry (Fe)
Concentration (mgkg-1)
1.5
1
0.5
0
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water



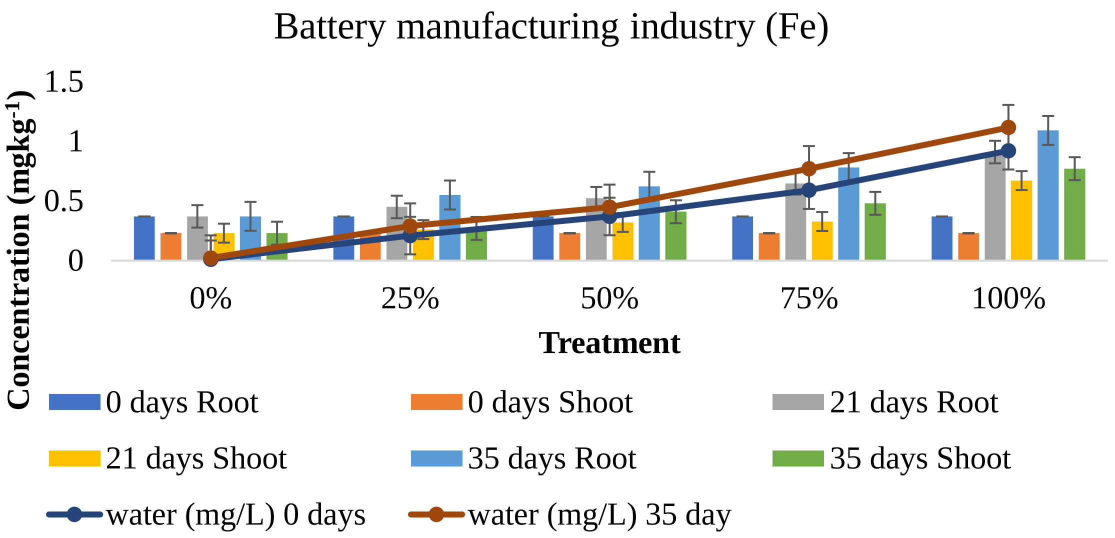
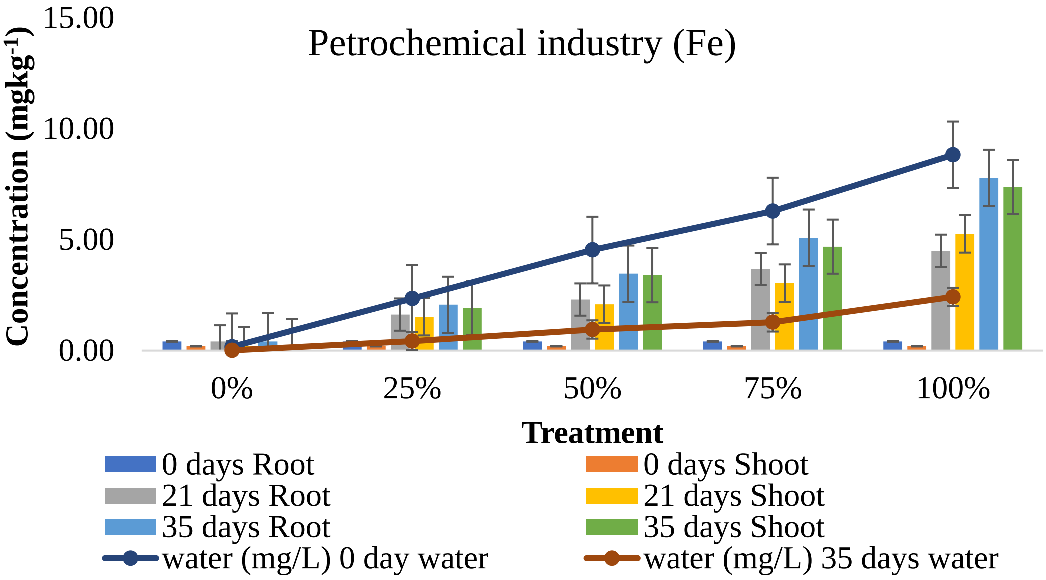


Fig. 4.3.2: Fe concentration in *L. gibba* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

P. stratiotes a floating macrophytes initially absorbed 0.41 and 0.19 mgkg-^{1} in their root and shoot respectively. After 21 days of exposure of petrochemical effluent it's accumulates 0.41, 1.62, 2.30, 3.68, 4.50 mgkg^{-1} and 0.20, 1.53, 2.09, 3.04, 5.26 mgkg^{-1} and after 35 days accumulation increase up to 0.42, 2.06, 3.47, 5.09, 7.79 mgkg^{-1} and 0.20, 1.92, 3.39, 4.68, 7.36 mgkg^{-1} in root and shoot at 0, 25, 50, 75, and 100% effluent concentration respectively. This plant showed highest accumulation of Fe after 5th week and remaining concentration in wastewater was 2.42 mgl^{-1} which is substantially lower than the CPCB prescribed limit of 3 mgl^{-1} for inland surface water. In treatment with chemical industry effluent, *P. stratiotes* accumulates 0.41, 0.48, 0.55, 0.64, 0.84 $mgkg^{-1}$ and 0.19, 26, 0.36, 0.45, 0.56 $mgkg^{-1}$ after 21 days and after 35 days its accumulates 0.41, 0.57, 0.69, 0.85, 0.97 $mgkg^{-1}$ and 0.20, 0.33, 0.40, 0.57, 0.76 $mgkg^{-1}$ in root and shoot respectively. When *P. stratiotes* exposed to effluent of thermal power plant then its accumulates 0.41, 0.50, 0.55, 0.61, 0.69 $mgkg^{-1}$ and 0.20, 0.28, 0.31, 0.36, 0.47 $mgkg^{-1}$ after 35 days of exposure. In battery manufacturing industry effluent, this macrophytes accumulates 0.41, 0.48, 0.54, 0.69, 0.89 $mgkg^{-1}$ and 0.19, 0.23, 0.28, 0.31, 0.71 $mgkg^{-1}$ Fe after 21 days and after 35 days its accumulates 0.41, 0.51, 0.69, 0.80, 1.09 $mgkg^{-1}$ and 0.19, 0.25, 0.31, 0.48, 0.77 $mgkg^{-1}$ Fe in their root and shoot respectively.



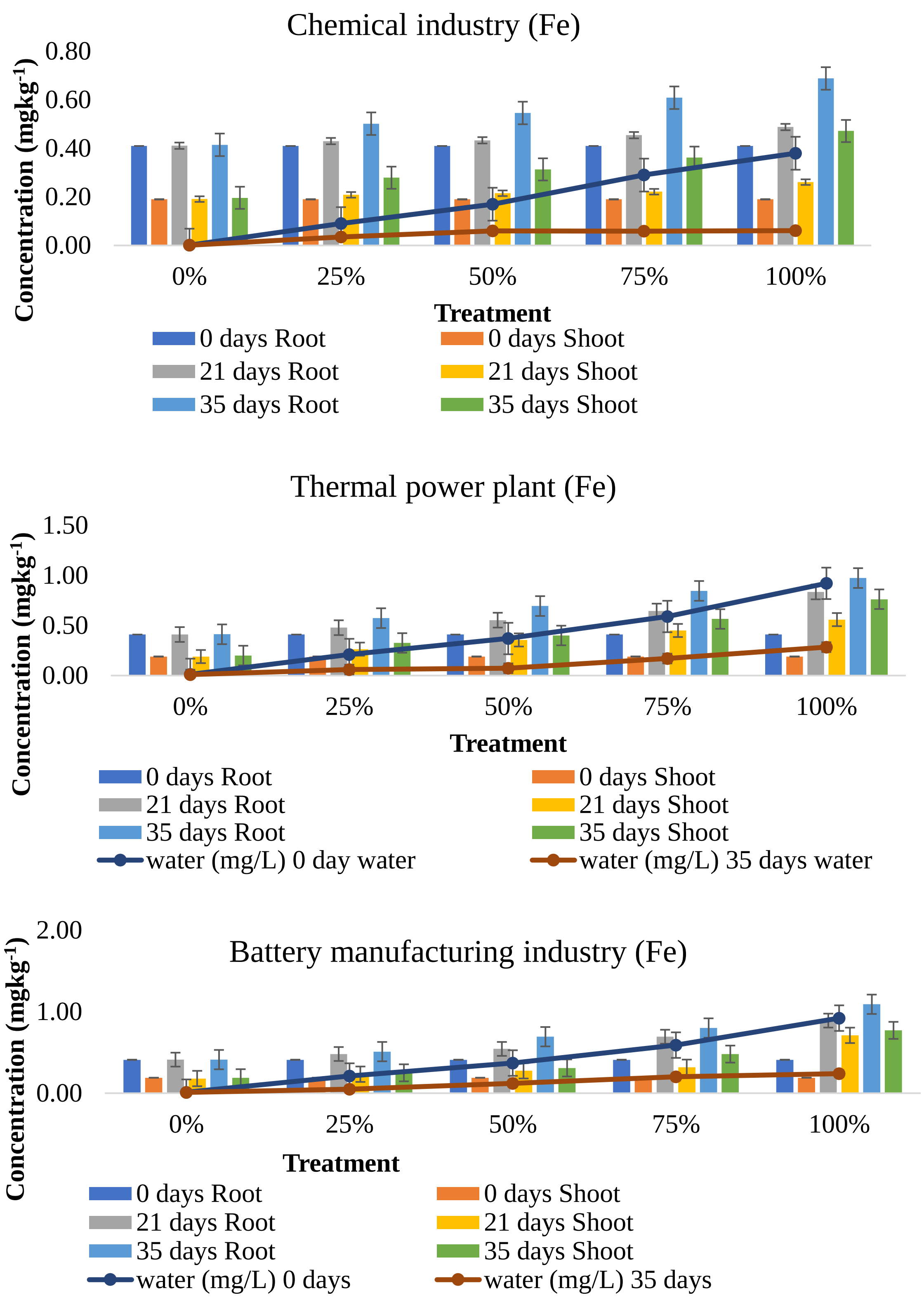
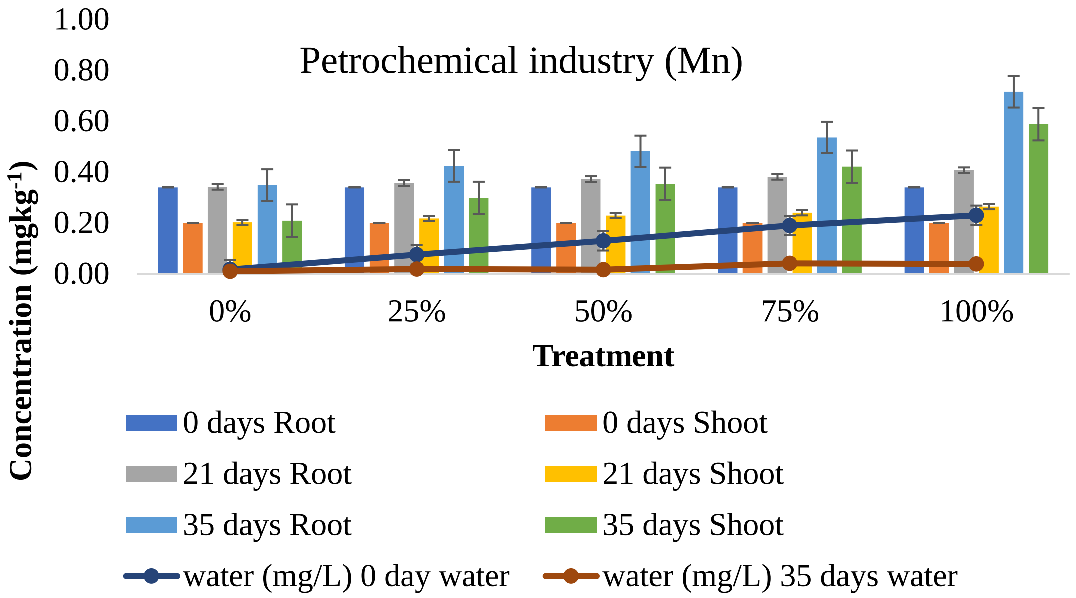
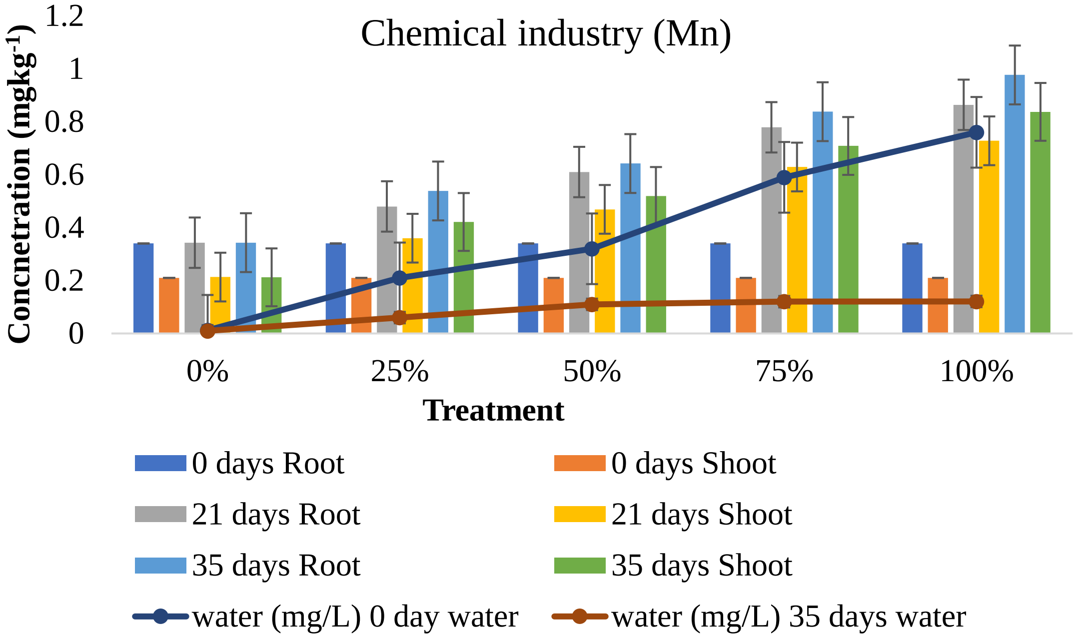
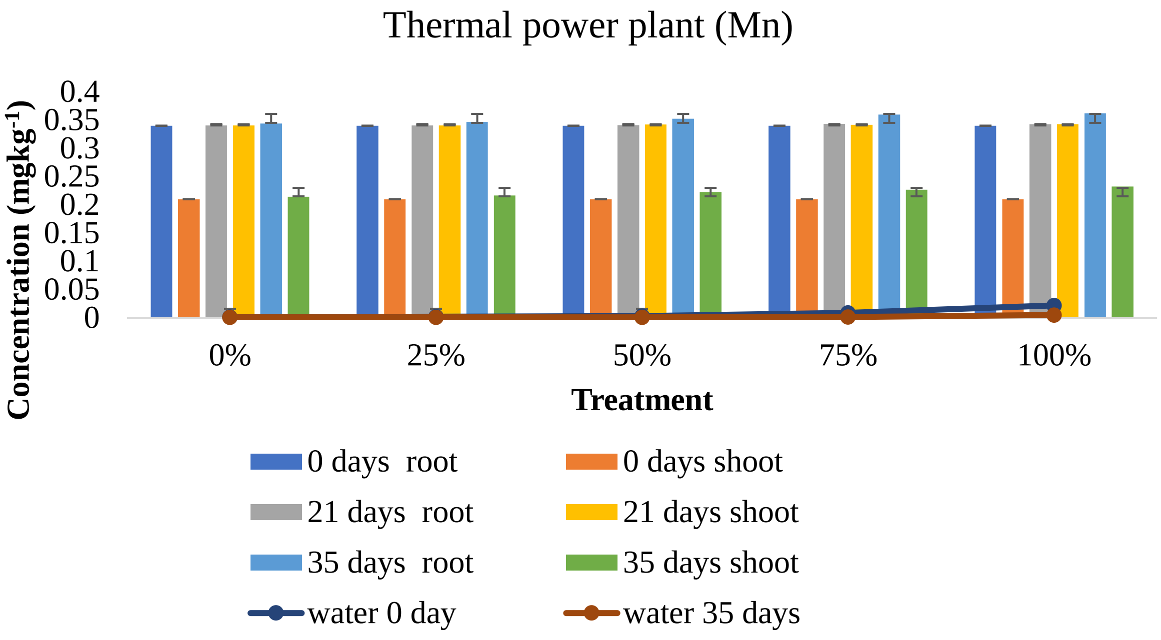


Fig. 4.3.3: Fe concentration in *P. stratiotes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

Mn is another essential metal used in enzymatic activity of plants. Initially *E. crassipes* gathers 0.34 and 0.21 mgkg^{-1} Mn in their root and shoot. After 21 days of exposure *E. crassipes* concentrates 0.34, 0.36, 0.37, 0.38, 0.41 and 0.20, 0.22, 0.23, 0.24, 0.26 mgkg^{-1} and after 35 days its concentrates 0.35, 0.42, 0.48, 0.54, 0.72 and 0.21, 0.30, 0.35, 0.42, 0.59 mgkg-^{1} Mn in root and shoot at 0, 25, 50, 75 and 100% of petrochemical industry effluent. In chemical industry effluent exposure this macrophyte accumulates 0.34, 0.48, 61, 78, 0.86 and 0.21, 0.36, 0.47, 0.63, 0.73 mgkg^{-1} Mn after 21 days of exposure and after 35 days its accumulates 0.34, 0.54, 0.63, 0.84 0.98 and 0.21, 0.42, 0.52, 0.71, 0.84 mgkg^{-1} Mn in root and shoot of plant respectively. In case of thermal power plant effluent in which Mn concentration was very low, so that Mn concentration after 21 and 35 days of exposure was not varying much basically in 0, 25 and 50% concentrated effluent. *E. crassipes* accumulates only 0.00, 0.01, 0.01, 0.02, 0.02 and 0.00, 0.01, 0.01, 0.02, 0.02 mgkg^{-1} Mn after 35 days of exposure. In battery manufacturing industry effluent, its accumulates 0.34, 0.64, 0.83, 1.32, 1.47 and 0.21, 0.44, 0.69, 1.21, 1.25 mgkg^{-1} Mn after 21 days of exposure and accumulates 0.35, 0.76, 1.04, 1.54, 2.07 and 0.22, 0.54, 0.76, 1.35, 1.41 mgkg^{-1} after 35 days of exposure.


Petrochemical industry (Mn)
Concentration (mgkg-1)
1.00
0.80
0.60
0.40
0.20
0.00
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Chemical industry (Mn)
Concnetration (mgkg-1)
1.2
1
0.8
0.6
0.4
0.2
0
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water



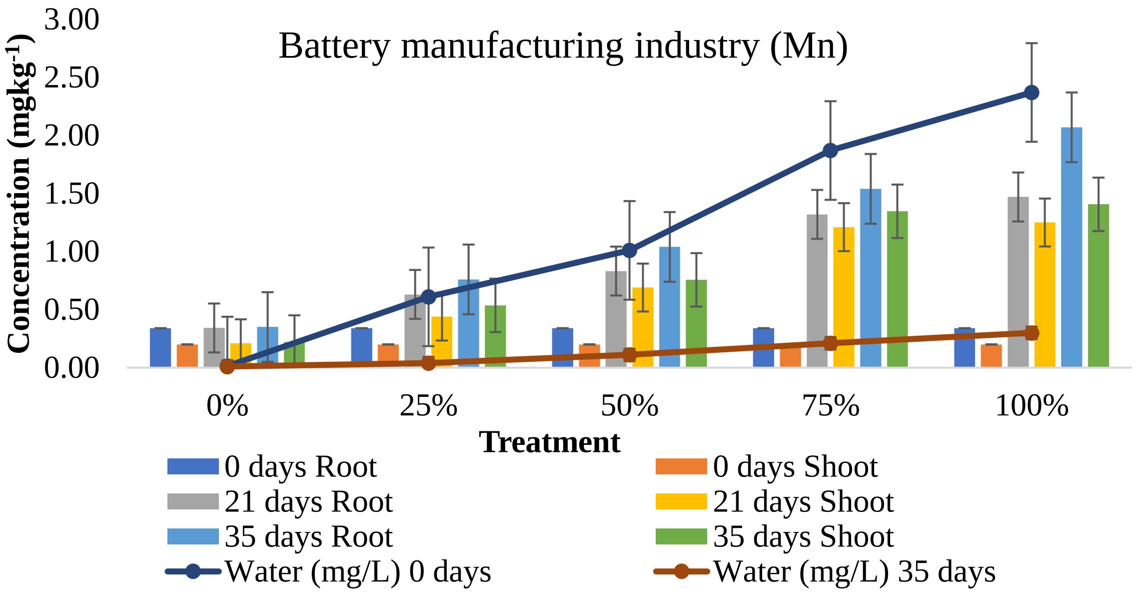
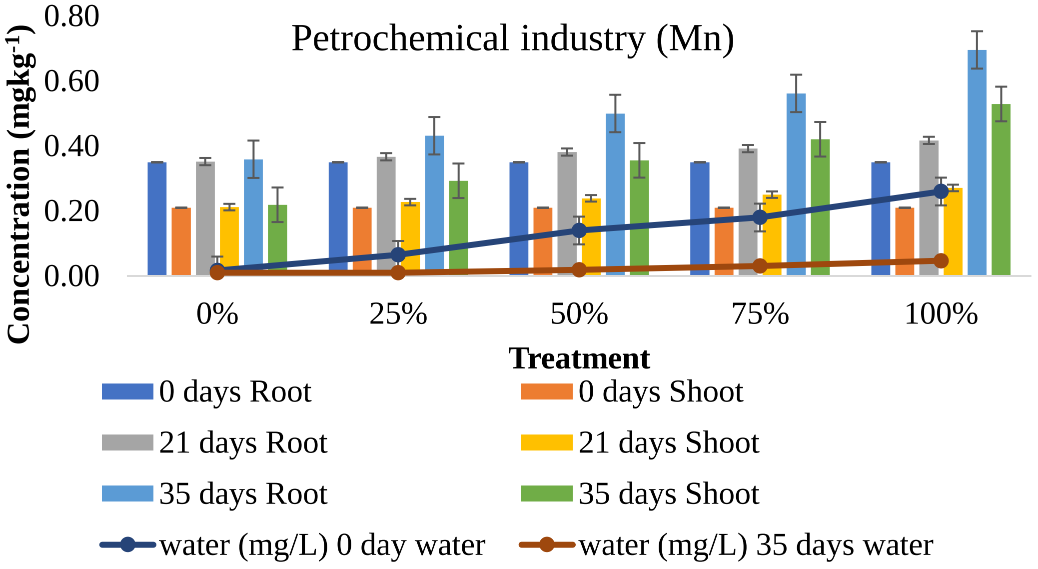


Fig.4.3.4: Mn concentration in *E. crassipes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

Initial Mn concentration was found to be 0.35 mgkg^{-1} and 0.21 mgkg^{-1} in root and shoot respectively. After 21 days it increases up to 0.35, 0.37, 0.38, 0.39, 0.42 in root and 0.21, 0.23, 0.24, 0.25, 0.27 mgkg^{-1} in shoot and after 35 days 0.36, 0.43, 0.50, 0.56, 0.70 mgkg^{-1} in root and 0.22, 0.29, 0.36, 0.42, 0.53 mgkg-1 from 0, 25, 50, 75, 100% petrochemical effluent. In chemical industry effluent exposure after 21 days *L. gibba* accumulate 0.59, 0.72, 0.78, 0.99, 1.09 $mgkg^{-1}$ 0.43, 0.56, 0.64, 0.90, 0.97 $mgkg^{-1}$ and after 35 days 0.59, 0.76, 0.88, 1.05, 1.24 $mgkg^{-1}$ and 0.43, 0.67, 0.77, 0.96, 1.13 $mgkg^{-1}$ in their root and shoot respectively at different concentration. In thermal power plants this plant accumulates 0.37, 0.39, 0.42, 0.45, 0.47 in root, 0.38, 0.44, 0.53, 0.62, 0.66 $mgkg^{-1}$ in shoot after 21days, and after 35 days of exposure its accumulates 0.38, 0.44, 0.53, 0.62, 0.66 $mgkg^{-1}$ and 0.24, 0.30, 0.38, 0.48, 0.53 $mgkg^{-1}$ in root and shoot. In exposure with battery manufacturing industry *Lemna* accumulates 0.37, 0.39, 0.42, 0.45, 0.47 $mgkg^{-1}$ in root 0.23, 0.25, 0.28, 0.31, 0.33 $mgkg^{-1}$ in shoot after 21 days, and 0.38, 0.44, 0.53, 0.62, 0.66 $mgkg^{-1}$ 0.24, 0.30, 0.53, 0.62, 0.66 $mgkg^{-1}$ after 35 days in their root and shoot respectively.



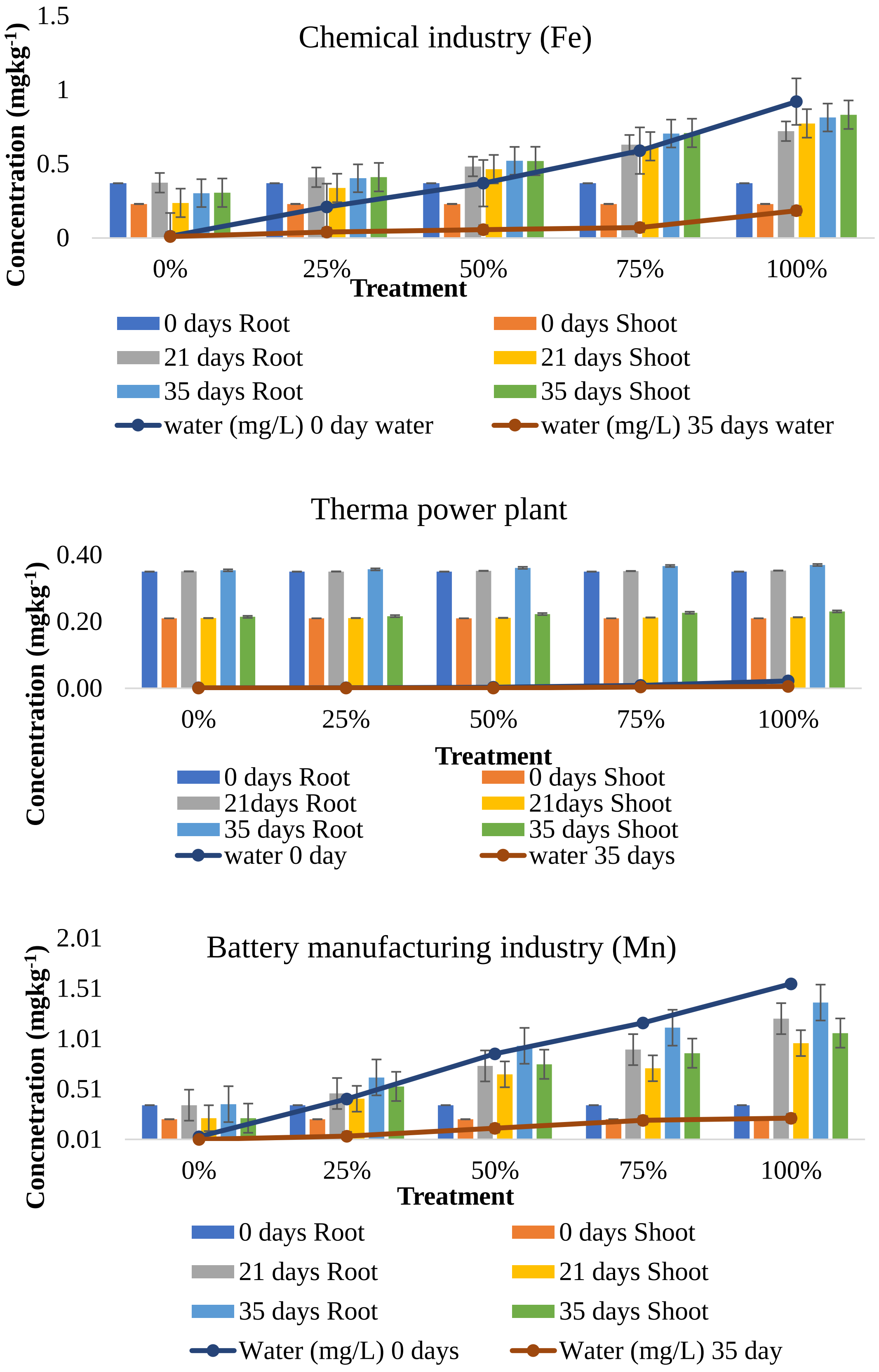
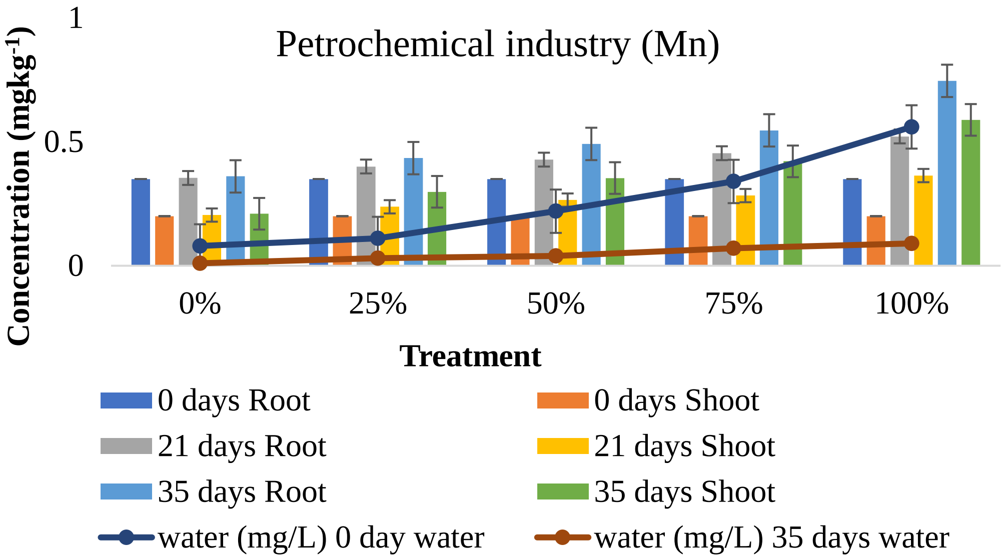
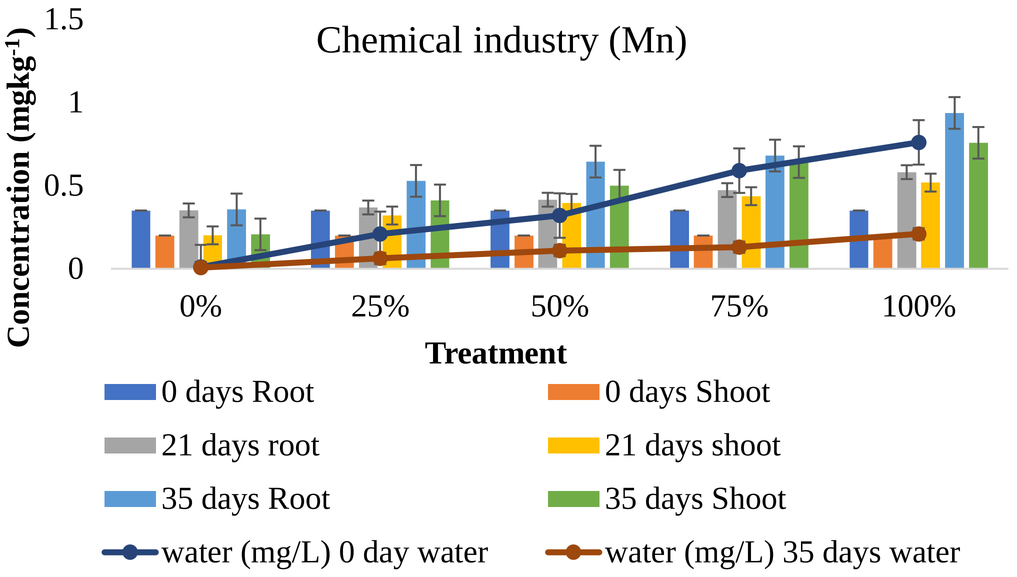


Fig. 4.3.5: Mn concentration in *L. gibba* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

P. stratiotes accumulates 0.35 and 0.20 mgkg^{-1} Mn initially in root and shoot. After 21 days treatment its accumulates 0.35, 0.40, 0.43, 0.45, 0.52 mgkg^{-1} and 0.20, 0.24, 0.26, 0.28, 0.36 mgkg^{-1} and after 35 days its accumulates 0.21, 0.30, 0.35, 0.42, 0.59 mgkg^{-1} and 0.21, 0.30, 0.35, 0.42, 0.59 mgkg^{-1} in their root and shoot. In chemical industry effluent, its accumulates 0.35, 0.37, 0.42, 0.47, 0.58 mgkg^{-1} and 0.20, 0.32, 0.40, 0.44, 0.52 mgkg^{-1} after 21 days and 0.36, 0.53, 0.64, 0.68, 0.94 and 0.21, 0.41, 0.50, 0.64, 0.76 mgkg^{-1} after 35 days in their root and shoot. From thermal power plant effluent, *P. stratiotes* absorbed 0.41, 0.43, 0.43, 0.45, 0.49 mgkg^{-1} and 0.19, 0.21, 0.22, 0.22, 0.26 mgkg^{-1} and after 35 days 0.41, 0.50, 0.55, 0.61, 0.69 and 0.20, 0.28, 0.31, 0.36, 0.47 mgkg^{-1} in root and shoot respectively. From battery manufacturing industry its accumulates 0.36, 0.58, 0.72, 0.96, 1.35 mgkg^{-1} and 0.23, 0.42, 0.64, 0.82, 0.89 mgkg^{-1} after 21 days and 0.36, 0.79, 0.81, 1.12, 1.64 mgkg^{-1} and 0.23, 0.56, 0.73, 0.97, 0.77 mgkg^{-1} Mn after 35 days in root and shoot of *P. stratiotes*. Mn concentration in shoot of *P. stratiotes* was decrease after 35 days, it's may be due to decomposition of plants.





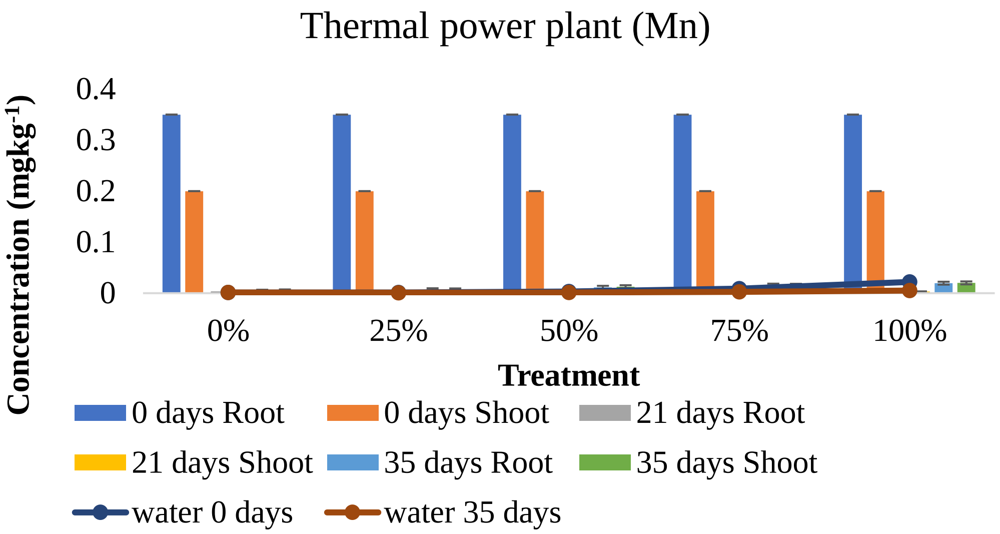
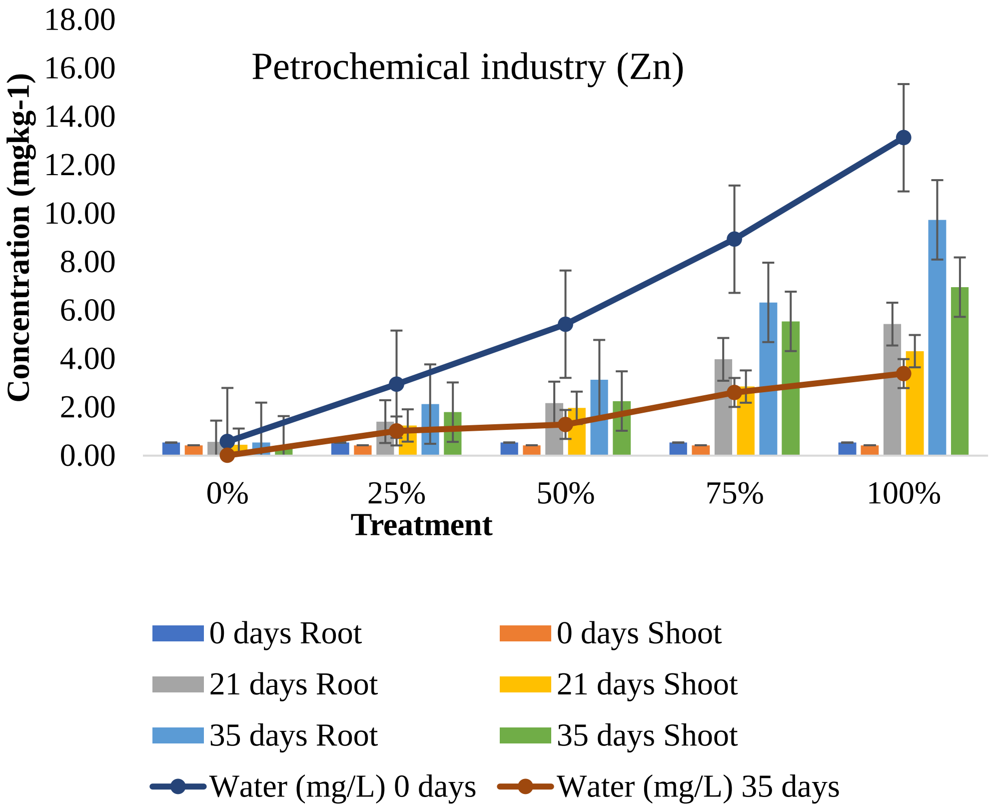


Fig 4.3.6: Mn concentration in *P. stratiotes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

In *E. crassipes* initial concentration of Zn was found to be 0.54 and 0.43 mgkg^{-1} in root and shoot respectively. Zn concentration after exposure for 21 days was found 0.56, 1.41, 2.17, 3.98, 5.43 and 0.45, 1.25, 1.98, 2.85, 4.32 mgkg^{-1} and 0.55, 2.13, 3.14, 6.33, 9.74 and 0.41, 1.80, 2.26, 5.55, 6.96 mgkg^{-1} Zn in root and shoot after 35 days of exposure with 0, 25, 50, 75 and 100% petrochemical industry effluent concentration respectively. In exposure of chemical industry effluent, *Eichhornia* plant accumulates 0.54, 0.79, 1.01, 1.48, 1.79 and 0.44, 0.68, 0.95, 1.40, 1.54 mgkg^{-1} after 21 days and 0.57, 1.02, 1.31, 1.90, 2.01 and 0.46, 0.90, 1.19, 1.77, 1.88 mgkg^{-1} after 35 days in their root and shoot. In exposure of thermal power plant effluent, Zn concentration in this macrophyte was found to be 0.54, 0.56, 0.60, 0.68, 0.87 and 0.43, 0.45, 0.50, 0.56, 0.69 mgkg^{-1} after 21 days and 0.55, 0.72, 0.84, 1.04, 1.19 and 0.44, 0.64, 0.74, 0.93, 1.06 mgkg^{-1} after 35 days in root and shoot from 0, 25, 50, 75 and 100% concentrated effluent. In battery manufacturing industry effluent, it accumulates 0.54, 0.61, 0.81, 1.27, 1.57 and 0.46, 0.51, 0.75, 0.98, 1.42 mgkg^{-1} in root and shoot after 21 days but after 35 days its accumulates 0.53, 0.72, 0.94, 1.43, 1.74 and 0.43, 0.60, 0.83, 1.05 and 1.50 mgkg^{-1} in root and shoot respectively.



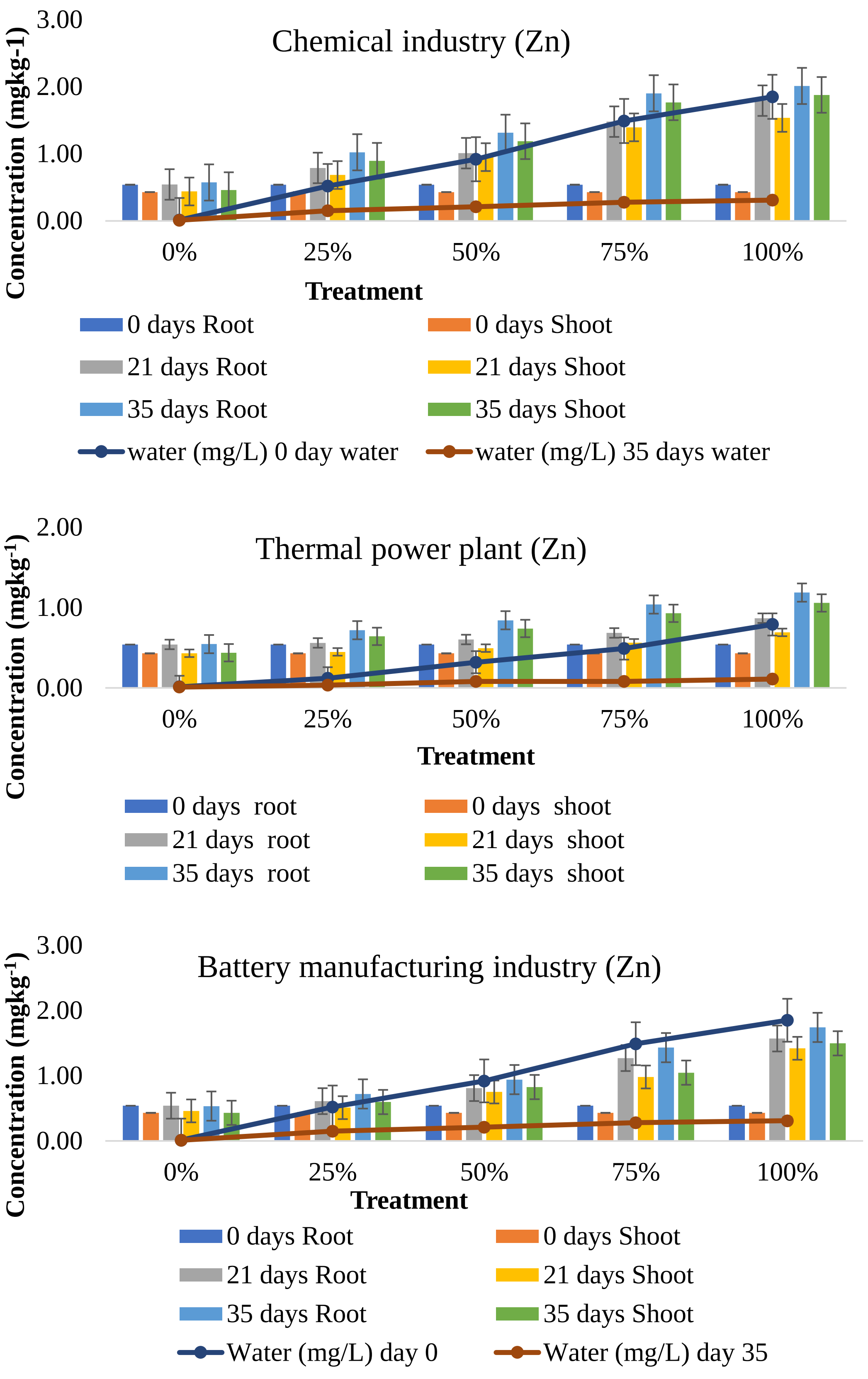
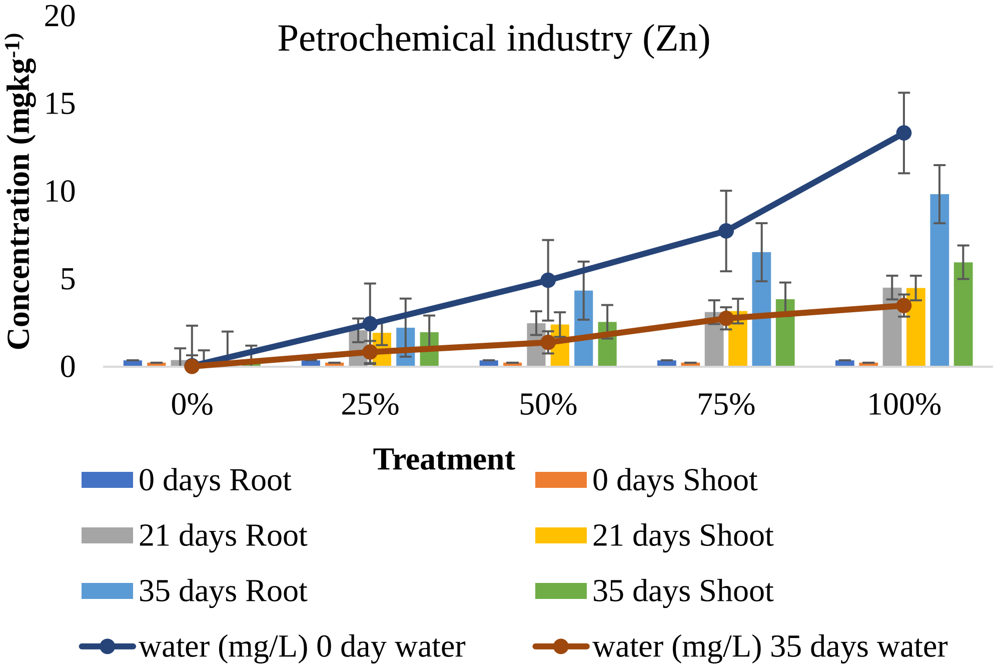


Fig. 4.3.7: Zn concentration in *E. crassipes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

L. gibba initially accumulates 0.37 and 0.24 mgkg^{-1} Zn in their roots and shoot respectively. After 21 days of treatment with petrochemical wastewater its accumulates 0.37, 2.08, 2.49, 3.11, 4.52 mgkg^{-1} Zn in root 0.23, 1.94, 2.40, 3.17, 4.49 mgkg^{-1} Zn in shoot and after 35 days of treatment its accumulates 0.35, 2.23, 4.34, 6.53, 9.84 mgkg^{-1} in root and 0.25, 1.96, 2.56, 3.85, 5.96 mgkg^{-1} in shoot from 0, 25, 50, 75, 100% effluent concentration. In chemical industry effluent its accumulates 0.37, 1.00, 1.26, 1.58, 1.74 mgkg^{-1} 0.23, 0.84, 1.09, 1.41, 1.51 mgkg^{-1} after 21 days and after 35 days its accumulate 0.37, 1.08, 1.37, 1.78, 2.14 mgkg^{-1} and 0.24, 0.95, 1.24, 1.61, 2.02 mgkg^{-1} from 0, 25, 50, 75 and 100% wastewater. In thermal power plant effluent exposure, its accumulates 0.37, 0.39, 0.42, 0.45, 0.47 mgkg^{-1} and 0.23, 0.25, 0.28, 0.31, 0.33 mgkg^{-1} after 21 days, 0.38, 0.44, 0.53, 0.62, 0.66 mgkg^{-1} and 0.24, 0.30, 0.38, 0.48, 0.53 mgkg^{-1} after 35 days of exposure. From battery manufacturing industry its accumulates 0.35, 0.47, 0.74, 0.90, 1.21 mgkg^{-1} 0.22, 0.41, 0.66, 0.72, 0.97 mgkg^{-1} after 21 days and 0.36, 0.63, 0.94, 1.12, 1.37 mgkg^{-1}, 0.22, 0.54, 0.76, 0.87, 1.07 mgkg^{-1} after 35 days.



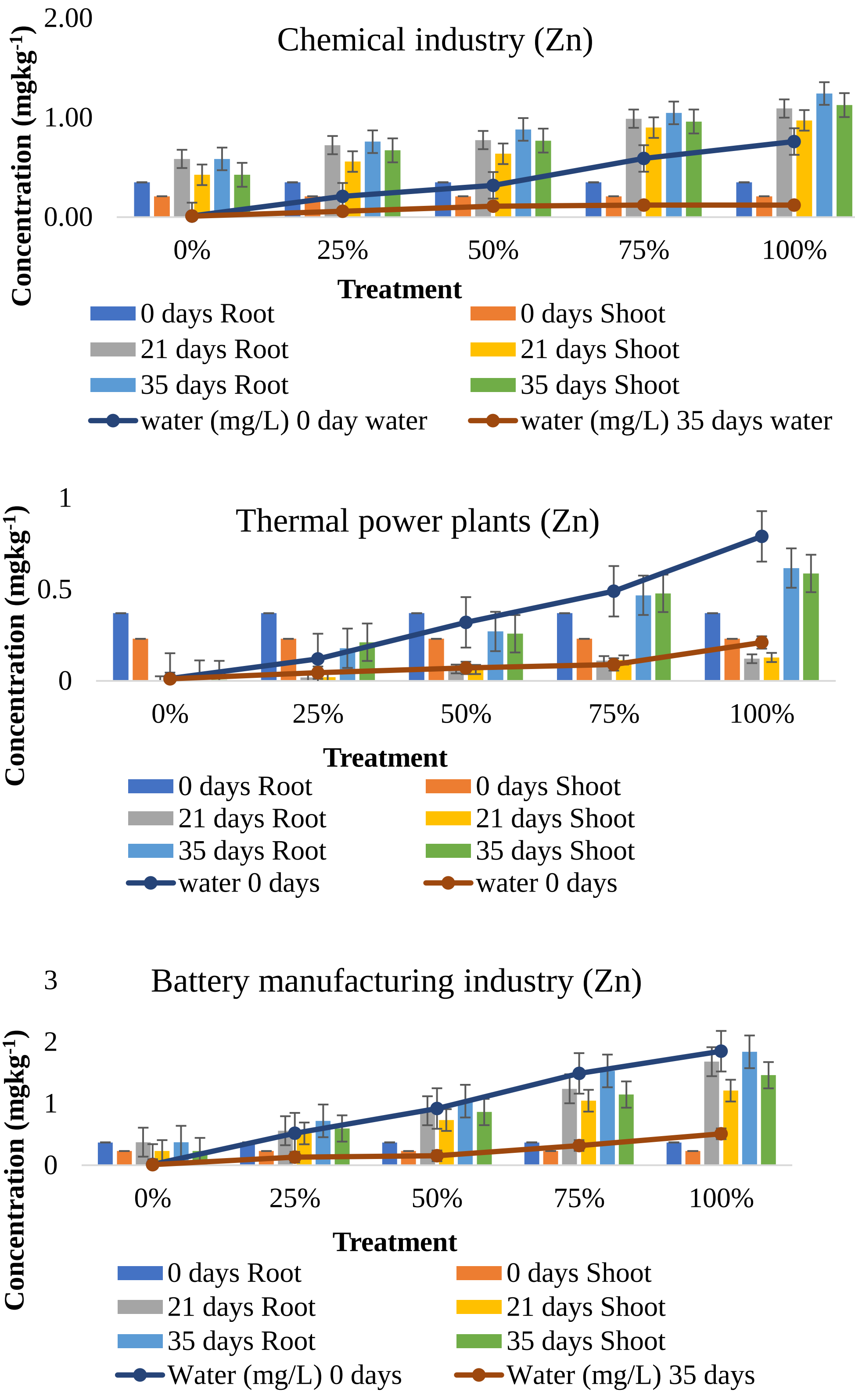
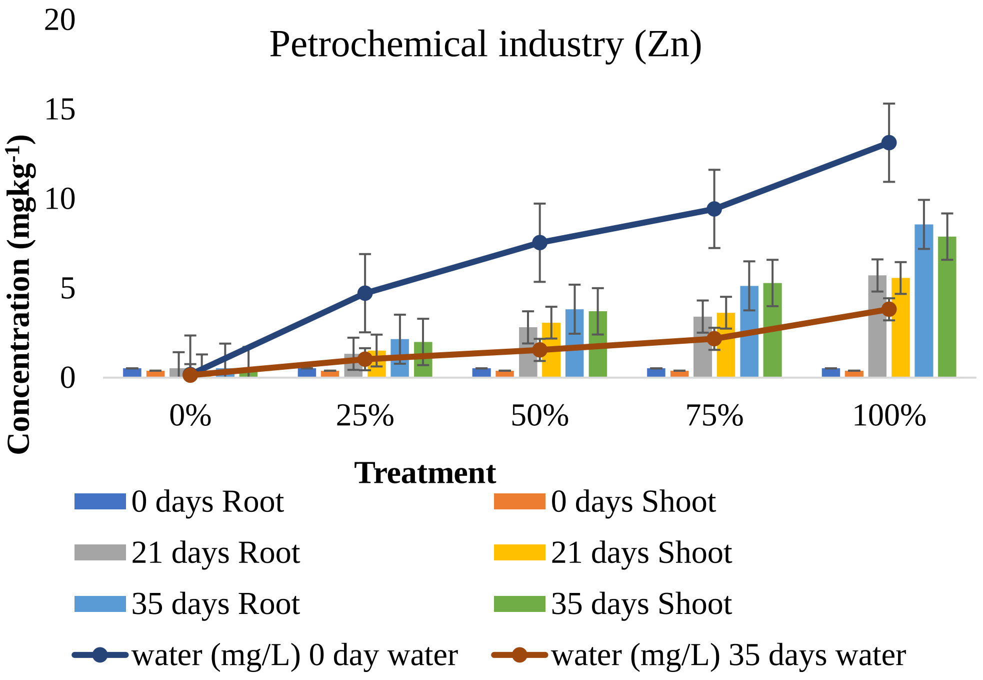
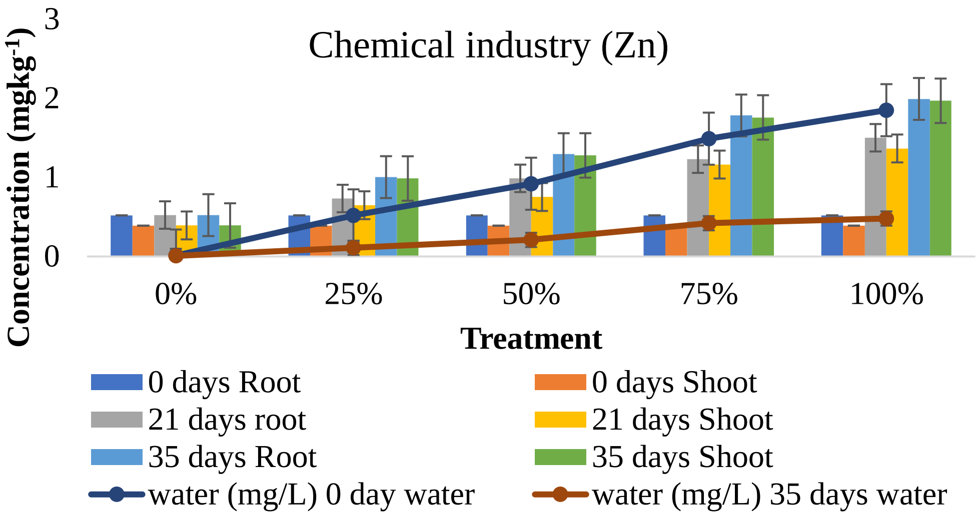


Fig 4.3.8: Zn concentration in *L. gibba* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

In *P. stratiotes*, 0.52 and 0.39 mgkg^{-1} Zn initially accumulated in root and shoot. After 21 days of treatment its accumulates 0.52, 1.33, 2.81, 3.41, 5.71 mgkg^{-1} and 0.40, 1.52, 3.07, 3.07, 3.63, 5.57 mgkg^{-1} Zn and After 35 days if treatment its accumulates 0.53, 2.15, 3.83, 5.13, 8.57 mgkg^{-1} and 0.41, 2.00, 3.17, 5.29, 7.88 mgkg^{-1} Zn in root and shoot at 0, 25, 50, 75 and 100% effluent concentration. Greger (1999) reported uptake of Zn by root and shoot of plant was increase with increasing metal concentration in medium and accumulation was linear correlated to the effluent concentration. From chemical industry effluent its accumulates 0.52, 0.73, 0.99, 1.23, 1.50 mgkg^{-1} and 0.39, 0.65, 0.75, 1.16, 1.37 mgkg^{-1} Zn after 21 days and 0.52, 1.00, 1.29, 1.78, 1.99 mgkg^{-1} and 0.39, 0.99, 1.28, 1.76, 1.97 mgkg^{-1} Zn after 35 days. In thermal power plant *P. stratiotes* accumulates 0.53, 0.70, 0.82, 1.02, 1.17 mgkg^{-1} 1 and 0.40, 0.60, 0.70, 0.89, 1.02 mgkg^{-1} in root and shoot after 35 days of experiment. From battery manufacturing industry its absorbed 0.54, 0.68, 0.83, 1.23, 1.36 mgkg^{-1} and 0.40, 0.49, 0.74, 0.89 mgkg^{-1} after 21 days and 0.53, 0.76, 0.91, 1.43, 1.64 mgkg^{-1} and 0.39, 0.56, 0.82, 1.05, 1.50 mgkg^{-1} after 35 days in root and shoot respectively.





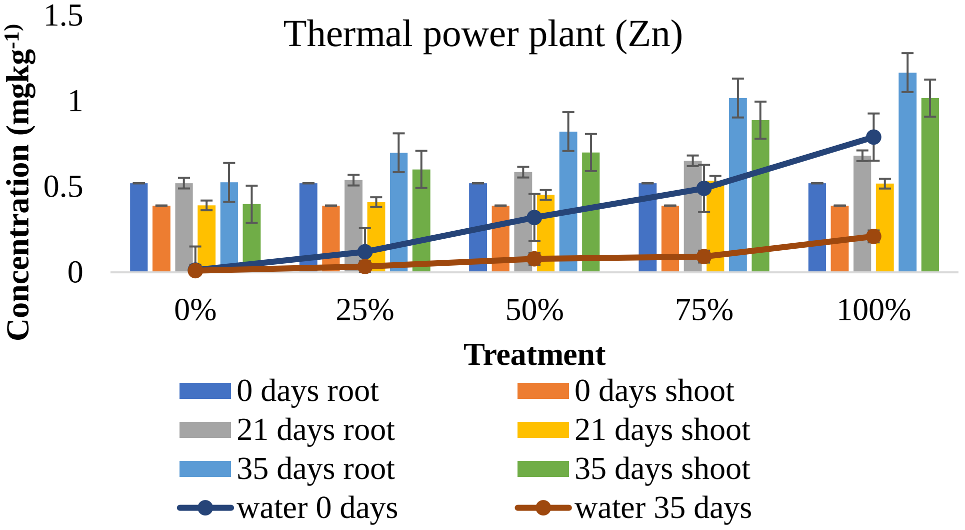
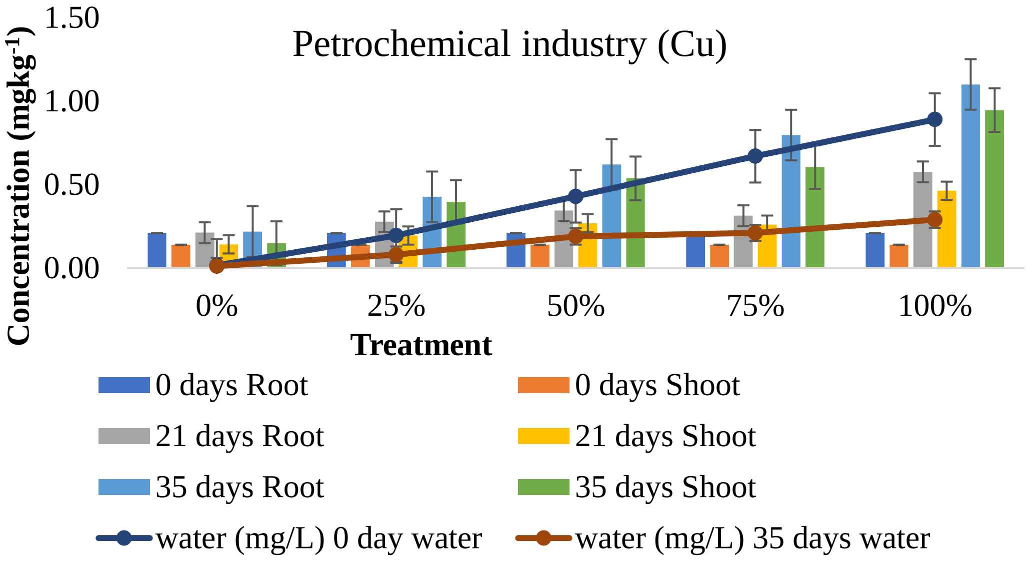


Fig. 4.3.9: Zn concentration in *P. stratiotes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

Cu is another essential metal for plants metabolism playing important role metalloenzymes, photosynthesis related membrane structures, also reported to be toxic metal (Li and Xiong, 2004). Its initial concentration in *E. crassipes* was found to be 0.21 and 0.14 mgkg^{-1} in their root and shoot respectively. After 21 days, exposure of petrochemical effluent, its accumulates 0.21, 0.28, 0.35, 0.31, 0.58 and 0.14, 0.19, 0.27, 0.26, 0.46 mgkg^{-1} in root and shoot of plant. After 35 days of exposure Cu was found to be 0.22, 0.43, 0.62, 0.80, 1.10 mgkg^{-1} and 0.15, 0.40, 0.54, 0.61 and 0.95 mgkg^{-1} in root and shoot respectively. At 100 and 75% effluent concentration, accumulation was low in comparison to 50 and 25 % effluent so we can say that accumulation was maximum at lower effluent concentration. In chemical industry effluent *Eichhornia* accumulates 0.21, 0.24, 0.25, 0.27, 0.28 mgkg^{-1} Cu in root and 0.14, 0.18, 0.19, 0.20, 0.20 mgkg^{-1} Cu in shoot, after 21 days and after 35 days Cu concentration increase, found to be 0.22, 0.25, 0.26, 0.29, 0.29 and 0.15, 0.18, 0.19, 0.22, 0.22 mgkg^{-1} in root and shoot respectively. In thermal power plant effluent, this macrophyte accumulates 0.54, 0.56, 0.60, 0.68, 0.87 mgkg^{-1} in root and 0.43, 0.45, 0.50, 0.56, 0.69 mgkg^{-1} in shoot after 21 days and 0.55, 0.72, 0.84, 1.04, 1.19 and 0.44, 0.64, 0.74, 0.93, 1.06 mgkg^{-1} after 35 days of exposure in their root and shoot respectively. In case of battery manufacturing effluent exposure to plant, it accumulates 0.54, 0.61, 0.81, 1.27, 1.57 mgkg^{-1} and 0.46, 0.51, 0.75, 0.98, 1.42 mgkg^{-1} after 21 days and 0.53, 0.72, 0.94, 1.43, 1.74 mgkg^{-1} and 0.43, 0.60, 0.83, 1.05, 1.50 mgkg^{-1} after 35 days.



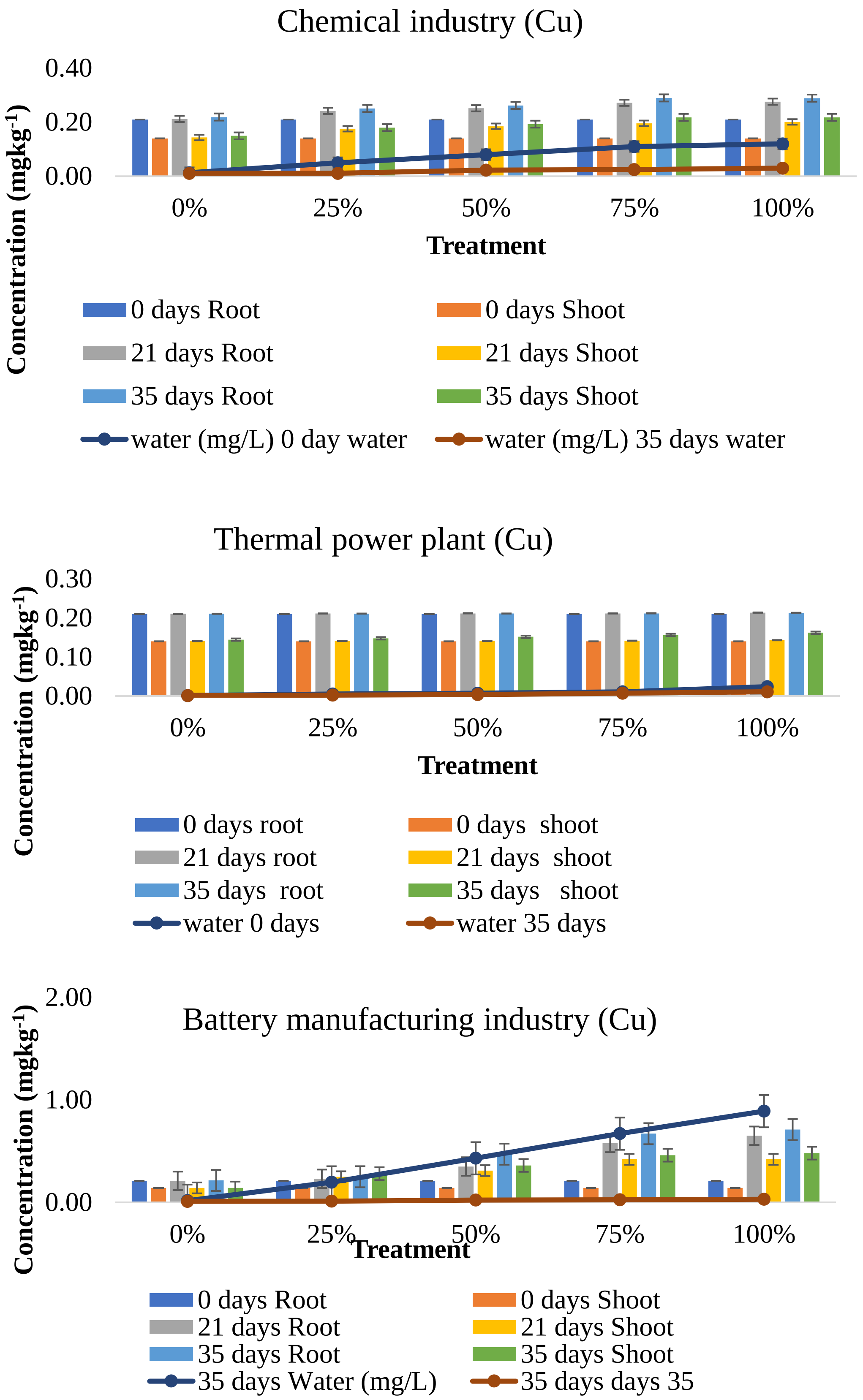
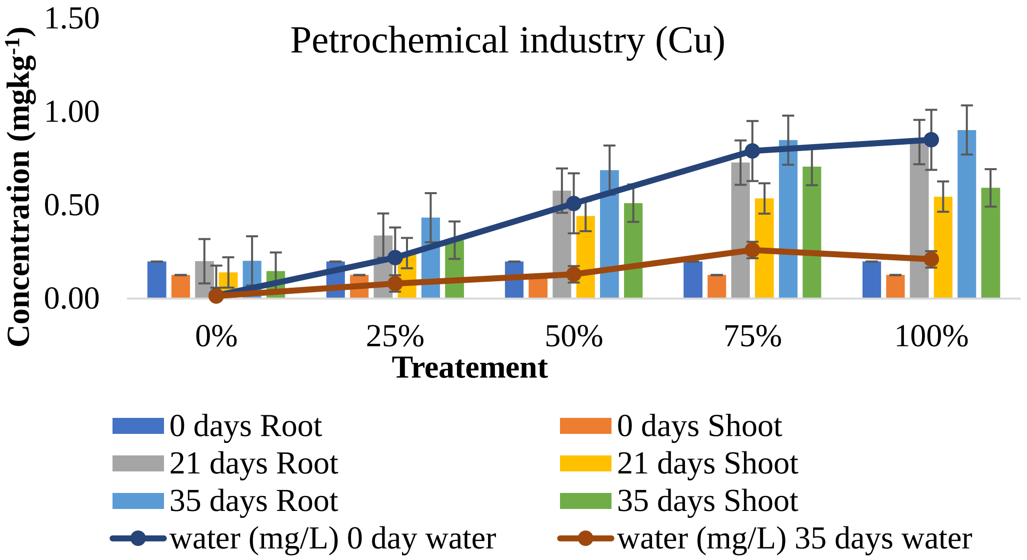
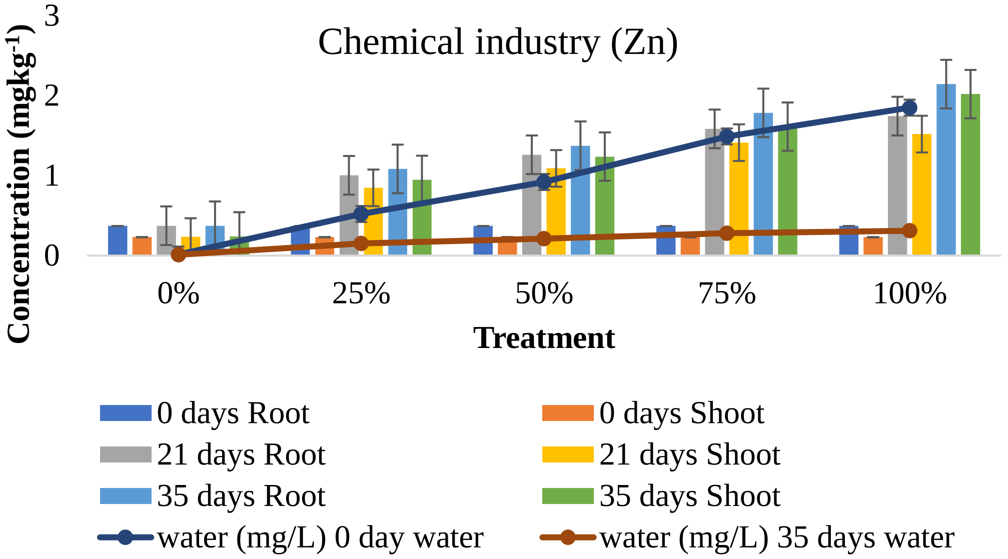
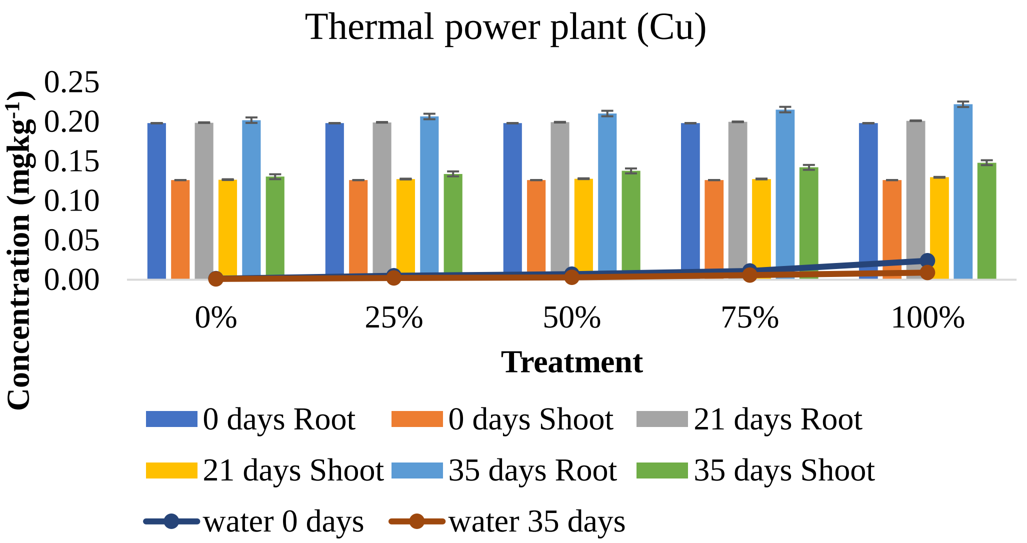


Fig 4.3.10: Cu concentration in *E. crassipes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

Initial concentration of Cu in *L. gibba* was found to be 0.20 and 0.13 in root and shoot respectively. After 21 Cu increases up to 0.00, 0.24, 0.49, 0.69, 0.70 mgkg^{-1} in root and 0.02, 0.19, 0.38, 0.58, 0.47 mgkg^{-1} in shoot and after 35 days it's found 0.00, 0.14, 0.38, 0.53, 0.64 mgkg-1 in root and 0.01, 0.12, 0.32, 0.41, 0.42 mgkg^{-1} shoot at 0, 25, 50, 75, 100% petrochemical effluent concentration. After 21 days Cu concentration was found 0.21, 0.21, 0.23, 0.25, 0.26 mgkg^{-1} and 0.13, 0.14, 0.16, 0.18, 0.19 mgkg^{-1} in root and shoot respectively. Chemical industry has found very low Cu concentration so, that after 35 days not much accumulation occurred found 0.21, 0.23, 0.24, 0.26, 0.27 mgkg^{-1} and 0.13, 0.16, 0.17, 0.19, 0.19 mgkg^{-1} in root and shoot respectively at 0, 25, 50, 75, 100%. Similarly, in thermal power plants effluent found very low Cu i.e. 0.02 mgl^{-1} so, accumulation was low. From battery manufacturing industry effluent *L. gibba* accumulates 0.20, 0.25, 0.37, 0.62, 0.78 mgkg^{-1} in root, and 0.14, 0.19, 0.24, 0.40, 0.52 mgkg^{-1} in shoot after 21 days and after 35 days it found 0.21, 0.31, 0.53, 0.78, 0.91 mgkg^{-1} in root and 0.14, 0.22, 0.36, 0.58, 0.66 mgkg^{-1} in shoot. Similar results revealed by Bokhari et al. (2016) they found increasing trend of accumulation of Cu in plant with increasing exposure time from sewage mixed industrial effluent. Initial copper concentration which is 2.0 mgkg^{-1} was increased to 7.13 mgkg^{-1}, 16.06 mgkg^{-1}, 24.4 mgkg^{-1}, 30.2 mgkg^{-1}, and 34.6 mgkg^{-1} at 3, 10, 17, 24, and 31 days of treatment respectively.


Petrochemical industry (Cu)
Concentration (mgkg-1)
1.50
1.00
0.50
0.00
0%
25%
50%
75%
100%
Treatement
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Chemical industry (Zn)
Concentration (mgkg-1)
3
2
1
0
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Thermal power plant (Cu)
Concentration (mgkg-1)
0.25
0.20
0.15
0.10
0.05
0.00
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water 0 days
water 35 days

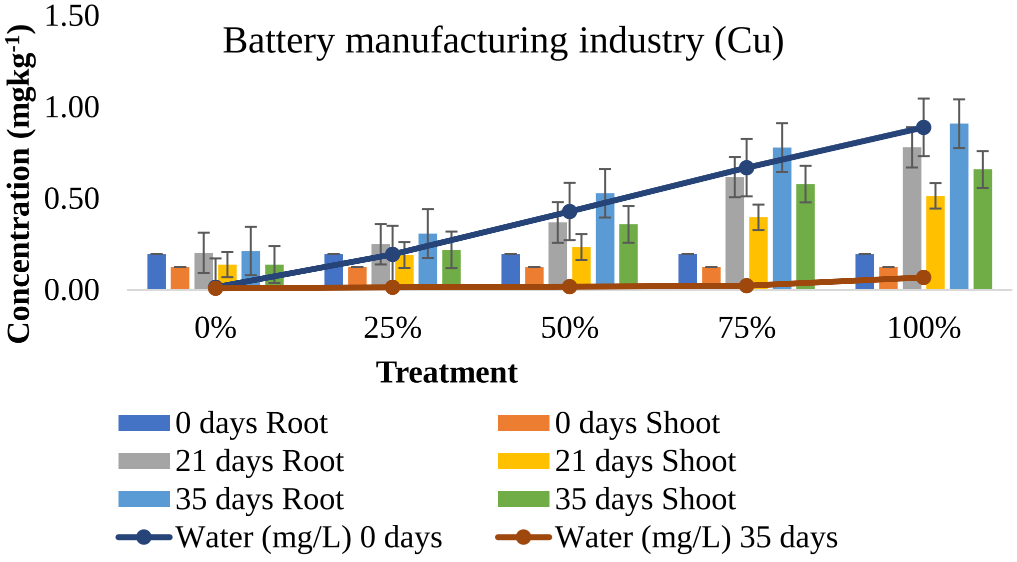
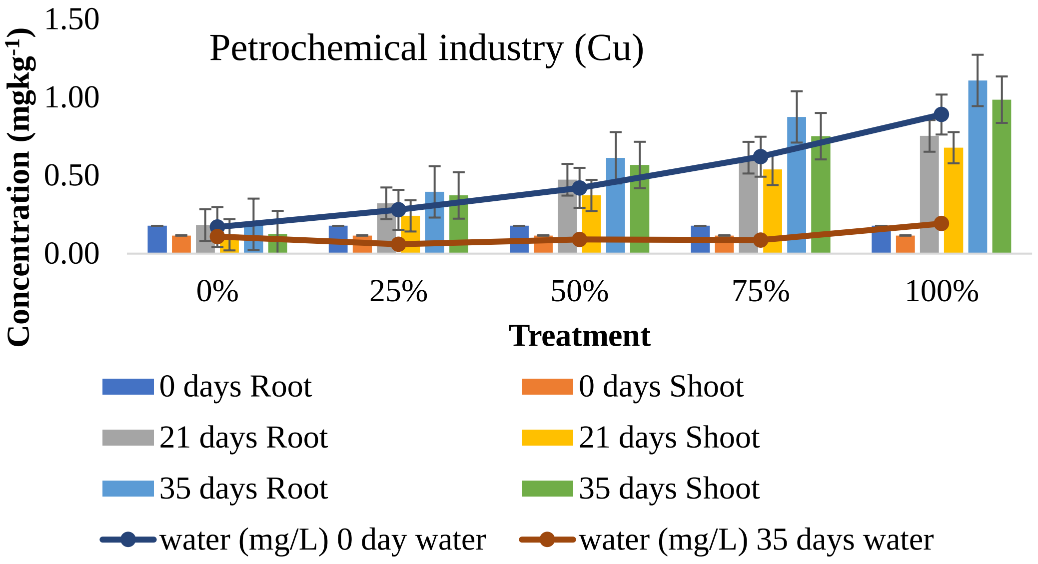
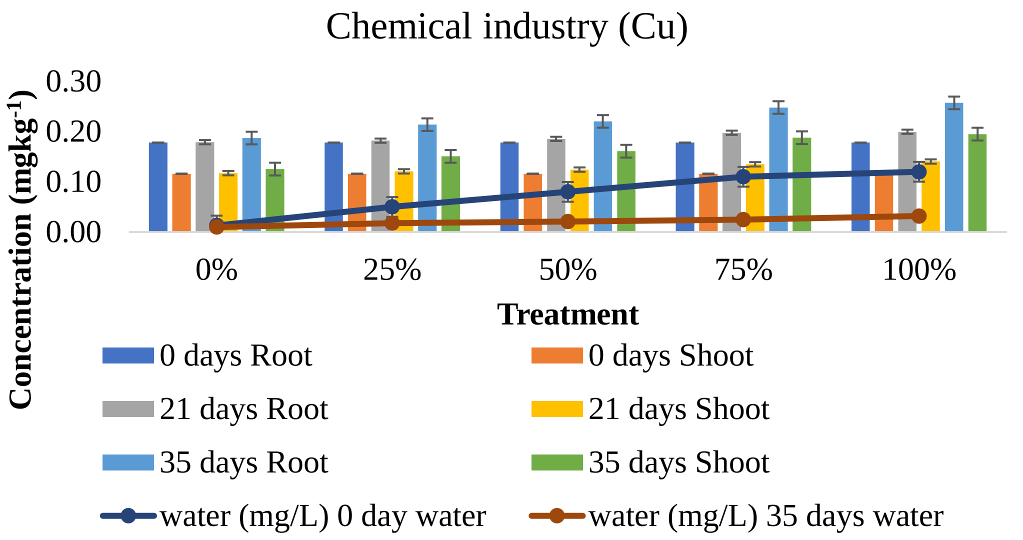
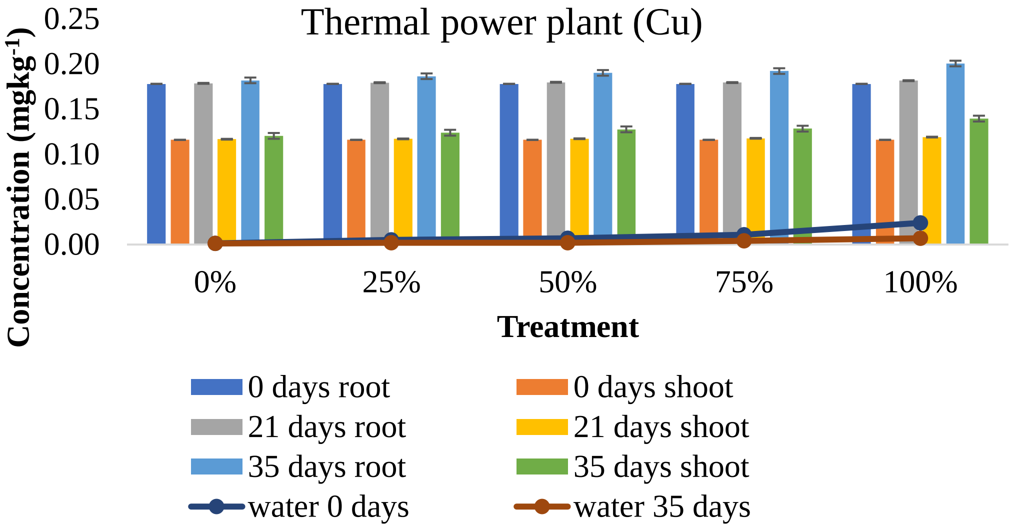


Fig. 4.3.11: Cu concentration in *L. gibba* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

Initially *P. stratiotes* accumulates 0.18 and 0.12 in root and shoot. After 21 days its 0.18, 0.32, 0.47, 0.61, 0.75 mgkg^{-1} and 0.12, 0.24, 0.37, 0.54, 0.68 mgkg^{-1} Cu and after 35 days its accumulates 0.19, 0.40, 0.61, 0.87, 1.11 mgkg^{-1} and 0.13, 0.37, 0.57, 0.75, 0. mgkg^{-1}. Cu from petrochemical industry effluent. From chemical industry effluent its accumulates 0.19, 0.21, 0.22, 0.25, 0.26 mgkg^{-1} and 0.13, 0.15, 0.16, 0.19, 0.20 mgkg-^{1} Cu in root and shoot after 35 days of treatment. In thermal power plant this macrophyte accumulates 0.18, 0.19, 0.19, 0.19, 0.20 mgkg^{-1} and 0.12, 0.12, 0.13, 0.13, 0.14 mgkg^{-1} after 35 days of exposure. From battery manufacturing industry *P. stratiotes* absorbed 0.19, 0.21, 0.39, 0.44, 0.63 mgkg^{-1} and 0.13, 0.21, 0.27, 0.37, 0.47 mgkg^{-1} Cu after 21 days and 0.19, 0.23, 0.45, 0.58, 0.71 mgkg^{-1} and 0.13, 0.21, 0.31, 0.47, 0.58 mgkg^{-1} Cu after 35 days in root and shoot at 0, 25, 50, 75 and 100% concentrated effluents.


Petrochemical industry (Cu)
Concentration (mgkg^{-1})
1.50
1.00
0.50
0.00
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Chemical industry (Cu)
Concentration (mgkg^{-1})
0.30
0.20
0.10
0.00
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Thermal power plant (Cu)
Concentration (mgkg^{-1})
0.25
0.20
0.15
0.10
0.05
0.00
0%
25%
50%
75%
100%
Treatment
0 days root
0 days shoot
21 days root
21 days shoot
35 days root
35 days shoot
water 0 days
water 35 days

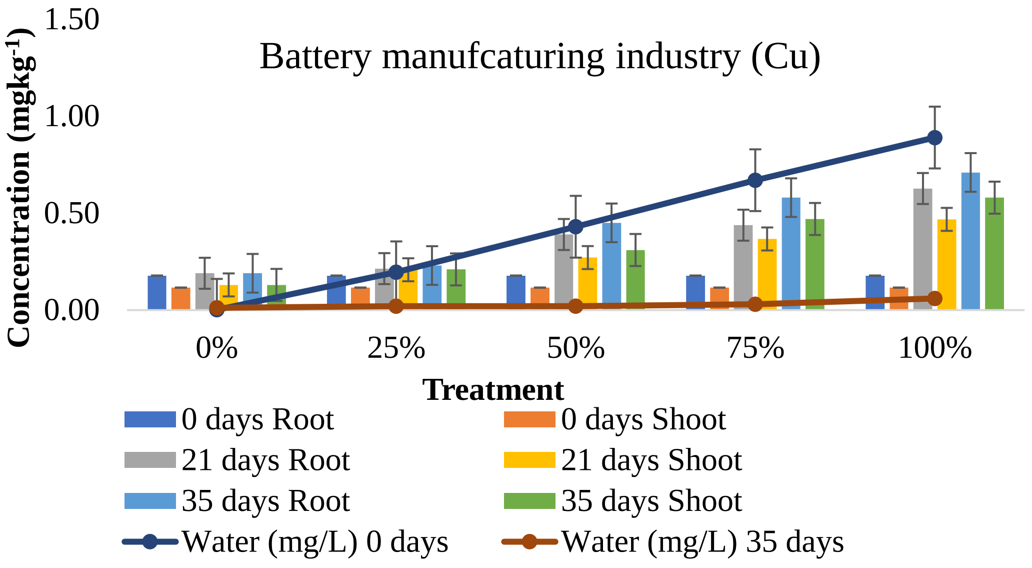


Fig. 4.3.12: Cu concentration in *P. stratiotes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

Pb is toxic and non-essential metal for plants. Initial concentration of Pb was found to be 0.10 and 0.07 mgkg^{-1} in root and shoot of the plant. After 21 days of exposure *E. crassipes* accumulates 0.10, 0.12, 0.12, 0.14, 0.17 mgkg^{-1} in root, 0.07, 0.08, 0.09, 0.10, 0.15 mgkg^{-1} shoot and after 35 days of exposure its accumulates 0.11, 0.19, 0.23, 0.29, 0.48 and 0.08, 0.17, 0.21, 0.29, 0.41 mgkg^{-1} in root and shoot from 0, 25, 50, 75 and 100% concentrated effluent. In battery manufacturing industry effluent exposure Pb recorded as 0.02, 0.32, 0.86, 1.21, 1.39 mgkg^{-1} in root and 0.02, 0.29, 0.75, 0.83, 1.25 mgkg^{-1} in shoot and after 35 days it was recorded as 0.03, 0.42, 1.07, 1.34, 1.77 mgkg^{-1} in root and 0.02, 0.31, 0.85, 0.94, 1.37 mgkg^{-1} in shoot.

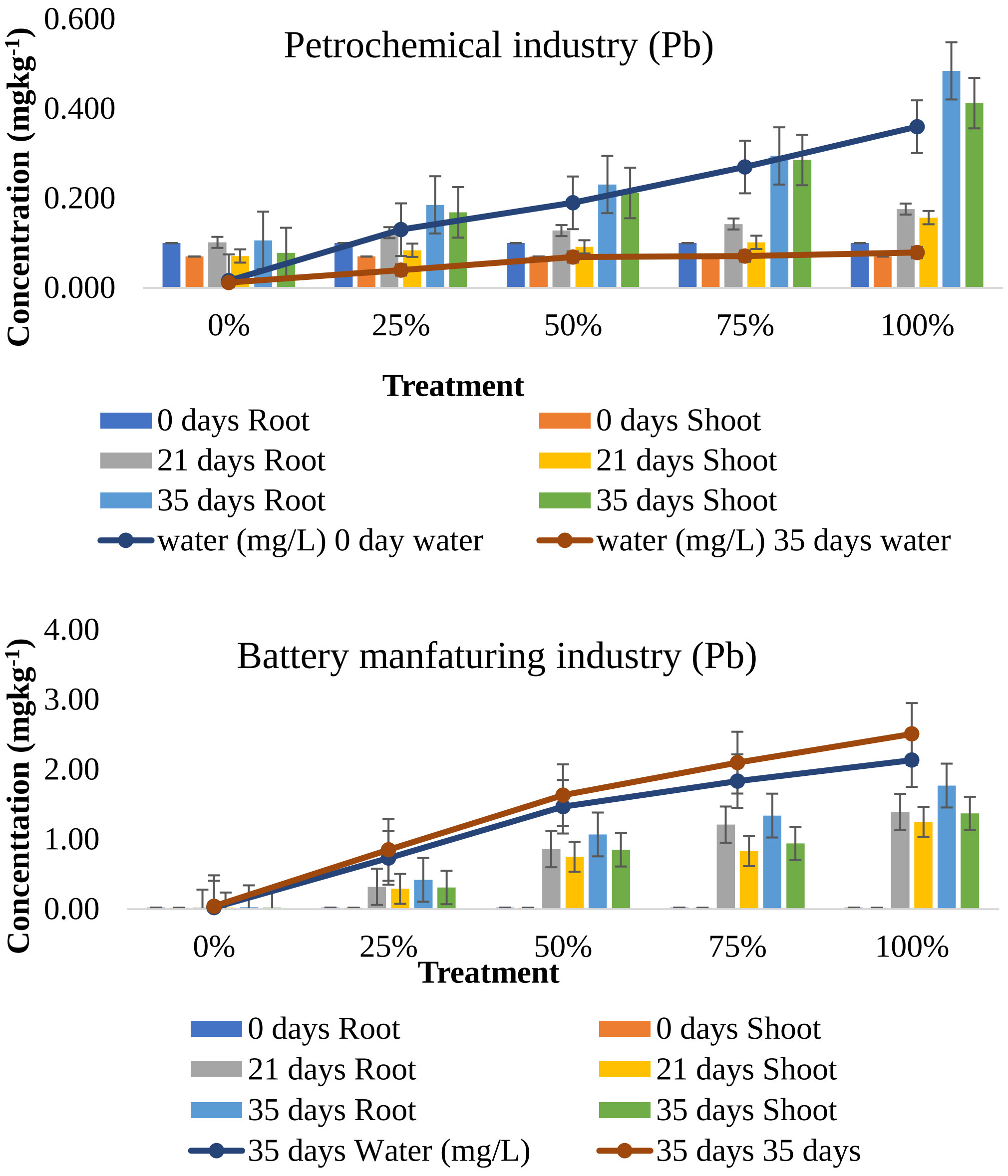
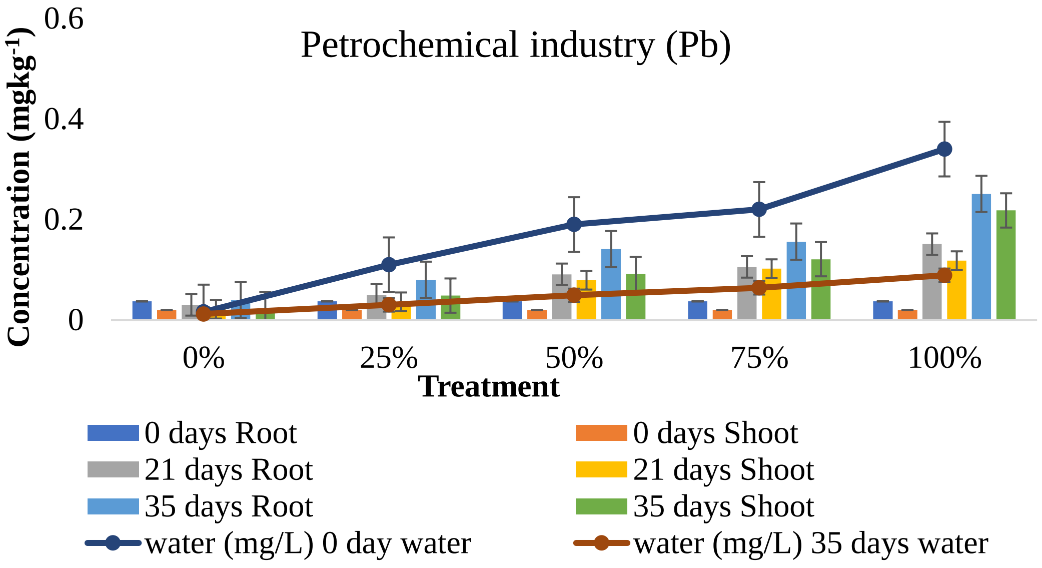


Fig. 4.3.13: Pb concentration in *E. crassipes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of petrochemical and battery manufacturing industries.

Initially Pb was found 0.03 and 0.02 in root and shoot of *L. gibba.* After 21 days accumulation was 0.03, 0.05, 0.09, 0.11, 0.15 mgkg^{-1} Pb and 0.02, 0.04, 0.08, 0.10, 0.12 mgkg^{-1} in root and shoot respectively. After 35 days it was found 0.04, 0.08, 0.14, 0.16, 0.25 mgkg^{-1}, 0.02, 0.05, 0.09, 0.12, 0.22 mgkg^{-1} Pb in root and root at 0, 25, 50, 75, 100% effluent respectively. From battery manufacturing industry effluent this macrophytes accumulates 0.03, 0.38, 0.87, 1.14, 1.26 mgkg^{-1} and 0.02, 0.25, 0.69, 0.81, 1.17 mgkg^{-1} in root and shoot respectively. After 35 days its accumulates 0.04, 0.52, 1.07, 1.34, 1.77 mgkg^{-1}, 0.02, 0.31, 0.85, 0.94, 1.22 mgkg^{-1} in root and shoot at 0, 25, 50, 75, 100% effluent respectively. Bhokhari et al. (2016) present that *L. minor* accumulated high concentration of lead from two different type of effluent i.e. municipal sewage effluent and sewage mixed industrial effluent. Lead has the highest initial concentration (0.41 and 0.60 mgl^{-1}) in the effluents. Initial concentration of lead in the plant was 5.8 mgkg-^{1} and after treatment concentration was found 318 mgkg^{-1} after 24 days which is maximum from sewage mixed industrial effluent. In others effluent concentration *L. minor* accumulates 81.3 mgkg^{-1}, 158 mgkg^{-1} and 237.8 mgkg^{-1} on days 3, 10 and 17 days respectively. At municipal sewage, Lead accumulation by *L. minor* was 15.6 mgkg^{-1},16.13 mgkg-^{1}, 53.46 mgkg^{-1} and 103.6 mgkg^{-1} on days 3 and 17, 24 and 31 days respectively. In a study by Leblebici and Aksoy (2011) *L. minor* was exposed to different Pb concentrations (0, 5, 10, 25, and 50 mgl^{-1}) under laboratory conditions for a period of 1, 3, 5, and 7 days. Results showed that *L. minor* accumulated 561 mgkg^{-1} Pb at 50 mgl^{-1} concentration on day 7.



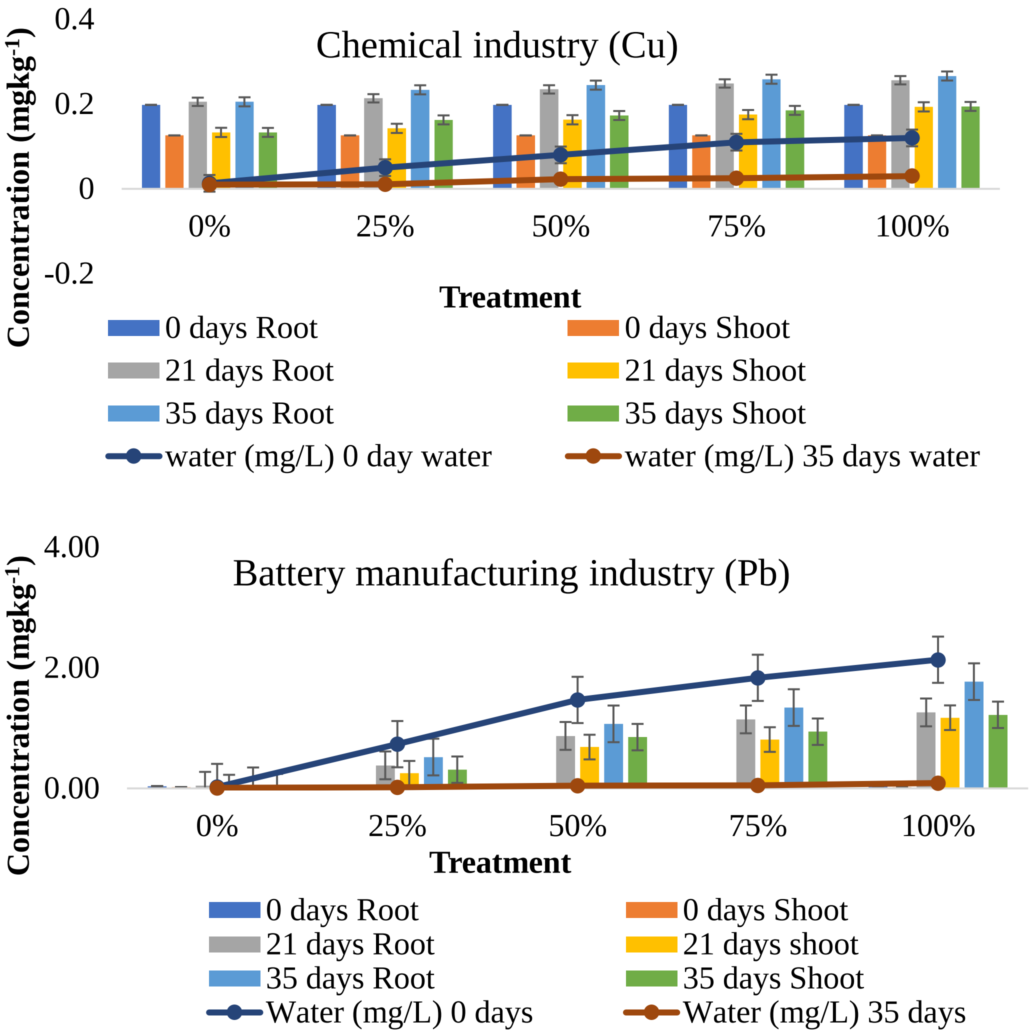
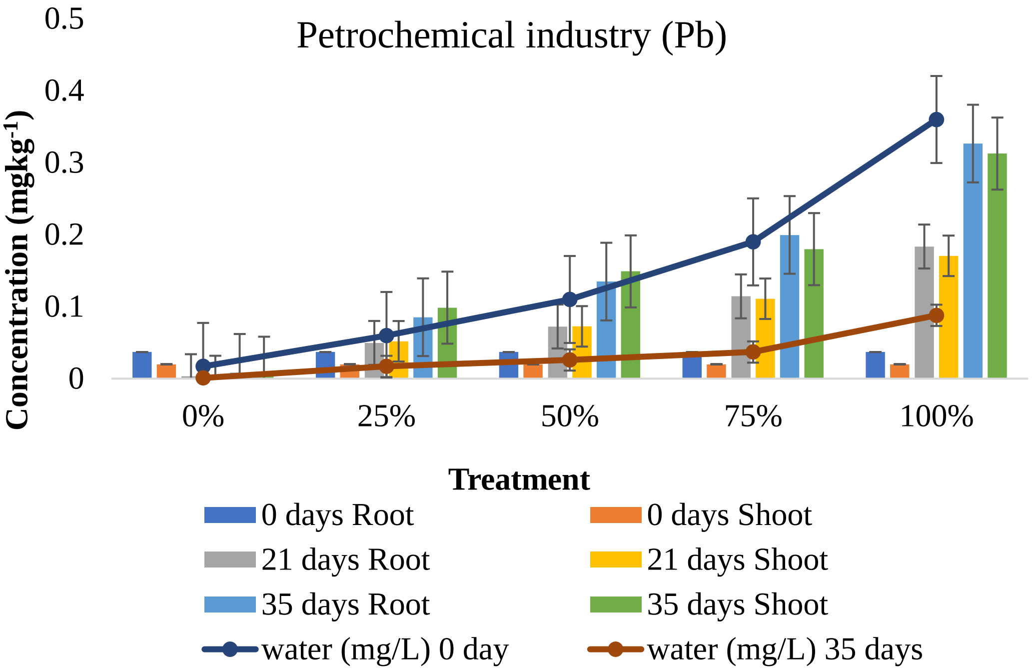


Fig. 4.3.14: Pb concentration in *L. gibba* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of petrochemical, chemical and battery manufacturing industries.

P. stratiotes accumulates 0.037, and 0.02 mgkg^{-1} Pb initially in root and shoot. After 21 days, its accumulates 0.00, 0.05, 0.07, 0.11, 0.18 mgkg^{-1} and 0.00, 0.05, 0.07, 0.11, 0.17 mgkg^{-1} Pb and after 35 days 0.01, 0.09, 0.13, 0.20, 0.33 and 0.01, 0.10, 0.15, 0.18, 0.31 mgkg^{-1} Cu in root and shoot respectively. From battery manufacturing industry *P. stratiotes* accumulates 0.03, 0.41, 0.51, 0.88, 1.26 mgkg^{-1} and 0.02, 0.29, 0.46, 0.74, 1.20 mgkg^{-1} after 21 days and 0.02, 0.47, 0.84, 1.28, 1.67 mgkg^{-1} and 0.02, 0.33, 0.61, 0.98, 1.27 mgkg^{-1} after 35 days.



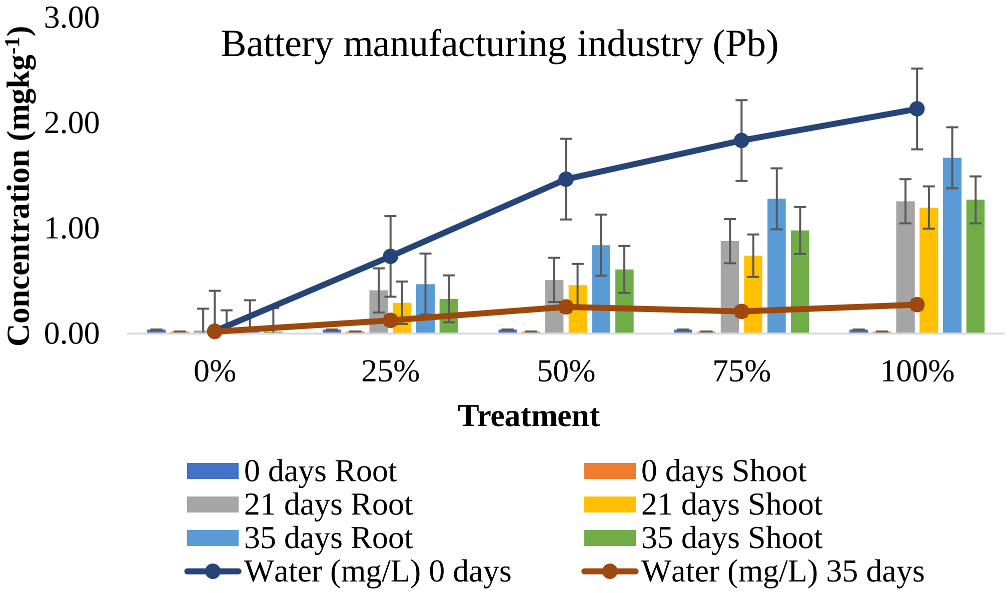


Fig. 4.3.15: Pb concentration in *P. stratiotes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of petrochemical and battery manufacturing industries.

Ni is essential trace metal for plant in low amount at higher amount cause significant effect in plants. Ni initially found to be 0.05 and 0.03 mgkg^{-1} in root and shoot of the *E. crassipes* and 0.33 mgl^{-1} in petrochemical industry effluent. After 21 days of exposure with petrochemical industry effluent *Eichhornia* accumulates 0.05, 0.07, 0.08, 0.09, 0.12 mgkg^{-1} in roots and 0.03, 0.05, 0.06, 0.07, 0.10 mgkg^{-1} in shoot and after 35 days of exposure 0.06, 0.13, 0.19, 0.25, 0.40 mgkg^{-1} in roots and 0.04, 0.12, 0.18, 0.23, 0.38 mgkg^{-1} in shoot.

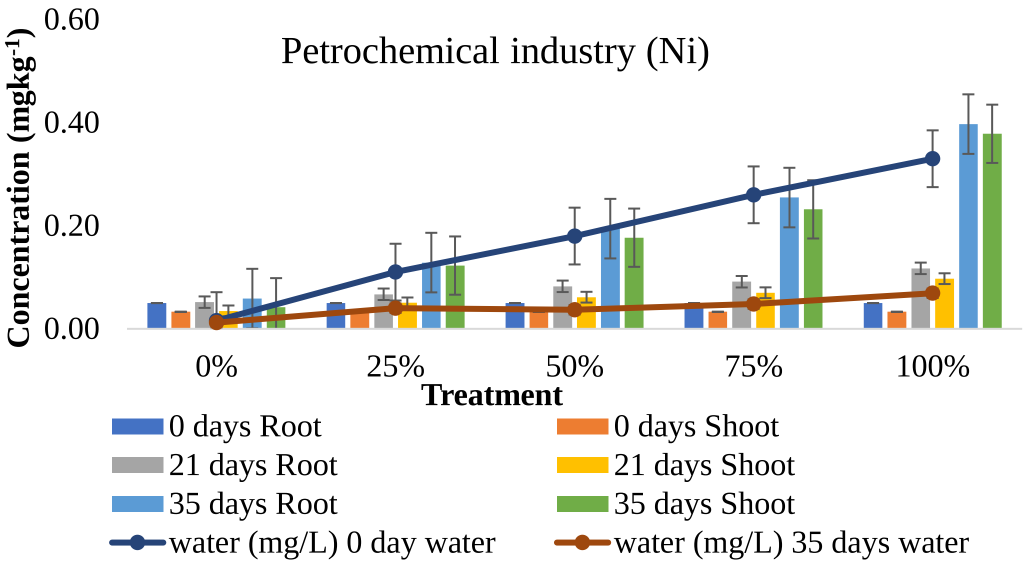


Fig 4.3.16: Ni concentration in *E. crassipes* in between root and shoot at 0, 21 and 21 days in 0, 25, 50, 75 and 100% effluent concentration of petrochemical industry.

L. gibba initially found 0.05 and 0.03 mgkg^{-1} Ni in their root and shoot respectively. After 21 days Ni concentration in root was found to be 0.05, 0.10, 0.14, 0.17, 0.25 mgkg^{-1} and in shoot 0.03, 0.09, 0.11, 0.14, 0.21 mgkg^{-1} and after 35 days it was found 0.05, 0.10, 0.19, 0.21, 0.31 mgkg^{-1} and 0.03, 0.11, 0.14, 0.16, 0.24 mgkg^{-1}. higher accumulation occurred at 100% effluent concentration. Similarly, Parinan et al. (2016) reported that Ni uptake value are higher in *L. gibba* than *Ceratophyllum demersum* and higher uptake attained at 6mgl^{-1} i.e. maximum Ni concentration.

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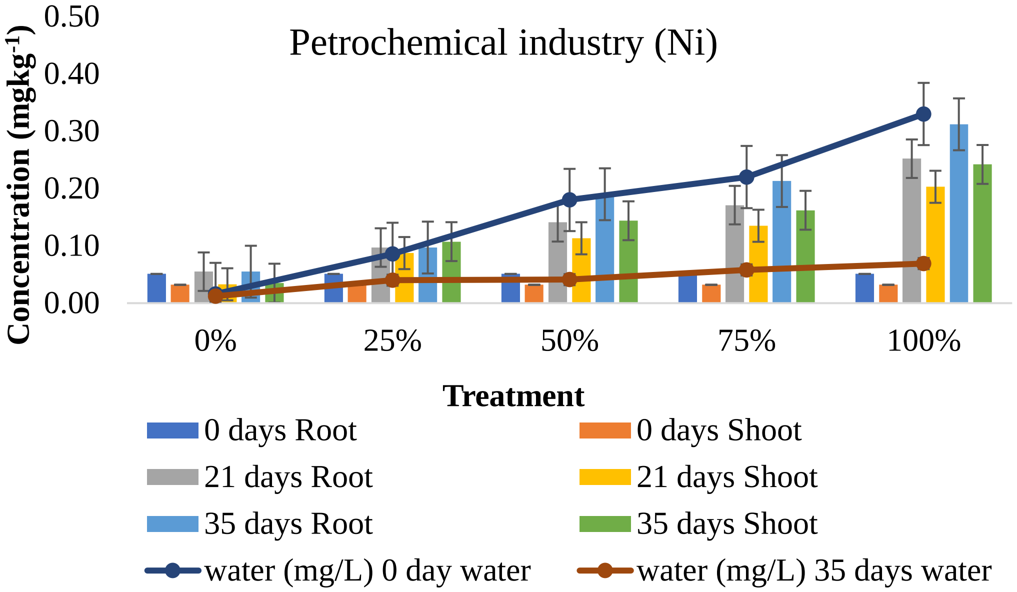


Fig 4.3.17: Ni concentration in *L. gibba* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of petrochemical industry.

P. stratiotes accumulates 0.05 and 0.03 mgkg^{-1} Ni initially in root and shoot. After 21 days of exposure its accumulates 0.06, 0.11, 0.14, 0.19, 0.27 mgkg^{-1} and 0.04, 0.08, 0.11, 0.14, 0.24 mgkg^{-1} Ni and after 35 days its accumulates 0.06, 0.13, 0.19, 0.25, 0.39 mgkg^{-1} and 0.04, 0.12, 0.17, 0.24, 0.37 mgkg^{-1} Ni in root and shoot of plant.

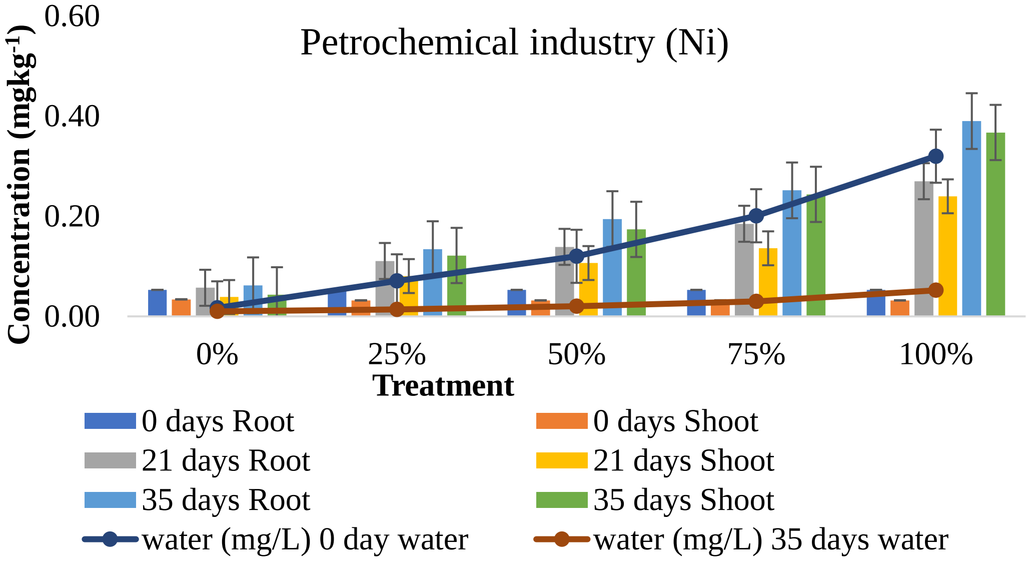
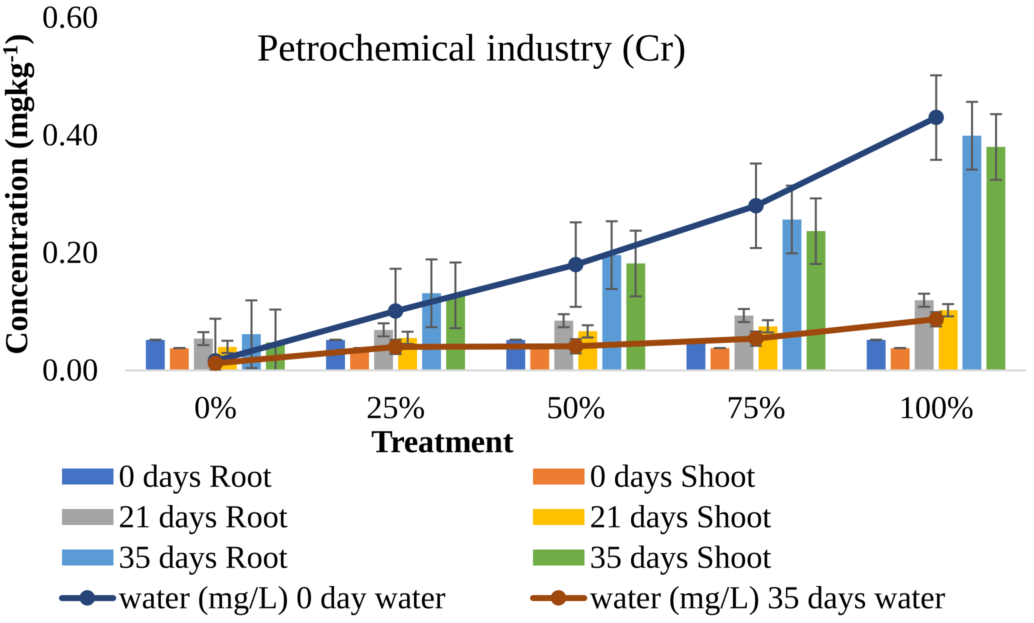
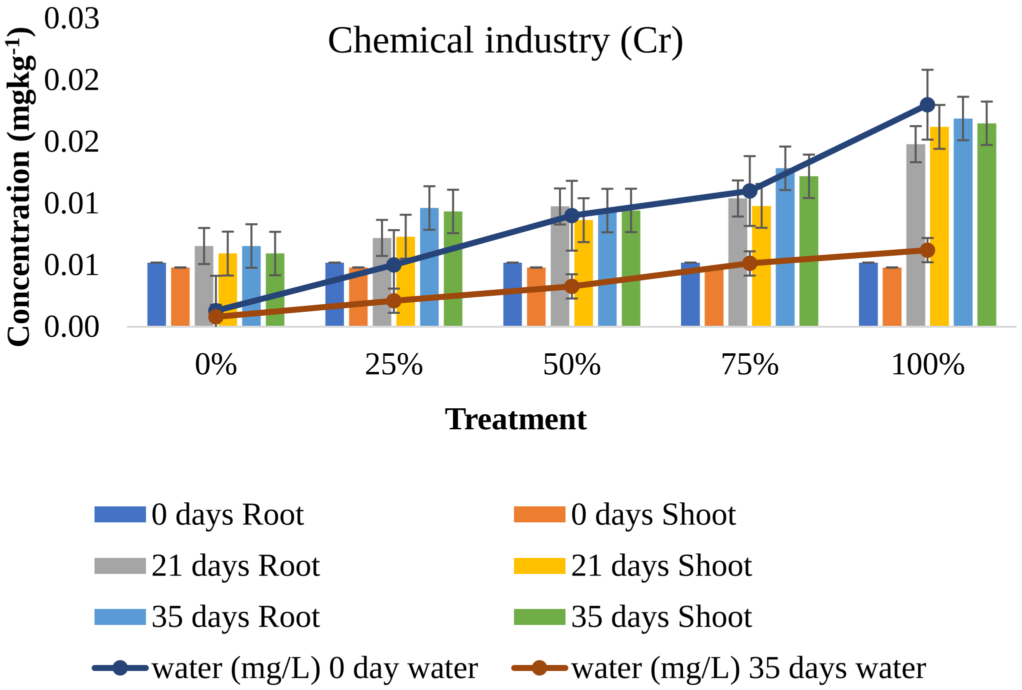
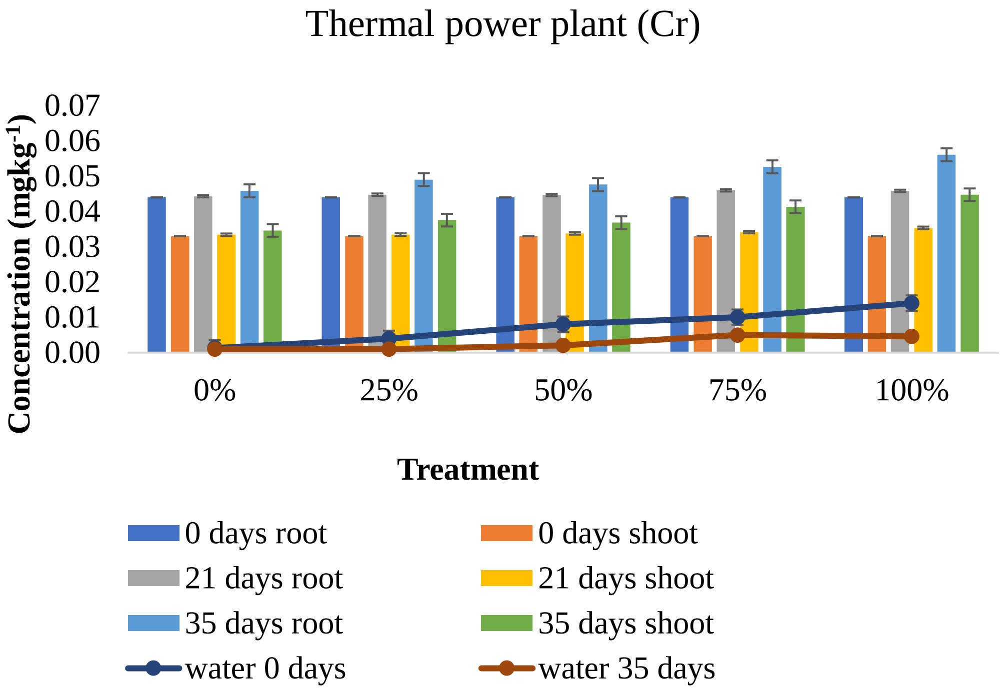


Fig 4.3.18: Ni concentration in *P. stratiotes* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of petrochemical industry.

Cr is non-essential, and its compound is highly toxic to plants and detrimental to their growth and development (Rai et al., 1995). *E. crassipes* initially absorbed 0.05 and 0.04 mgkg^{-1} Cr in their root and shoot respectively. After 21 days of petrochemical industry effluent its accumulates 0.05, 0.07, 0.08, 0.09, 0.12 mgkg^{-1} and 0.04, 0.06, 0.07, 0.08, 0.10 mgkg^{-1} Cr in their root and shoot respectively. After 35 days of exposure its accumulates 0.06, 0.13, 0.20, 0.26, 0.40 mgkg^{-1} and 0.05, 0.13, 0.18, 0.24, 0.38 mgkg^{-1} Cr in their roots and shoots. In exposure with chemical industry effluent this plant accumulates 0.007, 0.008, 0.01, 0.011, 0.015 mgkg^{-1} and 0.006, 0.007, 00.009, 0.01, 0.017 mgkg^{-1} Cr in root and shoot after 21 days and after 35 days its accumulates 0.007, 0.010, 0.009, 0.013, 0.017 mgkg-^{1} and 0.006, 0.009, 0.010, 0.012, 0.016 mgkg^{-1} Cr in their root and shoot of plants. Exposure with battery manufacturing industry its accumulates in the range of 0.01 to 0.02 mgkg^{-1} Cr after 21 days and after 35 days 0.01 mgkg^{-1} accumulation increase in their root and shoot respectively at 75, 100% effluent.




Chemical industry (Cr)
Concentration (mgkg-1)
0.03
0.02
0.02
0.01
0.01
0.00
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Thermal power plant (Cr)
Concentration (mgkg-1)
0.07
0.06
0.05
0.04
0.03
0.02
0.01
0.00
0%
25%
50%
75%
100%
Treatment
0 days root
0 days shoot
21 days root
21 days shoot
35 days root
35 days shoot
water 0 days
water 35 days

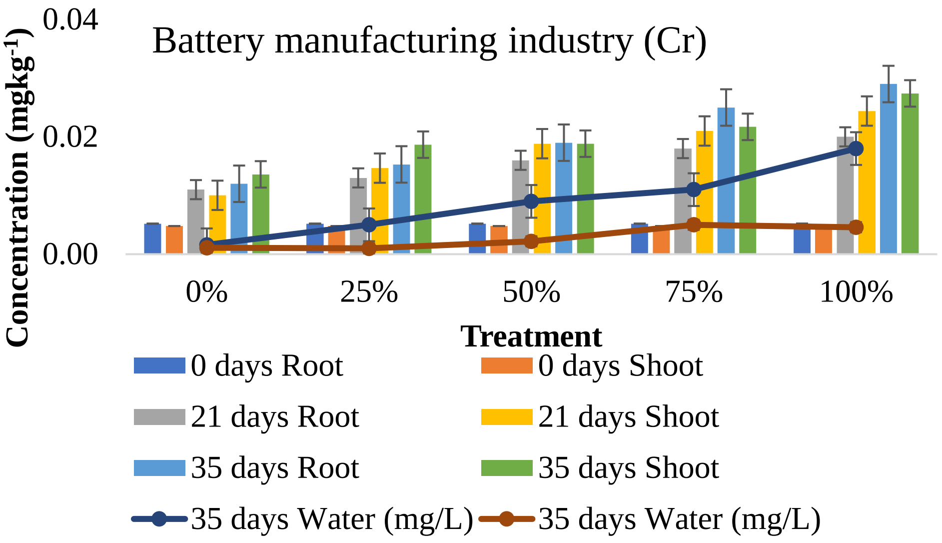
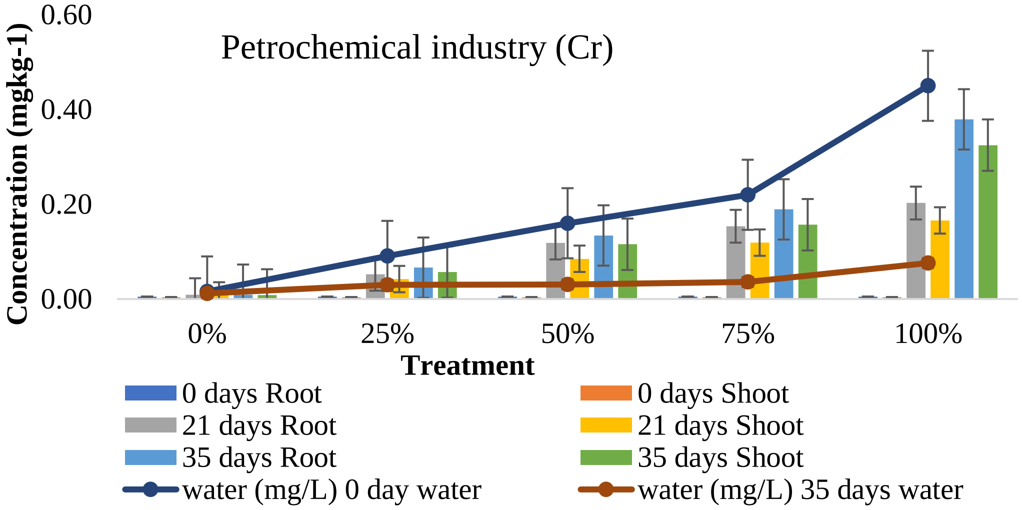
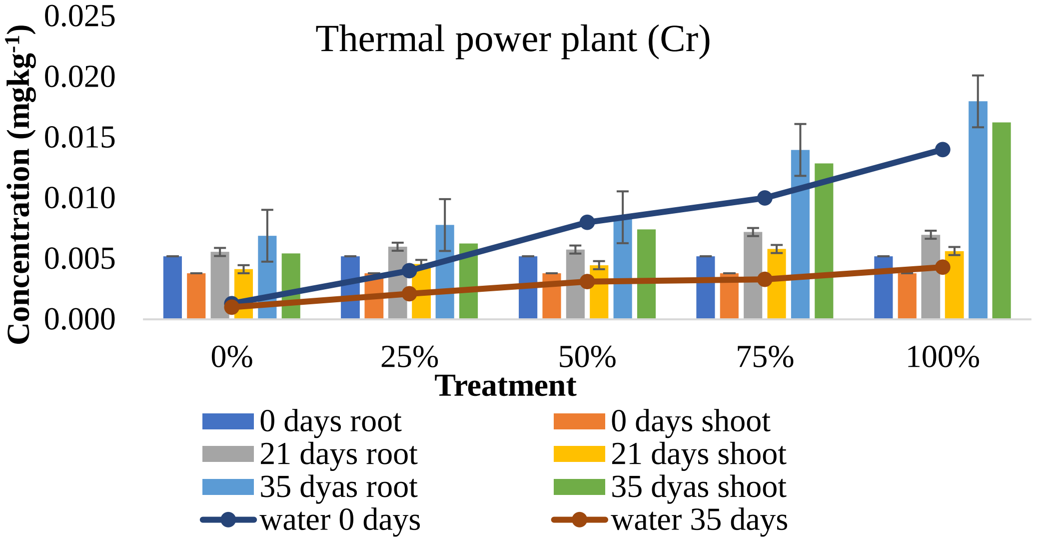
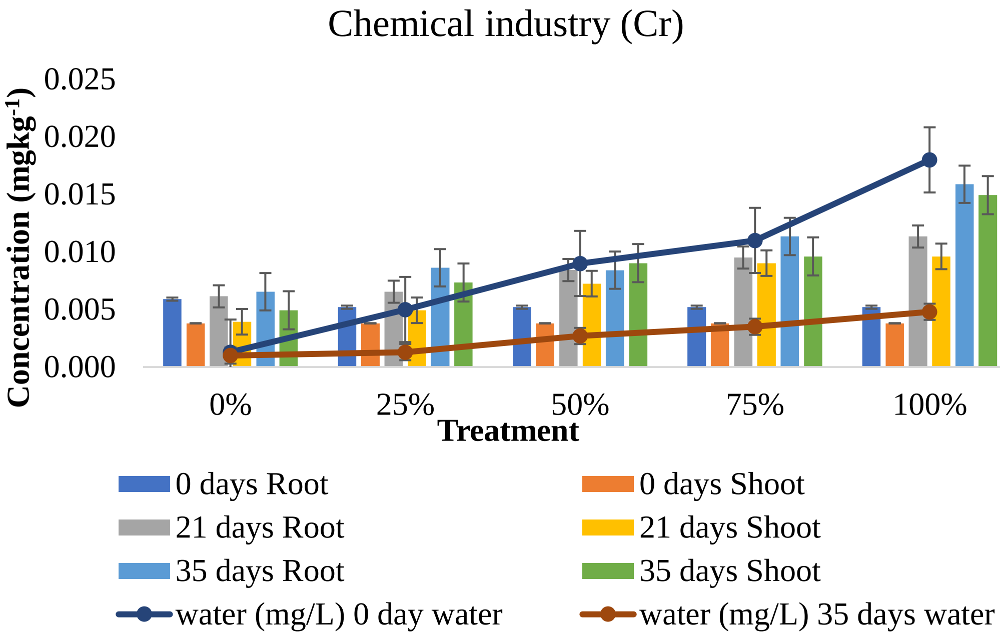


Fig. 4.3.19: Cr concentration in *E. crassipes* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of different industries.

Cr initially found 0.01 and 0.00 in root and shoot of *L. gibba*. After 21 days of exposure it was found 0.01, 0.05, 0.12, 0.15, 0.20 mgkg^{-1} and 0.00, 0.04, 0.08, 0.12, 0.17 mgkg^{-1} and after 35 days 0.01, 0.07, 0.13, 0.19, 0.38 mgkg^{-1} and 0.01, 0.06, 0.12, 0.16, 0.32 mgkg^{-1} in root and shoot at 0, 25, 50, 75, 100% petrochemical industry effluent respectively. In exposure of chemical industry *L. gibba* accumulates Cr in the range 0.01-0.02 mgkg^{-1} after 35days in root and shoot. In the exposure of thermal power plant effluent and battery manufacturing industry effluent, Cr accumulation was low because Cr concentration was very low i.e. 0.01 mgl^{-1}.




Thermal power plant (Cr)
Concentration (mgkg-1)
0.025
0.020
0.015
0.010
0.005
0.000
0%
25%
50%
75%
100%
Treatment
0 days root
0 days shoot
21 days root
21 days shoot
35 dyas root
35 dyas shoot
water 0 days
water 35 days


Chemical industry (Cr)
Concentration (mgkg-1)
0.025
0.020
0.015
0.010
0.005
0.000
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water

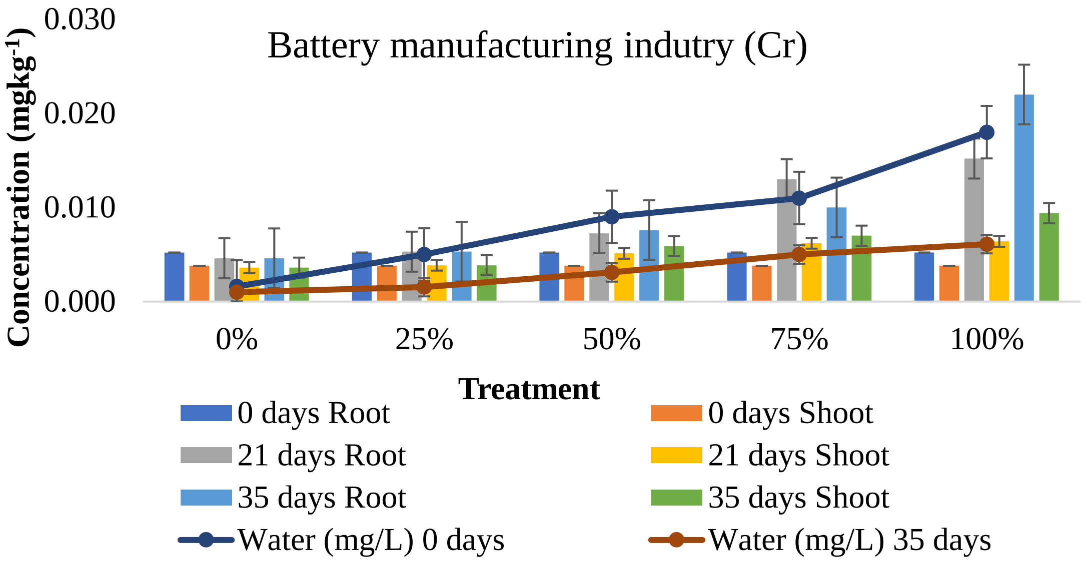
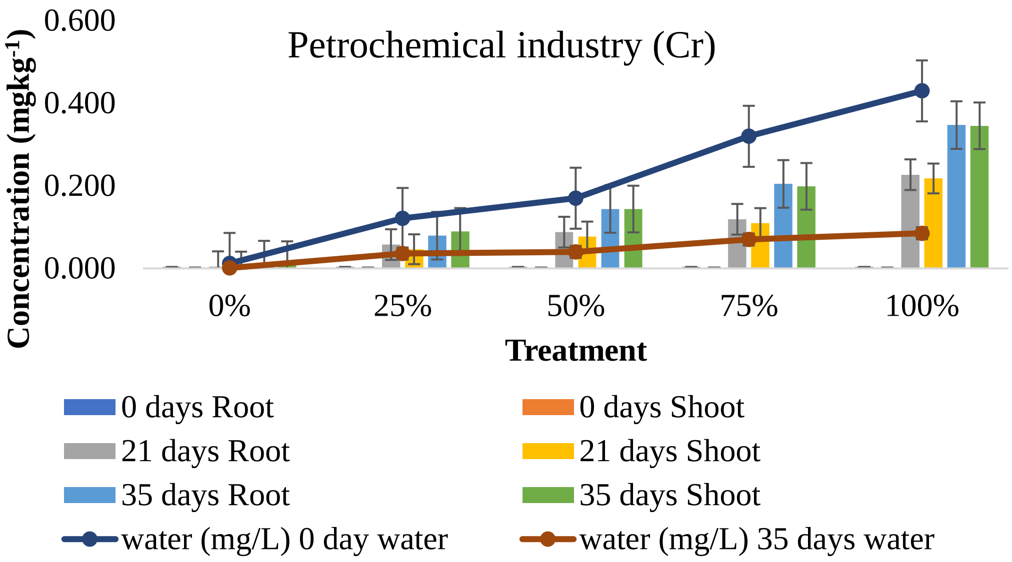
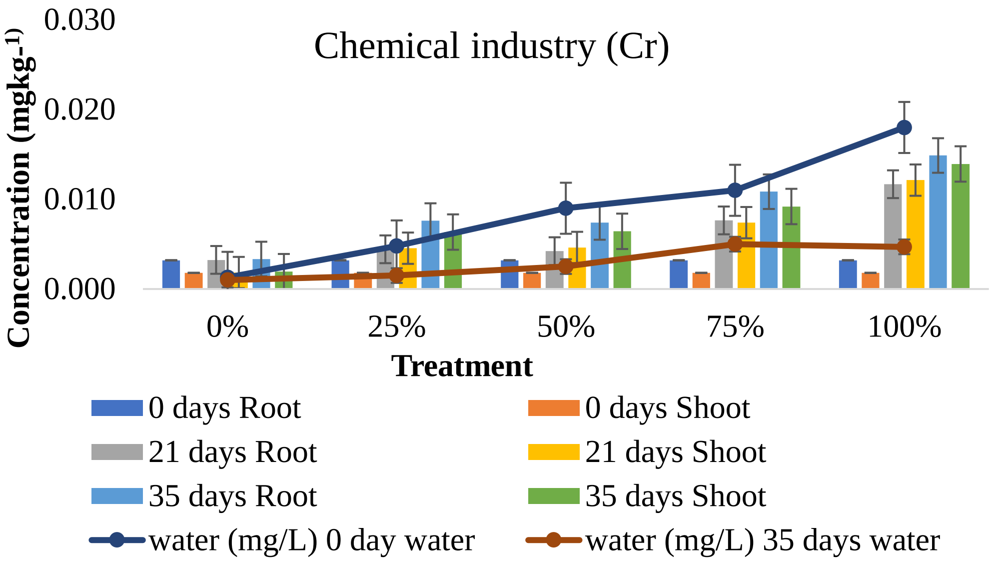
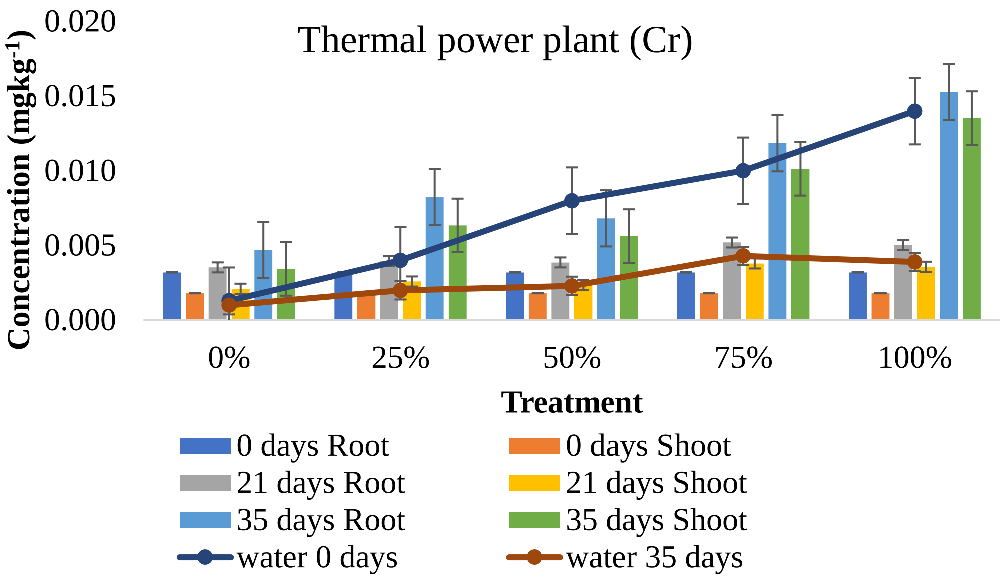


Fig. 4.3.20: Cr concentration in *L. gibba* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of different industry.

P. stratiotes accumulates 0.003, 0.05, 0.08, 0.11, 0.27 mgkg^{-1} and 0.002, 0.04, 0.07, 0.11, 0.21 mgkg^{-1} Cr after 21 days and 0.004, 0.07, 0.14, 0.20, 0.34 mgkg^{-1} and 0.003, 0.09, 0.14, 0.19, 0.34 mgkg^{-1} Cr after 35 days in root and shoot, from petrochemical industry effluent at 0, 25, 50, 75, 100% concentration, as initial concentration of Cr was 0.003 and 0.002 mgkg^{-1} in root and shoots. In chemical industry effluent its accumulates 0.003, 0.008, 0.007, 0.01, 0.01 and 0.002, 0.006, 0.006, 0.009, 0.01 mgkg^{-1} Cr in root and shoot after 35 days of treatment. From thermal power plant *P. stratiotes* accumulates 0.003, 0.008, 0.007, 0.01, 0.01 mgkg^{-1} and 0.003, 0.006, 0.006, 0.010, 0.014 mgkg^{-1} Cr after 35 days in root and shoot.


Petrochemical industry (Cr)
Concentration (mgkg-1)
0.600
0.400
0.200
0.000
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water


Chemical industry (Cr)
Concentration (mgkg-1)
0.030
0.020
0.010
0.000
0%
25%
50%
75%
100%
Treatment
0 days Root
0 days Shoot
21 days Root
21 days Shoot
35 days Root
35 days Shoot
water (mg/L) 0 day water
water (mg/L) 35 days water



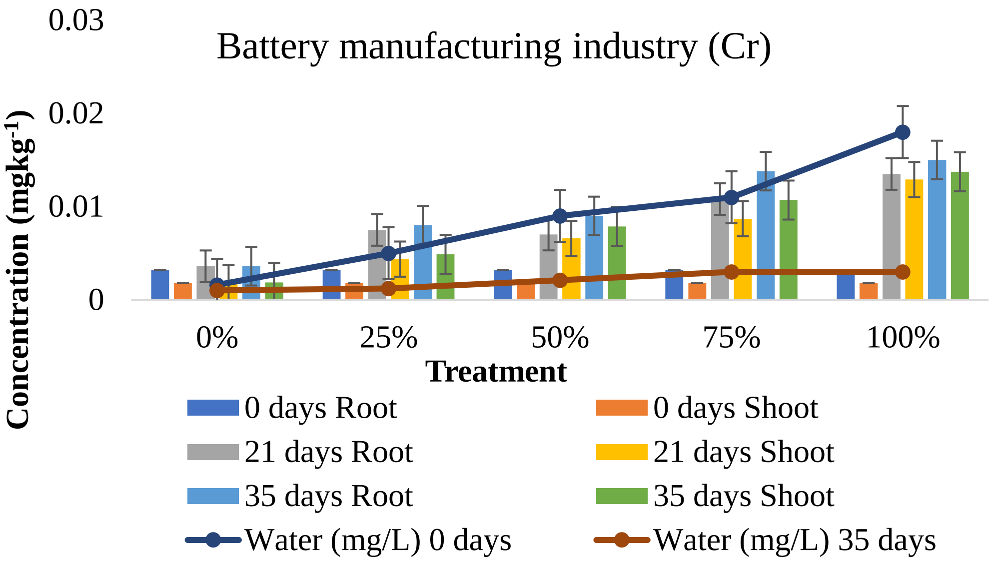


Fig 4.3.21: Cr concentration in *P. stratiotes* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of different industry.

Cd is non-essential heavy metals and significantly cause toxicity to higher plants (Sanita De tappi and Gabbrielli, 1999). Cd concentration was found to be 0.04 and 0.03 mgkg^{-1} before treatment in root and shoot of *E. crassipes*. After 21 days of treatment *E. crassipes* accumulates 0.04, 0.05, 0.05, 0.06, 0.06 mgkg^{-1} and 0.03, 0.04, 0.04, 0.05, 0.05 mgkg^{-1} Cd and after 35 days of exposure of chemical industry effluent, its accumulates, 0.04, 0.05, 0.05, 0.06, 0.06 mgkg^{-1} and 0.03, 0.04, 0.04, 0.05, 0.05 mgkg^{-1} Cd in root and shoot of plant respectively. In exposure of battery manufacturing industry Cd concentration was found 0.04, 0.05, 0.05, 0.06, 0.06 mgkg^{-1} and 0.03, 0.03, 0.04, 0.04, 0.05 mgkg-^{1} after 35 days in their root and shoot respectively. Oliveira et al. (2005) observed similar absorption rates between root and leaves in *Salvinia auriculata*.

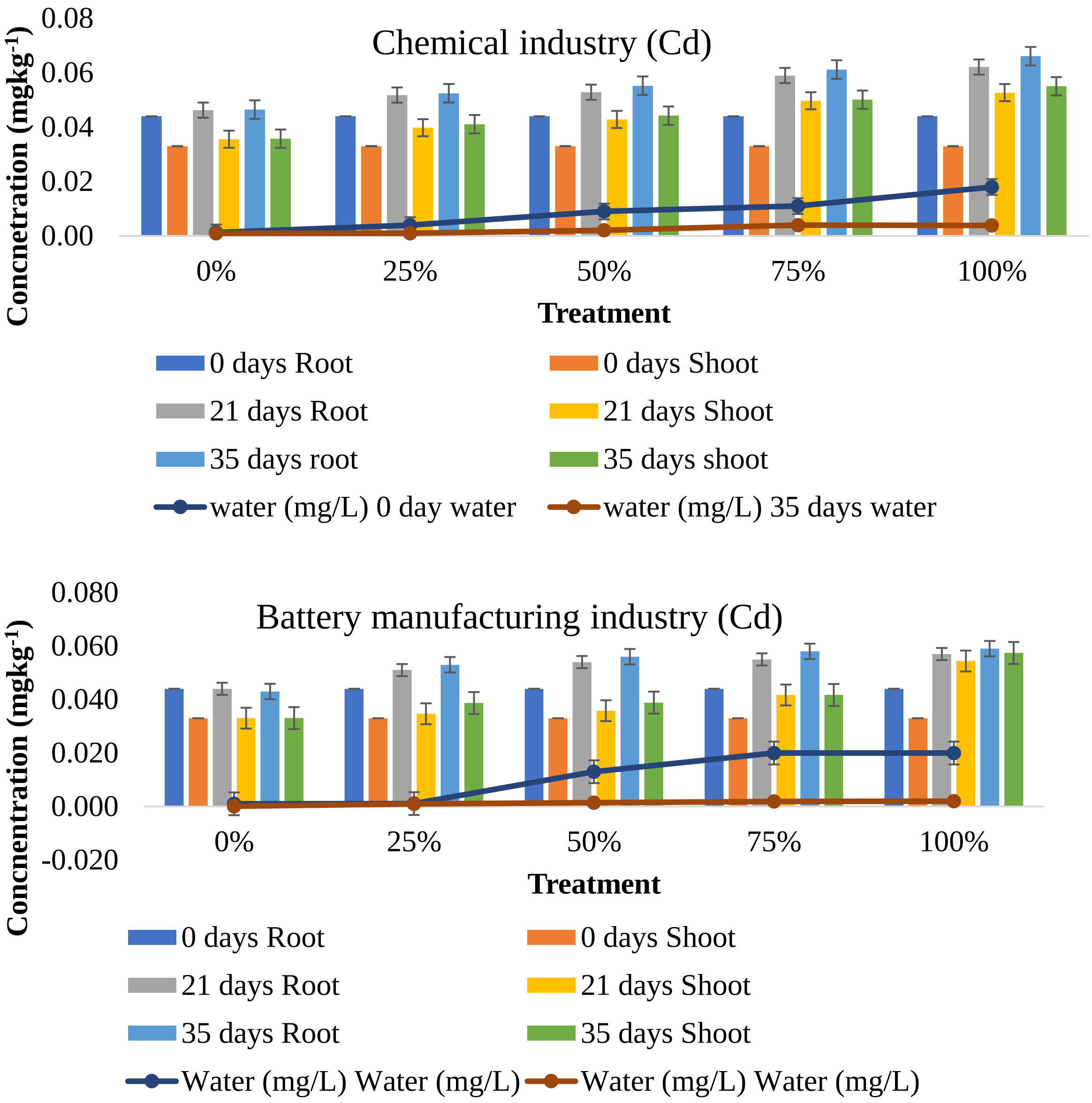


Fig 4.3.22: Cd concentration in *E. crassipes* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of chemical and battery manufacturing industries.

Cd is toxic metal initially found 0.005 and 0.003 mgkg^{-1} in root and shoot of *L. gibba*. After 35 days of exposure Lemna accumulates 0.01, 0.01, 0.02, 0.02, 0.02 mgkg^{-1} and 0.01, 0.01, 0.01, 0.02, 0.02 mgkg^{-1} Cd in root and shoot of plants at 0, 25, 50, 75, 100% effluent respectively. In battery manufacturing industry effluent exposure to plant accumulation occurred less than 0.01 mgkg^{-1} due to low concentration of Cd in effluent. Cd has become an increasing toxic problem because of its toxic effects on biological systems. It is non-essential for plant growth, taken up by root and shoot depressing growth by affecting photosynthesis, chlorophyll, nutrient uptake by plants. In the present study the toxicity of Cd was not up to level to affect the growth of plants.

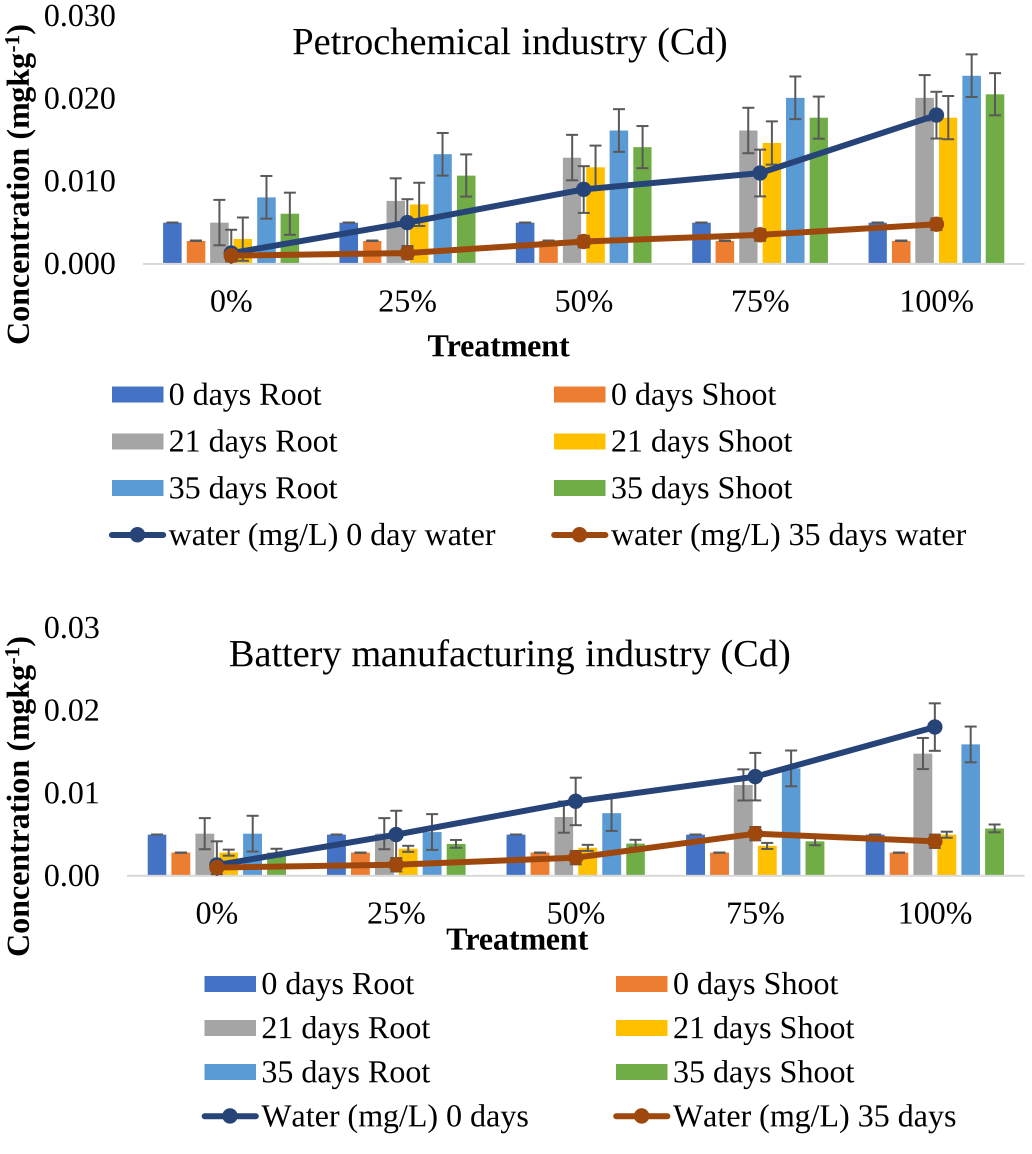
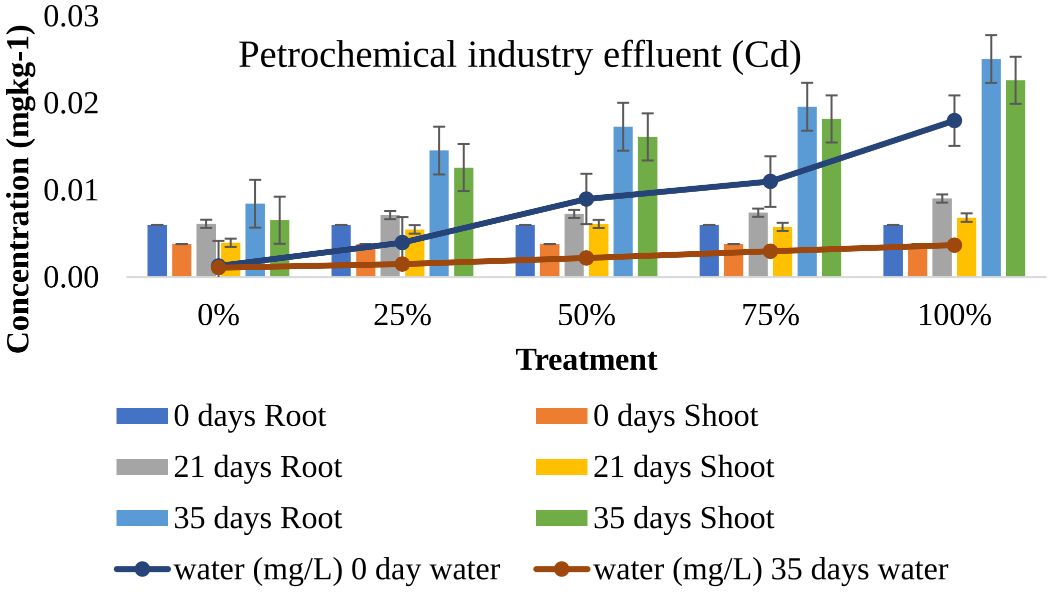


Fig 4.3.23: Cd concentration in *L. gibba* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of chemical and battery manufacturing industries.

Initially *P. stratiotes* absorbed 0.002 and 0.001 mgkg^{-1} in root and shoot respectively. after 21 days its accumulates 0.002, 0.05, 0.08, 0.13, 0.28 mgkg^{-1} and 0.001, 0.06, 0.11, 0.16, 0.27 mgkg^{-1} and 35 days of 0.001, 0.09, 0.17, 0.24, 0.46 mgkg^{-1} and 0.00, 0.08, 0.12, 0.21, 0.47 mgkg^{-1} in root and shoot. In chemical industry effluent, its accumulates 0.002, 0.01, 0.01, 0.02, 0.02 mgkg^{-1} root and 0.002, 0.013, 0.016, 0.018, 0.023 mgkg^{-1} shoot. after 35 days. From battery manufacturing industry effluent, *P. stratiotes* accumulates 0.00, 0.01, 0.01, 0.01, 0.02 mgkg^{-1} and 0.00, 0.00, 0.00, 0.00, 0.01 mgkg^{-1} Cd after 35 days of exposure.



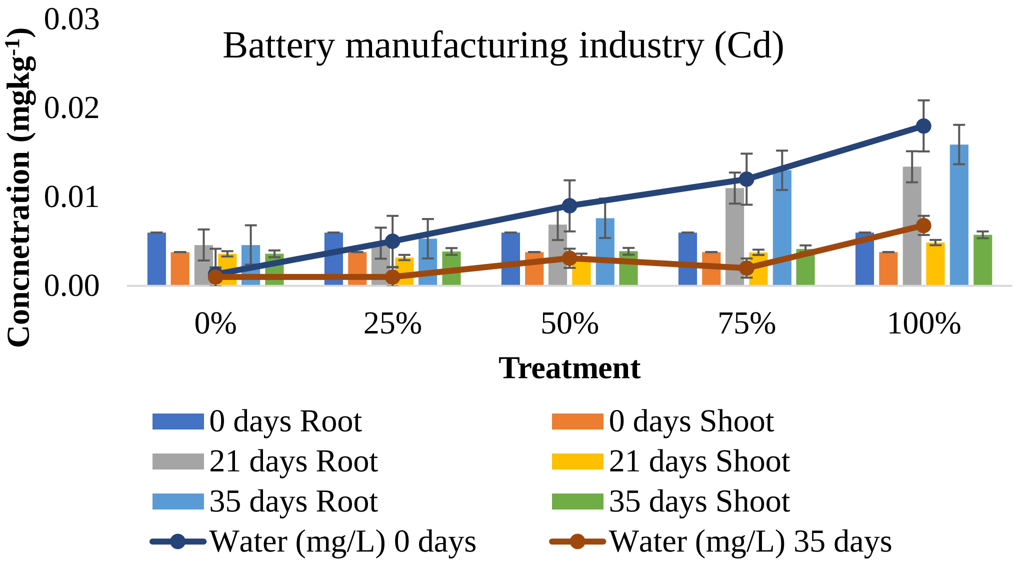


Fig. 4.3.24: Cd concentration in *P. stratiotes* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of chemical and battery manufacturing industry.

Hg initially found 0.02 and 0.02 mgkg^{-1} in root and shoot of *E. crassipes*. After 21 days of 0.02, 0.05, 0.06, 0.08, 0.15 mgkg^{-1} and 0.02, 0.04, 0.04, 0.05, 0.11 mgkg^{-1} and after 35 days 0.03, 0.11, 0.19, 0.26, 0.48 mgkg^{-1} and 0.03, 0.10, 0.13, 0.20, 0.49 mgkg-1 accumulates in root and shoot from 0, 25, 50, 75, 100% petrochemical effluent. In the exposure of Chemical industry effluent *E. crassipes* accumulates 0.00, 0.01, 0.01, 0.02, 0.02 mgkg^{-1} and 0.00, 0.01, 0.02, 0.02, 0.02 mgkg^{-1} Hg after 21 days and 0.01, 0.01, 0.02, 0.02, 0.03 mgkg^{-1} and 0.01, 0.02, 0.02, 0.03, 0.04 mgkg^{-1} Hg after 35 days in their root and shoot respectively. From battery manufacturing industry *E. crassipes* accumulates 0.01, 0.42, 0.51, 1.18, 1.35 mgkg^{-1} and 0.01, 0.22, 0.24, 0.66, 0.84 mgkg^{-1} after 21days and after 35 days its accumulates 0.01, 0.5, 0.86, 1.48, 1.87 mgkg^{-1} and 0.01, 0.38, 0.48, 0.84, 1.06 mgkg^{-1} in root and shoot respectively.

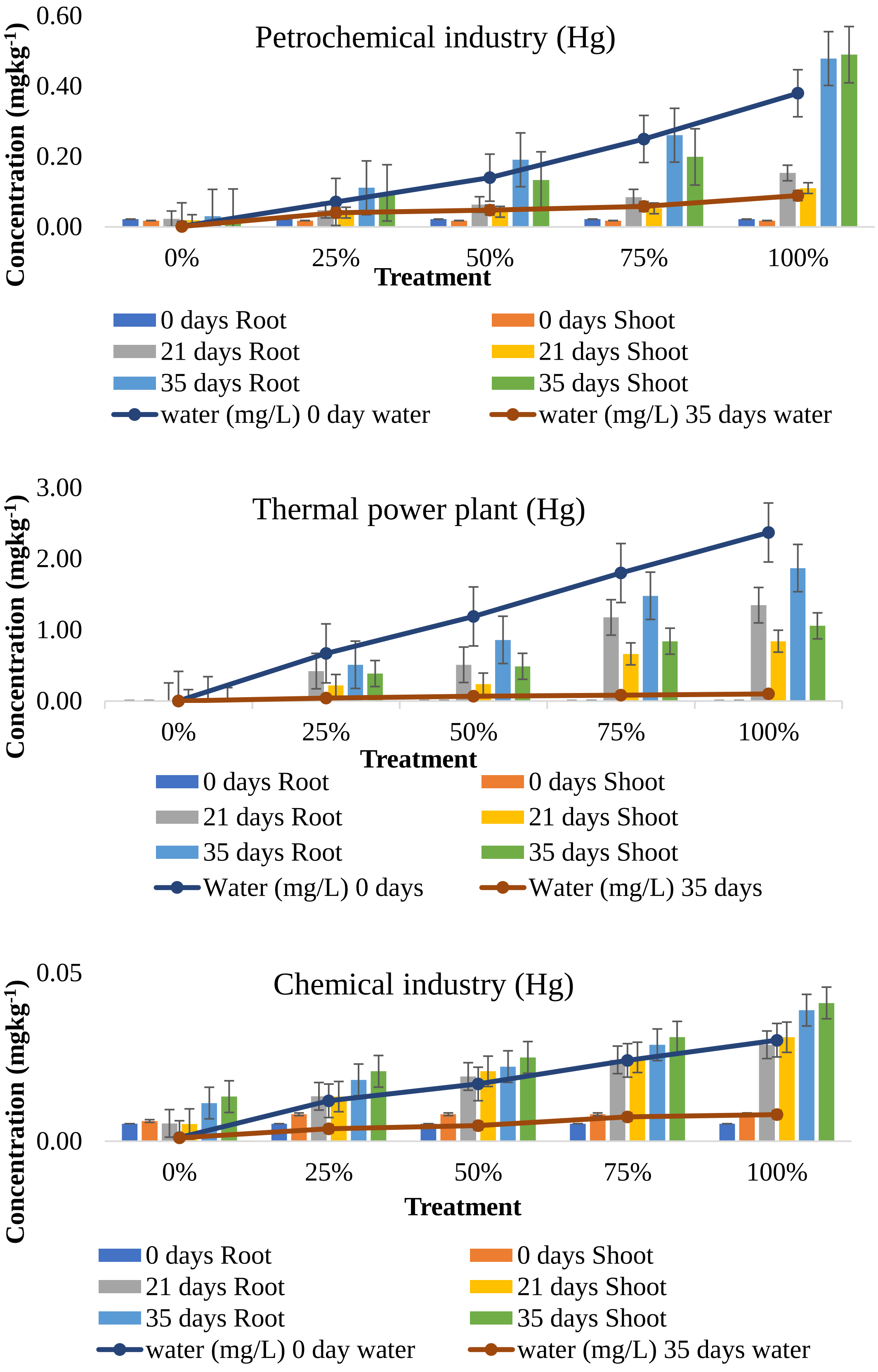
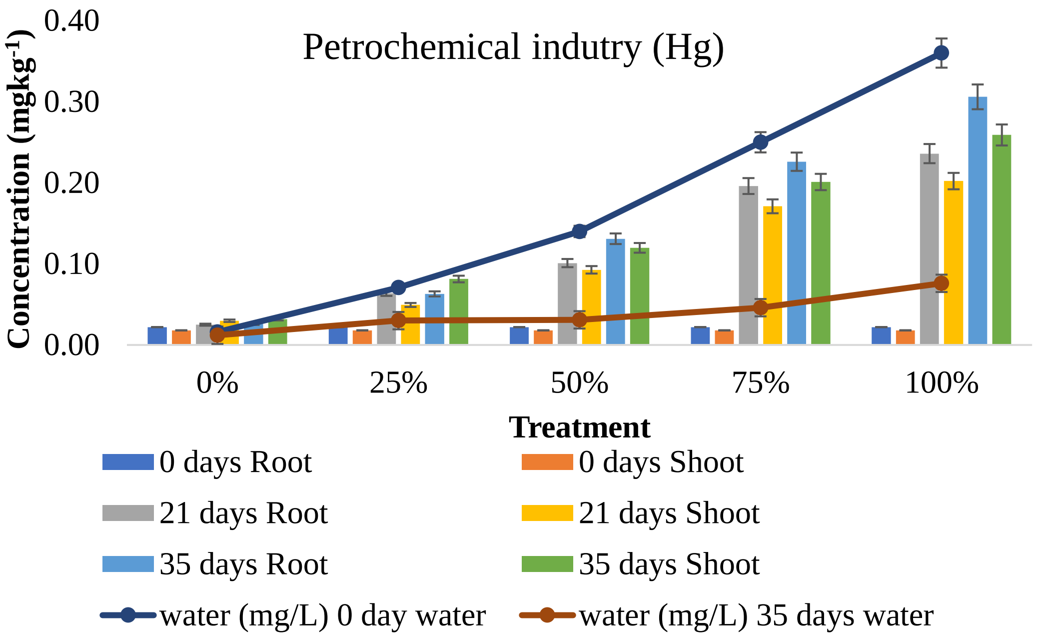
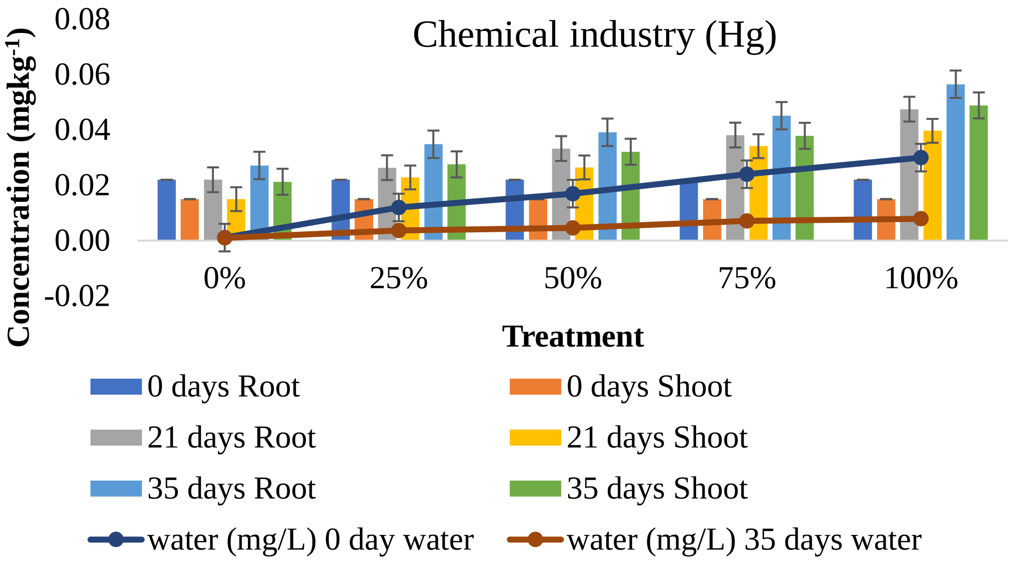


Fig:4.3.25: Hg concentration in *E. crassipes* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of different industry.

Initially Hg was found to be 0.02 and 0.02 mgkg^{-1} in root and shoot of *L. gibba* respectively. After 21 days of exposure of petrochemical effluent it accumulates 0.03, 0.06, 0.10, 0.20, 0.24 mgkg^{-1} and 0.03, 0.05, 0.09, 0.17, 0.20 mgkg^{-1} Hg in root and shoot and after 35 days it's accumulates 0.03, 0.06, 0.13, 0.23, 0.31 mgkg^{-1} and 0.03, 0.08, 0.12, 0.20, 0.26 mgkg^{-1} in root and shoot at 0, 25, 50, 75, 100% effluent respectively. In chemical industry effluent exposure Hg accumulates 0.02, 0.03, 0.03, 0.04, 0.05 mgkg^{-1} in root and 0.02, 0.02, 0.03, 0.03, 0.04 mgkg^{-1} in shoot, after 21 days and after 35 days its accumulates 0.02, 0.03, 0.04, 0.05, 0.06 mgkg^{-1} in roots and 0.02, 0.03, 0.03, 0.04, 0.05 mgkg^{-1} in shoot at 0, 25, 50, 75, 100% effluent. From battery manufacturing industry effluent *L. gibba* accumulates 0.02, 0.47, 0.65, 1.29, 1.37 mgkg^{-1} Hg and 0.02, 0.21, 0.38, 0.78, 1.05 mgkg^{-1} Hg in root and shoot after 21 days of exposure. After 35 days of exposure its accumulates 0.02, 0.53, 0.87, 1.58, 1.87 mgkg^{-1} Hg and 0.02, 0.24, 0.49, 0.094, 1.16 mgkg^{-1} Hg in root and shoot respectively.





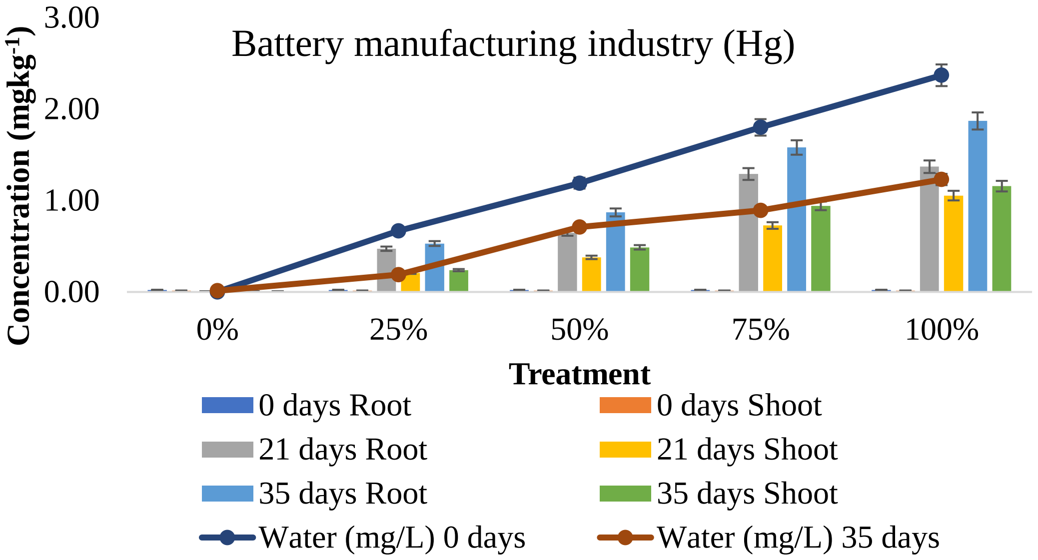
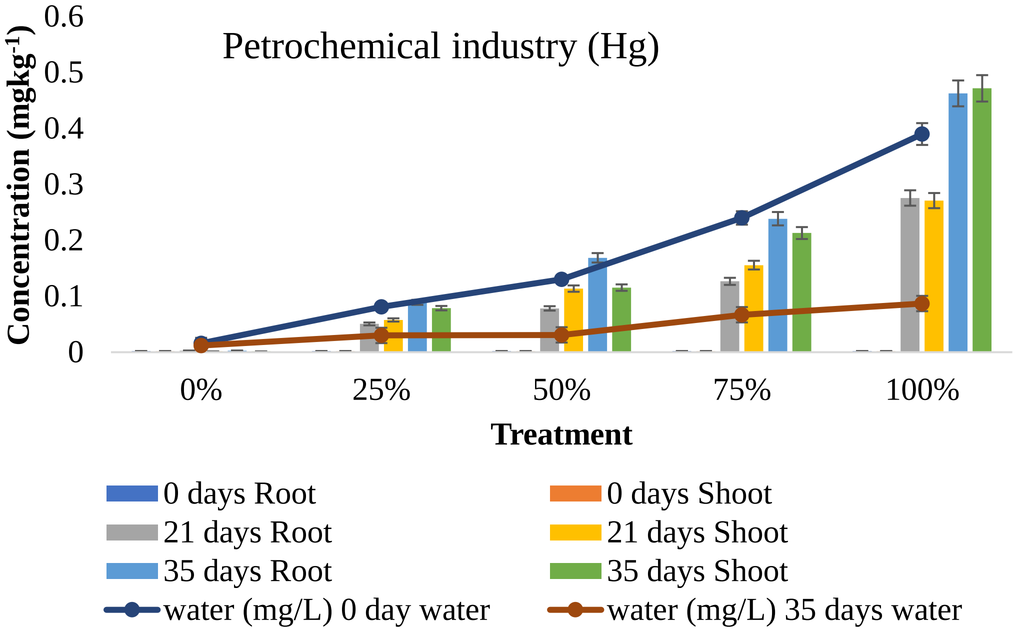
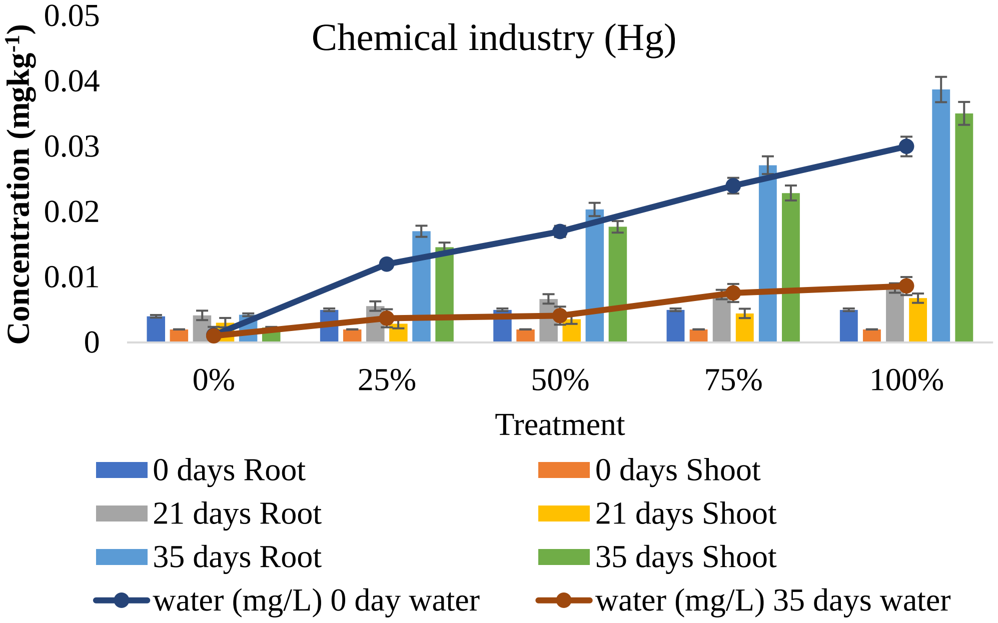


Fig. 4.3.26: Hg concentration in *L. gibba* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of different industry.

P. stratiotes initially accumulates 0.004 and 0.002 mgkg^{-1} Hg in their root and shoot. After 21 days of exposure its accumulates 0.00, 0.05, 0.08, 0.13, 0.28 mgkg^{-1} and 0.00, 0.06, 0.11, 0.16, 0.27 mgkg^{-1} Hg after 21 days and 0.00, 0.09, 0.17, 0.24, 0.46 mgkg-^{1} and 0.00, 0.08, 0.12, 0.21, 0.47 mgkg^{-1} Hg after 35 days in root and shoot from petrochemical industry effluent. In chemical industry effluent its absorbed 0.004, 0.01, 0.02, 0.027, 0.03 mgkg^{-1} and 0.002, 0.015, 0.018, 0.023, 0.035 mgkg^{-1} Hg in their root and shoot. From battery manufacturing industry *P. stratiotes* accumulates 0.002, 0.39, 0.73, 1.25, 1.45 mgkg^{-1} and 0.00, 0.19, 0.55, 0.79, 1.06 mgkg^{-1} after 21 days and 0.002, 0.54, 0.89, 1.54, 1.95 mgkg^{-1} and 0.00, 0.24, 0.69, 0.94, 1.16 mgkg^{-1} after 35 days in their root and shoot.





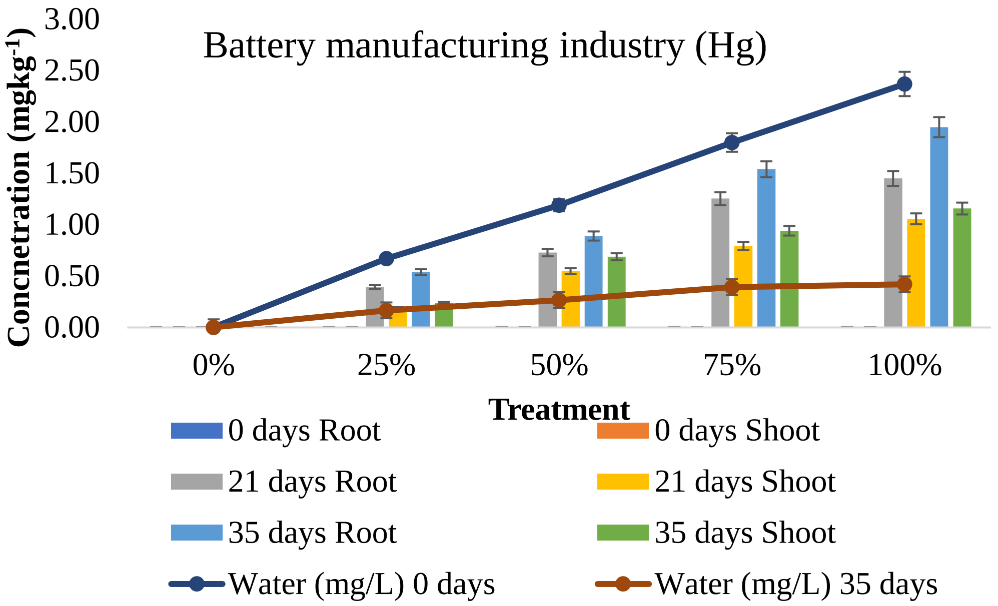


Fig 4.3.27: Hg concentration in *P. stratiotes* in between root and shoot at 0, 21 and 35 days in 0, 25, 50, 75 and 100% effluent concentration of different industry.

E. crassipes, P. stratiotes and *L. gibba* are classified as suitable for phytoremediation of heavy metals from industrial wastewater. These plants that are not eaten by any mans and animals and abundant in water system. Thus, its ability to accumulates metals in its tissues from wastewater system where its grows and metals concentration in root and shoot is indicator of its potential capacity to serve as a phytoremediation plant coupled with the non-existence of a sigh of the toxic effect of metals on the plant when sampled. The maximum concentration was found in roots of all the macrophytes, so root of macrophytes play key role in the accumulation of metals. When wide distribution, rapid production, adaptability, variety of groups, low cost and the possibility relating to the reusability of biomass, these plants are attractive and promising species in the remediation of affected environment.

In the studied heavy metals are efficiently accumulated from the effluent with great increase after 21 days. In sharp decrease of metals remaining in wastewater is inactivate of the first attainment of the saturation state. In conclusion, *E. crassipes, P.*

stratiotes and *L. gibba* used as eco technological wastewater treatment. These are preventing the spread of heavy metal from contaminated industrial site into fresh aquatic system. Higher metals accumulation occurred at 50% and 75% concentrated effluent. Thus, these macrophytes are an easy and cheap alternative of wastewater purification.

Chapter 4.4

Comparative study on the phytoremediation potential of various aquatic weeds to remediate heavy metals from different industrial wastewater

Comparative study on the phytoremediation potential of various aquatic weeds to remediate heavy metals from different industrial wastewater

India and other developing countries require phytoremediation to be an alternative process of wastewater treatment that is economical and cost-effective, less technical and plant-based technology (Andleeb et al., 2010). *E. crassipes, P. stratiotes* and *L. gibba* are invasive free-floating aquatic plants and used as the natural constituents to absorb heavy metals from various industrial wastewater (Vymazal and Kropfelova, 2008; Begum and Harikrishna, 2010). This study evaluates the efficiency of *E. crassipes*, *P. stratiotes* and *Lemna gibba* to remove heavy metals from the effluents of various industries of U.P.

E. crassipes is known for its ability to grow in various contaminated waters and their growth and photosynthetic activities are play key role in implementation in phytoremediation (Gopal, 1987; Jamuna and Noorjahan, 2009). *E. crassipes* is successful in removing heavy metals so, it grown in industrial effluents for evaluation of it potential (Lasat, 2002). *E. crassipes* efficiently removed 78.57, 83.04, 74.18, 70.81, 68.66, 73.70, 81.54, 80.71 and 76.60 % from 100% concentrated effluent, 88.71, 78.42, 70.81, 68.66, 73.70, 81.54, 80.71 and 76.80% from 75% concentrated effluent, 81.68, 87.15, 76.24, 55.81, 63.68, 79.44, 77.22 and 65.71% from 50% effluent, 79.15, 74.67, 65.42, 58.97, 69.23, 63.64, 60.40 and 43.66% from 25% and 23.53, 41.18, 37.50, 31.25, 25.00, 43.75, 36.26 and 18.75 % of Fe, Mn, Zn, Cu, Pb, Ni, Cr and Hg from 0% effluent (control tap water) of petrochemical industry effluent respectively. In petrochemical industry effluent heavy metal removal efficiency of *E. crassipes* is maximum show in 75% concentrated as compare to other dilution. In average *E. crassipes* maximally removed Fe (82.03%) followed by Mn (80.82%), Ni (75.93%), Cr (74.52%), Zn (71.66%), Pb (65.69%) and Cu (62.72%). Jayaveera et al. (2007), studied on *E.*

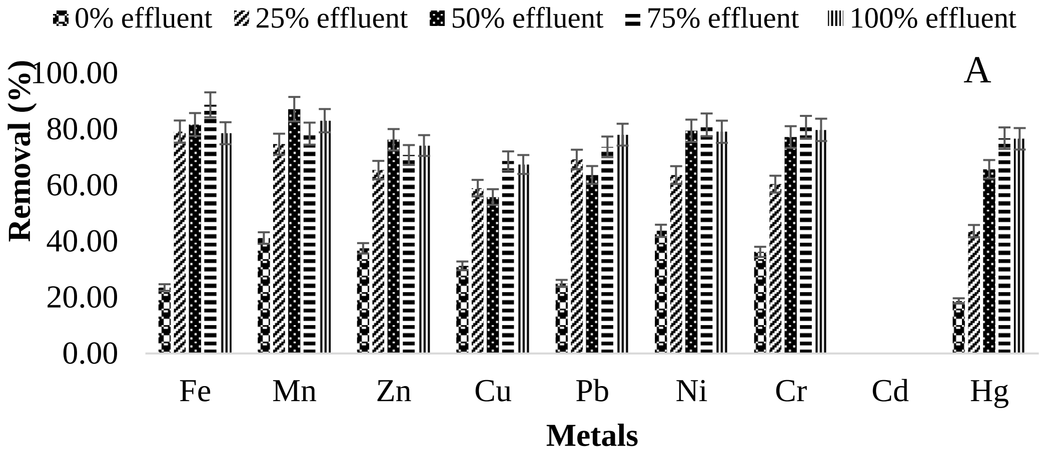
crassipes grown under nutrient and without nutrient condition for Fe rich effluent for 15 days and found that Fe was largely removed by rhizofiltration process under poor nutrient condition.

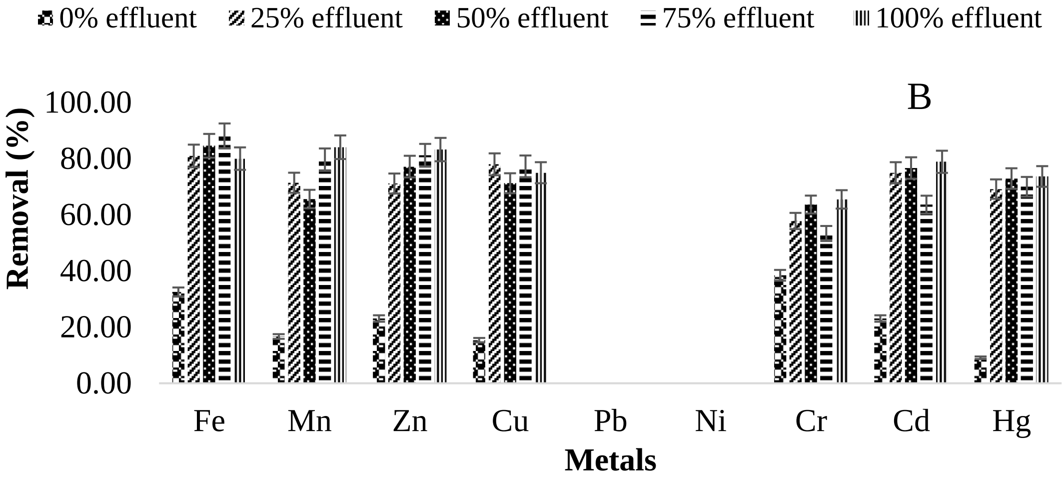
In chemical industry effluent Fe, Mn, Zn, Cu, Cr, Cd and Hg are removed 80.02, 84.08, 83.24, 75.00, 74.44, 78.89, 73.67% from 100% concentrated effluent, 88.14, 79.66, 81.21, 77.27, 67.27, 63.64 and 70.00 from 75% concentrated effluent, 84.59, 65.63, 77.17, 71.25, 67.27, 63.64, 70.00% from 50% concentrated effluent, 80.95, 71.43, 71.15, 78.00, 80, 75, 69.17% from 25% concentrated effluent and 32.50, 16.67, 23.08, 15.38, 38.46, 23.08 and 9.09% from 0% concentrated effluent respectively. In chemical industry effluent also, Fe is strongly removed (83.43%) followed by Zn (78.19%)> Cu (75.38%)> Mn (75.20%)> Cr (74.87%)> Cd (73.55%)> Hg (71.44%). Hernandez et al. (2015) reported that *E. crassipes* is possibly removed Cu (99.8%), Pb (97.9%) and Zn (94.37%) of 5 days. They also explained that it's has great capacity to remove metals due to more abundant and more surface availability. In general, various studies concluded that higher metals accumulated in roots mays be due to their absorption to the surface of root tissues (Mohamad and Puziah 2010; Victor et al., 2016a). Ajayi and Ogunbayo (2012) examined the efficacy of *E. crassipes* in textile, pharmaceutical and metallurgical wastewater for removal of Cd, Cu and Fe, and found that plants are good choice for Cd but not for so much for Fe and Cu. After 5 weeks of treatment *E. crassipes* remove 94.87, 95.59 and 93.55% cadmium from textile, pharmaceutical and metallurgical wastewater. Mohmood et al. (2005) studied the phytoremediation potential of *E. crassipes* on textile effluent collected from Lahore, Pakistan. It's feasibly treated wastewater and removed Cr, Zn, Cu after 5 days. Plants shows 94.78, 96.88 and 94.44 % reduction in Cr, Zn and Cu respectively. Biomass of plant is able to remove both chromium ions from aqueous solution (Elangovan et al.

2008; Hasan et al., 2010). Espinoza-Quinone et al. (2008) studies the removal of ionized Cr from water by using aquatic macrophyte with *E. crassipes,* found maximum removal efficiency for Cr^{3+} and minimum for Cr^{6+}. Worked modified by Swain et al., (2014) reported that highly capable for removal of Cd and Cu, also found that highest Cd concentration (230.39 $mgkg^{-1}$) accumulated in root and Cu (2314.2 $mgkg^{-1}$) accumulated in shoots. Priya and Selvan (2014) explains that *E. crassipes* attempts as remediation tools because of its high tolerance, higher reproduction rate, higher pollutant (organic and inorganic) removal.

In the thermal power plant wastewater, *E. crassipes* removed 80.02, 76.82, 86.08, 54.17, 67.14% from 100% concentrated wastewater, 88.14, 86.67, 83.67, 36.32, 50.00% from 75% concentrated wastewater, 84.59, 66.67, 74.69, 42.86 and 73.75% from 50% concentrated wastewater, 80.59, 44.44, 70.83, 58.00 and 75.00% from 25% concentrated wastewater and 16.67, 16.67, 23.08, 33.32 and 23.08% metals i.e. Fe, Mn, Zn, Cu and Cr respectively. These metals in 75 and 100% concentration are more effectively removed as compare to others dilution. Like petrochemical industry effluent, this effluent also shows maximum removal of Fe i.e. 83.43% and minimum of Cu i.e. 47%. In battery manufacturing industry effluent *E. crassipes* potentially removed 70.37, 84.08, 83.24, 75.00, 82.42, 74.44, 78.89 and 81.13% of Fe, Mn, Zn, Cu, Pb, Cr, Cd and Hg, at 100% concentrated effluent, 74.48, 79.66, 81.21, 77.27, 85.56, 54.55, 78.89 and 83.51% of Fe, Mn, Zn, Cu, Pb, Cr, Cd and Hg at 75% concentrated effluent, 73.09, 65.63, 77.17, 71.25, 88.82, 75.56, 76.67 and 85.71% of Fe, Mn, Zn, Cu, Pb, Cr, Cd and Hg at 50% concentrated effluent, 63.38, 71.43, 71.15, 78.00, 84.36, 80.00, 75.00 and 79.09% of Fe, Mn, Zn, Cu, Pb, Cr, Cd and Hg at 25% concentrated effluent, 55.88, 16.67, 22.31, 23.08, 20.43, 17.29, 23.08 and 28.57 % of Fe, Mn, Zn, Cu, Pb, Cr, Cd and Hg, at control respectively. *E. crassipes* effectively removed metals from 75% and

100% effluent of battery manufacturing industry effluent as compare to others dilution but comparison with metals wise Pb (85.29%) is maximally removed followed by Hg (82.36%) > Zn (78.19%)> Cd (77.36%)> Cu (75.38%)> Mn (75.19%)> Cr (71.13%)> Fe (70.33). According to results *E. crassipes* shows greater potential to remove heavy metals from different industrial wastewater. Liao and Cheng (2004) arranged the heavy metals according to the removal of heavy metals, that is, Cu> Zn> Ni> Pb> Cd, similarly in this investigation we find the pattern i.e. Fe>Pb>Zn>Ni>Cd>Mn>Hg>Cr>Cu. So, we can say that improvement and large scales utilization of the *E. crassipes* plant serve as positive approach to control water pollution especially in developing countries.





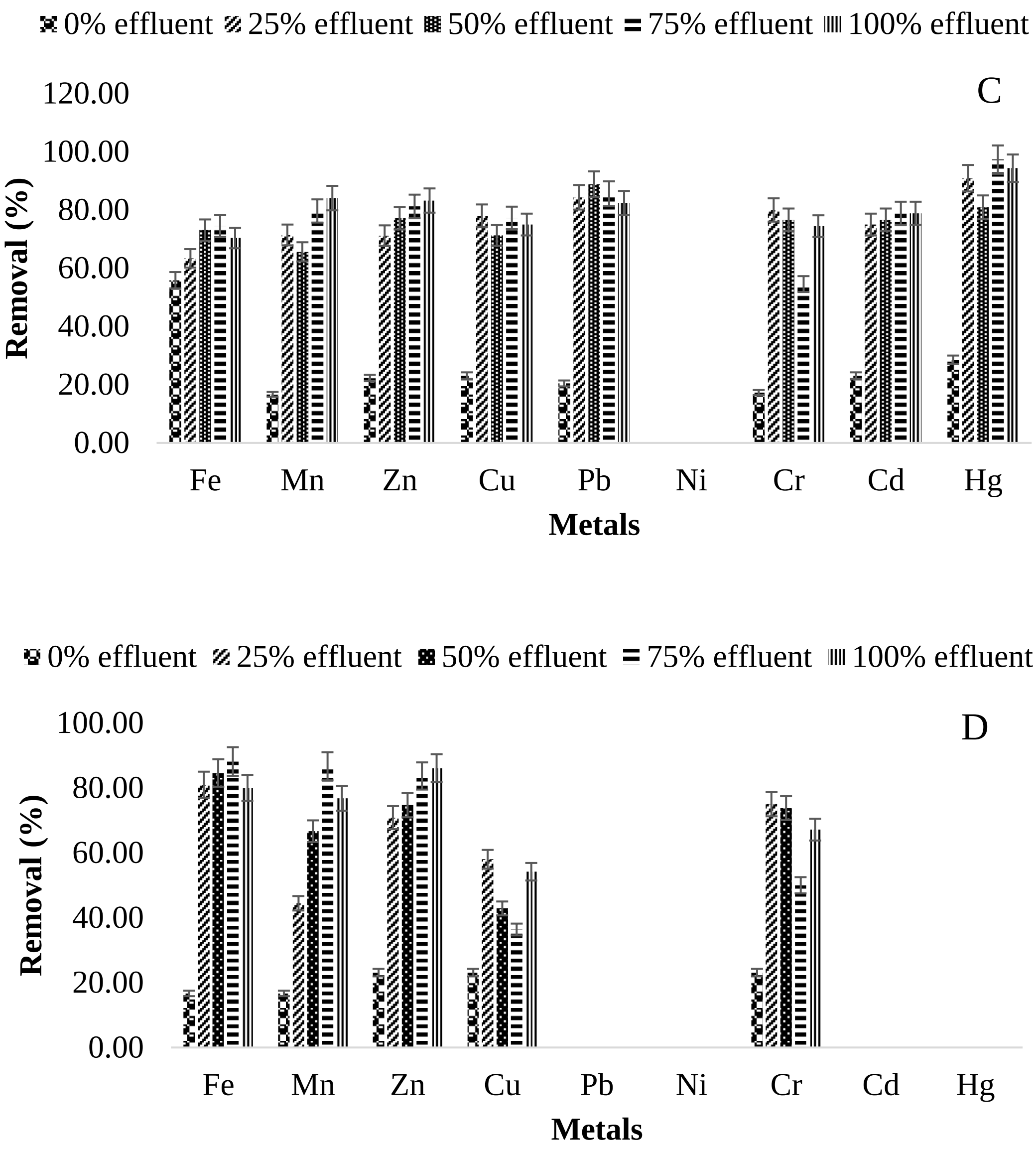


Fig. 4.4.1: Removal efficiency of heavy metals by *E. crassipes* from Petrochemica (A), Chemical (B), Thermal power plant (C) and Battery manufacturing industry (D).

L. gibba is small size macrophytes of Lemnaceae group. Due to its high multiplication rates and susceptibility to pollutant it is important in phytoremediation process for testing various toxicity procedure. In the study *L. gibba* removed 35.29, 72.20, 68.32, 76.06 and 65.01% Fe, 41.18, 84.61, 86.43, 82.78 and 81.92% Mn, 54.34, 65.71, 71.80, 64.32 and 73.82% Zn, 12.5, 63.18, 74.51, 67.09 and 75.29 % Cu, 25.00, 72.73, 74.21, 70.91 and 73.82% Pb, 24.37, 53.49, 77.22, 73.64 and 79.09% Ni, 25, 67.03, 80.63, 83.64 and 83.11% Cr and 25, 57.74, 77.85, 81.6 and 78.89% Hg of 0, 25, 50, 75 and 100% concentrated petrochemical industry effluent respectively. At 50 and 100% dilution *L. gibba* shows maximum removal efficiency of metals. Mn is strongly removed by *L. gibba* with 83.94% followed by Cr (78.60%)>Hg (74.02%)> Pb (72.92%)> Ni (70.86%)> Fe (70.40%)> Cu (70.02%)> Zn (68.92%).

Axtell et al. (2003) applied *L. minor* to determining the removal potential of Ni and Pb. Results showed that its removed larger i.e. 76 and 82% of Pb and Ni respectively. Mishra and Tripathi (2008) investigated the role of duckweed in removal of heavy metals such as Cu and Cd in 1,2 and 5 mgl^{-1} concentrated solution after 15 days in laboratory. Mant et al. (2006) studied removal rate of metals by *P. stratiotes* and *S. polyrrhiza* due to its efficient growth and high biomass accumulation in nutrient and metals contaminated environment. In the present study demonstrated that removal efficiency of *L. gibba* is mostly greater than 70% in all effluent at all dilution. Bokhari et al. (2016) also examined the potential of *L. minor* to remove Cd, Cu, Pb and Ni in two different effluent in glass house experiment for 31 days in hydroponic studied and overall plant demonstrated the ability to remove all metals. In comparison Pb is maximally removed by plants from both the effluents.

In chemical industry effluent its removed 16.67, 65.22, 81.39, 78.10 and 69.16% of Fe, 14.29, 68.10, 65.63, 71.19 and 65.66% of Mn, 12.14, 71.15, 76.09, 73.83 and 72.43% of Zn, 26.67, 78.00, 71.25, 77.27 and 75.00% of Cu, 15.38, 80.00, 77.78, 67.27 and 74.44% of Cr, 23.08, 75.00, 76.67, 63.64 and 78.89% of Cd, 9.09, 69.17, 72.94, 70.00 and 73.67% of Hg from 0, 25, 50, 75 and 100% concentrated effluent respectively.

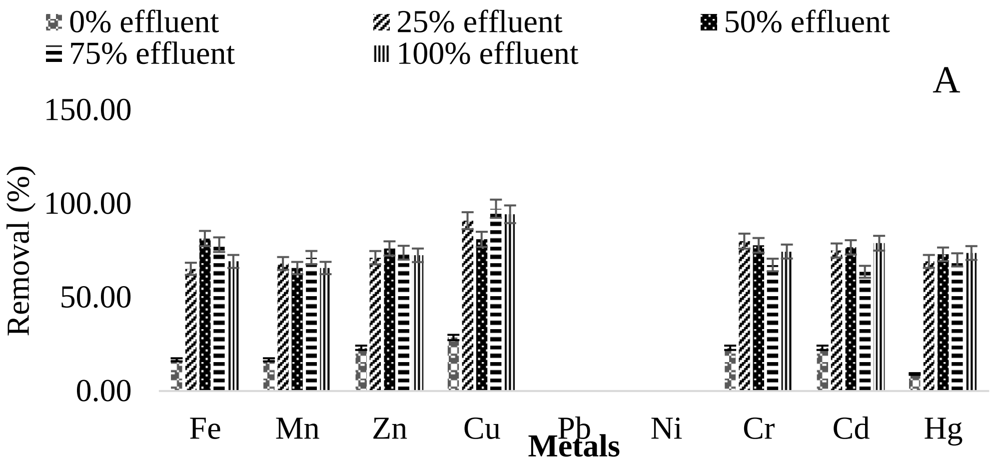
In chemical effluent, in average 50% dilution comparatively efficient for plant as compare other dilution. *L. gibba* maximally removed Cu with 75.38% potential followed by Cr (74.87%), Cd (73.55%), Fe (73.47%), Zn (73.37%), Hg (71.44%), Mn (67.64%). Leblebici and Aksoy (2011) used plant species *L. minor* and *S. polyrhiza* for purification of water contaminated with Pb with different concentration (0, 5, 10, 25 and 50 mgl $^{-1}$) and found Lemna is more effective than *S. polyshiza* and also that excess nutrient raised tolerance of plants to metals concentration. Higher tolerance would be useful in phytoremediation process and allowed plant growth in contaminated system.

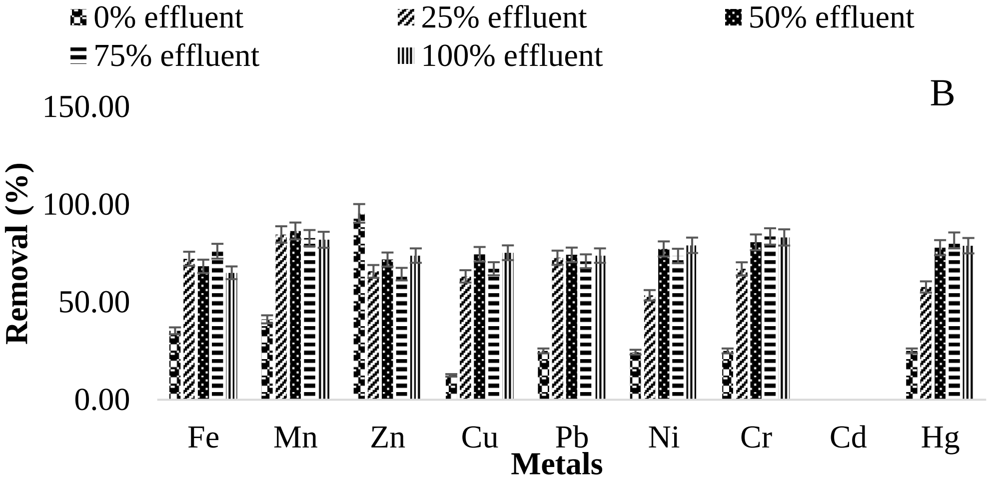
Gupta et al. (2011) studied the ability of *Vallisneria spiralis* and *Hydrilla verticillata* to absorbing contaminated from tannery effluent of Kanpur. Results shows maximum concentration found in root of *V. spiralis* i.e. 385.6 mgkg^{-1} and whole *H. verticillate* accumulates 201.6 mgkg^{-1} from 100% concentrated effluent after 9th day of exposure. Hawkins et al., (1995), studied on wetland plants for treatment of petrochemical industry effluent. After 7 days of laboratory experiment average 80%, Zn toxicity decrease by *Ceriodaphnia dubia* from effluent. According to Basile et al. (2012) three macrophytes *Elodea canadensis, L. minor,* and *Leptodictyum riparium* efficiently removed Cd, Pb, Zn and Cu, but *L. minor* most effective in comparison to other plants. Result shows that L. minor removed 95% Cd, 93% Pb, 86.5% Cu and 81.2% Zn from water.

In the case of thermal power plants effluent *L. gibba* removed 16.67, 64.44, 71.76, 80.34 and 82.34% Fe, 21.67, 28.57, 63.33, 52.22 and 74.55% Mn, 23.08, 62.50, 77.81 and 73.42% Zn, 23.08, 60.00, 57.14, 45.45 and 62.50 Cu, and 23.08, 47.50, 61.25, 67.00 and 69.29% Cr from 0, 25, 50, 75, and 100% concentration. In this power plant *L. gibba* is more efficient in 100% effluent for removal of metals and shows maximum removal of Fe with 74.72% potential and minimum removal of Mn with 54% potential.

In case of effluent from battery manufacturing industry *L. gibba* removed 16.67, 61.90, 79.19, 69.49 and 78.94% Fe, 18.03, 78.50, 62.50, 66.10 and 70.92% Mn, 28.57, 78.63, 81.74, 86.64 and 76.40% Zn, 21.86, 82.73, 85.71, 87.57 and 84.34% Cu, 27.86, 83.64, 79.52, 85.95 and 87.17% Pb, 32.5, 81.43, 80.80, 83.78, 79.77% Cr, 23.5, 77.86, 83.85, 85.86, 74.81% Cd and 13.75, 64.92, 68.25, 76.49 69.69% of Hg from 0, 25, 50, 75 and 100% concentration. In 75% dilution *L. gibba* shows maximum efficiency and it was maximally removed Cu with 85.05% efficiency followed by

Pb (84.06%)>Cr (81.14%)> Zn (80.85%)> Cd (80.51%)> Fe (72.38%)> Hg (69.83%)> Mn (69.50%).





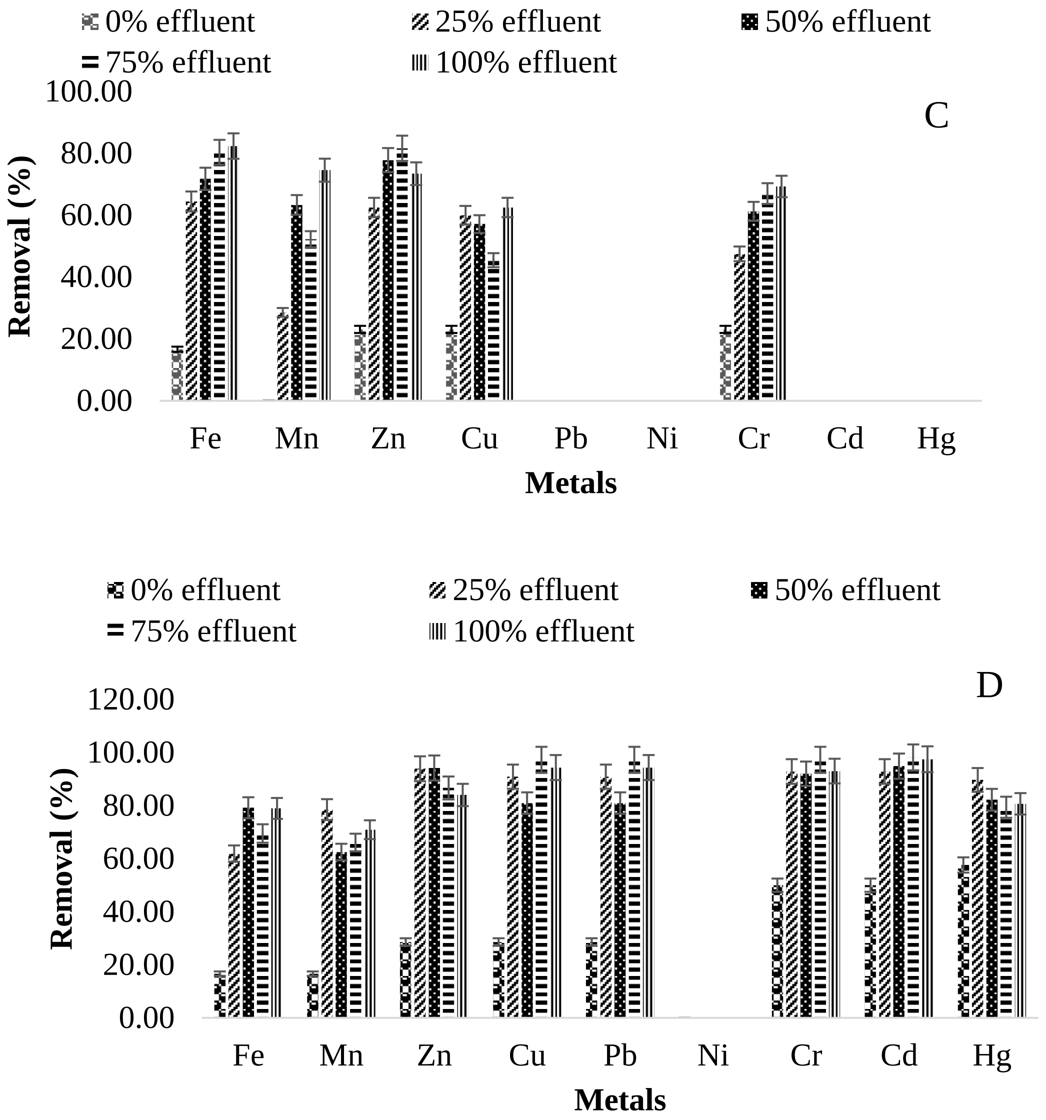


Fig 4.4.2. Removal efficiency of heavy metals by *L. gibba* from Petrochemical (A), Chemical (B), Thermal power plant (C) and Battery manufacturing industry (D).

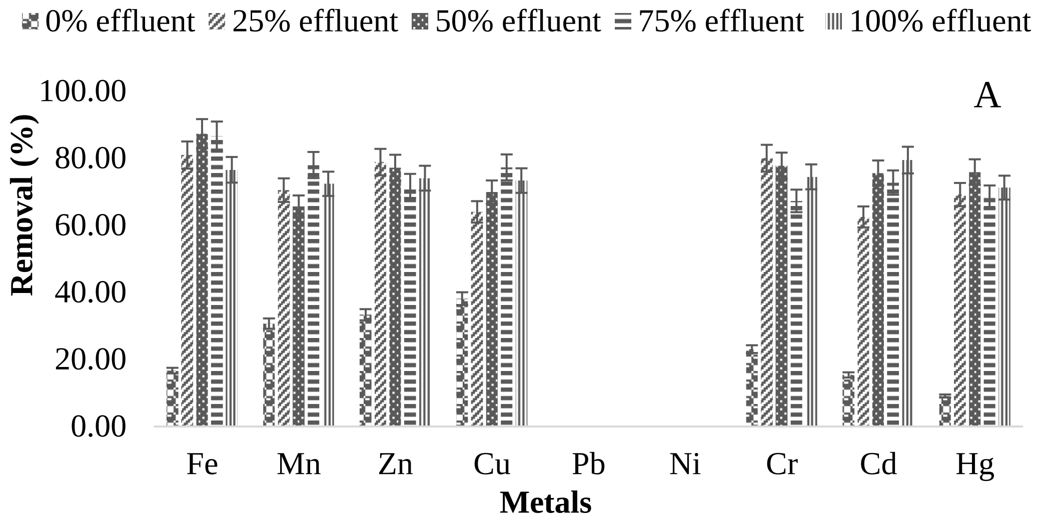
According to Vesely et al. (2011), *P. stratiotes* has good phytoremediation potential for removing Cd and Pb from contaminated solution. *P. stratiotes* removed 24.12, 81.70, 79.03,79.81 and 72.56% of Fe, 12.50, 72.73, 81.82, 79.41 and 83.98% of Mn, 17.65, 78.18, 79.44, 76.99 and 70.91% of Zn, 35.29, 78.18, 78.62, 85.97 and 78.43% of Cu, 23.12, 71.67, 76.36, 80.53 and 75.56% of Pb, 41.18, 80.28, 83.33, 85.07, and 83.75 % of Ni, 11.67, 70.25, 76.47, 78.13 and 80.19% of Cr and 25.00, 62.96, 76.15, 72.08 and 77.69% of Hg from 0, 25, 50, 75 and 100% concentrated petrochemical industry effluent respectively. 75% diluted effluent is efficiently removed heavy metals from petrochemical effluent. *P. stratiotes* strongly removed Ni (83.11%) followed by Cu (80.40%), Mn (79.47%), Fe (78.28%), Zn (76.38%), Cr (76.26%), Pb (76.03%) and Hg (72.22%). Das et al. (2014) investigated that *P. stratiotes* removed Cd from different concentration (5, 10, 15 and 20 mgl^{-1}) in hydroponic system for 21 days. Results shows that 48.08, 60.36, 65.19 and 70% Cd was decline by roots respectively. Similarly shoot decline 30.12, 54.8, 56 and 65.7% respectively. Ugya et al. (2015) also studied on *P. stratiotes* for heavy metals (Hg, Cd, Zn, Mn, Pb and Ag) of polluted stream contaminated by Kaduna refinery a petrochemical industry. Study shows that plant can be used effectively removed metals from stream.

In chemical industry effluent its removed 16.67, 80.95, 87.30, 86.61 and 76.55% Fe, 30.69, 70.48, 65.63, 77.97 and 72.37% Mn, 33.33, 78.85, 77.17, 71.74 and 74.05% Zn, 38.13, 64.00, 69.88, 77.27 and 73.33% Cu, 23.08, 80.00, 77.78, 69.27 and 74.44% Cr, 15.38, 62.50, 75.56, 72.73 and 79.44% Cd, 9.09, 69.17, 75.88, 68.46 and 71.23% Hg from 0, 25, 50, 75 and 100% concentrated effluent respectively. In this effluent, 100% concentration are effective for *P. stratiotes* then other dilution. *P. stratiotes* maximally removed Fe i.e. 83.43% followed by Zn (78.19%)> Cu (75.38%)>Mn (75.20%)>Cr (74.87%)>Cd (73.55%)> Hg (71.44%). Cd uptake by different floating macrophytes i.e. *S. herzogil*, *P stratiotes*, *Hydromistia stolanifera* and *E. crassipes* during low temperature periods of year and all species are considered efficient hyperaccumulator of Cd. *P. stratiotes* was found superior performer in all of them. Efficiency of *P. stratiotes* for Cd removal was found 63, 65, 72 and 74 % from 1, 2, 4, 6 mgl^{-1}. concentrated water solution respectively. Researchers also explain plant has ability to keep its capability removed Cd even some toxicity appeared in higher concentration (Maine et al., 2001). Shah and Reddy (2016) found that *P. stratiotes* is effectively used for removal of Cu from contaminated water bodies. Reddy and Kumari (2016) studies the phytosorption abilities of 2 macrophytes *E. crassipes* and *P. stratiotes* by used Cu, Cd and Zn at different concentration and suggested their used in the removal of heavy metals pollution in water system. *P. stratiotes* is efficient for metal sorption and hence potential candidate for treating water, contaminated with industrial wastewater. *P. stratiotes* can be used for removal of Mn, Zn and Pb and its biomass can be increase, scientist also explained that 10% concentration are tolerable but tolerability decrease with increasing concentration (Pillai, 2016).

In thermal power plants effluent *P. stratiotes* removed 16.67, 61.11, 64.71, 79.66 and 83.95 % Fe, 8.33, 28.57, 60.00, 67.78 and 76.82 % Mn, 23.08, 72.50, 75.63, 81.22 and 73.42% Zn, 7.69, 60.00, 71.43, 63.64 and 70.83% Cu, and 23.08, 50.00, 71.25, 57.00 and 72.14% Cr from 0, 25, 50, 75, and 100% concentration. *P. stratiotes* is effective most effective at 100 and 75% concentration i.e. 75.43% and 75%. It is maximally removed Zn with the 75.69% potential followed by Fe with 72.35%, Cu with 66.47%, Cr with 62.60% and Mn with 58.29%. *P. stratiotes* is used in laboratory experiment for rhizofiltration process for Al, Fe and Mn and found that its removed >90% of Al, Fe and Mn during 15 days of periods. Sasmaz et al., (2015), investigate removal efficiency of heavy metals (Cu, Pb, Zn, As) in gallery water in mining area of Keban, Turkey. Two species of Lemna (*L. gibba* and *L. minor*) are used to removal. Results explain that *L. minor* and *L. gibba* removed 36 to 87% Cu, 1015-1259% Pb, 382-628% Zn and 7070-19709% As during 2 to 3 days respectively. Study revealed that both species had high potential to remove Cu, Pb, Zn and As contaminated by different ores of mine.

In battery manufacturing industry wastewater *P. stratiotes* removed 16.67, 76.19, 67.57, 66.10 and 73.94% Fe, 28.57, 71.06, 85.36, 88.69 and 78.80% Mn, 15.38, 73.08, 74.78, 65.10 and 67.03 of Zn, 23.08, 75.00, 60.00, 74.55 and 66.67% Cu, 21.43, 70.91, 80.95, 87.57 and 84.53% Pb, 22.31, 69.75, 76.67, 72.73 and 84.44 % Cr, 23.08, 80.00, 65.56, 83.33 and 62.22% Cd and 27.27, 85.71, 79.30, 82.20 and 81.15% of Hg from 0, 25, 50, 75 and 100% concentration. At 75% concentration of wastewater, *P. stratiotes* is effective for all metals. Removal efficiency of *P. stratiotes* is very high i.e. >75% for heavy metals. Ni shows maximum removal percentage while Cr shows minimum. Removal percentage of the heavy metals through *P. stratiotes* are arrange in following sequence: Ni> Pb>Fe>Hg>Zn>Mn>Cd>Cu>Cr.

Three aquatic plants *P. stratiotes*, *H. verticilata*, and *S. molesta* were treated with different concentration of Hg ranging from 1 to 1000 mgl^{-1} during 1, 3 and 5 hr. exposure. Various perceptible effects like foliar injury, decrease of chlorophyll content and plant biomass was appear due to excess exposure (Mhatre and Chaphekar, 1985). Mishra and Tripathi (2008), investigated *P. stratiotes*, *E. crassipes* and *S. polyrrhiza* for removal of Fe, Cu, Zn, Cd and Cr from 1, 2 and 5 mgl^{-1} synthetic solution. Results revealed that plants removed >90% of metals during 15 days of experiment and maximum removal observed on 12th day than decrease. Results also revealed that *E. crassipes* is most effective plants of these heavy metals followed by *P. stratiotes* and *S. polyrrhiza*. Heavy metal accumulated in plants body without harming, shows higher tolerance to all metals. They proposed that these plants were also used for larger scale i.e. highly polluted wastewater.


0% effluent
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100% effluent
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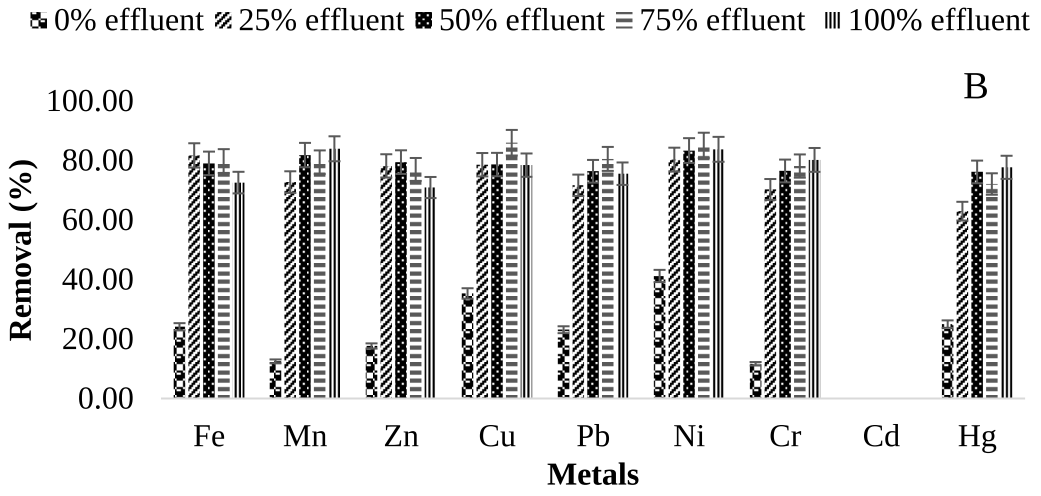
Metals


0% effluent
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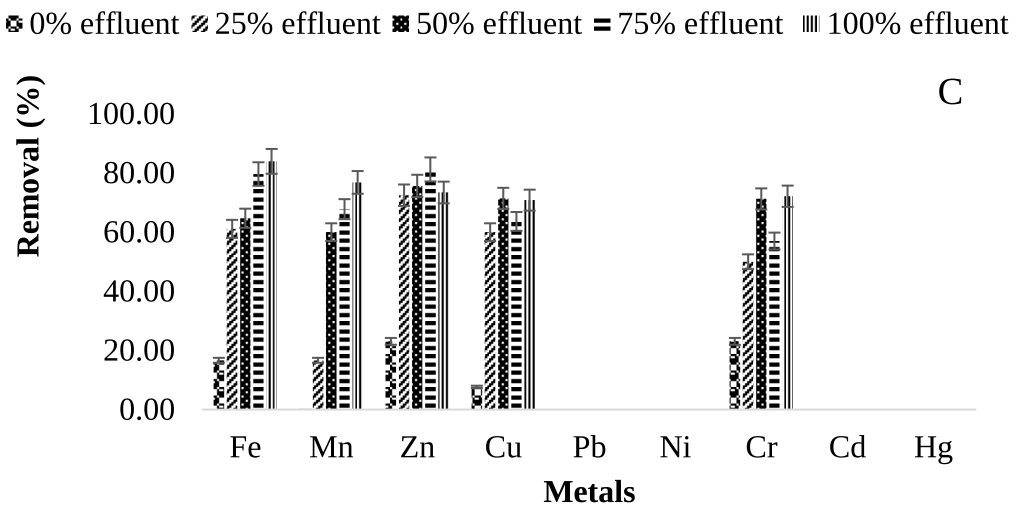
Pb
Ni
Cr
Cd
Hg
Metals


0% effluent
25% effluent
50% effluent
75% effluent
100% effluent
C
Removal (%)
100.00
80.00
60.00
40.00
20.00
0.00
Fe
Mn
Zn
Cu
Pb
Ni
Cr
Cd
Hg
Metals

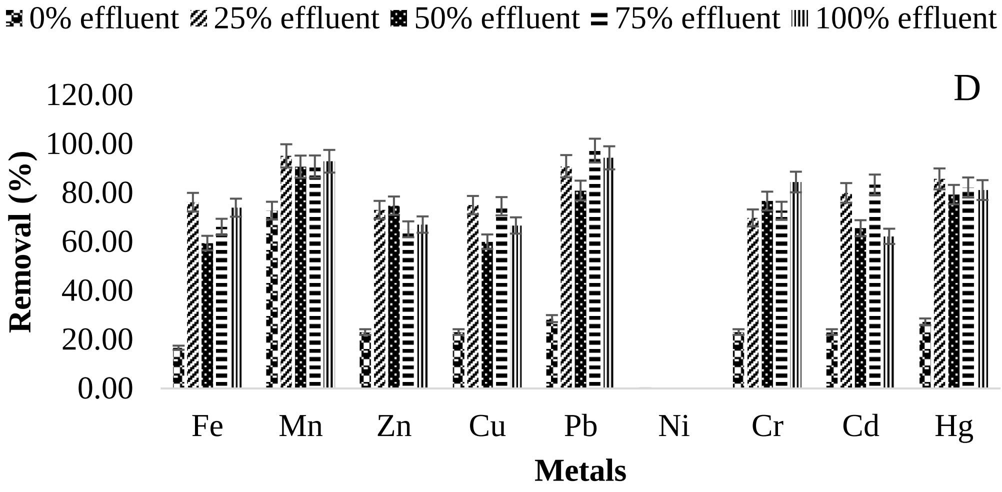


Fig 4.4.3: Removal efficiency of heavy metals by *P. stratiotes* from Petrochemical (A), Chemical (B), Thermal power plant (C) and Battery manufacturing industry (D).

The rate of the metal uptake is depended on the metal concentration in wastewater. The final metal concentration in wastewater and metal concentration in selected macrophytes ratifies that the main process of metal removal. *P. stratiotes* shows the highest removal efficiency resulting most appropriate for metal removal as compare to *E. crassipes* and *L. gibba*.

Enrichment coefficient factor (EC) is the ability of the aquatic macrophytes to accumulate heavy metals in their tissues which grown in different industrial wastewater. The greater the metal absorption, the higher the concentration of metals in the biomass, which is against their initial substrate concentration. As a basis for assessing the Enrichment coefficient of metals, the four-degree scale was adapted in which, when EC are < 0.01 means no accumulation; if value found in the range of 0.01–0.1 mean low EC; if range found in between 0.1–1.0 means medium EC; and when values is >1 means high EC (Michałowski and Gołas, 2001). It should be noted that the accumulation factors for all the examined metals were considerably higher for roots than for shoots.

E. crassipes accumulates higher level of Cd and their enrichment coefficient is calculated as 29.50, 30.53, 40.00, 53.00, 430 for root and for shoot it was calculated as 28.70, 21.95, 27.74, 38.67 and 330.33. After Cd, Cu and Cr are also have high enrichment coefficient values which is 21.40, 2.73, 20.43, 26.80, 23.67; 16.00, 18.40, 15.65, 25.45, 14.00 in Cu, 12.83, 11.60, 25.45, 53.00, 41.82; 12.83, 8.34, 17.65, 38.67, 32.76 in Cd for root and shoot respectively. *L. gibba* highly accumulates Cu their EC calculated as 46.19, 52.00, 28.04, 20.00, 21.40 for roots; 33.50, 38.67, 19.05, 14.19, 14.00 for shoots and 17.75, 20.71, 14.06, 5.61, 19.00 for roots, 14.50, 16.79, 9.69, 5.12 and 13.00 for shoots. Pb is also highly bioaccumulate in *L. gibba* i.e. 26.03, 23.85, 24.88, 20.00, 21.40 for roots and 17.94, 18.08, 19.77, 17.22, 2.08 for shoots. In case of *P. stratiotes* after Cu, Pb, Cr and Mn also show maximum enrichment coefficient values i.e. 6.07, 6.11, 3.32, 3.71, 1.45; 4.62, 4.68, 2.41, 2.61, 1.08 in Pb, 5.00, 4.60, 4.29, 6.61, 3.56; 4.58, 3.57, 3.75, 4.02, 1.84 in Cr and 5.47, 5.33, 7.40, 19.75, 35.00; 4.22, 4.60, 6.67, 14.08, 22.07 in Mn for root and shoots respectively.

Translocation factor (TF) is a ratio of heavy metals accumulated in root and shoot of the aquatic plants. TF is generally dependent on plant species, metals and other environmental factors i.e. pH, temperature, salinity etc. (Fritioff et al., 2005, Marbaniang and Chaturvedi 2014). When TF is >1 means plants is accumulator and if TF is <1 mean plant is excluder species. According to Lasat et al. (2000) If TF value is greater than 1, its indicates that metal transport from root to shoot through efficient metals transport system, and, metal sequestrated in shoot vacuole and apoplast. This type of plant potential known as hyperaccumulation and plant known as hyperaccumulators.

TF values for metals present in battery manufacturing effluent in all three macrophytes at different concentration are shown in table 4.4.1. In this effluent TF value for all the metals in all macrophytes at every concentration is greater than 1. Root of the macrophytes accumulates larger amount of the metals than shoot, which means limited translocation occur in macrophytes.

Table 4.4.1: Enrichment coefficient factor and translocation factor in macrophytes grown in Battery industry effluents

Eichhornia crassipes									
EC (Shoot)									
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Cd	Hg
100%	2.30	4.83	4.69	16.00	3.12		12.48	28.70	10.57
75%	2.77	3.74	6.41	18.40	3.55		8.34	21.95	13.80
50%	2.70	3.93	6.88	15.65	5.18		17.65	27.74	16.28
25%	2.48	3.98	13.41	25.45	2.70		38.67	38.67	10.38
0%	5.67	60.07	22.07	14.00	1.08		32.76	330.33	0.60
EC (Root)									
100%	2.51	5.61	6.90	21.40	4.19		12.83	29.50	18.70
75%	3.44	5.11	7.33	22.73	5.06		11.60	30.53	24.26
50%	3.62	4.48	9.45	20.43	6.52		25.45	40.00	28.67
25%	3.42	4.80	19.00	26.80	3.66		53.00	53.00	22.17
0%	6.17	52.87	35.00	23.67	1.45		41.82	430.00	0.60
TF									
100%	1.09	1.16	1.47	1.48	1.34		1.03	1.03	1.77
75%	1.24	1.37	1.14	1.46	1.43		1.39	1.39	1.76
50%	1.34	1.14	1.38	1.31	1.26		1.44	1.44	1.76
25%	1.38	1.21	1.42	0.89	1.35		1.37	1.37	2.14
0%	1.09	0.88	1.59	1.53	1.35		1.28	1.30	0.99
Lemna gibba									
EC (Shoot)									
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Cd	Hg
100%	3.97	2.86	4.83	33.50	17.94		1.54	1.37	0.60
75%	2.67	3.58	4.33	38.67	18.08		1.40	0.82	1.05
50%	5.32	5.68	6.30	19.05	19.77		1.90	1.77	0.54
25%	3.38	4.55	12.47	14.19	17.22		2.53	2.91	0.35

0%	60.00	23.07	22.07	14.00	2.08		3.60	2.80	0.26
EC (Root)									
100%	5.62	3.61	6.20	46.19	26.03		3.61	3.80	0.97
75%	4.33	4.78	5.60	52.00	23.85		2.00	2.55	1.77
50%	8.10	6.84	7.83	28.04	24.88		2.45	3.45	0.96
25%	6.88	5.50	14.56	20.00	28.89		3.46	3.98	0.77
0%	37.12	37.40	36.00	21.40	4.27		4.60	5.10	0.26
TF									
100%	1.42	1.26	1.28	1.38	1.45		2.34	2.77	1.62
75%	1.63	1.33	1.29	1.34	1.32		1.43	3.12	1.68
50%	1.52	1.20	1.24	1.47	1.26		1.29	1.96	1.78
25%	2.04	1.21	1.17	1.41	1.68		1.37	1.37	2.22
0%	0.62	1.62	1.63	1.53	2.05		1.28	1.82	0.99
Pistia stratiotes									
EC (Shoot)									
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Cd	Hg
100%	3.21	2.45	4.22	14.50	4.62		4.58	0.84	2.76
75%	2.40	2.01	4.60	16.79	4.68		3.57	2.09	2.39
50%	2.58	3.52	6.67	9.69	2.41		3.75	1.25	2.58
25%	5.00	4.02	14.08	5.12	2.61		4.02	3.87	1.43
0%	19.00	35.52	22.07	13.00	1.08		1.84	3.60	2.87
EC (Root)									
100%	1.18	2.69	5.47	17.75	6.07		5.00	2.34	4.64
75%	1.36	2.75	5.33	20.71	6.11		4.60	6.50	3.91
50%	1.87	3.92	7.40	14.06	3.32		4.29	2.45	3.34
25%	2.43	5.43	19.75	5.61	3.71		6.61	5.30	3.24
0%	34.33	48.55	35.00	19.00	1.45		3.56	4.60	2.86
TF									
100%	1.42	1.10	1.29	1.22	1.31		1.09	2.77	1.68
75%	1.67	1.37	1.16	1.23	1.31		1.29	3.12	1.64
50%	2.24	1.12	1.11	1.45	1.38		1.14	1.96	1.29
25%	2.04	1.35	1.40	1.10	1.42		1.64	1.37	2.26
0%	2.17	1.37	1.59	1.46	1.35		1.94	1.28	0.99

In the treatment of chemical industry effluent *E. crassipes* shows maximum Enrichment coefficient for Cd and Mn i.e. 5.81, 4.30, 5.35, 8.47 and 5.26, 4.15, 2.75, 2.97, 0.57 for roots and 5.18, 4.15, 2.81, 3.52 0.76 and 5.80, 4.28, 5.35, 8.10, 2.77 for shoot. After Cd and Mn; Zn and Hg i.e. 4.75, 4.87, 3.69, 3.22, 3.33; 4.67, 4.78, 3.61, 3.11, 2.90 in Zn, 4.27, 3.26, 3.69, 3.52, 6.13; 4.19, 3.18, 3.67, 3.45, 7.24 in Hg for roots and shoots both respectively. In case of *L. gibba* higher enrichment shown for Mn and Zn. In *L. gibba* EC was calculated as 5.48, 3.90, 2.75, 2.97, 0.55; 5.51, 4.18, 2.81, 3.52, 0.55 and 4.98, 4.23, 3.69, 3.22, 3.33 and 4.96, 4.04, 3.61, 3.11, 3.40 for roots and shoot in Mn and Zn respectively. After that Cd and Hg also shows higher level of enrichment coefficient factor which is 4.37, 3.21, 3.73, 3.46, 5.17; 4.29, 3.18, 3.72, 3.40, 6.28 and 3.70, 4.31, 4.31, 6.36, 3.06; 3.69, 4.26, 4.20, 6.08, 3.27 for root and shoot respectively. In *P. stratiotes* EC for Cd was calculated as 5.16, 4.53, 5.14, 5.71 2.24 and 5.09, 4.80, 5.61, 5.87, 2.52 for root and shoots respectively. After that Cd, Hg and Zn also higher bioacclumative their bioconcentration factor is calculated as 4.02, 2.9, 3.75, 3.25, 5.83; 3.83, 2.76, 3.83, 3.40, 6.93 and 3.07, 3.00, 3.69, 4.40, 4.17; 3.02, 2.94, 3.61, 4.24, 3.62 for roots and shoots respectively.

In chemical industry effluent TF of Fe at 75%, Mn at 50 and 25%, Cu at 50%, and Cr at 100, 50 and 25% effluent are less than 1 otherwise it is greater than one in *E. crassipes*. In *L. gibba*, Cr and Mn low TF, Cr at 50% effluent shows lowest translocation factor value i.e. 0.62. *P stratiotes* shows maximum translocation of Mn at all concentrated effluent except 100%. Mn is largely translocated in shoots part all three macrophytes may be because Mn is essential metal for plants.

Table 4.4.2: Enrichment coefficient and translocation factor in macrophytes grown in chemical industry effluent

Eichhornia crassipes										
EC (Shoot)										
		Fe	Zn	Mn	Cu	Pb	Ni	Cr	Hg	Cd
	100%	3.44	4.67	5.18	2.6	BDL	BDL	1.95	4.19	5.8
	75%	6.4	4.78	4.15	3.11			1.44	3.18	4.28
	50%	4.81	3.61	2.81	2.3			1.42	3.67	5.35
	25%	4.08	3.11	3.52	3.64			2.15	3.45	8.1
	0%	9.96	2.9	0.76	0.85			1.42	7.24	2.77
EC (Root)										

	100%	3.47	4.75	5.26	2.64			1.88	4.27	5.81
	75%	6.22	4.87	4.15	3.2			1.49	3.26	4.3
	50%	5.64	3.69	2.75	2.27			1.29	3.69	5.35
	25%	4.34	3.22	2.97	3.71			2.1	3.52	8.47
	0%	8.42	3.33	0.57	0.83			1.68	6.13	2.46
TF										
	100%	1.01	1.02	1.02	1.01			0.96	1.02	1
	75%	0.97	1.02	1	1.03			1.03	1.02	1.01
	50%	1.17	1.02	0.98	0.99			0.91	1	1
	25%	1.06	1.04	0.84	1.02			0.97	1.02	1.05
	0%	0.85	1.15	0.75	0.98			1.19	0.85	0.89
Lemna gibba										
EC (Shoot)										
		Fe	Zn	Mn	Cu	Pb	Ni	Cr	Hg	Cd
	100%	3.17	4.96	5.51	2.28			2.32	3.69	4.29
	75%	6.56	4.04	4.18	2.36			1.66	4.26	3.18
	50%	4.74	3.61	2.81	2.04			1.94	4.2	3.72
	25%	4.03	3.11	3.52	3.33			2.73	6.08	3.4
	0%	9.57	3.4	0.56	0.71			1.13	3.27	6.28
EC (Root)										
	100%	3.17	4.98	5.48	2.27			2.22	3.7	4.37
	75%	6.79	4.23	3.9	2.42			1.76	4.31	3.21
	50%	5.11	3.69	2.75	2.03	BDL	BDL	1.19	4.13	3.73
	25%	4.34	3.22	2.97	3.23			2.64	6.36	3.46
	0%	7.32	3.33	0.55	0.72			1.34	3.06	5.17
TF										
	100%	1	1	0.99	1			0.96	1	1.02
	75%	1.04	1.05	0.93	1.02			1.06	1.01	1.01
	50%	1.08	1.02	0.98	0.99			0.62	0.98	1
	25%	1.08	1.04	0.84	0.97			0.97	1.05	1.02
	0%	0.77	0.98	0.99	1.02			1.19	0.94	0.82
Pistia stratiotes										
EC (Shoot)										
		Fe	Zn	Mn	Cu	Pb	Ni	Cr	Hg	Cd
	100%	1.91	3.02	2.65	2.47			2.58	3.83	5.09
	75%	2.21	2.94	3.4	2.87			1.48	2.76	4.8
	50%	2.88	3.61	2.72	1.86			1.85	3.83	5.61
	25%	2.26	4.24	3.41	1.92	BDL	BDL	3.03	3.4	5.87
	0%	7.4	3.62	1	0.94			1.13	6.93	2.52
EC (Root)										
	100%	1.99	3.07	2.79	2.47			2.48	4.02	5.16
	75%	2.56	3	2.54	2.8			1.53	2.92	4.53

	50%	3.89	3.69	2.67	1.75		1.69	3.75	5.14
	25%	2.72	4.4	2.88	1.99		2.95	3.25	5.71
	0%	6.82	4.17	0.96	0.92		1.34	5.83	2.24
TF									
	100%	1.04	1.02	1.05	1		0.96	1.05	1.01
	75%	1.16	1.02	0.75	0.97		1.03	1.06	0.94
	50%	1.35	1.02	0.98	0.94		0.91	0.98	0.92
	25%	1.2	1.04	0.84	1.03		0.97	0.96	0.97
	0%	0.92	1.15	0.96	0.98		1.19	0.84	0.89

In petrochemical industry effluent *E. crassipes* and *L. gibba* shows maximum enrichment coefficient for Mn, and *P. stratiotes* for Ni. In *E. crassipes* it was found 9.64, 4.78, 8.47, 4.43, 0.91 and 9.96, 5.39, 9.21, 5.18, 0.91, in *L. gibba* it was calculated as 7.36, 6.84, 7.90, 8.17, 0.93 and 6.80, 6.80, 7.68, 8.30, 0.92 and in *P. stratiotes* it was calculated as 6.45, 7.06, 7.09, 6.41, 0.90 and 6.48, 6.62, 7.07, 5.81 and 0.92 for root and shoot respectively. After Mn *E. crassipes* also shows higher enrichment coefficient for Hg, Ni and Pb i.e. 5.13, 4.11, 3.52, 2.23, 6.31; 5.30, 3.12, 2.14, 1.97, 7.53 in Hg, 5.03, 4.26, 3.90, 1.96, 0.72; 5.01, 4.14, 3.89, 2.24, 0.74 in Ni and 4.87, 2.74, 1.90, 2.13, 0.53; 4.34, 3.04, 2.06, 2.47, 0.66 in Pb in both roots and shoots respectively. In *L. gibba* after Mn, Cr, Zn, Hg and Ni also shows higher level of enrichment coefficient factor which is 4.92, 5.11, 4.16, 2.03, 0.33; 4.22, 4.25, 3.60, 1.78, 1.16 in Cr, 3.50, 3.01, 4.25, 4.49, 0.07; 3.34, 2.7, 4.04, 4.26, 0.07 in Zn, 3.74, 4.43, 3.52, 1.37, 0.33; 3.17, 3.98, 3.28, 2.11, 1.16 in Hg, 3.78, 2.79, 3.39, 1.15, 0.32; 3.04, 2.24, 2.73, 1.88, 1.15 in Ni in roots and shoots respectively. In comparison to other metals *P. stratiotes* also shows higher enrichment coefficient for Hg, Cu, Mn and Cr i.e. 5.32, 3.56, 5.44, 2.97, 1.06; 5.42, 3.18, 3.73, 2.63, 1.10 in Hg, 4.85, 8.01, 4.84, 3.62, 0.15; 4.53, 7.30, 5.01, 4.27, 0.08 in Cu, 4.40, 2.80, 3.54, 281, 0.15; 4.32, 3.16, 3.85, 3.28, 0.14 in Mn, 4.07, 2.92, 3.60, 2.20, 9.07; 4.05, 2.84, 3.60, 2.49, 9.13 in Cr for both roots and shoot respectively.

Shoot of the *E. crassipes* translocate Fe at 50%, Zn at 100, 75 and 50%, Mn at all dilution, Cu at 25%, Pb at 75, 50 and 25%, Ni at 25%, Cr at 25% and Hg at 100% as compare to root which means higher translocation occur from root to shoots in these metals. *L. gibba* shows minimum TF values for Ni and Hg i.e. 0.61 and 0.65 at 25% effluent and maximum for Pb i.e. 1.65, at 25 % effluent. The decreased Zn at 75%, Mn at 75, 50 and 25, Cu at 50 and 25%, Pb at 50 and 25%, Ni at 75 and 25% C at 25 and Hg at 100% in the shoot of the *P. stratiotes* in study of chemical effluent may be due to casparian bands act effective barrier for movement of the metals in stele of the plants (Macfarlane and Burchett, 2000).

Table 4.4.3: Enrichment coefficient and translocation factor in macrophytes grown in petrochemical industry effluent

Eichhornia crassipes									
	EC (Shoot)								
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Cd	Hg
100%	3.72	3.69	9.96	2.78	4.34	5.01	3.97	BDL	5.30
75%	7.07	3.57	5.39	2.22	3.04	4.14	3.68		3.12
50%	4.33	4.20	9.21	2.09	2.06	3.89	3.51		2.41
25%	4.05	0.85	5.18	3.20	2.47	2.24	2.24		1.97
0%	0.35	0.92	0.91	0.76	0.66	0.74	0.80		7.53
	EC (Root)								
100%	3.85	3.04	9.64	3.07	4.87	5.03	3.99		5.13
75%	7.59	2.83	4.78	2.79	2.74	4.26	3.79		4.11
50%	4.16	3.75	8.47	2.16	1.90	3.90	3.51		3.52
25%	4.23	0.92	4.43	2.71	2.13	1.96	1.98		2.23
0%	0.38	1.82	0.91	0.76	0.53	0.72	0.80		6.31
	TF								
100%	1.03	0.82	0.97	1.10	1.12	1.01	1.01		0.97
75%	1.07	0.79	0.89	1.26	0.90	1.03	1.03		1.32
50%	0.96	0.89	0.92	1.03	0.92	1.00	1.00		1.46
25%	1.04	1.08	0.86	0.85	0.87	0.88	0.88		1.13
0%	1.09	1.98	1.00	1.00	0.81	0.98	0.99		0.84
Lemna gibba									
	EC (Shoot)								
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Cd	Hg

100%	2.19	3.43	6.80	2.29	2.45	3.04	4.22		3.17
75%	3.10	2.72	6.80	1.58	1.89	2.24	4.25		3.98
50%	2.17	4.04	7.68	2.43	1.87	2.73	3.60		3.28
25%	2.63	4.26	8.30	1.45	1.62	1.88	1.78		2.11
0%	0.49	0.07	0.92	0.99	1.16	1.15	1.16		1.16
	EC (Root)								
100%	2.26	3.50	7.36	3.05	2.82	3.78	4.92		3.74
75%	2.91	3.01	6.84	2.04	2.44	2.79	5.11		4.43
50%	2.15	4.25	7.90	2.92	2.88	3.39	4.16	BDL	3.52
25%	2.57	4.49	8.17	1.72	2.67	1.15	2.03		1.37
0%	0.48	0.07	0.93	0.14	0.33	0.32	0.33		0.33
	TF								
100%	1.03	1.02	1.08	1.33	1.15	1.24	1.17		1.18
75%	0.94	1.10	1.01	1.29	1.29	1.25	1.20		1.11
50%	0.99	1.05	1.03	1.20	0.15	1.24	1.15		1.07
25%	0.98	1.05	0.98	1.18	1.65	0.61	1.14		0.65
0%	0.99	1.00	1.01	0.14	0.29	0.28	0.29		0.29
					Pistia stratiotes				
	EC (Shoot)								
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Cd	Hg
100%	3.04	3.09	4.32	4.53	3.55	6.45	4.05		5.42
75%	3.69	3.78	3.16	7.30	4.86	7.06	2.84		3.18
50%	3.57	3.71	3.85	5.01	5.73	7.09	3.60		3.73
25%	4.46	3.18	3.28	4.27	5.80	6.41	2.49		2.63
0%	5.00	0.08	0.14	0.08	8.24	0.90	9.13		1.10
	EC (Root)								
100%	3.22	3.28	4.40	4.85	3.71	6.48	4.07		5.32
75%	4.01	3.67	2.80	8.01	5.40	6.62	2.92		3.56
50%	3.65	3.84	3.54	4.84	5.19	7.07	3.60	BDL	5.44
25%	4.80	3.32	2.81	3.62	5.02	5.81	2.20		2.97
0%	6.17	0.08	0.15	0.09	8.07	0.92	9.07		1.06
	TF								
100%	1.06	1.06	1.02	1.07	1.04	1.01	1.01		0.98
75%	1.09	0.97	0.89	1.10	1.11	0.94	1.03		1.12
50%	1.02	1.03	0.92	0.97	0.90	1.00	1.00		1.46
25%	1.08	1.04	0.86	0.85	0.87	0.91	0.88		1.13
0%	1.23	1.00	1.08	1.06	0.98	1.01	0.99		0.96

In thermal power plant effluent treatment *E. crassipes* shows maximum enrichment coefficient for Zn i.e. 5.88, 6.23, 3.73, 5.10, 0.51 and 5.70, 6.23, 3.82, 6.04, 0.79 in roots and shoots but *L. gibba* and *P. stratiotes* its shows for Fe i.e. 4.29, 4.40, 3.42, 2.33, 5.36 for roots 4.45, 4.32, 3.12, 2.31, 6.57 for shoots and 4.56, 3.37, 2.27, 2.63, 4.76 for roots and 4.62, 2.91, 2.06, 2.55 and 6.37 for shoots.

TF value of Fe at 100%, Zn at 50 and 25%, Mn at 100 and 25% and Cr at 50% in *E. crassipes* found less than 1 metals largely accumulated in shoots. In *P. stratiotes* and *L. gibba,* Mn shows minimum translocation factor for Zn at 25% concentrated effluent.

Table 4.4.4: Enrichment coefficient and translocation factor in macrophytes grown in thermal power plant effluent

				Eichhornia crassipes					
EC (Shoot)									
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Hg	Cd
100%	0.64	5.70	4.44	2.00	BDL	BDL	2.55	BDL	BDL
75%	3.93	6.23	13.92	2.34			1.67		
50%	4.27	3.82	12.73	2.91			1.82		
25%	6.04	6.04	6.40	3.76			4.55		
0%	3.19	0.79	4.40	0.44			1.63		
EC (Root)									
100%	0.48	5.88	4.31	2.17			2.63		
75%	4.37	6.23	16.67	2.60			1.73		
50%	6.31	3.73	12.54	3.05			1.72		
25%	7.05	5.10	6.77	4.03			5.04		
0%	3.79	0.51	3.90	0.39			1.84		
TF									
100%	0.75	1.03	0.97	1.08			1.03		
75%	1.11	1.00	1.20	1.11			1.04		
50%	1.48	0.98	0.99	1.05			0.94		
25%	1.17	0.84	1.06	1.07			1.11		
0%	1.19	0.65	0.89	0.89			1.13		
				Lemna gibba					
EC (Shoot)									
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Hg	Cd
100%	4.45	2.80	3.68	2.45	BDL	BDL	2.89	BDL	BDL
75%	4.32	5.32	3.88	2.68			2.74		
50%	3.12	3.63	11.58	3.88			1.17		
25%	2.31	4.70	6.40	3.95			1.17		
0%	6.57	0.61	4.40	0.43			1.63		

EC (Root)									
100%	4.29	2.94	3.57	2.65			2.97		
75%	4.40	5.20	3.95	2.87			2.66		
50%	3.42	3.80	10.37	4.07			1.04		
25%	2.33	3.97	6.77	4.23			1.23		
0%	5.36	0.37	3.90	0.38			1.69		
TF									
100%	0.96	1.05	0.97	1.08			1.03		
75%	1.02	0.98	1.02	1.07			0.97		
50%	1.09	1.05	0.90	1.05			0.89		
25%	1.01	0.84	1.06	1.07			1.05		
0%	0.82	0.61	0.89	0.88			1.04		
Pistia stratiotes									
EC (Shoot)									
	Fe	Zn	Mn	Cu	Pb	Ni	Cr	Hg	Cd
100%	4.62	2.99	3.95	3.36			3.01		
75%	2.91	5.42	5.34	3.10			1.94		
50%	2.06	3.97	10.61	5.82			1.67		
25%	2.55	6.40	6.40	3.95			2.27		
0%	6.37	0.79	4.00	3.67			1.63		
EC (Root)									
100%	4.56	3.08	3.84	3.23			3.10		
75%	3.37	5.41	5.45	3.55			2.01		
50%	2.27	3.87	9.62	6.10	BDL	BDL	1.57	BDL	BDL
25%	2.63	5.41	6.77	4.23			2.52		
0%	4.76	0.51	3.55	3.25			1.49		
TF									
100%	0.99	1.03	0.97	0.96			1.03		
75%	1.16	1.00	1.02	1.15			1.04		
50%	1.10	0.98	0.91	1.05			0.94		
25%	1.03	0.84	1.06	1.07			1.11		
0%	0.75	0.65	0.89	0.89			0.91		

In the study, it is proved that use of *E crassipes, P. stratiotes* and *L. gibba* are cheap and effective method for treatment of water. Due to their higher potential, it is also suitable for the cleaning industrial wastewater contaminated with heavy metals i.e. Zn, Mn, Cd, Cu, Cr, Fe, Ni, Pb and Hg. The sequence of heavy metal removal by *E. crassipes* are found Mn>Fe>Pb>Hg>Zn>Ni>Cd>Cr>Cu, by *P. stratiotes* are found Pb>Ni>Mn>Fe>Cd>Hg>Cr>Zn>Cu and *L. gibba* followed

Cd>Cu>Pb>Fe>Zn>Cr>Ni>Mn>Hg in all industrial effluent. Result of the study demonstrated that *E. crassipes* is highly efficient removal of Fe, Mn, Zn, Cr and Pb, *P. stratiotes* is highly efficient for Cu, Cd, Ni and Pb and *L. gibba* efficient only for Hg. The macrophytes roots accumulated heavy metal at the high level with small differences between individual water improvement modes. The Enrichment coefficient factors calculated for the above-ground parts, similarly as their counterparts for the roots, indicate a small effect of accumulation of metals in plants. Among the examined metals, the highest accumulation was exhibited by Cd in battery effluent treatment with *E. crassipes*. Translocation factor demonstrated that heavy metals are highly accumulated in roots of all the macrophytes because maximum. Therefore, growing macrophytes in water contaminated with industrial effluent can help to control heavy metal pollution and reduce health risks.

Chapter 4.5

Identification and application of suitable aquatic macrophytes for the removal of heavy metals from different industrial wastewater

Identification and application of suitable aquatic macrophytes for the removal of heavy metals from different industrial wastewater.

Macrophytes having ability to uptake, store, transport large quantity of heavy metals in their body parts such as root and shoots. This objective aims to identify suitable aquatic macrophytes that can uptake heavy metals from industrial wastewater and accumulates in its tissues or membranes. This study evaluates the efficiency of metals removals by identified species and retention time required by plants for their effective removal. Identification method depends on proper selection macrophytes that has higher translocation potential (Nazir et al., 2011). Which macrophytes shows more than 1 enrichment coefficient and higher root to shoot metal translocation are ideal for for phytoremediation and act as hyperaccumulator (Hadi et al., 2014).

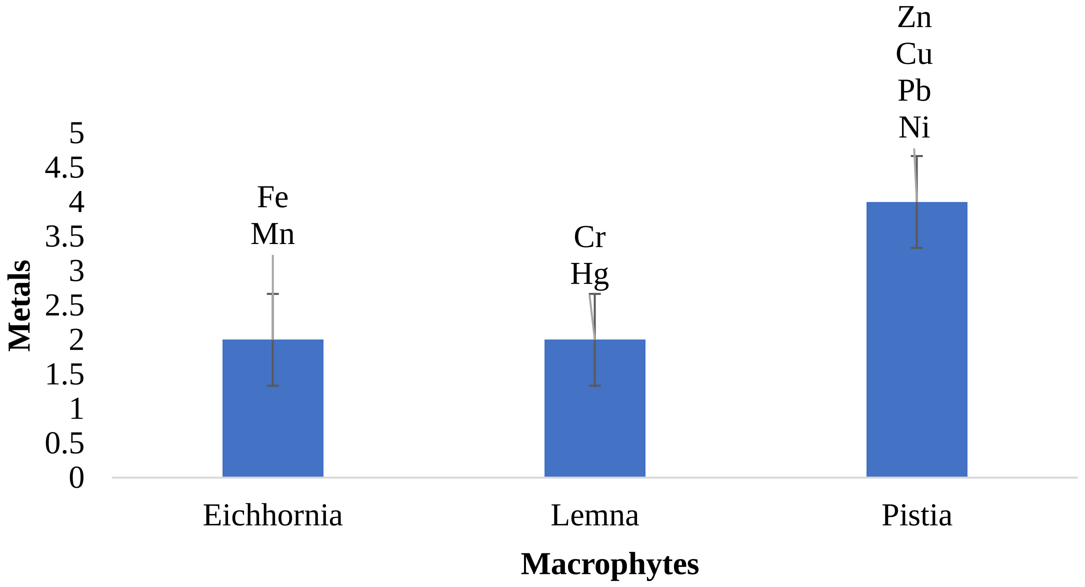


Fig. 4.5.1: Count of metals removed by macrophytes from petrochemical industry effluent

In petrochemical industry effluent, *P. stratiotes* identified as suitable macrophytes because it removes highest metals i.e. Zn, Cu, Pb and Ni as compare to *E. crassipes* (Fe, Mn) and *L. gibba* (Cr, Hg) from effluents. Ugya et al., (2015) studied on *P. stratiotes* used in wastewater of petrochemical industry and found that its effectively removed Hg, Cd, Mn, Ag, Pb and Zn. Vesely et al., (2011) suggested that *P. stratiotes* good rhizofiltration potential to remove Cd and Pb from contaminated solution accompanied by a high tolerance to heavy metal stress. Therefore, *P. stratiotes* a good capability for phytoremediation of wastewater.

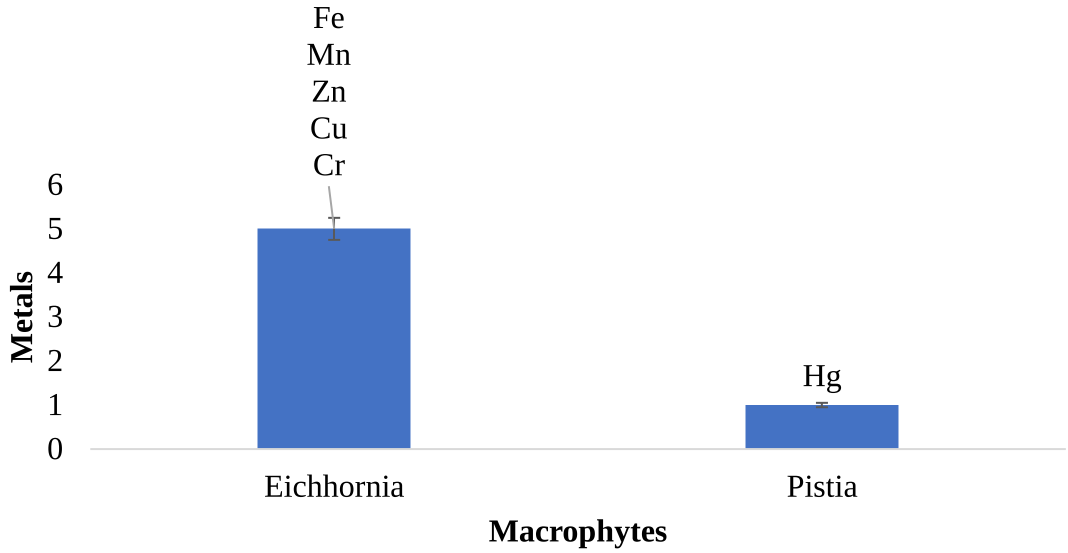


Fig. 4.5.2: Count of metals removed by macrophytes from Chemical industry effluent

In case of chemical industry effluent, *E. crassipes* shows suitable application which removes maximum metals Fe, Mn, Zn, Cu, and Cr. *P. stratiotes* applicable only for Hg removal and *L. gibba* is not applicable in any metals in chemical industry effluent. Ismail et al., (2015) used *E. crassipes* and *P. stratiotes* in treatment of wastewater and found *E. crassipes* is better performs in compare to *P. stratiotes.* Mishra and Maiti (2017) evaluates that E. crassipes is infamous macrophytes but being the most promising for removal of heavy metals from wastewate. It has been successfully used to accumulate heavy metals, dyes, radionuclides, and other organic and inorganic contaminants from water at laboratory, pilot, and large scale.

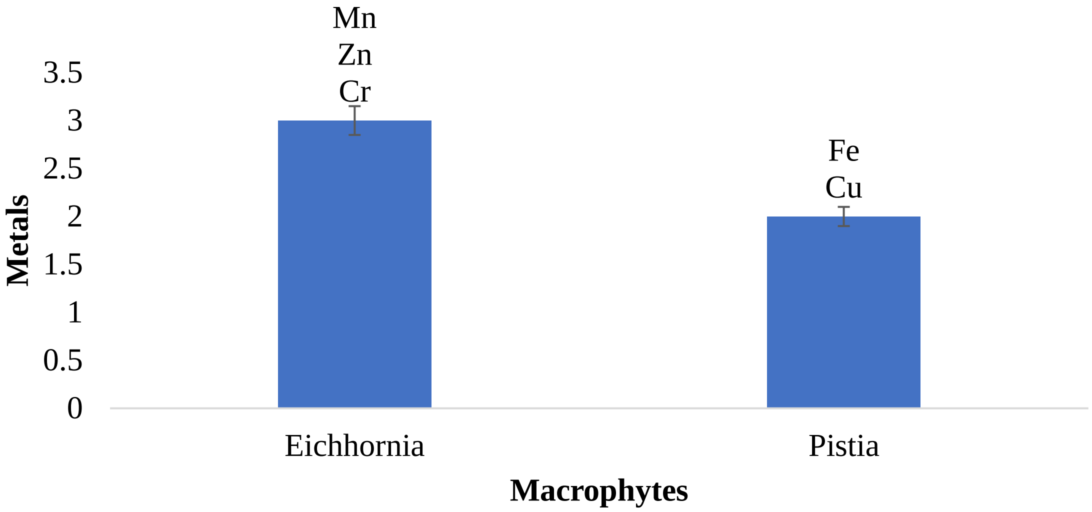


Fig. 4.5.3: Count of metals removed by macrophytes from Thermal power plant effluent

In case of thermal power plant in which only 5 metals i.e. Mn, Zn, Cu, Cr and Fe are detected in effluent, out of which Mn, Zn and Cr are efficiently removed by *E. crassipes* and Fe and Cu removed by *P. stratiotes*. So, we can say that *E. crassipes* are suitable in thermal power plant effluent. Ashok et al. (2014) studied that *E. crassipes* has the efficiency to with stand all metal contamination of the heavy metals and it can be used for removal of heavy metals from metal polluted water bodies. Ingole and Bhole (2003) also concluded that this plant could efficiently remove Zn and Pb from wastewater when its concentration is less than 10 mgl^{-1}.

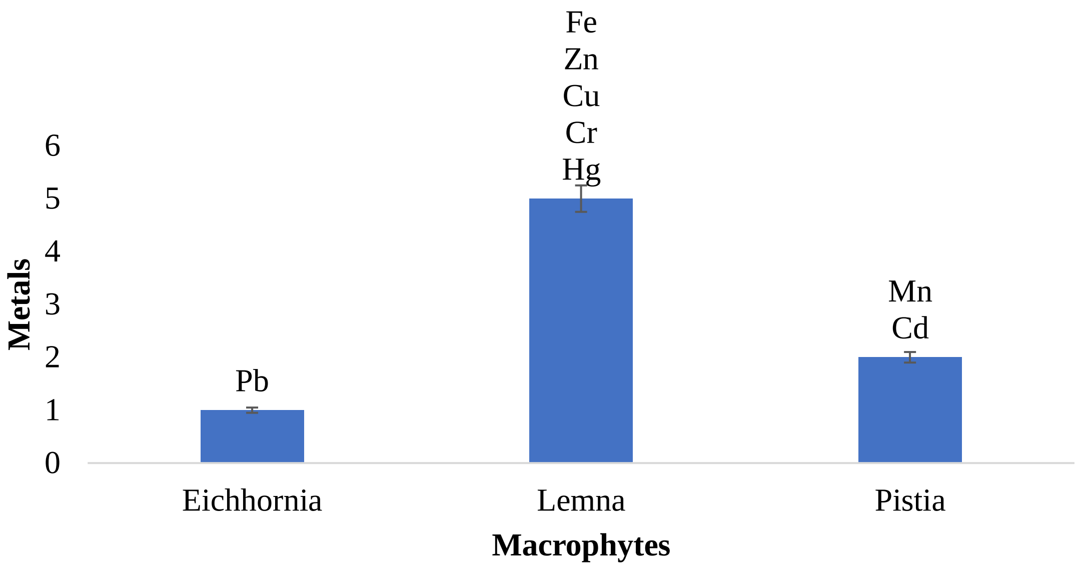


Fig. 4.5. 4 Count of metals removed by macrophytes from battery manufacturing industry effluent

In Battery manufacturing industry effluent, *E. crassipes* efficiently removed Pb and *P. stratiotes*, Mn and Cd while *L. gibba* removed Fe, Zn, Cu, Cr and Hg. Viet et al. (1988), proved that *L. gibba* is excellent bioaccumulator for various heavy metals and allowed it to treat a variety of wastewater industrial and municipal. Hammouda et al. (1995) evaluated the efficiency of *L. gibba* in various water system contaminated with heavy metals, suggested maximum reliability of system with mixture containing higher pollutants. Demirezen and Akbulut (2011) examined the growth characteristics of Lemna gibba in presence of wastewater with circulation. The effectiveness of *L. gibba* for removal of Pb, Mn, Ni and Cu also investigated and found best results.

In the conclusion *E. crassipes* are suitable for chemical industry and thermal power plant effluent, *P. stratiotes* are suitable for petrochemical industry, and *L. gibba* are suitable for battery manufacturing industry.

Chapter 5
Conclusion

CONCLUSION AND SUMMARY

Contamination in freshwater system due to direct discharge of industrial wastewater loaded with inorganic and organic contaminants such as heavy metals, metalloids, oil and grease and other petrochemicals etc. causes serious threats to plants and animals health. Constrains associated with heavy metal pollution includes their non-biodegradability, persistence nature, bioaccumulation in food chains. Since metal pollution affects the quality of water thereby affecting human and plant lives directly. Over last two decades many efforts have been done by several researchers in order to reduce the heavy metal contamination. In recent years, due to water scarcity, industrial wastewater is used for irrigation purposes which lead to detrimental effect on plants growth and reduction in crops yield and quality. Therefore, it is necessary to study the effects of industrial wastewater on crop system before irrigation.

Many conventional methods like chemical precipitation, ion exchange, coagulation, reverse osmosis, adsorption, flocculation, nanofiltration etc., has been used for wastewater treatment, however certain limitations like expensiveness and sensitive to operate, toxic sludge generation, extra operational cost for sludge disposal etc., make these techniques unaffordable and unsustainable. Therefore, a cost effective, more cleaner and eco-friendly alternative are required to sustainably remediate the industrial wastewater to minimize the health hazards. The efforts made in this area of wastewater treatment include the use of green technologies like bioremediation and phytoremediation. Most of the research work carried in this field has been conducted using hydroponic cultures with plant and most of which are invasive in character. For a successful phytoremediation technique, it is recommended that the selected plants should be perennial in nature, have high biomass production, tolerance and survival rate under different biotic and abiotic stress conditions.

Present work was undertaken to evaluate the wastewater quality from petrochemical, chemical, battery manufacturing industries and thermal power plant, situated at, Auriaya Kanpur, Lucknow and Kanpur respectively. The present study were also demonstrated the potential of aquatic macrophytes *viz. E. crassipes, P. stratiotes* and *L. gibba* for remediation of industrial wastewater contaminated with heavy metals. Comparative studies of selected macrophytes for their remediation potential for different industrial effluent concentration were also examined. Phytotoxicity caused by industrial effluents was also assessed in terms of seed germination. The selected seeds used for phytotoxicity assessment were seeds of Rice (Basmati- 198, Pusa sugandh and sambhamsoori), pulse (Chick pea, Moong N-1, and Massor VL-1) and Oil seeds (Mustard BR-40, Alsi KL-43 and Sesame G. til-3) varieties. The following studies were combined from these analyses.

Analyse the physicochemical characteristics including heavy metals of different industrial wastewater

Physico-chemical characteristics of industrial wastewater showed presence of high level of heavy metals *viz.* Fe, Zn, Mn, Pb, Cu, Cr, Cd, Ni and Hg and other organic and inorganic pollutants like chloride, oil and grease etc. which can cause major public health concern. The objective of the study was to evaluate the status of pollution created by industrial effluent. pH of the effluents was slightly alkaline nature and EC was slightly higher than prescribed level of CPCB. Total Suspended Solid (TSS) composed of silt, decaying matter and suspended waste was three times higher than prescribed limit of CPCB in three industries i.e. petrochemical, chemical and battery manufacturing industry effluent. However, in case of thermal power plant effluent TSS was found to be slightly lower than other industrial effluent but higher than the prescribed limits of CPCB. Chloride concentration was found to be higher in battery and chemical manufacturing industries due to application of chlorine compound. Dissolve oxygen (DO) of effluent were low because of high microbial activities resulting high level of biological oxygen demand (BOD). Oil and grease are important class of pollutant which is also toxic to aquatic plant and animals and carcinogenic to human beings. Effluent generated from petrochemical and chemical industries show high level of oil and grease. Nitrate and phosphate concentration were found under prescribed limits. Concentration of heavy metals *viz.* Hg, Mn, Pb, Cd in battery manufacturing Fe, Zn, Ni, Cr in petrochemical and Cu in chemical industry were found to be higher. Further, the thermal power plant effluent was found to be least polluted as compare to others effluent. The overall results showed that the presence of high level of heavy metals contamination and on prolong use of these effluent for irrigation purpose may leads irreparable ecological harm which could not be masked by short term economic prosperity. Therefore, the effluent generated from all selected industries should be treated prior to apply in agricultural field.

Evaluation of phytotoxicity of different industrial wastewater through seed germination test on different varieties of crop seeds

Presence of inorganic and organic contaminants has potential to reduce growth of microorganism, seed germination and can cause other environmental problems. Phytotoxic effects of different industrial wastewater were examined in terms of germination rate of rice, pulse and oil seeds. Seed germination rate is considered as an important parameter for evaluation of toxicity caused by wastewater. Seeds showed different sensitivities to toxic nature of effluents. Maximum seed germination was observed in control seed which are moisten by tap water. In the case of treatment of 50% effluent concentration a high percentage of seed germination was observed in compare to treatment of 100% effluent concentration. Result also indicated that the rice was more the susceptible crop while oilseeds were more tolerant crops towards toxicity caused by effluents. Salt (high TDS and TSS) concentration also reduced the seed germination rate. Rice, pulses and oil seeds varieties show maximum germination in petrochemical effluent except Moong and Alsi. Further, Moong and Alsi showed high percentage of seed germination in thermal power plant effluent. In case of battery manufacturing industry effluent all seeds showed lowest seed germination percentage. The results also indicated that 50% dilution of effluent was appropriate for irrigation of crops. On increasing the effluent concentration resulted decrease in seed germination rate, tolerance index, seedling vigor index. The obtained results also help in screening of seeds which was able to tolerate the toxic effects of various industrial effluents. Among all selected industries, effluent of petrochemical industry was found to be more toxic to chick pea and Sambha masuri while chemical industry effluent was more toxic for Pusa Sugandh and Sambha masuri. Similarily, thermal power plant effluent was found to be more toxic for Alsi while battery manufacturing industry showed its high toxicity potential to Basmati, Pusa sugandh, moong and Chick pea.

To study the partitioning of metals within different aquatic weeds exposed to different industrial wastewater.

Accumulation of Fe, Zn, Mn, Pb, Cu, Cr, Cd, Ni and Hg were investigated in three aquatic macrophytes *E. crassipes, L. gibba* and *P. stratiotes*. On the basis of obtained results, all the selected aquatic macrophytes can be considered good accumulators for all the tested heavy metals. Accumulations of metals were found to be higher in roots than shoots. Although, variations in level of heavy metal accumulation among selected macrophytes were observed. Metal concentration in tissue increased gradually with increase in the metal concentration in growing medium along with duration of treatment. Zinc (Zn) was the most accumulated metal among all metals because Zn is essential for plants requires for carbon dioxide utilization, carbohydrate and phosphorus metabolism and synthesis of RNA and auxin. Mercury (Hg) was least accumulated metal which was found to be highly toxic for plants. Study also concluded that *E. crassipes* was most effective in accumulating Fe, Mn, Zn and Pb while *L. gibba* was effective in accumulating Cr and Hg only. Similarly, *P. stratiotes* was found to be effective for accumulation of Cu, Cd and Ni. Further, *E. crassipes* can be considered as a most suitable species for phytoremediation due to their higher biomass, well developed fibrous root system and ability to absorb high amount of heavy metals from contaminated growing medium.

Comparative study on the phytoremediation potential of various aquatic weeds to remediate heavy metals from different industrial wastewater

The results shown that application of *E crassipes*, *P. stratiotes* and *L. gibba* for treatment of wastewater can be considered as an effective method. Due to their high accumulation potential, these macrophytes can be suitable candidates for the remediation of industrial wastewater contaminated with heavy metals *viz.* Zn, Mn, Cd, Cu, Cr, Fe, Ni, Pb and Hg. The sequence of heavy metal removal by *E. crassipes* was found as follows: Mn>Fe>Pb>Hg>Zn>Ni>Cd>Cr>Cu. The sequence of heavy metal removal by *P. stratiotes* was Pb>Ni>Mn>Fe>Cd>Hg>Cr>Zn>Cu while in case of *L. gibba* trend of metal removal efficiency was Cd>Cu>Pb>Fe>Zn>Cr>Ni>Mn>Hg in all industrial effluent. Result also revealed that the *E. crassipes* was highly efficient for removal of Fe, Mn, Zn, Cr, Pb; *P. stratiotes* for Cu, Cd, Ni, Pb and *L. gibba* for Hg. Enrichment coefficient factors were calculated for the roots and shoots separately. The results revealed that the accumulation of metals in roots was higher than the shoots. Among the examined metals, the highest accumulation was exhibited for Cd in battery effluent treatment with *E. crassipes*. Translocation factor demonstrated that heavy metals were highly accumulated in roots of all the macrophytes indicated the less mobilization of metal from roots to shoots.

Identification and application of suitable aquatic macrophytes for the removal of heavy metals from different industrial wastewater

In the present study, all the macrophytes were found suitable for removal of heavy metal from different industrial wastewater. Applicability of macrophytes for wastewater treatment may be different for different industrial wastewater. It was also found that the *E, crassipes* were suitable for chemical industry and thermal power plant effluent while *P. stratiotes* for petrochemical industry and *L. gibba* for battery manufacturing industry. Furthermore, accumulation of metals were much higher in roots than shoots indicating that these aquatic macrophytes may be a better option for the phytoremediation of heavy metals from industrial wastewater.

Chapter 6
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Publications

Publications

Research articles

1. Sangeeta Anand, Sushil Kumar Bharti, Dhananjay Kumar and Narendra Kumar* (2016). Phytoremediation of flashlight manufacturing effluent through aquatic macrophytes. International Journal of Science, Technology & Society (IJSTS).
2. Sangeeta Anand, Dhananjay Kumar, Sushil Kumar Bharti and Narendra Kumar*, (2017). Phytotoxicity assessment of petrochemical industry effluent and phytoremediation potential of plants growing naturally at contamination site. IJGHC, Sec. A; 6(4): 232-241.
3. Sangeeta Anand, Sanjeev Kumar and Narendra Kumar* Phytoremediation of inorganic and heavy metals through aquatic macrophytes from Flashlight manufacturing industry effluent, communicated in Geophytology. (Accepted)

Book chapter

1. Sangeeta Anand, Sushil Kumar Bharti, Neetu Dviwedi, S.C. Barman and Narendra Kumar. Macrophytes for the reclamation of degraded water bodies with potential for bio-energy production. Springer, New York.