Chapter 4
Phytoremediation of Heavy Metals and Pesticides Present in Water Using Aquatic Macrophytes

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Abstract Heavy metals occurring naturally on the earth are used in various industrial activities, whereas pesticides are man-made products used for protecting the crop. Heavy metals are inorganic contaminants and aggravated due to their long-term persistence, whereas pesticides encompass a variety of different types of chemicals including herbicides, insecticides, fungicides, and rodenticides. Hence, remediation of water contaminated by heavy metals and pesticides seeks urgent attention. Phytoremediation is an efficient alternative and less expensive method to strip heavy metals and pesticides directly from the water. Some of the aquatic plants used for removal of heavy metals and pesticides from water are duckweed (*Lemna minor*), water hyacinth (*Eichhornia crassipes*), *Hydrilla* (*Hydrilla verticillata*), water spinach (*Ipomoea aquatica*), water ferns (*Azolla caroliniana*, *Azolla filiculoides*, and *Azolla pinnata*), water cabbage (*Pistia stratiotes*), etc. Molecular tools are used to understand the mechanisms of uptake, sequestration, translocation, and tolerance in plants. The purpose of this review is to assess the current state of phytoremediation as an innovative technology and potential of aquatic macrophytes in remediation of water contaminated by heavy metals and pesticides.

Keywords Phytoremediation · Heavy metals · Pesticides · Water · Macrophytes

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4.1 Introduction

Water is very valuable for agriculture and as a natural resource. Unfortunately, during the recent decades, overexploitation of natural resources by various human activities such as industrialization, urbanization, disposals of wastewater, and unplanned agricultural practices has resulted in enormous amount of contaminants in water. Heavy metals and pesticides released by anthropogenic activities beyond toxic limits are continuously threatening the life of human beings (Zhang et al. 2009; Ishaq and Khan 2013; Arora et al. 2018). The point source contaminants include metal smelting and mining effluent from industries, while nonpoint sources include fertilizers and pesticides from agricultural run-off (Kumar et al. 2018a). Each pollutant has its own deleterious effects on flora and fauna, but the addition of heavy metals and pesticides into the water is a growing concern. Heavy metals such as lead (Pb), mercury (Hg), arsenic (As), copper (Cu), zinc (Zn), and cadmium (Cd) and pesticides like endosulfan, dichlorodiphenyltrichloroethane (DDT), mevinphos, ethion, copper sulfate, are highly toxic when absorbed in plants and animals (Kumar et al. 2016, 2018b).

Elements having density between 5.306 and 22.00 g/cm^2 are termed as heavy metals and these originate both from natural and anthropogenic sources (Gall et al. 2015). These metals are leading contaminants for environment because of being non-biodegradable and can be transferred through trophic levels and accumulate in the biota insistently (Nancharaiah et al. 2016; Kumar et al. 2018c). Some metals such as manganese (Mn), Zn, cromium (Cr), molybdenum (Mo), iron (Fe), and nickel (Ni) are essential at low concentrations for healthy function of biota but toxic at higher concentration, and some are non-essential and extremely toxic even at very low concentration including Pb, Hg, and Cd (Nagajyoti et al. 2010; Prasad 2011; Chibuike and Obiora 2014; Rezania et al. 2016). Heavy metals such as Cd, Pb, Zn, Hg, Mn, Cu, Cr, Ni, and Fe released from various industries are toxic and hazardous. They enter into food chain and, if are beyond limits, then can accumulate in plants, animals, and humans causing serious health hazards (Babel and Kurniawan 2004; Barakat 2011; Sood et al. 2012). A summary of several anthropogenic sources of heavy metals, their effects on health, and the available control techniques are presented in Table 4.1.

Pesticides consist of a large group of chemicals that are used throughout the world as insecticides, herbicides, fungicides, molluscicides, rodenticides, nematicides, and plant growth regulators to control unwanted plants, pests, and diseases to improve the productivity of food (Agrawal et al. 2010). The major groups of chemical pesticides include organochlorines, organophosphates, carbamates, and pyrethroids. Pesticides target different types of pests and their constant exposure also impacts non-target species, and this can lead to induced toxicity once it crosses the threshold limit in the system and food chain resulting in depleted biodiversity and health of ecosystems including humans (Kumar et al. 2018b; Arora 2018a) (Table 4.2).

Table 4.1 Sources of heavy metals, their health effects, and control techniques available

Metals	Sources	Health effects	Available control techniques	References
As	Paints, dyes, drugs, soaps, fertilizer	Weakness, pigmentation, nausea, peripheral nervous system failure, cardiovascular failure, DNA breakdown	Reverse osmosis or nanofiltration	Duarte et al. (2009) and Vaclavikova et al. (2008)
Cd	Mining and smelting activities, pigments, paints, electroplating, batteries	Hypertension, weight loss, hypochromic anemia, pulmonary fibrosis	Anaerobic digestion activated sludge process	Jaishankar et al. (2014)
Pb	Pigment, paints, batteries, industrial smelting, ceramics, Ayurvedic herbs, Troy	Osteoporosis, inhibits formation of hemoglobin, loss of IQ, high blood pressure, anemia, gastrointestinal effects	Chemical precipitation	Singh et al. (2011a, b) and Naseem and Tahir (2001)
Hg	Pesticide, dental filling, switches, light bulbs, batteries	Damage brain, neurological and renal disturbances, tremor, memory problem, lung damage	Ion exchange process, carbon adsorption	Jaishankar et al. (2014)
Selenium (Se)	Plastics, paints, rubber, preparation in drugs, feed additive, anti-dandruff shampoo	Hair loss, stomach pain, difficulty in breathing	Biosorption	Jaishankar et al. (2014)
Cr	Rocks, electroplating magnetic tapes, paints, cement, rubber and paper, etc.	Nose ulcers, breathing problems, asthma, damage kidney and liver	Stabilization pond	Jaishankar et al. (2014)
Ni	Cigarettes, diesel exhaust, electroplating, pigment, arc welding, dental materials	Lung cancer, nose cancer, sickness and dizziness, respiration failure, asthma and chronic bronchitis, allergic reaction such as skin rashes	Activated sludge process	Cempel and Nikel (2006)

4.2 Conventional Methods Used for the Removal of Heavy Metals and Pesticides

In order to maintain water quality standards, it is essential to remove heavy metals from wastewaters. Various conventional processes are being used for removal of heavy metals from wastewater such as chemical precipitation, reverse osmosis, ion exchange, and electrochemical deposition. Toxic heavy metals required to be removed from wastewater include Zn, Cu, Ni, Cd, Pb, and Cr (Fenglian and Wang 2011). Conventional physical and chemical methods for removal of heavy metals

Table 4.2 Pesticides, their health effects, and available control techniques

Pesticides	Health effects	Available control techniques	References
Organochlorine (endosulfan, DDT, dieldrin, alachlor, atrazine, lindane, and methoxychlor)	Cancer, eye, liver, and kidney problem	Activated carbon adsorption	Mnif et al. (2011)
Organophosphate (malathion, ethion, and phorate)	Nervous system problem	Filtration and centrifugation	Sullivan and Blose (1992)
Carbamate (aldicarb, carbosulfan, carbofuran, and carbaryl)	Muscle weakness, dizziness, sweating	Activated carbon adsorption	Sullivan and Blose (1992)
Pyrethroid (permethrin, deltamethrin, bifenthrin, cyfluthrin, and barthrim)	Toxic to nervous system, nausea, vomiting, headache	Activated carbon adsorption	Mnif et al. (2011)

are costly and time consuming and result in formation of secondary pollutants apart from being non-sustainable as well (Namasivayam and Ranganathan 1995; Mishra et al. 2017). However, chemical precipitation is still the most widely used method for heavy metal removal from effluents. A summary of various conventional techniques used for these treatments of wastewater along with the associated limitations are presented in Tables 4.3 4.4, 4.5, 4.6, and 4.7.

Although these techniques are effective for remediation purposes, they have significant risks in the excavation, transportation, handling, and disposal of toxic by-products. Other drawbacks are the extremely higher operational cost and small-scale application; lack of knowledge, especially for incineration; and also increase in the exposure rate. Therefore, the restoration of contaminated aquatic ecosystems requires ecological and cost-effective remediation technologies. Phytoremediation is a technique in which plants are used for remediation of contaminated water, soil, and sediments (Kumar et al. 2013a; Bauddh and Singh 2015). This technology is used for the removal of heavy metals, radionuclides, nutrients (nitrate, phosphate, etc.), solvents, explosives, crude oil, and organic pollutants such as persistent organic pollutants (POPs), polycyclic aromatic hydrocarbons (PAHs), and pesticides from wastewater and soil by using plants (Kumar et al. 2013b; Arora 2018b). Phytoremediation is a novel, eco-friendly, cost-effective, solar-driven, and in situ applicable remediation strategy (Kalve et al. 2011; Singh and Prasad 2011; Sarma 2011; Vithanage et al. 2012).

In the last two decades, using plants for metal and pesticides removal has attracted more attention (Jha et al. 2010). According to sciencedirect.com, a total of 5647 articles are published containing the term “phytoremediation” since the last 16 years (Fig. 4.1). Phytoremediation is also a set benchmark to assess the patent and research article development compared with other alternative strategies. The average annual percentage of phytoremediation is higher in patents and research (12% and 24%) versus bioremediation (4% and 12%), remediation (6% and 12%), and constructed wetland (14% and 16%) from 1999 to 2011 (Keomel et al. 2015).

Table 4.3 Various applicable conventional methods of wastewater treatment and their associated disadvantages

Methods	Application	Disadvantage	References
Reverse osmosis	Separated by semipermeable membrane	Clogging of membrane and expensive	Singh et al. (2011a, b)
Chemical precipitation	Metals and pesticides are removed by addition of coagulants such as lime, alum due to its availability and low cost	Large amount of sludge containing toxic compound is produced	Aziz et al. (2008), Fenglian and Wang (2011) and Singh et al. (2011a, b)
Ion exchange	The electrostatic force was responsible for the ion exchange of the diluted metal solution	High cost, partial removal of certain ions	Singh et al. (2011a, b)
Activated carbon adsorption	Remove disagreeable taste and color, chlorine, pores trapped microscopic particles and large organic molecules	Can generate carbon fines which are corrosive and abrasive	Sivakumar et al. (2012)
Nanofiltration	Nano-sized reactive agent removes organic contaminants	Rapidly clump with soil and limit the dispersal to their target	Dialynas and Diamadopoulos (2009)
Coagulation and flocculation	Coagulation destabilized colloidal particles by adding chemicals and neutralized negative charges and flocculation destabilized in floc and settle by gravity.	Large-volume sludge production, high chemical consumption	Fenglian and Wang (2011)
Flotation	Solids are removed by attaching air bubble, decreasing its density and float.	Increasing of ion strength flotation efficiency decreasing, higher cost	Ahmad et al. (2016)
Membrane filtration	Process commonly based on molecular size, charge, chemical nature, affinity, etc.; transmembrane pressure acts as a driving force for contaminants to transfer across the membrane	Limited flow rates process, complex operating system, low selectivity, higher maintenance cost	Qin et al. (2007)

4.3 Uptake Mechanisms of Contaminants by Plants

Macrophytes have specific and effective mechanisms for the removal of contaminants which vary with the plant type and whether the pollutant is organic or inorganic. Inorganic uptake is driven via membrane transporters, while the organic contaminants by diffusion. These absorbed contaminants then get detoxified by biochemical reactions using enzymatic mechanisms in the plant. Uptake of inorganic compounds is facilitated by active or passive mechanisms, whereas organic compounds are generally governed by hydrophobicity and polarity.

Table 4.4 Phytoremediation and its techniques

Techniques	Mechanisms of action	Medium	References
Rhizofiltration	Metals are taken up in aquatic plants through the roots	Water	Rawat (2012) and Rezania et al. (2016)
Phytoextraction	Uptake and concentration of metals via direct uptake into plant tissue with subsequent removal of plants	Soil/ water	Ali et al. (2013) and Thakur et al. (2016)
Phytostabilization	Root exudates cause metals to precipitate and become less bioavailability	Soil/ water	Wuana and Okieimen (2011)
Phytovolatilization	Plants evaporate volatile metals (Se, Hg) from the surface of leaves	Soil/ water	Sharma et al. (2015) and Thakur et al. (2016)
Phytotransformation	Plants uptake and degradation of organics with the help of enzymes	Soil/ water	Cacadore and Durate, (2015)

4.4 Uptake Mechanism of Metals

Metal accumulations in aquatic macrophytes have been reported in literature (Zhang et al. 2009; Rahman and Hasegawa 2011; Revathi and Venugopal 2013). In aqueous ecosystems heavy metals are directly or indirectly present as free ions, insoluble and soluble forms such as oxides, hydroxides, carbonates, chlorides, and humic substances. The roots of plants accumulate these metals through the plasma membrane to the cells, where detoxification and sequestration of metals take place at the cellular level. The heavy metals uptake/translocation mechanisms are likely to be closely regulated. Metals bind with peptides and proteins in plants and this results in enhanced accumulation. These peptides or proteins are preferentially metal specific such that metals with toxic effects, i.e., Cd, Hg, and Pb are also sequestered. Detoxification or sequestration process occurs after translocation in which a huge amount of heavy metals concentrate in organs without suffering any toxic effects (Ryu et al. 2003). Malate and citrate are excretion of plants which acts as metal chelators. As the pH decreases, the plants simultaneously increase the bioavailability of the metals by strong chelating agents (Ross 1994). Many scientists explained that cytoplasmic Ni is sequestered by histidine while vacuolar Ni is detoxified by binding with citrate (Kramer et al. 1996; Dhir et al. 2009). Zn forms more stable complexes with citrate and oxalate, while malate returns to the cytoplasm. Oxidative stress by heavy metals occurs after the formation of reactive oxygen species, such as superoxide ions, hydroxyl ions, and hydrogen peroxide. These ions are deactivated by enzymes, i.e., superoxide dismutase, ascorbate peroxidase, catalase, guaiacol peroxidase, and glutathione reductase, and nonenzymes, i.e., glutathione, phenolic compounds, and ascorbic acid (Parvaiz et al. 2008; Azqueta et al. 2009). In detoxification process heavy metals form complex with chelators and remove meta-

Table 4.5 Historical advances of phytoremediation using macrophytes

Decades	Summary	Scope of works	Highlight	References
1970s	Due to massive capacity of nutrient uptake from wastewater, aquatic macrophytes used for remediation	Basically research on remediation potential of aquatic plants specially submerged plants	Potential of uptake	Boyd (1970) and Cowgill (1974)
	Submerged and floating quickly uptake pollutant from water			
	Levels of potentially toxic elements in the plants were at least an order of amount higher than in the supporting aqueous medium			
1980s	Floating and emergent plants can uptake contaminants through roots while submerged plants by root and leaves both	Study on emergent and floating plants capability	Species determination	Denny (1980, 1987)
1990s	Rates of toxic metal uptake and removal of plants are greater than 1000 μg g^{-1} called as hyperaccumulator	Research on metal accumulation through root and foliar parts of aquatic plants	Importance of various mechanisms	Outridge and Noller (1991) and Sharma and Gaur (1995)
00–10	Aquatic plants can efficiently remove heavy metals which is the largest category of pollutants	Monitoring the effective role of macrophytes and developing of hyperaccumulator plants	Effectiveness of species	Hu et al. (2003), Kamal et al. (2004) and Rai (2009)
	Aquatic macrophytes also used as nonliving, for removal and monitoring of heavy metals			
10–18	Uptake mechanisms of green plants which can accumulate pollutants with high ability	Focus on mechanism and improvement of techniques efficiency	Optimization toward implication	Sharma et al. (2012), Ali et al. (2013), Sasmaz et al. (2008) and Kumar et al. (2018a)
	Effectiveness 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			

Table 4.6 Macrophytes and their phytoremediation potential for various heavy metals

Plant species	Metals	Accumulation	References
Eichhornia crassipes	Cu	6000–7000 mg kg^{-1}	Molisani et al. (2006)
	Cr	4000–6000 mg kg^{-1}	Hu et al. (2007)
	Ni	1200 mg kg^{-1}	Low et al. (1994)
	Cd	2200 µg kg^{-1}	Zhu et al. (1999)
	Zn	1677 mg g^{-1}	Kamel (2013)
	As	909.58 mg kg^{-1}	Delgado et al. (1993)
	Hg	119 ng g^{-1}	Molisani et al. (2006)
	Mn	300 mg kg^{-1}	Dixit et al. (2011)
Azolla pinnata	Ni	16,252 l µg g^{-1}	Arora et al. (2004)
	Cd	740 µg g^{-1}	Rai (2008)
	Cr	1095 µg g^{-1}	Rai (2010)
	Hg	940 µg g^{-1}	Rai and Tripathi (2009)
	Pb	1383 mg kg^{-1}	Thayaparan et al. (2013)
Azolla filiculoides	Cd	2608 µg g^{-1}	Arora et al. (2004)
	Cu	6013 µg g^{-1}	Zhang et al. (2008)
			Arora et al. (2006)
	As	>60 µg g^{-1}	Vesely et al. 2011
	Cr	12,383 µg g^{-1}	
	Pb	1607 mg kg^{-1}	
Azolla caroliniana	Cr	356 mg kg^{-1}	Bennicelli et al. (2004) and Zhang et al. (2008)
	Hg	578 mg kg^{-1}	
	As	284 mg kg^{-1}	
Hydrilla verticillata	As	231 mg kg$_{-1}$	Srivastava et al. (2010)
	Cu	770–30,830 mg kg^{-1}	Srivastava et al. (2011)
Typha latifolia	Ni	295.6 mg kg^{-1}	Afrous et al. (2011)
	Cu	1156.7 mg kg^{-1}	Nguyen et al. (2009)
Pistia stratiotes	Pb	519 mg kg^{-1}	Vesely et al. (2011)
Salvinia minima	Pb	5469 mg kg^{-1}	Vesely et al. (2011)
Salvinia natans	Cr	7.40 mg g^{-1}	Dhir (2009)
Lemna gibba	U	896.9 mg kg^{-1}	Mkandawire et al. (2004)
	As	1021.7 mg kg^{-1}	
Lemna minor	Pb	561 mg g^{-1}	Leblebici and Aksoy (2011) and Bokhari et al. (2016)
	Cu	34.6 µg g^{-1}	
Typha angustifolia	Mn	860 mg kg^{-1}	Sasmaz et al. (2008)
	Cu	50 mg kg^{-1}	
	Zn	56.47 µg g^{-1}	
Myriophyllum spicatum	Pb	8.94 mg g^{-1}	Kamel (2013)
	Zn	2.66 mg g^{-1}	
Ceratophyllum submersum	Pb	258.62 mg kg^{-1}	Kamel (2013) and Guo et al. (2014)
	Zn	1172.8 mg kg^{-1}	

(continued)

Table 4.6 (continued)

Plant species	Metals	Accumulation	References
Wolffia globosa	As	>1000 mg kg^{-1}	Zhang et al. (2009)
Phragmites communis	Fe	2813 μg g^{-1}	Chandra and Yadav (2011)
	Mn	814.40 μg g^{-1}	
	Zn	265.80 μg g^{-1}	
	Pb	92.80 μg g^{-1}	
Phragmites australis	Cu	16.55 μg g^{-1}	Salman et al. (2015)
	Pb	0.77 μg g^{-1}	
	Cd	33.115 μg g^{-1}	
Potamogeton pectinatus	Cd	964.75 μg g^{-1}	Salman et al. (2015)
	Cu	28.75 μg g^{-1}	

bolically active cytoplasm ions by moving them into vacuole and cell wall. In vacuoles hazardous metal ions are captured in limited sites. Therefore, other parts of the cell do not have access to these dangerous metal ions. Cd detoxification by inducing the synthesis of phytochelatins (PCs) forms a Cd-PC molecule, which is further transferred into the vacuoles by Cd/H antiport and ATP-dependent phytochelatin-transporter (Revathi and Venugopal 2013). MTP, a gene encoding a protein localized at tonoplast (separating vacuole from cell wall), is exceedingly expressed in plants of Zn/Ni hyperaccumulating plants (Dräger et al. 2004; Kim et al. 2004; Hammond et al. 2006; Gustin et al. 2009). It has been suggested that MTP play a major role in Zn tolerance and accumulation. Persant et al. (2001) explained that MTP also mediate the Ni vacuolar storage in *Thlaspi goesingense* shoots.

4.5 Uptake Mechanisms of Pesticides

Aquatic plants have capacity to uptake and accumulate organochlorine, organophosphorus, carbamate, and pyrethroid pesticides from water (Gobas et al. 1991; Rice et al. 1997; Macek et al. 2000). These pesticides pass through membrane between root symplast and xylem apoplast by diffusion and their entry depends on passive movement over membranes for their uptake into the aquatic plants (Nwoko 2010). No specific transporters are found in plants for these man-made compounds, so the speed of movement of pesticides in the plant depends to a large extent on their physicochemical properties. Three sequential phases are involved in metabolization of pesticides.

In the first phase, pesticides undergo hydrolysis, reduction, and oxidation (Eapen et al. 2007; Komives and Gullner 2005). Functional groups present in pesticides convert these into more polar, chemically active, and water-soluble compounds (Komives and Gullner 2005). In plants, oxidative metabolism is primarily mediated

Table 4.7 Macrophytes and their phytoremediation potential for various pesticides

Plant species	Contaminants	Accumulation	References
Hydrilla verticillata	Chlordane	1060.95 μg L^{-1}	Chaudhry et al. (2002)
Typha latifolia	Dieldrin	0.60 ng g^{-1}	Guo et al. (2014)
Pistia stratiotes	Chlorpyrifos	0.036 mg g^{-1}	Prasertsup and Ariyakanon (2011)
Lemna minor	Flazasulfuron,	27 μg g^{-1}	Olette et al. (2009)
	Dimethomorph	33 μg g^{-1}	
	Chlorpyrifos	0.23 g^{-1}	
Ceratophyllum submersum	Aldrin	0.38 ng g^{-1}	Guo et al. (2014)
	Endosulfan	0.73 ng g^{-1}	
Phragmites communis	α-HCH	0.89 ng g^{-1}	Guo et al. (2014)
	β-HCH	1.18 ng g^{-1}	
	γ-HCH	0.97 ng g^{-1}	
	DDTs	0.93 ng g^{-1}	
Schoenoplectus californicus	DDTs	30.2–45.7 ng g^{-1}	Miglioranza et al. (2004)
	HCHs	0.61 ng g^{-1}	
	Chlordane	4.04 ng g^{-1}	
Spirodela oligorrhiza	o,p′-DDT, p,p′-DDT	50–66%	Gao et al. (2000)
Plantago major	Cyanophos	76.91 μg g^{-1}	Romeh (2014)
Iris pseudacorus	Chlorpyrifos	1.88 μg g^{-1}	Wang et al. (2013)
	Triazophos	42.11 μg g^{-1}	Li et al. (2014)
Nymphaea amazonum	γ-Cyhalothrin	2.02 μg g^{-1}	Mahabali and Spanoghe (2014)
Eleocharis mutala	Imidacloprid	13.51 μg g^{-1}	Mahabali and Spanoghe (2014)
Cyperus rotundus	Triazophos	24.63 μg g^{-1}	Li et al. (2014)
Acorus calamus	Chlorpyrifos	15.3 μg g^{-1}	Wang et al. (2016)
Juncus effusus	Tebuconazole	720 μg g^{-1}	Lv et al. (2016)

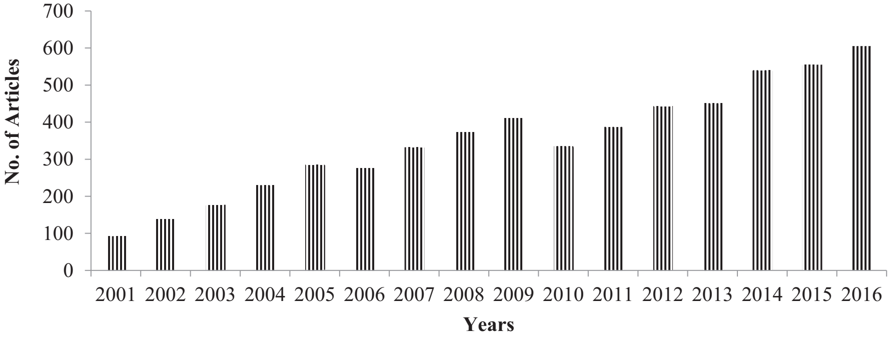

Fig. 4.1 Publications in the field of phytoremediation from the last 16 years (www.sciencedirect.com)

by cytochrome P_{450} monooxygenase (Sandermann 1994, Doty et al. 2007). These enzymes are very crucial during oxidative bioactivation process to emulsify the highly hydrophobic contaminants and convert them into chemically reactive electrophiles forming conjugates (Morant et al. 2003). In the second phase, conjugation takes place in the cytosol where pesticide gets conjugated with sugar, amino acids, and –SH group of glutathione and converts into hydrophilic forms. Conjugated compounds have a high molecular weight and are more polar and less toxic as compared to the parent compound. Transformation hydroxylation of organochlorine pesticides, i.e., 2,4-D, is followed by conjugation with glucose and malonyl and deposition in vacuoles. Every enzyme that participates in detoxification process has specific functions. Phosphatases that cleave phosphate groups from organophosphate are studied in *Spirodela polyrhiza*, and dehalogenases that cleave halogen group from organochlorine pesticides are noted in *Myriophyllum aquaticum* (Dhir 2009; Susarla et al. 2002). After this sequestration takes place. Capture of the pesticides such as organochlorine and organophoshate by plants includes physical (adsorption, absorption, partition) and chemical processes (complex formation), and reaction with cuticular membrane components helps in the sequestration of lipophilic organic compounds. Once man-made chemicals are taken up by plant, it can be transformed via metabolization, volatilization, lignification, and mineralization to carbon dioxide, water, and chlorides. Detoxification transforms the main chemical to non-phytotoxic metabolites, including lignin, that are stored in plant cells (Coleman et al. 1997; Dietz and Schnoor 2001). Then these metabolites are transported to the vacuoles by tonoplast membrane-bound transporters. Vacuolar compartmentalization is a major stage in detoxification of pesticides (Coleman et al. 2002).

4.6 Influencing Factors in Phytoremediation

There are several factors which can affect the uptake mechanisms of heavy metals and understanding about these factors can improve the metal removal capacity of plant. These factors are divided into two categories, biotic and abiotic, and are discussed below.

4.6.1 Biotic Factors

4.6.1.1 Plant Species

Phytoremediation techniques depend upon the suitable species that can accumulate heavy metals and produce higher biomass using established crop production and management practices (Rodriguez et al. 2005).

4.6.1.2 Plants Organs

Roots are important organs of the plants; they can absorb contaminants and bind to the cell wall or other macromolecules to prevent them from moving to other sensitive organs of the plant (Merkl et al. 2005). Zn and Cd get accumulated in the roots and the stem, while the accumulation of Cu was more in the leaves because the capacity of the roots gets exhausted due to the higher concentration of Cu in the wastewater (Rezania et al. 2015).

4.6.2 Abiotic Factors

4.6.2.1 pH

It is a very important abiotic factor controlling metal availability to the plant (Chen et al. 2015). Sanyahumbi et al. (1998) reported that Pb 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 on heavy metal uptake was investigated through *Potamogeton natans* and *Elodea canadensis*, and it was reported that metal removal efficiency increased with decreasing salinity and increasing temperature (Fritioff et al. 2005).

4.6.3 Chelating Agents

Chelating agents are commonly used to increase the bioavailability of heavy metals, accordingly enhancing their uptake by plants (Tangahu et al. 2011). Ethylenediaminetetraacetic acid (EDTA), a strong chelating agent and having strong complex formation capacity, has been widely used (Yen and Pan 2012). Phosphonates and phosphonic acids are also used as chelating agents in many applications, e.g., in paper, pulp, and textile industries and for heavy metals in chlorine-free bleaching solutions that could inactivate the peroxide (Gledhill and Feijtel 1992).

4.6.4 Other Environmental Factors

Climate is an important limiting factor for efficiency of phytoremediation at a particular site. Temperature is a key factor, affecting transpiration and growth metabolism, and ultimately leads to disruption of the plant's metal uptake capacity (Burken and Schnoor 1996; Bhargava et al. 2012). Removal efficiency of plants increases linearly with increasing temperature (Yu et al. 2011). The temperature affects the growth and consequently the length of the roots. The structure of the root under field conditions differs from that under greenhouse conditions (Merkl et al. 2005). Understanding mass balance analyses and the metabolic fate of contaminants in plants are the keys to maintain the applicability of phytoremediation (Mwegoha 2008). Metal uptake by plants depends on the bioavailability of the metal in the water, which in turn depends on the retention of the metal, as well as the interaction with other elements and substances in the water as well as on the prevailing climatic conditions (Tangahu et al. 2011).

4.7 Potential of Some Aquatic Macrophytes for Removal of Heavy Metals and Pesticides from Water

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 can be 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 that grow below the surface of water and include a few ferns, numerous mosses, and some angiosperms (e.g., *Hydrilla verticillata*, *Ceratophyllum demersum*, *C. submersum Myriophyllum aquaticum*, *Elodea canadensis*, *Vallisneria americana*, *Utricularia vulgaris*, *Najas graminea*).
3. Free-floating macrophytes: Plants that are non-rooted to the substratum and float 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-leaf macrophytes: Plants that are submerged or in sediment but with leaves that float with long flexible petiole on the surface (mainly include angiosperms, e.g., *Nymphea alba*, *Potamogeton crispus*, *P. natans*, *P. pectinatus*, *Nelumbo nucifera*, *Hydroryza aristata*). Boyd (1970), Stewart (1970), Wooten and Dodd (1976), and Conwell et al. (1977) were pioneers to demonstrate the pollutant removal potential of aquatic macrophytes. They considered these plants as important components of the aquatic systems in not only being food source, but because of the ability to act as effectual accumulators of heavy metals (Devlin 1967; Rai 2009; Deval et al. 2012; Sood et al. 2012).

Many scientists compared the efficiency of aquatic macrophytes for phytoremediation. Aquatic macrophytes absorb nutrients through their effective root systems. They are extensively used to remove nutrients, heavy metals, and pesticides from wastewater due to their relative fast growth rate and accumulation ability. Phytoremediation is an economic method with minimum maintenance and also helps in improving biodiversity. Several studies have revealed that aquatic plants are very effective in removing heavy metals and pesticides from polluted water (Khan et al. 2009; Yasar et al. 2013; Akter et al. 2014; Sasmaz et al. 2015). Discussed below are some important macrophytes which are potentially important for phytoremediation purposes.

4.7.1 Eichhornia crassipes

E. crassipes, commonly known as water hyacinth, is a rapidly growing aquatic macrophyte which can double its biomass in a few days and is one of the world's most troublesome weed. This quality has also made it an applicant for use in phytoremediation (Dhote and Dixit 2009). Many scientists proved that water hyacinth has high removal rates for various heavy metals like Fe, Zn, Cu, Cr, Mn, Hg, Cd, and As from aqueous solutions (Jadia and Fulekar 2009; Mohamad and Latif 2010; Priya and Selvan 2014; Rezania et al. 2015). The water hyacinths store metals in their bladders, followed by their translocation to stems, leaves, and roots (Rizwana et al. 2014). Mokhtar et al. (2011) used *E. crassipes* for the removal of Cd and Zn from water, as well measured the concentration of Cd and Zn absorbed in different parts of water hyacinth (leaves, roots, stem, and flowers). Ajayi and Ogunbayo (2012) studied the efficiency of *E. crassipes* in removing Cd, Cu, and Fe from water and found that transfer efficiency of Cd is more as compared to Cu and Fe. It was also investigated that this emergent plant is effective in removing mevinphos (insecticides) and ethion (phosphorus pesticides) from polluted water (Ramchandran et al. 1971; Wolverton 1975; Xia and Ma 2006).

4.7.2 Lemna

Lemna, commonly known as duckweed, is a free-floating macrophyte on the water surface. It is fast growing and adapts easily to various aquatic conditions and globally distributed in lakes, ponds, wetlands, and some effluent lagoons. It has been used to recover heavy metals since more than 30 years. Most of studies have been conducted with species *L. genus*, *L. minor*, and *L. gibba* (Guimaraes et al. 2012). 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. *L. minor* can remove up to 90% of soluble Pb from water (Singh et al. 2011a, b). Sasmaz and Obek (2009) reported that the aquatic plant *L. gibba* was used for the accumulation of As, B, and U from secondary effluents as an alternative method for treatment. The results demonstrate that As was quickly absorbed by *L. gibba* in the first 3 days of the experimental study. Other studies on duckweed showed that an excess of Cu interferes in respiration, photosynthesis, pigment synthesis, and enzyme activity of the plants (Teisseire and Guy 2000; Prasad et al. 2001; Frankart et al. 2002; Babu et al. 2003). Olette et al. (2009) have found that *L. minor* can effectively accumulate pesticides, viz., copper sulfate (fungicide), flazasulfuron (herbicide), and dimethomorph (fungicide), from water bodies.

4.7.3 Typha

Typha is an ordinary wetland plant that belongs to family Typhaceae and grows widely in tropic and warm regions. Most of the studies have been done with the species *T. latifolia*, *T. angustifolia*, *T. domingensis*, and *T. angustata. T. latifolia* has a high capacity to transport heavy metals to its tissue. Therefore, it also tolerates higher levels of metals in its tissue without serious physiological damage. Dunbabin and Bowmer (2009) reported that metal concentrations increased in the order of roots > rhizomes > nongreen leaf > green leaf and found that the accumulation was highest in the roots and the green leaves had the lowest concentrations of Cu, Zn, Pb, and Cd. Chandra and Yadav (2010) also checked *T. angustifolia* for remediation potential of various heavy metals (Cu, Pb, Ni, Fe, Mn, and Zn) and resolved that it could be a possible phytoremediator for heavy metals from industrial wastewater under optimized conditions. Miglioranza et al. (2004) observed significant differences in the DDT level between root and shoot of *Typha* tissues, indicating the capability of the plant to uptake pesticide.

4.7.4 Azolla

Azolla is a small aquatic fern belonging to family Azollaceae with monotypic genus (Sood and Ahluwalia 2009). Azolla occurs in the symbiotic association with N_2 fixing blue, green alga *Anabaena azollae* (Mashkani and Ghazvini 2009; Sood et al. 2011). This fern can hyperaccumulate a variety of pollutants such as heavy metals and pesticides from aquatic ecosystems (Padmesh et al. 2006; Mashkani and Ghazvini 2009; Rai and Tripathi 2009; Sood et al. 2011). This fern has several features which prove it to be a better plant for phytoremediation, which include fast growth rate, nitrogen-fixing ability, and easy biomass disposal. Both living and dead biomass have been used for the removal of heavy metals (Rai 2008; Mashkani and Ghazvini 2009). Three species of water fern (*A. caroliniana*, *A. filiculoides* and *A. pinnata*) have been studied for heavy metal uptake from water. Rai (2008) reported that *A. pinnata* removed up to 70–94% of heavy metals (Hg and Cd) from chlor-alkali and ash slurry effluent in Singrauli region of UP (India). Deval et al.

(2012) concluded that *A. caroliniana* showed maximum efficiency toward the accumulation of Zn. Photosynthesis pigment of *Azolla* was also observed to increase under the influence of Zn and other contents of the effluents.

4.7.5 Hydrilla verticillata

H. verticillata is a submerged aquatic macrophyte that can grow on the surface and forms dense mats in water bodies. For removal of inorganic and organic contaminants, the whole plant plays an important role. Scientists explained that *H. verticillata* has strong appetite for As and Cd, but its appetite for Pb is not so strong (Ghosh 2010; Singh et al. 2011a, b, 2012, 2013). Dixit and Dhote (2010) studied Cr, Pb, and Zn uptake along with morphological changes in *H. verticillata* which indicate that uptake of metals is dose dependent.

4.7.6 Salvinia

Salvinia is a free-floating aquatic macrophyte of Salviniaceae family. It is widely distributed, having a fast growth rate and close relation with *Azolla* and *Lemna*. Genus *Salvinia* represents several species, i.e., *S. herzogii*, *S. minima*, *S. natans*, and *S. rotundifolia*, which show potential to remove various contaminants including metals from wastewaters (Nichols et al. 2000; Olguin et al. 2005; Sune et al. 2007; Sanchez-Galvan et al. 2008; Xu et al. 2009). *S. minima* is able to remove Ni, Cu, and As from water (Mukherjee and Kumar 2005; Rahman et al. 2009). Fuentes et al. (2014) indicated that *S. minima* are a hyperaccumulator of Ni, although higher concentrations may affect the physiological performance of the plant. Espinoza-Quiñones et al. (2008) demonstrated that *Salvinia auriculata* can be used as biosorbent for heavy metal removal from industrial effluents in wetlands.

4.7.7 Pistia

Pistia, commonly called as water lettuce, is a genus of aquatic macrophytes in the family Araceae. It floats on the surface of the water and roots are hanging beneath floating leaves. They are natural hyperaccumulators of many toxic heavy metals. Odjegba and Fasidi (2004) reported that *Pistia* is a potential candidate for the removal of Zn, Cr, Cu, Cd, Pb, and Hg. It accumulates Zn and Cd at high concentrations, whereas Hg is moderately accumulated and is poor in Ni accumulation (Guimaraes et al. 2012). Miretzky et al. (2004) mentioned that the percentage of removal by *P. stratiotes* was very high (>85% for Pb, Cr, Mn, and Zn). They also explained that it can almost completely eliminate the metals in the first 24 h of exposure. Prasertsup and Ariyakanon (2011) investigated potential of *P. stratiotes* for removal of chlorpyrifos (organophosphate pesticide) under greenhouse conditions and found it to remove the pesticide by 82% from water. Recently, Kumar et al. (2019) also reported the efficient removal of Cu^{2+}, Fe^{3+}, and Hg^{2+} from aqueous solutions by *P. stratiotes*.

4.7.8 Ipomoea aquatica

I. aquatica belongs to the family Convolvulaceae, originated in China, and is usually consumed as a green leafy vegetable. It is mostly found in southern Asia, India, and southern China. Chen et al. (2009) investigated that *I. aquatica* can remove Cr (III) from aqueous solution in the presence of chelating agent EDTA and chloride. Chloride can increase the solubility of Cr and enhance the bioaccumulation in shoots and roots of the plant. Gothberg et al. (2002) estimated the accumulation of Pb, Cd, Hg, and methyl mercury in *I. aquatica*. However, concentrations of Hg were higher in leaves than in stems. Chi et al. (2008) observed that accumulation of di-*n*-butyl phthalate (phthalic acid esters) in five different genotypes of *I. aquatica* with their potential of phytoremediation.

4.7.9 Myriophyllum

Myriophyllum is a submerged perennial macrophyte, found in stagnant and slow-moving waters in the southern hemisphere. Several studies on heavy metal biosorption ability of species *M. spicatum*, *M. triphyllum*, and *M. aquaticum* have been done. This is applied for biomonitoring and water purification by accumulating heavy metals in their tissues (Ngayila et al. 2007). Accumulating capacity of this plant is higher due to rhizomatous stem that are able to capture pollutants from water (Orchard 1981). Grudnik and Germ (2010) used it as indicator for pollution by metals in lake and reported the concentrations of metals in *M. aquaticum* were higher than other plants indicating the concentrations of the metal pollutants in the lake. Harguinteguy et al. (2016) showed positive correlation between Co, Cu, Mn, and Zn concentration in water and leaves of *M. aquaticum.*

4.7.10 Phragmites australis

P. australis is an emergent aquatic macrophyte commonly called as reed. They are grown under extreme environmental conditions in presence of nutrients and organic carbon (Quan et al. 2007; Bonanno and Giudice 2010). The root of this plant accumulates higher quantity of heavy metals in the cortex parenchyma cells with large intracellular air space (Sawidis et al. 1995). Bonanno and Guidice (2010) studied the heavy metal accumulation in *P. australis* organs and also evaluated its suitability for biomonitoring. Concentration of heavy metal in aboveground parts depends largely in growing season; particularly accumulation may increase simply at the end of the growing season (Brogato et al. 2009). Highest metal accumulation was recorded in roots and shoots in September and April, whereas leaves expressed higher value in February (Salman et al. 2015). Bananno and Guidice (2010) explained that the root of *P. australis* acts as a filter for Cu because it accumulates 70% (in roots). So, this filter effect is the most effective strategy for protection of shoots and roots from Cu-induced injuries. According to the recent studies, *P. australis* has many benefits, such as good growth, worldwide distribution, and high levels of heavy metal tolerance (Salman et al. 2015).

4.7.11 Ceratophyllum demersum

C. demersum is a submerged aquatic macrophyte which can grow in low light and muddy water, may be oligotrophic or eutrophic. Various studies of the phytoremediation have shown that *C. demersum* is effective for accumulation of heavy metals and pesticides (Krems et al. 2013; Guo et al. 2014; Chen et al. 2015). This plant has positive adaptive strategy in response to heavy metals and pesticides in in situ studies (Borisova et al. 2014). Rai et al. (1995) reported that *C. demersum* was able to remove >70% Pb from pond water in 15 days. Abdallah (2012) explained that chlorophyll is an important factor which is sensitive to heavy metal concentration. A decrease of chlorophyll proves the toxic nature of Cd, which interacts with –SH group of enzymes involved in chlorophyll synthesis. According to Saygidegs and Dogan (2004), *C. demersum* accumulated more Pb than *L. minor* and chelating agent EDTA has the ability to increase bioavailability of Pb to increase accumulation in plants. Guo et al. (2014) reported that organochlorine pesticides hexachlorocyclohexae (HCH), DDT, aldrin, dieldrin, endosulfan, etc. are accumulated in *C. demersum* tissues.

4.8 Management, Treatment, and Disposal of Phytoremediating Aquatic Macrophytes

It has been validated by various scientists that phytoremediation is a cost-effective and eco-friendly technology for rehabilitation of polluted environments as compared to conventional methods, but it has its own drawbacks (Rahman et al. 2009; Sood et al. 2012; Emmanuel et al. 2014; Sharma et al. 2015). For example, plant growth and biomass production are good, but seasonality and poor tolerance are constraints of the technology, and affective process should involve regular harvesting and disposal of macrophytes since they will decompose and release heavy metals back to the environment (Rai 2008). Only accumulation of metals in macrophytes is not enough implementation of this emerging technique. The proper disposal of these macrophytes after phytoremediation is very essential; otherwise, these macrophytes will act as another source of pollutants in the environment. There are several processes by which phytoremediating plants can be converted into economically beneficial material.

4.8.1 Biogas Production

Biogas is a clean and environmentally friendly fuel formed by the anaerobic digestion of organic wastes, i.e., animal dung, vegetable wastes, municipal solid wastes, and industrial wastes (Weiland 2010). Anaerobic digestion is a biological process in which organic matter is degraded in the absence of oxygen. The biogas generated can be used directly for various purposes, i.e., cooking, heating, or production of electricity. There is a comprehensive literature significantly describing the use of aquatic plants used as a potential store for biogas production due to high quantity, high carbon-nitrogen ratio, and good content of fermentable materials. *Eichhornia*, *Pistia*, *Typha*, and *Trapa* can be degraded easily and produce high biogas yields (Elhaak et al. 2015). Singhal and Rai (2003) showed the use of *E. crassipes* and *Panicum hemitomon*, for phytoremediation of industrial effluents and subsequent production of biogas.

4.8.2 Ethanol Production

Ethanol is a liquid fuel which can be produced from phytoremediating aquatic macrophytes through hydrolysis and fermentation which can make them a good substrate as well. Hydrolysis and fermentation require fermentable sugars, which may be available in very small amounts in aquatic plants, so pretreatment is necessary for making sugar more easily available for chemical hydrolysis (Gunnarsson and Petersen 2007). Scientists generally follow three steps for production of ethanol from aquatic plants. In the first step, the cellulase enzyme was produced by the isolation and qualitative screening of microorganisms in the excreta of cow, pork, goat and municipal waste. However, this enzyme has also been produced by the addition of dry aquatic plants and micro-organisms. In the last step, ethanol is produced through fermentation process by hydrolysis of cellulose present in aquatic plant by the fermenting organism (Randive et al. 2015; Patel and Patel 2015). Rezania et al. (2015) studied the use of barley and malt extract enhancer for ethanol production from *E. crassipes* and *P. stratiotes* and found that use of these substrates increases the production.

4.8.3 Incineration

Incineration is the combustion of waste material in the presence of oxygen. In this process the phytoremediating aquatic plants may be used for making charcoal and the by-products can be used as a fuel (Rahman and Hasegawa 2011). Sun drying and direct burning product of water hyacinth are used as fertilizer on a small scale in certain parts of the world (Gunnarsson and Petersen 2007).

4.8.4 Composting and Vermicomposting

Compost improves soil nutrient and structure; hence, it can be an option for management of harvested macrophytes in developing countries, where chemical fertilizers are not affordable. Macrophytes contain nutrients like nitrogen, phosphorus, and potassium, and converting them into compost takes less than 30 days (Newete and Byrne 2016). This makes it feasible for farmers for improving soil condition by swiftly utilizing the waste converted to compost. Hussain et al. (2016) formed vermicompost by using *Salvinia* and *Eisenia fetida* and concluded that it is an effective technique to convert *Salvinia* into value-added product.

4.8.5 Other Uses

Many macrophytes such as *Eichhornia*, *Typha*, and *Cyperus* have been directly collected from experimental sites and used for making mats, hats, bags, baskets, and spoon holders in weaving industries. Stem of *Scirpus grossus* is used in manufacturing of hard rope and fine mats. These plants also reduce wave action impacts and hold the bottom sediments more efficiently which helps to reduce turbidity and suspension of nutrients bound in the sediment.

4.9 Conclusion

Since contamination of water by toxic heavy metals and pesticides is a serious environmental problem, therefore, effective remediation methods are necessary. Conventional methods for clean-up and restoration of heavy metals and pesticides from contaminated water have limitations like high cost and creation of secondary pollutants. Phytoremediation is a promising technology that can become a reliable, efficient alternative for remediation of contaminated water. Plants can take up heavy metals and pesticides by their roots, stems, and leaves and accumulate them in organs. The knowledge of several factors which affect the uptake mechanisms of heavy metals, like plant species, addition of chelating agents, and physical and climatic conditions can help in improving the efficiency of the process. It is now proven that many aquatic plants such as *Eichhornia*, *Pistia*, *Lemna*, *Salvinia*, *Typha*, and *Hydrilla* are capable of accumulating heavy metals and pesticides. The roots of these plants naturally absorbed heavy metals from water. Accumulation and

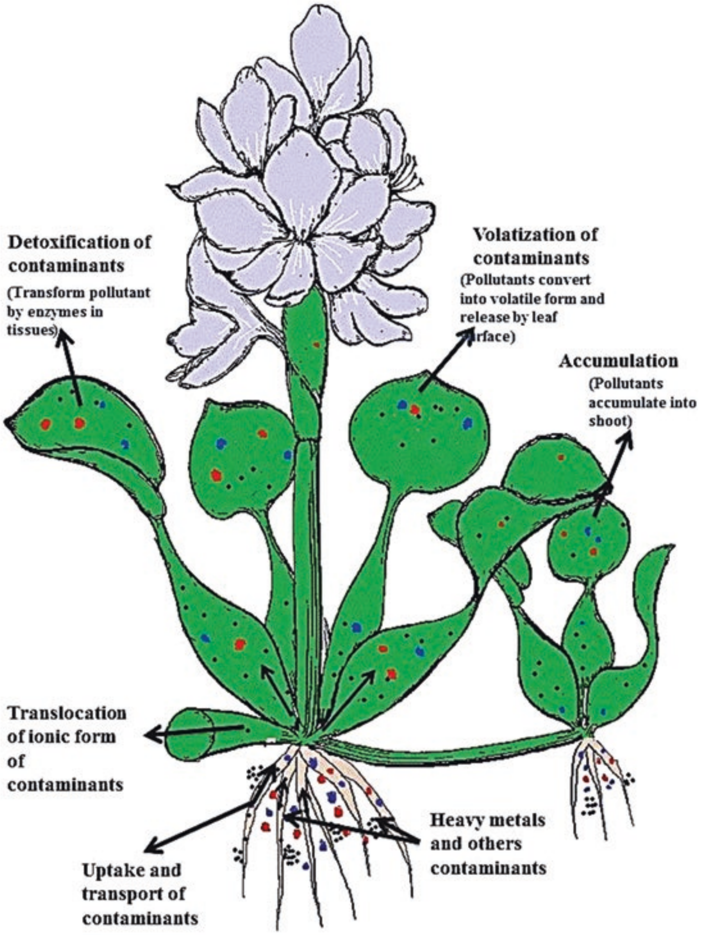

Fig. 4.2 Phytoremediation mechanism and its techniques in plant tissues

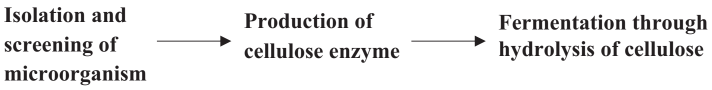

Fig. 4.3 Flow diagram for the three steps for production of ethanol from aquatic plants

remediation of heavy metals and pesticides are not enough for implantation of phytoremediation. Management, and treatment of the end product, i.e., the biomass is also a major concern. Some studies have now shown that there is possibility to use macrophytes' biomass for production of biogas, bioethanol, etc. This can pave the way for effective utilization of this technology for cleaning the contaminated sites by an eco-friendly and effective approach (Figs. 4.2 and 4.3).

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