Role of Phytoremediation as a Promising Technology to Combat Environmental Pollution

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Mehmooda Munazir, Rahmatullah Qureshi, Mubashrah Munir, and Hamid Mukhtar

Abstract

A wide variety of pollutants such as heavy metals, organic, and inorganic wastes are continuously being added to the environmental components globally. These pollutants are stressing our environment and badly eroding the biotic components of our ecosystems. Besides this, these are hazardous to human health. Phytoremediation is a promising environment friendly technology that has gained attention of researchers across the globe from the past few decades. Phytoremediation (also known as "green remediation" and "botanical bioremediation") utilizes plants to reduce, remove, degrade, or immobilize environmental toxins, primarily those of anthropogenic origin aiming at restoring polluted sites to a condition useable for private or public applications. Some of the heavy metals and pollutants such as lead, chromium, cadmium, copper, nickel, mercury, zinc, strontium, boron, selenium, arsenic, thallium, uranium, calcium, cobalt, manganese, nitrates, herbicides, and chlorinated compounds are highly toxic and lethal even in trace amounts which may be teratogenic, mutagenic, endocrine disruptive as well as behavioral and neurotoxic in nature. With ever-increasing urbanization

M. Munazir
Department of Botany, Government College Women University, Sialkot, Punjab, Pakistan

R. Qureshi (✉)
Department of Botany, Pir Mehr Ali Shah Arid Agriculture University, Rawalpindi, Punjab, Pakistan
e-mail: rahmatullahq@uaar.edu.pk

M. Munir
Department of Biological Sciences, University of Veterinary and Animal Sciences, Lahore, Punjab, Pakistan

H. Mukhtar
Institute of Industrial Biotechnology, Government College University (GCU), Lahore, Pakistan

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and advancement in technology, the addition of pollutants is expected to continue by many folds. Phytoremediation has been found effective in remedying the high concentration of these pollutants from the soil and groundwater. Some plant species have interestingly been found effective in absorbing radioactive and toxic elements from air as well. The concept of phytoremediation was well-known, and various plants are being used by the Neanderthal man for wastewater treatment from thousand years ago. Some of the species such as *Avena sativa, Brassica juncea, B. napus, Hordeum vulgare, Panicum virgatum, Thlaspi caerulescens,* and *Viola calaminaria* have successfully been used to absorb environmental pollutants. From the past decade, several methods of phytoremediation like phytoextraction/phytoaccumulation, phytotransformation, phytostabilization, phytostimulation, phytorhizodegradation, phytodegradation, and phytovolatilization have been under investigation. Besides, the role of different factors that affect phytoremediation such as EDTA, CDTA, DTPA, EDDS, NTA, HEDTA, EGTA, and citric acid have also been studied by various researchers globally. This chapter is an endeavor to provide a comprehensive overview on all aforementioned aspects of phytoremediation along with future prospects of this technology. In addition, limitations and advantages of the said technique are also discussed in detail that would help the readers to find answers to various questions pertaining to this potential technique.

Keywords

Phytoremediation · Phytotransformation · Phytostabilization · Phytostimulation · Chelating agents

16.1 Introduction

Phytoremediation is a promising environment-friendly technology that has gained attention of researchers across the globe from the past few decades. This is plant-based technology used either naturally or genetically engineered plants for cleaning up the polluted environments (Cunningham et al. 1997; Flathman and Lanza 1998; Sarma et al. 2021; Sonowal et al. 2022). This is supposed to be a low-cost technology that utilizes plants to reduce, remove, degrade, or immobilize environmental toxins, primarily those of anthropogenic origin for restoring polluted sites to a condition useable for private or public applications (Ensley 2000). Though the term, phytoremediation is a quite new discovery; however, it is practiced since ages (Cunningham et al. 1997; Brooks 1998). The use of semiaquatic plants for recycling the radionuclide-polluted water was found in practice in Russia at the initiating time of nuclear period (Timofeev-Resovsky et al. 1962). A number of plants have capability to accumulate significant amount of metals in their tissues while growing on metal deposited soils without showing toxicity (Baker et al. 1991; Entry et al. 1999). The effectiveness of phytoremediation depends on the type of pollutant, bioavailability, and soil properties (Cunningham and Ow 1996). Some of the heavy metals and pollutants such as lead, chromium, cadmium, copper, nickel, mercury, zinc, strontium, boron, selenium, arsenic, thallium, uranium, calcium, cobalt, manganese, nitrates, herbicides, and chlorinated compounds are highly toxic and lethal even in trace amounts which may be teratogenic, mutagenic, endocrine disruptive as well as behavioral and neurotoxic in nature (Duffus 2002). With ever-increasing urbanization and advancement in technology, the addition of pollutants is expected to continue by many folds. Phytoremediation has been found effective in remedying the high concentration of these pollutants from the soil and groundwater (Lone et al. 2008).

16.2 Environmental Pollution and the Need of Remediation

Healthy, prosperous, and successful life on earth is dependent on healthy environment. But the quality of environment has been deteriorated by means of environmental pollution. The environmental pollution can be defined as "addition of unwanted and undesirable elements to the biotic and abiotic components of an environment by means of anthropogenic activities which ultimately decrease the quality of life." The scarcity of drinking water and loss of soil fertility are the initial results of pollution. The situation becomes worse when it enters at the food chain level. It is pathetic to note that drinking water is not healthy in most of the parts of the world owing to the contamination by various environmental pollutants (Daud et al. 2017).

The environment is comprised of two types of components, i.e., abiotic and biotic. The three major abiotic components include air, water, and soil. The biotic components, on the other side, include human beings, flora, fauna, and the microbes. The abiotic components are affected first by environmental pollution in which they directly affect the biotic components. Addition of contaminants to the environment has been taking place since human existence on the planet. There are two types of heavy metal contamination, i.e., natural or anthropogenic caused by human beings (Fig. 16.1). However, majority of heavy metals are mainly added by human beings themselves, thus making the environment unfit for leading good quality of life. For example, industrial wastes badly pollute our environment. Distillery industries are one of the examples of such industries that add polluted water to the soil. This water contains a mixture of organic and inorganic pollutants which may gain entry to food chain and directly affect the quality of life (Chowdhary et al. 2019; Thakare et al. 2021; Prasad 2021).

The type and quantity of contaminants vary in different countries. Intensity of severity is found higher in the developing and poorly developed nations, since they are careless about their environment. Industrial effluents are usually present in the surrounding areas without any treatment, thus become major health hazard for the people dwelling in such areas. The water, soil, and air are badly polluted, and the contaminants can easily gain entry to the food chain. Regretfully, a huge number of deaths occur every year due to diseases and illnesses caused by environmental

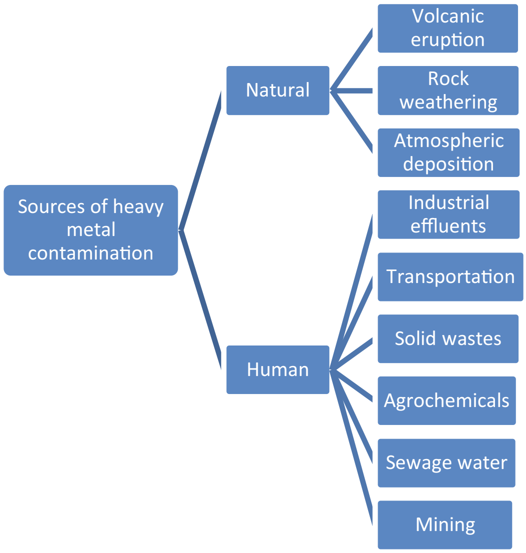

Fig. 16.1 Sources of heavy metal contamination

pollution. According to a report, nine million deaths occurred during 2015–2016 as result of environmental pollution (Gangamma 2018).

The severity of problem is increased where the people are usually illiterate, less educated, and totally unaware of the consequences of pollution. They manage to work in small industries and factories without bothering the extent of pollution that they are exposed to. Hence the problems of poor are aggravated by poor standards of life and health issues. The governments and administrative units, in such countries, are usually less concerned about the issues of environment. As a result, the environment gets more and more polluted without any check and control. Among different contaminants, heavy metals like lead, chromium, cadmium, copper, nickel, mercury, zinc, strontium, boron, selenium, arsenic, thallium, uranium, calcium, cobalt, manganese, nitrates, herbicides, and chlorinated compounds are highly toxic (Santos et al. 2018). Followings are sources of these toxic materials released in the environmental systems (Kanwar et al. 2020):

- Mining and smelters may cause the addition of As, Cd, Pb, and Hg metals.
- Various industries may add As, Cd, Cr, Co, Cu, Hg, Ni, and Zn metals.
- Atmospheric deposition may result into addition of As, Cd, Cr, Cu, Pb, Hg, and U.
- Agrochemicals may deposit As, Cd, Cu, Pb, Se, U, and Zn.
- Solid/liquid waste may cause addition of As, Cd, Cr, Cu, Pb, Hg, and Zn.

Besides, few bacteria may also add toxic mercury (mono- and/or dimethylmercury) to the environment that eventually polluted drinking water and food materials (Kumar et al. 2017). According to the United Kingdom Environment Agency (UKEA), there are some 1300 plus mining places that polluted soil and water reservoirs by adding different kinds of heavy metals like copper, cadmium, lead, and zinc (Foulds et al. 2014). Besides, the cosmetics and chemical fertilizers are also accountable for heavy metal pollution (Callender 2004).

Consequences of environmental pollution range from minor and negligible to serious problems for human. Among different problems that arise as a result of environmental pollution, the deterioration of human health is most eye-catching and alarming. A large number of diseases like renal dysfunction, alimentary canal problems strike human race every year, causing serious and irreversible health damage and even to death at times (Briggs 2003). Contamination of food is one of the major hazards that affect humanity worldwide. Environmental pollution has one more serious role and a potential threat to cause change at genetic level in any biotic component that resulted in life-threatening diseases and irreversible damages. Cancer is one of such devastating diseases, which owes large number of casualties every year in almost every part of the world (Boffetta 2006).

16.3 Types of Environmental Contaminants

It has been estimated that the pollution caused by heavy metals may surpass the other contaminants if it goes unchecked. A wide variety of contaminants exist that affect the quality of life to a great deal. The solid wastes and nuclear discharges are usually ranked as the worst pollutants, followed by heavy metals (Chen et al. 2003a). Following are the major types of pollutants that are predominantly found:

16.3.1 Inorganic Contaminants

An element which is found in periodic table cannot be further broken down into simpler parts. It is an entity in itself that has the potential to react with other elements to form compounds of various natures. The heavy metals are one of the major contaminants of soil that adversely affect the quality of soil and cause serious pollution, mostly affect the street and road-side soils (Christoforidis and Stamatis 2009; Li et al. 2001). Due to pure form, they cannot be broken down so they remain as such in the medium causing serious damages to the environment. Heavy metals, in minor quantities, work with enzymatic system of the plants to regulate physiological processes of plants, but at higher concentrations, they have negative impacts on plant growth and development. Arsenic, cadmium, zinc, copper, lead, iron, helium, neon, and solvents acetone, ethyl acetate, butanol, ethanol, methanol, deuterated water, hexane, chloroform, quercetin, and lots of chemicals are used in the laboratories of research institutes and hospitals (Charlesworth et al. 2017). Brief details about some of the heavy metals that pollute environment are given below:

16.3.1.1 Chromium

Chromium is abundantly found as part of rocks. In addition, it is found in the form of complexes with metals like lead (Pb), calcium (Ca), potassium (K), phosphorus (P), copper (Cu), aluminum (Al), sulfur (S), and others. It is found in different valent forms, the most reactive being Cr (VI) and Cr (III). Its natural forms are not serious environmental hazard as they are complexes of varying natures. Chromium may be released from rocks by natural weathering of rocks but this process is usually slow. On the other hand, anthropogenic activities add chromium to the environment as a reactive entity leading to serious health issues. It is mainly used in industries like alloying, tanning of animal hides, textile industry, lumber, and pigments. It has been observed that chromium in the form of chromate ions is most toxic due to its high solubility and ability to penetrate living membranes but its other forms like hydroxides, oxides, and sulphates are less toxic due to less solubility (Oliveira 2012). So, solubility of its chemical forms plays main role in its extent of toxicity. The contribution of leather industry as a source of chromium is now ranked first. Industrial cities such as Sialkot and Faisalabad (famous for their leather and textile industry) add the highest level of chromium to the environment compared to other cities of Pakistan. It is also worthwhile to mention that the incidences of cancer in such cities have increased many folds in the last decade.

16.3.1.2 Lead

Lead is another heavy metal which is toxic to living beings in many ways. The highest amount of lead is added by various industries to the environment leading to soil, water, and air pollution. Lead poisoning is a serious concern, and children are more severely affected by lead toxicity as compared with adults especially because they are not aware of the potential damages that it may bring. Lead toxicity may lead to a variety of health disorders ranging from abdominal pain, irritation, or lethargy to comma, anemia, and even neurological disorders (Hai et al. 2018).

16.3.1.3 Arsenic

Arsenic is one of the toxic contaminants of soil which is usually found in industrial wastes. It has the potential to damage human, plant, and animal health if it gains entry to the food chain (Prasad et al. 2013). Its bioavailability is predominantly affected by soil pH and can be increased by addition of organic chelating agents preferably citric acid (González et al. 2019).

16.3.1.4 Cadmium

Cadmium is added to the environment by natural processes from the earth's crust; however, anthropogenic activities lead to increase at higher levels. Processing of different metals like zinc, iron, and aluminum is the major way of its addition to environment. Besides, it is added through cigarette smoke, coal, and oil combustion from power plants and phosphate-based fertilizer applications. Higher cadmium levels not only affect plant physiology (especially respiration and transpiration), but they cause damages to microbial world and organisms dwelling in water (i.e., fish). The health damages from cadmium toxicity range from minor to major issues. Its higher levels may lead to cancer, birth defects, anemia, kidney damage, etc. Cadmium is mentioned as a "red list" metal as it may have serious impacts on health.

16.3.2 Organic Contaminants

An organic contaminant is the one that can be metabolized by plants and converted into inorganic constituents. Almost 30% of photosynthates of a plant are released into the rhizosphere by roots. A variety of phytochemicals and sugars are released by plant roots. The microbes that are associated with roots utilize such metabolites to gain energy. Such contaminants sometimes become serious hazard for the surrounding environment. They may gain entry to the water table and affect quality of biotic life.

16.4 In-Practice Strategies to Combat Environmental Pollution

Human beings have now realized the role of pollution in deteriorating and damaging health and overall life quality, and efforts are being made worldwide to protect environment of further damage. Following four main strategies are being used to control environmental pollution:

- To control the addition of contaminants to the environment. In this way, environmental pollution can be reduced by adopting procedures and strategies that add minimum pollutants. This strategy can play vital role in reducing environmental deterioration.
- To render such pollutants harmless or less toxic in an effective way by using different physical, chemical, and biological means. Extensive research may prove effective if all these means are studies in-depth.
- Increased awareness campaigns among masses thus educating them about environmental pollution and its disastrous results.
- Strict legislation and effective monitoring at the government level.

Some of the countries such as China, India, and Pakistan use the sewage water for growing vegetables and fruits due to lack of knowledge and awareness leading to transfer of contaminants to the food chain.

The abovementioned second strategy has been adopted by researchers worldwide to combat environmental pollution.

16.4.1 Bioremediation

American Environmental Protection Agency (EPA) defined bioremediation agents as “microbial cultures, enzymes and nutrient additives that significantly increase the rate of biodegradation to mitigate the effects of various pollutants” (Nichols 2001). Bioremediation can be performed both in situ and ex situ. In situ bioremediation can be performed at the contaminated site, while ex situ contamination involves removal of contaminated materials from one site and transfer to the other site after treatment.

Both approaches are successfully used but usually in situ approach is preferred being cost effective (April et al. 2000).

Some of the microorganisms have always been playing role as decomposers which decompose organic matter of all kinds and convert it to simpler inorganic substances. Similarly, some of the species of fungi and bacteria degrade hydrocarbons in the environment (Nilanjana and Chandran 2011). This is a natural decontamination process that takes place on its own (Venosa and Zhu 2003; Vinothini et al. 2015). The first reports on successful use of bioremediation report back to 1974 when a bacterial strain *Pseudomonas putida* was used to remove petroleum from contaminated sites. In those years, efforts were focused on identification of various microbial species in cleaning petroleum spills. The research work on microbial species got more attention and progressed further till 2000 A.D. Since this process takes place nicely at a fast rate under controlled conditions in vitro but it is comparatively quite slow in the field conditions (Venosa et al. 1996; Mearns 1997). So, the research concentrated more on phytoremediation, and the importance of plants as natural remediated captivated thoughts of researchers.

16.4.2 Phytoremediation

Phyto is derived from "*phyton*" which is a Greek word that means "plant," and "remediation" is derived from "*remedium*" a Latin word that means "to restore balance" (Cunningham et al. 1996). So, the term "phytoremediation" can roughly be defined as "a technique to restore balance by using plants." It can be properly defined as "a set of methods/technologies that employ living plants to clean up soil, air, and water contaminated with hazardous contaminants." Or it can be defined as "the use of plants along with other mechanical techniques to scavenge, remove or detoxify environmental contaminants" (Prasad 2017, 2018). The concept of phytoremediation was well known, and various plants are being used by the Neanderthal man for wastewater treatment for thousands of years ago (Rastogi and Nandal 2020).

The term "phytoremediation" was coined in 1991 by Ilya Ruskin. This technique has proved effective in removal of organic and inorganic pollutants from environment (Etim 2012). This technology has been widely accepted as an eco-friendly technology by researchers, academicians, and general public of different continents (Ghazaryan et al. 2019). The use of plants to deal with environmental pollution is not new. This concept has always been with human but it is practically caught more attention gradually. About 300 years ago, plants were used to clean water of contaminants. This technique has gained popularity across globe to such an extent that plenty of research activities, publications, conferences, and symposia are devoted to it every year for the past 50 years.

This technique employs natural physiological processes of plants (Etim 2012). Plants are unique organisms as they contain unique set of metabolic processes that can be used to tackle with environmental pollution. Growing plants in a contaminated matrix (soil, water, organic/inorganic debris) can fix problems of environmental pollution (USEPA 2000). They work in unique ways by fixing the contaminants in their bodies (immobilizing/binding the contaminants), degrading them by using their cellular machinery, or converting them in to less harmful or totally harmless forms (conversion). Once the plants have performed their role, they can either be removed or disposed off in appropriate ways. Over the years, plants have thankfully evolved in a way that they can deal with environmental pollutants by using their metabolic or physiological processes. Human beings can benefit from such processes. Plants can be used to clean up metals, herbicides, pesticides, solvents/toxic chemicals, explosives, crude oils, polyaromatic hydrocarbons, and landfill leachates from soil and/or water. They can be used to clean up river basins and even ocean bottoms that are accidentally polluted by oil spills, etc.

16.5 Types of Phytoremediation

Plants can work in different ways to deal with environmental contaminants. Phytoremediation can either be used alone or in combination with different chemical and/or mechanical procedures to clean up the environment (Etim 2012). The root system of plants plays vital role in absorbing contaminants from soil. The roots contain systems to protect themselves from harmful concentrations of contaminants. The roots provide larger surface area for absorption of such substances hence trees are considered better for this purpose as they have larger rhizosphere. Trees can play strong role in areas where contaminants are found in deeper layers of soil by pulling water up from their deep and wider rhizosphere. In some cases, the roots release few substances into their rhizosphere that play role in aggregation of soil particles, hence affecting the rate of absorption of contaminants from soil.

16.5.1 Phytoextraction

It is the type of remediation in which plants absorb contaminants from the environment in a harvestable form. It involves absorption of contaminants from soil and accumulation in above ground parts (preferably crown parts/foliage) of plant (Fig. 16.2). The soil can be used for growth of other plant species after proper remediation. The roots absorb the substances from soil and concentrate them in the above ground parts. Those plant species which can concentrate higher levels of contaminants in their bodies are called "hyperaccumulators." While those plant species which accumulate less contaminant may be cropped repeatedly to remove the medium of a contaminant. Such plants which hyper-accumulate toxic metals are generally regarded as metallophytes. Examples of such species include *Salix* and *Populus*. Phytoextraction has gained in popularity for the past few decades. It has been found effective in removal of inorganic substances or heavy metals. The contaminants usually accumulate in different plant parts. In case the plants have accumulated the contaminants up to a certain level, it becomes impossible for them to accumulate beyond that amount, and hence the remaining contaminants may leach down to the deeper soil layers.

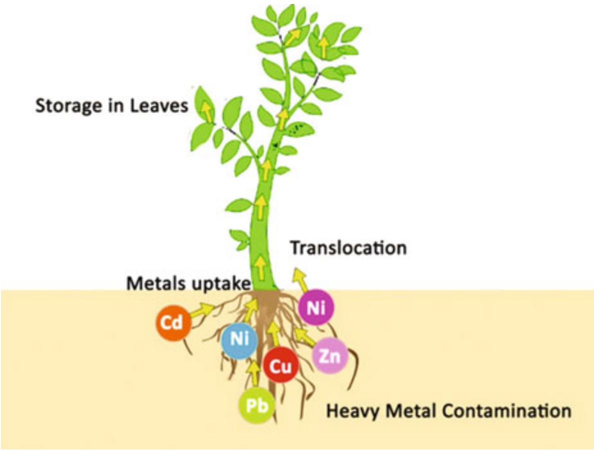

Fig. 16.2 Generalized figure to describe the process of phytoextraction in a plant. (Anoopkumar et al. 2020)

This strategy is helpful in remediation of soils contaminated with heavy metals, and a huge number of studies exist that reported use of different plant species to remediate soils affected with such metals. For example, Kaviani et al. (2019) used phytoremediation strategy to remediate Ni-contaminated soil. Their study is eye-catching as they not only measured the phytoextraction potential of a plant species (*Salicornia iranica*) but also measured the detoxification capacity of the said plant. This task was accomplished by measuring glutathione-S-transferase (GST) expression in addition to the other physiological parameters. They reported that the total chlorophyll and carotenoids were reduced after exposure to high dose of Ni (i.e., 500 mg/kg) recorded at different time intervals. They also reported a higher GST expression. The plant could accumulate higher levels of Ni in roots and aerial parts. The root and shoot lengths were reduced. The results showed that this plant species can be used for remediation of Ni-contaminated soils.

It is even more interesting to note that phytoremediation can be successfully applied where there is just single metal present in the medium. In case more than one metal are present, then only those metals can be absorbed which do not compete with each other for absorption by the roots. Not all metals in a medium can be absorbed simultaneously by the plant roots. This is because few metals antagonize the other metals absorption. This fact has been reported by many researchers. For example, in a latest research by Singha et al. (2019), iron plaque formation was observed on the roots of an aquatic macrophyte, *Pistia stratiotes* L. This iron plaque was formed by ferrous ions in the industrial wastewater. This plaque favored the extraction of iron and potassium and reduced the absorption of calcium from water. The absorption of cadmium was also suppressed but when the concentration of cadmium was raised to 500 μmol then its absorption by plaque containing roots increased. Cadmium detoxification was also observed in plants with iron plaque formation on roots. Kanwar et al. (2020) has reported various plant species which are used for eliminating different kind of heavy metals across the world (Table 16.1).

Helianthus annuus is known to absorb arsenic (Raab et al. 2005). *Pteris vittata* is another plant species that can accumulate arsenic (Fayiga et al. 2004). Willow (Salix smithiana) is good extractor of copper, zinc, and cadmium (Kacalkova et al. 2009). It has the ability to quickly transport the metal from points of absorption to upper parts of the plant. In addition, it produces high biomass which can be utilized for energy production. Alpine penny cress (*Thlaspi caerulescens*) has the ability to accumulate cadmium and zinc at higher levels (Cosio et al. 2004). But this species does not accumulate copper. *Salix viminalis* has been found effective in accumulating cadmium, another toxic metal (Mleczek et al. 2009). Different metal chelators can be used to increase efficiency of absorption of metals from medium. Among them, EDTA is one of the most famous and highly experimented chelators.

16.5.2 Phytovolatilization

Phytovolatilization involves the removal (by volatilization) of contaminants from a medium (from soil or water) and release in to the air (Limmer and Burken 2016). The substances are not released as such in to the air, rather they are converted in to less toxic and less harmful substances before their release. The contaminants are usually volatilized at the surfaces of leaves or stems but they may get evaporated from roots. Selenium and mercury are the metals that can be phytovolatilized by such plants. Plants with higher rates of transpiration can be effective in this regard, e.g., Poplar trees (Fig. 16.3).

16.5.3 Phytotransformation/Phytodegradation

Phytotransformation is also known as phytodegradation. In this process, the toxic elements are decomposed by the plants and rendered nontoxic or less harmful (Fig. 16.4). This method is advantageous as it can scavenge toxic substances from soil, water, and air also. In this method, the substances are not completely broken down into their simplest components instead they are transformed by the machinery of plants from one form to the other (Bock et al. 2002). The compounds that come in contact with plant are broken down either inside the body of plant or in the rhizosphere. It has been observed that special enzymes are released by such plants into the rhizosphere that decompose the organic matter in the matrix. It is effective in removal of various types of solvents from any matrix. We all know very well that plants are generally regarded as “lungs of nature” as they add oxygen to the environment. But the role of plants in this method resembles that of ‘human liver’ where liver has the role to detoxify the human body of harmful or toxic substances. So, the plants are usually regarded as “green liver” due to their metabolic capabilities that render different compounds nontoxic hence cleaning the environment of such contaminants. This method is under investigation by some of the research groups

Table 16.1 Uptake of various heavy metals by the higher plants (after Kanwar et al. 2020)

Toxic metal	Plant	Medium	Uptake of heavy metal (mg/kg)	References
As	*Pteris vittata* L.	Soil and water	8331	Kalve et al. (2011)
	Pteris ryukyuensis Tagawa	Soil	3647	Srivastava et al. (2006)
	Pteris quadriaurita Retz.		2900	
	Pteris biaurita L.		2000	
	Pteris cretica L.		1800	
	Eleocharis acicularis (L.) Roem. & Schult.	Water	1470	Sakakibara et al. (2011)
	Sedum alfredii Hance	–	9000	Xiong et al. (2004)
	Prosopis laevigata (Humb. & Bonpl. ex Willd.) M.C.Johnst.	–	8176	Buendía-González et al. (2010)
	Arabis gemmifera (Matsum.) Makino	–	5600	Kubota and Takenaka (2003)
	Salsola kali L.	Water	2075	de la Rosa et al. (2004)
	Azolla pinnata R.Br.	Water	740	Rai (2008)
	Deschampsia cespitosa (L.) P. Beauv.	Water	236.2	Kucharski et al. (2005)
	Corrigiola telephiifolia Pourr.	Soil	2110	García-Salgado et al. (2012)
Ni	*Alyssum bertolonii* Desv. [Syn. *Odontarrhena bertolonii* (Desv.) Jord. & Fourr.]	Soil	10,900	Li et al. (2003)
	Alyssum caricum T.R.Dudley & Hub.-Mor. [Syn.*Odontarrhena carica* (T.R. Dudley & Hub.-Mor.) Španiel, Al-Shehbaz, D.A.German & Marhold]		12,500	
	Alyssum corsicum Rob. ex Gren. & Godr. [Syn. *Odontarrhena robertiana* (Bernard ex Gren. & Godr.) Španiel, Al-Shehbaz, D.A.German & Marhold]		18,100	
	Alyssum pterocarpum T.R.Dudley [Syn. *Odontarrhena pterocarpa* (T.R.Dudley) Španiel, Al-Shehbaz, D.A.German & Marhold]		13,500	
	Alyssum heldreichii Hausskn. Syn. *Odontarrhena heldreichii* (Hausskn.) Španiel, Al-Shehbaz, D.A.German & Marhold	Soil	11,800	Bani and Pavlova (2010)
	Alyssum markgrafii O.E. Schulz [synonym *of Odontarrhena chalcidica*	Soil	19,100	

(continued)

Table 16.1 (continued)

Toxic metal	Plant	Medium	Uptake of heavy metal (mg/kg)	References
	(Janka) Španiel, Al-Shehbaz, D.A. German & Marhold]			
	Alyssum murale M.Bieb. [synonym of *Odontarrhena alpestris* (L.) Ledeb.]	Soil	4730–20,100	
	Alyssum serpyllifolium Desf.	Soil	10,000	Prasad (2005)
	Isatis pinnatiloba P.H. Davis	Soil	1441	Altinozlu et al. (2012)
Cd	*Phytolacca americana* L.	Soil	10,700	Peng et al. (2008)
	Sedum alfredi Hance		9000	Xiong et al. (2004)
	Prosopis laevigata (Humb. & Bonpl. ex Willd.) M.C.Johnst.	Soil	8176	Buendía-González et al. (2010)
	Arabis gemmifera (Matsum.) Makino [Syn. *Arabidopsis halleri* subsp. *gemmifera* (Matsum.) O'Kane & Al-Shehbaz]	–	5600	Kubota and Takenaka (2003)
	Salsola kali L.	Water	2075	de la Rosa et al. (2004)
	Azolla pinnata R.Br.	Water	740	Rai (2008)
	Deschampsia cespitosa (L.) P.Beauv.	Soil	236.2	Kucharski et al. (2005)
	Rorippa globosa (Turcz. ex Fisch. & C. A.Mey.)	Soil	>100	Wei et al. (2008)
	Thlaspi caerulescens J. Presl & C.Presl [Syn. *Noccaea caerulescens* (J.Presl & C.Presl) F.K.Mey.]	Soil	263	Lombi et al. (2001)
	Azolla pinnata R.Br.	Water	740	Rai (2008)
	Pteris vittata L.	Water and soil	20,675	Kalve et al. (2011)
	Eleocharis acicularis (L.) Roem. & Schult.	Water	11,200	Sakakibara et al. (2011)
	Thlaspi calaminare (Lej.) Lej. & Courtois [Syn. *Noccaea caerulescens* subsp. *calaminaris* (Lej.) Holub]	Soil	10,000	Sheoran et al. (2009)
	Deschampsia cespitosa (L.) P.Beauv.	Soil	966.5–3614	Kucharski et al. (2005)
Hg	*Achillea millefolium* L.	Soil	18.275	Wang et al. (2012)
	Marrubium vulgare L.	Soil	13.8	Rodriguez et al. (2003)
	Rumex induratus Boiss. & Reut.	Soil	6.45	Rodriguez et al. (2003)

(continued)

Table 16.1 (continued)

Toxic metal	Plant	Medium	Uptake of heavy metal (mg/kg)	References
	Silene vulgaris (Moench) Garcke	Soil	4.25	Pérez-Sanz et al. (2012)
	Festuca rubra L.	Soil	3.17	Rodriguez et al. (2003)
	Poa pratensis L.	Soil	2.74	Sas-Nowosielska et al. (2008)
	Helianthus tuberosus L.		1.89	
	Armoracia rusticana G. Gaertn., B.Mey. &		0.97	
	Juncus maritimus Lam.	–	0.315	Zheng et al. (2016)
	Cicer arietinum L.	Soil	0.2	Wang et al. (2012)
	Eleocharis acicularis (L.) Roem. & Schult.	Water and soil	20,200	Sakakibara et al. (2011)
	Aeollanthus biformifolius De Wild. [Syn. *Aeollanthus subacaulis* var. *linearis* (Burkill) Ryding]	Soil	13,700	Chaney et al. (2010)
	Ipomoea alpina Rendle [Syn. *Ipomoea linosepala* subsp. *alpina* (Rendle) Lejoly & Lisowski]	–	12,300	Mitch (2002)
	Haumaniastrum katangense (S.Moore) P.A.Duvign. & Plancke	Soil	8356	Sheoran et al. (2009)
	Pteris vittata L.	Soil	91.975	Wang et al. (2012)
Cr	*Pteris vittata* L.	Soil and water	20,675	Kalve et al. (2011)
Pb	*Medicago sativa* L.	Soil	43,300	Koptsik (2014)
	Brassica juncea (L.) Czern.		10,300	
	Brassica nigra (L.) W.D.J.Koch		9400	
	Helianthus annuus L.		5600	
	Betula occidentalis Hook.		1000	
	Euphorbia cheiradenia Boiss. & Hohen.	Soil	1138	Chehregani and Malayeri (2007)
	Deschampsia cespitosa (L.) P.Beauv.	Soil	966.5	Kucharski et al. (2005)
	Euphorbia cheiradenia Boiss. & Hohen	Soil	1138	Chehregani and Malayeri (2007)

across the globe, with special emphasis on phytodegradation of organic compounds, e.g., methyl-tert-butyl ether, herbicides, tri-chloroethylene, industrial substances, xenobiotics (Newman and Reynolds 2004).

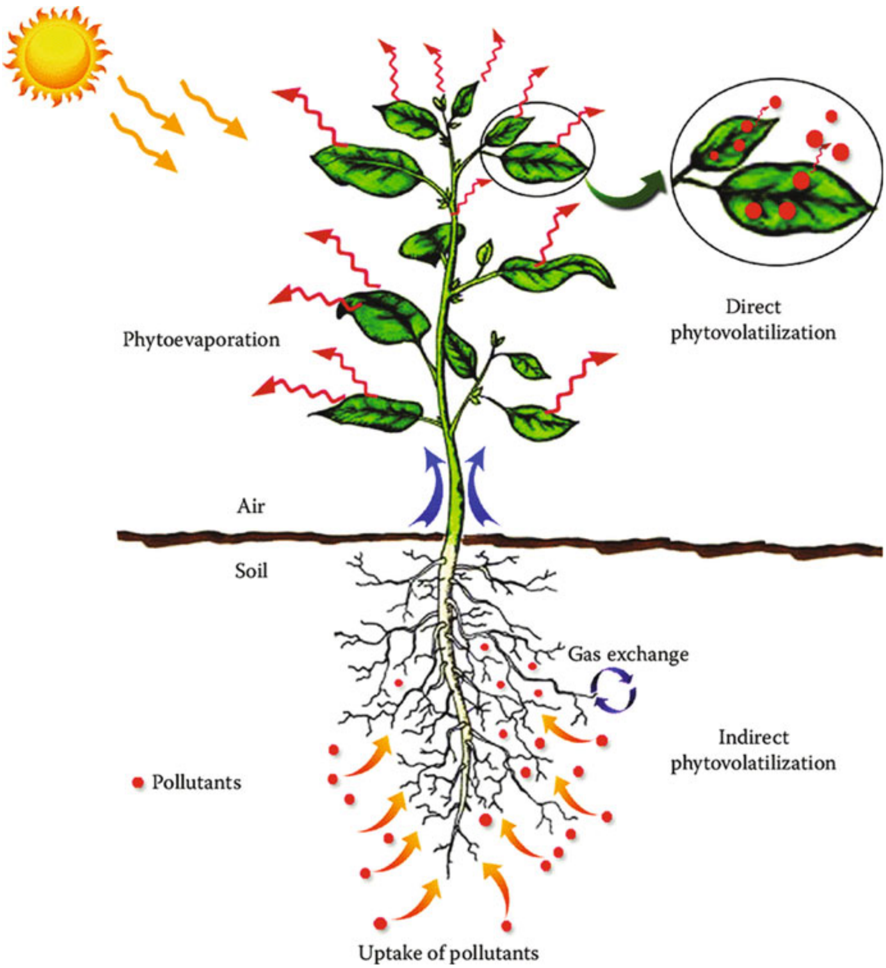

Fig. 16.3 Generalized figure to describe the process of phytovolatilization in a plant (Chandra and Kumar 2018)

Few plant species effectively degrade toxic substances and render them totally nontoxic using their cellular machinery while others immobilize and fix such chemicals in their bodies in nonextractable form. The compounds which are fixed/ stored in the plant body are dealt in a way that they do not affect the health of the plant itself. It is also interesting to note that, in some cases, the microbes in association with few plant species have the ability to metabolize and decompose such compounds in the rhizosphere. The recent studies are focused on finding the mechanisms of transformation in different plant species that are good at such transformation. The studies done so far reflect that there are three stages/phases of this transformation that usually start with adding polarity to the contaminants.

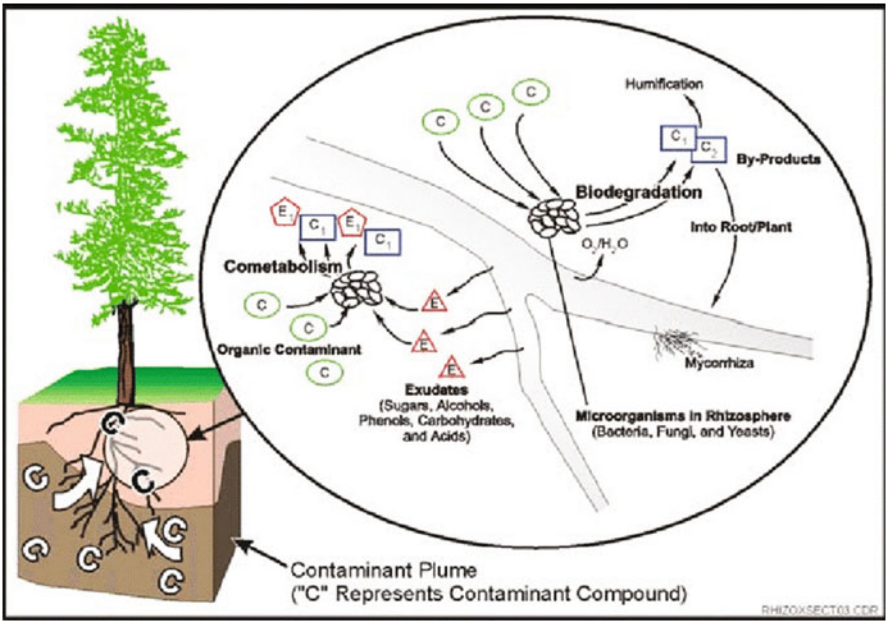

Fig. 16.4 Generalized figure to describe the process of phytotransformation/phytodegradation in a plant (Longley 2021)

Among all other contaminants, so far, the transformation mechanism of trinitrotoluene has been studied in-depth (Kiiskila et al. 2015).

This technique involves transformation of pollutants by enzymatic degradation. This technique has been used by US Army to remediate water contaminated with TNT and RDX at Milan Army Ammunition Plant at Tennessee. This approach has the potential to remediate water in situ or ex situ. The US Air Force has also employed such procedures to investigate the potential of this technology in remediation of environmental components (Best et al. 1997). It is interesting to note that few studies have reported that some of the transformed compounds are released into the air from plant surfaces (Newman and Reynolds 2004).

16.5.4 Rhizofiltration

It may be defined as "a process in which plant roots are employed to filter water/soil of contaminants." The pollutants are either absorbed (concentrated in the plant body) or adsorbed to the roots (Fig. 16.5). It actually involves transfer of pollutants from soil to the plant roots. The plants are generally grown first in a greenhouse (in pots) or in a hydroponic system. Then, this process can be performed either directly at the contaminated site to free water of contaminants or contaminated water can be collected and taken to the site where plants are growing (i.e., off-site area). Few

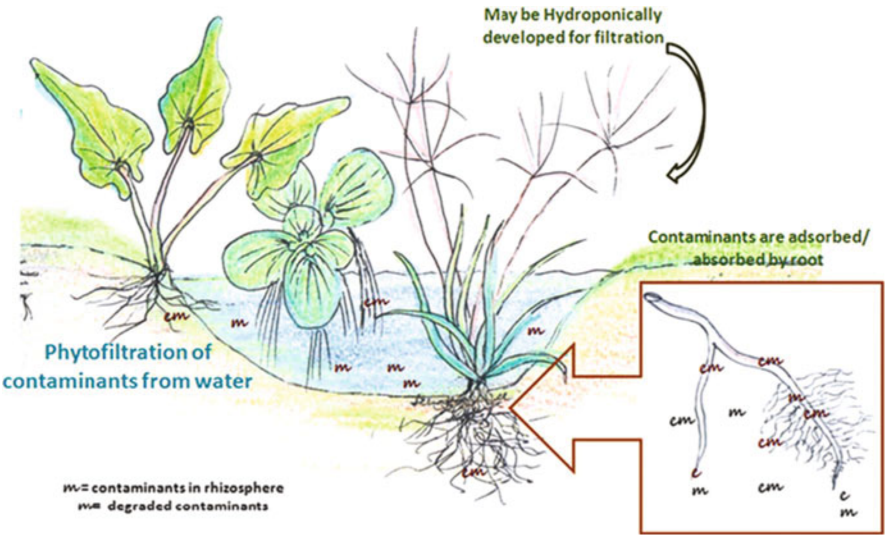

Fig. 16.5 Generalized figure to describe the process of rhizofiltration in a plant (Datta et al. 2013)

plant species that have gained popularity in this regard include *Helianthus annuus* L., *Brassica juncea* (L.) Czern., *Phaseolus vulgaris* L. var. *vulgaris,* and a number of members from Poaceae family. The metals that have been effectively removed so far include copper, zinc, chromium, cadmium, lead, and uranium. Among different radioactive metals, uranium (238 U) has become a serious concern for all nations as its mining and other activities keep adding it to the atmosphere. Some plant species are good at absorbing this radioactive contaminant. Such plants usually absorb it through their roots (Gupta et al. 2019). In a study, sunflower efficiently absorbed almost 80% of uranium from water contaminated with the said metal. Interestingly, the heavy metal was absorbed within just 24 h. It reflects the strength of this technique in removal of heavy metals from contaminated water.

16.5.5 Phytodesalination

This method may be described as "a method that employs the plants tolerant to higher concentrations of salts to clean a medium of excess salts" (Fig. 16.6). Such plants are generally regarded as "halophytes." The soil, after removal of salts, can be used for agricultural purpose. This technique has been studied extensively especially in countries with saline areas in order to free soil of excess salts and improve its qualities for growing food crops. Efforts have been made to identify salt-tolerant genes from different organisms including microbes and introduction of such genes into selected plant species to be used for phytodesalination (Walid et al. 2012).

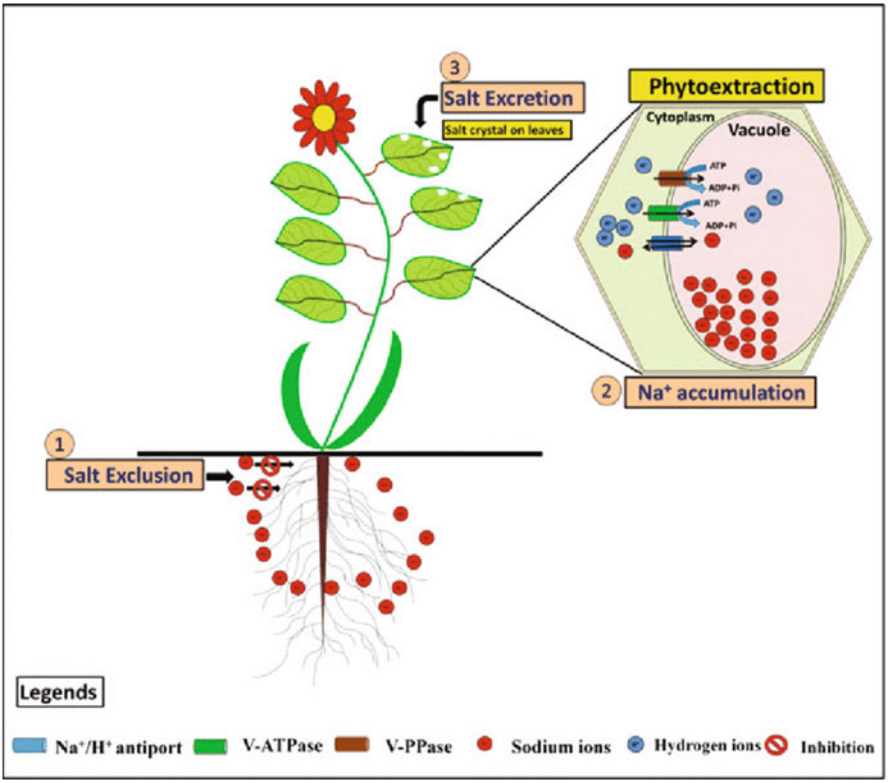

Fig. 16.6 Generalized figure to describe the process of phytodesalination in a plant. (Saddhe et al. 2020)

16.5.6 Phytostimulation

In this method, the plant roots enhance and support microbial growth in the near surface zone of earth crust, and the contaminants are degraded by those microbes. This approach is named as "plant-assisted remediation" by some researchers. The concept of this technique revolves around an increase in microbial growth and activity in rhizosphere and resultant degradation of contaminants in soil (Fig. 16.7). Since this process is bound to happen around any plant species provided that the soil structure favors microbial growth and degradation in rhizosphere.

It has been observed that the plant roots increase the growth of microbes in three major ways; (1) by adding organic matter to the rhizosphere (by death and decay of roots), (2) by respiration (thus adding oxygen), and (3) by secretion of roots exudates. Since the growth of microbes is enhanced by activity of plant roots so this method is dependent on plant growth in that contaminated area (Hussain and Hasnain 2011).

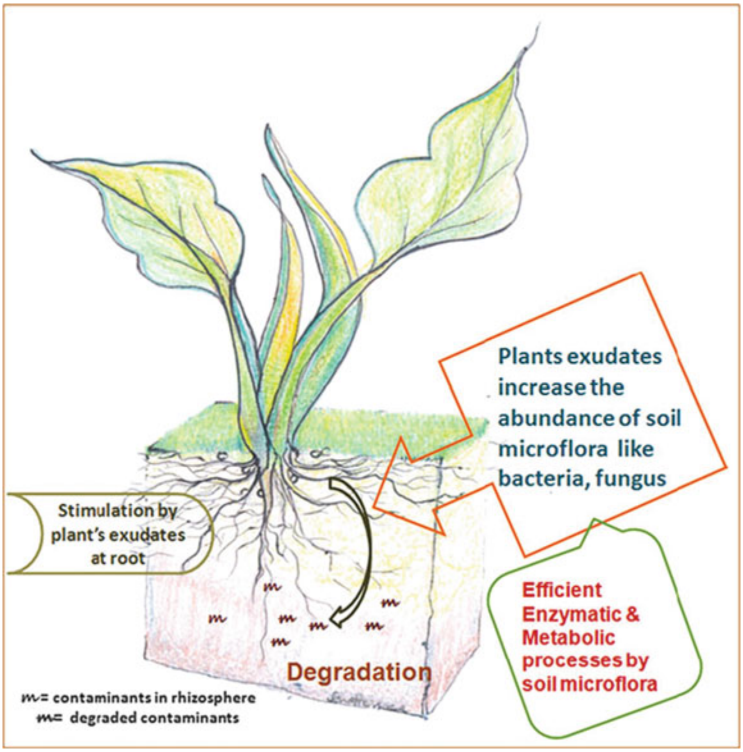

Fig. 16.7 Generalized figure to describe the process of phytostimulation (Datta et al. 2013)

16.5.7 Phytostabilization

In this method, heavy metals are stabilized and reduced at the plant–root surface and their mobility is reduced. It is just like captivating something and not letting it go anywhere else (Fig. 16.8). Soil erosion and leeching of contaminants are higher in areas with little vegetative cover. So, growth of contaminant-tolerant plants in such area helps fix the contaminants in the root zone. In this way, the spread and exposure of such contaminants to surrounding environment is reduced. The soil is gradually reclaimed by vegetative cover. Once the soil properties are restored to normal, different crops can be cultivated in routine (Bolan et al. 2011).

16.5.8 Hydraulic Control

In this method, tree roots control/limit water movement by strong pumping action. The water cannot move to deeper soil layers, rather it moves up by hydraulic pressure of roots. Greater volumes of water can be pulled by trees on daily basis. For examples, 30 gallons of water are pulled up by pumping action of Poplar tree

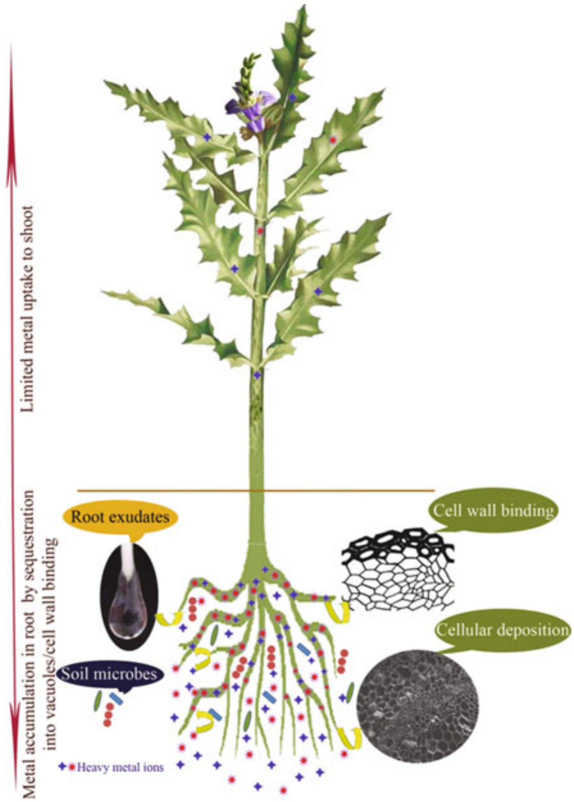

Fig. 16.8 Generalized figure to describe the process of phytostabilization (Shackira and Puthur 2019)

roots per day. Similarly, cotton wood tree can pull up to 350 gallons of water per day. It has been documented by Environmental Protection Agency of USA that Poplar trees have great potential to limit the leaching down of various contaminants by pulling water up. Poplar trees have great potential to remediate soil of toxic elements (Castro-Rodríguez et al. 2016). These trees have been shown to remediate soil of high levels of nitrates (O'Neill and Gordon 1994).

16.6 Factors Affecting Phytoremediation

Following factors have been studied so far that can affect the rate of phytoremediation. These factors, in simple words, play role in enhancement of efficiency of phytoremediation. These factors are discussed below:

16.6.1 Plant Species

The selection of plant species and proper planning of phytoremediation involves the study of the following:

1. The type of contaminants (organic/inorganic/metallic/mixtures of metallic, organic, and/or inorganic).
2. The nature of contaminated medium (soil/water/air/organic or inorganic debris/combination of soil, water and/or organic, and inorganic debris/sediments).
   (a) In case of soil, the depth of the soil that is contaminated helps determine the right type of plant species. For example, if the top layer of the soil is contaminated, then the use of trees is not favorable, rather shallow rooted plant species can be useful in this regard. While, in case of contamination of deeper soil layers, trees can play effective role in remediation of soil.
   (b) If contaminants are found in water, then it can be a water reservoir like a pond, lake, river, stream, ocean, or waterfall. For example, if contaminants have leeched down to the level of groundwater table, then tree species can play proper role in pumping the water up using their extensive root system.
   (c) Waste effluents of factories/industries can either be organic or inorganic debris or mixtures of one or more types of contaminants.
   (d) Wastes from residential areas (solids/liquids).
3. The climatic and environmental conditions of the contaminated site.
4. The nature of solid wastes, metallic, or nonmetallic.
5. The nature and type of soil particles along with its physical and chemical characteristics. Since the soil is mostly the medium of growth for most of the plants.
6. In case of air, the study of air quality index is also important.

It has been observed that the efficiency/rate of phytoremediation differs among different plant species. It has also been shown that different plant parts have varying potentials/tendencies to accumulate contaminants in them. For example, it has been reported by Firdaus-e-Bareen et al. (2019) that heavy metal phytoextraction capacity was different in two selected plant species, i.e., sorghum and pearl millet. They found that pearl millet had higher efficiency of extraction of heavy metals. And at the same time, different plant parts showed different specificity for accumulation of metals with roots ranked the first followed by stem and leaves.

Different plant parts accumulate different levels of heavy metals. It was observed by many researchers. For example, Khan et al. (2019) reported higher metal accumulation in roots than in foliar parts of *Petunia hybrida* L. copper got accumulated in roots and the other metals (Cd, Cr, Cu, Ni, and Pb) in above ground parts of the same plant. They reported that the plant underwent through heavy stress in contaminated water. Its physiological processes were disturbed to a great deal under heavy metal stress. The quality of plant aesthetics decreased when it was exposed to higher concentrations of heavy metals. Yet the said plant can be used for phytoextraction of selected metals. Similarly, some other species including *Helianthus annuus*, mustard plants (especially *Brassica juncea*), *Apocynum canabinum*, *Pteris vittata*, *Salix* spp., *Beta vulgaris*, Ragweed (*Ambrosia artimisifolia*), *Populus* spp. have also been found effective in this regard.

A comprehensive study of mechanisms/physiological processes of plant species should also be conducted prior to phytoremediation. It is important to mention here that few contaminants, if continue to stay in a medium/matrix (for some time at least), they support the growth of various types of microbial species. Hence, the selection of plant species depends on those microbial species also. In such a case, tolerance and response level of plant species to such factors must be studied prior to planning a phytoremediation project. It is also noteworthy that plant species may be selective in remedying a medium. In addition, a variety of plant species may be needed to grow simultaneously in a medium based on the type of contaminants found in it. Rotation of different plant species at the same site/area may be needed, depending upon the type of contaminants. Among different plant species halophytes are well known for their role as phytoremediators. The role of halophytes is acknowledged worldwide. They are known to remediate coastal and other areas due to their hyperaccumulation potential.

16.6.2 Soil Amendments

The research has accelerated on finding the mechanisms, strategies, and methods to enhance the phytoremediation efficiency/rate. Amendment of soils is considered as one of the major ways to do so. Many researchers across the world have conducted such researches which involve addition of different chemical, microbial, and other agents to the rhizosphere. It is noteworthy that researchers have proved that soil amendments have remarkable role in improvement of phytoremediation rate. Soil amendments that have been experimented so far include the use of chelating agents of various natures (ranging from organic to inorganic, natural to synthetic), microbial species and even varying combinations of such agents. All these agents are discussed below:

16.6.3 Chelating Agents

Chelating agents are those substances that chelate with the contaminants in soil/water and enhance their accumulation and bioavailability in the medium. Chelating agents mostly function by formation of coordination complexes with metals in the medium. Hence, we can say that the phytoremediation efficiency is enhanced by such agents. There are two major types of chelating agents, i.e., organic and inorganic. The inorganic ones are usually synthetic agents. The inorganic ones have been considered more effective in increasing bioavailability of contaminants in the medium (Dineshkumar et al. 2019). Prominent examples of inorganic chelating agents include EDTA, DOTA, EDDS, DTPA, EDDHA. On the other hand, organic ones are those that are derived from living organisms. Examples of such agents include proteins, carbohydrates, nucleic acids, and various types of organic acids. The organic acids that have been experimented so far to enhance phytoremediation rate include 2,3 Dihydroxy benzoic acid, citric acid, homo citric acid, and gluconic acid.

16.6.3.1 Organic Chelating Agents

It is the interaction of roots, soil particles, dissolved, and un-dissolved materials in the soil liquids that decides the overall nature of rhizosphere and absorption of contaminants from soil. Organic acids play crucial role in deciding the soil pH. The acidic environment has effects on solubility and subsequent bioavailability of contaminants in the soil. For example, lead oxides, carbonates, and sulphates are readily soluble in acidic medium (Traina and Laperche 1999).

Rhizosphere is modified not only by proton contributing properties of acids but by their action as ligands that chelate with metals. Redox reactions also occur in the rhizosphere as soon as the acids are added to it. Such reactions can affect metal mobility in the rhizosphere (Violante et al. 2010). These acids are usually weak and their pK_a values range from 3 to 9. Their molecular weight varies greatly. The lowest molecular weight of organic acids is possessed by oxalic and citric acids (Wei et al. 2009). The acids with lower pK_a values generally have carboxylic functional group while those with higher values have phenolic group. Irrespective of their functional groups, they generally increase metal solubility in the soil water and affect weathering process. Various researchers from different countries have been working on this aspect, e.g., Wu et al. (2012) tried to assess the effect of organic amendments on phytoremediation using *Sedum* spp. Among organic chelating agents, following three acids have been researched the most:

16.6.3.1.1 Citric Acid

Citric acid is a well-known organic agent that is famous among masses for its health promoting effects. It is a metabolite of almost all living aerobic organisms. It has high antioxidant activity and is usually found in high concentrations in fruits especially those with bitter/sour taste (like lemon and orange). It is a weak acid and is used by general public to add sour taste to foods. Its molecular formula is $C_6H_8O_7$ (Kaushik 2015). It is a tricarboxylic acid. It has great potential to chelate heavy metals in the medium and get absorbed by plant roots at a greater pace due to its small-sized molecules. The effect of citric acid supplementation has been found effective by few researchers, e.g., Turgut et al. (2004) and Chen et al. (2003b).

16.6.3.1.2 Oxalic Acid

Oxalic acid is yet another organic acid that is being studied for its chelating tendencies for different metals in the soil solution. It is a metabolite of human, algae, and plants as well. It is a dicarboxylic acid, produced in the living cells by metabolism of ascorbic or glyoxylic acid. Interestingly, the bodies cannot metabolize it and it is excreted out of the body as a waste product. It is a good reducing agent and chelates with metals thus increasing their phytoavailability in the soil solution (Wang et al. 2019). Oxalic acid has been shown to increase bioavailability of cadmium in the soil (Hou et al. 2019). Oxalates have been reported to impart tolerance to plants growing in soils contaminated with aluminum, lead, cadmium, and zinc (Rajendra and Yashbir 2017).

16.6.3.1.3 Gluconic Acid

Gluconic acid is also a naturally produced mild organic acid found mainly in honey, fruits, teas, and wine. Its molecular formula is $C_6H_{12}O_7$ and pK_a value is 3.7 (Kaushik 2015). It is generally produced in the bodies of microorganisms (e.g., *Aspergillus niger* and Gluconobacter) by degradation of glucose (Ramachandran et al. 2006). Gluconic acid and its derivative sodium gluconate have huge applications in food and pharmaceutical fields. It effectively chelates with metals including iron, aluminum, and calcium. It works best in alkaline environment.

In an interesting study by Hu et al. (2019), citric acid, oxalic acid, and EDDS (ethylenediamine disuccinic acid) were applied to the soil contaminated with higher uranium levels. They grew Macleaya cordata in those affected soils and observed the effect of aforementioned chelating agents. Citric acid was found to be the most effective chelating agent while oxalic acid was not much efficient in increasing the bioavailability of uranium in soil. The antioxidant system of the plant performed well against the oxidative stress caused by both chemical entities applied. Similarly, cadmium availability is improved by addition of oxalic acid in contaminated soils. Bioavailability of other metals has also been studied in organic acid supplemented soils.

In addition to the abovementioned organic acids, malic acid, tartaric acid, homocitric acid, and 2,3-Dihydroxy benzoic acid have also been investigated for their possible effects on phytoremediation. But only few studies can be observed in this regard. It is also interesting to note that organic acids, not always, increase the phytoremediation rate. Sometimes, their role has been either negligible or negative. This situation reflects that more studies are needed to properly investigate their role in remediation of contaminated sites/media.

16.6.3.2 Synthetic Chelating Agents

The effect of synthetic chelating agents on efficiency of phytoremediation of different plant species growing in soils contaminated with different metals has been studied extensively. Among different synthetic agents, EDTA has been experimented the most for its role in phytoremediation. It has been well-established that EDTA has strongly positive role in this regard as it does accelerate remediation of contaminated sites. Jiang et al. (2019) provided the first successful evidence in 2019 that lead absorption was enhanced by addition of EDTA to soil. The bamboo plants efficiently absorbed higher lead concentrations quickly when EDTA was employed as a chelating agent. They used such higher concentrations of lead up to 0–1500 mg/kg, and EDTA was used from 250 to 1000 mg/kg.

Recent research by Gul et al. (2019) also supported the view that EDTA has strongly positive effect on phytoremediation. They studied the effect of EDTA supplementation on phytoremediation efficiency of two selected plant species (*Pelargonium hortorum* and *P. zonale*) from Pb and Cd contaminated soil was evaluated.

Different concentrations of lead (0 to 1500 mg/kg), cadmium (0 to 150 mg/kg), and EDTA (0 to 5 mmol/kg) were employed. They reported marked difference in phytoextraction efficiency of both plant species where *P. hortorum* showed higher phytoremediation potential. In addition, the EDTA supplemented soils lead to higher phytoextraction of contaminants from soil.

In another recent study by Dou et al. (2019) reported an enhanced uptake of cadmium by plant species *Bidens pilosa* when EDTA was added as a chelating agent. They also demonstrated that cadmium could be efficiently absorbed at equal rate irrespective of its type (sulfate/phosphate/chloride of cadmium). The only factor that affected the rate of uptake was the chelating agent itself. In another valuable study by Chaturvedi et al. (2019), phytoremediation potential of *Brassica oleracea* L. and *Raphanus sativus* L. plant species was investigated. These plants were grown in soils contaminated with different heavy metals including zinc and lead. They documented that the metal uptake was enhanced when chelating agent was added. This study also supported the idea of soil amendment to improve heavy metal uptake by plants.

In yet another study, the metal uptake, tolerance to extracted metal, and biomass was increased when soil was amended with addition of EDTA as chelating agent. This study was conducted by Wasino et al. (2019). They employed EDTA as chelating agent for three heavy metals cadmium, zinc, and lead. They used plant species *Chrysopogon zizanioides* and *C. nemoralis* for phytoremediation. They recorded an increase in absorption efficiency of the said plant species when EDTA was employed. Both species showed significant remediation potential in case of soil contaminated with all three metals.

It must be kept in mind while experimenting with such agents that applied concentration does carry weightage in affecting solubility, extractability, translocation, and other parameters of phytoremediation. For example, EDTA and other chelating agents do increase phytoremediation efficiency but their higher concentrations have opposite effect. The study of Yu et al. (2019) is worth mentioning here. They reported that higher levels of EDTA had negative impacts on remediation of soil contaminated with manganese using *Polygonum pubescens*.

It is also noteworthy that addition of EDTA is not always fruitful. It may miraculously increase uptake of a metal by a plant species but at the same time it may not increase uptake of another metal. For example, Ghazaryan et al. (2019) found that the uptake of copper was enhanced in EDTA supplemented medium while there was no such increase in uptake of molybdenum by the same plant species under the same conditions. It shows that prior to planning a project, a pre-hand study must be done to investigate the effect of different factors that may influence phytoremediation efficiency of a plant species.

Interestingly, studies do exist which compare the effects of synthetic and natural chelating agents on phytoremediation (Wu et al. 2004). For example, Wu et al. (2004) reported that EDTA enhanced the uptake of two of the four experimented heavy metals, i.e., Cu and Pb by *Brassica juncea* while organic acids including oxalic, citric, or malic acid had no remarkable effect. All chelating agents were used at equal concentration, i.e., 3 mmol kg^{-1}. These results show superiority of the use of EDTA over organic acids in influencing the rate of phytoremediation. DOTA (1,4,7-Tetraazacyclododecane 1,4,7,10 Tetra acetic acid) is yet another popular synthetic agent that has shown its effects on phytoremediation. It is also known as Tetraxetan. Its IUPAC name is 2-[4,7,10-tris (carboxymethyl)-1,4,7,10-tetrazacyclododec-1-yl] acetic acid, and its molecular formula is $C_{16}H_{28}N_4O_8$. It is mostly used to chelate with lanthridine (Kaushik 2015). Other synthetic agents have also been investigated for their potential effects on phytoremediation but the highest impacts have been produced so far by EDTA that has already been discussed in this chapter.

16.6.3.3 Combined Effect of Organic and Inorganic Chemical Agents

Some other researchers have documented the idea of soil amendments to increase bioavailability of contaminants in soil. Majority of them agreed upon the use of fertilizers to support plant growth and subsequent increased absorption of contaminants. In a study, Zhang et al. (2019) has experimented the use of EDTA and silicon-based fertilizers to see their effect on phytoextraction by rice plants. They observed an increase in phytoextraction of cadmium by rice plants when EDTA and fertilizers were applied simultaneously. Shahid et al. (2019) had experimented the effect of EDTA and citric acid was evaluated on physio-biochemical traits of young and old bean leaves under cadmium stress. They reported that EDTA enhanced Cd uptake and accumulation and decreased its toxicity by controlling different physio-biochemical traits. But citric acid surprisingly reduced uptake of heavy metal. It shows that EDTA enhances metal uptake and protects plants against its damages while citric acid which is an organic acid reduces metal uptake.

Some researchers are now looking for some suitable additives that reduce the toxic effects of metals and EDTA on plants. In a latest and interesting study conducted by Revathi and Subhashree (2019), sodium nitroprusside has been used to see its effect on phytoremediation efficiency and physiological processes of the plant. It was observed by them that in absence of sodium nitroprusside, the antioxidant activity in plant increased upon addition of EDTA and heavy metal which means that the plant needed to get rid of free radicals. But when sodium nitroprusside was added, the antioxidant enzyme activities (catalase, superoxide dismutase, ascorbate peroxidase, and glutathione reductase) reduced. It clearly shows that the additives like sodium nitroprusside can reduce the stress levels in plants and indirectly enhance phytoremediation potential of the plant.

16.6.4 Microorganisms

Plant growth promoting rhizobacteria, several fungal species, mycorrhiza, endophytes, and algae have been shown to increase the rate of phytoremediation (Umesh et al. 2016). Among these, plant growth promoting rhizobacteria (PGPRs) play role in strengthening the plants against abiotic stresses by improving the efficacy of soil and plant growth promotion (Prasad et al. 2015). They also have role in influencing the crop sustainability by increasing production of various enzymes, nitrogen fixation, and solubilization of phosphorus and potassium in the soil. In a study, they have been proved to improve cadmium bioavailability in leguminous plants by bioaccumulation and by formation of complexes and chelates (Jebara et al. 2019).

Plant–microbe interactions have been extensively studied for the past two to three decades. But the research on this aspect has accelerated enormously for the past few years. At the same time, plant microbe and metal interaction has also seen extensive interest and research. Plant microbes usually include different bacterial species and mycorrhizae. Such microbes make symbiotic associations with plant roots, thus getting mutual benefits. This shows that plant-associated microbes can play vital role in remediation of contaminated environment. There are various studies available that comply with this statement and prove it. For example, it has been shown by Jan et al. (2019) that soil under high cadmium stress can be remediated by rice seedlings with the help of *Bacillus cereus,* a bacterial species.

Since this view has been well-established that biogeochemical interactions play vital role in bioavailability and uptake of environmental contaminants, researchers have been looking for appropriate microbial species to investigate their effect on phytoremediation potential of plants. Since microbial species are plant-species-specific and the properties of soil are also crucial to their growth and sustenance and overall performance, optimization of conditions for microbial-phytoremediation is the point of focus for researchers. Most of the researchers agree that different types of microbial metabolites have role in adjustment of rhizosphere and hence subsequent phytoremediation. Such metabolites include indole-3-acetic acid, organic acids, siderophores, and 1-amino-cyclo-propane-1-carboxylic acid deaminase (Rajkumar et al. 2012). Recently, microorganisms are being genetically engineered with two main objectives. The first one is to increase their efficiency of pollution control (which is by modification in their innate metabolic characteristics), and the other one is to regulate plant growth. Both these strategies ultimately strengthen phytoremediation efficiency (Mishra et al. 2019).

16.6.5 Combination of Chelating Agents and Microorganisms

Some researchers have reported that use of combinations of different factors improved phytoremediation efficiency. For example, Asilian et al. (2019) reported that combination of chemical and microbial approaches enhanced phytoremediation efficiency of maize plants. They used a bacterial (*Pseudomonads fluoresce*) and a fungal species (*Piriformospora indica*) along with Tween-80 surfactant for this purpose. They grew maize seeds in cadmium polluted soil and observed the status of plant growth and cadmium levels in plant tissues. Their study provided evidence for higher phytoremediation efficiency of maize plants after combined application of microbial and chemical factors.

In yet another latest study, Yasin et al. (2019) tested the effect of EDTA and a bacterium *Enterobacter* sp. CS2 on phytoextraction efficiency of a plant species *Impatiens balsamina* L. from soil. The researchers used soil contaminated with industrial effluents carrying different concentrations of nickel (Ni). The seeds of the said species were soaked in this contaminated soil for 50 days and Ni-tolerance index, bioconcentration, and translocation factor were observed. They expertly found out that Ni reduced plant growth and development in absence of both factors, i.e., bacterium and chelating agent. But the plants had higher tolerance level for the metal when the soil was supplemented with bacterial species *Enterobacter* sp. CS2. In addition, EDTA supplementation enhanced metal uptake by plant. Hence their study clearly indicated that combination of microbial and chelating agent supplementation has direct effect on efficiency of phytoremediation.

16.6.6 Combined Effect of Organic and Synthetic Chelating Agents

Few studies have focused on evaluation of combined effect of organic and inorganic chelating agents. Guo et al. (2018) reported application of chelating agents with the potherb *Brassica juncea* while growing in soil contaminated with a smelter. Two heavy metals viz., zinc and cadmium, were found in the soil which was efficiently absorbed by the plants after application of EDTA in combination with citric acid and oxalic acid. The chelating agents were added to soil 3–4 weeks after sowing. The accumulation of both metals was enhanced almost 1.5–3 folds in different experiments. While the plant physiology went through heavy stress and the antioxidant enzymes were produced at a higher concentration. Their study provided interesting results. For example, the highest phytoremediation efficiency was observed with single chelating agent, i.e., EDTA alone which was followed by combination of EDTA and organic acid. It is interesting to note that only organic acids were added to the medium before phytoremediation, and there was no significant increase in phytoremediation efficiency. McBride et al. (2019) noticed higher solubility and phytoavailability of cadmium and zinc prior the application of organic acids only (i.e., without EDTA) but there was no enhancement in uptake of any metal by *Phytolacca americana*. They elaborated that this reduced uptake might be due to presence of competent metals (copper and manganese) in the medium or less bioavailability of resultant metal complexes.

16.6.7 Plant Growth Regulators

Phytohormones, generally known as plant growth regulators (PGRs), are amazing compounds that have crucial role in the life of plants and without them, plants cannot exist. The reason being, they influence every cellular process from its formation, sustenance, growth, development, division, and so on (Rostami and Azhdarpoor 2019). It has been well-established and well-understood that plant hormones protect plants against all sort of biotic and abiotic stresses that hit their lives. They are involved in signaling and absorption of metals from soil too. Hence, their role in absorption of contaminants especially that of heavy metals is also under investigation. It has been proved that exogenous application of PGRs has positive impacts on plant growth and development and to alleviate heavy metals stress and management of their toxicity by great many researchers including Zhu et al. (2012, 2013), Agami and Mohamed (2013), and Masood et al. (2016). Among PGRs, auxins, cytokinins, gibberellins, and salicylic acids have shown potential in increasing the rate/efficiency of phytoremediation in plants; while brassinosteroids have been documented to play the same role in microalgae. The extent of their effectiveness depends on the type of plant species and its physiological conditions, their concentrations used, and the environmental conditions. Their direct effect is on the growth of plant which is enhanced thus adding to biomass of plants. The efficient plant growth increases the efficiency of absorption of contaminants from their environment (Bajguz 2019).

16.6.8 Intercropping Different Plant Species

The concept of intercropping is not new. Growing different plant species in a shared place, under the same field conditions has unique consequences. This technique is different from crop rotation in which more than one different plant species are grown in the same field area one after the other. Crop rotation is said to retain soil fertility. While, intercropping involves growth of different species simultaneously, this may have role in affecting the rhizosphere. This concept of intercropping has been widely used in case of phytoremediation. Recently, in the early months of 2019, few studies have reported that intercropping hyperaccumulator plant species which can have positive influence on phytoremediation potential of such plants. Shuzhen et al. (2019) reported a hyperaccumulator plant species *Sedum plumbizincicola* which was intercropped with *Oxalis corniculata* and *Buxus sinica* in soils affected with higher Cd levels. They used organic acids as chelating substances to increase efficiency of remediation, and they obtained positive results in this regard when oxalic acid was used at a higher concentration of 11 mmol kg^{-1}. Oxalic acid increased bioavailability of Cd in the soil, which was then absorbed by these aforementioned plant species.

16.6.9 Alterations in Plant Genome

It is quite evident that few plant species are more tolerant to metal toxicity. This tolerance may rightly be attributed to the genetic characteristics. Since the genome of a living being is the backbone of life. The genes are responsible for all morphological and biochemical characteristics in a living being. So, keeping this information in view, it can be concluded that any alteration in the genome may enhance the tolerance of a plant species for specific contaminants. Two different strategies such as: introduction of mutant in plant genome and introduction of tolerant genes to plant genome can be used to improve genetic traits of metal tolerant plants.

16.6.9.1 Introduction of Mutations in Plant Genome

Recently, the role of mutations in plant genome has also been investigated. Since mutations may lead to unique results. Experimentation carried out by Navarro-León et al. (2019) is valuable in this regard as they studied the role of a gene in affecting the efficiency of phytoextraction of cadmium from contaminated soil. They used *Brassica rapa* for phytoremediation considering greater efficiency of said plant species in tolerating heavy metals. They reported that the TILLING mutants of *B. rapa* (BraA.hma4a-3) had greater tolerance for Cd as they reflected lower reduction in biomass and higher quantities of the said metal in their foliar parts. This study clearly demonstrated that mutations in selected genes can be beneficial as they enhance phytoremediation efficiency of plants.

16.6.9.2 Introduction of Tolerant Genes to Plant Genome

Plant species that are more tolerant to heavy metal toxicity are being screened for their genes. Such genes may be introduced to other plant species which are better suited for phytoremediation projects. For example, *Prosopis juliflora* has strong tolerance for heavy metals. Keeran et al. (2019) have reported that this plant can be taken as ideal for gene mining for phytoremediation. Its genes can be transferred to other species for better performance. The research efforts are continuously being made to produce metal tolerant plants yet such plants have not reached field level. Some researchers have been trying to search such tolerant or heavy metal responsive genes from different plant species. For example, Abou-Elwafa et al. (2019) performed such an analysis on 107 accessions of sorghum using a set of 181 microsatellite markers. They reported 14 phytoremediation and heavy metals tolerance QTLs in the said plant species and 19 heavy metals stress tolerance genes.

16.6.10 Electrokinetics

The use of electrokinetics and phytoremediation approaches one after the other at different time intervals can influence the rate of phytoremediation. It has been observed by some researchers that alternate application of these two methods can substantially remediate the contaminated site in shorter span of time. For example, in a study by Chang et al. (2019), circulation-enhanced electrokinetics-phytoremediation (using corn plants)-circulation-enhanced electrokinetics was applied alternately to lead-contaminated area. They reported a reduction of 63% in initial lead levels.

16.7 Advantages of Phytoremediation

Following are the advantages of using phytoremediation to combat environmental pollution (Bock et al. 2002):

- Can not only be performed in situ but ex situ.
- Low cost.
- Solar energy-driven technique (natural process).
- In situ remediation of contaminated areas and subsequent restoration of a site.
- The plants can be easily monitored.
- It is much effective in areas with lower levels of contamination (less polluted areas).
- A huge variety of contaminants can be dealt by using this single technique, just right selection of plant species is crucial to this process.
- The fertility of soil is either maintained or improved.
- Effective where most of the other methods especially mechanical ones fail to work.
- Accepted and understood easily by public.
- The possibility of recovery and reuse of metals.
- Strengthens natural ecosystem.
- Eco-friendly technology.
- It may play its role in alteration of environmental or climatic conditions of an area depending on the type of plant species and the dimensions of area under cover. For example, if a huge area is affected by radiation, the phytoremediation strategies may involve the growth of big bare land area under cultivation of one or more plant species to free that zone of radiations, thus contributing to the overall weather patterns of the region.
- Phytoremediation has proved successful in reducing the levels of explosive compounds in soil and water. Few submerged plant species have effectively reduced the levels of RDX up to 40% which was simply doubled, i.e., up to 80% after addition of microbial species in the matrix. In the same way, 5% reduction in TNT concentration was found by submerged and floating plant species.
- Contaminants of various types have been successfully controlled from damaging the environment using phytoremediation. Yet further studies are required on this aspect to establish it as a promising approach in combating environmental pollution.
- Large areas of soil/water bodies can be remediated by using plants.
- Plants can be easily monitored.
- Possibility of recovery and reuse of contaminants.
- Expensive and complicated equipment are not required.
- Social acceptance.

16.8 Disadvantages/Limitations of Phytoremediation

Following are the disadvantages or limitations of using phytoremediation:

- Phytoremediation can work effectively in shallow groundwater, soils, or sediments. The area where roots cannot penetrate will remain affected of contaminants as such.
- Higher concentrations of contaminants can be fatal to the plant as well as the consumers of those plants. For example, the phytotransformers can be fatal to small animals like snails, so special care must be taken to keep animals from such plants prior to and during phytoremediation projects.
- Slow technique as compared with other conventional methods of waste management.
- The plant species with slow growth and higher biomass can be problematic.
- Only suitable for fully or at least partially hydrophilic contaminants.
- Contaminants may reach groundwater since the plant roots may fail at some points in absorbing the contaminants.
- The characteristics of soil and climatic and/or environmental conditions of the area can influence the rate and quality of remediation.
- If algae are used for this purpose, then excessive growth of algae in the top-most water layer may block the entry of light in deeper layers of water body hence suffocating the life underneath.
- Need of special attention on safe disposal of plants after remediation of environmental components, and special care is needed to prevent food chain from contamination since plants are the primary producers.
- Input of human resource that frequently and attentively keeps an eye on growth of plants and associated procedures in process.
- High input of labor and energy.
- Needs long-term commitment, care, and continuous study until the medium is restored to its normal.
- Long time period is required for complete recovery of contaminated environmental components (at least one growing season of planted species is required).
- The deeper layers of soil cannot be remediated as plant roots cannot penetrate to such depths (generally limited to top 3 ft of soil and top 10 ft of water).
- Addition of chelating agents is needed in some cases (in order to weaken/break the bonding between soil particles and the contaminant thus enhancing their availability for absorption).
- Large land areas are needed for phytoremediation.
- In some cases, contamination may shift from one environmental component to the other.
- The contaminants that get fixed in plant body may gain entry to food chain by herbivores, pollinators, and other consumers of plants.
- Climatic or any other factors may affect the growth of different plant species.
- It has been reported by almost every study on the said aspect that chlorophyll content decreased in every plant species that was exposed to heavy metal stress.

This reduction ultimately affects total photosynthates and hence plant life. This is a severe limitation of this technique as it depends mainly on plant.

16.9 Recent Research Trends in Phytoremediation

The recent research trends involve use of nanotechnology to enhance efficiency of phytoremediation (Zhu et al. 2019). The nanomaterials may remove contaminants from environment, promote plant growth and development, and increase availability of contaminants for absorption by plant roots. But it has been observed that still this technology is under investigation for its potential miraculous uses in phytoremediation. Most commonly used nanoparticles in this regard are that of iron. This nanoparticle has been under investigation to enhance environmental remediation. It is hypothesized that the said particle can increase phytoavailability of contaminants to the plant roots. For example, in a recent research, Mokarram-Kashtiban et al. (2019) have shown that zero valent nano particles of iron had positive effects on phytoremediation while higher concentrations had the reverse effect. Research on the use of nanoparticles is scarce and needs validation through in-depth study.

The use of phytoremediation is no doubt an effective method to remediate soil. But at the same time, slow speed of the said process has been taken as a serious limitation of it. Few researchers have come up with a solution by experimenting different methods in combination with phytoremediation to see if the efficiency of remediation increases. Interestingly, the efficiency increased when electrokinetic bioremediation method was used (Kim et al. 2005). They reported that use of electrokinetic bioremediation enhanced the efficiency of remediation. This approach was appreciated in the circle of researchers and they started different experimentations that employed phytoremediation in combination with other techniques to get better results in a short time span. In one of such latest studies, a group of researchers from Taiwan used circulation-enhanced electrokinetic-phytoremediation-circulation-enhanced electrokinetic approach to speed up the remediation process (Chang et al. 2019). Their research proved successful as they reported that the contaminated soil was remediated at a faster pace. They reported that the soils in Taiwan were contaminated with lead which could be removed efficiently by using this approach.

Since the soils are usually contaminated by more than one factor, research on phytoremediation strategies that hit more than one contaminant are being conducted. For example, in a recently published research work by Huang et al. (2019), oxalic acid-activated phosphate rock and bone meal were applied to copper- and lead-contaminated soil to investigate their effect on immobilization of both these contaminants.

Following are the main research topics under investigation nowadays:

- Selection of appropriate plant species for phytoremediation.
- Optimization of conditions for phytoremediation.

- Studies on the role of chelating agents in increasing bioavailability of contaminants (with special emphasis on heavy metals).
- Studies to enhance the rate of phytoremediation with special emphasis on different factors that can influence this process.
- Studies to find out metal-specific chelating agents to increase bioavailability of metals.
- Experimentation on the use of combinations of different factors for phytoremediation, e.g., use of combinations of organic/synthetic/natural agents.
- Research on hyperaccumulator plant genes with emphasis on mechanism of hyperaccumulation.
- Introduction of hyperaccumulator genes to other plant species, i.e., formation of transgenic plants.
- Studies on combined effect of mechanical, physical, chemical, and microbial agents in addition to phytoremediation to accelerate remediation of contaminated soils.
- Intercropping of different plant species to see its effect on phytoremediation.
- Crop rotation to enhance phytoremediation rate.

16.10 Future Prospects and Recommendations

Phytoremediation is being extensively studied at the level of universities and research institutes, yet its application is still limited to few areas on the globe. Phytoremediation being a natural process can be thought of as a miraculous method of remedying different environmental components and to find out solution to chronic environmental pollution in various world regions or planet as a whole. Efforts are being made in remedying environment with special emphasis on metals (especially uranium, arsenic, lead, chromium, and cadmium), pesticides, solvents, explosives, and oils of various kinds. Further research may lead to its full acceptance and wider applications, hence dealing environmental pollution in an effective manner. This technique can be foreseen as a strong weapon in combating environmental pollution worldwide. It is recommended that a researcher must gain proper knowledge of physiological processes and molecular mechanisms along with biological and engineering strategies as it can polish the quality of phytoremediation. Moreover, field trails should be performed to find out solutions to various problems and to find answers to various questions. Interestingly, on one side, a number of plant species can play vital roles in phytoremediation; while others cannot tolerate contaminants at all, an in-depth study of different plant species must be undertaken to prepare a list (preferably a database of plant species) that can be used for specific phytoremediation projects. Such a database should contain all relevant information with information on case studies to help select a plant species for phytoremediation in an area in case it is needed urgently. This may save time, energy, and resources to a great deal. Though this technique seems promising in dealing environmental pollution, care must be taken in order to gain maximum benefits and to eliminate its negative impacts on the biotic components of environment especially human beings. So, it is suggested that the use of such plant species which may lead to allergies of various kinds especially pollen allergy should not be brought into cultivation close to residential areas as they may solve one problem and affect human population in other ways. Proper study of the contaminated area must be conducted prior to planning a phytoremediation project. Such study may include investigation of climate, environment, weather conditions, and soil and/water characteristics. Type and properties of flora and fauna belonging to contaminated area must be focused. Initial testing of soil and/or water and a full-length experiment on a selected sample out of the infected site should be carried out to see the potential or chances of success in remediating the contaminated site. Most importantly, proper study must be conducted on proper disposal or dispositioning of the resultant plants. In case, the resultant contaminated plants are burnt, the properties of air and ash must be studied to see the effect of burning on metabolic products of contaminants. Extreme care must be taken in dealing plant samples so that the human and animal population is not harmed by them. The biodegraded compounds or by-products may reach groundwater or the food chain. So in-depth research is needed not only on proper disposal of such contaminants/contaminated plants but also the metabolic processes (including all biochemical reactions) must be studied so that the ecosystem can be protected from the possible damages that may be caused by them. Preferably those plants which grow by vegetative means must be preferred for phytoremediation. This is because the sex structures including pollens may get infected by contaminants. In case plants with sexual mode of reproduction are used, the palynological studies of such plants must be carried out to see if the contaminants got accumulated in pollens or other sex structures. The human beings and animals may come in contact with such pollens thus getting hurt in one way or the other. The hydrology and soil profile of contaminated site carry great weightage in planning a project so extreme care must be taken to study these parameters first.

16.11 Conclusion

An alarming, nonstop, and uncontrolled increase in release of wastes of diverse nature in the environment has been observed due to anthropogenic activities including increased urbanization, extreme lack of awareness of hazards of environmental pollution, deforestation, industrialization, and an increase in chemical and atomic warfare. This ever-increasing pollution is becoming the cause of concern for every nation on this planet as continuing healthy and comfortable life on this planet is becoming challenging. The consequences could be harsh if the situation goes unchecked. So, this matter must be taken seriously, and quick fix to the problem must be sorted out. On one side, strategies must be adopted to minimize the entry of pollutants to the environment, and on the other, such means must be investigated that could control the situation in an environment-friendly manner. Phytoremediation seems a method of choice as it has the potential to remediate the environment in most environment-friendly way. Though the researchers across globe have made efforts in this regard, yet in-depth research is needed on various aspects with special emphasis on understanding the mechanisms of phytoremediation, physiological processes of plants, mechanisms of uptake, translocation, accumulation, and tolerance. This would be made possible if interdisciplinary research is performed involving experts from physical, chemical, and biological sciences.

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