Bioremediation of Toxic Pollutants: Features, Strategies, and Applications

Arti Yadav, Deepika Goyal, Mrinalini Prasad, Teg Bahadur Singh, Preksha Shrivastav, Akbar Ali, and Prem Kumar Dantu

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1 Introduction

Any substance which has no further economic use for living beings and exists in the environment (open fields, water or air) is supposed to be a pollutant (Megharaj et al. 2011). Due to fast population growth, urbanization, and industrialization, the amount of hazardous waste is increasing annually. The dumping of hazardous waste, namely, rubber, plastics, pesticides, heavy metals, and industrial waste, into the environment is unsafe. The harmful effects of these substances on natural or man-made resources are due to their physicochemical and biological properties which led to the pollution, and as a result the resources become unfit for use and are of concern to the environmentalists (Fulekar 2010). The foremost sources of such hazardous substances are chemical industries. Approximately 6×10^6 chemical compounds have been synthesized, with 1000 new chemicals being synthesized yearly. Almost 60,000 to 95,000 chemicals are in commercial use. According to Third World Network Reports, more than one billion pounds (450 million kilograms) of toxins are released worldwide in air, water, and land (Shukla et al. 2010).

A. Yadav · D. Goyal · M. Prasad · T. B. Singh · P. Shrivastav · A. Ali · P. K. Dantu (✉)
Department of Botany, Dayalbagh Educational Institute (Deemed University),
Agra, Uttar Pradesh, India

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The chemical compounds causing ecological problems leading to environmental imbalance are of global concern now Garima and Singh (2014). The hazardous wastes from chemical industries and household sewage comprise noxious organic and inorganic chemicals containing heavy metals, high pH solvents, and salts. The oil spills and long-term use of fertilizers led to the accumulation of heavy metals in the soil and water affecting health of human beings and other organisms. After the green revolution, soil fertility as well as microbial flora and fauna was devastated due to accelerated use of chemical fertilizers and pesticides in order to increase agricultural productivity (Henis 1997).

Agricultural pollutants are classified in four broad groups, namely, fertilizers, pesticides, animal waste, and fossil fuels and its combustion products. Among them, fertilizers and animal wastes are easily recycled by nature itself; however, recycling of fuel and pesticides needs more time and energy due to their xenobiosis, recalcitrance, and potential toxicity hence are long-term persistent pollutants (Henis 1997).

There are various physical, chemical, and thermal methods adopted for the treatment of persistent toxic contaminants; however, they are not found effective to remove pollutants from all sites at satisfactory level of the Environmental Protection Acts. Further, cost of their renewal run into billions of US dollars (Roseberg 1993; Glass 2000; McIntyre 2003; Kuiper et al. 2004). Therefore, the biological approach popularly known as bioremediation received a special interest due to its low cost, high public acceptance, and eco-friendly nature. “Bioremediation” as the word itself explains is the process of treatment of environmental contaminants using living organisms. Bioremediation uses bacteria, fungi, algae, higher plants, or their enzymes to recover the environmental disturbances to its original state altered by hazardous wastes or pollutants (Glazer and Nikaido 1995).

Bioremediation is not a new process on Earth and perhaps has been there since the beginning of life (Okpokwasili 2007). As the waste is dumped into soil or water, the microbes get adopted for that changed environment, start degradation of that waste material, and use it for their own carbon and energy source. However, due to fast industrialization, the amount of hazardous waste increased beyond permissible limit; therefore, the same process has become challenging to microorganisms. Bioremediation is the most effective and promising method to resolve this environmental problem. Bioremediation can occur at the site of contamination or other places after excavation of contaminated soil or water (Sharma 2012). In bioremediation process, several technologies have been used to reduce contamination from the environment, such as bioventing, biopiling, bioaugmentation, biosparging, compositing, land farming, biopiles, bioleaching, bioreactor, composting, etc. Microbes have potential metabolic activities to remediate toxic wastes; however, the process is very slow, and satisfactory level of remediation does not take place. Hence, recent research is being focused toward molecular approaches in the development of transgenic microbes or consortia, engineered protein, metabolic engineering, whole-transcriptome profiling, and proteomics for remediation of environmental contaminants (Wood 2008). Cell surface expression of specific proteins allows the engineered microorganisms to transport, bioaccumulate, and/or detoxify heavy metals and to degrade xenobiotics (Arshad et al. 2007).

2 Salient Features of Bioremediation

- Bioremediation is a living organism (bacteria, fungi, yeasts, algae, and green plants) based natural environment cleaning technique that requires less time, cost, and efforts.
- When the pollutants are present in huge amount, microbial population increases using polluting substances as nutrient or energy source, and once the pollutants are degraded or imbibed, their population declines. The effectiveness of bioremediation depends on metabolic potential of microorganisms (Antizar-Ladislao 2010) and the environmental conditions which allows the microbial growth and activity.
- Bioremediation can occur at the site of contamination avoiding the human health risk which may be possible due to transportation of hazardous waste. It is generally carried out under aerobic condition though it is also possible under anaerobic conditions.
- Bioremediation is based on the principle of elimination, mineralization, attenuation, or transformation of toxic substances by the use of biological processes (Shannon and Unterman 1993).
- Bioremediation is the most promising method of pollution treatment than other traditional ones as it has the ability to completely abolish or render the pollutants from site of contamination.
- Conventional method may not completely destruct pollutants; rather, they only convert them into new waste such as incineration. On the other hand, bioremediation along with degradation transforms the toxic compounds to harmless products (water and harmless gases) eliminating the jeopardy of future liability related to the treatment and disposal of hazardous waste.
- Bioremediation is an eco-friendly and inexpensive technique. It does not use toxic chemicals for treatment of contaminants and hence has high environmental acceptance.
- Bioremediation process is affected by several factors such as nature of pollutants, pH and moisture content of soil, nutritional (contaminants) concentration and bioavailability, microbial density and diversity, oxygen content, temperature, and redox potential.
- Bioremediation activity can be enhanced by the addition of nutrients such as nitrogen and phosphorus.
- The microbial populations exposed to a specific pollutant (e.g., hydrocarbons) become adapted to that and develop genetic changes. When adapted microbial populations are used to treat hydrocarbon-contaminated sites, they respond very quickly (within hours) resulting in higher biodegradation rates than non-adapted ones (Leahy and Colwell 1990; Atlas and Bartha 1998).

3 Natural and Living Agents of Bioremediation

Environmental toxicants imposed on plants are a serious concern in most countries. These heavy metals imbalance the ecological harmony and cause disturbance to animals as well as to plants (Table 1). Removal of heavy metals from a contaminated site is known as remediation. Before the knowledge of microbes for remediation, there were some conventional methods which were being widely used for contaminant removal. These conventional methods include dredging (physical removal of the contaminated sediment layers), capping (covering the contaminated sediment surface with clean material, thus isolating the sediments), and incineration (waste treatment technology which involves the combustion of organic substances contained in waste materials). Nowadays, there are many strategies adopted by researchers to remediate heavy metals. There are certain biotechnological approaches that require the use of living organisms (Table 2) and cell manipulation to develop alternative and innovative methods to maintain natural environment. Living organisms that are used for remediation of contaminants from soil and water could be algae, bacteria, fungi, or plants. On the basis of types of biological organisms used for remediation processes, they are categorized as phycoremediation, bioremediation, mycoremediation, and phytoremediation (Fig. 1).

3.1 Bioremediation

Bioremediation is made up of two words: “bios” that means life and refers to living organisms and “to remediate” that means to solve a problem. Bioremediation is a biological process of the decontamination of contaminated environment. Microbes produce some enzymes which have the ability to degrade organic contaminants into nontoxicants. There are some microbes which are being widely used in remediation process as *Pseudomonas putida, Dechloromonas aromatica, Deinococcus radiodurans, Methylibium petroleiphilum,* and *Alcanivorax borkumensis.* However, there are some drawbacks/limitations in this process. One is that microbes (bacteria and fungi) do not act on a broad range of organic compounds. No organism is reported till now which can destroy a large percentage of the natural chemicals that exist. Another hurdle of bioremediation is that it takes a long period of time to act and impose its effect.

3.2 Mycoremediation

Besides the use of bacteria, fungal species as *Aspergillus niger*, *Aureobasidium pullulans*, *Ganoderma lucidum*, and *Cladosporium resinae* are found to be capable in mycoremediation (Mani and Kumar 2014). Fungi secrete more potent enzymes

Table 1 Living organisms involved in bioremediation

Living organism	Species	Metals	References
Bacteria	*Pseudomonas veronii*	Cd, Zn, Cu	Vullo et al. (2008)
	Burkholderia species	Cd, Pb	Jiang et al. (2008)
	Bacillus sp.	Cd, Pb, Cu	Guo et al. (2010)
	Kocuria flava	Cu	Achal et al. (2011)
	Serratia marcescens	U	Kumar et al. (2011)
	Pseudomonas aeruginosa	U	Choudhary and Sar (2011)
	Bacillus cereus	Cd, Zn	Hrynkiewicz and Baum (2012)
	Halomonas sp.	Sr	Achal et al. (2012b)
	Sporosarcina ginsengisoli	As	Achal et al. (2012a)
Fungi	*Penicillium canescens*	Cr	Say et al. (2003)
	Ganoderma lucidum	Ar	Loukidou et al. (2003)
	Aspergillus fumigates	Pb	Ramasamy et al. (2011)
Algae	*Chlorella pyrenoidosa*	U	Singhal et al. (2004)
	Chlorella fusca	Pb, Zn, Cd, Cr, Cu, Ni	Ahluwalia and Goyal (2007)
	Spirogyra sp.	Pb, Cu	Lee and Chang (2011)
	Spirulina sp.	Cr, Cu, Fe, Mn, Se, Zn	Mane and Bhosle (2012)
	Hydrodictyon sp.	V, As	Saunders et al. (2012)
	Oedogonium sp.	V, As	Saunders et al. (2012)
Lichen	*Cladonia rangiformis*	Pb	Ekmekyapar et al. (2012)
Plants	*Pteris vittata*	Cu, Ni, Zn, As	Ma et al. (2001)
	Brassica juncea	Se, Cd	Banuelos et al. (2005)
	Helianthus annuus	Cd	Mani and Kumar (2014)
	Populus sp.	Hg	Lyyra et al. (2007)
	Brassica napus	Cd	Selvam and Wong (2008)
	Typha latifolia	Pb	Tiwari et al. (2008)
	Nelumbo nucifera	Zn, Cu, Pb, Ni	Kumar et al. (2008)
	Amaranthus viridis	Cr	Liu et al. (2008)
	Helianthus annuus	Cu, Zn, Pb, Hg, As, Cd, Ni	Mani et al. (2012)
	Trifolium pratense	Cs	Wu and Tang (2009)
	Spinacea oleracea	Pb, Zn	Mani et al. (2012)
	Vetiveria zizanioides	Cd, Pb	Danh et al. (2009)
	Nicotiana tabacum	Cd	Wojas et al. (2009)
	Brassica juncea	Pb	Zarei et al. (2010)
	Pistia stratiotes	Cd, Pb, Zn	Vesely et al. (2012)
	Populus tremula	Zn, Cd, Cu	Ruiz et al. (2011)
	Gmelina arborea	Al	Dudhane et al. (2012)

Table 2 Developmental methods of bioremediation

Techniques	Principle requirement	Advantages	Disadvantages	References
Bioventing	Air (O_2) supply	Effective for treatment of petroleum-, diesel-, phenanthrene-, and hydrocarbon-contaminated sites Used in both aerobic and anaerobic bioremediation	Prolonged treatment; soil vapors of volatile compounds are formed due to high airflow rate which require additional off-gas treatment	Azubuike et al. (2016), Höhener and Ponsin (2014), Thome' et al. (2014), Frutos et al. (2010), Rayner et al. (2007), Mihopoulos et al. (2000, 2002), Shah et al. (2001), and Burgess et al. (2001)
Biopiling	Treatment bed, aeration system, nutrient or irrigation system, and leachate collection system	It transforms contaminants to carbon dioxide and water and completed in 3 to 6 months	Forced aeration system and excessive heating of air causes drying of contaminated soil resulting to inhibition of microbial activities which in turn promotes volatilization rather than biodegradation	Azubuike et al. (2016), Garima and Singh (2014), Niu et al. (2009), Wu and Crapper (2009), and Sanscartier et al. (2009)
Bioaugmentation	Metabolically more active microorganism	Natural attenuation processes Treats soil and water contaminated with chlorinated ethane such as tetrachloroethylene and trichloroethylene pollutants transformed to harmless ethylene and chloride	System that is difficult to monitor	Plangklang and Alissara (2010); Sei et al. (2001) and Bouwer and Zehnder (1993)
Biosparging	Air (O_2) supply under high pressure	Readily available, easy to install, noninvasive to operation site, and completed in short time Used for treatment of petroleum products like gasoline, diesel fuel, jet fuel, and kerosene Effectively treats benzene-, toluene-, ethylbenzene- and xylene (BTEX)-contaminated groundwater	It has environmental constraints as it requires uniform air sparging, permeable soil and unconfined aquifer, etc. No field and laboratory data available to support design consideration. Prediction of airflow direction is difficult	Azubuike et al. (2016), Garima and Singh (2014), Hajabbasi et al. (2011); Lambert et al. (2009), Kao et al. (2008), and Philp and Atlas (2005)

Compositing	Elevated temperature (55 to 65 °C°*C*) Straw, alfalfa, manure, agricultural waste, and wood chips used as bulking agents and supplementary carbon source	More convenient and inexpensive, self-heating by microorganisms useful to treat hydrocarbon-polluted soils, to make plants healthier, it is good alternative to land filling or incinerating practical	Extended treatment time (incubation periods range from months to years). Requires nitrogen supplementation. Requires periodic turning of piled polluted soil for effective treatment	Garima and Singh (2014), Blanca et al. (2007, 2008), and Barr (2002)
Land farming	Tillage (for aeration), nutrients, and irrigation Excavated polluted soil is sandwiched between clean soil (at bottom) and clay and/or concrete soil (at upper most layer)	Simple, self-heating, and cost-effective Requires less energy and has no negative impact on the environment Operative to any climate and site Useful to treat diesel- and hydrocarbon-polluted (polyaromatic hydrocarbons) site	Large operating space requirement, slow degradation rates, and requires long incubation periods, additional cost due to excavation and reduced efficacy in inorganic pollutant removal, not suitable for toxic volatiles due to its design and mechanism of pollutant removal (volatilization), especially in tropical climate regions, less efficient	Garima and Singh (2014), Cerqueira et al. (2014), Silva-Castro et al. (2015, 2012), Besaltatpour et al. (2011), Paudyn et al. (2008), Blanca et al. (2007, 2008), Khan et al. (2004), and Maila and Colete (2004)
Biopiles	Aboveground pilling of excavated polluted soil, aeration, irrigation, nutrient, leachate collection system, and treatment bed	Economic, requires less space, reduces volatilization of low molecular weight pollutants, operative to extreme cold climate and clay and sandy soils, used to treat total petroleum hydrocarbon, applicable to agriculture to municipal waste	Mass transfer problem, bioavailability limitation, robust engineering, maintenance and operation cost, lack of power supply at remote areas, need to control abiotic loss, excessive air heating causes soil drying which affects microbial activity, promotes volatilization rather than biodegradation	Dias et al. (2015), Whelan et al. (2015), Gomez and Sartaj (2014), Akbari and Ghoshal (2014), Chemlal et al. (2013), Sanscartier et al. (2009), and Blanca et al. (2007, 2008)

(continued)

Table 2 (continued)

Techniques	Principle requirement	Advantages	Disadvantages	References
Slurry bioreactors	Bioaugmenting or biostimulating agents (sewage sludge)	Fast degradation kinetics, optimized environmental parameters, toxicity of amendments Applicable for crude oil (total petroleum and polyaromatic hydrocarbons)-polluted sediment and soil polluted with volatile organic compounds such as BTEX (benzene, toluene, ethylbenzene, and xylene)	Soil requires excavation which can disrupt soil structure, due to ex situ treatment; it requires more manpower, cost inefficient due to transportation of pollutant to treatment site	Chikere et al. (2012, 2016), Zangi-Kotler et al. (2015), Mustafa et al. (2015), Plangklang and Alissara Reungsang (2010), Antizar-Ladislao et al. (2007, 2008); Arsam et al. (2007), Philp and Atlas (2005), Fuller et al. (2003)
Aqueous bioreactors	Bioaugmenting or biostimulating agents (sewage sludge)	Enhances mass transfer, effective use of inoculants and surfactant, effective to reduce toxic concentrations of contaminants. Used to treat laundry wastewater polluted with linear alkylbenzene sulfonate, BTEX-contaminated water and coal gasification wastewater	Relatively high cost capital and high operating cost; transportation may cause environmental problems	Delforno et al. (2015), Firmino et al. (2015), Xu et al. (2015), and Arsam et al. (2007)
Precipitations/ flocculations	It requires nondirected physicochemical complexation reaction between dissolved contaminants and charged cellular components (dead biomass)	Cost-effective and advantageous for removal of heavy metals	Yet to be exploited commercially	Natrajan (2008)

Microfiltrations	Microfiltration membranes and constant pressure	Remove dissolved solids rapidly, effective for wastewater treatment; more than 90% of original wastewater can be recovered and reused	Yet to be exploited commercially	Shukla et al. (2010)
Electrodialysis	Requires cation and anion exchange membrane pairs	Electrodialysis is reusable and can tolerate high temperature Useful to eliminate dissolved solids efficiently	Yet to be exploited commercially	Shukla et al. (2010)

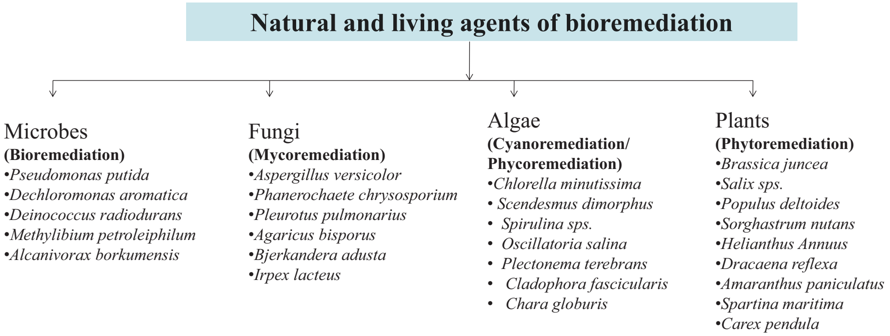

Fig. 1 Living organisms involved in bioremediation

even in nutrient-deficient conditions, and these enzymes act on a broad category of natural chemicals. Remediation through fungus may proceed faster than bacterial degradation, with hurdle suggested as the main mechanism of calcium mobilization (Gadd 2010). Many fungal species are reported to metabolize hydrocarbons, and some of them may be used in bioremediation of oil-polluted regions. These fungal genera include *Acremonium, Aspergillus, Aureobasidium, Candida, Cephalosporium, Cladosporium, Cunninghamella, Fusarium, Geotrichum, Gliocladium, Graphium, Hansenula, Mortierella*, etc. Few of the fungus, as *Trichoderma*, increases biomass of plant acting as a biocontrol agent as well as remediates agricultural waste (Pakdaman and Goltapeh 2006). *Lentinus edodes*, the gourmet mushroom, has potential of removing more than 60% of pentachlorophenol from soil (Pletsch et al. 1999). Such a potent fungus is being used as a boon in oil industries and refineries. *Phanerochaete chrysosporium* and other white-rot fungi degrade some xenobiotics as DDT and lindane (Kirk et al. 1992).

3.3 Phycoremediation or Cyanoremediation

Phycoremediation is defined as the "use of algae to treat solid wastes or wastewaters." There are few microalgae and macroalgae such as more commonly known as the seaweeds that have the ability of removing soil and water toxicants such as heavy metals, hydrocarbons, and pesticides through various mechanisms, ranging from biosorption, bioconcentration, biotransformation, to volatilization. The most common examples of microalgae are *Chlorella pyrenoidosa*, *Chlorella fusca*, *Spirogyra sp.*, *Spirulina sp.*, *Hydrodictyon sp.*, and *Oedogonium sp.* Microalgae are reported for potent remediation of pollutants from environments (Phang et al. 2015).

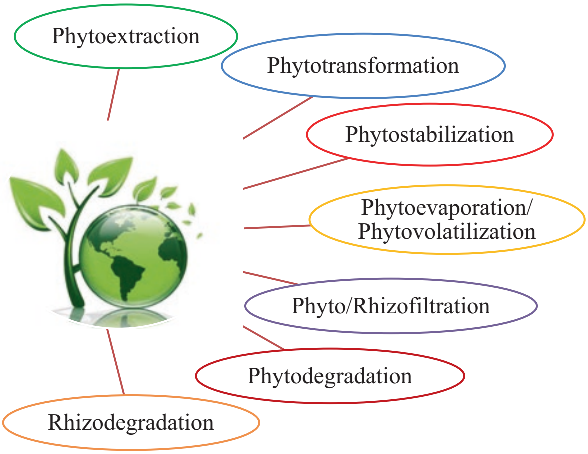

Fig. 2 Various processes used by plants for bioremediation

3.4 Phytoremediation

When plants or plant parts are involved in the removal of environmental toxicant, the process is called phytoremediation. Modern technology of phytoremediation includes phytoextraction, phytotransformation, phytostabilization, phytoevaporation or phytovolatilization, phyto−/rhizofiltration, phytodegradation, and rhizodegradation (Mahar et al. 2016), as depicted in Fig. 2.

3.4.1 Phytoextraction

Phytoextraction is a remediation process where pollutants are taken up by plant roots or algae from the contaminated soil, sediments, and/or water, and then they are accumulated in the shoots (harvestable plant biomass) (Sekara et al. 2005; Rafati et al. 2011; Razzaq 2017). Since the last two decades or so, phytoremediation technique has become more popular worldwide for extracting heavy metals from soil or water (Sulmon et al. 2007). Plants absorb pollutants from soil or water through roots and store them in root biomass or transport them up to shoot biomass or leaves. Plants continuously absorb pollutants until it is harvested. At the time of plant biomass harvesting, it was reported that plant concentrates the pollutants to much smaller volume than they were initially present in the polluted site. After the harvest, the level of pollutants is generally reduced in the soil which can be further removed through repeated process of plantation of several crops, and pollutant-free soil could become suitable for other vegetation. It was also reported that plants along with fungus (*T. atroviride*) showed more effective phytoextraction of Cd and Ni than without fungus (Cao et al. 2008). Phytoextraction is more advantageous than other traditional methods of bioremediation in several ways such as it is more eco-friendly process and prevents soil disruption or any other harm to soil quality; phytoremediation is less expensive than other cleanup processes. However, it is a more time-consuming process due to direct involvement of plants (Shukla et al. 2010).

3.4.2 Phytotransformation

In phytotransformation, complex organic molecules are converted into the simpler form through degradation or breakdown, and simple organic molecules can be retained in the plant tissues, soil, or water (Razzaq 2017). Thus, complete breakdown of the compound does not occur in phytotransformation. The complex organic pollutants such as pesticides, explosives, solvents, industrial chemicals, and other xenobiotic substances are metabolized to nontoxic forms by several plants (e.g., *Canas*), or sometimes microorganisms associated with plant roots may metabolize them in soil or water (Shukla et al. 2010). Hence, the term "green liver model" is used to explain phytotransformation, as plants behave analogously to the human liver when dealing with these xenobiotic substances (pollutant). The phytotransformation process completes in two phases of metabolism: In Phase I, the polarity of pollutants is increased by nitroreductase enzymes (Yoon et al. 2008), followed by phase II where glucose and amino acids are added to the polarized pollutants to further increase polarity (also called conjugation) (Mendez and Maier 2008); thus, the plants reduce toxicity and sequester the xenobiotics. Trinitrotoluene phytotransformation has been extensively researched, and a transformation pathway has been proposed (Vanderford et al. 1997).

3.4.3 Phytostabilization

Phytostabilization is a kind of phytoimmobilization technique in which plants are used for immobilization of soil or water pollutants (Singh 2012; Shukla et al. 2010). In this technique, pollutants are generally absorbed and accumulated in roots, adsorbed on roots, or precipitated in the rhizosphere which reduces contaminant mobility to groundwater or air, thus decreasing the bioavailability and preventing spread through the food chain (Yoon et al. 2008; Erakhrumen 2007; Ghosh 2010; Shukla et al. 2010; Wuana and Okieimen 2011). The major limitation of this technique is that it does not remove pollutants from soil or water completely, but it reduces only pollutant mobility to water stream or soil. Therefore, this technique alone is not sufficient for removal of contaminants; however, this technique can be used along with other bioremediation processes to manage the polluted sites (Vangronsveld et al. 2009; Razzaq 2017).

3.4.4 Phytovolatilization

Phytovolatilization refers to the removal of pollutants in volatile form where plants uptake water-soluble pollutants from the soil along with minerals that convert them into volatile form and then release them into atmosphere as they transpire water (Danika et al. 2005; Shukla et al. 2010; Razzaq 2017). The degree of success varies with plant as phytovolatilizers with one study showing poplar trees to volatilize up to 90% of the trichloroethylene (TCE) they absorb (Danika et al. 2005).

3.4.5 Phytofiltration or Rhizofiltration

Phytofiltration is the process where plants absorb or adsorb organic pollutants from wastewater in order to prevent its mixing with groundwater (Danika et al. 2005). Phytofiltration is slightly different in concept to phytoextraction as the former is related to the remediation of contaminated groundwater rather than polluted soils. Since plant roots are used in this technique, the term rhizofiltration is generally used in place of phytofiltration. However, rhizofiltration can be called blastofiltration when young seedlings are used or caulofiltration when excised plant shoots are used (Macek et al. 2000; Razzaq 2017). Earlier studies revealed that movement of toxic pollutants can be reduced in groundwater using this technique (Memon et al. 2001; Sakai et al. 2012). In rhizofiltration, acclimatized plants are used for remediation of contaminants (Marcia et al. 1999).

3.4.6 Rhizodegradation

Rhizodegradation refers to the degradation of organic pollutants in the soil by soil living microorganisms where the enzymatic activity of soil microbes is enhanced by plant root exudates (Razzaq 2017; Shukla et al. 2010). The plant root exudates such as sugars, alcohols, and other organic acids act as carbohydrate sources for soil microbes for enhancing their growth and activity. Few of these exudates also act as chemotactic signals for microflora. Since the biodegradation activity of soil microbes is stimulated by plant-derived exudates, the process is also called enhanced rhizosphere biodegradation, phytostimulation, and plant-assisted bioremediation (KudjoDzantor 2007).

3.4.7 Phytodegradation

It is the process by which plant-driven breakdown or degradation of toxic organic pollutants such as herbicides or trichloroethylene occurs. Degradation can take place by internal or external metabolic processes (Razzaq 2017). In external processes, complex organic compounds are hydrolyzed to simple and small units by plant enzymes. The simpler forms of contaminant can be absorbed by plants which can be incorporated and used as metabolites by the plant as it grows (Singh and Jain 2003).

4 Techniques Involved in Bioremediation

Bioremediation is broadly classified in to two groups (Fig. 3): in situ and ex situ which are further categorized into several techniques on the basis of amenability of the contaminants to biological transformation (biochemistry), availability of the pollutant to microorganisms (bioavailability), and opportunity for optimization of biological activity (bioactivity).

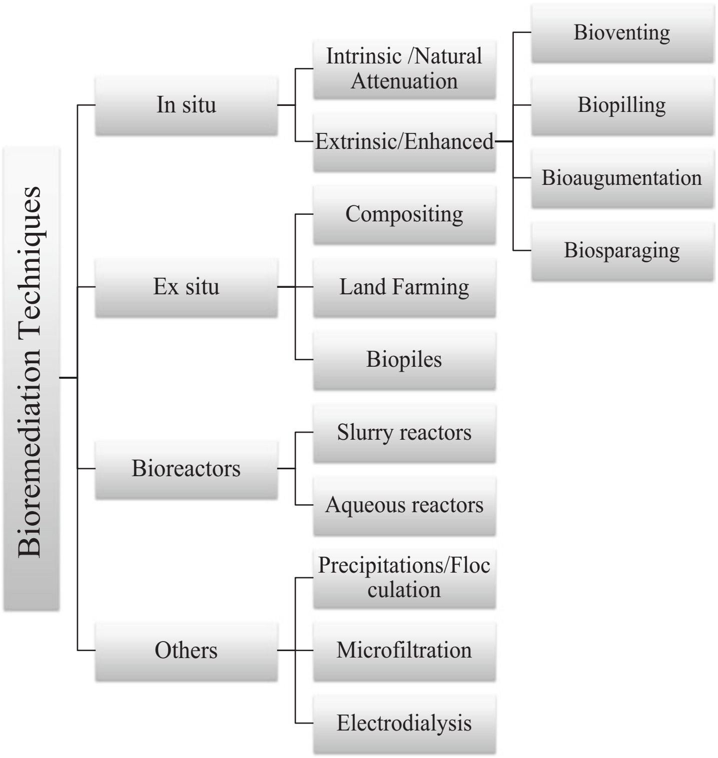

Fig. 3 Techniques of bioremediation

5 Recent Developments in Remediation Technology

Besides conventional methods as landfilling and leaching, excavation, and burial or soil washing which are time-consuming and less efficient, some advanced methods like the use of nanoparticles, nonliving biomass, and genetically modified plants are in trends for remediation Dhermendra et al. (2008). Nanoparticles are being used due to their small size and large surface area which can interact with heavy metals. High surface-to-volume ratio of nanoparticles makes them more suitable for adsorption of heavy metals. Nowadays, superparamagnetic iron oxide nanoparticles (SPION) are also being used for the separation of contaminants from soil and aquatic wastes due to their ultrafine structure and high competence and prepared iron nanoparticles for the remediation of heavy metals as Cd, Cr, Cu, and Ni. In another study, chitosan nanoparticles were formulated for the treatment of Cu from aquatic system (Yuwei and Jianlong 2011). Another approach is using nonliving biomass where no media or chemicals are required; thus, it is economical. *Cladonia rangiformis* (a nonliving lichen) is being used for the accumulation and remediation of lead from aqueous solution (Mohamad et al. 2012). Some other dead cells are also reported as *Mesorhizobium amorphae* and *Spirulina sp.* for the remediation of heavy metals as Cu and Pb, respectively (Aneja et al. 2010). Genetic engineering of plants is done to improve phytoaccumulation, phytoextraction, and phytosequestration. Recently, *Arabidopsis thaliana* was developed transgenically to increase the tolerance and accumulation of arsenic and cadmium by overexpression of AsPCS1 and YCF1 genes. These genes are derived from garlic and baker's yeast (Gaur et al. 2013). A metallothionein gene is transferred from yeast to *Nicotiana tabacum* to accumulate Cd in the roots of this transgenic plant (Krystofova et al. 2012).

6 Applications of Bioremediation

There are several advantages of bioremediation making this technique a preferred technology to remediate polluted sites:

- Bioremediation is a scientifically accepted natural process, which uses microorganisms and higher plants to remediate a wide range of organic and inorganic compounds and metabolize them to harmless products or into carbon dioxide and water.
- The complete elimination of contaminants reduces any chance of future liability associated with treatment and disposal of contaminated material.
- Microbes increase their numbers when a huge amount of contaminant is present, and once the contaminant is degraded, their population declines.
- Bioremediation can be employed on the site of contamination (in situ) without any environmental distraction. In situ bioremediation reduces the chance of environmental expose of pollutants, while transportation eliminates the threats to human health.

- On-site bioremediation with natural attenuation and fewer inputs makes it a less expensive technique for cleaning of toxic wastes (Hussain et al. 2009; Kumar et al. 2011).

7 Limitations of Bioremediation

Although bioremediation seems to be a good alternative for toxic contaminant removal, it is not fully developed/established method. Further, it requires continuous research due the involvement of microorganisms and toxic chemical compounds. Few limitations of bioremediation are as follows:

- Bioremediation is in general labor intensive and can take several months for the remediation of toxic waste to achieve at satisfactory levels.
- Bioremediation is limited to biodegradable compounds only; further complete degradation of all pollutants is not possible.
- Bioremediation involves degradation of hazardous wastes that possess a huge number of contaminants and toxicity which can inhibit the growth of microorganism or sometimes kill them.
- The pollutants which are converted to another form of chemical compound during the process of bioremediation may be more persistent or hazardous.
- Bioremediation is highly specific process that requires potentially active microorganisms, proper aeration, nutrients, irrigation, favorable pH, and temperature 20 °C to 30 °C (Vidali 2001).
- In order to enhance the activity of bacterium, fungi, or any other microorganisms, additives are supplemented which may be disruptive to other creatures inhabiting in same environment when done in situ (Vidali 2001). Thus, there is chance of more damage by bioremediation than the actual pollutant itself.
- The factors such as chemical composition, solubility, oxidation–reduction, and microbial interaction of waste likewise affect bioremediation process.
- It is time-consuming process as compared to excavation and removal of soil or incineration Kumar et al. (2011).
- It is difficult to extrapolate from bench and pilot-scale studies to full-scale field operations Hussain et al. (2009).
- Bioremediation is still a developing technology, and continuous research is needed to develop and engineer bioremediation technologies (genetically modified microorganisms) that are appropriate for sites with complex mixtures of contaminants that are not evenly dispersed in the environment Sharma (2012).
- Further, there is a problem after release of genetically engineered microorganism into environment because as time will pass it becomes difficult to remove them Garima and Singh (2014).

8 Conclusions

Organic and inorganic toxic pollutants are major problems to the environment and human health. Worldwide research on chemical contaminants helps to understand its recalcitrance and toxicity (Alcock et al. 2011). Although a variety of physical and chemical methods are used for the removal of these toxic wastes, the biological method (bioremediation) is the only one which is economic and eco-friendly technology for better and safe future (Uqab et al. 2016). A diverse group of metabolically active microorganisms are involved for in situ and ex situ bioremediation. However, response to environmental pollutants varies within a microbial guild (Ramakrishnan et al. 2010), and the presence of co-contaminants can elicit variable responses (Ramakrishnan et al. 2011). Reports reveal that nutrient supplement promotes microbial growth as well as pollutant degradation (Adams et al. 2015). Besides microorganisms, plants are also helpful to extract, degrade, transform, and store pollutants. Site characterization is the crucial step for effective bioremediation so that suitable technique (ex situ or in situ) can be employed. Geological characteristics of polluted site(s) including soil type, pollutant depth and type, site location relative to human habitation, and performance characteristics of each bioremediation technique should be incorporated in deciding the most suitable and efficient method to effectively treat polluted sites (Azubuike et al. 2016). Researchers are conducting pilot-scale bioremediation research which helps one to understand applications and limitations of this strategy. In this chapter, research finding on successful use of bioremediation to treat a variety of toxic waste has been discussed. Though bioremediation is recommended as an effective alternative for pollutant treatment, it has several practical limitations which need more research regarding soil–microbe–plant–contaminant interactions to translate effectively the bench- and pilot-scale findings to field scale (Hussain et al. 2009). However, the advantages of this technology generally compensate the disadvantages making it more reliable (Kumar et al. 2011) and have proved again and again its potential to degrade variety of pollutants (Garima and Singh 2014; Megharaj et al. 2011).

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