



# IMPACT OF AGRICULTURE ON ENVIRONMENT AND BIOREMEDIATION TECHNIQUES FOR IMPROVISATION OF CONTAMINATED SITE

Pragati Nayak<sup>1</sup> and Hitesh Solanki<sup>2</sup>

<sup>1</sup> Ph.D. Research scholar, Department of Environment science, School of Sciences, Gujarat University, Ahmedabad, Gujarat, India, Pincode- 380009; E-mail: pragatinyk@gmail.com

<sup>2</sup> Professor and Head, Department of Environment Science, University School of Sciences, Gujarat University, Navrangpura, Ahmedabad, Gujarat, India Pincode-380009  
Email:hasolanki@gujaratuniversity.ac.in

## ABSTRACT

Sustainable development requires the progress and preferment of environmental management and a persistent exploration and improvement for green technologies to treat a wide range of habitats contaminated by disorganized anthropogenic activities. Bioremediation is an excellent and effective cleaning technique to remove toxic waste from contaminated environment. Bioremediation is vastly involved in degradation, eradication, immobilization, or detoxification of diverse chemical wastes and physical hazardous materials from the surrounding through the all-embracing with the help of microorganisms and plants. Bioremediation technique is widely used to treat wastewater and to remove agricultural chemicals (pesticides and fertilizers) that leach from soil into groundwater. Certain toxic metals, such as mercury, selenium and arsenic compounds, can also be removed from water by bioremediation. Thus, bioremediation is a research and solution oriented technology that needs a prior thorough understanding of different types of available processes for improvisation and clean-up of contaminated sites. In this review, impact agricultural pollution, principle of bioremediation process and, different types of in-situ and ex-situ bioremediation techniques have been discussed.

**Keywords:** Agriculture, bioremediation, in-situ, ex-situ, phytoremediation

## INTRODUCTION

Environmental pollution is being the burning challenge of current living organisms on the earth. Pollution is the introduction of contaminants into an environment, may also defined as an unwanted alteration in the physical, chemical and biological characteristics of air, water and soil which affect human life, lives of other useful living plants and animals, industrial progress, living conditions and cultural assets (Sands, 2003). Pollution can take the form of chemical substances, or energy, such as noise, heat, or light energy. Pollutants as the main, element of pollution can be foreign substances or energies, or naturally occurring, they are considered contaminants when they exceed natural levels. Air, water and soil are being polluted differently. Soil being a “universal sink” bears the utmost burden of environmental pollution (Ashraf et al., 2014).

The different agricultural sources of soil pollutants include agrochemical sources, such as fertilizers and animal manure, and pesticides. Trace metals from these agrochemicals, such as, Cu, Cd, Pb and Hg, are also considered soil pollutants as they can disturb plant metabolism, thus crop productivity declines. Entry of pollutants directly (release of effluents on land) or indirectly (use of polluted water as irrigation to crops) has been reported to contaminate vast areas of soil resources and groundwater bodies, affecting crop production as well as human and animal health through food contamination (FAO & ITPS, 2015).

The report found that soil pollution has an adverse impact on food security in two ways – one is, it can decrease crop yields due to toxic levels of contaminants and second is, crops grown in polluted soils are unsafe for intake by animals and humans. It insisted government to reverse the damage and encourage better soil management practices to limit agricultural



pollution.

Bioremediation is an alternate, cost-effective and eco-friendly technology that provides sustainable ways to clean-up contaminated environments (Dados et al., 2015; Silva-Castro et al., 2015). Recently, a wide variety of organisms such as bacteria, fungi, algae, and plants with efficient bioremediating properties were successfully employed for effectual elimination of toxicants from the polluted sites (Vidali, 2001; Leung, 2004).

In addition, bioremediation helps to conserve soil texture and characteristics (Vidali, 2001; Adams & Guzmán-Osorio, 2008). Though bioremediation has many advantages, the success of its application depends on two major groups, namely, nature of pollutant (degree of pollution, aggregate, and oxidation state of crude oil) and environmental conditions (temperature, oxygen concentration, climatological conditions, pH, moisture content, presence of alternate carbon sources and microbes with degradation capability, soil property, and nutrient availability) (Agarry & Jimoda, 2013; Nwogu et al., 2015). Overall, bioremediation technique depends on having the right microorganisms/plants in the right place under the suitable environmental conditions in order for the degradation process to occur successfully. In order to select a clean-up technique, it is essential to understand mechanism of bioremediation processes in detail. Understanding of remedial processes supports to choose appropriate method for degradation of specific pollutant. It is required to understand that under which environmental conditions the pollutant possibly will be degraded or converted into non-toxic form. This paper provides the insight view about impact of agricultural practice on environment and different types of bioremediation techniques available for improvisation of contaminated sites. Recent bioremediation technologies and methods for removal of pollutants are also discussed.

## **EFFECT OF AGRICULTURE ON ENVIRONMENT**

Modern agricultural practices use many kinds of chemicals such as fertilizers, pesticides, cleaners, crop preservatives to produce and keep large amount of high-quality food. But every single of these chemicals has hazardous and unforeseen side-effects as like toxicity to non-target organisms which leads to ecological imbalance (Önder et al., 2011; Gangadhar et al., 2015).

Pesticides are applied to eliminate harmful insects, microorganisms and other pests which coalesce with soil, water, air and food, and contaminate the agricultural foods which in turn affect both human health and disturb the balance of ecosystem. Pesticide runoff is an important contributor to surface-water contamination (Önder et al., 2011). Additionally, fields, streams, lakes, ground water and sea altered to a kind of poison storage in time. They have the capacity of contaminating waters and plants and kill soil microorganisms as well as beneficial insects. Other harmful chemicals such as heavy metals can damage vital body organs, the immune system, and can terminate the working of nervous system. Parasites and bacteria from farm waste may also pollute water, contributing to diseases and fatality.

Excessive or wrong usage of the fertilizer which are used to improve plant growth, could be a reason for environmental pollution. Using high amounts of nitrogen fertilizer contaminates ground water, drinking water, stream and sea. This distresses the water organisms and when that kind of water is used somewhere they break the natural balance of environment. Unconscious using of phosphorus fertilizers also affects natural balance due to increasing phosphate value in water (Önder et al., 2011). Increased levels of chemical nutrients in aquatic systems, nitrogen and phosphorus, from manure and fertilizers give expansion to eutrophication when washed into nearby surface waters by rain or irrigation. Eutrophication is the dense growth of plant life and algae on the water surface and mainly leads to high occurrences of algal blooms. Eutrophication extensively depletes dissolved oxygen which can kill fish and other aquatic biota. It is also linked to increased rate of paralytic shellfish poisoning in humans, leading to death (Kremser & Schnug, 2002).

In arid and semi-arid regions, irrigation is a vital part to grow enough agricultural yield and quality. Incorrect irrigations cause environmental glitches. Rising of ground water, salinity, fertilizers and chemical additives remain to go deep with irrigation water, trace elements collect in water sources and cause soil erosion and these polluted waters make disease and are harmful on the whole living organisms and problematic for environment. In agriculture, extreme irrigation leads to high salinity and desertification of the land. It can be said, as agricultural policies affect land use, they have impact on the amount of soil erosion in



agricultural regions through changes of the economic conditions of agricultural production (Schuler & Sattler, 2010).

The persistent use of agro-chemical products degrades and destroys the soils, animals, plants, waters, and wildlife, gradually altering the ecosystems which support biodiversity. Furthermore, pesticides can kill beneficial insects, soil microorganisms, birds and some rare small species like butterflies which

have far-reaching effects on biodiversity. Since these chemicals remain in the soils for many years, their repercussions to biodiversity are massive (Gill & Garg, 2014).

Wrong soil tillage with regards to field location, soil structure and climate conditions cause soil moving with rain in other words cause erosion (Luo et al., 2010).

Agricultural applications which are without rotation due to lack of information or economic reasons entail to one-way consumption of soil plant nutrition elements, decrease to soil fertility, degradation, increasing of disease and harms in the soil and it also cause to erosion (Pellegrino et al., 2011). It is clear that stubble burning cause serious environment problems. It cause to wind and water erosion, product lose when it made uncontrolled applications, breaks the natural vegetation and makes the soil unfertile by destroying vitality on the top side of soil. For these harms on the environment, stubble burning prohibited with laws in many countries (Önder et al., 2011).

Taken together, agriculture, forestry and land-use change account for about one-fifth of global GHG emissions. Carbon dioxide emissions from agriculture are largely attributable to losses of above and below ground organic matter, through changes in land use, such as conversion of forests to pasture or cropland, and land degradation such as caused by overgrazing. The bulk of direct emissions of methane and nitrous oxide, two potent GHGs, are the result of enteric fermentation in livestock, rice production in flooded fields, and the application of nitrogen fertilizer and manure, all of which can be reduced through the implementation of better management practices to avoid consequences of climate change. (FAO, 2016).

Another issues include genetic, species and ecosystem diversity. The intensification of agriculture has directed to widespread reduction of species and habitats. Overgrazing particularly in mountain zones has led to the erosion of vegetation cover with the consequent loss of soil, the silting of rivers, etc. (Mishra, 2013).

Chemical pesticides, herbicides and agrochemicals used to control pests, diseases and weeds normally contaminate the soil and can persist for years. As a result, it gradually alters the soil microbial activities and soil chemistry, depleting soil fertility by killing soil microorganisms. Reports determine that millions of fertile soils are lost annually because of the use of synthetic fertilizers, pesticides, and herbicides combined with other farm practices (Donkova & Kaloyanova, 2008; Mahmood et al., 2016).

## **PRINCIPLES OF BIOREMEDIATION**

Bioremediation is the use of microorganisms for the degradation of hazardous chemicals in soil, sediments, water, or other contaminated materials. Often the microorganisms metabolize the chemicals to produce carbon dioxide or methane, water and biomass. Alternatively, the contaminants may be enzymatically transformed to metabolites that are less toxic or innocuous (Joutey et al., 2013; Azubuike et al., 2016). In bioremediation, living organisms, such as microorganisms (bacteria, fungi, and algae) or plants, are used to degrade and detoxify the hazardous pollutants present in the environment and convert them into CO<sub>2</sub>, H<sub>2</sub>O, microbial biomass, and metabolites (by-products which are less toxic than the parent compound) (Shah, 2014; Parte et al., 2017).

These microorganisms can be indigenous to that contaminated site or may be isolated and brought from outside to that contaminated site for bioremediation (Das & Adholeya, 2012). Microorganisms degrade and transform these pollutants through their metabolic reactions and utilize them for their growth.

Complete degradation of a pollutant needs the action of several microbes, so sometimes potential microbes can be added to the contaminated site for the effective degradation process and this process is called bioaugmentation (Tyagi et al., 2011). The biodegradation process depends on favourable environment conditions, the pollutant type and solubility, and the bioavailability of the pollutant to the microbes, thus the environmental conditions are controlled or manipulated to let sufficient microbial growth and thus fast and effective biodegradation of the pollutant (Chandra et al., 2013; Raj et al., 2014; Alin et al., 2016).



## TYPES OF BIOREMEDIATION

The bioremediation process is broadly categorized into ex situ remediation and in situ bioremediation, based on the origin, transportation, and removal of pollutants from contaminated sites (Soccol et al., 2003; Dadrasnia et al., 2013; Azubuike et al., 2016).

### **In situ bioremediation:**

In situ bioremediation is considered as the most appropriate option as it involves the treatment of pollutants in actual contaminated sites deprived of any excavation or disturbance (Vidali, 2001; Azubuike et al., 2016). It is divided into intrinsic in situ bioremediation (without any practice of enhancement) or enhanced in situ bioremediation (bioventing, biosparging, and phytoremediation). Although the in situ method is economically feasible, as there is no extra cost needed for any excavation and transportation processes (only the cost of designing and installing the equipment set-up used for the enhancement of microbial activity is required), it is less controllable and less effective than ex situ bioremediation methods. This method is used for effective treatment of sites contaminated with dyes, chlorinated solvents, heavy metals, and hydrocarbons (Kim et al., 2014).

In situ bioremediation techniques have been successfully used to treat chlorinated solvents, dyes, heavy metals, and hydrocarbons polluted sites (Frasconi et al., 2015; Roy et al., 2015). Notably, the status of electron acceptor, moisture content, nutrient availability, pH and temperature are amongst the important environmental conditions that need to be suitable for a successful in situ bioremediation to be achieved (Atlas & Philp, 2005). Some in situ bioremediation techniques might be enhanced (bioventing, bioslurping, biosparging and phytoremediation), while others might proceed without any form of enhancement (intrinsic bioremediation or natural attenuation) (Azubuike et al., 2016).

**Intrinsic in situ bioremediation:** This technique of bioremediation is also recognized as natural attenuation and it is one of the recognised in situ bioremediation methods. This method involves the unassisted and passive remediation of contaminated sites without any human interference. This technique includes both aerobic and anaerobic microbial processes for the treatment of biodegradable and recalcitrant pollutants. To form a successful and sustainable process, systematic monitoring is required in spite of the absence of any external forces in this process, therefore it is also termed as monitored natural attenuation (Azubuike et al., 2016).

### Enhanced in situ bioremediation:

This type of in situ bioremediation involves the enhancement of polluted sites by excavation or the addition of nutrients, air, and microbes to enhance microbial growth for the biodegradation process. Techniques such as bioaugmentation, biostimulation, bioslurping, biosparging, and bioventing are examples of the enhanced in situ bioremediation method (Tiwari & Singh, 2014).

**Bioaugmentation:** In this process, a group of indigenous microbes, microbes from other sites, or genetically engineered microbes are added to the contaminated site to improve the biodegradation process. This method is very efficacious when the native microorganism is unable to degrade the pollutants (Abatenh et al., 2017). This method is mostly used to treat municipal wastewater and soil contaminated with aromatic and chlorinated hydrocarbons (Nzila et al., 2016). It is also observed that non-indigenous microbes compete with indigenous ones for the growth of their population and therefore proper management of the land treatment unit is required (Sharma, 2012; Azubuike et al., 2016).

**Biostimulation:** In this method of in situ bioremediation, indigenous microbial population growth is stimulated by providing necessary nutrients in contaminated soil or groundwater for the effective treatment of pollutants. This is a beneficial method for the treatment of hydrocarbons and metal-contaminated sites (Kao et al., 2008). In most highly hydrocarbon-contaminated coastal systems, nutrient availability is the limiting factor for biodegradation (Lang, 2016; Mapelli et al., 2017).

**Bioventing:** In the bioventing bioremediation method, air (oxygen) and nutrients are supplied in a controlled way through wells to the contaminated soil to stimulate microbial activity for



the decay process. In this method, very low oxygen is given with low air-flow rates to the contaminated soil that is sufficient for effective microbial biodegradation and to minimize the volatilization and release of pollutants to the atmosphere (Atlas & Philp, 2005). This technique has been used successfully in the remediation of soils polluted by oil products (Brown et al., 2017). This technique can be effectively used for the removal of pollutants from deep below the surface (vadose zone). It is mostly used for the biodegradation of low-molecular-weight hydrocarbons, absorbed fuel residuals, spilled light petroleum, and volatile pollutants from the soil (Höhener & Ponsin, 2014).

**Bioslurping:** The technique of bioslurping combines many of processes, such as vacuum enhanced pumping, bioventing, and soil vapour extraction, for the removal of soil and groundwater pollutants using an indirect supply of oxygen and stimulating microbial biodegradation (Gidakos & Aivalioti, 2007). It is a useful way to remove products floating in the water table, also called light non-aqueous phase liquids (LNAPLs), which are low-density water-insoluble groundwater contaminants. It works like a tube that slurps LNAPLs from a tank and carries pollutants upwards. Bioventing is also used to biodegrade air and water contaminants after they are removed from the surface (Kim et al., 2014; Azubuike et al., 2016). The limitation of this technique is the low permeability of soil which reduces the oxygen transfer rate and further decreases the microbial activity. These techniques generally are fruitful for the removal of volatile and semivolatile organic contaminants from soil and liquid (Vidali, 2001). The bioslurping system should only be installed if the contaminants are at most 7m below the soil surface because the vacuum pump is inefficient in sucking LNAPLs (light non-aqueous phase liquids) at greater depth (Johnston, 2010; Azubuike et al., 2016).

**Biosparging:** Unlike the bioventing process, in the biosparging process air is supplied into the soil subsurface (i.e., saturated zone) which can cause the upward movement of volatile organic pollutants to the surface area for the biodegradation process and also augments pollutant removal from the polluted site by stimulating the microbial activity (Atlas & Philp, 2005; Waudby & Nelson, 2004). This process is facilitated by two main factors, that is, soil permeability and pollutant biodegradability (Bennett, 1999; Simpanen et al., 2016). The biosparging process has been extensively utilized for the treatment of aquifers polluted with hydrocarbons (benzene, xylene, toluene, and ethylbenzene) and petroleum products (Banerjee et al., 2016).

#### Ex situ bioremediation

As the name suggests, the ex situ bioremediation method involves the excavation and transportation of pollutants from the actual polluted site to another site and then the removal of pollutants using several bioremediation techniques (Aktaş, 2013). The ex situ bioremediation techniques are further classified into different categories, such as landfarming, composting, biopiling, bioreactors, and biofilters, based on the pollutant types, depth and degree of pollution, treatment cost, and the geographical and geological features of the polluted site (Koning et al., 2001; Atlas & Philp, 2005). Further details about these techniques are given below. After treatment with ex situ remediation techniques, the decontaminated soil could be useful for landscaping purposes (Shah, 2014; Azubuike et al., 2016; Oyetibo et al., 2016).

**Biopiling:** Biopiling or heap technique is a combination of landfarming and composting methods. The elements of this technology are watering, aeration, and leaching. Its usage is increasingly taken into consideration thanks to its construction characteristics and the favourable cost-benefit proportion that allow functional bioremediation process, provided that adequate control of nutrients, temperature and aeration is ensured. This process involves the lab testing of the contaminated soil sample to check the degradation potential, mechanical separation of the soil sample for the homogenization of the soil, piling of excavated polluted soil, additional amendment of soil with nutrients, and forced aeration to boost microbial degradation. Similarly, potent microbes could be added to the biopile for the effective remediation of the pollutant. The whole arrangement of this system includes a treatment bed and nutrient, aeration system, irrigation system buried under the soil, and a leachate collection system. The biopiling method is mostly used for treating soil contaminated with the low-molecular-weight pollutants and petroleum hydrocarbons, such as BTEX, PAHs, and



phenols, and could be effectively used in the remediation of an enormously contaminated environment such as in cold regions (Dias et al., 2015). This technique is used increasingly for bioremediation due to its cost-effectiveness, and the temperature, pH, and nutrient condition can be controlled for bioremediation (Whelan et al., 2015).

**Landfarming:** In the landfarming technique, contaminated soil is excavated, spread, and tilled from time to time over the fixed soil layer support above the ground to let the aerobic biodegradation of pollutants by autochthonous microbes. Tillage provides the aeration, nutrients, and irrigation to increase the microbial activity during landfarming (Volpe et al., 2012). It is seen that landfarming can be done only for the treatment of 10-35 cm of surface soil (USEPA, 2006). A neutral pH can be upheld using agricultural lime. While landfarming is considered to be an ex situ bioremediation method, in a rare cases it can be considered to be an in situ bioremediation method (Sanscartier et al., 2009). This technique is mostly used in the treatment of aliphatic and polycyclic aromatic hydrocarbon and PCBs contaminated sites (Silva-Castro et al., 2015). This technique is considered as the simplest and cheapest option as less equipment, low cost, and low maintenance is required for its operation, and therefore it is gaining the greatest consideration as an alternative dumping method (Carriger et al., 2006; Azubuiket al., 2016).

**Bioreactors:** In this method, usually a reactor or container is used to degrade the contaminant under controlled conditions. Engineered containment systems, such as slurry or aqueous reactors, are utilized for the treatment of contaminated solid material (soil, sludge, and sediment) and water (Vidali, 2001). There are different operating modes of bioreactor, which include: batch, fed-batch, sequencing batch, continuous and multistage. Factors such as controlled bioaugmentation, supplementation of nutrients, mass transfer, bioavailability of pollutants, and optimized reaction conditions make the bioreactor centered bioremediation technique most effective (Mohan et al., 2004). The bioreactor technique is used for the treatment of soil or water contaminated with volatile organic pollutants such as benzene, toluene, ethylbenzene, and xylene (BTEX) (Li & Goel, 2011; Azubuiket al., 2016).

**Biofilters:** In this technique, columns embedded with microbes are used for the removal of gaseous pollutants (Boopathy, 2000). Before the treated waste stream can be discharged, it must be clarified so that the number of present microorganisms in the discharge meets regulations. It is used to treat contaminated gas streams. For this form of treatment to be effective, the contaminant must be volatile (Frazar, 2000).

**Phytoremediation:** Phytoremediation is an emerging bioremediation techniques that utilizes plants and their roots for the remediation of contaminated soil and water (Tangahu et al., 2011). Most of the research studies have reported that, at the contaminated site, the pollutant removal mechanism by plants involves the uptake of pollutants by a passive process, translocation of the pollutant from root to shoot by xylem flow, and accumulation in the shoot (San Miguel et al., 2013). Vegetation-based bioremediation methods have enormous potential to degrade, accumulate, immobilize, and transform the persistent contaminants by acting as a biofilter and metabolizing the pollutant (Ismail, 2012). It is considered to be an innovative and cost-effective alternative bioremediation method to treat unsafe contaminated sites. The phytoremediation method involves various techniques depending on the pollutant type, such as elemental (toxic heavy metals and radionuclides) and organic (hydrocarbons and chlorinated compounds), and their fate (accumulation, degradation, stabilization, volatilization, transformation, filtration, and the combination of these processes) (Dhankher et al., 2012; Malik et al., 2016).

Depending on the kind of pollutant, it occurs through different mechanisms, namely biodegradation, vaporization, filtration, among others. Elemental contaminants like heavy metals or radioactive elements are mainly extracted, transformed and sequestered, while organic contaminants are eliminated mainly through rhizodegradation, biodegradation, vaporization or stabilization (Kuiper et al., 2004; Azubuiket al., 2016).

**Phytoextraction:** This mechanism involves removal of the contaminant from the soil and its accumulation in various parts of the plant (e.g., root, stem or leaf) (Dhankher et al., 2012). It refers to the ability of plants to accumulate pollutants in their aboveground tissue, which is

then harvested to eliminate the contaminants (McIntosh, 2014). Plants may accumulate pollutants into the roots, shoots, and leaves and produce a mass of plants containing pollutants (metals) that are further transported for disposal and recycling (Meagher, 2000). This technique is also known as the phytoaccumulation or rhizoaccumulation method of phytoremediation.

- **Phytotransformation or Phytodegradation:** After the absorption of the contaminant by the plant, it is transformed into a less toxic form. Such a transformation can also occur on the plant's own surface (Parket al., 2011). The phytodegradation of organic compounds can take place inside the plant or within the rhizosphere of the plant (Pivetz, 2001; Newman & Reynolds, 2004).

- **Phytovolatilization:** It refers to the plants' ability to uptake contaminants that are water soluble and eradicate them through transpiration of water into the atmosphere (Batty & Anslow, 2008; Ferro et al., 2013; Masciandaro et al., 2013). The use of this technology works with low molecular weight pollutants in the soil such as single-chain hydrocarbons that are more soluble (Gerhardt et al., 2009).

- **Phytostimulation:** Plant roots release substances (exudates) that can accelerate degradation of certain pollutants. When microorganisms contribute to degradation, the whole process is called phytostimulation (Dhankher et al., 2012). Rhizodegradation, sometimes called phytostimulation, rhizosphere biodegradation or plant assisted bioremediation, is the enhanced breakdown of a contaminant by increasing the bioactivity using the plant rhizosphere environment to stimulate the microbial populations (Prasad & Prasad, 2012; Patil et al., 2013). This method is useful in removing organic contaminants, such as pesticides, aromatics, and polynuclear aromatic hydrocarbons (PAHs), from soil and sediments. Chlorinated solvents also have been targeted at demonstration sites (Miller, 1996).

- **Phytostabilization:** It is a stabilizing process for contaminated soils and sediments in place using plants, thus preventing the lateral or vertical migration of toxic metals by leaching. This technique is best applicable in phytostabilizing metal contaminants of waste landfill sites (e.g. Pb, Cd, Zn, As, Cu, Cr, Se, and U) where the best option is to immobilize them in situ (Faerber & Herzog, 2010).

- **Rhizofiltration:** The technique is a form of phytoremediation which uses plant roots to remove target species from water. Through absorption and eventual sequestering of specified contaminants in the leaves or roots, toxic solutes can be easily and efficiently removed from water making it suitable for consumption. Specific removal of heavy metals such as  $\text{Cu}^{2+}$ ,  $\text{Cd}^{2+}$ ,  $\text{Cr}^{6+}$ ,  $\text{Ni}^{2+}$ ,  $\text{Pb}^{2+}$ , and  $\text{Zn}^{2+}$  have already been demonstrated in current research (Dushenkov et al., 1995).

## CONCLUSION

Biodegradation is a very productive and environmentally friendly option to remediate, clean, manage and recover a contaminated ecosystem. A working mechanism of bioremediation is to reduce, detoxify, degrade, mineralize or transform more toxic pollutants to a less toxic form. It is obvious that bioremediation can treat a wide range of compounds. The leading step to a successful bioremediation is site description, which helps create the most appropriate and promising bioremediation technique (ex-situ or in-situ). Ex-situ bioremediation techniques tend to be more expensive due to excavation and transportation from an archaeological site. The outcome of each degradation process depends on microbial (biomass concentration, population diversity, enzyme activities), substrate (physico-chemical characteristics, molecular structure, and concentration), and a range of environmental factors (pH, temperature, moisture content, EC, availability of electron acceptors and carbon and energy sources). It is important to examine geological characteristics of polluted sites like soil, pollutant type and depth, human habitation site for selection of the most appropriate and operative bioremediation technique to get better results and successful treatment of polluted sites. To increase the effectiveness of remediation the combined use of multiple techniques should also be considered, which makes it possible to



minimize the individual disadvantages of each of the techniques. Studies and planning must be conducted before implementing it on a large scale which helps to select the most suitable technique of bioremediation technique.

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