

Phytoremediation approaches to PAH contaminated soil

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Abstract

Petroleum based products are the current major source of energy and their transport across the world frequently result in oil spillage, contaminating the soil and water alike. There is an overwhelming need for in situ technologies that can efficiently remediate persistent contaminants from soil in a cost-effective, environmentally friendly method. In phytoremediation, plants are utilized to clean up contaminated soil, taking advantage of their natural ability to take up, accumulate and/or degrade constituents of their soil and water. Phytoremediation processes and techniques suggest their applicability to a broad range of contaminants including various organic compounds such as chlorinated solvents, BTEX (Benzene, toluene, ethylene & xylene), polycyclic aromatic hydrocarbons (PAHs), nutrients and surfactants offering a low-cost method of soil remediation technology. Chemical nature of organic contaminants, sources of contamination, microbial processes associated with plants and role of plants in PAH contaminated soil reclamation have been discussed to decipher the strategies to accomplish remediation to a large extent over short durations of exposure based on multi-process integration approaches.

Keywords: Phytoremediation, pollution, polycyclic aromatic hydrocarbons, oil spillage.

Introduction

Petroleum and its products are of specific concern in pollution studies due to their structural complexity, slow biodegradability, bio-magnification potential and above all the serious health hazards associated with their release into the environment. Plants are used successfully in the remediation of a wide range of contaminated soils, mainly due to their 2 way functioning-creating favorable conditions for microbial degradation and roots assessing the contaminants in the soil.

Nature & types of organic contaminants: Organic pollutants differ in charge and solubility. Non-polar compounds such as hydrocarbons are poorly soluble in water and sorbs readily to hydrophobic soil particles like soil organic matter. Organic contaminants include petroleum hydrocarbons, often introduced into the environment via the oil and gas explorations. Crude oil is a complex mixture of aliphatic, aromatic, heterocyclic and asphaltene/tar hydrocarbons, ranging in size from C6 to > C50. Total petroleum hydrocarbons (TPHs) are classified into different fractions. Fractions 1 (C6-C10) and 2 (C10-C16) are volatile or semi-volatile, whereas fractions 3 (C16-C33) and 4 (C34-C50) are hydrophobic and recalcitrant. Compounds from fractions 3 and 4 can be highly toxic and are regulated due to their mutagenicity and carcinogenicity. These hydrocarbons can be degraded by various processes, including photo oxidation, microbial action and natural rhizosphere action (Greenberg et al., 2007).

Petroleum or crude oil, a naturally occurring flammable liquid found in rock formations in Earth's crust consists of a complex mixture of hydrocarbons of various molecular weights and organic compounds. The proportion of these hydrocarbons is highly variable and ranges from as much as 97% by weight in the lighter oils

to as little as 50% in the heavier oils and bitumens. The hydrocarbons in crude oil are mostly alkanes (C_nH_{2n+2}), cycloalkanes or naphthenes (C_nH_{2n}) in addition to various aromatic hydrocarbons or asphaltenes (C_nH_n) while the other organic compounds contain nitrogen, oxygen and sulfur along with trace amounts of metals such as iron, nickel, copper and vanadium. The most common products of petroleum distillations are fuels which include: ethane and other short-chain alkanes, diesel fuel (petrodiesel), fuel oils, gasoline (petrol), jet fuel, kerosene and liquefied petroleum gas (LPG). Certain types of resultant hydrocarbons may be mixed with other nonhydrocarbons, to create other end products like alkenes (olefins) which can be manufactured into plastics or other compounds, lubricants which produce light machine oils, motor oils and greases, wax (used in the packaging of frozen foods), sulfur or sulfuric acid useful as industrial materials, petroleum coke used in solid fuel, paraffin wax etc. Bioremediation of oil components is restricted by their volatility, toxicity and bioavailability; which in turn depends on the length of the carbon chain. Oil components with a short length carbon chain can easily evaporate; whereas molecules with longer carbon chains or complicated (eg. branched or cycle) molecular structures persist in the soil due to less susceptibility to degradability, extractability and other related processes in a time-dependent manner.

Common sites of PAH contaminants: The typical contaminated sites are: unregulated former disposal sites (areas associated with spills, leaks, open storage areas); filling stations; electrical facilities, leaks and spills, firefighter training areas, ports and waterways where past industrial discharges contaminated sediment; lagoons used to store or "treat" industrial effluents, mine tailings; ponds, above and below ground fuel storage sites;

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municipal landfills; military training areas; abandoned buildings with inventory of fuel, transportation routes where spills or spraying of oil or chemicals have occurred and wood preserving sites (Site remediation technologies, 1997). Petroleum is also the raw material for many chemical products such as plastics, paints and cosmetics.

Phytoremediation: Phytoremediation is the green technology that uses plants to remediate contaminated soil, sediment and surface water. Pradhan et al 1998) \lt is a cost-effective, ecologically compatible tool for the environmental clean-up of a wide array of contaminants such as petroleum hydrocarbons, chlorinated solvents, pesticides and metals (Anderson et al., 1993; Cunningham & Lee 1995; Schwab et al., 1995). Plants preserve the natural structure and texture of soil using solar energy and hence this technology is suitable to diverse regions and climates.

Phytoremediation process

(a) Uptake of organic compounds from soil and water; (b) Accumulation or processing of these chemicals via lignification, volatilization, metabolization, mineralization; (c) Use of enzymes to break down complex organic molecules into simpler molecules (ultimately carbon dioxide & water) and (d) Increase the carbon and oxygen content of soil around roots (and so promote microbial/fungal activity) and decay of root tissues.

Phytoremediation techniques

There are 5 basic phytoremediation techniques:

- 1) Rhizofiltration, a water remediation technique involving the uptake of contaminants by roots of plants.
- 2) Phytoextraction, a technique involving uptake of contaminants from soil. 3) Phytotransformation, applicable to both soil and water and involves the degradation of contaminants through plant metabolism. 4) Phyto-stimulation, (plant-assisted bioremediation) used for both soil and water and involves stimulation of microbial biodegradation at the root zone. 5) Phytostabilization, using plants to reduce the mobility and migration potential of contaminants in soil (Ralinda & Miller, 1996).

Metabolism inside plants

Organic pollutants cause hindrance to uptake of compound in a plant as they are lipophilic and covalently bound to the soil components where log Kow is important for transport of substances in the transpiration stream. After reaching the plant rhizosphere contaminants migrate into the root, where lipophilic compounds either limit the uptake or cause accumulation in the partly suberised root cortex. Before the pollutant is translocated by the transpiration stream in the xylem into other tissues of the plant (root & shoot), it has to enter the symplast to overcome the barriers of suberised casparian strips in the root endodermis where detoxification and metabolism occur (Schroder *et al.*, 2002). P-450 enzymes catalyse phase-I transformation reactions like hydroxylation, sulfoxidation and N and O-dealkylation and Glutathione-

S-transferases are responsible for conjugation reations which play a central role in detoxification in plants. Phase-III of plant xenobiotics metabolism involves storage and compartmentation of soluble conjugates in the cell wall (Komossa *et al.*, 1995). Transformation of contaminants may occur outside the plant in the rhizosphere, inside the plant or sorbed to the leaf surface (photolysis) (Trapp & Karlson, 2001).

Microbial processes associated with plants: Plant enzymes aid in the degradation of contaminants during phytoremediation, but during natural attenuation or bioaugmentation, indigenous microbial population performs the degradation. However the hydrocarbon assemblage is resistant to soil micro-organisms under normal conditions and persist in the subsurface for decades. Since plant roots can supply readily available carbon sources for microorganisms they influence the soil microbial community by increasing microbial numbers, improve physical and chemical conditions of soil, increase humification and adsorption of pollutants onto the rhizosphere. Since different plants have varying effects on microorganisms, their degradation rate is dependent on individual composition of plant exudates (Merkl et al., 2005).

Filamentous fungi of the genera Fusarium, Penicillium. Trichoderma. Aspergillus, Neosartorya. Pseudallescheria, Cladosporium, Pestalotiopsis, Phoma and Paecillomyces have the ability to synthesize extracellular oxidative enzymes which can relate lignin degrading enzyme systems with crude oil contaminants (Naranjo et al., 2007). White-rot fungi contain all three lignin-modifying enzymes (LMEs): lignin peroxidase, manganese dependent peroxidase, and laccase enzymes and hence are able to breakdown and mineralize several environmental pollutants into nontoxic forms. Through intensive study of lignolytic fungi, it has been established that these organisms produce extracellular enzymes with very low substrate specificity. Petroleum hydrocarbon pollution severely effects the species composition and density of indigenous microalgae in polluted soil (Megharaj et al., 2000). D'Annibale et al. (2006) explain that external nutritional supply like saw dust, fertilizers or plant debris is always required in conjunction with fungus unlike bacteria. He isolated nine fungal strains from an aged and heavily contaminated soil, identified and screened to assess their degradative potential. Among them, Allescheriella sp. strain DABAC 1, Stachybotrys sp. Strain DABAC 3, and Phlebia sp. strain DABAC 9 were selected for remediation trials on the basis of Poly R-478 decolourization associated with lignin-modifying enzyme (LME) production. These autochthonous fungi were tested for the ability to grow under nonsterile conditions and to degrade various aromatic hydrocarbons in the same contaminated soil. The autochthonous fungi made a significant decrease in soil toxicity, as assessed by both the Lepidium sativum L. germination test and the Collembola mortality test. The large quantity of residual



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Common name of plant	Scientific name of plant	Research findings	References
Cocks foot Tall fescue Red fescue Rye grass Birdsfoot-trefoil Red clover White clover	Dactylis glomerata Festuca arundinacea Festuca rubra Lolium perenne Lotus carniculatus Trifolium pretense Trifolium repens	Naphthalene decreased to about 20% & other PAHs decreased with an increase in molecular weight, except with pyrene, the only PAH not to show any significant decrease.	Smith <i>et al.,</i> 2006
Cat claw	Mimosa monancistra	Dissipation of benzo[a]pyrene significantly faster in vegetated soil.	Bernal <i>et al.,</i> 2007
Baby bear	Cucurbita pepo ssp pepo	As soil moisture, plant density increased rates of contaminant accumulation by plant roots were increased. Concentrations of the compound in plant roots were inversely related to plant age whereas no change in the bioavailability of the compound was observed in successive generations of plants grown in the same contaminated soil.	Kelsey <i>et al.,</i> 2006
Annual ryegrass Tall Fescue Yellow Sweet Clover	Lolium multiflorum Festuca arundinacea Schreb. Melilotus officinalis Lam.	Maturity of plant root contributes to reduction in the bioavailability of target PAHs.	Parrish <i>et al.,</i> 2005
Sunflower Bermuda grass Southern crabgrass	Helianthus annuus L. Cynodon dactylon L. Digitaria ciliaris (Retz.)Koeler.	Vegetation increases total numbers of beneficial fungi and bacteria in contaminated soil.	Olson & Fletcher, 2000
Maize	Zea mays L.	Increase in hydrocarbon bioavailability, stimulates bacterial population.	Radwan <i>et al.,</i> 199 Chaineau <i>et al.,</i> 2000
Switch grass Little bluestem grass Alfalfa	Panicum virgatum Schizachyrium scoparium Medicago sativa L.	Reduction in total PAH concentration after six months of treatment.	Pradhan <i>et al.,</i> 1998 Wiltse <i>et al.,</i> 1998
Rice Naked Spinach Devil's beggartick	Oryza sativa L.cv. Spinacia oleracea L.cv.Ohrai. Pueraria lobata (Willd.) Ohwi Bidens frondosa L.	Significant decrease in TPH concentration under vegetated conditions.	Kaimi <i>et al.,</i> 2007
Slender oat	Avena barbata	A large phenanthrene degrader population in rhizosphere is related to root debris & soil exudates.	Miya & Firestone, 2001
Alfalfa Alpine blue grass	Medicago sativa L. Poa alpine L.	Stimulation of bioremediation around plant roots due to higher number of organic chemical degraders indicates potential.	Nichols et al., 199
Tall fescue	Festuca arundinacea	Greater total bacterial numbers & PAH-degrading bacteria in rhizosphere soil.	Ho & Banks, 2006
Alflalfa Reed	Medicago sativa L. Phragmitis australis	Rhizosphere microflora of alfalfa was less inhibited by hydrocarbon contamination with higher degradative potential compared to reed.	Muratova <i>et al.,</i> 2003

oil in microcosms with plants and fungi may suggest, however, that plants were able to take up greater quantities of nutrients with the assistance of fungi, intensifying competition between plants and bacteria (Vavrek *et al.*, 2001).

Based on the hypothesis that root exudates not only increase the rhizosphere microbial numbers but also community composition. Binet *et al.* (2000) investigated the significance of plant microbial interactions in the degradation of PAH. Dead and decaying roots (result of normal root turnover) produce air filled macro-pores in

soil which facilitate oxygen diffusion in the subsurface. As a result biodegradation of many organic chemical contaminants is stimulated by plants. Certain organic chemicals can be passively taken up through the roots and transported in the plant's vascular system (Ferro *et al.*, 1999). More efficient degradation of contaminant is reported in phytoremediation systems when plants associated themselves with endophytes. Plant endophyte partnerships are beneficial to improve phytoremediation of a mixture of contaminants (Weyens *et al.*, 2009).



Table 2. Studies on phytoremediation with spent oil contaminants in soil.

Common name of plant	Scientific name of plant	Research findings	References
Smooth brome grass Wheat Flax	Medicago truncatular Bromus mermis Secale cereale Triticum aestivum Linum ussitasimum Agropyron desertorum	Reduction in germination, above ground height & biomass for all species.	Sharifi <i>et al.,</i> 2007
Chilly Tomato	Capsicum annum L. Lycopersicum esculentum	Germination percentage, mean height, mean leaf area reduced. Premature death with all strengths of oil.	Anoliefo & Vwioko,1995
Castor	Ricinus communis L.	Stimulation of growth parameters- plant height, stem girth, leaf area, fresh and dry weights, and root length.	Vwioko & Fashemi, 2005
Slim amaranth	Amaranthus hybridus L.	Decrease in the total chlorophyll and protein levels.	Odjegba & Sadiq, 2002
Cowpea	Vigna ungiculata L. Walp)	Reduction in germination time, increase in germination percentage, plant height, leaf number and total biomass.	Ataga & Adedokun, 2007

Wolf et al. (2007) attribute rhizosphere effect to the very characteristics of the roots. The contribution of the rhizosphere microbes in the degradation of pollutants is referred to as rhizoremediation (Anderson et al., 1993; Schwab et al., 1995). However when trees establish rhizosphere colonization, the hydrocarbons become amenable to biodegradation (Phillips, 2004). This emerging technology has been reviewed by a number of authors (Salt et al., 1998; Schnoor 2002; Joner & Leyval, 2003a, b; Suresh & Ravi Shankar, 2004; Okoh, 2006). Nichols et al. (1997) and Radwan et al. (1998) demonstrated in hydrocarbon-degrading increase microorganisms under the influence of "rhizosphere effect" in the root zone of the plants which are utilized in the phytoremediation of hydrocarbon-contaminated soil.

Siciliano *et al.* (2003) demonstrated that phytoremediation systems that substantially decrease aged Total Petroleum Hydrocarbons (TPH) in soil do so mainly by increasing bacteria and that bacteria contain a bulk of hydrocarbon catabolic genes. However, promotion of catabolic activity by plants is strongly influenced by site factors that include available nutrients. Their results suggest that phytoremediation systems are better optional methods for increasing the potential of soil to degrade hydrocarbons.

Role of plants in soil reclamation: Though annual, biennial and perennial plant species were found during the survey of contaminated sites, annuals and biennials would be less useful for phytoremediation than perennial species since they require low management inputs (Godwin & Thorpe, 2000). When benzene and octane were absorbed through root cells there were concentration dependent variations in the level of sub cellular organelles structure in alfalfa and rye grass root cells. Destruction in cell homoeostasis was observed at

the basic metabolism enzyme levels in annual and perennial plants (Sadunishvili *et al.*, 2009). The role of plants in remediation of PAHs, spent oil and crude oil contamination is presented in Table 1-3.

Aprill and Sims (1990) established a mix of eight grasses in sandy loamy soil to demonstrate whether degradation of four PAH (benzo[a]pyrene, benzo[a]anthracene, dibenzo [a, h] anthracene & chrysene) was stimulated by plant growth. The eight grasses included Andropogon gerardii, Schizachyrium scoparium, Sorghastrum nutans, Panicum virgatum, **Elymus** canadensis

glaucifolius, Bouteloua curtipendula, Bouteloua gracilis and Pascopyrum smithii. Vicia faba plants (broad beans) are suggested as tools for the phytoremediation of oily desert areas because these can grow in poor soils and tolerate crude oil concentrations up to 10 % w/w (Radwan et al., 1998). Their rhizospheres are rich in hydrocarbon utilizing microorganisms which could affect oil attenuation in sand (Radwan et al., 2000).

Alfalfa recorded an increase in the number of both culturable. aerobic heterotrophic and petroleum degrading bacteria in the rhizosphere by 7 weeks, though its rhizosphere effect was not as substantial as perennial ryegrass due to varying root morphologies. Lolium perenne seemed to support an increase in microbial numbers as the rhizosphere effects were similar for both heterotrophic bacteria and petroleum degrading bacteria. In addition there was a large increase in hexadecane degrading bacteria (Kirk et al., 2005). Muratova et al. (2003) observed increase in total microbial numbers and high rate of PAH degradation in PAH-contaminated soil which was planted with *Medicago sativa L.* and reeds when compared with unplanted soil. Mangroves found in the areas with most industrial and shipping activity are subjected to the effects of PAH contamination. There are limited reports on phytoremediation of organic pollutants affected by mycorrhiza during the last decade. Most of these studies were concentrated on ectomycorrhizal fungi in pure culture, rather than in symbiosis with plants (Joner & Leyval, 2003b). A number of changes take place in soil in the presence of roots which could be mistaken as changes in its chemical characteristics, modified microbial composition and enhanced microbial activity. It has thus been demonstrated that ectomycorrhizal fungi may degrade several compounds such as 2,4-

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Table 3: Studies on	nhytoreme	diation with	crude oil	contaminants	in soil
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Common nama	†	diation with crude oil contaminants in soil	References
	Common name Scientific name of plant Research findings		
Alfalfa	Medicago sativa L.	Light crude oil at 1%-10% prevents normal growth and germination of Alfalfa in soil.	Dariush Minai-Tehrani <i>et al</i> ., 2007
Maize	Zea mays Psoralea esculenta Pursh.	Inhibited germination and growth at high concentrations.	Ogboghodo et al., 2004
Prairie turnip Crested Wheat Grass Yellow sweet clover White clover Timothy Prairie sagewort Pennsylvania Cinquefoil	Lathyrus venosus Muhl. Agropyron pectiniforme R. & S. Melilotus officinalis (L.) Lam. Trifolium repens L. Phleum pretense L. Artemisia frigida Willd. Potentilla pensylvanica L.	Lowest seedling mass in contaminated soil.	Robson <i>et al.</i> , 2004
Guinea corn	Sorghum bicolor	Inhibition of germination and growth in guinea corn, serves as a bio-indicator of crude oil polluted areas.	Akaninwor <i>et</i> al., 2007
Maize Alfalfa Ryegrass Wheat Soybean Winter vetch	Zea mays Medicago sativa Lolium perenne Triticum aestivum Glycine max Vicia villosa	Z. mays and G. max seedlings possess greater potential to enhance remediation based upon percent emergence and plant biomass production in contaminated soil.	Issoufi <i>et al.</i> , 2006
Bahia grass Dallis grass Pearl millet Centipede grass Brown top millet Weeping love grass	Digitaria sanguinalis (L.) Scop Paspalum notatum Flugge P. dilatatum Poir. Pennisetum glaucum (L.)R.Br. Eremochloa ophiuroides (Munro) Hack). Brachiaria ramosa L.	Optimum Nitrogen application increased plant root growth and extends rhizosphere potential. Nitrogen requirement for growth varies based on plant species.	Kirkpatrick <i>et</i> al., 2006
Sudan grass	Eragrostis curvula (Schrad.) Nees. Sorghum sudanense (Piper) Stapf Lespedeza cuneata (Dumont) G.Don. L.stipulacea Maxim. L.striata(Thunb.)H.&A.(Marion)		
American joint vetch Winter vetch Annual ryegrass Tall fescue Wheat Slender oat Crimpson clover Arrow leaf clover	Aschynomene americana L. Vicia villosa Roth Lolium multiflorum L.(Marshall) Festuca arundinacea Schreb.(KY31) Triticum aestivum L.(Roane) Avena sativa L.(Chapman) Trifolium incarnatum L.(Dixie) T. vesiculosum		
	Sorghum bicolor L.	All vegetated treatments resulted in higher remediation efficiency.	Banks <i>et al.,</i> 2003
	Brachiaria brizantha	The medium fertilizer concentration resulted in best root growth and highest absolute oil dissipation.	Merkl <i>et al.</i> , 2005
Cowpea	Vigna unguiculata	Total amylase, starch phosphorylase and mitotic activities were inhibited.	Achuba, 2006
	Phaseolus vulgaris	Can sustain growth in an oil polluted soil; Good candidate for phyto-remediation.	Nwoko <i>et al.,</i> 2007
Inflated-scale flat sedge Indian goose grass	Brachiaria brizantha (Hochst. ex. A. Rich.) Stapf Cyperus aggregatus (Willd.) Endl. Eleusine indica (L.) Gaertn.	Developed coarser roots larger specific root surface area in contaminated soil.	Merkl <i>et al.,</i> 2005
Broad beans	Vicia faba	Co-inoculation of <i>V. faba</i> plant roots with nodule bacteria and PGPR enhanced plant growth and nitrogen fixation.	Radwan <i>et</i> al., 2005
Okra	Abelmoschus esculetus	Combination of crude oil, microbes and fertilizers stimulated germination, growth, biomass, microbial population and rate of degradation.	Ogbonna <i>et</i> al., 2007



dichlorophenol, 2,4,6-trinitrotoluene, atrazine, polychlorinated biphenyls and some 3-5 ring PAHs. In a pot study, Joner and Leyval, (2003a) found that in PAH-contaminated soil planted with a mix of clover-ryegrass, PAH concentrations decreased as a function of proximity to the roots, and that mycorrhiza generally enhanced plant growth and favoured growth of clover than that of ryegrass.

Wang and Zhao (2007) from their studies on uptake and biodegradation of PAHs by marine seaweed Laminaria japonica reported that 0.1 mg/l conc. of PAH levels, less than 90% of phenanthrene and pyrene were removed from the seawater and that 70-90% of phenanthrene and 65-90% of pyrene which accumulated in the seaweed tissues were metabolised after 10 d of incubation. Therefore they suggest that in coastal waters contaminated with PAHs, Laminaria japonica could play an important role in removing PAHs and possibly other organic pollutants from sea water, thus serving as an environmentally-friendly' bioremediation system that protects ecological health and marine life. Their results also indicate that there is lower bioaccumulation of these PAHs in the seaweed tissues and get biodegraded rapidly to less or non-toxic forms of compounds by oxidant enzymes. This is an important public health factor considering the large quantities of seaweed consumption in China and other Asian countries.

Frick et al. (1999) stated that Triticum aestivum, Andropogon gerardii, Bouteloua curtipendula, Bouteloua gracilis, Buchloe dactyloides, Chloris gayana, Cynodon spp., Daucus carota, Elymus canadensis, Festuca aurundinacea, Festuca rubra, Glycine max, Lemna sp., Lolium multiflorum, Lolium perenne, Medicago sativa L., Panicum coloratum L., Panicum virgatum, Phaseolus vulgaris. Populus sp., Schizachyrium scoparium, Sorghastrum nutans, Sorghum vulgare, Sorghum bicolor subsp. drummondii and Zoysia japonica indicated potential significant phytoremediation for in-situ remediation of oil contaminated soils.

One commonly used plant for phytoremediation is tall fescue (*Festuca arundinacea*), a robust, broad-leaf grass. This plant is effective for enhancement of PAH dissipation in the rhizosphere (Banks *et al.*, 1999; Chen *et al.*, 2003; Chen & Banks, 2004; Huang *et al.*, 2004; Parrish *et al.*, 2005). These identified species provide the initial guidelines for remediation strategies in other tropical countries because phytoremediation of oil-contaminated soils is dependent on the ability of the selected species to enhance the degradation of petroleum hydrocarbons (Merkl *et al.*, 2004).

Challenges and future prospects

One component that has hindered widespread use of phytoremediation is the production of stress ethylene which results due to chemical toxicity, nutrient deficiency and/or water stress in impacted soils. However, Multiprocess phytoremediation system (MPPS) which can utilize plant/PGPR (plant growth promoting rhizobacteria)

interactions to mitigate the effects of stress ethylene is developed by plant biomass get enhanced particularly in the rhizosphere (Greenberg et al., 2007). Stressors and physical restrictions, heterogeneity in initial contaminant levels, root structure, soil structure, organic composition of soil, soil pH, moisture content and microbial activity and regulatory restrictions are the limiting factors in the use of genetically modified organisms for successful field trails. Yet, strategies to minimize ecological risks and improve methods for monitoring, sampling and analyzing experimental field data by addressing the inconsistencies in experimental protocols are needed for adequate performance of remediation systems at the field level (Gerhardt et al., 2009). Exploiting the natural biodiversity by recognizing suitable natural species that grow in the contaminated sites and understanding the mechanisms involved in the soil plant interactions may widen the scope of remediation attempts.

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