Bioremediation of Polycyclic Aromatic Hydrocarbons-Polluted Soils at Laboratory and Field Scale: A Review of the Literature on Plants and Microorganisms

Fabián Fernández-Luqueño, Fernando López-Valdez,
Cesar R. Sarabia-Castillo, Selvia García-Mayagoitia,
and Sergio R. Pérez-Ríos

Abstract The interrelationships between microbes and plants and the potential of utilizing these relationships to improve the dissipation of pollutants have been widely discussed during the last decades. However, to the best of our knowledge, there has been no prior study on the interrelationships between plants and microorganisms to degrade pollutants and shape a sustainable future. The characterization, identification, culturing, and management of plants and microorganisms suited for remediation techniques should be clearly defined, with the intention that the bioremediation techniques not only recover contaminated sites but also contribute to sustainable development and increasing social welfare. This chapter aims to provide the cutting-edge knowledge about the different biological interrelationships that are simultaneously taking place on a polluted site, prior, during, and after of the bioremediation strategies, taking into account and at the same time discussing the experimental findings at the laboratory and field scale by outstanding specialists.

Keywords Bioaugmentation • Biostimulation • Decontamination • Environmental Pollution • Phytoremediation • Sustainable Development

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F. Fernández-Luqueño (✉) • C.R. Sarabia-Castillo • S. García-Mayagoitia
Sustainability of Natural Resources and Energy Program,
Cinvestav-Saltillo, Coahuila C.P. 25900, Mexico
e-mail: cinves.cp.cha.luqueno@gmail.com

F. López-Valdez
Instituto Politécnico Nacional-CIBA, Tepetitla de Lardizábal, C.P. 90700 Tlaxcala, Mexico

S.R. Pérez-Ríos
ICAP–Instituto de Ciencias Agropecuarias, Universidad Autónoma del Estado de Hidalgo,
Tulancingo, Hidalgo C.P. 43600, Mexico

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Introduction

Living organisms such as plants, earthworms, and microorganisms have been recognized by their capacity to dissipate pollutants (Hong et al. 2015; Lu and Lu 2015; Xue et al. 2015). Some biochemical and physiological properties of these organisms are used to increase the dissipation of polycyclic aromatic hydrocarbons (PAHs) through biodegradation and bioremediation processes (Abbasian et al. 2015; Haritash and Kaushik 2009). Biodegradation is a natural way of recycling wastes or pollutants, which are usually used in relation to ecology, waste management, and mostly associated with bioremediation, a technology for environmental remediation. Bioremediation is defined as the treatment of pollutants or waste by the use of living organisms in order to eliminate, attenuate, degrade, transform, or break down (through metabolic or enzymatic action) the undesirable substances to inorganic components, such as CO_2, H_2O, and NO_3^- (Fernández-Luqueño et al. 2011; Pistelok and Jureczko 2014).

Organic pollution by PAHs is an increasing concern by the environmental scientists nowadays. Increasing concern for the environment has recently highlighted three major problems to resolved, namely, pollution, scarcity of resources, and unsustainable development of our societies. Pollution is defined as the introduction of elements, compounds, substances, or energy into the environment at concentrations that adversely alter its biological functioning or that present an unacceptable risk to humans or other targets that use or are linked to the environment (Fernández-Luqueño et al. 2011; Okparanma and Mouazen 2013; Berezina et al. 2015). In addition, PAHs pollution is a cause of many human and environmental health-related problems.

PAHs are organic molecules that often contaminate water (Fernández-Luqueño et al. 2013a; Leonov and Nemirovskaya 2011; Vodyanitskii 2014), soil (Alagic et al. 2015; Chen et al. 2015; Ibrahim et al. 2015; Wloka et al. 2015), sediments (Hall et al. 2011; Meng et al. 2015), and air (Ma and Harrad 2015; Szulejko et al. 2014). Although several hundred PAHs exist, most studies have been focused on a limited number of them, the so-called 16 EPA priority PAHs, seven of them might be mutagenic, carcinogenic, and teratogenic (Keith 2015). In the natural environment, the PAHs undergo transformations involving both biotic and abiotic processes such as volatilization, adsorption, photolysis, chemical oxidation, and the microbial degradation, among others. However, plants and microbial activities make up the primary pathway for PAHs removal from the environment (Fig. 1).

Recently, different papers have reviewed the biodegradation and bioremediation of soil, water, and air polluted with PAHs, e.g., Fernández-Luqueño et al. (2011), Abbasian et al. (2015), Alagic et al. (2015), and Xue et al. (2015). However, until now, there have been no reviews summarizing the relationship between microbial and vegetal populations under different PAHs-polluted ecosystems in order to enhance the degradation of PAHs, while the main biotechnological challenges to increase the biodegradation of PAHs at laboratory and field scale have neither been published. The objective of this chapter is to provide

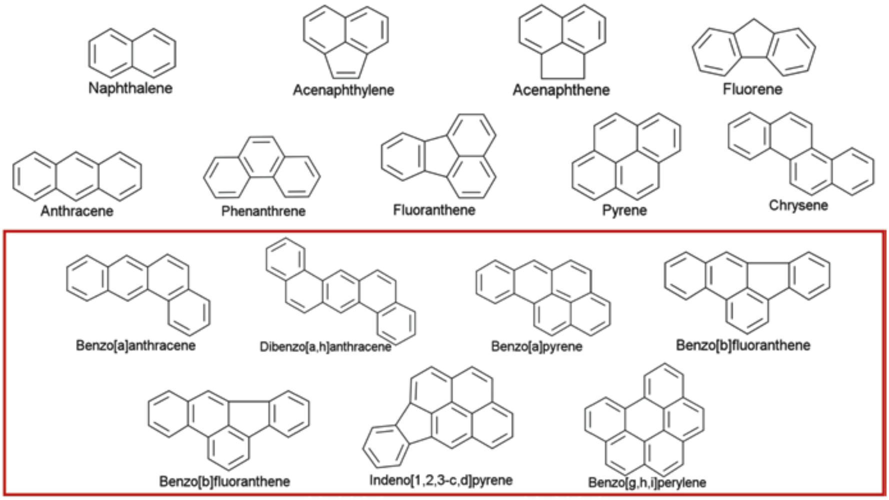

Fig. 1 List of 16 EPA priority polycyclic aromatic hydrocarbons (in the *red* box, there are seven PAHs that might be mutagenic, carcinogenic, and/or teratogenic)

the cutting-edge knowledge about the different biological interrelationships that are simultaneously taking place on a polluted site, prior, during, and after of the bioremediation strategies, taking into account and at the same time discussing the experimental findings at the laboratory and field scale by outstanding specialists.

Plants and Microorganisms Suited for Remediation Techniques

Pollution of soil, water, sediments, and air by PAHs is a common phenomenon across the globe, which may pose a great threat to the environment and human being at large. Different treatment methods have been employed to reclaim contaminated soils, water bodies, or air nowadays. However, plants and microorganisms have been recognized by their potential to dissipate PAHs within a very narrow range of climates and physical and biochemical characteristics of polluted substrates (soil, sediments, water, and air), e.g., Fernández-Luqueño et al. (2011) and Yavari et al. (2015).

Phytoremediation is a strategy that employs plants to degrade, stabilize, and/or remove PAHs, which can be an alternative green technology method for remediation of PAHs-polluted soils, water, and air. Phytoremediation, as a green technology option, is defined as the use of plants to remove pollutants from the environment or to render them harmless. This technique includes seven main strategies such as (Fig. 2):

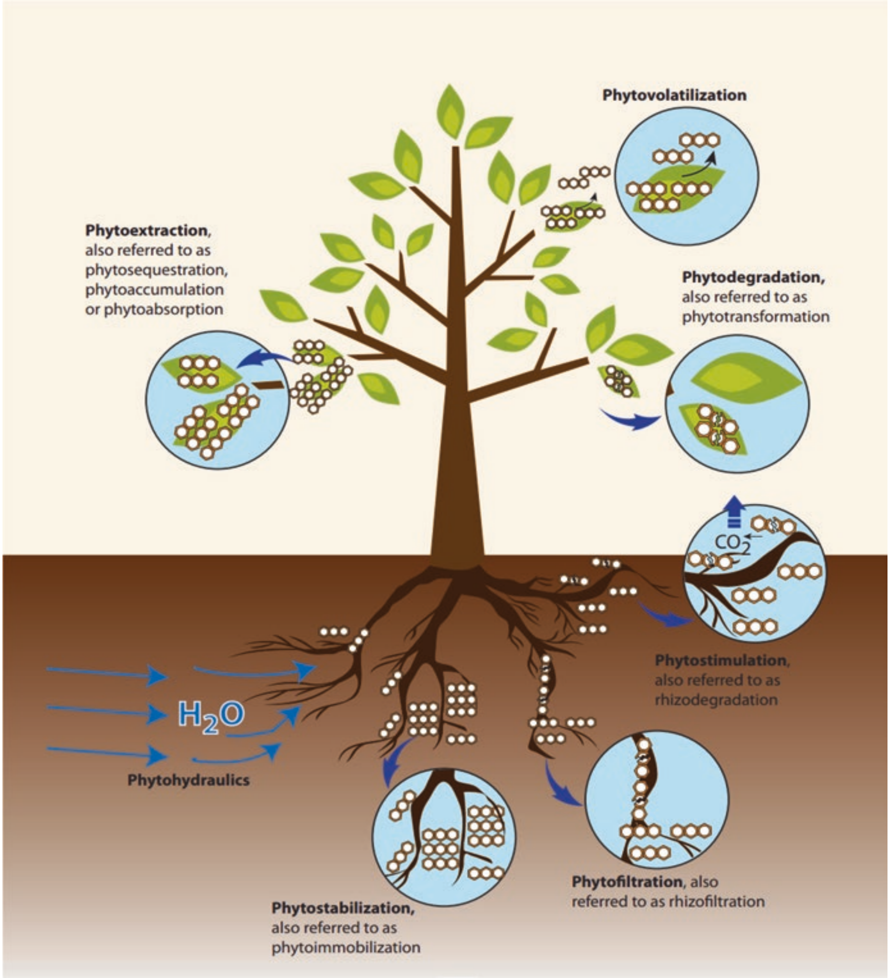

Fig. 2 Main strategies used to remediate contaminated soils, sediments, air, and water bodies

- Phytoextraction, also referred to as phytosequestration, phytoaccumulation, or phytoabsorption: plants remove PAHs from the soil and concentrate them in the harvestable parts of plants (Jiao et al. 2015).
- Phytodegradation, also referred to as phytotransformation: plants break down PAHs into simpler compounds that are integrated with plant tissue, which in turn, foster plant growth (Al-Baldawi et al. 2015).
- Phytofiltration, also referred to as rhizofiltration: plants and/or roots absorb, adsorb, concentrate, and/or precipitate PAHs. It involves filtering water through a mass of tissues to remove toxic substances or nutrients (Lee 2012).

- Phytohydraulics: this process is used to limit the movement of contaminants with water. Plants are used to increase evapotranspiration, thereby controlling soil water and contaminant movement (Hong et al. 2001).
- Phytostabilization, also referred to as phytoimmobilization: plants reduce the mobility and bioavailability of pollutants in the environment either by immobilization or by prevention of migration (Pulford and Watson 2003; Masu et al. 2014).
- Phytostimulation, also referred to as rhizodegradation: process where roots release compounds in order to enhance microbial activity in the rhizosphere through the rhizospheric associations among plants and symbiotic soil microorganisms (Gartler et al. 2014).
- Phytovolatilization: plants increase the volatilization of pollutants into the atmosphere via themselves through its ability to take up, translocate, and subsequently transpire volatile contaminants (Shiri et al. 2015).

It is well known that the plants may use more than one strategy of the abovementioned simultaneously during a common phytoremediation process. In addition, there are other strategies to improve the environmental quality and remove pollutants using plants, which are categories or variations of the abovementioned strategies. These include constructed wetlands, hydraulic barriers, phytodesalination, and vegetation covers.

Phytoremediation has now emerged as a promising strategy for in situ removal of many contaminants, while microbe-assisted phytoremediation including rhizoremediation appears to be particularly effective for the removal and/or degradation of organic contaminants from PAHs-polluted substrates (Zawierucha et al. 2014; Chen et al. 2016). Furthermore, root exudates from plants do help to dissipate PAHs and act as substrates for soil microorganisms, which result in increased rate of PAHs biodegradation. It has to be remembered that the strategies chosen for a phytoremediation project depend on the contaminant level, contaminant properties, and the contaminated matrix (Fig. 3).

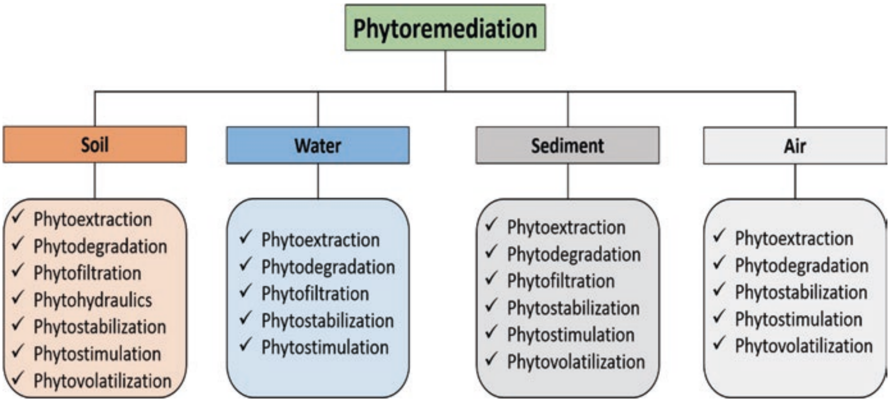

Fig. 3 Application of phytoremediation strategies as a function of the matrixes (such as soil, water, sediments, and air)

Different plants and crops have been found useful for phytoremediation of PAHs-polluted substrates (Table 1). Phytoremediation is a particularly useful in wetland environments because it uses plants and their associated microorganisms to recover PAHs-polluted soil and water (Table 2). Plant-associated rhizobacteria are involved in the PAHs degradation in contaminated substrates, while the plants themselves have the potential to enhance the rhizobacteria population. It is well known that many studies have been concentrating on the plant-microorganism interaction in phytoremediation, where the presence of autochthonous microorganisms can enhance the remediation efficiency of plants.

It has to be remembered that Macek et al. (2000) stated some advantages and disadvantages of phytoremediation. The main advantages of phytoremediation in comparison with classical remediation methods can be summarized as follows: (i) it is far less disruptive to the environment, (ii) there is no need for disposal sites, (iii) it has a high probability of public acceptance, (iv) it avoids excavation and heavy traffic, (v) it has potential versatility to treat a diverse range of hazardous materials, and (vi) it is cheaper than other techniques. However, the use of phytoremediation is also limited by the climatic and geological conditions of the site to be cleaned, temperature, altitude, soil type, and accessibility by agricultural equipment.

According to Macek et al. (2000), phytoremediation also has some disadvantages such as:

1. Formation of vegetation may be limited by extremes of environmental toxicity.
2. Contaminants collected in leaves can be released again to the environment during litter fall.
3. Contaminants can be accumulated in fuel woods.
4. The solubility of some contaminants may be increased, resulting in greater environmental damage and/or pollutant migration.
5. It may take longer than other technologies.
6. The plant biomass may require additional management prior to final disposition.
7. It may need the use of plants or microorganisms transgenic.
8. It requires technicians with strong academic skills about phytoremediation and about their economic, social, and environmental implications.

In addition, according to Eapen and D'Souza (2005), a plant suitable for phytoremediation should possess the following characteristics: (i) ability to tolerate, accumulate, or degrade pollutants in their aboveground parts, (ii) tolerance to pollutants concentration accumulated, (iii) fast growth and high biomass, (iv) widespread highly branched root system, and (v) easy harvestability.

Regarding the interactions among plants and indigenous rhizobacteria, Fernández-Luqueño et al. (2011) and Chen et al. (2016) stated that microbe-assisted phytoremediation has been well documented in scientific literature so that there is enough evidence to state that microbe-assisted phytoremediation has potential as an effective and inexpensive technique for removal, degradation, or dissipation of organic pollutants from polluted systems such as soils, water bodies, or air.

Table 1 Plants and/or crops used to phytoremediation, their rates of degradation, and the additional benefits

Plant	Degraded pollutant	Rate of PAHs degradation/ dissipation	Additional benefits	Reference(s)
Azolla caroliniana Willd.	Phenanthrene	80% in 49 days	Bioenergy production	Castro-Carrillo et al. (2008)
Bassia scoparia (L.) A.J.Scott	Crude oil (TPHs)	31.2–57.7% for natural soil and 28.7–51.1% for pre-sterilized soil after 5 months	Soil erosion control	Moubasher et al. (2015)
Bidens maximowicziana Oett.	Pyrene	79% in 50 days	Soil erosion control	Lu et al. (2010a)
Bidens pilosa L.	Crude oil (TPHs)	9% in 64 days	Soil erosion control	Kuo et al. (2014)
Brassica juncea (L.) Czern.	Pyrene	67% in 60 days	Can accumulate Cu	Chigbo and Batty (2013)
Brassica napus L.	Pyrene	30% in 90 days	Forage	D'Orazio et al. (2013)
Chromolaena odorata (L.) R.M. King & H. Rob.	Crude oil	80% in 180 days	It can degrade to 65% of HM in 180 days	Atagana (2011)
Cyperus brevifolius L.	Crude oil (TPHs)	61.2–86.2 in 360 days	Soil erosion control	Basumatary et al. (2012)
Echinacea purpurea (L.) Moench	Fluoranthene, pyrene, benzo(a) anthracene, chrysene, benzo(b) fluoranthene, benzo(k)fluoranthene, benz(a)pyrene, and dibenzo(a,h) anthracene	The removal rate of $\sum 8$ PAHs was 92.92% in 50 days	Soil erosion control	Liu et al. (2014a)
Eichhornia crassipes (Mart.) Solms	Naphthalene	66% after 5 h of experimental setup	Soil erosion control	Nesterenko-Malkovskaya et al. (2012)
Eleusine indica (L.) Gaerth.	PAHs	32% in 5 months	Soil erosion control	Lu et al. (2010b)
Festuca arundinacea Schreb.	PAHs	84% in 7 months	Soil erosion control	Soleimani et al. (2010)
Festuca pratensis Huds.	PAHs	64–72% in 7 months	Soil erosion control	Soleimani et al. (2010)

(continued)

Table 1 (continued)

Plant	Degraded pollutant	Rate of PAHs degradation/ dissipation	Additional benefits	Reference(s)
Fimbristylis littoralis Gaudich.	PAHs	92% in 90 days	Degrade 96% of as in 90 days	Oluchi-Nwaichi et al. (2015)
Fire Phoenix (is a combined Poaceae species including *Festuca arundinacea* Schreb., *Festuca elata* Keng ex E. Alexeev, and *Festuca gigantea* (L.) Vill.)	Fluoranthene, pyrene, benzo(a) anthracene, chrysene, benzo(b) fluoranthene, benzo(k)fluoranthene, benz(a)pyrene, and dibenzo(a,h) anthracene	The degradation rate of Σ8PAHs was 99.40% after 150 days	Soil erosion control	Liu et al. (2014b), Xiao et al. (2015)
Impatiens balsamina L.	TPHs	18.13–65.03% after 4 months	Soil erosion control	Cai et al. (2010)
Jatropha curcas L.	Lubricating oil (TPHs)	89.6–96.6% in 180 days	Bioenergy production	Agamuthu et al. (2010)
Juncus roemerianus Scheele	PAHs	84–100% in 1 year	Can degrade *n*-alkanes	Lin and Mendelssohn (2009)
Juncus subsecundus N.A.Wakef.	Phenanthrene and pyrene	97% and 43–63% after 10 weeks, respectively	Can accumulate Cd	Zhang et al. (2012)
Kandelia candel (L.) Druce	Phenanthrene and pyrene	56.8 and 47.7% after 60 days, respectively	Soil erosion control	Lu et al. (2011)
Lolium multiflorum Lam.	TPHs	59% in 80 days	Forage	Alarcon et al. (2008)
Lolium perenne L.	Pyrene	28% in 90 days	Livestock feed (forage)	D'Orazio et al. (2013)
Luffa acutangula (L.) Roxb.	Anthracene and fluoranthrene	98.2–98.9% and 85.9–96.9% in 45 days, respectively	Due to its fibrous characteristics, (the fruit) is used as exfoliating	Somtrakoon et al. (2014)
Medicago sativa L.	Pyrene, fluoranthene, benzo(a) anthracene, chrysene, benzo(b) fluoranthene, benzo(k)fluoranthene, benz(a)pyrene, and dibenzo(a,h) anthracene	32% removal of pyrene in 90 days and a removal rate of 98.11% for the ∑8PAHs in 150 days	Livestock feed (forage)	D'Orazio et al. (2013), Xiao et al. (2015)

Mirabilis jalapa L.	TPHs	41.61–63.20% in 127 days	Soil erosion control	Peng et al. (2009)
Onobrychis viciifolia Scop.	Pyrene and phenanthrene	74.1% and 85.02%, respectively, in 120 days	Forage	Baneshi et al. (2014)
Potamogeton crispus L.	Phenanthrene and pyrene	18.3–34.1% and 14.1–27.8%, respectively, in 54 days	Soil erosion control	Meng and Chi (2015)
Sagittaria trifolia L.	Diesel	54–85% in 50 days	Soil erosion control	Zhang et al. (2015)
Salix rubens Schrank	PAHs	98.56% in 36 months	Soil erosion control	Da Cunha et al. (2012)
Salix triandra L.	PAHs	98.65% in 36 months	Soil erosion control	Da Cunha et al. (2012)
Scirpus grossus L.f.	TPHs	66.6–81.5% in 72 days	Soil erosion control	Al-Baldawi et al. (2015)
Sorghum sp.	Pyrene and phenanthrene	73.84% and 85.2%, respectively, in 120 days	It used for human consumption and forage	Baneshi et al. (2014)
Tagetes patula L.	Benzo(a)pyrene	78.2–92.9% after 92 days	Can accumulate heavy metals	Sun et al. (2011)
Trifolium repens L.	Pyrene	77% after 60 days	Forage	Xu et al. (2009)
Triticum sp.	Pyrene and phenanthrene	65–70% and 98–100% in 90 days, respectively	Use of food	Shahsavari et al. (2015)
Vallisneria spiralis L.	Phenanthrene and pyrene	53.3–59.6% and 50–53.6% in 54 days, respectively	Soil erosion control	Liu et al. (2014c)
Zea mays L.	PAHs (crude oil)	52.21–72.84% in 60 days	Use of food	Liao et al. (2015)
Zostera marina L.	PAHs	73% after 60 weeks of treatment	Can degrade PCBs (polychlorinated biphenyls)	Huesemann et al. (2009)

Table 2 Microorganisms, identified in phytoremediation strategies, their rates of PAHs degradation, and the additional benefits

Microorganism	Degraded pollutant	Rate of PAHs degradation/ dissipation	Additional benefit	Reference(s)
Acinetobacter sp.	Phenanthrene and pyrene	90% of phenanthrene and 50% of pyrene in 6 days	It can degrade to 90% of fluorine	Shao et al. (2015)
Bacillus sp.	Diesel	74% in 60 days	Biohydrogen production	Cisneros- de La Cueva et al. (2014), Liu et al. (2015)
Cycloclasticus sp.	Phenanthrene, pyrene, and fluoranthene	98%–99% in 10 days and 52–63% and 49–65% in 21 days, respectively	Unreported	Cui et al. (2014)
Cycloclasticus sp. in association with *Marinobacter* sp.	Pyrene and fluoranthene	63–76% and 65–83% in 21 days, respectively	*Marinobacter* sp. is used for the production of α-amylases	Cui et al. (2014)
Halomonas sp. +Marinobacter sp.	Phenanthrene	90% in 12 days	*Halomonas* sp. has a high capability of bioremediation, due to they can be used as catalysts in different processes	Dastgheib et al. (2012)
Martelella sp.	Phenanthrene	100% in 6 days	Considered as PGPR	Feng et al. (2012)
Micrococcus sp.	Phenanthrene	99% in 21 days	They can degrade industrial substrates such as pyridine, herbicides, and polychlorinated biphenyls	Dellagnezze et al. (2014)
Ochrobactrum sp.	Anthracene, phenanthrene, naphthalene, fluorene, pyrene, benzo(k) fluoranthene, and benzo(e) pyrene	88, 98, 90, 97, 84, 57, and 50%, respectively, within 4–5 days	Considered as PGPR	Arulazhagan and Vasudevan (2011)
Ochrobactrum sp.	Phenanthrene	90% in 7 days	Considered as PGPR	Chang et al. (2011)
Penicillium sp.	Benzo(a)pyrene	83.84% in 5 days	Production of penicillin	Machin-Ramirez et al. (2010)
Pseudomonas sp.	PAHs and benzo(a)pyrene	3–5% of PAHs in 1 day and 12.73% of benzo(a)pyrene in 5 days	Considered as PGPR	Brito et al. (2015), Parray et al. (2015), Machin-Ramirez et al. (2010)

Pseudoxanthomonas sp.	Phenanthrene	100% within 120 h under optimized conditions	Unreported	Patel et al. (2012)
Rhodococcus sp.	Naphthalene and phenanthrene	100% and phenanthrene, respectively, within 9–11 days	Can degrade linear alkanes and branched alkanes	Yang et al. (2014)
Staphylococcus sp.	Phenanthrene	90% in 3 days	Unreported	Chang et al. (2011)
Streptomyces sp.	Naphthalene	81.03–85.23% in 12 days	Production of antibiotics	Ferrafji et al. (2014)
Achromobacter xylosoxidans	Fluoranthene	90% after 14 days	Unreported	Ma et al. (2015)
Acinetobacter venetianus	Diesel	>95% in 60 h	Has industrial applications for the production of the bioemulsifier emulsan	Lin et al. (2015)
Aspergillus niger	Benzo(a)pyrene	45% in 5 days	Used for the production of enzymes and chemicals	Machin-Ramirez et al. (2010)
Aspergillus sclerotiorum	Pyrene and benzo(a) pyrene	99.7% and 76.6% after 8 and 16 days, respectively	Unreported	Passarini et al. (2011)
Bacillus mycoides	Benzo(a)pyrene	27.06% after 5 days	They are in common pesticides and used to inhibit the growth of harmful bacteria and fungi	Machin-Ramirez et al. (2010)
Cladosporium cladosporioides	Pyrene and benzo(a) pyrene	42.1% and 45.3% after 8 and 16 days, respectively	Unreported	Passarini et al. (2011)
Dietzia maris	Phenanthrene	63% in 21 days	Production of canthaxanthin	Dellagnezze et al. (2014)
Ensifer meliloti (before *Sinorhizobium meliloti)*	Phenanthrene	46.3% in 5 days	Considered as PGPR	Muratova et al. (2014)
Fusarium solani	Pyrene	64.1–70.7% after 2 weeks	Acts as degrading plant material in soil	Hong et al. (2010)
Ganoderma lucidum	Phenanthrene and pyrene	>95% in 6 days	Has been investigated extensively for its pharmaceutical applications	Ting et al. (2011)
Hypocrea lixii	Pyrene	69.4% after 2 weeks	Improves plant growth	Hong et al. (2010)
Kocuria flava	Naphthalene	53% in 10 days	Considered as PGPR	Ahmed et al. (2010)
Kocuria rosea	Naphthalene and phenanthrene	36% and 9%, respectively, in 10 days	They can degrade bird feathers	Ahmed et al. (2010)

(continued)

Table 2 (continued)

Microorganism	Degraded pollutant	Rate of PAHs degradation/ dissipation	Additional benefit	Reference(s)
Methylobacterium populi	Phenanthrene	27% in 20 days	Considered as PGPR	Ventorino et al. (2014)
Mucor racemosus	Pyrene and benzo(a) pyrene	33.8$ and 51.7% after 8 and 16 days, respectively	It can be used in the fermentation for the production of bioethanol	Passarini et al. (2011)
Mycobacterium goodii	PAHs	3–4% day^{-1}	Unreported	Brito et al. (2015)
Novosphingobium pentaromativorans	Phenanthrene, pyrene, and benzo(a)pyrene	86.62% of the phenanthrene in 24 h, 31.81% of pyrene in 36 h, and 22.18 of benzo(a)pyrene in 48 h	Unreported	Lyu et al. (2014)
Penicillium commune	Industrial oil (PAHs)	95.4 in 5 days	Production of α-amylases	Esmaeili and Sadeghi (2014)
Pseudomonas aeruginosa	PAHs	100% degraded in 4 weeks	Considered as PGPR and biocontrol agent	Patowary et al. (2015), Goswami et al. (2015)
Rhizobium tropici	Phenanthrene and benzo(a)pyrene	50% and 45%, respectively, after 120 h	Contribute to biological nitrogen fixation	Gonzales-Paredes et al. (2013)
Saccharomyces cerevisiae	Benzo(a)pyrene	46% in 5 days	Industrial use	Machin-Ramirez et al. (2010)
Serratia marcescens	Benzo(a)pyrene	32.41% after 5 days	Unreported	Machin-Ramirez et al. (2010)
Sphingomonas koreensis	Naphthalene, phenanthrene, anthracene, and pyrene	100, 99, 98, and 92.7% within 15 days, respectively	Unreported	Hesham et al. (2014)
Trichoderma asperellum	Phenanthrene, pyrene and benzo(a)pyrene	74.4, 62.63 and 80.94% in 18 days, respectively	Biocontrol agent	Zafra et al. (2015)
Trichoderma harzianum	Benzo(a)pyrene	77% in 5 days	Biocontrol agent	Machin-Ramirez et al. (2010)
Trichoderma longibrachiatum	Phenanthrene	90% in 14 days	Used in biological control	Cobas et al. (2013), Zhang et al. (2014)
Trichoderma viride	PAHs	47% after 12 months of treatment	Biocontrol agent	Szczepaniak et al. (2015)
Yarrowia lipolytica	Crude oil (TPHs)	58–68% in 7 days	Production of lipases with application in biotechnology	Hassanshahian et al. (2012)

Biological Interrelationships Between Plant and Microorganisms in a Polluted Site: Insights into Prior, During, and After of the Bioremediation Strategies

For more than 120 years, the biological interrelationship among plants and microorganisms has been studied. However, the remediation techniques are not older than 30 years. Nevertheless, more and more studies have demonstrated the remediation's potential to recover polluted systems, which are becoming major environmental and human health concerns worldwide.

Lynch and Moffat (2005) were the first to use the term "phytobialremediation" in order to redefine phytoremediation assisted by microorganisms. Recently, it has been reported that plants and microorganisms help each other in the whole process of phytoremediation throughout phytobialremediation, which may be improved with transgenic technologies. Phytobialremediation is a technique, which can be carried out by free-living microorganisms or by symbiotic microbes, which live in the rhizosphere. In addition, it has to be remembered that plant microbial symbionts may constitute the "unseen majority" in phytoremediation of organic compounds (Fester et al. 2014). The rhizosphere is the microecological zone surrounding plant roots, i.e., it is a narrow region of soil that is directly influenced by root secretions and associated soil microbes. In the rhizosphere, the roots release a number of compounds establishing a highly dynamic and active microbial community distinctly different from the bulk soil microbial community. The exudates compounds increase contact among plant roots and the surrounding soil and prevent dehydration during dry spells. The functions of the plant root system include anchorage, the absorption of water and mineral nutrients, synthesis of various essential compounds, and the storage of food. Furthermore, the plant root system aerates the soil and provides a steady-state redox environment and a starting material for colonization of plant growth-promoting rhizobacteria (PGPR). PGPR are the rhizosphere bacteria that can enhance plant growth by a wide variety of mechanisms such as degradation of pollutants, phosphate solubilization, siderophore production, biological nitrogen fixation, antifungal activity, and induction of systemic resistance, among others (López-Valdez et al. 2015).

Chen et al. (2016) studied the potential of interplanting a Zn/Cd hyperaccumulator plant (*Sedum alfredii* L.) with a rhizospheric mediator (perennial ryegrass, *Lolium perenne* L.) for remediation of an actual wastewater-irrigated soil co-contaminated with PAHs and heavy metals in a 2-year greenhouse experiment, using *Microbacterium* sp. strain KL5 and *Candida tropicalis* strain C10. They found that the highest efficiency of PAHs removal, PAHs mineralization, and metal phytoextraction was obtained by interplanting ryegrass with *S. alfredii* associated with regular reinoculation with strain KL5 and C10 in the contaminated soil. Additionally, they reported that microbial inoculation promoted soil enzyme activity, PAHs removal, plant growth, and metal phytoextraction. Their date from qPCR and high-throughput sequencing suggest that reinoculation was necessary for the long-term remediation practice, and plants especially ryegrass were beneficial for PAHs degraders (Chen et al. 2016). As already explained, it has to be remembered that PAHs degradation in soils is dominated by bacterial and fungal strains belonging to a wide number of taxonomic groups (Fernández-Luqueño et al. 2011), i.e., it is well known that degradation/dissipation rates are strongly influenced by a wide number of soil microbial communities. Fernández-Luqueño et al. (2013b) studied the dynamics of the bacterial community composition, i.e., the diversity and abundance of microbial soil communities through PCR-DGGE of 16S rDNA gene fragments from a saline-alkaline soil polluted with PAHs. They found in a 56-days experiment that some microbial communities were harbored in the nine studied treatments. In addition, they found that the number of ribotypes increased in an alkaline-saline soil amended with wastewater sludge and spiked with phenanthrene and anthracene. Aertsen and Michiels (2005) noted similar results in a soil polluted with PAHs. They showed that both microorganism prokaryotes and eukaryotes possess mechanism that generates genetic and phenotypic diversity upon encountering stress such as PAHs spill.

Fernández-Luqueño et al. (2011) stated that the cutting-edge knowledge in the molecular genetics of plant and microorganisms and the knowledge-based methods of rational genetic modification suggest the possibility to develop plants and/or microorganisms that could decontaminate environments. The genomics and genetic engineering are the main biotechnological techniques to achieve this. Plants and microorganisms naturally respond differently to various kinds of stresses and gain fitness in the polluted environment. However, applying genetic engineering techniques can accelerate this natural process, but it has to be taken into account that ethical and social concerns are important. In addition, it has to be remembered that during the last several decades, plants and microorganisms have been widely investigated as unconventional systems for getting faster production of consumer goods and additional benefits. In genetic transformation processes, the gene of interest of donor plants, microorganisms, or viruses is transferred to host plants using methods such as *Agrobacterium* mediation, bombardment/biolistics, electroporation, a silicon-carbide fiber-based technique, polyethylene glycol-mediated protoplast fusion, and liposome-mediated gene transfer, among others. To date, transgenic plants have been engineered for the following purposes: to increase their tolerance to abiotic and biotic stresses, to improve the nutrient uptake, to reduce the effect of harmful agrochemicals, to increase their yield (grain production, growth rate, and biomass production), to increase the symbiotic interaction ships with soil microorganism, to increase the tolerance to pollutants, and to be used during phytoremediation processes (Abiri et al. 2016). Kotrba et al. (2009) published a review in which they summarize the state of the art on phytoremediation with genetically modified plants. Hannula et al. (2014) stated that the impact of genetically modified plants on natural or agricultural ecosystems showed that specific effects of single transformation events should be tested on a case-by-case basis in a natural setting where the baseline factors are all taken into consideration. In addition, Fernández-Luqueño et al. (2011) suggested that care should then be taken that the genetically modified microorganisms and plants do not outcompete the native ones or that negative traits spread through the soil microbial population.

Therefore, the environmental risk is latent when genetically modified microorganisms and plants are released in the environment in order to phytoremediate natural systems polluted with PAHs. New techniques such as stable isotope probing experiments, high-throughput sequencing, and meta-transcriptomics should be used in parallel with carefully designed field experiments considering a holistic review of the different individual reactions that are simultaneously taking place during the phytoremediation and that should be source of additional effects on the subsequent plant and microorganism species.

Increasing Social Welfare Throughout Remediation

Phytoremediation will become more economically feasible if the harvestable plant biomass results in financial returns (Mench et al. 2010). However, agronomic constraints, such as problems with crop rotation, climate, soil quality, and culture, must be considered. According to Mench et al. (2010), the commercial viability of a phytoremediating crop, which depends on total revenue (minus nonlabor variable costs) earned on the area to be cleaned up and calculated over an appropriate time period, is not decreased from what would be earned by conventional agricultural production. Decision making by the stakeholder must be assisted by a "cost-benefit analysis" accounting for the timely evolution of costs and benefits of phytoremediation. In addition, Mench et al. (2010) stated that assuming a predefined time period for the remediation, a cost-benefit approach could distinguish the cost of the phytoremediation action, capital, and operational costs connected with the contaminant removal, performance of the remediation crop, the soil or water conditions, and the difference between initial and target levels of contamination. Taken as a whole, these determine: (i) the remediation timescale, (ii) the income loss generated by the contaminated matrix, (iii) the potential income through biomass valorization, and (iv) the projected income from the remediated matrix, determined by its functional use (Mench et al. 2010; Ciesielczuk et al. 2014). However, the economics of phytoremediation is frequently favorable, but financial returns from produced biomass and element recycling have yet to be optimized. In addition, strategies for phytoremediation have to be relied on sustainable development, because environment protection does not preclude economic development, and economic development is ecologically viable today and in the long run.

Phytoremediation appears to be a feasible approach for cleaning contaminated matrix with PAHs, but technical hindrances have to be overtaken to shape a sustainable future throughout remediation techniques. In addition, a widespread lack of awareness among governments and societies about the current scale, pervasiveness, and risk to billions of people from environmental contamination hinders the establishment of strategies to stop/reduce the PAHs pollution. However it has to be remembered that phytoremediation is an efficient and cheap technique, but it is not free. Finally, site decontamination should be regarded as integral to bioeconomy and sustainability goals.

Conclusions and Perspectives

A substantially large body of information on the potential of phytoremediation for cleaning up the environment has been gathered together. Here we summarize the gained experience, which has helped to prove the suitability of plants and microorganism to remediate polluted environments. However, it has to be remembered that many technical hindrances currently limit the efficiency of phytoremediation. In addition, it has to be taken into account that to protect human health and the environment is necessary to develop and to promote innovative cleanup strategies that restore polluted sites/matrix to incorporate them to a productive use and promote the environmental stewardship and the sustainable development. Sharing scientific knowledge and technologies for assessing, cleaning, and preventing contamination is necessary, but the lack of environmental education in our society is evident, while the universities and research centers have the commitment of preparing young engineers with strong academic skills to address and decontaminate the increasingly polluted environment. We must not forget that the multidisciplinary nature of assessment and cleanup of polluted sites requires a complex and costly team of experts.

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