

Review

Rhizospheric microorganisms as a solution for the recovery of soils contaminated by petroleum: A review



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ABSTRACT

Petroleum is currently the world's main energy source, and its demand is expected to increase in coming years. Its intense exploitation can lead to an increase in the number of environmental accidents, such as spills and leaks, and an increase in the generation of environmental liabilities resulting from refining. Due to its hydrophobic characteristics and slow process of biodegradation, petroleum can remain in the environment for a long time and its toxicity can cause a negative impact on both terrestrial and aquatic ecosystems, with the main negative effects related to its carcinogenic potential for both animals and humans. The objective of the present review is to discuss environmental contamination by oil, conventional treatment techniques and bioremediation an alternative tool for recovery petroleum-contaminated soils, focusing on the rhizodegradation process, plant growth-promoting rhizobacteria (PGPR), a phytoremediation strategy in which the microorganisms that colonize the roots of phytoremediator plants are responsible for the biodegradation of petroleum. These microorganisms can be selected and tested individually or in the form of consortia to evaluate their potential for oil degradation, or even to measure the use of biosurfactants produced by them to constitute tools for the development of environmental recovery strategies and biotechnological application.

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1. Introduction

Among the numerous forms of energy, oil represents a considerable part of the world energy source. According to the

International Energy Agency (IEA, 2017), in order to meet world demand, in 2015 97 million barrels of oil were needed per day, and it is estimated that 100 million barrels of oil per day will be needed up to 2021. At the same time, as the use of petroleum is of great relevance to society, numerous environmental accidents, such as spills and leaks, can be caused during exploration activities, and they can generate waste from refining, which can cause both

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terrestrial and aquatic ecosystems damages when they are not adequately treated (Wang et al., 2014).

Soil and water contamination caused by accidents can occur in several environmental compartments: groundwater (Liu et al., 2015), soils (Li et al., 2015), rocky shores (Kankara et al., 2016), sediments (Burnes and Jones, 2016) and oceans (Broszeit et al., 2016), causing impacts mainly from a toxicological point of view. In this way, the oil entry into ecosystems is one of the major environmental concerns. According to Vaziri et al. (2013), oil contamination can cause cancer in living beings, especially by their recalcitrant properties.

When the environment with petroleum is contaminated, it is necessary to use remediation techniques to recover the degraded area. Most of these techniques are not feasible due to the extent of the contamination. Therefore, it is necessary to search for alternative decontamination technologies namely biological technologies that, according to Shekoohiyan et al. (2016), are feasible, easy to apply, cost-effective, and according to García-Sánchez et al. (2018), are effective in remediation of environmental contamination.

Considering all these characteristics, phytoremediation stands out as a promising alternative in terms of biological treatment. Brzeszcz et al. (2016) and Seckin (2016) define phytoremediation as an alternative technology that involves the use of plants and microorganisms associated with their roots in order to reduce environmental impact. According to Alaribe and Agamuthu (2015) and Hanks et al. (2015), in the case of petroleum, these depollution agents are able to degrade toxic compounds when they use them as energy source, releasing substances, often less toxic for the environment.

Rhizoremediation, composed by the combined use of phytoremediation and bioremediation, has been proposed as a gentle remediation option to rehabilitate multi-contaminated soils (Lacalle et al., 2018). The rhizodegradation is a strategy for phytoremediation of polluted environments with petroleum and its derivatives. In this process, plants act indirectly in phytoremediation, since their presence in the environment provides favorable conditions for the growth of microorganisms in the rhizosphere region. These microorganisms are able to biodegrade the toxic compounds. The microorganisms commonly isolated from the rhizosphere of phytoremediation plants according to Rein et al. (2016), are bacteria, fungi and yeasts. Thus, the purpose of this review is to discuss about the environmental contamination by oil, the treatment techniques conventionally used, and alternative treatments, more specifically the rhizodegradation process, plant growth-promoting rhizobacteria (PGPR), advantages and limitations of rhizodegradation. For this purpose, the search for information in the Web of Science - WoS database was performed, in the online version. The search was carried out in English language works and covered the years from 2000 to 2018. The research was based on the search for articles that had as descriptors: petroleum, petrol derivatives, soil contamination, bioremediation, phytoremediation, rhizodegradation, rhizosphere, plant-microbe association, PGPR, and microorganisms.

2. Environmental contamination by oil and negative impacts to environmental compartments

Several studies in recent years involve the analysis of environments contaminated by different pollutants from industrial activities. Among the most studied contaminants, pesticides, chlorophenols and heavy metals can be highlighted, and the same importance is verified when the pollutant is petroleum (Pi et al., 2016; Shahi et al., 2016; Shekoohiyan et al., 2016; Venkidusamy et al., 2016; Wang et al., 2016; Zhang et al., 2016).

Due to the technological development and capital accumulation,

occurred the world trade's expansion and dependence on oil and its derivatives (Carneiro and Gariglio, 2010), since they are the main products used in both domestic and industrial activities (Andrade et al., 2010).

Petroleum is composed of a complex mixture of aliphatic, aromatic and heterocyclic hydrocarbons (Falkova et al., 2016), formed by biogeochemical processes and its found at varying depths in the subsoil. Depending on its origin, most of its components (from 60% to 90%) are classified as biodegradable and the smallest part (from 10% to 40%) is characterized as crude or recalcitrant; in other words, not biodegradable, resulting in a delay to disappear from the environment.

The problems commonly associated with the disposal of petroleum in the environment involve accidental spills during the transport, management or storage of hydrocarbons in underground deposits, distribution pipes, or refining processes (Das and Chandran, 2010; Smith et al., 2015). Around these refineries and near the transport facilities, the impacts comprise a challenge of managing the oil production chain (Olajide and Ogbeifun, 2010; Vaziri et al., 2013).

Mena et al. (2016) report that when there is an oil spill, the contaminant fills the pores of the soil and it is rapidly adsorbed by its particles, moving vertically with capillary forces with the aid of gravity, altering the chemical, physical and biological compositions of the soil. The oil penetrates the soil easily until it reaches groundwater, which changes its ecological and geochemical states.

An initial modification occurs in the availability of oxygen, because oxygen is an electron acceptor molecule that stimulates the degradation of petroleum hydrocarbons (Ramírez-Pérez et al., 2015). Under reduced or absent oxygen conditions, aerobic microorganisms present in contaminated soil have their metabolism partially interrupted, which directly interferes with microbial activity, thus affecting ecosystems, causing negative impacts due to their toxic properties (Chen et al., 2015).

The negative impacts on different ecosystems include the impairment of altered biotic and abiotic conditions, mainly reflecting loss of habitat and serious consequences on biodiversity. According to Nardeli et al. (2016), the most common impacts related to the presence of petroleum in the environment include abiotic stresses such as heat, hypoxia, oxidative and osmotic stresses. Gargouri et al. (2015) mention that in general, the pollution of natural resources with petroleum hydrocarbons and their derivatives cause soil's impermeabilization and imbalance in the groundwater's quality. According to Abbasian et al. (2016), and Andreolli et al. (2015), changes in soil microbial composition and inhibition of activity are also highlighted, as a consequence of the alteration in soil's chemical composition and low nutrient availability.

In plants, the negative effects occur in the growth and development stages, in other words, from seed germination to reproduction. The presence of petroleum in the soil induces oxidative stress, which reduces growth and causes foliar deformation and tissue necrosis, as well as disturbances in the signalling of metabolic pathways of oxygen reactive species and responses related to defense against pathogens (Nardeli et al., 2016).

The animals are also harmed due to damaging to the conditions of their habitat, a fact that can lead to the development of cancer and other diseases, as well as imbalances in the food chain. Insufficiency in reproductive capacity and bioaccumulation in the food chain at levels that disturb biochemical and physiological processes are also reported. The sum of these environmental imbalances leads directly or indirectly to human health, since oil contamination has high toxicity and carcinogenic, mutagenic and teratogenic potential, as affirmed by Chen et al. (2015), and Rein et al. (2016). Depression of the nervous system, narcosis and irritation of the

mucous membranes of the eyes are some of the symptoms caused in humans by oil contamination (Bezza and Chirwa, 2015; Ameen et al., 2016; Bastida et al., 2016).

3. Treatment of environments contaminated by petroleum

Faced with the problem that involves the environmental contamination by oil, it is necessary to carry out its treatment. There are several forms of treatment of environments contaminated by oil and the choice of the most appropriate technique is decisive for the minimization of environmental impacts. Physical, chemical and biological methods are developed and employed to perform soil cleaning by reducing the concentration and/or toxicity of the contaminants, or even by eliminating them.

Studies based on the recovery of contaminated environments are becoming increasingly necessary, since several ecosystems are the ones that are the target of negative impacts caused mainly by accidents of petrogenetic origin (Moreira et al., 2011).

A variety of conventional technological options for treatment of oil-contaminated soils are available (Hu and Chan, 2015) and are based on thermal, chemical and physicochemical treatments, such as incineration, thermal desorption (Kastanek et al., 2016), electro kinetics, extraction of vapours (Covino et al., 2016; Ivshina et al., 2016; Lim et al., 2016), microwave heating, chemical oxidation, ultrasound, flotation, and solvent extraction (Lim et al., 2016). Informations about these technologies are summarized in Table 1.

These technologies can be classified into two groups: *in situ*, when the remediation is done in the contaminated site itself and *ex situ* when the remediation is carried out in a different place from where the contamination occurred (Mena et al., 2016).

The degree of success of remediation depends on a number of factors that influence oil degradation, such as: soil texture, pH, organic matter, nutrients, temperature, humidity, rhizospheric exudates, salinity, microbial community present, pollutant tolerance and concentrations of biodegradable hydrocarbons; in other words, their bioavailability determined by their accessibility to adsorption. The application of the selected treatment also influences the rate of degradation of the pollutant. According to Smith et al. (2015) and

Chen et al. (2015), the bioavailability of the contaminant varies along time. This process is called as aging or sequestration and, according to Muniz et al. (2004), it is a challenge for the rehabilitation of the environment, since the sorption becomes stronger with the passage of time, resulting in a fraction of persistent compounds in the environment (Rein et al., 2016).

Conventional treatment technologies have their own characteristics, advantages and limitations. Despite the proven efficiency of these techniques, when considering some aspects of their practical application, there are some disadvantages in their use, such as: limited application in difficult to reach places; the amount of contaminated soil to be treated can economically make the remediation impracticable; and complete destruction of soil microbiota.

For the remediation of soils contaminated with petroleum, it is necessary to integrate them with alternative technologies, with the objective of complementing and improving the efficiency of environmental decontamination. However, alternative technologies are considered to be less environmentally aggressive, have greater ease of practical application in any contaminated environment, and are cost-effective compared to conventional technologies. Specifically, in this study, the application aspects of bioremediation will be considered.

4. Bioremediation: an alternative tool for recovery petroleum-contaminated soils

Contamination of water and soil is a major problem for all organisms worldwide. Depending on the nature of the pollutant, diversity in the biosphere is diverse. Pollution can be attenuated with the use of bioremediation, which is a process in which living organisms or their products are used naturally, or artificially to remediate or reduce (remediable) pollutants in the environment (Gouda et al., 2018). For this purpose, plants and microorganisms that tolerate and have the capacity to grow in the presence of the pollutant are usually used.

Bioremediation is an important environmental service that allows the recovery of different ecosystems from the auto-depuration

Table 1
The main technologies conventionally employed for the treatment of environments contaminated with petroleum, their concepts, advantages and disadvantages adapted from Lim et al. (2016).

Technologies	Concept	Advantages	Disadvantages
Thermal			
Incineration	Destruction of the soil by temperature close to 1000 °C.	Can remove 100% of contaminant; Covers large volumes of areas.	Releases polluting by-products to atmosphere; Subsequent need for air filtration; High operating cost.
Thermal Desorption	Increased vapor pressure.	Removes up to 95% of the oil; Emits little or no contaminating gas.	Applicable only for volatile contaminants.
Microwave Frequency Heating	Conversion of energy to volatilize the contaminant.	Removes 100% of oil; Fast processing; Reduces total energy consumption.	It impoverishes the soil; Can cause problems for human health.
Chemical			
Chemical Oxidation	Oxidation of the contaminant by chemical compounds.	Minimizing the dispersion of the contaminant; Does not produce toxic byproducts.	Can destroy microbiota from soil; Does not support the vegetation.
Electro kinetics	Application of electric current for the removal of the pollutant.	Speed of execution; Feasible cost.	Ineffective in low contaminant concentrations; Creating hot spots around the electrodes.
Physical-Chemical			
Ultrasound	Microbubbles that break the chemical bonds of the contaminant.	Practically does not use chemicals.	High cost; High energy consumption.
Flotation	Separation of solid particles of different natures.	Simple technology; Low operating cost.	Requires large amounts of water; Aged oil reduces efficiency by up to 27%.
Extraction by Solvent	Solvent application for dilution of the contaminant.	Removes more than 97% of oil; Development of green solvents.	Low interaction between soil and solvent mixture; High operating cost.
Extraction by Steam	Induction of controlled airflow to remove contaminant.	It does not pollute the atmosphere; Effective in highly porous soils; short term; Stimulates the growth of the microbiota.	Abiotic factors affect efficiency; Not suitable for soils with low air permeability and / or high water content.

performed by the microorganisms. As reported by Papadaki and Mantzouridou (2016), microorganisms are capable of degrading or converting contaminants that can be used as an energy source, or they produce less and less toxic waste reducing their concentration over time. According to Tahir et al. (2016), the adaptation of microorganisms to the stresses caused by the presence of pollutants in the environment plays a fundamental role in recovery and environmental restoration due to characteristics such as resistance and metabolic potential that are inherited during natural selection.

This process can occur naturally, that is, without human intervention. In this case, this natural remediation is not considered as an alternative treatment, but can minimize the negative consequences and impacts on the environment and human health. Adetutu et al. (2013) report that, in 320 days the remediation was of only 46.4% of the oil in contaminated soil. According to Jacques et al. (2007), because it involves exclusively natural processes, this natural remediation can take years for total or partial recovery, which requires an intervention using assisted bioremediation strategies, as well as constant environmental monitoring.

In the last decades, the increasing recognition of the efficiency and economic viability of the use of bioremediation for the treatment of soil contaminated by petroleum and its derivatives has been witnessed (Broszeit et al., 2016; Laczi et al., 2015). Some microorganisms such as bacteria, fungi or yeasts, have a high capacity for degradation of pollutants, so they are widely used for environmental depollution (Kuppusamy et al., 2017).

Although it is time consuming, bioremediation is one of the most cost-effective means of remediating soil and water pollution. According to Ullah et al. (2015), regarding assisted bioremediation, different techniques can be applied, such as bio-ventilation, bio-lixiviation, bioreactors, composting, bioaugmentation, biostimulation, phytoremediation, and rhizodegradation. However, application of these techniques is unidirectional and need be further associated with each other to overcome such limitations.

In the case of bioremediation of soils contaminated with petroleum, these microorganisms can be isolated from the roots of phytoremediation plants. A phytoremediation plant of oil contaminated soils must first be tolerant to the presence of the pollutant and, when growing, it should provide favorable conditions for the growth of microorganisms in the rhizosphere, by increasing aeration, eliminating rhizospheric exudates, and consequently providing the biodegradation of the contaminants. Microorganisms, in turn, secrete enzymes that degrade oil, and as a result generate energy for cells and substances like water and carbon dioxide that are released into the environment. This process is called rhizodegradation, and the schematic representation of this process in soil contaminated by petroleum can be observed in Fig. 1.

The rhizodegradation provides the natural recovery of the environment, as the contaminant is gradually being biodegraded;

however, the rate of degradation cannot be slower than the negative consequences of oil on the environment. Thus, the analysis of the environment, type of vegetation and soil microbiota are predominant factors in the rate of degradation of the oil.

In situ bioremediation is notable for involving stimulation (biostimulation) of indigenous microorganisms capable of degrading petroleum contaminants and it is often used in association with other remediation technologies as a means of maximizing success in achieving positive results and reducing costs (Hu and Chan, 2015). Moreover, bioremediation is a biotechnological approach that allows the recovery of contaminated soil with lower environmental impacts when compared to non-biological methods (Martínez-Álvarez et al., 2015).

The success of bioremediation depends on several environmental factors that directly influence the recovery of the contaminated environment. Among these factors, the availability of oxygen and the concentrations of available nutrients necessary for the microbial community to metabolize the contaminants are highlighted (Martínez-Álvarez et al., 2015). However, adverse environmental conditions, such as extreme pH values, high salinity, imbalanced amounts of nutrients and the presence of toxic compounds may inhibit microbial growth and metabolism (Papadaki and Mantzouridou, 2016).

4.1. Microbe-assisted phytoremediation

Phytoremediation is an emerging technology that uses plants and their associated microbes to clean up pollutants from the soil, water, and air. Throughout the development phase of plants, there is an alliance between soil, plant and microorganisms. This association is developed when the microbial community in the rhizosphere is stimulated by release of plant rhizodeposits. Several groups of microorganisms depend on these mechanisms, thus establishing a gradient of interactions of the promotion of the growth of the plant to the parasitism (Tabassum et al., 2017).

These microorganisms may influence the availability of the contaminant and the uptake into the rhizosphere by plants. These associations may modify the chemical composition of root exudates and pH of the soil and, therefore, bioavailability of contaminants in the soil (Sarwar et al., 2017).

Plant growth-promoting rhizobacteria (PGPR) has been used as a co-evolution between plants and microbes showing antagonistic and synergistic interactions with microorganisms and the soil. Microbial revitalization using plant growth promoters had been achieved through direct and indirect approaches like invigorating root growth, bio-fertilization, rhizoremediation etc (Gouda et al., 2018).

The symbiotic and non-symbiotic relationships between microbes and plants are making it a single candidate for the

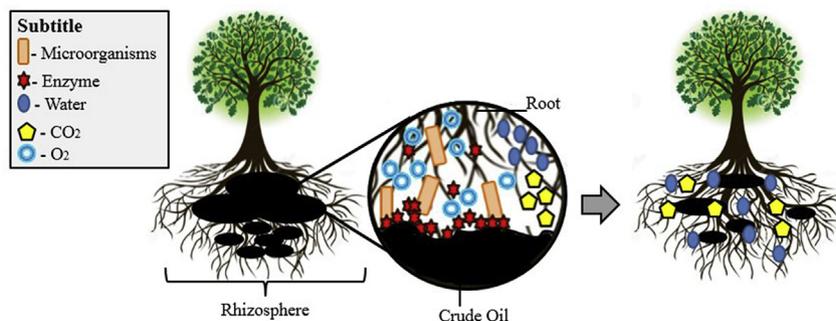


Fig. 1. Rhizodegradation process of soils contaminated with petroleum, which involves the growth of microorganisms associated to the rhizosphere of plants tolerant to the contaminant and, therefore, its biodegradation, with the release of water and carbon dioxide to the environment.

rhizoremediation. According to Etesami (2018), PGPR enhance plant growth and plant tolerance to biotic and abiotic stresses by different action mechanisms, often more than one mechanism. As report by Pérez-Montaño et al. (2014) and Tabassum et al. (2017), the PGPR promotes the growth of the plant by using its own metabolism (solubilizing phosphates, producing phyto hormones or fixing nitrogen), or can directly affect the plant metabolism (increasing the absorption of water and minerals) that consequently increases: root development; the enzymatic activity of the plant; provides the development of other microorganisms beneficial to plants; and may suppress plant pathogens. Some recent successful examples of PGPR application for the phytoremediation of PAH and other contaminants in soil are shown in Table 2.

One of the clearly beneficial the effects of PGPR include the suppression of deleterious microorganisms along with the promotion of plant growth attributes. PGPR can produce antibiotics, compete for nutrients with pathogens or induce systemic resistance in the host plant to defend it against pathogens.

In general, developing more detailed fundamental knowledge about the interactions between PGPR and plants, and their application as bioremediators would facilitate a better understanding of alleviating contaminants toxicity stress of sources of soil and water, and perhaps would allow better predictions regarding the plant response.

4.2. Mechanism of rhizodegradation

Plant species are colonized by a large number of microorganisms in several regions, including their tissues (endophytes), leaves (epiphytes) and rhizosphere, but it is in the rhizosphere that the mechanism of rhizodegradation occurs in the associated microbial community (Nardeli et al., 2016).

The rhizosphere is a complex ecosystem consisting of the narrow nutrient-rich soil zone (Razavi et al., 2016) ranging from the surface to a depth of 1–5 mm (Lim et al., 2016). Newman et al. (2016) and Sillen et al. (2015) argue that in the rhizosphere, the interdependence between microorganisms and plants results in a symbiotic association creating an interactive microbiome influenced by the roots of plants and their activities that, in association

with the roots, produce primary and secondary metabolites.

According to Venturi and Keel (2016), this variety of primary metabolites (organic acids, carbohydrates, and amino acids) and secondary metabolites (alkaloids, terpenes and phenolic compounds) interferes in some way with the rhizosphere microflora. Most of the metabolic activity occurs due to the interactions between energy and nutrients, exudates, loss of the border cells of the hood, gaseous losses and the deposition of substances (Razavi et al., 2016), providing the basis for the establishment of plant/microbe interactions, which increases the volume of the microbial biomass according to the irregularities of the root surfaces (Liang et al., 2016; Venturi and Keel, 2016). It is also important to highlight the release of oxygen by the roots of the plants, aerating the pores of the soil and allowing the development of aerobic organisms, which facilitates the rhizodegradation.

These factors make the rhizosphere densely inhabited by several organisms including bacteria, fungi, protists, nematodes and invertebrates, which will benefit the plant by providing the necessary vitamins, amino acids and cytokinins to support plant growth (Lim et al., 2016). Microbial density and enzymatic activity are high in the proximity of roots or residues and decrease with increasing distance, forming distinct millimeter gradients (Marschner et al., 2012).

When the soil is contaminated by oil, the amount of nutrients available in the rhizosphere decreases and the oleophilic microorganisms begin to degrade the contaminant generating energy for itself. As a result of this metabolism, products such as less toxic organic compounds, water and carbon dioxide (Wolicka et al., 2009) are excreted from the microbial cell, making it available both to the plant and to other microorganisms that begin to develop in this environment in the process of bioremediation (Fig. 2).

Studies show that distinct microorganisms that develop in these environments may have tolerance to the contaminant in an environmental accident. Bacteria, fungi, yeasts and algae have been used for the application of the bioremediation technique of soils contaminated with petroleum. In Poland, Wolicka et al. (2009) isolated aerobic microorganisms from an area contaminated by petroleum products. The degradation activity of benzene, toluene, ethylbenzene and xylene (BTEX) as evaluated in the field and in the

Table 2
Examples of successful remediation of hydrocarbons from soil by combined use of plants and bacteria inhabiting around/on the root surface - Plant growth-promoting rhizobacteria (PGPR) process.

Plant	Rhizobacteria	Action mechanism	Reference
<i>Lolium multiflorum</i>	<i>Glomus mosseae</i> and <i>Acinetobacter</i> sp.	PAH bioremediation	Yu et al. (2011)
<i>Axonopus affinis</i>	<i>Pseudomonas</i> sp. ITRH25, <i>Pantoea</i> sp. BTRH7 and <i>Burkholderia</i> sp. PsJN	Oil diesel removal	Tara et al. (2014)
<i>Scirpus triquetter</i>	<i>Pseudomonas</i> sp. J4A]	Oil diesel removal	Zhang et al. (2014b)
<i>Testuca arundinacea</i>	<i>Lysobacter</i> , <i>Pseudoxanthomonas</i> , <i>Planctomyces</i> , <i>Nocardioiodes</i> , <i>Hydrogenophaga</i> , <i>Ohtaekwangia</i>	Petroleum phytoremediation	Hou et al. (2015)
<i>Medicago sativa</i>	<i>Pseudomonas aeruginosa</i>	Heavy metals and petroleum bioaugmentation-assisted	Agnello et al. (2016)
<i>Jatropha curcas</i>	<i>Flavobacterium</i> (B7), <i>Serratia</i> (B8), <i>Pasteurella</i> (B1) and <i>Azotobacter</i> (B6)	1,4-dichlorobenzene degradation	Pant et al. (2016)
<i>Arabidopsis thaliana</i>	<i>Bacillus megaterium</i>	Overcome salt stress	Erice et al., 2017
<i>Medicago sativa</i>	Arbuscular mycorrhizal fungi	PAH rhizoremediation	Gao et al. (2017)
<i>Lolium multiflorum</i>	<i>Mycobacterium gilvum</i>	PAH degradation	Guo et al. (2017)
<i>Brassica nigra</i>	<i>Alcaligenes</i> , <i>Bacillus</i> , <i>Curtobacterium</i> and <i>Microbacterium</i>	Heavy metal phytoremediation	Roman-Ponce et al. (2017)
<i>Oryza sativa</i>	<i>Thalassobacillus denorans</i> (NCCP-58) and <i>Oceanobacillus kapialis</i>	Overcome salt stress	Shah et al. (2017)
<i>Raphanus raphanistrum</i>	<i>Bacillus</i> sp. CIK-512	Lead (Pb) phytoaccumulation	Ahmad et al. (2018)
<i>Raphanus sativus</i>	<i>Bacillus</i> sp. CIK-516	Decrease of negative impacts on soil contaminated with nickel	Akhtar et al. (2018)
Mangrove rhizosphere	<i>Kocuria flava</i> AB402 and <i>Bacillus vietnamensis</i> AB403	Arsenic bioremediation	Mallick et al. (2018)
<i>Armoracia rusticana</i>	<i>Rhizobium radiobacter</i> and <i>Diaphorobacter nitroreducens</i>	Carbamazepine degradation	Sauvêtre et al. (2018)

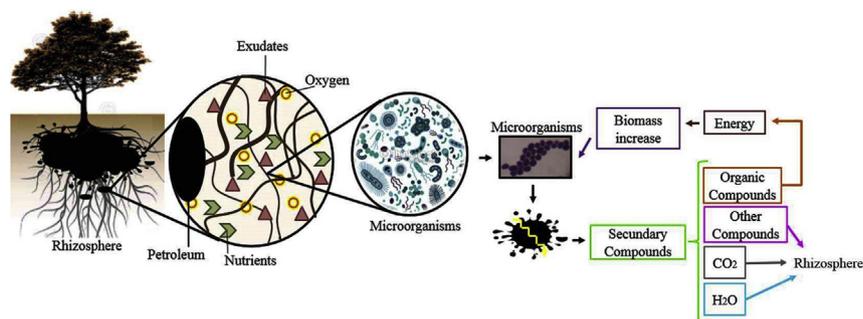


Fig. 2. Rhizodegradation mechanism in soil contaminated with petroleum. Exudates, nutrients and oxygen are released by the plants in their roots that are absorbed by the microorganisms. These tolerant microorganisms will transform part of this oil into secondary compounds such as less toxic substances, carbon dioxide and water that will be released into the rhizospheric environment, and another part, organic compounds will be used as a source of energy for the growth of its biomass.

laboratory. In the laboratory, a reduction of 84% of benzene, 86% of toluene, 80% of ethylbenzene and 82% of xylene was obtained, and under field conditions 95% reductions of benzene and toluene, 81% ethylbenzene and 80% xylene was obtained over a period of 210 days.

In the bioremediation of oil contaminated environments, the most widely used organisms are bacteria, as they are distinguished by high frequency, rapid growth and a broad spectrum of degradation of petroleum products (Wolicka et al., 2009). Almansoori et al. (2015) report the use of *Serratia marcescens* for the removal of gasoline in contaminated soil, and the results showed a removal of up to 85.4% of TPH compared to other additives. Bastida et al. (2016) used the biostimulation strategy for promoting the removal of up to 88% of polycyclic aromatic hydrocarbons (PAH) and alkanes in 50 days. Borah and Yadav (2016) in India reported the efficiency of *Bacillus cereus* in the degradation of 96% of kerosene making it a potential tool for the bioremediation of petroleum-based contaminants.

4.3. Microorganisms used in the bioremediation of crude oil and oil components

To date many studies have reported the ability of microorganisms to utilize crude oil and oil components as the growth substrates. Deng et al. (2016) cited the efficiency of pyrene degradation in different concentrations using the bacterium *Mycobacterium gilvum* immobilized in chemically modified peanut shell powder. Dueholm et al. (2015) demonstrated by means of a case study the degradation of aromatic hydrocarbons in mud with the application of the bioaugmentation strategy with the use of the *Pseudomonas monteilii* strain. Ortega-González et al. (2015) reported a reduction of 100% for naphthalene, 37.87% for anthracene, 25.10% for pyrene and 18.18% for fluoranthene over a period of 45 days using *Amycolatopsis* sp. for bioremediation of oil contaminated soil.

Fungi are also important organisms in the reduction of oil pollution, since they have the capacity of bioremediation due to their enzymatic activities, as well as bacteria. They are found in the roots of plants associated to phytoremediation, or in soils or water contaminated by oil (Mohsenzadeh et al., 2012). Andreoli et al. (2015) verified the efficiency of two bioremediation strategies: bioaugmentation and biostimulation, in burned soil. Using the fungus *Trichoderma* sp., bioaugmentation degraded 55% of the hydrocarbons in 60 days, and the biostimulation bio-remediated 70% of the contaminant in the same period. The authors concluded that, despite the proven efficiency of bioaugmentation, biostimulation appears to significantly reduce the time required for remediation, most likely due to increased microbial degradation through improved soil nutrient balance in burned soil.

In a field study, Mohsenzadeh et al. (2012) fungal strains were

isolated from refinery oil contaminated sites and evaluated the growth capacity and the degradation potential in a medium containing different oil concentrations (0–10% v/v). The data showed that *Aspergillus terreus* degraded 44% in the treatment containing 10% (v/v) of oil; *Acromonium* sp. degraded 50% in the treatment containing 2% (v/v) of oil; *Alternaria* sp. degraded 55% in the treatment containing 8% of the oil; *Penicillium* sp. degraded 54% in the treatment containing 8% (v/v) of oil, in a period of 90 days.

Yeasts are also known to degrade petroleum hydrocarbons due to the presence of the group of enzymes called Cytochrome P450, which have the ability to biodegrade this contaminant. According to Das and Chandran (2010), *Candida maltosa*, *Candida tropicalis*, *Yarrowia lipolytica*, and *Pichia* sp. are capable of degrading C₁₀–C₁₆ alkanes and fatty acids, and *Saccharomyces cerevisiae* has a remarkable ability to utilize petroleum as a source of carbon in aqueous solution (Hanano et al., 2015). Gargouri et al. (2015) verified the growth of two yeasts in long chain n-alkane, diesel oil and crude oil. The results showed that *Candida tropicalis* and *Trichosporon asahii* presented a degradation efficiency of TPH by about 97% and 95% respectively over a period of 20 days. The authors concluded that these yeast strains may be useful for the bioremediation process and decrease of oil pollution in contaminated wastewater.

Algae and protozoa are important members of the microbial community in aquatic and terrestrial ecosystems, but the extent of their involvement in the biodegradation of hydrocarbons is largely unknown. Chandra et al. (2013) and Das and Chandran (2010) cited a study involving *Prototheca zopfii*, capable of using petroleum as an energy source and a potential for degrading n-alkanes and iso-alkanes as well as aromatic hydrocarbons. In contrast, protozoa do not have this ability.

Individual organisms can only metabolize a limited range of hydrocarbon substrates. Therefore, it requires associations of mixed populations (consortium or guild) with different enzymatic abilities to degrade complex mixtures of hydrocarbons, such as crude oil in the soil, in freshwater, in the oceans, and in all environmental compartments (Chandra et al., 2013). Studies report the application of consortia as a strategy of bioremediation in environments polluted with oil and the results are satisfactory.

The consortium consisting of *Marinobacter* sp., *Pseudomonas* sp., *Halomonas* sp., *Hahella* sp., and *Alcanivorax* sp., bioremediation alkanes in the biostimulation strategy for oil removal in sediment (Abed et al., 2014). Brzeszcz et al. (2016) carried out a study applying the bioaugmentation strategy with *Mycobacterium frederiksbergense*, *Acinetobacter* sp., and the consortium consisting of the two microorganisms. *Acinetobacter* sp. removed 14.5% of petroleum hydrocarbons, while *Mycobacterium frederiksbergense* and the consortium removed 22.3% and 25.1% in 60 days, respectively. Through the use of n-hexadecane or diesel as the sole source of

carbon, Cai et al. (2014) identified and characterized 55 producers of marine degrading biosurfactants belonging to 8 genera, namely *Alcanivorax* sp., *Exiguobacterium* sp., *Halomonas* sp., *Rhodococcus* sp., *Bacillus* sp., *Acinetobacter* sp., *Pseudomonas* sp., and *Streptomyces* sp.

Some recent successful examples of use of consortia application for the remediation of oil and other contaminants in soil and sediments are shown in Table 3.

In some cases, organisms produce substances (biosurfactants) that are able to increase the bioavailability of the contaminant in the environment and allow the degradation action by these organisms. Biosurfactants are amphiphilic compounds secreted extracellularly that contain hydrophobic and hydrophilic portions, allowing them to accumulate between the fluid phases of an organism and thus decrease the surface and interfacial tension between the contaminant and the environment. Almansoory et al. (2015) report the use of biosurfactants produced by bacteria for the removal of gasoline in contaminated soil. The results showed a significant removal of up to 93.5% of the TPHs compared to the other additives.

The ability of the biosurfactant, produced by *Paenibacillus dendritiformis*, to desorb PAH from soils contaminated with engine oil had an efficiency of more than 96% for phenanthrene, 83% for pyrene in 5 days, and 81% for oil sludge of motor used in contaminated sands in a period of 24 h, improving its bioremediation by increasing the bioavailability of the contaminant (Bezza and Chirwa, 2015). In Russia, Ivshina et al. (2016) obtained a removal of 16%–69% of PAH with the use of a biosurfactant produced by *Rhodococcus ruber*.

França et al. (2015) isolated *Bacillus subtilis* from Brazilian mangrove with the objective of studying biosurfactant production and evaluating its functional properties and applicability to bioremediation. The use of agroindustrial residues (glycerol, sunflower oil, cheese whey and cashew) as alternative substrates for the production of biosurfactants was tested, and glycerol was the best source of carbon producing 1290 mg L⁻¹ of crude biosurfactant. The micellar critical concentration of the crude biosurfactant produced by *B. subtilis* was able to reduce water surface tension from 72 to 30 mN m⁻¹ and reduce the interfacial tension at a water/gasoline

ratio of 15 to 3 mN m⁻¹.

The use of a biosurfactant produced by the yeast *Yarrowia lipolytica* is used for the degradation of petroleum, and Csutak et al. (2015) state that the production of lipases may be related to the biodegradation of the contaminant. In Brazil, the use of the Rufisan biosurfactant, produced by the yeast *Candida lipolytica*, is used to accelerate the degradation of hydrophobic substances, capable of removing 98% of the motor oil in contaminated soil (Rufino et al., 2013).

4.4. Advantages and limitations of the rhizodegradation

As with any environmental recovery technique, rhizodegradation has advantages and limitations when it is chosen to be applied in oil contaminated environment. To date, there are many studies on the potential application of plants and microorganisms in petroleum bioremediation, and it is possible to measure the efficiency in the use of the rhizodegradation in soil contaminated with petroleum (Agnello et al., 2016; Borah and Yadav, 2016; García-Sánchez et al., 2018; Guo et al., 2017; Jia et al., 2016; Li et al., 2016; Pant et al., 2016).

Some of the advantages are: (i) rhizodegradation is easy to implement for contaminated site remediation, maintenance costs are minimal, in most cases not limited to contaminated site size and can be easily supported by plant growth; (ii) the biostimulation of the microorganisms found in the microbiota also helps to provide enough nutrients and organic matter to improve the quality of the soil to be remedied in the period of its application; (iii) plants also help to stabilize the soil due to their root structure (Lim et al., 2016); (iv) the bioavailability of the contaminant and the high rate of dissolution may increase the rate of rhizodegradation of the compound and may lead to its total or partial elimination or reduce its toxicity (Dey et al., 2016); (v) the contaminant can serve as a source of energy for the rhizospheric microbiota and cause the increase of indigenous microbial biomass (Dell'anno et al., 2009), which consequently accelerates its disappearance from the environment; (vi) the microbial variety with degrading potential may vary depending on the type of environment, increasing the range of toxic compounds to be biodegraded, as well as the use of

Table 3
Some examples of successful application of use of consortia application for the remediation of oil and other contaminants.

Type of addition/supplement specific name	Oil contaminant	Process duration	Removal efficiency (%)	Reference
Bacteria <i>Nocardia nova</i> ; yeast <i>Rhodotorula glutinis</i> var. <i>Dairensis</i> ; and fertilizer	Heavy crude oil	41 days	7.4	Trindade et al. (2005)
Bacteria <i>Pseudomonas</i> sp., <i>Gordonia alkanivorans</i> , <i>Rhodococcus erythropolis</i> , <i>Acinetobacter junii</i> , <i>Exiguobacterium aurantiacum</i> and <i>Serratia marcescens</i>	Petroleum	140 days	80.0	Liu et al. (2011)
Bacteria <i>Pseudomonas</i> sp. and <i>Achromobacter</i> sp. in association with the plants <i>Scirpus tripueter</i> , <i>Phragmites australis</i> , <i>Carex phacota</i> and <i>Sagittaria sagittifolia</i>	Diesel	60 days	80.0	Liu et al. (2012)
Bacterial consortium and nutrient mixture	Crude oil	18 months	99.9	Singh et al. (2012)
Bacteria <i>Marinobacter</i> sp., <i>Pseudomonas</i> sp., <i>Halomonas</i> sp., <i>Hahella</i> sp. and <i>Alcanivorax</i> sp.	Alkanes	88 days	67.2	Abed et al. (2014)
Bacteria <i>Pseudomonas aeruginosa</i> and <i>Acinetobacter baumannii</i> ; and organic waste (sugarcane bagasse and oil palm empty fruit bunch)	Crude oil	20 days	100.0	Hamzah et al. (2014)
Bacteria <i>Pseudomonas aeruginosa</i> and <i>Achromobacter xylosoxidans</i>	Crude oil	24 weeks	80.0	Roy et al. (2014)
Bacteria <i>Azotobacter</i> sp. and <i>Clostridium</i> sp.; yeast <i>Saccharomyces</i> sp.; fungi <i>Aspergillus</i> sp., <i>Penicillium</i> sp. and <i>Actinomycetales</i> sp.; and earthworms <i>Eisenia fetida</i> , <i>Eisenia andrei</i> and <i>Dendrobena veneta</i>	Engine oil	14 days	99.0	Chachina et al. (2015)
Bacteria <i>Staphylococcus saprophyticus</i> and <i>Serratia marcescens</i> ; and the yeast <i>Rhodotorula aurantiaca</i> and <i>Candida ernobii</i>	Diesel	7 days	69.0	Silva et al. (2015)
Fungi <i>Alternaria alternata</i> , <i>Aspergillus terreus</i> , <i>Cladosporium sphaerospermum</i> , <i>Eupenicillium hirayamae</i> and <i>Paecilomyces variotii</i>	Diesel	5 days	34.0	Ameen et al. (2016)
Bacteria <i>Mycobacterium frederiksbergense</i> and <i>Acinetobacter</i> sp.	Crude oil	60 days	25.1	Brzeszcz et al. (2016)
Bacteria <i>Burkholderia cepacia</i> , <i>Sphingomonas</i> sp. and <i>Pandoraea pnomenus</i>	Crude oil	40 days	64.4	Shen et al. (2016)

genetically modified organisms (GMO's) (Chandra et al., 2013); (vii) all the strategies of the bioremediation can be applied *in situ* (Mena et al., 2016) and the application of this biotechnological technique is viable to execute (Shekoochyan et al., 2016); and (viii) in a non-scientific context, plants are attractive for their aesthetics and widely accepted by the community.

However, rhizodegradation is a slow process, and it can only be considered as a long-term solution. According to Kim et al. (2014), it cannot be used in all contaminated sites due to limitations in biological activity. Abiotic parameters can affect the success of bioremediation, such as: type and concentration of contaminants; soil characteristics and porosity; water content and humidity; nutrient concentration; pH; temperature; oxygen availability. Type and character of plant diseases, phytotoxic resistance; types of microorganisms; contaminant tolerance; and other biological factors can affect the development of plants and consequently their microbiota. Huang et al. (2005) mention that if the plant species is unable to grow in an oil-contaminated environment, the plant would not be able to produce enough biomass to provide development of the microbe, which hinders the degradation of the contaminant.

Some microorganisms are capable of causing diseases to plants or may hinder their growth. Other factors, such as xenobiotics, may cause imbalance in the rhizosphere conditions, with both plant and microorganisms having direct and indirect consequences, and there may be no degradation of the pollutant (Sillen et al., 2015; Razavi et al., 2016). The low dissolution rate of the contaminant and/or its low bioavailability in the environment, partially or totally prevents the degradation of the pollutant; the results of its application may take months, years, or even decades to appear (Zhang et al., 2014a; Chen et al., 2015) when applied *ex situ*, the strategies of bioremediation can reach prohibitive cost, depending on the contaminated area and its extension, and, they have secondary environmental consequences with the destination of the residues of the first treatment; and in some cases environmental contamination by oil may have irreversible environmental impacts, with severe consequences for all environmental compartments (Martínez-Álvarez et al., 2015). Thus, the successful application of a bioremediation strategy for further degradation of environmental pollutants is often limited by the lack of methods to monitor the survival and activity of the organisms involved (Dueholm et al., 2015).

5. Future perspectives

Oil is the main source of world energy and is of great relevance for the development of various socioeconomic factors. It is highly abundant, the energy cost is cheap, and it is a precursor of a range of products such as fuels, fertilizers, plastics, paint, clothes, etc., essential for industrial development. However, overdependence of the human population on oil has led to the circulation of life-threatening chemicals, which are not only hazardous for human health, but can also disturb the ecological balance.

As long as there is the exploitation of oil and the use of its derivatives, there will be environmental accidents and consequently the contamination of ecosystems. Soils contaminated with organic pollutants especially with petroleum and its derivatives have attracted attention in the search for alternatives of environmental recovery. Several methods have been developed in the last decades, and although some have proven more efficiency, their limitations in the application are still reasons of doubt in the decision making, that is, which method is more suitable for the recovery of soils contaminated with petroleum.

New studies on these mechanisms will help improve strategies for the use of rhizospheric microorganisms in mediating oil toxicity tolerance. Based on the knowledge generated up to now, it is

suggested several future avenues of research approaches: (i) further studies are required to explore the effect of oil toxicity tolerant PGPR on plant biochemistry; (ii) comparative studies of interactions between plants and oil toxicity tolerant PGPR, now lacking, would shed light on the mechanisms governing abiotic stresses; (iii) further studies on oil toxicity tolerant PGPR in plant biology should be focused on making full use of the role of these PGPR and even fungi (co-inoculation of beneficial PGPR and mycorrhizal fungi) in conferring tolerance in plants against oil toxicity stress; (iv) understand the prospects of synergetic interactions of various plant-associated microbes in future climate change.

Nowadays, bioremediation, especially represented by the rhizodegradation strategy, stands out due to its sustainability, since it is a technique that unites: economic aspects, as it involves lower costs of application and operation; ecological aspects, because it allows the integration of several factors in the recovering environment; and social aspects, since it is well accepted socially by the community, as it improves the aesthetics of the contaminated site. Thus, the process that uses rhizospheric microorganisms for the remediation of environments contaminated by petroleum and its derivatives is the most effective biotechnological tool, being able to remedy the contaminate by up to 100%, reducing the residence time of the contaminant in the environment and the negative impacts. Finally, the bioremediation is an eco-friendly and economic method to control petroleum pollution.

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