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# Bioremediation of Petroleum-Contaminated Soil

*Shuisen Chen and Ming Zhong*

## Abstract

Petroleum is not only an important energy resource to boost the economic development, but also a major pollutant of the soil. The toxicity of petroleum can cause a negative impact on ecosystem, as well as the negative effects related to its carcinogenic for both animals and humans. In the present study, bioremediation as an alternative tool for restoration petroleum-contaminated soils was set forth, and focusing on the phytoremediator plants, petroleum-biodegradable microorganism are responsible for the biodegradation of petroleum. In the present chapter, the bioremediation of petroleum-contaminated soil, as well as the influence factors of bioremediation are elaborated based on the recently studies. This will provide a novel understanding on bioremediation and help improve strategies for petroleum-contaminated soils remediation.

**Keywords:** petroleum, contaminated soil, bioremediation, phytoremediation, rhizoremediation, biostimulation, bioaugmentation

## 1. Introduction

Petroleum is an important strategic resource that dominates the world economy [1]. Petroleum is composed of a complex mixture of aromatic hydrocarbons, aliphatic hydrocarbons, heterocyclic hydrocarbons, asphaltene and non-hydrocarbon compounds. And 60–90% of them are classified as biodegradable [2]. In the past decades, with the development of petroleum industry, petroleum has caused a severe environment contamination and relevant adverse effects during the exploration, transportation, management or storage of hydrocarbons in underground deposits, and refining processes, among which the soil contamination with petroleum is a serious global problem [3].

The petroleum contamination induces oxidative stress, causes the alteration in soil's chemical composition and low nutrient availability. The primary harmful effects of petroleum include inhibition of seed germination, reduction of photosynthetic pigments, slowdown of nutrient assimilation, inhibition of root growth, foliar deformation and tissue necrosis, as well as destroy biological membranes, disturb the signaling of metabolic pathways and disrupt plant roots architecture [4–7]. The low-molecular-weight hydrocarbons can penetrate plant cells resulting in plant death. In addition, the petroleum and its derivatives lead to the development of cancer and other diseases. Previous studies indicated that petroleum contamination caused the depression of the nervous system, narcosis and irritation of the mucous membranes of the eyes in humans [8–11]. In view of the high toxicity, carcinogenic, mutagenic and teratogenic potential of petroleum contamination,

the bioaccumulation of petroleum in the food chain would disturb biochemical and physiological processes which lead directly or indirectly to human health [12, 13]. Therefore, petroleum contamination is not only a negative impetus for plant growth and development but also an adverse factor for human and ecological health.

2. Technologies for petroleum-contaminated soil remediation

Faced with the serious environmental problems that involve the soil contamination by petroleum, an increasing attention has paid to the development and implementation of innovative technologies for the removal of petroleum from soil in the past decades. Multiple soil remediation technologies involve the physical remediation, chemical remediation and bioremediation were developed and employed for the restoration of petroleum-contaminated soil, particularly the eco-friendly bioremediation (Figure 1).

2.1 Physical remediation

Physical remediation uses the physical properties of the contaminants or the contaminated medium to destroy, separate, or contain the contamination, which include soil vapor extraction, flotation, ultrasonication, electro kinetics remediation, thermal desorption and biochar adsorption.

Soil vapor extraction is focus on inducing volatilization of nonaqueous-phase liquid and vapor-phase transport of volatile organic compounds form the subsurface to the surface for subsequent treatment. Soil vapor extraction is also known as in situ soil venting, in situ volatilization, enhanced volatilization, or soil vacuum extraction, in which the extraction well was used to create a pressure or concentration gradient to remove volatiles and some semivolatile contaminants from soil [14]. Soil vapor extraction can remove large quantities of volatile contaminants in uniform soils within a short time. Meanwhile, soil vapor extraction provides oxygen through the flow of air to stimulate the growth of microorganisms. However, the efficiency of soil vapor extraction is affected by soil properties and operational conditions, as well as contaminant properties [15, 16].

The flotation technology relies on the difference in surface properties of both contaminant and soil, which separate oil from soil via a gas–liquid–solid system. The flotation mechanism is dependent on (I) collision between contaminants and

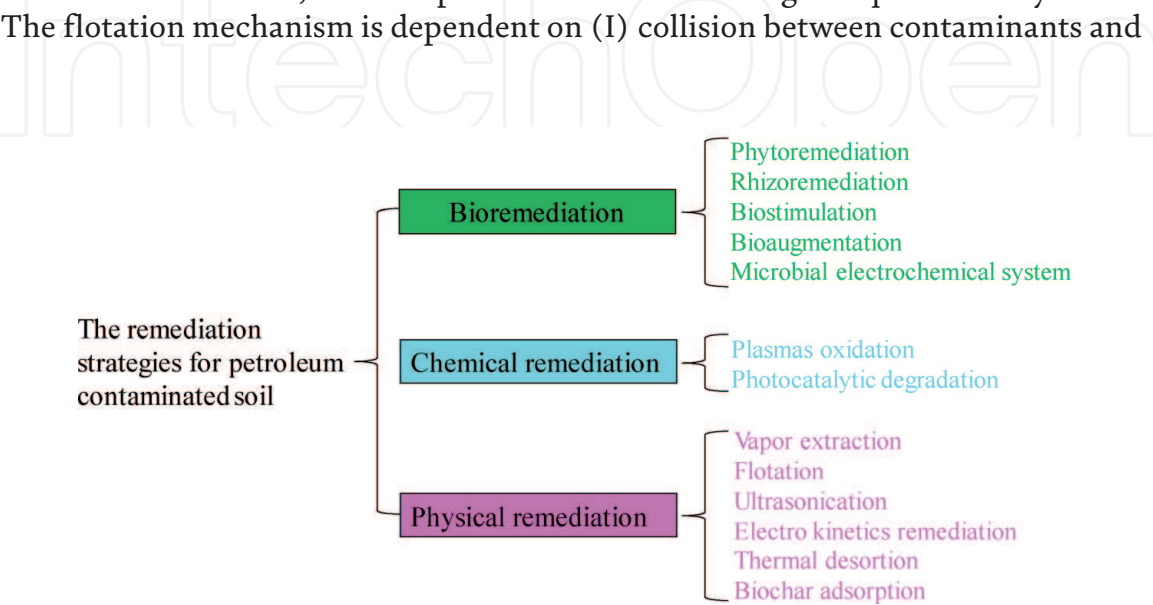


Figure 1. Schematic of the remediation strategies for petroleum-contaminated soil remediation.

bubbles, (II) form bubble-contaminant with the attachment of contaminants and bubble, (III) flotation of bubble-contaminant based on difference in buoyancy and detachment of contaminant from bubble-contaminant [17]. Flotation is characterized as simplicity, low operational cost and high efficiency for contaminants removal. It also can separate very small or light weight particles with low settling velocities. Nevertheless, large amounts of wastewater were produced during the flotation process. And the efficiency of aged or weathered contaminated soil was significantly decreased [18].

The ultrasonication helps desorption of the contaminant and promotes the formation of strong oxidant, hydroxyl radicals ( $\text{OH}\cdot$ ) which enhance the efficiency of pollutant removal [16, 19]. Ultrasonication can eliminate the hazardous pollutants without any chemicals. Moreover, the on-spot heating and intense agitation will enhance heat and mass transfer processes. However, the higher energy consumption for generation of acoustic makes it the costly setup [20].

Electro kinetics remediation employs direct electric current between appropriately distributed electrodes (cathodes and anodes) that embedded in petroleum-contaminated soil to form an electric field. The voltage potential gradient were formed in the electric field which cause the fluid medium to flow preferentially towards the cathode and drag the contaminant together with the bulk flow [21]. The advantage of electro kinetics remediation is speed of execution and low operating cost. Moreover, the electro-osmotic flow is constant through the entire soil mass during the remediation which more suitable for low permeability soils. However, the electro kinetic process is ineffective in low contaminant concentration. The alteration of soil pH and hot spots around the electrodes were induced after an extended period of time [16].

Thermal desorption based on the manipulation of temperatures to increase the vapor pressure of the contaminants, in which the contaminants were volatilized and subsequent desorption from contaminated soil [22]. Thermal desorption is very effective in destroying the oil pollutants under high heat condition. In addition, thermal desorption emits little or no contamination gas into the atmosphere. However, only the volatile contaminants were removed by thermal desorption [23].

Biochar is carbon-enriched and porous with high specific surface area and biodegradability. The biochar was employed as an amendment to implement organic contaminated soil remediation due to the surface adsorption, partition and sequestration [24, 25].

## 2.2 Chemical remediation

Chemical oxidation has the potential for rapidly deposing or preprocessing soil contaminants. Oxidants that can cause the rapid and complete chemical destruction of petroleum contaminants are employed in the chemical oxidation remediation. In chemical oxidation, the contaminants is oxidation chemically converts to non-hazardous, at least, biodegradable products or less toxic compound that more stable, less mobile or inert [16, 26].

Plasmas oxidation is also considered as highly competitive technology to remediating the pollutants from soils. Especially the plasma technology based on pulsed corona discharge and dielectric barrier discharge has aroused widely concerns in soil remediation [27, 28]. Plasma are macroscopically electrically neutral aggregates composed of numerous ions, electrons, atoms, molecules and unionized neutral particles. A number of active constituents such as  $\text{O}_3$ ,  $\text{H}_2\text{O}_2$ , the hydroxyl radicals ( $\text{OH}\cdot$ ) and high energy electrons were generated in the generation of plasma by ionization, in which a strong oxidizing environment was created for oxidative decomposition of contaminants [29].



Photocatalytic degradation is effective for decomposition of polycyclic aromatic hydrocarbon in the soils. This technology makes use of the semiconductor metal oxide as catalyst to degrading organic pollutant into small molecules directly [30]. Semiconductor molecule contains a valance band with stable energy electrons and an empty higher energy conduction band. The absorption of radiation can initiate the photocatalytic reaction, in which the formation of holes ( $h^+$ ) in valence band and electrons ( $e^-$ ) in conduction band in femtosecond time scale. During the photocatalytic process, the hydroxyl radicals ( $OH\cdot$ ) and superoxide radical anion ( $O_2^{\cdot-}$ ) are formed to degrading organic pollutant. However, the light absorption characteristics, humic substances content and moisture content of soil may affect the photocatalytic degradation [31, 32].

### 2.3 Bioremediation

The physical and chemical remediation have their own characteristics, even effective for higher contaminants removal. However, the practical applications are impracticable under some circumstances, such as the remediation of amount of contaminated soil is economically impracticable. Furthermore, most of the physical and chemical remediation technologies are unavoidable to destroy the soil microbiota that reduces the concentration of soil at the expense of damaging the integrity of soil ecosystem [33]. Therefore, alternative technologies that have less environmentally aggressive, greater ease of practical application, as well as more efficient and cost-effective for environmental decontamination are expected. In the past decades, significant advances on bioremediation have been achieved. Although it is time consuming, bioremediation techniques, due to their eco-friendly approach and very low cost, efficient and sustainable for restoring the contaminated soil in the context of sustainability, are extensively noticeable at present [34].

Bioremediation is a process that naturally or artificially take advantage of living organisms or their products to reduce (degrade, detoxify, mineralize or transform) the pollutants of the contaminated environment [35]. For this purpose, living organisms (plants and microorganisms) that tolerate and have capacity to grown under contaminated soil are usually used. Number of studies has revealed that selecting petroleum-tolerant plants for bioremediation in cases of soil petroleum pollution is a feasible and sustainable technology. Many plants, such as perennial ryegrass, alfalfa, *Mirabilis jalapa*, were considered as tolerant to petroleum stress [36–38]. The microorganisms that are utilized in petroleum pollutants removal can be bacteria, fungi or yeasts. These microbes are the essential component in soil ecological systems that play a vital role for the remediation of petroleum hydrocarbon and other pollutants [39, 40]. Some of them have high capacity to degrade contaminants and widely used for environmental depollution [41]. In the bioremediation of petroleum contaminated soils, the most widely used organisms are bacteria which have high frequency, rapid growth and a broad spectrum of degradation of petroleum products [42].

Phytoremediation is a kind of bioremediation. Phytoremediation is considered as an alternative technology that makes use of plants and microorganisms associated with their root to degrade or reduce soil contaminants [43]. The main factors to consider when choosing a plant as a phytoremediator are root system, plant survival and its adaptability to prevailing environmental conditions. Plants not only can degrade petroleum pollutants directly via enzymatic activities, but also can stimulate the rhizosphere microbial community to degrade petroleum contaminants [44]. The use of plant growth promoting rhizobacteria (PGRR) plays an important role in phytoremediation. The PGRR promote the growth of plant by providing phytohormones and mineral nutrition. PGRR can also generate antibiotics, compete for

nutrients with pathogens or induce systemic resistance in the host plant to protect it from pathogens [45]. To understand the interactions between PGRR and plants would be better reveal the alleviation of contaminants toxicity of soil.

Rhizoremediation is a strategy for phytoremediation, plant act indirectly in phytoremediation, since their presence in the environment provides favorable conditions for the growth of microorganisms in the rhizosphere region. The rhizosphere is a soil zone ranging from the surface to a depth of 1–5 mm, in which the interdependence between plants and microorganisms result in a symbiotic lead to form a symbiotic relationship [46]. Plants roots can release the organic acids, carbohydrates, amino acid and oxygen to the rhizosphere, which promote the development of rhizosphere microbe (including bacteria, fungi, protists, nematodes and in vertebrates). And the microbe benefits the plant by providing the necessary vitamins, cytokinins and amino acids to promote plant growth [16]. The development of microbial biotechnology is beneficial for screening and identifying microorganisms from petroleum contaminated soils [47]. Many microorganisms have been isolated and utilized as biodegraders for petroleum hydrocarbons disposal. More than 79 genera of bacteria that capable of degrading petroleum hydrocarbons have been identified [48]. Furthermore, some microorganisms were crucial for petroleum hydrocarbons since the abundance of these microorganisms were dominant increased after petroleum contamination [49]. In view of different indigenous bacteria have different catalytic enzymes, the combination of multiple functional bacteria were preferable to remediating the pollutants in contaminated soils. Previous studies showed that the joint action of indigenous bacterial consortium and exogenous bacteria were effectively accelerating the degradation of petroleum [50].

Biostimulation is one of the main strategy bioremediation for the decontamination of petroleum-polluted soil, which through adjusting the environmental conditions (temperature, moisture, pH, redox potential, aeration, mineral nutrition) to enhance the growth and the metabolic activity of indigenous degrading microbial populations. The microorganisms' activity in biostimulation practice is tolerant to various hydrocarbons and can utilized hydrocarbons as carbon sources for their growth [51].

Bioaugmentation is another strategy of bioremediation, which refer to the inoculation of exogenous microorganisms into the contaminated soils to degrade the target contaminants [52]. The inoculated microorganism can be one strain or a consortium of microbial strains with diverse functional degradation capacities [53]. Bioaugmentation was considered to be more effective for the degradation of the light fraction (C<sub>12</sub>-C<sub>23</sub>) of petroleum hydrocarbons [54]. Bioaugmentation can divided into cell bioaugmentation and genetic bioaugmentation based on the degradation mechanism of the inoculated strains. Cell bioaugmentation relies on the survival and catabolic activity of the inoculated strains to accelerate the degradation of target contaminants directly [55]. While genetic bioaugmentation based on the spread of catabolic genes (plasmids, integrons or transposons mediated) into native microbial populations. And then the native acquiring these genes achieve the ability to degrade organic contaminants [56]. As compare to the cell bioaugmentation, genetic bioaugmentation, especially plasmid-mediated bioaugmentation appears to have greater potential for the bioremediation of contaminated soils [57].

In addition, microbial electrochemical system was considered as an emerging technique for bioremediation, which integrates microbial and electrochemical processes to convert the pollutants to less-toxic or value-added products [58]. With various inherent advantages, microbial electrochemical system was mostly applied in remediation of petroleum contaminants in soil. Microbial electrochemical system

is considered as more flexible for various contaminants in bioremediation due to the oxidation and reduction transformation in remediation processes [59].

### **3. Influence factors of bioremediation**

Bioremediation is viewed as a technique to accelerate the natural biodegradation process in a cost-effective and environment friendly way. However, bioremediation is time consuming, and the contaminant concentration and composition, temperature, soil pH, oxygen condition and salinity are highly affected the bioremediation of the petroleum-contaminated soils.

The plants and microbes would unable to grow in a high petroleum soil. In that case, the bioremediation was inoperative or low efficiency. In addition, some of the petroleum derivatives with high solubility have higher cytotoxicity to biodegradation bacterial, while other compounds produced no significant inhibitory effects on bacterial growth [60].

The temperature plays a vital role in bioremediation and influence biodegradation reactions [61]. Indeed, temperature can indirectly affecting biodegradation efficiency by affecting bacterial growth and metabolism, altering soil matrix and the mode of occurrence of pollutants [62].

Petroleum and its derivatives can full fill the interstices of soil, which reduce the amount of oxygen in soils. Under reduced or absent oxygen conditions, the metabolism of aerobic microorganisms was partially interrupted, as well as the bioavailability and degradation efficiency of pollutant were reduced [12, 63].

The key components of biodegradation of petroleum hydrocarbons are various specific enzymes [61]. The alterations of pH value may influence the enzymes activities to reduce the effective of the biodegradation. In other hand, alterations of pH and high salinity may inhibit microbial growth and metabolism. In addition, the lack of technique to monitor the survival and activity of the organism in soil also limited the application of bioremediation.

### **4. Conclusions**

Bioremediation is an eco-friendly and economic method to remove the petroleum pollutants of soil. There were several kinds of bioremediation have been applied to remediate the petroleum hydrocarbon from contaminated soils, such as phytoremediation, rhizoremediation, biostimulation, bioaugmentation, and so on. The petroleum-tolerant plants and the high-effective petroleum degradation microbes are preferable used in bioremediation process. Alternatively, construction of the different functional bacterial consortium or genetic engineering bacteria, and potentially using integrated bioremediation approaches for bioremediation of petroleum have become a trend in future. However, some of the influence factors are significant reduced the degradation efficiency on bioremediation application. Therefore, use a combination of bioremediation and other technologies is effective strategy to accelerate for petroleum hydrocarbon pollutants removal. Furthermore, developing and enriching the novel technologies of bioremediation remains far off.

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## Conflict of interest

The author declares no conflict of interest.

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