



Research Progress on Remediation Technology for Polycyclic Aromatic Hydrocarbons (PAHs) Pollution

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ABSTRACT

Polycyclic aromatic hydrocarbons (PAHs) are a class of persistent organic pollutants (POPs) widely distributed in the environment, with carcinogenic, teratogenic and mutagenic properties, posing a serious threat to ecosystems and human health. This study provides a systematic review of the structural characteristics of PAHs, their sources and their environmental hazards, and focuses on the principles, applications and limitations of several remediation technologies. Among them, microbial remediation has become a research hotspot due to its high efficiency and environmental friendliness. The article further focuses on the research progress of microbial degradation of PAHs, and discusses the current research status of strain resources, microbial metabolic pathways and key degradation genes. To provide theoretical basis and technical reference for PAHs pollution control.

KEYWORDS

PAHs, Pollution remediation technologies, Microbial degradation.

1. INTRODUCTION

Polycyclic Aromatic Hydrocarbons (PAHs) are hydrocarbons that contain two or more benzene rings in their molecular structure, generally consisting of linear, angular, or clustered arrangements of densely packed aromatic rings[1-3]. It is widely distributed in the environment of soil, atmosphere, groundwater, and sediment[4], and is a typical Persistent Organic Pollutants (POPs) that cause environmental pollution[5]. The presence of PAHs in the environment is a serious hazard to human health due to their strong carcinogenicity. As early as 1979, the United States Environmental Protection Agency (the U.S. EPA) published 16 priority PAHs control[6]. It has attracted much attention and in-depth research worldwide. Therefore, how to safely and effectively remediate PAH-contaminated water bodies and soil environments and reduce ecological risks is now a common focus of attention in related fields and industries.

2. CHARACTERISTICS OF PAHS

2.1. Structure and properties of PAHs

Based on the number of benzene rings they can be classified into two categories: One group of low molecular weight PAHs, containing three or less benzene rings, such as naphthalene, phenanthrene and anthracene; One category is high molecular weight PAHs with four or more benzene rings, such as pyrene, fluorescent onion, benzo(alpha)pyrene[7]. The difference in molecular weight of PAHs and the difference in physicochemical properties of the substances lead to the difference in their toxicity and environmental hazards, and the number of benzene rings of PAHs is directly proportional

to their toxicity. The higher the number of benzene rings, the higher the chemical stability, persistence, resistance to biodegradation, genotoxicity and carcinogenicity, which increase with time[8, 9]. Thus, high molecular weight PAHs have higher toxicity relative to low molecular weight PAHs.

In addition, low molecular weight PAHs, due to their structural instability, are distributed in the atmosphere in the form of tiny particles that migrate with atmospheric movement; High molecular weight PAHs are chemically stable, adsorbed to soil and sediment, and partially dissolved in water. The physicochemical properties of PAHs allow for rapid mobility in the environment, making them widely distributed in air, soil, aqueous environments, sediments, terrestrial water, and groundwater[1, 10, 11].

2.2. Sources and hazards of PAHs

PAHs, as a representative, difficult to degrade and highly toxic organic pollutant, are mainly products of incomplete combustion and thermal decomposition of hydrocarbon-containing compounds in reducing atmospheres, and their sources are also very extensive. PAHs commonly detected in the environment can be categorized into two main groups, natural and anthropogenic sources, depending on the source of production. Natural sources mainly include volcanic activity, oil and gas spills, forest fires, and synthesis in living organisms; anthropogenic sources mainly include combustion of fossil fuels such as coal and oil[12], as well as emissions from automobile exhausts, industrial exhausts, and industrial wastewater[12-14]. Since PAHs come from a variety of sources, the interaction between natural and anthropogenic sources has led to a gradual increase in their quantity, resulting in their wider distribution worldwide, and the pollution of PAHs has become more and more serious.

PAHs have high toxicity, and the hazards to organisms and the environment are mainly reflected in their toxic effects on organisms and impacts on the ecosystem. Numerous studies have shown that PAHs are potentially harmful to ecology and human health due to their bioconcentration and "triple" (i.e., mutagenicity, teratogenicity, and carcinogenicity) effects. PAHs can enter the human body through the skin, respiratory tract and digestive tract, causing various hazards to the human body, such as damage to the respiratory system, circulatory system, nervous system, damage to the liver and kidney[15], and may even cause DNA damage, mutations and chromosomal aberrations, increasing the risk of carcinogenesis[16, 17]. Secondly, ecologically, PAHs are highly toxic and persistent and can cause damage to ecosystems [18]. In water bodies, PAHs can affect the growth and reproduction of aquatic organisms and accumulate in aquatic organisms, causing damage to their biofilms and damage to aquatic ecosystems. In soil, PAHs reduce microbial diversity and abundance as well as activity [18] while inhibiting plant growth [19, 20] and affecting soil quality and fertility [21]. In the atmosphere, volatilization and oxidation reactions of PAHs produce photochemical pollutants that cause serious pollution of the atmosphere.

3. RESEARCH PROGRESS ON PAHS REMEDIATION TECHNOLOGIES

3.1. Physical remediation

Remediation technologies in environments contaminated with PAHs include mainly physical, chemical and biological methods [22-24].

There are more methods of physical remediation, among which the more mature technology is thermodynamic remediation, i.e., thermal desorption technology, which involves heating the soil through an external heat source to volatilize the pollutants from the soil, and then concentrating on the exhaust gas to achieve the purpose of purifying the soil. It has been shown that high-temperature (>300°C) treatment can be highly effective in removing high-ring PAHs, but energy consumption is high, and high temperature also damages the soil structure; low-temperature (50-100°C) combined with microbial remediation can be considered, which ensures highly effective degradation and also

reduces energy consumption [25]. However, in addition to high energy consumption, thermal desorption technology also has problems such as higher equipment prices, higher treatment costs, and the risk of secondary pollution, which restricts its application in large-scale and large-scale environmental remediation.

3.2. Chemical remediation

Chemical remediation means mainly include chemical drenching and chemical oxidation.

Chemical drenching usually refers to the use of drenching agent to drench contaminated soil, through the dissolution or solubilization of pollutants in the soil PAHs, pollutants with the drenching agent outflow, and then the drenching agent that is the subsequent treatment of the technology. The research of this method focuses on the development and selection of drenching agents, mainly surfactants. For example, Barbati [26] et al. drenched PAHs in marine sediments with synthetic and natural surfactants (e.g., Triton X-100, rhamnolipids), and the surfactants increased the bioavailability of PAHs through solubilizing effect and reduced the adsorption of contaminants to soil particles, and the drenching efficiency was 30 times higher than that of water drenching alone. Zhu[27] et al. used a surfactant foam drenching of PAHs-contaminated soil with a mixture of rhamnolipid and humic acid, and the removal of naphthalene and phenanthrene reached 60.1% and 56.68%, respectively. However, chemical drenching still has many application limitations. Surfactant residues may be toxic to soil microorganisms and plants, thus posing the risk of secondary pollution; high-temperature drenching requires a large amount of chemical reagents, which is less economical.

Chemical oxidation technology, on the other hand, is a technology that degrades PAHs and reduces their toxicity by introducing oxidizing agents that cause oxidative reactions and the oxidative decomposition of pollutants in the soil into harmless substances. Commonly used chemical oxidizing agents include hydrogen peroxide, permanganate, TiO₂, Fenton's reagent and activated persulfate[28]. Iron-based/biochar-activated persulfate (e.g. Fe-Ni/AC) generates sulfate radicals with degradation efficiencies of 80%-96% for HAPHs[23]. Degradation of PAHs ranging from 84.7% to 99.9% using hydrogen peroxide and Fenton (H₂O₂/Fe²⁺) systems[29]. Rotondo et al[30] showed much the same, with the Fenton system generating hydroxyl radicals under acidic conditions (pH 2-4) and degrading low-ring PAHs (e.g., phenanthrene) with 70-85% efficiency. Chemical oxidation is more efficient and broad-spectrum compared to other treatments: it is applicable to a wide range of PAHs and complex contaminated soils. However, more toxic by-products such as quinones or environmentally persistent free radicals (EPFRs) may be generated[31]. Also higher cost and energy consumption.

3.3. Phytoremediation

Phytoremediation of PAHs is a method that reduces or removes PAHs by utilizing the biological properties of plants such as uptake, transport and metabolism. Plants of the same type differ in their ability to absorb PAHs and their treatment effects, representative of which include alfalfa[32], ryegrass [33], Sambucus cannablis[34], and bitterbrush [35]. Phytoremediation technology has the advantages of green low-carbon, economic efficiency, stable effect and safety of the remediation process, etc. It has great potential to be applied in the soil remediation industry, but it is greatly affected by external conditions. The effectiveness of plant degradation of PAHs was influenced by environmental factors such as derived nutrients, temperature, light, and salinity value. Meanwhile, the remediation cycle is long and not applicable to high concentration of polluted soil.

3.4. Zootechnical remediation

Zootechnical remediation is the process of utilizing soil animals to directly absorb, transform, or decompose contaminants in the soil. Devi [36] et al. found that earthworms improved soil aeration, microbial migration and metabolism through burrowing, and increased the removal of 4-6 cyclic

PAHs by 30%-50%. The results of Huang et al [37] and Pan et al [38] similarly illustrated that the activity of earthworms can promote the removal of PAHs. In addition, small soil animals such as nematodes and hoppers can accelerate the contact between PAHs and microorganisms through feeding, which indirectly promotes microbial degradation, but the remediation efficiency is relatively low, and it is mostly used as an auxiliary means [39].

3.5. Microbial remediation

Microbial remediation is the process of reducing the activity of harmful substances in soil or degrading them into harmless substances by utilizing the metabolic action of microorganisms with specific functions under suitable environmental conditions.

Bioaugmentation (biological enhancement) is the inoculation of a contaminated site with highly efficient degrading strains or colonies of bacteria for the purpose of degradation. Efficient degrading strains, which may be functional strains enriched and screened from contaminated sites, or genetically engineered bacteria with degrading functions obtained through gene editing [40]. In bio-enhanced remediation of PAH-contaminated soil or wastewater applications, the role of microorganisms as the dominant players in the remediation process is crucial. Biostimulation refers to the use of physicochemical means, such as the artificial addition of nutrients or electron acceptors such as H₂O₂, O₂, NO₃⁻, etc., to PAH contaminated sites to stimulate the growth and reproduction of indigenous microorganisms, enhance their activity and degradation functions, so as to achieve the purpose of removing PAH and remediation of contaminated sites [41, 42]. For example, some researchers have been able to achieve more than 98% degradation of Fe by changing physical and chemical conditions such as aeration, carbon, nitrogen and phosphorus ratio, mixing speed, rhamnolipid. Yang [43] et al. showed that the addition of nitrate (NO₃⁻), which promotes anaerobic metabolism in *Bacillus*, could increase the phenanthrene degradation rate from 38.7% to 93.9%.

4. PROGRESS IN RESEARCH ON MICROBIAL DEGRADATION OF PAHS

4.1. Current status of research on PAH-degrading bacterial strains

Currently, microbial remediation technology is quite mature, and has become a hotspot for the remediation of PAH pollution in recent years, and bio-enhanced technology, i.e., the introduction of functional bacterial strains or flora at the contaminated site is the main. Whereas the screening of efficient degrading microorganisms has been the research focus of microbial-enhanced remediation technology, microorganisms that can degrade PAHs in nature include bacteria, fungi [44] and algae [45]. Microbial species differ in the carbon and energy sources available to them. As summarized in Table 1, some of the PAH-degrading microorganisms reported in recent years' studies were summarized. It can be seen that bacteria are the most reported microorganisms in PAHs degradation studies. Among them, the largest number of strain species were involved in the phylum Actinobacteria, Ascomycetes. This is similar to the findings of Huang [46] et al. They analyzed the genomes of 95 reported degrading bacterial strains and showed that 95% of the degrading bacteria belonged to the phyla Proteobacteria and Actinobacteria, with the most abundant genera being *Pseudomonas* (14%), *Rhodococcus* (10%), *Mycobacterium* (9%), *Sphingobium* (7%).

Table 1 Summary of microbial resources for PAHs degradation

Strain	Source	Degradable contaminants	Reference
<i>Achromobacter xylosoxidans</i> IITR150	Oil sludge treatment plant	Naphthalene, phenanthrene, anthracene, benzo[α]pyrene, benzo[α]anthracene, fluoranthene	[47]
<i>Acidovorax</i> sp. strain NA3	Contaminated soil	Naphthalene, phenanthrene, quat, benzo[α]anthracene, benzo[α]pyrene.	[48]
<i>Agromyces</i> sp. PyB-10	Paddy soil	Pyrene, naphthalene, phenanthrene, anthracene, fluoranthene	[49]
<i>Alcaligenes ammonioxydans</i> VITRPS2	Oil contaminated soil	Naphthalene	[50]
<i>Arthrobacter</i> sp. P1-1	Contaminated site	Phenanthrene	[51]
<i>Bacillus</i> sp. PAH-2	Marine environment contaminated by oil spills	Benzo[α]anthracene, pyrene, benzo[α]pyrene	[52]
<i>Bacillus</i> sp. PyB-9	Paddy soil	Pyrene, naphthalene, phenanthrene, anthracene, fluoranthene	[49]
<i>Brevibacillus</i> sp. PDM-3	Sludge	Phenanthrene, anthracene, fluorene	[53]
<i>Fusarium</i> sp. H-H2	Contaminated soil from coal mining areas	Pyrene, benzo[α]pyrene	[54]
<i>Klebsiella michiganensis</i> EF	\	Phenanthrene, pyrene	[55]
<i>Mycobacterium</i> sp. 16F	\	Pyrene	[56]
<i>Mycobacterium</i> <i>austroafricanum</i> . MYC038	\	Pyrene	[57]
<i>Pelagerythrobacter</i> sp. N7	Mixed saline soils	Phenanthrene, naphthalene	[58]
<i>Phlebia acerina</i> S- LWZ20190614-6	Sediment	Benzo[α]pyrene	[59]
<i>Pigmentiphaga kullae</i> strain KIT-003	Nakdong River, Korea	Benzo[α]pyrene	[60]
<i>Pseudomonas aeruginosa</i> NG4	Oil contaminated area	Naphthalene, enthrone	[61]
<i>Pseudomonas fluorescens</i> S01	Crude oil contaminated sludge	Mixed PAHs and Heterocyclic PAHs	[62]
<i>Pseudomonas xizangensis</i> S4	Highland lake sediments	Fluoranthene	[63]
<i>Rhodococcus rhodochrous</i> ATCC 21198	\	Fluorene, phenanthrene, anthracene, pyrene	[64]
<i>Rhodococcus ruber</i> L9	\	Pyrene	[65]
<i>Sarocladium terricola</i> RCEF778	Sedimentation in the vicinity of petrochemical plants	Pyrene	[66]

Shigella sp. PyB-6	paddy soil	Pyrene, naphthalene, phenanthrene, anthracene, fluoranthene	[49]
Sphingobium sp. PM1B	contaminated soil	Phenanthrene	[67]
Sphingobium sp. SJ10-10	Abandoned coking plant	Phenanthrene	[68]

4.2. Microbial degradation pathways of PAHs

Existing studies have been relatively thorough on the degradation pathways of PAHs degradation by microorganisms such as bacteria, fungi and algae [69]. The process can be categorized into aerobic degradation and anaerobic degradation based on whether oxygen is required for the process. Although the metabolites generated during the degradation of different types of PAHs by different types of microorganisms may be slightly different, the intermediates and degradation pathways can be summarized to arrive at a universal mechanism, and the specific metabolic pathways are shown in Figure 1.

Bacterial degradation of PAHs is mainly through aerobic metabolism, where PAHs are first formed into dihydrodiol products under the action of an initial dioxygenase, and then metabolized to catechols under the action of dehydrogenase; this is then followed by ring-opening cleavage through the meso or adjoining sites, and so on, with gradual ring-opening [69, 70]. Aerobic degradation of PAHs by fungi mainly by means of cytochrome P-450 monooxygenase and lignin peroxidase. Cytochrome P450 monooxygenase catalyzes the generation of monooxygenated products from PAHs, which are then isomerized to form phenols and concatenated with sulfate, glucose, or xylose, or hydrated with epoxidase to form trans-dihydrodiols[69, 71]. Some bacteria also use this route. In contrast, lignin-degrading enzymes act by generating free radicals intracellularly, which are then used to oxidize PAHs to form quinone metabolites that are cleaved by ring opening and thus mineralized[72, 73]. Algae metabolize PAHs similarly to both bacterial dioxygenase degradation and cytochrome P450 monooxygenase degradation pathways. In contrast, under anaerobic or anoxic conditions, sulfate, nitrate, or metal ions are used as electron acceptors for respiration, resulting in the degradation of PAHs to low molecular weight substances[74].

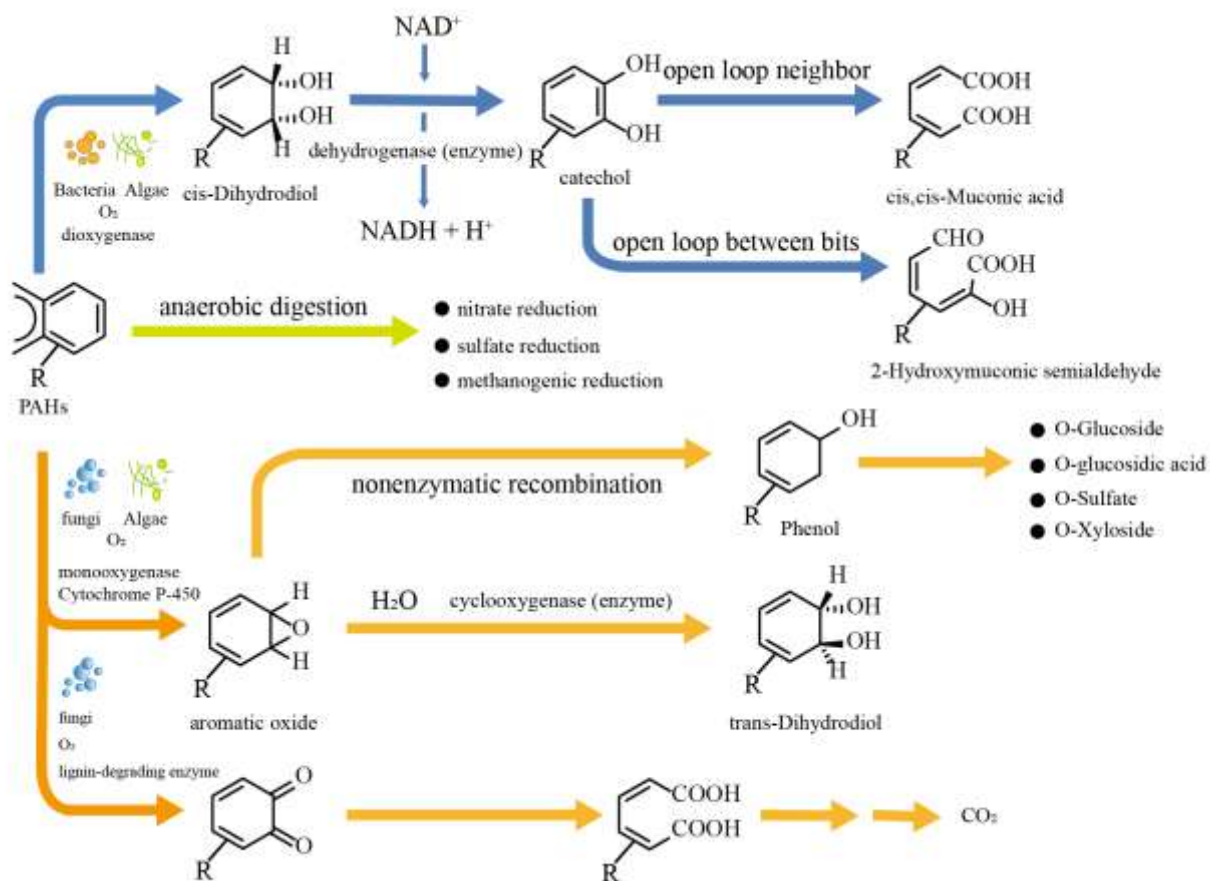


Figure 1 Mechanism of action of microbial degradation of PAHs

4.3. Microbial degradation genes for PAHs

Microorganisms activate specific degradation genes and synthesize corresponding degradation enzymes, triggering a series of metabolic processes that lead to the transformation and degradation of PAHs. The degradation of PAHs begins with the hydroxylation of the benzene ring by hydroxylated dioxygenase, so the multicomponent hydroxylated dioxygenase system is the key enzyme and rate-limiting enzyme in the whole degradation process, and based on the study of the structure and function of the gene, the genes or clusters of genes of the upstream and downstream degradation pathways of PAHs in many degrading strains have been discovered, localized and studied[75].

The first PAHs degradation genes identified were from the nah gene cluster on the *Pseudomonas putida* G7 large plasmid NAH7 in the order nahAa Ab Ac Ad B F C Q E D[76]. Mainly responsible for coding the enzymes required for the upstream degradation pathway of naphthalene. Genes at different sites in the nah-like gene cluster encode different enzyme families involved in the action of degrading bacteria, including ndo, dox, pah, and pdo genes[77].

In addition to this, there are many degradation genes in PAHs-degrading strains that are functionally identical to the classical nah-like genes but do not share homology[78]. For example, phn genes, phd genes, nid genes, nag genes, etc., these genes are more diverse and widely distributed, and the genes are not arranged in the same order. This also provides a glimpse into the specificity of different strains in PAHs degradation mechanisms and the diversity of PAHs genes[75].

5. CONCLUSION

The environmental pollution control of PAHs is one of the core issues in the field of environmental science and engineering at present. In this paper, the physicochemical properties and pollution sources of PAHs are summarized, and their environmental persistence and biotoxicity mechanisms are revealed. In terms of remediation technology, physical and chemical methods can quickly reduce the concentration of pollutants, but there are problems such as high cost and easy to produce secondary pollution. Plant and animal remediation technologies are limited by bottlenecks such as environmental adaptability and long remediation cycles, while microbial remediation technologies show significant advantages due to their degradation specificity and sustainability. Studies on microbial degradation of PAHs have shown that degrading strains mineralize PAHs into non-toxic small molecules through oxidation and hydroxylation, and their metabolic processes are precisely controlled by key genes and regulatory networks.

However, the mechanism of microbial community interactions under compound pollution conditions, the efficiency of heterologous expression of degradation genes, and the stability in practical engineering applications are still difficult points to be broken. Future research needs to combine multi-omics technology, bio-enhancement strategies and artificial intelligence-assisted design to construct an efficient and multi-functional degradation system, and promote the transformation of PAHs pollution remediation from laboratory research to large-scale application.

CONFLICTS OF INTEREST

We have no conflict of interest.

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