

Review

Mycoremediation as a Potentially Promising Technology: Current Status and Prospects—A Review

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Abstract: Global environmental pollutants are becoming intense because of the increasing human population, urbanisation, and industrialisation. Human health and the ecosystem are affected by soil and water contamination. Therefore, creating strategies is essential to tackle this persistent issue. In the process, the health and environmental risk associated with these pollutants can be significantly reduced. Previously, traditional remediation techniques have been employed in combating these environmental pollutants, proving ineffective. Mycoremediation, which uses fungi or their compounds to remediate environmental pollutants, has shown to be a cost-efficient, environmentally friendly, and effective method of environmental remediation that includes organic, inorganic, and emerging contaminants (antibiotics, pharmaceuticals). This review provides an overview of various mycoremediation approaches through fungi for biosorption, precipitation, biotransformation, and sequestration of environmental pollutants. In addition, the removal of metals, persistent organic pollutants, and other emerging contaminants by mycoremediation was highlighted. For example, fungi such as *Pleurotus dryinus*, *Trametes hirsuta* MK640786, and *Aspergillus niger* shows 91%, 94%, and 98.4% degradation of pollutants ranging from pesticides to azo dyes, respectively. Furthermore, prospects of mycoremediation to remove heavy metals and emerging pollutants from waters and soils were discussed. It was elucidated that fungi have great potential for the mycoremediation of emerging pollutants such as heavy metals, pharmaceuticals, polycyclic aromatic hydrocarbons (PAHs), pesticides, and weedicides. The findings suggested a knowledge gap exists to enhance the rate of the mycoremediation process. Therefore, a possible framework of mycoremediation was proposed to facilitate this promising technology for rectifying global environmental problems. For mycoremediation procedures to be as effective as possible, further studies are needed on fungal enzymes' role, activities, and regulation.

Keywords: Remediation; mycoremediation; fungi; environment; pollutants; heavy metals



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1. Introduction

Hazardous contaminants have a severe and challenging impact on the environment nowadays. The range of pollutants released into the atmosphere by natural processes and human economic activity is extensive. It includes several classes of compounds (polycyclic aromatic hydrocarbons (PAHs), synthetic dyes, detergents, chlorinated compounds, plastics, dioxins, pharmaceutical compounds, etc.) and the mixtures that they produce (creosote, oil, etc.). Leakage of contaminants and accidental discharge into the environment are

serious problems, especially when the biodegradation capacity of the natural microbiome is insufficient to remove or neutralise the pollutants. The physiology of cells, the composition of microbial cell walls, and physicochemical parameters, including temperature, pH, metal concentration, ionic strength, and time all play a role in the complex process of microbial metal remediation [1]. Metals, unlike organic contaminants, are not naturally destroyed, so their retention duration in the soil is measured in thousands of years. They undergo oxidation changes or complex organic transformations and remain in the soil [2].

Microorganisms can remove metals from aqueous solutions. The term for this phenomenon is biosorption. In recent research, mycosorption is defined as the biosorption of fungi [3]. Mycosorption results from the attraction of functional groups on the fungal cell wall and dyes, which occurs through a physical or chemical process and depends on the fungal biomass [4]. Mycosorption is a subject of significant interest to scientists all around the world [5]. Fungi have the ecological and biochemical ability to break down organic compounds in the environment and reduce the risk of radionuclides, metalloids, and metals through chemical modification or by affecting chemical bioavailability. In addition, these fungi are ideal for bioremediation due to their ability to form large mycelial networks; their catabolic enzymes are not highly selective, and they do not depend on pollutants as a growth substrate [6]. Most fungi can break down numerous pollutants due to their ability to synthesize extracellular enzymes to digest complex carbohydrates without prior hydrolysis [7].

Mycoremediation is a bioremediation subset that employs fungi to degrade, re-store, and heal contaminated ecosystems [8–10]. Mycoremediation uses fungi for bioremediation, and the long threads (hyphae) attach to roots, rocks, and soil particles, forming a filamentous body known to tolerate heavy metals and adapt its growth to extremes of temperature, pH, and nutrition. Bioremediation of contaminated environments favours fungi over bacteria due to the distinctive qualities of their hyphal network, biomass, and extended lifecycle. Furthermore, metal-resistant fungi compete with native bacteria under severe environments [11]. The extensive metabolic capacity of fungi enables numerous applications for eliminating various contaminants. The cell walls of fungi contain polysaccharides and proteins with amino, phosphate, hydroxyl, sulfate, and carboxyl groups that bind metal ions [12]. These functional groups provide the ligand atoms necessary to form complexes with metal ions, which attract and retain metals in the biomass. Metal removal potential is evaluated by selecting metal-tolerant fungi from a polluted environment. Bioaugmentation of potentially metal-adsorbing fungi can be an effective site-specific bioremediation technique. Several researchers have also noted that to develop effective bioremediation techniques that can maintain high metal concentrations and extract metals from the environment; research must be conducted on wild-type fungal strains [13,14].

Recent reviews have focused on the mycoremediation of petroleum-contaminated soils, heavy metals, and hydrocarbons, the parameters that influence these processes, and their broad applications. This article provides an overview of methods, prospects, and future perspectives of mycoremediation for removing inorganic and organic pollutants from contaminated land surfaces and water bodies. The role of various fungi in the degradation of pollutants such as PAHs, pharmaceutical and agricultural waste, and heavy metals was also investigated. The application of myco-nanotechnology is also recommended as a potential future initiative to enhance the effectiveness and rate of mycoremediation. Furthermore, this review provided insight into some fungal strains' mycoremediation/degradation pathways. This study also highlights the environmental degradation environment (aerobic and anaerobic) of the fungal alongside the metabolic pathway of the pollutants. To the best of our knowledge, this area has received little or no exposition in previous reviews on mycoremediation. Therefore, this study provides information on the current state of mycoremediation research that will guide future researchers on current trends in mycoremediation.

2. Classification of Fungi, Bacteria, and Microalgae Species and Their Remediation Performance

It is crucial to understand the biotransformation and biodegradation of hazardous and toxic substances interacting with ecological diversity and its behaviour [15]. Despite the rich ecology of the 69,000 fungi found globally, only a few species have been associated with mycoremediation (Figure 1) [16]. Various principal genera of fungi, bacteria, microalgae, and fungi involved in bioremediation are highlighted in Table 1. The biotic and abiotic variables, including moisture, aeration, temperature, metal ion concentration, phosphorus availability, nitrogen, carbon, and the presence and development of fungi, are caused by interspecific microbial competition [17]. More research is required on the fungal ecology related to mycoremediation, but literature is abundant on fungal ecology.

Table 1. List of the principal genera of fungi, microalgae, and bacteria used in bioremediation.

Organism	Genus	Ref.
Fungi	<i>Cryptococcus</i>	[18]
	<i>Trichoderma</i>	[19]
	<i>Rhizopus</i>	[20]
	<i>Penicillium</i>	[21]
	<i>Mucor</i>	[22]
	<i>Lasioidiplodia</i>	[23]
	<i>Fusarium</i>	[24]
	<i>Drechslera</i>	[24]
	<i>Curvularia</i>	[25]
	<i>Aspergillus</i>	[26]
Microalgae/Cyanobacteria	<i>Selenastrum</i>	[27]
	<i>Nannochloropsis</i>	[28]
	<i>Synechocystis</i>	[29]
	<i>Chlorococcum</i>	[30]
	<i>Oscillatoria</i>	[31]
	<i>Scenedesmus</i>	[32]
	<i>Spirogyra</i>	[33]
	<i>Chlorella</i>	[34]
	<i>Spirulina</i>	[35]
Bacteria	<i>Marinobacter</i>	[28]
	<i>Oleispira</i>	[36]
	<i>Cycloclasticus</i>	[37]
	<i>Thalassolituus</i>	[38]
	<i>Alcanivorax</i>	[39]
	<i>Pseudomonas</i>	[40]
	<i>Flavobacterium</i>	[41]
	<i>Enterobacter</i>	[42]
	<i>Bacillus</i>	[43]
	<i>Alcaligenes</i>	[44]

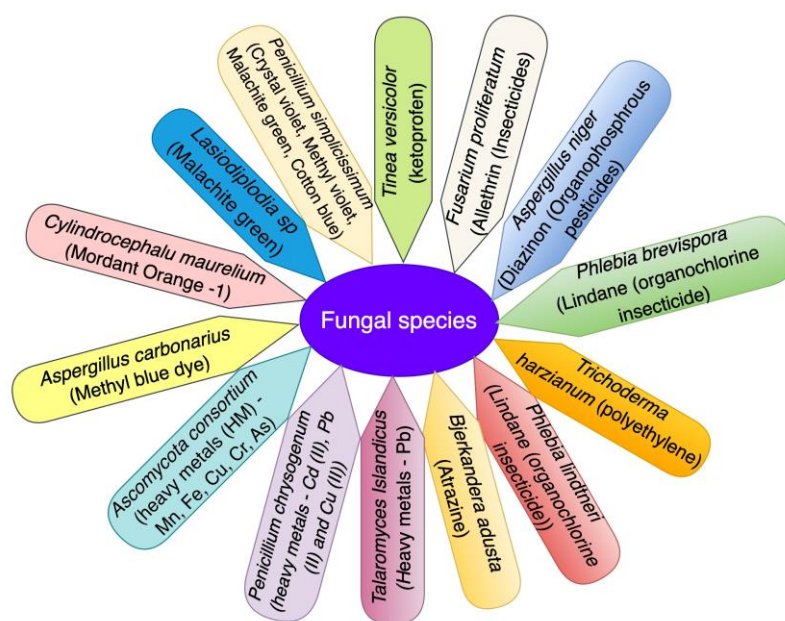


Figure 1. Bioremediation of various pollutants and their specific fungal species applied [45].

2.1. Wood Rot Fungi

Wood rot fungi are classified as *Basidiomycota* (*Basidiomycetes*) [46]. These species of fungus play an essential role in the bioremediation of organic contaminants in addition to their role in wood rooting. No preconditioning is necessary before the transformation of pollutants [47]. Therefore, they have extensive flexibility and the capacity to degrade tissues. The wood-degrading fungi species are distinguished as strong competitors like *T. versicolor*, *Phanerochaete* sp., *Pleurotus* ssp., etc., and weak competitors like *Ganoderma applanatum* and *Dichomitus squalens* based on their capacity to colonise new environments. According to their mode of action in wood tissues, they can be classified as brown rot fungi and white rot fungi [48]. The total absorption of spilled explosive, organochlorine insecticides, polychlorinated biphenyls (PCBs), petroleum hydrocarbons, and PAHs are achieved by white-rot fungi (*Polyporus* sp. and *Phanaerochaete*) [49,50]. In a study by Adenipekun and Isikhuemhen (2008), it was found that *Lentinus squarrosulus* can increase the carbon content, bioavailable phosphorus, and organic matter of soils contaminated with motor oil [51]. The Fenton reaction discovered that *Gloeophyllum trabeum*, *Fomitopsis pinicola*, and *Daedalea kinsii* effectively degraded Dichloro-Diphenyl-Trichloroethane (DDT) [52].

Numerous fungi have been shown to degrade PAHs, including, *Agrocybe dura*, *Agrocybe praecox*, *Gymnopilus luteofolius*, *Irpex lacteus*, *Mycenagaleri culata*, *Trametes ochracea*, *Stropharia aeruginosa*, *Stropharia rugosoannulata*, *Phanerochaete velutina*, and *Physisporinus rivulosus* [53]. According to Zafra et al. (2016) [54], the fungal consortium *Aspergillus nomius*, *Rhizomucor variabilis*, *Aspergillus flavus*, and *Trichoderma asperellum* played a vital role in the breakdown of pyrene and benzo [α] pyrene phenanthrene. Thus, bioremediation of contaminated resources is highly dependent on fungal colonies. Therefore, further research could be done to comprehend the precise mechanisms and processes employed to enhance the performer's inherent capacity.

2.1.1. Brown Rot Fungi

This group of cellulose-degrading fungi metabolises the hemicellulose and cellulose found in wood. Most brown-rot fungi are located in the *Gloeophyllales*, *Polyporales*, *Hymenochaetales*, and *Agaricales* [55]. Due to demethylation, oxidation, and depolymerisation, the lignin was partially modified via a Fenton-type catalytic system without enzymes. Partially altered lignin gives rotten wood its distinctive dark brown hue [56]. A brown rot fungi-induced oxidative process leading to the formation of hydrogen peroxide (H_2O_2) also aids in the production of hydroxyl (OH) free radicals, which smooth the mineralisation and

biodegradation of synthetic chemotherapeutics agents [57]. The formation of oxalic acid and its tolerance to antimicrobial drugs increases its capacity to transform metals. Brown rot fungi can significantly use this bioremediation potential [58].

2.1.2. White Rot Fungi

A few species of white-rot fungi have the distinctive ability to break down lignin along with hemicellulose and cellulose, causing deterioration and bleaching of wood [48]. These fungi were first identified in the group while studying mycoremediation, and they generate enzymes like laccase, H_2O_2 -generating enzymes, manganese peroxidase, and lignin peroxidase [59]. Extracellular oxidative ligninolytic enzymes from *Phanerochaete chrysosporium* have been extensively researched for the biodegradation of complex substances. *P. chrysosporium* breaks down insoluble or toxic chemicals into H_2O and CO_2 more effectively than other fungi or microorganisms. Recalcitrant chemicals are degraded or biotransformed using various oxidative and reductive techniques to make their presence in the environment less toxic. The variety of aromatic complexes and xenobiotic chemicals in contaminated soil is easily eliminated owing to the properties of the non-specific and flexible ligninolytic enzymes [60]. Besides *P. chrysosporium*, other white-rot fungi known for degrading these substances include *Irpex lacteus*, *Lentinula edodes*, *Bjerkandera adusta*, *Trametes versicolor*, and *Pleurotus ostreatus* (=*P. ostreatus*). Several studies have shown that at least 30% of mycoremediation processes involve white rot fungi [61].

2.2. Leaf Decomposing Fungi

One of the critical elements in forest ecology is the fungus that breaks down leaves. These fungi actively perform the mineralisation, humification, and degradation of wood and soil organic materials [62]. A rapid successional change occurs as this fungal population degrades the leaf litter. *Ascomycota* fungi predominate in the early phases of litter decomposition, but as decay progresses, their population steadily declines as the number of *Basidiomycota* fungi increases [63]. These fungi produce a variety of lignocellulolytic compounds from plant litter, which are degraded. *Basidiomycetous* litter fungi are required for the biodegradation of lignocelluloses using extracellular enzymes like peroxidase, laccase, and oxidase, producing hydrogen peroxide (H_2O_2) [64]. These enzymes also biotransform persistent organic pollutants in the soil, including pesticides and herbicides. Therefore, using these fungi for bioremediation will expand the spectrum of contaminants in the soil [6].

2.3. Endophytic Fungi

Endophytic microbial groups, including fungi and bacteria, can colonise plants without harm to the host or themselves [65]. They are found in particular plant tissues, including vascular bundles, apoplastic spaces, cortical roots, dead cortical cells, and young buds. They produce various compounds for nitrogen fixation and methane assimilation [66]. They possess the saprophytic properties necessary to survive in dead litter [67]. In addition, they generated numerous enzyme groups, including lipase and cellulase, peroxidase, and protease, for the deterioration of environmental chemicals, such as petrochemicals, polyaromatic hydrocarbons, herbicides, pesticides, polychlorobiphenyls, polyester polyurethane, insecticides, etc., and the biotransformation of heavy metals in their reduced states [68]. In this way, they enhance the ability and flexibility of tolerance to toxins and other poisons, including heavy metals. Therefore, these groups are valuable tools for the bioremediation [69].

2.4. Mycorrhiza

Interactions between fungi and plant roots occur in various forms, including ectomycorrhiza, orchid mycorrhiza, ericoid mycorrhiza, monotropoid mycorrhiza, arbutoid mycorrhiza, arbuscular mycorrhiza, ectendomycorrhiza [70]. They protect against environmental stressors such as metal toxicity stress, and water is also associated with plants'

nutrient supply. The plant system's toxicity of heavy metals was enhanced by lowering the metal translocation [71]. This indicates that translocation mechanism supports plants' capacity to adapt to and survive in environments with high levels of heavy metal contamination. The host plant also benefited the fungus by keeping a contaminated area around it and metabolising numerous petroleum products, PAHs, and chlorinated aromatic pesticides, like 2,4-dichloro phenoxy acetic acid (2,4-D), and atrazine through enzymatic degradation [72]. For potentially toxic components in the soil to be broken down, mycorrhizal is essential [73].

2.5. Soil Fungi

Zygomycota, Chytridiomycota, and Ascomycota are the soil fungi that comprise this complex group. These groups play essential roles in nitrogen and carbon cycling and soil organic matter degradation, the most critical elements of soil ecology [55]. Typically, they are saprophytes, non-ligninolytic organisms that can degrade cellulose [74]. Genera in this category include *Trichoderma*, *Stachybotrys*, *Rhizopus*, *Phlebia*, *Penicillium*, *Paecilomyces*, *Mortierella*, *Geomyces*, *Fusarium*, *Engyodontium*, *Cladosporium*, *Cunninghamella*, *Beauveria*, *Aspergillus niger*, *Alternaria*, *Microsporum*, *Allescheriella*, and *Acremonium* [68]. Non-ligninolytic fungi produce monooxygenase, an extracellular enzyme that degrades PAHs by hydroxylation [75]. The fungi serve as mediators for soil bioremediation and also tolerate a variety of contaminants, such as endosulfan, chlorinated biphenyls (CBA), PAHs, and PCBs. The fungus degraded the resistant polymers in further degradation phases. Consequently, these fungi are categorised as xenobiotic-degrading fungi, and their multispecies consortia demonstrate their exceptional effectiveness in soil bioremediation under laboratory conditions [76]. In a study conducted by Passarini et al. [77], researchers found that *Aspergillus sclerotiorum* CBMAI 849 and *Mucor racemosus* CBMAI 847 could metabolize between 50% and 90% of pyrene and benzo[a]pyrene. Table 2 lists recent species of fungi and their biodegradation performance under different environmental conditions.

Table 2. Biodegradation performance of recent fungal species under varying experimental conditions.

Fungal Species	Remediation Methods	Pollutants	Experimental Conditions	Degradation Environment	Treatment (mg/L)	Removal/Uptake	Ref.
<i>Fusarium proliferatum</i> CF2	Degradation	Allethrin (Insecticides)	Incubation conditions: 5 days, media—mineral slat at 26 °C, shaker speed—110 rpm and pH—6.	Aerobic	50 mg/L	95%	[78]
<i>Tinea versicolor</i>	Biosorption	Ketoprofen	100 µL of fungi incubation were injected for 21 days at 25 °C under 150 rpm shaking speed.	Aerobic	5 mg/L	80%	[79]
<i>Staphylococcus succinus</i> HLJ-10, <i>Aspergillus niger</i> MK640786	Degradation	Diazinon (Organophosphorous pesticides)	Culture conditions: T—30 °C, pH—5, shaker speed time: 7 days.	Aerobic	25 mg/L	91.8%	[80]
<i>Pleurotus dryinus</i> , <i>Trametes hirsuta</i>	Biosorption, and biotransformation	Phenol	150 rpm media cylindrical woodchips and 4 g/L glucose at 27 °C.	Anerobic	Biorefinery wastewater	94% and 100%	[81]
<i>Aspergillusterreus</i>	Adsorption and degradation	Azo dye	Incubation conditions: T—30 °C, and contact time 168 h	Aerobic	100 mg/L	98.4%	[82]
<i>Cylindrocephalum aurelium</i>	Biotransformation	Mordant Orange-1	Incubation conditions: pH—3, agitation speed (100 rpm), in the dark for 30 days	Anerobic	20,000 mg/L	86%	[83]
<i>Lasiodiplodia</i> sp.	Degradation	Malachite green	Incubation conditions: pH—7, T—30 °C.	Aerobic	50 mg/L	96.9%	[84]
<i>Talaromyces amestolkiae</i> , <i>Penicillium ludwigii</i> , <i>Penicillium citrinum</i> , <i>Gongronella butleri</i>	Biosorption	Uranium	Incubation conditions: media—potato dextrose broth, shaker speed—horizontally at 150 rpm at 25 °C for 7 days.	Anerobic	100 mg/L	60% (11 species out 57)	[85]
<i>Talaromyces islandicus</i>	Uptake	Pb	Incubated for 5 days at 30 °C.	Aerobic	100 mg/L	89.14%	[86]

rpm—revolutions per minute.

3. Bioremediation (In Situ and Ex Situ)

This is a simple, eco-friendly, long-lasting, and cost-effective way of restoring and cleansing contaminated soil. It entails the natural decomposition of petroleum hydrocarbon pollutants by microorganisms like fungi, bacteria, and yeast that degrade hydrocarbons. Microorganisms with adequate nutrients and optimized limiting factors convert contaminants in soil into nontoxic or simpler compounds, such as water and carbon (IV) oxide, through oxidation under aerobic conditions [87–90]. In addition, In situ and Ex-situ bioremediation are well-established technologies in contaminated environments (Figure 2) [91]. Before adopting bioremediation, it is critical to evaluate all the limiting elements that can influence the remediation process's effectiveness. It is easier for microorganisms to break down aliphatic hydrocarbons than branched or long-chain hydrocarbons, which are more difficult to biodegrade [92]. Microorganisms that degrade hydrocarbons use carbon compounds for reproduction, growth, and energy.

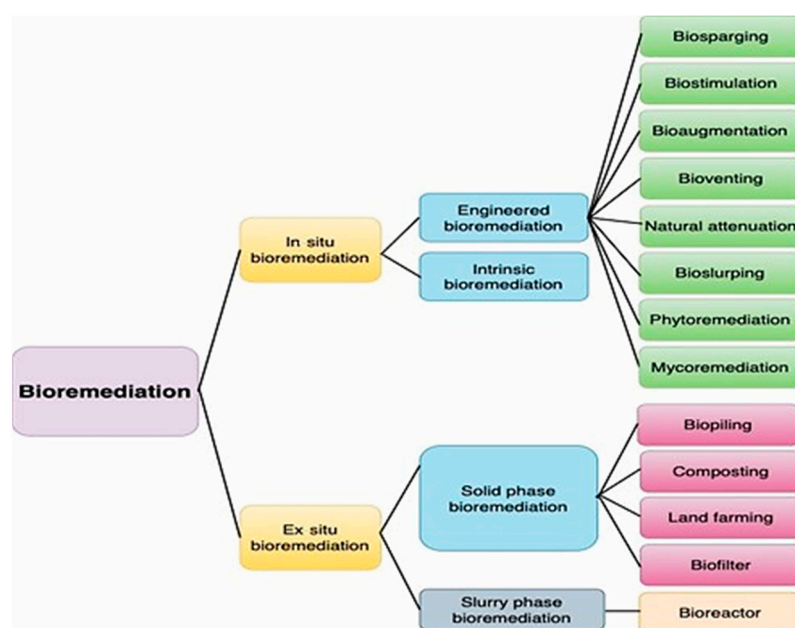


Figure 2. Different techniques of bioremediation.

Bioremediation utilizing chosen microorganisms to break down petroleum hydrocarbons is gaining interest among researchers. *Pseudomonas* bacteria efficiently break down petroleum hydrocarbons into simpler compounds [93,94]. In addition, fungi including *Rhizopus*, *Fusarium*, and *Penicillium* have been identified and employed in the bioremediation of sediments and soil contaminated with petroleum hydrocarbons [95,96]. However, bioremediation has been used for petroleum hydrocarbons since 1940 but gained a lot of attention after the Exxon Valdez oil spill in 1980 [97]. Vidali [98] categorized microorganisms participating in bioremediation processes as follows:

Aerobic microbes are responsible for biodegradation in the presence of oxygen, with *Mycobacterium*, *Rhodococcus*, *Sphingomonas*, *Alcaligenes*, and *Pseudomonas* being the aerobic bacteria identified for their degrading abilities. It has been observed that these bacteria degrade pesticides and hydrocarbons, including alkanes and polyaromatic compounds. Many of these bacteria rely solely on the contaminant for energy and carbon.

Anaerobic bacteria degrade without oxygen. Anaerobic bacteria that dechlorinate the solvent trichloroethylene (TCE), chloroform, and PCBs from river sediments are gaining increasing attention.

Ligninolytic fungi, including the white rot fungus *Phanaerochaete chrysosporium*, can break down various toxic or persistent environmental pollutants.

3.1. Ex Situ Bioremediation

Ex situ, as the name implies, refers to the removal of contamination to a remote treatment facility. Due to the significant labour-intensive task of excavating contaminated soil and transporting it offsite, this approach is not very popular. Ex situ remediation's fundamental element is introducing the suitable soil nutrient, moisture, and oxygen conditions offsite [99]. Nonetheless, the ex situ bioremediation procedure increases the risk of contamination spread or accidental spillage during transport [100]. Two different technique classes can be used, as explained below.

3.1.1. Slurry Phase

This process involves mixing contaminated water, soil, and other additives in a sizable bioreactor to maintain contact between the native microorganisms and the contaminants. The bioreactor environment is maintained at its ideal level to allow the microorganisms to break down the pollutants, adding essential nutrients and oxygen. After the treatment, the water is separated from the solids, and, if the wastewater is still contaminated, it is disposed of and further treated. The slurry phase is a relatively quick process for treating contaminated clay compared with other biological treatment methods [101].

3.1.2. Solid Phase

Soils are treated using solid phase treatment in a ground treatment area. This area is fitted with collection systems to prevent contaminants from eluding treatment. The degradation rate is increased by controlling variables such as moisture, heat, nutrients, and oxygen. Solid-phase systems require more space and processing time than slurry-phase processes, but they are still easy to operate and maintain. The following methods can be used to achieve this treatment [102].

Soil Biopiles

This biodegradation technique is applied to clean up excavated soil that contains petroleum-contaminated materials. Biocells are another name for soil biopiles. This method entails piling up contaminated soil and triggering microbial growth either aerobically or by adding moisture, nutrients, or minerals [101]. Biopiles can range in height from 3 to 5 feet. Additionally, oxygen is used in this technology to promote bacterial growth. Aeration of biopiles is accomplished by injecting air through piping with perforations strategically placed throughout the pile [102].

Composting

During composting, contaminated soil is mixed with biomass, such as corncobs, hay, and straw enhancing oxygen and water availability to microorganisms. The contaminated soil is placed in treatment containers and aerated during composting by mixing. When composting, a process known as "window composting" is used. Tractors mix the material frequently in the vast mounds of soil that make up the "windows." The ratio used for composting, which varies depending on soil type, contaminant concentration, and characteristics, is 75% contaminated soil to 25% compost. Since compost remediation can be completed within a few weeks, it is a faster remediation method [103].

Land Farming

This technique promotes native microorganism biodegradation and makes it easier for contaminants to degrade aerobically. This is accomplished by excavating contaminated soil and repeatedly spreading it on a prepared bed until the contaminants are degraded. To promote the development of native species, some minerals, and nutrients are also provided.

3.2. In Situ Bioremediation

The in situ method is characterized by applying bioremediation at the point of contamination. These techniques are typically the most suitable options due to their low cost and

minimal disruption, as they facilitate in situ treatment without excavating or transporting contaminants [104,105]. The soil depth that can be effectively treated in situ is a limitation for in situ remediation. In many soils, bioremediation only occurs at about 30 cm depth or less due to oxygen diffusion limits, but in some cases, greater depths of 60 cm and more have been successfully treated [106].

3.2.1. Types of In Situ Bioremediation

Intrinsic Bioremediation

Intrinsic bioremediation is a technique using the inherent propensities of the local microbial population to convert environmental pollutants into nontoxic forms. Typically, this process is applied to subsurface locations such as underground petroleum tanks. Intrinsic bioremediation utilizes the existing capabilities of microbial communities in the natural environment to degrade environmental pollutants without modifying them or accelerating the process through engineered processes [99]. The local microbial population is stimulated, and its metabolic activity is increased by supplying nutrients and oxygen.

Enhanced (Engineered) In Situ Bioremediation

As its name implies, this method introduces a microorganism to the contaminated area. By improving the physicochemical conditions to promote the growth of microorganisms, engineered in situ bioremediation speeds up the degradation process.

- Biosparging

Biosparging is a biological process that eliminates aromatic compounds such as benzene, toluene, ethylbenzene, xylene, and naphthalene. This process involves the reduction of aromatic compounds and mineral oil into a more straightforward, more useful form by adding appropriate aerobic bacteria. To promote microbial activity and improve the removal of pollutants from polluted sites, air is introduced into the subsoil, similar to bioventing. When air is pumped into the saturated zone during biosparging, volatile organic compounds may migrate upward into the unsaturated zone to speed up the biodegradation [107]. Two significant factors influence the biosparging procedure: the permeability of the soil (which determines the bioavailability of pollutants for microorganisms) and the biodegradability of the pollutant.

- Bioaugmentation

Bioaugmentation is a strategy to increase the effectiveness of the existing microorganism population in reducing contamination. This technique involves adding an organic culture to the contaminated soil to create a bioreactor-like environment. Two standard options for remediating a contaminated site are adding a pre-adapted pure bacterial strain and a pre-adapted consortium. Bioaugmentation is primarily utilized for bioremediation on oil-contaminated sites. Bioaugmentation is an inexpensive technique for treating wastewater and soil contamination compared with other methods [108].

- Bio-venting

Bio-venting is an in situ remediation technology that uses microorganisms to decompose organic soil elements [109]. This technique includes the controlled stimulation of airflow to enhance bioremediation by increasing the activity of indigenous microorganisms. Bio-venting consists of adding nutrients and moisture to enhance bioremediation and achieve the non-toxic transformation of pollutants by microorganisms. This method has gained importance among in situ bioremediation techniques, especially for the remediation of sites contaminated by light petroleum products. Bioventing is used primarily for degrading adsorbed fuel residuals, but it can also be used for degrading volatile organic compounds (VOCs) via biologically active soil.

- Bioslurping

The technique of bioslurping combines bioventing with vacuum-enhanced pumping to remediate soil and groundwater by indirectly supplying oxygen and promoting con-

taminant biodegradation [110]. This method employs a “slurp” that extends into the free product layer and draws up liquids (free products and soil gas) from this layer in a way similar to how a straw draws the fluid from any vessel. An adjustable-length “slurp tube” is installed in the well of the bioslurping system. As the slurp tube is connected to a vacuum pump, free product, and some groundwater can be extracted from the light non-aqueous phase liquids layer (LNAPL). The vacuum-induced negative pressure zone promotes the LNAPL flow to the well, which also attracts LNAPL trapped in tiny pore spaces above the water table. This method purifies soils contaminated with semi-volatile and volatile organic compounds.

- **Phytoremediation**

Phytoremediation is the direct application of green plants and the accompanying microorganisms to maintain or lessen contamination in sediments, sludges, soils, surface water, or groundwater. To reduce the toxic effects of pollutants, this technique relies on using plant interactions (physical, biochemical, biological, chemical, and microbiological) in contaminated sites. It can be used as a replacement technology alongside conventional mechanical clean-up methods, which frequently demand high capital investments and consume much energy. Particularly favorable conditions for phytoremediation can be found in areas with low contaminant concentrations over significant clearing areas and at shallow depths. According to the pollutant type (elemental or organic), phytoremediation involves several mechanisms (extraction or accumulation, volatilization, stabilization, filtration, and degradation) [111]. Extraction, transformation, and sequestration are the main methods to remove elemental pollutants (toxic heavy metals and radionuclides).

3.3. *Merits and Demerits of Bioremediation*

Microorganisms have been routinely used to treat and transform waste products for at least 100 years, although bioremediation has evolved into an innovative and environmentally friendly technology. Municipal wastewater treatment relies on the metabolic activities of microorganisms to break down organic matter in wastewater that arrives at treatment plants with selected and adapted populations of microorganisms. This industry depends on using microorganisms in engineered and controlled systems [112,113].

3.3.1. *Merits of Bioremediation*

Bioremediation has several advantages over conventional remediation methods, such as landfilling. It is often possible to perform bioremediation on-site, eliminating transportation costs. The site can continue to be used for production or industrial purposes during bioremediation. Bioremediation decomposes the waste, resulting in the long-term benefits associated with non-destructive treatment processes. In addition, bioremediation can be combined with other treatment technologies to form a treatment chain that allows for treating mixed and complex wastes [114,115].

The bioremediation of waste streams can also benefit from using renewable (waste) materials [116]. A variety of residues and by-products can be recovered and upgraded by chemical or biological processes to higher value and valuable products. These residues and by-products include chips, wood sawdust, waste paper, sunflower seed hulls, rice husks, peanut shells, cotton seed and husk, coffee pulp, bananas and coconuts, tequila bagasse and sugarcane bagasse, vine prunings, sorghum stover and maize, reed stems, grasses, cotton stalks, corn cobs, and cereal straw [117–119]. The chemical characteristics of such lignocellulosic agricultural residues make them a substrate of enormous biotechnological value. Through solid state fermentation (SSF), these products can be transformed into a variety of value-added products, including mushrooms, animal feed enriched with microbial biomass, compost that can be applied as a biopesticide, biofertilizer, flavours, ethanol, organic acids, enzymes, and biologically active secondary metabolites. Additionally, they can be used to bioremediate hazardous compounds, detoxify agroindustry residues, and process pulp [120,121]. Based on basidiomycetous cultures, SSF has been proposed to improve and add value to lignocellulosic residues by upgrading proteins and

converting residues into animal feed [122], or producing enzymes [122,123]. There has been considerable research on lignocellulolytic mushroom fungi such as *Pleurotus ostreatus* and *Trametes versicolor* for the bioremediation and biodegradation of hazardous and toxic compounds, such as caffeine residues [117], as well as poisonous chemicals found in polluted soils and groundwater, including pesticides, PAHs and PCBs and chlorinated ethenes (CIUs) [124,125].

3.3.2. Demerits of Bioremediation

Bioremediation has drawbacks and limitations, just like most treatment methods. Some chemicals, such as highly chlorinated compounds and heavy metals, are not readily susceptible to biological degradation and stabilization. Table 3 summarizes the classes of chemicals and their susceptibility to biodegradation. Additionally, microbial degradation of some chemicals may result in the production of substances that are more toxic or mobile than the parent compound(s). For instance, TCE undergoes a series of microbiologically mediated reactions in anaerobic environments, sequentially removing chlorine atoms from the molecule. This process is known as reductive dehalogenation. Vinyl chloride (VC), a known carcinogen, emerges as the final byproduct of this chain of reactions.

Table 3. Classification of chemicals and their bioremediation susceptibility.

Chemical Class	Examples	Biodegradability
Polyaromatic hydrocarbons	Benzo(a)pyrene, anthracene, creosote	Aerobic
Petroleum hydrocarbons	Fuel oil	Aerobic
Ketones and esters	MEK, Acetone	Anaerobic and aerobic
Aromatic hydrocarbons	Toluene, benzene	Anaerobic and aerobic
Asbestos		Not biodegradable
Corrosives	Caustics, inorganic acids	Not biodegradable
Radioactive materials	Cadmium, plutonium, uranium	Not biodegradable
Metals		Not degradable experimental biosorption
Organic cyanides		Aerobic
PCBs	Arochlors	Some evidence; not readily degradable
Chlorinated solvents		Anaerobic (reductive dichlorination), aerobic (methanotrophs)

In some cases, applying bioremediation without a thorough understanding of the microbial processes involved and the metabolic and chemical pathways could make the situation worse than it already is. Due to the complexity of the science of bioremediation, it must be tailored to a particular site's environmental and kinetic constraints to minimize their effects [126,127]. Therefore, the initial costs for site assessment, characterization, and feasibility evaluation for bioremediation might be more expensive than those linked to more traditional technologies such as air stripping. To evaluate the efficiency of the bioremediation technique in its clean-up performance, extensive site monitoring is required during project implementation, just as with remediation technologies [128]. Monitoring requirements may include microbiological and chemical monitoring associated with chemical/physical remediation techniques. The implementation of bioremediation is also impacted by regulatory restrictions [129]. The advantages and disadvantages of bioremediation technologies are outlined in Table 4.

Table 4. Merits and demerits of bioremediation technologies.

Factors	Merits	Demerits
Natural Process	Applies a biological strategy that uses microorganisms to remediate polluted areas	The biological mechanism is very delicate and necessitates the presence of microorganisms with metabolic activity, favourable growth conditions, and appropriate nutrients.

Table 4. Cont.

Factors	Merits	Demerits
Labour/Effort	It is easy and requires less labour.	It is difficult to transfer the mechanism from pilot-scale to large-scale application.
Cost-Effectiveness	Compared with more conventional methods for cleaning up toxic waste, it is a more affordable strategy.	Bioremediation techniques, such as reactor designs, can, however, be more costly than conventional methods.
Duration		A bioremediation treatment requires more time than other treatment options. Using little or no nutrient amendments can slow down the bioremediation process.
Nutrient amendments	There is constant availability of nutrients (organic and inorganic wastes) that are readily applied to encourage the rapid growth of microbes.	The bioremediation process can be hampered by amendments and nutrients that are toxic to the microorganisms.
Ease of application	As bioremediation occurs on-site, it eliminates the need for waste to be moved off-site, protecting human health and the environment at the same time.	
Environmentally friendly	It is non-intrusive, meaning site users can continue to use the site without interruption. The method is environmentally friendly and sustainable.	Some biodegradation products have the potential to be more harmful than the original compounds in some cases, and persist in the environment.
Contaminant type	A wide range of biodegradable contaminants can be treated with this technique.	Not all substances undergo a complete and rapid decomposition, particularly inorganic contaminants.
Legislation and Guidelines		Regulators continue to disagree about the proper performance standards for bioremediation.

4. Mycoremediation

Using fungi, this method reduces or eliminates environmental pollutants by breaking them into less toxic or non-toxic forms [130,131]. Furthermore, mycoremediation is restricted to the surface or the aerobic soil zone where mycelia of fungi are able to grow.

Fungi can degrade environmental pollutants because they produce and secrete enzymes that break down lignin and cellulose [132]. For bioremediation, it is essential to use ligninolytic fungi such as *Polyporus* sp. and *Phanaerochaete chrysosporium* because these fungi can degrade a wide range of toxic pollutants [133]. Various types of fungi degrade a wide range of materials in different environments. The cultivation of *Penicillium* sp. has caused the degradation of polyethylene [134]. Several species of filamentous fungi are hydrocarbonoclastic. It has been found that some white rot fungi use their mycelia to degrade petroleum hydrocarbons because they produce oxidative enzymes, extracellular enzymes, chelators, and organic acids. According to Ulfig et al. [135], the keratinolytic fungi *Trichophyton ajelloi* successfully removed hexadecane and pristane from crude oil. A similar study was conducted by Njoku et al. [136] utilizing *Pleurotus pulmonarius* to remediate soil contaminated with a 1:1:1:1 mixture of petrol, diesel and spent engine oil. According to the findings, after 62 days of incubation, the soil treated with 10% mycelium removed 68.34% of total petroleum hydrocarbons, while the soil treated with 2.5% removed 22.12%. Based on these findings, the fungus *Pleurotus pulmonarius* may be able to remediate soil contaminated with a moderate quantity of petroleum hydrocarbon mixture.

Merits and Demerits of Mycoremediation

Fungi can immobilize PTEs by chelating them to polymers, biosorbing them, or forming insoluble metal oxalates. The nonspecificity of fungal enzymes such as Mn peroxidase, lignin peroxidase and laccase, allows fungi to degrade various soil pollutants to obtain energy or food. Special enzymes released by fungi break down pollutants such as plastics,

pesticides, PTEs, PCBs, petroleum hydrocarbons, dyes, and PAHs. These pollutants are then transported into the mushroom fruiting bodies of the fungi [137]. The extracellular enzymes of the fungi reduce the risk to human health by preventing toxins from entering the food chain. The mushroom's fruiting body serves as a final repository for the contaminants [138,139]. The degradation of pollutants and reduction in toxic effects by mushrooms have been reported by Malachova et al. [140], Choi et al. [141], and Kulshreshtha et al. [142]. Such mushrooms can be consumed and used in future environmental remediation studies.

Large-scale mushroom availability, soil contaminants that can be absorbed by mycelium, and optimal environmental factors for mycelial growth are all prerequisites for the mycoremediation technology to be successful. These characteristics cover every aspect of mushroom cultivation's physiology, ecology, and biology. Compared with every other strategy currently used for PTE eradication, mycoremediation has many advantages. Mycoremediation is an effective and well-designed method for addressing PTE pollution because it has significant mycelial growth. It can be carried out either in situ or ex situ. It is cheap, requires little space, is environmentally friendly, and can be used in fields. Mushrooms may also be grown alongside a region's main crops. There have been a few reported limitations to this process, however. The time spent creating and cleaning up the contaminated environment is the primary constraint. Growing mushrooms on-farm waste, sludge, and industrial wastes could result in toxic byproducts in the food supply and endanger people's health. It is challenging to bioremediate multiple metal-contaminated sites due to the conflicting effects of microorganisms on trace element mobilization or immobilization [143,144]. Therefore, when growing mushrooms in polluted areas, the substrate's characteristics should be considered.

5. Comparative Analysis and Application of Bioremediation Technologies

The degradation of PAHs from soil can be accomplished in three phases as follows:

- (i) Inspection of the PAH-contaminated site and its associated risk assessment involves examining the extent of PAH contamination based on their permissible levels.
- (ii) Selection of cost-effective, feasible, and environmentally friendly soil PAH degradation techniques. Based on recent research, Table 5 lists some of bioremediation techniques' influencing parameters, merits, and demerits. To date, laboratory-scale treatment methods have been implemented successfully [145–147]. Several important factors must be considered when applying bioremediation techniques at the field scale, including (a) the physical and chemical properties of the contaminated soils, including their composition, temperature, water-to-soil ratio, environmental conditions, and oxygen availability [145]; (b) the activity, diversity, microbial community, resistance, and interaction; (c) the mass trajectories, toxicity, PAH concentration and interaction [146]. To optimize these parameters for field-scale applications, they must be adapted appropriately.
- (iii) A PAH-contaminated site requires a pretreatment and posttreatment assessment. This phase examines the biochemical conversion of PAH compounds after treatment, e.g., their removal or conversion to non-toxic compounds [147].

Table 5. A comparison of remediation techniques based on their influencing parameters, benefits, and drawbacks.

Techniques	Influencing Parameters	Merits	Demerits	Applicability	Duration	Ref.
Rhizoremediation	Soil type, texture, particle size, nutrients and organic matter content.	High production of biomass. Root exudation in the rhizosphere provides better nutrient uptake for rhizosphere microbiome. Efficient tolerance of plants towards PAHs.	Inability to determine an accurate degradation time for organic pollutants. Lack of field studies.	Small scale (long term)	Longer degradation time	[148]

Table 5. Cont.

Techniques	Influencing Parameters	Merits	Demerits	Applicability	Duration	Ref.
Phytoremediation	Root zone, characteristics of plant species, characteristics of PAHs, characteristics of medium, environmental conditions.	Increased soil fertility through the release of organic matter. Suitable for large-scale applications. Environmental and eco-friendly.	Time consuming, particularly in moderately and highly contaminated sites due to slow growth rate and low production of biomass.	Large scale (long term)	Longer degradation time.	[149]
Genetically modified microorganism (GEMs)	Chemical structure, microbial population composition, environmental conditions.	Low-technology equipment is required. Depending on the soil condition, in situ and ex situ methods can be employed. Equipment requirements are minimal in comparison with other remediation technologies. It is possible to completely break down organic contaminants into non-toxic chemicals.	Less information available on risk assessment of GEMs. Treatment takes a longer time. A volatile organic compound (VOC) cannot be controlled effectively using the ex situ method. Physico-chemical characteristics and toxicity of soil are extremely sensitive to these parameters. Presence of incomplete breakdown of organic contaminants if the process is not well controlled, managed and monitored.	Large scale (long term)	Longer degradation time.	[150]
Nano-remediation	Remediation time, initial concentration of PAHs, dosage of nanomaterial.	Good surface-coating lability. Due to the large surface area, there is a high level of reactivity and a large number of active sites. Enables remediation in deeper soil.	Exposure of nanomaterials to both humans and the environment.	Large scale (long term)	Shorter degradation time	[151]
Vermiremediation	Earthworm's life cycle (i.e., feeding, burrowing, metabolism, secretion).	Cost-effective remediation. Advantage of increasing earthworm biomass that can be harvested and used as livestock feed.	Earthworms may not be suitable as biomonitoring agents due to risk assessment. It is not suitable for cleaning up highly polluted soil.	Small scale (long term)	Very less	[152]
Electrokinetic remediation	Mixed nature of contaminant, electrolyte properties, voltage gradient, and soil heterogeneity.	Effective with low permeability soil. Low environmental impacts.	Not effective for all types of PAHs. Low solubility. Poor desorption ability.	Small scale (long term)	Longer degradation time	[153]
Mycoremediation	Temperature, pH, heavy metals, and redox potential.	It is economical, eco-friendly, and an effective strategy to combat the ever-increasing problem of soil and water pollution.	As a result, the process is often slow, and the proportion of contaminants removed rarely approaches 100%.	Small scale (long term)	Shorter degradation time	[154]

The field applications of the aforementioned bioremediation strategies have only been the focus of a small number published studies. According to Guo et al. [155], differences in bioavailability persist after bioremediation in agricultural and industrial soils with PAH contamination. *Mycobacterium* sp. and *Mucor* sp. were injected into agricultural soils and manufactured gas plant (MGP) soils to study PAH biodegradation. The bioavailability of PAH before and after biodegradation was estimated using Tenax-TA extraction and solid-phase microextraction (SPME), respectively, to determine the bioavailability and chemical activity of the compounds. During biodegradation in MGP soil, only PAHs with three and four rings were degraded. MGP soils and agricultural soils degrade PAHs differently. The use of earthworms to extract Tenax-TA from agricultural soils was found to be more

sensitive and effective than SPME. According to Wang et al. [156], metal accumulators and PAH remediators were co-planted to phytoextract and rhizoremediate soil contaminated with heavy metals. This is one of the most effective methods for removing heavy metal and PAH contamination from soils. By co-planting *Sedum alfredii* (*S. alfredii*) with ryegrass (*Lolium perenne*) or castor oil plant (*Ricinus communis*), metal and PAH contaminants from co-contaminated soils were reduced. Compared with a monoculture planting, co-planting *S. alfredii* with castor oil plant decreased shoot biomass. When planted with ryegrass or castor oil plant, the cadmium content of *S. alfredii* shoots decreased significantly compared with the monoculture, but there was no reduction in Pb or Zn concentrations in co-planted *S. alfredii* shoots. Co-planting ryegrass or castor oil plant with *S. alfredii* significantly increased pyrene and anthracene dissipation compared with bare soil or a *S. alfredii* monoculture.

Due to strict laws preventing the release of GEMs into the environment, only a few GEMs have been used in field trials [157]. The transfer of genetically engineered microorganisms from the laboratory to the field is complicated by a lack of knowledge about the population dynamics of genetically engineered microbes in the field and by inadequate physiological control of catabolic gene expression in genetically engineered organisms under nutritional and other stress conditions [158,159]. Before they can be used in the field, bioengineering and releasing these modified microorganisms into the environment must overcome several challenges, such as differences in hazard assessment protocols and public health issues. To increase biodegradability through genetic modification, other bacterial strains are preferred over indigenous bacteria that can proliferate and withstand regionally stressful conditions.

Rodriguez-Campos et al. [160] removed PAH, total petroleum hydrocarbons (TPH), and alkanes using a combination of bioaugmentation, phytoremediation, and vermiremediation. The bacterial consortium augmented the earthworm's ability to eliminate PAH (at more than 77% removal efficiency). Surfactants are responsible for desorbing and mobilizing the organic contaminants in soil [161,162], so adding surfactants to the vermiremediation process can be of utmost importance for the removal of PAH contaminants. Due to their low ecotoxicity, natural surfactants can offer a more cost-effective and trustworthy bioremediation process than synthetic ones [163].

Recently, there has been an increase in the use of composting for the degradation of PAH compounds. Ex situ aerobic stabilization of organic compounds requires moisture, oxygen, and porosity. Sayara and Sánchez [164] report that the compost/co-substrate matures in contaminated soil due to the microbial action within the mixture, resulting in PAH degradation.

6. Mechanisms of Mycoremediation

The synthesis and secretion of biological compounds, mode of action, metabolites, and growth needs are essential characteristics of fungal species that are known pathways and mechanisms for mycoremediation (Table 6). Figure 3 depicts multiple mechanistic paths for mycoremediation. The mechanisms of mycoremediation can be categorised as follows:

- Avoidance reduces metal accumulation via absorption, precipitation, and biosorption, which lowers metal toxicity.
- Extrusion is the process of transporting contaminants out of the fungal biomass.
- Sequestration mechanisms involve synthesising intracellular chelating compounds and subsequent chelation in the fungal cells to dilute the contaminants.
- Biotransformation includes the reduction, oxidation, demethylation, methylation, and evaporation processes which convert toxic compounds and heavy metals (HMs) into less harmful forms.

Table 6. Different mycoremediation mechanisms: merits and demerits.

	Pros	Cons	Remarks	Ref.
Biotransformation	A faster fungal growth rate shortens the time required for transformation. The biocatalyst operates between 20 and 40 °C and has a pH range of 5.0 to 8.0 at ambient conditions. It requires minimal operational control and time-saving technology.	Biocatalyst operating parameters must be precise. Enzymes are an expensive system. A high concentration of product or substrate can inhibit certain biocatalytic reactions, thus halting biotransformation.	Biological catalysts are used in bio-transformations, which are organic reactions.	[165,166]
Biosorption	Cost-efficient production of biomass. Simple and customisable method to remediate a wide range of contaminants. Simultaneous removal of many HMs.	Expensive regeneration. Numerous kinds of adsorbents are required. Reactor saturation and clogging.	Multiple metals can be removed at once, which is environmentally friendly but expensive.	[167,168]
Precipitation	Removal of high pollution loads is mainly achievable. Excellent at removing metal sulphide. The simplest and least expensive wastewater treatment system.	Unfeasible for the removal of small amounts of pollutants. It is difficult to maintain an environment conducive to growth and development.	Pollutants are eliminated, and the end product is separated.	[168,169]
Natural attenuation	Economically feasible. Following other bioremediation treatments, it can be utilised as a “polish” treatment. Reducing environmental contaminants without human intervention.	Long-term and extensive performance monitoring required. Not a time-efficient method. Natural attenuation’s removal of one contaminant can also remove other advantageous components.	Utilised for the co-precipitation, dispersion, immobilisation, and reversible and irreversible sorption of inorganic components.	[170–172]
Surface sequestration	Fungi secrete an extracellular enzyme that converts complex material into a simpler form which is then absorbed by its cell wall, so facilitating the remediation of a broad spectrum of persistent pollutants (PAHs, insecticides, and pesticides).	Few candidates from the fungal kingdom demonstrate their effectiveness in bioremediation under field conditions. Extracellular enzyme activity is impeded by high glucose concentration and stimulated by reducing glucose concentration.	Comprises chelation, complexation, coordination, and physical adsorption.	[173,174]

Various microorganisms, including fungi, algae, and bacteria, can break down PAHs. However, the most significant part of mineralisation is by far played by bacteria. On the other hand, fungi primarily biotransform PAHs, which involves detoxification to less toxic or nontoxic metabolites that can be utilized by other organisms [175]. Most bacteria that can break down high molecular weight PAHs are Actinomycetes from the genera *Mycobacterium*, *Rhodococcus*, and *Gordonia* [176]. The PAH is converted to a dihydrodiol by a multi-enzyme system as the first step in the aerobic catabolism of an aromatic molecule by bacteria. This hydroxylated intermediate can undergo further processing to become a central intermediate catechol or protocatechuate cleaved through ortho or meta cleavage pathways (Figure 4). In polynuclear aromatic compounds like PAHs, the rings are sequentially broken down through dihydroxylation and cleavage, producing more intermediates before being converted into tricarboxylic acid cycle intermediates [177,178].

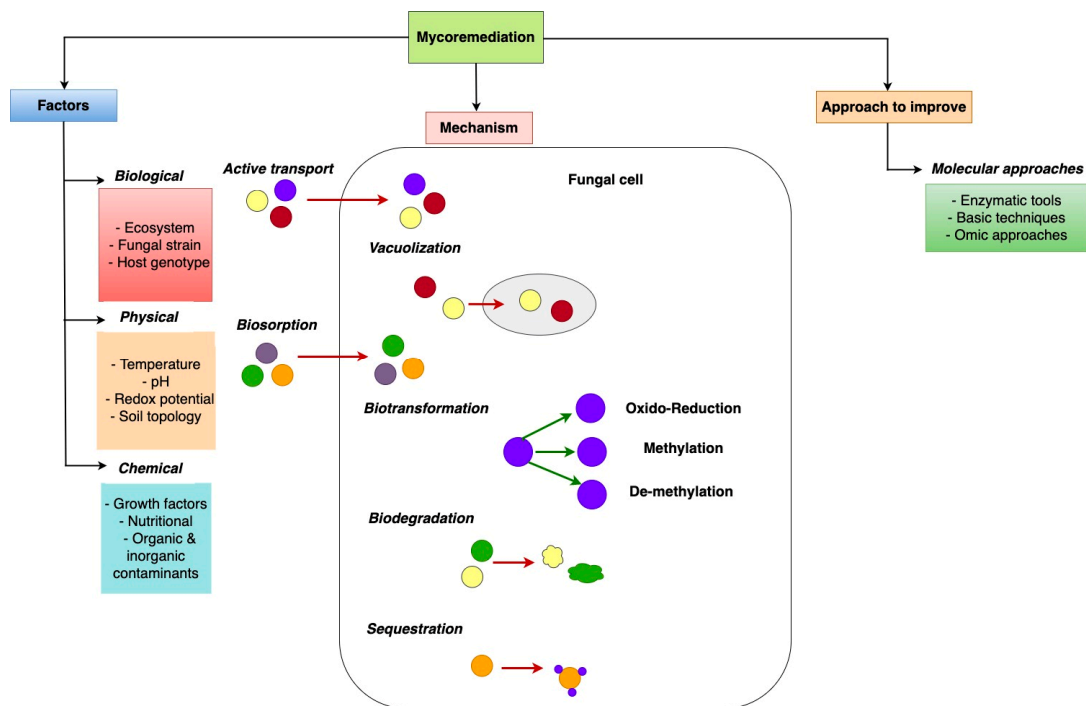


Figure 3. Diagram showing distinct cellular mycoremediation mechanisms. Adapted and modified from Kumar et al. [45]. Numerous cellular, molecular, and metabolic factors influence the mycoremediation process. Active transport or passive diffusion are two methods by which the fungus cell can absorb heavy metals (bioabsorption). Heavy metals are biotransformed, sequestered, etc., within the cell to minimize toxicity. The mycoenzymes involved in the mycoremediation process include hydrolase, oxido-reductase, and antioxidant enzymes. An extracellular enzyme, or exoenzyme, is a catalyst that acts on the exterior part of the cell. Additionally, they corrupt complex molecules like cellulose and hemicellulose and facilitate biosorption after catalyzing specific reactions (see Table 2 for a detailed discussion).

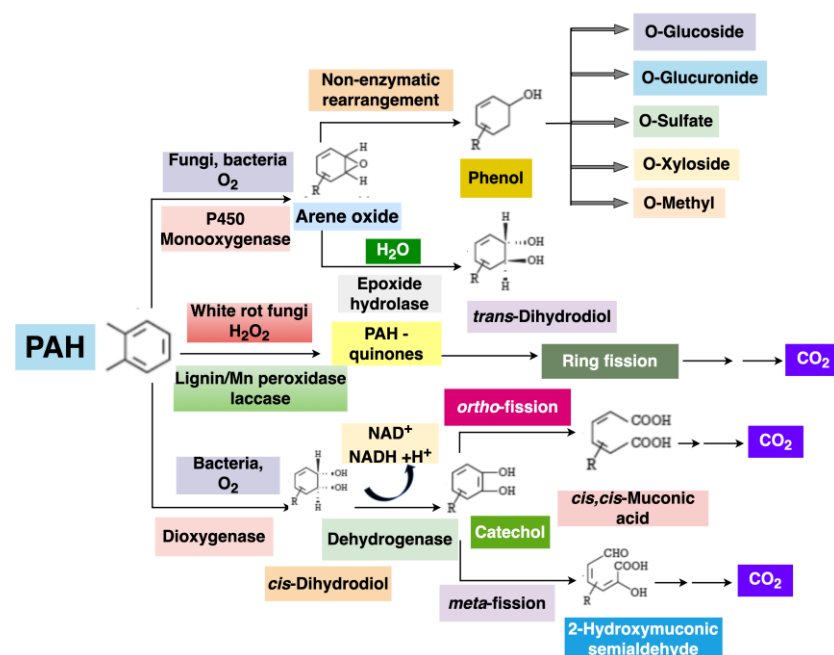


Figure 4. Degradation pathway of selected polyaromatic hydrocarbons (PAH) [179]. Adapted and modified from Dutta and Hyder (2019).

6.1. Immobilisation Process

Immobilisation reduces the mobility of pollutants by altering their physical and chemical properties. This is the optimal strategy for reducing the availability of harmful contaminants in biological systems [180]. Physically controlling interaction with pollutants or chemically modifying pollutants are viable methods of implementing this technique [181,182]. The cell of a fungus is very active, intricate, and composed of several functional groups, including thiol, amino, carbonyl, carboxyl, and hydroxyl groups. As Lewis bases, these functional groups bind metal cations and other hazardous compounds at the surface of the cell wall. They are finally trapped in filamentous fungal biomass [183]. Therefore, the structure and chemistry of the cell wall of fungi are essential for immobilising harmful contaminants. A minor alteration in the form of the fungal cell wall may considerably impact immobilisation efficiency. Solidification and stabilisation are the fungi's most effective mechanisms for immobilising pollutants [184]. Injecting appropriate substances into polluted places can precipitate non-degradable hazardous compounds, including metals, resulting in solid compounds like metal hydroxide.

The main factors determining the immobilisation of contaminants include the natural properties of polluted areas, such as water availability, pH, temperature, and soil type, [185]. In addition, immobilisation is considered one of the essential biogeochemical metallic procedures in sites contaminated with heavy metals [186].

6.2. Mobilisation

Multiple mobilisation methods, including leaching, siderophore chelation, alkylation, methylation, and redox transformation, can be used by microbes to activate contaminants. The proton efflux process microbial communities use for leaching best suits the acidic soil environment. By reducing and chelation, *Trichoderma harzianum* can oxidise Fe and Mn and solubilise Zn [187]. When iron is limited, microbes produce low molecular-weight siderophores [188]. The main task of these substances is to separate plutonium, magnesium, manganese, and iron(III) (Fe^{3+}) from other metals (Cr, Mg, and Mn) [189]. Alkylation moves an alkyl group, which can take the form of a carbene, carbanion, free radical, and an alkyl carbocation, from one molecule to another. To generate different metalloids, methylation requires incorporating methyl groups (CH_3) introduced into metal by enzymes. This mechanism is triggered by the synthesis and excretion of fungi that produce oxalic acid, an effective metal ion chelator, while citric acid in combination with metal ions forms insoluble oxalate [190].

6.3. Biosorption

Biosorption is a biotechnological technique to remediate heavy metals (HMs) using living and non-living fungi, bacteria, and algae. It is a physicochemical process in which hazardous substances from biological sources are absorbed through ion exchange, reduction, precipitation, chelation, and adsorption [191].

Biosorption uses a solvent (liquid phase) and a biosorbent (solid phase), and the solvent prevents the sorption of dissolved material [192]. Numerous industrial, agricultural, and natural waste products have been used as biosorbents; nevertheless, biomass from fungi has attracted much interest due to its significant amount of biosorbent. The fungal cell wall is the first biological component to interact with pollutants. It plays a crucial role as a barrier and protective layer that controls the uptake of potentially hazardous metals into the cell. The biosorption efficiency of fungal species can be significantly affected by changes in the cell wall structure or the environment of the fungal biomass. *Neosartorya fischeri* and *Aspergillus fumigatus*, for example, significantly reduced their As(V) biosorption capacity after acid treatment [183,193].

Similarly, the biosorption efficiency of Fe, Zn, and Ni(II) was significantly improved by *Phanerochaete chrysosporium* and *Aspergillus japonicus* at neutral to alkaline pH (7.0–9.0) [194,195]. At pH 3.0, Cu, Ni, and Zn biosorption are negligible. This might be due to *Aspergillus niger*'s cations and hydronium ions competing for binding sites [196].

Treatment of fungal biomass with FeSO_4 and FeCl_3 improved the biosorption ability of As(V) by *Aspergillus fumigatus* [193].

Metalloids and metals can be biotransformed by altering the microenvironment via catalysis, oxidation, and reducing mobility and metal solubility [185]. Methylation and decarboxylation are also involved in biotransformation, and these reactions can volatilise metals and thus reduce their toxicity. In addition to cytoplasmic vesicles and vacuoles, cytoplasmic vesicles and vacuoles can transport metals to the mycelium and plant symbionts of fungi. The biotransformation of numerous aromatic hydrocarbons by yeast, including long-chain phenyl alkanes, chlorinated phenols, ethylated benzene, dibenzofurans, ethers and their halogenated derivatives, dioxins, and polycyclic aromatic hydrocarbons, has been extensively reported [197]. However, it should be emphasised that many of these xenobiotics can be biotransformed into various compounds with unknown properties. Exploring the resulting products' risk assessments and biotransformation mechanisms is necessary to minimise the environmental impact. It has been observed that wood rot fungi can biotransform cadmium into cadmium oxalate trihydrate, zinc sulfate into zinc oxalate, copper sulfate into copper oxalate, and lead nitrate into lead oxalate [198].

A pseudo-ion exchange process, also called biosorption, involves exchanging metal ions for counterions in biomass or resin. Generally, filamentous fungi have a greater capacity to adsorb heavy metals, and Aquatic fungi have also been shown to accumulate heavy metals. Michelot et al. [199] described the uptake of metals, and a hypothesis was made about the mechanisms of bioaccumulation in fungi. It was discovered that the marine fungi *Corollospora lacera* and *Monodictys pelagica* accumulate lead and cadmium extracellularly in their mycelia [200]. Several external factors (such as the type of metal, functional site, and ionic form in solution) and an exothermic tendency are involved in biosorption. Other variables such as type of biomass preparation, biomass concentration, temperature, initial metal ion concentration and metal properties, pH, and ion concentrations of other interfering species are critical in determining the extent of biosorption. Biosorption and recovery can be enhanced by a magnetic field-induced stirring [201].

6.4. Role of Fungal Enzymes in Mycoremediation

Fungal enzymes can be used for production of organic materials, hydrolysis of lignin-related compounds, decolourising dyes and textile inks, wastewater treatment, laccase-based biosensors, pulp bleaching, and wood pulping [202,203]. Organic pollutants such as trinitrotoluene (TNT), PAHs, PCBs, phenols, and dyes can be broken down using white-rot fungi [204,205]. Reduced solids levels and pathogen burden can be achieved using fungal enzymes, including laccases, cellulases, peroxidases, xylanases, and proteases (Table 3). These enzymes are also used to help deflocculate the sludge more efficiently. Enzymes are often used in the mycoremediation of various pollutants, hydrolases, and oxidoreductases. Due to an aerobic flooded fermentation process, lignin peroxidase (an extracellular enzyme) can be released from fungi or be present in the aqueous phase [206]. Extracellular enzymes generally accelerate the breakdown of pollutants and can be applied successfully in industrial settings to treat biodegradable and organic waste [207]. Table 7 provides an overview of the most widely used fungal enzymes, the target chemical, and their potential use in bioremediation.

Table 7. Some widely used enzymes for mycoremediation produced by fungal species and targeted compounds.

Fungal Species	Enzymes Involved	Compound Degraded	Remarks	Ref.
<i>Fusarium oxysporium</i>	Endoglucanase	Transform silver	Grows on arid, temperate, and tundra soils.	[208]
<i>Bjerkandera adusta</i>	Lignin peroxidases	Xenobiotic compounds	Typically grows on decaying wood.	[209]

Table 7. Cont.

Fungal Species	Enzymes Involved	Compound Degraded	Remarks	Ref.
<i>T. versicolor</i>	Laccase	Toluene and benzene	Commonly grows in tilled layer.	[210]
<i>Aspergillus flavus</i>	Laccase	Dyes and surfactants	Legumes and cereals are excellent for promoting healthy growth.	[211]
<i>Trametes pavonia</i> , <i>Penicillium verruculosum</i> , <i>Penicillium piculispurus</i> , <i>Botryosphaeria laricina</i> , <i>Aspergillus glaucus</i>	Ligninolytic enzymes	Herbicides and pesticides	Degradation	[212,213]

7. Factors Influencing Mycoremediation

The efficiency of mycoremediation is controlled by variables including water content, nutrients, oxygen levels, pH, and temperature [214]. It has been found that ideal temperatures for mycoremediation are between 25 and 30 °C [215]. Aguilarivera [216] observed that 70% relative humidity is optimal for *P. ostreatus* mycoremediation. Brady [217] states that the ideal soil carbon-nitrogen ratio is 10. Nutrient requirements are typically met using inorganic and organic manures [215]. Gueren [218] revealed that the mixture of a sample for mycoremediation with compost increases the remediation effectiveness of PAHs by up to 50%. The addition of compost helps to optimise the temperature during the procedure [219]. Dickson et al. [216] states that phosphorus and nitrogen can become limiting factors. In addition to the fungal biomass, it is known that the duration of the remediation process, the type of substrates, and mobilising agents influence the effectiveness of the mycoremediation [220]. Various factors include a fungus life cycle, antifungal agents, soil geochemistry, surfactants, and chelating agents [75,221]. Table 8 illustrates the impact of some key parameters.

Table 8. An analysis of the biological and physicochemical influences on fungal bioremediation and their potential importance.

Fungal Species	Pollutants	Factor	Experiment Design	Remarks	Observation	Ref.
<i>Rhodotorula mucilaginosa</i> and <i>Beauveria</i>	Zn and Pb	Temperature, and time	Microcosm studies. Culture conditions: 21 days incubation, temperatures—4° and 30 °C, media—glycerol yeast extract agar (GYEA).	Accumulation efficiency: <i>Rhodotorula mucilaginosa</i> , Zn (2.50%), Pb (16.55%); <i>Beauveria bassiana</i> , Zn (0.64%), Pb (8.44%)	The growth of <i>B. bassiana</i> was not affected at a lower temperature (4 °C) and reached 74% (control), while at similar Zn concentrations at 30 °C, 62%, 70% and 88% were reached.	[222]
<i>Penicillium freii</i> and <i>Aspergillus niger</i>	PAHs (pHenanthrene, anthracene, flouranthene, and pyrene)	pH	pH range of soil microcosm (5.0–8.0).	At pH 7.5, anthracene, flouranthene, phenanthrene, and pyrene undergo 50% biodegradation.	Increasing the pH of Arthur Brower's top soils increases their bioremediation potential.	[223]
Fungi with filaments (species of <i>Rhizopus</i> and <i>Aspergillus</i>)	Cd and Cr	Metal type, pH and fungus species	YMS culture medium incubated at pH 4.5 for 4 h at 25 °C was used for the biosorption assay.	<i>Aspergillus</i> sp.1 accumulated Cr (1.20 mg g ^{−1}), and Cd (2.72 mg g ^{−1}), while <i>Rhizopus</i> sp. accumulated significantly more Cr (4.33 mg g ^{−1}).	Level of tolerance of filamentous fungi to metals were observed in the order Cu > Cr > Cd > Co > Ni. However, there was no direct relationship between level of metal resistance and biosorption capacity in <i>Aspergillus</i> isolates.	[224]

Table 8. Cont.

Fungal Species	Pollutants	Factor	Experiment Design	Remarks	Observation	Ref.
<i>Aspergillus foetidus</i>	Azo dye (reactive black 5)	Temperature, dosage and pH	0.1 M HCl/0.1 M NaOH/1.0 M was autoclaved with the fungal biomass for 1 h. Kinetics of biosorption of Azo reactive black (100 mg/L) onto fungal biomass prior to equilibrium were investigated.	At pH 2–3, <i>Aspergillus foetidus</i> showed a decolourisation efficiency of over 99%. The increase in temperature from 30 to 50 °C significantly increased the biosorption capacity.	The biosorption process was endothermic and spontaneous. Furthermore, the biosorption capacity is strongly dependent on the temperature and increases significantly with an increase in temperature from 30 to 50 °C	[225]

7.1. Temperature

Temperature affects the bioavailability of the pollutants and the development of fungal species, both of which are involved in bioremediation. Fungi are classified as mesophilic (5 °C to 35 °C), thermophilic (above 40 °C), or psychrophilic (below 5 °C) based on the temperature of their optimal growth. The degradation of organic matter was accelerated by higher temperatures, which also improved the bioavailability of contaminants and HM absorption. For mycoremediation, fungi can tolerate high temperatures [170,226]. At 30 °C, Purchase et al. [222] found that *Rhodotorula mucilaginosa* and *Beauveria bassiana* had higher levels of Pb and Zn in their bodies [222]. At 10–20 °C, *Yarrowia lipolytica* showed the most increased activity to degrade diesel oil (up to 41%). Similarly, temperatures around 15 °C resulted in 20% higher oil degradation activity of the inoculum (20%) [227]. In the temperature range from (30–50) °C, the biosorption of the fungi increases significantly [225]. At 37 °C, *Kluyveromyces marxianus* IMB3 successfully removed the most colour from the diazo dye. In addition, binding affinity and enzyme activity in fungal physiology are two other crucial factors that are influenced by temperature. The optimal temperature not only affects the growth of fungi and forms and the availability of pollutant uptake, but also affects the enzyme that converts or degrades the pollutant.

7.2. pH

pH controls the bioavailability of harmful pollutants to achieve remediation [228]. The solubility, precipitation, and bioavailability of individual toxic chemicals or components are based on their intrinsic toxicity under certain reactive conditions [229]. The bio removal of the violet-coloured (methyl violet 10B or hexamethyl pararosaniline chloride) textile dye by *Aspergillus niger* was 62.3%, 91.4%, 64.0%, and 92.4%, respectively, after a 24-h incubation period at pH 9, 8, 3, and 2 [230]. Studies have shown that pH is the most critical element in the fungal discolourisation of several azo dyes (Table 2). *Aspergillus foetidus* demonstrated the capacity to decolourise more than 99% of the reactive black dye five at pH 2–3; this ability was insignificant between pH 3.0 and 5.5 [231]. *R. arrhizus* and *A. versicolor* showed the maximum discolourisation of remazol blue (RB) (89.4%) at a pH of 6.0 [232]. 50% biodegradation of PAHs, such as pyrene, phenanthrene, fluoranthene, and anthracene, was optimal at soil pH 7.5 [223]. Furthermore, *Penicillium* species were dominant in soils with acidic pH, while *Aspergillus* populations were abundant in alkaline environments.

7.3. Heavy Metals (HMs) Bonded with Hydrocarbon

HM contamination often occurs together with polyaromatic hydrocarbons (PAHs) through car exhaust, waste incineration, and the use of fossil fuels, and this co-exposure to PAHs is extremely complicated [233,234]. Based on their binding, the diversity of potential ligands, and the different mobility of each metal ion, different PAH-bound HMs produced other deleterious environmental and microbiota-related impacts [235,236]. Since the constituting pollutants must first be separated, the transformation of HM in connection with PAH is highly challenging [237]. Moderate doses of pyrene have been found to alter

soil microbial populations and promote adjusted population growth, further assisting in reducing HM stress.

In contrast, Gautheir et al. (2004) focus on the adverse effects of PAH-metal mixtures on the microbial community [238]. Fungal species such as *Ganoderma lucidum*, *Pleurotus ostreatus*, and *Agaricus bisporus* have been described for the efficient degradation of petroleum hydrocarbons. Advantageously, polyaromatic hydrocarbons are degraded when *Pleurotus ostreatus* is present [239]. HMs (Pb, Cu, Cr, and Cd) are bonded to polyaromatic hydrocarbons and can be remediated using fungal species prevalent in natural communities (*Pleurotus*, *Fusarium*, and *Acremonium*). Furthermore, plant-microbe associations are particularly effective for the bioremediation of HMs linked to polyaromatic hydrocarbons [240].

8. Emerging Mycoremediation Processes

8.1. Myco-Nanotechnology

Myco-nanotechnology, which combines “mycology” and “nanotechnology”, offers much potential, in part because of how diverse and widely distributed fungi are [241]. It was discovered that nanoparticles with good monodispersed and consistent dimensions could be synthesized by focusing on the preparation of nanoparticles using fungi. Fungi have been found to produce significant amounts of protein, which may contribute to the mass productivity of nanoparticles. Many biomedical scientists are becoming interested in nanomaterials, one of the most recent and promising applied science fields [241].

Plant extracts, bacteria, and fungi can be used to synthesize nanomaterials. Applying mycology to the biosynthesis of nanoparticles has several benefits over using bulk plants because it eliminates the need to purify the nanoparticles after synthesis thoroughly [240]. They can also be grown quickly in laboratories and extensively in industrial settings. Fungi also create nanoparticles with precise dimensions and excellent mono-dispersion [242,243]. Numerous studies have shown the significance of fungi in the environmentally friendly synthesis of gold nanomaterials. Evidence indicates that certain fungi, including *Fusarium*, *Aspergillus*, and *Penicillium* sp., have been applied to synthesise nanoparticles like palladium, silica, platinum, gold, silver, etc. [244].

Certain bacteria, yeasts, and fungi are crucial for removing toxic metals by reducing the metal ions. For example, bio-friendly microorganisms could reduce the toxicity of metallic nanoparticles by reducing the metal ions or by creating colloidal particles containing insoluble complexes with metal ions (such as metal sulfides) [244]. It is proposed that the removal of heavy metals from wastewater and soils is achieved through biosorption-based biosynthesis of nanoparticles. Additionally, this process could support the production of heavy metal nanoparticles with potential technological applications [245]. An innovative method to fabricate metal nanoparticles has emerged: using the highly structured physical and biosynthetic activities of microbial cells (NPs) [246,247].

Mycoremediation is a cost-effective and ecologically dependable strategy for combating the escalating terrestrial and aquatic pollution crisis. The benefits of fungi are primarily attributable to their robust growth, resistance to complex pollutants, increased surface area to volume ratio, production of multifunctional extracellular enzymes, vast hyphal network, and ability to adapt to changes in temperature, metal-binding proteins, and pH [171,248–251].

8.2. Molecular Approach to Improve Mycoremediation

Fungi have a tremendous metabolic and physiological tendency to break down toxic waste in the environment via chemical modifications or influence chemical bioavailability [6]. Filamentous fungi such as *Penicillium*, *Mucor*, *Trichoderma*, and *Aspergillus* have shown resistance to inorganic and organic contaminants [252]. Mushrooms can bioaccumulate toxic metals from contaminated soils [253,254].

Terrestrial and marine species of *Penicillium*, *Trichoderma*, *Aspergillus*, and *Mortierella* have a high potential for the bioremediation [255]. Under the influence of ectomycor-

rhizal, the wild genotype of *Pinus pinaster* accumulated 30 mg g⁻¹ more Cd in its shoots than another genotype. According to Sousa *et al.* [256], diverse interactions of different *P. Pinaster* genotypes and mycorrhizal associations exist under various Cd concentrations. *Aspergillus sp.1* accumulated Cr (1.20 mg g⁻¹) and Cd (2.27 mg g⁻¹), while *Rhizopus sp.* accumulated higher amounts of Cr (4.33 g⁻¹ mg) and Cd (2.72 mg g⁻¹) and in fungal biomass [224].

Trichoderma has been shown to promote the bioconcentration factors (BCF) of Zn, Cr, and Cd when grown on Salix, Ni, and Cr with *Phalaris arundinacea* [250,257]. As a result, given the information above, it can be concluded that mycorrhiza may be a valuable tool for mycorrhizal mediation by changing the relationship between the fungus and the host, either by modifying one or more of the partners or controlling variables and accomplishing this through revegetation may result in more promising species on the contaminated sites. According to Garon and Sage [258], *Penicillium italicum* helped break down fluorine in the presence of cyclodextrins. Mycoremediation's potential may also be enhanced by adjusting several biotic and abiotic factors, including soil biological characteristics, plant, and fungal species, shaking rate, adsorbent dose, the concentration of pollutants, interactions between plants and soil, and the physicochemical properties of the soil. For example, *Aspergillus spp.* showed a more remarkable ability to remove copper and nickel at pH 4 [259]. According to Pundir *et al.* [259], the formation of metal hydroxides leads to a decrease in the metal removal rate at higher pH values (above pH 5). It was found that at higher pH, the negative charge forming strong bonds with a metal ion on the surface of the fungus occurs more frequently. But also, the dissociation of functional groups on the surface influences it.

The molecular strategy used in mycoremediation also involves the genetic modification of DNA molecule fragments containing one or more nucleotides that can be precisely inserted, removed, or replaced in the cells of an organism's genome through the use of scientific and technological advances that enable rational genetic modification at the global (genome) or local (gene) level [260]. Effector nucleases (TALEN), CRISPR-associated nucleases (Clustered Regular Interspaced Short Palindromic Repeat), and transcriptional activators are among the most widely used tools for the gene editing [261]. CRISPR-Cas technology for gene editing has been developed to be efficient and straightforward. The use of these tools may be advantageous for mycoremediation. According to recent publications, scientists have mainly used the CRISPR-Cas system in model organisms such as *Escherichia coli* or *Pseudomonas* [262] due to recent developments in CRISPR tools and the development of gRNA to express function-specific genes essential for remediation, non-model organisms such as *Achromobacter sp.* HZ01 and *Comamonas testosteroni* can also be used for the mycoremediation [263].

Another strategy for high-throughput genetic engineering methods is next-generation sequencing. By generating accurate sequence reads and ideally identifying genetic mutations, problems with microarrays can be overcome. By counting the sequence reads corresponding to a particular RNA molecule, as opposed to measuring an unreliable fluorescent stain and trying to account for all possible experimental variation, it is possible to determine the level of gene expression reflected in the amount of a particular RNA molecule. Counting sequence reads can even provide a more accurate estimate of gene expression. In studying the differences between stem cells and differentiated cells, such as cancer cells, next-generation sequencing offers distinct advantages in obtaining regulatory marks in chromatin, identifying neuronal regulatory protein binding sites in the genome, and providing information on how a signal from the outside world can alter a gene regulatory network. Next-generation sequencing is cost-effective, fast, and time-saving but computationally expensive. It is critical for future bioinformatics data mining to develop computational techniques under the circumstances.

9. Emerging Mycoremediation Applications

Mycoremediation application in bioremediation may be regarded as an emerging technology; however, numerous scientific works have been published recently. Figure 5

is a schematic illustration of emerging mycoremediation applications. Furthermore, remediation could be applied in other small-scale spillages such as soil spillage, in vitro oil contamination, etc.

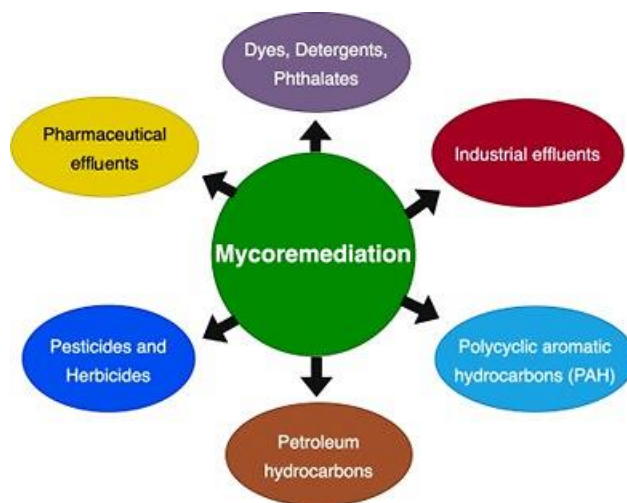


Figure 5. Schematic representation of the emerging applications of mycoremediation.

9.1. Fungal Bioremediation of Industrial Effluents

Filamentous fungi can be a reliable alternative to treat highly toxic wastewater. It is highly regarded that fungi's exceptional ability to generate organic acids, enzymes, and other metabolic intermediates gives them the capacity to survive in the most challenging environmental settings [264]. The textile and metal industries' wastewater and effluents containing cadmium, zinc, and cyno-metal compounds are both well suited for treatment by fungi enzymes. For instance, mushrooms have proven to be a viable alternative for the efficient removal and purification of industrial effluent because they generate a variety of metabolic intermediates, including citric acid, proteins of different types, and peroxidases, among other enzymes [265]. Numerous examples of fungi being used to treat wastewater have recently been shown, and yeasts can potentially adsorb wastewater from lemonade production. The relationship between *Mycorrhizal* and the soil's microorganisms are essential, and they have proven benefits in terms of increased crop quality, yields, and resistance to stress and heavy metals [266]. The bio-oxygen demand (BOD) and chemical oxygen demand (COD) can be reduced by up to 80% by several fungi, including *Saccharomyces fragilis* and *Fusarium sambucinum*.

9.2. Polycyclic Aromatic Hydrocarbon (PAHs) by Mycoremediation

The term "PAHs," also known as "polyarenes," refers to a large and diverse group of substances (more than 100 substances) that contain over two condensed benzene groups in a linear form or an angular cluster. Aromatic rings categorize PAHs; small PAHs have fewer than six rings, whereas large PAHs have more. PAHs' melting and boiling points rise along with their molecular weight while water solubility and vapour pressure decrease. The impacts of bioaccumulation and persistence on human health and the environment are severe. The accumulation of PAHs in adipose tissue, kidneys, and liver leads to the formation of even more dangerous compounds such as formaldehyde, benzene, and heterocyclic aromatic amines.

These substances present a more significant risk because of their widespread environmental presence, biodegradation resistance, and bioaccumulation capacity [267]. The risk persists due to the mutagenic and carcinogenic results of breathing in air containing PAHs. Mycoremediation is, therefore, a more effective method, and this procedure has a reasonable cost-to-benefit ratio compared to other methods. Fungi can degrade PAHs due to their intrinsic properties. For example, ligninolytic fungal enzymes are known to be nonspecific to their substrates, prompting researchers to explore this technique [268].

Various tests, such as dye decolouration, guaiacol, and gallic acid, evaluate the ligninolytic properties. Several other pollutants can also be broken down by enzymes that degrade lignin. These enzymes are located outside cells, are abundant in fungi, and are extracellular. These enzymes are ideal for bioremediation because they are not substrate-specific [269]. These fungi are believed to oxidize PAHs initially using their peroxidases in the extracellular environment. Some PAHs are immediately oxidized by fungal lignin peroxidases, while fungal manganese peroxidases indirectly oxidize others via enzyme-mediated lignin peroxidation [270].

Hydrocarbons derived from petroleum are so pervasive that we cannot avoid them. However, they are the world's most significant pollutants. Fungi are among the many microorganisms that have demonstrated the capacity to degrade petroleum hydrocarbon effluents. Generally, fungi are more effective than bacteria at degrading petroleum effluents. The fungus consumes petroleum as a source of carbon and removes it from the environment if it grows near an oil well.

Penicillium sp., *Rhizopus* sp., *Aspergillus niger*, and *Aspergillus terreus* are the fungi species that have been tested for bioremediation of hydrocarbons. *Cochliobolus lutanus* is the newest member of this group of fungi that degrade the oil. As previously stated, *Penicillium* sp. possesses the lowest elimination potential, and *Aspergillus niger* possesses the highest bioremediation potential among the species. There are four major groups of fungi, and Deuteromycetes and Ascomycetes both have a large number of members that contribute to the bioremediation of hydrocarbons. Numerous microorganisms in diesel oil sludge tanks degrade petroleum hydrocarbons and aliphatic hydrocarbons. *Aspergillus fumigatus* was more effective than other tested microorganisms [271]. Propionic acid and other metabolites were detected in the water phase of *A. fumigatus* growth assays after 60 days of incubation.

As a beneficial species for mycoremediation, *Pleurotus ostreatus* possesses anti-oxidant, anti-cancer, immunostimulatory, anti-inflammatory, and anti-diabetic therapeutic properties [272]. Numerous studies have demonstrated that it is effective at degrading petroleum hydrocarbons. The fungus *Pleurotus ostreatus* retains many enzymes that degrade multiple compounds. *P. ostreatus* can transmit water, nutrients, bacteria, and contaminants along the mycelium, dispersing resources for contaminant degradation within the soil matrix. *P. ostreatus*'s potential to move through air-filled soil crevices, metabolise nontoxic organic molecules, and infiltrate micropores allows the fungus to thrive in conditions that inhibit bacterial growth. For optimal performance, bioremediation depends on the fungal strain and pH, temperature, oxygen availability, and nutrient concentration. Between 30 and 40 °C is the optimal temperature for optimal bioremediation results. However, bioremediation agents are active at temperatures as low as 1 °C. The effectiveness of fungal bioremediation is also determined by a moisture range between 50 and 80%.

9.3. Mycoremediation of Pesticides

In agricultural practices, farmers use pesticides to increase crop quality and yield and combat pathogens, weeds, and insects. There is no doubt that the use of pesticides has increased production, but it has also increased environmental pollution.

Multiple studies demonstrate that pesticides harm human health and the planet's flora and fauna. Numerous researchers have reported diverse neurotoxic, mutagenic, and carcinogenic effects of pesticides, as well as disruptions to the nervous, respiratory, cardiovascular, and endocrine systems and inhibition of various enzymatic activities [273].

Research is ongoing to remove pesticides from the environment utilizing various strategies, including biological and chemical methods. Considering the magnitude of this problem, physicochemical methods are now obsolete. There is evidence that microbes can degrade pesticides, and fungi have many advantages. Due to their mycelial structure, they can absorb more pesticide; It possesses an extensive range of extracellular enzymes, is abundant in nature, is not very mutagenic, and their spores can withstand adverse conditions [274]. Due to all the benefits listed above, fungi are ideal for bioremediation. White-rot fungi degrade mono-aromatic xenobiotics, such as numerous pesticides, without depoly-

merizing them. However, it needs to be made clear how ligninolytic activity is involved. The extraction of many fungi from the soil is necessary for remediation of the excessive amounts of pesticides; these fungi need to be isolated, tested and completely characterized.

Numerous studies have shown that two *Fusarium* species (*F. solani* and *F. poae*) break down lindane by employing it as a carbon source. The cytochrome P450 monooxygenase system mediates this process. *P. chrysosporium* was found to have the potential to degrade lindane, the most extensively studied pesticide for fungal bioremediation. Production of ligninolytic peroxidase is not required for the activity of this enzyme. These strains rely on lindane as a carbon source. They could utilise all tested concentrations of lindane (0.600 g/mL), but the most significant increase was observed at 100 g/mL.

It was also observed that after an incubation period of 10 days, *F. solani* and *F. poae*, degraded the 100 mg/mL pesticide by up to 59.4% and 56.7%, respectively. Additionally, lindane was a suitable carbon source for *Fusarium verticillioides*. Within five days of incubation, the fungus *Ganoderma australe* could biodegrade 3.11 mg of lindane per gram of fungal biomass. Some reports claim that *Penicillium oxalicum* uses methamidophos as a source of nitrogen. Nearly all of the initially supplied methamidophos were destroyed by *Penicillium oxalicum* within 12 days of incubation [275]. *Trametes hirsuta*, a white rot fungus, was studied to determine its capacity for endosulfan biodegradation. It was discovered that this strain could utilize and degrade endosulfan sulfate produced during endosulfan biodegradation. When fungi use this pesticide as a source of sulfur, they degrade endosulfan into almost 16 different species.

9.4. Mycoremediation of Pharmaceutical Effluents

To maintain pharmaceutical safety and quality standards, effluents must be disposed of appropriately, especially from the pharmaceutical sector. Before treatment, it is essential to classify the nature and their components since the resulting effluents may contain teratogenic, mutagenic, or cancerogenic substances for humans [276]. Bioremediation provides a benefit over alternative chemical treatments that have failed and made the water poisonous due to the production of byproducts during treatment.

9.5. Mycoremediation of Dye

The white-rot fungus *Phanerochaete chrysosporium* mineralizes various primary aromatic pollutants, including nitrotoluenes, PAHs, dioxin, and chlorophenols, in addition to lignin [277]. Three possibilities exist for the fate of adsorbed dye: (1) association with hyphal systems, (2) physical desorption to the solution, and (3) enzymatic destruction via residing hyphal structures. Dye-saturated mycelium can be rebuilt for subsequent adsorption through physical desorption and enzymatic breakdown. Utilizing active mycelium and extracellular enzymes results in the quickest regeneration. This method could be applied to the continuous treatment of wastewater using *Trametes versicolor* by sequentially adsorbing and degrading dyes through the resident fungal mycelium. *Aspergillus foetidus* rapidly settles around the interior of biomass pellets, suggesting strong bioadsorption rather than the biotransformation of colours. By utilizing the dead macrofungi *Phellinus igniarius* and *Fomes fomentarius*, Rhodamine B's carboxylic and amino compounds help to increase sorption. Rarely do colours that are being deteriorated by fungus become mineralised. After adding *P. chrysosporium* for 12 days, mineralisation (23–48%) of radiolabeled azo dyes was achieved.

10. Future Prospects

Further focused research is needed to optimise mycoremediation methods that are environmentally friendly, economical, and can be used with or without plant associations in co-contamination. This will improve remediation effectiveness and decontamination rates in aquatic and terrestrial ecosystems. In the field of mycoremediation, numerous significant future research directions could be pursued and are outlined below.

- To improve bioremediation applications, competitiveness, and practicability, it is necessary to screen new species of fungal and microbial consortia for the biodegradation of multiple contaminants with higher ecological adaptation.
- Mycoremediation is still in its infancy at the laboratory/greenhouse level, which limits its effectiveness in the field. Therefore, before commercialising this green technology, the mycoremediation abilities of each species must be evaluated in their natural environment.
- Increased study of high-throughput techniques (e.g., enzyme engineering, NGS, and microarray technologies) must be undertaken to make mycoremediation more economical and practically feasible.
- Finding different, more advantageous biological techniques (such as Phytoremediation, natural attenuation, etc.) and mechanisms for remediating stress caused by contaminants is necessary.
- Microbes' genealogy and genetic modification must be investigated to better understand the remediation mechanisms.

11. Conclusions

This review discusses mycoremediation as a low-cost, eco-friendly, and effective technique to remediate environmental pollutants using fungi or their compounds. The different mycoremediation methods (biosorption, precipitation, biotransformation, and sequestration) were presented. Some recent fungal species and their biodegradation performance under varying environmental conditions have been highlighted. These consist of their practical and distinctive ability to be used in the bioremediation of organic contaminants. Apart from the fungal community and diversity, the biological, chemical, and physical influences on fungal bioremediation and their potential importance are analysed. In addition, there is a growing interest in studying myco-nanotechnology from different ecologies for the bioremediation of pollutants. Therefore, removing contaminants from contaminated land surfaces and water bodies by combining mycoremediation with myconanotechnology and other cutting-edge technologies such as enzyme engineering, next-generation sequencing (NGS), and microarray technology has a promising future.

Based on the findings of this review and the critical questions on mycoremediation, a synergistic strategy with a positive policy framework for mycoremediation, ranging from laboratory scale to pilot scale testing involving fungal species with suitable properties, is urgently needed. This approach must be tested on a laboratory scale before being replicated in real field situations. However, the study of mycoremediation still encounters certain limitations: The process is time-consuming, and the removal of contaminants rarely reaches 100%. The bioavailability of the pollutant and the soil matrix in question influence the final treatment result. To improve the application possibilities, competitiveness, and practicality of bioremediation, it is necessary to explore using bioinformatics techniques for new species of fungal and microbial consortia with higher ecological adaptability for the biodegradation of multiple pollutants.

Among the two complex global problems of climate change and pollution, this synergistic strategy also functions as a unique tool for the future security of natural resources and the health of ecosystems.

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