

BIOREMEDIATION OF PAHS CONTAMINATED AGRICULTURAL SOIL-A REVIEW PAPER

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ABSTRACT

Soil is an important environmental matrix that directly or indirectly supports the life of all creatures. Despite being the ultimate sink for all contaminants, it has been neglected for a long time, resulting in poor soil quality. Due to the contamination of various toxic polycyclic aromatic compounds (PAHs) in soil, it diverts the quality of soil and impacts the soil ecosystem. Henceforth, it is necessary to identify the ecologically sustainable treatment alternatives for contaminated site cleanup. Biological treatment of PAHs contaminated soil is emerging as a promising and sustainable treatment options because they are safe, cost effective and eco-friendly treatment solutions. When it comes to pollutant degradation, microorganisms are known for their enzyme-catalyzed catabolic activity, which can be advantageous in the decomposition of PAHs. There are various microbes which are extensively used for the removal of PAHs, in which *Cobetia marina*, *Rhodococcus soli* and *Pseudoalteromonas agarivorans* were found to be significant in degradation of PAHs. This review paper compiled a various recent in-situ and ex-situ bioremediation techniques used for the degradation and remediation of PAHs in agricultural soil.

Keywords: Polycyclic aromatic hydrocarbons, Soil, Degradation, Techniques.

INTRODUCTION

Polycyclic aromatic hydrocarbons (PAHs) are a class of chemically toxic compounds found in large quantities in agricultural soil as a result of waste water irrigation and industrial operations (Guo et al., 2017; Fernandez-Luqueno et al., 2011). PAHs pollution is a major global problem owing to its negative consequences. High persistence of polycyclic aromatic hydrocarbons (PAHs) in microbial breakdown and its negative impacts on the environment are the motivating attention to adopt various techniques for degradation and removal of PAHs in agricultural soil. Bioremediation is recognised as a relatively recent, successful technology and at the same time it is a cost-effective method for PAHs removal in contaminated site. It is a method of removing pollutants from the environment in order to restore the natural ecosystem and avoid further pollution (Sasikumar and Papinazath, 2003). Bioremediation is a method used for

removal of PAHs from contaminated environment by utilising microorganisms to convert toxins to harmless products through mineralization, or by the conversion of contaminants into microbial biomass (Baggott, 1993; Mentzer and Eber, 1996). Toxic PAHs are mineralized or bio transformed by using specific bacteria or certain enzymes in contaminated agricultural soil. Pseudomonas, Sphingomonas, Brevibacterium, Arthrobacter, Nocardioidea and Mycobacterium bacteria have been used to breakdown PAHs, especially naphthalene (NAP) and phenanthrene (PHE) (Ghosal et al., 2016). Anaerobic biodegradation of PAHs is also extensively used for PAHs removal under denitrifying and sulphate-reducing bacteria (Lu et al., 2019). The aerobic degradation pathway is the principal oxygen-dependent pathway in surface layer soils, and it relies on genes that code for dioxygenases and monooxygenases (Yagi et al., 2009). There have been some advancements in

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bacterial remediation of contaminated soil techniques, such as inoculation with bacteria picked from PAHs-affected locations or nitrogen supplementation of contaminated soils (Cunningham *et al.*, 1996; US EPA, 2000; Suthersan, 2002; Nzila, 2013). Modern biological methods have been widely employed to improve microbial PAHs degradation efficiency and clarify biodegradation metabolic pathways. Traditional ex situ approaches such as land farming, composting, and soil piles is also widely used for PAHs removal away from the contaminated site, however ex situ methods are quite expensive.

Bioremediation techniques for removal of PAHs in agricultural soil

Biological techniques have received the most attention for PAHs cleanup, followed by integrated methods, chemical oxidation, and physical procedures (Kuppusamy *et al.*, 2017). Bioremediation is gaining acceptance as a viable alternative treatment option for PAHs contaminated soil remediation because it is regarded as a safe, effective, environmentally acceptable, and cost-effective method of eliminating organic pollutants from contaminated soil. Bioremediation of PAHs in contaminated agricultural soil is found to be a complex

phenomenon because of the multi steps followed bioremediation process, and the environmental factors associated with PAHs degradation (Bosma *et al.*, 1997; Tang *et al.*, 2005).

Types of Bioremediations

On the basis of place where PAHs compounds are removed, there are principally two ways of bioremediation:

In Situ Bioremediation

In situ bioremediation is most commonly used to remove PAHs from contaminated soils at the source point and it can frequently be done onsite, with little or no disturbance to routine operations. This also avoids the need to transfer large amounts of waste off-site, as well as the potential health and environmental risks that might occur during transit (Vidali, 2001). It is a more successful way for cleaning contaminated site, since it saves money on transportation and eliminates chemical contamination with harmless microorganisms. The procedure is also recommended since it produces the least amount of damage to the contaminated area, however it is more time-consuming than other restorative methods.

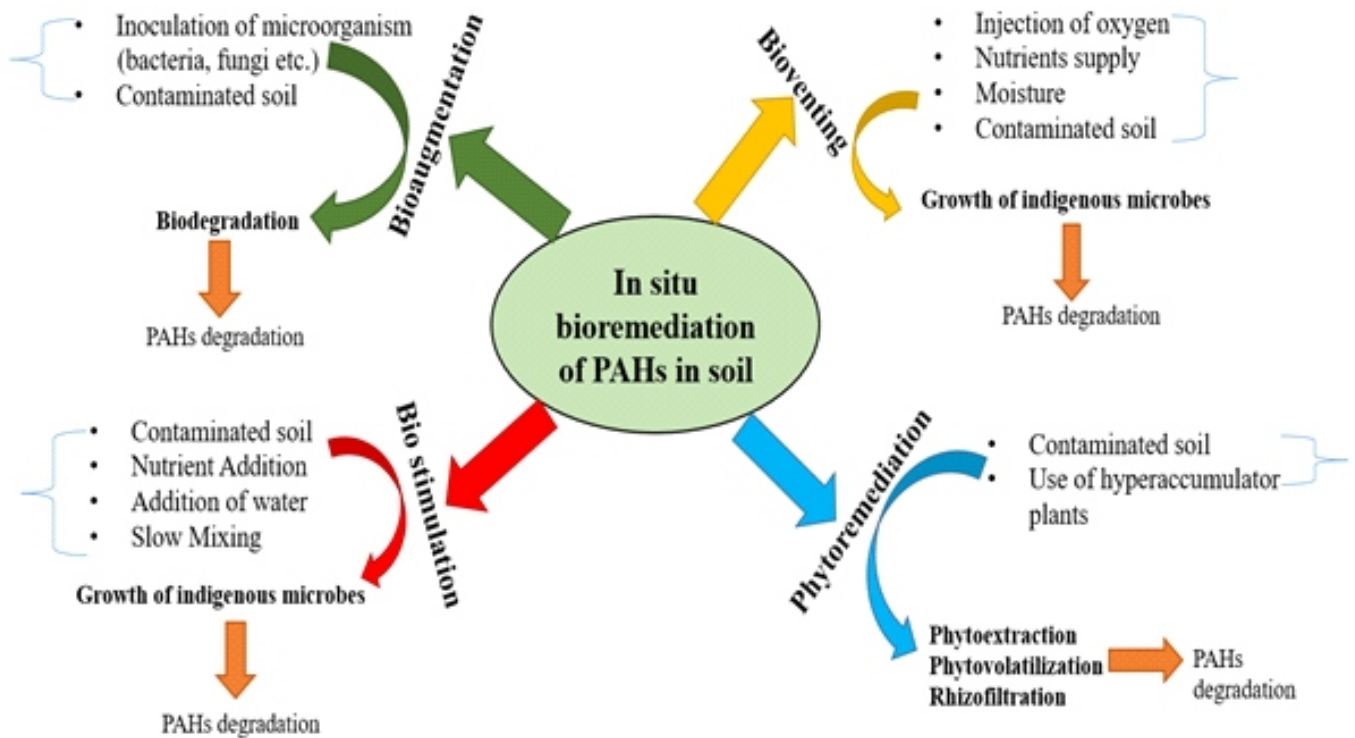


Fig. 1: In situ bioremediation techniques used for PAHs removal in agricultural soil.

Bioaugmentation

Bioaugmentation is defined as a bioremediation technique for enhancing the removal capacity of PAHs by introducing specific type of microorganisms to the polluted site for promoting biodegradation. Microbial PAHs remediation involves the application of particular microorganisms such as bacteria, archaea, fungus, and algae alone or in combination. Microbiological decontamination of PAHs contaminated soil is stated to be a more effective, cost-effective, and adaptable treatment option than physicochemical treatment (Rivas, 2006). Biodegradation of PAHs is a technique of recycling organic contaminants by employing living organisms to break down organic or inorganic matter, either in the presence of oxygen (aerobic biodegradation) or without oxygen (anaerobic biodegradation). Bioaugmentation can speed up the degradation process even in the circumstances where indigenous communities of degrading bacteria are not found (Alemzadeh, 2018; Cunningham et al., 2004). Garon et al. (2004) used fungus (*Absidia cylindrospora*) as a bioaugmentation supplement in contaminated soil to improve bioremediation in polluted soil, effected by fluorene. Bioaugmented microorganism also showed 70.8% and 72.2% reductions in benzo[a]pyrene and anthracene, while biostimulated and control microorganism showed substantially lower levels (Wu et al., 2008). Physical, chemical, and biological features of soils, as well as the physio-chemical characteristics of chemicals, have a significant impact on the degrading capability of naturally existing microbes for field bioremediation (Haritash and Kaushik, 2009). Bacteria have a metabolic flexibility that makes them ideal for degrading PAHs contaminants (Ma and Zhai, 2012). Unlike bacteria, archaea's degradation routes and methods for bioremediation have not been widely investigated (Khemili-Talbi et al., 2015). Mycoremediation, or fungal remediation, is a term that refers to the treatment or remediation of PAHs contaminated site, which involves a variety of fungal species. Some litter-decomposing fungi have been shown to colonise soil and degrade PAHs (Steffen et al., 2002), while native non-ligninolytic fungi isolated from soil have been found to significantly modify PAHs (D'Annibale et al., 2006; Potin et al., 2004).

Biostimulation

Biostimulation is a technique in which nutrients are added to a contaminated agricultural soil, to boost the growth of naturally occurring bacteria. It requires adding nutrients (nitrogen, phosphorus, and trace minerals) to the site, as well as adjusting the pH, soil moisture content, and aeration to encourage the proliferation of indigenous microorganisms and provide bacterial communities with a favourable environment in which they effectively degrade the PAHs (Salanitro et al., 1997). In order to boost the degrading

activity of native or foreign microbes, inorganic additions of macro nutrients (N, P, K) or micro nutrients (Mg, S, Fe, Cl, Zn, Mn, Cu, Na) are needed for the recovery of contaminated agricultural soil with PAHs (Samanta et al., 2002). The addition of fertilisers has been proven to increase the number and activity of microbial communities, resulting in improved soil deterioration (Breedveld and Sparrevik, 2000; Betancur-Galvis et al., 2006; Xu and Obbard, 2003). The addition of nutrients to encourage the activity of indigenous microorganisms can aid in the degradation of PAHs based contaminants (Alexander, 1999; Samanta et al., 2002).

Bioventing

The most popular in situ therapy is bioventing, which includes providing air and nutrients to the contaminated soil with PAHs via wells in order to promote the indigenous microorganisms. To increase microbial growth and activity, oxygen is drawn through the contaminated medium. Bioventing uses low air flow rates and only supplies the quantity of oxygen required for biodegradation, reducing volatilization or pollutant release into the atmosphere. It is effective for simple hydrocarbons and can be utilised in situations where pollution is found far beneath the surface (Vidali, 2001).

Phytoremediation

Phytodegradation appears to be the most promising method for organic pollutants like polycyclic aromatic hydrocarbons (PAHs), by the use of plants to extract, sequester, and detoxify polluted agricultural soil with PAHs. Hyperaccumulator plants are used in soil to remediate PAHs contamination. Phytoremediation also known as rhizoremediation, is now regarded a viable, low-cost option for treating large areas of organic chemical pollution (Susrala et al., 2002; Parrish et al., 2005). For a small number of plants, there has been evidence of increased polycyclic aromatic hydrocarbons (PAHs) breakdown in plant rhizosphere, as a result of increased microbial activity (Spriggs et al., 2005). Phytoremediation offers the following advantages over other approaches: it prevents the natural structure and texture of the soil, uses primarily solar energy, can attain large levels of microbial biomass in the soil and it is cost effective (Huang et al., 2004). Tall fescue (*Festuca arundinacea*) and switchgrass (*Panicum virgatum*) were shown to be capable of removing all PAHs with an average of 40% effectiveness, with the exception of Indeno(1,2,3-c,d)pyrene, which had a removal efficiency of only 1.5 % (Gan et al., 2009). Lee et al. (2018) discovered that the native Korean grass species *Panicum bisulcatum* and *Echinogalus crus-galli* for phytoremediation of PAHs in contaminated soil. Gan et al. (2009) chosen five tree species (red mulberry, black willow, rooted hybrid poplar, sycamore and black

locust) for phytoremediation, which have the capability of removing PAHs from soil. Liste and Alexander (2000) used three species of plant, such as jack pine (*Pinus banksiana*), red pine (*Pinus resinosa*), and white pine (*Pinus strobus*) for pyrene breakdown in soil. Phytoremediation has the following advantages over other approaches: it preserves the soil's natural structure and texture, greater abundance of microbial biomass in plant rhizosphere area, it is cost effective and has the potential to be rapid degradation for organic contaminants. Although there are certain advantages to utilising plants to remediate persistent toxins over other ways, there are a number of drawbacks to using this technology on a broad scale (US EPA, 2000; Suthersan, 2002). Many plant species are vulnerable to pollutants, particularly PAHs, which is a severe constraint (Huang et al., 2004; Burd et al., 1998). As a result, they grow slowly, making it difficult to produce enough biomass for effective soil remediation. Although land farming, bioremediation, and phytoremediation all have some efficacy in removing persistent PAHs from polluted soils.

Microbial Enzyme mediated Bioremediation

It is an advanced technique where isolated enzymes from fungus, bacteria, and other living organisms is used for PAHs elimination from contaminated soil for bioremediation. It performs over a wide range of temperatures and pH, henceforth the enzymatic activity is very efficient and selective. Enzymes involved for PAHs oxidation include

oxygenase, dehydrogenase, lignin peroxidase, manganese peroxidase, laccases, and phenoloxidases (Mohan et al., 2006). Fungi's oxidative enzymes are less substrate-specific, so they are more efficient (Harms et al., 2011; Zhang et al., 2020). The sole disadvantage of this approach is the high cost of enzyme synthesis, extraction, and purification (Kuppusamy et al., 2017). Specifically, the bacteria use oxygenase-assisted metabolism to degrade PAHs aerobically (comprising monooxygenase and dioxygenase enzymes). PAHs breakdown is carried out by the fungus using the monooxygenase enzyme (Gupta and Pathak, 2020). Ligninolytic fungi generate enzymes including lignin peroxidase, manganese peroxidase, and laccases that oxidise PAHs (Aydin et al., 2017).

Ex Situ Bioremediation

PAHs are naturally more resistant to biodegradation and stay longer in the environment for years due to the lack of a suitable endogenous microbial population and incompatible environmental circumstances, so ex situ methods are important for the degradation of PAHs in contaminated soil. Ex situ bioremediation process takes place away from the contamination site, it necessitates the transportation of contaminated soil to the bioremediation site. Ex situ bioremediation approaches adopt the inoculation of specific foreign microorganisms like bacteria and fungus with PAHs contaminated site. This approach has more drawbacks than benefits.

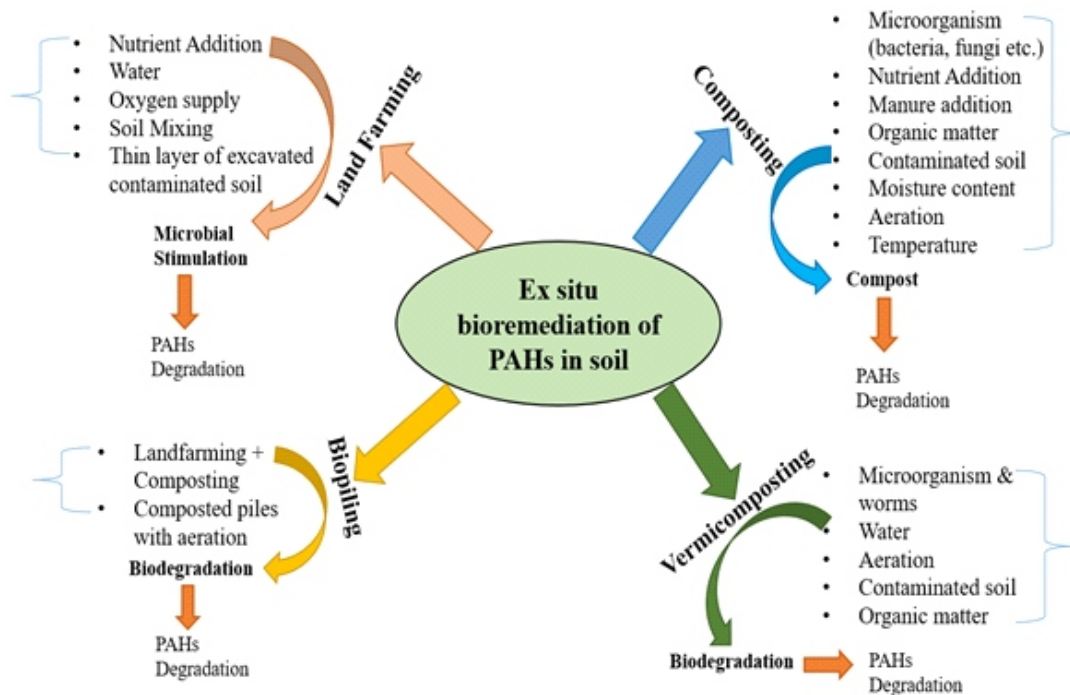


Fig. 2: Ex situ bioremediation techniques used for PAHs removal in agricultural soil.

Land farming

Land farming is a basic bioremediation approach, recently been investigated as a PAHs-remediation technology in which polluted soil is excavated and put over a prepared bed, then tilled on a regular basis to increase aeration until contaminants get degraded. The goal of this technique is to encourage indigenous bio degradative bacteria and make it easier for them to degrade pollutants aerobically. In general, the procedure is confined to treating the top 10–35 cm of soil (Vidali, 2001). Land farming will also oxygenate the soil, possibly enhancing the proliferation of aerobic soil bacteria and resulting in faster PAHs breakdown (Vidali, 2001). Land farming has a distinct advantage among other technologies for stimulating native soil microflora that are enriched in the soil by improving limiting factors such as inadequate aeration, poor microorganism contacts with the contaminants, and insufficient nutrients (Hansen et al., 2004).

Composting

Composting is the process of mixing polluted soil with non-hazardous organic additions like manure or agricultural waste. The presence of these organic materials encourages the growth of a diverse microbial population and the high temperatures associated with composting, hence promotes biodegradation of PAHs (Vidali, 2001). Temperatures in the compost range from 55 to 65 degrees Celsius. It is the decomposition of organic wastes by microbes, usually at a high temperature. Composting technology entails adding organic agents to the contaminated soil with PAHs, which increases porosity and allows for optimal air flow, optimal temperature and also providing a source of easily assimilated carbon for biomass growth (Eweis et al., 1998; Antizar-Ladislao et al., 2004). Composting technique is a successful and cost effective method for the degradation of PAHs, however it is more successful in degrading low molecular weight PAHs than high molecular weight PAHs (Cajthaml et al., 2002).

Biopiling

Land farming and composting are combined in biopiles. Engineered cells are essentially aerated with composted piles. Excavated soils are mixed with soil additives, placed on a treatment area, and bioremediated utilising forced aeration in biopile treatment, which is a full-scale technology.

Bioreactor

The polluted soil is excavated and transported to a treatment bioreactor. The bioreactor might have been built on-site or in a separate treatment facility. Ex situ method such as the use of bioreactors allow temperature and pressure control to promote PAHs breakdown in soil (Álvarez Bernal et al., 2006). Typically, the soil is slurried with water before being

treated in the reactor, which improves bioremediation conditions.

CONCLUSION

Bioremediation is the most prominence and successful techniques for the degradation of PAHs from agricultural soil, as compare to other physical and chemical techniques. Various ex situ and In situ methods have been used for the removal of PAHs from contaminated soil, for a long time. In recent year researchers are most focused to develop much efficient combinations of techniques or advancement in existing techniques to achieve the removal efficiency of PAHs from contaminated soil. PAHs in agricultural soil, damage human health and soil quality, henceforth it is necessary to identify ecologically sustainable treatment alternatives for contaminated site cleanup.

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