

Remediation of Polychlorinated Biphenyls (PCBs) through Bio and Phytoremediation in the Environment: A Review Study

Sunila Abdul Wassey¹, Sehrish Shafeeq², Jawad Abdullah Butt^{3*}, Ahmed Jamil³

¹ Sindh Environmental Protection Agency (SEPA) Karachi, Pakistan

² Department of Biochemistry, University of Karachi, Pakistan

³ Environmental Research Center, Bahria University Karachi Campus, Pakistan

* Email: jawadbutt.bukc@bahria.edu.pk

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Abstract: One of the persistent organic pollutants (POPs) utilized globally between the 1930s and 1980s was polychlorinated biphenyls (PCBs). Even though their usage has been severely constrained, a lot of PCBs are still present in the environment, including soils and air. This local, national, international and governmental organizations regarding polychlorinated biphenyls and how can resolve and treat the problem sustainably. This can be achieved if seriously working on degrading persistent organic pollutants, especially polychlorinated biphenyls biologically via thorough research and sound implementation. The methods opted for remediation by the use of choosing suitable types of microorganisms and plants. These methods are practically active for the identification of the source, concentration level and effects on the ecosystem. It would help in developing appropriate strategy and data generation in formulating the inventories for the management of polychlorinated biphenyls in the future. It is also noted through numerous research that polychlorinated biphenyls are also found in human blood commonly analyzed by indicator congeners such as PCB 138,153, 180, and in air and soil markers of PCBs pollution include PCB congeners 28, 52,101,138,153 and 180 as well. The methods and mechanisms described for degrading the persistent pollutant in this research review the solutions for mitigating PCB pollution in an environment. The best option is to strictly ban it in Pakistan or weightage its usage with its multiple benefits and disadvantages. Promote biological treatment if the payback is healthy and environmentally sustainable.

Keywords: Polychlorinated bi-Phenyls, bioremediation, phytoremediation, environment, pollution.

Introduction

Polychlorinated biphenyls (PCBs) are considered one of the most notorious persistent organic pollutants (POPs) and come under dirty dozen besides Hexa chlorobenzene, Dioxins, dichloro-Diphenyl-trichloroethane (DDT), Furans and others (Vergani et al., 2017). Since the 1930s, usage of PCBs has been started on a large scale for commercial purposes and mainly found in plastic, lubricating oils, carbonless copy paper, inks, impregnating, paints, additives, adhesives, waxes, immersion oils, fire retardants, sealing liquids, plaster and casting in different industries. Mostly found as insulating fluids in transformers and capacitors, vacuum pump fluids, and hydraulic fluids, that remain in use. Although PCBs manufacturing was banned by the USA in 1979 and 2001 by the Stockholm Convention on Persistent organic pollutants (POPs). Although the acceptance of restraining guidelines, the usage of PCBs levels is still seen in seawater, sediments of waterways and outdoor air.

PCBs are manmade, aromatic substances that are not naturally present in the environment. Their persistent nature in the environment is because of the physicochemical characteristics of the compounds and to easily affect nearby habitats through close interactions with a variety of environmental matrices, including sediments, soils, groundwater, surface water, and the food chain (Jing et al., 2018; Mao et al.,

2021). The chemical formula of PCB is represented as; $C_{12}H_{10-n}Cl_n$ (Erickson, 1997) and its chemical structure are shown in Fig. 1. Toxicity of PCB.

The persistent, accumulative, hydrophobic nature deteriorating the environment and its lipophilic nature have more affinity to bioaccumulation and bio-magnification process in an organism (Beyer et al., 2009; Devi, 2020). PCBs are a cluster of 209 congeners with an extensive variety of toxic and biological special effects. Congeners are divided into PCBs that are Dioxin-Like (DL) and Non-Dioxin-Like (NDL). Chemical examination is commonly used to analyze PCB combinations, and six PCBs that are frequently referred to as "indicator" PCBs are tested in this manner (PCBs; IUPAC Nos. 28, 52, 101, 138, 153 and 180). NDL-PCBs are less toxic than DL congeners, but these contaminants are nevertheless dangerous because they have been found at much higher concentrations than the latter congeners in the blood and tissues of animals, fish, and people who have been exposed to environmental PCBs through the food chain (Abella et al., 2016; Beyer et al., 2009). Numerous studies examined the amount of Organ Chlorines (OCs) in Pakistan's environment (Syed et al., 2011; Alamdar et al., 2014). A study results (table 1) found that PCB pollution and its indication caused by PCBs are six identified (Mahmood et al., 2014). Whereas Table 2 represented the most toxic PCBs tabulated according to their physical and chemical

properties where in only three indicators are identified out of 206 PCBs congeners.

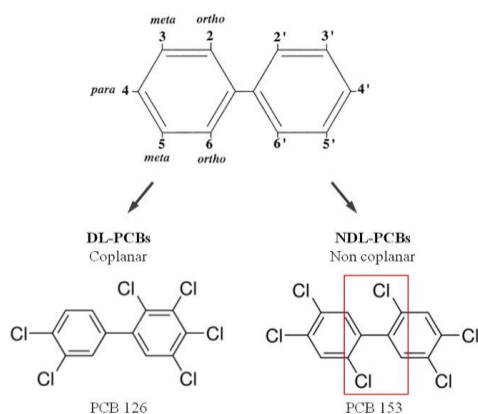


Fig. 1. Chemical structure of PCBs (Abella et al., 2016).

Table 1 Statistics analysis of PCB congeners in Soil and Air and markers of PCB pollution (Mahmood et al., 2014).

S.NO.	PCB Congeners	Soil (ng g ⁻¹)				Air (pg m ⁻³)			
		St. dev.	Mean	Min	Max	St. dev.	Mean	Min	Max
1	PCB-28	1.58	1.89	0.19	4.88	19.7	37.5	10.0	61.7
2	PCB-52	0.20	0.20	0.02	0.50	8.50	8.50	0.50	23.5
3	PCB-101	0.20	0.09	0.01	0.30	2.21	3.90	0.79	7.20
4	PCB-138	0.20	0.20	0.10	0.10	1.40	1.20	0.06	3.60
5	PCB-153	0.29	0.20	0.01	0.80	1.93	2.10	0.25	5.5
6	PCB-180	0.58	0.60	0.01	1.50	0.33	0.30	0.03	0.9

Table 2 Physical and chemical properties of some of the most toxic and environmentally predominant PCB congeners with three as markers congeners of PCB pollution.

S.NO.	Properties	PCB-138	PCB-153	PCB-180
1	Molecular Formula	C ₁₂ H ₄ Cl ₆	C ₁₂ H ₄ Cl ₆	C ₁₂ H ₃ Cl ₇
2	Boling Point	400 (Calc.)	---	240-280
3	Water Solubility g/L at 25°C	15.9 (Calc.)	0.91-0.86	0.31-6.56 (Calc.)

In three decades PCBs were extended due to their isolating and flammable properties (Erickson et al., 2011). PCBs might be discharged via accidental release, leaks, waste materials burning, and disposal activities, inadequate burning of landfills, sewage sludge (chlorine holding) and incineration sites (Kim et al., 2014). In Asia, improper mechanism of discarding municipal waste in suburban areas has likely bases POPs significantly in developing countries (Someya et al., 2010). PCB exposure via food in adults is about 2–6 ng/kg/day. In the US, 167 MSW incineration sites have 250 tons/day incineration capability or are more responsible for significant PCBs emission (Abella et al., 2016). PCBs

are consistent and stable in the environment; therefore, the US-EPA has declared them as probable human carcinogens (Judd et al., 2003) as having toxicological implications, and deleterious impacts during the last decades (Mahmood et al., 2014) and (Robertson et al., 2001).

In Pakistan, improper and unplanned agricultural and industrial development raised numerous problems relating to PCB contamination and affecting dangerously human health and ecology (Ahmed, 2003). Although the Government undersigned with Stockholm Convention to minimize the production and discharges of PCBs into the environment (Abu-Elabbas, 2013). In Punjab (Pakistan), unsympathetic use of goods and agrochemical cause PCB contamination in the environment and human being through bio-accumulation. The reconditioned products are used in the electronic market and their waste called e-waste (Naqvi et al., 2020) are re-used by illegal recycling and restoration of metals through swelting (Baqar et al., 2017) and subsequently pollute the environment through PCBs releases (Eqani et al., 2012).

Many research studie has been carried out in Pakistan to assess the status of Organo-chlorines (OCs) (Syed et al., 2011; Eqani et al., 2012; Alamdar et al., 2014), but few reports have mentioned the environmental PCB concentrations (Mahmood et al., 2014). Several studies also focused on the degradation of environmental pollutants by different bacteria which can feed exclusively on hydrocarbons (Yakimov et al., 2007).

Polychlorinated Biphenyls (PCBs) Remediation Techniques

A sustainable and technical process of degradation of PCBs in the environment depends on chemical treatment. Normally, sunshine or bacteria in the environment break down PCBs. When PCBs are in the air, shallow water, or surface soils, sunlight is crucial to their destruction process in the presence of microorganisms like bacteria, algae, or fungi biodegrader agents.

Biodegradation and Bio-Remediation

Biodegradation is a biologically declined procedure of complication of chemical complexes through which the organic materials are cracked down into simple composition via active microbial organisms and as a result, the term "mineralization" to describe mostly

biologically mediated variation in a substrate. The treatment uses naturally occurring organisms that break down the harmful substances into less toxic or innocuous substance and bring it into their natural state, this method is called biodegradation. Whereas, pollutants can be remediated by natural attenuation and biodegradable abilities of microorganisms, which can be boosted by engineering procedures, whichever by adding carefully chosen microbes (bioaugmentation) or using a biostimulation system wherever nutrients are added. Genetic work is used to increase the competencies of microbes but despite its effectiveness, there are numerous issues affecting the efficacy and associated risks of this procedure (GEM) (Joutey et al., 2013) as mentioned in Fig. 2.

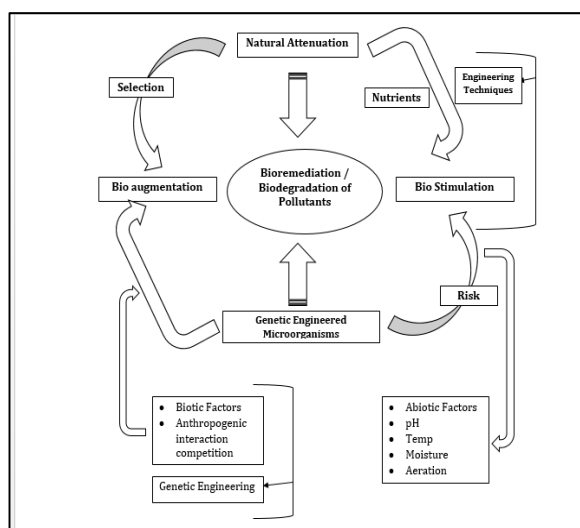


Fig 2. Pollutant bioremediation by utilizing the biodegradation abilities of microorganisms.

Biodegradable matters (plants and animals/bodies initiated from living creatures/or synthetic constituents like plants and animals place to usage by microbes) or certain of the microbes have a naturally microbial catabolic variety to transfigure a wide sort of mixtures containing Poly Aromatic Hydrocarbons (PAHs), hydrocarbons, Polychlorinated Biphenyls (PCBs) and metals (Leitao, 2009). Several published studies described PCB physiochemical treatment preferences for PCB degradation. Previously, studies for biological degradation of PCBs in which microorganisms produce such enzymes that transform the organic contaminant into simpler composites.

Biodegradation can be a mineralization or a co-metabolism. In mineralization, proficient organisms use the organic contaminant as a cause of energy and carbon as a result of the decrease of contaminant to its essential elements. Whereas, Co-metabolism requires

another constituent for the microbes and the targeted contaminant is transformed at a similar period. If the products of co-metabolism are responsive to additional degradation, then they might be mineralized then inadequate degradation produces metabolites that are further toxic than the parental molecule. Therefore, it requires conglomerate microorganisms that use as the basis of nutrients.

The effectiveness of biodegradation depends on the composition of the composite, the presence of unusual substituents, where those substituents are located within the molecule, and the quantity and solubility of the contaminants. The stable carbon-halogen bonds from microorganisms must be broken down by the aromatic halogenated chemicals under high energy conditions. Microorganisms use substitute chlorine to alter the rich characteristics of the aromatic ingredient and the electron density of specific locations in order to deactivate the complex's primary oxidation. Then substituted chlorines have stereochemical effects on the empathy between substrate molecule and their enzymes (Munawar et al., 2021).

Mechanisms for Degradation of PCBs

Microbial degradation of PCBs comprises two possible methods i.e., aerobic degradation and anaerobic de-halogenation. Aerobic biodegradation is an efficient process that may occasionally involve hydroxylation, chlorine atom removal from the biphenyl circle, ring cleavage, and oxidation of the following products. Only innocuous compounds like chlorine, carbon dioxide, and water are created when PCBs are completely aerobically mineralized (Furukawa, 2000).

Diversified microbial groups have the utmost power of bio-degrading potential since the genetic information of further than one organism is essential to reduce the complex combinations of organic composites existing in polluted parts (Passatore et al., 2014). Bacterial strains mostly gram-negative bacteria of the genus *Pseudomonas* are capable to reduce aromatic hydrocarbons insulated from the soil. On the other hand, *Aeromonas*, *Mycobacterium*, *Rhodococcus*, *Corynebacterium* and *Bacillus* genera are also considered as the Bio-Degradative pathways for degrading organic pollutants from soil. Biodegradation of PCBs ensues through several phases, alike a metabolic chain containing diverse microbial and fungal strains. PCB degradation scheme comprises two main microbial metabolic phases (Fig. 3).

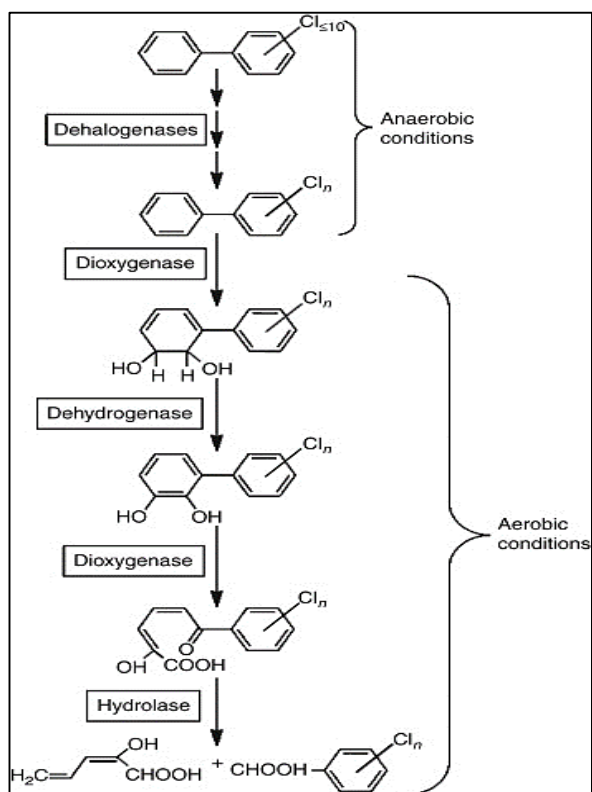


Fig. 3 Illustration of a few of the main pathways described for PCB biodegradation (Pathiraja et al., 2019).

Based on a comprehensive evaluation of previous study works, which measured the ability and growing features of facultative anaerobic microbes in degrading PCBs. Anaerobic, aerobic and two-stage cultivation environments are shown in Fig. 4.

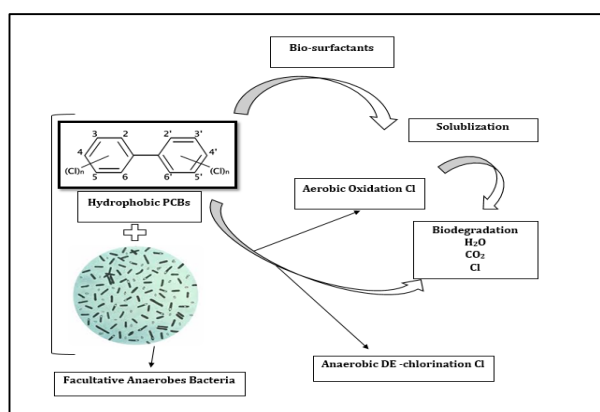


Fig. 4 Representation of some of the main pathways described for PCB biodegradation (Passatore et al., 2014). Newly Isolated Aerobic Bacteria and Anaerobic Pathways for Degrading PCBs.

Aerobic microbes cultivate quicker than anaerobes and can tolerate high degradation amounts resulting in compound mineralization. Hypothetically, the biological deficiency of PCBs would outcome to provide water, carbon dioxide and chlorine. This procedure comprises the elimination of chlorine from

the biphenyl ring trailed by oxidation and cleavage of the resultant complex (Fig. 5).

Based on the numerous bacterial strains and various primary biodegradability (depletion) of PCBs, general concepts were obtained concerning the correlation between PCB structures and biodegradability (Furukawa, 2000).

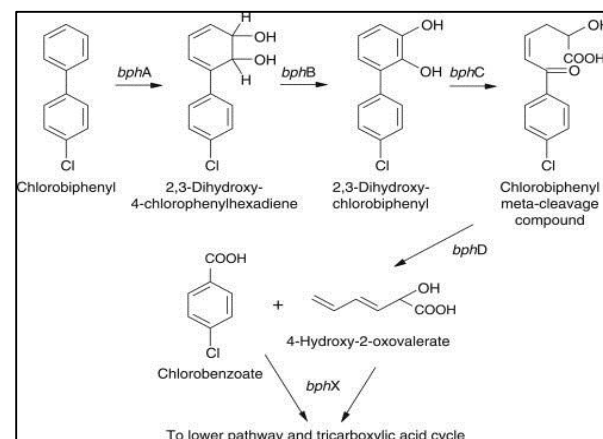


Fig. 5 Hypothetical Aerobic PCB biodegradation pathway of 4-monochlorinated biphenyl (Furukawa et al., 2004; Xiang et al., 2020; Elangovan et al., 2019).

Coupling of Anaerobic De-chlorination and Aerobic Degradation

Chlorinated PCBs compounds (electron acceptors) might be de-stabilized by anaerobic microorganisms, whereas aerobic strains degraded the benzene ring structures. Anaerobic consortia could also mineralize PCBs completely by de-chlorination (Meckenstock et al., 2016).

Combining aerobic and anaerobic processes speeds up the breakdown of highly chlorinated PCBs (Chen et al., 2014; Payne et al., 2013). This led researchers to design an experiment that involved drying food continuously while simulating a natural consecutive aerobic-anaerobic condition in a paddy field. It was found that 40% or more of the PCB congeners degraded while in constant-drying conditions degraded only 20% degradation. The action of three facultative anaerobic microorganisms was done under the anaerobic-aerobic degradation, so PCB was reduced from commercial-based PCB mixture Aroclor1260 and reduced to about 49% within 15 days (Pathiraja et al., 2019). (Payne et al., 2013) indicated that 80% of total PCB reduction takes place after 120 days in contaminated sediment with concurrently added aerobic *Paraburkholderia* LB400 and anaerobic De-Chlorinator De-Halobium DF1. According to the outcomes of the study, it is observed through review study that anaerobic and aerobic degrading microbes, and three facultative anaerobic bacteria, correspondingly, the influence of the degradation is simultaneous behavior mode was noticed to be

improved comparison of a consecutive procedure (Pathiraja et al., 2019; Payne et al., 2013).

Phytoremediation of PCBs

The process of cleaning up pollution with plants as the treatment of contaminated sites or a variety of organic compounds can be removed including PCBs, a method called phytoremediation (Aken et al., 2010). This process can be done by autotrophic organisms, sunlight and CO₂. Through various natural allelochemicals and xenobiotic compounds, plants have developed varied decontamination of organic compounds (Singer et al., 2003). This process is cost-effective and in simple terms, it is economical green technology through which, the materials in the soil become assimilated, metabolized, adsorbed, or removed (Wu et al., 2015). The photo-degradation and phytoextraction subsets of this technique are the most advanced for the phytoremediation of organics and metals in contaminated soils (Mani et al., 2014; Lee, 2013).

Phytoremediation Process

The studies on the bioremediation of PCBs by plants i.e. Reinholtz and Volpe, (1977) (aquatic plants), Weber and Mrozek, (1979), (uptake and translocation), Schwartz and Lehmann, (1982), (PCBs detection in plant tissues), and Bacci and Gaggi, (1985), were published for the first time in the late 1970s and early 1980s (volatilization and translocation of PCBs from soil). Later, tremendous progress was made, revealing the potential of plants and associated microbes for PCB degradation. Practices accepted in the phytoremediation of PCBs contain Phyto-extraction, Rhizo-remediation, and Phyto-transformation (Aken et al., 2010) and the process of phytoremediation of PCBs, organic pollutants is well-described in Fig. 6. This is also well explained by the fact that contaminants in soil and groundwater may be taken up by plant tissues (Phyto-Extraction) or adsorbed to the roots (Rhizo-filtration); contaminants in soil may be broken down by microbes in the root zone (Rhizo-remediation); and contaminants inside plant tissues may be converted to other substances by plant enzymes (Phyto- transformation) or may be volatilized into the atmosphere (phytovolatilization).

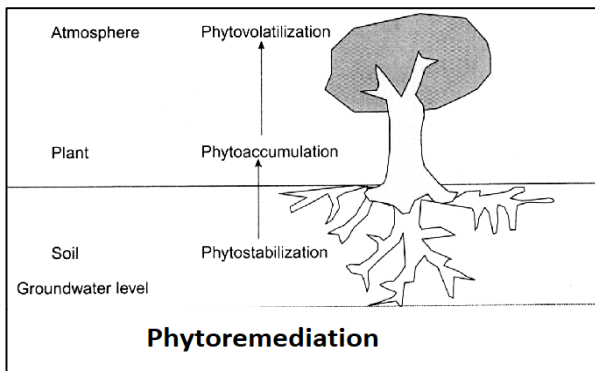


Fig. 6. Phytoremediation of organic pollutants, PCBs (Aken et al., 2010)

PCB reduction has been more detected in vegetated soils rather than in non-vegetated soils. Phenolic compounds including flavonoids and terpenes in roots and have been testified and PCBs were degraded by microbial activity in the soil. There is also an experiment done for testing phytoremediation of PCB-contaminated soil in the greenhouse by various plant kinds including tall fescue, reed canary grass, switchgrass, and alfalfa. It was found that Aroclor 1248 removed from all vegetated pots to a greater extent i.e 38 percent or not as much of PCB recovery, as compared to non-planted vessels where PCB retrieval was 82%. The effectiveness of PCB by plants can be decreased at a faster rate if the degree of chlorination increases. It is correlated with the enzymatic activities of plants in soil and levels of biodegradation of PCBs, based on their log Kow. Aroclor 1260 in contaminated soil was decontaminated by several plant species selected from three sites in Canada and exhibited distinct concentrations of PCBs in root tissues and less significant in shoot tissues. Despite estimations based on log Kow, hepta- and non-chlorinated biphenyls were found in shoots even though substantial amounts of tetra- to Hexachlorinated biphenyls were found. This demonstrated the rise in chlorinated compounds absorbed by plant roots. Whereas when present in plant tissues, PCBs were probably degraded by enzymes.

It was reported that the PCBs can be transformed via plant cell cultures (*Rosa* sp.) in the plants and revealed that more than 10% of 11 distinctive congeners were metabolized. As a result, numerous studies demonstrated that the breakdown of mono- to Tetra-Chlorinated congeners with the various mono- and Di-Hydroxylated metabolites have been discovered and confirmed using whole plants and plant cell cultures (Chroma et al., 2003; Harms et al., 2003). For instance, *Rosa* species and *Lactuca sativa* species cell cultures were used to evaluate the Mono-Hydroxylation of 3, 3', 4, 4'-Tetrachlorobiphenyl. There is a very little awareness about the metabolism of PCBs by plant enzymes like oxygenases, cytochrome, P-450 mono-oxygenases and peroxidase. The de-chlorination of 2, 2', 4, 4', and 5, 5'-hexachlorinated biphenyl was reported by the crude extract of *Medicago sativa*, even though no plant de-halogenated has yet been identified (Field et al., 2008).

Phytoremediation Status in Pakistan

Pakistan is rigorously polluted due to the consistent expulsion of industrial and domestic waste. Over-population and irregular industrialization alarm the environmental pollution level that threaten the environmental condition in general and deteriorate the public health as well as natural ecosystem (Ashaiekh et al., 2019; You et al., 2015). The waste water effluent from various sources contains both organic and inorganic pollutants (Arora et al., 2018; Bharagava et al., 2018). Chlorinated phenols, Azo dyes, Endocrine-disrupting chemicals, Polychlorinated Biphenyls, Poly-

aromatic Hydrocarbons, and Pesticides along with toxic metals (cadmium, chromium, mercury, lead, arsenic, etc.) that are remarkably persistent in the atmosphere and non-biodegradable. The poor biodegradability and high concentration of organic pollutants in waste water pose serious environmental hazards (Saxena et al., 2015 & 2016).

Using bacteria and plants to degrade PCBs and other POPs from contaminated soil and water is another promising strategy for environmental sustainability in Pakistan. Table 3 displays a list of papers on plants and microbes for the phytoremediation of PCBs. Through the detoxification and degradation of POPs/PCBs, the bacteria promote the growth of plants. Additionally, they enhance plant health and growth due to their unique processes for stimulating plant growth (Arslan et al., 2017).

Table 3. Phytoremediation of PCBs by plants and bacteria (Aken et al., 2010).

Pollutants PCBs	Species	Organism
PCBs	<i>Pseudomonas</i> sp. (strain LB400)	Sugar beed seed
PCBs	<i>Comamonas testosterone</i> B-356)	Tobacco (<i>Nicotiana tobacum</i>)
2,3,4-Trichloro-biphenyl	<i>Pseudomonas xenovorans</i> LB 400	<i>Sinorhizobium meliloti</i> colonizing
3-Chloro-biphenyl (PCBs)	<i>P. xenovorans</i> LB 400 <i>P. aeruginosa</i> strain 142	Alfalfa (<i>Medicago sativa</i>) Alfalfa roots
Individual PCBs congeners and contaminated soil	<i>Burkholderia xenovorans</i> LB 400	Colonizing Alfalfa (<i>Medicago sativa</i>) <i>Pseudomonas fluorescens</i> colonizing
4-chloro-biphenyl	<i>Burkholderia xenovorans</i> LB 400	<i>Nicotiana tobacum</i> , <i>Nicotiana benthamiana</i>
2,3-dihydroxy-bipgenyl	<i>Pseudomonas testosteroni</i> B-356	Tobacco (<i>Nicotiana tobacum</i>)

The direct sources of water and soil contamination in Pakistan are mainly the agriculture sector. The excessive uses of fertilizers, pesticides, and herbicides, spillages of hazardous substances and disposal problems of urban and industrial waste are the main causes of persistent pollutants as well as heavy metals pollution. Thus, a variety of phytoremediation strategies have been applied globally to clean contaminated soil and water. These technologies, which employ a variety of plant species to adapt to detoxify metals and chemical substances are undoubtedly promising. Pakistan has a unique terrain with numerous weather zones and a wide variety of

flowers. Additionally, more than 6,000 species of higher plants have been found for the phytoremediation process. Currently, more than 400 angiosperm plant species have been examined and identified as hyperaccumulators globally (Lee, 2013; Kamran et al., 2014).

Conclusion

This study reviewed the importance and treatment mechanisms of PCB remediation by biological methods. Microbial degradation and phytoremediation have effectiveness but limited remediation because of the long duration of remediation whereas; the major benefit is the relatively less implementation costs and environmentally friendly technologies. PCBs have complex structures so understanding their physical and chemical properties is much more important to know. So that, their fate and transport thereby selecting appropriate remediation approaches can easily be achieved. Polychlorinated Biphenyls (PCBs) are one of the principal members in the group of Persistent Organic Pollutants (POPs). PCBs have severe environmental and health effects, which can include immune system changes, reproductive impairment carcinogenicity and effects on wildlife that cause biological diversity loss. In keeping its accessibility wide-reaching and its environmental impacts worldwide, the proposed review is very much fruitful to evaluate the environmental risks related to PCBs and can identify the concentration level of PCBs. Environmental pollutant removal and the renewal of contaminated sites are major targets for current societies. As biodegradation process based on the catabolic action of PCBs-degrading microbes is deliberated as an environmentally friendly, economically, and encouraging approach to remediate PCBs pollution. It is essential for us to better comprehend the transference and outcome of PCBs in the environments and enzymes involved in the degradation trials of PCBs.

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