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Review on the role of recombinant DNA technology on environmental sanitation

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Abstract
A method for treating contaminated media, such as soil, subterranean material, and water, called bioremediation involves changing the environment in a way that encourages the growth of microorganisms while reducing the amount of the target pollutants. Evidence suggests that bioremediation is more cost-effective and environmentally friendly than other remediation options. Microbial and phytoremediation are two different types of bioremediations. Living microorganisms are utilized in microbial bioremediation to break down hazardous chemicals into innocuous by-products of cellular metabolism like CO2 and H2O. However, in phytoremediation, contaminated soil and water are removed using plants. Thanks to specialized jumping genes, microbes can evolve biological tolerance to any environmental toxin. Ex-situ and in-situ techniques are used to perform bioremediation on contaminated soils. Ex-situ is the term used to describe the removal of toxins from soil and water, whereas in situ is the treatment of contaminated places. Successful bioremediation has utilized GE microorganisms, recombinant DNA, and RNA technology. New metabolic pathways have been developed by modifying microbial genes to improve bioremediation procedures. The heavy metal that can be released into the environment that is the most hazardous is mercury. Mercury may be taken out of contaminated sediment, soil, or water using the GE Escherichia coli strain JM109. A place contaminated with mercury can be cleaned up using GE bacteria that have the Mer A gene. Bioremediation has been applied to transgenic plants such as *Arabidopsis thaliana*, *Nicotiana tabacum*, *Brassica juncea*, *Brassica oleracea var botrytis*, and *Lycopersicon esculentum* that express cytochrome P450 enzymes and have the potential to remove pollutants from soil and water. Mer A and Mer B-expressing transgenic plants can extract mercury and transfer it to the shoot. The metabolic breakdown of TCE from contaminated locations was accelerated by the genetically modified tobacco plants that expressed human cytochrome P4502E1. Two key elements influencing bioremediation procedures are the nature of the pollutants and the environmental circumstances. To increase bioremediation rates on contaminated sites, environmental conditions need to be changed. Therefore, the aim of this review is to highlight the use of recombinant DNA technologies in environmental sanitation.

Keywords: Bioremediations, DNA, recombinant, sanitation, environment

1. Introduction
Due to the population's rapid growth, more natural resources are being exploited to meet the population's high need for food, energy, and other necessities. The industrial revolution was a solution to these needs, but it also led to the manufacturing of a vast array of different organic and inorganic compounds, which have both directly and indirectly contributed to the ongoing contamination of habitats. Agriculture, industry, business, government, military, residential activities, and other human activities all contribute to the environmental release of these hazardous wastes (Chaudhry, 1994 [13].

The trend of environmental degradation is so rapid and pervasive that detectable levels of contamination are even found in the deepest ocean waters. Only around 10% of all waste was safely disposed of, according to estimates made by the Environmental Protection Agency (EPA) (Reddy and Mathew 2001) [51].

These wastes are overburdening the ecosphere on Earth, and the rate at which biodiversity is vanishing is worrying. When Rachel Carson wrote about how the use of pesticides like DDT damaged people's health and wiped-out animals to such an extent that spring arrived without the sound of birds, she was exposing the deadly effects of dangerous chemicals. This was in her 1962 book, Silent Spring. The United States banned DDT because of Carson's findings on the harmful impact of DDT residues on bird populations. Pesticides' observed ability to cause cancer was later discovered.

Then, hundreds of cases of paralysis and sensory loss were reported around Japan's Minamata Bay due to mercury poisoning. Thousands of individuals in Japan and Taiwan, respectively, were exposed to polychlorinated biphenyls (PCBs) through contaminated cooking oil in the late 1960s and mid-1970s. Exposure to a high concentration of the chemical resulted in miscarriage and congenital defects (Azad *et al.*, 2014) [5].

Twelve chemicals the so-called "Dirty Dozen" were outlawed in a deal that was finalized in December 2000 at the Stockholm Convention, which involved 122 nations. Eight of these (Aldrin, chlordane, DDT, dieldrin, endrin, heptachlor, minex, and toxaphene) are pesticides, two (PCBs and hexachlorobenzene) are industrial chemicals, and two (dioxins and furans) are byproducts of combustion and industrial processes. The first ever global agreement to ban a class of chemicals was this treaty. Most of the dirty dozen were still being used in India and Latin America despite being prohibited in industrialized nations (Hill, 2004) [29].

Physical, chemical, and physicochemical methods have been found to be ineffective, inadequate, and uneconomical for managing a sizable number of hazardous wastes. Here, the intermediate product that has been degraded occasionally might be more harmful, and the environmental issues might be partially replaced. Therefore, it is crucial to use biotechnological advances in the treatment of hazardous waste. In the field of biological research, new methods have been developed and used at an astonishing rate. The biotechnology used here ranges from traditional biotechnology (food fermentation, biological control, etc.) to contemporary biotechnology based on recombinant DNA technology, bioinformatics, DNA microarray, bioprocess technology, and immunoassay (Singh, 2015) [58].

As a result, bioremediation is a method for treating polluted media, such as soil, subsurface material, and water, by changing the environment to encourage the growth of microorganisms and reduce the concentration of the target pollutants. Compared to other remediation options, evidence suggests that bioremediation is more affordable and environmentally friendly (EPA, 2011) [24]. Alternatively, bioremediation is the use of microorganisms to sequester, degrade, or eliminate environmental toxins; as a result, bioremediation technology is gaining popularity. Compared to other traditional approaches of pollutant cleanup, it offers more efficient, targeted, alternative, and environmentally beneficial procedures. It all revolves on three techniques (phyto-, microbial, and nanotechnology-based remediation) that biodegrade diverse refractory chemicals and xenobiotics into simple organic molecules, salts, carbon dioxide, water, and other benign things (Ahluwalia and Sekhon, 2012) [2].

2. Review on Role of Recombinant Dna in Environmental Sanitation

Ecosystem health can be evaluated using biotechnology. Pollutants can be converted into benign compounds, renewable resources can be exploited to produce biodegradable materials, and environmentally friendly production and disposal techniques can be developed. To increase efficiency and cut costs, environmental biotechnology uses adequately qualified live creatures and genetic engineering. These two characteristics will be crucial in the widespread use of organisms in the future to lessen the burden of harmful compounds on the environment. Researchers have developed a method termed bioremediation, a developing strategy to restore areas damaged by pollution or in other ways due to ecosystem mismanagement, in response to the pressing need for an effective environmental biotechnology procedure (Azad *et al.*, 2014) [5].

2.1 Bioremediation

Two words make up the phrase "bioremediation": "bios" (which means life and refers to living things) and "to remediate" (which means to address a problem). The term "bio remediate" refers to the employment of biological organisms to address environmental issues such polluted soil or groundwater. Utilizing living microorganisms to remove toxins from the environment or stop pollution is known as bioremediation. In other terms, it is an environmental pollution removal technology (Sasikumar and Papinazath, 2003) [54].

According to Bennett *et al*. (2002) [9], harmful substances are transformed, degraded, or otherwise rendered harmless through the employment of biological systems. Inoculation of exogenous organisms into the site is another option for bioremediation, as is using the local microbial community with or without nutritional supplementation. It is now possible to clean up soil, water, and air by using a variety of bioremediation techniques to detoxify hazardous contaminants. Microbial and phytoremediation are two different types of bioremediations. Microbial bioremediation uses living microorganisms to break down hazardous substances into harmless by-products of cellular metabolism including CO2 and H2O. However, plants are utilized in phytoremediation to remove contaminants from the soil and water (Thakur, 2006) [63].

2.2 Bioremediation technique

Ex-situ and in-situ site of application bioremediation techniques can be used, at least on the surface When selecting a bioremediation method, factors such as the type of pollutant, the depth and volume of pollution, the type of environment, the location, the cost, and environmental regulations are considered. Temperature, pH, oxygen and nutrition levels, along with other abiotic factors, all affect how effective the bioremediation process is (Smith *et al.*, 2015) [59].

In Situ Bioremediation: The contaminated matrix is not removed when using bioremediation procedures "in place". To remove the contaminants from polluted soils and groundwater, in situ bioremediation is typically used. The fact that it uses harmless microbes to remove chemical contaminations and reduces transportation costs makes it a better way for cleaning contaminated settings. The chemotactic affinity of these bacteria towards pollutants should be positive. The likelihood of bioremediation is increased by this characteristic in nearby areas where bio-remediates have not yet been spread (Tarla *et al*., 2020) [62].

The approach is also chosen because it doesn't disturb the contaminated area as much. This would be especially important in locations with high levels of hazardous contaminants, such as those contaminated with radioactive or chemical substances, or in places where pollution and investment are discouraged (like factories). The ability to simultaneously treat soil and groundwater is another benefit of in situ bioremediation. However, there are some drawbacks to in situ bioremediation, including the fact that it takes longer than other remedial techniques, changes in seasonal microbial activity because of exposure to uncontrollable environmental factors, and the potential for further issues with the use of additives (Azad *et al*., 2014) [5].

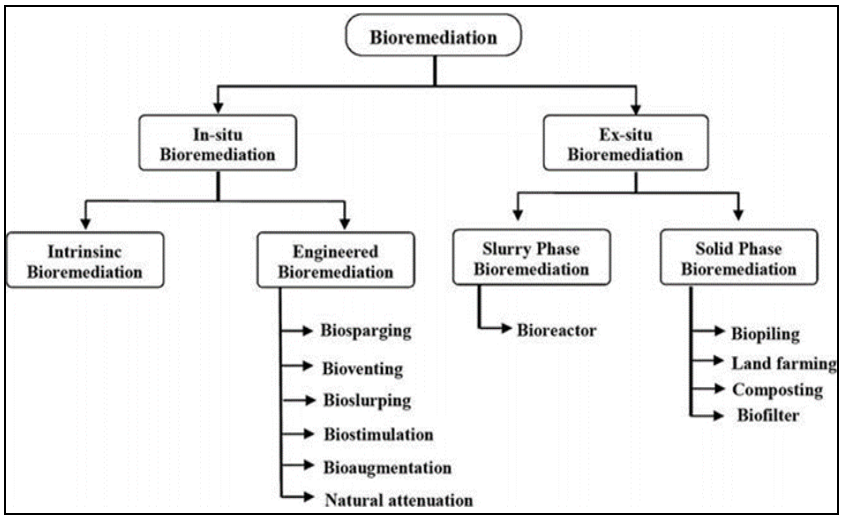

Fig 1: classification of bioremediation techniques Source: Smith *et al*., (2015) [59].

The type of waste materials determines the yield of bioremediation; specifically, if wastes could supply the necessary nutrients and energy, then microorganisms could bio remediate. However, the loss of bioactivity may be made up for by stimulating native bacteria in the absence of beneficial wastes. Applying genetically modified bacteria is another option with less appeal (Tarla *et al*., 2020) [62]. Based on where the microorganisms used as bio remediates come from, two different types of in situ bioremediation are defined.

Intrinsic bioremediation: This technique for in situ bioremediation entails altering the ecological conditions of the contaminated area without directly introducing new microorganisms, bolstering Indigenous populations, and enhancing the metabolic activities of native or naturally occurring micro fauna by providing better nutrition and ventilation (Azad *et al.,* 2014) [5].

Engineered in situ bioremediation: This type of bioremediation includes introducing microorganisms to a contaminated site. Because the conditions at pollution sites are commonly unfavorable for the establishment and bioactivity of the exogenously modified microorganisms, in this case, as in intrinsic bioremediation, the environment is changed in a way that improves the physio-chemical conditions. Oxygen, electron acceptors, and nutrients (such as nitrogen and phosphorus) are required for the acceleration of microbial growth. (Singh, 2015) [58]. Bioaugmentation comprises the addition (augmentation) of specialized microbial cultures, which are often grown separately under well-defined circumstances, to each environment (in situ or in a bioreactor) to carry out a particular restorative activity (Alvarez and Illman, 2006) [3]. Since the 1800s, agriculture has adopted this tactic, for instance by introducing nitrogen-fixing Rhizobium organisms to legume roots (Gentry *et al.*, 2004) [28] and is now increasingly being used to enhance the biodegradation of recalcitrant organic pollutants in groundwater and soils.

There have been two different Bioaugmentation strategies created. One is based on the addition or replacement of the native microbe population by the injection of microorganisms with the required catabolic capacity. In this instance, the chosen bacteria or consortia can thrive in the contaminated environment, outcompete local microorganisms, and occupy a particular metabolic niche (Vogel and Walter, 2002) [67]. The second Bioaugmentation method involves the insertion of a lot of cells that work briefly as biocatalysts and break down a lot of the target pollutant before going dormant or dying (Duba *et al.*, 1996) [22].

The inoculated cells are unable to thrive in situ in these circumstances due to the intrinsic abiotic and biological stress present in the new environment, which includes changes in temperature, pH, water activity, low levels of nutrients, harmful pollutant concentrations, and competition from local microbes (Gentry *et al*., 2004) [28]. To avoid this, regular biomass re-injection is required throughout time in these circumstances (Silva *et al.*, 2004) [56].

Mycoremediation: Mycoremediation is the term used to describe fungus-mediated remediation. Extracellular enzymes secreted by fungal cells break down harmful wastes to provide energy for their growth and development. The Earth's carbon cycle's most significant degradative event is likely the effective decomposition of lignocellulose. Extracellular enzymes are being used more frequently to test the fungi's ability to degrade no cellulosic wastes such plastic, petroleum hydrocarbon pollution, dyes, pesticides, and nutritional wastes. *Phanerochaete chrysosporium*, *Pleurotus floridia*, *Trametes hirsute*, and *Ceriporiopsis subvermispora* are a few of the fungi that are increasingly exploited in the degradation of hazardous wastes. Because of their flexible enzymatic system, *Pleurotus* species are currently employed to degrade synthetic colors effectively (Singh *et al.*, 2008) [57]. The mycelium and spores of many fungi can absorb Cd, Cu, Pb, Hg, and Zn. Sometimes dead fungus has stronger walls than living ones. For the treatment of U and Th, systems utilizing *Rhizopus arrhizus* have been created (Bennett *et al.*, 2002) [9].

2.2.1 Mechanisms of bioremediation by genetically engineered microbes

Microbes with specialized jumping genes can develop biological tolerance to any environmental poison. For the bioremediation of contaminated soils, both ex-situ and in-situ approaches are employed. Ex-situ means taking toxic substances out of the environment, whereas in situ means treating contaminated areas (Vidali, 2001) [66]. Since ex-situ procedures are expensive and produce ineffective metal extraction results, they have been utilized for soil excavation and groundwater purification. In situ is a practical and

environmentally sound method for the indirect reduction of metal by biologically produced H2S by sulphate-reducing bacteria. This strategy makes use of naturally occurring biogeochemical processes through bioremediation. It can immobilize or, to varying degrees, remove toxins rather than moving them from one environmental medium to another (Sari and Tuzen, 2009) [53].

When biological, chemical, and physical mechanisms combine to lessen the toxicity and mobility of subsurface contamination, this is known as natural attenuation. The bioremediation process can be impacted by a few microbiological activities. Because positively charged metal ions are drawn to negatively charge microbial cell membranes, the microbial activity known as bio sorption can affect bioremediation and remove them. With the help of bacteria' biosorption, heavy metals like Pb and Cd have been eliminated from aqueous solutions (Sari and Tuzen, 2009) [53].

Another method for promoting the bioremediation of contaminated places is called "bio stimulation," which entails enhancing the growth and development of microorganisms by modifying the pH, quantities of nutrients, and oxygen. By introducing genetically modified bacteria to boost the activity of insufficient native microbes, Bioaugmentation can aid in the bioremediation of contaminated environments (Vidali, 2001; Silva *et al.*, 2004) [66, 56].

On their exterior surface, most bacteria release polysaccharides that harden into slime and capsules. In contaminated locations, bacteria can, under the right circumstances, reduce the metal and oxide materials. While they cannot break down inorganic metals, microorganisms can alter their oxidation states. To detoxify metals from their inorganic to organic forms and back again, microbial reduction systems can be used. Microbes can turn harmful materials into energy. As a result, they are capable of geometric growth and, upon disintegration, enormous biomass production both aerobic and anaerobic (Azad *et al.*, 2014) [5].

Through aerobic and anaerobic respiration, they can degrade complex hydrocarbons. Enough oxygen is needed for the aerobic process, which is a quicker and more complete system. Unlike methane and hydrogen sulphide, it does not produce undesirable byproducts. Complex hydrocarbons are transformed into smaller molecules by the anaerobic process, a biological process. Higher rates of waste molecule destruction require anaerobic microorganisms, which are crucial since they are inexpensive and require little energy (Singh, 2015) [58].

2.2.2 Genetically engineered microbes for remediation

Microbes like yeast, filamentous fungi, and bacteria, according to environmental biotechnology, may be able to extract heavy metals from aqueous solutions. Utilizing the metabolic power of microorganisms allows for the safe and cost-effective removal of toxins from contaminated areas. GE microorganisms, recombinant DNA, and RNA technology have all been used for efficient bioremediation. Microbial genes have been manipulated to develop novel metabolic pathways, which will boost bioremediation processes. GE microorganisms might be the best approach because of the distinctive features of their metabolic pathways (Azad *et al*., 2014) [5]. The use of GE microbes, a cutting-edge technology, to remove heavy metals and toxic waste from contaminated areas has attracted public attention (Shukla *et al.*, 2010) [55]. Additionally, it has aided in the removal of heavy metals and other resistant substances (Muhammad *et al*., 2008) [45].

Bacteria can help turn dangerous forms of heavy metals into less toxic ones by using their metal regulatory genes. GE microorganisms that express metallothioneins (MT) can hasten the accumulation of heavy metals (Jan *et al.*, 2009) [32]. The most dangerous heavy metal that can be released into the environment is mercury. Mercury can be eliminated from contaminated water, soil, or sediment using the GE Escherichia coli strain JM109. Mercury can be removed from a contaminated site using GE bacteria that possess the MerA gene. Effective mercury bioremediation can be facilitated by transgenic bacteria that express metallothioneins and polyphosphate kinase (Ruiz *et al.*, 2011) [67].

GE Organic pollutants in contaminated locations can be broken down by *Deinococcus radiodurans* and *Pseudomonas putidia*. It has been established that using organophosphates as pesticides in agriculture seriously damages the ecosystem. GE bacteria may break down chlorinated organic substances like lindane and trichloroethylene (Kumar *et al.*, 1996) [36]. The chemical lindane (c-hexachlorocyclohexane) is bad for the environment and people. More than 98% of the lindane in paddy fields may be broken down by the recombinant Anabaena in about 6–10 days. By introducing several phenol catabolic genes (pheA, pheB, pheC, pheD, and pheR) into their transformed forms, GE E. coli and P. putida have been shown to digest trichloroethylene (Marconi *et al*., 1997) [42]. The ability of GE P. putida S12 to digest naphthalene, toluene, and biphenyl was demonstrated by Marconi *et al*. (1997) [42] after the insertion of plasmids harbouring genes for the catabolism of these pollutants. Industrial effluent may contain the metal chromium (Cr), which is extremely carcinogenic. Cr may be removed from industrial effluent using genetically modified microorganisms such *Ralston metallidurans*. Cadmium (Cd) can be eliminated from industrial effluent by the recombinant *Caulobacter* species strain JS4022/p723- 6H (Patel *et al.*, 2010) [48].

Arsenic (As) is a very poisonous metal that is present in nature. Arsenic can be eliminated from contaminated soil by GE bacteria that express the ArsM gene by volatilization (Liu- *et al.*, 2011) [39]. When present in contaminated soil, ArsR-expressing E. coli can facilitate the bioaccumulation of Arsenic. The GE E. coli SE5000 strain can absorb nickel (Ni), which is arguably the most difficult to remove from the environment, from an aqueous solution (Fulkerson *et al.*, 1998) [26]. Recombinant DNA technology is a promising technique for producing GEO that may resist environmental toxins for effective bioremediation. In 1985, Chakraborty published the first DNA-based bioremediation method for petroleum-related contamination. The removal of heavy metal, chlorinated hydrocarbon, pesticide, petroleum hydrocarbon, and explosive pollution from polluted locations is possible with the help of this technology. A potent mutagenesis method known as DNA shuffling can produce novel enzymes and biocatalysts with faster rates of degrading polyaromatic hydrocarbons and chlorinated ethane (Canada *et al.*, 2002) [10]. Numerous studies have shown that horizontal gene transfer, a part of bacterial evolution, has played a significant influence in this field (Dennis, 2005) [17]. In the larger context of horizontal gene transfer among bacteria, the horizontal transmission of recombinant DNA is regarded as natural and is likely a common occurrence (Davison, 2002) [16]. The density of the microorganisms affects horizontal gene transfer. The likelihood of genetic exchange between recombinant and native microorganisms is probable, but the risks of such genetic exchange will depend on the features

involved. The introduction of GEO may indirectly affect local wildlife and plants. By integrating plasmid addition methods into the cells, such as antisense RNA-regulated plasmid addition and protein plasmid addition, horizontal gene transfer to other extant bacteria may be avoided. Furthermore, according to Davison (2002) [16],

GEOs do not survive long when released into the environment and are broken down before any environmental effects have taken place. In laboratory research, horizontal gene transfer rates could be higher than in the natural world. Recombinant DNA technology can make it easier to create a variety of degradative pathways for the partial or complete breakdown of harmful contaminants. This feature of recombinant DNA might be the preferable method for the breakdown of xenobiotic contamination. Dioxygenases and monooxygenases have been developed using DNA methods for bioremediation. This technology can enhance catabolic pathways to counteract the harmful effects of contaminants (Mason *et al.*, 1997) [43].

Proetsky *et al.* (2005) [68] developed an RNA technology for bioremediation processes such as Sulphur oxidation, assimilation of C1 compounds, and the acquisition of nitrogen. For the synthesis of cDNA, RNA technology is applied in groundwater remediation (Parro *et al*., 2007) [47]. Trichloroethene (TCE) can be broken down by microbial communities that have 16SrRNA sequences. The most crucial method for lowering sulphates on contaminated locations is phylogenetic oligonucleotide. The phylogenetic characterization of bacteria that can be exploited for bioremediation of contaminated locations can really be obtained from 16S RRNA genes. With the decrease of Fe3, microorganisms with the 16S RRNA sequence, like the *Geobacter* species, can oxidize organic pollutants (Fennell *et al.*, 2001) [25].

2.2.3 Mechanisms of bioremediation by genetically engineered plants

Inexpensively and sustainably, various plant species can clean up soil and water. Heavy metal detoxification may be facilitated by some putative cellular and molecular pathways found in plants. Compounds may be eliminated or changed into biologically inert forms as part of phytoremediation. The idea of removing heavy metals and other chemicals via metal-accumulating plants was first proposed in 1983 (Chaney *et al.*, 1997) [12]. On contaminated locations, plants can breakdown pollutants in an efficient and environmentally friendly manner. According to Pollard *et al.* (2002) [49], plants can solubilize metals from the soil, take them up into their roots, and then transfer them to their shoots. The solubilization and uptake of metals can be aided by the chelating substances that some plants release into their root zones. The excretion of organic acids by several hyper-accumulator plants may help metal uptake (Ciurli *et al*., 2014) [14]. Plant roots can draw up metals or radioactive contaminants from the soil, contaminated water, or wastewater, which can then be transported and stored in various sections of the plant. Heavy metals like Pb and Cd from polluted soil can be eliminated using phytoextraction methods. This technique can also be used to eliminate excessive selenium (Se) (Eapen *et al*., 2006) [23]. Through the management of arsenic (As), zinc (Zn), copper (Cd), and uranium (U) in polluted areas, Phyto stabilization is a stabilizing process that can lower the bioavailability of dangerous elements in the soil. Metal absorption, clearance, and translocation capabilities are mediated by a variety of genes. Transgenic plants have MT genes that enable them to transport heavy metals from contaminated areas and synthesize peptides with 60–80 amino acids, 9–16 cysteine residues. For each metal, specific mechanisms for absorption, translocation, and sequestration should be devised. To increase the accumulation of zinc, calcium (Ca), cadmium (Cd), and manganese (Mn), metal transporter genes like ZAT and CAX-2 have been incorporated into transgenic plants (Hirschi *et al.*, 2000) [30].

2.2.4 Genetically engineered plants for remediation

Physical and chemical remediation methods present significant challenges in the removal of hazardous materials from contaminated sites because of their high costs and complex nature. Due to its low cost, high efficiency, and environmental friendliness, phytoremediation is superior to other methods. Through genetic engineering techniques, transgenic plants can take advantage of molecular detoxification mechanisms. Metal uptake, removal, translocation, and bioaccumulation are all regulated by many genes. These genes for metal uptake and accumulation have been transferred into transgenic plants. (Abhilash *et al.*, 2009) [1].

Transgenic plants expressing bacterial, or mammalian genes can be used in xenobiotic metabolism for effective phytoremediation (Kurumata *et al*., 2005) [38]. Transgenic plants expressing cytochrome P450 enzymes have the potential to remove pollutants from soil and water (Kumar, 2012) [37]. A transgenic cauliflower displayed a 16-fold greater cadmium deposition after the yeast CUP1 gene was added (Sriprang and Murooka, 2006) [68]. When mercury ion reductase was added to the roots of *A. thaliana*, this hazardous ion could be absorbed, and the amount of volatile mercury could be reduced. Transgenic *B. juncea* showed greater Cd, Cr, Cu, Pb, and Zn accumulation than wild type plants following the introduction of foreign genes (Zhu *et al*., 1999) [68]. Herbicide resistance was seen in transgenic rice plants expressing human cytochrome P450 genes, and they also removed agrochemicals from the soil (Kawahigashi *et al.*, 2007) [35]. Atrazine, chlorotoluron, and pyriminobac methyl tolerance was increased in transgenic potato plants that expressed human CYP1A1. Chloroacetanilide herbicide phytoremediation has been carried out using transgenic tobacco plants expressing maize glutathione S-transferase 1. High levels of trinitrotoluene were eliminated from transgenic tobacco plants that expressed type I nitro reductase (Karavangeli *et al.*, 2005; Travis *et al.*, 2007) [34, 65].

Cytochrome P450 and glutathione S-transferase are two important enzyme groups that contribute significantly to the increased breakdown of herbicides. According to Macek *et al.* (2008) [40], transgenic plants can lessen the buildup of agrochemicals in the environment and may help prevent and lessen chemical contamination. As a result, potentially hazardous locations could be turned into secure agricultural land. Transgenic trees have a faster rate of compound uptake and can improve the metabolism of organic contaminants. As a result of their robust development, vast root systems, and substantial biomass, they would be appropriate for bioremediation. For the removal of volatile hydrocarbons, poplar hybrids expressing rabbit CYP2E1 have been employed, and transgenic aspen trees were better at absorbing TNT than naturally occurring aspen trees (Doty *et al.*, 2007) [20].

According to Strand *et al*. (2003) [61], levels in areas planted with the control plant remained stable while transgenic plants like *A. thaliana* eliminated RDX (hexahydro-1, 3, 5-trinitro-1,3,5 triazine) from the chosen polluted regions. Mercury removal and transport are both possible in transgenic plants that express the merA and merB genes. The metabolic breakdown of TCE from contaminated locations was accelerated by genetically modified tobacco plants that expressed the human cytochrome P4502E1 (Doty *et al*., 2000) [21]. For instance, transgenic plants that have had xenobiotic degradation genes inserted into their roots can break down environmental contaminants. ACC deaminase-expressing transgenic plants can lower ethylene levels (Arshad *et al.*, 2007) [4].

Table 1: The development of transgenic plants for bioremediation

Name of genes	enzymes	Source of genes	Target plants
Biphenyl dioxygenase gene	Biphenyl dioxygenase	B. *xenovorans*	N. *tabacum*
bphc	Biphenyl catabolic enzyme	*Pandoraea pnomenusa*	N. *tabacum*
CYP71A10	Cytochrome p450	*Glycine max*	N. *tabacum*
Mn peroxidase	peroxidase	C. *versicolor*	N. *tabacum*
Tpxl	Peroxidase	L. *esculentum*	L. *esculentum*
Xpla and xplb	Cytochrome p450 monooxygenase	*Rhodococcus Rhodochrous*	A. *thaliana*
onr	Pentaerythritol tetranitrate reductase	*Enterobacter cloacae*	N. *tabacum*

Source: Doty S. (2008) [19]; Meagher R. (2000) [44]; Mackova *et al.,* (2006) [41].

3. Factors Influencing Bioremediation of Contaminated Sites: The nature of the pollutants and the environmental circumstances are the two main determinants that affect bioremediation processes. According to Gavrilescu (2005) [27], the nature of pollutants includes the chemical make-up of the contamination as well as its physical condition, such as concentration, solid, liquid, or gaseous, and chemical bond type. The environmental conditions present at contaminated locations affect how well bioremediation works. Different environmental conditions, such as temperature, pH, nutrients, oxygen, and water content, can affect the growth and activity of bacteria. To increase the rate of bioremediation on contaminated sites, environmental conditions should be changed (Baptista *et al*., 2005) [8]. The decomposition of residual hydrocarbons and microbial colonization are both significantly influenced by temperature. Temperature affects the decomposition of contaminated sites by altering the characteristics of oils, microorganisms, and hydrocarbon solubility. The rate of chlorophenol degradation was reduced below 20 C; the ideal temperature for this process is 30 C (Cho *et al.*, 2000) [14]. According to Balks *et al*. (2002) [7], PH is an important environmental factor that affects the competition between metabolic ions and the activity of functional groups in biomass. The effectiveness of bioremediation of contaminated sites is influenced by soil attributes, including soil texture, nutritional circumstances, moisture content, temperature, and levels of organic matter. These may have an impact on how well plants absorb pollutants. Compared to temperate regions, tropical countries may be better suited for phytoremediation (Kamath *et al.*, 2004) [33].

4. Conclusions

Significant prospects for the removal of environmental toxins have been made possible through transgenic techniques. Comparatively speaking, this technology is more affordable and environmentally friendly than traditional ones. It is important to consider environmental conditions that may affect the biodegradation of polluted sites. Additionally, GEO should be used and contained safely for bioremediation while following the correct regulatory processes. There are, however, several current challenges, including the dispersal of transgenic pollen, the horizontal transmission of plasmids among bacteria, and the poor survivability of GEO and transgenic plants.

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