

PHYTOREMEDIATION-REVIEW ON BIOTECHNOLOGICAL APPROACH FOR REMEDICATION OF EMERGING POLLUTANTS

Abstract

Exponential growth in human population, urbanization, and industrialization have led to a rise in environmental pollutants globally. These pollutants emerge as a result of the increase in human population as well as advancement in the use and application of agricultural chemical pesticides, medicinal drugs, personal care products (PPCPs), plastic polymers, and heavy metals. Although, the intent of this compound is to improve agricultural yields to ensure food and material supply to satiate the ever-growing need for a growing human population and also human health and better life in general. However, advancement in large-scale production of these compounds results in the generation and release of waste products to terrestrial and aquatic environments as a consequence of which this leads to devastating effects for the entire ecosystems. For addressing the environmental issue environmental-friendly and sustainable means of remediating contaminants is indispensable. To resolve this, biological strategy i.e. bioremediation (plants and microbes) can be utilized for removing this toxic material from the environment. However, extensive release of these pollutants which may be due to natural processes and human economic activity has a severe and challenging impact on the environment nowadays where leakage or accidental discharge of these hazardous contaminants are serious problems. The biodegradation capacity of the natural microbiome is insufficient in combating them. In this review, we will be focusing on the potential of plants and biotechnological

Author

Lalfelpuii Ruth
Department of Biotechnology
Pachhunga University College
Aizawl, Mizoram, India.
ruthpuii_kix@yahoo.co.in

exploitation for the improvement of plant's ability to tolerate different pollutants and phytoremediation efficiency and highlight future challenges.

Keywords: Phytoremediation, hyperaccumulator plants, transgenic plants, xenobiotics, oxidative stress

I. INTRODUCTION

The term "Phyto-" denotes plants, and "-remediation" signifies the reversal of damage. Therefore, phytoremediation refers to the use of plants for the remediation or cleanup of environmental pollutants. This approach involves the cultivation of plants in contaminated areas to either absorb or break down pollutants (1). It is often hailed as the "Green Revolution" in the realm of innovative cleanup technologies. Although the concept of using metal-accumulating plants to eliminate heavy metals and other compounds was first introduced in 1983, it has actually been practiced for the past 300 years in the treatment of wastewater discharges (69).

Phytoremediation employs various mechanisms to remediate pollutants from the environment. Phytoremediation techniques can be categorized as follows:

- **Phytostabilization:** This involves the use of plants to decrease the mobility of metals in the soil through processes like absorption and precipitation, thus reducing their bioavailability.
- **Phytoextraction:** This process entails plants extracting metals from the soil and concentrating them within their roots and shoots.
- **Phytovolatilization:** Contaminants are taken up by plants and subsequently released into the atmosphere through transpiration.
- **Phytodegradation:** This refers to the degradation of organic pollutants into less toxic forms, either in the soil or within the plant's tissues (2-7).

Phytoremediation, a natural biological process for degrading xenobiotic and recalcitrant compounds that accumulate in the environment is an emerging, eco-friendly green engineering phyto-technology where hyper-accumulator plants with their natural ability to remediate these pollutants are selected or are genetically engineered to improve their ability to tolerate as well as improve their ability to directly uptake pollutants from surface water, groundwater, soil, and sediments. Phytoremediation has received increased attention for sequestering and mineralizing organic and inorganic compounds present in contaminated soil which is of worldwide concern, and is applicable for a wide range of contaminants viz; heavy metals, radionuclides, organic compounds like chlorinated solvents, polycyclic aromatic hydrocarbons, pesticides/insecticides, explosive and surfactants, thus, prove to be an effective, economical and socially accepted technology (8-10). Conventional remediation technologies that use physiochemical and chemical methods are costly and difficult to implement, slow in the process, and emphasize separation rather than eliminating the hazardous substance from contaminated sites as a result of which causes a buildup of secondary pollutants damaging soil fertility that negatively impacts the agricultural environment (11,12). Because of their metabolic capacities, plants and microbes can both live in contaminated environments and break down contaminants [13, 14]. However, the natural processes of microbes and plants are unable to remove contamination caused by heavy metals like mercury. Heavy metals have carcinogenic effects and can cause DNA damage in humans and animals due to their mutagenic ability (15-17). Recalcitrant heavy metals area potential threat as they are nondegradable and stay in the soil for centuries and cleanup of heavy metal contaminated sites is obligatory to abate entry of toxic elements into the food chain. Development of genetically engineered plants by transfer or overexpression of detoxifying

genes or metal chelator genes into a candidate plant can improve the phytoremediation traits of hyperaccumulator plants (18-20), hence, could solve these problems.

II. TRANSGENIC APPROACH FOR BIOREMEDIATION- USE OF TRANSGENIC PLANTS

Genetically engineered organisms offer environmentally friendly and cost-effective alternatives for managing and eliminating pollutants in contaminated sites. Within plant cells, there exist cellular and molecular mechanisms with the potential for detoxifying pollutants by either removing or converting them into biologically inactive forms. The concept of using hyper-accumulator plants for removing heavy metals and other compounds was introduced in 1983 (22). Plants possess the ability to uptake pollutants from the soil through their roots and transport them to the above-ground parts (23). Some plants release compounds into their root zones, which can chelate metals, aiding in their solubilization and uptake (24), a process regulated by numerous genes. It is essential to introduce these genes responsible for pollutant mitigation into hyperaccumulator plants. For incorporating hyperaccumulation traits into fast-growing, high-biomass plants, traditional breeding techniques such as plant hybridization are utilized (64). Somatic hybridization allows for the transfer of the metal hyperaccumulation trait to high biomass plants. Notably, somatic hybrids derived from *T. caerulea* and *B. napus*, combining enhanced hyperaccumulation capabilities with increased biomass production (65), have demonstrated the capacity to accumulate substantial levels of Zn and Cd. Compared to traditional breeding, genetic engineering offers advantages in modifying plants with desirable traits for phytoremediation. Moreover, it enables the transfer of desirable genes from hyperaccumulator species to sexually incompatible plant species (66,67), showing promise in the field of phytoremediation. By employing *Agrobacterium tumefaciens*-mediated plant transformation, these genes can be introduced and expressed in recipient plants (25). Genes responsible for various metal-related processes, including uptake, removal, translocation, and bioaccumulation, have been successfully identified (26-28). Through the transfer or overexpression of these genes in candidate plants, transgenic plants with enhanced abilities to degrade xenobiotics and accumulate metals have been developed (29). Remarkably, transgenic plants have demonstrated the capability to degrade chlorinated solvents, explosives, and phenolic substances (30-32). For instance, transgenic cauliflower that incorporates the Yeast CUP1 gene accumulates cadmium levels 16-fold higher than unmodified cauliflower (33, 34).

In addition to cadmium tolerance, two novel rice genes, HPP (heavy metal-associated plant protein) and HIPP (heavy metal-associated isoprenylated plant protein), have been identified, conferring tolerance to copper, zinc, cadmium, and manganese (33). The co-expression of vacuolar proton pump (V-PPase) with a Na/proton antiporter (NHX1 transporter) enhances copper tolerance and accumulation in transgenic tobacco (34). The expression of Cytochrome P450 genes in transgenic plants has the potential to remove pollutants from soil and water (35). Moreover, various genes, including the bacterial biphenylchlorophenyldioxygenase gene (bphC), CYP71A10, Mn peroxidase gene, pentaerythritoltetranitratereductase (onr) gene in tobacco, basic peroxidase (tpx1) gene in tomato plants, and Cytochrome P450 monooxygenase (XplA and XplB) gene in *Arabidopsis thaliana*, have been generated as potential tools for phytoremediation of hazardous contaminants (36-41).

Furthermore, the introduction of the bacterial *merA* gene into the roots of *A. thaliana* has enabled the absorption of toxic mercury ions and their conversion into less hazardous volatile mercury (42). Transgenic *B. juncea*, overexpressing γ -glutamylcysteine synthetase, has exhibited greater tolerance and accumulation of cadmium, chromium, copper, lead, and zinc compared to wild-type plants (43).

Transgenic *Arabidopsis* plants were able to transport oxyanion arsenate to above ground levels, where it was subsequently reduced to arsenite and sequestered into thiol peptide complexes. This was achieved through the introduction of *E. coli* *ArsC* and γ -ECS genes (21). Heavy metal tolerance in plants is closely linked to the strength of their oxidative stress defense systems. Heavy metals can trigger the excessive production of reactive oxygen species (ROS), leading to oxidative stress. To bolster antioxidant activity, a common approach is to overexpress genes involved in the antioxidant machinery (68). Modifying oxidative stress-related enzymes can result in enhanced metal tolerance (44). The insertion of xenobiotic degradation genes into the root system of transgenic plants facilitates the degradation of pollutants in contaminated sites (45-46). By expressing ACC in transgenic plants, it was observed that ethylene levels were reduced (47). Phytotoxic nitroaromatic explosives, which are challenging for non-transgenic plants to deal with, can be more effectively remediated using transgenic plants when bacterial genes involved in their degradation are expressed (48). Reports indicate that metal transporter genes like ZAT and CAX-2 genes in transgenic plants enhance the accumulation of zinc, calcium, cadmium, and manganese (49,50). Altering oxidative stress-related enzymes can also lead to improved metal tolerance (51). Aluminium (Al) toxicity can inhibit root elongation (52-56). The introduction of the *AtGR1* gene in transgenic plants resulted in more rapid root elongation, even under various concentrations of Al treatment. This suggests that *AtGR1* gene expression alleviated Al-induced root growth inhibition by mitigating Al-induced oxidative stress, offering an effective approach to enhance Al tolerance (57).

Identifying and introducing metal transporter genes that encode transporter molecules capable of enhancing the plant's capacity to absorb metal ions represent a promising approach in phytoremediation. Several plant metal transporters have been identified, including the *Arabidopsis* *IRT1* gene, which encodes a protein regulating the uptake of iron and other metals (58), and the *MRP1* gene, which encodes the Mg-ATPase transporter (59). Co-expressing two bacterial genes, arsenate reductase (*ArsC*) and γ -glutamylcysteine synthetase (γ -ECS), in *Arabidopsis* plants significantly increased arsenic tolerance compared to wild-type plants or plants expressing γ -ECS or *ArsC* alone (60). Additionally, the overexpression of the *YCF1* yeast protein in *Arabidopsis thaliana* enhanced tolerance and increased the accumulation of Cd and Pb (61).

III. PERSPECTIVE

The existence of hazardous toxic substances in the environment has an excessive negative impact on the overall health of living organisms. The persistent nonbiodegradable nature of heavy metals could enter the food chain which might result in the rapid accumulation of these pollutants in living organisms through biomagnification (63). It also decreases soil richness altering nutrient cycling. Thus, efficient, environmental friendly and economical technologies are indispensable to promote detoxification in the recovery of affected biomes and for mitigation of pollutants from contaminated sites. Identification of

promising plant species and specific gene for detoxification and then transferring those genes to other species using genetic engineering tool can significantly enhanced the detoxification capabilities of hyperaccumulator plants as a result of which it can lead to more effective contaminated sites reclamation. Already existing scientific studies of several genes and the use of techniques for pollutant degradation provide hope for developing novel transgenic plants with improved tolerance to heavy metals and for detoxification or degradation of toxic substances into recipients with increased adaptability. Discovering novel genes that can break down new contaminants is an urgency to create new transgenic organisms that can remediate pollutants in a proficient manner as the industry continues to grow and there is an exponential increase in the amount of toxic material generated from these industries on a consistent basis. For the eco-rehabilitation of toxic recalcitrant substances, phytoremediation proves to be a promising technique. Further investigations must be carried out in this area to enhance our knowledge to identify genes and clarify metabolites and their mechanisms and their capacity to combat pollutants using modern scientific technology which can aid in discovering novel genes and metabolites for efficient phytoremediation of pollutants by transgenic hyperaccumulator plants (62). Understanding the underlying mechanism of the intrinsic detoxification methods, phytoremediation using transgenic plants will provide environmental friendly alternative to conventional remediation methods.

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