



Role of Microbial Enzymes in Environmental Bioremediation

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Abstract

The issue of environmental pollution by industrial waste, agricultural chemicals, synthetic plastics and oil products like petroleum hydrocarbons has become very serious worldwide because of its resistance to damage and toxicity. Traditional physicochemical cleaning methods often have problems of high cost of operation, ineffective removal, and causes secondary environmental pollution. For these reasons, bioremediation has become a viable alternative being environmentally friendly and sustainable and therefore finds application in the detoxification of polluted environments by using biological systems. Enzyme based bioremediation has gained much strong attention among other bioremediation strategies due to its high specificity, effectiveness and the capacity to convert complex and resistant pollutants into less harmful or environmentally friendly products. The role of the major microbial enzymes; oxidoreductases, peroxidases, hydrolases, oxygenases, dehalogenases as well as plastic-degrading enzymes are carefully analyzed in terms of their mechanism and environmental impacts.

Keywords: Microbial enzymes; Environmental bioremediation; Pollution control; Wastewater treatment; Sustainable remediation

Introduction

High rates of industrialization, urbanization, as well as intensive farmland farming have led to a continued discharge of dangerous pollutants into the land and water systems. Industrial effluents, petroleum hydrocarbons, pesticides, synthetic dyes, chlorinated compounds as well as plastic wastes are major concerns because such substances are chemically stable, toxic, and immune to natural degradation mechanisms.² These long-lasting pollutants are stored in the environment and present severe health hazards to human beings, biodiversity as well as ecological stability, such as carcinogenicity and mutagenicity.³

There have been classical methods of remediation that include excavation, sonication with chemicals, incineration, and physical segregation to control the polluted areas. However, these approaches are usually linked to high operation costs, partial elimination of polluted goods and the production of by-products that are toxic in nature.⁴ This leads to the growing demand of more environmentally friendly and cost-effective solutions which are capable of providing effective detoxification of the pollutants.

Bioremediation is an eco-friendly and effective method, which has been identified that makes use of biological systems to decompose or convert environmental pollutants to less toxic products.⁵ The process takes advantage of the metabolic capacity of microorganisms, plants, or their enzymatic systems to clean the contaminated environments either in the natural state or under controlled conditions. The enzyme-based bioremediation method is one of the many strategies that have received much interest because of its high specificity, catalytic efficiency, and independent action without need to grow cells.⁶

Microbial enzymes are the most important in the degradation of stubborn pollutants by reaction, in the form of oxidation, reduction, hydrolysis and dehalogenation.⁷ Complex organic pollutants which otherwise cannot be easily broken down in the environment can be broken down by enzyme classes such as oxidoreductases, hydrolases, oxygenases, and dehalogenases.⁸ This review is dedicated to the bioremediation using enzymes, their uses in the environmental cleanup, modes of action, and their potential in sustainable pollution management.

Concept of Bioremediation

Bioremediation is a process of restoration of environmental contaminants by the use of biological systems to degrade or detoxify or convert them into less harmful substances. This method uses the natural metabolic capabilities of microorganisms, plants, or their enzymatic constituents to purify contaminated environments either under laboratory or natural

environments.⁹ Biochemical reactions play the role of stimulating bioremediation processes whereby complex organic pollutants are changed to simple compounds which ultimately mineralize into carbon dioxide, water and inorganic salts.

There are two broad categories of bioremediation, intrinsic bioremediation, in which naturally grown microorganisms remove the contaminants without human help, and engineered bioremediation, in which the activity of microorganisms is enhanced through nutrient addition, aeration, or bioaugmentation.¹⁰ Environmental factors that are very critical to the success of bioremediation include the temperature, pH, availability of oxygen, availability of nutrients and the chemical nature of the pollutants.

Contaminants are degraded by microorganisms through either carbon and energy utilization or through co-metabolic mechanisms. Microbial strains change pollutants incidentally during the metabolism of other substrates.¹¹ Oxidation, reduction, hydrolysis and dehalogenation are among the enzymatic reactions that are predominant in these degradation processes. The overall rate and degree of removing the pollutants depend on the specificity and effectiveness of such enzymatic reactions.

The benefits of bioremediation over traditional ways of remediation are the cheaper, minimal disturbance to the environment and possibility of total detoxification of the pollutants.¹² However, it can be limited by its inefficient performance by adverse environmental factors or the unavailability of contaminants. Biochemical and ecological mechanisms in bioremediation are therefore important in optimizing the enzyme-based remedies in environmental restoration practices.

Types of Bioremediations

Bioremediation is a conceptual term that involves various operations that can be used to control environmental pollution using biological agents. Bioremediation may be categorized as microbial bioremediation, phytoremediation as well as enzyme-based bioremediation, depending on the organisms or biological component involved in contamination.¹³ Both methods differ in their mechanism, suitability and effectiveness with regard to site conditions and characteristics of pollutants.

I. Microbial Bioremediation

Microbial bioremediation involves utilizing bacteria, fungi or algae to decompose or neutralize pollutants in the environment. Microorganisms have different metabolic processes that allow them to consume organic pollutants as a way of obtaining carbon and energy or transform them through co-metabolic activities.¹⁴ Application of this method has been widely used in the

remediation of petroleum hydrocarbons, pesticides, industrial solvents and organic wastes. However, some microbial remediation may be limited by pollutant toxicity, nutrient insufficiency and unfavorable environmental conditions.¹⁵

2. Phytoremediation

The concept of phytoremediation utilizes vegetation to absorb, accumulate, stabilize or degrade contaminants within the soil and waters. These ways of using plants to treat contaminants are Phyto extraction, phytodegradation, Phyto stabilization, and rhizofiltration.¹⁶ This process is considered eco-friendly, and visually acceptable but in most cases limited by low rate of remediation, low depth of root penetration and intolerance of plants to heavy levels of contaminants.¹⁷

3. Enzyme-Based Bioremediation

In enzyme-based bioremediation isolated or immobilized enzymes are used to catalyze the breakdown of pollutants and no living cells are used. This methodology would overcome the restrictions caused by microbial survival and growth and as a result could afford to cure problems in extreme conditions like high toxicity, temperature, or pH.¹⁸ Microorganisms 'enzymes are highly specific to their substrate and catalytic, therefore suitable for the selective elimination of a pollutant. The advances in immobilization of enzymes and protein engineering has also enhanced the enzyme stability and reusability in the environmental applications.¹⁹ Overall, enzyme assisted bioremediation is emerging as a promising and manageable technique of environmental clean-up that can be combined with other biological or physicochemical techniques.²⁰

4. Enzyme Based Bioremediation

Enzyme based bioremediation is a advanced biological remediation technique, which uses isolated/immobilized enzymes to catalyze the transformation of environmental pollutants into less toxic or environmentally products. Unlike the whole cell microbial remediation this method does not rely on microbial growth, survival, and adaptability thus making it highly effective even to extreme environmental conditions, including high toxicity levels, high salinity, high temperatures or high pH levels.²¹

Microbial enzymes are highly specific biocatalysts that can be used to accelerate degradation reactions. These enzymes are involved in the catalysis of several important biochemical reactions such as oxidation

reduction, hydrolysis, oxygen addition, and dehalogenation which are most critical in the degeneration of structurally complex and persistent pollutants.²² Enzymes are highly specific and hence decrease the production of unwanted by products and allow selective remediation.

Enzyme based remediation may be installed in either in situ, with enzymes added into contaminated environment or ex situ where the contaminated material is exposed to enzymes in a controlled reactor.²³ Enzymes can be either used freely, or they may be attached on solid surfaces e.g. alginate beads, silica matrices or nanoparticles to improve stability, allow reuse and increase resistance to environmental pressure.²⁴

The recent developments in the field of biotechnology have helped to bring about a great enhancement in the relevancy of enzyme-based remediation. Protein engineering, directed evolution and metagenomic screening techniques have enabled the development of enzymes with improved catalytic activity, have broader substrate specificity and can survive in unfavorable environments.²⁵ Also, there has been better performance of enzyme consortia and multi-enzyme systems over that of single enzymes as it allows reactions of complex pollutants sequentially.²⁶

Enzyme based bioremediation despite its advantage has certain challenges among which are the cost of production, enzymes instability, and the challenge in applying it in large regions. However, research that has been done on continuous basis to improve enzyme immobilization, carrier formulation and cost-effective systems of enzyme production is still going on to eliminate these limits.²⁷ As a result, bioremediation through enzymes is increasingly recognized as a effective and practical approach of managing environmental pollution.

Role of Microbial Enzymes in Bioremediation

As they catalyze the specific biochemical responses that lead to the transformation and detoxification of the environmental pollutants, microbial enzymes are the main agents of enzyme-based bioremediation. These enzymes have a great substrate specificity and catalytic potential and allow the breakdown of structurally complex and persistent contaminants that remain in polluted environments.²⁸ Below are the key examples of enzymes involved in bioremediation.

1. Oxidoreductases

The oxidoreductases are one of the best examined types of

enzymes in bioremediation of the environment. These enzymes involve oxidation reduction reactions in which they are catalysts, which help break down aromatic compounds, phenols, dyes, and other industrial pollutants.²⁹ Laccases are also excellent examples of oxidoreductases that can oxidize a variety of organic substrates with the assistance of molecular oxygen as an electron donor. They are highly applicable in both wastewater treatment and dye degradation procedures because of their wide range of substrates apart from being applicable without any other cofactor in the initial process.³⁰

3. Peroxidases

Lignin peroxidase and manganese peroxidase as well as versatile peroxidase are important in the breakdown of high molecular weight aromatic pollutants, e.g. polycyclic aromatic hydrocarbons and lignin like compounds.³¹ These enzymes enhance oxidative processes in the presence of hydrogen peroxide to produce radicals that start the dissolution of the complicated pollutant structures. Fungal peroxidases have been found to be very effective in breakdown of persistent organic pollutants found in industrial effluents.³²

4. Hydrolases

Hydrolases are used to catalyze the hydrolytic binding of chemical bonds like ester, amide and glycosidic bonds. Particularly important are the enzymes like esterases, lipases, proteases, and amidases in the disintegration of pesticides, oils and polymeric compounds.³³ Hydrolases are utilized extensively to treat agricultural soils that are being affected by organophosphate and carbamates pesticides because they are able to lessen toxicity by cleaving of bonds.³⁴

5. Oxygenases

Important enzymes in biodegradation of petroleum based and hydrocarbons pollutants are oxygenases. These enzymes add molecular oxygen to organic compounds and trigger the degradation of the aliphatic hydrocarbons and aromatic hydrocarbons.³⁵ Monooxygenases and dioxygenases are usually related to microbial degradation processes of the crude oil molecules and polycyclic aromatic hydrocarbons that are also important in the bioremediation of oil spills.³⁶

6. Dehalogenases

Dehalogenases used as catalysts to dehalogenize the halogenated organic substances, the most toxic and long lived pollutants in the environment. These enzymes turn chlorinated and brominated compounds into less toxic compounds which are more easily degraded by microorganisms.³⁷ The dehalogenases are highly active in the treatment of contaminated ground water containing chlorinated solvents and industrial chemicals.³⁸

2. Plastic Degrading Enzymes

The identification of plastic degrading enzymes has provided new opportunities to solve the problem of plastic pollution. PETase, MHETase, cutinases and other enzymes are capable of breaking down synthetic polymers like polyethylene terephthalate into smaller and biodegradable using their enzymes.³⁹ Being mainly bacterial and fungal based enzymes, they show that enzymatic systems have the potential of reducing plastic and microplastic sources in the environment.⁴⁰

Emerging Enzymes and Future Perspectives

In recent developments in environmental biotechnology, novel enzymes have been identified having catalytic efficiency and extended substrate specificity in bioremediation as a starting point. Metagenomic, Meta transcriptomic, and proteomic systems have produced novel catalytic enzymes that have enlarged the list of biocatalysts that can degrade complex and previously persistent pollutants.⁴¹ These methods enable the uncultivable microbial communities to be explored which include enzymatic systems to extreme and polluted environments.

The use of protein engineering and directed evolution has also contributed to the creation of enzymes that are more stable, active, and have enhanced resistance to adverse environmental factors including extreme pH, temperature and salinity.⁴² A series of oxidoreductases, hydrolases and oxygenases that have been made through mutations and exhibit better catalytic activity have performed well at the pollutant degradation process, compared to their native forms. Such developments enhance greatly the practicability of the process of enzyme remediation in the field.

The other trend is the emergence of multi enzyme systems and enzyme consortia that mimic natural biodegradation products through a sequential and synergistic degradation of tough pollutants.⁴³ These systems increase the level of degradation and the level of reduction in the accumulation of intermediary toxic substances. Also, immobilization of enzymes on nanomaterials and biocompatible carriers has demonstrated potential in enhancing the reusability, stability of operations of enzymes as well as cost-effectiveness.⁴⁴

Plastic degrading enzymes are also among the most developing fields in the study of enzyme mediated bioremediation products. Ongoing finding and characterization of polyesterases and depolymerases has enhanced how fast plastics degrade, which provides a way forward in terms of plastic and microplastic pollution.⁴⁵

Even with these advancements, there are still challenges in the form of large scale production of enzymes,

economically viable and accepted by regulators. Future studies on enzyme-based systems push forward should concentrate on incorporating the existing remediation technologies with the enzyme-based system, offering more effective strategies of delivering the enzymes, and coming up with low-cost production platforms so as to enable wider applications of these enzyme-based systems to the environment.⁴⁶

Rationale and Objectives of the Review

The increasing nature and persistence of environmental contaminants and restrictions of traditional remediation technology have further intensified the requirement of effective and sustainable biological measures. Bioremediation using enzymes has been receiving more and more scientific and industrial interest because of its strong catalytic specificity, operational flexibility, and low ecological impacts, thereby making it a good substitute to environmental cleaning. Nevertheless, with great advancement, the current research on enzymatic remediation is mainly scattered among various types of pollutants and enzyme systems and their integrated use has been left unexplored.

The main aim of the review is to critically summarize and analyze the existing literature on enzyme-based bioremediation with specific reference on the mechanistic roles of microbial enzymes in degradation of various contaminants of the environment. This paper provides a systematic overview of several classes of enzymes commonly used in bioremediation such as oxidoreductases, peroxidases, hydrolases, oxygenases, dehalogenases, and plastic degrading enzymes with a focus on their catalytic mechanisms, and in certain cases, on the applications in pollutant removal.

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Conclusion

Bioremediation involving the use of enzymes has come out as an effective and viable approach to deal with complex types of environmental pollution. Contrary to traditional physicochemical methods such as remediation, enzymatic methods possess a high level of substrate specificity, catalysis, and disturbance of the ecological set is reduced and negligible compared to the other conventional methods

of remediation Microbial enzymes including oxidoreductases, peroxidases, hydrolases, oxygenases, dehalogenases, and synthetic polymer degrading enzymes have important roles in the degradation of industrial dyes, pesticides, petroleum hydrocarbons, halogenated compounds, to mention a few. The growing body of literature shows that enzymatic systems can reach a pollutant degradation efficiency of over 70-90 percent under favorable conditions, especially when enzyme immobilization and multi enzyme consortia are used Metagenomics, protein engineering and enzyme immobilization technologies have greatly contributed to the advancement of enzyme stability, activity and resistance to environmental stress, increasing their utility in real-life remediation processes It is worth mentioning that the production of plastic-degrading enzymes, including PETase, has provided fresh opportunities to reduce the plastic and microplastic pollution which is an increasing global environmental challenge. Although these have been achieved, there are still challenges with regard to large scale production of enzymes, cost of operation and deployment on a field scale. Research in the future must be done in creating cost effective platforms to produce enzymes, enhancing enzyme recycle, and adding enzymatic systems to the already available remediation systems. In sum, bioremediation by enzymes is an emerging flexible and promising tool to environmental management that can be used to make significant contributions to the global pollution mitigation process.

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Conflict of Interest

The author(s) declare no conflicts.

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