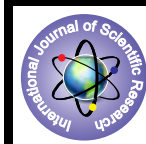


## Enzymatic Bioremediation: Global Scenario of Environmental Cleaning



### Environment

**KEYWORDS :** Enzyme bioremediation, Global scenario, Pollutant eradication.

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### ABSTRACT

*Pollution is the biggest menace to the living being on this planet today. The pollutant eradication through exploiting the enzyme potential by using the various techniques is being revolutionized in the present global scenario. Enzyme bioremediation is a breakthrough technology that has the potential to offer numerous benefits over conventional technologies. Enzyme bioremediation have greater positive effects and propose significant promise to pollutant bioremediation. In conclusion, the enzymatic bioremediation open the new era of pollutant eradication for clean, safe and green environment.*

### Introduction

The concept of using enzyme systems for enhancing bioremediation is relatively new and adopted globally. The effectiveness of enzymes in bioremediation is absolutely depends upon their efficiency and specificity. Enzymes are considered as eco-friendly catalysts and enzymatic techniques are considered as environmentally friendly processes (Gianfreda & Rao, 2008). Enzymes are stereo-specific with broad specificity and can be applied to a vast range of compounds. They can be used under extreme conditions with more mobility. All these characteristics render enzymes eco-friendly for bioremediation. Enzyme bio catalysis is referred as white biotechnology (Alcade et al. 2006) and work by green chemistry concept (Sheldon & van Rantwijk, 2004). "White biotechnology involves the use of biotechnology in the bulk production of fine chemicals such as amino acids, vitamins, antibiotics, enzymes, drugs, organic acids and polymers". It is appropriate to consider white biotechnology as green chemistry as it focuses on the development of clean bioprocesses that lead to reductions in greenhouse gas emissions, energy and water usage. The major enzyme groups engaged successfully in bioremediation are as follows-

#### 1. Mixed Function Oxidases (MFO)-

MFO also known as dependent cytochrome P-450 monooxygenases or P450 system which includes cytochrome P450 and NADPH-cytochrome P450 reductase. Cytochrome P450 enzymes have a broad substrate range and catalyses biochemically potent reactions such as the oxidation or hydroxylation of non-activated carbon atoms (Khaled et al. 2012). MFO is actively involved in the metabolism of both endogenous and exogenous substances and can metabolize organophosphates, carbamates, xenobiotics and pyrethroids.

#### 2. Oxygenases-

Oxygenase plays a key role in the metabolism of organic compounds by aromatic ring cleavage. Oxygenases are (I)

#### Monoxygenases-

The monooxygenases catalyzes oxidation of alkanes, steroids, fatty acids, aromatic and aliphatic compounds. Monooxygenase catalyzed the desulfurization, dehalogenation, denitrification, ammonification, hydroxylation, biotransformation and biodegradation of compounds. Methane monooxygenase is involved in the degradation of substituted methanes, alkanes, cycloalkanes, alkenes, haloalkenes, ethers and aromatic/heterocyclic hydrocarbons.

#### (II) Dioxygenases-

Different enantiomers of dioxygenases catalyze the oxygenation of a wide range of substrates and oxidize aromatic compounds into aliphatic products. Aromatic ring cleavage dioxygenases, and catechol dioxygenases degrade the chlorinated derivatives via meta and ortho pathways (Adams et al. 2006). These dioxygenase are dexteros in the breakdown of aromatic rings in a vari-

ety of pollutants such as catechols, protocatechuates, gentisates, salicylates, aromatic amino acids, 2-aminophenols, 3- hydroxy-anthranilates, hydroxyquinones and hydroquinones.

#### 3. Laccases-

Laccases (p-diphenol:dioxygen oxidoreductase) constitute a family of multicopper oxidases that catalyze the oxidation of phenolic, chlorinated biphenyls and aromatic substrates with concomitant reduction of molecular oxygen to water. Intra and extracellular laccases are capable of catalyzing the oxidation of ortho and para-diphenols, methoxyphenolic, aminophenols, polyphenols, polyamines, lignins, aryl diamines and inorganic ions (Couto et al. 2006). Laccases are also prominently involved in the depolymerization of lignin and degradation of compost, azo-dyes, phenols, anilines and aromatic thiols.

#### 4. Hydrolases-

Hydrolases catalyze the substrate via condensations and alcoholysis. Hydrolases are efficient for biodegradation of oil spills, organophosphate, carbamate insecticides, DDT and heptachlor. These enzymes also involves in the biotransformation of carbofuran, carbaryl, parathion, diazinon and coumaphos (Sutherland et al. 2004) into simpler products.

#### 5. Esterases-

Esterases includes carboxiesterases, amidases and phosphatases. Esterases degrade the insecticides mainly, organophosphates, carbamates, pyrethroids and xenobiotics via breakdown of carboxylic ester bond. The Carboxiesterases (type B esterases) are proficient for the hydrolysis of xenobiotics, amide, thioester, phosphate esters (parathion, paraoxon) and acid anhydrides (DIPFP).

#### 6. Lipases-

Lipases are ubiquitous enzymes which catalyze the hydrolysis of triacylglycerols to glycerol and free-fatty acids. Lipases degrade lipids derived and drastically reduce hydrocarbon from contaminated soil. Lipase activity is a useful indicator for testing hydrocarbon degradation in soil. These enzymes catalyze: hydrolysis, inter-esterification, esterification, alcoholysis and aminolysis. Lipase has many potential applications in food, chemical, detergent manufacturing, cosmetic and paper making industries.

#### 7. Cellulases-

Cellulases convert waste cellulosic material into foods to meet burgeoning population and have been the subject of intense research (Bennet et al. 2001). In paper and pulp industry, cellulases have been employed for recycling of paper. The cellulases are added during brewing to increase the juice liberation from fruit pulp and for the production of ethanol from cellulosic biomass.

#### 8. Oxidoreductases-

Oxidoreductases detoxify toxic xenobiotics, phenolic, and anilinic compounds through polymerization and copolymerization

(Park et al. 2006). The enzyme also reduces the radioactive metals from an oxidized soluble form to reduce insoluble form. The most promising application of oxidoreductase is the removal of endosulfan (1,2,3,5,6,7,7-hexachloro-5-norbornene-2,3-dimethanolcyclic sulfite), an organochlorine insecticide, which is highly toxic and endocrine disruptor.

### 9. Proteases-

Proteases such as serine endopeptidase, cysteine peptidase, aspartic endopeptidases and metallopeptidases has wide range of applications in food, leather, detergent, and pharmaceutical industry. The alkaline proteases are greatly used in leather industry for the removal of hairs.

### 10. Glutathione S-Transferase (GST)-

GST are involved in detoxification of endogenous and xenobiotic compounds (Khersonsky & Tawfik, 2010).

### 11. Phosphotriesterases (PTE)-

The most effective PTEs are: organophosphate hydrolase (OPH), methyl parathion hydrolase (MPH), organophosphorus acid anhydrolase (OPAA), di-isopropylfluorophosphatase (DFP) and paraoxonase 1 (PON1) (Bigley & Raushel, 2013). PTE are the most suitable pesticide degrading enzymes that hydrolyze and detoxify organophosphate pesticides (OPs).

### 12. Peroxidases-

Peroxidases catalyze the oxidation of lignin and other phenolic compounds at the expense of hydrogen peroxide. Cytochrome c peroxidase, ascorbate peroxidase (APX), catalase, lignin peroxidase (LiP), manganese peroxidase (Mnp) and horseradish peroxidases (HRP) are the members of this class. Three major classes of the peroxidase includes: (I) Lignin Peroxidases- Lignin peroxidase (LiP) plays a central role in the biodegradation of the plant cell wall constituent lignin and aromatic compounds. (II) Manganese Peroxidases-The  $Mn^{3+}$  chelate oxalate generated by MnP acts as a mediator for the oxidation of various phenolic compounds. (III) Versatile Peroxidases- VP enzymes directly oxidize  $Mn^{2+}$ , methoxybenzenes, phenolic and aromatic substrates. Class I peroxidases are biosynthetic enzymes involved in plant cell wall formation and lignifications.

### Properties of enzymes for bioremediation

The boost in enzymatic bioremediation can be exploited to engineer enzyme properties: active-site topology, enlarge binding pockets and alter the substrate specificity and stability. Consequently, the enzyme properties has been altered through modification in protein structure to make it more stable, more resistant to self destruction and make it target directed and function. Biological remediation can be rationalized by specific finite measurements of following enzyme properties: maximal enzymatic rate ( $V_{max}$ ), substrate specificity (Km), turnover number (kcat), enzyme efficiency (kcat/Km). Along with these acquaintances the pollutant eradication by enzymatic catalysis depends on the following properties-

1. Enzyme should target pollutant to substantially less toxic products.
2. Should not require cofactors, which would be prohibitively expensive.
3. Should have high  $K_m$  (substrate affinity) and Kcat (turn over) values with broad substrate specificity.
4. The enzyme must be robust to the wide range of temperature, pH and ionic environments.
5. The enzyme should fulfil all the required performance criteria in relation to stability, kinetics and cost of production for a bioremediation.

### Pros and cons of enzyme bioremediation

The benefit and limitation of enzyme bioremediation are as follows-

#### Pros

- I. Enzymatic bioremediation is a natural process and perceived as an acceptable waste treatment process.
- II. The enzymatic bioremediation does not generate toxic side-products.
- III. Following treatment, the enzymes are digested on-site by the indigenous microorganisms.
- IV. The enzymes significantly enhance their bio-availability efficiently with organic solvents.
- V. Enzymes are effective even in drastic operational conditions.
- VI. Enzymes are stereo-specific with narrow or broad specificity and can be applied to a vast range of compounds.

#### Cons

- I. Conversely, the enzymatic bioremediation is still expensive, having short lifetime and low stabilization.

### CONCLUSIONS

The development of knowledge about the enzymatic response to pollutant degradation expands the new horizon of cost effective and potentially significant advance environmental cleaning. Enzymes have great environmental advantages and are preferred over chemicals and microorganisms as they do not generate toxic side products. The main advantages of using enzymes in these scenarios are their favourable unique properties, basically their biodegradability and high chemo, regio and stereo-selectivity. This is opening exciting new panorama for enhancing bioremediation programs in the coming years. Future research should emphasize to evaluate the effectiveness of enzymes, the cost of enzyme production, culturing microorganism and develop engineering processes to design parameters and system for optimal enzyme kinetics. In conclusion, enzyme bioremediation has been implemented globally and opened the new era of pollutant eradication for clean, safe and green environment.

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