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Review

Study of Different Types of Enzymes for Biological, Agro-Food Processing and Industrial Wastewater Treatment: An Overview

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Abstract: The objective of the review study was carried out from different research data to find out the innovative latest information on the waste water treatment in agro, industrial and municipal areas. From the review study, it has been noted that cellulase, peroxidage, lignage, tyrosinase, pactinase and amylase have been widely used for waste water treatment. Besides, microbial enzymes such as microbial dioxygenase, oxygenase, laccase, peroxidase, lipase, cellulase, protease and fungal laccase have been applied for the waste water treatment in industrial and municipal areas and found effective results. Different techniques of application have been discussed using different hydrolytic and microbial enzymes. Therefore it seems that enzymatic treatments keep a significant role in waste water treatment from agro-food processing industries having low cost of production and treatment management process.

Keywords: Enzymes, Cellulase, Peroxidase, Ligninase, Waste Waster

Introduction

Biological means of waste water treatment in pulp and paper mills as well as other industries are a great and modern technology instead of chemical technology. Enzymes are used in many sources of waste water like agricultural, botanical, municipal, industrial and environmental (Hossain and Uddin, 2021).

Liu and Smith (2020) observed that enzymes were higher level industrial bio-catalysts having extensive applications in a wide range of manufacturing and processing sectors, including the agricultural, food and household care industries. The catalytic productivity of enzymes were higher as compared to the inorganic chemical catalysts receiving mild conditions. Besides, the nutrient medium was necessary for the biomass culture represented an important cost to industrial enzyme production. They also stated that Activated Sludge (AS) was a waste bio-product of biological wastewater treatment and was consisted of microbial biomass that was degraded organic matter manufacturing substantial quantities of hydrolytic enzymes. Liu and Smith (2020) exhibited that concentrated and purified AS enzymes were readily substitute inorganic and commercial bio-enzyme catalysts in many industrial utilization including leather processing and in detergent and animal feed formulation.

Robinson (2015) stated that enzymes were biocatalysts yielded by cells and were involved in most of the metabolic processes completed by living organisms. The maximum enzymes were globular proteins by comprising a amino acid conformation which might be bound to a non-protein coenzyme or metal ion cofactor. Enzymes promoted biochemical reactions by homogeneous mechanisms to inorganic chemical catalysts such as metals, metal oxides and metal ions, by taking molecules to overcome the energy limitation necessary having a reaction to proceed and increasing the correct oriented collision of molecules (Berg *et al*., 2002). Enzyme catalysis were involved by the substrate binding to the active site of the enzyme to build an enzyme-substrate complex as transitional state. As the reaction progresses, the enzyme was detached from the products without making consumed itself (Price and Stevens, 1999). Rupley *et al*. (1983) exhibited that commercial enzymes were extracted from the biomass of bacteria and different types of fungi, including yeast and then produced utilizing an industrial fermentation process. Industrial enzyme production was made by utilizing formulated culture media and to provide all the essential nutrients which important for microbial growth followed by further consolidation and purification (Rupley *et al*., 1983). Enzymes had been extracted by causing interrupted the biomass and were formed into marketable products by the addition of stabilizers (neutral salts like ammonium sulphate, glycerol or sorbitol) and mapped out to enable the protein conformation and standardizing components (salts or carbohydrates as for example, starch, maltodextrins and sugar alcohols) to mix the extracted enzymes to a standardized properties (Bisswanger, 2011). Particularly, synthetic culture media were represented as one of the most important costs for the commercial enzyme production, was equivalent to 30-40% of the total production cost (Hermes *et al*., 1987) and consequently, enzymes were produced by the standard industrial methods which were generally too expensive for the wide adoption in large-scale and ongoing industrial procedures (Li and Zong, 2010).

Enzymes Application in Pulp and Paper Waste Water

Previously, enzymatic hydrolysis techniques were not well developed in pulp and paper industries for the waste water treatment. Currently a lot of research activities have been done in the enzymology of lignin degradation. Hakulinen (1988) reported that ligninase, cellulase and peroxidase were the most significant enzymes utilized in pulp and paper industries for the waste water treatment. Among them, peroxidase was employed for the color abolition in the bleaching effluents. It had also been reported that combination of treatments like enzymes together with special microbes were possible to remove toxic and harmless compounds from wastewater (Hakulinen, 1988).

Enzymes Application in Food, Bakery and Agro Industry Waste Water

Different Enzymes

Zolghadri *et al*. (2019) exhibited that implementation of enzymes (tyrosinase) in the deletion of toxic organic compounds from the industrial wastewater which was similarly demonstrated by Klibanov *et al*. (1983) and coworkers (Malony *et al*., 1986) using enzymes of the peroxidase and polyphenol oxidase categories and compounds of the phenol and aniline types. The strengthen benefits of an enzyme-based treatment over traditional biological treatment included as application to a broad range of compounds, including those which were biorefractory, operation under wide temperature, pH and salinity ranges. It was reported that browning of fruits, fungi, vegetables and hyper-pigmentation in human skin were two common unexpected phenomena. Tyrosinase was the main enzyme identified as responsible for this enzymatic browning and melanogenesis in mammals (Dembitsky and Kilimnik, 2016, Maghsoudi *et al*., 2013). The encouraged researchers and scientists were to focus on the identification, isolation, synthesis and characterization of new potent tyrosinase enzyme for the various application in the food, pharmaceutical, cosmetics and medicinal industries.

It was reported that Mushroom tyrosinase had been employed to make antitoxic *waste water* or soil contaminated. Xu and Yang (2013) stated that a practical and less expensive immobilization technique had been developed for the mushroom tyrosinase to be used for the enzymatic treatment of phenolic wastewater. It had been catalyzed by the enzyme immobilized in the form of phenolic compounds as for example, phenol, p-cresol, p-chlorophenol and bisphenol. It could be efficiently eliminated using a complete conversion obtained within 0.5-3 h, superior to other processes catalyzed by the same enzyme. They also stated that the effects of reaction time, pH and enzyme concentration of the phenol solution were investigated. The sequence of dephenolization value was in accordance with the substrate suitability of the enzyme. The ability of reuse of the CLEA (Cross-linked tyrosinase aggregate) had been investigated in a batch reactor for each phenol. In a constant stirred tank reactor, the CLEAs encapsulated into calcium alginate gels were effective for removing phenol for 26h. The toxicity of the phenol-having solution was remarkably diminished after treatment with the tyrosinase CLEA as investigated by the Hydra Sinensis Test.

Zolghadri *et al*. (2019) stated that mashroom tyrosinases had been separated and refined from different sources like plants, animals and microorganisms. Although many of them, had been sequenced, except few of them had been characterized. Presently, a novel tyrosinase produced by actinobacteria had been isolated and attributed with the purpose of identifying novel enzymes having exclusive properties for the biotechnological implementation (da Silva *et al*., 2013, Dolashki *et al*., 2012). However, among different sources of tyrosinase, mushroom tyrosinase from *Agaricus bisporus* was a major and cheap resource of tyrosinase bearing high resemblance and equivalence as compared to the human tyrosinase (Vanitha and Soundhari, 2017). Due to these good properties the characteristics of mushroom tyrosinase had been investigated widely as a standard for investigating of tyrosinase inhibitor, enzyme-catalysed reaction, melanogenic and enzyme-inhibitor (Zekiri *et al* 2014). They also reported that tyrosinase from *Agaricus bisporus* was a 120 kDa tetramer having two different subunits like heavy and light which had three domains and two copper binding sites which bound to six histidine residues and interacted with molecular oxygen in the tyrosinase active site (Gasparetti, 2012).

Taylor *et al*. (1995) stated that several oxidative enzymes like peroxidases catalyzed the reaction of phenols. It had been exhibited that chloroperoxidase had been used to catalzye the removal of phenols from solution by a similar mechanism (Carmichael *et al*., 1985). Enzymes of the polyphenol oxidase type, represented using mushroom tyrosinase (Atlow *et al*., 1984) as well as the laccase type, from various soil microorganisms (Bollag, 1992). Both of them used molecular oxygen rather than peroxide as an oxidant had been explored for the similar technique. It had been studied with the Horseradish enzyme (HRP) by maintaining pH, temperature, incubation period, hydrogen peroxide and the enzyme and reported that different optimal pH and catalytic lifetimes of the enzyme were exhibited for the various phenols (Nicell *et al*., 1992).

Amylolytic Enzymes (Amylase)

Priya and Renu (2018) observed that amylolytic enzymes were a beneficial in bakery, food, automation dishwashing, fodder, wastewater treatment and poultry. They also stated the experiment by utilizing jack fruit seed as a cheap substrate for the manufacturing of amylase in solid substrate fermentation utilizing *Penicillium sp* SP2 and to treat wastewater. They also investigated that stagnant water was implemented to isolate Penicillium sp. SP2 for amylase production. The fungal isolate, *Penicillium sp.* SP2 was cultured in solid substrate fermentation employing jack fruit seed as a less expensive substrate. The process parameters were optimized to develop the production of enzyme. Amylase was fractionated using ammonium sulphate and purified utilizing sephadex G-75 gel filtration column. The crude amylase was implemented to treat wastewater. The isolated Penicillium sp. SP2 used jack fruit seed substrate and manufactured amylase. Among the carbon sources, glucose outstandingly improved the production of enzyme. Moreover, amylase production was found to be higher at 35°C. Considering as cheap cost jack fruit seed was a novel substrate for amylase production for various biotechnological implementations. The amylase using *Penicillium sp.* SP2 was found great implementation in wastewater treatment.

Pectinase Enzyme

Thakur and Kumar (2019) stated that rising water pollution was a major cause of diseases and other health related problems. Treatment of waste water generated through various industries such as paper processing industries, food processing industries and agro-based industries was a challenge since it released various plant polysaccharides such as starch and pectin. Waste water treatment could be done through chemical, physical and enzymatic methods. Chemical methods utilized for the treatment waste water were harsh on the environment and caused direct or indirect environmental pollution. Alternatively, enzymes such as pectinase was utilized for the pre-treatment of pectin rich wastewater generated through above mentioned industries before releasing the water to streams, rivers or sending the pre-treated water for secondary treatments.

Cellulase Enzyme

Libardi *et al*. (2020) stated that cellulose was gained from raw sanitary wastewater utilizing a fine-mesh sieve (0.35 mm) and quantified by enzymatic hydrolysis and thermo-gravimetric analysis. The manufacturing and formulation of cellulase enzyme which resulted in an enzymatic blend of endoglucanases cellobiohydrolases as well as beta-glucosidases. The content of the recaptured cellulosic material was 21.3% according to the enzymatic hydrolysis and 27.7% for thermo-gravimetric. The enzymatic hydrolysis of the WWTP residue employing the produced cellulase exhibited better results than the commercial cellulase complex used (Fig. 1).

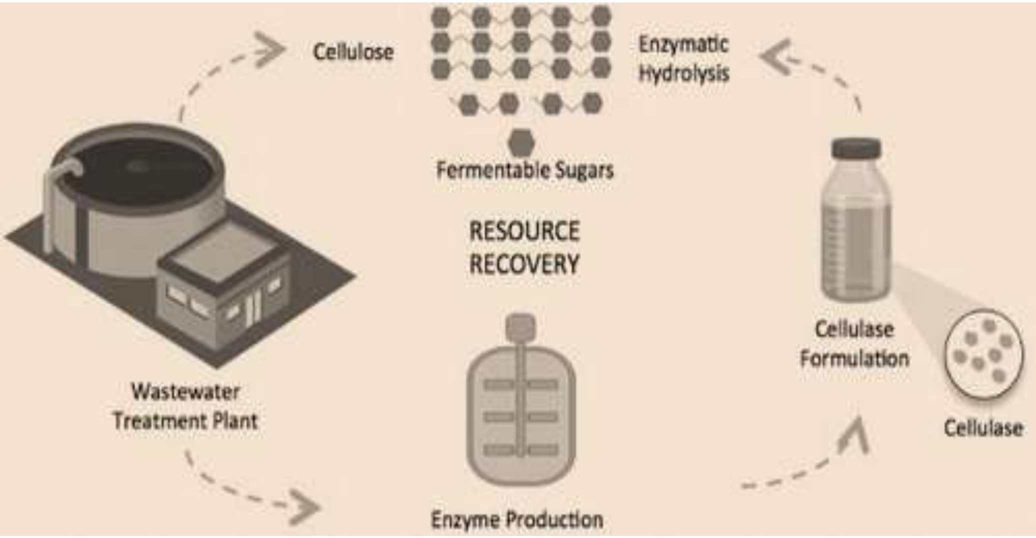

Fig. 1: Diagram shows the utilization of cellulase for the waste water treatment (Libardi *et al*., 2020)

Types of Microbial Enzymes

Microbial Oxidoreductases

Detoxification of toxic organic compounds utilizing various bacteria and fungi (Rani *et al*., 2019) and higher plants coming after oxidative coupling was moderated with oxidoreductase.

Microbial Oxygenase

It had been reported that oxygenase was under the oxidoreductase group of enzymes and participated in oxidation of reduced substrates transferring oxygen from molecular oxygen (O_2) employing FAD/NADH/NADPH as a co-substrate. These enzymes had a significant character in the metabolism of organic compounds as they increased their reactivity or solubility in water or brought about cleavage of the aromatic rings (Pandey *et al*., 2017). They also reported that oxygenases also mediated dehalogenation reaction of halogenated methane, ethane and ethylene in connection with multifunctional enzymes monooxygenases. These enzymes catalyzed oxidative reactions of substrates from alkane to complex molecule as steroid and fatty acid as well as required molecular oxygen for their function. These enzymes required only molecular oxygen for their activity and used the substrate as reducing agent (Pandey *et al*., 2017). Various aromatic and aliphatic compounds were catalyzed by monooxygenases such as desulfurization, dehalogenation, biotransformation, ammonification, denitrification, hydroxylation and biodegradation.

Microbial Dioxygenase

It had been exhibited that dioxygenases catalyzed the enantiosis specifically the oxygenation of extensive range of substrates (Rani *et al*., 2019). They also reported that aromatic compounds were primarily oxidized by using dioxygenases, reflecting the implementation of dioxygenases in the environmental reformation. Moreover, the dioxygenases were found in the soil bacteria and engaged in the transformation of aromatic precursor into the aliphatic product (Rani *et al*., 2019).

Microbial Laccase

Laccases (p-diphenol: Dioxygen oxidoreductase) belongs to the family of multicopper oxidases produced by certain plant, fungi, insect and bacteria that catalyzed the oxidation of an extensive range of decreased phenolic and aromatic substrates having concomitant reduction of molecular oxygen to the water. However, many microorganisms produced intra and extracellular laccases which had ability of catalyzing the oxidation of orthodiphenol, paradiphenol, aminophenol, polyphenol, polyamine, lignin, aryl diamine and some inorganic ions (Pandey *et al*., 2017).

Microbial Peroxidase

It had been stated that these enzymes catalyzed the oxidation of lignin and other phenolic compounds at the cost of hydrogen peroxide (H2O2) with a mediator (Siil *et al*., 2009). They also reported that lignin peroxidase (LiP) and manganese-dependant peroxidase (MnP) had been studied because of their high potentiality to degrade toxic substances in the environment, (Siil *et al*., 2009).

Microbial Lipase

It had been observed that these enzymes catalyzed various reactions as for example, hydrolysis, aminolysis, alcoholysis, inter-esterification and esterification (Rani *et al*., 2019). In accordance with its diagnostic utilization in bioremediation, lipase had many potential implementations in food, manufacturing, cosmetic, chemical, paper making industries and detergent industries but its manufacturing expenses had limited as its industrial applications (Siil *et al* 2009).

Microbial Cellulase

In the course of the enzymatic hydrolysis, cellulose was degraded by the cellulase enzyme to the reducing sugars that might be fermented by the utilization of yeasts or bacteria to the ethanol (Pandey *et al*., 2017). Besides, cellulase caused the deletion of cellulose microfibrils which were built at the time of washing and the utilization of cotton-based cloths. However, in paper and pulp industry, cellulase was used for the deletion of ink during recycling of paper (Pandey *et al*., 2017).

Microbial Protease

Proteases belong to the group of enzymes that hydrolyzed the peptide bonds in aqueous condition and synthesized them in non-aqueous conditional environment. It had been reported that proteases had wide range of implementationin food, leather, detergent and pharmaceutical industry (Rani *et al*., 2019). They also stated that industries and pollutants aromatic compounds, with phenols and aromatic amines, incorporated one of the major types of pollutants and were firmly conducted in many countries. Moreover, they were found in the waste water of a extensive variety of industries bearing resins and plastics, petroleum refining, dressing and pulp and paper, coal conversion, textile, dye and other chemicals, wood preservation, mining and metal coating (Pandey *et al*., 2017).

Kraft Process using Peroxidase and Laccase

It had been exhibited that the Kraft process which was extensively utilized in wood pulping leaves 5-8% (w/w) of the residual modified lignin in the pulp processing industry (Pandey *et al*., 2017). These residues were accountable for the attributes of brown color of the pulp and was commercially deleted by the utilization of bleaching agents as for example, chlorine and chlorine oxides. They also reported that bleaching operations turned out to the dark brown colored pollutant which contained toxic and mutagenic chlorinated products that caused an environmental threat (Pandey *et al*., 2017). Pandey *et al*. (2017) observed that the utilization of peroxidases and laccases was effective for the treatment of bleaching pollutant. They also stated that the configuration might alter having the harsh physical and chemical conditions of temperature, pH and ionic strength. However, this drastic conditions were often encountered in pollutant streams. Pandey *et al*. (2017) exhibited that the immobilization technology reduced the loss of enzymes by increasing their recycling ability and also minimized the chances of loss of enzyme activity under the harsh environmental condition. They also observed that the utilization of immobilized enzymes in waste treatment as compared to the free enzymes by resulting in many benefits like increased stability, recycling ability, easy handling, reduction in constant expenses. Moreover, horseradish peroxidase covalently immobilized on the magnetic pellet which preserved a high functionality and stability and exhibited higher phenol conversion.

Fungal Laccase

It had been reported that γ -aluminium oxide pellet based immobilization having had been shown to decolorize solutions of azo dyes (Pandey *et al*., 2017). Many researchers stated that an enzyme into the effluent directly was one of the simplest method of enzyme controlling to cause pollutant in the introduction of cells or tissues production. This method was implemented when suitably adapted strains of microorganisms were used to co-metabolize targeted pollutants. They also reported that cultures of the *Staphylococcus arlettae* were shown to decolorize solutions of four azo dyes in a microaerophilic sequential process and the average de-colorization obtained was of 97%. Moreover, in the cases of where the effluents were treated which contained pollutants that were not supported the growth, cell free or isolated enzymes were preferred for the use of all intact microorganism.

Conclusion

From the review study, it can be seen that cellulase, peroxidage, lignage, tyrosinase, pactinase and amylase have been found widely used nowadays for waste water treatment instead of chemical treatments. In addition to that microbial enzymes such as microbial dioxygenase, oxygenase, laccase, peroxidase, lipase, protease and fungal laccase have been utilized for the waste water treatment in industrial and municipal areas and found effective results using different technologies. Therefore, the enzymatic treatments have a significant role in waste water treatment from agro-food processing industries having low cost of production with management process.

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Author’s Contributions

ABM Sharif Hossain: Supervision the review data collection work, partially review writing and editing, final draft checking.

Musamma M Uddin: Literature review collection, Writing - original draft.

Ethics

The authors confirm that this article has no ethical problem.

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