



The Biocatalytic potential of Laccase enzyme

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ABSTRACT

Laccases have gained much attention of researches in the last decades due to their ability of oxidizing both phenolic and non-phenolic lignin related compounds as well as highly recalcitrant environmental pollutants. Laccases is commercially used to produce ethanol and also for the delignification of woody tissues. . Laccases are also used as catalysts for the manufacture of anti-cancer drugs and even as ingredients in cosmetics. Recently, the utility of laccases has also been applied to nanobiotechnology.

This paper reviews recent and important research paper related to the properties, heterologous production, molecular cloning, and applications of laccases within different industrial fields as well as their potential extension to the nanobiotechnology area. In this report we focused on the occurrence of laccase and its functions in physiological development and industrial utility. Laccase participates in cross-linking of monomers, degradation of polymers, and ring cleavage of aromatic compounds. It is also used in the synthesis of organic substance, where typical substrates are amines and phenols, the reaction products are dimers and oligomers derived from the coupling of reactive radical intermediates. In the recent years, these enzymes have gained application in the field of textile, pulp and paper, and food industry. Recently, it is also used in the design of biosensors, biofuel cells, as a medical diagnostics tool and bioremediation agent to clean up herbicides, pesticides and certain explosives in soil. \

Keywords: Laccases, biocatalyst, industrial applications

I. INTRODUCTION

Laccase was studied by Gabriel Bertrand in 1894 in the sap of Chinese lacquer tree. Laccases are the multicopper enzyme in higher plants and fungi. These are copper containing 1,4-benzenediol: oxygen oxidoreductases found in higher plants and microorganisms. These are glycosylated polyphenol oxidases that contain 4 copper ions per molecule that carry out 1 electron oxidation of phenolic and its related compound and reduce oxygen to water. When substrate is oxidized by a laccase, it loses a single electron and usually forms a free radical which may undergo further oxidation or nonenzymatic reactions including hydration, disproportionation, and polymerization . These enzymes are polymeric and generally contain 1 each of type 1, type 2, and type 3 copper centre/subunit where the type 2 and type 3 are close together forming a trinuclear copper cluster. In plants laccases are found in cabbages, turnip potatoes ,pears , apples and some other vegetables. They are also present in Ascomycetes, Deuteromycetes and basidiomycetes and. some bacteria such as *Theiophora terrestris*, *Phanerochaete chrysosporium*. These enzyme require oxygen as a second substrate for their enzymatic activity.



Recently laccases have been efficiently applied to nanobiotechnology due to their ability to catalyze electron transfer reactions without additional cofactor. The technique for the immobilization of biomolecule such as layer-by-layer, micropatterning, and self-assembled monolayer technique can be used for preserving the enzymatic activity of laccases.

II. POTENTIAL OF LACCASES

A few laccases are at present in market for textile, food and other industries, and more candidates are being actively developed for future commercialization. A vast amount of industrial applications for laccases have been proposed and they include pulp and paper, textile, organic synthesis, environmental, food, pharmaceuticals and nanobiotechnology. Laccases are highly specific, energy-saving, and biodegradable therefore laccase-based biocatalysts fit well with the development of highly efficient, sustainable, and ecofriendly industries.

2.1 Pulp and paper industry

In wood pulp the separation and degradation of lignin conventionally obtained using chemical oxidants. Conventionally based chlorine – based method of removing lignin from pulp has been removed from the use of laccase enzyme [1].The pretreatment of delignification of wood pulp with the help of laccases can give us milder and cleaner strategies [2].The physical, chemical as well as mechanical properties of pulp can be improved by laccases either by forming reactive radicals by lignin or by functionalizing lignocellulosic fibres. It can be used to delignify the pulp with the help of mediators [3]. Mediators are the compounds with high redox potential having low molecular weight and may be used to oxidize the non – phenolic residues from the oxygen delignification. The cross – linking and functionalizing ligninaceous compounds has been discovered [4]. Both fungal and bacterial laccases help in improving pulp properties and bioremediation of pulp and paper mill effluents.

2.2 Textile industry

Laccase is used to improve the whiteness in conventional bleaching of cotton . its potential benefits . Its potential benefits include chemicals, energy, and water saving. Indigo –dyed denim fabrics can be shaded to lighter shades by the use of laccases [5]. Dye precursors can be converted by the use of laccases for the better and more efficient fabric dyeing . Its potential use can be found in cloth and dish washing . The use of laccase can be found in removing the odor on fabrics including cloth, sofa surface and curtain. It is also added in the detergent to remove the odor that is generated during the cloth washing . Its use has been found to resist the wool shrinkage [6]. This seems to be the most amazing use of laccase enzyme. The process of chlorination has been conventionally used to resist the wool shrinkage. Previously proteinase treatment was commonly used for the anti shrinking of wool but now it has been replaced by the laccase enzyme due to its high specificity and much lower environmental impact. Moreover, proteinase leads to deterioration of fibre strength and limited anti shrinking property, and protein degradation. *T. versicolour* was used in the form of pellets by Blanquez to treat a black liquors discharge for detoxifying and reducing the colour, aromatic compounds, and chemical oxygen demand (COD). Romero found that bacteria *S. maltophilia* decolorizes some synthetic dyes (methylene blue, methyl green, toluidine blue, Congo red, methyl orange, and pink) as well as the industrial effluent.

2.3 Food industry

The components of food such as carbohydrates, unsaturated fatty acids, phenols and thiol-containing proteins act as substrates for the laccase enzymes. The functionality, quality or cost reduction can be done by the modification of these substrates by the laccase enzyme . Oxygen leads to the degradation of food and laccase enzyme can be used as oxygen scavengers to remove oxygen and our food will be saved . The dissolved oxygen in the vegetable oil can be removed by the use of laccase enzyme and due to this the flavor quality of the vegetable oil is increased . Oxidation food items derived partly or entirely from extracts of plant materials can be done by the use of laccase enzyme. Cacao was soaked in solutions containing laccase, dried and roasted in order to improve the flavor and taste of cacao and its product . It is used to enhance the colour of the tea based products [7]. Gels can be formed of food ingredients by the cross linking of sugar beet pectin and ferulic acid by oxidative coupling.[8]. Fruit juice stabilization can be done with the help of various enzymatic treatments, among which laccase can be used . Wine stabilization is one of the main applications of laccase in the food industry and is alternative to physical-chemical adsorbents [9].

Recently, a few bacterial laccases have been isolated from *Escherichia coli*, *Bacillus halodurans*, *Thermus thermophilus*, and several species of *Streptomyces*. Bacterial laccases are believed to play a role in melanin production, spore coat resistance, morphogenesis, and detoxification of copper [10]. The bacterial laccase from *Bacillus subtilis* was found to be an endospore coat protein with high thermostability [11]. Utilizing bacterial laccases for industrial production would allow for new biotechnological applications due to the ease of genetic improvements to expression level, activity, and selectivity [12].

Formation of Haze, oxygen content and temperature affects the storage life of beer. Laccase could be added for the removal of the unwanted oxygen in the finished beer, and thereby the storage life of beer is enhanced. Also, a commercialised laccase preparation named “Flavourstar

2.4 Bioremediation

Laccase also used for the bioremediation . Undesirable contaminants, byproducts or discarded material can be biodegraded with the help of laccases. Plastic waste having olefins can be degraded with the help of laccases . Laccase helps to initiate the radical chain reaction which leads to the breakdown of the plastic. Polyurathanes also degraded by laccases . It also facilitates the degradation of phenolic compounds from biphenol and alkylphenol derivatives and also the decomposition of fluorescent brighteners . Laccase may also be used to eliminate odor emitted from places such as garbage disposal sites, livestock farms, or pulp mills [13]. The effluents generated from house can be decolorized with the help of laccase enzyme that are hardly decolorized by conventional sewage treatment plants [14]. Laccases can decolorize waste present in waters from olive oil mills [15] and pulp mills . It also decreased the concentration of synthetic heterocyclic compound such as halogenated organic pesticides in the soil [16]. In this sense, Laccases are being included in several enzymatic bioremediation programs . Another potential application is the bioremediation of contaminated soils, toxic organic pollutants, such as various xenobiotics, PAHs, chlorophenols, and other contaminants [17].

2.5 Organic synthesis

Laccases also used as a biocatalyst in the organic synthesis [18]. Laccase-catalyzed cross-linking reaction of new urushiol analogues for the preparation of “artificial urushi” polymeric films (Japanese traditional coating) was demonstrated . It is also mentioned that laccase induced radical polymerization of acrylamide with or



without mediator [19]. Polymerization of various amino acids and phenolic compounds also done by laccase . Recently, to improve the production of fuel ethanol from renewable raw materials, laccase from *T. versicolor* was expressed in *S. cerevisiae* to increase its resistance to (phenolic) fermentation inhibitors in lignocellulose hydrolyzates [20].

2.6 Pharmaceutical sector

Laccases are also used for the production of antimicrobial detoxifying or active personal care agents. Due to their specificity and bio-based nature, potential applications of laccases in the field are attracting active research efforts. Anesthetics, anti-inflammatory, antibiotics sedatives can be produced with the help of laccase enzyme. Used in the synthesis of complex medical compounds as anesthetics, anti-inflammatory, antibiotics, sedatives, etc. , including vinblastine, mitomycin, penicillin X dimer, cephalosporins, and dimerized vindoline [21]. Laccase-based *in situ* generation of iodine, a reagent widely used as disinfectant . Also, laccase has been reported to possess significant HIV-1 reverse transcriptase inhibitory activity . A novel application field for laccases is in cosmetics. For example, laccase based hair dyes could be less irritant and easier to handle than current hair dyes. More recently, cosmetic and dermatological preparations containing proteins for skin lightening have also been developed. Laccases may find use as deodorants for personal-hygiene products, including toothpaste, mouthwash, detergent, soap, and diapers.

2.7 Laccase function in insects

Laccase has been found in the cuticles of many insect species. The laccase from insects is a long amino-terminal sequence characterize by a unique domain consisting of several conserved cysteine, aromatic and charged residues Laccase-1 was found to be present in the midgut, malpighian tubules and fat body as well as epidermis of the tobacco horn worm, *manduca sexta*, and may oxidize toxic compounds ingested by the insect. Laccase-2 was involved in cuticle tanning of the red flour beetle, *tribolium castaneum*. Laccase is also present in the salivary glands of *N. cincticeps* which is secreted in watery saliva , by using biochemical and histochemical approaches

III. CONCLUSION

Laccase are versatile enzyme having a great biocatalytic potential in several areas. They are multicopper enzymes which are widely distributed in higher plants, animals and fungi. They are capable of degrading lignin and widely present. They help in treatment of waste water by decolorizing and detoxifying the industrial effluents. They are also used in paper and pulp industry, textile industry, bioremediation and act as biosensor. However, one of the limitation for large scale applications of laccase is the lack of capacity to produce large volumes of highly active enzyme at affordable cost. Laccase has applied to various biotechnological applications. That's why laccase is receiving much attention of researchers around the globe.

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