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Actinomycetes as Tools for Biotransformations of Lignans

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Opinion

The current demands for novel and sustainable biotechnological processes, including new microbial enzymes with industrial potential are constantly required. As a form to address the growing need for industrially relevant enzymes, functional screenings of microorganisms and/or (meta)genome mining techniques have emerged as powerful strategies for the identification of promising enzymes to novel or improved industrial processes [1]. Once enzymes have been well characterized, they can be produced, studied, and engineered about their biocatalytic, including possible synergistic activities in multiple protein cocktails. Currently, there is an increased interest in exploring and exploiting microbial enzymes for selective degradation of plant biomass. The efforts are concentrated on the bioconversion of lingo-cellulosic material. While ample enzymes are nowadays available for the degradation and modification of the polysaccharide content of plant biomass, there is a need for effective lignin degrading enzymes.

Focus on novel ligninolytic enzymes from microbial lignin-degrading systems can be valuable biocatalytic tools for the valorisation lignocellulosic material, as energetic substrates for bio-products obtaining. Biocatalysis refers to the use of whole cells or enzymes to catalyze reactions or transformations can promote the generation of numerous human needs [2].

One of the most recently microbial groups explored in this area is actinobacteria, known about their special abilities to produce diverse bioactive compounds, including enzymes with multiple biotechnological applications. Due to its ability to produce secondary metabolites with widespread industrial applications, *actinomycetes* have attracted the attention of many research groups in the world [3].

Adapted to many environmental conditions, actinobacteria are particularly promising, since this group is a potential producer of antimicrobial compounds, enzyme inhibitors, immuno-modifiers, enzymes and growth promoting substances for plants and animals [4]. Additionally these bacteria can also participate in the degradation processes of recalcitrant organic matter, contributing to the ecological balance do carbon in the planet.

Lignin, as molecular compound, comes from the oxidative polymerization of hydroxycinnamic alcohol derivatives [5]. The term lignocellulose is related to the bound of cellulosic material and the phenolic polymer by ferrul oil bonds [6]. Davin et al. (2008) [7] conducted extensive reviews of various aspects of lignin and lignan formation, including a detailed biochemical evaluation of reactions by random or controlled coupling of 4-hydroxyphenyl (H), guaiacyl (G) and siringuil (S), as well monolignol derivatives. In this sense, due to the economic value added in the production of wood and biofuels, lignin biosynthesis, as well as its manipulation, has been described as the target of some research works [8]. Enzymes that lead to the formation of monomers with different compositions, resulting in different proportions of guaiacyl and siringuil units, involving gene and mutation studies [9-11]. In addition, informations about the enzymatic reactions involved in the formation of lignans from coniferous alcohols is still limited [5].

In this context, lignocellulolytic enzymes from actinobacteria are one of the most explored for their application in industries that use lingo-cellulose as raw material. In addition to cellulose, lignin is also considered as one of the most abundant polymers at Earth. Lignin, suber in and condensed tannins are polymers composed by phenyl-propanoids that can contribute significantly to the stability and robustness of gymnosperms and angiosperms, from mechanical or environmental damage, such as pruning or drying [7].

While in the past, it was thought that only (white-rot) fungi were responsible for the degradation of lignin, it is becoming clear that also bacteria can play an important role in lignin degradation [12]. The main bacterial enzyme actors in lignin degradation seem to be laccases and a recently identified family of heme-containing peroxidases known as dye-decolorizing peroxidases (DyPs, EC 1.11.1.19) [13,14]. These enzymes form a distinct group of heme-containing peroxidases and seem to offer attractive catalytic

properties for biotechnological purposes, including ligninolytic abilities [15]. Except for these bacterial peroxidases, laccases also can be potent lignin-modifying enzymes. These oxidoreductases act on various polyphenols which form the core of lignocellulosic material [16,17]. Laccases are classified as multicopper oxidases, with low substrate specificity, which allows their activity under a large scope of organic compounds. Laccases typically contain four copper atoms to support catalysis. Laccases from *actinomycetes* were also already described, including *Streptomyces griseus, S. cyaneus, S. coelicolor, S. ipomoea, S. Sviceus and Thermobifida fusca*. These proteins were found to represent so-called small laccases, containing two domains linked to copper atoms [18,19]. Genes from diverse lignocellulolytic actinomycete strains have been described, cloned and expressed in *Escherichia coli* [20-22]. Saini et al. (2015) [23] reported about a thermo-alkali stable laccase from *Thermobifida fusca*, which could promote oxidation of 2,6-dimethyl phenylalanine and p-aminophenol [24]. While some reports show that several bacterial peroxidases and laccases have been reported in literature, the number of available and well-characterized laccases and Dyp-type peroxidases is very limited.

In this sense, studies about lignans and their pharmacological properties have been gaining increased relevance, involving investigations on their cytostatic, antitumor, as well as antiparasitary activities. In this way, enzymatically directed reactions, promoting improvement of regal and diastereo-selectivity, for the desired products, have become elegant tools and extremely relevant in the study of lignans. In order to evaluate the effect of the microorganisms on the biotransformation of lignans, some studies have been conducted [5,25-28].

All of these allied to emerging issues, non-profit, over-the-counter potential efficient medicines, treatment of neglected diseases, sustainable production of fuels, making the use of viable technologies, low cost and reuse of plant biomass. In this way, more investigations are needed in order to converge with the evaluation of the same substrates to the microbial metabolism, like some phenylpropanoids isolated from *Necthandra neucantha*, belonging to the family *Lauraceae* [29]. Such a multidisciplinary characteristic of a project that is being conducted by our research group, we are aiming the pharmacological evaluation of the biotransformation of phenylpropanoids with historical parasitic activities, by actinobacteria isolated from distinct Brazilian environmental habitats. Conventional methods using plants to produce compounds are still considered as effective, but they imply in low concentration of the desired compounds and high dependence on agricultural productivity, which involves classical risk factors, including climatic conditions and plant pathogens [30].

Previous results from our group and partnerships, demonstrated that some *actinomycetes* can be able to promote the bioconversion of lignin compounds, motivating deep investigations about the enzymatic systems that are involved, as well as the characterization of reaction products. In terms of bioenergy view, the conducted study is innovating in the use of non described compounds (lignans) [29], as model substrate to evaluate ligninolytic abilities of microbial strains. At the clinic view, it is possible to verify new possibilities to biotransformed phenylpropanoids with improved activities.

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