Title: IN SILICO EVALUATION OF THE MARINE-DERIVED FUNGUS *Peniophora* sp. CBMAI 1063 TRANSCRIPTOME: LACCASE GENES

Authors: 1Otero, I.V.R.; 1Ferro, M.; 1Bacci, M.; 1Ferreira, H.; 1Sette, L.D.

Institution: 1Biosciences Institute, São Paulo State University - UNESP, Rio Claro-SP, Brazil.

Abstract:

Laccases (oxy-reductase, EC 1.10.3.2) belong to the group of ligninolytic enzymes and are able to catalyze reactions involving phenolic compounds and aliphatic and aromatic amines. The potential of these enzymes has been exploited in the treatment of industrial effluents (bioremediation), clarification of beverages, pulp bleaching, among others industrial sectors. Studies with the marine-derived fungus *Peniophora* sp. CBMAI 1063 demonstrated the ability of this organism to produce large amount of laccases under saline conditions in flask (50 mL) and in bioreactor (3.5 L) scales. The culture conditions for laccases production by this fungus were previously optimized and patented (INPI BR 10 2014 008502). These enzymes showed ability to degrade textile dyes and showed resistance to different ions, pH and temperatures. In this context, the present study aimed to identify laccase genes from the analysis of the transcriptome of the fungus *Peniophora* sp. CBMAI 1063. RNA was extracted using RNeasy Plant Mini Kit QIAGEN after cultivating the fungus under the optimized conditions for laccases production. Samples were sent to MACROGEN for paired-end sequencing on Illumina platform HiSeq2000. The reads obtained underwent a quality control using FastQC program and the trimming bases with less Phred 20 was taken by Trimmomatic program. The transcripts were mounted on the Trinity program and the annotation of the transcripts was held at Blast2GO. A total of 51 isoforms of the laccase gene were obtained, and among them, 10 presented complete ORFs. After manual curing of selected genes by observing the conserved domains (NCBI - Conserved Domain Database), 4 isoforms showed complete domains and multi-domains and similarity with other fungal laccases (*Trametes* sp., *Pleurotus ostreatus*, *Steccherinum murashkinskyi*, *Coprinopsis cinerea*). These 4 isoforms encoding between 58 and 61 Kda protein and all sequences have high GC content (above 55% -Gene Runner). The analysis of the transcriptome from the marine-derived fungus *Peniophora* sp. CBMAI 1063 was efficient, allowing the detection of laccase genes expressed by this organism. One of the isoforms was already cloned and will be submitted to sequencing in order to get heterologous expression in two different systems (eukaryote and prokaryote). Data from the present work encourage further studies aiming the reduction of time and costs in the laccases production process by the marine-derived fungus *Peniophora* sp. CBMAI 1063.

Key-words: Ligninolytic enzyme, Transcriptome Analysis, Bioinformatics, Marine-derived fungus

Support: FAPESP (project 2013/19486-0) and CNPq (IO scholarship 159488/2014 and LDS 304103/2013-6)