

Novel laccase-like multicopper oxidases from the *Myrothecium roridum* fungus – production enhancement, identification and application in the dye removal process

Anna Jasińska[✉], Aleksandra Góralczyk, Adrian Soboń and Jerzy Długoński

Department of Industrial Microbiology and Biotechnology, Faculty of Biology and Environmental Protection, University of Lodz, Łódź, Poland

The aim of this study was to overproduce, identify and apply novel laccase-like multicopper oxidases (LMCOs) from *Myrothecium roridum* in a dye removal process. LMCOs' production was enhanced by modifying the medium and adding copper ions. After purification, two proteins, LMCO1 and LMCO2, with molecular masses of 46.7 and 66.3 kDa were discovered. Peptide analysis by mass spectrometry revealed that they belong to the cupredoxin superfamily. Characteristic peptide sequences were obtained for MCOs and bilirubin oxidases. Crude enzymes were applied in a dye decolorization process. Supplementation with 1 mM of vanillin allowed an almost complete elimination of the Indigo carmine within 3 hours. The dye was removed from a solution containing metals, surfactants and organic solvents. The in-gel assessment of the activity and decolorization ability of MCOs, followed by protein extraction and SDS-PAGE, confirmed that only LMCO2 was responsible for the dye removal. MCOs produced by *Myrothecium* sp. have been poorly studied before. The obtained results broaden knowledge on this subject and may contribute to the development of an eco-friendly method of dye elimination.

Key words: multicopper oxidases, laccase, bilirubin oxidase, *Myrothecium roridum*, decolorization

Received: 19 December, 2017; revised: 13 March, 2018; accepted: 19 March 2018; available on-line: 25 April, 2018

[✉]e-mail: anna.jasinska@biol.uni.lodz.pl

Abbreviations: MCOs, multicopper oxidase; LMCOs, laccase-like multicopper oxidases; ABTS, 2,2-azinobis-3-ethylbenzothiazolin-6-sulfonic acid; DMP, 2,6-dimethoxyphenol; TEMPO, 2,2,6,6-tetramethylpiperidine 1-oxyl

Supporting information

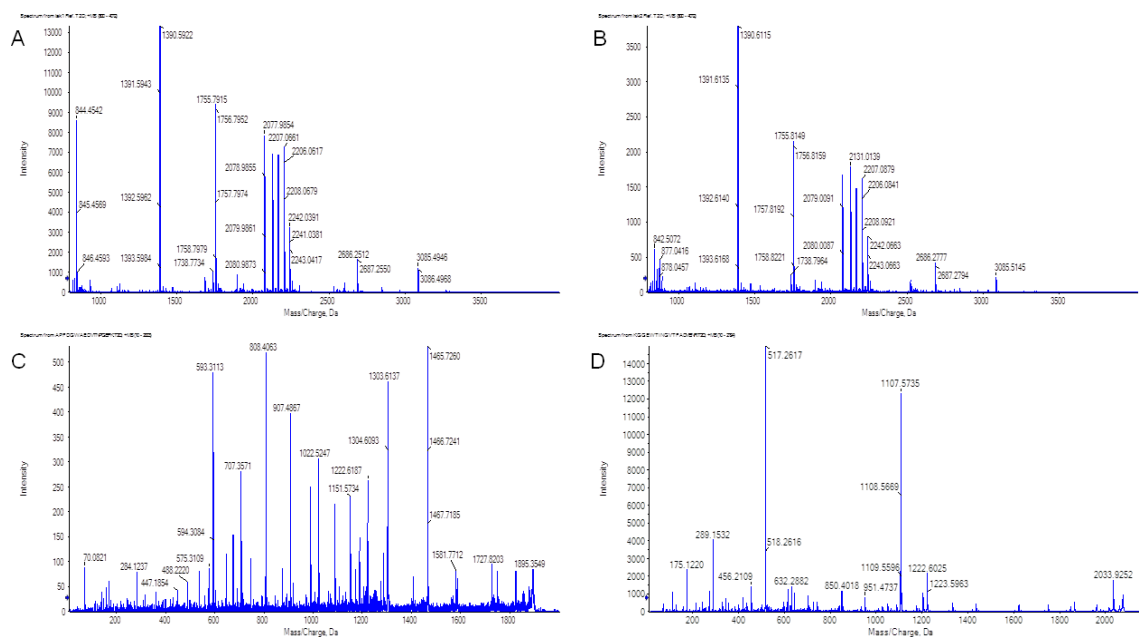


Figure S1. The mass spectra and tandem mass spectra of LMCO1 and LMCO2. (A) TOF MS spectrum of LMCO1. (B) TOF MS spectrum of LMCO2. (C) TOF MS/MS precursor 1895.82 (D) TOF MS/MS precursor 2076.99.