

Serbian Biochemical Society

Tenth Conference

with international participation

24.09.2021. Kragujevac, Serbia

“Biochemical Insights into Molecular Mechanisms”

Horseradish peroxidase C1A wild type gene and its variants expressed in *Pichia pastoris* KM71H strain

Ana Marija Balaz^{1*}, Milica Crnoglavac Popović², Marija Stanišić², Predrag Ristić², Milan Senčanski³, Tamara Todorović², Radivoje Prodanović²

¹ *Institute of Chemistry, Technology and Metallurgy, National Institute of the Republic of Serbia, University of Belgrade, Belgrade; Serbia*

² *University of Belgrade - Faculty of Chemistry*

³ *Vinča Institute of Nuclear Sciences, National Institute of the Republic of Serbia, University of Belgrade*

* *e-mail: anamarija.balaz@ihm.bg.ac.rs*

Enzyme immobilization enables maintenance of enzyme activity and structural stability even in adverse conditions ¹. Structural changes in enzymes that can occur due to the action of organic solvents, inhibitors or increased temperature can be prevented by immobilization of the enzymes in metal–organic frameworks (MOFs). It is reported that several enzymes, such as cytochrome c and horseradish peroxidase (HRP) have been successfully incorporated into MOFs ². The aim of this work is to produce wild type horseradish peroxidase, isoform C1A, and several mutants specially designed to increase the activity and stability of HRP while immobilized within selected MOFs. Wild type and its variants were produced in metalotrophic yeast, *Pichia pastoris* KM71H strain, their activity and basic kinetic parameters were determined and compared prior immobilization.

Acknowledgements

This study was funded by the Science Fund of the Republic of Serbia (program PROMIS, project SYMBIOSIS, #6066997). AMB and MCP contributed equally to this work.

References

1. Lee CH, Jin ES, Lee JH, Hwang ET. Immobilization and stabilization of enzyme in biomineralized calcium carbonate microspheres. *Front Bioeng Biotechnol* 2020;8:1191.
2. Sun Y, Shi J, Zhang S, Wu Y, Mei S, Qian W, Jiang Z. Hierarchically porous and water-tolerant metal–organic frameworks for enzyme encapsulation. *Ind Eng Chem Res* 2019;58:12835-44.