Serbian Biochemical Society
Sixth Conference

"Biochemistry and Interdisciplinarity: Transcending the Limits of Field"

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"***Biochemistry and Interdisciplinarity: Transcending the Limits of Field***"

Foreword

Dear Colleagues

It is my distinct pleasure to welcome you to the 6th Conference of the Serbian Biochemical Society, entitled "Biochemistry and Interdisciplinarity: Transcending the Limits of Field". It is an honor for me to be selected as the Editor of Proceedings of the Conference. I am grateful to the Steering Committee of Serbian Biochemical Society for giving me this opportunity to shape the premiere forum in biochemistry in the region. We have been tremendously fortunate to have Mihajlo B. Spasić as the first Editor. He nurtured this Conference (and Society) through its re-starting years as it grew in quality and relevance. Clearly, following in his footsteps is a challenge.

We have invited Djuro Josić from the University of Rijeka and eight experts from four major universities in Serbia to give lectures at the 6th Conference. The visit of our dear colleague from Croatia is a part of an initiative for closer collaboration within FEBS3+ (Croatia, Hungary, Slovenia, and Serbia) Meeting Programme that was established by FEBS in 2010. We have also invited students at the final years of PhD studies to present their work in our Proceedings as Abstracts. Official languages at the Conference will be Serbian, Croatian, and English.

I would like to express my gratitude to the members of the Scientific Board who suggested lecturers and to all respected colleagues who accepted the invitation.

Editor of the Proceedings
Ivan Spasojević

Semi rational design of cellobiose dehydrogenase from *Phanerochaete chrysosporium* for increased oxidative stability

Ana Marija Balaž[1*], Raluca Ostafe[2], Radivoje Prodanović[3]

[1]*Center of Chemistry, Institute of Chemistry, Technology and Metallurgy, University of Belgrade, Belgrade, Belgrade, Serbia*
[2]*Fraunhofer institute for Molecular Biology and Applied Ecology (IME), Aachen, Germany*
[3]*Faculty of Chemistry, University of Belgrade, Belgrade*

[*]*e-mail: ambalaz@gmail.com*

Cellobiose dehydrogenase (CDH, EC 1.1.99.18) from *Phanerochaete chrysosporium* belongs to a group of oxidoreductases and has the ability to degrade different components of woody plants. CDH is secreted by wood degrading, phytopathogenic and saprotrophic fungi and this widespread appearance implies hers important function and makes her an important enzyme for applications in industrial and biotechnological processes, as well as biosensors and biofuel cells. Cellobiose dehydrogenase is also used in industry for bleaching cotton and in food industry for lactose detection. CDH is monomeric enzyme consisting of two domains, flavin domain containing FAD as cofactor and smaller hem *b* containing cytochrome domain, connected via flexible linker. Physiological role of CDH is reflected in the degradation of cellulose and lignin in cooperation with other cellulolytic enzymes, because CDH catalyzes oxidation of celobiose (Glc –β–1,4 Glc) and other β–1,4–linked disaccharides and oligosaccharides to the corresponding lactons. Enzymes used in biosensors and for bleaching cotton should have high stability, especially toward reactive oxygen species. In order to improve oxidative stability of CDH, we have mutated CDH and tested its stability in presence of hydrogen peroxide. After successful cloning of the CDH gene in pYES2 vector, saturation mutagenesis was used to make library mutants where tree methionine residues were mutated. Residual activity of mutants was measured after the enzyme incubation in 0.3 M hydrogen peroxide for 0, 2 and 6h. After analysis of large number of mutants, it was observed that three mutants are showing higher oxidative stability compared to the wild – type enzyme. Residual activities of these mutants after 6 hour incubation in the hydrogen peroxide were over 50%, whereas wild-type has 30%. Selected mutants were expressed in *S. cerevisiae* and purified on DEAE column. Purity and activity of the enzymes were detected on the electrophoresis gel, oxidative stability of purified mutants was measured once again and characterization of these mutants was done. Mutants showing increased oxidative stability were sequenced and we have decided to

combine these mutations with each other in order to make combined mutants that will be tested for oxidative stability.