Journal of
Electrical Engineering and Electronic Technology

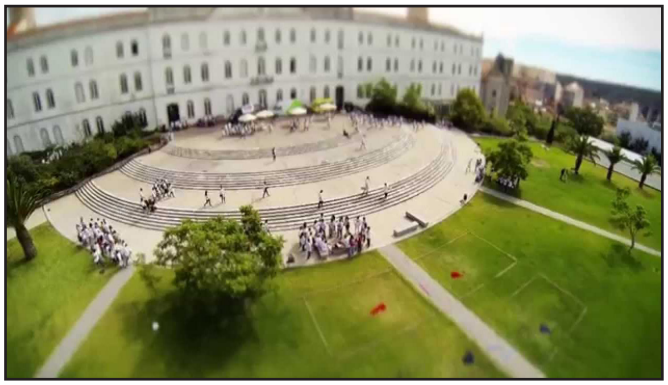
Abstract

Highly efficient hydrogen peroxide biosensors based on dye-decolorizing peroxidases

Catarina Barbosa
Universidade NOVA de Lisboa, Portugal

Abstract:
Dye-decolorizing peroxidases (DyP) are heme-containing enzymes that couple the oxidation of different substrates, such as phenolic and azo dyes and lignin related compounds with the reduction of H2O2 to water. Due to their broad substrate range, easy genetic manipulation and over-expression in E. coli, as well as their stability over wide ranges of pH and temperature, DyPs are considered an attractive target for development of biotechnological applications.1 In this work the DyP from Pseudomonas putida MET94 (PpDyP) and three variants obtained by directed evolution,2 which display increased resistance to H2O2 inactivation in solution, were explored for the construction of a H2O2 biosensor. The wild-type (wt) enzyme and variant forms 29E4 (E188K, H125Y mutations), 6E10 (E188K, A142V, H125Y mutations) and 25F6 (E188K, A142V, H125Y, G129D mutations) were immobilized on biocompatible silver electrodes. The structural properties of the adsorbed PpDyPs were addressed by surface enhanced resonance Raman (SERR) spectroscopy, and their electrocatalytic properties towards H2O2 reduction were studied by electrochemistry.

A successful immobilization maintaining the structural characteristics was achieved for all enzymes. From the four enzymes studied the wt and 29E4 PpDyP bioelectrodes displayed the best performance with a good dynamic response range (1-200 µM H2O2), high sensitivity (1.3–1.4 A□M-1□cm-2) for H2O2 and long term stability.3 With this work we show for the 1st time that DyPs can be used for the construction of 3rd generation H2O2 biosensors. Importantly, the proposed PpDyP devices display superior sensitivity in comparison to biosensors based on other peroxidases, such as horseradish peroxidase.4



Biography:
Catarina Barbosa is a BI fellow at the Raman BioSpectroscopy Lab at ITQB NOVA. She has a BSc. degree in Biochemistry from Faculdade de Ciências e Tecnologia da Universidade Nova de Lisboa (FCT/NOVA). In 2018 she obtained her MSc. degree in Biochemistry for Health at ITQB NOVA. Then in 2019 she started her present research fellowship. Her main research interest is the development of enzyme based electrochemical biosensors.

Recent Publications:

1. Colpa, D. I., Fraaije, M. W. & Bloois, E. Van. DyP - type peroxidases : a promising and versatile class of enzymes. J. Ind. Microbiol. Biotechnol. 41, 1–7 (2014).
2. Brissos, V., Tavares, D., Sousa, A. C., Robalo, M. P. & Martins, L. O. Engineering a Bacterial DyP-Type Peroxidase for Enhanced Oxidation of Lignin-Related Phenolics at Alkaline pH. ACS Catal. 5, 3454–3465 (2017).
3. Barbosa, C. et al. Immobilized dye-decolorizing peroxidase (DyP) and directed evolution variants for hydrogen peroxide biosensing. Biosens. Bioelectron. 153, (2020).
4. Koposova, E. et al. Bioelectrochemical systems with oleylamine-stabilized gold nanostructures and horseradish peroxidase for hydrogen peroxide sensor. Biosens. Bioelectron. 57, 54–58 (2014).

3rd International Congress on Biosensors and Bioelectronics; July 20-21, 2020; Paris, France

Citation: Catarina Barbosa; Highly efficient hydrogen peroxide biosensors based on dye-decolorizing peroxidases; Biosensors 2020; July 20-21, 2020; Paris, France.