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Biosensor based on CYP2D6-functionalised carbon nanotube transducer for continuous detection of xenobiotics

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The rising demand of personalised therapeutic products has led to an increasing interest in the investigation of sensing approaches for the detection of drugs, food compounds, and natural substances including potential mutual interferences. Although enzyme-based electro-chemical bio-sensors show significant potential towards the construction of a sensitive and selective sensor, a direct quantitative detection of the enzyme's catalytic activity still remains challenging. Distinct properties of the enzyme are required as well as an electrode in order to enhance the Direct Electron Transfer (DET) between the enzyme's catalytic active site and the electrode. Cytochrome P450 enzymes (CYPs) comprise for about 80% of the Phase-I drug-metabolizing enzymes in the human liver, whereas they functionalize chemicals that are foreign to the human body (xenobiotics) and endogenous substrates in order to increase their excretion. In our study, a screen printed working electrode with an additional carbon nanotube layer was functionalised with microsomes prepared from insect cells infected with recombinant baculovirus containing human isoenzyme cytochrome P450 2D6 (CYP2D6) and cytochrome P450 reductase, using a drop-casting technique and the immobilisation was additionally stabilised using two different cross-linkers, i.e., glutaraldehyde and poly(ethyleneglycol) diglycidyl ether, and an additional stabilising agent Nafion. The characterisation of the biosensor was conducted in a micro-cell, the activity of the immobilised CYP2D6 enzyme was measured by conversion of two model substrates, i.e., Luciferin-ME EGE and dextromethorphan, where the concentration of the products, i.e., Luciferin EGE and dextrorphan, were determined using high-performance liquid chromatography. The stability, selectivity and sensitivity of the biosensor were evaluated using a cyclic-voltametrical and chronoamperometrical processing of the induced signal.

Biography

Matic Krivec obtained his PhD degree in 2014 at Jozef Stefan International Postgraduate School (Slovenia) in the field of nanosciences and nanotechnologies and is currently a full-time Junior Researcher at CTR Carinthian Tech Research AG in Austria. His recent research topics are synthesis and characterization of nanoparticles, in particular TiO2 nanoparticles and carbon nanotubes, and their integration into smart devices (microreactors, gas sensors, electrochemical sensors). He is an author and co-author of 5 publications in peer-reviewed journals, 18 publications in conference proceedings and one PCT patent application.

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