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Integration of Horseradish Peroxidase/Carbon Nanotube to Construct a Novel Platform for Direct Electrochemistry of Enzymes and Biosensing Applications

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Abstract

In this work, we describe a new 3-dimensional (3D) network of crosslinked Horseradish Peroxidase/Carbon Nanotube (HRP/CNT) on a thiol-modified Au surface in order to build up the effective electrical wiring of the enzyme units with the electrode. This is achieved by the electropolymerization of aniline-functionalized carbon nanotubes (CNTs) and 4-aminothiophenol -modified-HRP on a 4-aminothiophenol monolayer-modified Au electrode. The synthesized 3D HRP/CNT network has been characterized with cyclic voltammetry and amperometry, resulting the establishment direct electron transfer between the redox active unit of HRP and the Au surface. Electrochemical measurements reveal that the immobilized HRP exhibits high biological activity and stability and a quasi-reversible redox peak of the redox centre of HRP was observed at about −0.355 and −0.275 V vs. Ag/AgCl. The electron transfer rate constant, KS and electron transfer co-efficient α are found to be 0.57 s-1 and 0.42, respectively. Based on the electrocatalytic process by direct electrochemistry of HRP, a biosensor for detecting H_2O_2 is developed. The developed biosensor exhibits excellent electrocatalytic activity for the reduction of H_2O_2. The proposed biosensor modified with HRP/CNT 3D network displays a broader linear range and a lower detection limit for H_2O_2 determination. The linear range is from 1.0×10^{-7} to 1.2×10^{-4}M with a detection limit of $2.2.0\times10^{-8}$M at 3σ. The Michaelies–Menten constant Kapp M value is estimated to be 0.18 mM. Moreover, this biosensor exhibits very high sensitivity, good reproducibility and long-time stability. Ease of fabrication, a low cost, fast response and high sensitivity are the main advantages of the new biosensor proposed in this study. These obvious advantages would really help for the real analytical applicability of the proposed biosensor.

Keywords: Redox chemistry, Biosensor, Carbon nanotube.