Characterization of an oxygen biosensor based on bilirubin oxidase enzyme efficiently tethered to a nanostructured carbon paper transducer

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Oxygen bioelectrocatalysis has potential relevancy for different fields namely industry, clinical and energy field, specifically through the development of efficient biosensors or biofuel cells capable to produce energy for low-power electronic devices. In an energy-demanding world, the design of self-powered biosensors is a very interesting possibility to be applied in the aforementioned fields. The recurrence to enzymes for catalytic processes enables high specificity to the substrate / target analyte at mild conditions. More concretely, the enzyme bilirubin oxidase (BOx) is a multicopper oxidase that performs the catalysis of oxygen naturally (acting as a co-substrate) and very efficiently. In electrochemistry, a direct electron transfer feature is easily attained with this enzyme even when immobilized simply by adsorption to a transducer. However the employment of certain tethering agents in immobilization can orientate the enzyme and lead to improved stability of the system [1]. Another important part in the bioelectrocatalytic system is the electrode nature itself. One relevant example is carbon fibre paper (CP) which is commonly used in both fuel and biofuel cells due to its high porosity and surface area, excellent mechanical and electronic properties [2]. In this sense, the objective of this work was the development of a simple but highly efficient oxygen biosensor that consisted in the immobilization of BOx to a multi-walled carbon nanotube (MWCNT) modified CP through pyrene-based bifunctional crosslinker. The electrode nanostructuration with MWCNT not only enables a higher electron transfer capacity but also allows anchoring of the enzyme. Cyclic voltammetry was used to assess enzyme immobilization, with the bioelectrode achieving a current density of -114 μA cm^{-2} at 0 V in the presence of O_2 (air-saturated) compared with only -31 μA cm^{-2} in the absence of O_2. The sensitivity was determined by amperometry with the biosensor achieving a value of 580 μA cm^{-2} mM^{-1} and the limit of detection corresponded to about 1 μM. To note also the great stability of the biosensor which practically maintained the sensitivity value after 5 days in storage at 4 ºC (106% compared to day 0).

REFERENCES

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