

A new nanocomposite amperometric biosensor for L-lactate determination

A. A. Vorobiienko, O. A. Biloivan

Institute of Molecular Biology and Genetics, NAS of Ukraine

150, Akademika Zabolotnoho Str., Kyiv, Ukraine, 03143

o.a.biloivan@edu.imbg.org.ua

biology20@ukr.net

Aim. The development of nanocomposite biosensor based on printed carbon electrode and immobilized lactate dehydrogenase for L-lactate determination in sweat. **Methods.** Electrochemical analysis of L-lactate provided by detecting reduced nicotinamide adenine dinucleotide (NADH) as a product of enzymatic conversion involving lactate dehydrogenase (LDH). In this regard, the study included the development of a high-performance nanocomposite sensor for monitoring NADH using screen-printed carbon electrodes (SPCEs). Modification of the surface of the working electrode was performed by drop method with a mixture of N-graphene with chitosan. An additional m-phenylenediamine membrane was applied by electropolymerization. The LDH layer was formed by covalent binding to BSA gel in glutaraldehyde vapors. The effects of SPCE modification on their electrochemical properties were investigated by cyclic voltammetry and amperometry. Spectrophotometric methods were used for the enzyme activity control and L-lactate determination in parallel with the biosensor measurements. **Results.** The laboratory models of an epidermal printed amperometric biosensor were developed for non-invasive determination of lactate in sweat. The analytical characteristics of the biosensors developed were investigated in model solutions (buffer, saline solutions, artificial sweat) in the mode of stationary cell. A comparative analysis of the analytical characteristics of the developed biosensors based on SPEs of different compositions and configurations and immobilized lactate dehydrogenase was performed. The influence of sweat components on the results of L-lactate measurements was studied. The composition of the biosensor and the measurement procedure for the determination of lactate in artificial sweat samples was optimized. The results of L-lactate measurements obtained using the developed laboratory model of sensor system and obtained by the traditional method of analysis are compared. **Conclusions.** The study has shown that the modification of the surface of the carbon electrode with a layer of 1 % N-graphene with 1 % chitosan increases the sensitivity three times and reduces the optimal potential for oxidation of NADH on the electrode surface (0.1–0.3 V). The developed and optimized model of the biosensor based on SPCE C110 (Drop Sens, Spain) and immobilized LDH allows the L-lactate analysis in the range of concentrations of 1–8 mM with a sensitivity of 0.145 $\mu\text{A}/\text{mm}$ and detection limit of 50 μM . It has been shown that an additional polymer membrane based on m-phenylenediamine significantly improves the selectivity of the biosensor. The operational stability of the biosensor was found to be 10 measurements of lactate and stability during storage at 4 °C in dry form — one month. The fundamental possibility of determining the concentration of lactate in samples of artificial sweat using the developed biosensors is shown.