SPECTROSCOPIC PROPERTIES AND ACTIVITY OF GLUCOSE OXIDASE

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Usage of biosensing systems is the promising, rapid and accurate method for detection and analysis of various compounds concentration. Nowadays the best-known and the most common biosensors are amperometric glucose biosensors. They can detect blood's free glucose in a short amount of time – it is very important for people with diabetes. The active part of the biosensor is glucose oxidase (GOx) enzyme immobilized on the surface of the electrode [1-3]. In order to create a non-invasive biosensor based on GOx, it is necessary to understand the properties of this enzyme related to the influence of the surrounding environment.

The purpose of this research was to evaluate absorption and fluorescence spectra changes and to associate them with changes of GOx activity.

During this work, the changes of the spectroscopic properties of GOx and flavin adenine dinucleotide (FAD) were investigated in three different cocentration citrate-phosphate buffering solutions with pH values in the range [3 ÷ 8]. During the study, changes in stationary absorption, fluorescence spectra, and fluorescence emission kinetics were observed during the 16-days period. The activity of the GOx enzyme was determined in two different ways – Spectrophotometric Benzoquinone method and Colorimetric method.

GOx enzyme selectively catalyzes the oxidation of D-glucose. Products of the reaction change the stability of the solution system. This leads to changes of GOx spectroscopic properties, which are associated with changes in the structure of the enzyme, during which the FAD is more quickly released from the active center of the enzyme [3].

During this study, we found that GOx enzyme fluorescence band at 530 nm belongs to coenzyme FAD fluorescence. In 16 days measurement period, the increase of GOx fluorescence, which is due to the fact that the enzyme has changed its structure and denatured, was visible.

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