

ENZYMATIC BIOSENSORS FOR POINT-OF-CARE TESTING OF α -AMYLASE AND β -GALACTOSIDASE

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Introduction. α -Amylase is a glycosidase that catalyzes the hydrolysis of α -1,4-glycosidic bonds in starch which liberates glucose, maltose, and limit dextrin. Salivary α -amylase has become one of the most promising biomarkers in clinical diagnostics. If the individual suffers from chronic psychosocial stress, the concentration of α -amylase in saliva increases ten times in comparison to the normal value [1]. β -Galactosidase (lactase) is a glycoside hydrolase enzyme that catalyzes the hydrolysis of β -galactosides into monosaccharides (galactose and glucose) by disrupting β -1,4-glycosidic bonds. Salivary β -galactosidase is associated with halitosis and reduction of enzyme levels plays an important role in therapy [2]. Therefore, it is essential to develop a simple and cost-effective way to monitor α -amylase and β -galactosidase in saliva.

Main part. This work is devoted to the development of biosensors for detecting the activity of enzymes listed above. The biorecognition element of the biosensors consists of specific carbohydrates (enzyme substrates), glucose oxidase, and nanosized titanium dioxide. The working principle of the biosensors is based on the evaluation of the color intensity of yellow-colored peroxy titanium complex, which is formed as a result of the interaction between titanium dioxide and hydrogen peroxide released during an enzymatic reaction.

To determine α -amylase, starch is cleaved by α -amylase producing maltose which is hydrolyzed by α -glucosidase to glucose that is then oxidized by glucose oxidase. As the result of oxidation, hydrogen peroxide is released and reacts with titanium dioxide. Whilst the determination of β -galactosidase involves the hydrolysis of lactose to glucose.

In this work, titanium (IV) oxide was synthesized by the sol-gel method and was subjected to dialysis for 1.5 days to adjust pH. Dialyzed sol was mixed with saccharides and glucose oxidase and then applied to the film. The reflection spectra and the calibration curves for different enzyme activity levels were obtained by diffuse reflectance spectroscopy.

Conclusion. This work explores a novel strategy for identification of α -amylase and β -galactosidase in saliva. It does not require expensive laboratory instrumentation, operator expertise and can be easily incorporated into routine point-of-care analysis.

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References:

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