Institute of Molecular Biology and Genetics National Academy of Sciences of Ukraine

POTENTIOMETRIC ENZYME BIOSENSOR MODIFIED WITH GOLD NANOPARTICLES

SHKOTOVA L.V.*, ZINCHENKO O.A., ARKHYPOVA V.M., DZYADEVYCH S.V.

*luda_shkotova@yahoo.com

The ever-increasing number of necessary clinical tests in medical diagnostics requires highly sensitive, selective, rapid and economic methods of analysis. The decisive factors of the development of laboratory diagnostics are the elaboration and implementation of new technologies to meet the main requirements to modern laboratory equipment - reliability, efficiency and ease-of-use.

The work is aimed on application of gold nanoparticles for the improvement of characteristics of potentiometric enzyme biosensors based on immobilized butyrylcholinesterase (BuChE).

pH-sensitive field-effect transistor

The responses of pH-FET sensors (Fig. 1.) were measured using a circuit of direct measurement of the current in the channel of field-effect transistor with an active load. The threshold voltage was about -2.5 V. The conditions of measurements were as follows: the channel current is about 500 μ A, the drain-source voltage is about 2 V, the substrate is connected to the drain.

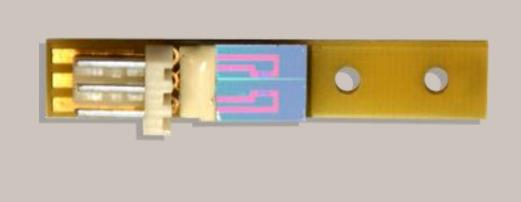


Fig. 1.

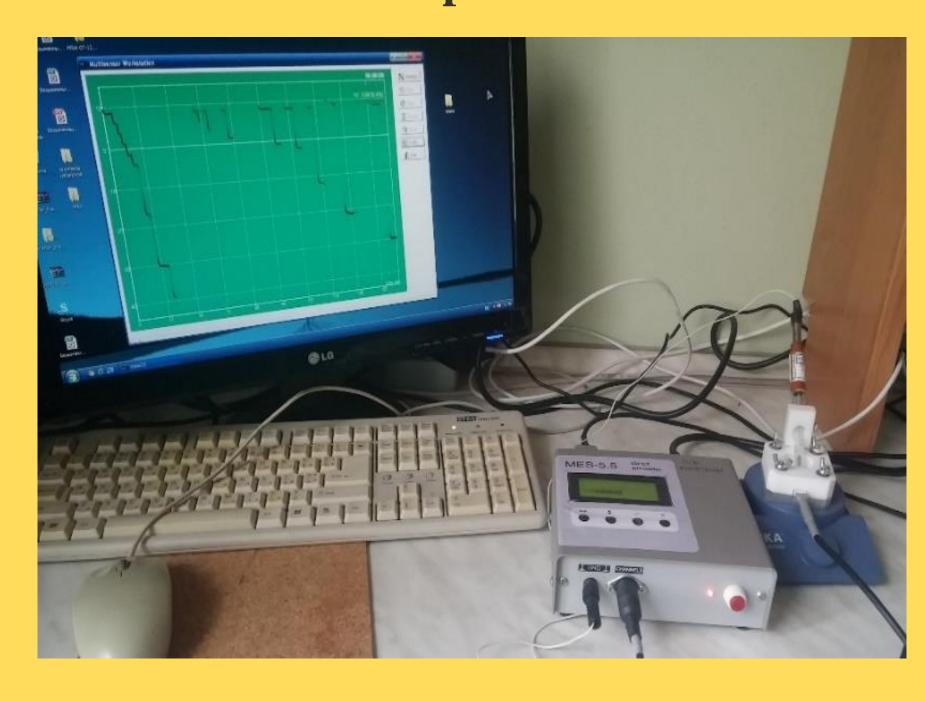
The composition of a butyrylcholinesterase-based bioselective membrane with gold nanoparticles was optimized. Optimal conditions of the biosensor functioning, such as pH and ionic strength of the working buffer solution, were selected. The signal reproducibility of the developed biosensor and its storage stability were studied (Tab.).

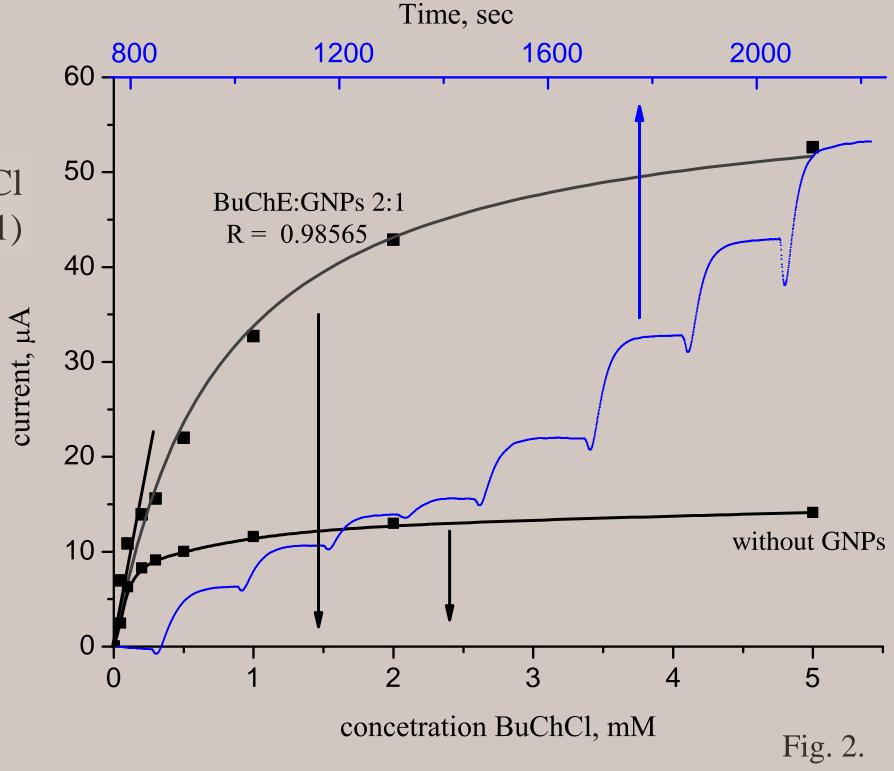
			_
Parameters	BuChE without GNPs	BuChE:GNPs 2:1	BuChE:GNPs 1:1
Minimum detection limit, mM	0.05	0.025	0.025
Linear range of measurement, mM (R correlation)	0.05 - 0.2 R=0.983	0.025 - 0.2 R=0.986	0.025 - 0.3 R=0.998
Response time, min	2.0	1.4	1.35
Storage stability, % residual activity after	84	27	56
20 days pH optimum	7.5	7.5	7.0

Fig.2. The calibration curve (the dependence of response on the BuChCl concentration) for the biosensor with GNP (the ratio of BuChE to GNP is 2:1)

A highly sensitive, selective potentiometric biosensor based on immobilized butyrylcholinesterase was developed; its laboratory prototype has been created. The enzyme served as a sensitive element and pH-sensitive field-effect transistors (FETs) were used as the transducers of a biochemical signal into electrical signal.

General view of potentiometric device





Acknowledgements

The work was carried out thanks to financial support from the National Research Fund of Ukraine in the framework of the competition of projects for research and development "Support of research of leading and young scientists" (project 2020.02/0097).