Kamila Spychalska, MSc

Construction of biosensor systems modified with semiconductor structures

Disturbances in the homeostasis of the human and animal organism, resulting from the presence of endocrine disrupting compounds (EDC) in aqueous solutions, can cause the appearance of many serious, long-lasting diseases. Hence, it is very important to develop more sensitive and faster analytical methods that enable the qualitative and quantitative control of these compounds. Classic biochemical tests very often turn out to be too time-consuming and insufficiently accurate, which is why increasingly attention has been paid to the development of modern diagnostic techniques, including biosensors.

Biosensors are devices belonging to the group of chemical sensors. These systems consist of two basic elements: the bioreceptor layer, also known as the biologically active layer (responsible for specific recognition of the tested substance) and the transducer part (converting the obtained chemical or biochemical signal into a measurable value, e.g. current intensity). The exceptional sensitivity and selectivity of biosensors results from the presence of a biologically active material (e.g. an enzyme, antibody or cell), which is an inherent element in the receptor layer. The construction of biosensors was qualified in 2012 by the Ministry of Economy of the Republic of Poland as one of the priority technologies indicated in the Foresight technological industry project - InSight 2030, related to the recognition of strategic technologies.

Due to the worldwide demand for fast and cheap analytical methods, the research carried out as part of my doctoral dissertation is aimed at intensifying research on the design of a new generation of miniature diagnostic devices with wide application possibilities. In my research work, I focus on the design and construction of electrochemical immunosensors and enzymatic biosensors for determining the concentration of steroid hormones in aqueous solutions, which could be used in the future as tools for environmental protection.

I started designing individual biosystems with the selection of an appropriate semiconducting compound used in the production of the chemosensitive layer. The main argument for creating a thin film layer covering the electrode surface was the establishment of a highly efficient contact between the immobilized biologically active element (enzyme protein,

antibody) and the transducer. In the course of the research, platinum electrodes were modified with heterocyclic derivatives of pyridine, dithiensilol and benzothiadiazole.

I tested selected heterocyclic compounds in terms of their electropolymerization or self-organization on the electrode surface. In the conducted research, I used electrochemical techniques, such as differential-pulse voltammetry, cyclic voltammetry, chronoamperometry and physical adsorption. I characterized the modified surfaces using scanning electron microscopy.

I used the electrodes prepared in this way in the further part of the research for the construction of bioreceptor layers. In enzyme biosystems, it is essential to maintain the catalytic activity of immobilized proteins. The material covering the electrode should not only keep the enzyme stable on its surface, but also prevent a possible loss of its catalytic activity. In the case of antibodies, it is important to create a strong and stable interaction between the antibody and the material covering the electrode - during measurements it is necessary to wash out the antigen associated with the antibody while maintaining a stable coverage of the semiconducting layer by proteins.

In subsequent research stages, proteins belonging to the oxidoreductase class (horseradish peroxidase, laccase and tyrosinase) as well as anti-17β-estradiol monoclonal antibodies were immobilized on previously modified substrates, using physical adsorption and covalent cross-linking methods. The effectiveness of the deposition of enzymes on the electrode surface and the measurement of their catalytic activity was carried out using colorimetric techniques with the use of a spectrophotometer.

The last stage of my research involved combining the created chemosensitive layer with an electrochemical transducer, and then carrying out amperometric measurements in the presence of selected analytes in order to define the analytical parameters of the constructed systems.

The above-mentioned research works allowed for the construction of 10 stable biosystems, enabling the monitoring of the concentration of steroid hormones belonging to the group of endocrine-active compounds in laboratory and pharmaceutical samples. Such devices have enormous potential to become commercially available sensors, which are an excellent alternative to traditional analytical methods enabling fast and sensitive in situ measurements.