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DEVELOPMENT AND ADAPTATION OF A BILIRUBIN-SENSITIVE BIOSENSOR FOR CLINICAL DIAGNOSTICS

Aim. Bilirubin is the final product of heme degradation, and its elevated levels are associated with hyperbilirubinemia, jaundice, hemolytic disorders, hereditary syndromes, and liver diseases. Owing to the clinical importance of bilirubin determination, this study focused on the development and optimization of an amperometric biosensor based on bilirubin oxidase for sensitive and selective bilirubin detection. **Methods.** An amperometric biosensor based on bilirubin oxidase (BOx) was employed, where BOx catalyzed the oxidation of bilirubin to biliverdin. The resulting product was further electrochemically oxidized at -0.15 V, generating purpurin, electrons and protons, which produced a current response proportional to the analyte concentration. **Results.** During the study, the composition of the bioselective element was optimized, in particular, the optimal concentrations of 0.75% glutaraldehyde and 15% BOx were established, and the working potential of -0.15 V was determined, at which the maximum amperometric response to bilirubin was observed. It was shown that increased concentrations of NaCl and protein components in the samples reduced the signal intensity, but this effect was eliminated by diluting the samples ten-fold before analysis. At the same time, biosensor demonstrated high selectivity to the main biological interferences (amino acids, glucose, urea, creatinine), a linear operational range of $7.5\text{--}1000\text{ }\mu\text{M}$, a detection limit of $7.5\text{ }\mu\text{M}$ of bilirubin and high reproducibility (RSD = 6.7%). All of this confirms its potential for clinical application in the future. **Conclusions.** In this work, an amperometric biosensor based on BOx was developed and optimized, which provides a selective and sensitive response in a wide range of bilirubin concentrations. The results obtained confirm the suitability of the proposed system for the accurate determination of bilirubin in biological fluids and its potential for clinical application. **Grants.** The work was carried out as part of project no. 225067 of the Ukrainian Swiss Joint Research Projects: Call for Proposals 2023 program. The part of work was funded by the Simons Foundation № SFI-PD-Ukraine-00017453.

Keywords: biosensor, clinical diagnostics, biomarker, bilirubin-sensitive sensor.

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