Urease inhibition-based biosensor for heavy metal ions determination to control wastewater treatment in textile industry

Bakhmat V. A.[1,2] , Soldatkin O.O [1,2] , Pyeshkova V.M. [1,2,3], Dzyadevych S.V. [1,2]

[1] Institute of Molecular Biology and Genetics of NAS of Ukraine, Zabolotnogo Street 150, 03143 Kyiv, Ukraine,

[2] Taras Shevchenko National University of Kyiv, Volodymyrska Street 64, 01003 Kyiv, Ukraine

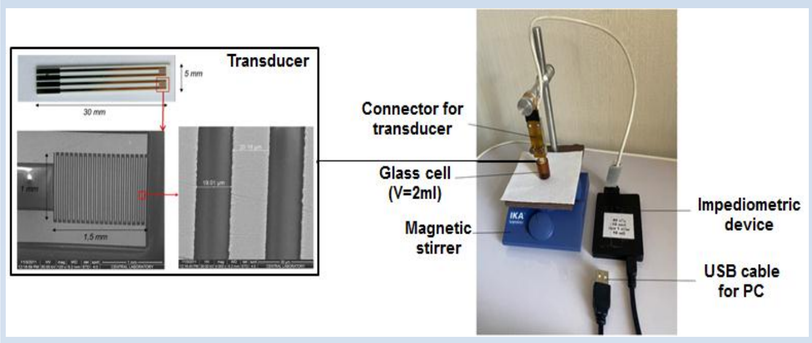
[3] School of Engineering and Innovation, The Open University, MK7 6AA, Milton Keynes, The United Kingdom

E-mail: veronikab2406@gmail.com

Textile wastewater heavy metal pollution has become a severe environmental problem worldwide. A lot of dyes that are used in the textile industry contain heavy metals. The high concentrations of heavy metals in effluents from textile industry cause severe toxicological implications on the environment with a dramatic impact on human health. Conventional heavy metal detection methods often require expensive equipment, highly skilled personnel, the application of complex operational procedures, long detection times, and they are not applicable for real-time detection.

The main goal of this work was to develop and optimize urease inhibition-based impediometric biosensor for the determination of heavy metal ions (HMIs) to control wastewater treatment in textile industry. As a result of this work new impediometric urease-inhibition based biosensor was developed for determination of HMIs in water. A differential pair of gold interdigitated electrodes deposited on a ceramic substrate was used as the impediometric transducer. As a bioselective element, urease was chosen, which was immobilized by cross-linking with glutaraldehyde on the surface of electrodes. The developed biosensor was characterized by high signal reproducibility (RSD < 3%). It was shown, that the developed biosensors can be stored at - 4°C for a 10 month without loss of activity more than 13%. The biosensors sensitivity toward 0.001-10 µM concentration of different heavy metal ions was investigated. The results showed the possibility of analysis of very low concentrations of silver ions (LOD = 1 nM). It was found that the optimal incubation time of bioselective elements in solution with heavy metal ions was 30 minutes. The possibility of reactivation of biosensors using EDTA was evaluated in order to reuse the biosensors for the inhibitory analysis. It was investigated how inhibitor concentration, time of incubation with the reactivator and other parameters influence the reactivation efficiency. The optimal incubation time of biosensors in a solution with EDTA was 30 min. The possibility of multiple reactivations of the developed biosensor was estimated with the purpose of its repetitive application for heavy metal ions determination in water solution.

The image of electrochemical transducers that consist of two pairs of gold interdigitated electrodes and photo of the impediometric device that was used in our work.



All measurements were carried out in 5 mM phosphate buffer solution, pH 7.35

The biosensor operation is underlain by the following enzymatic reaction:

Urease

$$H_2N\text{-}CO\text{-}NH_2 + 2H_2O + H^+ \rightarrow 2NH_4^+ + HCO_3^-$$

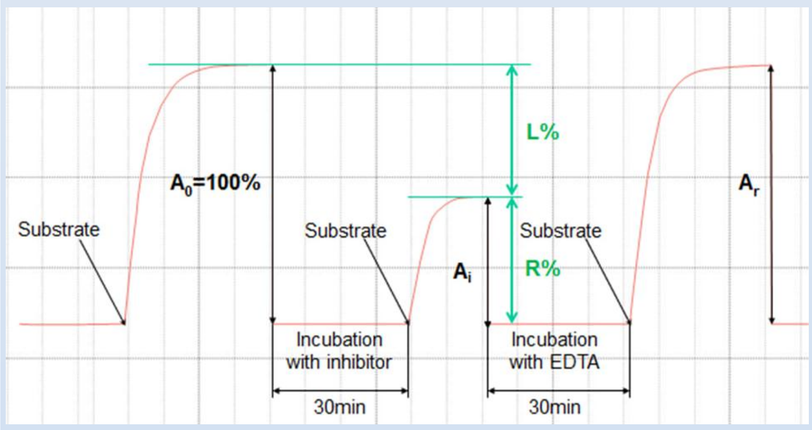
Heavy metal ions inhibit enzymes in following way:

$$Enz\text{-}SH + Me^+ \rightarrow Enz\text{-}S\text{-}Me + H^+$$

The initial enzyme activity after inhibition can be recovered by the reactivator EDTA:

$$Enz\text{-}S\text{-}Me + EDTA \rightarrow Enz\text{-}S\text{-}H + EDT\ [Me]$$

Inhibitory analysis



Biosensor signals:

A_0— before inhibition

A_i — after inhibition

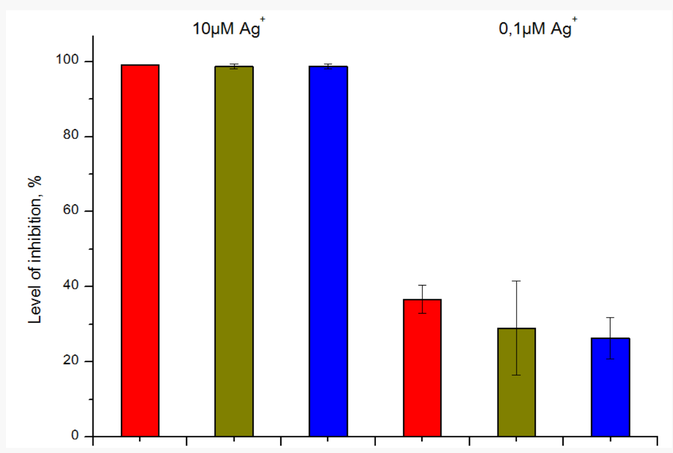
A_r — after reactivation EDTA

R — residual activity

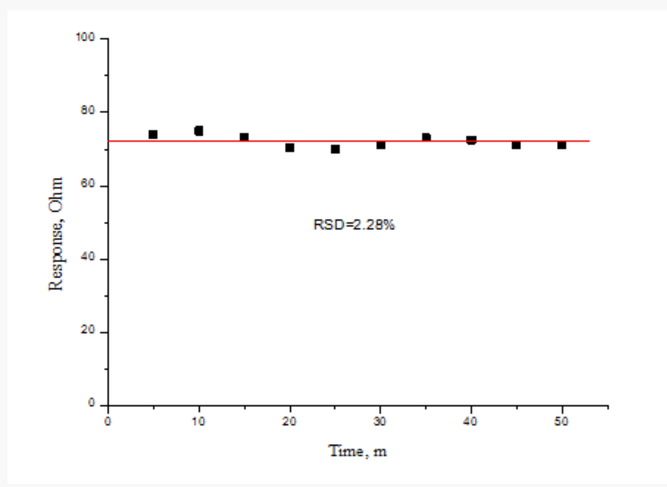
$R=A_i*100/A_0$

L — level of inhibition

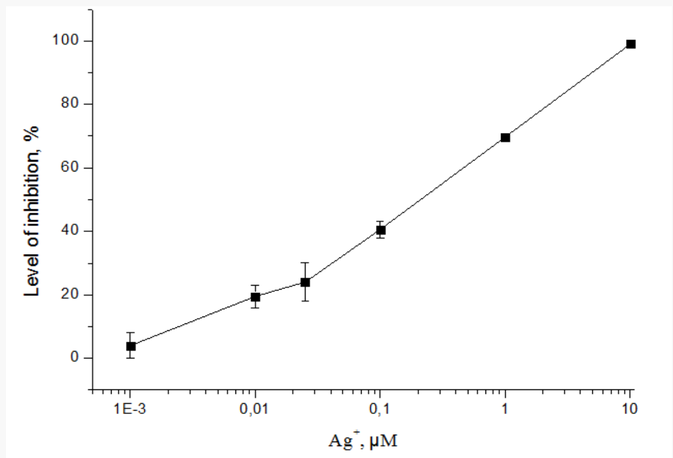
$L=(A_0-A_i)*100\%/A_r$



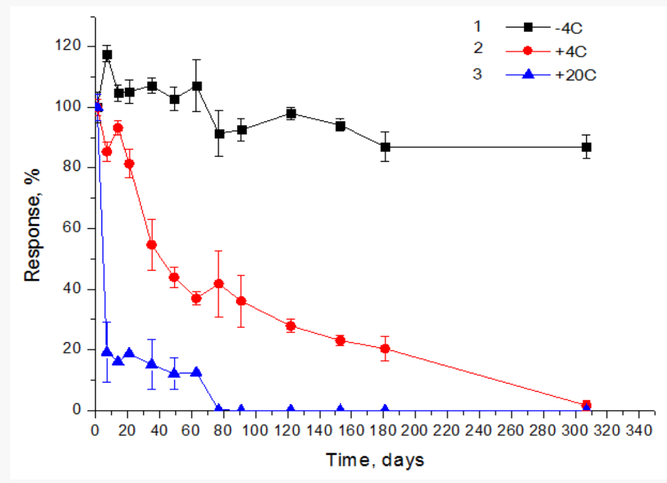
The level of inhibition by Ag^+ ions biosensors based on ureases with different activity (red –115, green – 31, blue – 67, U/mg).



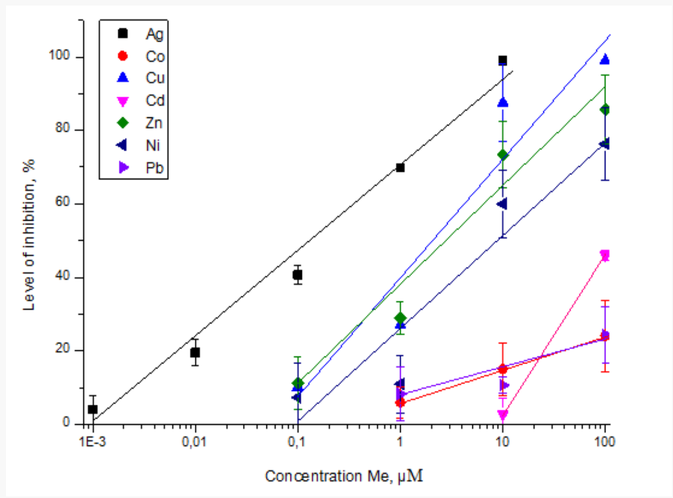
Reproducibility of urease biosensor.



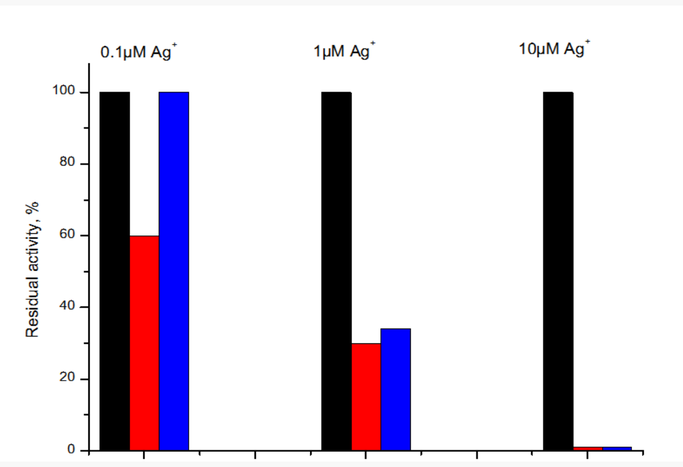
Calibration curve of urease biosensor for silver ions determination.



Storage stability of urease biosensor.



Dependence of level of inhibition of urease biosensors on different concentrations of heavy metal ions.



Residual activity of biosensor before (black), after inhibition by Ag^+ ions (red) and after reactivation by EDTA (blue) .

Conclusions

As a result of this work new impediometric urease-inhibition based biosensor was developed for determination of heavy metal ions in water. Sensitivity of urease biosensors toward different heavy metal ions was investigated. The possibility of biosensor reactivation with EDTA after inhibition was demonstrated. The biosensor was characterized by high reproducibility of responses and showed high storage stability. In future the developed biosensors can be used for heavy metal ions detection to control wastewater treatment in textile industry.

Acknowledgements

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Waste2Fresh