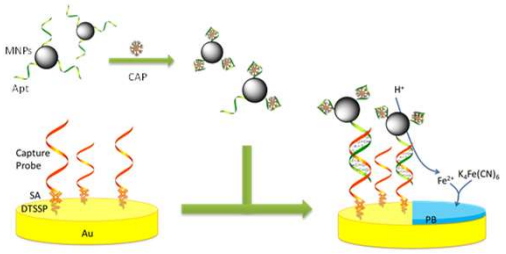
A novel electrochemical biosensor based on magnetic-separation/concentration-signal-amplification in-one method for chloramphenicol detection

Lulu Cao,[a] Yingchun Fu,[a,*] Qi Zhang,[a] Huang Dai,[a] Yibin Ying,[a] Yanbin Li,[a,b]
[a] College of Biosystems Engineering and Food Science, Zhejiang University, Hangzhou 310058, China
[b] Department of Biological and Agricultural Engineering, University of Arkansas, Fayetteville, AR 72701, United States

Abstract

Here, we propose a novel magnetic-separation/collection-signal-amplification in-one method to develop a facile and sensitive electrochemical biosensor for chloramphenicol (CAP) detection. Briefly, aptamer-modified magnetic nanoparticles (MNPs-Apt) was designed to capture CAP in sample, then the MNPs-Apt composite was conjugated to Au electrode through the DNA hybridization between the unoccupied aptamer and a strand of complementary DNA, followed by an electrochemical conversion method (ECC) of MNPs labels to electrochemically active Prussian blue (PB) the method significantly promoted the signal amplification. Therefore, the proposed biosensors exhibited linear detection range covering three magnitudes and a limit of detection down to 1 ng mL^{-1}



Scheme 1. Schematic representation of new method for electrochemical detection of CAP.

Introduction

- As well known, using magnetic materials, such as magnetic nanoparticles (MNPs), to separate and concentrate target has been one of the most common and promising methods for sample pretreatment due to their unique magnetism behavior and abundant surficial properties
- Electrochemical biosensor has attracted considerable attention for its high sensitivity and speed, simplicity, and low cost for signal output

Result and Discussion

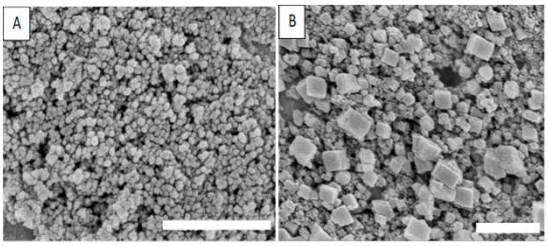


Fig. 1. SEM images of the MNPs-Apt modified electrode before (A) and after (B) the ECC treatment. Scale bar: 500 nm

For the pristine MNPs-Apt, nanoparticles of ca. 20 nm in diameter were observed with smooth surface. However, after the ECC treatment, it appeared plenty of particles with cube-like shape, which is a characteristic shape of PB crystal.

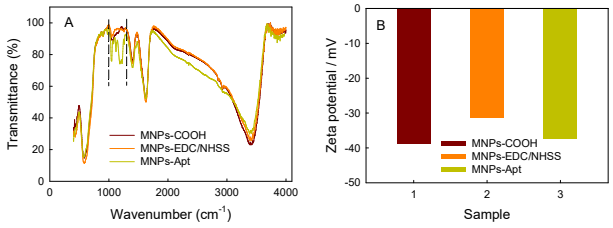


Fig. 3. FT-IR curves (A) and Zeta potentials (B) of MNPs-COOH, MNPs-EDC/NHSS and MNPs-Apt

To ensure the modification of aptamer on the MNPs, we adopted FT-IR and Zeta potential to investigate different processes of the modifications, including the pristine MNPs-COOH, EDC/NHSS activated MNPs-COOH and the MNPs-Apt. From the FT-TR curves as shown in Fig. 3A, the pristine and activated MNPs-COOH showed minor difference, compared with which, the MNPs-Apt presented series of newly appeared absorbance peaks located from 1000 cm^{-1} to 1243 cm^{-1}, which are ascribed to the phosphodiester groups of nucleic acid

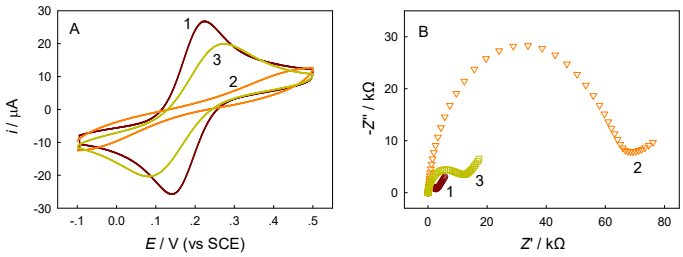


Fig. 3. CV (A) and EIS (B) of bare electrode, MNPs-Apt-conjugated electrodes before and after ECC process in the absence of $K_4Fe(CN)_6$. Characterization was conducted in a PBS solution containing 1.0 mM $Fe_3(CN)_6$/1.0 mM $K_4Fe(CN)_6$. Scan rate: 0.1 V·s^{-1}. EIS: 100 kHz-0.1Hz, 100 mV rms, 0.21 V bias

Interestingly, we also conducted the ECC treatment of the final MNPs-conjugated electrode in the absence of $K_4Fe(CN)_6$ (no PB yielded), and found a refreshing effect through the monitoring by CV and EIS with electrochemical probe of 1 mM $K_3Fe(CN)_6$/$K_4Fe(CN)_6$, as shown in Fig. 4.

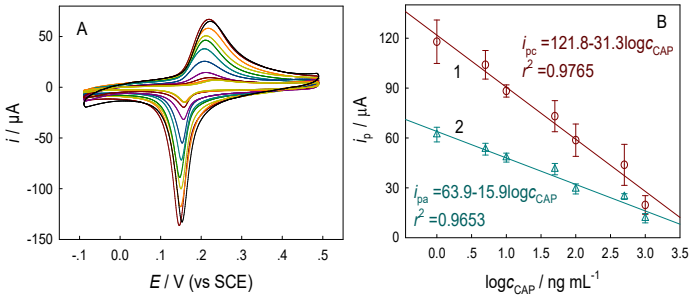


Fig. 5. (A) CV response curves of CAP at different concentrations (from bottom to top: control, 0.5, 1, 5, 10, 50, 100, 500, 1000, 5000, 10000 ng mL^{-1}). (B) The calibration plot of reductive (1) and oxidative (2) peaks currents to CAP concentrations.

Both the reduction and oxidation peak currents of PB were collected and analyzed. As shown in Fig. 5, along with the increase of the concentration of CAP, more aptamers were occupied by CAP and lower probability for the conjugation of MNPs labels to electrode remained. Therefore, the current responses presented decreased signal intensity. When adopting both the cathodic and anodic peak currents as the signals, the biosensor exhibited a linear detection range (LDR) from 1 ng mL-1 to 1000 ng mL-1, which is as wide as 3 orders of magnitude, as well as a limit of detection (LOD) down to 1 ng mL-1 (S/N=3).

Conclusion

- We have explored a magnetic-separation/concentration-signal-amplification in-one method to develop a facile and sensitive electrochemical biosensor for CAP detection. This method integrated the rapid separation and concentration function of MNPs, and signal readout/amplification ability by exploring the ECC process to obtain electrochemically active PB. Taking advantages of the high abundance of iron content in MNPs and the refreshing effect, the method significantly promotes the signal amplification without any additional labels. Therefore, the proposed biosensors exhibited satisfactory performance which was better than or comparable with those of most analogues. The developed method may lead to new concept for developing biosensor for rapid and facile detection in food safety, clinic diagnose/therapy and environmental monitoring fields.

Acknowledgements

This research was supported in part by Walmart Foundation through Walmart China Food Safety Collaboration Center (project # 0402-70013-21-0000).

References

- Yan, Z.D., Gan, N., Li, T.H., Cao, Y.T., Chen, Y.J., 2016. Biosens. Bioelectron. 78, 51-57.
- Yin, J.L., Guo, W.J., Qin, X., Zhao, J.L., Pei, M.S., Ding, F., 2017. Sens. Actuators, B 241, 151-159.