

A microfluidic impedance aptasensor based on immunomagnetic separation and urease catalysis for continuous-flow detection of pathogenic bacteria



Lan Yao¹, Lei Wang¹, Fengchun Huang¹, Gaozhe Cai¹, Xinge Xi², Jianhan Lin^{1*}

¹China Agricultural University, Beijing, China

²University of Arkansas, Fayetteville, AR, US

*corresponding author: Dr. Jianhan Lin, jianhan@cau.edu.cn



Abstract

Foodborne pathogens have resulted in serious public health issues and numerous economic losses, and early screening of foodborne pathogens in food supply chains is a key to control the outbreaks of foodborne illnesses. In this study, a microfluidic impedance aptasensor combined with the immune magnetic nanoparticles, the urease and the interdigitated microelectrode in the microfluidic channel was developed for rapid, sensitive and continuous-flow detection of target bacteria. The impedance was online measured and analyzed using the impedance normalization to determine the concentration of the target bacteria. A good linear relationship between the impedance relative change rate of the catalysate and the concentration of the bacteria was obtained with low detection limit of 12 CFU/mL. This aptasensor has the potential for online screening of foodborne pathogens.

Principle

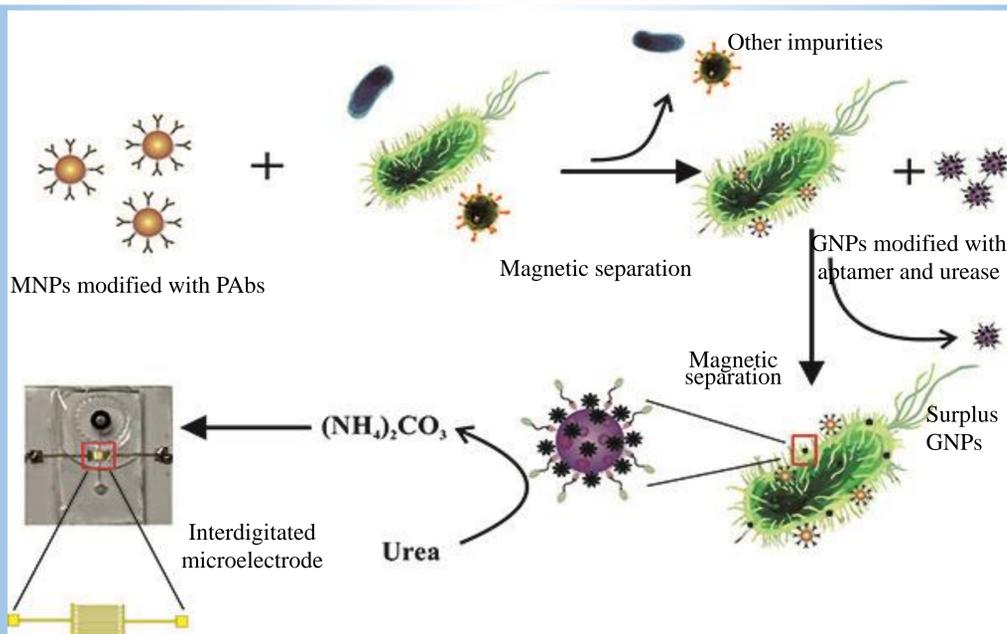


Fig. 1. The principle of the microfluidic impedance aptasensor for online detection of foodborne pathogen using immune-magnetic separation, urease catalysis and continuous-flow impedance measurement.

Results and Discussion

Proof of concept on online impedance measurement

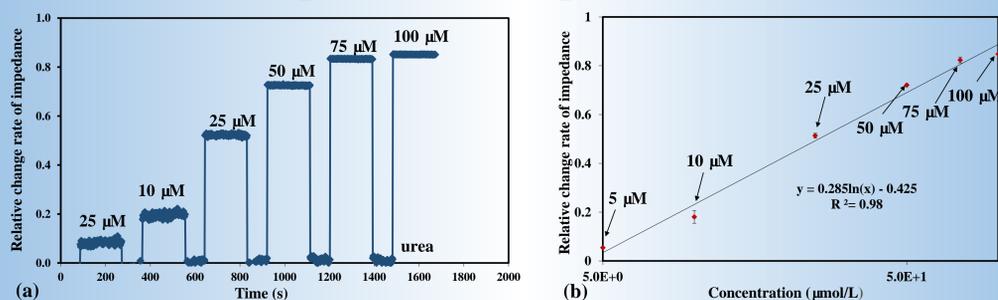


Fig. 2. (a) The relative change rate of impedance for ammonium carbonate; (b) The linear relationship between the relative change rate of impedance and the concentration of ammonium carbonate (N=3).

Bacteria separation

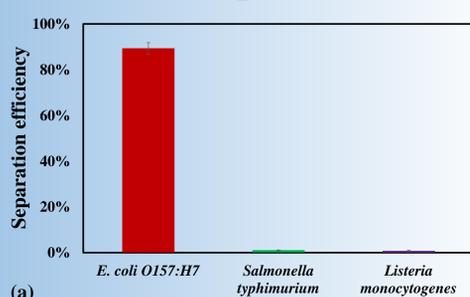
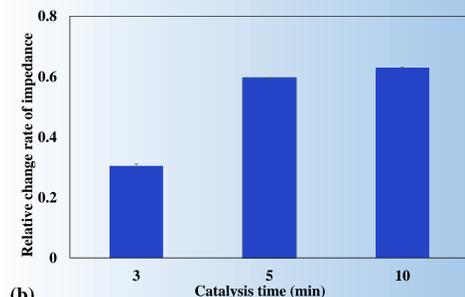


Fig. 3. (a) The separation efficiency of the target bacteria and the non-target bacteria (N=3); (b) The relative change rate of impedance measured at the characteristic frequency of 15 kHz for different enzymatic catalysis time in the detection of *E. coli* O157:H7 at the concentration of 10² CFU/mL (N=3).

Catalysis time optimization



Normalization of the impedance data

Table 1 Comparison of different data analysis methods

Electrode	Z _s (kΩ)	Z _c (kΩ)	ΔZ (kΩ)	RZ
Electrode 1	37.1	46.0	8.9	0.19
Electrode 2	80.5	102.6	22.1	0.22
Electrode 3	43.5	54.9	11.4	0.21
Average	53.7	67.8	14.1	0.21
Standard deviation	23.4	30.4	7.0	0.01
Relative standard deviation	44%	45%	50%	5%

Sensitivity and specificity of this aptasensor

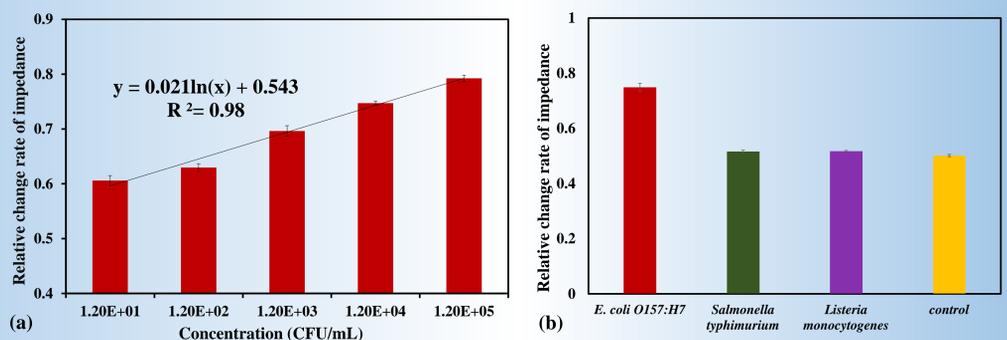


Fig. 4. (a) The separation efficiency of the target bacteria, *E. coli* O157:H7, and the non-target bacteria, *Salmonella typhimurium* and *Listeria monocytogenes* (N=3); (b) The relative change rate of impedance of the target bacteria, *E. coli* O157:H7, and the non-target bacteria, *Salmonella typhimurium* and *Listeria monocytogenes* (N=3).

Conclusions

- This aptasensor could accurately measure the impedance in continuous-flow condition.
- The impedance normalization could reduce the impact from microelectrode variation.
- This aptasensor was able to detect *E. coli* O157:H7 as low as 12 CFU/mL within 2 h based on that the urease catalysis was used to greatly amplify the detection signal.

Ongoing and Future Work

The impedance of the solution in the microfluidic chip can be detected by the hand-held detector and the data can be sent to the phone that had an App on it.

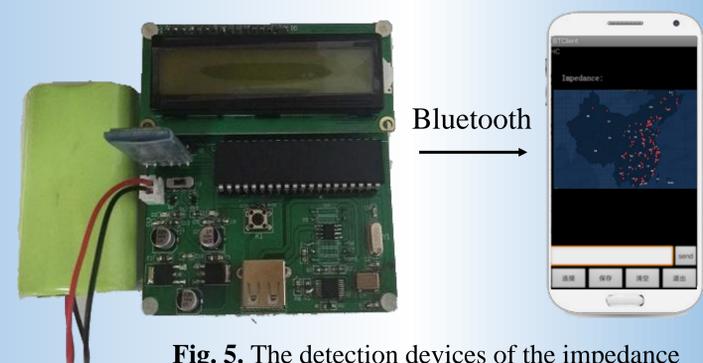


Fig. 5. The detection devices of the impedance

Acknowledgment

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