

## Abstract

In this study, we proposed a novel impedance biosensor combining immunomagnetic separation with urease catalysis for sensitive detection of foodborne pathogen. The target bacteria was first separated and concentrated using the monoclonal antibody modified magnetic nanoparticles, followed by reacting with the polyclonal antibody and urease modified gold nanoparticles to form the sandwich complexes. After the urease on the complexes were used to catalyze the hydrolysis of urea into ammonium carbonate, the impedance change was measured using the interdigitated array microelectrode. This impedance biosensor was able to detect target bacteria as low as  $10^2$  CFU/mL in 1 h.

## Principle

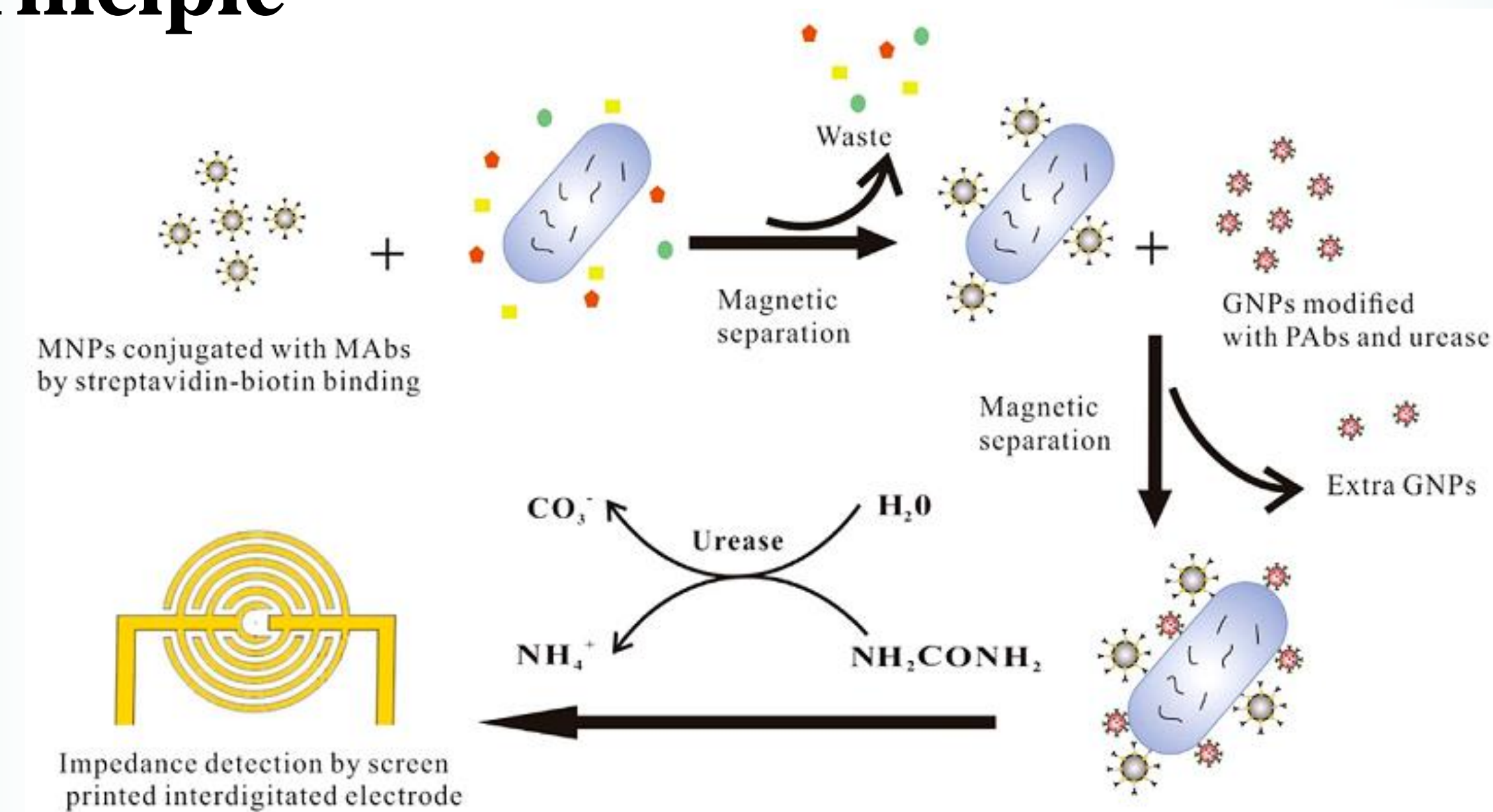


Fig.1. The principle of the impedance biosensor based on immunomagnetic separation and urease catalysis.

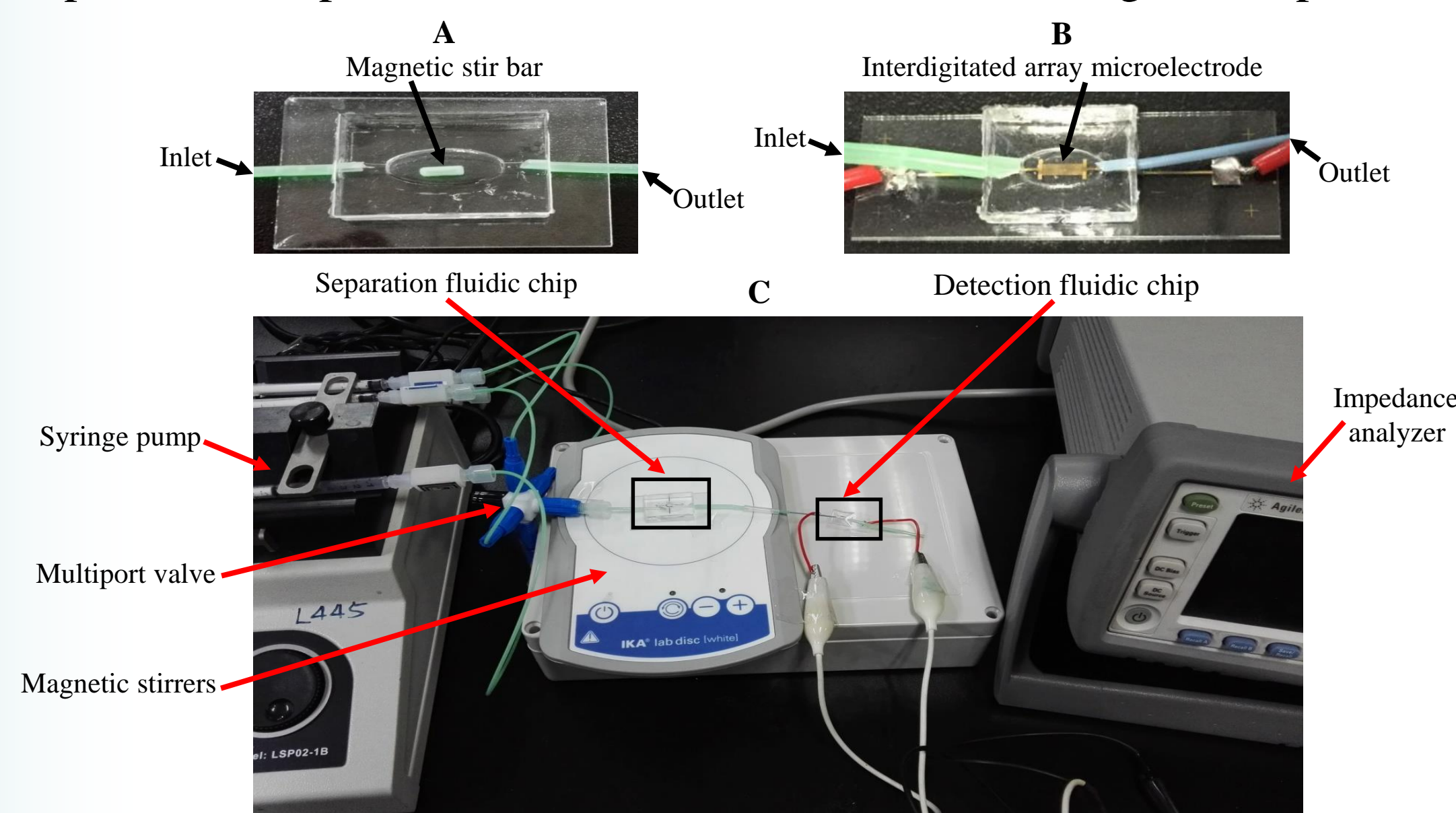


Fig.2. The proposed microfluidic biosensor for sensitive detection of foodborne pathogens.

## 3. The detection of *Listeria* in different concentrations

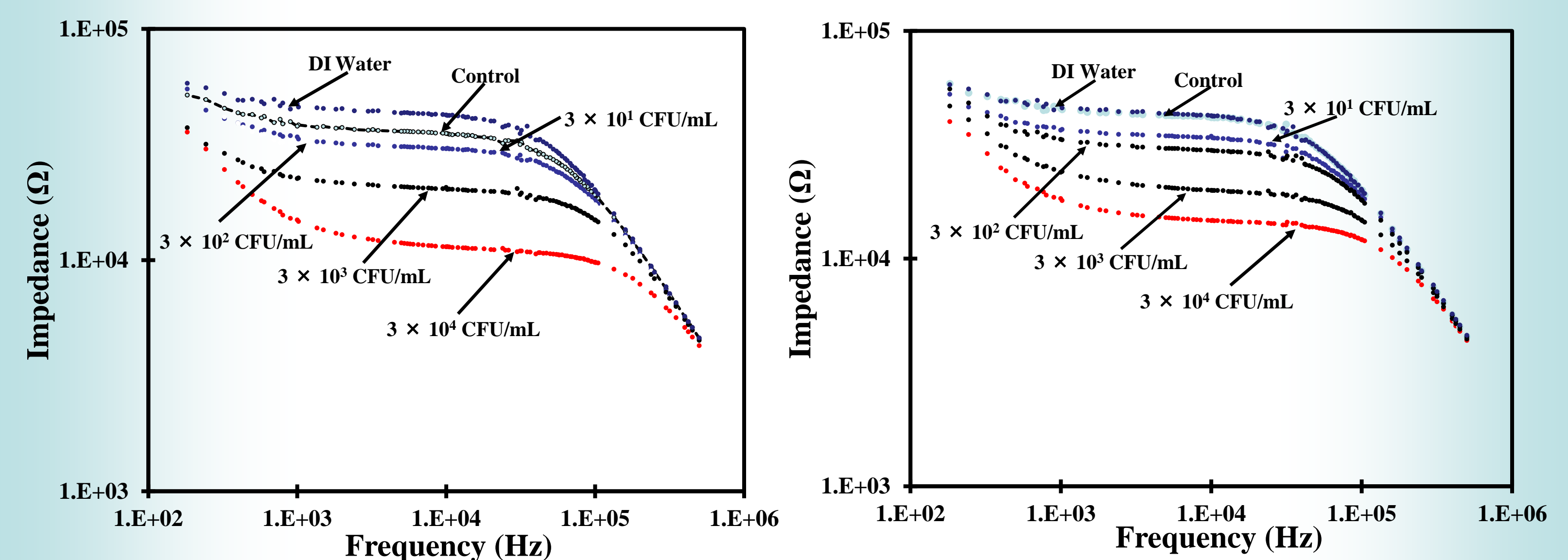


Fig. 5. (a) Bode plots of the EIS of *Listeria monocytogenes* at the concentrations of  $3.0 \times 10^1 - 3.0 \times 10^4$  CFU/mL in the pure cultures; (b) Bode plots of the EIS of *Listeria monocytogenes* at the concentrations of  $3.0 \times 10^1 - 3.0 \times 10^4$  CFU/mL in the spiked lettuce samples.

## 4. The TEM images

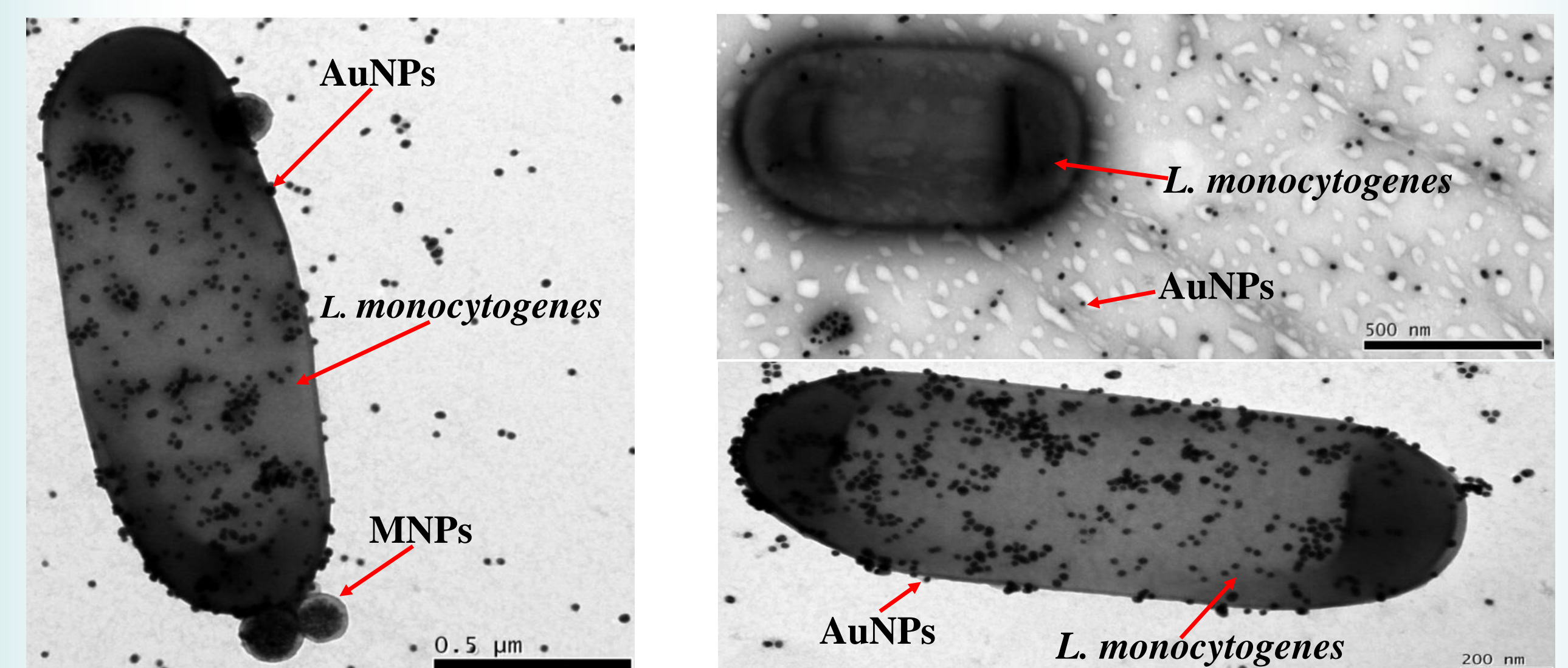


Fig.6. (a) TEM image of the MNP-*Listeria*-AuNP complex; (b) TEM image of *Listeria monocytogenes* incubated with the BSA blocked AuNPs; (c) TEM image of *Listeria monocytogenes* incubated with the PAb and urease modified AuNPs.

## 5. The detection of *Listeria* in the pure cultures and spiked lettuce samples

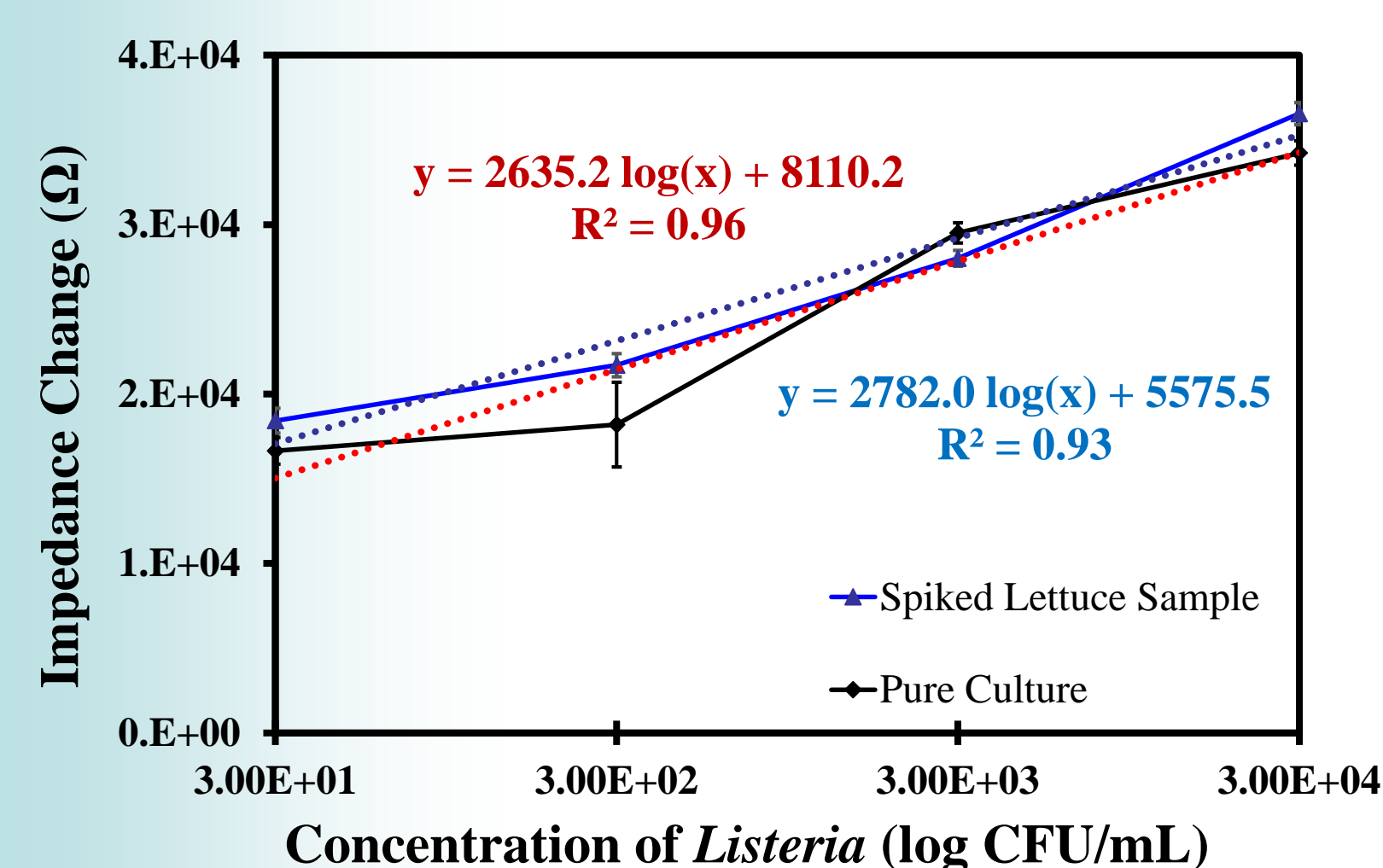


Fig.7. Linear relationship between the impedance change and the concentration of *Listeria* in the pure cultures and the spiked lettuce samples by the proposed method.

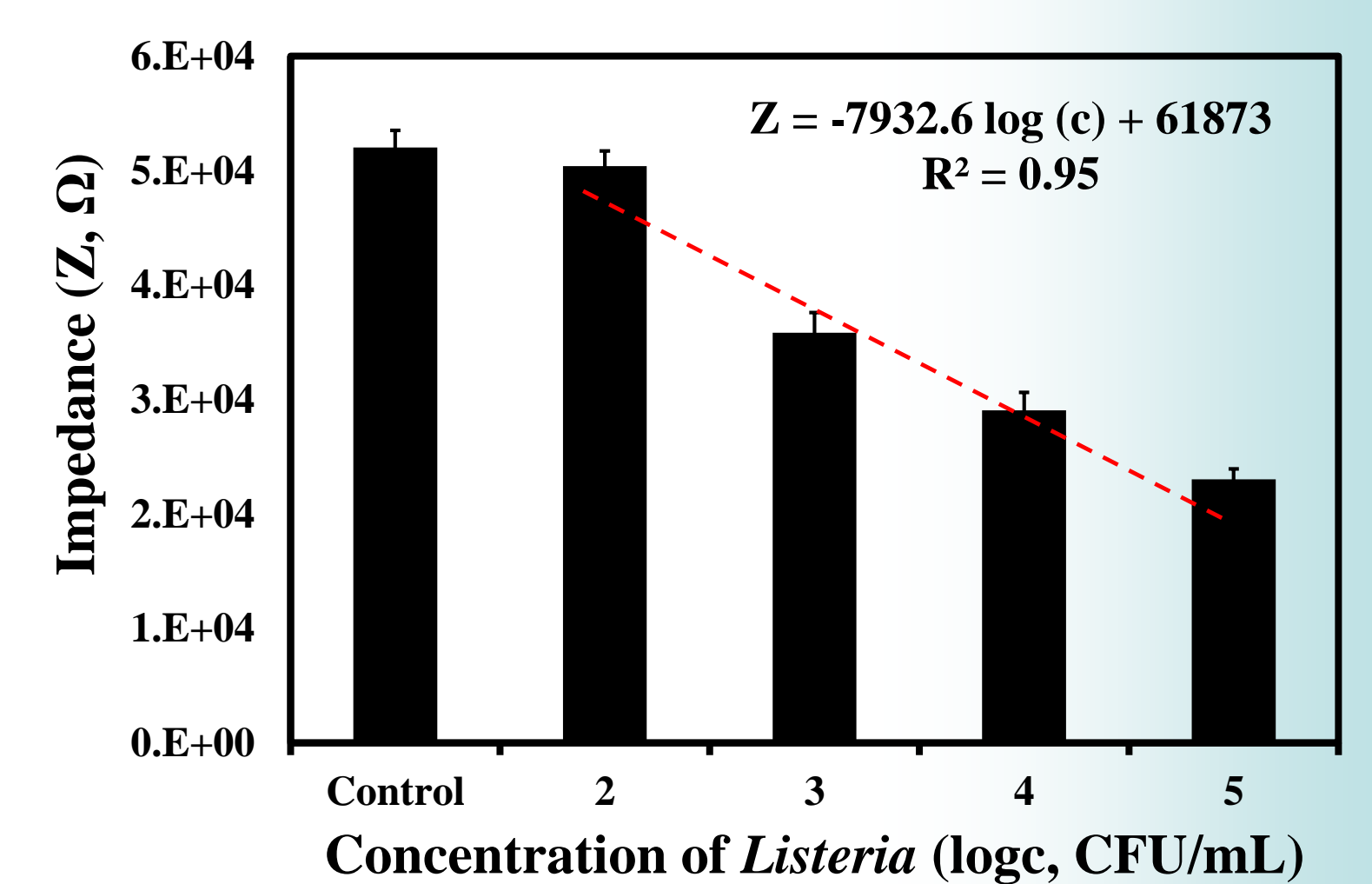


Fig.8. Linear relationship between the impedance at the characteristic frequency of 3 kHz by the proposed microfluidic biosensor.

## Results and Discussion

### 1. The immunomagnetic separation efficiency and specificity

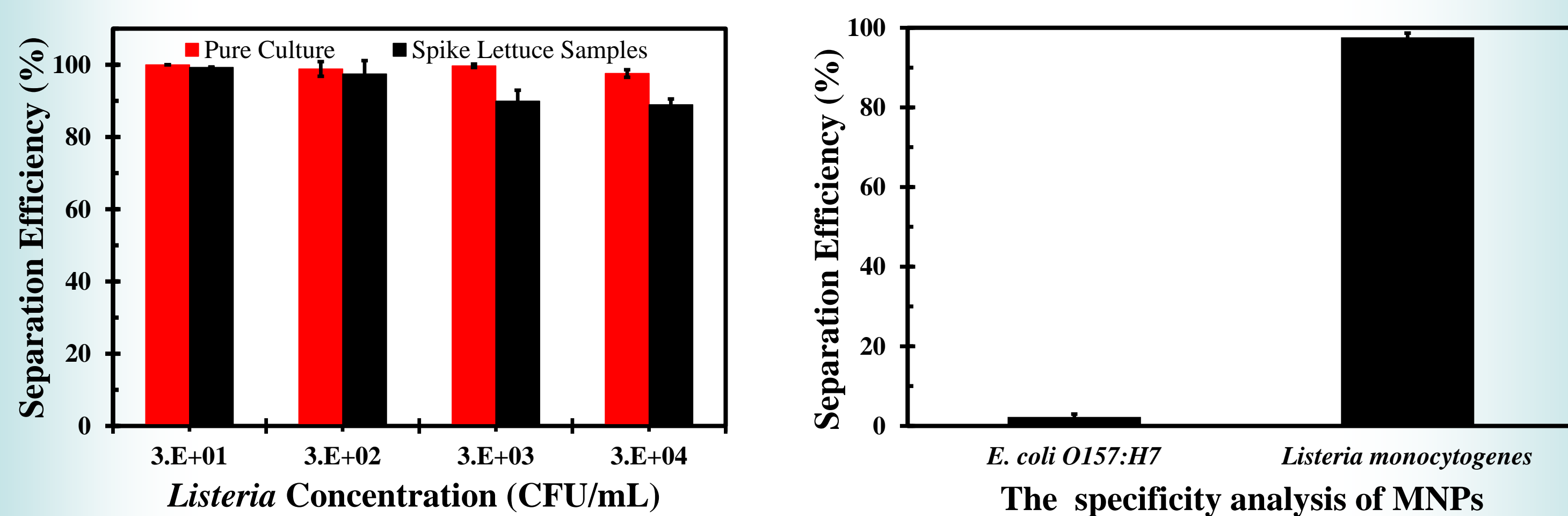


Fig.3. (a) Separation efficiency of *Listeria* at the different; (b) Separation efficiency of *Listeria monocytogenes* and *E. coli* O157:H7 at the same concentration.

### 2. The data simulation

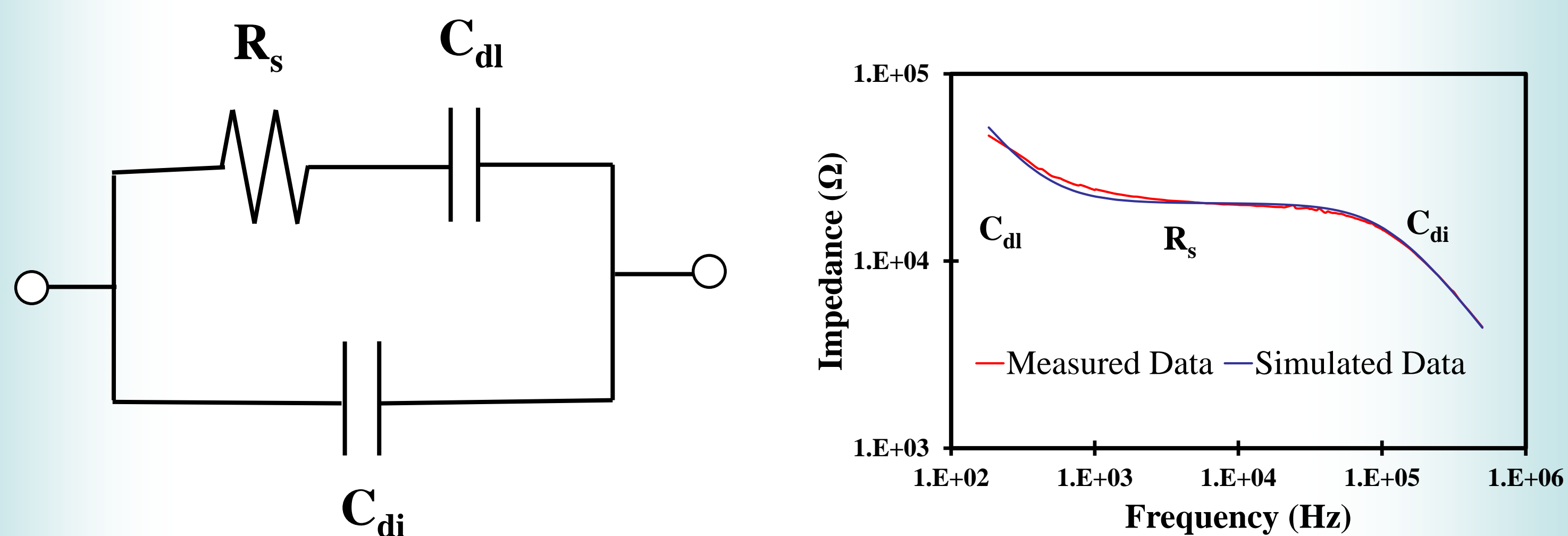


Fig. 4. (a) The equivalent circuit for fitting of the impedance data; (b) Bode plots of the impedance spectra of the measured and simulated data in the frequency range of 183 Hz to 500 kHz in the detection of  $3.0 \times 10^3$  CFU/mL of *Listeria*.

## Conclusions

This biosensor showed its potential to provide a simple, low-cost and sensitive method for rapid screening of foodborne pathogens and could be easily extended for other biological or chemical targets.

## Acknowledgments

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