Sensitivity enhancement of SERS-based immunosensors for influenza A by 2D arrays of Au@Ag coreshell nanoparticles

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1. Introduction

Surface-enhanced Raman scattering (SERS) spectroscopy has received much attention as a high sensitive detection technique of trace amount of biological and chemical samples owing to its advantages, e.g. nondestructive, no photobleaching, sensitive and fast detection. By combining SERS and immunoassay, a high sensitivity biosensor can be developed. In this study, a direct SERS-based immunoassay platform was adopted to detect influenza A by using gold core-silver shell (Au@Ag) nanoparticle (NP) two dimensional (2D) arrays as a SERS substrate, Antibody and 4,4'-thiobisbenzenethiol (TBBT) loaded AuNPs as SERS probes, and nucleoprotein as a bioanalyte. The completion of antigen-antibody reaction is confirmed by detecting the Raman signal of TBBT. As a result of the plasmon coupling between the SERS substrate and probe, the sensitivity of the immunoassay on the SERS substrate was enhanced by a factor of 4 relative to that on a flat Au film.

2. Experimental

The direct SERS-based immunoassay consists of two main components: SERS probes and a SERS substrate. The SERS probes were synthesized by labelling TBBT on the surface of 25 nm AuNPs. Then, the particles were coated with SH-PEG-COOH, followed by immobilization of influenza A antibody. The SERS substrate was fabricated by arraying 51 nm Au@Ag NP using a hybrid method [1]. Then, the SERS substrate was immersed in a mercaptohexadecanoic acid (MHDA) solution to obtain hydrophilic 2D arrays. After that, COOH at the substrate surface was activated with EDC/NHS, followed by nucleoprotein immobilization. At the end, the SERS probes were dropped and incubated on the substrate.

3. Result and discussion

To examine the sensitivity enhancement effect of the immunoassay arising from the SERS substrate, two kinds of substrates were used: Au@Ag 2D arrays and Au film substrates. The signal response of the TBBT characteristic peaks at 1065 and 1565 cm^{-1} [2] was correlated to dose of nucleoprotein for both SERS and Au substrates. As shown in Fig. 1, the calibration curves of immunoassay on the SERS and Au substrates were obtained by plotting the intensity of the 1565 cm^{-1} band as a function of the concentration of nucleoprotein. By comparing the slope of linear regression (sensitivity), we found that the immunoassay on the SERS substrate shows 4 times higher sensitivity than that on the Au film substrate. This result indicates that the enhancement field from the SERS substrate amplifies the signal of Raman reporter molecules [3].

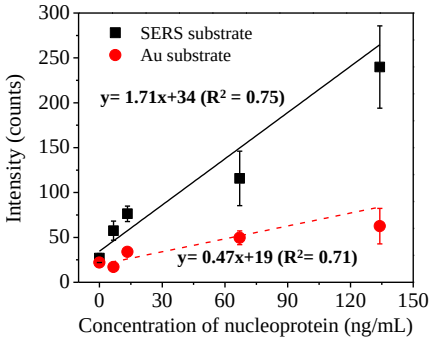

Fig.1 Calibration curves of immunoassays on the SERS and Au film substrates.

3. Conclusions

In this work, a direct SERS-based immunoassay with high sensitivity for influenza A was demonstrated using TBBT-labeled AuNPs as SERS probes and Au@Ag 2D arrays as a SERS substrate. The SERS substrate plays an important role for sensitivity enhancement through the plasmonic coupling between the SERS substrate and probe.

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