Supplementary materials

New Bi$_2$MoO$_6$ Nano-Tremellas for Promising Electrochemical Immunoassay of Carcinoembryonic Antigen

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S1. XRD pattern of Bi$_2$MoO$_6$

![XRD pattern of Bi$_2$MoO$_6$](image)

Fig. S1. XRD patterns of the as-fabricated samples Bi$_2$MoO$_6$ and pure Bi$_2$O$_3$.

S2. Optimization of experimental condition

In order to get preferable sensitivity and specificity of immunosensor, four factors

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affecting the performance of the Au@Bi$_2$MoO$_6$ NTs based immunosensor toward 1 ng/mL CEA were optimized with DPV, which included the concentration of Au@Bi$_2$MoO$_6$ NTs, pH values of PBS, the concentration of anti-CEA as well as the affinity reaction time between antibody and antigen.

Au@Bi$_2$MoO$_6$ NTs play an important role in the response of the prepared immunosensor to CEA. As seen in Fig. S2A, at the low concentration of Au@Bi$_2$MoO$_6$ NTs modified, the response signal is small, and the increasing concentration continuously increased the current till the concentration reached 2.0 mg/mL. However, when the concentration of Au@Bi$_2$MoO$_6$ NTs was higher than 2.0 mg/mL, the electron transfer rate of the immunosensor began decreasing. The current increase in the beginning could be attributed to the good conductivity and large surface area of Au@Bi$_2$MoO$_6$ NTs beneficial to the electron transfer and bimolecular modification. However, the reduced current was caused by the thickness of the material blocking the electron transfer. Therefore, the concentration of Au@Bi$_2$MoO$_6$ NTs was set as 2.0 mg/mL.

Therefore, we can know that when the pH was 6.5, the environment for the anti-CEA was the best and the maximum sensitivity of the antigen-antibody can be stimulated. The medium pH for CEA detection plays an essential role in the electrochemical performance of the designed Au@Bi$_2$MoO$_6$ NTs-based immunosensor. Five pH values (5.5, 6.0, 6.5, 7.0, and 7.5) were selected to examine the response of the immunosensor to CEA. As shown in Fig. S2B, as the pH value increased, the current went up and reached the highest value at 6.5. when the pH was higher than 6.5, the detection performance of the immunosensor was apparently influenced due to the pH-sensitive binding between Ab and CEA. Therefore, the optimal pH value was set as 6.5 for CEA detection.

Five different concentrations of anti-CEA (25, 50, 75, 100 and 125 µg/mL) were individually incubated with Au@Bi$_2$MoO$_6$ NTs modified electrode to achieve the favorable detection performance of the immunosensor toward 1.0 ng/mL CEA (Fig. S2C). It was found that the peak current greatly increased when more anti-CEA were immobilized on the electrode surface and the immunosensor exhibited the highest
response to CEA at the concentration of anti-CEA increasing to 75 µg/mL.

The incubation time of the antibody-antigen was established and the incubation time was selected to be 20, 40, 60, 80, 100 min, respectively. As can be seen from the Fig. S2D, the antigen-antibody reaction reached maximum at 60 min, and kept stable afterwards. So 60 min was chosen as the optimal incubation time between anti-CEA and CEA.

![Figures](image)

Fig. S2. Effects of (A) concentration of Au@Bi$_2$MoO$_6$ NTs, (B) pH of PBS, (C) concentration of anti-CEA and (D) antibody-antigen reaction time on reduction current responses of the immunosensor.