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Supplementary Material

A signal-on photoelectrochemical biosensor for detecting cancer marker type IV collagenase by coupling enzyme cleavage with exciton energy transfer biosensing strategy

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1. Characterization of AgNPs and Ag NPs labeled peptide

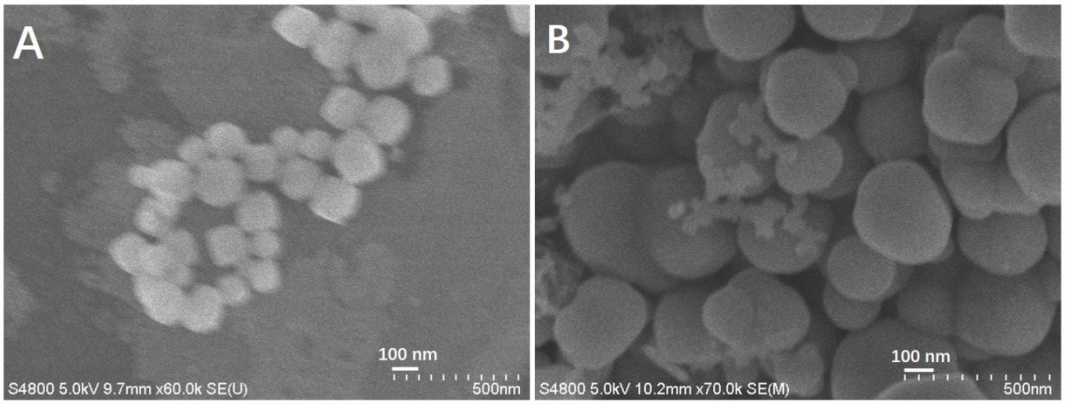

Fig.S1. Scanning electron microscopy (SEM) images of Ag NPs (A) and Ag NPs-peptide (B) nano-composites. Scale bars=500 nm.

2. Optimization of experimental conditions

To optimize the multi-layer film of CdTe QDs, different coating numbers of CdTe QDs were used to prepare ITO/$(PDDA/CdTe)_n$ electrodes, based on electrostatic attraction interaction between positively charged PDDA and MPA functionalized CdTe QDs with inverse charge.[1,2] Fig. S1A shows the effect of the number of layers (n) on the photocurrent intensity detected by PEC methods. It can be seen from the figure that the photocurrent intensity increases as the number of layers increasing, indicating that the PEC activity of the QDs depends on the number of CdTe QDs immobilized onto the electrode. However, when the number of molecular layers increased to 6, the anode photocurrent increased to the highest and then appeared a downward trend. There are two factors that affect the photocurrent intensity of the (PDDA/CdTe)n multilayer.[2,3] On one hand, as the coating number increased, the amount of photoelectrochemical active material increases, resulting in an increase in photocurrent. On the other hand, when more (PDDA/CdTe)n multilayer is assembled to their surface, the impedance effect is partially increased, which may induce a decrease in photocurrent. For n=6, the photocurrent reaches its maximum value. Therefore, we chose ITO/$(PDDA/CdTe)_6$ to build the PEC biosensor.

In the PEC experiments, the applied potential and excitation wavelength are two very important factors, which relevant to the anodic photocurrent intensity. We discussed the effect of these two factors on the photocurrent in the figure. During the experiment, the photocurrent response can be influenced by the applied potential.[4] In the range of -0.1 to -0.6 V, it can be seen from Fig.S1B that the magnitude of the current increases vs. the applied potential increases negatively. This indicates that the photogenerated electrons are effectively driven to the counter electrode by the negative potential. Meanwhile, when the applied potential at -0.5 V, the current intensity was 95% of that at -0.6 V, showing sufficient sensitivity for the PEC detection of type IV collagenase. A low constant potential is beneficial for the elimination of interference from other species in the samples. Therefore, we chose -0.5 V as the most suitable applied potential.

In the process of the illumination, the effect of the excitation wavelength on the intensity of the anode photocurrent was investigated. As shown in Fig. S1C, the photocurrent rises first and then falls in the range of 350 nm to 410 nm. This is accordance with the optical absorption properties of CdTe QDs. At 400 nm, the photocurrent intensity is about 95% of that at 380 nm. And it quickly decreases more than 400nm. Furthermore, a long wavelength means low energy consumption, which is beneficial for biological systems monitoring.[5] And the wavelength of 400nm is visible; the damage to the biomolecule is small. Therefore, 400nm as the optimum wavelength was selected in the subsequent PEC test.

Usually, the enzyme activity is relevant to the incubation temperature and time under certain

experimental conditions. The optimal temperature is usually around human body temperature (37 °C) for the enzymes in human cells.[6,7] Therefore, 37 °C was chosen as the optimal incubation temperature for the PEC biosensor. For a given enzyme concentration, the incubation time of the enzyme reaction was also necessary to determine. With increasing incubation time, enzyme activity begins to great, more and more of the free enzyme is converted into the substrate-bound complex. And then enzyme gradually saturated. Therefore, considering the sensitivity of detection, 60 minutes was used in the following experiment.

In the process of enzyme detection, the concentration of AA electrolyte and the pH value of buffer were also very important. In many previous tests studied by us and other teams, the AA concentration was optimized to be 0.1 mol·L^{-1} [1, 2, 8-10]. Therefore, we chose the optimal concentration of 0.1 mol·L^{-1} for AA in this test. For the pH of the buffer solution, we consider selecting human blood as the detection solution in the actual sample detection. Normal human blood has a pH between 7.35 and 7.45. Therefore, we selected pH=7.40 as the optimal pH for the subsequent detection.

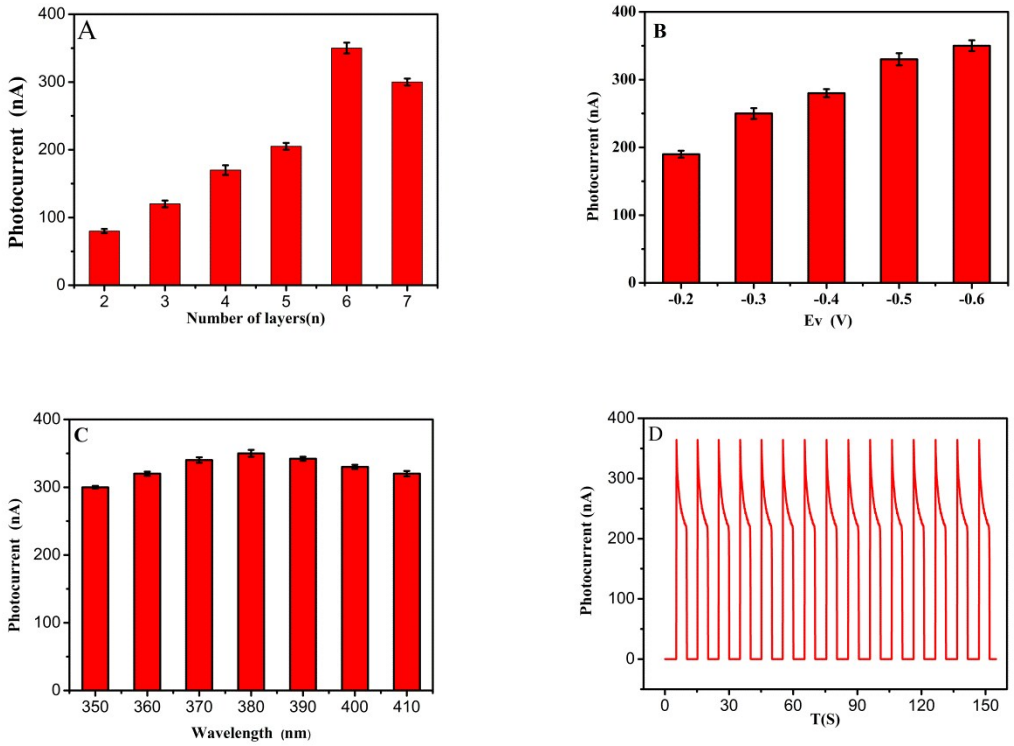

Fig.S2 Photocurrent intensity of the CdTe QDs modified ITO electrode vs. the number of bilayers (n) of (PDDA/CdTe)n multilayers (A), applied potential (B) and (C) wavelength (D) time-based photocurrent response of the sensor. The photocurrent response was measured in 0.1 M PBS (pH 7.40) containing 0.1 M AA. The applied potential was -0.5 V. The light wavelength was 400 nm.

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