Electronic Supplementary Material (ESI) for Analyst
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Label-free electrochemiluminescence immunosensor for cardiac troponin I using luminol functionalized gold nanoparticles as sensing platform

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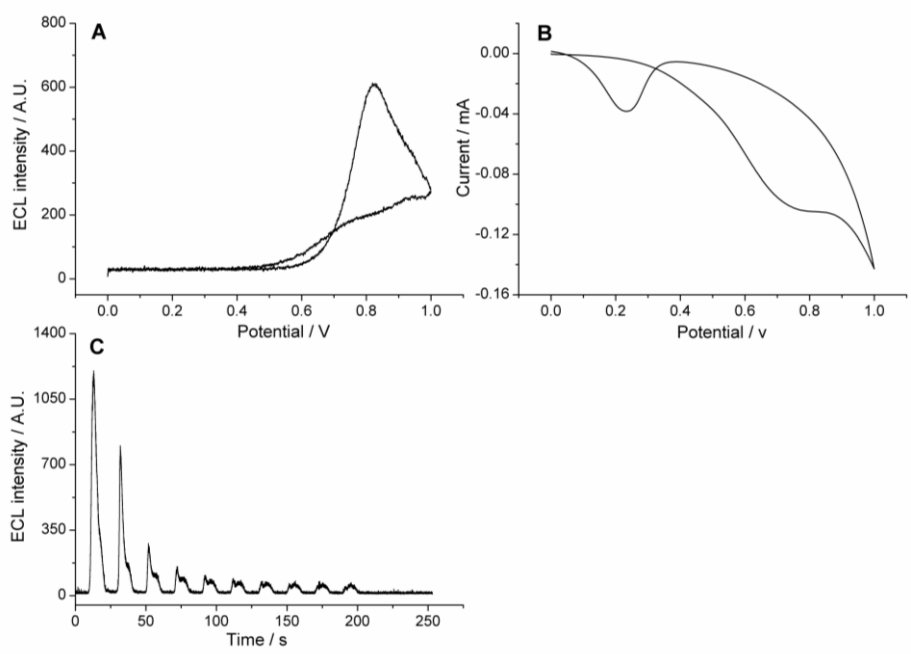

Fig. S1. Typical I_{ECL}-E curve (A) and cyclic voltammogram (B) of the immunosensor at the scan rate of 100 mV s^{-1}. (C) ECL signals of the immunosensor under CV from 0 to 1.0 V, scan rate of 100 m V s^{-1}, 10 cycles. cTnI: 10 ng mL^{-1}.

The ECL behavior of the immunosensor was studied by CV with a potential scan from 0 to 1.0 V in 0.02 M CBS containing 1.0 mM of H_2O_2. The I_{ECL}-E curve of the immunosensor is shown in Fig. S1A. And the corresponding cyclic voltammogram is shown in Fig. S1B. The ECL emission started to increase from 0.5 V and the ECL peak was found around 0.8 V (see Fig. S1A), which were related to electro-oxidation of luminol on the AuNPs according to the CV in Fig. S1B. Fig. S1C shows the ECL signals of the immunosensor under CV in 10 cycles. It can be seen that the ECL signals under CV decreases gradually in the next cycle. This phenomenon was attributed to that the diffusion layer on the surface of electrode of ECL signals generated by CV was difficult to recover in the next period, resulted in the decreasing of ECL signals.

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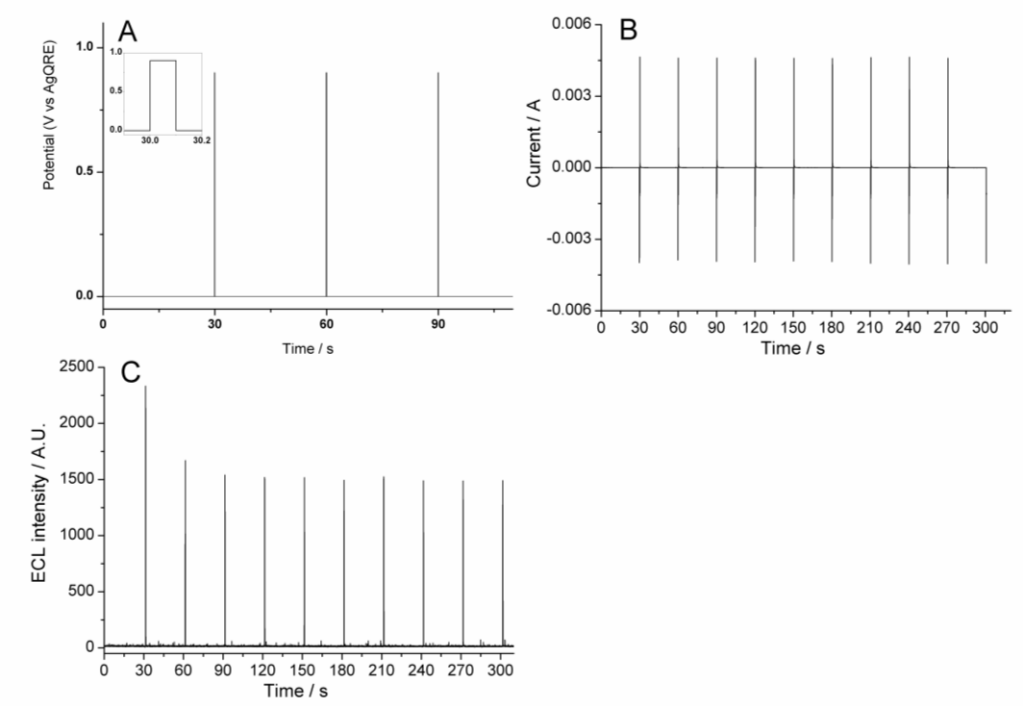

Fig. S2. (A) The potential-time curve potential and (B) the current-time curve of the double-step potential applied on the working electrode of the immunosensor. (C) ECL signals of the immunosensor under a double-step potential in 10 pulse periods. Experimental parameters: Initial potential, 0 V; pulse potential, 0.8 V; pulse period, 30 s; pulse time, 0.1 s; H_2O_2, 1.5 mM; CBS, 0.02 M (pH 10.18), cTnI: 10 ng mL^{-1}.

The ECL behavior of the immunosensor was studied with a double-step potential in the ECL working solution. Fig. S2A, B show the potential-time curve and the current-time curve of the double-step potential applied on the working electrode of the immunosensor. When a double-step potential was applied to the electrode, a pulse ECL signal was obtained. The pulse ECL intensity decreased rapidly at the first stage, then reached a stable value in ten periods in every experiment as shown Fig. S2C. The ECL signals generated by pulse potential were relatively stable when compared with those generated by CV (see Fig. S1C). Therefore, double-step potential was adopted to generate the ECL signals of the immunosensor in following experiments.

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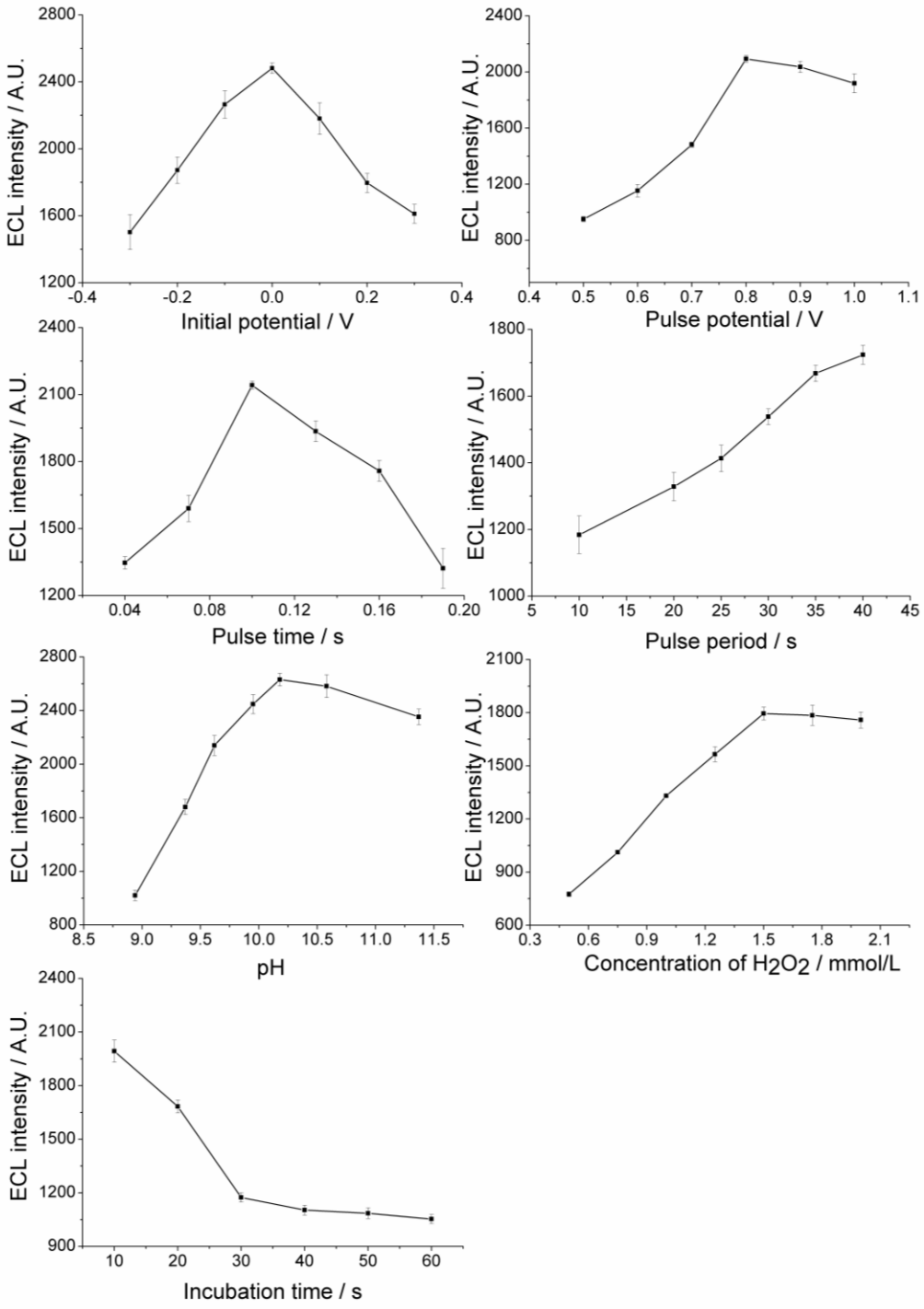

Fig. S3. Effects of (a) initial potential, (b) pulse potential, (c) pulse period, (d) pulse time, (e) pH of CBS, (f) H_2O_2 concentration and (g) incubation time on ECL intensity of the immunosensor. Initial potential, 0 V; pulse potential, 0.8 V; pulse period, 30 s; pulse time, 0.1 s; H2O2, 1.5 mmol/L; CBS, 0.02 M (pH 10.18), cTnI: 10 ng mL^{-1}.

The effect of the initial pulse potential in the range of –0.3 – 0.3 V was investigated (Fig. S3a). When the initial pulse potential was 0 V, a maximal ECL

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intensity was achieved. This is probably due to the initial pulse potential of 0 V has a better diffusion controlled reaction on the surface of the modified electrode. Therefore, an initial potential of 0 V was used in the following experiments. The effect of the pulse potential in the range 0.5 – 1.0 V was studied (Fig. S3b). The ECL intensity increased with the increase of the pulse potential since the electro-oxidation of luminol was much faster at higher electrode potential. However, if a high potential above 0.8 V was used, the stability and reproducibility of the modified electrode declined. This might due to the electro-oxidation of AuNPs on the surface of electrode and the break of Au-S bond between the 1,3-propanedithiol molecules and the gold electrode[1]. Thus, a pulse potential of 0.8 V was adopted. The effect of the pulse time on the ECL intensity was examined in the range of 0.04 – 0.19 s (Fig. S3c). The ECL intensity increased with the pulse time. However, when the pulse time was longer than 0.1 s, the ECL intensity decreased rapidly at the second period, and could not reach a stable ECL signal in the following period. This was due to that the diffusion layer on the surface of electrode became thicker in a relatively long pulse time, and was difficult to recover in the next pulse. Therefore, a pulse time of 0.1 s was selected. The effect of the pulse period on the ECL intensity was investigated in the range of 10 – 40 s (Fig. S3d). The ECL intensity increased with the increase of the pulse period, which might be due to more effective diffusion of H_2O_2 in a longer pulse period. To obtain shorter analytical time and higher ECL intensity, a pulse period of 30 s was adopted in the following experiments. The effect of pH on the ECL intensity was examined in the range of 8.9 – 11.4 in 0.02 M CBS. Fig. S3e shows the pH

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dependence of the ECL intensity. The maximal ECL intensity was obtained at pH 10.18. Fig. S3f shows the change of ECL intensity with the concentration of H_2O_2. The ECL intensity increased with the increase of the concentration of H_2O_2 and reached maximum at 1.5 mM. This trend might be caused by of the co-oxidation function of H_2O_2. Therefore, 1.5 mM H_2O_2 was selected in the following experiments. The effect of incubation time on ECL intensity was also investigated. As shown in Fig. S3g, ECL intensity decreased with the increasing of incubation time and trended to level off after 40 min, and longer incubation time could not cause obvious signal change. So, 40 min of incubation time was used for the detection of cTnI in this study.

References

1 X. B. Yin, B. Qi, X. P. Sun, X. R. Yang and E. K. Wang, *Anal. Chem.* 2005, **77**, 3525-3530.