Electronic Supplementary Material (ESI) for Analyst.
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Supporting Information:

Label-free analytical performances of peptide-based QCM biosensor for trypsin

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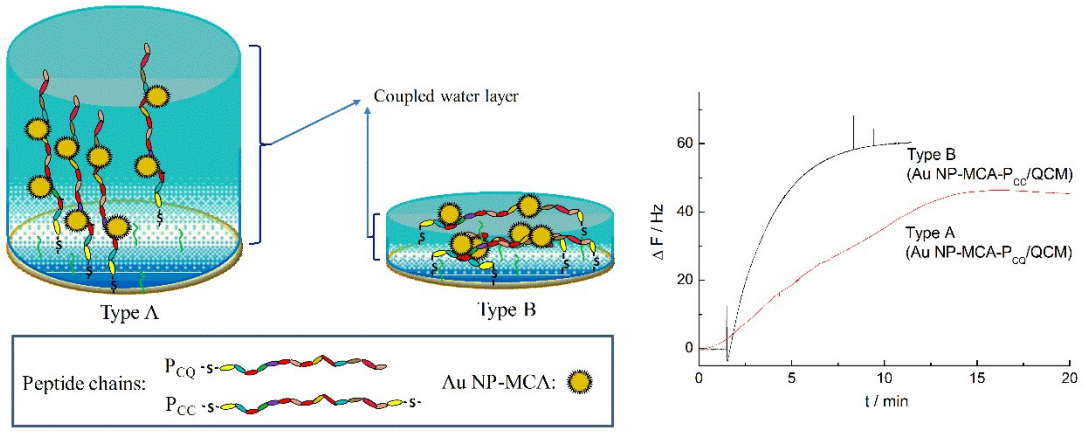

Fig.S1. The schematic of left shows the sensing interface with coupled water layer under the peptide different immobilizing methods (A: single-end immobilized P_{CQ}; B: double-end immobilized P_{CC}), the figure on the right shows the signal responses of the two sensors to the trypsin of 80 ng moL^{-1}.

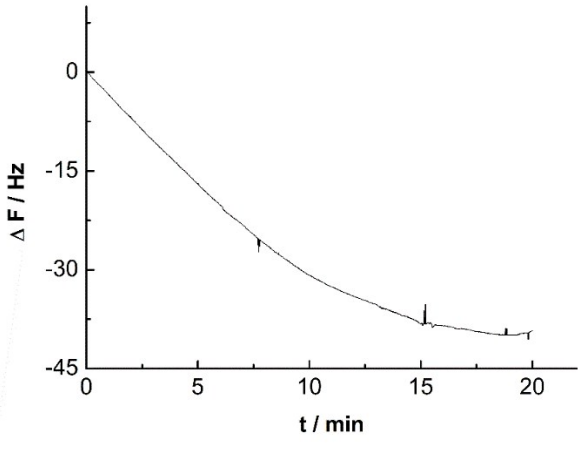

Fig.S2. Frequency response of QCM electrode for the immobilization of P_{CC}.

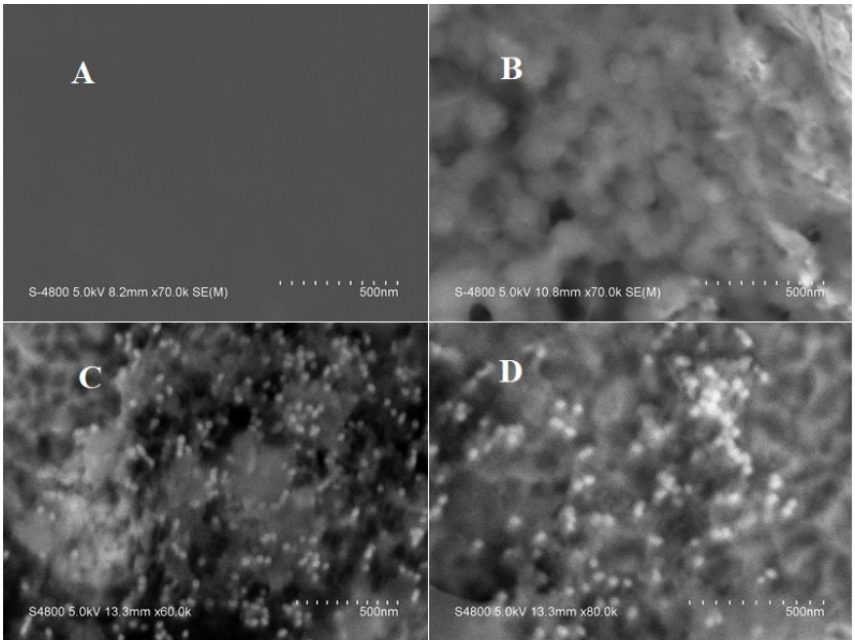

Fig.S3. SEM images of the unmodified QCM chip (A), P_{CC}/QCM chip (B), Au NPs-MCA-PCC/QCM chip (C), Au NPs-MCA-PCC/QCM chip in 60 ng·mL^{-1} trypsin solution after 600 s, the measure temperature was 37 ℃.

To investigate the fabrication of Au NPs-MCA-P_{CC}/QCM biosensor and the process of trypsin detection, SEM images were also taken for observing the surface change of bare QCM, P_{CC}/QCM, Au NPs-MCA-P_{CC}/QCM and Au NPs-MCA-P_{CC}/QCM. Compared with bare QCM (Fig. S3A), a flocculated molecular layer was observed (Fig.S3B), which revealed the successful immobilization of P_{CC} into QCM electrode. The coupling of Au NPs with P_{CC} is an important procedure, which plays an important role on the signal amplification for trypsin detection. As can be seen from Fig.S3C, individual Au NPs (white dots) were observed after the binding of Au NPs with P_{CC} on the QCM electrode. Moreover, the performances of Au NPs-MCA-P_{CC}-QCM for trypsin detection were also investigated using SEM. a trypsin solution (60 ng mL^{-1}) were tested and

the corresponding morphologies were represented in Fig.S3D, respectively. The injection of trypsin resulted the decrease of Au NPs from sensor surface, suggesting correlation between the cleavage rates of P_{CC} and the concentrations of trypsin.

In addition, the Au NPs were prepared according to a previously reported method that the chlorauric acid was reduced by citric acid (*Anal. Biochem.*, 2010, 397, 212). The Au NPs would be carried negative charges. However, we found that gold nanoparticles without functionalization could bind to the P_{CC} well at pH 9.0, but the trypsin activity would be inhibited when pH was too high. At a pH of 7.4, MCA-functionalized Au NPs combine best with P_{CC}, with the sensing signals of 3-5 times greater than those without using functionalized Au NPs. Therefore, we use MCA functionalized Au NPs as the signal amplification factor in our work.

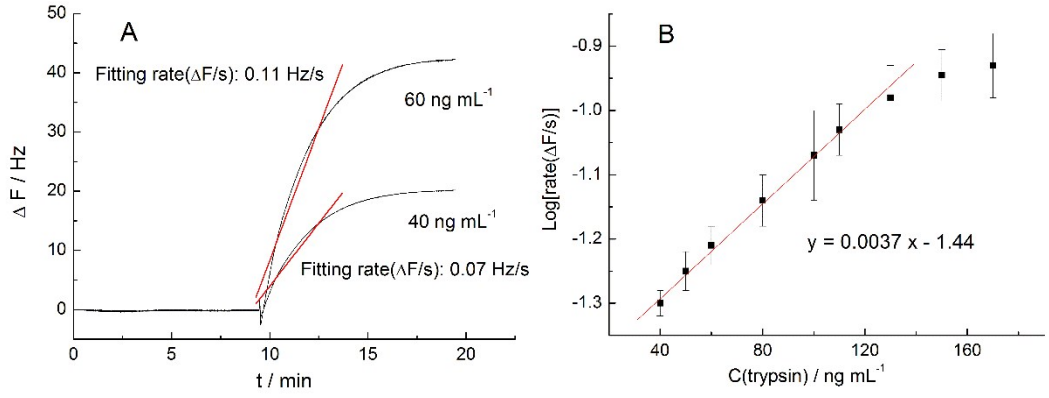

Fig.S4. The responses of the Au NPs-P_{CC}/QCM biosensor to different concentrations of trypsin from 40 ng·mL^{-1} to 60 ng·mL^{-1}. The red lines are the fitting values of frequency change within 5 minutes after injection (A). The linear relationship between the logarithm of rate of ΔF (log[rate(ΔF/s)]) and the concentration of trypsin in the range of 40-170 ng·mL^{-1}. The error bars

represent the standard deviation (S.D.) of three measurements.

Fig. S4 shows the logarithm of rate of ΔF (log[rate(ΔF/s)]) as a function of the concentration of trypsin (range from 40 to 170 ng·mL^{-1}). A linear relationship between the log[rate(ΔF/s)] and the concentration of trypsin was obtained in the range from 40 to 130 ng mL^{-1}. The linear relationship can be described as $y = -1.44 + 0.0037x$ with a correlation coefficient $r^2 = 0.989$, where y is the logarithm of rate of ΔF (log[rate(ΔF/s)]) and x is the concentration of trypsin.

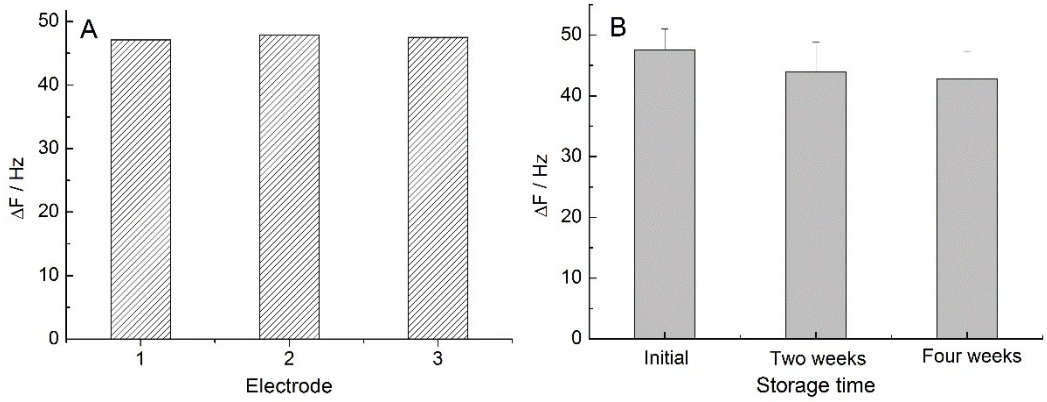

Fig.S5. (A) Repeatability and (B) storage stability of the Au NPs-MCA-P_{CC}/QCM biosensor against the determination of trypsin (60 ng mL^{-1}).