Supporting Information

Poly(β-cyclodextrin) enhanced fluorescence coupled with specific reaction for amplified detection of GSH and trypsin activity

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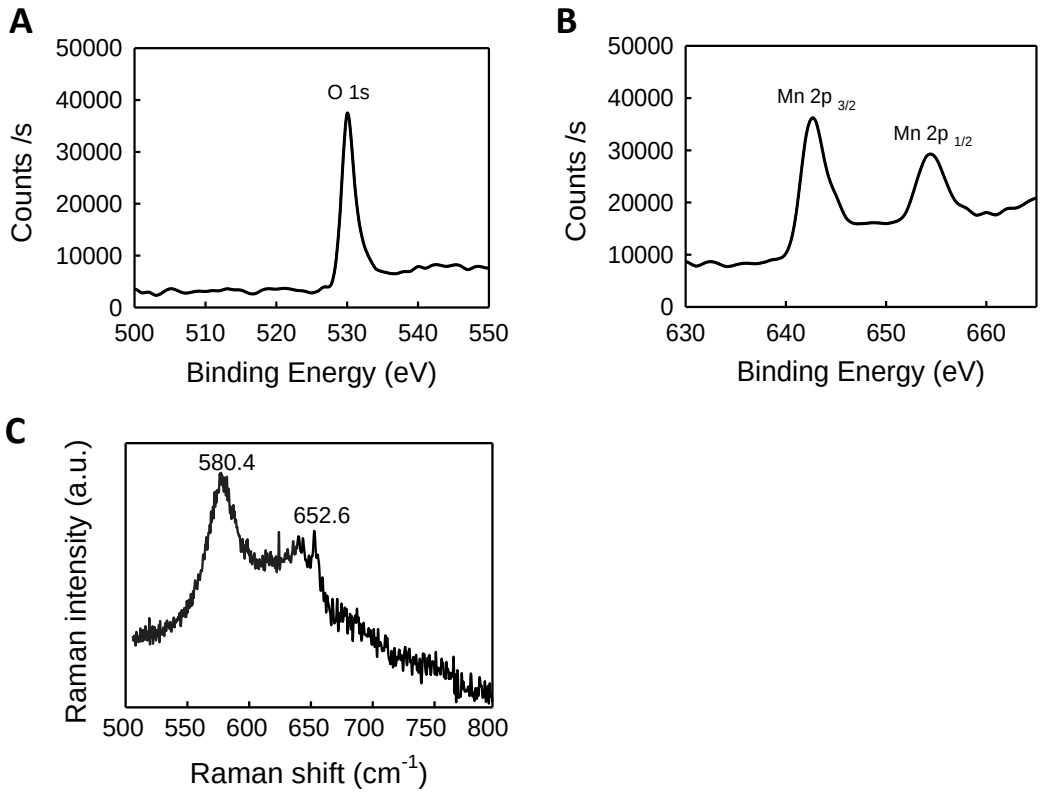

Fig. S1 The high-resolution XPS spectra of the O $_{1s}$ (A), Mn $_{2p3/2}$ and Mn $_{2p1/2}$ (B) peaks of MnO_2 nanotubes. (C) Raman spectrum of MnO_2 nanotubes.

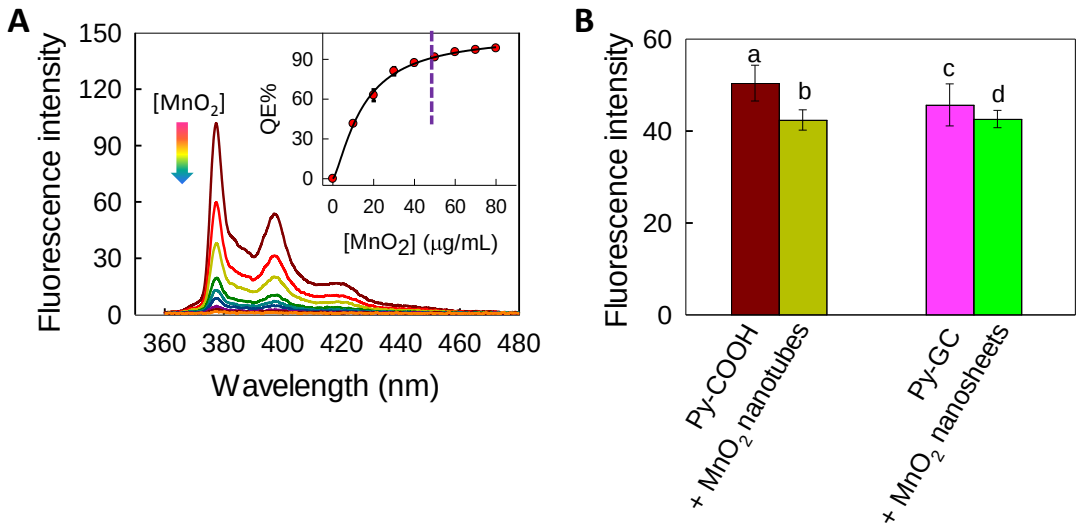

Fig. S2 (A) FL quenching curves of Py-GC (1 μM) by MnO_2 nanotubes with excitation at 345 nm. Inset: FL quenching efficiency (QE %) as a function of the Py-GC concentration. [MnO_2] = 0, 10, 20, 30, 40, 50, 60, 70, and 80 μg/mL. (B) FL intensity changes of Py-COOH without (a) and with (b) MnO_2 nanotubes, and (c-d) validation of the electrostatic interactions. [Py-COOH] = 1.0 μM; [MnO_2] = 50 μg/mL; [Py-GC]= 1.0 μM; [MnO_2 nanosheets] = 50 μg/mL.

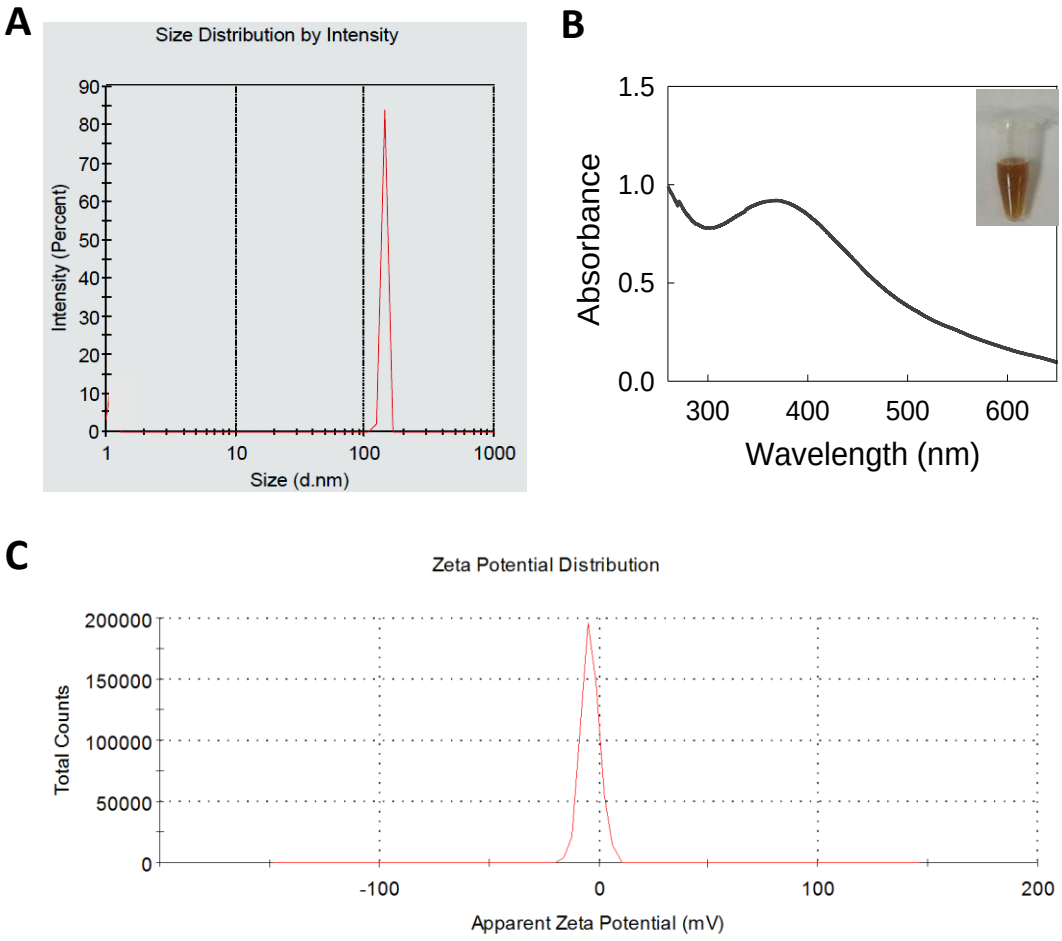

Fig. S3 (A) DLS data and (C) Zeta potential of MnO_2 nanosheets (-3.82 mV). (B) UV-vis absorption spectrum and photograph (inset) of MnO_2 nanosheets.

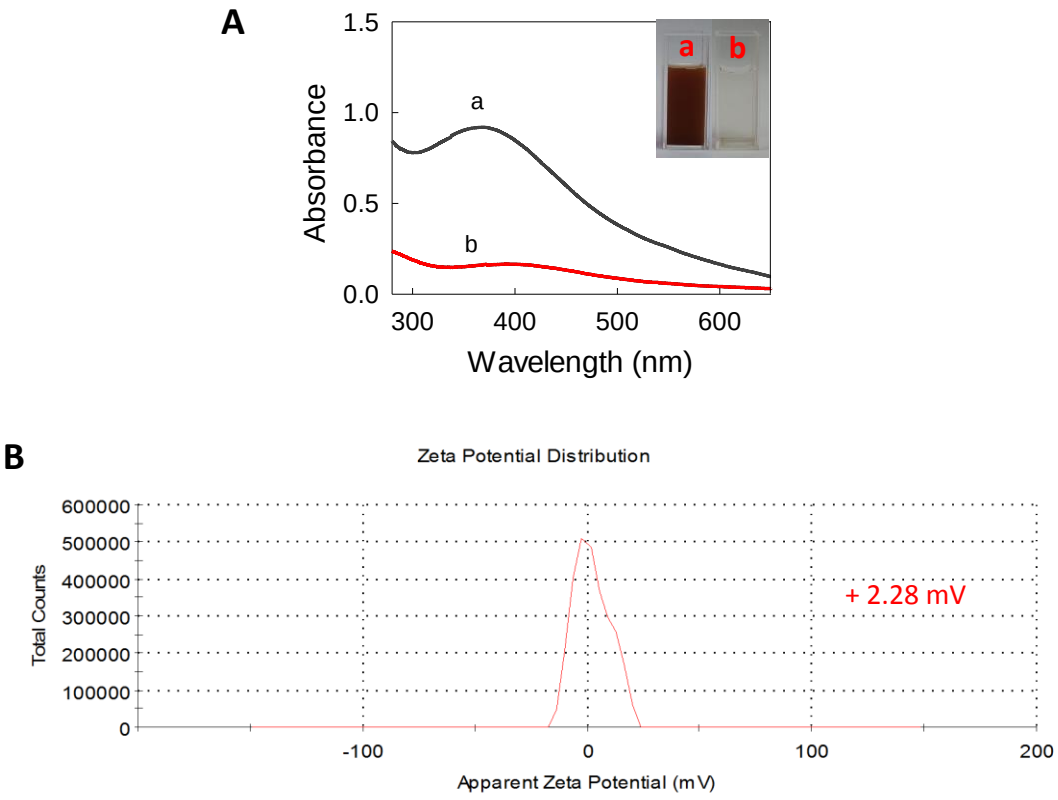

Fig. S4 (A) UV-vis absorption spectra and photographs (inset) of MnO_2 nanosheets in the absence (a) and presence (b) of GSH, respectively. (B) Zeta potential of MnO_2 nanotubes with GSH added.

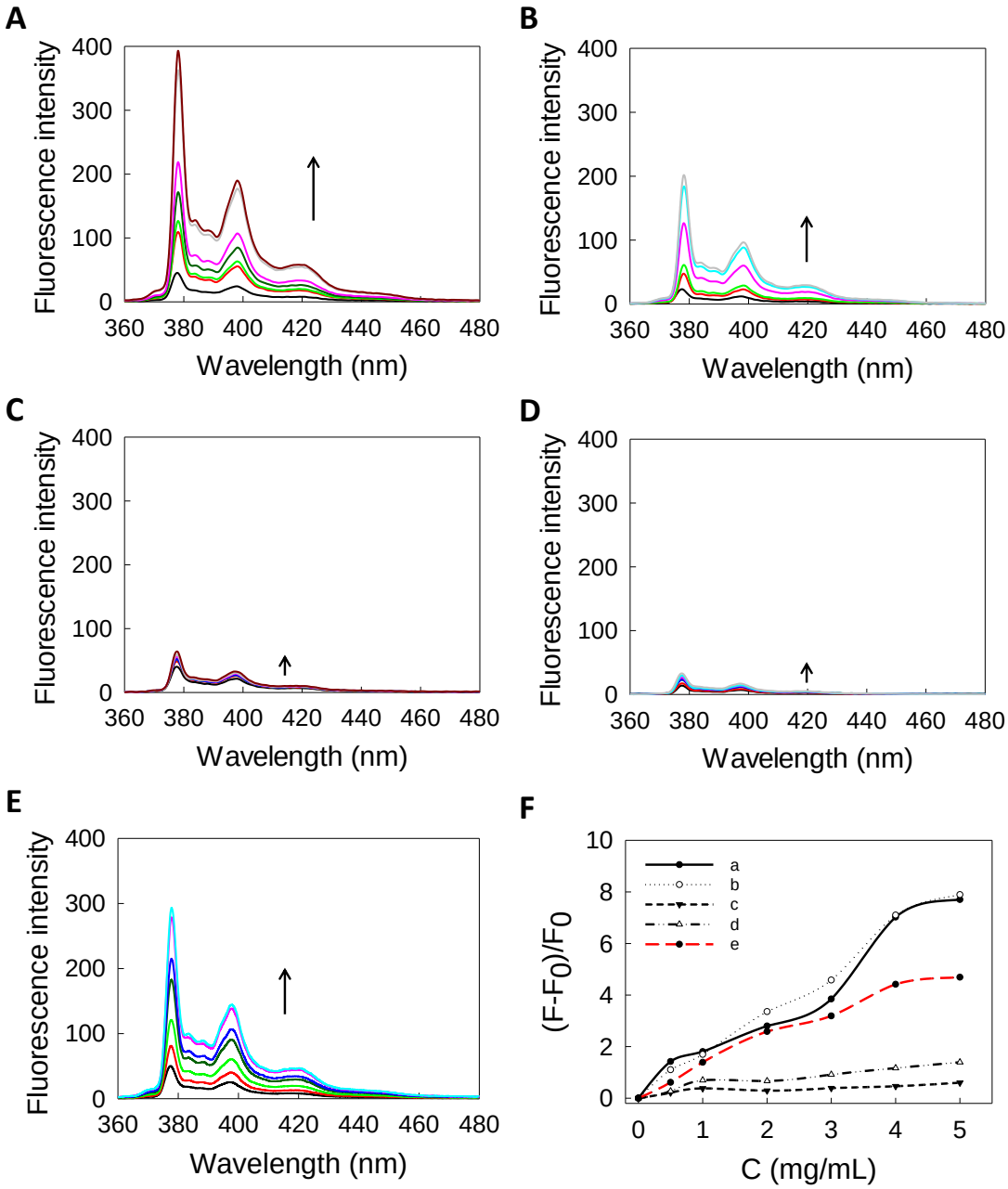

Fig. S5 Effect of different concentrations of β-CDP (or β-CD) (from bottom to up: 0, 0.5, 1.0, 2.0, 3.0, 4.0, and 5.0 mg/mL, respectively) on the fluorescence emission of Py-GC (or Py-COOH) in Tirs-HCl solution (A-D) and in biological fluids (E). A. Py-GC + β-CDP; B. Py-COOH + β-CDP; (C) Py-COOH + β-CD; (D) Py-COOH + β-CD. (F) Fluorescence enhancement as a function of the β-CDP (or β-CD) concentration obtained from A-E. F_0 and F are the fluorescence intensity of the sensing system at 378 nm in the absence and presence of β-CDP, respectively. [Py-GC] = 1 μM, [Py-COOH] = 1 μM. λex = 345 nm.

As a critical factor for the electrostatic interaction between Py-GC and Py-GC/MnO_2 nanoprobe, the effect of pH was first optimized (Figure S6A). Interestingly, in Tirs-HCl buffer solutions at a pH range from 7.0 to 9.5, the fluorescence intensity changes ($(F-F_0)/F_0$) of detection system in the present of β-CDP upon addition of GSH. It is worth noting that the fluorescence intensity of nanoprobe has scarcely affected by pH change, accordingly, 7.4 is adopted as the optimal pH value for the next sensing experiments. Owing to the possibility of β-CDP concentrations causing the fluorescence enhancement of Py-GC and thus influencing the signal amplify response of our constructed nanoprobe by adding GSH, we next investigated the effect of β-CDP concentrations in the sensing system. Figure S6B illustrates the increase of nanoprobe fluorescence intensity changes at 378 nm with increased β-CDP concentrations at the present of GSH, and 5 mg/mL is adopted as the optimized concentration.

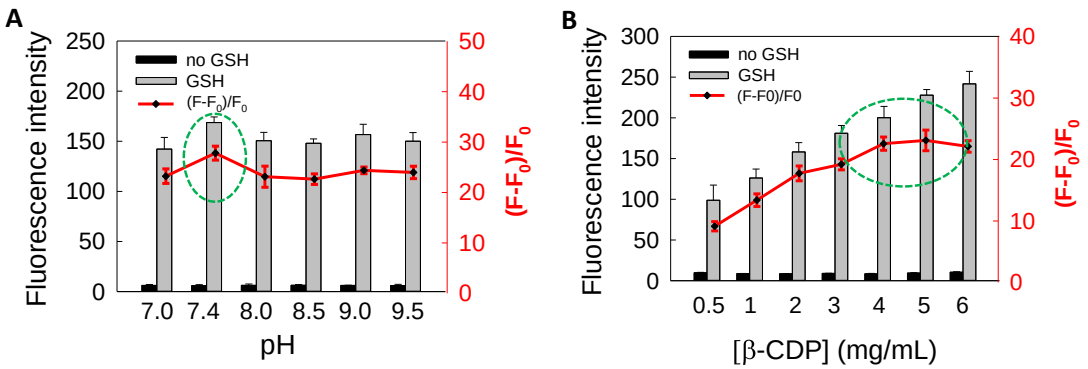

Fig. S6 Effects of pH (A) and β-CDP concentrations (B) on the fluorescence responses to different concentrations of GSH. [Py-GC] = 1 μM, [MnO_2] = 50 μg/mL; [GSH] = 200 μM; [β-CDP] = 0.5-6.0 mg/mL; pH = 7.0-9.5; $\lambda_{ex}/\lambda_{em}$ = 345 nm/378 nm.

Table S1. Comparison with some reported methods for the detection of GSH

Probe	Linear range	LOD	Reference
CdTe QDs-Hg^{2+} system	0.6-20 μM	0.1 μM	Analyst, 2012, 137, 924-931
GQDs/MnO_2 nanosheets	0.5-10 μM	0.15 μM	ACS Appl. Mater. Interfaces, 2016, 8 21990-21996
g-C_3N_4/MnO_2 nanocomposites	0-2000 μM	0.2 μM	Anal. Chem. 2014, 86, 3426-3434
MnO_2 NPs/TMB	0.26-26 mM	0.1 mM	J. Am. Chem. Soc., 2012, 134, 18928-18931
Py-GC/MnO_2 nanotubes	2-100 μM	0.06 μM	This work

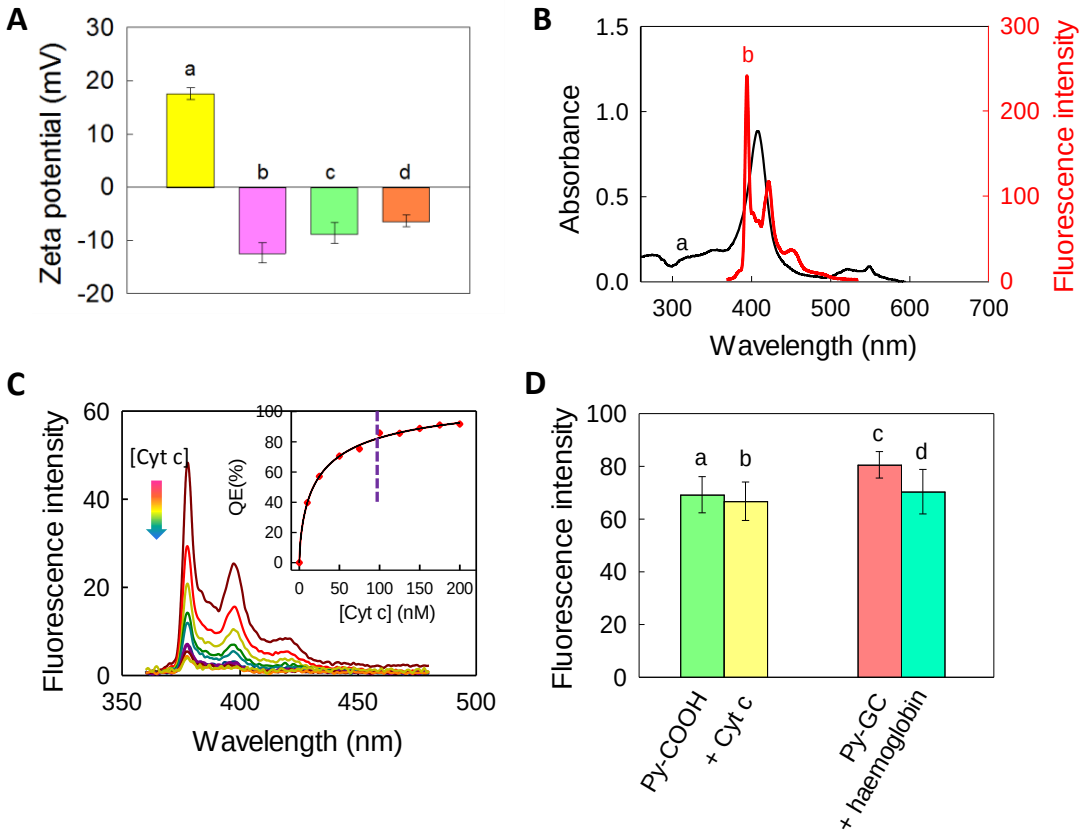

Fig. S7 Zeta potentials obtained in assays of trypsin activity by the Py-GC/Cyt c system under different conditions: (a) Cyt c (+17.6 mV), (b) Py-GC/Cyt c (-12.3 mV), (c) Py-GC/Cyt c + trypsin, (d) Py-GC/Cyt c + trypsin+ β-CDP. (B) UV-vis absorption spectrum of Cyt c (a) and emission spectrum of Py-GC (b), respectively. (C) FL quenching curve of Py-GC (1 μM) by Cyt c with excitation at 345 nm in 20 mM Tris-HCl buffer (pH 8.5). Inset: fluorescence quenching efficiency (QE %) as a function of the Py-GC concentration. [Cyt c] = 0, 10, 25, 50, 75, 100, 125, 150, 175, and 200 nM. (D) FL intensity changes of Py-COOH without (a) and with (b) Cyt c, and (c-d) validation of the electrostatic interactions. [Cyt c] = 100 nM; [Py-COOH] = 1.0 μM; [Py-GC] = 1.0 μM; [haemoglobin] = 100 nM.

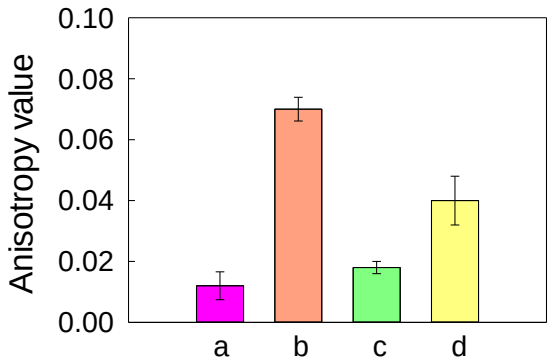

Fig. S8 FL anisotropy values at 378 nm obtained in assays trypsin activity by Py-GC/Cyt c system under different conditions: (a) Py-GC, (b) Py-GC/Cyt c, (c) Py-GC/Cyt c + trypsin, (d) Py-GC/Cyt c + trypsin + β-CDP. [Py-GC] = 1 μM, [Cyt c] = 100 nM; [trypsin] = 500 ng/mL.

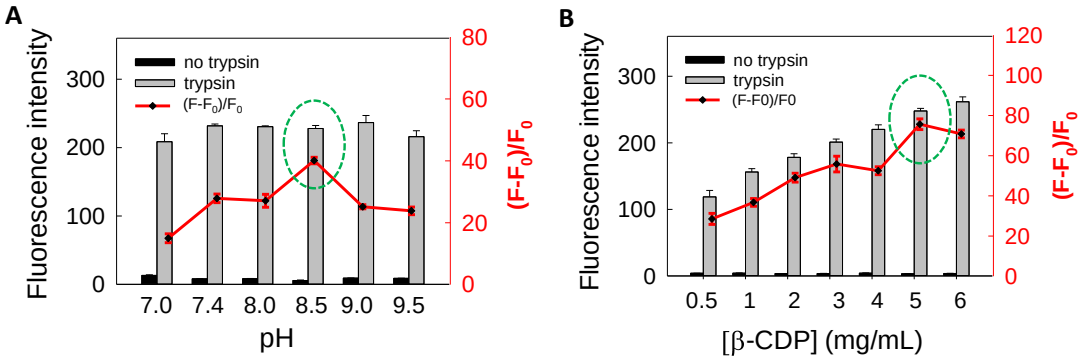

Fig. S9 Effects of pH (A) and β-CDP concentrations (B) on the fluorescence responses to different concentrations of trypsin. [Py-GC] = 1 μM, [MnO_2] = 50 μg/mL; [Cyt c] = 100 nM; [GSH] = 200 μM; [trypsin] = 500 ng/mL; [β-CDP] = 0.5-6.0 mg/mL; pH=7.0-9.5; λex /λem = 345 nm/378 nm.

Table S2. Determination of the trypsin in serum samples using Py-GC/Cyt c probe

Probe	Linear range	LOD	Reference
PFP-CO_2Na/Cyt c	0-40 nM	1.7 nM	Anal. Chem. 2010, 82, 8604-8610
TPE/Arg_6	0-8.0 μg/mL	0.2 μg/mL	Org. Lett. 2010, 12, 2274-2277
Peptide-CdSe/ZnS	0-500 μg/mL	0.1 μg/mL	Anal. Chem. 2007, 79, 208-214
Py-GC/Cyt c	0-2.5 μg/mL	0.04 μg/mL	This work