Simple fluorescence assay for trypsin through protamine-induced carbon quantum dots quenching aggregation platform

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Fig. S1. Fluorescence spectra of CQDs under different exciting wavelengths



Fig. S2. Variable fluorescence intensities of CQDs in NaCl with different concentrations. Error bars represents the SD of three parallel determinations.



Fig. S3. Variable fluorescence intensities of CQDs in different pH conditions. Error bars represents the SD of three parallel determinations.



Fig. S4. Variable fluorescence intensities of CQDs under the excitation of 345 nm at continuous 1 hour



Fig. S5. High-resolution XPS spectra of N1s (A), S2p (B), and C1s (C).



Fig. S6. Fluorescence image of CQDs, CQDs/Pro and the addition of Try into CQDs/Pro under a 365 nm UV lamp.



Fig. S7. Fluorescence spectra of CQDs without and with the presence of Try.



Fig. S8. The variable fluorescence intensities (A) or fluorescence spectra (B) of CQDs with the addition of 40 μ g/mL Pro in different period (A) and different contents (B).



Fig. S9. Curve of fluorescence quenching efficiency with different Pro concentration to 20 μ g/mL CQDs. Error bars represents the SD of three parallel determinations.



Fig. S10. The fluorescence recovery ($\Delta F=F-F_0$) of CQDs/Pro towards 500 ng/mL Try at different pH. Error bars represents the SD of three parallel determinations.



Fig. S11. The fluorescence recovery ($\Delta F=F-F_0$) of CQDs/Pro towards 500 ng/mL Try at different temperatures. Error bars represents the SD of three parallel determinations.



Fig. S12. The fluorescence recovery ($\Delta F=F-F_0$) of CQDs/Pro towards 500 ng/mL Try at different reacting time. Error bars represents the SD of three parallel determinations.

Detection method or Material	LOD	Linear range	reference
	(ng/mL)	(ng/mL)	
Mn-doped ZnSe quantum dots	40	100-12000	[S1]
Surface imprinting strategies	100	Not mentioned	[S2]
Gold nanoclusters	9	10-50000	[S3]
Redox cycling in the presence of L-ascorbic acid	100	100-1000	[S4]
Self-Powered Sensor for Naked-Eye	500	Not mentioned	[S5]
Silver nanoparticles	2	2.5-200	[S6]
DNA hosted Cu nanoclusters	0.0048	0.1-1000	[S7]
BSA-stabilized gold nanoclusters	2	10-10000	[S8]
Silver triangular nanoplates	1.8	5-80	[S9]
CQDs/pro	8.08	25-500	this work

Table S1: Comparison of different methods for determination of Try

Samples	Added	Found	Recovery	RSD
	(ng/mL)	(ng/mL)	(%)	(n=3, %)
1	25.00	27.25	109.04	2.90
2	50.00	50.01	100.02	2.39
3	250.00	247.68	99.07	1.55
4	500.00	517.39	103.47	1.42

Table S2: Detection of spiked Try in serum samples

Reference

[S1] X. Gao, G. Tang, Y. Li and X. Su, Anal Chim Acta, 2012, 743, 131-136.

[S2] O. Hayden, C. Haderspock, S. Krassnig, X. Chen and F. L. Dickert, Analyst, 2006, 131, 1044-1050.

[S3] X. You, Y. Li, B. Li and J. Ma, Talanta, 2016, 147, 63-68.

[S4] S. Park and H. Yang, Analyst, 2014, 139, 4051-4055.

[S5] B. A. Zaccheo and R. M. Crooks, Analytical Chemistry, 2011, 83, 1185-1188.

[S6] P. Miao, T. Liu, X. Li, L. Ning, J. Yin and K. Han, Biosens Bioelectron, 2013, 49, 20-24.

[S7] L. Wang, F. Shi, Y. Li and X. Su, Sensors and Actuators B: Chemical, 2016, 222, 945-951.

[S8] L. Hu, S. Han, S. Parveen, Y. Yuan, L. Zhang and G. Xu, Biosensors and Bioelectronics, 2012, 32, 297-299.

[S9] H. Chen, A. Fang, Y. Zhang and S. Yao, Talanta, 2017, 174, 148-155.