## **Supporting Information**

Ultrasensitive surface-enhanced Raman scattering detection of trypsin based on anti-aggregation of 4-mercaptopyridine–functionalized silver nanoparticles: an optical sensing platform toward proteases

Lingxin Chen,<sup>\*a</sup> Xiuli Fu,<sup>a,b</sup> and Jinhua Li<sup>a</sup>

<sup>a</sup> Key Laboratory of Coastal Zone Environmental Processes and Ecological Remediation,

Yantai Institute of Coastal Zone Research, Chinese Academy of Sciences, Yantai 264003,

China

<sup>b</sup> University of Chinese Academy of Sciences, Beijing 100049, China

\*Corresponding author. Phone/Fax: +86 535 2109130.

E-mail address: lxchen@yic.ac.cn (L. Chen)



Fig. S1. UV-Vis absorption of AgNPs.



Fig. S2. TEM images of (A) citrate capped AgNPs and (B) Au@Ag NPs.



Fig. S3. Raman spectra of 4-MPY modified on AgNPs in the presence of protamine. Concentration of protamine:  $0.3 \mu g/mL$ ; concentration of 4-MPY:  $2.5 \mu M$ .



Fig. S4. Raman spectra corresponding to 4-MPY modified different kinds of nanoparticles: (a) hydroxylamine ions-capped AgNPs, (b) Au@Ag NPs, (c) citrate-capped AgNPs.



Fig. S5. Raman spectra corresponding to different kinds of Raman reporters modified on AgNPs. Concentration of protamine: 0.3  $\mu$ g/mL; concentration of 4-MPY, MG and R6G: 2.5  $\mu$ M.



Fig. S6. Effect of the concentration of 4-MPY (A) and protamine (B) on the Raman intensity. Each error bars represent the standard deviations based on three independent measurements.



Fig. S7. Raman spectra of (a) AgNPs+4-MPY and (b) AgNPs+4-MPY+Protamine (5



Table S1. Comparisons of analytical performances of various typical methods for trypsin detection.

Method	Technique in detail	Linear range	Detection limit	Test medium	Ref.
Colorimetry	Arg6 leading to the aggregation of gold nanoparticles	Not given	1.6 ng/mL	PBS (2.0 mM, pH 8.5)	1
Colorimetry	Based on a derivative of Rhodamine B and copper ions becoming colorless in the presence of $\alpha$ -amino acids, and the hydrolysis of bovine serum albumin to produces $\alpha$ -amino acids	0–5.0 μg/mL	Not given	Tris–HCl (10 mM, pH 7.0)	2
Fluorometry	Design and synthesis of novel fluorescent probe based on photo-induced electron transfer	$\begin{array}{l} 4.40 \times 10^{-7} 7.04 \times \\ 10^{-5} \text{ g/mL} \end{array}$	40 ng/mL	PBS(20 mM, pH 8.00)	3
Fluorometry	Peptide-induced fluorescence quenching of conjugated polyelectrolyte	0–20 ng/mL	0.25 ng/mL	PBS (10 mM, pH 7.4) at 37 ℃	4
Fluorometry	Digestion of BSA-stabilized gold nanoclusters	0.01–100 µg/mL	2 ng/mL	Tris–HCl (50 mM, pH 8.0) containing 5 mM CaCl2	5
Electrochemistry	Flash chronopotentiometry using a reversible polycation-sensitive polymeric membrane electrode	20-6,100 ng/mL	20 nM	Phosphate buffer (10 mM, pH 7.4, containing 10 mM NaCl)	6
Electrochemistry	Potentiometric determination using a polymeric membrane polycation-sensitive electrode based on current-controlled reagent delivery	0.5 –5 U/mL	0.3 U/mL	Tris–HCl (50 mM, pH 7.4) containing 120 mM NaCl	7
SERS	SERS method based on anti–aggregation of 4-mercaptopyridine–functionalized silver nanoparticles	0.1–10,000 ng/mL	0.1 ng/mL	HEPES (10 mM, pH 7.4)	This work

## REFERENCES

- 1 W. X. Xue, G. X. Zhang and D. Q. Zhang, Analyst, 2011, 136, 3136–3141.
- 2 X. D. Lou, L. Y. Zhang, J. G. Qin and L. Zhen, Langmuir, 2010, 26, 1566–1569.
- 3 K. H. Xu, F. Liu, J. Ma and B. Tang, Analyst, 2011, 136, 1199–1203.
- 4 H. L. Fan, X. H. Jiang, T. Zhang and Q. H. Jin, *Biosens. Bioelectron.*, 2012, **34**, 221–226.
- 5 L. Z. Hu, S. Han, S. Parveen, Y. L. Yuan, L. Zhang and G. B. Xu, *Biosens*. *Bioelectron.*, 2012, **32**, 297–299.
- 6 K. L. Gemene and M. E. Meyerhoff, Anal. Biochem., 2011, 416, 67-73.
- 7 Y. Chen, J. W. Ding and W. Qin, Bioelectrochemistry, 2012, 88, 144-147.