## Lysozyme-stabilized bimetallic gold/silver nanoclusters as a turn on fluorescent probe for determination of ascorbic acid and acid phosphatase

Shu Pang $\dagger^{a^*}$  and Siyu Liu $\dagger^{b^*}$ 

\*Corresponding author

<sup>a</sup>College of chemistry, chemical engineering and environmental engineering, Liaoning

Shihua University, Fushun 113001, China. Email:Pangshu@lnpu.edu.cn.

<sup>b</sup>College of Life and Health Sciences, Northeastern University, Shenyang 110000,

China.

<sup>†</sup>These authors contributed equally to this work.



**Fig.S1** FT-IR of the free template lysozyme (curve a) and Lys-Au/Ag NCs (curve b) Inset: Zeta potential measurement of Lys-Au/Ag NCs solution.



Fig.S2 EDX spectrum collected for as-prepared Lys-Au/Ag NCs.



Fig.S3 The fluorescence emission intensity of Lys-Au/Ag NCs in different pH environments incubated for 30 minutes.



Fig.S4 The fluorescence emission intensity of Lys-Au/Ag NCs incubated with various NaCl or

KCl concentrations for 30 minutes. (10 mmol/L citrate-citric acid buffer solution pH 5.0)



**Fig.S5** The fluorescence emission intensity of Lys-Au/Ag NCs in the presence of various kinds of biomolecules (200 μmol/L Glu, His, Try, Gly, 100 μmol/L AA, 0.4 mg/mL HAS, HB or Pep). (10 mmol/L citrate-citric acid buffer solution pH 5.0)



Fig.S6 The UV-Vis absorption spectra of Lys-Au/Ag NCs respectively incubated with 0  $\mu$ mol/L(curve a), 50  $\mu$ mol/L(curve b), 100  $\mu$ mol/L (curve c) or 200  $\mu$ mol/L AA (curve d) for 10 minutes.



Fig.S7 The temporal evolution of fluorescence emission intensity of Lys-Au/Ag NCs incubated with of 500  $\mu$ mol/L PAA and 10  $\mu$ g/mL ACP.

Fluorophore	Sensing system	Dynamic range	Detection limit	Reference
Carbon quantum dot	Turn off-on/	5-200 µmol/L	1.35 µmol/L	33
	Cr(VI)ions-AA			
Carbon dots	Turn off/	0.057-4.0 µmol/L	18 nmol/L	34
	Cu <sup>2+</sup> ions-AA			
Graphitic carbon nitride	Turn off-on/	0.6-300 µmol/L	0.15 μmol/L	35
nanosheets	Cr(VI)ions-AA			
Carbon dots	Turn off/	24-40 μg/mL	_	36
	Fe <sup>3+</sup> ions-AA			
Carbon quantum dots	Turn off-on/	0.15-15 μmol/L	0.105 µmol/L	37
/AuNCs nanohybrid	Cd <sup>2+</sup> ions-AA			
Silver/carbon nanohybrid	Turn off-on/	0.2-60 µmol/L	25 nmol/L	38
	Fe <sup>3+</sup> ions-AA			
Gold nanoclusters	Turn off-on/	0.1-10 µmol/L	22 nmol/L	39
	I- ions-AA			
Au nanoclusters	Turn off/	3-40 µmol/L	1.5 μmol/L	40
-PbS quantum dot	AA-quenching			
CdTe quantum dot	Turn off-on/	0.022-0.44mmol/L	4.33 µmol/L	41
	4-AP-AA			
Lyz-Au nanoclusters	Turn-on/	0.2-200 µmol/L	0.12 μmol/L	This
	AA-enhancing			method

Table S1 Performance comparison of various fluorescent sensors for AA detection

Table S2 Performance comp	arison	of various	fluorescent	sensors for	ACP detection
---------------------------	--------	------------	-------------	-------------	---------------

Fluorophores	Sensing systems	Dynamic range	Detection	Reference
			limit	
CuInS <sub>2</sub> quantum dots	Turn off-on-off/	6.4-192 nU/ mL	3.1nU/mL	24
	Cu <sup>2+</sup> -ATP-ACP			
Carbon quantum dots	Turn off-on-off/	18.2-1300 U/L	5.5 U/L	25
	Ni <sup>2+</sup> -pyrophosphate-ACP			
Gold nanoclusters	Turn off-on-off/	1-30 nmol/L	1 nmol/L	26
	Fe <sup>3+</sup> -pyrophosphate-ACP			
Copper nanoclusters	Turn on-off/	3.1-100 U/L	0.8 U/L	27
	pH-Fe(III) pyrophosphate			
	(FePPi <sub>2</sub> ) complex-ACP			
Anionic polymer	Turn off-on-off/	4-28 nmol/L	_	28
	Fe <sup>3+</sup> -pyrophosphate-ACP			
Lyz-Au/Ag NCs	Turn-on/	100-12500ng/mL	53ng/mL	This
	PAA-ACP			method

			<u> </u>	( )
Serum samples	Added ACP	Detected ACP	Recovery	RSD
	(ng/mL)	(ng/mL)	(%)	(n=3, %)
1	100	94	94	4.8
2	500	532	106	3.9

**Table S3** Determination of ACP in fetal bovine serum samples according to equation (2)