Supporting Information for

Visual and efficient immunosensor technique for advancing biomedical applications of quantum dots on *Salmonella* detection and isolation

5 Feng Tang^{a,b}, Dai-Wen Pang^c, Zhi Chen^d, Jian-Bo Shao^e, Ling-Hong Xiong^{c,f}, Yan-Ping Xiang^g,

6 Yan Xiong^d, Kai Wu^h, Hong-Wu Ai^b, Hui Zhangⁱ, Xiao-Li Zheng^e, Jing-Rui Lv^e, Wei-Yong Liu^a,

7 Hong-Bing Hu^e, Hong Mei^e, Zhen Zhang^b, Hong Sun^b, Yun Xiang^{b,*}, Zi-Yong Sun^{a,*}

8 ^aDepartment of Laboratory Medicine, Tongji Hospital, Tongji Medical College, Huazhong
9 University of Science and Technology, Wuhan 430030, People's Republic of China.

^bDepartment of Laboratory Medicine, Wuhan Children's Hospital, Huazhong University of
Science and Technology, Wuhan 430016, People's Republic of China.

12 Key Laboratory of Analytical Chemistry for Biology and Medicine (Ministry of Education),

13 College of Chemistry and Molecular Science, State Key Laboratory of Virology, and Wuhan

14 Institute of Biotechnology, Wuhan University, Wuhan 430072, People's Republic of China.

¹⁵ ^dMicrobiological Laboratory, Wuhan Center for Disease Control and Prevention, Wuhan
¹⁶ 430015, People's Republic of China.

^eWuhan Children's Hospital, Huazhong University of Science and Technology, Wuhan 430016,
People's Republic of China.

¹ ^fShenzhen Center for Disease Control and Prevention, Shenzhen 518055, People's Republic of
 ² China.

³ ^gDepartment of Rehabilitation, Tongji Hospital, Tongji Medical College, Huazhong University
⁴ of Science and Technology, Wuhan 430030, People's Republic of China.

5 ^hJiangan Center for Disease Control and Prevention, Wuhan 430017, People's Republic of China.

⁶ ⁱMicrobiological laboratory, Qiaokou Center for Disease Control and Prevention, Wuhan
7 430030, People's Republic of China.

9	*Corresponding authors: Zi-Yong Sun: zysun@tjh.tjmu.edu.cn; 0086-027-83663639;				
10	Yun Xiang: xiangyun5272008@163.com; 0086-027-82433426				
11					
12					
13					
14					
15					
16					
17					
18					
19					
20					
21					

1 Supplementary Text

2 Supplementary Note 1

3 Reagents and instruments.

Mouse monoclonal antibodies (Product number: B343M) to Salmonella (1 mg/mL) were 4 purchased from Abcam (London). CdSe/ZnS QDs (Product number: Q4525) was purchased from 5 Wuhan Jiayuan Quantum Dots Co., Ltd., China. Xylose lysine desoxycholate agar (XLD, 6 7 Product number: R459902), Rappaport-Vassiliadis agar (RV, Product number: CM1112B), Tryptone Soy Agar (TSA, Product number: CM0277B) and buffered peptone water (BPW, 8 Product number: CM1049B) were purchased from Oxoid (London). Succinimidyl-4-(N-9 10 maleimidomethyl) cyclohexane-1- carboxylate (SMCC, CAS Number: 64987-85-5, Product Number: S0853) was purchased from TCI Europe (Shanghai, China). Dithiothreitol (DTT, 11 Product Number: 20290) and 50X TAE Buffer (Tris-acetate-EDTA) were purchased from 12 Thermo Fisher Scientific Inc (USA). Super-speed Super Absorbing Polymer (SSAP, Product 13 14 number: 127284) was purchased from Liaocheng City Yongxing Environmental Protection Material Co., Ltd. China. All chemicals were of analytical grade and all aqueous solutions were 15 prepared with double-distilled water. 16

Swabs were customized from the Hubei Qianjiang Kingphar medical material Co. Ltd.,
China. All serial dilutions of bacteria were prepared using a turbidity meter (DensiCHEK Plus,
bioMerieux). The biochemical identification of the pathogen in samples was performed using an
automatic microbiology analyzer (VITEK 2 COMPACT, Biomerieux, France).

The artificial excitation light source based on a blue light-emitting diode (LED) and a matched optical cut-off filter (Standard serial number: ZWB2) were customized from Beijing YongXing Information Sensing Technology Co., Ltd. China. The stereomicroscope (14× -90×, trinocular continuous zoom microscope, SZM-45T1) was ordered from the Ningbo Sunny Instruments Co., Ltd. China. Images of fluorescence emission and absorption spectra were recorded on a UV–visible spectrophotometer (UV-2550, Shimadzu, Japan) and a fluorescence spectrophotometer (LS-55, Shimadzu, Japan).

8 Laser scanning confocal microscope (LSCM), transmission electron microscopy (TEM) and 9 field emission scanning electron microscope (FESEM) analyses were conducted on an Olympus 10 FuoViewTM FV1000 system, a Hitachi 7000FA system and a Hitachi S-4800 system, 11 respectively.

2 Supplementary Note 2

3 Preparation of bacterial strains.

4 Bacterial strains were purchased from American Type Culture Collection (ATCC), the National
5 Center for Medical Culture Collections (CMCC, Beijing, China) and the Collection Center of
6 Agricultural Microbiology (CCAM, Hubei, China).

A total of 12 type strains were prepared, including Salmonella Paratyphoid A (CMCC 50093), Salmonella Typhimurium (ATCC 14028), Salmonella Infantis (ATCC 51741), Salmonella manhattan (CMCC 50152), Salmonella Enteritidis (ATCC 13076), Salmonella senftenberg (CMCC 50105), Salmonella Aberdeen (CMCC 50147), Salmonella typhi (ATCC 19430), Salmonella muenster (CCAM 090010), Escherichia coli (ATCC 25922), Proteus mirabilis (ATCC 12453) and Citrobacter freundii (ATCC 10787). Forty local Salmonella belonging to different serotypes were collected and identified by the Center for Disease Control and Prevention (CDC) in Wuhan, Hubei, China.

- -

- 1
- 2 Supplementary Note 3
- 3 Table S1

4 Technical Data of optical cut-off filter (ZWB2)

Test Items	Type/Unit	Parameters of Ultraviolet Glass	
Thickness	mm	1	
	Х	0.532	
A(2856k)	у	0.171	
	Y	0.900	
	Х	0.231	
D65(6504k)	У	0.032	
	Y	0.400	
ND		1.520	
α×10 ⁻⁷	°C	97	
Tg	°C	515	
Ts	°C	589	
S	g/cm ³	2.65	
DA	grade	2	
DW	grade	2	
Tansmitance(313nm)	%	≥ 38.0	
Transmittance(365nm)	%	≥ 80.0	
Transmiitance(405nm)	%	≤ 8.0	
Transmittance(700nm)	%	≤ 14.0	
Bubble	grade	С	
Stripe	grade	4	

		Stress	10 ¹² /Pa	3	
1 2 3 4	A(28 Coeff Propo accor	56k) and D65(6504k) a ficient of thermal expan ortion; DA, Acid resistan dance with the standard o	are the chromaticity sion; Tg, Transition ce; DW, Water resist of International Com	values; ND, Index of refraction; $\alpha \times 10^{-7}$, temperature; Ts, Softening temperature; S, ance; The relative parameters and units are in mission on Illumination (CIE).	, , 1
5					
6					
7					
8					
9					
10					
11					
12					
13					
14					
15					
10					
18					
19					
20					
21					
22					
23					
24					
25					
26					
27					

2 Supplementary Note 4

3 Table S2

4 SSAP (127284) Technical Data

Test Items	Type/Unit	Parameters		
Appearance		White powder		
Free Absorbency For Distilled Water	g/g	≥ 500		
Free Absorbency (0.9 % salty water)	g/g	≥ 50		
Retention Capacity	g/g	\geq 40		
Absorbing Capacity Under Load (0.3 PSI)	g/g	≥ 30		
Absorbing Rate (g/g/sec) (25 Times Salty Water)	seconds	≤ 40		
Moisture Content	%	≤7		
PH Value		6.5~7.5		
\leq 20 Mesh (On 830 µm)	%	0		
20~40 Mesh (830 μm -380 μm)	%	30±5		
20~60 Mesh (830 μm -250 μm)	%	≥ 70		
60~80 Mesh (250 μm -180 μm)	%	≤ 20		
80~100 Mesh (180 μm -150 μm)	%	≤5		
\geq 100 Mesh (Under 150 µm)	%	1		
PSI, Pounds per square inch; Mesh, Unit of granule diameter.				

2 Supplementary Note 5

3 Characterization of mAbs-conjugated QDs bioprobes.

4 As shown in **Figure S1A**, the mAbs-conjugated QDs run slower than the QDs. Under optimal 5 conditions, the concentration of mAbs-conjugated QDs was $98.6 \times 10^{-2} \,\mu\text{g/}\mu\text{L}$. The technique 6 using antibody- conjugated QDs has been designed to increase analytic sensitivity of the 7 detection method.^{1, 2}

8 We used an ultraviolet (UV) visible spectrophotometer and a fluorescence spectrophotometer 9 to record the fluorescence absorption and emission spectra of these mAbs-conjugated QDs 10 (**Figure S1B**). The major antigen on the outer cell wall of *Salmonella* is lipopolysaccaride (LPS), 11 which is about 8 - 10 nm thick. With TEM, we found that most mAbs-conjugated QDs bioprobes 12 attached onto the surface of *Salmonella* (**Figure S1C**).³ We observed that the nanoparticles 13 tended to aggregate on the outer cell wall of *Salmonella*.





Figure S1. Characterization of mAbs-conjugated QDs bioprobes. (A) Images of gel 2 electrophoresis of mAbs-conjugated QDs (lane a) and QDs (lane b). (B) Fluorescence absorption 3 and emission spectra of water-soluble QDs before and after the conjugation. The fluorescence 4 emission spectras of water-soluble QDs before and after the conjugation are almost entirely 5 consistent around 526 nm. (C) TEM micrograph (H-7000FA, magnification of 20.0 k, 75 kV) 6 of Salmonella incubated with mAbs-conjugated QDs for 15 min. UV scanning wavelength range 7 is 400 - 700 nm; scan spacing is 0.5 nm; scan speed is high speed; excitation wavelength is 380 8 nm; scan step is 1 nm; reference solution is 10 mM PBS (pH 7.4). 9

3 References

- 4 1. B. Tang, L. Cao, K. Xu, L. Zhuo, J. Ge, Q. Li, L. Yu, Chemistry, 2008, 14, 3637.
- 5 2. Clark, R. B., M. A. Lewinski, M. J. Loeffelholz, R. J. Tibbetts. Cumitech 31A: Verification

6 and Validation of Procedures in the Clinical Microbiology Laboratory. Coordinating ed., S. E.

- 7 Sharp. ASM Press, Washington, DC. 2009.
- 8 3. R. Schneider, C. Wolpert, H. Guilloteau, L. Balan, J. Lambert, C. Merlin, *Nanotechnology*,
 9 2009, 20, 225101.
- 10

11

12 Supporting Information Available: Description of the material. Refer to Web version on13 PubMed Central for supplementary material.