Supplementary Information

Fluorescent carbon dots with highly negative charge as sensitive probe for real-time monitoring of bacterial viability

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Author Contributions

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1. Experimental Section

1.1 Synthesis of non-doped carbon dots^{S1}

2 g citric acid was put into a 5 mL beaker and heated to 200 °C for 30 min. The obtained orange liquid was added drop by drop into 100 mL of 10 mg/mL NaOH solution under vigorous stirring. After neutralized to pH 7.0 with NaOH, the aqueous solution of non-doped carbon dots was obtained.

1.2 Synthesis of unary atom-doped carbon dots^{\$2,\$3}

The nitrogen-doped carbon dots (NCDs) were synthesized by hydrothermal treatment of CDs (obtained by the developed electrochemical etching method).

1.3 Synthesis of binary atom-doped carbon dots⁸⁴

The nitrogen and sulfur co-doped carbon dots (NSCDs) were synthesized according to a simple hydrothermal method. Briefly, 2 g of citric acid monohydrate and 2 g Lcysteine were mixed within 80 mL deionized water under continuous stirring. The mixture was heated hydrothermally in an autoclave at 80 °C for 72 hours after bubbling N_2 for 30 min. The resulting yellowish-brown solution was cooled to room temperature and filtered with 0.22 mm membranes. To further purify the as-prepared NSCDs, the filtrate was subjected to dialysis against pure water through a dialysis bag (500–1000 MWCO).

1.4 Surface passivation of NPSCDs

Concentrated nitric acid (14.4 mol/L) was added into the solution of NPSCDs (1:10). The mixture was heated at 60 °C for 30 min. The obtained solution was then dialyzed for 1 day (MWCO 1000) to remove unreacted nitric acid.

2. Additional Figures



Figure S1. PL spectra of NPSCDs with excitation wavelengths from 280 nm to 500 nm

with increment of 20 nm.



Figure S2. Fluorescence decay curve of NPSCDs. (300 nm excitation, monitored with

425 nm narrow bandpass filter)



Figure S3. The high-resolution XPS spectra of (a) C 1s and (b) O 1s.



Figure S4. Absorbance-based viability assay to HeLa cells incubated with different concentrations of propidium iodide for 2 h.



Figure S5. HeLa cell imaging with different (a) incubation time (200 $\mu g/mL)$ and (b)

concentration of NPSCDs (3 h).



Figure S6. PL spectra of NPSCDs during (a) heating and (b) cooling processes from 30 °C to 90 °C with increment of 10 °C.



Figure S7. CLSM images of *E coli* (60 °C for 15 min) incubated with (a) CDs, (b) NCDs and (c) NSCDs. (d) ζ potential of the four types of CDs (Blue) and calculated *E. coli* alive rate (red).

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