

Supplementary Materials for:

Lysosome targeting, Cr(VI) and L-AA sensing, and cell imaging based on N-doped blue-fluorescence carbon dots

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Fluorescence QY measurements

The relative fluorescence QY(Φ) of N-CDs was calculated using the equation of $\Phi_x = \Phi_{\text{std}} I_x A_{\text{std}} \eta_x^2 / (I_{\text{std}} A_x \eta_{\text{std}}^2)$. Quinine sulfate in 0.1 M H₂SO₄ was chosen as a reference with a quantum yield (Φ_{std}) of 0.54 at 360 nm. In the equation, I_x and I_{std} are FL intensities of N-CDs and the reference, respectively. A_x and A_{std} denote the optical densities of N-CDs and the reference, respectively. η_x and η_{std} represent the refractive indices of N-CDs and the reference, respectively. The absorbances of all samples in a 1.0 cm cuvette were kept under 0.1 at excitation wavelength to minimize re-absorption effects.

MTT assay

For cell cytotoxicity test, HeLa cells were first plated on a Costar 96-well tissue-culture cluster and cultured at 37°C with 5% CO₂ in air for 3 h to adhere cells onto the surface. The well without cells and treatment with N-CDs was taken as a zero set. The medium was then changed with 100 μ L of fresh DMEM supplemented with 10% FBS containing N-CDs, and cells were allowed to grow for another 24 h. At least five parallel samples were performed in each group. Cells without treatment with N-CDs were taken as a control. After adding 20 μ L of 5.0 mg mL⁻¹ MTT reagent into individual well, the cells were further incubated for 4 h, followed by removing the culture medium

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with MTT, and then 150 μ L of DMSO was added. The resulting mixture was shaken for 10 min at room temperature. The OD of the mixture was measured at 490 nm with a SunRisemicroplate reader (Tecan Austria GmbH, Grödig, Austria). The cell viability was estimated using the equation of Cell Viability (%) = $(OD_{\text{Treated}}/OD_{\text{Control}}) \times 100\%$, where OD_{Control} and OD_{Treated} were obtained in absence and presence of N-CDs, respectively.

Synthesis process of N-CDs

Blue-fluorescence N-CDs were fabricated through hydrothermal carbonization of folic acid and p-phenylenediamine. The formation process of N-CDs likely involve decomposition, polymerization, carbonization, and nuclear burst. Firstly, folic acid and p-phenylenediamine formed intermediates through dehydration and decomposition. Afterwards, these intermediates were polymerized, carbonized and aromatized to form aromatic clusters. Simultaneously, the aromatic clusters may undergo nuclear explosions to form N-CDs. To get good FL properties, we investigated the influence of reaction temperature and time on FL properties of resultant N-CDs. As shown in Fig. S1, the optimum reaction condition for N-CDs is 220°C for 4 h.

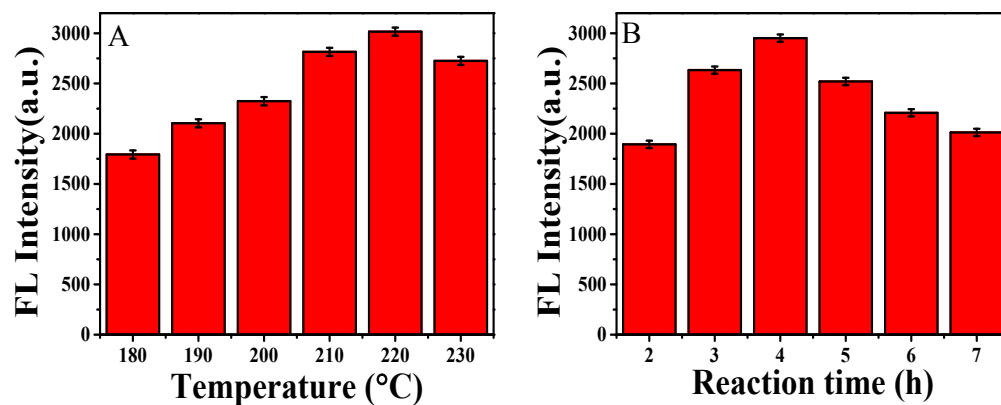


Fig. S1 FL intensity of N-CDs prepared under different reaction temperature (A) and time (B).

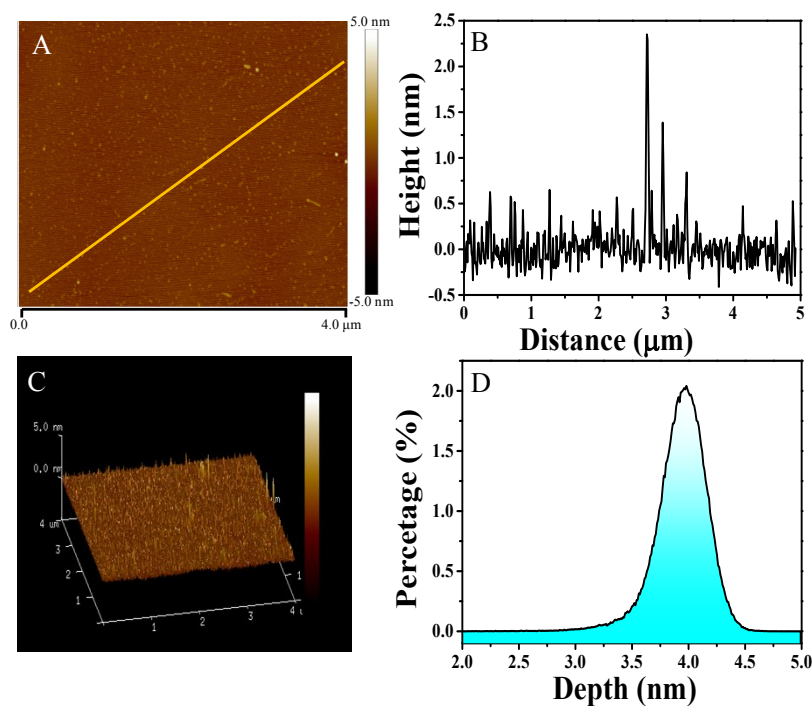


Fig. S2 (A) AFM topographic image of N-CDs. (B) Distribution of height profile along line in AFM topographic image. (C) AFM three-dimensional image of N-CDs. (D) The height distribution of N-CDs.

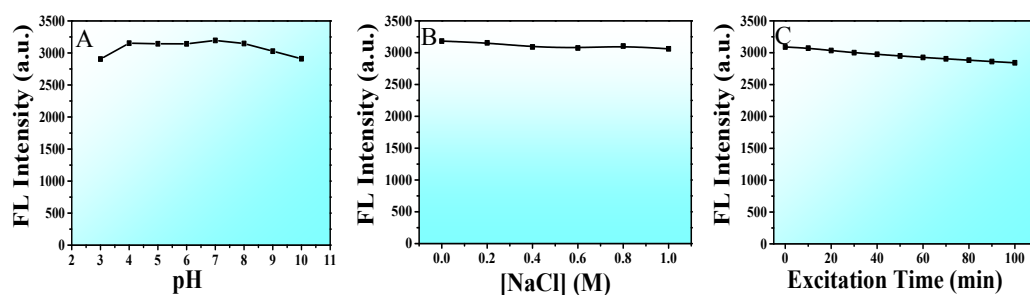


Fig. S3 (A) Effect of pH on FL intensity of N-CDs. (B) Effect of NaCl concentration on FL intensity of N-CDs. (C) Effect of excitation time on FL intensity of N-CDs.

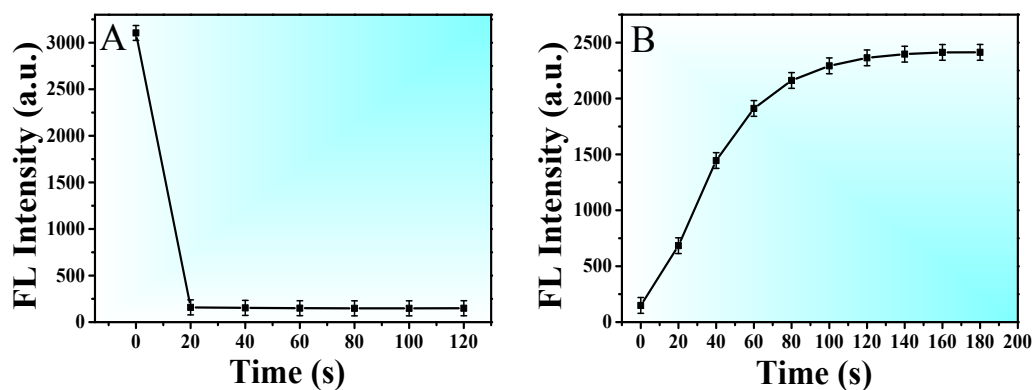


Fig. S4 (A) Time-dependence of FL intensity of N-CDs/Cr(VI) under 360 nm excitation. (B) Time-dependence of FL intensity of N-CDs/Cr(VI)/L-AA solution under 360 nm excitation.

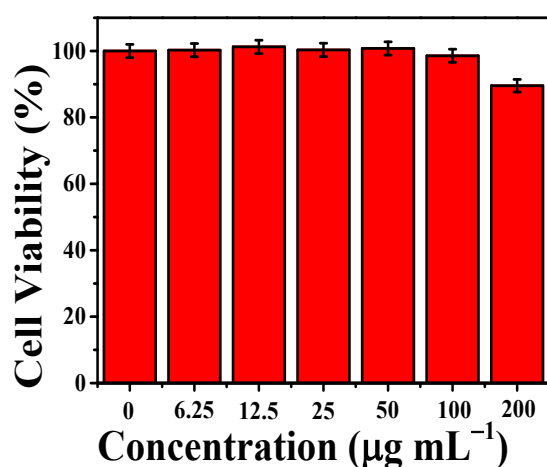


Fig. S5 Toxic effect of N-CDs on HeLa cells.

Table S1 Comparison of different CDs based Cr(VI) probes.

Fluorescent probes	CDs precursor	Linear range (μM)	Detection limit (nM)	Ref.
CDs	waste tea extract	1.9–115.4	810	[1]
CDs	flax straw, EDA	0.5–80	190	[2]
Ag-doped CDs	FA, PE, AgNO ₃	0.1–6	43.7	[3]
CDs	m-phenylenediamine, citric acid	0–5	140	[4]
N-CDs	citric acid, ethylenediamine	0–10	260	[5]
CDs	4-aminoacetanilide, hydrochloride, 4-acetamidobenzaldehyde	1–400	130	[6]
P,N-CDs	EDA, ethylenediamine	7–70	710	[7]
N-CDs	folic acid, p-phenylenediamine	0.10–150	9.4	This work

Table S2 Comparison of various CDs based L-AA probes.

Fluorescent probes	CDs precursor	Linear range (μM)	Detection limit (μM)	Ref.
Tea-CDs/Cr(VI)	waste tea extract	500–5000	87.02	[1]
CDs/Cr(VI)	flax straw, EDA	0–200	0.35	[2]
S,N-CDs/Cr(VI)	glucose, H ₂ SO ₄ , EDA	6.6–892	0.076	[8]
CDs/NO ₂ ⁻	nicotinic acid, folic acid	0.1–800	50	[9]
N-CDs/Cr(VI)	CA, Glu	1–750	0.3	[10]
N-CDs/Cr(VI)	p-PD, ammonia	5–50	0.32	[11]
CDs@EDTA/Cr(VI)	orange peels, EDTA	0.1–400	0.1	[12]
N-CDs/Cr(VI)	folic acid, p-phenylenediamine	750–2250	25	This work

References

- 1 K. Chen, W. X. Qing, W. P. Hu, M. H. Lu, Y. Wang and X. H. Liu, *Spectrochim. Acta. A*, 2019, **213**, 228–234.
- 2 G. K. Hu, L. Ge, Y. Y. Li, M. Mukhtar, B. Shen, D. S. Yang and J. G. Li, *J. Colloid Interface Sci.*, 2020, **579**, 96–108.
- 3 B. Hu, Y. Ma and N. Wang, *Microchem. J.*, 2020, **157**, 104855.
- 4 G. X. Qiao, D. Lu, Y. P. Tang, J. W. Gao and Q. M. Wang, *Dyes Pigm.*, 2019, **163**, 102–110.
- 5 S. H. Liu, J. L. Cui, J. B. Huang, B. S. Tian, F. Jia and Z. L. Wang, *Spectrochim. Acta. A*, 2019, **206**, 65–71.
- 6 F. P. Mutuyimana, J. Liu, S. Nsanzamahoro, M. Na, H. L. Chen and X. G. Chen, *Microchim. Acta*, 2019, **186**, 163.
- 7 B. Wu, X. F. Shi, W. Han, T. S. Wang, C. R. Wang and L. Jiang, *RSC Adv.*, 2018, **8**, 31793–31802.
- 8 S. M. Song, F. Liang, M. L. Li, F. F. Du, W. J. Dong, X. J. Gong, S. M. Shuang and C. Dong, *Spectrochim. Acta. A*, 2019, **215**, 58–68.
- 9 L. L. Gan, Q. Su, Z. B. Chen and X. M. Yang, *Appl. Surf. Sci.*, 2020, **530**, 147269.
- 10 Y. H. Zhang, X. Fang, H. Zhao and Z. X. Li, *Talanta*, 2018, **181**, 318–325.
- 11 Y. T. Meng, Y. Jiao, Y. Zhang, Y. Li, Y. F. Gao, W. J. Lu, Y. Liu, S. M. Shuang and C. Dong, *Talanta*, 2020, **210**, 120653.
- 12 M. Wang, R. Shi, M. J. Gao, K. L. Zhang, L. L. Deng, Q. F. Fu, L. J. Wang and D. Gao, *Food Chem.*, 2020, **318**, 126506.