

Shining a Light on Cells: Amine-Passivated Fluorescent Carbon Dots as Bioimaging Nanoprobes

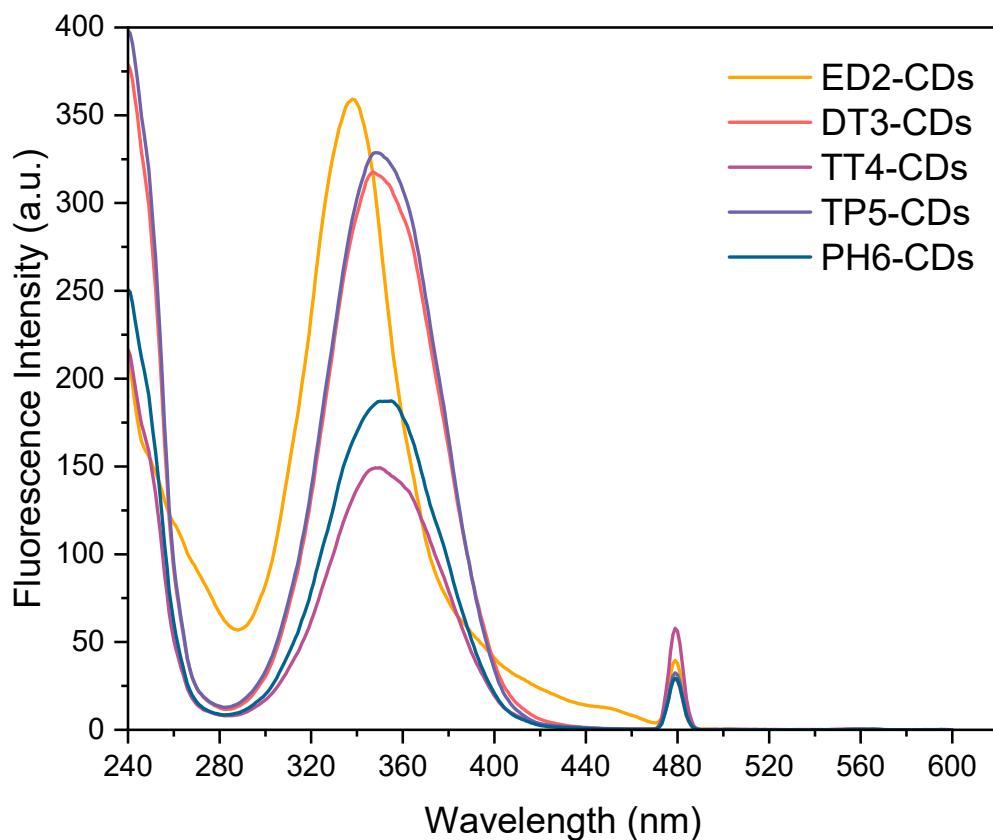
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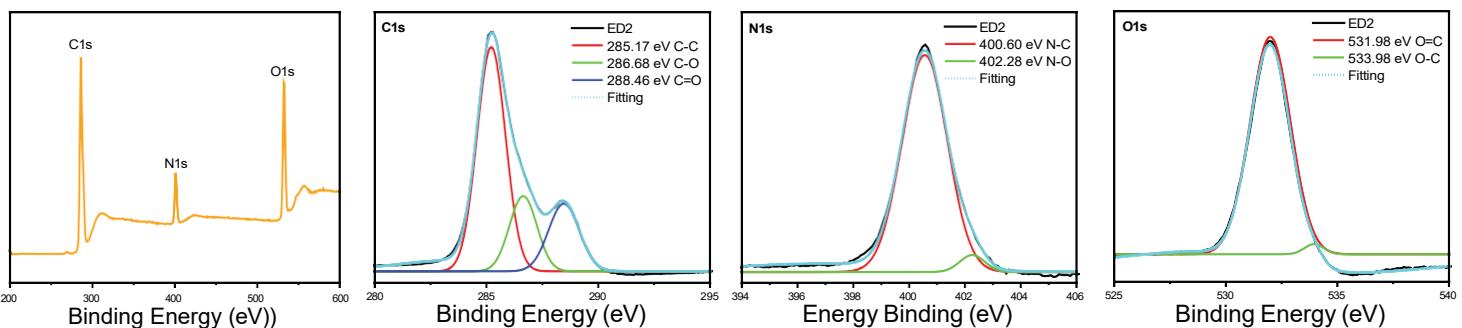
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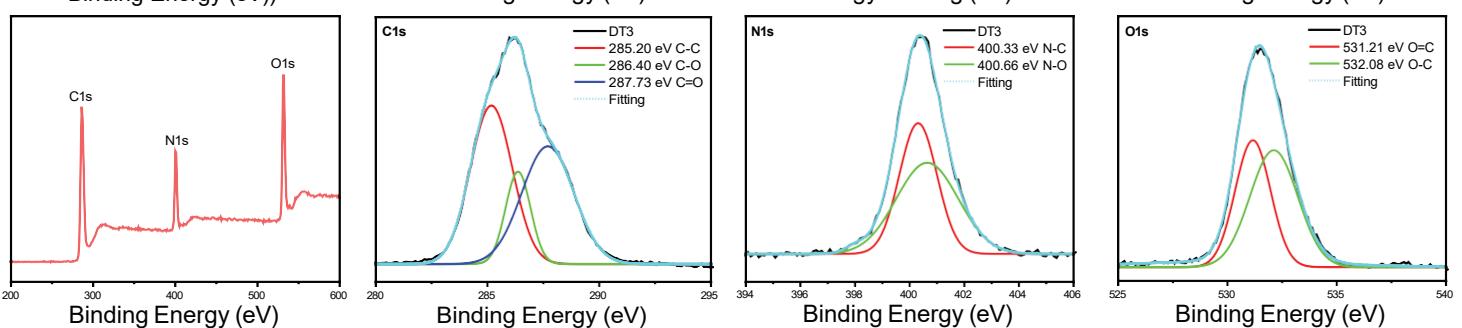


S1. Fluorescence excitation spectra of amine passivated CDs measured at 480 nm emission.

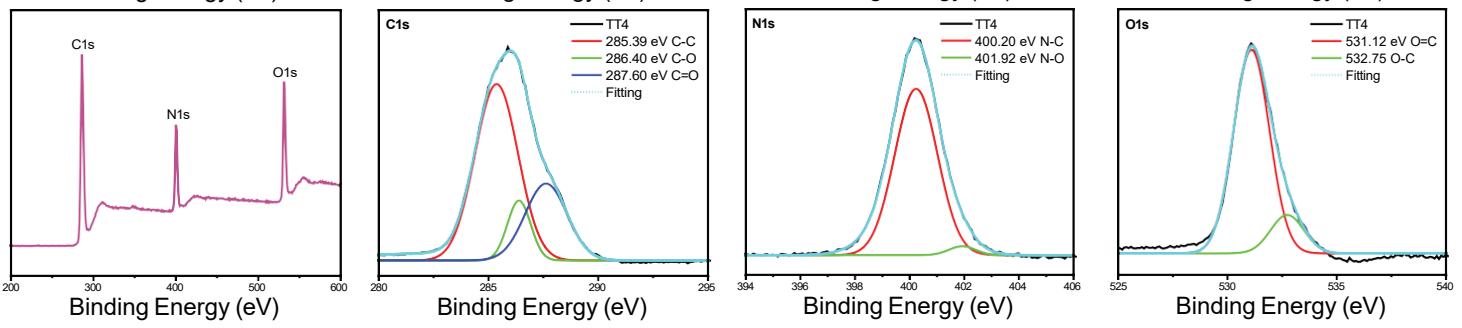
A



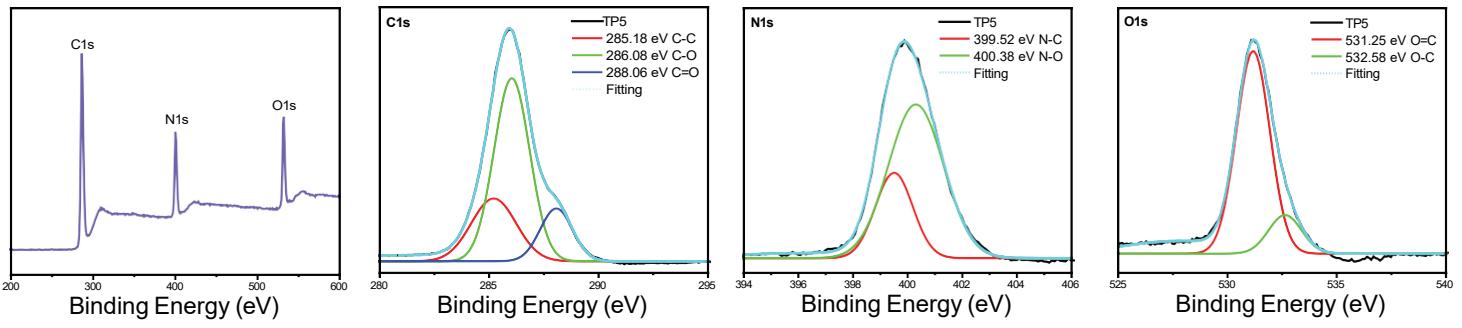
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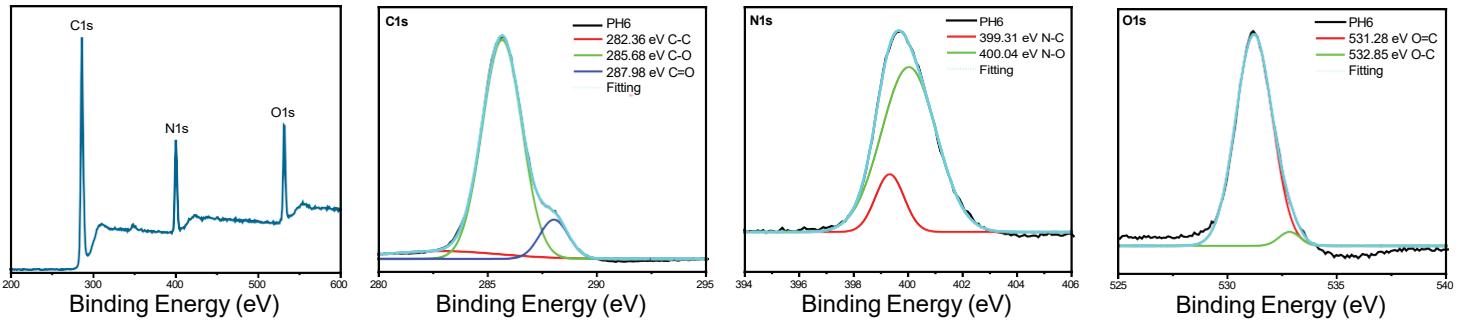
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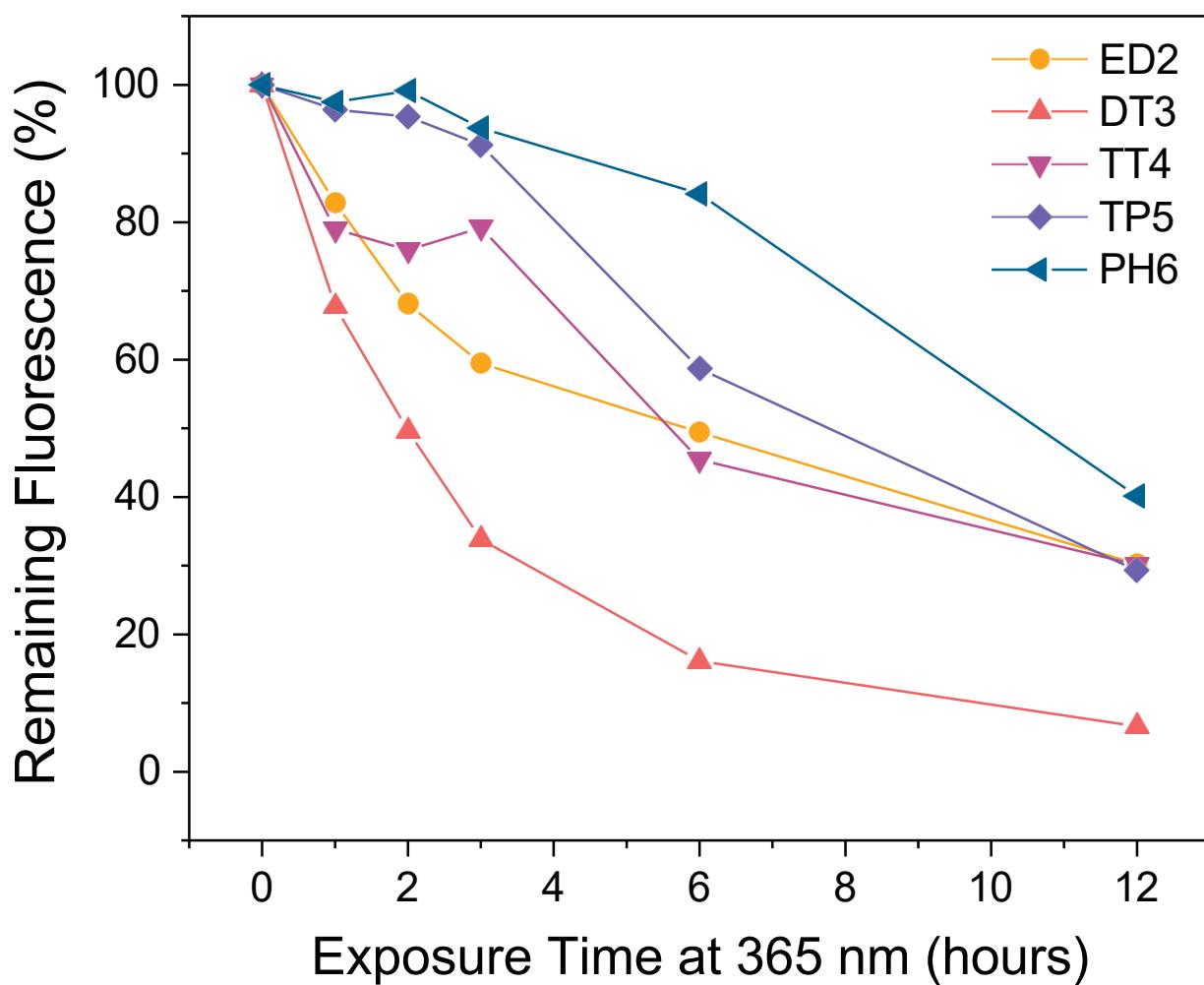
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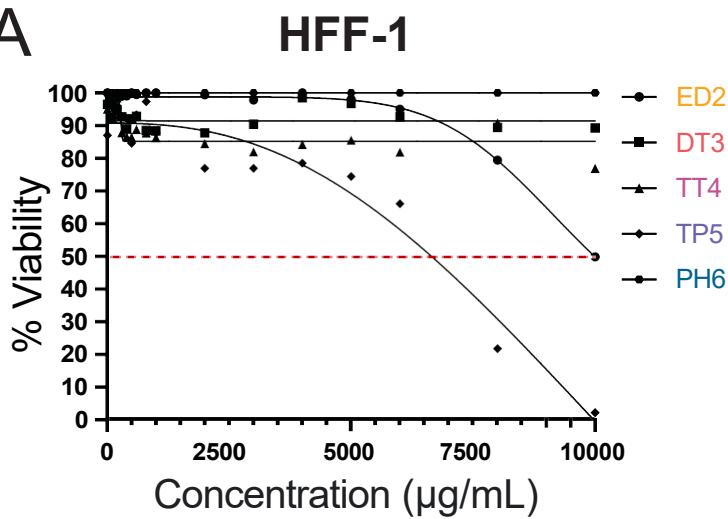
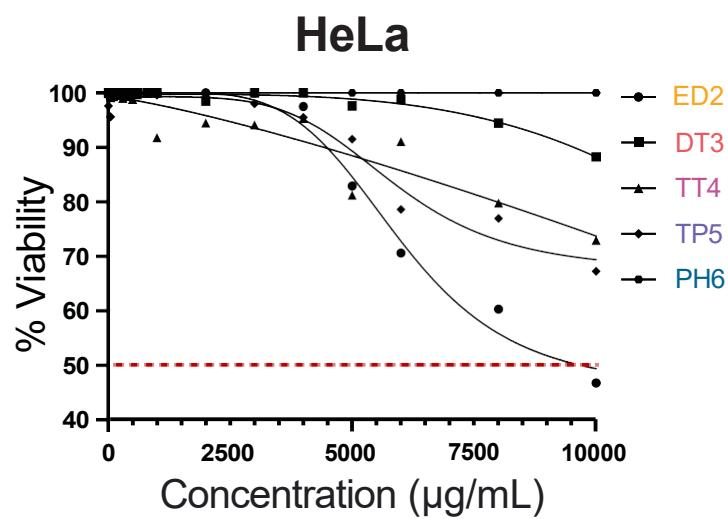
E



S2. XPS survey and deconvoluted spectra of all CDs showcasing 3 binding energies ascribed to C1s, N1s and O1s for (A) ED2-CDs; (B) DT3-CDs; (C) TT4-CDs; (D) TP5-CDs; (E) PH6-CDs



S3. Amine-passivated CDs are highly photostable. A graph shows the fluorescence (%) of CDs following exposure to 365 nm light over a period of 0-12 hours.

A**B**

S4. Cell viability in HFF-1 and HeLa cells after treatment with amine passivated carbon dots (A) The left graph shows the IC50 (dotted line) for HFF-1 cell viability, after treatment with varying concentrations of the different CDs for three population doubling times (7,500 $\mu\text{g/mL}$ and 10,000 $\mu\text{g/mL}$ for TP5-CDs and ED2-CDs respectively, and $>10,000 \mu\text{g/mL}$ for TT4-, DT3-, PH6-CDs). (B) The right graph shows the IC50 (dotted line) for HeLa cell viability, after treatment with various concentrations of the different CDs for three population doubling times (10,000 $\mu\text{g/mL}$ for ED2-CDs and $>10,000 \mu\text{g/mL}$ for DT3-, TT4-, TP5- and PH6-CDs).

Table S1. One-Way ANOVA and Paired t-test statistical analysis for cellular uptake of CDs in both HeLa and HFF-1 cells for Fig.3

Statistical Test	Cell Line	Groups	P-value	Data Interpretation
One-way ANOVA		ED2 DT3 TT4 TP5 PH6	1.76E ⁻³²	Population means are significantly different
Paired t-test	HeLa	ED2:PH6	1.33E ⁻⁹	Population means are significantly different
		DT3:PH6	1.81E ⁻⁹	Population means are significantly different
		TT4:PH6	1.09E ⁻⁹	Population means are significantly different
		TP5:PH6	5.58E ⁻⁷	Population means are significantly different
One-way ANOVA	HFF-1	ED2 DT3 TT4 TP5 PH6	7.40E ⁻⁸	Population means are significantly different
Paired t-test	HFF-1	ED2:PH6	3.69E ⁻⁴	Population means are significantly different
		DT3:PH6	7.78E ⁻⁷	Population means are significantly different
		TT4:PH6	4.01E ⁻⁵	Population means are significantly different
		TP5:PH6	6.47E ⁻⁵	Population means are significantly different

Table S2. One-Way ANOVA and Paired t-test statistical analysis for Lysosome and Cytosol co-localization of CDs in HFF-1 cells for **Fig.4**

Statistical Test	Co-Localization	Groups	P-value	Data Interpretation
One-way ANOVA	Lysosome	ED2 DT3 TT4 TP5 PH6	3.80E ⁻³²	Population means are significantly different
Paired t-test	Lysosome	ED2:PH6 DT3:PH6 TT4:PH6 TP5:PH6	9.71E ⁻¹⁰ 6.10E ⁻¹⁰ 3.01E ⁻⁸ 1.83E ⁻⁴	Population means are significantly different Population means are significantly different Population means are significantly different Population means are significantly different
One-way ANOVA	Cytosol	ED2 DT3 TT4 TP5 PH6	1.46E ⁻¹¹	Population means are significantly different
Paired t-test	Cytosol	ED2:PH6 DT3:PH6 TT4:PH6 TP5:PH6	2.36E ⁻⁸ 1.45E ⁻⁶ 1.44E ⁻⁴ 7.78E ⁻⁷	Population means are significantly different Population means are significantly different Population means are significantly different Population means are significantly different

Table S3. One-Way ANOVA and Paired t-test statistical analysis for co-localization in the cytosol of CDs in both HeLa and HFF-1 cells for **Fig.5**

Statistical Test	Co-Localization	Groups	P-value	Data Interpretation
One-way ANOVA	Lysosome	ED2 DT3 TT4 TP5 PH6	3.80E ⁻³²	Population means are significantly different
Paired t-test	Lysosome	ED2:PH6	9.71E ⁻¹⁰	Population means are significantly different
		DT3:PH6	6.10E ⁻¹⁰	Population means are significantly different
		TT4:PH6	3.01E ⁻⁸	Population means are significantly different
		TP5:PH6	1.83E ⁻⁴	Population means are significantly different
One-way ANOVA	Cytosol	ED2 DT3 TT4 TP5 PH6	2.39E ⁻¹¹	Population means are significantly different
Paired t-test	Cytosol	ED2:PH6	4.64E ⁻⁵	Population means are significantly different
		DT3:PH6	0.00172	Population means are significantly different
		TT4:PH6	0.02642	Population means are significantly different
		TP5:PH6	8.83E ⁻⁵	Population means are significantly different