

Facile Preparation of Low Cytotoxicity Fluorescent Carbon Nanocrystals by Electrooxidation of Graphite

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1. XPS of CNCs

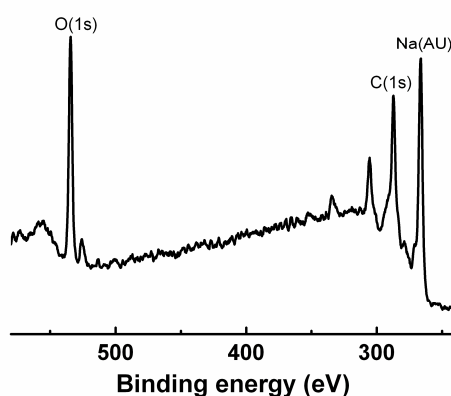


Figure S1. XPS of CNCs

2. Determination of quantum yield

The quantum yield (Φ) of CNCs was calculated by comparing the integrated photoluminescence intensities (excited at 330 nm) and the absorbance values at 330 nm of the CNCs with the reference quinine sulfate. The quinine sulfate (QS) (literature $\Phi = 0.54$) was dissolved in 0.1 M H_2SO_4 (refractive index (η) of 1.33) and the CNCs was dissolved in ultra pure water ($\eta = 1.33$).

The data was plotted in Figure S2. The slopes of the sample and the standards were determined. The data showed good linearity. The quantum yield was calculated using the below equation:

$$\Phi_x = \Phi_{ST} (m_x / m_{ST}) (\eta_x^2 / \eta_{ST}^2)$$

Where Φ is the quantum yield, m the slope, η the refractive index of the solvent, ST for the standard and X for the sample. The quantum yield of the CNCs was calculated to be 0.012.

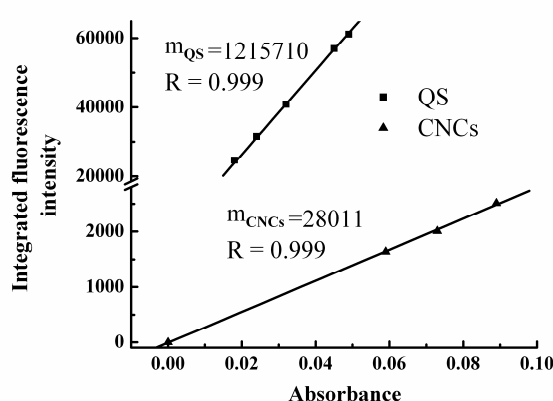


Figure S2. Plot of integrated photoluminescence intensity vs. absorbance of the blue CNCs and quinine sulfate.

3. Effect of excitation wavelength on the fluorescence spectra

Purified CNCs (<5 kDa) was dissolved in ultra pure water. The fluorescence spectra of the CNCs were measured with a fluorimeter (LS-55, Perkin Elmer), with a slit width of 10 nm and 15 nm for excitation and emission, respectively. The excitation wavelength increased by a 10 nm increment starting from 290 nm. Corresponding spectra were given in Figure S3. The emission peak did not shift with varying excitation wavelength. The emission spectrum of CNCs was excitation independent.

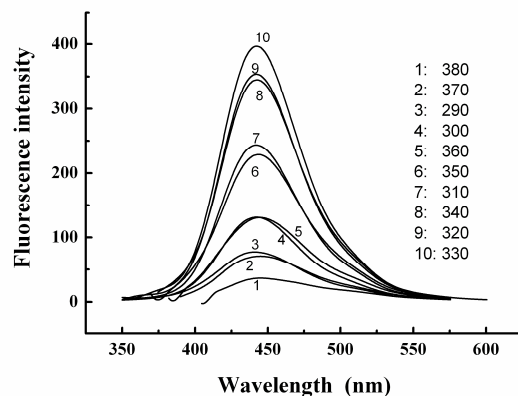


Figure S3. Emission spectra of blue CNCs at different excitation wavelengths as indicated.

4. Photostability of blue fluorescent CNCs

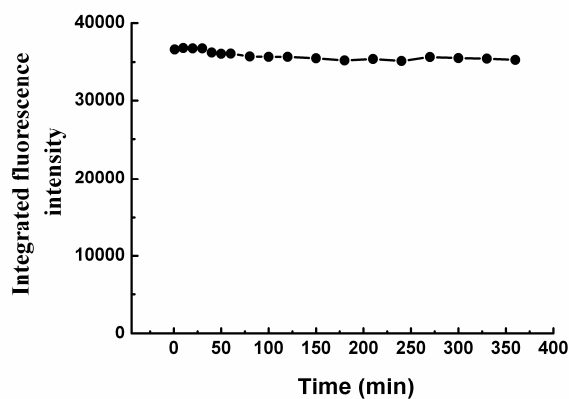


Figure S4. Dependence of fluorescence intensity on excitation time for blue CNCs in ultra pure water.

5. Influence of ionic strengths

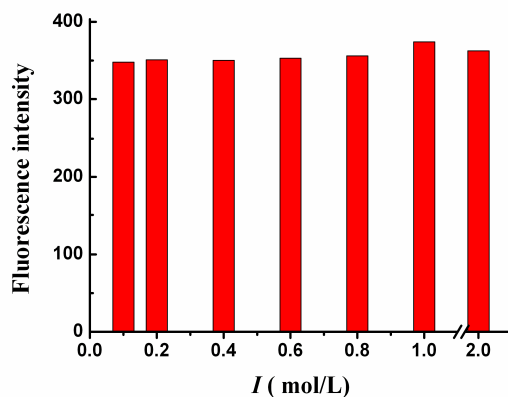


Figure S5. Fluorescence intensity of blue fluorescent CNCs in KCl aqueous solution (pH 7) against the ionic strength.

6. Effect of pH on the photoluminescence intensity of CNCs

The photoluminescence intensity of CNCs decreased when the pH value was larger or smaller than 4.5. The photoluminescence intensity could be recovered by adjusting the pH value back to around 4.5 (Figure S7). And the emission peak did not shift with varying pH value.

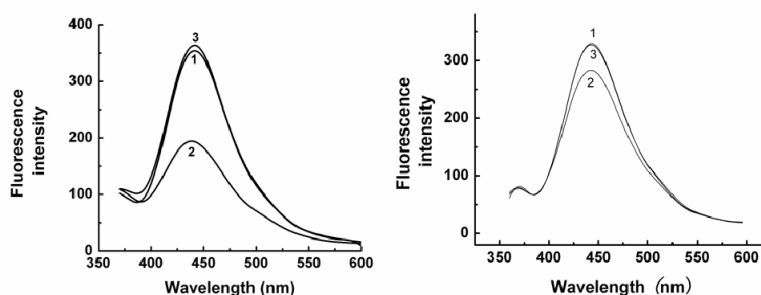


Figure S6. (Left) Spectra of CNCs at various pHs. (1) pH 5, (2) pH 2.0 by adding H_2SO_4 in (1) and (3) pH 5 by adding NaOH in (2). (Right) Spectra of CNCs at various pHs. (1) pH 6.0, (2) pH 9.2 by adding NaOH in (1) and (3) pH 6.0 by adding H_2SO_4 in (2).

7. Fourier-transform infrared (FT-IR) spectra of CNCs

The infrared spectra of the CNCs were obtained on a Thermo Nicolet 360 FT-IR spectrophotometer. Pressed pellets were prepared by grinding the powder samples with KBr in an agate mortar.

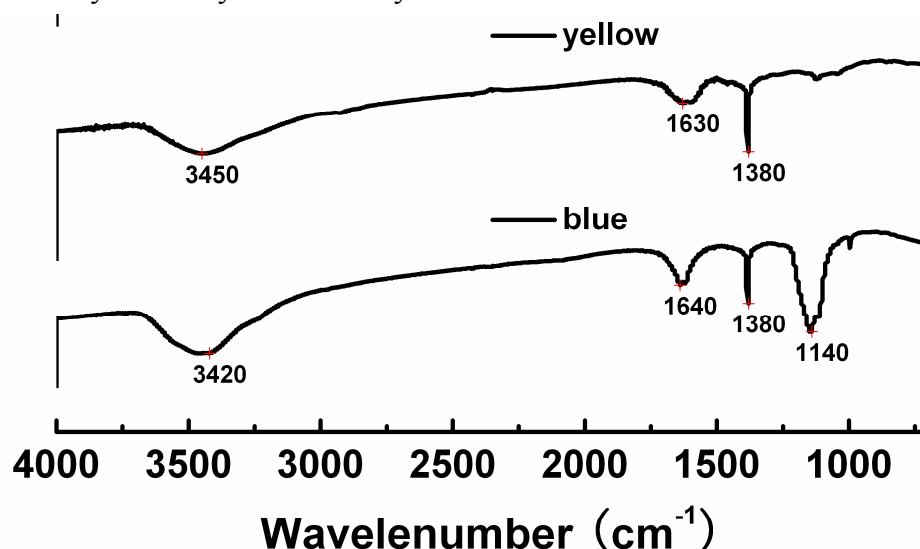


Figure S7. FT-IR spectra of CNCs.

8. Cytotoxicity Tests

100 μ l of suspension of 293T human kidney cells (8×10^4 cells/mL) in Dulbecco's modified Eagle's medium supplemented with 10% fetal bovine serum and 1% penicillin/streptomycin (DMEM) was added to each well of a 96-well plate. The cells were cultured first for 24 h in an incubator (37°C, 5% CO₂), and for another 24 h after the culture medium was replaced with 100 μ l of DMEM containing the CNCs at different doses. Then, 20 μ l of 5 mg/mL MTT solution was added to every cell well. The cells were further incubated for 4 h, followed by removing the culture medium with MTT, and then 150 μ L of DMSO was added. The resulting mixture was shaken for ca. 5 min at room temperature. The optical density (OD) of the mixture was measured at 570 nm with a Microplate Reader Model 550 (BIO-RAD, USA). The cell viability was estimated according to the following equation:

$$\text{Cell viability [\%]} = (\text{OD}_{\text{treated}} / \text{OD}_{\text{control}}) \times 100$$

where OD_{control} was obtained in the absence of CNCs, and OD_{treated} obtained in the presence of CNCs.