Supporting Information

A Low-Temperature Solid-Phase Method to Synthesize Highly Fluorescent Carbon Nitride Dots with Tunable Emission

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1. Preparation of graphitic Carbon Nitride Dots (g-CNQDs)

0.101 g (1.68 mmol) of urea and 0.081g (0.28 mmol) of sodium citrate were mixed in an agate mortar (6 cm) and grounded to a uniform powder. The mixture was placed in an autoclave and heated to 180 ^{o}C for 1 h in a drying oven. The resultant yellowish mixture was purified by alternative washing with ethanol and centrifugation at 12, 000 rpm for three times, and then dialyzing against pure water through a dialysis membrane for 24 h. Four species of g-CNQDs with distinct emission were synthesized by tuning the amount of sodium citrate. 0.101 g (1.68 mmol) of urea was separately reacted with 0.165 g (0.56 mmol), 0.081 g (0.28 mmol), 0.055 g (0.187 mmol) and 0.041 g (0.14 mmol) of sodium citrate, making the molar ratio of urea to sodium citrate vary from 3:1 to 6:1, 9:1, and 12:1. The as-obtained g-CNQDs were finely dissolved in water and demonstrated bright fluorescence performance under a UV-lamp. The stable aqueous solutions were further characterized by fluorescence and UV-Vis spectroscopy, dynamic light scattering, and transmission electron microscopy.

In the control experiments, 1.68 mmol of urea and 0.28 mmol of sodium citrate were separately placed in an autoclave and heated to 180 °C for 1 h in a drying oven. After being cooled to room temperature, both of the two reactants showed no obvious change. The as-obtained powders were white in colours and showed no fluorescence emission when dissolved in water.

2. Characterizations

A UV-2450 spectrophotometer (Shimadzu, Columbia, MD) and a Fluoromax4 fluorometer (Horiba Jobin Yvon, Edison, NJ) were used to record the absorption and emission spectra of the g-CNQDs. The surface charge of the g-CNQDs was measured with a Zetasizer Nano-ZS size analyzer (Malvern, Worcestershire, UK). HR-TEM images were carried out on a FEI Tecnai field emission electron microscope at an acceleration voltage of 200 KV. X-ray powder diffraction was collected on D8 Advance. X-ray photoelectron spectra were acquired on an imaging photoelectron spectrometer (Axis ultra, Kratos Analytical Ltd.). The atomic force microscopy (AFM) images were obtained using a tapping mode of Agilent 5500 Atomic Force Microscope. Infrared spectra were collected on a VERTEX 70 Fourier transform infrared (FT-IR) spectrometer (Bruker) in a KBr pellet, scanning from 4000 to 400 cm^{-1} at room temperature. The confocal fluorescence images of HEK 293T cells were acquired by a laser scanning confocal fluorescence microscopy (Leica TCS SP5).

3. Quantum Yield Measurements

Quantum yield was measured according to the established procedure (Lakowicz, J. R. *rinciples of Fluorescence Spectroscopy*, 2nd Ed., **1999**, Kluwer Academic/Plenum Publishers, New York). The optical densities were measured with the UV-vis spectra obtained by a UV5800

Spectrophotometer. Quinine sulfate in 0.1 M H2SO4 (literature quantum yield of 0.54 at 360 nm) was used as the standard. Absolute values were calculated using the standard reference sample that has a fixed and known fluorescence quantum yield value according to the following equation:

$$\varphi_X = \varphi_S (A_s / A_x)(I_x / I_s)(\eta^2{}_x / \eta^2{}_s)$$

where φ is the quantum yield, A is the optical density, and η is the refractive index.

4. Cell Culture and Cell Imaging

HEK 293T cells (approximately 1×10^5 cells/well) were seeded in a 24-well plate and cultured at 37 °C with 5% CO_2 overnight to adhere cells onto the surface. Then the culture medium was replaced by 0.5 mL of fresh medium containing 0.8 mg/mL g-CNQDs, and the cells were incubated for another 24 h. The cells were washed with PBS buffers (pH = 7.4) for three times, followed by being fixed with 4% paraformaldehyde solution for 10 minutes and being washed with PBS buffers for twice. Finally, the fixed cells were observed under a confocal fluorescence microscopy.

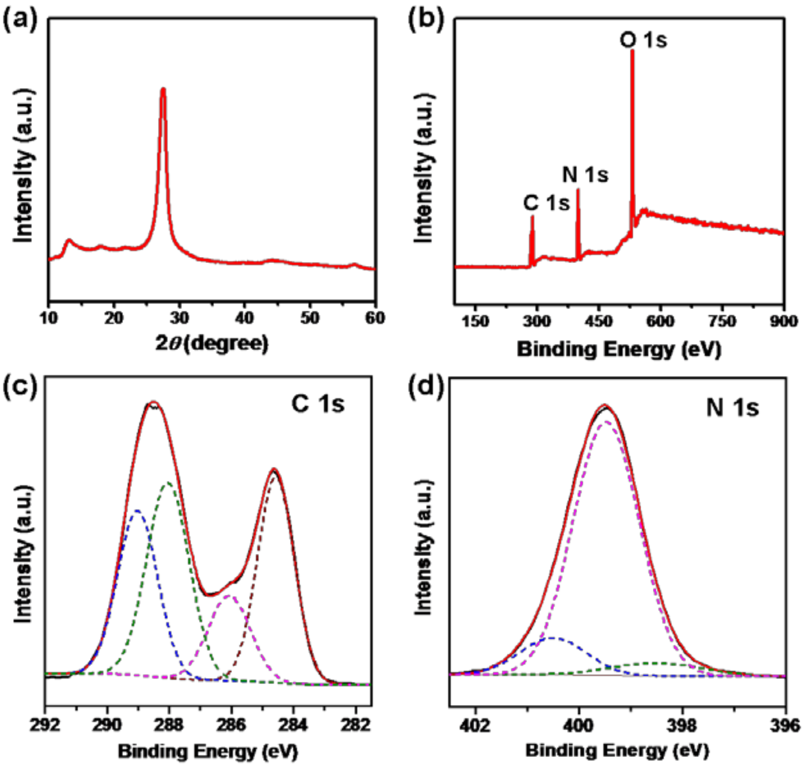

Fig. S1 (a) XRD spectra and (b) XPS spectra of the obtained g-CNQDs. (c) and (d) are the corresponding C1s spectrum and N1s spectrum of g-CNQDs.

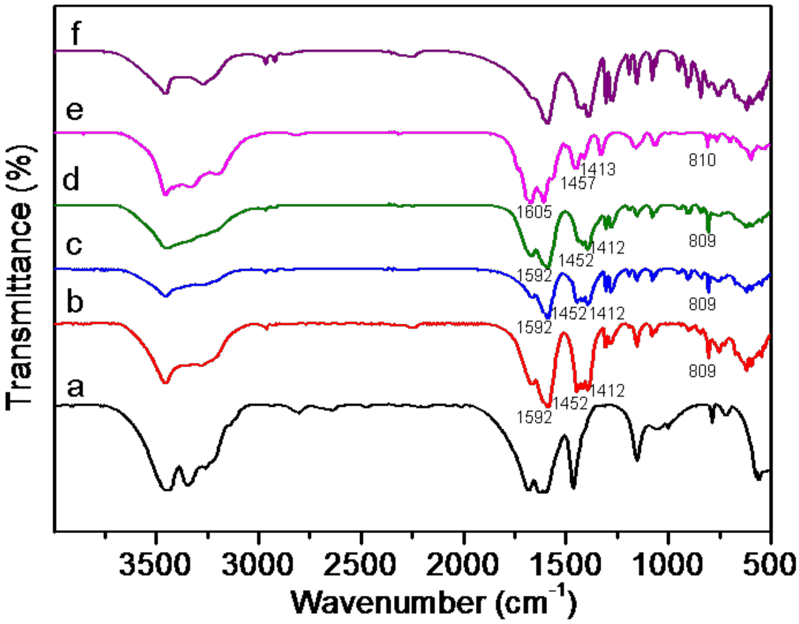

Fig. S2 FT-IR characterizations of the obtained g-CNQDs. (a) Pure urea; (b-e) g-CNQDs obtained by using different molar ratio of urea to sodium citrate: (b) 3:1, (c) 6:1, (d) 9:1, and (e) 12:1; (f) pure sodium citrate. The characteristic band around 809 cm^{-1} is attributed to the heptazine units, the bands at 1412 cm^{-1} and 1452 cm^{-1} correspond to the typical stretching modes of CN hetercycles.[1] The broad peaks between 3100 and 3450 cm^{-1} are attributed to N-H and O-H stretching, and the high-intensity peak at 1590 cm^{-1} corresponds to the asymmetric stretching vibrations of the carboxylate anions, indicating that carboxylate and hydroxyl groups are on the surface of the g-CNQDs.

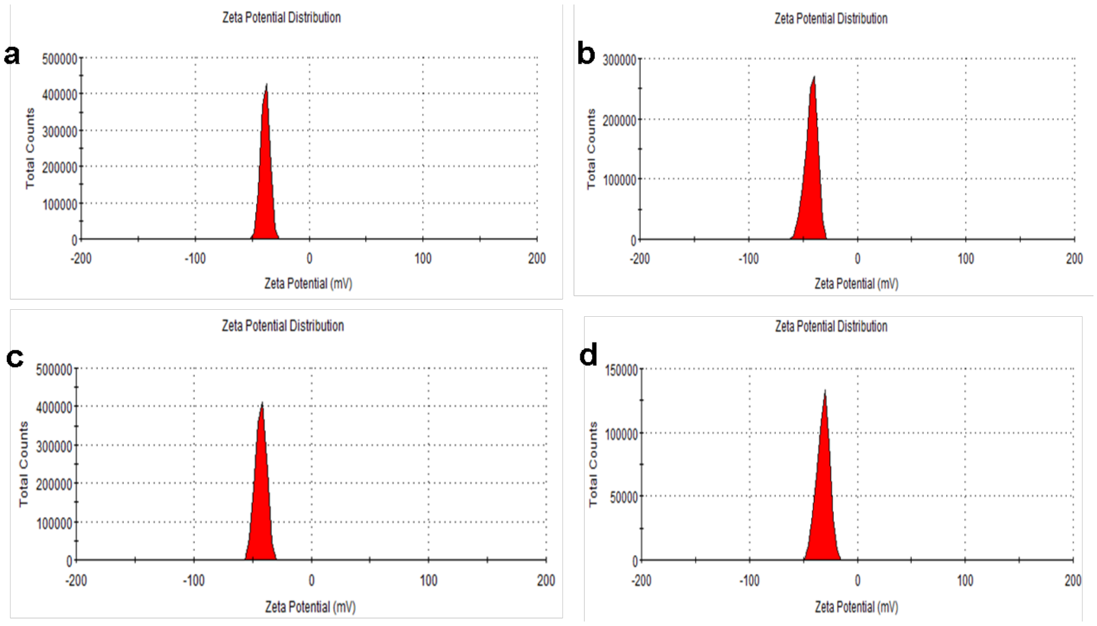

Fig. S3 Average zeta potential of the obtained carbon nitride dots. (a) -38.5 mV, urea and sodium citrate with the molar ratio of 3:1; (b) -42.4 mV, urea and sodium citrate with the molar ratio of 6:1; (c) -42.5 mV, urea and sodium citrate with the molar ratio of 9:1; (d) -31.6 mV, urea and sodium citrate with the molar ratio of 12:1.

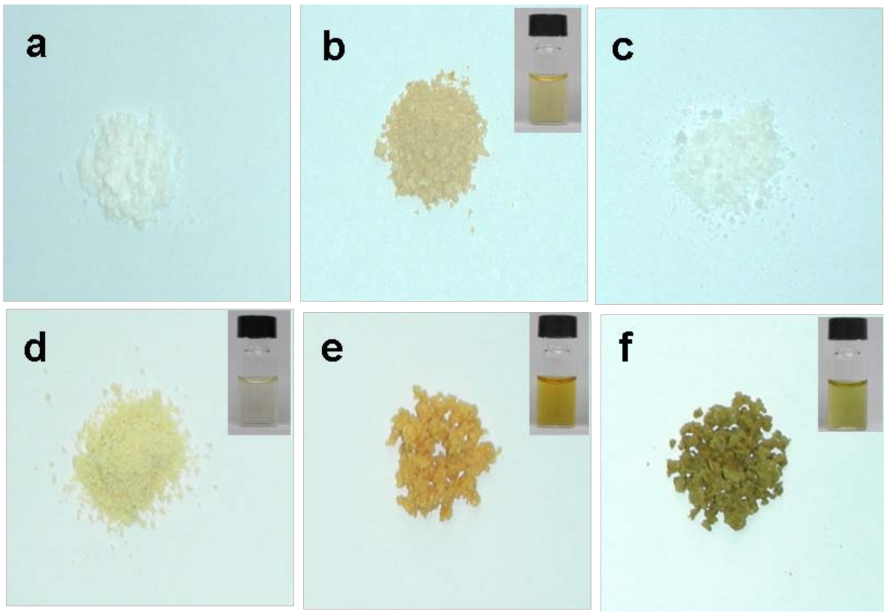

Fig. S4 Photos of dried carbon nitrides produced by altering the molar ratio of urea to sodium citrate. (a) only urea heating at 180 °C; (b) urea and sodium citrate with the molar ratio of 6:1; (c) only sodium citrate heating at 180 °C; d) urea and sodium citrate with the molar ratio of 3:1; (e) urea and sodium citrate with the molar ratio of 9:1; (f) urea and sodium citrate with molar ratio of 12:1. The inset pictures of b, d, e and f are the corresponding carbon nitride dots dispersed in water. The concentration of each solution is 5 mg/mL.

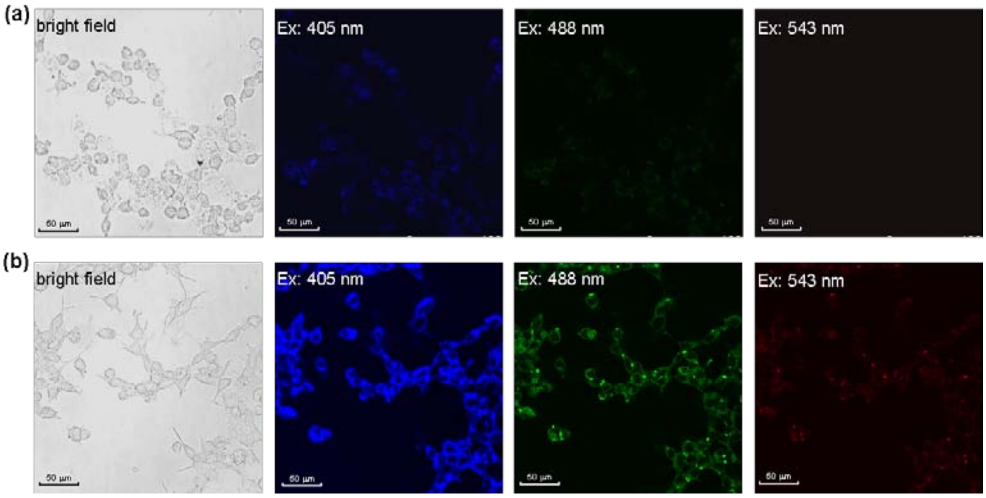

Fig. S5 Confocal fluorescence images of HEK 293T cells after incubation without (a) and with (b) g-CNQDs for 24 h.

References

1. (*a*) J. H. Liu, T. D. Zhang, Z. C. Wang, G. Dawson and W. Chen, *J. Mater. Chem.*, 2011, **21**, 14398; (*b*) M. J. Bojdys, J. –O. Müller, M. Antonietti and A. Thomas, *Chem.-Eur. J.*, 2008, **14**, 8177.