

## Electronic Supplementary Information

### Highly luminescent carbon nanodots by microwave-assisted pyrolysis

Xinyun Zhai,<sup>‡a</sup> Peng Zhang,<sup>‡ac</sup> Changjun Liu,<sup>ab</sup>, Tao Bai,<sup>a</sup> Wenchen Li,<sup>a</sup> Liming Dai,<sup>c</sup>  
Wenguang Liu<sup>\*a</sup>

<sup>a</sup> School of Materials Science and Engineering, Tianjin Key Laboratory of Composite and  
Functional Materials, Tianjin University, Tianjin 300072, PR China

<sup>b</sup> Institute of Medical Equipment, Academy of Military Medical Sciences, Tianjin 300161, PR  
China

<sup>c</sup> Department of Macromolecular Science and Engineering, Case School of Engineering, Case  
Western Reserve University, 10900, Euclid Avenue, Cleveland, Ohio 44106, United States

<sup>‡</sup> These authors contributed equally to this work.

### Materials

Citric acid (CA, 99+%) and 3-(4,5-dimethyl-2-thiazoyl)-2,5-diphenyl tetrazolium bromide (MTT, 98%) were purchased from Alfa Aesar. 1,2-ethylenediamine (EDA, 98%), 1,4-butanediamine (BDA, 98%), diethylamine (DEA, 98%) and triethylamine (TEA, 98%) were purchased from GuangFu Technology Development CO, LTD. Quinine sulfate (98%, suitable for fluorescence) was supplied by Fluka. All other reagents were of analytical grades and used without further purification.

### Synthesis procedure

Firstly, 1 g CA was diluted with 10 ml water and then mixed with different amount of EDA (58.5 mg – 625.6 mg) under vigorous stirring, the carbonization proceeded very fast and no inorganic salt or acid was needed. Then, the clear transparent solution was put into a domestic microwave oven (700 W) and heated for different time periods. When cooled down to room

---

\* To whom correspondence should be addressed  
E-mail: [wgliu@tju.edu.cn](mailto:wgliu@tju.edu.cn)

temperature, the obtained red-brown and foamy solid was dissolved and dialyzed against pure water through a dialysis membrane (MWCO of 100) for 3 days. Finally, the red-brown aqueous solution containing both CA and CDs was precipitated and rinsed with anhydrous ethanol twice, and vacuum-dried at ambient temperature to collect 0.98g product (the yield of the reaction was about 75%).

The CDs synthesized with DEA, TEA, BDA were obtained through the same procedure via microwave irradiation of an aqueous solution of CA (100 mg/ml) containing DEA (76.1 mg/ml), TEA (105.3 mg/ml) and BDA (45.9 mg/ml), respectively. The best reaction time for EDA, DEA, TEA and BDA was 2 min, 4 min, 4 min, and 3 min, respectively, and the best microwave time for CDs without passivation was 3 min. In order to obtain the optimum fluorescence, the molar ratio between  $-\text{NH}_2$  and  $-\text{COOH}$  was determined to be 2:3.

## Characterization

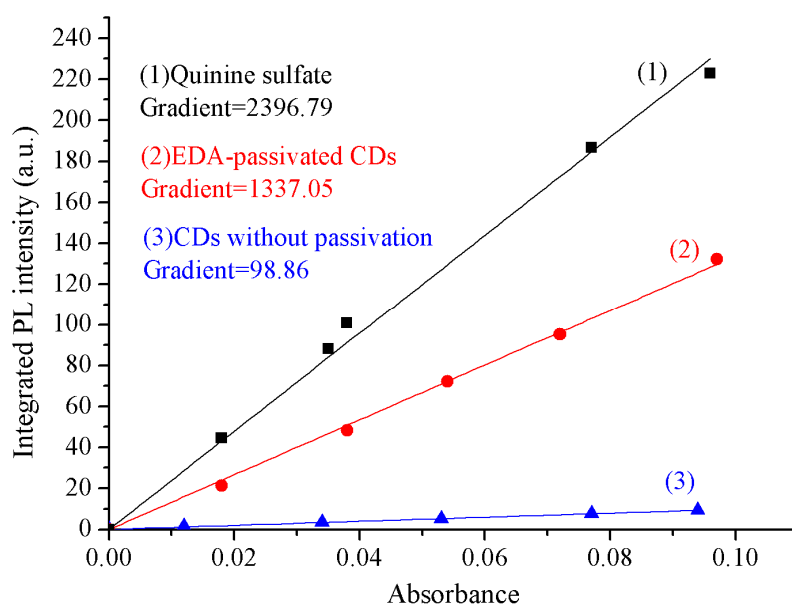
UV-Vis absorption was measured on a TU-1810 UV-Vis Spectrophotometer (Pgeneral, China). Photoluminescence (PL) emission measurements were performed using FLS920 fluorometer (Edinburgh Instruments, Britain). The normalized spectrum was obtained by divided each intensity of the PL spectrum by the maximum value of its own. The morphology and microstructure of the CDs were examined by high-resolution transmission electron microscopy (HRTEM) on a Philips Tecnai G2 F20 microscope (Philips, Netherlands) with an accelerating voltage of 200 kV. The samples for HRTEM were made by dropping an aqueous solution onto a 300-mesh copper grid coated with a lacy carbon film. The FTIR spectra of the samples were measured on a Nicolet 380 spectrometer (Thermo, America). The XPS spectra of the samples were measured on a Kratos AXIS Ultra DLD X-ray Photoelectron Spectroscopy (Shimadzu, Japan). X-Ray diffraction (XRD) profiles of the prepared samples were recorded on a Rigaku-D/MAX 2500 diffractometer (Rigaku, Japan) equipped with graphite monochromatized  $\text{CuK}\alpha$  ( $\lambda=0.15405\text{ nm}$ ) radiation at a scanning speed of  $4^\circ/\text{min}$  in the range from  $10^\circ$  to  $90^\circ$ . The composition of CDs was further confirmed by elemental analysis with Vanio-EL (Elementar Analysensysteme GmbH, Germany).

## Measurement of fluorescence quantum yields

The quantum yield of the Carbon Dots (CDs) was determined by a comparative method. Quinine sulfate in 0.1 M H<sub>2</sub>SO<sub>4</sub> (literature quantum yield: 54%) was used as a standard sample to calculate the QY of test samples which were dissolved in ultra pure water at different concentrations. All the absorbance values of the solutions at the excitation wavelength were measured with UV–Vis spectrophotometer. Photoluminescence (PL) emission spectra of all the sample solutions were recorded by FLS920 fluorometer at an excitation wavelength of 360 nm. The integrated fluorescence intensity is the area under the PL curve in the wavelength range from 380 to 700 nm. Then, a graph was plotted using the integrated fluorescence intensity against the absorbance and a trend line was added for each curve with intercept at zero. Absolute values were calculated according to the following equation:

$$\Phi_X = \Phi_{ST} \left( \frac{Grad_X}{Grad_{ST}} \right) \left( \frac{\eta_X^2}{\eta_{ST}^2} \right)$$

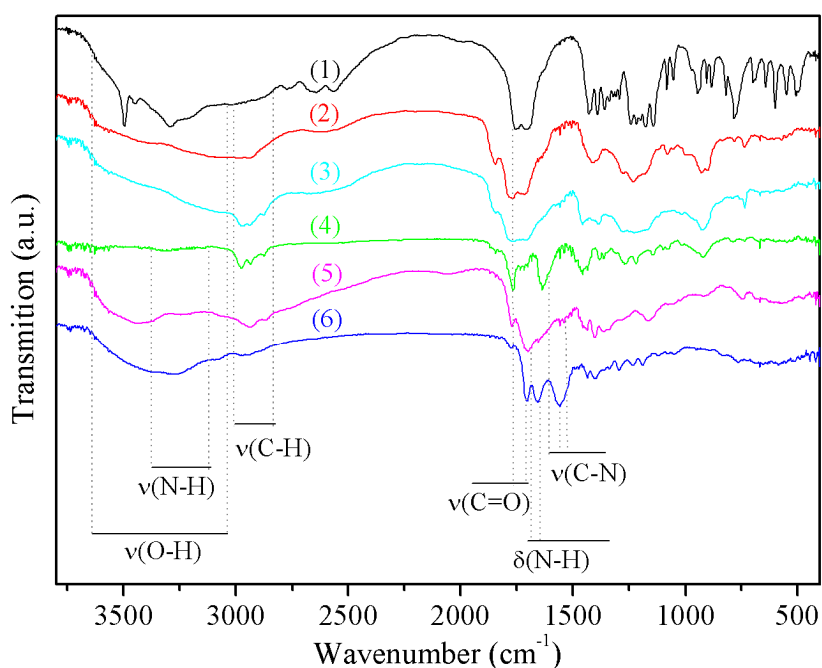
where the subscripts ST and X denote standard and test, respectively,  $\Phi$  is the fluorescence quantum yield, Grad is the gradient from the plot of integrated fluorescence intensity vs absorbance, and  $\eta$  is the refractive index of the solvent. In order to minimize the re-absorption effects, absorbance in the 10 mm fluorescence cuvette should never exceed 0.1 at the excitation wavelength.



**Figure S1.** Photoluminescence and absorbance of C-dots (results: EDA-CDs 30.2%; N-free CDs 2.22%, excited at 360nm).

## FTIR characterization of CDs

The pure CA exhibited characteristic absorption bands of  $\nu$  (O-H) and  $\nu$  (C=O) at  $3288\text{ cm}^{-1}$  and  $1732\text{ cm}^{-1}$ , respectively, which indicated the existence of  $-\text{COOH}$ . The FTIR spectra of the N-free CDs and TEA-CDs are very similar, both showing peaks of  $\nu$  (O-H),  $\nu$  (C-H) and  $\nu$  (C=O) at about  $3320\text{ cm}^{-1}$ ,  $2934\text{ cm}^{-1}$  and  $1764\text{ cm}^{-1}$  respectively. The EDA-CDs, BDA-CDs and DEA-CDs all exhibited characteristic absorption bands of  $\nu$  (C=O) ( $1732\text{ cm}^{-1}$ ) and  $\nu$  (C-N) ( $1580\text{ cm}^{-1}$ ), indicating the formation of  $-\text{CONR}$ , while the absorption bands of  $\nu$  (N-H) ( $3264\text{ cm}^{-1}$ ) and  $\delta$  (N-H) ( $1662\text{ cm}^{-1}$ ) of DEA-CDs diminished.



**Figure S2.** FT-IR spectra of CA (1), N-free CDs (2), TEA-CDs (3), DEA-CDs (4), BDA-CDs (5) and EDA-CDs (6).

**Table S1** The results of elemental analysis, QY and luminescence lifetime of CDs.

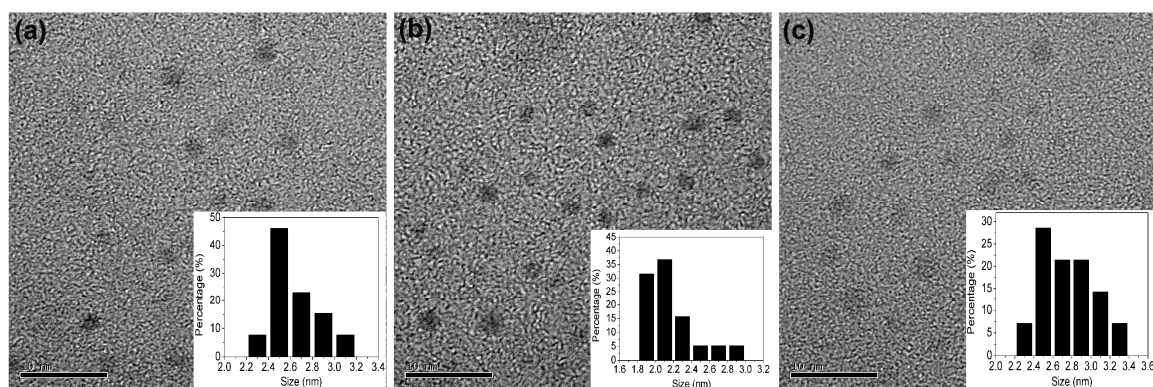
| Samples   | C%    | H%   | N%    | O<br>(Calculated, %) | QY (%) | PL lifetime (ns) |
|-----------|-------|------|-------|----------------------|--------|------------------|
| N-free CD | 50.59 | 4.93 | 0.10  | 44.38                | 2.2    | 3.65             |
| EDA-CD    | 53.39 | 6.00 | 20.15 | 20.46                | 30.2   | 15.96            |
| DEA-CD    | 62.26 | 8.70 | 6.21  | 22.83                | 4.2    | 4.74             |
| TEA-CD    | 61.25 | 6.64 | 0.58  | 31.53                | 3.2    | 4.14             |
| BDA-CD    | 54.10 | 7.05 | 12.07 | 26.78                | 7.9    | 10.79            |

## Cell culture, confocal microscopy and cytotoxicity assay

L929 (murine aneuploid fibrosarcoma cell line) was obtained from Peking Union Medical College (Beijing, China). The cells were cultured in RPMI-1640 Medium (1640, HyClone), containing 10% fetal bovine serum (FBS), 100 U/ml penicillin and 100 mg/ml streptomycin at 37 °C in 5% CO<sub>2</sub> humidified atmosphere.

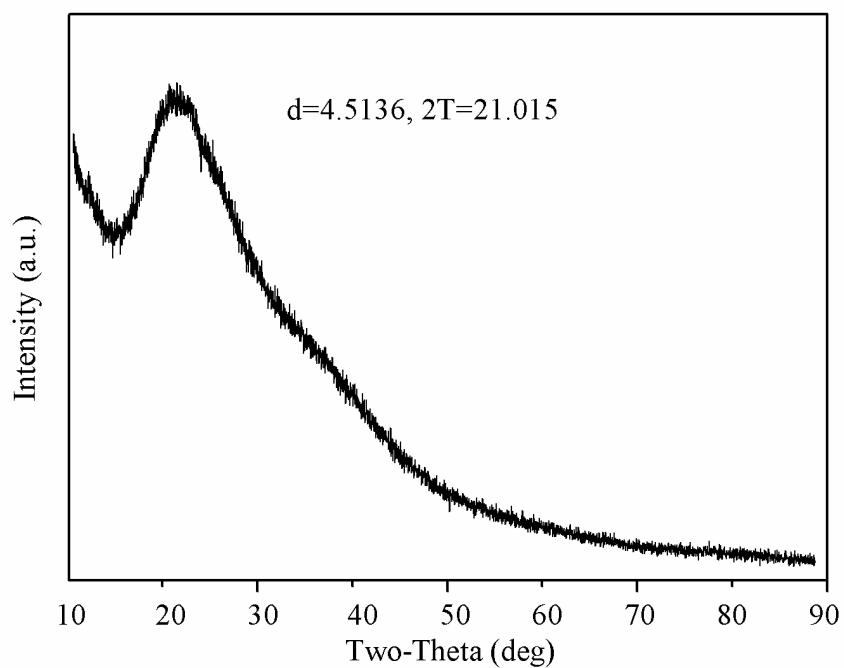
For confocal microscopy, L929 cells were seeded on a coverslip in 6-well plate 12 h before use. Then, the culture medium was replaced by 2.5 ml fresh medium containing 2 mg/ml EDA-CDs and the cells were incubated for another 24 h. The cells were then washed with isotonic PBS (pH 7.4) three times, and fixed with 4 % paraformaldehyde solution in PBS at 4 °C overnight. The samples were examined under a Leica confocal laser scanning microscope (Mannheim, Germany) equipped with a UV laser (351/364 nm), an Ar laser (457/488/514nm) and a HeNe laser (543/633 nm).

The cytotoxicity of EDA-CDs was assessed through MTT assay. L929 cells were seeded in a 96-well plate, at a density of  $2 \times 10^4$  cells/well and incubated overnight. Then, the culture medium was removed and the CDs at the increasing concentrations from 0.5 to 10 mg/ml were added into each well and incubated for 24 h before refreshing the medium with 200  $\mu$ l fresh complete medium containing 20  $\mu$ l MTT (5 mg/ml in PBS). The plate was further incubated for 4 h. Finally, all medium was removed and 150  $\mu$ l/well DMSO was added, followed by shaking for 15 min. The absorbance of each well was measured at 570 nm using a Synergy HT Multi-Mode Microplate Reader (BioTek, USA) with pure DMSO as a blank. Non-treated cell was used as a control and the relative cell viability (mean%  $\pm$  SD, n = 3) was expressed as  $\text{Abs}_{\text{sample}}/\text{Abs}_{\text{control}} \times 100\%$ .

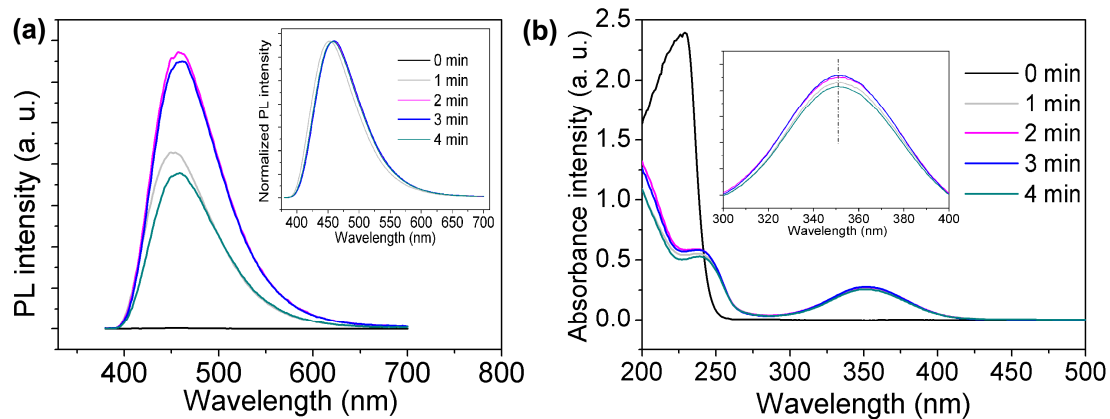


**Figure S3.** HRTEM images of (a) DEA-CDs, (b) TEA-CDs and (c) BDA-CDs. The scale bars are all 10 nm.

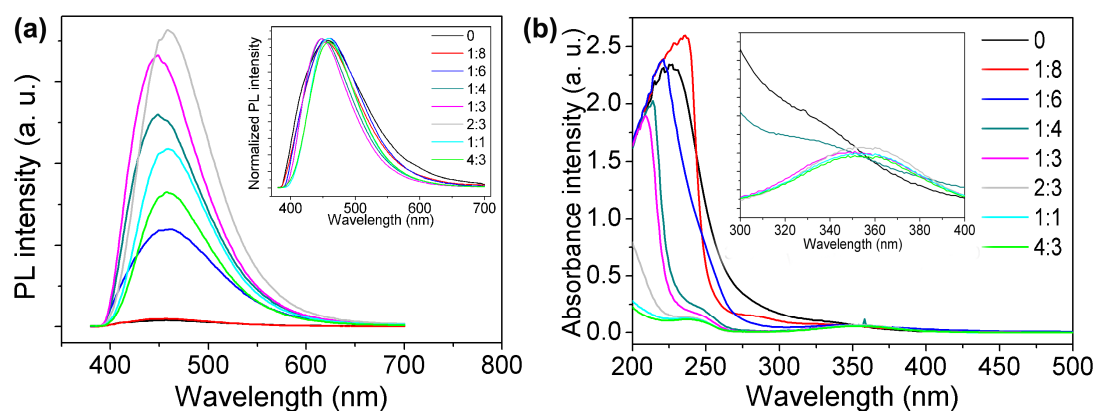




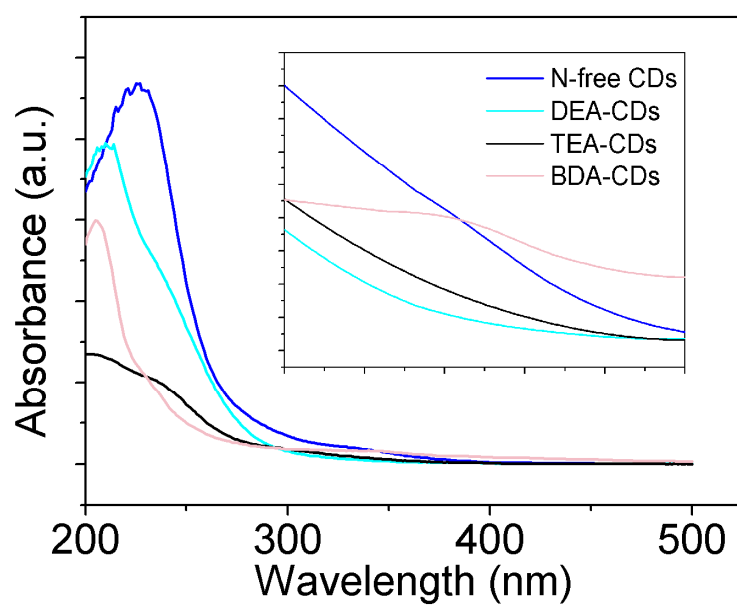
**Figure S4.** A typical XRD profile for EDA-CDs.



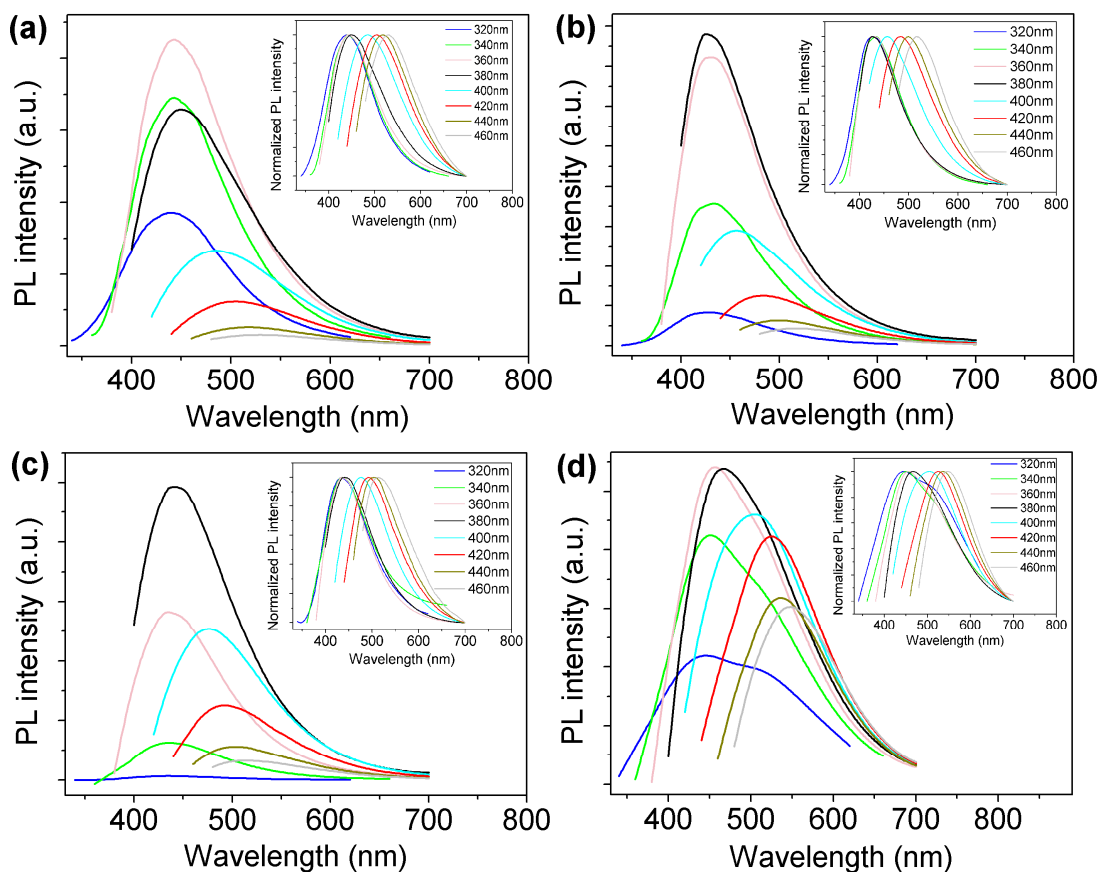
**Figure S5.** PL spectra ((a), excited at 360 nm) and UV-Vis absorbance spectra (b) of EDA-CDs prepared at different microwave pyrolysis time periods (1 g CA, 384  $\mu$ l EDA, 10 ml H<sub>2</sub>O); the insets are (a) normalized PL spectra, (b) magnified absorbance spectra from 300 nm to 400 nm exclusive of 0 time treatment, respectively. Each sample has the same absorbance value.



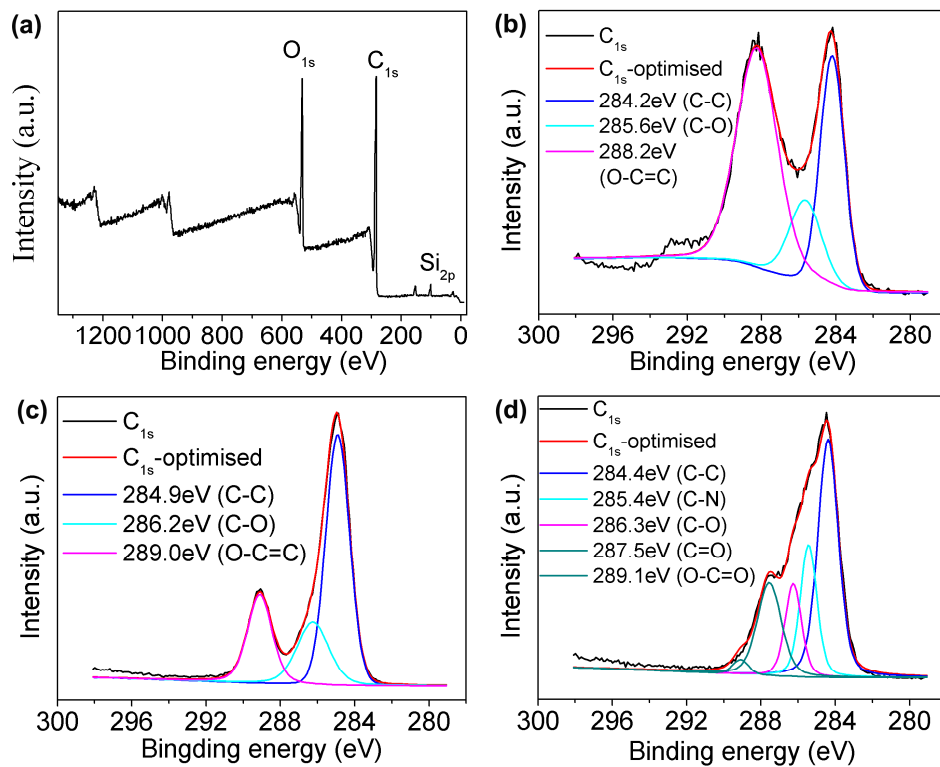
**Figure S6.** PL emission spectra ((a) excited at 360nm) and UV-Vis absorbance spectra (b) of EDA-CDs prepared with different ratio of amino groups to carboxyl groups (1 g CA, 10 ml H<sub>2</sub>O, microwave pyrolysis 2 min with different EDA amounts); the insets are (a) normalized PL spectra and (b) the magnified absorbance spectra from 300 nm to 400 nm. Each sample has the same absorbance value.



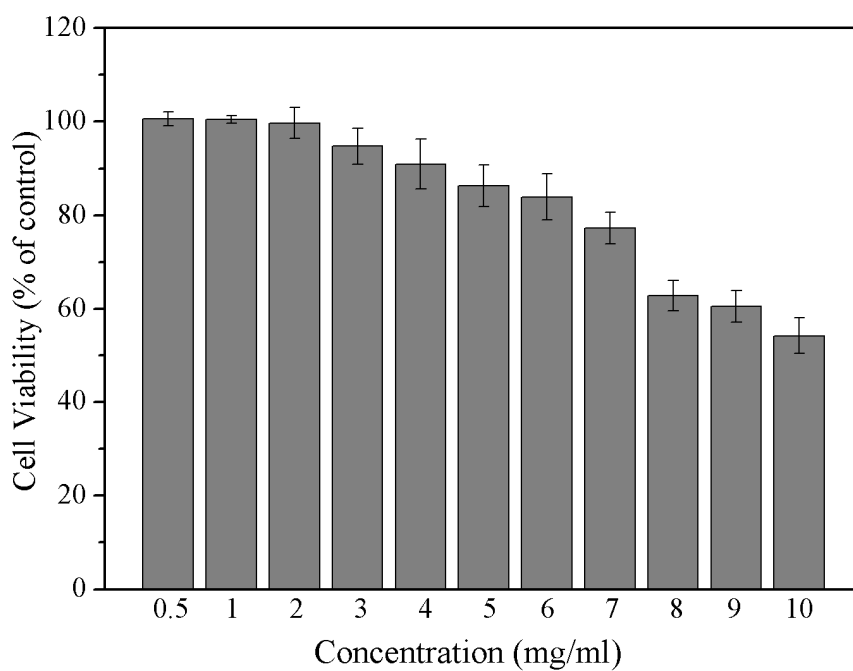
**Figure S7.** UV-Vis absorbance spectra of the different type of amines passivated CDs aqueous solution.



**Figure S8.** PL emission spectra (with progressively longer excitation wavelengths from 320 nm to 460 nm in 20 nm increments) of different type of amines passivated CDs (except EDA-CDs) (a) N-free CDs, (b) DEA-CDs, (c) TEA-CDs, (d) BDA-CDs (the inset is the normalized PL emission spectra).



**Figure S9.** XPS spectrum of N-free CDs (a), C<sub>1s</sub> spectrum of N-free CDs (b), TEA-CDs (c) and EDA-CDs (d).



**Figure S10.** Cytotoxicity testing results via a MTT assay. The values represent percentage cell viability (means%  $\pm$  SD, n = 3).