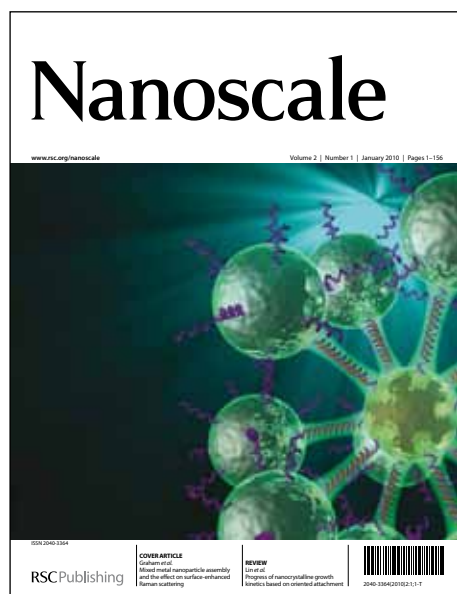


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ARTICLE

Nitrogen-doped, carbon-rich, highly-photoluminescent carbon dots from ammonium citrate

Cite this: DOI: 10.1039/x0xx00000x

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Received 00th January 2013,
Accepted 00th January 2013

DOI: 10.1039/x0xx00000x

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The synthesis of water-soluble nitrogen-doped carbon dots has received great attention, due to their wide applications in oxygen reduction reaction, cell imaging, sensors, and drug delivery. Herein, nitrogen-doped, carbon-rich, highly-photoluminescent carbon dots have been synthesized for the first time from ammonium citrate under hydrothermal condition. The obtained nitrogen-doped carbon dots possess bright blue luminescence, short fluorescence lifetime, pH-sensitivity and excellent stability at a high salt concentration. It is potential to be used for pH sensor, cell imaging, solar cells, and photocatalysis.

1 Introduction

Fluorescence is the emission of light by a substance that has absorbed light or other electromagnetic radiation. Fluorescent proteins,¹ organic dyes,² semiconductor quantum dots,³ modified carbon nanotubes,⁴ metal nanoclusters,⁵ silicon quantum dots,⁶ polymer dots,⁷ and nanodiamonds,⁸ are well-known fluorescent materials. Recently carbon dots (CDs) have drawn great attention of interdisciplinary scientists owing to their small sizes, excellent water solubility, strongly fluorescence, highly photostability, and non-toxicity.⁹⁻¹² Citric acid and its derivatives (nitrogen sources) are good candidates to synthesize nitrogen-doped (N-doped) CDs^{13,14} by bottom-up method. Giannelis *et al.*¹⁵ reported pyrolysis of citric acid and ethanolamine at 230 °C to obtain strongly photoluminescent N-doped CDs. They further carbonized tris(hydroxymethyl)-aminomethane and betaine hydrochloride at 180 °C to get luminescent and surface quaternized CDs.¹⁶ Chi *et al.*¹⁷ synthesized N-doped CDs by carbonizing citric acid at lower than 200 °C with branched polyethylenimine in one-pot step. In another work, Jiang *et al.*¹⁸ used hydrothermal method with sodium citrate and ammonium bicarbonate interacted at 180 °C to prepare N-doped CDs. However, carbon source (citric acid) and nitrogen source are used individually in the above-mentioned methods. In additions, some nitrogen-containing organic compounds¹⁹⁻²⁵ are also used to synthesize N-doped CDs, but they require a large amount of acid or base and tedious processes, which may result in restricting their applications.

There is no report about only a citric acid derivative containing both carbon source and nitrogen source to synthesize N-doped CDs. Ammonium citrate is a simple, low-cost, water-soluble compound, which can be used as an analytical reagent in determination of phosphate in fertilizer.²⁶ It can act as an “etching” reagent and influence a reductive reaction to convert Fe³⁺ to Fe²⁺ in the synthesis process of hematite hierarchical structures.²⁷ Leaching in ammonium

citrate has been extensively used to assess the fraction of water-soluble nickel compounds present in nickel producing and using workplace aerosols.^{28,29} To the best of our knowledge, ammonium citrate in present work is first used to synthesize N-doped CDs. Herein, we reported a novel and simple method using ammonium citrate, only a single compound, to synthesize N-doped, carbon-rich, highly-photoluminescent CDs. The as-synthesized CDs exhibit bright blue luminescence, short fluorescence lifetime, pH-sensitivity and excellent stability at a high salt concentration. The obtained N-doped CDs without any further surface modification could be functionalized with other active substances for specific applications in biomedical, sensor, solar cell, and catalysis.²¹

2 Experimental

2.1 Chemicals

All chemicals were analytical-reagent grade and purchased from Sinopharm Chemical Reagent Co., Ltd (Shanghai, China) without further purification. Deionized water with a resistivity of 18.1 MΩ·cm was used for all experiments.

2.2 Synthesis of N-doped CDs

Ammonium citrate (2 g, 8.2 mmol) was dissolved in 25 mL deionized water to form a transparent solution. The solution was sealed into a 50 mL Teflon equipped stainless steel autoclave followed by hydrothermal treatment at 160 °C for 6 h. The colour of the solution turned from colourless into dark blue in appearance. The obtained samples were concentrated into a small amount of liquid, transferred to a silica gel column, and eluted with methanol and dichloromethane to obtain bright blue fluorescent N-doped CDs.

2.3 Characterization

The morphologies of the samples were observed by transmission electron microscope (TEM, JEM-2100, JEOL, Japan). X-ray diffraction (XRD) analysis was carried out by D8 Advance, Bruker AXS Corporation, Germany. The photoluminescent (PL) spectra were performed through a fluorescent spectrophotometer (F-4600, Hitachi, Japan). The ultraviolet-visible (UV-Vis) absorption spectra were recorded by a Perkin-Elmer (USA) Lambda 950 UV-Vis-NIR spectrophotometer. Fourier transform infrared spectroscopy (FT-IR) was recorded on a VERTEC 70, Bruker, Germany spectrometer with room temperature deuterated L-alanine-doped triglycine sulfate (RT-DLaTGS) as a detector. The fluorescence lifetime was recorded by steady-state & time-resolved fluorescence spectrofluorometer (QM/TM/IM, PTL, USA). The X-ray photoelectron spectroscopy (XPS) was acquired with a Japan Kratos Axis UltraDLD spectrometer using a monochromatic Al K α source (1486.6 eV).

3 Results and discussion

N-doped, carbon-rich, highly-photoluminescent CDs were prepared through a one-pot hydrothermal treatment process of ammonium citrate at 160 °C for 6 h. The excess impurities, such as precursor residues and resulting small molecules, were removed by silica gel column. The resultant N-doped CDs were evaporated into solid states and then were dispersed into water for further characterization and use. Fig. 1A and B shows TEM image and particle size distribution by calculating the average sizes of 200 nanoparticles. The Gaussian fitting curve reveals that the average size of N-doped CDs is approximately 2.14 ± 0.34 nm. N-doped CDs are uniform in size and possess a nearly spherical shape.

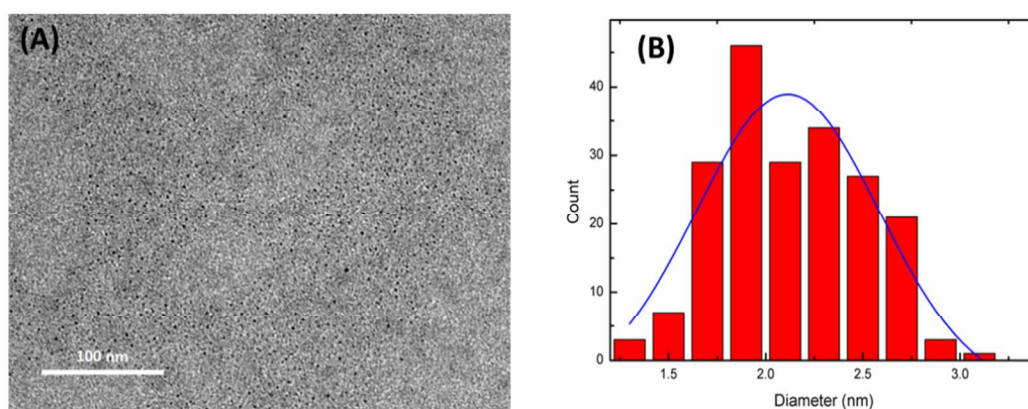


Fig. 1 (A) TEM image of N-doped CDs and (B) particle size distribution of N-doped CDs.

Fig. 2A shows bright blue fluorescence of N-doped CDs under UV light at 365 nm, and the absorption peaks at 235 and 327 nm, with a tail extending into the visible range. The two peaks are ascribed to π - π^* transition of C=C bond and n - π^* transition of C=O bond, respectively. Fig. 2B shows a detailed PL investigation with different excitation wavelengths. PL is one of most fascinating features of CDs. When the excitation wavelength is changed from 245 to 395 nm, the fluorescence emission peak at 437 nm remains unchanged. The excitation-independent PL behaviour are considered to be related to less surface defects and more uniform size of N-doped CDs, which is consistent with TEM observation. The wavelength-independent phenomenon makes the N-doped CDs useful for downconversion and upconversion cell imaging where the unwanted autofluorescence (e.g. tissue autofluorescence) can be avoided.²¹ The full width at half maximum (FWHM) of N-doped CDs calculated at 365 nm is about 66 nm. This good result is similar to that of CDs produced by electrochemical oxidation and microwave treatment.^{30,31} Comparing the second absorption peak in the UV-Vis spectrum with fluorescence peak in the PL spectra, a 110 nm red shift can be observed. Fig. 2C is the photoluminescent excitation (PLE) spectrum of CDs. It is found that two peaks are at 295 and 365 nm, respectively. This behaviour indicates that there are at least

two types of excitation energy trapped on the surface of N-doped CDs.²¹ When the excitation wavelength is 365 nm, the intensity of photoluminescent emission is maximum as shown in Fig. 2B. The PL properties of N-doped CDs may be classified to the anti-Stokes photoluminescence, in which the simultaneous absorption of two or more photons leads to the emission of light at a shorter wavelength than the excitation wavelength.³² As shown in Fig. S1, the fluorescence quantum yield of N-doped CDs was measured to be about 13.5% with quinine sulfate in 0.1 M sulfuric acid solution (quantum yield 54%) as the standard.³³ This quantum yield value is comparable to previous reports.^{11,31,34,35} The fluorescence lifetime of N-doped CDs was calculated to 10.6 ns according to fluorescence decay profile with excitation and emission wavelengths in 337 and 437 nm shown in Fig. 2D. It represents the fluorescence decay profile of N-doped CDs with single exponential decay kinetics. The short lifetime of N-doped CDs is indicative of the radiative recombination of the excitons giving rise to fluorescence.^{18,36} The suspension of CDs also shows extra-high stability. Even after being kept for 4 months in air at room temperature, it still exhibits a transparent appearance and strong PL, which offers another advantage for its future applications.

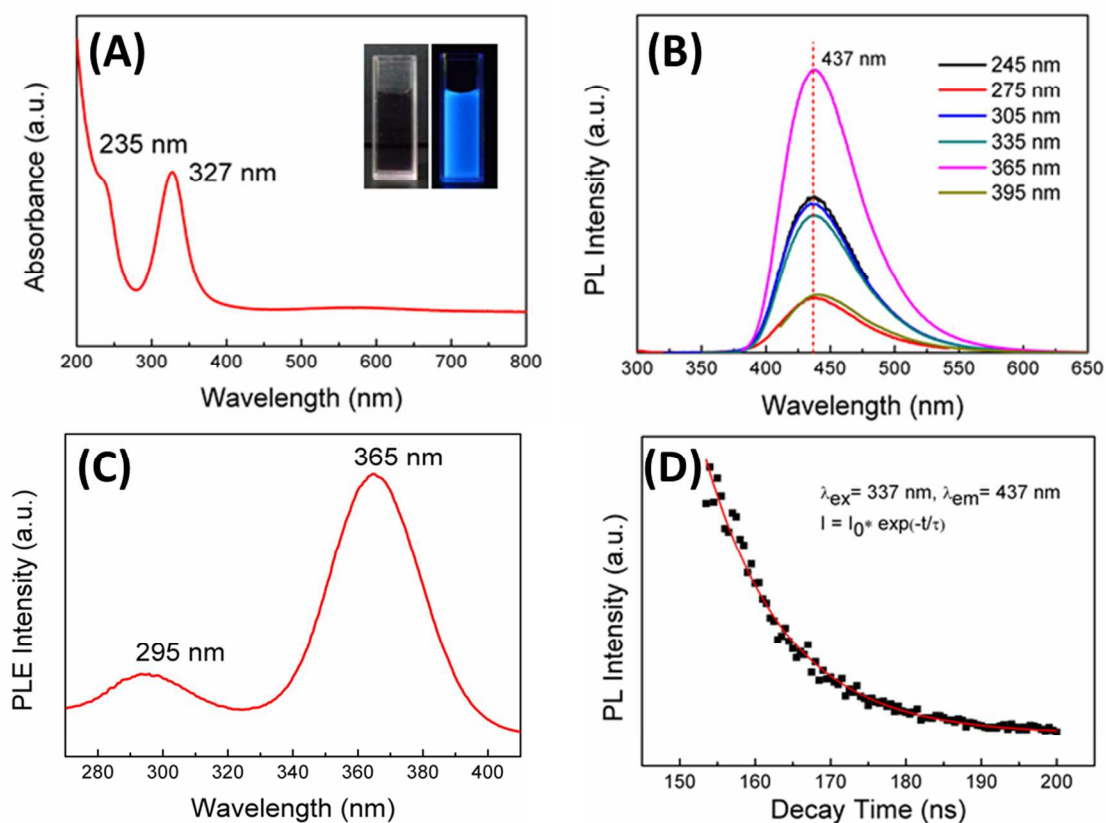


Fig. 2 (A) UV-Vis absorption spectrum of N-doped CDs, inset are photos of CDs under sunlight (left) and irradiated by 365 nm light (right); (B) PL spectra recorded for progressively longer excitation wavelengths from 245 to 395 nm in a 30 nm increment; (C) PLE spectrum of N-doped CDs with emission wavelength at 437 nm; (D) Fluorescence decay profile of N-doped CDs.

The structure and composition of N-doped CDs were characterized by FT-IR spectroscopy. As shown in Fig. 3A, the synthesized N-doped CDs show main absorption band of O-H stretching vibration at 3434 cm^{-1} and the amine bond from C=O stretching at 1625 cm^{-1} in the spectrum. The peaks at 2920 and 2851 cm^{-1} are related to the C-H bond stretching vibrations. The characteristic stretch band of the amine III C-N bond is at 1461 cm^{-1} . The bending vibration of C-O bond and stretching peak of C-O-C bond appear at 1400 and 1085 cm^{-1} , respectively.²² The FT-IR spectrum of reactant ammonium citrate is also shown in

Fig. 3A. Two different chemical environment's carboxyl groups at 1592 and 1719 cm^{-1} in ammonium citrate were changed into one C=O stretching peak under hydrothermal condition. N-doped CDs with amounts of hydroxyl and carboxyl groups on their surface possess an excellent water-solubility, which can be used in cell imaging and drug delivery. XRD pattern of N-doped CDs in Fig. 3B demonstrates that (002) interlayer spacing distance of CDs is 3.3 \AA , which is similar to that of the bulk graphite, showing good crystallization characteristics.

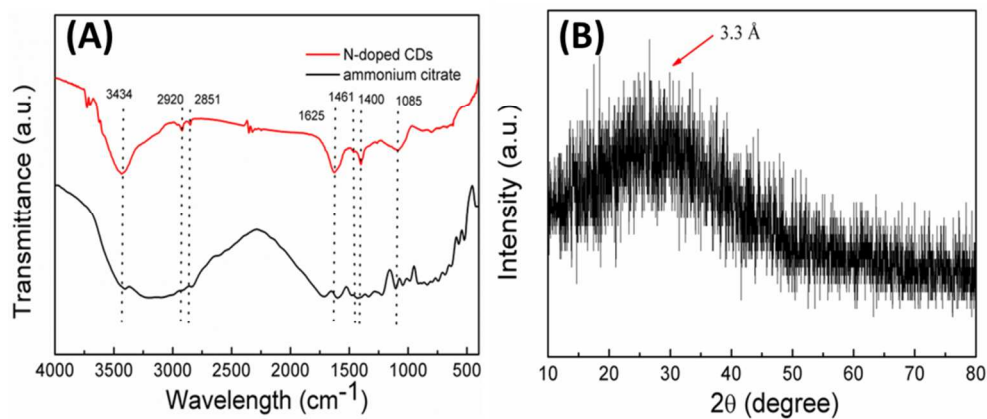


Fig. 3 (A) FT-IR spectrum of N-doped CDs; and (B) XRD pattern of N-doped CDs.

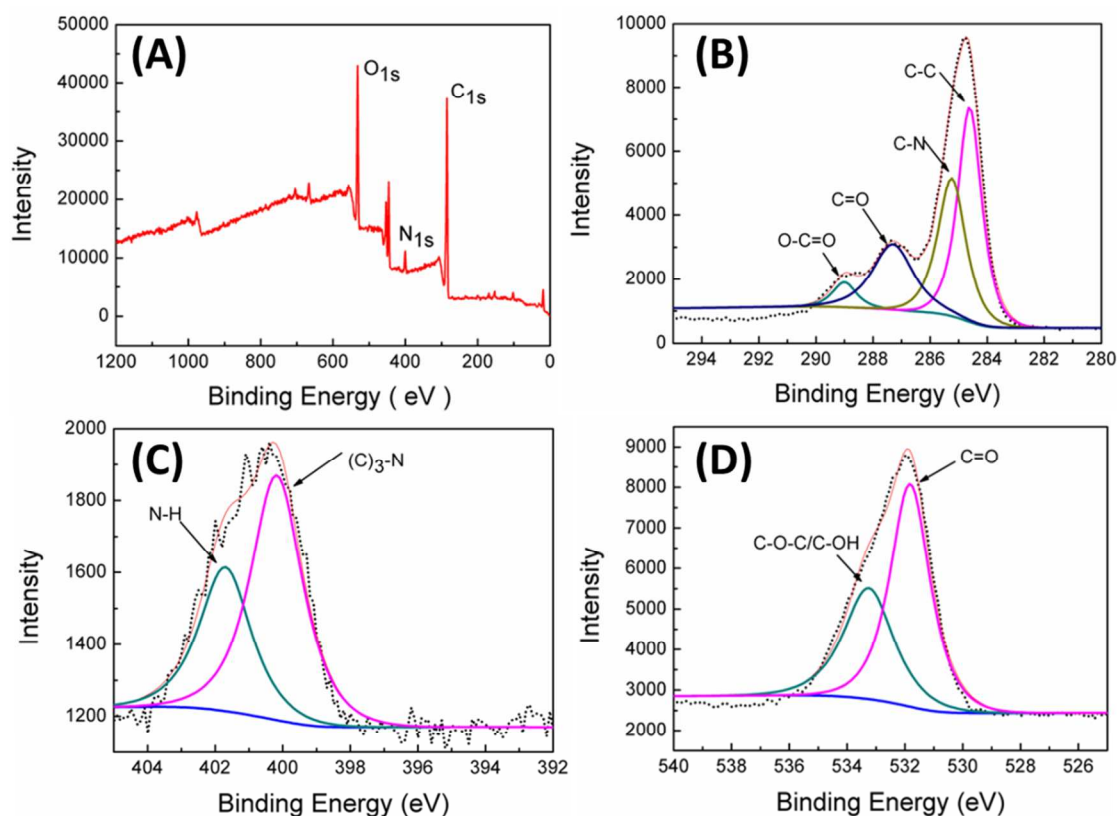


Fig. 4 XPS survey scan of N-doped CDs on indium foil. (A) XPS scanning spectrum shows three major peaks of carbon, nitrogen and oxygen. XPS high resolution survey scan of (B) C1s, (C) N1s and (D) O1s region of CDs

XPS measurements were performed for the surface element analysis of N-doped CDs. The XPS spectrum shown in Fig. 4A clearly reveals that carbon, nitrogen and oxygen are present at the surface of N-doped CDs. In the expanded XPS spectra, the C1s peaks at 284.6, 285.2, 287.3, and 289.0 eV shown in Fig. 4B can be assigned to carbon in the form of C-C (sp^3), C-N (sp^3), C=O (sp^2), and O-C=O (sp^2), respectively.^{37,38} The N1s peaks at 400.2 and 401.7 eV shown in Fig. 4C indicate that nitrogen exists mostly in the form of (C)₃-N (sp^3) and N-H (sp^3), respectively.³⁹

This verifies the synthesized CDs are nitrogen-doped CDs. The O1s peaks at 531.8 and 533.3 eV shown in Fig. 4D are associated with oxygen in the states of C=O and C-OH/C-O-C, respectively.³⁹ As shown in Table S1, 5.35 % of nitrogen and 68.75 % of carbon indicate that CDs are the nitrogen-doped and carbon-rich. Attempts to obtain Raman spectra were unsuccessful due to the intense fluorescence of CDs, resulting in covering the characteristics of the Raman signal.^{40,41}

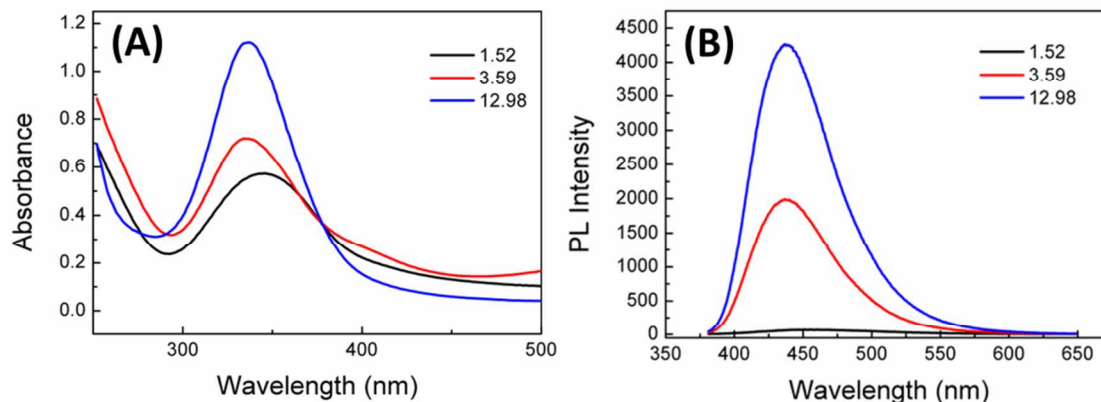


Fig. 5 (A) UV-Vis spectra of N-doped CDs and (B) PL spectra of N-doped CDs changed at different pH values.

We also investigated the absorbance change and photoluminescence response of N-doped CDs at different pH

value adjusted by adding NaOH and HCl. In Fig. 5A and B, N-doped CDs show lower absorbance and photoluminescence

intensity at lower pH, respectively. It is found that both the absorption and emission intensity of our obtained CDs are sensitive to the pH value. Meanwhile, the colour of the CD aqueous solution is darkened from light yellow to yellow-brown with the increase of pH value. The pH dependent properties may be attributed to aggregation of the synthesized CDs by their surrounding carboxyl acids at lower pH, resulting in the fluorescence quenching.¹⁸ This phenomenon also confirms that the hydrophilic carboxyl acid groups are present on the surface of N-doped CDs. The above results show that the CDs have potential as a pH sensor for pH measurement.

The salt effect of N-doped CDs on the fluorescent properties have also been investigated. In Fig. 6, when KCl concentration is lower than 0.6 mol/L, PL intensity increases sharply with the increase of KCl concentration. However, PL intensity remains almost unchanged as KCl concentrations increase from 0.6 to 2.0 mol/L. The influence of salt concentration on the PL intensity is attributed to the fact that the ionic strengths of salt can effectively control the aggregation of N-doped CDs and individually separate N-doped CDs under salt's interference. This demonstrates that the obtained N-doped CDs can be used in extreme environmental conditions because they can keep strong photoluminescence in higher salt concentration. As shown in Fig. 7, the formation mechanism of N-doped CDs might be that ammonium citrate molecules are firstly aromatized by intramolecular dehydration with a "polymerization" process. Next the intermediates are carbonized to form carbon nuclei of N-doped CDs that is composed of sp^2 carbon as revealed by XRD pattern. The carbon nuclei subsequently form nanocrystalline N-doped CDs with hydrophilic functional groups (hydroxyl group, carboxyl group, and so on). These groups located at the surface of the N-doped CDs act as a self-passivation layer to facilitate the water-solubility of N-doped CDs as well as the efficient photoluminescence properties.

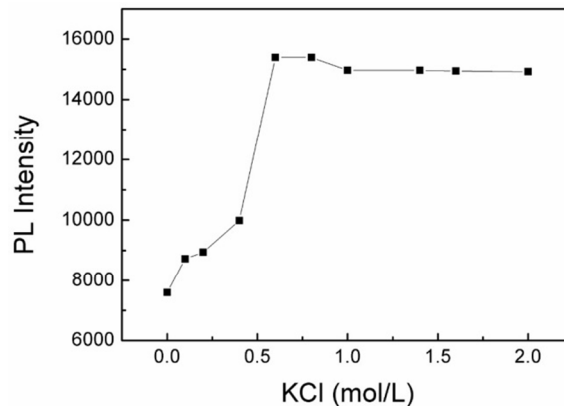


Fig. 6 Salt effect of KCl concentration on PL intensity.

4 Conclusions

In summary, we have developed a novel, green, and one-pot hydrothermal method to synthesize nitrogen-doped, carbon-rich, highly-photoluminescent CDs in a large scale only using an ammonium citrate precursor serving as both carbon source and nitrogen source, which is different from traditional synthesis method using two reactants. More importantly, the stable and tunable obtained N-doped CDs have excellent water solubility, favourable ionic strengths from salt effect, and unique PL properties such as pH-dependent luminescence. Therefore, combining a convenient synthesis route with attractive PL properties, CDs should have promising potential for serving as a candidate for a new class of pH sensor, biomedical sensors for physiological condition, fluorescence probes, cellular imaging as well as solution state optoelectronics devices.

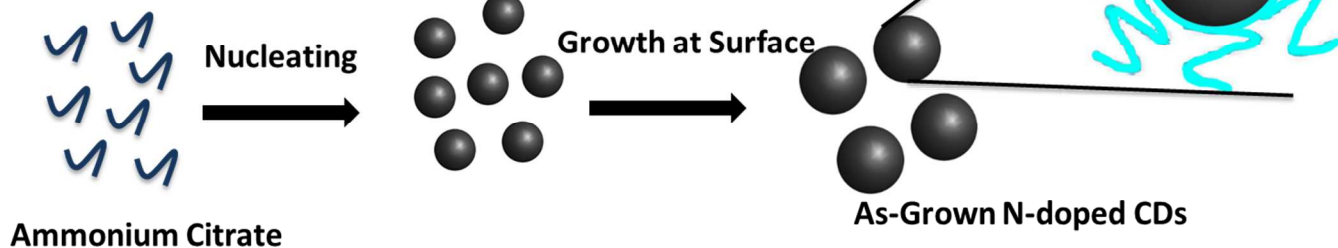


Fig. 7. Schematic diagrams of the formation mechanism of N-doped CDs.

Acknowledgements

This work was supported by the National High-Tech R & D Program of China (863 program, 2011AA050504), National

Natural Science Foundation of China (21171117 and 61376003), Program for New Century Excellent Talents in University (NCET-12-0356), Shanghai Science and Technology Grant (12JC1405700 and 12nm0503800), Shanghai Natural Science

Foundation (13ZR1456600), Shanghai Pujiang Program (no. 11PJD011), the Program for Professor of Special Appointment (Eastern Scholar) at Shanghai Institutions of Higher Learning, and Medical-Engineering Crossover Fund (YG2012MS40) of Shanghai Jiao Tong University. We also acknowledge the analysis support from the Instrumental Analysis Center of Shanghai Jiao Tong University.

Notes and references

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Electronic Supplementary Information (ESI) available: The curve of photoluminescence and absorbance of N-doped CDs and quinine sulfate, the table of XPS detailed information. See DOI: 10.1039/b000000x/

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N-doped carbon dots were synthesized by ammonium citrate as a solo precursor under hydrothermal condition.