

Green Synthesis of Carbon Dots from Lemon Juice with Precursor Ratio-Dependent Emissions

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Abstract

Carbon dots (CDs) are fluorescent nanomaterials with great potential for applications in sensing and imaging. The study explores the synthesis of CDs using a simple, low-cost, and rapid household microwave-assisted approach, utilizing lemon juice and urea as precursors. By modulating the precursor ratio, we obtained two types of CDs with different optical properties: green-emitting fluorescent CDs (g-CDs) and blue-emitting CD (b-CDs). Both particles showed quantum yields of approximately 10%. The synthesis process, conducted under solvent-free and ambient-pressure conditions, demonstrates an environmentally benign and sustainable route compared to conventional hydrothermal methods. Structural and optical characterizations confirmed that precursor ratio control directly influenced the emission behavior of the CDs. Preliminary investigations further revealed that the g-CDs exhibited selective and sensitive fluorescence quenching toward Hg^{2+} ions, with a limit of detection (LOD) of 0.054 ppm, indicating their promise as sustainable and cost-effective probes for heavy metal detection. This study highlights the potential of green-synthesized carbon dots for environmental sensing applications, contributing to developing simple early-warning tools for pollution monitoring

Keywords: Carbon dots, fluorescence, metal ion sensor, nanomaterial.

1. Introduction

Carbon dots (CDs) are a new class of carbon-based nanomaterials with sizes smaller than 10 nm [1-3]. CDs possess several superior properties compared to traditional semiconductor quantum dots and organic dyes, including low or no toxicity, good water solubility, excellent photostability, and the ability to tune their emission spectra through synthesis and surface modification. These properties enable CDs to be applied in various fields such as optoelectronics, biomedicine, sensing, and catalysis [1, 4].

To further improve the unique properties of CDs and enhance their chemical binding capabilities, CDs are often doped with heteroatoms, including nitrogen, phosphorus, sulfur,... [5, 6]. Among these, nitrogen is the most effective element in enhancing quantum yield and modifying electronic properties. The citric acid and urea system is one of the most typical reaction systems that has been shown to enhance the luminescent effect and alter the electronic properties of N-doped CDs [7]. Previous studies have demonstrated that thermal reactions of citric acid and urea produce molecular fluorophores such as citrazinic acid and 4-hydroxy-1H-pyrrolo[3,4-c]pyridine-1,3,6(2H,5H)-trione (HPPT) derivatives that govern the photoluminescence of carbon dots [8-10]. Specifically, precursor ratios and reaction conditions strongly determine the emission color and optical properties of CDs [10, 11]. These findings have established that the optical properties of CDs derived

from the citric acid–urea system are closely related to the chemical pathways occurring during synthesis [1, 12, 13].

One of the most important applications of CDs is as fluorescent sensors for heavy metal ions detection, which are serious environmental pollutants and highly toxic [14, 15]. In particular, mercury (Hg^{2+}) is classified by the World Health Organization (WHO) as one of the top toxic chemicals. Therefore, the development of sensitive, selective, and cost-effective sensors for Hg^{2+} detection is needed. CDs can function as fluorescent sensors through a fluorescence quenching mechanism, in which the interaction between the CDs and the target metal ion reduces the fluorescence emission intensity of the CDs [16, 17].

N-doped carbon dots are typically synthesized from carbon and nitrogen sources using methods such as pyrolysis, electrolysis [18], sonication, and hydrothermal [1, 19, 20]. However, these methods have some disadvantages such as high energy consumption, high temperatures, long synthesis times, high costs, and sometimes the use of toxic solvents. Aiming for sustainable development, green synthesis has emerged as a promising approach by designing chemical processes that minimize or eliminate the use and generation of hazardous substances, while optimizing the efficiency of raw material use. The green synthesis of carbon dots from natural materials or biological waste, combined with environmentally friendly reaction

techniques such as microwave treatment, further highlights the advantages of safety and sustainability [21, 22]. Among natural sources, lemon juice is the fruit juice with the highest citric acid content, ranging from 39 to 48 g/L [23]. Therefore, lemon juice can be considered an abundant, human-friendly source of citric acid that can be used to synthesize CDs.

In this study, nitrogen-doped carbon dots were synthesized using a household microwave-assisted method, employing lemon juice as the carbon source and urea as the nitrogen source. By adjusting the urea ratio, two types of CDs with different emission properties were obtained: green-emitting carbon dots (g-CDs) and blue-emitting carbon dots (b-CDs). Building on the established understanding of fluorophore formation in citric acid–urea systems, this study demonstrates a simple microwave-assisted route using lemon juice as a natural precursor and examines how precursor stoichiometry affects the resulting emission characteristics. The optical and structural properties of the obtained CDs were characterized, and their potential application for selective metal ion sensing was preliminarily evaluated.

2. Materials and Methods

2.1. Materials and Chemicals

Fresh lemons were obtained from local markets, and urea was purchased from GHTECH China; Quinine from Sigma-Aldrich; Fluorescein; 0.1M H₂SO₄; 0.1M NaOH; 0.001M EDTA. All chemicals were used without further purification. Deionized water was used in all experiments.

2.2. Methods

2.2.1. Synthesis of carbon dots and carbon dots

The CDs were synthesized according to the following procedure: 10 ml of lemon juice (pH 2-3), obtained after centrifugation at 5000 rpm for 10 minutes to remove all pulp, was mixed with 0.2 g or 2 g of urea for synthesizing b-CDs and g-CDs, respectively. The mixture was dissolved in a 100 ml beaker, then heated in a Sharp 800W household microwave oven for 5 minutes with a power output of approximately 400W. During the synthesis time, the mixture changed from a colorless liquid state to a light brown solid state and finally a dark brown color. The solid was then dissolved in 20 ml of deionized water, sonicated for 60 minutes, and centrifuged three times at 12000 rpm for 60 minutes to remove most of the aggregates. The CD solution was further purified by dialysis using a 3500 Da dialysis membrane for 72 hours at room temperature. The solution obtained outside the dialysis bag was stored in a sealed container at 4 °C in the dark for subsequent measurements.

2.2.2. Characterization

A Biochrom Libra S60 UV-Vis spectrophotometer was used to analyze the UV-Vis absorption spectra of the CDs. The fluorescence emission spectra of the CD solutions were measured on a Perkin-Elmer FL8500 molecular fluorescence spectrophotometer with a Single Cell Holder. The size of the CDs was analyzed by Scanning Electron Microscopy (SEM) and (Transmission Electron Microscopy (TEM) images. For SEM and Energy-Dispersive X-ray Spectroscopy (EDX) analysis, the carbon dot solutions were drop-cast onto aluminum stubs, dried, and subsequently sputter-coated with a thin layer of gold to prevent charging effects.

2.2.3. Quantum yield

The relative quantum yields (Q_y) of nanomaterials were determined based on following equation:

$$Q_{y(CDs)} = Q_R \left(\frac{Grad_{CDs}}{Grad_R} \right) \left(\frac{\eta_{CDs}^2}{\eta_R^2} \right) \quad (1)$$

where $Q_{y(CDs)}$ and Q_R are the quantum yield of CDs and the reference, respectively. $Grad$ is the gradient from the plot of integrated fluorescence intensity versus absorbance and η is the refractive of the solvent [24]. In which the Q_y of g-CDs and b-CDs were determined by using fluorescein in 0.1M NaOH and quinine sulfate in 0.1M H₂SO₄ as a reference, respectively.

2.2.4. Sensing of Hg²⁺

Stock solutions of metal salts including Ag⁺, Al³⁺, As²⁺, Cd²⁺, Cu²⁺, Fe³⁺, Hg²⁺, Mg²⁺, Mn²⁺, Ni²⁺, Pb²⁺, and Zn²⁺ were at a concentration of 1000 ppm. The g-CD and b-CD solutions were kept at an absorbance of less than 0.1 at 410 nm and 340 nm, respectively. The 1.8 mL CDs solution was mixed with 0.1 mL of a metal salt solution, and the fluorescence spectrum of the mixture was recorded after 30 minutes of incubation.

For Hg²⁺ sensing experiment, 1.5 mL of g-CDs solution was mixed with different volumes of a 10 ppm Hg²⁺ stock solutions and deionized water, giving a volume of 2.0 mL and final Hg²⁺ concentrations ranging from 0.5 to 5.0 ppm. The mixtures were thoroughly shaken and incubated at room temperature for 30 minutes to ensure binding equilibrium. Photoluminescence emission spectra were subsequently recorded in the range of 415–700 nm with an excitation wavelength of 410 nm. The limits of detection (LOD) and limit of quantification (LOQ) were calculated using the following equations:

$$LOD = 3.3\sigma/S \quad (2)$$

$$LOQ = 10\sigma/S \quad (3)$$

here, σ represents the standard deviation of the analytical response, which was obtained from ten replicate measurements of the blank solution (1.5 mL g-CDs and 0.5 mL deionized water), while S corresponds to the slope of the linear calibration curve relating fluorescence response to Hg²⁺ concentration.

3. Results and Discussion

3.1. Optical and Structural Properties of Carbon Dots

In this study, adjusting the reaction conditions while keeping the added urea ratio constant produced two types of nanoparticles. The optical properties of the g-CDs and b-CDs are shown in Fig. 1. The g-CDs have a maximum absorption peak at 280 nm, which is attributed to the characteristic π - π^* electronic transition of the C=C bond in the aromatic ring of the carbon core. Further observation in the visible region reveals an obvious difference between the two types of CDs: b-CDs

exhibit an absorption peak at 340 nm, whereas g-CDs have a maximum absorption peak at 410 nm. The photoluminescence emission spectra of b-CDs and g-CDs when excited at 340 nm and 410 nm, respectively, also show a distinct difference: b-CDs emit at 440 nm, resulting in a blue color, while g-CDs emit at 525 nm, resulting in a green emission. Interestingly, as the excitation wavelength increased, the emission peaks of both g-CDs and b-CDs exhibited a slight red shift, which suggests the co-existence of specific molecular fluorophore with the synthesized particles.

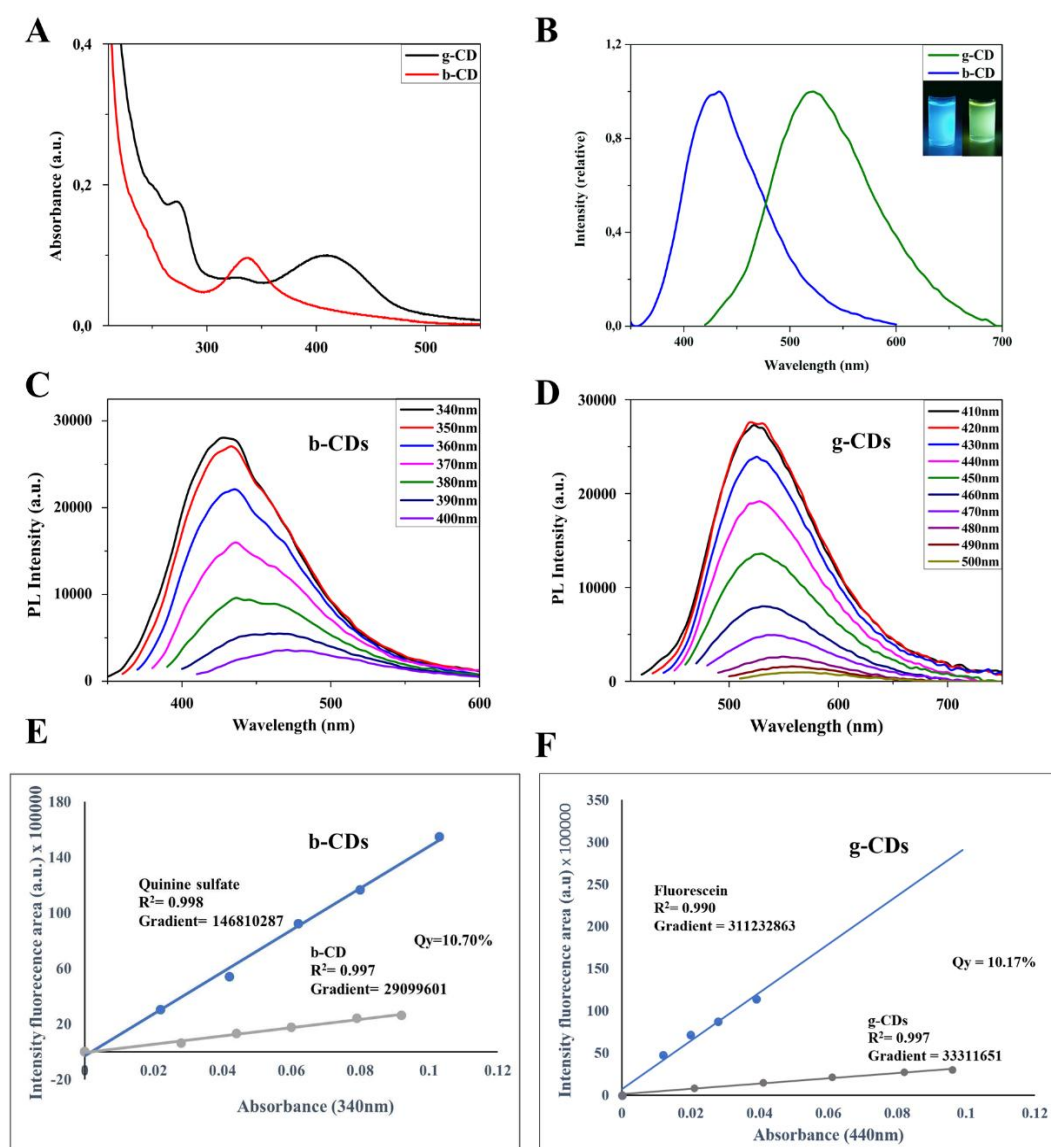


Fig. 1. Spectroscopic characterization of the as-prepared b-CDs and g-CDs: (1A) UV-Vis absorption spectra in aqueous dispersion; (1B) Photoluminescence (PL) spectra of b-CDs and g-CDs obtained under their respective optimal excitation conditions. PL emission profiles of (1C) b-CDs and (1D) g-CDs recorded at varying excitation wavelengths. Linear fitting of integrated PL intensity versus absorbance for the relative quantum yield calculations of (1E) b-CDs, and (1F) g-CDs.

Table 1. Comparison of the optical properties of g-CDs and b-CDs

Type of CDs	Precursor Ratio	λ_{abs} (max)	λ_{em} (max)	Qy
g-CDs	10 ml lemon juice: 2g urea	410nm	525nm	10.17%
b-CDs	10 ml lemon juice: 0.2g urea	340nm	440nm	10.70%

Fourier-Transform Infrared (FTIR) spectroscopy was utilized to determine the surface functional groups. The results demonstrated a remarkable similarity, as both types of CDs exhibited strong absorption peaks in the 2900–3600 cm^{-1} region, characteristic of the $-\text{COOH}$, N-H , and $-\text{NH}_2$ functional groups (Fig. 2). A medium absorption peak appeared in the 1600–1700 cm^{-1} region, characteristic of the C=O stretching vibration. Strong absorption was also observed in the fingerprint region. The FTIR results indicated that the type of functional groups on the surface of both CDs remained unchanged when the urea ratio was varied during synthesis. Therefore, the difference in photoluminescence emissions in both types of CDs could be due to the difference in the carbon core structures, instead of functional groups on CDs' surfaces.

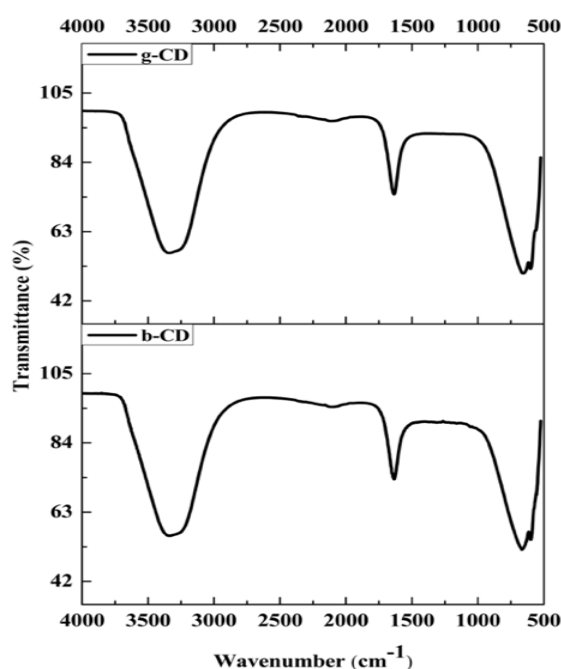


Fig. 2. FTIR spectra of g-CDs and b-CDs

The morphological and structural characteristics of the g-CDs were determined by SEM and TEM (Fig. 3). Obviously, the g-CDs exhibit non-uniform shapes and sizes. Although the particle size of the synthesized g-CDs is distributed within a diameter range of 5–25 nm, the g-CDs tend to agglomerate into particle clusters. This agglomeration phenomenon is common for carbon dot materials synthesized from natural precursors [25]. This morphological result is

considerably larger than reported values for other CDs; however, it also suggests potential for further optimization processes.

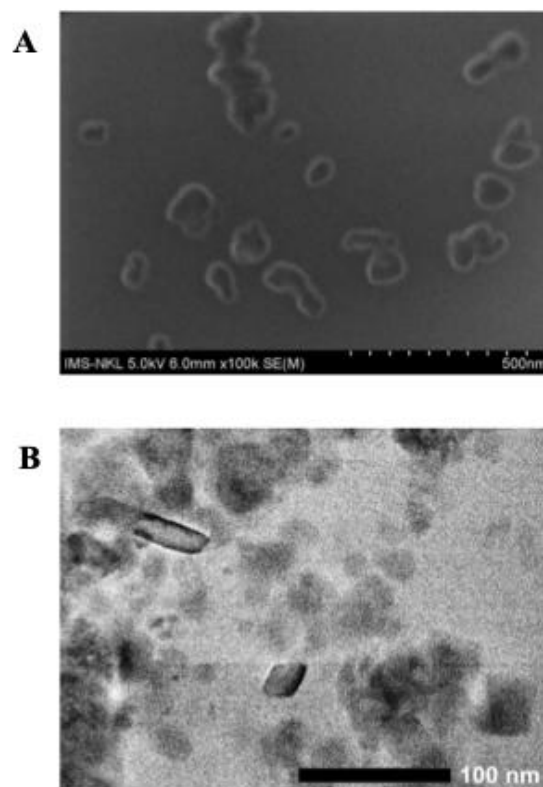


Fig. 3. (3A) SEM and (3B) TEM images of g-CDs

To investigate the elemental composition and validate the successful heteroatom doping of the synthesized materials, Energy-Dispersive X-ray (EDX) spectroscopy was performed. The EDX spectra reveals that both g-CDs and b-CDs are predominantly composed of carbon, nitrogen, and oxygen. Notably, the g-CDs exhibit a significantly higher nitrogen content (32.36 atomic%) compared to the b-CDs (19.67 atomic%), while the b-CDs display a higher oxygen content (32.27 atomic % compared to 19.67 atomic% in g-CDs) (Fig. 4). This distinct variation in elemental composition directly correlates with the precursor ratios used during synthesis. The higher nitrogen incorporation in g-CDs, facilitated by a greater urea concentration, likely promotes the formation of specific nitrogen-rich molecular fluorophores and surface states, thereby driving the observed red-shift in emission from blue to green.

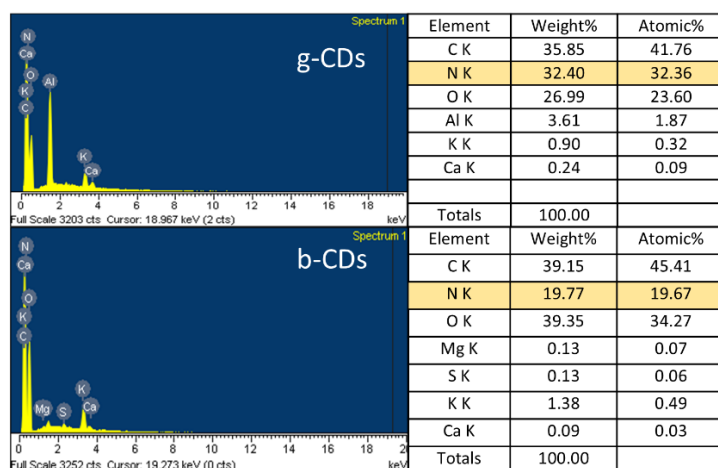


Fig 4. EDX results of g-CDs and b-CDs

3.2 Selective Sensing Ability for Hg^{2+} Ions

To evaluate the potential application of the synthesized CDs as a fluorescent probe, the selectivity of the material was investigated in the presence of various metal ions. The results demonstrate the high selectivity of the g-CDs for Hg^{2+} ions, whereas the photoluminescence of b-CDs changed significantly when adding both Fe^{3+} and Hg^{2+} (Fig. 5). Further studies were focused on the selectivity of the g-CDs

for Hg^{2+} ions. The fluorescence quenching efficiency (F_0/F) exhibited a good linear relationship with the Hg^{2+} concentration in the range of 0–5.0 ppm (Fig. 5D). The LOD and LOQ were calculated to be 0.054 ppm and 0.181 ppm, respectively. These results suggest that the as-prepared g-CDs serve as a sensitive and selective fluorescent probe, demonstrating great potential for the analysis and quantification of trace mercury ions.

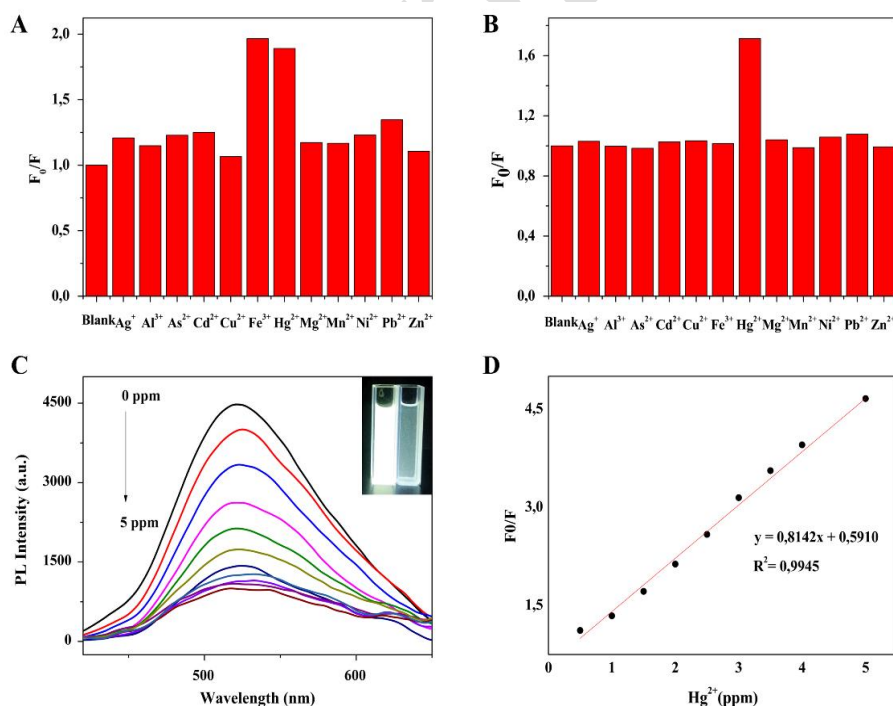


Fig. 5. Selectivity of the b-CDs (5A) and (5B) g-CDs sensing towards various metal ions, (5C) the fluorescence spectrum of g-CDs corresponding to different concentrations of Hg^{2+} ions (with inset showing the g-CDs without and with 5ppm Hg^{2+}), (5D) linear relationship of F_0/F and the Hg^{2+} concentration in the range of 0.5–5.0 ppm

3.3 Discussion

3.3.1. Formation of the two Synthesized Carbon Dots

This study demonstrates a green and efficient microwave-assisted synthesis of CDs from lemon juice and urea and examines that precursor ratio control enables simple tuning of optical properties: low urea produced b-CDs, while higher urea yielded g-CDs in a complex natural system. To preserve the facile and environmentally benign nature of the proposed green synthesis, no external pH adjustments were performed before microwave irradiation. However, lemon juice contains numerous organic compounds that can yield additional products, necessitating further purification.

The formation of CDs from citric acid and a nitrogen source through hydrothermal treatment is a multi-step, complex process. Initially, condensation and cyclization occur between carboxylic acid groups of citric acid and the amine groups of the nitrogen source, forming small fluorophore molecules [26, 27]. Subsequent heating drives polymerization and carbonization, which constructs the final carbon core of the CDs. The resulting product is therefore highly dependent on both the nitrogen precursor and the reaction conditions. For example, ethylenediamine directly forms a blue fluorophore (imidazo[1,2-a]pyridine-7-carboxylic acid, 1,2,3,5-tetrahydro-5-oxo or IPCA), whereas urea first decomposes into ammonia, which then reacts with citric acid to produce citrazinic acid [8, 26, 28]. Furthermore, slight changes in conditions are critical; a change from a sealed aqueous environment (yielding blue-emitting CDs from citrazinic acid) to a solvent-free environment (yielding green-emitting CDs from the fluorophore HPPT) produces entirely different fluorophores [10, 29]. Based on previous studies, the blue-emitting CDs (b-CDs) in this work are assumed to have optical properties similar to citrazinic acid (absorption ~ 340 nm, emission ~ 440 nm), while the

green-emitting CDs (g-CDs) resemble HPPT (absorption ~ 410 nm, emission ~ 520-540 nm). The formation of the as-synthesized CDs (Fig. 6) is therefore proposed to depend on the nitrogen source concentration. Increased nitrogen content may promote the formation of different molecular fluorophores, leading to distinct emissive species and variations in the carbon core structure, while the surface functional groups remain largely unchanged. Further studies are needed to confirm this mechanism.

3.3.2. Sensing Hg^{2+} by g-CDs

Previous studies have demonstrated that the fluorescence quenching of carbon dots is primarily driven by specific coordination interactions between Hg^{2+} ions and electron-rich surface functionalities, such as hydroxyl, carbonyl, and amino groups [30-33]. The quenching behavior observed in our g-CDs is probably governed by a similar pathway; however, detailed mechanistic elucidation remains a subject for our future investigations.

As summarized in Table 2, the as-prepared g-CDs exhibit a reasonable limit of detection (0.054 ppm or 269 nM) and a working linear range (0.5–5 ppm) for Hg^{2+} sensing. While this analytical performance is moderate and comparable to existing standard carbon-based probes, it remains sufficient for preliminary screening purposes. The primary advantage of our system lies not in unprecedented sensitivity, but rather in its sustainable and time-efficient synthetic methodology. By utilizing abundant natural lemon juice in a rapid microwave-assisted approach, this method avoids high energy consumption and complex chemical precursors typical of conventional hydrothermal treatments. Therefore, the g-CDs can serve as a simple, cost-effective, and eco-friendly tool for basic heavy metal detection.

Table 2. Comparison of different fluorescent nanomaterials of determination of Hg^{2+}

Fluorescent probbe	Precursor source	LOD (nM)	Linear range (nM)	Ref.
NCDs (hydrothermal)	Citric acid	226	10^3 – 1.2×10^4	[34]
N/S-CDs (hydrothermal)	Gardenia fruit	320	0.2 – 2×10^4	[35]
N-CDs (hydrothermal)	Highland barley and ethanediamine	480	1×10^4 – 16×10^4	[36]
N, S/C-Dots (microwave)	Citric acid/urea/L-cysteine	2000	0 – 4×10^4	[37]
CDs (microwave)	Lemon juice and urea	269	2.49×10^3 – 2.49×10^4	This work

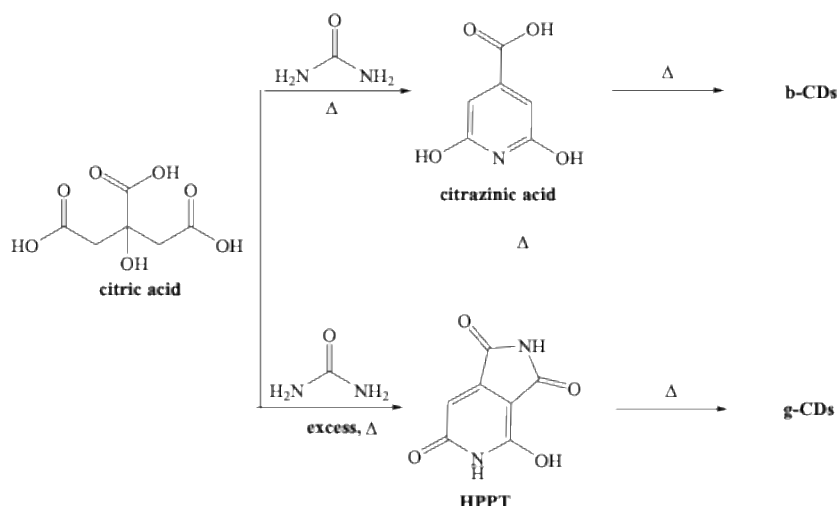


Fig. 6. Proposed formation of b-CDs and g-CDs from lemon juice with citric acid as a primary precursor

4. Conclusion

In summary, we have performed a green, rapid, and low-cost microwave-assisted protocol for fabricating fluorescent carbon dots from lemon juice and urea. By tuning the precursor ratio, we were able to obtain two types of CDs with distinct optical properties. This finding demonstrates that precursor ratio control offers a straightforward strategy to tune CD emission. More importantly, the g-CDs were demonstrated to be a high-potential fluorescent probe with sensitivity and selectivity for the detection of Hg^{2+} ions. The sensor displayed a good linear response in the 0.5–5 ppm concentration range with a *LOD* of 0.054 ppm. This highlights the potential of sustainable, natural-source nanomaterials for environmental sensing applications. Future work should focus on purification improvement, optimization of sensitivity, including pH effect, other metals interference, and real-sample testing to advance practical deployment.

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Declaration of competing interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work.

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