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Green synthesis of carbon quantum dots and its composite with Ag/chitosan using citrus fruit extracts

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ABSTRACT

Quantum dots are small semiconductors less than 50 nanometers in size, exhibiting unique optical and electronic properties due to quantum mechanics. Carbon quantum dots (CQDs) are particularly popular due to their size-dependent fluorescence, non-toxicity, biocompatibility, and ease of access. CQDs have significant potential applications in various chemical fields. In this study, CQDs were successfully synthesized using a green, one-step hydrothermal method involving citrus (tangerine). The synthesized CQDs were characterized using UV-VIS, XRD, IR, and TEM methods. The CQDs were then reacted with silver oxide to form CQD/Ag composite, and chitosan polymer was used to synthesize carbon quantum dots/Ag/Chitosan (CQD/Ag/Chit) bio-composite. The product was characterized using ultraviolet and infrared spectroscopy to confirm the bonds. This composite has potential applications in biological fields, as chitosan and silver have strong antibacterial properties, and their combination with active CQDs can enhance these properties.

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1. Introduction

Carbon, as the fundamental element in organic chemistry, has a profound impact on human daily life. Furthermore, carbon's versatility in combining with other elements makes it one of the most important and valuable elements in the pharmaceutical and food industries (Demming, 2010; Egbedina et al., 2022). Carbon quantum dots (CQDs) are the smallest members of the carbon family and have emerged as a powerful alternative to numerous fluorophores due to their exceptional electrical, optical, and physical properties (Huang et al., 2019b; Tajik et al., 2020; Kumar et al., 2022; Soumya et al., 2023; Zhao et al., 2023). In addition, CQDs are a desirable alternative to conventional materials in a wide array of applications, spanning from biological to industrial fields. This is due to their advantageous properties compared to other materials, such as extremely low cytotoxicity and excellent biocompatibility (Li et al., 2020; Tungare et al., 2020; Nammahachak et al., 2022; Mat Zaid et al., 2023). The luminescence of CQDs exhibits promising optical properties that function effectively even in solid-state conditions (Magesh et al., 2022; Shan et al., 2023; Sharma et al., 2023; Shan et al., 2024).

Among their notable features, the luminescence and, in particular, the photoluminescence (PL) emission of CQDs have garnered significant attention in recent years. CQDs are capable of emitting photons across a wide spectrum, from the ultraviolet to the near-infrared region (Zhang et al., 2019). Several plausible mechanisms have been proposed to explain the factors influencing the PL emission of CQDs, including surface passivation, aggregation-induced emission (AIE), proximity effects of other CQD particles, and CQD size (Tang et al., 2019; Lai et al., 2020; Ghasedi et al., 2022). Additionally, impurities can significantly alter the optical behavior of

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CQDs by introducing new energy states within the band gap between the highest occupied molecular orbital (HOMO) and the lowest unoccupied molecular orbital (LUMO), thereby affecting electronic transitions (Yang et al., 2018; Kou et al., 2020). An excited electron can jump to an energy state within the middle of the band gap and then return to its initial state. In the study of Tang et al. (2019), white-emitting CQDs were prepared using a direct one-pot synthesis approach, which offers the advantage of an *in situ* cellulose matrix in the synthesis medium.

On the other hand, the use of green resources to produce nanoparticles and quantum dots has been a focus of extensive research in recent years (Haji and Barzinjy, 2023; Khajavi et al., 2023; Kakhki et al., 2024). The key advantages of these green synthesis methods include simplicity, low cost, and biocompatibility. Recently, our group has synthesized CQDs using the leaves of Aloe Vera (Montazeri et al., 2024). The durability of cellulose fibers in the Aloe Vera paste allowed us to utilize them as *in-situ* matrix networks. In a recently published paper, Hong et al. (2019a) synthesized CQDs through a one-step hydrothermal process using straw plant and bamboo plant residues at 180 °C. Subsequently, they characterized the synthesized sample using spectroscopic methods, demonstrating its high optical properties and quantum performance. Finally, they presented their product for advanced biological imaging applications. In another study, CQDs were synthesized from candle soot and dispersed in a chitosan matrix, yielding a homogeneous CQD-Chit nanocomposite. After extensive experiments and subsequent spectrometric analysis, it was determined that the proposed sensor can be used to measure insulin with high selectivity, accuracy, and sensitivity. Additionally, this sensor offers a simple, cost-effective construction and superior stability compared to other sensors (Abazar and Noorbakhsh, 2020).

In the present study, CQDs were prepared under hydrothermal conditions (in an aqueous environment at 160 °C) using an easily accessible citrus species as a precursor. This method is simple, environmentally friendly, and potentially scalable for mass production. The obtained CQDs contain a significant amount of oxygen-containing functional groups, as confirmed by infrared spectroscopy. A series of analyses, including transmission electron microscopy (TEM), ultraviolet-visible (UV-Vis) spectroscopy, X-ray diffraction (XRD), and Fourier-transform infrared (FT-IR) spectroscopy were conducted to provide sufficient evidence supporting the claims regarding the white emission of CQDs. Additionally, the final product of this study is a CQD-Ag-polymer bio-composite which has potential applications in biological fields, as compounds such as chitosan and silver have strong antibacterial properties, and their combination with active CQDs can enhance these properties.

2. Experimental

2.1. Synthesis of CQDs

We synthesized CQDs using a hydrothermal/solvothermal method using citrus (tangerine) juice as the carbon source. For this propose, 25 mL of the filtered fruit juice and 25 mL of deionized water were poured into a teflon autoclave, which was protected by a stainless steel. After shaking, the autoclave was tightly lidded reaction container and placed in an oven at 160 °C for 4 hours (Bhunia et al., 2013). After the completeness of the reaction, the autoclave was cooled to ambient temperature. Next, the obtained product was placed in a centrifuge for 10 minutes at a speed of 12000 rpm. Then, the obtained solution was filtered with a 0.22 mm smooth syringe head filter and was finally kept as CQDs. The sediment was also dried and stored in a suitable place for analysis.

2.2. Synthesis of CQDs/Ag

To synthesize CQDs/Ag, 10 mL of $AgNO_3$ (1.0 M) was mixed with 10 mL of NaCl (1.0 M). After shaking, the mixture was centrifuged at 8000 rpm for 5 min. Next, 5 mL of NaOH (1.0 M) was added to the residue and shaked vigorously. The centrifuging process was then continued for over 5 min. The residue, as silver oxide, was kept to be used in the next step. In the next step, 0.06 g of silver oxide was added to 5 mL of deionized water and centrifuge was placed in an ultrasonic bath for 30 min at 60 °C. Then, 1 mL of the silver oxide solution and 1 mL of CODs were poured into an autoclave containing 10 mL of deionized water. After tightly lidding, the autoclave was put in an oven at 160 °C for 2 h. The obtained yield was kept cold in a dark place for the synthesis of CQDs/Ag/Chitosan.

2.3. Synthesis of CQDs/Ag/Chitosan composite

For the successful synthesis of CQDs/Ag/Chitosan composite, 0.2 g of chitosan was first dissolved in 50 mL of acetic acid (1.0%). Then, 10 mL of CQDs/Ag was added dropwise to chitosan solution at 40 and the mixture was stirred moderately for 24 h. Finally, the obtained powder (CQDs/Ag/Chitosan composite) was kept in a cold place for the relevant analyses (Esmaeilzadeh et al., 2020).

2.4. Characterization methods

To prove the formation of CQDs, a UV chamber with a lamp in the wavelength range of 200 to 300 nm was used. For this purpose, a solution of CQDs and NaOH with the ratio of 1 to 2 was poured in a test tube and exposed to UV radiation. In this relation, UV-VIS spectra were used to determine the amount and range of absorption of CQDs. About 1 mL of the product diluted 12 times with water to completely become transparent which was then transferred to the special quartz cell of the device and the absorption spectrum was recorded over the wavelength range of 200 to 800 nm. Absorption spectra were also recorded for the synthesized products of other steps in the same way using a NanoDrop 2000 UV-Vis spectrophotometer. A 150 kV Philips CM30 transmittance electron microscopy (TEM) was used to take TEM images. To obtain Fourier Transform Infrared (FT-IR) spectra of the sample, a PerkinElmer

spectrometer (ASTM E 1252-98) was employed.

3. Results and Discussion

3.1. Impact of addition of different volumes of CQD/Ag composite to chitosan

The experiment was carried out by adding 1, 4, 7 and 10 mL of CQD/Ag composite to chitosan, and it was found that the higher the volume of CQD/Ag composite, the darker the color of the obtained product that is due to the presence of more CQDs representing optical properties of the sample. As shown in Fig. 1, the product with 4 mL CQD/Ag has lower optical absorption than the product with 10 mL. The greenish photoluminescence of CQDs has been shown in Fig. 1.b and Fig. 1.d.

3.2. UV-Vis. absorption spectra of the prepared CQDs, CQDs/Ag composite and Ag NPs

UV-visible spectroscopy is regarded as a simple approach for characterizing carbon quantum dots (CQDs), implying some valuable insights into their optical properties and structural features. The UV-visible spectra of CQDs typically exhibit absorption peaks in the UV region, often accompanied by emission peaks in the
visible range, which can be used to determine the size, shape, and composition of the particles (De and Karak, 2013; Zulfajri et al., 2019). The position and intensity of these peaks can provide information about the functional groups present on the surface of the CQDs, such as carbon, nitrogen, and oxygen, which are critical for their optical and biological applications (De and Karak, 2013; Zulfajri et al., 2019). Furthermore, the UV-visible spectra can be used as proper criteria to evaluate the photoluminescence mechanism of CQDs, which is of paramount importance for understanding their fluorescence properties and potential uses in sensing, imaging, and photocatalysis (Guo et al., 2017; Zhu et al., 2017; Rawat et al., 2023; Danawala et al., 2024). In the literature, UV-visible spectroscopy is referred as a powerful tool for probing the optical and structural characteristics of CQDs, enabling researchers to optimize their synthesis and applications in various fields.
The absorption spectrum of CQDs has shown two peaks between 200 nm and 300 nm, which are caused by π to π* electron transitions related to sp^2 orbitals and n to π* electron transitions related to C=O bonds. Fig. 2 (A-E) depicted UV-Visible spectra of all the products in this study.

3.3. IR spectra of the synthesized CQD and CQD/Ag/Chit

IR spectroscopy was used to determine the functional groups of the prepared products. For this purpose, the synthesized product (CQD and CQD/Ag/Chit) was placed in a dark place at room temperature for 48 hours to ensure the absence of any water in the environment. Then, 0.1 g of the powdered sediment with 1.0 g of potassium bromide was pounded and homogenized into a tablet and the spectra were recorded.
In order to identify functional groups on the surface of CQDs, the absorption of synthesized nanoparticles in the IR spectral region was taken into consideration. CQDs are composed of a core of two-dimensional spherical structures whose surface are covered by hydrophilic groups in the raw materials and cause the optimal solubility of CQDs in the aqueous environment (Lv et al., 2017). After the synthesis and filtration, the CQDs sample of the sediment obtained was dried and the FT-IR spectrum was recorded (see Fig. 3). Using FT-IR, the spectral analysis shows a strong and relatively broad peak in the range of 3400 cm^{-1} related to the stretching vibrations of the O-H groups. The stretching vibrations of the C-H group (CH_2/CH_3) also appeared around the wavenumber of 2930 cm^{-1}. The peaks appearing or the range of 1600-1700 cm^{-1} belong to C=C and C=N bonds, and the peaks related to N=O and C-O of ester groups were observed at 1400 and 1200 cm^{-1}, respectively. The peak at 1730 cm^{-1} is related to the C=O groups of the carboxylic acid in the relevant structures.
In the infrared spectrum of chitosan, the peak appearing at 3440 cm^{-1} is related to the O-H group and intramolecular hydrogen bonds, while that at 2900 cm^{-1} shows the prisons of C-H group of the carboxylic acid group in chitosan. On the other hand, the peak at cm^{-1} 1600 is related to N-H bending vibrations of the amine group in this polymer. Also, peaks in the range of 1040-11000 cm^{-1} are attributed to the C-O amine and ring vibrations (Dimzon and Knepper, 2015; Negrea et al., 2015).
In the infrared spectrum containing CQDs, silver and chitosan, the peak at the wavelength of 11650 cm indicates the C=O stretching vibrations of the amide bond and the possibility of forming a bond between the carboxyl group of CQD and the amine group of chitosan. The two peaks appearing in the range of 3400-3500 cm^{-1} have shown the stretching vibrations of N-H and O-H group compared to the spectrum of chitosan alone (Dimzon and Knepper, 2015; Negrea et al., 2015) (Fig. 3).

3.4. XRD pattern of that prepared CQDs

X-ray diffraction (XRD) spectroscopy has been proven as a powerful technique for characterizing the structural properties of carbon quantum dots (CQDs). XRD analysis can provide information about the crystalline structure, phase composition, and average crystallite size of CQDs (Shaikh et al., 2019; Hamid Abd and Ibrahim, 2022; Sharma et al., 2022). The XRD pattern of CQDs typically exhibits a broad peak around 20-30° 2θ, indicating the presence of amorphous or nanocrystalline carbon (Shaikh et al., 2019; Sharma et al., 2022). The position and width of this peak can be used to estimate the average size of the CQD core, with smaller CQDs exhibiting broader peaks due to quantum confinement effects (Hamid Abd and Ibrahim, 2022; Das et al., 2024). Additionally, the presence of sharp peaks in the XRD pattern may suggest the formation of graphitic or crystalline domains within the CQDs (Das et al., 2024). By comparing the experimental XRD data with

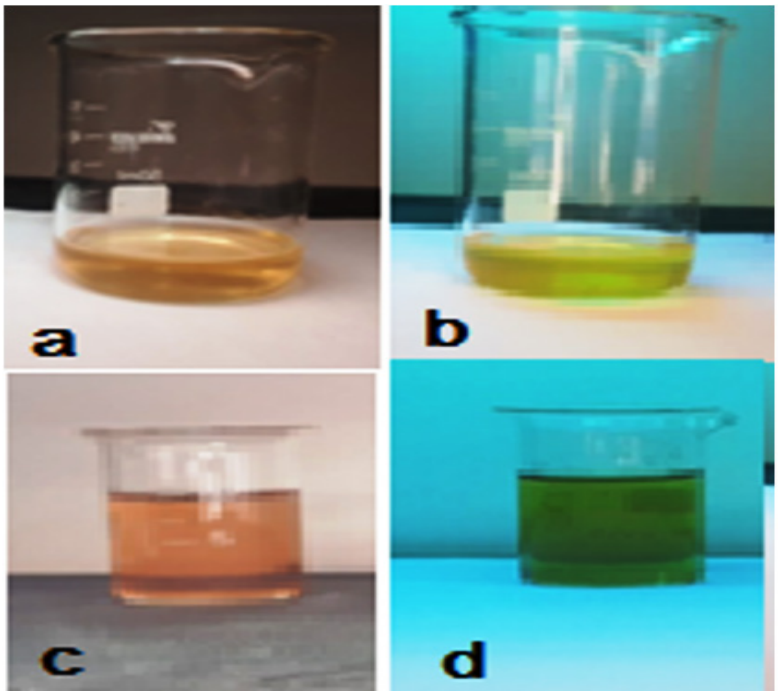

Fig. 1. a: CQD/Ag/Chit with 4 mL of CQD/Ag . **b:** Sample a under UV lamination. **c:** CQD/Ag/Chit with 10 mL of CQD/Ag . **d:** Sample c under UV lamination.

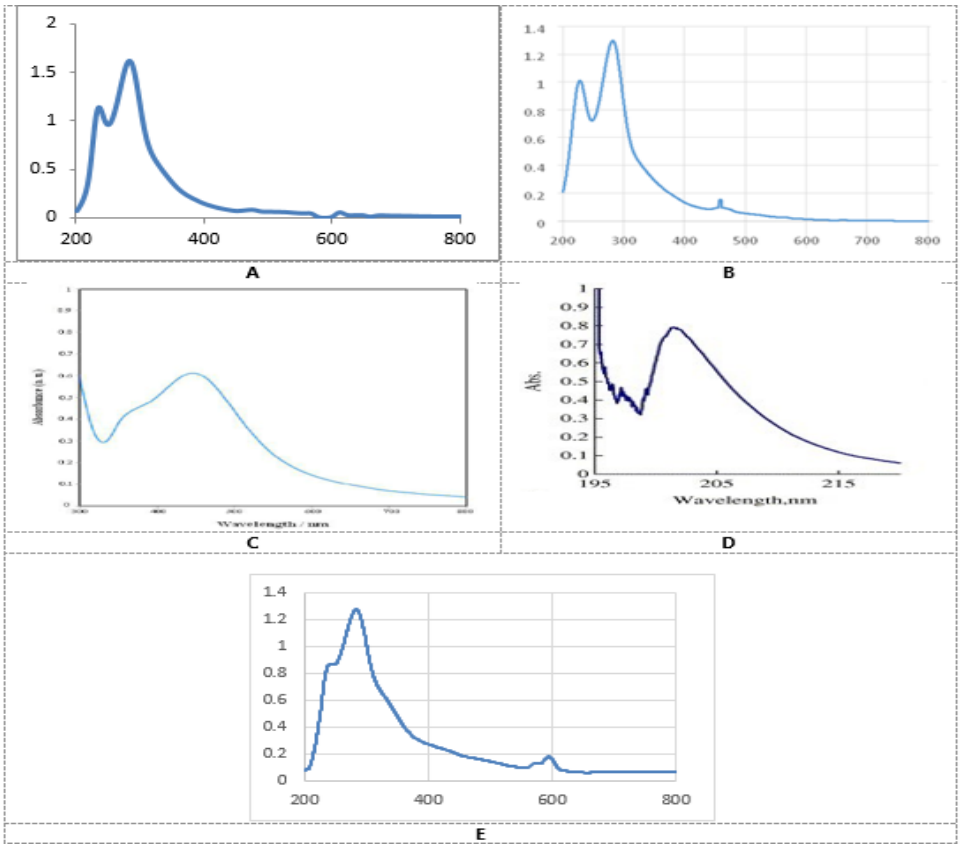

Fig. 2. UV-Visible spectra of **A:** CQDs, **B:** CQDs/Ag composite, **C:** AgNPs, **D:** Chitosan and **E:** CQDs/Ag/Chit.

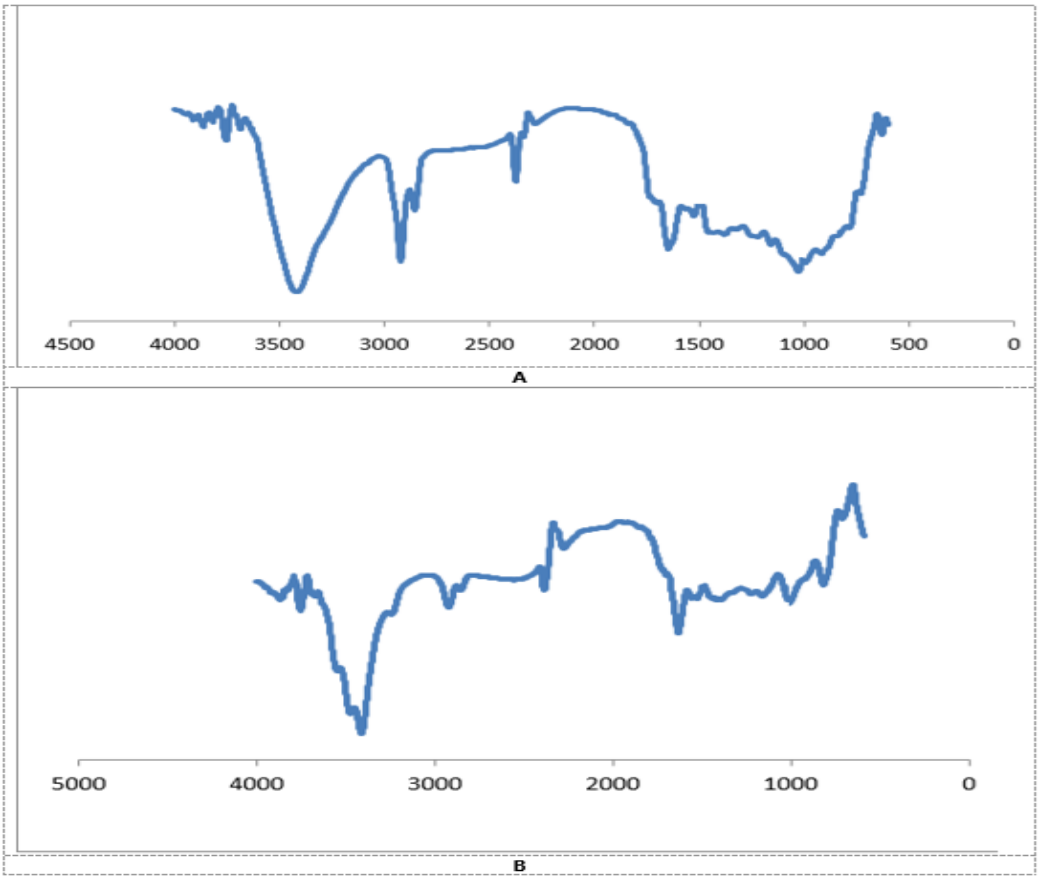

Fig. 3. IR absorption spectra of **A**: CQDs; **B**: CQDs/Ag/Chit.

reference patterns, it is possible to identify the presence of specific carbon allotropes or impurities in the CQD sample (Shaikh et al., 2019; Hamid Abd and Ibrahim, 2022; Sharma et al., 2022). In fact, XRD spectroscopy is a valuable tool for probing the structural characteristics of CQDs and understanding their formation mechanisms and properties.

As shown in Fig. 4, according to the XRD pattern of activated carbon, which has a strong peak in the range of Θ_2 = 20˚ and the characteristic of the presence of activated carbon, it can be attributed to CQDs. This type of carbon has an irregular and mostly amorphous structure.

3.5. TEM images of the prepared CQDs

Transmission electron microscopy (TEM) is often considered as an efficient technique for characterizing nanoparticles, offering detailed structural and chemical insights at the nanoscale (Anandharamakrishnan and Anandharamakrishnan, 2014; Mast et al., 2020). TEM enables the analysis of 2D projections of individual particles, providing information on their size, shape, and composition (Jain et al., 2023). This technique is ideal for nanoscale studies, offering high-resolution imaging, electron diffraction, and chemical analysis in a single instrument. With advancements like highly coherent field emission electron sources, TEM enhances its analytical capabilities, allowing for the study of diverse nanomaterials such as carbon nanocages, metal nanoparticles, and soft materials used in drug delivery systems. The versatility of TEM in providing comprehensive information about nanoparticles makes it an indispensable tool in nanomaterial characterization (Kumar, 2013; Anandharamakrishnan and Anandharamakrishnan, 2014; Mast et al., 2020; Vladár and Hodoroaba, 2020; Jain et al., 2023).

To check the morphology and determine the average size of the particles, transmission electron microscopy (TEM) is used. As can be seen in the Fig. 5, the synthesized CQDs have shown a spherical crystal structure that is well dispersed in water and the particles do not have significant molecular aggregation. Also, the average diameter of each spherical particle is 25 nm, which is comparable to other works. In this context, the particles have the right size.

4. Concluding remarks

CQDs were prepared under hydrothermal conditions (in an aqueous environment at a temperature of 160 °C) using an easily available citrus species as a precursor. This method is an easy and environmentally friendly method that may be possible for large-scale production. The obtained CQDs contain a large amount of oxygen-containing groups, which were detected by infrared spectroscopy. According to the analyzed results, hydrothermal forms points close to polyaromatic

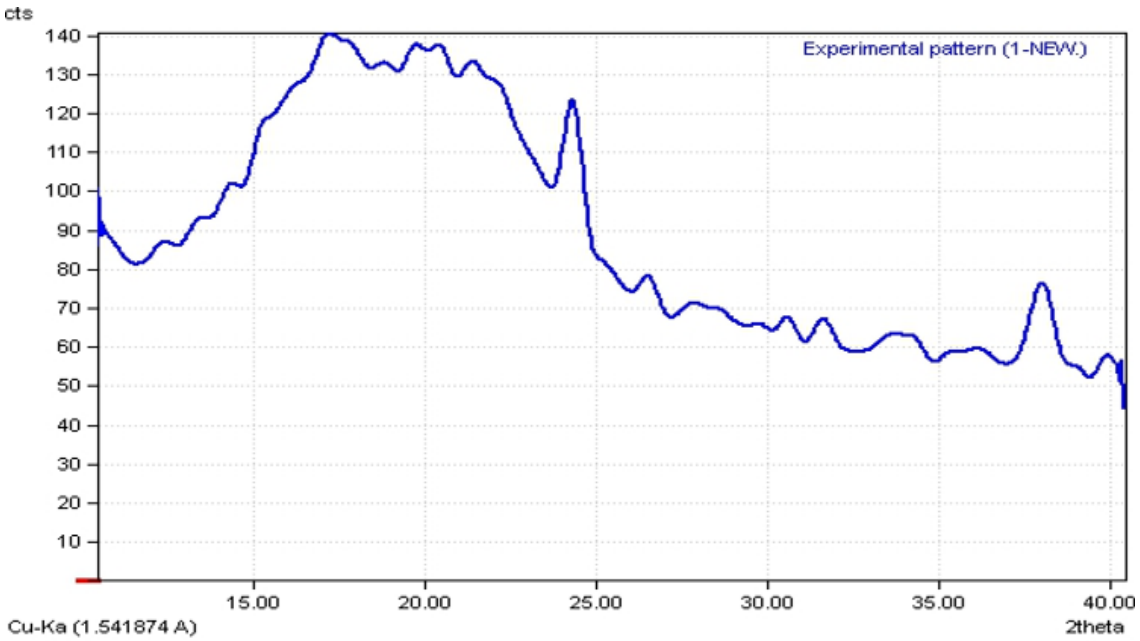

Fig. 4. XRD diagram of CQDs.

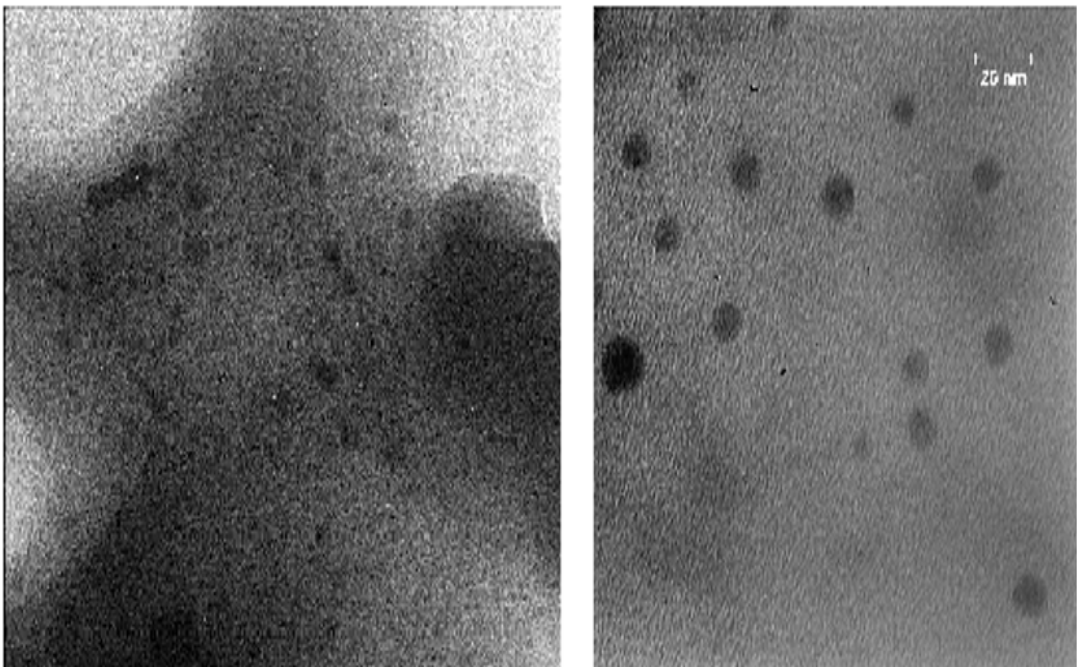

Fig. 5. TEM images of CQDs.

hydrocarbon clusters, such as furanocarbon-based polymers, and sp^2 and sp^3 points are formed with attractive luminescence properties. The prepared CQDs are worthy of attention due to their advantages in green synthesis, high solubility in aqueous environment and luminescence properties, and they can have potential applications as fluorescent markers and efficient catalysts in life and energy sciences. Also, the final product of this study is CQD-Ag-polymer biocomposite. Chitosan has also kept its optical and photoluminescence properties. This composite can be suitable in order to biological aspect, because according to literature, compounds such as chitosan and silver have strong antibacterial properties, and their combination with active CQDs can

intensify their antibacterial properties.

Abbreviations

AIE: Aggregation-Induced Emission; **CQD/Ag/Chit:** Carbon Quantum Dots/Ag/Chitosan; **CQDs:** Carbon Quantum Dots; **FT-IR:** Fourier-Transform Infrared; **HOMO:** Highest Occupied Molecular Orbital; **LUMO:** Lowest Unoccupied Molecular Orbital; **TEM:** Transmission Electron Microscopy; **UV-Vis.:** Ultraviolet-Visible; **XRD:** X-Ray Diffraction.

Data availability

The data can be provided from the authors upon reasonable request.

Author contribution statement

Conceptualization and literature search were performed by Ehsan Koushki and Behnam Mahdavi. The first and final drafts of the manuscript were prepared by Behnaz Noori Dolouie and Majid Mohammadhosseini. Ehsan Koushki and Behnam Mahdavi critically analyzed and gave suggestions to finalize the manuscript. All authors read and approved the final manuscript.

Conflict of interest

The authors declare that there is no conflict of interest.

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