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Performance Advantages and Applications of Carbon Dots in Biological Detection

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Abstract. Carbon dots (CDs), as zero-dimensional carbon-based nanomaterials, have emerged as a research focus in biomedical fields due to their superior properties compared to traditional fluorescent materials. This paper first outlines the background of CDs, including their discovery timeline and core characteristics such as tunable photoluminescence, nanoscale size, non-toxicity, and good biocompatibility. It then systematically analyzes three key performance advantages of CDs in biological applications: excellent fluorescent properties, such as tunable colors, high stability, red/near-infrared emission, nanoscale size-enabled benefits, such as good water solubility, easy surface functionalization, deep tissue penetration, and non-toxicity Subsequently, the paper explores specific application scenarios in biological detection, including ion detection, in vitro cell imaging, and in vivo biological imaging. Finally, the paper discusses current challenges and future outlooks, emphasizing that CDs hold great potential to advance clinical diagnosis and life science research with further technological development. In the future, the development of carbon dots will further advance the progress of the biomedical field.

Keywords: Carbon dots; biological detection; fluorescent probes; bioimaging; nanomaterials.

1. Introduction

Carbon is the fundamental element of all organic life and plays a crucial role in the development of nanomaterials. Ranging from the three-dimensional structure of graphite to two-dimensional graphene and graphene oxide, and further to one-dimensional carbon nanotubes, carbon-based nanomaterials have attracted researchers with their outstanding properties and broad application potential. Recently, there has been a surge in interest in carbon-based nanomaterials such as carbon nanotubes (CNTs), fullerenes, graphene, and nanodiamonds [1].

Carbon dots (CDs), discovered in the early 2000s, are new members of the "carbon family" and have quickly gained attention due to their unique properties. People first discovered CDs in 2004. During the process of Xu and his team using electrophoretically separating, and purifying single-walled carbon nanotubes (SWCNTs), they discovered fluorescent nanoparticles. However, in 2006, Sun and his colleagues officially introduced the term "carbon quantum dots" which ignited widespread interest in this field. More importantly, it marked the beginning of an era of in-depth research as well as innovation on CDs [2].

CDs are one of the Nanomaterials which are zero-dimensional carbon-based. Because it is a non-toxic alternative to traditional semiconductor quantum dots, so they are often associated with toxicity and environmental issues. Recently, CDs have attracted widespread attention across various scientific fields. The growing interest can be caused by a number of factors, such as their exceptional optical properties, versatile surface chemistry, biocompatibility, cost-effective preparation and environmentally friendly synthesis methods. Typically, the measuring of CDs is less than 10 nm. And they possess a number of remarkable characteristics like the ability to adjust the photoluminescence, the emission being dependent on the excitation and a high level of stability. This has led to these nanoparticles being a focal point of research in a wide range of disciplines, from chemical sciences to engineering to biomedical sciences.

The topic investigated by this paper is the applications of CDs in biological detection. For the body of this paper, it is divided into three main parts. The initial section is dedicated to the performance advantages of CDs in biological applications. This includes a thorough examination of their strengths in three distinct aspects: fluorescence, nanoscale size, and non-toxicity. The subsequent section explores the application of CDs in the biological field, covering the analysis of three specific scenarios which are how to use CDs to detect ions, image in vitro cell, and image in vivo biology. Ultimately, this work aims to provide theoretical and practical references for the development of CDs for the use in biomedicine.

2. Performance Advantages of CDs in Biological Applications

2.1. Fluorescence

When it is compared with other traditional fluorescent materials, CDs have many obvious advantages. For example, different with semiconductor quantum dots (QD), it can avoid heavy metal toxicity and photobleaching. They are also differing from rare earth nanomaterials, since it can control fluorescence precisely without complexing doping. In addition, CDs have higher solubility in polar solvents than conjugated polymers. CDs possess numerous unique characteristics. The first one is the tunable fluorescent colors: by regulating the species of carbon sources as well as the conditions of reaction, it can achieve the purpose of full-spectrum fluorescence regulation from blue to red to satisfy the needs of fluorescence in various scenes. The second one is the excellent optical and chemical stability: as for the optical aspect, when 365 nm ultraviolet light (100 mW/cm^2) was continuously irradiated for 12 hours, the fluorescence quantum yield (QY) of CDs only decreased by 3.2%, which was much better than that of QDs which decrease by 28.5% and conjugated polymers which decrease by 41.7%. As for chemical aspect, CDs can withstand high temperatures of 80°C and pH ranges of 1-13. They dissolve in polar solvents such as ethanol and water with a solubility of >10 mg/mL, and there is no aggregation quenching problem like conjugated polymers [1].

In recent years, it is suprising that fluorescent carbon quantum dots have became one of the most popular fluorescent carbon nanomaterials. They are named in this way, since they possess optical properties, which are similar to those of traditional quantum dots. For example, small particle size, a wide excitation wavelength range, tunable emission wavelength, high fluorescence efficiency, good photostability, and so on. However, carbon dots can be regarded as a novel, 'friendly' type of quantum dot due to their excellent biocompatibility and low toxicity, compared with traditional quantum dots. When carbon dots first came into people's view in 2004, they became a research hotspot for scholars not only at home but also at abroad. Their continuously explored superior optical properties have enabled them to make significant progress in research areas such as living cell labelling, biological imaging and the medical field. The following brief introduction covers the structure, preparation methods, optical properties and application prospects of CDs. [3].

2.2. Nanoscale Size

Typical CDs are quasi-spherical zero-dimensional nanomaterials with a diameter of approximately 10 nm, featuring abundant functional groups on their surface, such as carboxyl, hydroxyl, and amino groups. This nanoscale dimension confers multiple advantages. The first advantage is excellent water solubility. Because the nanoscale size ensures CDs can be uniformly dispersed in biological systems, facilitating their application in aqueous environments. The second advantage is surface functionalization. Since the rich functional groups enable facile conjugation or modification with biomolecules, making them ideal for constructing targeted bioprobes. The third advantage is deep-tissue penetration. Because their small size enhances penetration and diffusion through biological tissues, rendering CDs suitable for deep-tissue bioimaging and detection.

2.3. Non-Toxicity

Bioimaging is a technique that enables real-time, non-invasive direct observation of physiological conditions inside living organisms using cameras and detectors. Traditional fluorescent materials are limited in biomedical applications due to issues like toxicity or poor fluorescent performance. In contrast, CDs possess advantages including photostability, excellent biocompatibility, and simple synthesis routes, making them next-generation fluorescent probes for in vitro and in vivo bioimaging—thus highlighting the advantage of CDs' non-toxicity in the field of bioimaging .

In experiments on the characterization of red and near-infrared fluorescence-switchable carbonized polymer dots and their bioimaging applications, researchers applied CD solutions under acidic conditions to fluorescent imaging of the mouse stomach, achieving favorable imaging results. This experimental outcome indirectly reflects the non-toxic advantage of CDs: if CDs were toxic, they would impair mouse health, interfere with the imaging experiment, or even render the experiment unfeasible. This proves that the non-toxicity of CDs is a prerequisite for their successful application in bioimaging.

In studies on the application of hydrophobic near-infrared fluorescent CDs in bioimaging, researchers used CD super particles for stomach imaging and whole-body imaging of mice, enabling clear identification of the mouse stomach and hindlimb blood vessels. Because throughout the entire experiment, everything proceeded smoothly, and there is nothing which can prove CDs are toxic were observed. This phenomenon emphasizes the importance of CDs' non-toxic properties in bioimaging applications, as these properties ensure the feasibility of experiments and the accuracy of results.

3. Analysis of Application Scenarios in the Biological Field

3.1. Ion Detection

Carbonized polymer dots (CPDs) are widely used to detect cations and anions in the environment. The principle of the detection is closely related to the charactristis of the surface of CDs. The surface groups of CDs can form complexes with ions or bind to them through electrostatic interactions. Researchers have further designed a reversible "on-off" nanosensor. It utilizes CDs which is treated with Zn^{2+} for the selective detection of ethylenediaminetetraacetic acid (EDTA) and Zn^{2+}, as shown in Fig. 1 [1].

Relying on the specific recognition ability of CDs for chromium ions and cysteine, as well as the advantageous signal of red fluorescence emission. The investigators constructed an analytical method based on the dual-mode sensing of these kind of CDs. This method delivers superior performance and impact in real-time monitoring applications and has potential applications in clinical diagnosis and environmental analysis. It not only has the speacial ability of fast responding, but can also improve the accuracy of detection by using the dual-mode signal cross-validation. These characteristics of CDs offers significant advantages and unprecedented convenience in the ares of both clinical diagnosis and environmental analysis. It holds great promise for widespread use in these areas.

Moreover, thanks to the response to pH changes by dual-modal and the well-established analytical method, the pH value of wounds can be detected intuitively and conveniently in theory and in practice. In addition to applications in ion and pH detection, CDs also demonstrate selectivity for various biomolecules, including amino acids, glutathione and formaldehyde. Therefore, fluorescent probes prepared from CDs can provide valuable information for the early diagnosis and prevention of disease.

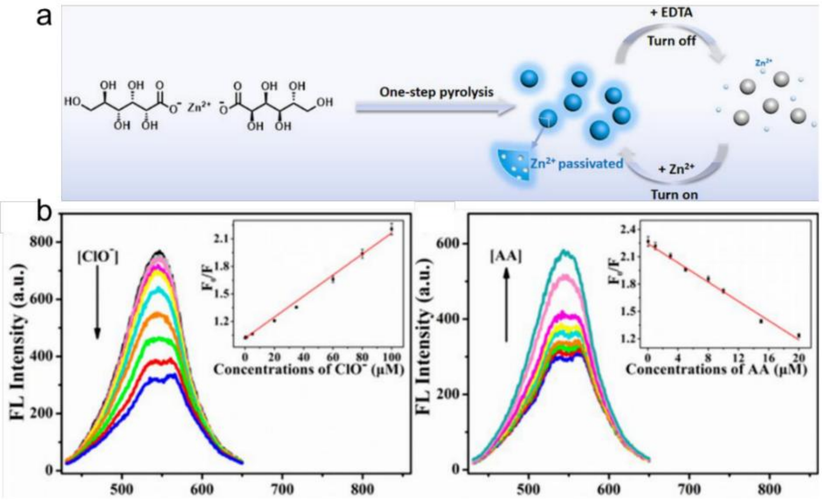

Fig. 1 (a) Illustration of the Possible Formation and Sensing Mechanism of CDs (b) The fluorescence intensity of carbon spots changed with the increase of ClO [1]

3.2. In Vitro Cell Imaging

In basic cell biology research, CDs can serve as fluorescent probes to achieve long-term dynamic imaging of living cells. Their excellent photostability effectively avoids the issue of photobleaching that plagues traditional organic dyes, enabling continuous tracking of morphological changes and behavioral characteristics of cells during processes such as proliferation and migration.

At the organelle level, precise targeted imaging of key organellescan be achieved by regulating the surface functional modification of CDs—such as conjugating targeting peptides, specific antibodies, or small-molecule ligands. This facilitates researchers to intuitively observe the structural dynamics, functional states of organelles, and their changes under pathological conditions. For instance, it allows monitoring of changes in mitochondrial membrane potential or the degradation function of lysosomes under stress [4].

Furthermore, CDs also demonstrate significant application value in cell-level disease-related research. For example, leveraging the fluorescent response properties of CDs, intelligent probes sensitive to specific reactive oxygen species (ROS), pH values, or biomolecules in cells can be designed. These probes enable real-time monitoring of abnormal changes in the cellular microenvironment, providing visual evidence for the early cellular diagnosis of diseases such as cancer.

3.3. In Vivo Biological Imaging

In mouse stomach imaging experiments, CDs exhibit excellent near-infrared (NIR) fluorescence performance and high quantum efficiency after acid stimulation, laying a foundation for bioimaging applications. When acidified CDs are used as contrast agents for real-time imaging of the mouse stomach, bright NIR fluorescence in the stomach area and high-quality imaging results are clearly observed. Meanwhile, the radiation power under the applied conditions is far below the safe exposure limit, fully demonstrating the excellent NIR imaging capability and deep-tissue penetration ability of CDs in living organisms.

In experiments related to CD superparticles, the superparticles formed by encapsulating CDs with amphiphilic polymers show outstanding performance. For stomach imaging, after perfusing the CD superparticle solution into the mouse stomach, fluorescence in the stomach area can be observed within a short time; even after a certain period, the fluorescence remains strong and the imaging remains clear—indicating the superparticles have good stability, high fluorescence intensity, and strong penetration. For whole-body imaging, after intravenous injection of CD superparticles, blood vessels and muscle tissues can be clearly identified. By observing the metabolic status at different time points, the distribution changes of CD superparticles in the body can be tracked, and it is inferred that they may be metabolized through the liver and intestines.

The excellent performance of CDs and CD superparticles overcomes the limitations of traditional contrast agents in NIR imaging, such as weak fluorescence, shallow penetration, and low safety. They provide a new tool for the accurate imaging diagnosis of gastric diseases, the visual observation of in vivo tissues and organs, and the tracking of drug metabolism. Their good biocompatibility and metabolic properties also provide key support for subsequent clinical translation, holding great promise for promoting the further development of NIR bioimaging technology in the fields of medical diagnosis and life science research [1].

4. Outlook

Compared with quantum dots and other carbon materials, the research on CDs is still in its relatively early stages. Currently, one of the most fundamental and critical issues is the lack of systematic and scalable synthesis protocols for producing high-quality CDs with desired structures—such as controlled size, shape, crystallinity, number of functional groups, and type and location of defects. Additionally, due to unstandardized synthesis routes and impurities, their exact reaction mechanisms, nucleation mechanisms, and formation processes remain unclear. Therefore, for the large-scale production of high-performance CDs via efficient approaches, systematic exploration should be conducted on the effects of precursors and reaction conditions on the properties of CDs, and purification protocols based on size or polarity need to be developed [5].

As a new type of carbon-based nanomaterial, CDs exhibit important and significant potential in a wide range of application fields, thanks to their unique fluorescent properties, excellent biocompatibility, low cost, non-toxic safety, and functionalizability. However, from the perspective of the practical application of CD-based materials, much more research work remains to be done. Several deepen opinions on the preparation, mechanisms, structure, properties, and applications of CDs have been proposed to provide potential guidance. Thanks to the development of advanced technologies and characterization methods, it is believed that controllable synthesis methods and large-scale production can be achieved, as can a better understanding of the structure-property relationship. This will greatly expand the range of applications for CD-based materials. The unique functions of CDs will pave the way for a bright future.

5. Conclusion

As a type of zero-dimensional carbon-based nanomaterial, CDs contain a significant application value in the field of biological detection by using their advantages such as tunable fluorescence, water solubility and modifiability derived from nanoscale size, good biocompatibility, and low toxicity.

In specific application scenarios, the ability of detection of CDs relies on specific mechanism of action. For instance, their surface groups can specifically bind to target ions (such as Hg^{2+}, Cu^{2+}) or biological molecules through coordination and electrostatic interactions, and the fluorescence signal changes can be used for precise detection. When they are made into fluorescent probes, they can use their stability of light to support for long-term dynamic cell imaging and even the targeted observation of organelles like mitochondria or lysosome. This kind of long-term and stable ability can provide a basis for early disease diagnosis. When it comes in vivo imaging, acid-stimulated CDs can take advantage of their inherent pH 1-13 tolerance property. And polymer-encapsulated CD superparticles allow clear imaging of the mouse stomach, whole-body vascular visualization, and metabolic tracking. With safe radiation and strong penetration of CDs, they can sucssefully overcome the limitations of traditional quantum dots.

Although there are currently challenges in the synthesis of CDs like the specific pathway by which the carbon source is converted into carbon dots in the reaction mechanism is still unclear. While during the purification process, the aggregation of nanoparticles is prone to cause low separation efficiency. However, existing research progress has fully demonstrated its broad prospects in the field of biomedicine: with the optimization of synthesis processes and breakthroughs in functional modification technologies, CDs are expected to further promote the development of the integrated technology of "real-time detection - precise imaging" in clinical diagnosis. At the same time, they will provide safer and more efficient tools for dynamic tracking of cells as well as the analysis of molecular interactions in life science research. It can accelerate the transformation of basic research into clinical applications.

References

[1] Mei Xuetian. Preparation of Red and NIR Emissive Carbonized Polymer Dots for Bioimaging. Jilin University, 2021.

[2] Sen S, Bose A. Carbon dots: A review of innovations, applications, challenges, and future prospects. Inorganic Chemistry Communications, 2025, 173: 113852.

[3] Wang D. Preparation and Properties of Fluorescent Carbon Dots. Lanzhou University, 2015.

[4] Mao Q, Meng Y, Feng Y, Li H, Ma T. Organelle imaging with carbon dots: strategies, challenges, and perspectives. Inorganic Chemistry Frontiers, 2023, 11(3): 713–734.

[5] Liu J, Li R, Yang B. Carbon Dots: A New Type of Carbon-Based Nanomaterial with Wide Applications. ACS Central Science, 2020, 6(12): 2179–2195.