#### **REVIEW ARTICLE**



## Food safety aspects of carbon dots: a review

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#### Abstract

Discovered in 2004, carbon dots have garnered a major attention due to their unique optical properties, nanoscale size, and cost-effectiveness. Their potential uses are applicable for bioimaging, electronics, and the food industry. Carbon dots are promising tools for detecting contaminants, identifying harmful bacteria, and monitoring essential nutrients. Here, we review the safety risks associated with applying carbon dots in the food industry, focusing on their integration into global food safety frameworks. We highlight recent advancements in the detection capabilities of carbon dots, showcasing their sensitivity and specificity in identifying foodborne pathogens and contaminants. We discuss strategies to mitigate potential health risks, such as optimizing carbon dot synthesis to minimize their toxicity and ensuring thorough regulatory assessments. Current research shows that carbon dots improve food safety, but research is needed to address safety concerns and ensure consumer confidence.

**Keywords** Carbon quantum dot  $\cdot$  Cytotoxicity  $\cdot$  Human health  $\cdot$  Food industry  $\cdot$  Nanosensor  $\cdot$  Safety risk  $\cdot$  Bioaccumulation  $\cdot$  Carbon nanodot

## Introduction

Carbon dots, also known as carbon nanodots, are minuscule luminescent particles with diverse potential applications in the food industry (Shi et al. 2019). These particles consist of carbon atoms organized in a nanoscale arrangement, typically measuring less than 10 nm in diameter, smaller than most viruses and bacteria (Xu et al. 2004). Their unique characteristics include intensive fluorescence, large surface area, biocompatibility, low cost, one-step production, and stability (Kang et al. 2020). In recognition of their

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Prize in Chemistry was awarded to three scientists (Nobel-Prize 2023). Because of these exceptional attributes, carbon dots have the potential to revolutionize the food industry, especially in the context of ensuring food safety by providing new and effective ways to detect contaminants, foodborne pathogens, measure nutrients, and extend the shelf life of food products (Ezati et al. 2022; Gao et al. 2024; Rossini et al. 2019; Zhang et al. 2022; Zhao et al. 2021a). For example, carbon dots-based sensors are being developed to detect foodborne pathogens such as Staphylococcus aureus or Escherichia coli (Gao et al. 2024; Zhao et al. 2021a), and carbon dots-coated packaging materials are being used to extend the shelf life of avocados until 14 days (Ezati et al. 2022). Carbon dots also can form inclusion complexes with a wide range of molecules. This property has been exploited to develop probes and sensors for detecting contaminants in food samples, such as pesticides, herbicides, or heavy metals (Bera and Mohapatra 2020; Chen et al. 2020; Hoang et al. 2023; Pajewska-Szmyt et al. 2020). However, integrating carbon dots into food products necessitates a comprehensive understanding of its safety aspects.

groundbreaking research on carbon dots, the 2023 Nobel

Food safety is a major global concern, with approximately 600 million people falling ill each year due to contaminated food (WHO 2022). To address this, key interventions include selecting appropriate foods, following good hygiene practices, ensuring food ingredient safety, and preventing cross-contamination during food processing (Njoagwuani et al. 2023). In the case of carbon dots, research on their toxicity is still in its early stages, and findings are often conflicting. While some studies have suggested that carbon dots are biocompatible and non-toxic (Emam et al. 2017; Yang et al. 2009b), others have found that their cytotoxicity can be influenced by factors such as size, dosage, and exposure to light (Liu et al. 2021; Wang et al. 2011). Research on the cytotoxicity of carbon dots has revealed that their photodegradation under light exposure can lead to the production of toxic molecules, posing a potential risk to human cells (Liu et al. 2021). The surface functionalization of carbon dots also plays a crucial role in their cytotoxicity, with carboxylic groups and polyethylene glycol-modified carbon dots with neutral charge showing the most promise for biological applications (Havrdova et al. 2016). Regarding biodistribution and clearance, carbon dots are efficiently excreted from the body, with different injection routes resulting in varying blood clearance patterns and tumor uptakes (Huang et al. 2013). These findings highlight the need for further research to fully understand the potential risks and benefits of using carbon dots in food applications.

While previous review articles have highlighted the potential of carbon dots in various food applications (Li et al. 2021; Sharma et al. 2021; Shi et al. 2019; Zhang et al. 2022), an essential consideration often overlooked is the safety risks associated with their usage. Several strategies were proposed to mitigate the risks associated with the use of carbon dots in prior review articles. For instance, one

approach involves synthesizing carbon dots from biomaterials (Wu et al. 2023). This review article aims to provide a comprehensive understanding of the food safety aspect of carbon dots, including their potential applications in the food industry, along with addressing the associated safety risks and proposing mitigating strategies, illustrated in Fig. 1.

## Carbon dots

## **Carbon dots and properties**

Carbon dots are a new class of nanomaterials with unique properties, including tunable fluorescence, biocompatibility, and surface state energy-gap tuning, making them promising candidates for various applications. These nanomaterials exhibit bright photoluminescence, resembling the properties commonly found in semiconductor quantum dots (Cao et al. 2013). The unique fluorescence characteristics of carbon dots can be tailored for applications in bioimaging, intracellular imaging, and optical sensing, with the ability to create "artificial" tunable carbon dots through composition and surface state modifications (Bao et al. 2015; Fu et al. 2015). Furthermore, carbon dots have been found to possess physicochemical and photochemical stability, making them suitable for applications in bioimaging and fluorescence imaging in vivo (Yang et al. 2009a). The potential use of carbon dots for in vitro and in vivo applications has been discussed, highlighting their non-toxic and high-performance fluorescence imaging capabilities (Yang et al.

Fig. 1 Potential applications of carbon dots in the food industry include monitoring nutrients, detecting food contaminants, detecting food pathogens, and enhancing food packaging. The safety of carbon dots is assessed in terms of cytotoxicity, genotoxicity, bioaccumulation, and biodistribution. Strategies to mitigate these risks include selecting appropriate precursors, optimizing synthesis methods, modifying surface agents, and developing specific protocols to detect carbon dots in foods



2009a). Additionally, the biocompatibility and antioxidant capabilities of carbon dots have been investigated, further emphasizing their characteristic properties, including luminescence and ease of synthesis (Rodríguez-Varillas et al. 2022). Overall, the multifaceted properties of carbon dots, encompassing fluorescence, luminescence, biocompatibility, and tunability, render them versatile nanomaterials applicable in bioimaging, sensing, drug delivery, pharmacetical analysis and optical devices (Ali et al. 2022). Figure 2 illustrates the procedure for synthesizing carbon dots, listing the most commonly applied method in recent studies.

## Synthesis of carbon dots

Carbon dots can be synthesized through two primary approaches: top-down and bottom-up. The top-down approach involves breaking down larger carbon materials into smaller carbon dots, aiming for a narrow size distribution and controlled optical properties (Wang et al. 2023). However, achieving precise control over size and structure can be challenging with this method. In contrast, the bottomup approach assembles carbon dots from smaller molecular precursors, offering greater versatility and control over size, structure, and optical properties, making it more suitable for specific applications (Shin et al. 2015; Wang et al. 2019, 2015). Top-down approaches transform macroscopic carbon structures into carbon dots using methods such as arc

Fig. 2 Common procedures for carbon dot synthesis. Precursors can be chosen from natural or synthetic materials (i) Various techniques are employed to synthesize carbon dots based on two primary approaches: top-down and bottom-up (ii). The synthesized carbon dots are purified using techniques such as, dialysis, ultrafiltration, and column chromatography (iii). The purified carbon dots exhibit intense fluorescence and nanoscale size (iv). Carbon dots are characterized using methods, including transmission electron microscope (TEM), Raman spectrophotometer, particle size distribution analyzer, and fluorescence spectrophotometer (v)



discharge, laser ablation, electrochemical oxidation, chemical oxidation, and ultrasonic synthesis (Wang et al. 2017). On the other hand, bottom-up approaches create carbon dots from molecular precursors like citric acid, sucrose, and glucose through methods such as microwave synthesis, thermal decomposition, hydrothermal treatment, template-based routes, and plasma treatment (Wang et al. 2017).

While various methods have been employed for carbon dots synthesis, recent studies have emphasized distinctive properties, with hydrothermal and microwave methods standing out in top-down approaches, and chemical oxidation and electrochemical oxidation in bottom-up methods (Fig. 2). Each method presents a unique set of benefits and challenges (Wang et al. 2017). For instance, chemical oxidation offers ease of operation and suitability for large-scale production but is associated with nonuniform size distribution. Microwave synthesis provides a short reaction time and easy size control, yet it incurs a high energy cost. Hydrothermal treatment is known for high quantum efficiency and cost-effectiveness but yields lower quantities of carbon dots (Wu et al. 2021). Electrochemical oxidation, while offering high-purity carbon dots with controllable size and good reproducibility, requires careful optimization for high yields. Selecting the most appropriate synthesis approach depends on specific requirements and considerations in the production of carbon dots.

## **Purification of carbon dots**

The purification of carbon dots is a pivotal step to ensure their quality and suitability for diverse applications. Various purification methods have been proposed in the literature, each offering unique advantages to address specific challenges related to impurities and by-products, including dialysis, ultrafiltration, and column chromatography. González-González et al. (2022) proposed a dialysis methodology to purify carbon dots synthesized by a chemical oxidation method for 360 h. This approach underscores the importance of overcoming purification challenges to obtain high-quality carbon dots. Similarly, Essner et al. (2018) conducted carbon dot syntheses using hydrothermal and microwave routes, employing citric acid paired with urea or ethylenediamine as a nitrogen source. They followed this with purification steps involving dialysis or ultrafiltration, highlighting the significance of purification in ensuring the quality of the resulting carbon dots. Furthermore, Sato et al. (2022) demonstrated the purification of surface-modified carbon dots using silica gel column chromatography, resulting in an increased photoluminescence quantum yield. This purification method illustrates the impact of purification on enhancing the optical properties of carbon dots. Additionally, Otten et al. (2022) emphasized the role of purification in removing impurities and enhancing the optoelectronic properties of carbon dots,

underscoring the importance of consistent characterization during the purification process.

# Application of carbon dots in the food industry

Carbon dots are a class of nanomaterials with unique properties, such as fluorescence, biocompatibility, and antimicrobial activity (Zhang et al. 2022). These properties make carbon dots promising candidates for a variety of applications in the food industry, including (1) developing probes and sensors for detecting contaminated toxins and other hazardous substances, (2) developing antimicrobial and antifungal food packaging materials, (3) detecting foodborne pathogens and (4) measuring the essential nutrients in food, which were visualized in Fig. 3. Residuals of these toxic elements can be found in any steps of food production, such as cultivation, production, storage, transportation, and consumption (Hoehl et al. 2012). Several harmful contaminated substances in food products were listed by the European Commission such as metal ions, mycotoxins, plant toxins, processing contaminants, banned additives, organic pollutants, and pesticides (EU-Commission 2023), which need to be detected and controlled within certain limits.

#### **Probes and sensors**

#### **Detection of metal ions**

The most concerning metal ions include chromium, lead, mercury, arsenic, and cadmium (EFSA 2023). Mercury (II) ion is one of the contaminants in milk that causes irreversible damage to neurological and renal systems. It is known as a neurotoxicant and a poison for the liver and kidneys (Wise et al. 2022). The presence of lead (II) ions in drinking water can cause neurodevelopmental effects on children (Levallois et al. 2018), whereas cadmium (II) ions present in rice can also pose a health risk like "itai-itai disease," a bone disease (Yu et al. 2017). These metal ions can be contaminated in foods and accumulated over time, causing actual disease.

The development of carbon dots-based sensors to detect metal ions has several advantages over traditional methods, which reported higher sensitivity and their ability to be used to detect metal ions in real time (Shi et al. 2019). Carbon dots are fluorescent nanomaterials that can be functionalized to bind to specific metal ions. When carbon dots bind to a metal ion, its fluorescence is quenched. This change in fluorescence can be used to detect the presence of metal ions in food samples, which was reported in various studies (Table 1). Carbon dots-based probes and sensors have shown a significant effect on detecting these commonly contaminated metal ions in water with a low

## **Applications of Carbon Dots in the Food Industry**



Fig. 3 Applications of carbon dots in the food industry. Carbon dots-based sensors monitor primary nutrients such as carbohydrates, amino acids, peptides, vitamins, and bioactive compounds. Carbon dots are incorporated into food packaging to enhance quality or monitor the freshness of foods. Carbon dots sensors detect food con-

taminants such as metal ions, residual pesticides, or food additives. Carbon dots are used to detect foodborne pathogens indirectly and directly, including *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtillis*, or *Salmonella typhimurium* 

limit of detection, including chromium (VI) with 2.3 nM, lead (II) with 0.08 ppb, cadmium (II) with 0.29 nM, iron (III) with 0.07  $\mu$ M and 63 nM (Du et al. 2023; Pandey et al. 2020; Sahu and Khan 2020; Zhang et al. 2020; Zhao et al. 2021b). Lately, multifunctional fluorescent probes have been developed to detect metal ions simultaneously to reduce time with low detection limits. For example, lead (II) and iron (III) could be detected in water simultaneously by dual emission carbon dots based on the different fluorescent intensity changes with a detection limit of 0.80 ppm and 4.74 ppm, respectively (Hoang et al. 2023). Furthermore, modifying the surface of nitrogen-carbon dots can lower the detection limit of the sensor to detect mercury (II) to 10 nM (Aziz et al. 2019). Carbon dots-based sensors to detect metal ions have been developed using two main quenching mechanisms, illustrated Fig. 4 based on the detailed explanation (Albrecht 2008). Most of these sensors were developed based on fluorescent quenching, where metal ions combine with carbon dots, reducing fluorescence (Zhang et al. 2022). Briefly, one common mechanism is static quenching (Fig. 4a). Initially, the fluorophore and metal ions (the quenchers) form a nonfluorescent complex (Fig. 4b), which results from the diffusion of both molecules, which is a time-dependent reaction. When this complex is excited with energy, it reaches the excited state. It comes back immediately to the ground state without the emission of photos but in the form of fluorescence (Fig. 4c). This difference between the non-fluorescent

| Type of analyte | Analyte                     | Probes   | Linear range | Limit of detection                          | Measured sample                    | References                 |
|-----------------|-----------------------------|--|--------------|---|------------------------------------|----------------------------|
| Metal Ions      | Zinc (II)                   | Chelation-enhanced carbon dots                 | 0.02–5 μM    | 5 nM  | Fetal bovine serum,<br>milk powder | Lu et al. 2023             |
|                 | Mercury (II)                | Nitrogen–Carbon<br>dots                        | 2–14 µM      | 0.44 µM                                     | Breast milk                        | Pajewska-Szmyt et al. 2020 |
|                 | Chromium (VI)               | Nitrogen–Carbon<br>dots                        | 0.01–4.5 μM  | 2.3 nM                                      | Tap water                          | Zhang et al. 2020          |
|                 | Iron (III)                  | Nitrogen–Carbon<br>dots                        | 0.8–27 µM    | 0.07 μΜ                                     | Environmental water                | Zhao et al. 2021b          |
|                 | Iron (III), lead (II)       | Carbon dots                                    | 0–128 µM     | 63 nM                                       | Tap water                          | Du et al. 2023             |
|                 | Lead (II), cadmium          | Carbon dots                                    | 10–200 µM    | 59 µM                                       | -                                  | Boobalan et al. 2020       |
|                 | (II)                        | Carbon dots                                    | 1–8 nM       | 0.29 nM                                     | Tap water, pond water              | Pandey et al. 2020         |
|                 | Cobalt (II)                 | Nitrogen and Sulfur<br>co-doped carbon<br>dots | 1–50 µM      | 26 nM                                       | -                                  | Sun et al. 2021            |
|                 | Cobalt (II), Copper<br>(II) | Nitrogen and Sulfur<br>co-doped carbon<br>dots | 1–100 µM     | 200 nM                                      | Water                              | Bisauriya et al. 2022      |
|                 | Copper (II), iron (II)      | Nitrogen-Doped<br>Carbon Dots                  | 0–40 µM      | Copper (II): 0.5 μM<br>Iron (II): 0.31 μM   | Animal feed, tablets               | Guo et al. 2023            |
|                 | Lead (II), iron (III)       | Dual emission car-<br>bon dots                 | 0–100 ppm    | Lead (II): 0.80 ppm<br>Iron (III): 4.74 ppm | Water                              | Hoang et al. 2023          |
|                 | Manganese (II)              | Carbon dots                                    | 0–500 µM     | 0.58 μΜ                                     | Tap water                          | Kumar et al. 2023          |

Table 1 Recent studies on carbon dots sensors for detecting metal ions

Carbon dots are used to develop sensors that detect numerous metal ions in foods and waters with high sensitivity. Doping carbon dots-based sensors is a common technique to enhance their sensitivity. The growing interest in the simultaneous detection of multiple metal ions is high-lighted



Fig.4 Mechanisms of carbon dots sensors for detecting metal ions in foods. (a) Static Quenching: The fluorescence of carbon dots is quenched by metal ions forming a non-fluorescent complex (b).

This non-fluorescent complex becomes fluorescent under excitation light (c). (d) Dynamic Quenching: The fluorescence of carbon dots changes due to collisions with metal ions (e)

metal ion complex state and the fluorescent metal ion complex state gives the concentration of the unknown metal ion in the food samples (Albrecht 2008). Another common mechanism is dynamic quenching (Fig. 4d), in which metal ions (quencher) are added to the fluorophore. The non-fluorescent complex is formed due to the collision between the quencher and the fluorophore (Fig. 4e). The unknown metal ion (quencher) concentration can be determined by measuring the difference between the fluorescence (Galletto and Bujalowski 2002).

#### **Detection of pesticide residues**

Pesticides can be categorized into four main classes, including herbicides, fungicides, insecticides, and acaricides (Fernandes et al. 2023). Pesticide residues can not only cause disease but also lead to death beyond a specific limit. This information was well-summarized by El-Nahhal and El-Nahhal (2021), who stated that exposure to pesticides can cause reproductive toxicity, cardiotoxicity, an increased risk of various types of cancers, and Parkinson's disease. Pesticide residues in tea have become a threat to human health; therefore, they are strictly controlled with specific maximum limits detailed in the legislation of the European Union, USA, and Japan (Fernandes et al. 2023).

Unlike the mechanism of carbon dots-based sensors to detect metal ions, these sensors detect pesticides based on an "off-on-off" quenching mechanism, which is illustrated in detail in Fig. 5 (Shi et al. 2019). There are two main pathways to explain the mechanism of this sensor, where both start from forming the non-fluorescent complex between

carbon dots with metal ions or nanoparticles (Fig. 5a). For the first pathway, carbon dots were fluorescent again when pesticides were combined with metal ions or nanoparticles bound with carbon dots (Fig. 5b). The change in fluorescent intensity was converted to the residual pesticide concentration in food samples. Enzymes play a central role in the second pathway (Fig. 5c). Enzymes react with substrates, forming the products of enzyme reaction, and then they will bind with metal ions or nanoparticles bound with carbon dots, making carbon dots fluorescent again (Fig. 5d). When the sensors interact with pesticides, pesticides will inactivate the enzyme reaction so that the metal ions and nanoparticles are combined again with carbon dots, forming a non-fluorescent complex (Fig. 5e).

The design of carbon dots-based sensors to detect pesticides was modified depending on the specific case; however, all cases were based on the "on-off" principles of carbon dots, as illustrated in Fig. 5. The fluorescence of carbon dots could be turned on or turned off by different compounds like metal ions, nanoparticles, or products of enzyme reactions, described in detail by Shi et al. (2019). While Chen et al. (2020) developing a carbon dots sensor with gold nanoparticles, enzymes can be used to develop these sensors to detect pesticides, which could act as fluorescent probes or quenchers in enzyme-based reactions (Huang et al. 2019). These sensors detect changes in the fluorescence of carbon dots caused by the presence of pesticides in a sample, enabling the determination of the pesticide concentration (Zhang et al. 2022). Cao and Guo (2024) developed carbon dots sensors to detect imidacloprid based on Förster Resonance Energy Transfer (FRET) methods, where the emission



**Fig. 5** Mechanisms of carbon dots sensors based on "off–on-off" quenching for detecting pesticides, modified from Shi et al. (2019) with two proposed mechanisms. In the first mechanism, fluorescent carbon dots react with metal ions to form a non-fluorescent complex (**a**). The fluorescent turns again in the presence of pesticides as the metal ions preferentially react with the pesticides (**b**). In the second

mechanism, metal ions react with the enzymatic product instead of carbon dots, turning on the fluorescence (c). When pesticides are present, they inhibit the enzyme–substrate reaction, preventing the formation of the fluorescent product and causing the fluorescence of the carbon dots to turn off again by forming a non-fluorescent complex (d and e)

spectra of carbon dots overlap with the absorption spectrum of Imidacloprid, resulting in fluorescent quenching of carbon dots. Table 2 lists the recently published studies about carbon dots-sensors for detecting pesticides.

Carbon dots-based sensors have shown potential in rapidly detecting pesticides with broad linear correlations and reasonable detection limits. Carbon dots were applied to detect several pesticides in water, including carbaryl (Chen et al. 2020) and dimethoate (Liu et al. 2020). Furthermore, imidacloprid, a pesticide contaminant in vegetables, was detected with a carbon dots-based sensor with a detection limit of 1.87 ng/kg (Cao and Guo 2024). In terms of herbicides, glyphosate, one of the most common herbicides worldwide, was detected in cucumber, pepper, and ginger with carbon dots- cadmium telluride probes with excellent linear correlations ranging from 10 to 1000 nM and a detection limit of 2 pM (Bera and Mohapatra 2020). Yang et al. (2023) synthesized carbon dots from mulberry leaves and sodium hydroxide, subsequently, a carbon dotssensor was developed to simultaneously detect glyphosate and parathion-methyl in herbal samples with the detection limit of 0.60  $\mu$ M and 0.14  $\mu$ M, respectively. Recently, Vadia et al. (2023) studied a multipurpose sensor to detect iron (III) ion and propiconazole in pharmaceutical and vegetable samples with a linear correlation range from 0.5 to 180  $\mu$ M for iron (III) and 0.1–40  $\mu$ M for propiconazole with the detection limit of 0.18  $\mu$ M and 0.054  $\mu$ M, respectively.

 Table 2
 Recent studies on carbon dots sensors for pesticide detection

| Type of analyte            | Analyte   | Probes                                       | Linear range                     | Low of detection                                 | Measured sample  | References                    |
|----------------------------|---|--|----------------------------------|--|--|-------------------------------|
| Pesticides                 | Carbaryl  | Red emissive carbon dots                     | 0–20 μg/mL                       | 0.52 ng/mL                                       | Tap water, lake<br>water                                 | Xu et al. 2024                |
|                            | Dimethoate  | Carbon dots /<br>Dithizone                   | 0.15–5 μM                        | 0.064 µM   | River water, farm water                                  | Liu et al. 2020               |
|                            | Imidacloprid  | Carbon dots                                  | 0.037–0.2 mg/L                   | 0.00187 mg/kg                                    | Lettuce, cole,<br>spinach, and<br>pakchoi with<br>spiked | Cao and Guo 2024              |
|                            | Organochlorine<br>pesticides:                               | The surface-<br>engineered                   | 1–19 µM                          | Heptachlor:<br>0.002 µM                          | Water, soybean sprout, mung                              | Nethaji et al. 2024           |
|                            | Heptachlor,<br>Endosulfan,                                  | fluorescent blue<br>emissive-carbon<br>dot   |                                  | Endosulfan:<br>0.099 µM                          | bean sprout  |                               |
|                            | Chlordimeform<br>and 2,4-dichlo-<br>rophenoxyacetic<br>acid |  |                                  | Chlordimeform:<br>0.16 μM                        |  |                               |
|                            |   |  |                                  | 2,4-dichlorophe-<br>noxyacetic acid:<br>0.082 µM |  |                               |
| Herbicide                  | Glyphosate  | Carbon dots / Cad-<br>mium telluride         | 10–1000 nM                       | 2 pM   | Cucumber, pep-<br>per, ginger                            | Bera and Mohapa-<br>tra 2020  |
|                            | Glyphosate  | A dual emissive<br>carbon dot                | 0–10 ppm                         | 0.03 ppm   | Water  | Clermont-Paquette et al. 2023 |
| Herbicide,Pesticide        | Glyphosate,<br>Parathion-methyl                             | Carbon dots                                  | Glyphosate:<br>1.0–110.0 µM      | Glyphosate:<br>0.60 µM                           | Food/herbal samples                                      | Yang et al. 2023              |
|                            |   |  | Parathion-methyl:<br>0.3–65.0 µM | Parathion-methyl:<br>0.14 µM                     |  |                               |
| Metal Ions, Pesti-<br>cide | Iron (III) ion,<br>Propiconazole                            | Carbon dots                                  | Iron (III) ion:<br>0.5–180 μM    | Iron (III) ion:<br>0.18 μM                       | Pharmaceutical,<br>and vegetable                         | Vadia et al. 2023             |
|                            |   |  | Propiconazole:<br>0.1–40 μM      | Propiconazole:<br>0.054 µM                       | samples  |                               |
| Fungicide                  | Dodine  | Bamboo stem bio-<br>mass carbon dots         | 0.1–10.0 nM                      | 4.3 nM   | Peach, plum,<br>apple, onion<br>leaf, kidney<br>beans    | Adaikalapandi et al.<br>2024  |
|                            | Thiophanate-<br>methyl                                      | Yellow-green fluo-<br>rescent carbon<br>dots | 0–10 µM                          | 50.7 nM  | Pear, orange<br>tomato                                   | Wang et al. 2024              |

Various studies focus on developing carbon dots-based sensors to detect pesticides, herbicides, and fungicide residues in food samples. The primary objectives in this field include increasing sensor sensitivity, lowering detection limits, and developing sensors capable of detecting multiple pesticides simultaneously

### **Detection of food additives**

Food additives have been used in many processed foods for several years to maintain their structure, enhance their appearance, and improve their flavor. While food additives can have remarkable benefits for the food industry, consuming large amounts of these compounds can have adverse effects on food quality and even consumer health. Additionally, several food additives have been banned in some countries due to health concerns. There are varied ways to produce carbon dots sensors to detect food preservatives, however, static quenching (Fig. 4) and "off–on-off" quenching (Fig. 5) are the two most common methods, which were described in subSects. "Detection of metal ions" and "Detection of pesticide residues". Sensors made from carbon dots can be used to detect a variety of food preservatives, including nitrile, formaldehyde, borax, and sulfite, listed in Table 3. Nitrile, a common preservative used to extend the shelf life of meat products, can be rapidly detected with carbon dots sensors with a linear range of 0–4.3  $\mu$ M and a limit of detection of 0.5 nM (Hu et al. 2019). Formaldehyde, a banned preservative in several countries, is still used in some food products, such as noodles, fish, and meat, to increase elasticity and shelf life. Nitrogen, phosphorous-carbon dots can detect formaldehyde in sprouted beans with a detection limit of 0.47  $\mu$ M (Qu et al. 2020). Borax, which has been

Table 3 Recent studies on carbon dots sensors for detecting food additives

| Type of analyte                    | Analyte                             | Probes   | Linear Range  | Low of detection                         | Measured sample  | References               |
|------------------------------------|-------------------------------------|--|---------------|--|--|--------------------------|
| Preservatives                      | Nitrite                             | Crystal violet car-<br>bon dots                              | 0–20 mg/L     | 0.6 mg/L                                 | Mustard, sausage,<br>water                                   | Liu et al. 2024          |
|                                    |                                     | Carbon dots  | 0–4.3 μM      | 0.5 nM                                   | Ham  | Hu et al. 2019           |
|                                    |                                     | Carbon dots  | 0.1–100 μM    | 31.6 nM                                  | Bacon, sausage,<br>pickle, milk<br>samples                   | Liu et al. 2019          |
|                                    | Potassium sorbate,<br>vitamin B12   | Boron-carbon dots  | 0.2–24 µM     | 6.1 nM                                   | Vinegar and bread  | Jia et al. 2019          |
|                                    | Formaldehyde                        | Nitrogen, phospho-<br>rous-carbon dots                       | 0–40 µM       | 0.5 μΜ                                   | Sprouted beans   | Qu et al. 2020           |
|                                    | Borax                               | Carbon dots  | 100–500 μM    | 1.5 μΜ                                   | Fishball   | Prathumsuwan et al. 2019 |
|                                    | Bisulfite, sulfite                  | Carbon dots /<br>zeolitic imida-<br>zolate framework         | 10 μM-8.5 mM  | 2.7 μΜ                                   | Sugar  | Wang et al. 2023         |
|                                    |                                     | Carbon dots / silver<br>nanoparticles /<br>hydrogen peroxide | 20–200 µM     | 3 μΜ                                     | Food samples and herbs                                       | He et al. 2023           |
| Artificial colors                  | Tartrazine (Yellow #5)              | Nitrogen-carbon<br>dots / Iron (III)<br>ion                  | 0.10–30.00 μM | I 48 nM                                  | Orange juice   | Yang et al. 2020         |
|                                    | Sunset yellow (Yel-<br>low #6)      | Nitrogen-carbon<br>dots                                      | 0.5–40 µM     | 28 nM                                    | Soft drinks, wine,<br>sugar rolls, dried<br>plum, swiss roll | Su et al. 2022           |
|                                    | Sudan Red II                        | Nitrogen, oxygen-<br>carbon dots                             | 0-8 mg/L      | 0.6 mg/L                                 | Spiked spice sam-<br>ples                                    | Ramoğlu et al. 2021      |
|                                    | Sudan Red I                         | Nitrogen, phospho-<br>rous-carbon dots                       | 43 nM–52 μM   | 43 nM                                    | Paprika  | Zhao et al. 2021b        |
|                                    | Indigo Carmine                      | Nitrogen-carbon<br>dots                                      | 0.73–10 μM    | 0.24 µM                                  | Fruit juice and soft drink                                   | Ali et al. 2021          |
|                                    |                                     | Europium-carbon<br>dots                                      | 1.5–10 μg/ml  | 0.4 µg/ml                                | Juice samples  | Albalawi et al. 2023     |
|                                    | Sunset Yellow                       | Carbon dots  | 0–60 µM       | 0.4 µM                                   | -  | Huang et al. 2017        |
| Artificial colors<br>and Metal Ion | Allura red (Red<br>#40), Iron (III) | Carbon dots  | 0–30 µM       | Red #40: 0.61 μM;<br>Iron (III): 0.26 μM | Juice, syrups, tap<br>water, wastewater,<br>distilled water  | Vijeata et al. 2022      |

Carbon-dot-based sensors detect various food additives, including preservatives and artificial colors. This is essential for maintaining food safety and mitigating consumer risks. Doping carbon dots in these sensors increases their sensitivity. A wider range of carbon dots has been studied and measured for this purpose

used as a food preservative in some countries to prevent the growth of bacteria and molds, is now illegal for use in foods in the USA following regulations from the Food and Drug Administration (FDA 2023). The combination of carbon dots and silver nanoparticles for biosensors can detect both sulfite and bisulfite in several food and herb samples with a detection limit of  $3.02 \,\mu$ M (He et al. 2023). Although carbon dots-based sensors can find the presence of several food preservatives, there is still early research about this, there are hundreds of preservatives that need to be studied.

Not like preservatives, carbon dots can detect almost primary artificial colors in food including tartrazine and sunset yellow for yellow color; sundan red I and sundan red II for red color; Indigo carmine for blue color (Albalawi et al. 2023; Ali et al. 2021; Ramoğlu et al. 2021; Su et al. 2022; Yang et al. 2020; Zhao et al. 2021a). For example, in India, sudan red I and sudan red IV are permitted for use in certain food products, such as chili powder and turmeric powder, which can be measured using carbon dots-based sensors with low detection on 43 nM for sudan red I (Zhao et al. 2021a) and 0.6 mg/L for sudan red II (Ramoğlu et al. 2021). Tartrazine was detected in orange juice by nitrogen-carbon dots / iron (III) ion sensor with a linear range from 0.1 to  $30 \mu$ M with a detection limit of 48 nM (Yang et al. 2020).

## Active packaging

The mechanism of carbon dots-based packaging is based on two main properties of carbon dots: their sensitivity to different pH and their antimicrobial and antioxidant activity. The development of active packaging with carbon dots is still in the early stages, but the reported results are remarkable, which are listed in Table 4. Due to their sensitivity to pH, the fluorescence of carbon dots could

Table 4 Recent studies on carbon dots sensors for active packaging

| Packaging materials  | Research Objects   | Results   | References              |
|--|--------------------|---|-------------------------|
| Carbon dots / chitosan / gelatin-based compos-<br>ite films                    | Avocado            | Extended shelf life of more than 14 days; Anti-<br>microbial activity on <i>Listeria mon ytogenes</i><br>and <i>Escherichia coli</i> ; Antifungal activity on<br>mold ( <i>Aspergillus flavus</i> and <i>Colletotrichum</i><br><i>orbiculare</i> ); negligible cytotoxicity to L929<br>cells (after 72 h) | (Ezati et al. 2022)     |
| Carbon dots / cellulose nanofiber / essential oil nanoemulsion / gelatin films | Tomato             | Extended shelf life up to 6 days; Antimicrobial effects against <i>Escherichia coli</i>   | (Bao et al. 2023)       |
| Nitrogen-carbon dots / chitosan  | Pork               | Maintained pork freshness; Antioxidant activ-<br>ity; Antibacterial activity of <i>Staphylococcus</i><br><i>aureus</i> and <i>Escherichia coli</i>  | (Lin et al. 2022)       |
| Carbon dots / carboxymethyl cellulose / agar-<br>based film                    | Not available      | The addition of carbon dots reduced the<br>mechanical strength and surface hydropho-<br>bicity, maintained the water vapor barrier<br>and improved UV blocking, antioxidant, and<br>antibacterial activities  | (Tammina and Rhim 2023) |
| Carbon dots / chitosan   | Blueberries        | Extended shelf life until 15 days; Stabilized<br>the anthocyanin content (chitosan / 3%<br>nitrogen-carbon dots); Delayed the spoilage  | (Chen et al. 2023)      |
| Zinc-carbon dots / carrageenan films   | Shrimps            | UV-blocking; Antimicrobial activity; Antioxi-<br>dant activity; Extended shelf life   | (Khan et al. 2024)      |
| Carbon dots / alginate   | fiordilatte cheese | Against <i>Escherichia coli</i> ; Extended the shelf life until 10 days   | (Lacivita et al. 2023)  |
| Carbon dots / antimicrobial bacterial cellulose membranes                      | Ground beef        | Significantly reduced the growth of <i>Escherichia coli</i>   | (Ghorbani et al. 2024)  |
| Carbon dots / gelatin / poly(vinyl alcohol)-<br>based functional films         | Ground pork        | Improved shelf life during the 48 h storage period at 20 °C; Prevented bacteria growth  | (Min et al. 2023)       |
| Carbon dots / anthocyanin / cellulose nanofiber                                | Pork, fish, shrimp | Improved UV barrier; Maintained antioxidant properties  | (Wagh et al. 2023)      |
| Carbon dots / alginate   | Banana             | Enhanced surface hydrophobicity of films;<br>Damaged slightly gas barrier proper-<br>ties; Increased the anti-browning ability;<br>Extended the shelf life  | (Mao et al. 2023)       |

Incorporating carbon dots with other packaging materials enhances packaging quality by increasing UV-blocking capabilities. Additionally, carbon dots improve the antimicrobial activity of packaging, thereby extending the shelf life of various food types, including meat products, seafood, and fruits

be changed depending on different pHs. Carbon dots have been combined with several packaging materials, such as chitosan, gelatin, and alginate to create active packaging that can be controlled by changes in fluorescence (Bao et al. 2023; Chen et al. 2023; Lacivita et al. 2023). Combining carbon dots and chitosan as modified active packaging for fresh-cut cucumber has been shown to significantly prolong the overall appearance and inhibit the growth of Staphylococcus aureus and Escherichia coli with increasing carbon dots concentration (Fan et al. 2019). Carbon dots have also been used as an edible coating in combination with alginate, which doubled the shelf life of fiordilatte cheese to 10 days (Lacivita et al. 2023). Moreover, carbon dots-based packaging has been shown to positively extend the shelf life of fruits, including avocados up to 14 days (Ezati et al. 2022), tomatoes up to 6 days (Bao et al. 2023), and blueberries up to 15 days at 25 °C (Chen et al. 2023). Adding carbon dots in the packaging even prevented the browning reaction of banana peel during ripening (Mao et al. 2023).

The incorporation of carbon dots in active packaging created a positive effect for not only fruit products but also meat products. The freshness of pork can also be monitored by the increase in fluorescent intensity of packaging made from carbon dots and chitosan (Lin et al. 2022). Carbon dots added into antimicrobial bacterial cellulose membranes significantly reduced the growth of Escherichia coli and extended the shelf life of ground beef up to 9 days at 4 °C (Ghorbani et al. 2024). In addition to its antioxidant and antimicrobial activity, carbon dots-based packaging can also maintain UV-blocking properties (Khan et al. 2024; Tammina and Rhim 2023). Tammina and Rhim (2023) concluded that incorporating carbon dots into an agar-based film can reduce the film's mechanical strength and surface hydrophobicity, but the effects on the mechanical strength of films may depend on the type of film. For example, in Khan et al. (2024) it was reported that zinc-carbon dots did not affect the strength of the carrageenan-zinc-carbon dots film.

#### Foodborne pathogen detection

Foodborne pathogens are a crucial problem that needs to be controlled in food safety. In recent years, researchers have made significant advancements in the development of detection methods for foodborne pathogens using carbon dots. Carbon dots were reported to detect several major foodborne pathogens including Staphylococcus aureus (Gao et al. 2024; John et al. 2020; Zhao et al. 2021a), Escherichia coli (John et al. 2020; Zhao et al. 2021a). The representative studies focusing on foodborne pathogen detection of carbon dots are shown in Table 5. Aflatoxin B, a main toxic produced by certain molds, particularly Aspergillus flavus and Aspergillus parasiticus, also can be detected with carbon dots probes (Li et al. 2024). The antimicrobial properties of carbon dots, combined with their nanoscale size, make them an ideal candidate for pathogen detection. Carbon dots have a core of carbon nanoparticles and a thin surface passivation layer, allowing them to interact with bacteria and toxins effectively (Abu et al. 2020). Research has shown that carbon dots can be used in conjunction with other nanomaterials, such as dyefilled nanoparticles, magnetic nanomaterials, and silver nanoshells, to enhance detection sensitivity and reduce detection time (Gao et al. 2024; John et al. 2020).

The integration of nanosensors into food packaging has emerged as a key application in the detection of pathogens and toxins in food products. Fluorescent nanoparticles, such as carbon dots, have shown great potential in this field. For example, studies have demonstrated that carbon dots coupled with immunomagnetic separation can effectively detect foodborne pathogenic bacterial species like *Salmonella typhimurium* and *Escherichia coli* in milk and apple juice (Zhao et al. 2009). Another approach to detecting the presence of *Escherichia coli, Staphylococcus aureus, Bacillus subtilis,* and *Proteus vulgaris* in food products is through pH-sensitive detection (Pathak et al. 2020). This change can be easily detected and quantified, providing a rapid and sensitive method for pathogen detection.

| Foodborne pathogen    | Probes                            | Minimum Inhibitory Concen-<br>tration/ Low of detection | Measured sample | References        |
|-----------------------|-----------------------------------|---|-----------------|-------------------|
| Aflatoxin B           | Nitrogen-doped carbon dots        | 77 pg/mL  | Coix seed       | Li et al. 2024    |
| Escherichia coli      | Nitrogen, phosphorous-carbon dots | 0.5 mg/mL   | paprika         | Zhao et al. 2021a |
|                       | Carbon dots / amoxicillin         | $1.5 \times 10^{-8}$ CFU/ml                             | _               | John et al. 2020  |
| Staphylococcus aureus | Carbon dots / amoxicilin          | $1.5 \times 10^{-8}$ CFU/ml                             | _               | John et al. 2020  |
|                       | Carbon dots / manganese dioxide   | 9 CFU/mL  | _               | Gao et al. 2024   |
|                       | Nitrogen, phosphorous-carbon dots | 7.5 μg/mL   | paprika         | Zhao et al. 2021a |

Table 5 Recent studies on carbon dots sensors for detection of foodborne pathogens

Carbon dots sensors can detect foodborne pathogens, including aflatoxin B, *Escherichia coli*, and *Staphylococcus aureus*. In addition to detecting pathogens, staining bacteria with carbon dots solutions can differentiate between bacterial types

## **Monitoring of food nutrients**

Food nutrient monitoring is essential for ensuring the quality and safety of food products. Additionally, it plays a crucial role in assessing the nutritional content of food and enabling individuals to make informed dietary choices. Traditional methods for food nutrient monitoring, such as ion exchange chromatography and mass spectrometry, often require complex procedures, expensive equipment, and lengthy analysis times. Therefore, the development of rapid measurement for control of the nutrient content in foods is necessary. One promising food nutrient monitoring approach is carbon dots (Table 6). Carbon dots have been used to develop novel nanosensors and nano biosensors for food quality control including glucose, amino acids, and vitamins  $B_2$  (Lin et al. 2019; Rossini et al. 2019; Tabaraki and Abdi 2019; Yuxin et al. 2022). These carbon-based nanomaterials have shown great potential in enhancing the sensing signals and improving the accuracy of food nutrient monitoring devices (De Paula et al. 2019). Besides the essential nutrients, the combination of carbon dots detects other nutrients like quercetin, curcumin, and tea polyphenol (De Paula et al. 2019; Han et al. 2019; Wei et al. 2020).

## Food safety assessments of carbon dots

Besides the remarkable application of carbon dots in the food industry, the food safety of carbon dots needs to be fully understood. One risk is their potential cytotoxicity, which refers to the ability of carbon dots to cause harm or damage to living cells at a certain dose. Furthermore, the genotoxicity of carbon dots is also an important aspect to be aware of, as it refers to their potential to damage genetic material and cause mutations. The accumulation and distribution of carbon dots in biological systems also need to be discussed. The overall food safety assessment of carbon dots is illustrated in Fig. 6.

Table 6 Recent studies on carbon dots sensors for monitoring food nutrients

| Type of nutrients        | Nutrients                  | Monitoring methods  | Linear range                            | Low of detection                     | Samples           | References             |
|--------------------------|----------------------------|---|---|--------------------------------------|-------------------|------------------------|
| Carbohydrates            | Glucose                    | Carbon dots /<br>glucose oxidase<br>/ horseradish<br>peroxidase       | 10 <sup>-6</sup> -10 <sup>-5</sup> M    | _                                    | Biofluids         | Rossini et al. 2019    |
|                          | Gluocose, glu-<br>tathione | Silver-Carbon dots<br>/ peroxidase                                    | Gluocose:<br>50–800 µM                  | Gluocose:<br>11.30 µM                | Saliva, urine     | Haiyang et al. 2024    |
|                          |                            |   | Glutathione:<br>1–60 μM                 | Glutathione:<br>3.54 µM              |                   |                        |
|                          | Glucose, lactase           | Carbon dots /<br>oxidase  | Gluocose:<br>21–38×10 <sup>-6</sup> M   | Gluocose:<br>2.60×10 <sup>-6</sup> M | Saliva            | Rossini et al. 2021    |
|                          |                            |   | Lactate: 1.0–<br>7.5×10 <sup>-4</sup> M | Lactate:<br>8.14×10 <sup>-7</sup> M  |                   |                        |
|                          | Glucose                    | Iron-Carbon dots /<br>tetramethylben-<br>zidin / hydrogen<br>Peroxide | 0.08–10.00 mM                           | 0.029 mM                             | Urine             | Yuxin et al. 2022      |
| Amino acids,<br>Peptides | Aspartic acid              | Nitrogen-Carbon<br>dots   | 0.5–50 μΜ                               | 90 nM                                | Sport supplements | Tabaraki and Abdi 2019 |
| Vitamins                 | Riboflavin                 | Zinc and Chlorine<br>Co-doped carbon<br>dots                          | 0–10 µg/mL                              | 12.5 ng/mL                           | Apple juice       | Meng and Wu 2024       |
| Others                   | Quercetin                  | Nitrogen- and<br>Sulfur-codoped<br>carbon dots                        | 0–29.7 μM                               | 17.3 nM                              | Red wine, onion   | Sasikumar et al. 2023  |
|                          | Curcumin                   | Nitrogen- and<br>Sulfur-codoped<br>carbon dots                        | 2.0–18.0 μM                             | 0.04 µM                              | Urine             | Han et al. 2019        |
|                          | Polyphenol                 | Carbon dots /<br>copper(II) ion                                       | 1–30 µM                                 | 0.31 µM                              | Green tea         | Wei et al. 2020        |

Carbon dots-based sensors can quantify the nutrient content in foods, such as carbohydrates, amino acids, vitamins, and other bioactive compounds. Doping carbon dots with nitrogen, sulfur, silver, or copper enhances sensor sensitivity. Sensors are designed to optimize performance depending on the specific nutrient being detected



Fig. 6 Food safety assessment of carbon dots. The cytotoxicity of carbon dots is influenced by various factors such as size, dosage, precursors, and surface passivation agents. The genotoxicity of carbon dots is primarily associated with dosage and does not cause acute effects

## Cytotoxicity

Carbon dots, while generally considered to have lower cytotoxicity than semiconductor quantum dots, can still exhibit cytotoxic effects depending on various factors involved in their synthesis and design (Hola et al. 2014; Sahu et al. 2012). Wang et al. (2011) emphasized in a systematic review of the cytotoxicity of carbon dots that while carbon dots have shown great potential for imaging applications, their cytotoxicity needs to be carefully evaluated to ensure their safe use. The cytotoxicity of carbon dots was affected by several factors, including dose, precursor materials, surface passivation agents, and size of carbon dots. First, the cytotoxicity of carbon dots was affected by the precursor materials. For instance, different organic precursors like glucose, sucrose, glycol, glycerol, and citric acid can introduce impurities or toxic components during the carbonization process, thereby affecting the cytotoxicity of the resulting carbon dots (Sahu et al. 2012). Additionally, the composition of carbon dots has been identified as a key determinant of their cytotoxic behavior. Studies have shown that carbon dots synthesized from natural precursors, such as orange juice, demonstrated excellent biocompatibility and low cytotoxicity (Sahu et al. 2012). The cytotoxicity of carbon dots was also dose-dependent (Li et al. 2018a). The results of this study indicated that 80% of cells survived at 1 mg/mL, whereas 90% of cells experienced

on genetic materials. Bioaccumulation and biodistribution studies indicate that small-sized carbon dots can cross the blood-brain barrier and accumulate in the heart, brain, liver, and tumors; however, no adverse effects have been detected

death at 10 mg/mL. However, carbon dots obtained from Kvass, a type of beverage, showed no cytotoxicity below 20 mg/mL (Liao et al. 2015). Different precursor materials with different doses can affect the cytotoxicity of carbon dots.

The cytotoxicity of carbon dots is also dependent on the selection of the surface passivation agent and the size. Surface modifications with biocompatible molecules can enhance the biocompatibility of carbon dots and reduce their cytotoxic effects (Nygård et al. 2000). Mukherjee et al. (2022) mentioned that the presence of heteroatoms on the surface of carbon dots, such as amine, hydroxyl, carboxyl, or thiol functional groups, can improve their physicochemical qualities, quantum yield, and likelihood of visible light absorption, eliminating the need for additional surface passivation (Mukherjee et al. 2022). Similarly, the size of carbon dots plays a crucial role, with smaller dots exhibiting higher cellular uptake and potentially inducing cytotoxic effects (Nygård et al. 2000). The significance of size-dependent cytotoxicity has been observed in studies on carbon dot quantum dots, where smaller quantum dots displayed higher toxicity than larger ones (Derfus et al. 2004). While it is essential to acknowledge that carbon dots generally exhibit low cytotoxicity (Du et al. 2023), it's important to note that the specific cytotoxic effects can vary depending on experimental conditions and cell types (Hola et al. 2014). Some studies have indicated that high concentrations or prolonged exposure to carbon dots can lead to cell death or affect cellular functions (Hola et al. 2014; Li et al. 2018a; Liao et al. 2015). Nonetheless, such effects are typically observed at concentrations significantly higher than those typically used for imaging or therapeutic applications.

## Genotoxicity

Genotoxicity refers to the ability of a substance to cause damage to genetic material, such as DNA, leading to mutations or other genetic alterations. In the case of carbon dots, several studies have investigated their genotoxicity. Carbon dots do seem not to cause any acute damage to the genetic materials, according to the results of published articles about the genotoxicity of carbon dots. One study by Pelaz et al. (2017) evaluated the genotoxicity of carbon dots in animals. The study found that carbon dots dosages of 51 mg/kg body weight did not cause any genotoxic effects or abnormalities in the organs of the animals. Another study by Zaidi et al. (2022) conducted a biosafety evaluation of photoluminescent carbon dots produced by nitric acid oxidation. This study found no acute toxicity, genotoxicity, or abnormalities in the organs of mice. The genotoxicity depends on the dose of carbon dots. For example, Havrdová et al. (2021) investigated the genotoxic effects of carbon dots on cells. The study found that quantum carbon dots up to 400 µg/ mL concentration did not significantly affect cell viability or DNA content. However, at higher concentrations, carbon dots could lead to changes in the morphology of cells, causing cell death.

## **Bioaccumulation and biodistribution**

In terms of the accumulation and distribution of carbon dots in biological systems, Yang et al. (2009a) a study was conducted on the biodistribution of carbon dots in vivo. The study results indicated minor accumulations of carbon dots in the organs, suggesting low bioaccumulation. This finding is consistent with the absence of any significant damage to the organs. However, the study also emphasized the need for further systematic investigations on the in vivo biodistributions of different types of carbon dots (Yang et al. 2009a). Carbon dots were reported to cause neurodegenerative effects by Song et al. (2018). The study found that carbon dots were distributed in the cytoplasm but not the nucleus after 24 h of incubation. In mice experiments, carbon dots were orally administered, reaching the brain within 2 h, as observed by the increased fluorescent intensity in the mouse brain, indicating passage through the blood-brain barrier. A recent study reported that carbon dots can accumulate in both the nucleus and cytoplasm causing death for L929 cells (Shabbir et al. 2022). Li et al. (2018a) demonstrated that carbon dots derived from Maillard reaction products can readily penetrate both plant and animal cells, distributing themselves either within the cell wall or the cytoplasm.

Additionally, significant accumulation was found in the liver, brain, and heart 2-6 h after administration, indicating potential blood-brain barrier crossing for ultra-small nanoparticles smaller than 5 nm (Li et al. 2018b). Although Li et al. (2018b) proved that carbon dots can be quenched by saliva, gastric juice, and duodenal-bile juice in vitro digestion experiments, the accumulation and distribution of carbon dots need to be studied deeply to understand their fate and effects on biological systems (Li et al. 2018b). Due to this potential health risk, further investigation is necessary into the association between carbon dots and bioactive molecules. Moreover, Liao et al. (2021) focused on the in vivo biodistribution, clearance, and biocompatibility of multiple carbon dots containing nanoparticles for biomedical applications. The study investigates the accumulation and fluorescence intensity of carbon dots over time, which found that single carbon dots took 5 h to reach maximum accumulation at the tumor site, and the fluorescence intensity of carbon dots gradually decreased over time. Although the accumulation and distribution of carbon dots have been studied, the conversion and excretion of carbon dots have not been thoroughly researched to obtain a comprehensive database on how carbon dots affect our health.

## Strategies for mitigating the risk

To ensure the safe use of carbon dots in various applications, researchers have developed or proposed various mitigating strategies, including surface modification and functionalization (Yao et al. 2019), using natural precursors (Sahu et al. 2012), and controlling the size of carbon dots via different synthesis and purification methods (Derfus et al. 2004). After studying the toxicity and the possibility of accumulating carbon dots in organs, developing standardized carbon dots with a reliable analytical method must be a concern. Figure 7 illustrates the overall diagram of the four main mitigating strategies.

### Selecting the appropriate precursors

Selecting the appropriate precursors is an effective strategy for mitigating the risks associated with carbon dots. The selection of precursors can strongly influence the properties and performance of carbon dots, including their optical properties, surface chemistry, and biocompatibility (Hola et al. 2014; Peng et al. 2016; Yao et al. 2019; Zeng et al. 2021). Several carbon dots were synthesized from various precursors listed in Table 7 and divided into two main groups, including natural materials and synthetic Fig. 7 Strategies for mitigating the risk of carbon dots. 1 Selecting appropriate precursors, such as natural materials, can produce smaller carbon dots with lower cytotoxicity, thereby reducing potential toxicity. 2 Optimizing synthesis methods can significantly impact the size, morphology, surface chemistry, and optical properties of carbon dots, directly influencing their potential toxicity. 3 Surface modification by adding functional groups or incorporating heteroatom doping can enhance solubility, stability, and biocompatibility while reducing cytotoxicity. 4 Developing protocols for detecting carbon dots in foods is crucial for comprehensively understanding their formulation and assessing their toxicity



compounds. The choice of precursors can influence the structural diversity, properties, and cytotoxicity of carbon dots. Different precursors, such as small organic molecules, polymers, or biomass-derived materials can lead to the formation of carbon dots with different structures and functionalities (Zeng et al. 2021). For example, the use of different carbon sources and surface passivation agents can result in carbon dots with varied optical properties (Yao et al. 2019). Sahu et al. (2012) mentioned that using synthetic precursors can produce carbon dots with higher cytotoxicity compared with natural precursors. However, carbon dots synthesized from natural sources obtain a smaller size (1.5–8 nm) than synthetic ones (2.5–17.5 nm) (Table 7).

Furthermore, the appropriate choice of precursors can also allow for controlling other physicochemical properties of carbon dots. For instance, the use of specific synthesis precursors can influence the size, surface charge, and stability of carbon dots (Peng et al. 2016). This allows for the customization of carbon dots for different applications, such as biomedical or optoelectronic applications. Moreover, the selection of precursors can also impact the biocompatibility and toxicity of carbon dots. By choosing safe and non-toxic precursors, the resulting carbon dots can be safer for use in biological applications (Hola et al. 2014). By carefully selecting the precursors, it is possible to tailor the properties of carbon dots to meet specific application requirements and reduce the risk of carbon dots, especially in the food industry.

## **Optimizing synthesis methods**

The synthesis method can significantly impact the properties and performance of carbon dots, including their size, morphology, surface chemistry, and optical properties (Atabaev 2018; De and Karak 2013; Wang et al. 2014). As discussed in Sect. "Synthesis of carbon dots", various methods have been utilized to produce carbon dots, each with its own set of advantages and disadvantages. Hydrothermal and microwave methods are two common techniques for producing carbon dots, involving high temperatures and energy to carbonize the precursors, resulting in carbon dots with high yield and smaller in size (Table 7). Different synthesis methods, such as hydrothermal, solvothermal, microwaveassisted, or pyrolysis methods, can result in carbon dots with varying sizes and shapes (Wang et al. 2014). Our previous study reported that lowering the synthesis temperature and using assist solvents like ethanol can obtain stronger fluorescent carbon dots, smaller size without increasing toxicity (Nguyen et al. 2024). Derfus et al. (2004) reported that the smaller carbon dots displayed higher cytotoxicity, similar to the conclusion of Nygård et al. (2000) higher cellular uptake and cytotoxicity. Furthermore, carbon dots smaller than 5 nm were recorded, passing the blood-brain barriers and accumulating in the brain, heart, and liver after 2-6 h (Li et al. 2018b). By carefully selecting the synthesis method, uniform and well-defined carbon dots can be achieved, enhancing their performance and reducing the potential variability in their properties (De and Karak 2013).

## Table 7 Synthesized carbon dots in recent publications

| Precursors  | Type of precursors | Synthetic method                      | Size (nm) | Quan-<br>tumn<br>yield (%) | Excitation/ Emis-<br>sion wavelength<br>(nm) | References                 |
|---|--------------------|---------------------------------------|-----------|----------------------------|--|----------------------------|
| Aloe, Urea  | Naturals           | Hydrothermal                          | 5.0       | 60.8                       | 323/441                                      | Yang et al. 2020           |
| Bael Patra fruit  | Naturals           | Hydrothermal                          | 6.0       | 57.1                       | 321/350                                      | Vijeata et al. 2022        |
| Citric acid, Urea   | Synthetics         | Microwave                             | 10.0      | -                          | 360/450                                      | Tabaraki and Abdi 2019     |
| Citric acid, tyramine   | Synthetics         | Microwave                             | 17.5      | 11.0                       | 355/440                                      | Rossini et al. 2019        |
| Citric acid, Urea, boric acid                                 | Synthetics         | Microwave                             | 5.0       | -                          | -  | Sahu and Khan 2020         |
| Citric acid, ethylenedi-<br>amine;                            | Synthetics         | Microwave                             | 5.0       | 73.7                       | 350/445                                      | Wei et al. 2020            |
| Citric acid, melamine   | Synthetics         | Hydrothermal                          | 4.9       | 44.0                       | 352/427                                      | Pajewska-Szmyt et al. 2020 |
| Citric acid, phenylenedi-<br>amine                            | Synthetics         | Hydrothermal                          | 3.2       | 30.0                       | 385/489                                      | Chen et al. 2020           |
| Citrate, Urea   | Synthetics         | Hydrothermal                          | 4.1-6.3   | 40.0                       | -  | Zhao et al. 2021b          |
| Citric acid, acetoguan-<br>amine, trimethylene<br>glycol      | Synthetics         | -                                     | _         | 5.3                        | 350/470                                      | Ramoğlu et al. 2021        |
| Citric acid, glycine  | Synthetics         | Hydrothermal                          | 0.5-4.0   | -                          | 340/430                                      | Yuxin et al. 2022          |
| Citric acid, glucose,<br>ethylenediaminetet-<br>raacetic acid | Synthetics         | Hydrothermal                          | 4.08      | -                          | 440/513                                      | Hoang et al. 2023          |
| Coconut husk  | Naturals           | Hydrothermal                          | 3.3       | _                          | 350/440                                      | Chunduri et al. 2016       |
| Corn stalk powder   | Naturals           | Microwave                             | 1.5-6.0   | 2.1                        | 326/445                                      | Du et al. 2023             |
| 2,3-Diaminobenzoic<br>acid hydrochloride,<br>sulfuric acid    | Synthetics         | Hydrothermal                          | 7.6       | 18.0                       | 595/644                                      | Liu et al. 2019            |
| Europium chloride, tan-<br>nic acid                           | Synthetics         | Hydrothermal                          | 7.0       | -                          | 307/340                                      | Albalawi et al. 2023       |
| Glucose, polyethyl-<br>eneimine, phosphoric<br>acid           | Synthetics         | Low Temperature                       | 12.0      | 14.0                       | 430/530                                      | Zhao et al. 2021a          |
| Glucose   | Synthetics         | Hydrothermal                          | 5.9-11.0  | -                          | 350/450                                      | Ezati et al. 2022          |
| Glycine, ethylenedi-<br>aminetetraacetic acid,<br>sodium salt | Synthetics         | Hydrothermal                          | 7.2       | 28.3                       | 360/428                                      | Ali et al. 2021            |
| Glycine   | Synthetics         | Hydrothermal                          | 3.3       | -                          | 390/470                                      | He et al. 2023             |
| Glycine, sugars   | Synthetics         | Low temperature, long time            | 2.5       | -                          | 360/438                                      | Nguyen et al. 2024         |
| Lemon peel  | Naturals           | Hydrothermal                          | 2.0       | 32.0                       | 378/468                                      | Vadia et al. 2023          |
| Locust powder, nitric<br>acid, diethylenetri-<br>amine        | Naturals           | Chemical Oxidation                    | 2.3       | 3.1                        | 390/470                                      | Su et al. 2022             |
| Litchi chinensis  | Naturals           | Hydrothermal                          | 4.1       | 12.0                       | -  | Tang et al. 2018           |
| Murraya koenigii  | Naturals           | Hydrothermal                          | 2.0 - 8.0 | 5.4                        | 390/450                                      | Pandey et al. 2020         |
| Mushroom  | Naturals           | Hydrothermal                          | 8.0       | -                          | -  | Boobalan et al. 2020       |
| Mulberry leaves   | Naturals           | Hydrothermal                          | 7.0       | -                          | 320/440                                      | Yang et al. 2023           |
| Phenylenediamine  | Synthetics         | Solvothermal                          | -         | 26.7                       | -  | Zhang et al. 2020          |
|   | Synthetics         | Hydrothermal                          | 10.0      | -                          | 365/470                                      | Cao and Guo 2024           |
| Polyethyleneimine imi-<br>nodiacetic acid,                    | Synthetics         | Hydrothermal                          | 6.7       | 15.9                       | 360/458                                      | Qu et al. 2020             |
| Phenylboronic acid  | Synthetics         | Hydrothermal                          | 3.3       | 12.0                       | 247/323                                      | Jia et al. 2019            |
| Pork rib bones  | Naturals           | Hydrothermal, Chemi-<br>cal Oxidation | 4.2       | _                          | 315/453                                      | Liu et al. 2020            |

#### Table 7 (continued)

| Precursors                             | Type of precursors | Synthetic method             | Size (nm) | Quan-<br>tumn<br>yield (%) | Excitation/ Emis-<br>sion wavelength<br>(nm) | References               |
|--|--------------------|------------------------------|-----------|----------------------------|--|--------------------------|
| Saccharomycetes, ethyl-<br>enediamine; | Synthetics         | Hydrothermal, Micro-<br>wave | _         | 16.0                       | 380/460                                      | Yu et al. 2019           |
| Sodium citrate                         | Synthetics         | Hydrothermal                 | -         | 9.3                        | 380/450                                      | De Paula et al. 2019     |
| Sodium citrate, thioure                | Synthetics         | Hydrothermal                 | 2.5       | 26.9                       | 350/440                                      | Han et al. 2019          |
| Sugarcane molasses                     | Naturals           | Heat treatment               | 1.9       | 5.8                        | 305/390                                      | Huang et al. 2017        |
| Sucrose                                | Synthetics         | Microwave                    | 5.3       | -                          | 365/520                                      | Hu et al. 2019           |
| Tryptophan, ethylenedi-<br>amine       | Synthetics         | Chemical Oxidation           | 6.2       | 48.0                       | 350/400                                      | Mintz et al. 2019        |
| Tryptophan, glucose                    | Synthetics         | Hydrothermal                 | 4.0-5.0   | 18.0                       | 300/450                                      | Ma et al. 2020           |
| Unripe peach                           | Naturals           | Hydrothermal                 | 8.0       | 15.0                       | 325/404                                      | Atchudan et al. 2016     |
| Water hyacinth leaves                  | Naturals           | Chemical Oxidation           | 3.7       | 27.0                       | 400/420-700                                  | Prathumsuwan et al. 2019 |
| Wheat brain                            | Naturals           | Hydrothermal                 | -         | 33.2                       | 400/500                                      | John et al. 2020         |

Numerous precursors are used to synthesize carbon dots, categorized into two main groups: natural and synthetic materials. The size, quantum yield, and fluorescent properties of the synthesized carbon dots vary depending on the synthesis method employed, such as hydrothermal, micro-wave, chemical oxidation, or heat treatment

Furthermore, the choice of synthesis method can also influence the surface chemistry and functionalization of carbon dots. For example, using surface passivation agents or functionalization during the synthesis process can modify the surface properties of carbon dots, such as their stability, dispersibility, and biocompatibility (Atabaev 2018). This can be particularly important for applications in bioimaging or sensing, where the interaction of carbon dots with biological systems is crucial. Moreover, the synthesis method can also impact the optical properties of carbon dots. Different synthesis methods can lead to carbon dots with distinct photoluminescence properties, such as emission wavelength, quantum yield, and stability (Wang et al. 2014). By selecting the appropriate synthesis method, it is possible to tailor the optical properties of carbon dots for specific applications, such as fluorescence imaging or optoelectronics. Additionally, the choice of synthesis method can also affect the scalability and reproducibility of carbon dot production. Some synthesis methods may be more suitable for large-scale production, while others may be more suitable for smallscale or laboratory-scale synthesis (De and Karak 2013). Considering the scalability and reproducibility of the synthesis method is essential to ensure the consistent and reliable production of carbon dots.

## Surface modification and functionalization

Surface modification and functionalization of carbon dots are important strategies to enhance their properties and reduce risks in various applications (Anilkumar et al. 2011; Ding et al. 2014; Park et al. 2016). By introducing functional groups to the surface carbon dots, their stability, biocompatibility, dispersibility, and functionality can be enhanced (Anilkumar et al. 2011; Ding et al. 2014). Functional groups such as amino, carboxyl, hydroxyl, or thiol groups can be added to enhance the solubility, stability, and reactivity of carbon dots (Park et al. 2016). These functional groups can also serve as anchor points for further functionalization or conjugation to biomolecules, dyes, or targeting ligands (Liu et al. 2015). In addition, these surface modification can even lower the detection limit of mercury (II) using nitrogen-carbon dots-based sensors (Aziz et al. 2019). Another approach to surface modification is doping carbon dots with heteroatoms like nitrogen, sulfur, or phosphorus. Doping can alter the electronic structure and surface chemistry of carbon dots, leading to improved photoluminescence, charge transfer, and catalytic activity (Dong et al. 2013; Hu et al. 2016). Nitrogen-doped carbon dots, for example, have shown enhanced fluorescence and improved biocompatibility, making them suitable for bioimaging and sensing applications (Wang et al. 2014). Furthermore, surface modification of carbon dots using appropriate precursors can improve their biocompatibility and reduce cytotoxicity (Yao et al. 2019). For example, Min et al. (2023) synthesized carbon dots from coffee grounds and seed extracts can be used to make active packaging to increase the shelf life of ground pork and prevent the growth of microorganisms.

In addition to functional groups and heteroatom doping, surface modification can involve the encapsulation or coating of carbon dots with polymers, silica, or other materials. This provides additional protection, stability, and functionality to carbon dots (Sawant et al. 2016). For instance, encapsulating carbon dots with polymers can enhance their biocompatibility and enable the controlled release of encapsulated drugs or biomolecule carbon dots (Sawant et al. 2016). Surface modification and functionalization of carbon dots can be tailored to specific applications. In bioimaging, surface modification by targeting ligands or biomolecules enables specific cellular or tissue targeting (Liu et al. 2015). In sensing applications, surface modification enhances the selectivity and sensitivity of carbon dots toward specific analytes (Shen and Xia 2014). Furthermore, surface modification can improve the performance of carbon dots in energy storage, catalysis, and optoelectronic devices (Balogun et al. 2016).

# Development of specific protocols to quantify carbon dots

Developing a quantitative method for measuring the amount of carbon dots is an important strategy for mitigating risks and ensuring accurate dosing in various applications. Several studies have focused on developing such methods to provide reliable and precise quantification of carbon dots. One approach to quantifying carbon dots involves spectroscopic techniques. Fluorescence spectroscopy is commonly used to measure the emission intensity of carbon dots, which can be correlated with their concentration (Li et al. 2012; Nguyen et al. 2024; Pan et al. 2020). By establishing a calibration curve using known concentrations of carbon dots, the emission intensity can be used to determine the amount of carbon dots in a sample. This method allows for rapid and non-destructive quantification of carbon dots. Another method for quantifying carbon dots is based on their absorbance properties. UV-Vis spectroscopy can be used to measure the absorbance of carbon dots at specific wavelengths, which can be correlated with their concentration (Nie et al. 2014). The amount of carbon dots can be determined by comparing the absorbance of a sample to a calibration curve (Churchill et al. 2009). This method is relatively simple and widely applicable.

In addition to spectroscopic techniques, other methods have been explored for quantifying carbon dots. For example, electrochemical methods have been developed, where the current or potential response of carbon dots is measured and correlated with their concentration (Zhao et al. 2022). This approach offers the advantage of high sensitivity and selectivity. Furthermore, advanced techniques such as atomic force microscopy (AFM), scanning electron microscopy (SEM), transmittance electron microscopy (TEM), and Raman microscope can be used to visualize and quantify carbon dots based on their morphology and size (Saraswat and Yadav 2020). These imaging techniques provide valuable information about the size distribution and density of carbon dots in a sample.

### Perspective

Moving forward, a concerted effort is needed to establish comprehensive safety guidelines and regulatory frameworks for incorporating carbon dots in food applications. Furthermore, research is needed to elucidate the biodistribution, degradation pathways, and long-term health impacts of carbon dots. By pursuing these endeavors, we can ensure the responsible and beneficial integration of carbon dots in various food technologies while safeguarding consumer health and environmental sustainability. Exciting applications lie ahead, including intelligent food packaging with quality monitoring, enhanced food preservation by precisely targeting and eliminating foodborne pathogens, and delivery systems for essential nutrients. Carbon dots could be the key to creating intelligent food colorants and pathogen sensors. By pursuing these research directions and exploring these applications, we can unlock the full potential of carbon dots for a safer, healthier, and more sustainable food future.

## Conclusion

Carbon dots have emerged as a noteworthy nanomaterial with various applications in various industries, including electronics, bioimaging, and pharmaceutical analysis. Recently, significant attention has been directed toward exploring their potential in the food technology sector. However, the absence of specific guidelines for determining toxicity thresholds and permissible levels of carbon dots in food remains a challenge. In this review, we have evaluated the potential risks associated with carbon dots and proposed mitigation strategies to address these concerns. These strategies encompass (1) selecting safe precursor materials, (2) adopting appropriate synthesis methods, and (3) modifying the surface agents to regulate surface properties, size, and characteristics of carbon dots. (4) Furthermore, it is imperative to develop standardized procedures for quantifying carbon dot levels in food products and to establish guidelines for research on the decomposition and potential contaminants introduced by carbon dots in food matrices.

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### **Declarations**

**Conflict of interest** The authors have no relevant financial or nonfinancial interests to disclose. The authors have no other competing interests to declare.

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