Carbon Quantum Dots: A Potential Candidate for Diagnostic and Therapeutic Application

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S. Sharath Shankar, Vishnu Ramachandran, Rabina P. Raj, T. V. Sruthi, and V. B. Sameer Kumar

Abstract

Among various quantum dots, carbon quantum dots (CQDs) are getting much more attention due to their nontoxicity and high water solubility. CQDs with or without functionalization/passivation possess different fluorescent properties. Moreover, simple and economical procedure involved in their preparation makes them a reliable substitute for nano-materials/semiconductor quantum dots. Due to their tunable fluorescent properties, these carbon quantum dots find application in anticancer studies, tissue engineering, nano-medicine and targeted drug delivery, electrochemical sensing, bio-sensing, and bio-imaging. Also, these CQDs are capable of showing high conductivity and therefore can be useful for improving the electrical/conductive nature of different materials. The CQDs can find significant role in LEDs. CQDS can be prepared by top-down approaches or bottom-down approaches. Top-down approaches involve arc discharge method, laser ablation, electrochemical oxidation, and chemical oxidation, and the bottom-down approaches include combustion, pyrolysis, microwave-assisted method, ultrasonic method, oxidative acid treatment, and hydrothermal methods. In this chapter we have tried to give an insight to the preparation and biological applications of CQDs.

Keywords

Carbon quantum dots · Fluorescent nano particles · Applications of Carbon quantum dots · Electrochemical/ bio-sensing · Bio-imaging

S. S. Shankar · V. Ramachandran · R. P. Raj · T. V. Sruthi · V. B. S. Kumar (✉)
Department of Biochemistry & Molecular Biology, Central University of Kerala, Kasaragod, Kerala, India

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3.1 Introduction

Carbon quantum dots (CQDs) are a new class of fluorescent carbon nano-materials of less than 10 nm size. CQDs are also referred to as carbon nano-dots (CNDs) and carbon dots (CDs/C-dots), which can be defined as having a carbogenic nucleus with functional surface groups. Discovery of CQDs was serendipity, when Xu and co-workers in 2004 saw some residual fluorescent nanoparticles during the purification process of single-wall carbon nanotubes (SWNTs) (Xu et al. 2004). It was Sun and team who named these particles as carbon quantum dots (Sun et al. 2006). The chemical character of CQDs varies from amorphous to nanocrystalline and has predominant sp^2 carbon, the spacing of which consists of graphitic or turbo-static carbon (Baker and Baker 2010; Zheng et al. 2011). CQDs are typically composed of 53.93% carbon, 2.56% hydrogen, 1.30% nitrogen, and 40.33% oxygen. The intriguing features of carbon dots are their chemical inertness, low photo-bleaching and photodegradation, low cytotoxicity, and excellent biocompatibility. Due to the existence of oxygen moieties, the dots have proven to be an extremely soluble material in aqueous solution (Baker and Baker 2010; Zheng et al. 2011). Functional groups such as –OH, -COOH, and -NH_2 and other CQD surface groups make them more water soluble (Cao et al. 2007). For these reasons, CQDs had drawn broad attention for various ranges of applications in technology, engineering, and particularly biomedical fields.

One of the most interesting characteristics of CQDs is the size-dependent optical absorption. Carbon dots have strong absorption in both ultraviolet and visible region (Li et al. 2010a). CQD's luminescence characteristics include electroluminescence and photoluminescence (PL), the most prominent of which is the latter. Carbon quantum dots have outstanding optical properties such as elevated fluorescence stability, non-blinking, adjustable excitation, and wavelengths of emission (Li et al. 2010a; Jia et al. 2012; Wang et al. 2013a, 2014). The precise understanding behind the emitting mechanism of CQDs is uncertain, but scientists speculated it involved quantum containment impact, surface trap stabilization, or exciton recombination radiation (Li et al. 2012). The presence of surface energy traps is stated to result in surface passivation emissions ascribed to CQD's photoluminescence (PL) (Sun et al. 2006). Moreover the photo-excited CQDs act as efficient electron donors and acceptors; therefore, electron acceptor or electron donor molecules in alternatives can be readily quenched. The CQDs also act as an appropriate candidate for light energy conversion, photovoltaic devices, and associated applications owing to their capacity to display photo-induced electron transfer property (Shen et al. 2012). CQDs with their simplistic methods of synthesis displayed a plethora of immense implications in the field of chemistry, engineering, biology, as well as medicine. Various precursors such as chemical, green, and waste products can be used to synthesize CQDs. Carbon dot synthesis is widely divided into two classifications, top-down and bottom-up. Top-down technique usually includes splitting bigger carbonaceous materials (Xu et al. 2004), whereas bottom-up synthesis process includes supported path and carbonization where a smaller precursor molecule forms nano-carbon dots through a series of chemical reaction (Baker and Baker

2010). Supported route is referred as blocking of nanoparticle agglomeration during high-temperature treatment (Wang et al. 2015a). Both approaches of synthesis require particle size control, proper measures against agglomeration during carbonization, and surface passivation. Method of arc discharge, chemical oxidation, laser ablation, electrochemical oxidation, etc. are included under top-down approaches. Generally dots synthesized by means of these methods lack fluorescence; an additional passivation is necessary to produce light emission. Bottom-up approaches include combustion, pyrolysis, oxidative acid treatment, microwave, ultrasonic, and hydrothermal methods, which serve more attraction and attention in recent decades due to their simplest preparation routes and inexpensive carbon sources (Li et al. 2012). Nowadays green synthesis is gaining more for the CQD synthesis. In the present chapter, we mainly focus on various methods of synthesis of CQDs and discuss their potential applications in different fields of therapeutics.

3.2 Synthesis

During the synthesis of single-walled carbon nanotubes by arc discharge technique (Xu et al. 2004), Xu et al. found carbon nano-dots in 2004. Laser ablation and arc discharge are altered technique of physical vapor deposition (PVD) involving the condensation of the strong carbon atom formed by warm gaseous carbon atom. Laser ablation is a simplistic, environmentally friendly, and effective technique for producing CQDs in which the surface conditions of particles can be adjusted. In this process, the carbon material is irradiated under elevated pressure and temperature with a laser beam in the presence of an inert gas as a carrier. Usually, carbon dots are prepared by dispersing the carbon precursor in a solvent by ultra-sonication and dropping the resultant suspension onto a glass cell; later those glass cells are treated as carbon target for laser irradiation/ablation. The surface of CQDs may be changed by choosing appropriate organic solvents to achieve tunable light emission. After laser irradiation, for the development of fluorescent carbon dots, the reaction mixture is subjected to centrifugation, purification, and surface passivation. Li et al. declared an easy laser ablation method to produce CQDs employing a common solvent as the liquid medium (such as acetone, ethanol, or water) and nano-carbon materials (less than 50 nm) as the carbon precursor (Thongpool et al. 2012; Qu et al. 2013). By adding nitryl with arc discharging fuming nitric acid, Xu et al. enhanced the hydrophilicity of the synthesized dots. The electrophoretic separation eventually resulted in three groups of fluorescent nanoparticles emitting blue-green, yellow, and orange at distinct molecular weights. Hu et al. in 2011 merged the multistep synthesis process into one-step reaction for simplifying the reaction as well as obtaining CQDs with better fluorescence. They combined laser ablation and passivation using polyethylene solution under laser radiation for 2 h and finally obtained fluorescent CQDs of size 3.2 nm with a quantum yield of 12.2%. Furthermore they developed CQDs of varied fluorescence by choosing different organic solutions (Hu et al. 2011).

Electrochemical synthesis is one of the CQD synthesis approaches by applying direct current from comparatively big carbon products such as graphene, graphite, carbon fiber, etc. The significant factors for generating CQDs through the electrochemical oxidation method are hydroxyl groups and an alkaline atmosphere. In electrochemical synthesis, three electrode systems are imparted; two Pt sheets were used as operating and counter-electrodes, and one calomel electrode installed on the lugging capillary was used as reference electrodes. In such a manner that the distance between the two Pt sheets is about 3 cm, the electrodes are fixed with a rubber plug. The precursor solution is to mix alcohol and water in a fundamental medium, and the length of the response is until the transparent solution becomes black. Later, equal ethanol volume is added to salting out sodium hydroxide (added for alkalinity). The mixture is kept overnight for evaporation of solvent, and a solid product is formed which after separation and purification yields carbon quantum dots. Zhao and Xie effectively synthesized CQDs using electrochemical method employing a 3.0 V electro-oxidized graphite column electrode (GE) with a Pt counter electrode in 0.1 M potassium dihydrogen phosphate aqueous solution as the electrolyte supporting solution (Zhao and Xie 2017). Subsequently, the resulting oxidant solution needs ultra-sonication, ultra-filtration through a 22.0 mm filter membrane, washing, and drying. Kang and his colleagues revealed another electrochemical operation for producing CQDs of 1–4 nm using alkali-assisted electrochemical techniques (Li et al. 2010a). Another simple strategy of producing high-quality CQDs is the use of small graphite pieces by accurate cutting into ultrafine particles of a graphite honeycomb layer. In 2016, Canaveri and his colleagues discovered that the CQD characteristics depended considerably on the moment spent in the method of electrolysis. The benefit of this synthesis is that by tuning the intensity of the applied present, size-dependent photoluminescent characteristics can be accomplished. Zhou et al. developed 2.8 nm large fluorescent carbon dots from multi-walled carbon nanotubes as the working electrode and later cultivated in carbon diaphragm using chemical vapor deposition (CVD) method and variable cycle voltage (Zhou et al. 2007). In another study, Hou et al. acquired water-soluble functionalized fluorescent CQDs with two Pt sheets ($1.5 \times 2\ cm^{-2}$) as positive and negative electrodes through electrochemical carbonization of sodium citrate and urea (Hou et al. 2015). Despite of all the advantages, major disadvantages in electrochemical synthesis are the tedious purification procedures of the synthesized carbon nanoparticle and lesser production yield.

Other strategies in carbon quantity dots synthesis involve (Fig. 3.1) hydrothermal method (Wang et al. 2013b; Xu et al. 2014; Dong et al. 2014; Gao et al. 2013; Shen et al. 2018), microwave-assisted pyrolysis method (Tang et al. 2014; Zhu et al. 2009; Wang et al. 2012; Rodríguez-Padrón et al. 2018; Yang et al. 2012), ultrasonic method (Oza et al. 2015), and acid dehydration method (LeCroy et al. 2014), and among those, the former two are the most widely used ones. Hydrothermal method of CQD preparation was first introduced by Liu and co-workers in 2007. It is the most common and easiest technique for the synthesis of carbon dots using both chemical and green precursors. Typically, in the presence of either air or organic solvent, the precursor is carbonized in an autoclave at elevated temperature. The blend is permitted to cool at room temperature after full carbonization, and products

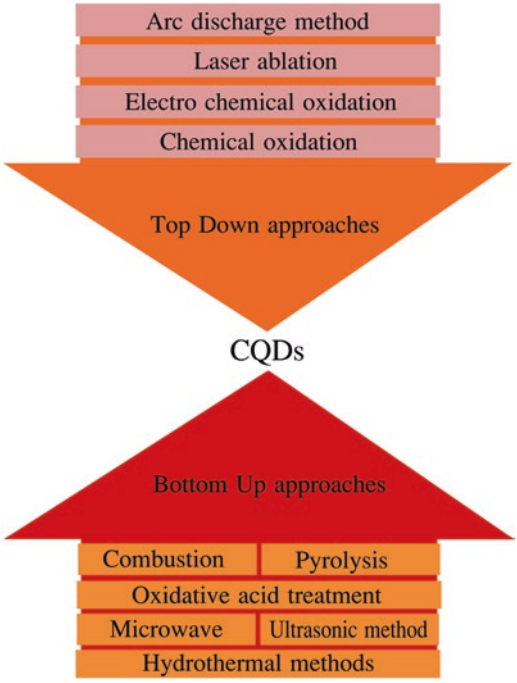

Fig. 3.1 Schematic representation of various methods used in CQD synthesis

can be extracted with an organic solvent. They chose candle ash as a carbon source and blended it with nitric acid via heat reflex and produced a black homogeneous solution and purified it further through a sequence of centrifugation, dialysis, and gel electrophoresis to achieve CQDs of distinct particle sizes. They discovered it even under the same excitation wavelength (λex, 315 nm); particles of different sizes showed different emission wavelengths in its fluorescence which signifies the better optical character of florescent CQDs (called elementary excitation and multi-emission). Many CQDs were subsequently synthesized using hydrothermal methods and using various carbon sources that considerably improved the quantity output of CQDs. Depending upon the precursor, hydrophilicity and hydrophobicity of the synthesized florescent CQDs can be manipulated. Normally surface passivation is not required; however, there is limited control over the particle sizes. In 2013, Gao et al. used C^{60} as a carbon source and CTAB as a passivator to prepare elevated fluorescence CQDs with a quantum yield of up to 60%.

Microwave method of CQD synthesis was introduced in 2009 by Zhu and co-workers, to synthesize novel CQDs showing that fluorescence depends up on the reaction time. Sugar moieties are generally used as a source of carbon and polymeric oligomers as a response medium. In their experiment, Zhu et al. chose carbohydrates as a source of carbon and PEG-200 as a solvent and coating agent, and the response took 2–10 min under 500 W microwave power radiation (Zhu et al. 2009). The colorless solution transforms into pale yellow and then dark brown in a couple of minutes, depicting the formation of CQDs. Wang and co-workers synthesized CQDs from eggshells by initially burning eggshell into ashes and mixing it with sodium

hydroxide solution and treating in microwave radiation for 5 min (Wang et al. 2012). Chandra et al. synthesized CQDs of shining bright green fluorescence in 3 min 40 s from sucrose using microwave radiation in phosphoric acid environment (Chandra et al. 2011). These low-cost economic syntheses of carbon dots are now widely used for various industrial and biological applications. The carbon dot synthesis of oxidative acid therapy utilizes waste soot as a carbon precursor. Waste soot is a mass of carbon particles from incomplete hydrocarbon combustion. The therapy includes reflux ionization in acidic medium of waste soot, followed by centrifugation, neutralization, and purification. Typically, burned waste soot was mixed with nitric acid and refluxed for 12 h. The carbon particles are collected by centrifugation after room temperature is reached. The collected mixture undergoes neutralization with sodium carbonate and extensive dialysis against water for obtaining pure carbon dots. The advantages of these acid treatments are efficient introduction of functional groups like carbonyl, carboxyl, amines, and epoxy to the synthesized CQDs which bestow them requisite properties for imaging application. However, the storage stability of thus synthesized carbon dots is minimal.

To remove amorphous carbon, catalyst, and other impurities introduced during the synthesis, it is essential to purify carbon nano-material for biological implementation. The typical purification processes used after synthesis are plasma oxidation, high-temperature annealing, and some chemical treatments. Surface passivation and functionalization are two critical steps for selective implementation in bio-imaging, drug delivery, etc. in post synthesis of carbon dots. Surface passivation decreases the damaging impact of surface contamination on their optical properties and gives them a high level of fluorescence. It is also a significant step in achieving PL property for CQDs with a size of 1.5–2 nm (Dimos 2016). It is typically performed by covering the carbon quantum dots surface with a thin layer of oligomers (PEG), thionyl chloride, thiols, and spiropyrans. Both the acid therapy and the passivation of the surface enhance the quantum yield of synthesized carbon dots as well as enhanced florescent emissions. Similarly, surface functionalization is very important for the synthetic carbon dots' florescent behavior. Wet chemical treatment involves acid reflux, and dry treatments such as RF-plasma treatment and treatment with strong acids such as H_2SO_4, HNO_3, $KMnO_4$, OsO_4, $K_2Cr_2O_7$, CCl_4, CF_4, $O_2(g)$, and SF_6 are employed. It introduces functional groups like carbonyl, ketone, hydroxyl, ester, alcohol, fluorine, thiol, amines, and carboxyl which can function as surface energy traps and lead to fluorescence emission behavior variations in CQDs. Numerous studies to functionalize and modify the surface of CQDs such as covalent bonding have been carried out to date (Dong et al. 2012; Yin et al. 2013) along with p–p interactions (Li et al. 2011), sol–gel (Mao et al. 2012; Wang et al. 2011), coordination (Zhao et al. 2011), which renders them solubility, biocompatibility and low toxicity. CQDs have big quantities of oxygen-containing groups that enable them to bind covalently to other functional groups. The latest strategy for surface alteration of CQDs is covalent bonding with chemical agents comprising amine groups to recover the photo luminescence of CQDs, which has been shown to be highly efficient in changing the characteristics of CQDs (Liao et al. 2013). Li et al. studied that the PL of prepared CQDs originated from carboxylate groups produced on the particle surface (Li et al. 2010b). They stated

that on the surface of the preliminary carbon precursors generated by laser irradiation, several oxygen-containing radicals could potentially be the cause of PL. The primary features of these functional CQDs are high stability and outstanding photo-reversibility. Another attractive advantage for creating functional group is that it can load smaller therapeutic molecule by covalent conjugation or non-covalent adsorption onto the CQDs. Doping with heteroatoms, sulfur, nitrogen, and magnesium ion, other than surface passivation and functionalization, can increase the quantity yield up to 83%. The most preferential use of nitrogen as a dopant is that it can donate its electron to carbon dots so that the change in electronic configuration contributes in increased quantum yield. It can either be accomplished by selecting precursor-containing nitrogen or by post-functionalization.

3.3 Applications

Since the discovery of CQDs and their fluorescent properties and tenable surface-related properties, research has been done with the material for its implementation in different fields of applications (Fig. 3.2). All the current fields of applications of CQDs are discussed below.

3.3.1 Bio-imaging

Live cell bio-imaging is an imperative tool for elucidating the dynamics of biological mechanism. Maintaining the cellular ambience is very critical in live cell imaging, as the environmental factors can duly impose abiotic stresses. When light

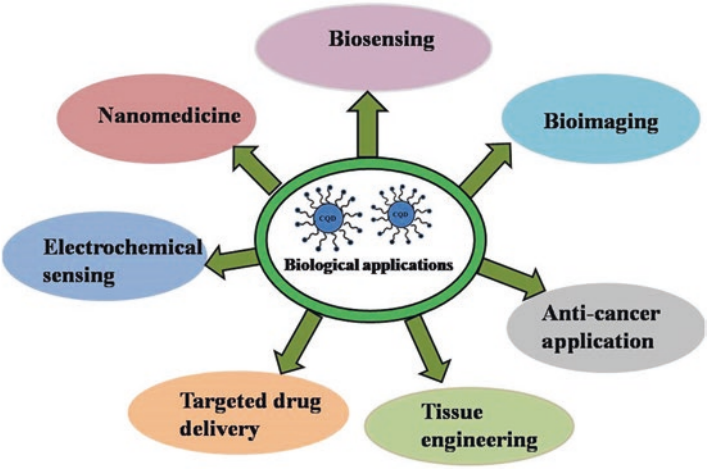

Fig. 3.2 Biological implementation of carbon quantum dots; CQDs display adjustable physical and chemical properties and could therefore be used in different biological areas such as bio-sensing, bio-imaging, etc.

radiation interacts with the cell, temperature rises owing to the excitation of light active molecules that can boost free radical formation. It is therefore essential to require minimum energy or a source of low intensity light radiation to decrease oxidative stress. Traditionally, fluorescent probes were used in bio-imaging studies, to understand cellular processes (Luo et al. 2013). Rhodamine 6G is one of the best commercially available organic fluorophores with quantum yield up to 80%; however, the major drawbacks of such probes are photo-bleaching, less photostability, narrow excitation, and emission wavelength and cytotoxicity. One of the remarkable properties of doped CQDs that distinguish them from conventional organic dyes is their resistance from photo-bleaching and photodegradation. The unique fluorescent properties, excitation-dependent multicolor emission, high photostability, high aqueous stability, superior cell permeability, surface modification capability, and good biocompatibility have made carbon dots a versatile material for bio-imaging and sensing. Unlike semiconductor nanocrystal-like quantum dots, CQDs are non-toxic in nature. Semiconductor quantum dots have been advantageous over florescent samples but are less biocompatible because most QDs have toxic heavy metals such as Cd, Pb, and Hg in their structure. Even if they have ground coverage, the likelihood that toxic metals will be released into the medium is not negligible (Jaishankar et al. 2014). CQDs could address effectively all of the aforementioned complications. Internalization of carbon dots by the cell is a temperature-dependent process, and it takes place at ambient temperature of 37 °C. They translocate into the cell via endocytosis; hence, coupling with membrane-translocating peptides can enhance the process. CQDs are used as fluorescent markers to test multiple cellular organs such as the endoplasmic reticulum, Golgi body, nucleolus, mitochondria, lysosome/endosome, etc. (Fig. 3.3), and CQDs can be efficiently removed from the body via excretion, within an hour of their intravenous injection (Longmire et al. 2008).

Carbon dots are very specific in their visible lengths of excitation and emission waves, and they also have elevated brightness at individual dot concentrations. Yang et al. first discovered the applicability of using CQDs in bio-imaging through the deployment of CQDs as a FL contrast agent in 2009. Several studies revealed the use of different CQDs in cellular fluorescence imaging using different cell lines, including Caco-2 (Dias et al. 2019), Ehrlich ascites carcinoma (Ray et al. 2009), HEK293 (Luo et al. 2013), pig kidney (Ray et al. 2009), B16F11 (Luo et al. 2013), murine P19 (Liu et al. 2009), BGC823 (Wang et al. 2011), and HeLa (Xu et al. 2015). Yang et al. used pegylated CQDs with adequate contrast for in vivo optical imaging (440 μg in 200 μL) by subcutaneously injecting CQDs into mice (Yang et al. 2009). Tao et al. used the same protocol and then collected images from 455 nm to 704 nm at distinct wavelengths after subcutaneous injection of aqueous CQD solution (Tao et al. 2012). In 2012, Hahn et al. developed diamine-capped pegylated CQDs to label B16F1 and HEK293 (Tao et al. 2012). In addition, real-time bio-imaging was produced possible by Hang's and colleagues' job, where they reported that CQDs could be used for real-time bio-imaging by delivering hyaluronic acid (HA) derivatives specifically for the purpose (Goh et al. 2012). HA/CQD hybrids are synthesized through amidation reaction between amino

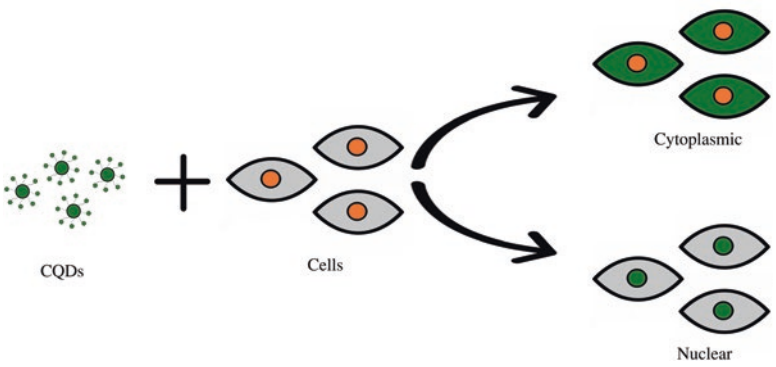

Fig. 3.3 CQDs' ability as a fluorophore in bio-imaging: The CQDs exhibit fluorescence under UV lights. Due to their smaller size, they can easily enter into the cells. Based on its functionality, it may locate into the nucleus or in the cytoplasm. Hence, the nucleus or cytoplasm can be easily visualized through the fluorescent microscope

groups on the surface of CQDs and carboxylic groups of HA. The in vivo imaging results revealed that these hybrids could employ target-specific delivery in the liver (Goh et al. 2012).

A combo of optical imaging (OI) and magnetic resonance imaging (MRI) is the latest appealing technology in bio-imaging using CQDs, where optical imaging provides quick screening, while MRI provides physiological and anatomical data and elevated spatial resolution (Lee et al. 2012). Because of its small cytotoxicity compared to commercial Gadovist VR, Gd(III)-doped CQDs could be used in biomedical research for multimodal imaging. In 2012, Bourlinos AB et al. prepared ultrafine water-dispersible Gd(III)-doped CQDs with dual MRI/FL personality through heat decomposition (Bourlinos et al. 2012) and thus acquired particles showing bright FL in the visible region and showing powerful T1-weighted MRI (Bourlinos et al. 2012). In the presence of small Fe_3O_4 nanoparticles for MR/FL multi-imaging, a 6 nm iron oxide double CQD was recently synthesized by pyrolysis of organic molecules (Srivastava et al. 2012). The CQDs produced by hydrothermal chitosan carbonization with amino acids were used to evaluate the cyto-compatibility of human lung adenocarcinoma cells (A549) (Yang et al. 2012). Zhang and Huang et al. revealed a tiny molecular fluorescent carbogenic complex that could selectively stain the nucleolus rich in RNA (He et al. 2016). Recently it has been revealed that CQDs generated by the hydrothermal one-pot response of m-phenylenediamine and L-cysteine can efficiently target the nucleolus. It offers high-quality nucleolus imagery in living cells, enabling nucleolus-related biological behavior to be tracked in real time, as opposed to SYTO RNASelect, a widely used commercial nucleolus imagery in fixed cells (Hua et al. 2018). These multifunctional CQDs prevent lysosomal/endosomal trapping and target nucleolus selectively and bring protoporphyrin IX (PpIX, a commonly used photosensitizer) into the nucleus effectively (Hua et al. 2018).

By its excitation-dependent fluorescence emission, numerous studies revealed the cell imaging ability of doped CQDs. CQDs are shown as multicolor nano-samples

due to excitation-dependent fluorescence emission, which can be excited with distinct excitation wavelengths. For example, on 543, 488 and 405 nm excitation, Zhai et al. (2012) reported that C-dots incubated with L929 cells could emit red, green, and blue fluorescence (Zhai et al. 2012). The in vivo fluorescence image of CQDs injected into the nude mouse with different agitation wavelengths shows a better contrast between the signal and background signal over 595 nm long, which is also efficient for in vivo imaging (Yang et al. 2009; Tao et al. 2012) (Timur 2018).

3.3.2 Bio-sensing

Biosensors are the type of analytical device which converts biological signals/response to electrical signals (Shankar et al. 2012, 2013). There are various types of biosensors present at the moment, like DNA sensor, enzyme sensors, protein sensors, etc. Nano-biosensors are considered to be the most important of different biosensors because they are capable of detecting biochemical signals and/or biophysical signals related to a specific disease at the level of one or a few molecules (Chandra et al. 2017). Nano-biosensors were effectively used to detect several biomolecules in vitro as well as in vivo. This technology is anticipated to revolutionize point-of-care and personalized diagnostics and be highly useful for early identification of disease (Chandra and Segal 2016). CQDs are capable of serving as either great electron donors or electron acceptors, making them suitable for monitoring/sensing different materials and parameters such as cell iron, copper, pH, proteins, enzymes, vitamins, and nucleic acid (da Silva and Gonçalves 2011). The mechanism behind the detection of ions is through the surface functional groups on CQDs, which show distinctive affinities to different target ions, resulting in an electron or energy transfer process quenching of PL intensity (Fig. 3.4). When the interaction is broken by external force, the PL is restored. Qu et al. established CQDs in 2013 using dopamine as the raw material that could be used as a Fe^{3+} detector with a stronger 0.32 μM detection threshold (Qu et al. 2013). For example, Xu et al. created a CQD-based ultrasensitive sensor in 2015 through a one-step hydrothermal therapy of potatoes for phosphate detection (Xu et al. 2015). Recently, Yang et al. built a fluorescent turn-on scheme in which a novel oligodeoxyribonucleotide (ODN)-CDs and graphene manufactured the optical sensor to identify Hg^{2+}. The method behind the sensing was FRET, graphene oxide could quench the fluorescence of ODN-CQDs via FRET, and luminescence was later retrieved with the addition of Hg^{2+} by removing ODN-Ds from graphene oxide owing to the creation of T-Hg^2-T duplex (Cui et al. 2015). Tian and colleagues produced aminomethylphenylterpyridine (AE-TPY) CQDs to determine changes in the pH value of physiological circumstances such as tumors. Thus, manufactured PL sensors with great selectivity and sensitivity could be used to monitor pH value gradients in a range of 6.0–8.5 (Kong et al. 2012). They showed the applicability of these sensors in mice's living cells and tumor tissues, which in the near future will require in vitro and in vivo applications. The CQDs have recently been used to identify the importance of intracellular pH in living pathogenic fungal cells (Cui et al. 2015).

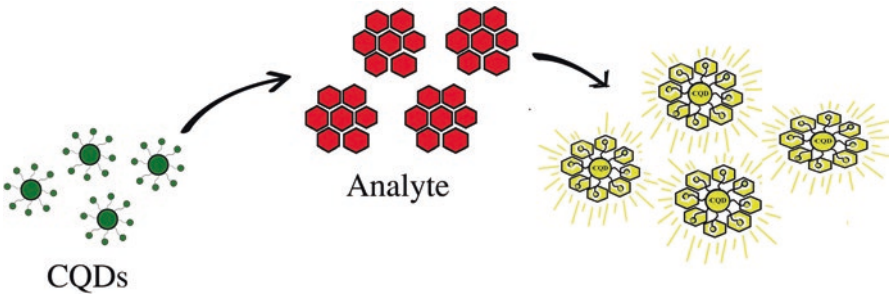

Fig. 3.4 Functional groups present in the functionalized CQDs have the ability to target certain biological molecules; binding to these molecules could result in the quenching of PL of CQDs which forms the basis of bio-sensing application

Bio-sensing of nucleic acid is based on adsorption by CQDs (as a fluorescent quencher) of the fluorescent single-stranded DNA (ssDNA) probe through p–p interactions. When ssDNA is hybridized with its target and double-stranded DNA (dsDNA) is formed, the desorption of the hybridized dsDNA from the surface of the CQDs causes its florescence to recover, resulting in the detection of the target DNA (Li et al. 2011). DNA-labeled CQDs have been created to detect proteins and enzymes; Maiti and colleagues have created CD-dsDNA sensors for histone detection (Maiti et al. 2013). The PL of this sensor would be quenched in the presence of dsDNA, while PL could be retrieved with the addition of histone owing to the powerful connection between histone and dsDNA through unwinding of dsDNA from CQDs. Quantitative protein detection can therefore be calculated from restored fluorescence. C-dot-based sensors widely detect biomolecules such as amino acids and vitamins. Xu et al. and Wang et al. in 2015 detected riboflavin and vitamin B12 using surface functionalized CQDs. Ratio-metric sensing protocol was adopted for detection, and CQD-based ratio metric sensors could detect riboflavin with high sensitivity even in 1.9 nM (Xu et al. 2015; Wang et al. 2015b). Novel nano-sensors composed of CQDs and gold particles were developed to detect cysteine, by Liu and Zang et al. in 2015 (Deng et al. 2015). In addition, Jana et al. in 2015 fabricated a turn-on CQD-based sensors that could detect cysteine with better sensitivity and selectivity (Jana et al. 2015).

3.3.3 Electrochemical Sensors

The biomolecules which are electrochemically active can be easily determined by means of electrochemical techniques such as cyclic voltammetry (Shankar and Swamy 2014), differential pulse voltammetry, etc. (Shankar et al. 2009). In the field of electrochemical sensing, the electrochemical characteristics of CQDs have recently been studied. In the presence of other molecules such as ascorbic acid and serotonin, Shereema and crew launched CQD-based carbon paste electrode for the electrochemical determination of dopamine (Shankar et al. 2015). The electrochemical and capacitive properties of CQDs were reported by another team. They observed

that rGO's capacity was enhanced by adding a suitable quantity of CQDs to the material. They claimed that the CD/rGO electrode in 1 M H_2SO_4 had good reversibility, excellent rate capability, fast charging, and high specific capacity (Dang et al. 2016). He et al. claimed that a composite of CQDs and poly-ortho-aminopyridine was capable of improving the electrochemical sensing properties of glassy carbon electrode. Further, they used this fabricated electrode for simultaneous detection of guanidine and adenine in a mixture (He et al. 2018). Kakaei's team studied the potential CQDs toward the oxidation-reduction reaction of methanol (Kakaei et al. 2016). Identification of metanil yellow and curcumin from a mixture of real sample was made possible with the help of CQD-fabricated electrodes, thus opening an application in food industry, for the detection of commonly employed adulterant (Fig. 3.6) (Shereema et al. 2018). Rene Kizek et al. manufactured CQDs for the electrochemical determination of anticancer drug, i.e., etoposide (ETO), and modified glass carbon electrode (GCE). They also suggested three distinct methods to modify GCE with CQDs. The electrode reported was capable of detecting etoposide with the smallest LOD relative to current detectors (Nguyen et al. 2016). Shunxing Li et al. in 2014 synthesized CQDs/octahedral Cu_2O nanocomposites and coated it on the GCE surface. Surface and electrochemical properties of the developed electrode were carried out and reported that the CQDs/octahedral Cu_2O-based electrode exhibits low electron transfer resistance. They also launched the electrode to detect glucose and hydrogen peroxide electrochemically (Li et al. 2015). Houcem Maaoui and his colleagues built and reported similar types of CQDs/Cu_2O/GCE in 2016 and used the same to determine glucose in patient serum (Maaoui et al. 2016). The Liu Junkang group reported a detailed investigation into the modification strategy of glassy carbon electrode with a hybrid material consisting of graphene aerogel and octadecylamine-functionalized carbon dots and their application in the electrochemical determination of acetaminophen (Ruiyi et al. 2018). Sundramoorthy and his team recently revealed electrochemically exfoliated carbon quantum electrodes modified in 2018 (Devi et al. 2018). In their method, electrochemical exfoliation method synthesized CQD using graphite rods in alcoholic NaOH was cast on a GCE or screen-printed carbon electrode (SPCE) for electrode manufacture. Electrochemical studies with these electrodes disclosed that the reported electrode could act as a dopamine electrochemical sensor with a 0.099 μM detection threshold (Fig. 3.5).

3.3.4 Nano-medicine and Targeted Drug Delivery

CQDs have been proved to be nontoxic and been reported for in vivo studies; this made it valid for nano-medicine application. Bechet and colleagues indicated that CQDs can be used in photodynamic therapy, a clinical procedure used for surface tumor treatment. This technique includes the localization and accumulation of photosensitizers in the tumor tissue, followed by irradiation using an accurate wavelength, producing single oxygen species (Bechet et al. 2008). Andrius et al. in Du145 cells indicated implementation of CQDs in radiotherapy, where PEG-CQDs

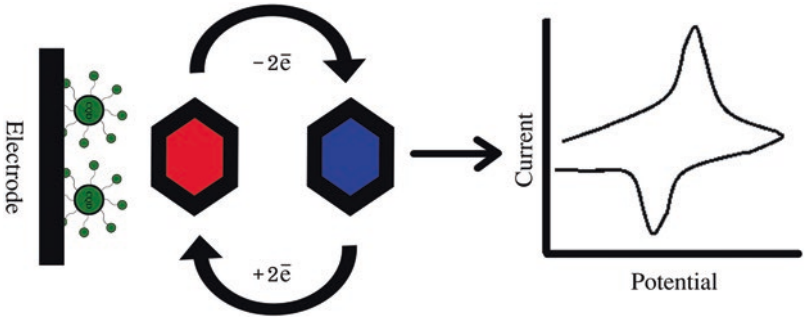

Fig. 3.5 Electrochemical sensing mechanism of electroactive molecules with CQDs: The CQDs catalyze the electrochemical redox process of biomolecules, and the resulting current is plotted against the potential

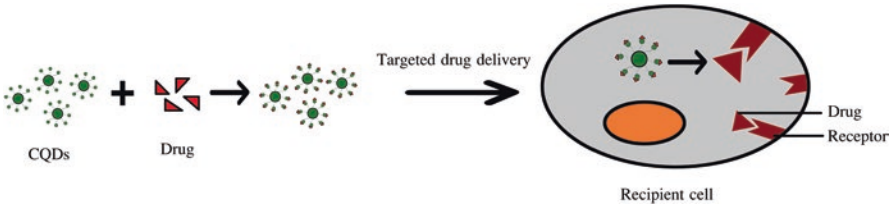

Fig. 3.6 Schematic representation of CQD-guided drug delivery system: Drugs conjugate with CQDs through its functional groups and directly deliver to the recipient cell. The released drug binds to the receptor site and acts specifically

covered with a silver shell were used as radio sensitizers (Kleinauskas et al. 2013). When exposed to low-energy X-rays, electrons were ejected from the coated silver shell, forming free radicals in the cancer cells adjacent to the CQDs-bPEI-coated CQDs (bPEI-CQDs), which have a large number of amino acids on their surface to condense DNA, making them suitable for gene carrier function (Hu et al. 2014). The nanotechnology-based drug delivery system has been gradually developed in recent years. The most explored drug delivery scheme is based primarily on AuNPs, but because of its toxicity, it limits its clinical treatment applications (Alkilany and Murphy 2010). Additionally, the requirement of thiol group for drug loading and the difficulty in tracking the AuNPs in in vivo system due to the quenching of fluorophores impose more limitation of drug choice (Kumar et al. 2013; Dulkeith et al. 2002). Therefore, CQDs serve as a good substitute for AuNPs, and different functionalizations could lead to better options for drug conjugation in conjunction with target agents and thus to a rise in drug delivery decisions (Fig. 3.6) (Kumar et al. 2013).

Zheng and team synthesized CQDs conjugated with platinum-based anticancer prodrug oxidized oxaliplatin through chemical coupling and were able to demonstrate the distribution of the conjugated CQDs by tracking the fluorescence signal (Zheng et al. 2014). The introduction of quinolone (photosensitive molecule) into

the CQD-based drug delivery scheme revealed a controlled release mechanism (Zheng et al. 2014). In this system, fluorescent characteristics of CQDs helped to activate photo-regulated drug release as a monitoring mechanism for drug delivery and quinoline molecules. Similarly, the release of pH-induced drugs was also recorded (Zheng et al. 2014; Karthik et al. 2013). A study on controlled release mechanism of DOX conjugated CQDs in HeLa cells showed that it drastically increased the drug efficacy. The CQDs-DOX conjugate did not exhibit any visible negative effect on normal cells, unlike on cancer cells (Gogoi and Chowdhury 2014). In addition, PEG-functionalized CQDs enabled longer circulation time in physiological systems and localized treatment there (Lai et al. 2012).

3.3.5 Tissue Engineering

CQDs hold excellent platform for tissue engineering, particularly mesenchymal stem cell (MSC)-based therapy (Shao et al. 2017a). CQDs were also apparently researched for implementation in bone tissue engineering, where the synthetic viable nanocomposite exhibited elevated load-bearing capacity and bioactivity. It is collectively called carbon dot decorated hydroxyapatite nanohybrid, a manufactured novel biomaterial. *Colocasia esculenta*'s aqueous extract of corms was taken as a precursor to CQDs and CaO from the shell of the egg served as a precursor to hydroxyapatite. It showed great biocompatibility, proliferation of cells, and activity of alkaline phosphatase against an osteoblast cell line, MG-63. The final nanocomposite verified its effectiveness as a bone-regenerating material by further enhancing its mechanical characteristics (Gogoi et al. 2016). In 2017, Dan Shao and colleagues created rat bone marrow mesenchymal stem cells (rBMSCs) based on citric acid carbon dots and their derivatives. The CQDs support long-term monitoring and differentiation of rBMSCs into osteoblasts by encouraging osteogenic transcription and mineralization of matrixes without influencing cell viability (Shao et al. 2017b). Alginate-derived CQDs have excellent gene delivery applicability as well as a fluorescent sample for visualizing the process of gene uptake. For the one-step green synthesis method, sodium alginate, a polysaccharide separated from seaweed, was used as a carbon source (Zhou et al. 2016). These CQDs which are positively charged have low toxicity, the ability to condense plasmid DNA, and the ability of CQDs produced by microwave-assisted pyrolysis to deliver SOX9 plasmid DNA in a non-viral manner. The successful delivery of SOX9 gene to the mouse embryonic fibroblasts (MEFs) induces chondrogenesis after in vitro transfection (Cao et al. 2018). Recently, collagen-derived carbon quantum dots with outstanding photo stability were also reported. Synthesized CQDs are reported to be deficient in photo-bleaching and are successfully introduced for the cell imaging in the 3D printed scaffold. It has an excellent application in the field of tissue engineering to monitor the efficiency and the success of regenerative medicine (Dehghani et al. 2018).

3.3.6 Anticancer Study

CQDs with hydroxyl group synthesized from styrene soot have been reported for its anticancer property. In this study, it was found that the CQDs have deleterious effect on the cancer cells (A549) and it showed negligible influence in non-cancerous cell (HEK 293T) (Fig. 3.7). The angiogenic study using these dots showed that it possesses anti-angiogenic properties too, as evidenced by the downregulation of angiogenic markers and fewer vessel density, when analyzed by CAM assay (Shereema et al. 2015). Walnut oil-synthesized CQDs through green synthesis also demonstrated anticancer activity against cell lines MCF-7 and PC-3. By activating caspase-3, the CQDs caused apoptosis in these cells (Arkan et al. 2018). CQDs produced from *L. plantarum* LLC-605 are capable of inhibiting the development of *E. coli* biofilm through a one-step hydrothermal carbonization technique and act as highly compatible and less toxic anti-biofilm material (Lin et al. 2018). Chang et al. produced CQDs from fresh tender ginger juice having on the surface groups of hydroxyl and carboxylate. They explain the efficient function of prepared CQDs in selective growth inhibition of hepatocarcinoma cell lines without influencing the growth of normal mammalian epithelial cells (MCF 10A) and liver cells (FL83B) in the suggested research. The flow cytometric assessment disclosed that a significant uptake of CQDs and the production of reactive oxygen species have been observed and induce apoptosis in HepG2 cells and are pointless in A549, MDA-MB-231, and HeLa cells (Li et al. 2014). In vivo studies were also carried out to prove the efficacy of CQDs to reduce tumor size that proposed the new strategy for therapeutic applications in liver cancer. Arkan et al.'s research showed that CQDs produced from walnut oil using hydrothermal techniques have cytotoxic and apoptotic potential on cells of prostate and breast cancer. Studies were conducted with PC-3 and MCF-7 cell lines, and the impact of prepared CQDs on caspase-3 and caspase-9 activity was verified. The findings acquired indicate that the CQDs specifically

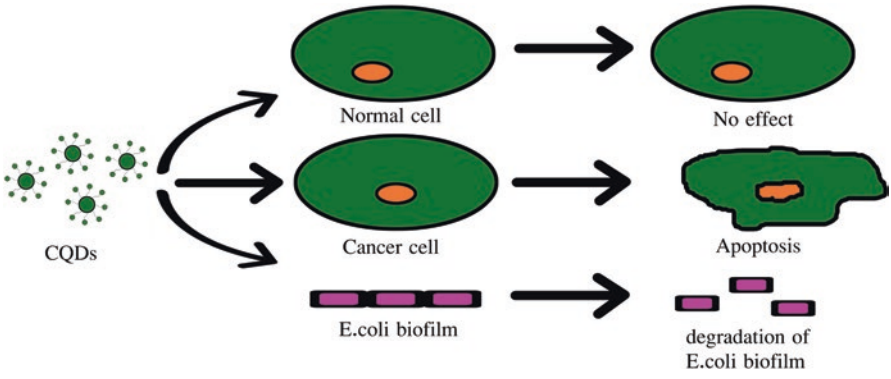

Fig. 3.7 The interaction of CQDS with anticancer property and ability to distinguish between a normal cell and cancer cell make it a perfect candidate for anticancer therapy, CQDs also possess potential antibiotic properties

boost the MCF-7 and PC3 cell lines caspase-3 activity. In addition, CQD levels also play a vital role in the operation of caspase-3. On the other hand, CQDs in the MCF-7 and PC-3 cell lines could not increase the activation of caspase-9 (Arkan et al. 2018). All these studies suggest that the starting material decides the functionality of the synthesized CQDs to a great extent and hence their capability to inhibiting the growth of different cancer cells. Other than the applications mentioned in this chapter, CQDs have been exploited for a plethora of application in different fields of science but mostly for its tunable fluorescence property. The application of CQDs holds promising prospects, and a plenty of applications are still to be explored in both biological and non-biological field.

3.4 Conclusion

- This carbon material possesses size less than 10 nm, which on surface passivation shows strong and tunable fluorescence emission properties and physical properties.
- High stability, excellent conductivity, and low toxicity among others are among their features.
- Different chemical, electrochemical, and physical methods can achieve CQD synthesis, which can be widely categorized into two kinds: top-down and bottom-up. Other methods, such as laser ablation, electrochemical synthesis methods, hydrothermal method, microwave-assisted pyrolysis method, ultrasonic method, acid dehydration method, etc., are frequently used.
- CQDs get great attraction due to their optical stability, nontoxicity, and solubility and can be easily functionalized with various functional group moieties or can be doped with heteroatoms on the surface, making them suitable candidate for biological applications like bio-imaging, nano-medicine and targeted drug delivery, bio-sensing, and electrochemical sensing.
- CQDs can be used as a significant material for in vitro and in vivo live cell imaging studies due to fluorescence and cell viability.
- It directly tags to the cellular organelles and reduces the drawbacks of organic fluorophore with high toxicity, photo-bleaching, etc.
- It is also known as multicolor nano-probe, due to excitation at different excitation wavelengths. Through the surface functional group they intract with different molecules in different way, this intraction helps to sense the presence of various materials, making it another promising application of CQDs known as bio-sensing.
- The ability to work as a drug delivery system makes CQDs an important factor in clinical and anticancer applications. Electrochemical sensor application of CQDs recently gained more attraction; tissue engineering and anticancer study is an important application.

- Due to the diversity in synthesis, surface functionalization and biological application make CQDs an important target for future applications.
- In a nutshell, CQDs are proving to be a good replacement for a plethora of nanomaterials present at this time period. CQDs have overcome the limitation with the currently used materials in all the mentioned applications. Thus, in a period of time, these dots may bring more refinement and more applications.

References

Alkilany AM, Murphy CJ (2010) Toxicity and cellular uptake of gold nanoparticles: what we have learned so far? J Nanopart Res 12(7):2313–2333

Arkan E, Barati A, Rahmanpanah M, Hosseinzadeh L, Moradi S, Hajialyani M (2018) Green synthesis of carbon dots derived from walnut oil and an investigation of their cytotoxic and Apoptogenic activities toward cancer cells. Adv Pharma Bull 8(1):149

Baker SN, Baker GA (2010) Luminescent carbon nanodots: emergent nanolights. Angew Chem Int Ed 49(38):6726–6744

Bechet D, Couleaud P, Frochot C, Viriot ML, Guillemin F, Barberi-Heyob M (2008) Nanoparticles as vehicles for delivery of photodynamic therapy agents. Trends Biotechnol 26(11):612–621

Bourlinos AB, Bakandritsos A, Kouloumpis A, Gournis D, Krysmann M, Giannelis EP, Polakova K, Safarova K, Hola K, Zboril R (2012) Gd (III)-doped carbon dots as a dual fluorescent-MRI probe. J Mater Chem 22(44):23327–23330

Cao L, Wang X, Meziani MJ, Lu F, Wang H, Luo PG, Lin Y, Harruff BA, Veca LM, Murray D, Xie SY (2007) Carbon dots for multiphoton bioimaging. J Am Chem Soc 129(37):11318–11319

Cao X, Wang J, Deng W, Chen J, Wang Y, Zhou J, Du P, Xu W, Wang Q, Wang Q, Yu Q (2018) Photoluminescent cationic carbon dots as efficient non-viral delivery of plasmid SOX9 and chondrogenesis of fibroblasts. Sci Rep 8(1):7057

Chandra P, Segal E (2016) Nanobiosensors for personalized and onsite biomedical diagnosis. The Institution of Engineering and Technology, London

Chandra S, Das P, Bag S, Laha D, Pramanik P (2011) Synthesis, functionalization and bioimaging applications of highly fluorescent carbon nanoparticles. Nanoscale 3(4):1533–1540

Chandra P, Tan YN, Singh SP (eds) (2017) Next generation point-of-care biomedical sensors technologies for cancer diagnosis. Springer, Singapore

Cui X, Zhu L, Wu J, Hou Y, Wang P, Wang Z, Yang M (2015) A fluorescent biosensor based on carbon dots-labeled oligodeoxyribonucleotide and graphene oxide for mercury (II) detection. Biosens Bioelectron 63:506–512

da Silva JCE, Gonçalves HM (2011) Analytical and bioanalytical applications of carbon dots. TrAC Trends Anal Chem 30(8):1327–1336

Dang YQ, Ren SZ, Liu G, Cai J, Zhang Y, Qiu J (2016) Electrochemical and capacitive properties of carbon dots/reduced graphene oxide supercapacitors. Nanomaterials 6(11):212

Dehghani A, Ardekani SM, Hassan M, Gomes VG (2018) Collagen derived carbon quantum dots for cell imaging in 3D scaffolds via two-photon spectroscopy. Carbon 131:238–245

Deng J, Lu Q, Hou Y, Liu M, Li H, Zhang Y, Yao S (2015) Nanosensor composed of nitrogen-doped carbon dots and gold nanoparticles for highly selective detection of cysteine with multiple signals. Anal Chem 87(4):2195–2203

Devi NR, Kumar TV, Sundramoorthy AK (2018) Electrochemically exfoliated carbon quantum dots modified electrodes for detection of dopamine neurotransmitter. J Electrochem Soc 165(12):G3112–G3119

Dias C, Vasimalai N, Sárria MP, Pinheiro I, Vilas-Boas V, Peixoto J, Espiña B (2019) Biocompatibility and bioimaging potential of fruit-based carbon dots. Nanomaterials 9(2):199
Dimos K (2016) Carbon quantum dots: surface passivation and functionalization. Curr Org Chem 20(6):682–695
Dong Y, Wang R, Li H, Shao J, Chi Y, Lin X, Chen G (2012) Polyamine-functionalized carbon quantum dots for chemical sensing. Carbon 50(8):2810–2815
Dong H, Kuzmanoski A, Gößl DM, Popescu R, Gerthsen D, Feldmann C (2014) Polyol-mediated C-dot formation showing efficient Tb 3+/Eu 3+ emission. Chem Commun 50(56):7503–7506
Dulkeith E, Morteani AC, Niedereichholz T, Klar TA, Feldmann J, Levi SA, Van Veggel FCJM, Reinhoudt DN, Möller M, Gittins DI (2002) Fluorescence quenching of dye molecules near gold nanoparticles: radiative and nonradiative effects. Phys Rev Lett 89(20):203002
Gao MX, Liu CF, Wu ZL, Zeng QL, Yang XX, Wu WB, Li YF, Huang CZ (2013) A surfactant-assisted redox hydrothermal route to prepare highly photoluminescent carbon quantum dots with aggregation-induced emission enhancement properties. Chem Commun 49(73):8015–8017
Gogoi N, Chowdhury D (2014) Novel carbon dot coated alginate beads with superior stability, swelling and pH responsive drug delivery. J Mater Chem B 2(26):4089–4099
Gogoi S, Kumar M, Mandal BB, Karak N (2016) A renewable resource based carbon dot decorated hydroxyapatite nanohybrid and its fabrication with waterborne hyperbranched polyurethane for bone tissue engineering. RSC Adv 6(31):26066–26076
Goh EJ, Kim KS, Kim YR, Jung HS, Beack S, Kong WH, Scarcelli G, Yun SH, Hahn SK (2012) Bioimaging of hyaluronic acid derivatives using nanosized carbon dots. Biomacromolecules 13(8):2554–2561
He H, Wang Z, Cheng T, Liu X, Wang X, Wang J, Ren H, Sun Y, Song Y, Yang J, Xia Y (2016) Visible and near-infrared dual-emission carbogenic small molecular complex with high RNA selectivity and renal clearance for nucleolus and tumor imaging. ACS Appl Mater Interfaces 8(42):28529–28537
He S, He P, Zhang X, Zhang X, Dong F, Jia L, Du L, Lei H (2018) Simultaneous voltammetric determination of guanine and adenine by using a glassy carbon electrode modified with a composite consisting of carbon quantum dots and overoxidized poly (2-aminopyridine). Microchim Acta 185(2):107
Hou Y, Lu Q, Deng J, Li H, Zhang Y (2015) One-pot electrochemical synthesis of functionalized fluorescent carbon dots and their selective sensing for mercury ion. Anal Chim Acta 866:69–74
Hu S, Liu J, Yang J, Wang Y, Cao S (2011) Laser synthesis and size tailor of carbon quantum dots. J Nanopart Res 13(12):7247–7252
Hu L, Sun Y, Li S, Wang X, Hu K, Wang L, Liang XJ, Wu Y (2014) Multifunctional carbon dots with high quantum yield for imaging and gene delivery. Carbon 67:508–513
Hua XW, Bao YW, Zeng J, Wu FG (2018) Ultrasmall all-in-one Nanodots formed via carbon dot-mediated and albumin-based synthesis: multimodal imaging-guided and mild laser-enhanced Cancer therapy. ACS Appl Mater Interfaces 10(49):42077–42087
Jaishankar M, Tseten T, Anbalagan N, Mathew BB, Beeregowda KN (2014) Toxicity, mechanism and health effects of some heavy metals. Interdiscip Toxicol 7(2):60–72
Jana J, Ganguly M, Pal T (2015) Intriguing cysteine induced improvement of the emissive property of carbon dots with sensing applications. Phys Chem Chem Phys 17(4):2394–2403
Jia X, Li J, Wang E (2012) One-pot green synthesis of optically pH-sensitive carbon dots with upconversion luminescence. Nanoscale 4(18):5572–5575
Kakaei K, Javan H, Mohammadi HB (2016) Synthesis of carbon quantum dots nanoparticles by cyclic voltammetry and its application as methanol tolerant oxygen reduction reaction electrocatalyst. J Chin Chem Soc 63(5):432–437
Karthik S, Saha B, Ghosh SK, Singh NP (2013) Photoresponsive quinoline tethered fluorescent carbon dots for regulated anticancer drug delivery. Chem Commun 49(89):10471–10473
Kleinauskas A, Rocha S, Sahu S, Sun YP, Juzenas P (2013) Carbon-core silver-shell nanodots as sensitizers for phototherapy and radiotherapy. Nanotechnology 24(32):325103

Kong B, Zhu A, Ding C, Zhao X, Li B, Tian Y (2012) Carbon dot-based inorganic–organic nanosystem for two-photon imaging and biosensing of pH variation in living cells and tissues. Adv Mater 24(43):5844–5848

Kumar V, Toffoli G, Rizzolio F (2013) Fluorescent carbon nanoparticles in medicine for cancer therapy. ACS Med Chem Lett 4(11):1012–1013

Lai CW, Hsiao YH, Peng YK, Chou PT (2012) Facile synthesis of highly emissive carbon dots from pyrolysis of glycerol; gram scale production of carbon dots/mSiO_2 for cell imaging and drug release. J Mater Chem 22(29):14403–14409

LeCroy GE, Sonkar SK, Yang F, Veca LM, Wang P, Tackett KN, Yu JJ, Vasile E, Qian H, Liu Y, Luo P (2014) Toward structurally defined carbon dots as ultracompact fluorescent probes. ACS Nano 8(5):4522–4529

Lee DE, Koo H, Sun IC, Ryu JH, Kim K, Kwon IC (2012) Multifunctional nanoparticles for multimodal imaging and theragnosis. Chem Soc Rev 41(7):2656–2672

Li H, He X, Kang Z, Huang H, Liu Y, Liu J, Lian S, Tsang CHA, Yang X, Lee ST (2010a) Water-soluble fluorescent carbon quantum dots and photocatalyst design. Angew Chem Int Ed 49(26):4430–4434

Li X, Wang H, Shimizu Y, Pyatenko A, Kawaguchi K, Koshizaki N (2010b) Preparation of carbon quantum dots with tunable photoluminescence by rapid *laser passivation in ordinary or*ganic solvents. Chem Commun 47(3):932–934

Li H, Zhang Y, Wang L, Tian J, Sun X (2011) Nucleic acid detection using carbon nanoparticles as a fluorescent sensing platform. Chem Commun 47(3):961–963

Li H, Kang Z, Liu Y, Lee ST (2012) Carbon nanodots: synthesis, properties and applications. J Mater Chem 22(46):24230–24253

Li CL, Ou CM, Huang CC, Wu WC, Chen YP, Lin TE, Ho LC, Wang CW, Shih CC, Zhou HC, Lee YC (2014) Carbon dots prepared from ginger exhibiting efficient inhibition of human hepatocellular carcinoma cells. J Mater Chem B 2(28):4564–4571

Li Y, Zhong Y, Zhang Y, Weng W, Li S (2015) Carbon quantum dots/octahedral Cu2O nanocomposites for non-enzymatic glucose and hydrogen peroxide amperometric sensor. Sensors Actuators B Chem 206:735–743

Liao B, Long P, He B, Yi S, Ou B, Shen S, Chen J (2013) Reversible fluorescence modulation of spiropyran-functionalized carbon nanoparticles. J Mater Chem C 1(23):3716–3721

Lin F, Li C, Chen Z (2018) Bacteria-derived carbon dots inhibit biofilm formation of Escherichia coli without affecting cell growth. Front Microbiol 9:259

Liu R, Wu D, Liu S, Koynov K, Knoll W, Li Q (2009) An aqueous route to multicolor photoluminescent carbon dots using silica spheres as carriers. Angew Chem Int Ed 48(25):4598–4601

Longmire M, Choyke PL, Kobayashi H (2008) Clearance properties of nano-sized particles and molecules as imaging agents: considerations and caveats. Nanomedicine (London) 3(5):703–717

Luo PG, Sahu S, Yang ST, Sonkar SK, Wang J, Wang H, LeCroy GE, Cao L, Sun YP (2013) Carbon "quantum" dots for optical bioimaging. J Mater Chem B 1(16):2116–2127

Maaoui H, Teodoresu F, Wang Q, Pan GH, Addad A, Chtourou R, Szunerits S, Boukherroub R (2016) Non-enzymatic glucose sensing using carbon quantum dots decorated with copper oxide nanoparticles. Sensors 16(10):1720

Maiti S, Das K, Das PK (2013) Label-free fluorimetric detection of histone using quaternized carbon dot–DNA nanobiohybrid. Chem Commun 49(78):8851–8853

Mao Y, Bao Y, Han D, Li F, Niu L (2012) Efficient one-pot synthesis of molecularly imprinted silica nanospheres embedded carbon dots for fluorescent dopamine optosensing. Biosens Bioelectron 38(1):55–60

Nguyen HV, Richtera L, Moulick A, Xhaxhiu K, Kudr J, Cernei N, Polanska H, Heger Z, Masarik M, Kopel P, Stiborova M (2016) Electrochemical sensing of etoposide using carbon quantum dot modified glassy carbon electrode. Analyst 141(9):2665–2675

Oza G, Oza K, Pandey S, Shinde S, Mewada A, Thakur M, Sharon M, Sharon M (2015) A green route towards highly photoluminescent and cytocompatible carbon dot synthesis and its separation using sucrose density gradient centrifugation. J Fluoresc 25(1):9–14

Qu K, Wang J, Ren J, Qu X (2013) Carbon dots prepared by hydrothermal treatment of dopamine as an effective fluorescent sensing platform for the label-free detection of iron (III) ions and dopamine. Chem Eur J 19(22):7243–7249

Ray SC, Saha A, Jana NR, Sarkar R (2009) Fluorescent carbon nanoparticles: synthesis, characterization, and bioimaging application. J Phys Chem C 113(43):18546–18551

Rodríguez-Padrón D, Algarra M, Tarelho LA, Frade J, Franco A, de Miguel G, Jiménez J, Rodríguez-Castellón E, Luque R (2018) Catalyzed microwave-assisted preparation of carbon quantum dots from lignocellulosic residues. ACS Sustain Chem Eng 6(6):7200–7205

Ruiyi L, Haiyan Z, Zaijun L, Junkang L (2018) Electrochemical determination of acetaminophen using a glassy carbon electrode modified with a hybrid material consisting of graphene aerogel and octadecylamine-functionalized carbon quantum dots. Microchim Acta 185(2):145

Shankar SS, Swamy BK (2014) Detection of epinephrine in presence of serotonin and ascorbic acid by TTAB modified carbon paste electrode: a voltammetric study. Int J Electrochem Sci 9(3):1321–1339

Shankar SS, Swamy BK, Ch U, Manjunatha JG, Sherigara BS (2009) Simultaneous determination of dopamine, uric acid and ascorbic acid with CTAB modified carbon paste electrode. Int J Electrochem Sci 4:592–601

Shankar SS, Swamy BK, Chandrashekar BN (2012) Electrochemical selective determination of dopamine at TX-100 modified carbon paste electrode: a voltammetric study. J Mol Liq 168:80–86

Shankar SS, Swamy BK, Chandrashekar BN, Gururaj KJ (2013) Sodium do-decyl benzene sulfate modified carbon paste electrode as an electrochemical sensor for the simultaneous analysis of dopamine, ascorbic acid and uric acid: a voltammetric study. J Mol Liq 177:32–39

Shankar SS, Shereema RM, Prabhu GRD, Rao TP, Swamy KB (2015) Electrochemical detection of dopamine in presence of serotonin and ascorbic acid at tetraoctyl ammonium bromide modified carbon paste electrode: a voltammetric study. J Biosens Bioelectron 6(2):1

Shao D, Lu M, Xu D, Zheng X, Pan Y, Song Y, Xu J, Li M, Zhang M, Li J, Chi G (2017a) Carbon dots for tracking and promoting the osteogenic differentiation of mesenchymal stem cells. Biomater Sci 5(9):1820–1827

Shao D, Lu MM, Zhao YW, Zhang F, Tan YF, Zheng X, Pan Y, Xiao XA, Wang Z, Dong WF, Li J (2017b) The shape effect of magnetic mesoporous silica nanoparticles on endocytosis, biocompatibility and biodistribution. Actabiomaterialia 49:531–540

Shen J, Zhu Y, Yang X, Li C (2012) Graphene quantum dots: emergent nanolights for bioimaging, sensors, catalysis and photovoltaic devices. Chem Commun 48(31):3686–3699

Shen T, Wang Q, Guo Z, Kuang J, Cao W (2018) Hydrothermal synthesis of carbon quantum dots using different precursors and their combination with TiO2 for enhanced photocatalytic activity. Ceram Int 44(10):11828–11834

Shereema RM, Sruthi TV, Kumar VS, Rao TP, Shankar SS (2015) Angiogenic profiling of synthesized carbon quantum dots. Biochemistry 54(41):6352–6356

Shereema RM, Rao TP, Kumar VS, Sruthi TV, Vishnu R, Prabhu GRD, Shankar SS (2018) Individual and simultaneous electrochemical determination of metanil yellow and curcumin on carbon quantum dots based glassy carbon electrode. Mater Sci Eng C 93:21–27

Srivastava S, Awasthi R, Tripathi D, Rai MK, Agarwal V, Agrawal V, Gajbhiye NS, Gupta RK (2012) Magnetic-nanoparticle-doped carbogenic nanocomposite: an effective magnetic resonance/fluorescence multimodal imaging probe. Small 8(7):1099–1109

Sun YP, Zhou B, Lin Y, Wang W, Fernando KS, Pathak P, Meziani MJ, Harruff BA, Wang X, Wang H, Luo PG (2006) Quantum-sized carbon dots for bright and colorful photoluminescence. J Am Chem Soc 128(24):7756–7757

Tang Y, Su Y, Yang N, Zhang L, Lv Y (2014) Carbon nitride quantum dots: a novel chemiluminescence system for selective detection of free chlorine in water. Anal Chem 86(9):4528–4535

Tao H, Yang K, Ma Z, Wan J, Zhang Y, Kang Z, Liu Z (2012) In vivo NIR fluorescence imaging, biodistribution, and toxicology of photoluminescent carbon dots produced from carbon nanotubes and graphite. Small 8(2):281–290

Thongpool V, Asanithi P, Limsuwan P (2012) Synthesis of carbon particles using laser ablation in ethanol. Procedia Eng 32:1054–1060

Timur SA (2018) Doped carbon dots for sensing and bioimaging applications: a minireview. Nano 8:342–352

Wang F, Xie Z, Zhang H, Liu CY, Zhang YG (2011) Highly luminescent organosilane-functionalized carbon dots. Adv Funct Mater 21(6):1027–1031

Wang Q, Liu X, Zhang L, Lv Y (2012) Microwave-assisted synthesis of carbon nanodots through an eggshell membrane and their fluorescent application. Analyst 137(22):5392–5397

Wang K, Gao Z, Gao G, Wo Y, Wang Y, Shen G, Cui D (2013a) Systematic safety evaluation on photoluminescent carbon dots. Nanoscale Res Lett 8(1):122

Wang CI, Periasamy AP, Chang HT (2013b) Photoluminescent C-dots@ RGO probe for sensitive and selective detection of acetylcholine. Anal Chem 85(6):3263–3270

Wang L, Zhu SJ, Wang HY, Qu SN, Zhang YL, Zhang JH, Chen QD, Xu HL, Han W, Yang B, Sun HB (2014) Common origin of green luminescence in carbon nanodots and graphene quantum dots. ACS Nano 8(3):2541–2547

Wang J, Wei J, Su S, Qiu J (2015a) Novel fluorescence resonance energy transfer optical sensors for vitamin B 12 detection using thermally reduced carbon dots. New J Chem 39(1):501–507

Wang J, Su S, Wei J, Bahgi R, Hope-Weeks L, Qiu J, Wang S (2015b) Ratio-metric sensor to detect riboflavin via fluorescence resonance energy transfer with ultrahigh sensitivity. Phys E 72:17–24

Xu X, Ray R, Gu Y, Ploehn HJ, Gearheart L, Raker K, Scrivens WA (2004) Electrophoretic analysis and purification of fluorescent single-walled carbon nanotube fragments. J Am Chem Soc 126(40):12736–12737

Xu Y, Jia XH, Yin XB, He XW, Zhang YK (2014) Carbon quantum dot stabilized gadolinium nanoprobe prepared via a one-pot hydrothermal approach for magnetic resonance and fluorescence dual-modality bioimaging. Anal Chem 86(24):12122–12129

Xu J, Zhou Y, Cheng G, Dong M, Liu S, Huang C (2015) Carbon dots as a luminescence sensor for ultrasensitive detection of phosphate and their bioimaging properties. Luminescence 30(4):411–415

Yang ST, Wang X, Wang H, Lu F, Luo PG, Cao L, Meziani MJ, Liu JH, Liu Y, Chen M, Huang Y (2009) Carbon dots as nontoxic and high-performance fluorescence imaging agents. J Phys Chem C 113(42):18110–18114

Yang Y, Cui J, Zheng M, Hu C, Tan S, Xiao Y, Yang Q, Liu Y (2012) One-step synthesis of amino-functionalized fluorescent carbon nanoparticles by hydrothermal carbonization of chitosan. Chem Commun 48(3):380–382

Yin JY, Liu HJ, Jiang S, Chen Y, Yao Y (2013) Hyperbranched polymer functionalized carbon dots with multistimuli-responsive property. ACS Macro Lett 2(11):1033–1037

Zhai X, Zhang P, Liu C, Bai T, Li W, Dai L, Liu W (2012) Highly luminescent carbon nanodots by microwave-assisted pyrolysis. Chem Commun 48(64):7955–7957

Zhao Z, Xie Y (2017) Enhanced electrochemical performance of carbon quantum dots-polyaniline hybrid. J Power Sources 337:54–64

Zhao HX, Liu LQ, De Liu Z, Wang Y, Zhao XJ, Huang CZ (2011) Highly selective detection of phosphate in very complicated matrixes with an off–on fluorescent probe of europium-adjusted carbon dots. Chem Commun 47(9):2604–2606

Zheng H, Wang Q, Long Y, Zhang H, Huang X, Zhu R (2011) Enhancing the luminescence of carbon dots with a reduction pathway. Chem Commun 47(38):10650–10652

Zheng M, Liu S, Li J, Qu D, Zhao H, Guan X, Hu X, Xie Z, Jing X, Sun Z (2014) Integrating oxaliplatin with highly luminescent carbon dots: an unprecedented theranostic agent for personalized medicine. Adv Mater 26(21):3554–3560

Zhou J, Booker C, Li R, Zhou X, Sham TK, Sun X, Ding Z (2007) An electrochemical avenue to blue luminescent nanocrystals from multiwalled carbon nanotubes (MWCNTs). J Am Chem Soc 129(4):744–745

Zhou J, Deng W, Wang Y, Cao X, Chen J, Wang Q, Xu W, Du P, Yu Q, Chen J, Spector M (2016) Cationic carbon quantum dots derived from alginate for gene delivery: one-step synthesis and cellular uptake. Actabiomaterialia 42:209–219

Zhu H, Wang X, Li Y, Wang Z, Yang F, Yang X (2009) Microwave synthesis of fluorescent carbon nanoparticles with electrochemiluminescence properties. Chem Commun 34:5118–5120