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Effects of representative quantum dots on microorganism and phytoplankton: A comparative study

Received 00th January 20xx,
Accepted 00th January 20xx

DOI: 10.1039/x0xx00000x

www.rsc.org/

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With the increasing applications of semiconductor particles, especially metal-based quantum dots and carbon quantum dots (CQDs), these quantum dots would inevitably be released into the environment, therefore their effects on biota should be assessed. Few studies on the effects of CQDs in microorganism and phytoplankton were performed. In this paper, we put forward more effective and convenient approaches to prepare three kinds of high-quality CQDs.Then we assessed their effects on *Staphylococcus aureus* and *Microcystis aeruginosa* firstly which were the representative of microorganism and phytoplankton,respectively,and compared with the effects of metal-based QDs (CdSe-QDs or CdTe-QDs).The results showed that CQDs at low concentrations (<50 $mg.L^{-1}$) had insignificantly effects on the growth of *Staphylococcus aureus* and *Microcystis aeruginosa*, while the influence of the CQDs was observed significantly when the concentration was increased up to 100 $mg.L^{-1}$. However, the negative impact of metal-based QDs was observed at any given concentration. In conclusion, the results demonstrated that any kind of carbon quantum dots has lower ecological risks than metal-based QDs, which may provide reference to utilize carbon quantum dots better and safely.

Introduction

Due to the advantages in its physical characteristics, quantum dots (QDs) have increasing use for commercial purposes during the last decades. As a semiconductor particle, quantum dots may be divided into two large subcategories, carbon-based quantum dots and metal-based quantum dots, which have been employed for many applications in photoelectric equipment, biological imaging, chemical sensors, and drug delivery. However, these semiconductor particles would be discharged, directly or indirectly, in the aquatic and terrestrial ecosystems with the extensive applications. For example, CdTe-QDs are increasingly measured in the nature water body. [1,2]Three sources of CdTe-QDs have been considered as the industrial wastewater, medical sewage and research usage. Thus, the potential environmental risks of these semiconductor particles should be concerned. So far, a variety of research have been published to evaluate the potential toxicity of quantum dots with heavy mental on species, such as [3]human cell, [4]rats, [5]invertebrate and [6,7]so on. Compared with metal-based quantum dots, carbon-based quantum dots have attracted higher attention owing to superior properties, as well as their promising application in various fields. Up to now, [8,9]there are also many investigations focused on the toxicity of carbon quantum dots on human cell. But their conclusion could not be applied on the all species, especially the single-celled organisms, such as microorganism and phytoplankton. [10]Because the microbes play an important role in ecological environment system, and the proportion of bacteria is as high as 90%. In addition, phytoplankton serves as an important component of the aquatic ecosystem. [11]Potential environmental impacts on these autotrophs could decrease the primary productivity, influence the entire food chain and change the structure and function of the whole ecosystem. Both of microorganism and phytoplankton are widely existed, and their reflection on environmental change is also most sensitive. So far, few literatures investigated the biosecurity of carbon quantum dots in microorganism and phytoplankton[12-15]. For instance, [12]nanocarbons synthesized from acetylene and benzene can destroy the *Escherichia coli* and *Staphylococcus aureus*, and limit the oxygen transport from environment. In addition, [13]Pereira et al. and [14]Zhang et al. have investigated the interactions of carbon nanotubes with green algae. The results showed that NPs inhibited algae growth by disturbing inducing oxidation, disturbing ATP production, decreasing photosynthetic activity and physical stress. Yet, the knowledge on the ecotoxicological effects of carbon quantum dots is still limited. Therefore, more experiments should be carried to assess the effect of carbon quantum dots on microorganism and phytoplankton.

In this work, we put forward more effective and convenient approaches to prepare three kinds of carbon quantum dots with superior optical properties. All of them exhibited small size, high aqueous solubility and bright blue fluorescence. Then, a series of assays were conducted to

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DOI: 10.1039/x0xx00000x

assess the effects of CQDs and metal-based QDs (CdSe-QDs or CdTe-QDs) on *Staphylococcus aureus(S.aureus)* and *Microcystis aeruginosa(M.aeruginosa)* which were the representative of microorganism and phytoplankton, respectively. [17,18] *S. aureus* is a kind of widely existed Gram-positive bacteria with high sensitivity to poisonous substance, while *M. aeruginosa* as a kind of freshwater cyanobacteria can reflect the condition of aquatic ecosystem. It is the first time to report the effects of carbon quantum dos passivated various agent on *S. aureus* and *M. aeruginosa*, hoping to provide reference to understand the effect of carbon quantum dots on ecosystem and utilize them better and safely.

Experimental

Materials

Activated carbon was made in the laboratory. Acetic acid (A.R., Nanjing Chemical Reagent Co.,Ltd), Hydrogen peroxide(30%,A.R.,Sinopharm Chemical Reagent Co., Ltd),Diethyl ether(A.R., Nanjing Chemical Reagent Co., Ltd.) PEG_{2000}(A.R., Xilong Chemical industry Co.Ltd), Citric acid (CA, Nanjing chemical reagent co., Ltd, AR), Glycine (Gly, Yika biotech, Nanjing, BR), N-butyl alcohol (Chinasun specialty products co., Ltd, AR), sodium hydroxide (West Long chemical co., Ltd, AR). All the chemicals were of analytical grade and used as received. Double distilled water was used throughout the experiments.

S. aureus (GIM1.142) sample were provided by the Guangdong Biological Germplasm Resource Bank, China. *S. aureus* were cultivated in Luria-Bertani (LB) culture medium (pH 7.4) which contains Tryptone, 10g/L Yeast extract and 5g/L NaCl.

The freshwater cyanobacteria *M. aeruginosa* (FACHB-1005) was obtained from the freshwater algae culture collection of the Institute of Hydrobiology (FACHB-Collection) of the Chinese Academy of Sciences. Cells of the cyanobacteria was incubated and maintained at 26 ± 1 °C under an illumination intensity of 2000 lux, with a 12 h/12 h light/dark interval. BG-11 medium was applied as the culture media which was adjusted to pH 8.0 with NaOH and HCL.

Instrumentation

Teflonlined autoclave (Zhenghong Plastics Co.,Ltd.) was used to prepare the citric acid-passivized carbon quantum dots and PEG_{2000}-passivated carbon quantum dots *S. aureus* were cultivated by a rotary shaker(Peiying Co.,Ltd.). Electronic microscope (JEM-2100 120 kV, JEOL), UV-vis absorption spectrophotometer (UV 2100, Shimadzu, Kyoto, Japan) and fluorescence spectrometer (RF-5301PC, Hitachi, Tokyo, Japan) were used to characterize the properties of CQDs. All pH values were measured with a pHS-25 pH meter (Shanghai INESA Scientific Instrument Co.,Shanghai, China).

Preparation of PEG_{2000}-CQDs, CA-CQDs, Gly-CQDs

PEG_{2000}-passivated carbon quantum dots(PEG_{2000}-CQDs) were prepared via a simple chemical oxidation proposed earlier by our group[19].Briefly, activated carbon was dissolved in a mixture of hydrogen peroxide and acetic acid, then refluxed at 100℃ for 12h.The obtained bare CQDs were purified by diethyl ether and passivized by PEG_{2000}.

Citric acid-passivated carbon quantum dots (CA-CQDs) were prepared by hydrothermal treatment of citric acid (CA). In a typical synthesis, CA (0.5 g) was added into H_2O (20 mL). Then the mixture was transferred into a 60 mL Teflonlined autoclave and heated at 200℃ for a period of 4 h and then cooled down to ambient temperature naturally. The obtained CA-CQDs were dispersed in water for further characterization and use.

Glycine-passivated carbon quantum dots (Gly-CQDs) were prepared using method reported earlier by our group[20]. Briefly, 3.6g CA and 1.5g Gly were dissolved in 30mL of distilled water and transferred to a 60mL Teflonlined autoclave. The reaction was maintained at 200℃ for 4h. The obtained burgundy solution was purified by extracting with N-butyl alcohol. Finally, the purified Gly-CQDs solution was dried and stored for further study.

The comparative assays on *S. aureus* and *M. aeruginosa*

In order to evaluate the effects of the three kinds of CQDs on *S. aureus* and *M. aeruginosa*, CdSe-QDs and CdTe-QDs were adopted as representative metal-based materials, which were synthetized by our lab[21, 22, 23]. Typically, CdTe-QDs modified with TGA were prepared from cadmium chloride and NaHTe solutions. Tellurium powder was mixed with sodium borohydride under N_2 for 30 min at 45℃.Then the mixture was injected into an N_2-saturareted Cd^{2+}-TGA precursor solution (the molar ratio of Cd:Te:TGA was 1:0.5:2.4). Finally, the mixture was refluxed at a temperature of 100℃ for 30 min. The synthesis of CdSe-QDs stabilized with TGA was similar to CdTe-QDs. Briefly, the mixture of Se powder and sodium borohydride were injected into Cd^{2+}-TGA. CdSe-QDs were obtained from the mixture refluxed for 60min under a nitrogen atmosphere.

Exposure experiments were carried out under the same temperature as the stock cultures. The bacterial suspension and phytoplankton inoculum were prepared for each experiment from their respective fresh culture stocks sampled during the exponential growth phase. The experiments of effects were performed in two parts: effects of the nano-materials on *S. aureus* and effects on *M. aeruginosa*. In the Part I, a quantity of CA-CQDs, Gly-CQDs and CdSe-QDs were dissolved in LB culture medium (final concentration=500mg/L), respectively. 0, 0.5, 1, 5, 10, 50mL of above three kinds of solution were added into 250mL Erlenmeyer flasks (final concentration: 0, 5, 10, 50, 100, 500mg/L), respectively. For each of these flasks, a certain volume of *S. aureus* solution was added accurately and placed on a rotary shaker (37℃, 250rpm) for different times ranging between 0 h and 24 h. Then, absorptivity in 610nm of each specimen was measured using UV-vis absorption spectrophotometer.

In the Part II, an equal volume of the fresh culture medium of 100 mL in the presence of PEG_{2000}-CQDs at various concentrations was added to the phytoplankton pellets in 250 mL of the previously sterilized conical flask. The corresponding PEG_{2000}-CQDs concentrations were 5, 10, 50, 100, 500 $mg \bullet L^{-1}$ respectively. As for CdTe-QDs, all treatments were processed

to be similar to PEG_{2000}-CQDs while with the corresponding CdTe-QDs concentrations were 0.2, 0.5, 1.0, 10.0, 20.0 mg•L^{-1} respectively. The phytoplankton culture without any quantum dots was used as a control. Samples were removed from the culture vessels at a predetermined time every 24 h. The cells were observed microscopically. [24]Chlorophyll and soluble proteins were analyzed using the standard method. The population growth rate (r) was calculated from the formula (1):

$$r = \frac{lnN_t - lnN_0}{t} \quad (1)$$

Where N_t and N_0 are population sizes at day 0 and day t, and t is time in days when the population size is maximum. Each experiment was replicated in three times. All the data analyses were carried out with the SPSS analytic package 16.0. Data were first tested for homogeneity (Levene's test). Variables from the results of Experiment I and II were examined by one-way analysis of variance (ANOVA) to identify the significant differences. All the figures were produced using Sigmaplot Version 12.0.

Results and discussion

Characterization

The characterization of PEG_{2000}-CQDs was reported earlier by our group[25]. As shown in Fig.1 and Fig.2, CA-CQDs and Gly-CQDs were characterized by TEM images, UV absorption spectra, FL spectra and FT-IR spectrum. It can be seen that two kinds of spherical particles were less than 10 nm in size and dispersed evenly (see Fig.1).

As shown in Fig.2a, CA-CQDs emitted blue light (485 nm) when excited with a 360 nm UV beam, while CQDs functioned by glycine emitted blue light (485nm) when excited with a 380nm UV beam (see Fig.2b).Using quinine sulfate as a reference, the quantum yield of the prepared PEG_{2000}-CQDs, CA-CQDs and Gly-CQDs measured by the equation were found to be 19.6%, 9.8% and 47%.

FT-IR spectrum was recorded to provide further structural insights about the CA-CQDs and Gly-CQDs (Fig. 2b).As shown in curve a, CQDs sample treated with CA showed characteristic absorption peaks of OH at 3460cm^{-1} and the stretching vibration band of C=O bond in the carboxylic acid at 1610cm^{-1},[26,27]which contributes to good water-solubility. [28, 29]According to literature, the O-H bond,-COOH and C=O of CA might interacted with each other under surpressure and high temperature, causing incomplete dehydration and carbonization. It may lead to form fluorescence CQDs with the hydroxyl group, carbanyl group and carboxyl group on the surface and carbon-carbon bond at the core. Compared with the curve a, the FT-IR spectrum of Gly-CQDs showed many characteristic absorption bands containing amide groups at 1628 cm^{-1} and 3458cm^{-1}, methylene group at 2936 cm^{-1}, and cyano group at 1186 cm^{-1}, but nearly no characteristic absorption of CA. Moreover, the stretching vibration of C=O bond at 1713 cm^{-1} was also detected, indicating that the Gly functioned CQDs with amide linkage (–CONH–).

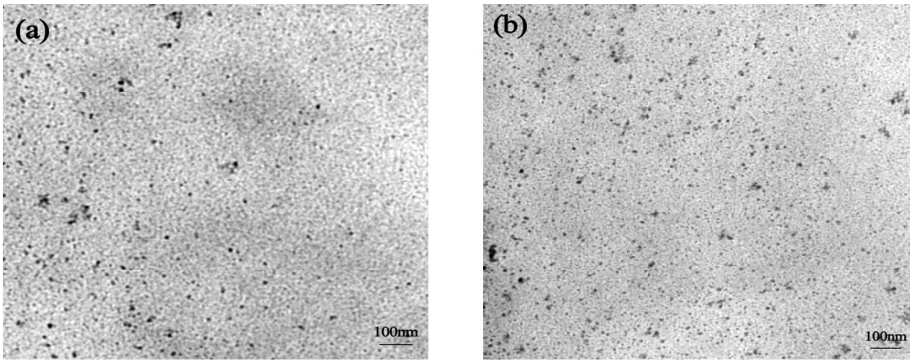

Fig.1 TEM images of CA-CQDs(a) and Gly-CQDs(b)

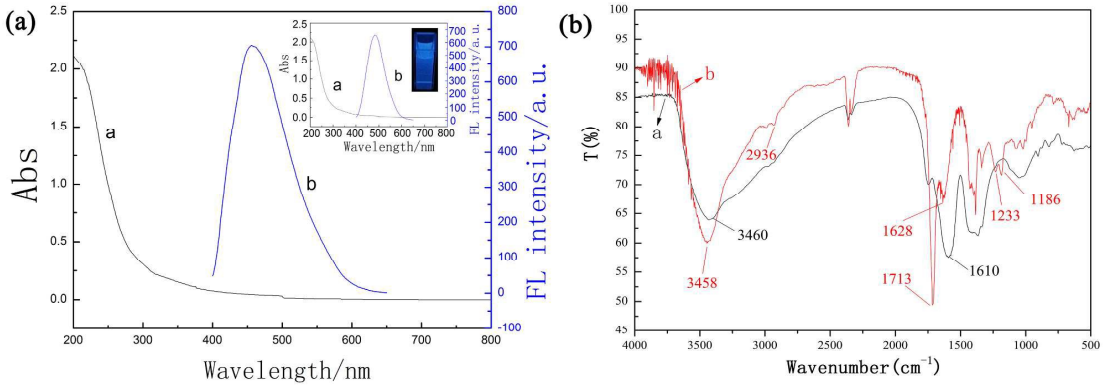

Fig.2 (a).UV absorption spectra and FL spectra of CA-CQDs with the excitation of 360 nm (a is UV spectra, b is FL spectra). Inset: UV absorption spectra and FL spectra of Gly-CQDs and image of the solution of Gly-CQDs obtained under 365 nm UV light. (b)FT-IR spectra of CA-CQDs (curve a), Gly-CQDs (curve b)

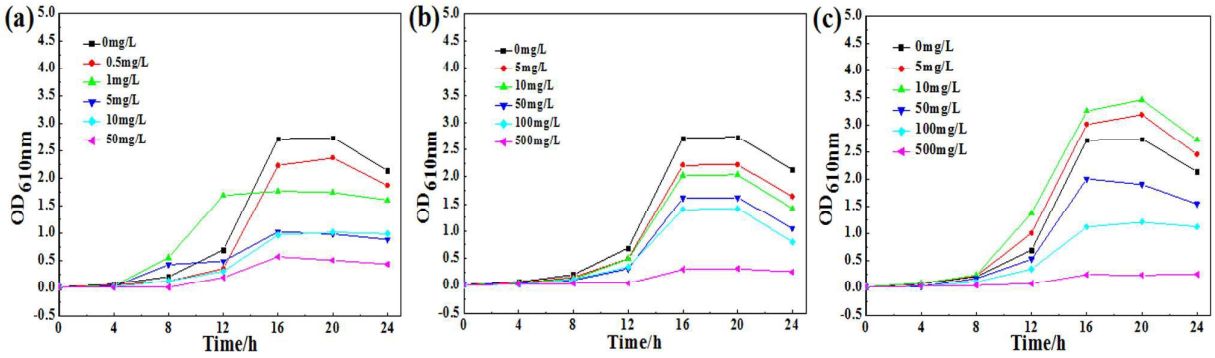

Fig.3 Growth curves with (a)CdSe-QDs-treated *S.aureus*,(b) CA-CQDs-treated *S.aureus*,(c) Gly-CQDs-treated *S.aureus*

Analysis of the effect on *S. aureus*

A series of assays were conducted to assess the effects of three different QDs (CA-CQDs, Gly-CQDs and CdSe-QDs) to the *S. aureus* respectively by means of the optical density method.

A series of LB medium spiked with different concentration of CdSe-QDs were also determined optical density to investigate the effect of CdSe-QDs, which is a representative of quantum dots constituted heavy metals. As shown in Fig.3(a), *S.aureus* was significantly inhibited by even a low concentration of CdSe-QDs. For bacteria grown with 1 mg/L CdSe-QDs, bacteriostatic rate was approximately 40%, suggesting growth environment was fairly unsuitable for *S.aureus*. Growth rates of bacteria had declined with the concentration of CdSe-QDs increased, and then the biomass stopped growing with concentration over 5mg/L. It was indicated that CdSe-QDs exhibited strong inhibition on *S.aureus*. [30]The different core materials of QDs were the principal reason for their different toxicities. Cadmium is toxic, even at a low concentration, and it can be released form the CdSe-QDs, while carbon was less toxic to the strain than cadmium.

Series of the same amount of *S. aureus* suspensions were inoculated into fresh LB medium spiked with CA-CQDs ranging in initial concentration from 0 to 500mg/L and the results are shown in Fig.3(b). It turned out that the growth tendencies of *S. aureus* in different concentrations of CA-CQDs are nearly in accord with *S. aureus* suspensions without any CQDs. All of these growth curves peaked nearly at 20h and the inhibition rates of *S. aureus* in that hour respectively were 18.82%, 25.40%, 40.75%, 48.17%, 8.56%(different concentration of CA-CQDs: 5, 10, 50, 100, 500mg/L).The result indicated that *S. aureus* was insusceptible when the concentration was less than10mg/L, and the inhibition effect had strengthened with the increase of CA-CQDs. As the initial concentration of CA-CQDs increased to 500mg/L, the level of growth declined, indicating high-concentration can significantly inhibit *S. aureus*.

Compared with the blank control group, growing status of *S.aureus* were different under different concentrations of Gly-CQDs. Not only have *S. aureus* been suppressed by Gly-CQDs (<10mg/L), they have increased better than those of the blank control group, which may be partly due to Gly-CQDs containing nitrogen element contributes to the growth of bacteria (See Fig.3(c)).The inhibitory effects under low concentration had been dulled by positive effects of nitrogen element, hence, *S. aureus* increased rather than decreased. Growth inhibition of bacteria can be observed obviously with increase of Gly-CQDs concentration ranging from 50mg/L to 500mg/L. The results shows that bacteriostatic rates of Gly-CQDs at 20h were 30.59%, 56.14%, 91.45% corresponded to serial concentration respectively (50、100、500mg/L) .It was indicated that positive effects of nitrogen element cannot be enough to offset toxicity effects on bacteria with the increase of Gly-CQDs concentration which can make the microflora to decrease. However, the investigation on *S. aureus* was still at the primary stage, effects of CQDs on other aspects of *S. aureus* (eg. metabolism) were unclear. We will carry more experiments to deepen the research in the follow-up study.

Analysis of the effect on *M. aeruginosa*

The population growth curves of *Microcystis aeruginosa* under five concentrations of PEG_{2000}-CQDs and CdTe-QDs were presented in Fig.4. In general, the population increased during 144 h (6 days) under most concentrations except 500 mg•L^{-1}(Fig.4(a)). The population dynamic at 5, 10, 50 mg•L^{-1} indicated the PEG_{2000}-CQDs at the three concentrations did not produce significant inhibition on the growth of *M. aeruginosa* ($p > 0.05$, F-test). The cyanobacteria population in the three concentrations grew slowly in the first 48 h and then increased faster until the end of the experiment. The maximum population size was 10.2 ± 0.14, 10.07 ± 0.18 9.25 ± 0.39 × 106 cell•mL^{-1}, respectively, which was only 0.79%, 1.34%, 4.96% lower than that in control. However, PEG_{2000}-CQDs produced a negative impact on the growth of *M. aeruginosa* at 100 mg•L^{-1}, the maximum population size was 6.06 × 106 cell•mL^{-1} on average and the population growth rate was 0.30 d^{-1}, which was 76.97% and 58.33% of that in control, respectively. In addition, the negative trend in the growth of *M. aeruginosa* was observed significantly when the concentration was 500 mg•L^{-1}. The maximum population size was only 0.14 ± 0.02 cell•mL^{-1}, which was only 1.35% of that in control. In comparison, CdTe-QDs produced a negative impact on the growth of *M. aeruginosa*, regardless of the concentration (Fig.4(b)). When the concentrations were lower than 1.0 mg.L^{-1}, the population increased until the end of the experiment. The population dynamics at 0.2 and 0.5 mg•L^{-1} indicated that CdTe-QDs had a significant effect on the growth of *M. aeruginosa*, respectively ($p < 0.05$, F-test). The maximum population size was 6.31 ± 0.67 and 3.48 ± 0.68 × 106 cell•mL^{-1} on average, which was 61.62% and 33.98% of that in control, respectively. In addition, the impact strengthened with the concentration increasing. CdTe-QDs produced a significant inhibition on the growth of *M. aeruginosa* when the concentration at 1.0, 10 or 20 mg•L^{-1}. The population growth rate was -0.37, -0.68 and -0.69 d^{-1} respectively. Most broken cells were observed microscopically.

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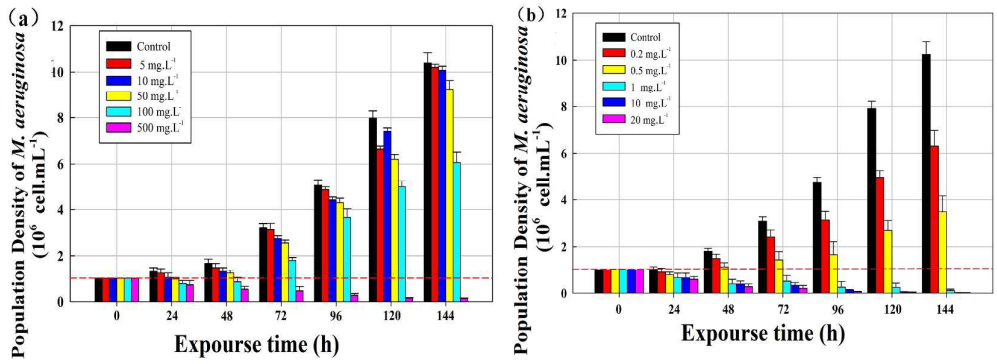

Fig.4 The population growth curves of *M. aeruginosa* under five concentrations of PEG_{2000}-CQDs (a) and CdTe-QDs (b).

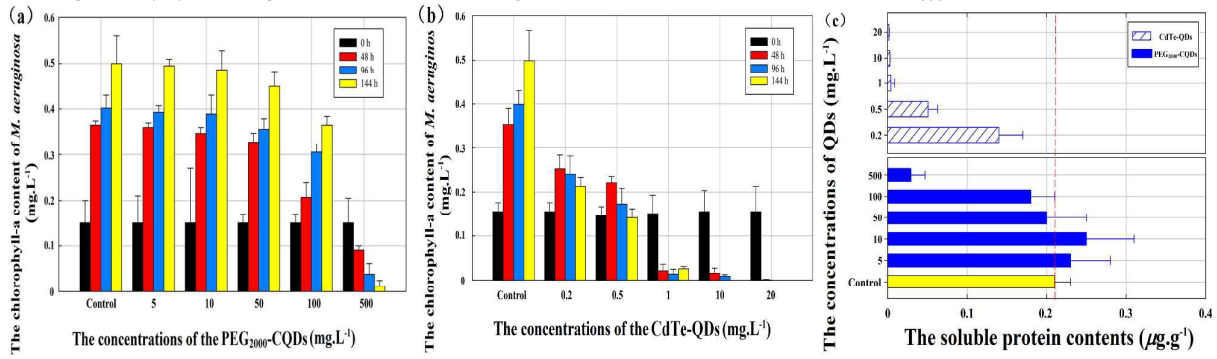

Fig.5 (a), (b) The chlorophyll-a content of *M. aeruginosa* varied under given concentrations of the PEG_{2000}-CQDs and CdTe-QDs.
(c) The soluble protein contents in the cell of *M. aeruginosa* varied under given concentrations of the PEG_{2000}-CQDs and CdTe-QDs at 144 h.

The chlorophyll-a content of *M. aeruginosa* varied under the given concentrations of the PEG_{2000}-CQDs and control were presented in Fig.5(a). The trend of the PEG_{2000}-CQDs was observed clearly. Chlorophyll-a was accumulated during 144 h (6 days) under most concentrations except 100 and 500 mg•L^{-1}. Statistical analysis showed that the PEG_{2000}-CQDs at 5, 10, 50 mg•L^{-1} had no significant effect on the chlorophyll-a increasing ($p > 0.05$, F-test). In addition, the chlorophyll-a content of *M. aeruginosa* was accumulated under 100 mg•L^{-1} of the QDs during the whole experiment time. The influence of the PEG_{2000}-CQDs was observed clearly. The content of chlorophyll-a at 144 h was 0.37 ± 0.02 mg•L^{-1}, which was 73.00 % of that in control. However, the chlorophyll-a content of *M. aeruginosa* declined significantly when exposed to the PEG_{2000}-CQDs at 500 mg•L^{-1}. It was 0.012 ± 0.01 mg•L^{-1} at the end of the experiment, only 2.42% of that in control. In comparison, CdTe-QDs produced obvious impacts on the photosynthesis of *M. aeruginosa*, regardless of the concentration. The content of chlorophyll-a was almost lower than the control at any given concentration. Although the chlorophyll-a of *M. aeruginosa* was accumulated in 48 h at 0.2 and 0.5 mg•L^{-1}, it declined rapidly from the second day. The content of chlorophyll-a at the end of the experiment was 0.21± 0.02 and 0.14 ± 0.02 mg•L^{-1} respectively, only 42.69% and 28.26% of that in control. When the concentration of CdTe-QDs arrived up to 1 mg•L^{-1} or higher one, the contents of chlorophyll-a dropped near to zero at 144 h finally.

The soluble protein contents in the cell of *M. aeruginosa* under given concentrations of the PEG_{2000}-CQDs and CdTe-QDs at 144 h were presented in Fig. 5(b). The soluble protein contents of the cell was induced at the relatively low concentrations (5 and 10 mg•L^{-1}), whereas was inhibited at the relatively high concentrations (> 10 mg•L^{-1}). The results showed that the soluble protein content was 9.52% and 8.70% more than that of the control at 5 and 10 mg•L^{-1}, respectively, while the value was 95.24% and 85.71% of that in control at 50 and 100 mg•L^{-1}. However, the concentration of 500 mg•L^{-1} produced a significant impact on the *M. aeruginosa*, which made the soluble protein content decline to 0.03 μg•g^{-1}, only 13.81% of the control. In comparison, the content of the soluble protein was almost lower than the control at any given concentration of CdTe-QDs. The CdTe-QDs exhibited a significantly negative influence on the soluble protein of the plankton ($p < 0.01$, F-test). The soluble protein content decreased with the CdTe-QDs

concentrations increasing. When the concentration arrived up to 1 mg•L^{-1} or a higher one, the contents dropped near to zero.

The median effective concentrations (EC_{50}) of the PEG_{2000}-CQDs and CdTe-QDs on the growth of *M. aeruginosa* were present in Figure 6. For the PEG_{2000}-CQDs, the inhibition effect was declining during the exposure time. At the first 24 h, the EC_{50} was 8.73 mg•L^{-1}, impliying a moderate toxicity. After the 48 h, the EC_{50} value increased rapidly and arrived at the maximum value (57.64 mg•L^{-1}) at 144 h, seven times of 24 h-EC_{50}, indicating a low toxicity. Oppositely, the EC_{50} of CdTe-QDs on the growth of cyanobacteria was much lower than that of the PEG_{2000}-CQDs at any exposed time.

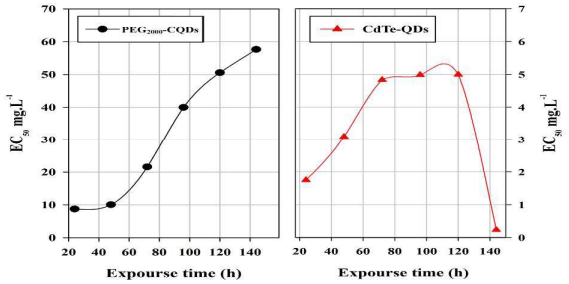

Fig. 6 The EC_{50} of the PEG_{2000}-CQDs and CdTe-QDs on the growth of *M. aeruginosa*

A three-stepwise toxic effect was observed. When exposed to CdTe-QDs in the first 48 h, the EC_{50} increased 76.57%, which implied a reduced toxicity. In the second step, the EC_{50} had no significant change from 72 to 120 h while declined rapidly to 0.23 mg•L^{-1} finally, indicating a latent high-toxic.

The growth inhibition of nanoparticles is related to their chemical composition. [31]For example, QDs usually release their core metals into water. [32,33]The dissolved metal ions are known to be toxic to aquatic organism even if at a relatively low concentration. The toxicity of CdTe-QDs is attributed to the core material, Cd. [34]Stepwise stress model (SSM) indicated that a serial sequence response of organisms was activated regularly by increased toxicant concentration or exposure time. In the present study, CdTe-QDs impacted the cyanobacteria in three steps. A declined inhibition in the first step implied the tolerance of *M. aeruginosa* when exposed to chemicals. However, stress gradually decreased the adaption as time passed until *M. aeruginosa* could not overcome the threshold, which caused the inhibition effect occurred again. In contrast, the PEG_{2000}-CQDs were synthesized from biological activated carbon, which was derived from the organic matrix such as twigs and peels. It suggests that cyanobacteria had a possible metabolic effect on PEG_{2000}-CQDs. The median effective concentrations (EC_{50}) for PEG_{2000}-CQDs and CdTe-QDs, respectively, resulted in a 50% reduction in the growth rate of cyanobacteria within the given exposure time compared to the control. The EC_{50} values comparison suggested that CQDs had much less inhibition effect and no latent impact, compared to CdTe-QDs.

[32]Hormesis is the affected function, which is characterized as a response to toxicants from low-concentration stimulation to a high-concentration inhibition. [35]In the previous research, the population growth rate of *P. tricornutum* was stimulated when [QDs] $\leqslant$ 0.2 Nm. [36,37]This effect is also reported in vivo with CdSe/ZnS-QDs and other nanoparticles. However, the effects of CQDs on organism were less characterized. It's worth noting that the positive effects on the growth and photosynthesis of *M. aeruginosa* at 5 and 10 mg•L^{-1} was not significant (Fig.4 and Fig.5), whereas the soluble protein in cell was induced at the relatively low concentrations. The organism, not the chemical, is considered as the key factor in the hormesis. The impact or disruptions in homeostasis induced finally the organism to respond in different levels. Thus, this response can be considered a signal of cellular stress. We presume there could have the possible occurring of some interaction between cyanobacteria and CQDs, which should be considered deeply in future studies.

Conclusions

In this work, PEG_{2000}-CQDs, CA-CQDs and Gly-CQDs were prepared via facile chemical oxidation and one-step thermal pyrolysis routes, respectively. All of these CQDs exhibited excellent good water solubility, favourable photostability. Whereafter, serials of comparative tests were conducted in order to investigate the inhibitory effects of different CQDs and metal-based quantum dots on *S. aureus* and *M. aeruginosa*. In the part I, the effects of CA-CQDs and Gly-CQDs compared with CdSe-QDs were investigated on *S. aureus* by [38]the optical density method. Results showed that bacterial abundance was of positive relevance with inhibitory effect of CQDs as the concentration increase. A low concentration of Gly-CQDs was benefited to *S.aureus*. However, *S.aureus* was more susceptible to CdSe-QDs which even in low concentration can inhibit bacteria significantly. It was demonstrated the CA-CQDs and Gly-CQDs were much less toxic compared with CdSe-QDs. In the part II, the research was the first time to adopted *M. aeruginosa* to evaluate the potential environmental risks of PEG_{2000}-CQDs and CdTe-QDs. The growth, chlorophyll-a accumulation and the soluble protein contents of the phytoplankton were evaluated. In general, CdTe-QDs had a significantly inhibitory effect on the population growth and chlorophyll-a accumulation of *M. aeruginosa*, whereas CQDs had much less toxicity and latent impact than CdTe-QDs. In summary, any kind of carbon quantum dots has low ecological risks than metal-based QDs on *S. aureus* and *M. aeruginosa*, which may provide reference value to utilize carbon quantum dots better and safely. However, more experiments should be done to deepen the research in the future.

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