

## Green synthesis of water-soluble quantum dots

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### Abstract

Quantum dot (QDs) or colloidal semiconductor nanocrystals is a nanoscale particle of semiconducting materials that is made from metallic binary compounds. The intrinsic optical properties of QDs provide long-term stability and high intensity of fluorescence than traditional organic fluorescent dyes and ability of various targeting QDs to different biomolecules via surface modifications. Therefore, the QDs have desirable properties for applying in fluorescence-based assay especially for pathogenic bacteria detection which require high sensitivity, selectivity and fluorescence sensor stability. There are several approaches for QD synthesis such as using different precursors, solvents and temperature.

In this study, we focus on green synthesis of luminescent quantum dots, CdTe, in aqueous solution to avoid using less toxic solvent with shorter reaction time and lower temperature (90°C) when is compared with traditional processes (over 200°C). In addition, this method used mercaptosuccinic acid (MSA) as a stabilizing agent that provided carboxylic functional group on the particles surface in situ resulting in highly water soluble quantum dots (MSA-capped QDs). Physical properties of the obtained nanoparticles were characterized by size, zeta potential, fluorescence spectrum, quantum yield and Transmission Electron Microscopy image (TEM image). The results showed narrow fluorescent spectrum (emission peak at 547 nm) and wide range of excitation (absorbance curve shift at 530 nm). This technique was reproducible and allowed particles to stabilize in aqueous solution with zeta potential of  $-22.6 \pm 7.3$  mV and average size around  $72.76 \pm 13.92$  nm. The advantage of this approach relied on its active site on the surface (carboxylic group) which could be easily modified by various biomolecules binding for biomedical diagnosis.

### Introduction

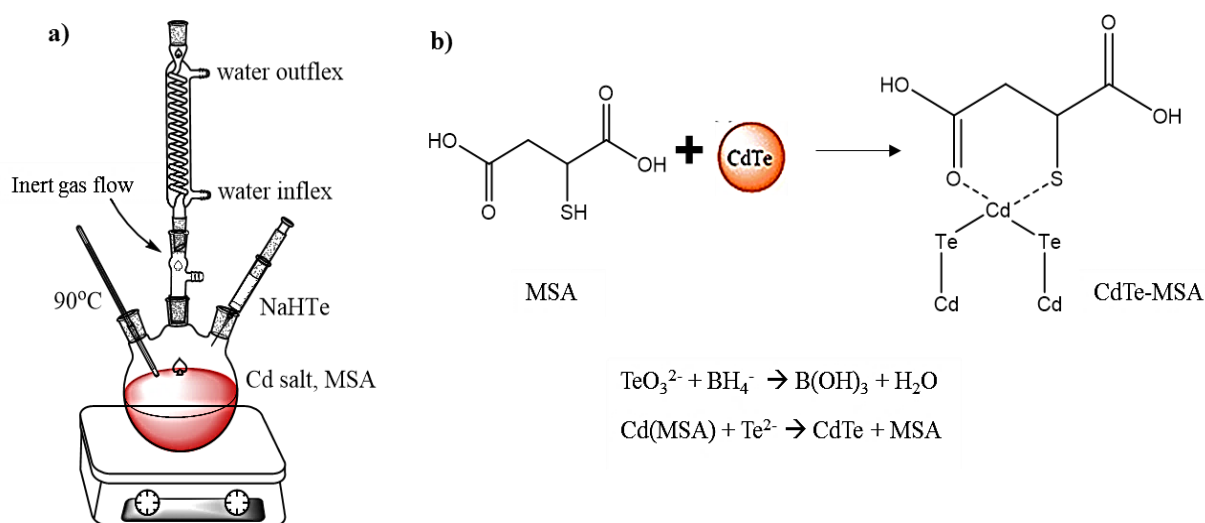
Quantum dots or colloidal semiconductor nanocrystals are nanoscale particles of semiconductor made from metallic compounds such as Cd, Pb, Te, or Se.<sup>1</sup> These particles have different intrinsic optical and electronic properties depending on particle size, therefore, various fluorescence spectrum could be observed from the same materials.<sup>2</sup> QDs have been used for bio-labeling and sensor applications because they provide high fluorescence intensity, long-term photostability and easy surface modifications which can increase multifunctional efficiency and diagnostic tools in many biomedical applications.<sup>3,4,5</sup>

High quality QDs could be synthesized using organic solvent, however, there are drawbacks of this method such as using high temperature (over 200 °C), inert atmosphere and toxic organic solvents e.g. dimethylcadmium and trioctylphosphine oxide.<sup>1</sup> In addition, the obtained QDs from organic based method have hydrophobic surface resulting in additional ligand exchange step to produce water soluble QDs which are necessary for biomedical

applications. Therefore the new approach for synthesis QDs has been introduced which is faster, easier and safer called aqueous route or water phase system<sup>1</sup>.

The aqueous route for QDs synthesis is a favor method that improves hydrophilic property of the particles. The sizes and fluorescence properties are controlled by using stabilizing agents such as thiols. Salted semiconductor and stabilizer are directly set up in water through the addition of freshly prepared NaHX solution (where X is the chalcogenide elements).<sup>1, 3,6</sup> Therefore, this method could be accomplished in one simple step. Moreover, this method uses water instead of toxic organic solvent and low temperature (90 °C) which not only can reduce the generation of toxic waste to environment but also could produce the water soluble QDs without additional ligand exchange process. Therefore, this system is a simple, fast and eco-friendly approach for generation of metallic nanoparticles.<sup>7</sup>

In this study, water-soluble QDs have been synthesized using CdTe as core particles in aqueous route system and the solubility was enhanced with MSA as thiol stabilizer which provided carboxylic acid functional group. The summarized synthesis scheme is show in figure. 1



**Figure. 1.** Schematic illumination of QDs synthesis, a) Aqueous route system; CdTe QDs are synthesized by mixing cadmium salt and oxygen-free NaHTe solution through the presence of MSA using low temperature (90 °C) b) The equation of CdTe formation and chemical structure of MSA-capped CdTe QDs.

## Method

### *Water-soluble CdTe QDs synthesis*

CdTe QDs synthesis via aqueous route system was modified from previous protocol by Ebrahim, S., et al. (2015) and the ratio of Cd:Te:MSA was adjusted to 1:1.6:2.3, respectively. The reaction was done as described below.

### *Preparation of oxygen-free NaHTe solution*

NaHTe solution was freshly prepared under restricted oxygen-free condition by mixing NaBH<sub>4</sub> (80 mg) and Te powder (31 mg) in 10 mL of distilled water under N<sub>2</sub> at 60 °C. The mixture was stirred until the solution turn to pink color.

### *Synthesis of MSA-capped CdTe QDs*

MSA-Cd solution was prepared by mixing 0.0548 g of CdCl<sub>2</sub> and 0.083 g of MSA in 100 mL of distilled water. The previously prepared NaHTe solution was added into MSA-Cd solution and the mixture was stirred at 90 °C at 450 rpm for 1 hour under the water-phase reflux system. The obtained QDs were washed with ethanol to remove excess reactants before re-suspending in distilled water. The aqueous MSA-capped CdTe QDs were kept at 4 °C under dark condition.

### *Characterization of QDs particles*

Following the synthesis synthesized, the obtained MSA-capped CdTe QDs were characterized as described below.

### *Absorption and fluorescence determination*

The absorption spectra of the QDs were determined using a UV-visible spectrophotometer (Agilent 8453 UV-visible spectroscopy) with ranging wavelength between 400 to 700 nm. The wavelength at first absorption peak ( $\lambda$ ) was used to calculate the average diameter (D) of particles using the equation reported by Yu, W.W., et al. (2003) as shown below.<sup>8</sup> Moreover, the absorbance value can be used to quantify the concentration of particles with Lambert-Beer's law after determining the extinction coefficient ( $\epsilon$ ) in equation (2). The fluorescence spectra were measured using a spectrofluorometer (Cary Eclipse fluorescence spectrophotometer) to record the emission spectrum (nm) with the excitation wavelength at 350 nm.

$$D \text{ (nm)} = (9.8127 \times 10^{-7})\lambda^3 - (1.7147 \times 10^{-3})\lambda^2 + (1.0064)\lambda - (194.84) \dots (1)$$

$$\epsilon = 10,043 \times 2.12D, D \text{ is size in nm from equation (1)} \dots (2) \text{ and}$$

$$A = \epsilon bC,$$

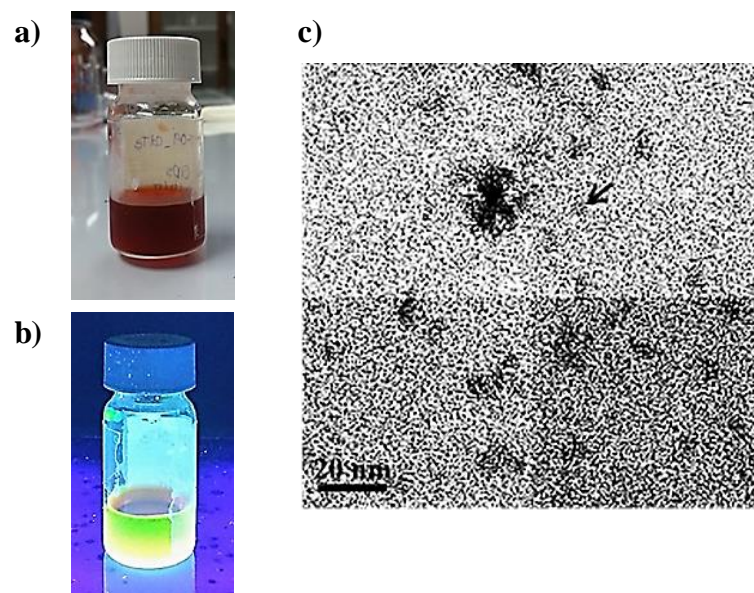
Where A is the absorbance value at the wavelength at first absorption peak, b is the path length (cm) and C is the concentration in molarity (mole/L).

### *Size, zeta potential measurement and Transmission Electron Microscopy (TEM)*

Size and zeta potential of particles were measured using Dynamic light scattering (DLS) technique (Malvern Zeta sizer Nano ZS). Each experiment was performed in triplicate. The size of QDs were confirmed using TEM (Fei Tecnai T20 G<sup>2</sup>).

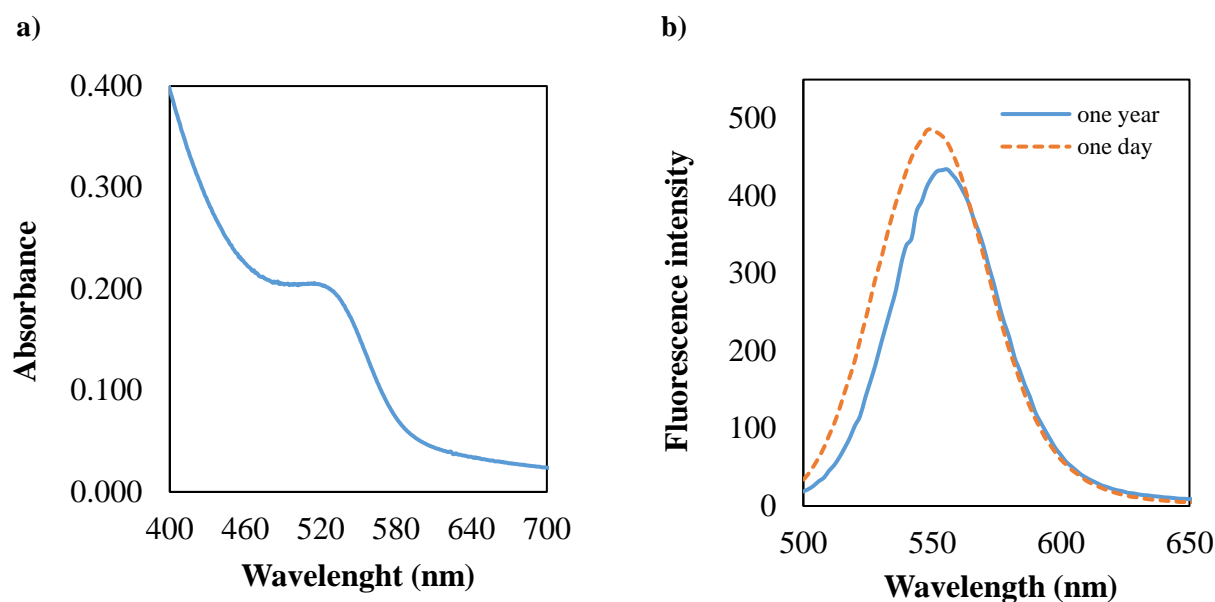
## **Results and Discussion**

Synthesis of CdTe QDs using aqueous route approach to obtain water soluble CdTe QDs in one step was performed as illustrated in Figure 1. The results showed that the developed method was simple and successfully achieved water soluble QDs with higher fluorescence (Figure 2). The clear red wine solution of QDs was observed under ambient light and provide strong green fluorescence under exposure with UV light (Figure 2a and 2b). Morphology of particles observed using TEM image showed small spherical particles with average size less than 5 nm (Figure 2c).



**Figure 2.** Illuminations of water-soluble QDs, a) MSA-capped CdTe QDs at ambient condition b) MSA-capped CdTe QDs under UV-excitation c) TEM image of MSA-capped CdTe QDs. Scale bar represents in 20 nm and black arrow indicates the individual particle.

Optical properties of the QDs were determined using spectrophotometer (Figure 3). This MSA-capped QDs had broad absorption spectrum with the first absorption peak at 530 nm (Figure 3a). This value was used to calculate concentration and approximate the size of CdTe QDs using the equation provided in the method section. The concentration was calculated to be 39.3  $\mu\text{M}$ . The approximated core size of CdTe from equation was found to be around 2.98 nm which correlated with the size from TEM image. The fluorescence spectrum of this obtained QDs could be observed the maximum emission wavelength at 554 nm (Figure 3b, dashed line) which also showed the narrow emission spectrum (full width at half-maximum (FWHM) was around 40 nm)<sup>9</sup>. Moreover, the stability of fluorescence intensity has been investigated and result showed that fluorescence intensity was slightly decreased after one year storage (figure 3b, solid line). This result indicated that the developed water-phase system can provide the fluorescence CdTe QDs with long term photostability.



**Figure 3.** Optical spectra of MSA capped CdTe QDs, a) Absorption spectrum b) Fluorescence spectra, dashed line and solid line represent measured data after one day and one year, respectively.

In order to use the CdTe QDs for biological applications, the hydrodynamic size and stability of QDs in water are necessary to be evaluated. The hydrodynamic size was observed using DLS technique. The obtained size was around  $72.76 \pm 13.92$  nm which was different from the previous calculation since the calculation focused only on the size of core CdTe but did not include the size that the capping reagent might contribute to. Whereas, the hydrodynamic size from DLS provide the information of the whole MSA-capped CdTe QDs in water, therefore showing larger size. In addition, the QDs showed polydispersity index (PDI) value around 0.2 indicating that the particles were monodispersed (Table 1). Zeta potential was also confirmed the successful coating of CdTe with the negatively-charged surface coating of MSA and high colloidal stability of the particles with the value of  $-22.6 \pm 7.3$  mV.

**Table 1.** The average size and zeta potential of the particles in aqueous solution

<b>MSA-capped CdTe QDs</b>	
Size (d.nm $\pm$ S.D.)	$72.76 \pm 13.92$
PDI $\pm$ S.D.	$0.219 \pm 0.031$
Zeta potential (mV $\pm$ S.D.)	$-22.6 \pm 7.3$

### Conclusion

The water soluble and highly fluorescent MSA-capped CdTe QDs was successfully obtained from this developed green synthesis process using one step in aqueous phase with low temperature (less than 100 °C). The obtained QDs were monodispersed, stable in water with high and stable fluorescence up to one year. This MSA-capped QDs also provide available carboxylic surface functionality which can be easily further modified with other biomolecules to increase specificity and apply for various bio-labelling or biosensor applications.

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