

Cite this: *Nanoscale*, 2012, **4**, 4103

www.rsc.org/nanoscale

## Sonochemical synthesis of highly fluorescent glutathione-stabilized Ag nanoclusters and S<sup>2-</sup> sensing†

Tingyao Zhou,<sup>a</sup> Mingcong Rong,<sup>a</sup> Zhimin Cai,<sup>a</sup> Chaoyong James Yang<sup>a</sup> and Xi Chen<sup>\*ab</sup>

Received 24th March 2012, Accepted 3rd May 2012

DOI: 10.1039/c2nr30718a

**A facile one-pot sonochemical approach is presented to prepare highly blue-emitting Ag nanoclusters (AgNCs) using glutathione as a stabilizing agent in aqueous solution. The as-prepared AgNCs can be applied in the selective detection of S<sup>2-</sup> with a limit of detection of 2 nM based on fluorescence quenching.**

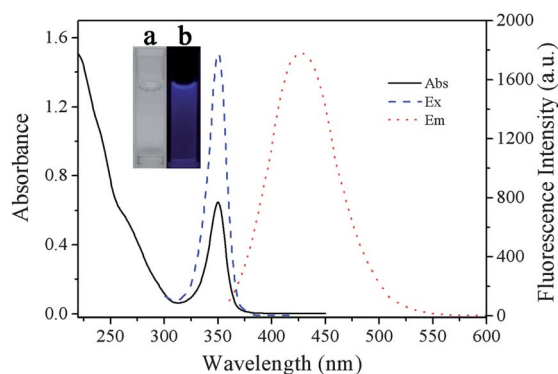
Silver nanostructures, due to their unique physical, electrical and optical properties, have attracted considerable attention in recent years for applications in catalysis, chemical sensing and bio-diagnostics.<sup>1</sup> For example, Ag nanoparticles exhibit localized surface plasmon resonance and are applied to the colorimetric detection of DNA,<sup>2</sup> metal ions,<sup>3</sup> proteins<sup>4</sup> and enzymatic activity.<sup>5</sup> In general, when the size of Ag nanoparticles decreases to the level of the electron Fermi wavelength (~0.5 nm for Ag), the plasmon absorption disappears completely and a molecule-like property of discrete electronic states is generated. These smaller Ag nanoparticles are referred to as Ag nanoclusters (AgNCs), and strong fluorescence can be obtained due to their molecule-like electronic states and a nonzero HOMO–LUMO gap.<sup>6</sup> However, AgNCs are generally not stable and easily aggregate to form larger nanoparticles in aqueous solution.

To avoid this aggregation, several stabilizers are applied as scaffolds for AgNCs, such as dendrimers,<sup>7</sup> polyelectrolytes,<sup>8</sup> DNA,<sup>9</sup> proteins,<sup>10</sup> as well as small molecules containing carboxylic groups or thiols.<sup>11</sup> Water-soluble highly fluorescent AgNCs have been prepared using several methods including radiolytic,<sup>12</sup> chemical reduction<sup>9,10,11c,d</sup> and photochemical (using UV light) approaches.<sup>8</sup> For example, Dickson and co-workers report highly fluorescent AgNCs with the emission colour being tuneable *via* a DNA sequence using sodium borohydride as a reducing agent.<sup>9c</sup> A variety of AgNCs are also prepared by reduction with sodium borohydride using different thiolates, such as 2,3-mercaptosuccinic acid,<sup>11a</sup> bovine serum albumin<sup>10c,e</sup> or dihydrolipoic acid<sup>11c</sup> as scaffolds. The production of fluorescent AgNCs in polymer microgel hosts by UV irradiation has

also been reported.<sup>8a</sup> Chemical reduction and photochemical (using UV light) approaches are simple and effective methods to prepare fluorescent AgNCs, however, they use environmentally harmful UV irradiation or toxic sodium borohydride.

It is important, therefore, to develop environmentally friendly methods for the preparation of fluorescent AgNCs. Sonochemistry is a well-established method for the synthesis of various nano-materials.<sup>13</sup> Recently, Suslick's group reported a sonochemical synthesis method for fluorescent AgNCs in the presence of the capping agent polymethylacrylic acid.<sup>14</sup> However, Ar protection during preparation is necessary, and more than one emitter is obtained in the products.

Glutathione (GSH) is a natural tripeptide consisting of glutamic acid, cysteine and glycine, and is widely used as a stabilizing agent in the synthesis of fluorescent AuNCs<sup>15</sup> and AgNCs.<sup>16</sup> In our work we described a facile sonochemical route for the preparation of blue-emitting and well-dispersed AgNCs using GSH as a capping agent in aqueous solution. Using ultrasound radiation, the one-pot preparation of fluorescent AgNCs in the presence of GSH could be achieved. The as-prepared AgNCs were characterized by UV-vis absorption and fluorescence spectra, as shown in Fig. 1. From the UV-vis absorption spectrum, it can be observed that fluorescent AgNCs exhibited a sharp absorption peak at 350 nm and a weak absorption peak at 274 nm due to inter-band transitions between the ligand/d-band and the sp band,<sup>17</sup> but no characteristic surface plasmon band of larger Ag nanoparticles at around 380–500 nm. The fluorescence



**Fig. 1** UV-vis absorption spectrum and fluorescence spectra of fluorescent AgNCs. Inset: photographs of fluorescent AgNCs under (a) daylight and (b) UV light at 365 nm.

<sup>a</sup>Department of Chemistry and Key Laboratory of Analytical Sciences of Xiamen University, College of Chemistry and Chemical Engineering, Xiamen University, Xiamen 361005, China. E-mail: xichen@xmu.edu.cn; Fax: +86 592 2184530; Tel: +86 592 2184530

<sup>b</sup>State Key Laboratory of Marine Environmental Science, Xiamen University, Xiamen 361005, China

† Electronic supplementary information (ESI) available: Experimental procedures of fluorescent AgNCs synthesis and Fig. S1–S6. See DOI: 10.1039/c2nr30718a

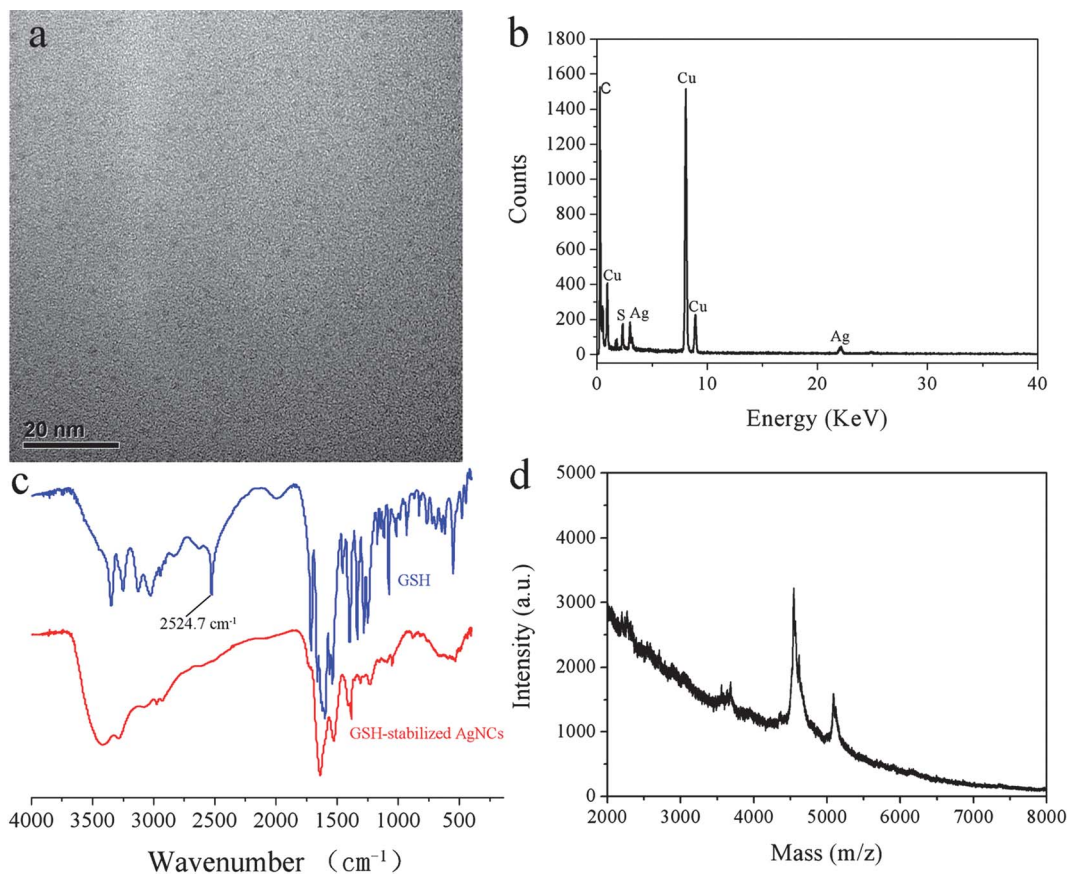
spectra show that an emission peak could be found at 430 nm and the maximum excitation wavelength was 350 nm. These optical properties of the as-prepared AgNCs are similar to the AgNCs obtained by Xie's group.<sup>16b</sup> As shown in the inset of Fig. 1, the solution of fluorescent AgNCs appeared colourless under daylight, while it emitted blue fluorescence under the UV light at 365 nm. The quantum yield of the fluorescent AgNCs was 1.9% calculated with quinine sulfate as a reference with a 350 nm excitation source.<sup>18</sup> Furthermore, the as-prepared AgNC solution was stable and had no obvious change (only a 13% decrease) in fluorescence intensity even after six weeks' storage without inert gas protection in the dark at 4 °C, as shown in Fig. S1 in the ESI.†

As shown in Fig. 2a, the transmission electron microscope (TEM) image shows that the fluorescent AgNCs were well-dispersed and uniform with an average size of 1.8 nm. An energy dispersive X-ray (EDX) spectrum (Fig. 2b) reveals the presence of Ag and S atoms. This result verified that the AgNCs were surrounded by the stabilizing agent GSH. Fourier transform infrared (FT-IR) spectra of GSH and GSH-stabilized fluorescent AgNCs are shown in Fig. 2c. The S–H stretching band (2524.7 cm<sup>-1</sup>) of GSH disappeared from the surface of the AgNCs, indicating the formation of covalent bonds between the GSH and AgNCs. The composition of fluorescent AgNCs was analyzed using matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) mass spectroscopy. As shown in Fig. 2d, a major peak at ~4545 Da could be assigned to the species [Ag<sub>12</sub>GSH<sub>10</sub> – 8H<sup>+</sup> + 8Na<sup>+</sup>] for the fluorescent AgNCs, which agrees

with an earlier report.<sup>16b</sup> A low-intensity peak at ~5093 Da might be assigned to the species [Ag<sub>15</sub>GSH<sub>11</sub>], which might be attributed to the by-product produced under ultrasonic irradiation.

The relationship between the fluorescence emission spectrum of the as-prepared AgNCs and the excitation wavelength was investigated. As shown in Fig. S2,† for an excitation wavelength between 320 nm and 370 nm, an emission peak at 430 nm with a maximum intensity could be observed at the excitation wavelength of 350 nm. The emission wavelength remained constant but the fluorescence intensity changed only with the increase of the excitation wavelength, indicating that the as-prepared fluorescent AgNCs contain only a single emitter.

The temporal evolution of the absorption spectra and the corresponding fluorescence emission spectra of the AgNCs under ultrasound radiation is shown in Fig. S3.† There were two broader absorption peaks at 350 nm and 274 nm, and a small emission could be observed before the ultrasonic irradiation. With increasing ultrasonic time up to 105 min, the absorption peak at 350 nm gradually became sharp and the peak at 274 nm gradually decreased, while the corresponding emission intensity gradually increased. After that, the absorption peak at 274 nm appeared again and the peak at 350 nm became broader. In addition, longer ultrasonic irradiation could reduce the emission intensity due to the generation of larger numbers of non-fluorescent AgNCs under longer ultrasonic irradiation times. The best ultrasonic irradiation time for the synthesis of fluorescent AgNCs was 105 min.



**Fig. 2** (a) TEM image and (b) EDX spectrum of the fluorescent AgNCs. (c) FT-IR spectra of free GSH and the GSH-stabilized AgNCs. (d) MALDI-TOF mass spectroscopy of the fluorescent AgNCs (positive-ion mode).

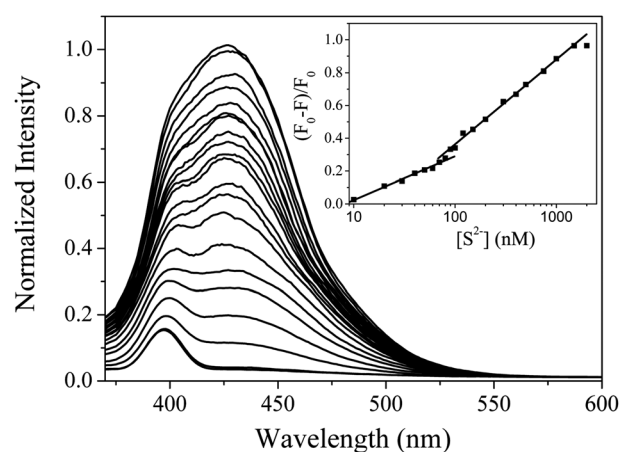
The influence of parameters such as the molar ratio of  $\text{Ag}^+$  to GSH and the pH value on the synthesis of fluorescent AgNCs was investigated. Fig. S4† shows the UV-vis absorption and fluorescence emission spectra of fluorescent AgNCs prepared with different ratios of  $\text{Ag}^+$  to GSH under the same  $\text{Ag}^+$  concentration and ultrasonic time. The optimal ratio was quite well-defined at 1 : 4, which can be considered either as the ratio of silver to GSH, corresponding to the chemical composition of the AgNCs, or at least as the ratio at which a possible excess of GSH is beneficial for the cluster formation and stability.<sup>11d</sup> The pH value, which was controlled by the addition of NaOH or  $\text{HNO}_3$  in the experiments, also played an important role in the synthesis of the fluorescent AgNCs. As shown in Fig. S5,† the maximum emission and absorption intensities at 350 nm could be synchronously obtained when the initial pH was about 5.0. Higher or lower pH values caused the emission intensity to decrease due to the cluster formation change.

Based on a previous report,<sup>19</sup> a possible mechanism was proposed for the preparation of fluorescent AgNCs. Firstly,  $\text{Ag}^+$  reacted with GSH to form a kind of silver thiolate complex, then ultrasonic irradiation destroyed the C–S bond of the silver thiolate complex and released a radical R. After the radical R reduced  $\text{Ag}^+$  to  $\text{Ag}^0$ , AgNCs were generated and kept stable in the presence of excess GSH, which was taken as a capping agent.

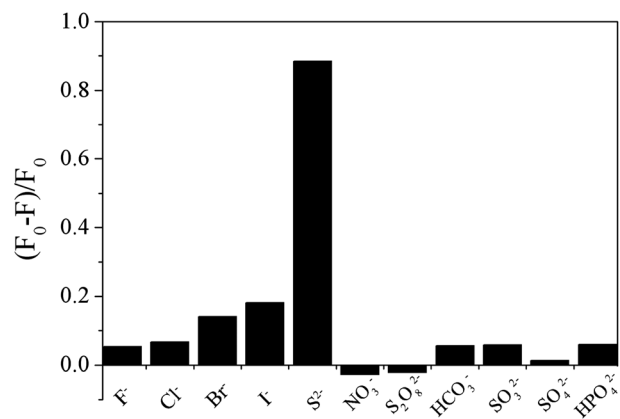
$\text{S}^{2-}$  is an important element in environmental monitoring due to its wide distribution in natural water and wastewater.<sup>20</sup> Note that the solubility product constant of  $\text{Ag}_2\text{S}$  (ref. 21) and the stability constant of silver thiolate<sup>22</sup> are  $6.3 \times 10^{-50}$  and  $1 \times 10^{12.3} \text{ M}^{-1}$ , respectively. As a result of the formation of  $\text{Ag}_2\text{S}$  (ref. 23), the as-prepared AgNCs could be considered in the determination of  $\text{S}^{2-}$  concentration using their fluorescence quenching. The response mechanism of fluorescent AgNCs towards  $\text{S}^{2-}$  was investigated. Fig. S6† shows that the solution of the Ag nanoclusters gradually changed to yellow in colour as the  $\text{S}^{2-}$  concentration increased, indicating the generation of  $\text{Ag}_2\text{S}$  formation or large Ag nanoparticles. From the UV-vis absorption spectra, it could be observed that a broad absorbance band for the  $\text{Ag}_2\text{S}$  particles appeared as the  $\text{S}^{2-}$  concentration increased, and no characteristic surface plasmon band for large Ag nanoparticles was seen, confirming the existence of  $\text{Ag}_2\text{S}$  formation.<sup>24</sup> To evaluate the sensitivity of this sensing system, different concentrations of  $\text{S}^{2-}$  (0.01–2.0  $\mu\text{M}$ ) were added to the solution containing the as-prepared AgNCs. As shown in Fig. 3, the fluorescence intensity of the AgNCs gradually decreased as the concentration of  $\text{S}^{2-}$  increased. Two linear correlations existed between the value of  $(F_0 - F)/F_0$  and the  $\text{S}^{2-}$  concentration in the logarithmic scale, 0.01–0.09  $\mu\text{M}$  and 0.1–1.5  $\mu\text{M}$ . The two linear equations were as follows:  $(F_0 - F)/F_0 = 0.2667 \log[\text{S}^{2-}] - 0.2434$  and  $(F_0 - F)/F_0 = 0.5159 \log[\text{S}^{2-}] - 0.6687$ . The detection limit at a signal to noise ratio of 3 for  $\text{S}^{2-}$  was found to be 2 nM, which was much lower than the maximum level (15  $\mu\text{M}$ ) of  $\text{H}_2\text{S}$  in drinking water permitted by the World Health Organization.<sup>25</sup>

To evaluate the selectivity of fluorescent AgNCs towards  $\text{S}^{2-}$ , a series of anions were investigated under similar conditions for the detection of  $\text{S}^{2-}$ , such as  $\text{F}^-$ ,  $\text{Cl}^-$ ,  $\text{Br}^-$ ,  $\text{I}^-$ ,  $\text{NO}_3^-$ ,  $\text{S}_2\text{O}_8^{2-}$ ,  $\text{HCO}_3^-$ ,  $\text{SO}_3^{2-}$ ,  $\text{SO}_4^{2-}$  and  $\text{HPO}_4^{2-}$ . As shown in Fig. 4,  $\text{S}^{2-}$  showed a significant quenching intensity of the AgNCs, while other anions presented only a slight quenching effect. These results revealed that the AgNCs were highly selective towards  $\text{S}^{2-}$ .

In conclusion, we have presented a facile one-pot sonochemical synthesis of blue-emitting AgNCs using GSH as a capping agent in aqueous solution. The results reveal that it is a rapid and



**Fig. 3** Emission spectra of fluorescent AgNCs in the presence of various concentrations of  $\text{S}^{2-}$  at a pH of 5.0 (from top: 0, 0.01, 0.02, 0.03, 0.04, 0.05, 0.06, 0.07, 0.08, 0.09, 0.10, 0.12, 0.15, 0.20, 0.30, 0.40, 0.50, 0.75, 1.0, 1.5 and 2.0  $\mu\text{M}$ ). Inset: relationship of relative fluorescence intensity with varying  $\text{S}^{2-}$  concentration.



**Fig. 4** Selectivity of fluorescent AgNCs towards  $\text{S}^{2-}$  over other anions at a pH of 5.0. The concentration of each anion was 1.0  $\mu\text{M}$ .

environmentally friendly method for preparing fluorescent AgNCs. The AgNCs consist of 12 silver atoms and the experimental conditions have been optimized to derive AgNCs with high emission. The AgNCs exhibit excellent water solubility and good dispersion, as well as high stability, and have an excellent sensitivity and high selectivity towards  $\text{S}^{2-}$ , with a detection limit of 2 nM.

## Acknowledgements

This research work was financially supported by National Nature Scientific Foundation of China (No. 20975085 and 21175112), the National Basic Research Program of China (2010CB732402) and the NFFTBS (No. J1030415), which are gratefully acknowledged. Furthermore, we would like to extend our thanks to Professor John Hodgkiss of The University of Hong Kong for his assistance with English.

## Notes and references

- (a) N. L. Rosi and C. A. Mirkin, *Chem. Rev.*, 2005, **105**, 1547; (b) G. V. Hartland, *Chem. Rev.*, 2011, **111**, 3858; (c) N. J. Halas,

- S. Lal, W.-S. Chang, S. Link and P. Nordlander, *Chem. Rev.*, 2011, **111**, 3913.
- 2 S. Liu, Z. Zhang and M. Han, *Anal. Chem.*, 2005, **77**, 2595.
- 3 K. Yoosaf, B. I. Ipe, C. H. Suresh and K. G. Thomas, *J. Phys. Chem. C*, 2007, **111**, 12839.
- 4 C. L. Schofield, A. H. Haines, R. A. Field and D. A. Russell, *Langmuir*, 2006, **22**, 6707.
- 5 H. Wei, C. Chen, B. Han and E. Wang, *Anal. Chem.*, 2008, **80**, 7051.
- 6 C. M. Aikens, *J. Phys. Chem. Lett.*, 2011, **2**, 99.
- 7 J. Zheng and R. M. Dickson, *J. Am. Chem. Soc.*, 2002, **124**, 13982.
- 8 (a) J. G. Zhang, S. Q. Xu and E. Kumacheva, *Adv. Mater.*, 2005, **17**, 2336; (b) L. Shang and S. J. Dong, *Chem. Commun.*, 2008, 1088; (c) Z. Shen, H. W. Duan and H. Frey, *Adv. Mater.*, 2007, **19**, 349.
- 9 (a) J. T. Petty, J. Zheng, N. V. Hud and R. M. Dickson, *J. Am. Chem. Soc.*, 2004, **126**, 5207; (b) C. M. Ritchie, K. R. Johnsen, J. R. Kiser, Y. Antoku, R. M. Dickson and J. T. Petty, *J. Phys. Chem. C*, 2007, **111**, 175; (c) C. I. Richards, S. Choi, J. C. Hsiang, Y. Antoku, T. Vosch, A. Bongiorno, Y. L. Tzeng and R. M. Dickson, *J. Am. Chem. Soc.*, 2008, **130**, 5038; (d) P. R. O'Neill, L. R. Velazquez, D. G. Dunn, E. G. Gwinn and D. K. Fyngenson, *J. Phys. Chem. C*, 2009, **113**, 4229.
- 10 (a) J. Yu, S. A. Patel and R. M. Dickson, *Angew. Chem., Int. Ed.*, 2007, **46**, 2028; (b) L. Peyser-Capadona, J. Zheng, J. I. Gonzalez, T. H. Lee, S. A. Patel and R. M. Dickson, *Phys. Rev. Lett.*, 2005, **94**, 058301; (c) C. Guo and J. Irudayaraj, *Anal. Chem.*, 2011, **83**, 2883; (d) S. S. Narayanan and S. K. Pal, *J. Phys. Chem. C*, 2008, **112**, 4874; (e) A. Mathew, P. R. Sajanlal and T. Pradeep, *J. Mater. Chem.*, 2011, **21**, 11205.
- 11 (a) Z. K. Wu, E. Lanni, W. Q. Chen, M. E. Bier, D. Ly and R. C. Jin, *J. Am. Chem. Soc.*, 2009, **131**, 16672; (b) T. Udaya Bhaskara Rao and T. Pradeep, *Angew. Chem., Int. Ed.*, 2010, **49**, 3925; (c) B. Adhikari and A. Banerjee, *Chem. Mater.*, 2010, **22**, 4364; (d) N. Cathcart and V. Kitaev, *J. Phys. Chem. C*, 2010, **114**, 16010.
- 12 (a) M. Treguer, F. Rocco, G. Lelong, A. Le Nestour, T. Cardinal, A. Maali and B. Lounis, *Solid State Sci.*, 2005, **7**, 812; (b) W. T. Wu, Y. S. Wang, L. Shi, W. M. Pang, Q. R. Zhu, G. Y. Xu and F. Lu, *J. Phys. Chem. B*, 2006, **110**, 14702.
- 13 J. M. Patete, X. H. Peng, C. Koenigsman, Y. Xu, B. Karn and S. S. Wong, *Green Chem.*, 2011, **13**, 482.
- 14 H. Xu and K. S. Suslick, *ACS Nano*, 2010, **4**, 3209.
- 15 (a) Y. Negishi, Y. Takasugi, S. Sato, H. Yao, K. Kimura and T. Tsukuda, *J. Am. Chem. Soc.*, 2004, **126**, 6518; (b) Y. Negishi, K. Nobusada and T. Tsukuda, *J. Am. Chem. Soc.*, 2005, **127**, 5261; (c) W. B. Chen, X. J. Tu and X. Q. Guo, *Chem. Commun.*, 2009, 1736; (d) R. C. Jin, *Nanoscale*, 2010, **2**, 343.
- 16 (a) X. Yuan, Z. Luo, Q. Zhang, X. Zhang, Y. Zheng, J. Y. Lee and J. Xie, *ACS Nano*, 2011, **5**, 8800; (b) X. Yuan, T. J. Yeow, Q. Zhang, J. Y. Lee and J. Xie, *Nanoscale*, 2012, **4**, 1968.
- 17 (a) S. Kumar, M. D. Bolan and T. P. Bigioni, *J. Am. Chem. Soc.*, 2010, **132**, 13141; (b) O. M. Bakr, V. Amendola, C. M. Aikens, W. Wenseleers, R. Li, L. Dal Negro, G. C. Schatz and F. Stellacci, *Angew. Chem., Int. Ed.*, 2009, **48**, 5921.
- 18 A. N. Fletcher, *Photochem. Photobiol.*, 1969, **9**, 439.
- 19 G. W. Yang and H. L. Li, *Mater. Lett.*, 2008, **62**, 2189.
- 20 T. Bagarinao, *Aquat. Toxicol.*, 1992, **24**, 21.
- 21 D. V. Goia, *J. Mater. Chem.*, 2004, **14**, 451.
- 22 (a) N. W. H. Adams and J. R. Kramer, *Aquat. Geochem.*, 1999, **5**, 1; (b) J. Liu, D. A. Sonshine, S. Shervani and R. H. Hurt, *ACS Nano*, 2010, **4**, 6903.
- 23 W. Y. Chen, G. Y. Lan and H. T. Chang, *Anal. Chem.*, 2011, **83**, 9450.
- 24 L. Motte, F. Billoudet and M. Pileni, *J. Phys. Chem.*, 1995, **99**, 16425.
- 25 [http://www.who.int/water\\_sanitation\\_health/dwq/guidelines/en/](http://www.who.int/water_sanitation_health/dwq/guidelines/en/).