ORIGINAL PAPER

Folic acid-CdTe quantum dot conjugates and their applications for cancer cell targeting

Preethi Suriamoorthy · Xing Zhang · Guiyang Hao ·
Alan G. Joly · Surya Singh · Marius Hossu ·
Xiankai Sun · Wei Chen

Abstract In this study, we report the preparation, luminescence, and targeting properties of folic acid-CdTe quantum dot conjugates. Water-soluble CdTe quantum dots were synthesized and conjugated with folic acid using 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide-N-hydroxysuccinimide chemistry. The influence of folic acid on the luminescence properties of CdTe quantum dots was investigated, and no energy transfer between them was observed. To investigate the efficiency of folic acid-CdTe nanoconjugates for tumor targeting, pure CdTe quantum dots and folic acid-coated CdTe quantum dots were incubated with human nasopharyngeal epidermal carcinoma cell line with positive expressing folic acid receptors (KB cells) and lung cancer cells without expression of folic acid receptors (A549 cells). For the cancer cells with positive folate receptors (KB cells), the uptake for CdTe quantum dots is very low, but for folic acid-CdTe nanoconjugates, the uptake is very high. For the lung cancer cells without folate receptors (A549 cells), the uptake for folic acid-CdTe nanoconjugates is also very low. The results indicate that folic acid is an effective targeting molecule for tumor cells with overexpressed folate receptors.

Keywords Quantum dots · Luminescence · Cancer · Folic acid receptors · Bioconjugation · Imaging · Labeling · Targeting

P. Suriamoorthy · X. Zhang · M. Hossu · W. Chen (✉)
Department of Physics, The University of Texas at Arlington,
Arlington, TX 76019-0059, USA
e-mail: weichen@uta.edu

G. Hao · X. Sun
Department of Radiology,
The University of Texas Southwestern Medical Center,
Dallas, TX, USA

X. Sun
Advanced Imaging Research Center,
The University of Texas Southwestern Medical Center,
Dallas, TX, USA

A. G. Joly
Pacific Northwest National Laboratory,
Richland, WA 99352, USA

S. Singh
Department of Integrative Biology, University of California,
Berkeley, CA 94720, USA

1 Introduction

Cancer is the second major cause of death in the USA, killing approximately half a million people in the USA alone every year (ACS 2009). Early detection and effective treatment provide the best hope for cancer patients. For example, early detection of cancer can avoid approximately 3–35% of cancer deaths (Colditz and Sellers 2006). Therefore, there is a crucial need to develop cancer-specific imaging probes for the early diagnosis of cancer. Cells are almost transparent to visible light thereby making their direct observation using a conventional microscope a challenge. Cells therefore need to be labeled with a fluorophore to enhance the contrast, thereby making imaging a simpler task. The major limitations in using fluorophores are the phenomena of photobleaching and blinking (Morgan et al. 2005), rendering them poor contrasting agents. To overcome these limitations, semiconductor nanocrystals, also known as quantum dots (QDs), have been extensively studied for the last decade. Some of the advantages of quantum dots over fluorescent probes are their efficient fluorescence, chemical stability, broad excitation bands, narrow emission bands, good water

solubility, compatible surface chemistry, high photostability, and low photo-bleaching rate (Morgan et al. 2005; Chen 2008). Even though Cd^{2+} containing quantum dots have high cytotoxicity (Zhang et al. 2006) that limits their practical application, in cell imaging they have tremendous potential as sensitive, nano-scale probes for early detection of cancer (Morgan et al. 2005; Chen 2008; Chen et al. 2006a; Smith et al. 2006). The applications of quantum dots for cell imaging and animal model studies can not only obtain insightful information for cancer detection but also for drug delivery and targeting (Chen and Sun 2008). Tumor targeting may be accomplished by using a tumor-specific ligand, such as folic acid. Folates are low molecular weight pterin-based vitamins required by eukaryotic cells for one-carbon metabolism and de novo nucleotide synthesis. The folate receptor is a glycosylphosphatidylinositol-anchored, high-affinity membrane folate-binding protein that is overexpressed in a wide variety of human tumors, including more than 90% of ovarian carcinomas (Sudimack and Lee 2000; Wang and Low 1998). On the other hand, normal tissue distribution of the folate receptor is highly restricted, making it a useful marker for targeted drug delivery to tumors. This methodology is currently being used for the selective delivery of imaging and therapeutic agents to tumor tissues (Leamon and Low 2001). Folic acid, a high-affinity ligand for the folate receptor (K_d=~10^{-10} M), retains its receptor binding property when covalently derivatized by its gamma-carboxyl group. Studies have shown that folate conjugates are taken into receptor-bearing tumor cells via folate receptor-mediated endocytosis (Antony 1996). Folic acid is potentially superior to antibodies as a targeting ligand because of its small size; lack of immunogenicity; ready availability; and simple, well-defined conjugation chemistry (Wang and Low 1998).

The conjugation of folic acid to quantum dots or nano-particles has been investigated and reported by several groups (Bharali et al. 2005; Yang et al. 2009; Hu et al. 2009; Manzoor et al. 2009). However, several questions regarding the application of quantum dot-folic acid conjugates for tumor targeting remain to be answered. For example, the mechanism responsible for quantum dot luminescence quenching is not yet clear. In addition, the targeting efficacy of folic acid-quantum dots to folate receptor positive (FR+) and folate-negative (FR−) tumors also needs to be identified. The present research is aimed to answer these questions.

2 Materials and methods

Materials Cadmium perchlorate hydrate ($Cd(ClO_4)_2.H_2O$), thioglycolic acid (TGA), aluminium telluride (Al_2Te_3), sulfuric acid (H_2SO_4), sodium hydroxide (NaOH), folic acid (FA), N-hydroxysuccinimide (NHS), 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC), Eagle's minimum essential medium (EMEM), F-12 K medium, fetal bovine serum (FBS; 10%), sterilized phosphate buffered saline (PBS), and Trypsin were purchased from Aldrich. Human nasopharyngeal epidermal carcinoma cell line - KB cells (ATCC number CCL-17) and human lung carcinoma cell line A549 cells (ATCC number CCL-185) were purchased from ATCC.

Synthesis Thioglycolic acid-stabilized cadmium telluride quantum dots were synthesized by a method described in the literature (Gaponik et al. 2002; Joly et al. 2005; Liu et al. 2006). Briefly, 1.463 g (4.70 mmol) of $Cd(ClO_4)_2.H_2O$ was dissolved in 125 ml of deionized (DI) water, and 0.793 ml (11.4 mmol) of thioglyocolic acid was added to the above solution under stirring. The pH of the solution was adjusted to 11.5 by dropwise addition of 0.5 M NaOH. The solution was transferred to a three-necked flask and de-gassed by bubbling Ar gas for ~10 min; 0.4 g of Al_2Te_3 was charged into a small three-necked flask. H_2Te_3 gas generated by the addition of 3 ml of 0.5 M H_2SO_4 to Al_2Te_3, was bubbled through the solution for ~5 min. The solution turned orange in color due to the formation of the CdTe precursors. The CdTe precursor solution was refluxed at 100°C, under open-air conditions, with condenser attached to promote the growth of nanocrystals. Folic acid was conjugated to TGA-coated CdTe quantum dots using EDC-NHS chemistry (Hermanson 1996; Wang et al. 2002); 0.05 M EDC-NHS (EDC/NHS=1:10) and 0.05 M QDs were mixed for 5 min. Then an equal molar ratio of 0.05 M folic acid was added to the above solution and stirred gently at room temperature over night. The unreacted chemicals (EDC, NHS, folic acid, and QDs) were removed by dialysis against pH adjusted (pH 11–12) de-ionized water for 1 day. The cut off molecular weight of the dialysis membrane was 12,000 Da.

Characterization The Fourier transform infrared (FTIR) spectra were measured on a Shimadzu RF-5301 attenuated total reflectance—FTIR spectrometer. The folic acid conjugation with QDs was monitored by a high performance liquid chromatography (HPLC) method performed on a Waters 600 Multisolvent Delivery System equipped with a Waters 2996 photodiode array detector and a Waters BioSuite 250 size-exclusion (SEC) column (300× 7.5 mm, 13 μm). The optical absorption spectra were recorded on a Shimadzu 1501 ultraviolet–visible (UV– VIS) spectrophotometer. The luminescence emission spectra were measured using a Shimadzu RF-5301PC fluorescence spectrophotometer. Luminescence lifetimes were collected using the frequency-doubled output of a synchronously pumped picosecond (ps) dye laser operating at 610 nm. The doubled output was focused onto the samples and emission collected at right angle

to the input. The emission was spectrally filtered, and the lifetime measured using time-correlated single photon counting. The instrument resolution was determined to be about 50 ps FWHM using a standard scattering material.

Cell culture and fluorescence imaging Human nasopharyngeal epidermal carcinoma cell line—KB cells (ATCC number CCL-17) and human lung carcinoma cell line A549 cells (ATCC number CCL-185) were cultured in EMEM and F-12 K medium respectively, supplemented with 10% FBS at 37°C (5% CO_2). The cellular uptake of the folic acid-conjugated quantum dots was studied as described by (Bharali et al. 2005). Briefly, glass cover slips were sterilized in ethanol for 1 h and air dried. The sterilized cover slips were placed in 6 well plates. Then, the KB and A549 cells were trypsinized and re-suspended in their corresponding growth media at a concentration of around 7.5×10^5 mL^{-1}; 60 μl of the cell suspension was placed on the cover slip and 2 ml of corresponding complete medium was added. The 6 well plates were incubated at 37°C with 5% CO_2 for 24 h. After 24 h of incubation, the media was removed and the cells were rinsed with PBS, and 2 ml of corresponding fresh media was added to the wells. Finally, 150 μl of quantum dots were added to one set of wells and 150-μl-conjugated quantum dots were added to the other sets of wells, and mixed properly. The plates were returned to the incubator. After 2 h, 4 h, and 8 h of incubation the plates were taken out of the incubator and rinsed several times with sterile PBS. The cover slips were carefully taken out of the wells, placed on glass slides, and covered with another sterile cover slip for fluorescence imaging observation which was conducted on a fluorescence imaging system with a Meiji Trinocular Fluorescent Microscope and a Pixera "Cooled" Digital Camera.

3 Results and discussion

3.1 Bioconjugation

In the conjugation, the addition of EDC and NHS to the QD solution resulted in the formation of a highly reactive intermediate (NHS-carboxylate) as reported by Bharali et al. (2005). This activated ester then reacted with the free amino group of folic acid, giving the nanoconjugates of folic acid with the CdTe quantum dots as illustrated in Fig. 1. FTIR and HPLC were used to confirm the successful conjugation of folic acid to CdTe quantum dots. Figure 2 represents the IR spectra of the CdTe QDs and CdTe-folic acid nanoconjugates. The FTIR spectrum of CdTe quantum dots shows no absorption band corresponding to the SH vibration at 2,570 cm^{-1}, illustrating that the thioglycolic molecules were anchored on the CdTe nanoparticles' surface through the sulfur atom in the mercapto group. (Gaponik et al. 2002) The FTIR spectrum of CdTe-folic acid nanoconjugates shows an OH stretching band at 3,368 cm^{-1} and an asymmetric CO_2 stretching band at 2,356 cm^{-1}. The amide bond can be characterized by the complex bands –$CO-NH_2$ and –CO-NH at 1,655 and 1,558 cm^{-1}, respectively. The –CO-NH mode contains contributions from –the C-N-stretching vibration and the –NH-bending vibration, which originates from bonding between the carboxyl groups of the TGA-capped QDs and the amide groups of the folic acid (Lei et al. 2008). The aromatic ring stretching of the pteridine ring and *p*-amino benzoic acid groups of the folic acid appear within the range of 1,476–1,695 cm^{-1} (Manzoor et al. 2009). The symmetric vibrations of the carboxylic group at 1,556 cm^{-1} and 1,377 cm^{-1} in the CdTe-folic acid nanoconjugate spectrum masks the bending vibrations of the NH group of the folic acid around the 1,500–1,600 cm^{-1} region (Zhang et al. 2002). Therefore, the IR
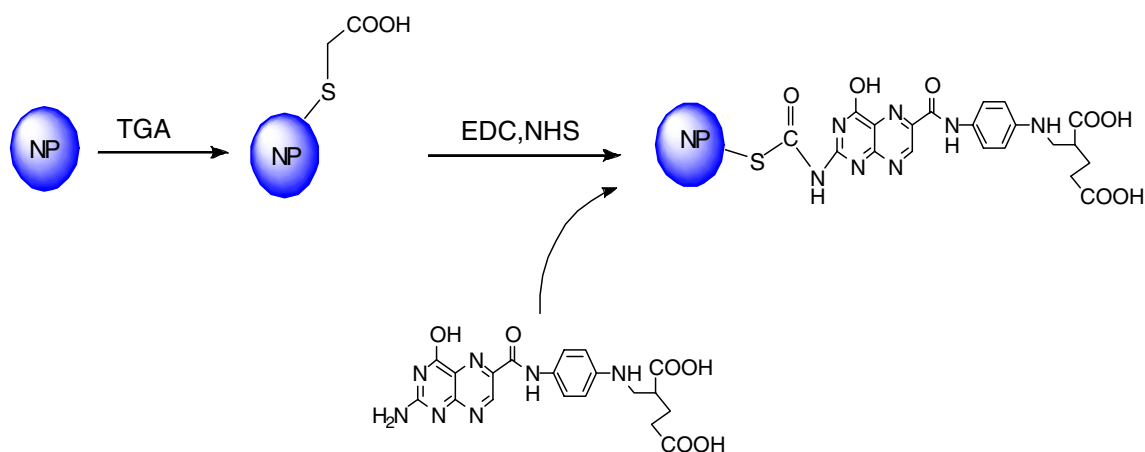

Fig. 1 An schematic illustration depicting the conjugation chemistry of folic acid to TGA-coated CdTe quatum dots via EDC-NHS reactions
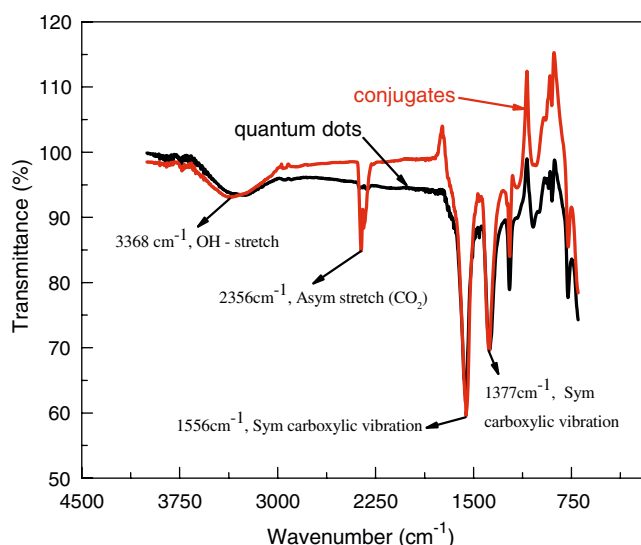
Fig. 2 FTIR spectra of CdTe quantum dots and CdTe quantum dots conjugated to folic acid

Fig. 4 Optical absorption spectrum of CdTe quantum dots (*red*) and emission spectrum of folic acid excited at 380 nm (*blue*)

spectrum demonstrates clearly that folic acid has been successfully conjugated to the CdTe quantum dots.

Eluted by 0.1 M phosphate buffer containing 0.15 M NaCl (pH 7.4) at the flow rate of 1.0 mL/min, the retention time of folic acid on the SEC-HPLC column was 12.7 min with the peak UV absorbance at 281.6 nm. Interestingly, the QDs exhibited very weak UV signals without characteristic absorption peaks. A fluorescence detector would be more appropriate for the QD detection (Wan et al. 2004). As expected, the folate-conjugated QDs showed a UV peak at 10.4 min with a UV–VIS spectrum in the range of 210 to 400 nm similar to that of folic acid, indicating the successful formation of folate-QD nanoconjugates.
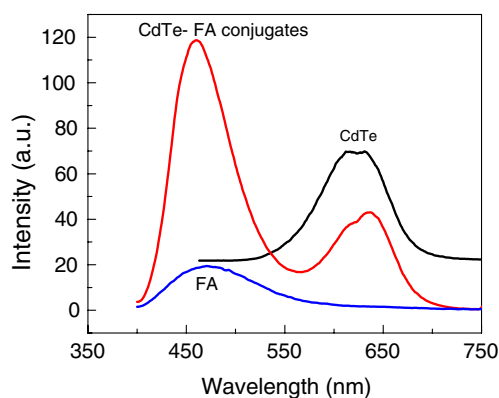
3.2 Luminescence properties

The luminescence spectra of CdTe quantum dots, folic acid, and CdTe-folic acid nanoconjugates are shown in Fig. 3. TGA-coated CdTe QDs have a narrow, symmetric emission peaking at 631 nm that has been reported and discussed extensively (Carbone et al. 2006; Chen et al. 2005; Chen et al. 2002; Chen et al. 2006b; Chou et al. 2006). Folic acid has an emission band peaking at around 458 nm which is the same as reported in the literature. (Tyagi and Penzkofer 2010; Thomas et al. 2002) The fluorescence spectrum of CdTe-folic acid nanoconjugates has two emissions at 460 and 630 nm, respectively. The 460 nm emission can be attributed to folic acid and the 630 nm emission to CdTe quantum dots.

It is noticed that the emission peaks of folic acid overlap with the absorption band of CdTe quantum dots as shown in Fig. 4. In principle, there could be energy transfer from

Fig. 3 Photoluminescence spectra of CdTe quantum dots (*black*), CdTe quantum dot-folic acid conjugates excited at 380 nm (*red*), and folic acid (*FA, blue*). The excitation wavelength is 380 nm for all measurements
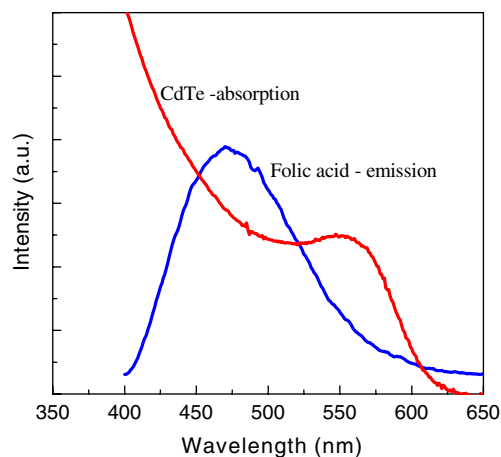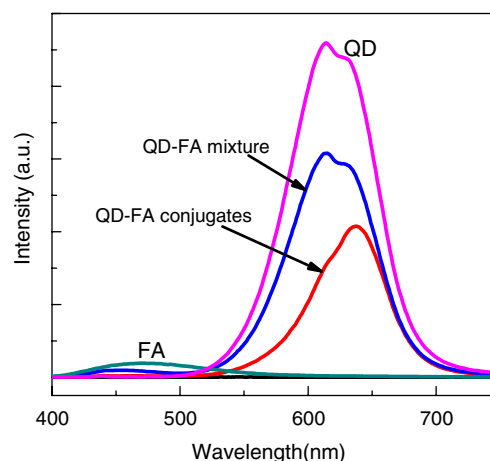

Fig. 5 Emission spectra of folic acid in water (*FA*), CdTe quantum dots (*QD*), CdTe-folic acid conjugates, and a CdTe-folic acid mixture (excitation at 380 nm)
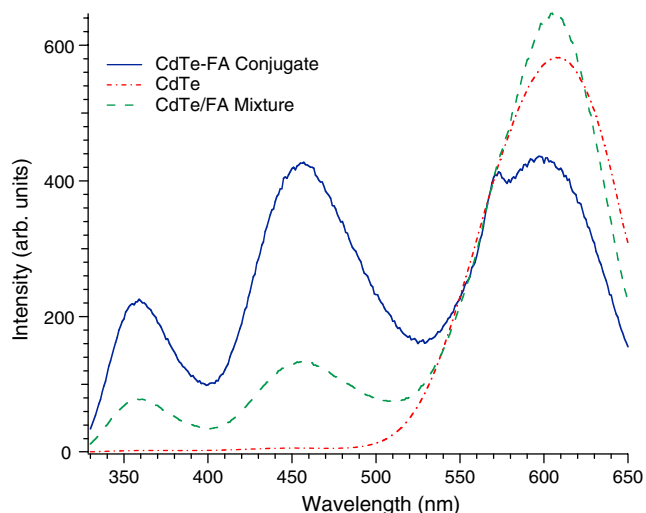
Fig. 6 Emission spectra of CdTe quantum dots, CdTe-folic acid conjugates, and CdTe-folic acid mixture (excitation at 285 nm)

Fig. 8 An schematic illustration of the energy transfer process from folic acid to CdTe quantum dots. η_{ET} is the energy transfer rate, τ_d is the emission decay rate of the donor (folic acid), and τ_a is the emission decay rate of the acceptors (CdTe quantum dots). In this process, if the lifetime of the donor is extremely short relative to the lifetime of the acceptor, no appreciable energy transfer occurs

folic acid to CdTe quantum dots in CdTe-folic acid nanoconjugates. Figures 5 and 6 display the emission spectra of CdTe quantum dots, CdTe-folic acid nano-conjugates and a CdTe-folic acid mixture excited at 380 (Fig. 5) and 285 nm (Fig. 6). In all the three samples, the concentration of CdTe quantum dots is the same. For excitation at 380 nm, the emission of CdTe quantum dots in both the CdTe-folic acid conjugates and mixture is weaker than in CdTe quantum dot solution. For excitation at 285 nm, folic acid shows two emissions at 360 and 450 nm, respectively. Both of the two emissions have been reported in the literature. (Tyagi and Penzkofer 2010) For excitation at 285 nm, the emission of CdTe quantum dots in the CdTe-folic acid mixture is slightly stronger than in CdTe quantum dot solution but the emission of CdTe quantum dots in the conjugates is still weaker than in CdTe

quantum dot solution. It was expected that the emission of CdTe quantum dots in CdTe-folic acid nanoconjugates would be enhanced as a result of energy transfer from folic acid. On the contrary, the luminescence of CdTe quantum dots in CdTe-folic acid nanoconjugates is quenched as compared with that of CdTe quantum dots. This indicates that energy transfer from folic acid to CdTe quantum dots likely does not happen in the nanoconjugates. To further investigate the possible energy transfer from folic acid to CdTe quantum dots, the luminescence lifetimes of folic acid at 450 nm were measured in folic acid solution and CdTe-folic acid nanoconjugates. As displayed in Fig. 7, the luminescence lifetimes from the two samples are almost identical and the lifetimes of folic acid in the conjugates and mixture are the same as reported by Tyagi and Penkofer (Tyagi and Penzkofer 2010). Energy transfer from the folic
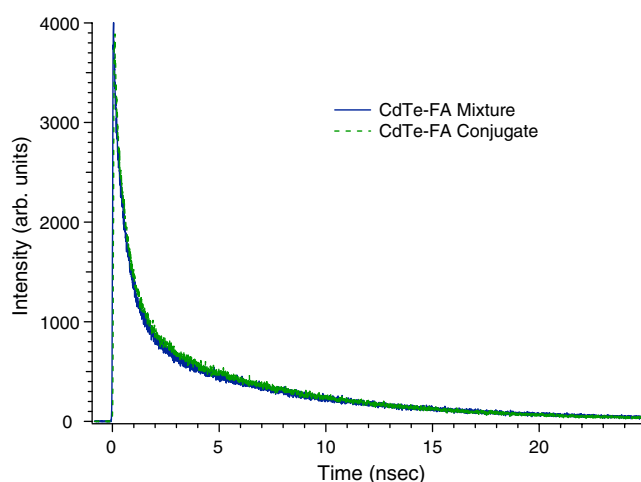

Fig. 7 Luminescence lifetimes at 450 nm following 285 nm excitation for the CdTe/folic acid mixture and conjugated sample. Both samples show near-identical lifetimes
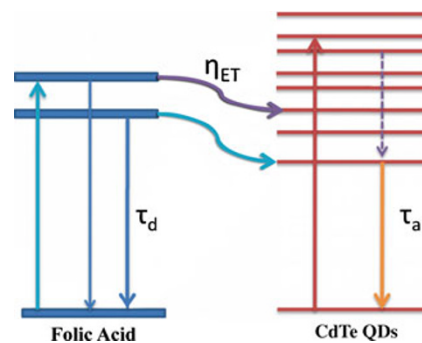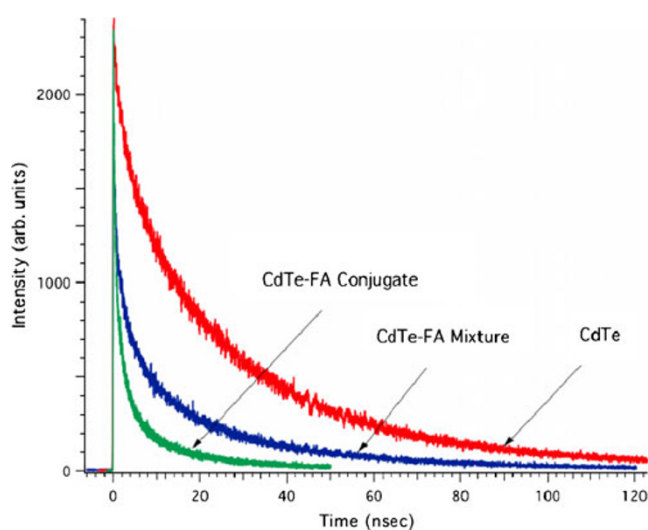

Fig. 9 Luminescence lifetimes at 600 nm following excitation at 285 nm for CdTe, a CdTe-FA conjugated sample, and a mixture of CdTe and folic acid

acid to the CdTe would be manifest by a shortening of the folic acid lifetime in the nanoconjugates. Thus, there is no evidence of energy transfer in this system.

The pre-conditions for energy transfer from a donor to an acceptor are that the emission of the donor must effectively overlap with the absorption of the acceptor, and the donor and acceptor must be close together with separation less than 10 nm (Clegg 1996). If these two conditions are met, energy transfer can occur. However, the energy transfer efficiency (η_{ET}) is not only determined by the energy overlap and separation between the donor and acceptor, but also related to the luminescence lifetimes of the donor and acceptor excited states (Clapp et al. 2004; Clapp et al. 2005). As shown in Fig. 8, the energy transfer from the excited states of the donor to the excited states of the acceptor is a competitive process with the relaxation to the ground state of the donor. Obviously, if the lumines-

cence lifetime of the donor (τ_d) is long and that of the acceptor (τ_a) is short, energy transfer can be very efficient. On the contrary, if the luminescence lifetime of the donor (τ_d) is short and that of the acceptor (τ_a) is long, energy transfer would be very inefficient. In the case of CdTe-folic acid system, the luminescence lifetime of folic acid is about five times shorter than that of CdTe quantum dots, and this is likely one reason why there is no energy transfer from folic acid to CdTe quantum dots in the conjugates. As there is no energy transfer from folic acid to CdTe quantum dots, the conjugation is not favorable for the luminescence enhancement of the quantum dots. On the contrary, during the conjugation, the CdTe quantum dot surface coating might be damaged and surface charges might be reduced because folic acid can reduce the pH value of the quantum dot solution. It is known that TGA-coated CdTe quantum dots are only stable at high pH

Fig. 10 Micrographs of KB cells incubated with QD/QD-FA obtained through fluorescence microscopy as observed under the 20× objective. *Green* Unlabeled human nasopharyngeal epidermal carcinoma cell line over-expressing surface receptors for folic acid. *Red* KB cells labeled with folic-acid-conjugated quantum dots
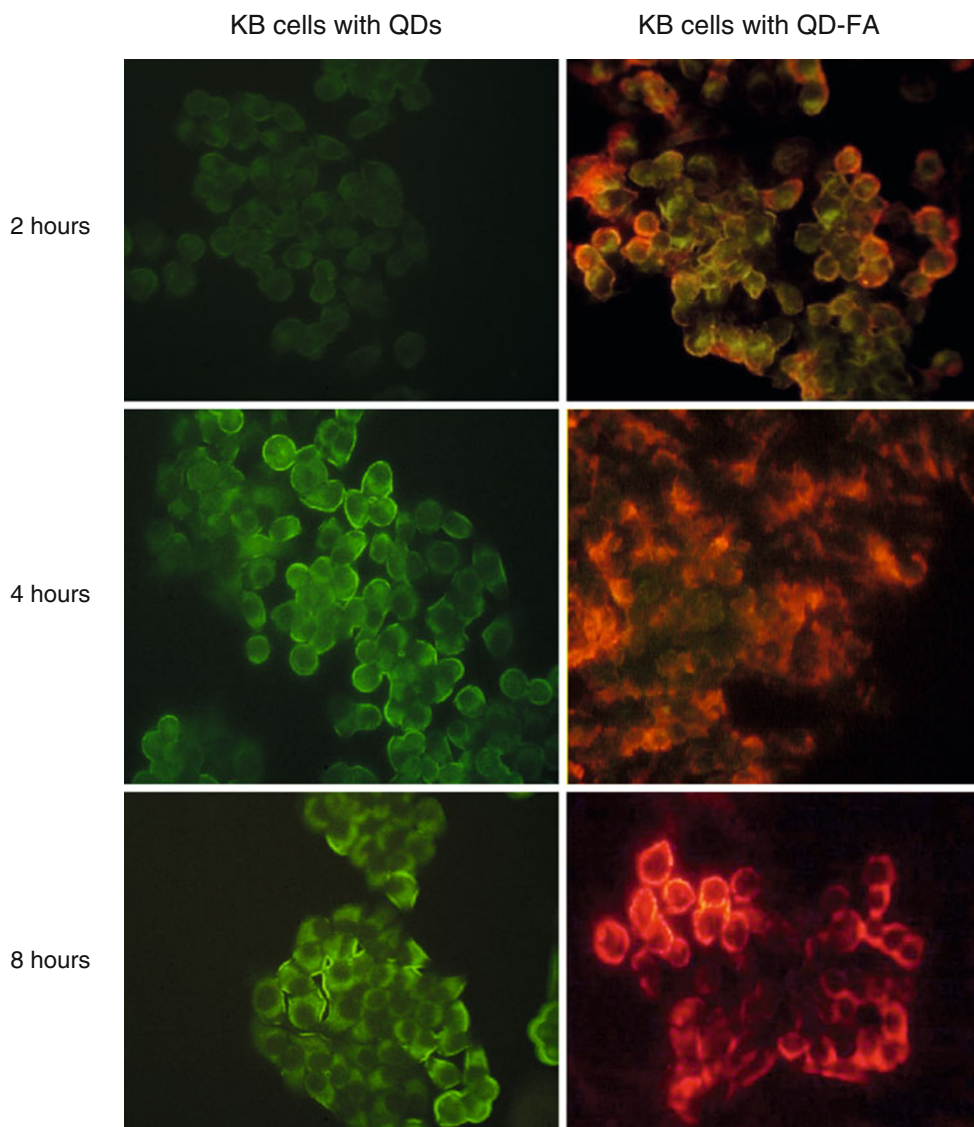
values (higher than 9.0) (Gaponik et al. 2002). Thus, the changes in the quantum dot surfaces and the pH value are likely responsible for the fluorescence quenching. The damage of the CdTe quantum dot surface coating by folic acid can be also reflected in the luminescence lifetime studies as shown in Fig. 9. The luminescence lifetime of CdTe quantum dot emission at 600 nm is shorter in the CdTe-folic acid mixture than that of the CdTe quantum dots. In the CdTe-folic acid conjugates, the CdTe quantum dot luminescence lifetime is much shorter than pure CdTe quantum dots. It is well-known that the increase of surface states or defects can shorten the luminescence lifetime and quench the luminescence.

In Figs. 3 and 5 the CdTe emission shows a dual peak between 600 and 650 nm. In the QD-FA conjugates the stronger peak is near ~640 nm while in CdTe QDs alone or mixed with FA the stronger peak is close to ~610 nm. This is a very interesting phenomenon that might give some information about the luminescence quenching of the QDs after conjugation with folic acid. It has been reported that the emission from II-VI semiconductor quantum dots is actually a combination of surface defect and excitonic emission (Joly et al. 2005; Liu et al. 2006; Chung et al. 2006). Most likely, the long-wavelength side is mainly due to surface defects, and the short-wavelength side is mainly due to the excitons. That the QD-FA conjugates show a stronger peak close to ~640 nm while CdTe alone or mixed with FA shows a stronger peak close to ~610 nm implies that more surface defects are produced in the quantum dots during the conjugation. More surface defects quench the excitonic luminescence and enhance the luminescence long-wavelength component. This is in agreement with the conclusion from the lifetime measurements that the production of surface defects during conjugation with folic acid is a major cause of the luminescence quenching.

Fig. 11 Micrographs of A549 cells incubated with QD/QD-FA obtained through fluorescence microscopy as observed under the 20× objective. *Green* Human lung carcinoma cell line lacking folic acid receptors
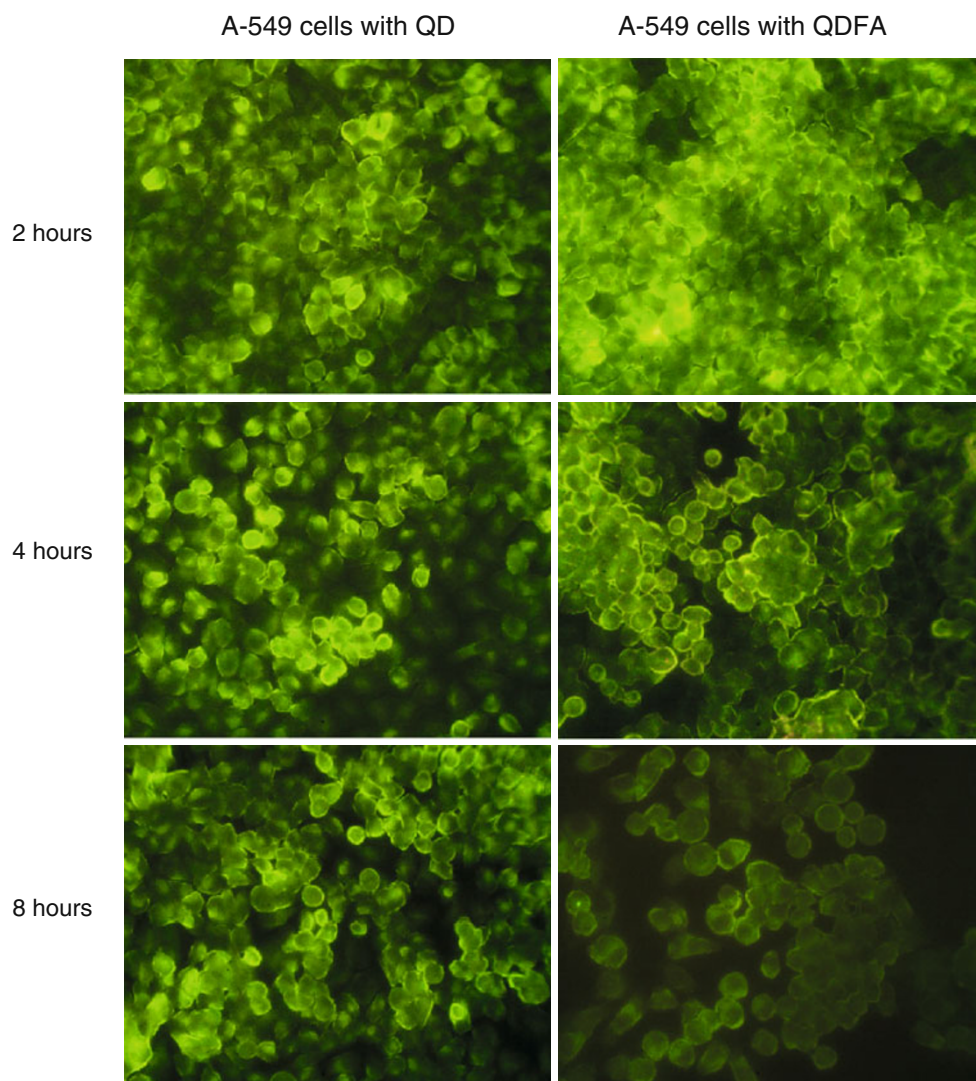
3.3 Tumor cell targeting observations

Even though the quantum dot luminescence is quenched somewhat by folic acid, the conjugates still have fairly intense luminescence after conjugation, which allows for cell imaging or labeling. To investigate the efficiency of folic acid-CdTe nanoconjugates for tumor targeting, pure CdTe quantum dots and folic acid coated CdTe quantum dots were incubated with human nasopharyngeal epidermal carcinoma cell line with positive folic acid receptors (KB cells) and lung cancer cells with negative folic acid receptors (A549 cells). Figure 10 displays the results of the uptake of the CdTe quantum dots (on the left) and the CdTe-folic acid nanoconjugates (one the right) by the KB cells after incubation for 2, 4, and 8 h, respectively. It is noted that the colors in the images are arbitrary and the green emission from the cells is probably from proteins or other molecules attached to the cells. Only the red or yellowish emissions are from the CdTe quantum dots. Clearly, the uptake of the CdTe-folic acid nanoconjugates by the KB cells is very high, while the uptake of the KB cells to the pure TGA-coated CdTe quantum dots is negligeable. Figure 11 shows the the uptake of the CdTe quantum dots (on the left) and the CdTe-folic acid nanoconjugates (one the right) by the A549 cells after incubation for 2, 4, and 8 h, respectively. The results show that almost no CdTe quantum dots or CdTe-folic acid conjugates were uptaken by the A549 cells. Figure 12 shows the uptake of CdTe-folic acid nanoconjugates by KB cells and A549 cells after incubation for 2, 4, and 8 h, respectively. As expected, the uptake of the CdTe-folic acid conjugates by KB cells is very high, but by A549 cells is almost nonexistent. Our observations demonstrate clearly the affinity and selectivity of folic acid as a

Fig. 12 Column 1: represents the ability of the folate receptors to bind to the folic acid coated QDs, thereby labeling the cells. Green Unlabeled KB cells. Orange/red KB cells labeled with folic acid-coated QD. Column 2: represents the inability of the A549 cells to bind the folic acid-coated QD, due to the lack of folate receptors on their surface. Green (on row 2)= A549 cells

KB Cells with QD FA   A-549 cell with QDFA

2 hours

4 hours

8 hours
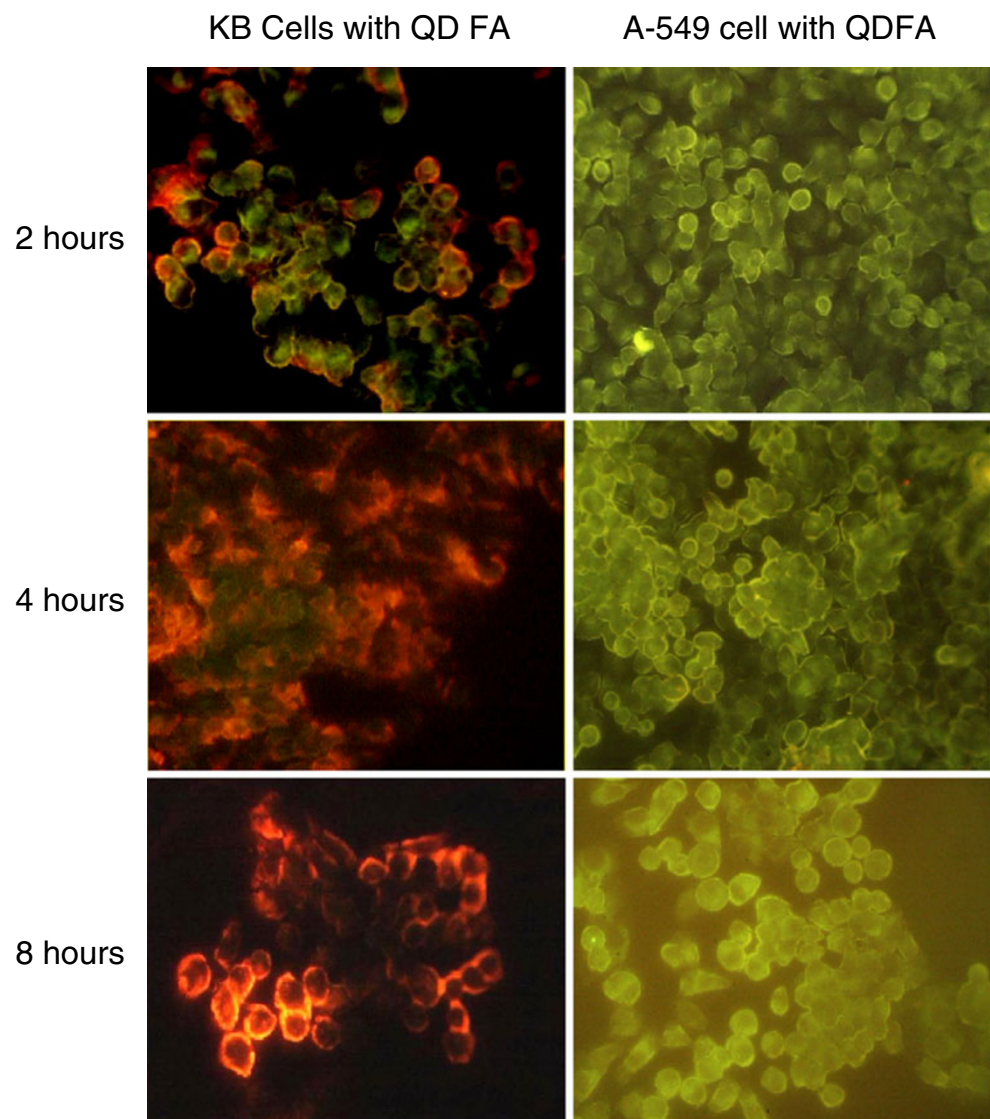
targeting ligand for tumor cells with positive folate receptors.

4 Conclusion

In summary, the luminescence and targeting properties of folic acid-CdTe quantum dot conjugates were studied and the interaction between folic acid and CdTe quantum dots was investigated. No energy transfer is observed from folic acid to CdTe quantum dots in CdTe-folic acid nanoconjugates and this is attributed to the fact that the luminescence lifetime of folic acid is much shorter than the lifetime of the CdTe quantum dots. The change of the pH values and possible damage of the quantum dot surface coating are likely the direct causes to the luminescence quenching of CdTe quantum dots after conjugation. The specific tumor targeting efficiency of the folic acid-CdTe nanoconjugates was evaluated by a comparative uptake study of the targeted and non-targeted QDs in KB cells (FR+) and A549 cells (FR−). The observations further demonstrate the promising potential of the folic acid-CdTe QDs for targeted tumor cell detetction.

References

ACS (2009) Cancer Facts & Figures. American Cancer Society, Atlanta, GA

Antony AC (1996) Folate receptors. Annu Rev Nutr 16:501–521

Bharali DJ, Lucey DW, Jayakumar H, Pudavar HE, Prasad PN (2005) Folate-receptor-mediated delivery of InP quantum dots for bioimaging using confocal and two-photon microscopy. J Am Chem Soc 127(32):11364–11371

Carbone L, Kudera S, Carlino E, Parak WJ, Giannini C, Cingolani R, Manna L (2006) Multiple wurtzite twinning in CdTe nanocrystals induced by methylphosphonic acid. J Am Chem Soc 128(3):748–755

Chen W (2008) Nanoparticle fluorescence based technology for biological applications. J Nanosci Nanotechnol 8(3):1019–1051

Chen W, Sun XK (2008) Luminescent nanoparticles for biological applications: imaging, therapy, and targeting strategies. In: Chen X (ed) Recent Advances of Bioconjugate Chemistry in Molecular Imaging Kerala. Research Signpost, India

Chen W, Grouquist D, Roark J (2002) Voltage tunable electroluminescence of CdTe nanoparticle light-emitting diodes. J Nanosci Nanotechnol 1(1):47–53

Chen Q, Ma Q, Wan Y, Su X, Lin Z, Jin Q (2005) Studies on fluorescence resonance energy transfer between dyes and water-soluble quantum dots. Luminescence 20(4–5):251–255

Chen W, Joly AG, Morgan NY (2006a) Optical Physics and Applications of Luminescent Nanoparticles. In: Balandin AA, Wang KL (eds) Handbook of Semiconductor Nanostructures and Devices, vol 2. American Scientific Publishers, Los Angeles, pp 295–334

Chen W, Zhang J, Westcott SL, Joly AG, Malm JO, Bovin JO (2006b) The Origin of X-ray Luminescence from CdTe Nanoparticles in CdTe/BaFBr:Eu2+ Nanocomposite Phosphors. J Appl Phys 99 (34302):34301–34305

Chou PT, Chen CY, Cheng CT, Pu SC, Wu KC, Cheng YM, Lai CW, Chou YH, Chiu HT (2006) Spectroscopy and femtosecond dynamics of type-II CdTe/CdSe core-shell quantum dots. Chemphyschem 7(1):222–228

Chung I, Witkoskie JB, Cao J, Bawendi MG (2006) Description of the fluorescence intensity time trace of collections of CdSe nano-crystal quantum dots based on single quantum dot fluorescence blinking statistics. Phys Rev E Stat Nonlin Soft Matter Phys 73(1 Pt 1):011106

Clapp AR, Medintz IL, Mauro JM, Fisher BR, Bawendi MG, Mattoussi H (2004) Fluorescence resonance energy transfer between quantum dot donors and dye-labeled protein acceptors. J Am Chem Soc 126(1):301–310

Clapp AR, Medintz IL, Fisher BR, Anderson GP, Mattoussi H (2005) Can luminescent quantum dots be efficient energy acceptors with organic dye donors? J Am Chem Soc 127(4):1242–1250

Clegg RM (1996) Fluorescence resonance energy transfer. In: Wang X, Herman B (eds) Fluorescence imaging spectroscopy and microscopy. Wiley, New York, pp 179–252

Colditz GA, Sellers TA (2006) E ET. Epidemiology—identifying the causes and preventability of cancer. Nat Rev Cancer 6:75

Gaponik N, Talapin DV, Rogach AL, Hoppe K, Shevchenko EV, Kornowski A, Eychmuller A, Weller H (2002) Thiol-capping of CdTe nanocrystals: an alternative to organometallic synthetic routes. J Phys Chem B 106(29):7177–7185

Hermanson GT (1996) Bioconjugate techniques. Academic, San Diego

Hu H, Xiong LQ, Zhou J, Li FY, Cao TY, Huang CH (2009) Multimodal-luminescence core-shell nanocomposites for targeted imaging of tumor cells. Chem Eur J 15:3577–3584

Joly AG, Chen W, McCready DE, Malm J-O, Bovin J-O (2005) Upconversion luminescence of CdTe nanoparticles. Phys Rev B 71(16):165304

Leamon CL, Low PS (2001) Folate-mediated targeting: from diagnostics to drug and gene delivery. Drug Discov Today 6:44

Lei Y, Tang H, Zhou C, Zhang T, Feng M, Zou B (2008) Incorporating fluorescent quantum dots in water soluble polymer. J Lumin 128(3):277–281

Liu Y, Chen W, Joly AG, Wang Y, Pope C, Zhang Y, Bovin JO, Sherwood P (2006) Comparison of water-soluble CdTe nano-particles synthesized in air and in nitrogen. J Phys Chem B Condens Mater Surf Interfaces Biophys 110(34):16992–17000

Manzoor K, Johny S, Thomas D, Setua S, Menon D, Nair S (2009) Bioconjugated luminescent quantum dots of doped ZnS: a cyto-friendly system for targeted cancer imaging. Nanotechnology 20 (65102):65101–65113

Morgan NY, English S, Chen WW, Chernomordik V, Russo A, Smith PD, Gandjbakhche A (2005) Real time in vivo non-invasive optical imaging using near-infrared fluorescent quantum dots. Acad Radiol 12:313–323

Smith AM, Dave S, Nie S, True L, Gao X (2006) Multicolor quantum dots for molecular diagnostics of cancer. Expert Rev Mol Diagn 6(2):231–244

Sudimack J, Lee RJ (2000) Targeted drug deliver via the folate receptor. Adv Drug Deliv Rev 41:147

Thomas AH, Lorente C, Capparelli AL, Pokhrel MR, Braun AM, Oliveros E (2002) Fluorescence of pterin, 6-formylpterin, 6-carboxypterin and folic acid in aqueous solution: pH effects. Photochem Photobiol Sci 1:421–426

Tyagi A, Penzkofer A (2010) Fluorescence spectroscopic behaviour of folic acid. Chem Phys 367:83–92

Wan Y, Wang LP, Lin BZ, Chen QD, Zhang H, Yang B, Su XG, Jin QH (2004) Studies on quantum dots synthesized in aqueos solution for biological labeling. Can J Anal Sci Spectros 49(2):1–6

Wang S, Low PS (1998) Folate-mediated targeting of antineoplastic drugs, imaging agents, and nucleic acids to cancer cells. J Control Release 53:39

Wang SP, Mamedova N, Kotov NA, Chen W, Studer J (2002) Antigen/antibody immunocomplex from CdTe nanoparticle bioconjugates. Nano Lett 2(8):817–822

Yang C, Ding N, Xu Y, Qu X, Zhang J, Zhao C, Hong L, Lu Y, Xiang G (2009) Folate receptor-targeted quantum dot liposomes as fluorescence probes. J Drug Target 17(7):502–511

Zhang Y, Kohler N, Zhang M (2002) Surface modification of superparamagnetic magnetite nanoparticles and their intracellular uptake. Biomaterials 23:1553–1561

Zhang Y, Chen W, Zhang J, Pope JL, Chen G, Pope C (2006) In vitro and in vivo Toxicity of CdTe nanoparticles. J Nanosci Nanotechnol 7:497–503