Supplementary Material (ESI) for Medicinal Chemical Communications

Supporting Information

Effect of Particle Shape on Phagocytosis of CdTe quantum dot-cystine Composites

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Experimental section

Synthesis of CdTe QD-cystine composites: All commercially obtained chemicals and materials were used directly without further purification. Deionized water (resistivity over 18 MΩ cm) from a Millipore Q water purification system was used in all experiments.

CdTe QD-cystine composites were fabricated with a procedure similar to the approach used in our previous quantum dots synthesis, with minor modifications.[1,2] In brief, 2 mL of cadmium chloride solution (0.04 mol L^{-1}) was diluted in 22.5 mL of deionized water, and then 800 mg of trisodium citrate dehydrate, 0.5 mL of sodium telluride solution (0.04 mol L^{-1}), certain amounts (see below) of L-cysteine, and 15 mg of sodium borohydride were added with vigorous stirring. After 15 min, the resulting mixture solution was loaded into a Teflon-lined stainless steel autoclave with a volume of 75 mL. The autoclave was maintained at 180°C for 40 min and then cooled to room temperature with flowing water. As described in our previous work,[3] L-cysteine was oxidized to L-cystine, in which CdTe nanocrystals were incorporated to form precipitates. By controlling the amount of L-cysteine added (6.0 mg, 12.5mg and 25 mg), different shaped CdTe QD-cystine composite particles were produced. The precipitates were collected by centrifugation at 6000 g for 10 min. After washing with water three times, the composite particles were suspended in water for further characterization.

Material Characterizations: 10 µl of composite suspensions were dropped on the smooth surfaces of aluminum films and dried overnight. After coating with a thin layer of platinum, the samples were examined with FESEM (JOEL, JSM-6700F, Japan). EDS spectra were recorded using the same samples. The dried composites were mixed with KBr to produce pellets for FTIR spectroscopy. FTIR spectra were obtained with a Perkin Elmer FT-IR Spectrum GX. Dry

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powders of the composites were also utilized for X-ray diffraction (XRD, Bruker AXS X-ray diffractometer) spectrometry. PL spectra of different composite suspensions were collected using an Aminco Bowman II luminescence spectrometer (Thermo Electron, USA) with an illumination source at 350 nm. Fluorescent images were acquired with a Zeiss LSM 510 Meta confocal microscope.

Cell culture: RAW 264.7 (Mouse leukaemic monocyte macrophage cell line) cells were used as model macrophages in this work. The cells were cultured in Dulbecco's Modified Eagle's Medium (DMEM) supplemented with 10% FBS in a humidified atmosphere containing 5% CO_2 at 37^{o}C.

Time-lapse LSCM: The cells were cultured at a density of 2×10^5/cm^2 on 5cm round glass coverslips for 24 h under standard culture conditions. Before each measurement, the coverslip was dammed with a home-made chamber that allowed the cell-attached surface of the coverslip to be immersed in 1 ml fresh culture medium. 50 μl colloidal suspension of composite particles was injected into the culture medium and bright-field images as well as fluorescent images were captured every 1 min for 1 h by the confocal microscope. The observation area was randomly selected from the coverslip. Bright-field and fluorescent images were merged to display the behaviors of the particles and macrophage cells. Typical stages of phagocytosis were selected to compose time-series images to better exhibit the internalization process.

SEM of the cells: After interaction with the particles for 1 h, the cells were washed with 0.01 M phosphate buffered saline (PBS, pH 7.4) several times to remove free particles. Then, 2.5% glutaraldehyde PBS solution was used to fix the cells at 4 ^{o}C for 12 h. The samples were dehydrated with a diluted ethanol series and dried under vacuum. The cell samples were coated with a thin layer of platinum prior to SEM. The SEM images are colored to better differentiate particles (purple) and cells (olive).

Internalization experiment: The internalization experiment was conducted following a published procedure, with modifications.[4] Particles were incubated with the cells over a time course range from 30 min to 4 h. After specific periods of incubation, the cells were washed with PBS three times to remove free particles. A cell scraper was applied to detach the cells from the culture plate. The collected cells were observed under a microscope. The internalization percentage was calculated with formula (1). 3 individual tests were repeated and at least 50 cells were counted in each test.

$$Internalization\ percentage(\%) = \frac{Number\ of\ cells\ with\ particles}{Number\ of\ counted\ cells}\times 100\% \quad (1)$$

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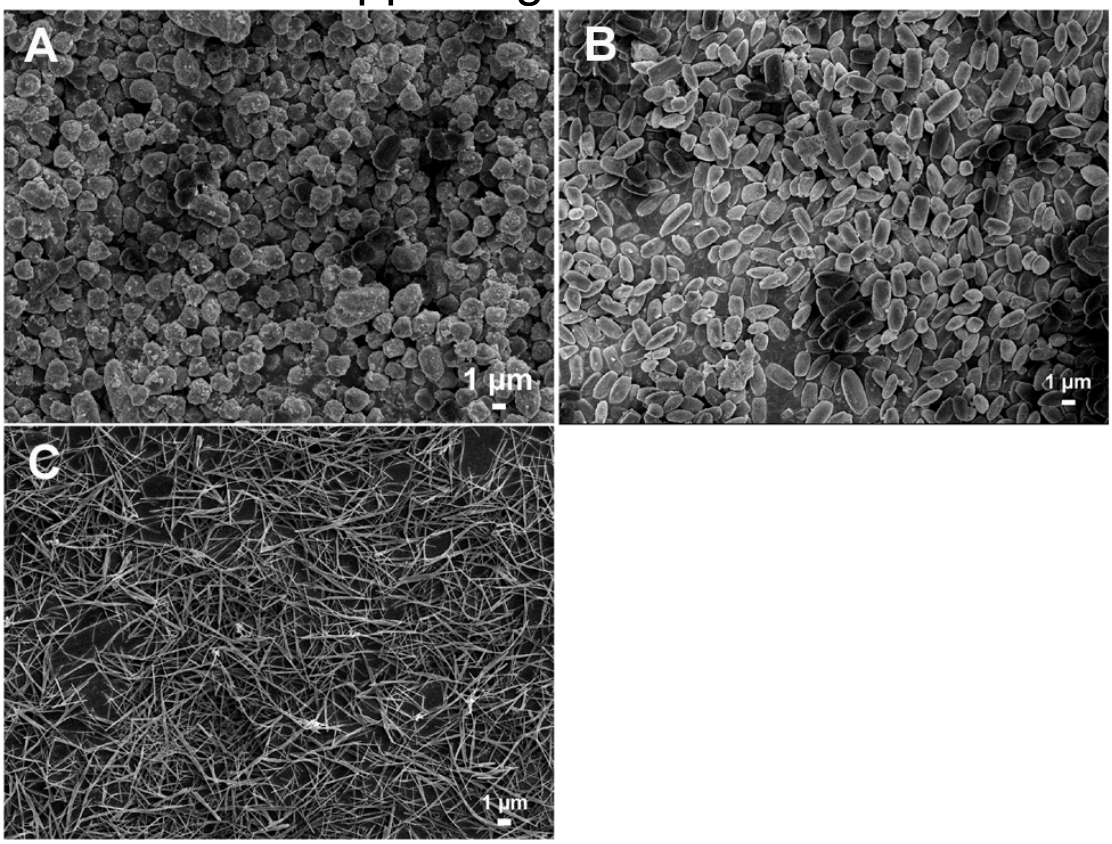

Figure S1. SEM micrographs of CdTe QD-cystine composites with controllable shapes: (A) microspheres, (B) microrods and (C) microneedles.

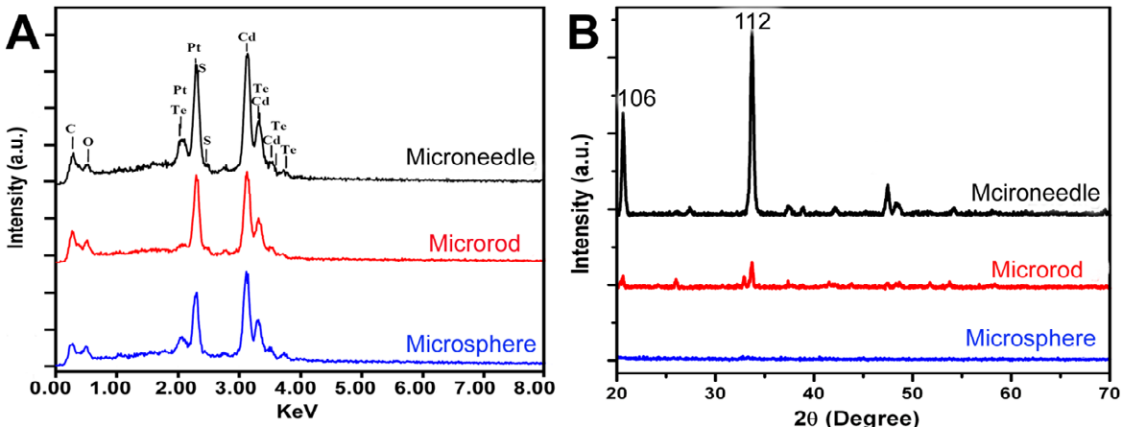

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Figure S2. Energy dispersive x-ray (A) and x-ray diffraction (B) spectra of CdTe QD-cystine microcomposites.

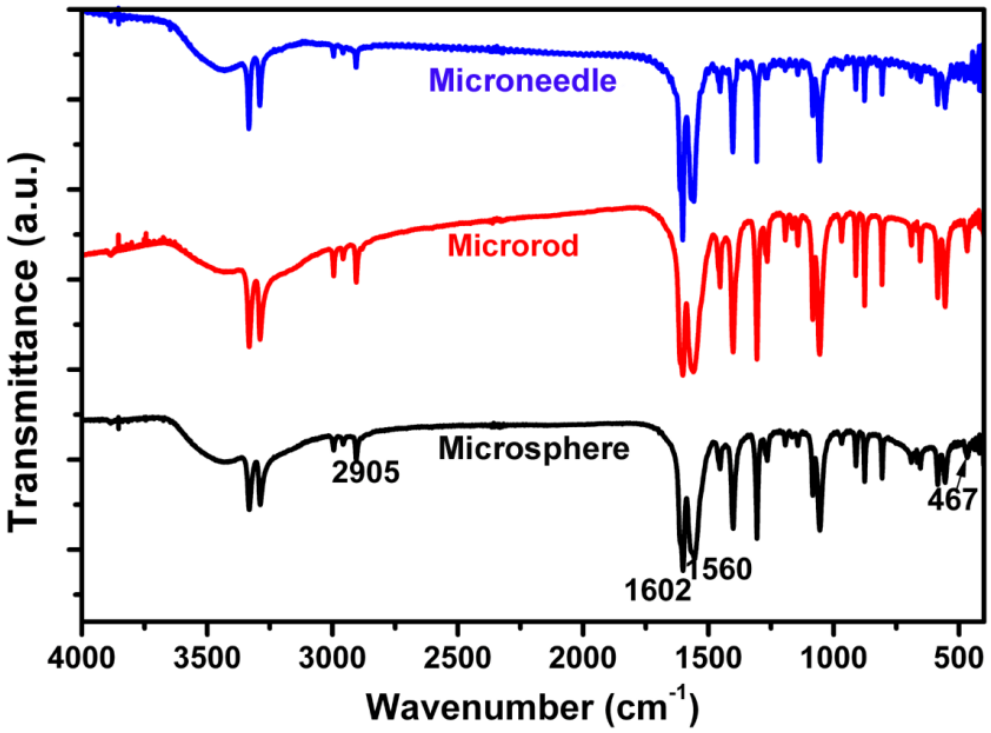

Figure S3. Fourier transform infrared spectra of CdTe QD-cystine composites with sphere, rod and needle shapes.

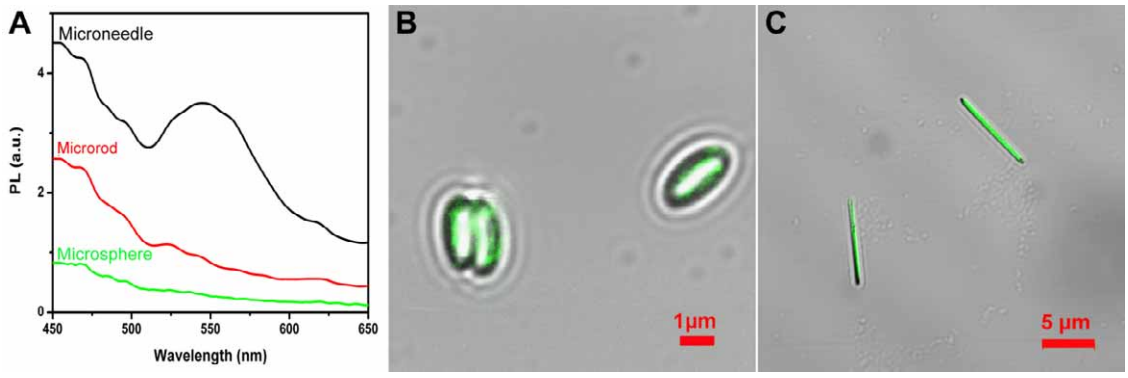

Figure S4. (A) Photoluminescence spectra of CdTe QD-cystine composites with sphere, rod and needle shapes. (B) and (C) Confocal microscopy images of microrod and microneedle CdTe QD-cystine composites, respectively.

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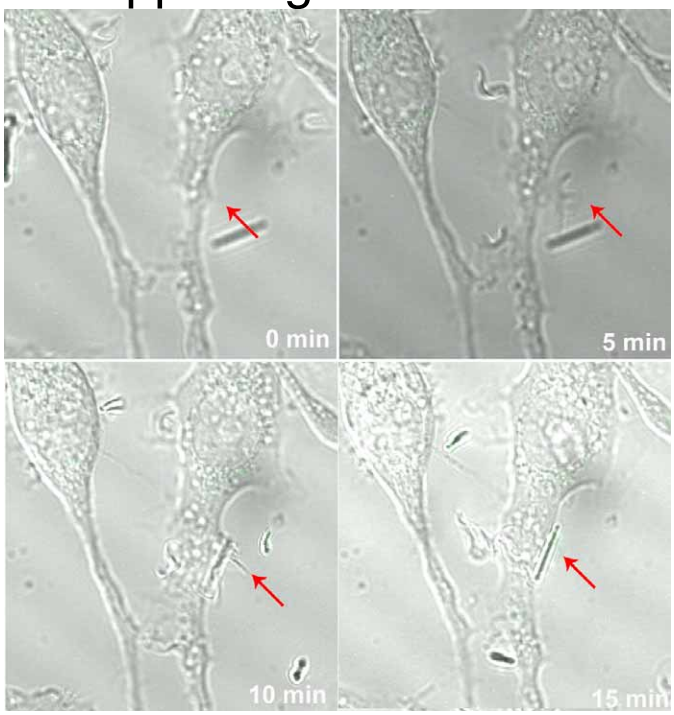

Figure S5. The active capture of a microneedle by the cell surface lamellar-structure. The arrows indicate the movement of the cell surface lamellar-structure.

Table S1. Cellular internalization profiles of different shaped CdTe QD-cystine microcomposites

Incubation Time (hour)	Cellular internalization percentage (%)		
	Microsphere	Microrod	Microneedle
0.5	54.7±3.5	15.66667±5.7	0±3.0
1	81.7±2.3	27.0±2.0	7.66667±3.1
2	81.7±2.3	39.66667±6.1	10.33333±5.0
4	81.7±1.2	30.33333±6.1	5.0±2.0

References

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2. Lu, Z. S.; Li, C. M.; Bao, H. F.; Qiao, Y.; Toh, Y. H.; Yang, X. *Langmuir* **2008**, *24*, 5445-52.

3. Bao, H. F.; Cui, X. Q.; Li, C. M.; Zang, J. F. *Nanotechnology* **2007**, *18*, 455701.

4. Gratton, S. E. A.; Ropp, P. A.; Pohlhaus, P. D.; Luft, J. C.; Madden, V. J.; Napier, M. E.; DeSimone, J. M. *Proc. Natl. Acad. Sci. U. S. A.* **2008**, 105, 11613-8.