Supporting Information

Facet energy and reactivity versus cytotoxicity: the surprising behavior of CdS nanorods

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1. Synthesis of CdS-L

To synthesize the CdS nanorods with the {101} facets, 1 mmol of cadmium acetate ($Cd(Ac)_2{\cdot}2H_2O$) and 2 mmol of L-cysteine ($C_3H_7NO_2S$) were added to a Teflon-lined stainless steel autoclave of 50 ml capacity. Then, 20 ml ethanolamine (EA) and 20 ml distilled water was added, and the mixture was stirred for 20 min at room temperature. Next, the autoclave was sealed and maintained at 180 °C for 24 h, and then cooled to ambient temperature. The resultant product was collected, washed with distilled water and absolute alcohol several times, vacuum-dried, and stored for further characterization and use.

2. Computational details

Our periodic density functional theory (DFT) computations were performed by using a plane wave basis set with the projector augmented plane wave (PAW) to model the ion-electron interaction as implemented in the Vienna ab initio simulation package (VASP). The generalized gradient approximation (GGA) with the PW91 functional and a 480 eV cutoff for the planewave basis set were adopted in all the computations. The Brillouin zone was integrated by using Monkhorst-Pack generated sets of k-points. 5×5×5 k-point meshes, and 2×2×2 supercell with 16 CdS pairs were benchmarked to be sufficient to reach the convergence for bulk computations. The surface computations were performed by using the slab model, in which a finite number of crystal layers in a three dimensional periodic cell were used to generate two surfaces via the introduction of a vacuum gap perpendicular to the surface. 2×2×1 k-point grid was used for the surface relaxation, with the third vector perpendicular to the surface, and the energy cut-off of the converged bulk computations (480 eV).

3. Toxicity tests with *R. niger*, *C. neoformans*, *C. albicans*, *E. coli* and *S. aureus* exposed to CdS nanorods

The strains *R. niger*, *C. neoformans*, *C. albicans*, *E. coli* and *S. aureus* used in this study were obtained from the Laboratory of Modern Mycology, Nankai University. The fungal strains were cultured in YPD medium (1% yeast extract, 2% peptone, and 2% dextrose), and the bacterial strains were cultured in LB medium (1% tryptone, 0.5% yeast extract, and 1% NaCl). The CdS nanomaterials (640 mg/L) were suspended in the respective medium containing 10^6 cells/mL of the strains. The mixtures were incubated by shaking for 24 h at 30 °C. The cultures were then diluted with sterile water, plated at YPD plates. The colony forming units (CFU) were then counted, and the percent of CFU were calculated as CFU of the treatment groups divided by CFU of the control (samples receiving no treatment of CdS). Each test was run in triplicate.

4. Adsorption experiments

First, a series of 40 ml vials each containing 640 μl 10 mg/mL CdS stock suspension were prepared. Then, different amounts of a BSA stock solution (in H_2O) were added to the vials, and the total volume was adjusted to 10 ml with double-distilled water. The vials were shaken at 180 rpm for 24 h to reach adsorption equilibrium. Afterward, the vials were left undisturbed at room temperature for 1 d, and the supernatant was withdrawn to analyze the concentrations of BSA using coomassie brilliant blue assay. The blank control samples of BSA showed no adsorption to the vial, and pH remained constant during all the experiments. The adsorbed mass at each equilibrium concentration was calculated based on a mass balance approach.

5. Fluorescence diacetate (FDA) staining method to assess cell viability

After a 6-h treatment with/without 640 mg/L CdS in YPD medium, the yeast cells were harvested, washed with PBS buffer and re-suspended in 1 ml of PBS buffer. Then, 2 μl FDA (1 mg/L in acetone, Sigma) was added to the cell suspensions. The mixture was equilibrated for 5 min and subsequently observed using a fluorescence microscope (BX-41, Olympus, Japan) with the green filter set. The numbers of FDA-stained cells and total cells were counted, and the percentages of the FDA-stained (viable) cells were calculated. At least 20 fields were analyzed.

6. Cell staining for endocytic phenomenon observation

After incubation with/without 640mg/L of CdS, the cells (wild-type and the endocytic deficient mutant *end3*Δ) were harvested, washed and suspended in YPD to an OD_{600} of 1.0. Four microliter of the lipophilic styryl dye, *N*-(3-triethylammoniumpropyl)-4-(p-diethylaminophenyl-hexatrienyl) pyridinium dibromide (FM4-64, 50 μg ml^{-1} in DMSO, Sigma) was added to 100 μl of the suspension, and the mixture was incubated at 30 °C by shaking at 50 rpm for 40 min in the dark. The cells were then centrifuged, and the harvested cells were re-suspended in 100 μl YPD medium and stained with 1 μl CFW(10 mg/L in distilled water, Sigma) and observed with a confocal laser scanning microscope (FV1000, Olympus, Japan) with a TRIT-C/Texas red filter set for the vacuolar membrane and a blue filter set for the cell wall.

7. Additional experiments with PEG-coated nanorods to determine the impact of surface passivation on nanoparticle bioavailability

To prepare the PEG-coated CdS nanorods, 1 g of CdS-H or CdS-L nanorods were added to PEG solution (PEG 3350, Sigma Aldrich, 40% (W/V), dissolved in dH_2O). After magnetic

stirring at room temperature for 24 h, the mixtures were centrifuged at 12,000 rpm for 10 min. The pellets were dried at room temperature, obtaining PEG-coated CdS-H nanorods (PEG-CdS-H) and PEG-coated CdS-L nanorods (PEG-CdS-L). The obtained nanorods were used for characterization, toxicity test, endocytosis assay and ER stress assay, using the procedures described in the main text and above. The concentration of each kind of nanorods used was 640 mg/L for CdS content.

PEG-coated CdS nanorods, together with the used PEG, were characterized by FITR (FTS6000, Bio-Rad, USA) and TG–DTA (Thermo plus EVO2, TG8121, Rigaku, Japan).

The concentration of each kind of nanorods used was 640 mg/L for CdS content: the concentration of uncoated CdS nanorods was 640 mg/L; that of PEG-coated CdS-H nanorods (CdS-H% = 75.3%) was 850 mg/L; that of PEG-coated CdS-L nanorods (CdS-L% = 76.56%) was 836 mg/L.

Particle uptake of PEG-coated and uncoated CdS nanorods was determined as described in the main text. The concentration of each kind of nanorods used was 640 mg/L for CdS content.

PEG-coated CdS-H and CdS-L were similarly coated by PEG

After 24 h of PEG coating, both nanorods were coated by PEG, which was confirmed by FITR (Figure S12(a)), since both PEG-coated nanorods showed similar absorbance peaks. Thermogravimetric analysis further revealed that CdS-H and CdS-L nanorods were coated with similar contents of PEG. The coating percent of PEG was 24.70% for CdS-H and 23.44% (Figure S12(b)).

PEG-coated CdS-H and CdS-L had similar endocytosis potential

Figure S13 revealed that PEG-CdS-H and PEG-CdS-L were biosorbed and/or internalized to a similar extent, which were higher than for uncoated CdS-H and CdS-L.

Table S1. Selected characteristics of CdS-H and CdS-L nanorods.

	CdS-H	CdS-L
Length (nm)	110 ± 26	108 ± 11
Width (nm)	25 ± 3	22 ± 4
Surface energy (J/m^2)	0.627	0.451
Surface area (m^2/g)	42.6	49.0
ζ potential (mV) [a]	-12.3 ± 1.6	-9.92 ± 1.18

[a] ζ potential measured for CdS in YPD medium.

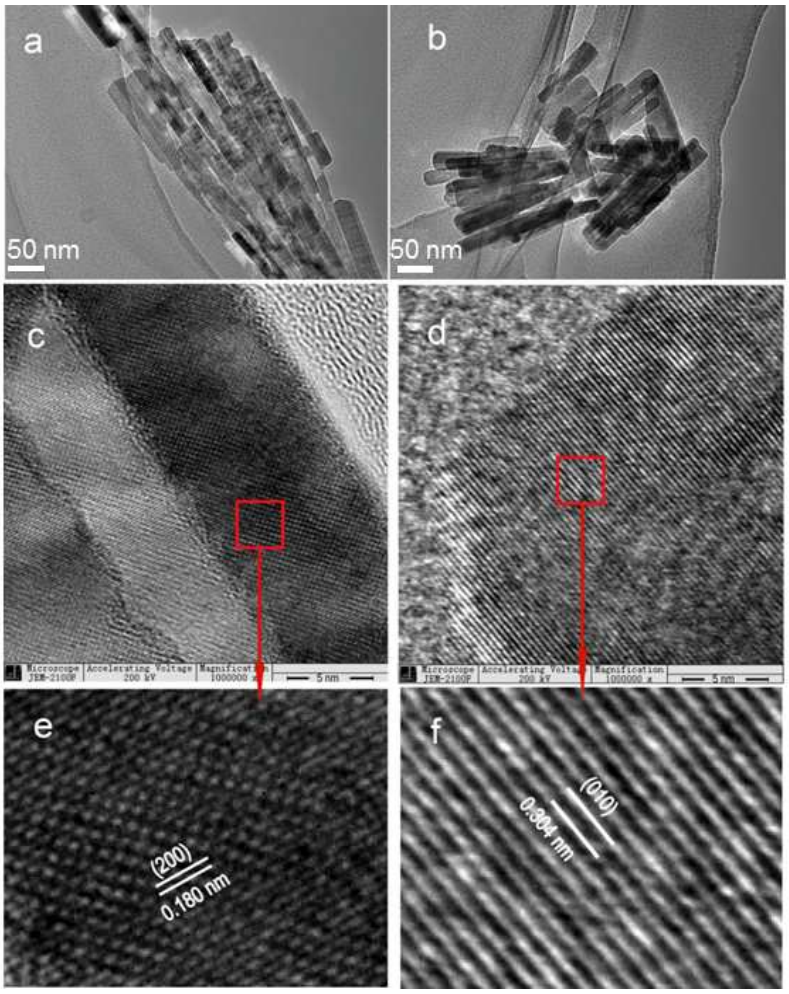

Figure S1. HRTEM images. (a, c, e) CdS-H. (b, d, f) CdS-L. e and f show the fringe spacings of the nanocrystals.

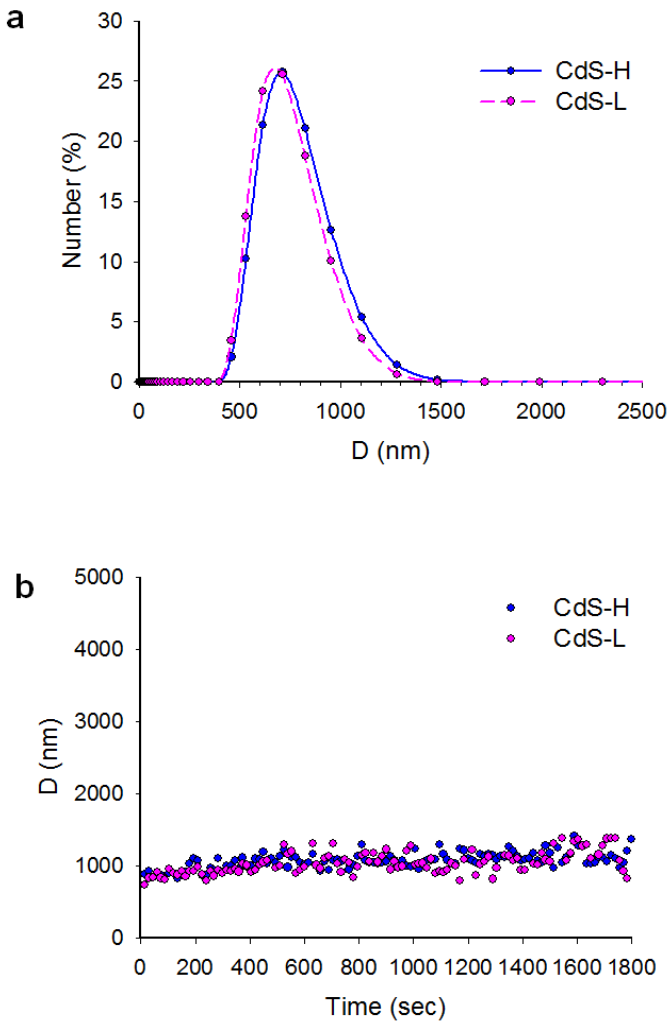

Figure S2. Particle size distribution (a) and aggregation kinetics (b) of CdS-H and CdS-L (20 mg/L) in YPD medium, determined with dynamic light scattering. D represents hydrodynamic diameter.

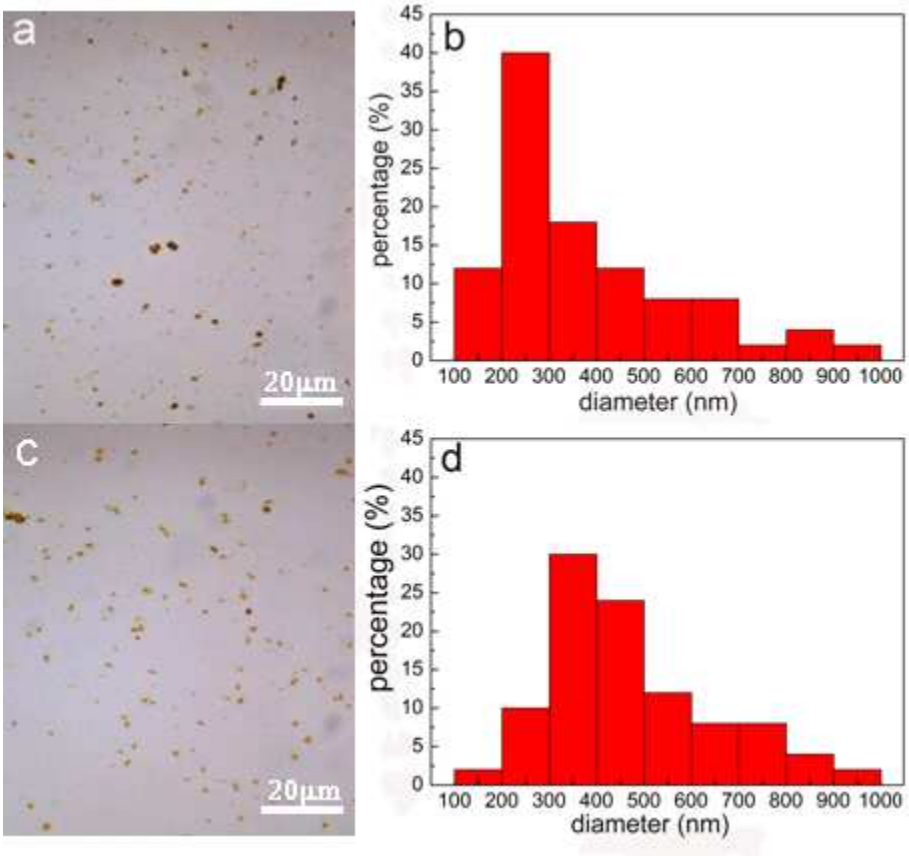

Figure S3. Optical microscopy images (left panel) and particle size distribution (right panel) of CdS nanorods in YPD medium. (a, b) CdS-H. (c, d) CdS-L.

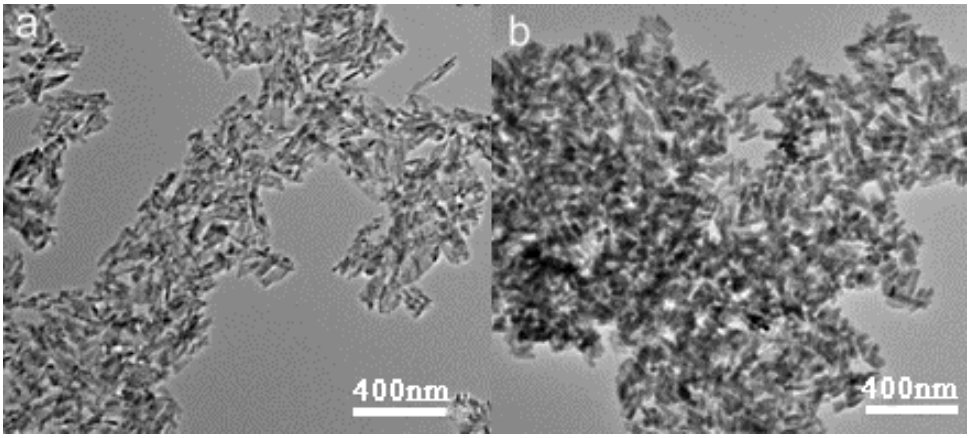

Figure S4. Representative TEM images of CdS nanorods in YPD medium. (a) CdS-H. (b) CdS-L.

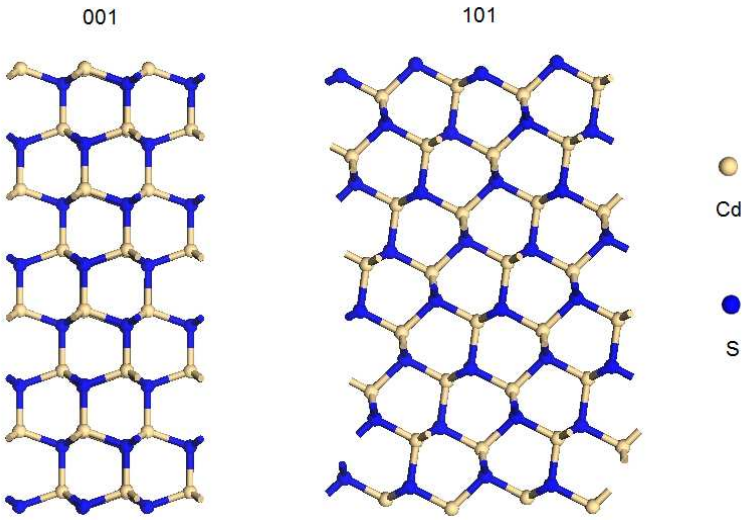

Figure S5. Slab models for {001} and {101} surfaces of CdS crystal.

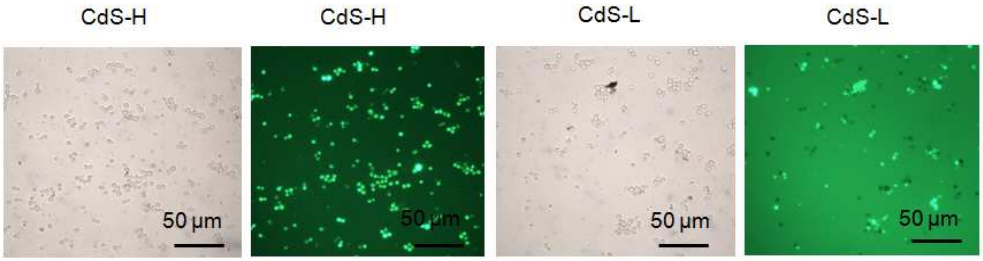

Figure S6. Fluorescence diacetate (FDA)-stained cells with treatment of CdS-H or CdS-L (640 mg/L) for 6 h.

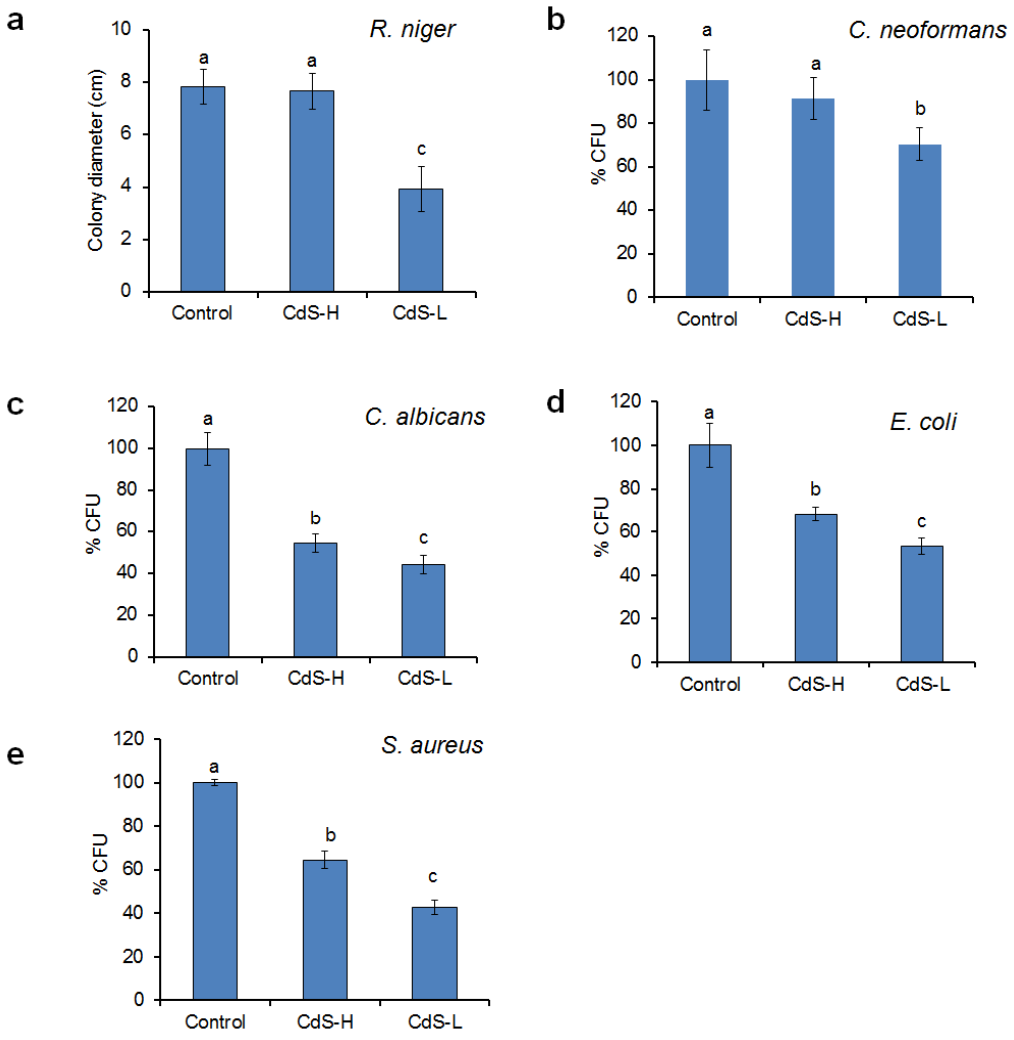

Figure S7. Inhibition effects of CdS-H and CdS-L (640 mg/L) on (a) *Rhizopus niger* and (b) *Cryptococcus neoformans* (c) *Candida albicans* (d) *Escherichia coli* and (e) *Staphylococcus aureus*. The incubation time was 24 h. "Control" represents cells receiving no treatment of CdS. CFU is a colony-forming units.

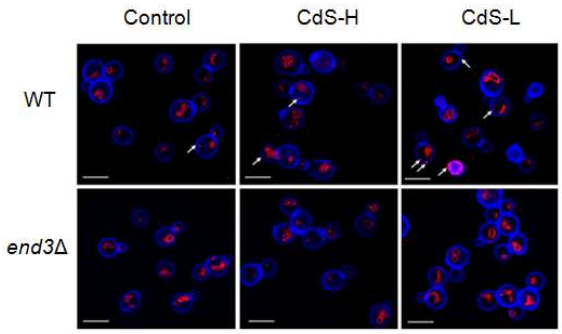

Figure S8. Effects of CdS on endocytosis and vacuole morphology. For CdS-H and control (cells receiving no treatment of CdS), the differences in amounts of endocytosis structures observed between wild-type (WT) and *end3*Δ strains were insignificant. In contrast, for CdS-L, significantly more endocytosis structures were observed for wild-type strain than for *end3*Δ strain.

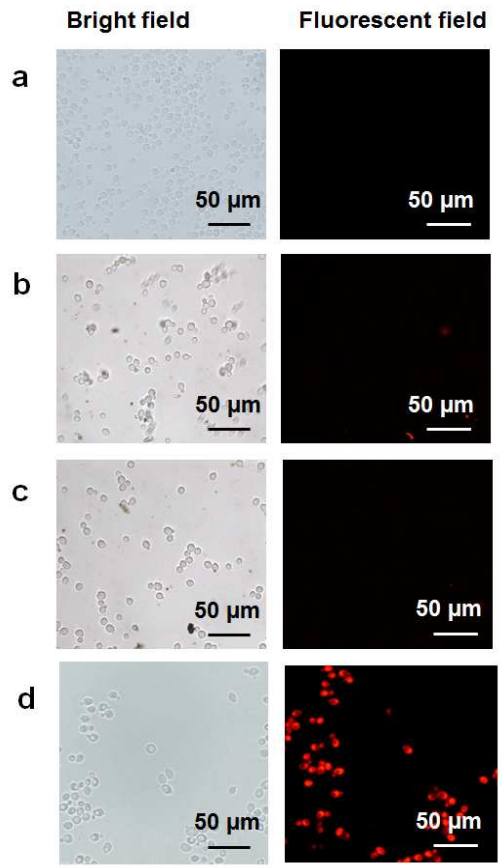

Figure S9. Fluorescence microscope images of yeast cells stained by propidium iodide (PI) show no significant damages to cell membranes. (a) negative control (cells receiving no treatment of CdS). (b) Yeast cells were treated with 640 mg/L CdS-H. (c) Yeast cells were treated with 640 mg/L CdS-L. (d) Positive control (yeast cells were incubated at 65 °C water bath for 30 min).

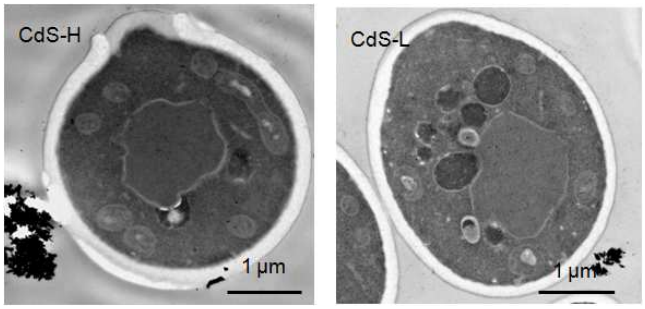

Figure S10. TEM images of *end3*Δ cells treated with CdS-H or CdS-L (640 mg/L) showing no detection of CdS inside cells.

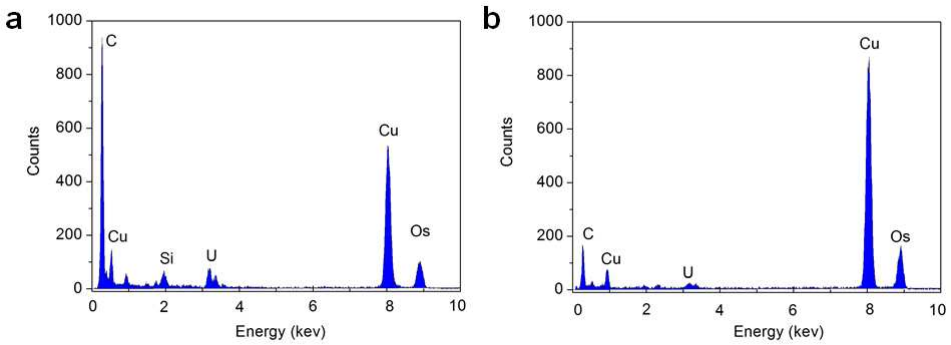

Figure S11. Energy dispersive spectrometry (EDS) analysis of *end3*Δ cells. (a) Cells treated with 640 mg/L CdS-H. (b) Cells treated with 640 mg/L CdS-L.

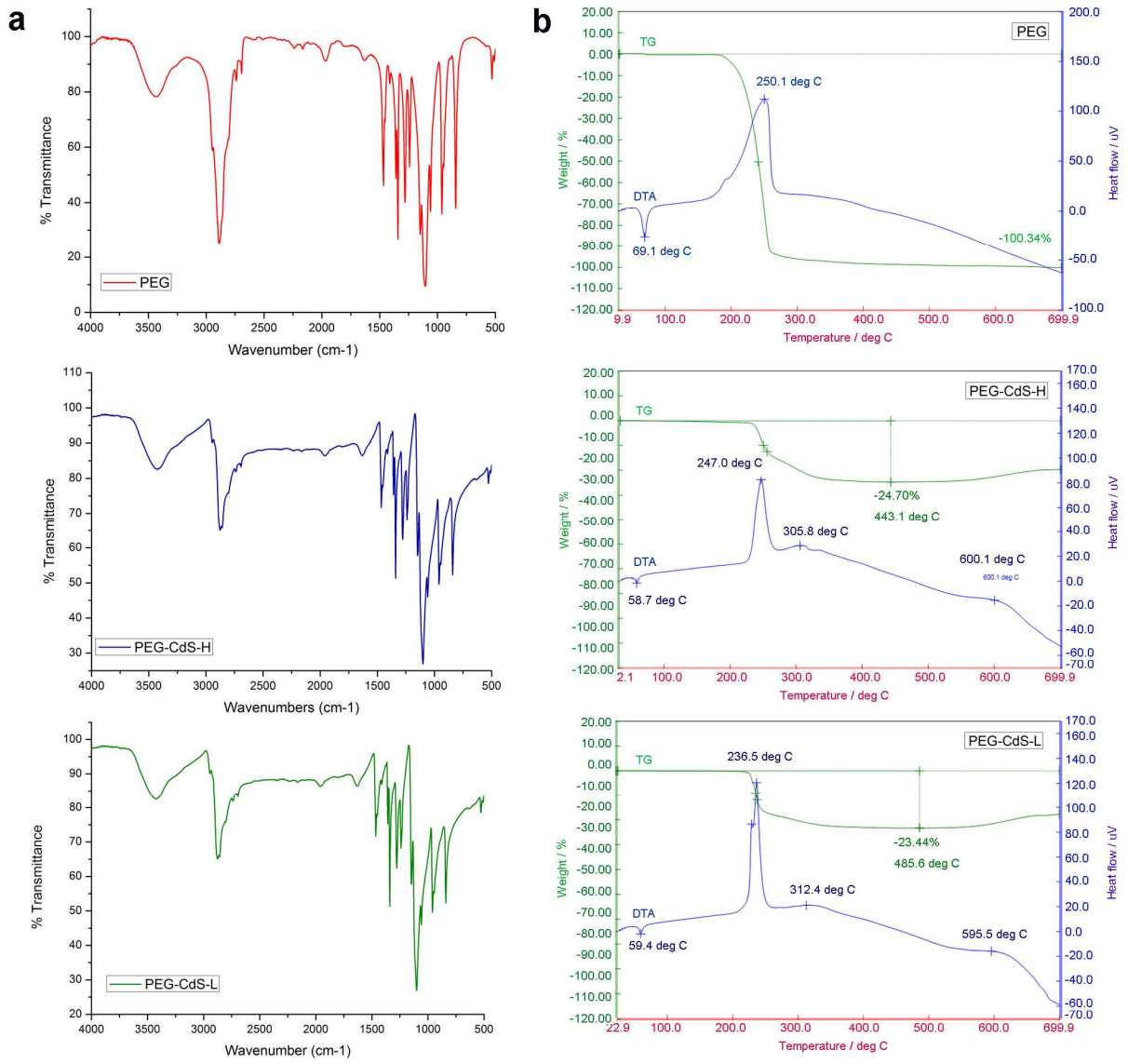

Figure S12. Characterization of PEG-coated CdS nanorods show similar extent of coating for CdS-L and CdS-H. (a) FITR of PEG, PEG-coated CdS-H and PEG-coated CdS-L. (b) Thermogravimetric analysis of the PEG-coated and uncoated nanorods.

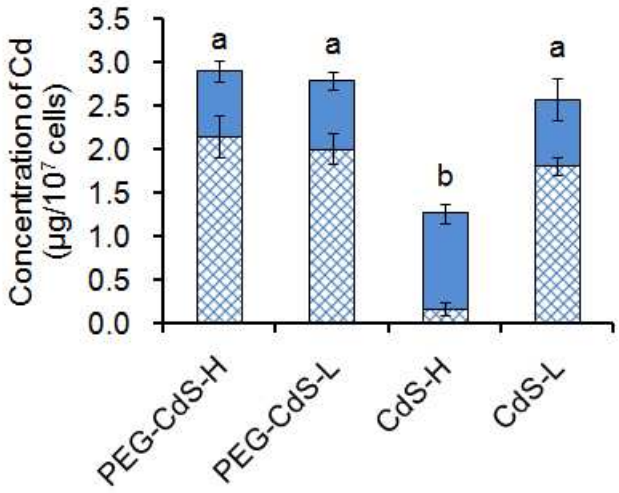

Figure S13. Total Cd accumulation in wild-type *Saccharomyces cerevisiae* exposed to 640 mg/L PEG-CdS-H or PEG CdS-L for 24 h. The PEG-coated CdS nanorods had undistinguishable endocytosis potential. No difference was observed for both intracellular Cd (checked patterns) and adhering Cd (unchecked patterns) between yeast cells exposed to PEG-CdS-H and the cells exposed to PEG-CdS-L. A significance ($p < 0.05$) difference in uptake was observed between coated CdS nanorods and uncoated CdS-H. The error bars indicate one standard deviations from the mean (n = 3).