Supporting Information

Amine Functionalized ZrO_2 Nanoparticles as Biocompatible and Luminescent Probes for Ligand Specific Cellular Imaging

Muhammad Nawaz Tahir[a†*], Ruben Ragg[a†], Filipe Natalio[a], Jugal Kishore Sahoo[a], Daniel Phillip[a], Kaloian Koynov[c], Dennis Strand[b], Susanne Strand[b], Wolfgang Tremel[a]*

a) Institut für Anorganische Chemie und Analytische Chemie, Johannes-Gutenberg-Universität, Duesbergweg 10–14, D-55099 Mainz, Germany.

b) I. Medizinische Klinik, Universitätsmedizin, Johannes-Gutenberg Universität, Langenbeckstrasse 1, 55131 Mainz, Germany.

c) Max Planck-Institut für Polymerforschung, Ackermannweg 12, 55128 Mainz, Germany.

CORRESPONDING AUTHOR FOOTNOTE. Corresponding author: Phone: +49 6131 392 5135; Fax. +49 6131 392-5605; e-mail: tremel@uni-mainz.de, tahir@uni-mainz.de

†These authors have equal contribution to this work.

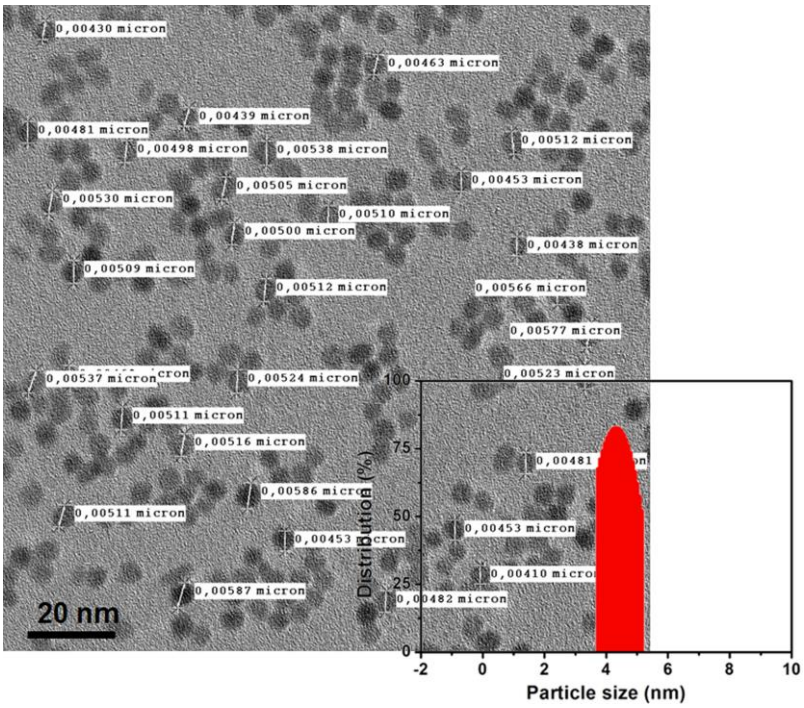

Figure S1. TEM image of unfunctionalized ZrO_2 NPs. Particle size distribution histogram obtained by averaging the sizes of approx. 100 nanoparticles.

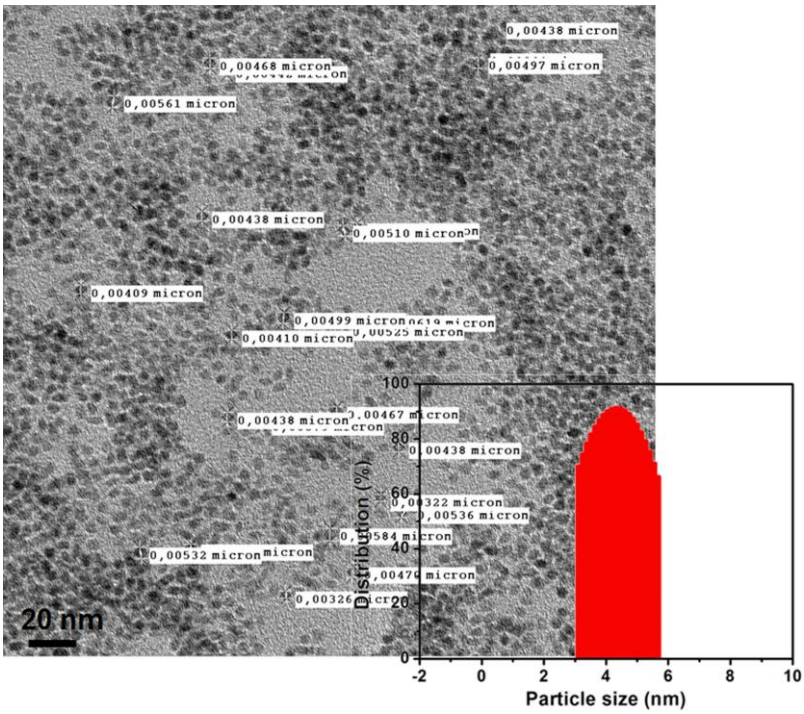

Fig. S2. TEM image of functionalized ZrO_2 NPs. Particle size distribution histogram obtained by using the average size of approx. 100 nanoparticles.

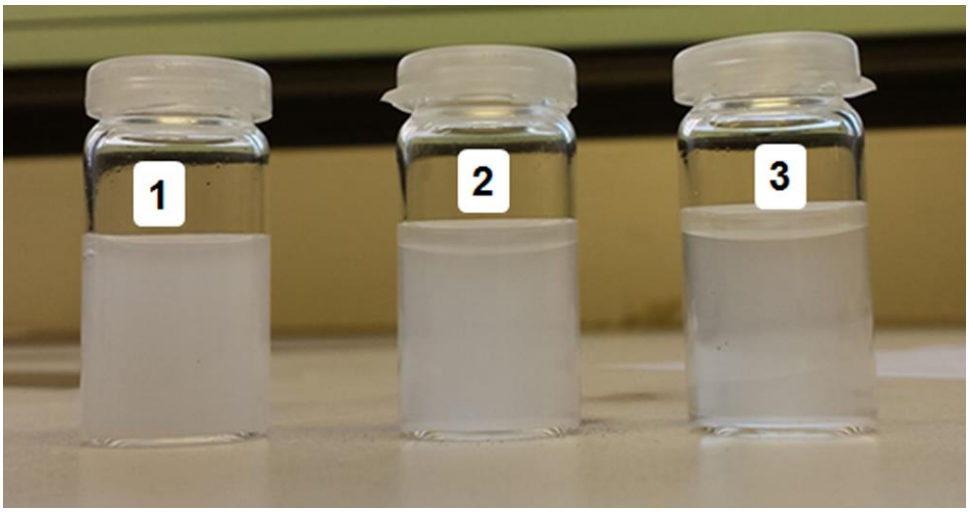

Fig. S3. Digital photograph showing the colloidal stability of 1) unfunctionalized ZrO_2 nanoparticles, 2) NH_2 functionalized and 3) TPP conjugated ZrO_2 nanoparticles. All three samples were prepared with a concentration of 1 mg/ml.

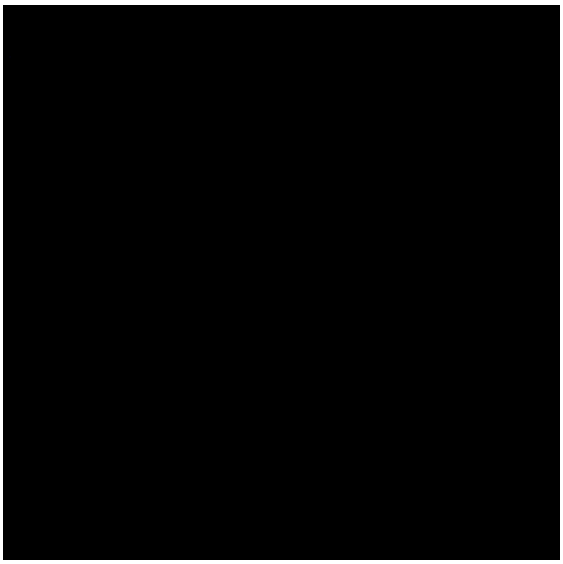

Fig. S4. CLSM image of unfunctionalized ZrO_2 NPs under excitation at 405 nm. No emitting signal was observed in agreement with the spectral data.

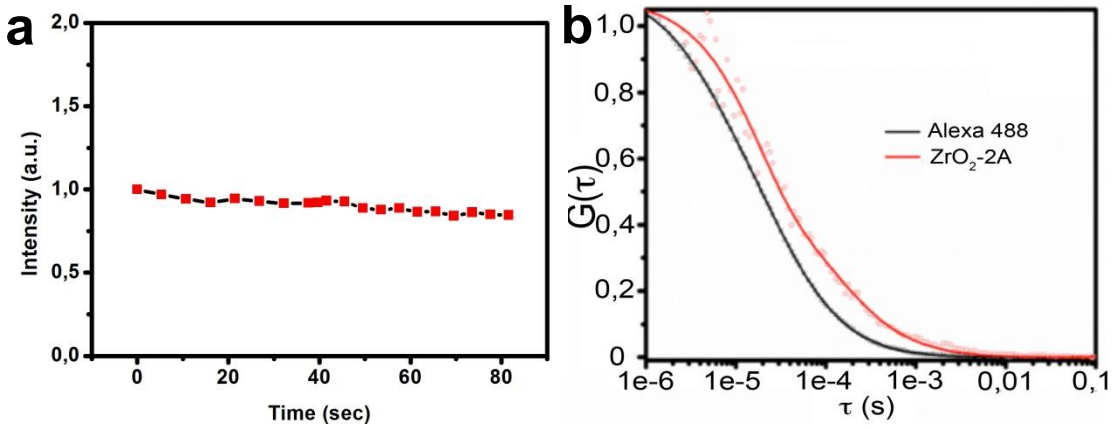

Fig. S5. a) Photo stability of F@ZrO_2 NPs when exposed to a laser (λ_{ex}=408 nm) with a power of 70% of 30 mW over 80 s. Almost no PL decay was observed under normal imaging conditions (4% of 30 mW). b) FCS autocorrelation curves for F@ ZrO_2 NPs (red circles) and Alexa 488 (black squares).

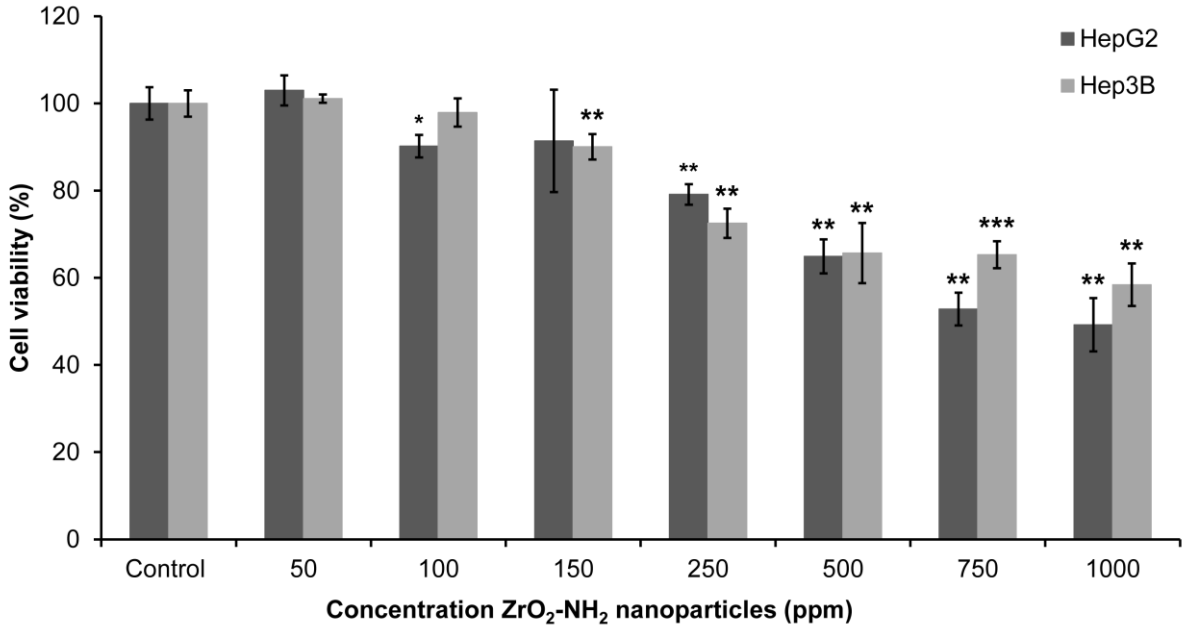

Fig. S6. Cytotoxicity assay with 1,4-butanediamine functionalized ZrO_2 NPs (ZrO_2-NH_2) in HepG2 and Hep3B cells after 24 h. No toxic behavior was observed up to 250 ppm for both cell lines, whereas higher NP concentrations ranging from 500 to 1000 ppm led a low cytotoxic response.

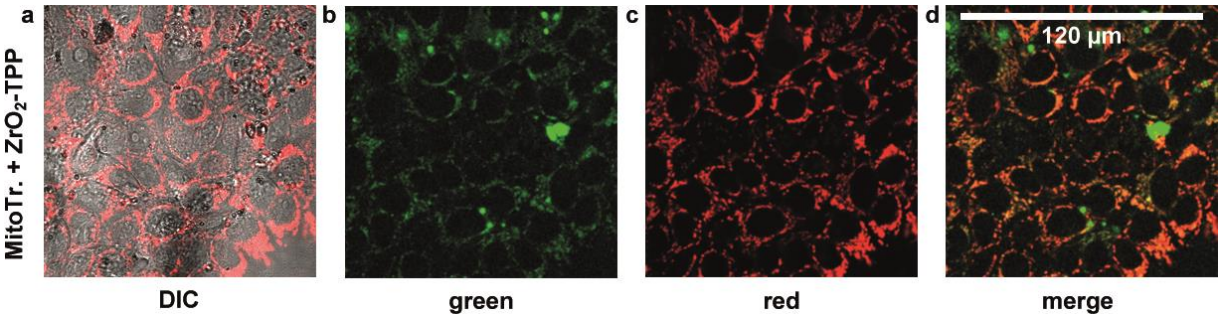

Fig. S7. CLSM colocalization studies of ZrO_2-TPP NPs in HepG2 cells. (a) Superimposed DIC image of treated HepG2 cells (b) Photoluminescent ZrO_2-TPP (λ_{ex} = 488 nm, green) and (c) MitoTracker Red (λ_{ex} = 543 nm, red) are taken up by the cells and achieve subcellular organization. (d) By merging the red and green channels the selective mitochondria targeting of the ZrO_2-TPP NPs is clearly visible and a PCC of 0.7 can be calculated.

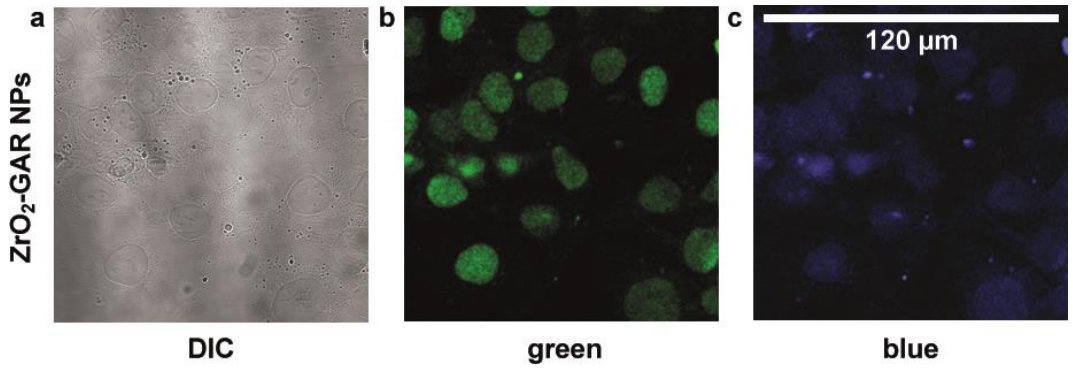

Fig. S8. CLSM localization studies of GAR-conjugated functionalized ZrO_2 NPs in Hep3B cells ectopically overexpressing Sirt6. (a) Superimposed DIC images of Hep3B+Sirt6 cells treated with Anti-Sirt6 primary antibody. (b) Subcellular localization of photoluminescent ZrO_2-GAR NPs after excitation at λ_{ex} = 488 nm (green) and (c) after excitation at λ_{ex} = 405 nm (blue).

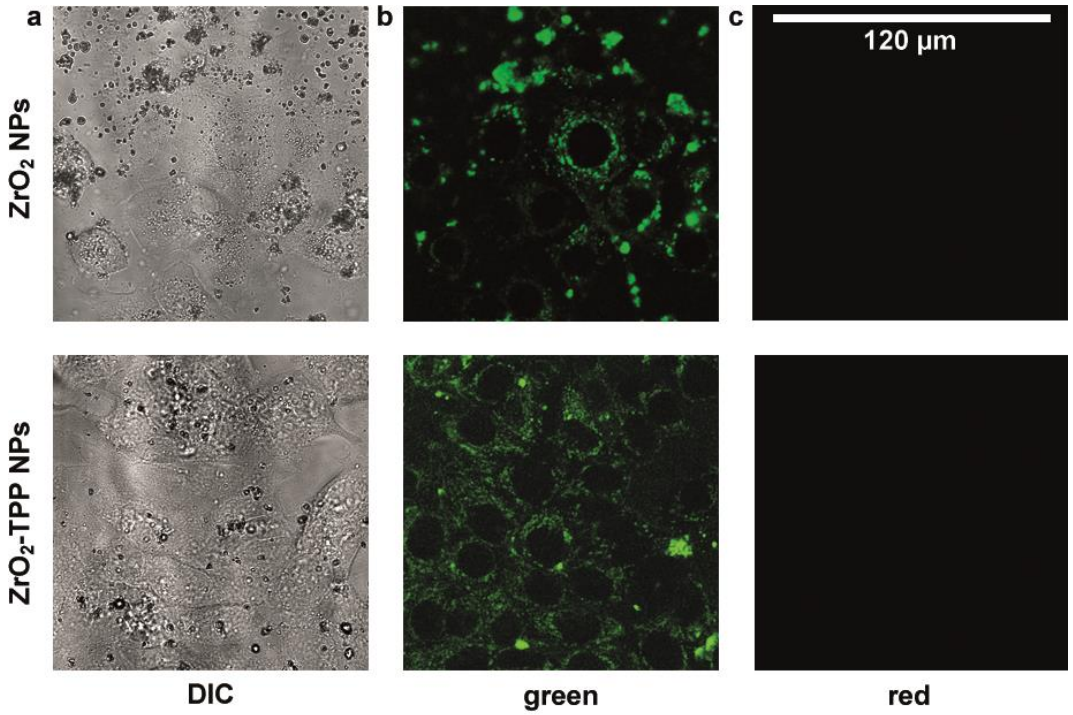

Fig. S9. CLSM colocalization studies of ZrO_2 and ZrO_2-TPP NPs in HepG2 cells. (a) Superimposed DIC image of treated HepG2 cells (b) Photoluminescence of unfunctionalized ZrO_2 and functionalized ZrO_2-TPP NPs (λ_{ex} = 488 nm, green). ZrO_2 NPs (top) achieve no subcellular localization compared to the mitochondria targeting ZrO_2-TPP NPs (bottom). (c) No luminescence was found in the red channel.