Supplementary Information

Theranostic icelles based on upconversion nanoparticles for dual-mode imaging and photodynamic therapy in hepatocellular carcinoma

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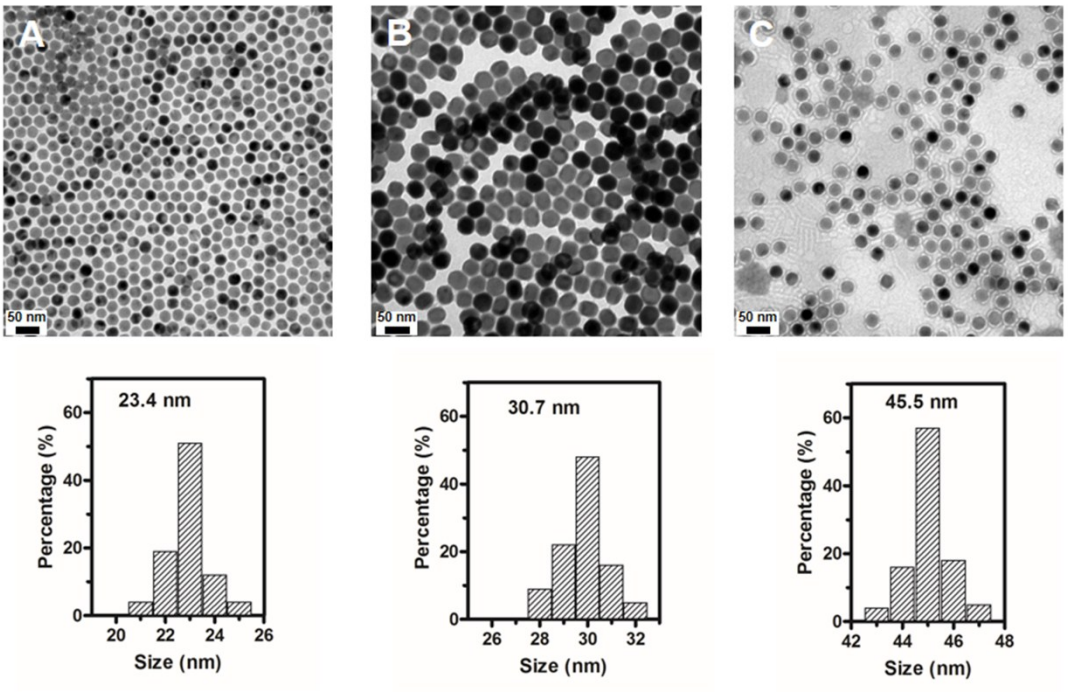

Figure S1 TEM images and size histograms of $NaYF_4$:Yb,Er nanoparticles (**A**), $NaYF_4$: Yb,Er@$NaGdF_4$ (UCNPs) nanoparticles (**B**) and PEGylated UCNPs (UPGs) micelles (**C**).

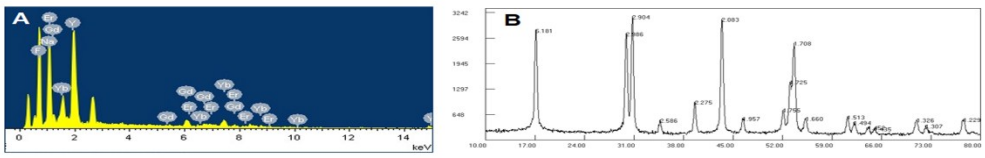

Fig. S2 (**A**) X-ray powder diffraction (XRD) and (**B**) emission diffraction X-ray (EDX) spectrum of $NaYF_4$:Yb,Er@$NaGdF_4$ nanoparticles (UCNPs).

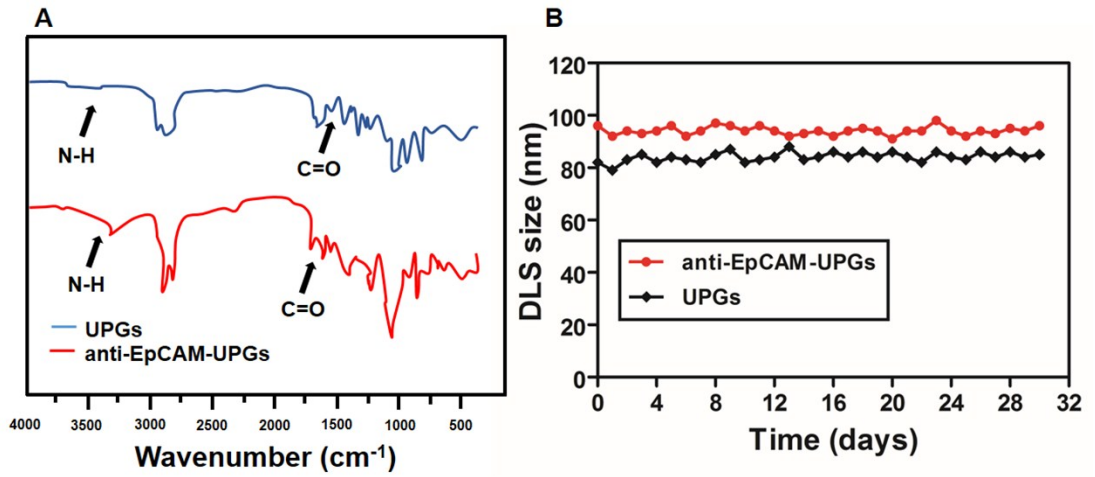

Figure S3 (**A**) Fourier transform infrared (FTIR) spectra of UPGs (blue line) and anti-EpCAM-UPGs micelles (red line). (**B**) Dynamic light scattering (DLS) size measurements of UPGs and anti-EpCAM-UPGs micelles dispersed in PBS for varied time durations (0 ~ 31 days).

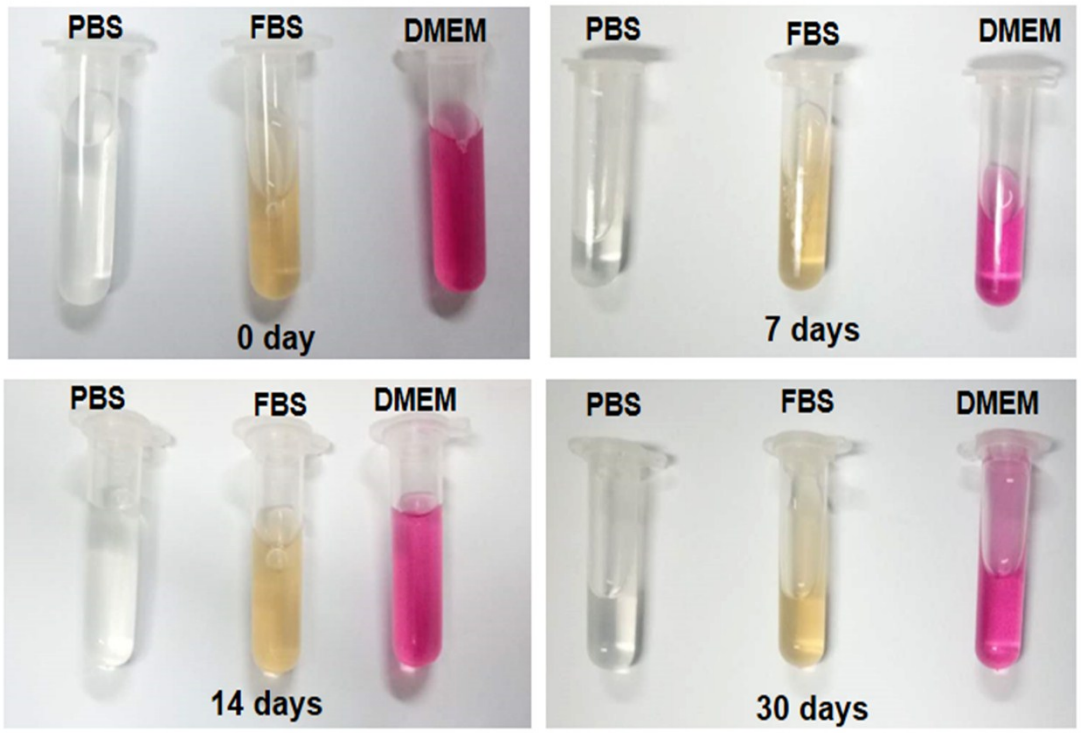

Fig. S4 Photographs of UPGs micelles in various solutions including phosphate buffered saline (PBS), fetal bovine serum (FBS), and DMEM cell medium with different time intervals.

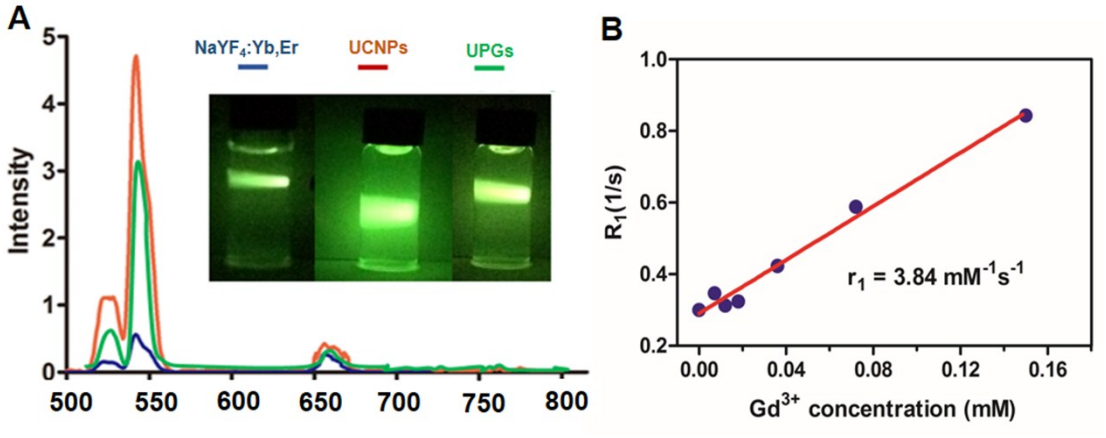

Figure S5 (**A**) Normalized upconversion luminescence spectra of $NaYF_4$:Yb,Er nanoparticles (blue line), UCNPs (red line) in cyclohexane, and UPGs micelles (green line) in PBS under 980 nm excitation. The insets are full-color photographs of the cyclohexane solutions of the NaYF4:Yb,Er (left), the UCNPs (middle) and aqueous solution of UPGs (right) taken in the dark under excitation of a 980 nm laser beam. (**B**) Plots of R_1 versus Gd^{3+} concentrations for UPGs in aqueous solution. The corresponding relaxivity value of UPGs is estimated to be r_1= 3.84 $mM^{-1}s^{-1}$.

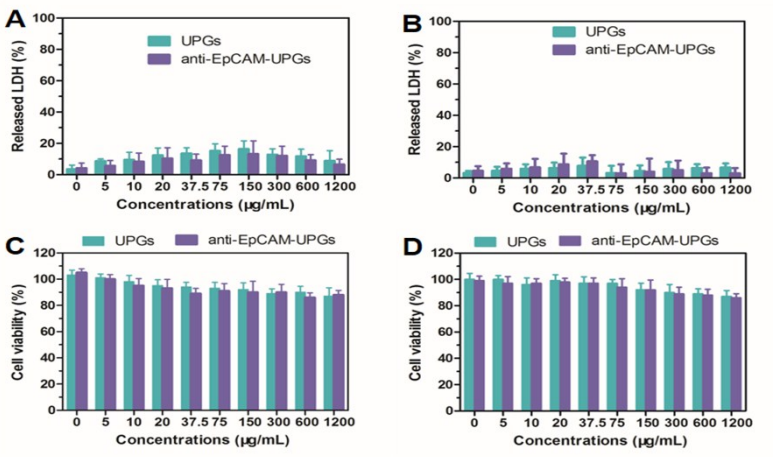

Fig. S6 *In vitro* toxicity evaluation of RAW 264.7 macrophages after coincubation with anti- UPGs/anti-EpCAM-UPGs micelles for (**A**) 24 h and (**B**) 48 h by lactate dehydrogenase (LDH) release assay. *In vitro* toxicity evaluation of RAW 264.7

macrophages after co-incubation with UPGs/anti-EpCAM-UPGs micelles for (**C**) 24 h and (**D**) 48 h by Cell Counting Kit-8 (CCK-8) method. All results show that UPGs and anti-EpCAM-UPGs micelles have little cytotoxicity to RAW 264.7 macrophages cells even with the high concentration of 1200 μg/mL.

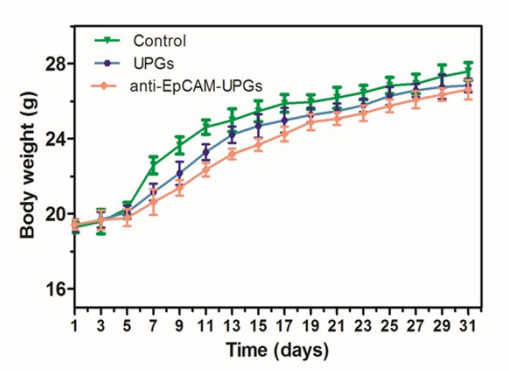

Fig. S7 Changes in body weight of mice treated with normal saline, UPGs and anti-EpCAM-UPGs micelles at different time points over time.

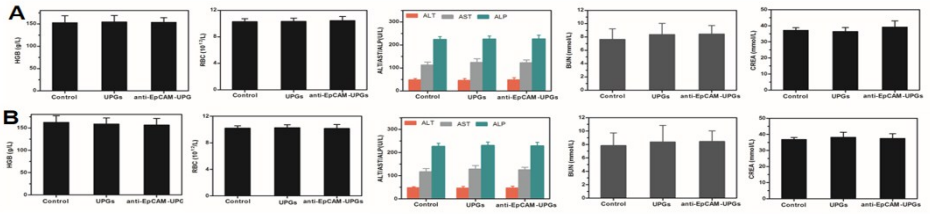

Fig. S8 *In vivo* toxicology study and serum biochemistry results obtained from balb/c nude mice after 7 and 31 days postintravenous injection with 150 μL physiological saline of UPGs (**A**) and anti-EpCAM-UPGs micelles (**B**) (40 mg/mL). Blood panel markers including: hemoglobin (HGB), red blood cells (RBC); liver function markers including: alanine aminotransferase (ALT), aspartate aminotransferase (AST), and alkaline phosphatase (ALP); kidney function markers including: blood urea nitrogen (BUN), and creatinine (CREA); untreated healthy mice were injected with 150 μL physiological saline via tail vein alone as the control. Statistics were based on six mice

per data point.

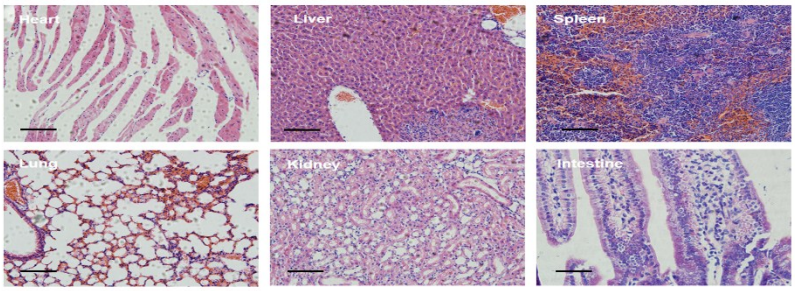

Fig. S9 H & E-stained tissue sections from major organs (heart, liver, spleen, lung, kidney and intestine) of mice. Mice were intravenously injected with 150 μL physiological saline of anti-EpCAM-UPGs-MX (40 mg/mL). No noticeable abnormality was observed in these major organs. A: heart; B: liver; C: spleen; D: lung; E: kidney; F: intestine. Scale bar: 40 μm.

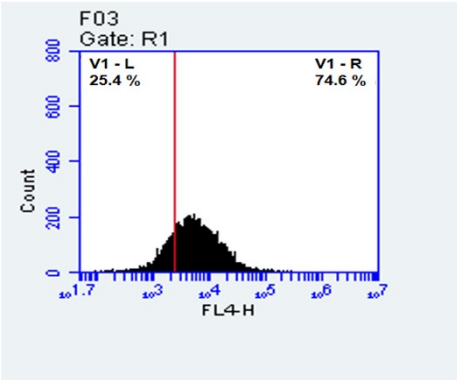

Fig. S10 Percentage of EpCAM positive BEL-7404 cells by flow cytometry.

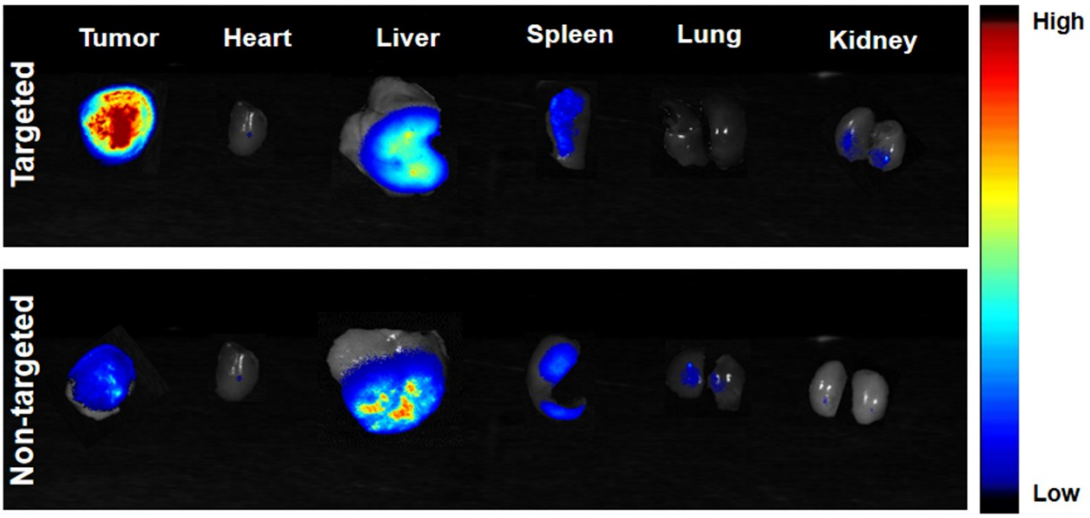

Fig. S11 Ex vivo upconversion luminescence (UCL) images of tumor tissue and various organs tissue from mice injected with anti-EpCAM-UPGs (i.e., Targeted) and UPGs (i.e., Nontargeted) micelles. The mice were sacrificed at 48 h postinjection.

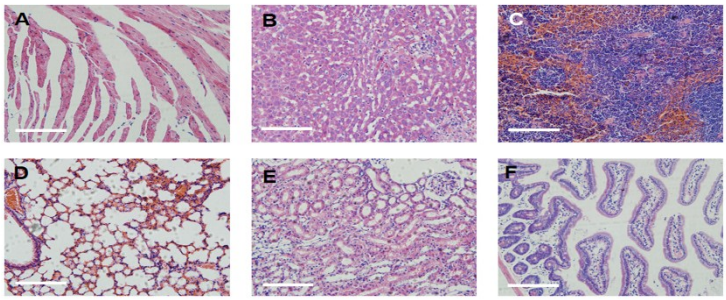

Fig. S12 Histological studies of tumors subject to different modes of therapies. The images are hematoxylin and eosin (H & E) sections from BEL-7404 tumor-bearing mice treated with anti-EpCAM-UPGs-MX + NIR irradiation. Scale bar: 40 μm.

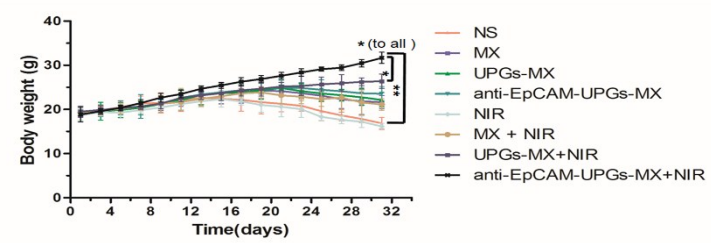

Fig. S13 Changes in body weight of mice treated with different interventions as a function of time.

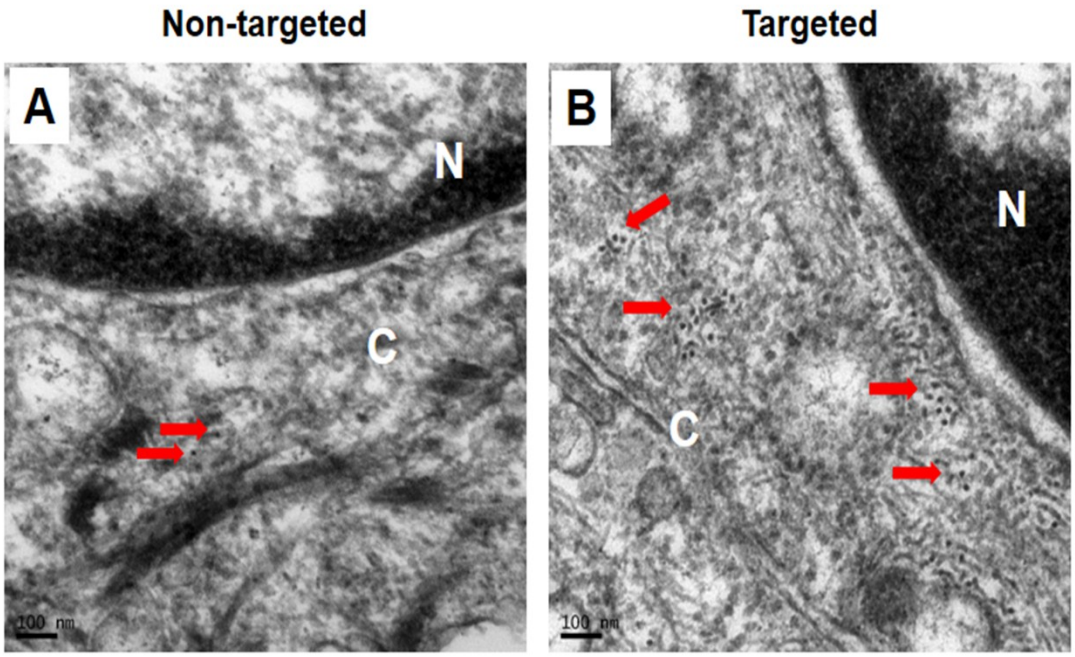

Fig. S14 Bio-TEM images of BEL-7404 tumor tissues of mice treated with UPGs (**A**) and anti-EpCAM-UPGs (**B**) micelles at 48 h post injection. N: nucleus, C: cytoplasm, red arrow: the micelles reside in the cytoplasm.