Supporting information:

Renally excretable silver telluride nanoparticles as contrast agents for x-ray imaging

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4 nm GSH-coated silver telluride nanoparticle synthesis:

The 4 nm core, GSH-coated Ag_2Te NPs were synthesized by preparing an aqueous solution as described above, followed by the addition of 1 mL of N_2H_4. The reaction was allowed to proceed for 15 minutes at room temperature under constant magnetic stirring. The synthesis was then quenched by placing the flask on an ice bath. The NPs were then washed, and suspended in PBS, as described above. The NPs were stored at 4 °C until further use.

Thermogravimetric analysis:

The thermal gravimetric analysis was conducted using a SDT Q600 TGA equipment (TA Instruments, New Castle, DE). Nitrogen gas was used to purge the instrument at a flow rate of 100 mL/min. The samples were heated up to 500 °C at a rate of 20 °C/min.

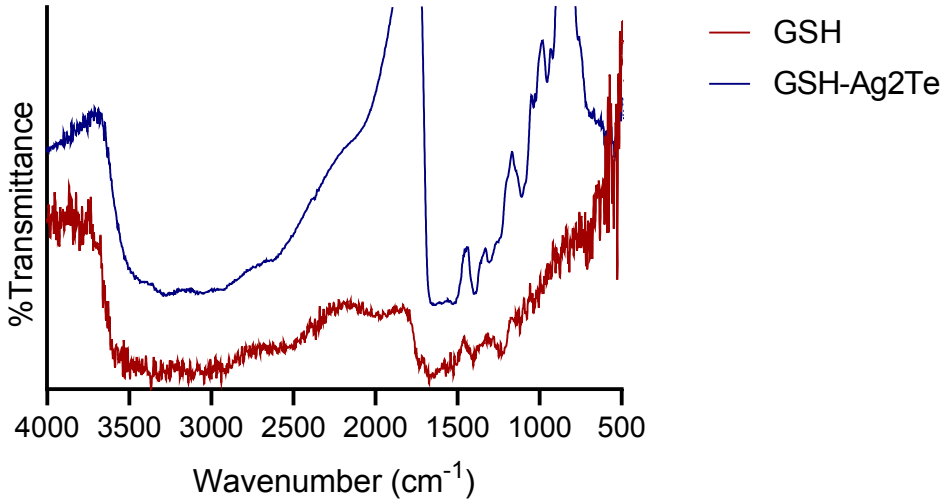
MTS assay:

MTS ((3-(4,5-dimethylthiazol-2-yl)-5-(3-carboxymethoxyphenyl)-2-(4-sulfophenyl)-2H-tetrazolium)) assay (Promega, Madison, WI) was used to further evaluate the cytotoxicity of GSH-Ag_2Te NPs according to the manufacturer's recommended protocol. In brief, the assay was performed in 96-well flat bottom plates with 20,000 cells per well seeded 24 hours before the experiment. The cells were incubated for 4 hours with GSH-Ag_2Te NPs at concentrations of 0, 0.1, 0.25, 0.5, or 1 mg of Ag per mL. After 4 hours of incubation, the treatment was removed and 20 µL of the MTS reagent and 100 µL of fresh media were added to each well and incubated for 30 minutes. A plate reader was then used to measure the absorbance at 490 nm. Three independent experiments were performed for each condition. The data is presented as mean cell viability relative to control ± standard deviation (n = 3).

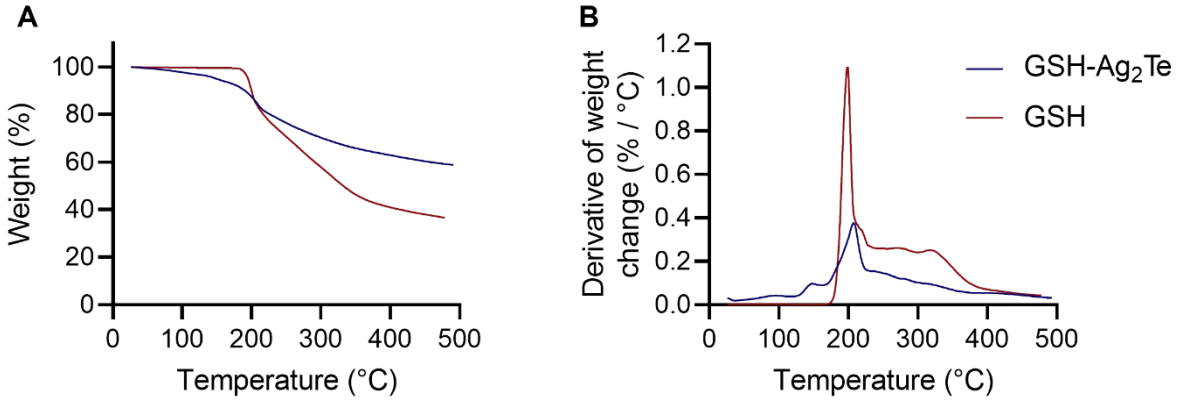
Table S1: Reactions for the synthesis of sub 5 nm Ag_2Te nanoparticles.

Coating	Reaction time	Temp.	Core size (nm)
GSH 30mM	10 min	RT	4 ± 1
	12 min	RT	3.6 ± 0.9
	13 min	RT	4 ± 1
	15 min	RT	4 ± 1
	Overnight	RT	5 ± 1
	5 min	Ice	3 ± 1
	10 min	Ice	4 ± 1
GSH 60 mM	15 min	RT	2 ± 1
3-MPA 30 mM	10 min	RT	3 ± 2
	12 min	RT	5.5 ± 0.7
	13 min	RT	4 ± 1
	15 min	RT	7 ± 2
	15 min	Ice	2.1 ± 0.7
	10 min	Ice	4 ± 1
	5 min	Ice	3 ± 1
3-MPA 90 mM	15 min	RT	2 ± 1
	15 min	Ice	1.8 ± 0.7
	10 min	Ice	8 ± 1

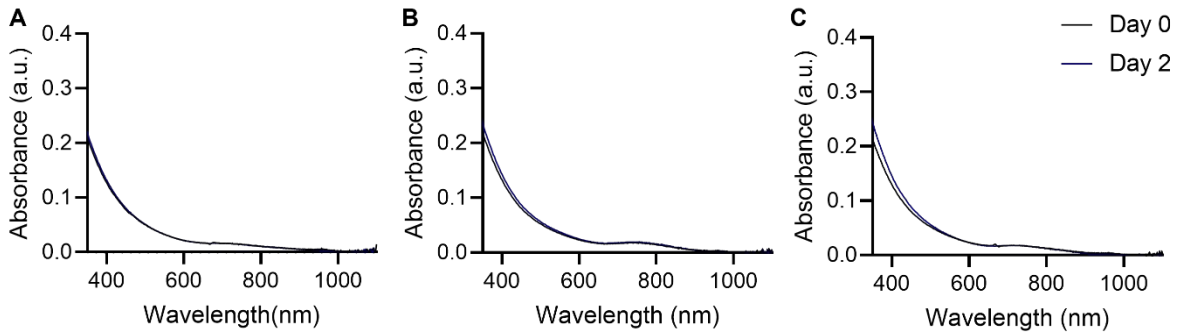
RT = room temperature; Ice = ice bath.



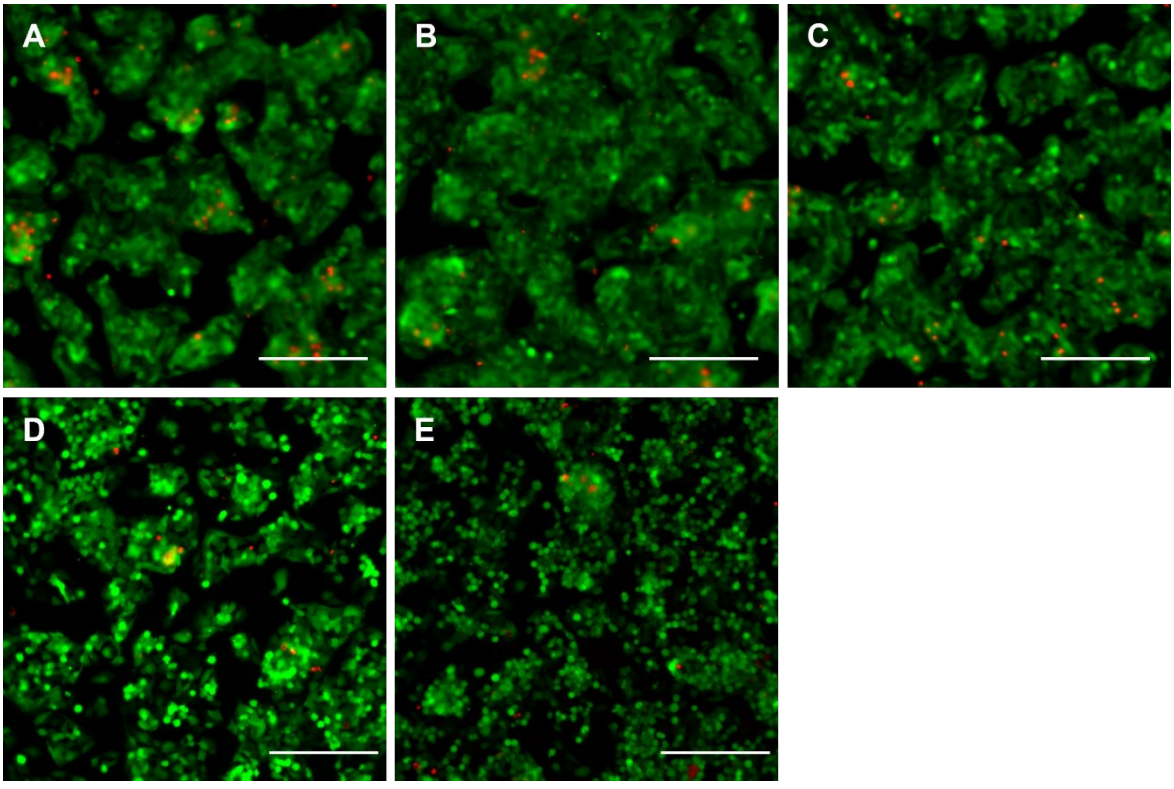
Supplemental Figure 1: FT-IR spectra of GSH and GSH-coated Ag_2Te nanoparticles.



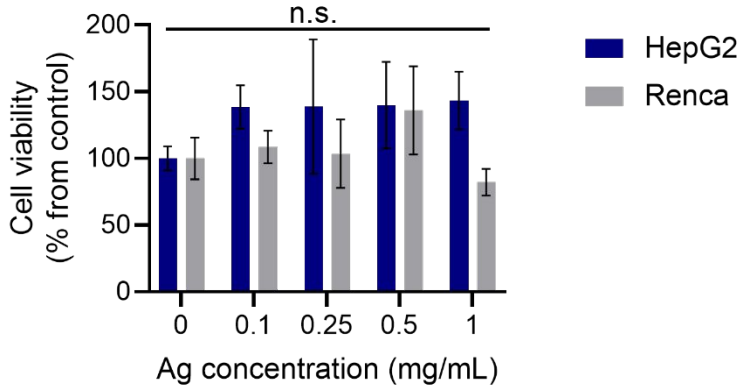
Supplemental Figure 2: Thermo-gravimetric analysis of GSH-Ag_2Te (blue) and GSH (red). A) Weight (%) change and B) derivative of weight change (% / °C) of the two samples as the temperature was increased to 500 °C.



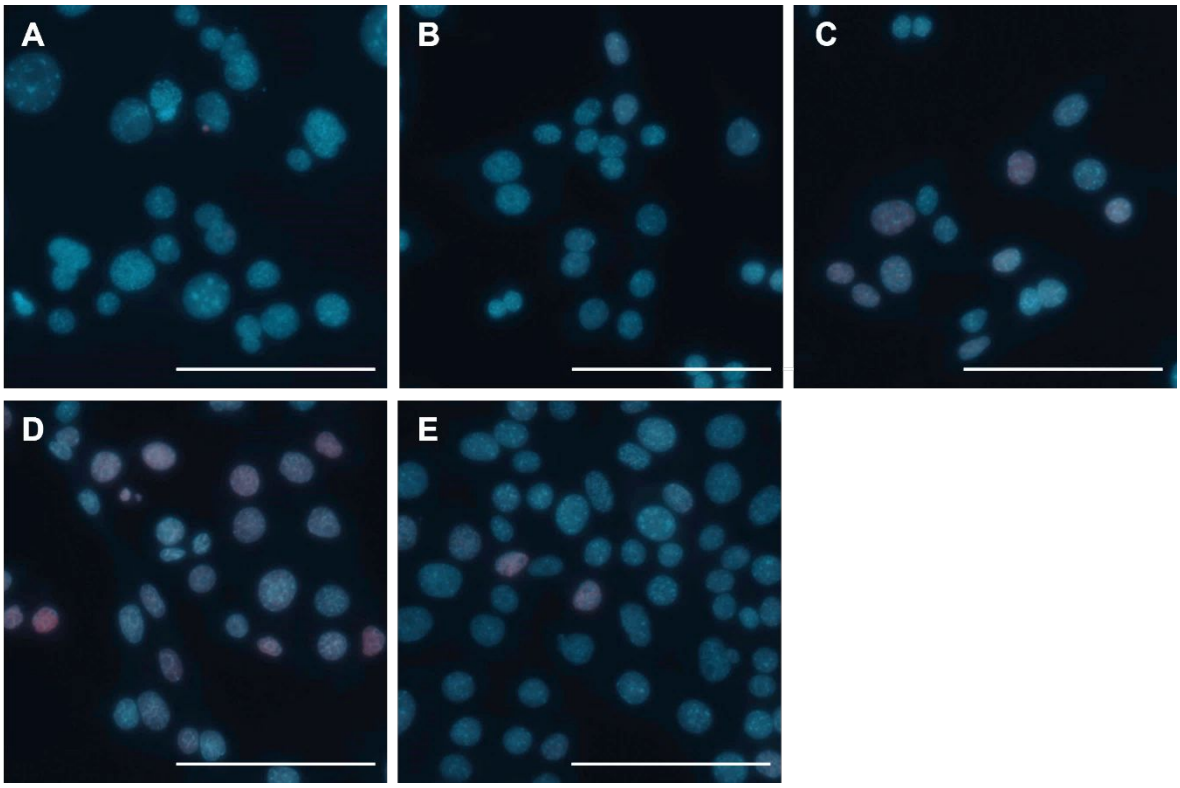
Supplemental Figure 3: GSH-Ag_2Te nanoparticle stability over time. UV-vis of GSH-Ag_2Te nanoparticles dispersed in A) water, B) PBS, and C) PBS + 10% FBS.



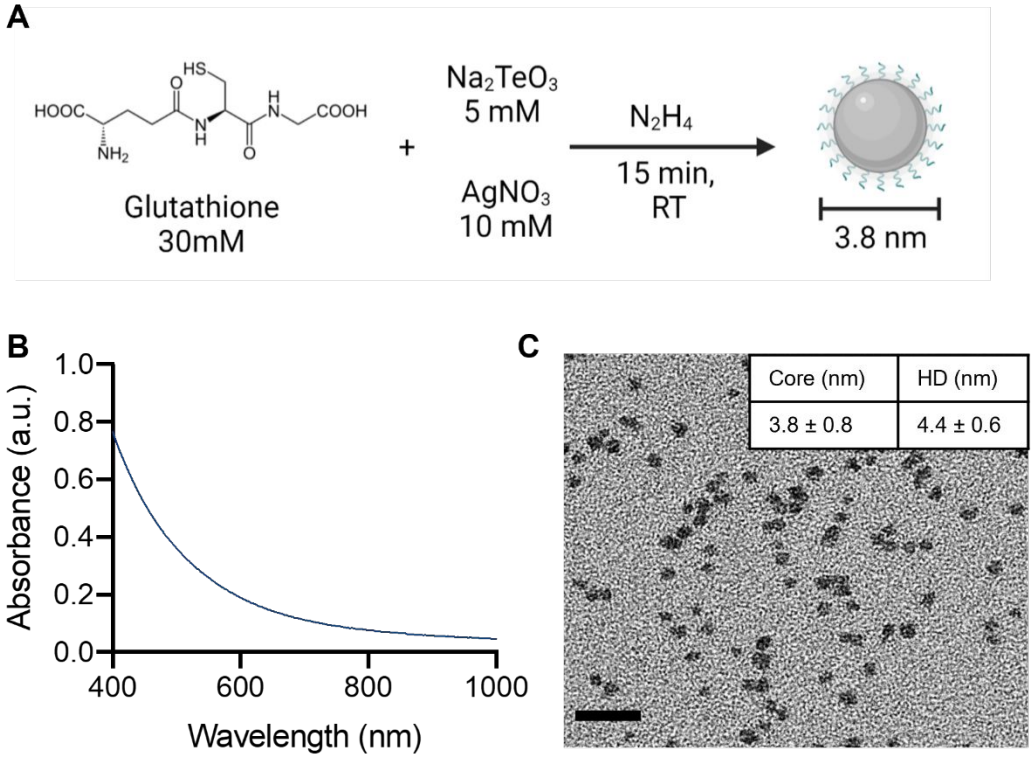
Supplemental Figure 4: Representative micrographs of HepG2 cells LIVE/DEAD assay at a GSH-Ag_2Te nanoparticle concentration of A) 0, B) 0.1, C) 0.25, D) 0.5, and E) 1 mg of Ag per mL. Scale bar = 200 µm; green = live cells; red = dead cells.



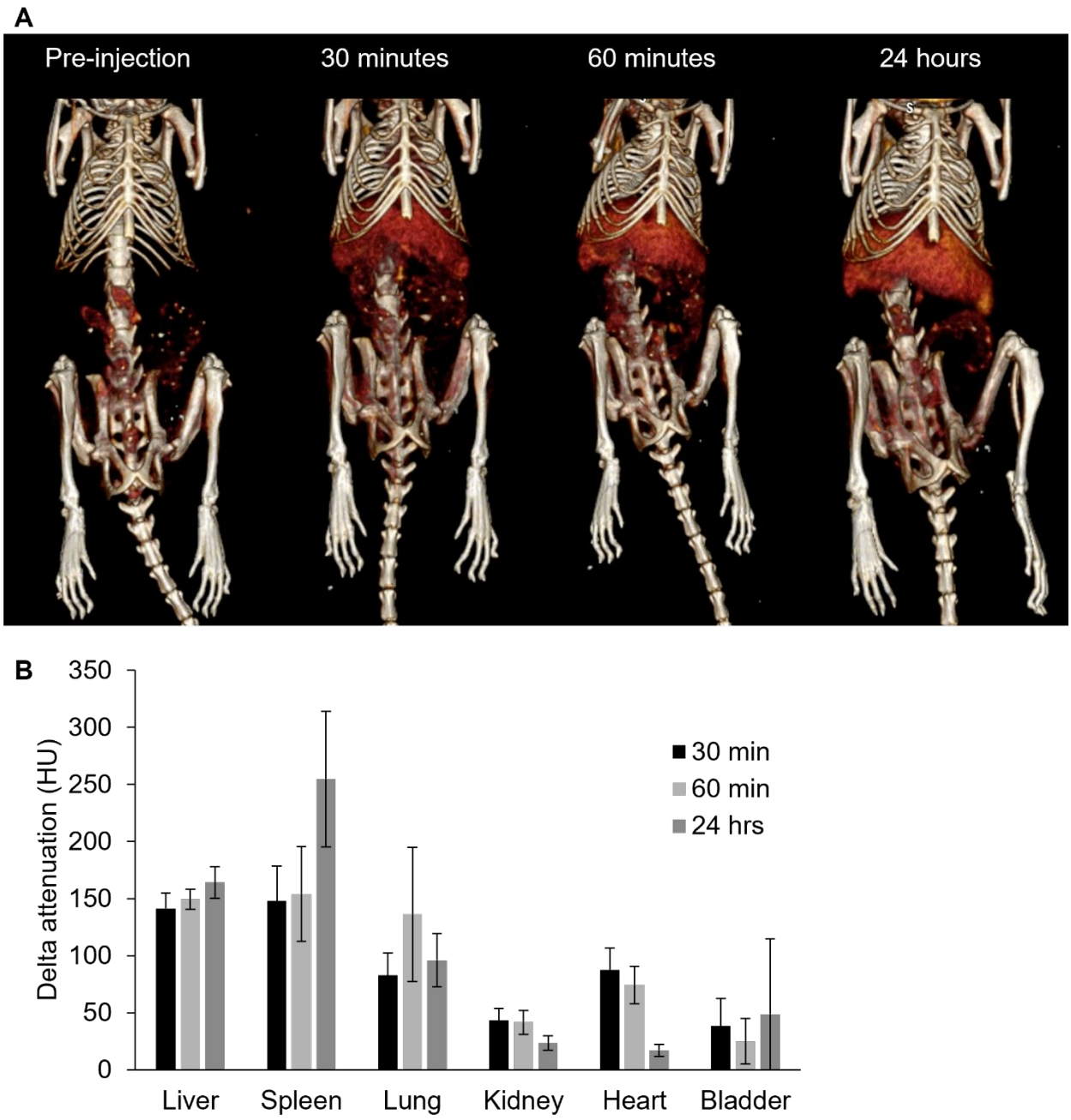
Supplemental Figure 5: MTS assay of HepG2 and Renca cells after 4 hours of incubation with GSH-Ag_2Te nanoparticles at different concentrations. Data presented as mean ± SD.



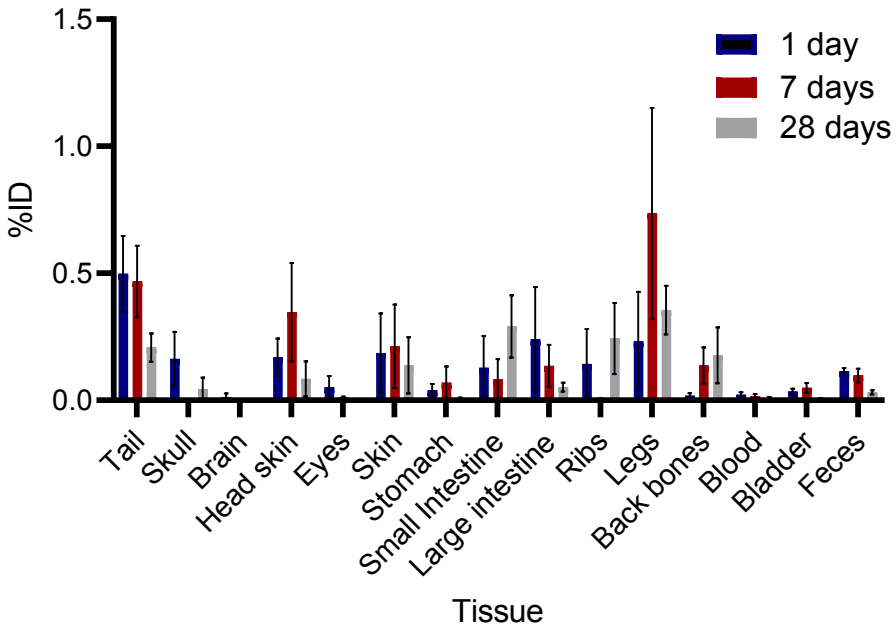
Supplemental Figure 6: Representative micrographs of γH2AX immunostaining of Renca cells after 4 hours of incubation with A) 0, B) 0.25, C) 0.5 or D) 1 mg of Ag per mL of GSH-Ag_2Te NPs.Renca cells were irradiated (6 Gy dose) as a positive control (E) for DNA damage. Scale bar = 100 μm. Blue = nuclei and red = γH2AX. Quantification was done in unedited images.



Supplemental Figure 7: Control GSH-Ag_2Te nanoparticle synthesis schematic and characterization: A) Nanoparticle synthesis schematic. B) UV-vis spectrum of control GSH-coated Ag_2Te NPs. C) TEM micrograph of control GSH-coated Ag_2Te NPs. Inset shows the core size and hydrodynamic diameter (HD) data for control GSH-coated Ag_2Te NPs.



Supplemental Figure 8: A) Representative CT images of 4 nm GSH-coated Ag_2Te NPs injected mice pre-injection, 30 min, 60 min, and 24 hours post-injection. B) Quantification of the CT attenuation in the different organs of mice injected with 4 nm GSH-Ag_2Te NPs at different time points (pre-injection, 30 min, 60 min, and 24 hours post-injection). n=5 per group. Data is presented as mean ± SEM.



Supplemental Figure 9: Biodistribution of GSH-Ag_2Te nanoparticles in other tissues at 1 day, 7 days, and 28 days post-injection. Data is presented as mean ± SEM.

Supplemental Figure 10: Photograph of GSH-Ag_2Te nanoparticle solution.