

Supplementary Materials for

GSH depletion liposome adjuvant for augmenting the photothermal immunotherapy of breast cancer

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Figs. S1 to S9

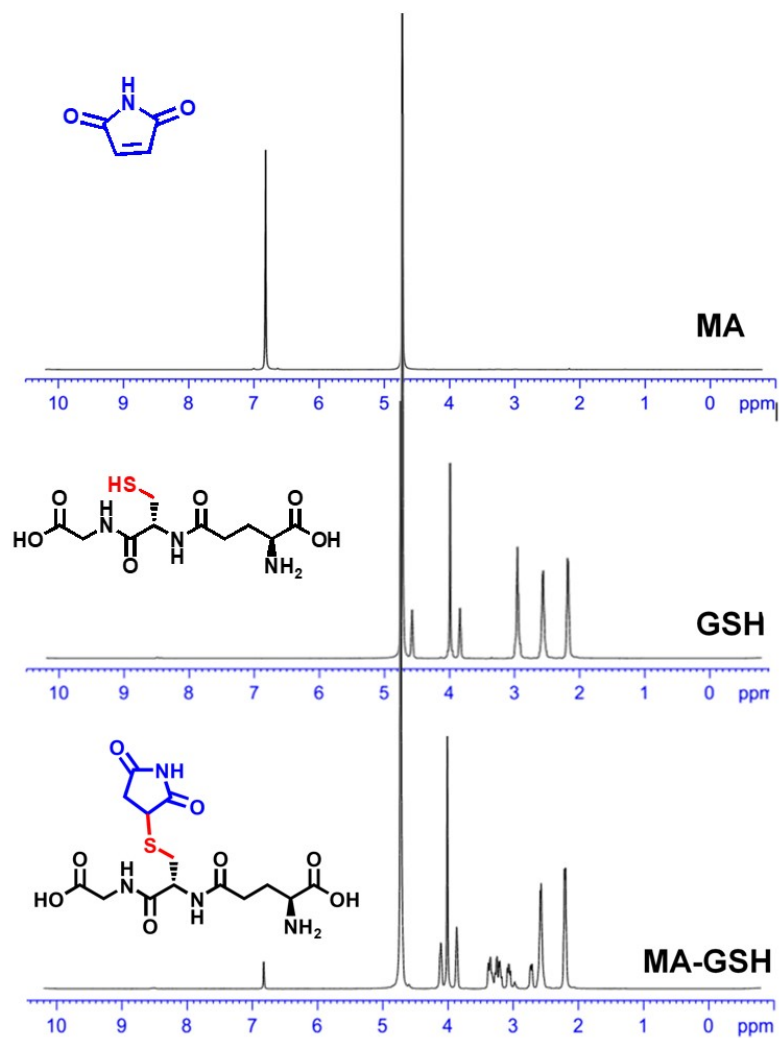


Fig. S1. ¹H-NMR spectrum of MA, GSH and MA-GSH (solvent: D₂O).

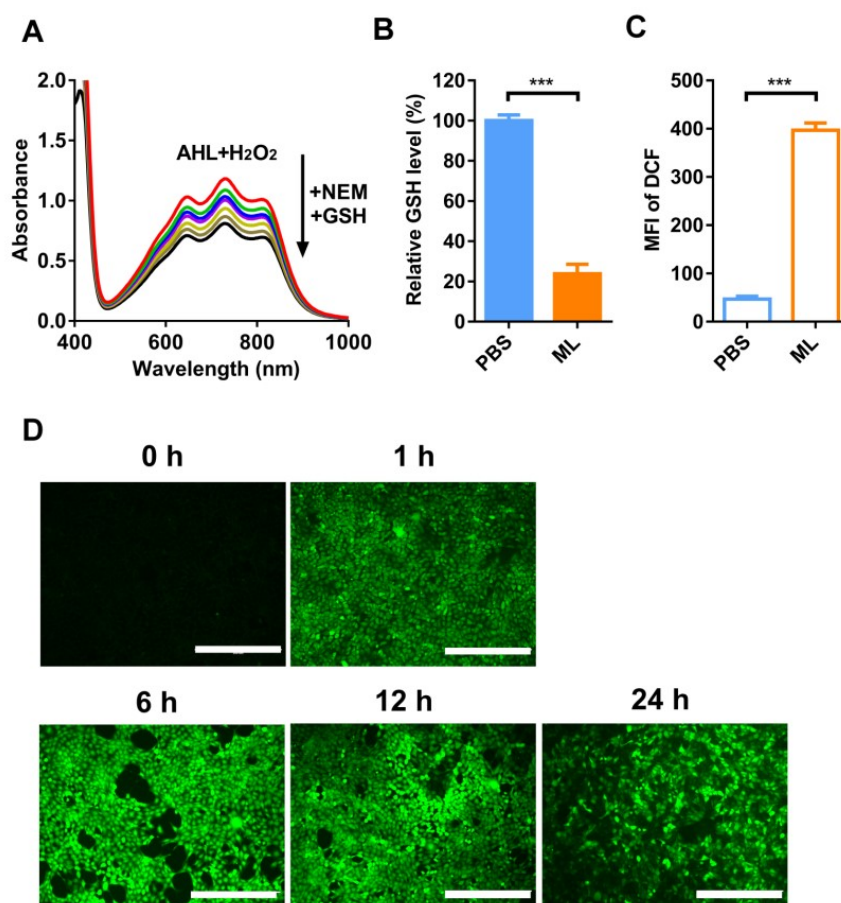


Fig. S2. GSH and ROS modulation. (A) Absorbance changing of AHL+H₂O₂+NEM in the presence of increasing GSH. (B) Statistic analysis of the relative GSH level in 4T1 cells after treatment with PBS or ML (n = 3). ****P*<0.001. (C) Statistic analysis of the MFI of DCF in 4T1 cells after treatment with PBS or ML (n = 3). ****P*<0.001. (D) Intracellular ROS monitoring in 4T1 cells after treatment with ML. Scale bar: 400 μ m.

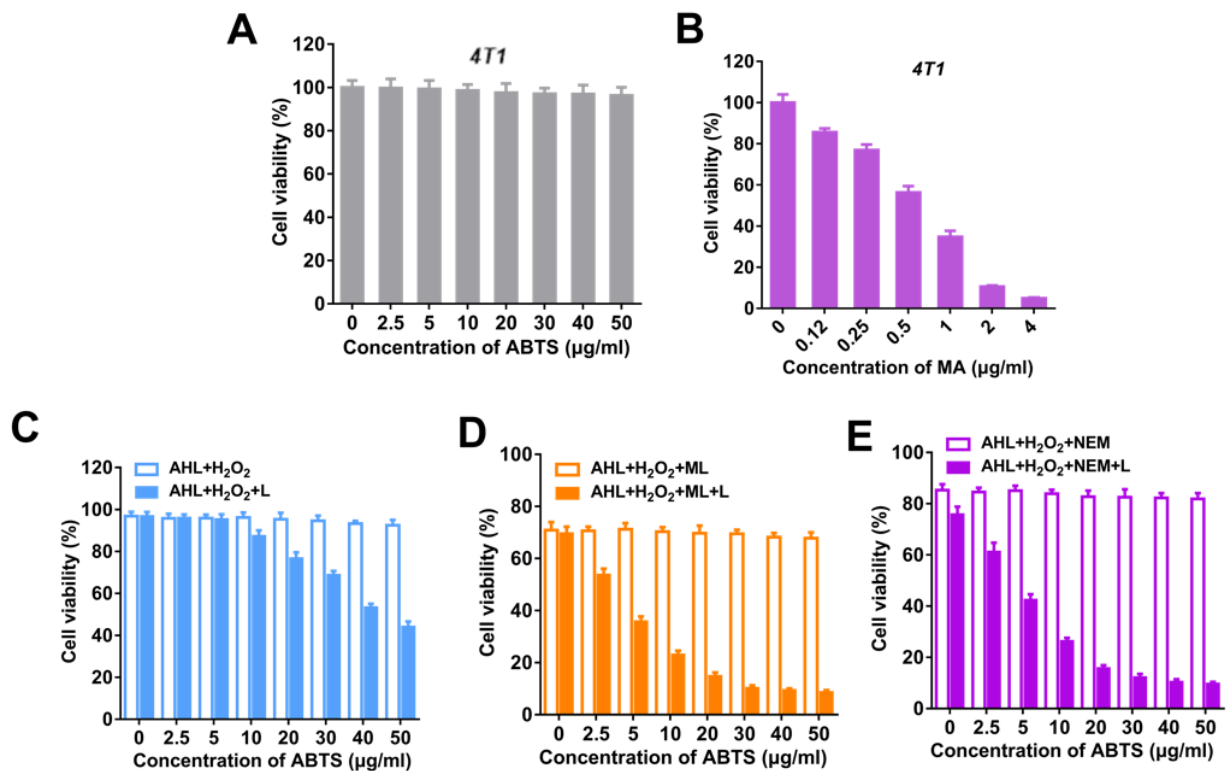


Fig. S3. Cell viability determination of 4T1 cells after various treatment. Cell viability of 4T1 cells after treated with (A) AHL or (B) ML by MTT assay (n=3). Cell viability of 4T1 cells after treated with (C) AHL+H₂O₂, (D) AHL+H₂O₂+ML or (E) AHL+H₂O₂+NEM in the presence or absence of laser irradiation by MTT assay (n=3).

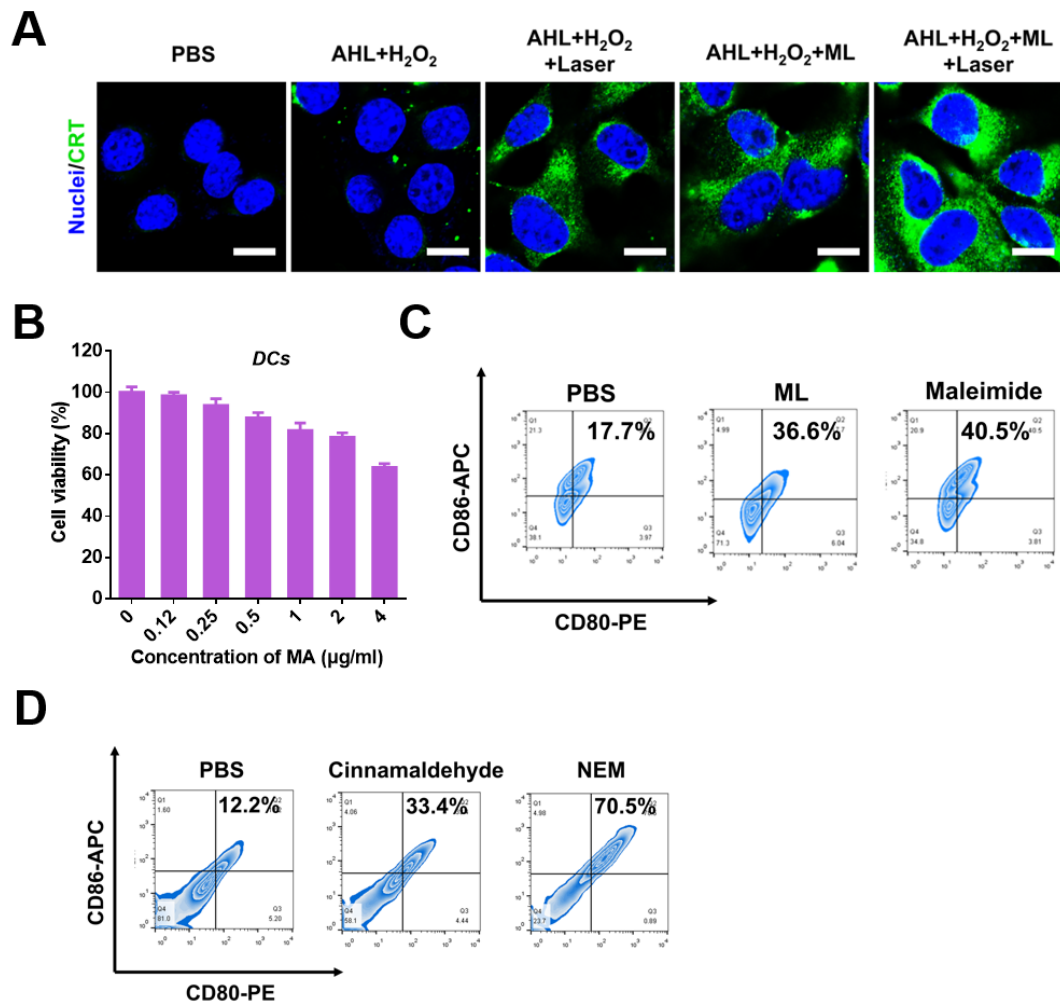


Fig. S4. Immune analysis in vitro. (A) CRT exposure of 4T1 cells after different treatments. FITC labelled CRT was indicated as green fluorescence. Nuclei was stained by DAPI and shown as blue fluorescence. Scale bar: 10 μm . (B) Cell viability of DC cells after treated with ML by MTT assay (n=3). (C) DCs maturation after incubation with maleimide or maleimide liposomes (ML) (gated on CD11c⁺ cells). (D) DCs maturation after incubation with cinnamaldehyde or N-ethylmaleimide (NEM) (gated on CD11c⁺ cells).

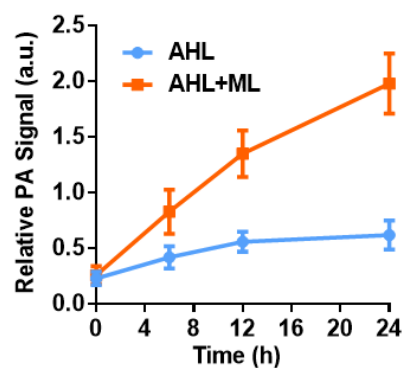


Fig. S5. The relative PA signal analysis of 4T1 tumor after i.v. injection of AHL or AHL+ML (n = 3)

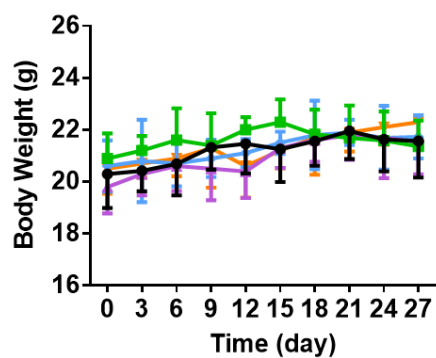


Fig. S6. Body weight monitoring during treatments (n = 5).

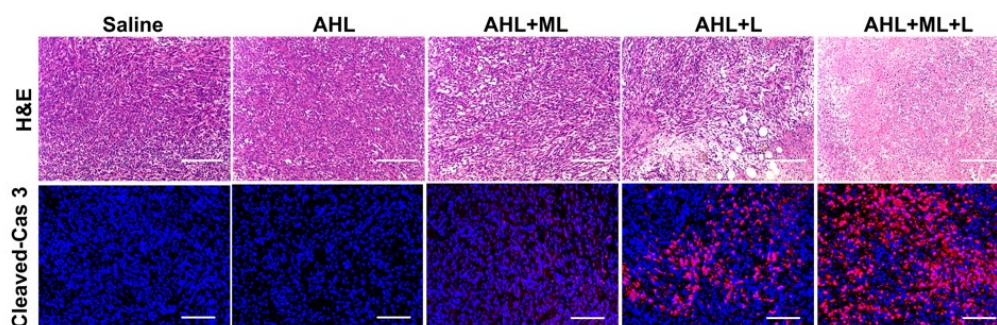


Fig. S7. H&E staining and cleaved-caspase 3 level analysis of tumor tissues on day 5 post treatment. Scale bar of H&E staining: 200 μ m, scale bar of cleaved-caspase 3 analysis: 100 μ m.

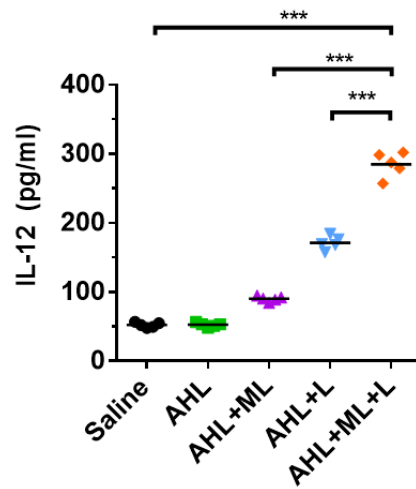


Fig. S8. Contents of the IL-12 in plasma on day 5 after treatment. Data represent mean \pm SD (n = 5). *** P <0.001.

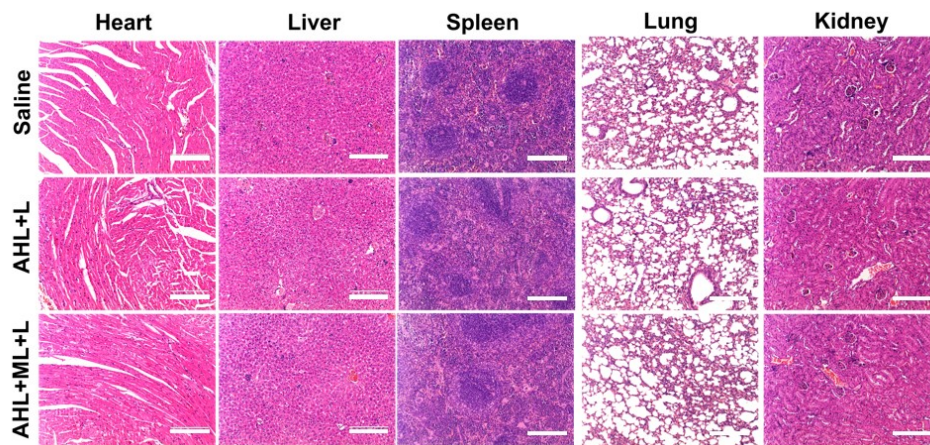


Fig. S9. Histological images of the HE-stained heart, liver, spleen, lung, and kidney harvested from the mice after treatment on day 5. Scale bar: 200 μ m.