

## Supporting Information

### **Iron-Based Nanoparticles for MR Imaging-Guided Ferroptosis in Combination with Photodynamic Therapy to Enhance Cancer Treatment**

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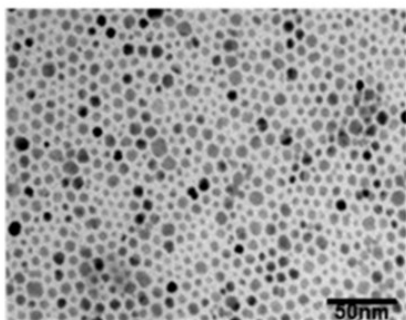
#These authors contributed equally

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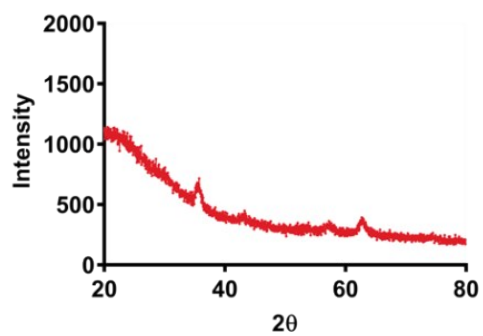
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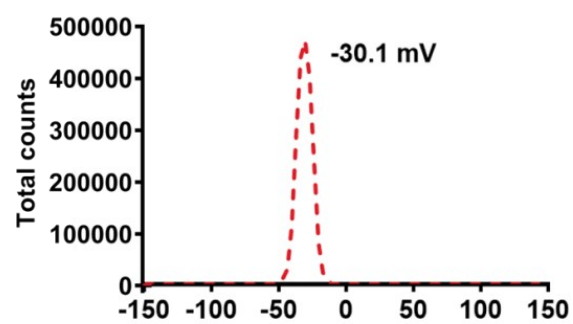
(A)



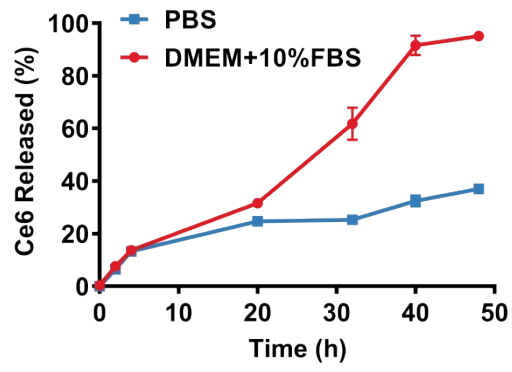
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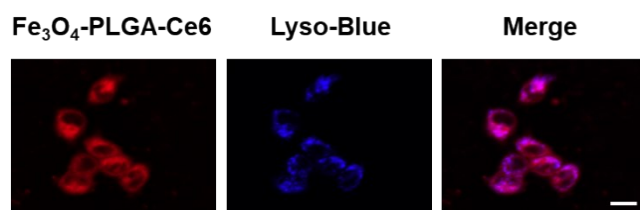
**Figure S1.** Characterization of citric acid (CA) coated  $\text{Fe}_3\text{O}_4$  NPs. (A) TEM image of CA-coated  $\text{Fe}_3\text{O}_4$  NPs (scale bars: 50 nm). (B) XRD measurement of CA-coated  $\text{Fe}_3\text{O}_4$  NPs.



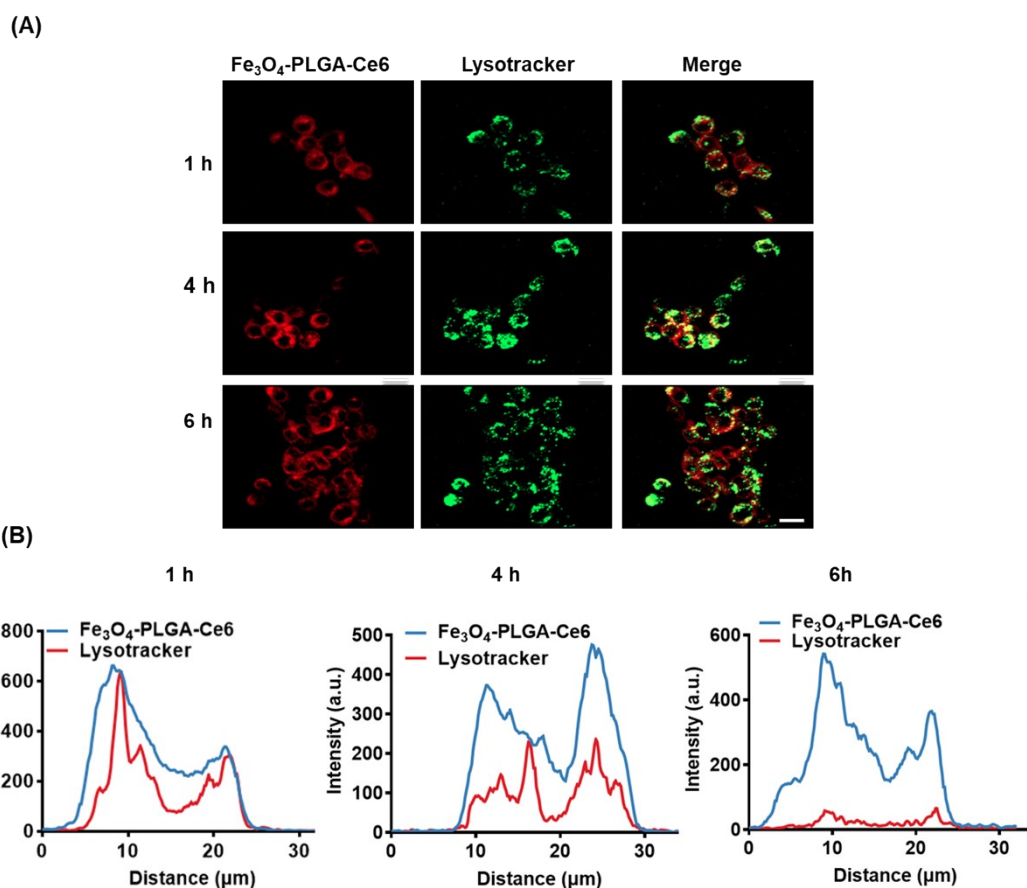
**Figure S2.** The zeta potential of Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs.



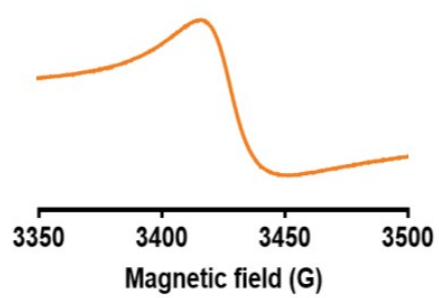
**Figure S3.** Release profiles of Ce6 from Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs in different cultivation environment (PBS, DMEM+10% FBS) with the time, n=3.



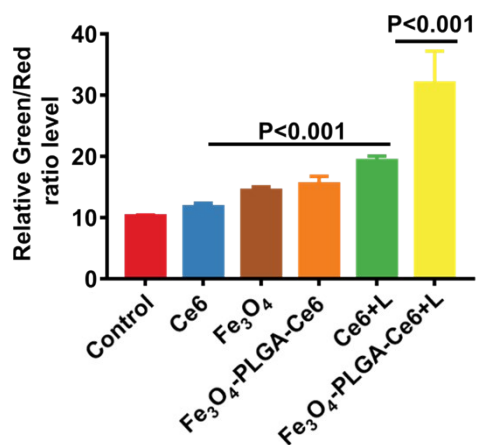
**Figure S4.** CLSM images of 4T1 cells after 1 h incubation with Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs. Lysosome was stained with LysoTracker Blue. Scale bar: 10  $\mu$ m. Drug dose: Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 10  $\mu$ g mL<sup>-1</sup> Ce6.



**Figure S5.** (A) CLSM of lysosomal escape in different time for 1, 2, and 4 h (Red fluorescent dots indicated Ce6 from Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs, green fluorescent dots indicated lysosome). (B) The line chart analysis of lysosomal escape at 1 h, 4 h and 6 h by single cell scanning. Scale bars: 10 μm. Drug dose: Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 10 μg mL<sup>-1</sup> Ce6.

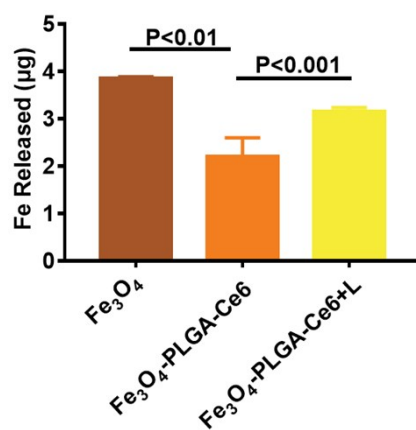


**Figure S6.** The EPR spectra of  $\text{Fe}_3\text{O}_4$ -PLGA-Ce6 NPs in PBS (pH 7.4) solution.

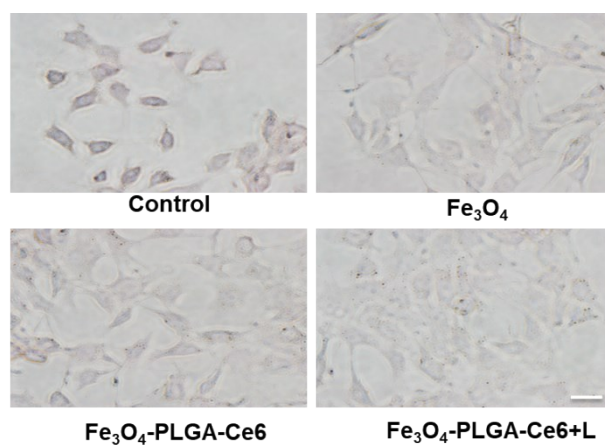


**Figure S7.** 4T1 cells were treated with Ce6, Fe<sub>3</sub>O<sub>4</sub>, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs, Ce6+L, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs +L for 24 h. The FITC/PI (Green/Red) ratio level was determined after staining with JC-1 probe by flow cytometry analysis, which indicated the mitochondrial damage, n=3. L represents laser. Drug dose: Ce6 10 μg mL<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub> 12.5 μg mL<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 10 μg mL<sup>-1</sup> Ce6, Ce6 plus laser 10 μg mL<sup>-1</sup> and Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs plus laser with 10 μg mL<sup>-1</sup> Ce6.

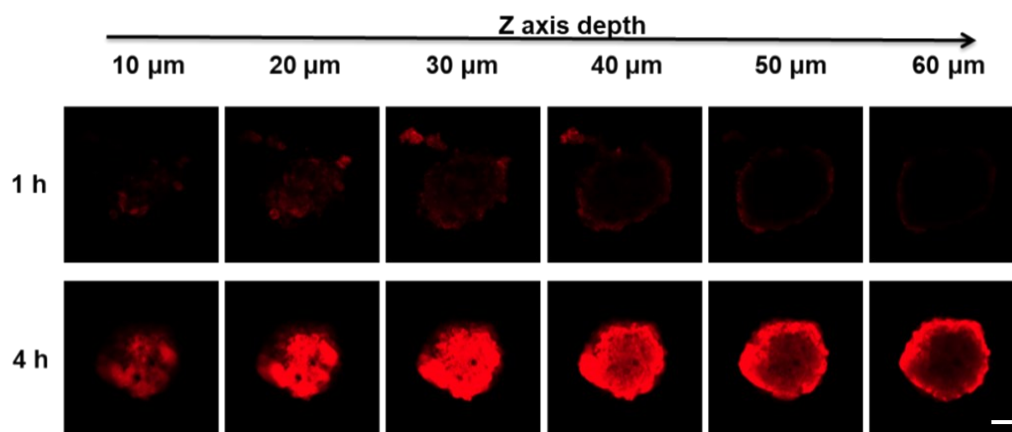




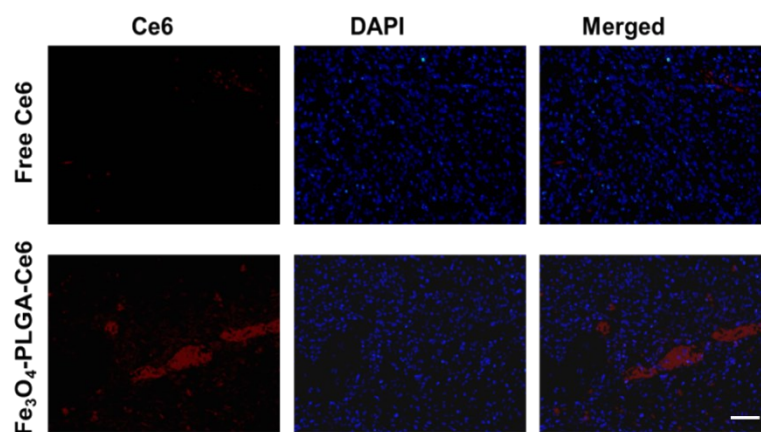
**Figure S8.** AAS detected iron ions released from nanoparticles in different formulas such as Fe<sub>3</sub>O<sub>4</sub>, Fe<sub>3</sub>O<sub>4</sub>+L, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 and Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6+L, L represents laser, n=3. Drug dose: Fe<sub>3</sub>O<sub>4</sub> 12.5 µg mL<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub> 12.5 µg mL<sup>-1</sup> plus laser, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 12.5 µg mL<sup>-1</sup> Fe<sub>3</sub>O<sub>4</sub>, and Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 12.5 µg mL<sup>-1</sup> Fe<sub>3</sub>O<sub>4</sub> plus laser.



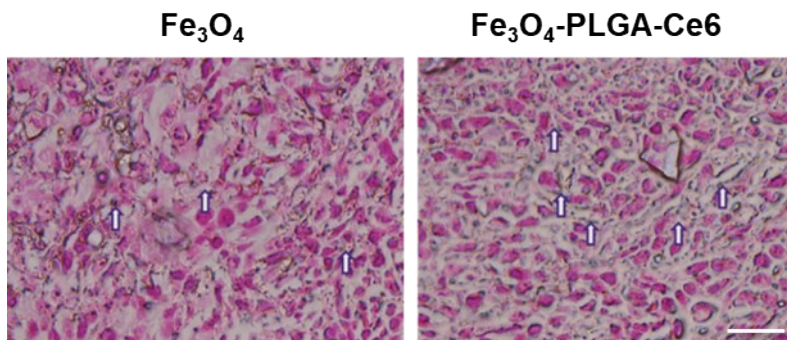
**Figure S9.** Lillie ferrous ion staining of different formulas in 4T1 cells. Blue foci indicated the ferrous iron. L represents laser. Scale bars: 10  $\mu\text{m}$ . Drug dose: Fe<sub>3</sub>O<sub>4</sub> 12.5  $\mu\text{g mL}^{-1}$ , Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 12.5  $\mu\text{g mL}^{-1}$  Fe<sub>3</sub>O<sub>4</sub> and Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 +L NPs with 12.5  $\mu\text{g mL}^{-1}$  Fe<sub>3</sub>O<sub>4</sub>.



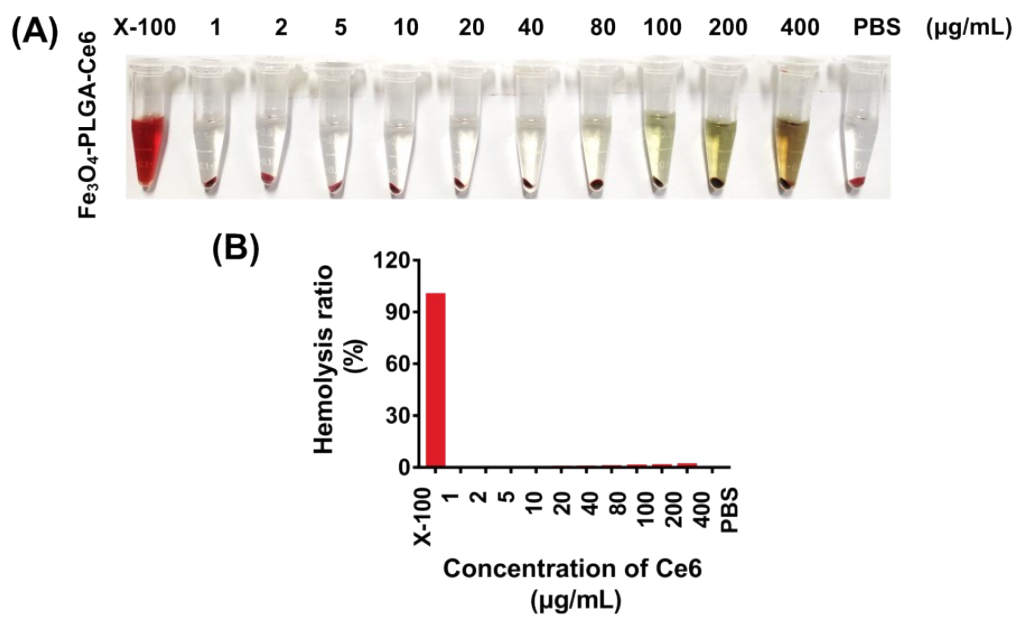
**Figure S10.**  $\text{Fe}_3\text{O}_4$ -PLGA-Ce6 NPs of tumor penetration study. Drug dose:  $\text{Fe}_3\text{O}_4$ -PLGA-Ce6 NPs with  $10 \mu\text{g mL}^{-1}$  Ce6. Scale bars:  $200 \mu\text{m}$ .



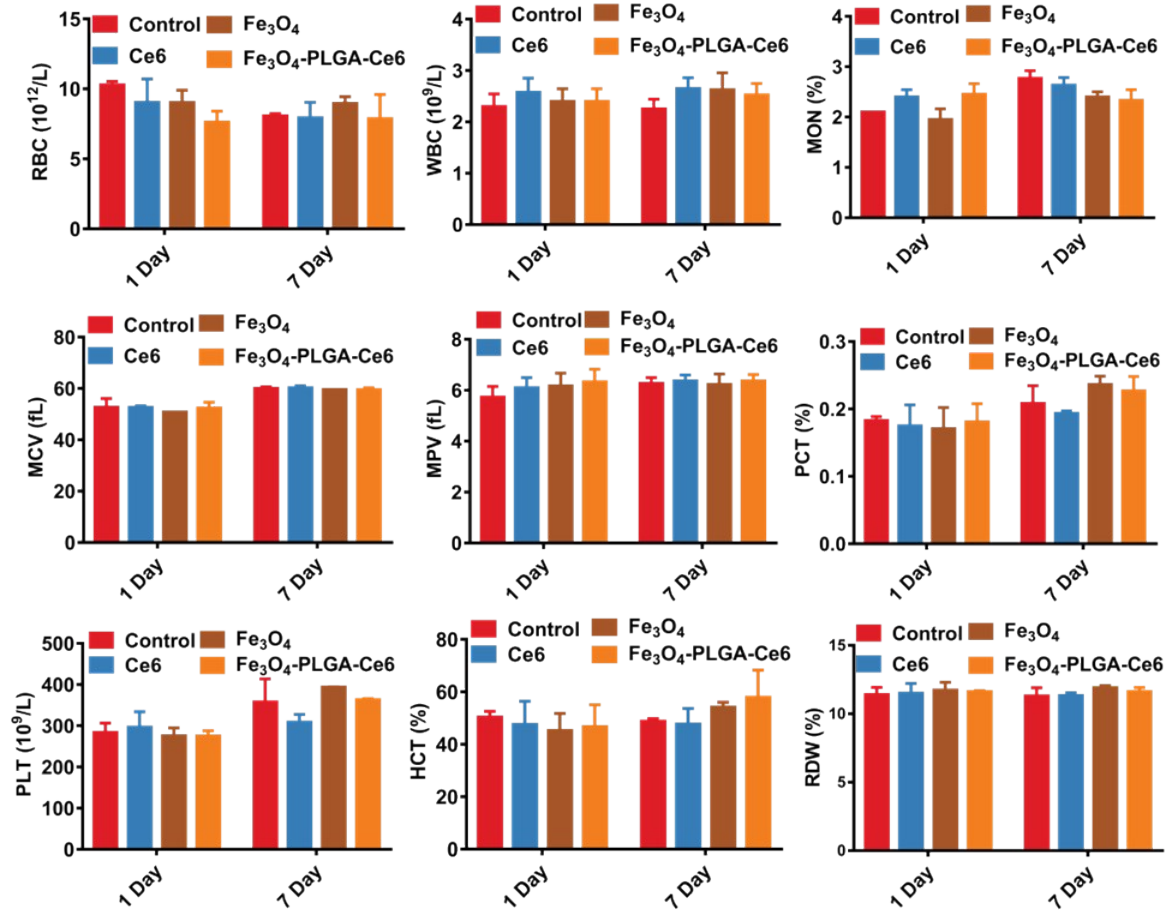
**Figure S11.** Accumulation of Ce6 in *ex vivo* tumor tissues treated with free Ce6 or Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs. Blue (DAPI), Red (Ce6). Scale bars:50  $\mu$ m. Drug dose: Ce6 5 mg kg<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub> 6.25 mg kg<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 5 mg kg<sup>-1</sup> Ce6.



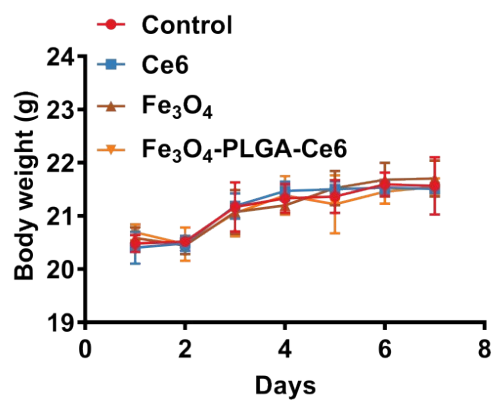
**Figure S12.** Iron ions accumulation in *ex vivo* tumor tissues treated with free  $\text{Fe}_3\text{O}_4$  or  $\text{Fe}_3\text{O}_4\text{-PLGA-Ce6}$  NPs. Blue ( $\text{Fe}^{2+}$ ), Red (Nuclei). Scale bars: 50  $\mu\text{m}$ . Drug dose:  $\text{Fe}_3\text{O}_4$  6.25  $\text{mg kg}^{-1}$ ,  $\text{Fe}_3\text{O}_4\text{-PLGA-Ce6}$  NPs with 6.25  $\text{mg kg}^{-1}$   $\text{Fe}_3\text{O}_4$ . The white arrow indicated the ferrous iron.



**Figure S13.** Hemolysis analysis. (A) The photos of hemolysis assay treated with  $\text{Fe}_3\text{O}_4\text{-PLGA-Ce6}$  NPs at different concentrations. (B) Hemolysis ratios of  $\text{Fe}_3\text{O}_4\text{-PLGA-Ce6}$  NPs with different Ce6 concentration.

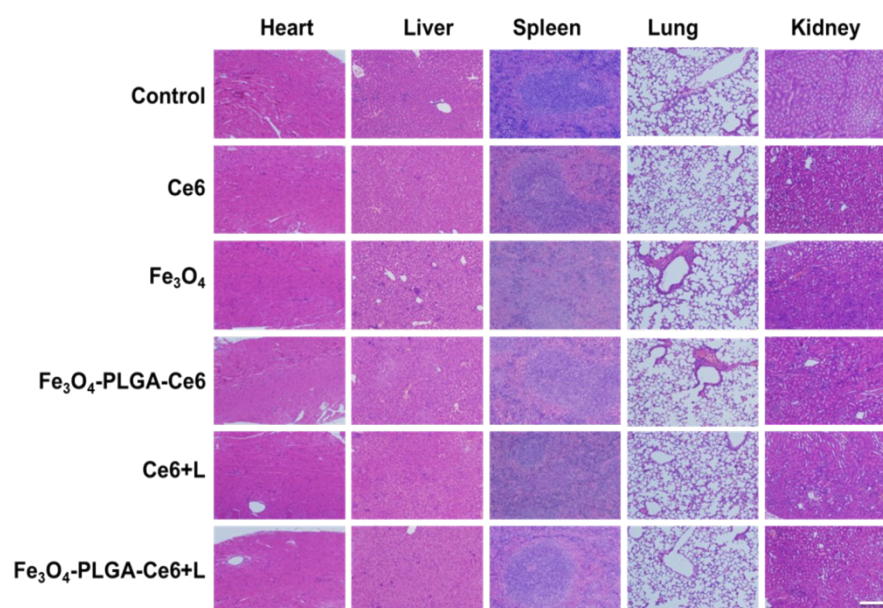


**Figure S14.** Nine blood routine index analysis for 4T1 tumor-bearing mice treated with different formulas after 1 day and 7 days. Drug dose: Ce6 5 mg kg<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub> 6.25 mg kg<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 5 mg kg<sup>-1</sup> Ce6. The group intravenously injected with PBS was defined as the negative control. n=4.



**Figure S15.** *In vivo* body weight curve of the hematological assays of BALB/c mice from 1 to 7 days post-treatment with different formulas (n=4). Drug dose: Ce6 5 mg kg<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub> 6.25 mg kg<sup>-1</sup>, Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 5 mg kg<sup>-1</sup> Ce6. The group intravenously injected with PBS was defined as the negative control.





**Figure S16.** Histological observation of 4T1-bearing mice organs with H&E staining after 12 days treatment with different formulas. (Scale bars: 150  $\mu\text{m}$ ). Drug dose: Ce6 5  $\text{mg kg}^{-1}$ , Fe<sub>3</sub>O<sub>4</sub> 6.25  $\text{mg kg}^{-1}$ , Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs with 5  $\text{mg kg}^{-1}$  Ce6, Ce6 plus laser 5  $\text{mg kg}^{-1}$  and Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 NPs plus laser with 5  $\text{mg kg}^{-1}$  Ce6. The group intravenously injected with PBS was defined as the negative control.

**Table S1.** Loading data of Fe<sub>3</sub>O<sub>4</sub>-PLGA-Ce6 nanoparticles.

<b>NPs</b>	<b>Drug</b>	<b>Encapsulation efficiency (%)</b>	<b>Loading content (%)</b>
Fe <sub>3</sub> O <sub>4</sub> -PLGA-Ce6	Fe <sub>3</sub> O <sub>4</sub>	70.1±2.5	25.5±0.4
	Ce6	75.5±3.7	22.1±1.1