

# Image-Guided Enhanced PDT/PTT Combination Therapy Using Brominated Hemicyanine Loaded Folate Receptor Targeting Ag<sub>2</sub>S Quantum Dots

*Eda Celikbas<sup>1</sup>, Ayca Saymaz<sup>1</sup>, Hande Gunduz<sup>1,2</sup>, Irem Koc<sup>3</sup>, Ece Cakir<sup>3</sup>, Alphan Sennaroglu<sup>4,5</sup>, Safacan Kolemen<sup>1,4\*</sup>, Havva Yagci Acar<sup>1,4\*</sup> and Kubra Onbasli<sup>6\*</sup>*

<sup>1</sup>Department of Chemistry, Koç University, Rumelifeneri Yolu, Sariyer, Istanbul 34450, Türkiye

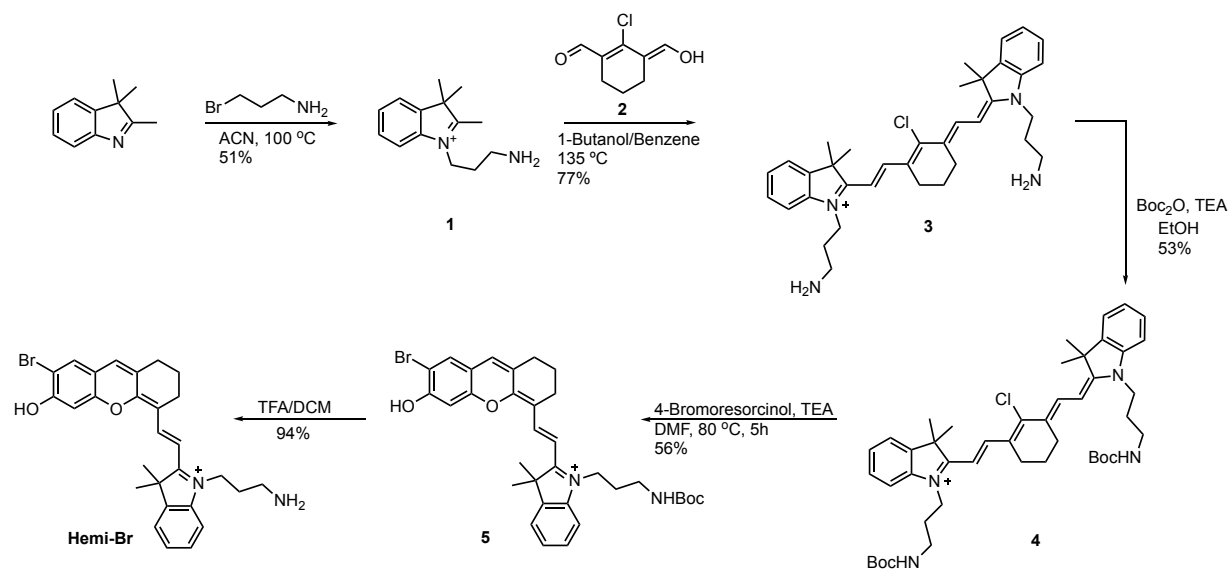
<sup>2</sup>Nanofabrication and Nanocharacterization Centre for Scientific and Technological Advanced Research, Koç University, Istanbul, Türkiye

<sup>3</sup>Graduate School of Materials Science and Engineering, Koç University, Rumelifeneri Yolu, Istanbul 34450, Türkiye

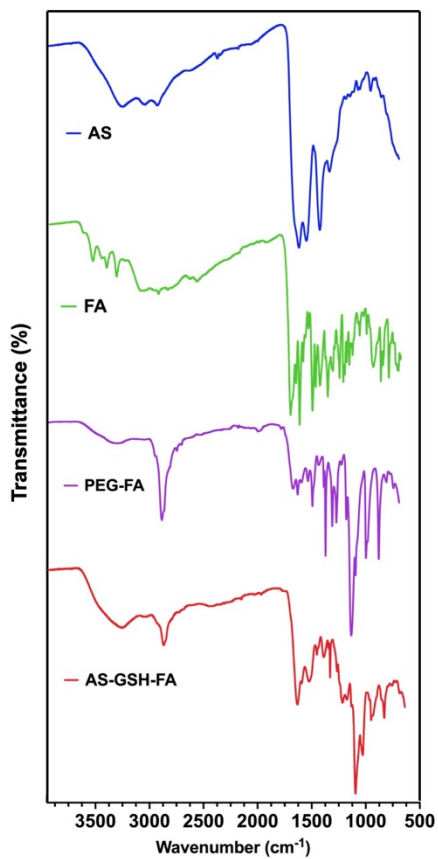
<sup>4</sup>KUYTAM, Koç University Surface Science and Technology Center, Koc University, Rumelifeneri Yolu, Istanbul 34450, Türkiye

<sup>5</sup>Departments of Physics and Electrical-Electronic Engineering, Koç University, Rumelifeneri Yolu, 34450 Istanbul, Türkiye

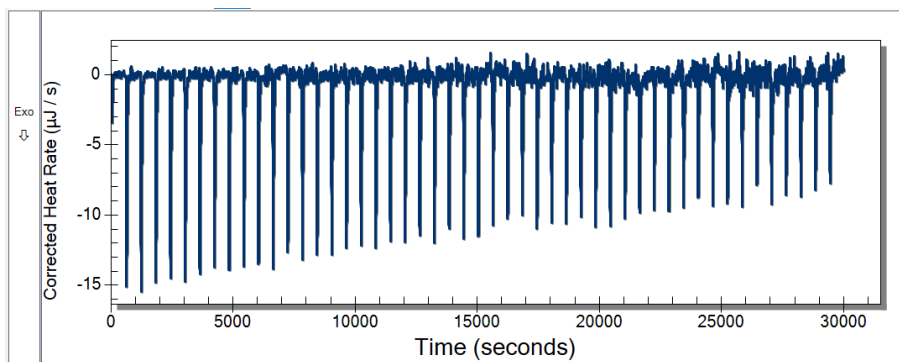
<sup>6</sup>Department of Metallurgical and Materials Engineering, 34469 Istanbul Technical University, Istanbul, Türkiye



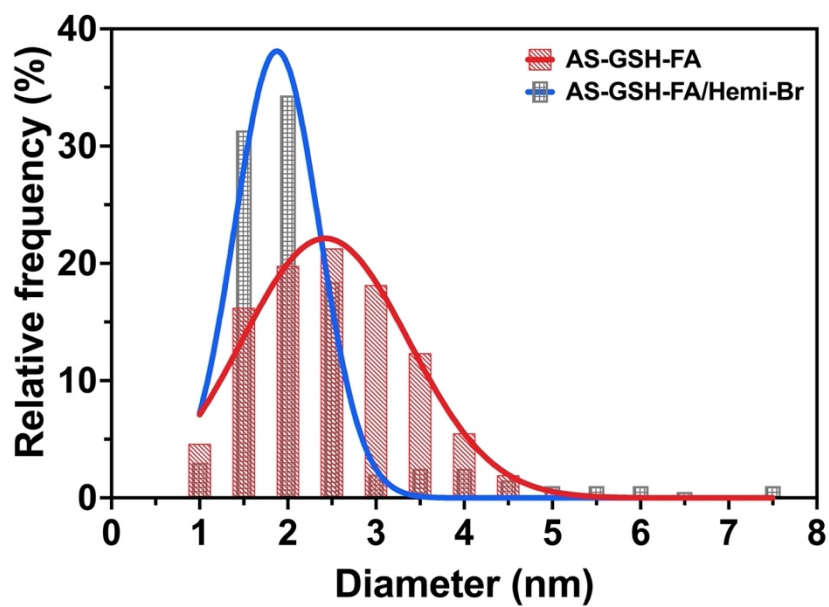
**Scheme S 1** Synthetic pathway of Hemi-Br.



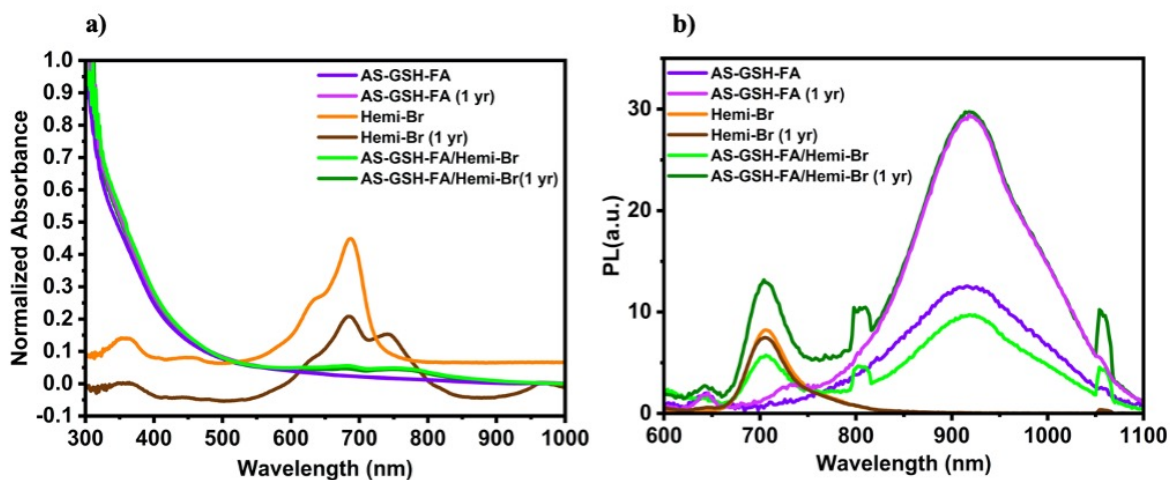
**Figure S 1** FTIR spectrum of AS-GSH QDs, FA, PEG-FA and AS-GSH-FA QDs.



**Figure S 2.** ITC exotherm profile of the titration of AS-GSH-FA with Hemi-Br.



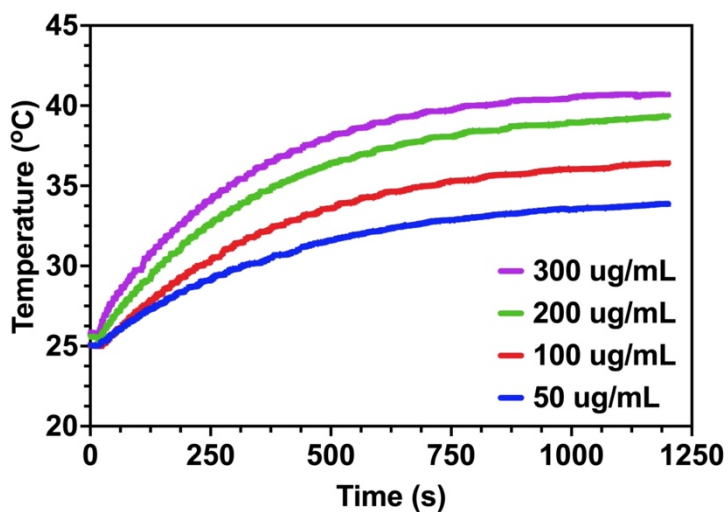
**Figure S 3** TEM histograms of the QDs corresponding to Figure 2c-d.



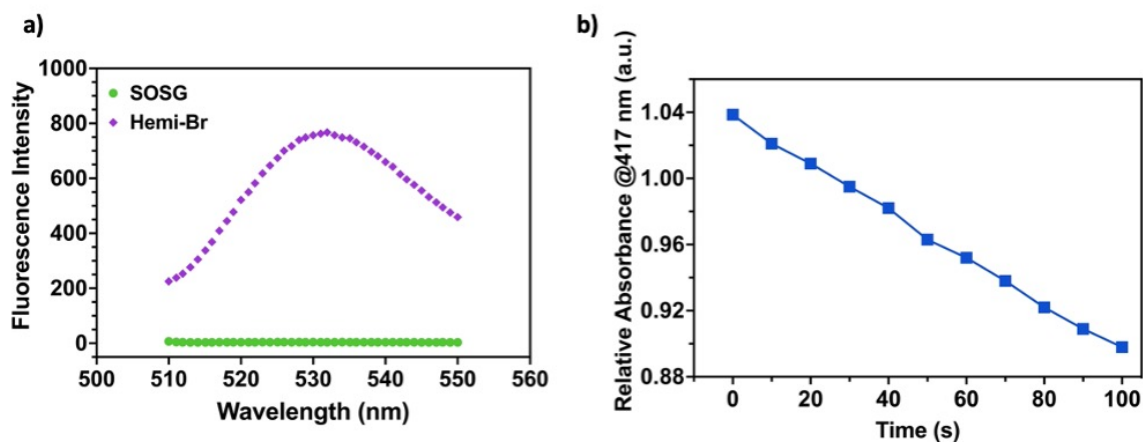
**Figure S 4** Absorbance (a) and photoluminescence (b) spectra of the QD conjugates on day one and year one of the synthesis.

**Table S 1** Hydrodynamic size and zeta potential of prepared QDs on day one and year one of the synthesis.

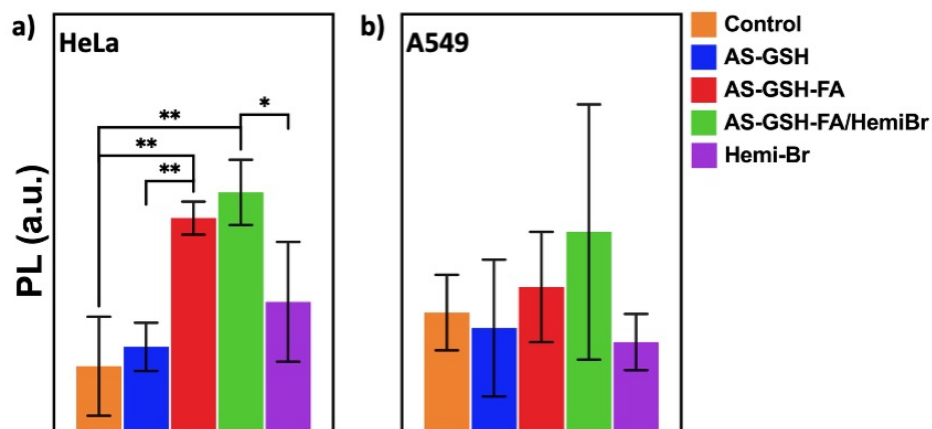
Sample Name	PDI	Size by number (nm)	Size by intensity (nm)	Zeta Potential (mV)
AS-GSH	0.17	2.94	7.21	-42.20
AS-GSH-FA	0.60	5.22	7.75	-34.70
AS-GSH-FA/Hemi-Br	0.47	75.50	112.20	-21.60
AS-GSH-FA (1 year)	0.16	5.07	7.61	-14.90
AS-GSH-FA/Hemi-Br (1 year)	0.55	87.34	107.4	-16.70



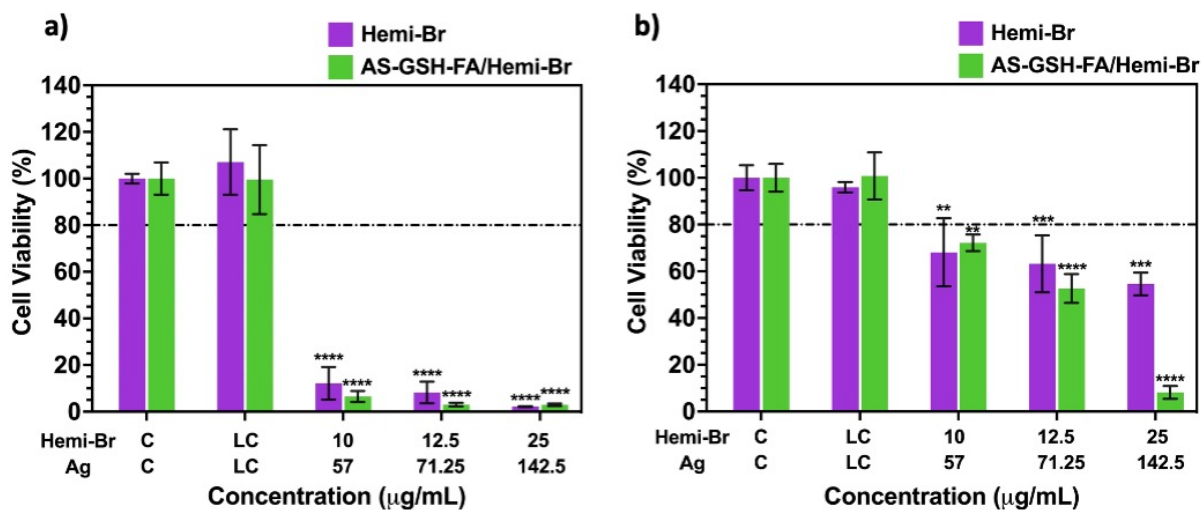
**Figure S 5.** Time and [Ag] concentration-dependent change in the temperature of AS-GSH-FA/Hemi-Br solutions irradiated at 640 nm, 215 mW.



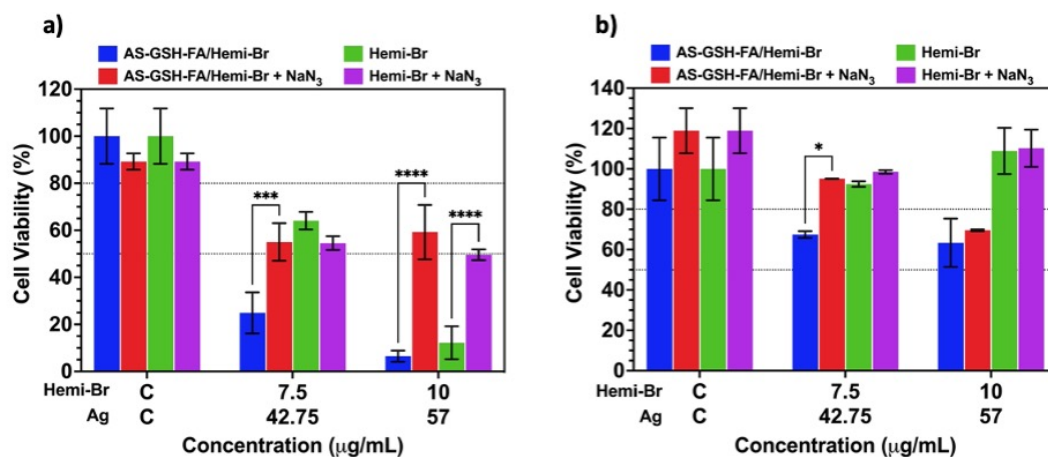
**Figure S 6.** (a) Fluorescence signal of SOSG at 525 nm upon irradiation at 640 nm (215 mW) of free Hemi-Br in 10% MeOH/PBS buffer (pH 7.4) at 7.5  $\mu$ g Hemi-Br/mL as a spectrum. (b) Decrease in the absorption signal of DPBF with respect to time upon irradiation of Hemi-Br with a 640 nm light in DMSO:PBS (99:1, pH 7.4).



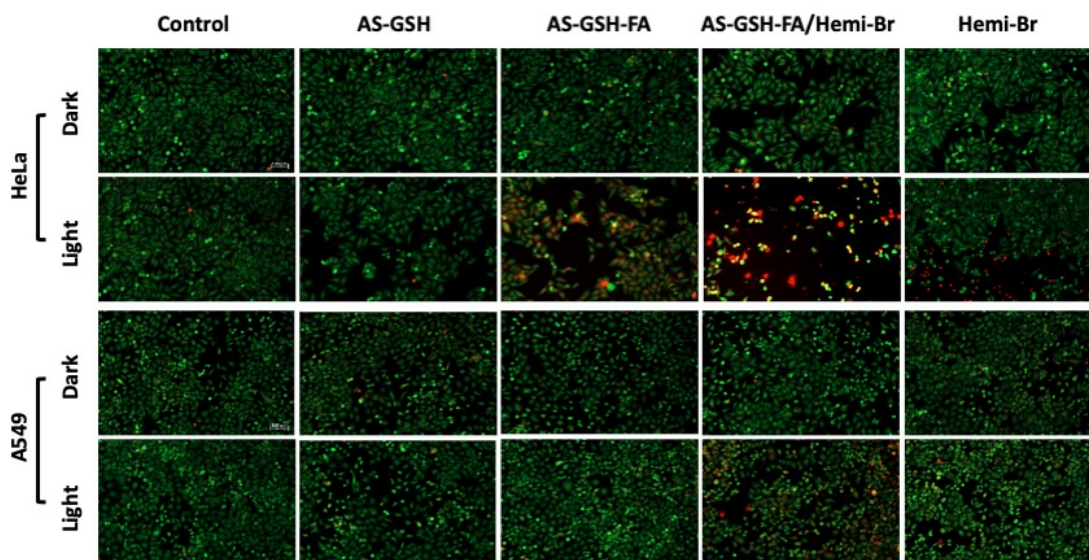
**Figure S 7.** Quantified intracellular NIR fluorescence signal measurement from (a) HeLa and (b) A549 cells treated with AS-GSH, AS-GSH-FA, AS-GSH-FA/Hemi-Br and free Hemi-Br for 6 h at 57  $\mu\text{g}$  Ag/mL and 10  $\mu\text{g}$  Hemi-Br/mL dose. The data are expressed as mean  $\pm$  S.D. (n=3). (0.0332 (\*), 0.0021 (\*\*), 0.002 (\*\*\*), < 0001 (\*\*\*\*))



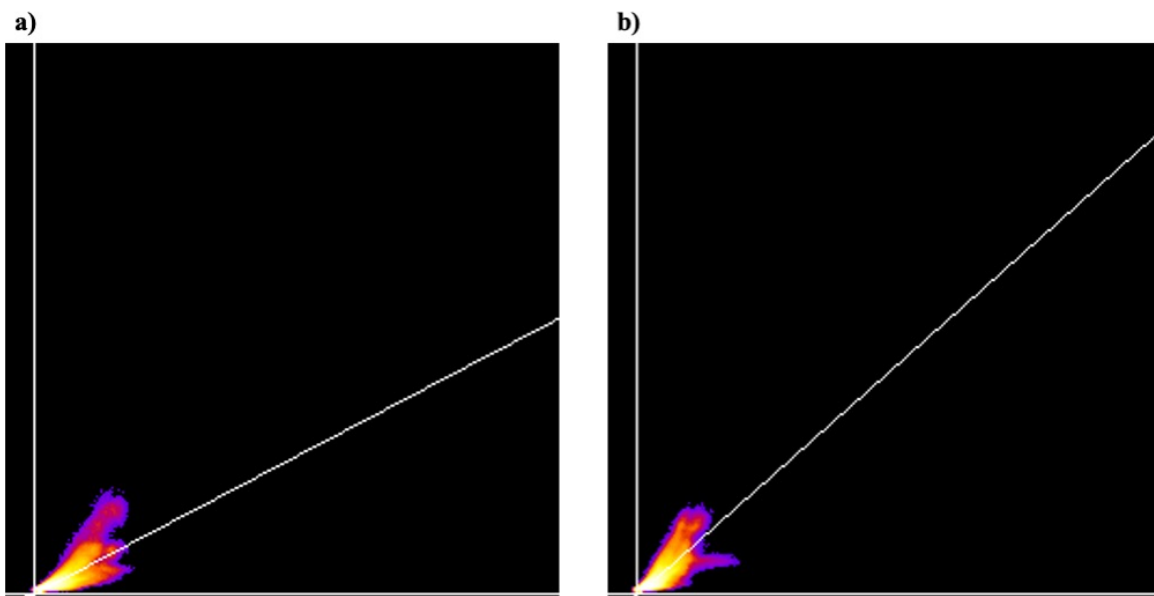
**Figure S 8.** Dose-dependent viability of (a) HeLa and (b) A549 cells treated with free Hemi-Br and QD-conjugates for 6 h and irradiated at 640 nm for 5 min at high doses. The data are expressed as mean  $\pm$  S.D. (n=3). (0.0332 (\*), 0.0021 (\*\*), 0.002 (\*\*\*), < 0001 (\*\*\*\*))



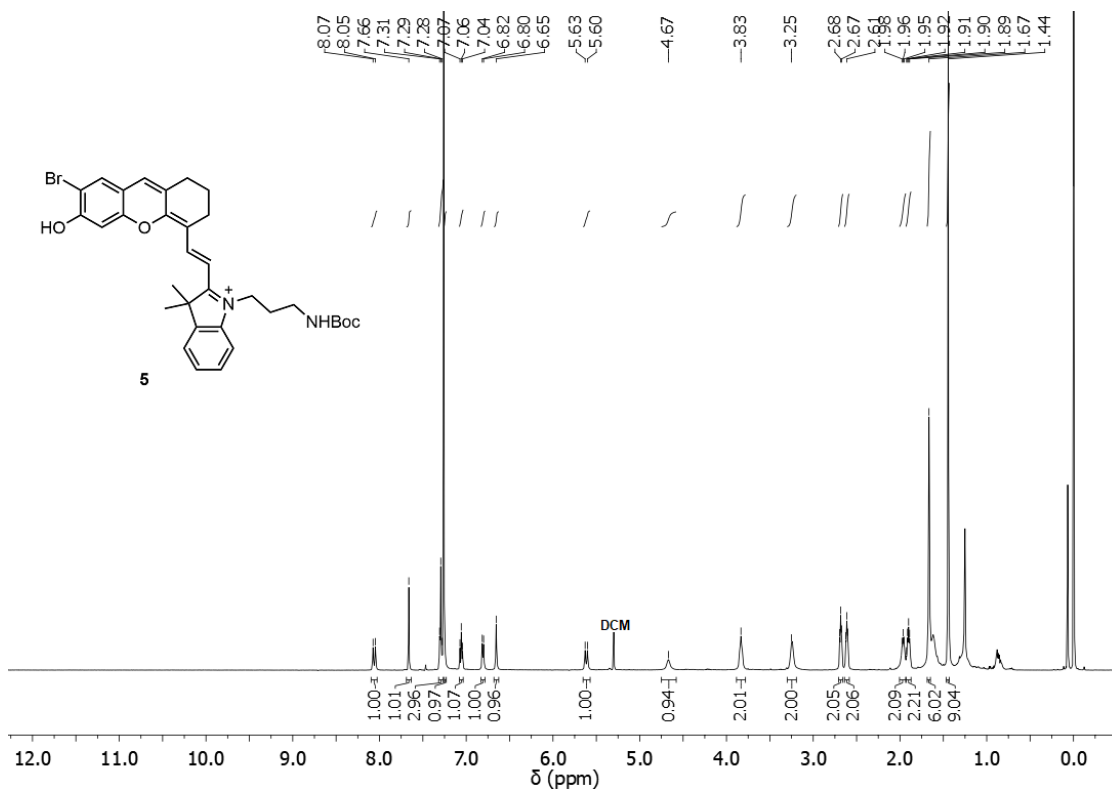
**Figure S 9.** Cell viabilities of (a) HeLa and (b) A549 cells treated with Hemi-Br and AS-GSH-FA/Hemi-Br upon laser irradiation (640 nm, 300 mW) for 5 min in the absence or presence of NaN<sub>3</sub> (5 mM). (0.0332 (\*), 0.0021 (\*\*), 0.002 (\*\*\*), < 0001 (\*\*\*\*))



**Figure S 10.** Fluorescence microscopy images of HeLa and A549 cells treated with QDs at 42.75 µg Ag/mL and 7.5 µg Hemi-Br/mL concentrations for 6 h (Dark) followed by laser irradiation (640 nm, 300 mW, 5 min) (Light) after live/dead staining. “Control” indicates untreated cells.



**Figure S 11.** Pearson correlation plots of MTG and (a) free Hemi-Br and (b) AS-GSH-FA/Hemi-Br. Images were processed and analyzed with ImageJ software.



**Figure S 9.**  $^1\text{H}$  NMR spectrum of compound 5 in  $\text{CDCl}_3$ .



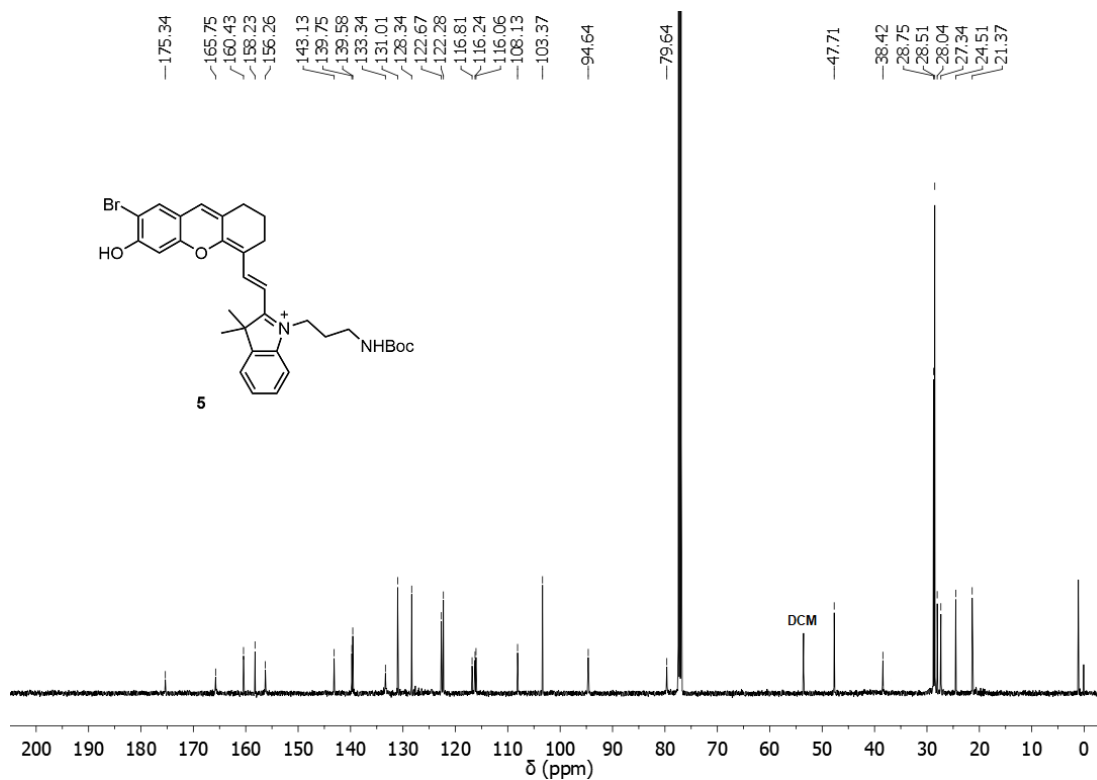


Figure S 10. <sup>13</sup>C NMR spectrum of compound **5** in CDCl<sub>3</sub>.

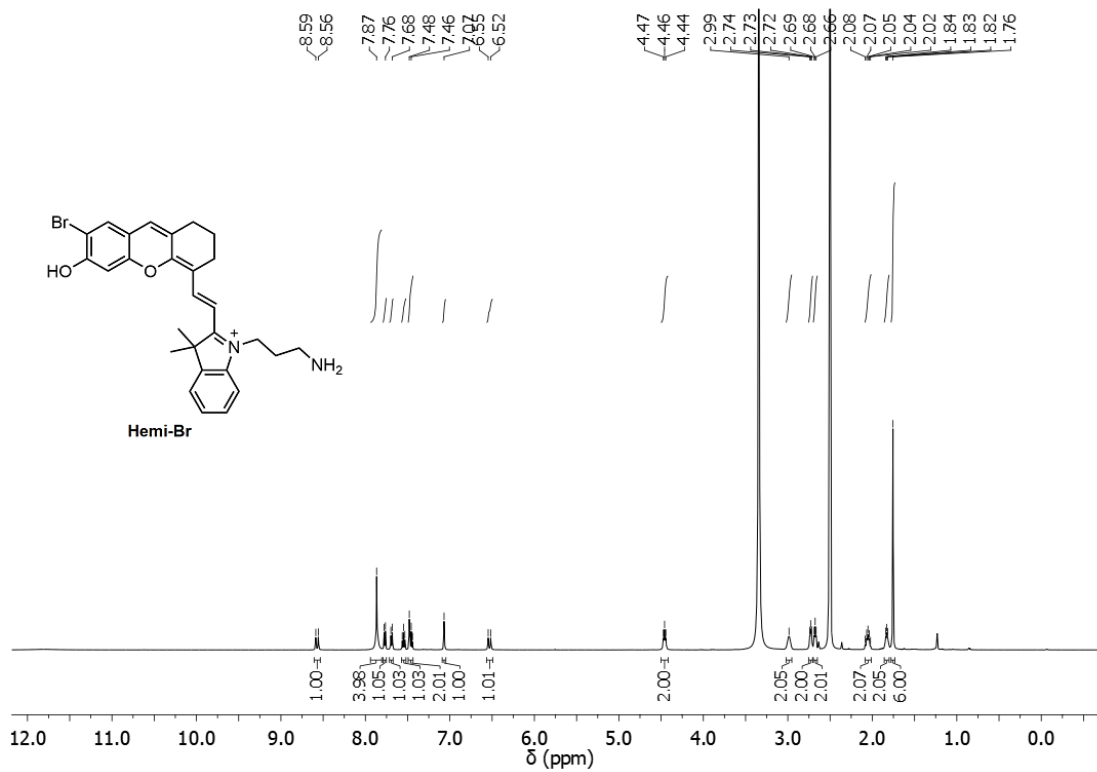
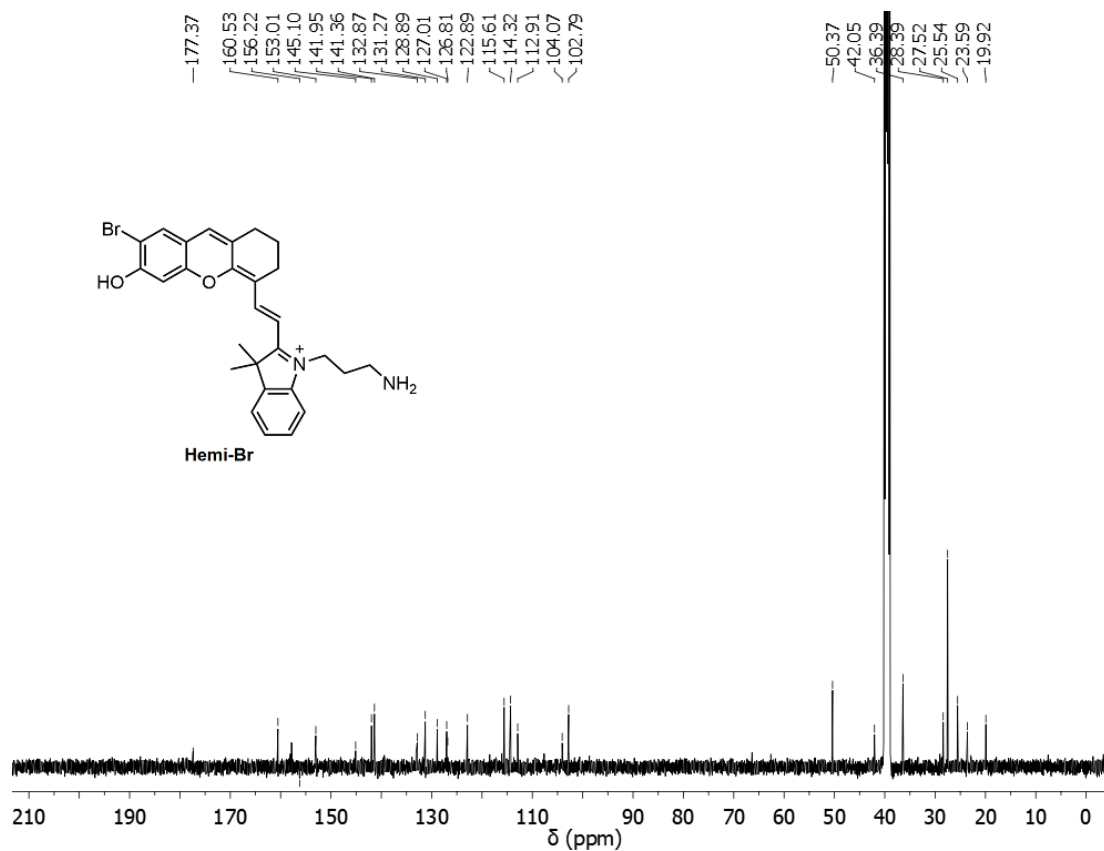
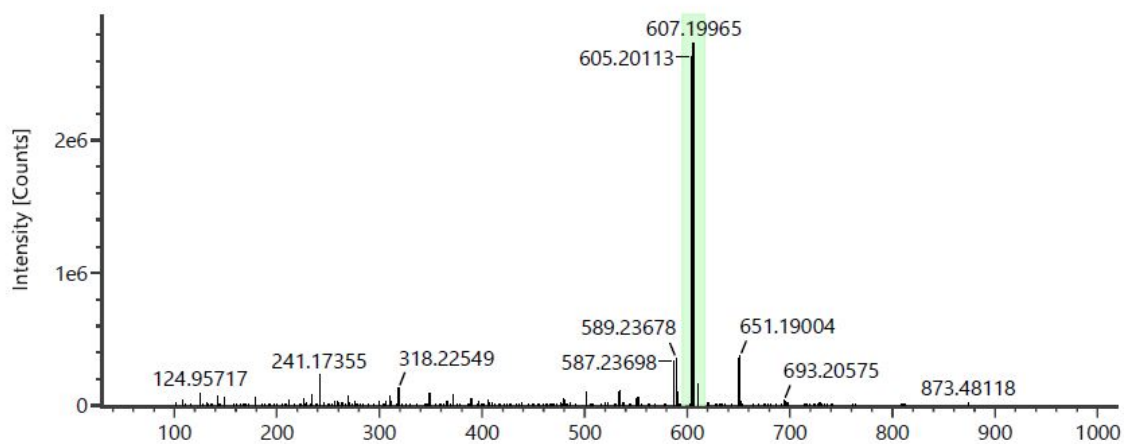


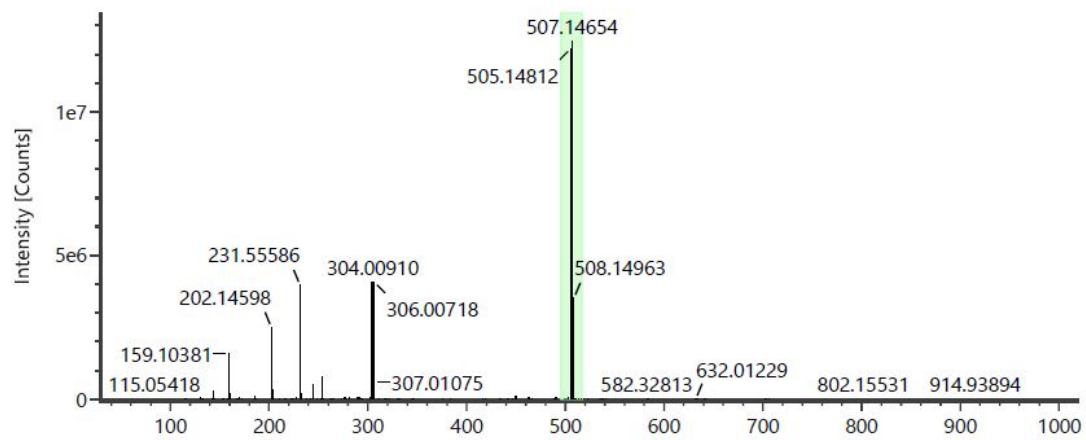
Figure S 11. <sup>1</sup>H NMR spectrum of compound **Hemi-Br** in DMSO-*d*<sub>6</sub>.



**Figure S 12.** <sup>13</sup>C NMR spectrum of compound **Hemi-Br** in DMSO-*d*<sub>6</sub>



**Figure S 13.** HRMS spectrum of compound **5**.



**Figure S 14.** HRMS spectrum of compound **Hemi-Br**.