

Supporting Information

Double-ratiometric fluorescence imaging of H_2Se and $\text{O}_2^{\cdot-}$ under hypoxia for exploring Na_2SeO_3 induced HepG2 cells' apoptosis

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1. Excitation and emission spectra of NIR-H₂Se with H₂Se, DHE with O₂^{•-} and Rhodamine 110.

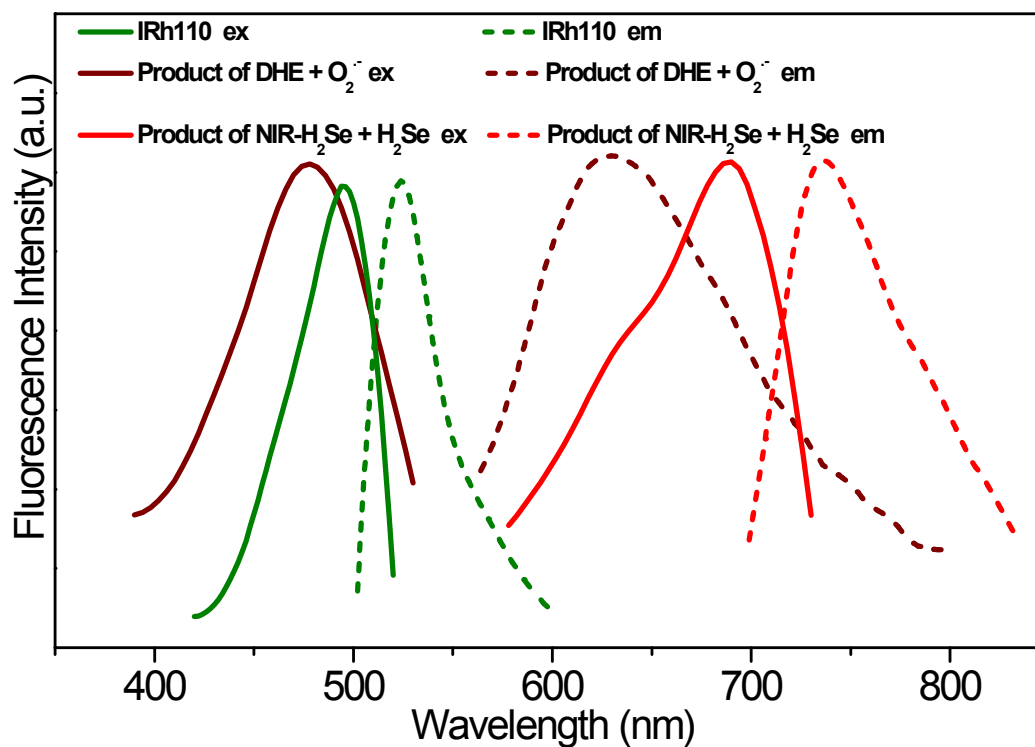


Figure S1. Excitation and emission spectra of the NIR-H₂Se with H₂Se, DHE with O₂^{•-} and Rhodamine 110, and their excitation and emission wavelengths are as follows respectively: $\lambda_{\text{ex}}/\lambda_{\text{em}} = 688/735$ nm, $\lambda_{\text{ex}}/\lambda_{\text{em}} = 488/638$ nm, $\lambda_{\text{ex}}/\lambda_{\text{em}} = 496/532$ nm.

2. Confocal fluorescence images of endogenous H_2Se and $\text{O}_2^{\cdot-}$ in living HepG2 cells treated with various concentrations of Na_2SeO_3 under normoxic (20% O_2) conditions.

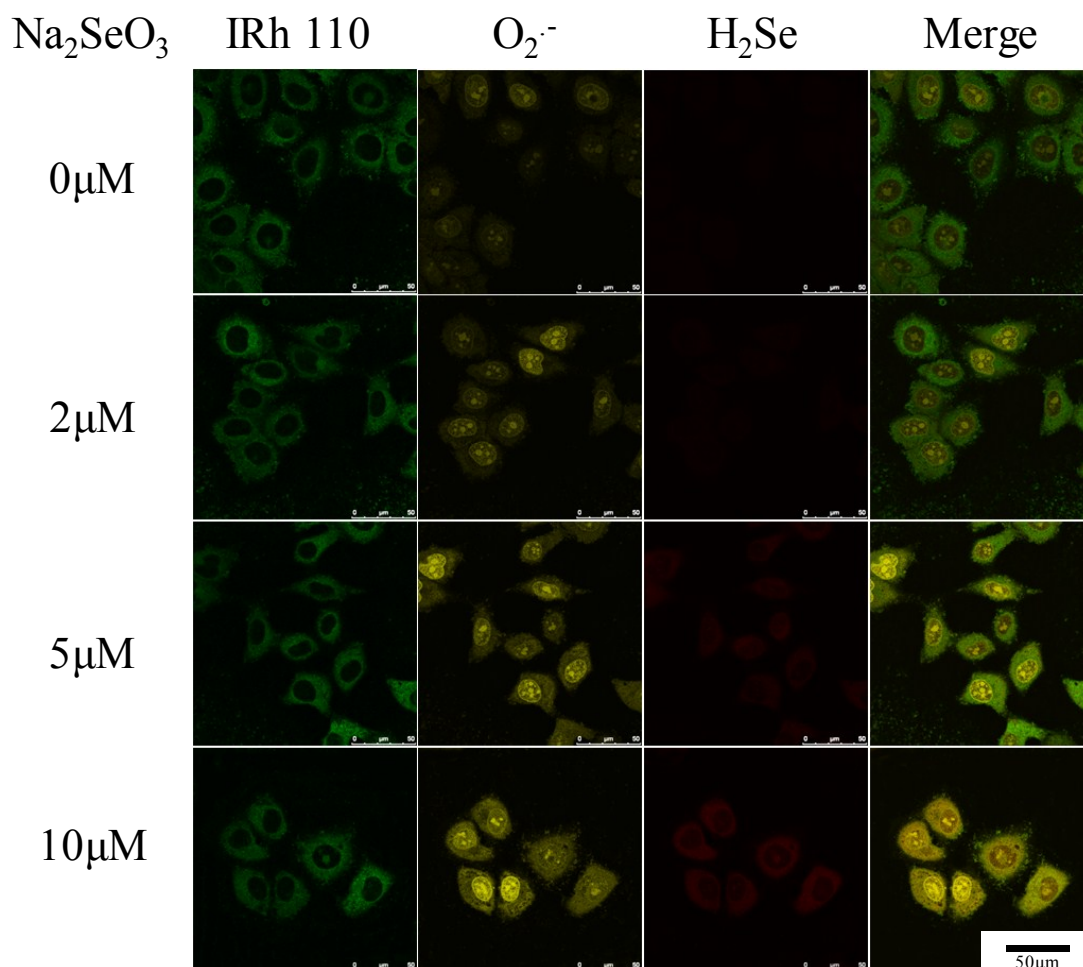


Figure S2. Confocal fluorescence images of endogenous H_2Se and $\text{O}_2^{\cdot-}$ in living HepG2 cells treated with various concentrations of Na_2SeO_3 under normoxic (20% O_2) conditions. The living HepG2 cells were treated with various concentrations of Na_2SeO_3 (0, 2, 5, 10 μM) for 12 h and then incubated with the mixture of 10 μM NIR- H_2Se , 5 μM DHE and 1 μM Rhodamine 110.

3. Confocal fluorescence images of endogenous H_2Se and $\text{O}_2^{\cdot-}$ in living HepG2 cells treated with various concentrations of Na_2SeO_3 under hypoxic (5% O_2 and 1% O_2) conditions.

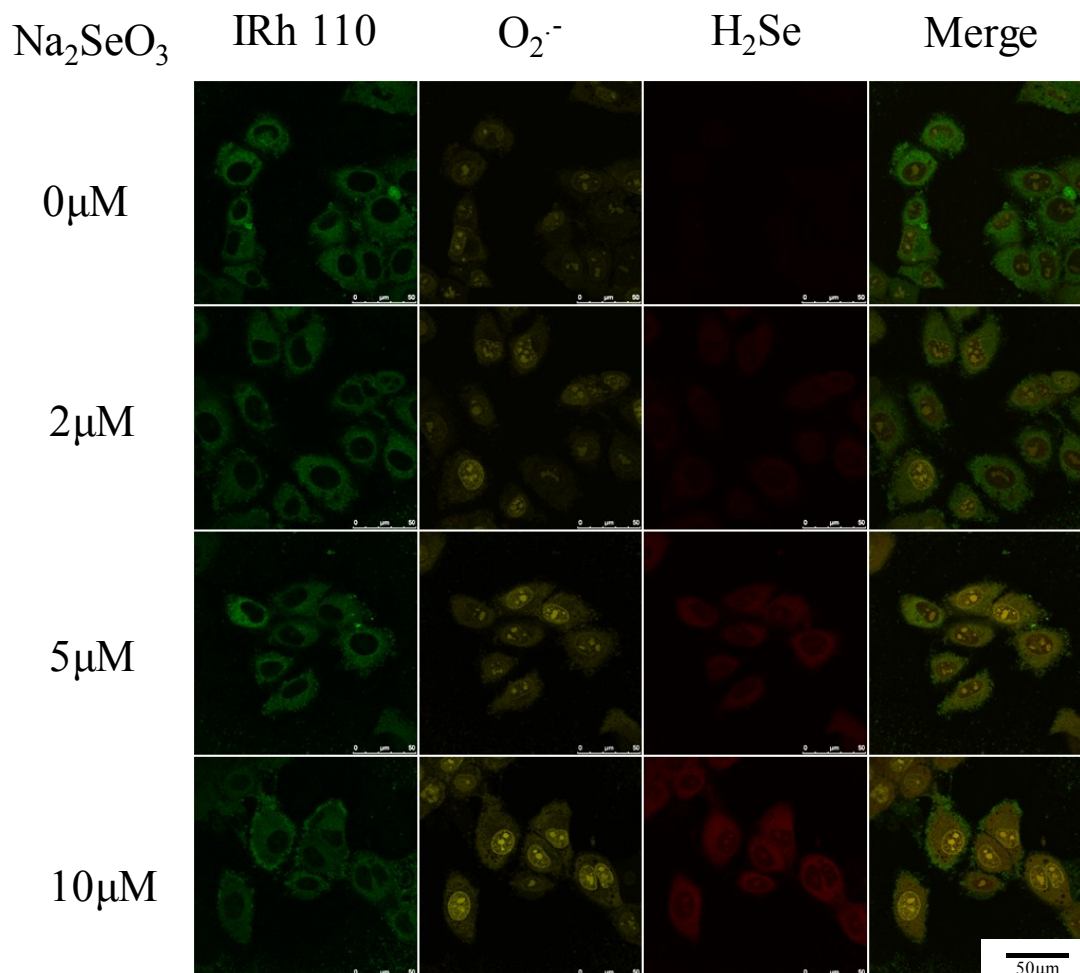


Figure S3. Confocal fluorescence images of endogenous H_2Se and $\text{O}_2^{\cdot-}$ in living HepG2 cells treated with various concentrations of Na_2SeO_3 under hypoxic (5% O_2) conditions. The living HepG2 cells were treated with various concentrations of Na_2SeO_3 (0, 2, 5, 10 μM) for 12 h and then incubated with the mixture of 10 μM NIR- H_2Se , 5 μM DHE and 1 μM Rhodamine 110.

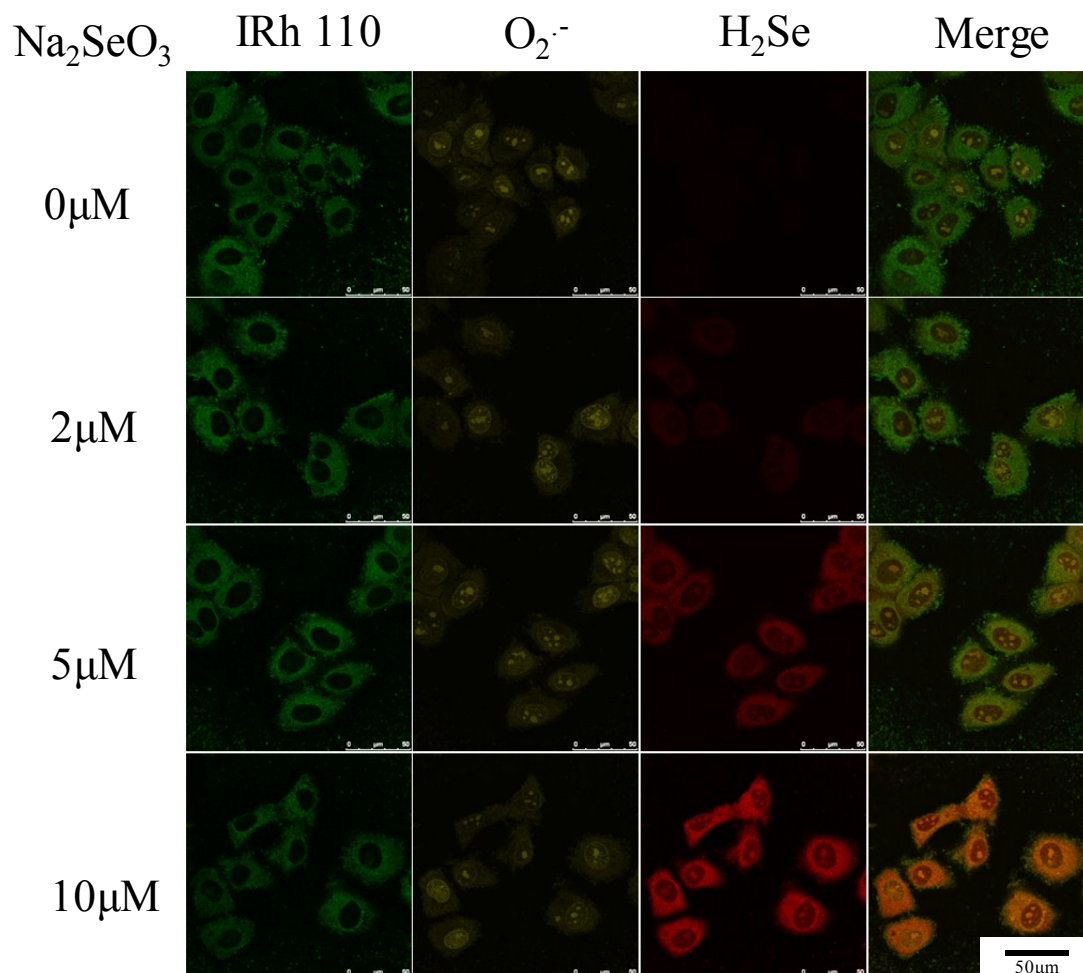


Figure S4. Confocal fluorescence images of endogenous H_2Se and $\text{O}_2^{\cdot-}$ in living HepG2 cells treated with various concentrations of Na_2SeO_3 under hypoxic (1% O_2) conditions. The living HepG2 cells were treated with various concentrations of Na_2SeO_3 (0, 2, 5, 10 μM) for 12 h and then incubated with the mixture of 10 μM NIR- H_2Se , 5 μM DHE and 1 μM Rhodamine 110.

4. **Scheme S1:** The chemical formula of NIR-H₂Se, DHE and their respective reaction product.

