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Supporting Information

Carbonyl Sulfide (COS) Donor Induced Protein Persulfidation Protects against Oxidative Stress

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Figure S1: Structures of Mesalamine and **7a-7c**.

Figure S2. Representative plots for the time course of H₂S release as monitored by methylene blue complex formation assay (λ_{max} - 676 nm) a) Kinetics of H₂S release from compound **1a**. b) H₂S generation plot for compound **1b**.

Figure S3. Representative plot for the time course of H₂S release from NQO1-Mes as monitored by methylene blue assay (λ_{max} - 676 nm). The rate constant for H₂S release was calculated to be 0.012 min⁻¹.

Dicoumarol; **8**

Figure S4. Structure of dicoumarol, **8**, a known inhibitor of NQO1 enzyme.

Figure S5. a) HPLC trace for compound **1a** in buffer. b) Area under the curve for the formation of *p*-anisidine. c) Area under the curve corresponding to the formation of lactone. d) Area under the curve for decomposition of compound **1a**.

Figure S6. Representative HPLC trace for compound NQO1-Mes in ACN (Retention time (RT) = 13.4 min). b) Representative HPLC traces for the decomposition of compound NQO1-Mes in the presence of NQO1 and NADH with concomitant release of lactone (RT = 5.8 min) and compound **9** (RT = 3.9 min). c) Area under the curve corresponding to the formation of mesalamine methyl ester, **9**.

Figure S7. Detection of H₂S using sulfide selective electrode. Signal for H₂S was observed from compound NQO1-Mes in buffer containing NQO1 and NADH. No current corresponding to H₂S release was observed from NQO1-Mes alone in buffer or RPMI media containing 10% FBS.

Figure S8. a) Structures of CN-BOT and methyl sulfonylbenzathiazole (MSBT) used for the detecting cellular persulfidation levels. b) Protein persulfidation induced by NQO1 activated COS/H₂S donors.

Figure S9. Cell viability assay conducted with **1a** for 72 h in human breast cancer, MCF-7 cells.

Figure S10. Cell viability assay conducted with compound **1a** for 24 h in human breast cancer, MCF-7 cells was measured using LDH assay.

Figure S11. a) Structure of JCHD. b) Induction of cell death using JCHD in DLD-1 cells in 24 h measured using MTT assay.

Figure S12. a) Cell viability of compound **1a** in human colon carcinoma, DLD-1 cells. b) Cytoprotective effects of varying concentrations of **1a** against JCHD induced stress in DLD-1 cells in 24 h. c) Cell viability of compound **1b** in DLD-1 cells after 24 h. d) Cytoprotective effects of varying concentrations of **1b** against JCHD induced stress in DLD-1 cells in 24 h.

Figure S13. a) Cell viability for compounds **1a** and **1b** in wild type mouse embryonic fibroblast, WT-MEF cells. b) Protection against JCHD induced stress in WT-MEF cells in 24 h using varying concentrations of compounds **1a** and **1b**.

Figure S14. a) Cell viability for NQO1-Mes in wild type mouse embryonic fibroblast, WT-MEF cells. b) Cytoprotective effects of NQO1-Mes against JCHD (15 μM) induced oxidative stress in WT-MEF cells over a period of 24 h. Results are expressed as Mean $\pm$ SEM (n = 3). [ns – non significant]. Compound **5** (50 μM) represents the negative control and Mes represents mesalamine (50 μM).