A ratiometric fluorescent probe for real-time monitoring of intracellular glutathione fluctuations in response to cisplatin

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Fig. S1. Absorption spectra of **1** (10 μ M) upon gradual addition of GSH (0-10 mM) in 0.2M PBS (30% ethanol). The excitation wavelength is 514nm. Each spectrum was recorded 15 minutes after GSH addition.



Fig. S2.Fluorescence spectra of 1 (10 μ M) upon gradual addition of GSH (0-10 mM) in 0.2M PBS (30% ethanol). The excitation wavelength is 514nm. Each spectrum was recorded 15 minutes after GSH addition.



Fig. S3. Time-course fluorescence response spectra of **1** (10 μ M) towards GSH concentration (2.5 mM -5 mM) both in forward and reverse reactions in 0.2 M PBS (30% ethanol) at 37 °C. λ_{ex} = 488 nm, λ_{em} = 544 nm. GSH was depleted by adding N-Ethylmaleimide (NEM).



Fig. S4. Time-course fluorescence response spectra of 1 (10 μ M) towards GSH (10 mM) in 0.2 M PBS (30% ethanol). λ_{ex} = 488 nm, λ_{em} = 544 nm.



Fig. S5. Fluorescence spectra of **1** (10 μ M) upon gradual addition of cysteine (0-10 mM) in 0.2 M PBS (30% ethanol). The excitation wavelength is 514 nm. Each spectrum was recorded 15 minutes after cysteine addition.



Fig. S6. Fluorescence spectra of **1** (10 μ M) upon gradual addition of homocysteine 0.1-1 mM in 0.2 M PBSd (30% ethanol). The excitation wavelength is 514 nm. Each spectrum was recorded 15 minutes after homocysteine addition.



Fig. S7. Fluorescence spectra of **1** (10 μ M) upon gradual addition of taurine 20 mM in 0.2 M PBS (30% ethanol). The excitation wavelength is 514 nm. Each spectrum was recorded 15 minutes after homocysteine addition.



Fig. S8. Fluorescence ratio of **1** after equilibrated with various of physiologically relevant species.



Fig. S9. Fluorescence spectra of 1 (10 μ M) in different pH in 30% ethanol solution. The excitation wavelength is 514 nm. Each spectrum was recorded 15 minutes after pH adjustment.



Fig. S10. Cell viability after 24 hours incubated with different concentration of 1.



Fig. S11. ¹H NMR of 1



Fig. S12. ¹H NMR of 1+2-mercaptoethanol.



Fig. S13. ¹³C NMR of 1



Fig. S14. ¹³C NMR of 1+2-mercaptoethanol.



Fig. S15. H-H COSY of NMR of 1



Fig. S16. H-H COSY of NMR of 1+2-mercaptoethanol.



Fig. S17. H-H COSY of NMR of 1+2-mercaptoethanol (zoom)



Fig. S18. HSQC of NMR of 1



Fig. S19. HSQC of NMR of 1+2-mercaptoethanol.



Fig. S20. Cell viability in A549 and A549-DDP cells upon treatment with cisplatin (20 μ M) at different time in the same culture conditions as the confocal experiments. Data are expressed as mean (%) ± standard deviation (S.D.) of at least three independent assays.



Fig. S21. Cell viability in A549 cells upon oxoplatin and oxclplatin (10 μM) treatment at different time. Data are expressed as mean(%)±standard deviation(S.D.) of at least three independent assays. *,p<0.05,**;p<0.01



Fig. S22. HRMS of [1+Na]+



Fig. S23. HRMS of [1+H]⁺



Fig. S24. Linear relationship between $(R - R_{min})/(R_{max} - R)$ and GSH concentration. R is defined as the ratio of the fluorescence intensities between 1-GSH (544 nm) and the apparent fluorescence isosbestic point (603 nm), R_{min} and R_{max} correspond to the R values at zero and saturated GSH concentrations, respectively.