Supporting Information

Thiazole Orange Styryl Derivatives as Fluorescent Probes for G-Quadruplex DNA

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EXPERIMENTAL SECTION

Materials. All oligonucleotides used in this study were synthesized by Sangon Biotech Co. Ltd. (Shanghai, China). 4-chloro-2-methylquinolin, 4-(diethylamino)benzaldehyde, 4-(4-methylpiperazino) -benzaldehyde and 4-(4-pyridinyl)benzaldehyde were purchased from Energy Chemical. 4-methl- piperidine, 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonicacid) diammonium salt (ABTS), 2-methylbenzothiazole, and iodomethane were purchased from J&K Company. Hemin was purchased from Beijing XinJingKe Biotechnology Ltd. Stock solutions of **5a-5d** (10.0 mM) were prepared in DMSO.

Instruments. Electropray ionization mass spectrometry (ESI-MS) was recorded on an LC-MS 2010A system (Shimadzu). High-resolution MS was conducted on a Bruker Daltonics Flex-Analysis spectrometer. ^{1}H nuclear magnetic resonance (NMR) spectra were measured on Avance III 500 WB nuclear magnetic resonance spectrometer (Bruker). Absorption spectra were collected on UH5300 spectrophotometer (Hitachi). Fluorescence spectra were recorded on a Hitachi FL-4600 fluorescence spectrofluorometer. Oligonucleotide concentrations were determined on a ThermoFisher NanoDrop 2000 instrument. Circular dichroism spectra were recorded on a JASCO J-815 circular dichroism spectropolarimeter. Fluorescence images were recorded on an FV3000-IX83 confocal microscope (Olympus).

Synthesis of 1-4 (Scheme 1). 4-chloro-1,2-dimethylquinolin-1-ium Iodide (1). A mixture of 4-chloro-2-methylquinolin (0.40 g, 2.24 mmol), iodomethane (0.84 mL, 13.48 mmol) and sulfolane (4.0 mL) was heated to 75 ℃ and stirred for 9 h. After reaction mixture cooling to room temperature, anhydrous ether was added, the solids were collected by filtration, washed with anhydrous ether several times, and dried under reduced pressure. MS(ESI): m/z calcd for $C_{11}H_{11}ClN^{+}$ $[M-I]^{+}$ 192.05, found 192.01.

1,2-dimethylbenzothiazol-1-ium Iodide (2). 2-methybenzothiazole (0.30 g, 2.0 mmol) mixed with iodomethane (0.40 mL, 4.0 mmol) in 10.0 mL dichloromethane and refluxed overnight. The crude was filtered and washed with anhydrous ether, then obtained compound 2 by dried under vacuum and used for the next reaction without further purification. MS(ESI): m/z calcd for $C_{9}H_{10}NS^{+}$ $[M-I]^{+}$ 164.05, found 164.10.

(Z)-1,2-dimethyl-4-((3-methylbenzo[d]thiazol-2(3H)-ylidene)methyl)quinolin-1-ium-Iodide (3).

Compound 1 (1.0 g, 3.20 mmol), Compound 2 (1.0 g, 3.44 mmol) and aqueous sodium bicarbonate solution (0.5 mol/L, 4.0 mL) were dissolved in 10 mL methanol and stirred at room temperature for 1 h. After then, 6.0 mL saturated KI solution was added to the reaction mixture and continue stirred for 15 min. The precipitate was collected via vacuum filtration, washed with water-acetone and vacuum-dried to give compound 3. MS(ESI): m/z calcd for $C_{20}H_{19}N_2S^+$ $[M-I]^+$ 319.12, found 319.10.

4-(4-formylphenyl)-1-methylpyridin-1-ium (4)**.** 4-(4-pyridinyl)benzaldehyde was dissolved in 2.0 mL sulfolane, 0.5 mL iodomethane was added. The reaction was heated to 75 ℃ and stirred for 9 h. After cooled to room temperature, the precipitant was filtered and washed with anhydrous ethyl ether. MS(ESI): m/z calcd for $C_{13}H_{12}NO^+$ $[M-I]^+$ 198.09, found 198.10.

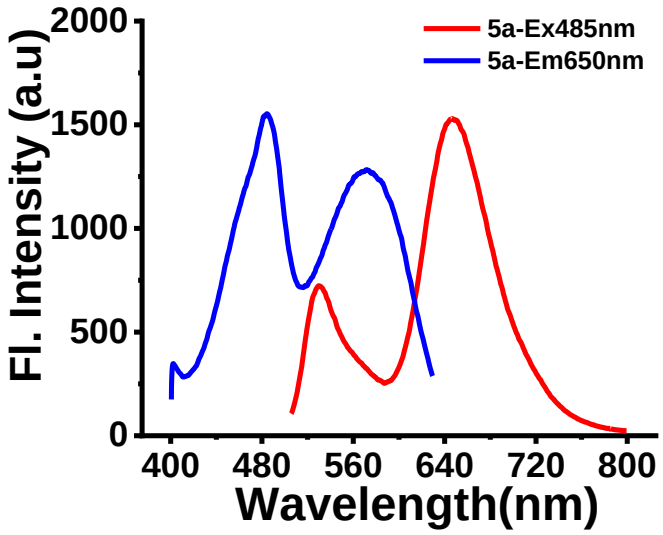

Figure S1. Excitation and emission spectra of **5a** (5.0 μM) in the presence of pu22 (10.0 μM).

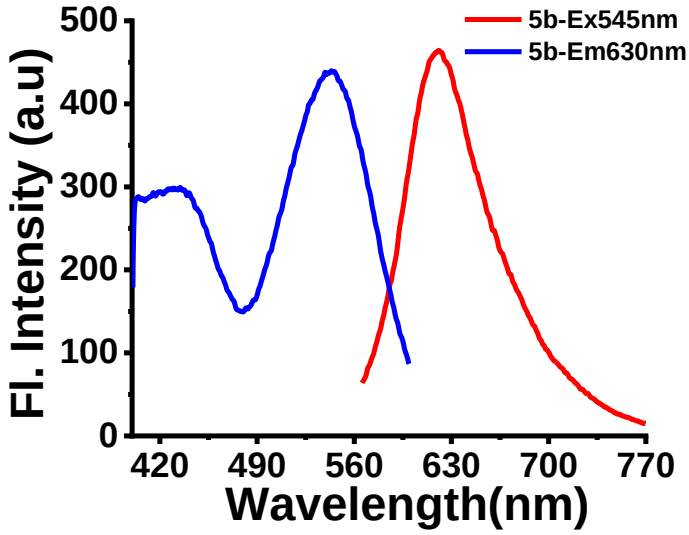

Figure S2. Excitation and emission spectra of **5b** (5.0 μM) in the presence of pu22 (10.0 μM).

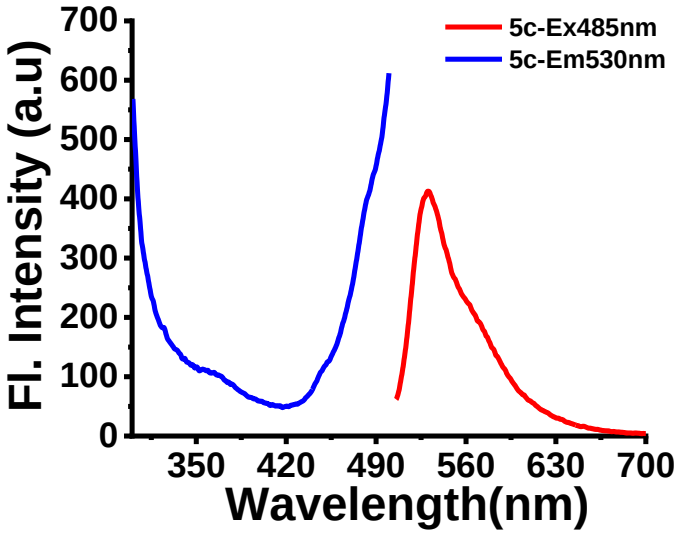

Figure S3. Excitation and emission spectra of **5c** (5.0 μM) in the presence of pu22 (10.0 μM).

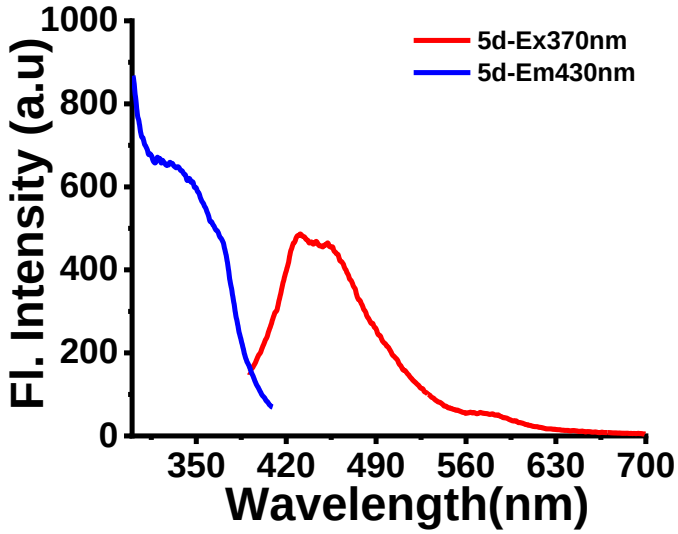

Figure S4. Excitation and emission spectra of **5d** (5.0 μM) in the presence of pu22 (10.0 μM).

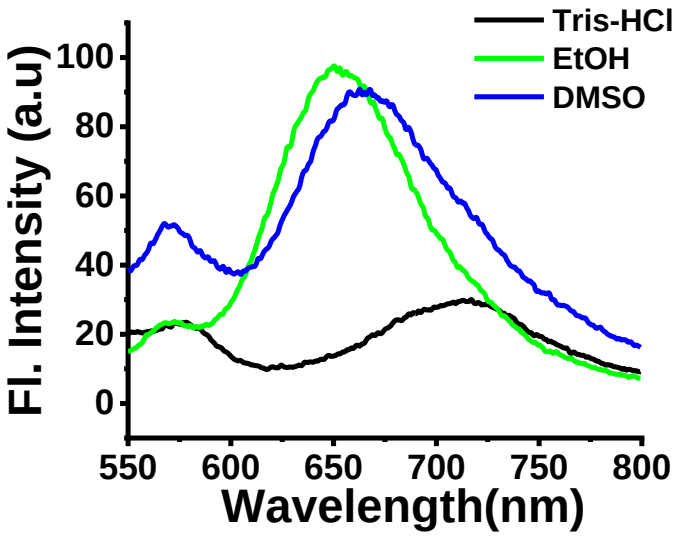

Figure S5. Fluorescence spectra of **5a** (5.0 μM) in different solvents (λ_{ex} = 485 nm).

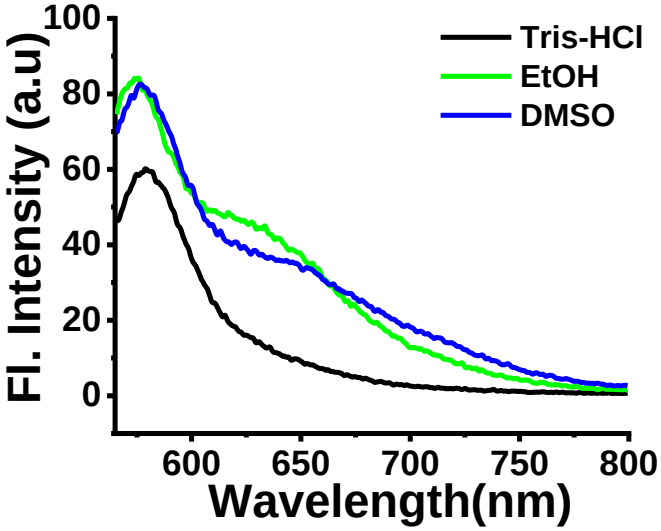

Figure S6. Fluorescence spectra of **5b** (5.0 μM) in different solvents (λ_{ex} = 545 nm).

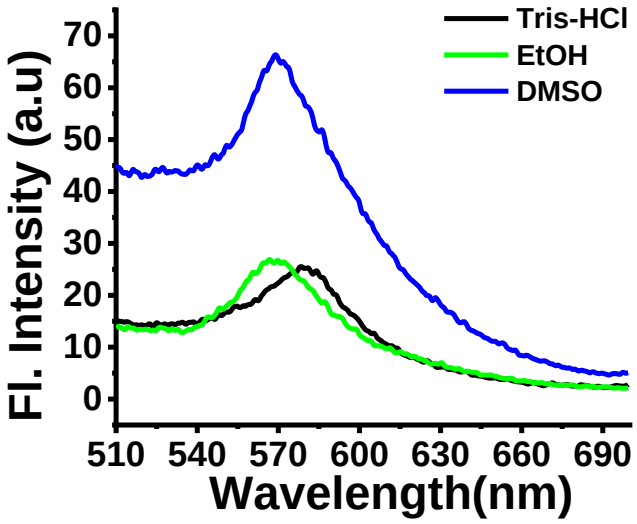

Figure S7. Fluorescence spectra of **5c** (5.0 μM) in different solvents (λ_{ex} = 485 nm).

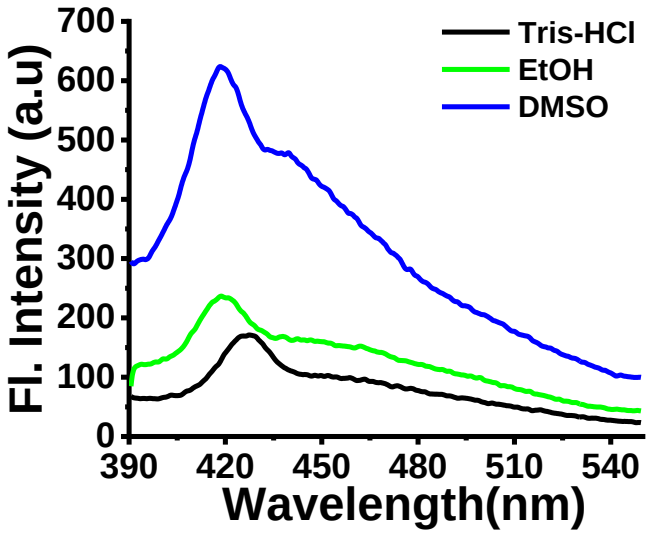

Figure S8. Fluorescence spectra of **5d** (5.0 μM) in different solvents (λ_{ex} = 370 nm).

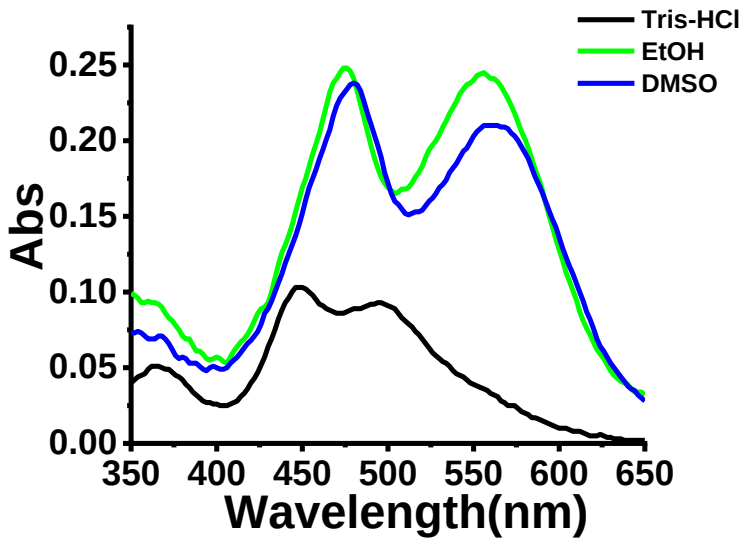

Figure S9. Absorption spectra of **5a** (25.0 μM) in different solvents.

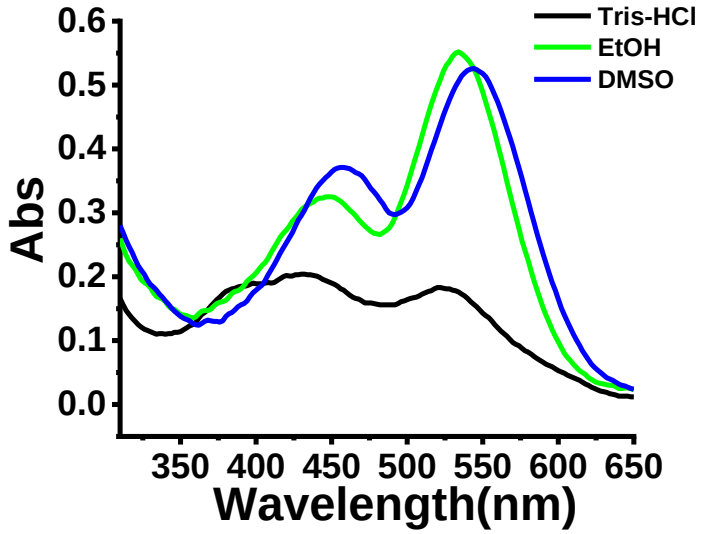

Figure S10. Absorption spectra of **5b** (25.0 μM) in different solvents.

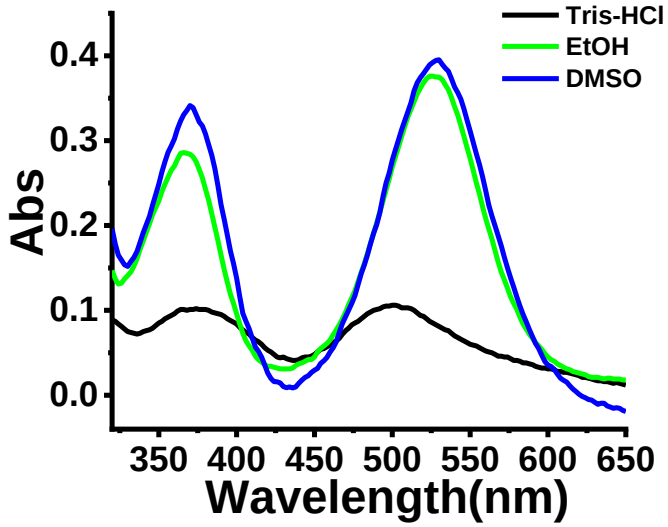

Figure S11. Absorption spectra of **5c** (25.0 μM) in different solvents.

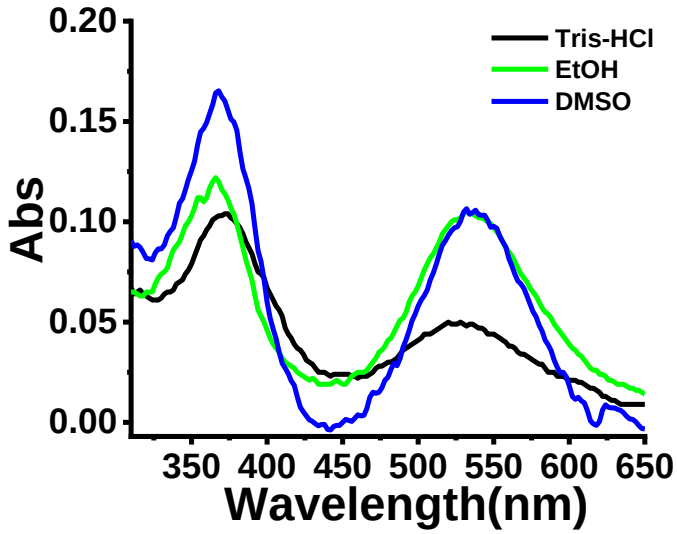

Figure S12. Absorption spectra of **5d** (25.0 μM) in different solvents.

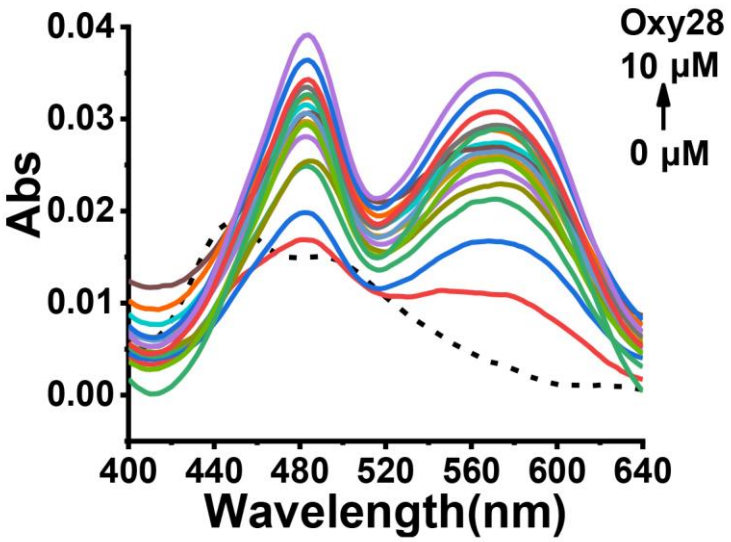

Figure S13. Absorption titration spectra of **5a** with Oxy28, the concentration of **5a** was 5.0 μM and the concentration of Oxy28 was 0, 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.5, 3.0, 4.0, 6.0, 8.0, 10.0 μM; dash lines represent the spectra of **5a** in the absence of Oxy28.

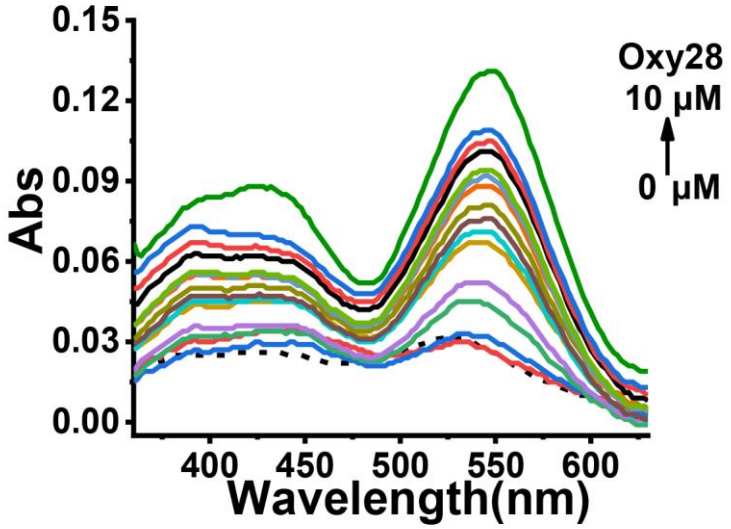

Figure S14. Absorption titration spectra of **5b** with Oxy28, the concentration of **5b** was 5.0 μM and the concentration of Oxy28 was 0, 0.25, 0.5, 0.75, 1.0, 1.25, 1.5, 1.75, 2.0, 2.5, 3.0, 4.0, 6.0, 8.0, 10.0 μM; dash lines represent the spectra of **5b** in the absence of Oxy28.

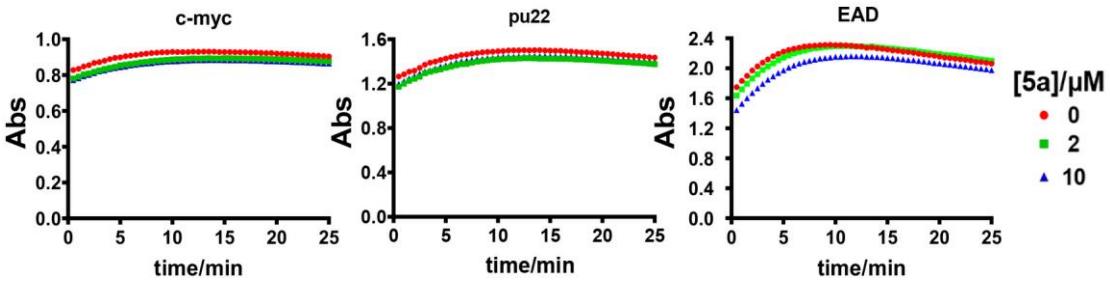

Figure S15. Inhibition activity of **5a** on the peroxidase activity of different G4/hemin complexes. The curves represent the change of $ABTS^{\bullet-}$ (absorbance at 415 nm) along with the reaction time.

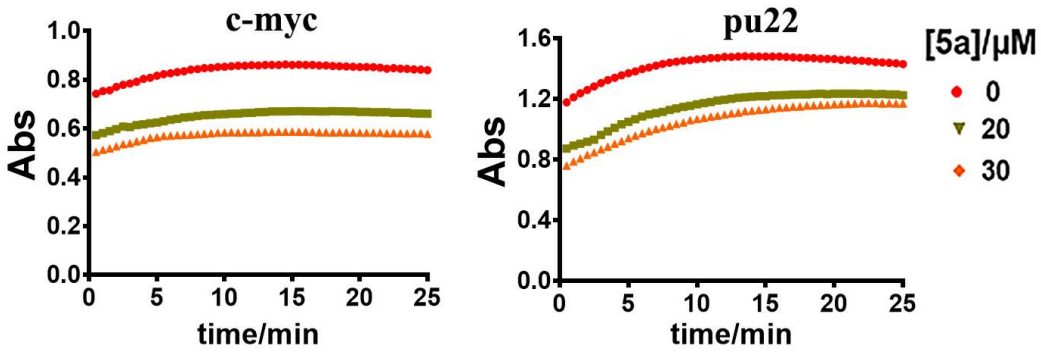

Figure S16. Inhibition activity of **5a** on the peroxidase activity of c-myc/hemin and pu22/hemin complexes. The curves represent the change of $ABTS^{\bullet-}$ (absorbance at 415 nm) along with the reaction time.

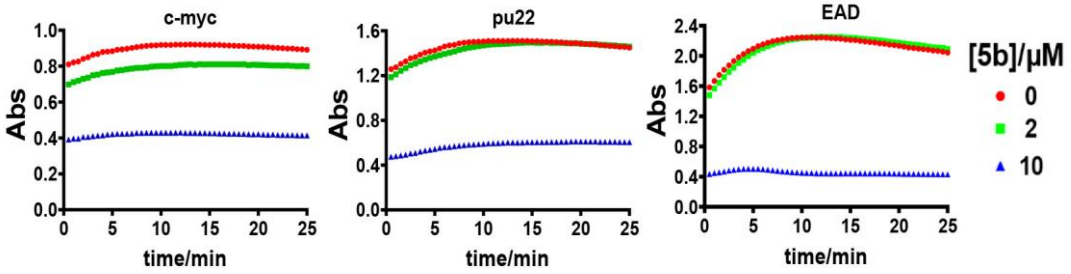

Figure S17. Inhibition activity of **5b** on the peroxidase activity of different G4/hemin complexes. The curves represent the change of $ABTS^{\bullet-}$ (absorbance at 415 nm) along with the reaction time.

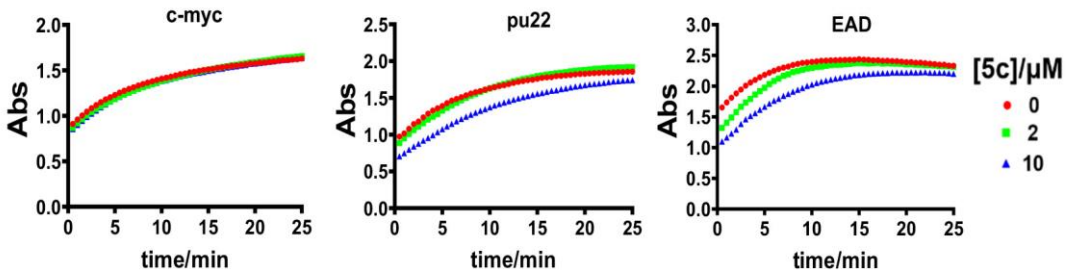

Figure S18. Inhibition activity of **5c** on the peroxidase activity of different G4/hemin complexes. The curves represent the change of $ABTS^{\bullet-}$ (absorbance at 415 nm) along with the reaction time.

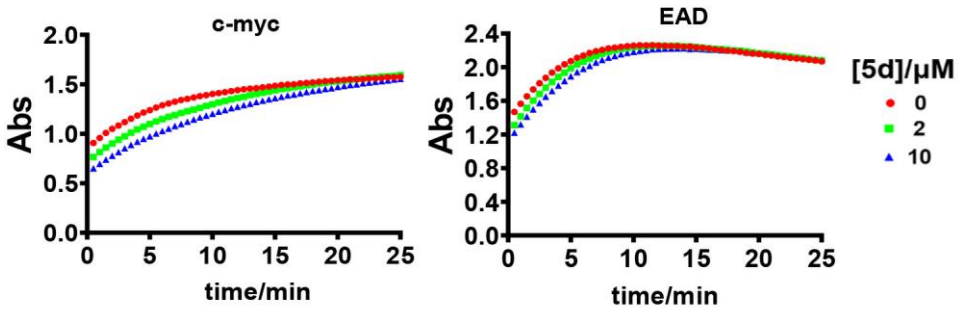

Figure S19. Inhibition activity of **5d** on the peroxidase activity of different G4/hemin complexes. The curves represent the change of $ABTS^{\bullet-}$ (absorbance at 415 nm) along with the reaction time.

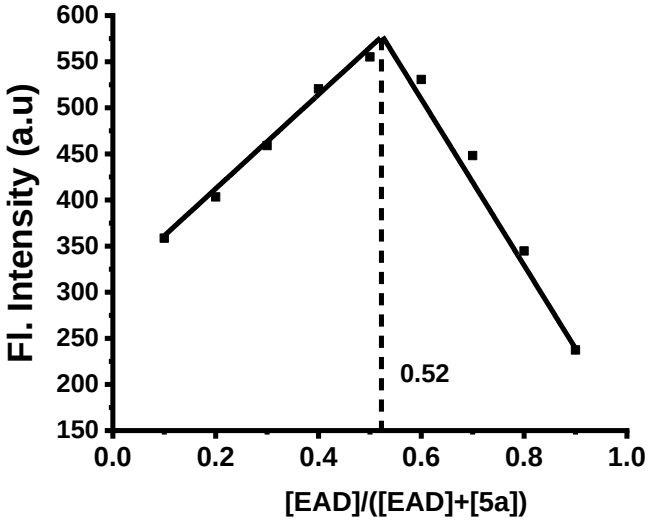

Figure S20. Jobs' plot obtained from fluorimetric analysis of mixtures of **5a** with EAD. The total concentration of **5a** and DNA is 5.0 μM; $\lambda_{ex/em}$ = 485/650 nm.

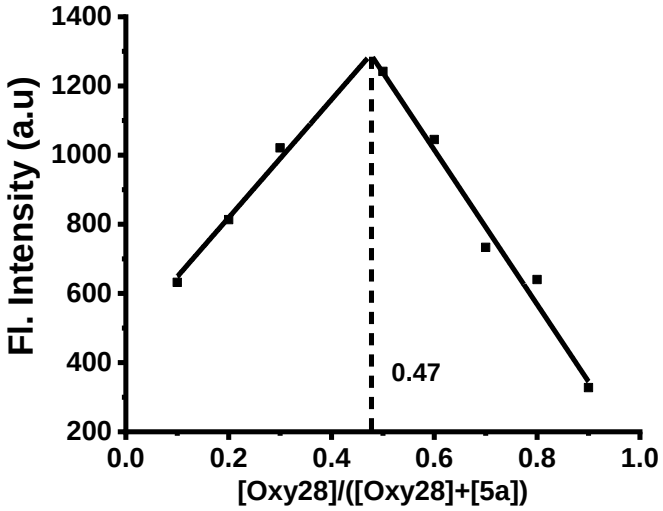

Figure S21. Jobs' plot obtained from fluorimetric analysis of mixtures of **5a** with Oxy28. The total concentration of **5a** and DNA is 5.0 μM; $\lambda_{ex/em}$ = 485/650 nm.

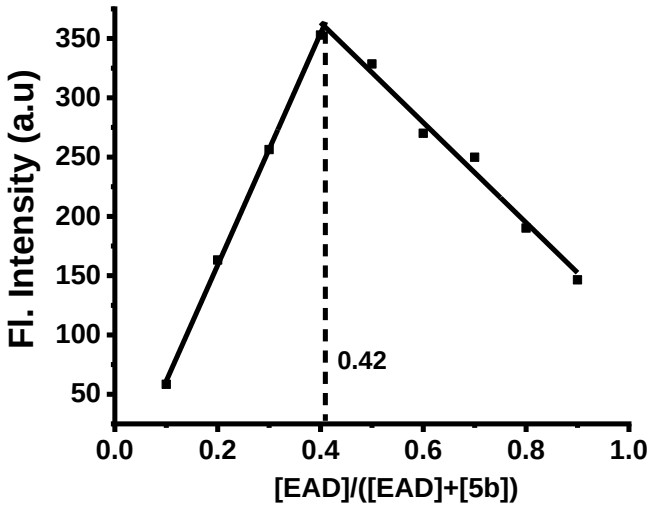

Figure S22. Jobs' plot obtained from fluorimetric analysis of mixtures of **5b** with EAD. The total concentration of **5b** and DNA is 5.0 μM; $\lambda_{ex/em}$ = 545/630 nm.

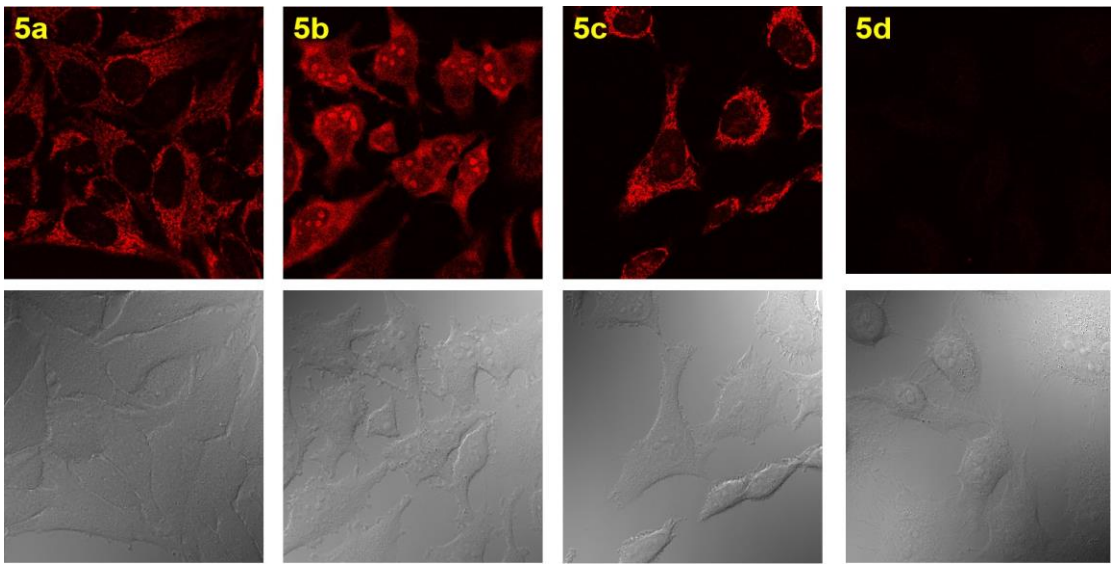

Figure S23. Confocal images of HeLa cells stained by **5a** (1.0 μM), **5b-5d** (5.0 μM) for 30 min. (**5a**/**5c** λ_{ex} = 488 nm; **5b**, λ_{ex} = 561 nm; **5d**, λ_{ex} = 405 nm).

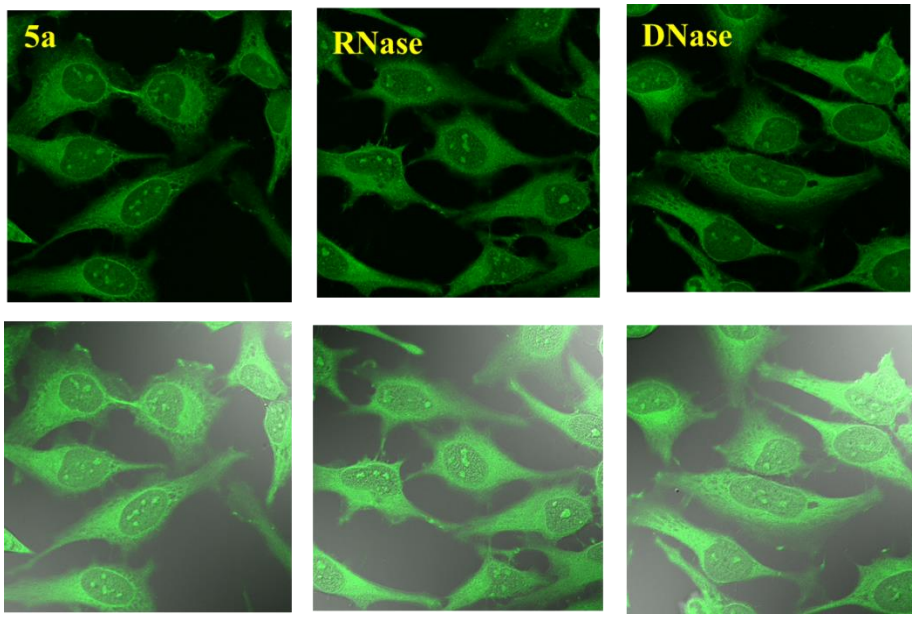

Figure S24. Confocal images of HeLa cells (fixed) stained with **5a** (1.0 μM) for 1h and treated without and with DNase I or RNase A.

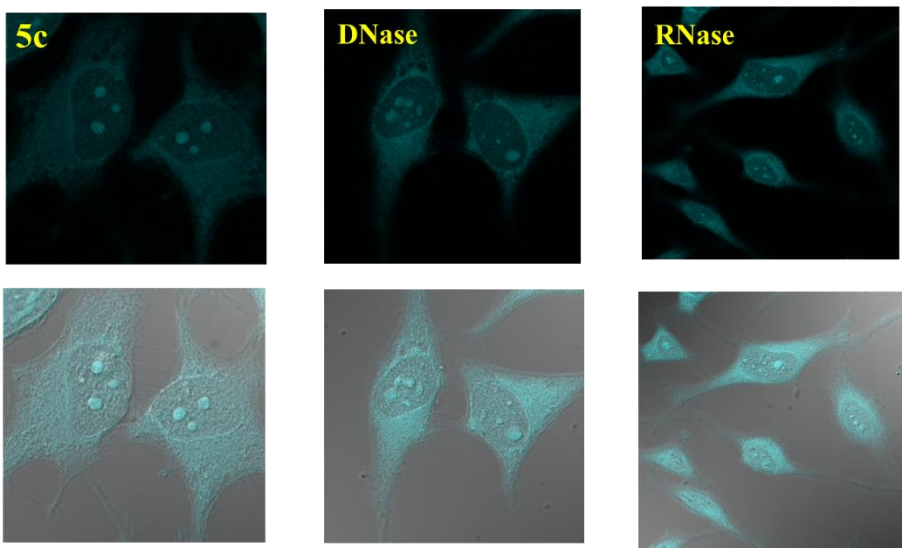

Figure S25. Confocal images of HeLa cells (fixed) stained with **5c** (1.0 μM) for 1h and treated without and with DNase I or RNase A.

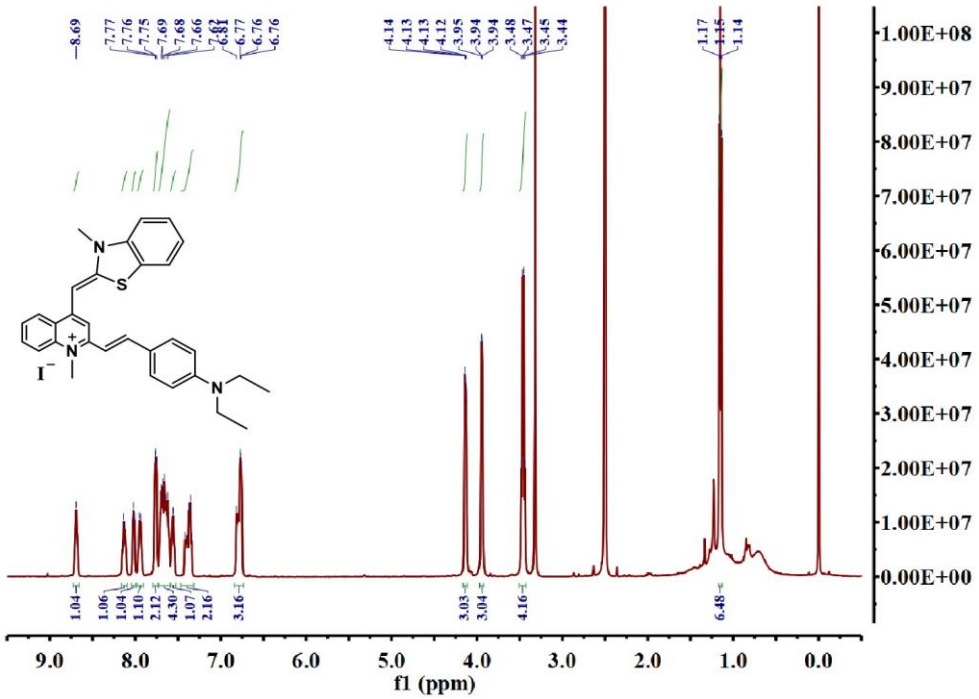

Figure S26. ^{1}H NMR spectra of **5a**.

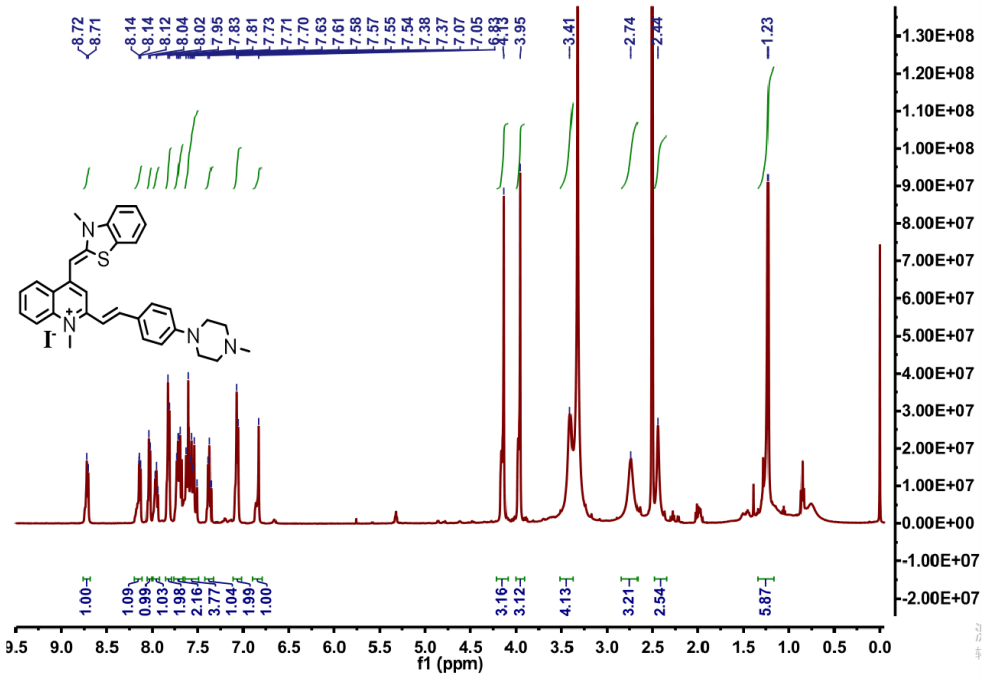

Figure S27. ^{1}H NMR spectra of **5b.**

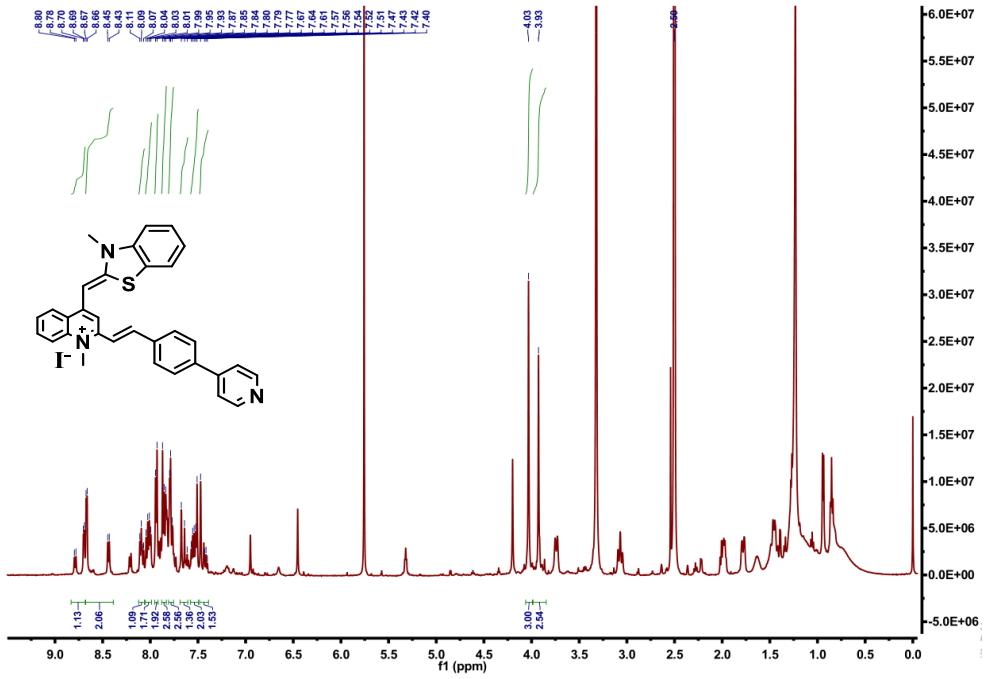

Figure S28. ^{1}H NMR spectra of **5c.**

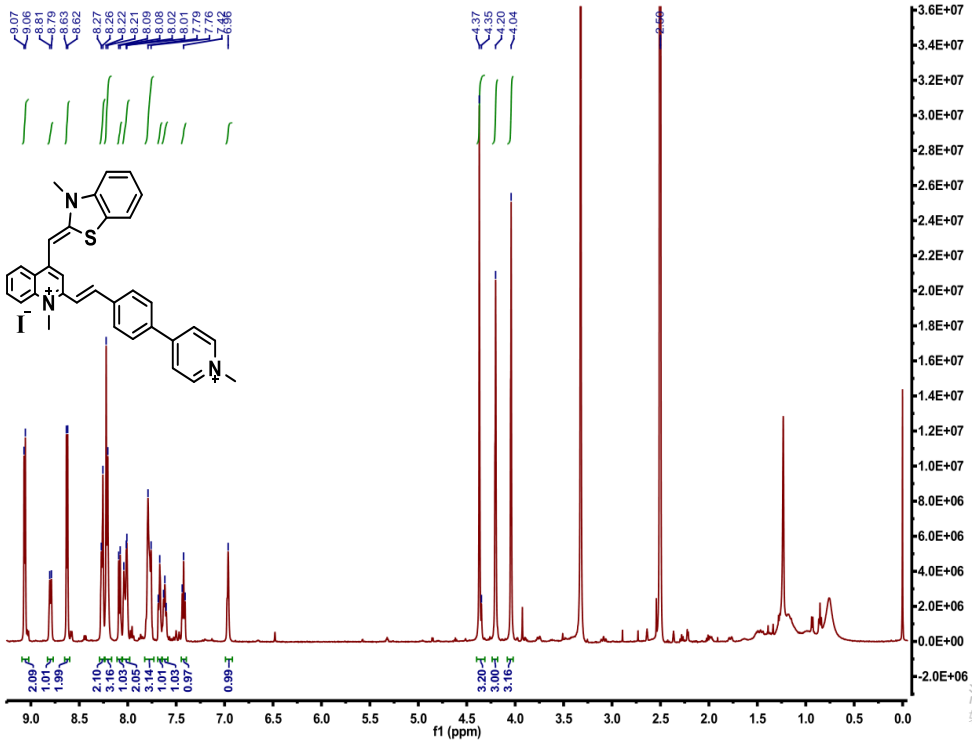

Figure S29. ^{1}H NMR spectra of **5d.**