Supplementary Information

Mitochondria-targeted turn-on fluorescent probe based on a rhodol platform for the detection of copper(I)

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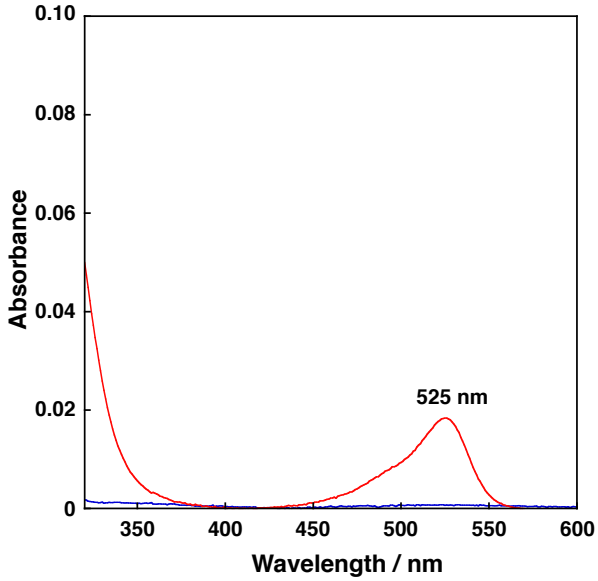

Fig. S1 UV-vis spectrrum of 1 μM RdlTPA-Et_2 (blue line) in 50 mM HEPES (pH 7.20) containing 2 mM glutathione (GSH). Red spectrum was observed upon reaction with 20 μM $[Cu^{I}(CH_3CN)_4]PF_6$ for 3 h.

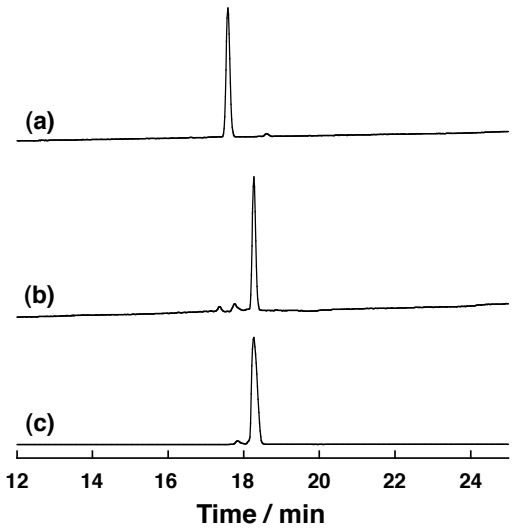

Fig. S2 HPLC chromatograms of the reaction mixture of RdlTPA-Et_2 and $[Cu^{I}(CH_3CN)_4]PF_6$. (a) Before addition of $[Cu^{I}(CH_3CN)_4]PF_6$. (b) Reaction mixture after incubation for 3 h at room temperature. (c) HMDER as the authentic sample. The reaction was carried out in 50 mM HEPES (pH 7.20) containing 2 mM GSH. The HPLC gradient was as follows: solvent A (0.1 % TFA in water), solvent B (0.1 % TFA in CH_3CN); 12-25 min, 0-100% B. The peaks were monitored by UV absorbance at 525 nm.

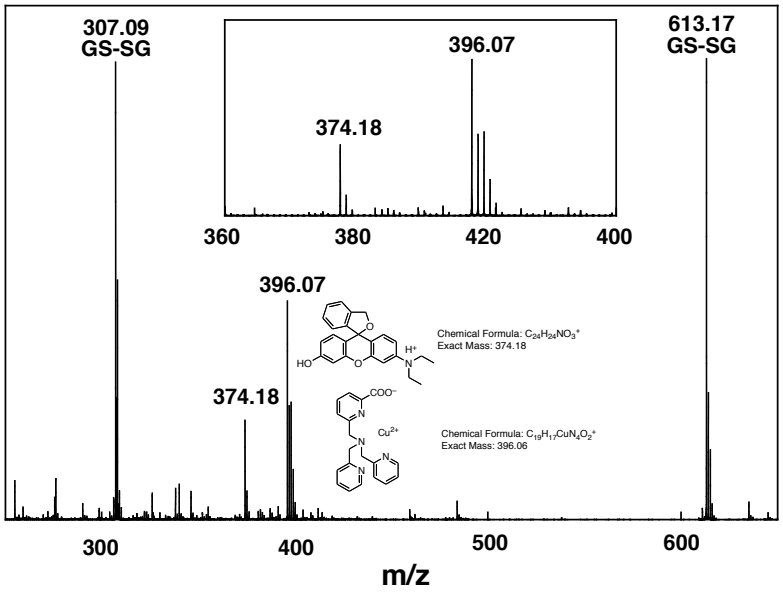

Fig. S3 ESI mass spectra (positive ion mode) for the reaction of 2 μM RdlTPA-Et_2 and 10 μM $[Cu^{I}(CH_3CN)_4]PF_6$ in 10 mM NH_4OAc in the presence of 100 μM GSH. The incubation time was 3 h. Inset: expansion of ESI-MS chromatogram in the range between 360 and 400. Mass peaks observed at 374.18 and 396.07 were attributed to HMDER ($[M+H]^+$) and the copper complex with oxidized TPA ligand ($[M]^+$), respectively. Peaks at 307.09 ($[M+2H]^{2+}$) and 613.17 ($[M+H]^+$) were attributed to the oxidized form of GSH (GS-SG).

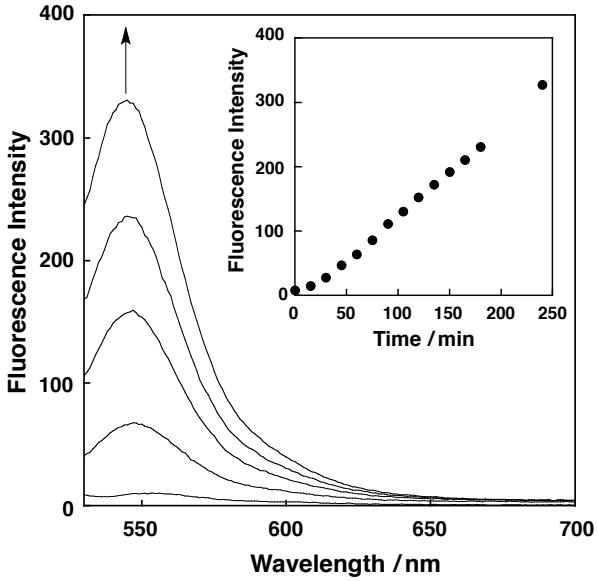

Fig. S4 Fluorescence spectral changes of 1 μM RdlTPA-Et_2 upon addition of 1 μM $[Cu^{I}(CH_3CN)_4]PF_6$ in 50 mM HEPES (pH 7.20) containing 10 mM GSH. The spectra were measured every 60 min with the excitation wavelength at 525 min. Inset: Time course of the fluorescence intensity at 542 nm.

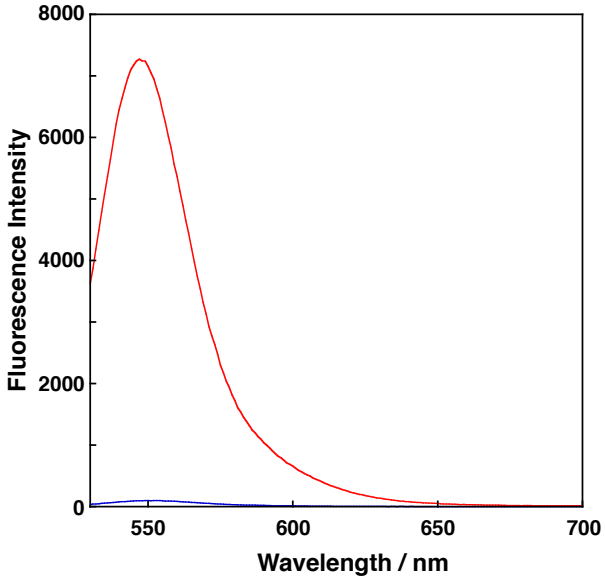

Fig. S5 Fluorescence response of 2 μM RdlTPA-TPP (blue line) to 20 μM $[Cu^{I}(CH_3CN)_4]PF_6$. A red spectrum was recorded after a 3 h reaction of RdlTPA-TPP with Cu^{I} in 50 mM HEPES (pH 7.20) containing 2 mM GSH. The excitation wavelength was 525 nm.

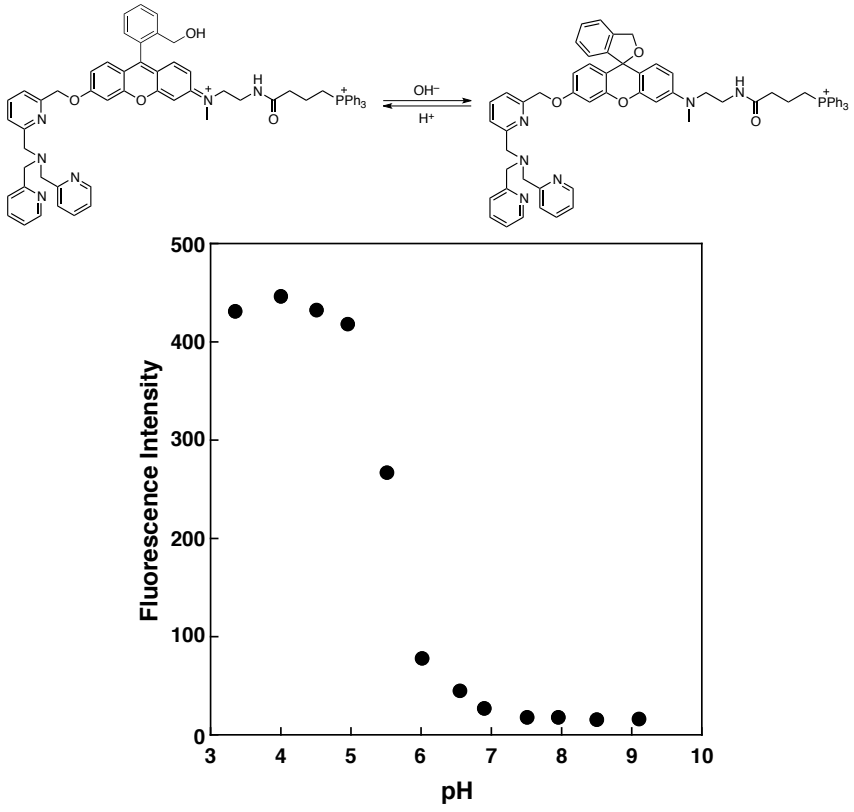

Fig. S6 Effect of pH on emission intensity at 542 nm of 1 μM RdlTPA-Et_2 in aqueous solutions, λ_{ex} = 525 nm.

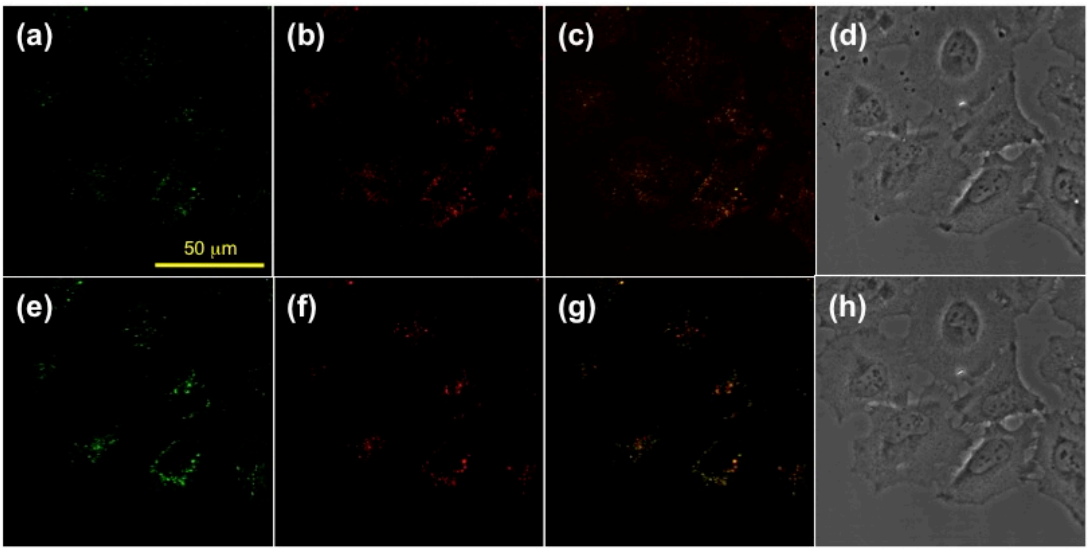

Fig. S7 Confocal fluorescence images of HeLa cells (a-d) before and (e-h) after treatment with CCCP. The cells were co-incubated with 10 μM RdlTPA-TPP and LysoTracker Red for 3 h at 37 °C. (a, d) RdlTPA-TPP; (b, f) LysoTracker Red; (c, g) merged image of RdlTPA-TPP and LysoTracker Red; (d, h) brightfield.

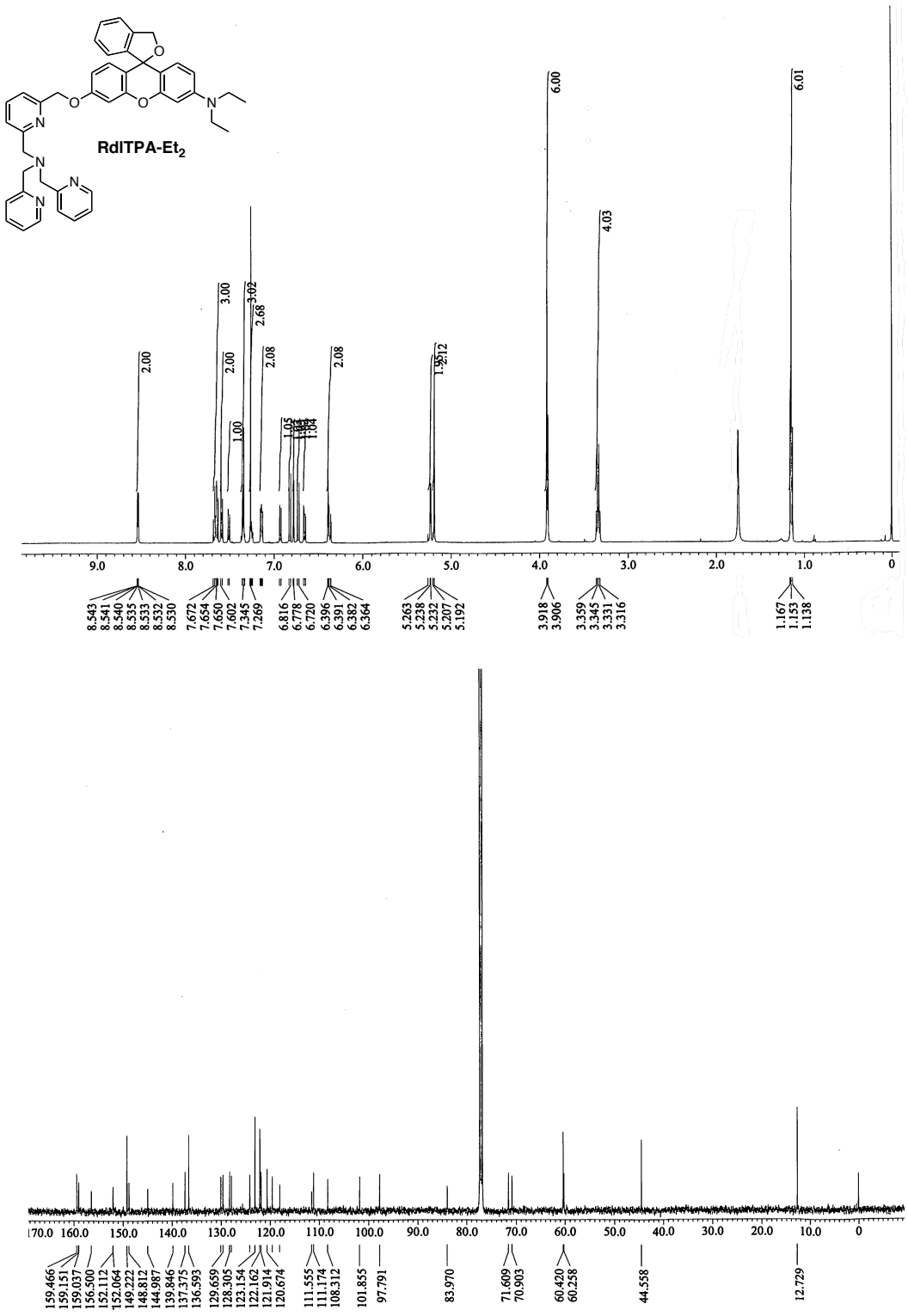

Figure S7. ^{1}H (top) and ^{13}C (bottom) spectra of RdlTPA-Et_2 in $CDCl_3$.

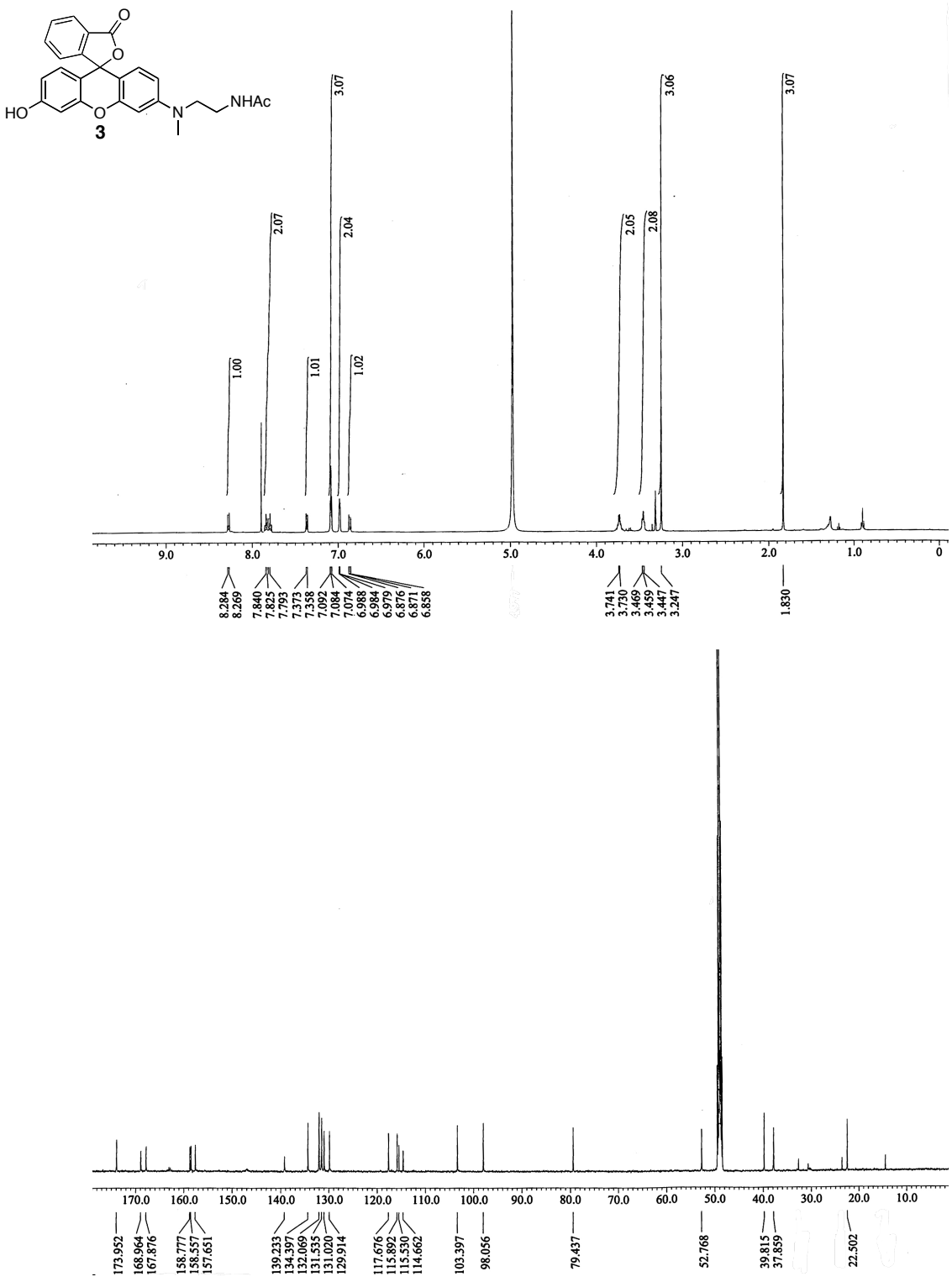

Figure S8. ^{1}H (top) and ^{13}C (bottom) spectra of **3** in CD_3OD.

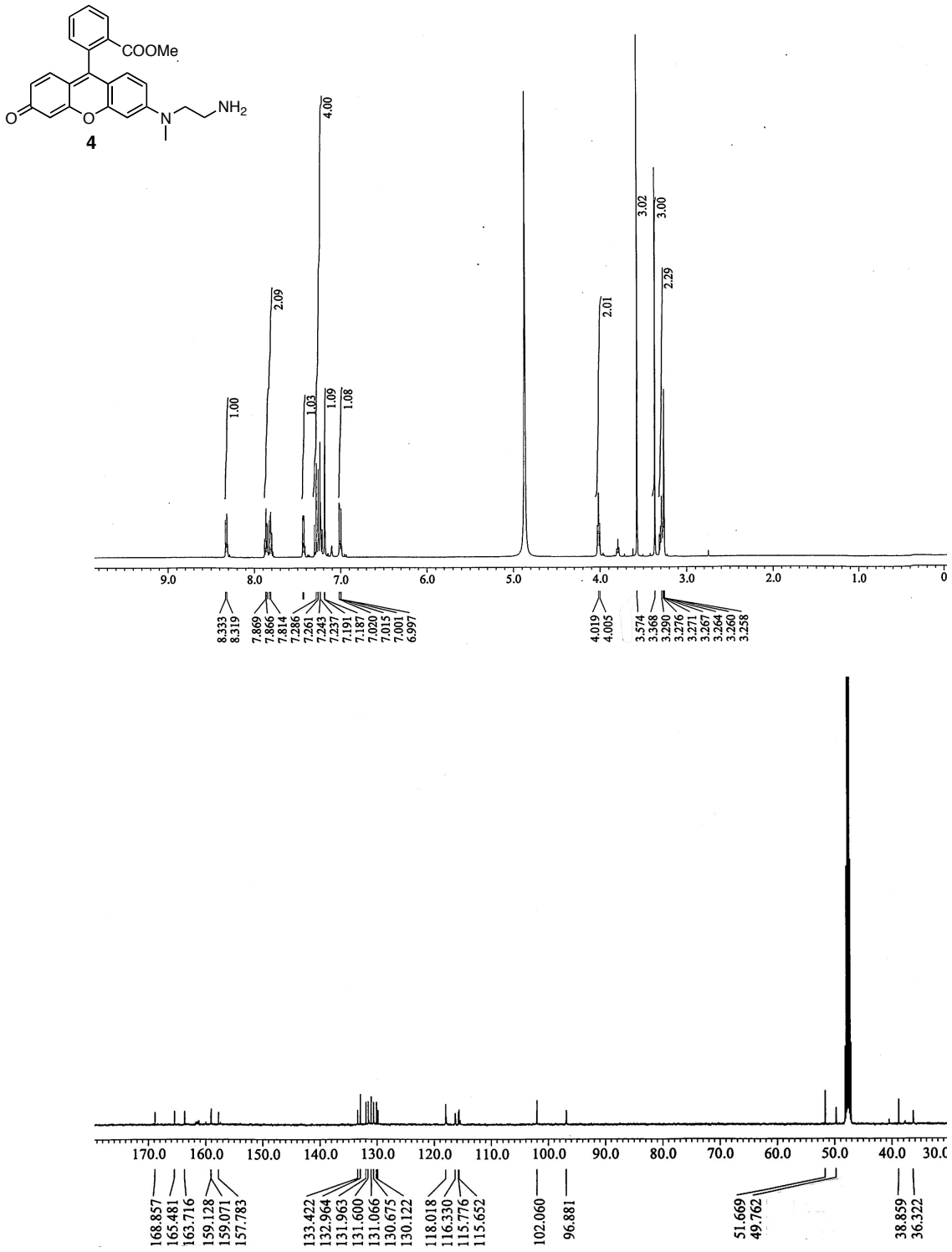

Figure S9. ^{1}H (top) and ^{13}C (bottom) spectra of **4** in CD_3OD.

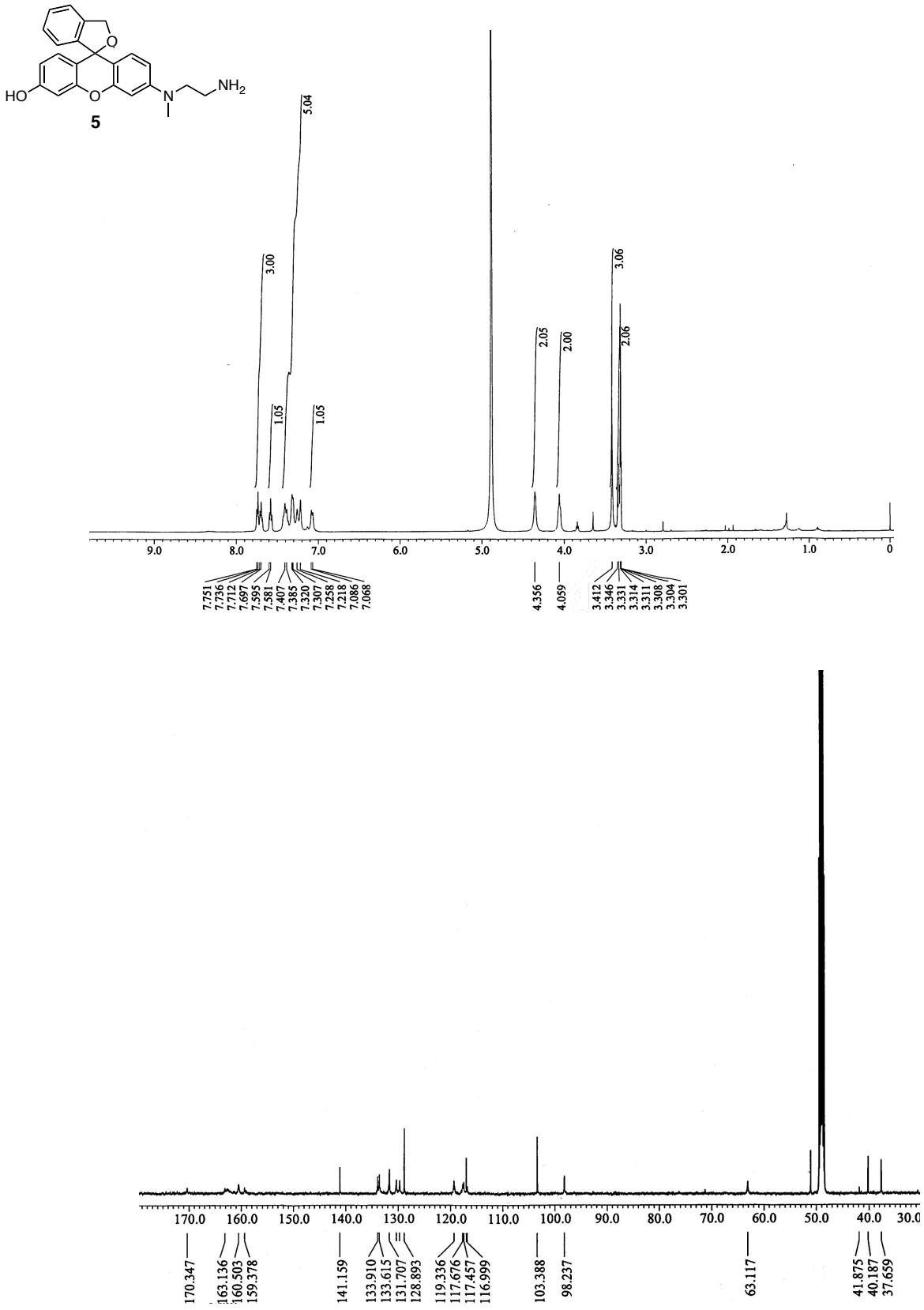

Figure S10. ^{1}H (top) and ^{13}C (bottom) spectra of **5** in CD_3OD.

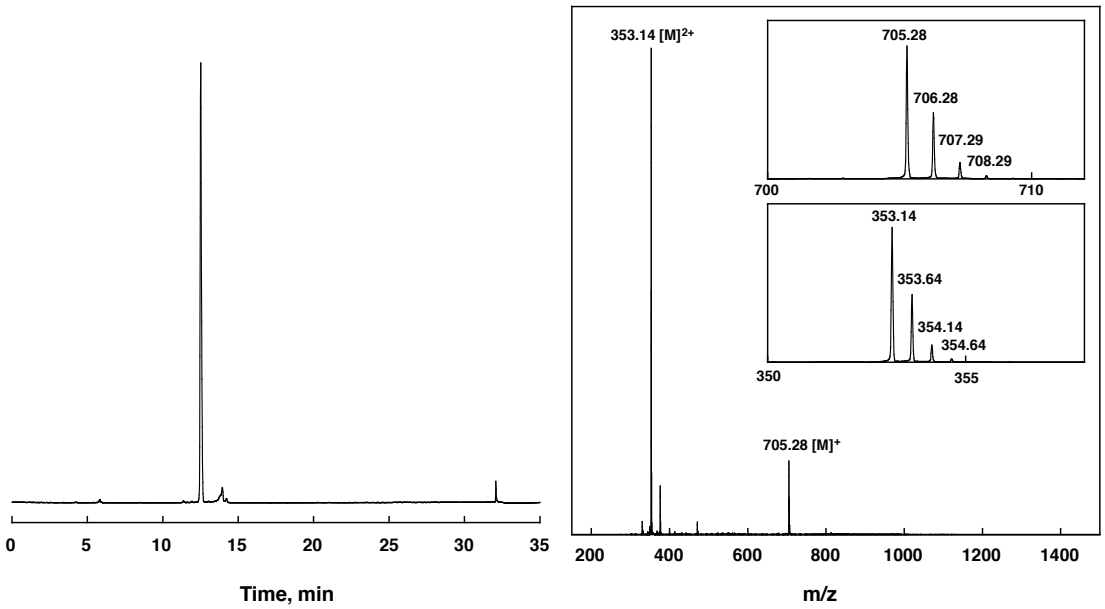

Figure S11. (left) HPLC chart and of isolated Rdl-TPP eluted with water/CH_3CN containing 0.1% TFA. (right) HR-ESI mass spectrum (pos.) of Rdl-TPP.

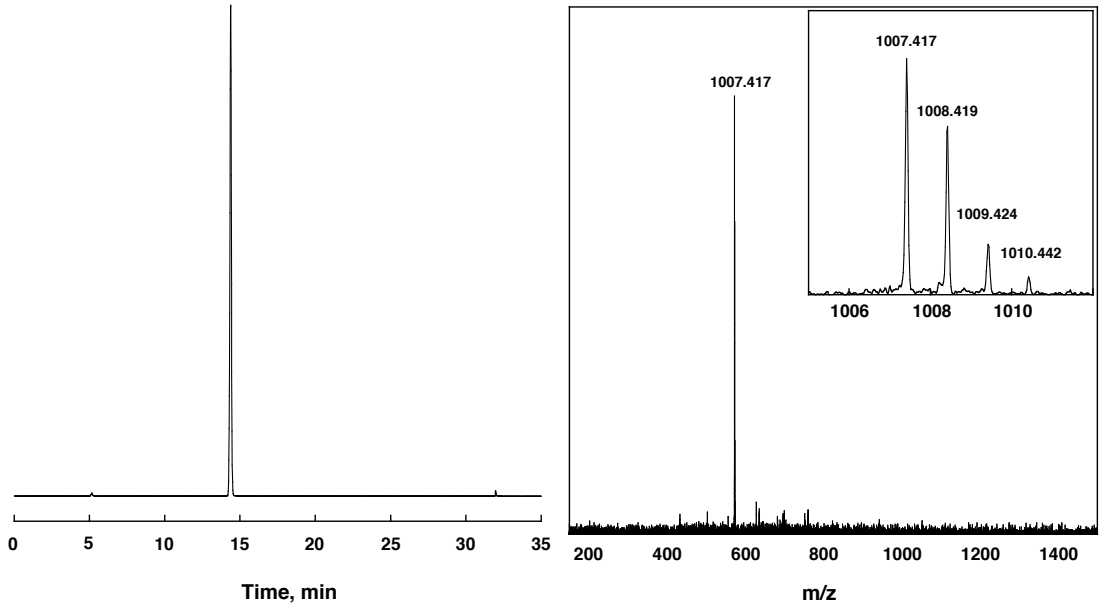

Figure S12. (left) HPLC chart and of isolated RdlTPA-TPP eluted with water/CH_3CN containing 0.1% TFA. (right) HR-ESI mass spectrum (pos.) of RdlTPA-TPP.