

Electronic Supporting Information

Design and synthesis of BODIPY-Clickates based Hg²⁺ sensors: Effect of triazole binding mode with Hg²⁺ on signal transduction

Mani Vedamalai,^a Dhaval Kedaria,^b Rajesh Vasita,^b Shigeki Mori^c and Iti Gupta^{a,*}

^{a,*}Indian Institute of Technology Gandhinagar, VGEC Campus, Chandkheda, Ahmedabad-382424, India.

Corresponding author E.mail: iti@iitgn.ac.in.

^b School of Life Sciences, Central University of Gujarat, Gandhinagar, Gujarat, India.

^c Integrated Centre for Sciences, Ehime University, Matsuyama, 790-8577, Japan.

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Empirical Formula	C ₁₉ H ₁₈ BCl ₂ F ₂ N ₃
Formula Weight	408.08
Crystal Color, Habit	orange, platelet
Crystal Dimensions	0.250 X 0.160 X 0.020 mm
Crystal System	orthorhombic
Lattice Type	Primitive
Lattice Parameters	a = 25.408(9) Å b = 8.297(3) Å c = 8.766(3) Å V = 1848.0(11) Å ³
Space Group	Pca2 ₁ (#29)

Table S1. Crystal data refinement parameters of Compound **3**

Empirical Formula	C ₁₉ H ₁₈ BF ₂ N ₉
Formula Weight	421.22
Crystal Color, Habit	orange, needle
Crystal Dimensions	0.200 X 0.100 X 0.020 mm
Crystal System	monoclinic
Lattice Type	Primitive
Lattice Parameters	a = 12.295(6) Å b = 20.765(10) Å c = 7.858(4) Å β = 100.122(9) ° V = 1975.1(17) Å ³
Space Group	P2 ₁ /c (#14)

Table S2. Crystal data refinement parameters of Compound 4

Solvents	F1			F2			4			3		
	λ_{abs} nm	λ_{em} nm	Φ_{f}	λ_{abs} nm	λ_{em} nm	Φ_{f}	λ_{abs} nm	λ_{em} nm	Φ_{f}	λ_{abs} nm	λ_{em} nm	Φ_{f}
Ethanol	495	509	0.004	495	513	0.004	494	500	0.002	495	513	0.003
Acetonitrile	491	513	0.001	492	495	0.001	492	515	0.001	493	500	0.001
Methanol	494	515	0.002	494	514	0.002	493	513	0.001	493	504	0.001
DCM	498	605	0.072	498	605	0.114	497	615	0.020	497	616	0.050
Hexanes	497	511	0.107	509	512	0.136	497	511	0.190	497	511	0.162
THF	496	623	0.028	496	618	0.036	496	511, 627	0.013	496	629	0.019
DMF	494	504	0.001	494	508	0.001	493	506	0.001	495	505	0.001

Table S3. Photophysical properties of **F1**, **F2**, **4**, and **3** in various solvents.

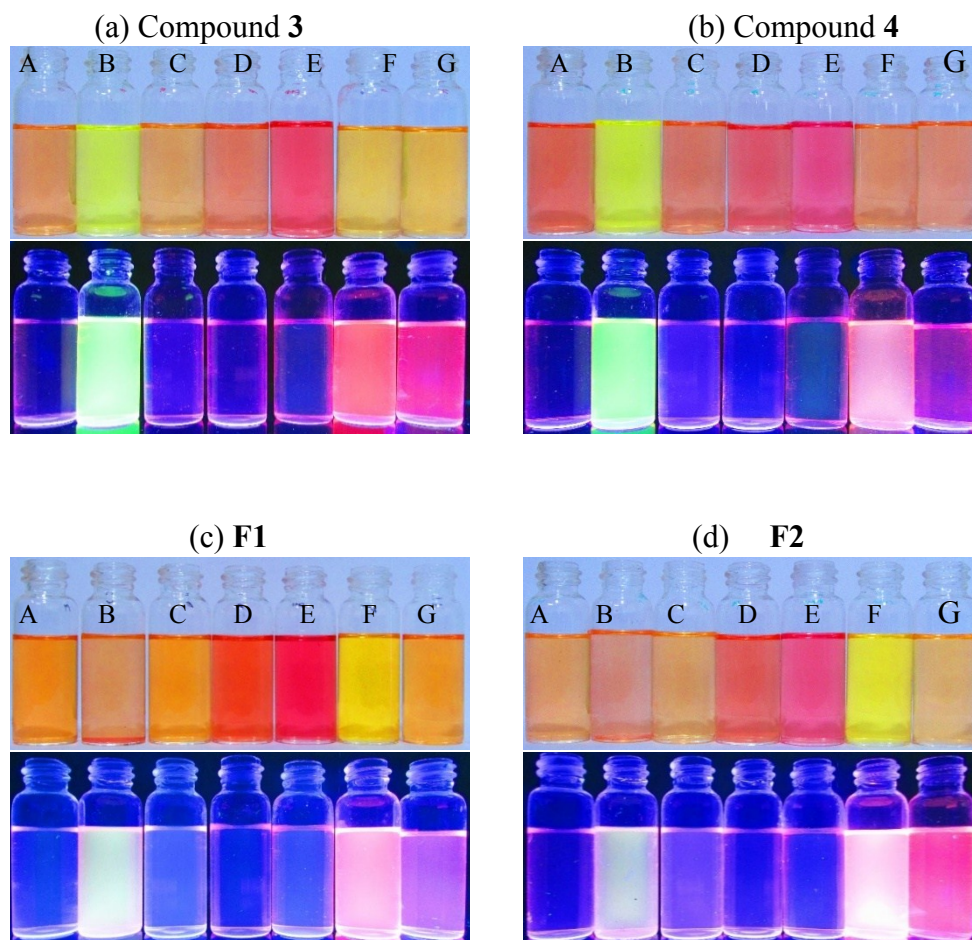


Figure S1. Solvatochromism of compound **3**, **4**, **F1** and **F2** in various solvents (A) methanol, (B) hexanes (C) ethanol, (D) acetonitrile, (E) dimethyl formamide, (F) dichloromethane, and (G) tetrahydrofuran. Visible (top) and long wavelength (below) pictures of compound **3** (a), **4** (b), **F1** (c) and **F2** (d) in various solvents.

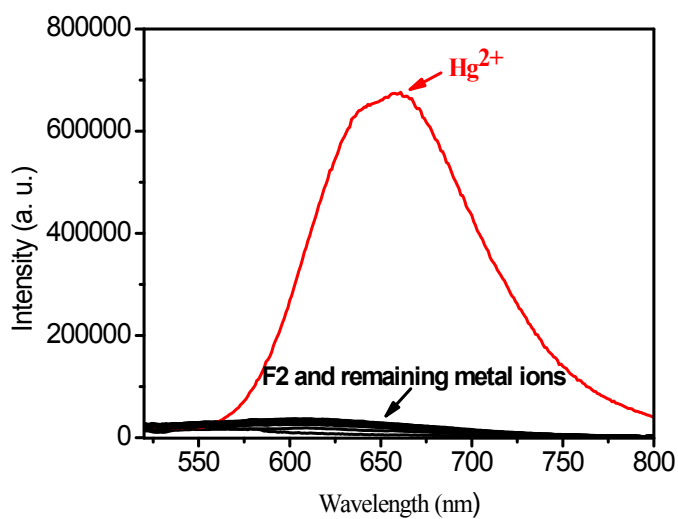


Figure S2. Fluorescence spectra of **F2** (10 μM) upon addition of 3 equivalent of various metal ions (Ag^+ , Ca^{2+} , Cd^{2+} , Co^{2+} , Cu^{2+} , Fe^{3+} , Fe^{2+} , K^+ , Mg^{2+} , Mn^{2+} , Ni^{2+} , Pb^{2+} , Zn^{2+} and Hg^{2+} in methanol ($\lambda_{\text{ex}} = 494 \text{ nm}$).

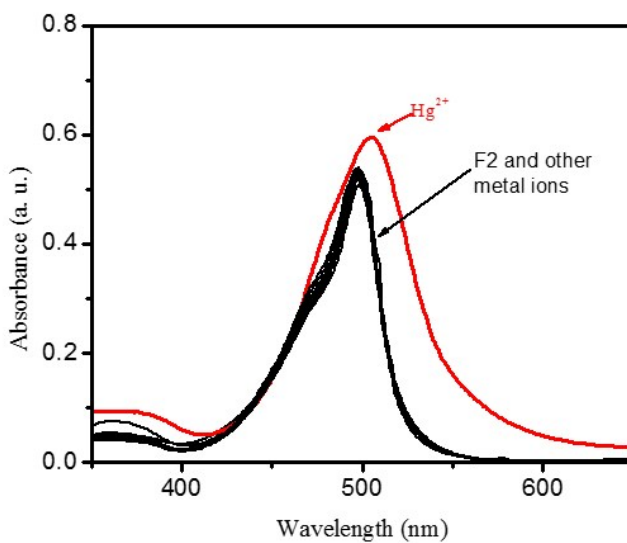


Figure S3. UV-visible spectra of **F2** (10 μM) upon addition of 3 equivalent of various metal ions (Ag^+ , Ca^{2+} , Cd^{2+} , Co^{2+} , Cu^{2+} , Fe^{3+} , Fe^{2+} , K^+ , Mg^{2+} , Mn^{2+} , Ni^{2+} , Pb^{2+} , Zn^{2+} and Hg^{2+} in methanol.

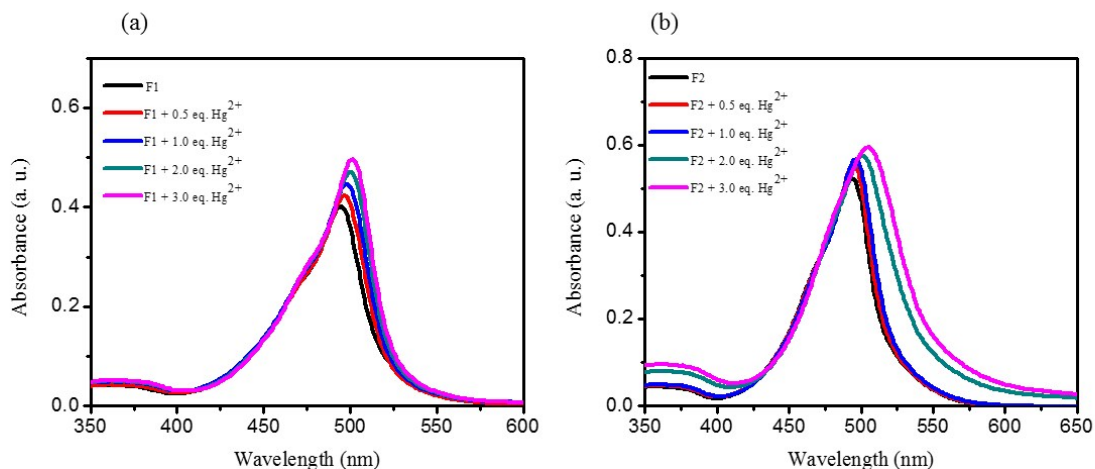


Figure S4. (a) UV-visible response of **F1** (10 μM) in the presence of different amounts of Hg²⁺ in methanol. (b) UV-visible response of **F2** (10 μM) in the presence of different amounts of Hg²⁺ in methanol.

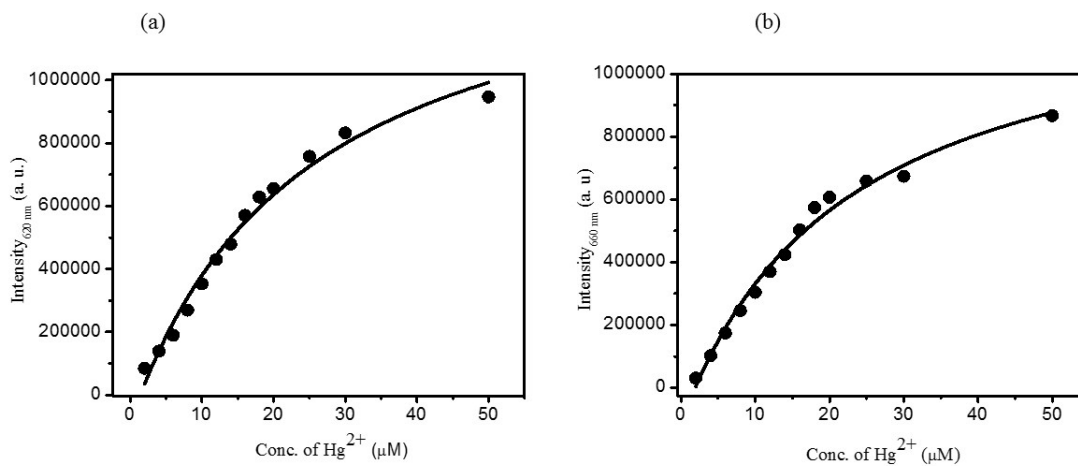


Figure S5. (a) Fluorescence responses of **F1** (10 μM) to Hg²⁺ solutions for K_d value calculation. Excitation was 494 nm. Spectra were acquired against increasing amount of Hg²⁺ 0, 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 25, 30, and 50 μM. The apparent K_d value was found to be 24.4 ± 5.1 μM. (b) Fluorescence responses of **F2** (10 μM) to Hg²⁺ solutions for K_d value calculation. Excitation was 494 nm. Spectra were acquired against increasing amount of Hg²⁺ 0, 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 25, 30, and 50 μM. The apparent K_d value was found to be 22.0 ± 3.9 μM.

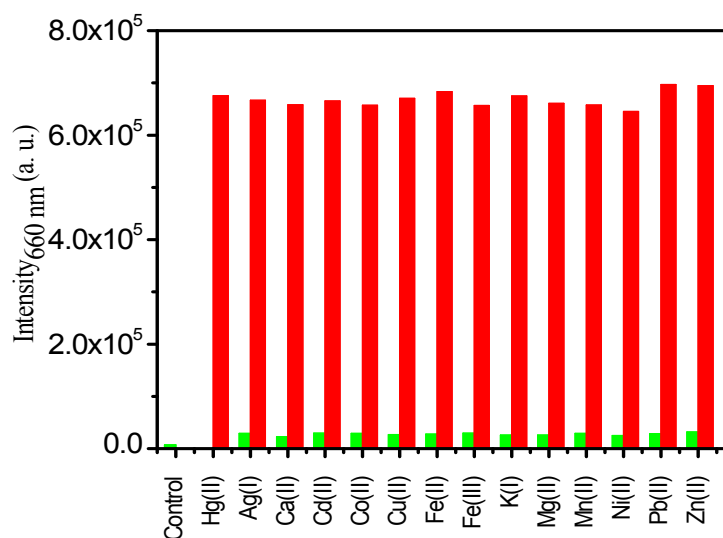


Figure S6. Relative fluorescence intensities of **F2** (10 μM) in the presence of Hg^{2+} (3 equiv.) and various metal ions (15 equiv.) in methanol, ($\lambda_{\text{ex}} = 494 \text{ nm}$). Green bars represent **F2** and 150 μM of other competing metal ions, red bars represent subsequent addition of 30 μM of Hg^{2+} to **F2** and **F2** with competing metal ions (150 μM).

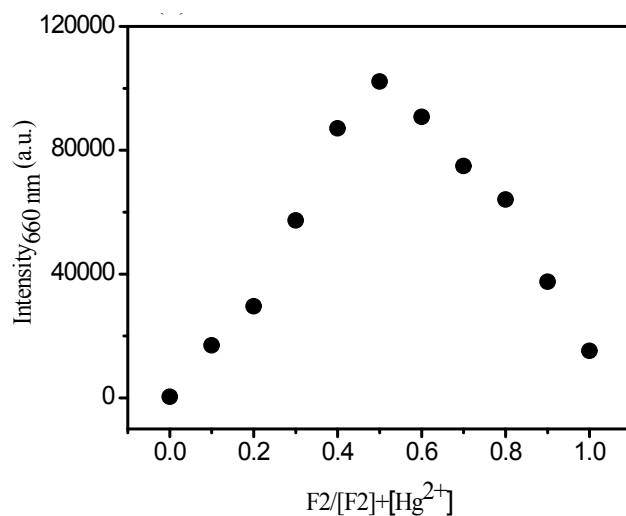


Figure S7. Job plot of **F2-Hg²⁺** complexes in methanol ($\lambda_{\text{ex}} = 494 \text{ nm}$). The total concentration of **F2** and Hg^{2+} was 10 μM

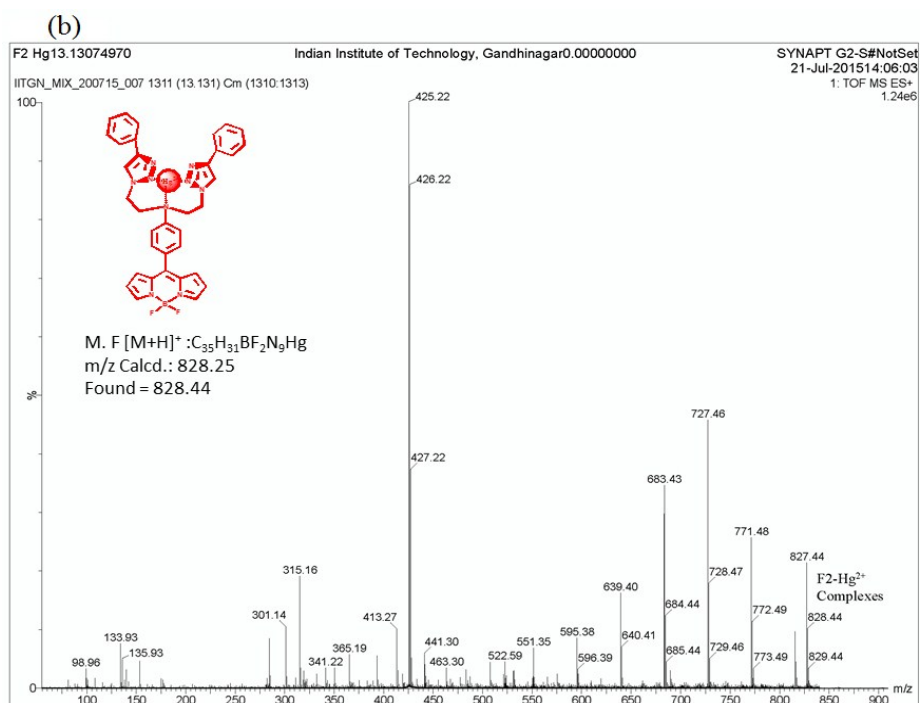
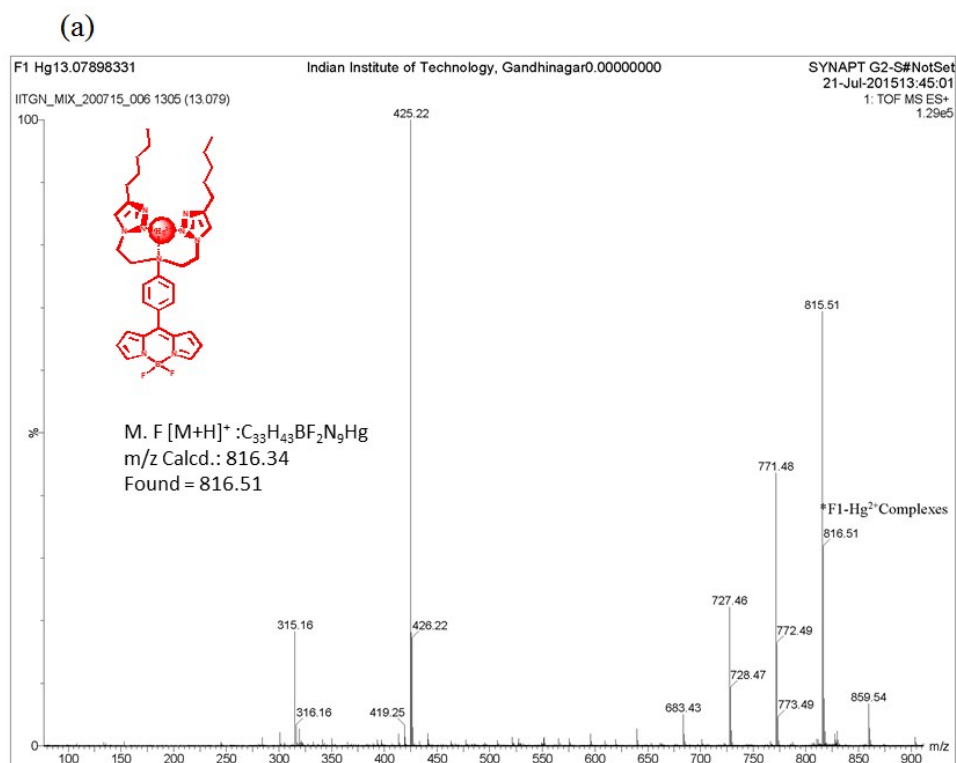


Figure S8. (a) ESI MS spectra of **F1-Hg²⁺** Complexes. (b) ESI MS spectra of **F2-Hg²⁺** Complexes.

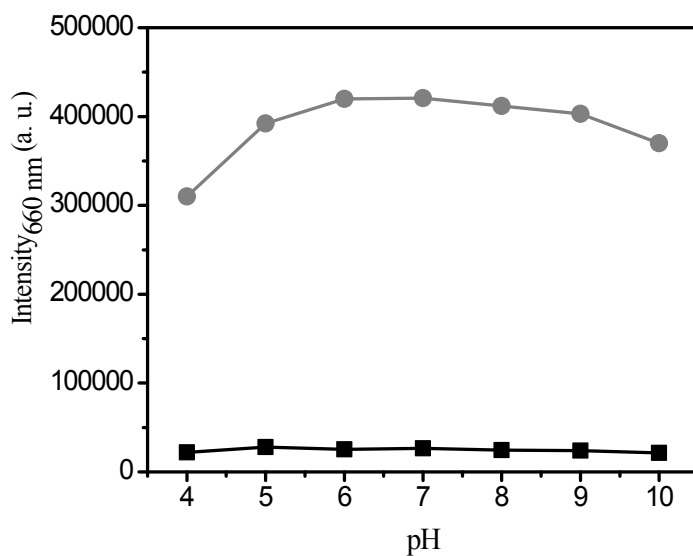


Figure S9. Fluorescence intensity of **F2** (10 μM ; ■) and after addition of Hg^{2+} (30 μM , ●) in methanol-water (9:1, v/v, 2 mM HEPES) medium as a function of different pH values. Excitation wavelength was 494 nm.

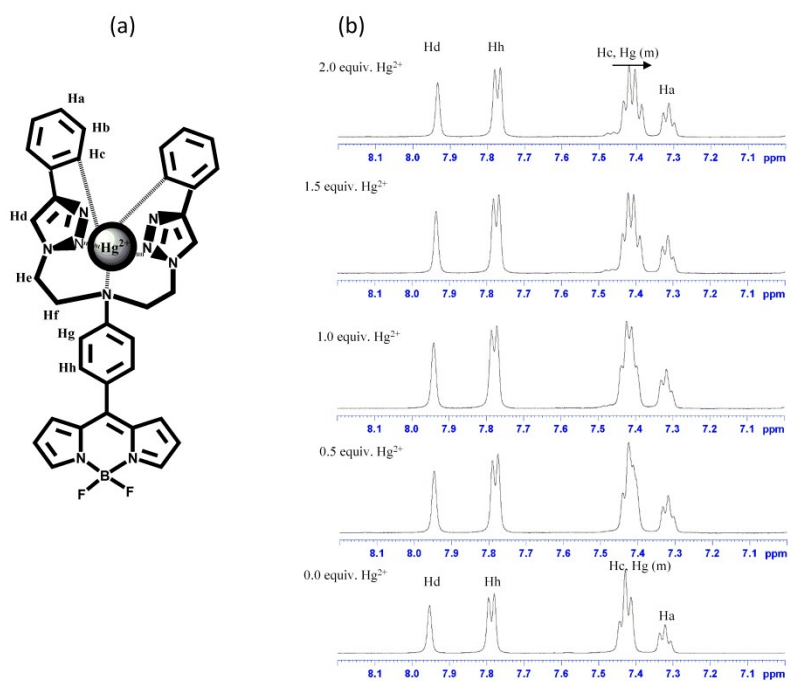


Figure S10. (a) Binding model; (b) Partial ^1H NMR spectra of **F2** (5mM) in the absence or presence of increasing Hg^{2+} in DMSO-d_6 .

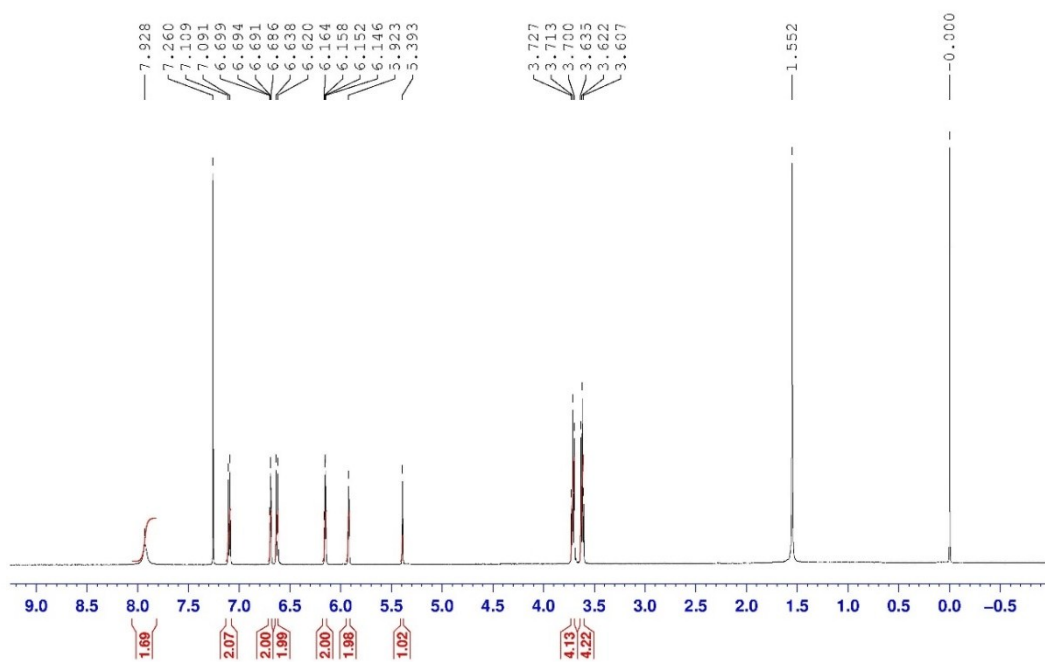


Figure S11. ^1H spectrum of Compound **2** in CDCl_3

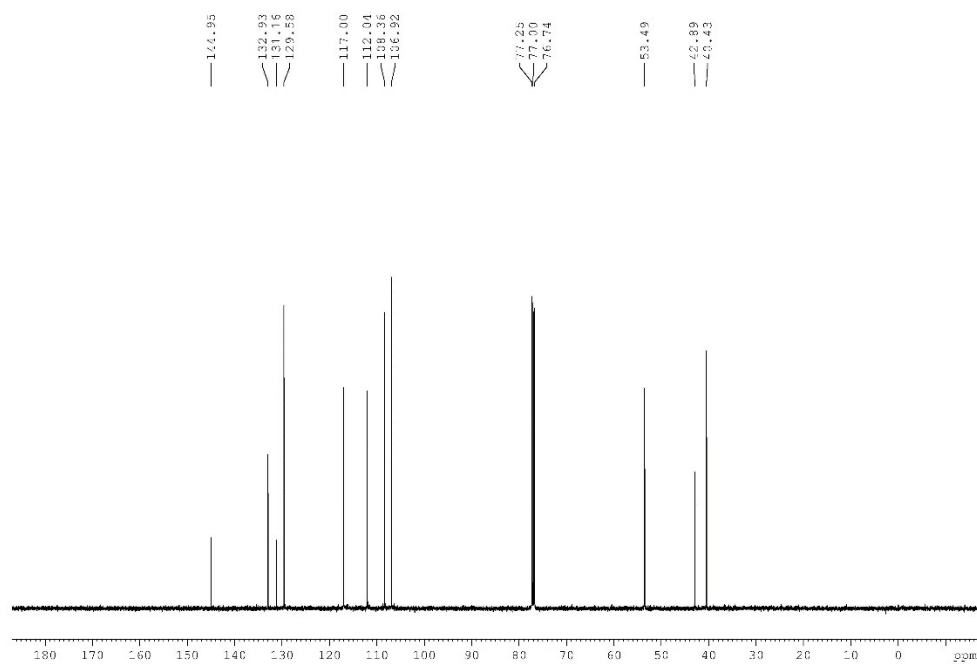


Figure S12. ^{13}C spectrum of Compound **2** in CDCl_3

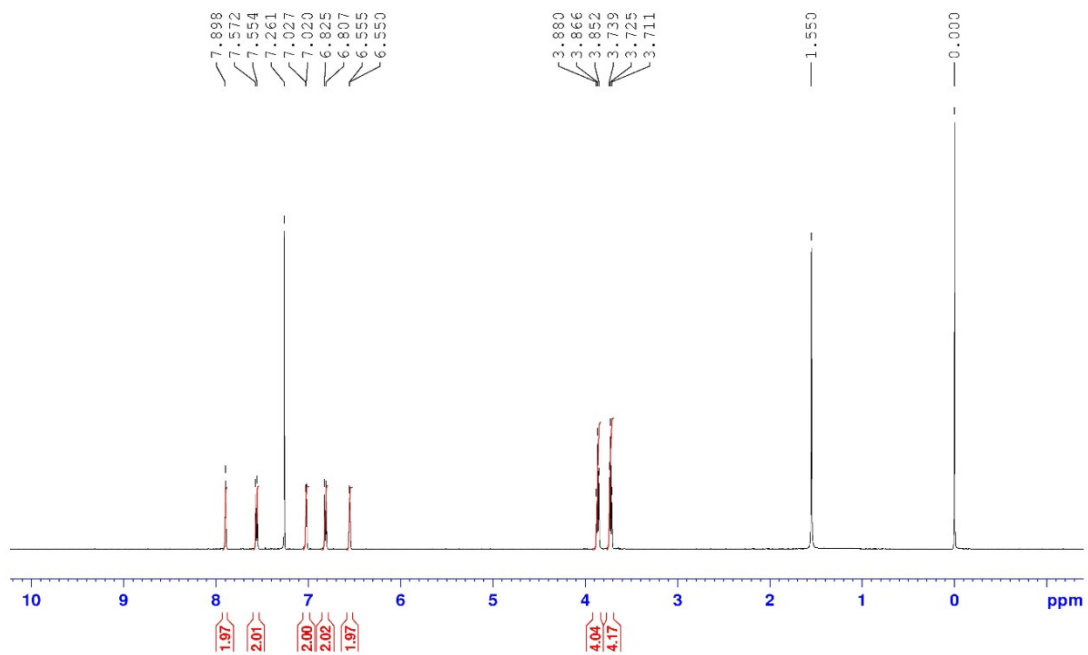


Figure S13. ^1H spectrum of Compound **3** in CDCl_3

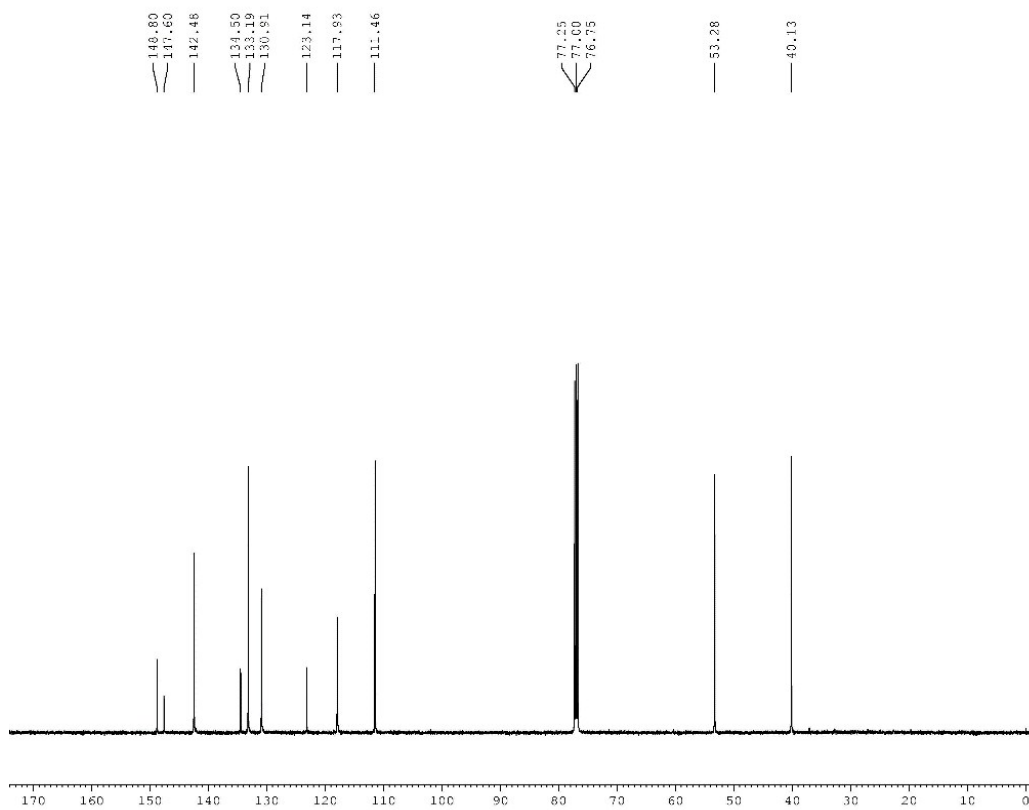


Figure S14. ^{13}C spectrum of Compound **3** in CDCl_3

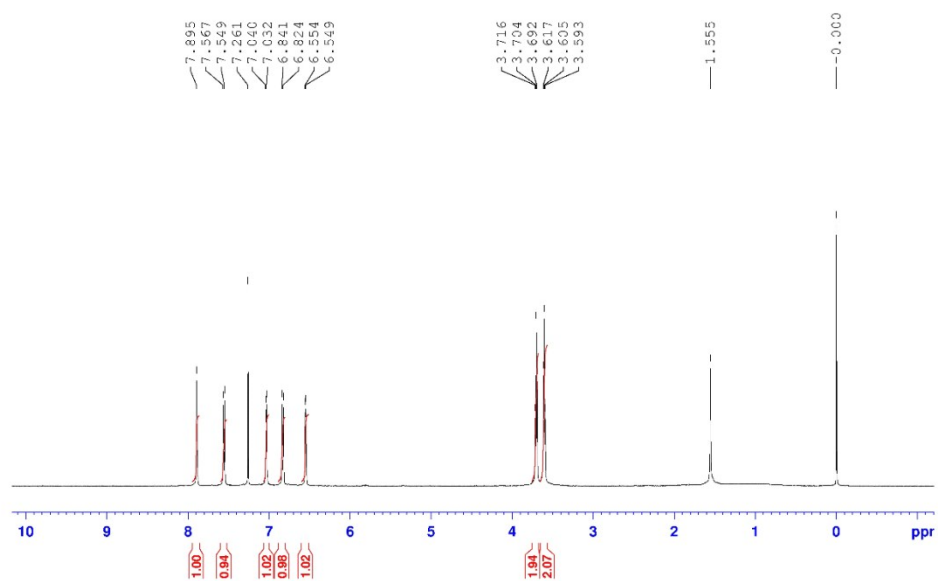


Figure S15. ^1H spectrum of Compound **4** in CDCl_3

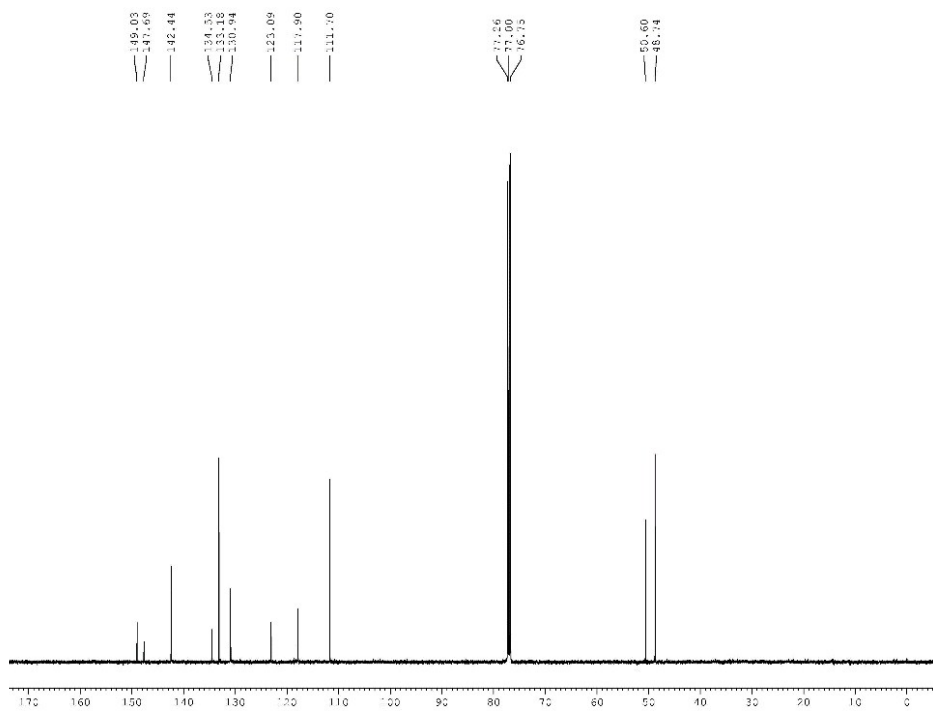


Figure S16. ^{13}C spectrum of Compound **4** in CDCl_3

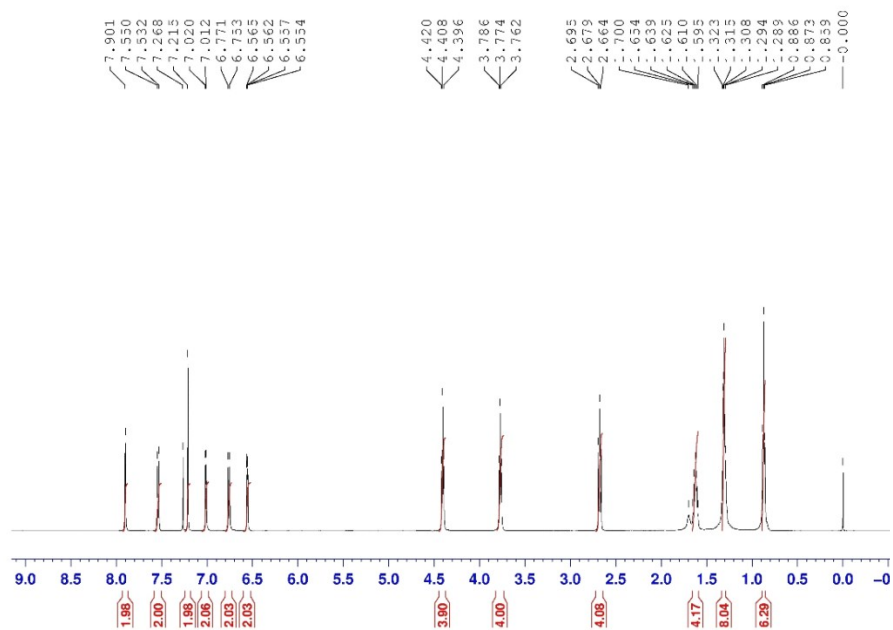


Figure S17. ^1H spectrum of F1 in CDCl_3

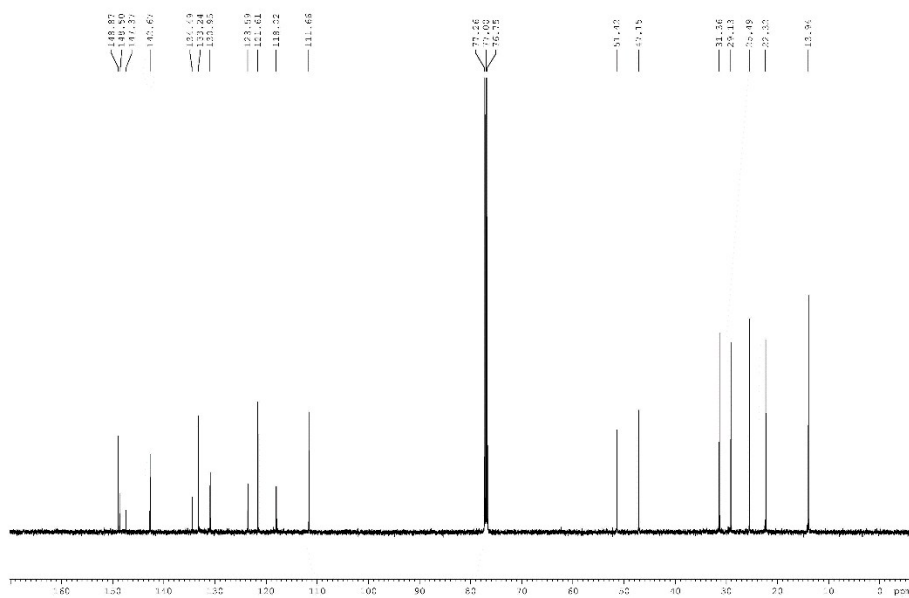


Figure S18. ^{13}C spectrum of F1 in CDCl_3

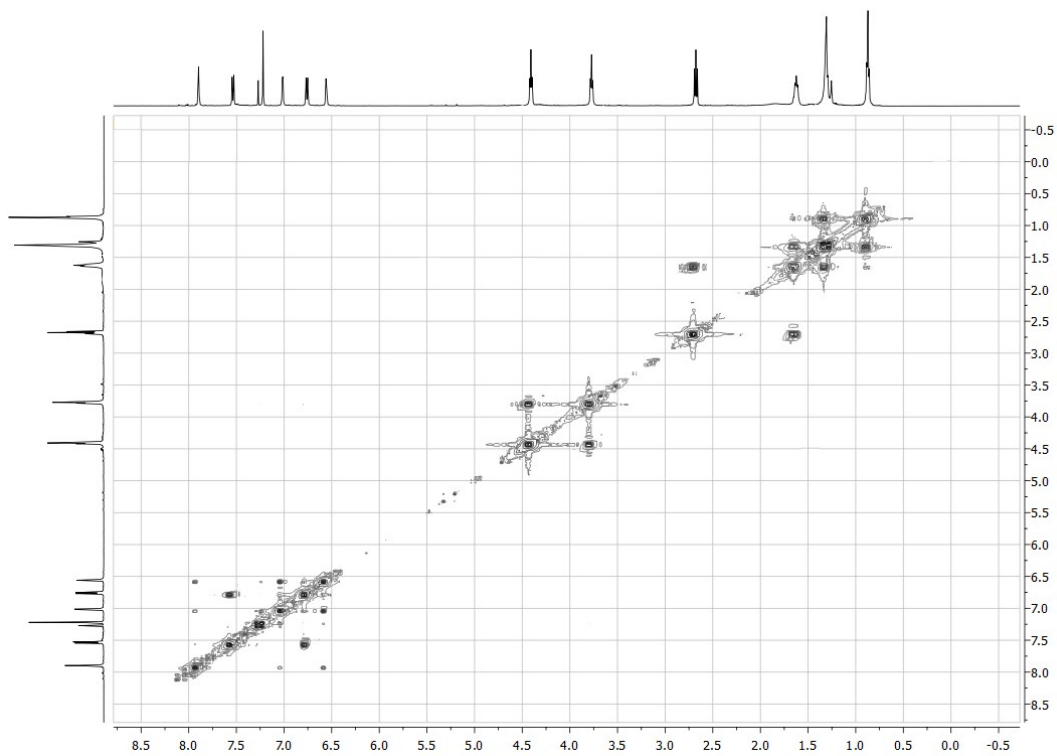


Figure S19. ^1H COSY spectrum of F1 in CDCl_3

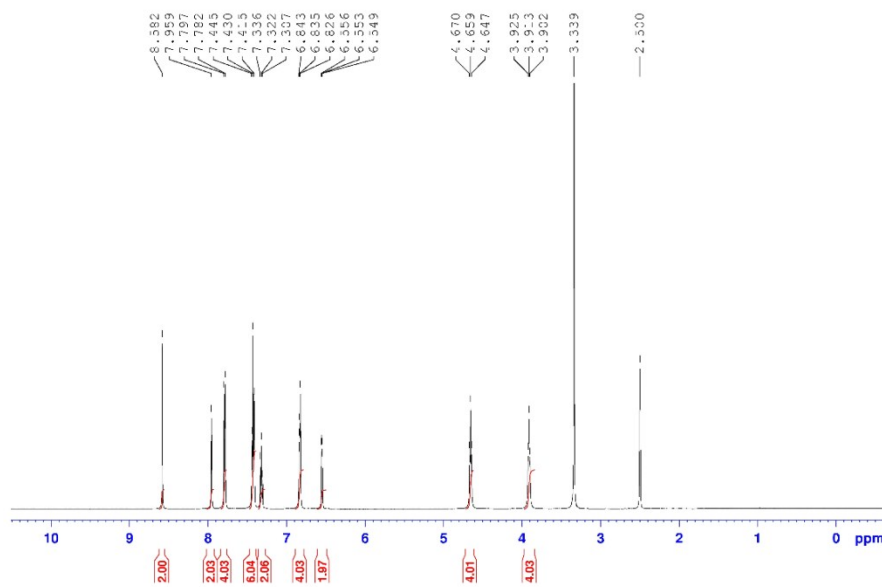


Figure S20. ^1H spectrum of F2 in DMSO-d_6

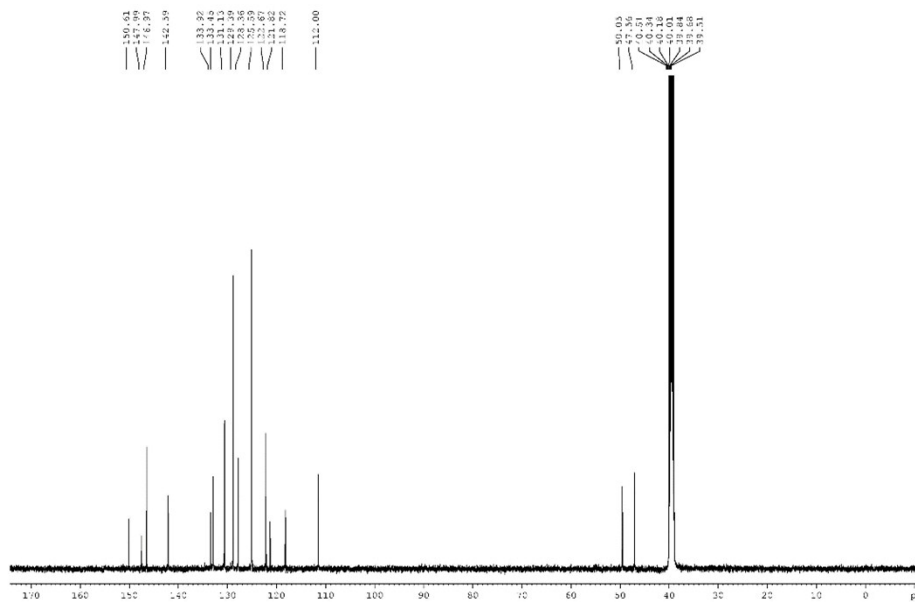


Figure S21. ^{13}C spectrum of **F2** in DMSO-d_6

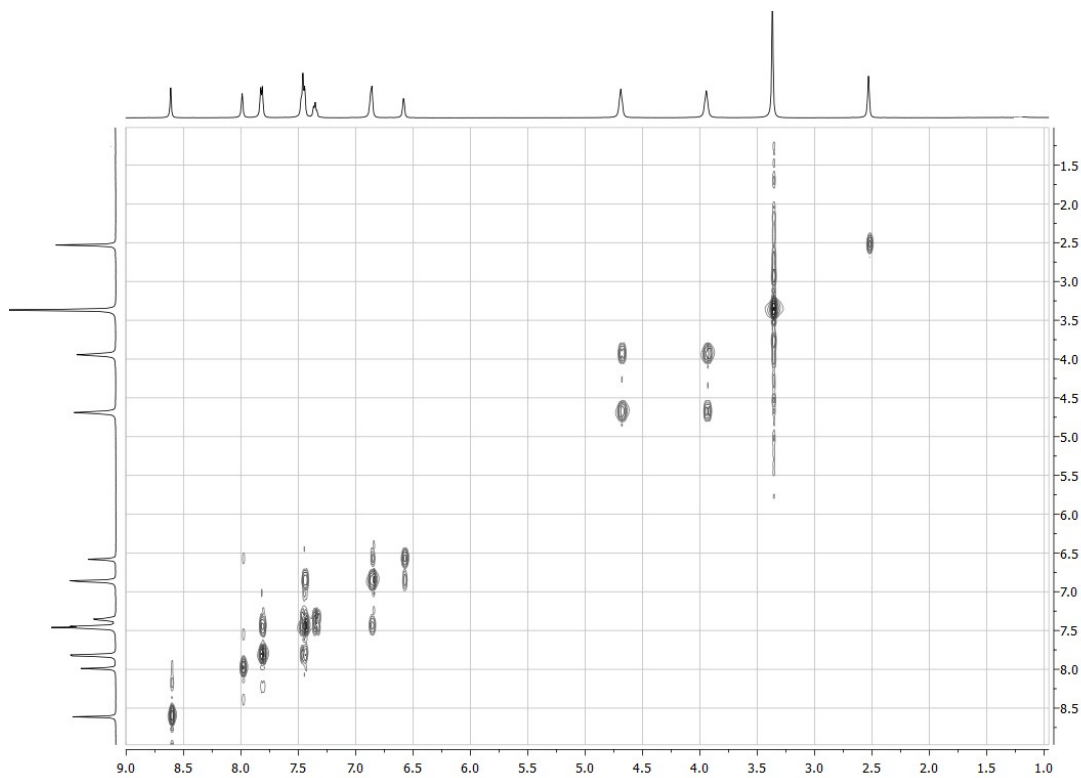


Figure S22. ^1H COSY spectrum of **F2** in DMSO-d_6