Electronic Supplementary Material (ESI) for RSC Advances.

2,1-Benzothiazine - (Quinolin/Thiophen)yl Hydrazone Frameworks as New Monoamine oxidase Inhibitory Agents; Synthesis, in *vitro* and in *silico* Investigation

Noman Javid[a,b †], Saquib Jalil[c †], Rubina Munir[d]*, Muhammad Zia-ur-Rehman[e], Amna Sahar[d], Sara Arshad[d], Jamshed Iqbal[c]*

[a] *School of Chemistry, University of the Punjab, Lahore-54590, Pakistan*

[b] *Chemistry Department (C-Block), Forman Christian College, Ferozepur Road Lahore, Pakistan*

[c] *Centre for Advanced Drug Research, COMSATS University Islamabad, Abbottabad Campus, Abbottabad-22060, Pakistan*

[d] *Department of Chemistry, Kinnaird College for Women, Lahore 54000, Pakistan*

[e] *Applied Chemistry Research Centre, PCSIR Laboratories Complex, Lahore-54600, Pakistan*

***Corresponding Authors:**

- e-mail: organist94@gmail.com; rubina.munir@kinnaird.edu.pk (R. Munir)
- Tel.: +92-992-383591-96; fax: +92-992-383441; e-mail: drjamshed@cuiatd.edu.pk (J. Iqbal)

† The authors have equal contribution

Synthesis of methyl 2-(methylsulfonamido)benzoate (2)

A neat mixture of methyl anthranilate **1** (100 mmol; 15.12 g, 13.2 mL) and triethylamine (100 mmol; 10.12 g, 13.9 mL) was added dropwise to methanesulfonyl chloride (100 mmol; 11.44 g, 8.0 mL) maintained at 0°C with continuous stirring. After addition, the reaction mixture was stirred continuously at room temperature under moisture free conditions for 45 minutes. Pale yellow crude product was isolated by washing the resulting mass with cold water which was subsequently recrystallized from ethanol to yield pure white crystals that were dried and weighed. Yield: 98%; mp 89-90 °C (Lit mp 88-90 °C).[1]

Synthesis of methyl 2-(*N*-methylmethylsulfonamido)benzoate (3)

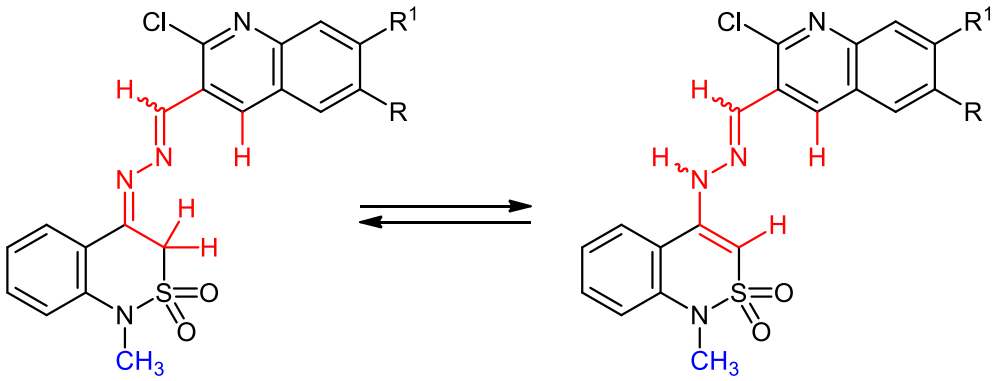
A solution of the reactant **2** (100 mmol; 22.9 g) in 25 mL dry DMF was allowed to cool in a round bottom flask under moisture free conditions. NaH (150 mmol) was added in the ice cold solution in six portions with continuous stirring. Afterwards methyl iodide (120 mmol; 17.04 g, 7.5mL) was introduced in the reaction mixture drop wise and the solution was stirred overnight. Upon completion of the reaction, the reaction mixture was poured into crushed ice and pH of the resulting solution was maintained at 4 by the addition of 5N HCl solution, following the stirring for 30 minutes. Vacuum filtration of the resulting precipitates followed by washing with distilled water gave the product **3**. The filtrate was extracted with chloroform and the chloroform layer was evaporated to obtain more yield of **3**.[2] Yield: 68%; mp 55 °C (Lit mp 55 °C).[2]

Synthesis of 1-methyl-1*H*-benzo[*c*][1,2]thiazin-4(3*H*)-one 2,2-dioxide (4)

30 mL dry DMF and **3** (50 mmol; 12.15 g) were added to a round bottom flask. **3** was dissolved and the solution was cooled to 0 – 5 ºC in freezing mixture. NaH (100 mmol) was added to the above solution in portions over a period of 1.5 hours under moisture free condition. The reaction mixture was left on stirring at room temperature. After completion of the reaction (as indicated by TLC), the mixture was plunged into ice cold and its pH was adjusted at 5 by adding 5N HCl solution. White precipitates were formed which were filtered, washed with water and air dried to obtain pure product.[2] Yield: 77%.

Tautomers and Stereoisomers

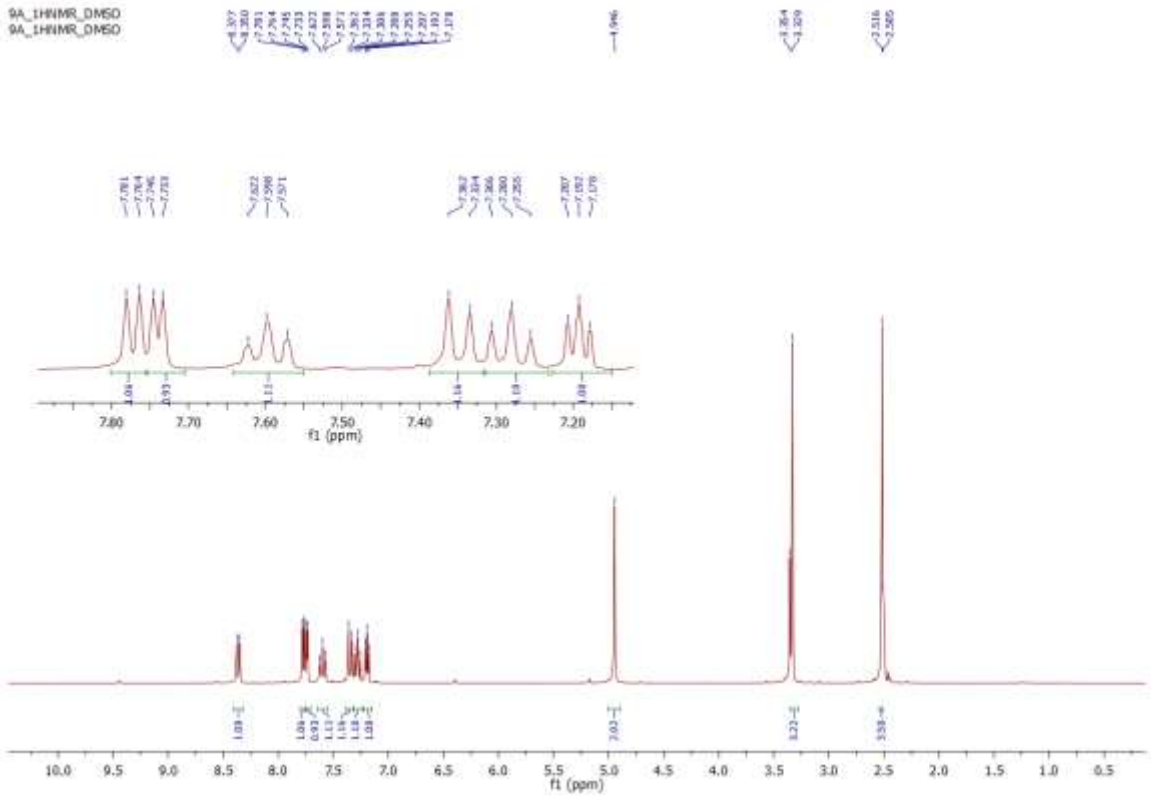
The highlighted skeleton in the following figure shows duplication of peaks for hydrogen and carbon atoms in ^{1}H NMR and ^{13}C NMR spectra due to different environment of nuclei.

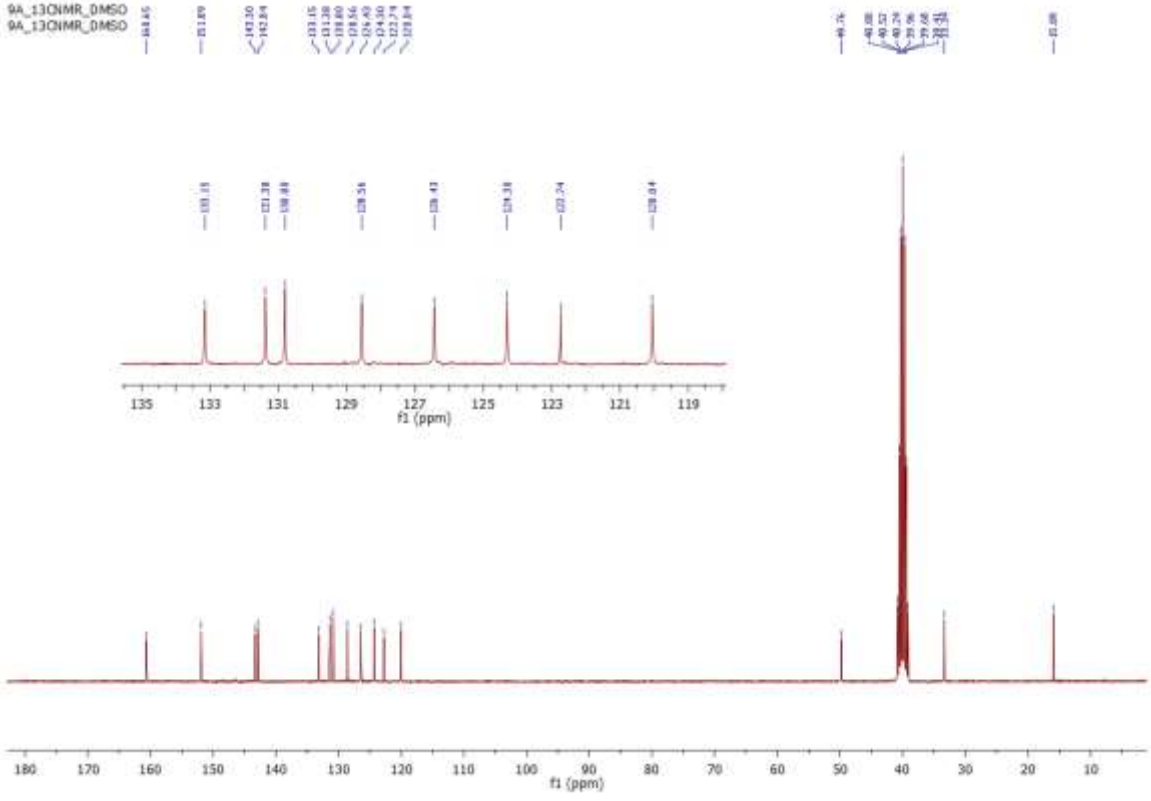


Imine Form **Enamine Form**

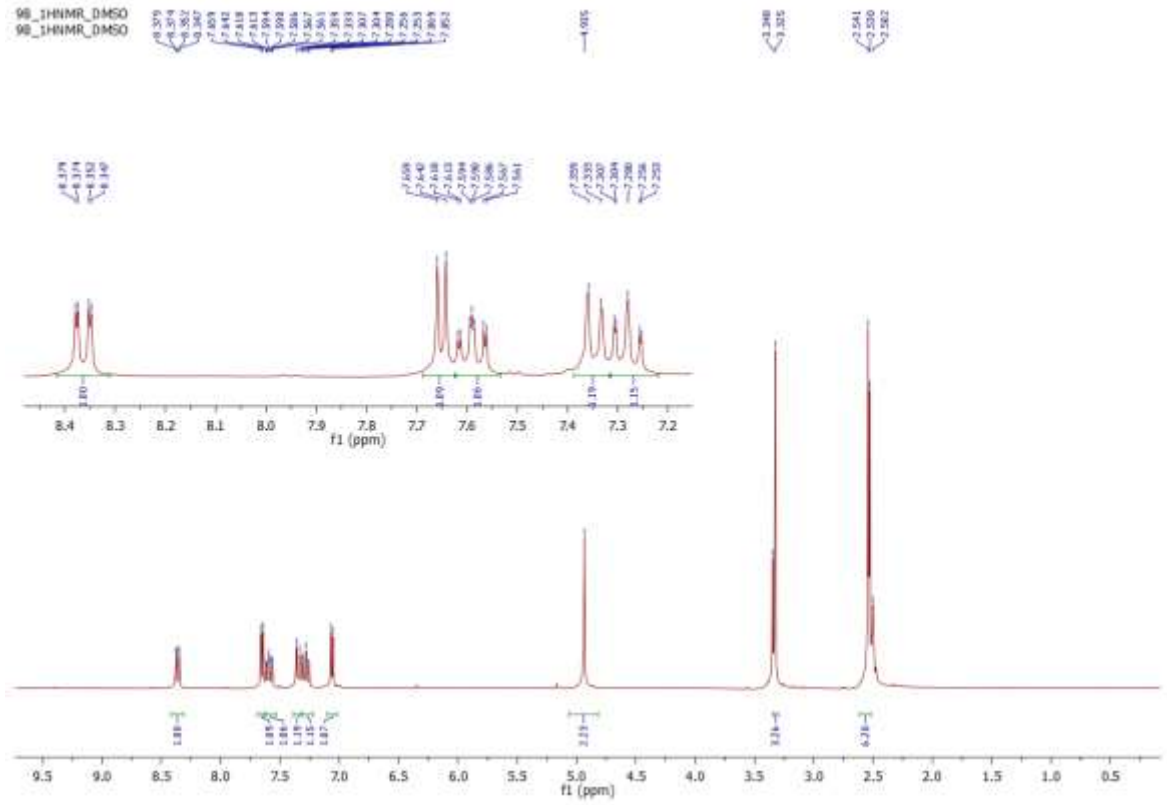
Figure S1: Tautomeric forms of Compounds **7(a-f)**

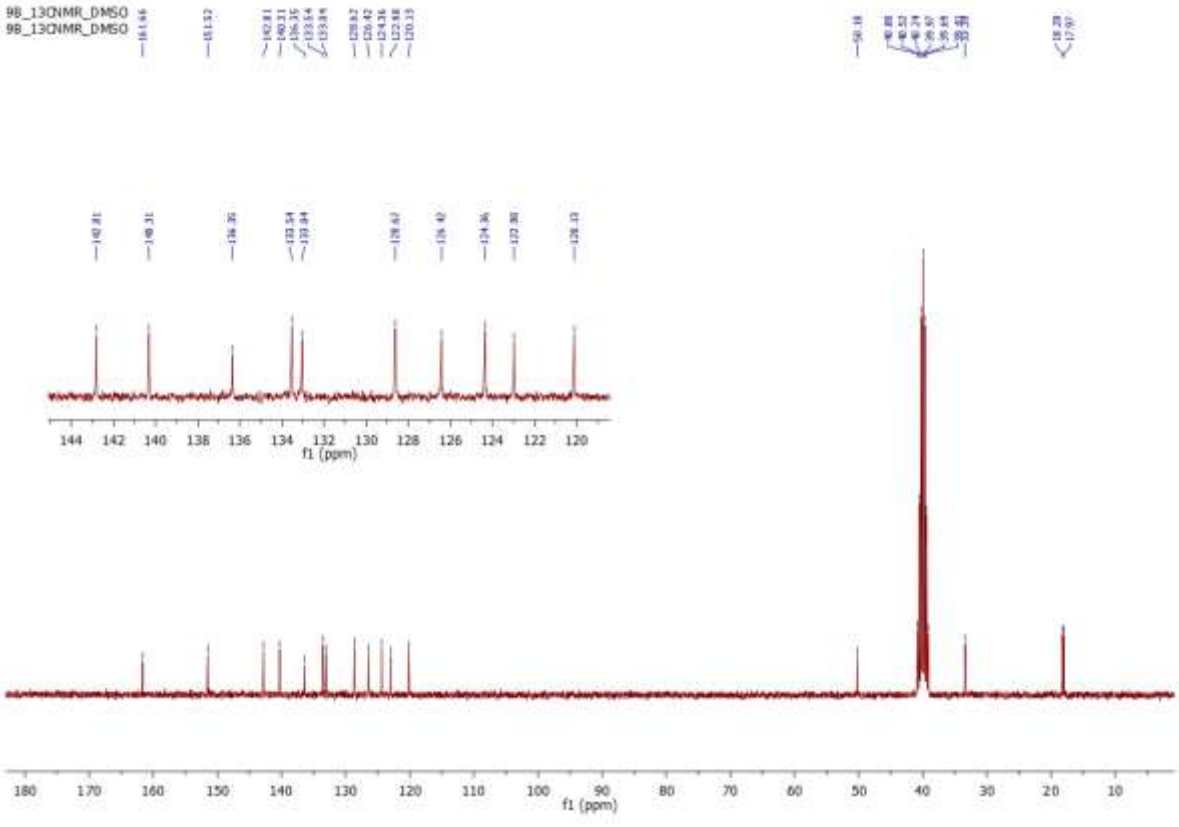
[1]HNMR and [13]C NMR Spectra of 9a



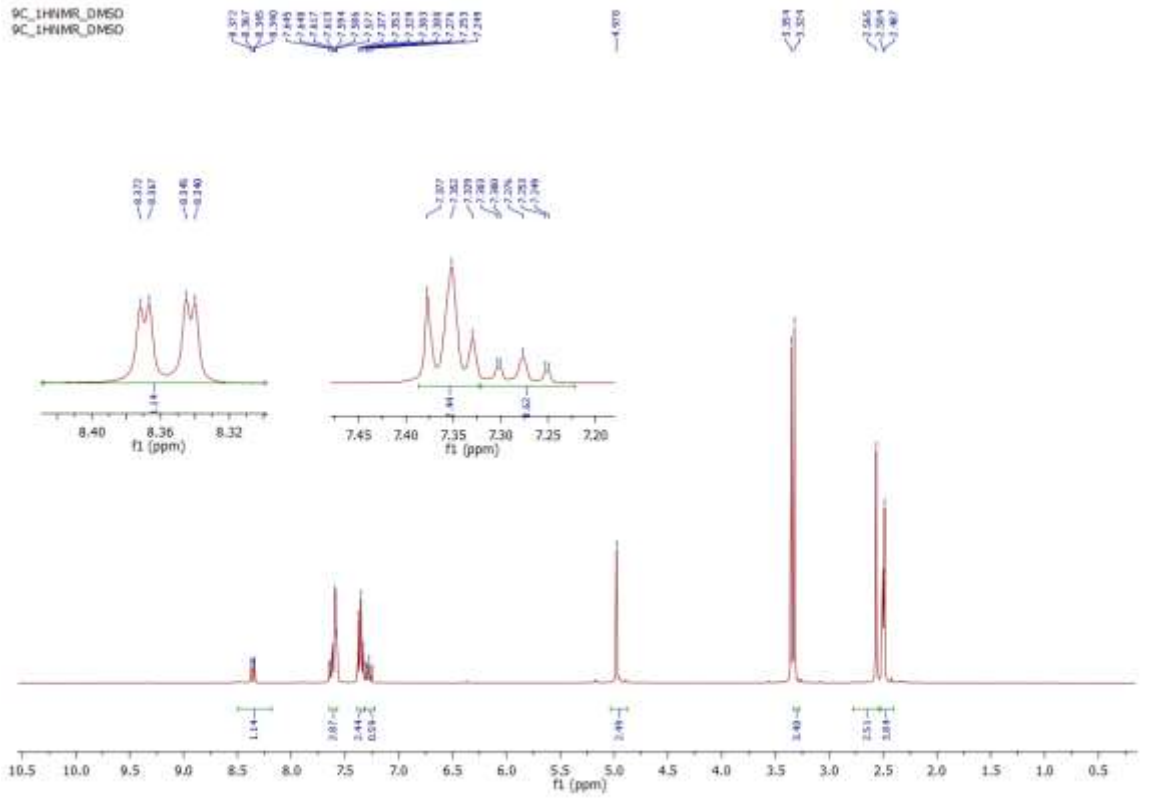


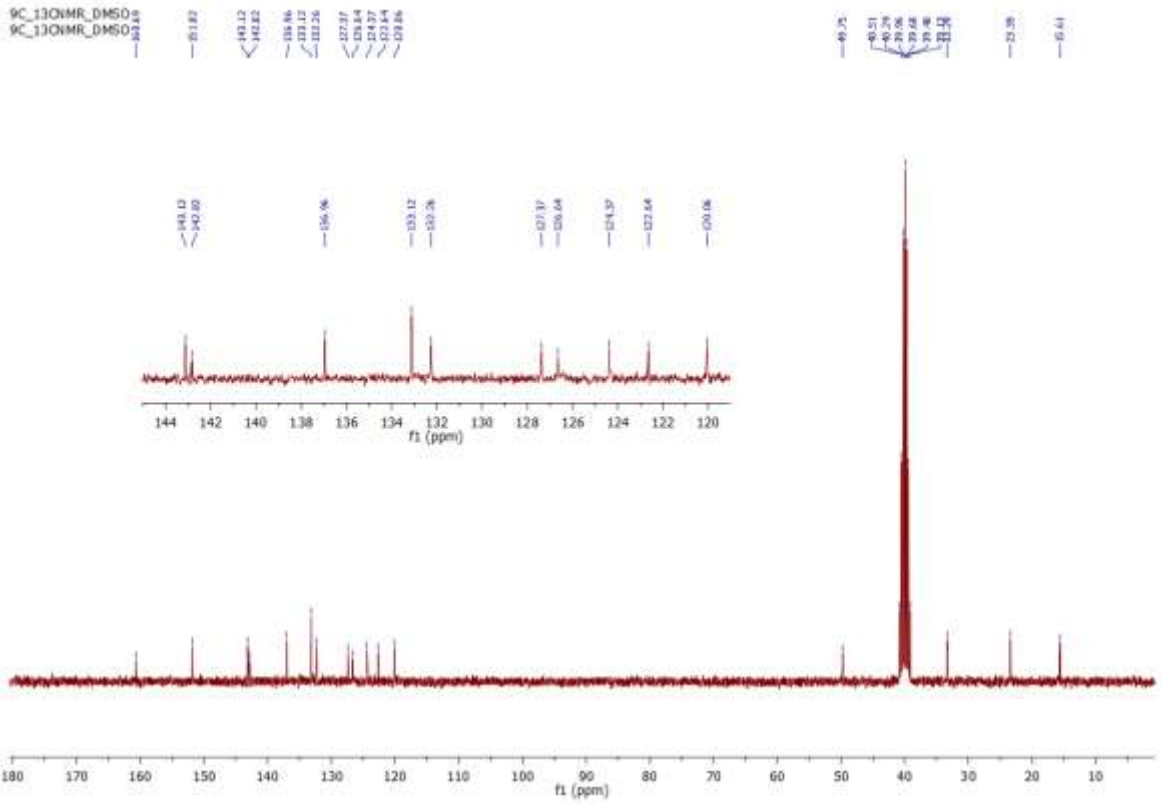
[1]HNMR and [13]C NMR Spectra of 9b



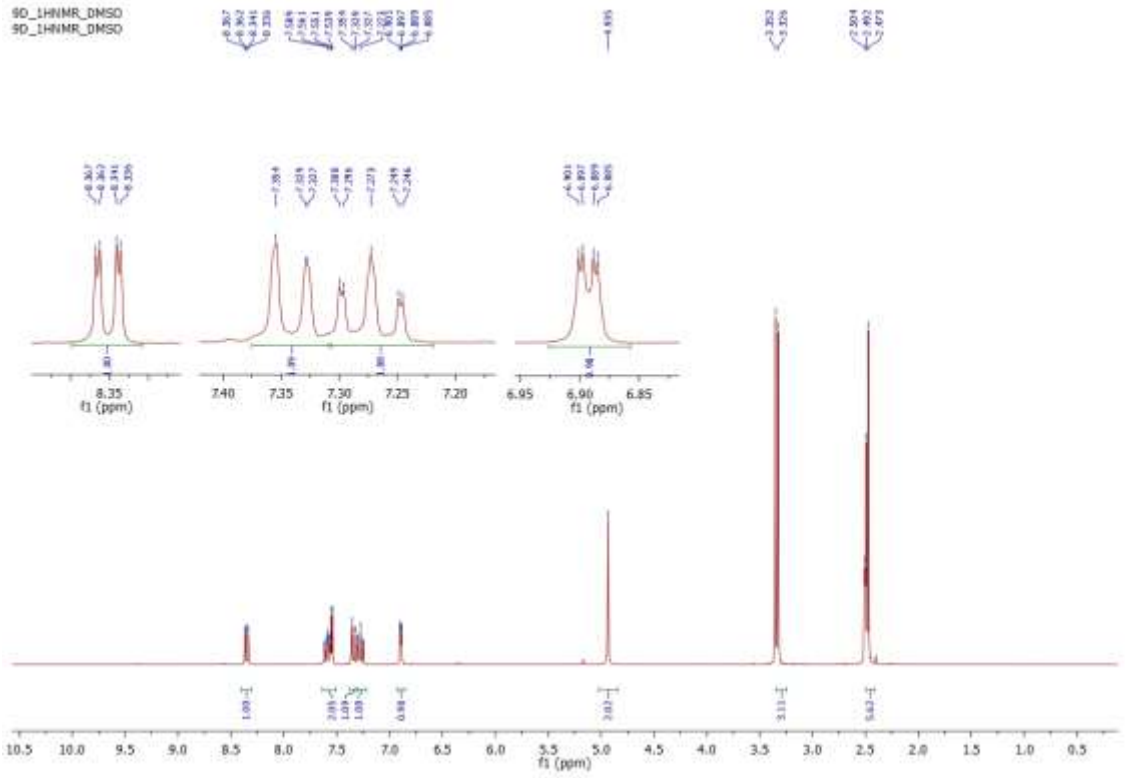


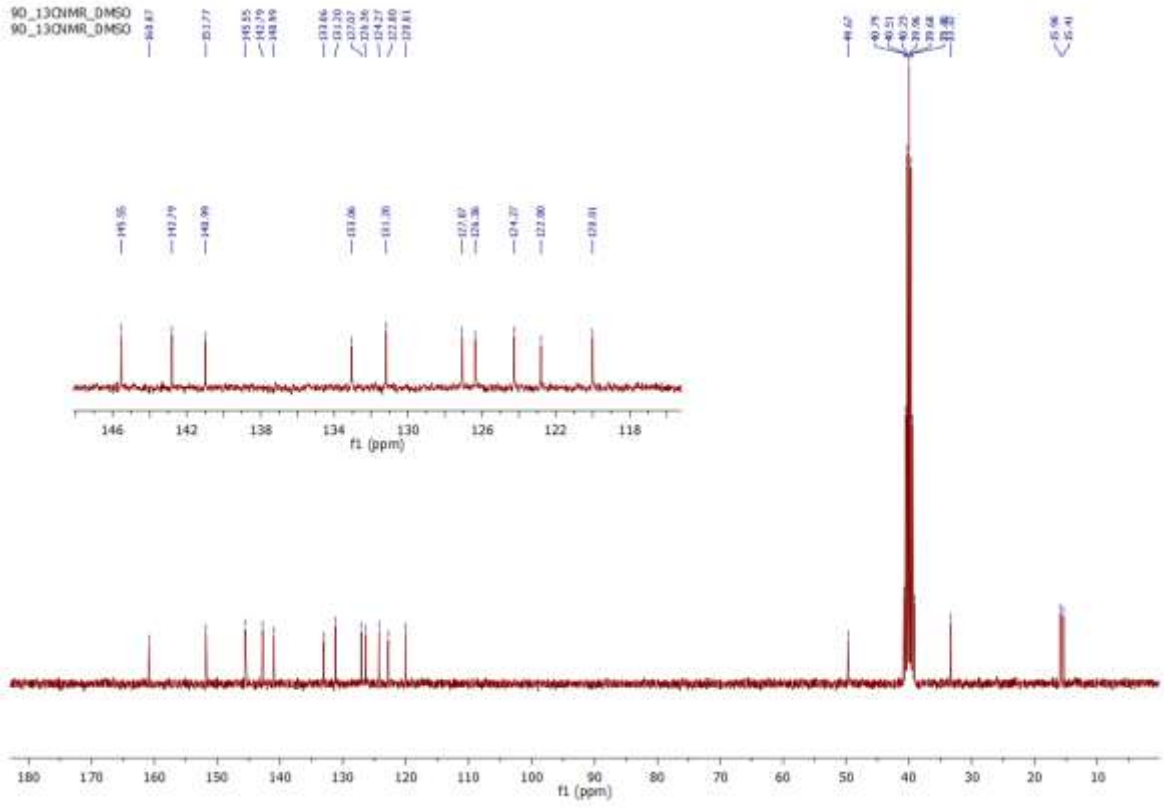
[1]HNMR and [13]C NMR Spectra of 9c



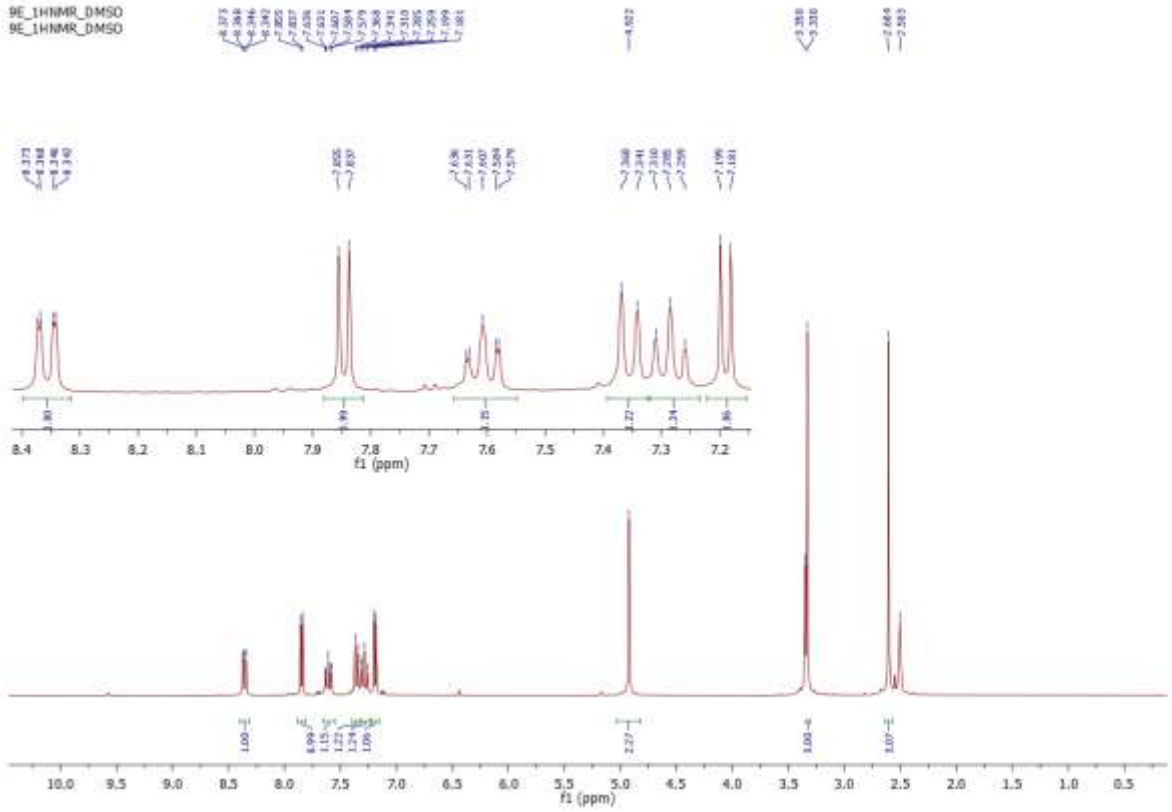


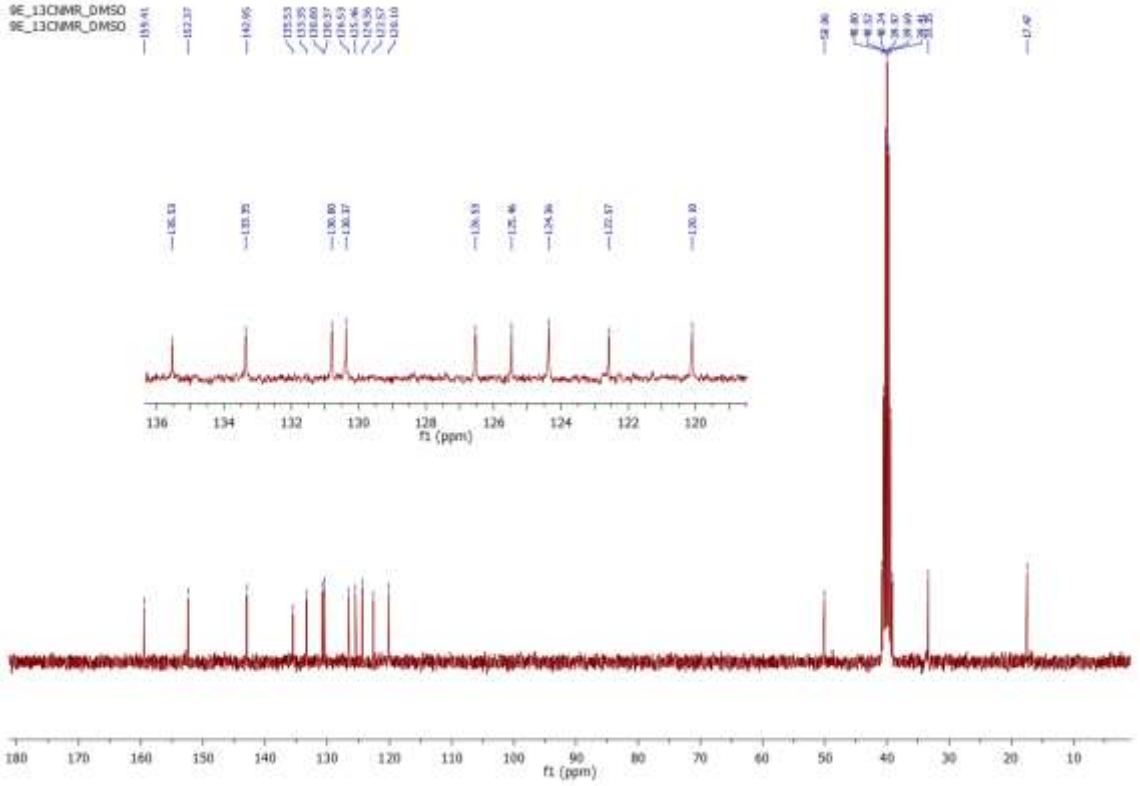
^{1}HNMR and ^{13}C NMR Spectra of 9d



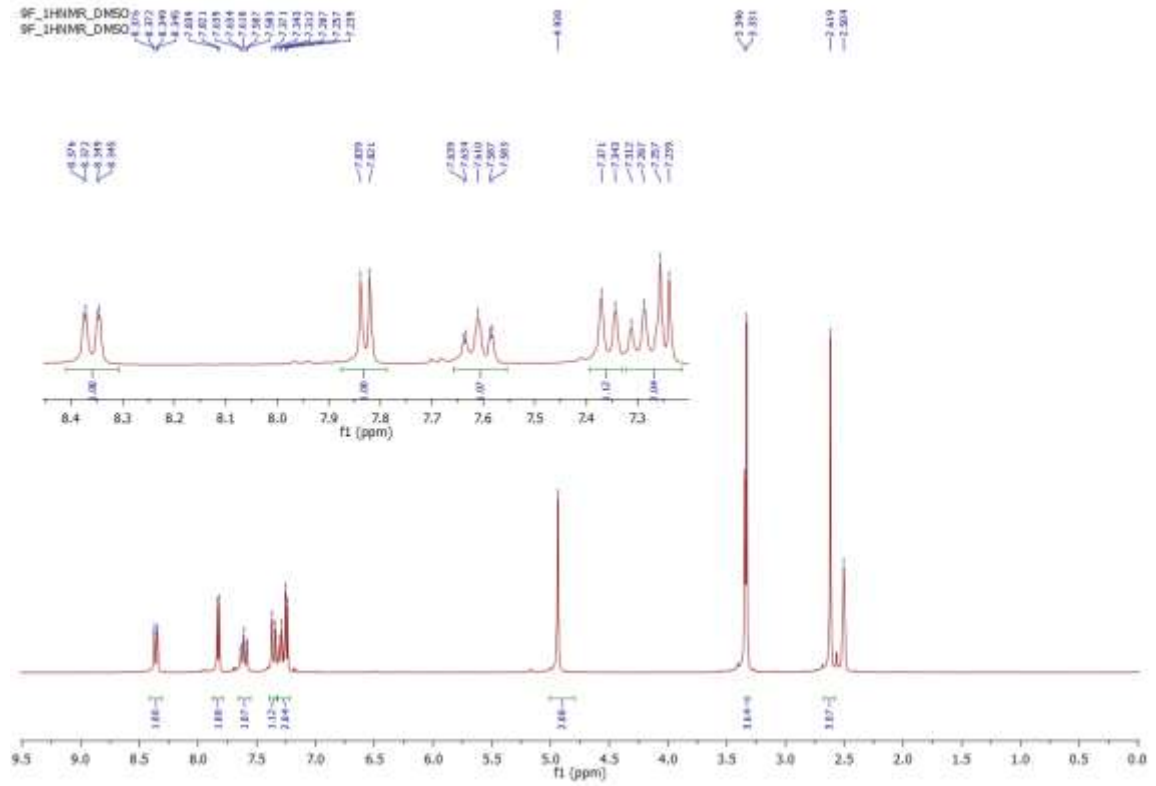


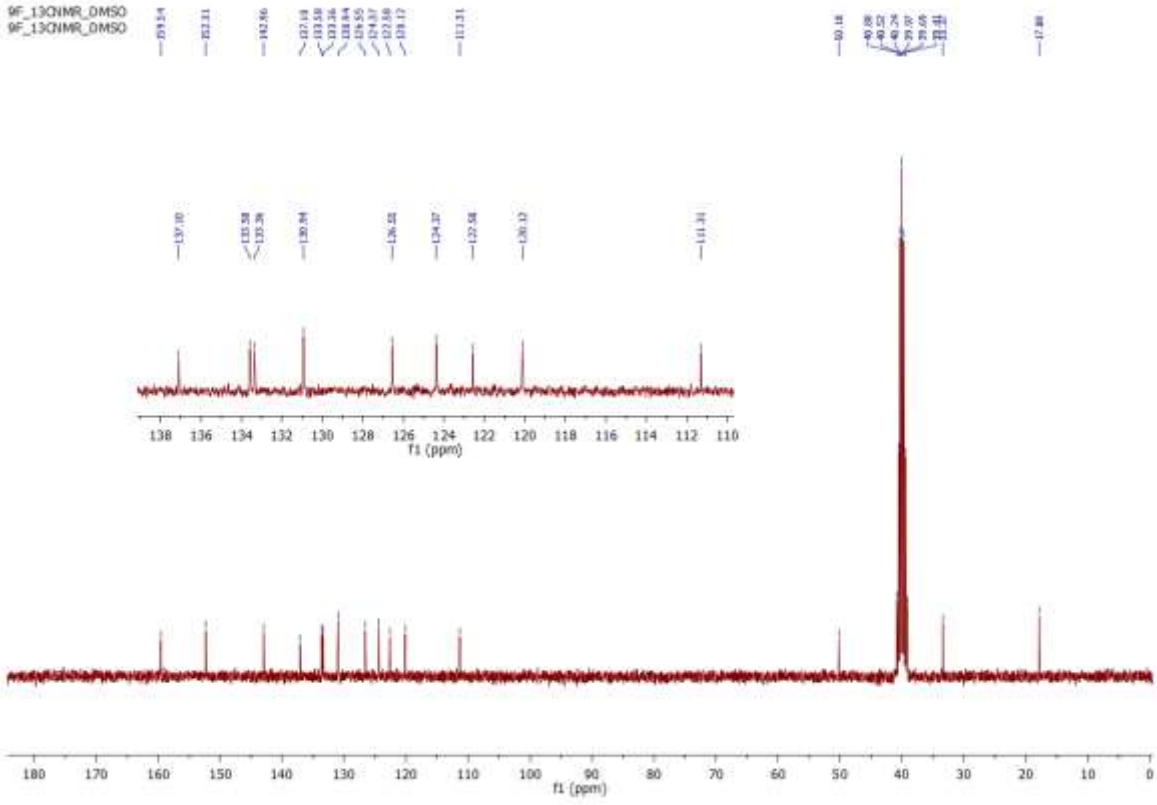
^{1}HNMR and ^{13}C NMR Spectra of 9e



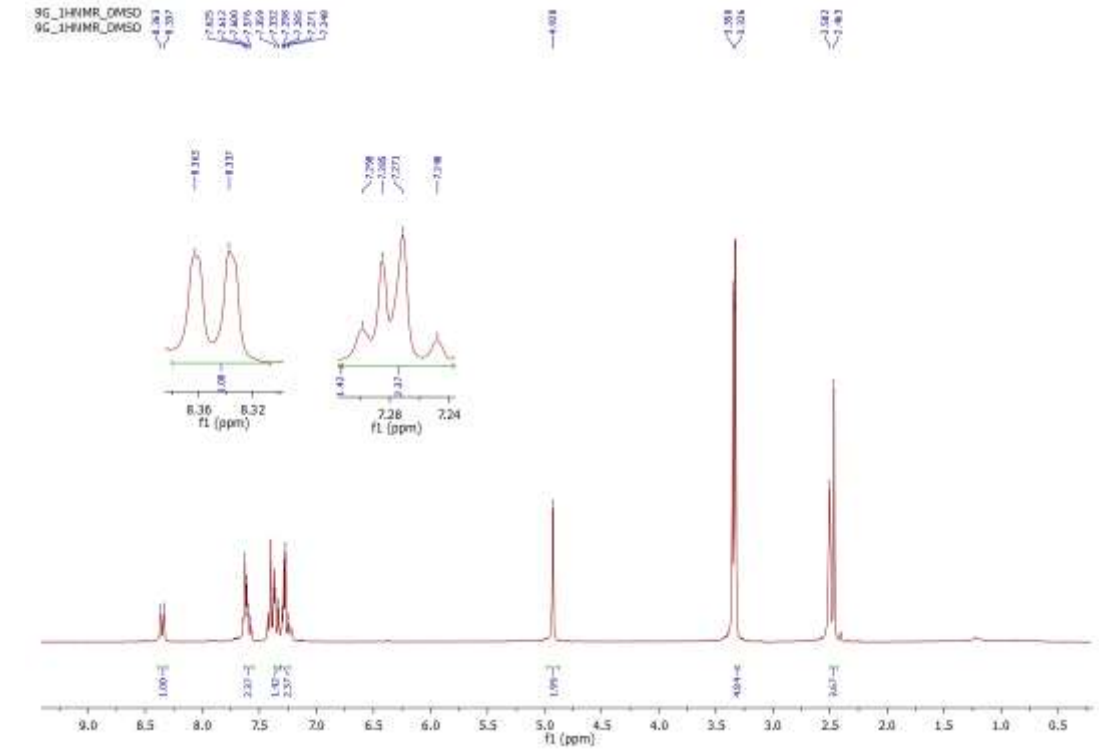


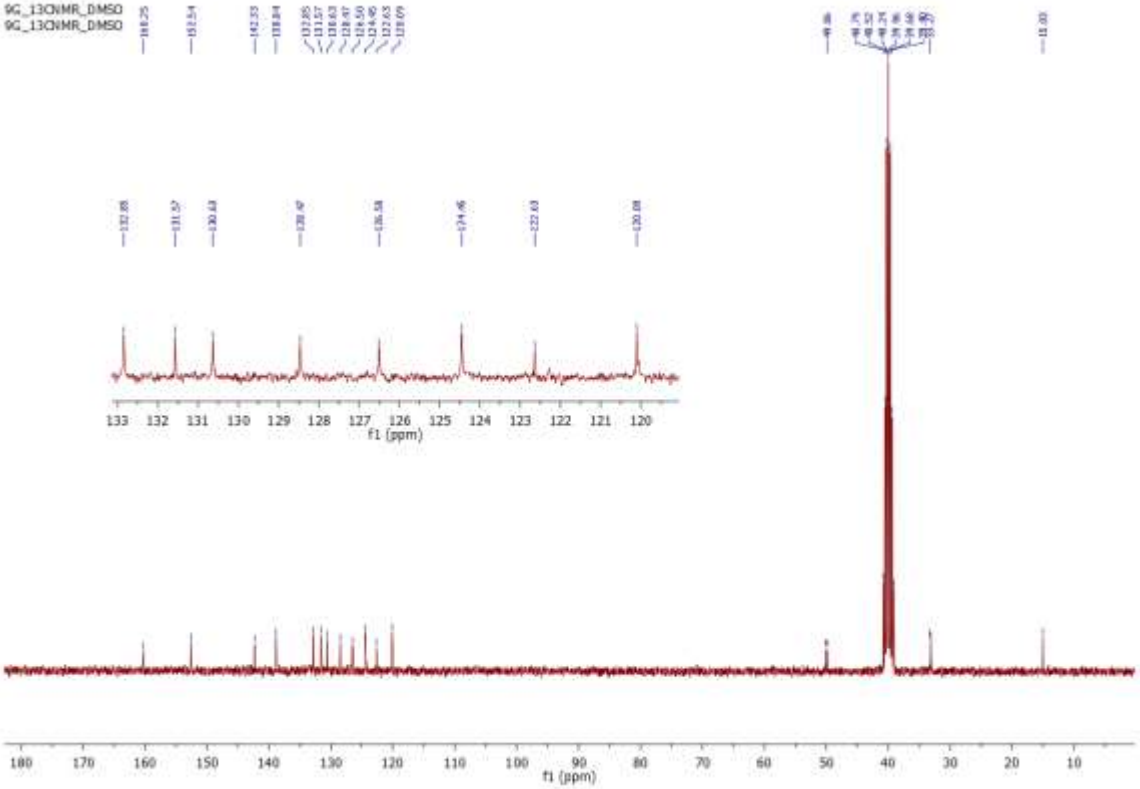
^{1}HNMR and ^{13}C NMR Spectra of 9f



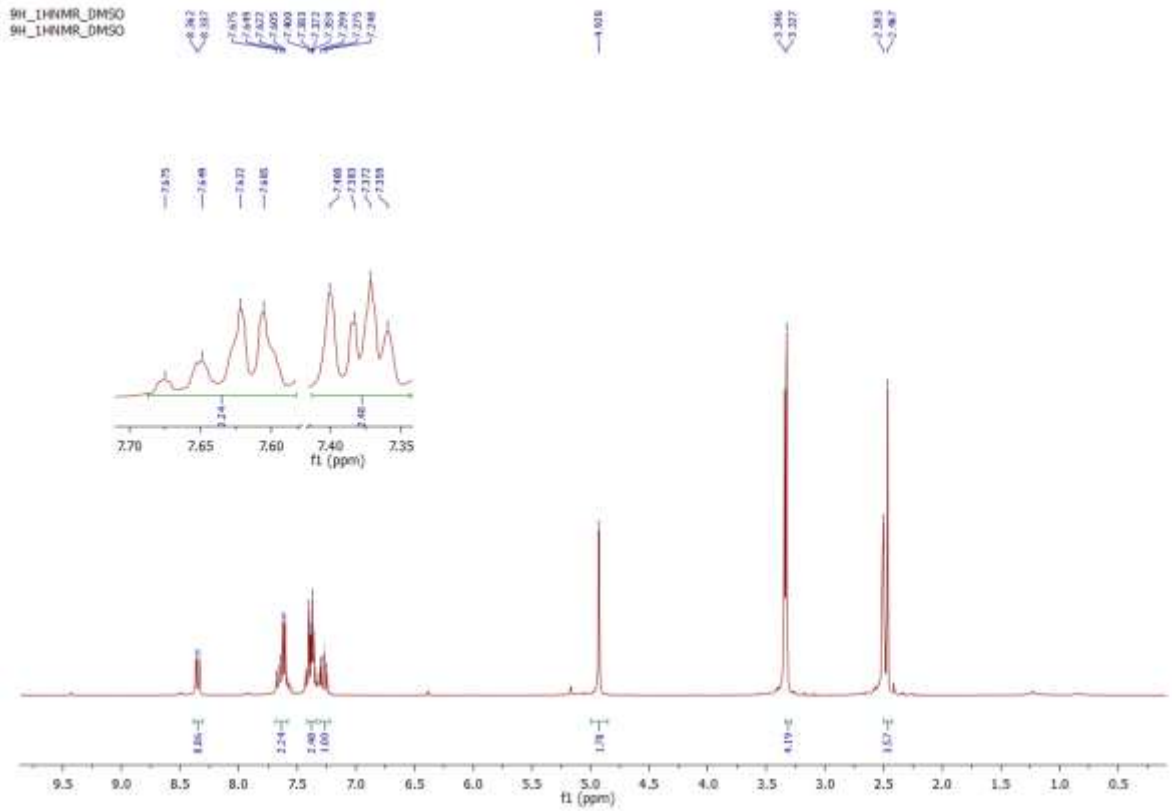


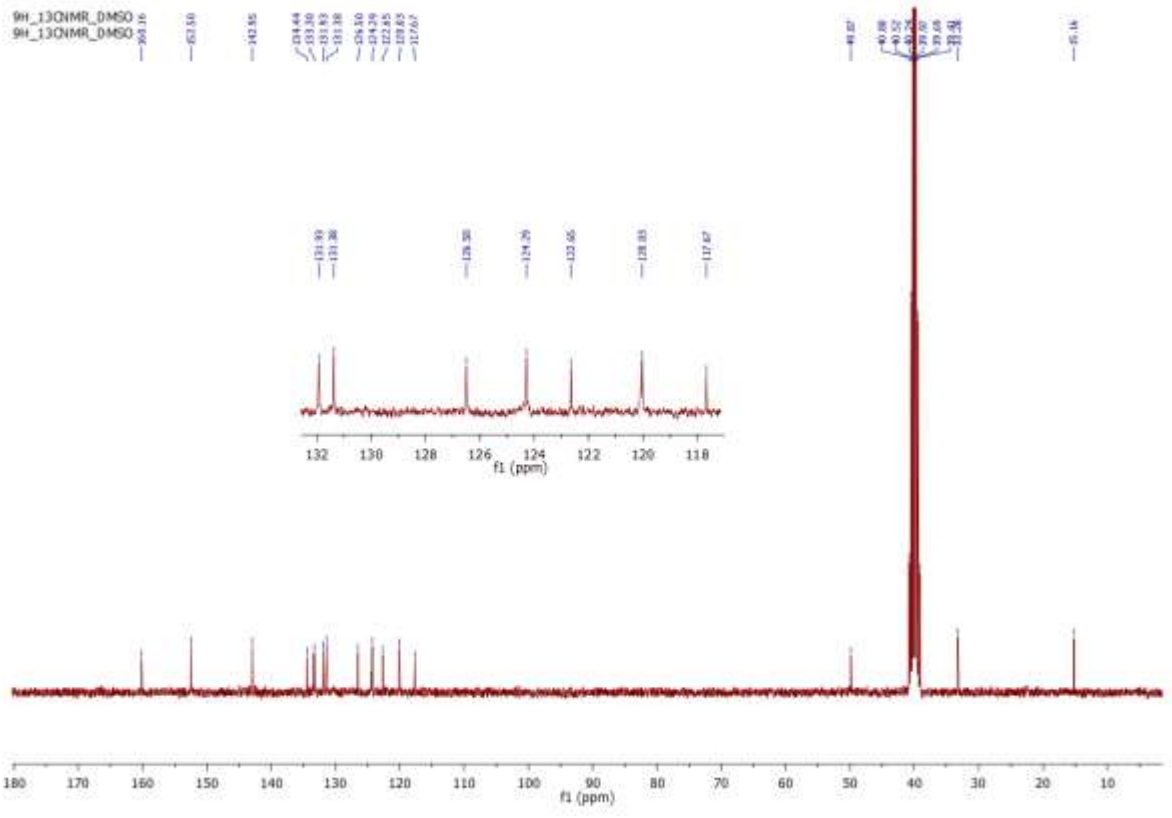
[1]HNMR and [13]C NMR Spectra of 9g



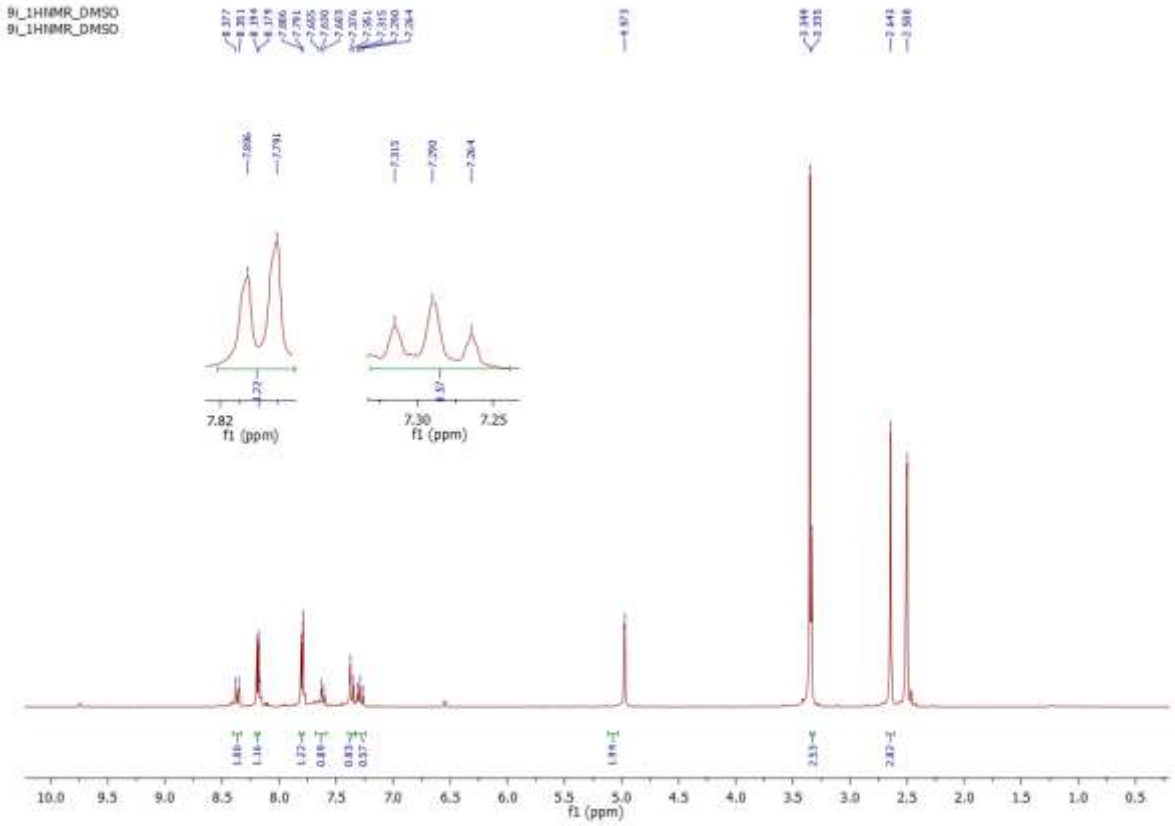


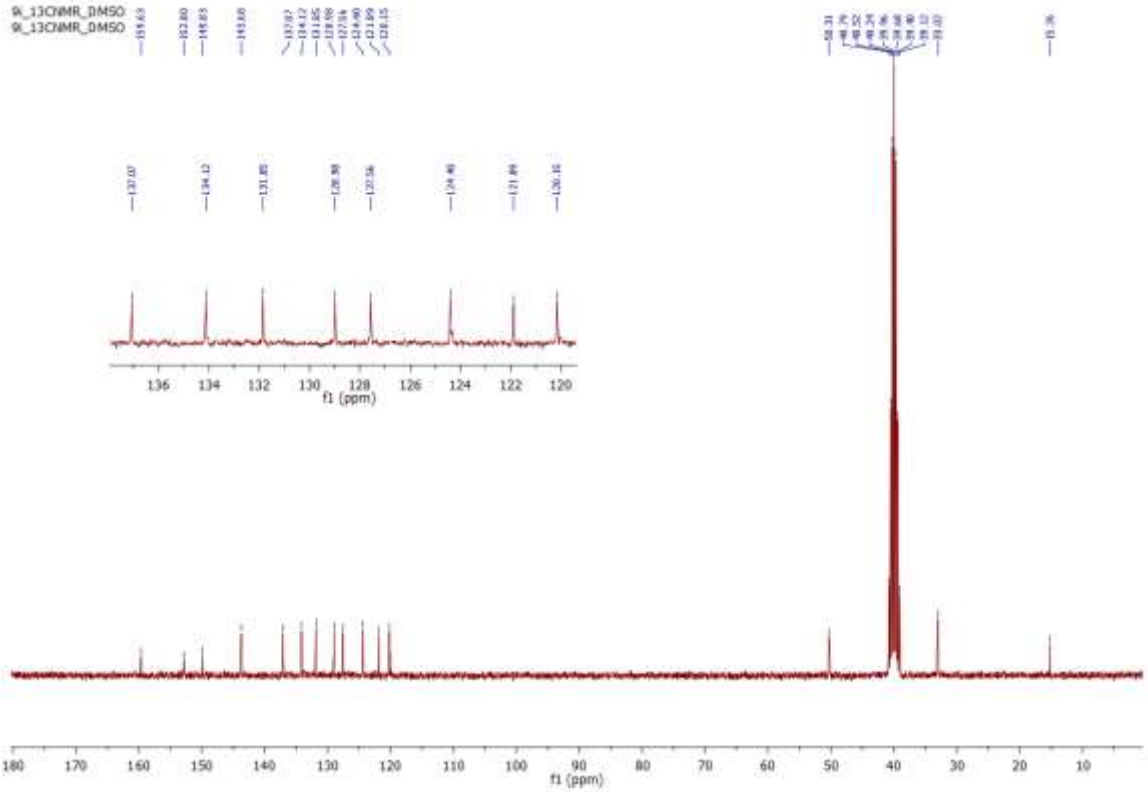
^{1}HNMR and ^{13}C NMR Spectra of 9h



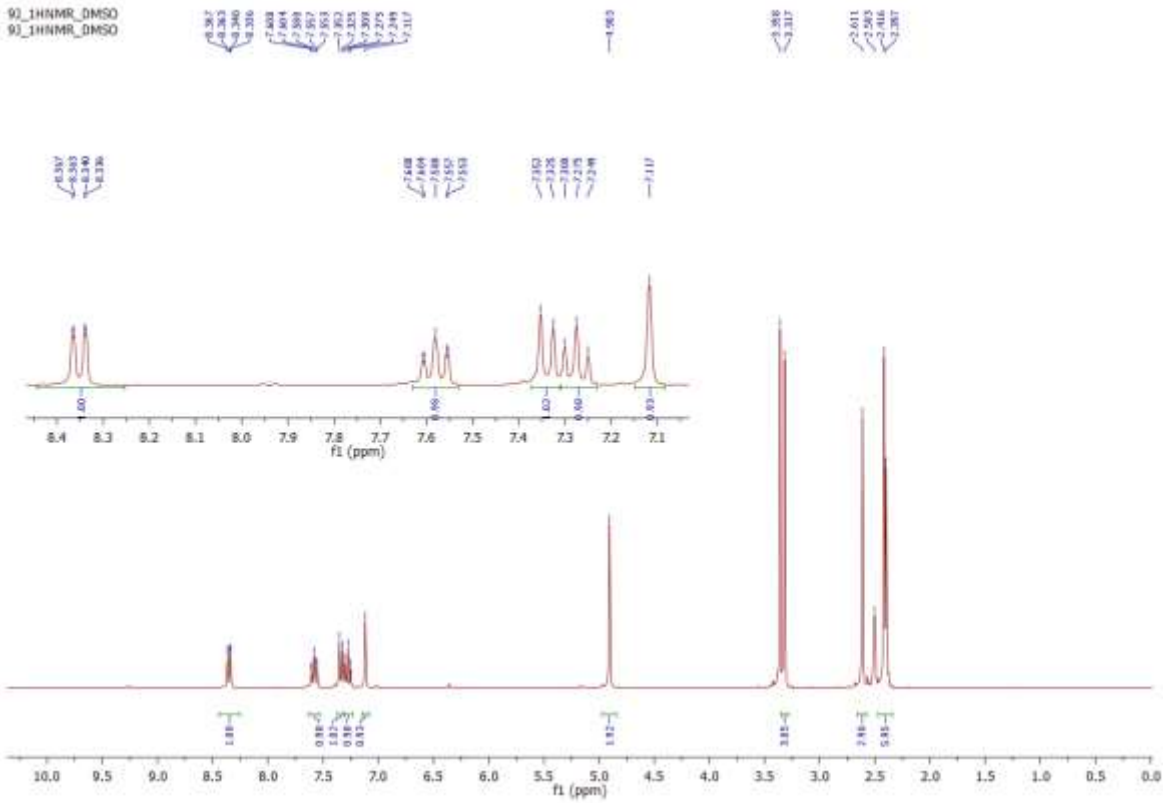


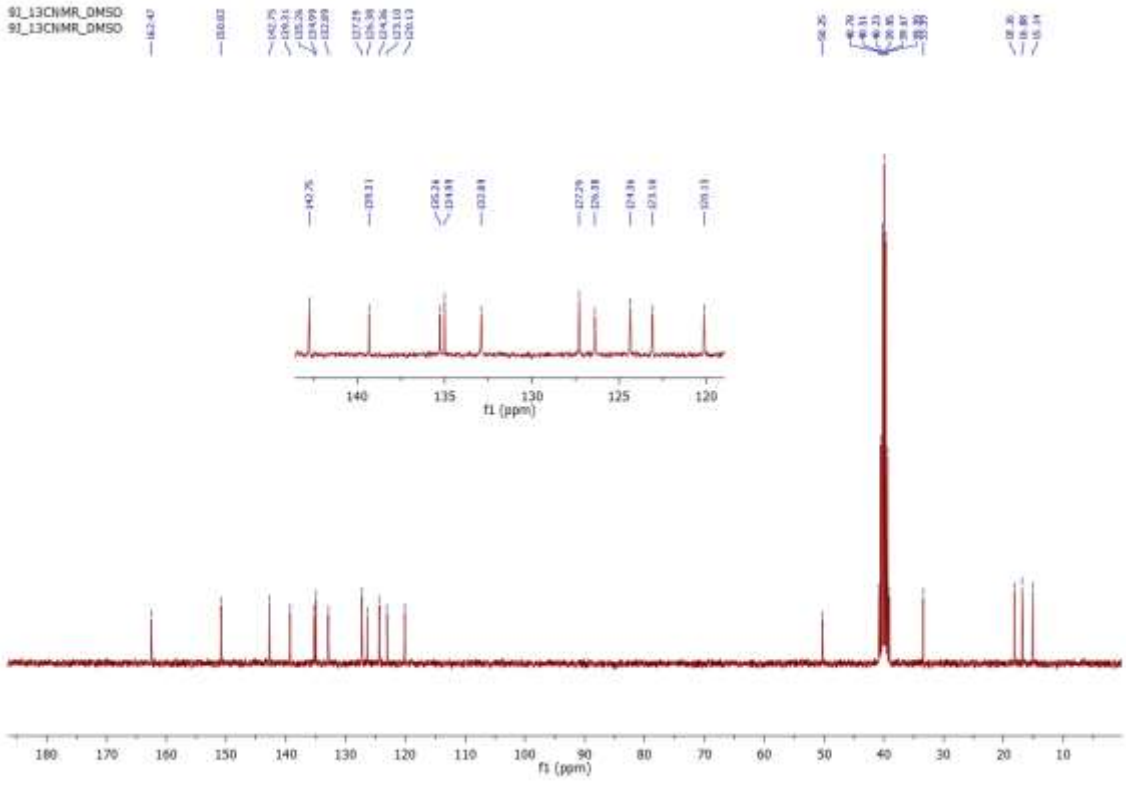
^{1}HNMR and ^{13}C NMR Spectra of 9i



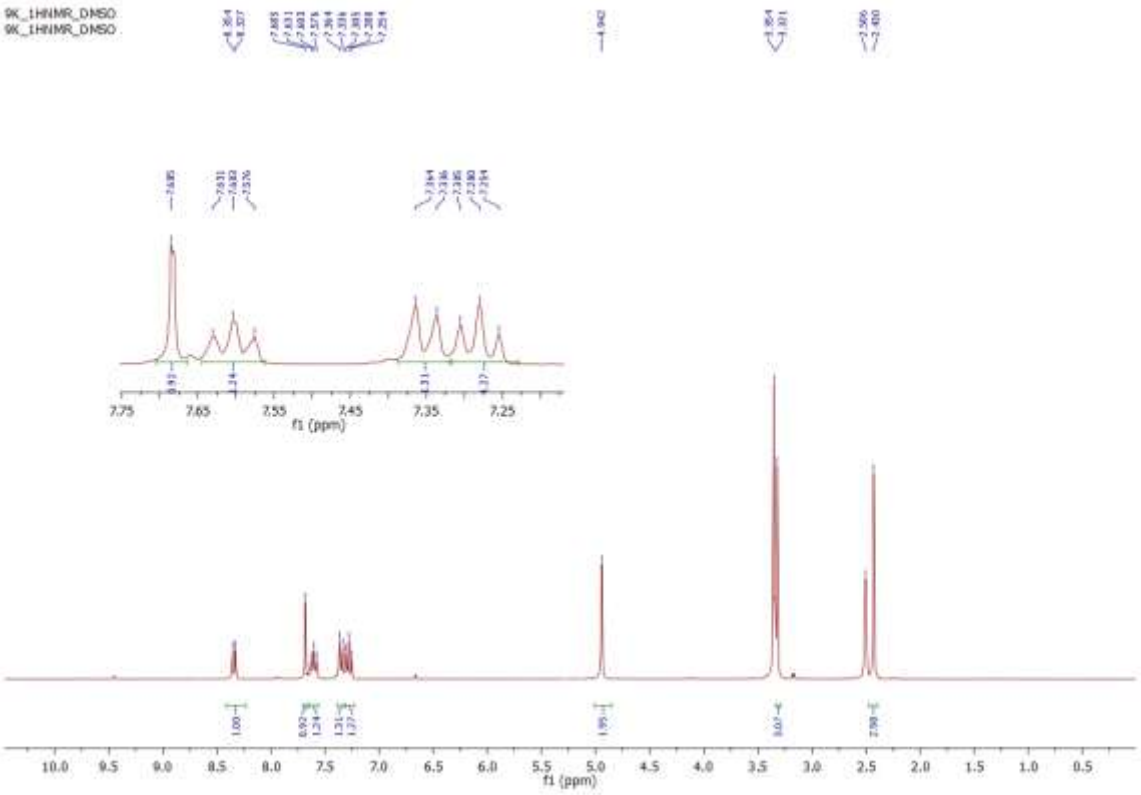


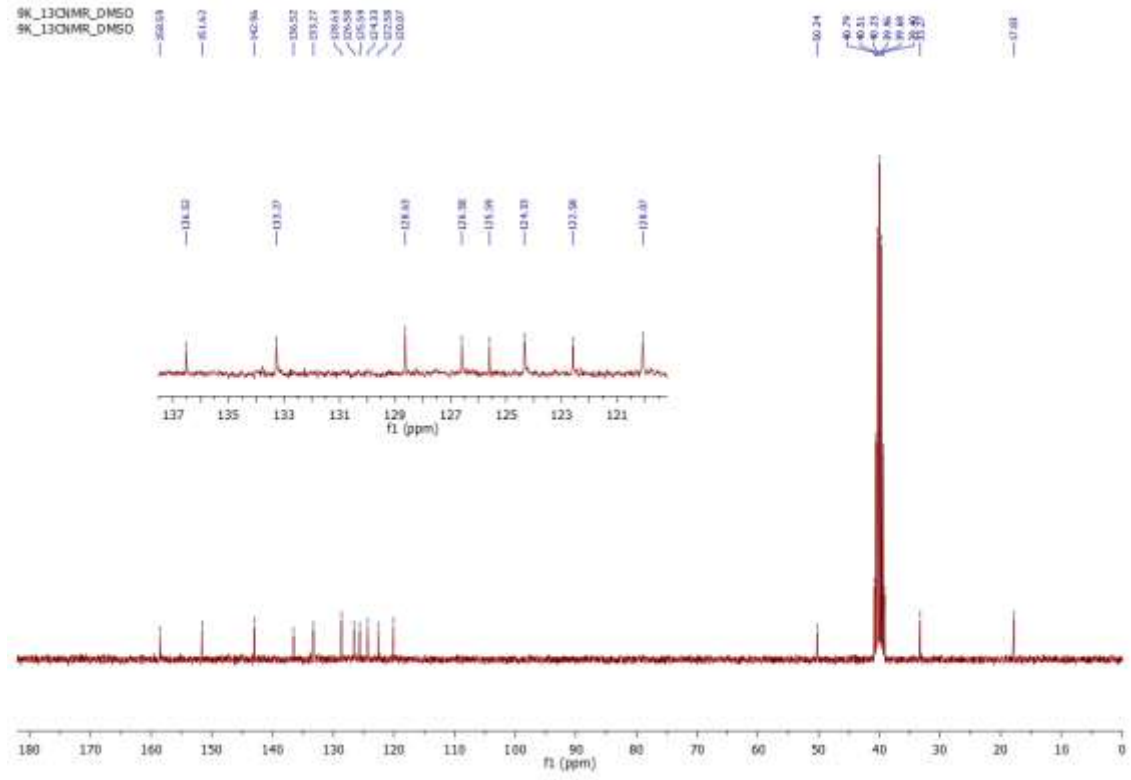
[1]HNMR and [13]C NMR Spectra of 9j



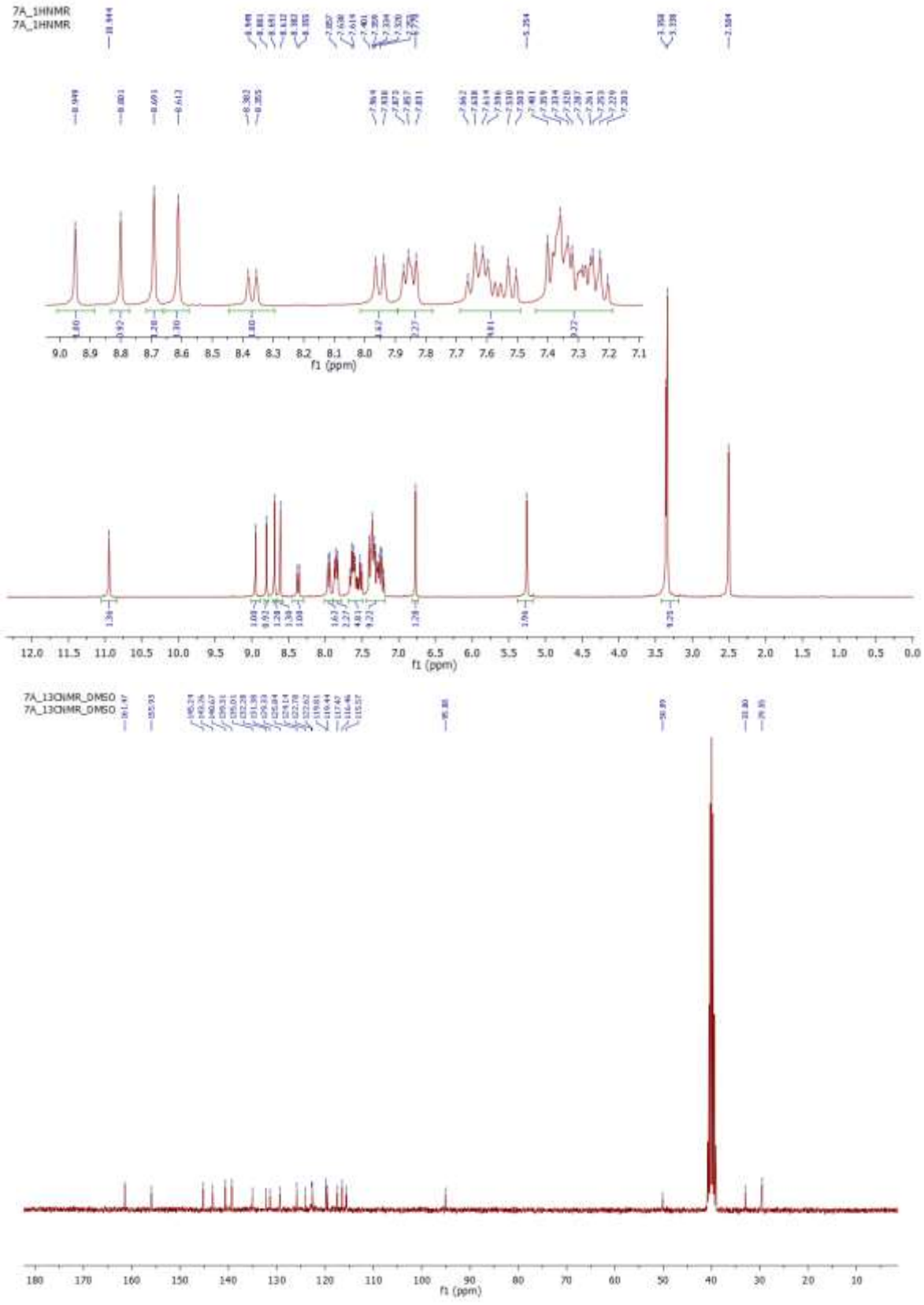


^{1}HNMR and ^{13}C NMR Spectra of 9k

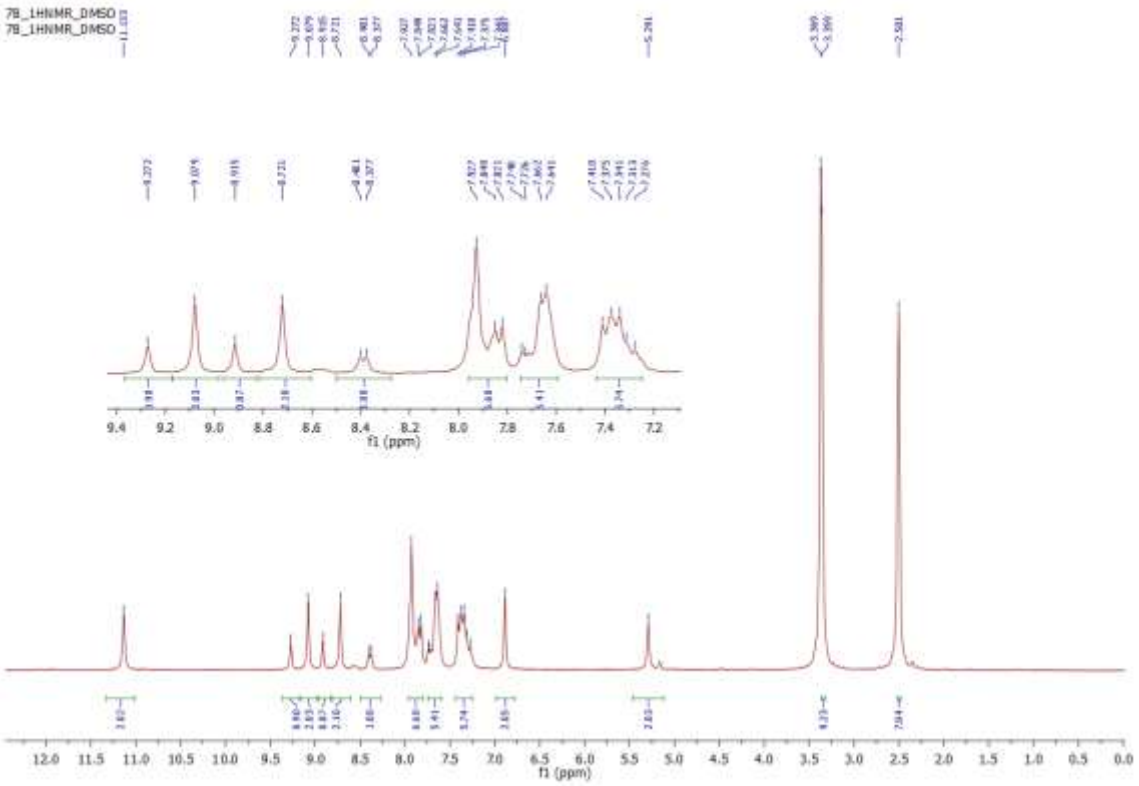


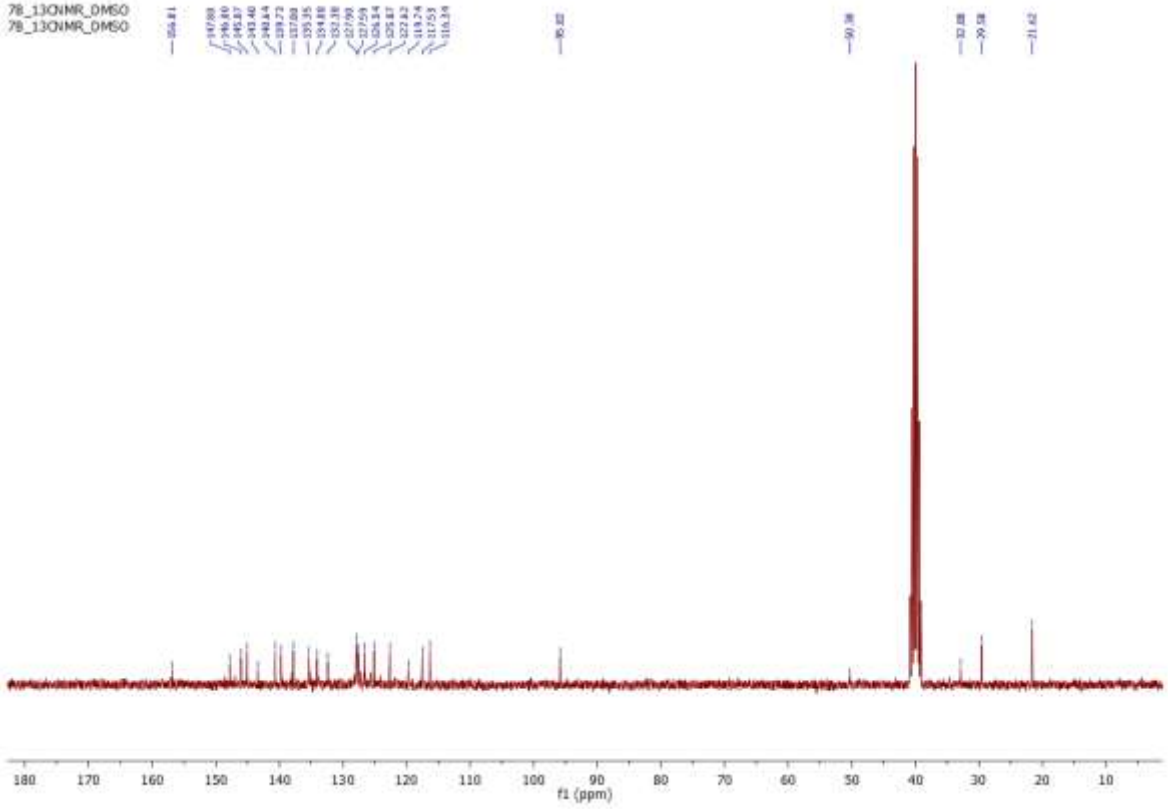


^{1}HNMR and ^{13}C NMR Spectra of 7a

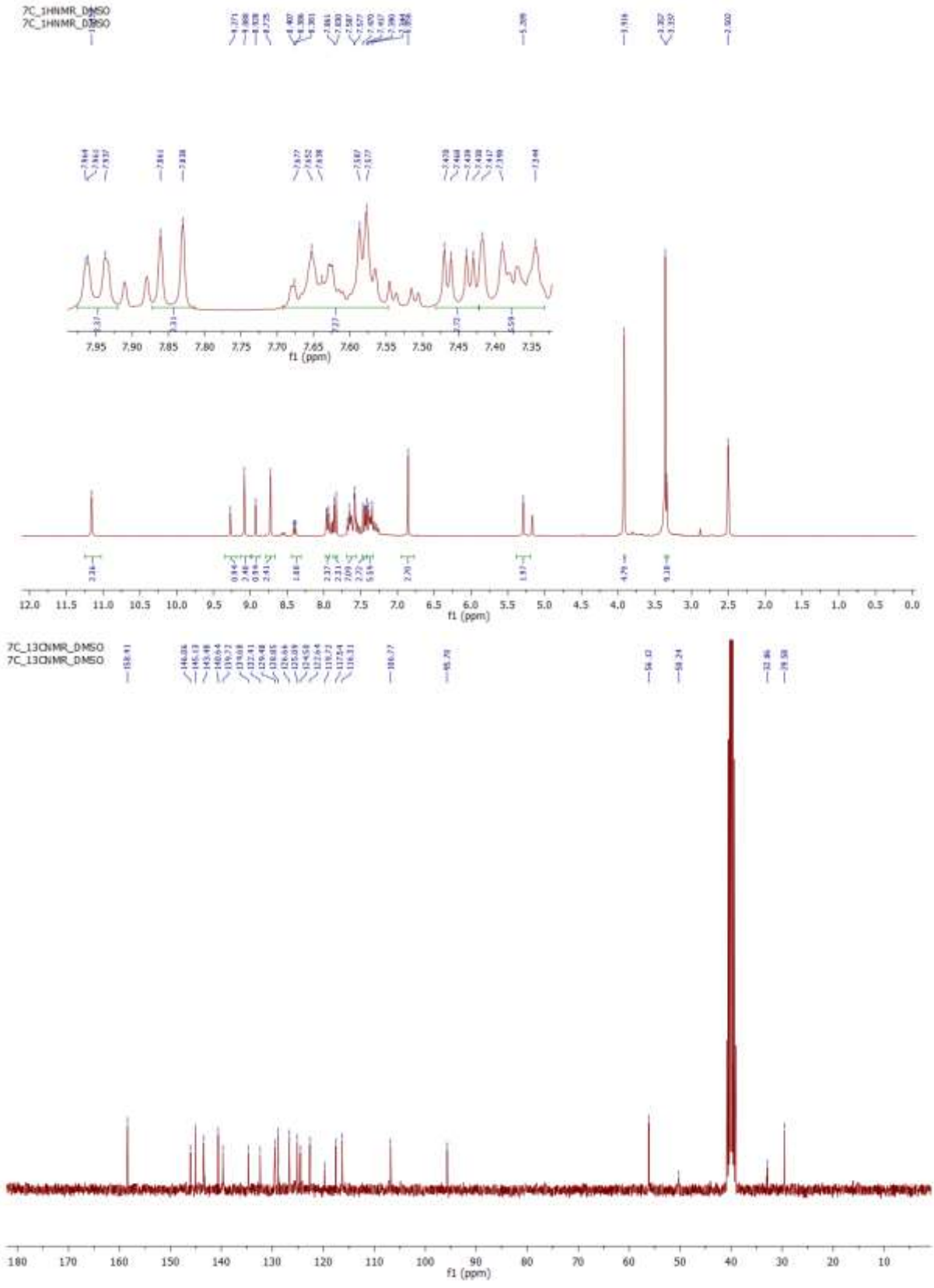


^{1}HNMR and ^{13}C NMR Spectra of 7b

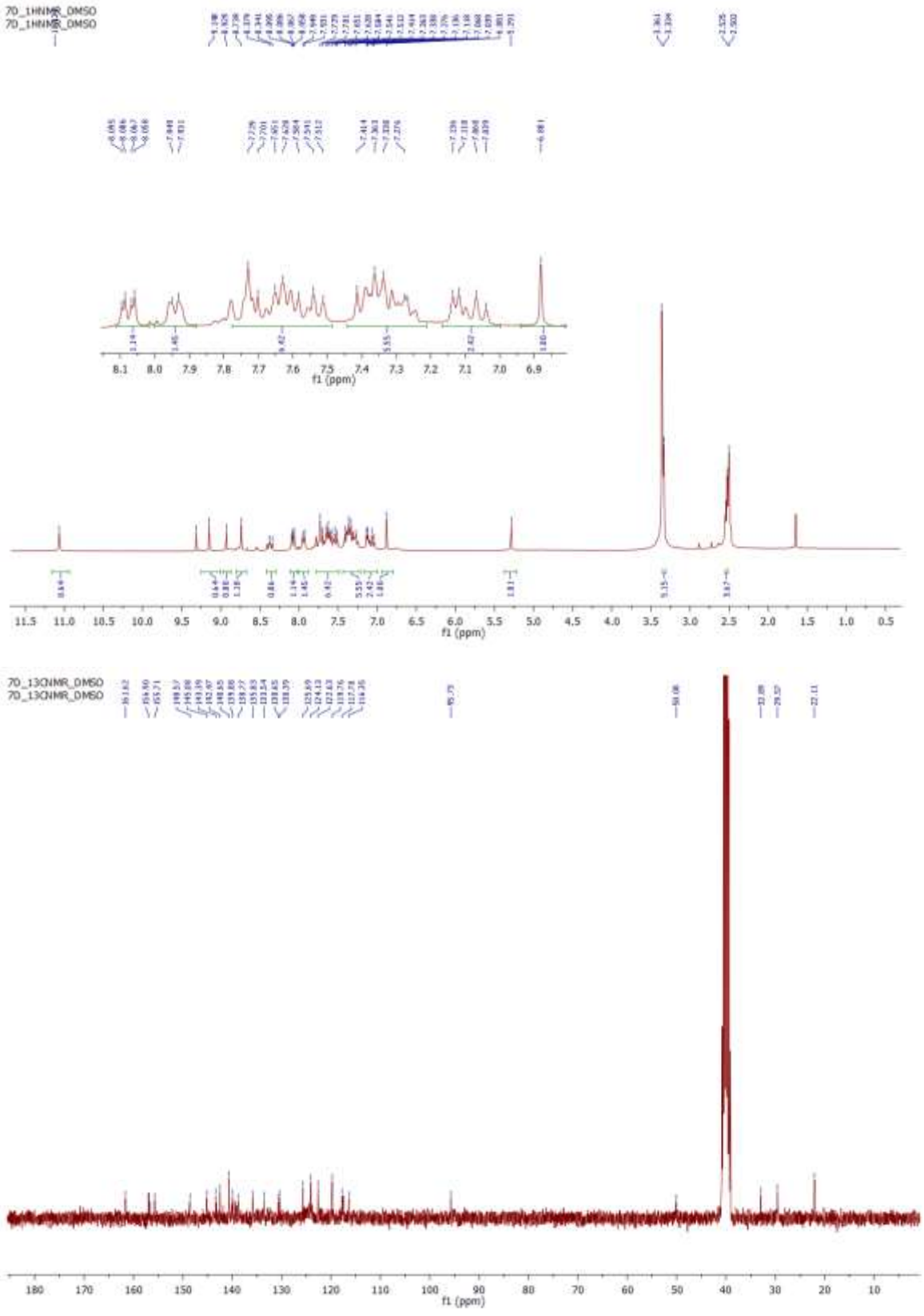




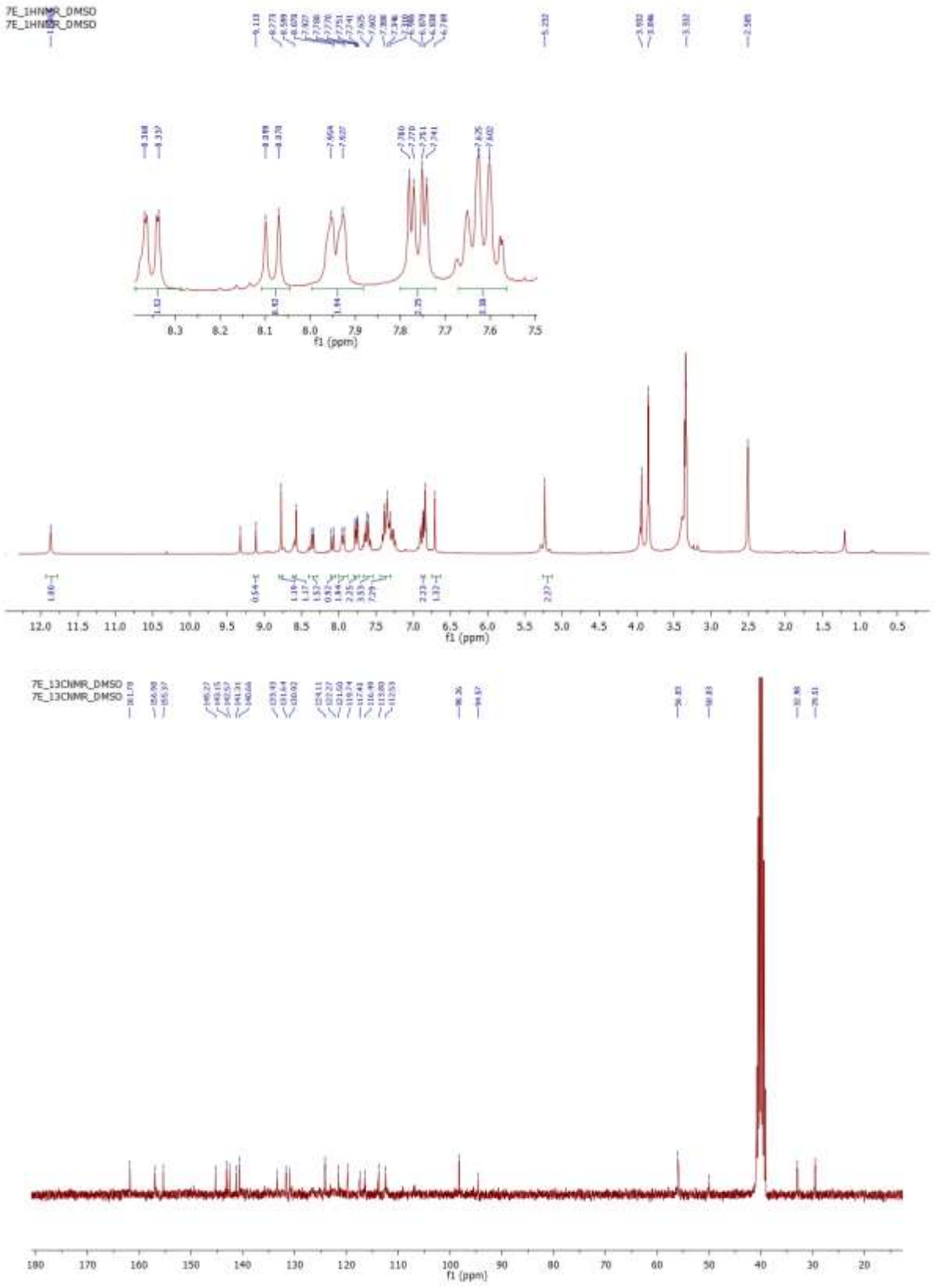
^{1}HNMR and ^{13}C NMR Spectra of 7c



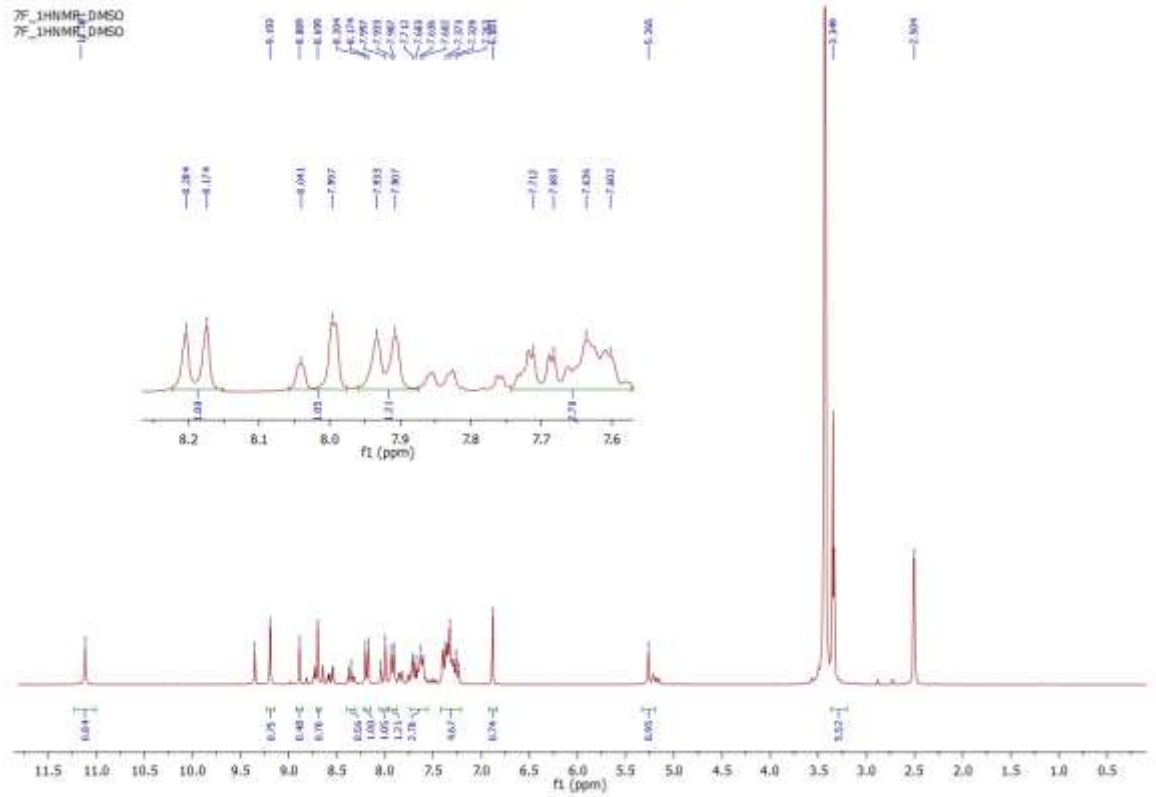
^{1}HNMR and ^{13}C NMR Spectra of 7d

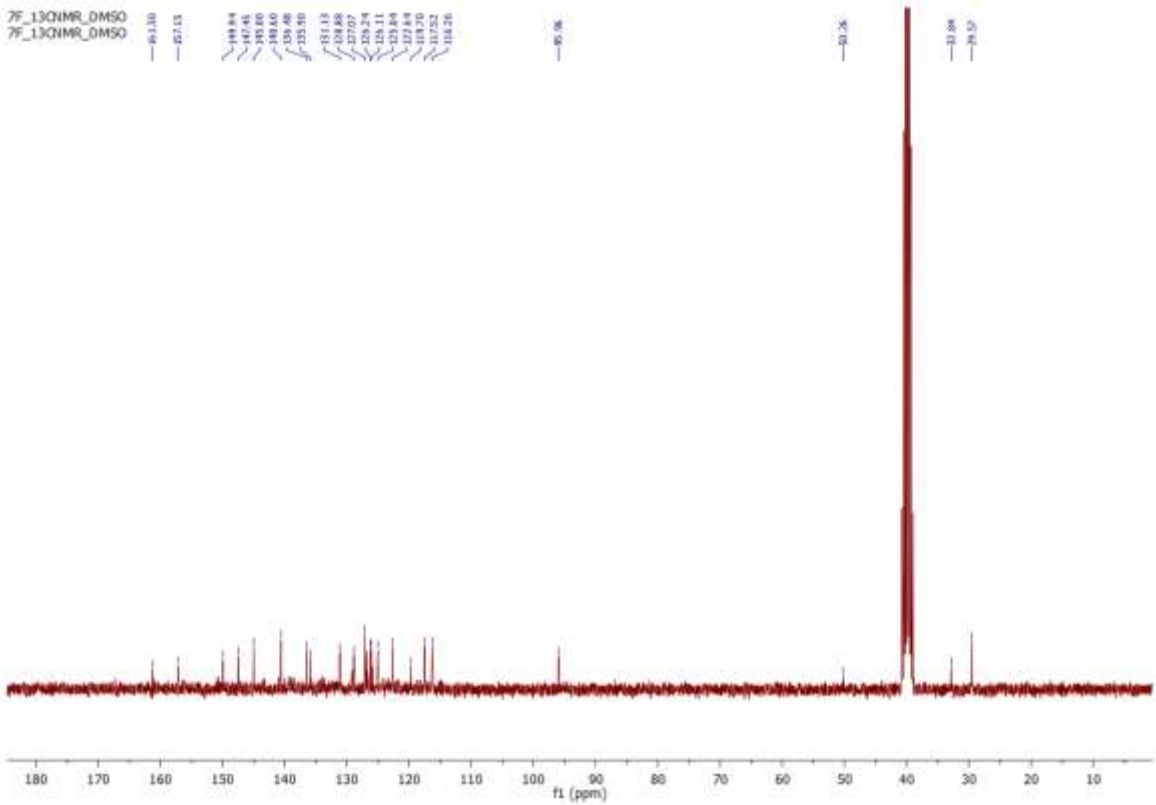


^{1}HNMR and ^{13}C NMR Spectra of 7e



^{1}HNMR and ^{13}C NMR Spectra of 7f



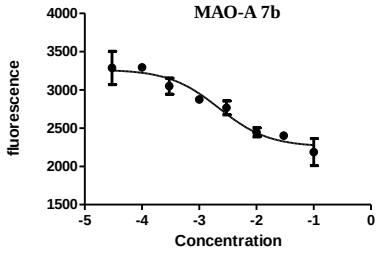
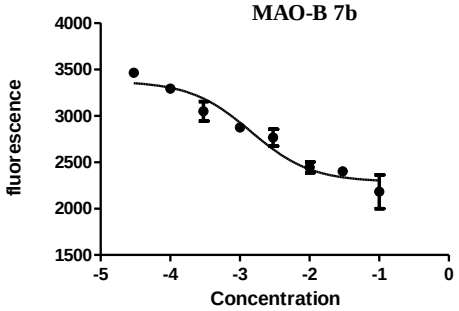
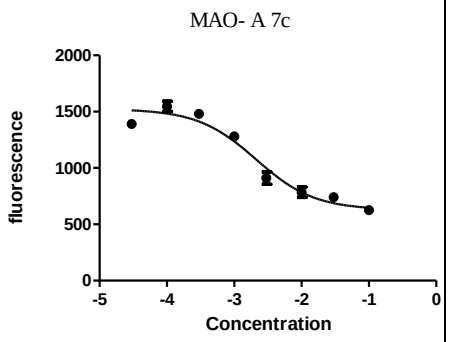
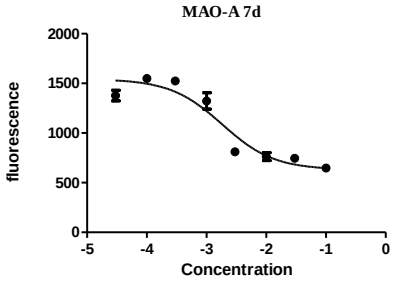
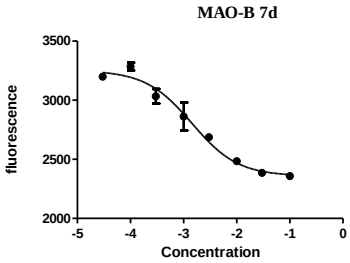
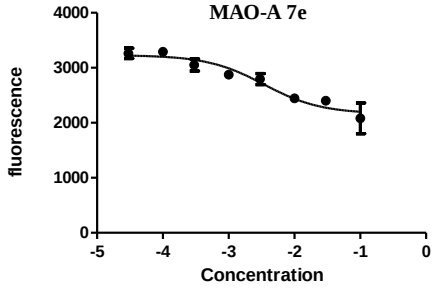
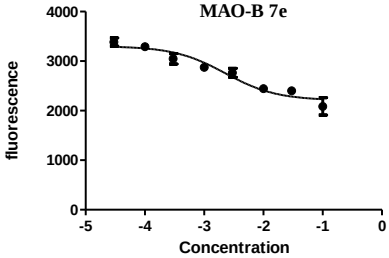


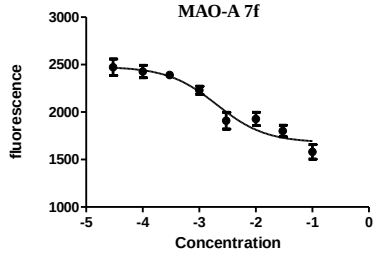
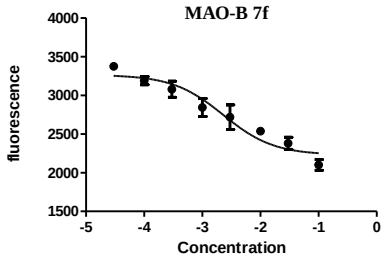
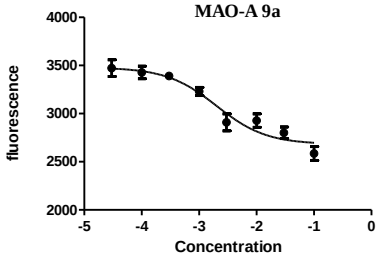
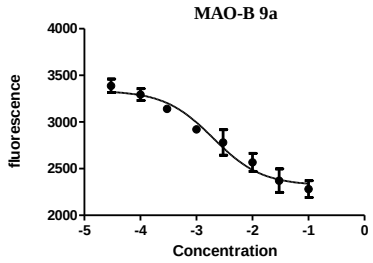
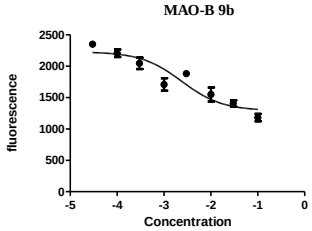
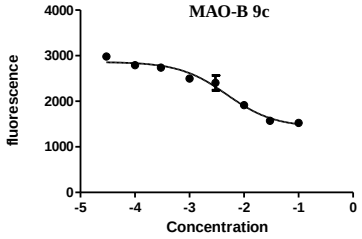
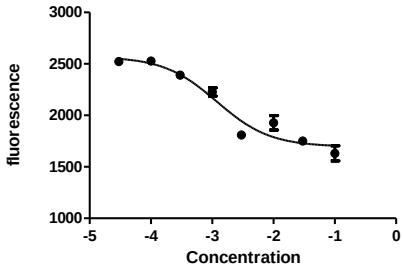
Biological activity

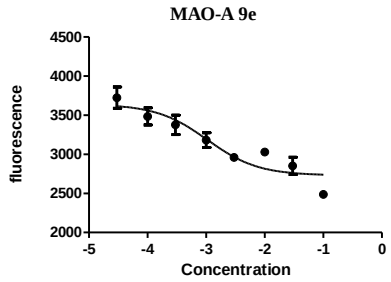
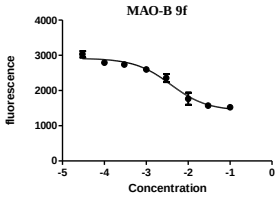
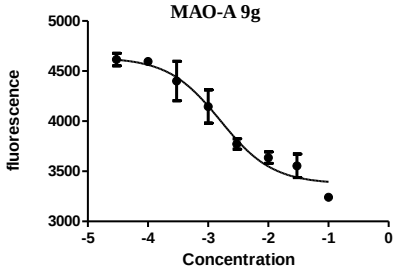
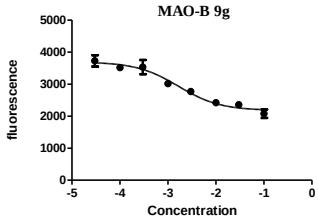
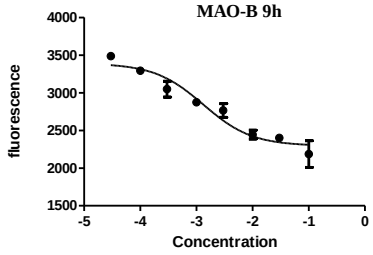
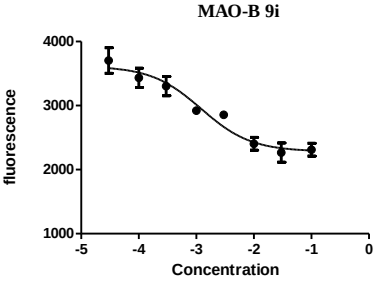
Monoamine oxidase (MAO-A and MAO-B) inhibition assay

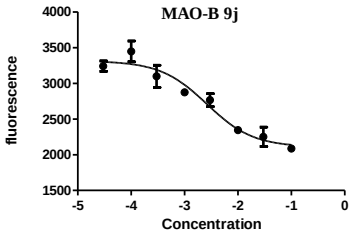
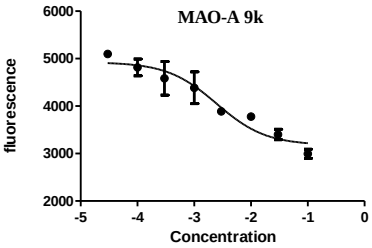
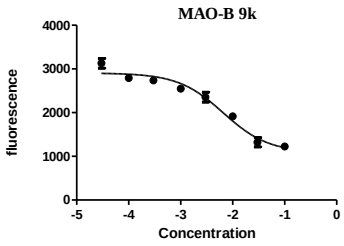
For the newly synthesized compounds, were analyzed on MAO A and MAO B activity was measured as per previously reported protocol. Freshly enzyme was prepared 15-20min before, at cool room temperature. Clorgyline (60 nM) or Deprenyl (300 nM) were used accordingly to block of MAO A and MAOB activity irreversibly. For preforming assay white 96 well plate was used. The assay volume was 100 μL having 60 μL buffer (pH 7.4) 10 μL test compound (0.1mM, 10% DMSO) followed by adding enzyme 10 μL (26μg of protein for MAO A and 5.0 μg for MAO B). The mixture was incubated for 15, 20 min for MAO B and MAO A respectively, 10 μL of substrate and 10 μL of freshly prepared Amplex red was added in the mixture. The final concentration of clorgyline and Deprenyl was 0.1mM used to determine non-MAO A and MAO B activity accordingly. The change in the fluorescence was determined by using fluorescence plate reader (BMG Labtech GmbH, orten berg Germany). The compounds which exhibited over 50% inhibition of either the MAO A or MAO B activity were further evaluated for determination of IC50 values. All experiments were repeated twice in triplicate. IC50 values were calculated by non-linear curve fitting program PRISM 5.0 (GraphPad, San Diego, California, USA)

Table S1: IC50 Values and % Inhibition of the Synthesized Derivatives

S. No.	Code	MAO A	IC_{50} & % inhibition	MAO B	IC_{50} & % inhibition
1s	7a		38.8[a]		43.6[a]
2	7b		2.32 ±0.91[b]		1.43 ± 0.89[b]
3	7c		2.10± 0.09 b		35.75[a]
4	7d		1.82 ± 0.24 [b]		1.05 ± 0.66[b]
5	7e		3.43 ±0.52[b]		2.22 ± 0.94[a]

6	7f		2.03 ±0.91[a]		1.21 ± 0.17 [b]
7	9a		2.09±0.89[a]		1.98 ± 0.14 [b]
8	9b		38.5[a]		2.38 ± 0.19 [b]
9	9c		39.1[a]		3.83± 0.48 [b]
10	9d		1.27 ± 0.10 [b]		31.09[a]

11	9e		1.04± 0.01 [b]		41.93[a]
12	9f		41.6[a]		3.82 ± 0.37 [b]
13	9g		1.52± 0.56 [b]		1.25 ± 0.28 [b]
14	9h		28.4[a]		1.03 ± 0.17 [b]
15	9i		39.8[a]		1.21 ± 0.13 [b]

16	9j		26.3[a]		2.61 ±0.37 [b]
17	9k		2.48 ± 0.70 [b]		2.58 ± 0.48 [b]

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