

## **Supplementary Materials**

### **Structure-based design of potent pyrazolo[1,5-*a*]pyrimidine CDK4/6 inhibitors: Biological evaluation and computational validation**

Faizah A. Binjubair<sup>a</sup>, Mahmoud S. Elkotamy<sup>b,\*</sup>, Amr A. Mattar<sup>c</sup>, Bjad K. Almutairy<sup>d</sup>,  
Sara T. Al-Rashood<sup>a</sup>, Mohamed M. Eldesouki<sup>e</sup>, Jalloul Bouajila<sup>f</sup>, Hatem A. Abdel-Aziz<sup>g</sup>,  
Mariam M. Fakhry<sup>h,\*</sup>

<sup>a</sup> *Department of Pharmaceutical Chemistry, College of Pharmacy, King Saud University, P.O. Box 2457, Riyadh 11451, Saudi Arabia*

<sup>b</sup> *Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Alsalam University, Kafr Alzayat 31611, Algharbia, Egypt*

<sup>c</sup> *Pharmaceutical Chemistry Department, Faculty of Pharmacy, El Saleheya El Gadida University, Egypt*

<sup>d</sup> *Department of Pharmaceutics, College of Pharmacy, Prince Sattam Bin Abdulaziz University, Al-Kharj, Saudi Arabia*

<sup>e</sup> *Scientific Research and Innovation Support Unit, Faculty of Pharmacy, Kafrelsheikh University, Kafrelsheikh, 33516, Egypt*

<sup>f</sup> *Laboratoire de Génie Chimique, Université de Toulouse, Toulouse INP, CNRS, LGC, Toulouse, France*

<sup>g</sup> *Applied Organic Chemistry Department, National Research Centre, Dokki, Cairo, 12622, Egypt*

<sup>h</sup> *Department of Pharmaceutical Chemistry, Faculty of Pharmacy, Egyptian Russian University, Badr City, Cairo, 11829, Egypt*

#### **Correspondence:**

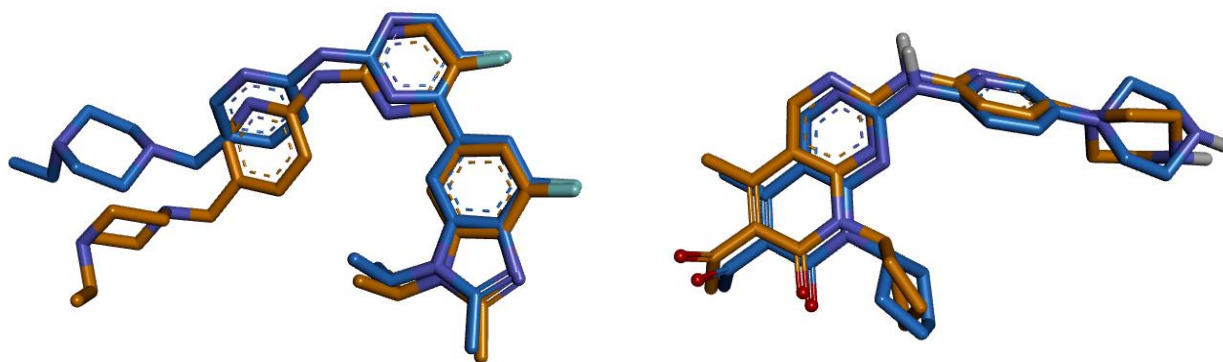
[mahmoudekotami@gmail.com](mailto:mahmoudekotami@gmail.com), [mariam-medhat@eru.edu.eg](mailto:mariam-medhat@eru.edu.eg)

# Content

<b>Docking Studies</b> .....	<b>4</b>
Figure S1. Docking protocol validation for CDK4 and CDK6. Superposition of the co-crystallized ligand (orange) and the redocked pose (blue) within the active site of CDK4 (left) and CDK6 (right), demonstrating the reliability of the applied docking protocol through the close overlap of binding conformations.....	5
<b><sup>1</sup>H NMR and <sup>13</sup>C NMR Spectra</b> .....	<b>6</b>
Figure S2. <sup>1</sup> H NMR of compound 19a .....	7
Figure S3. <sup>13</sup> C NMR of compound 19a.....	8
Figure S4. <sup>1</sup> H NMR of compound 19b .....	9
Figure S5. <sup>13</sup> C NMR of compound 19b .....	10
Figure S6. <sup>1</sup> H NMR of compound 19c.....	11
Figure S7. <sup>13</sup> C NMR of compound 19c.....	12
Figure S8. <sup>1</sup> H NMR of compound 19d .....	13
Figure S9. <sup>13</sup> C NMR of compound 19d .....	14
Figure S10. <sup>1</sup> H NMR of compound 19e .....	15
Figure S11. <sup>13</sup> C NMR of compound 19e .....	16
Figure S12. <sup>1</sup> H NMR of compound 19f .....	17
Figure S13. <sup>13</sup> C NMR of compound 19f .....	18
Figure S14. <sup>1</sup> H NMR of compound 19g .....	19
Figure S15. <sup>13</sup> C NMR of compound 19g.....	20
Figure S16. <sup>1</sup> H NMR of compound 19h .....	21
Figure S17. <sup>13</sup> C NMR of compound 19h .....	22
Figure S18. <sup>1</sup> H NMR of compound 19i .....	23
Figure S19. <sup>13</sup> C NMR of compound 19i .....	24
Figure S20. <sup>1</sup> H NMR of compound 19j .....	25
Figure S21. <sup>13</sup> C NMR of compound 19j .....	26
Figure S22. <sup>1</sup> H NMR of compound 19k.....	27
Figure S23. <sup>13</sup> C NMR of compound 19k.....	28
Figure S24. <sup>1</sup> H NMR of compound 19l .....	29
Figure S25. <sup>13</sup> C NMR of compound 19l .....	30
Figure S26. <sup>1</sup> H NMR of compound 19m .....	31
Figure S27. <sup>13</sup> C NMR of compound 19m .....	32

Figure S28. $^1\text{H}$ NMR of compound 19n .....	33
Figure S29. $^{13}\text{C}$ NMR of compound 19n .....	34
Figure S30. $^1\text{H}$ NMR of compound 19o .....	35

# Docking Studies



**Figure S1.** Docking protocol validation for CDK4 and CDK6. Superposition of the co-crystallized ligand (**orange**) and the redocked pose (**blue**) within the active site of CDK4 (**left**) and CDK6 (**right**), demonstrating the reliability of the applied docking protocol through the close overlap of binding conformations.

# **$^1\text{H}$ NMR and $^{13}\text{C}$ NMR Spectra**

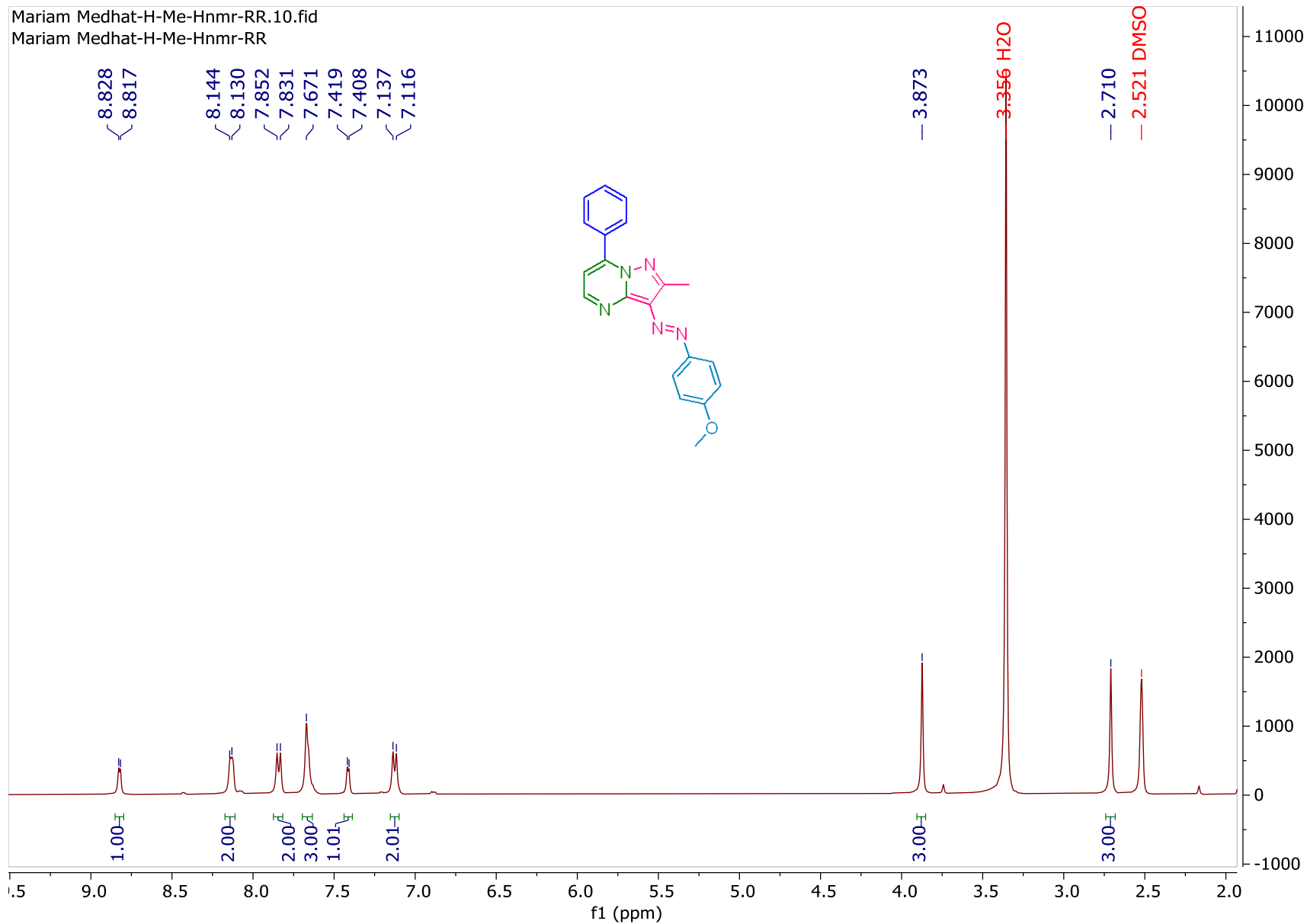


Figure S2. <sup>1</sup>H NMR of compound 19a

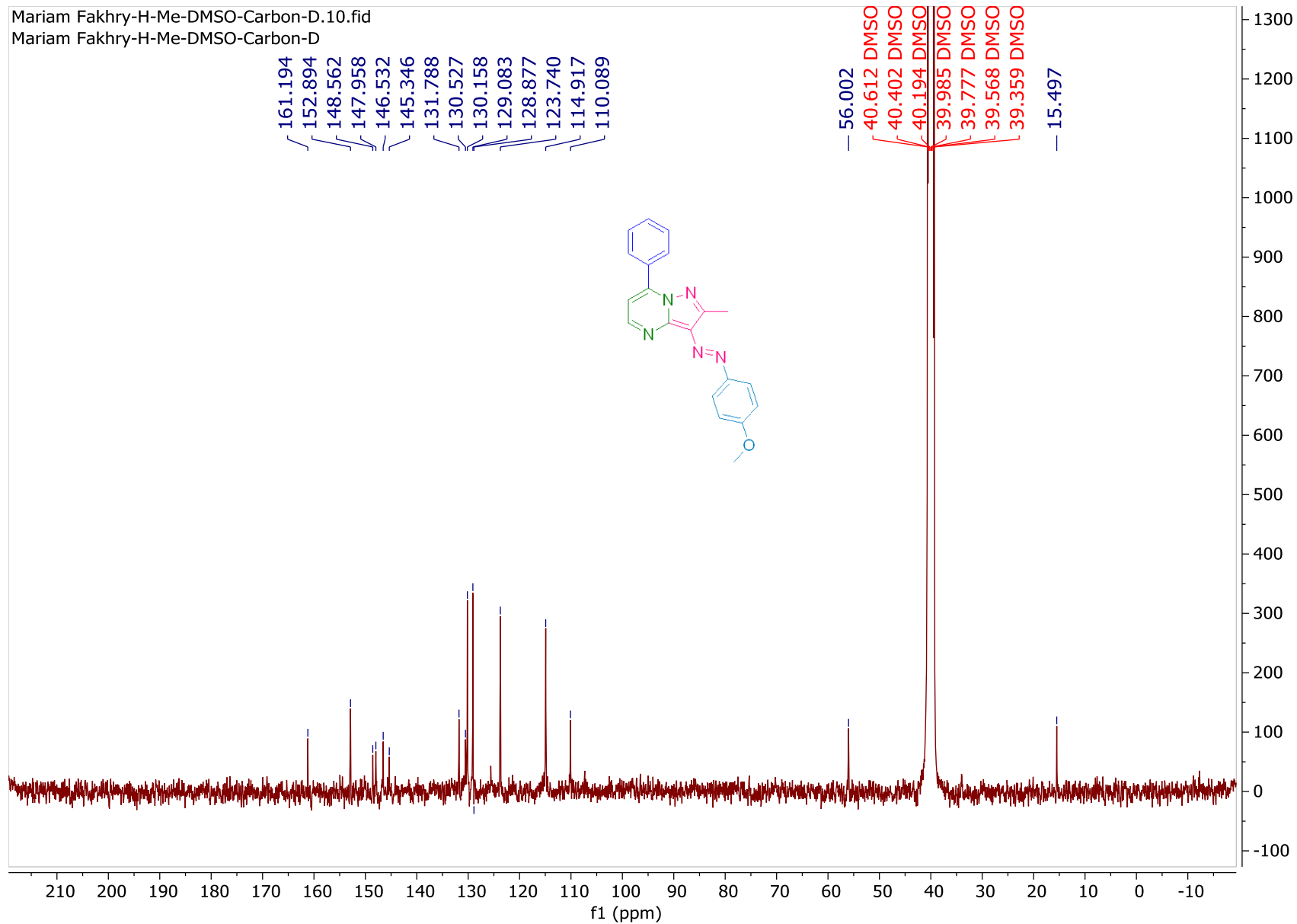


Figure S3.  $^{13}\text{C}$  NMR of compound 19a

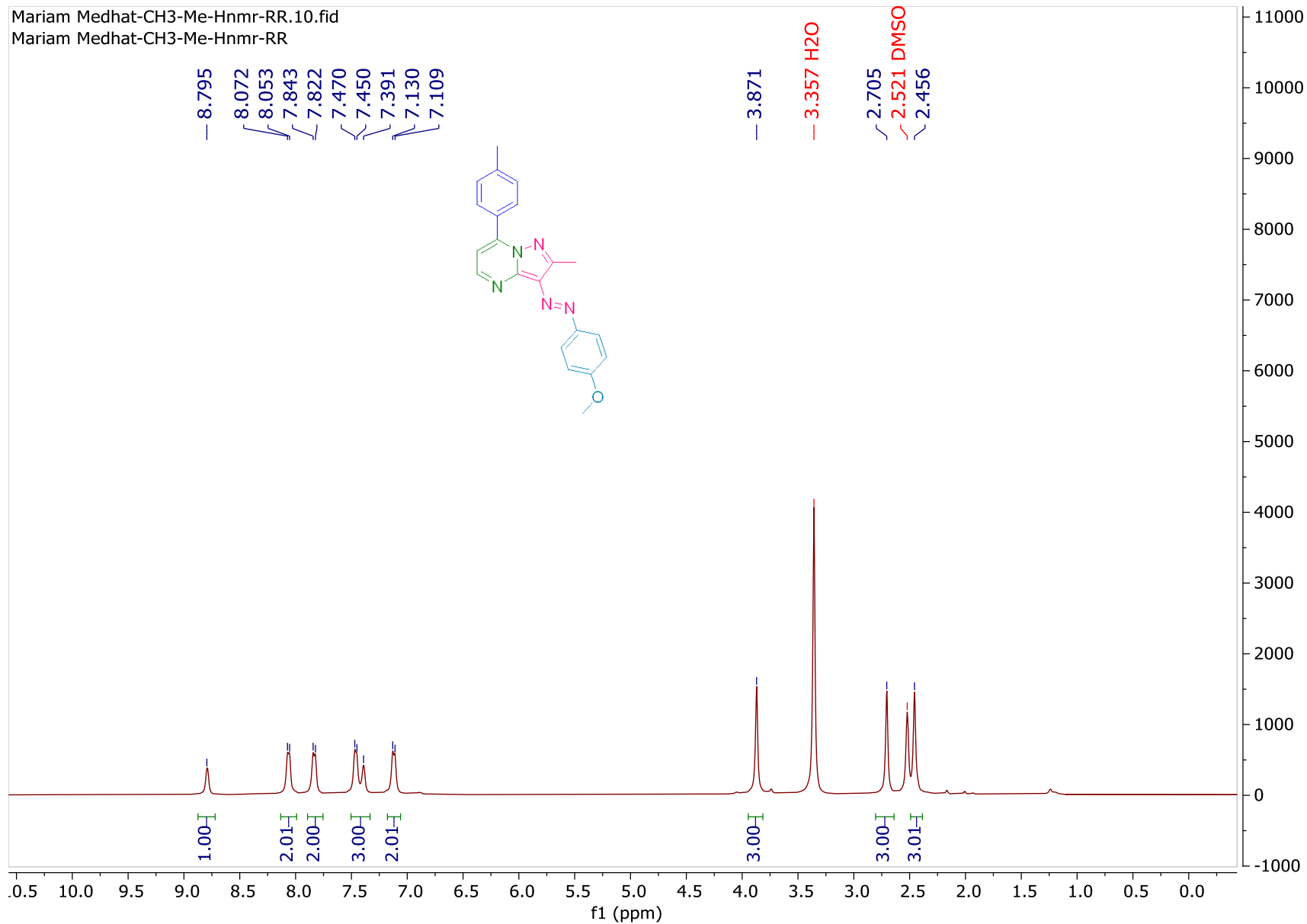


Figure S4. <sup>1</sup>H NMR of compound 19b

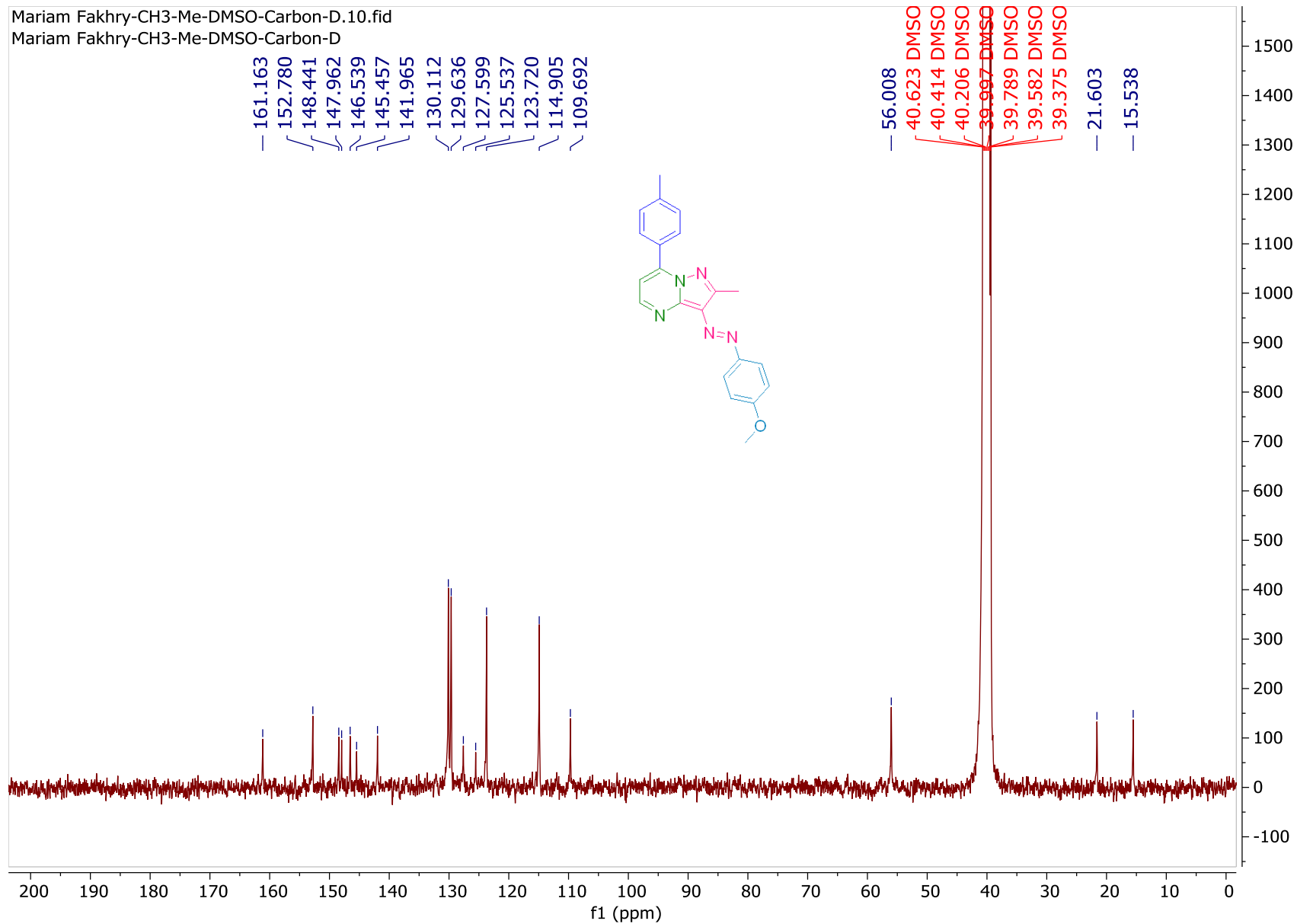


Figure S5.  $^{13}\text{C}$  NMR of compound 19b

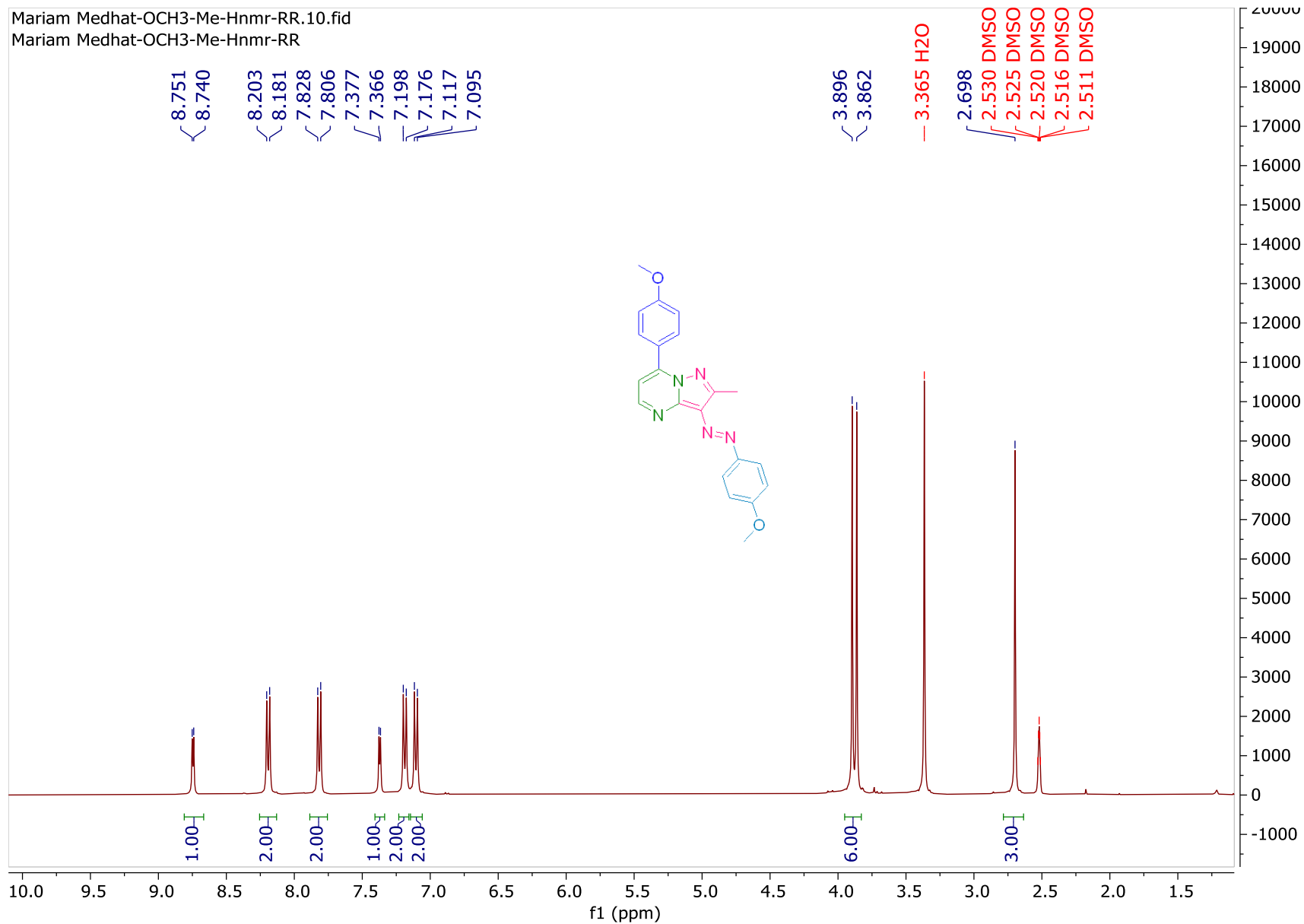


Figure S6. <sup>1</sup>H NMR of compound 19c

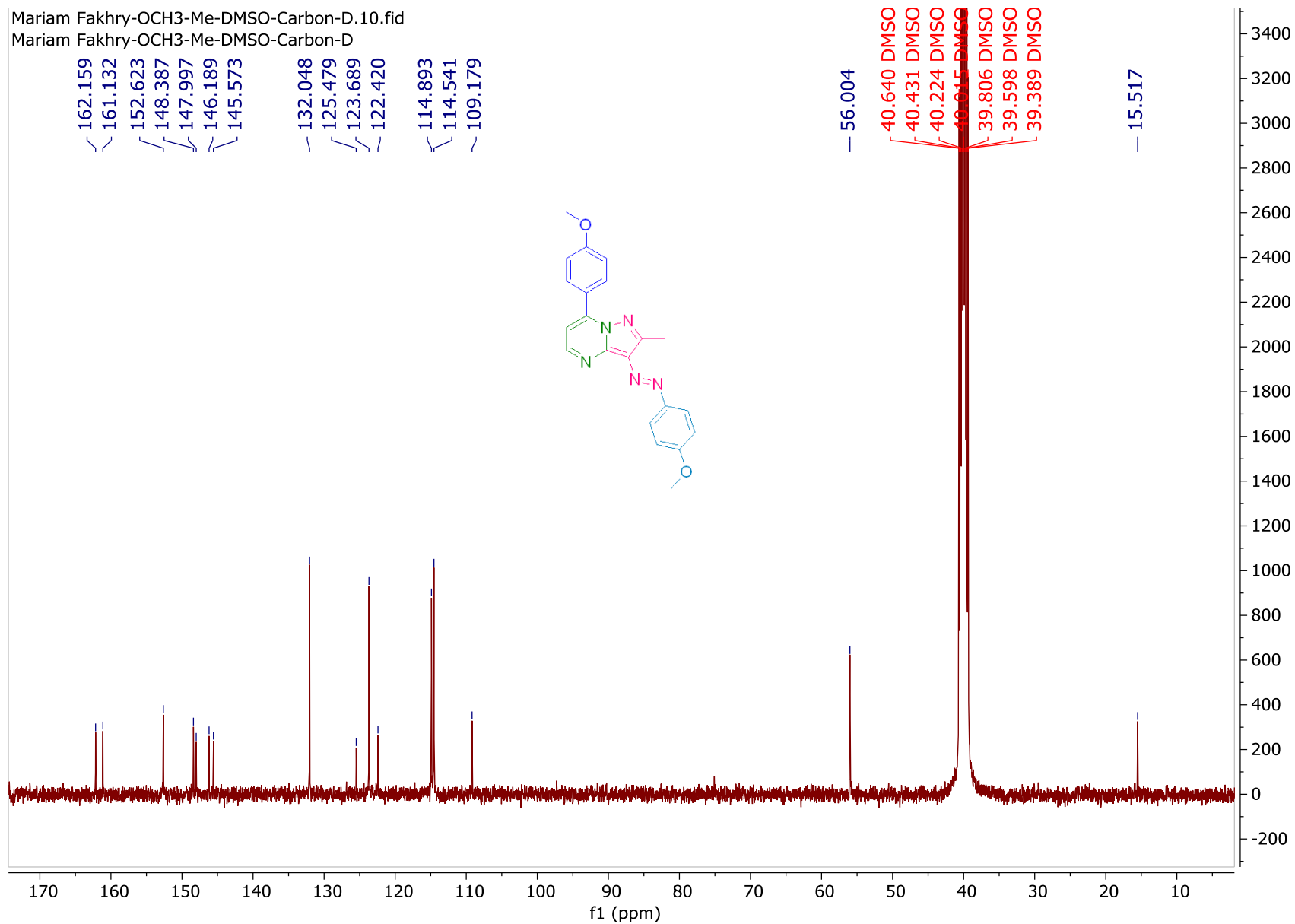


Figure S7. <sup>13</sup>C NMR of compound 19c

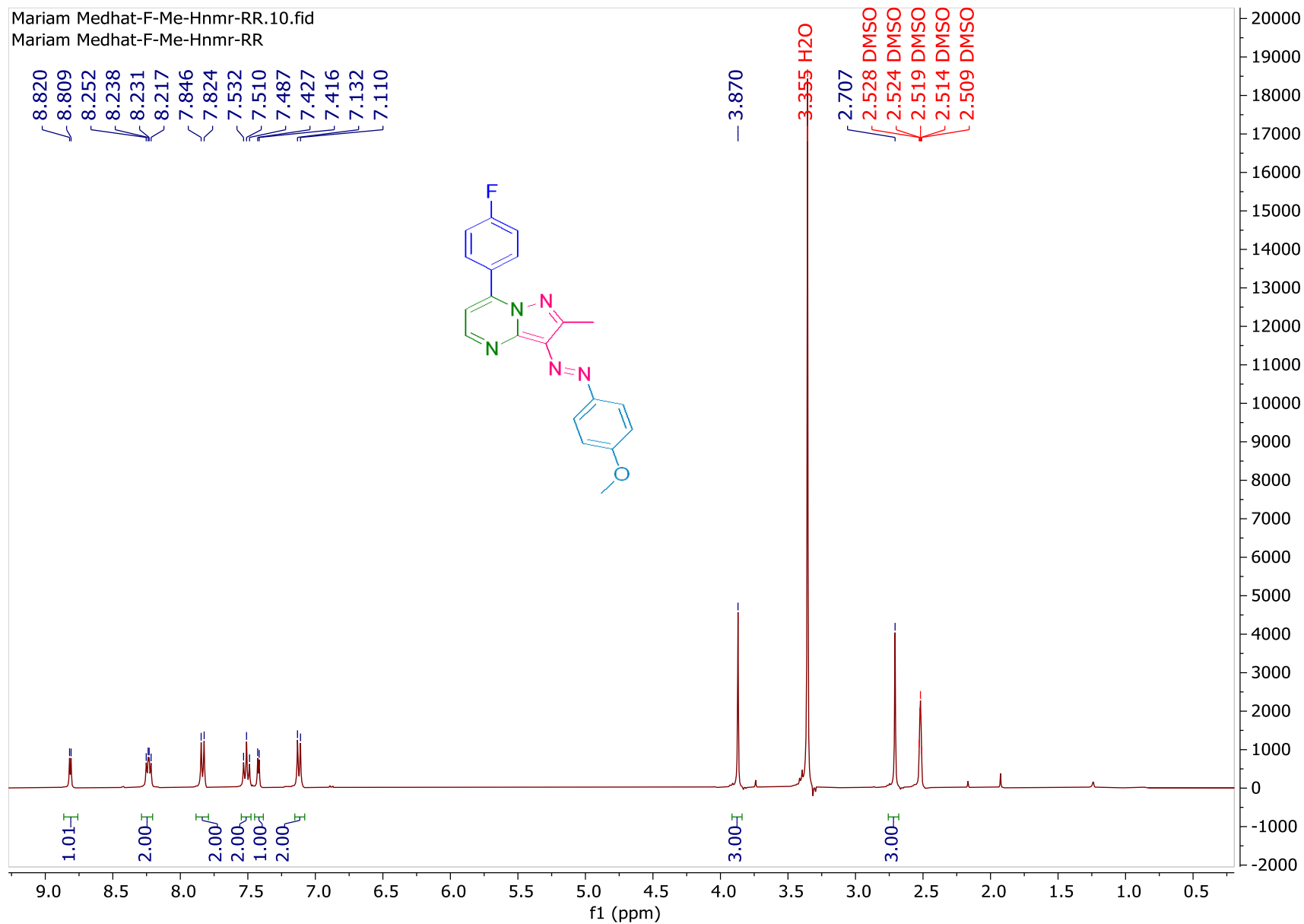


Figure S8. <sup>1</sup>H NMR of compound 19d

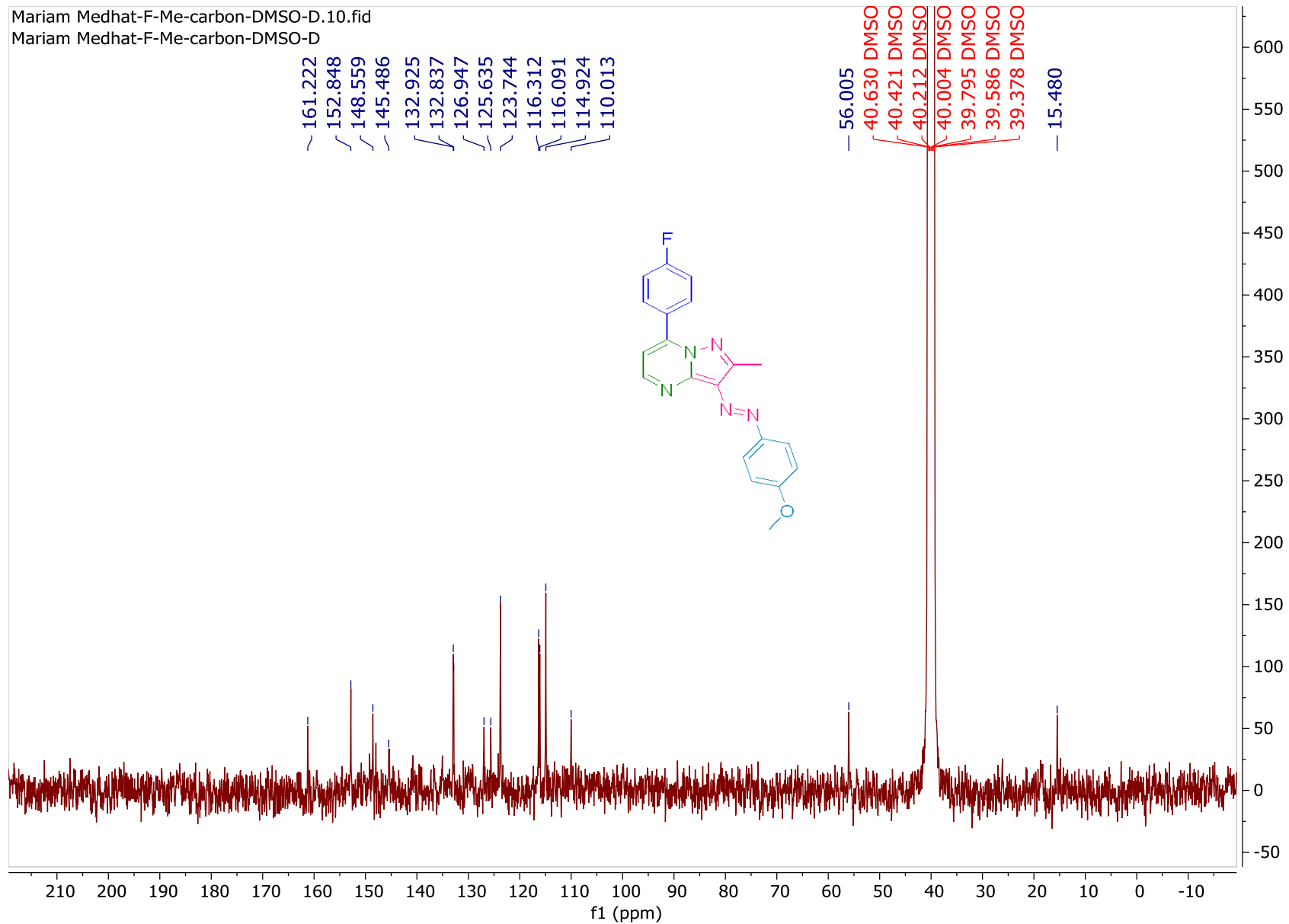


Figure S9. <sup>13</sup>C NMR of compound 19d

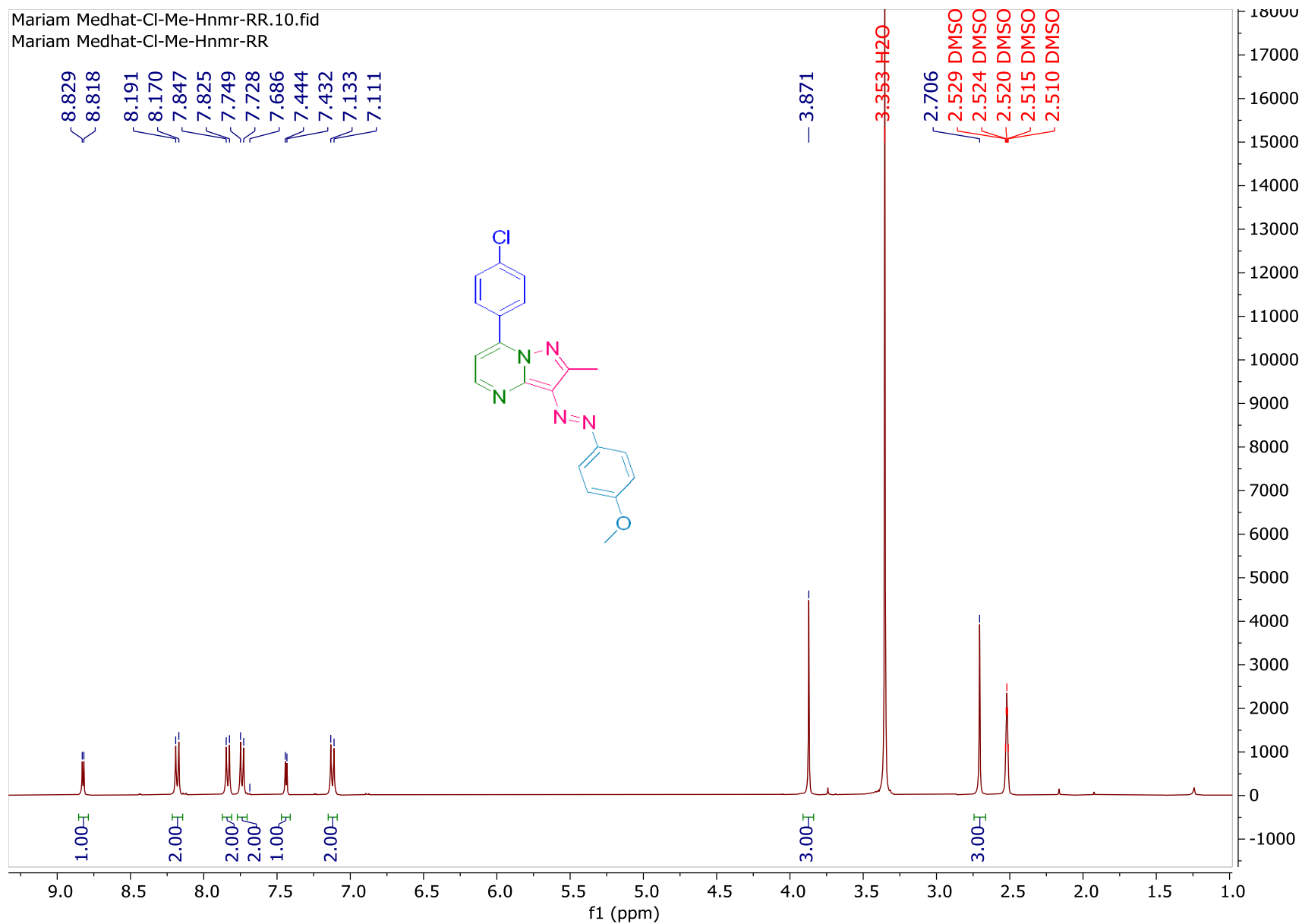


Figure S10. <sup>1</sup>H NMR of compound 19e

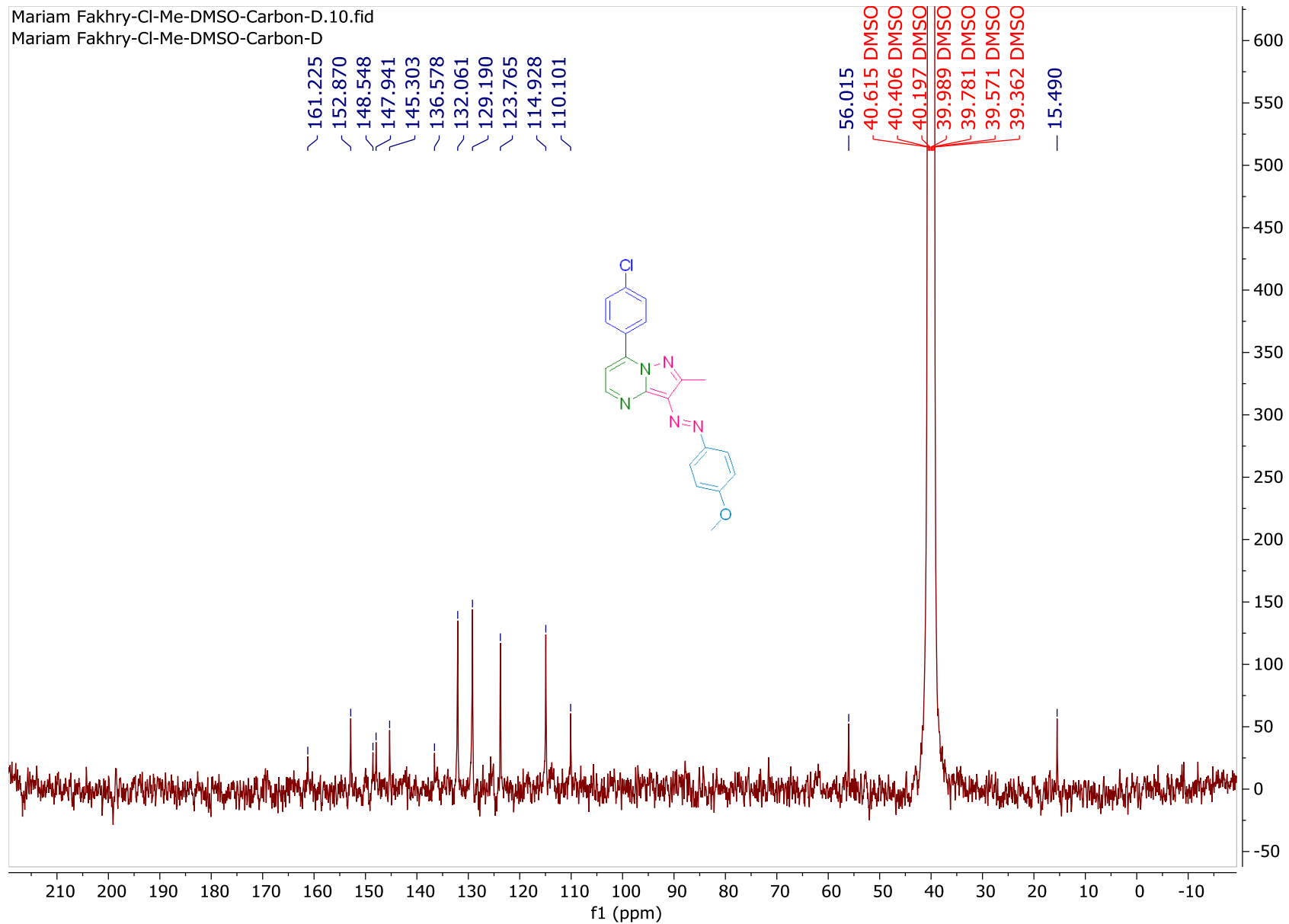


Figure S11. <sup>13</sup>C NMR of compound 19e

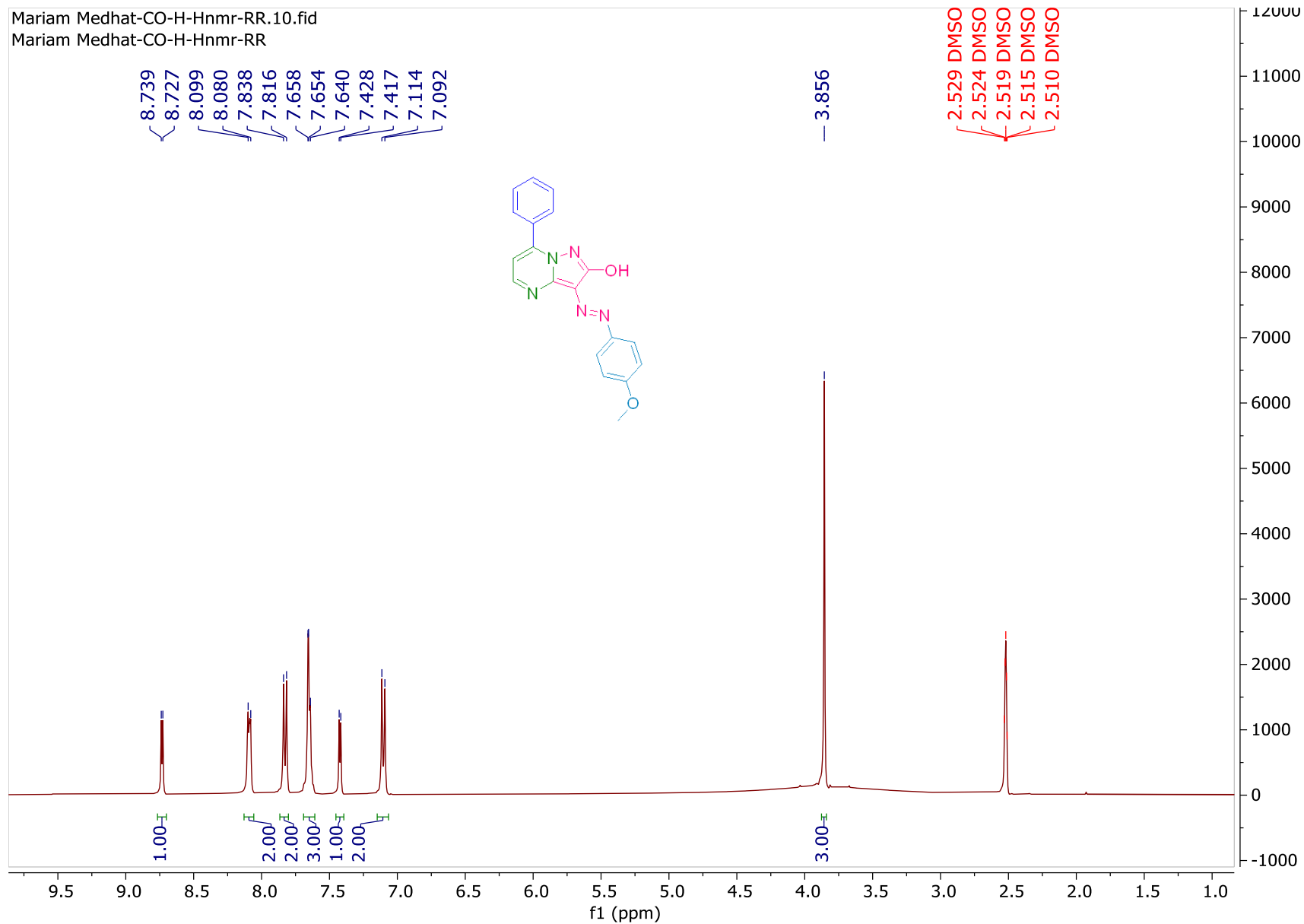


Figure S12.  $^1\text{H}$  NMR of compound 19f

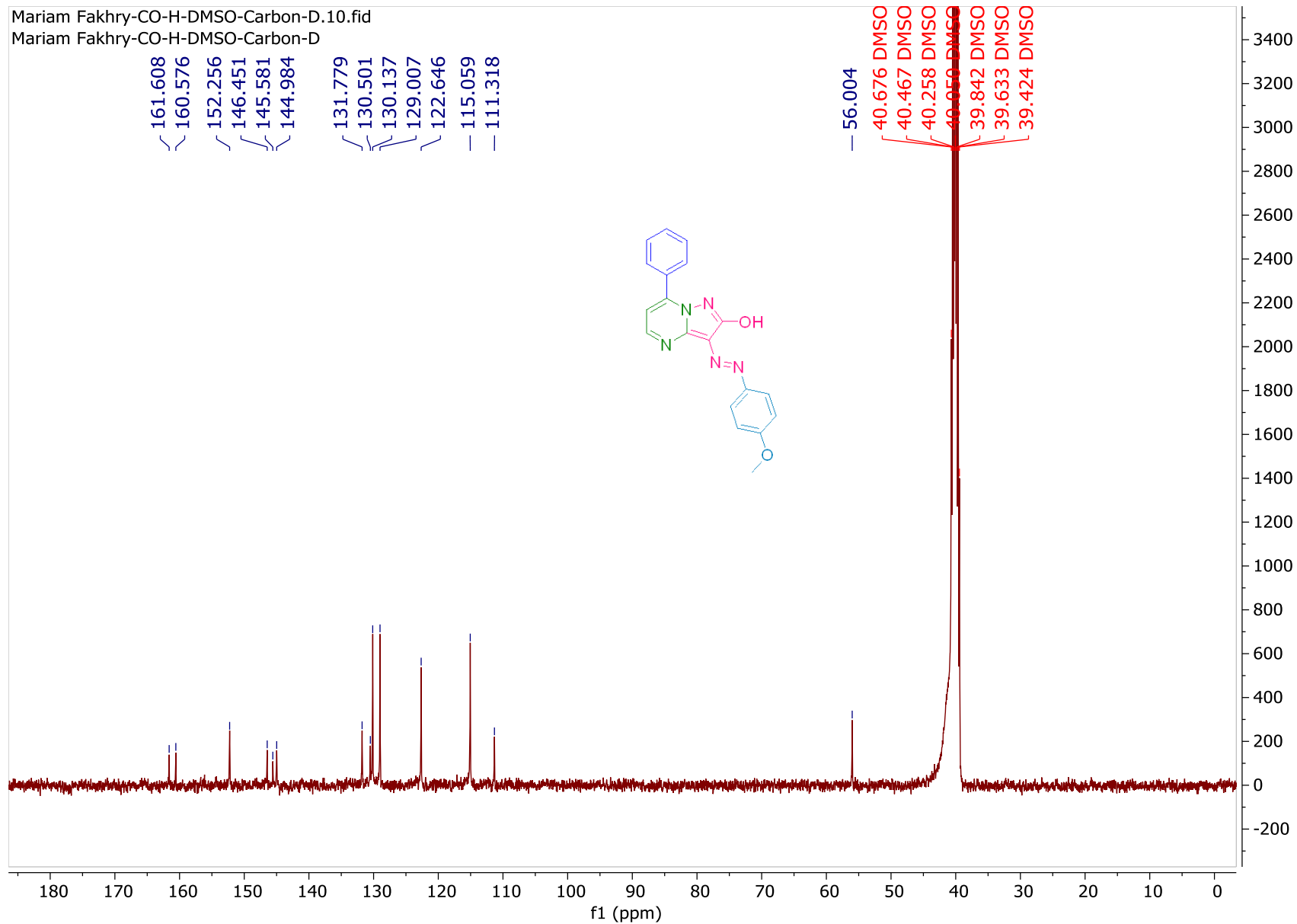


Figure S13.  $^{13}\text{C}$  NMR of compound 19f

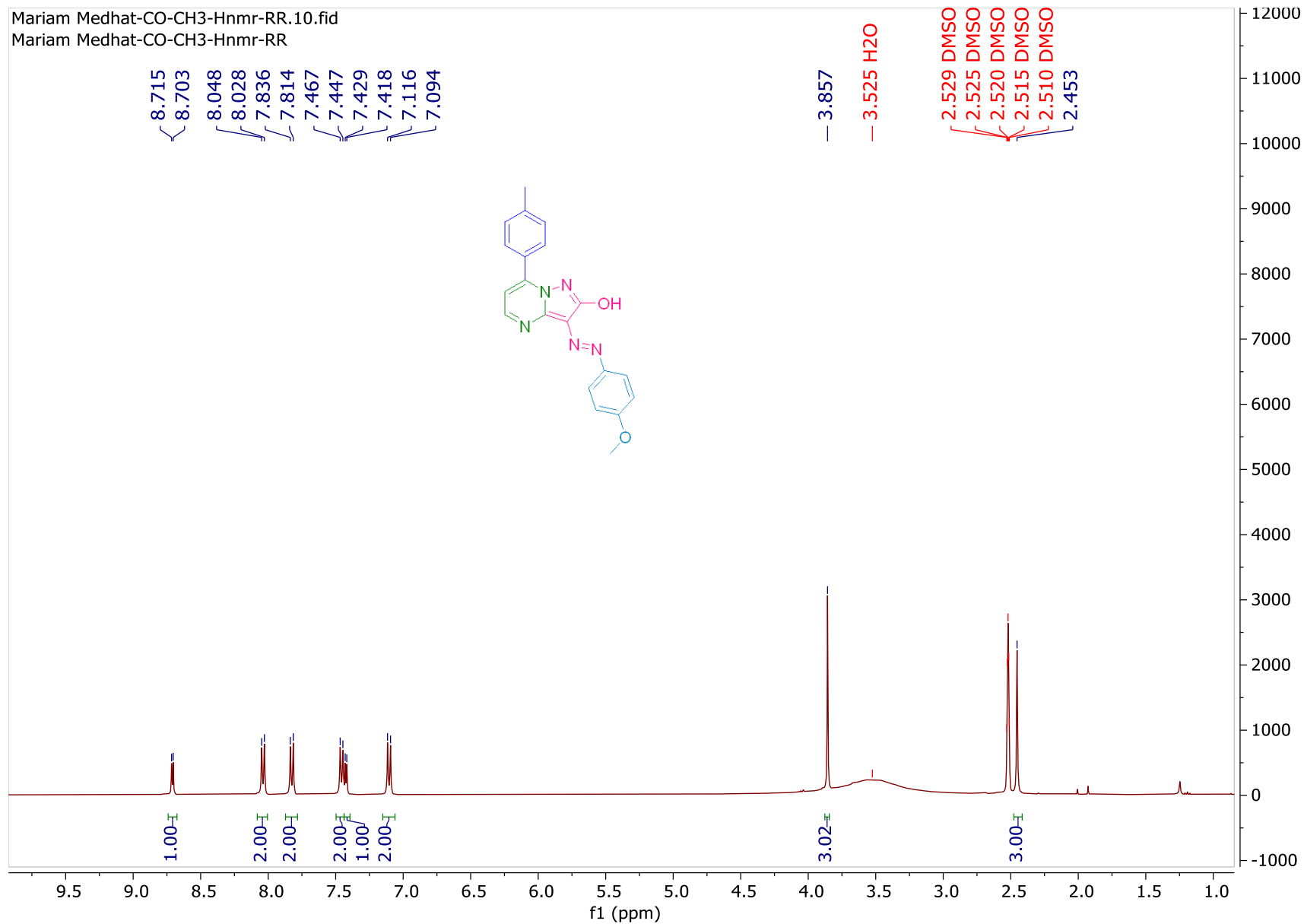


Figure S14. <sup>1</sup>H NMR of compound 19g

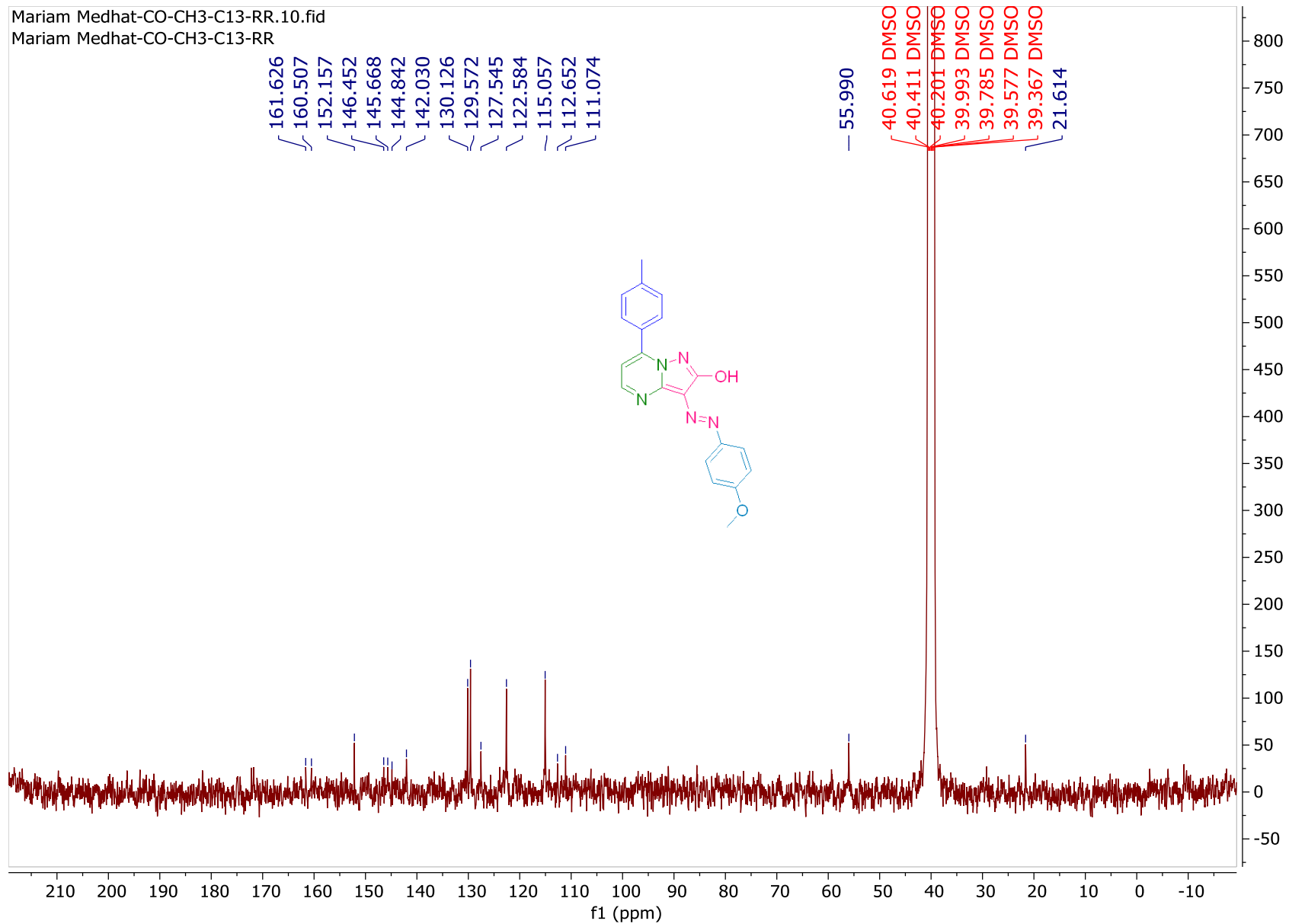


Figure S15.  $^{13}\text{C}$  NMR of compound 19g

Mariam Medhat-CO-OCH3-Hnmr-RR.10.fid  
Mariam Medhat-CO-OCH3-Hnmr-RR

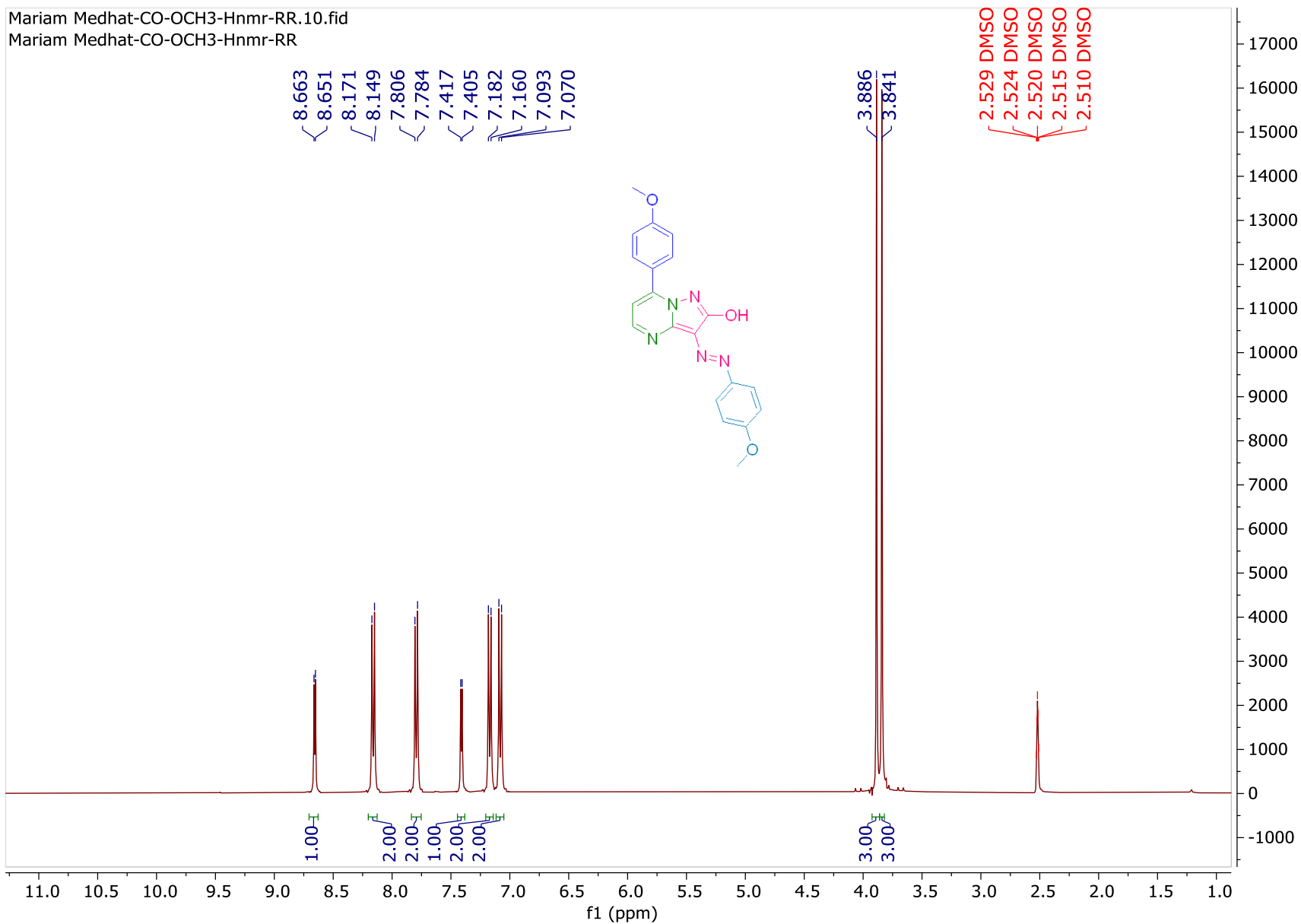


Figure S16.  $^1\text{H}$  NMR of compound 19h

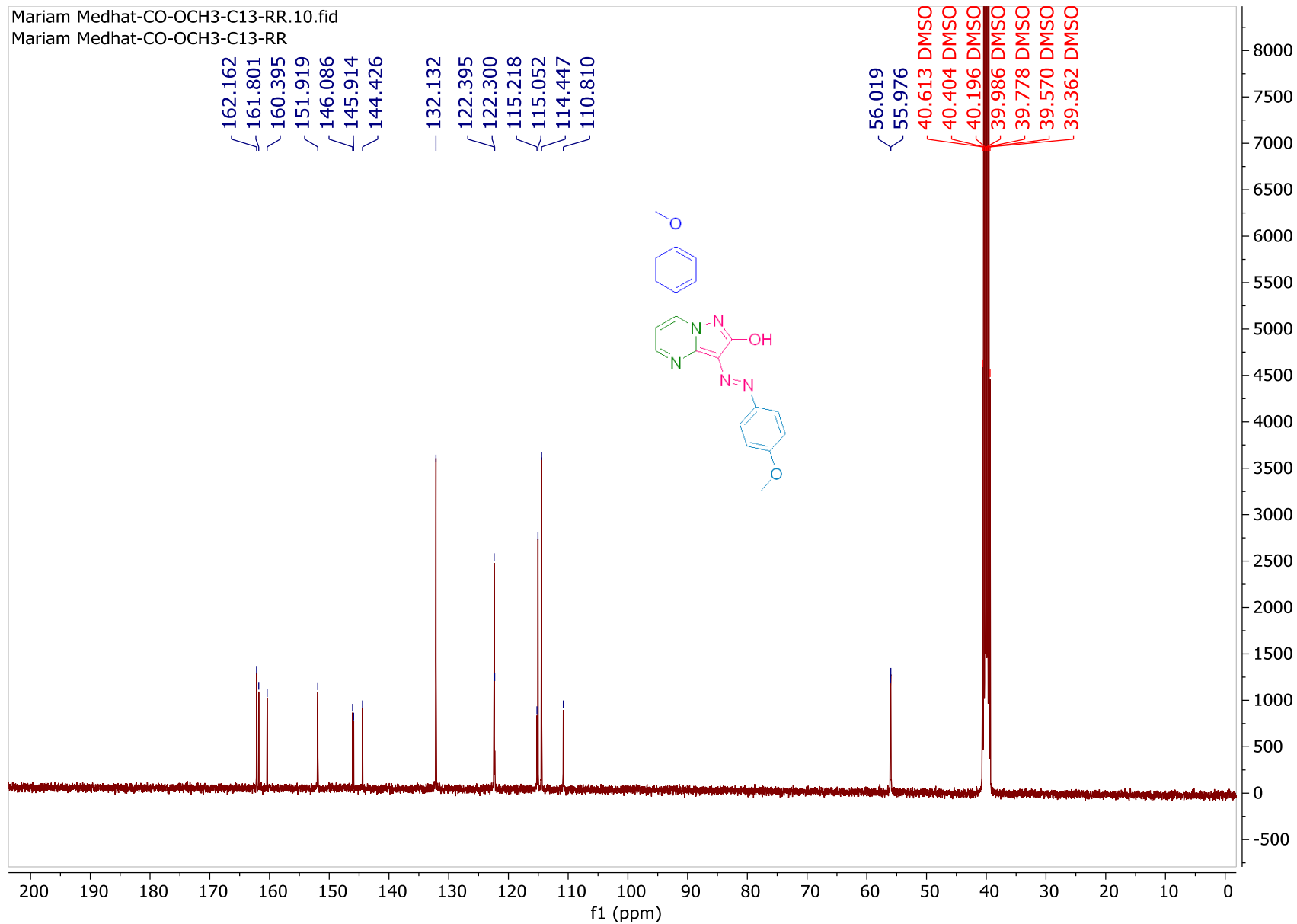


Figure S17. <sup>13</sup>C NMR of compound 19h

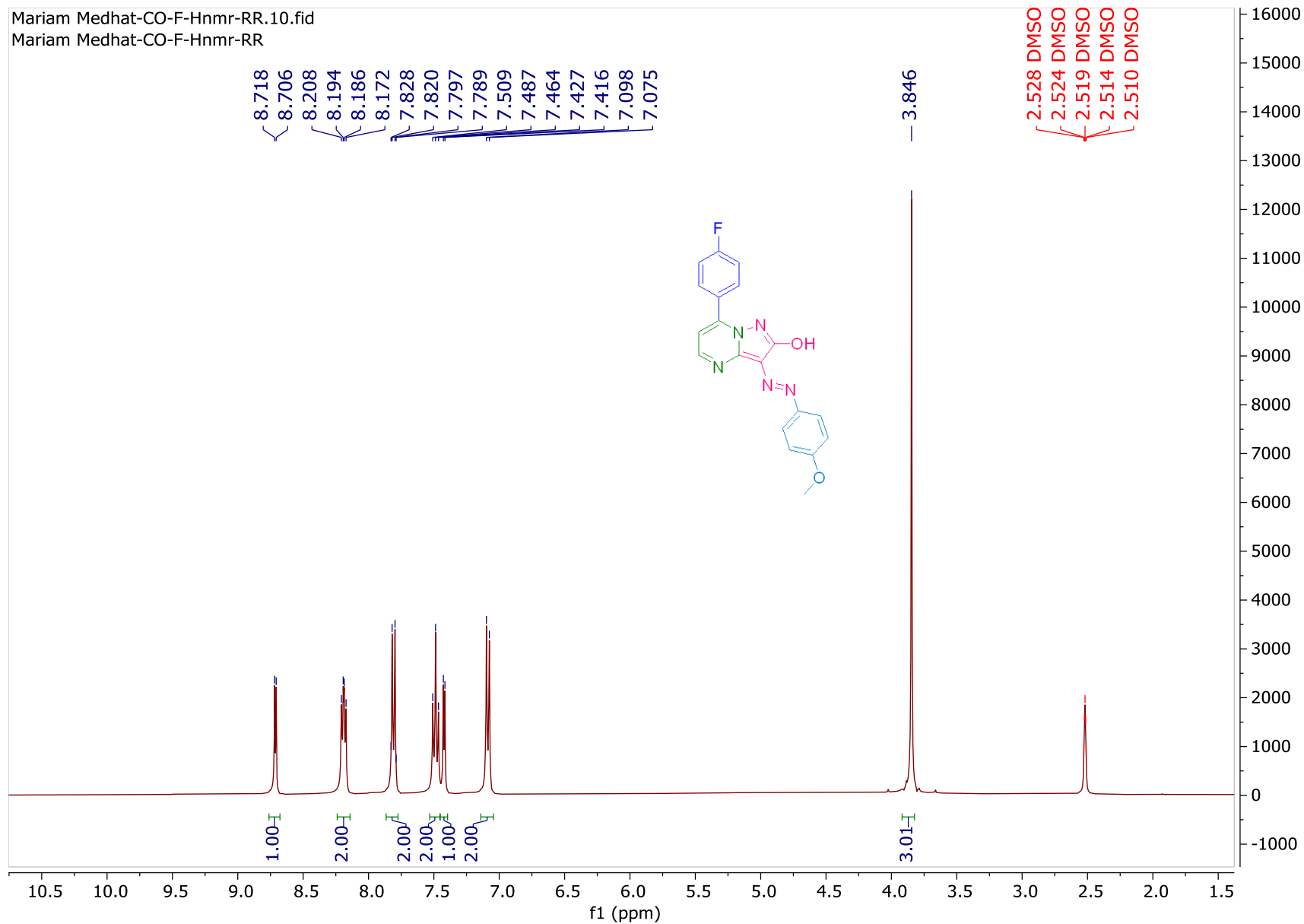


Figure S18. <sup>1</sup>H NMR of compound 19i

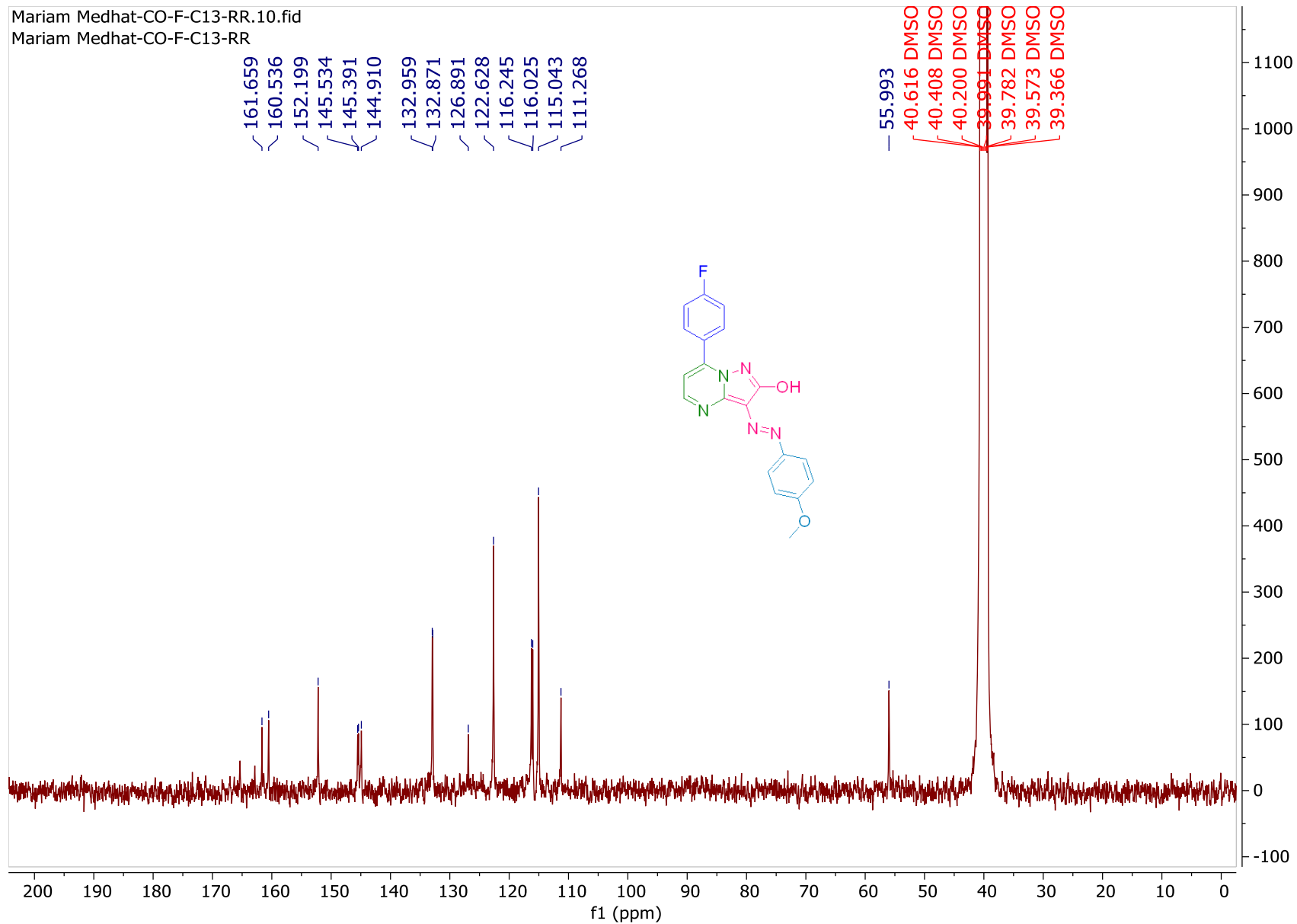


Figure S19.  $^{13}\text{C}$  NMR of compound 19i

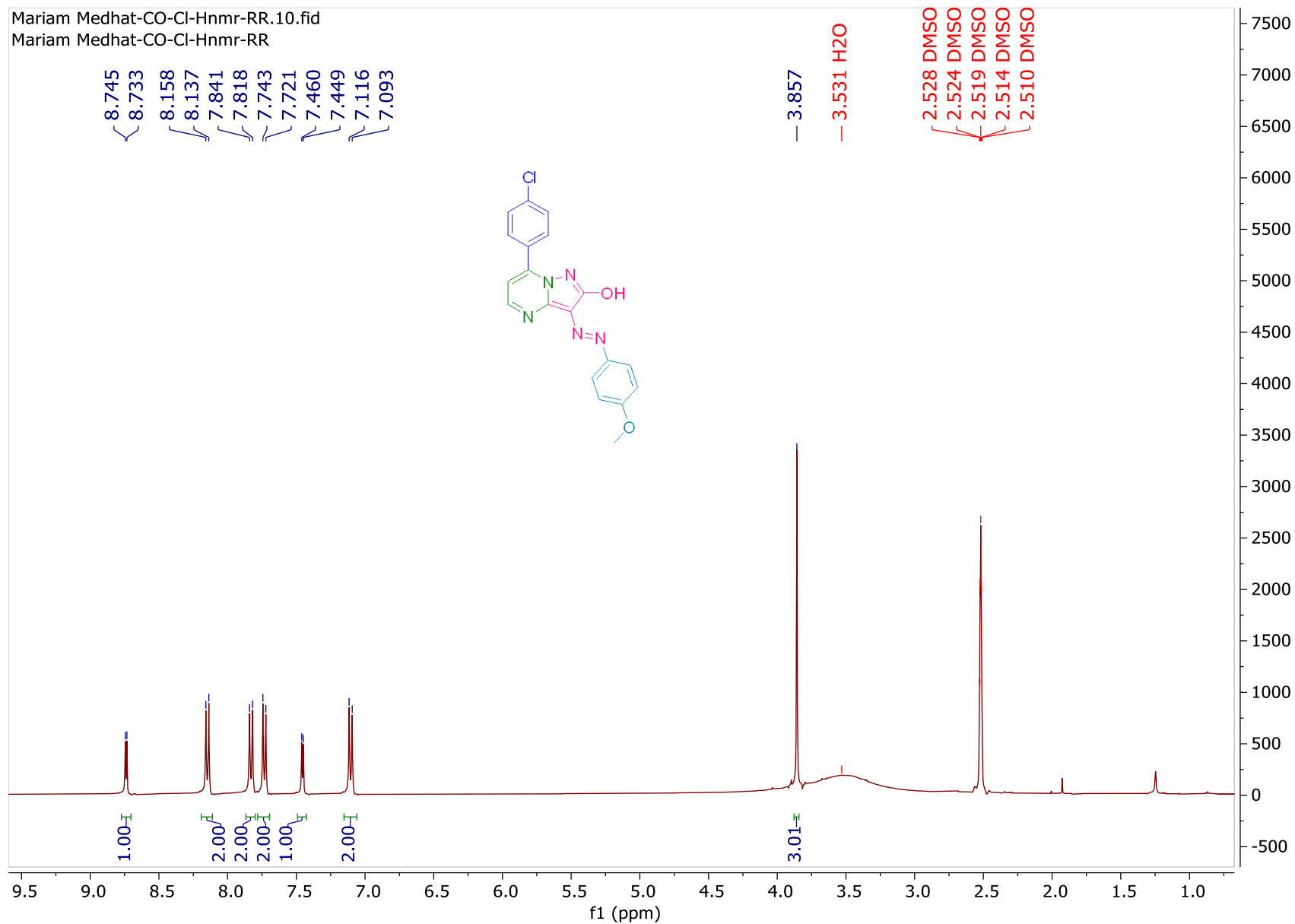


Figure S20. <sup>1</sup>H NMR of compound 19j

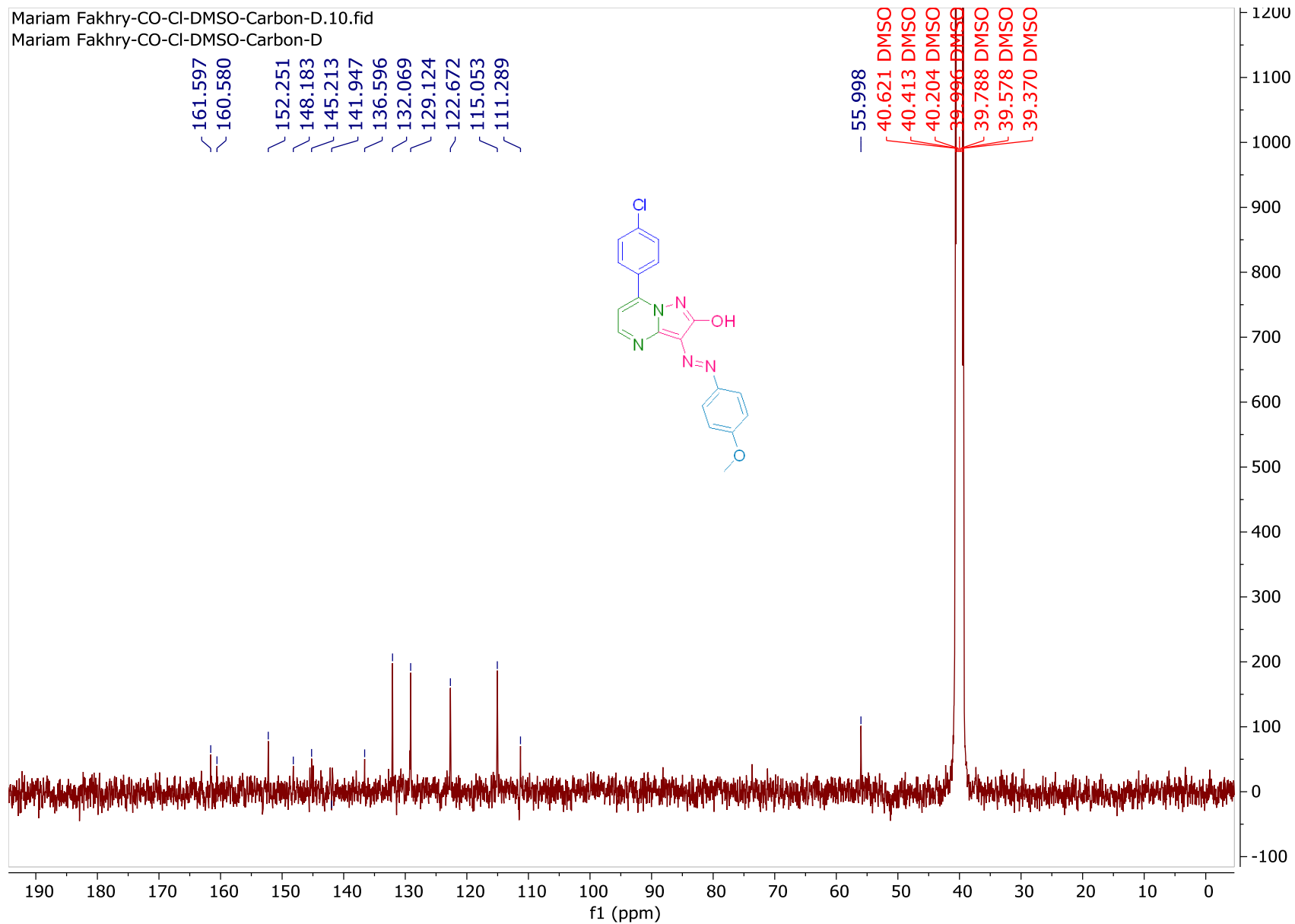


Figure S21.  $^{13}\text{C}$  NMR of compound 19j

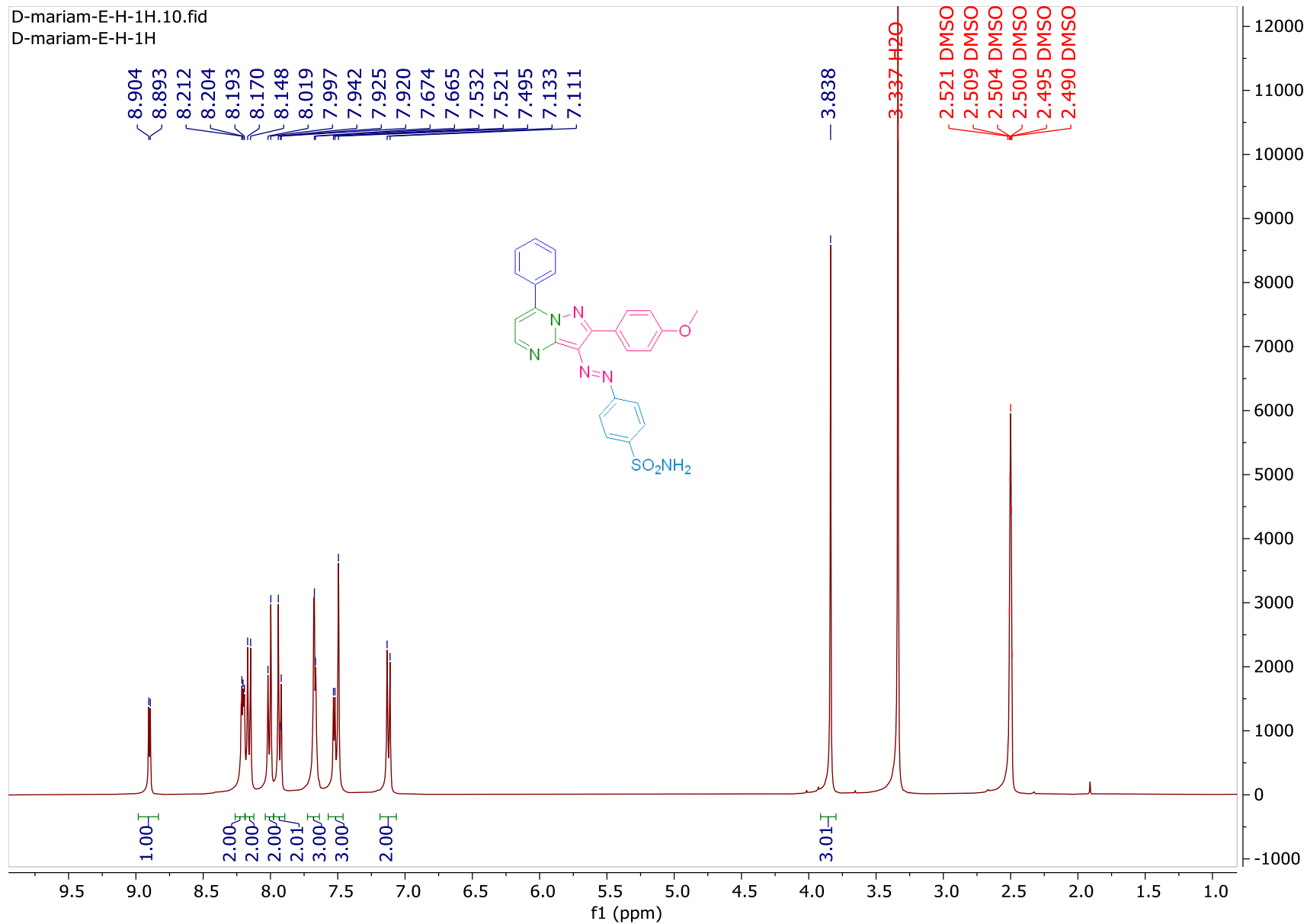


Figure S22. <sup>1</sup>H NMR of compound 19k

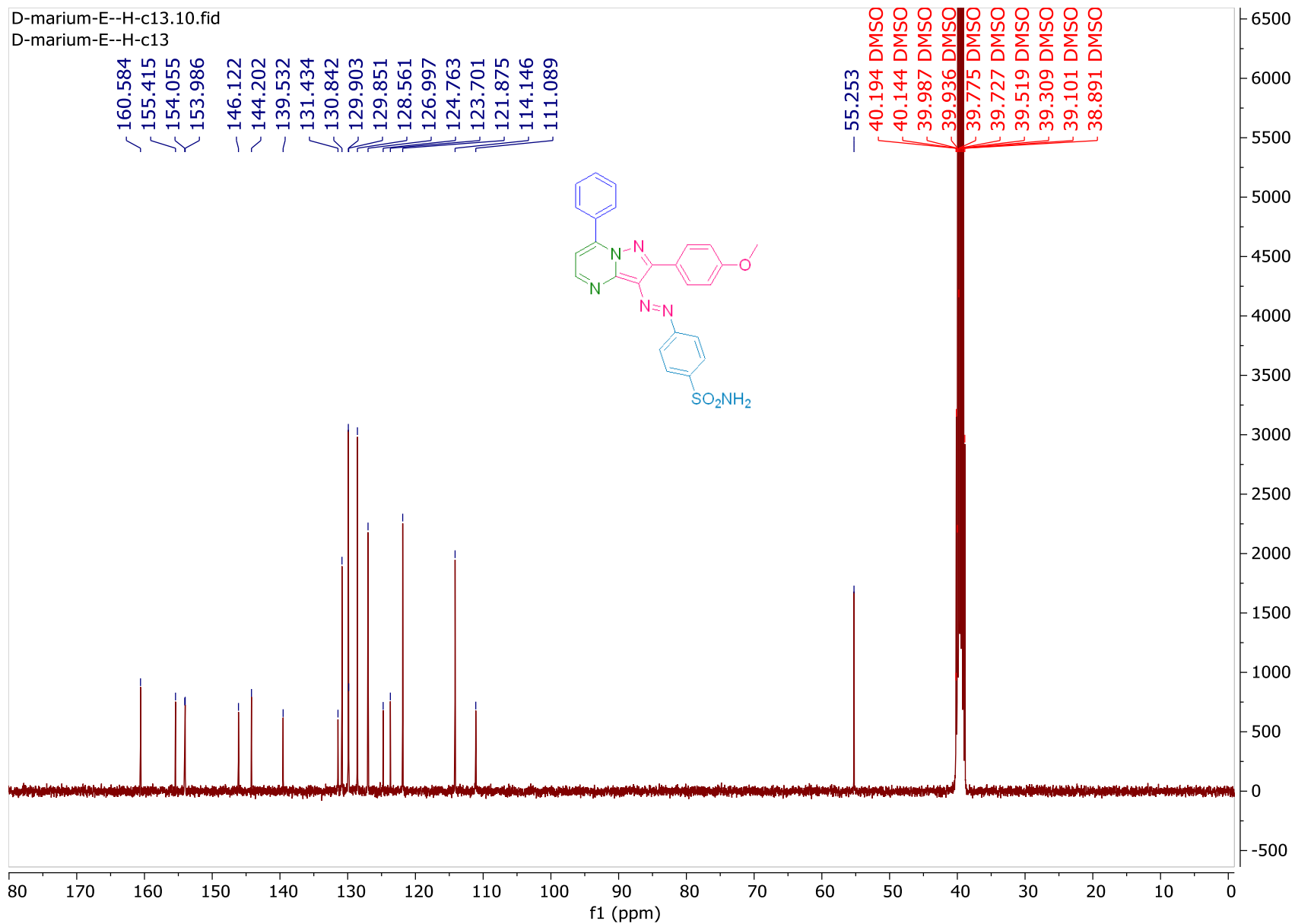


Figure S23.  $^{13}\text{C}$  NMR of compound 19k

D-mariam-E-CH3-1H.10.fid  
D-mariam-E-CH3-1H

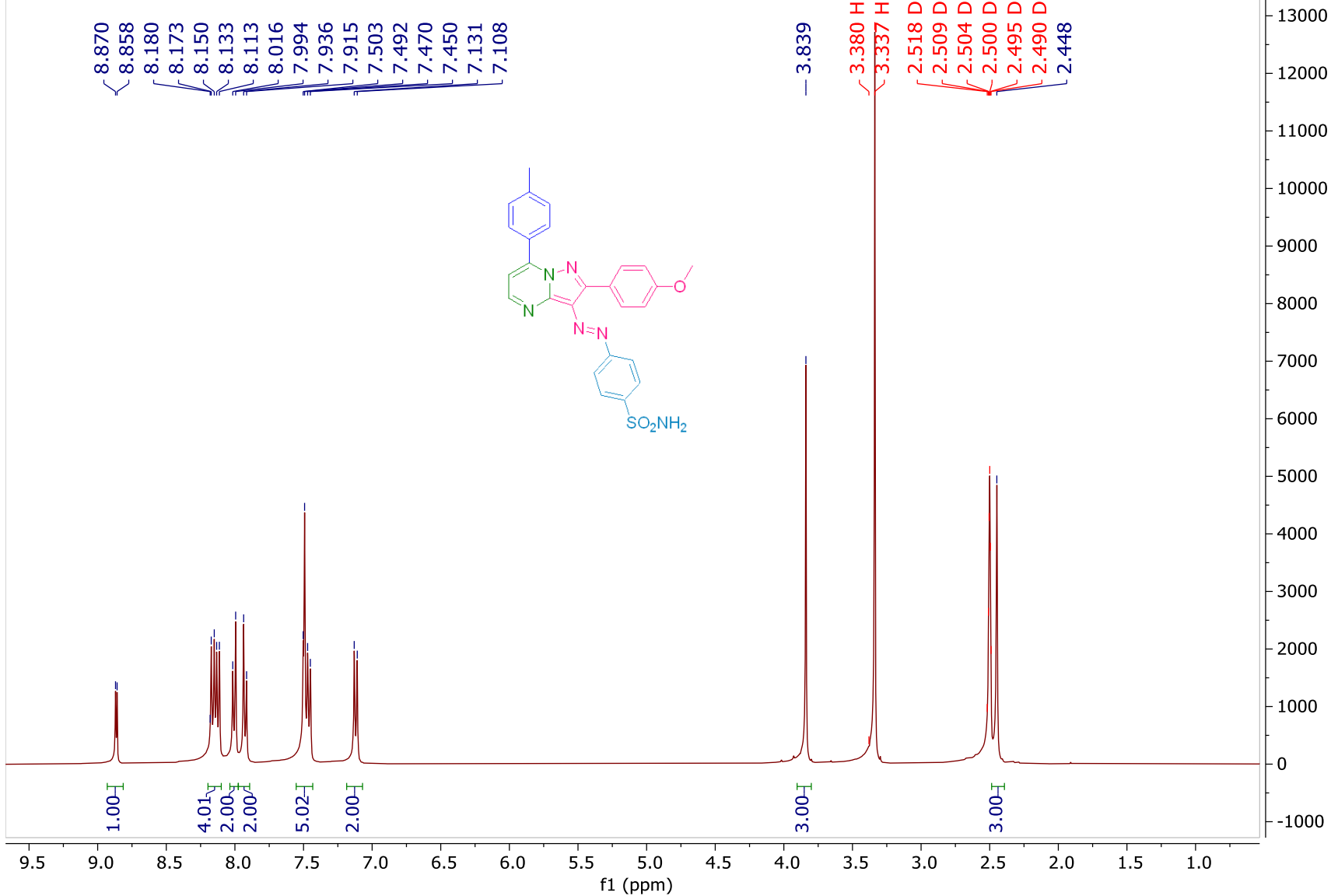


Figure S24. <sup>1</sup>H NMR of compound 191

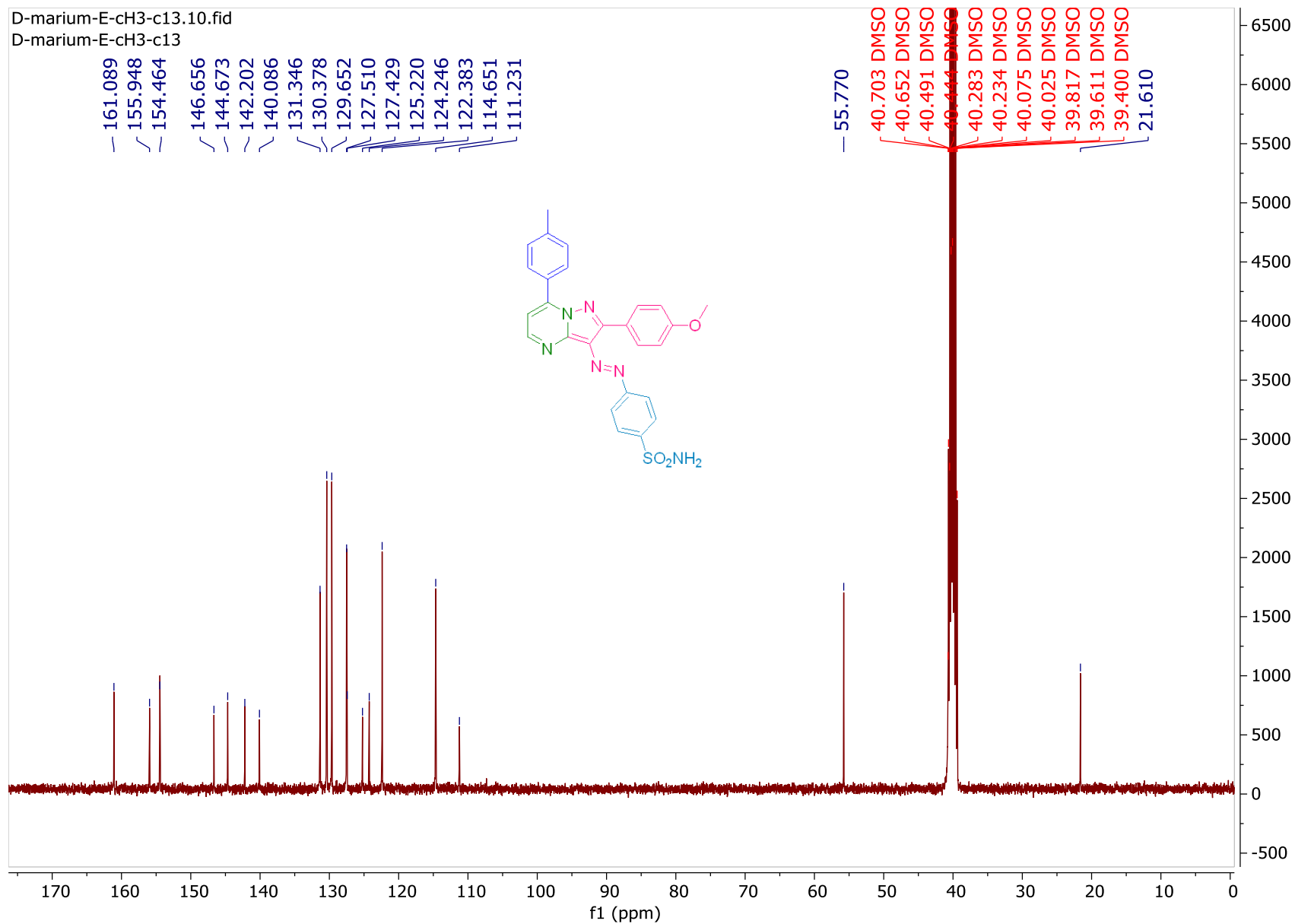


Figure S25. <sup>13</sup>C NMR of compound 19l

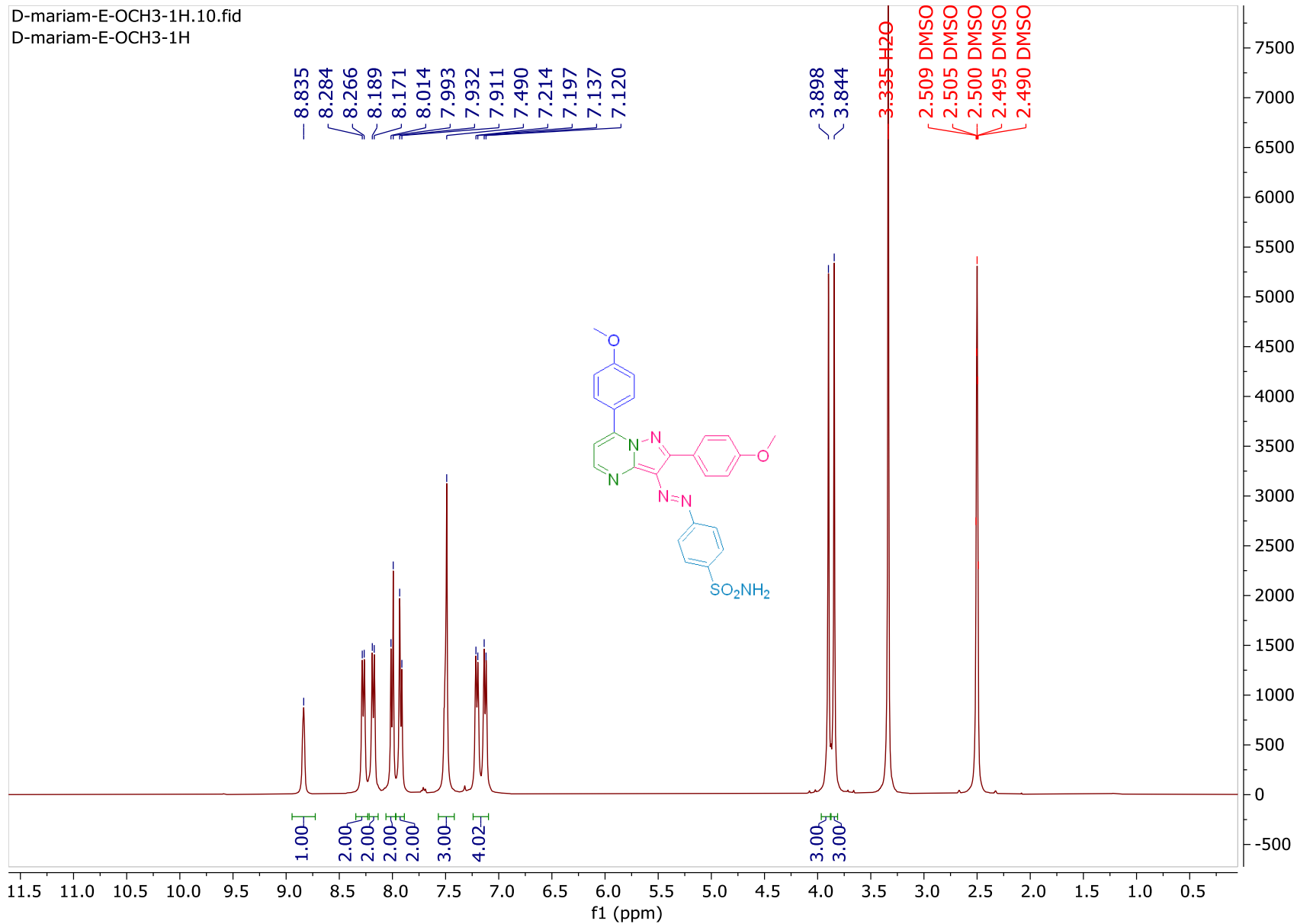


Figure S26. <sup>1</sup>H NMR of compound 19m

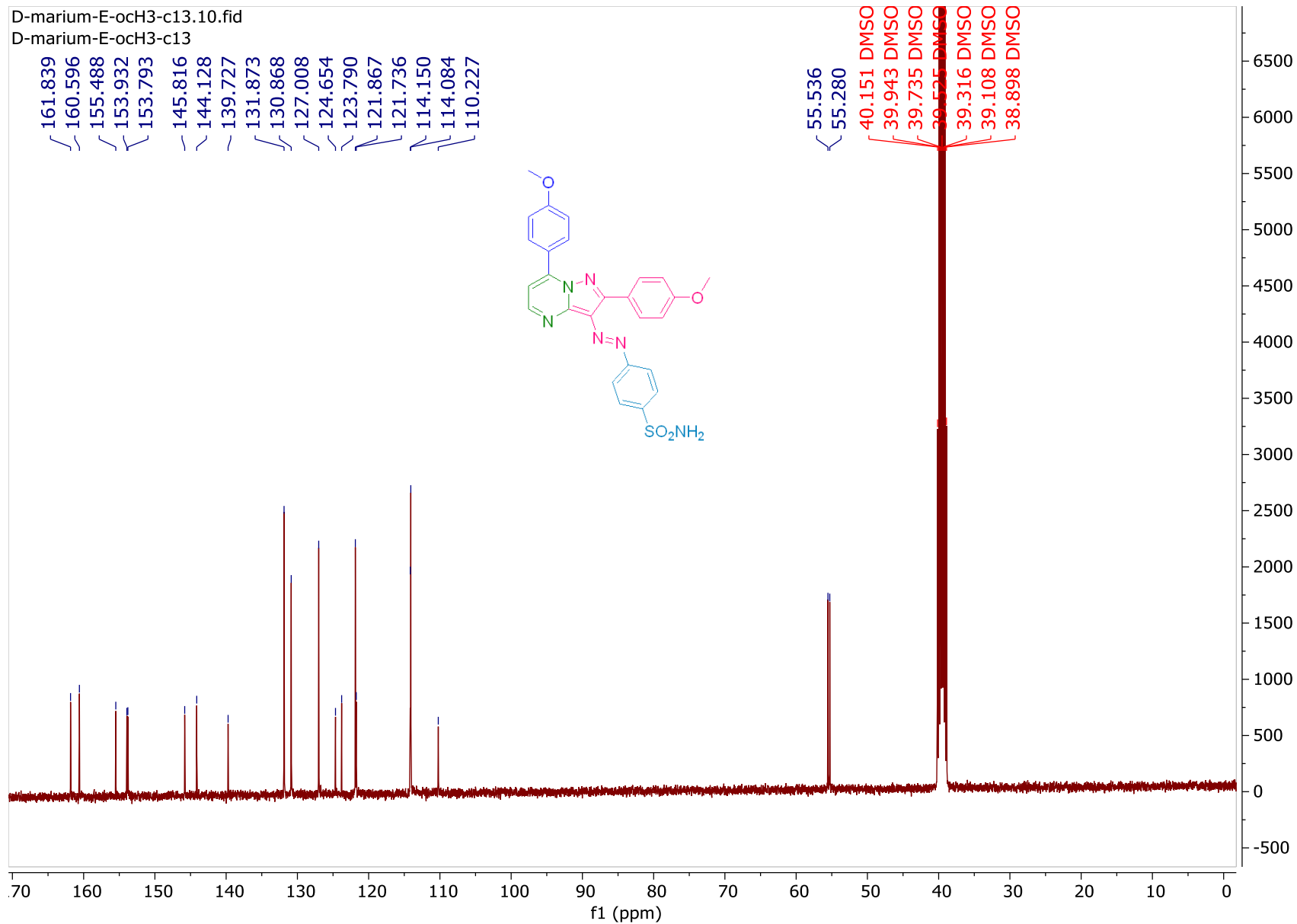


Figure S27. <sup>13</sup>C NMR of compound 19m

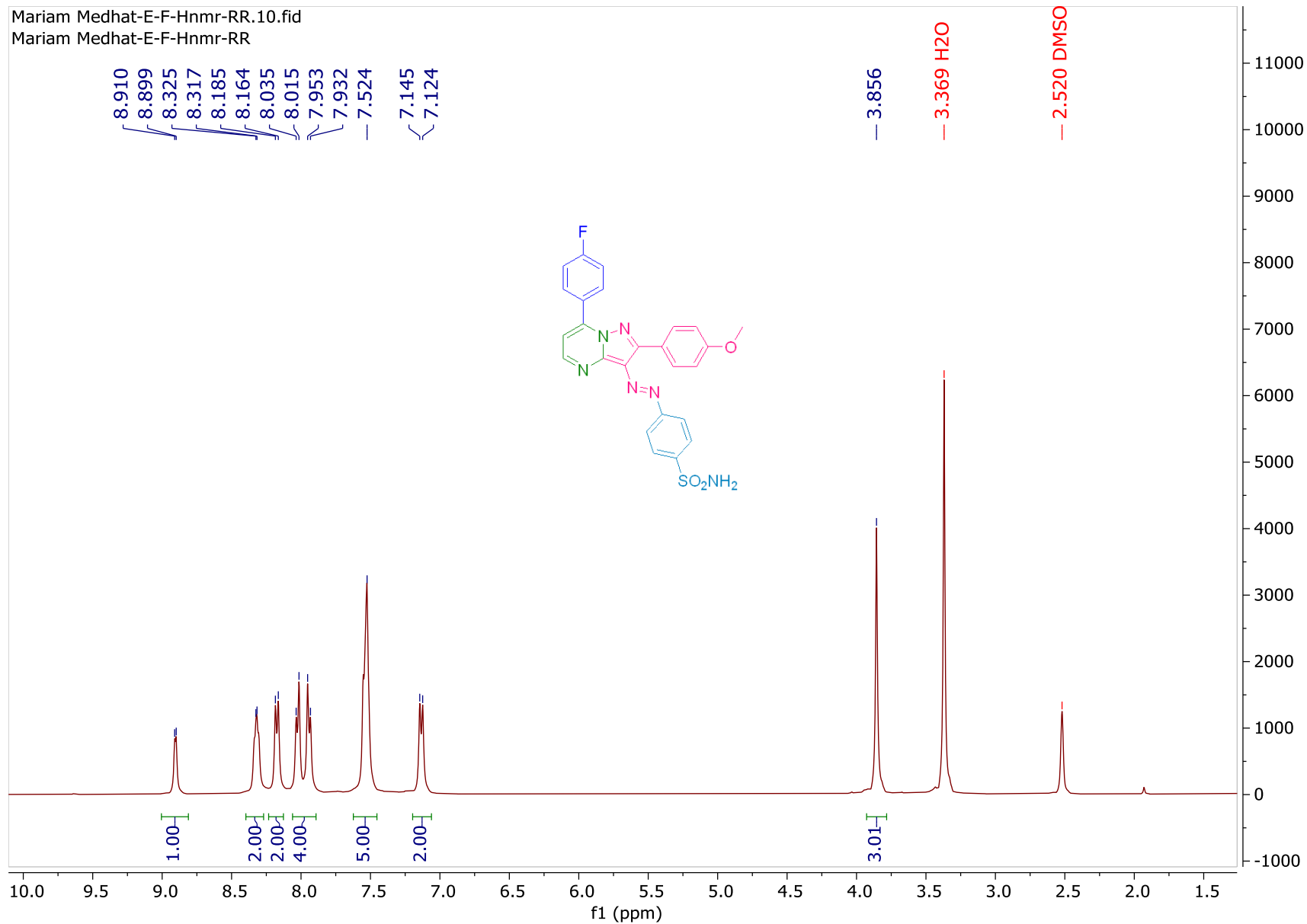


Figure S28. <sup>1</sup>H NMR of compound 19n

Mariam Medhat-E-F-C13-RR.10.fid  
Mariam Medhat-E-F-C13-RR

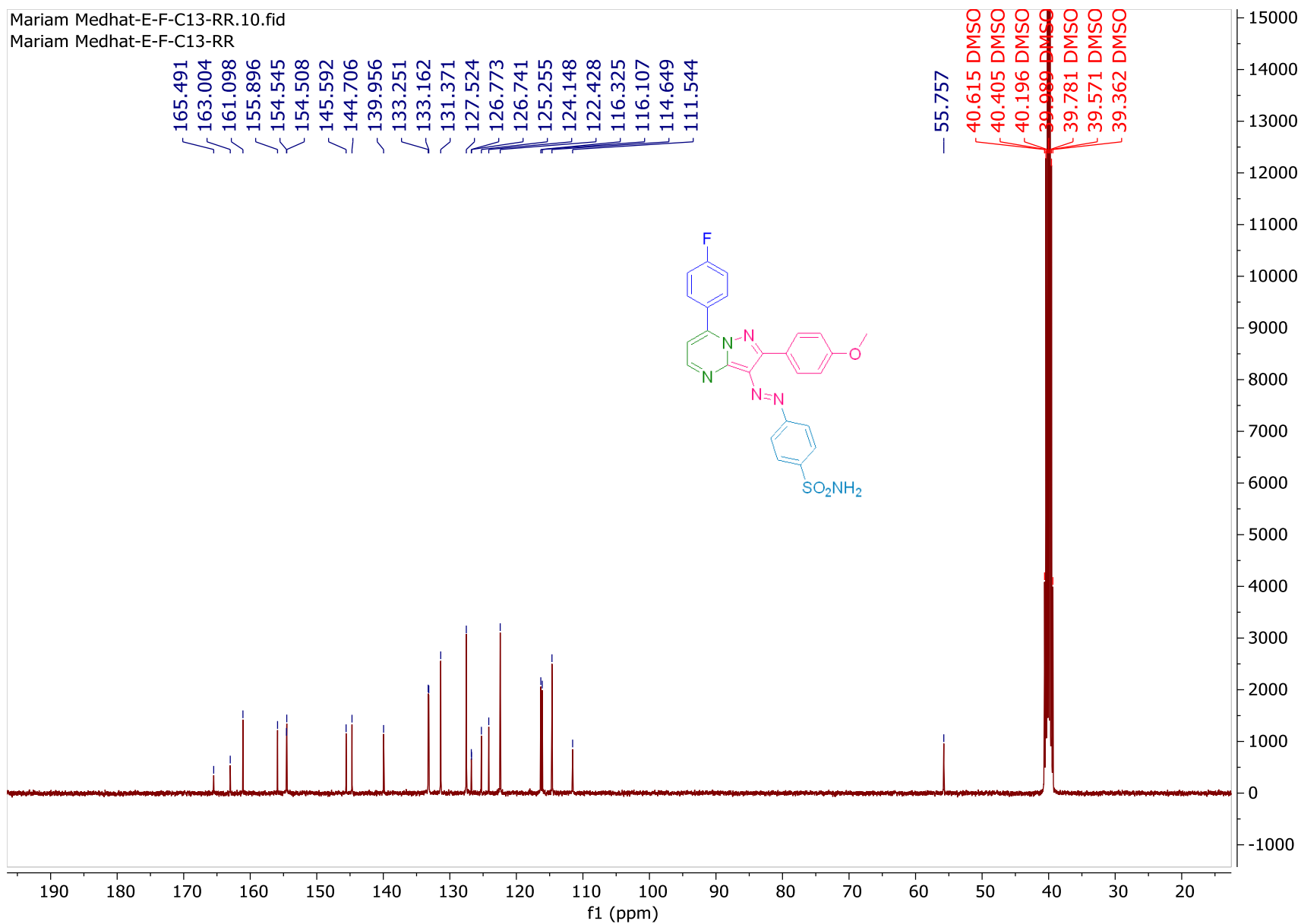


Figure S29.  $^{13}\text{C}$  NMR of compound 19n

Jul04-2024-Dr.Dina.90.fid

MARIAM-ECL

PROTON DMSO {C:\data\ Dr.Dina 6

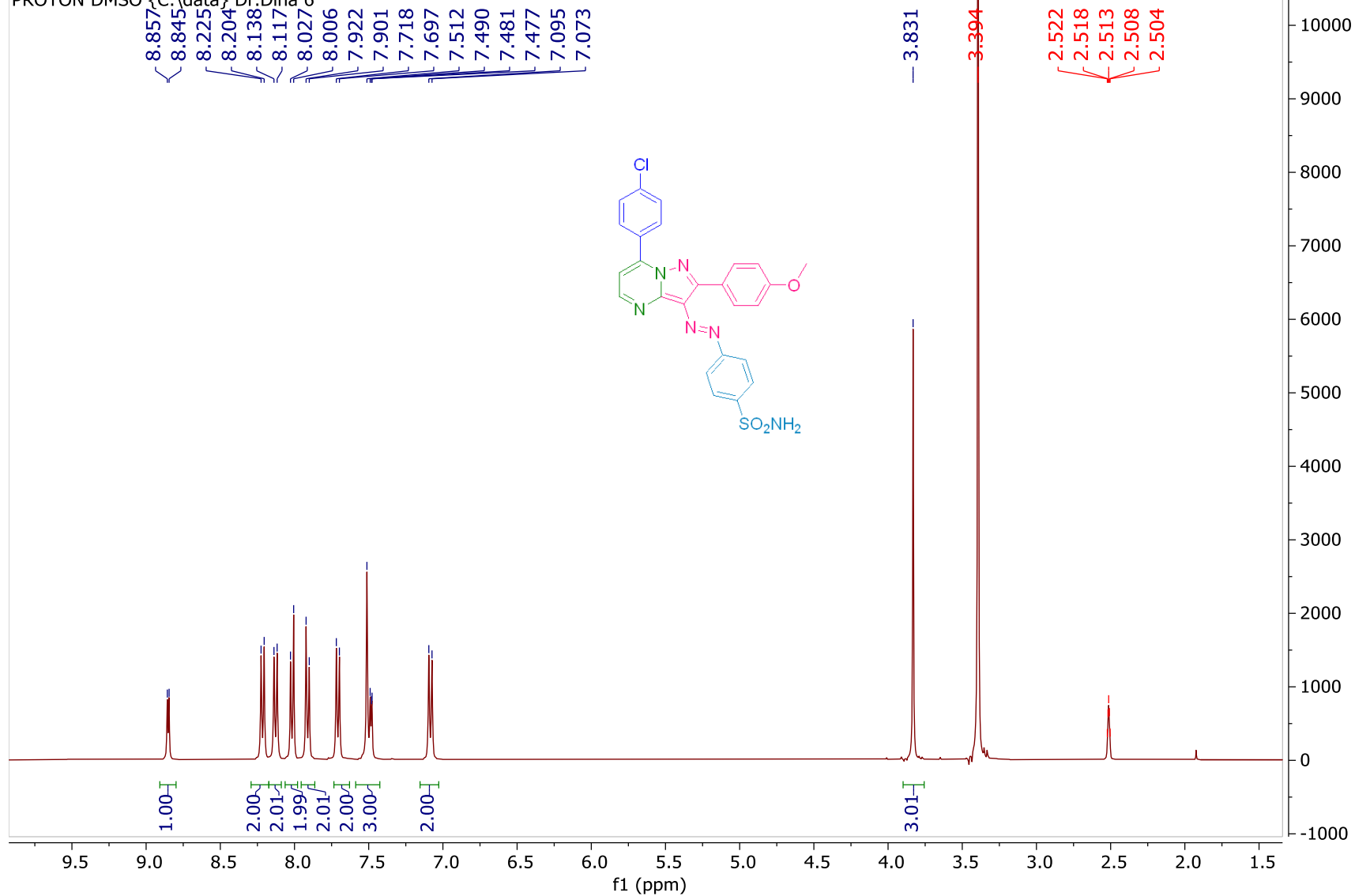


Figure S30. <sup>1</sup>H NMR of compound 19o

