

Supporting Information

Structure-activity relationship studies on highly functionalized pyrazole hydrazones and amides as antiproliferative and antioxidant agents

Matteo Lusardi ¹, Maria Grazia Signorello ¹, Eleonora Russo ¹, Debora Caviglia¹, Marco Ponassi ², Erika Iervasi ², Camillo Rosano ², Chiara Brullo ¹ and Andrea Spallarossa ^{1,*}

¹ Department of Pharmacy, University of Genova, viale Benedetto XV, 3, 16132, Genova, Italy; matteo.lusardi@edu.unige.it (M.L.); mariagrazia.signorello@unige.it (M.G.S); chiara.brullo@unige.it (C.B.); eleonora.russo@unige.it (E.R.); debora.caviglia@edu.unige.it (D.C.).

² Proteomics and Mass Spectrometry Unit, IRCCS Ospedale Policlinico San Martino, Largo R. Benzi 10, 16132, Genova, Italy; erika.iervasi@hsanmartino.it (E.I.); camillo.rosano@hsanmartino.it (C.R.); marco.ponassi@hsanmartino.it (M.P.).

*Correspondence: andrea.spallarossa@unige.it

Table of contents

Figure S1. ¹H-NMR (400 MHz, d₆-DMSO) spectrum of compound **10a**

Figure S2. ¹³C-NMR (101 MHz, d₆-DMSO) spectrum of compound **10a**

Figure S3. ¹H-NMR (400 MHz, d₆-DMSO) spectrum of compound **10b**

Figure S4. ¹³C-NMR (101 MHz, d₆-DMSO) spectrum of compound **10b**

Figure S5. ¹H-NMR (400 MHz, d₆-DMSO) spectrum of compound **11a**

Figure S6. ¹³C-NMR (101 MHz, d₆-DMSO) spectrum of compound **11a**

Figure S7. ¹H-NMR (400 MHz, d₆-DMSO) spectrum of compound **11b**

Figure S8. ¹³C-NMR (101 MHz, d₆-DMSO) spectrum of compound **11b**

Figure S9. ¹H-NMR (400 MHz, d₆-DMSO) spectrum of compound **11c**

Figure S10. ¹³C-NMR (101 MHz, d₆-DMSO) spectrum of compound **11c**

Figure S11. ¹H-NMR (400 MHz, d₆-DMSO) spectrum of compound **11d**

Figure S12. ¹³C-NMR (101 MHz, d₆-DMSO) spectrum of compound **11d**

Figure S13. ¹H-NMR (400 MHz, d₆-DMSO) spectrum of compound **12a**

Figure S14. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **12a**

Figure S15. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **12b**

Figure S16. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **12b**

Figure S17. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **12c**

Figure S18. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **12c**

Figure S19. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **12d**

Figure S20. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **12d**

Figure S21. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **13a**

Figure S22. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **13a**

Figure S23. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **13b**

Figure S24. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **13b**

Figure S25. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **13c**

Figure S26. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **13c**

Figure S27. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **13d**

Figure S28. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **13d**

Figure S29. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **14**

Figure S30. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **15**

Figure S31. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **15**

Figure S32. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **16**

Figure S33. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **16**

Figure S34. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **17**

Figure S35. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **18**

Figure S36. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **18**

Figure S37. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **19**

Figure S38. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **19**

Figure S39. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **20**

Figure S40. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **20**

Figure S41. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **21**

Figure S42. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **21**

Figure S43. ^1H -NMR (400 MHz, d₆-DMSO) spectrum of compound **22**

Figure S44. ^{13}C -NMR (101 MHz, d₆-DMSO) spectrum of compound **22**

Table S1. Inhibitory effect of compounds **10-22** on platelet aggregation and ROS production

Figure S1. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **10a**

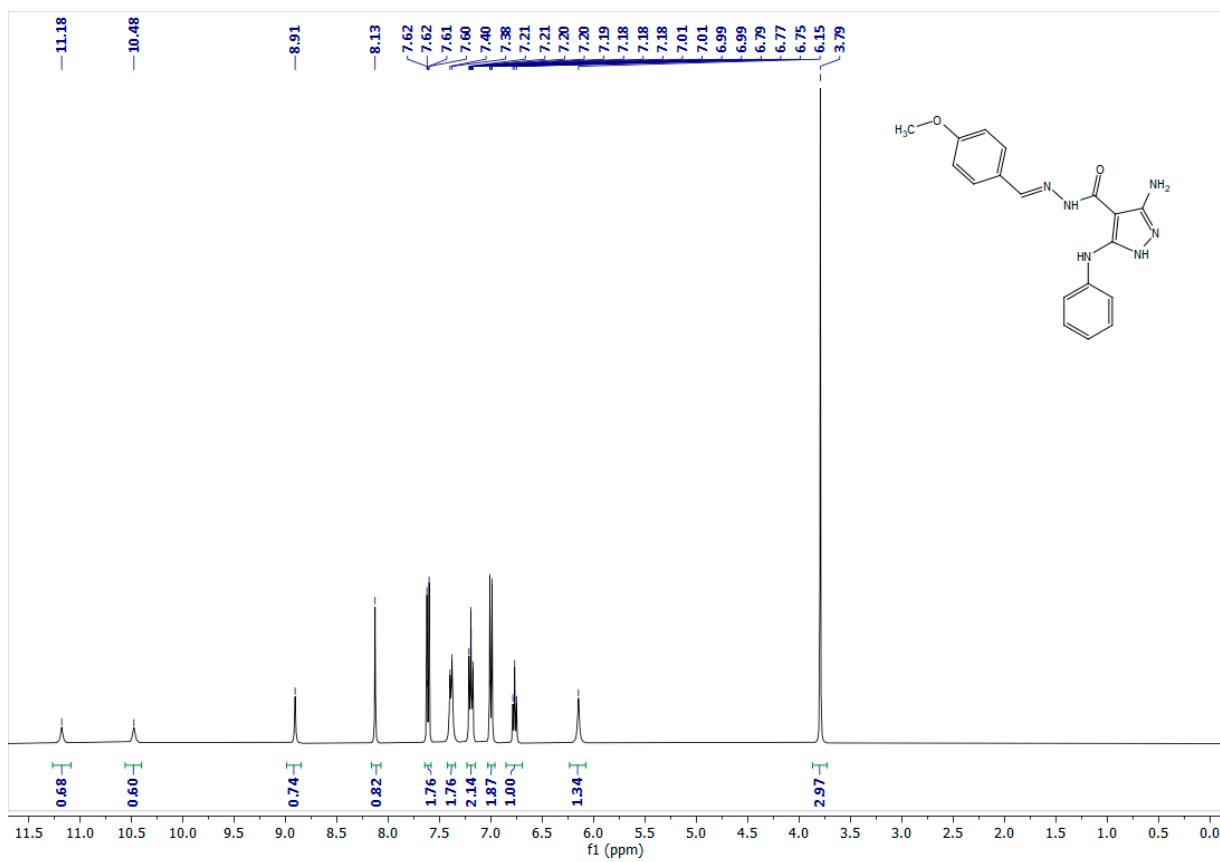


Figure S2. ^{13}C -NMR (101 MHz, $\text{d}_6\text{-DMSO}$) spectrum of compound **10a**

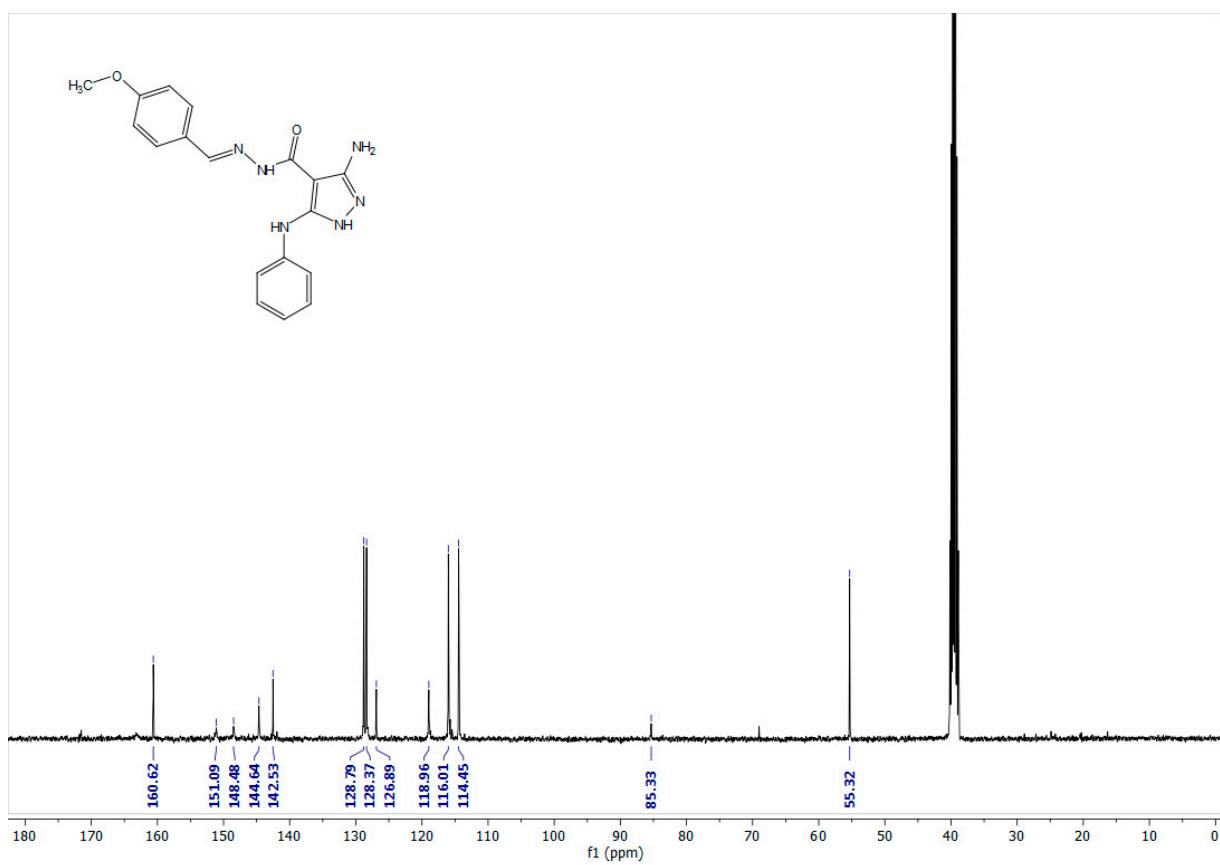


Figure S3. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **10b**

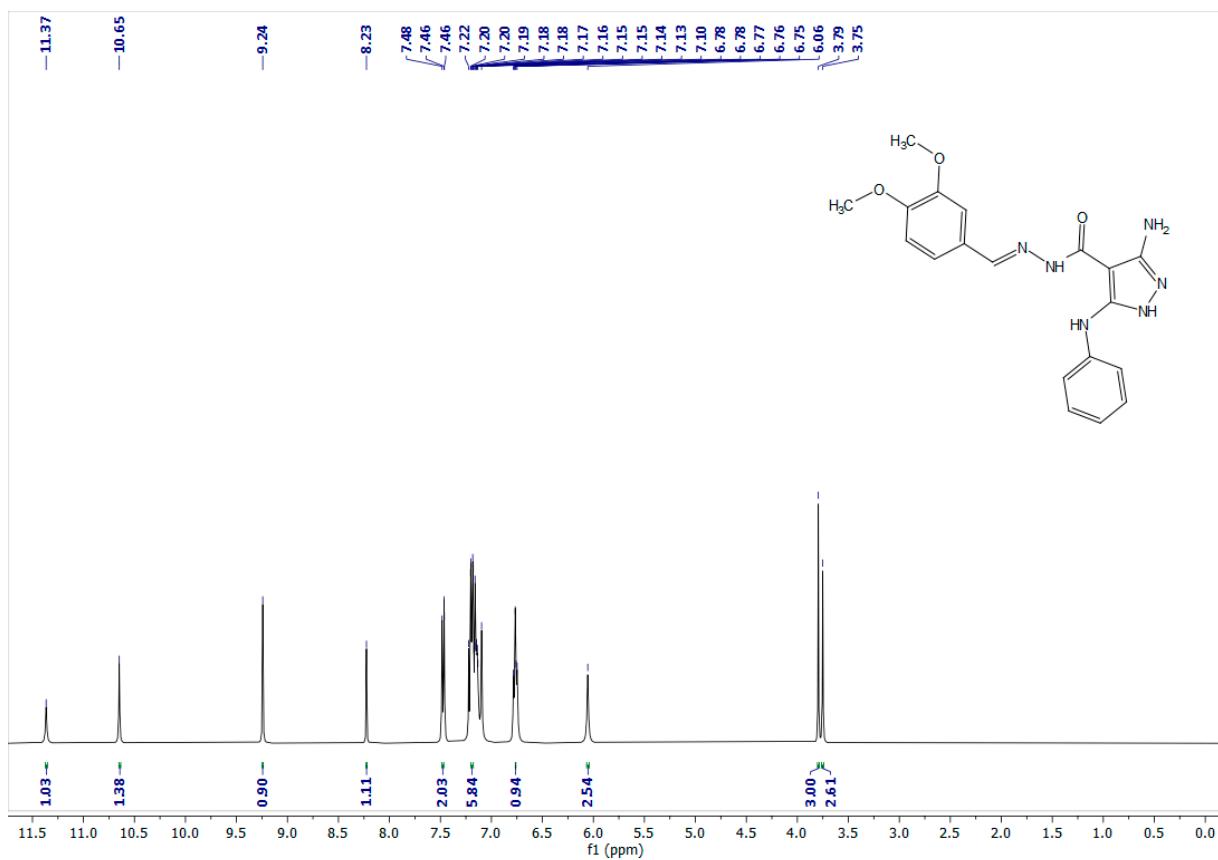


Figure S4. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **10b**

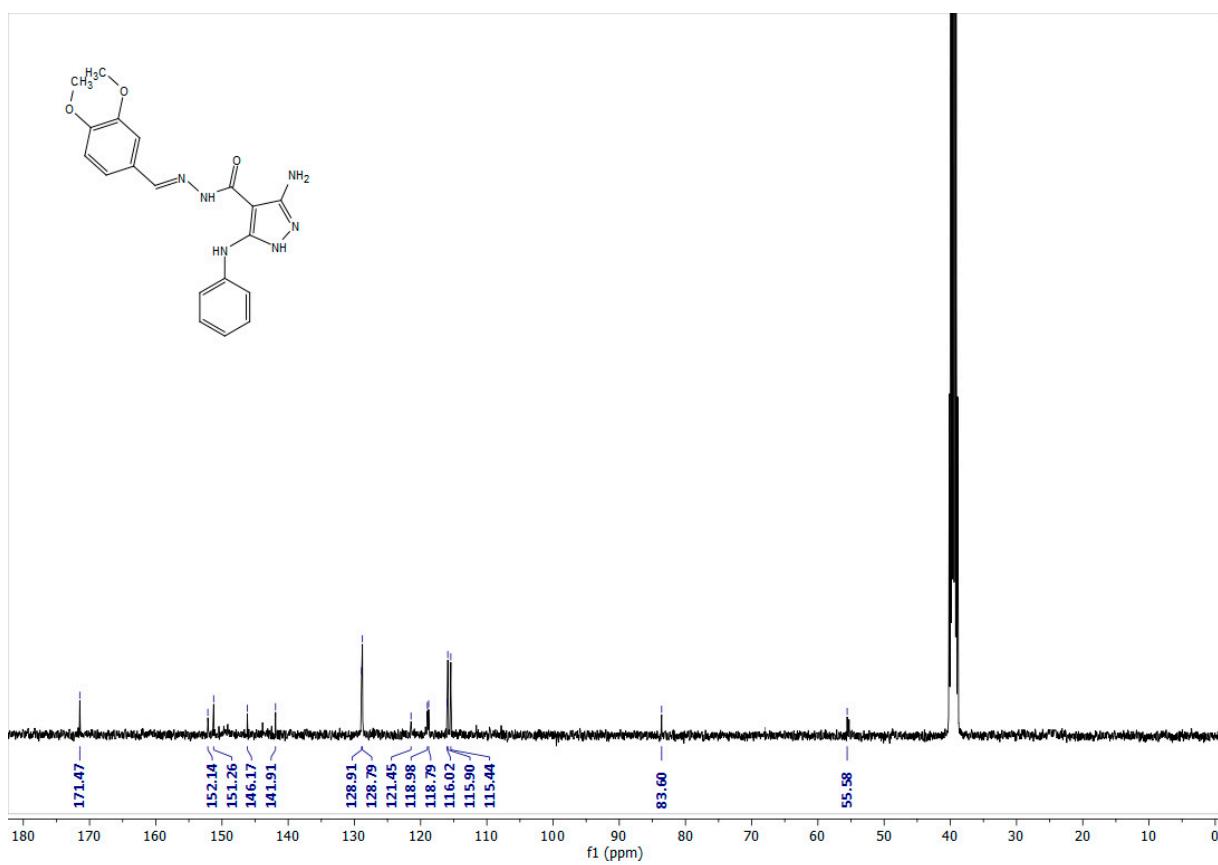


Figure S5. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **11a**

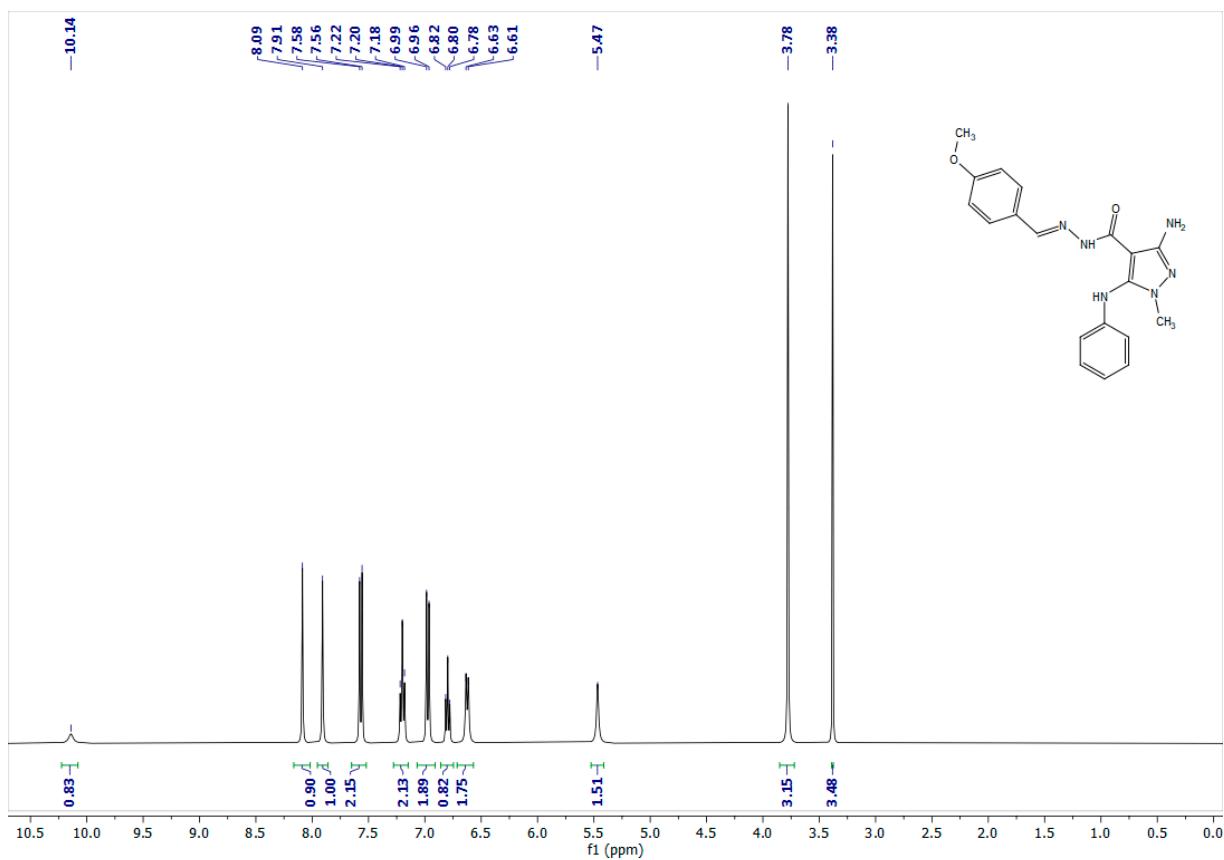


Figure S6. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **11a**

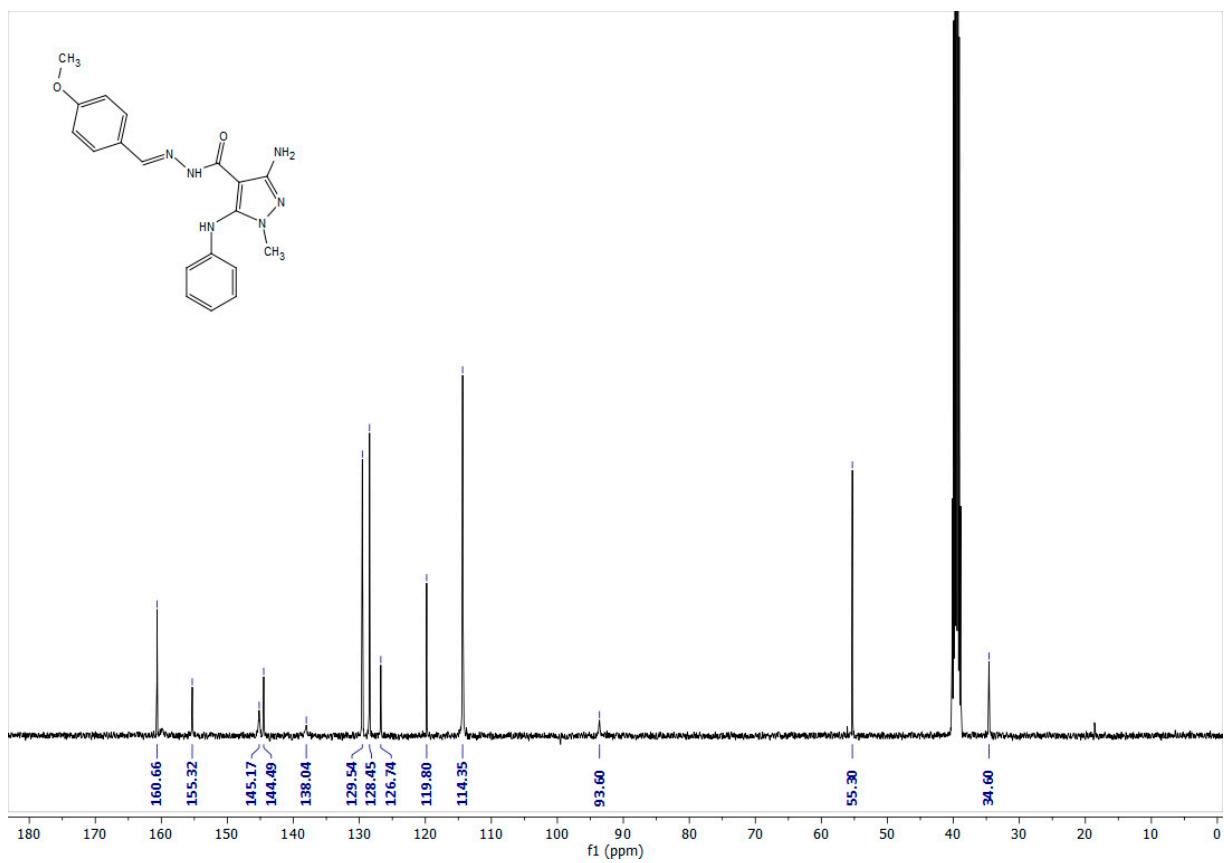


Figure S7. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **11b**

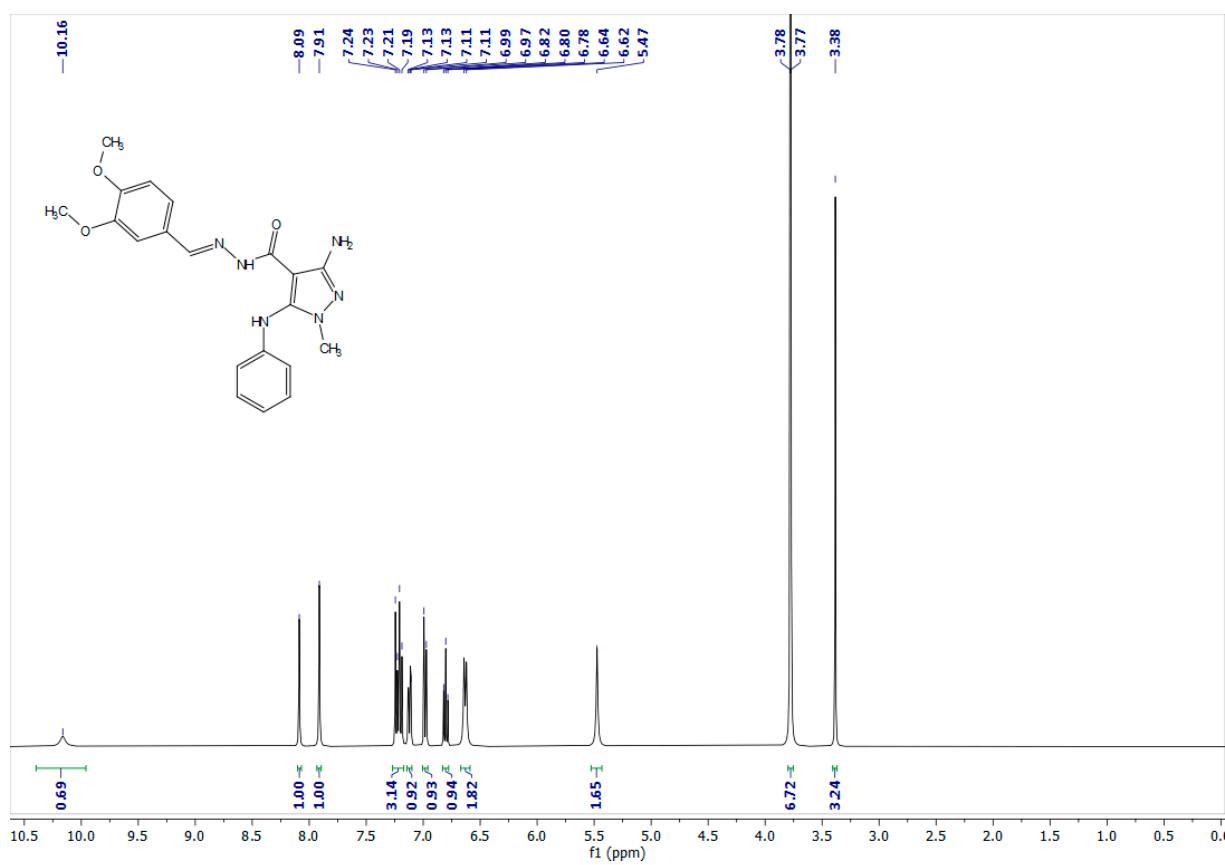


Figure S8. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **11b**

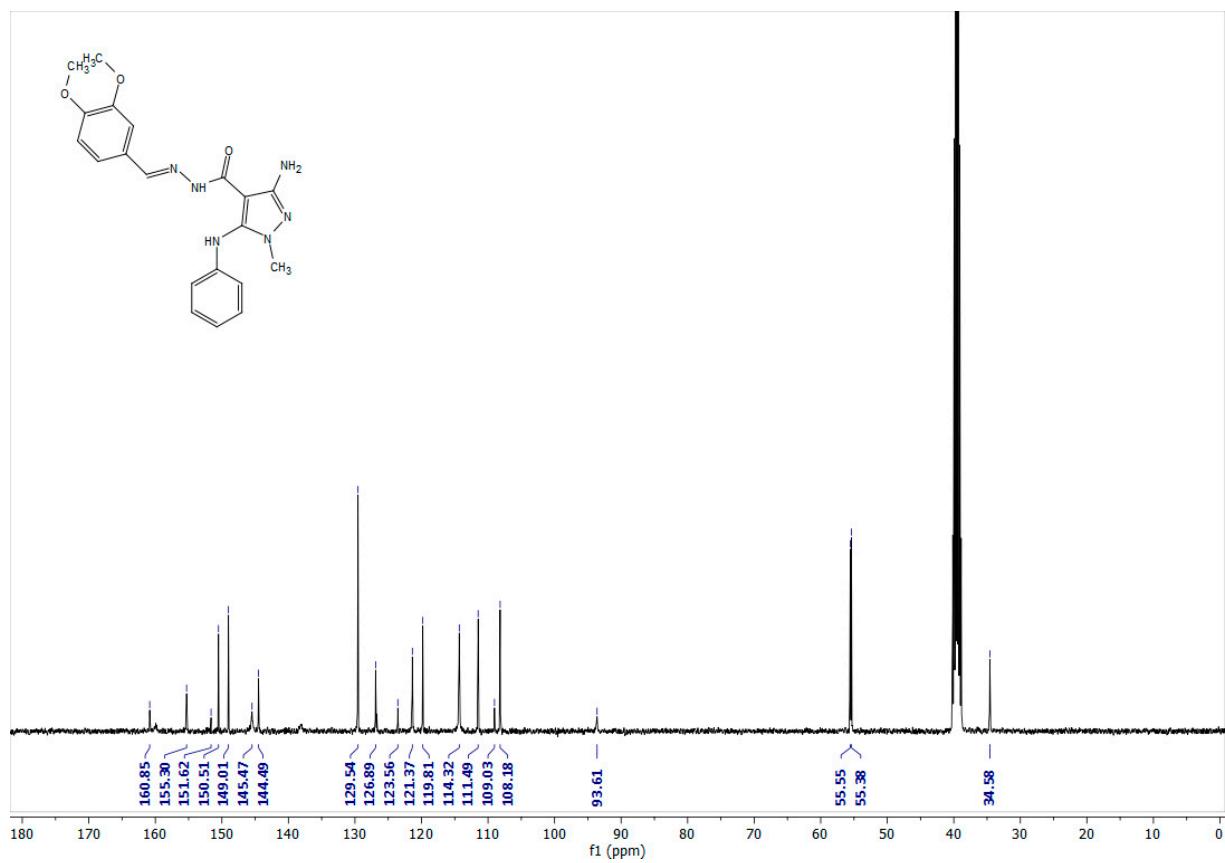


Figure S9. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **11c**

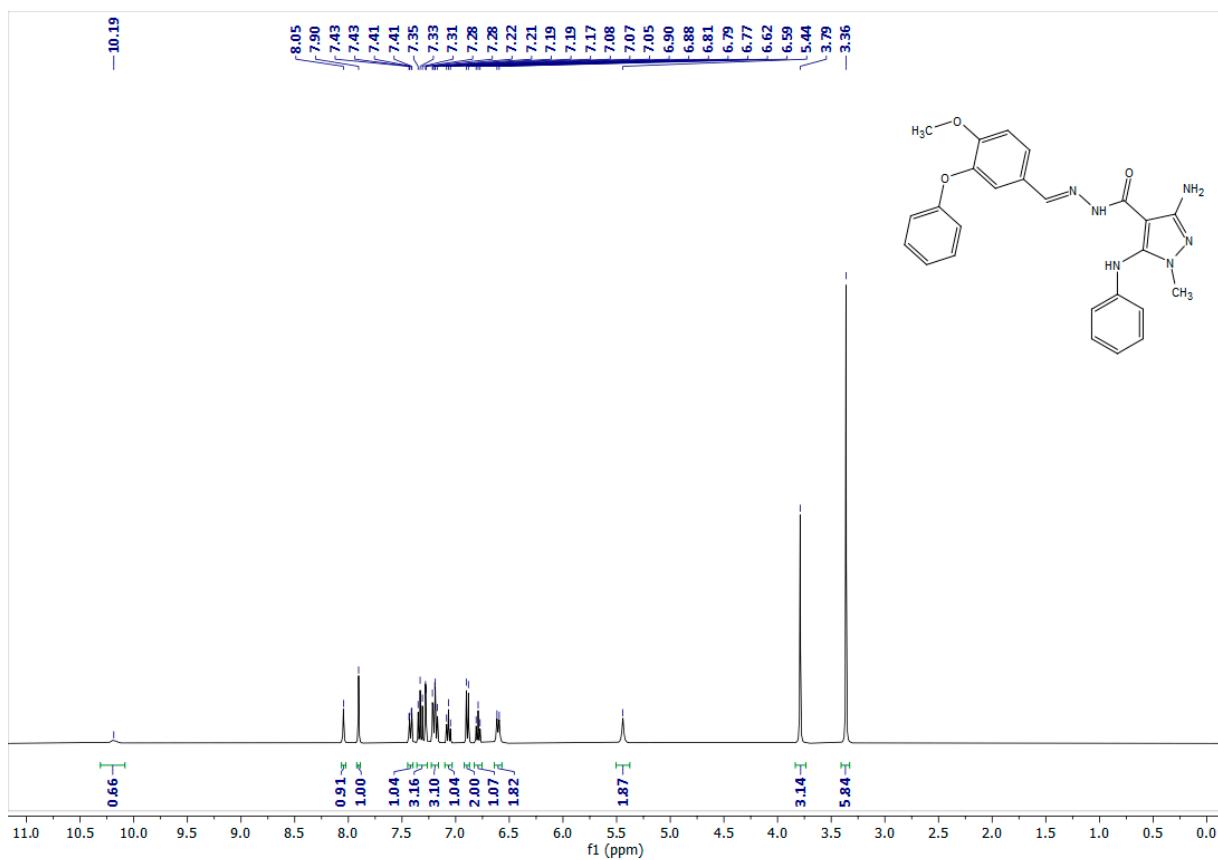


Figure S10. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **11c**

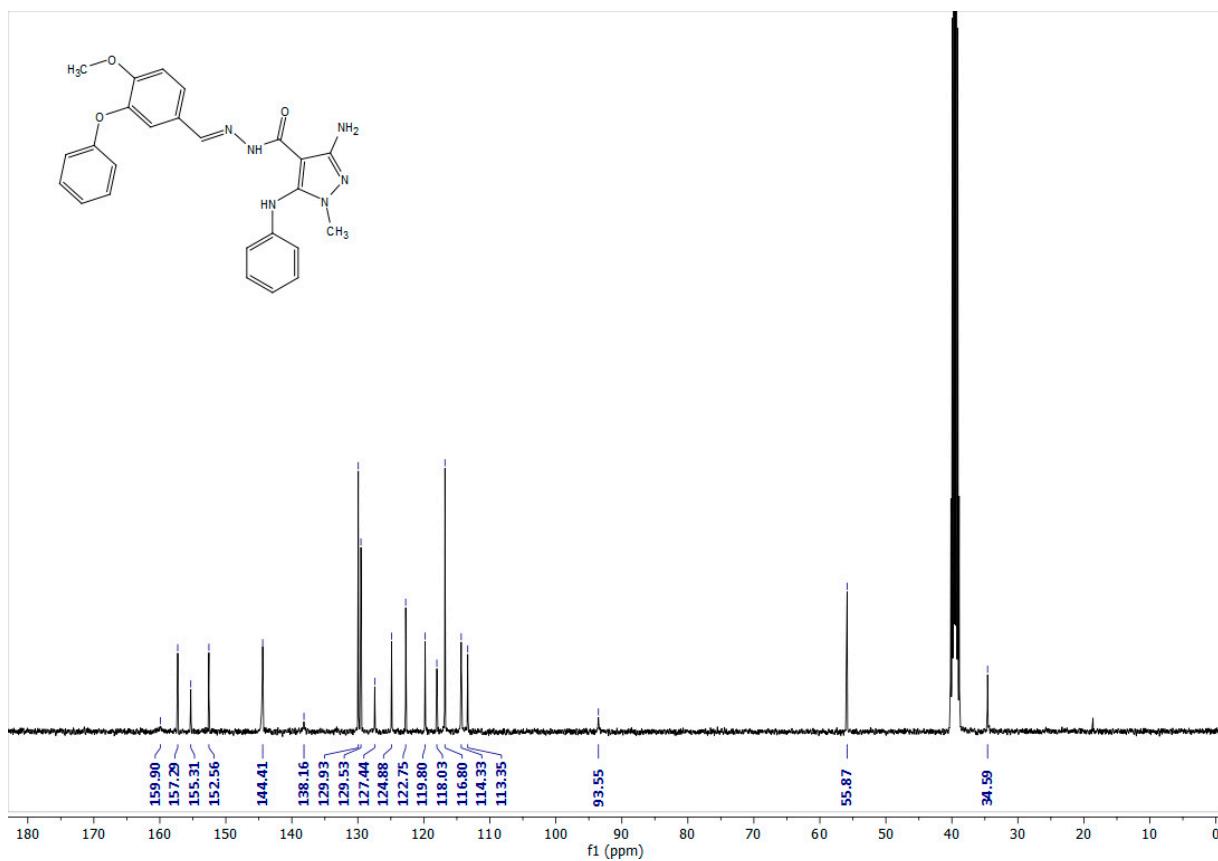


Figure S11. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **11d**

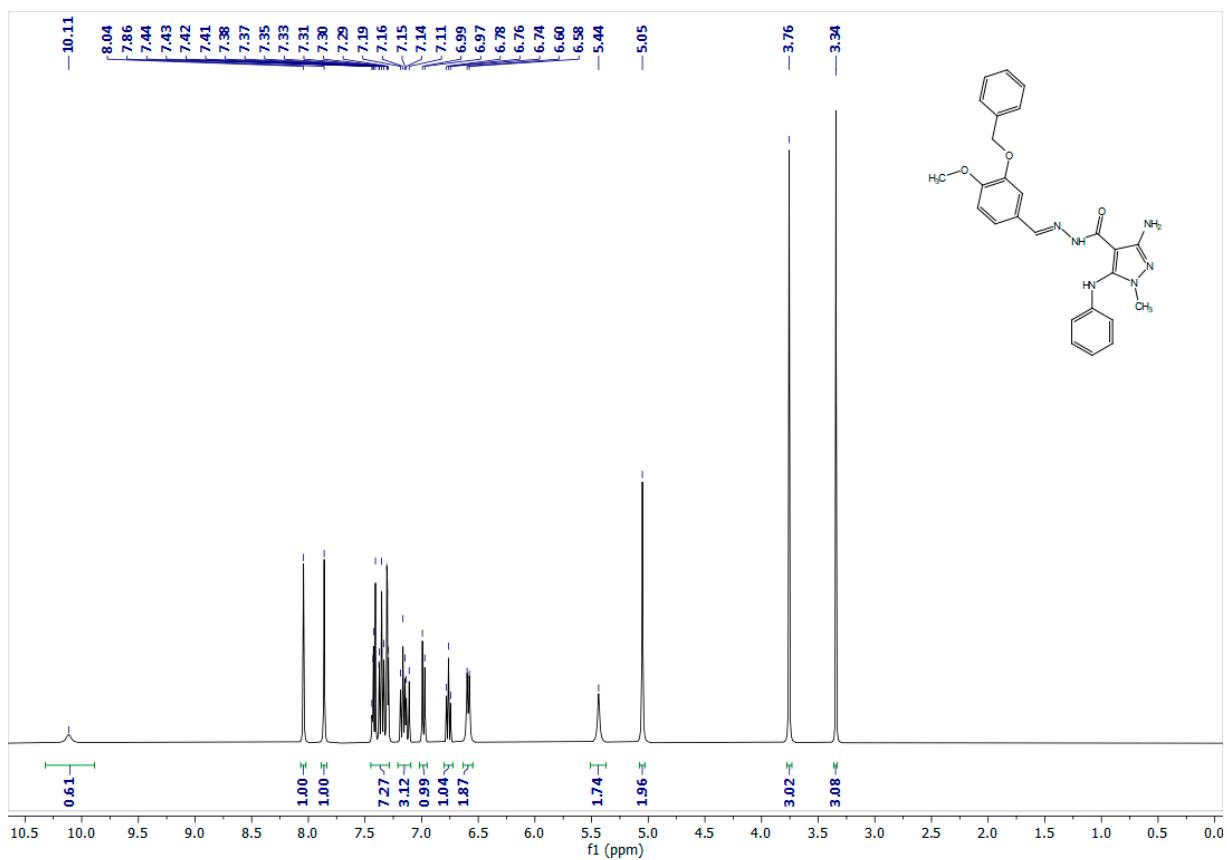


Figure S12. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **11d**

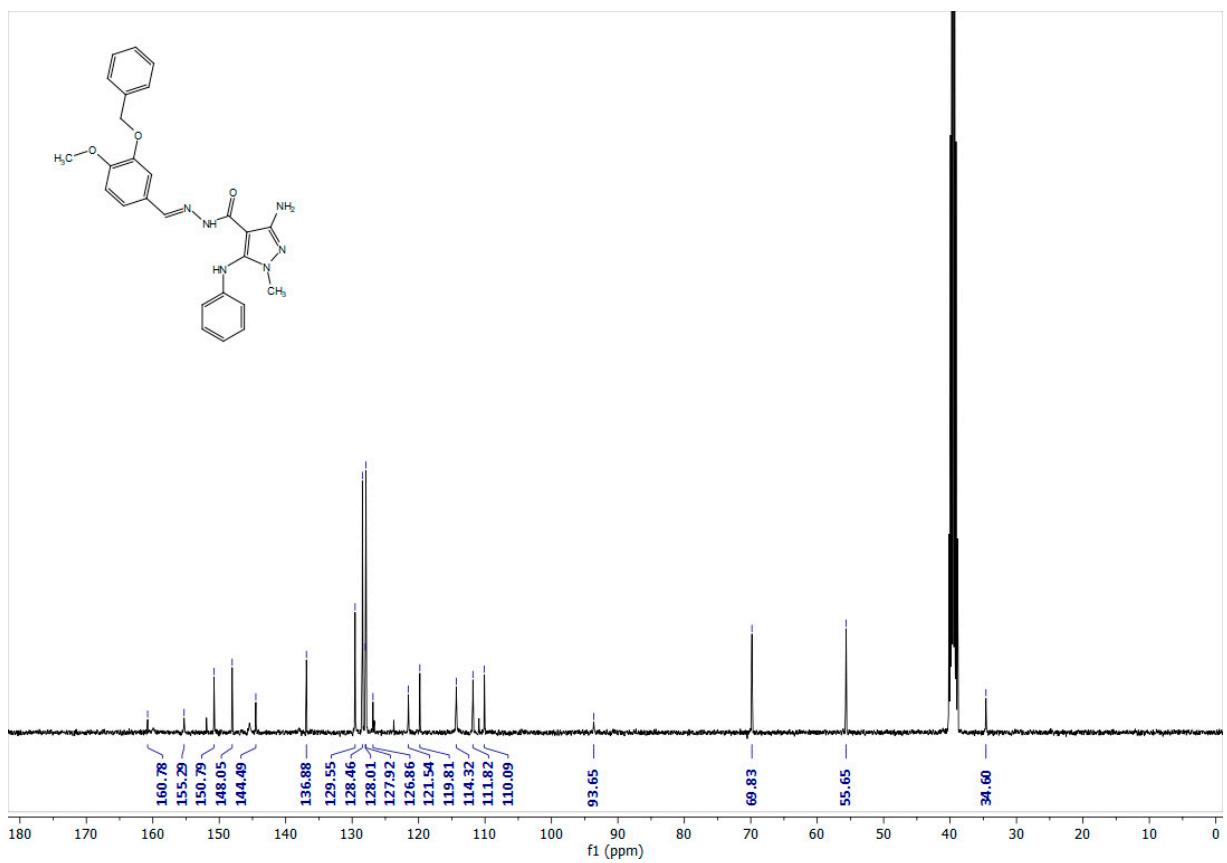


Figure S13. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **12a**

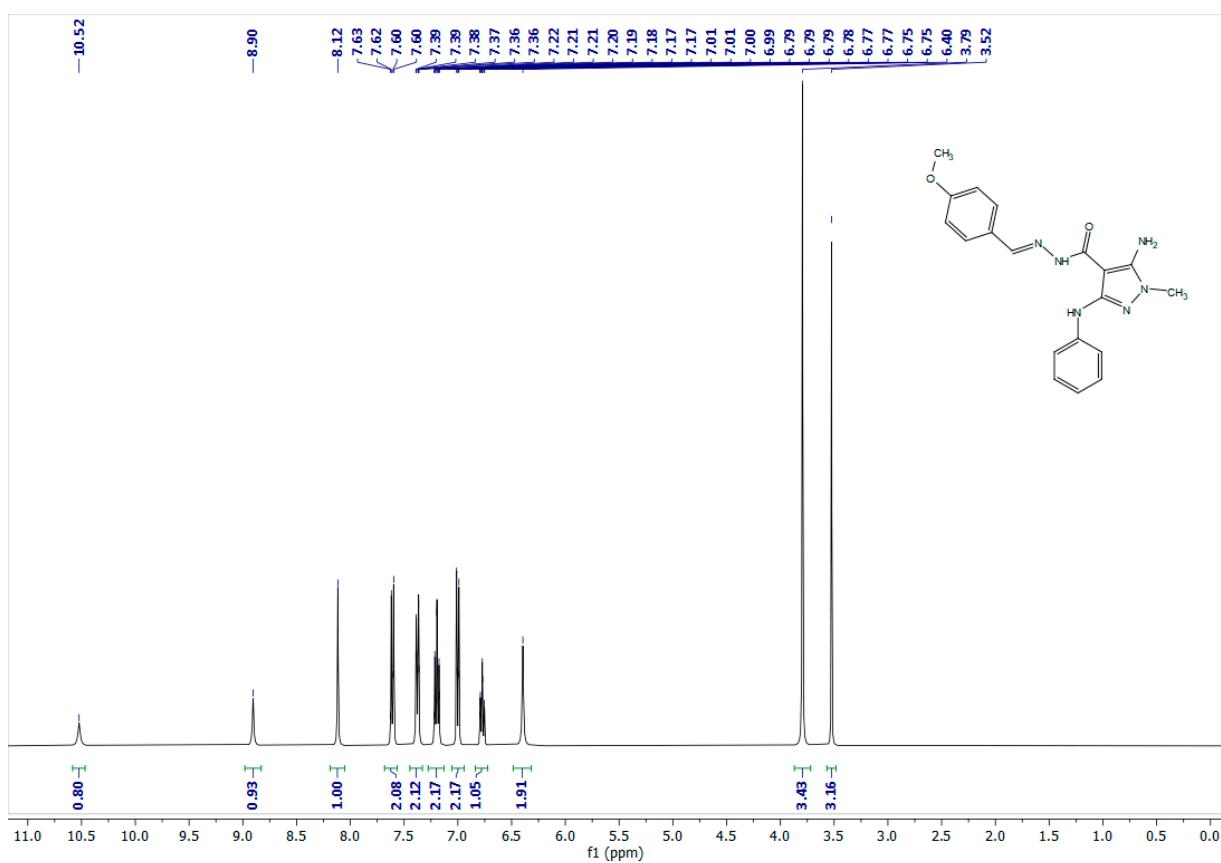


Figure S14. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **12a**

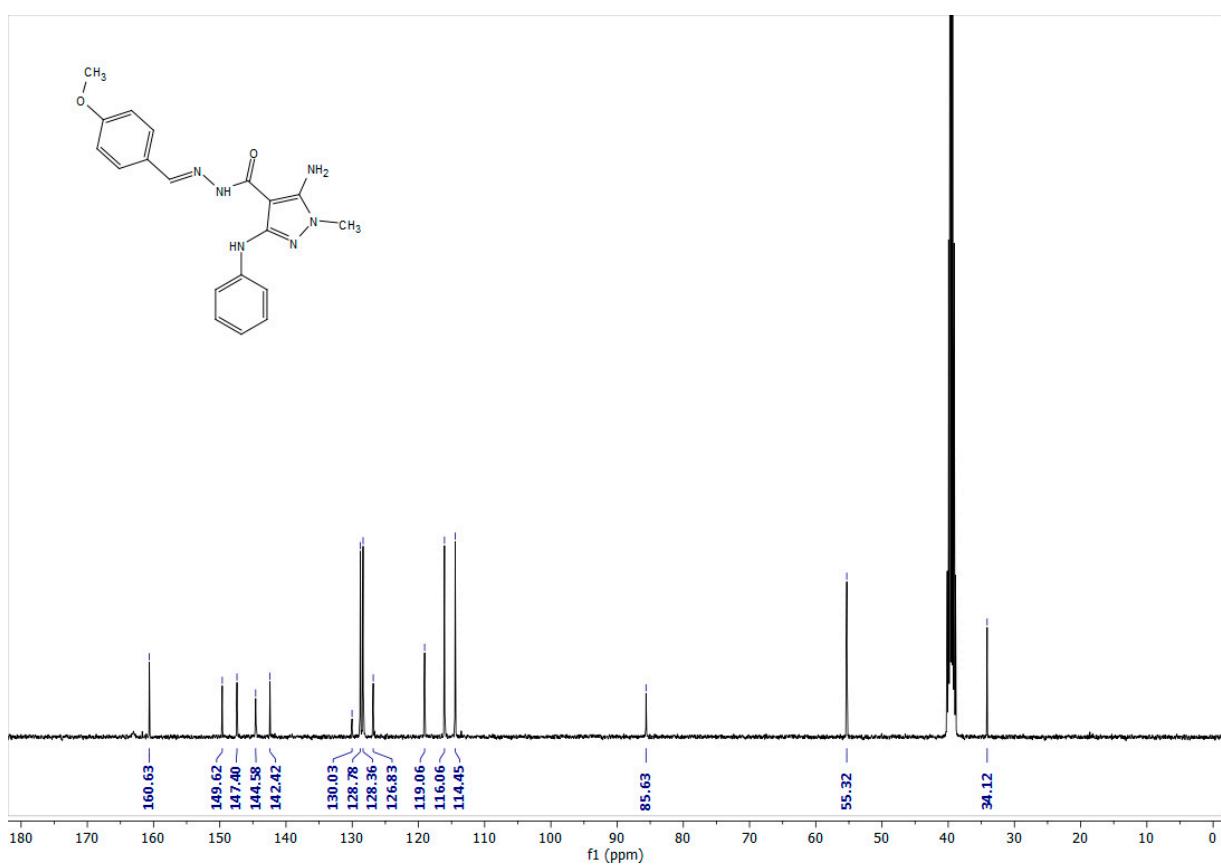


Figure S15. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **12b**

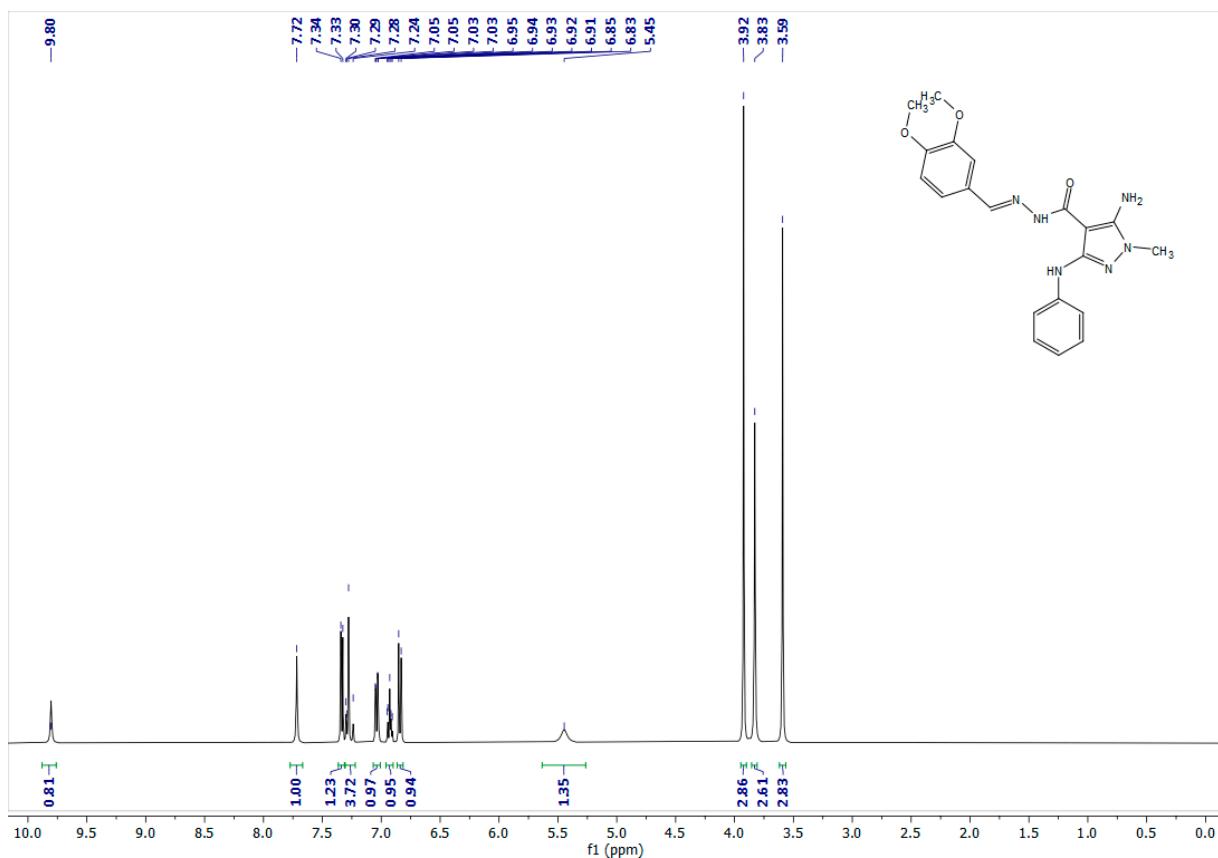


Figure S16. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **12b**

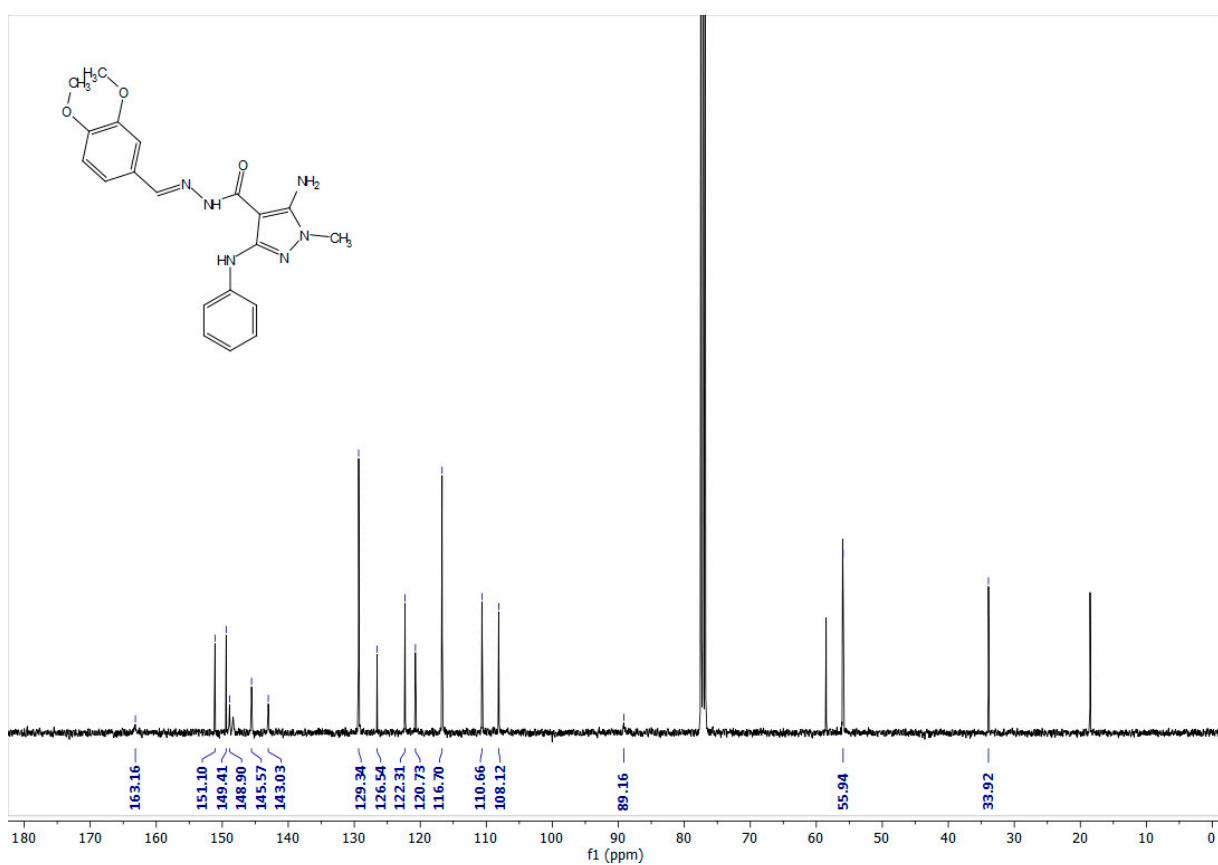


Figure S17. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **12c**

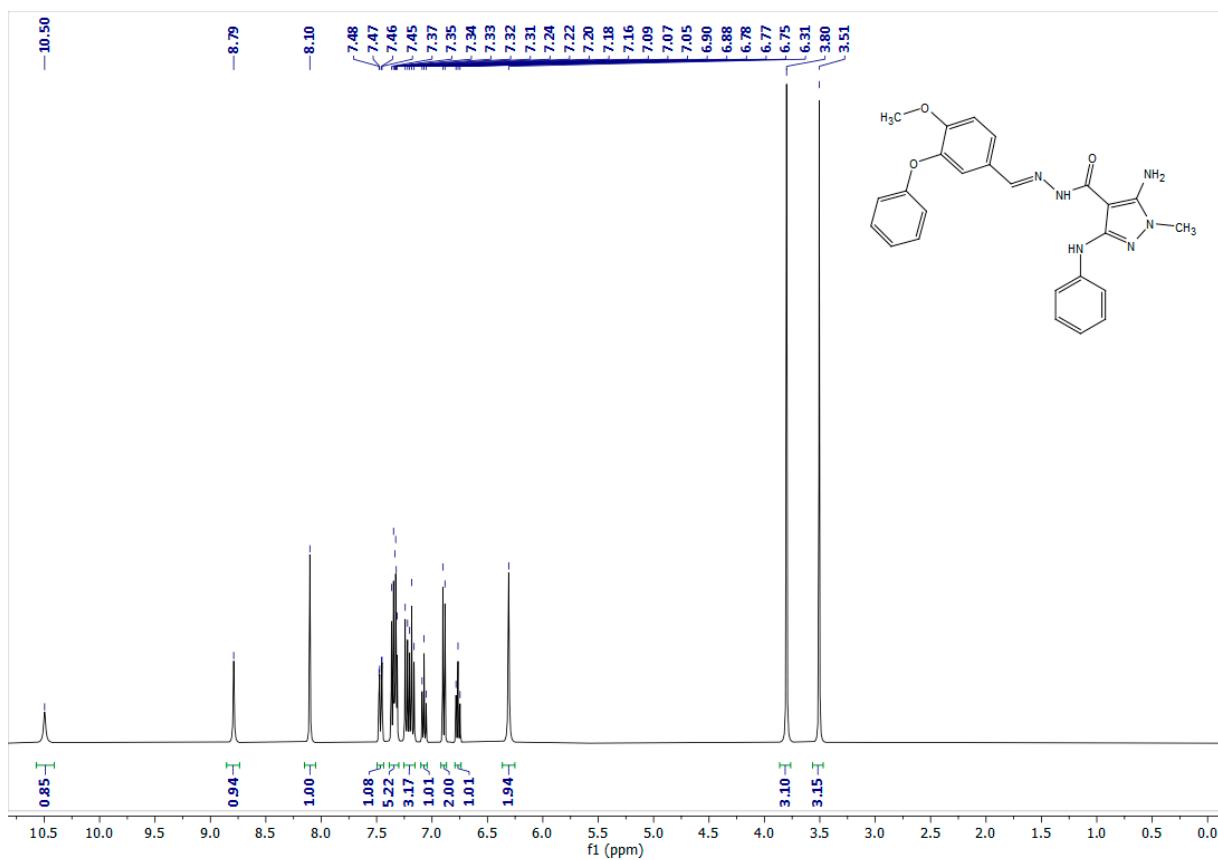


Figure S18. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **12c**

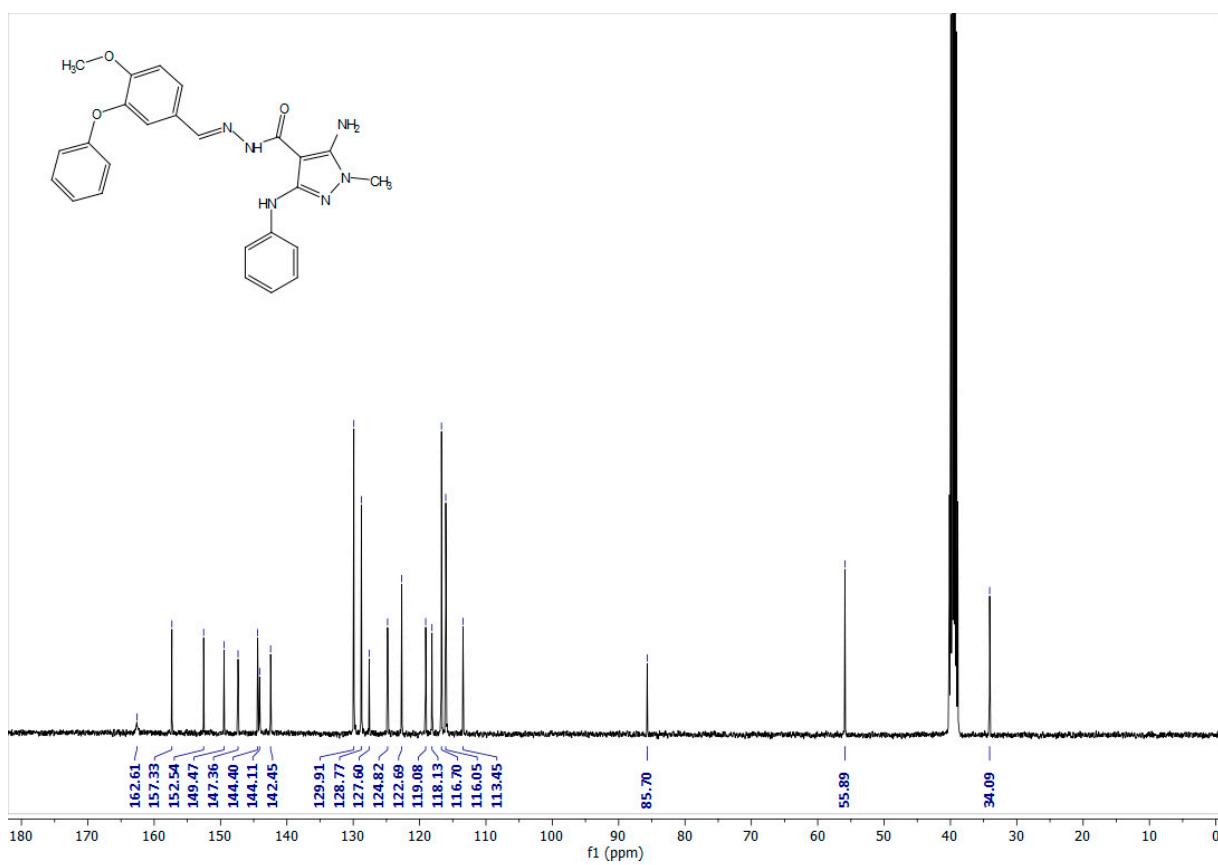


Figure S19. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **12d**

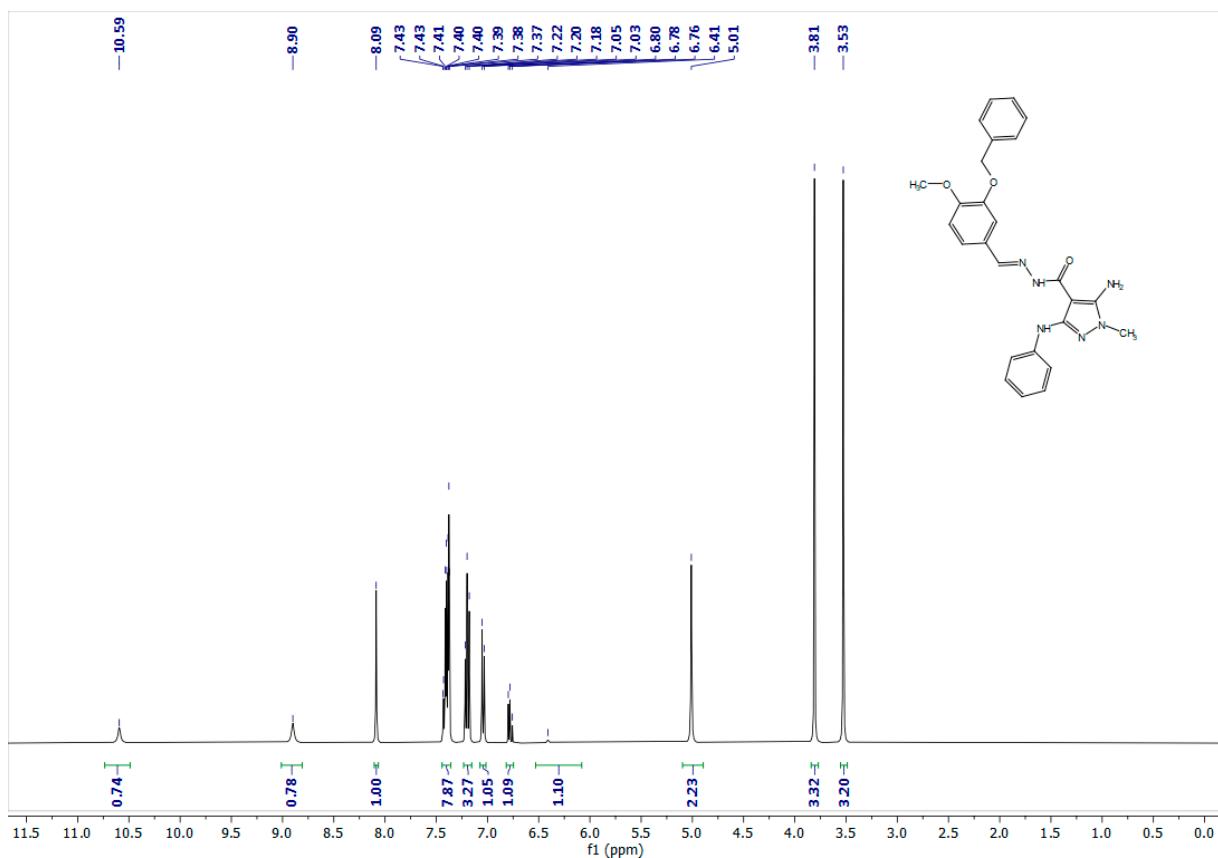


Figure S20. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **12d**

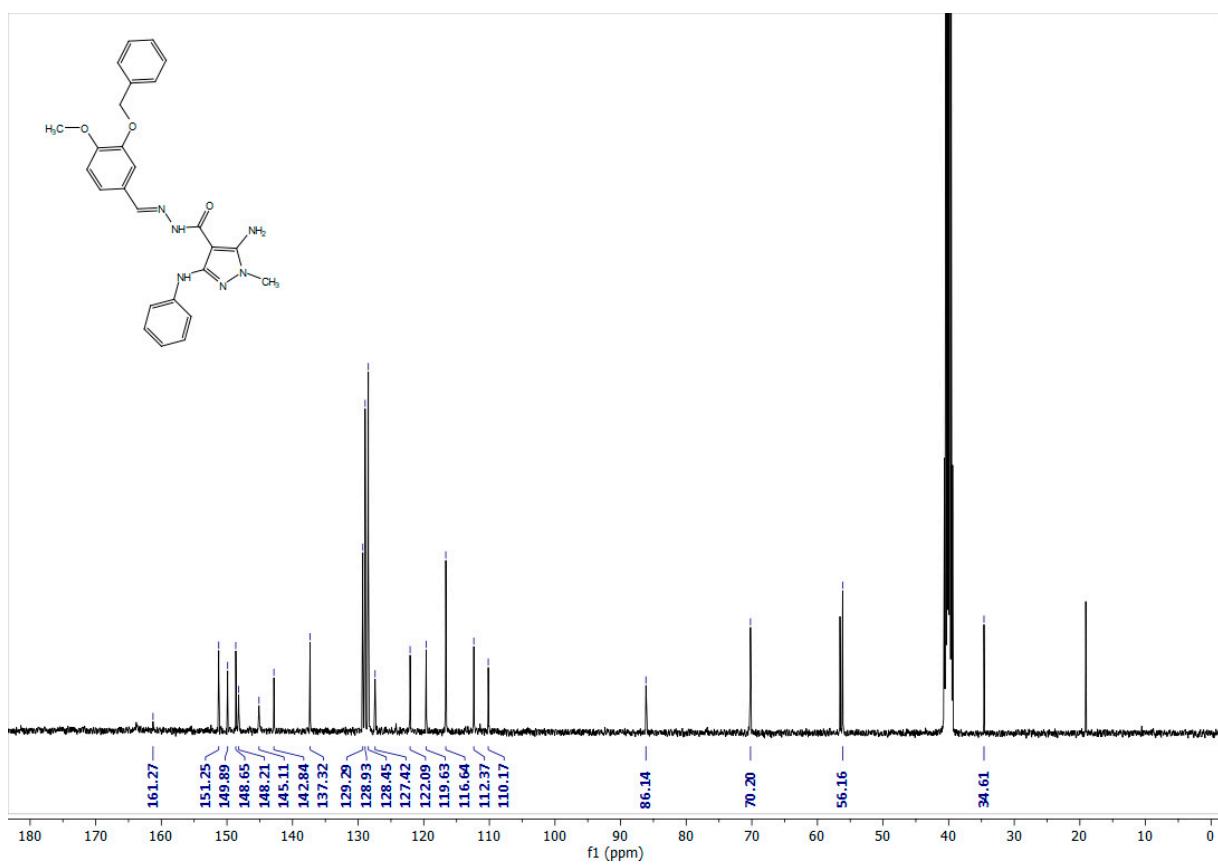


Figure S21. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **13a**

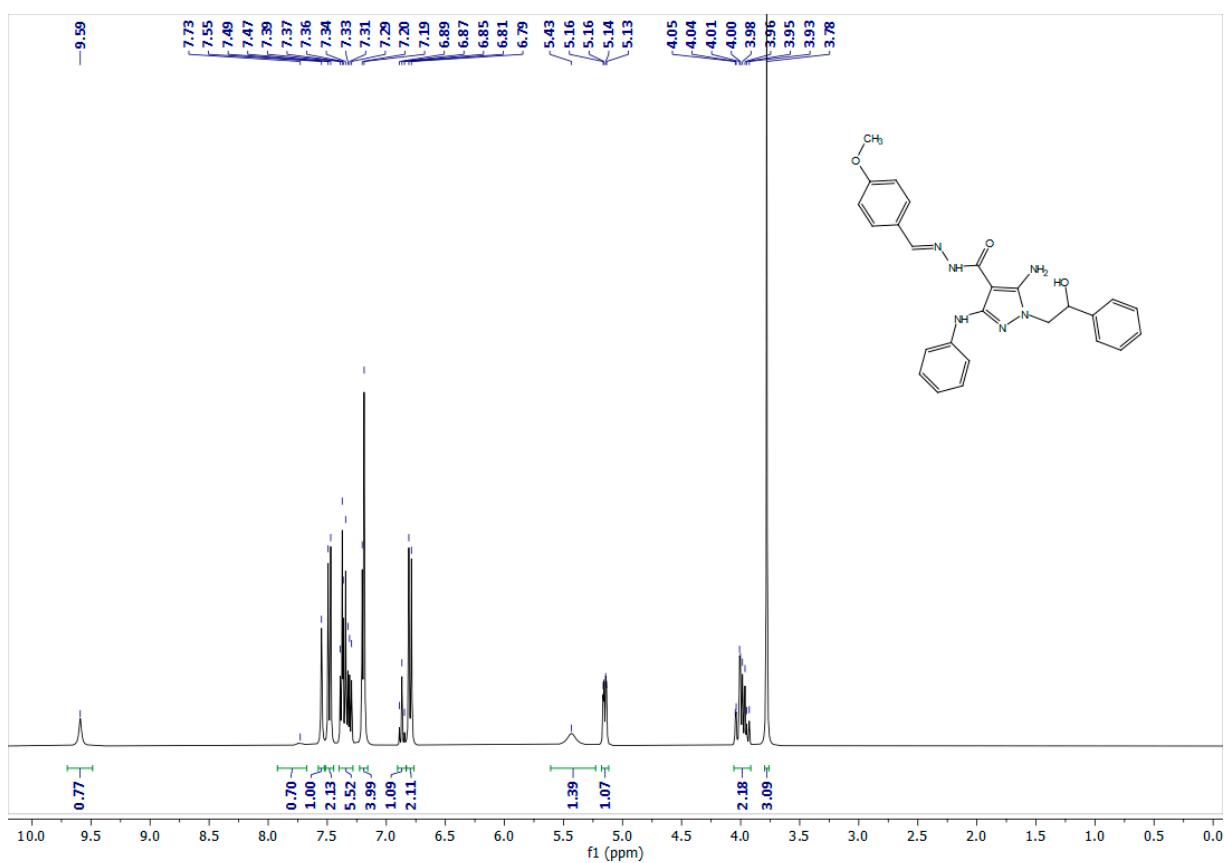


Figure S22. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **13a**

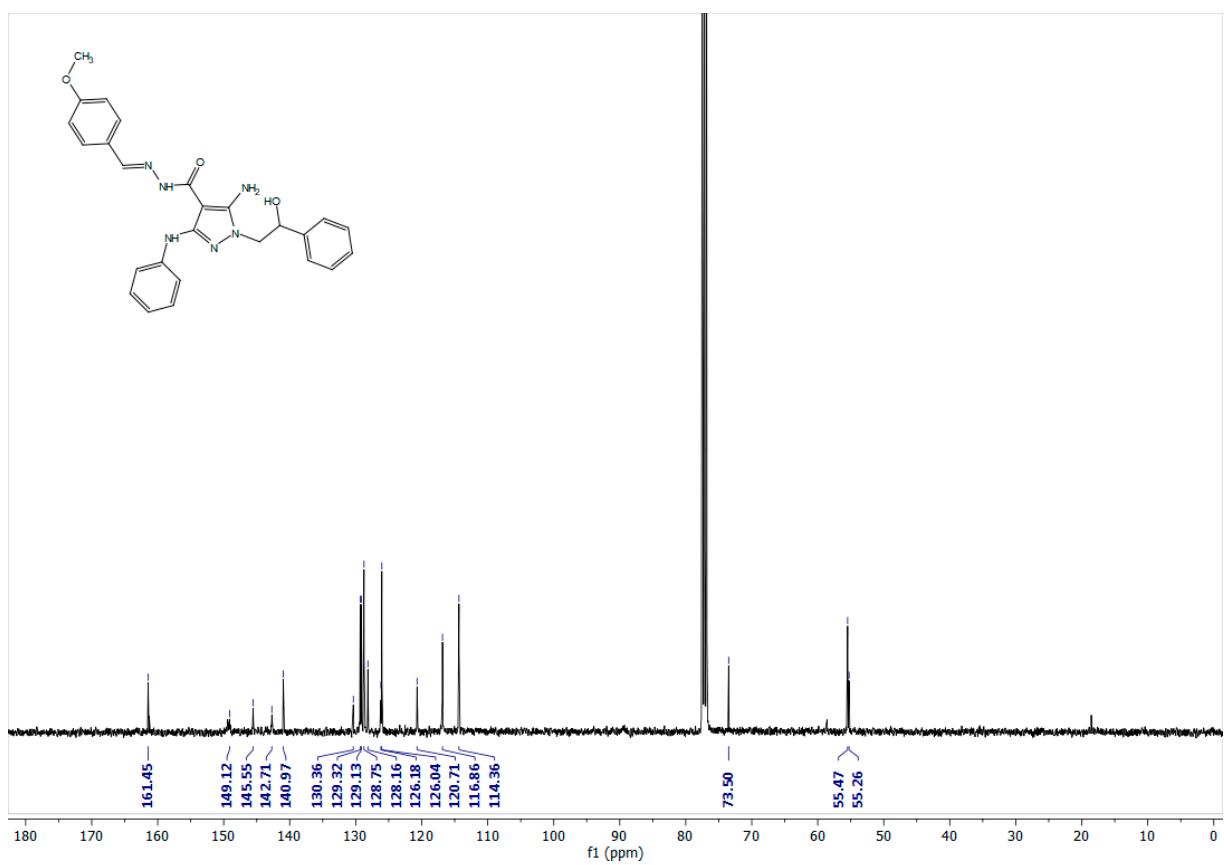


Figure S23. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **13b**

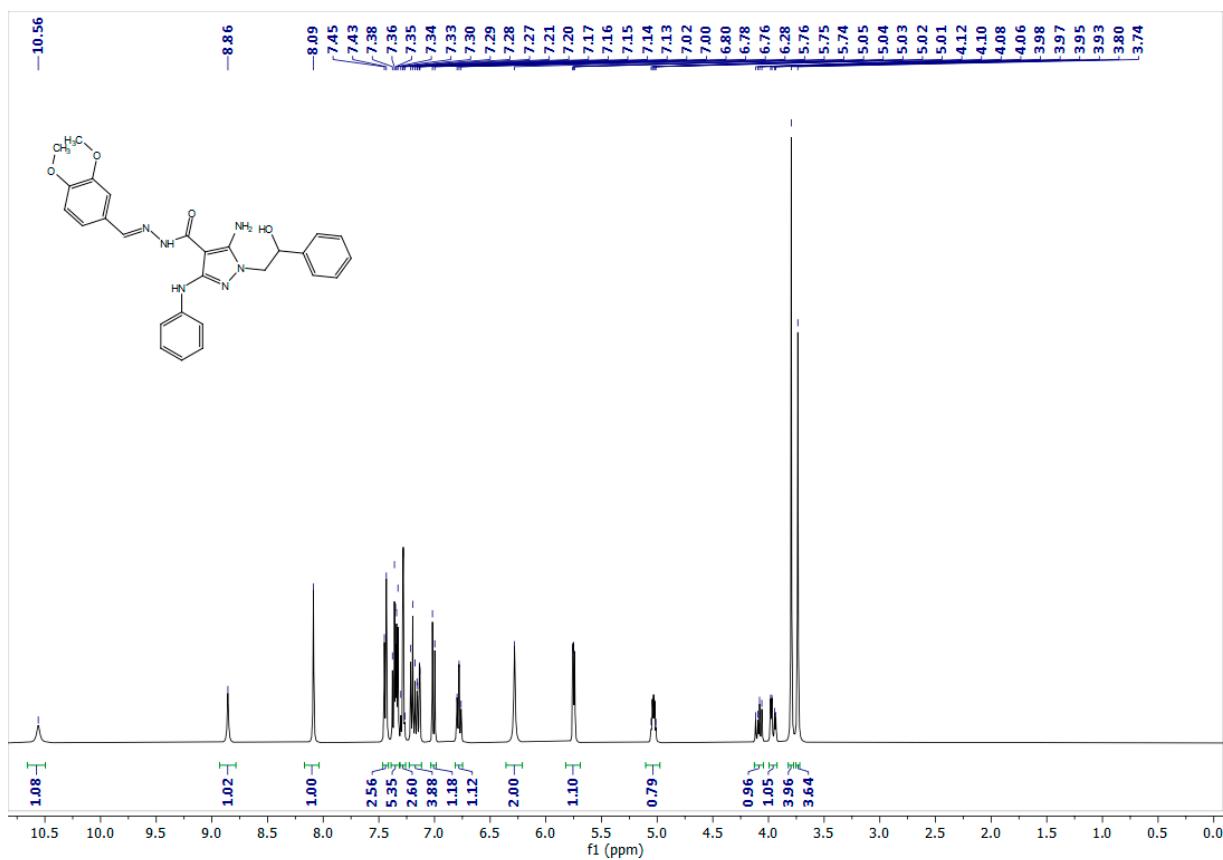


Figure S24. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **13b**

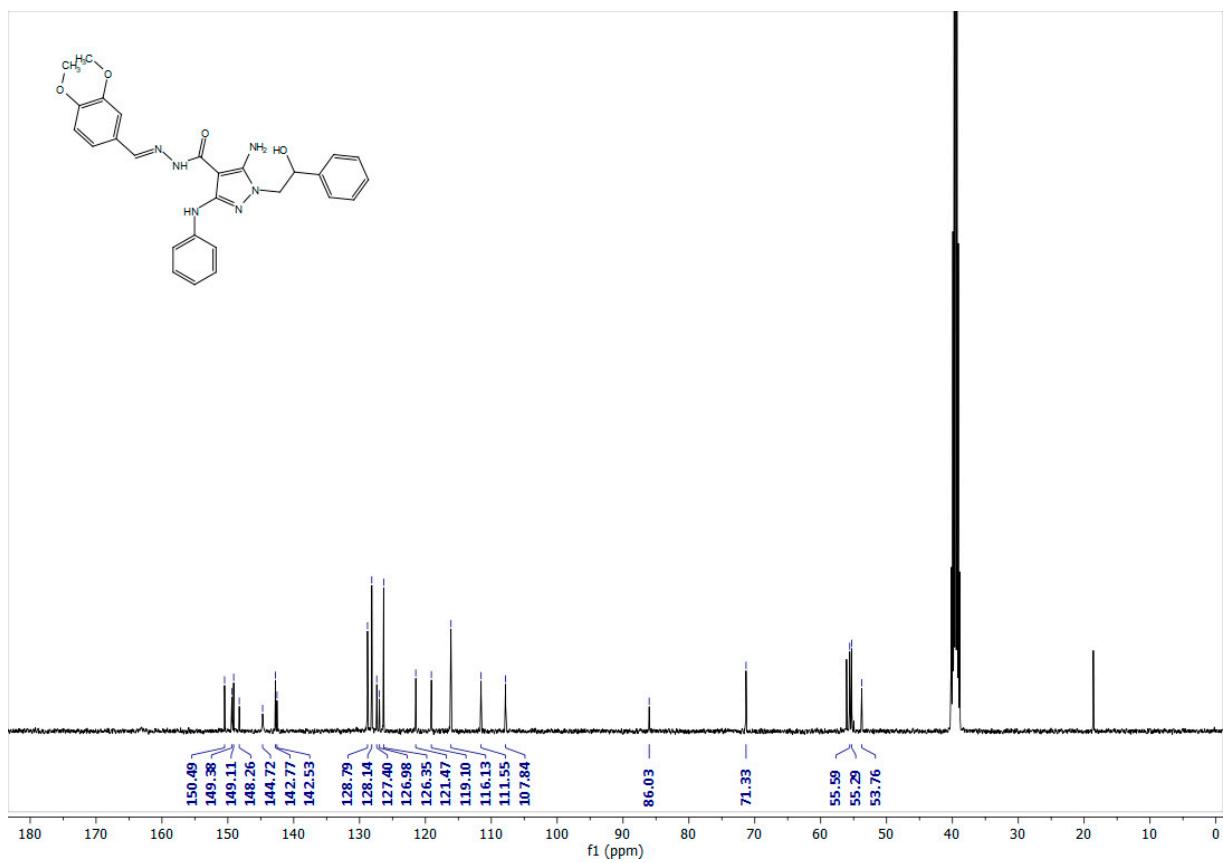


Figure S25. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **13c**

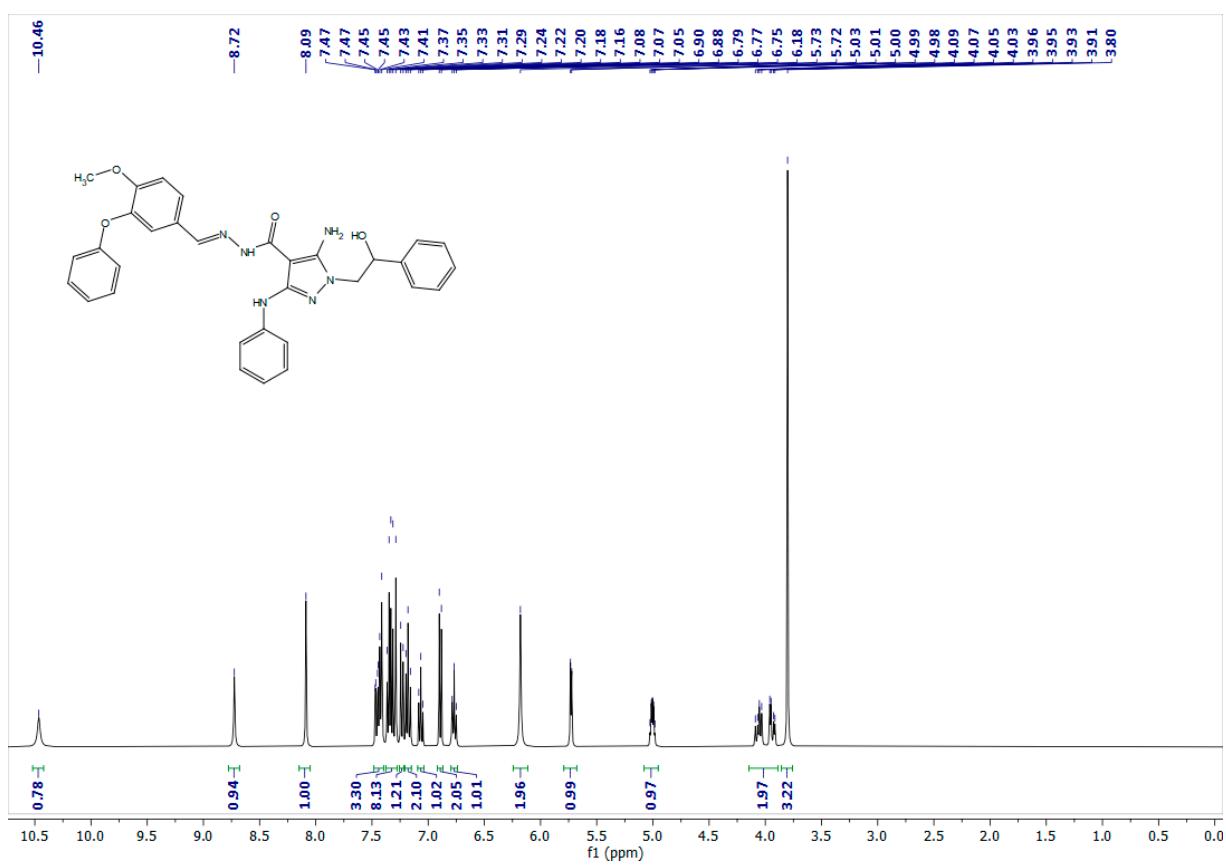


Figure S26. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **13c**

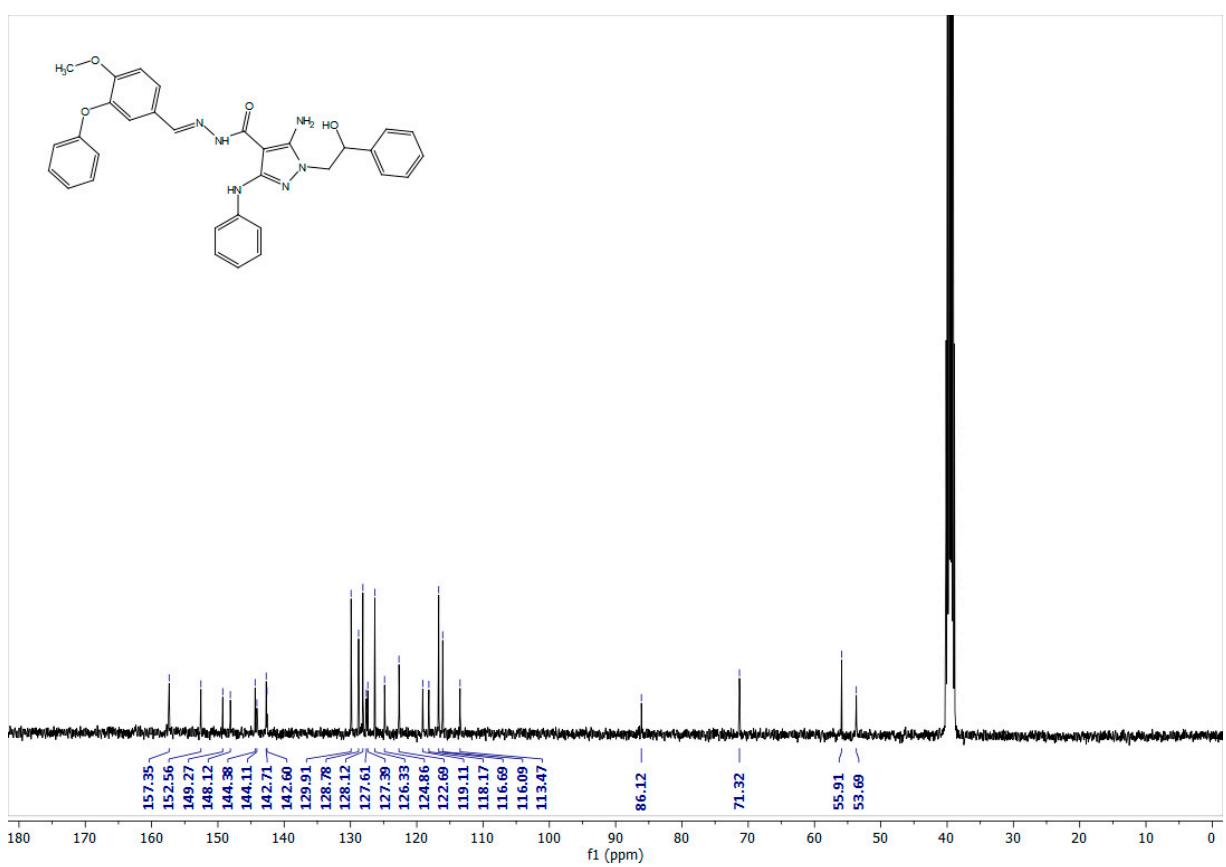


Figure S27. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **13d**

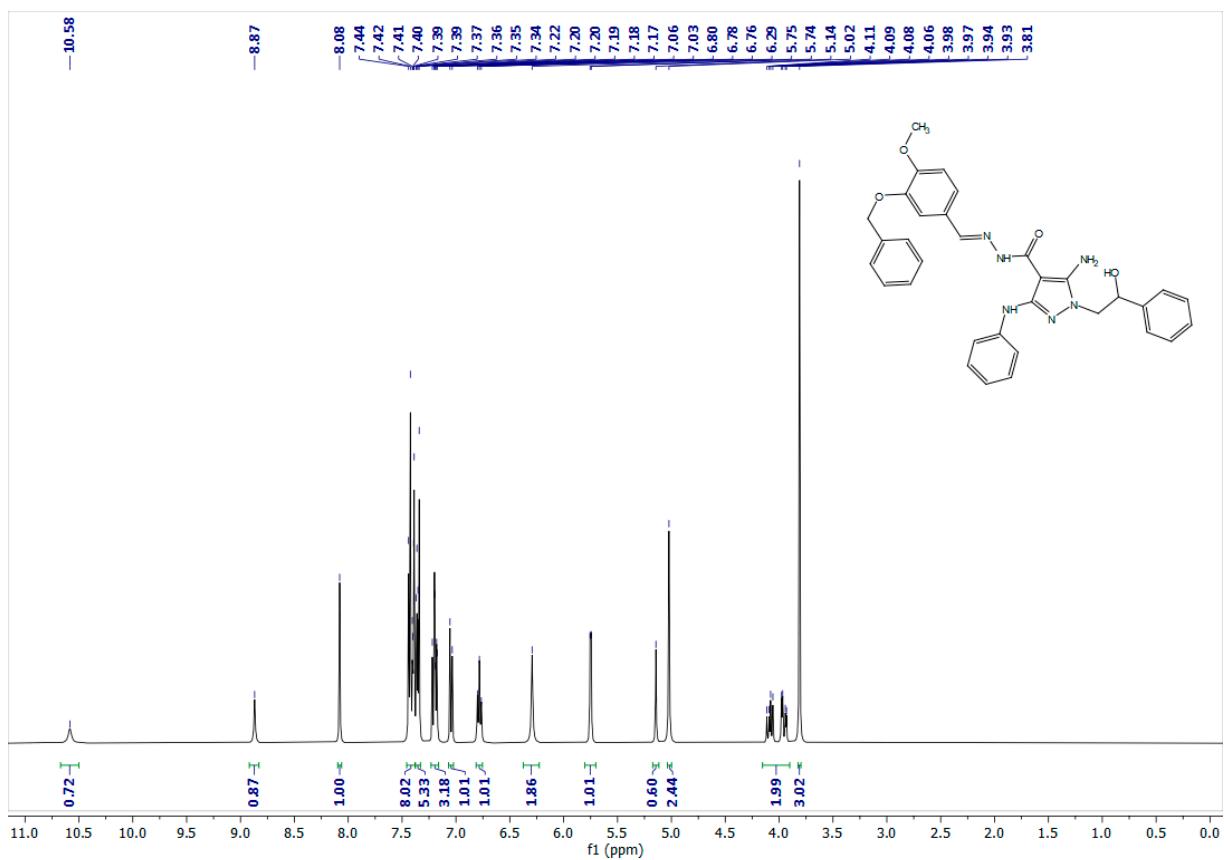


Figure S28. ^{13}C -NMR (101 MHz, $\text{d}_6\text{-DMSO}$) spectrum of compound **13d**

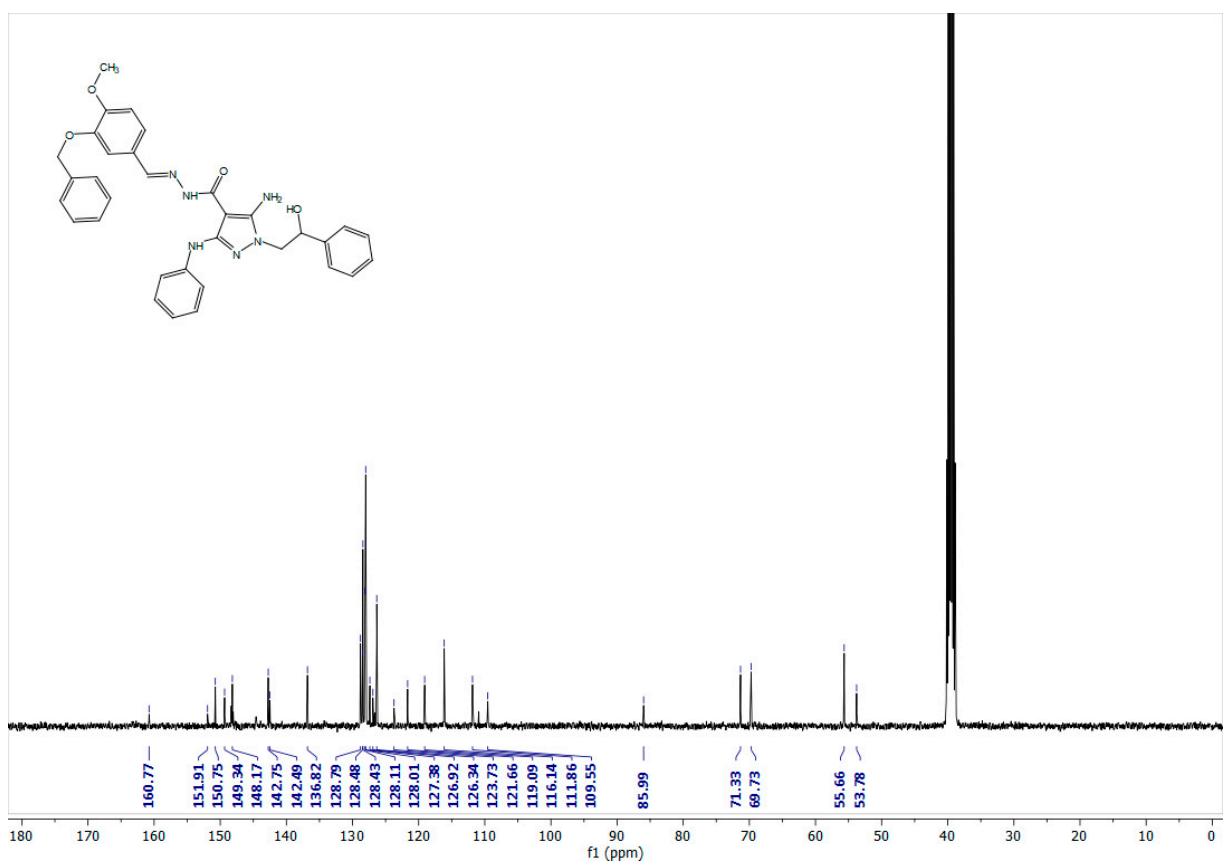


Figure S29. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **14**

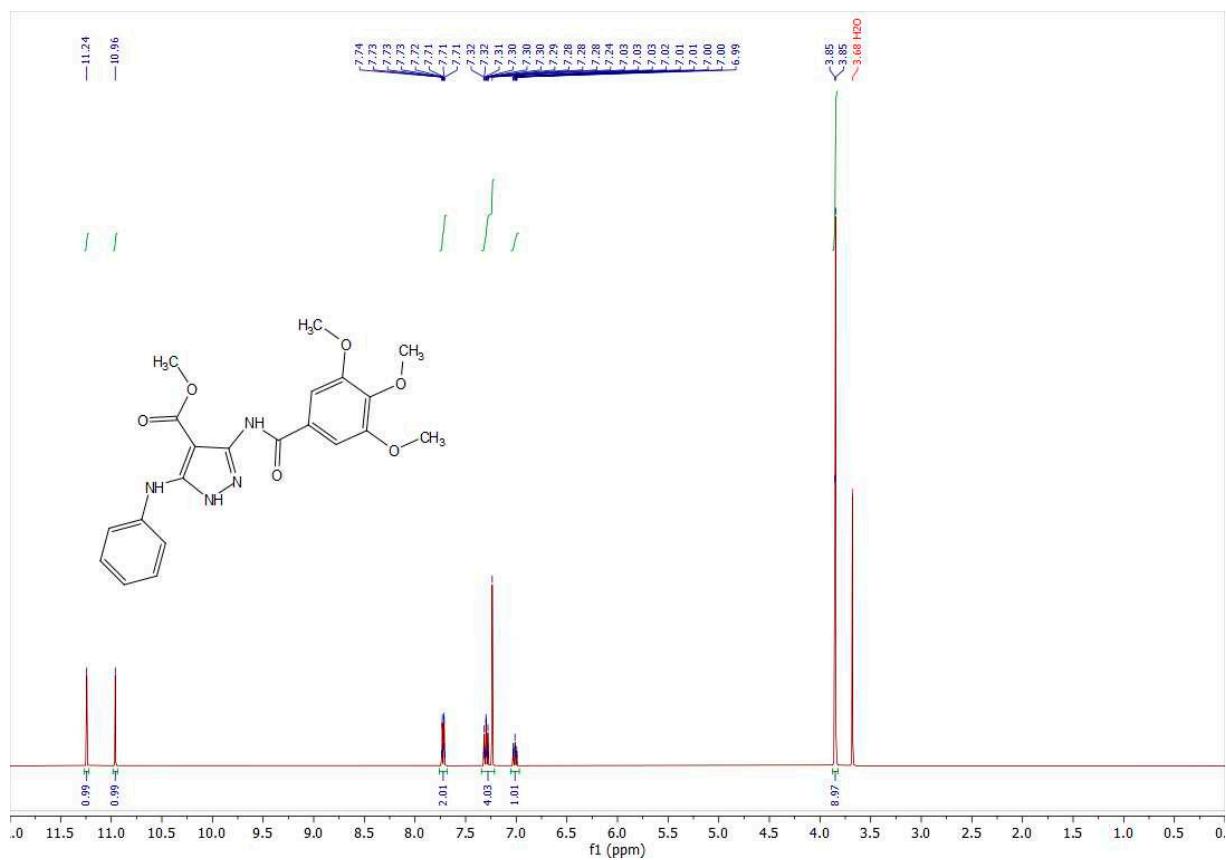


Figure S30. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **15**

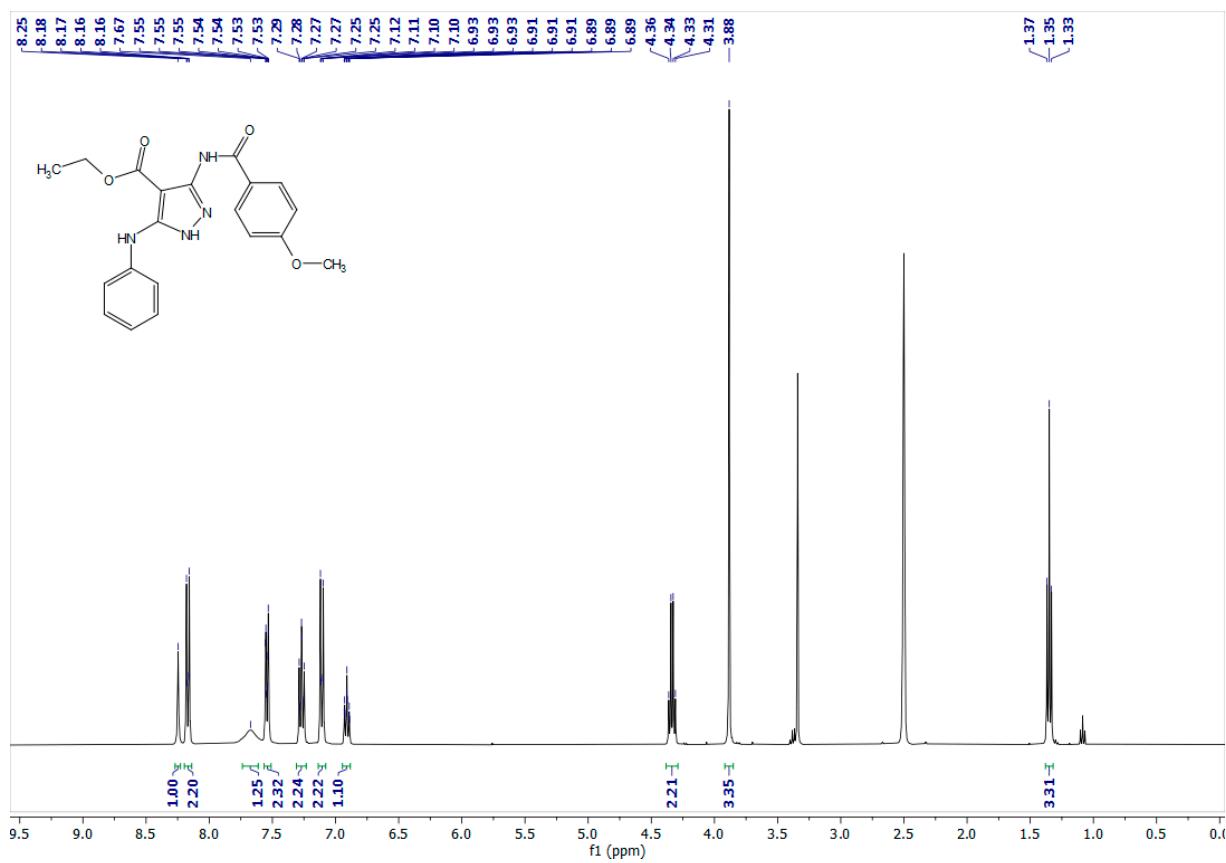


Figure S31. ^{13}C -NMR (101 MHz, $\text{d}_6\text{-DMSO}$) spectrum of compound **15**

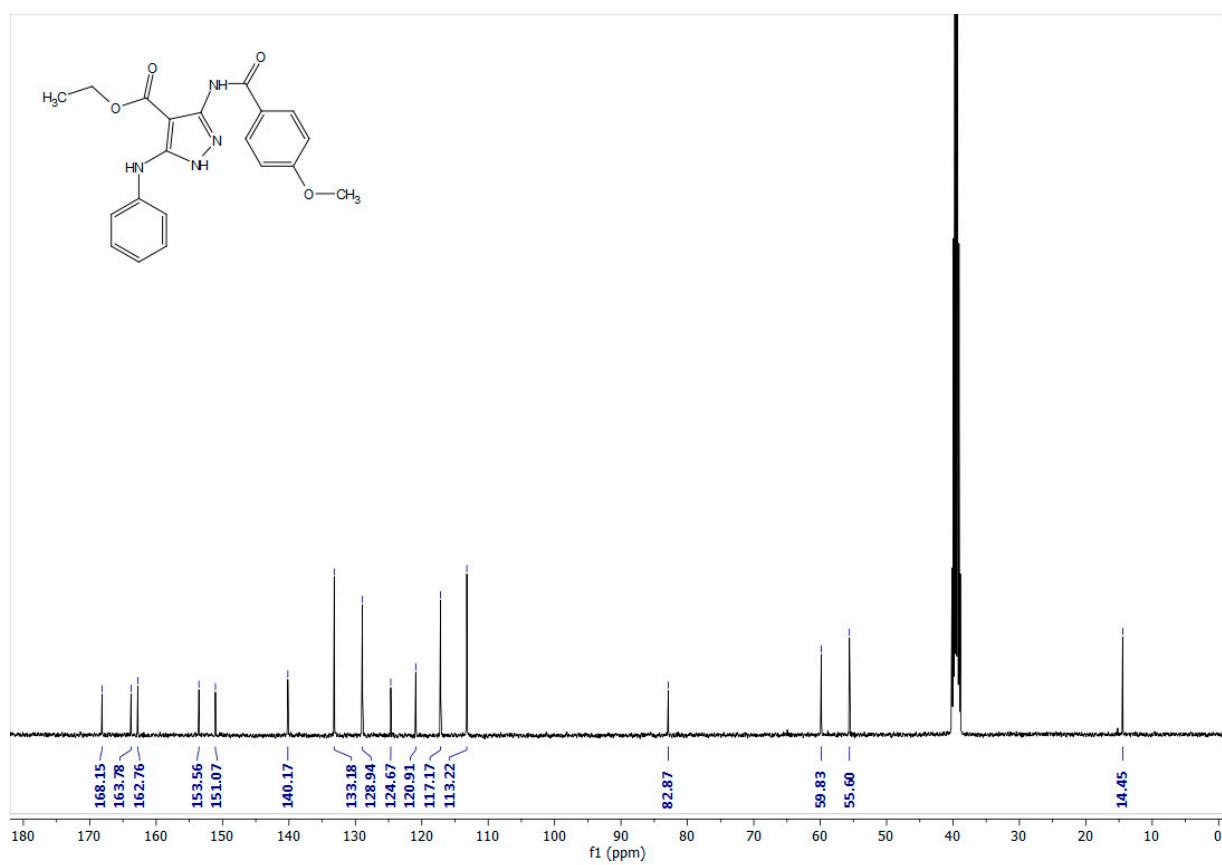


Figure S32. ^1H -NMR (400 MHz, $\text{d}_6\text{-DMSO}$) spectrum of compound **16**

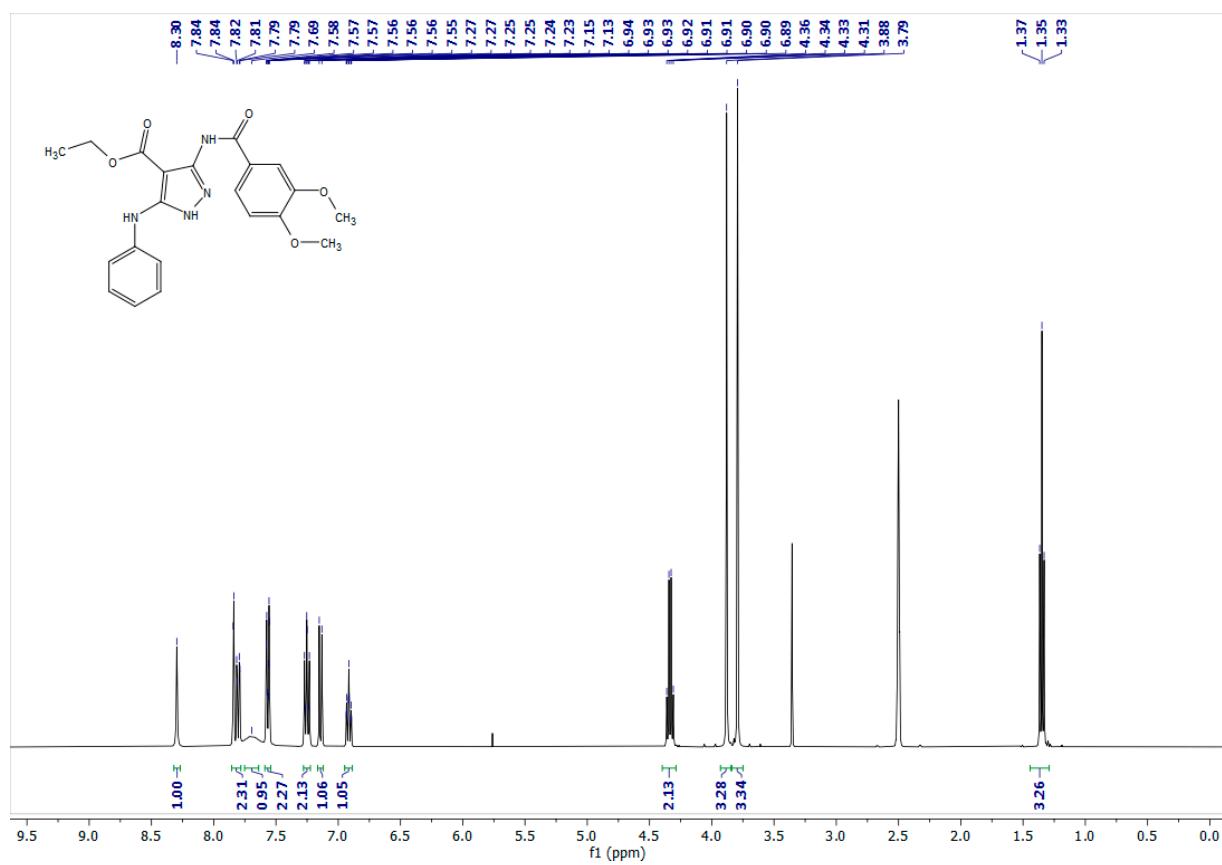


Figure S33. ^{13}C -NMR (101 MHz, $\text{d}_6\text{-DMSO}$) spectrum of compound **16**

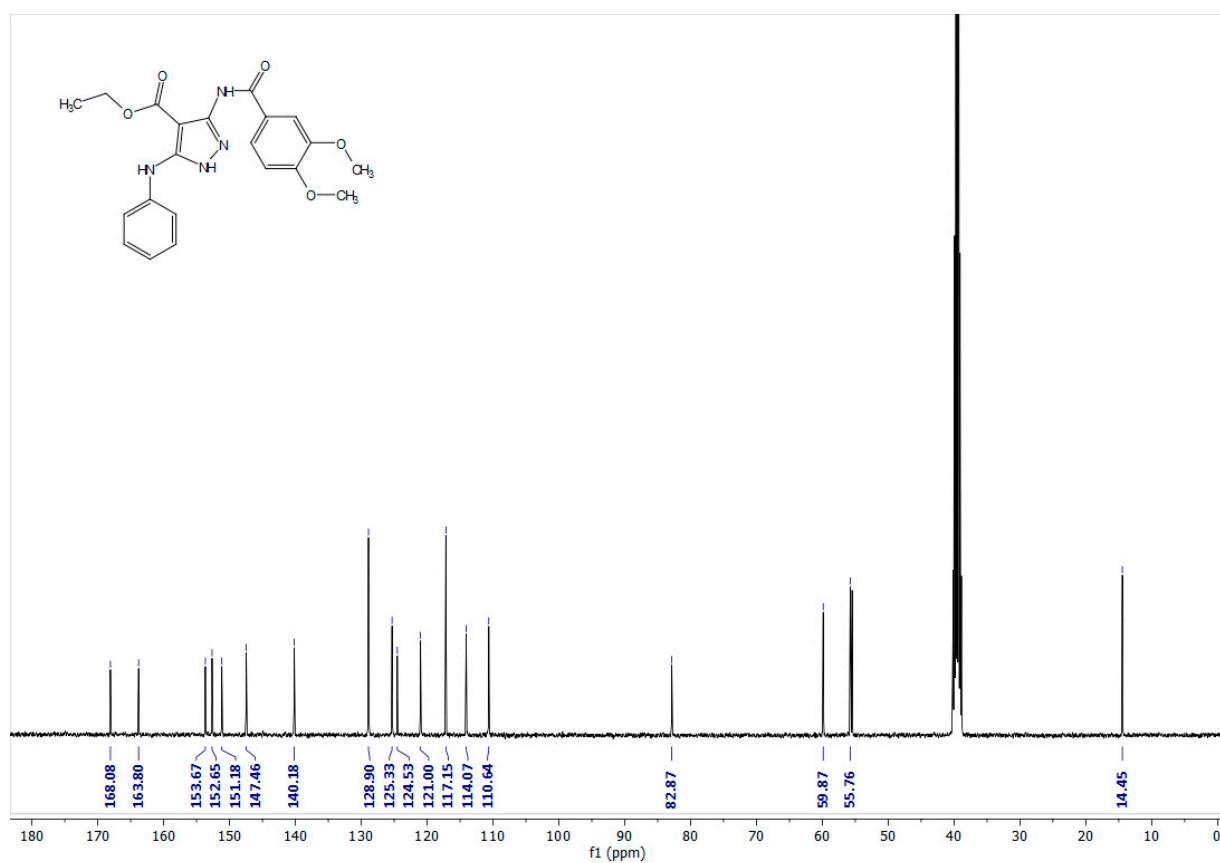


Figure S34. ^1H -NMR (400 MHz, $\text{d}_6\text{-DMSO}$) spectrum of compound **17**

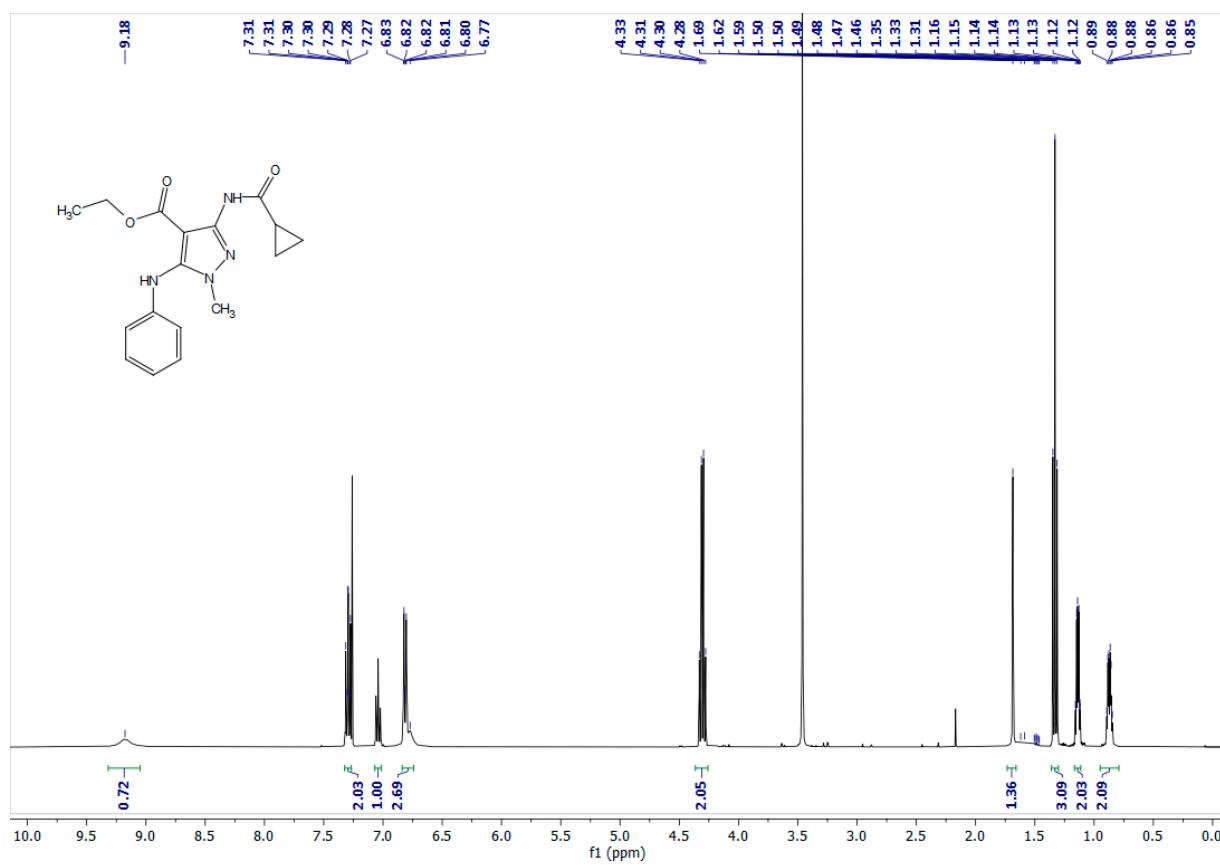


Figure S35. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **18**

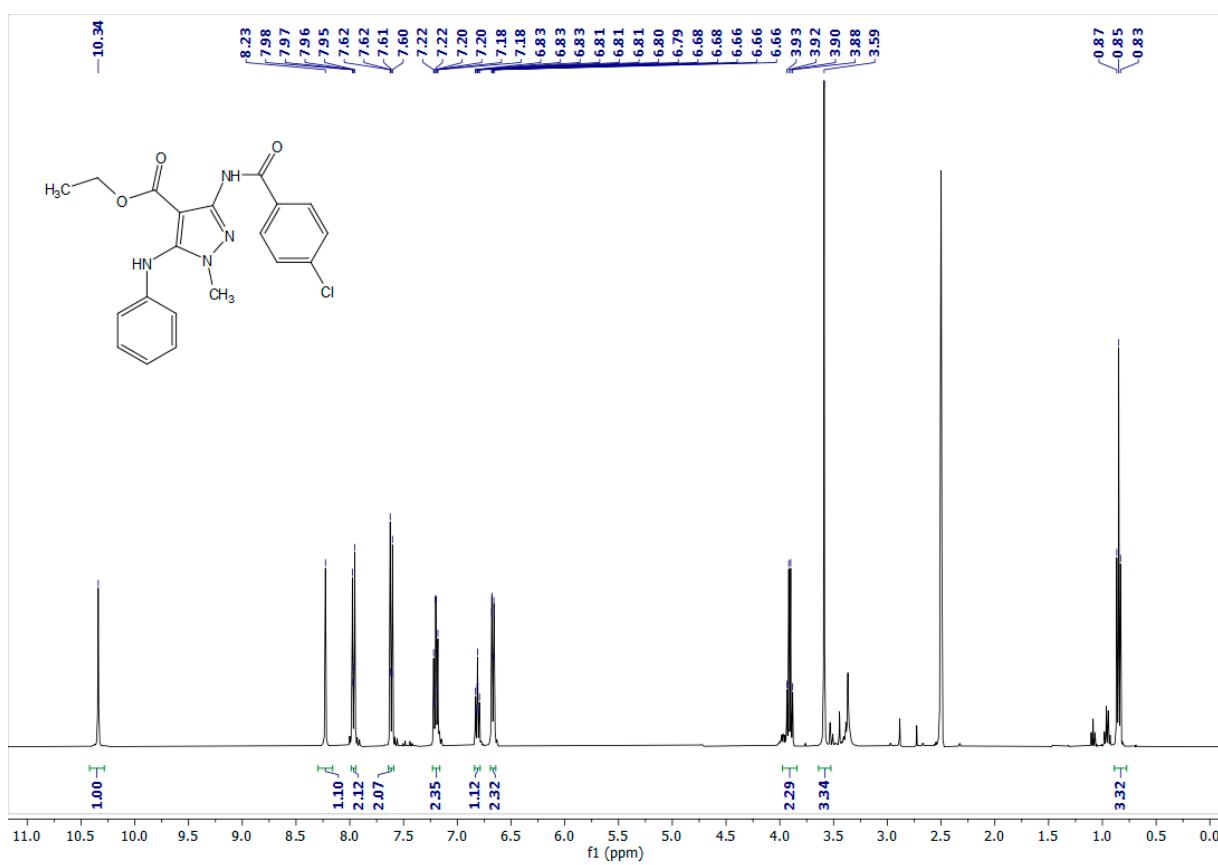


Figure S36. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **18**

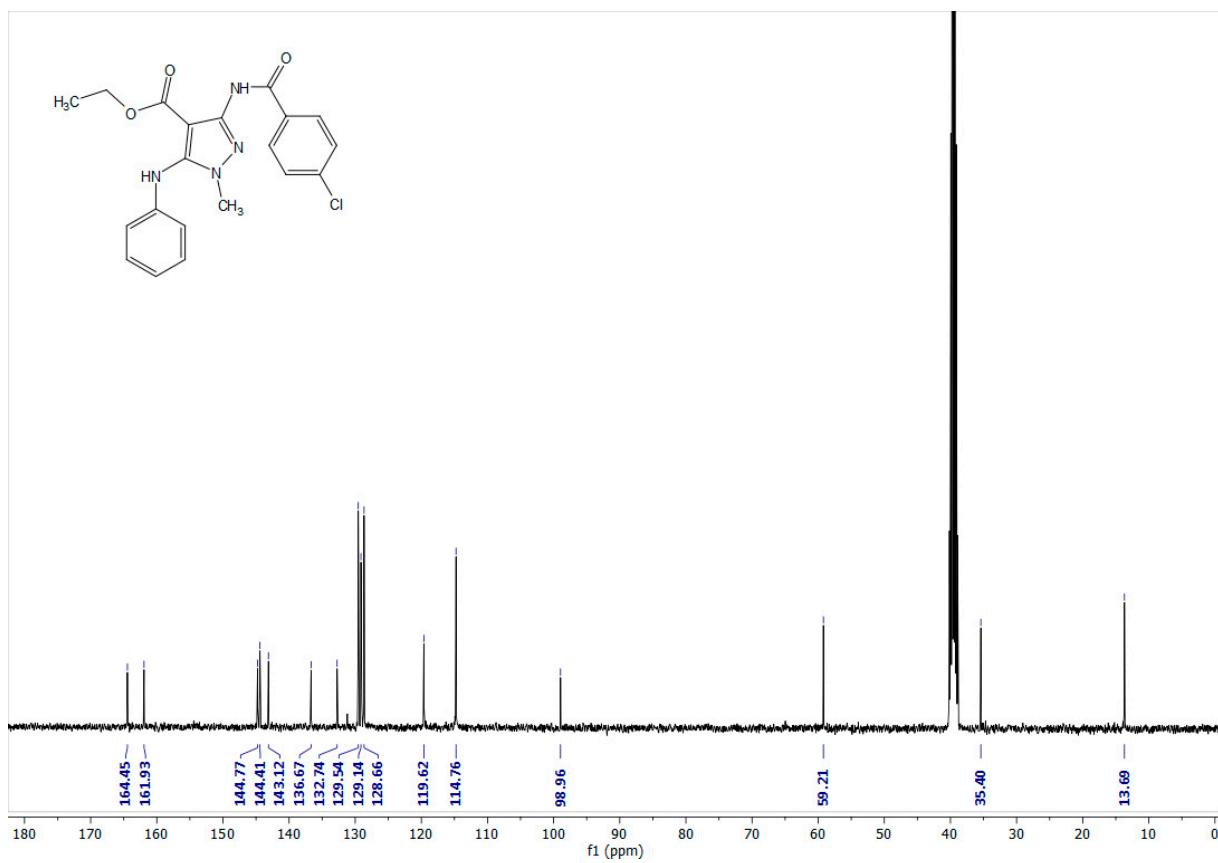


Figure S37. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **19**

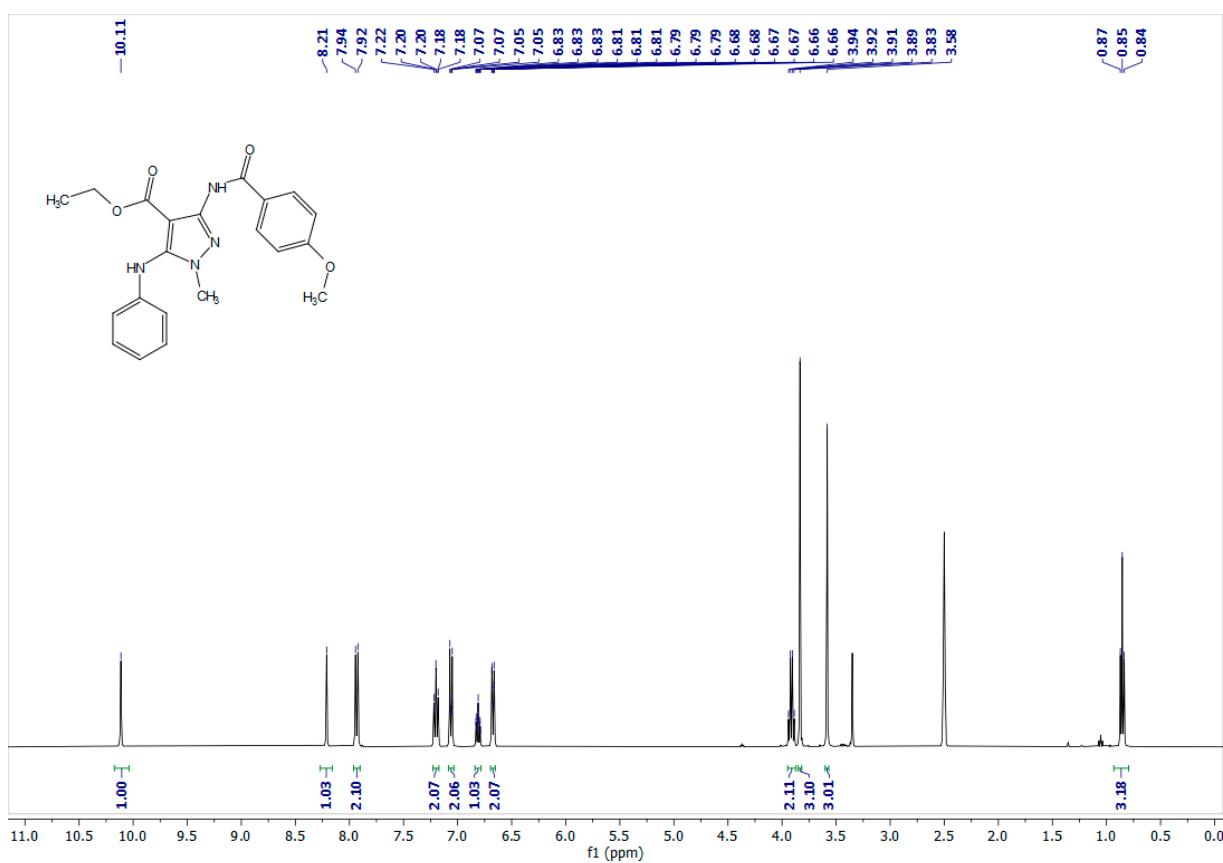


Figure S38. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **19**

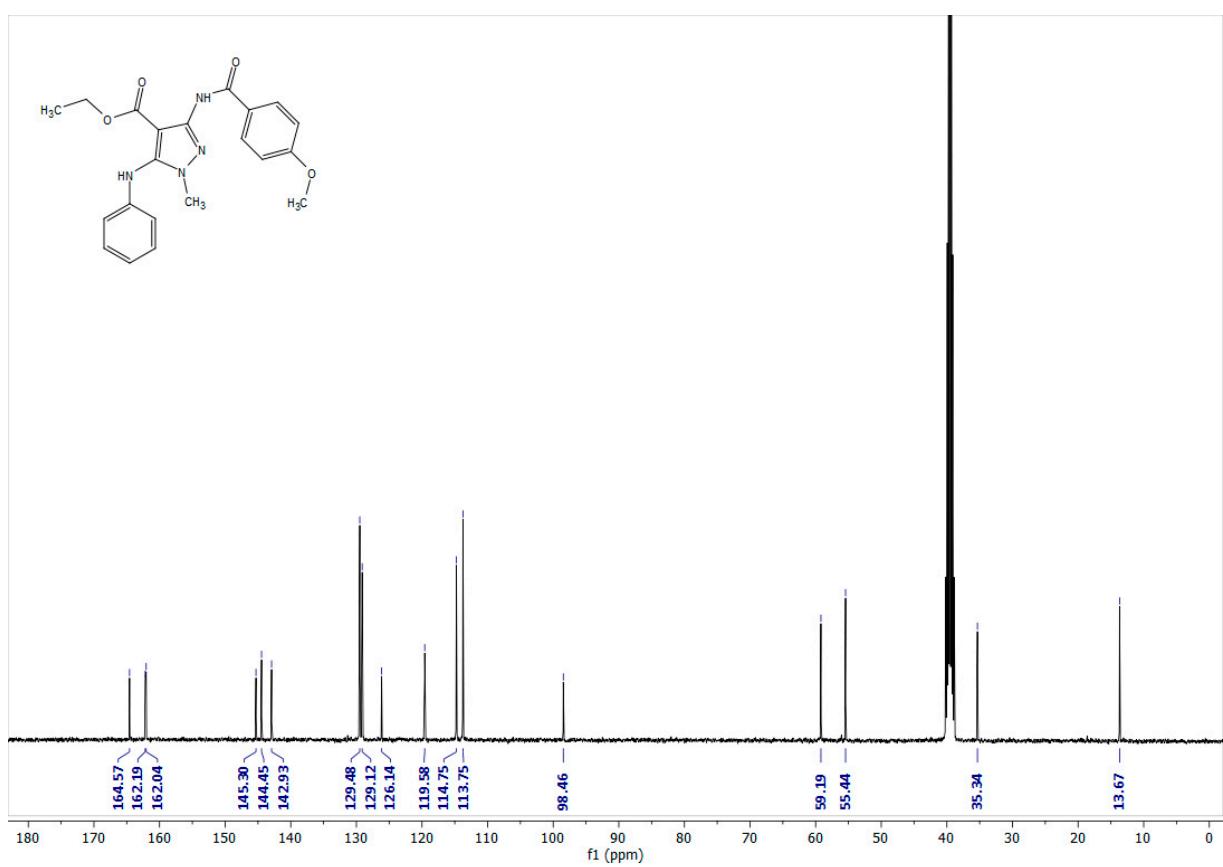


Figure S39. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **20**

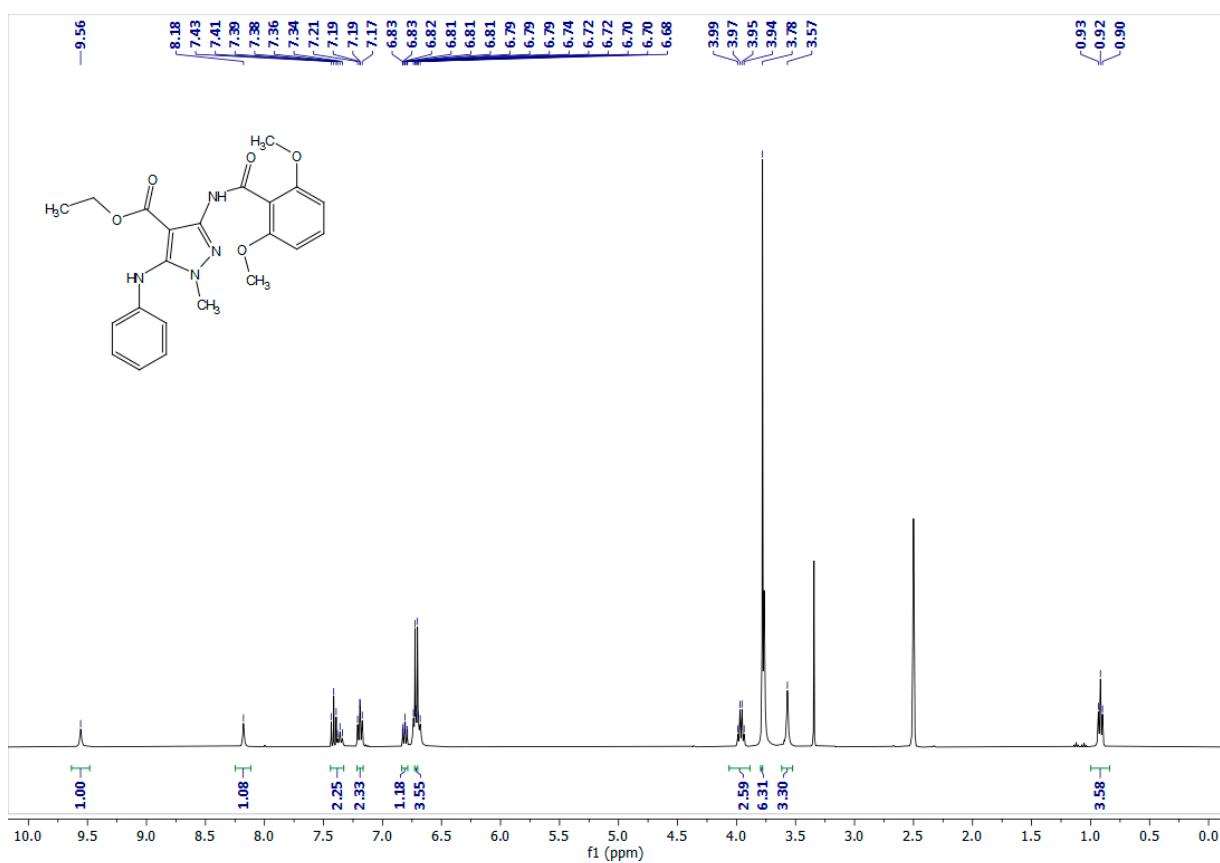


Figure S40. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **20**

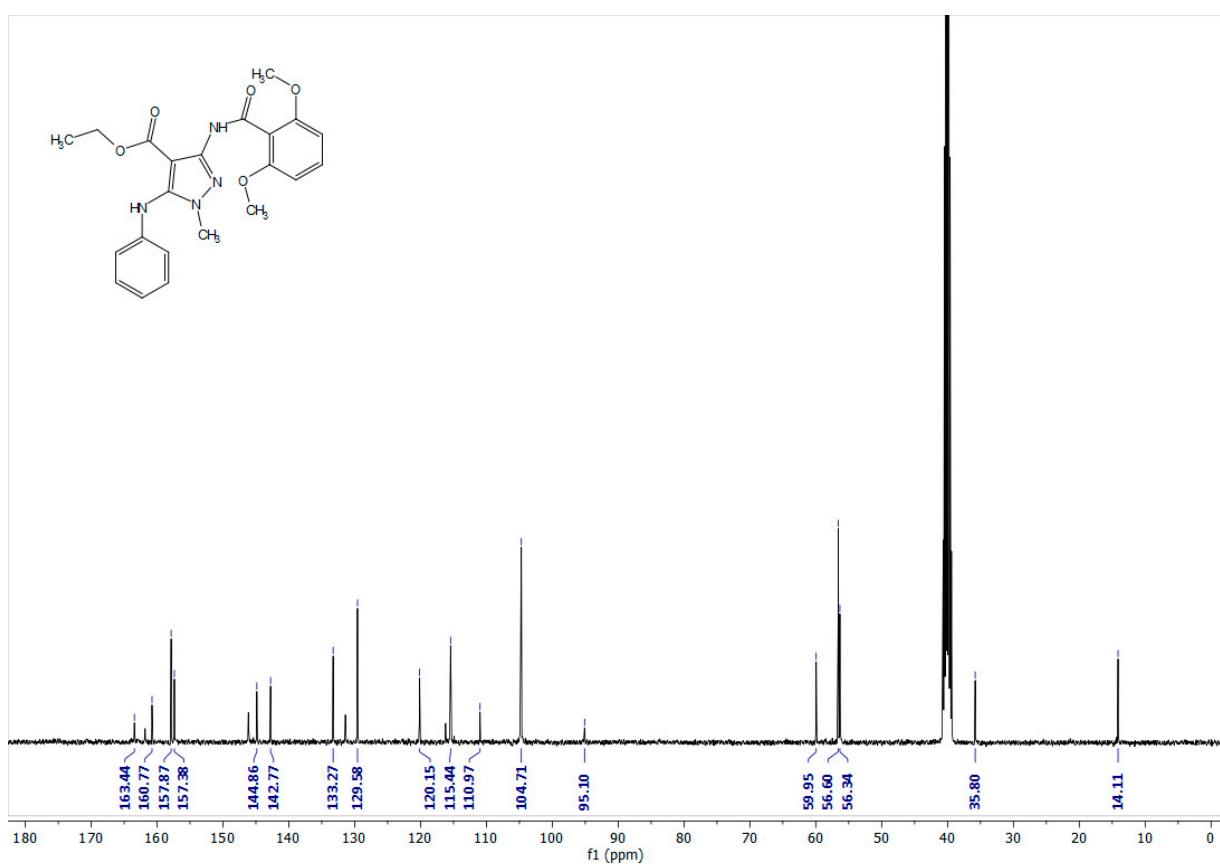


Figure S41. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **21**

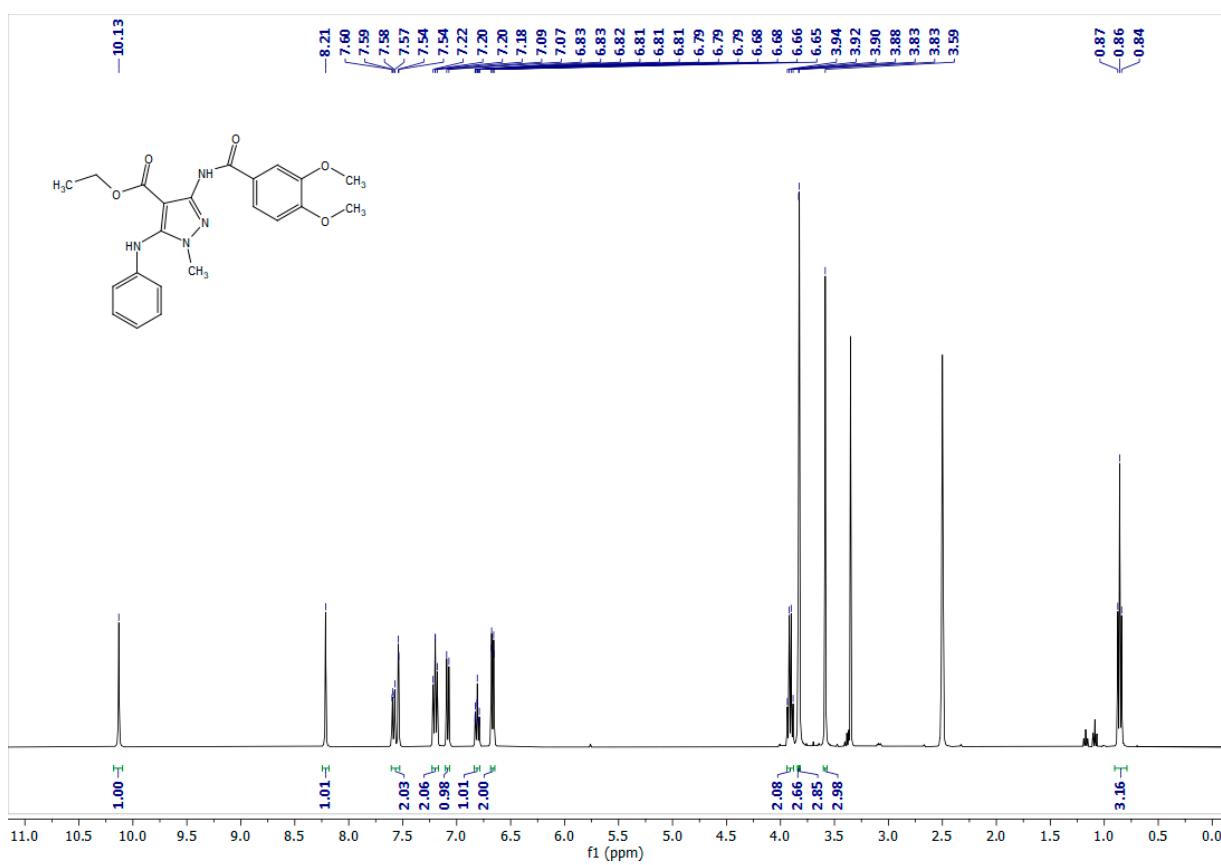


Figure S42. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **21**

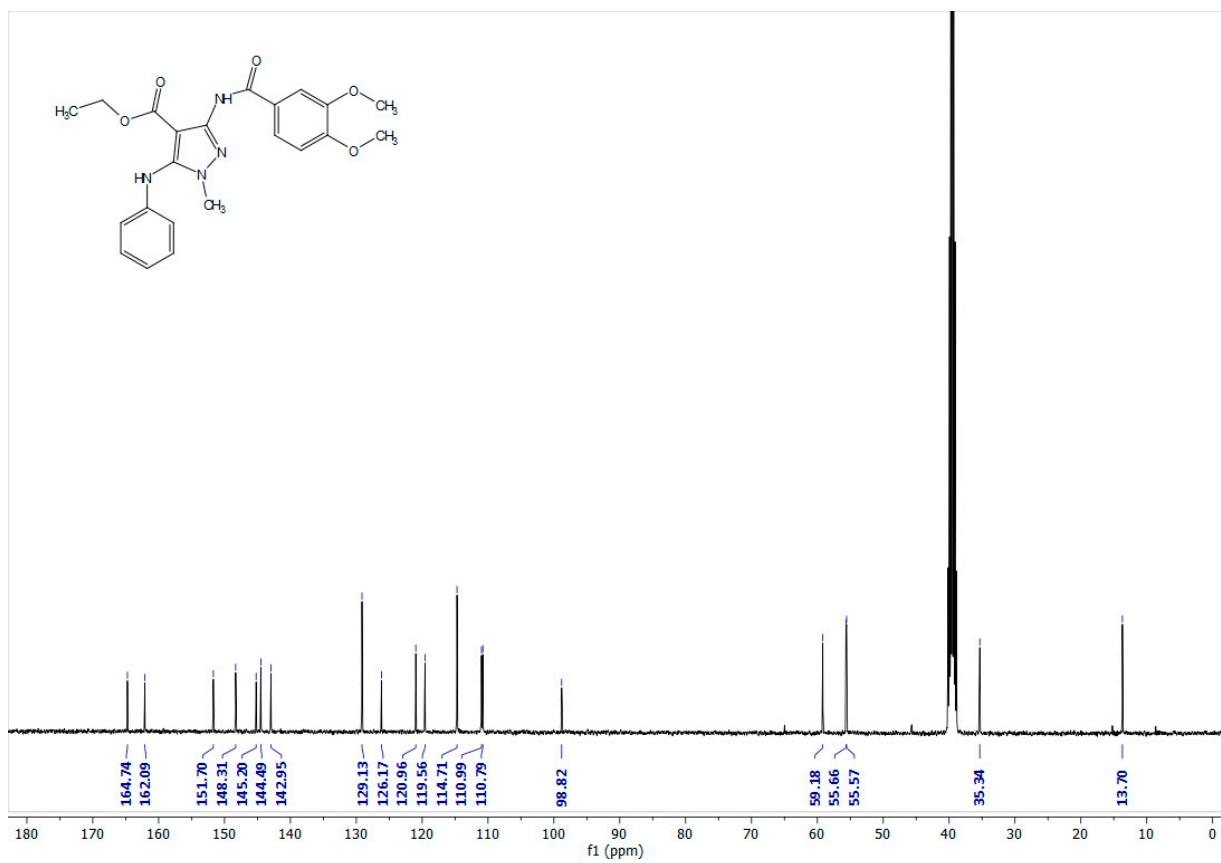


Figure S43. ^1H -NMR (400 MHz, d_6 -DMSO) spectrum of compound **22**

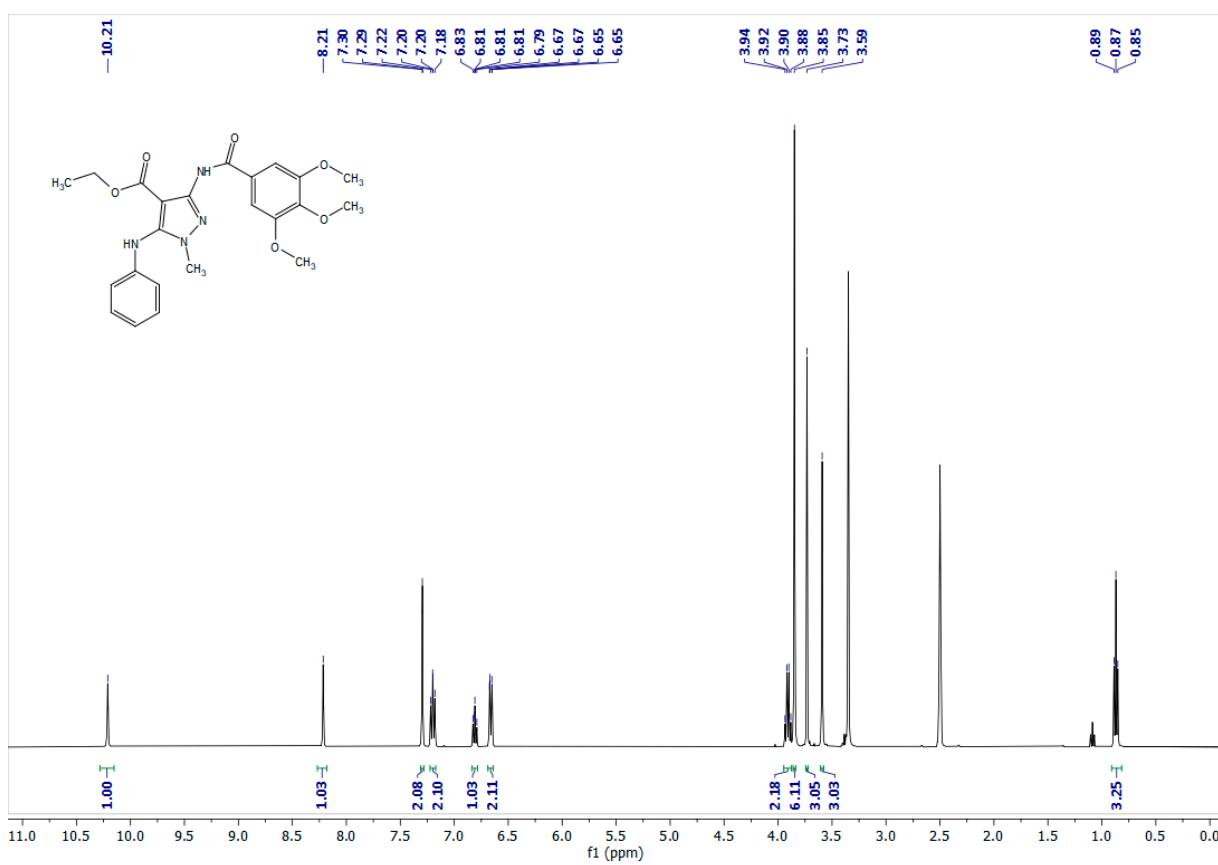


Figure S44. ^{13}C -NMR (101 MHz, d_6 -DMSO) spectrum of compound **22**

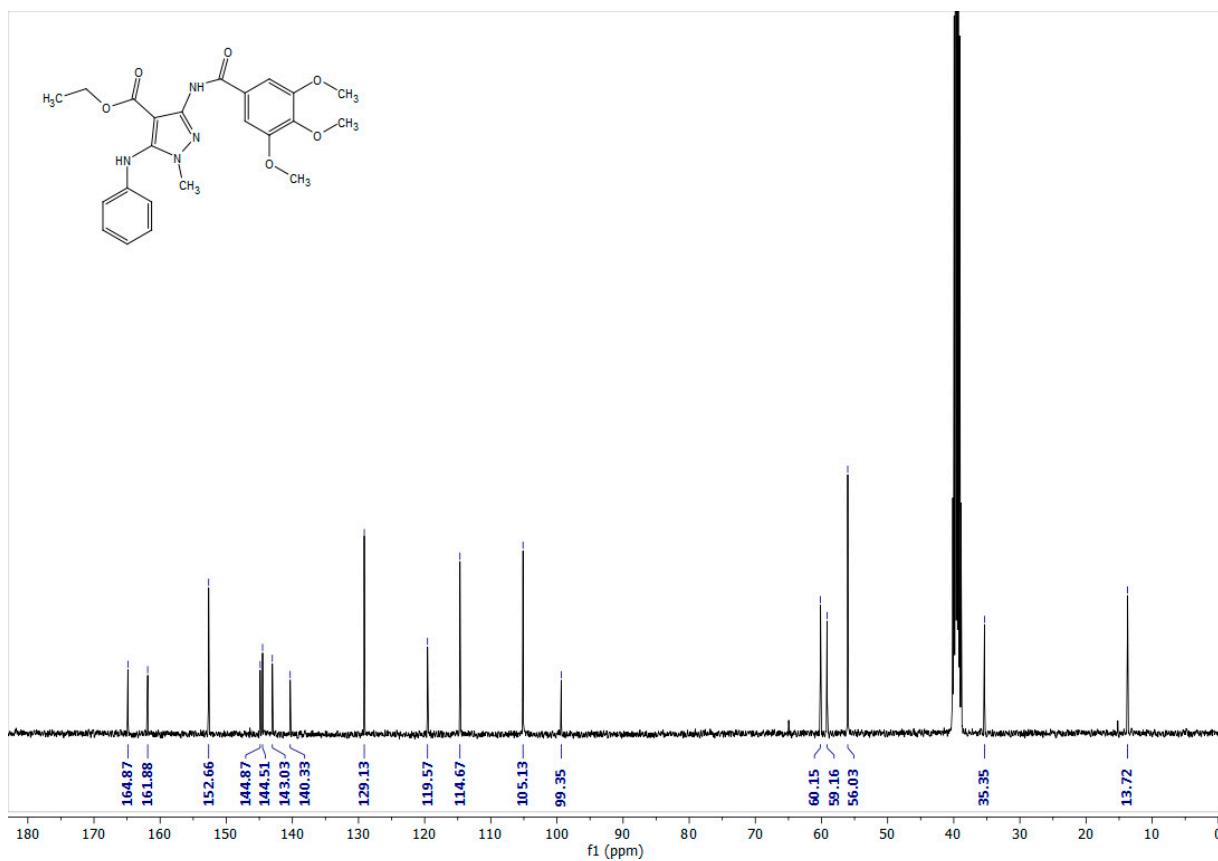


Table S1. Inhibitory effect of compounds **10-22** on platelet aggregation and ROS production

Cpd	IC ₅₀ (μ M \pm SD) ^a	
	Aggregation inhibition	ROS production inhibition
10a	204 \pm 12	111 \pm 9
10b	94 \pm 7	104 \pm 4
11a	183 \pm 13	123 \pm 12
11b	129 \pm 10	113 \pm 7
11c	841 \pm 43	148 \pm 13
11d	890 \pm 19	182 \pm 11
12a	265 \pm 23	113 \pm 7
12b	187 \pm 16	106 \pm 9
12c	331 \pm 14	193 \pm 16
12d	433 \pm 16	171 \pm 9
13a	883 \pm 39	213 \pm 17
13b	909 \pm 87	433 \pm 32
13c	313 \pm 29	209 \pm 15
13d	437 \pm 25	405 \pm 29
14	365 \pm 28	387 \pm 35
15	250 \pm 19	323 \pm 14
16	292 \pm 28	310 \pm 24
17	460 \pm 35	573 \pm 43
18	268 \pm 25	273 \pm 17
19	294 \pm 26	293 \pm 18
20	301 \pm 24	289 \pm 27
21	278 \pm 27	262 \pm 13
22	249 \pm 22	313 \pm 25
NAC	ND	872 \pm 26
ASA	438 \pm 18	ND

^aReported data are the mean value \pm standard deviation (SD) obtained in at least six different experiments each performed in duplicate.