

Supplementary information for

Antibacterial Thiopeptide GE2270-Congeners from *Nonomuraea jiangxiensis*

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Figure S45. Dose response curve of compounds **1-9** against A549 Human lung carcinoma cells (ATCC® CCL-185TM).

Table S1. NMR spectral data of **3** and **4**.

Residue	Position	3		4	
		¹³ C	¹ H, mult. (<i>J</i> = Hz)	¹³ C	¹ H, mult. (<i>J</i> = Hz)
Proline (Pro)	α	58.6, CH	4.25, m	58.5, CH	4.24, m
	β	27.9, CH ₂	1.92, m; 2.19, m	27.9, CH ₂	1.90, m; 2.19, m
	γ	22.1, CH ₂	1.88, m; 1.91, m	22.1, CH ₂	1.84, m; 1.91, m
	δ	44.6, CH ₂	3.40, m; 3.51, m	44.8, CH ₂	3.41, m; 3.51, m
	CO	169.4, C	-	169.3, C	-
Oxazoline (Oxa)	α	53.9, CH	4.55, m	53.9, CH	4.55, m
	β	63.2, CH ₂	4.54, m; 4.68, m	63.2, CH ₂	4.54, m; 4.68, dd (16.0, 8.0)
	CN	160.5, C	-	160.1, C	-
	CO	163.2, C	-	163.2, C	-
Thiazole A	2	167.9, C	-	167.9, C	-
	4	147.2, C	-	147.2, C	-
	5	132.5, CH	8.76, s	132.8, CH	8.75, s
Pyridine (Py)	2	150.1, C	-	150.1, C	-
	3	127.77, C	-	127.6, C	-
	4	141.2, CH	8.44, d (8.2)	141.3, CH	8.45, d (8.2)
	5	118.3, CH	8.27, d (8.2)	118.4, CH	8.28, d (8.2)
	6	150.0, C	-	150.0, C	-
Thiazole B	2	160.5, C	-	160.3, C	-
	4	153.2, C	-	153.2, C	-
	5	122.8, CH	8.31, s	123.1, CH	8.31, s
Thiazole C	2	170.7, C	-	171.0, C	-
	4	146.4, C	-	146.7, C	-
	5	116.4, CH	7.37, s	116.3, CH	7.37, s
Phenylserine (PheSer)	α	58.1, CH	5.24, m	58.0, CH	5.24, m
	β	73.3, CH	5.03, m	73.6, CH	5.02, m
	1	141.1, C	-	141.6, C	-
	2,6	126.5, CH	7.29, (2H, m)	126.6, CH	7.31, (2H, m)
	3,5	127.80, CH	7.29, (2H, m)	127.8, CH	7.31, (2H, m)
	4	127.4, CH	7.24, m	127.5, CH	7.25, m
	NH	-	9.00, d (7.7)	-	9.01, d (8.0)
	OH	-	6.03, d (4.3)	-	6.03, d (4.5)
Glycine (Gly)	α	41.0, CH ₂	3.88, dd (16.6, 4.0); 4.25, m	41.0, CH ₂	3.81, dd (17.2, 4.0); 4.30 dd (17.2, 8.6)
	CO	169.6, C	-	169.4, C	-
	NH	-	8.56, m	-	8.46, m
	2	168.2, C	-	165.4, C	-
Thiazole D	4	148.0, C	-	143.7, C	-
	5	124.4, CH	8.29, s	140.6, C	-
	5-CH ₂	-	-	67.2, CH ₂	4.98, s
	5-CH ₂ OMe	-	-	58.4, CH ₃	3.39, s
	CO	160.4, C	-	161.2, C	-

	α	55.4, CH	5.29, dd (8.0, 4.9)	55.5, CH	5.26, s
	β	33.8, CH	2.22, m	33.9, CH	2.21, s
Valine (Val)	γ	18.5, CH ₃	0.87, d (6.9)	18.4, CH ₃	0.86, d (6.6)
	γ'	17.7, CH ₃	0.89, d (7.0)	17.7, CH ₃	0.89, d (6.8)
	NH	-	8.76, d (8.0)	-	8.70, d (8.4)
	2	173.0, C	-	173.0, C	-
Thiazole E	4	148.4, C	-	148.6, C	-
	5	123.4, CH	8.10, s	123.3, CH	8.08, s
	CO	159.0, C	-	159.6, C	-
	α	48.0, CH	5.42, m	48.2, CH	5.37, m
	β	37.5, CH ₂	1.50, m; 2.76, m	37.7, CH ₂	1.35, m; 2.74, dd (16.5, 3.7)
Asparagine (Asn)	CONHMe	169.3, C	-	169.5, C	-
	NHMe	-	7.48, m	-	7.40, m
	Me	25.3, CH ₃	2.46, d (4.6)	25.6, CH ₃	2.46, d (4.7)
	NH	-	8.81, d (9.0)	-	8.74, d (8.0)
	2	164.5, C	-	164.6, C	-
Thiazole F	4	149.3, C	-	149.2, C	-
	5	126.8, CH	8.61, s	126.9, CH	8.62, s
	CO	160.0, C	-	160.2, C	-

¹H (400 MHz) and ¹³C (100 MHz) in DMSO-*d*₆. Assignments based on COSY, HSQC and HMBC and comparison with literature compounds. Chemical shifts (δ) in ppm. s: singlet; br s: broad singlet; d: doublet; br d: broad doublet; t: triplet, m: multiplet. One proton unless otherwise stated.

Table S2. ^1H and ^{13}C NMR data of **5** and **6**.

Residue	Position	5		6	
		^{13}C	^1H , mult. ($J = \text{Hz}$)	^{13}C	^1H , mult. ($J = \text{Hz}$)
Proline (Pro)	α	59.8, CH	4.33, dd (8.6, 3.8)	59.8, CH	4.33, dd (8.8, 3.6)
	β	29.1, CH_2	2.08, m; 1.86, m	29.3, CH_2	1.87, m; 2.09, m
	γ	24.4, CH_2	1.92, (2H, m)	24.4, CH_2	1.92, (2H, m)
	δ	47.0, CH_2	3.77, (2H, m)	47.1, CH_2	3.77, (2H, m)
	CO	173.5, C	-	173.6, C	-
Serine (Ser)	α	53.0, CH	4.90, dd (12.0, 4.0)	53.0, CH	4.90, dd (12.0, 4.0)
	β	61.7, CH_2	3.76, m; 3.82, m	61.8, CH_2	3.80, m; 3.84, m
	CO	168.9	-	168.9, C	-
	NH	-	8.47, m	-	8.47, m
	OH	-	5.28, br t (6.0)	-	5.28, m
Thiazole A	2	167.3, C	-	167.3, C	-
	4	150.8, C	-	150.7, C	-
	5	127.6, CH	8.55, s	127.5, CH	8.55, s
	CO	160.1, C	-	160.1, C	-
Pyridine (Py)	2	150.3, C	-	150.2, C	-
	3	127.7, C	-	127.6, C	-
	4	141.3, CH	8.44, d (7.0)	141.2, CH	8.46, d (6.8)
	5	118.5, CH	8.40, d (7.0)	118.6, CH	8.41, d (6.8)
	6	150.1, C	-	150.0, C	-
Thiazole B	2	160.4, C	-	160.3, C	-
	4	153.1, C	-	153.2, C	-
	5	123.0, CH	8.30, s	123.0, CH	8.30, s
Thiazole C	2	170.9, C	-	171.0, C	-
	4	146.7, C	-	146.7, C	-
	5	116.3, CH	7.36, s	116.3, CH	7.36, s
Phenylserine (PheSer)	α	58.0, CH	5.24, m	58.1, CH	5.24, m
	β	73.5, CH	5.03, br t (4.0)	73.7, CH	5.01, dd (6.2, 5.0)
	1	141.6, CH	-	141.7, C	-
	2,6	126.6, CH	7.30, (2H, m)	126.6, CH	7.31, m
	3,5	127.8, CH	7.27, (2H, m)	127.8, CH	7.30, m
	4	127.8, CH	7.25, m	127.7, CH	7.25, m
	NH	-	9.01, d (8.3)	-	9.01, d (7.8)
	OH	-	6.02, d (4.5)	-	6.03, d (4.6)
Glycine (Gly)	α	41.4, CH_2	3.87, m; 4.21, dd (16.6, 7.9)	41.1, CH_2	3.79, m; 4.26, dd (17.5, 8.6)
	CO	169.3, C	-	169.3, C	-
	NH	-	8.55, m	-	8.44, m
Thiazole D	2	168.4, C	-	165.5, C	-
	4	148.3, C	-	143.6, C	-
	5	124.5, CH	8.28, s	140.8, C	-
	5- CH_2OMe	-	-	67.3, CH_2	4.98, s
	CO	161.0, C	-	58.5, CH_3	3.38, s

	α	55.4, CH	5.24, m	55.2, CH	5.20, m
	β	34.0, CH	2.20, m	34.0, CH	2.18, m
Valine (Val)	γ	18.4, CH ₃	0.86, d (8.0)	18.3, CH ₃	0.85, d (6.8)
	γ'	17.7, CH ₃	0.88, d (8.0)	17.8, CH ₃	0.88, d (6.8)
	NH	-	8.73, d (7.8)		8.67, d (8.2)
	2	167.9, C	-	168.3, C	-
	4	141.7, C	-	141.9, C	-
Thiazole E	5	139.6, C	-	139.3, C	-
	5-Me	11.8, CH ₃	2.60, s	11.8, CH ₃	2.59, s
	CO	160.3, C	-	159.8, C	-
	α	47.9, CH	5.37, m	47.9, CH	5.31, m
	β	37.6, CH ₂	1.50, m; 2.73, dd (16.4, 3.7)	37.5, CH ₂	1.37, m; 2.72, dd (16.4, 3.2)
Asparagine (Asn)	CONHMe	169.6, C	-	169.6, C	-
	NHMe	-	7.48, m	-	7.38, m
	Me	25.6, CH ₃	2.46, d (4.6)	25.7, CH ₃	2.47, d (4.5)
	NH	-	8.77, d (9.0)	-	8.70, d (8.7)
	2	164.7, C	-	164.7, C	-
	4	149.3, C	-	149.3, C	-
Thiazole F	5	126.8, CH	8.60, s	126.9, CH	8.61, s
	CO	159.8, C	-	161.1, C	-

¹H (400 MHz) and ¹³C (100 MHz) in DMSO-*d*₆. Assignments based on COSY, HSQC and HMBC and comparison with literature compounds. Chemical shifts (δ) in ppm. s: singlet; br s: broad singlet; d: doublet; br d: broad doublet; t: triplet, m: multiplet. One proton unless otherwise stated.

Table S3. ^1H and ^{13}C NMR data of **7** and ^1H NMR data of **8** and **9**.

Residue	Position	7	8	9
		^{13}C	^1H , mult. ($J = \text{Hz}$)	^1H , mult. ($J = \text{Hz}$)
Thiazole A	2	168.1, C	-	-
	4	147.3, C	-	-
	5	132.3, CH	8.78, s	8.79, s
	CO	161.1, C	-	-
	COOMe	52.2 CH ₃	3.91, s	3.91, s
Pyridine (Py)	2	150.1, C	-	-
	3	127.6, C	-	-
	4	141.3, CH	8.41, d (8.2)	8.43, d (8.1)
	5	118.5, CH	8.30, d (8.2)	8.30, d (8.1)
	6	150.0, C	-	-
Thiazole B	2	160.4, C	-	-
	4	153.1, C	-	-
	5	123.0, CH	8.29, s	8.29, s
Thiazole C	2	170.8, C	-	-
	4	146.6, C	-	-
	5	116.3, CH	7.32, s	7.33, s
Phenylserine (PheSer)	α	58.3, CH	5.23, m	5.20, m
	β	73.2, CH	5.06, m	5.04, m
	1	141.6, C	-	-
	2,6	126.5, CH	7.26 (2H, m)	7.28 (2H, m)
	3,5	127.7, CH	7.25 (2H, m)	7.27 (2H, m)
	4	127.5, CH	7.21, m	7.22, m
Glycine (Gly)	NH	-	9.46, d (7.6)	9.45, d (6.2)
	OH	-	6.70, br s	6.65, br s
	α	41.3, CH ₂	3.89, dd (17.2, (17.2, 8.9)	3.80, dd (16.7, (16.7, 8.6)
Thiazole D	CO	169.4, C	-	3.6); 4.31, dd 8.45, m
	NH	-	8.57, m	4.31, dd 8.45, m
	2	168.3, C	-	-
	4	148.4, C	-	-
	5	124.5, CH	8.28, s	-
Valine (Val)	5-CH ₂	-	-	4.98, s
	5-	-	-	3.39, s
	CH ₂ OMe	-	-	3.39
	CO	161.1, C	-	-
	α	55.4, CH	5.24, m	5.24, m
Thiazole E	β	34.0, CH	2.19, m	2.18, m
	γ	18.4, CH ₃	0.86, d (6.8)	0.89, d (6.8)
	γ'	17.8, CH ₃	0.89, d (6.8)	0.85, d (6.8)
	NH	-	8.76, d (8.2)	0.85, d (6.8)
	2	167.9, C	-	-
Thiazole E	4	141.7, C	-	-
	5	139.6, C	-	-
	5-Me	11.8, CH ₃	2.60, s	2.59, s
	CO	160.5, C	-	-

	α	47.9, CH	5.34, m	5.29, m	5.30, dd (8.6, 4.6)
Asparagine (Asn)	β	37.6, CH ₂	1.43, m; 2.75, m	1.29, m; 2.72, m	1.34, m; 2.72, dd (16.0, 3.4)
	CONHMe	169.6, C	-	-	-
	NHMe	-	7.50, m	7.42, m	7.41, m
	Me	25.7, CH ₃	2.45, d (4.5)	2.46, d (4.6)	2.47, d (4.6)
	NH	-	8.73, d (9.4)	8.68, d (8.8)	8.67, br s
Thiazole F	2	164.6, C	-	-	-
	4	149.3, C	-	-	-
	5	126.9, CH	8.61, s	8.61, s	8.61, br s
	CO	160.2	-	-	-

¹H (400 MHz) and ¹³C (100 MHz) in DMSO-*d*₆. Assignments based on COSY, HSQC and HMBC and comparison with literature compounds. Chemical shifts (δ) in ppm. s: singlet; br s: broad singlet; d: doublet; br d: broad doublet; t: triplet, m: multiplet. One proton unless otherwise stated.

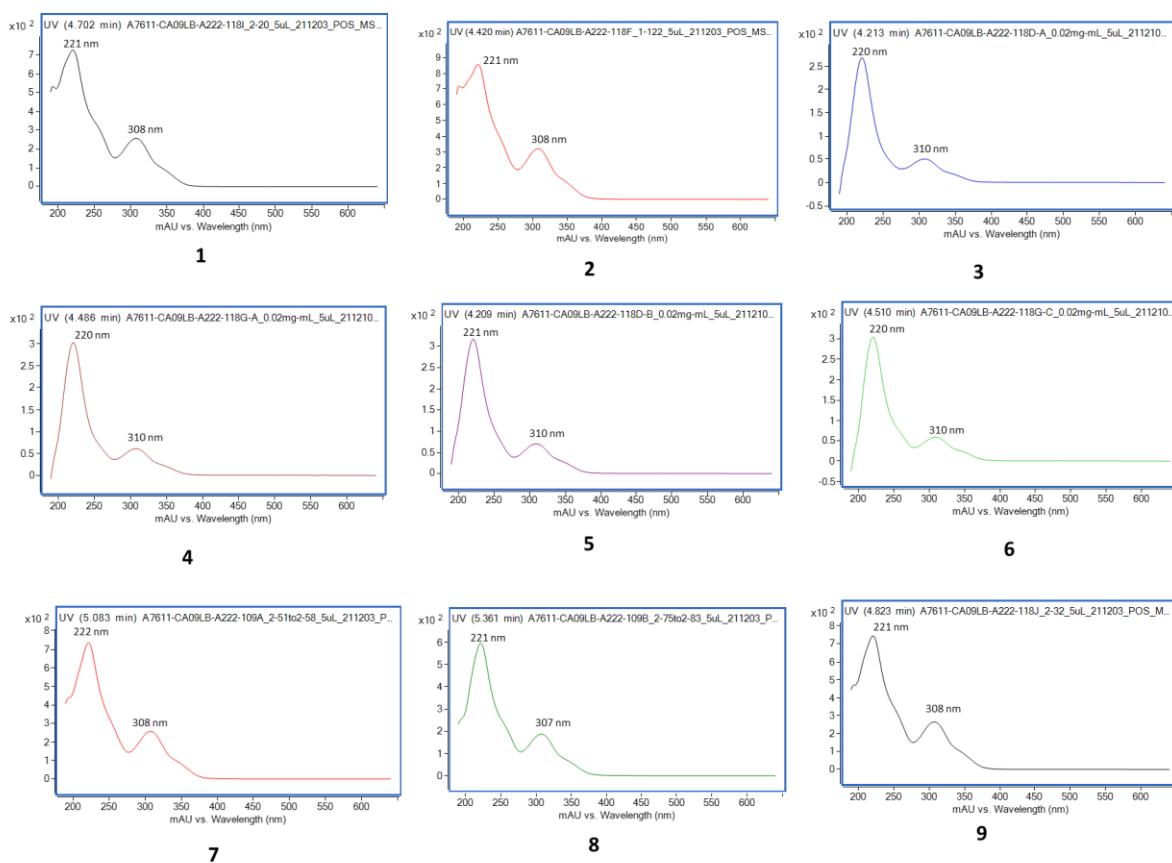


Figure S1. UV spectra for compounds **1–9**.

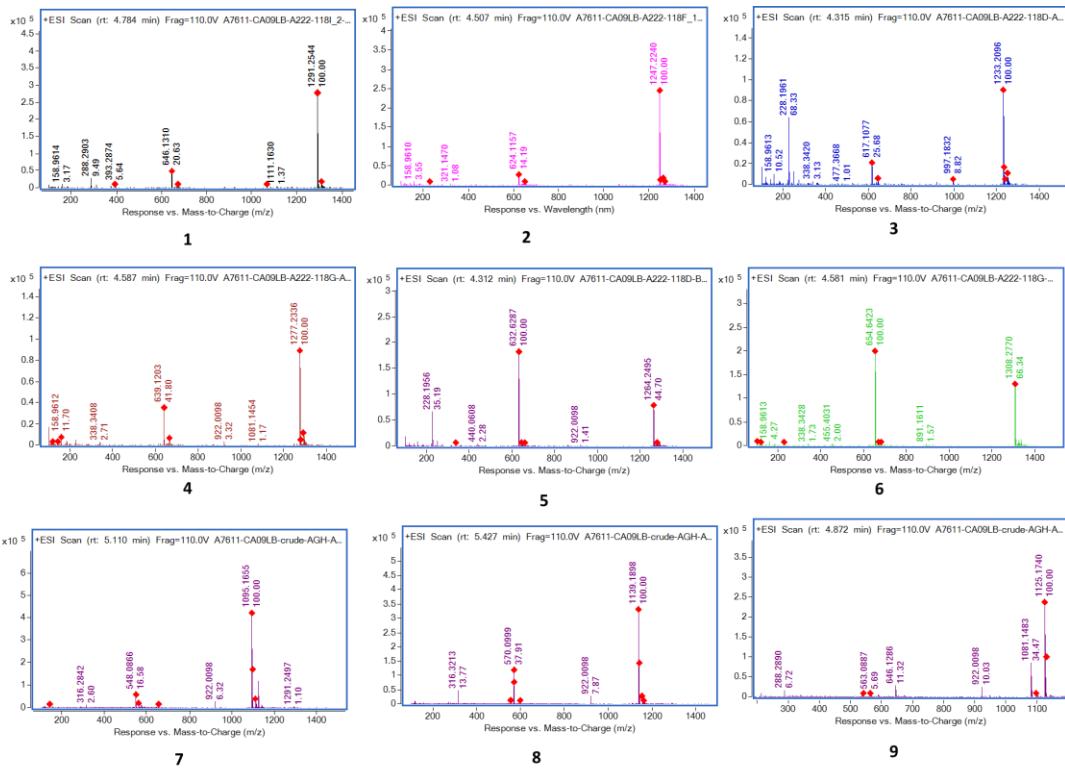


Figure S2. (+)-HRESIMS spectra for compounds **1–9**.

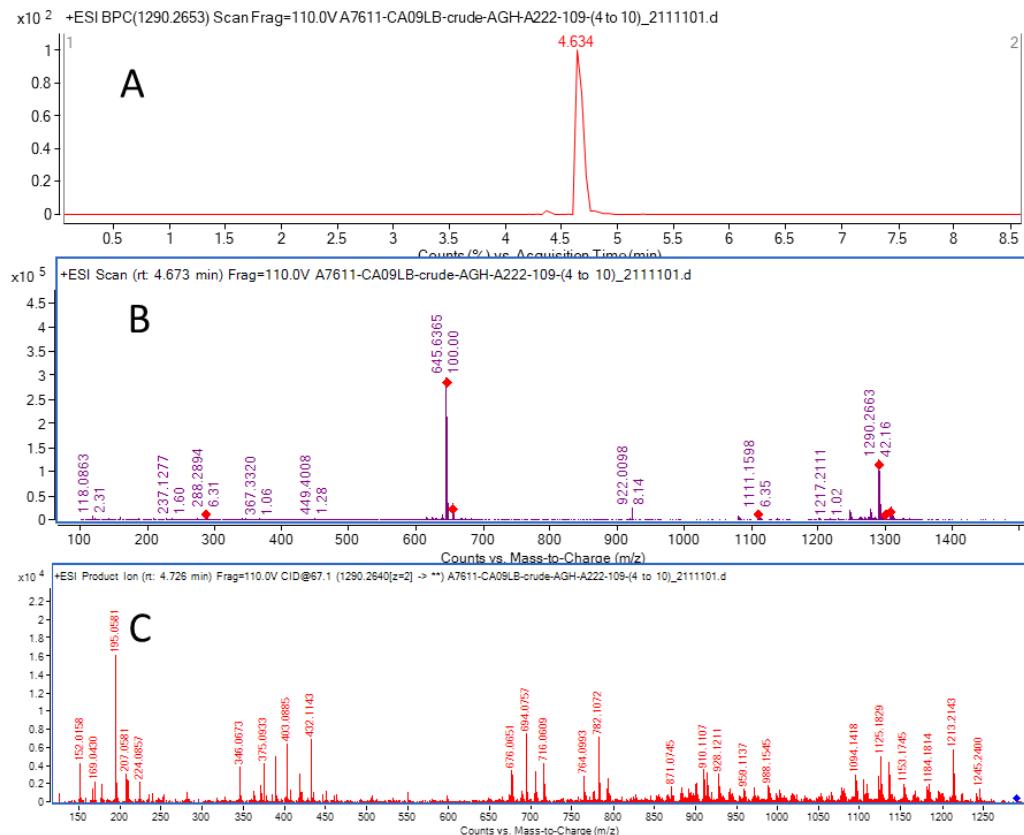


Figure S3. **A.** Extracted ion chromatogram (m/z 1290.2653 ± 5 ppm), **B.** (+)-HRESIMS spectrum, and **C.** MS/MS spectrum of GE2270A in the extract of *Nonomuraea jiangxiensis* strain A7611.

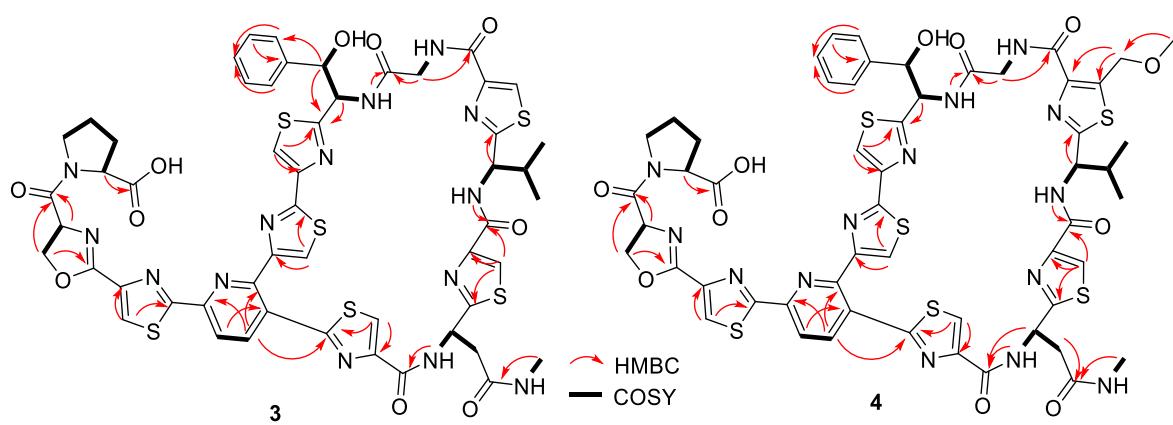


Figure S4. Selected COSY and HMBC correlations of **3** and **4**.

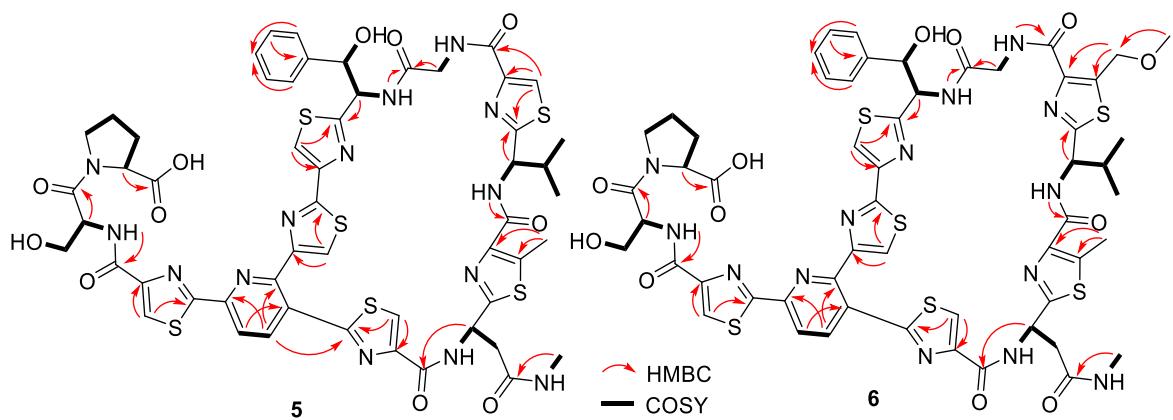


Figure S5. Selected COSY and HMBC correlations of **5** and **6**.

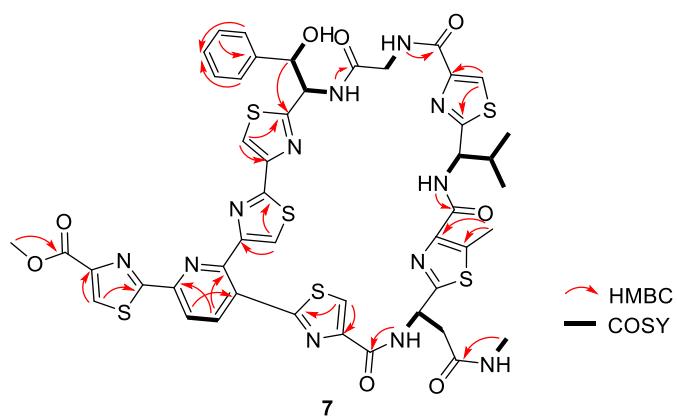


Figure S6. Selected COSY and HMBC correlations of **7**.

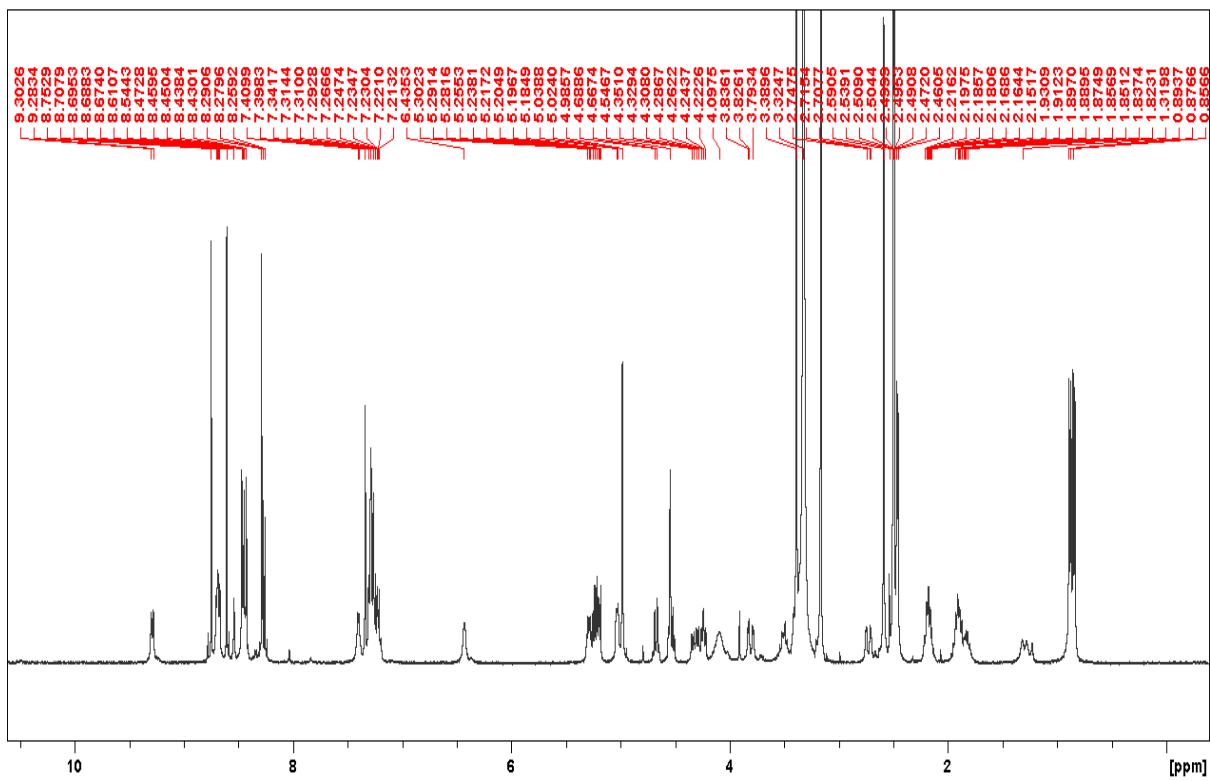


Figure S7. ¹H NMR spectrum (DMSO-*d*₆, 400 MHz) of **1**.

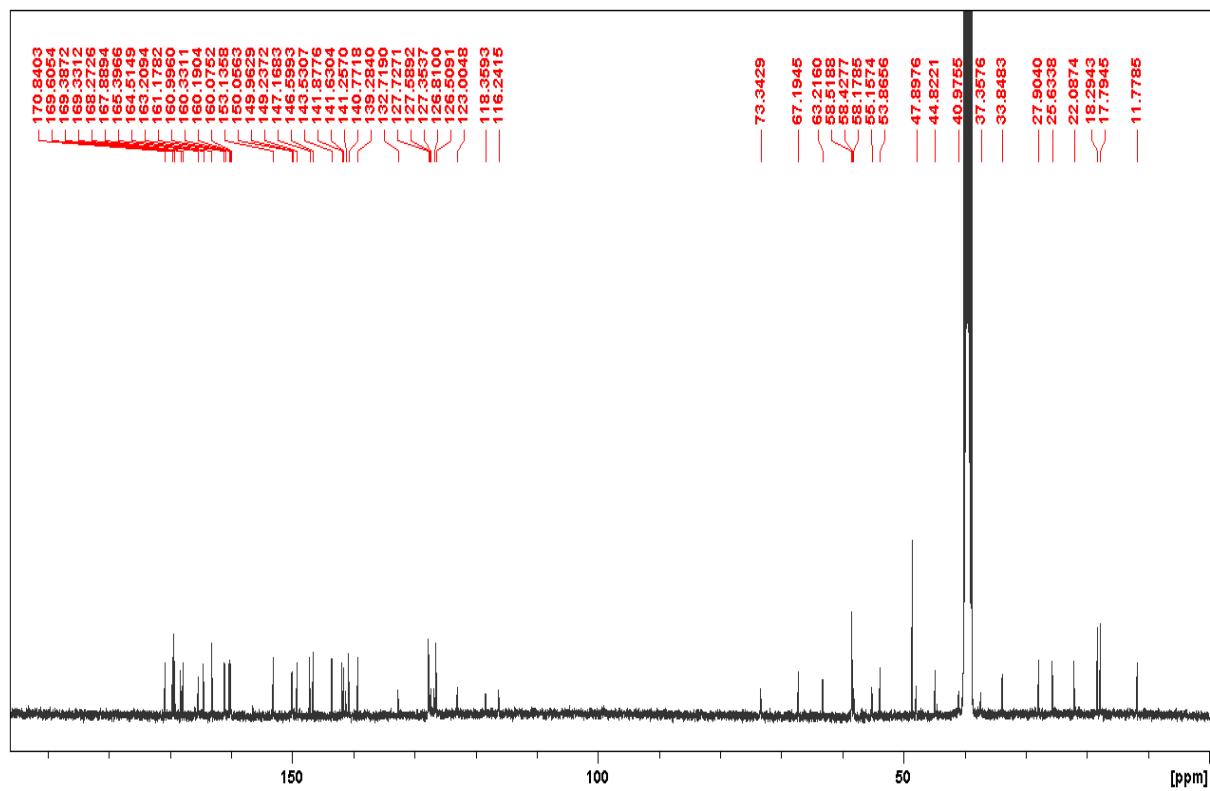


Figure S8. ¹³C NMR spectrum (DMSO-*d*₆, 100 MHz) of **1**.

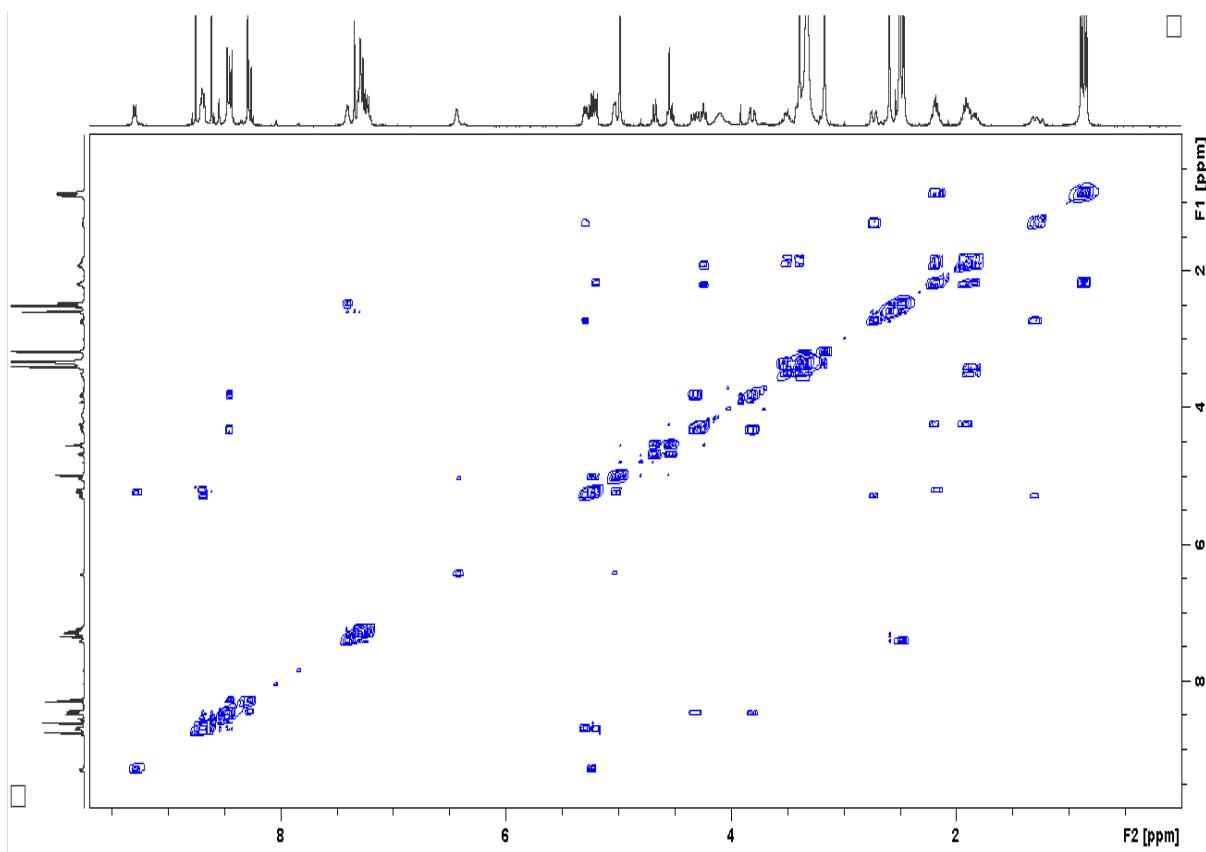


Figure S9. COSY spectrum of **1**.

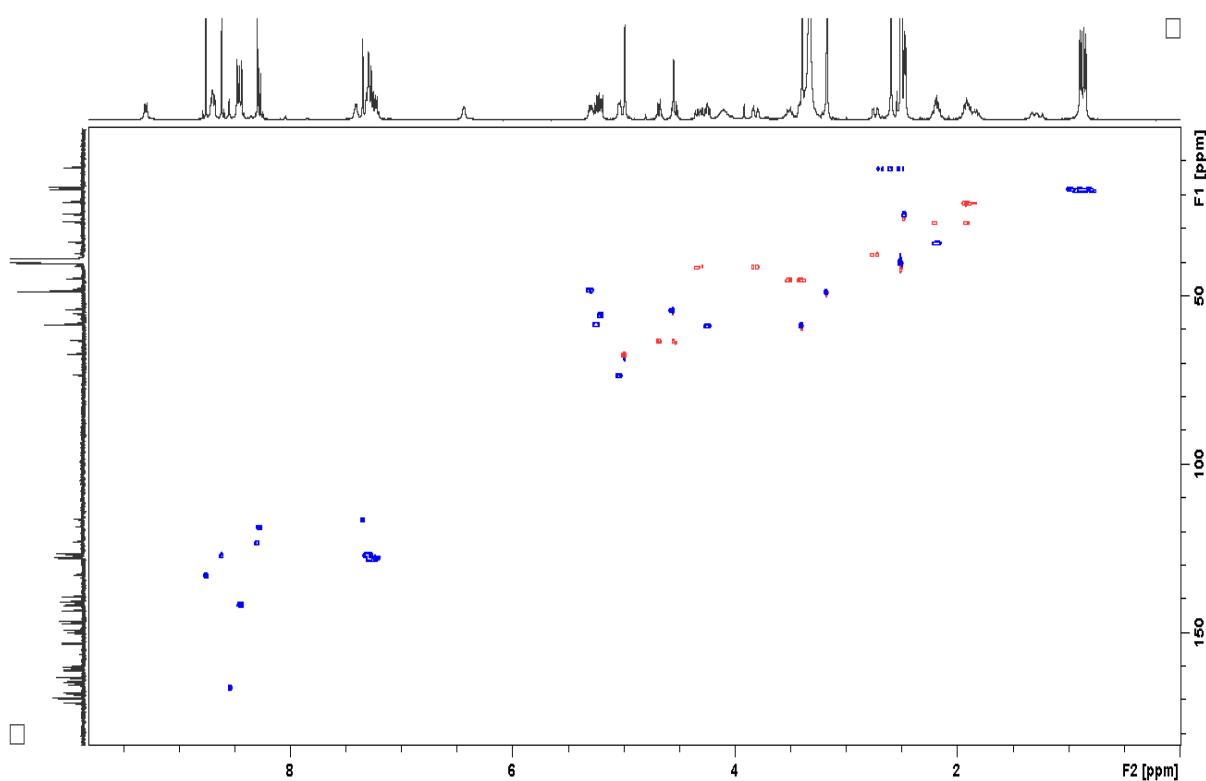


Figure S10. HSQC spectrum of **1**.

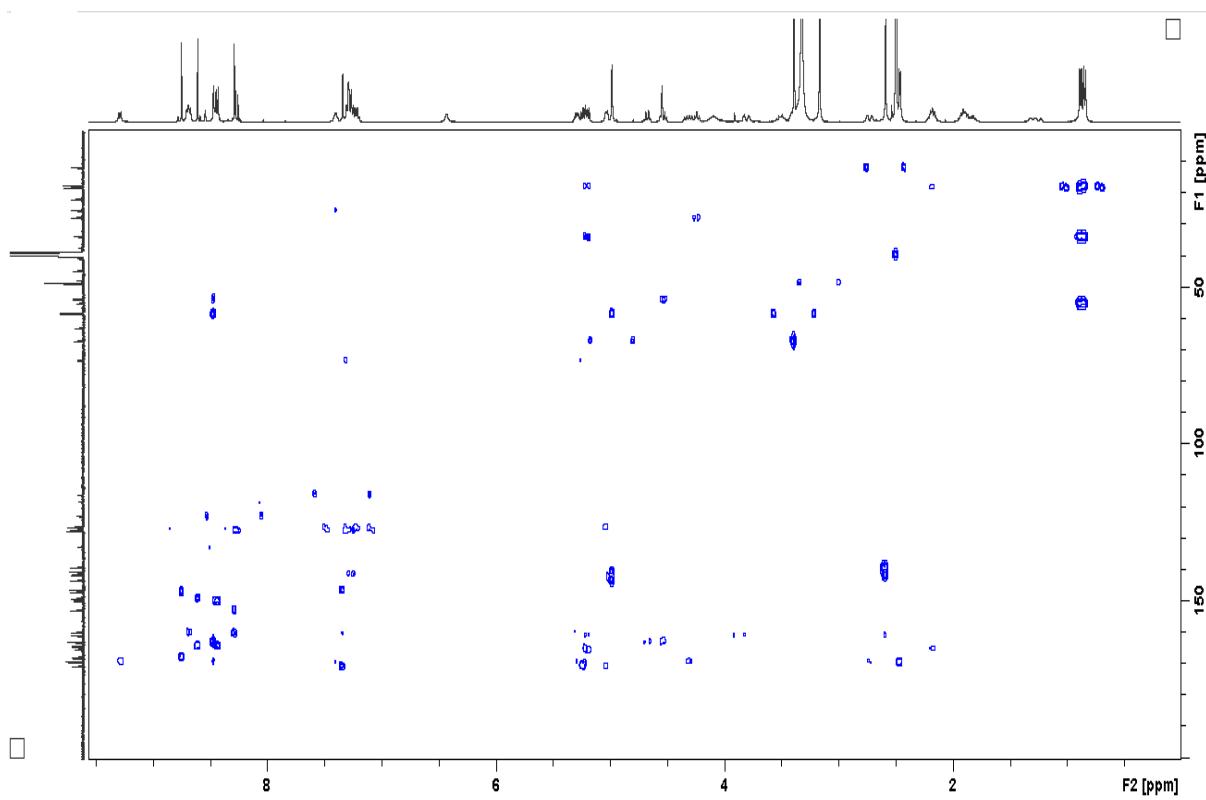


Figure S11. HMBC spectrum of **1**.

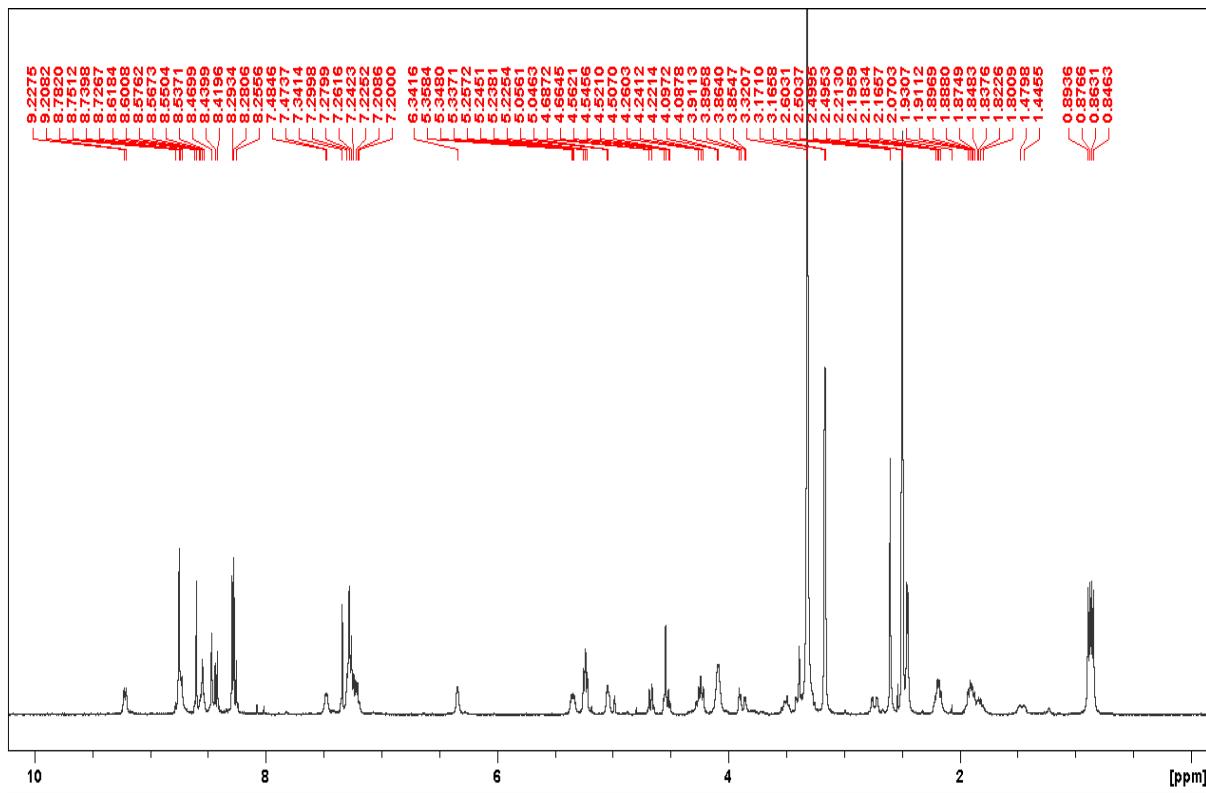


Figure S12. ^1H NMR spectrum ($\text{DMSO}-d_6$, 400 MHz) of **2**.

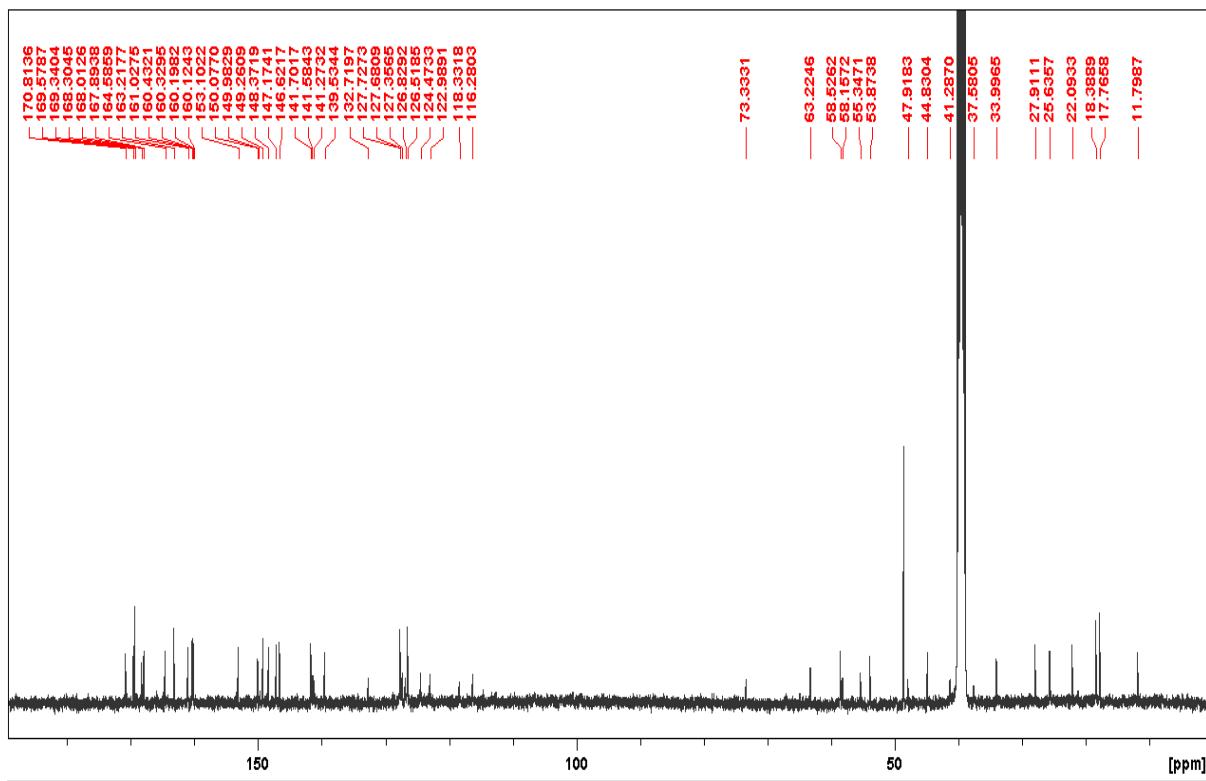


Figure S13. ^{13}C NMR spectrum (DMSO- d_6 , 100 MHz) of **2**.

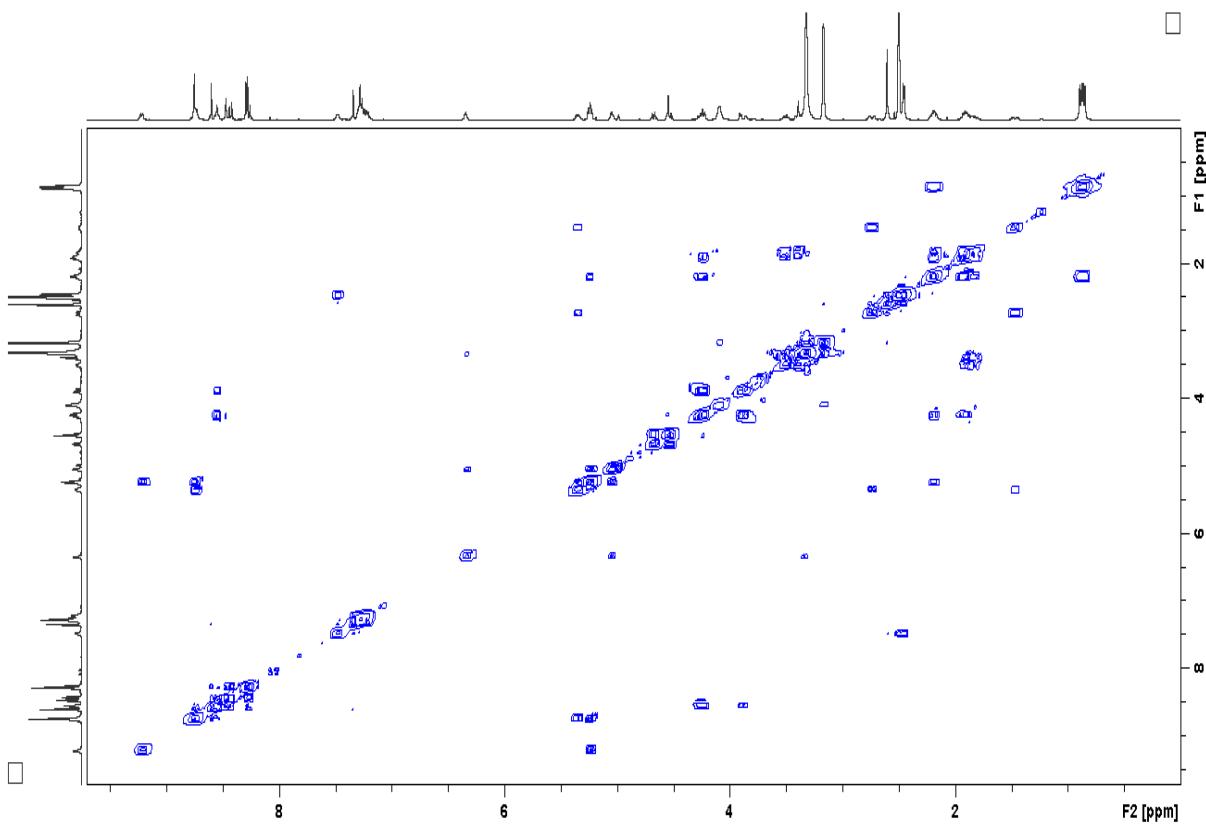


Figure S14. COSY spectrum of **2**.

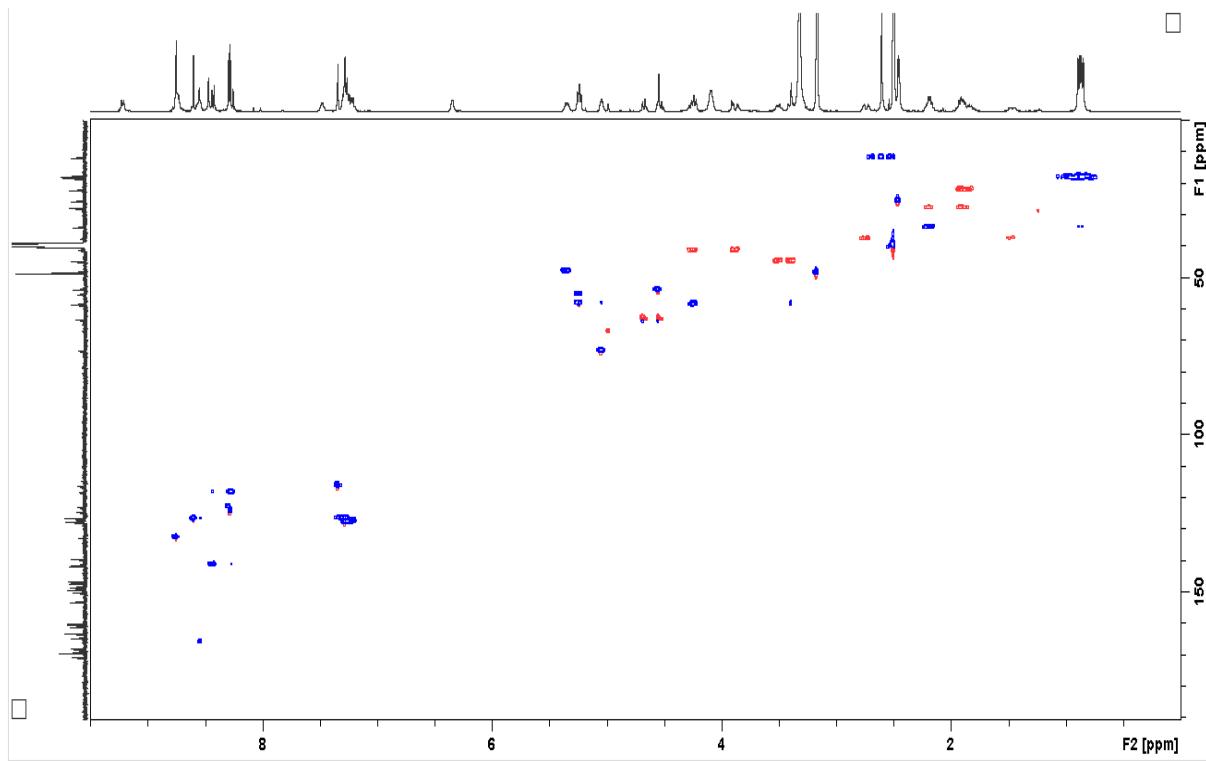


Figure S15. HSQC spectrum of **2**.

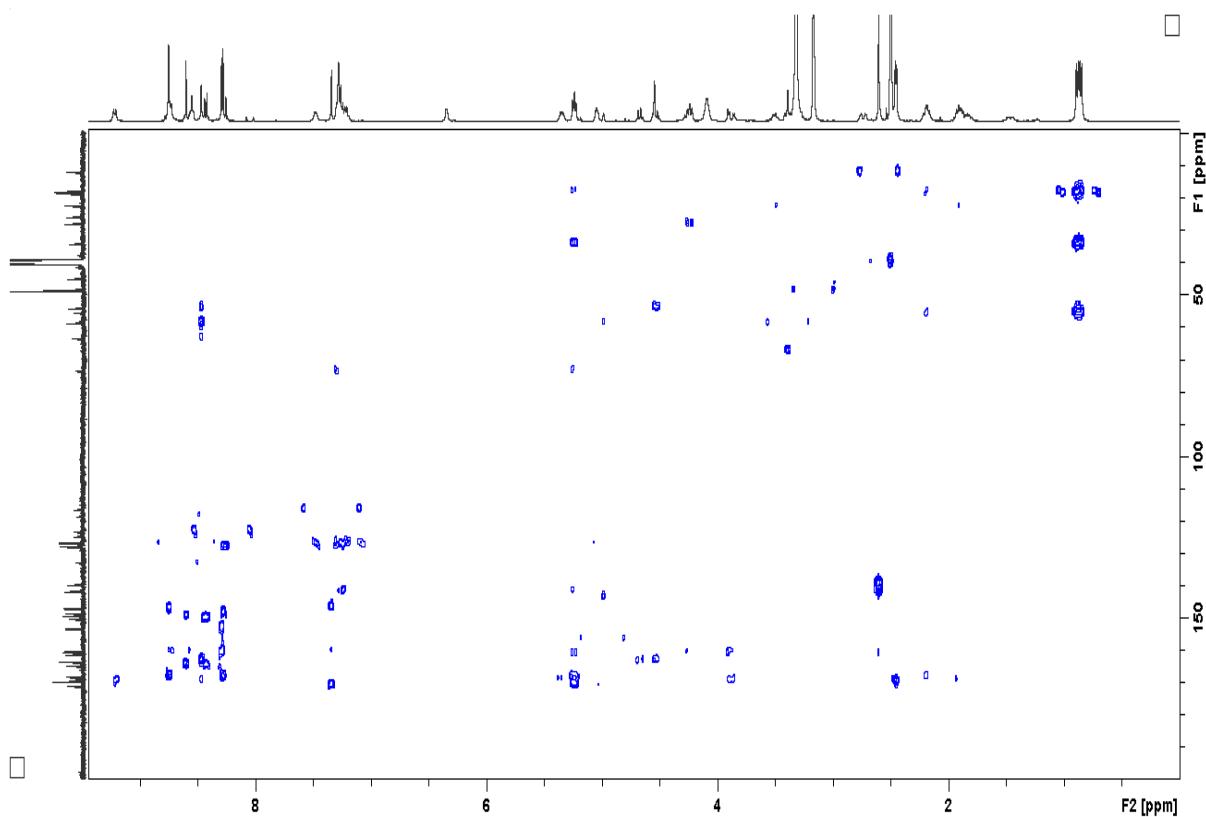


Figure S16. HMBC spectrum of **2**.

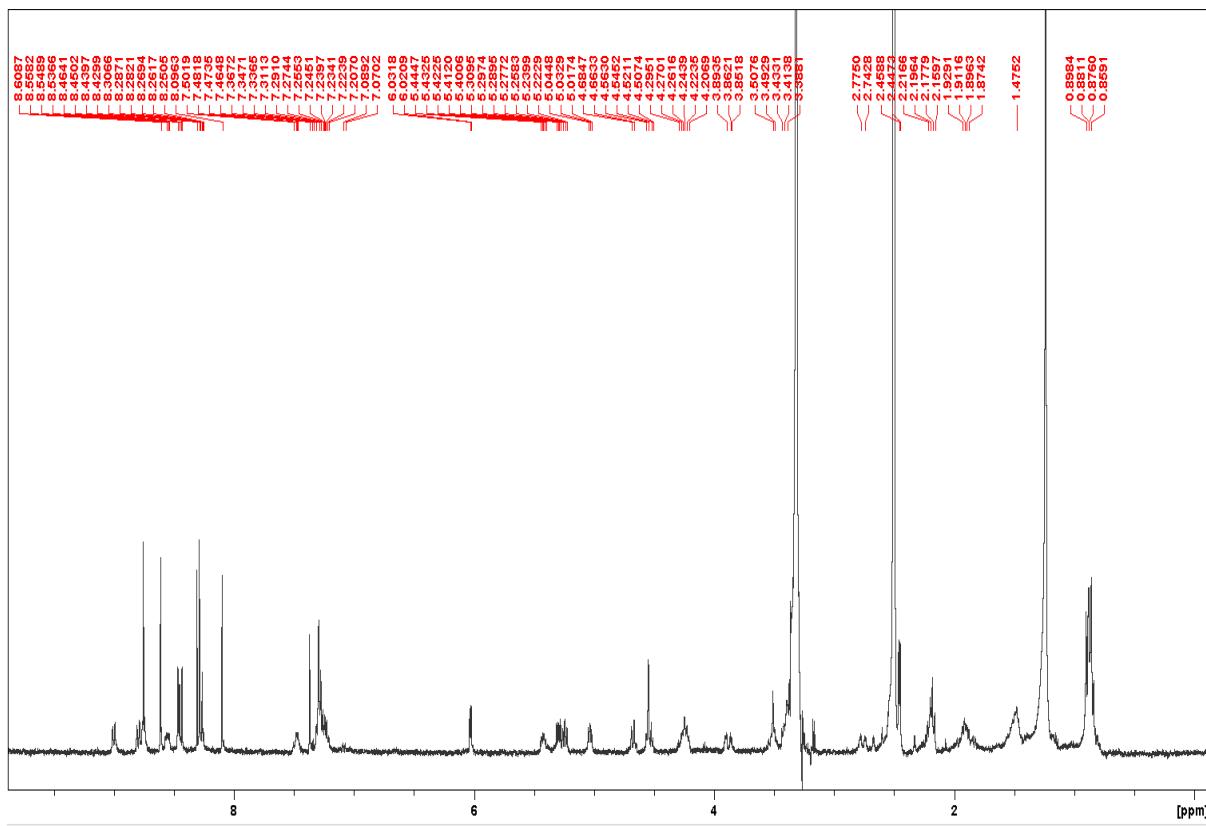


Figure S17. ^1H NMR spectrum (DMSO- d_6 , 400 MHz) of **3**.

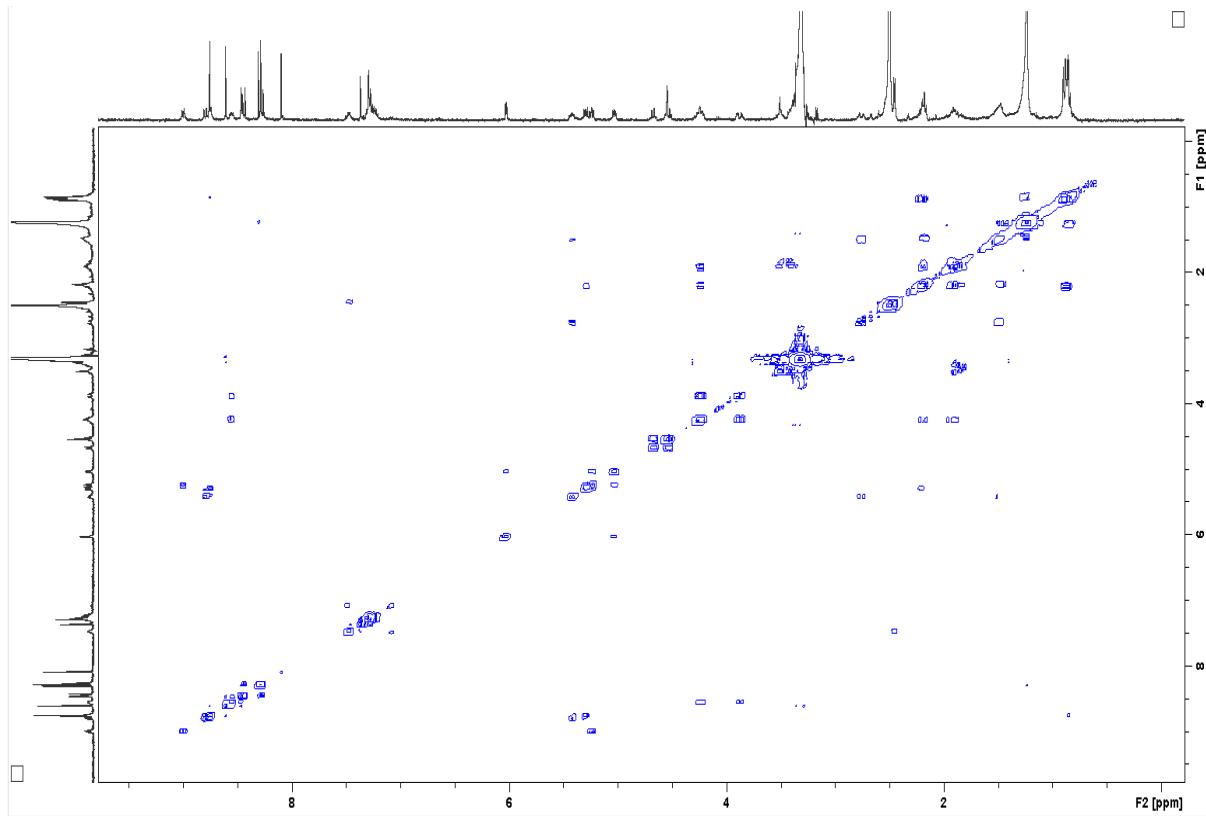


Figure S18. COSY spectrum of **3**.

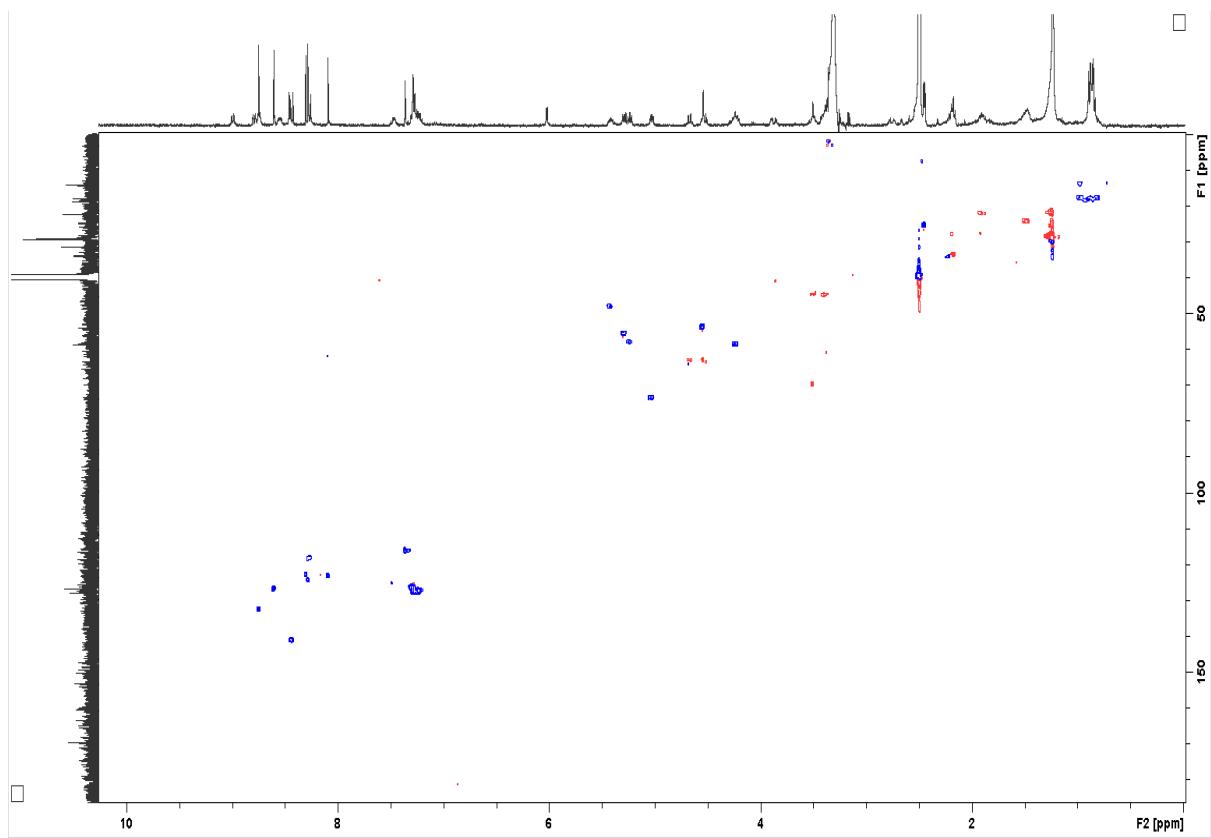


Figure S19. HSQC spectrum of **3**.

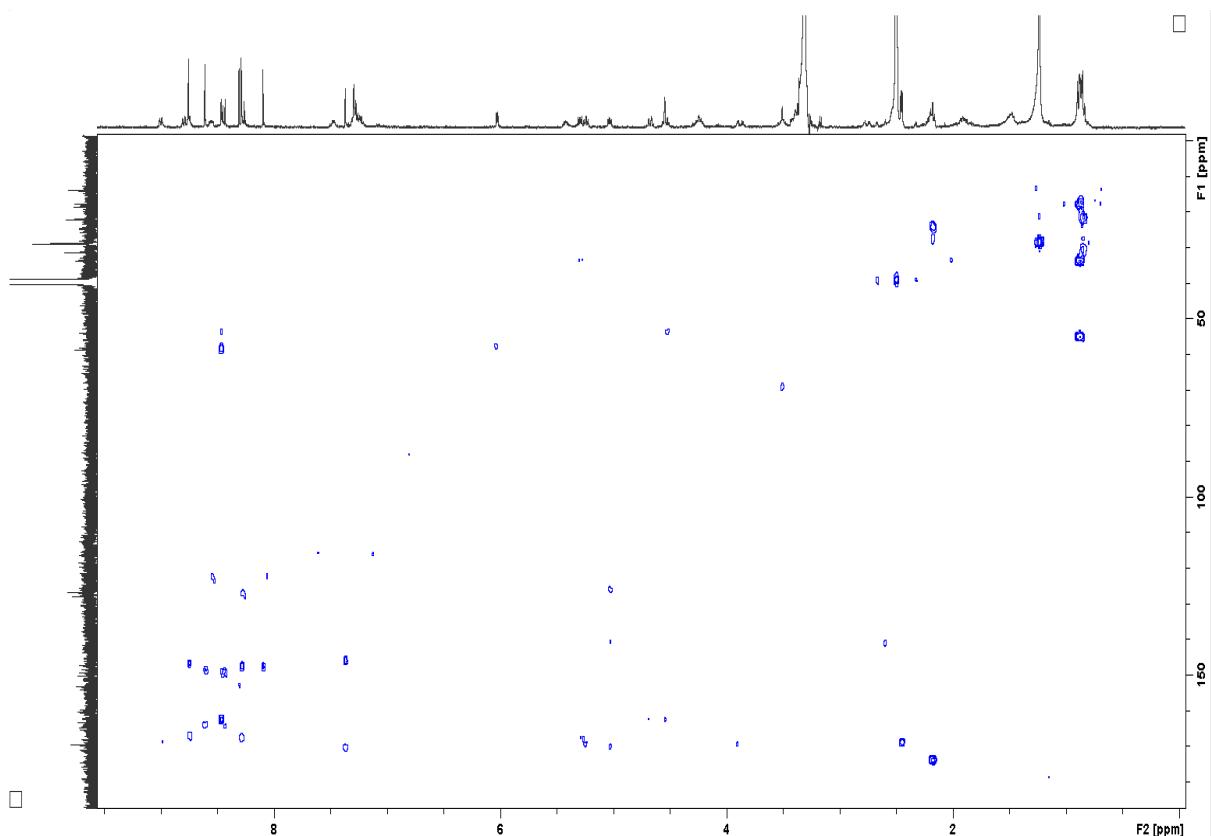


Figure S20. HMBC spectrum of **3**.

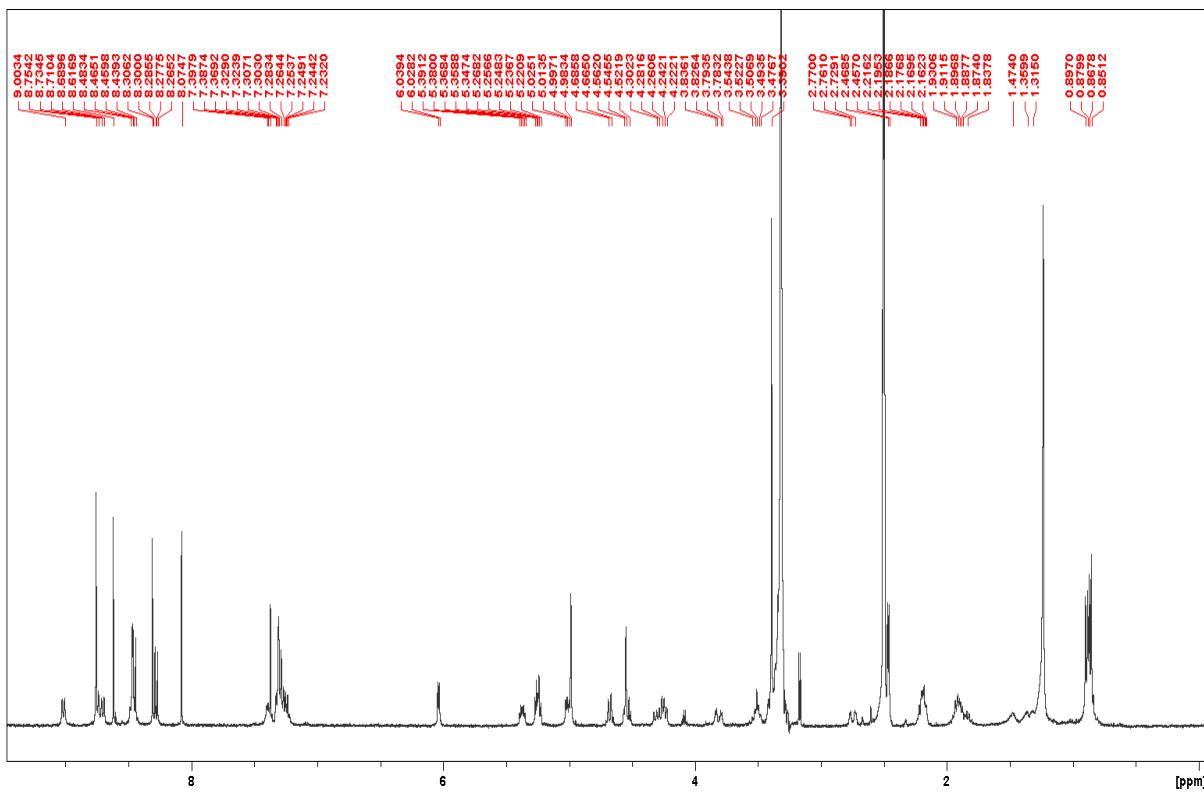


Figure S21. ^1H NMR spectrum (DMSO- d_6 , 400 MHz) of **4**.

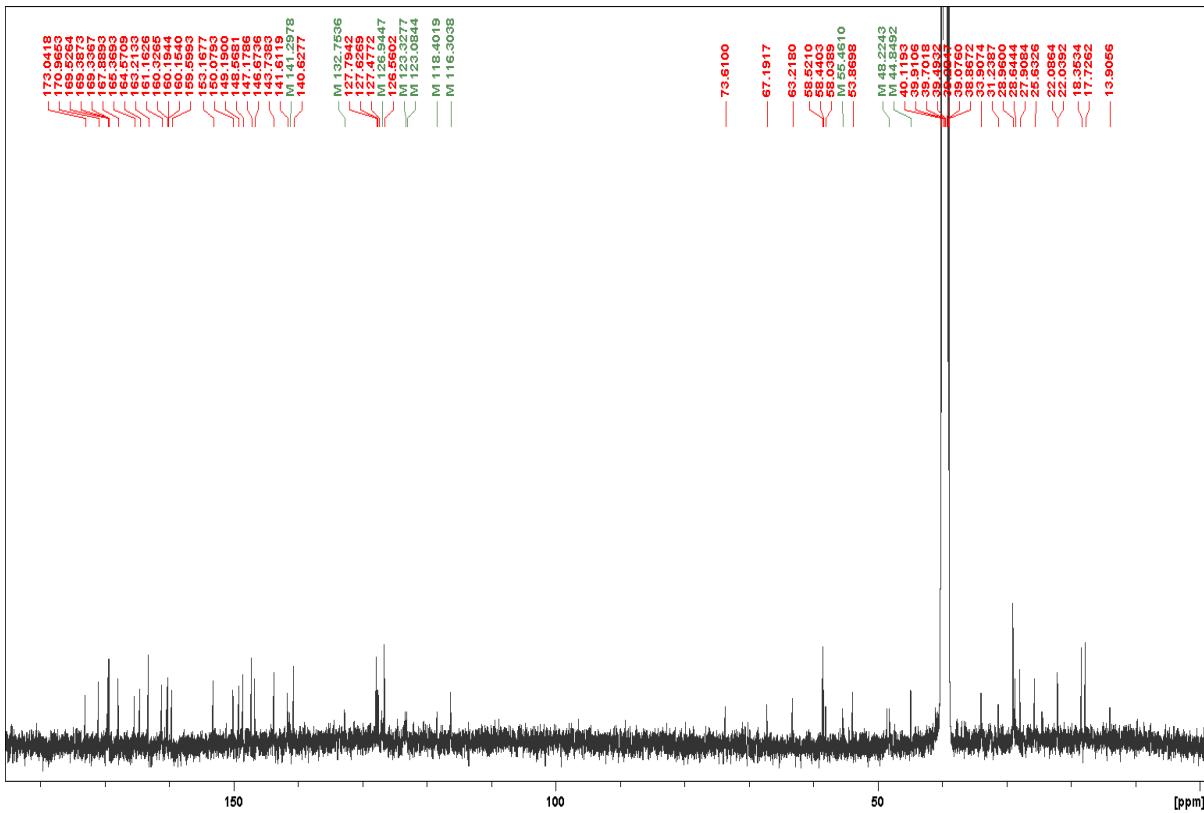


Figure S22. ^{13}C NMR spectrum (DMSO- d_6 , 100 MHz) of **4**.

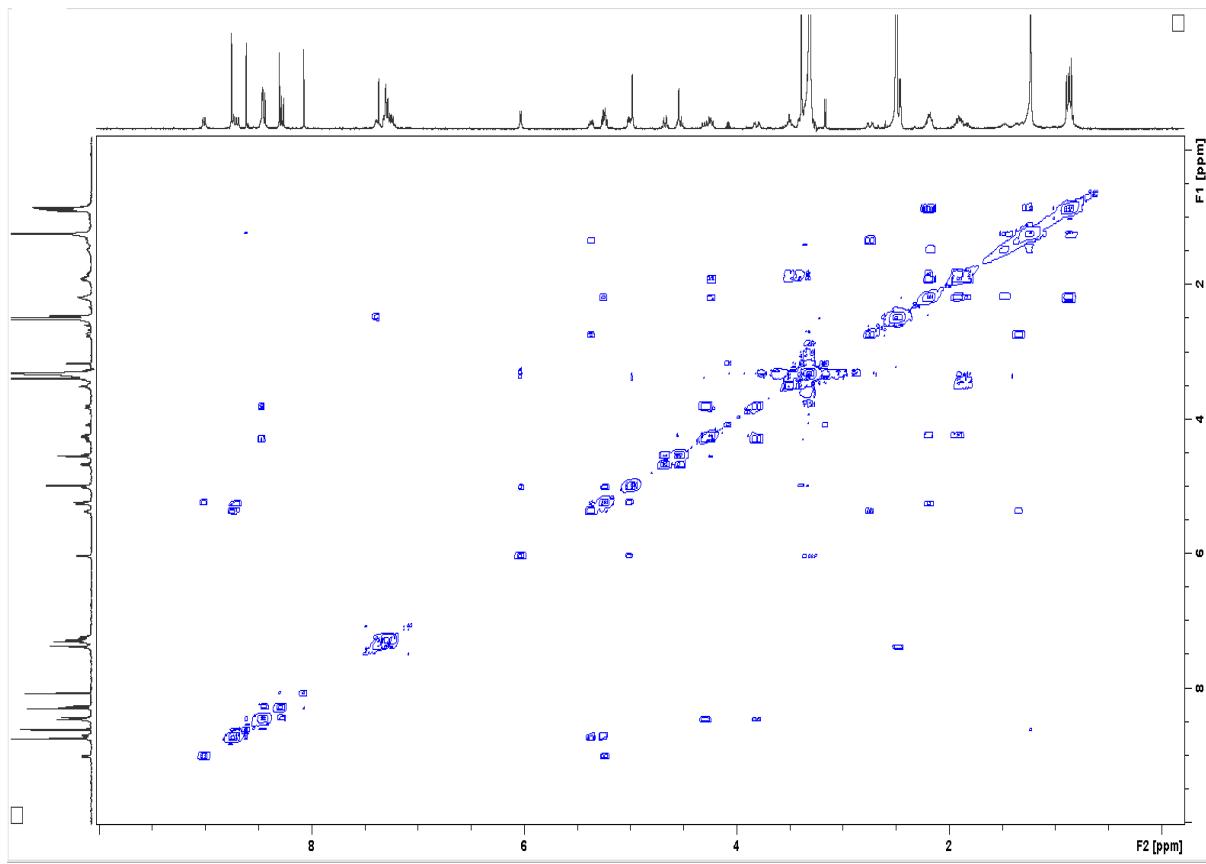


Figure S23. COSY spectrum of **4**.

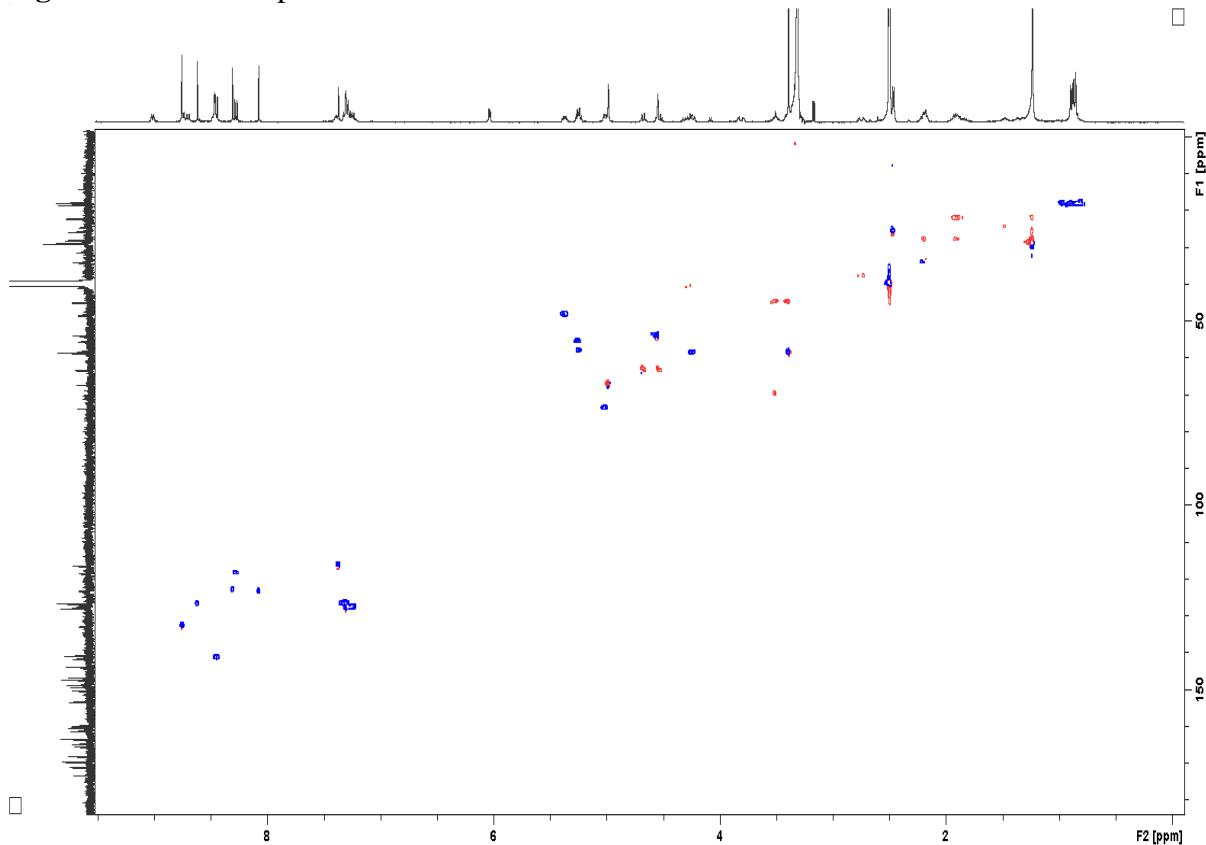


Figure S24. HSQC spectrum of **4**.

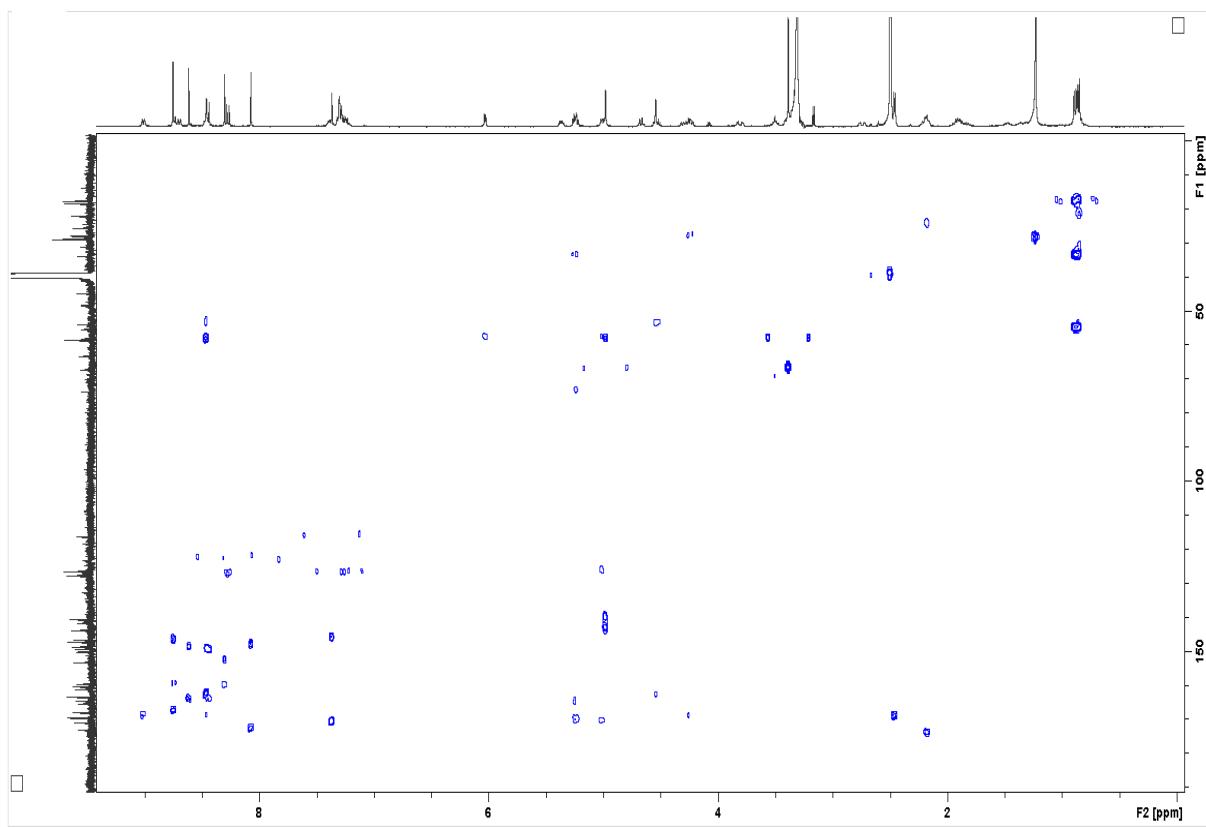


Figure S25. HMBC spectrum of **4**.

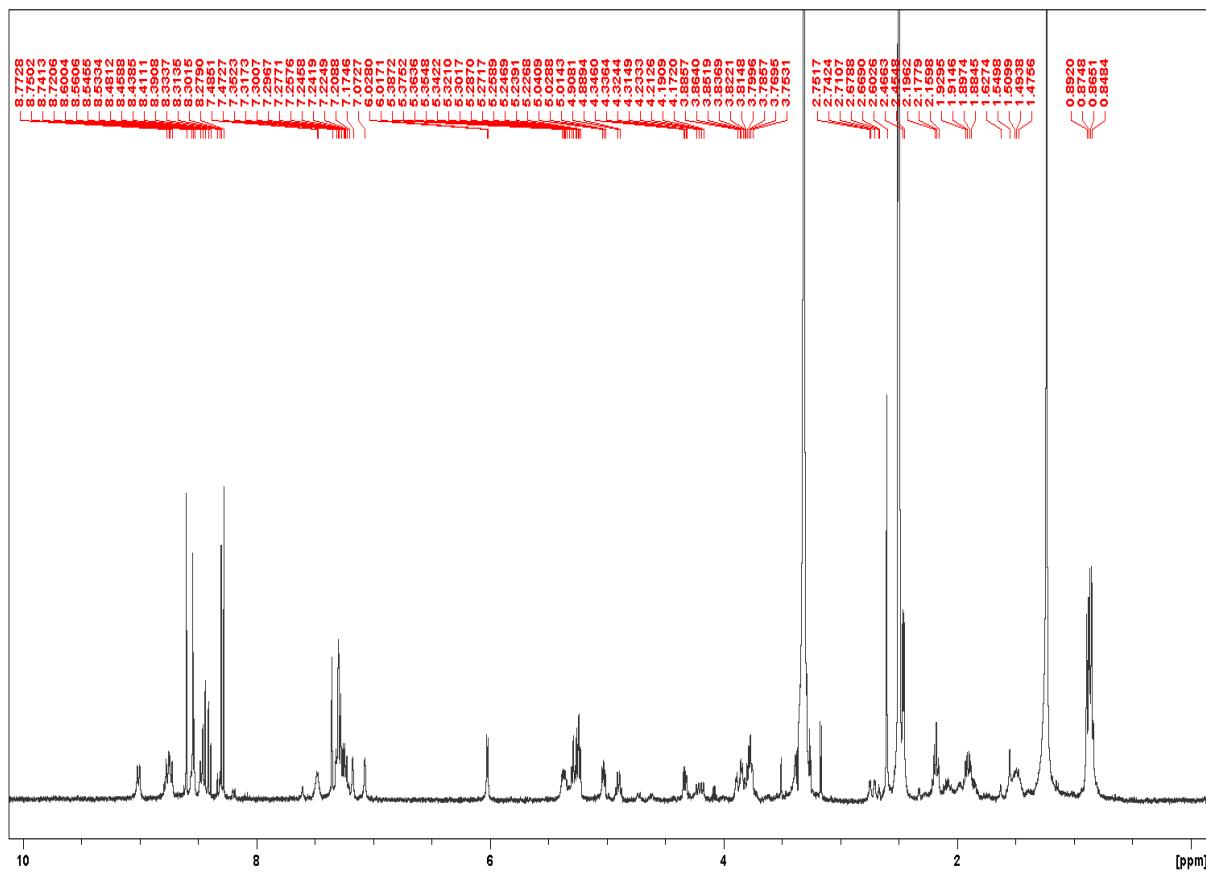


Figure S26. ^1H NMR spectrum ($\text{DMSO}-d_6$, 400 MHz) of **5**.

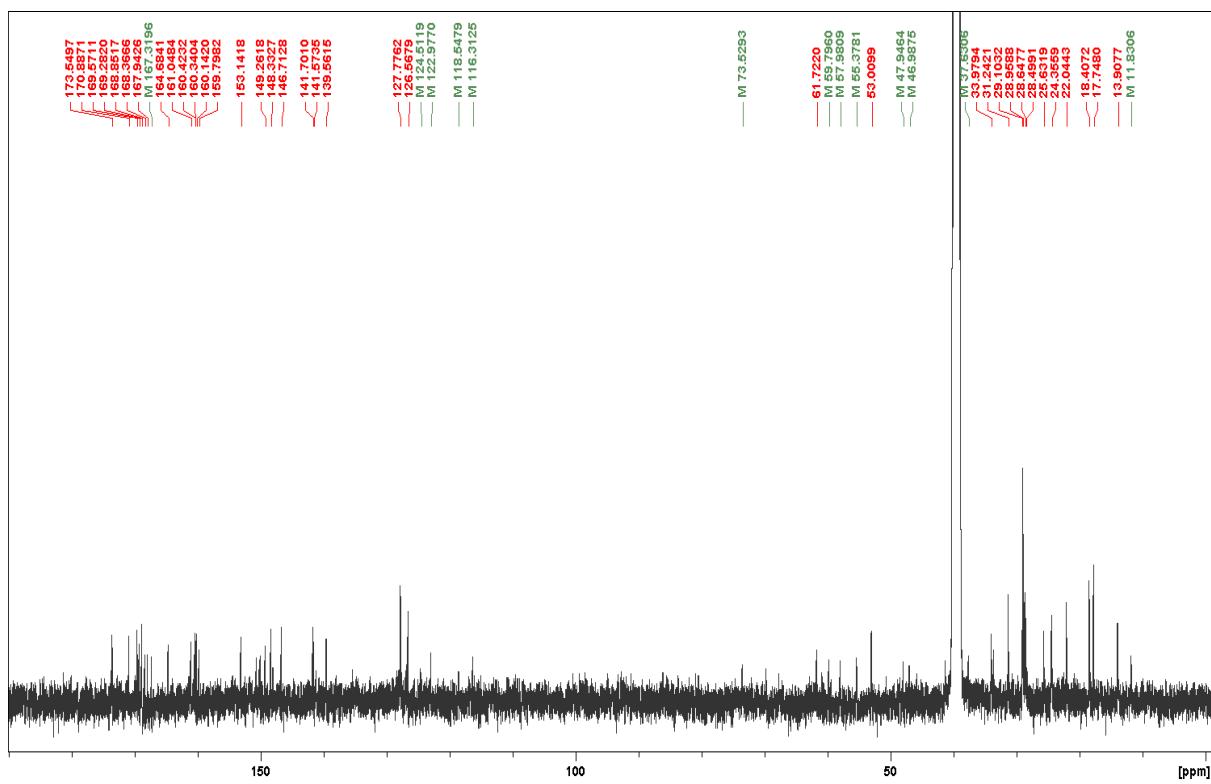


Figure S27. ^{13}C NMR spectrum (DMSO- d_6 , 100 MHz) of **5**.

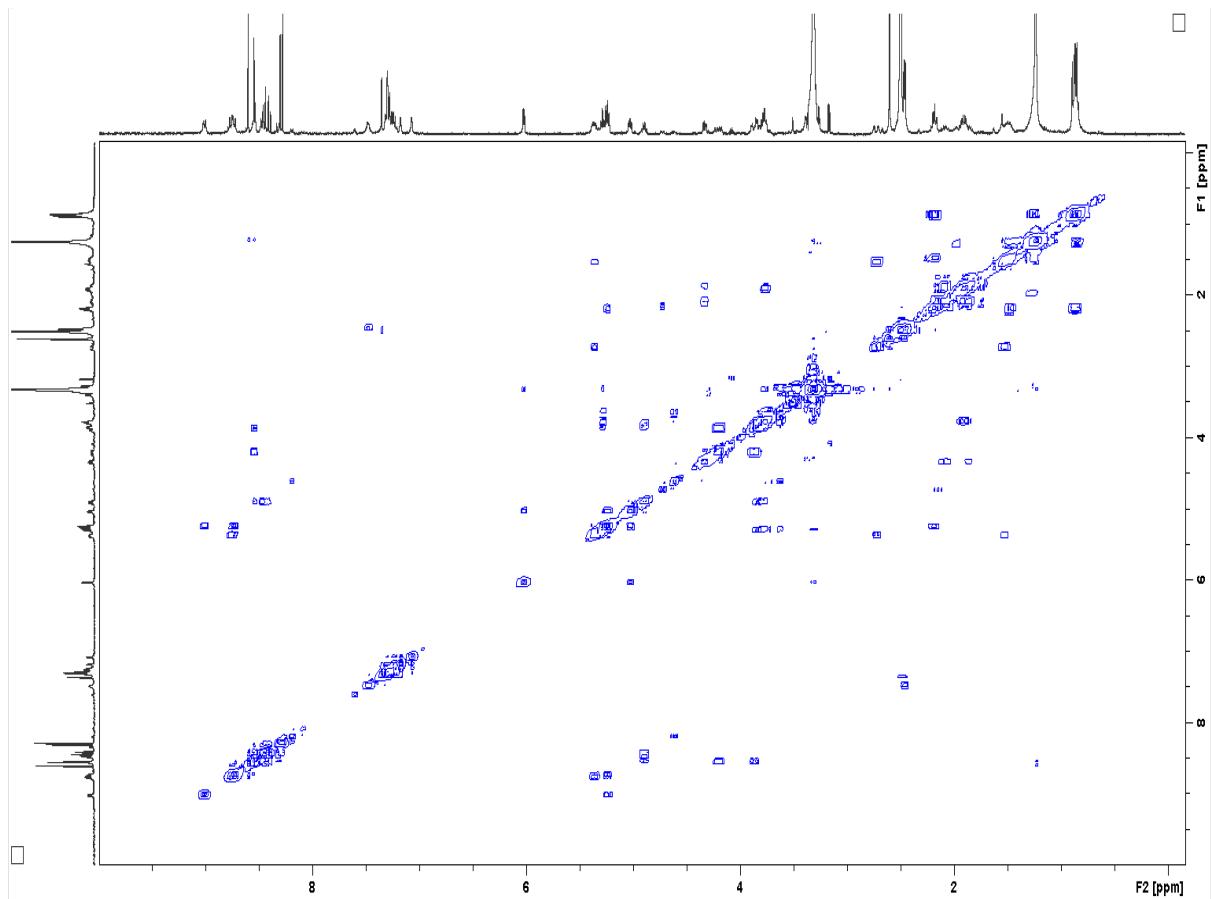


Figure S28. COSY spectrum of **5**.

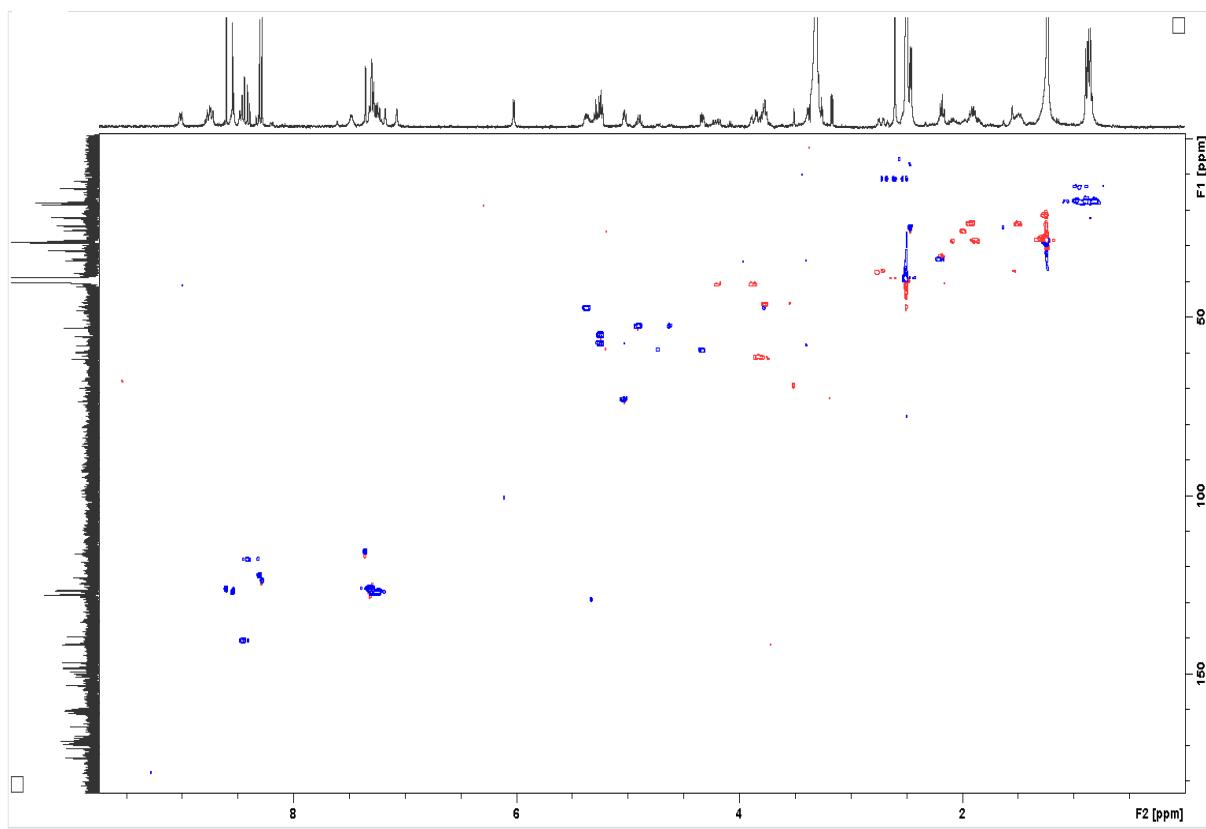


Figure S29. HSQC spectrum of **5**.

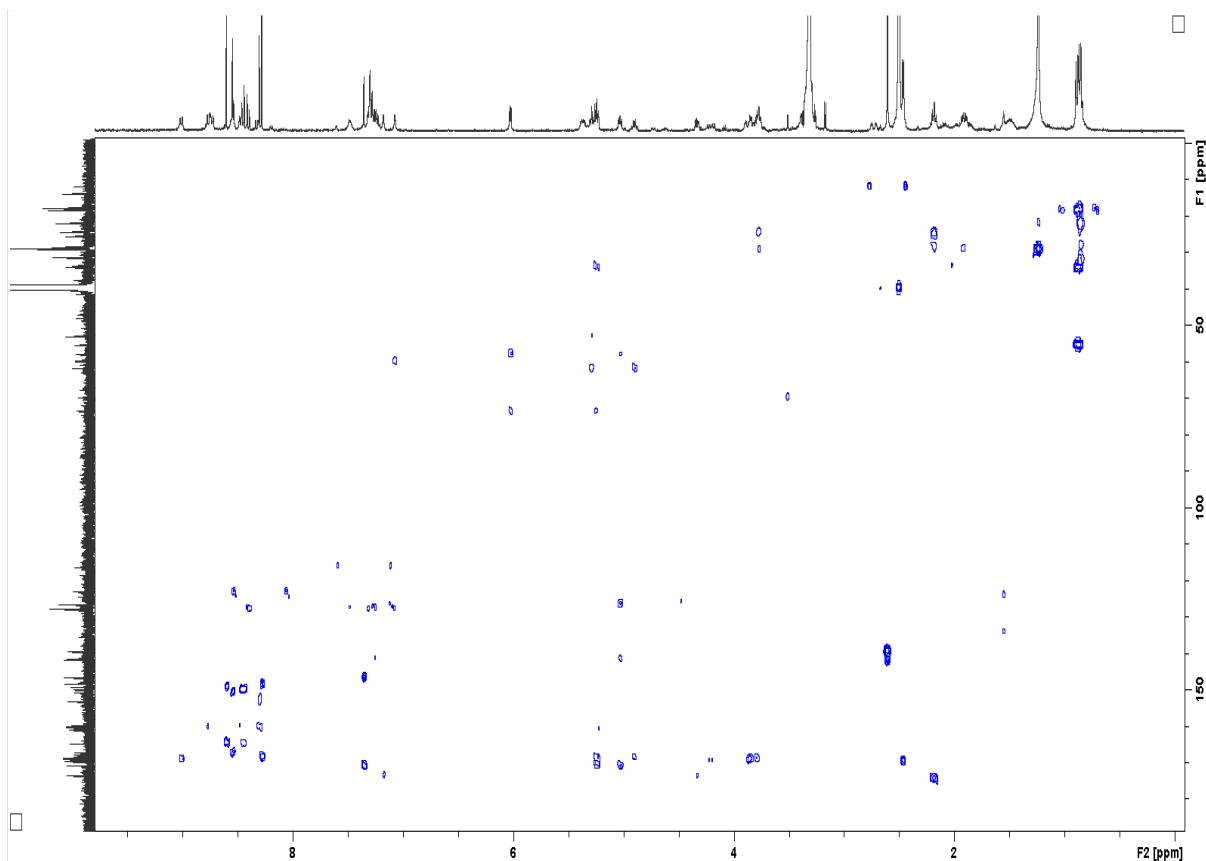


Figure S30. HMBC spectrum of **5**.

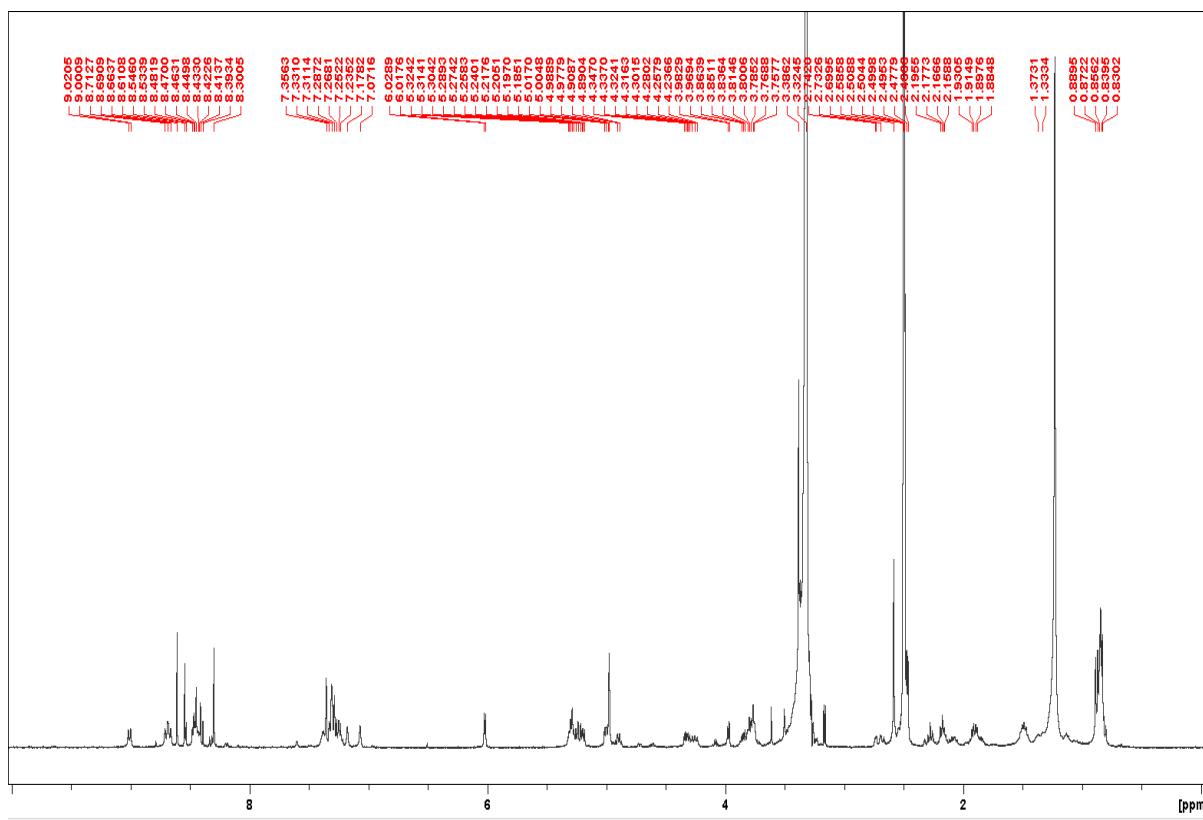


Figure S31. ^1H NMR spectrum (DMSO- d_6 , 400 MHz) of **6**.

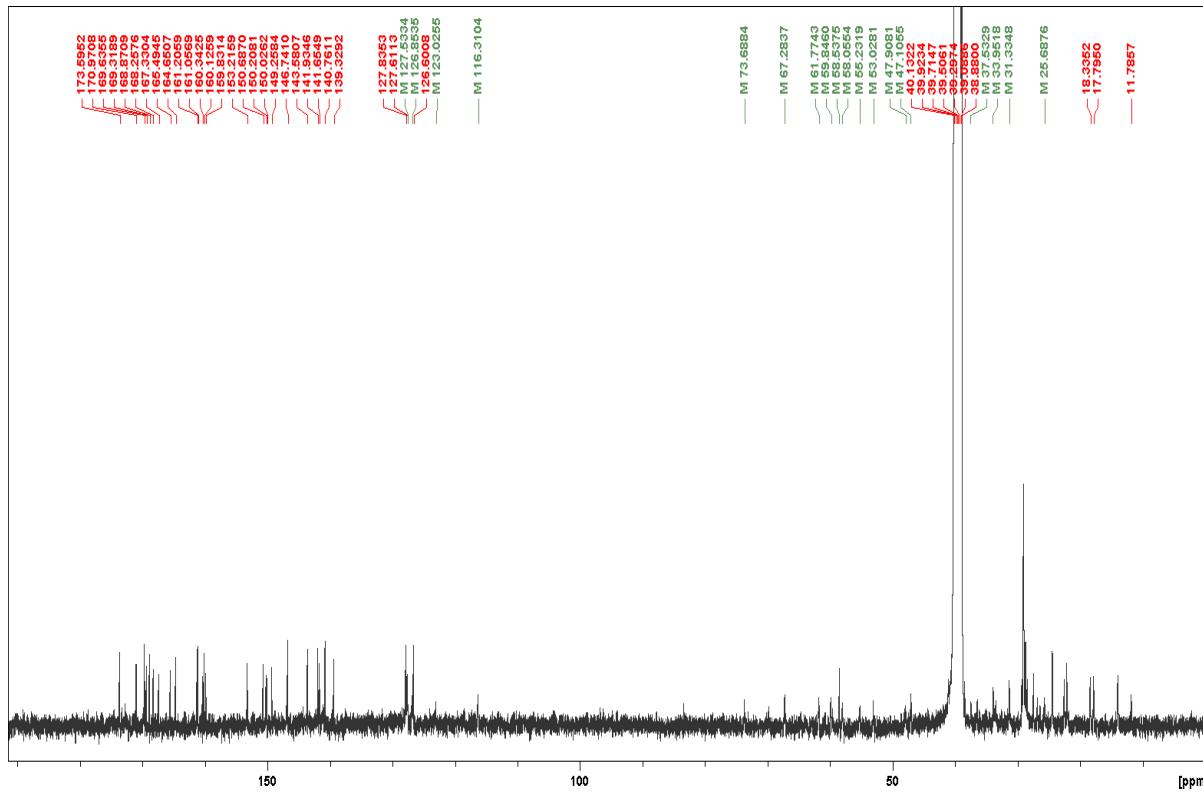


Figure S32. ^{13}C NMR spectrum (DMSO- d_6 , 100 MHz) of **6**.

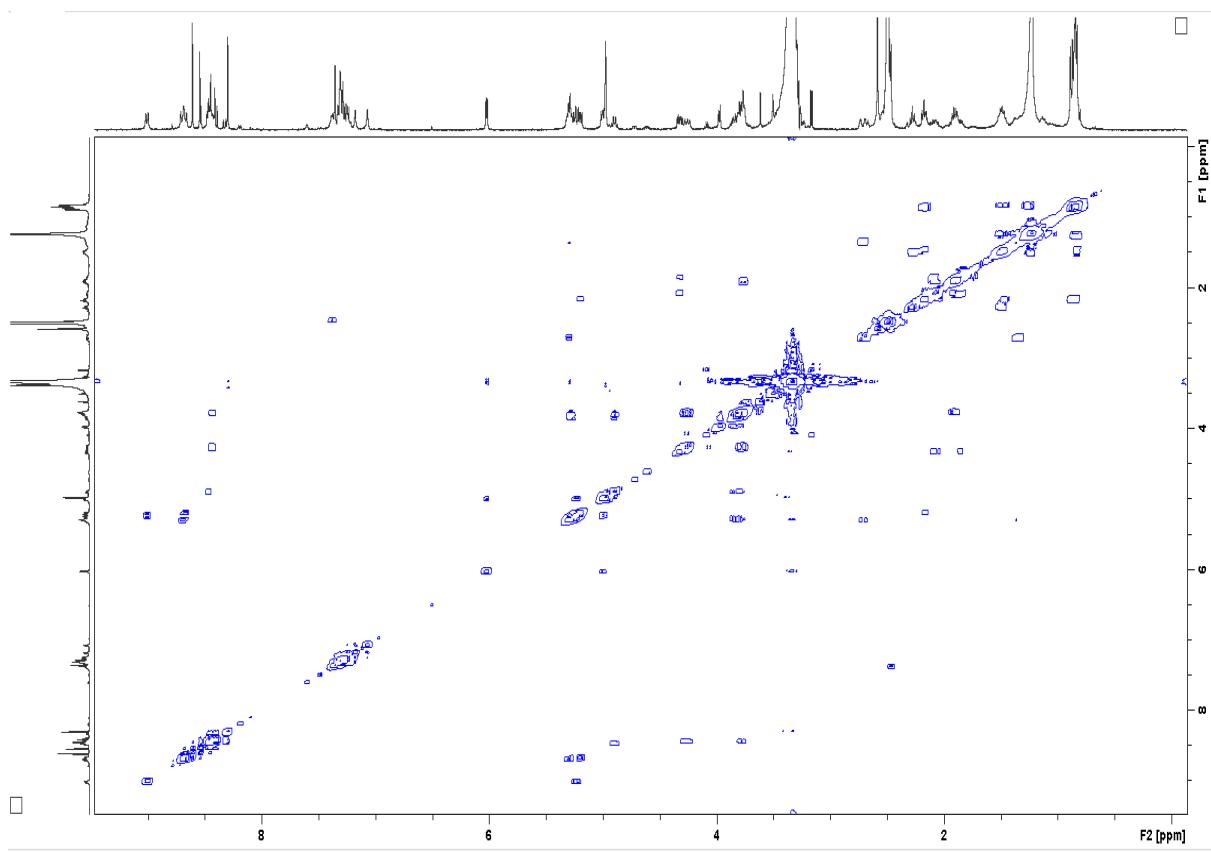


Figure S33. COSY spectrum of **6**.

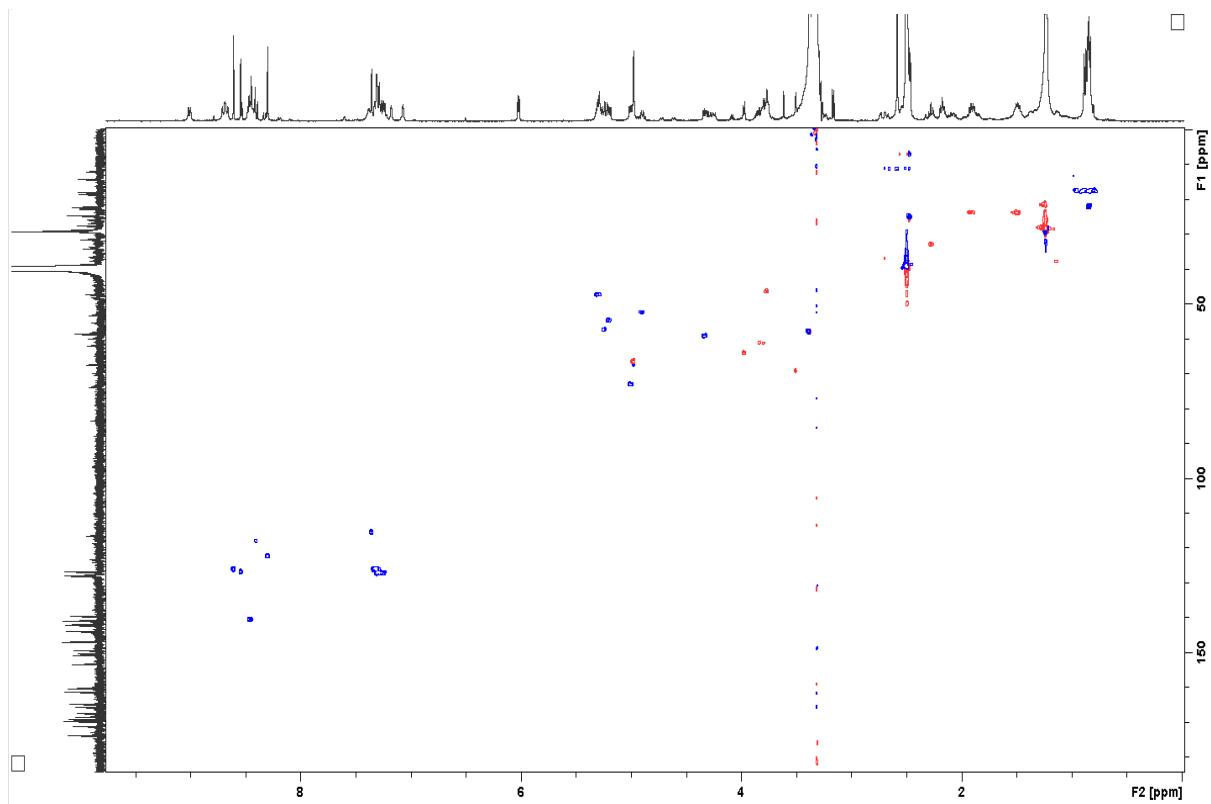


Figure S34. HSQC spectrum of **6**.

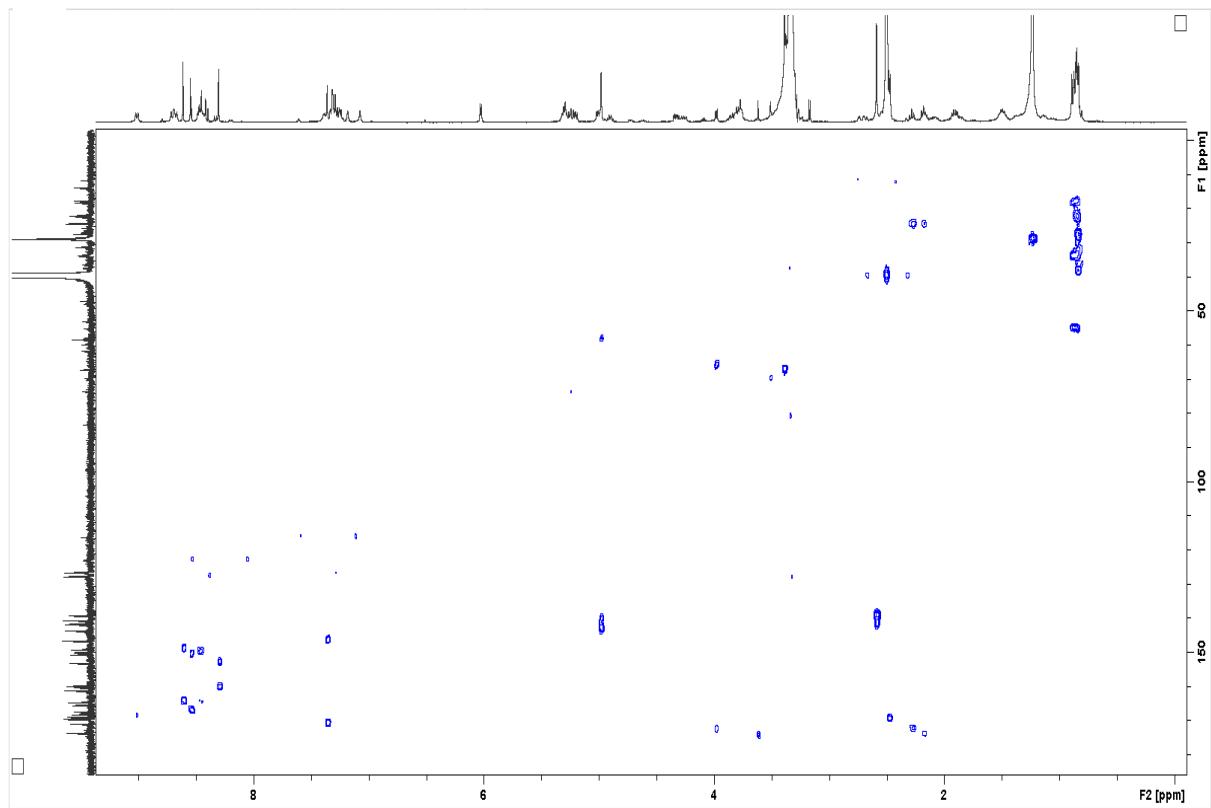


Figure S35. HMBC spectrum of **6**.

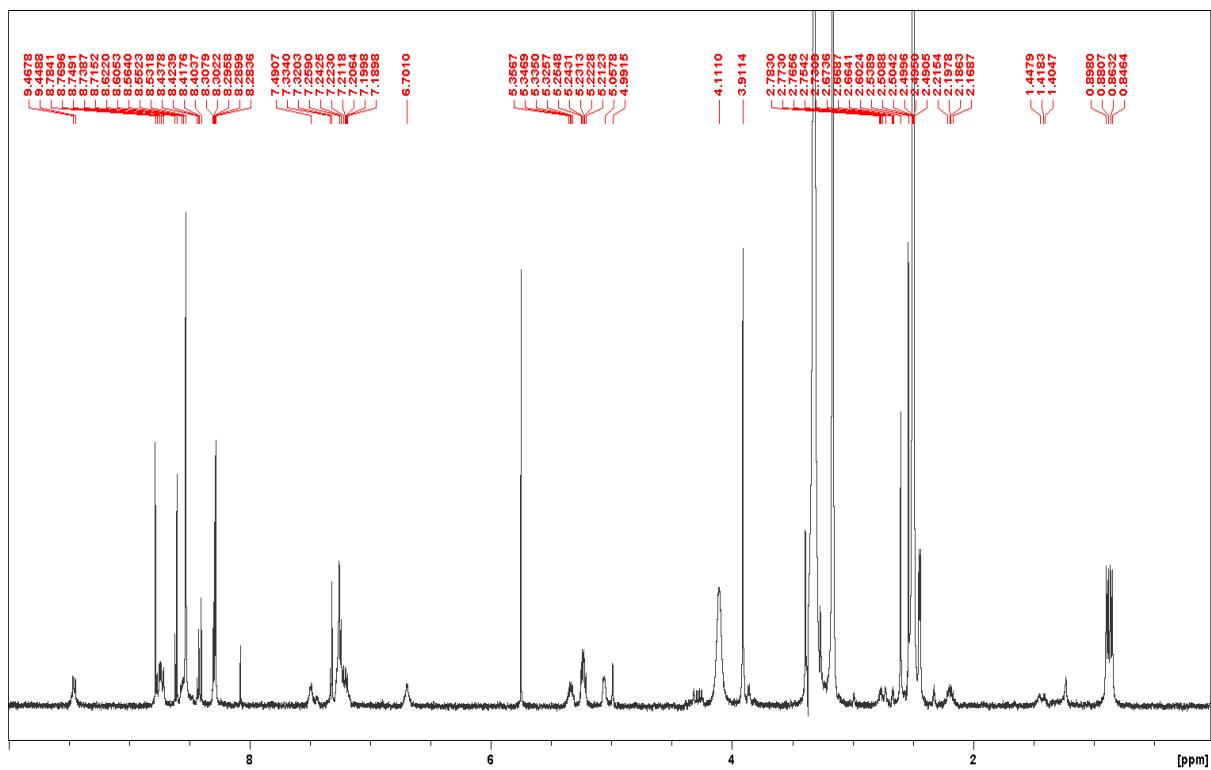


Figure S36. ^1H NMR spectrum (DMSO- d_6 , 400 MHz) of **7**.

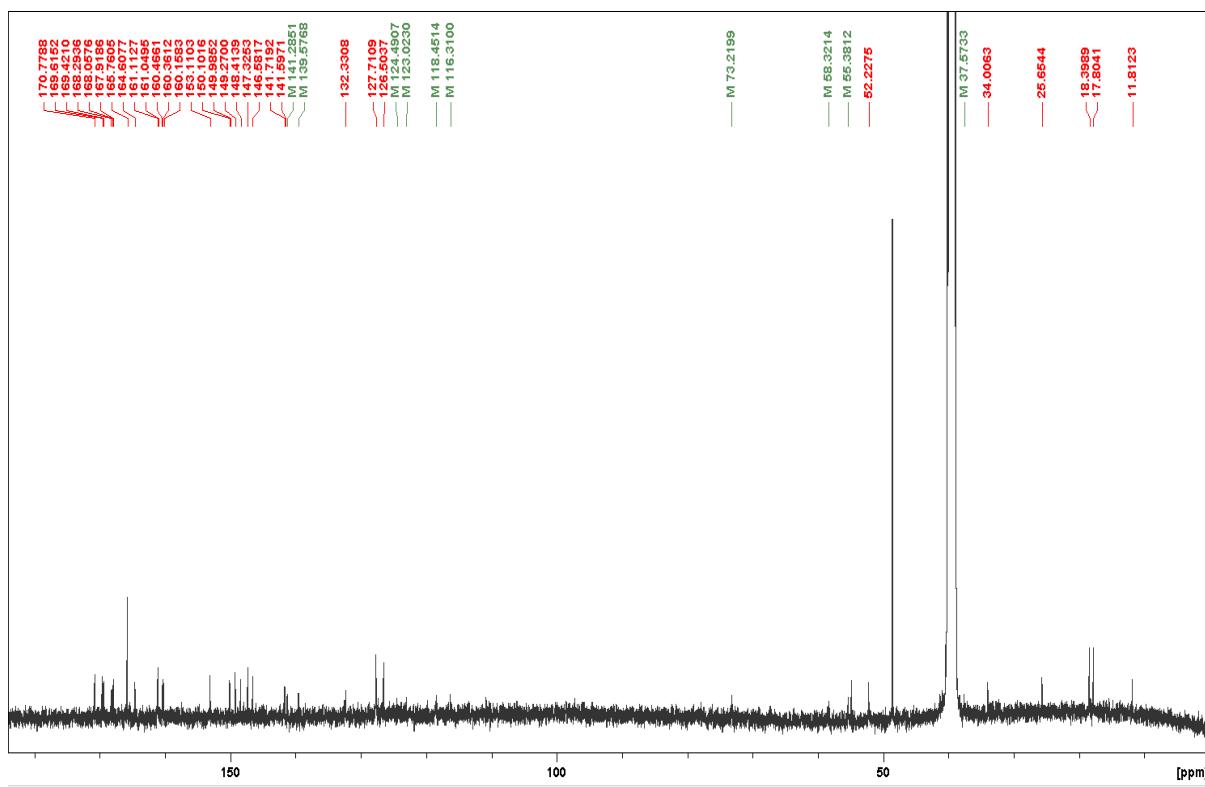


Figure S37. ^{13}C NMR spectrum (DMSO- d_6 , 100 MHz) of **7**.

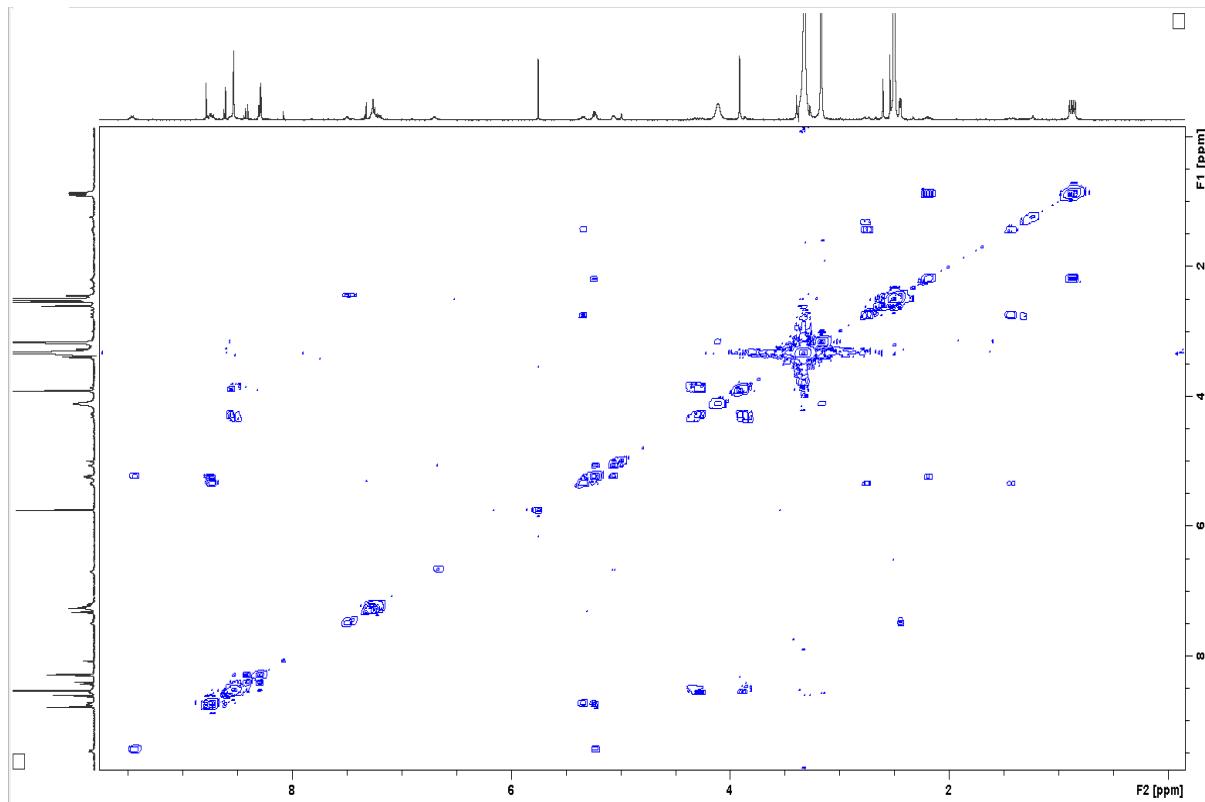


Figure S38. COSY spectrum of **7**.

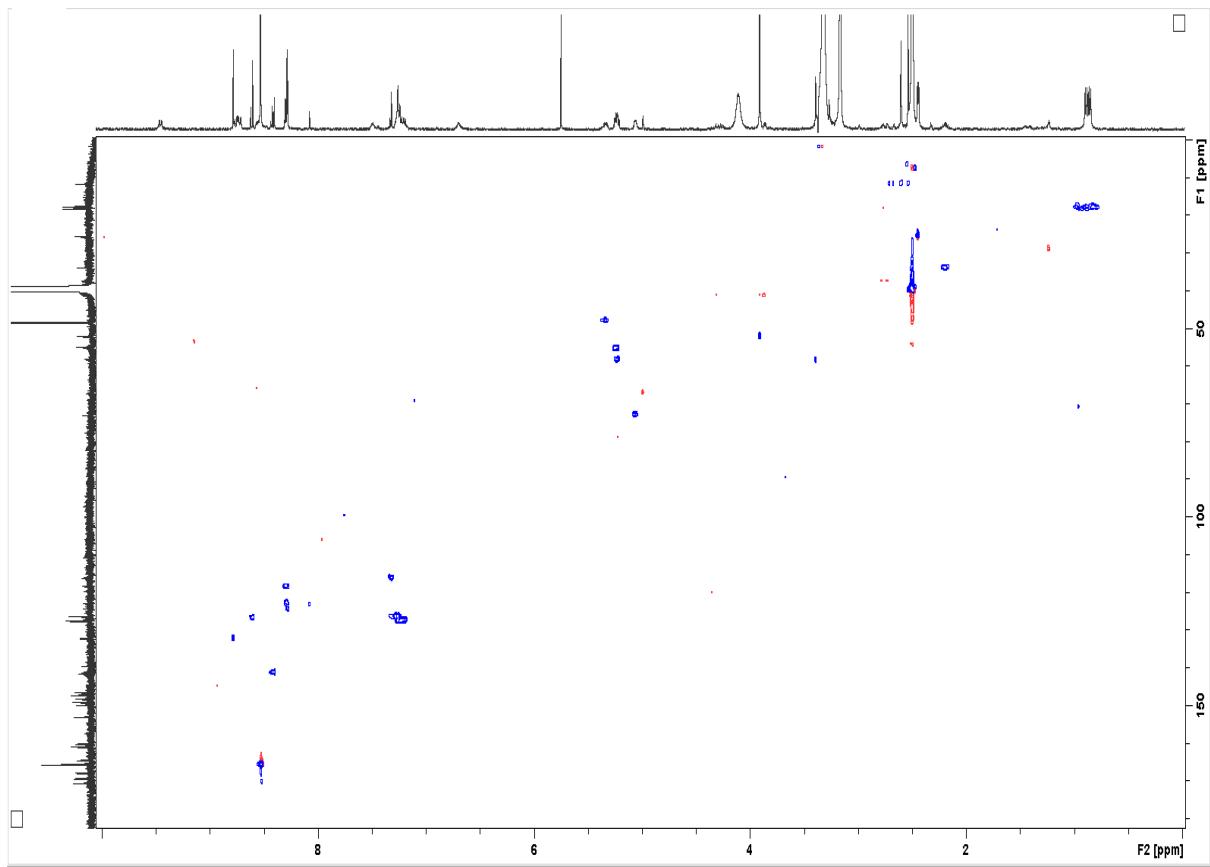


Figure S39. HSQC spectrum of 7.

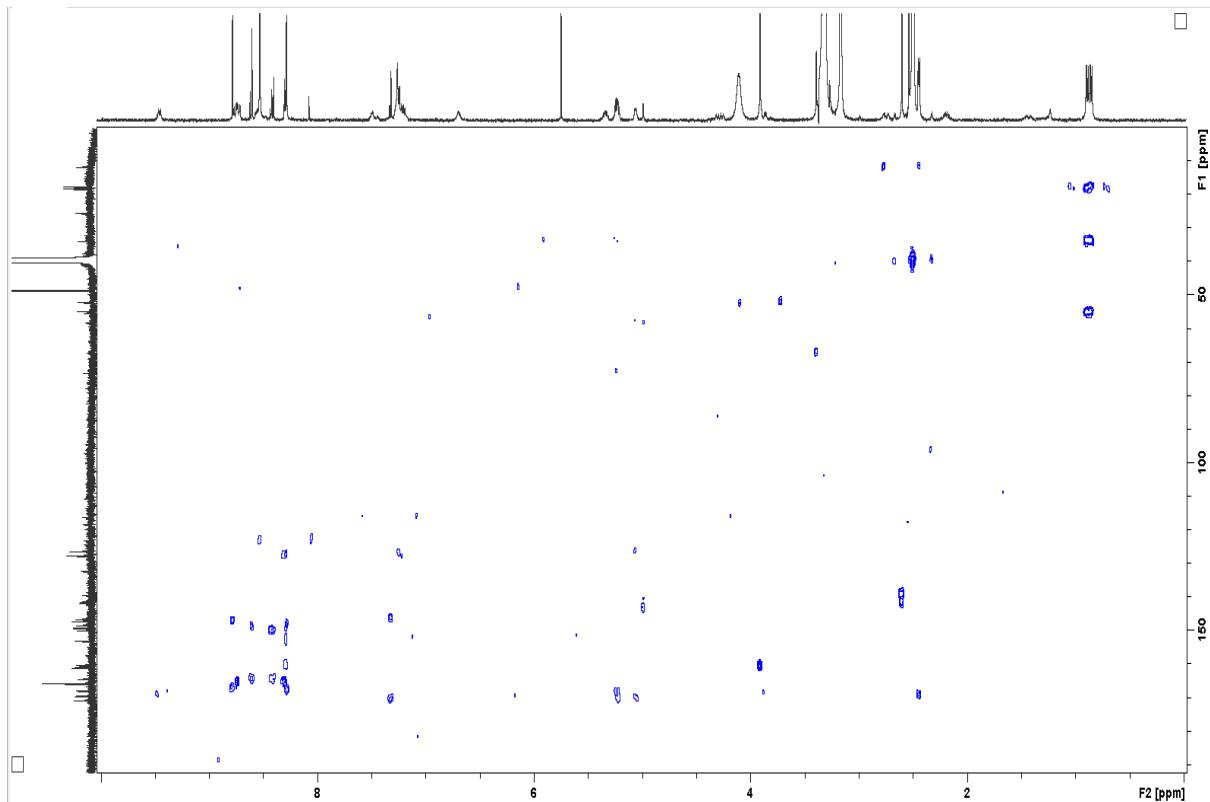


Figure S40. HMBC spectrum of 7.

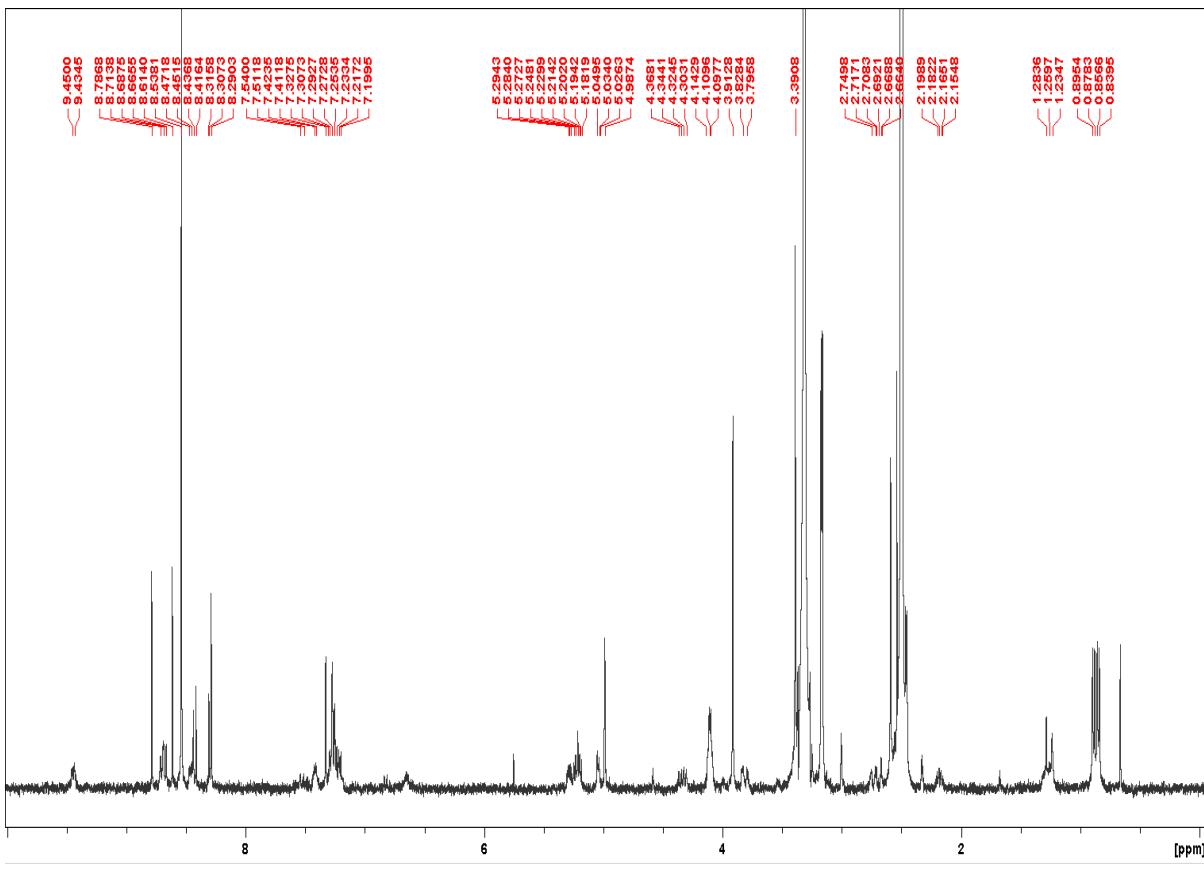


Figure S41. ^1H NMR spectrum (DMSO- d_6 , 400 MHz) of **8**.

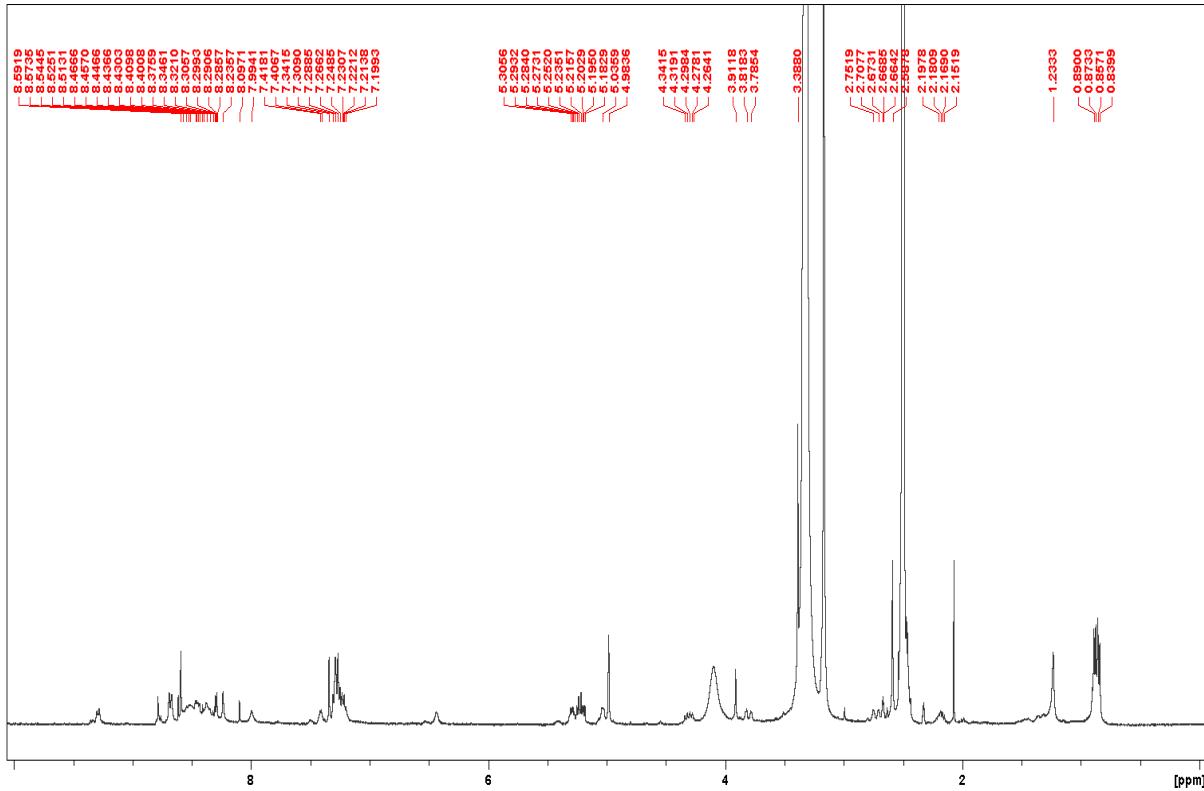


Figure S42. ^1H NMR spectrum (DMSO- d_6 , 400 MHz) of **9**.

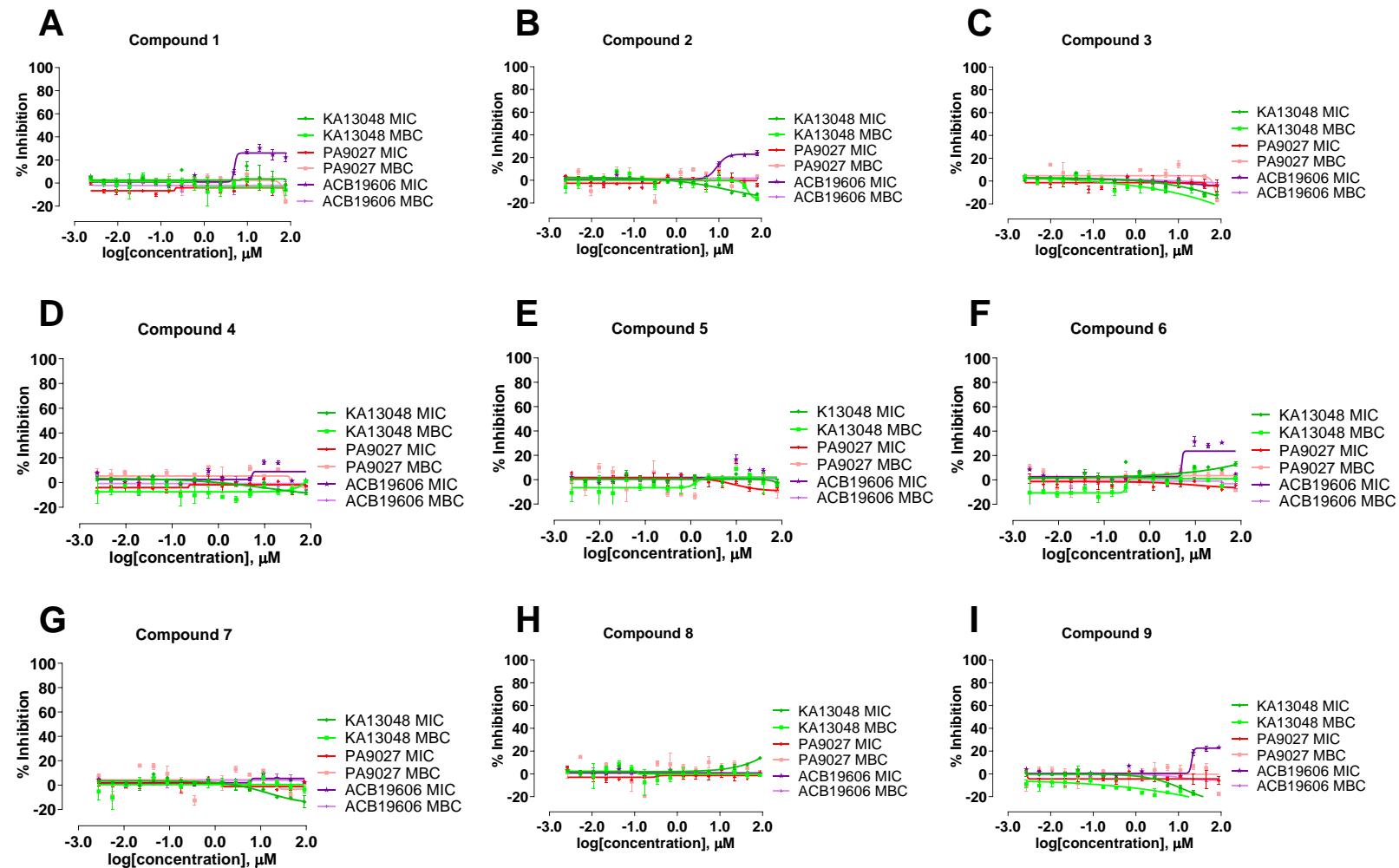


Figure S43. Inhibitory effect dose response curve against *Klebsiella aerogenes* (ATCC® 13048™), *Pseudomonas aeruginosa* (ATCC® 9027™) and *Acinetobacter baumannii* (ATCC® 19606™). **A)** Compound 1, **B)** Compound 2, **C)** Compound 3, **D)** Compound 4, **E)** Compound 5, **F)** Compound 6, **G)** Compound 7, **H)** Compound 8, **I)** Compound 9.

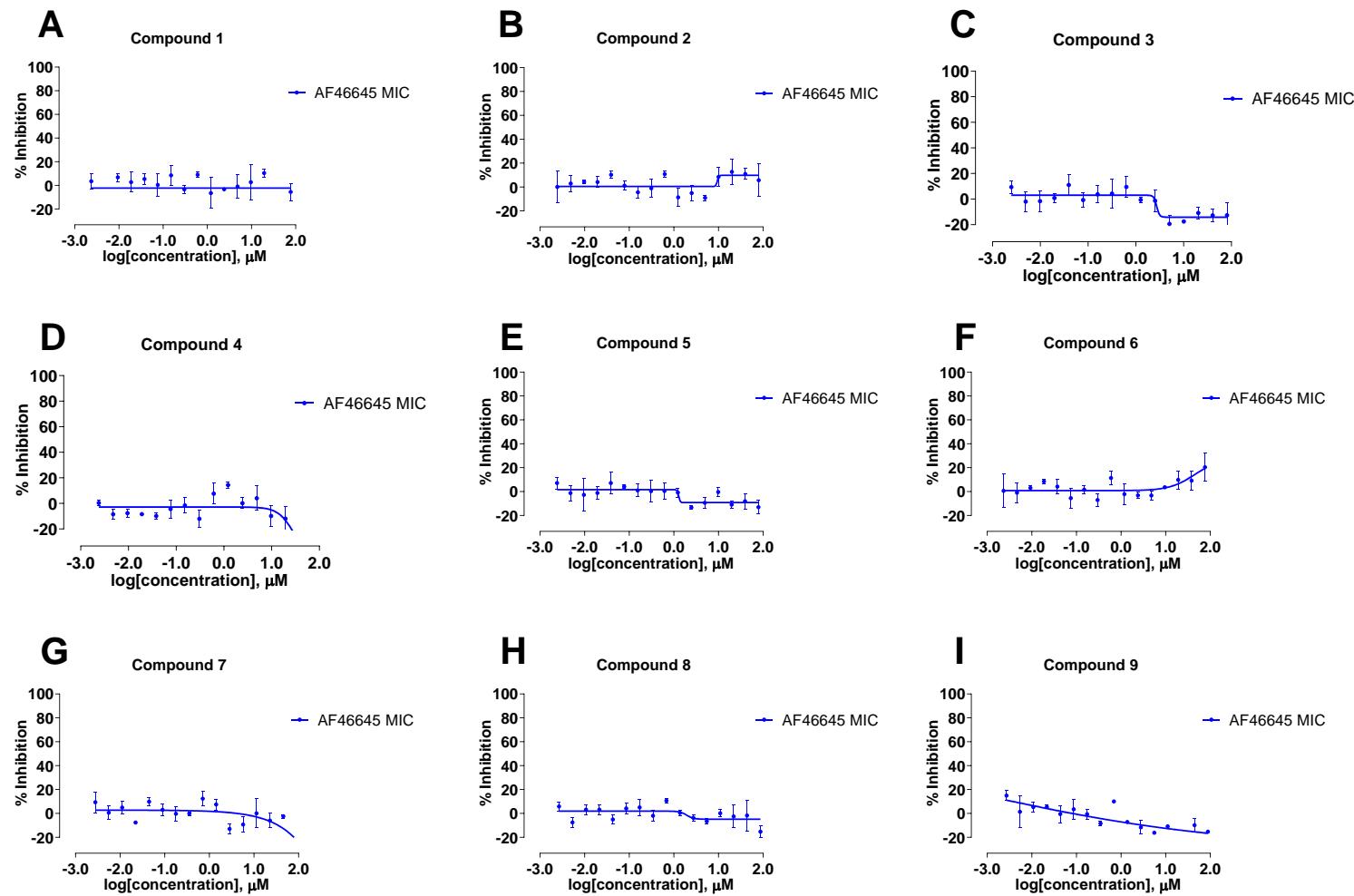


Figure S44. Inhibitory effect dose response curve against *Aspergillus fumigatus* (ATCC® 46645™). **A)** Compound 1, **B)** Compound 2, **C)** Compound 3, **D)** Compound 4, **E)** Compound 5, **F)** Compound 6, **G)** Compound 7, **H)** Compound 8, **I)** Compound 9.

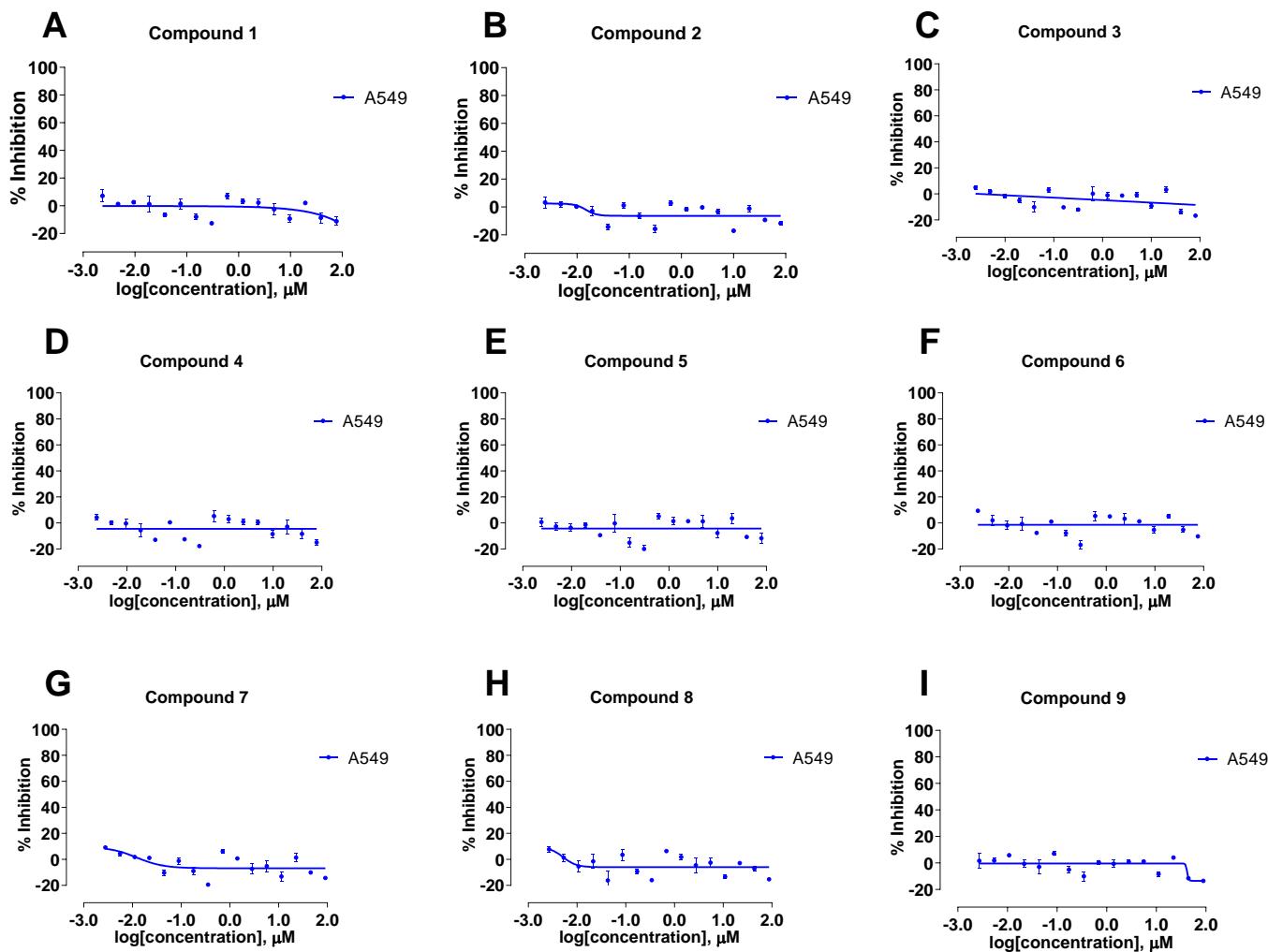


Figure S45. Inhibitory effect dose response curve against A549 Human lung carcinoma cells (ATCC® CCL-185™). **A)** Compound 1, **B)** Compound 2, **C)** Compound 3, **D)** Compound 4, **E)** Compound 5, **F)** Compound 6, **G)** Compound 7, **H)** Compound 8, **I)** Compound 9.