Supporting Information

Alkoxy-functionalised dihydropyrimido[4,5-*b*]quinolinones enabling anti-proliferative and anti-invasive Agents

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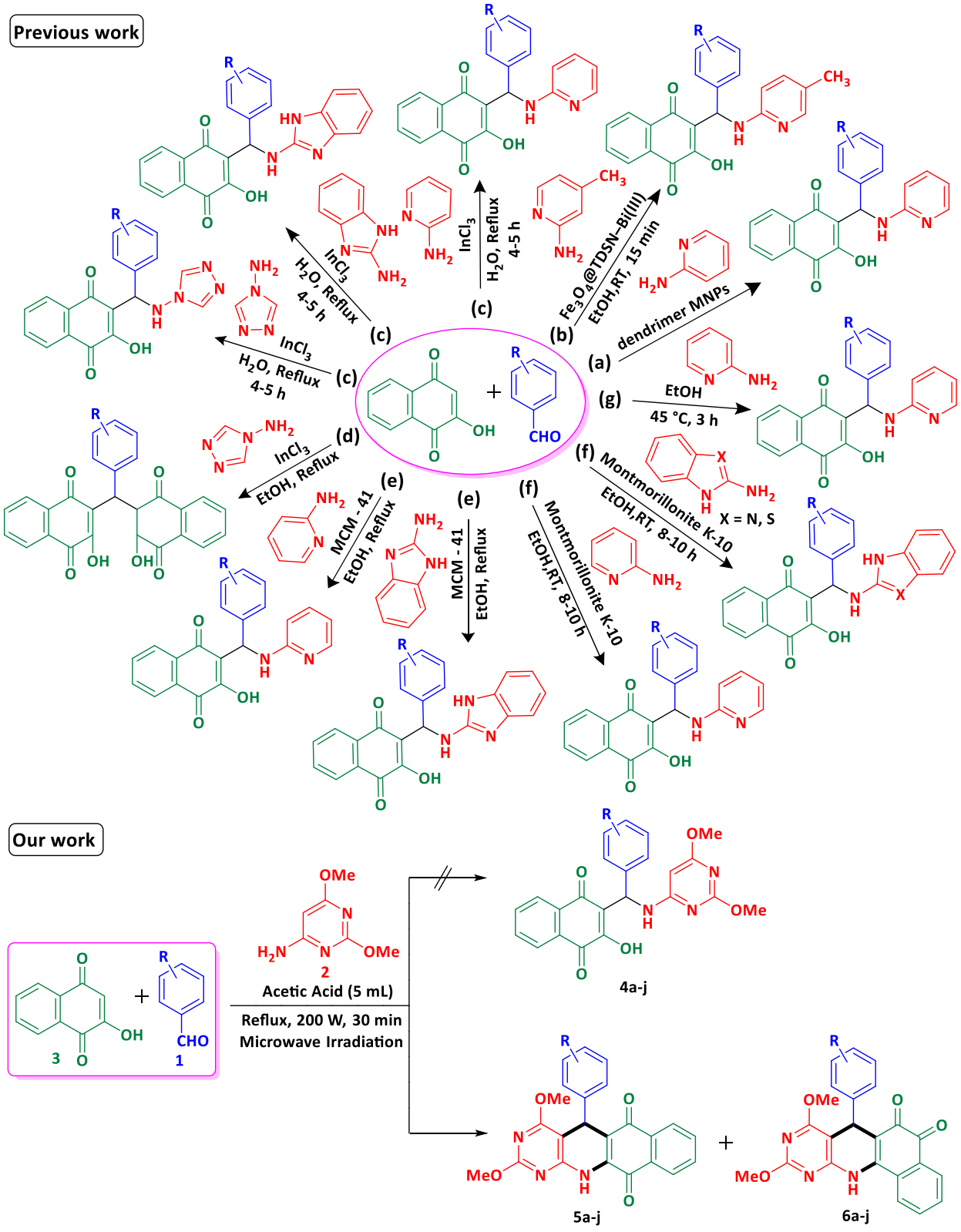
Table of Contents



Scheme S1 Multicomponent reaction of aryl aldehyde, heteroaromatic amine and lawsone.

Table S1. Optimization of solvent for the reaction.[a]

Entry	Solvent [b]	Temperature	Time	Conversion relative to aldehyde [c]
1	Solvent-free	100 °C	12 h	NR
2	Water	reflux	12 h	Incomplete
3	Ethanol	reflux	6 h	Full
4	Methanol	reflux	8 h	Full
5	**Acetic acid**	**reflux**	**1.5 h**	**Full**
6	Ethyl L-lactate	reflux	3 h	Full
7	Acetonitrile	reflux	6 h	Incomplete
8	DMF	reflux	4 h	Incomplete

[a] Reaction condition: 1 mmol *p*-methoxybenzaldehyde **1a**, 1 mmol 6-amino-2,4-dimethoxypyrimidine **2** and 1 mmol lawsone **3,** reflux

[b] 5 mL solvent

[c] Observed from TLC analysis

Table S2 Optimization of time and microwave irradiation power for the reaction.[a]

Entry	Power (w)	Time (min)	% Yield [b]
1	300	45	88
2	250	40	91
3	200	40	94
4	**200**	**30**	**94**
5	150	45	86

[a] Reaction condition: 1 mmol *p*-methoxybenzaldehyde **1a**, 1 mmol 6-amino-2,4-dimethoxypyrimidine **2** and 1 mmol lawsone **3,** reflux, 5 mL glacial acetic acid, reflux

[b] Crude product yield.

1. Experimental section

1.1. General methods:

For synthesis, all chemical reagents were purchased from the TCI, Sigma - Aldrich and Sisco Research Laboratories Pvt. Ltd. and used without further purification. The microwave-assisted reactions were performed in a "SINEO UWave-1000 Microwave, UV, US Synthesis Extraction Reactor". The progress of all chemical reactions was monitored by thin-layer chromatography (TLC, on aluminium plates pre-coated with F254 silica gel 60). Melting points of all solid compounds were determined by the open capillary tube method and are uncorrected. The LCMS analysis was collected on an MS–Agilent 6120 quadrupole. HRMS was determined on Waters Micro mass Q-Tof Micro 4000 quadrupole spectrometer. Nuclear magnetic resonance spectra (^{1}H NMR & ^{13}C NMR) were recorded on a Bruker 400 MHz WB FT-NMR spectrometer having proton noise decoupling mode with a standard 5mm probe using DMSO-d6 solution. Abbreviations are used for the ^{1}H NMR signal are as follows: s = singlet, d = doublet, t = triplet, q = quartet, dd = doublet of doublets, td = triplet of doublets, qd = quartet of doublets, qt = quartet of triplet,s m = multiplet. The chemical shifts are reported in parts per million and coupling constants (*J*) are provided in Hertz.

1.2. General procedure for synthesis of alkoxy-functionalised DHPQs 5a-j and 6a-j.

A mixture of aromatic aldehydes (**1**, 1 mmol), 6-amino-2,4-dimethoxypyrimidine (**2**, 1 mmol), lawsone (**3**, 1 mmol) and acetic acid (5 mL) were charged into a microwave vessel. The reaction mixture was heated at 110 °C (200 W) by MW irradiation for 30 min. The reaction progress was monitored on TLC using n-hexane:ethyl acetate (50:50, v/v) as mobile phase. After complete consumption of starting material, the reaction mixture was cooled to room temperature and poured into 20 mL cold water for complete solidification. The obtained crude product was filtered and dried in an oven. Further isolation and purification, the crude product was separated by flash chromatography using 50% ethyl acetate and 50% n-hexane as a mobile phase.

1.2.1. 2,4-dimethoxy-5-(4-methoxyphenyl)-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5a)

Orange solid, mp. 228 – 230 °C; ^{1}H-NMR (500 MHz, DMSO-d_6) (δ, ppm): 9.89 (s, 1H, NH), 8.02 (dd, *J* = 7.0, 2.0 Hz, 1H, Ar-H), 7.90 (dd, *J* = 7.5, 2.0 Hz, 1H, Ar-H), 7.80 (pd, *J* = 16, 7.5, 1.5 Hz, 2H, Ar-H), 7.15 (d, *J* = 8.5 Hz, 2H, Ar-H), 6.78 (d, *J* = 9.0 Hz, 2H, Ar-H), 5.25 (s, 1H, CH), 3.88 (s, 3H, OCH_3), 3.86 (s, 3H, OCH_3), 3.65 (s, 3H, OCH_3); ^{13}C-NMR (125 MHz, DMSO-d_6) (δ, ppm): 181.4 (C=O), 180.0 (C=O), 168.0, 163.2, 157.8, 155.7, 139.1, 137.1, 134.7, 133.2, 131.7, 130.2, 128.6, 125.8, 125.5, 118.2, 113.6, 94.2, 54.9 (OCH_3), 54.3 (OCH_3), 54.0 (OCH_3), 33.6 (C_{chiral}); HRMS(ES+): 430.1097 $[M + H]^+$.

1.2.2. 8,10-dimethoxy-7-(4-methoxyphenyl)-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6a)

Brownish orange solid, mp. 220 – 222 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 10.78 (s, 1H, NH), 8.52 (d, *J* = 8.0 Hz, 1H, Ar-H), 7.98 (d, *J* = 7.3 Hz, 1H, Ar-H), 7.83 (t, *J* = 7.5 Hz, 1H, Ar-H), 7.67 (t, *J* = 7.6 Hz, 1H, Ar-H), 7.14 (d, *J* = 8.6 Hz, 2H, Ar-H), 6.77 (d, *J* = 8.6 Hz, 2H, Ar-H), 5.16 (s, 1H,CH), 3.93 (s, 3H, OCH_3), 3.89 (s, 3H, OCH_3), 3.65 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 179.2 (C=O), 176.4 (C=O), 168.7, 163.8, 159.5, 156.7, 146.8, 146.4, 135.3, 131.7, 131.1, 130.2, 129.9, 129.1, 125.3, 120.0, 114.3, 113.8, 111.8, 95.7, 55.4 (OCH_3), 55.1 (OCH_3), 54.7 (OCH_3), 34.2 (C_{chiral}); HRMS(ES+): 430.1097 $[M + H]^+$.

1.2.3. 2,4-dimethoxy-5-(3-methoxyphenyl)-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5b)

Orange solid, mp. 230 – 232 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.99 (s, 1H, NH), 8.06 – 8.02 (m, 1H, Ar-H), 7.94 – 7.90 (m, 1H, Ar-H), 7.86 – 7.78 (m, 2H, Ar-H), 7.14 (t, *J* = 8.2 Hz, 1H, Ar-H), 6.83 – 6.79 (m, 2H, Ar-H), 6.74 – 6.70 (m, 1H, Ar-H), 5.31 (s, 1H,CH), 3.89 (s, 3H, OCH_3), 3.88 (s, 3H, OCH_3), 3.68 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 182.0 (C=O), 179.5 (C=O), 168.7, 163.8, 159.6, 156.4, 146.7, 140.1, 135.2, 133.8, 132.2, 130.9, 130.0, 126.5, 126.2, 120.3, 118.3, 114.4, 112.0, 94.5, 55.4 (OCH_3), 54.9 (OCH_3), 54.6 (OCH_3), 35.0 (C_{chiral}); HRMS(ES+): 430.1097 $[M + H]^+$.

1.2.4. 8,10-dimethoxy-7-(3-methoxyphenyl)-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6b)

Brownish orange solid, mp. 222 – 224 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 9.97 (s, 1H. NH), 8.07 – 8.01 (m, 1H, Ar-H), 7.92 (dd, *J* = 7.2, 1.9 Hz, 1H, Ar-H), 7.83 (qd, *J* = 7.0, 1.8 Hz, 2H, Ar-H), 7.18 – 7.11 (m, 1H, Ar-H), 6.85 – 6.69 (m, 2H, Ar-H), 5.31 (s, 1H, CH), 3.89 (s, 3H, OCH_3), 3.88 (s, 3H, OCH_3), 3.68 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 178.7 (C=O), 175.9 (C=O), 168.2, 163.3, 159.0, 156.2, 146.3, 145.9, 134.8, 131.2, 130.6, 129.7, 129.4, 128.7, 124.8, 119.6, 113.8, 113.3, 111.3, 95.2, 54.9 (OCH_3), 54.6 (OCH_3), 54.2 (OCH_3), 33.8 (C_{chiral}); HRMS(ES+): 430.1097 $[M + H]^+$.

1.2.5. 5-(2,4-dimethoxyphenyl)-2,4-dimethoxy-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5c)

Orange solid, mp. 232 – 234 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.76 (s, 1H, NH), 8.05 – 7.98 (m, 1H, Ar-H), 7.89 – 7.75 (m, 4H, Ar-H), 7.21 – 7.14 (m, 1H, Ar-H), 6.44 – 6.37 (m, 2H, Ar-H), 5.39 (s, 1H, CH), 3.86 (s, 3H, OCH_3), 3.79 (s, 3H, OCH_3), 3.68 (s, 3H, OCH_3), 3.65 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.8 (C=O), 179.8 (C=O), 168.4, 163.4, 159.9, 158.9, 156.7, 140.2, 135.2, 133.6, 132.4, 131.6, 130.6, 126.3, 126.0, 125.6, 117.9, 105.2, 99.3, 94.2, 56.0 (OCH_3), 55.5 (OCH_3), 54.8 (OCH_3), 54.4 (OCH_3), 32.0 (C_{chiral}); HRMS(ES+): 460.1247 $[M + H]^+$.

1.2.6. 7-(2,4-dimethoxyphenyl)-8,10-dimethoxy-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6c)

Brownish orange solid, mp. 218 – 220 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 10.72 (s, 1H, NH), 8.52 (d, *J* = 8.1 Hz, 1H, Ar-H), 7.95 (d, *J* = 7.5 Hz, 2H, Ar-H), 7.65 (t, *J* = 7.5 Hz, 1H, Ar-H), 7.19 (d, *J* = 8.1 Hz, 1H, Ar-H), 6.41 (d, *J* = 7.8 Hz, 2H, Ar-H), 5.21 (s, 1H, NH), 3.91 (s, 3H, OCH_3), 3.82 (s, 3H, OCH_3), 3.67 (s, 3H, OCH_3), 3.60 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 179.0 (C=O), 175.8 (C=O), 168.0, 162.9, 159.2, 158.5, 156.5, 146.1, 134.9, 131.4, 131.0, 130.5, 130.1, 128.5, 124.7, 124.6, 112.7, 104.5, 98.9, 94.8, 55.5 (OCH_3), 55.0 (OCH_3), 54.5 (OCH_3), 54.0 (OCH_3), 31.6 (C_{chiral}); HRMS(ES+): 460.1247 $[M + H]^+$.

1.2.7. 5-(2,5-dimethoxyphenyl)-2,4-dimethoxy-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5d)

Orange solid, mp. 238 – 240 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.85 (s, 1H, NH), 8.05 – 7.98 (m, 1H, Ar-H), 7.89 – 7.75 (m, 3H, Ar-H), 6.86 – 6.79 (m, 2H, Ar-H), 6.70 (dd, *J* = 8.9, 3.0 Hz, 1H, Ar-H), 5.40 (s, 1H, CH), 3.86 (s, 3H, OCH_3), 3.80 (s, 3H, OCH_3), 3.67 (s, 3H, OCH_3), 3.59 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.8 (C=O), 179.7 (C=O), 168.5, 163.5, 156.8, 153.2, 152.4, 140.4, 135.3, 134.0, 133.6, 132.3, 130.6, 126.3, 126.1, 117.5, 113.5, 112.5, 93.9, 56.7 (OCH_3), 55.7 (OCH_3), 54.8 (OCH_3), 54.4 (OCH_3), 33.1 (C_{chiral}); HRMS(ES+): 460.0569 $[M + H]^+$.

1.2.8. 7-(2,5-dimethoxyphenyl)-8,10-dimethoxy-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6d)

Brownish orange solid, mp. 218 – 220 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 10.77 (s, 1H, NH), 8.53 (d, *J* = 8.0 Hz, 1H, Ar-H), 7.96 (dd, *J* = 7.6, 1.5 Hz, 1H, Ar-H), 7.83 (td, *J* = 7.7, 1.5 Hz, 1H, Ar-H), 7.69 – 7.63 (m, 1H, Ar-H), 6.86 (d, *J* = 3.1 Hz, 1H, Ar-H), 6.78 (d, *J* = 8.9 Hz, 1H, Ar-H), 6.69 (dd, *J* = 8.8, 3.1 Hz, 1H, Ar-H), 5.21 (s, 1H, CH), 3.91 (s, 3H, OCH_3), 3.83 (s, 3H, OCH_3), 3.67 (s, 3H, OCH_3), 3.54 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 178.9 (C=O), 175.7 (C=O), 168.1, 163.0, 156.6, 152.6, 152.1, 146.3, 134.8, 133.2, 131.1, 130.5, 130.0, 128.5, 124.6, 117.4, 113.1, 112.3, 111.8, 94.4, 56.2 (OCH_3), 55.3 (OCH_3), 54.5 (OCH_3), 54.0 (OCH_3), 32.6 (C_{chiral}); HRMS(ES+): 460.0569 $[M + H]^+$.

1.2.9. 5-(3,4-dimethoxyphenyl)-2,4-dimethoxy-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5e)

Orange solid, mp. 238 – 240 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.94 (s, 1H, NH), 8.04 (d, *J* = 6.5 Hz, 1H, Ar-H), 7.92 (d, *J* = 7.0 Hz, 1H, Ar-H), 7.85-7.78 (m, 2H, Ar-H), 6.89 (s, 1H, Ar-H), 6.77 (d, *J* = 8.4 Hz, 1H, Ar-H), 6.68 (d, *J* = 8.2 Hz, 1H, Ar-H), 5.26 (s, 1H, CH), 3.89 (s, 6H, OCH_3), 3.70 (s, 3H, OCH_3), 3.64 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.6 (C=O),

179.1 (C=O), 168.1, 163.2, 155.8, 148.3, 147.6, 139.4, 137.5, 134.7, 133.3, 131.8, 130.4, 126.0, 125.7, 119.5, 118.0, 111.8, 111.7, 94.3, 55.5 (OCH_3), 55.4 (OCH_3), 54.4 (OCH_3), 54.1 (OCH_3), 34.0 (C_{chiral}); HRMS(ES+): 460.0644 $[M + H]^+$.

1.2.10. 7-(3,4-dimethoxyphenyl)-8,10-dimethoxy-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6e)

Brownish orange solid, mp. 224 – 226 °C; ^{1}H-NMR (500 MHz, DMSO-d_6) (δ, ppm): δ 10.77 (s, 1H, NH), 8.50 (d, *J* = 7.5 Hz, 1H, Ar-H), 7.99 (d, *J* = 6.0 Hz, 1H, Ar-H), 7.83 (t, *J* = 7.5 Hz, 1H, Ar-H), 7.67 (t, *J* = 7.5 Hz, 1H, Ar-H), 6.90 (s, 1H, Ar-H), 6.77 (d, *J* = 8.5 Hz, 1H, Ar-H), 6.62 (d, *J* = 8.5 Hz, 1H, Ar-H), 5.16 (s, 1H, CH), 3.93 (s, 3H, OCH_3), 3.91 (s, 3H, OCH_3), 3.69 (s, 3H, OCH_3), 3.64 (s, 3H, OCH_3); ^{13}C-NMR (125 MHz, DMSO-d_6) (δ, ppm): 178.7 (C=O), 175.9 (C=O), 168.1, 163.1, 155.9, 148.1, 147.4, 145.6, 137.5, 134.7, 131.1, 130.5, 129.7, 128.5, 124.6, 118.9, 113.4, 111.7, 111.6, 95.5, 55.3 (OCH_3), 54.3 (OCH_3), 54.5 (OCH_3), 54.1 (OCH_3), 33.2 (C_{chiral}); HRMS(ES+): 460.0644 $[M + H]^+$.

1.2.11. 5-(3,5-dimethoxyphenyl)-2,4-dimethoxy-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5f)

Orange solid, mp. 236 – 238 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.86 (s, 1H, NH), 8.03 (d, *J* = 6.6 Hz, 1H, Ar-H), 7.87 (d, *J* = 7.1 Hz, 1H, Ar-H), 7.84-7.78 (m, 2H, Ar-H), 6.85 (d, *J* = 3.1 Hz, 1H, Ar-H), 6.82 (d, *J* = 8.9 Hz, 1H, Ar-H), 6.71 (dd, *J* = 8.9, 3.0 Hz, 1H, Ar-H), 5.40 (s, 1H, CH), 3.87 (s, 3H, OCH_3), 3.81 (s, 3H, OCH_3), 3.67 (s, 3H, OCH_3), 3.60 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.3 (C=O), 179.2 (C=O), 167.9, 162.9, 159.3, 158.4, 156.2, 139.6, 134.7, 133.1, 131.8, 131.1, 130.1, 125.8, 125.5, 125.1, 117.4, 104.7, 98.8, 93.7, 55.5 (OCH_3), 55.0 (OCH_3), 54.3 (OCH_3), 53.9 (OCH_3), 31.5 (C_{chiral}); HRMS(ES+): 460.1247 $[M + H]^+$.

1.2.12. 7-(3,5-dimethoxyphenyl)-8,10-dimethoxy-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6f)

Brownish orange solid, mp. 222 – 224 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 10.78 (s, 1H, NH), 8.54 (d, *J* = 8.0 Hz, 1H, Ar-H), 7.97 (d, *J* = 7.6 Hz, 1H, Ar-H), 7.84 (t, *J* = 7.7 Hz, 1H, Ar-H), 6.87 (d, *J* = 3.1 Hz, 1H, Ar-H), 6.79 (d, *J* = 8.9 Hz, 1H, Ar-H), 6.70 (dd, *J* = 8.8, 3.1 Hz, 1H, Ar-H), 5.22 (s, 1H, CH), 3.92 (s, 3H, OCH_3), 3.84 (s, 3H, OCH_3), 3.68 (s, 3H, OCH_3), 3.55 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 179.4 (C=O), 176.2 (C=O), 168.5, 163.5, 157.1, 153.1, 152.5, 146.8, 135.3, 133.7, 131.5, 131.0, 130.4, 129.0, 125.1, 117.9, 113.6, 112.8, 112.2, 94.9, 56.7 (OCH_3), 55.7 (OCH_3), 54.9 (OCH_3), 54.5 (OCH_3), 33.11 (C_{chiral}); HRMS(ES+): 460.1247 $[M + H]^+$.

1.2.13. 5-(4-hydroxy-3-methoxyphenyl)-2,4-dimethoxy-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5g)

Orange solid, mp. 234 – 236 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.86 (s, 1H, NH), 8.81 (s, 1H, OH), 8.04 (dd, *J* = 7.3, 1.6 Hz, 1H, Ar-H), 7.92 (dd, *J* = 7.4, 1.6 Hz, 1H, Ar-H), 7.81 (pd, *J* = 7.4, 1.6 Hz, 2H, Ar-H), 6.85 (d, *J* = 1.9 Hz, 1H, Ar-H), 6.62 – 6.54 (m, 2H, Ar-H), 5.23 (s, 1H, CH), 3.89 (s, 3H, OCH_3), 3.88 (s, 3H, OCH_3), 3.71 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.6 (C=O), 179.1 (C=O), 168.1, 163.2, 155.7, 147.1, 145.4, 139.3, 136.0, 134.7, 133.3, 131.8, 130.4, 125.9, 125.7, 119.8, 118.2, 115.3, 112.0, 94.5, 55.6 (OCH_3), 54.4 (OCH_3), 54.0 (OCH_3), 33.9 (C_{chiral}); HRMS(ES+): 446.0551 $[M + H]^+$.

1.2.14. 7-(4-hydroxy-3-methoxyphenyl)-8,10-dimethoxy-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6g)

Brownish orange solid, mp. 252 – 254 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 10.74 (s, 1H, NH), 8.77 (s, 1H, OH), 8.51 (d, *J* = 8.0 Hz, 1H, Ar-H), 7.99 (d, *J* = 7.7 Hz, 1H, Ar-H), 7.83 (t, *J* = 7.5 Hz, 1H, Ar-H), 7.67 (t, *J* = 7.5 Hz, 1H, Ar-H), 6.87 (s, 1H, Ar-H), 6.59 (d, *J* = 8.1 Hz, 1H, Ar-H), 6.51 (d, *J* = 8.1 Hz, 1H, Ar-H), 5.14 (s, 1H, CH), 3.93 (s, 3H, OCH_3), 3.92 (s, 3H, OCH_3), 3.70 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 178.8 (C=O), 176.0 (C=O), 168.2, 163.1, 156.0, 147.0, 145.7, 145.2, 136.1, 134.8, 131.1, 130.6, 129.8, 128.6, 124.7, 119.3, 115.3, 113.7, 112.1, 95.8, 55.6 (OCH_3), 54.5 (OCH_3), 54.1 (OCH_3), 33.1 (C_{chiral}); HRMS(ES+): 446.0551 $[M + H]^+$.

1.2.15. 2,4-dimethoxy-5-(3,4,5-trimethoxyphenyl)-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5h)

Orange solid, mp. 228 – 230 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.92 (s, 1H, NH), 8.06 (dd, *J* = 6.9, 2.0 Hz, 1H, Ar-H), 7.94 (dd, *J* = 6.9, 1.9 Hz, 1H, Ar-H), 7.84 (qt, *J* = 7.4, 3.6 Hz, 2H, Ar-H), 6.54 (s, 2H, Ar-H), 5.29 (s, 1H, CH), 3.92 (s, 3H, OCH_3), 3.90 (s, 3H, OCH_3), 3.69 (s, 6H, OCH_3), 3.58 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.3 (C=O), 179.2 (C=O), 168.0, 163.0, 156.3, 152.7, 151.9, 139.9, 134.8, 133.5, 133.1, 131.8, 130.1, 125.8, 125.6, 117.0, 113.0, 112.0, 93.4, 56.2 (OCH_3), 55.2 (OCH_3), 54.3 (OCH_3), 53.9 (OCH_3), 32.6 (C_{chiral}); HRMS(ES+): 490.0702 $[M + H]^+$.

1.2.16. 8,10-dimethoxy-7-(3,4,5-trimethoxyphenyl)-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6h)

Brownish orange solid, mp. 220 – 222 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 10.76 (s, 1H, NH), 8.51 (d, *J* = 7.9 Hz, 1H, Ar-H), 8.00 (dd, *J* = 7.6, 1.5 Hz, 1H, Ar-H), 7.83 (td, *J* = 7.7, 1.5 Hz, 1H, Ar-H), 7.67 (td, *J* = 7.5, 0.9 Hz, 1H, Ar-H), 6.50 (s, 2H, Ar-H), 5.19 (s, 1H, CH), 3.94 (s, 3H, OCH_3), 3.93 (s, 3H, OCH_3), 3.66 (s, 6H, OCH_3), 3.57 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 179.1 (C=O), 176.4 (C=O), 168.7, 163.7, 156.4, 153.1, 146.3, 140.9, 136.8, 135.2, 131.6, 131.1, 130.1, 129.0, 125.2, 113.6, 105.3, 105.0, 95.7, 60.3 (OCH_3), 56.2 (OCH_3), 55.0 (OCH_3), 54.6 (OCH_3), 34.5 (C_{chiral}); HRMS(ES+): 490.0702 $[M + H]^+$.

1.2.17. 5-(6-bromobenzo[*d*][1,3]dioxol-5-yl)-2,4-dimethoxy-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5i)

Orange solid, mp. 224 – 226 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.91 (s, 1H, NH), 8.03 (s, 1H, Ar-H), 7.89 (s, 1H, Ar-H), 7.82 (d, *J* = 6.0 Hz, 2H, Ar-H), 7.07 (s, 1H, Ar-H), 6.80 (s, 1H, Ar-H), 5.95 (s, 2H, OCH_2O), 5.60 (s, 1H, CH), 3.88 (s, 3H, OCH_3), 3.81 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.2 (C=O), 178.9 (C=O), 168.4, 163.3, 155.5, 147.3, 146.8, 139.8, 137.7, 134.7, 133.2, 131.8, 130.3, 125.9, 125.6, 117.7, 113.0, 111.7, 110.2, 101.8 (OCH_2O), 93.9, 54.4 (OCH_3), 53.8 (OCH_3), 35.6 (C_{chiral}); HRMS(ES+): 521.9385 $[M + H]^+$.

1.2.18. 7-(6-bromobenzo[*d*][1,3]dioxol-5-yl)-8,10-dimethoxy-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6i)

Brownish orange solid, mp. 252 – 254 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 10.72 (s, 1H, NH), 7.99 (dd, *J* = 7.6, 1.5 Hz, 1H, Ar-H), 7.89 – 7.82 (m, 2H, Ar-H), 7.68 (dd, *J* = 8.3, 6.0 Hz, 2H, Ar-H), 7.04 (s, 1H, Ar-H), 6.77 (s, 1H, Ar-H), 5.95 (s, 2H, OCH_2O), 5.45 (s, 1H, CH), 3.92 (s, 3H, OCH_3), 3.83 (s, 3H, OCH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 179.1 (C=O), 176.4 (C=O), 169.0, 163.8, 156.3, 147.6, 147.2, 146.3, 137.9, 135.2, 131.7, 131.1, 130.2, 129.0, 126.2, 125.5, 113.8, 112.2, 110.6, 102.3 (OCH_2O), 95.4, 55.0 (OCH_3), 54.4 (OCH_3), 35.8 (C_{chiral}); HRMS(ES+): 521.9385 $[M + H]^+$.

1.2.19. 5-(4-ethoxyphenyl)-2,4-dimethoxy-5,12-dihydrobenzo[*g*]pyrimido[4,5-*b*]quinoline-6,11-dione (5j)

Orange solid, mp. 208 – 210 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): 9.95 (s, 1H, NH), 8.05 – 8.01 (m, 1H, Ar-H), 7.93 – 7.89 (m, 1H, Ar-H), 7.81 (pd, *J* = 7.3, 1.7 Hz, 2H, Ar-H), 7.14 (d, *J* = 8.7 Hz, 2H, Ar-H), 6.76 (d, *J* = 8.7 Hz, 2H, Ar-H), 5.26 (s, 1H, CH), 3.92 (m, 2H, CH_2), 3.89 (s, 3H, OCH_3), 3.86 (s, 3H, OCH_3), 1.25 (t, *J* = 7.0 Hz, 3H, CH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 181.5 (C=O), 179.1 (C=O), 168.1, 163.2, 157.2, 155.8, 139.2, 137.0, 134.7, 133.3, 131.8, 130.3, 128.6, 125.9, 125.6, 118.3, 114.2, 94.3, 62.8 (OCH_2), 54.4 (OCH_3), 54.1 (OCH_3), 33.6 (C_{chiral}), 14.6 (CH_3); HRMS(ES+): 444.0575 $[M + H]^+$.

1.2.20. 7-(4-ethoxyphenyl)-8,10-dimethoxy-7,12-dihydrobenzo[*h*]pyrimido[4,5-*b*]quinoline-5,6-dione (6j)

Brownish orange solid, mp. 226 – 228 °C; ^{1}H-NMR (400 MHz, DMSO-d_6) (δ, ppm): δ 10.80 (s, 1H, NH), 8.52 (d, *J* = 7.9 Hz, 1H, Ar-H), 7.98 (d, *J* = 7.6 Hz, 1H, Ar-H), 7.83 (t, *J* = 7.7 Hz, 1H, Ar-H), 7.67 (t, *J* = 7.6 Hz, 1H, Ar-H), 7.12 (d, *J* = 8.3 Hz, 2H, Ar-H), 6.75 (d, *J* = 8.3 Hz, 2H, Ar-H), 5.15 (s, 1H, CH), 3.93 (s, 3H, OCH_3), 3.91 (d, *J* = 6.9 Hz, 2H, CH_2), 3.89 (s, 3H, OCH_3), 1.25 (t, *J* = 7.0 Hz, 3H, CH_3); ^{13}C-NMR (100 MHz, DMSO-d_6) (δ, ppm): 178.8 (C=O), 175.9 (C=O), 168.2, 163.2, 157.1, 156.0, 145.6, 137.1, 134.8, 131.1, 130.5, 129.8, 128.6, 128.4, 124.7, 114.1, 113.8, 95.6, 62.8 (OCH_2), 54.6 (OCH_3), 54.2 (OCH_3), 32.9 (C_{chiral}), 14.6 (CH_3); HRMS(ES+): 444.0575 $[M + H]^+$.

1.3. Cell Based Assays

1.3.1 Materials and cell lines

Primary patient glioblastoma cells GBM6, GBM22 and GBM143 were acquired from the Brain Tumour PDX National Resource, Mayo Clinic, USA. Propidium iodide, insulin and epidermal growth factor were purchased from Sigma Millipore. Temozolomide was from Accord Healthcare. Immunoblot analyses were carried out as stated previously[1-3]. The following antibodies were used for immunoblotting: total AKT (Cell Signalling #9272), phospho Thr308 AKT (Cell Signalling #2965), total p44/42 ERK1/2 (Cell Signalling #4695), phospho Thr202/Tyr204 p44/42 ERK1/2 (Cell Signalling #4376), total STAT3 (Cell Signalling #9139), and phospho Tyr705 STAT3 (Cell Signalling #9145).

1.3.2 Cell Culture

Mammalian cells were grown in a humidified incubator with 5% CO_2 at 37°C. Primary patient glioblastoma cell lines were cultured in GBM media (DMEM supplemented with 10% FBS, 1% penicillin and streptomycin, 10 µg/ml insulin, and 20 ng/ml hEGF) as reported previously[4-6].

1.3.3 Cell viability assay

Cell viability was measured as stated previously[7, 8]. Briefly, equal number of cells were seeded per well in 96 well plates. After 72 hours treatment of indicated drugs or DMSO control 10 µl of MTS was added. After one hour incubation at 37 °C, absorbance was measured at 450 nm using a Tecan multi-well plate reader and data was represented as % viability compared to DMSO treated control as stated previously.

1.3.4 Proliferation and colony formation assays

Cell proliferation was measured as stated previously [1, 9]. Briefly, GBM6 cells were seeded in a 96 well plate at 3,000 cells per well in GBM media and cells were allowed to attach and enter log phase before drug treatment. DMSO, 2.9 µM (**5c**) and 3.3 µM (**5h**) were added. Every 24 hours afterwards, cell viability was assessed using CellTiter 96® AQueous One Solution Cell Proliferation Assay (MTS) kit following manufacturer's instructions and data was represented as % viability compared to DMSO treated control for a total of 5 days.

For colony formation assay, approximately 500 cells/well of GBM143 were seeded onto a six- well plate. After overnight incubation cells were treated with **5c** or Temozolomide (TMZ) at 3 µM and 50 µM concentrations respectively. The control wells were treated with DMSO. Media was replaced every 3 days along with drug treatment. After 15 days colonies were fixed with methanol and stained using crystal violet (0.5% w/v in deionized water).

1.3.5 Spheroid invasion assay

3D spheroid invasion assay was carried out as stated previously[10]. Briefly, 2,000 GBM6 cells were seeded per well in a low attachment u-bottom 96 well plate (Thermo #174925) and spheroids were allowed to form over four days. These spheroids were embedded in Geltrex (Thermo #A1413302) which is a basement membrane extract that contains laminin, collagen IV, entactin, and heparin sulphate proteoglycans. The embedded spheroids were supplemented with complete media at top with or without drugs. The photomicrographs of each well containing spheroid were taken at x100 resolution starting from day 0 (day of embedding the spheroid in matrix) through day 4 or day 6. The relative invasion was calculated using Image J software.

1.3.6 Cell cycle analyses

Cell cycle analyses using propidium iodide and flow cytometry were carried out as described previously[3, 9]. Asynchronous GBM6 cells were treated with either DMSO or **5c** at 3 µM for 16 hr. Post treatment, cells were washed with PBS+1% FBS and resuspended in flow cytometry tubes. Cells were then fixed with 70% ice-cold ethanol and propidium iodide (50 µg/ml) was added to the cells and incubated in the dark at room temperature (25°C) for 30 min. The cell populations were then subjected to quantitative measurement of DNA content by flow cytometry using a FACSFortessa (BD Biosciences) and cell cycle distribution and the percentage of G_2/M–S–G_1 cells to the total cell events were determined by the BD FACS Diva software. Stacked bar graphs were derived using Graphpad Prism.

1.3.7 Statistical Analysis

All statistical analysis was done using GraphPad Prism statistical package and presented as mean ± SD unless otherwise stated. Figure legends contain details of the statistical tests and multiple comparisons conducted throughout. Experiments were repeated 2-3 times with multiple technical replicates for the appropriate statistical tests to be conducted.

Figure **S1**: ^{1}H-NMR (500 MHz, DMSO-d_6) of Compound **5a**.

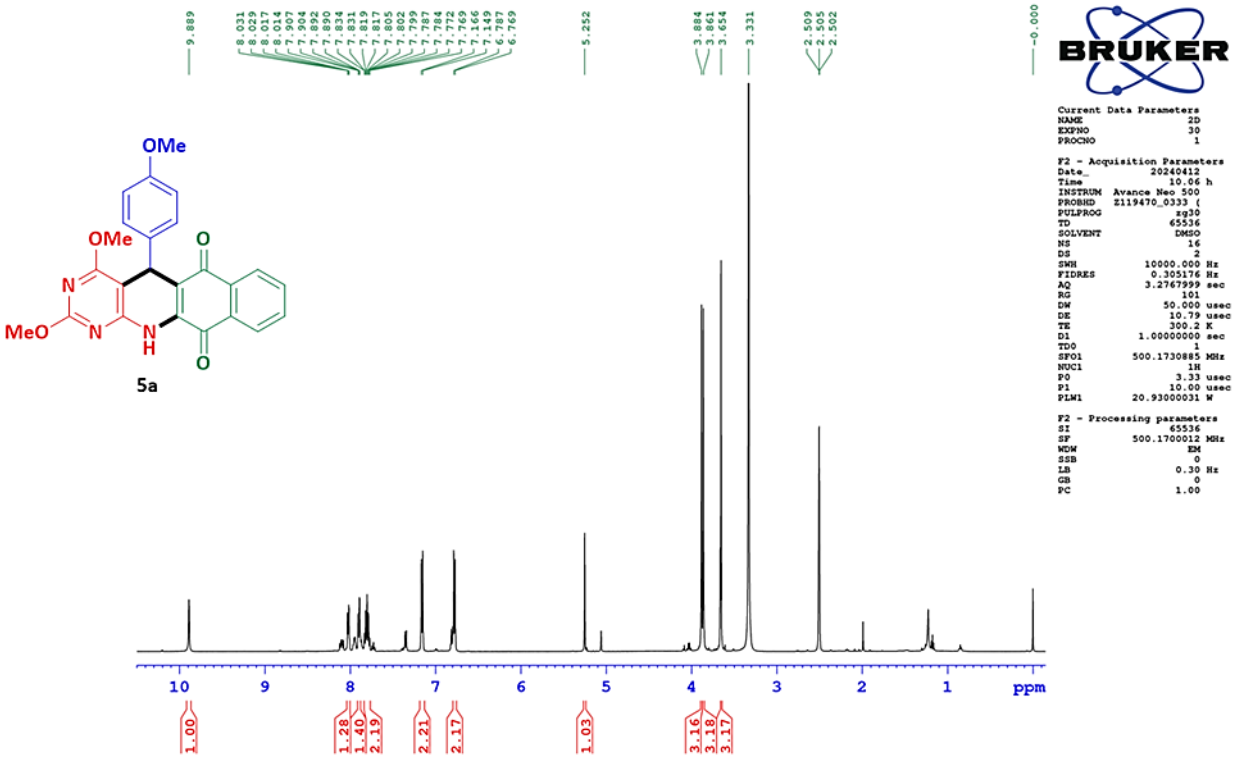


Figure **S2**: ^{13}C-NMR (125 MHz, DMSO-d_6) of Compound **5a**.

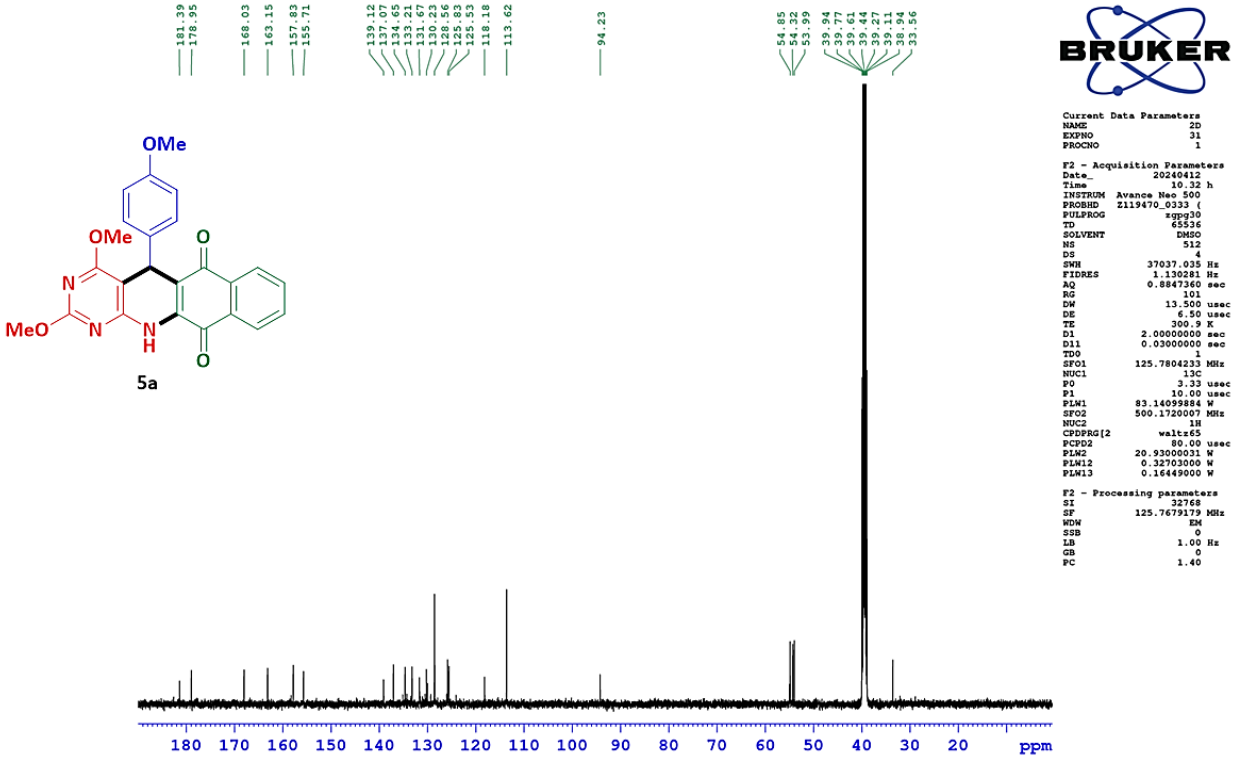


Figure **S3**: ^{13}C-DEPT 135 NMR (125 MHz, DMSO-d_6) of Compound **5a**.

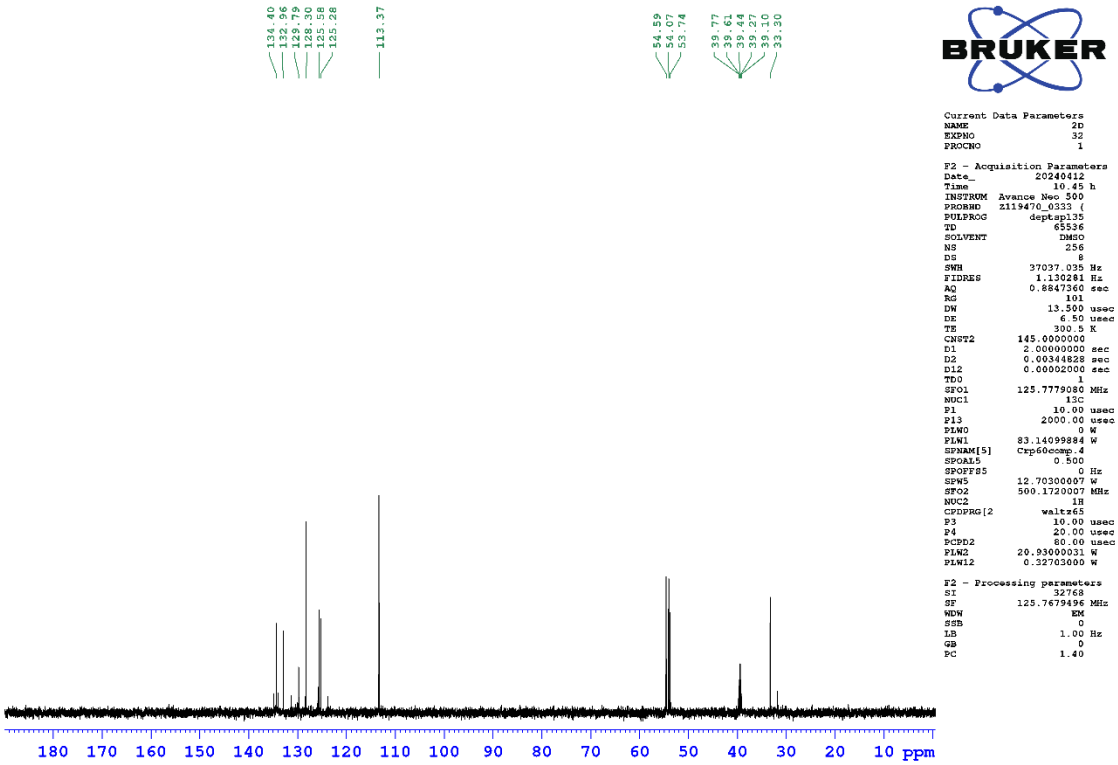


Figure S4: Structure of **5a** with labelled C & H atoms and selected HMBC correlation

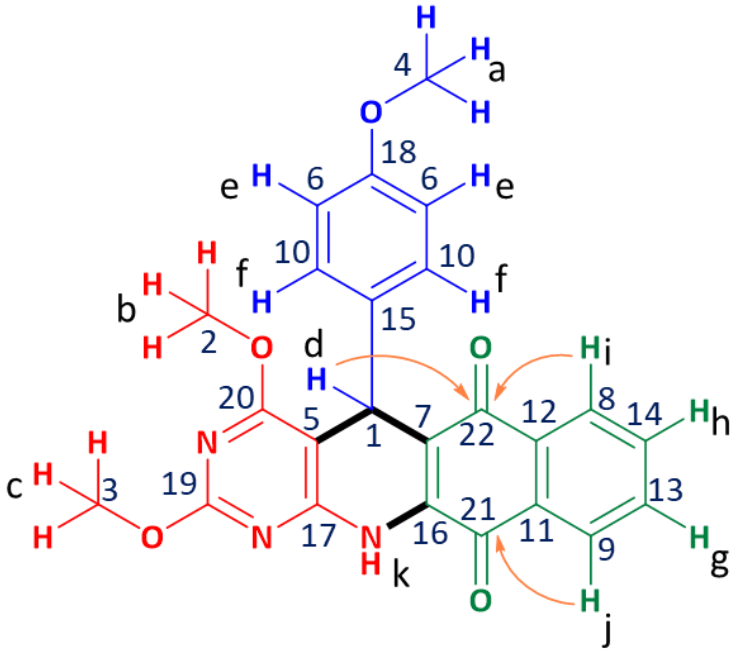


Table S3. HSQC & HMBC NMR data of Compound **5a**.		
	^{1}H - ^{13}C HSQC	**^{1}H - ^{13}C HMBC**
Proton (δ ppm)	Carbon (δ ppm)	Carbon (δ ppm)
H-a (δ 3.65)	C-2 (δ 53.99)	C-18 (δ 157.83)
H-b (δ 3.86)	C-3 (δ 54.32)	C-20 (δ 168.03)
H-c (δ 3.88)	C-4 (δ 54.85)	C-19 (δ 163.15)
H-d (δ 5.25)	C-1 (δ 33.56)	C-10 (δ 128.56), C-20 (δ 168.03), C-17 (δ 155.71), C-16 (δ 139.12), C-5 (δ 94.23), C-7 (δ 118.18), C-15 (δ 137.07) & C-22 (δ 181.39)
H-e (δ 6.77)	C-6 (δ 113.62)	C-6 (δ 113.62), C-15 (δ 137.07), C-10 (δ 128.56),
H-f (δ 7.15)	C-10 (δ 128.56)	C-1 (δ 33.56), C-6 (δ 113.62), C-10 (δ 128.56),
H-g (δ 7.78)	C-13 (δ 133.21)	C-11 (δ 130.23), C-8 (δ 125.53)
H-h (δ 7.80)	C-14 (δ 134.65)	C-12 (δ 131.67), C-9 (δ 125.83)
H-i (δ 7.89)	C-8 (δ 125.53)	C-11 (δ 130.23), C-13 (δ 133.21) & C-22 (δ 181.4)
H-j (δ 8.02)	C-9 (δ 125.83)	C-12 (δ 131.67), C-14 (δ 134.65), C-21 (δ 178.95)

Figure **S5**: HSQC Spectrum of Compound **5a**

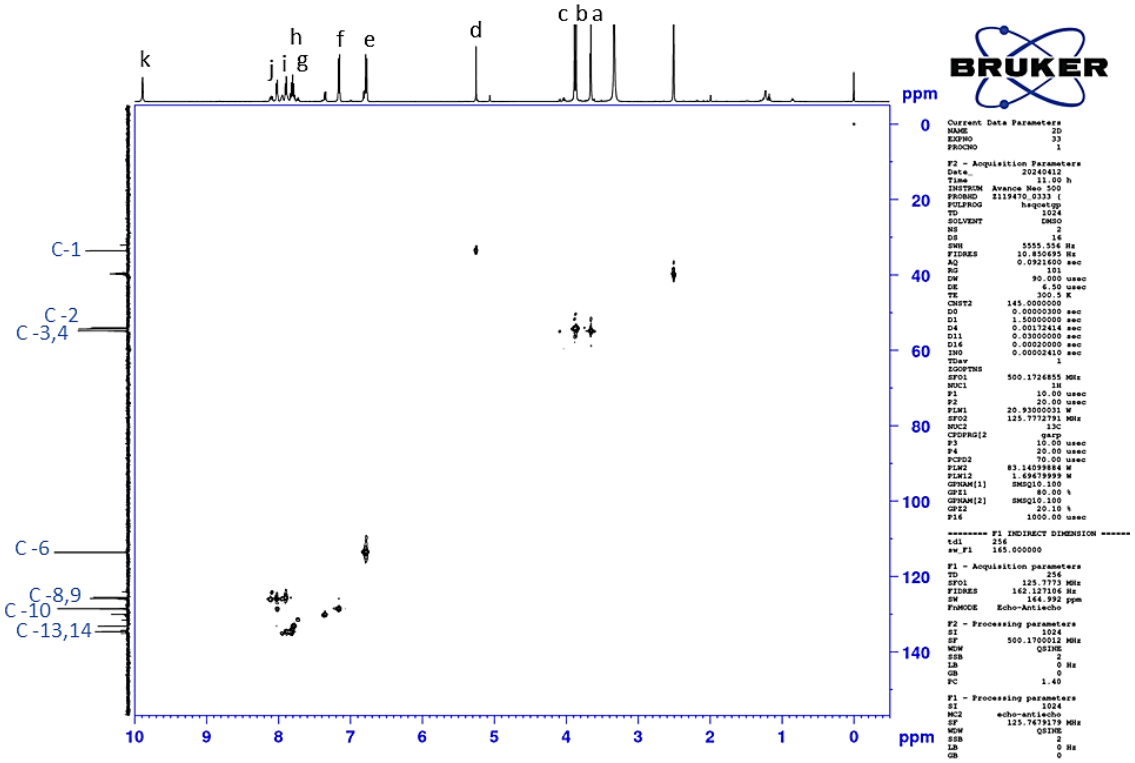


Figure **S6**: HMBC Spectrum of Compound **5a**.

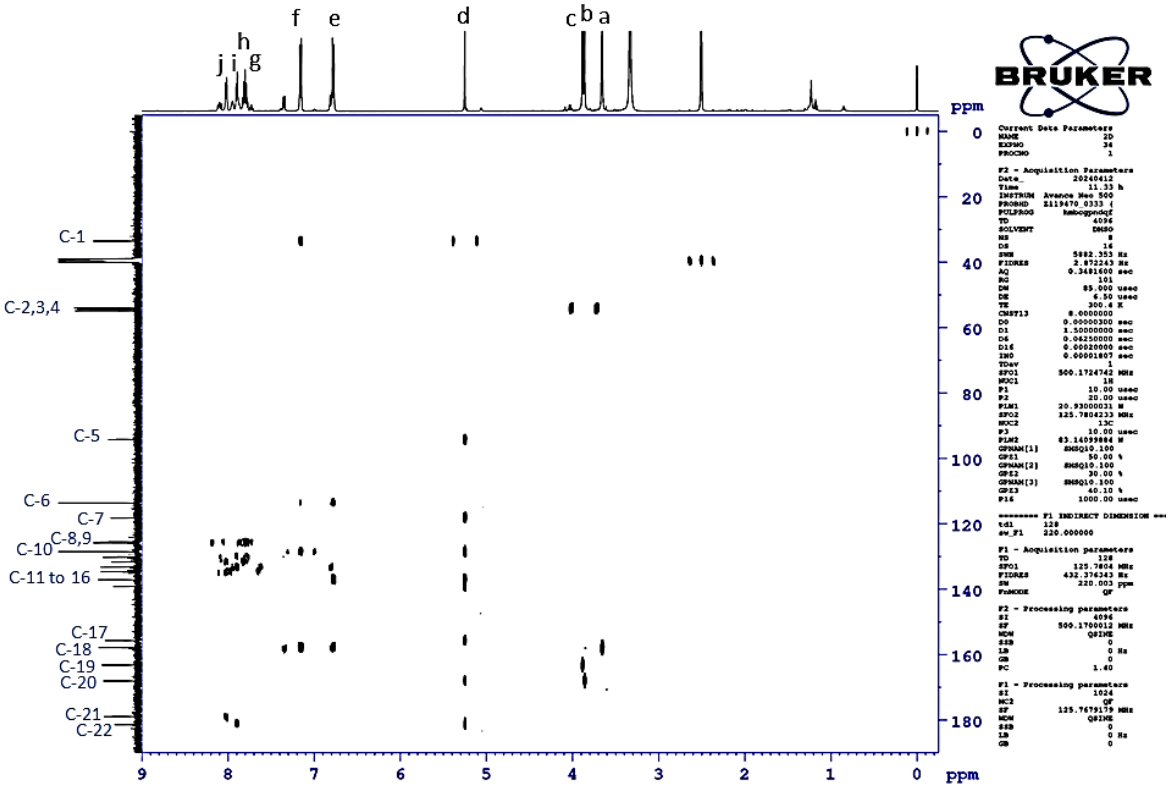


Figure **S7**: ^{1}H-NMR (500 MHz, DMSO-d_6) of Compound **6e**.

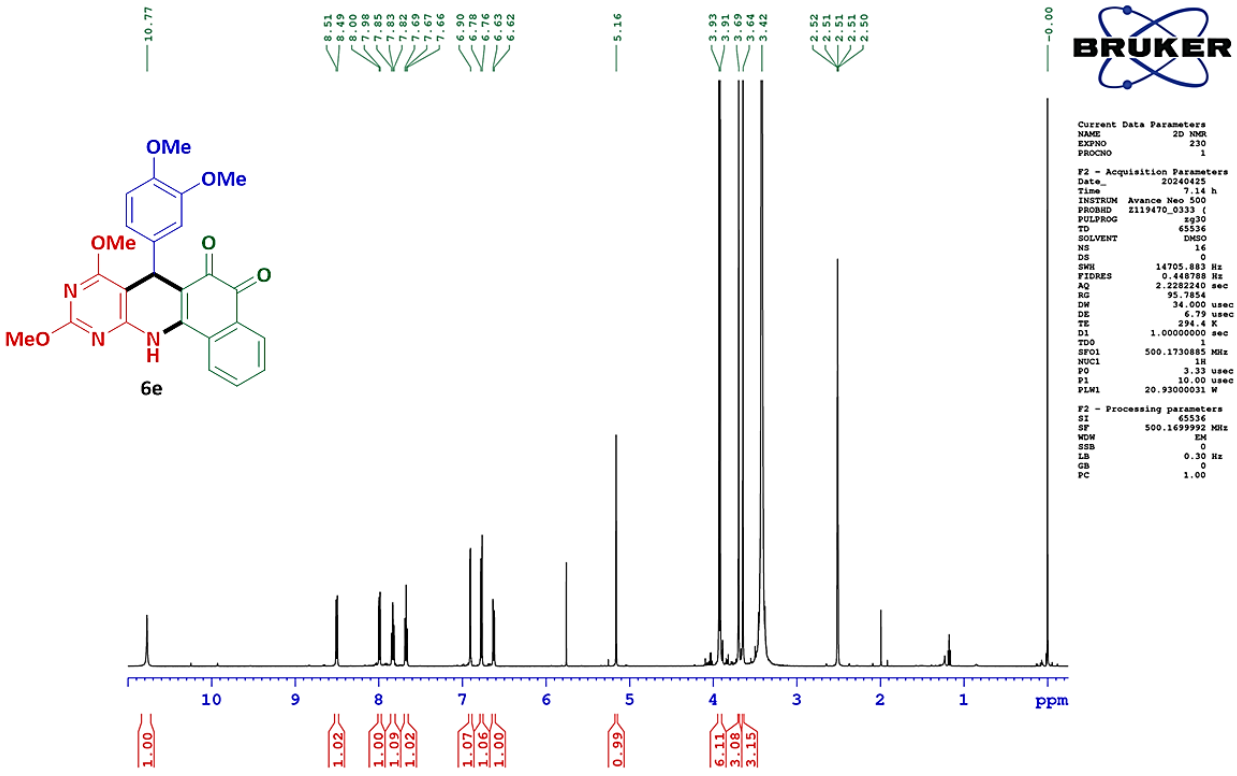


Figure **S8**: ^{13}C-NMR (125 MHz, DMSO-d_6) of Compound **6e**.

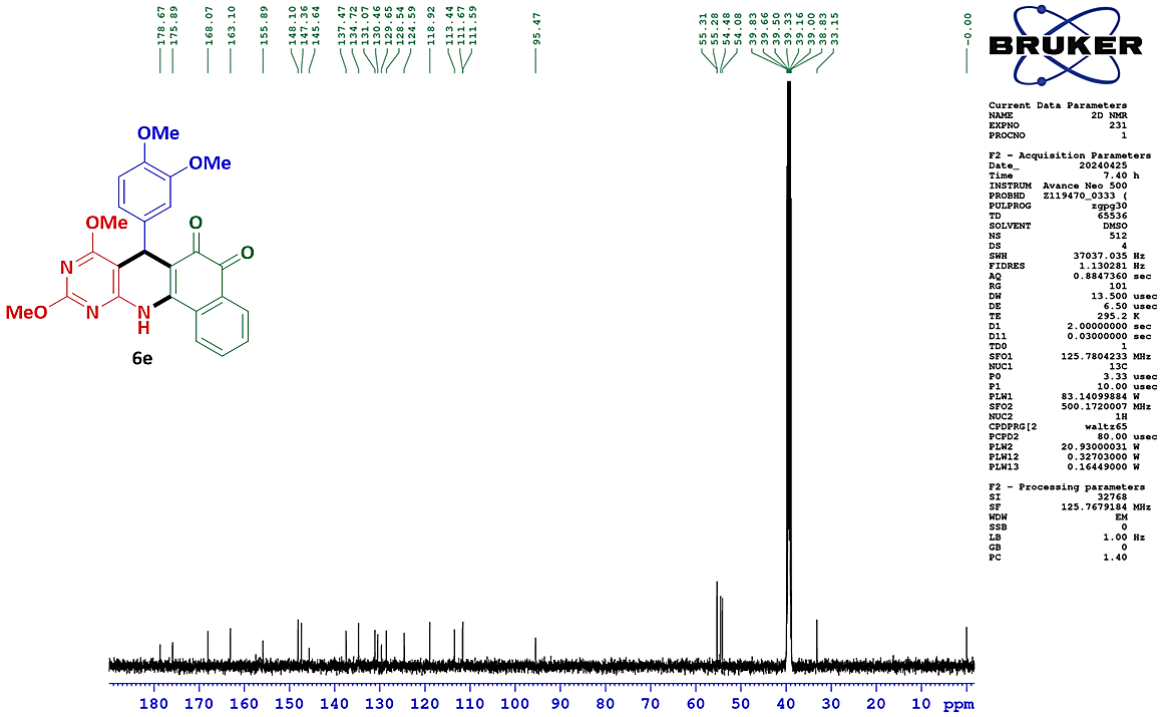


Figure **S9**: ^{13}C-DEPT 135 (125 MHz, DMSO-d_6) of Compound **6e**.

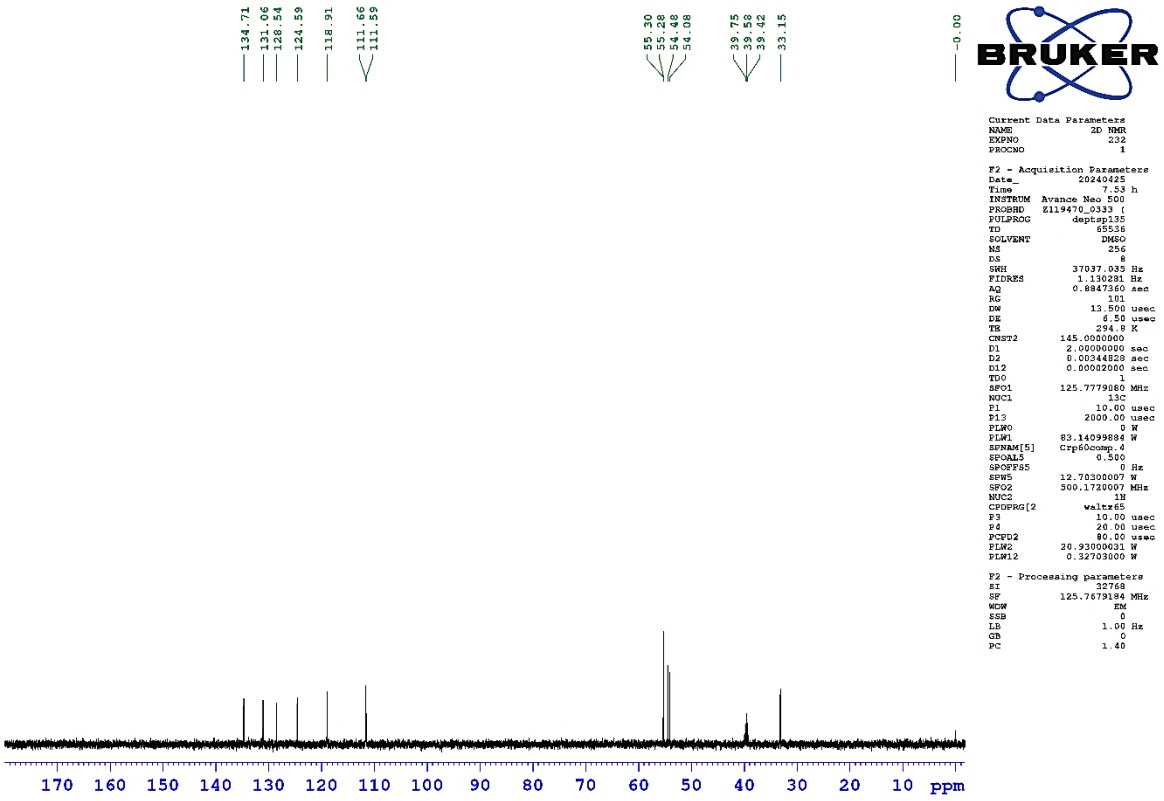


Figure S10: Structure of **6e** with labelled C & H atoms and selected HMBC correlation

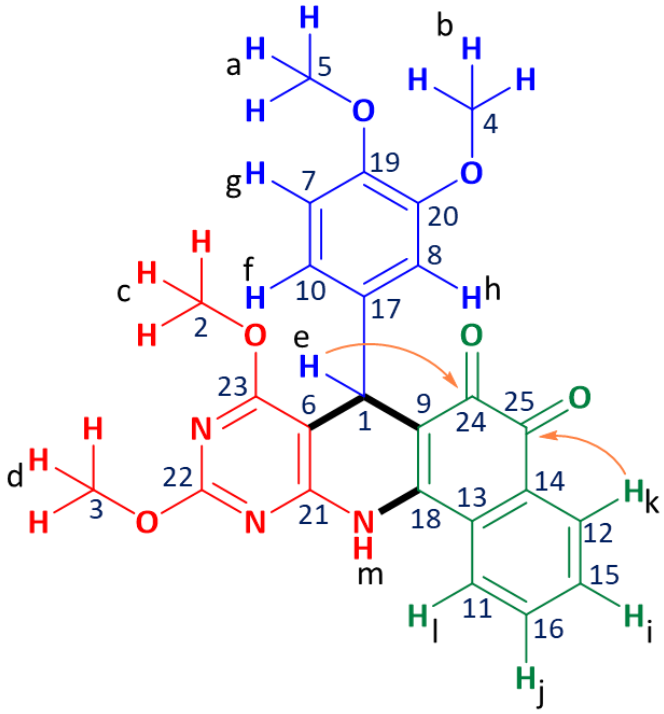


Table S4. HSQC & HMBC NMR data of Compound **6e**.		
	1H-13C HSQC	**1H-13C HMBC**
Proton (δ ppm)	Carbon (δ ppm)	Carbon (δ ppm)
H-a (δ 3.64)	C-5 (δ 55.31)	C-19 (δ 147.36)
H-b (δ 3.69)	C-4 (δ 55.28)	C-20 (δ 148.10)
H-c (δ 3.91)	C-2 (δ 54.08)	C-23 (δ 168.07)
H-d (δ 3.93)	C-3 (δ 54.48).	C-22 (δ 163.10)
H-e (δ 5.16)	C-1 (δ 33.15)	C-8 (δ 111.67), C-10 (δ 118.92), C-23 (δ 168.07), C-21 (δ 155.89), C-18 (δ 145.64), C-6 (δ 95.47), C-9 (δ 113.44), C-17 (δ 137.47) & C-24 (δ 175.89)
H-f (δ 6.63)	C-10 (δ 118.92)	C-1 (δ 33.15), C-19 (δ 147.36), C-8 (δ 111.67),
H-g (δ 6.77)	C-7 (δ 111.59)	C-17 (δ 137.47), C-20 (δ 148.10)
H-h (δ 6.90)	C-8 (δ 111.67)	C-1 (δ 33.15), C-10 (δ 118.92), C-19 (δ 147.36)
H-i (δ 7.67)	C-15 (δ 131.07)	C-11 (δ 124.59), C-14 (δ 130.46)
H-j (δ 7.83)	C-16 (δ 134.71)	C-13 (δ 129.65), C-12 (δ 128.54)
H-k (δ 7.99)	C-12 (δ 128.54)	C-16 (δ 134.72), C-13 (δ 129.65) & C-25 (δ 178.67)
H-l (δ 8.50)	C-11 (δ 124.59)	C-18 (δ 145.7), C-14 (δ 130.46) & C-15 (δ 131.07)

Figure **S11**: HSQC spectrum of Compound **6e**.

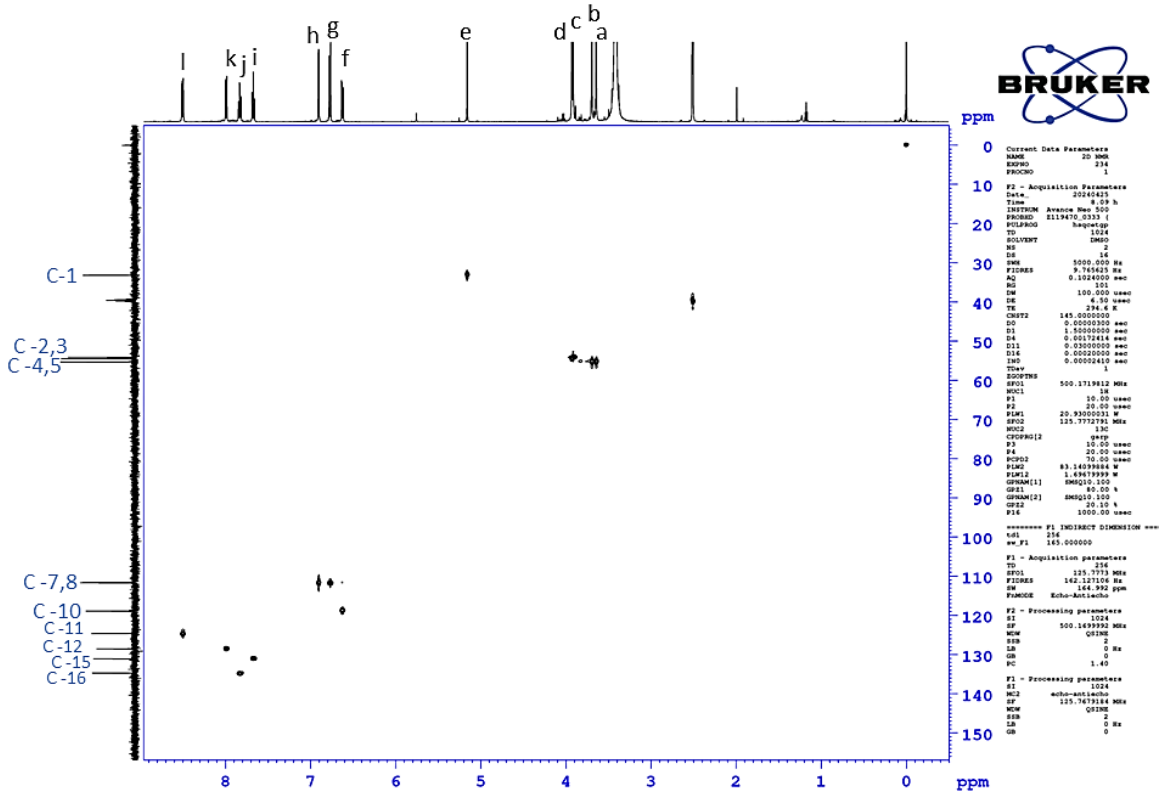


Figure **S12**: HMBC spectrum of Compound **6e**.

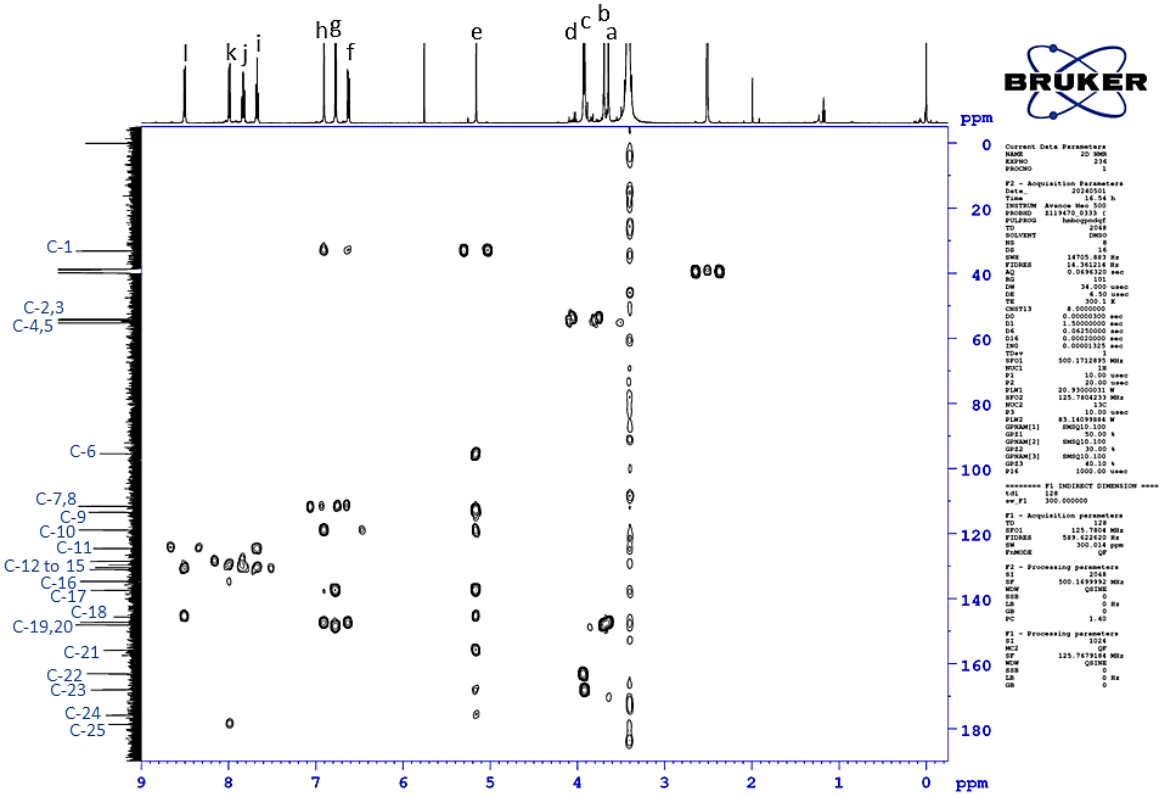


Figure **S13**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6a**.

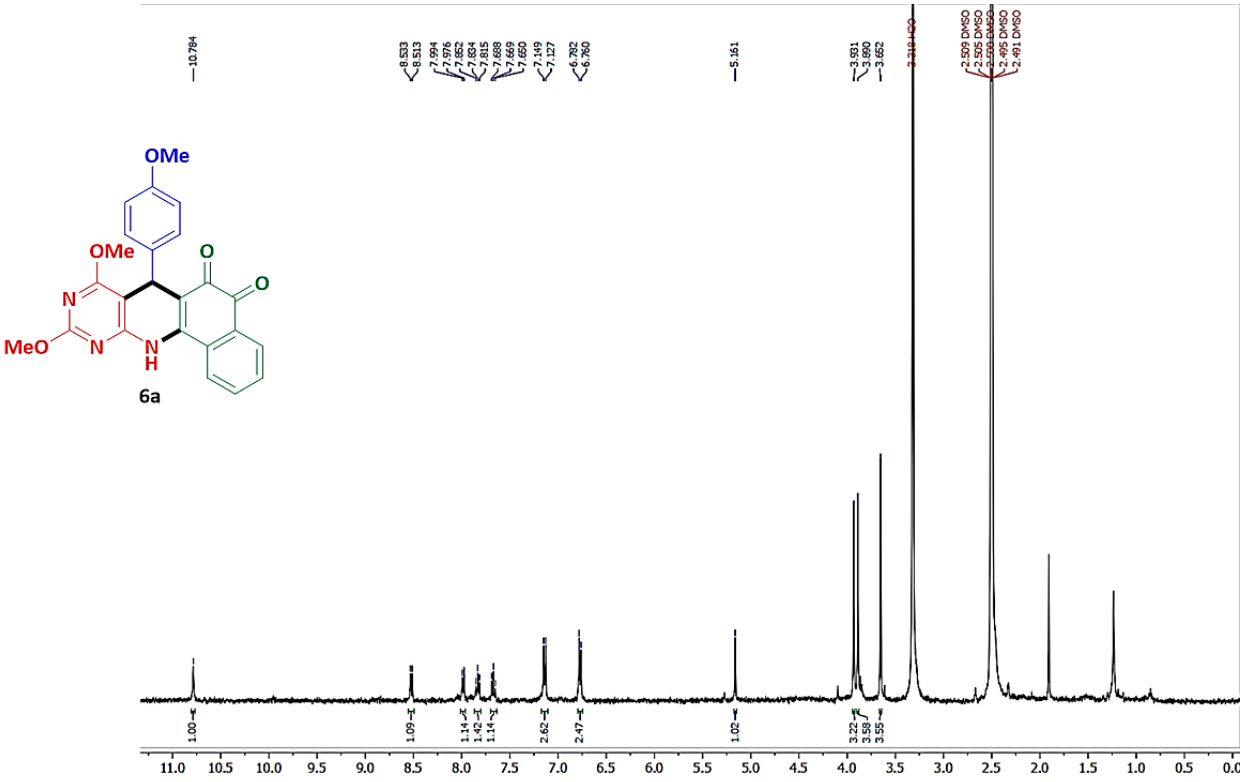


Figure **S14**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6a**.

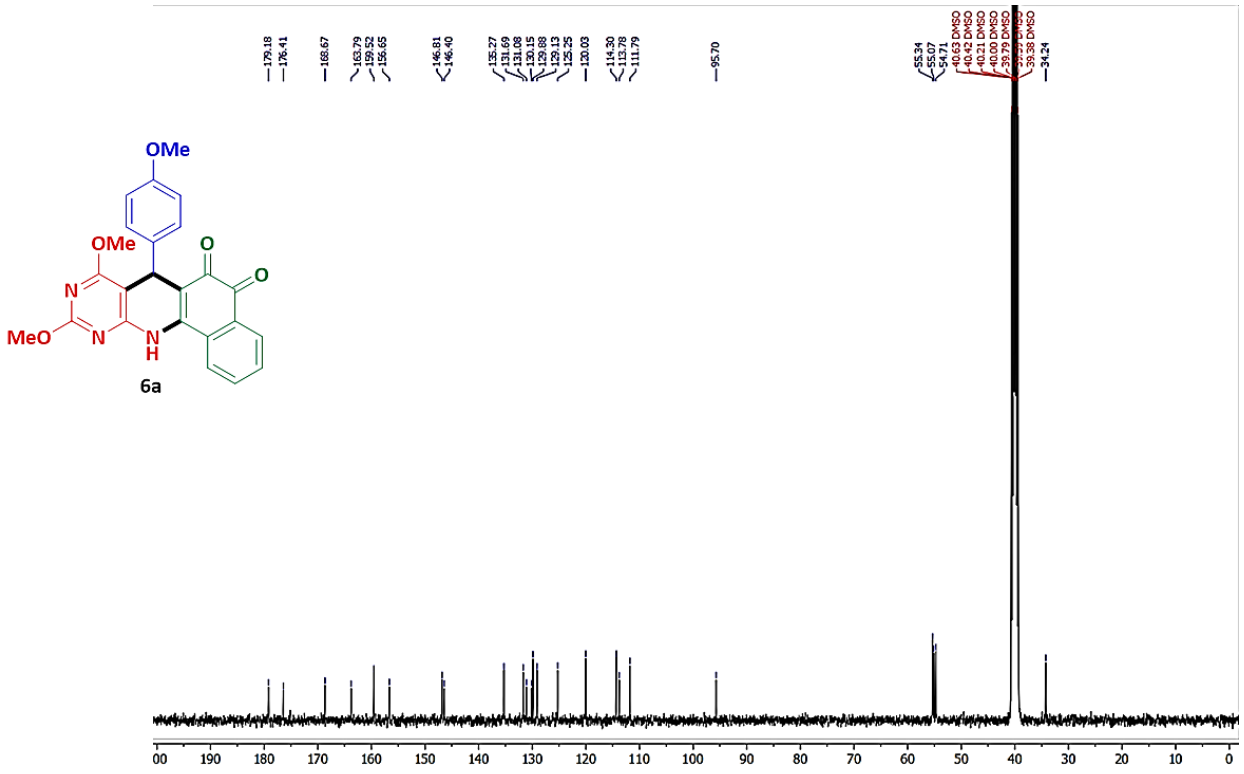


Figure **S15**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5b**.

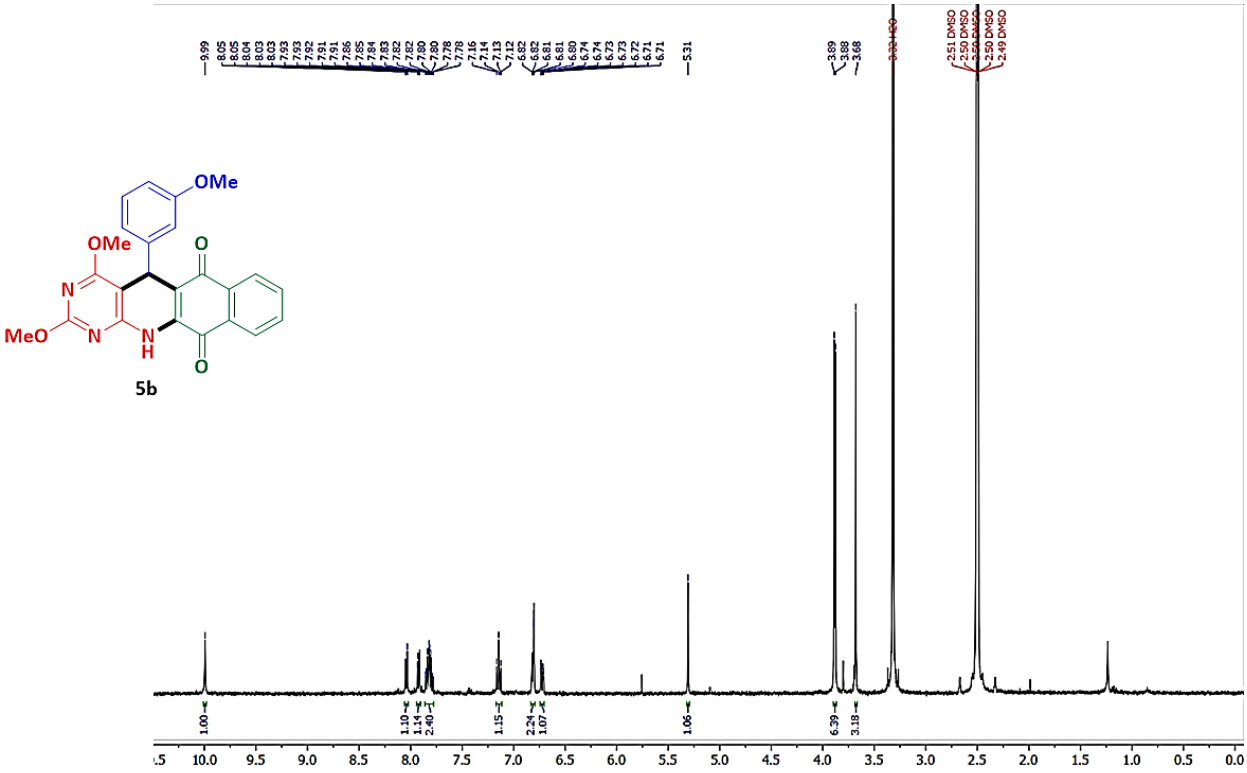


Figure **S16**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5b**.

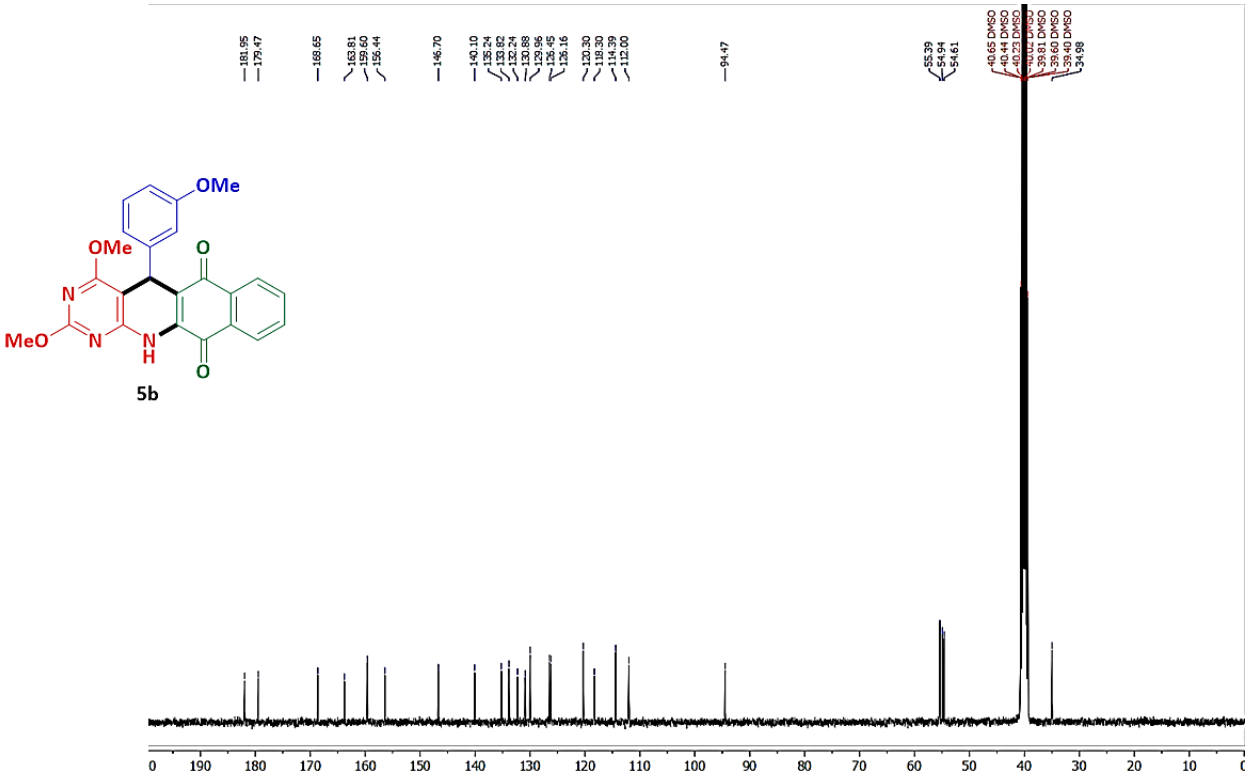


Figure **S17**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6b**.

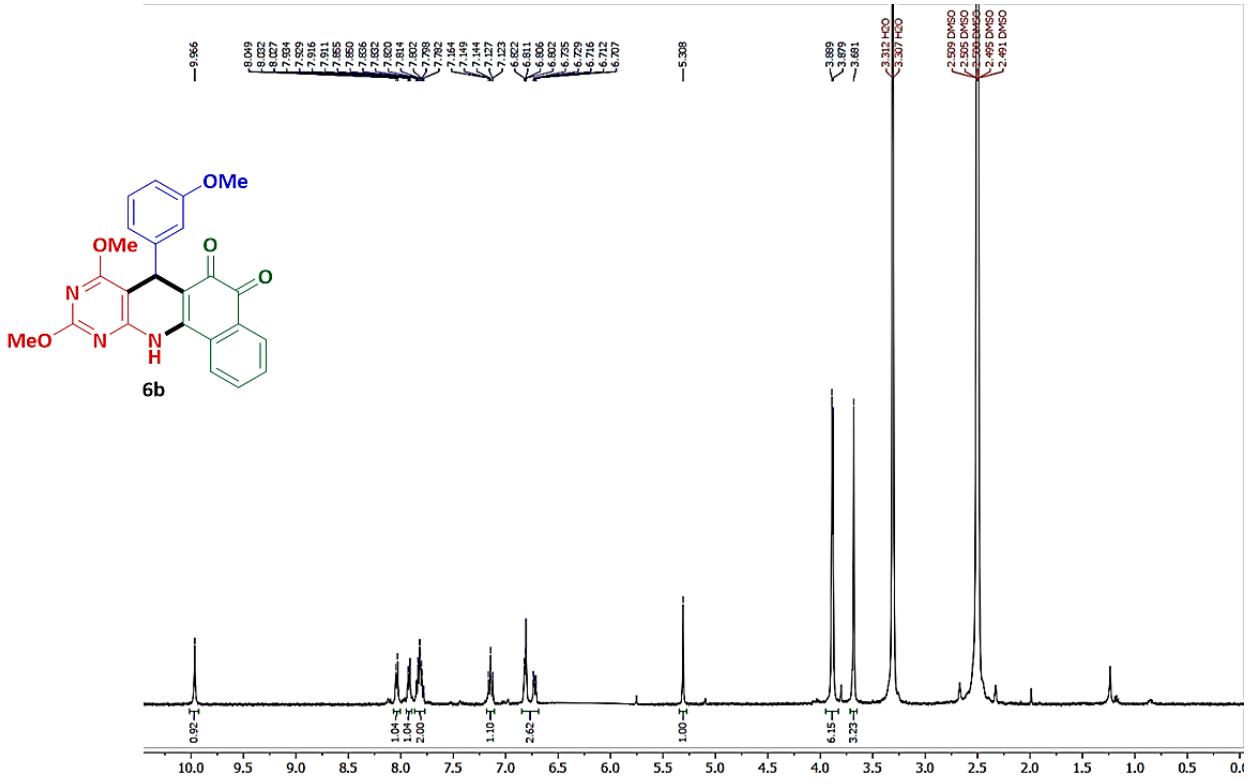


Figure **S18**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6b**.

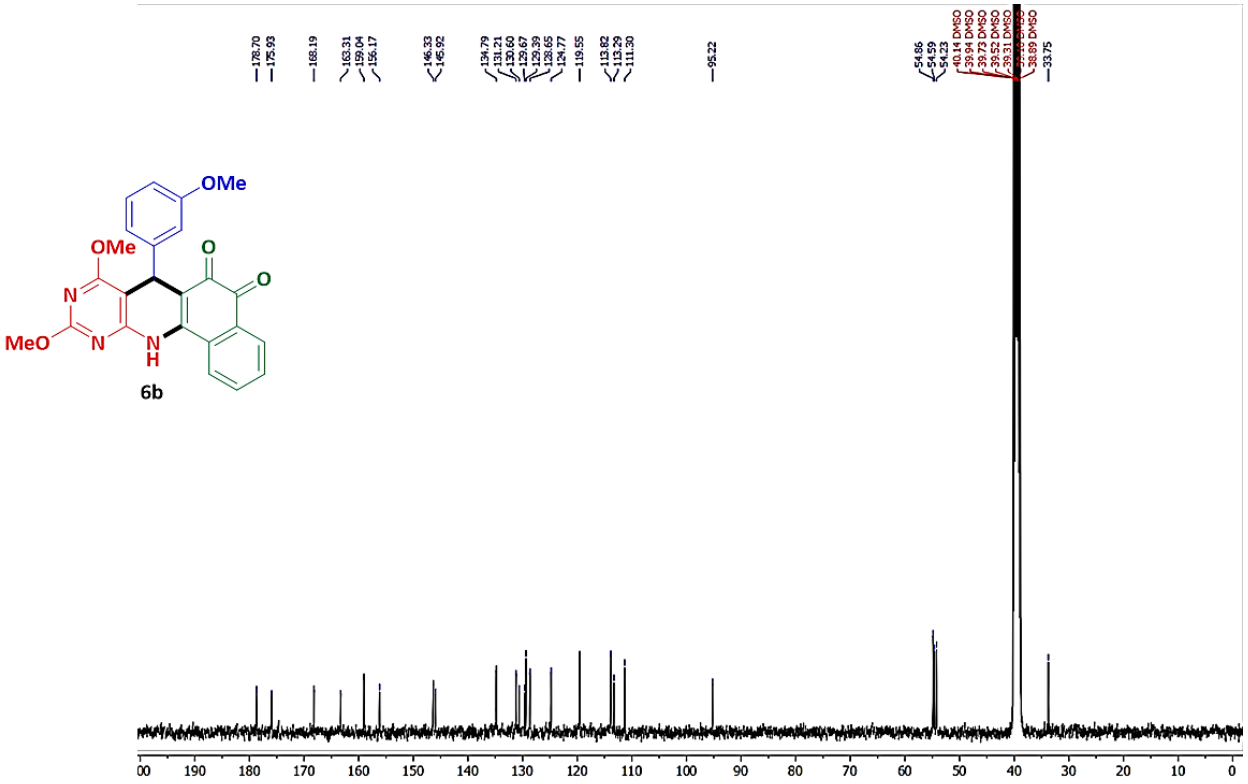


Figure **S19**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5c**.

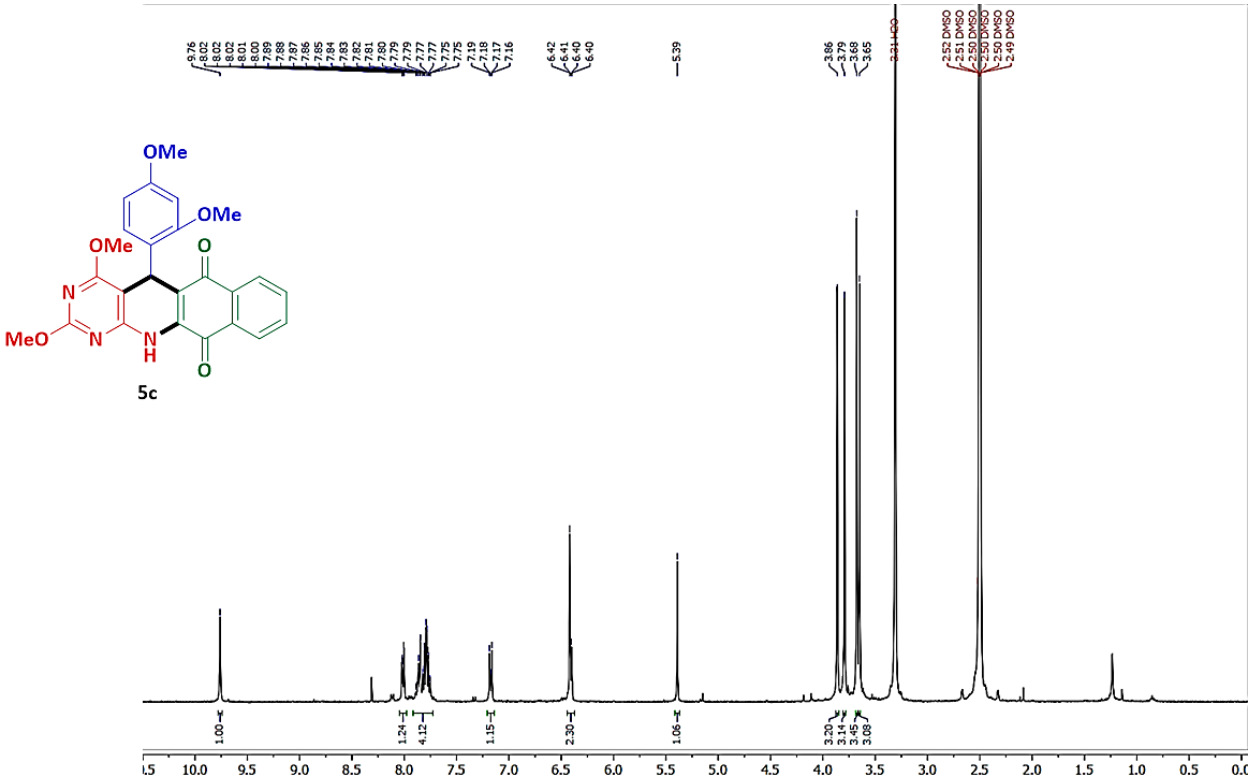


Figure **S20**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5c**.

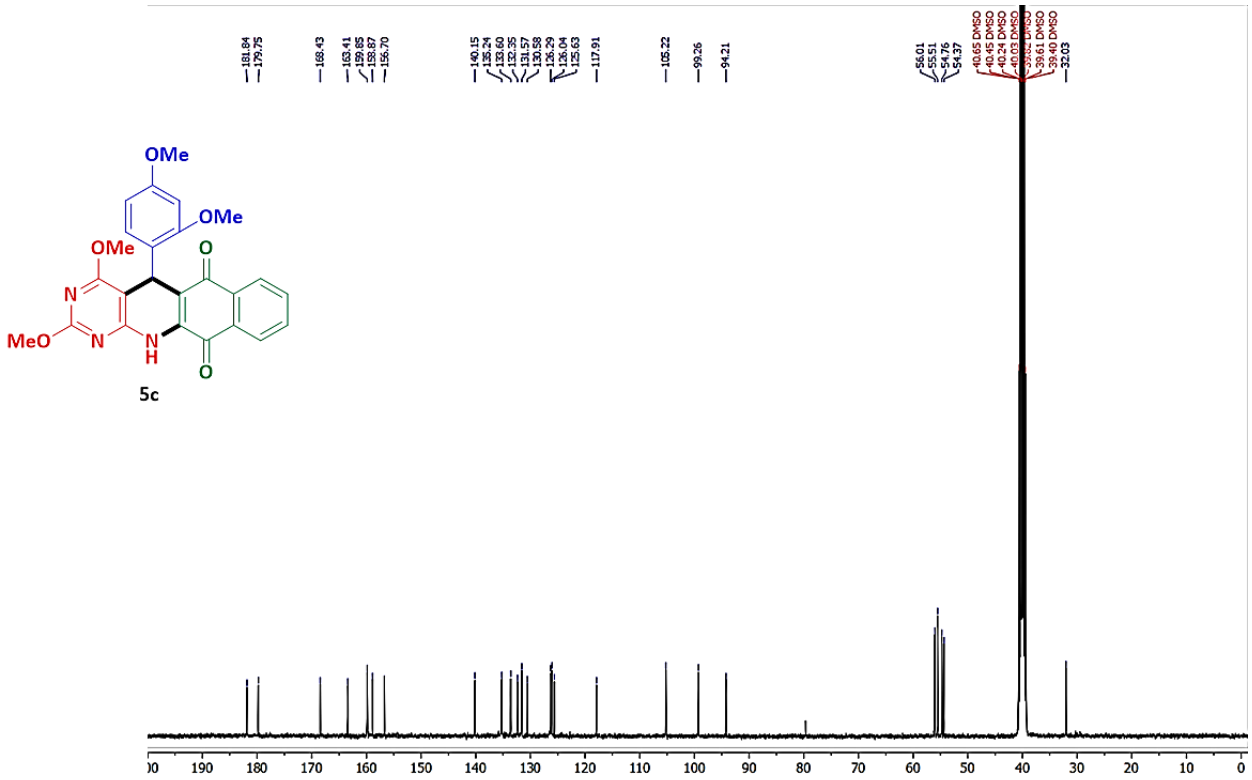


Figure **S21**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6c**.

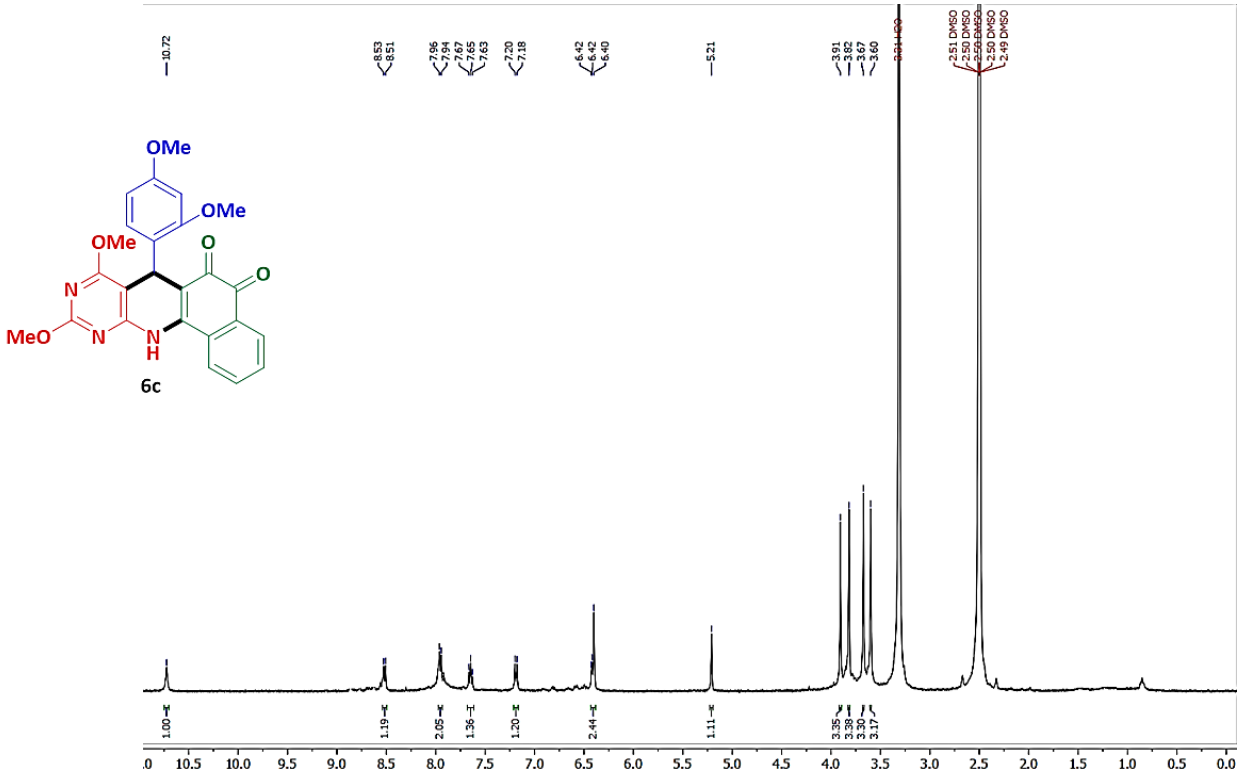


Figure **S22**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6c**.

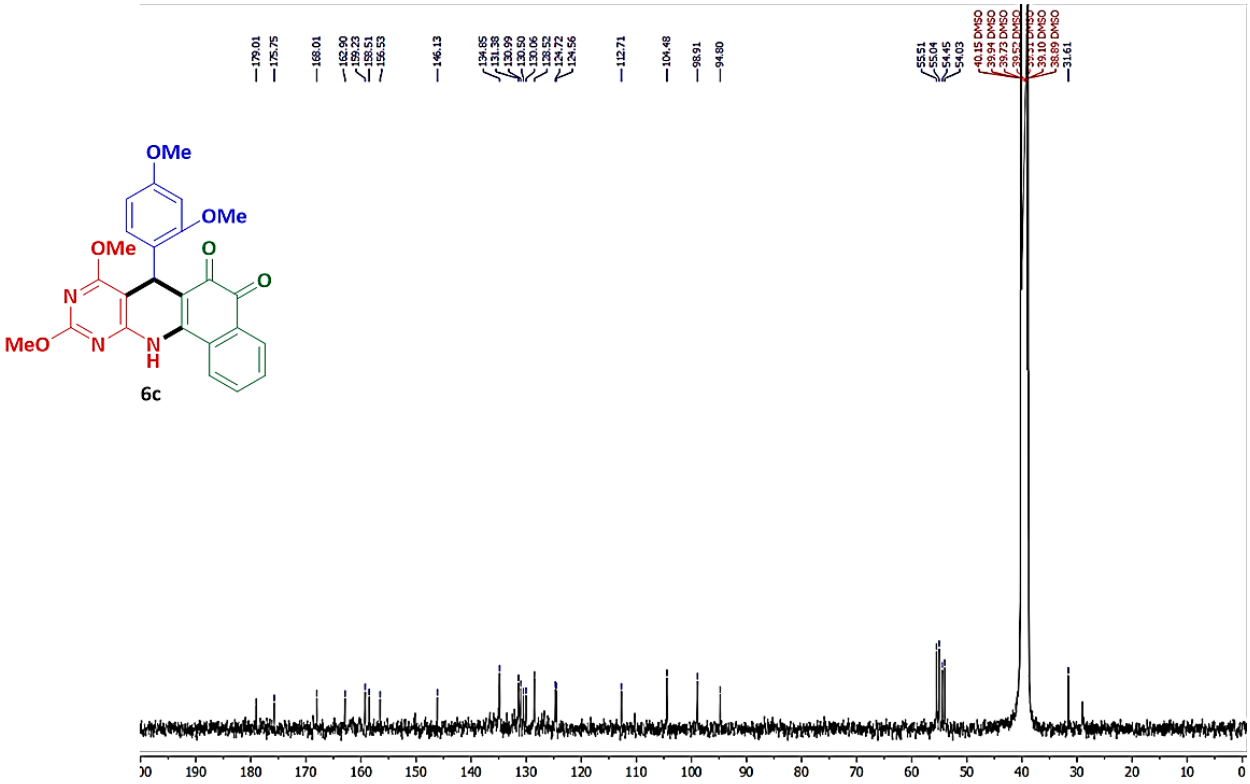


Figure **S23**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5d**.

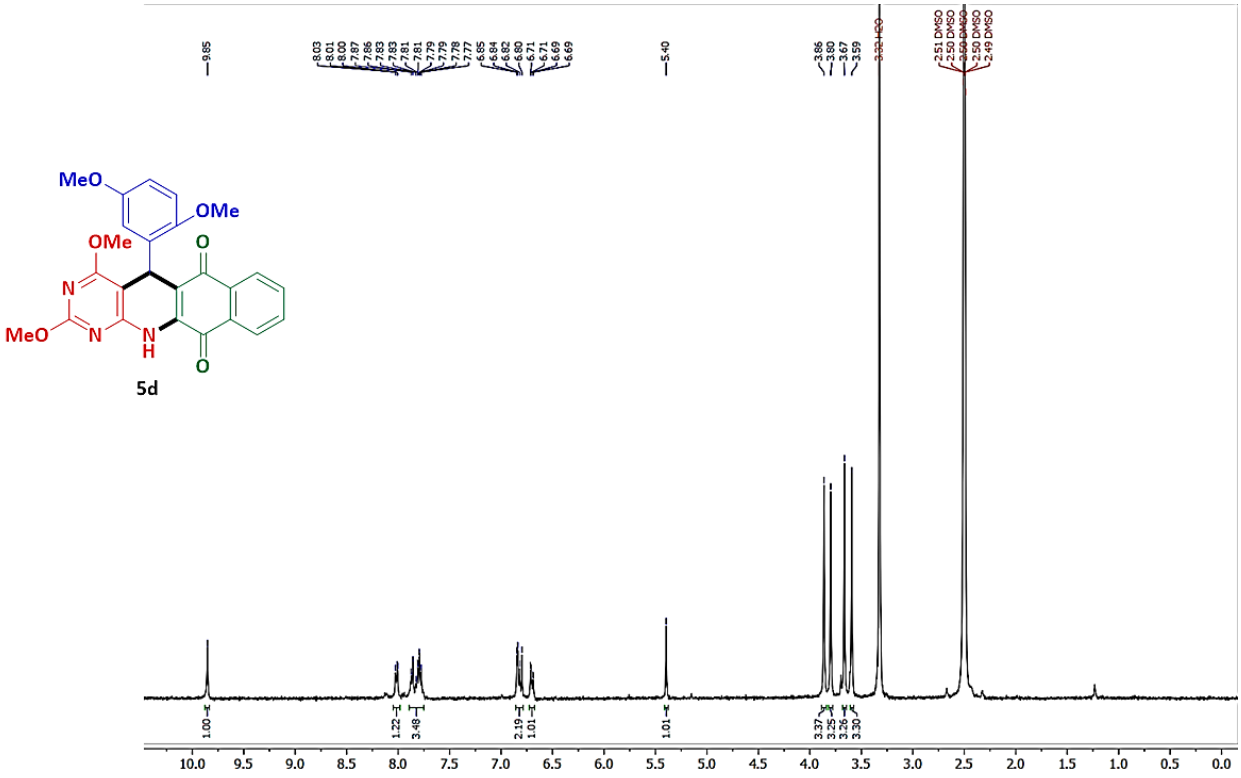


Figure **S24**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5d**.

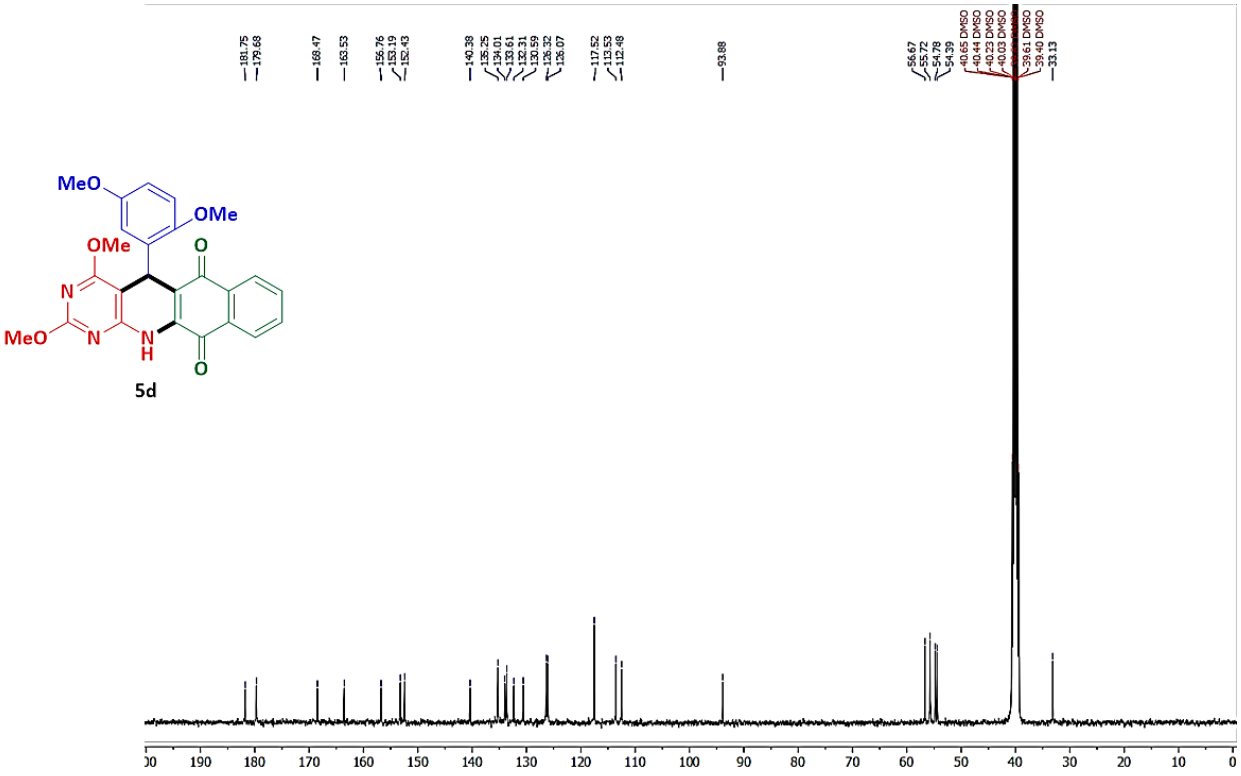


Figure **S25**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6d**.

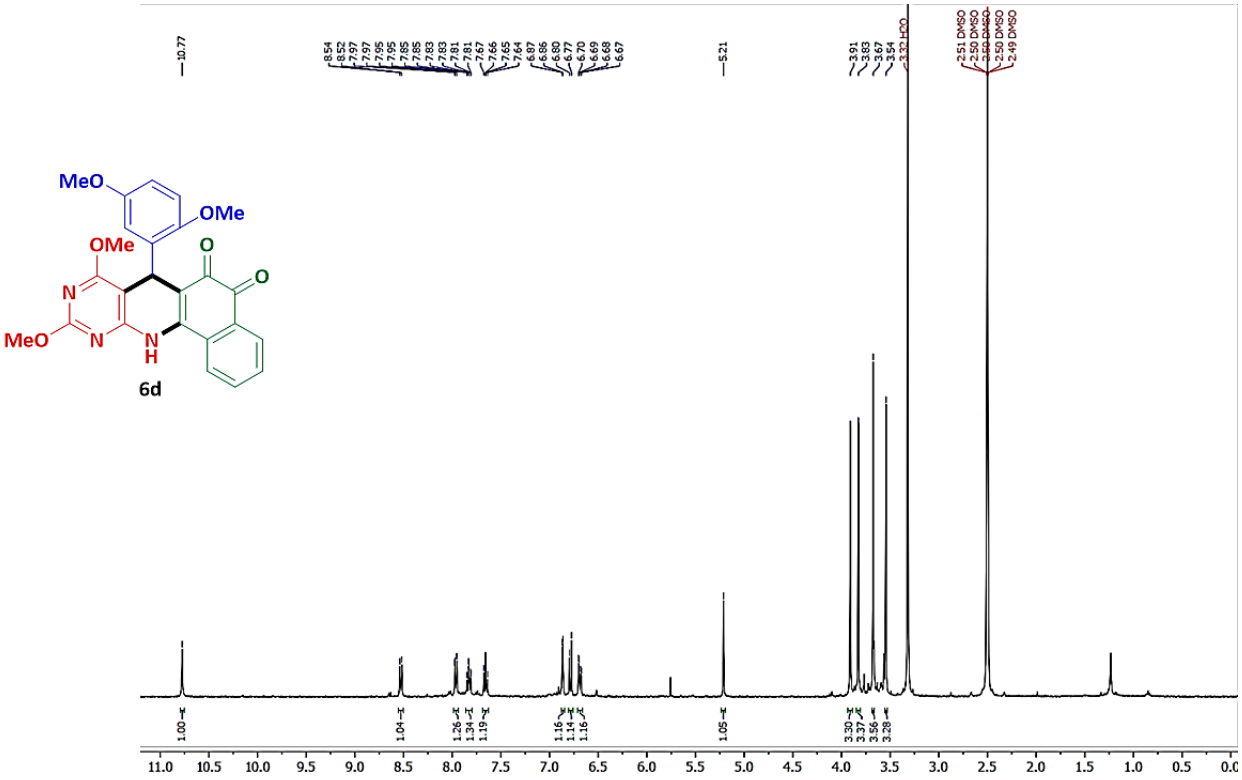


Figure **S26**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6d**.

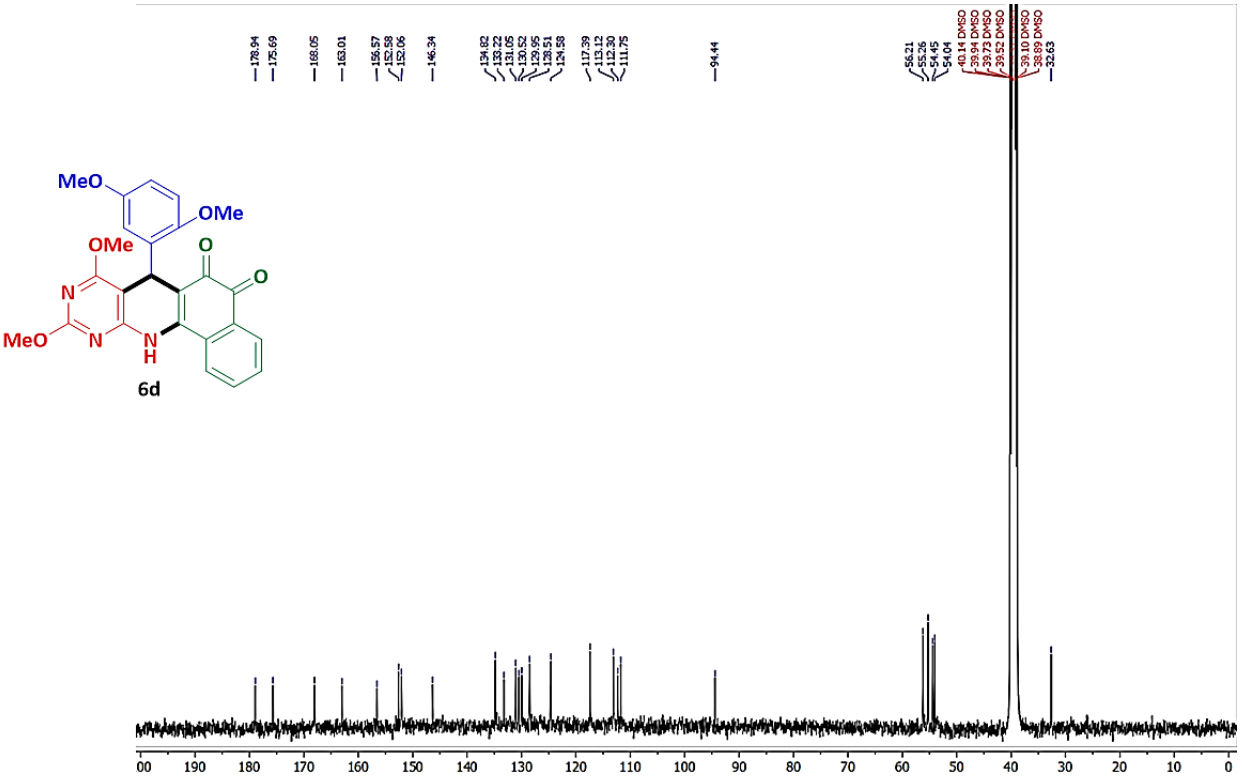


Figure **S27**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5e**.

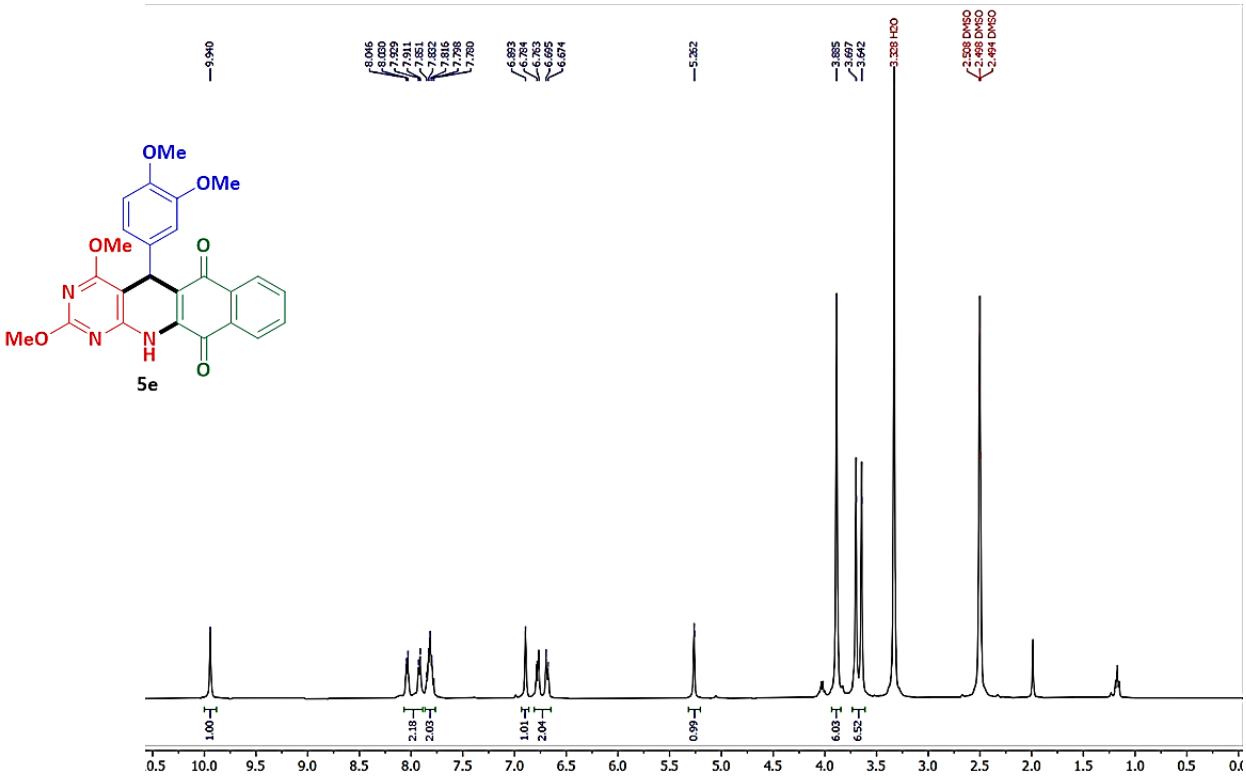


Figure **S28**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5e**.

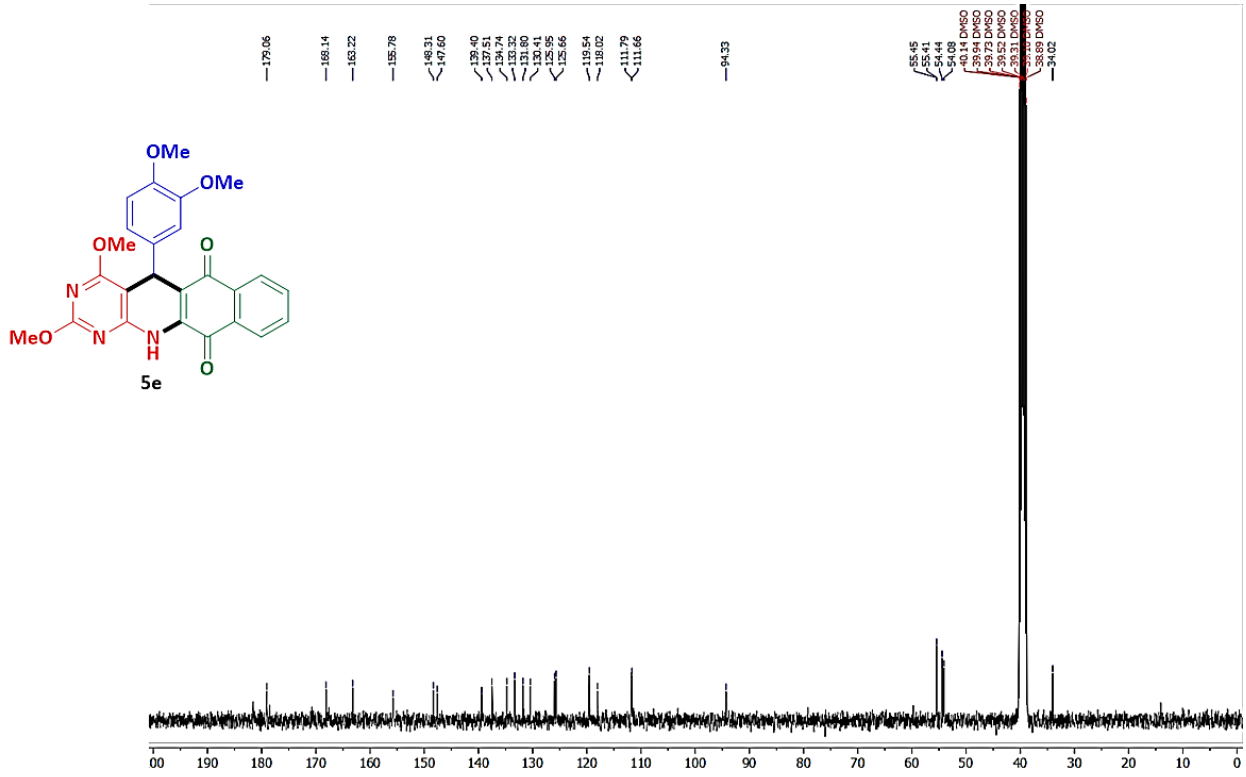


Figure **S29**: ^{1}H-NMR (500 MHz, DMSO-d_6) of Compound **6e**.

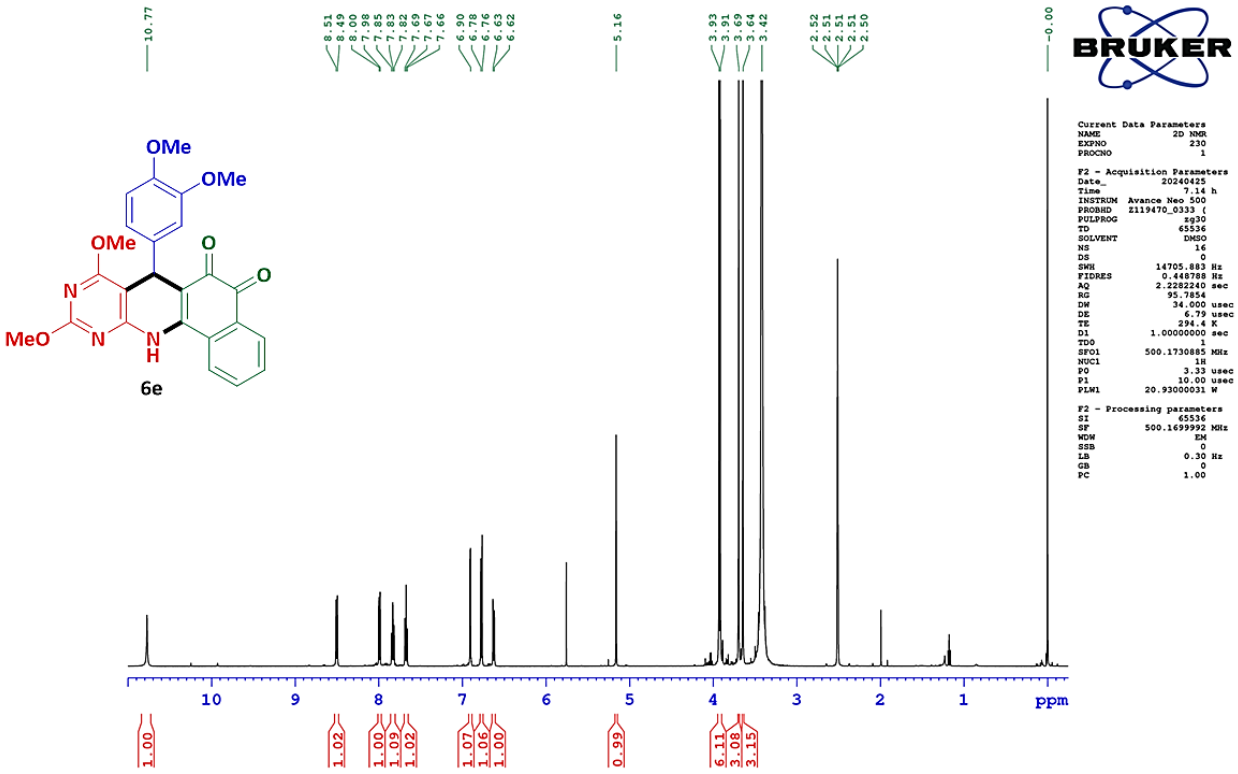


Figure **S30**: ^{13}C-NMR (125 MHz, DMSO-d_6) of Compound **6e**.

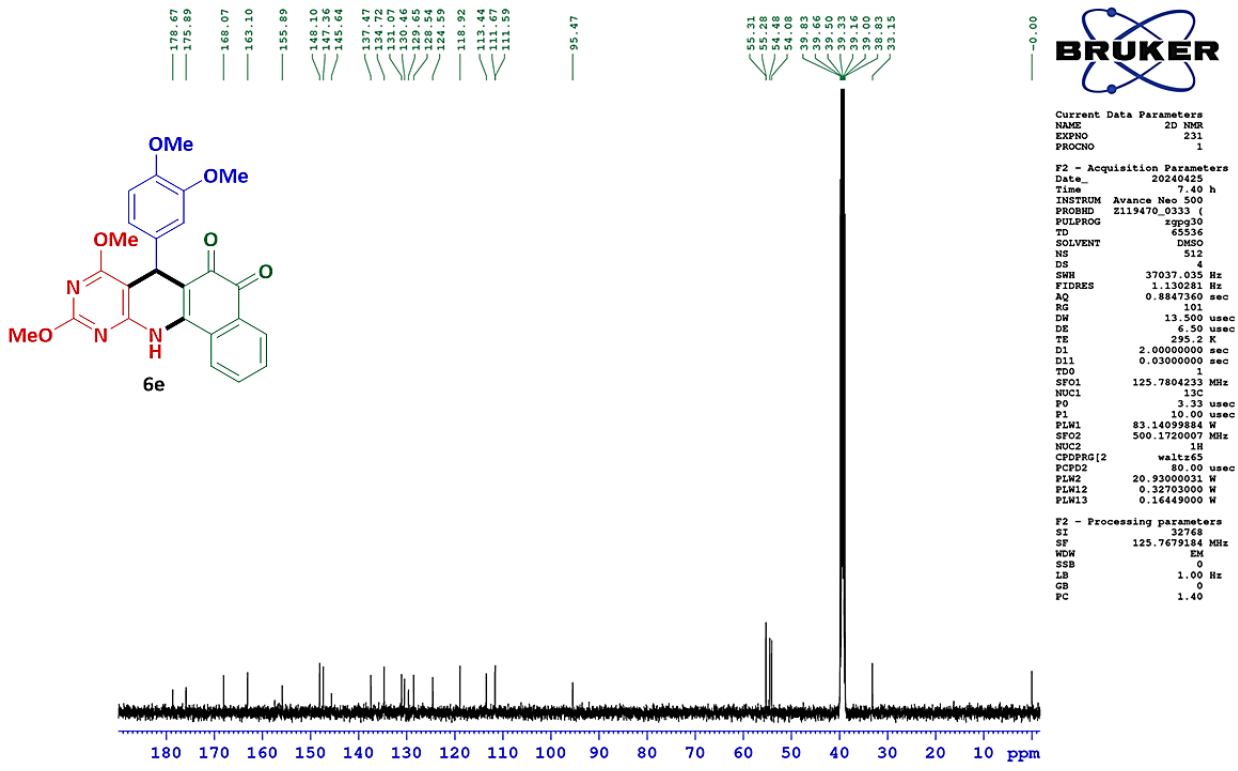


Figure **S31**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5f**.

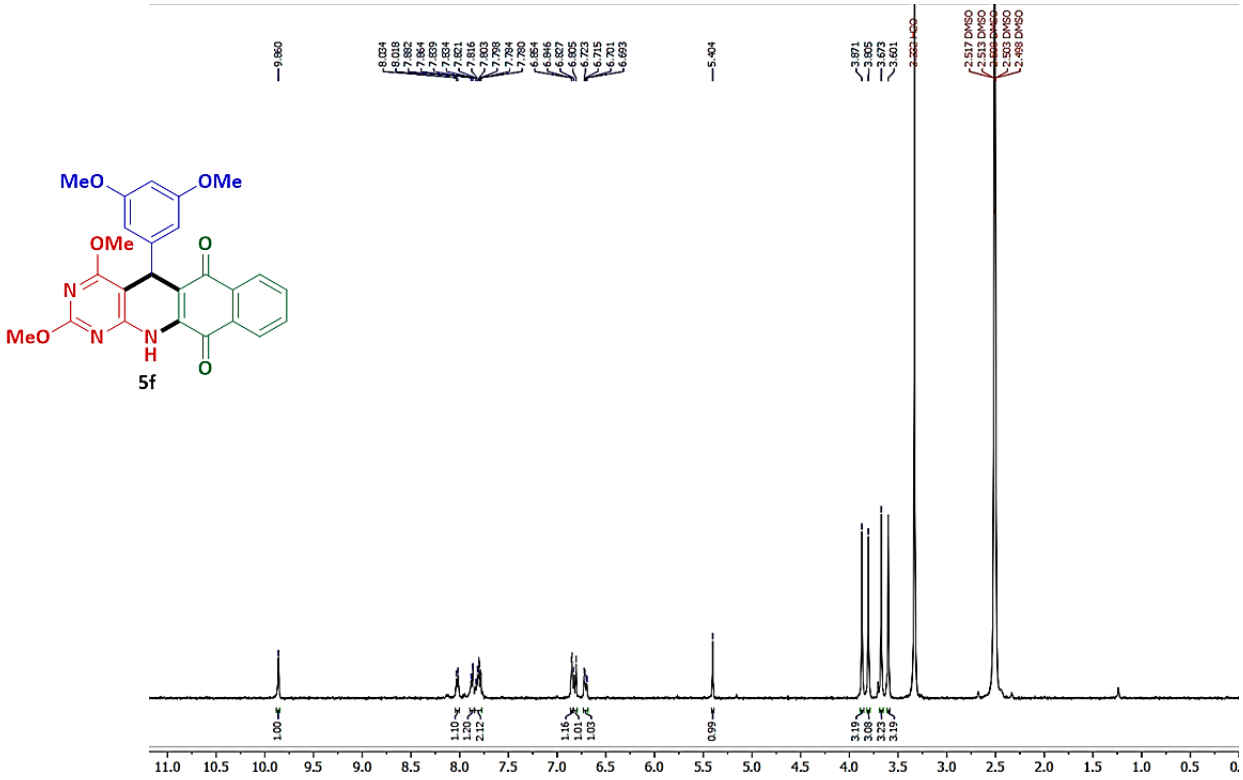


Figure **S32**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5f**.

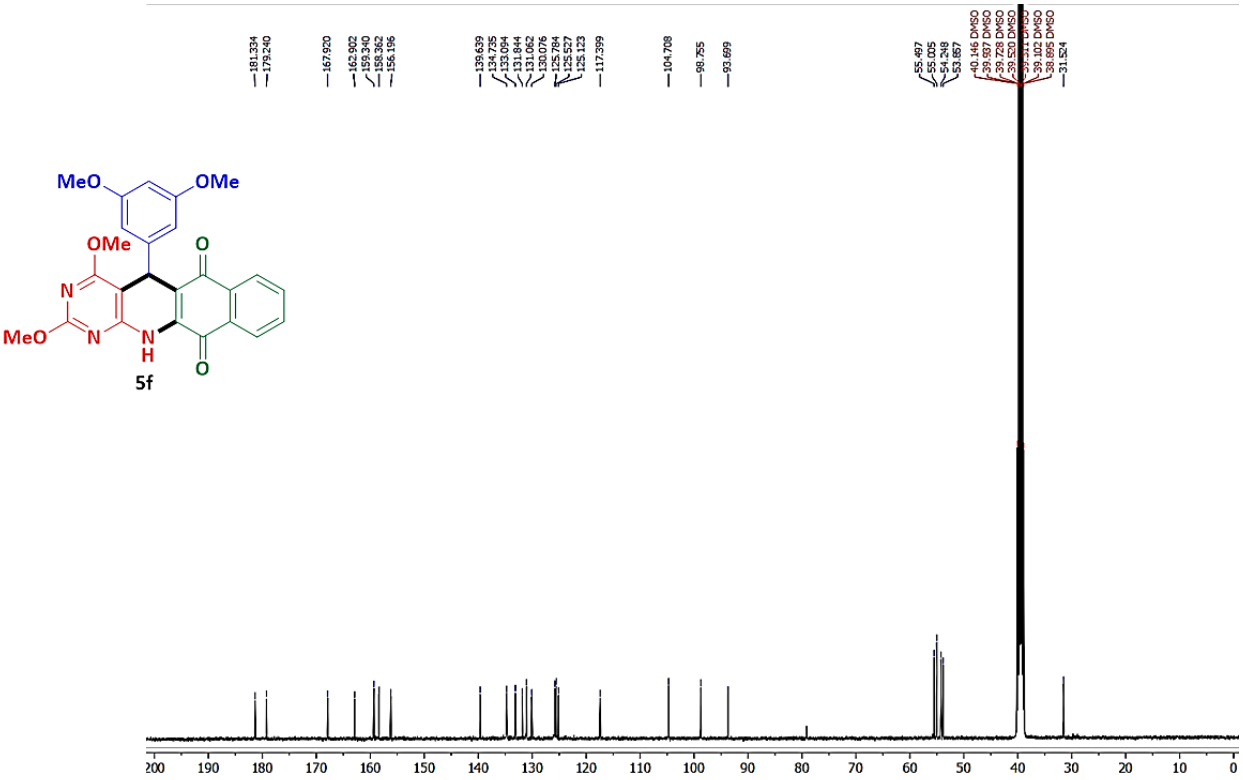


Figure **S33**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6f**.

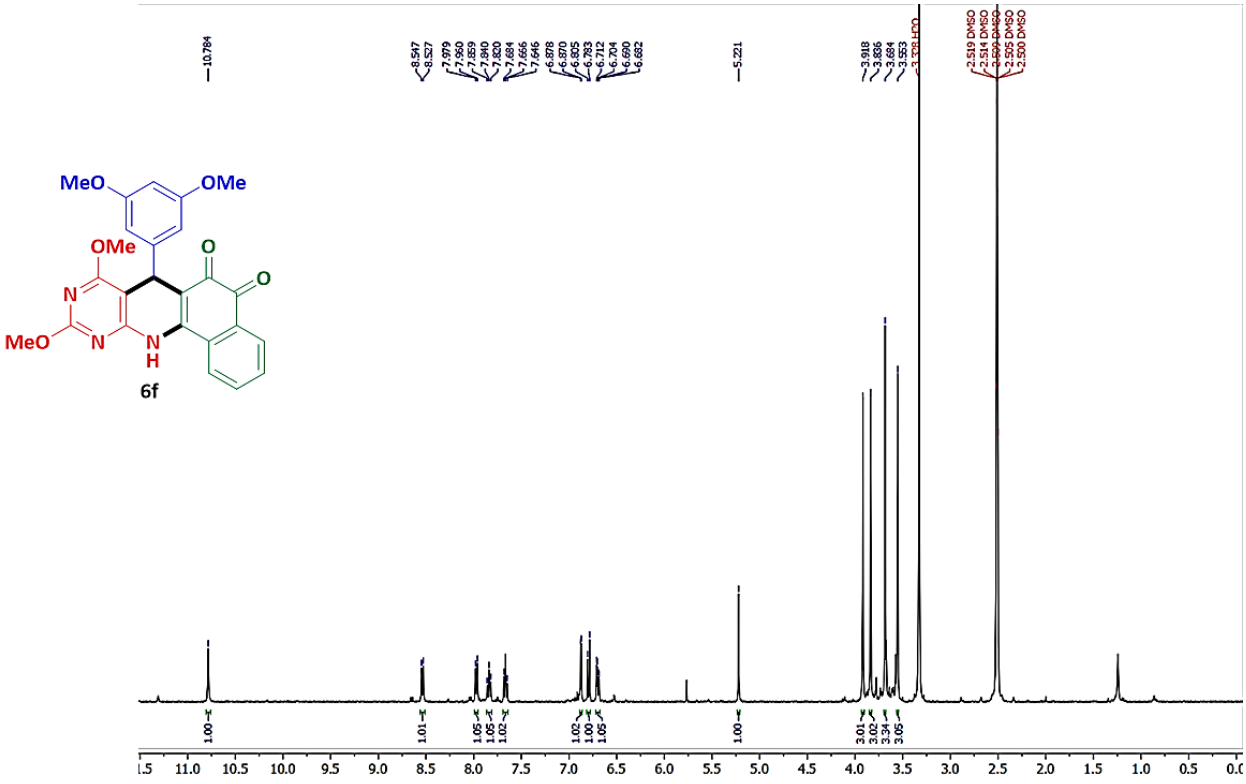


Figure **S34**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6f**.

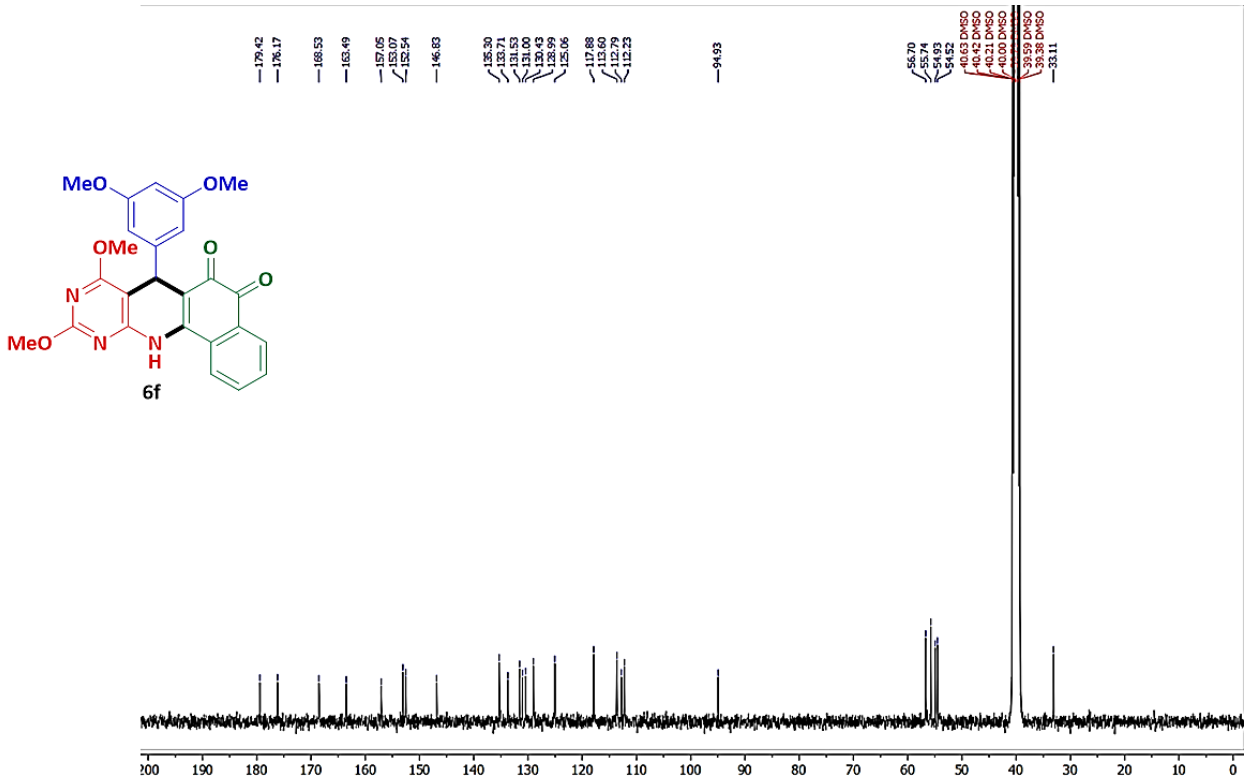


Figure **S35**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5g**.

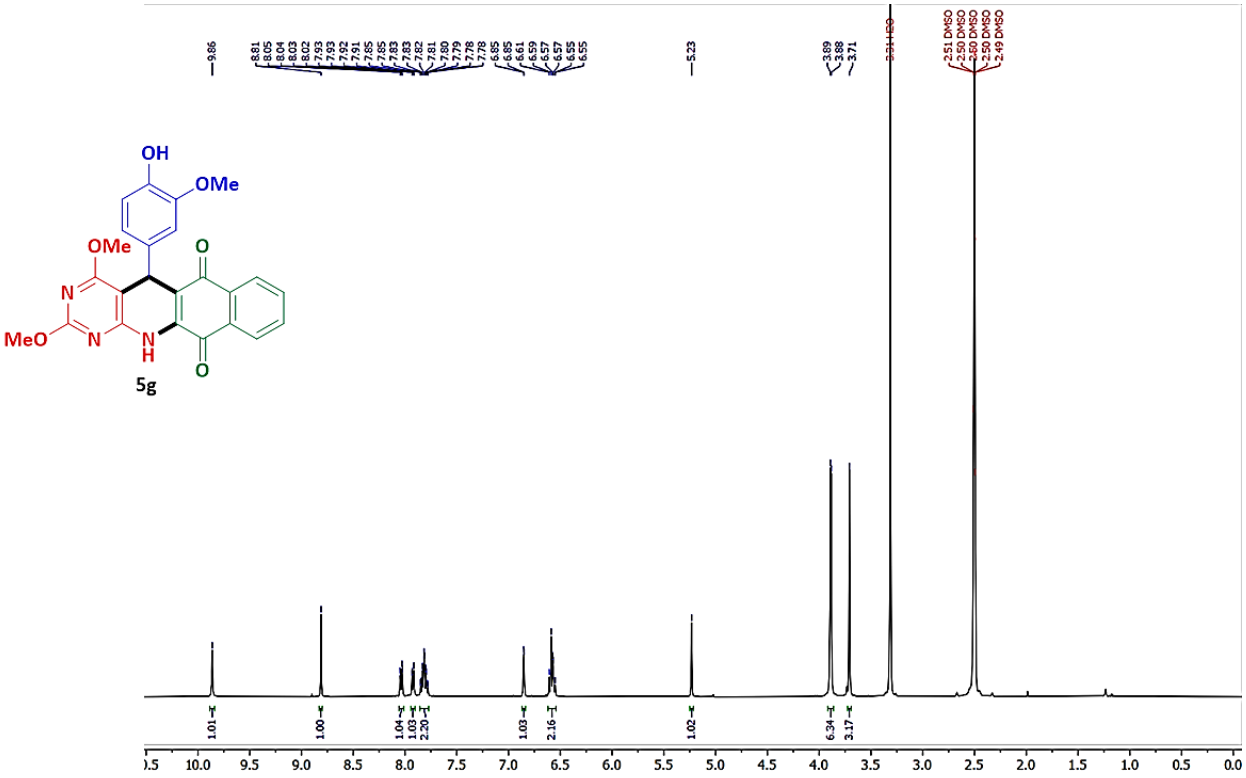


Figure **S36**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5g**.

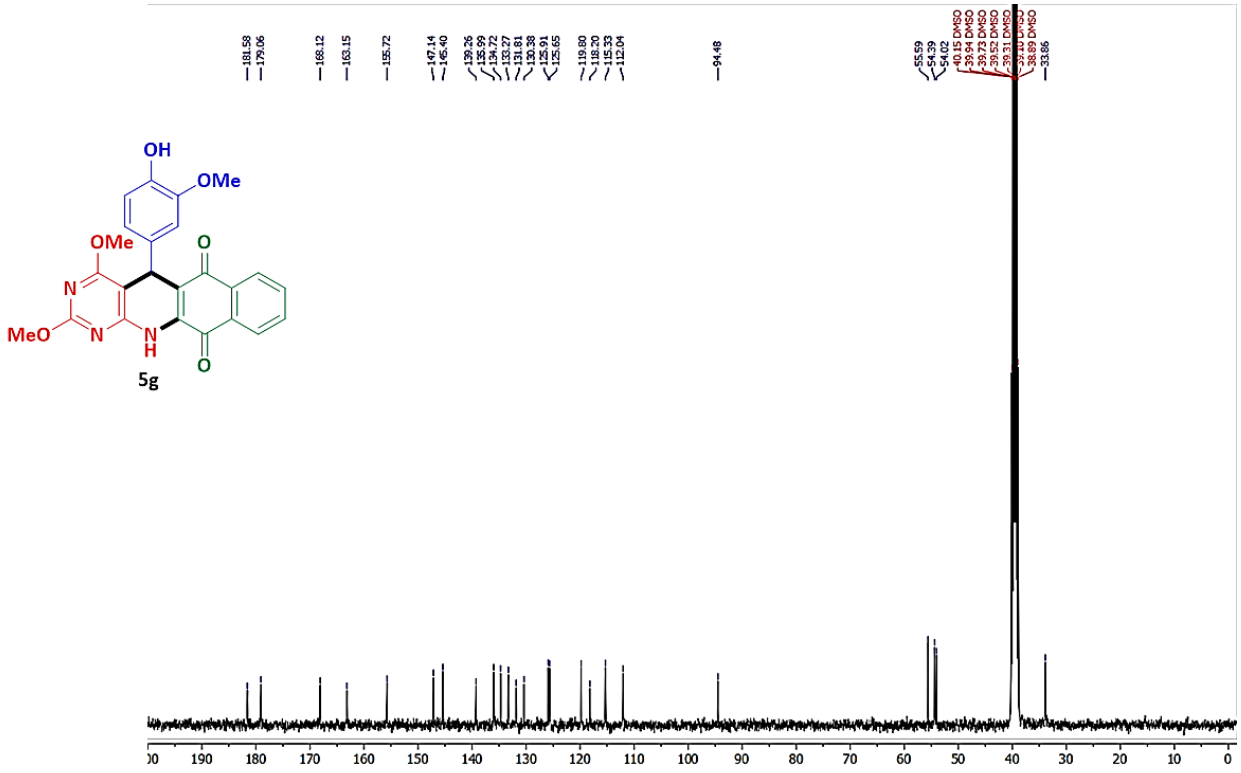


Figure **S37**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6g**.

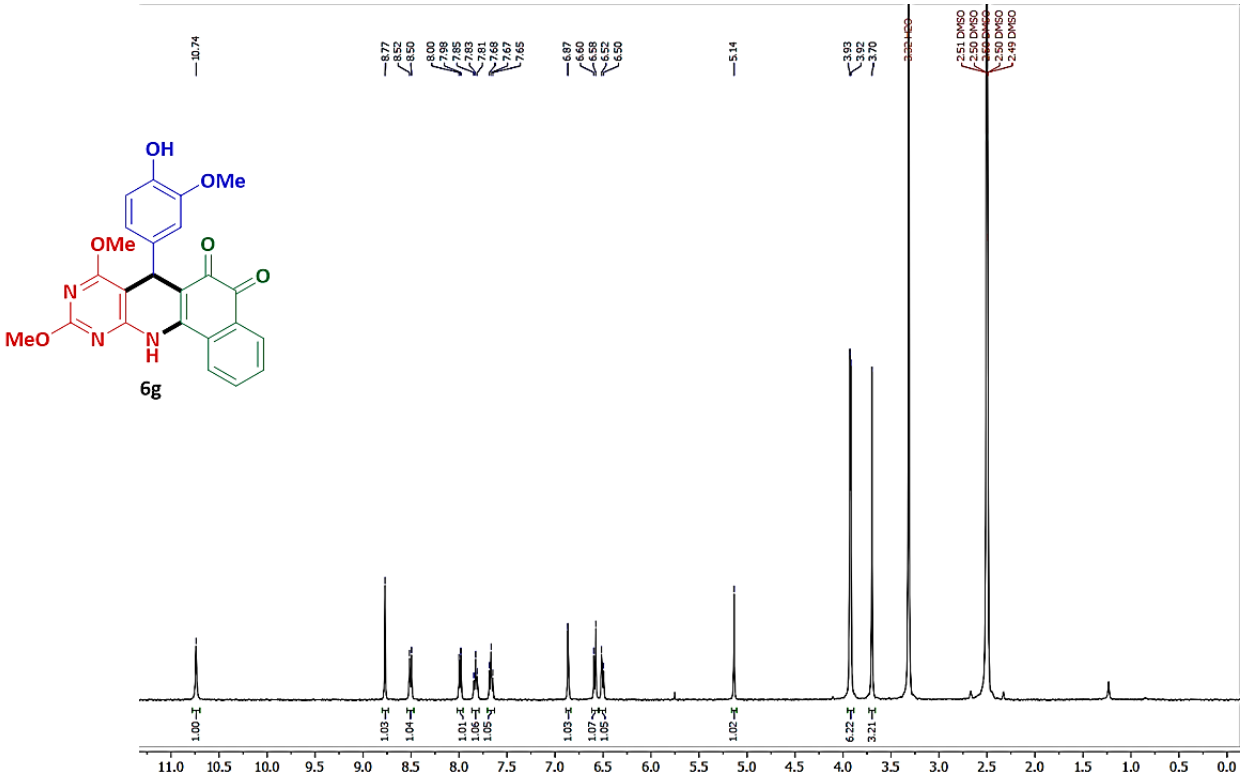


Figure **S38**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6g**.

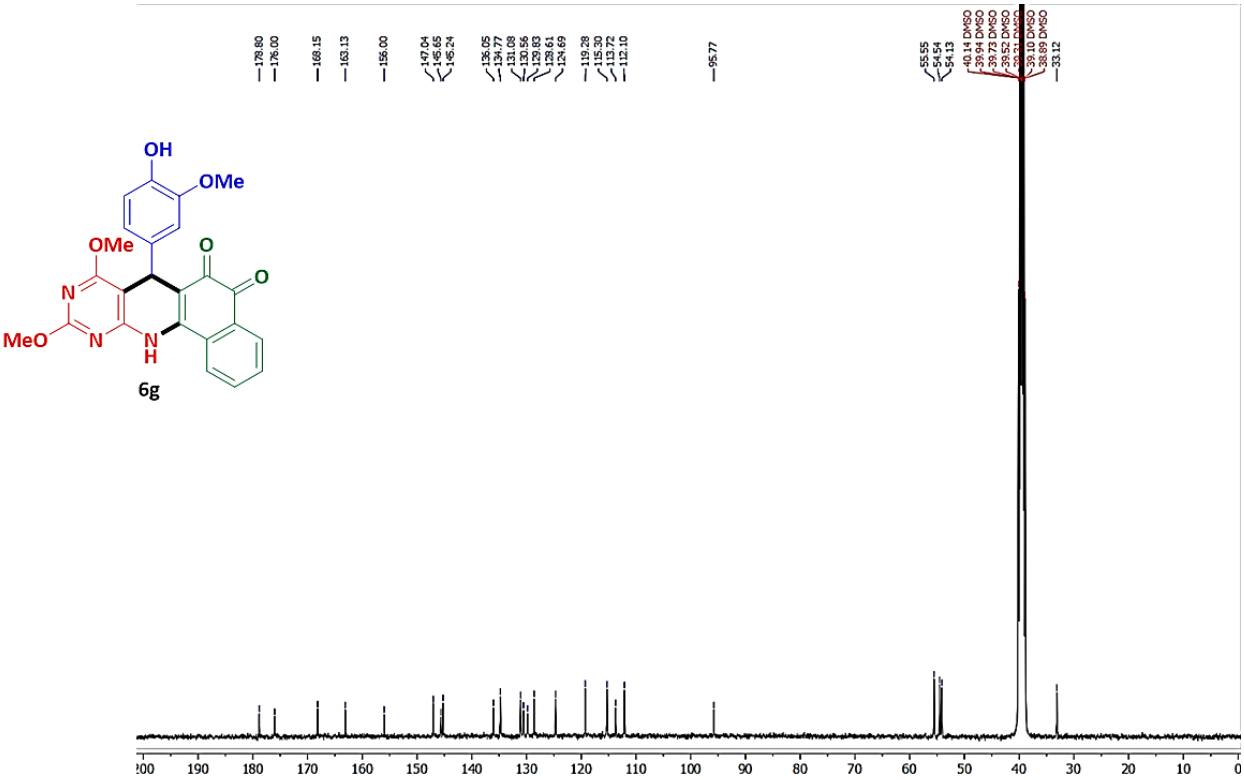


Figure **S39**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5h**.

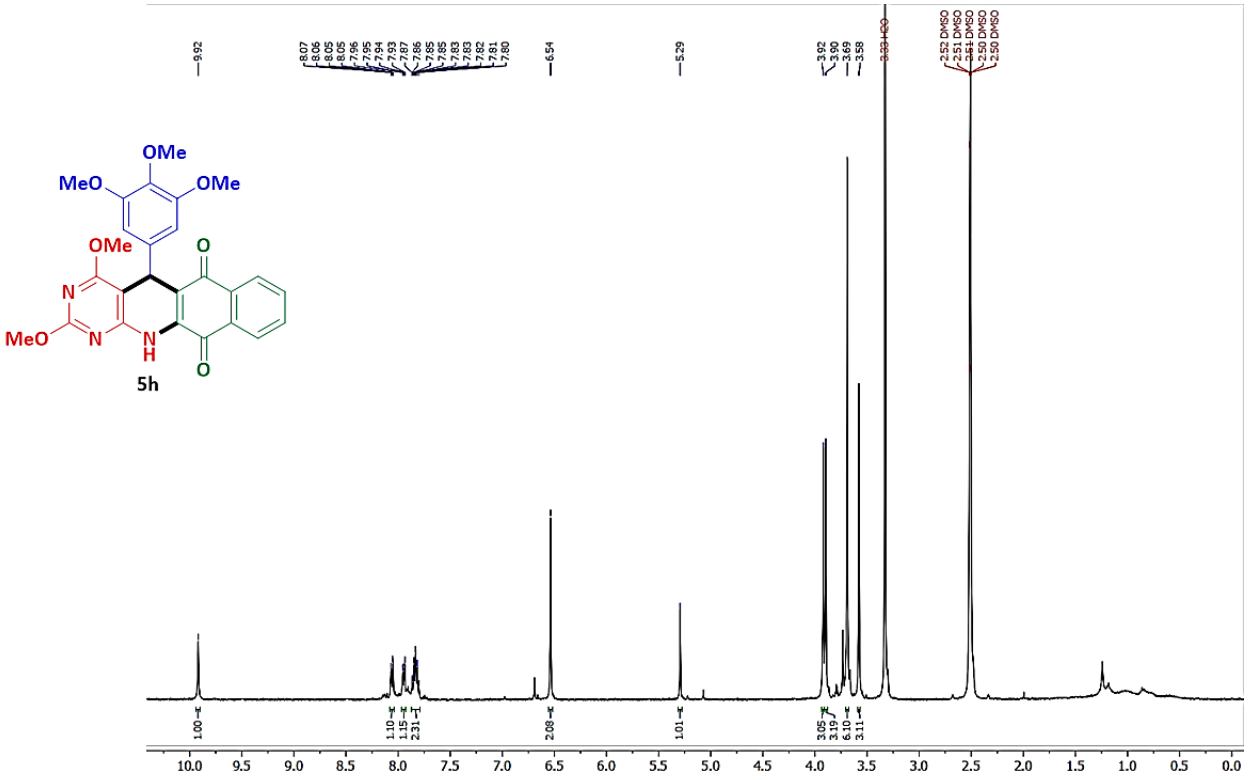


Figure **S40**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5h**.

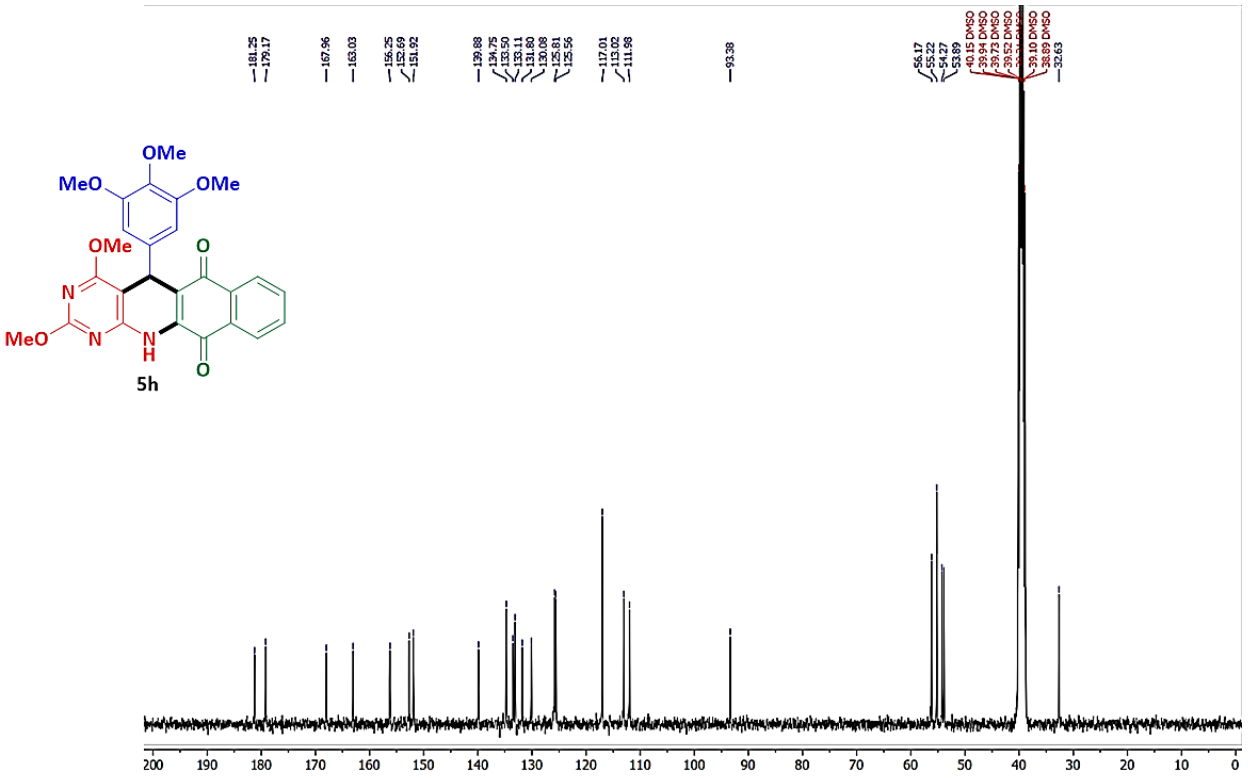


Figure **S41**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6h**.

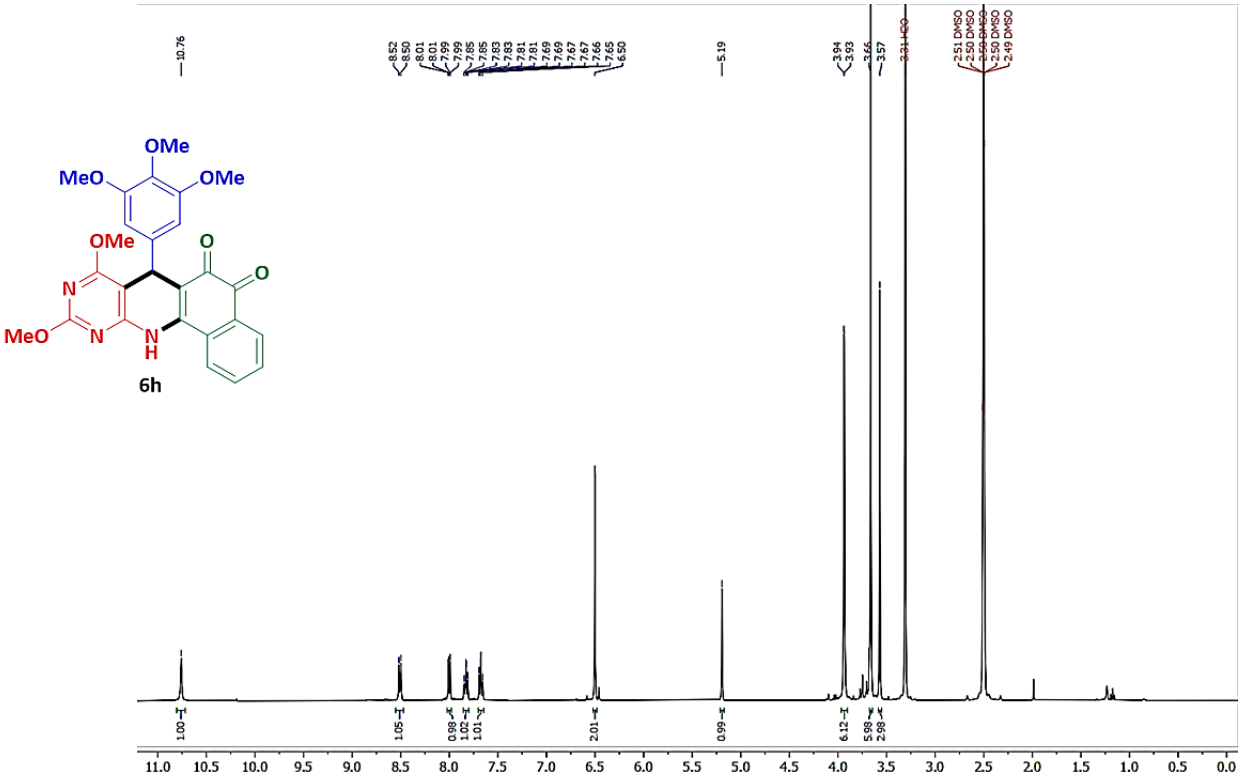


Figure **S42**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6h**.

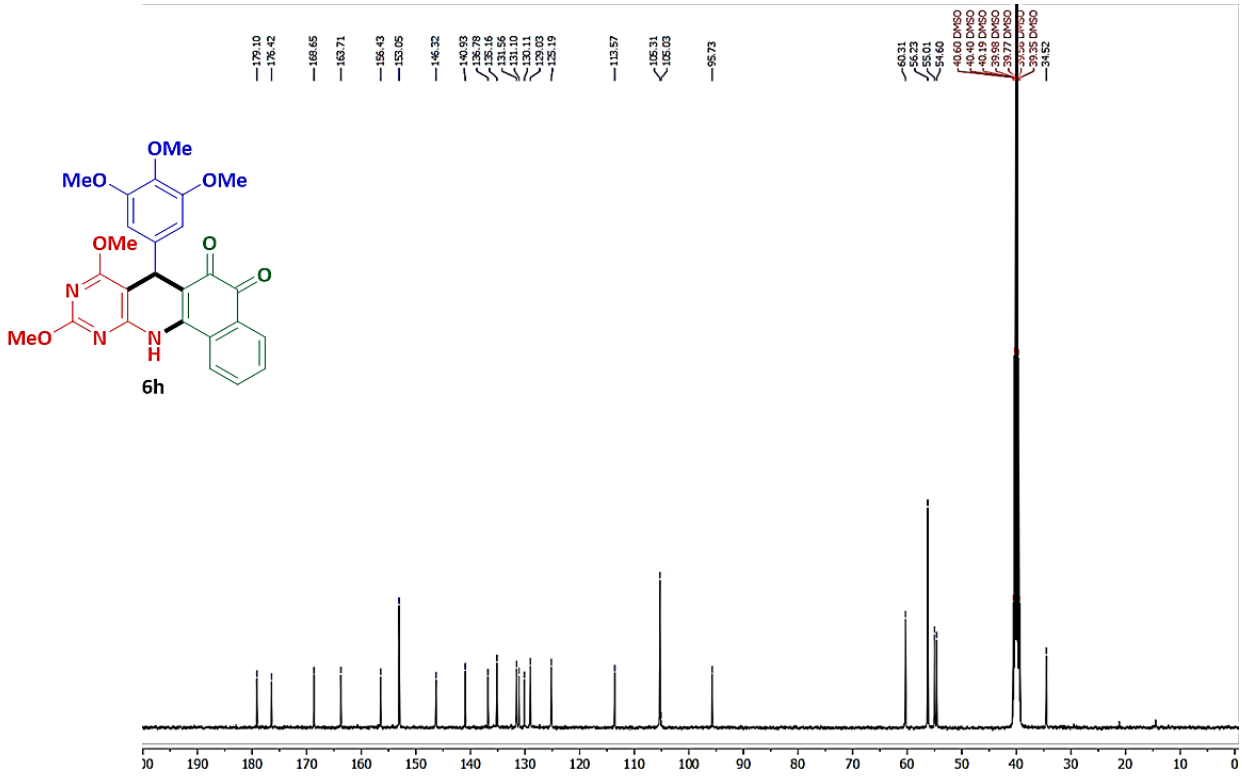


Figure **S43**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5i**.

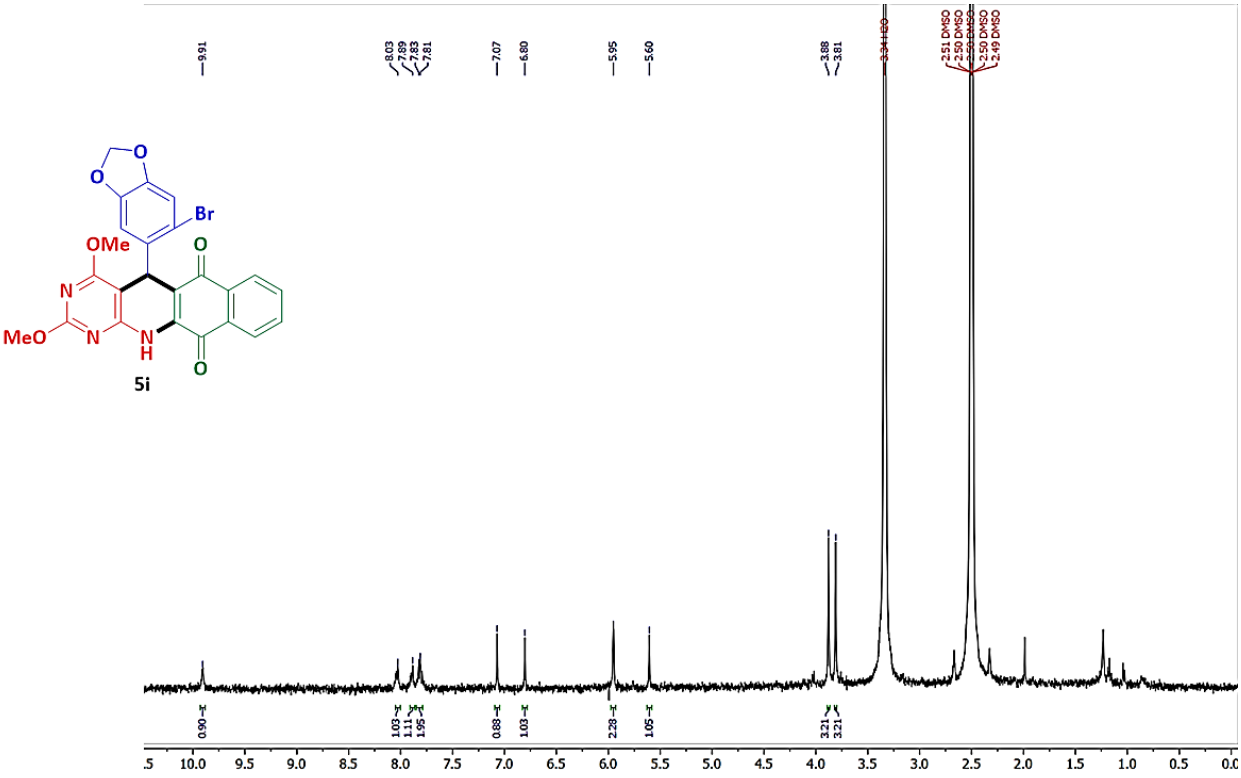


Figure **S44**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5i**.

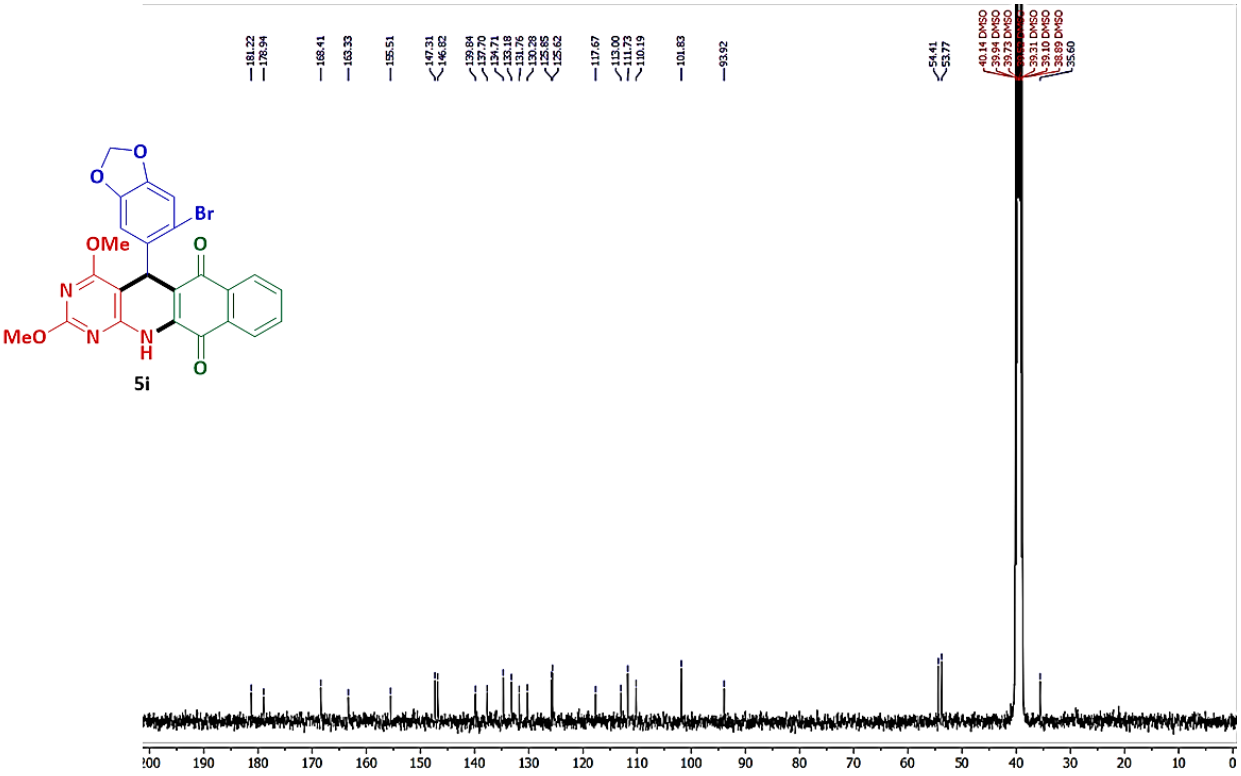


Figure **S45**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6i**.

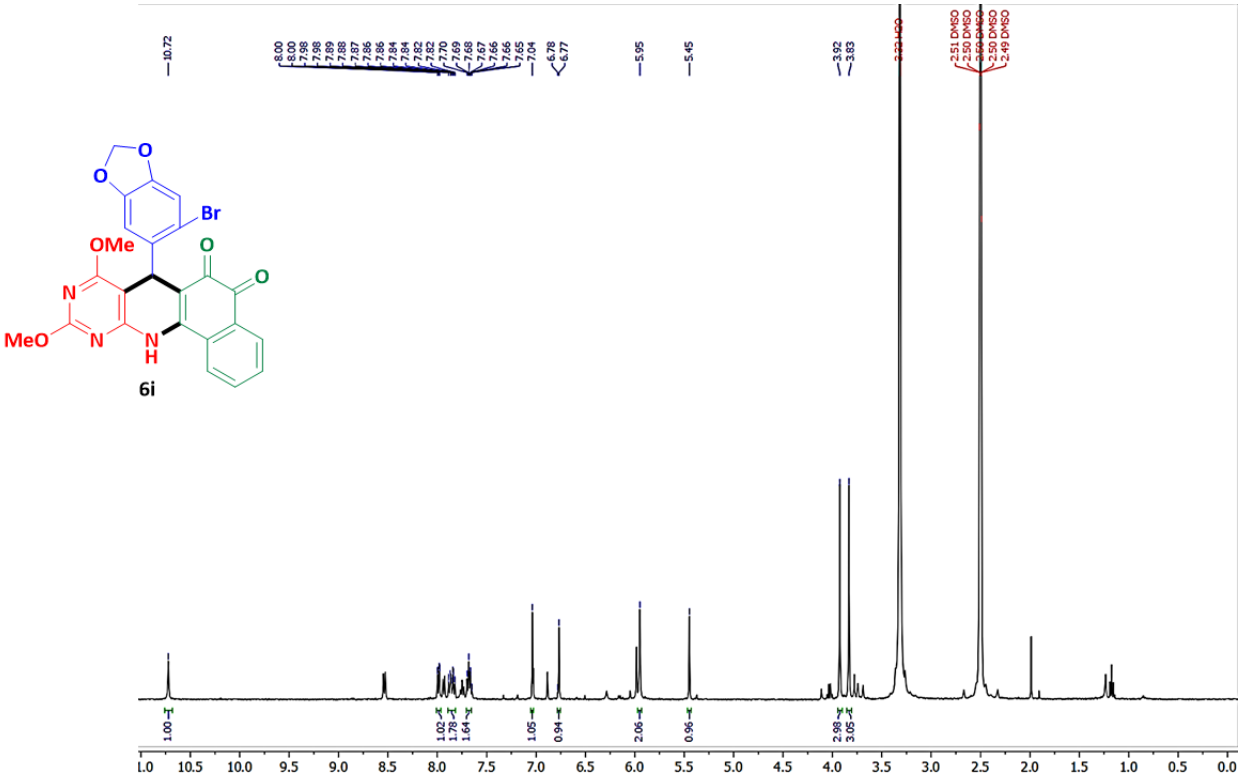


Figure **S46**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6i**.

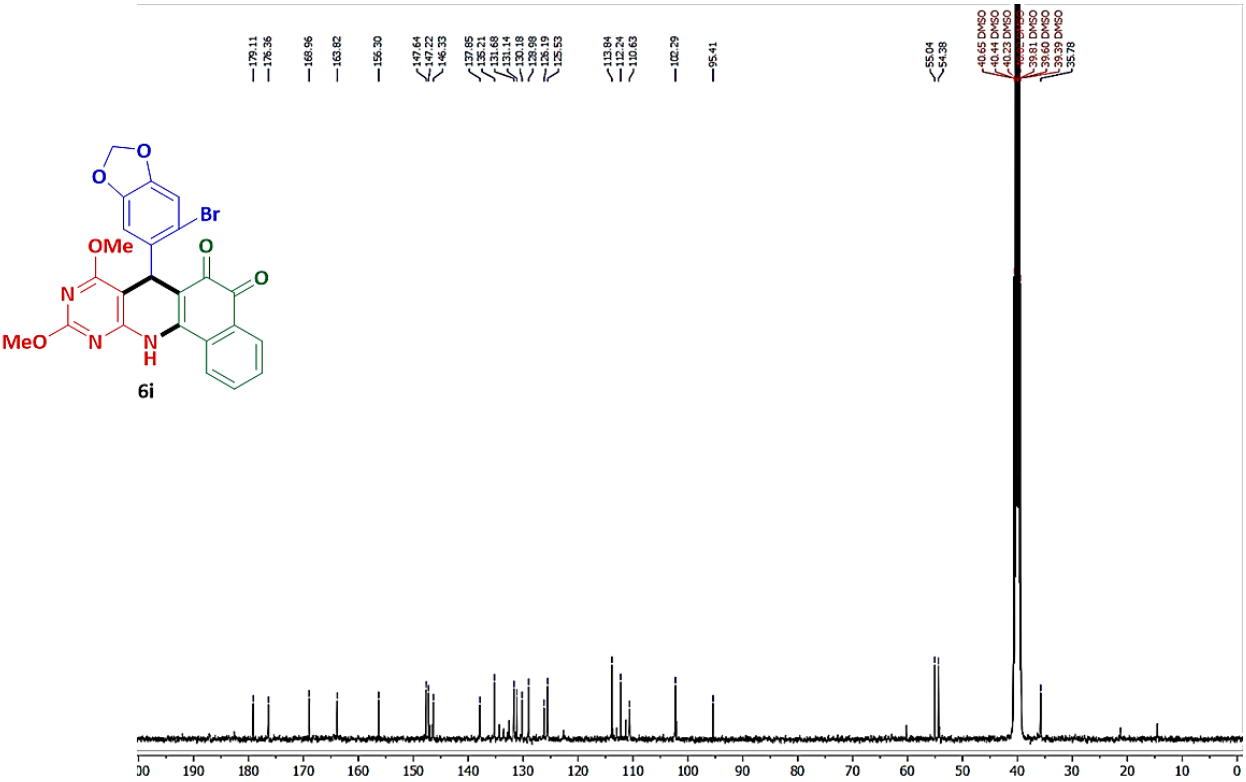


Figure **S47**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **5j**.

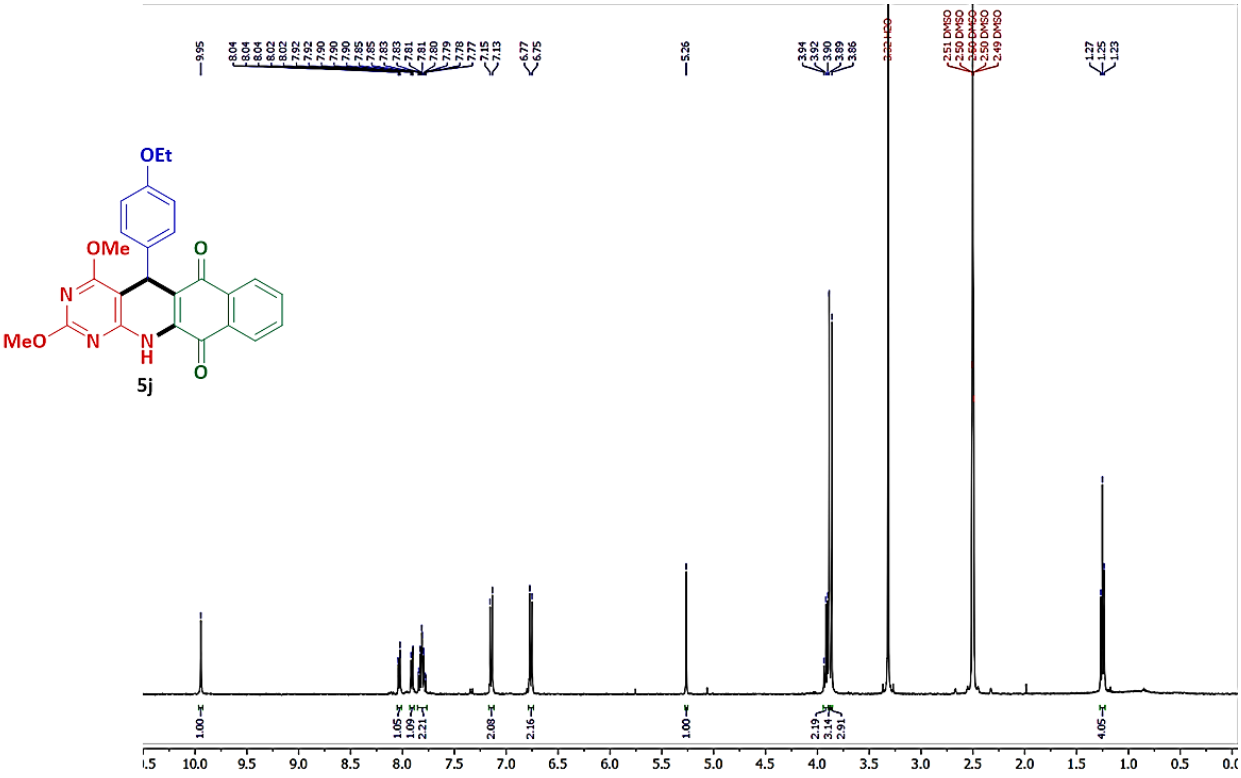


Figure **S48**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **5j**.

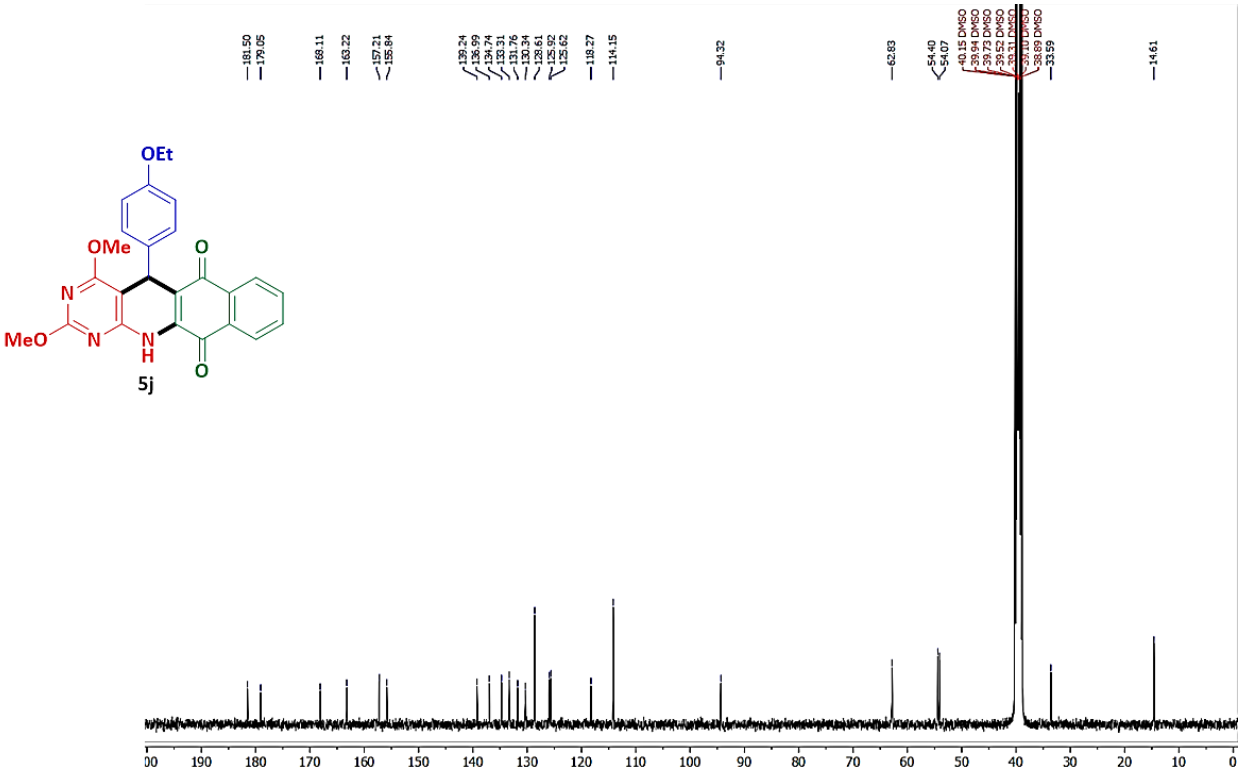


Figure **S49**: ^{1}H-NMR (400 MHz, DMSO-d_6) of Compound **6j**.

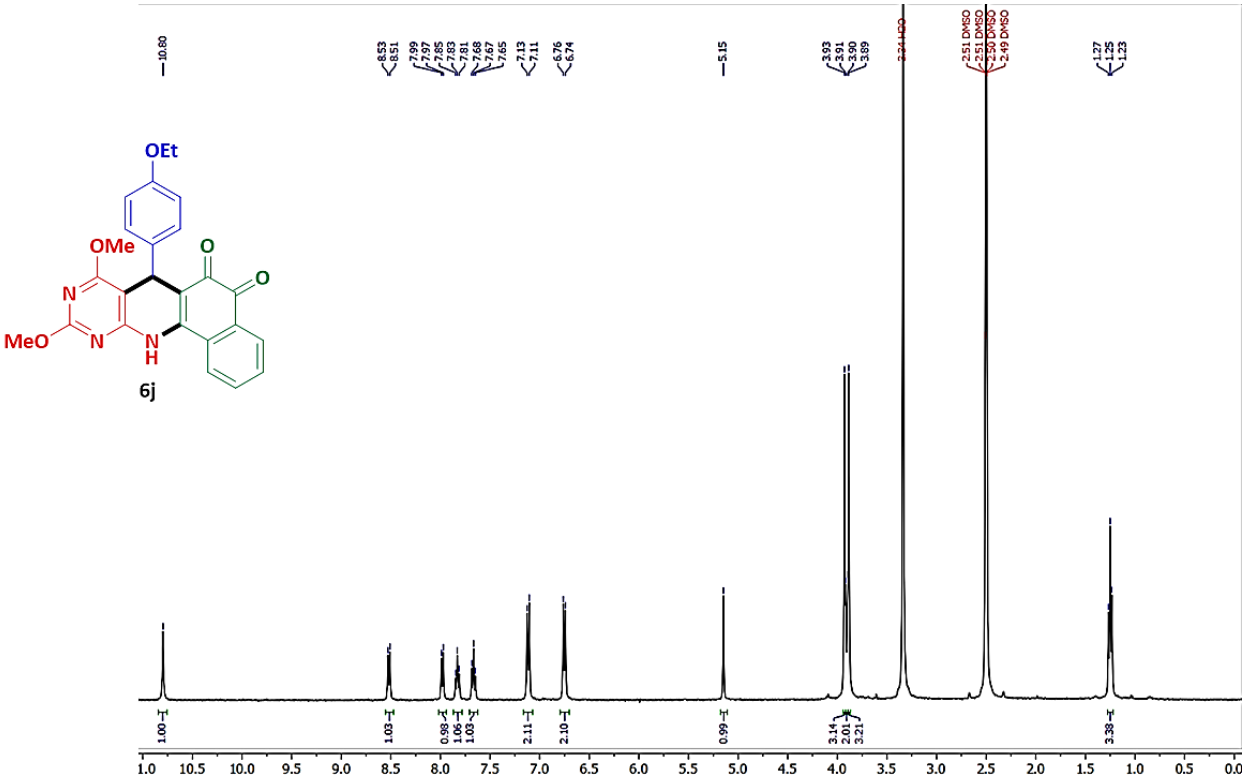


Figure **S50**: ^{13}C-NMR (100 MHz, DMSO-d_6) of Compound **6j**.

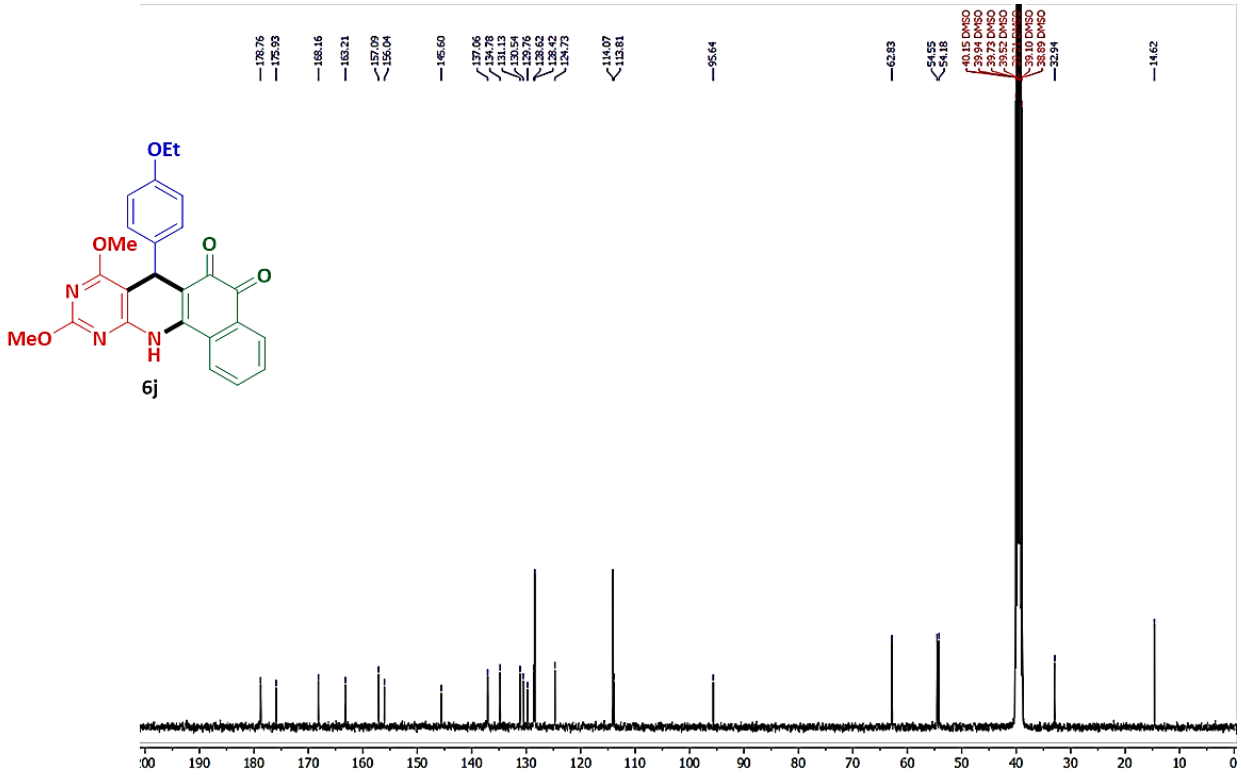
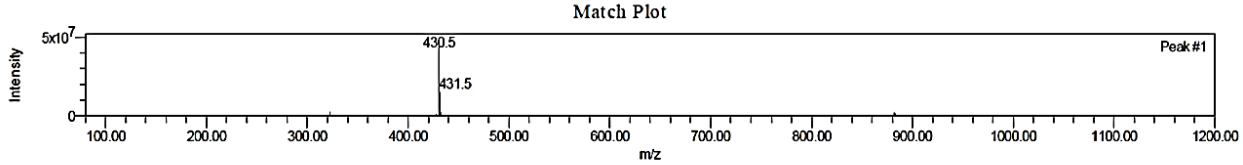


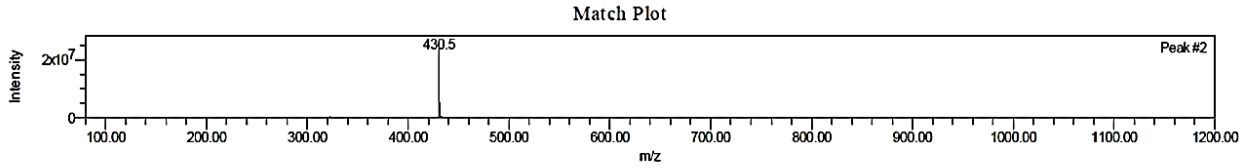
Figure **S51**: LCMS of Compound **5a** and **6a**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.775	430.48	120045799	319391859	70.06	SQ 1: MS Scan	MS TIC
2	2.047	430.47	65459618	136509394	29.94	SQ 1: MS Scan	MS TIC



Base Peak 430.48 Channel Description 1: 80.00-1300.00 ES+, Centroid, CV=30 - AVG (2.3:2.7;0.0:0.9) x 66.000 - AVG (2.3:2.7;0.0:0.9) x 66.000 Retention Time 1.77

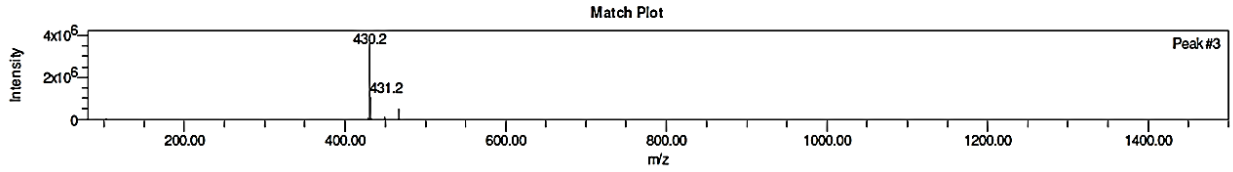


Base Peak 430.47 Channel Description 1: 80.00-1300.00 ES+, Centroid, CV=30 - AVG (2.3:2.7;0.0:0.9) x 66.000 - AVG (2.3:2.7;0.0:0.9) x 66.000 Retention Time 2.04

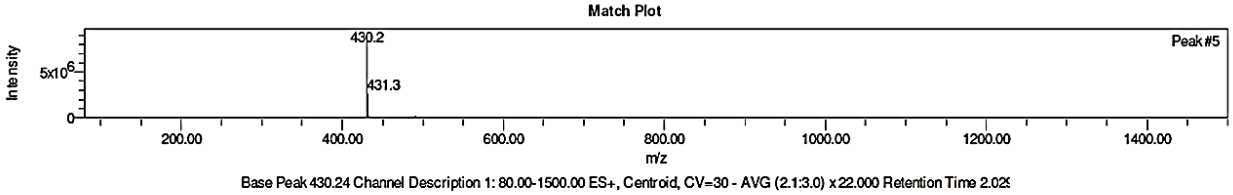
Figure **S52**: LCMS of Compound **5b** and **6b**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.561	102.19	172175	334436	0.35	SQ 1: MS Scan	MS TIC
2	1.637	306.35	205584	427779	0.45	SQ 1: MS Scan	MS TIC
3	1.765	430.20	15259236	22292748	23.44	SQ 1: MS Scan	MS TIC
4	1.778	430.20	9518544	7104535	7.47	SQ 1: MS Scan	MS TIC
5	2.029	430.24	35556402	64947101	68.29	SQ 1: MS Scan	MS TIC



Base Peak 430.20 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (2.1:3.0) x 22.000 Retention Time 1.76

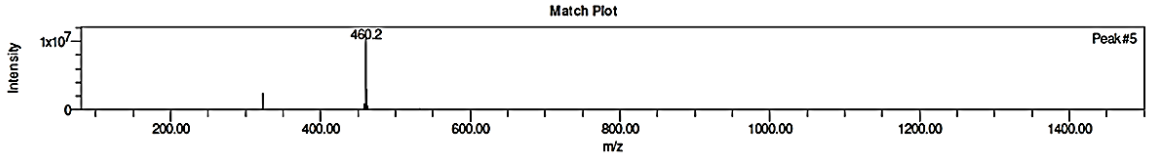


Base Peak 430.24 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (2.1:3.0) x 22.000 Retention Time 2.02

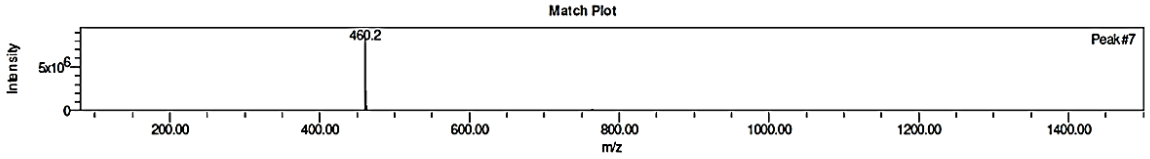
Figure **S53**: LCMS of Compound **5c** and **6c**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.480	308.60	47210	181248	0.10	SQ 1: MS Scan	MS TIC
2	1.568	430.29	362232	817619	0.47	SQ 1: MS Scan	MS TIC
3	1.603	430.38	166971	199488	0.11	SQ 1: MS Scan	MS TIC
4	1.667	477.42	753061	941422	0.54	SQ 1: MS Scan	MS TIC
5	1.746	460.25	47271310	98149562	56.56	SQ 1: MS Scan	MS TIC
6	1.842	446.24	5625311	11315750	6.52	SQ 1: MS Scan	MS TIC
7	2.011	460.24	37731053	61920977	35.68	SQ 1: MS Scan	MS TIC



Base Peak 460.25 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (2.1:3.0) x 22.000 Retention Time 1.74

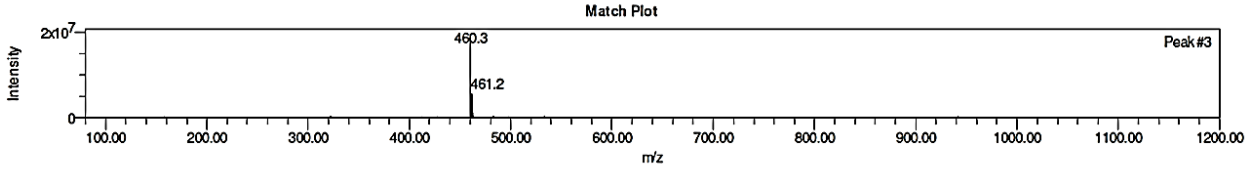


Base Peak 460.24 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (2.1:3.0) x 22.000 Retention Time 2.01

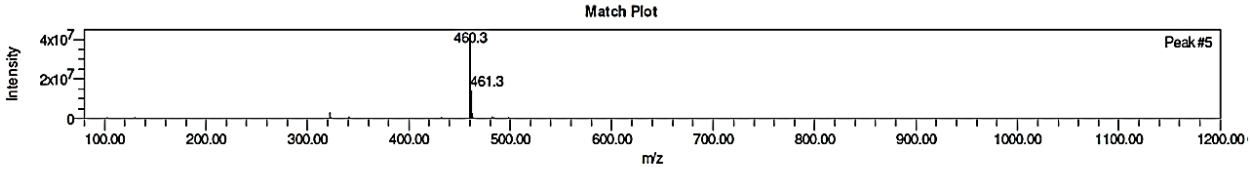
Figure **S54**: LCMS of Compound **5d** and **6d**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.530	568.52	4665761	8554236	1.35	SQ 1: MS Scan	MS TIC
2	1.598	427.29	26013484	53791895	8.48	SQ 1: MS Scan	MS TIC
3	1.677	460.29	74401742	175796194	27.71	SQ 1: MS Scan	MS TIC
4	1.739	446.23	12687620	45825705	7.22	SQ 1: MS Scan	MS TIC
5	1.902	460.29	113603228	350435664	55.24	SQ 1: MS Scan	MS TIC



Base Peak 460.29 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 Retention Time 1.677

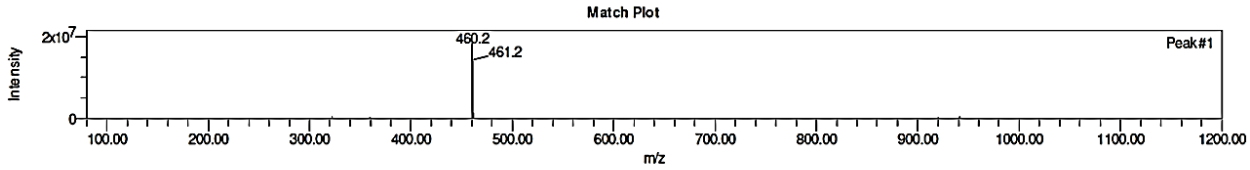


Base Peak 460.29 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 Retention Time 1.902

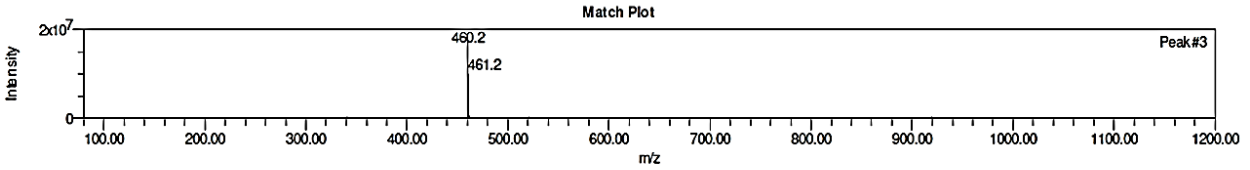
Figure **S55**: LCMS of Compound **5e** and **6e**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.605	460.24	64449269	194362951	53.19	SQ 1: MS Scan	MS TIC
2	1.703	306.22	3966713	11321953	3.10	SQ 1: MS Scan	MS TIC
3	1.798	460.24	49453877	159751545	43.72	SQ 1: MS Scan	MS TIC



Base Peak 460.24 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (1.9:3.0;0.0:1.5) x 66.000 Retention Time 1.60

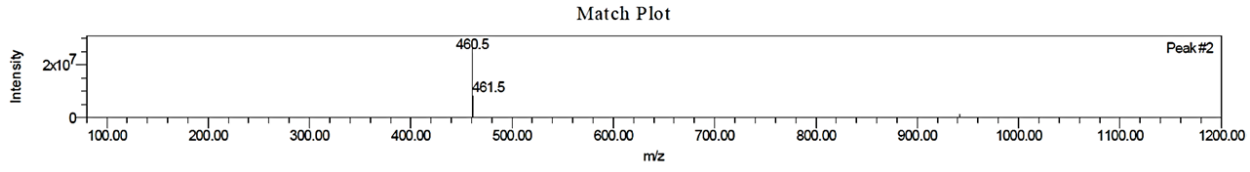


Base Peak 460.24 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (1.9:3.0;0.0:1.5) x 66.000 Retention Time 1.79

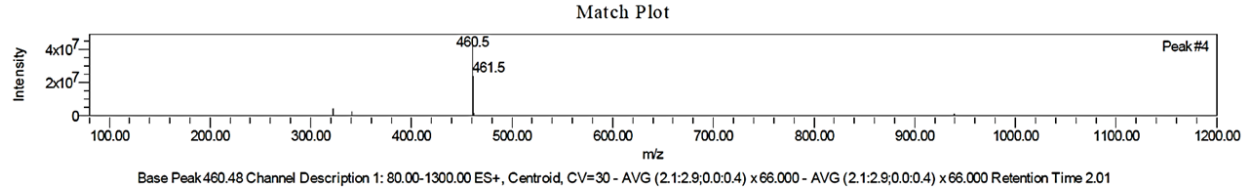
Figure **S56**: LCMS of Compound **5f** and **6f**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.619	427.50	7443571	11307606	2.10	SQ 1: MS Scan	MS TIC
2	1.746	460.48	79594254	172800007	32.12	SQ 1: MS Scan	MS TIC
3	1.819	446.44	3120204	6782431	1.26	SQ 1: MS Scan	MS TIC
4	2.019	460.48	125194840	347142214	64.52	SQ 1: MS Scan	MS TIC



Base Peak 460.48 Channel Description 1: 80.00-1300.00 ES+, Centroid, CV=30 - AVG (2.1:2.9;0.0:0.4) x 66.000 - AVG (2.1:2.9;0.0:0.4) x 66.000 Retention Time 1.74

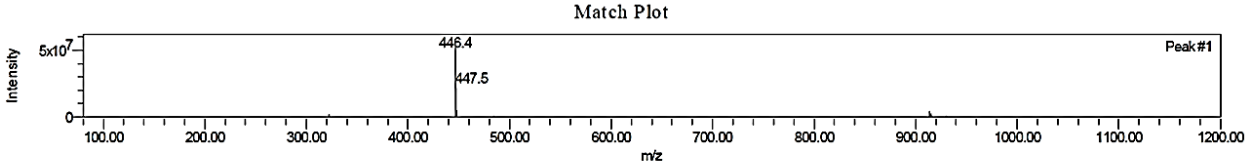


Base Peak 460.48 Channel Description 1: 80.00-1300.00 ES+, Centroid, CV=30 - AVG (2.1:2.9;0.0:0.4) x 66.000 - AVG (2.1:2.9;0.0:0.4) x 66.000 Retention Time 2.01

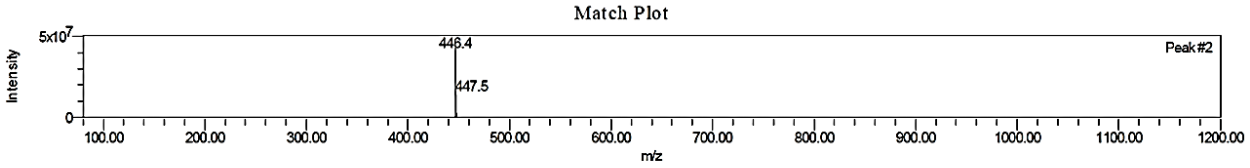
Figure **S57**: LCMS of Compound **5g** and **6g**.

Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.623	446.44	144367079	432452286	60.19	SQ 1: MS Scan	MS TIC
2	1.824	446.44	114637787	286017972	39.81	SQ 1: MS Scan	MS TIC



Base Peak 446.44 Channel Description 1: 80.00-1300.00 ES+, Centroid, CV=30 - AVG (2.0:2.6;0.0:0.5) x 55.000 - AVG (2.0:2.6;0.0:0.5) x 55.000 Retention Time 1.62

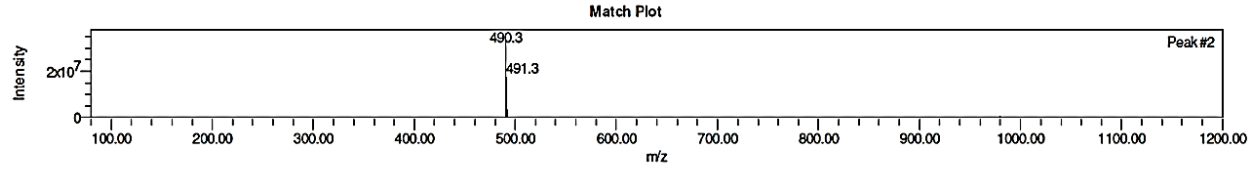


Base Peak 446.44 Channel Description 1: 80.00-1300.00 ES+, Centroid, CV=30 - AVG (2.0:2.6;0.0:0.5) x 55.000 - AVG (2.0:2.6;0.0:0.5) x 55.000 Retention Time 1.82

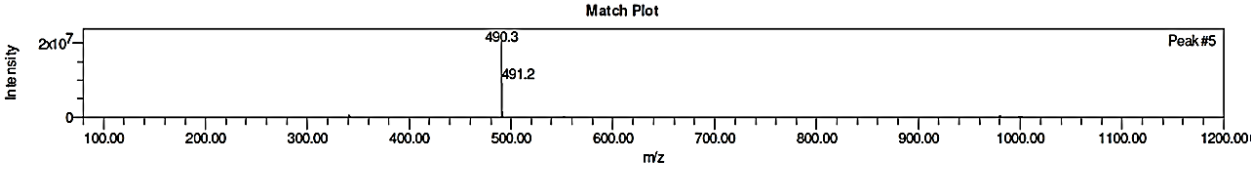
Figure **S58**: LCMS of Compound **5h** and **6h**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.591	506.29	2546194	5311093	0.95	SQ 1: MS Scan	MS TIC
2	1.666	490.28	94390202	288480013	51.55	SQ 1: MS Scan	MS TIC
3	1.749	306.24	3572833	6101509	1.09	SQ 1: MS Scan	MS TIC
4	1.805	506.26	8962906	19785782	3.54	SQ 1: MS Scan	MS TIC
5	1.881	490.28	85477559	239957867	42.88	SQ 1: MS Scan	MS TIC



Base Peak 490.28 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (2.1:2.9;0.0:1.4) x 66.000 Retention Time 1.66



Base Peak 490.28 Channel Description 1: 80.00-1500.00 ES+, Centroid, CV=30 - AVG (2.1:2.9;0.0:1.4) x 66.000 Retention Time 1.88

Figure **S59**: LCMS of Compound **5i** and **6i**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.249	155.95	553771	1078715	0.35	SQ 1: MS Scan	MS TIC
2	1.418	197.90	468304	1034610	0.33	SQ 1: MS Scan	MS TIC
3	1.613	506.22	737945	1693407	0.54	SQ 1: MS Scan	MS TIC
4	1.679	521.19	14327577	56034450	18.01	SQ 1: MS Scan	MS TIC
5	1.787	522.16	42511360	123620331	39.74	SQ 1: MS Scan	MS TIC
6	1.923	101.67	1010807	7185034	2.31	SQ 1: MS Scan	MS TIC
7	2.055	522.18	45597397	120431084	38.71	SQ 1: MS Scan	MS TIC

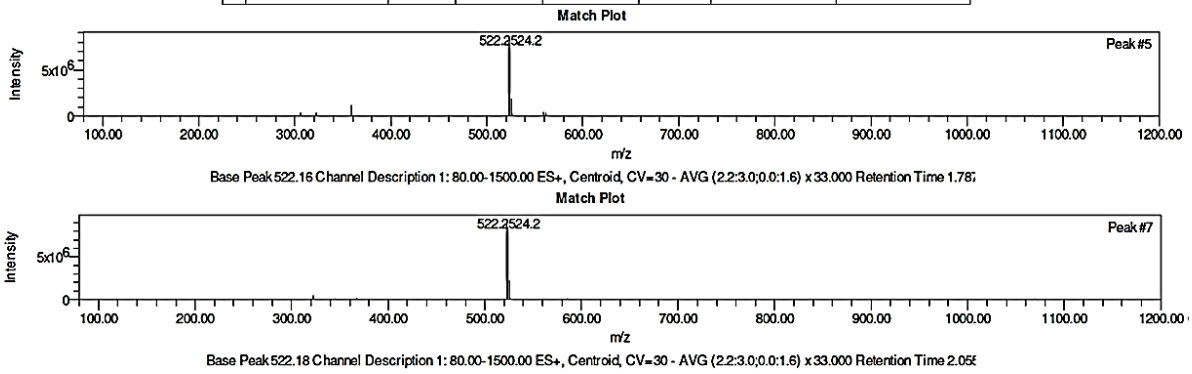


Figure **S60**: LCMS of Compound **5j** and **6j**.

Peak Results
Channel: SQ 1: MS Scan

	Retention Time (min)	Base Peak (m/z)	Height (µV)	Area (µV*sec)	% Area	Channel	Channel Name
1	1.807	444.24	87805798	275187089	73.85	SQ 1: MS Scan	MS TIC
2	2.080	444.27	34512088	97462691	26.15	SQ 1: MS Scan	MS TIC

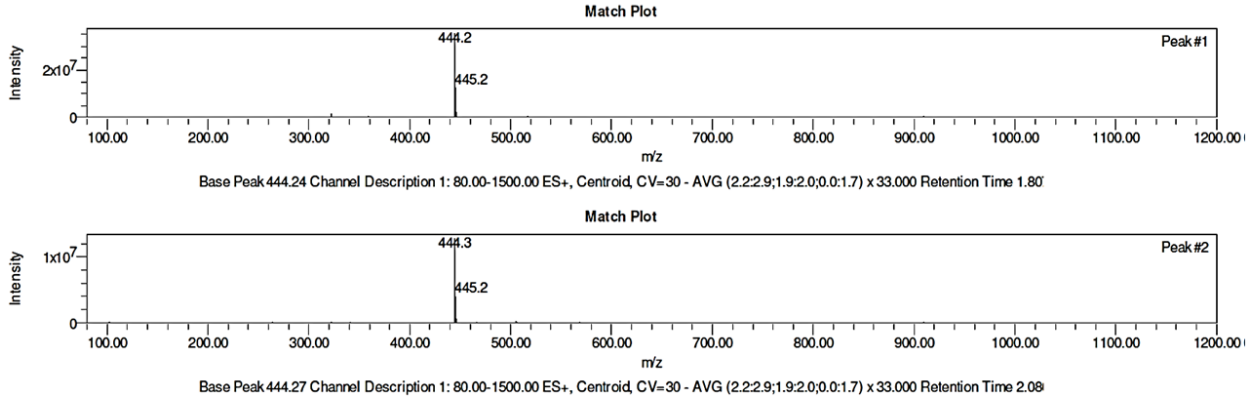


Figure **S61**: HRMS of Compound **5a**.

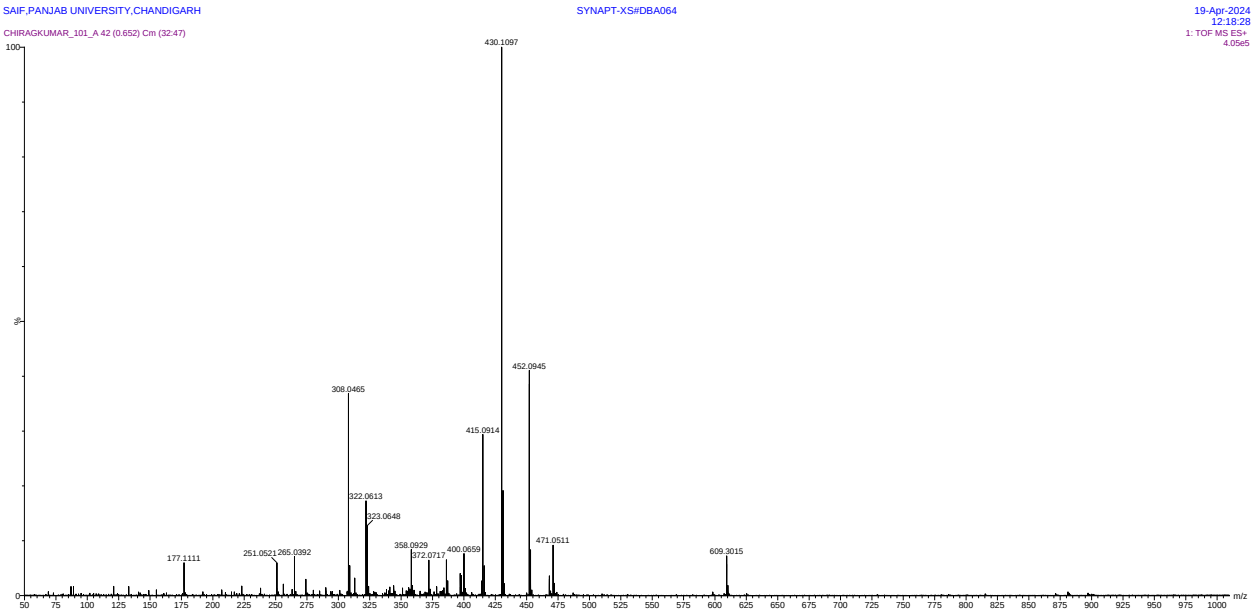


Figure **S62**: HRMS of Compound **6a**.

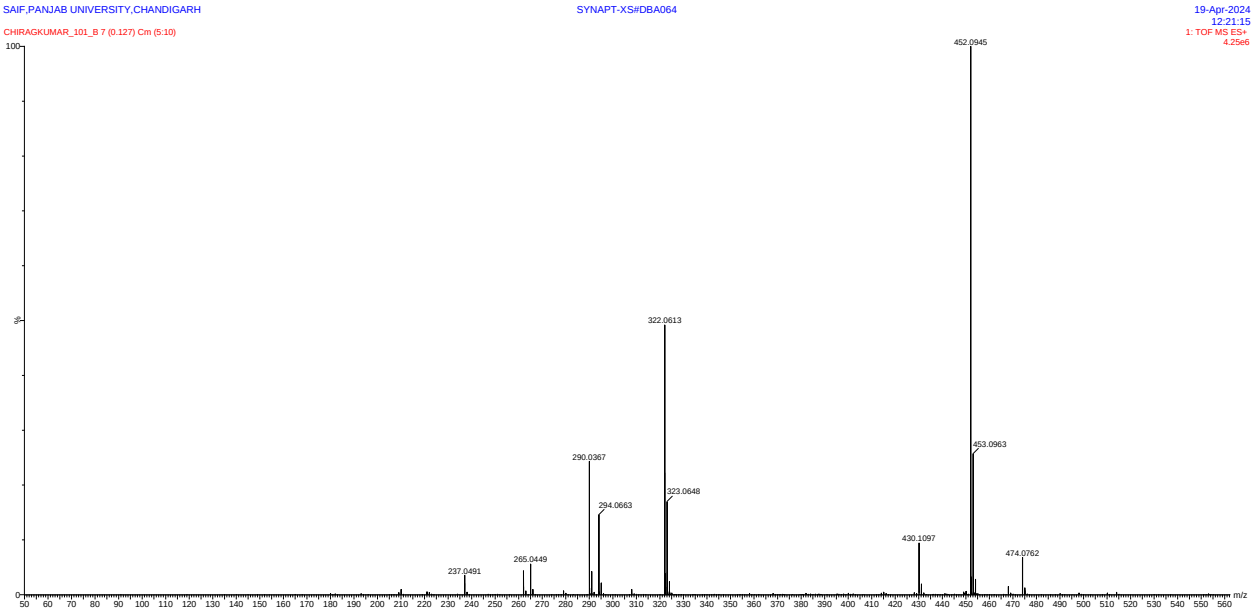


Figure **S63**: HRMS of Compound **5b**.

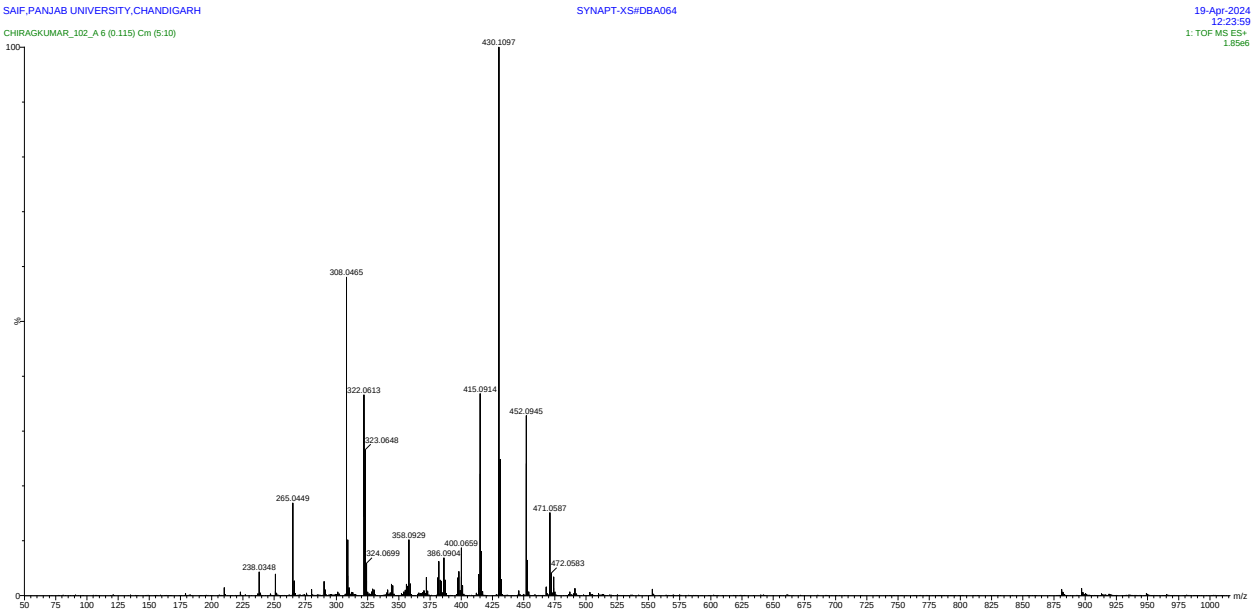


Figure **S64**: HRMS of Compound **6b**.

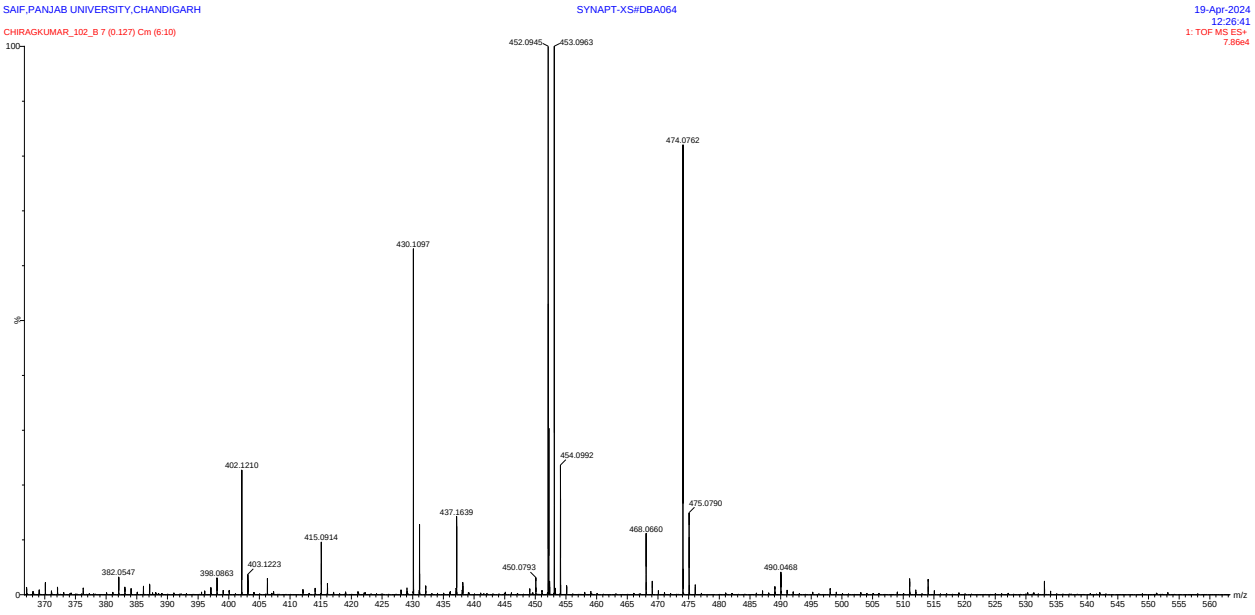


Figure **S65**: HRMS of Compound **5c**.

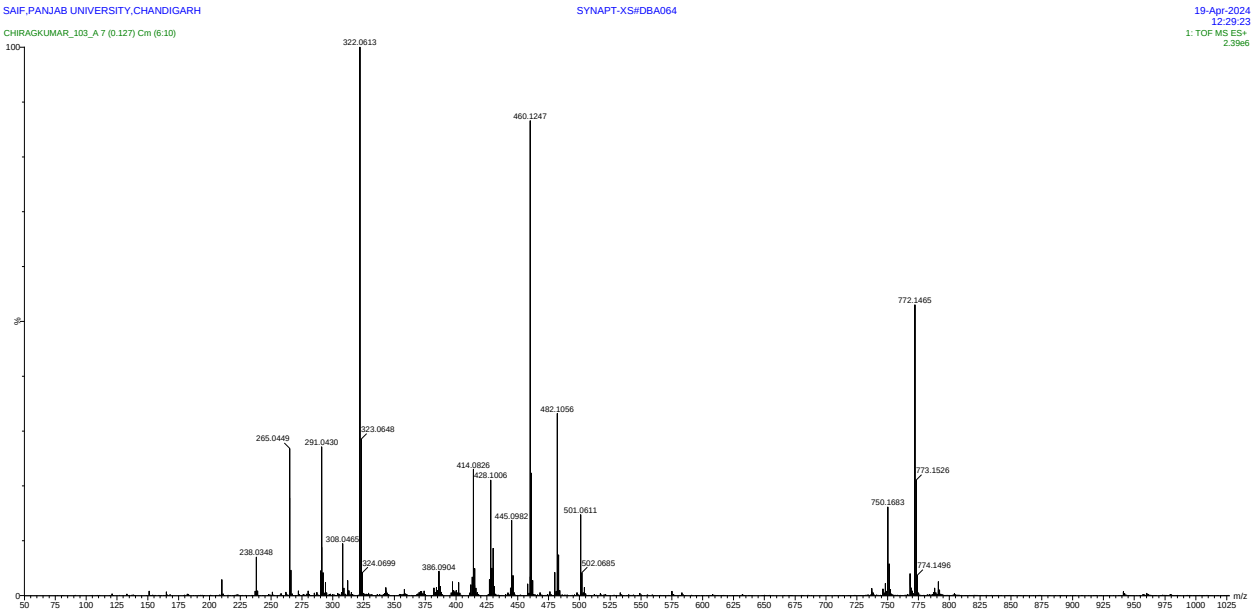


Figure **S66**: HRMS of Compound **6c**.

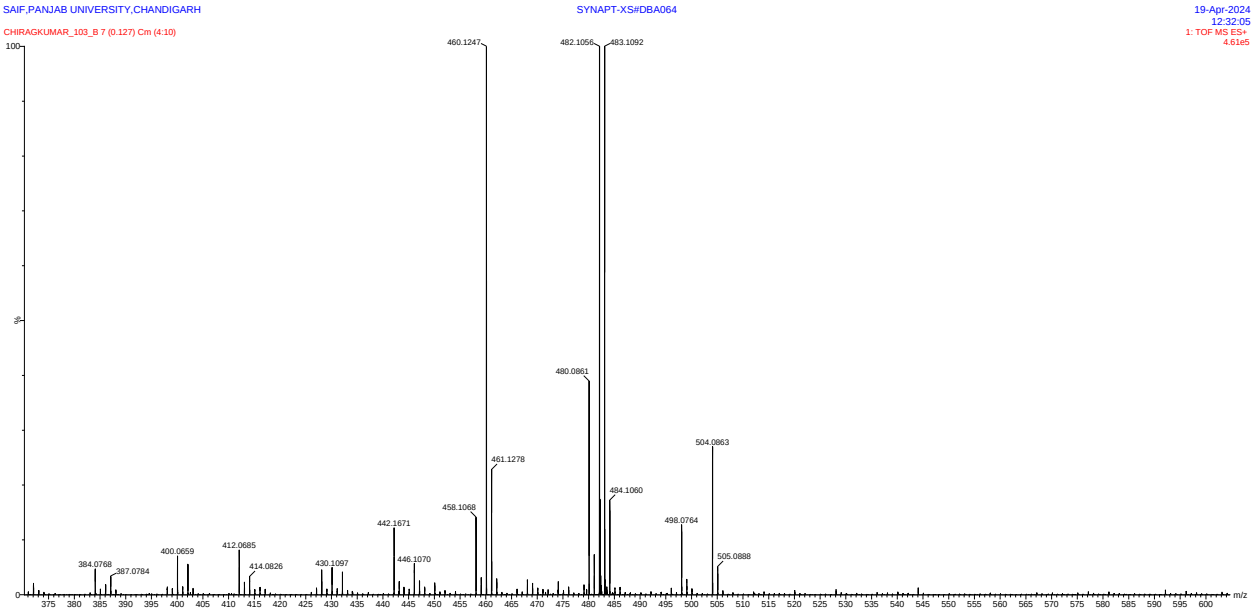


Figure **S67**: HRMS of Compound **5d**.

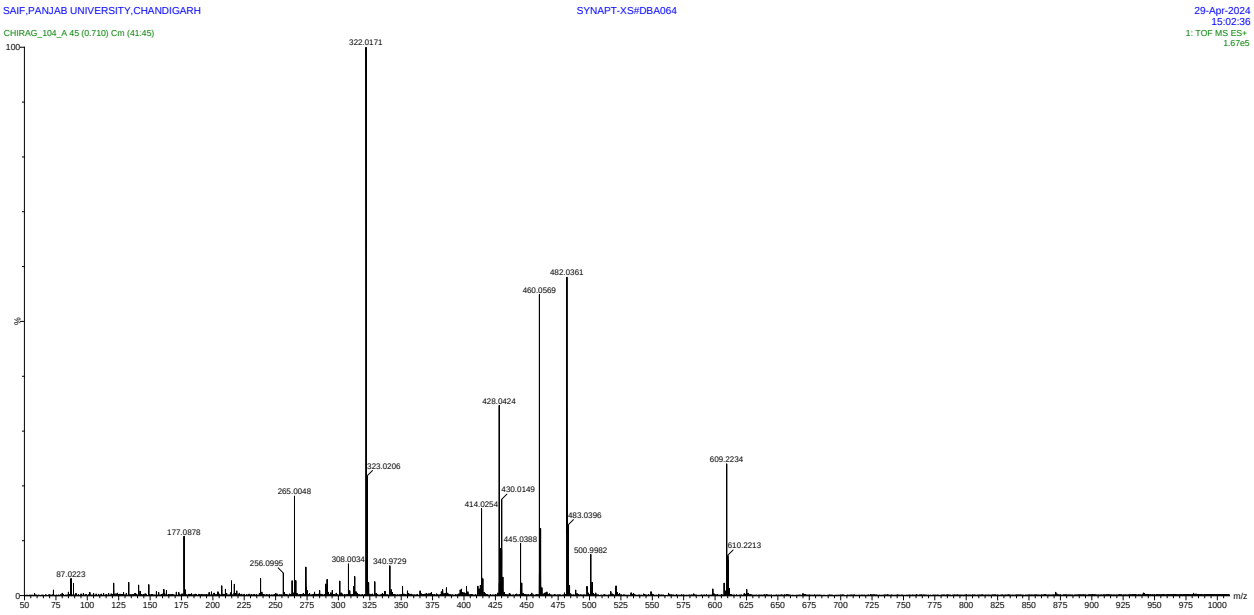


Figure **S68**: HRMS of Compound **6d**.

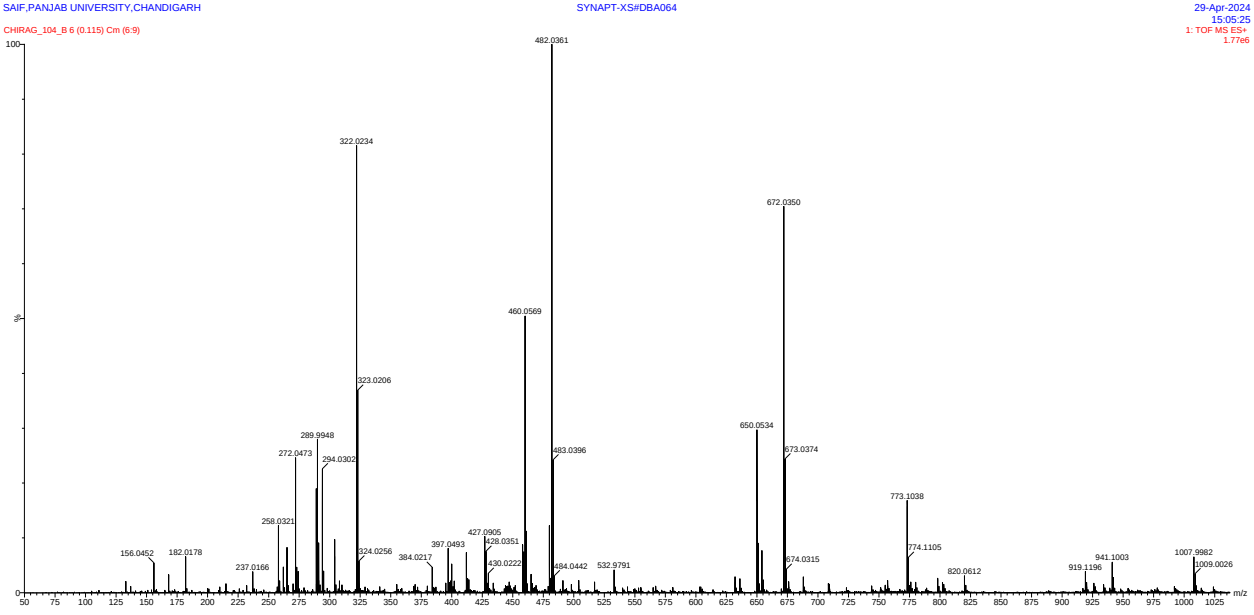


Figure **S69**: HRMS of Compound **5e**.

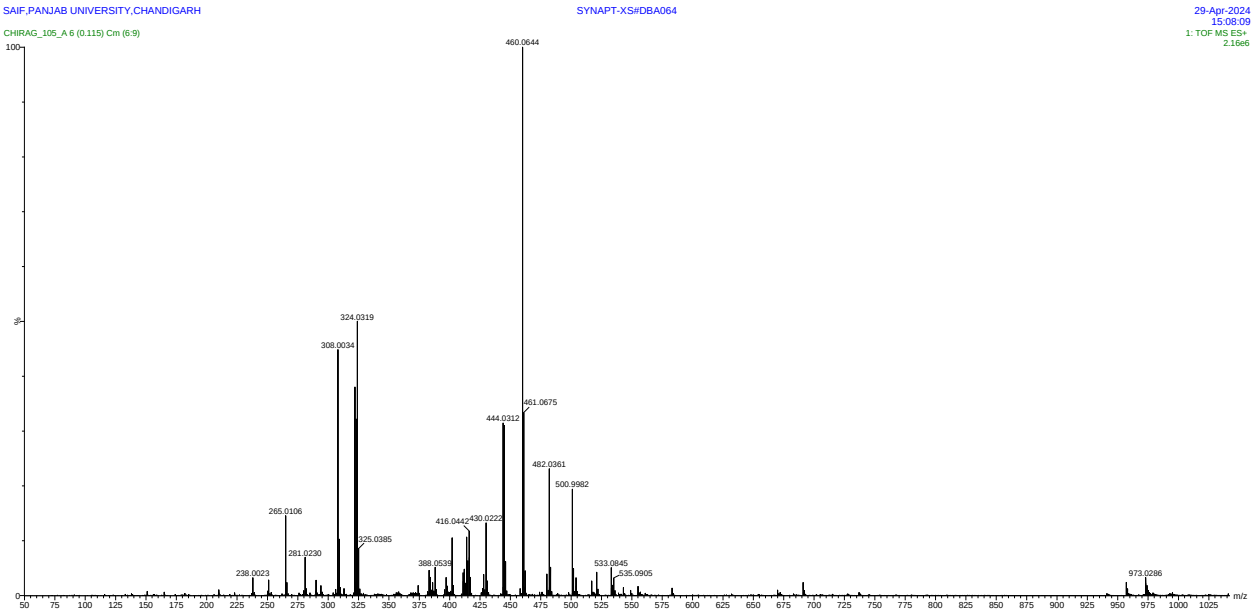


Figure **S70**: HRMS of Compound **6e**.

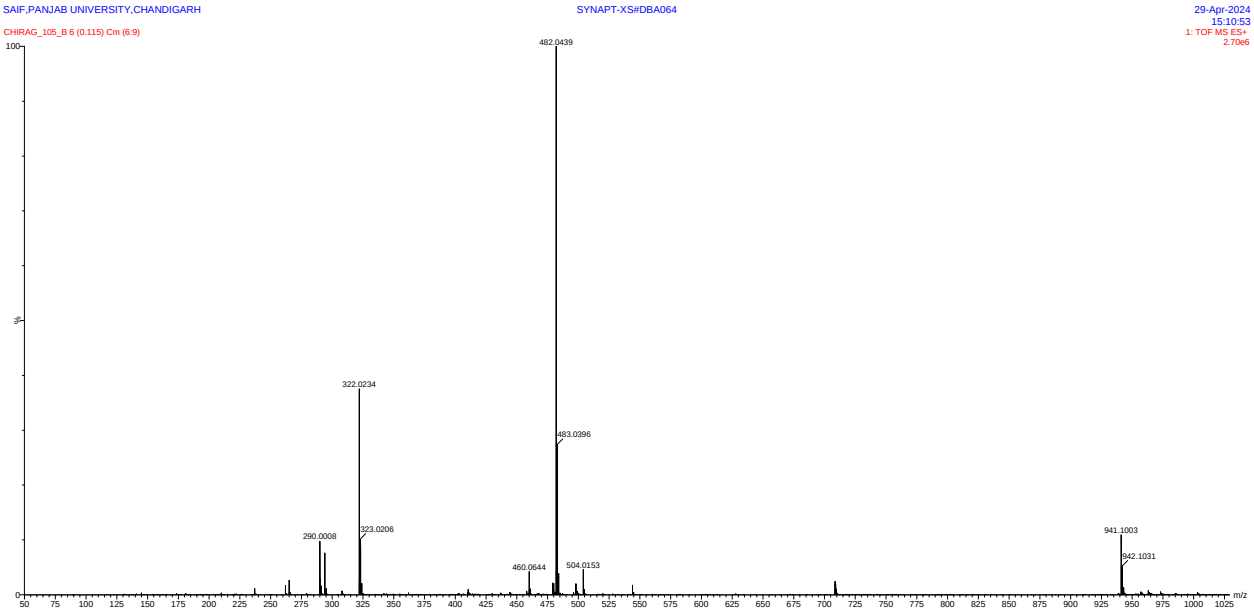


Figure **S71**: HRMS of Compound **5f**.

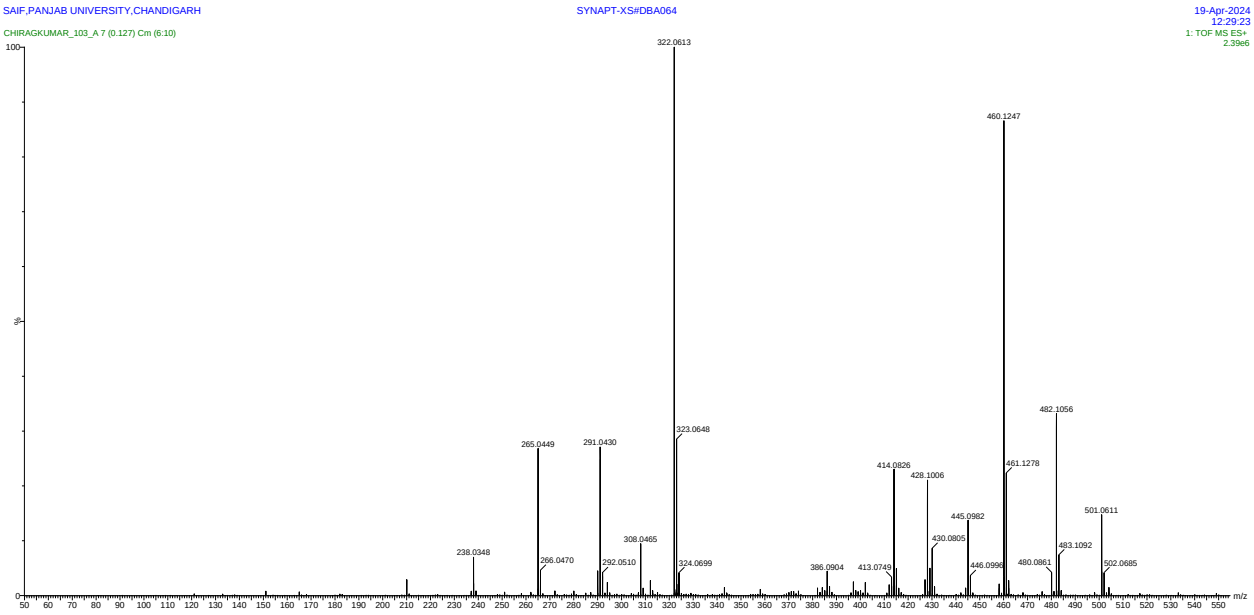


Figure **S72**: HRMS of Compound **6f**.

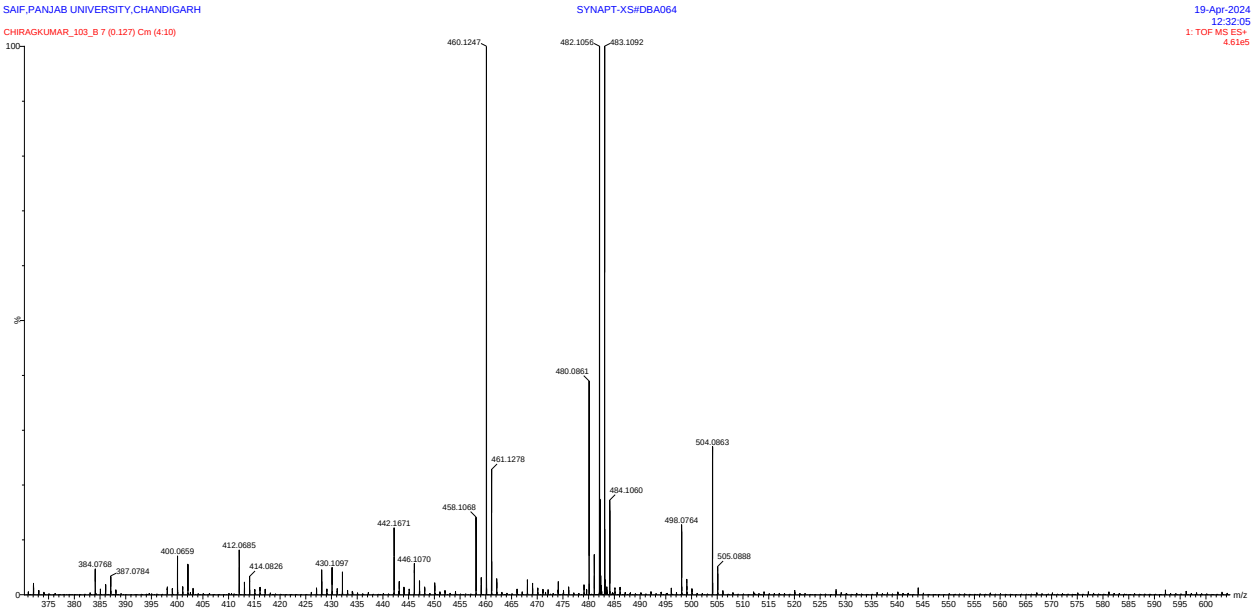


Figure **S73**: HRMS of Compound **5g**.

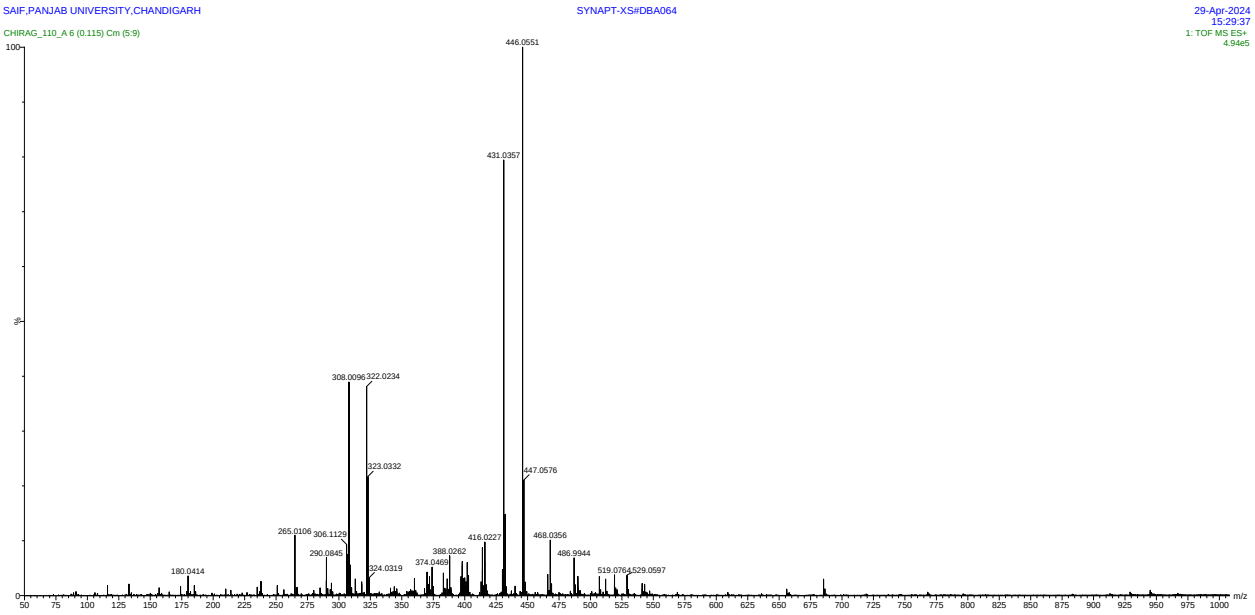


Figure **S74**: HRMS of Compound **6g**.

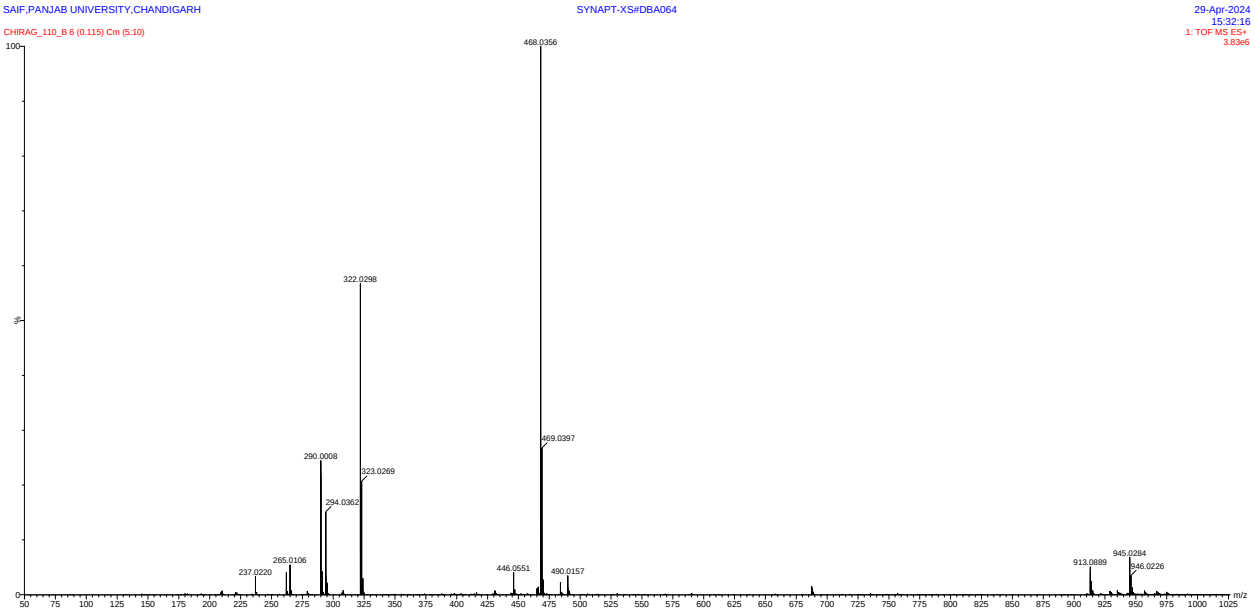


Figure **S75**: HRMS of Compound **5h**.

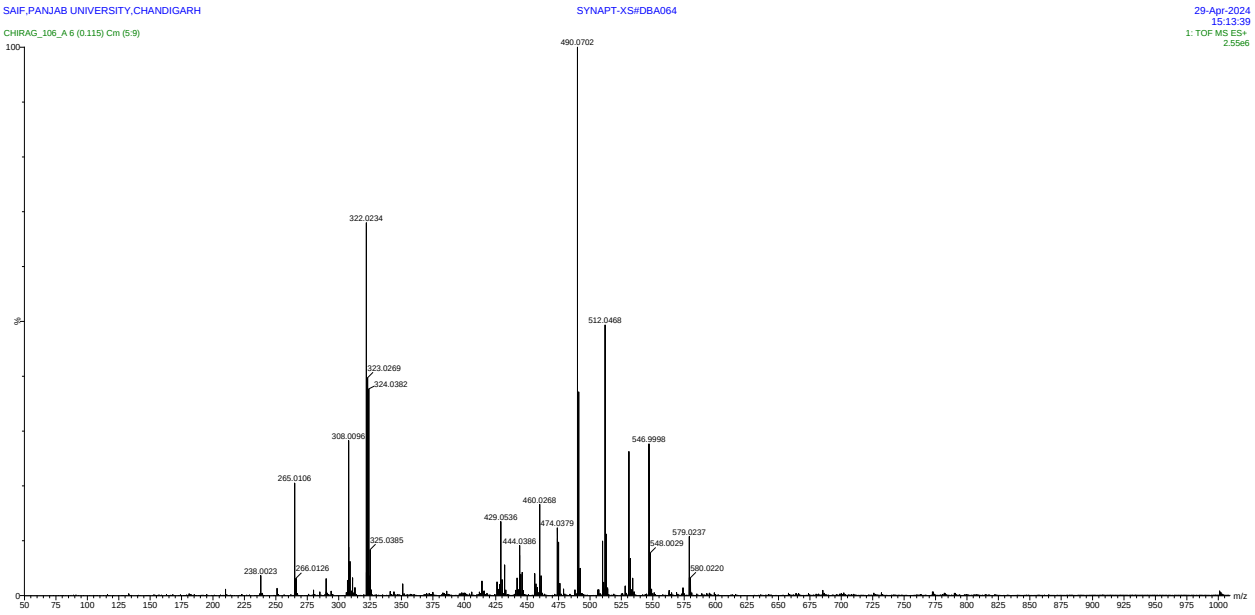


Figure **S76**: HRMS of Compound **6h**.

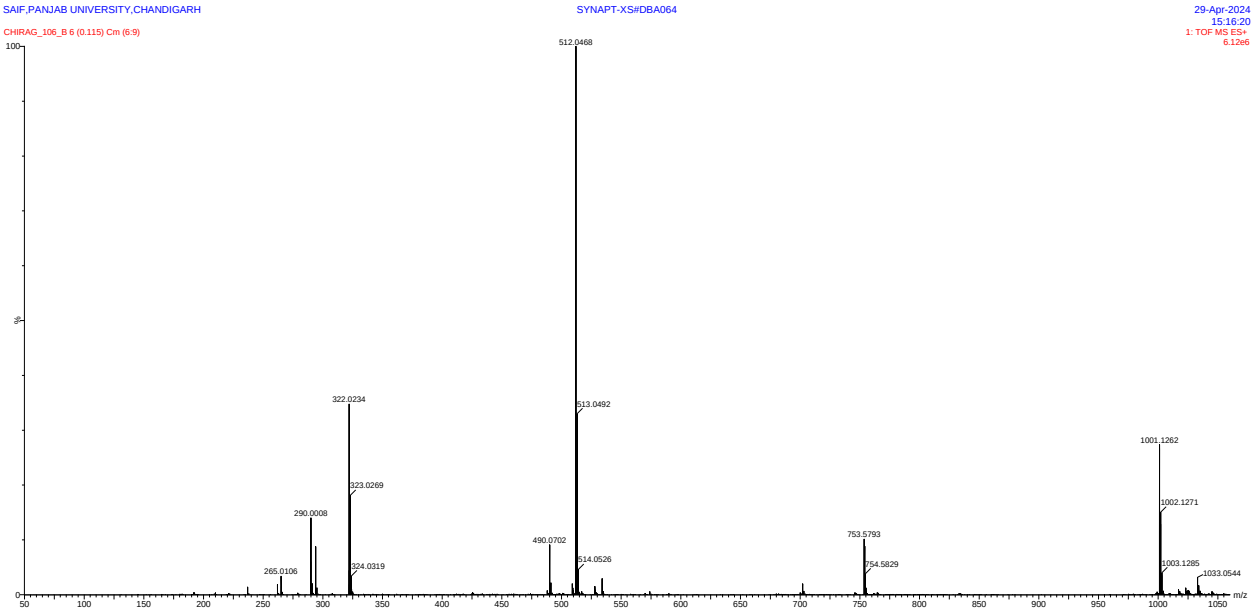


Figure **S77**: HRMS of Compound **5i**.

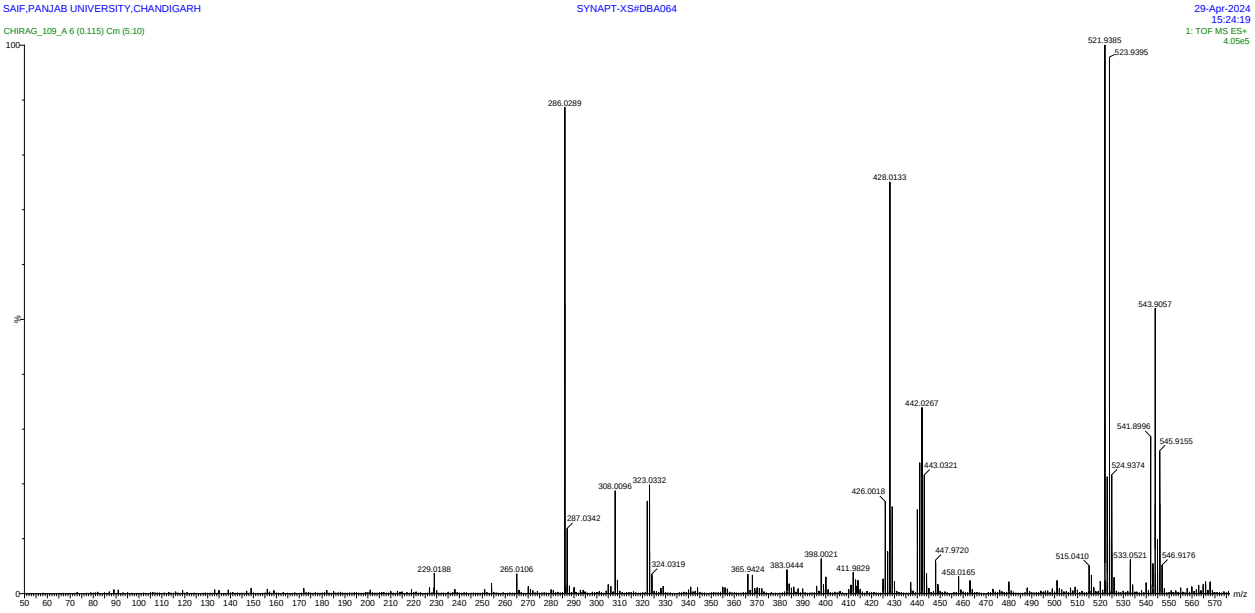


Figure **S78**: HRMS of Compound **6i**.

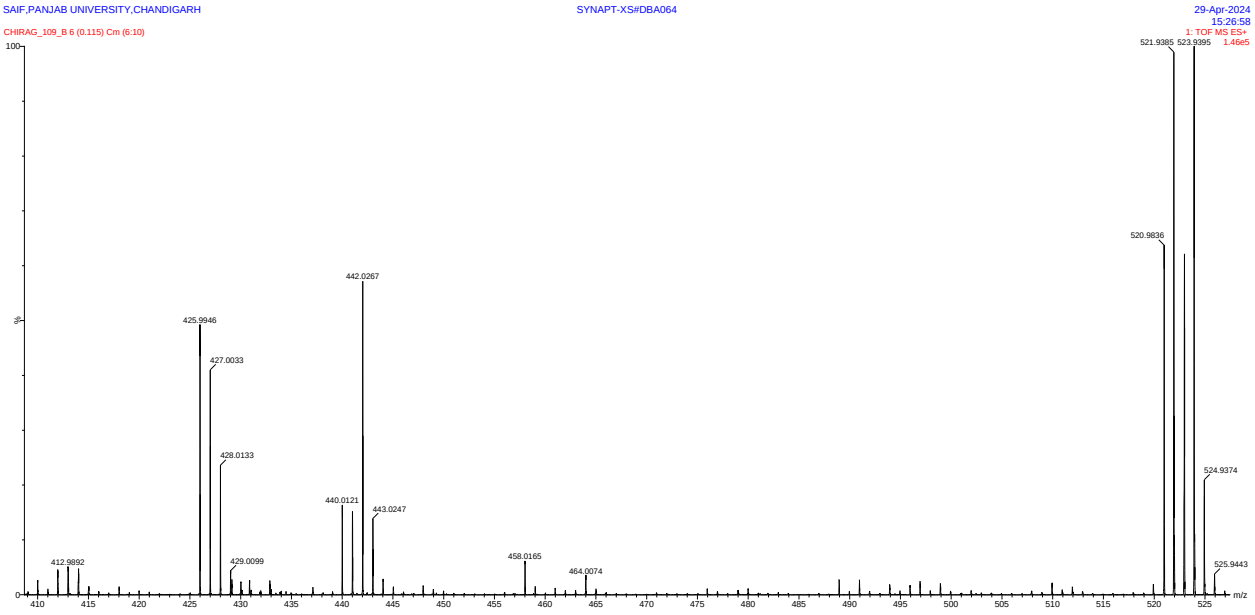


Figure **S79**: HRMS of Compound **5j**.

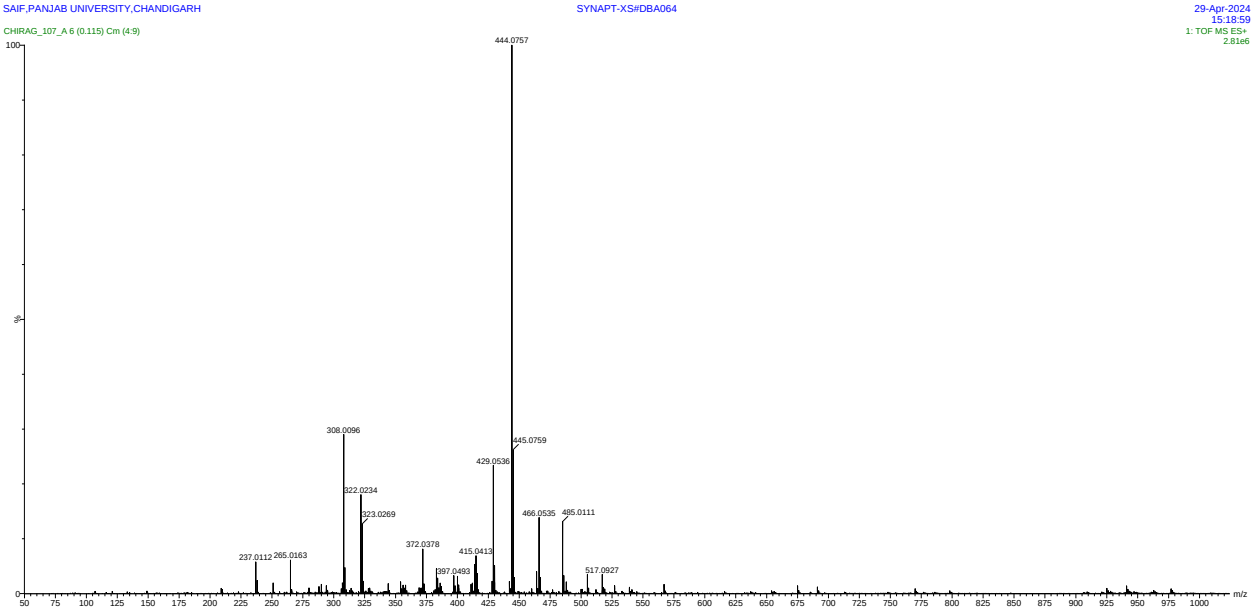
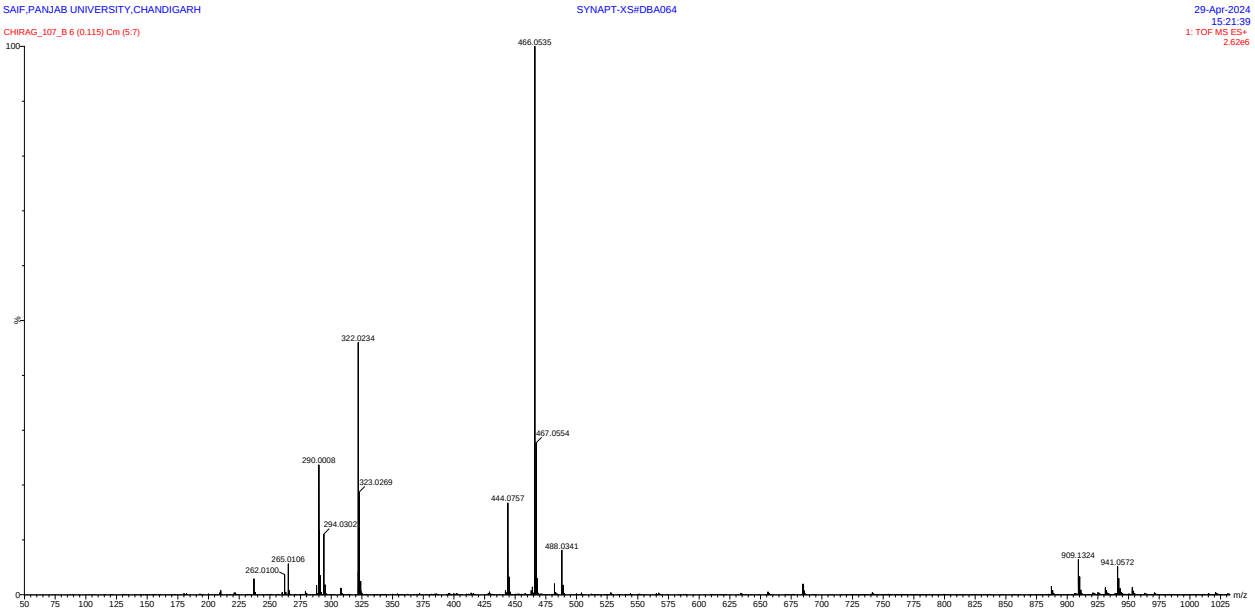


Figure **S80**: HRMS of Compound **6j**.



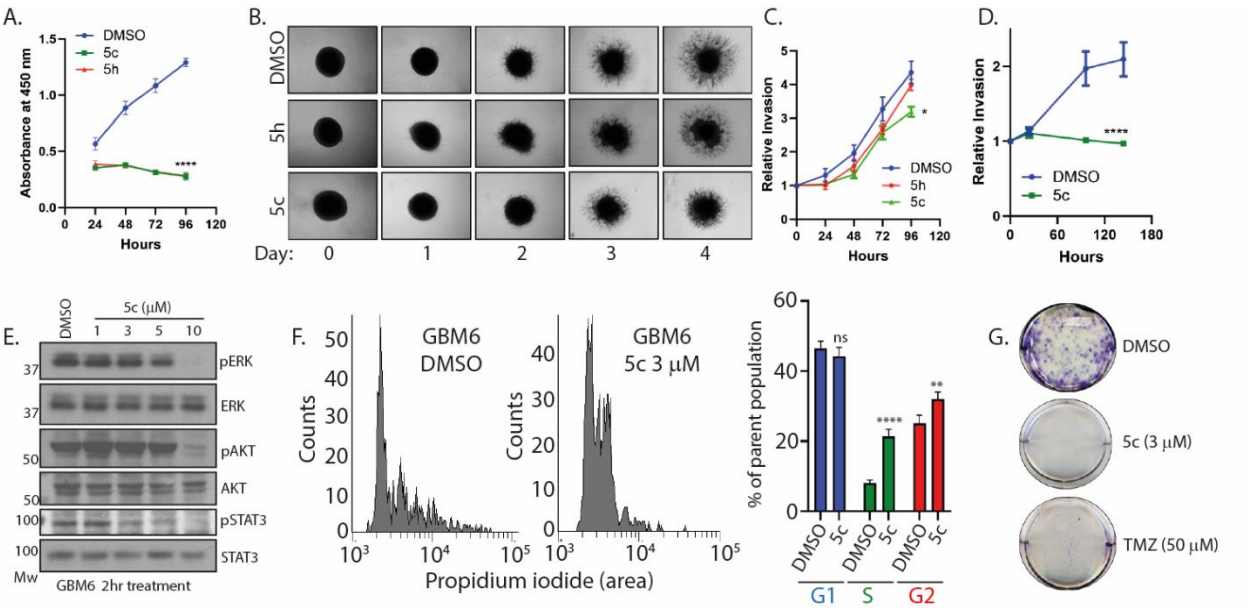


Figure **S81:** Effect of **5c** and **5h** compounds on proliferation and invasion in GBM cells: (A) GBM6 cell proliferation was measured over 5 days in the presence or absence of **5c** or **5h**. Error bar indicates standard error calculated from three independent experiments in triplicate; ****p<0.0001. (B) Panel of representative images of spheroids generated from GBM6 cells, invading basement membrane matrix post treatment with DMSO in control group and **5c** at 2.9 μM concentration and **5h** at 3.3 μM concentration in test groups, respectively. Photomicrographs of each spheroid was taken at x100 at different time points. (C) Relative invasion was calculated and quantified. (D) Quantification of invasion in DMSO treated control and **5c** at 5 μM concentration at day 0 and day 6. Statistically significant difference is represented as *p< 0.05, and ****p<0.0001. (E) Western blot analysis was carried out with the indicated antibodies for GBM6 cell lysates treated with **5c** for the indicated concentrations for 2 hr. (F) Asynchronous GBM6 cells were treated with various doses of **5c** for 16 hours and the cell cycle distribution was analysed using propidium iodide and quantified using flow cytometry. Statistically significant difference is represented as **p< 0.01, and ****p<0.0001 compared to DMSO-treated control cell population; two-way ANOVA with Sidak's multiple comparison; ns: not significant. (G) GBM143 colony formation assay with or without treatment with **5c** or temozolomide (TMZ) over 15 days.

Abbreviations

PQ	Pyrimido[4,5-*b*]quinolinone
DHPQ	Dihydropyrimido[4,5-*b*]quinolinone
TLC	Thin Layer Chromatography
LCMS	Liquid Chromatography Mass Spectrometry
HRMS	High-Resolution Mass Spectrometry
NMR	Nuclear Magnetic Resonance
DEPT	Distortionless Enhancement by Polarization Transfer
HSQC	Heteronuclear Single Quantum Coherence Spectroscopy
HMBC	Heteronuclear Multiple Bond Correlation Spectroscopy
Ar	Aromatic
DMSO	Dimethyl Sulphoxide
TMZ	Temozolomide

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